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Significant Research Achievements of Post Graduate Students (2007-2016)

Post Graduate School
Indian Agricultural Research Institute
New Delhi - 110 012

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Editors

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Printed: **February, 2017**

Citation:

T.K. Behera, Sunil Pabbi, V.K. Baranwal, K.M. Manjaiah & R.K. Jain (2017), Significant Research Achievements of Post Graduate Students (2007-2016), Post Graduate School, Indian Agricultural Research Institute, New Delhi - 110 012, India pp x+608

ISBN: 978-93-83168-24-8

Published by

Dean & Joint Director (Education) on behalf of the Director & Vice Chancellor
ICAR-Indian Agricultural Research Institute, New Delhi - 110012

Printed at:
Venus Printers and Publishers
B 62/8, Naraina Industrial Area, Phase-II, New Delhi - 110 028
Phone : 011-25891449, 45576780, Mobile : 9810089097

सत्यमेव जयते

त्रिलोचन महापात्र, पीएच.डी.
एफ एन ए, एफ एन ए एस सी, एफ एन ए ए एस
सचिव एवं महानिदेशक
TRILOCHAN MOHAPATRA, Ph.D.
FNA, FNASc, FNAAS
SECRETARY & DIRECTOR-GENERAL

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भारतीय कृषि अनुसंधान परिषद
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Foreword

The Indian Agriculture Research Institute (IARI), popularly known as 'Pusa Institute', is India's premier institute in the field of agriculture research, education and human resource development. In the 1958, IARI was accorded the status of a "Deemed University" under Section 3 of University Grants Commission Act of 1956 and thus became the front-runner of the national agricultural system in India. The unique feature of education at IARI is that research, teaching and extension are fully integrated. This has helped to make the post-graduate education not only more relevant to solve farmer's problems but also made it competitive at national and international levels. So far, 8395 students (3812 M.Sc. and 4583 Ph.D. students) including 366 foreign students from 38 countries have received their degrees from this premier institute. It is well known that this institute is the mother of various agricultural research institutes and State Agricultural Universities of our country. It has been ranked as number one Institution with AAAA^{+} among agricultural universities in the country according to Careers 360 University Ranking 2016.

The IARI alumni have been significantly contributing towards agricultural development not only in India but in many parts of the world. They occupy key positions in different agriculture universities, research institutes, state department of agriculture, government, as well as private companies. I learn that the PG School of IARI has successfully produced 1683 post-graduates students during the past 10 years period (2007-2016) and trained about 5000 scientists from different disciplines of agriculture from various parts of India and abroad.

I am pleased to learn that the Institute is bringing out a publication highlighting the significant research achievements of Post-graduate students that constitutes a significant portion of the research output of the Institute. It is noteworthy that many of these research achievements have been published in national and international journals.

The information included in the publication will serve as a ready reference and would be of great value to the scientists, students and administrators. This publication will motivate and inspire young research scholars pursuing their studies to strive towards excellence. Bringing out such a publication needs team work and enormous amount of efforts. I greatly appreciate the contribution of the co-editors Dr. T.K. Behera, Dr. Sunil Pabbi, Dr. V.K. Baranwal, Dr. K.M. Manjaiah and Dr. R.K. Jain, Dean & Joint Director (Education) as well as all the professors and faculty members in its timely publication.

New Delhi
February 9, 2016

(Trilochan Mohapatra)
Secretary, DARE & Director General, ICAR

Preface

The Indian Agriculture Research Institute (IARI) is one of the largest agricultural research establishments in the world. IARI was established in 1905 at Pusa, Bihar and shifted in 1935 to New Delhi with five major sections *viz*; Agricultural & Cattle Breeding, Chemistry, Economic Botany, Entomology and Mycology. The role of the Institute was greatly enhanced in 1958 when IARI received the status of a Deemed University under the University Grants Commission Act 1956. Currently the post graduate programme of IARI has been organized with mono-faculty and multi-crops mandate having 26 disciplines and 647 faculty members including 24 adjunct faculty and 159 faculty members from ICAR Institutes outside Delhi. The education system of IARI is largely based on course credit system, comparable to Land Grant Universities of USA where research, teaching, and extension are fully integrated. Till date the Institute has awarded M.Sc. degree to 3812 and Ph.D. to 4583 students.

The Post Graduate School is continuously striving to scale newer heights in agriculture research, education and extension through the adoption of relevant courses, novel teaching methods as well as continuous capacity renewal of its learned and committed faculty members. In continuation of our earlier publication entitled "Advances in Post Graduate Research for Improving Agricultural Growth and Prosperity" during Golden Jubilee celebration, the present volume on "Significant Research Achievements of Post Graduate Students (2007-2016)" provides an update of research accomplishments made by the post graduate students of IARI with the constant support and guidance of the faculty members. We believe that this volume will not only motivate the scholars towards achieving excellence in research endeavour but also provide valuable research information to the scientific community including planners and academicians.

We sincerely thank the professors, scientists, staff and students of the Institute. We express our deep sense of gratitude to Dr. Trilochan Mohapatra, Secretary, DARE & Director General, ICAR; Dr. N.S. Rathore, DDG (Education), ICAR and Dr. (Mrs.) Ravinder Kaur, Director, IARI for their constant support and guidance in bringing out this volume.

We are highly grateful to the Professors and Faculty members of all the Divisions who have contributed to this volume. We also appreciate the efforts and help rendered by the administrative staff of the Post Graduate School in bringing out this volume.

New Delhi
February 9, 2017

T.K. Behera, Sunil Pabbi, V.K. Baranwal,
K.M. Manjaiah & R.K. Jain

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School of
Crop Improvement

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Vinod, Niharika Mallick, Rama Prashat G. and Vignesh Muthusamy

Introduction

The Division of Genetics at ICAR-Indian Agricultural Research Institute widely regarded as the "Seat of Green Revolution" in India, is one of the important pillars of the institute. The Division has been in existence at IARI since 1960. Since its inception, the Division has been making significant contributions to basic, strategic and applied research in genetics and plant breeding of various crops as well as model genetic organisms. The leadership and vision of several eminent scientists like Drs. BP Pal, MS Swaminathan, AB Joshi, HK Jain and VL Chopra, have immensely contributed to the development of the Division of Genetics at IARI as a center of excellence in teaching and research related to genetics and plant breeding.

The Division of Genetics has nurtured a large contingent of post-graduate students, both from India and abroad. Alumni of the Division have either served or have been serving diverse national and international research organizations as geneticists and plant breeders of high repute. It is a matter of great pride that the alumnus and faculty of this division are decorated with World Food Prize and Padma awards. In its endeavour to develop highly trained human resources, the Division has been continuously upgrading its post-graduate education and research programmes, keeping in view the recent developments in crop genetics and breeding. The mandates of division are:

- Basic and strategic research for genetic enhancement of field crops
- Applied research/varietal development in various field crops
- Transfer of technologies developed by the division
- Post-graduate teaching and research/human resource development
- Developing and strengthening collaborative linkages with national/ international organizations in mutual areas of interest in research and post-graduate teaching

Significant Student's Research Achievements

The significant achievements of this division are primarily contributed by the students research work guided by eminent faculty members with exemplary support from Professors and Heads of the Division. A large number of publications have been made from their work in high impact factor journals. Some of the significant works are presented below under different headings.

Diversity Analysis

Genetic diversity analysis in bread wheat (*Triticum aestivum* L. em. Thell.*)* was carried out to study the variation, association among traits of economic importance and genetic diversity at morphological, biochemical and molecular levels which grouped 36 genotypes into 5, 7 and 6 clusters respectively. Genetic diversity for terminal heat tolerance in bread wheat was carried out using Mahalanobis D^2-statistics for 17 morphological and physiological traits which grouped 35 genotypes in 4, 5 and 5 multi-genotypic clusters in E1, E2 and E3

environments respectively. Twenty three new plant type (NPT) wheat derivatives along with three standard check varieties were investigated for molecular diversity analysis of using a set of 40 STMS markers. Pair wise similarity coefficients among the genotypes based on STMS markers ranged from 0.22 to 0.77 indicating greater extent of diversity among the genotypes which can be utilized in various breeding programmes. The genotypes DL 886, DL 901, DL 924, DL 927, DL 966 and DL 960 were found to be stable, diverse and suitable to wide range of environments for grain yield. The genotypes DL 910, DL 919, DL 960 and DL 940 produced superior bread based on bread loaf volume and other textural characteristics. A study was conducted to work out the genetic divergence for yield, morpho-physiological and quality traits under two sowing conditions namely restricted irrigated and rainfed conditions in wheat. 150 genotypes were grouped into 5 clusters. Genetic diversity and association studies for physiological traits under moisture stress conditions in wheat grouped 294 wheat genotypes into six clusters. Parentage of 294 genotypes revealed that genotypes belonging to different eco-geographical areas were included in the same cluster. This indicated that there is no association between clustering pattern and eco-geographical distribution of genotypes.

Maize (*Zea mays* L.) harbours significant genetic diversity not only in its centre of origin (Mexico) but also in several countries worldwide, including India, in the form of landraces. (Wasala *et al.,* 2013) conducted a study to DNA fingerprint 48 landrace accessions from diverse regions of India using 42 fluorescent dye-labeled Simple Sequence Repeat (SSR) markers, followed by allele resolution using DNA sequencer and analysis of molecular diversity within and among these landraces. The study revealed a large number of alleles (550), with high mean number of alleles per locus (13.1), and Polymorphism Information Content (PIC) of 0.60, reflecting the level of diversity in the landrace accessions Analysis of Molecular Variance showed that 63 % of the total variation in the accessions could be attributed to within-population diversity, and 37 % represented between population diversity. Cluster analysis of SSR data using Nei's genetic distance and UPGMA revealed considerable genetic diversity in these populations, although no clear separation of accessions was observed based on their geographic origin

Rakshit *et al*. (2011) characterised maize inbreds differing for resistance to pink borer (*Sesamia inferens*) using SSR markers and identified eight least susceptible, eight moderately susceptible and 32 highly susceptible inbred lines against pink borer, under artificial infestation conditions. The study has revealed considerable diversity among inbred lines differing for resistance against PFSR and pink borer; and provided ample scope for selection of parents for utilization in heterosis breeding. Muthusamy *et al.* (2015a) characterised a set of 48 maize inbreds differing for kernel carotenoids and identified potential cross combinations that can be exploited for development of biofortified maize hybrids rich in provitamin A carotenoids and antioxidants. Zunjare *et al*. (2015) characterised 48 maize inbreds with varying level of resistance to stored grain weevil (*Sitophilus oryzae*) using 63 SSR markers and grouped them in to three distinct clusters. A total of 177 alleles were detected with mean polymorphism information content of 0.44 and dissimilarity coefficient of 0.62. The diverse genotypes with high degree of resistance has been identified that can be used to develop weevil resistant hybrids. Chaudhary *et al.* (2015) carried out genetic diversity analyses among 24 diverse maize inbreds possessing rare allele of *crtRB1*. The mean β-carotene among the inbreds was 9.3 µg/g, with 14 inbreds having 9.1–18.8 µg/g. Among the inbreds of exotic origin, HP704-23, HP704-22, HP467-15, HP465-41 and HP467-20 had high β-carotene. In case of inbreds developed in India, VQL-2-PV, VQL1-PV, V345-PV, V335-PV, HKI161-PV, MGU-PV-2, HKI1105-PV, HKI323-PV and MGU-PV-3 were identified to possess high kernel β-carotene. These inbreds with rare allele of crtRB1 are unique germplasm and therefore holds immense promise in the biofortification programme worldwide. Molecular profiling of these inbreds using 65 SSRs distributed throughout the genome generated 268 alleles with a mean of 4.12 alleles per locus. The polymorphism information content varied from 0.21 to 0.82 with a mean of 0.58. The study detected 12 each of unique and rare alleles. Genetic dissimilarity ranged from 0.40 to 0.94 with an average of 0.79. Cluster analyses grouped 24 genotypes into four major clusters that depicted the diverse nature of the genotypes consistent with their pedigree. The study identified potential hybrid combinations for higher β-carotene that can be directly utilized in the biofortification programme.

A set of 30 pearl millet genotypes was assessed through D^2 statistics indicating presence of significant differences among the genotypes for various morphological and quality traits. Plant height, days to maturity, leaf blade length and days to 50% flowering contributed maximum to the diversity among the genotypes. Similarly, varying concentrations of tryptophan, lysine, iron, zinc and protein contributed towards nutritional quality diversity among the genotypes. Based on morphological and nutritional traits, the genotypes were clustered in to three clusters at 70% similarity level. Genotypes identified with desired nutritional quality components are: High iron content: PPMI 295 (140.05mg/kg), PPMI 893 (131.75 mg/kg), PPMI 162 (130.65 mg/kg). High zinc content: PPMI 721 (150.80mg/kg), 189 B (126.30 mg/kg). High protein: PPMI 721 (12.37%). High oil content: PPMI 69 (4.43%). High lysine: PPMI 654 (356 mg/100g). High tryptophan: PPMI 872 (338.5 mg/100g). Low phytic acid: PPMI 775 (0.53 g/100g). High reducing sugar: WGI 145 (3.56%). Such genotypes will be valuable materials for breeding program improvement of quality and yield.

Forty eight *Vigna* genotypes were characterized using 17 ISSR, 7 URP and 4 SSR markers. genotypes were grouped in 5 clusters indicating no association between clustering pattern and eco-geographical distribution of genotypes. Based on intercluster distance hybridization between genotypes of cluster I and cluster IV is recommended. RAPD and SSR markers were used to survey genetic diversity among 83 lentil genotypes that included 23 wild types, 19 indigenous varieties, 5 exotic varieties and 36 advanced lines. Dendrogram based on Jaccard similarity coefficient and UPGMA analysis showed the presence of two major clusters. Cluster I comprised 21 wild accessions of *orientalis* and 1 e*rvoides* subsp., Subcluster II A comprised of 19 released Indian varieties, which confirmed their low genetic base, as they are derived mostly from few parents. Subcluster IIB consisted of 41 genotypes including 5 exotic and 36 advanced breeding lines however; Cluster III had only one genotype of *orientalis* subsp. exhibiting only 45 % similarity with cluster I and II. The comparison of both marker system showed that RAPD markers were more efficient in detecting the variability among and between *Lens* species. The narrow genetic base of released cultivars emphasized the need for broadening of genetic base of breeding material using exotic and wild spp.

Morphological and molecular diversity analysis of soybean [*Glycine max* (L.) Merrill] germplasm lines with special reference to water use efficiency identified genotype EC457196 with highest average similarity (0.7417±0.1195) with all the genotypes. EC472130 exhibited lowest average similarity (0.5917±0.1071) with all the genotypes. The genotypes did not cluster as per the WUE or CID values.

Morpho-Physiological and Molecular characterization of chickpea (*Cicer arietinum* L.) genotypes for terminal drought tolerance based on RWC and MSI as well as on gene amplification with EREB and SPS medi identified genotype L550, Pusa Green 112 and ICC 92944 as most drought tolerant. These can be used in chickpea breeding programme for development of genotypes with greater resilience to terminal drought stress. Morphological, physiological and molecular characterization of chickpea (*Cicer arietinum* L.) genotypes for salt tolerance identified genotypes as most salt tolerant based on RWC, MSI and Na:K ratio as well as on gene amplification with CAD, Myb transcription factor and DHN were ICCV00104, ICCV06101, ICCV 101316 along with previously reported CSG 8962, JG 62 and ICCV 10. These can be further utilized in chickpea breeding programme for development of genotypes with greater resilience to salt stress environments. Morphological, physiological and molecular characterization of chickpea (Cicer arietinum L.) genotypes identified genotypes ICCV00104, ICCV06101, ICCV 101316 along with previously reported CSG 8962, JG 62 and ICCV 10 as most salt tolerant. These can be further utilized in chickpea breeding programme for development of genotypes with greater resilience to salt stress environments.

Genetics and Breeding for Biotic Stress Resistance

Pusa Basmati 1121 (PB1121) is a widely grown variety known for its excellent grain and cooking quality in the international and domestic market but is highly susceptible to bacterial blight (BB) disease. A novel BB resistance gene *Xa38* was incorporated in PB1121 from donor parent PR114-*Xa38* using a modified marker-

assisted backcross breeding (MABB) scheme (Ellur *et al.,* 2016). Phenotypic selection prior to background selection was instrumental in identifying the novel recombinants with maximum recovery of recurrent parent phenome (Fig. 1). The strategy was effective in delimiting the linkage drag to <0.5 mb upstream and <1.9 mb downstream of *Xa38* with recurrent parent genome recovery upto 96.9% in the developed NILs. The NILs of PB1121 carrying *Xa38* were compared with PB1121 NILs carrying *xa13* + *Xa21* (for their resistance to BB. Both NILs showed resistance against the Xoo races 1, 2, 3 and 6. Additionally, *Xa38* also resisted Xoo race 5 to which *xa13* + *Xa21* was susceptible. The PB1121 NILs carrying *Xa38* gene will provide effective control of BB in the Basmati growing region.

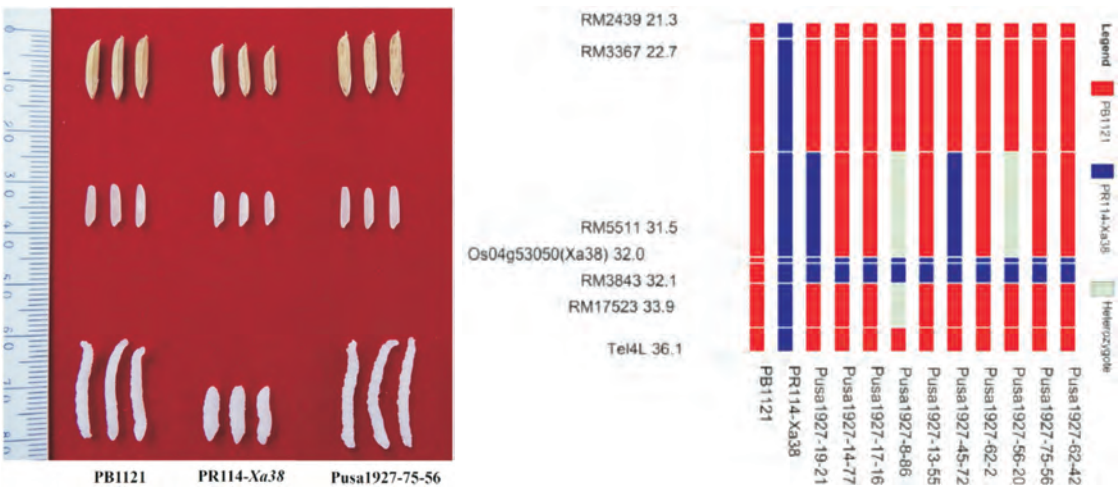

Fig. 1. (A) Grain and cooking quality of the recurrent parent PB1121, donor parent PR114-Xa38 and the PB1121-NIL (Pusa1927-75-56) possessing *Xa38*. (B) Depiction of extent of linkage drag in the selected NILs carrying BB resistance gene *Xa38*

Genetic analysis showed that leaf rust resistance in Sel.T3336 is controlled by one dominant gene against races 77-5 and 104-2 and in WR95 by a single dominant gene against race 77-5 and one dominant and one recessive genes against race 104-2 (Mallick et al. 2010). Expression profiling of leaf rust resistance (*Lr*) gene in wheat (*Triticum aestivum* L.) revealed specific amplification for DDTS 6 and DDTS 9 in the compatible interaction. The transcripts identified and differentially expressed during wheat and leaf rust interaction will be helpful in manipulating the pathways involved during appearance of resistance in host and emergence of virulence in pathogen.

Among various factors for post-harvest loss in maize, infestation of stored grain weevil (*Sitophilus oryzae* L.) is recognized as the major one. The weevil under favourable storage conditions can cause damage up to 80%. Chemical control is associated with serious health and environmental hazards. Thus, breeding for resistance to stored grain weevil holds significant. In the present study, a set of 230 inbred lines were evaluated to assess the genetic variability for resistance to weevil. Grain weight loss (3.40 to 42.32%), number of insect progeny emerged (5.00 to 76.33), germination (0 to 81.33%), pericarp thickness (36.18 to 194.83 µm) and grain hardness (62.33 to 1171.67 N) showed significant variations (Zunjare *et al.,* 2016). Grain weight loss and number of insect progeny emerged was positively correlated, while germination percentage was negatively correlated with grain weight loss and number of insect progeny emerged. Pericarp thickness and grain hardness did not show association with the above mentioned traits. Inbred lines CML394, SKV21, LM13, V351, Pant112, Pant109, Pant124, CML442, MGB1 and CML207 were resistant inbred lines with less grain weight loss, insect progeny and high germination percentage. Pop corn inbreds were more resistant, while QPM inbreds were highly susceptible (Zunjare *et al.* 2015b). Genetic Analyses of Resistance to Stored Grain Weevil (*Sitophilus oryzae* L.) in maize revealed that heritability for grain weight loss and number of insect progeny emerged was 29.41% and 32.55%, respectively. Based on mean performance and combining ability of both the traits, Pant110 and CM135 among

the lines and MGP462 among the testers were identified as promising inbreds for weevil resistance (Zunjare *et al.,* 2015). Turcicum leaf blight (TLB), caused by *Exerohilum turcicum*, is among the most important diseases affecting maize production and productivity in India.A set of 80 diverse inbred lines and landrace accessions were investigated for its reaction to TLB. Parental lines of some of the public-sector maize hybrids in India were found to be highly vulnerable to TLB at both test locations; these include CM138, CM139, CMM140, CM212, V341 and V345. Significantly, entries that a resistant to TLB at both locations, including NAI147, CM145 and one landrace accession from Assam (IML241) has been identified.

Soybean mosaic virus (SMV) is an important disease of soyabean. On the basis of natural incidence of SMV, infection index varied from 5.5 to 100%. Four genotypes namely SL 900 (5.5%), PS 1505 (5.5%), SL 958 (11.1%) and DS 2706 (11.1%) were found to be HR (Highly Resistance) whereas Bragg showed R (Resistance) reaction (29.6%), rests were susceptible. Out of 23 genotypes which were found to be susceptible under field screening only seven were found to be susceptible under challenge inoculation method (OD value of ELISA at 405nm > 0.342). The rate of seed-transmission varied depending on the genotype, the viral strain and the interaction between them.

Genetics and Breeding for Abiotic Stress Tolerance

Cultivar GW 322of wheat (*Triticum aestivum* L.) was improved for drought tolerance using Marker Assisted Back Cross Breeding (MABB) by introgressing QTLs controlling chlorophyll content, leaf temperature BARC68-BARC101, NDVI Xgdm93, staygreen Xgwm111, yield and yield components Xwmc 89 from HI1500. MABB was carried out to introgress heat tolerant QTLs controlling traits like days to anthesis (qANTH) and grain yield under stress (*qGrain yield*) linked with *barc186* and *gwm190* SSR markers, respectively from identified heat tolerant variety WH730 into an most popular, high yielding but high temperature stress susceptible Indian elite variety HD2733. Genetic analysis of high temperature tolerance in wheat (*Triticum aestivum* L.) was carried out using genotypes HD2808 and DL711 which revealed that high temperature tolerance of bread wheat is controlled mostly by epistatic alleles and suggested that selection must be employed at different locations and time during varietal development. Based on stability parameters wheat genotypes DW 1505, HD 2967, Vidhisa, HD 2851, HD 3055, PBW 590, DW 1508, HD 3052, NB 650, DW 1510, HD 3081, CL 3090 and WH 1021, Kundan identified as stable genotypes for grain yield and other yield and heat tolerance related traits across the environments. These genotypes can be utilized for introgression of useful genes to obtain new combinations for developing new genotypes for terminal heat stress affected areas.

24 modern cultivars of wheat were subjected to genetic and physiological characterisation in order to ascertain their use as parents in drought and heat tolerance breeding (Ramya et al. 2015). The physiological traits, canopy temperature and chlorophyll content of flag leaves were found to be widely distributed. Population structure analysis grouped them into seven clusters with optimum k value=7 (Fig. 2). AMOVA and PCoA (Principal Coordinate Analysis) revealed higher level of polymorphism among the resistant and susceptible

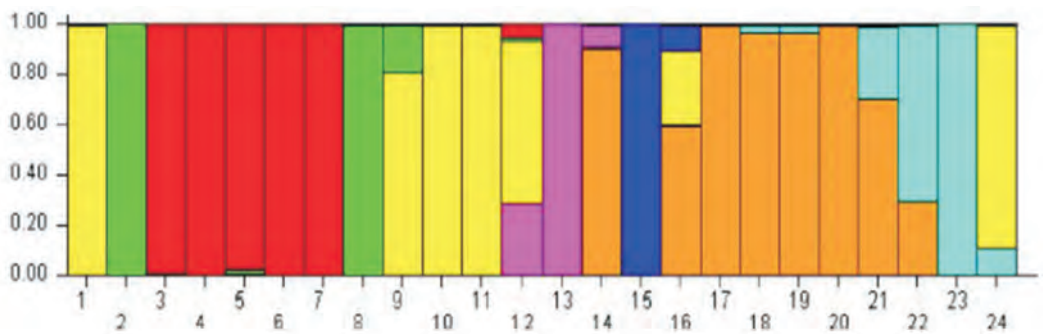

Fig. 2. Population structure of 24 wheat varieties grouped into 7 sub populations at k=7.

groups. The study revealed that the varieties used in breeding program are highly divergent pure breeding lines with unique pattern of alleles.

Genetic analysis of moisture stress tolerance in wheat (*Triticum aestivum* L.) was carried out which revealed positive and significant correlations for thousand kernel weight (0.436), grain weight per spike (0.486), canopy temperature depression at anthesis (0.500) and relative water content (0.502), and yield. Days to maturity, Canopy temperature depression, membrane injury and hectoliter weight had direct positive effects on yield both at genotypic and phenotypic levels across the three environments.

Genetic studies on salt tolerance in chickpea identified a complex type of inheritance. Parent Pusa 1103 was found to be good general combiner for seed yield per plant, RWC, MSI and harvest index, hence this genotype may be used as a potential source in future salinity breeding programme. Morphological, physiological and molecular characterization of chickpea genotypes were undertaken and results indicated the presence of variability among the genotypes. Correlations under stress conditions identified PSY (plant seed yield) to be significantly positively correlated with FPP (Filled pods per plant), RWC (relative water content), MSI (membrane stability index) and significantly negatively correlated with Na:K ratio. These traits can be used as selection indices for selecting genotypes with specific adaptation to salt stress conditions. The genotypes that emerged as most salt tolerant in the study in morphological, physiological scoring based on RWC, MSI and Na:K ratio as well as on gene amplification with CAD, Myb transcription factor and DHN were ICCV00104, ICCV06101, ICCV 101316 along with previously reported CSG 8962, JG 62 and ICCV 10. These can be further utilized in chickpea breeding programme for development of genotypes with greater resilience to salt stress environments.

Candidate genes for drought tolerance that had been validated in chickpea were deployed to identify the most promising genotypes. Only three out of these viz., SPS medi, CAD and EREB genes amplified. The association analysis of these genes with the physiological parameters indicated membrane stability index (MSI) in case of EREB and MSI and RWC in case of SPS medi to be associated with drought stress. The genotypes that emerged as most drought tolerant in the study in both physiological scoring based on RWC and MSI as well as on gene amplification with EREB and SPS medi were L550, Pusa Green 112 and ICC 92944. These can be used in chickpea breeding programme for development of genotypes with greater resilience to terminal drought stress.

Seventy Indian soybean genotypes were evaluated for their resistance to iron deficiency chlorosis (IDC) in the hydroponics solution culture. The study clearly depicted sufficient variation present in the Indian soybean for iron deficiency chlorosis resistance. Based upon the relative iron content, the genotypes were classified into four categories: category I- Fe stress responsive and inefficient (EC 25720, KYUNG-NOW-R2, EC389178, IC 24069, EC 113396, UPSL 656, UPSV 12, and UPSV 24), category II- Fe stress responsive and efficient (PK 1169, UPSV 27, EC439608), category III - Fe stress nonresponsive and efficient (PK 464, PKV 25), and category IV- Fe stress nonresponsive and inefficient (DS 74, EC 114526, IC-76151-W, DS-76-1-2-3, G2650, UPSV 12, EC 439606, EC389170, EC 389179, L 416, UM 77, TGX-1831-3E). The genotypes viz. PK 1169, UPSV 27 and EC 439608 identified as promising material for IDC (iron deficiency chlorosis) in soybean for resistance breeding.

An investigation into phenotypic and genotypic variance, heritability, genetic advance, correlation coefficients and path analysis for yield, morpho-physiological and agronomic traits in pearl millet genotypes under rainfed conditions for 13 quantitative characters indicated ample genetic variability among genotypes particularly for thermo-tolerance (Sankar *et al.,* 2013). Heritability and genetic advance estimates suggested higher heritability (84.8%) and predominance of additive genetic variance for membrane stability index (MSI). Positive and significant correlation was recorded for 1000 seed weight (0.504*), spike girth (0.346*), leaf blade width (0.342*) with grain yield per plant whereas days to 50% flowering (-0.359*) and days to maturity (-0.354*) showed significant negative correlation. The path coefficients indicated 1000 seed weight, spike girth, leaf blade width to have positive direct effects on grain yield per plant. This indicated the scope for improvement

of these characters through the selection process. Gene Expression Profiling of Heat Tolerance Gene/s form Pearl Millet [*Pennisetum glaucum* (L.) R.Br] identified that differential accumulation of heat shock responsive gene transcript can be a useful trait to select heat tolerant pearl millet genotypes even at seedling stage and can be further used for development of high temperature stress resistant genotypes.

An Indian mustard (*Brassica juncea*) germplasm consisting of 165 genotypes was screened for heat tolerance at seedling stage in the field under early sown conditions for two seasons and in the growth chamber using an artificial screening protocol (Fig. 3). A wide genetic variability for high temperature tolerance at seedling stage was observed in the germplasm. Based on the consistent identification of thermo-tolerant and thermo-susceptible types and the correlation between the field and the growth chamber data, the artificial screening protocol used in the study was standardized, that can be used as an alternative to field screening in the future experiments (Azharudheen *et al.,* 2014). Five thermo-tolerant and three thermo-susceptible genotypes selected based on the preliminary screening were re-evaluated in order to characterize high temperature tolerance at seedling stage in this crop. Two parameters, the extent of lipid peroxidation and specific leaf area were found to be clearly differentiating the thermo-tolerant and thermo-susceptible genotypes and which could be used as reliable selection criteria for thermo-tolerance at seedling stage in this crop.

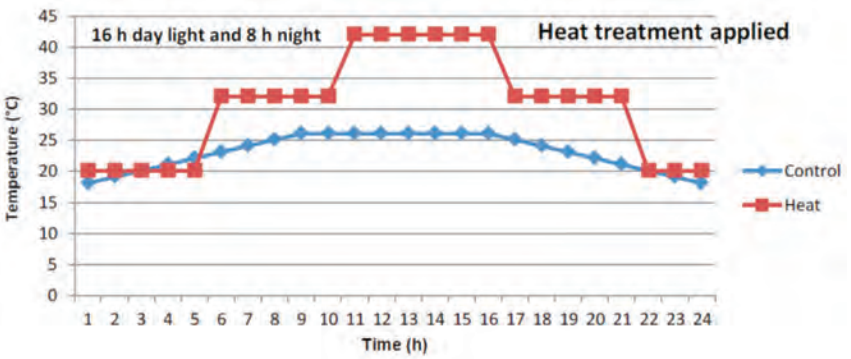

Fig. 3. Heat treatment applied in the treatment and control experiments

Inheritance and Gene Mapping Studies

Cytological analysis in *Aegilops speltoides* derived Selection 2427 in bread wheat during meiotic metaphase showed that the frequency of pollen mother cells with 21^{II} in different plants ranged from 0.67 to 1.00 with an average of frequency of 0.8. Molecular mapping of leaf rust resistance gene in sel. 2427 identified marker Xgpw4044 at map distance of 3.8cM on chromosome 3BL. The bread wheat line WR95 showed seedling resistance to several Indian stem rust (*Puccinia graminis* Pers. *f. sp. tritici*) isolates. Genetic analysis of stem rust resistance in WR95 was undertaken in populations derived from crosses NI5439/WR95 and Agra Local/WR95 (Gireesh *et al.,* 2015). The F1, F2, and F3 generations derived from the cross NI5439/WR95 tested against stem rust isolate 40A showed a recessive gene for resistance. The results were also confirmed in another F2 population derived from the cross Agra Local/WR95. Interestingly, genetic analysis in F2 population from the later cross i.e. Agra

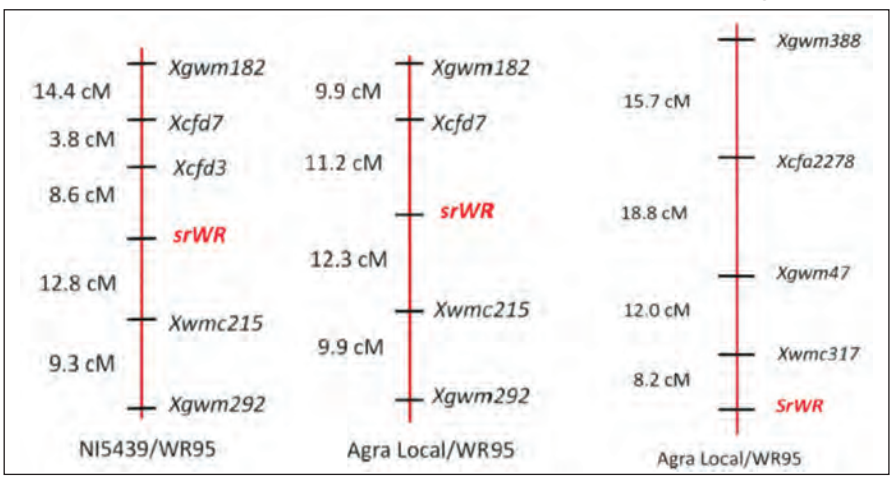

Fig. 4. (A) Linkage map of recessive stem rust resistance gene srWR in WR95 on 5DL chromosome of wheat; (B) Linkage map of dominant stem rust resistance gene SrWR in WR95 on 2BL chromosome of wheat

Local/ WR95 identified a dominant gene against isolates 11 and 11A. The recessive gene against isolate 40Awas mapped to the long arm of 5D chromosome. WR95 showed resistance to virulent isolates of Sr30 which is also located on 5DL chromosome (Fig. 4A). The dominant gene in WR95 that conferred the resistance to isolate 11 and 11A was mapped towards telomeric region of the long arm of 2B chromosome (Fig. 4B). The long arm of chromosome 2B also carries stem rust resistance genes Sr9, Sr16 and Sr28. WR95 showed resistance to virulent isolates to Sr9b (11, 11A, 21A-2 and 40A), Sr9e (15-1 and 40A) and Sr28 (11, 11A, 15-1, 21A-2 and 40A). The map position of dominant gene identified inWR95 is different from Sr9 and Sr28 while Sr16 is towards telomeric region and not mapped so far. Therefore, the dominant gene in WR95 may be either Sr16 or a new gene.

Mapping was carried out with 92 microsatellite markers specific to chromosome 2A on the F2 population of the cross Agra Local x Tc+Lr45 (Naik et al. 2015). Out of seven markers linked to the gene, four (gwm372, gwm275, gpw3167 and gwm122) were co-dominant and the other three (cfd168, cfd6 and gwm249) showed dominance, amplifying the allele only in the susceptible parent. The genetic map of 13.1 cM was constructed based on the results in 140 homozygous resistant and homozygous susceptible plants (Fig. 5). cfd168 was the closest marker linked to Lr45, followed by gwm372. These markers were validated on the NI5439 9 Tc+Lr45 F2 population, 12 different backcross lines carrying Lr45 and nearisogenic lines, mostly in Tc background isogenic for 46 different Lr genes belonging to both native and alien species. The marker gwm122 was found to be monomorphic. The closest co-dominant marker gwm372 showed reduced polymorphism. Two sequence-based primer pairs, G37294 and G372185, were designed and validated which can serve as robust co-dominant markers for utilization of Lr45 in wheat improvement.

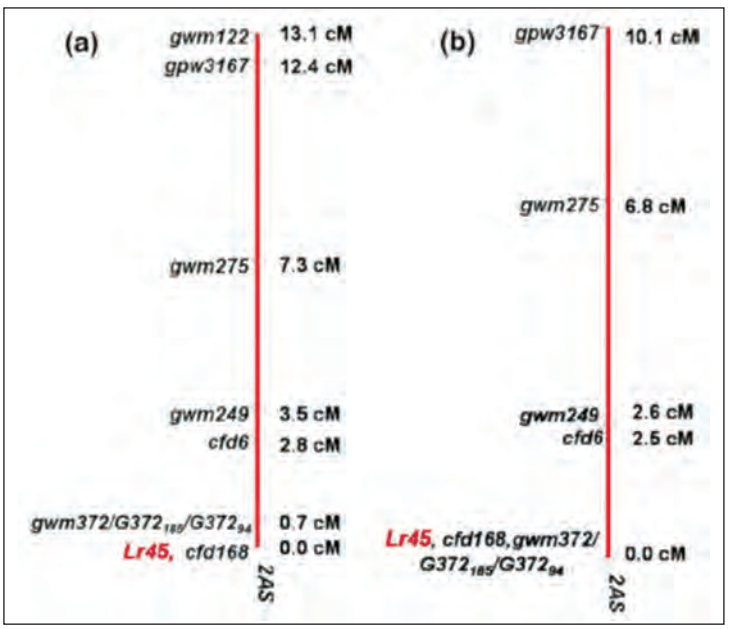

Fig. 5. Genetic linkage map of leaf rust resistance gene Lr45 and relative distance to markers located on chromosome arm 2AS in the populations a Agra Local x Tc+Lr45 and b NI5439 x Tc+Lr45

The SSR markers Satt600 and Satt285 were identified of having association with seed storability in soybean [(*Glycine max* (L.) Merril]. A set of 96 diverse soybean genotypes were investigated for population structure and to identify molecular markers associated with important agronomic traits (Kumar *et al*. 2014). Large phenotypic variability was observed for the agronomic traits under study indicating suitability of the genotypes for association studies. The population structure studies depicted the presence of seven subpopulations which nearly corresponded with the source of geographical origin of the genotypes (Fig. 6). Linkage disequilibrium (LD) between the linked markers decreased with the increased distance, and a substantial drop in LD decay values was observed between 30 and 35 cM. Genomewide marker-traits association analysis carried out using general linear (GLM) and mixed linear models (MLM) identified six genomic regions (two of them were common in both) on chromosomes 6, 7, 8, 13, 15 and 17, which were found to be significantly associated with various important traits viz., plant height, pods per plant, 100-seed weight, plant growth habit, average number of seeds per pod, days to 50% flowering and days to maturity (Fig. 7). The phenotypic variation explained by these loci ranged from 6.09 to 13.18% and 4.25 to 9.01% in the GLM and MLM studies, respectively.

Inheritance and mapping of disease resistance genes in Lentil (*Lens culinaris* Medik) identified dominant monogenic control in all the ten crosses (including reciprocals) involving 3 resistant parents viz. Precoz, PL5

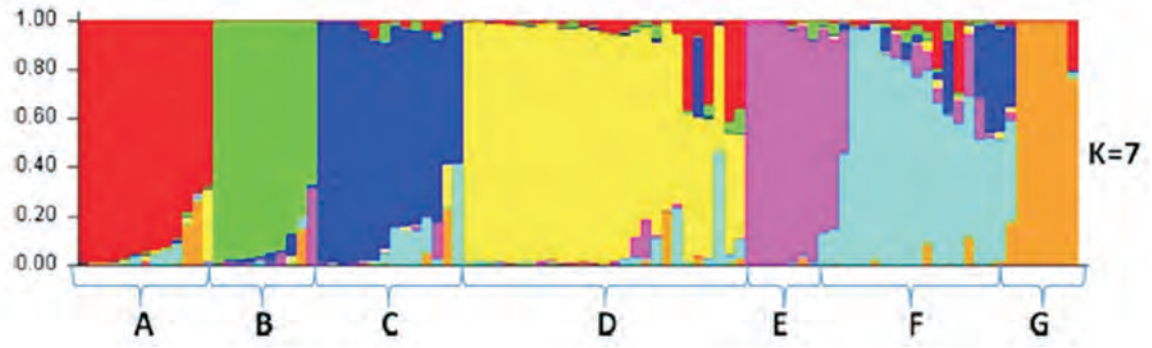

Fig 6. Graphical depiction of the seven subpopulations detected in the population structure study. Partial substructure has been observed in the population corresponding to geographical origin of the genotypes.

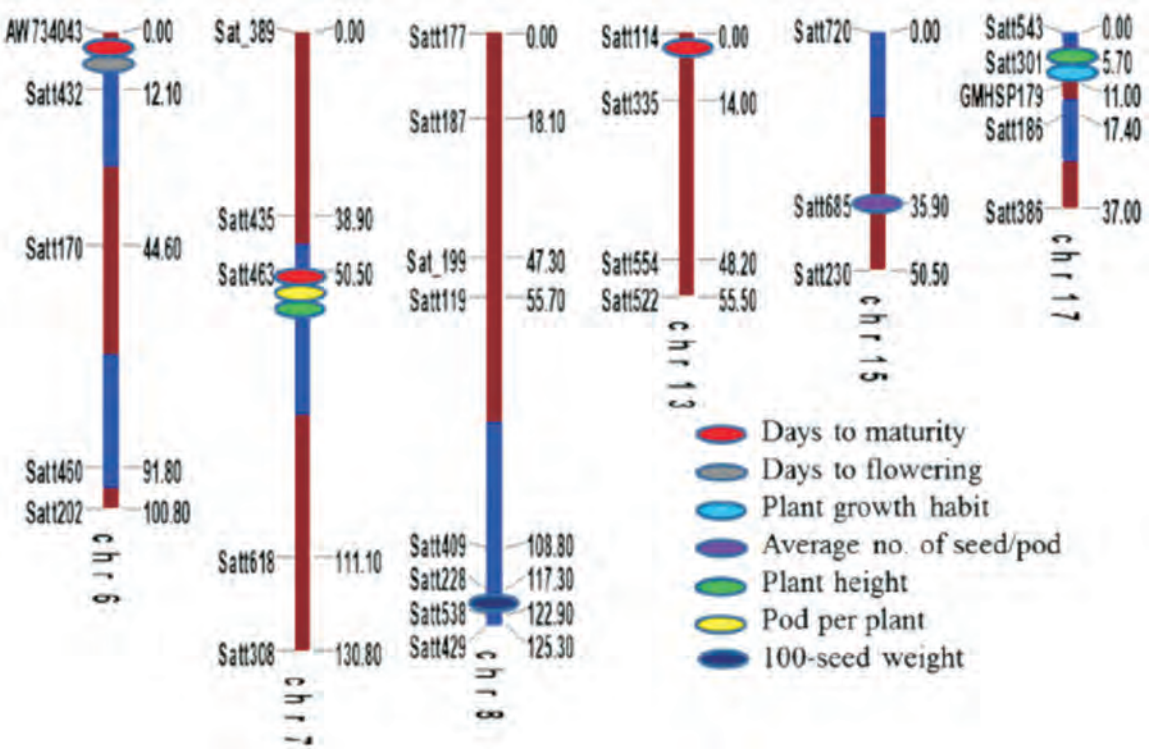

Fig. 7. Localization of six putative loci, AW734043, Satt463, Satt538, Satt114, Satt685 and Satt301 on their respective chromosomes 6, 7, 8, 13, 15 and 17 which were found to be associated with seven important agronomic traits

and DPL58. Dominant x dominant cross between Precoz and PL5 showed F_2 segregation ratio of 15 : 1 (15 Resistant : I Susceptible). This indicated involvement of at least two different genes between them. Molecular tagging of rust resistance gene was carried out in an F_2 mapping population involving 100 individuals from cross DPL 58 x ILL 8006 following bulked segregate analysis (BSA) using RAPD markers. More than 250 RAPD markers were used in the present investigation. Only 10 were found polymorphic between the parents. Out of these 10 markers, only 2 markers (OPS 10 and OPO 12) were able to differentiate the bulk too. The marker OPS 10 differentiated the bulks for rust resistance and the marker OPO 12 differentiated the bulks for blight resistance.

Genetic and molecular analysis of stem rust (*Puccinia tritici*) resistant gene in synthetic hexaploid lines of wheat (*Triticum aestivum* L.) revealed common resistance gene in both the Ug99 resistant varieties Kundan and UP 2338 conferring resistance against the stem rust race 122.One SSR marker Xgwm131 was identified as linked to unknown gene for stem rust resistance in UP 2338, from the analysis of the entire F_2 population of 195 individuals. Genetic analysis of leaf rust resistance in Selection G12 found to be governed by a single recessive gene. This gene was mapped to chromosome 3BL with a closely linked SSR marker X*gwm*547 located at a distance of 6.0 cM. As no other leaf rust resistance gene has been reported on chromosome 3BL so far, the leaf rust resistance gene in Selection G12 is tentatively named as *LrSel.G12* and inferred to be novel.

The trait seedling thermo-tolerance index (STI) in *Brassica spps* was mapped using the method of bulked segregant analysis. The trait was linked to the SSR marker BN12A at a distance of 33.2 cM. Findings of the present study can be used in the future breeding programmes aimed at the development of varieties with high temperature tolerance at seedling stage in Indian mustard. High temperature tolerance in Indian mustard was mapped and SSR marker BN12A at a distance of 33.2 cM was found to be linked with the trait. Findings of the study can be used in the future breeding programmes aimed at the development of varieties with high temperature tolerance at seedling stage in Indian mustard.

Gene Pyramiding

Marker assisted backcross breeding was employed to incorporate the blast resistance genes, *Pi2* and *Pi54* and bacterial blight (BB) resistance genes *xa13* and *Xa21* into the genetic background of Pusa Basmati 1121 (PB1121) and Pusa Basmati 6. Foreground selection for target gene(s) was followed by arduous phenotypic and background selection which fast-tracked the recovery of recurrent parent genome (RPG) to an extent of 95.8% in one of the near-isogenic lines (NILs) namely, Pusa 1728-23-33-31-56, which also showed high degree of resemblance to recurrent parent, PB6 in phenotype. The phenotypic selection prior to background selection provided an additional opportunity for identifying the novel recombinants viz., Pusa 1884-9-12-14 and Pusa 1884-3-9-175, superior to parental lines in terms of early maturity, higher yield and improved quality parameters (Ellur *et al*. 2016). There was no significant difference between the RPG recovery estimated based on SSR or SNP markers, however, the panel of SNPs markers was considered as the better choice for background selection as it provided better genome coverage and included SNPs in the genic regions (Fig. 8). Multi-location evaluation of NILs depicted their stable and high mean performance in comparison to the respective recurrent parents (Fig. 9). The Pi2 + Pi54 carrying NILs

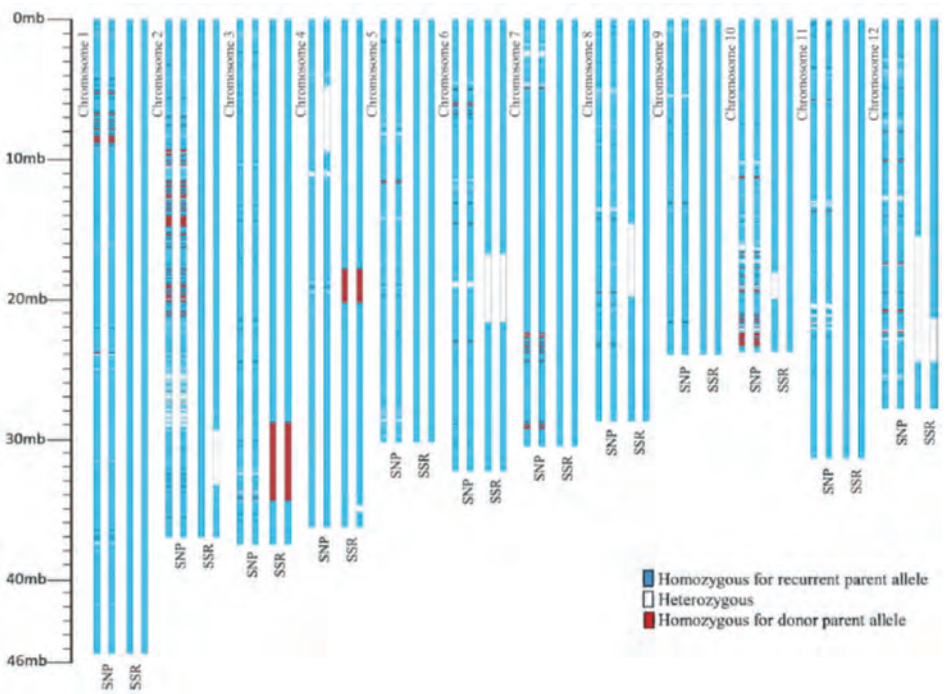

Fig. 8. Graphical representation of RPG recovery using SNP and SSR markers in PB1121-NILs carrying Pi2 + Pi54. In each of the chromosomes first bar represents the NIL Pusa1883-28-16-360 and second bar represents the NIL Pusa 1883-19-9-408.

were effective in combating a pan-India panel of *Magnaporthe oryzae* isolates with high level of field resistance in northern, eastern and southern parts of India. Alongside, the PB1121-NILs and PB6-NILs carrying BB resistance genes xa13+Xa21 were resistant against *Xanthomonas oryzae pv. oryzae* races of north-western, southern and eastern parts of the country. Three of NILs developed in this study, have been promoted to final stage of testing during the *Kharif* 2015 in the Indian National Basmati Trial.

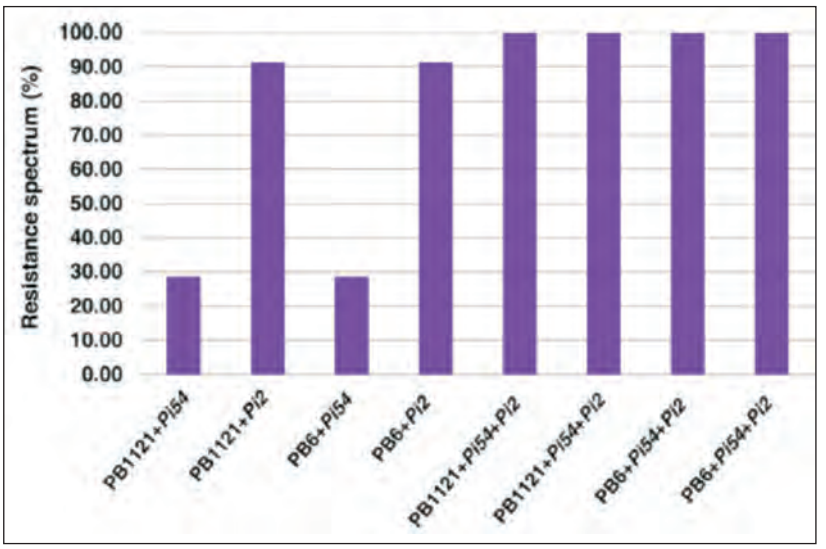

Fig. 9. Per cent of isolates to which NILs carrying blast resistance genes Pi2 and Pi54 or in combination in the genetic background of PB1121 and PB6, showed resistance reaction.

Marker assisted pyramiding of bacterial blight resistant alleles *Xa4, xa5, xa13* and *Xa21* were done into Basmati rice lines of Pusa 1121, Pusa 2511 and advanced breeding lines from PB 1/IRBB 60/Pusa 1302 using IRBB 60 as donor line for all the four genes. A total of 174 plants among three populations were selected based on their desirable bacterial blight resistant gene combinations and better grain and cooking quality attributes.

A widely grown but rust susceptible Indian wheat variety HD2932 was improved for multiple rust resistance by marker-assisted transfer of genes Lr19, Sr26 and Yr10 (Fig. 10). Foreground and background selection processes were practised to transfer targeted genes with the recovery of the genome of HD2932. The near-isogenic lines (NILs) of HD2932 carrying Lr19, Sr26 and Yr10 were individually produced from two backcrosses with recurrent parent HD2932. Marker-assisted background selection of NILs with 94.38–98.46% of the HD2932 genome facilitated rapid recovery of NILs carrying Lr19, Sr26 and Yr10 (Mallick *et al.,* 2015). In the BC2F2 generation, NILs were intercrossed and two gene combinations of Lr19+Yr10, Sr26 + Yr10 and Lr19+Sr26 were produced. A total of 16 progeny of two gene combinations of homozygous NILs of HD2932 have been produced, which are under seed increase for facilitating the replacement of the susceptible HD2932 with three of the sixteen improved backcross lines with resistance to multiple rusts.

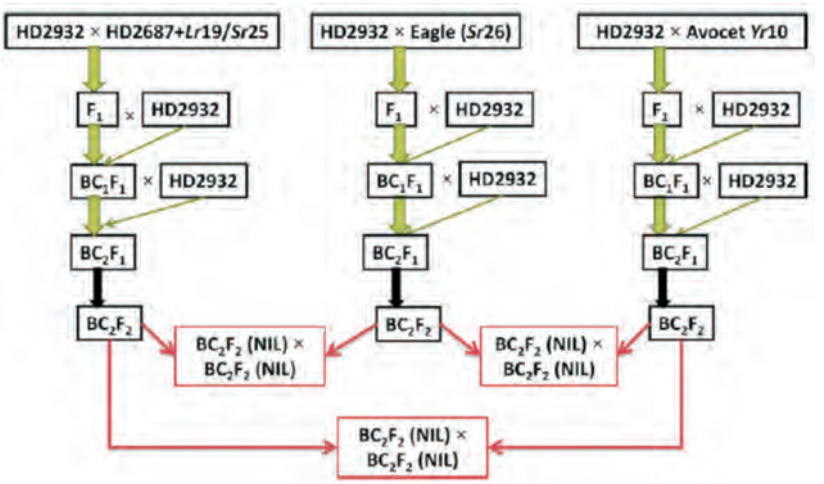

Fig. 10. Breeding scheme for transfer of multiple rust resistance in wheat variety HD 2932 (Foreground and background markers were scored from BC1F1 generation onwards)

Leaf rust, (*Puccinia triticiana Puccinia recondita tritici.*) is a major disease of wheat and breakdown of single major gene for resistance necessitates the search for new combinations of resistant genes. Alternatively, pyramiding of rust resistance genes is useful strategy enhance durability of resistance. Two near isogenic lines HD2687 + *Lr*19 and HD2687 + *Lr*28, were taken to pyramid the leaf rust resistance genes *Lr*19 and *Lr*28 in the genetic

background of HD2687. 51 plants were identified of carrying both the genes in homozygous state with 88.6 to 94.55% recurrent parent background. Foreground selection was done using molecular marker *Xwmc*221, SCS253 and SCS265 for *Lr*19 and SCS421 for *Lr*28. Molecular markers and leaf rust resistance genes followed the expected inheritance pattern in F_2. Among sixty three plants homozygous for leaf rust resistance allele *Lr*19, 51 carried *Lr*28. These 51 plants were analyzed for background similarity with the original recipient variety HD2687. For background selection 69 polymorphic SSR markers distributed throughout the genome were used and could detect 88.6 to 94.55% recurrent parent alleles in F_2 plants carrying both *Lr*19 and *Lr*28. Pyramided plants were tested for leaf rust resistance as adult plants stage against race 77-5 and were found to be highly resistant under artificial inoculation.

Breeding for Quality Traits

Wheat landraces are considered to provide useful sources of genetic variability for various traits including those for quality. Screening of indigenous collection of wheat at Grain Quality Laboratory led to identification of five novel variants on *Glu-B1* and *Glu-D1* loci. Among these, two indigenous collections IC 296923 and IC 296924 which was found to have novel subunit pairs 5_2 & 12_3 (in IC 296923) and 5_3 &12_3 (in IC 296924) investigated for finding out the location of these novel subunits using three way (top) cross and molecular markers. The studies confirmed that the allele for subunit 5_2 and 5_3 is located at the *Glu-D1* locus and that these subunits are suggested to be coded by the *Glu-D1x* gene. It was also concluded that the absence of any subunits of *Glu-D1y* locus in the lines IC 296923 and IC 296924 is due to a deletion mutation at the *Glu-D1y* locus leading to a Null allele at *Glu-D1y* locus in these lines. Combining the genetic and molecular studies, it was confirmed that the Null subunit at *Glu-D1y* has arisen due to a deletion of the DNA in this region. The allelic designations have been proposed for these subunits as *Glu-D1-1w* for 5_2, *Glu-D1-1 x* for 5_3, *Glu-D1-bu* for 5_2+ Null and *Glu-D1-bv* for 5_3+Null. Lines such as 'NapHal' with double null subunits and 'Pissi Local' with subunit 17* and soft grain can serve as donors for introducing better biscuit quality. The new lines IC 296923 and IC 296924 with the novel subunit pairs 5_2+Null and 5_3+Null can also be used as alternatives to these lines as these lines have better agronomical and morphological traits. 306 RILs (F_7) developed from a biparental cross between an old cultivar WH542 and a synthetic derivative (*Triticum dicoccon*PI94624/*Aegilopssqarrosa* (409)//BCN was used to map QTLs governing iron anz zinc in wheat. QTLs for grain iron were termed as *QGFe.iari-2A, QGFe.iari-5A, QGFe.iari-7A and QGFe. iari-7B* and as *QGZn.iari-2A, QGZn.iari-4A, QGZn.iari-5A, QGZn.iari-7A* and *QGZn.iari-7B* for grain zinc. The QTLs explained 2.3-6.8% phenotypic variation for grain iron and 3.2-14.4% phenotypic variation for grain zinc. Two markers, *Xbarc186, Xbarc74* showed an association with grain iron concentration and four markers, *Xgwm3, Xgwm538, Xgwm400* and *Xwms149* showed an association with grain zinc concentration. The identified QTLs in the present study represent novel genomic regions associated with grain zinc and iron concentration.

One of the landmark achievement has been the recent success in enriching kernel β-carotene in four commercial maize hybrids using marker-assisted introgression of *crtRB1* allele (Muthusamy et al. 2014). A favourable allele (543 bp) of the β-*carotene hydroxylase* (*crtRB1*) gene was introgressed in the seven elite inbred parents, which were low (1.4 mg/g) in kernel β-carotene, by using a *crtRB1*-specific DNA marker for foreground selection. About 90% of the recurrent parent genome was recovered in the selected progenies within two backcross generations. Concentration of β-carotene among the crtRB1-introgressed inbreds varied from 8.6 to 17.5 mg/g – a maximum increase up to 12.6-fold over recurrent parent (Fig. 11). The reconstituted hybrids developed from improved parental inbreds also showed enhanced kernel β-carotene as high as 21.7 mg/g, compared to 2.6 mg/g in the original hybrid. The reconstituted hybrids evaluated at two locations possessed similar grain yield to that of original hybrids (Fig. 12). These MAS-derived hybrids are also agronomically promising and currently under final year of national testing. This study was the result of comprehensive genetic analyses of kernel carotenoids in maize (Muthusamy *et al.* 2015a), molecular characterization of these inbreds (Muthusamy *et al.*

2015b), and identification of inbreds with favourable allele of *crtRB1* and *lcyE* for enrichment of provitamin A carotenoids (Muthusamy *et al.,* 2015c).

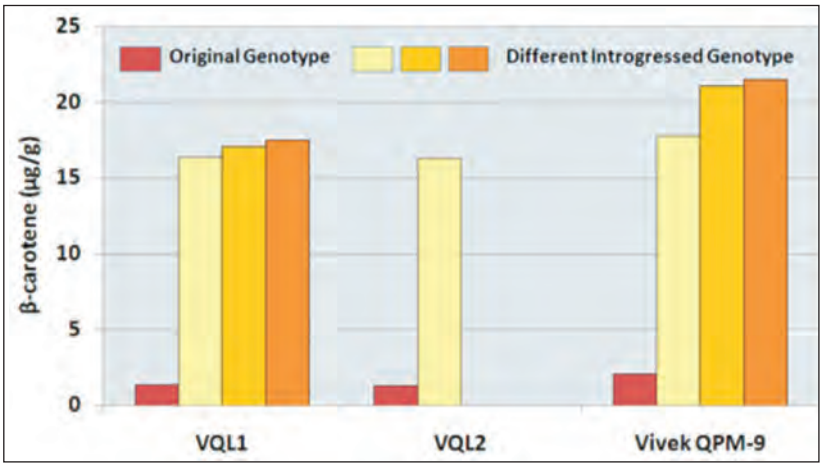

Fig. 11: Kernel β-carotene in original and introgressed versions of Vivek QPM-9 and its parental inbreds

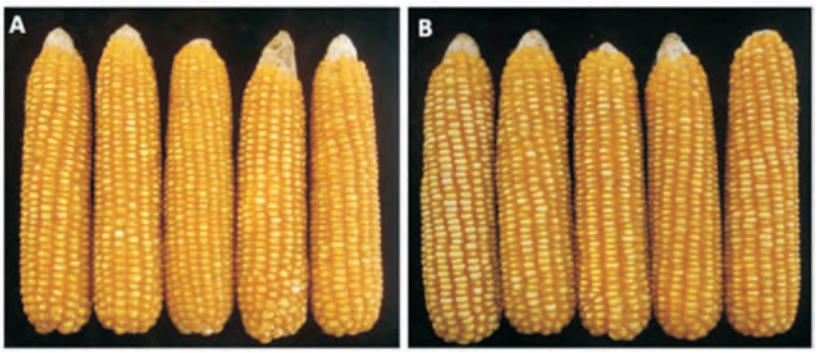

Fig 12. Ear- and grain- characteristics of the original and reconstituted version of hybrid. A: Vivek QPM-9; B: Improved Vivek QPM-9.

As a first on analyses of genetic variability and inheritance of kernel micronutrient (Fe and Zn) concentrations of maize in India, through two experiments, a total of 59 diverse inbred lines (including 46 non-QPM and 13 QPM lines) were evaluated at four environments. The experiments revealed: (i) significant variability for kernel Fe and Zn concentrations in the maize genotypes; (ii) influence of environment, genotype × environment as well as genotype × location interactions on kernel micronutrient status; and (iii) the potential of QPM genotypes for enhanced kernel Zn content. The overall ranges for kernel Fe and Zn were 6.50-43.20 mg/kg (mean: 23.38) and 14.05-49.67 mg/kg (mean: 28.31), respectively, across the environments. Stability analysis of kernel micronutrients indicated significant effects of genotypes, environments, genotype x environment as well as genotype x location interactions (Chakraborti *et al.,* 2011). Inheritance of kernel micronutrient concentrations was analyzed through: (i) analysis of a 7 x 7 full-diallel set at Delhi and Hyderabad; and (ii) generation mean analyses of crosses between two contrasting kernel micronutrient traits. Diallel analysis indicated the importance of both additive and non-additive gene action for the kernel micronutrient traits. Generation mean analysis of kernel micronutrient traits was undertaken using six generations in two sets (CM145 × V334; CM 128 × V340), revealing the importance of both additive and non-additive gene actions on target traits. However, additive gene action was of relatively higher magnitude compared to the dominance for kernel Fe, while the dominance component was greater than the additive for kernel Zn. Additive × dominance and additive x additive components played an important role in expression of kernel Fe and Zn concentrations, respectively (Chakraborti et al. 2010). Phenotyping of kernels using inductively coupled plasma mass spectrometry (ICP-MS) revealed considerable variability for kernel minerals concentration (iron: 18.88 to 47.65 mg kg^{-1}; zinc: 5.41 to 30.85 mg kg^{-1}; manganese: 3.30 to17.73 mg kg^{-1}; copper: 0.53 to 5.48 mg kg^{-1}) and grain yield (826.6 to 5413 kg ha^{-1}). Significant positive correlation was observed between kernel iron and zinc within ($r = 0.37$ to $r = 0.52$, $p < 0.05$) and across locations ($r = 0.44$, $p < 0.01$). Variance components of the additive main effects and multiplicative interactions (AMMI) model showed significant genotype and genotype × environment interaction for kernel minerals concentration and grain yield. Most of the variation was contributed by genotype main effect for kernel iron (39.6%), manganese (41.34%) and copper (41.12%), and environment main effects for both kernel zinc (40.5%) and grain yield (37.0%) (Mallikarjuna et al. 2016). Studies on Genetics and Genomics of Kernel Iron and Zinc in Maize (*Zea mays* L.) identified several candidate genes for iron and zinc accumulation such as *OPT, metal binding protein like, nramp3* and *NAS.*

Genetic analysis of yield, yield attributes and micronutrient concentration and their stability analysis in pearl millet (Pennisetum glaucum (L.) R. Br.) revealed genotype ICTP 8203 Fe was stable over environments for grain iron content. Genotype ICMR 07999 x IPC 1518 was found to be widely adaptable for plant height and 1000-grain weight. Hybrid IPC 1518 x PPMI 701 was also consistent in its performance over locations for 1000-grain weight. For single panicle weight, hybrid ICMR 07999 x PPMI 295 and ICMR 07999 x PPMI 701 were observed to be stable in its performance over environments. Thus, this study helped to identify hybrids that are widely adaptable for more than one trait. Genotypes identified with desired nutritional quality components are: High iron content: PPMI 295 (140.05mg/kg), PPMI 893 (131.75 mg/kg), PPMI 162 (130.65 mg/kg). High zinc content: PPMI 721 (150.80mg/kg), 189 B (126.30 mg/kg). High protein: PPMI 721 (12.37%). High oil content: PPMI 69 (4.43%). High lysine: PPMI 654 (356 mg/100g). High tryptophan: PPMI 872 (338.5 mg/100g). Low phytic acid: PPMI 775 (0.53 g/100g). High reducing sugar: WGI 145 (3.56%). Such genotypes will be valuable materials for breeding program improvement of quality and yield.

Earlier reported SNPs (*ea1ea2*) were validated in LES 39 (zero erucic acid variety) and further used for MAS. SNPs converted to Sequence Polymorphism Derived (SPD) markers were able to differentiate between the high and low erucic acid parents. The LES 39 specific 591 SPD marker was selected for identifying the low erucic acid segregants in the F_2 population (Singh *et al.* 2015). All the plants positive for the LES 39 specific SPD marker had no (zero) erucic acid. The present study is an important step in marker assisted breeding of zero erucic acid varieties in Indian mustard. To tag genes for low glucosinolates, simple sequence repeats (SSR) were used in RILs developed by using two contrasting parents for total glucosinolates. SSR marker Ni4H03 from 'B' genome has been tagged at a distance of 23.8 cM and At3g63420 from 'A' genome has been tagged at a distance of 37.3cM (Pushpa et al. 2016). Effect of selfing on oil quality traits under self vs. open pollinated conditions in Indian mustard revealed no significant change in erucic acid content between base material and selfed and open pollinated plants grown under nets upto two cycles of maintenance breeding, whereas, in all four low erucic acid varieties there was significant increase in mean erucic acid content by 0.197% and 0.527%, between selfed and out pollinated seeds of variety Pusa Karishma, 0.245% and 0.597% in Pusa Mustard 21, 0.207% and 0.687% in Pusa Mustard 22 and 0.217% and 0.561% in Pusa Mustard 24 after first and second cycles of open pollination, respectively.

Forty-one elite lentil lines were studied for stability of grain Fe and Zn concentration across three locations and highly significant differences between genotypes, locations and genotype × location interaction was observed. The maximum mean for grain Fe concentration over the locations was obtained for L 4704 (136.91 mg/kg grain), while for grain Zn concentration was highest for VL 141 (81.542 mg/kg grain). The highest mean grain Fe and Zn was recorded at Indian Agricultural Research Institute, New Delhi (Fe-87.30 mg/kg and Zn-68.602 mg/kg). Although both micronutrients were influenced by environment, grain Fe showed more G × E interaction in comparison to grain Zn concentration. On the basis of genetic diversity for micronutrient concentration, ten crosses are suggested for hybridization purpose to obtain the transgressive segregants. The combined analysis of multi-location phenotyping and genetic diversity further suggests six potential crosses for developing micronutrient rich varieties for the future (Kumar et al. 2014).

Breeding for Quantitative Traits

The favourable alleles for yield and yield contributing traits are distributed among two subspecies i.e., *indica* and *japonica* of cultivated rice (*Oryza sativa* L.). Identification of novel favourable alleles in *indica*/*japonica* will pave way to marker-assisted mobilization of these alleles in to a genetic background to break genetic barriers to yield. A new plant type (NPT) based mapping population of 310 recombinant inbred lines (RILs) was used to map novel genomic regions and QTL hotspots influencing yield and eleven yield component traits. Parental polymorphism was conducted with 1063 markers (STMS, RGNMS, HvSSR and gene based markers and 339 found to be polymorphic. 162 polymorphic markers) were utilized to genotype the entire RIL population. Twenty three markers showed segregation distortion from expected 1:1 segregation ratio. A

linkage map was constructed by using Mapmaker/EXP v3.0 with 126 markers data of 310 RILs. The total map length was estimated to 2023.1 cM with an average marker interval of 19.64 cM ignoring eleven large genetic intervals on eight chromosomes. The average physical length per cM genetic distance was estimated to be 169.97 Kb. Marathi et al. (2012) identified major quantitative trait loci (QTLs) for days to 50% flowering (R^2 = 25%, LOD = 14.3), panicles per plant (R^2 = 19%, LOD = 9.74), flag leaf length (R^2 = 22%, LOD = 3.05), flag leaf width (R^2 = 53%, LOD = 46.5), spikelets per panicle (R^2 = 16%, LOD = 13.8), filled grains per panicle (R^2 = 22%, LOD = 15.3), percent spikelet sterility (R^2 = 18%, LOD = 14.24), thousand grain weight (R^2 = 25%, LOD = 12.9) and spikelet setting density (R^2 = 23%, LOD = 15) expressing over two or more locations by using composite interval mapping. The phenotypic variation (R^2) ranged from 8 to 53% for eleven QTLs expressing across all three locations (Fig. 13). 19 novel QTLs were contributed by the NPT parent, Pusa1266. 15 QTL

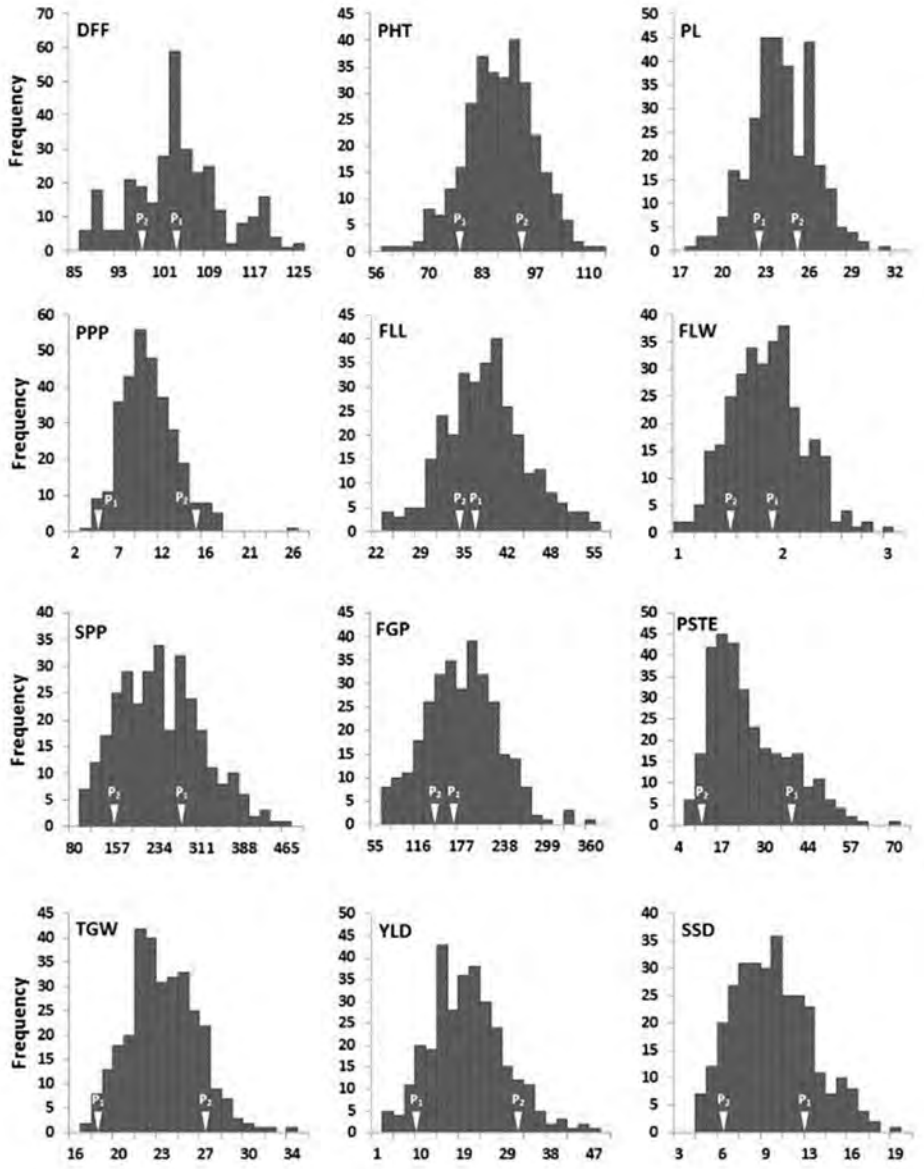

Fig. 13. Frequency distribution of Pusa 1266 × Jaya derived recombinant inbred lines for twelve traits at RBGRC, Aduthurai during *kharif*, 2006 Parental trait means are indicated by arrows

hotpots on eight chromosomes were identified for the correlated traits. Six epistatic QTLs effecting five traits at two locations were identified. A marker interval (RM3276-RM5709) on chromosome 4 harboring major QTLs for four traits has been identified (Marathi et al. 2012). The molecular genetic map developed using RIL population is comprehensive and will serve as a reference map for future studies. One of the RILs (No. 102) was found to be 10.6% higher yielding and 6 days earlier than national check Jaya. This genotype was nominated for testing in the Initial Varietal Trial of the All India Coordinated Rice Improvement Programme.

A mapping population of 310 recombinant inbred lines (RILs) derived from cross Pusa 1266/Jaya was used for mapping QTLs for grain chalkiness. The population showed normal distribution for rice grain chalkiness traits during all three seasons and transgressive segregants were observed for PGWC and CS. Parental polymorphism survey was carried using 1151 markers providing genome wide coverage. A total of 116 polymorphic markers were used to construct the linkage map and the total map length was estimated to be 1560.7cM with an average marker interval of 13.45cM. Two QTLs, one each for PGWC and CS, were prominently detected by composite interval mapping between a marker intervals RM 6273-RM204 on the short arm of chromosome 6. The QTL for PGWC, *qPGWC6.1* was consistently detected during all the three seasons, explaining a phenotypic variance of 17.75%, 18.79% and 12.20%, respectively. Similarly, the QTL for CS, *qCS6.1* was also consistent across three seasons, accounting for a phenotypic variation of 24.3%, 24.75% and 24.03%, respectively. Both these QTLs were found to be located in the vicinity of the genomic region harboring Granule bound starch synthase1 (GBSS1) locus. The positive allele for the reduced grain chalkiness was contributed by Jaya. Marker-QTL association influencing grain chalkiness traits identified in present study and those reported earlier, were validated in a set of 87 diverse rice germplasm lines. Two markers namely, RM324 and RM3294, which were reported linked with white core area (wca, R^2= 10.11 and Basal white (WB, R^2=6.25), respectively, showed significant association with PGWC and CS in the germplasms set. These markers can be used in MAS to develop rice varieties with reduced grain chalkiness.

A study was conducted to identify and characterize the genotype x management interaction in wheat, to identify the traits for specific adaptation and to validate the already identified QTLs for zero till and other production environments. Forty two genotypes with indication of differential adaptability were included in the study and highly significant variability was found for all traits under study with environment being the most important source of variation for yield, biomass, HI and number of tillers followed by the variation shown by genotypes for traits related to spike. Genotype x tillage seeding system contributed more than 20 per cent of variation for most of traits under study, indicating thereby existence of specific adaptation in different genotypes (Sagar *et al.,* 2016). According to GGE analysis, E1 is the most representative and discriminatory environment whereas for identification of specific adaptation E2 and E4 are better. Environments E5 and E6 are least discriminatory. Genotypes, CSW2 and HD3117 are showing specific adaptation for E1, E3, E5, and E6, whereas, CSW16, 18, 23, 25 and HD3115 for environment E4. Genotypes CTRB1813, CTFB4566, CSW35, CTRB1849, and CTRB817 observed specific adaptation for E2. In comparatively stressed year (2012-13), except biomass and height none of the other traits correlated with yield under permanent bed condition. Dendrogram constructed on the basis of markers for Rht, Vrn and Ppd alleles, pooled some of the genotypes bred under zero till in one group and PBW550 with other selections for conventional tillage in another and therefore can be used as selection criterion for specific adaptation.

Wheat varieties recommended for North Western Plain Zone were evaluated and the results showed that average annual genetic gain in grain yield was 0.88% or 25 kg ha-1 yr-1 during last 100 and more years (Gupta *et al.*, 2016). This is world level progress rate and is directly attributed to continuous and linear increment in biomass and HI. However no significant change has been observed for number of tillers per m row length. Biomass increase has been contributed mainly by phenological manipulations. Regarding physiological traits significant contributions has been made by increasing leaf area index, and decreasing trend over the years for traits like stomata / cm^2. Decreasing trend has been indicated for peduncle length and last node length as well. No trend in terms of end use quality characters was found (Fig. 14). Traditional continuous trend indicated less

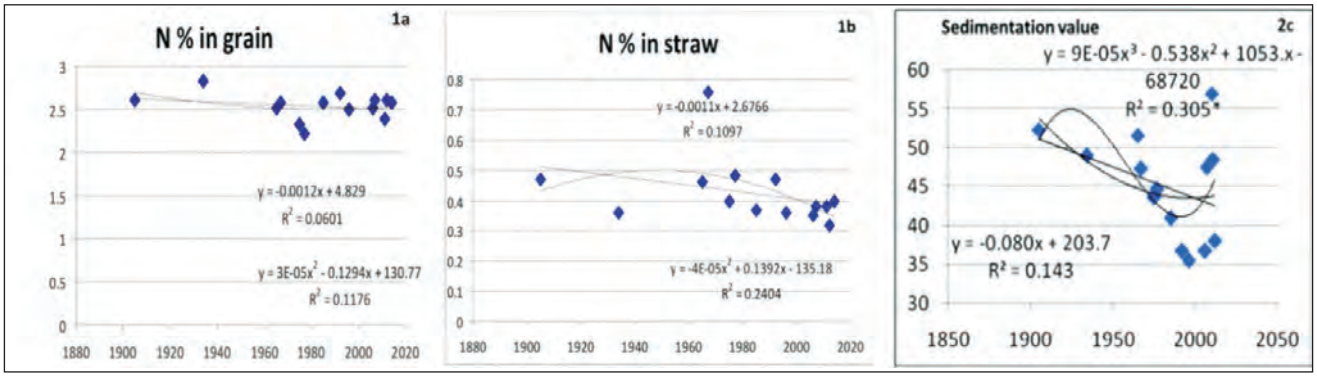

Fig. 14. Regression equations for nitrogen percentage in grain and straw weight with year of release

future scope for phenological manipulations while much possibility for HI is left untapped and has still further scope for exploitation. Plant breeding has played magnificent role and will do so in coming future.

Genetics of yield, yield components & resistance to banded leaf and sheath blight in maize (*Zea mays* L.) identified the genotypes L_4 (DMB-14) and L_8(DMB-22) were good general combiners for yield and yield associated characters. L5 x T3 (DMB-16 X DMB-28) and L9 X T5 (DMB-23 X DMB-30) with high sca effects and better parent heterosis were identified. L_2 X T_2 (DMB-9 X DMB-27) showed high degree of tolerance to BLSB with high productive potentiality. Genetic analysis of sugary trait and yield components in sweet corn (*Zea mays* var. *Saccharata*) revealed that the good general combiners for yield were IPSA-6133 and IPSA-6134 and for sugar content were IPSA-6134, IPSA-6140 and IPSA-6141. On the yield basis, the best parents were IPSA-3832, IPSA-3855, IPSA-3856, IPSA-3858and IPA-3044, IPA-3107, IPA-3135, IPA-3172 among the lines and testers, respectively.

Six populations developed and maintained by the pearl millet breeding section of the division were evaluated to identify the two most promising composite populations for further improvement. Pusa-266 and pusa-383 were identified as most promising and may be the suitable source populations where population improvement may be effective. Genetic components of variance and other associated genetic parameters were assessed in the two selected populations namely pearl millet composite pusa-226 and pusa-383 following North Carolina mating Design-1 (NCD-1). It exhibited preponderance of additive variance for yield and yield components in both the populations. Therefore, population improvement may be rewarding as selection responds to additive variance. The genetic architecture and character associations in the selected populations also has been studied to guide for their improvement and to analyze the selection efficiency of three simple intra-population improvement methods applied on the selected populations. To analyze the relative efficiency of three different selection methods namely mass selection, grided mass selection and bulk sibbing, least significant difference (LSD) comparison analysis at 5% probability was carried out. LSD comparison revealed that though mass selection is numerically superior to other selection methods applied, all the selection methods are statistically equally effective in improving the characters under study in these populations.

Sixty five genotypes of were grouped into 8 distinct clusters and the clustering pattern was more distinct for seed size (medium and bold seeded) than seed type *(desi and kabuli.).* Stability analysis identified the high performing *desi* and *kabuli* genotypes with specific and wide adaptability for different yield components for their utilization in future *desi-kabuli* introgression breeding. Four breeding methods viz. pedigree method (PM), single seed descent method (SSD), bulk method (BM) and selected bulk method (SBM) were evaluated for their efficiency in producing superior genotypes. The inferences of this study indicates that the yield superiority of selected materials on the basis of percent increase of highest yielder over check the selected bulk method (SBM) of breeding was the best method over the crosses followed by bulk method (BM) and on the basis of percent increase of mean of selected plants the pedigree method (PM) and bulk method (BM) of breeding

showed equal yield superiority followed by selected bulk method (SBM). Thus, overall selected bulk method (SBM) was identified to be the best.

Evaluation of genotypes for drought and yield related traits in chickpea were done during years 2007-08. The materials used were twenty two genotypes and one check under irrigated and rainfed conditions. The *per se* performance of chickpea genotype revealed that there was a substantial variability among genotypes for all the characters except number of seeds per pod under irrigated and rainfed conditions. Based on the mean performance, DG-115 was the best for seed yield per plant and DG-110 for harvest index under irrigated condition and genotype DG-119 was the best for seed yield per plant and harvest index under rainfed condition. For relative water content (RWC %), the genotype DG-117 showed highest mean values and genotype DG-104 had highest mean values for membrane stability index (MSI %) under rainfed condition. Twenty two chickpea genotypes and one check were also studied for various genetic parameters such as heritability, genetic advance, phenotypic (PCV), genotypic (GCV) coefficient of variation and different stress indices *viz.,* drought, dry matter, plant height and yield stability index. The high estimates of phenotypic and genotypic coefficient of variation expressed as percentage of mean were observed for seed yield per plant, number of pods per plant, harvest index and biological yield. High heritability coupled with high genetic advance was observed for seed yield per plant, biological yield, number of pods per plant and harvest index under irrigated and rainfed conditions indicating the scope for improvement and genetic gain through the selection of these traits. The genotypes, DG-119, DG-114, DG-116 and DG-118 were found drought tolerant based on different drought indices parameters. The results of correlation analysis revealed that that grain yield per plant had significant genotypic and highly significant phenotypic relationship with number of pods per plant, number of seeds per pod, harvest index and biological yield per plant. Therefore, this study suggested that chickpea improvement programme could be based on these characters as selection criteria.

Two hundred ten mini-core germplasm collections of chickpea collected from ICRISAT were evaluated at the Division of Genetics, Indian Agricultural Research Institute, New Delhi under field conditions during the years 2007-08. This collection was evaluated for nine qualitative characters. In this study most of the germplasm was semi-spreading type, pink flower, medium seed size, brown seed colour, rough seed surface and desi type of seed shape was reported. Correlation coefficient suggested that selection of plants with high or more plant height, total number of branches per plant, number of pods per plant and number of seeds per pod would show result in progenies with high seed yield potential in chickpea.

A recombinant inbred line mapping population developed from the cross Pusa Dwarf x H2001-4 was evaluated over two environments and was genotyped using SSR markers to identify QTLs for seed yield attributing traits (Fig. 15). This was the first ever RIL mapping population generated and evaluated for the QTL mapping of seed yield related traits in pigeonpea. A genetic map was constructed with 52 SSR (20 ASSR and 32 HASSR) markers which were grouped into 11 linkage groups covering a total length of 1397.2 cM. Two QTLs were identified for days to flowering *viz.,* qFL2 and qFL8 explaining 17.6 % and 8.4 % of phenotypic variance, respectively (Geddam et al. 2014). For plant height, two QTLs qPH1 and qPH2 were identified with R^2 value of 19.2 % and 9.3 %, respectively. The validated marker- QTL association is of immense

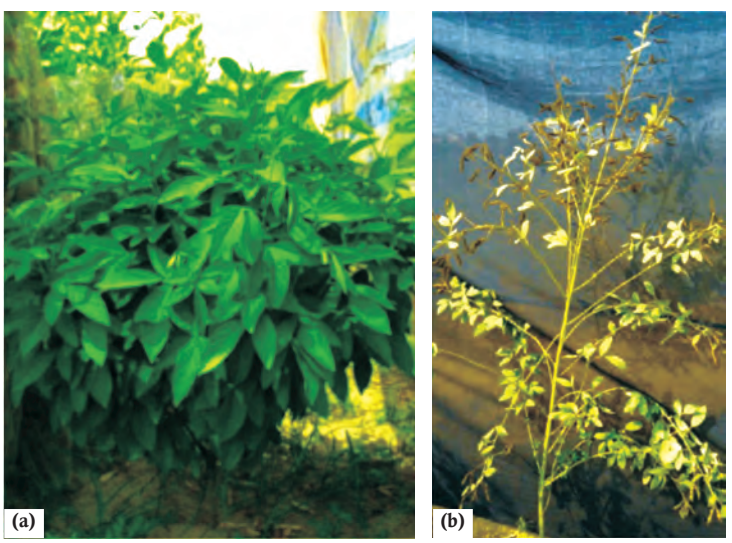

Fig. 15. Field view of the pigeonpea genotypes used as parents (a) Pusa Dwarf (b) H2001-4 in QTL mapping studies

importance for improvement of varieties in pigeonpea for earliness and plant type characteristics. The QTLs identified in the present study might be highly useful in marker assisted selection for the development of early maturing and high yielding genotypes in pigeonpea (Geddam et al. 2014).

The six genetrations (P_1,P_2, F_1, F_2, B_1 and B_2) of eleven primary cross combinations of Indian mustard *Brassica juncea*, viz., NPJ-102 x RGN-48, NPJ-102 x Laxmi, NPJ-102 x Pusa Jaganath, NPJ-102 x BEC-144, pusa Agarni x Laxmi, Pusa Agarni x pusa Jaganath , RGN-48 x Laxmi, RGN-48 x BEC-144, Ornamental Brassica x Pusa Jaganath, Pusa Jaganath x Ornamental Brassica, Pusa Jaganath x NPJ-50 were studied for seventeen characters namely days to 50 per cent flowering, days to maturity, plant height, point to first branch, number of primary branches, number of secondary branches, length of main shoot, point to first siliqua, number of siliqua on main shoot, siliqua length, seeds per siliqua, seed yield per plant, biomass, harvest index, leaf colour, flower colour and seed colour on single plant samples. All the generations of each cross combination were evaluated in a randomized block design with three replications. The mean of six generations was subjected to scaling test, joint scaling test to determine epistasis and genetic parameters m, d, h, i, j and l were evaluated along with heterosis studied as well as inheritance studies for leaf colour, flower colour and seed colour. An epistatic digenic model including all types of interactions played a major role for all the primary cross combinations. The study revealed the importance of both additive and non-additive type of gene action for all the characters studied. Duplicate epistasis played a relatively greater role than complementary epistasis suggesting reciprocal recurrent selection for development of a superior variety. The F_2 segregation pattern of all the primary crosses studied for inheritance study revealed that digenic ratio of 15 : 1 (Brown : Yellow seed coat colour), monogenic ratio of 3 : 1 (Yellow : Creamy flower colour) and monogenic ratio of 3:1 (Purple : Green leaf colour) plants.

Hybrid Breeding

Efforts were made to identify potential restorers in aromatic rice germplasm using molecular markers for fertility restorer genes in WA CMS, to assess their fertility restoration potential and extent of heterosis in test cross hybrids. Genotyping with gene linked markers, RM 3873 and RM 6100; and candidate gene based markers, DRRM-RF3-10, DRRM-RF3-5 and DRCG-RF4-14 for two genes, Rf3 and Rf4 restoring fertility in WA CMS lines, and assessing the pollen and spikelet fertility in test crosses with a set of 59 diverse genotypes, the efficacy of these markers in identifying potential restorers was estimated (Fig. 16). Overall, the candidate gene based markers were found to be efficient in identifying potential restorers with DRRM-RF3-10 and DRCG-RF4-14 showing 90% and 88.2%, respectively as compared to gene linked markers, RM 3873, RM 6100 showing 73.2%

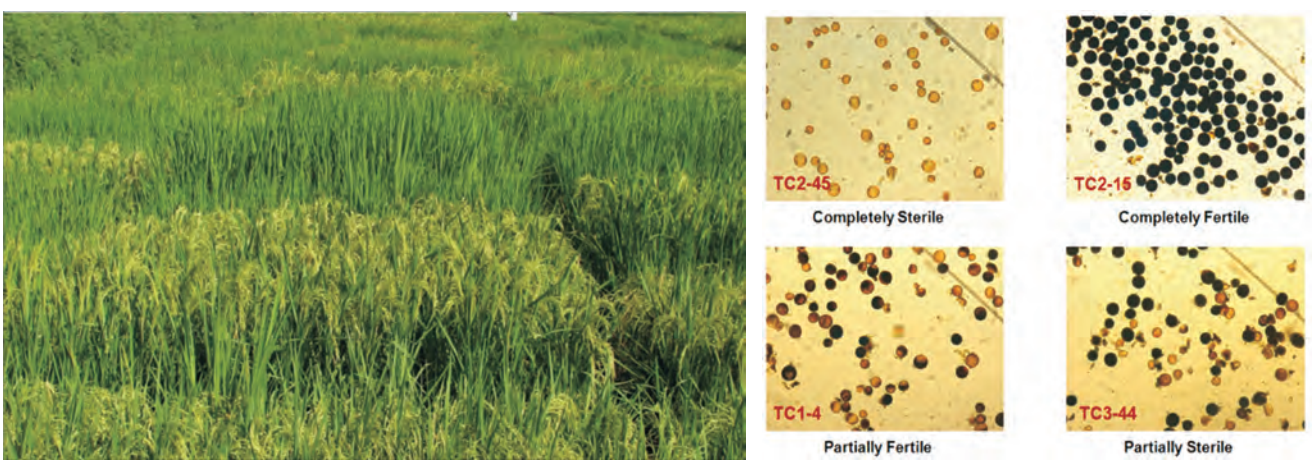

Fig 16: (a) Agro-morphological evaluation of 59 rice genotypes; (b) Differences in pollen fertility observed among the genotypes being used in the study

and 82.2%, respectively for Rf3 and Rf4. Standard heterosis in the 90 test cross hybrids ranged from -100 to 120.59%. Out of the 59 genotypes assessed, 45 restorers were identified as potential restorers and 18 of them produced heterotic test cross hybrids showing more than 20 % heterosis over standard hybrid check, Pusa RH10. Besides this, a set of 14 genotypes have been identified as potential maintainers. These genotypes can also be utilized in parental line improvement for developing heterotic rice hybrids.

Analysis of genetic diversity and heterotic potential in popcorn (*Zea mays everta*) inbred lines has been identified Three crosses namely, IPPA-20 x IPPA-37, IPPA-33 x IPPA-37 and IPPA-35 x IPPA-37 as the potential hybrids in popcorns for popping quality as well as yield. Inbred lines differing for starch components were taken for estimation of heterosis and combining ability in maize. Among *ae* mutant inbreds IPAM2, IPAM3 and IPAM6 exhibited good general combining ability for most of the important characters. Similarly IPAW1, IPAW2, IPAW6 exhibited good general combining ability for most of the characters. Crosses IPAM4×IPAM7, IPAM3×IPAM5 and IPAM5×IPAM7 were identified as the best hybrids with high amylose content having high SCA. Similarly, crosses IPAW2×IPAW9, IPAW5×IPAW8 and IPAW3×IPAW9 were identified as the best hybrids with higher amylopectin than normal corn and high SCA.

Analysis of Restorer and maintainer relationship of some Elite Inbred Line VIS-À-VIS A_4 and A_5 Cytoplasm in Pearl Millet [*Pennisetum glaucum* (L) R. Br.] showed that all the seven cytoplasmic male sterile lines (3A1; 2-A4 and 2 – A5 cytoplasm) coming from different cytoplasmic sources are capable of producing new superior hybrids. Combining ability analysis identified A_1 cytoplasm male sterile lines (ICMA 98333, 91444, 88004, 89111 and 95111) are good combiners for plant height. Similarly for days to 50% flowering and days to maturity the male sterile A_1 cytoplasm lines (ICMA 92111, 89111, MS 843 A and 5141 A) were found to be good combiners. The male parent PPMI 761 identified of completely restoring the fertility in two male sterile lines 843A and ICMA 93333 which belong to A_1 cytoplasm. Inheritance of fertility restoration in A_1 based Cytoplasmic Genetic Male Sterility in pearl millet [*Pennisetumglaucum* (L). R. Br.] identified involvement of the digenic supplementary or an epistasis with recessive gene action for these crosses 576A/IPC 1518, 411A/IPC 1518, 576A/ICMR 06111 and 411A/ICMR 06111.

In Pigeonpea (*Cajanus cajan* (L.) Millsp.) the parental combinations of A and R lines viz., ICPA2047 X ICPR2740, ICPA2043 X ICPR3472, ICPA2043 X ICPR4013, MS10A X AKPR9, MS10A X AKPR100 X GT288A X AKPR417 were identified as most distinct and divergent. Therefore, identified hybrid parental combination could be effectively used as candidate parental genotypes in hybrid development programme. Characterizing 32 soybean lines having differential fertility under different photo-period and temperature situations indicated pollen fertility as low (0.38 to 22.05%). One line (TNAUS56) showed no deviation in days to flowering and the percentage gain in response rate was found to be very low (2.38).

Genetic diversity among 44 Indian mustard (*Brassica juncea*) genotypes of indigenous and exotic origin was studied using 134 molecular markers (SSR) and 12 yield and yield contributing traits. Jaccard's similarity coefficients and Manhattan dissimilarity coefficients grouped the genotypes into four clusters as per their pedigree and origins. However, the correlation between phenotypic and genetic distance matrices was very low (r = 0.11). Principal Coordinate Analysis revealed that the grouping of the genotypes based on SSR marker data is more informative and realistic than that of phenotypic data. To determine the relationship of parental distances estimated from phenotypic traits and SSR markers with F_1 performance, average heterosis and heterobeltiosis in Indian mustard (*B. juncea*) involving same set of genotypes was taken. Jaccard's genetic distances (JD) and Manhattan genetic distances (MD) were taken as a criterion to classify the genotypic pair in to four diversity groups *viz*., high, intermediate high, intermediate low and low. Placement of higher number of significantly better hybrids is in extreme diversity groups created using JD values, while, it was higher in intermediate diversity groups generated through MD values. Low regression values were observed between JD among genotypic pairs and mean performance (R^2 = 0.02), average heterosis (R^2 = 0.046) and heterobeltiosis (R^2 = 0.15). Similarly, low regression values were also observed between MD among genotypic pairs and mean performance (R^2 = 0.033), average heterosis (R^2 = 0.046) and heterobeltiosis (R^2 = 0.009). The slope of linear

regression curve, placement of hybrids on the plot and low regression values in all the cases revealed that there is no significant association between genetic distances and hybrid performance.

Drosophila Genetics

Wnt family of signaling molecules plays many important roles during development, adult homeostasis and maintenance of stem cells. Any perturbations in Wnt genes leads to cancer and auto immune disorders in human. Drosophila has seven Wnt genes, out of which four Wnt genes *DWnt4*, *wingless* (*wg*), *DWnt6* and *DWnt10* are clustered on left arm of second chromosome and their synteny is conserved for more than 500 million years. Very little information is available for how these clustered Wnts interact or co ordinate their expression during different developmental stages. To address this, novel mutations have been isolated for *DWnt4* using EMS mutagenesis and were validated using genetic complementation, DNA sequencing and qPCR. These mutations are first ever null mutations in *DWnt4* gene, showing reduced denticles during ventral ectoderm pattering of embryos. There is loss of *Armadillo* in the mutants of *DWnt4* indicating that *DWnt4* may not be functioning in canonical pathway, while expression of *Engrailed* is not changed, indicating that *DWnt4* is not interacting with *Engrailed* but rather involved in cuticle patterning with some other gene which need further investigation. Though the null alleles isolated in lab are not showing any sterility phenotypes, but germline clones of *DWnt4* mutant allele are arrested at stage 6 of ovariole development (Fig 17). Genetic studies using *DWnt4* and *wingless* (*wg*) mutations revealed that *DWnt6* and*DWnt10* are not expressed in ventral ectoderm of embryo but they are able to rescue the defects in cuticles in absence of *DWnt4* or *wg* (Fig. 18). Knockdown

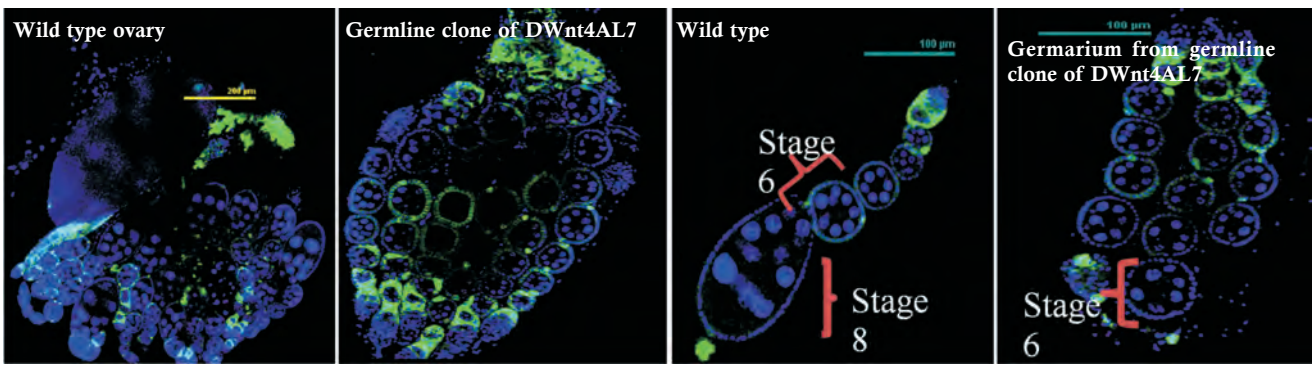

Fig 17: Germarium from DWnt4 mutant germline clone Ovarioles are arrested in stage 6 of oogenesis

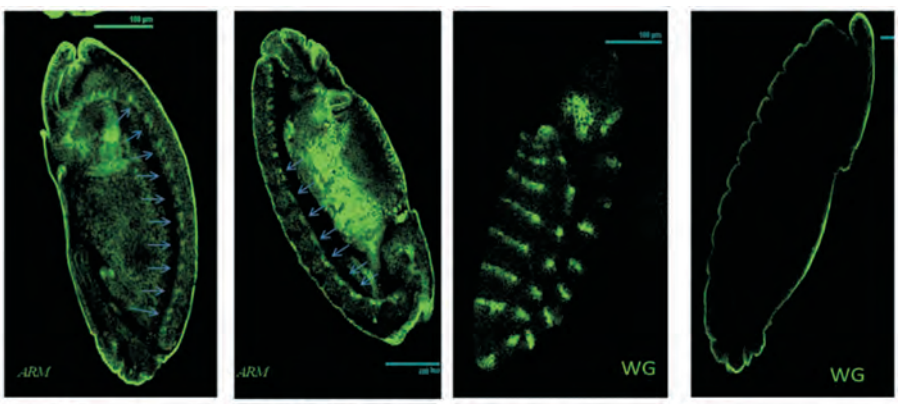

Fig 18: *DWnt6* and *DWnt10* are able to rescue *DWnt4* with canonical Wnt pathway in presence of *Armadillo* (*Arm*)and by suppressing *Wingless* (*Wg*) expression

of *DWnt6* and *DWnt10* transcripts in *DWnt4* and *wg* epidermal/cuticular mutants resulted in increased defects in cuticles. Expression studies involving immunostaining of embryos of mutants from *DWnt4* where *DWnt6* or *DWnt10* are over expressed suggests that *DWnt6* and *10* are able to rescue theses mutations by suppressing *wg* and activating Armadillo which in turn starts canonical Wnt pathway.

Conclusion

The student's research work carried out at the Division of Genetics emphasized on both basic and applied aspects of crop improvement. Starting from genetic diversity studies to mapping of various genes related to biotic and abiotic stresses were carried out using molecular marker technology. Emphasis was also given to hybrid breeding in different crops like rice, wheat and pearl millet. SEveral genes for biotic and abiotic stresses were pyramided to develop products in rice, wheat and maize. Molecular markers also have been succesfully deployed that led to rapid development cultivars. Besides, yield improvement, breeding for improvement of nutritional quality traits were also accomplished in different crops like rice, wheat, maize, pearlmillet, brassica and lentil etc. Division of Genetics houses laboratory for Drosophila genetics where many students have undertaken research on classical genetics.

Future Perspectives

Being the forerunner in the institute, Division of Genetics will continue its research both in basic and applied aspects with an aim to enhance resilience of Indian agriculture to climate change and climate vulnerability as there is enormous effect of climate change on food security. Apart from this, emphasis is also directed towards some cutting edge technologies like genome editing, TILLING, genome-wide association study (GWAS), use of MAGIC (Multi-parent advanced generation inter-cross) populations etc. This will be largely benefiting our research programme which would lead to development of newer technologies and generation of trained manpower in the discipline of genetics and plant breeding.

Acknowledgments

We wish to thank all the Faculty Members of the Division of Genetics and Plant Breeding for extending support and cooperation in preparing the research contributions of M.Sc. and Ph.D. students made during the period of 2007-16.

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Plant Genetic Resources

Rekha Chaudhury

Introduction

The postgraduate research at PGR discipline has been addressing many relevant aspects of PGR management including A) seed storage behaviour of crop species B) farmer management of crop population structure C) monitoring the loss of genetic diversity from traditional production systems D) diversity studies in crops especially landrace populations E) Transgenics detection

Significant Student's Research Achievements

Seed storage and related research including cryopreservation

Effect of temperature and relative humidity on seed storage of senna (*Cassia angustifolia* Vahl.) ds was undertaken using relative humidities (75 %, 33 %, 11 % and 5.5 %) and temperatures (5° C, 20° C and ambient). All physiological parameters were adversely affected by high RH. Storage temperatures of 5° C and 20° C were found to be optimum. Electrical conductivity and lipid peroxidation showed increase with loss of viability. Total soluble proteins and activity of dehydrogenase and acid phosphatase decreased with storage period.

Conclusions : Senna seeds are tolerant to ultra desiccation and can be used successfully used for cost effective conservation of this species. In future, ultra desiccation technique can be standardized for cost effective conservation of senna seeds.

Characterization and cryopreservation of genetic resource of *Citrus macroptera* collected from Tripura and Mizoram was conducted. Thirty two accs collected were characterised using morphological and Random Amplified Polymorphic DNA (RAPD) markers. Both morphological and molecular analysis showed a high degree of variation among analyzed accessions. Basic studies on seed physiology, seed storage behaviour, cryopreservation of embryonic axes and characterization were undertaken. The cryopreservation of embryonic axes of *C. macroptera* was attempted using air desiccation-freezing, vitrification and encapsulation-dehydration

Conclusions : Seeds of *C. macroptera* exhibited intermediate seed storage behaviour. Successful cryopreservation was achieved using all these three techniques.

Studies on water and oil contents in relation to seed cryobiology in Pilu (*Salvadora oleoides* Decne.) was done. Vaious aspects of germplasm management including studies on seed physiology, seed storage behavior, cryopreservation of seeds and embryonic axes, electrical conductivity, oil content and germplasm characterization was done. The seed storage behavior of *S. oleoides* was ascertained and cryopreservation of their embryonic axes was attempted using air desiccation-freezing method. Accessions were cryopreserved in the cryogenebank for long term conservation.

Conclusions: Seeds revealed recalcitrant seed storage behavior with short longevity however cryobanking could be achieved.

Optimizing seed recovery conditions in terms of physiological and morphological aspects after cryopreservation was studied in *Jatropha curcas, Salvadora oleoides* and *Citrus sinensis*. The effect of cooling and thawing and repeated

cooling and thawing on long term storage of germplasm was studied. Seeds and embryonic axes of different crop groups and of different seed storage behaviour were retrieved from cryostorage and their viability was retested after long term storage (7-24 years of storage).

Conclusions : Liquid nitrogen along with conventional method was found beneficial to break the dormancy of hardseeded species to some extent. By determining the seed coat ratio, the storage behavior of the seeds can be fairly predicted. On the basis of seed coat ratio determined for 7 different species with different seed shape and sizes, the accessions were classified into orthodox, intermediate and recalcitrant seeds.

Dynamics of on-farm management of crop landrace populations in traditional production systems

Population genetic studies using STMS primers on hullless barley (*Hordeum vulgare* L.) landraces from North-western Himalayas for promoting their use and *in situ* conservation on-farm demonstrated population structure of landraces from higher Himalayan ranges representing three administrative regions, Jammu & Kashmir, Himachal Pradesh and Uttarakhand. This classified the accessions into two major groups. Populations from Jammu & Kashmir were grouped together at sub-cluster level. On-station trials revealed considerable diversity for quantitative morphological and agronomic traits.

Conclusions : Scientific and institutional challenges for on-farm management of barley landrace diversity in traditional production systems and various "adding value" options for farmers to local crop diversity were documented.

In an another study, farmer management of population structure in traditional production systems of parts of Western Ghats of Kerala and Tamil Nadu states in context of on-farm management of rice landraces was also undertaken.

Population structure of rice landraces under farmer management; range and pattern of landrace diversity based on morphological characterisation, and variations due to environmental adaptations in different populations of the same named landrace from parts of Indian Himalayas were studied. The STMS markers fully differentiated the inter- and intra-population diversity of rice landraces from higher Himalayan ranges, both north-western and north-eastern regions.

Conclusions: The clustering pattern in rice landraces revealed clear association between geographical diversity and genetic diversity. The diverse and highly differentiated rice landraces found in the Himalayas can be primarily attributed to the vast eco-geographical diversity prevailing in the region. Investigating the population genetic structure can therefore help monitor change in diversity over time and space, and also devise a rational conservation plan.

Diversity analysis

Molecular profiling of Indian mango cultivars (255) and analysis of their dispersal pattern and geographical differentiation was done. Germplasm from 20 districts representing 6 major ecogeographical regions using 24 microsatellite primer pairs resulted in comparison of a total of 226 alleles with a mean of 9 alleles per locus. Results indicated that the most divergent cluster contained some wild types from south west India. Further, 25 cultivars investigated for intra-cultivar variation showed low to moderate variations.

Amaranths (*Amaranthus* spp.), was investigated for morphological and molecular (using STMS primer pairs) diversity analysis. Wide variations were recorded for the 13 morphological and three quality traits. Greater diversity was recorded for *A. hypochondriacus* accessions.

Conclusions : Promising accessions could be identified for yield and quality parameters for use by breeders in varietal improvement programme. The STMS primers were effective in differentiating accessions particularly at species level.

Assessment of genetic diversity in small and medium-grained aromatic rice (*Oryza sativa* L.) landraces/ varieties from Orissa using morphological, quality and molecular traits was done through morphological and molecular characterization. High and comparative levels of phenotypic variations were found among genotypes for majority of the traits studied. Correlation study among yield and quality traits revealed that L/B ratio had high positive correlation with grain length and 100-grain weight. Number of grains per panicle had negative and significant correlation with grain length and L/B ratio. Strong aroma genotypes had lower number of grains per panicle and number of effective tillers per plant.

Conclusions: Molecular characterization of aromatic rice genotypes using STMS markers generated high polymorphism and suggested their suitability for molecular diversity analysis of rice genotypes. The STMS markers were also effective in discriminating the aromatic rice genotypes on the basis of their local adaptations.

Dynamics of landrace diversity in 33 rice landraces from parts of Western Ghats of Tamil Nadu and Kerala States was studied alongwith a set of salt tolerant "pokkali" landrace populations using STMS primers. The UPGMA dendrogram classified the Western Ghat accessions into five groups. Clear association between genetic diversity and geographical origin was revealed. Similarly, STMS marker diversity in "pokkali" landraces assembled mainly from Ernakulam and Alappuzha districts of Kerala was assessed.

Conclusions: The differences in the agronomic potential of rice landraces suggested enhanced use of desired types for certain marketing interventions.

Molecular analysis of genetic variation and population differentiation in 44 landraces of aromatic rice (*Oryza sativa* L. sub-species *indica*) of Uttar Pradesh, Manipur and Odisha was done. A high amount of genetic variation was observed between the landraces. Unique/rare alleles could be used for DNA fingerprinting and differentiation of individual/group of genotypes from the rest. A large number of genotypes (287) exhibited two alleles per genotype, while 14 genotypes showed amplification of three alleles per genotype. The landraces were discriminated from each other in the dendrograms constructed using Nei and Li, and Jaccard's similarity and Nei's distance coefficients.

Conclusions: The presence of two or more alleles per locus within an accession reveals that some of the landraces are heterogeneous and the heterozygote genotypes are present in the accessions analyzed. The findings of present study are more relevant to rice improvement specially the non-Basmati aromatic types as there is large amount of genetic variation is present in the accessions analyzed.

Genetic resource diversity in *Luffa* using morphological, biochemical and molecular markers was studied Six taxa (5 species) of cultivated and wild were investigated for different morphological, quality traits and molecular characters. A set of 80 accessions from Bihar, Karnataka and Andhra Pradesh were characterized for 33 morphological traits. The results showed high degree of variation among analyzed accessions using morphological, quality traits and molecular characters and indicated vast genetic differences among the genotypes available in populations within the cultivated and wild taxa across regions in India.

Differential reaction of cowpea genotypes to pulse beetles under artificial seed infestation and biochemical basis of resistance to identify the sources of resistance against *Callosobruchus chinensis* and *C. maculatus* and to understand the physical and biochemical basis of resistance. Significant differences among the accessions were observed in terms of number of eggs laid, development period, adult emergence, number of emergence holes, weight loss and growth index (GI) of two species. Based on GI, a few resistant and moderately resistant accessions to both pulse beetles, *C. chinensis* and *C. maculates,* were identified. Significant differences among the cowpea accessions were observed for all the physical and biochemical parameters. Correlation coefficients were worked out between growth index (GI) of pulse beetles and physical and biochemical parameters. The accessions IC107466 and IC106815 identified as resistant to pulse beetles could be used in breeding programme for development of resistant cultivars.

Conclusions: However, the relationship between bruchids and their host plant is complex and not dependent on a single mechanism, but that there are overlaps between the morphological and biochemical bases of resistance.

Molecular and morphological analysis of genetic variation and population differentiation in two local maize (*Zea mays* L.) landraces namely Malan and Sathi of Mewar region of Rajasthan was studied. A wide range of phenotypic variability was exhibited for most of the quantitative and qualitative traits studied in 68 accessions of these landraces. Malan and Sathi landrace accessions were classified into four groups based on cluster analysis, while in PCA approach two groups were formed with nine outlying accessions.

Conclusions : The estimates for phenotypic coefficient of variation were higher than the genotypic coefficient of variation. Significant variability was observed at morphological and molecular levels which can further be utilized in crop improvement programmes. Promising landraces accessions identified in this study can be used in maize improvement programmes.

Molecular genetic diversity analysis of 68 genotypes of pomegranate (*Punica granatum* L.) field genebank collection using ISSR markers was undertaken using morphometric and ISSR markers. DNA fingerprint pattern of 68 pomegranate accessions based on 43 ISSR marker loci was successfully generated.

Conclusions : The findings of the study suggested the existence of high variability in the indigenous semi-feral germplasm accessions maintained in the pomegranate field genebank.

Genetic variation in 48 genotypes of bread wheat (*Triticum aestivum* L.) was analysed for genetic variation using microsatellite markers especially for terminal heat stress, an important climatic factor reducing the yield in wheat. A panel of The 22 primer-pairs amplified a total of 95 alleles with an average of 4.3 alleles per locus. These unique/rare alleles could be used for DNA fingerprinting and differentiation of individual/ group of genotypes from the rest. A partial region of heat-shock protein gene of wheat, *HSP101* was amplified and sequenced from 48 genotypes of bread wheat. The multiple sequence alignment revealed three types of *TaHSP101* alleles and SNPs at numerous places, which needs to validate using a more genetically diverse core set of bread wheat germplasm.

Conclusions: The findings of present study are more relevant to the trait-specific wheat improvement especially for heat-stress tolerance.

There is rich genetic diversity available in sacred or holy basil (*Ocimum tenuiflorum* L.) grown in different parts of the country. A set of 49 accessions was investigated for diversity analyses using morphological and molecular tools.. Characterization data of thirty-two morphological traits during vegetative to reproductive cycle of plant growth viz. qualitative (18 traits) and quantitative (14 traits) were recorded. Analysis showed high degree of variation among vegetative characters (habit, leaf) as compared to reproductive characters (flower, seed) indicating high level of diversity represented among the populations from different areas. Seed characters exhibited very little variations. In cluster analysis, the accessions formed two groups, the major group with 39 accessions mainly from Uttar Pradesh and from Assam.

Conclusions: Results of the analysis from morphological and molecular tools indicated vast genetic differences among the genotypes available in populations from different phytogeographical regions.

One of the major constraints in storage of lentil is damage caused by pulse beetles (bruchids). Wild lentils possess substantial genetic diversity are potentially important source of genetic variation for improvement of cultivated lentil. Hence, different *Lens* spp. were analyzed for genetic diversity assessment using PCR based molecular markers and evaluated for differential reaction to *C. chinensis* to identify the sources of resistance. Genetic diversity was assessed in 50 accessions of different *Lens* species using ISSR markers. The collection included accessions of one cultivated species, *L. culinaris* and six wild species, *viz.*,

L. culinaris subsp. *odemensis, L. culinaris* subsp. *orientalis, L. orientalis, L. nigricans, L. lamottei* and *L. ervoides.* 23 ISSR primers amplified a total of 368 bands with an average 16 bands per primer.

Most of the cultivated *Vigna* species have a narrow genetic base resulting in limited marker polymorphism within the germplasm. Due to this major limitation, most of the genetic linkage maps in *Vigna* species have been constructed using inter-specific or inter-subspecific crosses to increase the level of polymorphism. The use of Conserved Orthologous Sets (COS) markers as a starting point for marker development was motivated by their expected low copy number in the genomes of various legume species for which genomic information is not abundant. It was concluded from the study that, i)Expressed sequence tags (ESTs) are best source for developing COS markers in *Vigna* species as whole genomic sequence data in these species is not available. The newly identified COS markers shown orthology in *V. radiata* as revealed from amplification profile. This infers that there is scope to find more and more *Vigna* COS markers which help in comparing genomic information across species; ii) Polymorphism in intronic region is noteworthy as revealed by intron spanning COS markers (such as CPOX-2, SHMT-1 and SUSY-8) showing varied Indel (Insertions-Deletions) sites. This depicts that, there exists polymorphism in intron length among *Vigna* species which can further be explored for better understanding genome structure; iii) Statistical findings from DNA sequence data of three *Vigna* species (V. radiata, V. mungo, V.umbellata) suggests that there exists orthology as most of the genomic region in species were having equal level of conservation threshold as 1; iv) The Tajima's D and Fu Li's D tests were in the range of negative values and though this did not indicate any statistical significance, but can be verified further increasing large number of representative sequence datasets in each *Vigna* species. The information generated from this study helps *Vigna* researchers to identify the most critical attributes COS markers in understanding genome structure and comparative mapping of genes for desired traits.

Variability studies in morphological and biochemical characters of selected germplasm lines of faba bean (*Vicia faba* L.). A total of 43 selected accessions comprising of 23 indigenous and 20 exotic collections with two national checks (Vikrant and PRT-12) were taken up for characterization of agro-morphological and biochemical traits using minimal descriptors and descriptor states. These parameters shows good range of variation for days to 50% flowering, days to 80% maturity, plant height, number of branches per plant, number of leaflets per leaf, pod length, pod width, number of pods per plant, number of seeds per pod, hundred seed weight, seed yield per plant, seed characters and for biochemical parameters. Under the study, some of the indigenous (HB 20, HB 45, HB 47, HB 85, HB 175, HB 188, HB 608, RFB-4) and exotic (EC107842, EC 108908, EC 243608, EC 243794, EC 331564, EC 591792) lines showing promising results over the check varieties for both morphological and biochemical parameters have been identified as donor to use in faba bean crop improvement.

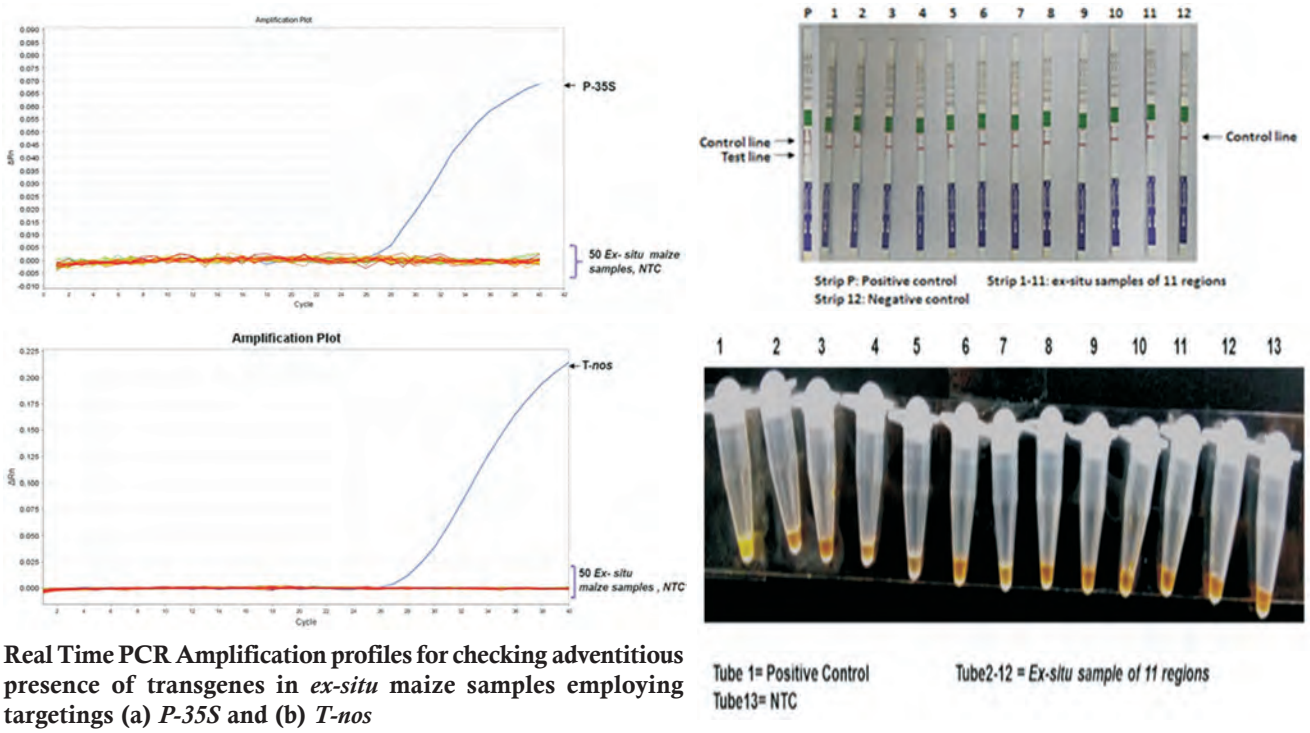
Genetic diversity was analyzed in 96 cotton genotypes which include the groups of released varieties (old and new), hybrids (*Bt* and non-*Bt*) and parental lines using SSR and ISSR markers. The 31 SSR and 4 ISSR primers generated 75 and 23 markers, respectively. The Polymorphism Information Content (PIC) value of SSR and ISSR primers was found to be 0.469 and 0.784, respectively. On the basis of statistical parameters such as % polymorphism, PIC, Resolving Power (RP) and Marker Index (MI) the primers namely, MUCS 400, JESPR 1197, BNL 3594, JESPR 153, JESPR 204, MUCS 164, BNL 3649 and NAU 1255 were found to be useful. Cluster analysis revealed two main clusters for the *G. hirsutum* genotypes at 65% genetic similarity. Genetic diversity was found to be highest within the 'Non-*Bt*' group, whereas, it was found to be the lowest for the '*Bt*' group genotypes. A comparison of genetic dissimilarity values between '*Bt*' and 'Non- *Bt*' genotypes showed that the absolute dissimilarity value for 'Non- *Bt*' is higher as compared to the '*Bt*' genotypes. However, these differences are statistically non-significant as revealed by the Student's T-test.

Strategy for Monitoring Adventitious Presence of Transgenes in Maize Collections Employing Different GM Diagnostic Assays

Protein- and DNA-based methods were employed to check specific targets in certified reference materials (CRMs) of selected GM maize events. Protein-based Enzyme-linked Immunosorbent Assay (ELISA) targeting

Cry1Ab and Cry1Ac and Immuno strips detecting Cry1Ac, Cry2A, Cry1Ab proteins were employed using CRMs of four GM maize events. DNA-based screening methods, *viz.*, Polymerase Chain Reaction (PCR) and Real-time PCR targeting *P-35S* promoter and *T-nos* terminator, Loop-mediated Isothermal Amplification (LAMP) targeting *P-35S*, were employed using CRMs of six GM maize events. Single plex PCR- and real-time PCR assays targeting *P-35S* and *T-nos* were used to check adventitious presence of transgenes in 50 *ex-situ* maize collections representing 11 different regions. Applicability of LAMP, ELISA and Immunostrip based screening assays were also checked for monitoring adventitious presence of transgenes using selected *ex-situ* accessions representing different regions. Based on the tests conducted, none of the samples were found positive for respective screening target, hence, confirming absence of adventitious presence of transgenes in these samples. Different protein and DNA-based methods were compared and these methods were high throughput, with rapid on-site applicability in case of Immunostrips and visual LAMP. Based on cost of testing, visual LAMP was found efficient. To ensure genetic purity and GM-free conservation of germplasm in genebanks, all these methods could be easily employed to check adventitious presence of transgene in *ex-situ* collections of maize and for other crops with rich diversity, where field trials of GM events of crops are being conducted in the close proximity.

Checking adventitious presence of transgenes in selected *ex-situ* maize samples representing 11 different regions employing QuickStix™ Combo Kit



Real Time PCR Amplification profiles for checking adventitious presence of transgenes in *ex-situ* maize samples employing targetings (a) *P-35S* and (b) *T-nos*

Checking adventitious presence of transgenes in 11 selected *ex-situ* maize samples employing LAMP assay targeting *P-35S*

The objective of the study was to detect viruses in the *in vitro* cultures in banana germplasm conserved in the IVGB at ICAR-NBPGR and to study the efficacy of cryotherapy for generating virus-free material. Fifty accessions of banana germplasm were virus indexed, for two viruses, namely, BBTV and BBrMV using polyclonal antisera for DAS ELISA. Subsequently nine accessions were cryopreserved using droplet-vitrification protocol on proliferating meristems. Post-cryotherapy, all samples (9 accessions) were tested with DAS-ELISA to test for BBTV and BBrMV and selected samples

(5 accessions) were subjected to PCR/RT-PCR additionally for all *BSV* and *CMV* viruses. TEM studies were undertaken in six accessions. Out of the 50 banana samples tested for the two viruses, 100% were free from BBTV whereas 94% were free from BBrMV, based on DAS-ELISA test. Cryopreservation of nine accessions yielded mean shoot regeneration rate from 3.3 to 38.9%, depending on genotype. A success rate of cryotherapy of 25% for BSV elimination and 33% for BBrMV was achieved in accession K44-B. The demonstrated that cryotherapy is a potential technique for virus elimination.

Acknowledgement

I wish to thank all the Faculty Members of the Division of Plant Genetic Resources for extending support and cooperation in preparing the research contributions of M.Sc. and Ph.D. students made during the period of 2007-16.

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Seed Science and Technology

S.K. Jain and Dharam Pal

Introduction

Division of Seed Science and Technology, IARI, a premier institution for development of improved varieties, seed production and imparting training in the areas of seed science and technology, took the lead and established a separate section of seed testing in the erstwhile Division of Botany in 1961. This section of seed testing was upgraded as a full-fledged Division of Seed Technology in 1968 to provide leadership in maintenance breeding, seed production, certification, quality evaluation, storage, physiology, pathology, variety identification, genetic purity evaluation and DUS testing for plant variety protection, development of seed quality assessment procedures and human resource development. The Division was renamed as Division of Seed Science and Technology in 1984.

Significant Student's Research Achievements

Significant contributions have been made by the students of the Division in seed production research, seed quality evaluation, policy intervention in national seed sector, human resource development and extension. During the period (2007-16) a total of 67 students were awarded degree with MSc: 39 and PhD: 28 and their the significant contribution of their research is given below.

Seed Production Technology

The investigation was carried with the objectives of optimizing steckling population densities and foliar nutrient applications for maximizing carrot (*Daucus carota* L) seed yield and quality, optimizing harvest maturity of seed in relation to changes in physiological and biochemical parameters and improving carrot seed germination and stand establishment through seed enhancement treatments. Seed yield primary umbel^{-1} was observed significantly higher and on par in all the plant densities other than 13plants m^{-2}. Higher number of secondary (11.57) and tertiary umbels (30.1) plant^{-1}, seed yield secondary^{-1} (1.57) and tertiary umbels^{-1} (0.089g), seed yield plant^{-1} (25.2g) was recorded in lowest plant density (4 plants m^{-2}). Higher per cent contribution of secondary umbels to total seed yield was recorded at lower steckling densities (4 & 5plants m^{-2}). Plant density had no significant effect on germination per cent of seeds collected from primary umbels. Significantly higher seedling length (15.16cm), dry weight (1.513g) with respect to primary umbel and higher Vigour index I and II (1135 and 113 respectively) were recorded in 5 plants m^{-2} density. Among the foliar sprays, 0.1% borax at 30 and 60 DAT recorded highest number of seeds in all the three umbel orders, seed yield plant^{-1} (19.95g) and ha^{-1} (12.35q). With the advancement of seed development and maturation in carrot seeds from 10 to 50 DAA, chlorophyll a, b and total chlorophyll, seed water content of fresh seeds and reducing sugar content showed decreasing trend. Seed dry weight, non-reducing sugars and total soluble carbohydrates content increased from 10 DAA to 50 DAA. Maximum germination in fresh, shade dried and rapidly dried seeds of primary and secondary umbels were observed from 40 DAA. This indicates that the seeds reached physiological maturity and attained desiccation tolerance form 40 DAA. In shade dried seeds SDS-PAGE analyses showed early appearance of protein bands with molecular weight of 55kDa and 36 kDa. Flowcytometry analysis showed accumulation of higher level

Quality seed production in carrot

2C DNA content in seeds collected at 45DAA.The results suggest that planting density of 5 plants m^{-2}, foliar spray with 0.1% borax or 0.1% $MgSO_4$ at 30 and 60 DAT, harvesting seeds from 45 DAA produce high quality seeds in carrot cv. Pusa Rudhira. However, under adverse conditions umbels can be harvested as early as 40 DAA and shade dried for obtaining optimum quality seeds.

The present study was undertaken in maize [*Zea mays* L] to assess the performance of parental lines under different sowing dates and the feasibility of hybrid seed production under climatic change regime. Parental lines of single cross hybrids namely, BML6, BML7, VQL1, VQL2, HKI193-1, HKI163, CM150, CM151 were sown on six planting dates in *kharif, rabi* and spring-summer seasons. Hybrid seed production potential was compared with two parental lines namely,CM150 and CM151 in *kharif* and spring-summer seasons. The temperature and RH had significant influence on field emergence, vegetative growth, flowering behaviour (days to flowering, flowering, duration, anthesis, silking interval), seed setting and seed yield of the parental lines. Among parental lines: CM150 and CM151 were least; VQL-1 and 2, HKI193-1, HKI163 were moderate and BML6

Effect of temperature on silking in different parental lines of maize

and BML7 were most affected by weather conditions (different planting dates). The results showed that *kharif* season was ideal for *parental* line multiplication followed by spring-summer season under Delhi conditions. Non-synchronization of flowering in parental lines was less during *kharif* (1.81 days) than spring-summer (4.21 days) season. Thus, among parental lines, hybrid seed production was feasible of single cross hybrid: PEHM-5 in *kharif* and spring-summer season. For higher seed quality and yield, sowing should be undertaken during second fortnight of July and first fortnight of March for *kharif* and spring-summer seasons, respectively. Seed quality parameters *viz.,* seed germination, vigour indices and incidence of mycoflora were found significantly higher in seeds harvested from *kharif* season. Seeds produced in all the seasons could safely be stored for 12 months from harvest under ambient conditions. Among fungal pathogens, *Aspergillusflavus* infection recorded was maximum, whereas *Fusarium moniliforme* was minimum.

Among the time of planting in onion [*Allium cepa* (L)] cv Pusa Riddhi, 15 October planting (T_1) showed significantly higher values for growth characters *viz.,*seed scape height (101.74 cm), umbel diameter (6.54 cm) productive umbellates per umbel (414.73) seed yield attributes such as seed setting (83.69%), seed yield per plant (8.71 g) and seed yield/ha (6.86 q). Higher seed quality attributes *viz.,* seedling length (9.48 cm), seedling dry weight (1.90 mg/seedling), germination (88.33%), vigour index-I (833.32), vigour index-II (167.08) and lower disease infection (38.06%) were observed in 15 October planting. Among the treatments spacing 60×30 cm had recorded significantly higher number of leaves/plant (40.89), seed scape height (102.26 cm), productive

Optimization of quality seed production in onion

seed scapes per plant (8.35), productive umbellates per umbel (419.4), seed setting (83.24%), seed yield per plant (12.43 g) and 1000-seed weight (3.01 g). The seedling length (10.37 cm), seedling dry weight (1.95 mg/seedling), germination (89.76%), vigour index-I (918.56) and vigour index-II (172.96) were significantly higher in S_3 (60×30 cm). The lower disease incidence (27.65%), PDI (5.81%) and EC of seed leachates (2.19 ì mhos/cm/g) were also recorded in S_3. Whereas, seed yield/ha (7.17q) was maximum in 60×10cm spacing (S_1). The chlorophyll content was also recorded higher in 15 October planting and 60×30 cm spacing. The foliar spray in combination of B+Zn+Ca+Mg (at 30 & 60 DAP) showed superiority in productive umbellates per umbel (579.35), umbel diameter (7.38 cm), 1000-seed weight (3.87 g), seed yield per umbel (4.18 g), seed yield per plant (16.70 g) and seed yield/ha (8.35 q). Among the quality attributes *viz.,* germination (91.84%), seedling length (10.79 cm), seedling dry weight (2.62 mg/seedling), vigour index-I (990.34), vigour index-II (240.73) and chlorophyll content were significantly higher in T_{15} than others. Whereas, lower EC from seed leachates was recorded in T_{14} (1.50 µmhos/cm/g) followed by T_{15} (1.65 µmhos/cm/g). The paclobutrazol treatment (T_6) showed significant lower number of leaves per plant (24.58) and lower seed scape height (69.9 cm) than other treatments. The T_6 also showed higher umbel diameter (7.31 cm), number of umbellates per umbel (628.05), 1000 seed weight (3.32 g) and seed yield/umbel (3.48 g). The triadimefon treatments T_{13} and T_{14} significantly reduced the disease incidence (16.5% & 17%) and severity (4.50% & 5.16%), respectively resulting into higher seed yield per plant (12.67 g) and seed yield/ha (6.34 q) than other treatments. Paclobutrazol treatments (T_1 to

T_6) showed higher chlorophyll content and antioxidant enzymes *viz.,* SOD, catalase and glutathione reductase than other treatments.

Significant difference among the methods of growing for leaf area was observed in trailing method of bottle gourd cv. Pusa Naveen at 45 and 55 DAS (9576.3 cm^2 &42807 cm^2), respectively. Significantly higher vine length (4.56 m), number of leaf per vine (40.48) and leaf nodes (40.48) were recorded in trailing in comparison to traditional method on 55 DFS, whereas these characters were non-significant on 45 DFS. Significantly less

Methodolgy and direction of seed planting for quality seed production in bottle gourd

number of days was taken for anthesis of first male flower (49.82) and female flower (54.67) in trailing. Non-significant differences for male flower, female flower/plant and sex ratio were observed among the trailing and traditional method of growing. The fruit development attributes i.e. fruit set, fruit developed to maturity, fruit weight, fruit length, fruit width and thickness of fruit wall were significantly higher in trailing than traditional method. Similarly, results were recorded for seed yield attributes *i.e.* total seed/fruit (723.01), filled seed per fruit (651.88), seed yield per fruit (98.33 g), seed yield per vine (346.16 g) and seed yield per acre (410.13 kg) were significantly higher in trailing in comparison to traditional method. The seed quality attributes *viz*, germination percentage, root & shoot length, seedling dry weight, vigour index I and II and embryo weight were also significantly superior in trailing method as compared to traditional method. Electrical conductivity from seed leachates, and seed coat weight showed non-significant difference among the growing methods. Seed physical parameter like breadth, eccentricity and axial width showed significant differences among the growing methods.

Rice (*Oryza sativa* L.) is one of the world's most important cereal, particularly in Asia, but increasingly so in Africa and Latin America as well. However, seed yields often fluctuate due to various environmental stresses.

Among the abiotic stresses, heat stress is emerging as the most serious threat to hybrid rice seed production as a consequence of climate change. Global climate change is likely to increase the current vulnerability of the crop to climate, with a projected global average surface temperature increase of 1.4 – 5.8ºC by 2100. Hybrid rice with 15-20% yield superiority over HYVs play a very important role in increasing the production of rice. More than 45 rice hybrids have been released for commercial cultivation in India. Date of flowering in the parental lines need to occur in synchrony for successful pollination and hybrid seed set. Little is known about the flowering behavior of the parental lines of Indian rice hybrids in high temperature conditions and its effect during hybrid seed production. An experiment was conducted to study the flowering pattern of parental lines of three Indian rice hybrids *viz.*, DRRH-2 (IR 68897 A, IR68897 B, DR714-1-2 R), DRRH-3 (APMS 6A, APMS 6B, RPHR-1005), PRH-10 (PUSA 6A, PUSA 6B, PRR 78) with three different sowing dates (I: 26/03/12; II: 30/04/12 and III: 13/06/12) at the experimental field of IARI, New Delhi. The three different date of sowing i.e., I, II and III showed variation in average temperature as 32, 29.4 and 28.6 ºC respectively from nursery sowing to physiological maturity. The parental lines of DRRH 3 were found susceptible to high temperature; however, parental lines of DRRH 2 and PRH 10 showed tolerance up-to some extent against high temperature. The seed quality was assessed after harvesting. Seeds produced under high temperature showed low in vigour index I (29%), vigour index II (20%) and germination (8%) as compared to those produced under normal sowing conditions. The study identified optimum time of sowing of parental lines of the three hybrids under Delhi and similar conditions elsewhere in India for the diversification of hybrid rice seed production areas.

Results revealed that late sowing often advances early flowering and maturity period of Indian mustard (*Brassica juncea* (L.) Czern & Coss). Morphological characterization revealed that out of seventeen descriptors, six visually assessed descriptors were monomorphic and out of eleven measurable characters, three were monomorphic, seven dimorphic and one polymorphic in both the years of the study. All the six visually assessed descriptors and eleven measurable characteristics were observed to be stable in performance over both seasons. Out of three OP varieties, two were grouped together whereas the third one was stood alone. The protogynous plants were grouped and clustered separately from that of varieties for measurable characters. The protogynous plants studied were totally distinct, uniform, stable and novel from that of OP varieties of brassica species. The variation in floral morphology, especially the stigma features i.e. stigma exertion from the floret prior to anthesis and protogyny interval, which ranged from 8-10 days, formed phenotypically distinct classes. Studies on stigma receptivity and pollen viability revealed significant variability and were at its peak three days after anthesis which reduced drastically thereafter. Compatibility crosses in protogyny plants, post pollination events starting from pollen deposition on the surface of the stigma and culminating with the entry of the pollen tube into the embryo sac, was rapid and completed in two days after pollination. The incompatible crosses did not show even single pollen germination on stigmatic surface and consequently no pollen tube growth upto the six days after pollination. Average seed set ranged from 3-8 seeds per pod from cross pollination and nil in self pollinated plants. The investigation also revealed that variation exists for protogyny interval, stigma receptivity and compatibility and these findings demonstrated that protogyny can be used successfully to produce controlled hybrids in brassica species without emasculation. For overcoming the incompatibility, chemical treatments of stigmas just before pollination was found to be effective in Pg and SI lines of Indian mustard. The chemical treatments appeared to mask the self-incompatibility recognition molecules of the pistil in overcoming the incompatibility. Among various treatments IAA (40 ppm), sucrose(4%) and NaCl (2%) were found to be more effective in inactivating/overcoming SI barriers compared to others. Application of molecular markers linked to sterility and fertility restorer genes was validated. The results of individual amplifications of these genes indicated the presence of four sequence-characterized amplified region (SCAR) markers, which enabled rapid and reliable identification of the CMS. Expressions of the *orf* genes were detected in the CMS line, while SCAR primers generated amplicons in the fertile plants. SCAR markers linked to the male fertility gene are located on the *Rf* locus and were found potential for genetic purity assessment of hybrids.

Black cumin (*Nigella sativa* L) is a multipurpose rabi crop cultivated for its seeds. The crop is sown during October-November and harvested during March-April under North and Central Indian conditions. Flower of

Nigella sativa basically shows unique mechanism to ensure self pollination, although abundant pollinators also visit flowers. However, the flower of *Nigella sativa* possess characteristic feature of both entomophyllus and anemophyllus flowers which makes it ambophyllus in nature where pollination is done by both biotic and abiotic pollinators in the same flower, providing some kind of reproductive assurance in the absence of pollinators to ensure successful reproduction and better seed setting. Our study shows that *N. sativa* was able to produce seeds under open condition (plants tagged & un-covered, T0), under selfing (plants covered by net cloth, T1), and by manual pollination of emasculated flowers (T2). Under T0, T1, and T2, seed yield of 8.46, 8.10 and 7.83 g/plant was obtained respectively. Seed development and maturation, onset of germination, desiccation tolerance and period required for seed maturation was also studied. Onset of germination took place after 35-40 days of pollination, while acquisition of desiccation tolerance took place 40-45 days after pollination when seeds reach harvesting maturity.

Morphological characterization and floral biology of parental lines of pumpkin cv. Pusa Hybrid – 1 were studied during summer seasons of 2008 and 2009 at Seed Production Unit farm, IARI. The parental lines were evaluated for a total of 40 phenotypic characters (3 plant, 2 stem, 3 tendril, 7 leaf, 4 flower, 19 fruit and 2 seed) following NBPGR descriptor list at vegetative, flowering and fruit maturity stages. Parental lines showed considerable variation in early plant vigour, plant growth habit, petiole length, leaf size, leaf blotches and number of primary branches (vegetative); node number at which first female flower appears, days to 50% flowering, sex ratio (floral characters); fruit colour (at immature stage), shape, blossom end shape, ridges on fruit, rind thickness; fruit weight, length, breadth, flesh thickness (fruit characters) and 100 seed weight. Flower anthesis began between 3.00 AM and 4.00 AM with peak reaching between 5.00 AM and 6.00 AM in both parental lines. The flower closure was initiated at about 8.00 AM and finally closed between 11.00 AM and 12.00 Noon in both the male and female flowers. The flower remained open for 3 h 30 min in the male and female parents. There was, however, a period of overlap in the flower anthesis of both sexes, which coincides with the peak of pollen viability and pollinator activity. Pollen viability was about 97.64 - 98.55% in freshly opened flowers but decreased to about 74.73% after 48 h and crashed to 23.40% after 96 h of its storage under ambient conditions. Pollens stored under refrigerated conditions (4^0 C temperature and RH – 40%) maintained 63% viability after 96 h of storage. Stigma receptivity, measured by fruit set percentage, was noted from 12 h before anthesis with highest at the time of anthesis.

In bottle gourd the high morphological diversity was observed among public sector genotypes has shown possibility of expanding the genetic base. Significantly higher vine length (4.96 m), number of fruit set (6.65) and number of matured fruit per vine (5.30) were recorded in trailing than the traditional method. The fruit development attributes i.e. fruit weight, fruit length and fruit width was significantly higher in trailing. Similarly, results recorded for seed yield attributes, the number of filled seed per fruit (584.7), seed yield per vine (517.5 g) and seed yield per acre (689.6 kg) were significantly higher in trailing in comparison to traditional method. The seed quality attributes viz, germination percentage, seedling length, seedling dry weight, vigour index I and II and seed moisture content immediately after harvest were also significantly superior in trailing method as compared to traditional method. The economic analysis of hybrid seed production showed higher BC ratio in trailing 3.87 and lower in traditional method (2.10). The significantly higher fruit set percentage (78.84%), number of filled seeds per fruit (505.73), 100 seed weight (15.72 g) and seed yield per fruit (79.53 g) were recorded at 2.00 PM pollination. However, the lower number of unfilled seeds per fruit (41.48) was also recorded at 2.00 PM pollination as compared to pollination at 12.00 noon, 4.00 PM and 6.00 PM. The seed quality parameters viz., germination (98.03%), seedling length (32.36 cm), seedling dry weight (0.4108 g), seedling vigour index-I (3172.48) and seedling vigour index-II (40.26) were significantly higher at 2.00 PM pollination. The electrical conductivity was significantly lower (45.15 µmhos/cm/g) at 2.00 PM pollination as compared to pollination at 12.00 noon, 4.00 PM and 6.00 PM. Highly significant differences of harvesting stages on fruit weight, fruit length, fruit width, number of filled seed, 100 seed weight and seed yield per fruit were noticed in the fruits harvested at 60 days after anthesis. The seed quality parameters i.e. germination percentage, seedling length, seedling dry weight, protein and oil content of seed kernel were recorded significantly higher in the fruits

harvested at 60 days after anthesis. Significantly higher fruit weight, fruit length, number of filled seed per fruit, 100 seed weight and seed yield per fruit was recorded at 10 whereas number of filled seed per fruit was higher at 40 days of post harvest ripening period. Similarly, there was significant effect of post harvest ripening period on seed quality parameters. Significantly higher germination (94.55%), seedling length (31.44 cm), seedling dry weight (0.363 g) weight of seed coat (0.3763 g) and weight of cotyledon (0.370) were noticed in the fruits kept for 30 days of post harvest ripening period. Similarly, higher fruit and seed quality parameters were recorded in 30 days ripened fruits (P3) over other post harvest ripening periods and also in fruits harvested at 60 days after anthesis and kept for 30 days of post harvest ripening period (H3P3) over other treatment combinations.

The success of crop production technology of scented varieties primarily depends on quality seeds. One of the important factor which affects the production of good quality seed in rice is the maturity stage of the crop which in turn is prone to modifications in agronomic practices. The three varieties of basmati rice *(Oryza sativa* L*)* viz., Pusa Basmati-l, Pusa Basmati-1121 and Pusa Basmati-6, were transplanted during *kharif 2009,* from I July to 1 August at an interval of ten days. Seed yield and yield parameters alike, total number of productive tillers, panicle length, number of filled grains/panicle, spikelet fertility, harvest index, and seed recovery (%) were significantly affected by time of transplanting. Pusa Basmati-6 gave the maximum yield (76.58 q/ha) when transplanted on 10 July. However, the seed yield reduced drastically, if transplanted on 1 August (41.46 q/ ha). Similar trend in seed yield were recorded in Pusa Basmati-l and Pus a Basmati-1121. The maximum seed recovery (87.33%) was recorded in variety Pusa Basmati-6 of 10 July transplanting. The maximal 1000-seed weight (29.87 g) was observed in Pusa Basmati-1121 in 1st July transplanting, whereas, the lowest 1000-seed weight (27.47 g) was observed in 1st August transplanting. Pusa Basmati-6 and Pusa Basmati-I showed drastic reduction in 1000-seed weight with delayed transplanting. Seed quality in terms of germination was higher in the early transplanting dates of 1 July. Pusa Basmati-1121 recorded highest percentage of germination (94.33%). The superior seed quality in relation to electrical conductivity was recorded when transplanting was done on 1 July. Significant reduction in seedling vigour was registered in 1 August transplanting. Among three varieties, Pusa Basmati-1121 showed highest seedling vigour (2578.1).

Seed yield of Indian mustard is mainly affected by prevailing temperature especially at flowering and seed setting. It is important to choose the right time for sowing. Late sown early varieties produce less number of siliqua and early sown late varieties give more leafy growth and produce siliqua very late. Therefore, it is necessary to evaluate the effect of different sowing times on the plant growth, seed yield and quality. Standardization of optimum planting ratio is also essential to facilitate effective pollination and to accommodate more female plants in the seed production plots to get maximum hybrid seed yield. Our present study showed that the optimum sowing time of parental lines of NRCHB 506 was third week of October to get good quality seed of maximum yield. Optimum planting ratio of male to female for hybrid seed production was 2:8, but for obtaining higher seed yield per unit area it may be extended up to 2: 1. Honeybee population was found in an increasing mode with rise in temperature up to a certain limit, indicating a relatively less bee population in early sowing plants for which flower period coincides with relatively low temperature conditions. Few beehives near the growing field may compensate this deficit in hybrid seed production of mustard in October. Female plants produced seeds of higher test weight under low yield condition. Strict maintenance breeding is must to maintain the genetic purity of the parental lines. Our study emphasized to provide an optimum seed production practice to the seed growers under Delhi conditions.

The present study was conducted to investigate the effects of zinc fertilization on seed yield and quality parameters of basmati and non-basmati rice cultivars. The experiment was conducted in two factorial randomized block design consisting of 4 treatments and 4 varieties, each having 3 replications. The treatments include control (without zinc), 2.5 kg zinc ha^{-1}, 5.0 kg zinc ha^{-1} and 7.5 kg zinc ha^{-1} tested for different rice varieties *viz.,* Pusa-44, Pusa-834, Pusa Basmati-1121 and Pusa Basmati-6. The source of zinc used was zinc sulphateheptahydrate. The experimental findings showed that zinc application @ 7.5 kg ha^{-1} had a significant effect in increasing the yield parameters, alike the total number of tillers, total number of productive tillers, plant height, number of

grains/panicle, number of filled grains/panicle, percentage of spikelet fertility, number of days to 50 per cent flowering, days to maturity and seed yield. Seed quality parameters namely, germination percentage, test weight, seedling length, shoot length, root length, seedling dry weight and vigour indices had increased significantly with zinc application @ 5.0 kg ha^{-1} and 7.5 kg ha^{-1}, whereas, the electrical conductivity decreased with zinc application. Seed health was improved in all rice varieties when zinc was applied @ 2.5 kg ha^{-1} and 5.0 kg ha^{-1}. Hence, it is suggested that zinc application @ 5.0 kg ha^{-1} can be economical for improving seed yield and quality of rice seed.

Studies on standardization of seed production of bitter gourd hybrids; Pusa hybid-1 and Pusa hybrid-2 were undertaken in spring-summer and *kharif* season under Delhi conditions. To modify sex expression of female parental lines of above hybrids growth regulators namely GA_3 @ 50 ppm, NAA @ 200 ppm, Maleic Hydrazide @ 100 ppm and etherel @ 50 ppm were sprayed on seedlings at three leaf and tendril initiation stage. Pollen viability and stigma receptivity studies were undertaken and pollination was done at different times of the day (7, 9, 11 am, 1 and 3 pm) to identify best time for pollination to achieve higher seed yield. Results showed that growth regulators significantly increased vegetative growth (vine length, number of branches, total number of nodes), sex expression (induction of female flowers at lower node, more number of female flowers, higher sex ratio), fruit traits (total number of fruits/plant, fruit weight, fruit length, fruit width), seed yield (total number and weight of seeds/fruit) and seed quality (germination and seedling vigour). Weather conditions had immense effect on hybrid seed production and quality. Among seasons compared, *kharif* season yielded higher fruit and seed yield but seed quality was better in spring-summer season. Time of pollination studies showed that fruit setting percentage, fruit traits (weight, length and width), seed yield (number and weight of filled seeds/fruit) and quality (germination and seedling vigour) were higher when pollination was undertaken between 7.00-11.00 am. The better treatments for modification of sex expression identified were: GA_3 @ 50 ppm, NAA @ 200 ppm and etherel @ 50 ppm. Economical hybrid seed production of bittergourd is feasible during *kharif* season under Delhi conditions.

The present investigation entitled in cherry tomato grown under different environmental conditions. Significantly higher seed yield per plant was measured in semi-climate controlled polyhouse (7.11 g) as compared to naturally ventilated polyhouse (4.08 g) and lower berry yield per plant in insect proof net house (3.38 g). In general, the crop duration was longer in semi-climate controlled polyhouse, compared to naturally ventilated polyhouse and insect proof nethouse. Significantly higher values for germination (84.63%), speed of germination (1.58), seedling shoot length (6.34 cm), seedling root length (6.49 cm), vigour index-I (532.53), vigour index-II (1.29) and less solute leakage (0.0070 ìS/g/cm) was measured under semi-climate controlled polyhouse condition of seed, in comparison to naturally ventilated polyhouse, and insect proof nethouse. Different methods of pollination were studied under different structures and found that semi-climate controlled polyhouse recorded higher number of berry set per truss (70.13), seeds per berry (67.87), seed yield per berry (0.0876 g), germination (80.57%) in comparison with naturally ventilated polyhouse and insect proof nethouse. The air blow method of pollination had highest number of berries per truss (72.91), berry weight (6.17 g), berry polar diameter (2.22cm), berry radial diameter (2.36 cm), 100 seed weight (0.129 g), seed yield per berry (0.0872 g), germination (80.71%) followed by vibration method.

High temperature stress adversely affects plant growth and seed quality, reducing grain yield of wheat. Hence, the material was raised under three different dates of sowing during the month of November, December and January to expose the plants to different range of temperature. The study revealed significant differences among dates of sowing, thereby indicating sensitivity to heat stress. However, the degree of sensitivity varied; as the crop was much more susceptible to heat stress under extremely late sowing conditions. The genotypic differences were significant for all traits under investigation, suggesting that the material was genetically diverse. Among the plant morphological traits, there was a maximum reduction in plant height under extremely late sown conditions (January sown) as compared to December sown crop when compared with normal conditions. Phenology of wheat crop was also significantly affected by higher temperature. Wheat varieties sown during the

month of November took more time to reach the anthesis stage than sown in December and January, wherein days to anthesis got reduced. In addition, all varieties sown in November took maximum days to mature, while last sowing date recorded lesser crop growth duration. The traits viz. flag leaf area, spikelet fertility and pollen fertility were comparatively less affected due to higher temperature. With respect to seed parameters, the study revealed that seed yield and yield contributing traits viz. tiller number and 1000-seed weight were most susceptible to heat stress.The spike characters viz. spike density, spike length and number of spikelets per spike were least effected by Heat stress and hence identified as heat tolerant traits. The traits viz. tiller number, grain yield, biomass, 1000-grain weight and vigour Index II were found to be most susceptible to heat stress.The study also identified tolerant and susceptible genotypes with respect to all the morphological and seed traits, as revealed by Heat Susceptibility Index Values.

Okra seeds of Pusa A-4 cultivar were harvested 10, 17, 24, 28, 32, 36 and 40 days after anthesis (DAA). At 40 DAA when seed moisture content dropped to around 8%, 20% and 26% fresh and rapidly dried seeds, respectively tested were hard seeded. Maximum seed germination in fresh developing seeds was attained at 36 DAA, when physiological maturity and maximum seed dry weight were attained. With seed maturation, there were rapid decline in leachate conductivity and seed moisture content. The desiccation tolerance in fresh okra seeds was attained after 28 DAA and after this stage, non-reducing sugar concentration increased many fold. One of the major problem of okra growers is difficulty in obtaining proper seedling emergence under low temperature. Solid matrix seed priming (SMP) was performed by mixing seed with moist Vermiculite for 6, 12, 18, 24, 30, 36 and 48 h incubation at 20°C and 25°C. Based on seed germination, speed of germination and seedling vigour parameters, solid matrix priming for 24 h duration at 20°C was optimum for improving seed quality in okra cv Pusa A-4.

The seed quality and storability of tomato as affected by the growing conditions, stage of fruit picking, seed extraction and drying methods were also investigated. The parental lines of Pusa Hybrid-4, performed better for the plant growth, flowering behaviour, fruit and seed set in green house condition than the open field conditions. The mean temperature below 20°C was found crucial for fruit and seed set and when the night temperature goes below 12°C reduced the pollen production and fertility drastically. The total number of days available for pollination work was almost double in green house condition in comparison to open field. In green house condition the stigma remained receptive upto seven days and as the age of the stigma increased the receptivity decreased. The fruit set percent was more when the pollination done one day after emasculation (71.11 percent) followed by 2nd day (65.88 percent) and 3rd day (63.33 percent) and lowest in the 7°' day pollinations (43.33 percent). The increased seed set per fruit was observed with the repeated pollination of the same emasculated flower bud twice. Pollen can be stored uptothree days and upto ten days without much reduction in pollen viability in ambient and in refrigerator respectively. The parental line seed produced in green house and open field had 5 to 10 percent dormancy and it was significantly higher in green house produced seed. No dormancy was noted in F, seed in any of the environments. The seed could be stored upto 12 months in ambient conditions without significant reduction in germination. However, seed vigour showed significant decrease after six months storage.

The proposed work was undertaken to evaluate the effect of sowing dates (early November, Mid November and early December) and nitrogen levels (0, 45 and 90 Kg N/ha) on seed yield and its quality of Isabgol. Seed testing protocols were evaluated by germinating the seeds in top of the paper and between paper and under 20°C, 25°C and 30°C. Dormancy status and its reduction methods were determined by using GA_3, KNO_3, prechilling, alcohol, acetone. Seed quality parameters were analysed of different seed lots collected from various agroclimatic locations of India. Seed storage behavior and desiccation sensitivity was studied by testing germination and vigour under different moisture contents and temperature regimes. Finally diversity analysis of cultivated and wild species of *Plantago* using RAPD markers was done. It was seen that Yield of Isabgol is moderately responsive to Nitrogen as we have seen no effect in 2007, but 45 Kg/ha N gives a higher yield, but for sowing dates, late sowing in December hampers seed yield: Must be sown in Oct-Nov. Plant height

decreased significantly in the late sown plants again emphasizing the earlier result, with no effect on N level. Inflorescence/spike length increases in late sown plants. Germination is not affected byN-dose but significantly reduces in the late sown plants, similar are the results for Vigour Indices.

The experiments were conducted using parental lines of pumpkin cv. Pusa hybrid-1 of pumpkin. Highly significant differences among the method of pollination for number of fruit set (3.27) and number of mature fruit (1.63) per plant was observed in hand pollination. Significant decrease in numbers of unfilled seed (35.58) and an increase innumber of seeds per fruit (392.41) and number of filled seed (356.83) per fruit were recorded in hand pollination. Seed yield/fruit (52.33 g), seed yield/plant (83.66 g) and 1000 seed weight (148.08 g) was significantly higher in hand pollination than natural pollination viz. 44.08 g/fruit, 46.00 g/plant and 134.90 g, respectively. Non significant difference for seed length, seed width, seed thickness and electrical conductivity was observed between the methods of pollination. Significantly higher germination (96 %) vigour index-I (3209), vigour index-II (40.47) was recorded in hand pollination than natural pollination in which germination percentage, vigour index-I and vigour index-II were (91), (2564) and (33.08) respectively. Significantly higher germination (94 %) vigour index-I (2520), vigour index-II (33.95) was recorded in hand pollination after six months of ambient storage. Maximum pollinator activity was observed at 8.00 am and minimum at 1.00 pm. Among parental lines the maximum pollinator activity was found in female flowers (24.68 /day/flower) and minimum male flowers (5.27 /day/flower). Honey bees were noted major pollinating agent.

For hybrid seed production of Pusa Rice Hybrid-10 (PRH-10), experiments with three dates of planting and six treatment (GA_3 application) in the row ratio of 2:8 (R:A) with three replication was undertaken in the experimental plot and one date of planting in seed production field.GA_3 dose 150 g/ha along with 1% boric acid in three split doses was found most effective in the maximum expression of characters favouring out crossing resulting in higher hybrid seed production in PRH-10 in both experimental as well as seed production plot. The optimum synchronization between parental lines and favourable environmental conditions during flowering contributed positively for higher yield of (3.17 t/ha) in 1stdate of sowing (1st week of June) in *kharif* 2004. The 1st and 2nd DOS harvested seed could maintain mean germination of more than 85% in different packaging material up to 18 months of storage. An yield level of 3.51 t/ha of hybrid seed can be obtained with the application of GA_3 @150g/ha along with 1% boric acid in 25:50:25 ratio. GA_3 application has no adverse effect on seed quality and the hybrid seed dried below 9% moisture can be stored in the ambient conditions for at least two planting seasons.

Seed Quality Assessment

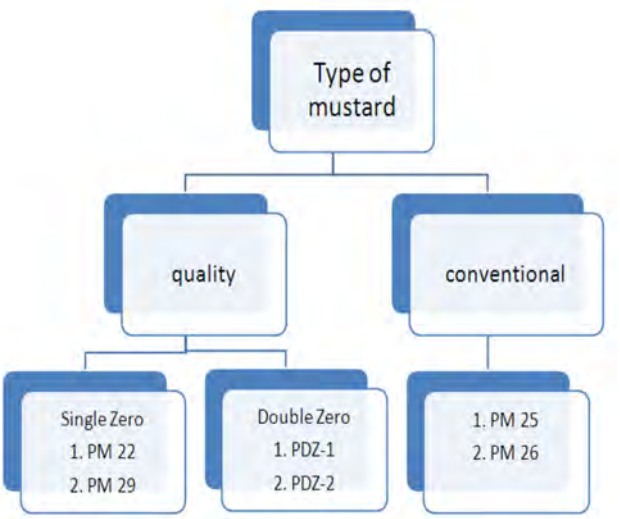
The study was undertaken to identify the SSR markers that could be used to test the genetic purity of six brinjal hybrids and their perspective parental lines. Among the 89 SSR markers studied, five markers (*viz.,* emk02K24, emb01M15, emg01B17, Eemd5F05 and CMS31) were found to be polymorphic between parental line of four respective hybrids (*viz.* PH-5, PH-9, NDBHL-20 and Kashi Komal) and found to be suitable for testing the genetic purity of these hybrids. A set of 5 identified polymorphic markers differentiated four of the six hybrids from each other, which can be used as referral markers for unambiguous identification and protection of these hybrids. The efficacy of any markers in ensuring the genetic purity of hybrids depends on homogeneity/uniformity and stability of the marker in respective parental lines. the analysis of plant to plant variation within the parental line of all hybrids, using the identified hybrid specific markers, showed highly homogenous profile of SSR markers, which further indicated the scope of application of these markers in maintenance and purity testing of parental lines and hybrid. Genetic purity analysis of certified hybrid seed lot of PH-9 was done using the identified hybrid specific marker, which explained 18% selfed seeds admixture in hybrid seed lot, which was reconfirmed through GOT.

Hybrid rice varietal identification was studied through molecular fingerprinting. About ten most informative sequence tagged microsatellite markers were employed for fingerprinting of Pusa RH 10. Out of ten STMS

markers analyzed, only two microsatellite markers (RM 206 and RM 228)depicted homozygous pattern in one seed lot of Pusa RH 10. The occurrence of inter-plant variation at non-linked marker confirmed the molecular heterogeneity. This highlights the importance of STMS markers in maintaining the genetic purity of the parental lines. Thus, .these STMS markers can be used with high degree of confidence for establishing distinctness of Pusa RH 10.

The present study was undertaken to identify the SSR markers that could be used to test the genetic purity of eleven pearl millet (Pennisetumglaucum) hybrids and their respective parental lines. Among the 40 SSR markers studied, 9 markers (viz., PSMP2084, PSMP2203, PSM P2040, PSlviP2089, PSMP2202, 13SME'2237, PSMP2273, PS1vf.P2270 and PSMP2263) were found to be polymorphic between parental lines of 10 respective hybrids (viz., RHRBH8609, REIRBH8924, GHB538,, GHB732. CHB744, GF1B719,. CliF1558, PUSA605, P1JSA23 and 11111167 improved) and found to be suitable for testing the genetic purity of these hybrids. Cluster analysis based on Jaceard's similarity coefficient using UPOMA grouped the hybrids into four major clusters. Within the cluster all the hybrids shared a cytoplasmic male sterile line as female (A) parent that were related to each other in their lineage. The genetic similarity between the hybrids ranged from 0.68 to 0.94 with an average similarity index of 0.79. A set of 5 polymorphic markers (Viz, PSMP2084,. PSMP2237, PSMP2202, P5MP2273 arid PS1 v IP2089) differentiated six other eleven hybrids from each other, which can be used as referral markers for unambiguous identification and protection of these hybrids.

The material comprised six genotypes of Indian mustard, in which two genotypes each were of conventional, single zero and double zero (quality types). The results showed significant differences amongst the conventional, single zero and double zero genotypes for imbibitions and electrical conductance (EC) from seed leachates. Double zero genotypes imbibed faster than single zero and/or conventional type genotypes; higher percentage of imbibitions (113.08%) with 24 hr of imbibition than those to single zero genotypes (87.24%) and/or conventional type of genotypes (62.76%). Yellow seeded genotypes (double zero groups) had also shown higher leakage (led to imbibitional damage) than those to black seeded genotypes. Significant differences between double zero and single zero and/or conventional type genotypes were also noticed for melanin, phenol, ascorbic acid, and antioxidant enzymes. The results indicated that imbibition pattern was significantly correlated with melanin and phenol content. The amount of melanin (55.99U) and phenol content (13.41mg GAE g^{-1}) in conventional type genotypes was significantly higher than single zero (48.32U and 12.07 mg GAE g^{-1}) and/or double zero (22.52U and 9.74 mg GAE g^{-1}) genotypes, respectively. The mean value for ascorbic acid was significantly higher in single zero (28.32mg/100g of seeds) than those to double zero (27.68 mg/100g of seeds) and/or conventional (23.69 mg/100g of seeds) type genotypes. The superoxide dismutase (SOD) and peroxidase (POD) activity was more in conventional type than single zero and/or double zero genotypes.

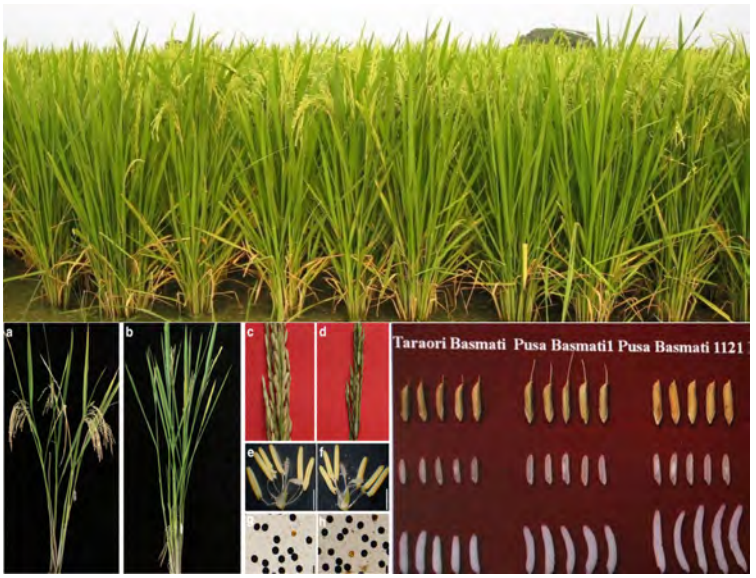


Biochemical variability in conventional and quality genotypes of Indian mustard

DUS Testing

The study grouped the 28 varieties of rice into five different clusters based on these DUS parameters. An exhaustive image library for various plant parts *viz.,* seed, leaf, stem and panicle was also developed for the material studied. Thus, a complete digital database comprising of 84 images each for seed, stem and panicle; and

448 leaf images was generated. Two different types of softwares were used for extraction of features from the images *viz.*, Grain Analysis Software (for size and shape features) and MATLAB software (for textural features). The varieties were grouped on the basis of these features generated from seed and leaf images. The size and shape features extracted by the Grain Analysis software as well as derived shape factors clearly distinguished the varieties. The additional textural features extracted from seed images (about 27 textural features) further aided in the differentiation of varieties. Thus, seed imaging features differentiated the varieties into six clusters. Similarly, image features were extracted from the leaf images. The MATLAB software extracted 27 textural features from both flag leaf and penultimate leaf with both ventral as well as dorsal side. Seven clusters were thus formed on the basis of leaf imaging features. The images were of immense help to distinguish the varieties on the basis of visual differences.



Morphological characterization of rice varieties

Sixty one rice cultivars showed overlapping of descriptor expression in various combination traits, but still the identity of all the cultivars in non-basmati as well as basmati group could be established individually. However, for characteristics like lemma: anthocyanin colouration of apex, density of pubescence of lemma and pubescence of blade surface; the categorization was subjective and varied from person to person; since the genotypes had to be grouped into weak, medium, and strong state of expression. Hence, the state of expression for these characteristics needs to be modified from the present state to include only two categories *viz.* absence or presence so as to rule out the ambiguity in recording the same, and to reduce the level of subjectivity involved. The assessment of biochemical characters, *viz.* phenol content, gelatinization temperature and amylose content is based on ripened seed onwards as per the DUS guidelines. However, changes occur even after the seed is stored. Hence, there is need to determine optimum period after ripening of grain for recording these observations. The observations on all these aspects were recorded beginning from freshly harvested seed vis-à-vis stored seed. The observations on the stored seed were recorded after every three months up to a period of one year. For phenol reaction, all the sixty one varieties showed intra-varietal variation and the final colour was recorded on the basis of predominant colouration of replicates. Four

Morphological characterization of rice varieties

varieties showed nil colour change of lemma (phenol reaction) and fifty seven varieties showed colour change of lemma from light (brown) to dark colour (brown or blackish brown) during the entire storage period. With

respect to gelatinization temperature, thirty five varieties showed constant alkali spreading value (ASV) even after twelve months of storage, whereas twenty six showed decrease in ASV. Hence, an increase in gelatinization temperature was observed with the decrease in alkali spreading value of rice starch and vice versa during various storage periods. The mean value of amylose content of freshly harvested paddy varieties ranged from 16 to 26%. The amylose content of rice varieties significantly increased during six months of storage period. Since amylose content is inversely related with glycemic index, hence, the varieties with high amylose content *viz.* Jyothi and Mahamaya were identified as valuable material for theurapatic use.

The morphological, physicochemical and cooking properties of milled rice were examined in 60 Farmers' varieties of rice which revealed significant difference with respect to grain dimensions, physicochemical and cooking properties. The length and breadth of milled raw rice varied from 3.99 to 7.65 mm and from 1.43 to 3.09 mm, respectively. Based on the L/B ratio, varieties were grouped into seven classes, short, slender (4), short bold (23), medium slender (4), long slender (6), long bold (21), Basmati type (1) and extra long slender (1). The amylase content ranged from 6.82% in variety Chipdo to 29.69% in sulendas. The gelatinization temperature (GT) was medium to high in 13 varieties and high in 47 varieties. Significance of wide differences with respect to grain sizes, amylase content and GT, which strongly influence the cooking quality of rice, in the farmers' varieties is discussed. Out of 40 SSR primers, which gave successful amplification of the genomic DNA, only a set of 8 (20%) primers were polymorphic, yielding 16 bands (alleles) among these 60 varieties. The size of the amplicons ranged from 120 bq (RM 238) to 240 bp (RM 551). The number of alleles per microsatellite ranged from 1 (RM 238, RM 119 and RM 120) to 3 (RM 259, RM 234 and RM 551) with an average of 2 alleles per locus. Major allele frequency ranged from 0.51 to 0.97 averaging 0.74. Genetic diversity ranged from 0.045 to 0.588 with a mean of 0.34. Considering the entire genotypic array, the mean value for polymorphic information content (PIC) for all microsatellite was 0.27. Microsatellite RM 122 with 2 alleles had the maximum PIC value (0.37) and the microsatellite RM 205 with 2 alleles had the minimum (0.27) value. The UPGMA cluster analysis grouped varieties into three main cluster with 58% genetic similarity and homing 24, 23 and 13 varieties, respectively.

Ascochyta blight is one of the most important diseases in NWPZ areas of our country, where favourable climatic conditions prevail during cropping season. Based on the screening results two promising genotypes under Desi chickpea namely, ICCV-13645 and ICCV04523 and one genotype under Kabuli chickpea namely ILC 3279 can be recommended to the farmers. Maximum growth and sporulation of *Ascochyta rabiei* was seen at 20°C in Potato Dextrose Chickpea Seed Extract Agar medium in comparison to Potato Dextrose Agar medium. A total of 63 RAPD primers were used for preliminary primer screening for this pathogen. Of these, only 22 RAPD primers gave amplification against *Ascochyta rabiei*; and only 7 promising primers identified (with reproducible results against all 10 isolates). Two primers namely OPS-5 and OPN-18 could be developed into SCAR marker based on the specificity and reproducible banding pattern. SCARs were advantageous over RAPD markers in specificity and reproducibility.

Fig. 11A: Effect of Ascochyta blight on chickpea seed quality varieties

Sheath rot in rice had maximum growth and sporulation was seen at 30°C in CDA medium. Thirty isolates were purified and used in the present investigation. Based on the growth of various isolates on CDA medium at 300C these thirty isolates can be grouped into three major categories of slow, moderate and fast growing cultures; similar

grouping can also be done for sporulation behaviour into low, medium and highly sporulating cultures. Based on the colony characteristics of various isolates representing different geographical locations of our country, these thirty isolates can be divided into six groups which represents different geographical locations. 7 RAPD and 8 ISSR primers were used against all 30 isolates. All RAPD primers showed 100% polymorphism except OPS13 (61.4%) and OPD5 (74%). Among ISSR primers ISSR5 and ISSR18 showed 100% polymorphism. In the combined cluster analysis of RAPD and ISSR markers, the similar geographical region isolates fits into same cluster as evident from the cluster I, comprised Kadapa, Ragolu, Maruteru, Westgodavari and Adutharai; all were from southern plain region of the country. The work has the potential of being exploited further to characterize the variability spectrum of this important pathogen with many other available marker systems.

Sheath rot symptoms on paddy

In the experiments, 12 isolates of brown spot of rice collected from different agro-ecological zones were used for the study. Results obtained showed that Patchouli oil was the most effective in 4 of the 5 isolates and it even surpassed the effect of *Thiram Royalflo* used as a recommended dose for seed treatment. Patchouli oil was the most efficient followed by Citronella oil and Lemongrass oil. Neem oil and Chinaberry extract were ineffective against all the 5 groups of isolates while Garlic extract and Basil oil were ineffective against B.o. 5 and B.o11. However they were effective to some extent against B.o 3 and B.o 7. Thiram. Royalflo recorded a significant zoneo Tiaimorrout it was less if compared to Patchouli oil. As far as Kalisena was concerned the maximum dose of 4x 10^8cfu resulted in effective control of *Bipolaris oryzae.* On the basis of present studies it has been found that Patchouli oil @ 1 per cent is highly effective in controlling the brown spot pathogen. The oil is easily available in the market @ Rs 3600 per litre. About 200 ml of Patchouli oil will be required to treat 20 kg of paddy seeds required for 1 hectare.

Seed Viability and Vigour During Storage

The study was conducted onphenotyping of soybean genotypes for seed quality and seed vigour, where seeds of 129 soybean genotypes were procured from one cycle of multiplication and used for morphological

characterization. The results registered a genotypic variability with respect to seed quality and vigour.100-seed weight was found to be negatively correlated with seed quality. There was a positive and significant but weak association registered between field emergence and seed storability (r=0.38); plant height and seed storability (r=0.52); days-to-50% flowering and seed storability (r=0.35). Of these 59 soybean genotypes, having sufficient quantity of seeds were stored for eight months under laboratory ambient environment (av 25±2^{0}C & 65±5% RH). Based on various physiological parameters, 10 genotypes each from good and poor storer were chosen and used for detailed physical, physiological, biochemical and molecular parameters. Seed germination, seedling growth, seedling dry weight, vigour indices, in general, decreased with increased period of seed storage or seed deterioration. Of 46 SSR markers used, 23 were polymorphic. Genetic similarity coefficients obtained using 23 SSR markers, grouped the studied 20 genotypes into four clusters. The PIC values ranged between 0.163 (Satt201) to 0.553 (Satt573) with an average of 0.329. SSR marker, Satt423 was found to discriminate two groups' *i.e.*, good and poor storer distinctly.

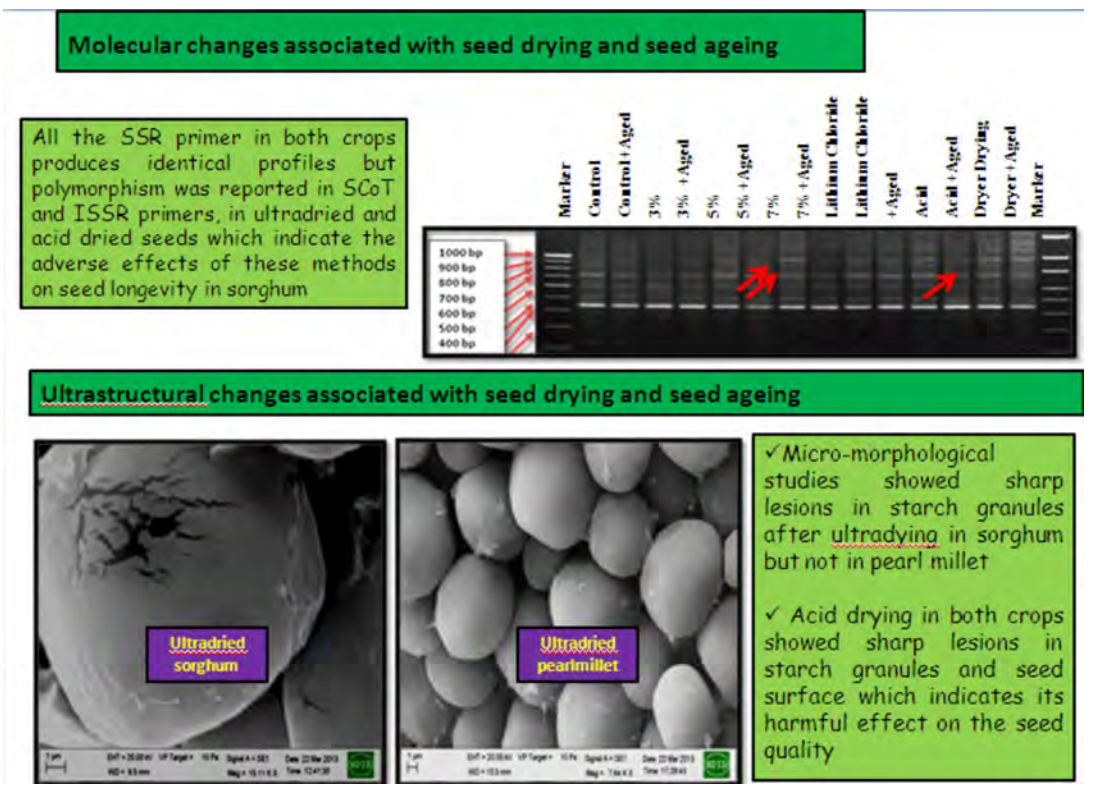
The study was conducted to evaluate and standardize different vigour tests to determine physiological potential of cauliflower and radish seed lots. Fourteen seed lots of cauliflower and 16 seed lots of radish were evaluated by the following laboratory vigour tests: standard germination, speed of germination, vigour index, mean germination time, accelerated ageing (AA), saturated salt accelerated ageing (SSAA), controlled deterioration (CD), electrical conductance (EC) from seed leachates and radicle emergence. The seedling emergence percentages, in the field, ranged from 44.6 to 65% and 50.3 to 61.3% for cauliflower and radish seed lots, respectively. The correlation coefficient (r) of following laboratory vigour tests in case of cauliflower *viz.,* standard germination (r=0.675), field emergence index (r=0.818), accelerated ageing (r=0.964), saturated salt accelerated ageing (r=0.878) at 72 hr, electrical conductance from seed leachates (r=0.844) at 6 hr and radicle emergence (r= 0.844) at 54 hr were significantly correlated with field emergence. The vigour index I (r=0.158), vigour index II (r= 0.495) and speed of germination (r=0.389) showed non- significant correlation with field emergence. In case of radish, standard germination (r=0.685), controlled deterioration (r=0.888), electrical conductance from seed leachates (r= 0.901) at 6 hr and radicle emergence (r= 0.781) at 44 hr significantly correlated with field emergence. Determination coefficient (R^2) of cauliflower seed lots forstandard germination, field emergence index, accelerated ageing, saturated salt accelerated ageing and electrical conductance from seed leachates were found significant and explains 45%, 67%, 92%, 63% and 69% of variation for field emergence, respectively, whereas in radish, determination coefficient (R^2) of seed lots forstandard germination, controlled deterioration and electrical conductivity were found significant and explains 46%, 79% and 70% of variation for field emergence, respectively. The correlation coefficient (r) and the coefficient of determination (R^2) of AA, SSAA, EC and controlled deterioration were found to be most significant among all studied vigour tests in predicting the seedling emergence of cauliflower and radish seed lots.

Deterioration of seed is inevitable and irreversible process. The seed vigour is affected by storage time, storage temperature, seed moisture content and environmental humidity. The longevity of seed depends on the amount of water it contains and on the temperature under which it is stored. In general, lower the moisture content higher is the seed longevity. The genotypes of cauliflower and radish were dried using silica gel to obtain the moisture content 2-3 and 1-2% and then stored for 4 and 6 months at both ambient and medium term storage (MTS) condition with control (moisture content 5-6%). The same seeds samples were also subjected to accelerated ageing for 1, 3 and 6 days. The seeds physiological characteristics, which include germination percentage, seedling length, root and shoot length, vigour index I and II, electrical conductivity and the activities of superoxide dismutase (SOD), peroxidase (POD), catalase (CAT) and dehydrogenase, varied with different moisture contents of seed.. The electrical conductivity of ultra-dry seeds was higher than that of control. The same result was also observed in the case of vigour index I and vigour index II. The result showed that all the species of both the genotypes were tolerant to ultra-dry condition and gave better result relating to physiological indices than the control condition. Under the same ageing condition, storage regimes and duration the seeds quality with control moisture content (5-6%) decreased greatly, whereas that of ultra-dry seeds still maintained at higher level. 2-3% moisture content samples in both the genotypes of

cauliflower and radish gave better result than 1-2% and control. The results manifested that ultra-dried seeds in both the species could maintain longevity and ultradrying treatment was an ideal method of seed storage. This research showed that the ultradrying technique could enhance the aging-resistant capability and storability of radish and cauliflower seeds. This technique would be potentially useful for the storage of the genotypes of cauliflower and radish.

Seed deterioration was hastened by retention of high moisture at higher relative humidity by the poor storersoyabean genotypes. Higher activities of lipoxygenase (LOX I and II) and maintenance of hydroperoxideslyase activity during storage aggravated the peroxidation of PUFAs in the cell membrane and resulted in higher release of volatile aldehydes in soybean seeds. The level of volatile aldehydes released and lipoxygenase II activity, concomitant with the reduction in germination under ambient conditions of storage indicated the role of lipid peroxidation in seed longevity behavior (r = -0.6638 and r = -0.7639, respectively). Good storer genotypes were protected from highly reactive free radicals, by significantly higher activity of free radical scavenging enzymes *viz.*, superoxide dismutase, glutathione reductase and catalase activity. Though the mean peroxidase activity was higher in poor storer genotypes, it cannot clearly differentiate a good storer from the poor, and irrespective of initial status, the peroxidase activity declines drastically during storage. The peroxidase spot test, which is used for grouping soybean varieties into positive and negative reaction groups, which is not indicative of their storability behavior. It was also evidenced that the spot test reaction changes drastically with storage, revealing the importance of sampling only fresh/high vigour seeds for peroxidase test.Genetic similarity coefficients obtained through SSR data analyses grouped the genotypes into two major clusters representing black and yellow seeded genotypes. SSR markers Satt371, Satt453 and Satt618 produced specific allelic bands with respect to their contrasting storability behavior and testacolour making them candidate markers linked to seed storability and testacolour.

The genotypes of sorghum and pearl millet were dried using silica gel, saturated salt of lithium chloride, conc. sulphuric acid and dryer (conventional method used to dry seeds for conservation in seed gene banks at



15°C and 15% RH) to obtain the moisture content up to 6%. Among the drying methods, the seed desiccated using acid and silica gel attained a moisture content of 6 + 0.2% in 8 to 12 days duration in different genotypes, whereas the seeds dried using lithium chloride and seed dryer attained the similar moisture content after 19 to 24 days. In case of acid and silica gel drying, rate of drying was quick, but acid drying had pronounced harmful effects on seed quality during storage whereas silica gel drying was found to be effective and quicker as compared to seed dryer and maintained the high seed quality which was comparable with slow drying methods using seed dryer and lithium chloride, over the storage period. The sorghum and pearl millet genotypes were dried using silica gel and to obtain about 7%, 5% and 3% mc and were stored under ambient and MTS (15°C and 15% RH) conditions for six months. In case of sorghum, the mc of 7% and 5% gave higher values for germination and associated seed quality parameters studied as compared to 3% mc, whereas in case of pearl millet, the mc of 5% and 3% gave higher values for seed quality parameters as compared to 7% mc, irrespective storage conditions. Biochemical parameters such as EC, WSS which are negatively correlated with seed quality, showed significantly higher values in control followed by seeds stored with 3% mc in sorghum and 7% in pearl millet. Higher values of SOD, POX, amylase, dehydrogenase and protein content, which are positively correlated with seed quality, was observed in seeds stored with 5% and 7% mc in sorghum while 3% in pearl millet over the control after six months of storage. The study reveals that optimum moisture content for extending seed storability and to ensure maximum viability is between 5-7% in sorghum, while 3-5% in case of pearl millet. The present study on sorghum and pearl millet seeds using different parameters, namely, physical, physiological, biochemical, molecular and micro-molecular parameters, reveals that pearl millet seeds can be dried upto 3% mc without affecting the structural integrity and storage potential, whereas, in case of sorghum, safe mc for storage is 5%, as below this, the structural integrity and storage potential is adversely affected.

Twenty one commercial seed lots of three varieties of Pusa Sawani, Pusa A-4 and Arka Anamika, were evaluated following the laboratory vigour tests: germination, speed of germination, vigour index, brick gravel test, paper piercing test, accelerated ageing (AA), controlled deterioration (CD), dehydrogenase test, electrical conductivity (EC) and cold test. The vigour tests were able to assess vigour differences among the seed lots of all three varieties. The seedling emergence percentage in the field ranged from 35 to 63% for the investigated seed lots. The correlation coefficient (r) of following laboratory vigour tests *viz.,* standard germination (r=0.693), speed of germination (r=0.700), brick gravel test (r=0.719), paper piercing test (r=0.905) and controlled deterioration (r=0.931) were significantly and positively correlated with field emergence. The vigour index I (r=0.206), vigour index II (r=0.298) and dehydrogenase test (r=0.101) were non-significant with field emergence. Determination coefficient (R^2) of seed lots for standard germination, speed of germination, brick gravel test, paper piercing test and controlled deterioration were found significant and explains 49%, 50%, 52%, 83% and 87% of variation in field emergence, respectively. The regression analysis result for vigour indices (I and II), dehydrogenase test and cold test explains 4%, 9%, 2% and 7% of the variation in field emergence, respectively. The correlation coefficient (r) and determination coefficient (R^2) of controlled deterioration test and paper piercing test were found highly significant among all other vigour tests in predicting seedling emergence of okra seed lots.The results suggest that controlled deterioration (CD) and paper piercing test can be successfully used to predict seedling emergence in okra seed lots.

Seeds of 26 soybean genotypes were grown in one cycle of multiplication to bring uniform physiological status of seeds and were stored for a period of 24 and 36 months at controlled environment (8°C and 35% RH). Seeds retrieved from storage and conditioned to lab temperature before use. Black-seeded small soybean genotypes, showed better storability. A significant reduction in seed coat hardiness was observed with increase in storage period; however the reduction was much more under ambient seed storage than those of controlled storage. Seed germination, AA, RSG, seedling growth, seed vigour index I & II, irrespective of genotypes, decreased with increased period of seed storage, whereas EC, gradually increased with increase storage period. Under Delhi ambient storage environment, cream-/yellow-seeded poor storer genotypes registered marginal increase in 100-seed weight with increase storage period, whereas it decreased under controlled storage environment. Volatile aldehydes, esterase enzyme activity increased with increase period of seed storage, whereas superoxide

dismutase activity consistently declined with the increased period of seed storage based on detailed physiological and biochemical phenotyping, four genotypes, namely DS-12-13, DS-2614, SL-799 and SL-525 were identified as good performers/storer. Based on detailed physical, physiological and biochemical, investigations on freshly harvested and stored seed; the volatile aldehydes and/or seed hardiness are suggested as better indices of soybean seed quality. From the above two, any parameter be chosen – as a good marker for seed quality in soybean.

The relationship between seed quality characters and field emergence of 21 seed lots of three garden pea cultivars Pusa Pragati, Arkel and Azad P3 was evaluated. The seedling emergence percentage of garden pea seed lots was positively and significantly correlated with standard germination (r = 0.618), vigour index I (r = 0.599), vigour index II (r = 0.575), Tetrazolium test (r = 0.576) and accelerated ageing (r = 0.560). The electrical conductivity test (r = - 0.721) was significantly and negatively correlated with field emergence. The coefficient of determination for electrical conductivity test (R2 = 0.52) was found to be significant followed by standard germination test (R2 = 0.38) vigour index I (R2 = 0.35), vigour index II (R2 = 0.33) and Tetrazolium test high vigour lot (R2 = 0.33) in predicting the variations in the field emergence of pea seed lots under study and it explained 52%, 38%, 35%, 33% and 33% of the variation in field emergence, respectively. The high vigour lot after hydropriming showed significantly highest germination (96%) and seed yield (15.79 q/ha). Maximum emergence (69%) was recorded in high vigour lot after treating with 50mM $CaCl_2$. Halopriming with 50mM $CaCl_2$ improved field emergence of high vigour lots by 12% whereas hydropriming improved emergence by 13.3% and 8.0% in medium and low vigour lots over control, respectively. The results suggest that hydropriming and halopriming with 50mM $CaCl_2$ can be successfully used to improve seedling emergence and morphological characters of aged garden pea seed lots.

The germination of cauliflower variety Pusa Deepali seeds containing 7.5% moisture fell to about 70% after 12 months of storage whereas at 5.5% moisture content maintained germination above 95% when stored under low temperature. Under ambient temperature the germination of Pusa Deepali seeds with 7.5% moisture fell below the Indian minimum certification standards after 9 months of storage whereas at 5.5% mc, it remained above 80% after 12 months of storage. The germination of Pusa Synthetic seeds containing 7.5% moisture fell to less than 70% after 9 months of storage and in seeds with 5.5% moisture it remained above 80% after 12 months of storage under low temperature. Under ambient temperature the germination of Pusa Synthetic seeds containing 7.5% moisture could not be maintained above the Indian minimum seed certification standards beyond 6 months and at 5.5% moisture it was 76% after 12 months of storage suggesting thereby that Pusa Synthetic is a poor storerthan Pusa Deepali. The packing material and CO_2 concentration did not affect the germination and other qualities of the seeds. It was found that packets made of LDPE lined aluminium foil were capable of holding modified atmosphere for one year. An exposure to 20% CO_2 + 2% 0_2 for 7 days was able to control 20 days old rice moth larvae age completely in the seed store. At this combination of gases the pathogen incidence on the seeds reduced to nearly half. Although there is an increase in *Penicillium* and *Aspergillus flavus,* the increase is lowest in 20% CO_2 + 2% 02. 1-lydropriming for 6 hrs was found the best treatment for PusaDeepali whereas osmopriming with -1 MPa PEG was found to be the best for Pusa Synthetic when sowing is to be done immediately following the treatment. In case the seed needs to be stored for 9 months or more halopriming with 20 mM KNO_3 was found best for cauliflower seeds.

The study was conducted on two pairs of CMS (A) and maintainer (B) lines of rice vartieties Pusa-6A and Pusa-6B, and IR58025A and IR58025B.Ageing resulted in changing pattern of water uptake with longer duration of *lag* phase and decreased rate of water uptake. This was mainly ascribed to enhanced solute leakage, decrease in the stored food reserve and decreased conversion of starch to sugar upon imbibition. This led to decreased water uptake, low vigour and slow germiantion. These changes occurred more rapidly in CMS lines than maintainer lines. Decrease in the activities of antioxidant enzymes viz., superoxide dismutase (SOD), catalse (CAT) and peroxidase (PDX) and repiratory enzymes i.e. malate dehydrogenase (MDH) and cytochrome oxidase (COX) during ageing, indicated their crucial role in the process. However, higher level of activities of PDX, MDH and COX in the seeds of maintainer line, especially after 12 month of storage, when the difference in

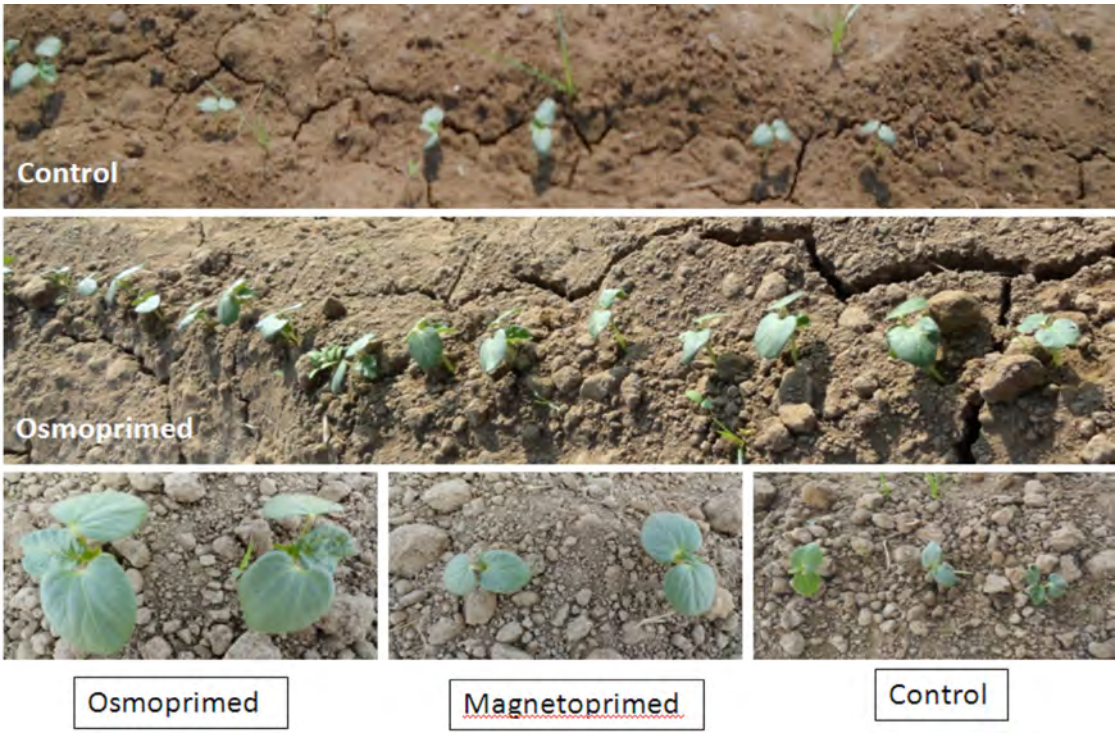
germination between CMS and maintainer line was clear suggested their role in differential storability. Potential germination and longevity/vigour was found not be highest at physiological maturity in rice, but was achieved at a later stage of seed maturity. Owing to early attainment of field maturity, CMS lines should be harvested earlier than their respective maintainer lines for achieving highest seed quality.

Seed Quality Enhancement

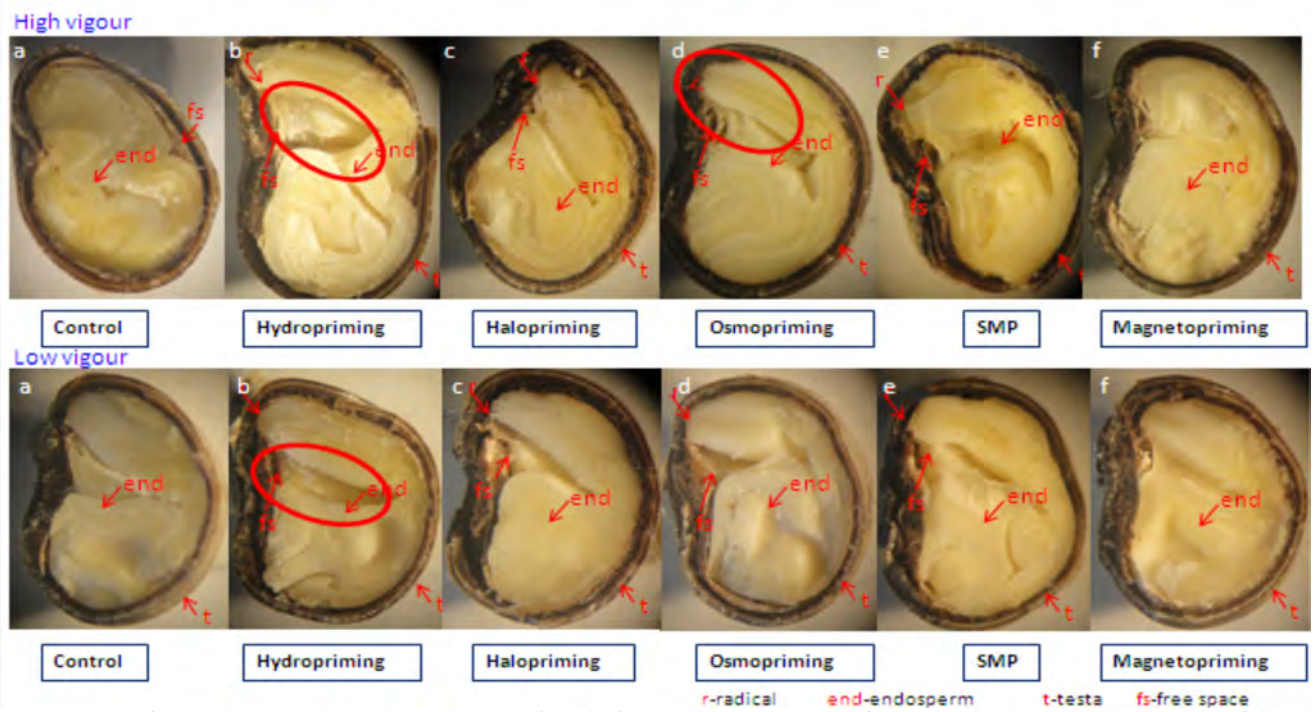
Heat stress was found to have a significant influence on the wheat seed quality parameters. Germination decreased from 94.49 to 84.94%, whereas vigour Index I reduced from 2645.4 to1885.4 with delayed sowing (increased heat stress) Vigour Index II also followed the similar trend. Seed harvested from heat stressed environment showed a significant decline in the seed quality parameters when stored at room temperature; however it was able to maintain the minimum seed certification standards, during storage under cool and dry conditions. For heat stress mitigation, seed priming and foliar sprays using different chemicals were tried. Based on the speed of germination, three better seed priming treatments were identified *viz.,* halo priming with KH_2PO_4 (1%); osmopriming (PEG 6000 at– 1Mpa) and plant growth regulator *ie.,* GA_3@50 ppm; however, seed priming with GA_3 was found to be the best? The study showed the enhancement in various crop morphological and seed quality parameters with seed priming. Foliar sprays using different chemicals, at vegetative (booting stage) and seed filling stage, significantly decreased the MDA content, EC values and hydrogen peroxide activitiy, whereas increased dehydrogenase activity. Hence, all foliar sprays studied, could successfully mitigate the effect of heat stress, however, salicylic acid @400ppm was found to be the most effective among the various chemical used.

It was also found to increase the activities of antioxidant enzymes *ie.,* catalase, peroxidase, superoxide dismutase and reduce the activity of hydrogen peroxide (free radical) in onion seeds. The results of post-priming storage indicated that the extent of seed deterioration was more in case of fresh seed lots of both varieties as compared to control. However, the carryover seed lot showed better performance as compared to control (unprimed seed but reduced performance was observed in four months stored seeds. Simultaneously, accelerated ageing test was conducted on the seeds of different priming treatments. The reduced performance of primed seed as compared to control was observed; the former were more susceptible to oxidative damage. It was concluded that solid matrix priming (SMP) was identified as the best treatment in terms of higher speed of germination and reduced mean germination time, resulting in enhanced seed performance and planting value of onion seeds.

In okra, 15 seed lots were tested under laboratory and field condition, to assess the relationship between different vigour tests (germination, pot germination, vigour indices, controlled deterioration, accelerated ageing, saturated accelerated ageing, electrical conductivity, complex stress vigour test) with field emergence. The controlled deterioration test (24% mc, 45°C, 24 h) could be recommended as the vigour test for prediction of field performance in okra. The effect of different seed enhancement treatments *i.e.* hydropriming (17 h, 25°C), halopriming (NaCl, 24 h, 25°C), osmopriming (PEG 6000, -1.0 MPa, 24 h, 25°C), solid matrix priming (vermiculite, 24 h, 25°C), magnetopriming (150 mT, 1 h) and seed coating (Incotec Disco Agro SP Red Guard L-200G) along with control was undertaken in *spring-summer* season. Among the seed quality enhancement treatments, osmopriming followed by magnetopriming was effective in improving field emergence, speed of emergence, seedling shoot and root growth, plant height, total chlorophyll content, fruit & seed yield and induced early flowering. The effect of seed enhancement treatments was more effective under sub-optimum conditions and in low vigour lots. In seed enhancement treatments involving hydration, improvement in membrane integrity was evident by reduction in electrical conductivity, water soluble sugars and total free amino acids. The activity of dehydrogenase, á-amylase, protease, esterase activity was higher in osmoprimed and magnetoprimed seeds as compared to control. The seeds stored at 16±2°C exhibited significantly higher quality than storage at 30±2°C in low and high vigour lots. Among the treatments, osmoprimed seeds maintained high



Effect of seed enhancement treatments on early seedling emergence and growth under field condition



Compound microscopy (300X) photographs showing the internal morphology of okra seed. (a) Dry control seeds without any free space; embryo and endosperm are filling the whole seed. (b) hydroprimed seed with large amount free space after dehydration; (c) Haloprimed seed with moderate free space; (d) Osmoprimed seed with moderate free space; (e) Solid matrix primed seeds with moderate free space; (g) Magnetoprimed seeds without any free space.

vigour throughout the storage period in high vigour lot. However, 6 and 9 months from storage, treated seeds of low vigour lots started showing decline in their quality. Overall, the decline in germination was moderate up to 9 months, which declined sharply up to 12 months especially in low vigour lot, however, high vigour lots of treated seeds maintained seed germination above certification standard (>65%) even after 12 month of seed storage. It could be concluded that osmopriming with PEG (-1.0 MPa, 24 h) could be used for better field performance and higher yield in okra.

Role of seed enhancement treatment in Indian mustard of three mustard varieties each; CS 54, RGN 73 and Pusa Mustard 28 tolerant and NRCDR 2, Bio YSR and Pusa Karishma susceptible to salt, moisture and temperature. Stresses influenced various seed quality parameters in all treatments. In moisture stress, the variety RGN 73 showed significantly higher first count, germination percentage, shoot length vigour index I and II than Bio YSR. Under temperature stress condition varieties showed no significant difference except vigour index I, root length and shoot length. PM 28 resulted in higher vigour index I and shoot length than Pusa Karishma. In salt stress variety CS 54 showed significantly higher germination percentage, root length, vigourindex I and II than NRCDR 2. The maximum first count percent, germination percent, root length, shoot length, seedling dry weight, vigour index I and II and minimum percentage of abnormal seedlings, hard seeds and dead seeds were observed in Salicylic acid, KNO_3 and hydropriming seed treatments. Salicylic acid had significant effect on all above parameter other than KNO_3 and hydropriming under all stress conditions.Effect of controlled deterioration on quality enhanced seeds showed that variety CS 54 had significantly higher percent germination and normal seedling than all other varieties, whereas NRCDR 2 had significantly higher abnormal seedling percentage than all other varieties.

Magnetic treatment is a potential physical seed enhancement treatment reported to improve field emergence, seedling vigour, seed yield and quality. A comparison was made between thepulsed (100 mT for 2h with 6 min. interval) and static(200 mT for 1h) magnetic treatmentson two differentially aged seed lots of Pusa composite-3 in spring-summer season. Significant increase in field emergence, speed of emergence, leaf area and root characteristics were observed in both the treatments but pulsed magnetic treatment was more effective than static magnetic field. A noticeable increase of root growth in terms of total root length, surface area, mean root diameter and root volume was observed in magnetically treated seeds as compared to control in germination test under laboratory conditions. There was reduction in the electrolyte leakage (13.82%) and increased in the activities of dehydrogenase and scavenging enzymes (catalase, superoxide dismutase, ascorbate peroxidase, peroxidase) in magnetically treated seeds as compared to control seeds. Higher and early free radical production and increased levels of hydrogen peroxide ions promoted early seedling emergence.It is concluded that both static and pulsed magnetic seed treatments are effective as pre-sowing treatment for improving field performance of maize both in *kharif* and *rabi* season.

For seed enhancement of seeds of speciality maize varieties (HQPM-l, VL Amber pop com, Win orange sweet com) and common maize (Parkash) were subjected to pre-sowing seed enhancement treatments. All treatments showed beneficial effects to varying degrees, in total emergence, speed of emergence, seedling weight (fresh and dry weight), plant height and seed yield. These treatments also advanced the days to flowering by 1-2 days over control. Most effective seed enhancement treatments identified were seed treatment with fulvic acid, hydropriming with or without thiram, $KN0_3$, ceolomic fluid and magnetic field in improving the above traits. Though all the genotypes showed significant improvement with treatments, Win sweet com and Parkash were more responsive than others, for field emergence, its speed and early seedling growth. To understand the physical, physiological and biochemical processes associated with seed enhancement, six treatments *viz.,* hydropriming (17 hr/20°C), slow hydration using hydrogel, halopriming (0.3% KNO_3), priming with 0.1 % fulvic acid, ceolomic fluid (17 hr/20°C) and magnetic field treatment (1000 G for 2 hr) were given to seeds of different vigour levels of above varieties. Increase in emergence, speed of emergence, seedling dry weight and root length, width and surface area were recorded in treated seeds as compared to control. There was a marginal reduction in the leakage of electrolytes and water soluble sugars from treated seeds. Increased activity

of germination related enzymes *viz.,* amylase. dehydrogenase and superoxide dismutase was recorded in treated seeds. There was also increase in protein and DNA content in the treated seeds as compared to control. The water absorption-desorption patterns and water activity in primed seeds showed modification of seed water-binding properties and reorganization of seed water in treated seeds. Pimed seeds, during storage up to six months at 25°C, showed no significant difference in seed quality.

Carrot and onion seeds were osmo-primed using PEG 6000 at -0.5, -1.0 and -1.5 MPa potential for 2, 4 and 6 days. Solid matrix priming (SMP) of the seed was performed by mixing seed with moist vermiculite followed by incubation for 12, 24, 36 and 48 hr at 15° and 20°C. Osmo-priming using PEG 6000 at -1.0 MPa for 2 days improved germination by 17.8 and 14.3 per cent and field emergence by 12 and 13 per cent over control in carrot and onion, respectively. Solid matrix priming for 24 hr at 20°C also improved germination by 14.3 per cent and field emergence by 12.2 per cent over control in carrot. In onion, SMP for 24 hr at 20°C also improved standard germination by 13 % and field emergence by 11.3%. The results suggested that non-standard temperatures of 15°C and SSAA (RH 75%) in carrot and standard germination, AA and SSAA (RH 75%) in onion are useful vigour tests to predict seedling emergence. Osmo- and solid matrix priming can be successfully used to improve seedling emergence in carrot and onion.

Studies on various vigour parameters including accelerated ageing and electrical conductivity of seed leachate in relation to field emergence and storability were conducted in cotton seed. The treated seeds were evaluated for first count, germination (%), seedling length, seedling dry weight, vigour index I and II, field emergence, field emergence index and speed of emergence. The above studies revealed that the H-1226 was having relatively higher vigour compare to H-1117. The standardization of magnetic energy treatment for seed quality enhancement for cotton revealed that 2500G for 1 hr exposure treatment gave higher root length and seedling length. Among all the seed enhancement treatments, the best treatments were royalflo @ 5ml/kg and thiram @ 2gm/kg + imidachloprid @ 7.5ml/kg as they significantly increased the first count, germination (%), vigour index I, field emergence, field emergence index and speed of emergence compared to control. They also maintained higher first count, germination (%), vigour index I, and vigour index II after 6 months of storage. The hydro priming treatment significantly increased seedling dry weight and vigour index II and priming

Higher returns from cucumber (Pant Shankar Khira-1) by growing crop under insect proof net house (during kharif season): The benefit-cost ratio was higher (1:2.25) than naturally ventilated polyhouse (1:0.37) and open field condition (1:1.13)

treatment with ceolomic fluid + thiram + imidachloprid revealed higher first count. Among different polymer treatments, polymer along with thiram or imidachloprid has shown good results than polymer alone. Electrical conductivity showed significant negative correlation with most of the vigour parameters studied. Hence it is suggested that seeds treated either with royal flo or thiram + imidachloprid can be used for enhancing the planting value in cotton.

Based on seedling vigour index, accelerated ageing, controlled deterioration, multiple stress (a new test developed in this lab) and EC of seed leachate were significantly correlated with the field emergence and storability (upto 12 months) in onion, the highest significant correlation was obtained with the first count (r = 0.924) and final count (r = 0.937) of the standard germination test. In case of soybean, the highest correlation was obtained between the field emergence and germination after accelerated ageing test (r = 0.775), though the other vigour tests, as well as the first and final counts of standard germination were also significantly correlated. However, for predicting the storability of soybean seeds, highest significant correlation was obtained between the final count of standard germination test and the germinability after storage. Hence, AAT and standard germination can be used for predicting the planting value and storability of soybean seed. Based on the comparative evaluation of 12 and 19 seed lots of onion and soybean, respectively, it was observed that seed lots recording more than 80% (onion) and 85% (soybean) germination after multiple stress test (MST) or more than 50% (onion) or 60% (soybean) germination after AAT, retained more than 70% germination at least upto 9 months and recorded at least 50% emergence in the field. Several seed treatments, including priming, polymer coating and fungicidal and botanical applications, were found to enhance the germinability and vigour of soybean and onion seeds. Among these, solid matrix priming/conditioning was most effective both as pre-sowing and mid-storage corrective treatment. This was followed by halopriming (0.3% KNO3) in onion and coating with Royalflo or polymer + thiram in soybean. A linear relationship between the activities of the hydrolytic, respiratory and antioxidative enzymes and vigour status of onion and soybean seeds during storage established their role in maintenance of seed quality.

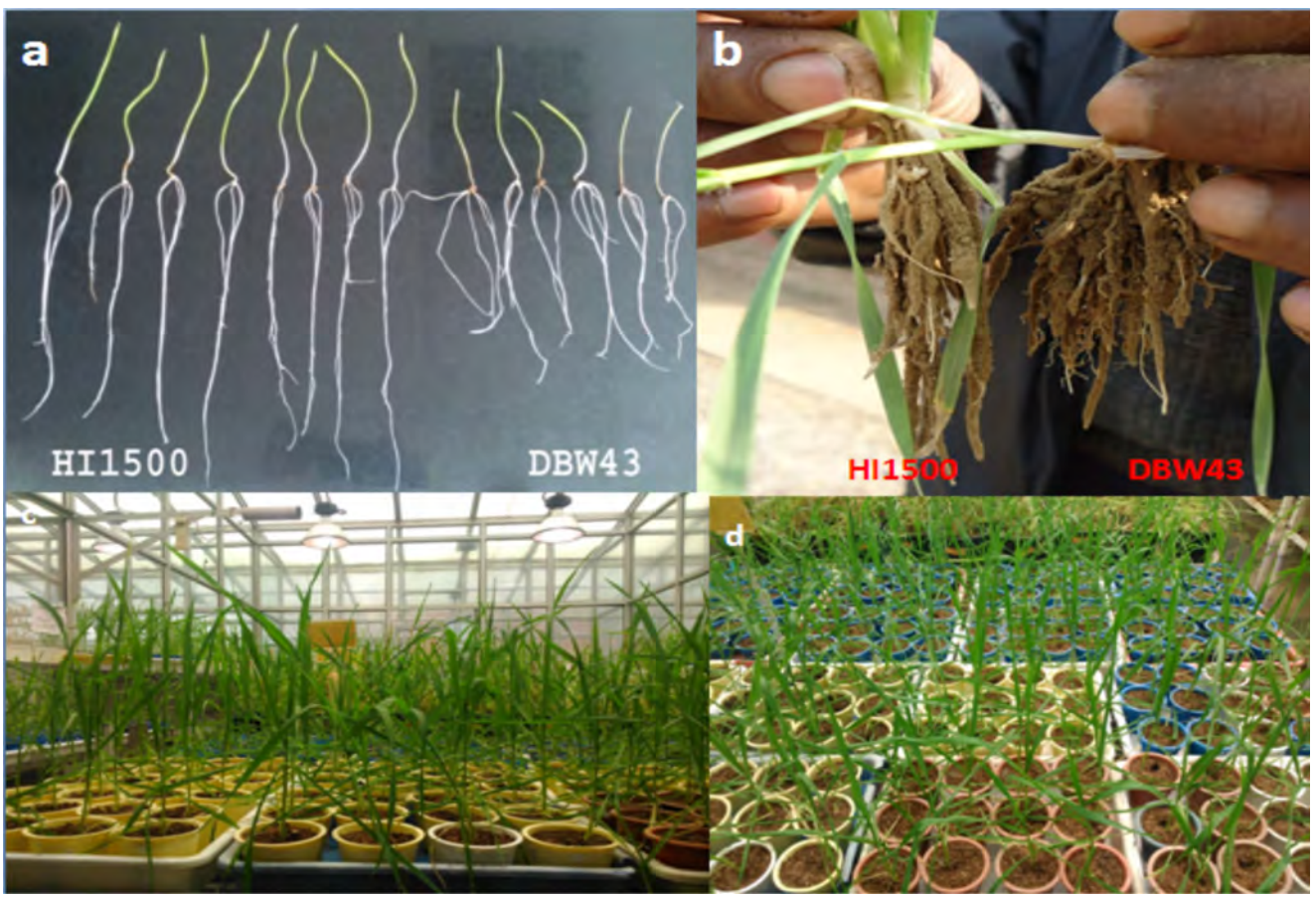
The initial seed quality parameters of onion were investigated and depending upon the germination percentage and incidence of mycoflora, the seed lot from Rajkot was selected for further studies. Further, the seeds were subjected to specific gravity separation and divided into five different grades. The specific gravity separation improved the seed quality parameters significantly over the original seed lot. The grades having germination percentage above Indian Minimum Seed Certification Standards (70 %) were mixed and used for further experiments. Also, the seeds were primed with different chemicals and the best treatment was selected. Priming treatments were found to enhance the speed of germination and other seed quality parameters to a significant extent. The seed germination and vigour indices were estimated at an interval of three months and it was observed that there was a gradual decline in both the parameters due to seed deterioration over the storage period. However, the extent of deterioration was lesser in case of polymer coated seeds owing to its specific property of regulating the moisture content. The polymer coated seeds were also associated with lower moisture content and lesser incidence of seed borne fungi as compared to the uncoated ones. The efficacy of seed treatments was evaluated through seedling emergence at the end of the storage period and it was found that the fungicidal seed treatments proved to be superior in the maintenance of the seed quality. The bioagents, especially *Trichoderma harzianum*, were also found effective.

The study of genetic purity testing was conducted in one intra-hirsutum hybrid (Nathbaba) and one intra-arboreum hybrid (PKVDH-1), 21 commercial seed lots were taken for vigour assessment and three lots (high, medium and low vigour were used for seed quality enhancement. Among the three fractions of proteins used for testing genetic purity of hybrids by electrophoresis, tris-soluble protein fraction was found useful in testing the genetic purity of hybrid PKVDH-1. The electrophoresis profiles of trissolubule proteins, globulins and methanol precipitated fractions failed to differentiate the parents from the hybrid in the Nathbaba hybrid and hence not useful for genetic purity testing. The RAPD analysis was found useful in differentiating the two parents, as well as the female parents from their respective hybrids. Primer OPA-07 and OPA-10 were selected for genetic purity testing of commercial seed lots of Nathbaba and PKVDH-1 hybrids respectively. The results of the genetic purity analysis were comparable to the field grow out tests, which proved the ability of reproducible markers such as OPA-07 and OPA-10 in genetic purity testing of intra-hirsutum and intra-arboreum hybrids.

Studies on various seed vigour parameters including accelerated ageing test and electrical conductivity of seed leachate in relation to field emergence and storability were conducted on chickpea. All the varieties of fresh

and old seed lot s were subjected to 11 seed enhancement treatments. The treated seeds were also evaluated for field emergence, field emergence index and speed of emergence in addition to all other vigour parameters studied. The initial assessment revealed that the materials were diverse for the various vigour parameters studied. Among all the treatments, it was found that thiram alone or in combination with polymers gave significantly high field emergence over control. The kabuli varieties (Pusa 1053 and Pusa 1108) showed sharp decline in field emergence (less than 20% in control) as compared to desi varieties (Pusa 256 and Pusa 2028). It was also observed that the old seeds were more responsive to treatments than fresh seed and the priming treatments were specific to varieties. Field emergence index was also improved significantly in the thiram or polymers + thiram treated seed. The electrical conductivity (EC) was recorded on untreated seed. It was found that EC values were significantly high in old seed lots. Accelerated ageing test was also done at 40^0 C and 100% RH for period of 36, 48 and 72 hrs. The correlation of various vigour parameters revealed that EC was significantly correlated with all the vigour parameters followed by accelerated ageing test. In the studies of storability, it was observed that storage of primed seed led to increased number of abnormal seedlings more frequently in kabuli varieties, Pusa 1053 and Pusa 1108 of old seed lots.

Two RIL populations developed for drought tolerance in wheat were used to screen for seed vigour traits along with validation of known QTLs reported for their contribution to drought tolerance. Experiments were conducted in laboratory, glasshouse and field conditions under control and water-deficit stress or rain-fed conditions. In first set of RIL (WL711× C306) field experiment was conducted in two consecutive *rabi* seasons from 2012-13 and 2013-14. Parent C306 performed better than the parent WL711 for NDVI, CTD, and SPAD-



Variation in root length of Parent DBW43 and HI1500, b: variation of root in parents under field condition c & d: seedling growth in pot under glasshouse condition

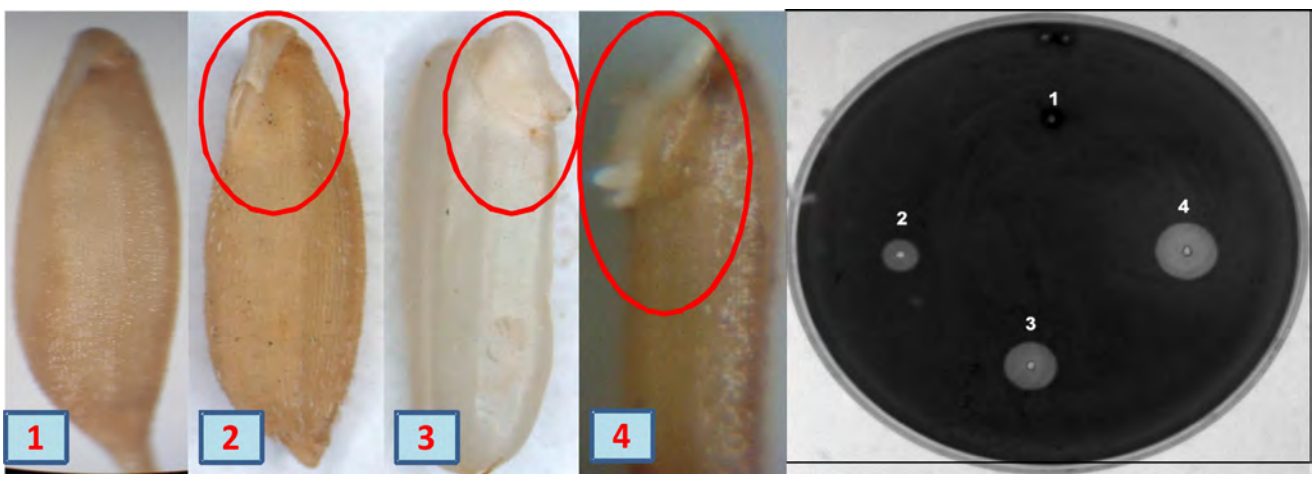
readings in water-deficit stress condition. Seed weight, seedling traits, like shoot length, primary root length, secondary root length, seedling vigour indices (SVI I) and (SVI II) were higher in C306 indicated the drought tolerant characteristics. Root weight density in this population at different soil depths (0-15 cm, 15-30 cm and 30-45 cm) showed positive association between root weight density (at all the depths) with grain yield. Reduction in grain yield was higher in WL711 than C306 during both the seasons, under rain-fed condition. This indicated that C306 was able to sustain under varying intensity and duration of water scarcity environments during both the cropping seasons. Direct and significant association was established between grain yield with biomass, harvest index, 1000-seed weight, root weight density and total root dry mass. In second set of RIL population, parent HI1500 exhibited higher root length than DBW43. Total root length as measured with the help of root image analyzer in DBW43 was higher than HI1500, as there were more roots. Further, root and shoot parameters were evaluated both under field and glasshouse conditions under irrigated and simulated water-deficit stress by withholding irrigation water to 50%. Under PEG solution and normal condition parent HI1500 showed higher speed of germination than those to DBW43, further due to water-deficit stress coleoptile length increased under PEG solution in HI1500 parent thus these traits were highly adapted to the drought tolerance. Under irrigated and rain-fed conditions, significant and direct associations were established between GY with BIO, TSW and HI. Seed weight was found to have significant association with root length, root volume, root surface area, average root diameter, number of root tips and root dry mass. Out of many QTLs reported for drought tolerance from earlier reports three QTLs were validated in second set of RIL population for example, Xbarc184 (root length), Xbarc156 (root numbers) and CFA2099 (coleoptile length); these QTLs could be further used for marker assisted breeding for drought tolerance in wheat. The present study indicated important key traits such as root length, root number and root volume, speed of germination, vigour indices I and II, could be considered for selection of drought tolerant lines under laboratory and glasshouse conditions.

The study was conducted for comparative analysis of conventional and quality type of Indian mustard genotypes differing in seed coat colour, based upon the imbibitional behaviour, enzymatic and non-enzymatic antioxidants and bio-molecules affecting seed germination and vigour. The material consisted of eight varieties of Indian mustard, in which four genotypes were of each conventional and quality types. The seed of these genotypes varied for seed coat colour *i.e.* four black and yellow. Thus, the experimental material had two genotypes each with black and yellow seed coat from conventional as well quality groups. The results of the study showed significant differences for imbibition and electrical conductivity (EC) amongst the black and yellow seeded conventional or quality types of genotypes. The results indicated that imbibitional behaviour was significantly correlated with seed colour, melanin and phenol content. Yellow seeded genotypes imbibed faster than those to black seeded genotypes as well quality type genotypes showed higher percent of imbibition (119.08%) than those to conventional type genotypes (81.20%) during 24 hr of water imbibition. Yellow seeded genotypes which had low melanin and phenol content, with weak seed coat, imbibed faster as well had higher leakage. Significant differences between yellow and black seeded genotypes were noticed for EC, melanin, phenol, ascorbic acid, tocopherol content and antioxidant enzymes. Mean EC from fresh yellow seeded genotypes was 95.54 ìS/cm/g of seed increased to 170.82 ìS/cm/g of seed, similarly EC from fresh black seeded genotypes was 61.18ìS/cm/g of seed increased to 97.79ìS/cm/g of seed under controlled deteriorated (CD) seeds. The amount of melanin (58.80 U) and phenol content (11.11 mg GAE g^{-1}) in black seeded genotypes was significantly higher than yellow seeded genotypes (29.25 U and 6.86 mg GAE g^{-1}), respectively. The mean value for ascorbic acid and tocopherol content were higher in black seeded genotypes (28.42 mg/100 g and 50.26 µg/g of seeds) than those to yellow seeded genotypes (23.11mg/100g and 43.37µg/g of seeds), respectively. The superoxide dismutase (SOD) and peroxidase (POD) activity was more efficient in black over yellow seeded genotypes as well in conventional type Indian mustard.

In rice non-structural carbohydrate (NSCs) get accumulated in stem during pre-anthesis when there is no active reproductive sink. Later on NSCs are remobilized to developing seed when supply of photosynthates becomes limiting factor due to onset of natural senescence. This study was conducted with 10 rice varieties varying in their maturity period (crop duration) and drought tolerance (upland and lowland cultivation), under

irrigated conditions. Dry mass accumulation during late embryogenesis up to physiological maturity indicated that genotypes maintaining higher remobilization of NSC are able to retain linearity in accumulation of dry mass during seed maturation. Association between NSC remobilization and biomass accumulation rate (r=0.50) was established and efficient genotypes were identified, such as, Vandana, Sahbhagi Dhan and Satyabhama. These genotypes are also known for the potential for drought tolerance. Seed germination and vigour including total amylase activity, were determined in fresh-harvested seeds and one month stored seeds, and influence of remobilization of NSC during seed maturity was evaluated. Initial seed germination percentage, speed of germination, seedling vigour index II, which is based on dry mass accumulation, and total amylase activity up to 48 hr after imbibitions recorded were higher in efficient genotypes. In addition, harvest index was associated with speed of germination (r=0.61). This indicated that higher remobilization of NSC had decreased starch content in stem reserves due to higher supply of sucrose to developing seed. It is postulated that sucrose might be required for seed development during desiccation for 'glassy-state' formation. It is concluded that genotypic variation in storage and remobilization of NSCs from stem reserves be utilized for enhancing both grain yield and seed quality, however further investigations are required to illustrate exact role of sucrose during seed desiccation.

To test this hypothesis and the effect of gibberellins (GA) and Abscisic acid (ABA) on enzyme activity, studies were conducted on ten rice genotypes comprising of five early germinating (which have taken less than 30 hours for radicle emergence) and five late germinating (which have taken more than 44 hours for radicle emergence) genotypes. Activities of galactomannan hydrolysing enzymes (viz., α-mannanase, α-mannosidase and α-galactosidase) increased with germination in both early and late germinating rice genotypes. The average enzyme activities of the genotypes was higher in early germinating group in comparison with the late germinating group for all the three enzymes and at all the three stages of germination (*viz.,* dry seed, lemma rupture and radicle emergence). Among the enzymes, activity of α-mannosidase was found higher in dry seeds of both the group followed by α-galactosidase and α-mannanase. GA supplement (at 50, 100 and 200 µM) in the germinating medium has increased the activities of all the three enzymes that manifested in faster lemma rupture and radicle emergence, in both early and late germinating rice genotypes, by 3-5 hrs and 8-12 hrs respectively. The increase in α-galactosidase activity with increased concentrations of GA was marginal in



Lemma rupture occurring during paddy seed germination, due to the growth potential or expansion of the embryo, which is under the control of GA and is unaffected by ABA

both the group and hence it is hypothesized that GA is facilitating in attaining the required threshold activity of α-galactosidase in shorter time, which has manifested in faster germination of all genotypes. Supplement of ABA (at 50, 80 and 100 ìM) in the germination medium has decreased the activities of all the enzymes due

to which time taken for radicle emergence was enhanced. However, no changes were observed with respect to time taken for lemma rupture, which corroborates the fact that lemma rupture could be due to expansion of embryonic axis or cell elongation which is under the control of expansins regulated by GA.

The study was conducted to evaluate the comparative effect of hydropriming on the performance of low and high vigour lots of maize hybrid PEHM 5 and its parental lines (Female – CM 150, Male - CM 151). Seed performance was evaluated on the basis of physiological and biochemical parameters viz., germination test, vigour index, electrical conductivity of seed leachate and antioxidant enzyme activities. Genetically pure and fresh seeds of low and high vigour lots of maize hybrid PEHM 5 and its parental lines (Female – CM 150, Male - CM 151) were subjected to hydropriming for 30 h. Seeds were hydroprimed in two sets, where in first case, seeds were dried back near to original moisture contents under shade at room temperature at 25±2°C after treatment. In the other case, seeds were only surface dried using blotting paper and evaluated immediately. Both the hydropriming treatment strategies improved the germination, seedling growth and reserve metabolism. Use of surface drying was more effective for maize seed invigoration, as evident from earlier and synchronized germination. Moreover, shoot and root length, seedling dry weight, α-amylase activity, protease, esterase, soluble sugars and dehydrogenase activity were also improved. Results suggested that surface drying, rather than re-drying close to original weight, was more effective. In addition, the activities of antioxidant enzymes of unprimed and primed seeds in hybrid and its parental lines were determined to investigate their relationship with germination characteristics. A steady increase was noted in the activities of four key antioxidant enzyme systems; superoxide dismutase (SOD), peroxidase (POX), catalase (CAT) and glutathione reductase (GR) in both high and low vigour seed lots of hybrid and its parental lines upon hydropriming irrespective of non-dried or redried strategies.

Acknowledgement

We wish to thank all the Faculty Members of the Division of Seed Science and Technology for extending support and cooperation in preparing the research contributions of M.Sc. and Ph.D. students made during the period of 2007-16.

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M.Sc. and Ph.D. theses submitted by the students in the Division of Seed Science and Technology, ICAR-Indian Agricultural Research Institute, New Delhi 110012 during the period of 2007-2016.

School of Crop Protection

Agricultural Chemicals

Irani Mukherjee and Dhruba Jyoti Sarkar

Introduction

In India, crops are affected by over 200 major pests, 100 plant diseases, hundreds of weeds and other pests like nematodes, rodents, etc. To minimise the losses due to these pests and diseases, use of pesticides is imperative. During past decades tremendous progress has been made in the field of crop protection products and technologies. Newer and safer molecules of agrochemicals and biopesticides, pesticide residue products, safety to the environment have gained more importance. But under the WTO regime, access to new agricultural chemicals and technologies discovered elsewhere in the world is difficult. India therefore must strengthen its crop protection and pesticide development programme to keep pace with the goal of sustainable agriculture.

Recognising the importance of pesticides in agriculture sector Division of Agricultural Chemicals was created in 1966. Since then, the Division has generated substantial knowledge and information on agrochemicals in general and crop protection chemicals in particular, for reference and use by the national system. The Division has proved a trendsetter in planning and execution of basic and applied research of national importance in multifarious domains of development, formulation and safety aspects of agrochemicals *viz.* pesticides and pesticide adjuvants, super absorbent hydrogels and composites, nitrification inhibitors, hybridizing agents, nutraceuticals, plant growth regulators, smart agro-inputs delivery systems, nano products, new analytical methods for pesticide residues, etc. Division has been registered as a source of environmental information in the International Directory of Source of the United Nations. Divisional laboratories are well equipped with advanced instruments and various type of services offered besides research and teaching. From 1989, division has been granted the status of an independent teaching discipline, and IARI has started conferring M.Sc. and Ph.D. degrees in Agricultural Chemicals. The mandates of the Division are:

- Development of agrochemicals from natural and synthetic resources
- Agrochemical formulation, research and development
- Safety evaluation of plant protection schedules on agricultural crops
- Develop human resource of excellence

Significant Student's Research Achievements

Students of this Division have been contributing significantly in the field of agrochemical research under the supervision of the consummate faculty members and the Professors of this Division. Some of the major research contribution of the students is discussed in theme wise and presented below.

Agrochemicals Developments

Eighteen new substituted amides were biocatalytically synthesised using *Candida antartica* lipase at 60-90°C without the use of either solvent or activating agents. The yield of compounds varied from 77-82%. These compounds were tested for nematicidal activity against J_2 of *Meloidogyne incognita*. Amongst the eighteen compounds, three compounds viz. N-propyl-butyramide, N-propyl-pentamide and N-propyl-hexamide showed significant activity with LC_{50} 67.5, 83.5 and 96.5 ppm, respectively. The structure-activity relationship revealed

that optimum chain length appears to be 4-carbon (Butyl) and further increase in chain length leads to decrease in activity.

The chemical constituents from *Stevia rebaudiana* were isolated and identified by GC-MS and tested for fungicidal and plant growth regulatory activity. Hexane extract of dried leaved showed maximum antifungal activity *Rhizoctonia bataticola* (ED_{50} = 26.62 ppm) and *Fusarium oxysporum* (ED_{50} = 41.14 ppm) and *Sclerotium rolfsii* (ED_{50} 40.61 ppm) as compared to water, ethyl acetate, methanol and dichloromethane extract. Maximum increase in seed germination, shoot length and root length was observed with hexane extract as compared to dichloromethane and methanol extract when tested for rice and lentil seedlings. Germacrene D, 1-naphthalenol, humulen-6, 7-epoxide, and β-ionone showed activity against *F. oxysporum* (ED_{50} – 52.88 ppm) and *R. bataticola* (ED_{50}-96.50 ppm). Extraction of freeze-dried *S. rebaudiana* leaves with hexane, followed by a series of chromatographic separations, yielded a mixture of sterebins, I, J, K, L, M and N besides several other compounds. Maximum antifungal activity was exhibited by sterebin I and K against *F. oxysporum* with ED_{50} = 26.80 and 36.96 ppm, respectively, whereas sterebin L was found to be effective against *R. bataticola* (ED_{50} = 59.90 ppm). Steviol, isosteviol, ethyl ester of isosteviol, 1,3-diol of isosteviol ethyl ester of 1,3-diol of isosteviol and oxime ethyl ester of 1,3-diol of isosteviol and oxime derivative of isosteviol synthesized from stevioside were tested for antifungal and plant growth regulatory activity. Maximum antifungal activity as well as plant growth regulatory activity was exhibited by isosteviol derivative.

Substituted amides and amines have been reported to possess antifungal, herbicidal, antioxidative, insecticidal, antiviral, nematicidal and antibacterial properties. Synthesis of these compounds have mainly been achieved by conventional methods, which generally involve the generation of a reactive carboxy derivative, either an acid chloride or anhydride, followed by aminolysis with amine. Enzyme-catalyzed organic reactions have provided a great impetus to organic synthesis during the last two decades. Enzymes, especially lipases, are known for their low cost and great tolerance towards their substrates. Environmentally benign character of the enzymatic processes is desirable for large scale industrial applications. A series of N-alkyl substituted amides, based on various phenolic acids, namely salicylic acid, 3-hydroxy cinnamic acid, p-coumaric acid, caffeic acid, ferulic acid, o-coumaric acid and cinnamic acid, have been synthesized by the condensation of equimolar amounts of phenolic acids with different alkyl amines viz propyl amine, hexyl amine, heptyl amine, undecyl amine, hexadecyl amine and octadecyl amine in presence of *Candida antarctica* lipase at 60-90°C in 16-20 hours. The reactions were carried out in a non - solvent system without the use of any activating agents. All the products were obtained in appreciable amounts and the yields for different compounds varied between 76% - 84%. The synthesized compounds were characterized using spectroscopy techniques namely IR and NMR (^{1}H and ^{13}C). All synthesised compounds were tested against rice root knot nematode, *M. graminicola*. Although all the compounds possessed promising activity, N-hexyl-salicylamide 9b, N-hexyl-2-hydroxycinnamamide 14b and N-hexyl-4-hydroxycinnamamide 11b were found to be more potent with LC_{50} values of 54.75, 61.791 and 74.11 ppm respectively. But they were found less active than carbofuran. Increasing chain length beyond six C-atoms causes a decrease in activity significantly. Order of nematicidal activity among different phenolic group was found to be salicylamides > 2-OH cinnamamides > 4-OH- cinnamamides > 3-OH- cinnamamides > cinnamamides > ferulic amides > caffeic amides.SAR study revealed that all the plant growth parameters (shoot length, shoot weight, root length and root weight) was found to follow the order 9b > 14b> 11b > SA both at 7DAI and 14DAI. As the nematicial activity increased, plant growth parameters also increased. 9b @ 500 ppm was found to be most effective with 64.29% reduction in gall formation. All the compounds were found more effective at 500 ppm and showed better results than the commercially recommended salicylic acid. The in vitro bioassay revealed that all synthesised compounds showed the activity against the BPH but the compounds 11a, 14a, 11b, 11c, 14b, 9a, 11d, 14c, 10a and 15a were more potent with LC_{50} values of 55.43, 63.4, 70.81, 73.14, 79.60, 83.38, 96.48, 97.39, 101.20 and 106.74 ppm respectively. But they were found less active than fipronil. It was found that increase in alkyl chain beyond propyl significantly decreased the activity of amides against *Nilapravata lugens*. Order of insecticidal activity among different phenolic group was found to be 4-OH- cinnamamides > 2-OH cinnamamides > salicylamides > 3-OH- cinnamamides > cinnamamides >

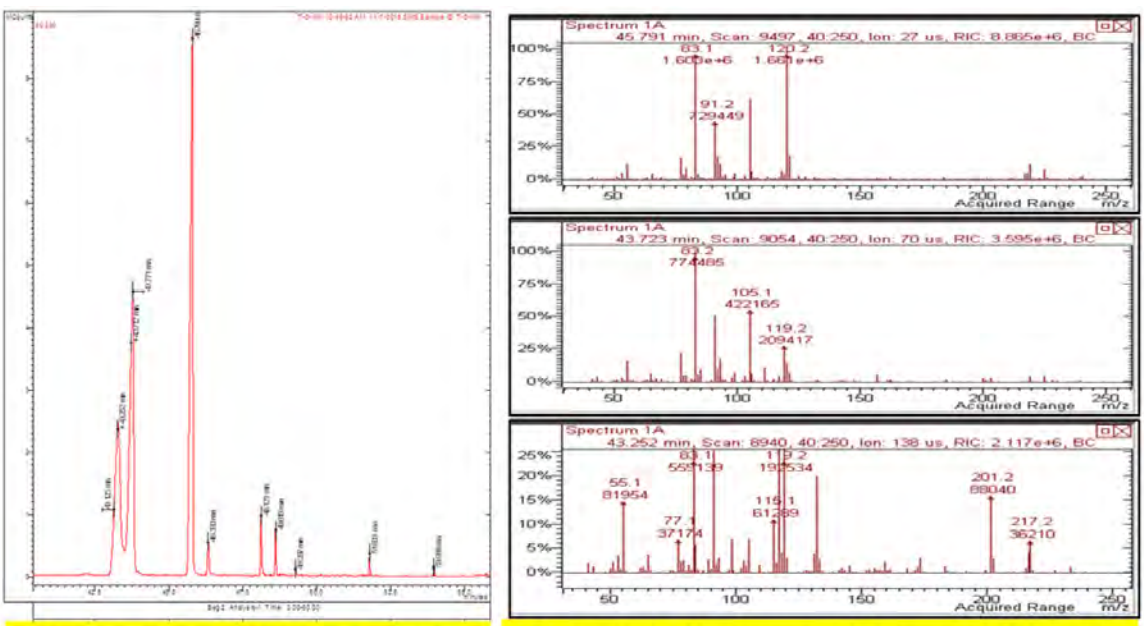
ferulic amides > caffeic amides.In pot experiment, 11a showed the highest activity with LC_{50} 94 ppm followed by 14a, 11b, 11c, 14b, 9a, 11d, 14c, 10a and 15a with LC_{50} values of 104.15, 118.01, 131.80, 150.13, 167.73, 199.26, 224.61, 250.91 and 290.89 respectively. But all compounds were found to be less active than fipronil (LC_{50} 4.4 ppm). However, the synthesised amides can be used in rice against BPH, root knot nematode and for inducing SAR as one complete package.

To optimize the structure to improve the bioactivity of azomethine (Schiff Base) derivatives, 23 novel Schiff bases derived from 2, 4, 5-trichloroaniline were synthesized and characterized using spectral techniques. These molecules were further explored for their antifungal activity against *R. bataticola* and *S. rolfsii* and nitrification inhibitory activity under laboratory conditions. All test compounds were highly effective as nitrification inhibitors. The potent compounds emerged from the series were (2,4-dimethoxybenzylidene)-(2,4,5-trichlorophenyl)-amine (C23) (4-isopropylbenzylidene)-(2,4,5-trichlorophenyl)-amine (C21) and (3,4,5-trimethoxybenzylidene)-(2,4,5-trichlorophenyl) amine (C3). In case of antifungal activity all test compounds were highly effective against both the pathogenic fungi except (2-nitrobenzylidine)-(2, 4, 5-trichlorophenyl) amine. In general, test compounds were more effective against *R. bataticola* (ED_{50}, 9.4-66.2 µg/ml) as compared to *S. rolfsii* (ED50, 10.6-117.1 µg/ml). The maximum antifungal activity was observed with compound, (3, 4, 5-trimethoxybenzylidine)-(2, 4, 5-trichlorophenyl) amine (C3) with ED_{50} 9.4 µg/ml against *R. Bataticola*. On the other hand, (4-tert-butyl-benzylidine)-(2, 4, 5-trichlorophenyl)-amine (C-15) was best against *S. rolfsii* with ED_{50}, 10.6 µg/ml with high unit activity against *R. bataticola* also (ED_{50}, 11.4 µg/ml).

Berries of *Berberis lyceum* Royle were utilized for the extraction of a potential anticancerous compound, anthocyanin. *B. lycium* berry tissues were extracted using methanol containing 0.1% HCl. Extract was purified by passing through XAD-7 macroporous adsorption resin column and purified crystals were obtained after lyophilization. Purified anthocyanin mixture was analysed using HPLC at λ_{max} of 520nm and found two major peaks contributing more than 80% of the anthocyanins. Seven minor peaks were also observed in the chromatogram. For the identification of anthocyanidins, anthocyanin mixture was subjected to acid hydrolysis using 4N HCl followed by LC/LC-ESI-MS analysis. Two major peaks, based on mass/UV spectrum and co-elution with standard compound characterized as delphinidin-3-glucoside (43.7%) and cyanidin-3-glucoside (40.1%). Out of seven minor peaks, five peaks were characterized based on their mass fragmentation pattern and their characteristic UV spectrum. Five minor anthocyanins were delphinidin-3-rutinoside (0.7%), pelargonidin-3-rutinoside (1.36%), cyanidin-3-rutinoside (5.08%), petunidin-3-rutinoside (1.38%), and peonidin-3-rutinoside (6.19%). Apart from anthocyanins, a number of phenolics like chlorogenic acid, coumaric acid, syringic acid, caffeic acid, vanillic acid and quercetin was recorded in methanolic extract. These phenolics were identified by triple quad LC/MS/MS analysis using MRM mode. Purified anthocyanin was evaluated for its antioxidant activity using four assay techniques. DPPH assay revealed that IC_{50} value of anthocyanin mixture was 25.3µg mL^{-1}, whereas it was 26.1 and 8.1 for purple cabbage and ascorbic acid, used as positive control. FRAP and CUPRAC assay revealed that FRAP value of *B. lyceum* anthocyanins was 80.3 VCE (µg mL^{-1}) and 350.4 µM Trolox Equivalent. In MTT assay the absorbance value was recorded 0.53 at 250 µg mL^{-1} for anthocyanin from *B. lycium*, whereas, purple cabbage (positive control) showed absorbance of 0.49.In order to enhance the stability of the extracted anthocyanin, inclusion complex was developed using β-cyclodextrin following lyophilization method. Confirmation of inclusion of anthocyanin inside the core of cyclodextrin was done by ^1H NMR, IR and SEM analysis. In ^1H NMR analysis, changes in chemical shift (δ) of all the six hydrogens of sugar unit present in β CD, confirmed the inclusion. In FT-IR analysis, suppression/disappearance of peaks (2855 and 1736 cm^{-1}) in the inclusion complex confirms the development of the complex. In dissolution study, after 90 minutes, 98.4% release was recorded in anthocyanin, whereas, only 72.4% release was recorded in the developed complex.

Rhizome of turmeric (*Curcuma longa*) is one of the richest sources of bioactive compounds. Curcuminoids, the active constituent are one of the most researched topicworldwide for its broad spectrum of biological activities.

Efforts have been made to incorporate the molecules into commercial food products as a pharmaceutical and nutraceutical agent but the low bioavailability limits its application. Bioactive terpenoid constituents present in turmeric fixed oil was utilized for the improvement of bioavailability of curcuminoids. Turmeric fixed oil (yield 5.7% w/v) from rhizome powder was extracted with hexane and purified by column chromatography for separation of terpenoids. Purified oil mixture was analyzed by GC-MS and found three major peaks in the chromatogram. a, ß- turmerone was separated together and their structure was confirmed by GC- EI-MS. ar-turmerone was separated as pure peak and characterized by mass, FT-IR and ^{1}H-NMR. For the separation of curcuminoids, de-fatted turmeric powder was extracted with ethyl acetate and the extract was purified using column chromatography, followed by prep-TLC to get individual compounds (R 0.78, 0.53, 0.25). Purified curcuminoids were analyzed by HPLC using RP-18 column with PDA detector (λ_{max} 425 nm) under isocratic conditions using mobile phase of acetonitrile 0.1% formic acid (60:40, v/v). Three purified curcuminoids were characterized by FT-IR, λ_{max}, H-NMR and HRMS data. Based on spectroscopic data, the purified compounds were confirmed as 1,7-bis(4hydroxy-3-methoxyphenyl)-1,6-heptadiene-3,5-dione Or curcumin-I(C);1-(4-hydroxyphenyl)-7(4-hydroxy-3-methoxyphenyl)-1,6-heptadiene-3,5-dione or curcumin-II (DC); and 1,7-bis(4hydroxyphenyl)-1,6-heptadiene-3,5-dione or curcumin-III, (BDC). The HPLC method was validated; LOD and LOQ values were calculated as 0.27, 0.33 & 0.42 µg mL -1, and 0.80, 1.00 & 1.25 µg mL -1 for C, DC and BDC, respectively (Fig. 1). Accelerated solvent extraction experiment was conducted using ethanol, ethyl acetate and acetone as extraction solvent, keeping homogenization technique as a control. Ethanol provided maximum physical yield (9.2 %) by ASE experiment followed by acetone (6.1 %) and ethyl acetate (5.8 %), although there is significant differences between ethyl acetate and acetone. Similarly, ethanol yielded maximum extract upon extraction by homogenization. But, opposite trend was recorded in case of acetone and ethyl acetate. No significant differences were observed in ASE as well as homogenization in terms of purity of individual curcuminoids. Antioxidant assay evaluated using DPPH and ABTS assay found that IC value of curcuminoids mixture were 78.8 and 145.7 µg mL -1 respectively. The antioxidant activity of three curcuminoids, followed the order C > DC > BDC in both assays. The bioavailability study was carried out individually with each curcuminoids and along with turmerones to observe their effect on bioavailability of curcumin. In-vitro study revealed that in the



GC-chromatogram of turmerones (fraction 15-56)

TIC spectrum of β-turmerone (A), α-turmerone (B) and Ar-turmerone (C)

Fig. 1: GC chromatogram of turmerones and MS spectrum of β-turmerone (A), α-turmerone (B) and Ar-turmerone (C)

presence of turmerone and turmerone mixture the bioavailability of curcumin has increased 2 fold and more. The information might be useful for the development of improved bioavailable curcumin formulation.

Pomegranate (*Punica granatum* L.) contains exceptional amounts of polyphenols throughout the whole parts of the fruit. Its peel extract have recently attracted interest as natural food preservatives due to its strong antioxidant potential as well as other bioactive properties. So, searching for the indispensable value of waste material, extraction of ellagitannins from pomegranate rind was standardized after comparing different extraction techniques. Ellagitannins belong to a large group of phytochemicals collectively known as polyphenolics or tannins. Extraction was made by low frequency bathsonication for 1 hour using ethanol provided punicalin yield of 2.10 %. High speed homogenization (2.08 %) and ultrasonication (1.71 %) proved as better extraction techniques among others in terms of punicalin yield after variation in time upto 30 min. It also serves the information that extraction efficiency of punicalin can be increased depending on the time of extraction. For purification of ellagitannins rich extract, different absorbent resins (DIAION HP-20, XAD-2 and XAD-16) were examined and finally DIAION HP-20 polyaromatic absorbent resin was selected for purification. The yield of punicalin in purified extract was 7.3 %. A significant purity improvement (38.40 %) was made by heating the purified extract with ethylacetate. After standardizing HPLC method for quantification of punicalinand ellagitannins content, characterization was made by HRMS/MS (ESI). The detailed HRMS study of purified extract confirmed the presence of two major compounds namely punicalin A, punicalin B (24.9 %) and one minor compound, hexose sugar of ellagic acid. The major two peak with m/z at 783.0658 and 783.0660 and their fragment ions at m/z of 765.0555 ([M+H] +-H O), 603.0032 ([M+H] Osugar), 556.9981 ([M+H] + -H2O-sugar-COOH), 513.0084 ([M+H]O-sugar COOH-COO)and 300.9976 ($C_{14}H_5O_8$) confirmed the structure of punicalin A andpunicalin B, respectively. Similarly characterization of the ethanolic extract byHRMS spectrum revealed that five compounds namely gallocatechin, dalbergionidin,ellagic acid, pentose sugar of ellagic acid and deoxyhexose sugar of ellagic acid were present. Difference in expected and measured m/z value was below 3 ppm for all the identified compounds. The purified ellagitannins were microencapsulated using maltodextrin as a carrier by spray drying technique. Purified ellagitannins were evaluated for in-silico prediction assay (Schrodinger Maestro v10.5software) which revealed that hexose sugar of ellagic acid and punicalin act as a better substrate for Mitochondrial Superoxide Dismutase (gliding score 7.780) and catalase enzyme (gliding score 9.741), respectively. For validation of the prediction assay, the formulated sample, ethanol extract and purified extract were evaluated for four nonenzymatic antioxidant assay namely DPPH, ABTS, FRAP and total phenolic contents. The IC_{50}values for DPPH assay of formulated sample provided better activity (4.99 µg mL^{-1}) than ethanolic extract (29.16 µg mL^{-1}) and purified extract (11.73 µg mL^{-1}). In ABTS assay, a best response of formulated sample (25.39 µg mL^{-1}) was observed whereas, in FRAP assay, formulated sample showed better antioxidant potency (1.51 µmole TBHQ g^{-1}) than ethanolic extract (0.58 µmole TBHQ g^{-1}) and purified extract (0.99 µmole TBHQ g^{-1}). The bioavailability study carried out using the standardized protocol revealed that bioavailability of punicalinin both the formulated sample and ethanolic extract was around 5%.+-H2+-H.

A series of novel nanosuperabsorbent composites (NSAPC) were prepared by *in situ* solution polymerization of a novel biopolymer of plant origin and clay mineral using a green chemistry technique. The structure of NSAPC was characterized by elemental analysis, FT-IR, solid state ^{13}C NMR spectroscopy, X-ray diffraction (XRD) and Scanning Electron Microscopy (SEM). The NSAPC exhibited significant swelling in various environments. Addition of NSAPC significantly improved the moisture characteristics of plant growth media (both with and without soil), indicating its potential for diverse applications in conservation agriculture. Study on the effect of NSAPC on sorption and leaching behaviour of metribuzin indicated significant increase in the sorption of metribuzin in NSAPC amended soil. Leaching losses of were drastically reduced up to 36.4% as compared to 58.5% in control. The findings indicate the possibility of utilization of test hydrogels in development of slow release formulations.

Guar gum (GG), a galactomannan polysaccharide of plant origin was employed to prepare GG-*g*-cl-polyacrylate superabsorbent (GG-SAP) and superporous hydrogels (GG-SPH; Fig. 2). Reaction parameters such

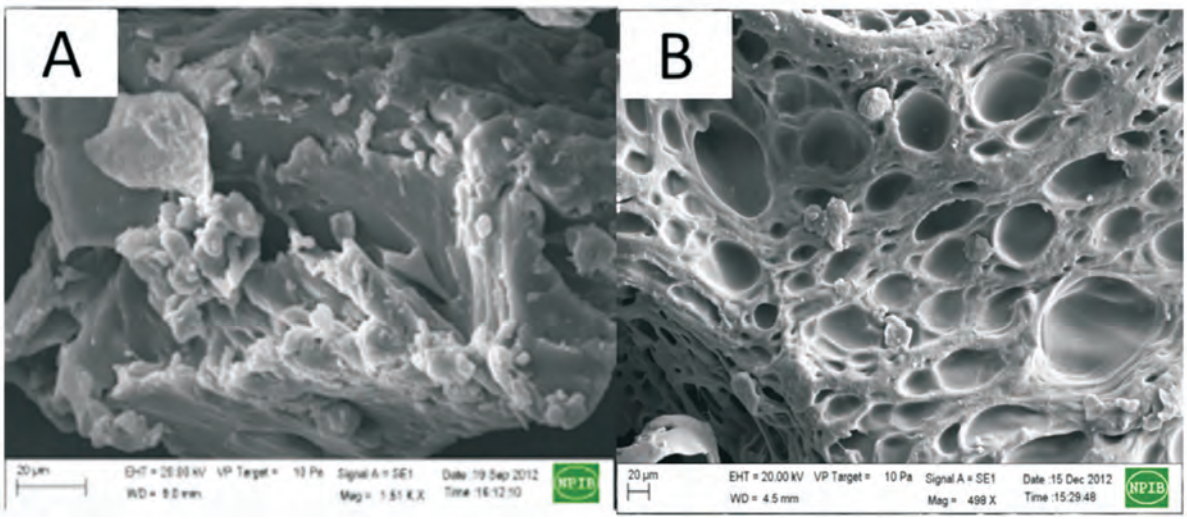


Fig. 2. SEM images of GG-*g*-cl-polyacrylate superabsorbent (GG-SAP) and superporous hydrogels (GG-SPH)

as guargum particle size, guargum-acrylate ratio, cross linker and initiator concentrations, volume of water per unit feed, foaming aid, porogen, foam stabilizer etc. were optimized. Microwave synthesis compared with the thermally initiated polymerization technique was found uneconomical under the process conditions employed. Structures of hydrogels were established by FT-IR and solid state ^{13}C NMR spectroscopy, X-ray diffraction (XRD) and scanning electron microscopy (SEM). Swelling behaviour of the representative GG-SAP and GG-SPH was evaluated w.r.t. ionic strengths of salt and fertilizer solutions, temperature, water quality and pH under laboratory conditions. GG-SAP exhibited significantly high swelling in acidic and alkaline environments as well as in the presence of salts and fertilizers. GG-SPH exhibited its pH-sensitive behaviour and faster swelling rate as compared to the corresponding GG-SAP. Addition of GG-SAP to plant growth media (sandy loam soil and soilless medium) significantly improved their moisture characteristics.

GG-*g*-cl-polyacrylate/clay superporous hydrogel composites (GG-SPHC) were prepared with improved water absorption and retention properties (Fig. 3). Guargum was also employed to develop core-shell nanogels (GG-CSH). Different core shell nanogels namely (GG-shell-NIPAAM-co-VP core); (GG-shell-PVA-core); (Poly NIPAAM-shell-GG-cl-PVA core) were synthesised by solution and emulsion polymerization techniques, that

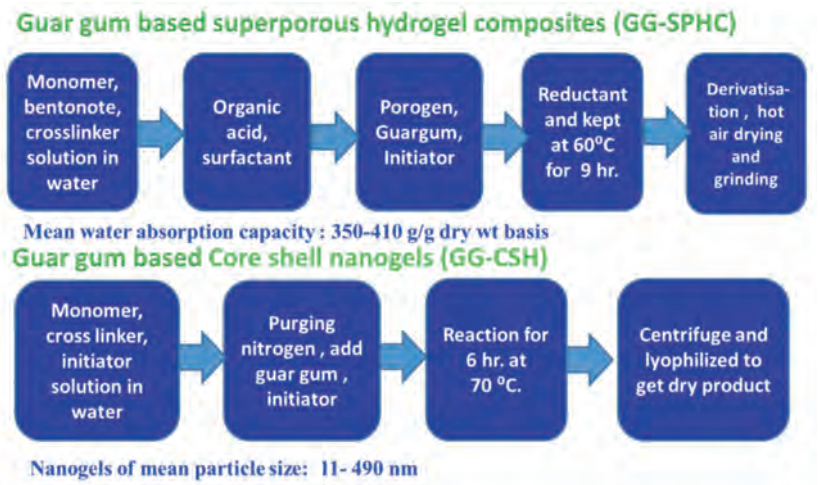


Fig. 3: Polymerisation technique of GG-SAPC and GG-CSH

exhibited mean particle size in the range of 11 nm to 490 nm. The prepared composites exhibited very fast swelling in distilled water (30 minutes), almost similar to the corresponding clay free hydrogel.

Nano-ranged amphiphilic block copolymers have attracted a great deal of attention in terms of their characteristic properties in solution, such as adsorption at interfaces and surfaces, self-assembly into micellar aggregates with a wide variety of geometries. These polymers are obtained by the polymerization of more than one type of monomers, typically one hydrophobic and one hydrophilic, so that the resulting molecule is composed of regions that have opposite affinities for an aqueous solvent. These materials, when intended for use in drug delivery, are generally composed of biocompatible, biodegradable constituents.

Amphiphilic polymers were synthesized using poly (ethylene glycols) (PEG) of different molecular weights, viz. 300, 600, 1000; as hydrophilic block and aliphatic diacids namely glutaric acid, adipic acid, pimelic acid and suberic acid as hydrophobic block in presence of catalyst Conc. H_2SO_4 (Fig. 4). Synthesized polyesters were characterized by using ^1H NMR, ^{13}C NMR and IR spectroscopy. Micellar sizes of the polymers were determined using Dynamic Light Scattering (DLS) which ranged from 127.5-354 nm. Transmission Electron Microscope (TEM) results confirm the findings of DLS. Critical Micelle Concentrations (CMC) of the synthesized polymers was determined using electrical conductivity meter which ranged from 112 to 155 mg L^{-1}.

Other aliphatic acids & aromatic diesters viz. azelaic acid, sebacic acid, dimethyl isophthalate acid and dimethyl terephthalate as hydrophobic block were reacted with PEG (Molecular weights, 1000, 2000 & 4000) to develop amphiphilic block copolymers. Micellar sizes of the polymers were determined using Dynamic Light Scattering (DLS)which ranged from 51.6-174 nm for aliphatic esters and 135.5-371 nm for aromatic esters. Transmission Electron Microscope (TEM) results confirm the findings of DLS. CMC were found to be ranged from 95 to 130 mg L^{-1} for aliphatic esters and 420-1500 mg L^{-1}.

Dimethyl 5-hydroxyisophthalate, as hydrophobic block, was used to synthesise amphiphilic block copolymers with PEG of molecular weights *viz.* 600, 1000, 1500 and 2000 in presence of conc. H_2SO_4 as catalyst and further alkylating the resulting polymers by attaching heptyl and tetradecyl chains to phenolic hydroxyl group. These polymers, when dissolved in water, aggregate to form micelles, giving size ranging from 26.50 to 85.10 nm as determined by DLS instrument. The molecular weight as calculated from the DLS were found to be in the range 8.8×10^3 to 8.5×10^4KDa (Kilo Daltons). CMC of the synthesized polymers was ranged between 115 to 148 mg L^{-1}(milligrams per litre).

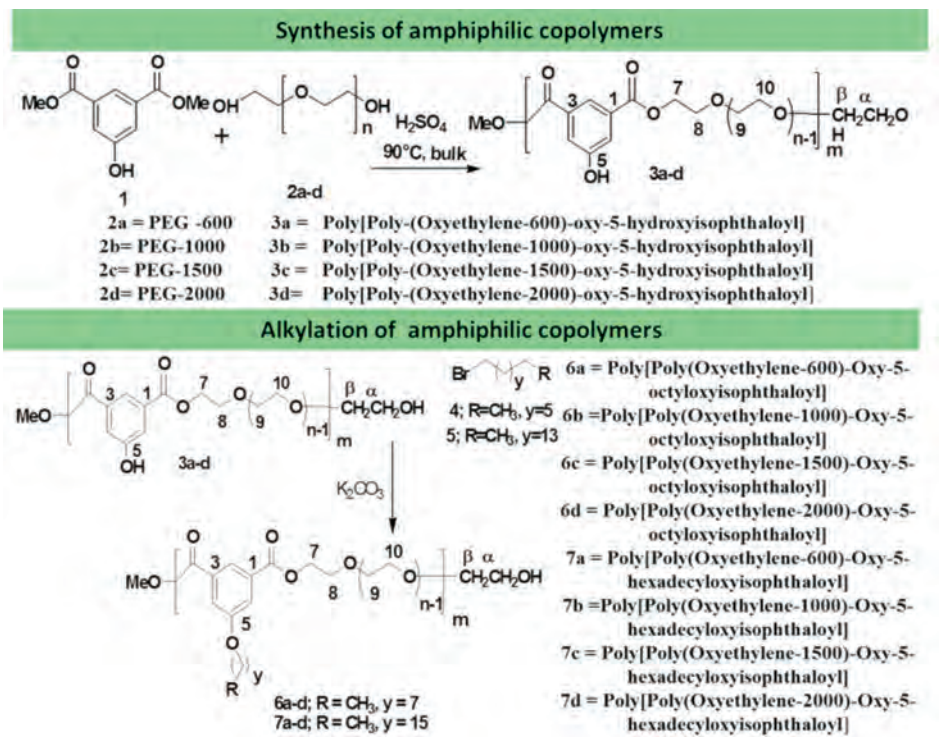


Fig. 4: Scheme of synthesis of amphiphilic polymer based on dimethyl hydroxyisophthalate as hydrophobic block and further alkylation by attaching heptyl and tetradecyl chains to phenolic hydroxyl group.

Agrochemical Formulation

Controlled release (CR) formulations of imidacloprid were prepared using amphiphilic copolymer using encapsulation technique. Encapsulation efficiency, loading capacity and stability after accelerated storage test of the developed formulations were checked. The kinetics of imidacloprid, release in water from the different formulations is reported. Release from the commercial formulation was faster than the CR formulations. The diffusion exponent (*n* value) of imidacloprid, in water ranged from 0.22 to 0.37 in the tested formulations. While the time taken for release of 50% of imidacloprid ranged from 2.32 to 9.31 days for the CR formulations. Seed coating was performed with these CR formulations of imidacloprid along with commercial formulation and control. The percent imidacloprid recovery ranged from 85.4-94.3% from different CR formulation seed coats. Better imidacloprid retention was observed on soybean seed coats treated with CR formulations. Different seed quality parameters viz., seed germination, seed vigour, seed moisture and seed storability, etc. were evaluated during the six month experimentation. The coated seeds with different nano-formulations had better seed quality over control and commercial formulated coated seeds during the experimentation. The bioefficacy of the prepared CR formulations was evaluated against major pests of soybean namely stem fly, Melanagromyza sojae Zehntmer and white fly, *Bemisia tabaci* Gennadius along with a commercial formulation at the experimental farm of Indian Agricultural Research Institute (IARI), New Delhi during kharif 2009 and 2010. Most of the CR formulations of imidacloprid gave significantly better control of the pests compare to its commercial formulations, however the CR formulations, Poly [poly (oxyethylene-1000)-oxy suberoyl] amphiphilic polymer based formulation performed better over others for controlling of both stem fly incidence and Yellow Mosaic Virus (YMV) infestation transmitted by white fly.

Controlled release (CR) formulations of thiamethoxam (3-(2-chloro-1,3-thiazol-5-ylmethyl)-5-methyl-1,3,5-oxadiazinan-4- ylidene (nitro)amine) were prepared using different amphiphilic copolymer using encapsulation technique (Fig. 5). Formulations were characterized by FT-IR spectroscopy, DLS, SEM and TEM. The kinetic study of thiamethoxam release from developed formulation showed slower release in water and soil as compare to commercial formulation. The diffusion exponent (*n* value) of thiamethoxam, in water ranged from 0.346 to 0.764 and in soil ranged from 0.532 to 0.881 in the test formulations. Thetimes taken for release of 50% of thiamethoxam form the CR formulations were 2.04 to5.46 days and 3.56 to 6.07 days in water and soil, respectively. The developed CR formulations of thiamethoxam were used to coat soybean seeds to evaluate the physiological quality of thiamethoxam-treated soybean seeds stored for three different periods. Better thiamethoxam retention was observed on soybean seed coats treated with CR formulations. More specifically formulation based on Poly [poly (oxyethylene-4000)-oxy sebacoyl] showed best retention capacity. The bioefficacy of the prepared CR formulations as seed coat was evaluated against major pests of soybean (JS335) namely white fly, *Bemisia tabaci* Gennadius and stem fly, *Melanagromyza sojae* Zehntmer along with a

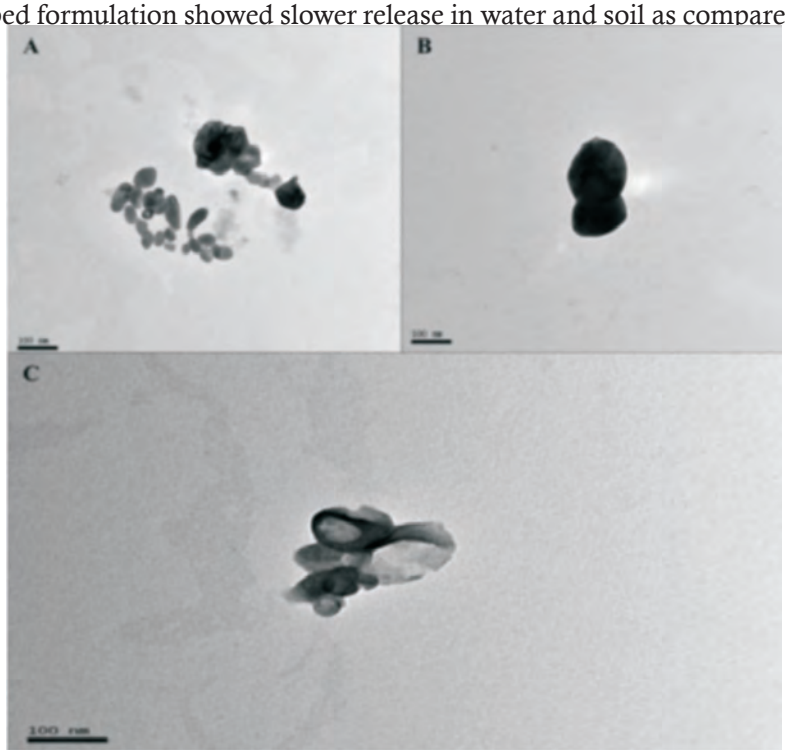


Fig. 5: TEM images of micelle based on amphiphilic polymer (A) and nanocapsules of thiamethoxam (B &C)

commercial formulation at the experimental farm of Indian Agricultural Research Institute (IARI), New Delhi during kharif 2011. Most of the CR formulations of thiamethoxam provided superior better control of white fly which transmit Yellow Mosaic Virus (YMV) compared to commercial formulation and control. Most of the developed CR formulations recorded higher yield over commercial formulation and control. The seed coating formulation based on amphiphilic polymer Poly [poly (oxyethylene-4000)-oxy sebacoyl] with recommended dose of thiamethoxam i.e. @ 3.0g a.i. kg^{-1} seed showed best performance in terms of yield (2134.41 kg ha-1) compare to commercial formulation (1752.65 kg ha-1).Nodulation pattern of soybean was not affected due to treatment of CR formulations and commercial formulation of thiamethoxam. Also the residues of thiamethoxam in seed andsoil at harvest were below the detection limit (0.25 µg mL-1) for both CR and commercial formulations. The study revealed that the CR products of the thiamethoxam may be usedfor seed quality enhancement and efficient pest management (Karmakar *et al.,* 2012)

CR formulations of carbendazim (Methyl *1H*-benzimidazol-2-ylcarbamate), a systemic fungicide, were prepared using amphiphilic polymers. The kinetics of carbendazim from developed controlled release (CR) formulations were studied and compared with that of the commercially available 50% Wettable Powder (WP) and the bio efficacy evaluation of developed formulations was done against the plant pathogenic fungi *Rhizoctonia solani* by the poison food technique. The developed formulations showed fungicidal activity at par with the commercially available formulation of carbendazim.

CR formulations of azoxystrobin using amphiphilic polymers were prepared by encapsulation technique. Formulations were characterized by FT-IR, DLS and TEM. The kinetics of azoxystrobin release in water from the developed formulations confirmed slower release azoxystrobin from amphiphilic matrix as compare to commercial formulation. The diffusion exponent (*n* value) of azoxystrobin, in water ranged from 0.21 to 0.56 in the tested formulations. The time taken for release of 50% of azoxystrobin ranged from 2.05 to 7.63 days for the CR formulations. The bioefficacy of the prepared CR formulations was evaluated *in vitro* and in pots against major pathogens of rice namely *Rhizoctonia solani* and *Fusarium fujikuroi*. Most of the CR formulations of azoxystrobin provided superior control of both fungi compared to commercial formulation. The percent inhibition ranged from 55.4-86.0% and 55.8-87.5% against *Rhizoctonia solani* and *Fusarium fujikuroi* respectively from different CR formulation of azoxystrobin obtained in *in vitro* study. In the pot experiments all the treatments resulted in a significant reduction of foolish seedlings infestation as compared to untreated control. The seedling treated formulation based on amphiphilic polymer Poly [poly (oxyethylene-4000)-oxy glycoyl] and Poly [poly (oxyethylene-1500)-oxyglycoyl] with recommended dose of azoxystrobin *i.e.* @ 1000 ppm against *Fusarium fujikuroi* showed best performance in terms of number of survived seedlings compared to commercial formulation. The study revealed that the CR products of the azoxystrobin may be used for efficient pest management of rice.

The loading of thiram in nano spheres was achieved by encapsulation in amphiphilic polymers. The release of thiram in water was checked together with the commercial formulation (75 WS) of the thiram using HPLC. The release of a.i from the encapsulated product was analyzed at different time intervals viz. 0, 1, 3, 7, 14, 21, 28, 35, 42 and 49 days. Data analysis for the release of the the a.i. from the encapsulated products at different time intervals showed that the release for all the developed formulations was slow where maximum release was observed at 15th day for PEG-600, 21st day for PEG-1000, 28th day for PEG -1500 and 35th day for PEG-2000 based formulation as compared to the commercial formulation in which the maximum release was upto 7rd day. The half- life calculation also indicated that the commercial formulation had minimum value as compared to the developed formulations. The period of optimum availability (POA) also confirmed the above findings (POA for commercial formulation was 7.79 while that of PEG – 2000 based formulation 25.15 days).

Nanoformulations of pyridalyl, was prepared for better efficacy and safe usage. Compatibility of pyridalyl with sodium alginate was ascertained and a new formulation was obtained by optimizing various parameters required for its preparation. Size of the optimized formulation was around 138 nm and size of pyridalyl was less than 100 nm. Spherical shape and nanosize of the capsules was confirmed by TEM. Developed nanoformulations

were characterized by using FTIR spectroscopy. Insecticidal activity of pyridalyl nano suspension was evaluated in comparison to its technical material and commercial formulation against larvae of *Helicoverpa armigera in vitro* as well as in field. The effectiveness of nanosized material increased remarkably, as evidenced by recording LC_{50} value of nanopyridalyl as 40 µg mL^{-1} in comparison to LC_{50} values of technical material (90 µg mL^{-1}) and commercial product (250 µg mL^{-1}). Bioassay results by leaf dip method showed that nanoformulation is 2.26 and 6.25 times more effective against *H. armigera* as stomach poison than the technical product and commercial formulation, respectively. By topical method, the respective LC_{50} values obtained were 80, 150 and 250 µg mL^{-1} for nanoformulation, technical material and commercial formulation. Independent diet incorporation study validated this trend.

Cucumber (*Cucumis sativus*) is an important high value vegetable grown under polyhouse conditions. Nematode and magnesium management are two most important aspects to realize higher productivity and superior quality in cucumber. Root-knot nematode, (*M. incognita*) causes severe damage to the crop leading to dramatic yield losses. Marigolds (*Tagetes* sp.) established to possess toxic principles effective against nematode pests offer ecofriendly alternative to hazardous chemical nematicides. Besides nematodes, cucumber crop is highly sensitive to availability of magnesium in soil. Integrated test formulations were prepared by incorporating hexane fraction of root extract of *Tagetes patula* and magnesium sulphate in Pusa Hydrogel, a superabsorbent polymer used as carrier. Release study of extract and magnesium from test products in soil and water showed that release followed first order kinetics and test products containing extracts behaved as slow release formulations with $T_{1/2}$ in water and soil 22.97 and 36.87 days respectively. Bioassay study showed that magnesium sulphate enhanced the nematicidal activity of extracts. Performance of test formulations vis-à-vis carbofuran and conventionally applied magnesium was evaluated in cucumber under polyhouse conditions. Leaf area, number of flowers per plant, percent magnesium in leaf tissue, leaf chlorophyll content and yield were determined. Integrated test formulations exhibited significant increase in yield over control. Similar trend was reflected in magnesium and chlorophyll content. The correlation between magnesium, yield, and chlorophyll was 83.2% and 85.3% respectively. In vivo bioefficacy evaluation of test products showed nematicidal activity (37.4%) comparable to carbofuran (47.83%). The findings indicate potential of hydrogel based integrated soil borne pest and nutrient management approach which needs to be further established under practical use situations.

Fungal diseases pose a major challenge in the productivity of fruits and vegetables. Root rot disease, caused by *Pythium aphanidermatum* is of major concern. In view of the environmental concerns related to crop protection chemicals, biocontrol approaches for pest management are of current interest in crop protection programmes. However, the potential of bioagents remains underutilized due to shelf and field life constraints such as high temperature and limited moisture in the soil. Development of suitable formulations to overcome these constraints is thus, imperative. The main objective of present thesis was to develop biopolymeric hydrogels based bioformulations of *Trichoderma harzianum* and *Pseudomonas fluorescens* and to evaluate their shelf lives and bioefficacy against *Pythium aphanidermatum* under laboratory conditions. Guargum based superabsorbent (GG-SAP) and guargum based superporous hydrogel (GG-SPH) with optimized matrix properties were used to prepare 12 novel bioformulations of *Trichoderma harzianum* and *Pseudomonas fluorescens* (Fig. 6). All the test compositions contained > 5×10^8 colony forming units per gram of carrier at three different storage temperatures, 5^0, 25^0 and 45^0C for a study period of 180 days. Bioefficacy evaluation (*in vitro*) of test compositions stored at different temperatures, against *Pythium aphanidermatum* showed that the test compositions stored at 5^0 and 25^0C were able to inhibit >50% *Pythium* population (*in vitro*) up to 180 days storage period. Compositions stored at 45^0C exhibited >50% (or equal) inhibition up to 90 days storage period. Finding of the present work point towards a promising hydrogel based bioformulation approach for integrated water and disease management in horticultural crops, which will be further, established under practical use conditions.

Guar gum based novel GG-*g*-polyacrylate/claysuperporous hydrogel composites (GG-SPHC) and core-shell nanogels (GG-CSH) were developed as carrier materials for agrochemical formulations. The optimized

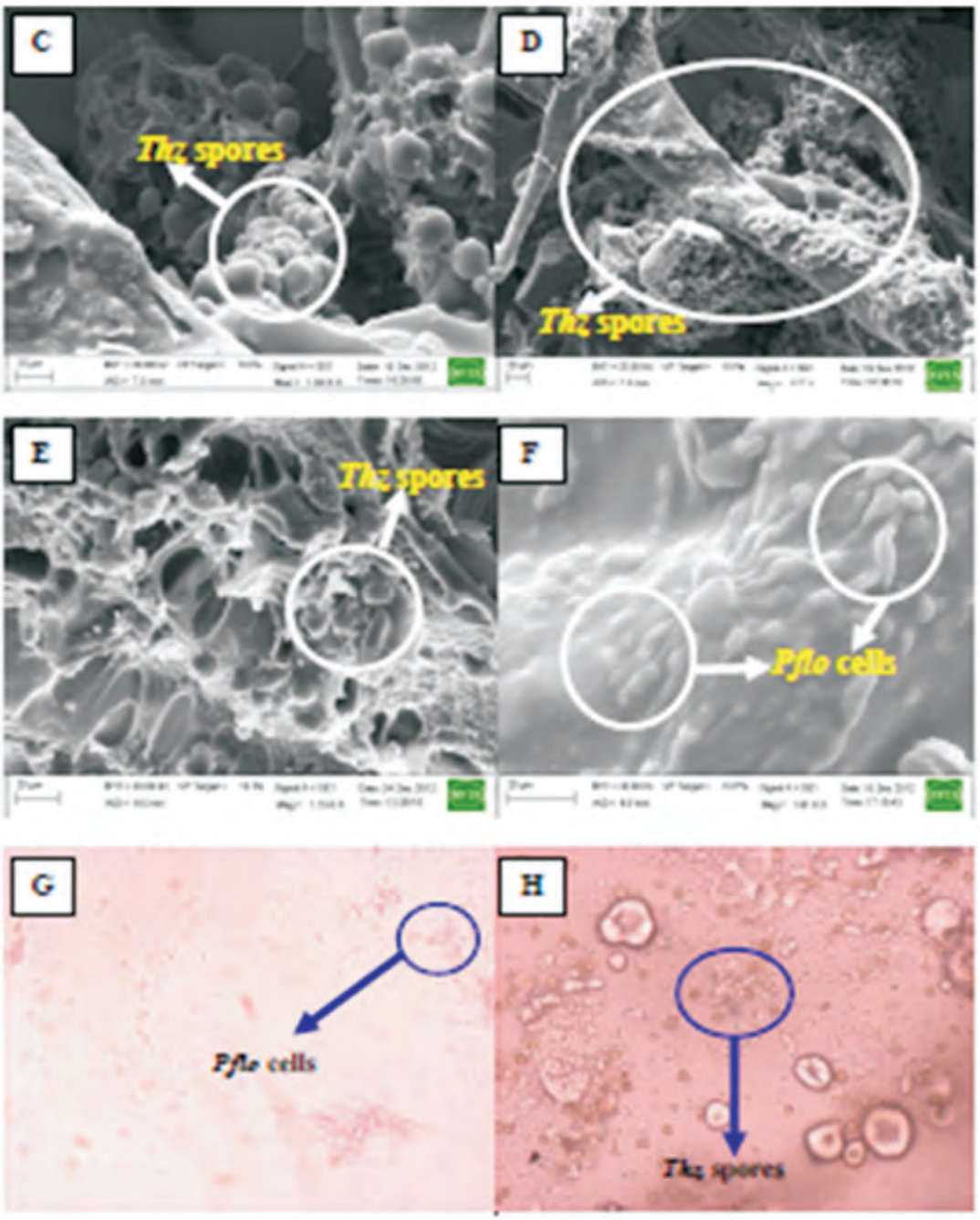


Fig. 6: SEM and optical microscope images of bioformulations of *Trichoderma harzianum* and *Pseudomonas fluorescens* based on guargum hydrogels

polymer composites and nanogels were used to prepare formulations of imazathapyr under ambient conditions. The release of *a.i.* from the all the test compositions in water followed Fickian mechanism and slow release behaviour as compared to commercial formulation. Field appraisal on bioefficacy of prepared formulations *vis-a-vis* commercial formulations showed very high weed control index of test formulation (96.7- 99.1% on 90 DAS) comparable to that of commercial formulations (98-99.4%). Hydrogel Composite based formulations of imazethapyr applied at pre-emergence stage, exhibited significantly higher weed control as compared to other formulation treatments.

Safety Evaluation of Agrochemicals

The use of pesticides for protecting crops from pest infestation may lead to the accumulation of their residues in food and feed crops. Increased public awareness on the use of pesticides and the concerns about their toxic residues, and effects on non-target organisms demand in-depth studies on the detection and quantification of pesticide residues on different crops, soils, water and other substrates. Besides establishing safe waiting periods, such studies yield useful information on the translocation, degradation and persistence of pesticides and their metabolites in different environmental segments.

Degradation and mobility behavior of fipronil in soil was studied under laboratory conditions. Metabolites formed in different treatments were identified and quantified. In general, the insecticide persisted for 15-90 days. The half-life for total residues (fipronil + sulfone + sulfide + desulfinyl) varied from 3.2-79.2 days under different treatments. The dissipation of the compound in soil was found to vary with the soil moisture regimes, fortification levels and light conditions. Trend in dissipation was submerged (T1/2 3.2-10.9 days)>field capacity (T1/2 27.6-36.7 days)> dry (T1/2 51.0-79.2days). The dissipation was faster at 1.0 µg g^{-1} level as compared to 10 µg g^{-1} level dissipation. Residues dissipated faster when exposed to ultra-violet light followed by sunlight and no light condition.. Under anaerobic condition, fipronil sulfide, a reduction product of fipronil, was detected. In UV and sunlight exposure, fipronil desulfinyl, major photo product of fipronil was detected. Leaching studies revealed low downward movement of compound. Even after leaching with water equivalent to 400 mm rainfall, more then 90% residues remained within top 10 cm layer in both packed and intact columns. Two toxic metabolites of fipronil viz desulfinyl and sulfone was found to be less mobile than fipronil. Reduced leaching was observed in case of formulation treatments as compared to analytical grade material.

The effect of nanoformulation on the persistence behaviour of pesticide as compared to their commercial formulation was studied through residue analysis of nanocapsule suspension of pyridalyl and its commercial formulation on vegetables. Methods were developed for the estimating the pyridalyl in tomato and okra. Residue study of commercial and nanoformulation of pyridalyl was done on two crops (okra and tomato).Ultra High Performance Liquid Chromatography (UHPLC) method has been standardized to analyze pyridalyl in different matrices with high resolution and sensitivity. Methods for estimating the insecticide in okra (*Abelmoschus esculantus*) and tomato (*Solanum lycopersicum*) were developed. Limit of detection (LOD) of pyridalyl in UPLC was 0.01 µg mL^{-1} and limit of quantification (LOQ) of the methods were 0.02 µg g^{-1} for okra and 0.04 µg g^{-1} for tomato.In case of okra, traditional extraction and cleanup method was used with recoveries ranging from 79-87%. Half-life and residue levels of pyridalyl were determined in okra over a period of 15 days. Bioefficacy of the laboratory synthesised nanocapsule suspension of pyridalyl along with its commercial formulation (10 EC) was also evaluated under natural incidence of the fruit borers of okra (boll worms, *Erias vitella and Helicoverpa armigera*) at the experimental farm of Indian Agricultural Research Institute (IARI), New Delhi during *kharif* 2012. Okra (Pusa A-4 variety) was grown and at the time of fruiting, the vegetable was sprayed with commercial and nanoformulation of pyridalyl at recommended (75 g a.i. ha^{-1}) and double the recommended dose (150 g a.i. ha^{-1}). Dissipation pattern of pyridalyl in okra was determined in the field as well as in pot for commercial and laboratory made nanoformulation of pyridalyl. Effect of the treatments was also recorded as percentage of fruit damage. Nanopyridalyl gave significantly better control of fruit borers, in comparison to commercial formulation and control. However, residues of pyridalyl were not significantly different for both the formulations. Half-life of pyridalyl residue was slightly higher in net house than in field. Similar study was undertaken to evaluate the rate of dissipation of the residues of pyridalyl in tomato (Rohini variety) grown in two different cultivation systems: open field (conventional cultivation) and net house (pot experiment) during *Rabi* season of 2012-13. Samples were extracted and cleaned up using state of the art QuEChERS methodology and analyzed by UHPLC with recoveries ranging from 90-116%. In Field study initial residues on tomato were found to be lower than that found in okra, while in pot study nearly similar residues were obtained. Safety evaluation using acceptable daily intake (ADI) showed that maximum permissible intake (MPI) (1.4 mg/person/day) for human is more than the theoretical maximum residue contribution (TMRC) calculated using maximum residues

recorded on the vegetables. Though multi locational field trials were not done, based on this study a tentative MRL can be proposed as 0.3 µg g^{-1} and a waiting period of one day can be suggested for nanoformulation of this lipophilic insecticide. It is not different from that of available commercial formulation. The study confirmed that nanoformulation of pyridalyl gives better control of fruit borers of tomato

Persistence behaviour of clothianidin was studied two different soil types, namely Delhi and Manipur under different moisture regimes, with and without amendment. The half-life values ranged from ($t_{1/2}$ 33.4-367.0 days). A highly persistent nature of the clothianidin was observed. The results revealed that clothianidin was moderately persistent in anaerobic soils ($t_{1/2}$ 143.3-103.7 days) and highly persistent in aerobic soils ($t_{1/2}$ 273.63-301.0 days). The compost amendement greatly enhanced the rate of dissipation of clothianidin in Delhi and Manipur soils ($t_{1/2}$ 111.4-158.4 and 150.2-188.4 days;167.2 and 231.5 days in unamended soils). 3-methyl-1-nitroguanidine (MNG) was is the major metabolite of clothianidin, was prepared in the laboratory, to serve as analytical standard in the soil persistence study. Exposure of clothianidin to different light conditions as thin film on petri plates indicated that clothianidin was more prone to degradation under UV light ($t_{1/2}$ 7.0 days) than under sunlight ($t_{1/2}$ 12.0 days) and dark light ($t_{1/2}$ 33.4 days). Adsorption percent of clothianidin in normal Delhi soil was 46.26-49.37%, as compared to less adsorption in normal Manipur soil with adsorption percentage 52.53-26%. Adsorption studies conducted in Delhi and Manipur soil amended with 0.2% FYM, indicated that amendment attributed to more adsorption than the corresponding normal soils without amendment (54.75-55.28% Delhi soil; 59.38-60.55% Manipur soil). Desorption was difficult in Manipur soil (desorption 22.50-55.19%) and soils with amendment (desorption 26.02-33.35%) having high organic matter content as compared to normal Delhi (desorption 52.36-71.12%) and FYM amended soil (32.55-42.74% desorption). Leaching carried out with increasing volumes of water indicated that clothianidin is more mobile in sandy loam soil of Delhi as compared to clayey soil of Manipur. The FYM amendment soils certainly decreases the leaching potential of clothianidin to lower depth of soil (Fig. 7).

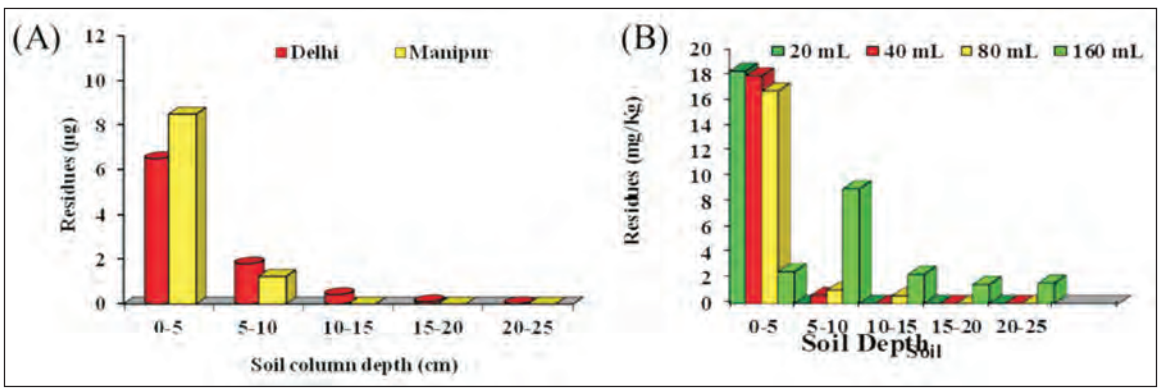


Fig. 7: Persistence of clothianidin in soil affected by different moisture regime (A) continuous flow conditions and (B) discontinuous flow conditions

Kresoxim-methyl is a broad spectrum foliar fungicide of the strobilurin group. A simple and reliable analytical method using HPLC was developed for analysis of kresoxim methyl and its acid from soil and water. Dichloromethane (DCM) was found to be the best solvent for extracting the residues from water. It was found that acidification of aqueous phase prior to DCM extraction gave optimum recovery of both kresoxim methyl (89.3-92.3%) and acid metabolite (79.9-84.8%) at 0.5 and 0.01 µg ml^{-1} fortification levels. The recovery data obtained following different methods for soil samples revealed that dipping and shaking with 20% aqueous acetone followed by partitioning of the acidified extract with dichloromethane and column clean up over Florisil is best for extraction and clean up with recovery percentage of 86.4-93.0% for kresoxim methyl and 82.7-84.4% for acid metabolite at 1.0 and 0.01 µg g^{-1} fortification levels. The developed method can be used for quantifying the residues of kresoxim methyl and its metabolite in field samples of water and soil. The

adsorption studies revealed that different soils had moderate to high adsorption capacities for kresoxim methyl and acid metabolite. The trend in adsorption in different soil was Almora > Kerala > Kolkata > Nagpur > Delhi. Adsorption was found to be higher in sludge amended soil as compared to normal soil. Desorption of kresoxim methyl was found to be highest in Delhi soil followed by Nagpur, Kolkata, Kerala and Almora soil. Kresoxim methyl acid metabolite showed much higher desorption than parent molecule. Low value of hysteresis coefficients suggests high hysteresis effect indicating easy and strong adsorption of kresoxim-methyl in soils and slow desorption. The results suggest that the organic carbon play a major role in sorption of kresoxim-methyl in soil.In leaching study, kresoxim methyl was found to undergo very fast dissipation in column soil and form acid metabolite. Acid metabolite has much more leaching potential as compared to parent molecule. Leaching of both parent and metabolite increases with the amount of rainfall. Amending the soil with sludge reduces the leaching potential of both parent and the acid metabolite. Because of the easy transformation of parent molecule into acid metabolite and high leaching potential of acid metabolite, it must be used cautiously in the high rainfall areas. Dissipation behaviour of kresoxim-methyl in soil and water has been investigated. Among the two soil types, kresoxim-methyl and total residues dissipated at a faster rate in Inceptisol than in Ultisol. Faster dissipation was observed in submerged conditions followed by field capacity and air dry conditions. Amending the soil with sludge fastens the rate of dissipation. Exposure to light and increase in the atmospheric CO_2 level also has a direct relation with the kresoxim dissipation. In water, rate of dissipation of kresoxim-methyl increased with the increasing pH of the medium. Exposure to light, increase in temperature and increase in the atmospheric CO_2 level increases the dissipation rate of kresoxim-methyl. The total study suggests that kresoxim-methyl alone has low to moderate persistence in soil and water. Under different conditions, it readily undergoes hydrolysis and transform into acid metabolite. Because of the slow dissipation of acid metabolite, the total residues (kresoxim methyl+acid metabolite) persist for a longer period in soil and water. Statistical analysis using SAS 9.3 software and Duncan's Multiple Range Test revealed the significant effect of soil type, moisture regime, organic matter, microbial population, light exposure, pH, temperature and atmospheric CO_2 level on the dissipation of kresoxim-methyl from soil and water (at 95% confidence level $p<0.0001$). It can be recommended from above research that kresoxim-methyl in high rainfall area can be used in proper way by using suitable management practices.

Persistence behaviour of tricyclazole, a systemic fungicide, belonging to triazolobenzothiazole class of compounds was studied in two different soil types form Delhi and Karnataka under different moisture regimes, pH, CO_2 level and using amendments. The study indicated that tricyclazole dissipated faster in submerged soil ($t_{1/2}$ 188.13–286.15 days) followed by field capacity ($t_{1/2}$ 266.37–334.44 days) and dry soil ($t_{1/2}$ 301–376.25 days) in both soils. The results revealed that tricyclazole was moderately persistent in anaerobic soils and highly persistent in aerobic soils. 2-hydrazinyl-4-methyl-1,3-benzothiazole, the major metabolite of tricyclazole,was not detected in the soil persistence studies. Tricyclazole was found to be stable in water over a pH range of 4 to 7 and dissipated faster in pH 9.2 with a half-life of 261.74, 281.31 and 231.54 days respectively. Under elevated CO_2 (550 and 750 $\mu molmol^{-1}$) conditions tricyclazole dissipated faster as compare to ambient CO_2 (395 $\mu molmol^{-1}$). It was found that tricyclazole is strongly adsorbed in inceptisol of Delhi having relatively higher organic matter as compared to ultisol of Karnataka. The downward mobility studies indicate that tricyclazole is highly mobile in ultisol as compared to inceptisol where it is relatively less mobile (Fig. 8). Experiments revealed that the potential of ground water contamination can be minimized by blue-green algae (BGA) amendment in soil columns where most of the pesticide is retained in the top layer of soil column. Sorption study indicated that adsorption of tricyclazole on Delhi soil was higher (43.09%) than Karnataka soil (18.27%) and it increased with 0.05% BGA amendment in both the soils (55.14% and 32.52% respectively). Desorption of tricyclazole in Delhi soil was less (7.16–20.11%) as compare to Karnataka soil (8.59–32.18%) and amendment with BGA decreased the pesticide desorption in both soil types . Desorption was slower than adsorption indicating a hysteresis effect. Leaching study was carried out in packed soil column. The downward mobility of tricyclazole was found to be decreased from 0–5 cm to 20–25 cm due to high adsorption by soil particles. In case of Karnataka soil, the mobility was found to be more than Delhi soil as around 0.08 μgmL^{-1}residueof tricyclazole was detected in the leachate, indicating high leaching potential in sandy loam Karnataka soil under continuous flow pattern.

Addition of BGA as amendment greatly influences the mobility behavior of the tricyclazole leading to retention of 85.30% and 65.40% of applied fungicide within top 0–5 cm layer of Delhi and Karnataka soil respectively. The leaching of tricyclazole under field condition indicated that it remained in upper soil profile (0–15 cm) during the initial 7 days after spraying and moved to lower soil profile slowly with time. Downward mobility of tricyclazole significantly increased with the time after application, dose and depth of application. Tricyclazole was found to be dissipated faster from the rice foliage with a half-life of 2.5–3.1 days and residue in grains at harvest was below detectable limit (<0.05 mg kg^{-1}) (Kumar and Mukherjee, 2013; Kumar *et al.,* 2012; Kumar *et al.,* 2015; Kumar *et al.,* 2017).

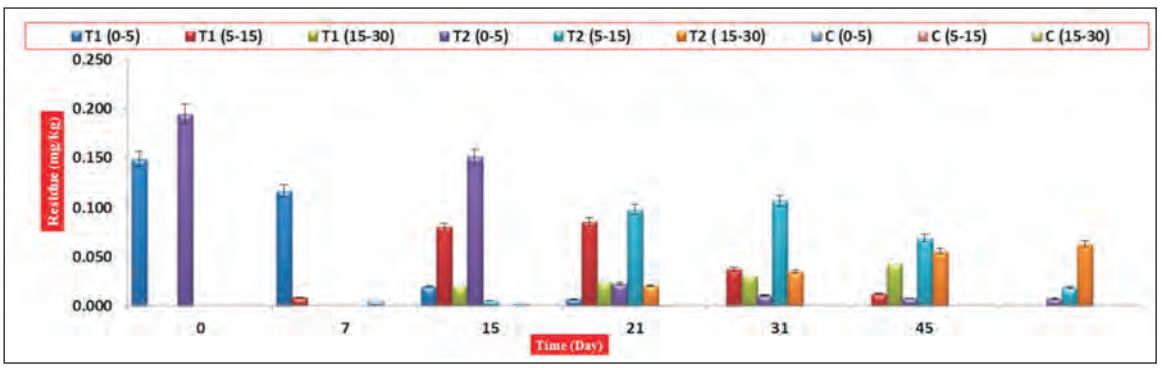


Fig. 8: Downward mobility of tricyclazole in paddy field

A study was undertaken to monitor the selected PAHs, PCBs and surfactants in influent and effluent water of the two sewage water treatment plants present in IARI farm. The two sewage treatment systems are Horizontal sub surface flow (HSSF) and vertical sub surface flow (VSSF). Methods have been developed for identification and quantification of selected contaminants PAHs -Naphthalene, Phenanthrene and Benzo(a) anthracene; PCBs-PCB 52 and PCB 44; surfactants - SDS and SDBS using GC-MS for PAHs and PCBs and LC-MS-MS for surfactant determination. The methods were found to be simple and selective with average recovery of 72.8, 75 and 86% for PAHs, PCBs and surfactants, respectively. The LOQ for PAH and PCB was 0.0025 µg/mL while for surfactant it was 0.01 µg/mL. Recovery experiments were conducted using 2 different solvents ethyl acetate and DCM:Hexane(1:1). The analysis of influent sewage water showed the presence of all the selected compounds except PCB 44 with concentrations ranging from 18.42-32.18, 4.0-7.70, 2.37-5.46 µg/L of naphthalene, phenanthrin, pyrene and benzo(a)anthracene, respectively; 3.91-8.73 µg/L of PCB 52 and 151.9-1026.5 µg/mL of SDS and 889.1-3163.5 µg/mL of SDBS. Both the sewage water treatment systems were found to remove theses contaminants from sewage water. In VSSF, the removal varied from 16-99% whereas in HSSF system, the removal of contaminants varied from 20-95%. In both the system, higher removal was recorded for naphthalene than other PAHs. A among the surfactant, SDBS showed higher removal than SDS. The removal of PCB 52 was lower than naphthalene but comparable to other PAHs. The removal of contaminants was almost similar but HSSF system was found to be slightly better for removal of PAHs and PCBs removal.Both the treatment systems were found to efficient in removing the selected contaminants (four PAHs, one PCBs and two surfactants).

Antibiotics are a group of compounds which are widely used in agriculture for the treatment of diseases and also as growth promoters in animals. Tetracyclines (TC) and oxytetracyclines (OTC) are mainly applied in livestock farming. Only small amount of TCs and OTCs are absorbed during metabolism, and the majority are excreted via feces and urine as unchanged form. Due to the application of animal manure and sewage sludge in agriculture as fertilizers, these antibiotics have the potential to reach soil and aquatic environment. Their residues have been frequently detected in soils, sediments, surface water, ground water and waste water. Exposure to low level antibiotics and their transformation products in the environment could be toxic and cause

dissemination of antibiotic resistant genes among microorganisms. Simultaneous study is not yet reported of tetracycline and oxytetracycline dynamics in different soils and manures under Indian conditions. Keeping this aspect in view, persistence and mobility of tetracycline and oxytetracycline in soils and manures was carried out under laboratory conditions. Persistence and leaching studies have been conducted in order to understand the dynamics of tetracycline and oxytetracycline in the soil and manures under laboratory conditions as influenced by different factors mainly water content, physical and chemical properties of soil and manures. Standardization of analytical methods was done for both tetracycline and oxytetracycline from soil, water and plant samples. Dip and shake method, partitioning using dichloromethane followed by column cleanup was found most suitable and HPLC parameters were standardized separately for each compound. Limit of Detection (LOD) for both tetracycline and oxytetracycline were found to be 0.01µgmL^{-1} (1 ng with 10µL injection). Limit of Quantification (LOQ) for tetracycline and oxytetracycline in water and soil samples were found to be 0.05µg mL^{-1}. Comparative study of persistence and mobility of tetracycline and oxytetracycline was studied in Delhi soil, Meghalaya soil, FYM and poultry at 20 µg g^{-1} under three different moisture regimes. In persistence study residues persisted beyond 90 days and undetected after 120 days in all the treatments. The half-life values ranged from (t1/2, 60.2, 56.79, 40.67 and 38.59 days) under various moisture conditions for tetracycline and t1/2, 42.39, 37.63, 32.72 , 31.03, in Delhi soil, Meghalaya soil, FYM and poultry manure, respectively for oxytetracycline. The results showed that tetracycline and oxytetracycline have highest persistence in Delhi soil in aerobic conditions followed by Meghalaya soil and least persistent in FYM and poultry manure under anaerobic conditions. Leaching studies showed that the mobility of tetracycline and oxytetracycline were highly influenced by types of soil and amount of organic matter content present. 8.7 and 5.5% of residues of tetracycline and oxytetracycline were present in leachate in Delhi soil and 6.22 and 3.68% in Meghalaya soil, respectively under continuous flow conditions, whereas in FYM and poultry manure residues were not detected in the leachate. Antibiotics showed highest mobility in Delhi soil, followed by moderate movement in Meghalaya soil and least in FYM and poultry manure. Uptake studies revealed that, the uptake of antibiotics also increased as the number of days increased whereas in soil the concentration of antibiotics decreased. For tetracycline there was about 5.54 -18.42 % uptake in plant by 3- 7 daywhereas in soil the decline recorded was 78.3-47.2 % for the same period. Likewise for oxytetracycline an uptake of 6.70-19.15 % in plant and decrease in soil by 75.49-42.53 %, during the period of 3 to 7 day of study was recorded.

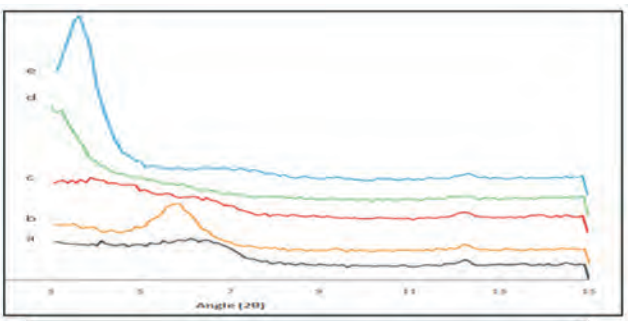
Spiromesifen, (Oberon) is a novel non-systemic insecticide belonging to the new chemical class of tetronic acid derivatives discovered at Bayer Crop Science during the 1990s. It was registered in 2006 for crops like chilli, tomato and okra for protection especially against whiteflies (*Bemisia* spp. and *Trialeurodes* spp.) and mites. It shows excellent ovicidal activity against mites wherein egg hatch in whiteflies is markedly reduced. It is a foliar applied insecticide. There is no report available on persistence and mobility behavior of spiromesifen in soil under Indian condition. In view of the above, persistence and leaching study of spiromesifen in soil was carried out under laboratory conditions. Analytical methodologies for analysis of residues of spiromesifen from soil and water were standardized. Partitioning with dichloromethane for aqueous samples and dipping and shaking with aqueous acetone followed by column clean up with neutral alumina for soil samples were found suitable. Spiromesifen residues were analyzed by HPLC fitted with UV detector. Persistence of spiromesifen was studied in soil collected from IARI campus; at two concentrations, 1.0 and 10 µg g^{-1}, under three different moisture regimes viz. air-dry, field capacity and submerged. It was found that the dissipation of the compound in soil varies with the soil moisture regimes, fortification levels and light conditions. Trend in dissipation was submerged ($t_{1/2}$ 14.3-16.7 days) > field capacity ($t_{1/2}$ 18.7-20 days) > dry ($t_{1/2}$ 21.9-22.9 days). Slightly faster dissipation was observed at 1.0 µg g-1 level as compared to 10 µg g-1 level. Persistence experiments of spiromesifen were also conducted under sterilized dry and submerged conditions at 10 µg g^{-1} concentration level, the half-life values were, sterilized dry 35.8 days and sterilized submerged 17.7 days. Spiromesifen residues persisted in water beyond 120 day with a half-life ranging from 5.7-12.5 days. Dissipation from water was faster at pH 9.2 ($t_{1/2}$ 5.7 days), followed by pH 4 (t1/2 9.7 days) and 7.0 ($t_{1/2}$ 12.5 days). Application of 2.5% compost to soil enhanced degradation of spiromesifen under field capacity moisture regime ($t_{1/2}$14.3 days).

Effect of light on degradation of spiromesifen was studied as thin film on petri plates and soil thin film under field capacity condition at 10 µg g^{-1} by exposure to sunlight , UV-light and dark light. Residues dissipated faster under UV-light ($t_{1/2}$ 3-4 days), in comparison to sunlight ($t_{1/2}$ 5.2-8.1 days) and dark light ($t_{1/2}$ 11.5-13.6 days). Leaching experiments were carried out in packed soil columns. Effect of analytical grade and its formulation on leaching behavior of spiromesifen was studied. Reduced leaching was observed in case of formulation as compared to analytical grade material. Leaching with 400 mL water equivalent to 1156 mm rainfall, 68.08 and 77.5 % residues remained in top 0-10 cm soil layer in case of analytical grade and formulation of spiromesifen, respectively. Effect of various components like FYM, vermicompost and nano clay on mobility of spiromesifen was also studied. In all the cases no residues were observed in the leachate. The total recovery in 0-10 cm soil depth with FYM, vermicompost and nano clay were 84.10, 86.10 and 97.7 %, respectively, indicating high adsorption by nano clay followed by vermicompost and FYM. The results showed that the nano clay adsorbs most of the compound when compared with FYM and vermicompost . Leaching studies conducted with, enol spiromesifen, indicated it to be more mobile than the parent insecticide, spiromesifen. The enol metabolite constituted 17.3 % in the leachate (Mate *et al.,* 2014; Mate *et al.,* 2015).

Decontamination of Pesticides

After industrial revolution, the effluent related pollution problems increased tremendously. Large volume of water is being utilised in the industries for various processes and washing purposes which in turn lead to the generation of large amounts of waste waters containing toxic substances such as heavy metals, pesticides, dyes, organic chemicals and other pollutants. When these effluents are released into the water bodies without proper treatment, they pose serious environmental concerns. Among those industries, pesticide industry is a major culprit in the environmental pollution problem. The wastes generated from the pesticide industries are very toxic and needed to be treated effectively. Various treatment processes are employed in the pesticide industrial effluent treatment plants. Among those techniques, adsorption is an important method for the removal of pesticides from water. Various adsorbents have been exploited for the removal of pesticides from waste water. Clay polymer composites are new area of adsorbents which should be exploited for its pesticide adsorption capacity.

Modified clays and microorganisms were employed to remediate soil and water contaminated with atrazine, the most widely used herbicide. As natural bentonite clay is a poor adsorbent of organic contaminant, it was modified using various quaternary ammonium cations *viz.*phenyltrimethylammonium (PTMA), hexadecyltrimethylammonium (HDTMA), trioctylmethylammonium (TOMA) and stearylkonium (SK, also known as dimethylbenzyloctadecylammonium) to enhance its atrazine adsorption capacity. All organoclays were prepared using organic cation equivalent to 100% of cation exchange capacity (CEC) of the bentonite and SK(2.5)-bentonite was prepared using 250% of CEC (Fig. 9). Modification was confirmed by elemental analysis,X-ray diffraction (XRD) patterns, Fourier transform infrared (FT-IR) and scanning electron microscopy (SEM) analysis.Compared to *d*-spacing of 14.47Å in bentonite, the least increase in *d*-spacing was observed in PTMA-bentonite (14.95Å) while SK-bentonite showed maximum increase (28.52 Å).Adsorption studies suggested that PTMA-modification did not improve atrazine adsorption, however, HDTMA-, TOMA- and SK-bentonites showed 50–70% adsorption at 1:200 clay-solution ratio. The Freundlich adsorption



XRD pattern of:	D (001) Å
a) Bentonite	14.47
b) PTMA-bentonite	14.95
c) HDTMA-bentonite 22.65	
d) SK-bentonite	28.52
e) TOMA-bentonite	24.52

Fig. 9: X-ray diffraction (XRD) patterns of modified clays

constant (K_f) values were 262.9, 373.3 and 298.4 in HDTMA-, TOMA- and SK (1)-bentonite, respectively. Increasing the SK cation loading in SK (2.5)-bentonite decreased the atrazine adsorption. Adsorption of atrazine in organoclays was nonlinear and slope (1/n) values were <1. Desorption of atrazine from organoclays showed hysteresis and TOMA- and SK (1)-bentonites were the best organoclay to retain the adsorbed atrazine. Adsorption of atrazine from waste water effluent of pesticide manufacturing units confirmed the atrazine retention potentials of these organoclays. Atrazine degrading enrichment culture was prepared by its repeated addition to an alluvial soil. Enrichment culture utilized atrazine as sole source of carbon and nitrogen and degradation slowed down when additional sources of carbon and nitrogen were added. Biuret was detected as the only metabolite of atrazine while deethylatrazine, deisopropylatrazine, hydroxyatrazine and cyanuric acid were not detected. Enrichment culture degraded atrazine in an alkaline alluvial soil while no degradation was observed in acidic laterite soil; probably microbial consortium was ineffective at low pH. Enrichment culture was able to withstand high concentrations of atrazine (110 μg g^{-1}) in alluvial soil and complete atrazine degradation was observed within 15 days. Culture independent PCR-DGGE technique was employed to study the change in bacterial community diversity in soil following repeated atrazine application. DGGE Separation pattern confirmed that atrazine untreated control soil had large microbial diversity and repeated atrazine applications resulted in selective enrichment of only those bacteria which could tolerate/utilize atrazine. Sequencing of 16S rDNA region revealed that the enrichment culture was a consortium of bacteria belonging to genus *Bacillus*, *Pseudomonas* and *Burkholderia*. Atrazine degrading potential of enrichment culture was evaluated in industrial waste water effluent. Enrichment culture was effective in degrading atrazine (27.7 μg mL^{-1}) and cyanuric acid (400 μg mL^{-1}) in waste water effluent while higher levels of atrazine/cyanuric acid were found to be toxic to the microbial consortium (Ghosh and Singh, 2009a; Ghosh and Singh, 2009b; Ghosh and Singh, 2013).

Clay embedded porous polymer materials were prepared for simultaneous removal of pesticides and heavy metals. Three modified clays were prepared from commercially available bentonite clay using quaternary salt of cetyltrimethyl ammonium bromide and sodium hexametaphosphate, separately as well as combined and characterized by FTIR and XRD. Four clay embedded polymer materials were prepared by using normal bentonite with different monomers namely styrene, methyl methacrylate, methyl acrylate, acrylamide separately by suspension polymerization technique. Out of these four prepared polymeric material, composite prepared from acrylamide was found hydrophilic. The remaining three prepared polymer materials were characterized by using FTIR, SEM and TEM. The SEM analysis revealed that the porous property was present only in the polymer prepared by using methyl acrylate as monomer. Therefore, three modified clay embedded porous polymer materials were prepared using modified methyl acrylate as monomer. All composite materials prepared from normal bentonite, modified bentonite and modified clay embedded hydrophobic polymers with styrene, methyl acrylate and methyl methacrylate were screened for adsorption of pesticides (chlorpyriphos and pretilachlor) and heavy metals (lead and chromium) in a mixture. In general, all the materials adsorbed pesticides as well as heavy metals. The materials containing cetyltrimethyl ammonium bromide as modifier either alone or in combination recorded high pesticides adsorption (> 85%) whereas materials containing hexametaphosphate either alone or in combination recorded high heavy metals adsorption (65-85% for chromium and 56-62% for lead).

Natural polymer-clay composites were prepared for adsorption of nine selected pesticides, atrazine, butachlor, carbendazim, carbofuran, imidachloprid, isoproturon, pendimethalin, thiophanate methyl and thiamethoxam. For that a multiresidue method has been standardised for the analysis of these nine selected pesticides, using LC-MS-MS. The LOD for the pesticides ranged between 0.0001 and 0.0002 μg/mL and LOQ between 0.0003 μg/mL and 0.06 μg/mL for the selected pesticides. The method recovery was found to be more than 80% at different fortification levels.Three natural polymers, cellulose, carboxy methyl cellulose and chitosan, were used for the preparation of clay polymer composites. Among the clays, normal bentonite, nano bentonite, nano bentonite modified with 25-30 wt % octadecylamine (ODA), nanobentonite modified with 35-45 wt % dimethyl dialkyl (C14-C18) amine (DMDA) and nanobentonite modified with 15-35 wt % octadecylamine and 0.5-5 wt% aminopropyltriethoxysilane (ODAAPS), DMDA showed the maximum adsorption of pesticides (42.8 to

99%) and was selected for the preparation of composites. for the comparison purpose, normal bentonite and nano bentonite were also utilised for the preparation of composites. Clay polymer composites were prepared by solution intercalation method by maintaining the polymer to clay ratio of 1:2. Among the prepared composites, 1:2 CMC-DMDA was found to be the best and detailed adsorption study was carried. Favourable adsorption of the pesticides on the composite was observed. The pesticides showed higher adsorption at basic pH. In order to improve the pesticide removal efficiency of the composites, the polymer to clay ratios were varied and screened. As the clay amount increased, the pesticide adsorption was also 105 increased. 1:5 CMC-DMDA composite showed the maximum adsorption of pesticides (55.0 to 99.7%). For further improvement in the removal efficiency of the composites for atrazine, mixed clay composites were prepared using CMC and combinations of organoclays, DMDA and ODAAPS. When ODAAPS was used instead of DMDA in 1:5 composite, the removal efficiency for atrazine was improved from 55.0% to 60.9%. Further improvement in the overall pesticide removal efficiency was observed when 1:2.5:2.5 CMC-DMDA-ODAAPS was used (64.9-99.9%). The natural polymer composite 1:2.5:2.5 CMC-DMDA-ODAAPS was found to be the better than the synthetic ones prepared from poly acrylic acid (1:2.5:2.5 AA-DMDA-ODAAPS) and poly acrylamide (1:2.5:2.5 AM-DMDA-ODAAPS). Detailed adsorption study was carried out using 1:2.5:2.5 CMC-DMDA-ODAAPS and the data were fitted into different isotherms. Thiophanate methyl showed the maximum adsorption capacity and the adsorption of pesticides on the composite was found to be favorable and followed physisorption process. The composite was further utilised for the decontamination of pesticides. As the composite dose increases, the adsorption of pesticide also increases. The prepared composites were characterised by infrared spectroscopy (FTIR), X-ray diffraction spectroscopy (XRD), Scanning electron microscopy (SEM) and Electron dispersive X-ray (EDX) analysis and Transmission electron microscopy (TEM). The basal spacing was increased when the composite was prepared by incorporating CMC and clays. Intercalation of polymer into the clay layers is confirmed. The composites formed were found to be flaky. Pesticide industrial effluent collected from the nearby industry was characterised by GC-MS and LC-MS-MS. The before treatment sample was found to contain only one pesticide, thiamethoxam at a concentration level of ~50 µg/mL. So the effluent was fortified with other pesticides at the same level and decontamination study was carried out using the prepared composite. The effluent treatment using the composite dose of 5g (thrice) gave the highest removal efficiency for the selected pesticides (72.9 to 100.0%). Efficient regeneration of the used composite was obtained by acetone washing followed by thermal regeneration at 200°C. Even after second reuse, the composite was able to maintain 57 to 97% of its pesticide removal efficiency. The pesticides generated during the regeneration process can be effectively degraded by using 2N NaOH. The prepared composite can be utilised at some stages of the effluent treatment process in order to bring down the pesticide concentration considerably. The use of the biopolymer nano organoclay composite prepared is an efficient method for the decontamination of water containing pesticides. The preparation and regeneration of the composite is easy and economical making it a promising adsorbent for the pesticides.

Detection of pollutants in complex environmental matrix with high selectivity and sensitivity constitute a challenging task. Recent advancements in molecularly imprinting polymers (MIPs) technology have provided a versatile tool for the highly selective quantification of these toxicants in food matrices. In the present work molecularly imprinted and non-imprinted polymers were synthesized and utilized for selective detection of chlorpyrifos and imidacloprid in brinjal and honey. Analytical methods involving GC-ECD for chlorpyrifos and HPLC-PDA and LC-MS/MS for imidacloprid were developed for quantification of residues. The calibration curves were found to be linear for chlorpyrifos from 0.01 to 5 µg mL-1 with R^2 value of 0.967 and from 0.1 to 10 µg mL-1 with R^2 value of 0.993 for imidacloprid. Recovery of chlorpyrifos from water and water: methanol mixture revealed that dichloromethane could recover more than 85% of the added pesticide from freshly fortified solution. Nine molecularly imprinted and non-imprinted polymers (MIPs) each, including magnetic MIP, were prepared for chlorpyrifos and imidacloprid using different combinations of functional monomer (MAA, MA, MMA, AM or AA) and cross-linker (EGDMA or TRIM). Prepared materials were characterized by FT-IR, SEM and TEM techniques. Screening of the prepared MIPs and NIPs revealed much higher sorption and selectivity of MIPs (30.67-92.67%). This may be due to the presence of cavities complimentary in shape and size to the template molecule on MIP surface, which serves as specific recognition sites for target molecule.

TRIM based MIPs showed less sorption capacity (78.23-83.04%) as compared to EGDMA (77.52-92.67%) based MIPs probably due to high concentration of high molecular weight TRIM in the polymerization solution resulting in the formation of denser and less porous material having less recognition sites. Adsorption studies with the MIPs and NIPs revealed increased sorption with increase in adsorbent amount. The sorption data for both chlorpyrifos and imidacloprid fitted well to pseudo second order kinetics model with correlation coefficient value of 0.999 indicating involvement of chemisorption process in adsorption. Fitting of adsorption data into Freundlich and Langmuir isotherms revealed that the data fitted well into linearized Freundlich equation (R^2>0.98) indicating multilayer adsorption of pesticides onto the MIPs. Two straight-line curves obtained for chlorpyrifos and imidacloprid in Scatchard chart is suggestive of the presence of two different types of binding sites on MMIP. Chlorpyrifos MMIP shows much higher absorption and selectivity for chlorpyrifos as compared to lindane, cypermethrin, quinalphos and triazophos. Similar studies conducted with imidacloprid MMIP, in presence of carbofuran, azoxystrobin, acetamiprid and thiamethoxam also revealed high sorption and selectivity of MMIP towards imidacloprid. High selectivity towards template is indicative of the formation of specific recognition sites in the MMIP. Regeneration and reusability studies conducted with MMIP revealed that washing with methanol: acetic acid with sonication could remove more than 85% of the bound template. Regenerated MMIPs were successfully used for removal of chlorpyrifos and imidacloprid from solution in three successive adsorption cycles without any significant loss in their removal efficiency. Percent removal of chlorpyrifos and imidacloprid in three adsorption cycles varied from 89.73-92.02% and 88.68-91.10 % respectively. The feasibility and applicability of MMIP were tested in fortified honey and brinjal samples. Calculation of recovery percent revealed 90.02% and 87.89% of the added chlorpyrifos was recovered from MMIP in case of brinjal and honey respectively. Similarly 87.07% and 90.55% of the added imidacloprid in brinjal and honey samples was recovered from MMIP. Results revealed that these MMIP can be used as SPE adsorbent for cleanup of complex matrices for recovering chlorpyrifos and imidacloprid (Pritam and Mukherjee, 2010; Pritam and Mukherjee, 2013).

Fruits are often found with unsafe level of pesticide residues. Besides washing the most recent method being popularized for post-harvest removal of pesticide residues in fruits is the use of ozone water dips. Ozone is a strong oxidant. Citrus fruits are good source of antioxidant compounds which are good for health. Keeping this in view studies were conducted to evaluate the effect of ozonation on phytochemicals during pesticide removal in two citrus fruits i.e. grapefruit and kinnow. For this a commercial ozonizer was used. Fruits were taken from IARI orchard where three pesticides – dimethoate, imidacloprid and carbosulfan were applied during the growth period of plant and fruit formation. GC method was standardized for analysis of dimethoate and HPLC method for imidacloprid and carbofuran (as carbosulfan is a pro pesticide) in fruit juice and pulp+peel. Analysis of the fruits showed only the residues of imidacloprid in fruits. Fruits were subjected to ozonation treatment for 15 and 30 minutes. Washing treatment was also included as third technique. It was found that both ozonation and washing decreased residues of imidacloprid. Ozonation for 15 minutes reduced the pesticide residue in grapefruit and kinnow up to 48.58 and 47.76% respectively which were at par to washing. Similarly, 30 minutes ozonation removed the pesticides by 64.15 and 57.20%. Effect of ozonation was evaluated on different phytochemicals of citrus fruits e.g. organic acids (oxalic, tartaric, malic, malonic, ascorbic and citric), β-carotene, lycopene, hydroxycinnamic acids (ferulic acid and p-coumaric acid), and polyphenols in grapefruit and kinnow. For this methods were standardized for the extraction and HPLC analysis of these phytochemicals from both the citrus fruits. Organic acids were not much affected by ozonation except ascorbic acid in kinnow where 24.04% reduction of ascorbic acid content was found on 30 min ozonation. On ozonation lycopene content in grapefruit was decreased nearly 2.35% in comparison to 24.8% of β-Carotene. Direct ozonation of juice samples showed that organic acids were also affected by ozonation. In general oxalic acid increased, ascorbic acid degraded too fast, citric acid decreased while tartaric acid is least affected. But due to the thick and hard outer peel and structure of citrus fruits the organic acids were not very much affected on ozonation in whole fruit except ascorbic acid. Ferulic acid and p-coumaric acid both were decreased, however in grapefruit percent decrease was less. In kinnow nearly 38.8 and 34.8% decrease was observed for ferulic acid and p-coumaric acid respectively. Study of polyphenols also revealed that ozonation decreased most of the polyphenols; however

some showed increase as well. Individual compounds of polyphenols could not be identified due to the non-availability of authentic samples. Some polyphenols increased with ozonation treatment while others were found to be decreased.Thus, ozonation was proved to be an effective method for post-harvest reduction of pesticide residues from citrus fruits without affecting many of the organic acids. However, the concentrations of other phytochemicals have been altered by ozonation. The reduction of antioxidants such as ascorbic acid, ferulic acid, p-coumaric acid, and β-carotene are a matter of concern (Swami *et al.,* 2016a).

Studies were conducted to evaluate the efficiency of ozonation method of pesticide removal in tomato, apple, capsicum and grapes using commercial ozonation equipment. As ozone is a strong oxidant, its potential effect on nutritional quality of these fruits and vegetables has also been investigated. Multi-residue GC method was standardized for analysis of six pesticides – methyl parathion, chlorpyrifos, cypermethrin, chlorothalonil, azoxystrobin and hexaconazole in tomato, apple, capsicum and grapes. Fruits and vegetables were fortified by dipping in 10 µg g-1 aqueous solution of pesticides and then subjected to ozonation treatment for 15 and 30 minutes. Washing treatment was done by dipping fortified fruits/vegetables in water for 30 minutes. It was found that both ozonation and washing decreased residues of all pesticides, reduction by ozone increased with time. Ozonation for 15 minutes reduced the pesticide residue in tomato, apple, capsicum and grapes in the range of 20.18-69.84, 26.91-73.58, 18.25-96.81 and 12.44-87.09 % respectively. Similarly, 30 minutes ozonation removed the pesticides by 38.78-90.25, 39.39-95.14, 48.67-96.95 and 45.8295.43 % respectively in tomato, apple, capsicum and grapes. Washing method was more effective than 15 minutes ozonation in some of the experiments. However, ozonation for 30 minutes gave the best removal for all the pesticides. Different fruits behaved differently in pesticide removal. In capsicum all the pesticides except methyl parathion were best removed by 30 minute ozonation. Non systemic and non-polar pesticides are better removed from fruit surface. Chlorothalonil was the most removed pesticide in all the fruits. Degradation of pesticides by ozonation in water was also studied. Methyl parathion was fastest degrading pesticide with half-life of 0.33 minutes followed by chlorpyrifos (1.75 min). Azoxystrobin showed biphasic degradation with first rapid phase(0.18 min) followed by second slow phase (3.11min). Half-life values of other pesticides ranged from 2.15 to 2.84 minutes. GC-MS analysis of aqueous samples showed the formation of methyl paraoxon as pesticide degradation product of methyl parathion. As the paraoxon is a toxic compound the presence of such degradation products is also a matter of concern. Effect of ozone on ascorbic acid, ß-carotene, lycopene, cyanidine-3-glucoside and polyphenols in fruits and vegetables was studied. Ozonation resulted in reduction of ascorbic acid content of all the fruits and vegetables ranging from 13.37-65.72% and 24.8-77.46% for 15 and 30 minutes respectively. Lycopene content in tomato increased from 145.53 µg g-1 to 177.55 and 196.54 µg g-1 in 15 and 30 minutes ozonation 121 respectively. ß-Carotene increased from 5.25 µg g-1 to 5.49 and 8.61 µg g in 15 and 30 minutes respectively. Cyanidin-3-glucoside content in apple was reduced by 29 and 62.8 % respectively by 15 and 30 minutes ozonation. Eleven polyphenolic compounds – gallic acid, 3,4-dihydroxybenzoic acid, catechin, epicatechin, syringic acid, p-coumaric acid, rutin, quercetin-3-O-glucoside, quercetin, resveratrol and kaempferol analyzed by LCMS in non-ozonated and ozonated samples of fruits and vegetables showed different trends in different fruits. Some polyphenols increased with ozonation treatment while others were found to be decreased. Thus, ozonation proved to be an effective method for post-harvest reduction of pesticide residues. However, the concentration of nutritional components has been altered by ozonation. The reduction of antioxidants such as ascorbic acid and anthocyanins like cyanidin-3-glucoside are a matter of concern (Swami *et al.,* 2016b).

Biochar, carbon rich residue obtained from pyrolysis of biomass, have been recommended as amendment in agricultural soils for carbon sequestration and improve their physico-chemical properties and nutrient status. Soil amendments including biochar play an important role in managing the pesticide residues in the environment. Pyrazosulfuron-ethyl, a sulfonylurea herbicide, is recommended for weed control in transplanted- and direct seeded-rice cultivation. Therefore, efforts have been made to study the effect of wheat and rice biochars on the sorption, degradation and leaching behaviour of pyrazosulfuron-ethyl in organic carbon poor Inceptisol. Wheat (WBC400 and WBC600) and rice (RBC400 and RBC600) straw biochars were prepared at 400°C and 600°C temperature and were characterized for their physico-chemical properties using XRF, XRD, FTIR,

^{13}C-NMR and SEM-EDX. Herbicide adsorption capacity of biochars was affected by the pyrolysis temperature and nature of feed stock and rice biochars were better than the wheat biochars. Deashing of biochars further enhanced their herbicide adsorption potential. Adsorption of pyrazosulfuron-ethyl on biochars was affected by the pH, total organic carbon, polarity, surface area and porosity of biochars.Amendment of biochars at 0.1, 0.2 and 0.5% levels significantly enhanced the pyrazosulfuron-ethyl adsorption capacity of soils and effect varied with nature/dose of biochar amendment. Sorption data fitted well to the Freundlich adsorption isotherm and isotherms were non-linear. Isotherm non-linearity increased with increase in dose of biochars and in soil amended with 600°C temperature biochars. The Freundlich adsorption coefficient (Kf) values were higher in rice biochar-amended soils and increased with increase in level of biochars. Desorption of pyrazosulfuron-ethyl in biochar amended soil showed hysteresis and biochar amendment significantly reduced herbicide desorption. Pyrazosulfuron-ethyl was poorly retained in the Inceptisol, but biochar-amendment significantly reduced herbicide leaching losses. Effect of biochars on herbicide leaching corresponded to their herbicide adsorption potential. Biochar-amendment increased herbicide retention in 0-15 cm soil profile. Biochar amendment to soil increased herbicide persistence in soils, especially nonflooded soils and effect was affected by the dose / nature of biochar. Hydrolysis at sulfonylurea and ester linkage was the main mechanism of pyrazosulfuron-ethyl degradation. These findings are significant in assaying the effect of biochars on fate of pyrazosulfuron-ethyl in agricultural soil (Fig. 10).

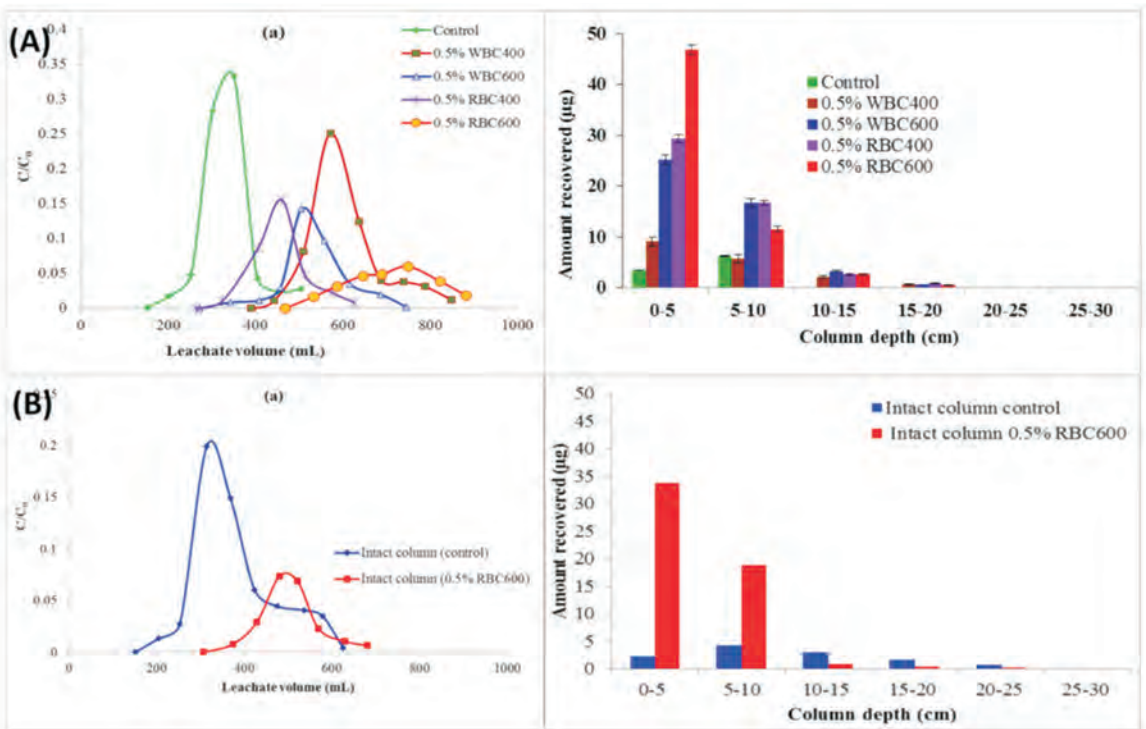


Fig. 10: Leaching of pyrazosulfuron-ethyl in biochar amended soil columns (A) Packed column and (B) Intact column

Performance of granular carbon has been evaluated for the removal of two triazine herbicides i.e. atrazine and metribuzin and two acetanilide herbicides i.e. alachlor and metolachlor. Adsorption desorption studies of atrazine, metribuzin, alachlor and metolachlor by batch method indicated the highest adsorption of triazine than acetanilide herbicides. Between atrazine and metribuzin, adsorption capacity of atrazine (K_f =17378) was found more than metribuzin (K_f =16710) when studies individually. Similarly, metolachlor (K_f =16033) has higher adsorption capacity than alachlor (K_f =4217). The trend of adsorption was atrazine> metribuzin > metolachlor> alachlor. This trend was found when adsorption studies were conducted with three sequential

adsorption. As the number of adsorption cycle increased more of metolachlor was sorbed on the carbon, which may be due to presence of available sites on granular carbon. The value of 1/n for alachlor indicated that there was a proportionate increase in adsorption of alachlor with concentration. The percent adsorption of alachlor was not affected by concentration. While for metolachlor 1/n value (0.34) value of intercept indicates that adsorption does not increase proportionately with concentration, although its adsorption intensity is more than alachlor. Desorption studies revealed that atrazine, metribuzin and metolachlor were not desorbed, but alachlor was desorbed in each cycles of desorption.When adsorption was carried out with the mixture of these four herbicide, metribuzin was the most adsorbed herbicide with a trend of metribuzin> atrazine> alachlor> metolachlor. In mixture there remains a competition between herbicides for adsorption sites and metribuzin having the more adsorption intensity (n= 2.38) than atrazine (n= 1.75) is more adsorbed followed by metolachlor and alachlor. In desorption atrazine and metribuzin were no desorbed and only metolachlor and alachlor were desorbed. Percent desorption of alachlor was more than metolachlor. Although water solubility of metolachlor (488 µg mL-1) is much higher than alachlor (242 µg mL-1), still alachlor was more desorbed which could be explained on the basis of adsorption intensity of alachlor (n=1) which was lower than metolachlor (n=3.). In column studies at higher concentration (1 and 5 µg mL-1), metolachlor was highly detected herbicide with a trend of metolachlor> alachlor> atrazine> metribuzin. But at lower concentration (0.1 µg mL-1), the most mobile and least adsorbed herbicide was alachlor. More elution of metolachlor at higher concentrations may be due to the less available site of adsorption in the presence of higher concentration. The performance of horizontal column was found better than vertical column.

Summary

N-propyl-butyramide, N-propyl-pentamide and N-propyl-hexamide, synthesised by using *Candida antartica* lipase at 60-90°C, exhibited nematicidal activity against J_2 of *Meloidogyne incognita* with lower LC_{50} values,67.5, 83.5 and 96.5 ppm, respectively.

Hexane extract of dried leaves of *Stevia rebaudiana* showed maximum antifungal activity *Rhizoctonia bataticola* (ED_{50} = 26.62 ppm) and *Fusariumoxysporum* (ED_{50} = 41.14 ppm) and *Sclerotium rolfsii* (ED_{50} 40.61 ppm) as compared to water, ethyl acetate, methanol and dichloromethane extract.

Sterebins were isolated from hexane extraction of *Stevia rebaudiana* with effectivity against *Rhizoctonia bataticola* (ED_{50} = 59.90 ppm)

N-alkyl substituted amides, based on various phenolic acids, namely salicylic acid, 3-hydroxy cinnamic acid, p-coumaric acid, caffeic acid, ferulic acid, o-coumaric acid and cinnamic acid, have been synthesized by reacting with different alkyl amines viz. propyl amine, hexyl amine, heptyl amine, undecyl amine, hexadecyl amine and octadecyl amine in presence of *Candida antarctica* lipase at 60-90°C in 16-20 hours.

All the N-alkyl substituted amides possessed promising activity, N-hexyl-salicylamide, N-hexyl-2-hydroxycinnamamide and N-hexyl-4-hydroxycinnamamide were found to be more potent with LC_{50} values of 54.75, 61.791 and 74.11 ppm respectively, against *Meloidogyne graminicola.*

Schiff bases were derived from 2, 4, 5-trichloroaniline and explored for their antifungal activity against *R. bataticola* and *S. rolfsii* and nitrification inhibitory activity under laboratory conditions.

These Schiff bases were more effective against *R. bataticola* (ED_{50}, 9.4-66.2 µg/ml) as compared to *S. rolfsii* (ED50, 10.6-117.1 µg/ml).

Anthocyanins were extracted from berries of *Berberis lyceum* Royleusing methanol containing 0.1% HCl.

Major anthocyanins extracted from berries of *Berberis lyceum* Royle were characterised as delphinidin-3-glucoside (43.7%) and cyanidin-3-glucoside (40.1%). Five minor anthocyanins were delphinidin-3-rutinoside (0.7%), pelargonidin-3-rutinoside (1.36%), cyanidin-3-rutinoside (5.08%), petunidin-3-rutinoside (1.38%), and peonidin-3-rutinoside (6.19%).

Turmeric fixed oil (yield 5.7% w/v) from rhizome powder was extracted with hexane and purified by column chromatography for separation of bioactive terpenoids (a, ß- turmerone).

Curcuminoids (curcumin-I, curcumin-II and curcumin-III) were extracted from de-fatted turmeric powder with ethyl acetate and purified using column chromatography.

Ellagitannins wereextracted from pomegranate rind and the process was standardized after comparing different extraction techniques.

Nanosuperabsorbent composites (NSAPC) were prepared by *in situ* solution polymerization of a novel biopolymer of plant origin and clay mineral using a green chemistry technique.

Guar gum (GG), a plant origin polymer was employed to prepare GG-*g*-cl-polyacrylate superabsorbent (GG-SAP) and superporous hydrogels (GG-SPH).

GG-*g*-cl-polyacrylate/clay superporous hydrogel composites (GG-SPHC) were prepared with improved water absorption and retention properties.

Guargum was also employed to develop core-shell nanogels (GG-CSH) that exhibited mean particle size in the range of 11 nm to 490 nm.

Amphiphilic polymers were synthesized using poly (ethylene glycols) (PEG) of different molecular weights, as hydrophilic block and aliphatic diacids and aromatic diesters as hydrophobic block in presence of catalyst Conc. H_2SO_4.

Amphiphilic polymers were used encapsulate different pesticides (imidacloprid, thiamethoxam, carbandazim,azoxystrobin and thiram) to develop controlled release pesticide formulations.

Sodium alginate based nanosuspension of pyridalyl was developed with improved bioefficacy against *Helicoverpa armigera.*

Hydrogel based formulation of hexane fraction of root extract of *Tagetes patula* and magnesium sulphate was prepared. These integrated formulations showed better nematicidal activity and nutrient management strategies.

Guargum hydrogels based bioformulations of *Trichoderma harzianum* and *Pseudomonas fluorescens* were prepared with improved shelf life and these bioformulations showed better management strategy for controlling *Pythium aphanidermatum* under laboratory conditions.

Core-shell nanogels based on guargum were developed as carrier materials for agrochemical formulations.

Modified bentonites were prepared using quaternary ammonium cations*viz.*phenyltrimethylammonium (PTMA), hexadecyltrimethylammonium (HDTMA), trioctylmethylammonium (TOMA) and stearylkonium (SK, also known as dimethylbenzyloctadecylammonium)to enhance its atrazine adsorption capacity.

Porous polymeric materials were prepared by reacting different monomers namely styrene, methyl methacrylate, methyl acrylate, acrylamide with organically modified bentonite clay by suspension polymerization technique. Hydrophobic polymeric matrix with styrene, methyl acrylate and methyl methacrylate were screened for adsorption of pesticides (chlorpyriphos and pretilachlor) and heavy metals (lead and chromium) in a mixture.

Natural polymer-clay composites were prepared, by reacting bentonite, nanobentonite and organically modified nanobentonite with cellulose, carboxy methyl cellulose and chitosan, for adsorption of nine selected pesticides, atrazine, butachlor, carbendazim, carbofuran, imidachloprid, isoproturon, pendimethalin, thiophanate methyl and thiamethoxam.

Molecularly imprinting polymers (MIPs)were prepared for chlorpyrifos and imidacloprid using different combinations of functional monomer and cross-linker.

Effect of ozonation on phytochemicals during pesticide (dimethoate, imidacloprid and carbosulfan) removal in two citrus fruits i.e. grapefruit and kinnow were studied. Organic acids were not much affected by

ozonation except ascorbic acid in kinnow where 24.04% reduction of ascorbic acid content was found on 30 min ozonation. On ozonation lycopene content in grapefruit was decreased nearly 2.35% in comparison to 24.8% of β-Carotene.

Efficiency of ozonation method of pesticide removal in tomato, apple, capsicum and grapes using commercial ozonation equipment were studied. It was found that both ozonation and washing decreased residues of all pesticides, reduction by ozone increased with time. Ozonation resulted in reduction of ascorbic acid content of all the fruits and vegetables ranging from 13.37-65.72% and 24.8-77.46% for 15 and 30 minutes respectively. Lycopene content in tomato increased from 145.53 µg g-1 to 177.55 and 196.54 µg g-1 in 15 and 30 minutes ozonation 121 respectively. ß-Carotene increased from 5.25 µg g-1 to 5.49 and 8.61 µg g in 15 and 30 minutes respectively.

Wheat and rice biochars on the sorption, degradation and leaching behaviour of pyrazosulfuron-ethyl in organic carbon poor Inceptisol was studied. Pyrazosulfuron-ethyl was poorly retained in the Inceptisol, but biochar-amendment significantly reduced herbicide leaching losses.

Performance of granular carbon has been evaluated for the removal of two triazine herbicides i.e. atrazine and metribuzin and two acetanilide herbicides i.e. alachlor and metolachlor.The trend of adsorption by granular carbon was atrazine>metribuzin>metolachlor>alachlor (Sarkar *et al.,* 2012).

Future Perspective

Agrochemicals are an essential component of pest management strategies, but their indiscriminate use has led to the development of resistance in pests and environmental contamination. Rational use of pesticides which are selective and are potent even in low doses and their advanced formulation and application technology are more critical for plant protection. The challenges lie in the use of pesticides as a common strategy to manage insect, mite and nematodes for holistic protection. It will also be necessary to adopt other methods like biological control, behavioral manipulation, and use of plant products as behavior modifying chemicals in a judicious blend taking in to account the conditions of farming and farmers. Currently, the pesticides used in the country are imported as technical or ready to use formulations. There is a need to develop new novel agrochemicals with increased activity, selective toxicity and non-target organisms. India has made pioneering contributions to the use of botanicals as pesticides, particularly, the products derived from *Neem*. The plant biodiversity in the India needs to be extensively explored using modern tools to discover new botanicals and bio-control agents. Strategies need to be put in place to mitigate the problems of pest resistance, resurgence and out breaks of secondary pest through development and promotion of multipesticide formulations, pesticide adjuvants, such as synergist and integrating their use with other pest control strategies. There is a urgent need to generate holistic data on the bio-efficacy, bio-safety, toxicology and persistence of the pesticides in use in the country, with the help of research institutions, industry and regulatory agencies.

Acknowledgment

We wish to thank all the Faculty Members of the Division of Agricultural Chemicals for extending support and cooperation in preparing the research contributions of M.Sc. and Ph.D. students.

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Entomology

Subhash Chander

Introduction

The Division of Entomology established in 1905 as one of the five major Divisions of the then Imperial Agricultural Research Institute located at Pusa, Bihar was shifted to its present premises in 1936. Eminent entomologists like H.M. Lefroy, T. B. Fletcher, H.S. Pruthi, S. Pradhan and K.N. Mehrotra laid strong foundation for basic and applied research in Insect Science.Their pioneering contributions resulted in the publication of a monumental reference work *viz.,* 'Indian Insect Life' in 1906 by Lefroy and 'Text Book of Agricultural Entomology' by Pruthi. Since then the identification of the insecticidal principles of neem, the concept of Integrated Pest Management (IPM), periodicity of locusts, mode of action of DDT and development of 'Pusa Bin' stand out prominently in the annals of entomological research of our country. The Division has pioneered in investigations in Insect Biosystematics, Insect Physiology and Toxicology, Biological Control and Economic Entomology.

National Pusa Collections (NPC) with over five lakh specimen serves as a national repository for insect pests of crops. Currently, CD-ROM based diagnostics are being developed for various crop pests. Over the last 50 years, 1500 new species of insects have been described from NPC. As a national service for pest diagnostics, every year, over 4000 specimens are identified by the taxonomists of this Division. Several taxonomic treatises on agriculturally important insects belonging to orders Lepidoptera, Coleoptera, Hemiptera, Orthoptera and Hymenoptera and class Acarina have been made from this Division. Significant work is being undertaken on description/redescription of economically impotant species of ants, non *Apis* bees, hymenopterous parasitoids, leaf hoppers and white grubs that has also resulted in discovery of new species of insects.

Insect toxicology studies brought out relative toxicity of insecticides against major crop pests and technology for development of their formulations. Besides, studies were also carried out on persistent toxicity, residue limits and safer waiting periods etc. Besides elucidating the mode of action of azadirachtin, the antifeedant principle of neem seeds, a number of indigenous floras have been investigated for identification of natifeedant and insect growth regulatory properties (IGR). The Division was the first to report insecticide resistance (IR) in Singhara beetle, *(Galerucella birmanica)* and has played important role in monitoring IR management in *Helicoverpa armigera*. Biochemical basis of insecticide resistance was extensively investigated. Carboxyl esterases were found to impart an ability to degrade pyrethroids in the resistant strains of *H. armigera*. Likewise, studies on baseline susceptibility of the diamondback moth and the American bollworm to *Bacillus thuringiensis (Bt)* have led to the understanding of genetics of *Bt* resistance in crop pests. Insecticide resistance has been evaluated in *Spodoptera litura*, *Bemisia tabci* and *Tribolium castaneum* populations collected from different parts of the country. Several native strains of *Bt* from diverse habitatsare being characetrized and evaluated against impotant pests including defoliators, storage pests and sucking pests. Likewise, studies on diversity of gut bacteria in white flies and fruit flies, virus-vector relationship and on nutritional biochemistry of pests have been initiated.

The Division has established a strong unit of biological control. *Apanteles flavipes* was exported to Barbados for the control of sugarcane borer, *Diatrea saccharalis* in the 1970s. Presently more than two-dozen parasitoids, predators and microbial pathogen species are reared for investigations. Predators *viz.*, *Crypolaemus montrouzieri*

and *Scymnus coccivora* were introduced in Caribbean countries for the control of mealy bug in 1996. The discovery of a nucleopolyhedrovirus (NPV) of tobacco caterpillar in 1969 led to intensive research on insect pathogenic viruses, bacteria and fungi. Genomic mapping of baculoviruses of insect pests during 1980s laid strong foundation for insect molecular biological research in the Division. A major attempt is being made to develop temperature insensitive strains of *Trichogramma* for our climatic conditions. Likewise, effect of thermal stress on host-parasitoid interaction involving mealy bugs and their parasitoids is being assessd. Studies on tritrophic interactions to enhance parasitoid activity are one of the current thrusts of this Division.

Studies on the the biology and bionomics of major insect pests laid the foundation for adoption of eco-friendly IPM approaches. Significant contributions were made to understand the biology and host plant resistance of major insect pests. Biochemical basis of host plant resistance in maize against *Chilo paretellus* has been investigated thoroughly. Significant work has been done on development of insect forewarning and monitoring tools, spectral signatures and crop-pest simulation models. Likewise, research on key factor analysis, pest risk analysis, and climate change impact assessment and required pest management adaptations is also being undertaken. Effect of habitat management practices such as inter- and trap-crops on conservation of natural enemies of pests has also been evaluated. Besides, role of herbivore-induced plant volatiles (HIPV) in enhancing natural enemy populations is being investigated.

Recently, this Division has established a 'National facility for insect rearing' for developing standalone technologies for rearing insect pests for analytical studies and for developing novel artificial diets for several crop pests. The mandates of the Division are:

- To conduct basic and strategic research in Entomology
- To develop as centre for academic excellence in the area of research, post-graduate educational and human resource development
- To provide services in insect science and technologies.

Significant Student's Research Achievements

Biosystematics

Investigation on the subfamily, Myrmicinae from India included taxonomic studies on 42 species under 16 genera belonging to 9 tribes. Diagnostic keys for quick identification of all taxa up to the level of species have been formulated. The spatial and temporal biological diversity of ants belonging to subfamily Myrmicinae vis-à-vis total diversity of ants was investigated through various diversity indices. Species richness fluctuated between 1 and 4 at each sampling site throughout the sampling duration and its diversity was restricted to only 6 different species across all the sites. During the *kharif* season *Pheidole indicum Monomorium, scabriceps*, *M. indicum* and *Meranoplus bicolor* were found in the rice field among which *P. indicum* was the most dominant while in the soybean field. *Aphaenogaster beccarii, M. himalayanus* and *M. indicum* were abundant with *M. himalayanus* dominating over the others. However during the *rabi* season, *M. himalayanus* was the only species found in both chick pea and mustard fields. Further, *M. himalayanus* was found to act as a seed harvester ant in the mustard field (Meshram, 2008).

In another study in the subfamily, Formicinae on 43 species were studied under the genera, *viz.*, *Acropyga, Anoplolepis, Camponotus, Cataglyphis, Formica, oecophylla, Lepisiota, Paratrechina, Polyrhachis* and *Pseudolasius. Camponotus (Orthonoto myrmex) sericeus* was found to possess metapleural gland, which happened to be the first species being recorded from India and only third species of *Camponotus* in the world to possess this gland. A new species *Camponotus dapoliensis* was also reported and described. The spatial and temporal biological diversity of ants belonging to subfamily Formicinae vis-à-vis total diversity of ants collected was studied at four diverse sampling sites. A total of only five different species could be collected from the various sites,

viz., C. compressus, C. parius, C. sericeus, L. frauenfeldi and *C. setipes*. The species *C. compressus* was found at all microhabitats as a dominant species. The *C. sericeus* was captured only at the grassy lawn, while *C. setipes* was found at all microhabitats except at the grassy lawn there by indicating the influence of the vegetation present at the site (Coumar and Dey, 2008).

Accurate identities were established for 29 species under 9 genera of halictid bees (Family Halictidae) including five species new to science *viz.*, *Pseudapisindorenis*, *Lipotriches ma kaliammani*, *L. anaiyurensis, L. sathiarensis* from Tamil Nadu and *Lipotriches jhajhraensis* from Uttarakhand under subfamily Nomiinae. Two species *Nomia strigata* and *Nomioides fortunatus* were recorded for the first time from India. An up to date annotated checklist of the Indian species has been compiled from all the published literature available on family Halictidae for the last three and a half centuries with inclusion of all the available synonyms. Sub family Halictinae was the most abundant that included six genera and 120 species under tribe Halictini. The genus *Lasioglossum* Curtis of Halictinae was more diverse with 75 species, followed by the genus *Lipotriches* Gerstaecker and *Nomia* Latreille of Nomiinae (Balaji and Dey, 2016).

Genus *Andrena*, a well-known genus of family Andrenidae commonly known as sand bees or mining bees, was investigated based on 300 specimens including those available at National Pusa Collection (NPC) and those collected from Delhi, Rajasthan, Uttarakhand and Himachal Pradesh. This has led to streamlining the concept of this genus based on several novel characters. Consequently, an up to date annotated checklist of the Indian species has been compiled. An in depth analysis of the checklist indicated that genus *Andrena* is represented in India by 23 subgenera and 54 species including the three new species described during the current studies. Species of this genus were found to be distributed in the states of Rajasthan, Punjab, Himachal Pradesh, Gujarat, Uttarakhand, West Bengal, Uttar Pradesh, Bihar, Jammu and Kashmir and Delhi.Three species new to science *viz.*, *A.* (*Zonandrena*) *pantnagarensis* sp. nov., *A.* (*Z.*) *dehradunensis* sp. nov. and *A.* (*Euandrena*) *indiaensis* sp.nov. have been described (Meena and Dey 2015).

Subfamily Mutillinae (Family Mutillidae) with 29 genera having 169 species from India was studied. The studies done on the approximately 400 specimens led to several new records viz., *Trogaspidia pilosella* (Magretti), *Pristomutilla pauli* (André) and*Trogaspidia intermedia* (Saussure) from India. The study accomplished numerous new distribution records within India, cataloging of all the known 169 species under 29 genera with synonyms, taxonomic accounts of 31 species under 12 genera including a new species *Trogaspidia karnatakaensis*, and description of the male of *Karlissaidia sexmaculatus* for the first time. Keys have been formulated for all the genera and species studied, separately for both males and females augmented with valid characters. Present study could thus bridge some of the gaps in the taxonomy of Indian Mutillinae (Chandran and Dey, 2013).

In field surveys, hymenopterous parasitoids and predators belonging to five families were recorded on *Pieris brassicae* Linnaeus. The parasitoids were *Cotesia glomerata* (Braconidae), *Pteromalus puparum* (Pteromalidae) and *Brachymeria lasus* (Chalcididae), and predators *viz.*, *Polistes hebraeus* and *Vespa orientalis* (Vespidae), and *Eumenes dimidiatipennis* and *E. petiolata* (Eumenidae) could be identified. As high as 38% parasitism was recorded due to the various parasitoids, among which *C. glomerata* was the most predominant accounting for 33% parasitism. All the natural enemies recorded during the present studies proved to be new records for Delhi. Further, *C. glomerata* and *P. puparum* as larval and pupal parasitoid respectively of *P. brassicae* and *T. apanteloctena* as hyperparasitoid of *C. glomerata* were recorded for the first time from Iran. Morphometric comparisions of 3 population of *C. glomerata* from 3 regions of 2 countries India and Iran based on 39 important diagnostic characters with the statistical software SAS 9.2 revealed that populations of Umiam, Meghalaya and Urmia, Iran were morphologically closer to each other. A diagnostic key for the important parasitoids, predators and hyperparasitoids associated with *P. brassicae* was formulated (Mirfakhraie and Dey, 2012).

Likewise, redescription of *Diaeretiella rapae***,** collected from nine different parts of India, was undertaken based on 40 quantitative characters including two meristic and thirty-eight ratio characters from 167 specimens/11 populations. Based on their coefficient of variation (CV) values, 16 characters were found to be essential for

discriminating different populations. Analyses of measurements showed that, three ratio characters *viz.* length of flagellomere I to width of flagellomere I; width of mesopleuron to length of mesosoma and length of M+Cu to length of 1Cua had heavier principal component weights in the first principal component and served as the main contributing variables in the diagnostic differentiation of the populations. Following cluster analysis two groups were formed: Group I dominated by populations from plains and Group II dominated by populations from higher altitude. Two parasitoids, *viz*., *D. rapae* and *Aphidius colemani* Viereck, along with a hyperparasitoid *Pachyneuron aphidivora* were recordedon *Lipaphis erysimi* on mustard (*Brassica juncea* variety Pusa bold) (Pramanik *et al.,* 2012).

White grubs are the serious pests of several economic crops and ubiquitous in nature, among which the genus *Maladera* is one of the largest groups consisting of more than 500 described species. Twelve species of genus *Maladera* viz., *M. insanabilis*, *M. iridescens, M. fumosa*, *M. burmeisteri*, *M. discrepens*, *M. cardoni*, *M. simlana*, *M. nilgirensis*, *M. atratula*, *M. affinis, M. indica*, 1850)and *M. coxalis*, from India have been redescribed and diagnostic keys for their identification have been developed. The characters such as clypeal margin, punctuation and bristles; length of antennal club; scutellar punctuations and bristles; length of superior meta-tibial spurs in relation to first metatarsomere; male genitalia were found to be distinct and important in *Maladera* species identification.

In another study, 10 species *viz.*, *Gynaecoserica pellecta*, *Leuroserica lateralis*, *Neoserica flavoviridis*, *Oxyserica pygidialis*, *Trichoserica umbrinella*, *Microserica viridicollis*, *Microserica affinis*, *Microserica marginata*, *Microserica interrogator* and *Microserica* sp. indet., were studied and diagnostic keys were developed. The selected species represented six genera viz., *Gynaecoserica, Leuroserica, Microserica, Neoserica, Oxyserica* and *Trichoserica*. The major distinguishing characters included length of the lamellate club to rest of the segments, number of segments in lamellate club, pubescence, punctation and shape of clypeus, bristles on elytra and punctation on pygidium. The important characters used for delineation of *Microserica* species included number of antennal segments, anterior and posterior angles of pronotum with distribution of bristles. In addition, descriptions of male genitalic characters have also been provided (Dileep Kumar, 2015).

Likewise, white grub species associated with groundnut and potato ecosystems were investigated for the intraspecific variations among the different populations of *Holotrichia* spp. Studies on intraspecific variations were carried out in the predominant species, *H. serrata* and *H. consanguinea,* by considering three geographical populations from India. Univariate, multivariate and discriminant function analysis based on morphological characters (18) and morphometric characters (23 in males and 19 in females) revealed significant differences among three populations of both the species. The principle component analysis showed that characters such as total body length, length of antennal and tarsal segments, elytra length, pronotum width and clypeus width accounted for nearly 50% variation in *H. serrata,* while 14 characters accounted for 43-64% variation in males and females, respectively, in *H. consanguinea.* The discriminant function analysis confirmed the worthiness of selected characters in differentiating the three populations of both the species.

Biosystematic study was undertaken on the leafhoppers associated with mango in different parts of the country. Consequently, *Amrasca splendens Ghauri*, *Amritodus atkinsoni (Lethierry)*, *A. brevistylus* Viraktamath, Busoniomimus manjunathi Viraktamath & Viraktamath, Idioscopus anasuyae Viraktamath and Viraktamath, *I. clypealis* (Lethierry), *I. decoratus* Viraktamath, *I. dworakowskae* Viraktamath, *I. jayashriae* Viraktamath & Viraktamath, *I. nagpurensis* (Pruthi), *I. nitidulus* (Walker) and *I. spectabilis* Viraktamath were described. A new species belonging to the genus Idioscopus was described from Nauni, Himachal Pradesh. Emphasis was given to male and female genitalia variation. Line diagrams and in depth photography were included for description. Checklist and diagnostic keys were developed for easy identification of the species *(Srinivasa, 2015).*

Similarly, Genus *Maiestas* of leaf hoppers belonging to subfamily Deltocephalinae was investgated. Surveys were conducted in various parts of country for the collection. Redescription / description of *Maiestas albomaculatus, M. belonus, M. dorsalis, M. fletcheri, M. hospes, M. intermedius, M. krameri, M. maculatus, M. pruthii, M. subviridis, M. systenos, M. tareni, M. truncatus, M. veinatus* and *M. xanthocephalus* was undertaken. Emphasis was given to

male genitalia variation. Line diagrams and in depth photography were included for description. Checklist and diagnostic keys were developed for easy identification. The molecular characterization was done using mitochondrial genes (Mt COI) for certain species. The morphological and molecular characterization data was integrated accordingly so that the species identity could be confirmed and established.

Insect Microbe Interaction

Gut bacterial association was studied in two populations of whitefly, *Bemisia tabaci* (Gennadius), Asia IIi and Asia I collected from Delhi and Andhra Pradesh, respectively. A total of 17 genera with 32 species were identified from the two populations. Among these, six genera viz., *Bacillus*, *Lysinibacillus*, *Kocuria*, *Acinetobacter*, *Pseudomonas* and *Staphylococcus* were found to be common in both the populations. *Bacillus* in Asia IIi, and *Bacillus* and *Lysinibacillus* in Asia I were associated with all the developmental stages revealing their importance. *Bacillus* was thus predominant in both the populations. Phylogenetic analysis of 16S rDNA sequences of isolated gut bacteria showed that few of these bacterial strains formed separate cluster when compared to their respective counterparts from other parts of the world. Alkaline phophatases (ALP) activity was significantly higher in first two instars in Asia IIi than Asia I population (Fig. 1), perhaps indicating relatively high bacterial diversity in the former (Poddar and Subramanian, 2015).

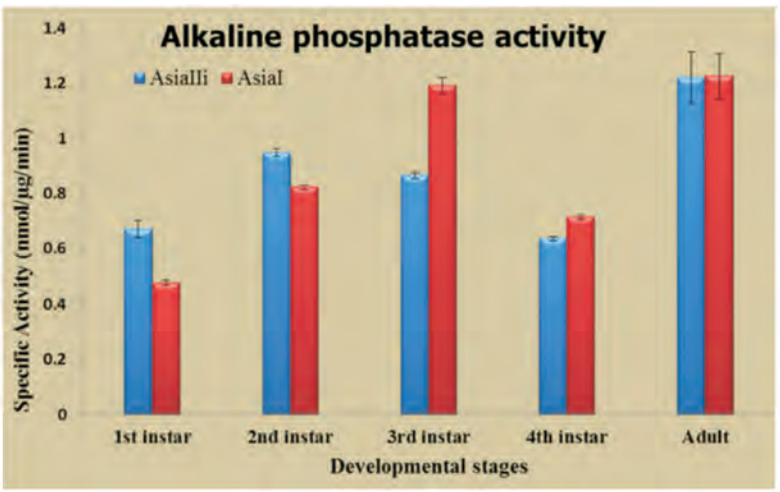

Fig. 1: Alkaline phosphatase activity in white fly populations

Likewise, bacteria were characterized from the mid gut of laboratory and field adult flies of *Bactrocera zonata* (Saunders) using the culture-dependent and 16S rRNA gene sequence analysis. Nine bacteria were identified as members of genera *Bacillus, Enterobacter, Klebsiella* and *Stenotrophomonas*. Bacterial diversity in the gut of the fruit fly adults differed between the laboratory and field flies but *Stentrophomonas maltophilia*, *Enterobacter cloacae* and *Klebsiella pneumoniae* were present in both populations. Among bacteria, *E. cloacae* and *K. pneumoniae* were more attractive to the adults of the fruit fly as compared to *S. maltophilia*, *E. asburiae*, *B. subtilis* and *B. cereus*. Out of seven antibiotics screened against 10 bacterial cultures, kanamycin, streptomycin, chloramphenicol and tetracycline proved to be highly sensitive. Incorporation of the antibiotics in maggot diet @ 5 to 20 ppm did not exhibit any negative effect on survival, development and fecundity of the fruit fly. Information so generated would help in development of improved management strategies against the pest.

Against neonates of red flour beetle, *Tribolium castaneum* (Herbst), out of 51 native *Bacillus thuringiensis (B.t.)* isolates collected from different habitats, 11 isolates proved more effective than *B.t. tolworthi* @100 µg/g of diet on 7th day, while nine isolates performed better than *B.t. tolworthi* against the adults. The most effective isolate was VKK-GJ-4 (LC_{50}=7.02 µg/g of diet) against neonates followed by VKK-GA-6 (LC_{50}=19.03 µg/g of diet) against adults (Fig. 2). Biochemical and molecular characterization revealed occurrence of *Cry* genes as: *cry3* in four isolates; *cry7*, *cry8* and *cry 9* in two isolates each; *cry11+cry3+ cry* 28 in VKK-GA-3; *cry24* in VKK-GA-7; *cry28+ cry3* in VKK-MGA-3; *cry28+cry8* in VKK-GJ-4. The isolates *viz.,*VKK-GA-6, VKK-GA-7, JK-5, MPII5-1 and VKK-GJ-4 can be utilized for developing bio-insecticides to manage storage pests (Veeranna, 2015).

Similarly, against adult cotton aphid, *Aphis gossypii,* a total of 30 native *Bt* strains along with four reference *Bt* strains were screened for the insecticidal activity in three different forms viz., pre solubilized form (spore crystal complex), solubilized form (pro-toxin form), trypsinized form (toxin form) by feeding bioassays. In trypsinized form, 13 Bt strain were found to be highly toxic (70-100% mortality) followed by pre-solubilized form with 9 strains as highly toxic and solubilized form only with two strains as highly toxic. Out of 12 Bt strains isolated from hemipteran insects, four strains viz., VKK-AC1, VKK-AC2, VKK-BB1 and VKK-BB2 and one strain (VKK-PX1) isolated from a lepidopteran insect showed consistent mortality in all the three forms. Further, based on LC50 values, VKK-AC2 and VKK-BB1 were found to be most effective strains against adults of *A. gossypii* in pre-solubilized, solubilized as well as in trypsinized form followed by VKK-PX1 and VKK-BB2. Protein level characterization based on the SDS-PAGE gel showed the banding patterns ranging from 20-135 kDa. Further evaluation of these isolates can be carried out for the insecticidal activity against other important insect pests as these isolates possess diverse group of cry genes (Rajashekhar, 2015).

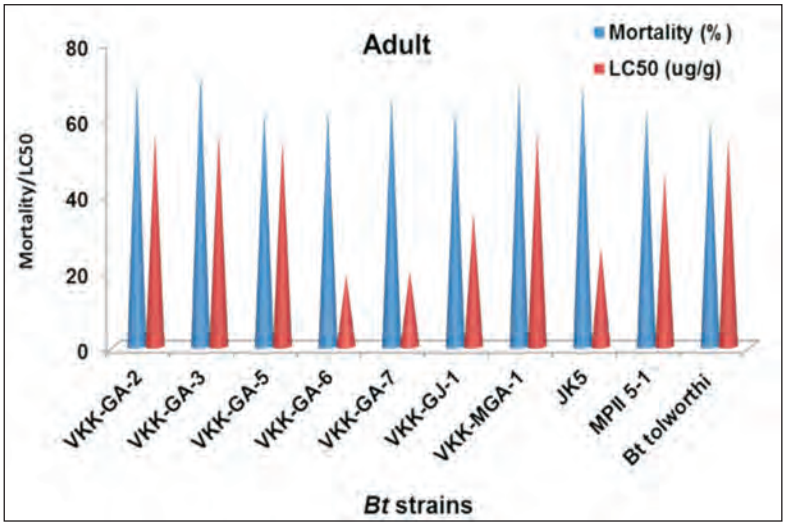

Fig. 2 Efficacy of Bt strains against Tribolium castaneum

Likewise, 40 native *Bt* strains from various habitats were screened for their insecticidal activity against neonates of economically important polyphagous pests, *Spodoptera litura* and *Spodoptera exigua* by feeding assays. LC_{50} values of *Bt* strains varied from 0.87 µg/g of diet (VKK-AC1) to 14.41 µg/g of diet (VKK-OL1) against neonates of *S. litura,* whereas against *S. exigua* LC_{50} values varied from 1.00 µg/g of diet (VKK-AC2) to 40.78 µg/g of diet (VKK-LE1). Six strains (VKK-LO, VKK-SO, VKK-AC2, VKK-BB1, VKK-LE1, and VKK-AG2) were effective against both *S. litura* as well as *S. exigua.* Out of 22 strains, different genes were detected as, *Lep1gene* in 10 strains, *cry2* in VKK-AC1, *cry1C* in VKK-BB1, *cry8* in VKK-GJ4 and GA4, and *cry28* in VKK-GA4, VKK-GJ10 and VKK-GJ4. In the study *cry28* was also found to be effective against lepidopteran insects, besides *cry1, cry2* and *cry8* genes. Further protein level characterization showed the banding pattern ranging from 32 ≥ 236 kDa. Protein band of 105-115 kDa was prominent in most of these strains that may be responsible for toxicity against *Spodoptera*.

Investigations were undertaken on the virus-vector relationship between two begomoviruses *viz.*, Tomato leaf curl New Delhi virus (ToLCNDV) and Mungbean yellow mosaic virus (MYMV) and two vector genotypes, Asia I and Asia II-1 populations of white fly, *Bemisia tabaci*. Distinct differences in transmission efficiency were observed between Asia I and Asia II-1 as Asia I recorded relatively higher transmission efficiencies of 73.3 and 66.6% as compared to 66.6 and 56.6% Asia II-1 for ToLCNDV and MYMV diseases, respectively. Likewise, distinct differences in transmission parameters i.e. acquition access period (AAP) and inoculation access period (IAP) were consistently observed between Asia I and Asia II-1 for both the viruses. The enzyme kinetics of alkaline phosphatase (ALP), esterase, lipase and α-amylase were also investigated. Asia II-1 was found to have significantly higher reaction rate for ALP with Vmax as 1.338±0.141 nmol/µl/min) compared to 1.021± 0.044 nmol/µl/min in Asia I. However, the substrate affinity for this enzyme was more in Asia I as exemplified by lower Km value (0.1826±.036 nmol) compared to the Km value in Asia II-1 (0.394 ± 0.1247). Asia I was found to have significantly higher enzyme velocity for esterase, lipase and α-amylase in comparison to Asia II-1 (Anokhe, 2015).

Eight isolates of *Beauveria bassiana* and *Metarhizium anisopliae* obtained from IARI and National Centre for Integrated Pest Management (NCIPM), New Delhi were tested against fruit fly, *Bactrocera dorsalis* (Hendel). ITCC No. 6628, ITCC No. 6645 and B. NCIPM were found to be pathogenic to adult fruit flies with 100% mortality in 4-6 days and LC_{50} value of $2.5x10^5$,$1.2x10^9$ and $7.5x10^6$ conidia/ml, respectively. The isolate B. NCIPM proved relatively safe to *Coccinella septumpunctata.* Efficacy of commercial myco-insecticides, Bio-power®, Bio-magic® and Bio-catch® against adults of fruitflyvaried from 26.6- 46.6%. The isolate ITCC No. 6645 exhibited dose dependent mortality against third instar maggots with LC_{50} as $9x10^9$ conidia/ml. Isolate ITCC No. 6628, the most effective isolate with the least LC_{50},was mass produced on sorghum grains and 18 formulations based on talcum powder and pyrophylite were tested for viability of the fungus. Only three formulations based on pyrophylite showed germination above 90%, which were subsequently bioassayed against the pest (Elbashir, 2014).

Insecticide Resistance Management

Resistance in the pink bollworm, *Pectinophora gossypiella* (Saunders) to *Bacillus thuringiensis* Cry toxins was studied in populations collected from different locations in India. Screening of *P. gossypiella* populations with Cry2Ab showed 9-fold difference between the most susceptible (Bharuch Non-*Bt*) and the resistant (Jalgaon) population. The F_1 progeny obtained from the mating of resistant and susceptible parent showed autosomal inheritance of Cry2Ab resistance. The heritability (*h*) value for F_1 crosses depicted the semi-dominant inheritance of Cry2Ab resistance. The number of alleles responsible for conferring the Cry2Ab resistance in *P. gossypiella* were found to be more than one. The midgut receptor enzyme alkaline phosphatase, which acts as scaffold for insertion of toxin in the midgut membrane was found to be associated with Cry2Ab resistance in *P. gossypiella*. The alkaline phosphatase activity was significantly higher in the final instar larvae treated with Cry2Ab and resistant parent also showed higher alkaline phosphatase activity which seemed to inherit in F_1 progeny (Malthankar, 2015).

Studies on molecular markers associated with Bt resistance in *Helicoverpa armigera* (Hübner) was undertaken with the field-collected populations from Bangalore, Dharwad, Raichur and New Delhi. Out of 146 isofemale lines created, 42 isofemale lines were used for Cry1Ac resistant alleles and 65 iso-female lines, separately for screening susceptibility. The expected Bt resistance allele frequency was found to be 0.085 with 95% confidence interval. The mode of inheritance of Cry1Ac resistance was autosomal and was inherited as a semi-dominant trait. The reciprocal genetic crosses between the resistant, DNDT-R and the susceptible strain, M4-S showed dominance value (h) between 0.58 and 0.68, and 231.33 fold resistance was observed based on LC50 value. The alkaline phosphatase activity of the resistant and susceptible parents ranged from 93.78 to 123.93 µM/min/µg of protein and 129.61 to 185.89 µM/min/µg of protein, respectively. The study would help in monitoring Bt resistance in the pest (Kennedy *et al.,* 2015).

The tobacco caterpillar, *Spodoptera litura* (Fabricius) larvae collected from Delhi, Sonepat and Varanasi from cauliflower were bioassayed topically with profenophos, quinalphos and cypermethrin. The results revealed that the Delhi and Varanasi populations showed the least susceptibility to profenophos, whereas Sonepat population showed least susceptibility to cypermethrin. The significant variations in the activity of detoxification enzymes *viz.*, carboxylesterase (CarE), glutathione S-transferase (GST), cytochrome P450 monooxygenase and acetyl cholinesterase (AChE) were observed among the populations. Enhanced level of acetylcholinesterase activity was observed in Sonepat and Delhi populations which showed better response to profenophos and quinalphos. Host plants such as castor, cauliflower and soybean also influenced the susceptibility of larvae to insectcides and activity of different detoxifying enzymes. All the three populations were found to be more susceptible to chlorantraniliprole, followed by emamectin benzoate and indoxacarb as compared to other insecticides. Thus, due to existing variation in the pesticide susceptibility of *S. litura*, novel insecticide could be the potential candidates than conventional insecticide in near future (Karrupaiah and Srivastava, 2013).

During two years, populations from Sriganganagar, Ludhiana and Amravati exhibited higher tolerance level to all three neonicotinoids, imidacloprid, acetamiprid and thiamethoxam, whereas Khandwa poulation showed moderate tolerance followed by New Delhi population. A close correlation between cytochrome P450 enzyme and neonicotinoid susceptibility was observed as elevated levels of cytochrome P450 existed in Sriganganagar (494.20 and 503.21 nmol/mg of protein), Ludhiana (465.48 and 482.02 nmol/mg) and Amravati (595.19 and 669.27 nmol/mg) population during two years (Fig. 3). On the other hand, Khandwa (393.70 and 404.16 nmol/mg) and New Delhi (289.26 and 360.50 nmol/mg of protein) populations recorded lower activity of cytochrome P450 enzyme. The *B. tabaci* populations reared on three different host plants, tomato, cotton and brinjal had significant variation in the levels of detoxifying enzymes, *viz.*, esterases, glutathione S- transferase and cytochrome P450. Based on LT_{99} data, a diagnostic time was determined to be 20 minutes to separate a susceptible from tolerant population, while 24 hr were needed to cause 99% kill in highly tolerant populations. Time increase beyond 20 min to bring 99% kill may be the first and quick diagnostic symptom for development of a tolerant population. This method can be used to diagnose the development of tolerance to neonicotinoids in *B. tabaci* (Hanskumar, 2016).

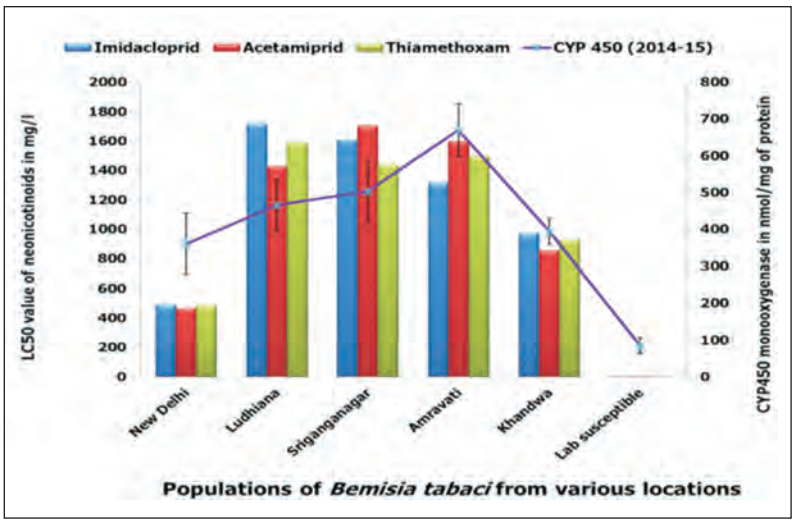

Fig. 3 Efficacy of insectcides against white fly populations

Significant variations in the susceptibility levels of *T. castaneum*to phosphine were observed in populations collected from five states viz., Uttar Pradesh, Punjab, Haryana, Madhya Pradesh and Rajasthan along with a susceptible strain collected from TNAU, Coimbatore. Based on median dose values, populations from Ajmer (1.901 mg/l), Indore, Jaipur, Hapur, Jhansi, Ludhiana Narwana, Malanpur and Sirsa showed higher degree of tolerance; populations from Sangrur, Pratapgarh, Sanwer, Patiala and Bikaner region had moderate level of tolerance, while population from Gohana depicted low level of tolerance when compared with the susceptible strain (0.071 mg/l). Molecular screening using DLD primers revealed the population from Ajmer having the highest resistant allele frequency (83.9%), while the lowest R allele frequency (29.3%) was recorded in the susceptible population. A close correlation between the median lethal values and the resistant allele frequency was observed. Resistance to phosphine in *T. castaneum* was found to be governed by more than one major genes and was inherited as an incompletely recessive trait. The susceptibility levels of the reciprocal F1 hybrids did not show any significant difference indicating that the resistance was inherited in an autosomal manner. The calculated degree of dominance (-0.70 and -0.68) further revealed that the resistance trait is an incompletely recessive one (Ramya, 2015).

Evaluation of Plant Products Against Insect Pests

Hexane, methanol and chloroform extracts of *Andrographis paniculta* were evaluated for insect growth regulatory (IGR) activity against *Spodoptera litura* larvae through diet mixing method and compared with that of Azadirachtin 50%. Five-day old larvae of *S. litura* were fed with different concentration of extracts and Aza mixed with diet and parameters such as larval mortality, larval pupal intermediate, pupal mortality, pupal adult intermediates, abnormal adults and normal adults were observed. It was observed that normal adult did not form under methanol extract and Azadirachtin treatments. Normal adult emergence with hexane and chloroform treatments was reduced compared to control. Hexane and chloroform extract could form only

33.3% normal adults at highest concentration of 0.07%. GI_{50} for Hexane and chloroform extracts were 0.014 and 0.026, respectively (Berin *et al.,* 2007).

Likewise, methanol extract of tubers of glorylily, *Gloriosa superb* (Colchicaceae), a well known medicinal plant, was observed to have seven promising fractions against insects. These fractions were found to contain stigmastenone, β-sitosterol, α-amyrin, colchicine, colchicoside and 2-demethyl colchicine. Their antifeedant activity was higher in comparison to azadiracthin against *Spodoptera litura* and also showed maximum larval weight reduction but abnormalities were not dose dependent. Insecticidal activity of various extracts against adults of *Tribolium castaneum* was also observed and repellent activity of various extracts was low to medium against both larval and adults tages (Nebapure *et al.,* 2014).

Millettia pachycarpa, a perennial climbing tree (Leguminoseae), found only in the north-east region, was studied for its bioefficacy against *Spodoptera litura*. The dried leaves were extracted with hexane and methanol. Out of the 151 fractions and a pure compound, lupeol were obtained. The GC-MS studies revealed the fractions to be caryophyllene,1,4,7- cycloundecatriene, alpha-selinene, pentadecanoic acid, hexa decanoic acid, ethyloleate, ethylpalmitate, eicosane, and heptacosane. Crude dichloromethane (DCM) extract contained four bioactive isoflavonoids, identified as millewanin-G, millewani- H, pyranochalcone and diprenyl trihydroxyisoflavone. Antifeedant activity compared through AI50 values and growth inhibitory activity compared though GI50 values revealed that all the fractions were comparable to azadiractin. Among all the extracts and fractions, methanol and butanol extracts showed good antifeedant activity inbothno-choiceandchoice methods, while DCM extracts showed good insect growth inhibitory activity (Arati, 2012).

Antifeedant and growth regulatory effects of Gudmar, *Gymnema sylvestre* was also studied against *Spodoptera litura*. Dry leaves of *G. sylvestre* were subjected to extraction with hexane and methanol**,** partitioned and characterized using NMR and HPLC. Anti feedant activity of two extracts, GS6 and GS9 was higher in comparison to azadiracthin through no-choice test method after 24 hours. Through choice method after 24 hours extracts**,** GS3, GS6, GS8 and GS9 showed better antifeedancy compared to azadiracthin. Formation of deformities like larval-pupal intermediates, pupal-adult intermediates, abnormal pupae and abnormal adults were not dose dependent. Normal adult emergence was reduced compared to control with all extracts. In topical application method most of extracts showed lower GI_{50} value compare to azadirachtin. Through diet feeding method, three extracts GS6, GS8 and GS9 showed high level of larval weight reduction after azdirachtin. The ability of females, developed from larvae fed on treated diets, to produce the next generation, as exemplified by fertility per female data, was highly impaired with all the extracts at highest concentration (Choudhary *et al.,* 2014).

Various extracts dried leaves of *Clerodendron infortunatum*, pure compounds and azadirachtin were evaluated for their antifeedant and insect growth inhibitory activity against *Helicoverpa armigera*. Pure compounds were identified as clerodin, 15-methoxy-14, 15-dihydroclerodin and 15-hydroxy-14,15-dihydroclerodin. Antifeedant activity of CI 4 and pure compounds (CI 5, CI 9 and CI 10) was higher in comparison to azadirachtin through no-choice as well as choice test method after 24 hours. Through topical application maximum larval and pupal weight reduction was recorded with CI 13 followed by CI 4 at the highest dose, but it was less than azadirachtin. Formation of abnormalities was not dose dependent. Normal adult emergence was reduced with all extracts compared to control and it was minimum with CI 4. Through diet feeding method, CI 4 and CI 10 showed good larval weight reduction. The highest pupal mortality occurred with extracts CI 4, CI 6, CI 9 and CI 10. CI 4 and CI 13 showed complete reduction of adult emergence at all doses. Thus, extract CI 4 and pure compounds CI 5, CI 9 and CI 10 showed very high antifeedant as well as insect growth inhibitory activity (Abbaszadeh *et al.,* 2012).

The effect of various extracts of seeds of *Caesalpinia cristae*as grain protectant was evaluated against *Callasobruchus chinensis* infesting green gram. Significantly less oviposition was observed in all the treatments compared to untreated and solvent treated control. Amongst the various extracts, methanol extract was more

effective at different concentrations at 30, 60 and 90 days after treatment followed by butanol and hexane extracts. No adult emergence was observed in any of the treatments after 30 and 60 days except in seeds treated with hexane extract of *C. cristae.* However, adult emergence was observed in all the treatments 90 days after treatment except in higher concentration of butanol extract of *C. cristae*. There was no adverse effect of these extracts on the viability of the seeds even after 180 days of treatment (Dhingra *et al.,* 2007).

Similarly, various extracts obtained from *Lantana camara viz.*, LC-1, LC-2, LC-3, LC-4, LC-5 and azadiractin technical and essential oil were evaluated for their bioefficacy against almond moth, *Cadra cautella*. Application of extracts of *L. camara* and azadirachtin against different stages of *C. cautella* showed that toxicity of extract was dose dependent. Matured eggs were more tolerant than young ones against LC-1, LC-2, LC-3 and LC-4 extracts. Extracts LC-3 and LC-4 showed lowest LC_{50} values against egg stage of *C. cautella.* Only extract LC-3 was effective against larval stage of *C. cautella* irrespective of their age, while only LC-3 and LC-4 extracts were effective on adults. Application of extracts of *L. camara* on wheat seeds to control infestation of *C. cautella* resulted in reduction in seed weight loss and seed damage. Fumigant activity of essential oil of *L. camara* against *C. cautella* showed that mortality of larval and adult stage was dose dependent. Essential of *L. camara* was found to be most effective against adult stage compared to egg and larval stage (Gotyal *et al.,* 2010).

Eighty seven germplasm accessions of cereals-rice, maize, barley, wheat and oats were assayed in the laboratory for trypsin and chymotrypsin inhibitory activity using synthetic substrate BApNA and expressed as TIU and CIU, respectively. Among all the accessions evaluated, five barley accessions had high trypsin inhibitory activity (~ 40 TIU/mg protein). Trypsin inhibitory activity was more compared to chymotrypsin inhibitory activity in all the cereals tested. Only one accession of maize (51705) had comparable activity (30.50 TIU) as that of barley. The trypsin inhibitory activity of rice, wheat and oats accessions was below 10 TIU/mg of seed protein. Barley seed is hence a potential source for isolation of protease inhibitors for use in transgenic studies aimed at insect pest management.

Ecological Studies

Abundance and diversity of predator species during different growth stages of rice crop in an untreated field was analysed through Simpson index, Shannon-Wiener index and Berger-Parker index. Fifteen species observed to predate upon rice pests during the crop season included nine spider species, one species each of mirid bug and rove beetle, and two carabid and coccinellid species each. Spiders were dominant during early crop stages, while mirid bug dominated during post-flowering period. Spider and mirid bug populations were positively correlated with planthopper population and these exhibited a sort of population regulatory behaviour. Based both on species richness and equitability, Simpson index and Shannon-Wiener index found the predator community to be the most diverse between 39-51 days after transplanting (DAT) and the least diverse at 79 DAT. Dominance of any single species in the community, as revealed by Berger-Parker index, led to decline in community diversity. Life table analysis of six cohortsof brown planthopper (BPH), *Nilaparvata lugens* (Stal) showed that around 70% of BPH mortality occurred by 2nd nymphal instar, which resulted in type A survivorship curve. Regression coefficients (b) of egg (K_E), 1stnymphal instar (K_{N1}), 2ndnymphal instar (K_{N2}), 3rdnymphal instar (K_{N3}), 4th

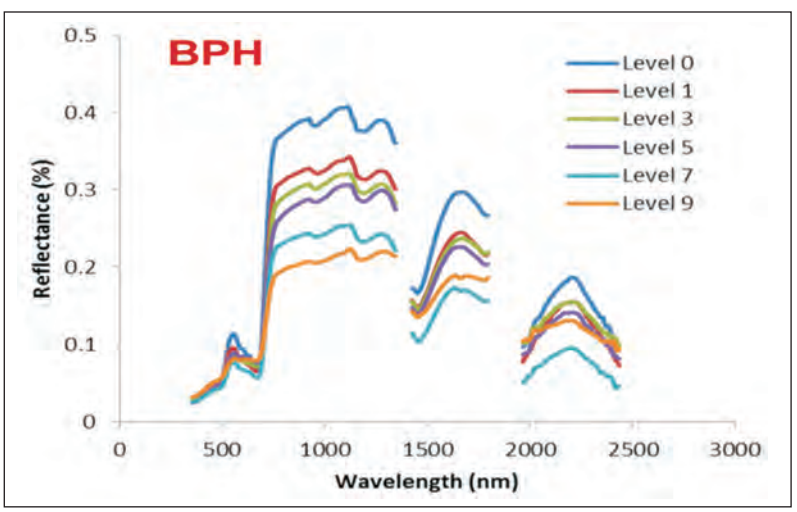

Fig. 4 Rice plant reflectance under differential BPH population

nymphal instar (K_{N4}), and 5thnymphal instar (K_{N5}) were found to be 0.061, -0.034, 0.826, 0.027, 0.0002 and 0.165, respectively. The highest b value (0.826) for 2ndnymphal instar indicated it to be the major contributor to the BPH mortality (Rajesh, 2013).

Spectral signatures were established for leaf folder, *Cnaphalocrosis medinalis* damage on rice with remote sensing using spectroradiometer. Spectral reflectance of infested and un-infested rice crop did not differ in visible range (350 – 700 nm). The major difference in spectral reflectance was found in near infrared (NIR) (780 – 1350 nm) where un-infested crop had higher reflectance compared to infested crop (Fig. 4). Difference between signatures of un-infested and infested crop increased with infestation severity and crop age. Likewise, remote sensing study revealed that BPH infested rice plants had higher reflectance in the visible (VIS) region and lower reflectance in the near infrared (NIR) region as compared to uninfested plants. Sensitive wavelengths in relation to BPH damage detection were found to be 1986, 665, 1792 and 500 nm. Rice plant reflectance corresponding to the sensitive wavelengths was used to develop a multi linear regression model. The model were satisfactorily validated (R^2= 0.94, RMSE =0.79%). These model would facilitate assessment of BPH damage based on rice plant reflectance (Prasannakumar *et al.,* 2013).

Pest-weather models through multi-linear regression were developed between monthly mean BPH light trap catches and monthly mean minimum temperature (Tmin), maximum temperature (Tmax), morning relative humidity (RH_1) and evening relative humidity (RH_2) observed at Maruteru, Andhra Pradesh during 2000-2007 *kharif* seasons. Agro-ecological zoning of rice BPH incidence was done for the state of Andhra Pradesh by interpolating the aforesaid pest-weather model with the help of the 'Geographic Information System (GIS)'. A map of BPH incidence was developed and categorized into four severity classes *viz.*, severe, high, moderate and low for ecological zoning purpose. Comparison between predicted and observed BPH light trap catches at Nellore (*kharif* 2004 and 2005), Ragolu (*kharif* 2003-2007) and Rajendra Nagar (*kharif* 2005 and 2007) evinced very high level of congruence between them, thereby validating the agro-ecological zoning of BPH incidence in Andhra Pradesh (Yadav *et al.,* 2010). Likewise, pest-weather modules were developed for yellow stem borer at Coimbatore (Tamil Nadu), Pattambi (Kerala) and Warangal (Andhra Pradesh.) by regression analysis of stem borer dead heart damage (DH) and weather parameters *viz.* maximum temperature (T_{max}), minimum temperature (T_{min}), morning relative humidity (RH_1) and afternoon relative humidity (RH_2). Agro-ecological zoning for rice stem borer was donefor the states of Tamil Nadu, Kerala and Andhra Pradesh trough GIS by extrapolating the pest-weather relationships developed for Coimbatore, Pattambi and Warangal, respectively. The maps thus prepared showed the areas of equal epidemic potential in relation to stem borer infestation within the states.

Planthoppers, BPH and white backed planthopper (WBPH), *Sogatella furcifera* (Horvath) as well as their predators, spiders and mirid bugs followed aggregated distribution Pusa 1121 rice in field. Sequential sampling plans based on Taylor's distribution parameters (a = 0.398, b = 1.614) and economic injury level (10 hoppers/hill) were formulated for rice planthoppers with and without consideration to predation (Fig. 5). Sequential sampling plans with predator effect suggested need for management measures at higher planthopper population. This would be

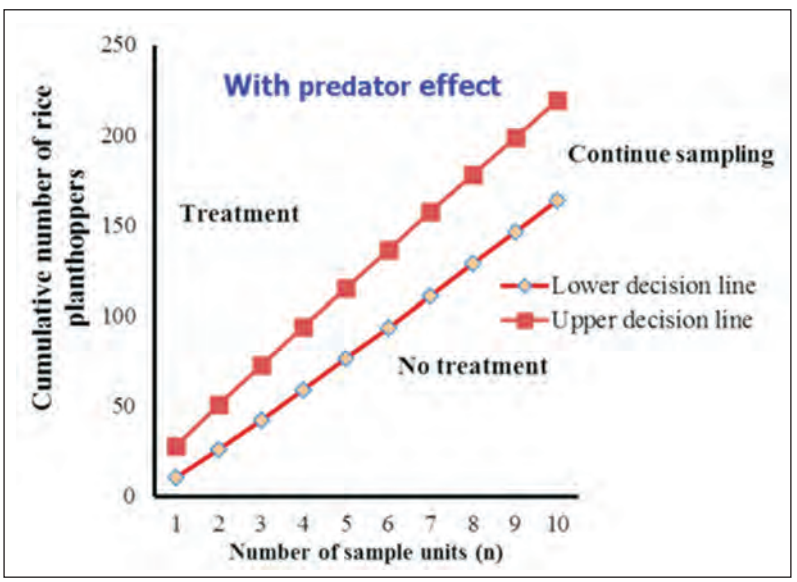

Fig. 5 Sequential sampling plan for rice planthoppers

helpful in avoiding unwarranted pesticide application thereby ensuring natural enemy conservation and favourable benefit- cost to farmers (Rajna and Chander, 2013).

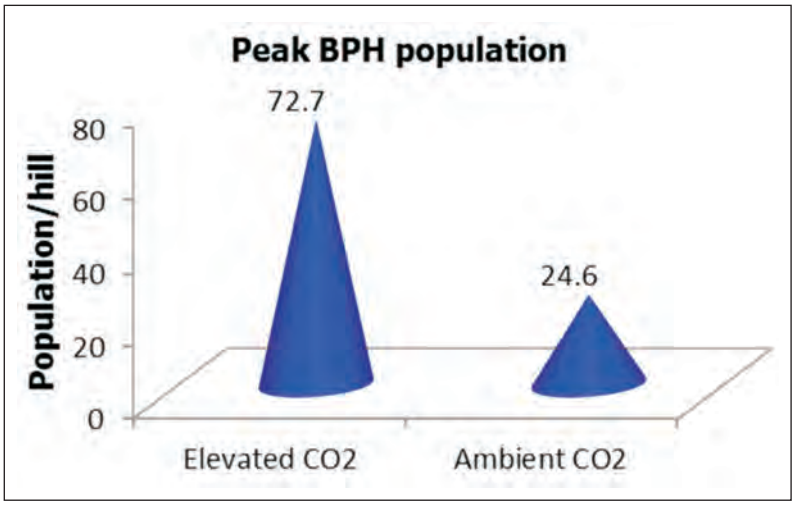

Fig. 6 Impact of elevated CO2 on BPH population

Elevated CO_2(570±25 ppm) exhibited positive effect on BPH on Pusa Basmati 1401 through its enhanced fecundity (29-31.6%) that resulted in more than doubling of its population at peak incidence compared to ambient CO_2 during two years (Fig. 6). The BPH females also excreted more honeydew (68.2-72.3 %) under elevated CO_2 compared to ambient CO_2 during two years. Higher canopy circumference (13.2-16.3 cm) was recorded under elevated CO_2 compared to ambient CO_2 (10.1-12.5 cm) at different phenological crop stages. Elevated CO_2 exhibited nutritive effect on rice crop plants through increase in various yield contributing parameters. However, despite the positive effect of elevated CO_2 on crop, rice suffered higher yield loss due to BPH infestation under elevated CO_2 (29.9-34.9%) compared to ambient CO_2 (17.0-23.1%). Study under Free Air CO_2 Enrichment (FACE) revealed that elevated CO_2 enhanced BPH population irrespective of early, normal and late transplanting, thus resulting in higher yield loss compared to ambient CO_2 under all the transplanting dates. Imidacloprid (0.006%) was found to be the most effective @ 700 l/ha) followed by 600, 500 and 400 l/ha under elevated CO_2. On the other hand, 500-700 l/ha spray volume proved more effective than 400 l/ha under ambient CO_2. The study revealed that more BPH population under elevated CO_2 may demand insecticide application with increased spray volume for effective management of the pest under changing climatic conditions (Gurupirasanna *et al.,* 2016).

InfoCrop, a generic crop–pest simulation model, was used to validate yellow stem borer, *S. incertulas*, damage mechanism on two rice cultivars, Pusa Basmati 1 and Ratna. The model simulated the pest damage appropriately under natural injury as well as under artificial detillering. Validated InfoCrop was used to simulate economic injury levels (EILs) of the stem borer at different rice growth stages for varied control expenditure, incurred on two and threesprays of monocrotophos and one granular application of carbofuran, and for three market prices ofproduce. The simulated EILs were observed to be comparable to empirical injury levels that were previously established. The validated model was also used to devise iso-loss curves that depicted same yield loss for different combinations of the stem borer injury and crop age. Crop simulation model-based EILs and iso-losscurves would be useful in monitoring the stem borer injury and in need assessment for pesticide application, thereby avoiding unnecessary expenditure and environmental contamination.

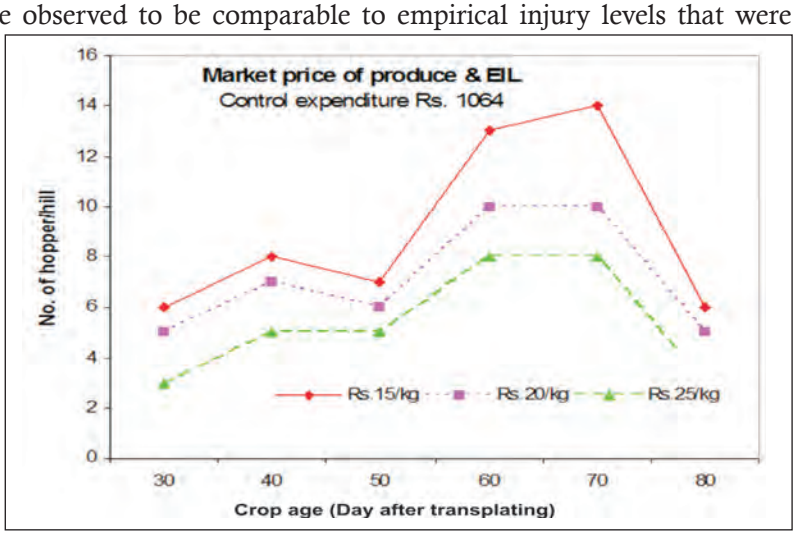

Fig. 7 Simulated economic injury levels of brown planthopper (BPH)

Likewise, rice planthoppers' damage on Pusa Basmati 1 cultivar was simulated with InfoCrop. The model was calibrated and validated with two experimental data sets on planthopper population and rice yield

that were generated through differential insecticide application. Economic injury levels (EILs) of planthoppers were simulated with two control expenditures involving two applications with each of monocrotophos and imidacloprid, and three market prices of Pusa Basamti 1 rice (Fig. 7). Simulated EILs were comparable to earlier established empirical EILs, indicating utility of simulation models for developing location specific EILs that may help in doing away with the use of blanket EILs. Iso-loss curves were also devised through validated model.

Similarly, leaf folder damage on Pusa Basmati1 rice was simulated with InfoCrop through field experiments. Simulated yields and total dry matter in different treatments over the experiments were close to their observed counterparts, indicating an appropriate validation of the leaf folder damage mechanisms onPusa Basmati1 rice. The leaf folder economic injury levels (EILs) were simulated based on control expenditure on application of cartap hydrochloride 50SP and quinalphos 25SC, and market price of Pusa Basamti1. Likewise, iso-loss curves were also formulated to ensure judicious pesticide application (Arya and Chander, 2012).

Based on temperature-dependent development of pink stem borer, *Sesamia inferens* (Walker), thermal constants for eggs, larvae and pupae were determined as 47.6, 700 and 166.7 degree days (DD), respectively through a linear model with corresponding lower development thresholds being 13.8°C, 10.6°C and 12.7°C. Besides, optimum temperature and upper developmental threshold, respectively were found to be 34.6 and 36.2 °C for eggs, 34.5 and 36.4°C for larvae, and 31.7 and 37.0°C for pupae of the pink stem borer through a non-linear model. Based on the thermal requirements, and biotic and abiotic mortality factors, a mechanistic holometabolous population simulation model for *S. inferens* was developed and coupled to InfoCrop-rice model. Validated model was used to simulate the impact of elvated CO_2 and temperature on *S. inferens* population in accordance with four standard special report on emissions socio-economic scenarios, A1, A2, B1 and B2. Simulations revealed that *S. inferens* population might decline to the extent of 5.82-22.8% by 2020 and 19.0-42.7% by 2050 under Delhi conditions. Following decline in pest population, *S. inferens* induced yield losses also revealed a declining trend under changed climate.

Likewise, thermal constants for small nymph (1st-2nd instar), large nymph (3rd–5th instar) and adult of BPH were determined to be 126.6, 140.8 and 161.3 DD, respectively with corresponding development thresholdsof 8.8, 9.5 and 9.6 °C. A thermal constant-based mechanistic hemimetabolous- population model was adapted for BPH and linked with InfoCrop to simulate climate change impact on both the pest population and crop-pest interactions. The model was validated with field data at New Delhi and Aduthurai (Tamil Nadu, India), (R^2=0.96, RMSE=1.87 %). Climate-change-impact assessment through coupled BPH-InfoCrop model, in the light of the projected climate-change scenario for Indian subcontinent, showed a decline of 3.5 and 9.3–14 % in the BPH population by 2020 and 2050, respectively, during the rainy season at New Delhi (Fig. 8), while the pest population exhibited only a small decline of 2.1–3.5 % during the winter at Aduthurai by 2050. However, the study considered the effect of only CO2 and temperature rise on the BPH population and crop yield, and not that of probable changes in feeding rate and adaptive capacity of the pest.

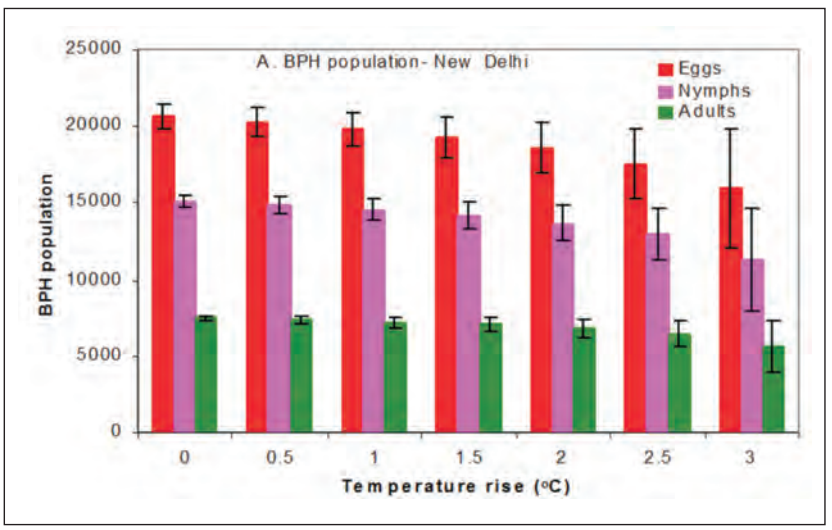

Fig. 8 simulation of impact of temperature rise on BPH population

The effect of different temperature regimes on the papaya mealybug, *Paracoccus marginatus* and its parasitoid *Acerophagous papayae* was investigated. The developmental duration of all developmental stages of melay bug decreased with increase in temperature from 20 to 32°C. Conversely, developmental rate of papaya melaybug increased with increase in temperature. Adult male and female required 344.82 and 332.58 degree-days (DD), respectively, for development with corresponding developmental threshold as and 9.7 and 13.0°C. The highest egg and nymphal survival (%) of mealy bug was recorded at 20°C and 30°C, respectively. Pre-oviposition period was longest (14.8 days) at 20°C and shortest at 30°C (4.6 days). Adult emergence in melaybug parasitoid, *A. papayae* had a positive correlation with temperature increase and female: male sex ratio increased with increase in temperature until 30°C. Parasitism due to *A. papayae* drastically decreased with increase in temperature and no parasitization was found above 32°C. Enzyme activity in melay bugs showed a decreasing trend with declining thermal stress (Laneesha, 2016).

Host Plant Resistance

The extraction of pure fraction of 2, 4-dihydroxy-7-methoxy-1, 4-benzoxazine-3-one (DIMBOA) in a cost-effective manner from 2-day old maize plants grown in dark was standardized. Also, other plant biochemicals such as chlorophyll, protein and sugar were quantified and correlated with leaf injury rating (LIR) due to the stem-borer. The correlation of LIR was found to be maximum with DIMBOA at 1.31 mg/g fresh weight of seedlings of HKI 193-2. Comparatively higher concentration of primary metabolite such as chlorophyll, carotenoid, protein, sugar and moisture was observed in susceptible germplasm. The study concluded that germplasm with high DIMBOA and low total chlorophyll, carotenoid and sugar content may act as a good source of resistance against *C. Partellus* (Chandra, 2012).

Similarly, phenolic acids (ferulic and *p*-coumaric) were explored against *C. partellus* in maize and quantified from 17 maize inbred lines. The *p*-coumaric acid was observed to be predominant compared to the ferulic acid and their concentration ranged from 1.3 to 4.7 mg/g, which varied with plant age. Based on LIR and tunnel length, the inbreds HKI 577 and HKI 323 were identified as least susceptible. Phenolic acid content in maize inbred lines was negatively correlated with the LIR and tunnel length at all the plant ages studied. p-*Coumaric* acid was found to inflict higher larval mortality (41.5 %) than ferulic acid (17.70 %) over control besides reducing the larval and pupal weights, while both the phenolic acids prolonged the development period, reduced successful pupation and delayed adult emergence thus retarding growth and development. Significant negative correlation was found between the cell wall bound phenolic acids and total soluble sugar content. Crude protein and total soluble sugar contents in maize inbreds were observed to be positively correlated with the damage parameters (Gundappa, 2012).

Thirty diverse maize genotypes were evaluated for ovipositional antixenosis to identify genotypes with high level of resistance to spotted stem borer, *Chilo partellus* Swinhoe. In both multi and no-choice tests, genotypes, WNZPBTL 2, CM-500, PFSR 51016/1, WNZPBTL 6, AEB (Y) C5 F 38-1, HK I-PC-5 and CM-202 recorded significantly lower mean number of egg masses per plant. Principal component analysis (PCA) identified the stem tunnelling (ST, %), dead hearts (DH, %) and leaf injury rating (LIR) as most important traits with regard to genotypic response to stem borer resistance. Correlation between morphological traits and damage parameters revealed negative association of seedling vigour with LIR and DH, plant height with LIR, and leaf width with DH. Individuals of mapping population (F_2) developed from WNZPBTL 2, resistant parent and HKI 1352, susceptible parent, were phenotyped for the LIR and classified on 1 to 9 scale. F_2 distribution of LIR indicated monogenic control of spotted stem borer resistance in maize with dominance of resistance over susceptibility. Screening of parents with 200 simple sequence repeat (SSR) markers, identified 37 polymorphic markers across the genome. Chromosomes 1, 5 and 6 were revealed to carry most distinct genome, whereas chromosome 1 was more similar between parents. Only one marker (*bnlg1057*) at map location *bin1.06* showed polymorphism between parents as well as bulks with distinct frequency of resistant and susceptible alleles. One potential simple sequence

repeat marker (*bnlg 1057*) linked with the gene of interest was identified using bulk segregant analysis (Cholla, 2016).

Based on survival and development of spotted stem borer, *Chilo partellus* (Swinhoe), white and yellow kernel maize genotypes were rated as resistant to the pest compared to quality protein maize (QPM) and sweet corn genotypes. Protein content in maize genotypes was lower as compared to stem borer larvae reared on them, while amounts of amino acids were higher in maize seedlings as compared to stem borer larvae. The fatty acid *viz*., cinnamic acid, linolenic acid, behenic acid and lignoceric acid were found present only in maize seedlings, while the fatty acids *viz*., methyl-3-methoxytetradecanoate, palmitoleic acid methyl-14-methylhexadecanoate, methyl-16-methylheptadecanoate, oleic acid and erucic acid were found only in stem borerlarvae in spite of being absent in the maize seedlings, indicating their requirement for different life processes. These could be used as biomarkers for understanding host plant-insect interactions for resistance to the stem borer.

Of 85 green gram accessions evaluated against three species of pulse beetle species,accession KM-11-10 was found to be resistant to *Callosobruchus maculatus* and *C. chinensis*, while PS-16 showed resistance against *C. analis* and *C. chinensis*, based on the growth index (GI). The GI had negative relationship with mean developmental period and significant positive relationship with adult emergence and seed weight loss. Removal of the testa increased the oviposition rate on the decorticated seeds indicated seed coat properties hindered the egg deposition of pulse beetles. Among the physical parameters, the GI of all the three bruchids had positive relationship with 100-seed weight and negative relationship with seed coat hardness. Among biochemical parameters, the GI showed positive relationship with protein and starch content, while it had negative relationship with tannin, α-amylase inhibitor and phenol content of seed. Artificial seed bioassay revealed that increasing proportion of resistant seed powder in mixture of susceptible seed powder adversely affected the bruchid growth and development. The maximum number of SSR bands was amplified by the primer CEDG-305 and it proved to be useful for diversity assessment of the green gram. Results thus showed that green gram accessions KM-11-10 and PS-16, found to be relatively resistant against pulse beetle, can be used as promising donors for developing bruchid resistant varieties (Soumia *et al.,* 2016).

Pest management

The two biorationals *viz.,* emamectin benzoate and Neem baan® (a neem based biopesticide) were tested for their physical compatibility, emulsion stability and bioefficacy against shoot and fruit borer on brinjal and okra. The biorationals were physically compatible and did not produce any phytotoxic symptoms. The combination treatments had phytotonic effect on plant height and number of flowers and shoots, besides reducing the pest damage. Habitat management studies in brinjal with coriander intercrop, reduced leaf hopper population significantly and had highest number of coccinellids in brinjal. Among the other natural enemies, it also attracted *Paederus* sp., *Chrysoperla* sp., *Eristalis* sp and *Brachymeria* sp. (Lekshmi *et al.,* 2011).

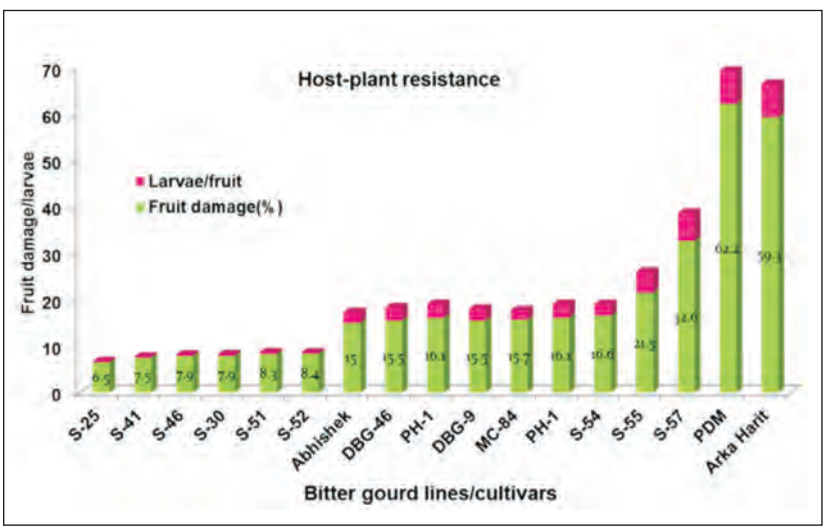

Fig. 9 Reaction of bitter gourd germplasm against melon fruit fly

Out of 50 bitter gourd genotypes screened during spring-summer season against melon fruit fly, *Bactrocera cucurbitae* a very difficult pest to manage, *viz.*, S-25, S-30, S-41, S-46, S-51 and S-53 proved to be promising with less than 10% fruit infestation and very low pest survival as compared to cultivars such as Abhishek, Pant Karela 2 and Nakhra Local with 15-23% fruit damage, and Arka Harit and PDM that suffered around 60% fruit damage (Fig. 9). These can thus be utilized in resistance breeding programme against the pest.

Insecticides, spiromesifen @ 150 g ai/ha and buprofezin @ 250 g ai/ha proved to be more effective against white fly, *Bemisa tabaci* than imidacloprid (35 g ai/ha), acetamiprid (20 g ai/ha), thiamethoxam (50 g ai/ha), bifenthrin (80 g ai/ha), spinosad (75 g ai/ha) and triazophos (250 g ai/ha) on spring-summer crop of bitter gourd. On the other hand, spinosad (75 g ai/ha) + protein autolysate (4%) was most effective against the melon fruit fly on bitter gourd followed by spinosad (75 g ai/ha) alone, dimethoate (200 g ai/ha) + protein autolysate (4%) and malathion (750 g ai/ha) + protein autolysate (4%). Integration of host-plant resistance and bio-rational pesticides would help in managing bitter-gourd pests in an effective and eco-friendly manner (Lekshmi *et al.,* 2014).

Investigations under staggered planting revealed that *Spodoptera litura* preferred to lay more number of eggs on maturing plants, particularly 60-80 days after sowing (DAS), under choice condition as well under no-choice condition. The larval parasitoid, *Cotesia plutellae* on *Plutella xylostella* and *Cotesia glomerata* on *Pieris brassicae* were found to be predominant. Parasitisation and weather parameters together could account for 68.5-77.8 % and 72.7-79.2 % variability in *P. xylostella* and *P. brassicae* incidence, respectively. The intercrops *viz.*, onion, radish and coriander recorded significantly lower insect pests population than cauliflower monocrop. The cauliflower + coriander intercropping system proved superior over cauliflower + radish andcauliflower + onion intercropping systems. The cauliflower + coriander had the highest build-up of natural enemies that resulted in a reduction in pest infestation, thereby resulting in the highest cauliflower head weight (921 to 938g). It is suggested that constant monitoring during critical age of 60-80 DAS rainy season cauliflower and intercropping in cauliflower ecosystem could prove to be useful for better pest management (Mahendran, 2015).

Use of seed treatments in crops such as wheat, maize, chickpea, groundnut and soybean has increased considerably. Laboratory evaluations were thus made with imidacloprid (600FS), fipronil (5FS) and chlorpyriphos (20EC) to assess deleterious effects of seed treatments on seddlings. Phytotoxicity was observed in chlorpyriphos treatments, while imidacloprid exhibited phytotonic effects on the seedlings. Following seedtreatments were found to be safe as no deleterious effects on seedlings were observed: Wheat – imidacloprid@ 3-5 ml/kg seed; fipronil@ 4 and chlorpyriphos @2 ml/kg seeds; Maize - imidacloprid@1.5-5; fipronil @ 5 ml/kg seeds.; Chickpea – imidacloprid@ 5-10; fipronil @ 5-7 and chlorpyriphos @7 ml/kg seeds; Groundnut – imidacloprid@ 3-7; fipronil @ 5 and chlorpyriphos @6 ml/kg seeds; and Soybean – imidacloprid@ 4-6; fipronil @ 3-5 and chlorpyriphos @4 ml/kg seeds.

Toxicity of different CO_2 concentration was evaluated on different stages of *Trogoderma* infesting wheat. The toxicity of CO_2 on different stages of *T. granarium* indicated that the egg stage was the most susceptible followed by adult and pupa. The larvae (fifth instar) were found to be the most tolerant stage. Young eggs, pupae and adults were relatively more tolerant compared to the older ones and their tolerance decreased as age increased. The mortality of the different stage was dose dependent. Exposure period was thus found to be directly proportional to toxicity of CO_2 against *T. granarium*. Activity of CO_2 significantly increased as temperature was increased from 25-35°C, showing a positive relationship between mortality and temperature. Lactate content of treated larvae and pupae showed a linear increase compared to untreated ones. The larvae being the most tolerant stage were able to accumulate more amounts of lactate and still tolerate high doses of CO_2. Thus, for the complete kill of all the stages of *T. granarium*, 55 per cent CO_2 was required with 5 days exposure period irrespective of the evaluated temperatures (Cecily, 2007).

Acknowledgement

I wish to thank all the Faculty Members of the Division of Entomology for extending support and cooperation in preparing the research contributions of M.Sc. and Ph.D. students made during the period of 2007-16.

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Nematology

Tushar Kanti Dutta and Anil Sirohi

Introduction

Nematological developments in India are marked by discoveries of a few important nematode pests such as root-knot nematode (*Meloidogyne* spp.) on tea from Tamil Nadu; Ufra nematode (*Ditylenchus angustus*) on rice from West Bengal; ear-cockle nematode (*Anguina tritici*) on wheat from Punjab and white tip disease of rice *Aphelenchoides besseyi* in Central India during the early part of the 20th century. Phytoparasitic nematodes are the important constraints in profitable crop production and were only realized around the early sixties following discovery of the potato golden cyst nematode (*Globodera rostochiensis)* on potato in the Nilgiris of Tamil Nadu; the cereal cyst nematode (*Heterodera avenae*) on wheat and barley in Rajasthan; the burrowing nematode (*Radopholus similis*) on coconut, pepper and banana in Kerala and so on. The organized work on phytonematodes in India started only during 1960's. The Division of Nematology at IARI was established in the year 1966. Currently the Division is regarded as one of the best referral centre for Nematological research in country and worldwide. The Division executes full-fledged research in basic and applied aspects on nematology and is equipped with high-end laboratory support.

The mandates of the Division are:

- Nematode biodiversity and biosystematics using conventional and molecular approaches.
- Nematode genomics and its utilization in the development of transgenic.
- Exploitation and use of beneficial and entomopathogenic nematodes for the management of insect pests of crops.
- Intensification of researches on rice root-knot nematode, *Meloidogyne graminicola* under rice-wheat cropping system in the Indo-gangetic plains.
- Climate change and its impact on population dynamics of nematodes.
- Nematode problems and their management under protected horticulture and different cropping systems.
- Nano technology and identification of newer molecules for their nematicidal action.

Significant Student's Research Achievements

The research achievements of the Division have been contributed significantly by the students sincerely guided by the experienced faculty members and the Professors of this Division. The research contribution of the students is discussed head-wise and presented below.

Nematode Biosystematics

Plant-parasitic nematodes are a great constraint in the productivity and production of various economically important crops and are estimated to cause damage of almost $173 billion worldwide. Cereal cyst nematode, *Heterodera avenae* group contains at least thirteen valid species. Out of these thirteen species *H. avenae, H. filipjevi, H. spinicauda* and *H. iri* have been reported from India. *H. avenae* are widespread in India and has been reported

from the states of Rajasthan, Haryana, Delhi, Himachal Pradesh, Jammu and Kashmir, Madhya Pradesh, Punjab, Uttarakhand and Uttar Pradesh. Physiological variations in *H. avenae* populations have been reported earlier. Therefore, 3 Indian populations of cereal cyst nematode from Delhi, Kangra (Himachal Pradesh), Leh (Jammu and Kashmir) were studied for their morphological and morphometric characters through light and scanning electron microscopy and sequences of Internal transcribed spacer (ITS)-ribosomal (r) DNA and Cytochrome oxidase c Submit 1 (COI) of DNA. Morphological and morphometric characteristics of Delhi population matched with *Heterodera avenae.* Sequence of ITS region of Delhi population showed more than 98% similarity with *H. avenae* accession no KC152906. Well-developed under bridge was present in the Kangra population. Compared to *H. iri* it had small tail and hyaline region. Based on morphological and morphometric character Kangra population (from Tihri village) was identified as *H. filipjevi*. Leh population was characterized by comparatively short juveniles (489 µm); with small sylet (25 µm), tail (57 µm) and Hyaline region (35 µm); small cysts (L=669 µm) and vulval slit length (5.8 µm). ITS region (626 characters) in this population was much smaller compared mainly due to the deletions observed in the ITS1. Based on morphological, morphometric and molecular evidences Leh population was suggested to be a new variant (species :?) in *H. avenae* complex. COI sequences were highly unpredictable and BLAST results were erratic w.r.t. its taxonomic importance in this nematode. In silico analysis with NEB cutter showed that restriction enzymes AgeI, BcgI, BsrFI and BaaWI; Tth111I, Pf1FI, NciI and BstXI; and AhdI and FauI had unique restriction sites for Kangra, Delhi (IARI) and Leh ITS sequences respectively and hence are useful for differentiating these populations.

Maize cyst nematode (*Heterodera zeae*) is a major nematode pest of economically important monocotyledonous crops, causing considerable yield losses to the tune of millions of dollars annually worldwide. It has been observed that the populations of these nematodes from different countries and within our country differ considerably in their virulence and host preferences. So variability in the con-specific populations of *H. zeae* from Samastipur, Ambala, Kulu, Jammu, Udaipur and Wellington was studied using physiological, morphological and molecular characters. Physiological variations depicted that none of the cultivar/lines of maize showed any resistance against the populations. Vetiver showed susceptibility to Samastipur, Udaipur, Kulu and Wellington populations and moderately resistant reaction to Ambala and Jammu populations. All the six populations were not able to infest sorghum cultivars PC-1 and PC-9. Only Udaipur population was able to produce cysts in the sorghum cultivar PC-23. Based on the reactions of these populations on the test plants, Samastipur and Wellington; Udaipur; Kulu; Ambala and Jammu had been suggested as four groups among the populations studied. Virulence studies using maize cv. Kiran showed that plant growth parameters were significantly reduced at 2 J_2/g of soil for Samastipur, Ambala and Kulu populations, whereas 4 J_2/g of soil was the pathogenic level for Udaipur, Jammu and Wellington populations. Results on the morphometric variations showed that Wellington, Udaipur, Kulu, Ambala, and Jammu populations shared more or less similar characters with the type measurement of the species. Intraspecific variations were, however, observed in Jammu, Ambala and Samastipur populations in many characters in both second stage juveniles and cone top structure. Molecular analysis using PCR-RFLP analysis of Internal Transcribed Space (ITS) region of ribosomal DNA and sequencing showed that that Jammu, Ambala and Samastipur populations were distinct variants of *H. zeae*. They produced maximum number of fragments when the PCR amplified ITS region was digested with eight restriction enzymes. Maximum numbers of fragments were produced in these populations showing heterogeneity, when digested with restriction enzymes *HinF1, Mspl* and *Ava1.* But overall *Alul* and *Rsal* produced maximum number of restriction fragments with no distinction in *Dra 1.* Physiological, morphological and molecular analyses showed Jammu, Ambala and Samastipur populations as variants and may be separate biotypes.

Cyst nematodes belonging to subfamily Heteroderinae are one of the most important group of sedentary plant parasites. Many cyst nematodes including *Heterodera schachtii, H., glycines*, *Globodera* spp. etc. are quarantine pests in most of the countries. Two cyst forming nematodes (KR1 and KR2) from Kangra and one (KU1) from Kullu district were found in a survey conducted in Himachal Pradesh. An effort was made to characterize *Heterodera* spp. isolated from Kangra and Kullu districts of Himachal Pradesh, based on morphological, morphometric and molecular characteristics of the cysts and juveniles. ITS region of KR1 showed 100% similarity with

100% query coverage to the *H. schachtii* from USA. Morphological and morphometric characteristics of the cysts, vulval cone structure, and second stage juveniles of KR1 population showed many similarities with the *H. schachtii* populations reported from other countries. KR1 population was characterized as *H. schachtii* type. Morphological characters of KR2 cysts and their vulval cone structures showed resemblance to the *H. skohensis*. Morphometric characters of *H. skohensis* (KR2 isolate) were also compared with original description of the species and a new range of variations in the morphometric characters was proposed. No data was available on molecular characterization of *H. skohensis*, therefore, ITS region of this species was sequenced for the submission to NCBI. Based on the morphological and morphometric characters of the Cysts and second stage juveniles KU1 population was characterised as morphotype of *H. trifolii.*

Entomopathogenic nematodes belonging to the families *Steinernematidae* and *Heterorhabditidae* are soil dwelling lethal parasites of insects that are used for inundative, augmentative or inoculative biological control. Entomopathogenic nematodes of the genus *Heterorhabditis (*Rhabditida: *Heterorhabditidae*) are symbiotically associated with the bacterium *Photorhabdusluminescens.* Though several indigenous strains of *Heterorhabditis* have been isolated from various localities, few among them have been identified with proper taxonomic studies. A strain of *Heterorhabditis* was isolated from extreme cold desert condition of Leh (Ladakh) Jammu & Kashmir, India. Present investigation were therefore undertaken to identify and describe this strain (named L22) with morphological, morphometric and molecular inputs. Results suggested that *Heterorhabditis* L22 strain was close to *H. bacteriophora.* Bacteria in L22 strain was identified as *Photorhabdusluminescens* ssp. *laumondii*. Presence of this subspecies of bacteria has only been reported in *H. bacteriophora*. Several differences in morphological

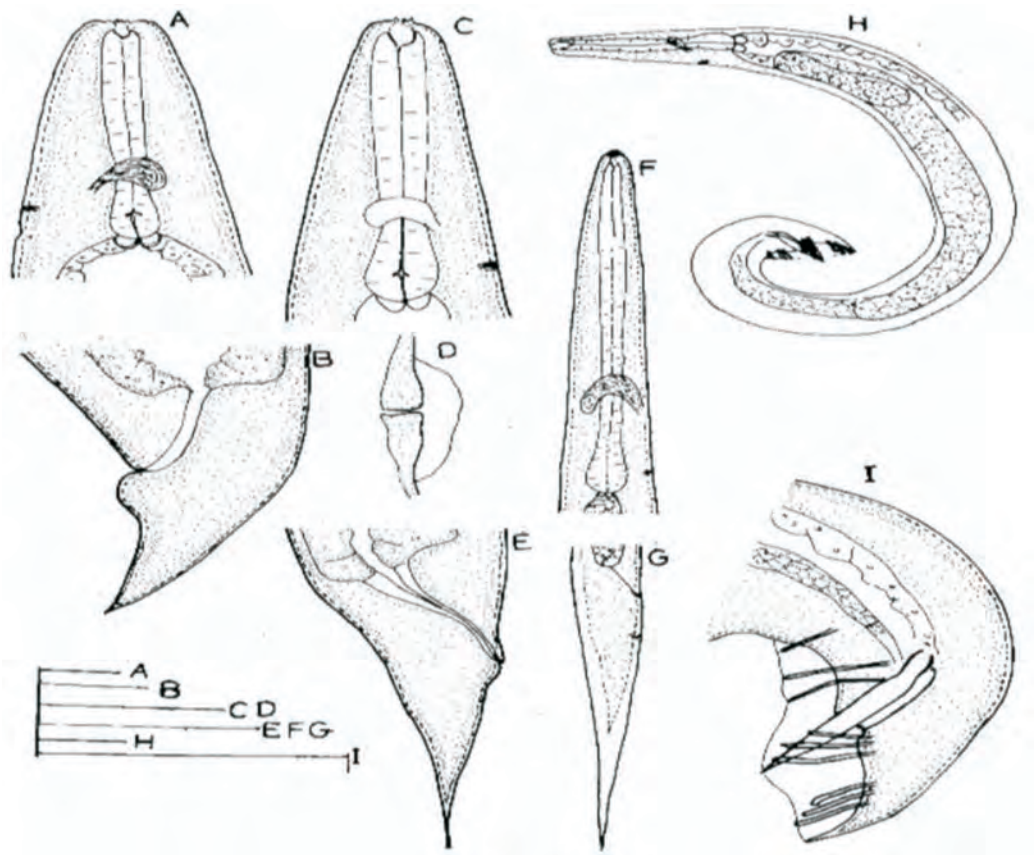

Fig. 1: *Heterorhabditis* sp. population from Leh, Ladakh, India.A,B- First generation hermaphrodite female, C,D,E,H,I- second generation female, F,G- infective juvenile. Scale bar=50µ

and morphometric characters of L22 isolate with *H. bacteriophora* were pointed out (Fig. 1). Comparison of morphometric characters of L22 with 17 other species of *Heterorhabditis* was presented in tabular form. Comparisons of Cox1 and partial rDNA sequences with other species also suggest it to be an unreported species. Based on the morphological and morphometric characters, *Heterorhabditis* L22 strain was found to be a new species of this genus. This uniqueness needs to be further supported with the comparisons of full ITS regions of L22 with other *Heterorhabditis* species.

An investigation was undertaken to characterize the entomopathogenic nematodes isolated from Tamil Nadu and to identify a highly virulent *Steinernema* species for the management of homopteran insect pests. A new species of entomopathogenic nematode, collected from Chennai in Tamil Nadu, India was identified and described as *Steinernema harryin sp.* The new species can be separated from all other species of *Steinernema* by a combination of infective juvenile, male and female characteristics. Its infective juveniles are characterized by having body length of 486 (435-508) µm and lateral field with 6 or 7 incisures. The first generation males are distinguished by the GS% 79 (71-85) and 25 genital papillae. Females of this species are recognized by the presence of well developed, double flapped epiptygmata and protruding vulval lips. Furthermore the new species is characterized by the sequence length of the ITS regions (1014 bp), ITS1 (354 bp), ITS2 (363 bp) and also by the nucleotide composition and pairwise distance between species. The susceptibility of homopteran insects to four indigenous strains of entomophathogenic nematodes was evaluated under laboratory conditions. The results evinced that *S. thermophilum* caused significant mortality of mealybugs (100%) and aphids (83%) within two days at an inoculums level of 500 IJ_S/individual. Against whitefly, *S. riobrave* proved to be effective as it caused 66% mortality within three days at 500 IJ_S. Further, the highly virulent species *S. thermophilum* was tested for its bioefficacy against solenopsis mealybug, *Phenacoccu solenopsis* under field conditions. Nematodes were sprayed in three concentrations (1000, 2000 and 3000 IS_Sml) along with an adjuvant APSA80 (0.033%). An insecticide Chloropyriphos (20% EC) and water alone were included as controls in the study. The results showed that *S. thermophilum* at 3000 IJ_S/ml caused the highest mortality of 34%, followed by insecticidal and 2000 IJ_S/ml treatment with 32% and 29%, respectively. *S. thermophilum* has shown high potential for incorporation into integrated pest management strategies along with other control measures for the control of *P. solenopsis.*

Root lesion nematodes, *Pratylenchus* sp. is a globally distributed nematodes causing severe root damage in many crops at economic level. Taxonomic separation of species in this genus is always difficult for high intra specific variability of characters. An investigation entitled "Morphological and Molecular Characterization of *Pratylenchus* species Occurring at IARI Farm, New Delhi" was undertaken. The Population of *Pratylenchus* species were collected from the rhizosphere of different crops, viz. wheat, maize, cotton, brinjal, pigeon pea, cowpea, chickpea, mango and citrus from IARI farms, New Delhi for detailed morphological and morphometric characterization. The character recorded were body length, number of lip annuli, stylet length, tail length, tail shape, number of tail annuli, vulval position and de Man's ratios. The characterization was further strengthened by molecular data with sequence variability of ITS (Internal Transcribed Spacer) region of ribosomal DNA. Integration of morphological and molecular data gave a concrete base of separation and identification of the two species as *Pratylenchus zeae* and *Pratylenchus thornei* present at IARI farms, New Delhi. Presently the genus comprises 74 valid species globally. This study also provided a check-list and compendium of globally known species of Pratylenchus, including the described species from India and analysis on species variability within the genus. Also the list of synonyms, species inqurendae and nomina nuda have been incorporated. The analysis of different traits among all species within this genus shows the diversity of *Pratylenchus* globally, making way for advances in taxonomic approaches in both conventional and modern techniques in species diagnostics.

A study was undertaken with the objectives of recording the diversity of plant parasitic nematodes in the rhizosphere of the mango and guava at IARI farm. In all, a total of 7 species of plant parasitic nematodes were found in mango and guava orchard at IARI farm. These were *Helicotylenchus indicus, Hemicriconemoides strictathecatus, Rotylenchulus reniformis, Hoplolaimus indicus*, *Mesocriconema sphaerocephala Tylenchorhynchus mashhoodi* and *Tylenchulus semipenetrans*. Amongst the plant parasitic nematodes in mango orchard *H. strictathecatus* was in

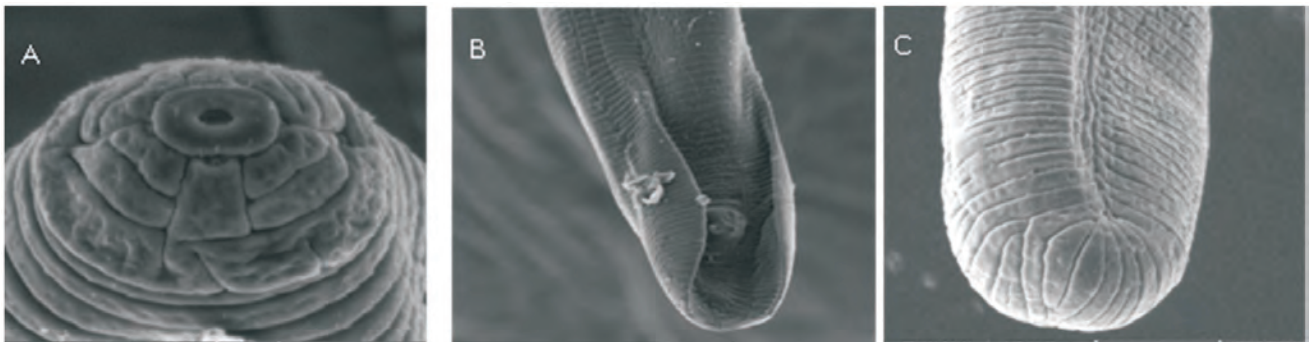

Fig. 2: SEM Photographs of *Hoplolaimus indicus*, A: enface view; B: Male tail; C: Female tail.

high numbers followed by *Helicotylenchus indicus. T. mashhoodi* was in low numbers. *M. Sphaerocephala* was recorded for the first time on guava and mango. In guava, *Helicotylenchus indicus* was having maximum density followed by *H. Strictathecatus* and *Hoplolaimus indicus.* Morphometric characters of different nematode species encountered were also compared with original description of species along with any subsequent study made by any other author on the same species. As a result of this study new range of variations in morphometric characters of *H. Strictathecatus* and *Hoplolaimus indicus* were suggested. Host specific variability in the morphometric characters of nematode species were found in the present study. Scanning electron microscopic observations were also made on indigenous populations of *Hoplolaimus indicus, Helicotylenchus indicus* and *H. strictathecatus.* The identity of citrus nematode, *Tylenchulus semipenetrans* juvenile from the rhizosphere of guava was confirmed based on the ITS region. Further, the similarities in the morphological and morphometric characters of *H. strictathecatus* from this work to that of the populations from Florida also supports proposition by that the introduction of *Hemicriconemoides* sp. to Florida perhaps occurred along with the mango seedlings imported from India during 1889 (Fig 2 and 3).

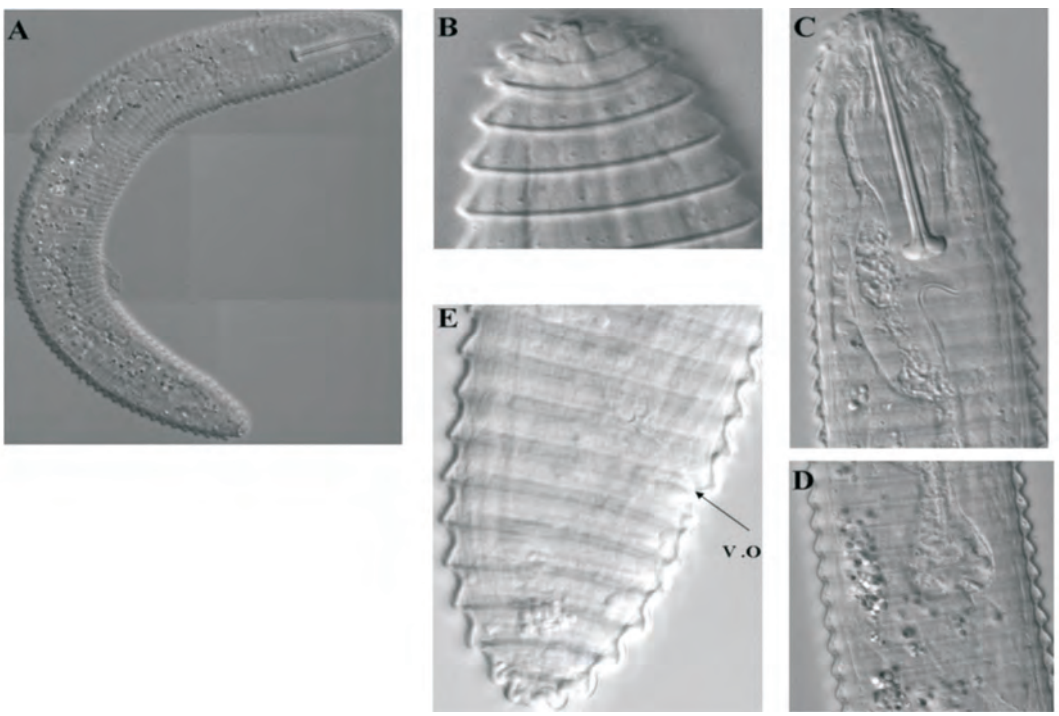

Fig. 3: *Mesocriconema sphaerocephala* (female)- A) Full body length B) Lip region C) Anterior part (lateral view) D) Basal bulb E) posterior part; arrow indicates- vulval opening.

Nematode Management

Neem seed (NS) and neem seed kernel (NSK) granules at 0.2 and 0.4% significantly reduced root-galling and improved growth of tomato plants. NSK was more effective than NS in suppressing nematodes and reducing damage due to *M. incognita* infecting tomato. Both neem products were more toxic to *M. incognita* than to *C. persegnis* followed by *H. indica.* NSK was more toxic than NS to all the three nematodes (*M. incognita, C. persegnis* and *H. indica*). In case of indirect toxicity of neem seed and kernel formulations of *H. indica,* the sand bioassay showed higher mortality in *G. mellonella* due to neem treated *H. indica* than the filter paper bioassays. *H. indica* @ 160 IJ3/g soil resulted in significant reduction in galling and J2 population in soild and improved plant growth characters compared to control and lower inoculum levels, but requirement of such high population densities is viewed as a limitation for practical biocontrol of *M. incognita.* Combination of *H. indica* at 160 IJ3/g soil and NSK @ 1 g/kg soil showed significant reduction in galling and J2 population of *M. incognita* in soil and also improved plant growth characters compared to individual treatments. The results on compatibility of different bioagents and neem seed granules showed that combination of need seed + *T. viride* + *P. fluorescens* was not compatible. Combination of NS + *T. viride* was most effective in reducing *M. incognita* in tomato and cucumber and it was at par with carbofuran @ 1 kg a.i./ha to improving plant growth parameters. Combination of NS + *T. viride* + carbofuran reduced galling and improved plant growth significantly and was the best treatment in field condition. *In vitro* study proved that neem seed granules inhibited *P. fluorescens* and *T. viride* inhibition of *T. viride* was temporary. Also *P. fluorescens* inhibited growth of *T. viride.*

In the backdrop of hazards imposed by chemical control methods, biocontrol can be important component for integrated nematode management in an eco-friendly and target specific way. Rhizobacteria have the potential to become efficient bio agents for the management of plant-parasitic nematodes. With this background the present investigation was undertaken to evaluate the efficacy of rhizobacteria, *Pseudomonas fluorescens,* as biocontrol agent of *Meloidogyne incognita* infecting brinjal (cv. Pusa Shyamla Long). Eight experiments, including six *in vitro* and two *in vivo* were conducted to test the efficacy of the bacteria, *P. fluorescens* on hatching and mortality of *M. incognita;* to evaluate the application methods of the bacteria on plant growth of brinjal and penetration and multiplication of *M. incognita;* to ascertain the shelf life of *P. fluorescens* in talc based formulations and alginate beads and also to test the ability of bacteria to induce systemic resistance in brinjal against the root-knot nematode. Effect of four strains (Pf 1-12, Pf 1-20, Pf 2-1 and Pf-ws) of *P. fluorescens* using cell suspension concentrations (10^6, 10^7, 10^8, 10^9 and 10^{10} cells/ml) and cell-free filtrate concentrations (S, S/4, S/8, S/16 and S/20) on the hatching and mortality of *M. incognita* was investigated *in vitro.* Results indicated that both the bacterial preparations of all the four strains significantly reduced the hatching and mortality at each level of concentration compared to control (distilled water). However, strain Pf 2-1 caused relatively higher inhibition of hatching and mortality. After 72h, lowest hatching of 13.2% and 18.8% was observed in cell suspensions (10^{10} cells/ml) and cell-free filtrate (S), respectively. Similarly, the maximum mortality of 49.8% and 49.4% was observed after 72h in cell suspensions (10^{10} cells/ml) and cell-free filtrate (S), respectively. Inhibition of hatching and mortality were positively correlated with bacterial concentrations and exposure periods. Experiments on the effect of application methods of the bacteria viz. seed bacterization, soil drench, and bare root dip either alone, or seed bacterization in combination with soil drench, or root dip clearly showed that the presence of bacteria enhanced seed germination, root colonization, chlorophyll content and also improved plant growth parameters. A reduction of 47.8% in nematode penetration and 60.7% in nematode population/kg soil was observed in treatment that received seed bacterization, followed by soil drench. Results on the shelf life of bacteria showed that maximum cfu/g (5.5×10^9, 5.9×10^9 and 9.0×10^8) was recorded after 180 days of storage at 5^0 C while lowest cfu/g (5.0×10^4, 8.0×10^4 and 2.0×10^3) was observed at 35^0 C temperature in talc, talc + straw (2%) and alginate beads, respectively. The bacterial population declined markedly with storage time (60 to 180 days) irrespective of storage temperature in all the three carriers. The results on the ability of *P. fluorescens* to induce systemic resistance in brinjal using split root technique indicated that the bacteria was able to reduce nematode penetration and increase growth characters of the brinjal plant through induced systemic resistance as a defensive mechanism against the nematode infection.

Pasteuria penetrans is a Gram positive, endospore forming bacteria with biocontrol potential against root-knot nematode, *Meloidogyne sp.* It suppresses or inhibits the reproductive potential of the nematode pest and reduces the mobility of juveniles (J_2s) at high spore levels (Fig 4). Attempts were made to isolate indigenous populations of *Pasteuria* infecting root-knot species from arid, semiarid, humid and sub humid ecosystems. *Pasteuria* spores detected in the Hisar population of *M. incognita* were multiplied on cowpea, cv Pusa Komal in pouches in an incubator at 25^0C for 32±2 days. The *Pasteuria* infected females varied from 38.4 to 91.3%

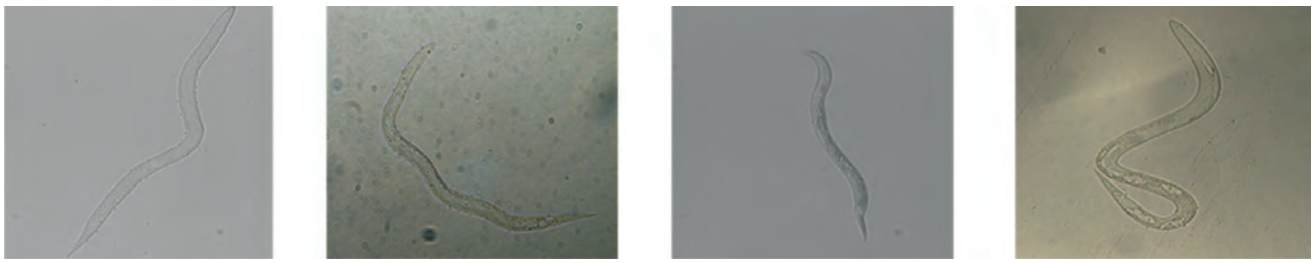

Fig. 4: *Pasteuria* spore encumberance in root-knot nematode.

per plant. A nematode inoculum of 15 per root tip with a spore load of 15 per J_2 resulted in significantly high number of infected females than a nematode inoculum level of 5 or 10 J_2s per root tip with a spore load of 10 per J_2. Inoculation of the plants with *Pasteuria* attached J_2s also resulted in significantly reduced number of egg laying females, compared to untreated control. Spore attachment bioassays performed against four species of *Meloidogyne*, stages of *M. incognita* and other plant parasitic nematode species, indicated host specificity, with the highest number of spores on *Meloidogyne incognita* (average of 6.25) followed by *M. Javanica* 3.90, *M. hapla* 1.10, *M. graminicola* 0.90at a spore density of6.9 X 10^5/spores mL . No other species, *Heterodera cajani, H. avenae*or *Rotylenchulus reniformis* showed spore attachment with the isolate. No developmental stage of *M. incognita* (J_3 or$J_{4)}$ or males showed any attachment of spores on their cuticle. A pre-exposure of spores to high temperature (50^0C, 10 minutes) resulted in significant decline in spore attachment (spores/J2)at 27^0C. Incubation of spores in polyhouse soil too revealed a gradual reduction in spore attachment on nematode juveniles. The preincubation of *Pasteuria* isolate with other bacteria viz. *Bacillus cereus*, *B. subtilis* and *B. pumilus*, resulted in a per cent decline of 97.23, 94.60 and 96.57, in average spore attachment (spores/J2), compared to untreated control.

Okra, *A. esculentus L.* is an important vegetable crop grown throughout the world including India. Presently, it occupies about 4 lakh ha with 4.19 million tones of production and productivity of 10.3 MT/ha in India. Okra production in our country is still dominated by the locally available genotypes/ open-pollinated varieties. Now-a-days okra hybrids are highly preferred by the farmers due to its higher yield compared to varieties. Okra hybrids are grown in 20,000 ha with a share of 5.38% to the total area of the crop. With awareness of advantages for cultivation of F1 hybrids the area is steadily increasing. Plant parasitic nematode is one of the important factor which limiting the production of okra. Reniform nematode, *R. reniformis* is considered as an economically important pest of okra after root-knot nematode. Although some work has been carried out on *R. reniformis* mainly on varieties of okra there is no planned work reported on biology, pathogenecity, host resistance and suitable management practices for reniform nematode especially infecting okra hybrids. Considering the value of the crop and paucity of information on reniform nematode infection on hybrids of this crop which are mainly grown by the farmers for commercial cultivation, it is planned to carry out investigations with the following objectives, 1) To screen the available okra hybrids of identify sources of resistance to reniform nematode, *R. informis* and if resistant found to study DNA polymorphism for resistance. 2) To study the biology and pathogenecity of reniform nematode, *R. reniformis* (race A) on okra hybrids and varieties. 3) To evaluate suitable management options against renifrom nematode, *R. reniformis* infecting okra hybrids and validation of best treatment option under nematode sick plots. Experiment no. 1 was carried out with 208 okra genotypes (73 hybrids, 1 variety, 134 wild-types) to study their reaction against *R. reniformis.* Results indicated that none of the genotypes showed resistance to the nematode. In the present study, we could not to find any resistance in

okra hybrids and wild types against *R. reniformis.* Experiment no. 2 was undertaken to study the life cycle and pathogenecity of *R. reniformis* on okra variety and hybrid. The experiment consisted of okra variety Pusa A4 and hybrid MHOK 10 and laid out in a horizontal block design with three replications. The results indicated that the reniform nematode was able to complete its life cycle in hybrids 3 days earlier than variety and was pathogenic to both hybrid and variety at the lower inoculum level of less than 1 larva per gram of soil. Experiment no. 3 was carried out to find out suitable management option for the management of reniform nematode on okra hybrid, MHOK 10 in three parts viz., first part: soil application of organic amendments (vermicompost @ 25 and 50 g/kg, neem cake @ 5 and 10 g/kg, castor cake @5 and 10 g/kg and FYM @ 50 and 100 g/kg of soil). Second part: Seed soaking with five products (Neemark @ 5%, Neem Seed Kernel Extract (NSKE) @ 10%, phosalone (35 EC) 0.1%, triazophos (40 EC) 0.1% and carbosulfan (25 EC) 0.1% for one hour) and third part: incorporation of three bio-agents (*P. fluorescens, T. viride,* and *G. mossae* @ 125 mg and 250 mg (2.5 and 5 kg/ha) in soil. The results on three different parts of the pot experiment indicated that application of organic amendments as a whole improved plant growth at their higher dosages and inhibited the nematode population significantly. Among them neem cake at 10 g/kg of soil was found much superior. Similar type of results was obtained with seed soaking experiment on all five materials. Carbosulfan (25 EC) at 0.1% performed better among them. Incorporation of bio-agents in soil also gave better growth acceleration and reduced the nematode population as compared to control. The higher dose (250 mg/kg of soil) of *p. fluorescens* was very much effective than others.

The cyanobacterium, *Synechococcus nidulans* is reported to possess nematotoxic effects against *M. incognita.* Preliminary tests revealed that the cell culture or its filtrate caused insignificant immobility in nematodes. In an attempt to detect the toxin principle, the cynobacterium was initially multiplied in BG-11, Bothe's and Fogg's media to identify the medium supporting maximum cell proliferation in minimum time. The maximum cell count ($10.25x10^6$cells/ml) and chlorophyll conent ($76.27x10^{-5}$mg/ml) was observed in Bothe's medium at 30± 2^0C in 15 days, following which the cell culture was harvested, sonicated and tested for toxicity against plant parasitic and beneficial nematodes *in vitro* trails, at S, S/10 and S/100 dilutions for 24, 48, 72, 96 h. The sonicated cell extracts (S) showed an average per cent immobility of 62.9 in *Meloidogyne incognita*, 56.1 in *Heterodera avenae*, 25.1 in *Heterorhabditis indica* and 33.1 in *Cephlobus persegnis*. The per cent revivals ranged from 0.16-10, causing mortality of 59.1% in *M. incognita*, 48.9% in H. avenae and relatively lower mortality of 23.5% in *H. indica* and 30.2% in *C. persegnis*. These trials confirmed the presence of an endotoxin. Air dried cell culture of *S. nidulans*, when subjected to a polar and a non-polar solvent in Soxhlet's apparatus, yielded two fractions, both of which showed enhanced immobility and mortality, compared to the sonicated cell extracts. Methanol fraction was more toxic than the hexane fraction, at all five dilutions (4000, 2000, 1000, 500 and 250 ppm) tested. In 4000ppm methanol fraction, the average per cent immobility was higher in plant-parasitic nematodes (100.0 in *M. incognita* and 88.4 in *H. avenae*), followed by free living bacterivorous nematode (70.7 in C. persegnis) and entomopathogenic nematode (52.1 in *H. indica*), with per cent mortality of 100.0 in *M. incognita,* 84.4 in *H. avenae*, 68.9 in *H. indica* and 50.9 in *C. persegnis.* The LC_{50} values (ppm) were 176.4, 824.2, 3980.0 and 1890.0 in methanol fraction, compared to 1121.0, 1393.0, 7007.0 and 3066.0 in hexane fraction for *M. incognita, H. avanae* and *C. persegnis,* respectively. Chemical analysis of methanol fraction revealed the presence of lipids, carbohydrates and proteins. Thin layer chromatography showed the development of a clear spot from the methanol fraction with an R_f value of 0.37, and a trail of spots from the hexane fraction, with an R_f value of 0.43. The fractions need to be purified further to explore the chemical nature of the nematotoxic biomolecule.

Sonicated and unsonicated heterocystous and non heterocystous cyanobacterial extracts of *Hapalosiphon luteolus, Anabaena variabilis, Calothrix brevissima, Nostoc punctiforme, Tolypothrix nodosa, Scytonema bohmeri, Synechococcus nidulans, Microcoleus vaginatus, Phormidium tenue* and *Microchaete tenera* were tested against second stage juveniles ($J2_S$) of root-knot nematode, *M. incognita.* The extracts of all the ten unsonicated cyanobacteria caused an average immobility of 4.8-22.9% and mortality of 1.5 - 5.6% which increased to 88.2 - 96.5% and

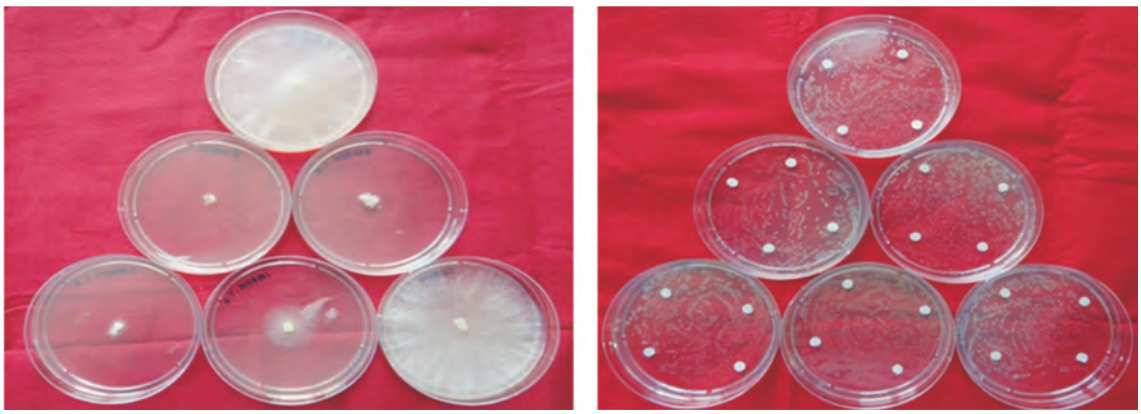

Fig. 5: Effect of carbofuran on growth of *Trichoderma harzianum and Pseudomonas fluorescens*.

4.2- 29.3% on sonication, respectively. The highest per cent mean mortality was observed in *S. nidulans* (29.3) followed by *P. tenue* (26.9), *M. tenera* (25.8), *T. nodosa* (22.3), *M. vaginatus* (21.3), *S. bohmeri* (17.5), *N. punctiforme* (6.5), *A. variabilis* (5.6), *C. brevissima* (5.1), and least in *H. luteolus* (4.2) with an increase in exposure time from 24 to 72h. *S. nidulans* culture collected at weekly intervals for five weeks revealed that sonication of 14 day culture caused the maximum immobility (94.2%) and mortality (29.3%) in *M. incognita* $J2_S$, compared to controls (medium and water). This extract was tested *in vitro* against infective stages and egg hatch of other plant-parasitic nematodes viz. *M. graminicola, Heterodera cajani, H. avenae* and *Rotylenchulus reniformis*. Extracts of sonicated *S. nidulans* caused an average mean immobility in the range of 91.3-98.4% in infective stages of the nematodes with no significant difference with an increase in exposure time from 24 to 72h. The highest per cent mean mortality was observed in *M. graminicola* (31.5) followed by *M. incognita* (29.3), *H. avenae* (20.9) and *R.*

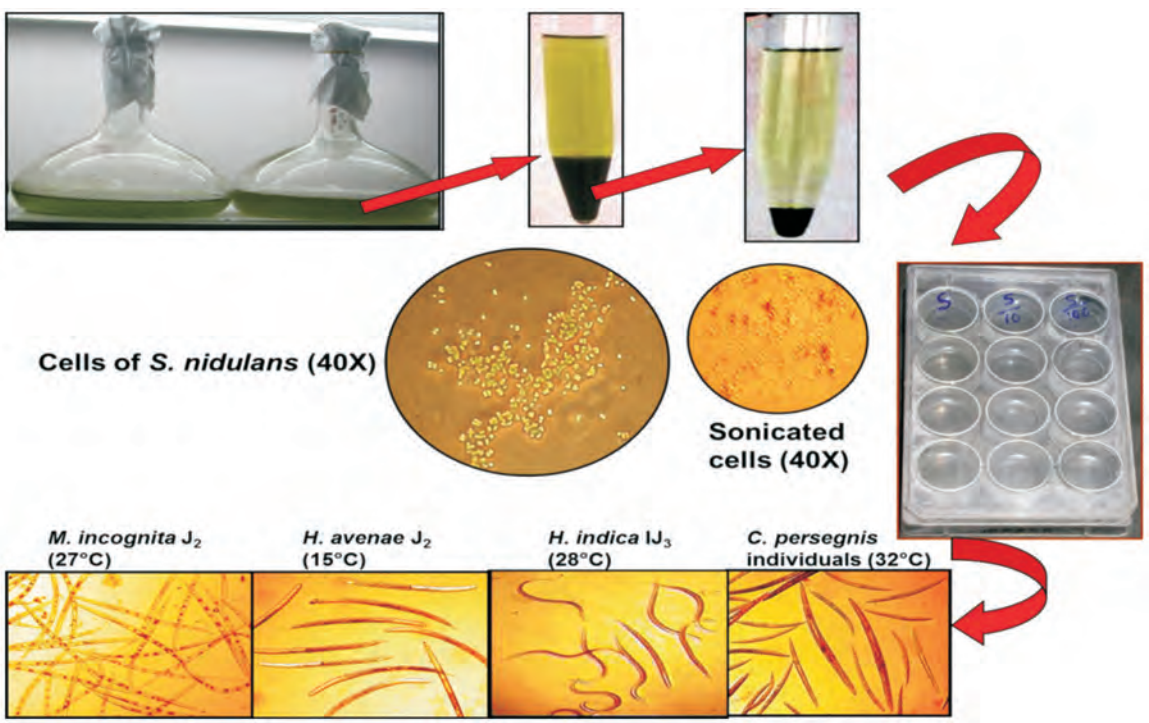

Fig. 6: Effect of sonicated cell cultures of *S. nidulans* on immobility and mortality of plant-parasitic and beneficial nematodes.

reniformis (17.4) and/or *H. cajani* (17.4) with a significant increase with the period of exposure from 24 to 72h. No significant differences in mortality were observed between *M. graminicola* and *M. incognita* and between *H. avenae* and *H. cajani.* The percent between hatch inhibitions over control (water) was maximum in *M. incognita* (94.2), followed by *H. avenae* (91.6), *H. cajani* (72.3) and *M. graminicola* (70.6) and least in *R. reniformis* (58.6). Three carriers' viz. fuller's earth, vermiculite and talc were used for formulating *S. nidulans* and evaluated for their bionematicidal efficacy in microplots. Significant suppression of root galling (84.7%), number of eggmasses (91.7%) per root and nematode multiplication rate (2.2) were observed along with a significant increase in root and shoot length and fresh weight of brinjal, at 0.5% w/v in treatments with fuller's earth formulation, compared to inoculated control. The shelf life of the three formulations evaluated at 15, 25 and 35 ± 1^0C for 6 months revealed significantly higher number of colony forming units of *S. nidulans* in fuller's earth followed by vermiculite and talc. The storage temperature did not have significant effect on shelf life of *S. nidulans.*

Investigations were aimed at selecting a suitable medium for rapid multiplication of *Streptomyces purpeofuscus* MTCC 6473 and to evaluate its cell free filtrates (CFF), polar and non-polar extracts for nematicidal activity against root-knot nematode *Meloidogyne incognita* (J2) *in vitro. S. purpeofuscus* was grown on five culture media, namely nutrient broth, starch casein broth, oatmeal broth, Bennett's broth and Growth Medium-93. The 12 day old culture on Growth Medium-93 showed a significantly higher biomass (95.5mg/l) compared to other media. The culture was collected at two days' interval for nematicidal bioassays. An average per cent immobility of 9.25 and mortality of 4.9 in *M. incognita* $J2_S$, at 24h increased significantly to 63.75 and 59.42 respectively at 96h. The maximum per cent hatch inhibition over control in CFF from 12 day old culture was 41.5 in 1:80 dilution (water: cell free filtrate). The average per cent J2 immobility and mortality was significantly higher in ethyl acetate fraction than in hexane fraction at all five concentrations tested. The crude extract of *S. purpeofuscus* MTCC 6473 was applied as soil drench (1&2% w/v) and root dip treatment of tomato seedling for (30 & 60 minutes) which were compared with carbofuran (1kgai/ha) as nematicide. The treatments were found effective in different aspects. Among the actinomycete treatments, soil drench at 2% w/v caused maximum reduction (83.91%) in nematode invasion, at par with carbofuran application (86.1%). Root dip for 60 minutes resulted in minimum average number of adults/plant (48.75), egg masses/plant (4.25), root-galling (37.75) and J2 population in soil (123.25), compared to 119.3, 10.75, 100.25 and 220.0 per plant, respectively in inoculated control, with increased fresh shoot length and weight, root length and fresh weight. The soil nematode (J2) density was at par in the root dip treatments and significantly lowers than the two drench treatments. The average number of galls/plant and number of egg masses/root that developed in root dip treatment for 60 minutes were at par with carbofuran treated plants. The above observations support the nematodes antagonistic potential of the actinomycete.

Among the sixteen isolates of *Streptomyces* from compost samples collected from different locations, isolate 16 showed the highest antagonistic effects against *M. incognita* juveniles ($J2_S$) and was identified as *S. lavendulae* MTCC 706. Its cell free filtrate collected at 13 days of incubation caused an average immobility of 85.2 and mortality 71.2 against *M. incognita* J2 within 72h with 88.9% hatch inhibition in the standard extract. Ethyl acetate extracts (EAE) of *S. lavendulae* MTCC 706 showed an average per cent immobility and mortality of 82.6 and 80.1, respectively which was significantly more than 57.5 and 45.9 observed in hexane extract (HE). The bioefficacy of the EAE and HE on egg hatch bioassays recorded upto 21 days, too indicated that the nematicidal moieties were more soluble in the polar solvent than in the non polar solvent; an average of 87.8% inhibition was observed in EAE and 41.8 in HE in the standard extract, although inhibition was observed in all 6 dilutions of the extracts. The effects on immobility, mortality and hatch inhibition were positively related to the concentrations tested in all the above treatments. The drench application of *S. lavendulae* MTCC 706 @ 2% v/w (SLT) resulted in less than 50% juvenile invasion of *M. incognita* $J2_S$ in tomato (cv. Pusa Ruby), delayed development of $J4_S$, 50.7% reduction in root galling and 42.7% reduction in eggmass production, compared to control. The reproduction factor was 1.2 in SLT, compared to 2.4 in control in 45 days. SLT was compatible with the nematicide carbofuran, as increase in bioefficacy of SLT was observed with 60% reduction in root galling, 61.5% reduction in eggmass production and a reproduction factor of 1.1 in the combined application

with carbofuran @ 0.5Kg a.i/ha. Replanting of tomato in the pretreated soil revealed a further reduction in root galling (81.3%) eggmass production (80.5%) and reproduction factor (0.6%), indicating establishment of the actinomycete in the soil.

Biopesticdes are an important option for nematode management in an ecofriendly and target specific approach. *Trichoderma harzianum* Rifai and *T. viride* Pers, are beneficial fungi that are present in nearly all soils and other diverse habitats and are very potential biological control agents, known to influence a number of pathogens including nematodes. The biological control ability of *Trichoderma spp* seems to be due to multiple factors, as they have the ability to produce a variety of extracellular lytic enzymes and the production of many secondary metabolites. The metabolites of both *Trichoderma harizianum* and *T. viride* were extracted with acetone, benzene, methanol, and ethyl acetate using soxhlet extractor. The different concentrations (1000, 500, 250 and 125 ppm) of all these extracts were tested separately against second stage juveniles of *Meloidogyne incognita in vitro* by immersion bioassay method. Observations on mortality were recorded after 24, 48 and 72 hours of exposure. Based on the mortality data, LC_{50} values were calculated by probit analysis and LC_{50} values of all these extracts were compared with that of standard pesticide/nematicide (triazophos). The ethyl acetate extract of both the fungal bio-agents were found most effective, while the extracts of *Trichoderma harzianum* was found better to that of *T. viride* irrespective of the solvents used. Methanol was found as the second best solvent whereas acetone and benzene were the least efficient. As the ethyl acetate and methanol extracts of both the fungal bio-agents were found most effective in bio-assay tests, so that these two extracts of both the fungal bio-agents were used for the pot culture experiments. It revealed from pot experiments that the different concentrations of these extracts were found effective against *M. incognita* infecting okra under glass house conditions. 1000 ppm concentrations of the ethyl acetate extracts of both the fungal bio-agents and 1000 ppm concentration of methanol extract of *Trichoderma harzianum* significantly improved various plant growth parameters of okra, whereas 1000 and 500 ppm concentrations of ethyl acetate extracts and 1000 ppm concentrations of methanol extracts of both the fungal bio-agents significantly reduced total number of galls and nematode population in soil.

Management of root-knot nematode with carbofuran @1 & 2 kg a. i./ha, *Trichoderma harzianum* @ 1 & 2% w/w and neem seed oil as seed treatment of mungbean against *M. incognita* was carried out and observation were recorded on the number and function of nodules, chlorophyll content, plant growth and nematode multiplication. It was observed that neem oil treatment had positive effect on nodule functioning and formation. Nutrition status of plant revealed that shoot has better content nutrition such as N, P, K, Ca and Mg, while root has lower level of these elements than the inoculated control plant. Treatments had improved nutrition level of plant. Neem oil was best in giving highest nutrition to both in shoot and root of plant. Plant growth was better with neem oil while carbofuran had minimum plant growth though better than the treated inoculated control plant. Consequently the yield of plant in term of number and weight of pod per plant was translated higher than the other treatments. Nematode reproduction indicated that number of galls, egg mass were least with carbofuran followed by neem oil and bioagent. Similarly, reproduction factor was affected. Visible radiation reflectance pattern of both the experiments showed that reflectance was higher in treatments with low chlorophyll content and *vice versa.*

Investigations were carried out to study the *in-vitro* bioefficacy of *Crotalaria juncea* extracts against *M. graminicola* infesting rice. *In vitro* studies showed that the mortality of $J2_S$ increased with an increase in concentration of methanol and hexane extracts and maximum mortality was observed in both the extracts @ 500 µg/ml at 96 h. There was an improvement in the plant growth parameters of rice with the application of *Crotalaria* extract as a root dip as well as soil drench. The *Crotalaria* extracts provided better initial protection to the rice plants as it was evident from the less penetration of $J2_S$ till day after inoculation (DAI). The numbers of females were least in both the concentrations at 500 µg/ml. The percent reduction in no. of females ranged between 42.99-70.89% and 39.00-63.50% in methanol and hexane extracts respectively compared to 15.8% reduction in carbofuran in root dip treatment and similar trend was observed in case of soil drench treatment. This was perhaps due to the nematoxic principle in extracts. In another experiment, wherein plant parts of *C. juncea* were incorporated into the soil, maximum increase in plant growth parameters rice was observed in treatment (T3) where leaf

and roots of *C. juncea* was incorporated simultaneously during transplanting of rice seedlings. This showed an added advantage of incorporating the plant parts of *C. juncea* into the soil. Although, specific polar and non-polar active principles in *C. juncea* leaves responsible for mortality/immobility remain to be determined, their identification should assist in studying the mechanism involved with enhanced nematode antagonism. Such studies would have value for recommendations (for the use of *C. juncea*) to the farmers who need an effective method for reducing *M. graminicola* populations in rice-wheat cropping system.

The cell free extract (CFF) of eleven native isolates of *Trichoderma virens* from Indian Type Culture Collection (ITCC) were screened against *Meloidogyne incognita*. The CFF isolate of ITCC 7351 caused maximum mortality (100 per cent) in minimum time (60 minutes) of the infective juveniles (J2) as revealed by a sequential time dilution bioassay. Egg masses exposed to the CFF of ITCC caused 89 per cent egg hatch inhibition. No egg parasitisation was observed by the isolate till 15 days of incubation. The ethyl acetate extract (EAE) and butanol extract (BE) of 7day old culture exhibited significant immobility till 1000ppm and mortality in the juveniles till 5000ppm. GCMS analysis of these extracts gave nine compounds, three of which were common with the non-nematicidal isolate ITCC 6122. Column chromatography resulted in 5 compounds in EAE, none of which caused juvenile mortality and eight compounds in BE, one of which caused 78 per cent mortality. Petriplate bioassay on pluronic gel exhibited reduced J2 movement towards tomato (cv Pusa Ruby) roots dipped in CFF of the extract, compared to roots treated with water or media as controls. The isolate was rhizosphere competent as shown by bioefficacy trial in pots using unsterilized soil on tomato cv Pusa Ruby. Significant reduction in root galls, egg mass production, eggs/eggmass and reproduction factor indicate that *T. virens* ITCC 7351 has a potential to be a biocontrol agent for *Meloidogyne* species in tropical regions.

Entomopathogenic Nematology

The compatibility of entomopathogenic nematode (EPN), *Stenernema thermophilum* with different agrochemicals including 12 insecticides, 6 herbicides, 3 fungicides, 7 biopesticides and 8 sufractants/adjuvants, different biogents and organic amendments were evaluated. Among the agrochemicals, all except Hostathion, Dursban and Thiodan in their recommended doses were found compatible. The bacterial bioagents namely *Bacillus thuringiensis* and *Pseudomonas fluorescens* were found to be harmful to the survival of infective juveniles, whereas all the fungal bioagents studied were found to be safe. In contrast, *S. thermophilum* and its symbiotic bacterium *Xenorhabdus indica* could suppress the growth of fungal bioagents namely *Metarhizium anisopliae* and *Beauveria bassiana.* It indicated the incompatibility of *S. thermophilum* with *M. anisopliae* and *B. bassiana* especially when both target the sme host. The organic amendments like vermicompost, farm year manure, bone meal and neem cake were compatible with *S. thermophilum.* The experiment on interspecific interaction of different EPN species namely *S. thermophilum, S. glaseri* and *Heterorhabditis indica* showed dominance of *S. glaceri* at 27^{0}C and *S. thermophilum* at 35^{0}C. But combined infections in all the combinations resulted in reduced progeny production as compared to single infections. The studies on nontarget effects of these three species of EPNs showed no harmful effect on earthworms, whereas ants and spiders were observed to be affected at higher doses. The synergistic interaction of *S. thermophilum* and biopesticidal formulation Tracer (spinosad) was observed in the pot experiment on *Helicoverpa armigera* infecting tomato. This result indicated that the EPNs like *S. thermophilum* in combination with lower doses of safer pesticidal formulations like Spinosad could be a possible alternative to reduce the pesticide load on the environment. The toxic metabolites produced by the symbiotic bacterium *X.indica* were isolated with different solvents like, hexane, ethyl acetate and butanol. As ethyl acetate fraction showed more fungicidal action against *Sclerotium rolfsii,* it was further studied for the chemical nature of biotoxin. The studies revealed the presence of xenocoumacins, nematophins and xenorhabdins in the ethyl acetate extract of *X. indica.*

Experiments were conducted to screen the insecticidal virulence of seven indigenous strains of *Xenorhabdus,* the symbiotic bacteria associated with the populations of entomopathogenic nematode *Steinernema.* All the

nematode populations were able to impart 100% mortality to 4-5th instar of *G. mellonmella* within 48h. Based on the color phenotype of the *Galleri* cadaver, bacterial growth on the NBTA media and upper-threshold temperature the seven *Xenorhabdus* strains were divided into Group A (strains X1, X3 and X5) and Group B (strains X2, X4, X and X7). Strains X1, X3 and X5 were heat tolerant with the upper-threshold temperature of 39^{0}C to 40^{0}C followed by X2, X4 and X7 (32^{0}C). The exception was strain X6 from Group B with an upper threshold temperature of 35^{0}C. The oral insecticidal activity of all seven strains of *Xenorhabdus* was poor against the neonates of *H. armigera* after 24 h and 144h growth of the bacteria in Nutrient Broth. A significantly high mortality of the neonates was observed when the insects were fed with diet mixed with 72h old culture broth. Strain X6 was found to be the most virulent strain causing 60% mortality after 96 h closely followed by the strain X5 which resulted in 53.3% mortality. Out of the seven strains X6 and X7 were classified as insecticidal as they were found to be orally toxic to the neonates of *H. armigera* and were able to impart mortality of 50% and above. Further characterization of the insecticidal activity of the strain X6 showed that the sonicated cells were equally effective in causing (82%) and reducing the weight of the larvae (91.17%) in comparison to healthy larvae. The supernatant possessed poor oral toxicity (38% mortality) but there was significant decline in the weight of the larvae to up to 53.52%, over healthy, which suggested a strong anti-feedant effect.

The pathogenecity of fresh and anhydrobiotic IJ of *Heterorhabditis indica* against *Galleria mellonmela* was evaluated through bioassay test using different dose and time intervals and also the physiological examination was done after drawing of haemolymph from *G. mellonella* larvae infected with both forms of IJ in the laboratory. A comparison of the pathogenicity of the fresh and rehydrated IJ of *H. indica* in causing mortality of *G. mellonella* show that the mortality was generally higher due to the fresh IJ than that due to the rehydrated IJ at the same time interval and inoculums dose. However, these differences were only apparent at lower doses and up to 36 h after inoculation. At higher doses and longer incubation all or most of the *G. mellonella* larvae were killed. The rehydrated IJ of *H. indica* retained their virulence potential and were as infective as the fresh IJ to impart mortality to the IV instar larvae of *G. mellonella.* However, at equivalent inoculums levels and post-inoculation incubation periods, the mortality of *G. mellonella* IJ as well as their progeny production was slightly reduced. This indicated that either some of the *H. indica* IJ died and failed to revive upon rehydration, or they suffered some level of injury or depletion of energy reserves. It would therefore, be advisable that a slightly higher dose of rehydrated anhydrobiotic *H. indica* may be used. The protein content in haemolymph of *G. mellonella* larvae infected with fresh and rehydrated IJ of *H. indica* was lesser compared to healthy larvae. Rehydrated IJ caused relatively lesser decrease of protein content in haemolymph of the insect larvae. Both fresh and rehydrated IJ also caused pronounced decrease in free amino acids as compared to untreated larvae. There was no difference in the quantum of decrease of free amino acids due to infection by fresh and rehydrated IJ. *G. mellonella* larvae infected with both forms of IJ of *H. indica* suffered decrease in the total haemocyte count as compared to non-infected larvae, however, the decrease was relatively greater due to infection by the fresh IJ.

Releasing the activated anhydrobiotic *H. indica* IJ_S in soil and comparing their movement with the fresh IJ_S in the presence and absence of host insect larvae-*Galleria mellonella,* it was found that the nematodes dispersed both vertically and horizontally from the point of application. The tendency to migrate vertically was more evident with a faster rate, than to migrate horizontally. With increase in the distance (2<4<6<8 cm) and time (12<24<36<48 h) the percent vertical and horizontal migration also increased, although rate was fast in case of former than the later. This suggests that the IJ_S follow a positive geotropic movement along with the gravity. Comparison within all the treatments suggests that maximum of 80.7% fresh IJ_S migrated vertically up to 4cm in presence of host insect in 48h, whereas the maximum migration in horizontal plane was recorded up to 61.9% till 2cm in 48h in case of fresh IJ_S. Anhydrobiotic *H. indica* IJ_S aggregated to from large clumps but remained straight and did not coil. Comparative quantification on four biochemical factors reveal that under moisture stress condition at 97% RH when IJ_S enter into partial anhyrobiosis, there was an increase in the accumulation of total sugar (3.0%) and non-reducing sugar (171.9%) while decrease in the levels of reducing sugar (25% and total soluble proteins (52.8%), in comparison to fresh nematodes. There was no detrimental effect on the *Photorhabdus* cells due to anhydrobiotic stress on the IJ_S

as the bacterial symbiont could be isolated on nutrient agar plates. The bacterial retained their virulence as 100% mortality was found in *G. mellonella* larvae injected with bacterial cells.

The production of *H. indica* raised either on P or M form *Photorhabdus* cells was studied with an *in vitro* system. The bacteria proliferated in the media to provide food source for the sterile nematode eggs to replicate. Though, initially the moulting was slow in plates supplemented with M form bacteria, the growth rate of the first generation J1, J2, J3, J4, males and second generation females was statistically at par with the P form cells. Significant difference was observed in the development of hermaphrodites, followed by the subsequent second generation of J1, J2, J3 and J4 stages between 10 to 30 days. The hermaphrodites developed in P form plates were larger in size and produced more number of eggs than those recovered in M form plates. Both, amphimictic and automictic reproduction occurred in the second generation, in both the treatments, between 25 and 35 days. *Endotokia matricida* was also a common feature in both nematode generations. In the control plates without any bacterial supplement, the eggs hatched to J1 and could not develop beyond J2 stage even till 35 days. Pathogenicity of P, M and phase II form variants of *Photorhabdus* was tested by injecting 1, 10, 100, 500 and 1000 cells/ 4th instar *G. mellonella.* Though mortality was relatively faster in P form cells as compared to M form, with respect to all the 5 doses tested, there was no statistical difference between the two. Higher bacterial doses of 100, 500 and 1000 cells/larva were significantly at par in their mortality pattern, whereas, the lower two doses of 1 and 10 cells/ larva were found to be significantly different. Injection with 1 cell/ larva resulted in a maximum of 40% mortality after 42h in both the forms. In comparison to P and M form variant, the P II cell variant resulted in significantly poor mortality.

Out of eight cryoprotectants viz. ethylene glycol (10 and 20%), glycerol (15 and 20%), dimethyl sulfoxide (10 and 20%), glycerol (15%) + methanol (70%) and glycerol (20%) + methanol (70%) and glycerol (20%) + methanol (70%) tested for cryopreservation, for a period of one week, 10% ethylene glycol for *M. incognita* and 20% ethylene glycol for *H. indica* were found to be the most efficient in providing 14.43 and 11.25% survival, respectively. It was followed by 20% DMSO in providing 11.15% survival of *H. indica* and 14.13% survival of *M. incognita.* Uniformly, in all the treatments the dead juveniles of *M. incognita* and *H. indica* were found lying straight and in a few cases they found to be ruptured after removing from liquid nitrogen. The cryo infective juveniles of *M. incognita* and *H. indica* recovered after 14, 28 and 56 days to preservation in liquid nitrogen did not lose their infectivity and were able to penetrate and reproduce on tomato seedlings and *Galleria* larvae, respectively. Effect of vitrification of *M. incognita* eggs did not provide any encouraging results. After thawing only 0.43, 0.50 and 0.40% eggs hatched in 10% ethylene glycol, 20% glycerol and 20% DMSO, respectively, as compared to 51.82% hatching observed in untreated control.

Basic Research

Investigation was conducted to know the differences in host recognition, invasion, development and reproduction of the root-knot nematodes, *Meloidogyne graminicola* and *M. incognita* of rice and tomato (Dutta et al., 2011). Attraction bioassays in Pluronic gel showed clearly that *M. incognita* preferred tomato roots more than rice while *M. graminicola* was attracted more towards rice compared to tomato. By providing two routes to a source of attachment, one long and one short in Y-chamber, nematodes took the most direct route towards their preferred hosts but often took the longer route towards poor hosts. The root exudates which might have regulated the nematode movement towards specific hosts, was collected from tomato and rice, extracted through Solid Phase Extraction in C18 column. GC-MS analysis of the exudates showed presence of some small lipophilic molecules (SLM). These molecules from tomato (ToSLM) and rice (RiSLM) showed motility inhibitive and nematotoxic effects in neat concentration and nematostatic effect in diluted concentration on both *M. incognita* and *M. graminicola.* Western blot analysis with different Polyclonal and Monoclonal antibodies showed major differences among the protein profiles of the two nematode species. Thus evolutionary adaption of these root-knot nematodes to their preferred hosts might have led to variability in their gene/protein profile which could also contribute to their differential behavior outside and inside of the different host crops.

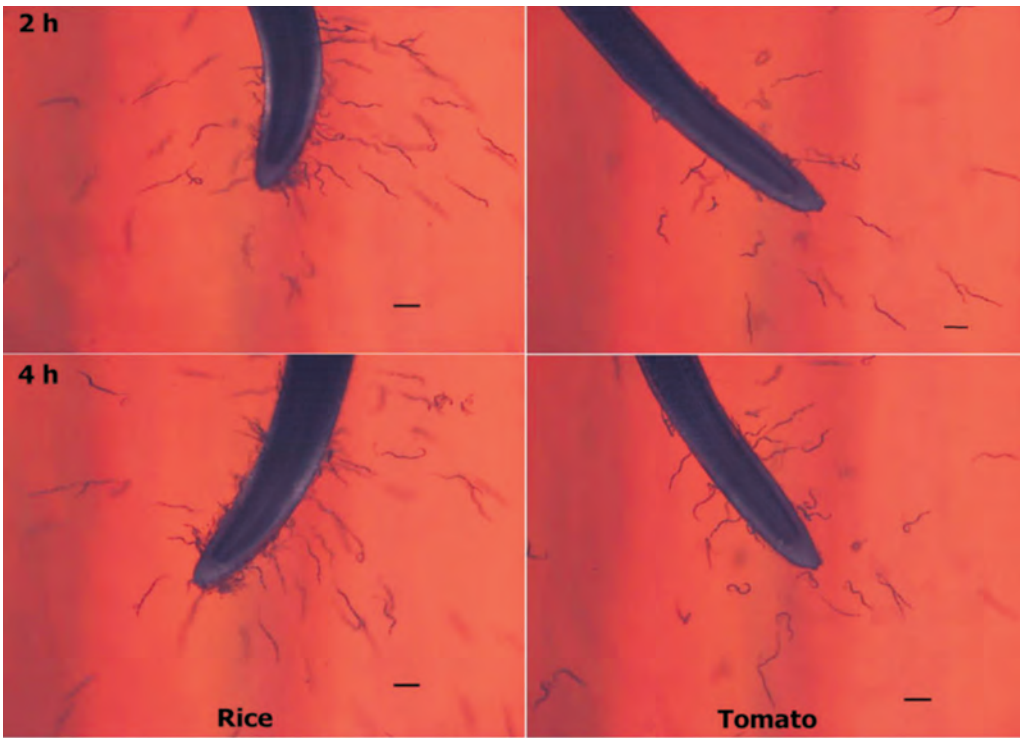

Fig. 7: Attraction of *M. gramainicola* towards the root tip of rice and tomato at different time periods in Pluronic gel medium.

Investigations were carried out to know the effect of different population densities of *Meloidogyne graminicola* on nutrient uptake and physiological parameters of the rice plant and quality of grain and vigour of seed produced (Patil et al, 2013) (Fig 7). The effects of the ammonical and nitrate forms and concentration of nitrogen on attraction and infection of root was also studied. Electrophysiological and ^{15}N nitrate movement studies were carried out to observe changes in cellular nitrate levels in nematode infected rice root. The concentrations of N, P, K, Fe and Zn in rice leaves were significantly reduced and were negatively correlated to the nematode infestation level. The plant height and shoot dry weight were also decreased with increasing nematode inoculation in pots and infection levels in field. The residual contents of N, P, K, Fe and Zn in soil were greater at higher levels of nematode infestation than in nematode-free or lower infestation levels, with a strong positive correlation. The macro- and micronutrients remained unutilized in the soil while the plant suffered severe nutrient deficiencies and growth reduction in the presence of nematode infestation. The reduced photosynthesis of the host plant, chlorophyll content, photosynthetic water use efficiency, transpiration and increased respiration rates were recorded at very different population densities of nematode. Attraction bioassays in X and Y chambers showed that *M. graminicola* preferred rice roots grown in 0.1 mM and 2.85 mM of NO_3 as $Ca(NO_3)_2$ compared to 10 mM nitrate as $Ca(NO_3)_2$ and 2.85 mM NH_4Cl, and the rice seedling supplied with NH_4Cl as a sole N source were found to be much less susceptible to nematode invasion under the very high nematode inoculation rates. Apparently, rice plants grown in NO_3 (0.1, 2.85 and 10 mM NO_3 as $Ca(NO_3)_2$) resulted in an alkaline rhizosphere pH supporting *M. graminicola* infection, while plants grown in NH_4NO_3 and NH_4Cl supplied media resulted in acidic pH. The pH differences at the root may be an important factor influencing the ability of a nematode to both locate and invade the rice root. The change in pH might influence the nematode attraction and repulsion of the nematodes. The $^{15}NO_3$ influx and electrophysiological measurements clearly showed that the root NO_3 transport activity was severely decreased in galled roots infected with nematodes. The results also prove that rice grains produced on plants infected by the nematode, *M. graminicola* were lighter in weight and had poorer nutrient qualities such as amylase and protein contents that would affect consumer interests. Further, if these

grains were used as seed, the germination percentage was lower and the seed vigour was poor compared to the seed obtained from plants grown in *M. graminicola* free soil, harming crop performance and farmers' economic interests. Investigations on physiological parameters of plants and performance of varieties would be vitiated if the roots are infected by nematodes. Similar effects can be expected in case of other crops and root parasitic nematodes.

An investigation was carried out to find the effect of elevated CO_2 levels on root-knot nematode, *Meloidogyne incognita* lifecycle, histopathology, host defense enzymes and pathogenicity on tomato varieties Pusa Ruby and SL-120 in National Phytotron Facility, IARI, New Delhi. Results revealed that the lifecycle decreased by 3.5 and 4 days in Pusa Ruby and SL-120, respectively under elevated CO_2 levels. The CDH values confirmed the role of temperature in determining this duration. Penetration increased by 40.41% and 42.74% at HAI and fecundity decreased by 8.08% and 38.84% in Pusa Ruby and SL-120 respectively under elevated CO_2 level. Histopathological studies confirmed normal giant cell development and early development of nematode under elevated CO_2 level. Peroxidase activity increased by 86.55% & 85.07% and superoxide dismustase activity increased by 74.12% & 57.34% in Pusa Ruby and SL-120 under elevated CO_2 at 3rd DAI. Pathogenicity tests revealed that the minimum damaging threshold level of initial nematode inoculums was at 1000 J_2^S per cent or 2 J_2/cc soil. The rare of nematode multiplication was maximum at 100 J_2^S/plant in both CO_2 levels, however the multiplication rate was less under elevated CO_2 level in both varieties.

In order to adopt resistance breeding approach, 64 germplasm of Asian rice (*Oryza sativa*) were evaluated for resistance against rice root-knot nematode on the basis of root-knot index. Our study showed a large variation in susceptibility and sensitivity to *M. graminicola* infection among the rice cvs./landraces examined. Out of the total 64 rice cvs., seven (Abhishek, Khaja, Super Sugandhamati, Kishori Dehraduni, Gaudeshwari, Tuniaslet, Chima Kamin) had less than 10 galls/plant and of these, Abhishek exhibited a strong resistance response with least number of galls (2 galls/plant) and necrotic browning of roots which is typical of R-gene mediated resistance response. The nematode females in the roots of cv. Abhishek were smaller in size compared to those on the highly susceptible cv. Bangla Patni. The histopathological studies showed that the giant cells collapsed and degenerated before the J2s developed into adults. In order to map the gene governing resistance to root-knot nematode, the cultivar Abhishek was crossed with the susceptible genotype Bangla Patni to generate an F_2 population. A total of 419 STMS markers were used in the parental polymorphism survery, among which 94 markers were found to be polymorphic. Resistant and susceptible bulks with 10 plants each were generated for bulked segregant analysis using 94 identified polymorphic markers. The marker *HvSSR10-21* was identified to be linked with the R-gene at a distance of 18.1 cM with the LOD score value indicates the tight linkage between identified marker and the R-gene against *M. graminicola.* The gene was designated as *Mgl(t).*

Fig. 8: Root morphology of 14-day-old rice seedlings grown in sterile culture on agar in Petri dishes supplied with three different forms of N supply and 1% sucrose and infected with *Meloidogyne graminicola*.

Thermal death kinetics of root-knot nematode *Meloidogyne incognita* was determined using hot-water bath at temperature range between 45-60^{0}C. Minimum time required to kill 100% second stage juvenile ($J2_S$) decreased with the increase in temperature; it was 2 min for 55 ^{0}C treatments. Minimum hatching of root nematode eggs (1.02%) was recorded at 60 ^{0}C for 4 min treatment. Treatment of egg masses and $J2_S$ to three radio frequencies

(RF) *viz.,* 13, 27 and 42 MHz showed that the effect of 27 and 42 MHz on inhibition of the egg hatch and mortality was statistically at par and better than that of 13 MHz treatment. The thermogram of tuberose after RF heating at 27 MHz showed that the rise of temperature was not uniform throughout the bulb tissues. Often the rise of temperature was more in the narrower part (point of emergence of shoot) of the bulbs compared to the broader equatorial region. RF heating also resulted in the higher temperature at the centre of the bulb compared to its peripheral regions. The bulbs of tuberose, treated at 27 MHz (1.2 KW power) for 8 and 10 min showed delayed germination and poor plant growth compared to untreated bulbs. Infected bulbs were treated keeping the same treatments. Egg mass from infected bulbs were isolated and their viability was evaluated by observing egg hatch. Hatching of eggs occurred in the egg masses from all the treatments. Minimum (15%) hatch was recorded with maximum time of RF exposure (i.e. 10 min) to infected bulbs.

Investigations were carried out to study the bioefficacy of controlled-release nano-formulations (CRNF) of Salicylic acid (SA) as foliar spray and seed dresser which induce defense responses in wheat against the cereal cyst nematode, *Heterodera avenae. In vitro* studies showed that the mortality of $J2_S$ increases with increase in concentration of all the three $CRNF_S$ of SA i.e. A, P and S and 100% mortality was observed at 500 µg/ml with all the formulations after 96 h. There was an improvement in the plant growth parameters of wheat with the application of CRNF of SA (foliar as well as seed treatment). The developed CR formulations provided better initial protection to the wheat plants as it was evident from the less penetration of $J2_S$ during 7 DAI till 14 DAI. The percent penetration of $J2_S$ ranged between 0.24 to 0.77%, 0.44 to 0.55% and 0.11 to 0.55% with the foliar application of CRNF of SA: A, P and S respectively compared to control (34.6%). Later on, restricting the further development of $J2_S$ to white females. This was perhaps due to the sustained SA release from these formulations. The above study was confirmed with the changes in biochemical parameters, where in, the data indicated the peroxidase (PO) and phenyl alanine ammonia lyase (PAL) activities enhanced in all the treatments over control at 7 and 14 DAI in both shoots as well as roots. Among the three formulations tested, S @ 125 µg/ml or above was highly effective when applied as foliar spray and as seed dresser respectively. Thus, CRNF of SA (A, P & S) induces tolerance in wheat plants against *H. avenae* and these formulations could be further utilized under microplot/field conditions for the management of *H. avenae* in wheat.

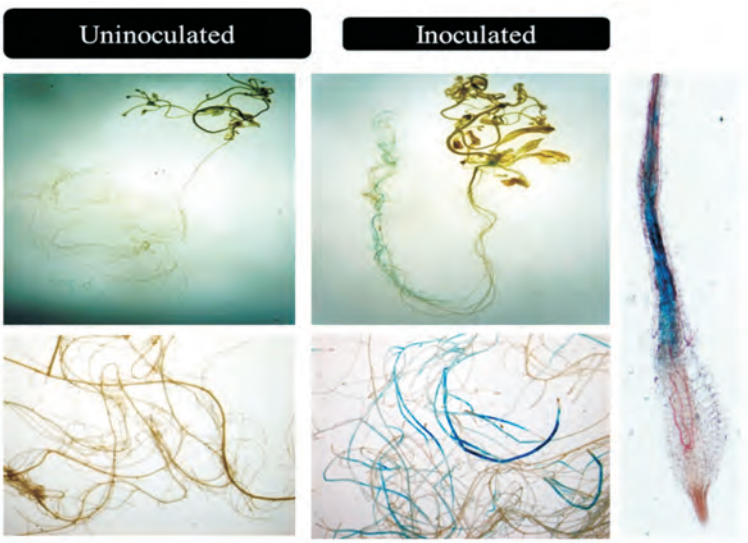

Fig. 9: Histochemical GUS assay in Arabidopsis plants for *M. incognita* responsive promoter

In a study, 33 direct seeded rice (DSR) genotypes of Asian rice (Oryza sativa) were evaluated for resistance against rice root-knot nematode, *M. graminicola* on the basis of root-knot index and multiplication factor. Our study showed a large variation in susceptibility and sensitivity to *M. graminicola* infection among the rice cvs./ landraces examined. Out of the total 33 rice cvs, 6 cultivars had less than 10 galls/plants and of these, cv. NDR-97 exhibited a strong resistant response with least number of galls (1.5 galls/plant) and Multiplication factor (MF) which reflects the overall ability of the nematode to be a successful parasite, varied significantly among all the cvs./land races. Calculated MF value of *M. graminicola* was highest in PB-1121 (39.86) and lowest in NDR- 97 (0.10). Nematode females in the roots of resistant cv. PB-1121. The histopathological studies of root galls of susceptible and resistant plant showed significant differences in the development of females as well as giant cells and egg production. Thus, the fecundity of the nematode was suppressed in the resistant cv. In susceptible plants

cv. PB-1121, the presence of well-developed group of abnormally enlarge cells (giant cells) was common in vascular region whereas, in NDR-97 female development was poor. In order to gain insight of differential response of susceptible and resistant rice cultivars upon root knot nematode, *M. graminicola* infection (Kumari et al., 2016), expression of OsEDSI, OsPAD4 and OsWRKY genes involved in plant innate immunity was investigated. The SSR marker HvSSR10-20 linked to the nematode resistance gene in the resistant variety "Abhishek" was used in the current study to validate in another nematode resistant variety NDR-97.

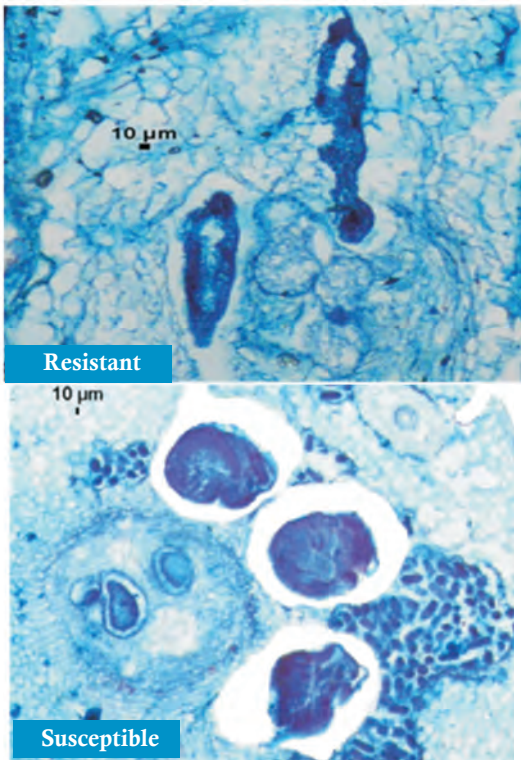

Fig. 10: Differences in giant cell morphology of *M. graminicola* in susceptible and resistant cultivars of rice.

Molecular Nematology

Genome sequencing projects of several PPNs including *Meloidogyne incognita* provide an invaluable wealth of gene sequences which can be used for disturbing the nematode life cycle that in turn can help in target identification for their management options. In the present study, we have given transcriptional evidence for 13 Neuropeptide like protein (nlp) genes in M. incognita. The differential gene expression of two nlp genes (nlp-3 and nlp-12) in five different developmental stages revealed their higher expression in the mature females followed by pre parasitic juveniles. Functional validation using RNAi (Dutta et al., 2015a) was carried out for two nlp genes, *nlp-3* and *nlp-12*. Higher expression of these genes in the pre-parasitic stages and mature females revealed that, these genes might be involved in the host recognition and reproduction process. The in situ hybridization studies of these genes revealed their expression in neurons associated with the basal bulb and tail region of the nematodes. Both the nlp genes were silenced through RNAi which significantly reduced the number of nematodes attracted towards host at different time intervals. Silencing of nlp-12 gene was most effective in preventing the penetration up to 34.63 and 19.33 per cent at 24 and 72 hours respectively after inoculation. Further, the silencing effects on the phenotype could be correlated with the transcript abundance of the target genes by qRT-PCR. After dsRNA soaking, *nlp-3* and *nlp-12* were down-regulated 54 and 7 folds respectively. The long term effects of *in vitro* RNAi was evident as it reduced the disease intensity in terms of galling in the range of 23 to 31 per cent due to reduced nematode infection. There was a 22 to 31 per cent reduction in the number of endoparasites. Additionally, it was impressive to note that a one time dsRNA soaking lasted till the end of nematode life cycle as reflected by reduced nematode reproduction in the range of 24 to 32 per cent and fecundity by 17 to 30 per cent. Based on the present findings, it could be concluded that RNAi of *nlp-3* and *nlp-12* genes indicated them as potential gene targets for effective nematode management through host delivered RNAi for enhanced level of disease reduction.

The FMR Famide-like peptides (FLP_S) are the most diverse and widely studied group of nematode neuropeptides (Papolu et al., 2013). FLP_S play a significant role in the nematode nervous system as a neurotransmitters or neuromodulators role in the nematode system as a neurotransmitters or neuromodulators and proposed to be one of the important targets. The present study was undertaken to examine the stability of transgenic brinjal expressing dsRNA of *flp-18* against different population densities of *M. incognita.* T_1 plants of transgenic brinjal carrying single copy to flp-18 were challenged with 1, 2, 4 and 8 second stage juveniles per gram of soil along with wild type for comparison. Bioefficacy studies performed revealed 40-70% reduction in host invasion compared to wild type plants. Likewise, reproduction and multiplication was also reduced considerable by 41-50 and 60-75% respectively irrespective of the population density. Molecular analyses by PCR and qRT-PCR of the T_1 transgenic plants used in the bioefficacy confirmed both for the presence of transgene and its

expression. Comparison of total proteins of transgenic and wild type plants did not show any difference. This is a pioneer study to demonstrate the performance of host delivered RNAi based transgenics against various population densities. Generally, nematode populations under field conditions are not evenly distributed and quite variable. The population density in given field could be either at economic threshold level or below or above these levels (i.e. under ETL, ETL and above ETL). In view of this, the study suggests that transgenic brinjal plants expressing dsRNA of *flp-18* gene could provide stable resistance of *M. incognita* at various densities. It may prove promising for the management of *M. incognita* under field conditions with varying population densities (Fig 11).

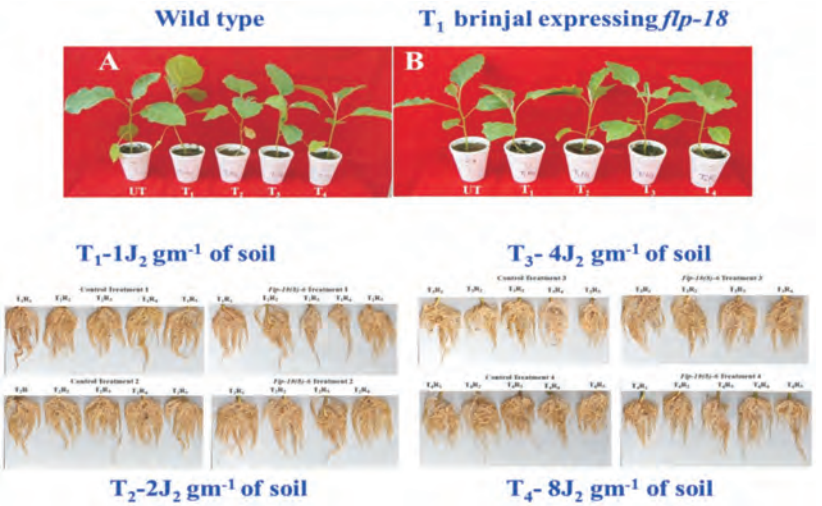

Fig. 11: Bioassay of transgenic eggplants against *M. incognita* infection.

Transformed tomato (*Solanum lycopersicon*) plants (cv. Pusa Ruby) producing dsRNAs targeting AY134442, a dorsal esophageal gland gene and AY134444, a subventral esophageal gland gene of *M. incognita*, were used to study nematode penetration, gall formation and their multiplication. Apart from this giant cell anatomy was also studied to understand the effect of silencing of these genes on the feeding cells (Fig 10) . The *M. incognita* infection analysis was done on molecularly confirmed T_1 tomato plants. The overall reduction in penetration of root-knot juveniles over control ranged between 68.7% and 65.7% in T_1 generation respectively after 2 and 4 days of nematode inoculation. The maximum percent reduction in the development of galls in T_1 was 62-64% over control. Apart from affecting the penetration and development, the silencing strategy also affected the overall giant cell number, their area and the number of nuclei per giant cell and area of giant cells was in the range of 26 to30% and 4 to 7%; 20 to 28% and 39-55% and 12% and 51 to 71% in the tomato plants transformed with AY 134444 and AY134442 genes respectively. The results demonstrated that *in plant* produced dsRNA is ingested by the feeding nematode considerably affected the parasitism capability of the root-knot nematode, *M. incognita.* This resistance engineering technology has a great potential for nematode management as it is eco-friendly and specific against a particular pathogen.

The investigations were carried out to evaluate the response of three synthetic elicitor molecules, namely, DL-β-amino-n butyric acid (BABA) (@ 2000, 4000, 6000 and 8000 µg/ml) as foliar application on wheat *(Triticum aestivum)* crop against the cereal cyst nematode, *H. avanae.* In general, plant growth parameters (shoot length, shoot weight, root length, root weight) of wheat plant increased with the increase in concentration of elicitor molecules. However, the plant growth showed variable response irrespective of the dosage and the type of elicitor molecules. There has been negative correlation between the plant growth parameters and the nematode infestation observed during the spray of different molecules, thus increase in tolerance in plants, against *H. avenae.* This was confirmed by the quantitative estimation of defence related enzymes like peroxidase (PO), polyphenol oxidase (PPO), phenylalanine ammonia lyse (PAL) and lipoxigenase (LOX) were carried out following the foliar application of three synthetic elicitor molecules, 5 and 10 days after inoculation. It was observed that enzyme activity increased 5 days after inoculation of *H. avenae.* The percent increase or decrease of the enzymes activity for of LOX, PO, PPO and PAL over the control in the range of (10-27%). Among the three elicitor molecules, BABA @ 8000 µg/ml was highly effective in reducing the nematode penetration and further development of *H. avenae* and increase in plant growth parameters, followed by JA and SA @200 µg/ml. Thus, these molecules could be exploited for the management of *H. avenae* in wheat under microplot/field conditions.

In a study salicylic acid (SA), Rose Bengal (RB) and β-aminobutyric acid (BABA), spray and root-dip applications were used to induce resistance in eggplant *(Solanum melongena)* cultivar Pusa Purple Long against *M. incognita.* Two enzymes, peroxidase and phenylalanine ammonia lyase, associated with resistance responses in crop plants against nematodes and other pathogens were also assayed so as to understand biochemical basis of resistance. The study results indicated significant increase of plant growth parameters in eggplant after application of the three chemicals @ 25, 50, 100 and 200 µg/ml. The best growth parameters were recorded on treatment with SA followed by RB and BABA. Root dip treatments by SA and BABA were more effective in reducing root-knot nematode population whereas spray treatment by RB proved better in reducing nematode build-up. Overall, SA (200 µg/ml) root dip was most effective in inducing resistance in eggplant as least nematode multiplication was observed. This was corroborated with increase in peroxidase and phenylalanine ammonia lyase (PAL) enzymes across the period of 24, 48, 72 and 96 h w.r.t. to control. Both these enzymes are associated with resistance responses against nematodes and other pathogens in several crops (Pankaj *et al.,* 2013).

The proteinase inhibitors (PI_S) are important elements of natural plant defence strategy against various pathogens including plant parasitic nematodes (Papolu et al., 2016). The PI_S form stable complex with proteolytic enzymes and cause disruption in nematode feeding. The genes encoding PI from Chickpea, *Cicer arietinum,*(*CaPI*) and Black gram, *Vigna mungo,*(*VmPI*) were cloned, expressed in bacterial expression vector pET28a and pure proteins were isolated. The proteins of ~30 kDa and ~25 kDa molecular weight were isolated from *CaPI* and *VmPI* gene expression, respectively and were tested for its efficacy against root-knot nematode (RKN), *Meloidogyne incognita,* one of the most proliferous nematode pests of vegetable crops all over the world. The *in vitro* feeding of *CaPI* and *VmPI* proteins at different concentrations to the second stage juveniles ($J2_S$) of RKN caused upto 27.73% and 55.50% mortality respectively. Further, the potential of *CaPI* (*in vitro*) fed J2s to infest the roots of tomato cv. Pusa Ruby and its effect on root galling was tested. After 10 and 35 day of inoculation, no. of J3/J4 and the root galls on tomato were reduced by 70% and 76.71% respectively. The number of egg masses reduced by 87.50% after 35 days of inoculation. Thus both the PIs showed anti-nematode properties against *M. incognita* J2s upon *in-vitro* feeding. In another study the tomato cv. Pusa Ruby was transformed with *CaPI* gene using *Agrobacterium tumefaciens* mediated transformation. The gene was successfully transformed in tomato which was confirmed by polymerase chain reaction (PCR), RT-PCR and southern blotting. The effect of *CaPI* gene expression on *M. incognita* penetration, development and reproduction was studied in the transformed vis a vis non-transformed tomato plants. The *M. incognita* J2 penetration in tomato roots was 40.94% less whereas; number of third/fourth stage juveniles (J3/J4) in roots was 42.14% less when compared with wild type control. The root gall numbers reduced up to 50.64% and the final population of nematode decreased by 48.64%. The significant difference in growth of transformed plants over wild type upon nematode infection was observed. These studies indicate that PIs can be helpful in developing new management strategies against root-knot nematode, *M. incognita.*

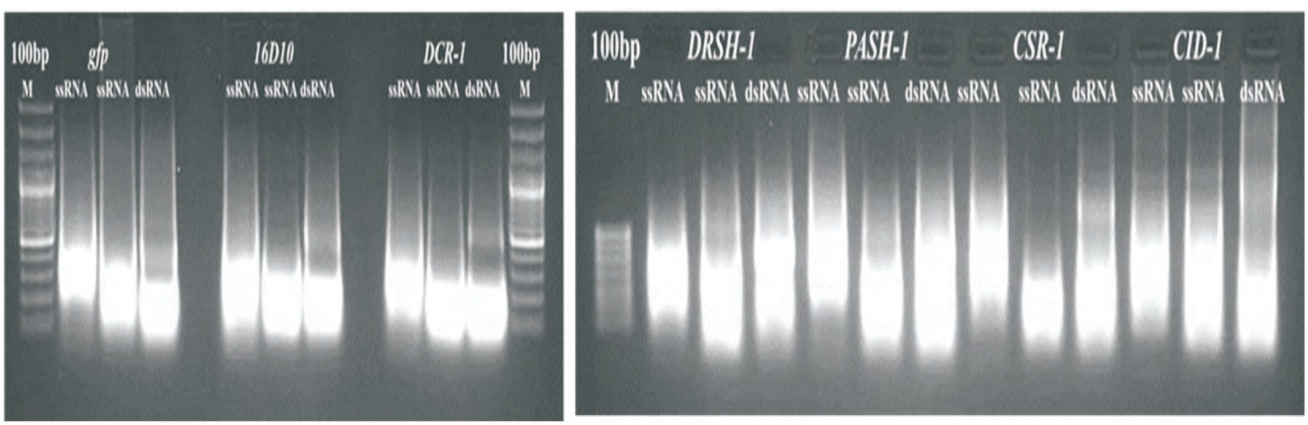

Fig. 12: DsRNA amplification of several RNAi pathway genes in *M. incognita.*

Comparative genomics using bioinformatics could help in identification of the appropriate and potential gene targets but they have to be functionally validated. RNA interference (RNAi) has proved to be very promising for understanding the role of genes so that they could be the candidates to design nematode resistant crops. In the present study, for the first time we had given transcriptional evidence for twenty eight RNA interference patway genes including four new genes, *CSR-1, NRDE-3, SAGO-1* and *PPW-1* in *Meloidogyne incognita.* The differential gene expression of these genes in four developmental stages revealed more expression of these genes in the egg stage. Further, *in vitro* RNAi silencing of the five key pathway genes affected the RNAi processing of a target gene in *M. incognita* juveniles confirming the role of these key genes that also indicated in active RNAi pathway in *M. incognita.* Hence, functional validation using RNAi was carried out for three FMRF amide-like peptide (FLP) genes, *flp-1, flp-12, flp-18* and a fusion of all these three *flp* genes named as *fusion* gene. Higher expression of these genes in the pre-parasites stages revealed that, these genes may be involved in the host recognition process as compared to other developmental stages. The *in situ* hybridization studies of these three genes revealed their expression in sensory, motor and interneurons which are associated with the amphids and phasmids of the nematodes. First time we showed processed exo-dsRNA in to siRNA in PPNs by northern analysis and also quantified the florescence in the nematodes soaked in Alexa fluor 488 dye labeled double stranded RNA (dsRNA) by using fluorescent multimode plate reader. All the three *flps* singly and a *fusion* gene were silenced using RNA. It was found that RNAi silencing of each of the genes significantly reduced the attraction of nematodes at different time intervals both in single and *fusion* gene. Silencing of *fusion* gene was the most effective in preventing the penetration up to 66 and 55% at 24 and 72 hours respectively after inoculation. Further, the silencing effects on the phenotype could be correlated with the transcript abundance of the target genes by real time PCR. The long term effects of *in vitro* RNAi was evident as it reduced the disease intensity in terms of galling in the range of 8 to 54% due to reduced nematode infection. There was a 19 to 63% reduction in the number of endoparasites. Additionally, it was very impressive to note that a onetime dsRNA soaking lasted till the end of nematode life cycle as reflected by reduced nematode reproduction in the range of 15-54% and fecundity by 33-58%. Ultimately, we found an additive effect of the *fusion* gene on disease score. Based on the present findings, it could be concluded that a combinatorial RNAi in the form of a single sequence having multiple genes targeting different crucial nematode functions could be very potential for effective nematode management through host delivered RNAi (Dutta et al., 2015b) for silencing the target genes continuously for enhanced level of disease reduction.

Acknowledgment

We wish to thank all the Faculty Members of the Division of Nematology for extending support and cooperation in preparing the research contributions of M.Sc. and Ph.D. students.

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Plant Pathology

V. K. Baranwal, Saritha R.K. and Bishnu Maya Bashyal

Introduction

The Division of Plant Pathology is more than 100 years old. It was originally established in 1905 as Mycology section of Imperial Agricultural Research Institute (IARI) at Pusa, Bihar to initiate mycological and plant pathological research in India. The sectional status was raised to the status of Division of Mycology in 1943-44 after the Institute was shifted to New Delhi. Subsequently, it was named as Division of Mycology and Plant Pathology in 1947-48 and later it was designated as Division of Plant Pathology. Two Regional Stations, one at Pune (Maharashtra) and other at Kalimpong (West Bengal) were established in 1939 and 1956 for the research on plant viruses.

Disease diagnosis, detection of pathogens and management of disease risks have been the main fabric of Divisional research. The Division has evolved over the years with four major sections, i.e., Mycology, Fungal Pathology, Bacteriology and Virology. *Herbarium Cryptogamae Indiae Orientalis* (HCIO) and Indian Type Culture Collection (ITCC), Center for Advanced Faculty Trainng (CAFT) in Plant Pathology and Referral Center for virus indexing of tissue culture raised plants are the "Life Lines". The Division has highly trained scientific, technical and field staff. It has well equipped laboratories to work on diagnosis and characterization of plant pathogens, electron microscopy, cloning of genomes, electrophoresis, sequencing, use of PCR in disease diagnosis, tissue culture, plant transformation etc.

Division evolved over the years with four major sections Mycology, Fungal Pathology, Bacteriology and Plant Virology. The mandates of the Division are

- To conduct basic and applied research leading to detection, identification and management of plant pathogens
- To serve as a Centre for academic excellence in the area of Post-Graduate education and training in Plant Pathology
- To provide national leadership in plant pathological research through the development of new concepts and technologies

Significant Student's Research Achievements

Taxonomy and pathogen characterization

The phylogenetic relationship of *Curvularia* (imperfect stage) with *Bipolaris* (imperfect stage) and *Cochliobolus* (perfect stage of both the genera) was established where *Cochliobolus* showed as the perfect state of both the genera as the species of *Cochliobolus* clustered with the respective anamorphic (*Curvularia* and *Bipolaris*) species. Through the phylogenetic analysis, *Bipolaris spicifera* clustered with *Cuvularia* and not with *Bipolaris.* Therefore, this species can be transferred and named as *Cuvulariaspicefera* (Pirti *et al.,* 2016).

Potential DNA barcode regions were selected for the identification of *Trichoderma* spp. (β-tubulin), *Bipolaris* spp. (ITS), *Chaetomium* (ITS as primary barcode and β-tubulin as secondary barcode) and *Curvularia* spp. (ITS) and the sequences were deposited in the BOLD (Barcode Of Life Database).

Twenty four (12 isolates from chickpea and 12 isolates from other hosts) isolates of *Sclerotinia sclerotiorum* were grouped into 7 pathotypes and differential cultivar, namely, DCP 92-3, Pusa 212, Vishal, JG 74, GPF 2 and WR 315 for each group were identified. Based on the mycelial compatibility grouping (MCG), 24 isolates were placed into 7 MCGs.

Wilt affected cucumber samples collected from different parts of the country were characterized morphologically, pathologically and based on vegetative compatibility groups (VCGs). All fifty isolates were grouped into eight VCGs arbitrarily named 1-A, 1-B, 1-C, 1-D, 1-E, 1-F, 1-G and 1-H. No self-incompatibility was observed in any of the isolates tested. Based on pathogenicity, isolates were divided into 4 pathogenic groups I, II, III and IV.

Twenty isolates of *Alternaria porri* were collected from different major onion cultivating regions of India were characterized for cultural, morphological, pathogenic and molecular diversity, which demonstrated the existence of considerable variation.

A total of forty isolates of *Alternaria brassicae* were collected from different parts of the country were characterized at morphological and pathogenic level and the isolates showed high level of variability *in vitro* in respect to mycelial growth, growth pattern and sporulation. Protein profiling studies revealed that the isolates were grouped into three distinct groups.

Nucleotide sequence of the S and M RNA segments of a Groundnut bud necrosis virus isolate from *Vigna radiata* (mungbean isolate, GBNV-MB) was determined and compared with another isolate from *Arachis hypogaea* (groundnut isolate, GBNV-type). Comparative sequence analysis revealed that the genome organization of the S and M RNA segments of both GBNV-MB and GBNV-type isolates was similar. However, considerable differences were observed in their intergenic regions (IGRs) and the glycoprotein precursors (Gn/Gc) of the M RNA segments (Fig. 1).

Eight begomovirus isolates causing tomato leaf curl disease originating from different geographical locations of India were characterized. These isolates were identified by common and species specific primers designed on the basis of available sequences. Of the eight isolates two each were of ToLCBV and ToLCGV and one of ToLCNDV and the remaining three isolates appeared to be new viruses. A new begomovirus species and tentatively named Tomato leaf curl Rajasthan virus (ToLCRV) was reported. tomato; *Datura metel, Eclipta alba, Tageteserecta, Phyllanthus niruri, Parthenium hysterophorus* and *solanum nigrum*, were established as alternative hosts of begomovirus.

Three isolates *of Citrus tristeza virus* (CTV), a member of the genus *Closterovirus* (family *Closteroviridae*), D1, D7 and D15 from Delhi were characterized based on sequencing of 5′ORF. The phylogenetic analyses indicated extensive genetic diversity in CTV in Delhi. From analysis of the 3′ half 8.4 kb sequence of three Indian CTV isolates, B5 (Bangalore), D1 (Delhi) and Kpg3 (Darjeeling hills) sequence of Indian isolate B5 is unique and isolates D1 and Kpg3 are recombinant.

Ring spot disease in citrus is caused by *Indian citrus ringspot virus* (ICRSV) a type species of *Mandarivirus*. Distinct symptomatology, transmission, ELISA and RT-PCR assay results indicated these

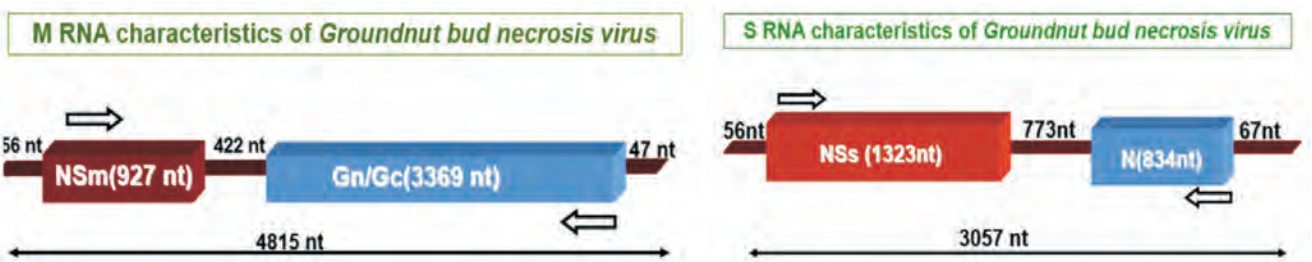

Fig. 1: Characterstics of M and S RNA of groundnut bud necrosis virus

isolates can be either different strains of ICRSV or different species of genus *Mandarivirus.* Analysis of 3' region of ICRSV isolates at molecular level from major citrus growing regions of western India was undertaken which revealed occurrence of diversity in CP as well as NaBP genes of ICRSV among different isolates. Full genome of an isolate of ICRSV was cloned and sequenced. A study was undertaken to determine conclusively whether ICRSV is seed transmitted or not. The screening of the seeds using DAC-ELISA revealed presence of virus in seed coats but not in embryo and endosperm. The seedlings in the grow-out test did not show any symptoms for eighteen months and the virus was not detected in seedlings by DAC-ELISA and RT-PCR. The results indicate that ICRSV is not seed transmitted in Kinnow mandarin.

Garlic common latent virus (GarCLV) is a member of the genus *Carlavirus* belonging to the family *Betaflexiviridae* of the order *Tymovirales*. GarCLV has flexuous filamentous particles similar to the other members of *Carlavirus* containing a single molecule of linear ssRNA of ~8.6 kb. GarCLV does not induce any visible symptoms of its own on garlic but plants show severe yellowing and mosaic when GarCLV infection is associated with other potyviruses The coat protein (CP) gene of five Indian GarCLV isolates was determined. The length of all the CP genes was found to be 960 bp, encoding a protein of 319 amino acids. Comparative nucleic acid sequence analysis revealed 4.3% diversity among the Indian isolates with an overall 11.9% diversity among all isolates worldwide. Amino acid sequence comparison showed significant variability in the N terminal of CP of GarCLV (Pramesh and Baranwal, 2013).

Grapevine leafroll disease (GLD) Fig. 2 is a complex viral disease of grapevine accounting for more than 60 % of yield losses due to viruses. It is caused by distinct filamentous viruses belonging to the family *Closteroviridae*. Despite being known to be present since one decade in Indian vineyards, no authentic study on GLD has been carried out. On the basis of serology, electron microscopy, RT-PCR (reverse transcriptase polymerase chain reaction) and sequencing, we report for the first time, the association of GLRaV-1 and -3 with GLD symptomatic vines of the vineyards of Nashik and Pune regions of India. During the investigation, p24 gene of GLRaV-1; partial HSP70h (heat shock protein 70 homologue), coat protein (CP) and entire p19.7 genes of GLRaV-3 were characterized.

Fig. 2: Symptoms of Grapevine leaf roll disease

Streak disease of banana and plantain (Musa spp.) caused by several badnaviruses, collectively known as banana streak viruses (BSV), is a major constraint in banana production and genetic improvement worldwide. Complete genome sequences of three episomal *Banana streak MY virus* (BSMYV) isolates sampled from triploid banana hybrids (Chini Champa: AAB; Malbhog: AAB and Monthan: ABB), grown in North-East and South India was achieved by sequence independent improved rolling circle amplification (RCA). RCA coupled with restriction fragment length polymorphism (RFLP) revealed diverse restriction profiles of five BSMYV isolates (Fig. 3) (Sharma *et al.,* 2015).

Chirke and foorkey disease is an important constraint of large cardamom cultivation in eastern sub-Himalayan mountains. However the genome of the chirke virus has not been fully characterized and molecular diagnostics were not available. In this study, genome sequence of 5961 nucleotides of the virus associated with chirke disease was achieved. Phylogenetic analysis showed that the virus associated with the chirke disease of large cardamom is a new species under the genus *Macluravirus* in the family *Potyviridae* for which the name Large cardamom chirke virus (LCCV) has been proposed.

Soybean crops showing systemic mottling, mosaic and leaf deformation were observed at high disease incidences (25.1–71.0%) in the kharif season of 2011 and 2012 in the experimental farm of the Indian Agricultural Research Institute (IARI), New Delhi. Symptomatic soybean leaves contained flexuous particles (650 9 12 nm), suggesting an infection by a Carlavirus. The causal virus was characterized as a strain of *Cowpea mild mottle virus* (CPMMV) on the basis of mechanical inoculation, whitefly transmission, seed transmission and sequencing of the viral genome. This is the first report of natural infection by a distinct strain of CPMMV in soybean in India.

Papaya ringspot virus (PRSV), vectored in nature by aphids, continues to be a major limiting factor affecting the production of papaya as well as on cucurbits in India (Fig. 4). Coat protein (CP), the helper component proteinase (HC-pro) and nuclear inclusion protein-a (NIa-pro) coding regions from 12 PRSV (P and W) isolates originating from different locations were analyzed.

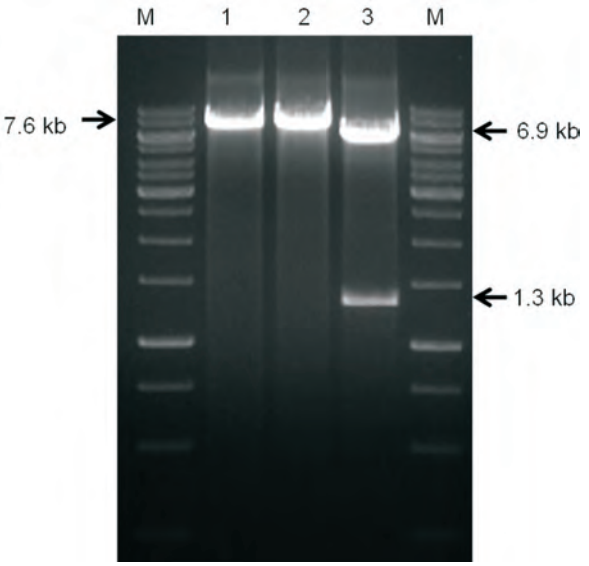

Fig. 3: Improved random primed rolling circle amplification (RCA) of badnaviruses genome. *Kpn*1 digested RCA product derived from Banana streak MY virus infected banana hybrid plants Lane 1: Chinichampa (AAB), lane 2: Monthan (ABB), 3: Malbhog (AAB), M: 1 Kb maker (Fermentas, USA).

The results indicated that the CP is more diversed than NIa-pro and HC-pro coding regions. Further, the Indian isolates are more variable (upto 12%) than the other Asian (upto 7%) and American isolates (upto 4%). Based on HC-pro and NIa-pro sequence-based phylogeny, Indian isolates showed more relatedness with the other Asian isolates than American isolates. In contrast, CP-based phylogeny revealed that the Indian isolates were more closely related with American than the other Asian isolates and formed three separate clusters as Indian, other Asian & American clusters. Interestingly, three sub-clusters North-Indian, South-Indian and North-East-Indian were formed within Indian isolates. Recombination analysis revealed seven putative recombination events in CP, five in NIa-Pro and none in HC-Pro.

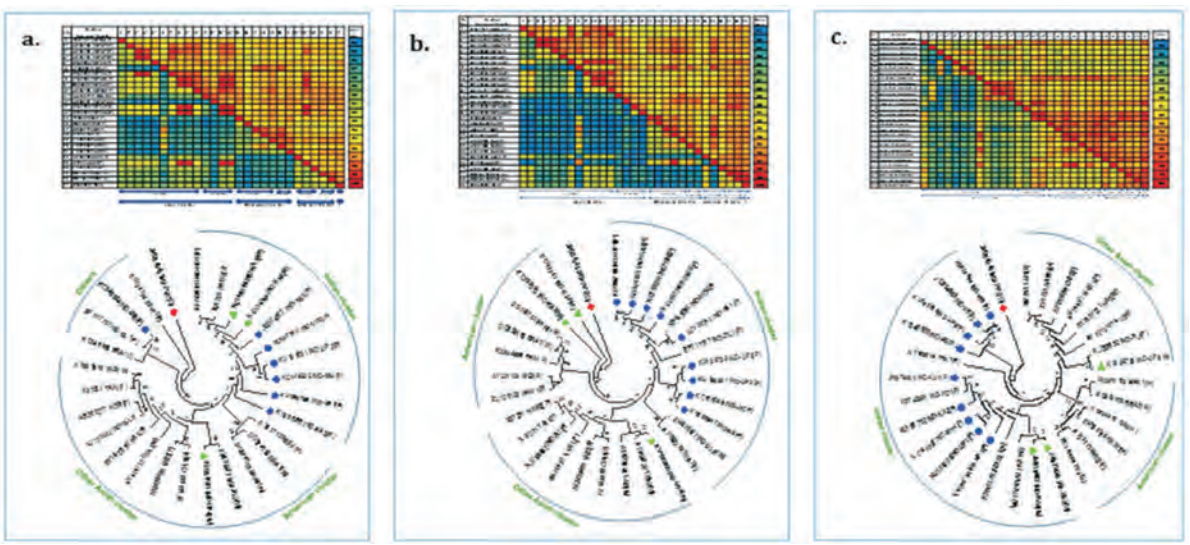

Fig. 4. Comparative sequence and phylogenetic analysis of HC-pro (a); Nia-pro (b) and CP coding regions (c) of PRSV isolates

Chirke and foorkey disease is an important constraint of large cardamom cultivation in eastern sub-Himalayan mountains. However, the genome of the chirke virus has not been fully characterized and molecular diagnostics were not available. In this study, genome sequence of 5961 nucleotides of the virus associated with chirke disease was achieved. Phylogenetic analysis showed that the virus associated with the chirke disease of large cardamom is a new species under the genus *Macluravirus* in the family *Potyviridae* for which the name large cardamom chirke virus (LCCV) has been proposed.

Incidence of toria (*Brassica rapa* cv. toria) phyllody (TP) disease with phyllody, virescence, witches broom, extensive malformation of floral part, formation of bladder like siliques and flower sterility symptoms was observed in the range of 4.8% to 9.8 % in ten different lines in experimental fields of Indian Agricultural Research Institute, New Delhi, India during the years 2008 and 2009. TP could be transmitted either by grafting or by dodder to toria and other 9 rapeseed/mustard species as confirmed by nested PCR. Transmission of TP phytoplasma to periwinkle was successful only through dodder. Molecular characterization of TP samples collected from four different locations (New Delhi, Meerut, Ludhiana and Bharatpur) in India revealed all to be identical and belonged to 16SrIX Pigeon pea witches-broom group (Mehdi and Baranwal, 2011).

The phytoplasma etiology was confirmed using direct and nested polymerase chain reaction (PCR) with amplification of a fragment of 1.5 and 1.25 kb of 16SrRNA gene of the eleven seasamephyllody and witches broom isolates. *Sec* A and *In silico* restriction enzyme digestion of 1.25 product of 16Sr DNA with *Bam*HI,*Bfa*I, *Eco*RI, *Hha*I, *Hpa*II, *Rsa*I, *Alu*I, *Hae* III, *Taq*I and virtual *iphy* classifier analysis with 17 restriction endonucleases grouped the SP and WB isolates into 16Sr I-B and 16Sr II-C subgroups, from Uttar Pradesh , while, the isolates from Delhi and Bihar belonged to16Sr I-B subgroup (Fig. 5). *Hishimonus phycitis* (Disant) could transmit the SP phytoplasma from diseased to healthy sesame plants. A weed *Sclerocarpus africanus* Jacq

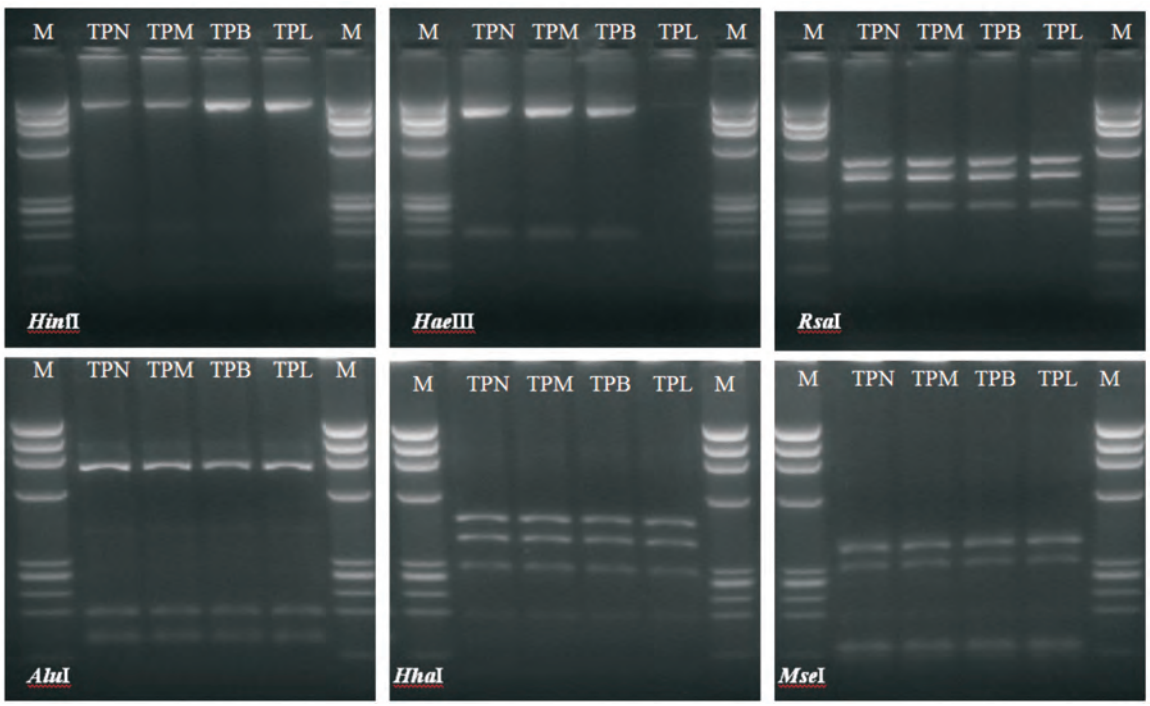

Fig 5: Actual RFLP analyses of 1.25 kb of 16S rDNA nested-PCR products of TP phytoplasma (amplified using primer pair R16F2n/R16R2) digested with *Hinf* I, *Hae*III, *Rsa*I, *Alu*I, *Hha*I and *Mse*I restriction enzymes. TPN: toriaphyllody strain New Delhi, TPM: toriaphyllody strain Meerut, TPB: toriaphyllody strain Bharatpur, TPL: toriaphyllody strain Ludhiana. M: phi X174/HaeIII Marker (1353, 1078, 872, 603, 310, 281, 271, 234, 194, 118, 72).

of family astercaeae showing little leaf and witches' broom symptoms was reported as putative alternate host for SP phytoplasma.

The survival or increased fitness of the vector *Bemisia tabaci* is attributed to endosymbiont present in their body parts. Whitefly population from cotton field contains *Bacillus* species, *Ralstonia* species in all developmental stages, egg, four instars and in adults. However, in the case of glasshouse reared population, the predominant bacteria were *Ralstonia* species besides *Bacillus pumilus* present in instars only.

Development of detection techniques for plant pathogens

PCR using species specific SCAR primers were developed for karnal bunt (*Tilletia indica*) and *Puccinia triticina* of wheat; wilt pathogens such as *Fusarium udum, F. oxysporum* f. sp. *cucumerinum, F. Oxysporum* f. sp. *meloni,* and *F. oxysporum* f. sp. *ciceri*, and other fungal pathogens such as *Rhizoctonia solani* and *Fusarium fujikuroi*. RAPD primer OPA 8 can be used for identification of *Sclerotinia* species whereas, primer OPC 3 was able to distinguish sclerotia and non-sclerotia forming isolates. PCR based detection was developed for *Xanthomonas campestris* pv. *campestris.* An easy, cost effective and rapid nitrocellulose membrane (NCM) based DNA template preparation for detection of toria phyllody phytoplasma has been developed also tested in different crops. LAMP based detection assaywas also developed for the detection of *P. triticina* having sensitivity of 100fg, which shall be helpful in rapid detection and diagnosis of wheat leaf rust (Fig. 6).

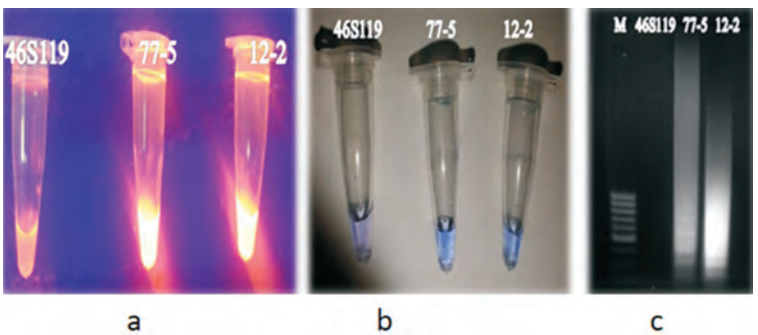

Fig. 6: Visual detection of LAMP reaction for specific detection of *P. striiformis* using (a) EtBr (b) HNB dye (c) Agarose gel electrophoresis of LAMP products. Lane M: 100 bp marker 1: *P. striiformis tritici* (46S119); 2: *P. triticina* (77-5) and 3: *P. triticina* (12-2)

In vitro expressed virus coat protein (CP) provides an easy alternative for raising antisera against viruses were successfully employed for raising specific antisera against *Large cardamom chirke virus* (LCCV) and *Cardamom bushy dwarf virus* (CBDV)associated with foorkey disease of large cardamom, *Groundnut bud necrosis* virus (GBNV) Garlic *common latent virus* (GarCLV) and GLRaV-3 and Cardamom bushy dwarf virus (Fig 7).

In badnaviruses precise CP sequences are not known and two putative CP coding regions (p48 and p37) of *Banana streak MY virus* (BSMYV) were identified *in silico* by comparison with caulimoviruses, retroviruses and *Rice tungro bacilliform virus*. The putative CP coding region (p37) was *in vitro* expressed in pMAL system and affinity purified. The purified fusion protein was used as antigen for raising polyclonal antiserum in rabbit. Two immune dominant linear epitopes were identified at N and C-terminal of putative CP of BSMYV (pep-I and

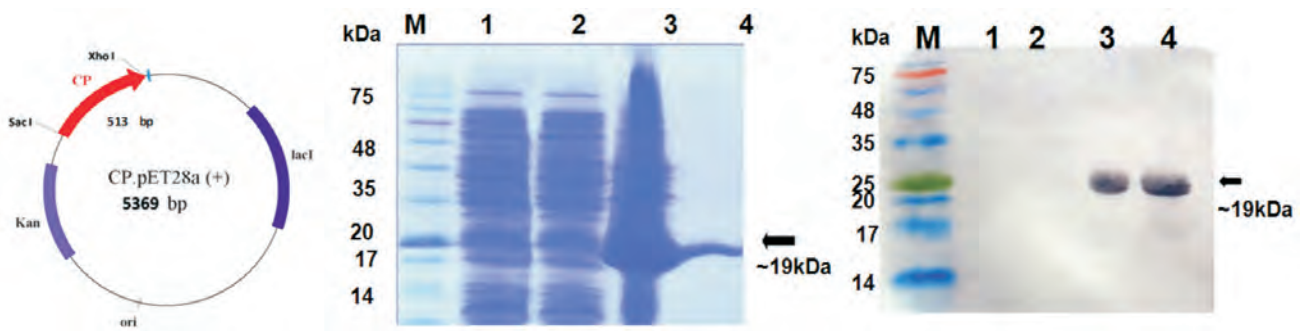

Fig. 7: Expression of capsid protein of cardamom bushy dwarf virus in *E. coli*

pep-II), synthesized and used for polyclonal antiserum production. Only anti pep-I antiserum strongly reacted with BSMYV virions in ISEM and ACP-ELISA.

Immunocapture-RT-PCR (IC-RT-PCR), more sensitive than ELISA and RT-PCR alone, is a technique where the virus can be detected without isolating the RNA. A modified extraction and RNA release protocol was developed for IC-=RT-PCR and was validated for specific detection of *Grapevine leaf roll-associated virus* 3 (GLRaV-3) in the vines of five infected grapevine cultivars (Kumar et al. 2015). In onion mixed infection of *Onion yellow dwarf virus* and allexi viruses viruses could be established by duplex RT-PCR.

A sensitive duplex-immunocapture-PCR (D-IC-PCR) was standardized for the sensitive, reliable and accurate routine indexing of episomal BSV infection in tissue cultured and field banana samples (Sharma *et al.,* 2014).

Virulence analysis

Twenty five Indian wheat genotypes were evaluated for stem, leaf and stripe rust resistance genes using nine different pathotypes *viz.,* 40-1, 40-A and 117-6 of stem rust; 77-5, 77-8, 104-2, 12-1 of leaf rust and 46S119 and 78S84 of stripe rust. The most commonly detected stem resistance genes were Sr2 and Sr31 which were present singly or in combination with other resistance genes in wheat genotypes (Gurjar *et al.,* 2012). The presence of Sr 2 (40%) gene was observed in ten genotypes, HD 2894, HD 2687, VL 616, UP 2425, Raj 3765, HI 1531, HI 977, DBW 16, PBW 343, WL 711 (+*Lr*57). The presence of *Lr26* gene was observed in 9 genotypes (36%) followed by *Lr13* (28%), *Lr34* (24%), *Lr23* (20%) and *Lr* 24 (12%). *Yr9* gene was observed in 9 genotypes (36%) followed by Yr 2 (28%) and *Yr18* (20%).

Analysis on virulence pattern of Basmati rice blast (*Magnaporthe oryzae*) isolates from North-western regions of India was made by testing twenty four monogenic rice entries, each carrying single *R*-gene for blast resistance. Mawana isolate exhibited the maximum virulence showing compatibility with 83% of blast resistance genes. This isolate was avirulent only on four *R*-genes, *viz. Pi-11, Pi-s*^{h}*, Pi-ta*^{2}and *Pi-z*^{t}. With regard to virulence spectrum of other *M. oryzae* isolates, NDPB1 and Nagina isolates were virulent on 75% and 72.7% *R*-genes respectively. All these three highly virulent isolates were avirulent on two *R*-genes, *i.e.Pi-ta*^{2} and *Pi-z*^{t}. Further, *Pi-ta*^{2} was identified as the most effective resistance gene on which all the isolates, except one from Aligarh, were avirulent (Dubey *et al.,* 2014).

Race profiling of rice blast isolates revealed that the *M. oryzae* race U77-i7-k177-z17-ta733 (Mo-ni-0068-Panipat, Mo-si-mnd1-Mandya, Mo-ei-ggt1-Gangtok and Mo-ei-ctc4-Cuttack) is highly virulent and compatible to all the tested *R*-genes as well as widely used blast resistant donor Tetep. 60 rice landraces have been validated phenotypically and molecular level of major blast *R*-genes (*Pi1, Pi5, Pi9, Pib, Pita, Piz5* and *Pi54*), 6 accessions *viz.* BAM845, IC133156, IC124676, IC346849, IC371862 and IC369532 expressed high degree of resistance to majority isolates of both regions. The molecular validation revealed that, *Pi1*, *Pi5*, *Pi9*/*Piz5*, *Pib*, *Pita*/*Pita2*and*Pi54*genes were found in these land races, respectively. Genetic analysis for blast resistance was carried out in two land races, *viz.* Vanasurya and IC471862. The F2 population derived fromIC371862 x Co 39 (blast susceptible rice cultivar) and Vanasurya x Co 39 crosses showed a 3R:1S segregation ratio, indicating the resistance response being governed by a single dominant gene.

Virulence analysis of 36 isolates of *Fusarium oxysporum* f. sp. *ciceris*, wilt pathogen of chickpea, collected from 12 states of India, were grouped into eight raceson a new set of 10 differential cultivars of chickpea, namely, C104, JG 74, CPS 1, PUSA 212, WR 315, KWR 108, GPF 2, DCP 92-3, Chaffa and JG 62. Diversity among Indian populations of *Foc* representing 8 races was determined by vegetative compatibility grouping (VCG). Out of 39 isolates, 18 isolates were self compatible and the rest were self incompatible. Based on pairing behaviour, all *Foc* isolates were placed in to single VCG 0280 (Saabale and Dubey, 2014).

Isolates of *Rhizoctonia solani* from rice, maize and mungbean were studied for their variability with respect to cultural, morphological, pathogenicity and genetical characteristics. Reciprocal inoculation of the hosts with

the isolates of *R. solani* found positive and proved cross infectivity. Four rice isolates, two isolates each from maize and mungbean were more virulent. Maize isolate, MRS12 was non-sclerotial in nature and could not infect any of the test hosts.

Bacterial blight (BB) caused by *Xanthomonas oryzae* pv. *oryzae* (Xoo) is a potential threat to rice production worldwide. 60 Xoo isolates were pathotyped into 6 races. Among the six races, race 4 and 6 were found to be predominant in north-western part of India while race 2 and 3 were in north-eastern states including West Bengal, Assam, and Meghalaya. Race 1, and race 2 comprises less virulent population producing mean BB lesion <10 cm on most of the NILs, compared to the other four races. Among the R-genes, the most vulnerable were Xa1 and Xa2 showing susceptible reaction to all the races while *Xa*3, Xa11, *Xa*13, and *Xa*21 were found to be stable showing resistant to moderately resistant type BB reaction.

Host pathogen interactions

In order to identify the early-defense induced genes of wheat in response to *Bipolaris sorokiniana* infection, forward suppression subtractive hybridization (SSH) cDNA library was prepared using wheat genotype Chiriya 7 (resistant) as tester and Sonalika (susceptible) as driver, therefore, this library mainly contained up-regulated genes during incompatible interaction between wheat and *B. sorokiniana*. More than1500 clones could be obtained from SSH cDNA library, out of which around 500 clones after colony PCR were subjected to sequencing. The sequences were analysed by BLAST for homology search against the Genbank non-redundant database. Finally 46 gene sequences after removing repeats were analysed and categorized based on their putative functions and the hosts. Out of them, 15 (33 %) were classified as plant defense responsive genes, while 22% genes were having metabolism related functions, 9% genes were related to transcription/translation and 6% genes had cellular transport related functions (Fig. 8). Thirteen percent clones were categorised under miscellaneous category. Only 4% genes were related to signal transduction while 13% were unknown genes. Some of the significant defense genes identified are: Cysteine protease (CP), Calreticulin (CRT), Predicted omega amidase NIT-2 like, Dual specificity protein kinase pyk-1, Pacifastin like, Della (Rht-B1), and UDP- glycosyl transferase (UGT). Calreticulin (CRT) is a key Ca^{2+} binding protein residing in endoplasmic reticulum (ER) which is highly conserved in all eukaryotic organisms (Honnareddy *et al.,* 2014).

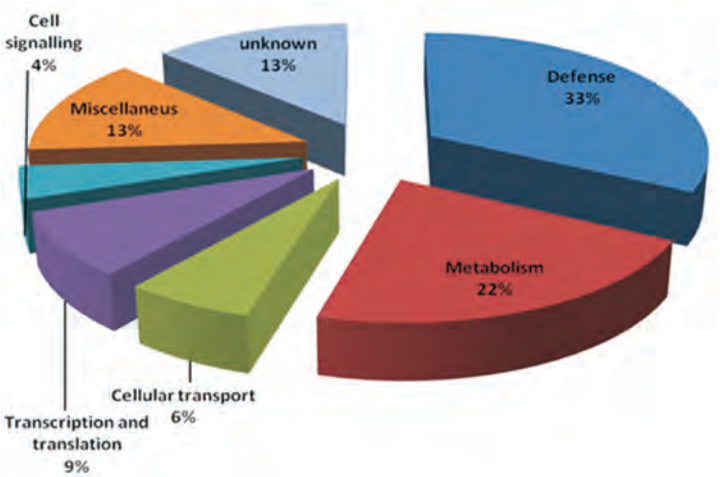

Fig. 8: Functional cataloguing of differential expressed genes in resistant wheat genotypes Chiriya 7 post inoculation with *B. sorokiniana*

Mitogen activated protein kinase (MAPK) cascades are universal signal transduction modules that play crucial role in resistance response of plants against biotic stresses. MAP kinases involved in defense response in wheat against *Bipolaris sorokiniana viz.,* TaMAPK2, TaMAPK1, TaFLRS and TaDSPK were isolated, cloned and sequenced from wheat genotype Chiriya 7and expression analysis using semi-quantitative RTPCR and qRT-PCR, upon challenging with *B. sorokiniana*, in wheat resistant genotypes Chiriya 7 and susceptible genotypes Sonalika showed that the expression of TaMAPK1, TaFLRS and TaDSPK was induced rapidly during the first 19 hour of post inoculation in incompatible interaction than in compatible interaction in wheat seedlings. In contrast expression of TaMAP2 was activated rapidly in compatible interaction during 19 hours of post inoculation. These results suggest that TaMAPK1, TaFLRS and TaDSPK may play an important role in wheat disease resistance (Fig. 9).

The SSPs having putative role in pathogenicity were selected for expression analysis, as effectors for *B. sorokiniana* and *B. oryzae*. The expression study of the selected six SSPs was observed under fungal mycelium growth of *B. sorokiniana* and *B. oryzae* as well as *in planta* after inoculation on rice and wheat. Among them *B. sorokiniana* specific effector protein (BS_SSP2) did not express *in vitro* but in presence of host it showed expression, which is a character for being an effector. Therefore, this could be a potential effector protein for *B. sorokiniana*.

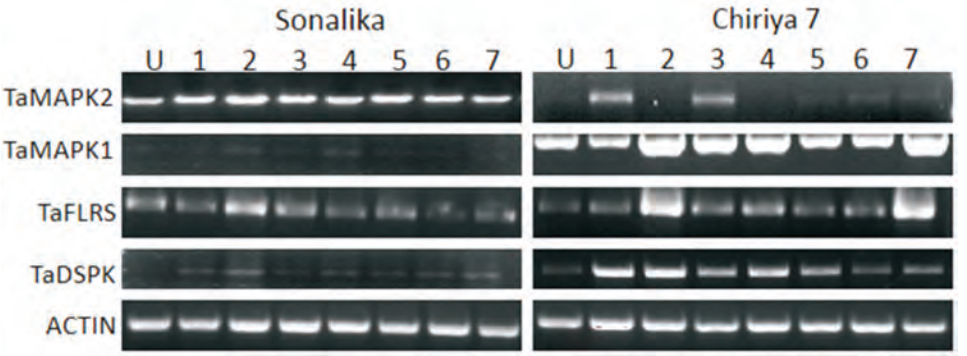

Fig. 9: Time course expression analysis of the TaMAPK 2, TaMAPK1, TaFLRS and TaDSPK amplified through semi-quantitative RT PCR in the genotype Sonalika and Chiriya 7 at specified periods of post inoculation with *B. sorokiniana* BS-75 isolate. Lane U – uninoculated; lane 1- 2 hours post inoculation (hpi); lane 2-19 hpi; lane 3- 24hpi; lane 4- 48 hpi; lane 5- 4 days post inoculation (dpi), lane 6-6dpi; lane 7-8dpi;and actin is taken as internal control

Melanin isolated from twenty isolates each of *B. sorokiniana* and *B. oryzae* showed a significant positive correlation observed between pathogenicity and melanin content. *In vitro* expression for melanin pathway genes *BRN1* and *SCD1* using qRT-PCR revealed high expression in *B. sorokiniana*. Functional analysis of melanin pathway gene *SCD1* through homologous recombination mediated gene knockout in *B. sorokiniana* showed that knockout mutants displayed an albino phenotype and no infection was observed on inoculation on wheat (Fig. 10).

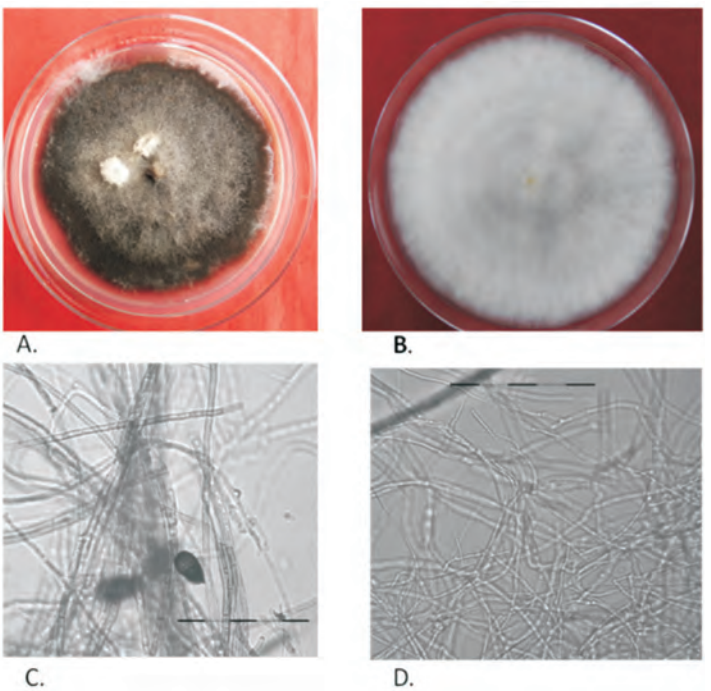

Fig. 10: Comparison of branching pattern of WT and Δ*SCD1* knockout strains. Morphological features of (A). WT strain (B). Δ*SCD1* knockout strains. Microscopy was carried out after 8 days of fungal growth in PDA at 40X lens. Normal growth was observed in (C). WT strain (D). Δ*SCD1* knockout strains, both in terms of extent of branching and lengths of the branches

Next generation sequencing and qRTPCR techniques were explored to identify the transcripts and verify the expression level of genes involved in host pathogen interaction wrt*Lr24* based resistance to *Puccinia triticina* in wheat. *De novo* transcriptome assembly showed 66,415 and 68,688 transcripts in resistant and susceptible genotypes respectively. Transcriptomics study of *Lr*24 based resistance revealed 5873 genes unique to resistant genotype having *Lr*24, 6782 genes unique to susceptible genotype, while 10841 genes were common to both. The Top-BLAST hit result of the predicted coding domain sequence (CDS) for both resistant and susceptible genotype showed maximum similarity with *Aegilops* sp. followed by *Triticum* sp. and *Hordeum vulgare*. A total of 1407 and 1078 SNPs were detected in susceptible and resistant genotype respectively having potential application in marker assisted breeding for leaf rust resistance (Fig. 11). A total of 118 and 113 dinucleotide SSRs, 974 and 872 tri-nucleotide, 20 and 8 tetra-nucleotide

SSRs were identified in susceptible and resistant genotype respectively, which can be further utilized in *Lr*24 based marker aided selection for leaf rust resistance and genetic mapping of *Lr*24 linked genes using mapping populations. A total of 659 genes were found to be differentially expressed of which 349 genes were upregulated and 310 genes were down regulated in resistant genotype having *Lr*24 gene. Most of these defense related genes upregulated in early hours (before 72hpi) indicating that a cascade of defense starts early in *Lr*24 mediated resistance which successfully inhibited pathogen establishment.

Fig. 11: Ven diagram depicting number of unique and differential genes in HS 240+*Lr* 24 and HS240

Resistant Gene Analogues (RGAs) from resistant genotype of pigeon pea against Fusarium wilt have been isolated, cloned and sequenced. Based on nucleotide sequence similarity, seven classes of RGAs have been identified with their constitutive nature of expression. Sequence alignment has shown motif of P-loop and Kinase 2a and most of the Kinase 2a motifs were showing TIR homology.

Polymerase chain reaction based cDNA subtraction method termed as Suppression Subtractive Hybridization (SSH) technique, was used to selectively amplify the transcripts corresponding to the gene expression induced by *Foc*, belonging to race 4, which were differentially expressed in resistant (WR315) and susceptible (JG62) chickpea genotypes. One hundred eighty one recombinant clones were sequenced based on colony PCR results out of 1078 and 1274 clones from resistant and susceptible cultivars. The sequences identified from present study were classified into 8 functional categories namely, defense, signal transduction, metabolism, hypothetical, transcription and translation, transportation, fungal origin and sequences unknown homology with database. Defense and signal related genes occurred at 24% and 11% from resistant and 18% and 24% from susceptible cultivars, respectively, these include many oxidative burst and pathogenesis related genes. Expression study of defense related genes by semi quantitative and qRT-PCR analysis proved that the resistant cultivars showed many fold higher expression as compared to the susceptible cultivar. Higher expression of super oxide dismutase, metallothionein and cysteine protease in the resistant cultivar indicated that oxidative burst plays a major role against *Fusarium* wilt disease. The partial sequences identified from the present study offers a great potential for developing transgenic chickpea against Fusarium wilt disease (Saabale and Dubey, 2012).

A study was carried out on histopathological and molecular characterization of maize genotypes against Maydis Leaf Blight caused by *Bipolaris maydis* using five susceptible and five resistant genotypes. The leaf hair density and wax deposition was high in resistant and significantly less in susceptible. Average stomatal count was found less in resistant than susceptible genotypes. The photosynthetic rate was significantly high in resistant genotypes immediately after infection. Among 30 SSR primers used, only 3 primers viz. umc1335, umc1590 and y1 located on 1.06, 1.06 and 6.02 bin locus, showed polymorphism. The identified markers umc1590 and y1 could further be exploited in marker assisted selection for MLB resistance.

Leaf blast pathogen *Magnaporthe grisea* has been reported to be the causal agent in rice and pearl millet as well as other grasses. Cross infectivity test indicated pearl millet leaf is more conducive for formation of comparatively higher number of appressoria than on rice irrespective of isolates used for inoculation. It appeared that appressoria formation is largely influenced by host surface. Leaf surface comparison between pearl millet and leaf surface based on contact angle of water droplet, wettability and surface retention indicated significant variation between the surfaces. Low contact angle values on pearl millet surface indicated that the liquid spread up and retention on the surface while high contact angle values on rice surface is the reason for poor spreading and retention. Higher ratio of number of appressoria to conidia on pearl millet leaf surface than on rice leaf surface indicated that rice leaf surface is less conducive to

pathogen *M. grisea* than pearl millet leaf surface. It is evident that leaf surface characteristics of the two host play significant role probably by making difference in water or moisture availability for the pathogen required for infection and symptom development.

Seeds, seedlings and florets of rice genotype Pusa Basmati 1509 and Pusa1342 were inoculated with conidial suspension of bakanae disease pathogen *Fusarium fujikuroi* (F250) and progress of pathogen as well as the localization was observed at 6, 12, 18,24,48 and 72 hrs post inoculation through scanning electron micrcoscope and light microscopes. The micro conidia of *F. fujikuroi* started germ tube formation at six hours after incubation; germination percentage was maximum at 9 hrs after incubation. *F. fujikuroi* colonized glume, embryo and husk of rice seed. Colonization of *F. fujikuroi* was maximum in husk followed by embryo of rice seed (Fig. 12). The different types of infectious structure such as swollen tip hyphae, infectious hyphae, appressorium and infection cushion were observed in *F. fujikuroi* rice interactions. Maximum transmission percentage was found in floret inoculation (45.00 %) followed by seedling (33.25 %) and seed inoculation (30.50 %) in rice genotype Pusa Basmati 1509.

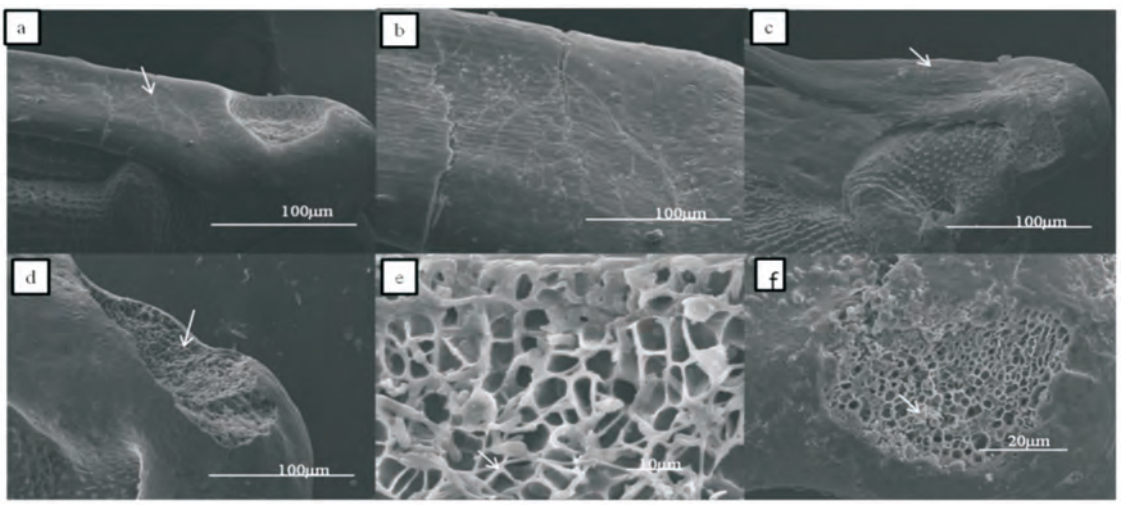

Fig. 12: Scanning micrographs showing colonization of glume and embryo by *Fusarium fujikuroi.***a, b, and c, colonization of glume of rice seed PB 1509, a ,b of 24 hpi and c of 72 hpi, d, e, and f, colonization of embryo after, 24 and 72 hours of inoculation respectively**

Constitutive expression of uncharacterized pathogenicity genes of *Colletotrichum orbiculare* in a minimal medium indicated that the genes are not redundant and may function under stress conditions. Among the genes, significant expression of *ENH87556* in menodione (1.3-fold) together with a weak expression in hydrogen peroxide in relation to untreated control indicated its role in oxidative stress generated due to the superoxide radicle of menodione. Additionally, a strong expression (3.8-fold) of the gene in ferulic acid greater than that of either control, minimal medium or minimal medium with leaf extract indicated its role in phenol metabolism. Thus, the gene could be a potential molecular target for anthracnose control upon validation by functional analysis.

Bacterial blight caused by *Xanthomonas axonopodis* pv. *punicae* (Xap) is an important disease of pomegranate. We identified six Xop type T3SS effectors in Xap (*i.e.* XopC2, XopE1, XopL, XopN, XopQ and XopZ) using gene-specific PCR. A null mutant of XopN effector (XapΔ*xopN*) was developed using double crossover based homologous recombination to understand its possible role in bacterial virulence. Wild type Xap but not XapΔ*xopN* produced intense water soaking in infiltrated pomegranate leaves. The *in planta* population count of XapΔ*xopN* was reduced approximately 32-fold relative to the wild strain indicating that *xopN* is required for maximal growth and virulence of Xap in pomegranate. Besides, full length sequence of XopN was generated and a XopN specific marker was developed.

Bacterial wilt caused by *Ralstonia solanacearum* is a serious disease that causes severe yield and quality losses in many plants. In order to understand the phenotypic response of *A. thaliana* against *R. solanacearum* race 4 / biovar 3, different dilution of bacterial suspension were inoculated on seeds and roots of plants, and found that at seedling wilted in a density dependent manner. Transcriptional changes associated with wilt disease development in *A. thaliana* due to *R. solanacearum*, was analyzed in microarray based gene expression profiling using ATH1 Affymetrix gene chips representing over 22,500 probe sets representing approximately 24000 genes of *A. thaliana* (Fig. 13). Over 1000 genes were found differentially expressed in Col-0 inoculated with bacteria on roots at 1.0 OD of bacterial cells whereas 443 were differentially at 0.1 OD of bacterial suspension. Upon seed inoculation only 85 were differentially expressed in the Col-0 inoculated. The up-regulated genes included genes related to abscission, aging, defense and immune response, auxin stimulus, ethylene stimulus, jasmonic acid and salicylic acid biosynthesis and signalling, water depriviation, tap and lateral development process. Significantly up-regulated genes such as phytoalexin deficient 3 (pad3), arabidopsis thaliana centroradialis (atc), dark inducible 11 (din) were further validated using real time PCR experiment that clearly validated the microarray data.

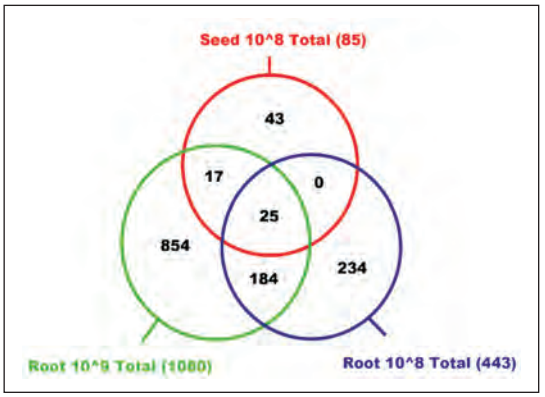

Fig. 13: Altered gene expression in *Arabidopsis* Col-0 upon inoculation with *Ralstonia solanacearum* race 4 Venn diagram showing number of differentially expressed common and unique genes in arabidopsis upon *Ralstonia* interaction on seed or roots with varying density of bacterial cells

Potato virus Y (PVY) of and *Potato virus X* (PVX), are two RNA viruses that together result in synergistic interaction. In the present study, two well defined strains, PVY-Del66 (N-O strain) and PVX-ptDel9 that is asymptomatic on potato were used to study their interactions during mixed infection in *Nicotiana benthamiana* and *Nicotiana tabacum*. Mixed infection caused severe necrosis and death of plants. Synergistic symptoms in tobacco were more prominent under low temperature conditions than at high temperature. In low temperature conditions, the coat protein (CP) level of PVX in *N. benthamiana* was found to be two-fold higher than the CP of PVY. However, when PVY and PVX viruses were inoculated together, the level CP of both the viruses increased and reached to the peak before that in single virus inoculations. The levels of suppressor genes of the same samples revealed that*p*25 of PVX was higher than HC-Pro of PVY in single inoculations. Under mixed inoculation of PVY and PVX, expression of *p*25 declined to the level of HC-Pro, which co-related to the declining levels of CPs of PVY and PVX.

siRNA generating sequences of 21 nucleotides were identified *in-silico* from coat protein and silencing suppressor genes of PVY and PVX. The target sequences were used to generate artificial microRNA constructs by inserting them in sense and antisense directions on the base of the stem of miR159a backbone. Four artificial microRNAs (YCPmiR, XCPmiR, HC-PromiR and *p*25miR) constructs were developed and validated by transient assays in *N. benthamiana*. *N. benthamiana* plants treated with artificial microRNA constructs showed no symptom of PVY and PVX. The ELISA revealed that the constructs were effective in reducing the respective virus load in tobacco and potato.

A partial tandem dimer containing 1.8-mer of BSMYV-IN1 was constructed in binary vector. The agroinoculated banana plants were not positive for BSMYV DNA in RCA three months post-inoculation which might be due to the homology dependent silencing because of the presence of integrant eBSV sequences in inoculated banana plants.

Biological control

Morphological, molecular and biological control analyses were used to evaluate variation in the *Trichoderma* spp. and to assess whether these isolates have any teleomorphic link with Hypocrea (Fig. 14). The morphological

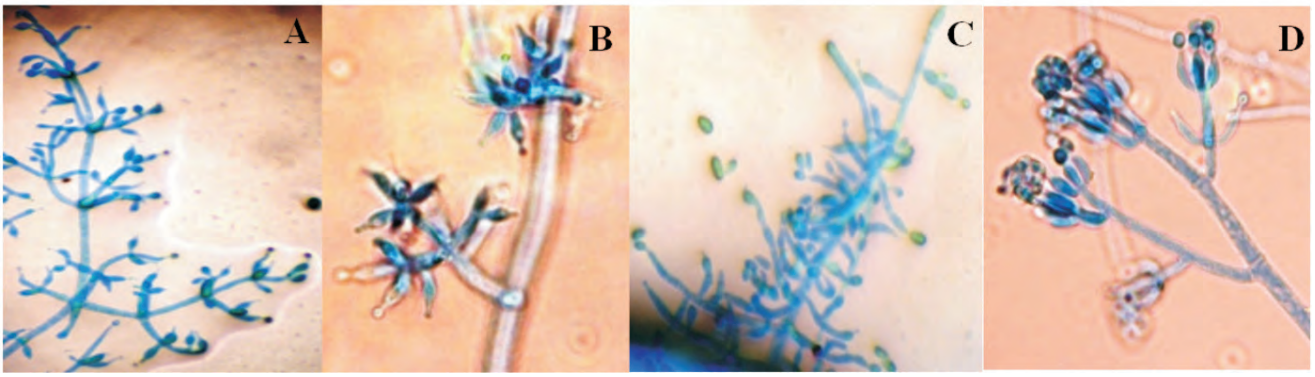

Fig. 14. Morphology of different species of *Trichoderma*. *A: T. asperellum; B: T. harzianum; C: T. longibrachiatum* and D: *T. virens*

characters revealed the major difference between the isolates of *T. harzianum* and *T. viride.* The cluster analysis from RAPD and ISSR markers showed good comparison of ISSR marker with morphological marker than RAPD. ITS1 and ITS2 nucleotide sequence analysis compared with 20 other GenBank accessions of *T. harzianum* and *T. viride* facilitated molecular identification and genetic cataloguing of twelve *Trichoderma* isolates into two groups i.e. *T. harzianum* and *T. viride.* An attempt was made to artificially induce Hypocrea in laboratory under different growth conditions by crossing twenty nine intraspecific crosses among the isolates of *T. harzianum* and *T. viride* and observed for the presence of fruiting bodies. The morphological characters made the anamorph of Hypocrea to closely relate with Tv12 isolate. The pair wise similarity index also showed more than 90% similarity between them (Rajesh *et al.,* 2013).

Among twenty nine different biocontrol agents tested against sheath blight pathogen of rice *Trichoderma koningii* 5201 showed maximum inhibition of radial colony growth, sclerotial formation, sclerotial size and their germination. In combination studies the treatment of *Trichoderma koningii* 5201 + *Chaetomiumcochliodes* 3319 showed maximum inhibition of radial growth, sclerotial formation, sclerotial size and germination. In glasshouse experiments, pre-inoculation foliar spray of *T. Koningii* 5201 + *C. Cochliodes* 3319 showed maximum reduction in infected tillers, relative lesion height and disease severity. In field experiments, pre-inoculation foliar spray of *T. Koningii* 5201 + *C. Cochliodes* 3319 showed maximum reduction in infected tillers, relative lesion height and disease severity. Further, the plots treated with the above treatment showed maximum quantity of 1000-grain weight, grain yield/plot, and straw yield.

1, 4-β-D-glucan cellobiohydrolases gene (*cbh*) was amplified from the genomic DNA of *Trichoderma harzianum* and *T. reesei* isolates which yielded a 500bp product. Purified PCR products were ligated to pGEMT cloning vector, transferred to *E. coli* DH5α separately and both the strands of the insert in this plasmid were sequenced. BLAST analysis of the *cbh-1* sequences from *Trichoderma* species under study has three close homologues: *cbh-1* of *Hypocrea lixii*, *cbh-1* of A-25 strain of *Hypocrea lixii*. It also showed homology with conserved domain of Glycosyl Hydrolases family. Comparison of amino acid sequences of *cbh-1* gene of both *T. harzianum* and *T. reesei* shows that the resultant sequence of *T. harzianum cbh-1* can be translated to 133 amino acids and that from *T. reesei* has 124 amino acid sequences and has 75% sequence similarity. When compared with the *T. viride*homologs, it shows that there were 99.19 homology with *T. Reesei* and 73.6 homology with *T. harzianum*.

Fifty one isolates of *Trichoderma* (23 isolates of *T. virens* and 28 isolates of *T. harzianum*) were morphologically characterised and used to screen some promising bio-control agents against soil borne pathogens. These isolates were made into different groups based on three bio-efficacy tests (dual culture, volatile and non-volatile). Eight high potential and two low potential isolates from *T. virens* and 12 high potential and 2 low potential isolates from *T. harzianum* were selected as promising isolates against three different soil-borne pathogens *viz., Fusarium oxysporum*, *Rhizoctonia solani* and *Sclerotium rolfsii* (Srinivasa and Prameela Devi, 2014).

Phyllosphere microbiomes in relation to rice blast was analysed using next generation sequencing approaches. Sequence reads of conserved ribosomal DNA generated in Pyrosequencing of metagenome of genotypes led to identification of 9207 and 3199 species with Shannon diversity index of 6.78 and 2.79 respectively for susceptible and resistant genotypes. The susceptible genotype harboured significantly more bacterial communities than resistant cultivar. Bacterial species belonging to phylum Proteobacteria was found to be most dominating over other group of bacteria. Quantum of microbial community and its composition significantly varied between the collection sites. The major bacterial species encountered in blast endemic field are *Microbacterium testaceum* and *Pantoea ananatis* whereas *Brevundimonas subvibrioides* and *Comamonas testosteroni* were found associated with leaf grown in climate chamber.

Integrated disease management

Pattern of Sheath blight *Rhizoctonia solani* Kuhn disease development in rice cultivars was evaluated by determining degree of vertical and horizontal spread in terms of relative lesion height and percent infected tillers respectively. Six elite rice cultivars viz. Swarna, Pusa Basmati 1, Pusa 1121, Tetep,Vikramarya and Pankaj were inoculated with Kapurthala (RS-1), New Delhi (RS-2) and Moga (RS-3) isolates in open field pot experiments. Swarna was found to be most susceptible with maximum relative lesion height and cultivar, Tetep the most resistant, Pusa Basmati 1 and Pusa1121 were either susceptible or moderately susceptible. Vikramarya and Pankaj were either resistant or moderately resistant. Isolate RS-1 was found to be the most virulent isolate.

Three susceptible (PRR78, Pusa1460 and Pusa6B) and one resistant parent (Tetep) along with 186 Tetep derived blast resistant Basmati rice breeding lines (BC_1F_4 and BC_2F_5)were evaluated against sheath blight under field conditions. The significant difference in resistance level was observed between the parental lines. Tetep was resistant (grade 1), while Basmati entries Pusa1460, Pusa6B and PRR78 were susceptible (grade 5-7). Among the Tetep derived blast resistant Basmati genotypes, thirteen BC_1F_4 lines from the cross Pusa6B x Tetep were resistant, while, eleven BC_2F_5 lines from the cross Pusa1460 x Tetep and twelve BC_2F_5 lines from the cross PRR78 x Tetep exhibited only moderate level of resistance (grade 3). Nine homozygous lines derived from the cross Pusa6B x Tetep and seven homozygous lines derived from the cross Pusa1460 x Tetep were identified to possess both, blast resistance gene *Pi54* as well as the major sheath blight QTL *qSBR11-1*. Finally, field evaluation data and QTL analysis revealed that only nine Tetep derived blast resistant Basmati lines in the background of Pusa6B were resistant to sheath blight disease.

A set of 102 rice lines were evaluated for blast as well as sheath blight resistance. None of the rice genotypes was resistant to sheath blight. However, four entries were observed to possess high degree of tolerance to sheath blight and were validated for major sheath blight QTL (*qSBR11*-1). When the same set of 102 rice genotypes were screened for blast resistance with *M. Oryzae* isolate (Mo-ni-0066) from Meerut, 32 entries were identified phenotypically resistant. These entries were subjected to molecular validation for the presence of five major *R*-genes, *viz.*, *Piz5/ Pi9*, *Pita, Pi54, Pi1,* and *Pib*. Individually none of the entries could be observed to possess all these major *R*-genes .However some of the resistant genotypes were validated for one or more *R*-genes. Further, evaluation of these genotypes with highly virulent *M. oryzae* isolate (Mo-simandya- hr-12) from Karnataka revealed that Bhusan San and P 1490-03 were resistant to blast. Interestingly, molecular validation of these two genotypes did not show any of the above *R*-genes, indicating the possible role of some other *R*-gene/s.

Combined application of fungicide (carbendazim) with insecticide (endosulfan) was effective in controlling sheath blight. Disease severity reduced up to 71.87% with significant increase in grain yield (85.68%) with carbendazim and endosulfan treatment. This was followed by hexaconazole + endosulfan which reduced disease severity (67.84%) and increased grain yield up to 71.11%. Changes in peroxidase (PO), polyphenol oxidase (PAL), phenylalanine ammonialyase (PAL) and total phenols were observed in sheath blight inoculated plants sprayed with fungicides (carbendazim and hexaconazole), insecticides (endosulfan and chlorpyriphos) and herbicides (thiobencarb and butachlor) when applied singly or in combination. Significant induction of PO, PPO, PAL and Phenols were detected from 1st day up to 3rd day after spraying on rice plants inoculated with *R. solani*and thereafter the activity declined.

For the management of *Sclerotinia sclerotiorum* disease in legumes a combination of soil application with Pusa Biopellet 4G (*T. viride*), seed treatment with Pusa 5SD (*T. viride*) + carboxin (Vitavax) + *Rhizobium cicero* and foliar spray with iprodion at 40 days after sowing provided maximum seed germination, shoot and root lengths and pod yield with minimum incidence of stem rot.

In a comprehensive *in vitro* biocontrol agent efficacy testing programme seventeen fungal and six bacterial bio-control agents were screened against *A. porri* by dual plate technique. Bio-agents (Th-3, Th-30, Tv-12, Tv-15, Psf 3 and Bs-1) were selected and the mode of action was studied by means of volatile and non-volatile compounds production and vigour index under *in vitro*. Th-3 isolate showed high level of disease reduction, growth promotion when subjected to seed treatment, seedling dip and three foliar spray of onion bulb crop under glasshouse and field conditions. Th-3 isolate could also reduce disease incidence on the flower stalk and paved the way in increasing onion seed yield under *in vivo* studies.

For the management of bakanae disease various fungicides and biocontrol agent were evaluated under *in vitro*, glass house and field conditions. Among seven fungicides evaluated, maximum disease inhibition was observed in Propiconazole treated seedlings (67.60%) followed by Contaf plus (39.79%) and Flubendiamide (32.22%) under *in vitro*, glasshouse as well as field conditions. Among biocontrol agents, maximum disease inhibition was observed for *Talaromyces flavus* treated seedling (46.96%) followed by *Trichoderma harzianum* (44.97%) and *Cheatomium globossum* (44.83%) under laboratory as well as field conditions.

A study was conducted to know the effect of two formulations of hexaconazole *viz.*, commercial and newly synthesized nano-hexaconazole molecule on phenotype, phytotoxin and ergosterol biosynthesis of *R. solani* f. sp. *sasakii*. A total of 58 isolates belonging to maize, rice, moong and pigeon pea collected from different parts of the country were subjected to pathogenicity analysis. Among all isolates, two isolates, M25 and M16 of *R. solani* f. sp. *sasakii* were superior in percent disease index, relative infected area parameters. The effect of nano-hexaconazole on phenotypic characters of *R. solani* f. sp. *sasakii* M25 and M16 isolates revealed higher radial growth inhibition, delayed sclerotia formation, reduced number and weight of sclerotia, decreased infectivity of nano-hexaconazole exposed fungi on host when compared with commercial-hexaconazole. *In vitro* bioassay of phytotoxin collected from nano-hexaconazole amended growth medium produced reduced necrotic lesion on the leaves of susceptible maize cultivar. Tissue necrosis due to phytotoxin was also observed on different host species *viz.*, barley, wheat, rice, tomato, onion, field bean, citrus and pomegranate apart from maize. Infrared (IR) spectrum of phytotoxin resulted in possible presence of lactone, ketone and benzene groups in the crude phytotoxin. The HPLC study revealed less phytotoxin in the pathogen exposed to nano-hexaconazole than in commercial-hexaconazole. Biosynthesis of ergosterol was also less in nano-hexaconazole treated *R. solani* f. sp. *sasakii*.

In order to screen maize genotypes under field condition for resistance to PFSR disease, tooth pick method has been used for creating artificial epiphytotics. In this study, thirty four maize inbred lines were screened in field by toothpick method of inoculation. Under laboratory condition, twenty maize genotypes were screened by two new methods of inoculation of the cut stems. Split stems were inoculated in the first method and un-split stems were inoculated by toothpick method in the second. The split method produced recordable data within fifteen days of post inoculation (DPI) where as the un-split stem inoculation method produced the result at 20 DPI. Both the new techniques employed in the laboratory were faster in producing results as compared to the field screening of maize genotypes by the standard toothpick method. Split open method was better than the un-split method of cut stem inoculation *in vitro*. A new scale ranging from 1 to 16 cm was developed based on the existing 1-9 scale of PFSR for rating of the disease severity *in vitro*.

Studies were conducted to find out on the correlation of disease cycle and pathogen. In artificial inoculation studies, the typical symptoms of rice false smut disease were produced on the inoculated rice cultivar (PRH-10) by injecting 2ml of conidial and mycelia bit suspension @ $2x10^5$cfu/ml in leaf sheath at booting stage. Spraying spore suspension on the inflorescence was found to be successful in producing disease symptoms on artificially inoculated rice cultivars (Pusa rice hybrid-10, Pusa Sugandh-2 and Pusa Sugandh-3). Transmission electron

microscopy (TEM) examination of the ultra thin section of the samples revealed the presence of compact and orderly arranged hyphae of *U. virens* filled with fat bodies. It was observed that the host cells were degraded but their cell walls were intact.

Disease epidemiology and forecasting

Spot blotch (*Bipolaris sorokiniana*) once endemic to eastern Gangetic plains has now emerged as major disease in wheat throughout Indo-Gangetic plains. Criteria for spot blotch infection and developmental rate model has been worked out to explain spot blotch development in wheat under climate change scenario. Number of spot /cm^2 leaf (infection index) has been modelled in relation to temperature and duration of relative humidity 95% or above (RH-duration). The surface response in relation to temperature (*T*) and RH-duration (*D*) which could explain variation in infection index and thus proposed as a model: $Y = \left(\frac{36-T}{7}\right) \times \left(\frac{T-16}{13}\right)^{2.6} \times (1 - 0.8114^{D-m})$ where *m* is dependent on *T*. Temperature response on incubation period (IP_{20}) has been estimated and hourly IP_{20} rate was established as *(0.002 Temp –0.03) {1 – exp[0.151(Temp – 36)]}* which could reasonably explain the rate of spot development tested under natural infection in the field. Increase in lesion size, infected area/cm^2/leaf and decrease in incubation and latent period were noted under combined elevated CO^2 (450-550 ppm) and temperature (+1.5°C) level. Based on spot blotch favorable hours and rate for IP_{20} completion under current situation indicated that eastern region of Indo-Gangetic plains is relatively more favorable during February-March than western region. Addition of temperature (1.5°C) above the ambient temperature has shown to increase favorable hours and rate of incubation period completion during February and March. Based on the distribution of favorable periods and developmental rate, it indicated that likelihood of spot blotch has further increased in eastern plains whereas in western plains borderline risk for the disease has been crossed. A criterion for monitoring of infection favorable period and a developmental rate has been proposed to develop a spot blotch forecasting system for scheduling protective fungicide applications as the severity of the disease is likely to increase further under climate change scenario.

For detection and monitoring of *Mungbean Yellow Mosaic India Virus* (MYMIV) in soybean spectral leaf reflectance properties of were investigated over the wavelength range 350-2500 nm. Due to MYIMV infection reflectance increased in visible range (VIS, 350-740 nm) and decreased in near infra red (NIR, 750-1400 m). Significant reflectance difference between infected and healthy leaves has been in green and red region (560-691nm) and 750 nm of NIR region ($p>0.5$). However, sensitivity analysis has indicated wavelength 622, 688 in red region and 750 in NIR regions are most variable region occurs due to infection. Relationship between WMI and LAI and wavelength of the red peak (λ_{re}) and amplitude (dr_{re}) and also area of red edge peak ($\sum$ (dr 680-780)) indicated predictability of MYIMV infection in terms of these parameters. Reflectance at 622, 688 and 750 nm have been found to increase as the mosaic severity increased with significant slope coefficients ($p<0.001$) and coefficient of determination were highest for regression between R688 and mosaic severity. Ratios calculated as R750/R445 and R (750-445)/ (750+445) has been found to decrease with increase in mosaic severity. Most effective spectral reflectance ratio or index for estimating per cent yield loss in soybean due to MYMV infection was (R750 – R445)/(R750 + R445). Therefore, red edge parameters as well as spectral indices could serve as good indicator for monitoring of stress caused by MYMIV infection in soybean.

Disease progression studies under field conditions of black leaf spot caused by *Alternaria brassicae* (Berk.) Sacc. revealed that there was periodical increase in lesion number and Per cent Disease Index (PDI). PDI progression was higher in mustard varieties followed by cauliflower and cabbage varieties. The speed of progression of disease among the crop cultivars was calculated by using Area Under Disease Progress Curve (AUDPC) and apparent rate of infection (*r*-value). Highest AUDPC value was recorded on Pusa Bold variety of mustard and the lowest was recorded on CJ-182 variety of cabbage. This may enable to select the slow disease progressing varieties for the management of the disease. Relative humidity was found negatively correlated with the development of disease. On the contrary, disease development was positively correlated with maximum and minimum temperature.

An attempt was made to eliminate garlic viruses through meristem culture after thermotherapy.G-1, most commonly grown garlic cultivar was found to be infected with potyviruses (*Onion yellow dwarf virus*, OYDV; *Leek yellow stripe virus*, LYSV), carlaviruses (GarCLV; *Shallot latent virus*, SLV) and *Allexivirus* (*Garlic virus* X, GarVX). Solar heat treatment resulted in lower survival and regeneration of plants, but successfully eliminated all the viruses in 66% of the regenerated plants after 10 days treatment (Pramesh and Baranwal, 2015).

The coat protein mediated resistance (CPMR) strategy has shown promise in Hawaii (USA) in managing PRSV and thus explored One CP-based construct (pBI121:PRSV-FullCP) and two marker-free hairpin constructs using 341bp CP (pCAM:341CP:hp:MF) & 310bp HC-pro (pCAM:310HC:hp:MF) were used in papaya (cvs. Pusa Delicious) transformation through *Agrobacterium* using somatic embryos as explants. Putative transformants derived from pBI121:PRSV-FullCP construct (two), pCAM:341CP:hp:MF (24) and pCAM:310HC:hp:MF (5) constructs were subjected and found positive through molecular evaluation based on PCR and Southern hybridization.

The full length Sup gene of TK1 isolate of *Citrus trizteza virus* was selected for development of gene construct in antisense orientation in pBinAR vector. The construct was mobilized to *Agrobacterium* cell EHA105. The regeneration protocol of Kagzilime (*Citrus aurantifolia*) was standardized using 0.75-1.0 cm long epicotyl segments of *in vitro* raised 21-28 days old seedlings. Total number of 30 plants was initially regenerated in selective medium and finally, four putative transgenic plants were selected.

Acknowledgement

We wish to thank all the Faculty Members of the Division of Plant Pathology for extending support and cooperation in preparing the research contributions of M.Sc. and Ph.D. students made during the period of 2007-16.

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School of Natural Resource Management

Agricultural Engineering

D.K. Singh, Indra Mani, Roaf Ahmad Parray and Satish D. Lande

Introduction

The major challenges before Indian agriculture are to increase production and productivity to meet the food requirement of burgeoning population while maintaining sustainability without inflicting harm to environment. In the backdrop of increasing labour scarcity, declining surface and water resource availability for irrigation, post harvest losses, abiotic stresses on India's agricultural system due to climate change; the role of Agricultural Engineers will be of paramount importance to ensure sustainable and climate-resilient agriculture for ensuring food security and profitability. The same can be achieved by use of precision farm machines, efficient irrigation and soil & water management technologies, appropriate post harvest technologies and smart storage facility. *"According to Forbes magazine, climate change, shrinking land availability to farming, population growth, demand for higher-priced, better tasting foods will put more pressure on a straining agricultural system and will lead to the rise of Agricultural Engineering - a profession focused on developing better and more sustainable ways to grow food to meet the needs of the world (Knapp , 2012)."*

Ever since its establishment in the year 1945, Division of Agricultural Engineering has been striving to address the problems faced by Indian farmers. In the year 1966, Division started postgraduate teaching programme. The Division has been offering M.Tech. and Ph.D. in three sub-disciplines namely Soil and Water Conservation Engineering, Farm Power and Equipment, and Agricultural Processing and Structures. Post graduate research of the Division includes all important aspects such as soil and water conservation, ground water development and management, irrigation and drainage, micro-irrigation, rainwater harvesting, climate change impacts, hydrology, design and development of modern farm machinery, precision agriculture machines, application of drone and robotics in agriculture, machines for horticultural mechanization, processing for value addition, technologies for efficient biomass utilization and use of solar energy in agriculture.

Significant Student's Research Achievements

Researches conducted by the students under the able guidance of faculty members have contributed significantly in the research outputs of the Division. The research Contributions made by the students categorized under various themes are presented in following paragraphs.

Management of Surface and Groundwater Resources

A study was conducted in semi-arid region to simulate the recharge processes. All the important processes which control the groundwater recharge in semi-arid region such as rainfall, infiltration, evapotranspiration, soil moisture storage in vadose zone and surface runoff were considered to estimate the recharge rate at water table and total recharge using HYDRUS-1D and MODFLOW models. The average numbers of rainy days observed in the last 37 years were 26. Number of consecutive dry spells of 7 and 14 days were 4 and 1 respectively in the monsoon season of year 2008. The long term average of consecutive dry spells of 7 and 14 days were 7 and 3 respectively. Consecutive dry spells exhibited increasing trend. The average surface runoff estimated from IARI campus using Curve Number method was 90.6 mm, which is about 13% of the total rainfall. Simulation

results revealed that maximum and minimum daily recharge rates in various sub areas varied from 1.4 cm/day to 2.5 cm/day and 0.01 cm/day to 0.05 cm/day respectively. Cumulative recharge at water table (cumulative flux) in various sub areas varied from 20.01 cm to 23.43 cm. Cumulative recharge was found to be varying from 29 to 34% of monsoon rainfall. Total surface runoff in various sub areas varied from 3.39 cm to 14.36 cm. Reference evapotranspiration was estimated to be 41.6 cm. Natural recharge under prevailing pumping rate and pumping schedule was 0.99 m which was 23.3% of monsoon rainfall of year 2008. Average rise in water table under artificial recharge conditions varied from 1.43 m to 1.54 m, provided that all the surface runoff is used to recharge the aquifer. Average rise in water table under increased daily pumping rate varied from 0.90 m to 0.96 m which was lower than the natural recharge with prevailing pumping rate and schedule (Anilkumar, 2011, Dandekar *et al.,* 2011 Anilkumar *et al.,* 2013).

Seawater intrusion in coastal aquifer of Minjur was modeled to suggest the options for managing the this aquifer (Fig. 1). Groundwater salinity and water level measured through observation wells and piezometers were subjected to geo statistical analysis to study the spatio-temporal variability of sea water intrusion for delineating the affected zone in coastal aquifer of Minjur. The variograms and krigged spatial maps were generated for pre monsoon and post monsoon seasons of 1999 and 2008. The variogram analysis of salinity showed a nugget to sill ratio of less than 0.25, indicating that the groundwater salinity has very strong spatial structure. The average ranges of variograms for spatial and temporal analysis were about 11.28 km and 10 years, respectively. Spatial analysis revealed that the area where EC exceeded 3.0 dS m^{-1} increased from 37.8 % in 1999 to 56.7 % in 2008 in unconfined aquifer. In semi confined aquifer, the area with EC more than 3.0 dSm^{-1} increased marginally from 62.2 % in 1999 to 63.3% in 2008. Groundwater quality improved during post monsoon in both the aquifers. However, improvement in groundwater quality was more in unconfined aquifer. Groundwater models MODFLOW and MT3D were used to simulate seawater intrusion and groundwater salinity in affected area under various management scenarios (Fig. 2). Simulations were done to predict the seawater intrusion and groundwater quality in the year 2025 starting from 2009. Simulation results showed that if the groundwater pumping and recharge rates prevailed in 2008 were continued, the severely affected area would extend to 11.25 km and 12.0 km away from coast in unconfined and semi confined aquifers respectively compared to 10.50 km and 11.25 km in 2008. Simulation results also showed that seawater intrusion can be controlled to varying levels by increasing recharge, reducing groundwater pumping and relocation of pumping. Combination of management options namely reduction in ground water pumping by 25% from unconfined aquifer, increase in pumping by 25% from semi confined aquifer, increased recharge

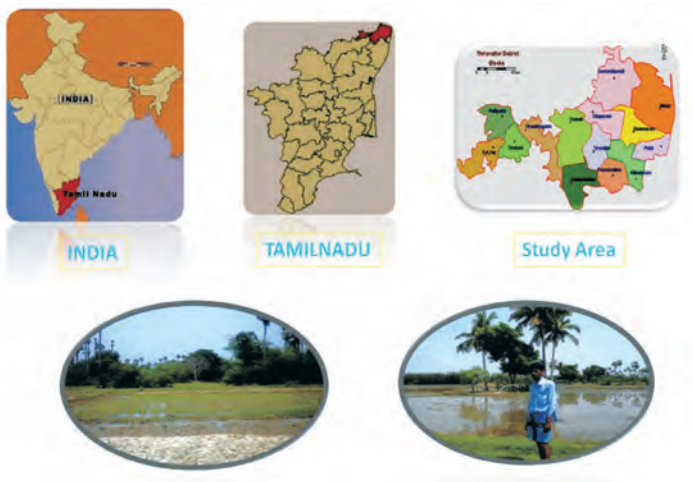

Fig 1. Aquifer of Minjur in Tamil Nadu

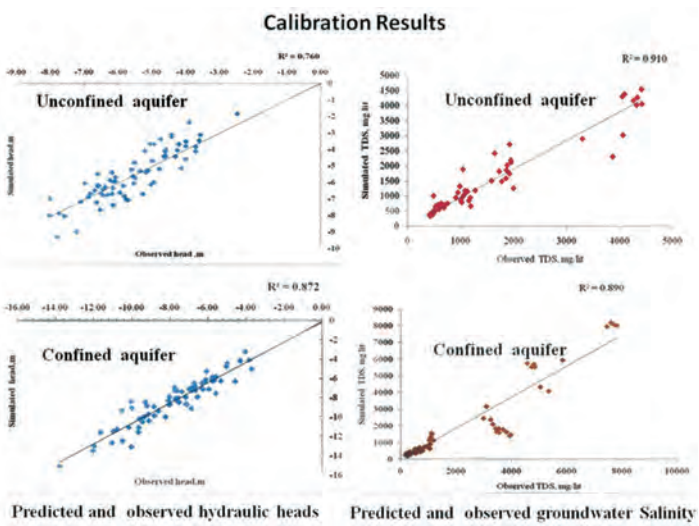

Fig 2. Calibration and validation results

by maintaining stage in river to 0.5 m during monsoon and 0.25 m during non monsoon have potential to control seawater intrusion and improve groundwater quality in the aquifers of Minjur (Mini, 2012, Mini *et al.*, 2014, Mini *et al.,* 2015, Mini *et al.,* 2015).

A study was conducted in Arvari River catchment in Alwar district of Rajasthan state of India to quantify the hydrologic impacts of construction of large scale Rain Water Harvesting Structures (RWHS) for assessment of water resource potential for future utilization in agriculture. It was found that 109.3 MCM water could be harvested from the entire catchment but only 3.78 MCM water could be harvested from the existing RWHS. The spatial variability of decadal ground water table fluctuation ranged between 5.67 to 33.6 m bgl during 1982-1991. While the upper and middle reaches of the catchment have shown a rise in the water table the lower reaches have shown a constant decline in the water table of approximately 2 m during 1992-2001 over the preceding decade. In 2002-2012 however, in the SW side of the catchment a sudden drop of 54 m was observed as a result of combination of factors. Larger recharge (R_{ep}) volumes during 2012 (>30 % as compared to 2011) could be attributed to a 20 % increased amount of rainfall in 2012 as compared to 2011. Some structures could retain water up to 273 days indicates that there was a great possibility of utilizing harvested surface runoff. After the detailed survey and study it was proposed to construct 10 numbers of rockfill dams, 11 check dams and one small reservoir to harvest the excess runoff from the catchment. (Mahato, 2013)

An effort was made to link Hydrus-1D model with the GIS to assess the nitrate leaching through the vadose zone in cropped regions. An interface of Hydrus-1D with ArcGIS was developed and simulated for experimental blocks of IARI farm under rice, wheat and maize crops. The model was calibrated up to 120 cm depth (0-30, 30-60, 60-90, and 90-120 cm) using the observed data of soil water and nitrate content of one cropping season. The model predicted soil moisture flux moving up to 180 cm and 270 cm below ground surface during *rabi* and *kharif* season, respectively. Nitrate movement in the vadose zone was up to 180 cm for both wheat and maize during both *rabi* and *kharif* seasons amounting to 10.8 and 16.0 kg ha^{-1}, respectively. In rice, nitrate leaching below 120 cm was observed to be 28.5 kg ha^{-1}. Further, the nitrate leaching to groundwater was predicted to be 6.4, 7.9 and 9.1 mg l^{-1} during one crop growing season with prediction error below ± 20 percent and model efficiency (*E*) of 0.88. Further, the nitrate leaching scenarios to groundwater generated with fertilizer application rates exceeding the optimal dose up to 50 percent showed an increasing trend fitted with second order polynomial equation ($0.96 \leq R^2 \leq 0.99$). (Dash *et* 2011, Dash, 2012, Dash *et al.,* 2012, Dash *et al.,* 2015, Dash *et al.,* 2015, Dash *et al.,* 2015, Dash *et al.,* 2016,).

Performance of filtration unit of groundwater recharge shaft was evaluated for suggesting appropriate filtration unit for recharging groundwater. The effect of variable thickness of coarse sand (CS), gravel (G) and pebble (P) layers of the filtration unit on the recharge rate and the sediment concentration of effluent water was evaluated. An experiment was carried out with laboratory-scale models having varying depths of CS, G and P layers in five different thickness combinations, *viz.* 1: 1.5: 3, 1.5: 1: 3, 3: 1: 1.5, 3: 1.5: 1, 1: 1: 1 (CS: G: P). These models were operated with six different treatments having varying concentrations of turbid water, similar to the sedimentation level of the surface run-off ranging from 6 to 16 g/l. It was observed that higher thickness of CS resulted in reduction of the recharge rate, but improved the filtration of the effluent. Overall, considering both the recharge rate and sediment concentration of the effluent, the filtration layer thickness ratio of 1. 5: 1: 3 (CS: G: P) would be the optimal design of the filtration unit to facilitate higher recharge and perform better filtration of the turbid water (Kambale, 2007, Kambale *et al.,* 2009).

Climate Change, Irrigation and CO_2 emission

A study was undertaken to evaluate the trend and predict the changes in climate parameters, assess the impact of climate change on crop water requirement and groundwater availability for irrigation under various climate change scenarios and suggest coping strategies to climate change impacts on irrigated crops in an agriculturally dominant area under National Capital Territory of Delhi. Time series weather data was analysed using Mann-Kendall test. Auto Regressive Integrated Moving Average (ARIMA) model was used for forecasting the future

climate trend. One dimensional variably saturated flow model HYDRUS-1D and groundwater flow model MODFLOW were calibrated and used to simulate the impact of climate change on groundwater recharge and availability. Scenarios considered for assessment were based on ARIMA, Indian Network for Climate Change Assessment (INCCA) and Inter-Governmental Panel for Climate Change (IPCC) predictions. Analysis of time series weather data indicate that average annual temperature and relative humidity increased marginally during 1975 to 2010. However, wind speed, sunshine hours and rainfall decreased during this period. Results also indicate that crop water requirement did not increase if it was estimated using all important climatic parameters even though the average temperature increased during this periods. However, if only rise in temperature was considered, crop water requirement increased under all scenarios considered in the study. The simulation results suggest that average groundwater recharge in 2030s may increase marginally (0.03 m) compared to the recharge in 2005 in case of scenario based on ARIMA predictions. However, in case of groundwater recharge estimates based on IPCC and INCCA predictions, the groundwater recharge may decrease by 0.09 m to 0.21 m and 0.11 m, respectively with respect to groundwater recharge in the year 2005.The results indicated decrease in groundwater recharge with increase in rainfall intensity. Based on water requirement and ground water availability under various climate change scenarios, appropriate coping strategies to climate change impact on irrigated crops have been suggested (Kambale, 2012, Kambale et al 2015, Kambale *et al.,* 2016).

A study was under taken to analyse the climate variability and groundwater level fluctuation trends, simulate the climate induced water level fluctuations, and assess the carbon foot print of groundwater irrigation in Karnal district of Haryana for suggesting appropriate mitigation strategies. Mann-Kendall test and Sen's slope estimator were used for trend analysis of climatic parameters, potential evapotranspiration and groundwater levels. ARIMA model was used for time series modelling and forecasting of pre and post-monsoon groundwater levels. HYDRUS-1D and MODFLOW were used to simulate the impact of climate change on groundwater fluctuations. During 1981-2011, the mean daily minimum and mean daily temperature increased by 0.02^{0}C/yr and 0.01^{0}C/yr, respectively (Fig. 3). Mean daily maximum temperature remained constant. Mean daily relative humidity and wind speed increased by 0.11 % per year and 0.57 km/yr, respectively. Mean daily sunshine hours and reference evapotranspiration decreased by 0.06 h/yr and 0.01mm/yr, respectively. During 1974 to 2010 average rates of water table decline in pre-monsoon and pos-monsoon season were 0.23 m/yr and 0.27 m/yr, respectively. Pre and post-monsoon groundwater levels in 2050 would decline by 12.97 m and 12.00 m, respectively with respect to 2010. Groundwater recharge would increase by 0.25 m over the baseline recharge under the scenario based on ARIMA predictions which considered the effect of all climatic parameters. However, under the scenarios based on INCCA and IPCC predictions which considered only rise in temperature, groundwater recharge would reduce by 0.06 to 0.07 m and 0.16 to 0.22 m, respectively. Rise in temperature by 3.5^{0}C and 4.3^{0}C along with 9% and 16% increase in rainfall over the base year would increase the recharge by 0.09 m and 0.14 m, respectively. CO_2 emission were estimated using the procedure presented in Fig. 4. Study revealed that the carbon foot print of groundwater irrigation for baseline scenario was highest for sugarcane (93.15 kgCO_2/ha/m) followed by rice (40.39 kgCO_2/ha/m), wheat (27.71kgCO_2/ha/m), mustard (26.16 kgCO_2/ha/m), pigeon pea (13.49 kgCO_2/ha/m) and pearl millet (3.99 kgCO_2/ha/m). However, total CO_2 emission under baseline scenario was highest for rice (140655.97 mt)

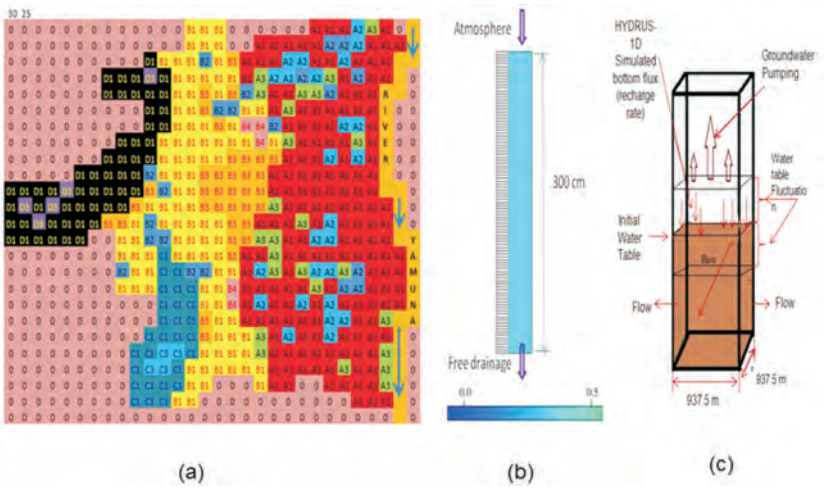

Fig 3: (a, b, c) Conceptual framework for simulation with MODFLOW

followed by wheat (98153.48 mt) and sugarcane (18415.98 mt). Emission of CO_2 can be reduced by 31.96% through improvement in pump efficiency from 34.7 to 51%. Improvement in irrigation efficiency by 15% over the baseline efficiency would reduce CO_2 by 23.06% in rice and 24.99 % in other crops. By improving the pumpset and irrigation efficiencies together up to the achievable level, CO_2 emissions can be reduced up to 48.35% (Patle, 2013, Patle et al 2013, Patle et al 2015, Patle et al 2016, Patle *et al.,* 2016).

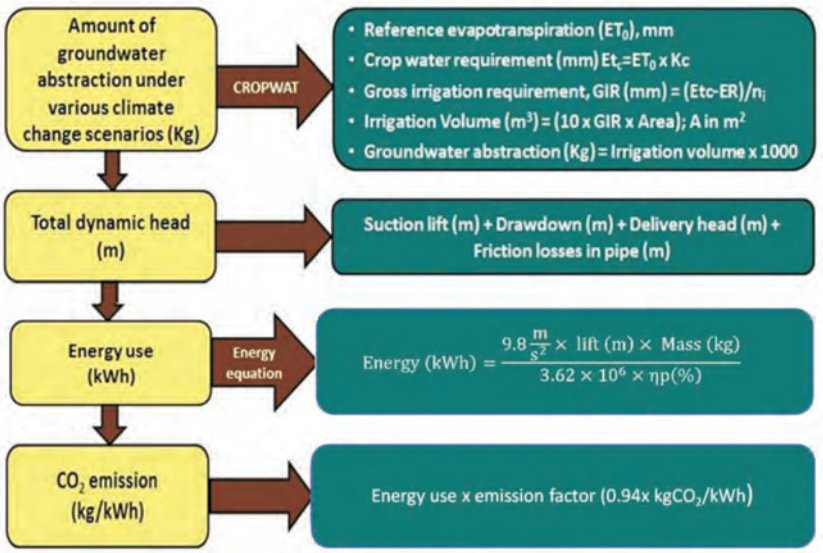

Fig. 4: Steps for assessment of carbon foot print

Impact of climate change on drought occurrences in Pune district, Maharashtra, India was investigated using the climatic parameters of Pune district from 1958 to 2004. The interface for 'Climate Change Trend Analysis (CCTA)' was developed in MATLAB® to analyze the trends in annual, seasonal (*kharif*) and monthly rainfall depths during *kharif* season rainfall using non-parametric Mann-Kendall (MK) and modified Mann-Kendall (MMK) trend test with the Sen's slope estimator. It was observed that, the rainfall during *kharif* had been decreasing**,** June rainfall had been increasing while July rainfall had been decreasing at 99, 95 and 90% confidence level, respectively from 1958 to 2004. The changing rainfall trend during monsoon months necessitates judicious management of inputs for rainfed agriculture in Pune district. Further, a newly conceptualized index named as Standardized Effective Rainfall and Crop Evapotranspiration Index (SERCEI)" was developed for agricultural drought assessment which uses the climatic water balance derived from effective rainfall and crop evapotranspiration. The developed drought index SERCEI was validated for two rainfed crops *i.e.* pearl millet and groundnut grown in the Pune district. Yield deviation (%) from average and the SERCEI values for September resulted in correlation coefficient (r) of 0.61 for Pearl millet and 0.72 for Groundnut. The SERCEI analysis for ClimGen generated future data set with climate change adjustment through GCM anomalies predicted more droughts as compared to the series generated without climate change adjustments. The results indicated that climate change will largely affect drought duration, magnitude and frequency in the study region and subsequently the availability of future water resources for agricultural production (Patil, 2013, Patil *et al.,* 2013).

Irrigation Water Management in Canal Command

Water Delivery Schedule for Jhajjar distributary in the Western Yamuna Canal Command of Haryana was developed using Geospatial Tools and CROPWAT model. The geo-database of different soil, water and crop parameters in the command area were developed and analysed to estimate the irrigation water requirement of rice and wheat crops grown on the delineated rice-wheat cropping system encompassing twelve outlets of the distributary. Further, the comparison of canal release roster of the Jhajjar water services division with the crop water demand based delivery schedule developed in this study showed that about 74.5% of total command area located in the middle and tail reaches are facing water deficit during the crop growth season. The outlets at the head reaches of the distributary received canal supply exceeding the crop water requirement, which results in waterlogging problem in the region. Besides this the ground water use scenarios in conjunction with the canal water supply to meet this deficit was also prepared for all the outlets of the distributary (Vibhute 2012, Vibhute, 2016)

A study was undertaken to design balancing reservoir in canal command for micro irrigation at Sirsa district, Haryana, India. Water balancing reservoir stored either canal water or tube well water or both to reduce the gap between demand and supply of water nearby areas. The deficit of demand can be replenished by adopting water efficient micro irrigation system at the outlets of water balancing reservoir. The chemical properties of canal water revealed that direct application of canal water is safe for irrigation while the chemical properties of ground water revealed that direct application of ground water is not safe for irrigation. The ratio of 1:2 (1GW: 2CW) was suggested for irrigation. In view of poor quality of ground water and uneven supply of canal water, drip irrigation through balancing reservoir can be considered as an feasible option (Pawan Jeet, 2013)

Enhancing water use efficiency through fertigation, mulching and irrigation scheduling

Dynamics of Moisture and Nutrients Distribution under Subsurface Drip Irrigation was studied through field experiment and by use of Hydrus-2D model . It was found that the downward movement of water in sandy-loam soil was more because of predominant role of the gravity, therefore shallow depth (10 cm) of placement of drip tape and drip lateral was recommended in potato and onion crop to get higher yield. Maximum yield of potato (33.6 t /ha) and onion (25.7 t/ ha) were obtained by applying 23.6 and 60.7 cm of irrigation water and by placing the drip tape and drip lateral at 10 cm soil depth in potato and onion, respectively. The risk of NO_3-N leaching can be reduced by adopting the subsurface drip irrigation. NO_3-N availability was more in the upper soil (0-15 cm soil depth) when drip lateral was placed at 5, 10 and 15 cm reducing the chances of leaching beyond the root zone of crop (Patel, 2007, Patel and Rajput, 2008).

A Fuzzy neuro model for drip irrigation scheduling of greenhouse cut flower rose was developed. Field experiments under greenhouse condition was done to evaluate the best treatment and collect information on weather, soil and plant parameters. Adaptive neuro fuzzy inference system (ANFIS) was used for development of fuzzy neuro model, which integrated Sugeno type fuzzy inference system and neural network architecture. The results obtained showed that highest yield (48 flowers/plant/year) was recorded in treatment with 100% crop evapotranspiration. The Sugeno type fuzzy inference system was found to be the best inference system for the fuzzy-neuro model. Three membership functions in low, medium and high classes with degree of membership ranging between 0 to 1 were developed. Four fuzzy rules were developed employing four input parameters and three membership functions. Fuzzy Neuro drip irrigation scheduling model was developed with four input parameters, four fuzzy rules, three input and output membership functions. Validation of the model showed close agreement between predicted and observed values of irrigation quantity and irrigation interval and can be used for greenhouse irrigation scheduling. Coefficient of determination (R^2) between predicted and observed irrigation quantities and model efficiency were 0.97 and 0.98, respectively. Results of the sensitivity analysis showed that model was very sensitive to leaf area, which was followed by solar radiation, temperature and humidity. Model was used for prediction of irrigation quantities and irrigation interval in different months. The predicted values of the irrigation quantities and interval varied from 3.6-8.1 m^3/day/1000m^2 and 2-5 days respectively in different months of the year (Hasan, 2007, Hasan *et al.,* 2008, Hasan *et al.,* 2009).

Soil water and salt distribution under subsurface drip irrigation in cotton crop was studied during 2009 and 2010 under loamy soil. Experimental design was strip split plot with 3 replications including different levels of saline water as main plot (EC_1= 2 dS m-1(ground water), EC2= 5 dS m^{-1}, EC_3= 8 dS m^{-1} and EC_4=11 dS m^{-1})) and different depth of laterals placement (lateral placed at soil surface, 15 cm and 30 cm below the soil surface) as sub plot. Saline water was made by mixing salts of NaCl and $CaCl_2$ in irrigation water (1:1 ratio). Composite analysis of two years showed that mean boll weight and boll number per plant were significantly different while yield and earliness percentage were not significant between two years. Saline water up to 8 dS m^{-1} did not decreased cotton yield but by applying salinity up to 11 dS m^{-1} yield decreased 32.6%, 40.3% and 39.2% as compared to EC_1, EC_2 and EC_3 treatments. Lateral depths had no effect on yield. Different levels of saline water and lateral depths had no significant effect on cotton fiber quality except micronaire. Treatments of saline water 11 dS m^{-1} and lateral buried at 15 cm had low micronaire than other treatments. The results of

soil water distribution showed that soil water content decreased by horizontally distance 20 and 40 cm away from drip line in all lateral depths. Distribution of soil moisture and electrical conductivity was simulated by Hydrus 2D model. The model was evaluated by R^2, AE, RMSE and E. There was good correspondence on soil water distribution between observed data from field experiment and simulated by model (Ghorban, 2011).

A farm pond operational model was developed using soft computing technique known as Neuro-Fuzzy modeling. It is to be used to formulate operating policies of storage reservoir at farm level. The developed model was validated statistically with new sets of input-output data set. The result of validation revealed that the developed Neuro-Fuzzy model was able to predict outflow with high accuracy. Coefficient of determination (R^2) between observed and predicted value was found to be 0.98. Results showed that the model has high accuracy and predictability (Sonawane, 2011).

Investigations were done to evaluate the effect of deficit irrigation and split dose of N-fertilizer at different growth stages of wheat on its growth and yield. Water use efficiency (WUE) and nitrogen use efficiency (NUE) were estimated. The maximum yield of wheat was obtained under full irrigation and with split-N dose as 25% basal, 25% CRI and 50% tillering during the year 2010-11 and at 25% basal, 50% CRI and 25% tillering growth stages during 2011-12. The WUE exhibited a decreasing trend with application of 25% DI and full irrigation as compared to 50% DI for both years of experiment. Moreover, the average yield loss varied from 7 to 15% at 25% DI as compared to full irrigation during both years of experiment. Results revealed that split-N doses of 25%:50%:25% and 25%:25%:50% as basal, at CRI and tillering stages resulted in higher wheat grain yield. The water use efficiency of wheat crop under deficit irrigation levels was more than that at full irrigation (Pathan, 2012).

A study was conducted to evaluate the effect of drip lateral lengths (25, 50, 75 and 100 m) and system operating pressures at 0.5, 1.0 and 1.5 kg cm-2 on the water and nitrogen distribution uniformity of a drip irrigation system. Drip-line lengths and system operating pressures significantly ($P<0.01$) affect coefficient of variation and distribution uniformity. Highest value of distribution uniformity for irrigation water and different fertilizer solutions (urea, MoP and Phosphoric acid) were observed at 1.5 kg cm-2 with 25 m long drip-line and lower values of coefficient of variation were observed at the same operating pressure and drip-line length. Growth parameters and yield of baby corn varied significantly ($P<0.05$) with fertigation frequencies whereas these were not affected significantly at different system operating pressures. All the growth parameters and yield of baby corn were recorded highest under biweekly fertigation frequency with 1.0 kg cm-2 system operating pressure (T4). Whereas, lowest values of these parameters were observed under control treatment (conventional fertilizer application with furrow irrigation). Highest yield of cob, baby corn and fodder were recorded in biweekly fertigation schedule at system operating pressure of 1.0 kg cm-2, respectively. The lowest yields of cob, baby corn and fodder were recorded in control (conventional fertilizer application and furrow irrigation). Higher values of baby corn weight, length, girth with and without husk along with total and reducing sugar of fresh baby corn were recorded under biweekly fertigation and lowest values of these were observed in control. The highest benefit cost ratio was found under treatment biweekly fertigation frequency during April-July. (Kumar, 2012, Kumar *et al* 2012, Kumar *et al* 2012).

Potassium dynamics and Fertigation scheduling using Hydrus 2D model for Kinnow was studied Based on the experiment conducted to study the effect of different levels of Fertigation on fruit yield and quality parameters, the treatment with 80% ETc and 700 g K per plant per year of was chosen as optimum dosage to increase the yield and quality of kinnow fruits. This study helped in choosing the optimum dosage of water and fertilizer which was used for developing an optimal fertigation schedule for kinnow plants (Desai, 2011).

Artificial Neural Network Model was used to predict the crop evapotranspiration. Climatic variables namely, maximum and minimum temperature, relative humidity I and relative humidity II, sunshine hours, pan evaporation, wind speed, rainfall, solar radiation and days after sowing were used as input parameters, and lysimeter measured ET as output parameter. The data partition, input parameter selection and selection

of input model strategies were carried out based on the statistical data analysis results. The ANN model was developed in MATLAB 7.0 software, which is high performance language for technical computing. It included different field of functions and toolbox, from which the neural network toolbox was used to create ANN. The performance of ANN model was observed by the statistical criteria of Coefficient of Correlation (r), Root Mean Square Error (RMSE), Mean Absolute Error (MAE) and Nash–Sutcliffe Coefficient (E). It was done for both, the development data set and the evaluation data set. To get more exposure for the developed ANN model predicted ET, the Penman-Monteith estimated ET was compared with ANN predicted ET and lysimeter measured ET. ANN predicted ET values were used for the application of irrigation scheduling of wheat crop (Nikam, 2008, Nikam *et al.,* 2010).

An experimental was conducted to estimate the bare soil evaporation from different soil types and develop bare soil evaporation prediction equations. The ambient temperatures was found to have maximum influence on bare soil evaporation in both soil types. There were marked differences in the evaporation behaviour of silty clay loam (SCL) and sandy loam (SL) soils wherein the evaporation rates were higher in SCL as compared to SL soils. Multiple linear regression models developed could be used to estimate evaporation from silty clay loam (SCL) and sandy loam (SL) soils (Ingle, 2009, Ingle *et al.,* 2013).

Drip irrigation is an efficient method of irrigation in which water supplied directly to the root zone of the plant so as to maintain the soil moisture near the field capacity of the soil for most of the time. The combination of polyhouse farming and drip irrigation can save about 20- 25% of water compared to the open drip irrigated farming system. Efficiency of irrigation in polyhouse can be enhanced by accurate estimates of evapotranspiration. Keeping this in field experiment was carried out in a naturally ventilated polyhouse to determine the effect of degree of ventilation on climatic parameters in a naturally ventilated polyhouse and to compare the evapotranspiration inside and outside the naturally ventilated polyhouse. The results revealed that evaporation under empty polyhouse condition was 51% as compared to outside the polyhouse. The evaporation in the cropped polyhouse was 31.6 % as compared to outside of the polyhouse. The evapotranspiration of the capsicum inside the polyhouse was calculated as 282 mm as compared as 408 mm of the open field condition. It was about 69.2% of the open field condition which can save about 30% of water compared to the open drip irrigated system (Balraj, 2009).

Water resources are limited worldwide for agriculture and therefore there is a need to develop and evaluate water-saving irrigation techniques. Regulated deficit irrigation (RDI) and partial root zone drying (PRD) are latest techniques used for saving irrigation water and maximize water use efficiency of different crops grown in open field and greenhouse. In this study both these techniques were evaluated for greenhouse grown tomato (Lycopersicon esculentum) with low pressure drip irrigation system. The following five irrigation treatments were tested: (1) FULL, control treatment where the full amount of irrigation water was measured using Class-A pan evaporation data, was applied uniformly on the two side of plant-root zone; (2) PRD30, (3) PRD50, 30% and 50% deficit irrigation in which wetted and partially dry sides of the root-zone were interchanged in every irrigation respectively; (4) RDI30, (5) RDI50, 30% and 50% conventional deficit irrigation maintained at 30% and 50% deficit, compared to FULL irrigation, where water applied on the both side of the root-zone respectively. The highest fruit yield (245 t/ha) was measured under FULL irrigation treatment, followed by PRD (242.72 t/ha) and RDI (236.06 t/ha) treatments respectively. Water use efficiency (WUE) was found 40-45% higher for PRD and 30-35% higher for RDI compared to FULL irrigation without much yield reduction. Therefore, PRD practice can save up to 45% of irrigation water with only marginal yield reduction in tomato. It was found that in PRD leaf area index and vegetative growth was reduced for greenhouse tomato. Also PRD, significantly improved fruit quality in terms of total soluble solids content and titratable acidity without decreasing marketable yields in comparison to fully irrigated treatments. PRD practice not only saved irrigation water but also enhanced fruit quality with marginal yield reduction. Therefore, PRD could be used for greenhouse tomato in areas where water is scarce and expensive (Randhe, 2013).

An experiment was carried out during October to March 2010-11 to study the system performance, soil water and nitrate distribution and effect of irrigation levels and fertigation frequency on yield and growth of cabbage crop. Drip irrigation system consisted pressure compensating drippers (PC) and non-pressure compensating drippers (NPC). Biweekly and weekly fertigation frequency showed no significant difference in yield of cabbage, both in PC and NPC systems. The highest yield was recorded in weekly fertigation (91.2 t ha^{-1}) followed by bi-weekly fertigation (90.3 t ha^{-1}) under pressure compensating drippers, whereas the highest yield was recorded in biweekly fertigation (73.3 t ha^{-1}) followed by weekly fertigation (61.5 t ha^{-1}) under Non-Pressure compensating drippers. It was observed that irrigation levels of 80% of ET_c was best irrigation level which saves the water without compromising the yield and weekly fertigation frequency was best frequency which saves the time in fertigating the field without compromising the yield (Gajanan, 2011)

A study was carried out in the research farm of Precision Farming Development Centre, Water Technology Centre, Indian Agricultural Research Institute, during October to March (2009-10). Cabbage (Brassica oleracea L. capitata) seedlings were transplanted during the third week of October with plant to plant and row to row spacing of 40 cm x 60 cm. Five irrigation frequencies: N1 (daily), N2 (once in 2 days), N3 (once in 3 days) and N5 (once in 5 days) were considered as main treatment and three levels of irrigation i.e. 100, 80 and 60 % of crop evapotranspiration (ETc) as the sub treatments. The water requirement of cabbage crop varies, respectively, from 1.2 to 6.9 mm day-1 from early stage to peak demand period. Soil matric potential in the range of -25 to -32 kPa at 30 cm depth and irrigation frequency of once in two days can be used as an index for drip irrigation scheduling during cabbage growth period in sandy loam soils for attaining higher yields. Drip irrigation frequency significantly affected the cabbage yield. The maximum yield (70.1 t ha-1) was obtained by applying water @ 80% of ETc once in 2 days irrigation followed by 100% of ETc at once in 3 days irrigation frequency. Maximum B-C ratio (2.3) was found in once in two days irrigation with 80% of crop Etc (Kumar 2010).

A field experiment was conducted to study the characteristics of municipal wastewater available at IARI and to investigate the feasibility of its safe use through surface and subsurface drip system. The study included the determination of water distribution pattern in soil and quality of produce in terms of heavy metal content and *E.coli*, coliforms contamination under drip irrigation. The different type of drip laterals resulted in significantly different spatial distribution of soil water in the root zone. The heavy metal contents found in the cauliflower were within the safe limit prescribed by FAO/WHO as well as the Indian standard. The maximum yield (80.44 t ha^{-1}) and Minimum curd yield (59.31 t ha^{-1}) were recorded with wastewater application under bioline drip laterals with subsurface placement and inline lateral with surface placement, respectively. Analysis of nutrients in cauliflower curd showed that all the heavy metals content was significantly higher under wastewater irrigation as compared to groundwater irrigation. Study revealed that there was no *E. coli* and total coliform detected in the cauliflower curd using groundwater in irrigation. The presence of *E. coli* was detected in soil with both the laterals (inline and bioline) using wastewater and similar trend was observed at all growth stage (Deepak Singh, 2011).

A field experiment was carried out at Centre for Protected Cultivation Technology (CPCT), to study the response of different colored plastic mulches on yield and crop water requirement of drip fertigated capsicum under greenhouse and to evaluate water dynamics using Hydrus-2D model. Three colored plastic mulches (silver, yellow and black) and three levels of irrigation (100 %, 80 % and 60 % ET_c) were evaluated. The treatments were replicated three times following a randomized block design. The maximum yield was observed for silver mulch at 100 % ET_c (63.9 t ha^{-1}) followed by yellow mulch at 100 % ET_c (60 t ha^{-1}). Statistical analysis showed that treatment combination of mulch color and irrigation level had significant effect on yield of capsicum. Mulch using inside greenhouse condition saved around 60 % of the water requirement than the open field. Water productivity (35.53 kg m^{-3}) highest for yellow color plastic mulch at 60 % ET_c, followed by 34.38, 33.83 and 33.33 kg m^{-3} for silver color plastic mulch at 80%, 60 % and 100 % ET_c. The use of plastic mulch in naturally ventilated greenhouse decreased the surface soil temperature up to 2 to 3°C during summer and during winter increased the surface soil temperature up to 3 to 5°C in comparison with control condition. Water dynamics in

the root zone of capsicum were simulated for 100 % and 80 % ET_c irrigation level for colored plastic mulches. The simulation results done with Hydrus-2D model showed that water content for sandy loam soil was more in the second layer of soil (15 to 30 cm) and was near the field capacity (Kumari, 2014).

Soil moisture sensor was developed and tested during the year 2011 and 2012 at Water Technology Centre, Indian Agricultural Research Institute (IARI), Pusa, New Delhi. Integration of sensors network with tele-communication technologies having modified tensiometer, level sensor, controller, GSM, receiver, transmitter, solenoid valve, water meter, pump were integrated to develop the automated irrigation system. The developed system was evaluated and tested successfully in Okra (abelmoschus esculentus) crop. The use sensor network, global system for mobile communication (GSM) and short message service (SMS) carry out data from sensor and make data base information to directly alert the farmer through their mobile phone and control the soil moisture status in the field. System allows the user to effectively monitor and control the water application in the field via sensor and/or through mobile phone set by sending command in the form of message and receiving the appliances status. This system is having potential of integrating the precision farming with sensor based application in fully automated system to interpolate over an area for spatial decision making need to be tapped for making agriculture attracting in future (Kumar, 2012)

A low cost, user friendly soil moisture sensor (Gypsum Block) for real time monitoring of irrigation was developed and inked with Wireless Sensor Network (WSN) using GSM module comprising with a microcontroller unit. The developed WSN system was installed in field for monitoring of soil moisture and irrigation scheduling. The Microcontroller was programmed to run the pump whenever the moisture content sensed by sensors installed in the field reached to threshold value (less than 15 %). Thereafter, microcontroller sends a text message to user mobile in the form of "Motor ON" through transmitter and receiver via use of GSM and SMS technology. When moisture content crosses field capacity (greater than 18-20%) microcontroller sends signal to motor as a result the motor and solenoid value gets OFF. Through the receiver and transmitter a text message is revived by the user in the form of "Motor OFF". The developed wireless sensor technology can be used by the rural farming community to save labor, efficient management of nutrition's and fertilizers through mobile phone (Veranna, 2016).

A LM35 IC based sensor circuit capable of measuring leaf -air temperature differential was designed, developed and tested in field during the year 2013 and 2014. A relationship between soil moisture content and leaf-air temperature differential was developed. The microcontroller unit of the sensor circuit was programmed to irrigate the crop automatically. Sensor circuit was integrated with drip irrigation headwork and tested in Kinnow (*Citrus reticulata Blanco)* crop. The microcontroller checks the leaf-air temperature differential measured by LM35 ICs at an interval of three days and irrigate the crop either for 0.5 hour or for 1 hour as per requirement. The developed automated irrigation system was able to maintain the soil moisture content nearer to field capacity throughout the experimental period and used 8 % less water in Kinnow crop. The cost of the developed automated system was ₹10,750 (Debnath, 2014).

An experiment was carried out during November to February 2015-16 at PFDC, Water Technology Centre, IARI, New Delhi to study the performance of different soil moisture sensors (FDR, tensiometers and gypsum blocks), soil water distribution and to develop the guidelines for use of soil moisture sensors at field level in a Cole crop (broccoli). Results revealed that FDR sensor remains a good tool for irrigation scheduling with limited drawbacks. Results also revealed that water content after irrigation exceeds above field capacity and remains around field capacity up to the 24h in root zone of broccoli. At the end of 48 h, water content was found to be slightly less than the field capacity in the active root zone. Therefore, irrigation of broccoli crop in sandy loam soil on alternate day basis will maintain adequate moisture in the root zone. Drip irrigation frequency significantly affected the broccoli yield. The maximum yield (24.46 t/ha) was obtained with 80% of ET_c with once in 2 days irrigation followed by 100% of ET_c with once in 2 days. Lowest yield (16.53 t/ha) was obtained at 60% of ET_c at once in 3 days irrigation (Kumari, 2016).

Effect of full and deficit irrigation regimes and nitrogen fertilizer levels on the yield and growth of maize was investigated in the semi-arid environment through conduction of experiment at research farm of Water Technology center (WTC), ICAR-Indian Agricultural Research Institute (IARI), New Delhi, India**.** The experiment was carried out during the *kharif* season of 2009 and 2010 with four irrigation levels *i.e.* rainfed (W_1); 50% FC) (W_2); 75% FC (W_3) and full irrigation: (W_4) and three nitrogen fertilization levels *i.e.* not fertilized (N_1); 75 kg N ha^{-1} (N_2) and 150 kg N ha^{-1} (N_3). It was observed that varying levels of irrigation and nitrogen resulted in significant difference in yield of maize. Maximum grain yield, biomass, water productivity (WP) and irrigation water use efficiency (IWUE) were observed for full irrigation (W_4) and N_3 treatment amounting 5930 kg ha^{-1}, 18150 kg ha^{-1}, 12.9 kg mm ha^{-1} and 15.7 kg mm^{-1} ha^{-1}, respectively. Subsequently, the water driven crop growth model AquaCrop was calibrated and validated using field experiment data of 2009 and 2010, respectively. The model was successfully calibrated for simulating maize grain and biomass yield for all treatment levels with the prediction error statistics model efficiency (E); $0.95<E<0.99$ and coefficient of determination R^2:$0.9<R^2<0.91$. Further, the validated AquaCrop model was evaluated in predicting the yield of *kharif* maize under future water availability scenarios. Future climatic data were generated using a climate generator ClimGen, which was parametrized using 36 year's historical data of the study area. The generated climatic data were used first in the CROPWAT model to estimate the irrigation schedule, which was then used in the validated AquaCrop model to predict the grain yield for future years. It was observed that the sowing date of the crop played a significant role in maize yield variations under the future climate change scenarios. The ClimGen generated rainfall depths during years 2012 (739 mm) and 2014 (604.9mm), resulted in significant yield difference of 1600 kg ha^{-1} and 5670 kg ha^{-1}, respectively under rainfed with full fertilization levels. Thus the process of using the outputs of ClimGen and CROPWAT in AquaCrop can be standardized as a model linking protocol to estimate future maize yield and irrigation water requirement for sustainable production and as an adaptation measure to climate change (Abedinpour, 2011).

Single and dual crop coefficients of mustard (Brassica Juncea) cultivar Pusa Vijay (NPJ-93) were determined using daily data acquired during 2013-14 and 2014-15 from weighing type field lysimeters located at the research farm of Water Technology Centre, ICAR-IARI, New Delhi, India. Weather parameters acquired from the automatic weather station located near lysimeters were used for estimation of reference evapotranspiration using modified Penman-Monteith formulae. Single crop coefficient (Kc) for mustard cultivar Pusa Vijay (NPJ-93) during rabi 2013-14 was 0.39, 0.72, 1.02 and 0.5, for initial (0-30 days after sowing (DAS)), development (31-70 DAS), mid (71-110 DAS) and late (111-130 DAS) stages, respectively. While in dual Kc [Kcb (basal crop coefficient) + Ke (soil evaporation coefficient)], the value of Kcb was 0.19, 0.55, 0.91 and 0.24 and the Ke was 0.20, 0.17, 0.11 and 0.26 for initial, development, mid and late season stages, respectively. Whereas, during rabi 2014-15 the single Kc was 0.36, 0.63, 1.04 and 0.44, and for dual Kc (Kcb+Ke) the value of basal crop coefficient (Kcb) was 0.17, 0.46, 0.91 and 0.23. Soil evaporation component (Ke) was 0.19, 0.17, 0.13 and 0.21 for initial, development, mid and late season stages, respectively. Ratio of transpiration to evapotranspiration was 0.75, which implies that the evaporation component was only 25%, whereas the transpiration component was 75% of the total crop evapotranspiration. Water productivity was estimated to be 14.9 kg/ha-mm corresponding to grain yield of 2.34 t/ ha and total depth of water applied 157 mm (i.e. two irrigations of 30mm each besides effective rainfall depth of 97mm) during rabi 2013-14. Whereas, during rabi 2014-15, the water productivity was 15.4 kg/ha-mm with grain yield of 2.89 t/ ha and total amount of water applied 187 mm (i.e. three irrigation of total 75mm besides effective rainfall depth of 112mm). A software was developed in JAVA programming language using experiment generated data and secondary data of mustard, wheat, maize and soybean crops acquired from lysimeter experiment to estimate crop coefficient and subsequent irrigation scheduling. Single and dual crop coefficients of mustard and the developed software can be used for judicious irrigation scheduling and enhancing crop productivity in this region (Gupta, 2015).

A field experiment with split-split plot design (SSPD) was conducted to study the response of two winter wheat (*Triticumaestivum L.*) cultivars (*viz.* salt tolerant cultivar KRL-1-4 and salt non-tolerant cultivar HD-2894) on growth and grain yield of wheat under saline irrigation regimes with and without foliar potassium

fertilization during *rabi* 2011-12 and 2012-13 in the research farm of the Water Technology Center (WTC), ICAR-IARI, New Delhi, India. Potassium in the ratio of K^+: Na^+ (1: 10) was applied as foliar application during the heading stage of the crop. It was observed that the maximum plant height under different treatments showed a decline trend with increase in salinity levels from 1.7 to 12 dSm^{-1} for both cultivars (KRL-1-4 and HD 2894). The maximum LAI value for salt tolerant (KRL-1-4) cultivar showed minimal variation (*i.e.* 3% to 16%) from 4 dSm^{-1} to 12 dSm^{-1}salinity regimes as compared to the control (1.7 dSm^{-1}) under non-foliar treatment. Results showed that the grain yield of KRL-1-4 and HD-2894 cultivars with foliar potassium fertilization at the heading stage increased on an average by 5.5 to 21% and 2.5 to 15% during both years of experiment, respectively under different saline irrigation regimes as compared to the control. Overall, it was observed that the foliar potassium fertilization increased the grain yield of both wheat cultivars, while the salt tolerant cultivar performed better than the salt non-tolerant cultivar under irrigated saline regimes. Further, The AquaCrop model was calibrated using experiment data of *rabi* 2011–12 and validated with data of *rabi* 2012–13. It was observed that the AquaCrop model prediction for grain yield was better [*i.e.* Model efficiency (ME)=0.86, index of agreement (d)=0.95 and Coefficient of determination (R^2)=0.96] as compared to biomass yield (*i.e.* ME= 0.91, d=0.97 and R^2=0.93) and water productivity (*i.e.* ME= 0.6, d=0.82 and R^2=0.93) for all treatment levels. It was also observed that the model could predict grain yield more accurately upto 8 dSm^{-1} as corroborated by lower prediction error ranging from 1.77 to 6.52%. Further, the production functions to predict the grain yield under different salinity regimes, irrigation and rainfall depths besides the quantity of foliar potassium fertilizer was developed. Predicted grain yield of both KRL-1-4 and HD-2894 wheat cultivars under different irrigation water salinity regimes and doses of foliar potassium to be applied at the heading stage of wheat is presented in the dissertation as management alternatives to assist stakeholders in enhancing productivity of wheat under irrigated saline environment. Mukesh Kumar, 2016)

Field experiment was carried out to study the effect of electrical conductivity on the crop water uptake, simulation of the water uptake and to develop the irrigation guidelines for the greenhouse capsicum grown in soilless media. The experiment was conducted during the year of 2015-2016 under semi-climate greenhouse condition at Center for Protected Cultivation Technology (CPCT), farm at Indian Agricultural Research Institute (IARI), New Delhi. The yellow colored popular and widely accepted capsicum variety Swarna was selected for the study. Four strategies/treatments based on electrical conductivity of the fertigation solution were applied with three replications during the entire growing period. Treatments were taken as (2.4,2.6,2.8), (2.3,2.5,3.0), (2.7,3.0,3.2) and (2.5,2.5,2.5) dS/m respectively for vegetative, flowering and fruiting stages. Crop water uptake was found to be maximum 246 kg and minimum 17.85 kg during fruiting and vegetative stage respectively corresponding to EC 2.8 and 2.7 dS/m for entire crop duration. Capsicum colored fruit yield was found to be maximum 41.15 Kg and minimum 20.05 Kg corresponding to the treatments T1 and T3 respectively having 15 plants each. Crop water productivity was found to be maximum 119.28 kg/m^3 and minimum 72.12 kg/m^3 corresponding to the treatments T1 and T3 respectively having 15 plant each. Water Uptake by the capsicum for entire growing period were Simulated by the Water uptake Model developed by Carmassi *et al.*, 2007. Determination coefficient for the linear regression between estimated and measured values of daily transpiration was found to be 0.836 with a slope of 0.748. The measured and estimated value of transpiration followed common trend throughout the entire crop duration and varied from 0.8-3.8 kg/m^2/day during 5-45 days after transplanting for greenhouse capsicum. The results indicated that water uptake model could be used to predict water requirement and improve irrigation control in soilless culture. However the model coefficients require parametric adjustments for specific climate and crop conditions. The total amount of irrigation water, no of irrigation, irrigation interval and irrigation duration varied from 1000-6400 ml per plant per month, 4-8, 3-6 days and 15-50 min for entire crop duration Oct-April (Ahirwar, 2016).

Crop Water Foot Print Assessment for Sustainable Agriculture

A study was undertaken to develop a methodology for WF assessment at rivers basin scale, to assess the VWC and WF of crops and to assess the sustainability of WFs within the Gomti and Betwa river basins of

India (Fig. 5. Impact of crop management options on basins' WF was also evaluated. Spatial Resolutions Units (SRU) delineated on the basis of homogeneity in soil, agro-climatic sub-regions, district boundaries and spatial variability in crop evapotranspiration (ETc). Optimization models were used to develop the cropping patterns to reduce the WF within the basins. Results showed that WF of Gomti and Betwa river basins was 12773 million m^3 and 9186 million m^3, respectively. The agricultural sector accounted for 96.1% and 97.8% of the total WF in Gomti and Betwa basins, respectively. Paddy, wheat and sugarcane had largest WF accounting for 88.3% of the total WF in Gomti basin. In Betwa basin, wheat, soybean and chickpea accounted for 75.3% of the total WF. At present, about 5054 million m^3 and 3590 million m^3 of water is being exported from Gomti and Betwa basins, respectively. Optimal cropping pattern with relocation of crops from high to low WF areas would result in savings of about 9.26% and 25.12% of blue water in Gomti and Betwa basins, respectively. Sustainability analysis of WFs indicated that water consumption and pollution pattern in Gomti basin are sustainable while in Betwa basin there is need for reduction of WF. With the adoption of improved irrigation methods, laser land leveling and optimal cropping pattern at basin scale, it is possible to reduce the WF and VW imports to the basins (Mali, 2015).

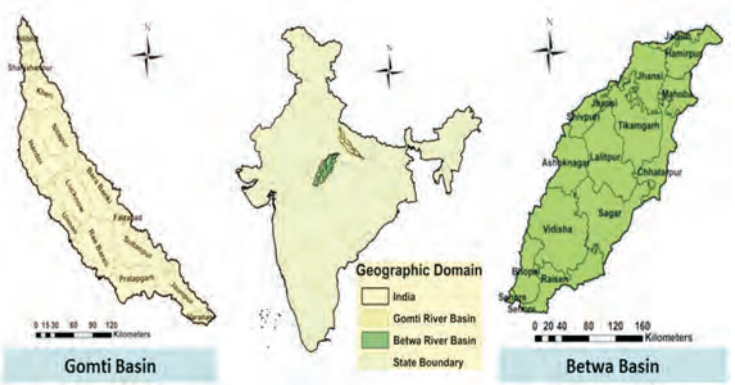

Fig. 5 Gomti and Betwa basins

Management of Saline and Waterlogged Areas

A decision support system for reclamation and management of salt-affected soil was developed. The DSS provides the user with the design of productive bunds/dykes to check the ingress of sea water in coastal saline soils and tidal gates to prevent the associated drainage problem. The DSS for the design of irrigation practices incorporate the leaching requirement for saline soils and concept of thin and frequent irrigations for alkali soils. Provision was made in the DSS whereby all the inputs from various modules were stored in a separate log file for future reference. Different modules were validated by comparing the calculated values from modules with the field observations by using student's t-test of significance. In general the decision support system performs well for deciding reclamation and management techniques for salt-affected soils. Visual Basic and MS office (including MS Excel and MS Access) are the software pre-requisites for proper functioning of this DSS on any computer (Singh, 2007).

Spatial prediction of soil salinity for subsurface drained agricultural field in *Western Yamuna Canal Command*(WYCC) area, Haryana, India was carried out using geostatistical approach to evaluate the impact of drainage on soil salinity. Semi-variogram analysis of soil salinity dataset revealed that the spherical semi-variogram model was the best to describe the spatial variability of soil salinity in sub surface drained agricultural field. Soil salinity maps for pre and post drainage periods were generated using Ordinary Kriging. Spatial analysis of the krigged maps for these periods showed that the salt affected area decreased from 56 % to 35 % and 78.5 % to 37.7 % after operation of subsurface drainage system during *kharif* and *rabi* seasons, respectively. Moreover, out of 35-38 % area where problem of soil salinity still exist, only 1.73 - 3.30 % lands were strongly saline (8 dS m^{-1}<ECe<16 dS m^{-1}) and remaining land area was moderately saline (4 dS m^{-1}<ECe<8 dS m^{-1}) after drainage. Overall, this study conclusively demonstrated the need and importance of spatial modeling of soil salinity using geostatistical technique for evaluation and management of subsurface drainage system (Kumar, 2007).

An experiment was conducted with four irrigation water salinity levels *viz.*Control (S_1, ground water) with EC of 4 dSm^{-1}(S_2); 8 dSm^{-1} (S_3) and 12 dSm^{-1} (S_4)and four wheat varieties including three salt tolerant [*i.e.*KRL-210 (V_1), KRL-1-4 (V_2), KRL-19 (V_3)] and one salt non-tolerant variety HD2894 (V_4).It was observed that the

salt tolerant variety KRL-1-4 resulted in highest yield under irrigation saline regimes for both the years with increase in average yield by 26% and 41% as compared to the salt non tolerant variety HD2894 at irrigation water salinity of 8 and 12 dSm^{-1}, respectively. Salt dynamics and its budgeting in the crop root zone, the conversion factors and regression equations for estimation of salinity of saturation paste extract (EC_e) from $EC_{1:2}$ and $EC_{1:5}$ were developed to minimize the estimation time without use of laboratory procedures. Salt budgeting during the growing season of both the years showed higher salt deposition (*i.e.*11.81 tha^{-1}) during *rabi* 2009-10 as compared to 6.91 tha^{-1} during *rabi* 2010-11. Production functions under different salinity levels and wheat varieties were also developed using the experiment data with R^2ranging from 0.94 to 0.99. Further, the AquaCrop ver. 4.0 with salinity moduleand SWAP (Soil-Water-Atmosphere-Plant) models were calibrated and validated using the experimental data of *rabi*2009-10 and 2010-11, respectively. It was observed that with limited data set, the AquaCrop model was able to simulate the grain yield in all treatments with prediction error statistics *i.e.* model efficiency (ME), index of agreement (d) and coefficient of determination (R^2) of 0.85, 0.96 and 0.94, respectively. SWAP model simulated the salt dynamics in the root zone with ME, d and R^2 of 0.95, 098 and 0.96, respectively. Moreover, the crop model WOFOST (World Food Study), a simple crop module in SWAP could able to simulate the grain yield using both the experiment generated and generic data with ME, d and R^2 values of 0.75, 0.93 and 0.95, respectively. Nonetheless, the study revealed that the salt tolerant variety resulted in higher yields and water use efficiencies with saline irrigation. Besides this, the salt dynamics in the root zone depths and crop yield simulated by the crop models would assist in scheduling of irrigation, leaching of salts from crop root zone and conjunctive water use planning for enhancing productivity under irrigated saline environment (Kumar, 2013, Kumar et al 2013, Kumar et al 2014, Kumar et al 2014, Kumar et al 2015).

A study was undertaken for monitoring and simulation of waterlogging and salinity in irrigation command using GIS and Remote Sensing from area located in Gohana Tehsil. Three spectrum indices NDVI, NWDI and BI were created. Also Digital Elevation Model (DEM) was prepared by digitizing the contour map under GIS environment for assessing the surface flooding with irrigation command. Along with this location wise risk prone areas of water logging salinization were simulated by salt balance approach. An area of 777.5 ha would remain unaffected due to relative elevation difference. At drainage coefficient of greater than 2 mm/day the rate of areal increase in salinity would be very low and the normal area would be unaffected in areal spread. At a drainage coefficient of 2 mm/day, the rate of salinity increase of saline land would be very small. Therefore, a drainage system designed for a drainage coefficient of more than 2 mm/day would be incommensurately expensive than the benefits (Pattanaaik, 2007, Pattanaaik, 2007, Pattanaaik, 2012)

Design and Development of Farm Machinery and Mechanization

An experimental plug seedling transplanting mechanism that can eject seedlings at the same speed of forward travel, but in an opposite direction was designed and fabricated. Two independent furrow opening and closing mechanism *viz.* 1) shoe type furrow opener with press wheel furrow closing cum compaction system were tested in soil bin using dummy seedlings. The effect of soil parameters viz. moisture content, bulk density and machine parameters *viz.* speed of operation on furrow closure, uniformity in spacing and success of transplanting plug seedlings were studied. A prototype vegetable plug seedling transplanter was then designed using CAD software "Pro Engineer Wildfire 4.0" based on optimum operating conditions and design values obtained in soil bin studies. A two row tractor mounted, semi-automatic vegetable transplanter for plug seedlings was fabricated and tested in field for transplanting one month old tomato and brinjal seedlings. Transplanting efficacy of 94.48% and 97.12% were obtained for tomato and brinjal plug seedlings, respectively (Gaikwad, 2010).

Mechanical hydrogel applicator was developed and evaluated. The physical and engineering properties of hydrogel were determined. Average angle of repose for each hydrogel sizes i.e. MS18, MS26, and MS36 were 39.6°, 42°, 43.9° respectively. The coefficient of friction for respective three sizes were 0.83, 091, 0.97. The average quantity of hydrogel granules obtained per hectare for MS25 size for three level of mixture (N1, N2, N3) with star wheel metering mechanism were 2535g, 2743g, 2896g; with the screw feed 2708g, 2917g, 3104g

and with brush feed 3542g, 3639g and 3403g for N1,N2 and N3. Thus the screw feed metering mechanism was selected design of the applicator. Field experiments were laid down using Factorial Randomized block design. Thus, the average hydrogel size of MS36 helped to attain maximum yield and improved crop growth parameters. Whereas MS 18 hydrogel size resulted in least increase in yield for treatments 1 to 6 taking different hydrogel sizes. As the particle size of hydrogel from mesh size MS 18 to MS36 decreased, the water absorption capacity increased and thus had longer existence in soil. The crop yields, straw yield, growth of the plant and moisture conservation were highest for the treatment T_3 and the lowest for control plot (Patil, 2011).

A web based custom hiring business model (Custom-Mach) was developed to suggest the fleet size, investment required, economic machinery size, operation scheduling and to facilitate decision making for custom hiring of combine harvester**.** Variation in farm power machinery demand supply gap for rice-wheat cropping system across different land holdings was assessed from the primary data collected from Aligarh Division.. It was found that mechanization has picked up only of selective operations stage-I mechanization requiring more energy and less intelligence. The existing custom hiring models were found unsatisfactory in terms of addressing the problem of mechanization gap existing in the area and were inefficient on economic, technological and managerial aspects. The developed fleet size selection model suggested a fleet of 3 cultivators, 3 harrows, 3 rotavator, 3 wheat seed drill, 2 combine harvesters, 1 straw combine, 2 balers, 1 laser leveler and 6 tractors with a total investment requirement of Rs. 98.7 lakh for a catchment area of 200 ha under paddy and 300 ha under wheat. The annual use hours for harrow, cultivator, rotavator, wheat seed drill, combine harvester, straw combine, baler, laser leveler and tractor predicted by the model for same catchment area and based on the selected fleet size were respectively 347, 278, 514, 250, 563, 375, 286, 417 and 922 respectively. For an area of 20 ha (10Paddy+ 10Wheat) size selection model gave the machinery size requirement of 0.61 m MB plough, 0.9 m disc plough, 1.63 m disc harrow, 2.42 m cultivator, 1.5 m rotavator, 2.42 m seed cum ferti-drill and a power source requirement of 33.56 kW. The combine harvester decision model was found to suggest the owning of combine harvester at 408 own acres and 200 custom acres or at 560 custom acres and 50 own acres. The optimum path selection model was found to guide the service operator the minimum travel distance and maximum coverage area. The results of the study in the form of custom hiring model is of great use for proper implementation of mechanization plan of any area (Parray, 2016, Parray *et al.,* 2016).

A study was conducted to develop a tractor drawn onion digger, which could dig the bulbs, lift the dug bulbs and finally windrow the crop after separating the soil. Biometric, physical and mechanical properties of onion bulbs and leaves (variety Pusa white round) relevant to the design of onion digger were first determined. Based on the determined parameters, a prototype scale experimental tractor drawn onion digger was fabricated with provision of changing length from 1.0 to 1.5 m, elevator slope from 15° to 25° and elevator speed ratio from 1.0 to 1.5. The influence of these variables on the performance parameters such as digging efficiency, separation index and damage percentage of the prototype onion digger were determined. The machine, operated by 35-45 hp tractor, performed satisfactorily and gave field capacity of 0.32 ha/h at the forward speed of 3.0 kmh^{-1}. The estimated cost of the developed onion digger was Rs.12,500/- . The operational cost of the digger was found to be Rs 992/- per hectare against the manual digging cost of Rs. 2162/ha resulting in saving of Rs 1170/- per hectare. The breakeven point for mechanized operation was 122 hours which was of 49 % of annual utility with payback period 3.9 years (Khura, 2008).

A low cost foliar applicator was developed for application of Urea Ammonium Nitrate (UAN). Physical properties i.e. dynamic viscosity, surface tension and specific gravity of UAN were determined at 3% N of UAN and it was found nearly equal to water. The reciprocating type PVC pump (2 no.) with capacity of 4.53 l/min each was selected for the UAN sprayer. Pressure relief valve (PRV) was used to maintain pressure level. Three type of nozzles i.e. flat fan, stream jet and stream jet were selected for study on the basis of application rate and pump discharge at different pressures. Spray pattern and uniformity of spray distribution of nozzles (flat fan, stream jet SJ3 and SJ7) were determined on patternator test rig. Flat fan nozzles showed most uniform pattern at 2 kg cm^{-2} pressure and 500 mm nozzle height, whereas streamjet showed maximum uniformity at 1.6 kg cm^{-2}

and 600 mm height. Streamjet nozzles were found suitable for low boom height and directed application of fertilizer. Variation in discharge of nozzles among the boom was observed less than 10 per cent while operated in field. Field capacity of the developed foliar applicator was found 0.3 ha h^{-1}. The average heart rate and energy expenditure for operating the prototype were observed as 119 beats min^{-1} and 26.7 kJ min^{-1}, respectively at operating pressure of 1.5 kg cm^{-2}. The estimated cost of the research prototype foliar applicator was 13000/-. The operational cost of the equipment was Rs 94.68 ha^{-1}. The breakeven point (BEP) and payback period(PBP) of developed foliar applicator were estimated as 113.43 h $year^{-1}$ and 4 years (Hitesh, 2016).

An algorithm was developed to determine soil nitrogen which, in turn, may be used for developing soil nitrogen sensor. Three different soils with the high percentage of sand, silt and clay respectively, were collected and analyzed in order to provide optimal algorithm for using in any soil type in future. In this study attempt has been made to firstly classify the three different types of soil based on the soil triangle with the relative percentage of sand, silt and clay respectively in each of these three soil samples. Physical properties i.e field capacity, permanent wilting point, available water capacity and chemical properties i.e pH, available nitrogen and total nitrogen, organic carbon, phosphorus and potassium were determined. Four levels of moisture content keeping in view the best moisture levels between tillage moisture and field capacity were selected. Moisture levels were controlled and the same were measured by moisture sensor as well as oven dry method. The electrical conductivity of these soil samples were measured by using EC meter. A significant relation between electrical conductivity and moisture levels of soil was observed. Four levels of nitrogen were selected based on common practice of nitrogen application of 50, 100, 150, 200 kg/ha. The electrical conductivity of these selected samples with varying nitrogen levels were measured by EC meter. A significant relation was observed between electrical conductivity of soil and levels of nitrogen therein. An algorithm was developed using a polynomial relationship between EC of each selected soil samples and moisture levels. Also a polynomial relationship was developed between EC of each selected soil samples and nitrogen levels. A significant relation was observed among EC and different levels of nitrogen as well as different levels of moisture. The coefficient of determination for moisture levels were 0.985, 0.988 and 0.981 for clay loam (46 % Clay), sandy loam (61 % Sand) and sandy loam (41% Silt) respectively in that order. The coefficient of determination for nitrogen levels were 0.9832, 0.9 and 0.99 for clay loam (46 % Clay), sandy loam (61 % Sand) and sandy loam (41% Silt) respectively in the same order. This algorithm maybe used to develop a sensor for real time measurement of available soil nitrogen which, in turn, will enable a farmer precise application of nitrogenous fertilizer (Hassan, 2016).

A study was envisaged to investigate the effects of rotary tillage on soil properties with special attention to sub-soil compaction. The limited pass effects on soil pulverization pattern were studied under three different levels of soil moisture content and speed ratio. Rotary tillage was found to significantly increase the bulk density and cone index at the depths of 15-30 cm. The rotary tilling created a compacted soil layer with soil cone index exceeding 2.0 MPa (2,135 MPa in sandy loam soil and 2.08 MPa after 40 passes and 60 passes, respectively. Large number of passes of rotary tilling led to the soil structure index of 2.29-2.41, which exceeded the theoretical upper limit for favorable seedbeds and declined root growth of maize in finer soil aggregates. The observations suggested that continuous use of rotary tillage deteriorated the soil structure and hindered root growth. Rotary ploughing gave maximum soil tilth indices and maize grain yield than rotary tilling. Tilth index model suggested that range of 0.79 to 0.94 as potential model indicator for maize crop for maximum yields.90.8-0.94 for sandy loam and 0.79-0.89 for sandy clay loam). Therefore the optimum rotary tillage operating parameters can be rotary tilling and ploughing at higher speed ratio with one pass in sandy loam soil and two passes in sandy loam soil. With view of soil pulverization and management of soil quality in sub-soil layers, rotary plough can be the alternative. Considering the effect of rotary tilling on sub-soil compaction and soil quality degradation, limiting its use to two operations per year or sub-soiling once in 4 years can be the option for sustained management of soil environment (Yadachi, 2016).

A study was undertaken to design and develop a tractor operated semiautomatic onion transplanter to mechanize onion transplanting operation. The physical properties of 50, 60 and 70 days old onion seedlings

relevant for the design of metering mechanism and prototype transplanting machine were determined. An experimental plug and finger type transplanting mechanisms were designed and fabricated. Soil bin studies were conducted on experimental transplanting mechanisms to study the effect of seedling parameters i.e. age of seedlings and machine parameters (speed of operation, height of seedling drop, finger material on plant spacing, planting depth, success of transplanting, furrow closure, filling efficiency and damage). A six row prototype onion transplanter was then and fabricated based on the findings of soil bin studies and optimum operating conditions and design values obtained. In plug metering mechanism, the observed plant spacing was 12.14 to 13.35 cm whereas in finger type metering mechanism it was 16.79 to 19.50 cm. The performance of plug type metering mechanism was found better and closer to the recommended practices than finger type metering mechanism. Therefore, plug type metering mechanism was selected for development of tractor operated onion seedling transplanter. The average field capacity and field efficiency of developed six row semiautomatic onion seedling transplanter was 0.078 ha/h and 76.53% respectively. The labor requirements in mechanical transplanting was 115.38 man.h/ha as against 400 man.h/ha in manual transplanting (Pandirwar, 2016).

A precision planter was designed and developed for direct seeding of paddy. The physical properties were studied for dry, 1-day soaked and 2-day soaked condition of paddy as per farmer practices, to design different seed metering plates. An electronic experimental test rig was developed to evaluate different performance parameters. From the analysis of performance parameters, a mean seed spacing of 15.1 cm, highest quality feed index of 90.5%, lowest miss and multiple index of 5.7 % and 6.2% respectively with a minimum seed damage of 0.38% were observed at a forward speed of 2.0 km/h, 350 angle of inclination with slanting type cell shape. A prototype precision planter with electronic seed metering system was developed and evaluated in the field condition. The performance parameters i.e seed placement index, seed rate, seed spacing and variation in spacing observed from field evaluation of designed electronically control planter were 86.39%, 19.97 kg/ha and 14.8 cm and while for mechanical planter the observed values were 74.28%, 22.7 kg/ha and 14.4 cm, respectively. Thus, the speed synchronization based electronic metering mechanism with proximity sensor and PWM based microcontroller was led to a saving of 12.04% in seed rate,16.3% increase in seed placement index, spacing closer to recommended spacing with a variance of 1.032 as compared to 4.984 variation observed in mechanical planter. The break -even point (BEP) and payback period (PBP) were estimated 128 h/year and 2.2 years for electronic and 137 h/year & 2.4 years, respectively for mechanical planters. Based upon the results it was concluded that a planter with electronic metering mechanism would lead to more precision and thus could be recommended for farmers use to reap the benefits of direct seeding. The developed prototype mechanical as well as electronic metering plate planters could be used separately according to the need and field situation (Rajaiah, 2016, Rajaiah et al, 2016).

An Aqueous fertilizer placement with seed drill was designed for dryland agriculture. Design consisted of component of design of main frame, metering system for both seed and aqueous fertilizer, furrow opener, ground wheel drive and a power transmission system. The aqueous fertilizer metering was attained by designing a suitable pumping system to deliver a measured quantity of aqueous fertilizer. Based on information available on pumping system and requirements of aqueous fertilizer pumping, a rotator gear pump assisted constant head gravity feed of aqueous fertilizer was found suitable. Increased rate of aqueous fertilizer gave enhanced growth performance parameters in addition to better germination. For 8000/ha aqueous fertilizer rate, increase in germination, no of shoot per plant, no of earhead, plant height, grain yield and straw yield were 51%, 48%, 38%, 11%, 38% any 60% respectively in comparison to those in plots with no aqueous fertilizer. It would make possible the sowing of wheat in dryland areas where seeding is not possible in many instances (Kant, 2008).

A study was undertaken to design and develop an experimental field plot drill to achieve precision in drilling of seeds and fertilizers on permanent raised bed. The physical properties of five wheat varieties (HD-2967, DBW-17, PBW-550, HDCSW-16 and HDCSW-18) and two granular fertilizers (Prilled urea and DAP) relevant for the development of metering mechanism were determined. The four types of furrow openers (Inverted T-type furrow opener with plain rolling coulter, single disc furrow opener, double disc furrow openers and

double disc furrow opener with plain rolling coulter) were developed and soil bin studies were conducted to study the effect of furrow openers at three forward speeds (1.5, 2 and 2.5 km h^{-1}) on soil physical properties i.e. moisture retention, bulk density and penetration resistance; and maize stalk residue cutting in three densities (3, 4 and 5 t ha^{-1}) to optimize furrow opener and speed of operation. The double disc furrow opener with plain rolling coulter cut 100% maize stalk residue at maximum speed of 2.5 km h^{-1}, but due to limitation of manual feeding of seed and fertilizer, the operational speed was kept at 1.5 km h^{-1}. The four row distributor with 100 kg ha^{-1} seed rate was found suitable for Oyjord metering mechanism. A prototype permanent raised bed seed cm fertilizer plot drill with four row distributor and the plain rolling coulters followed by double disc furrow openers was designed and fabricated based on the findings of laboratory and soil bin studies and optimum operating conditions and design values obtained. The field capacity and field efficiency of permanent raised bed seed-cum-fertilizer plot were 0.172 ha h^{-1} and 80% respectively. The plant stand of HD-2967, DBW-17, PBW-550, HDCSW-16 and HDCSW-18 were 225, 282, 225, 276 and 314 per square meter in case of conventional experimental plot drill and 312, 366, 340, 332, 375 per square meter in case of permanent raised bed seed-cum-fertilizer plot drill , respectively. Similarly the average grain yield in case of permanent raised bed seed-cum-fertilizer plot drill of HD-2967, DBW-17, PBW-550, HDCSW-16 and HDCSW-18 were 4.6, 5.5, 4.8, 5.4 and 6.3 t ha^{-1} respectively, compared to 3.7, 4.2, 3.7, 4.9 and 5.1 t ha^{-1} for conventional experimental plot drill respectively (Sawant, 2016).

A study on design parameters effecting mechanical carrot harvesting was undertaken. An experimental set-up of carrot harvester having provision to vary machine parameters like rake angle, length of soil separator and angle of soil separator was designed and fabricated. The experiment was conducted at three levels of moisture (15, 12 and 9 per cent) for different length of soil separators (40cm, 60 cm, 80 cm), three levels of rake angle (15°, 25° and 35°) and three different angle of soil separator (0°, 10° and 20°) using factorial design. The most suitable soil moisture was determined with all different combination of machine design parameters for best performance in terms of percentage of carrot harvested, percentage of carrot damage, soil separation index and power requirement. The design values of machine were determined at optimum soil moisture content. The maximum percentage of carrots harvesting of 97.4 per cent was obtained at 60 cm length of soil separator, 25° of rake angle and 20° of angle of soil separator. Minimum percentage of carrots damage of 4.87 per cent was obtained at 40 cm length of soil separator and 20° of soil separator angle. An average power requirement for the operation of carrot harvester at a speed of 2.3 km/h was 4.44, 5.3 and 5.75 kW at 15°, 25° and 35° of rake angle. Overall, soil separator length of 60 cm, rake angle of 25° and soil separator angle of 20° was considered design values of the harvester for efficient carrot harvesting at 12 per cent optimum moisture content. These design values are recommended for fabrication and evaluation of prototype. The prototype gave a carrot harvesting percentage of 97.8, carrot damage percentage of 4.56, soil separation index of 0.21, power requirement of 5.18 kW and field capacity of 0.21 ha/h when operated at a speed of 2.3 km/h. The estimated cost of single unit carrot harvester was Rs 6000 and operating cost of Rs 1481/ha (Shirwal, 2010).

A study to bridge the mechanisation gap by developing paddy straw collector-cum-chopper to find the long lasting solution for paddy straw after harvesting to enrich the exhausted soil was undertaken. The machine was fabricated after determining the physical and mechanical properties of paddy straw by Ti + DA texture analyzer. The fabricated prototype of paddy straw collector-cum-chopper comprised of three sub-systems i.e. straw collection and conveying unit, straw chopping unit and discharging mechanism with provision to apply optimised dose of fungal inoculum. The Duncan multiple range test disclosed that optimum conditions occurred at 40 % moisture content operating at 3 kmph and 40 cm height of cut. At optimum conditions, percentage of less than 5 cm, 5-10 cm, 10-15 cm and > 15 cm was 63.5 %, 8.12 %, 6.44 % and 17.92 %, power consumption of 16.14 kW, energy requirement of 7.82 kWh/t consuming 5.78 l h^{-1} fuel and 2.06 t h^{-1} machine productivity. The chopped paddy straw was decomposed with/without fungal inoculant above and below the soil. The treated chopped paddy straw incorporated within the soil by rotavator resulted in an increase in NPKC from 182.0 kg ha^{-1}N, 63.5 kg ha^{-1}P, 1862.5 kg ha^{-1} K and 0.51 % from initial value of 110.6 kg ha^{-1} N, 42.8 kg ha^{-1} P, 1068.4 kg ha^{-1} K and 0.33 per cent. Due to uniform mixing, the total biomass content increased from 273.3

to 290.1 µg of biomass / g of soil, resulting in an increase in dehydrogenase activity from 57.51 to 111.04 µg TPF g^{-1} 24 per hour and emission of CO_2 from 272.8 to 319.1 milli gram. As the degradation progressed, the total biomass content, dehydrogenase activity and emission of CO_2 decreased to 266.4 µg of biomass/ g of soil, 105.04 µg TPF g^{-1} per 24 hour and 307.1 milli gram after 45 days of degradation. The developed paddy straw collector-cum-chopper has an annual utility of 1146 tonnes, break even point of 556 hours per annum and pay back period of one year. It also resulted in 85.09 % cost saving in comparison to manual paddy straw management (Muzamil, 2016, Muzamil *et al.,* 2015).

A ridge profile power weeder was developed with 2.20 kW petrol-start kerosene-run engine as prime mover. The ridge profile power weeder was evaluated in terms of weeding efficiency, plant damage percentage and field capacity for different soil-machine parameters such as soil moisture content (15.26±0.96, 12.42±0.52 and 9.44±0.68%), types of blade (L- type, C-type and Flat-type) and gang speed (160, 180 and 200 rpm) at three different levels. It was observed that weeding percentage ranged from 74.47 to 93.89% for different soil- machine parameter combinations. Highest weeding percentage of 93.89% was recorded for L-type of blade and 200 rpm of gang speed at 15.26±0.96% soil moisture content (d.b). While it was lower (74.47%) in Flat-type of blade with 160 rpm of gang speed at 9.44±0.68% soil moisture content. Type of blade and gang speed had influence over the plant damage. Results showed that plant damage percentage varied from 0-7.33% for different soil machine parameter combinations. Plant damage percentage had shown positive correlation with gang speed. Lowest plant damage was observed at gang speed of 160 rpm for C-type of blade. Average field capacity of ridge profile power weeder was 0.08 ha.h-1at 15.26±0.96% soil moisture content for C-type of blade. C-type of blade was most suitable one with little compromise for weeding efficiency with minimum power requirement. Maximum performance index was estimated for C- type blade as 192.34 while, lowest was estimated for Flat-type of blade as 153.94. Average field machine index computed was 66.51 per cent. The total cost of machine was Rs. 27600/- and its estimated cost of operation was Rs. 640/- per ha. The saving in cost of operation was 88.3% as compared to manual weeding (Rs. 5470). Time Saving with ridge profile power weeder as compare to manual weeding was 92.97 per cent. The ridge profile power weeder had a breakeven point at 179.38 h.yr-1 with a payback period of 2.74 years (Thorat, 2013).

A pressurized metering system was designed and developed for precision seeding by controlled application of aqua-fertilizer according to available soil moisture. An experimental set up was used to optimize different pump variables for required discharge rate. The design values of different variables were selected for calibration of pumping system. Based on the design values, aqueous fertilizer metering mechanism was fabricated and mounted on existing tractor operated seeder after suitable modifications. The power to operate pressurized pumping system was derived from its PTO. Different components of seed drill were selected as per BIS standard. Power transmission system from tractor PTO to seed drill and pump variable speed drive were designed and fabricated to suit the requirement of aqua ferti seeder with pressurized aqueous metering system. The functional test evaluation of aqua ferti seeder was done for discharge and uniformity. The developed machine is capable of delivering a maximum discharge of 2664 l/h aqueous fertilizer with uniformity (Lande, 2008).

A comprehensive study was undertaken in four blocks of Bulandshahr districts of Uttar Pradesh to assess the use pattern of tractors and machinery in terms of age, use hours, use patterns and economic factors. Based on the machinery use pattern in the selected area a custom hiring model in Visual Studio C# programme was developed for selection of tractor and associated equipment for a particular operation. The model could select appropriate machinery system according to area under a crop, number of operations for each crop, available time gap between crop rotation and time available for each operation. The choice of opting for custom hiring on economic criterion was also offered by the model. Validation of the model indicated that the model outputs did not significantly vary from actual field observations. The model suggested that for paddy-wheat crop rotation, all four blocks of the district would require 33-87% additional tractors for timely completion of field operations, which can be made available through custom hire of existing fleet in the district. Present annual

uses and economics of uses suggested custom hiring of improved implement as laser leveller, rotavator and seed drill a better option for the district (Shyam Nath, 2012).

Physical and engineering properties of urea briquettes UB1 (3g) and UB2 (2g) were studied in relevance to the design of mechanical urea briquette applicator. Size and shape of metering cup were designed on the basis of maximum size of briquettes. Performance of metering mechanism of the applicator for uniformity of placement was tested in the laboratory on sticky belt, and performance parameters as average spacing, multiple index, missing index, quality of feed index and precision were evaluated. urea briquettes UB1 (3g) could provide recommended rate of N, optimum metering cup size for the briquette size was selected as 6.5 mm for final development of the applicator. A prototype urea briquette applicator was designed, developed and subsequently evaluated for its performance in sandy loam puddled soil at the Division of Agricultural Engineering, IARI, New Delhi. The mechanical urea briquette applicator recorded average draft of 92.41 N during the operation in the field at forward speed of 0.87km.h-1. The average placement depth of 7.66 cm in the range of 6.6-8.6 cm was observed, which was in the range of recommended 5-10 cm depth for deep placement of urea briquette. The developed urea briquette applicator could thus place the briquette at required depth and at required distance in a row. The power required to operate the applicator was 22.33 W, which was within the human capability of a person. Average field capacity of 0.05ha.h-1 was obtained for continuous operation of urea briquette applicator at an average speed of 0.87 km.h-1. Field efficiency of 69% was observed, which was in prescribed range of 65-75 % for planter, a similar machine (Deo, 2013).

A study was undertaken to develop and evaluate a two-row power operated multicrop planter for planting maize and soybean. Inclined plate metering mechanism was designed, fabricated and tested on the sticky belt set-ups for uniformity of seed spacing, performance indicators using different combinations of design variables such as metering plate of different cell shape (semi-circular B1, rectangular B2 and L-shape B3) and different cell size (7, 8 and 9 mm for maize and 10, 11 and 12 mm for soybean seeds). With the optimum design values selected from lab experiment, a prototype multicrop planter was designed and fabricated using necessary components for planting maize and soybean seeds. The field evaluation of the prototype recorded an average draft and power requirement of 1.56 kN and 0.61 kW at soil moisture content of 12 to 12.5 % and bulk density of 1.2 to1.3 g.cm-3 with an average fuel consumption of 0.75 l.h-1. An average field capacity of 0.11 ha.h-1 was obtained for continuous operation of multicrop planter at an average forward speed of 1.36 km.h-1 for both the crops. The machine had an overall average field efficiency and field machine index of 80.74 and 71.78 %, respectively. The average depths of seed placement were 2.25 cm and 2.15 cm for maize and soybean, respectively. The average plant spacings on the 21st day post sowing were 16.74 cm and 8.51 cm for maize and soybean, respectively, against the theoretical seed spacings of 17.48 cm and 9.65 cm. The average plant population per meter length of bed was 6 and 12 as against theoretical plant population of 5 and 10 for maize and soybean, respectively. Average germination percentages in field were 76.67 and 73.33 % for maize and soybean, respectively. The machine weighed 65 kg, and thus suitable for hilly regions (Singh, 2013).

A study was done to optimise the design and operational parameters of the turner-cum-mixer-machine used for rapid compost preparation. The experiment was conducted at three different levels of blade shape (Straight, L-shape and Knife edged) with three different rotor speeds (220, 300 and 350 rpm), three levels of forward speed (1.26, 2.26 and 4.17 km/h) and three different pile heights (0.8 m, 1.0 m and 1.2 m). The optimised performance parameters were tested with 1000 ml culture dose per tonne of the material of the compost. The optimum conditions were found with Straight shaped blades at a rotor speed of 300 rpm operating with the tractor forward speed of 2.26 km/h and pile height maintained at 1.0 m. This resulted in decrease of the compost density from 514.3 kgm-3 to 299.1 kgm-3 The uniform mixing at the optimum parameters caused moisture to vaporize from the top surfaces and loss was from 19.4% to 6.66% with the maximum temperature of 60^0C from the initial ambient temperature of 39^0C. The electrical conductivity increased from 1421 µS/m to 1531 µS/m. The analysis indicated 0.578% N, 1.17% K and 1.24% P, and variation in C: N ratio from 54:1 to 21:1. The reduction of the particle size with less moisture content helps the smaller particles to pass easily

through 4 x 4 mm sieve. All the nutrient contents and various parameters studied were in accordance with the standards set by BIS. The operation hourly cost of compost turner-cum-mixer with a 55.95 kW tractor was Rs 700, the payback period 1.0 year and Break Even Point 722 hours per year and the annual utility of the machine could be 0.3 lakh tonne of compost. The mechanized pile method of composting saved 90.4% cost of production. Also, there was saving of 75 days per cycle in compost preparation using this method in comparison to traditional pit method (Muzamil, 2012).

An experimental set up of garlic harvesting system was fabricated to determine the influence of soil-machine parameters on mechanical harvesting of garlic at three levels of soil moisture content (15, 12 and 9%), rake angle (100,150 and 200) and speed of operation (1.5, 3.0 and 4.5 km.h-1). Field experiments were conducted to determine optimum values for design of garlic harvester. The design values were incorporated in design of a garlic harvester and field evaluated. It was observed that soil moisture significantly affected harvesting percentage, soil separation and power requirement. Rake angle influenced overall performance parameters. Machine speed of operation also had effect on soil separation index and power requirement. Soil moisture of 12.23±0.35% (d.b), rake angle of 150 and 1.5 km.h-1 speed of operation were recommended as design operational parameters of a garlic harvester. A 4-row tractor operated garlic harvester was accordingly designed and field evaluated in sandy loam textured field. Harvesting percentage, damage percentage, soil separation index, power requirement and field capacity were observed to be 96.12%, 5.94%, 0.26, 4.54 kW and 0.24 ha.h-1, respectively, and were satisfactory. Operational cost of machine harvesting was estimated to be 55 % lower than that of manual harvesting. The prototype garlic harvester had a breakeven point at 218.12 h.yr-1, with a pay back period of 3.63 years (Khambe, 2012).

Variations in design parameter of trailers and their operating conditions in Haryana and Maharashtra were assessed to ascertain different factors causing instability. Survey revealed that Haryana had almost equal share of single and double axle trailers whereas in Maharashtra use of single axle trailer was 36 % more. Inter-state variation in instability response of the operators was higher on sloppy conditions vis-a vis normal road conditions and also the material loaded ranged from 0.5 tonne to 12 tonne in the single axel trailer whereas 0.8 to 15 tonne in double axel trailer. A scaled physical model with scale factor 2.44 was designed and fabricated to determine factors affecting both static and dynamic stability. Static load test on varying slopes showed that the angle of inclination interacted with the normal load in influencing the load shift from outer wheel to inner wheel and thus causing instability of the tractor trailer system. An optimum normal load less than 4271.3 N and the centre axle position of trailer may be safe operating conditions for tractor trailer system when angle of inclination less than 15°. The safe level of stability was found to be more in case of double axle trailer operation than that of single axle trailer in terms of normal load carrying capacity, operation of system up to 5° with higher forward speed up to 4.056 km/h as compared to single axel trailer. The critical limits of inclination for operation of single axle trailer with centre axle were in the range of 7.52° to 8° with forward speed of 1.26 to 3.12 km/h. The critical limits for operation of double axle trailer are 7.24° to 7.6° of angle of inclination with forward speed of 1.27 to 3.23 km/h (Lande, 2015 , *Lande et al.,* 2015).

Fig. 6. Development of Self Propelled Garlic Planter

A study was undertaken to design and fabricate self propelled garlic planter and evaluate its performance (Fig. 6). A inclined plate type

metering device with cell size of 15 % more than maximum clove dimension was designed based on the physical and engineering properties of the garlic cloves. The various performance parameters such as average seed spacing, miss index, multiple index, quality of feed index, precision and seed damage were 9.42cm, 6.8 %, 12.72 %, 80.48 %, 22.67 % and 8.26 % respectively. A petrol engine of 2.65 kW was used as the power unit. Field capacity of the planter was found to be 0.09 $ha.h^{-1}$ with a field efficiency of 77.7 % when operated at operational speed of 1.5 $km.h^{-1}$. The estimated cost of the machine was Rs. 50550 /- with the cost of operation per hectare was Rs.2370 /- as compared to manual hand planting with cost of Rs.22500 /- per hectare. The breakeven point was found to be 79 $h.yr^{-1}$, and it was having a payback period of 3 years (Barik, 2014).

Fig. 7 Oscillating Soil Separator for Garlic Harvester

A study was undertaken to design and develop oscillating soil separator for garlic harvester. A prototype garlic harvester was designed using CAD software. The length and width of soil separator was kept as 1000 and 900 mm. Rod spacing was kept as 50 mm. Highest garlic harvesting percentage (98.07%), maximum soil separation index (0.20) and minimum damage percentage of 3.47% was found in sandy loam soil at moisture of 12.88±0.35 percent (d.b). Minimum power requirement (2.43 kW) for operation garlic harvesting system also occurred at 12.88±0.35 %. Highest garlic harvesting percentage, maximum soil separation and minimum damage percentage of 97.78%, 0.19 and 3.76% respectively was observedat 25" rake angle. The garlic harvester's average harvesting efficiency was 94.76%, similarly the damage percentage (4.81), soil separation index (0.23) and optimum power requirement was 5.74 kW for soil moisture content of 12.88±0.35 per cent. The total cost of machine was Rs.24700/- and its estimated cost of operation Rs.1670/- per ha. This cost of operation is much lower than cost of manual harvesting (Rs.8250/- per ha). The garlic harvester had a breakeven point at 125.36 $h.yr^{-1}$ with a pay back period of 2.6 years (Borkar, 2014)

A study was undertaken to design and evaluate a single row mechanical carrot planter. Biometric properties of carrot seeds helped determine the design values of different parts of metering mechanism, such as cell size (2.8±0.42 mm), cell diameter (1.2±0.36 mm), thickness (0.61±0.09 mm), hopper wall slope (40°) and material for metering plate (Aluminum). Inclined plate metering mechanism was designed, fabricated and tested on the sticky belt for uniformity of seed spacing, performance indicators using different combinations of design variables such as plates of different cell shape (Semi-circular P_1, Triangular P_2 and slant P_3) and different treatments of carrot seed (uncoated S_1, biogas slurry coated S_2 and Thirame coated seed S_3) at three inclinations of metering device (40°, 50° and 60°). The combination of design variables viz, 50° plate angle, slant cell shape performed the best with respect to planting and gave 83.9 per cent of quality of feed index, 20.9 per cent of precision, and with the least level of miss and multiple indices of 6.11 per cent. With the above design values, the prototype planter was designed and fabricated using necessary components for raised bed planting of carrot seeds. The field evaluation of prototype recorded a minimum draft of 378 kgf and 4.3 l/h fuel consumption. The optimum plant population per meter length of bed was 72 for treatment S_2 against theoretical plant population of 80 plants and resulted in high yield of 20.14 tonnes/ha. Mechanical carrot planter could reduce 53.88 % of cost of operation with break even points at 122 h/year and the payback period of 3.71 years (Yadachi, 2011).

A variable rate fertilizer metering mechanism was designed by changing speed of the feed shaft. A microcontroller unit programmed with a PWM motor driver was used to run the feed shaft at varying speed. Two types of fluted rollers i.e. straight and helical flutes were used with varying number of flutes having same volume each flute.There were no significant difference between Helical and straight fluted roller with varying numbers of flutes (6,8,10,12) with respect to discharge rate. The discharge rate of SSP fertilizer was significantly higher than that the DAP. There was significant difference in applied Torque and Power requirement for straight and helical fluted roller with different number of flutes, as well as types of fertilize. Helical shaped roller with 6 flutes was having minimum Torque and Power requirement in comparison other fluted rollers. Based on the result obtained from experiment speed of the metering feed shaft can be recommended for application of different granular fertilizers (Gurjar, 2015).

Ergonomic Design Modification of Manual Tools

A study was undertaken to determine the dust concentration and develop the safe interventions for farm workers. Four observation points were selected for the measurement, such as workers harvesting with sickle, workers feeding crop into thresher chute, workers giving crop to feeder and workers collecting threshed grains. Dust concentration was measured in the breathing zone of workers using a real - time personal dust monitor. The average values of environmental parameters such as temperature (^{0}C), wind velocity (m/sec) and relative humidity (%) were also recorded on each day of experiment. The measured dust levels were compared with standard exposure limits. The particle size distribution of dust during above operations was also measured using AEROCET 531 sampler. Three filter materials namely cloth, foam and tissue paper which are commonly used for the protection against solid aerosols in the disposable type masks were selected for the study. An experimental set-up was fabricated to determine the filter efficiency and pressure drop across the filters. Effect of varying filter area and dust concentration on filter efficiency and pressure drop across the filter was also studied in the fabricated set-up. The personal dust protectors with the simple design were developed from the tested filter materials. All the three dust protectors were subjected to suitability tests of one hour each on twenty workers performing wheat harvesting and threshing during the season as per the test procedure mentioned in BIS 9473-1980 (Pandirwar, 2010).

Interventions to reduce vibration transmission to power tiller operator in various field operations were developed. VST shakti 130 DI, PCB piezotronics, adapter and the data analyzing system (AIDAVibra, Italy) were used. Three different operational conditions and three different speeds were selected to determine the hand-transmitted vibration. The magnitude of vibration was measured in terms of rms acceleration in different axis, as outlined in ISO-5349. The physiological and postural parameters included in the study were heart rate, oxygen consumption, energy consumption, body part discomfort score and overall discomfort score. To reduce vibration magnitude, three different vibration reduction materials were selected. Considering the properties, availability in different shape and sizes and cost effectiveness, the rubber and the polyurethane were selected for interventions development. Depending upon the nature of contact between handle bar and chassis two types of interventions were developed. The nut and bolt contact vibration transmission was reduced by developing collar bushes. In surface to surfaces contact, plane sheets interventions were developed for vibration reduction (Chaturvedi, 2010).

A ergonomic variable height platform for training and harvesting of tomato crop in greenhouse was designed and developed. Five different work heights including 6 different subjects and 2 tools i.e. scissor and secateurs was performed to find the optimum work height for least discomfort and physiological cost. It was found that working at elbow height caused least discomfort and required minimum energy for training of vines by the subject. To attain this optimum work height, an ergonomic variable height platform was developed which provided versatility of height adjustment with help of foot operated hydraulic pump. Heart rate and energy expenditure were found to be minimum i.e.98bpm and 12.9 KJ/min which put this work in the category of light work. There was significant difference in the Heart Rate, Body Part Discomfort Score, Overall Discomfort Score

and Energy Expenditure on increasing the working height of the subject. There was no significant difference in the heart rate and energy expenditure between the 2 tools i.e. scissor and secateur. Significant difference was found between the Body Part Discomfort Score and Overall Discomfort Score between the 2 tools i.e. scissor and secateur. Vertical movement of the platform in one time during pruning was found to be 46.1 cm as it was the vertical length of the workspace envelope of hand at elbow height of 5th percentile of Indian population. On the basis of this study subject is suggested to work within the workspace envelope of hand at elbow height resulting in lesser discomfort, energy expenditure and greater productivity (Choudhary, 2016).

A power tiller operator model was developed for the vibration study. Model results showed that intervention in the form of handle grip on the handle bar(I1) of the power tiller may reduce vibration by 8.97 per cent. Another intervention applied at the interface between handle bar and the chassis (I2) reduced vibration by 33.76 per cent. A vibration reduction of 61.54 % was achieved by incorporation of engine isolators in between the chassis and the engine (I3). With these results from the model it was found that the vibration reduction was highest when the interventions were applied at location between engine and chassis. The vibration reduction was further enhanced with the incorporation of the interventions at all the places together. By getting this vibration reduction the interventions were designed and developed for different interfaces in the power tiller. Based on the model data the interventions were developed and the experiments were carried out for the determination of vibration reduction. Eight types of interventions were developed with the model output. It was observed that the vibration reduction were higher with the developed interventions I_1, I_2 and I_3. but the stability of the system reduced. For adequate stability of the system two support systems were designed for the intervention I_3. It was observed that the vibration reduction was high (60-70 %) with the developed interventions. Further daily exposure limit for safe operation of power tiller without having chances of HAVS were also determined. The total duration of 10 % prevalence of power tiller was also determined with the help of the observed data. The measured data was also compared with the ISO 5349 (1986) to get the daily safe exposure limit for the operator (Chaturvedi, 2016).

Comparative study of three types of manual weeder namely wheel hoe, crescent hoe and dry land weeder were ergonomically evaluated for women farm worker. The highest heart rate, oxygen consumption and energy expenditure was observed in the operation of wheel hoe. Working with wheel hoe was categorized in heavy work and body part discomfort and overall discomfort score was higher than other weeders, and also field capacity of wheel hoe was highest. A wheel hoe weeder with geometric reduction in the cutting blade (S_1 (5%) and S_2 (10%)) with modified handle dimensions (W_1 (400mm), W2 (440mm) and W_3 (480mm)) was evaluated for work output and ergonomic parameter. It was observed that heart rate with modified wheel hoe reduced by 11 beats per minutes as compared to the existing wheel hoe. There was shift in the category of workload from "heavy" to "moderate". It was observed that heart rate, oxygen consumption and energy expenditure were lowest for (S_2-W_2).Resting period for existing weeder reduced from 33.33min to 20.63 min with modification. There was no significant difference in the field capacity. The postural parameter like average twisting velocity, maximum sagittal flexion and maximum lateral velocity were lowest in modified tool with (S_1-W_3), (S_2-W_2) and(S_1-W_2).The modified tool minimizes shoulder, elbow and wrist deviation and moves towards the neutral posture. The body part discomfort score and overall discomfort of workers also reduced to 18.38 and 1.47 for (S_2-W_2) compared to the existing tool (body part discomfort and overall discomfort score i.e. 31.01 and 2.51) (Sinha, 2014).

Tractor operator protective system was designed to mitigate the dust and temperature exposure. The cab is cooled by passing air through a wet evaporative surface, made up of 35-mm-thick layer of shredded wood packaged between the wire mesh. The evaporative surface is kept continuously wet with recirculation of water by a low-capacity, submersible water pump installed at the bottom of a water tank. At tractor speed of 5 kmph, relative air velocity at the face of the pad was about 1.1 m/s. Evaluation of protective enclosure was done with different field operation and at different environmental parameters level. The temperature significantly reduces from 43°C to 33.25°C with the relative humidity 32% to 39% for secondary tillage operation and 42°C

to 35.3°C with relative humidity of 34% to 41% for transportation operation. The dust concentration level for PM 10 reduced significantly from 18.5 mg m^{-3} to 0.35mg m^{-3} for secondary tillage operation and 4.4mg m^{-3} to 0.2 mg m^{-3} for transportation when compared with without productive enclosure and with protective enclosure. Adaptability of protective system was analyzed through physiological parameter of tractor operators heart rate reduced from 138 beats min^{-1} to 119 beats min^{-1} and overall discomfort score from 4 to 2.9 for secondary tillage operation, 117 beats min^{-1} to 107 beats per min and overall discomfort score to 2.9 to 1.9 for transportation operation. The evaporative cooling efficiency achieved through the system was 80% (Kumar, 2012).

A study was undertaken to find out best possible human posture for converting human power into rotational power. The study was undertaken with objectives to develop an experimental setup for rotary power application, to determine the physiological and postural parameters of operators discomfort in different modes of rotary power application and to evaluate optimal human efforts in different modes of rotary power application. For this study, six subjects were selected. The measured data was used to calculate increase in heart rate over resting heart rate (Δ HR), oxygen consumption (l min^{-1}), energy expenditure (kJ min^{-1}), % VO_2max, human efficiency, BPDS and ODS (Potdar, 2008).

A study was undertaken to find out best possible human posture for converting human power into rotational power using leg muscles. The postural dynamometer comprised of a frame on which flywheel was mounted which is rotated by different modes i.e. pedal, stepper and bicycle mode. The experiments on postural dynamometer for foot operated rotary power generation were conducted with these subjects through the three operation modes i.e., pedal, stepper and bicycle, 5 levels of mechanical load conditions and with three replications. The measured data was used for calculation of increase in heart rate over resting heart rate (Δ HR), oxygen consumption (l min-1), energy expenditure (kJ min-1), % VO2max, human efficiency, BPDS and ODS (Ram, 2008).

A study was conducted to find out the best forearm posture in ideal combination of muscles to optimize push/pull strength. Experiments were conducted for seven forearm positions *viz.* neutral, pronation, dorsiflexion, palmerflexion, radial deviation, ulnar deviation and supination. Ergonomic evaluation in terms of heart rate, oxygen consumption, energy expenditure, body part discomfort (BPDS) and overall discomfort score(ODS) were assessed for different forearm position at different loads. The maximum aerobic capacity (VO2 max) of the selected subjects varied from 2.05 to 2.35 l min-1. The working heart rate (beats min-1), oxygen consumption (l min-1), energy expenditure (kJ min-1), ΔHR (beats min-1) ,%VO2 Max, BPDS and ODS were highest for radial deviation and lowest for neutral deviation of forearm position. Higher load output condition under all the seven forearm position force application the ΔHR (beats min-1) values exceeded the limit for continuous performance level (40 beats min-1). Based on the result in terms of maximum push pull strength and ergonomic parameters, it was found that neutral forearm posture is the best mode of force application. However there was similar push pull strength in the range of handles orientation from 00 to 350 from the neutral position (Kumari, 2015)

A house to house survey of forty farm women workers was conducted in Thanakalan village, Kharkhoda Block of Sonipat district in Haryana to study the load carrying activities. The result of the survey showed that musculoskeletal problem was the major health problem among the women farm workers in different age groups. It was observed that 38 out of 40 woman farm workers (95 %) had musculoskeletal disorder. Carrying load (10 kg to 40 kg) by women farm workers was an everyday activity which contributed to musculoskeletal disorders. Body parts discomfort shows that lower back is the most affected body part, as 87.5 per cent women farm workers complained of back pain, followed by neck pain (72.5 per cent) and knee and foot pain. An experimental study of different modes of carrying different load (i.e. Head, Back and Shoulder and 10, 15, 20 kg) on different ground inclination (i.e. 0, 5, 10 %) was conducted in laboratory set up. Women working on local farm were selected and prepared to walk on treadmill. The results showed that head mode of carrying load on horizontal surface is economical in terms of energy consumption irrespective of quantity of load as compared to back and shoulder mode. On inclined surfaces (i.e. 5 and 10 % ground inclination) back mode of carrying load was less energy consuming as compared to head and shoulder mode. A Biomechanical model was developed to predict metabolic energy cost for carrying load in different modes and inclinations. Model had

high co-relation (co-efficient of 0.95) between measured and predicted values of metabolic energy consumption (kJ/min). User friendly interactive software was also developed. A Shoulder mounted harness was designed for women farm workers for carrying load which did facilitate load carrying similar to head loading without introducing element of eccentricity. The harness was made up of aluminium material to make it sturdy and light weight (Jena, 2016¸ Jena et. al, 2016).

Rural Storage Structures and Processing Equipment

A study was conducted to evaluate the different solar coatings on the efficiency of solar dryer for sapota drying. Ten solar coatings were selected such as GI, Al, Cu, black synthetic coated GI, black synthetic coated Al, black synthetic coated Cu, black synthetic enamel coated GI, black synthetic enamel coated Al, black synthetic enamel coated Cu as non-selective coatings while black chrome as selective coating. The spectro radiometer and Fourier's transform infrared spectroscope (FTIR) and infrared thermometer were used to measure the reflectance and emissivity and temperature difference between coatings and ambient temperature of solar coatings respectively. The reflectance of these coatings was found in the range of 0.01 to 0.56 while emissivity was in range of 0.93 to 1. The temperature difference (Ts-Ta) of non-selective coatings with ambient temperature was found maximum. The collector efficiency with black synthetic enamel coated Al was found as 25.67% and the average mid-day thermal efficiency of non-selective coatings was around 23% that keeps the collector temperature almost constant for some minutes when the radiation falls. It was found that the black chrome gives the quick response to temperature change so random variation in thermal performance of solar air heater. So black synthetic enamel coated Al was found best coating among selective and non-selective coatings (Sawant, 2011) .

An evaporative cooled storage structure (ECSS) was designed to assess the performance with three different pad materials and four distinctive pad areas in no load condition. The SPV system consisting of 4 solar panels of 100Wp each, one inverter and a battery bank (including 2 batteries) were used. The total energy produced in a by PV modules to the battery bank was 2.4 kWh. From the load data, consumption of energy was observed to be 2.212 kWh/day which was about 92.16 % of the power supplied by solar PV system. This showed that the SPV system is suitable to supply electricity to cover the loads requirement without utilizing power from the electrical grid during the sunny day. The materials examined were wood wool, *khas* and CELdek. The areas for the cooling system considered were of 2.25 m^2, 4.5 m^2, 6.75 m^2 and 9 m^2. The performance criteria included measuring temperature, relative humidity and cooling efficiency. For testing the performance of the ECSS, produce having low shelf life such as spinach, green beans, pointed gourd, cucumber, bottle gourd, capsicum, tomato and papaya were used. The results obtained for fruits and vegetables storage indicated that there was significant difference in PLW (%) and firmness values of the fruits and vegetables when stored in ECSS than those in room (ambient condition). The shelf life for the fruits and vegetables stored in ECSS was 1-3 days more compared to room storage. Evaporative cooling system is easy to operate, efficient and affordable especially in the areas where electricity is not available or its supply is erratic (Mansuri, 2015, Mansuri et al, 2016).

A study was conducted to evaluate the effect of thermal and oil pre-treatment on the recovery of green gram dhal. The dhal recovery was found to increase by about 15% after oil treatment and decreased after thermal treatment. Cooking time also decreased by 27.40 % over untreated sample in case of oil treatment and 32.55 % in case of thermal treatment. There was significant difference in dhal recovery and cooking time in case of untreated, oil treated and thermally treated samples (Wani, 2009)

An equipment was developed for hydro-priming of vegetable seeds. With the help of CAD software "Pro Engineer Wildfire 4.0" detailed manufacturing drawing was prepared before final fabrication of the equipment. The developed equipment was then evaluated for its performance on priming of okra and pea seeds. Okra (*Abelmoschus esculentus* L. Moench) and pea (*Pisum Sativum*) seeds were subjected to accelerated aging process

to further obtain lots having different germination percentage and moisture content. The moisture content and germination percentage for okra seed lots was in the range of 10.98 to 16.89 % d.b and 80 to 50%, while for pea seeds lots varies from 14.94 to 28.04 % d.b and 80 to 60 %, respectively. Physical properties of all the three seed lots of both okra and pea were found dependent on its moisture content as well as germination percentage. The effect of hydro-priming process parameters was determined on responses viz. moisture content after hydro-priming (MC), germination percentage (FGP), seedling length (SL), seedling dry weight (SDW), vigour indices (VI- I and VI- II) and electrical conductivity (EC). Multiple regression models were found to be significant for all the process responses. Optimal solutions were found to be 73 % (IGP), 6h (SD), 23°C (Temp), 330 (rpm) and 2.50 m^3/min (air flow rate) for okra seeds while 75 % (IGP), 55 min (SD), 20°C (Temp), 320 (rpm) and 0.50 m^3/min (air flow rate) for pea seeds. The fabrication cost of the seed priming equipment of 5 kg for okra/ 3kg of pea was estimated Rs. 12,000. Total operating cost was calculated as 55 Rs/h. Operating cost was Rs. 66 Rs/kg for okra and 17 Rs/kg for pea seeds. BEP was found to be 31.76 h /yr which is 12.70 per cent of annual utility of 250 hours (Mahawar, 2015).

Study was undertaken to develop a firmness sensor to assess ripening of banana fruit during storage. Green, unripe, mature bananas were stored at 11 different storage conditions and physiological properties relevant to ripening were measured. During storage fruit firmness, pulp firmness and peel thickness decreased while TSS, peel thickness, pulp to peel ratio, physiological loss in weight and degreening increased with storage period. Correlations between firmness and other measured properties were evaluated. Firmness sensor was developed based on the firmness behavior observed during storage. It consisted of an especially designed electronic sensor supported on a frame. It was observed that damage to the fruit surface was negligible when firmness sensor was used to measure fruit firmness. Correlation coefficient between firmness measured with firmness sensor and other physiological properties were found more than 0.9 indicating that firmness sensor developed during work could be used to assess the ripening of banana (Kale, 2009)

An experiment was conducted on flexible hermetic storage system for paddy. Five packaging materials viz. jute bag, HDPE woven lined, HDPE woven un-lined bags, polypropylene bags and super grain bags were selected for the study. Two varieties of paddy, fine (pusa uugandh5) and coarse grains (suvarna) were selected for the study. The observations noted were, moisture content, relative humidity, temperature, oxygen, carbon dioxide and insect count. Highest moisture content (17.41% w.b.) was observed in grain stored in HDPE woven unlined bag and lowest (13.41 % w.b.) in grain from super grain bag. The super grain bag resisted moisture migration due to its lowest water vapor transmission rate compared to other packages. Lowest oxygen content (3.98%) was measured in super grain bag. And highest carbon dioxide measured in the same bag of value 16.67%. Whereas, insect dropped up to 3 in numbers in super bags. Jute bag, woven lined bag and woven unlined bags were found unable to stop moisture migration to and from the grain stored in them. Super grain bag is the best among the treatments for creating storage environment of low oxygen and high carbon dioxide content. The super grain bag is economically viable to store paddy for full one year or more. The polypropylene bag was economically viable to use for less than a year of paddy storage (Divekar, 2016; Divekar *et al.,* 2016)

A method has been developed which allows continuous measurement of the moisture content of maize grains. It consists of Support frame, Pulley, String, Sample container and Top pan weighing balance. The weight of sample container was transferred with the help of inelastic string passing over the pulleys. This system was used to weigh the sample up to 2kg. Experiments were conducted at four Inlet air temperature levels (40, 50, 60, and 70^{0}C) and three bed air levels (20, 30, 40 m/min). The results show that temperature is an effective factor on the drying rate. With increase in drying air temperature drying rate increases. The moisture ratio reduced sharply up to 25 min of drying time, after that it decreases with drying time but at slow rate. The drying constant increased with increasing air temperature and was maximum (0.03 min^{-1}) for a combination of 70^{0}C air temperature and 40 m/min air flow rate. The drying of maize grains occurred during the falling rate period and that no constant rate period was observed. The effect of air flow rate was negligible for all the drying air temperature (Yadav, 2012).

A study was undertaken for to Develop and evaluate the Garlic Peeler. The engineering properties of garlic relevant to peeling operation were evaluated. The length, width, thickness, geometric mean diameter and sphericity of garlic segments at moisture content of 40.50 percent (w.b.) were observed to be, on an average, 26.25, 10.36, 8.73, 13.34 and 0.51 mm respectively. The angle of repose, terminal velocity and static coefficient of friction were varied from 25.50 to 37.50 degree, 7.18 to 12.24 and 0.337 to 0.532 m/s respectively. The developed garlic peeler consisted of a feed hopper, roller and concave mechanism, blower and power transmission system. The peeling efficiency, yield of peeled garlic, yield of unpeeled garlic, damage, and peel separation were found to be 86.60, 86.20, 4.70, 9.15 and 96 % respectively with a machine capacity of 27 kg/h and the energy requirement of 1.15 kw-h (Manjunatha, 2007).

The present work was undertaken to study the production of onion flakes using solar dehydrator. It was observed that the variety, chemical and chemical concentration level influenced the drying rate and the physic-chemical and sensory attributes of dehydrated onion. The increased chemical concentration level bettered the product with regard to physic-chemical and sensory attributes. Air velocity (0.014-0.044 m/s) had negligible effect on drying since most of the drying occurred in the falling rate period. Model was developed to find out the heat available in solar collector as a function of ambient conditions *viz*. temperature, relative humidity and solar radiation. Also model for predicting the drying time based on moisture content, to find out weight of drying onion based on drying time and to predict the weight loss of dehydrated product based on drying time was developed (Edukondalu,2007)

A solar powered screen cleaner with higher cleaning efficiency and minimum cleaning losses was designed and fabricated using mild steel angle iron and flats for frame, hopper, screen cradle, screen, screen oscillating mechanism, power transmission system and solar power pack. Screen cleaner was operated with DC motor; 24 V, 250 W and energized with direct coupling to three solar photo voltaic panel of 210 Wp connected in parallel. Its cleaning capacity was found in the range of 100-150 kg/h. It had feature for adjusting the slope, pitch, screen type and size. The optimum performance was achieved at screen cradle slope of 5^0, screen oscillation of 250 strokes per minute and feed rate of 150 kg/h. The highest cleaning efficiency for chickpea, soybean and lentil was 88.27%, 87.14% and 84.3%, respectively at 90% purity level. Losses were found 2.12%, 1.25% and 1.73% at 90% purity level, respectively. The total cost of machine was Rs.47100/- and its estimated cost of operation was Rs. 0.269/- per kg. This cost of operation is much lower than cost of manual cleaning (Rs. 12.73/- per kg). The solar powered screen cleaner had a breakeven point at 393.33 $h.yr^{-1}$ with a pay- back period of 2.48 years (Aradwad, 2014)

A study was undertaken to study the milling characteristics of pigeonpea and to optimize the premilling treatment for maximum dal recovery with IARI mini dal mill. Milling by IARI Mini Dal Mill yielded finished product recovery : 72.83% and hulling efficiency: 90.47% with pre-milling heat treatment at 85 °c for 4 minute and hydrothermal treated lot resulted finished product recovery: 74.96% and milling efficiency: 90.77%. Further increase of product recovery and dehulling efficiency was observed with scoring the kernel before treatment. Cost of processing was to be 1.25Rs./kg and 1.60 Rs/kg for heat and hydrothermal pre-milling treatment and milling by IARI Mini Dal Mill, respectively (Indore, 2013).

Engineering properties of selected seeds (botle gourd, sponge gourd, garden pea and radish) were determined and results depicted that size, angle of repose, coefficient of friction, terminal velocity increased with an increase in moisture content. On the other hand, bulk density and true density decreased with moisture content. For bottle gourd seeds, the size was found to be maximum (7.03±0.19 mm), sphericity value was highest for garden pea seeds (0.83±0.05). Angle of repose was highest for garden pea seeds (26.37° ±1.23°) and coefficient of friction was highest for bottle gourd seeds (0.582±0.047). Terminal velocity, drag coefficient, bulk density and true density values were highest for garden pea seeds (10.13±0.21 m/s, 0.92±0.062, 729.66±7.36 kg/m^3 and 1258.37±5.91 kg/m^3, respectively). Number of blades and tilt angle were optimized to get maximum velocity and minimum power consumption. Result revealed that 4 blades with 70° tilt angle yielded maximum air velocity with minimum power consumption. Based on the selected parameters a solar based pneumatic seed cleaning

system was developed**.** Developed machine was found to have more than 90% of the cleaning efficiency for all the seeds under study. Besides, all the seeds had more than 99% physical purity after cleaning. The cost of operation of solar operated screen cleaner was calculated to be Rs 10.08 per h with through put capacity of 80 kg/h, (Mishra, 2016)

A weight Grading Machine for Oranges was developed. It consists of elements such as orange feeding unit, main frame and chain conveyor, electronic weighing assembly, laser sensing assembly, dropping and collection unit and power transmission system. The P CB was designed and developed with microcontroller, IC's and other electronic components mounted on it. The software programme in C language was written for microcontroller. The machine is capable of individually feeding and weighing fruits and grading them in four different commercial weight grades of less than 100g, 100-150g, 150-200g and greater than 200g. The different grader variables such as speed (12.2, 15 and 20 rpm) and microcontroller settings at three levels were evaluated for their effect on performance of orange grading machine. The capacity in number of fruits/h, grading efficiency in performance parameters studied. The final performance evaluation was carried out for selected speed of 15 rpm and microcontroller setting II. The grading machine performed satisfactorily while testing. The capacity of grading machine was found to be 2275 fruits/h, grading efficiency was 90%, energy requirement was 0.5 kw-h and mechanical injury was 2% (Gaikwad, 2010).

Summary

During the period of 2007-2016, numbers of innovative and adoptive technologies and equipments were developed by the M.SC./M.Tech. and Ph.D. students of all three sub-disciplines. In the sub-discipline of Soil and Water Conservation Engineering, studies were conducted under the themes namely management of surface and groundwater resources, climate change; irrigation and CO_2 emission, irrigation water management in canal command, enhancing water use efficiency through fertigation; mulching and irrigation scheduling; crop water foot print assessment for sustainable agriculture, management of saline and waterlogged areas. In the sub-discipline of Farm Power and Equipment studies were conducted under themes of design and development of farm machinery and mechanization, ergonomic design modification of manual tools. Sub-disciplines of Agricultural Processing and Structures focused on rural storage structures and processing equipment. Brief summary of the studies conducted are given below.

- Modelling procedures and protocol for assessing the ground water recharge, seawater intrusion, and nitrate pollution of groundwater and impact of climate change on groundwater were developed
- Combination of management options namely reduction in ground water pumping by 25% from unconfined aquifer, increase in pumping by 25% from semi confined aquifer, increased recharge by maintaining stage in river to 0.5 m during monsoon and 0.25 m during non monsoon was suggested as an alternative to control seawater intrusion and improve groundwater quality in the aquifers of Minjur in Tamil Nadu
- Rainwater harvesting structure were suggested for the Arvari River Catchment
- Nitrate Pollution of Groundwater at IARI Farm was assessed
- Performance of filtration unit of groundwater recharge shaft was evaluated and appropriate filtration layer thickness ratio was suggested
- CO_2 emission from groundwater irrigation under various climate change scenarios were assessed and appropriate mitigation strategies were suggested
- Impact of climate change on drought occurrences in Pune district, Maharashtra, India was investigated
- Water Delivery Schedule for Jhajjar distributary in the Western Yamuna Canal Command of Haryana was developed using Geospatial Tools and CROPWAT model

- Studies were conducted water and nitrogen movement under drip fertigation using simulation model.
- Appropriate guidelines for design and operation of drip fertigation system were developed for various crops
- Fuzzy neuro model for drip irrigation scheduling of greenhouse cut flower rose was developed

  Operational guideline for utilization water stored in pond was developed using Neuro-Fuzzy modeling
- Effect of deficit irrigation and split dose of N-fertilizer at different growth stages of wheat on its growth and yield was evaluated and fertilizer application and irrigation schedule was developed
- Multiple linear regression models to estimate evaporation from silty clay loam and sandy loam soils was developed
- Regulated deficit irrigation (RDI) and partial root zone drying (PRD) techniques of water saving were evaluated. Based on the results, PRD was recommended for greenhouse tomato in areas where water is scarce and expensive
- The characteristics of municipal wastewater available at IARI was characterized for the possible use for irrigation through surface and subsurface drip system
- Response of different colored plastic mulches on yield and crop water requirement of drip fertigated capsicum under greenhouse were evaluated and best colour of plastic mulch was suggested
- Low cost, user friendly soil moisture sensor (Gypsum Block) for real time monitoring of irrigation was developed and inked with Wireless Sensor Network (WSN) using GSM module comprising with a microcontroller unit.
- Automated irrigation system was developed by integrating sensors network with tele-communication technologies having modified tensiometer, level sensor, controller, GSM, receiver, transmitter, solenoid valve, water meter and pump.
- Low cost, user friendly soil moisture sensor and LM35 IC based sensor circuit capable of measuring leaf -air temperature differential was developed
- Performance of different soil moisture sensors (FDR, tensiometers and gypsum blocks), were evaluated to develop the guidelines for use of soil moisture sensors at field level in a Cole crop (broccoli)
- Performance of AquaCrop Model to predict the grain yield of Maize under Different Nitrogen and Water Availability scenarios was evaluated.
- Single and dual crop coefficients of mustard were determined from the experiment conducted in lysimeters
- Wheat yield under irrigated saline environment was simulated using AquaCrop model and the production functions to predict the grain yield under different salinity regimes, irrigation and rainfall depths besides the quantity of foliar potassium fertilizer was developed
- Irrigation guidelines for the greenhouse capsicum grown in soilless media was developed based on field experiment on effect of electrical conductivity on the crop water uptake and simulation results of water uptake
- Methodology for crop water foot print assessment at rivers basin scale and virtual water content was developed and the sustainability of water foot prints within the Gomti and Betwa river basins of India was determined
- A decision support system for reclamation and management of salt-affected soil was developed
- Spatial prediction of soil salinity for subsurface drained agricultural field in Western Yamuna Canal Command(WYCC) area, Haryana, India was carried out using geostatistical approach and the impact of drainage on soil salinity was evaluated

- Salt dynamics and its budgeting in the crop root zone, the conversion factors and regression equations for estimation of salinity of saturation paste extract (EC_e) from $EC_{1:2}$ and $EC_{1:5}$ were developed to minimize the estimation time without use of laboratory procedures. Salt tolerant variety resulted in higher yields and water use efficiencies with saline irrigation. Besides this, the salt dynamics in the root zone depths and crop yield simulated by the crop models would assist in scheduling of irrigation, leaching of salts from crop root zone and conjunctive water use planning for enhancing productivity under irrigated saline environment
- Monitoring and simulation procedures for assessing waterlogging and salinity in irrigation command using GIS and Remote Sensing were developed
- Seedling transplanting mechanism that can eject seedlings at the same speed of forward travel, but in an opposite direction was designed and fabricated.
- A mechanical hydrogel applicator was designed and fabricated
- A web based custom hiring model to suggest the fleet size, investment required, economic machinery size, operation scheduling and to facilitate decision making for custom hiring of combine harvester was developed
- Tractor drawn onion digger was designed and developed
- Foliar Applicator for Urea Ammonium Nitrogen was designed and Developed
- Feasibility of soil nitrogen detection using electrical conductivity for site-specific application was studied
- Effect of rotary tillage on soil physical properties and crop response was investigated. It was observed that continuous use of rotary tillage deteriorated the soil structure and hindered root growth
- Tractor operated semiautomatic onion transplanter to mechanize onion transplanting operation was designed and developed
- Mechanical transplanter for onion seedlings was designed and developed
- Precision planter for direct seeding of paddy was designed and developed.
- Design parameters of Aqueous Fertilizer placement with Seed Drill for dryland agriculture were studied. It was observed that increased rate of aqueous fertilizer gave enhanced growth performance parameters in addition to better germination
- Permanent raised bed seed-cum-fertilizer plot drill was designed and developed. A prototype permanent raised bed seed cm fertilizer plot drill with four row distributor and the plain rolling coulters followed by double disc furrow openers was designed and fabricated based on the findings of laboratory and soil bin studies and optimum operating conditions and design values obtained.
- Design parameters effecting mechanical carrot harvesting was studied. The maximum percentage of carrots harvesting of 97.4 per cent was obtained at 60 cm length of soil separator, 25° of rake angle and 20° of angle of soil separator. Minimum percentage of carrots damage of 4.87 per cent was obtained at 40 cm length of soil separator and 20° of soil separator angle
- Collector-cum-chopper for paddy straw management was developed. The machine was fabricated after determining the physical and mechanical properties of paddy straw by Ti + DA texture analyzer. The fabricated prototype of paddy straw collector-cum-chopper comprised of three sub-systems i.e. straw collection and conveying unit, straw chopping unit and discharging mechanism with provision to apply optimised dose of fungal inoculum
- Ridge profile power weeder was designed and developed
- Pressurized Aqua-Fertilizer Metering system of a seeder was designed and evaluated
- Mechanical Urea Briquette Applicator for System of Rice Intensification was developed
- Multi-crop planter for hill agriculture was developed

- Design and operational parameters of compost-turner-cum-mixer was optimized
- Design parameters of mechanical harvesting of garlic were studied
- Dynamic Stability of Tractor-trailer system was studied. It was observed that the safe level of stability was more in case of double axle trailer operation than that of single axle trailer in terms of normal load carrying capacity, operation of system up to 5° with higher forward speed up to 4.056 km/h as compared to single axel trailer.
- Self Propelled garlic planter and oscillating soil separator for garlic harvester were developed
- Metering system for variable rate fertilizer applicator was design and developed
- Design parameters of a mechanical carrot planter were studied and the prototype of planter was designed and fabricated using necessary components for raised bed planting of carrot seeds
- Ergonomic assessment of manual load carrying among women farm workers was done and ergonomic design modification in intercultural hand tools to make female worker friendly was done
- Agricultural- Dust Protective Interventions for farm workers was developed,
- Interventions to reduce vibration transmission to power tiller operator in various field operations were developed
- Ergonomic variable height platform for training and harvesting of tomato crop in greenhouse was design and developed
- Safe enclosure for tractor operators was designed and developed
- Comparative study of three types of manual weeder namely wheel hoe, crescent hoe and dry land weeder were ergonomically evaluated for women farm worker. It was observed that heart rate with modified wheel hoe reduced by 11 beats per minutes as compared to the existing wheel hoe
- Shoulder mounted harness was designed for women farm workers for carrying load which did facilitate load carrying similar to head loading without introducing element of eccentricity.
- Evaporative cooled storage structure (ECSS) was designed to assess the performance with three different pad materials and four distinctive pad areas in no load condition
- Effect of thermal and oil pre-treatment on the recovery of green gram dhal was evaluated. The dhal recovery was found to increase by about 15% after oil treatment and decreased after thermal treatment
- Equipment for hydro-priming of vegetable seeds was developed and its performance was evaluated for priming of okra and pea seeds.
- Firmness sensor to assess ripening of banana fruit during storage was developed. It was observed that damage to the fruit surface was negligible when firmness sensor was used to measure fruit firmness
- Experiment conducted on flexible hermetic storage system for paddy revealed that the super grain bag was economically viable to store paddy for full one year or more
- Cross-flow thin bed drying characteristics of maize (zea mays) grains using continuous sample weight measurement were studied and method for continuous measurement of the moisture content of maize grains
- Garlic peeler was developed and evaluated. Garlic peeler consisted of a feed hopper, roller and concave mechanism, blower and power transmission system.
- Model for predicting the drying time based on moisture content, to find out weight of drying onion based on drying time and to predict the weight loss of dehydrated product based on drying time was developed
- Solar powered screen cleaner with higher cleaning efficiency and minimum cleaning losses was designed and fabricated using mild steel angle iron and flats for frame, hopper, screen cradle, screen, screen oscillating mechanism, power transmission system and solar power pack

- Milling characteristics of pigeonpea were studied and premilling treatment for maximum dal recovery with IARI mini dal mill were optimized
- Solar based pneumatic seed cleaning system was developed
- A weight grading machine for oranges was developed. It consists of elements such as orange feeding unit, main frame and chain conveyor, electronic weighing assembly, laser sensing assembly, dropping and collection unit and power transmission system. The capacity of grading machine was found to be 2275 fruits/h, grading efficiency was 90%, energy requirement was 0.5 kw-h and mechanical injury was 2%

Future Perspective

Division will like to focus on following issues in future

Soil and Water Conservation Engineering

- Assessment, utilization and management of groundwater resources, surface and groundwater interaction, groundwater recharge and impact of climate change on regional hydrology
- Irrigation planning and enhancing water productivity in water scarce and water excess areas.
- Nexus between water, energy and nutrient
- Watershed hydrology for rainwater harvesting and management; river basin hydrology and flood routing
- Modern tools and techniques (Remote Sensing and GIS) in watershed/ natural resource management in varying Bio-geo-hydrologic settings
- Water measurement devices for irrigation scheduling
- Fertigation scheduling and nutrient dynamics under green house cultivation and design of micro Irrigation systems for canal commands
- Farm Power and Equipment
- Design and development of machinery for biomass utilization, accelerated compost making and applications
- Agricultural mechanization pattern and modeling, soil compaction study, modeling and machinery design for its management
- Design and development of precision machinery
- Ergonomic assessment of agricultural operations and machines/tools and intervention for development of gender friendly farm equipment
- Variable rate application systems for seed and fertilizer
- Drone assisted disease monitoring and spraying system
- Design and development of Machines for horticultural mechanization, hill agriculture and Small farm mechanization
- Automation of agricultural machines for precision application through use of mechatronics and embedded system design
- Design and development of senor based precision planting system for row crops
- Agricultural Structure and Process Engineering
- Solar powered technologies for farm operations and rural structures
- Low cost, low carbon foot print cool storage structures for perishable products
- Solar energy based technology for farm protection

Acknowledgments

Authors wish to thank the Faculty Members of the Division of Agricultural Engineering for the support and cooperation extended by them in preparing the research contributions of M.Sc. and Ph.D. students.

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Agricultural Physics

Vinay Kumar Sehgal

Introduction

The scientific understanding about the role played by the physical processes and their significance for sustenance of the soil-water-plant-atmosphere continuum (SPAC) could be realized only when demarcation of science was clearly made into biological (living) and physical (non-living) streams. In the history of Agricultural Sciences, Agricultural Physics is perhaps the youngest and the only discipline which deals with the basic laws of physics, and energy/matter interactions, in the context of crop growth and development. Agricultural production can be sustained on the soil, if all the agricultural operations are planned keeping in view the physical, climatological and hydrological factors in relation to the physical forces and conditions in the atmosphere at optimum dynamic equilibrium, under which, seeds and plants survive and remain productive.

Starting from a small unit "Physics in Agriculture" created in 1948, the Division of Agricultural Physics was established as a full-fledged teaching and research discipline in 1962. Endowed with a mission to study Soil-Water-Plant-Environment Energetic for eco-friendly and sustainable exploitation of agricultural resources, the Division has four sub-disciplines: Agricultural Meteorology, Plant-Biophysics, Remote Sensing and Soil Physics. During the last five decades or so, all these formed the major pillars of the edifice of Agricultural Physics as a Discipline. The mandates of the Division are:

- To conduct strategic multi-disciplinary research for understanding the physical processes of crop plants grown under varying soil and atmospheric environments for enhanced and sustainable agricultural productivity
- To organize upgraded post-graduate research and teaching activities in the areas of Soil Physics, Agricultural Meteorology, Plant-Biophysics and Remote Sensing applications.
- To maintain meteorological observatory, create agro-physical information database and disseminate it for use by students, researchers, farmers and planners.
- To prepare real time medium range weather-based agro-advisory bulletins for the farmers of NCR-Delhi region.
- To conduct regular training courses on Agricultural Meteorology, Soil Physics and Remote Sensing and their applications in agricultural resources planning and management at National and International levels.

Significant Student's Research Achievements

The master and doctorate students of the discipline, under the able guidance of faculty, have contributed significantly towards the research achievements of the Division. The sub-discipline wise significant research achievements of the students during 2007-16 are presented below:

Agricultural Meteorology and Crop Modelling

An experiment was conducted on three wheat cultivars (HD-2967, W-544, PBW-502) to study the change in quantity and quality of incoming solar radiation on their growth and yield in rabi 2015-16. Four radiation regimes (Control, 25%, 35%, 50% and 75% reduction as compared to control) were created by using successively increasing layers of shade net which resulted in decrease in direct but increased diffuse radiation. Measurements

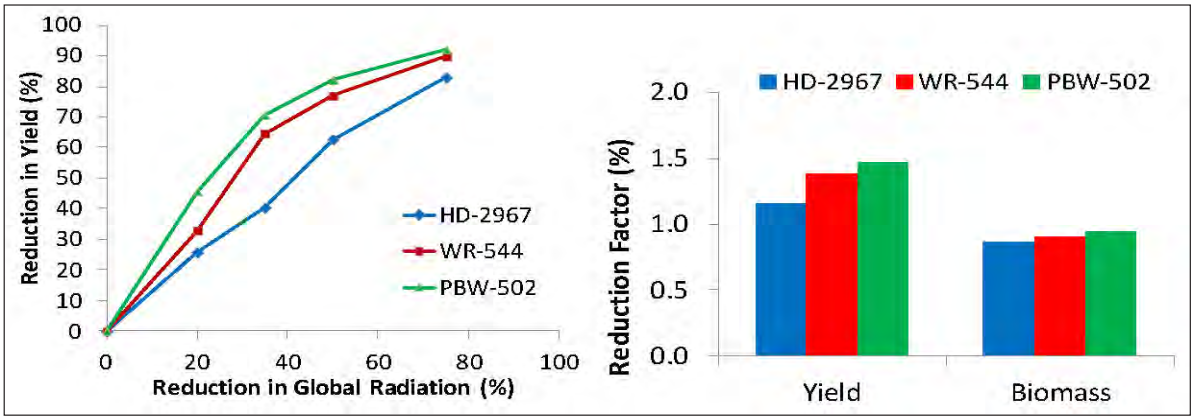

Fig. 1: Reduction in wheat yield with reduction in incident seasonal radiation for three wheat cultivars and their corresponding reduction factor.

on total and diffuse radiation, crop LAI, fIPAR and biomass were measured at different growth stages. The results of experiment showed though there was an increase in LAI at 35% reduction but crop biomass, photosynthesis rate, stomatal conductance and yield decrease with reduction in global radiation though radiation-use-efficiency increased. It was found that with the reduction in radiation, the wheat yield reduced least in HD-2967 and maximum in PBW-502. Upto 40% reduction in radiation, the yield reduction factors varied between 1.0 to 1.5 while for above ground biomass it varied from 0.87 to 0.95, implying that harvest index also reduced with the reduction in radiation.

The effect of different row directions, viz. north-south (N-S) and east-west (E-W) on micrometeorological parameters and its impact on crop growth parameters were studied in the experimental farm of the IARI) during rabi season of 2013-14 with three oilseed *Brassica* cultivars (Pusa Vijay, Pusa Mustard-21 and Pusa Bold). The micrometeorological parameters like IPAR, fIPAR, temperature and humidity within crop canopy differed significantly for different row directions during pod development and seed filling stages. N-S direction intercepted PAR significantly higher than E-W direction. Soil moisture extraction was also better in N-S direction.

Fig. 2: Mustard sown in two different row directions at different growth stages.

LAI, biomass, seed yield and oil content was also significantly higher in N-S direction probably due to better interception of PAR and soil moisture extraction. Temperature with in canopy was higher in case of N-S direction and relative humidity was lower in case of N-S direction. Aphid infestation and white rust infection were significantly lower in N-S direction probably due to lower relative humidity. Traditionally, mustard crop is sown by broadcasting or line sowing in E-W direction in N-S India. If it's sown in N-S direction, better yield and oil content expected. It can be a component of IPM.

To characterize the microenvironment of rice-mustard cropping system and the relationship of micrometeorological parameters with growth, yield, pest and diseases of rice-mustard cropping system under

conservation agriculture a study was conducted in the experimental farm of IARI, New Delhi during *kharif* and *rabi* seasons of 2014-15 with rice cultivar PRH-10 and mustard cultivar PM-25 with following treatments: T1 – ZT DSR (Zero tillage direct seeded rice) – ZTM (Zero tillage mustard); T2 - ZT DSR + BM (Brown manuring) – ZTM (Zero till mustard); T3 - MR (Mustard residue) + ZT DSR – RR (Rice residue) + ZTM; T 4 - MR + ZT DSR + BM –RR + ZTM; T5 - MBR (Moong bean residue) + ZT DSR – ZTM; T6 - MBR + ZT DSR – RR + ZTM ; T7 - TPR (Transplanted rice) – ZTM; T8 - TPR – CTM (Conventional till Mustard). Highest LAI was observed in one conventional treatment of rice (T8) and in one conservation agriculture treatment (CA) in case of mustard (T6). PAR interception also followed similar pattern as above. More interception of PAR produced higher biomass and yield. Profile temperature in clear and calm day was in increasing order from bottom to top. The profile temperature was cooler in CA treatments in Mustard but in conventional plots in case of rice. Profile Relative Humidity showed the reverse trend. Soil surface moisture was more in CA plots in case of mustard while in conventional plots in case of rice. Soil surface temperature was more in conventional plots in case of mustard. Pests and diseases were found more in conventional although T2 of conservation treatment showed highest incidence. T6 (MBR+ZTDSR-RR+ZTM) showed overall best result in this rice mustard cropping system under conservation agriculture.

Energy/heat fluxes over a cropland are important to understand the interaction between the crop surface and atmosphere including practical applications in crop yield prediction and water resources management. A large aperture Scintillometer (LAS) instrument, complemented with an automatic weather station, has been installed in the experimental covering a path length of 990 m of intensively cultivated irrigated agricultural landscape. The diurnal and seasonal patterns of radiation, sensible and latent heat fluxes and evaporative fraction were measured analyzed for maize in *kharif* and wheat in *rabi* seasons of 2014-15. The biophysical parameters (Leaf area index, soil moisture, crop height) were recorded at a temporal resolution of fortnight basis along the LAS path length at usual sampling distance. Leaf area index had a significant positive correlation with latent heat flux (r = 0.56) and a negative correlation with sensible heat flux (r = -0.63). Soil moisture had a significant negative correlation with sensible heat flux (r = -0.77). The average evapotranspiration from cropland was 1.58 mm d^{-1} and total evapotranspiration was 543 mm over the twelve months study period. The Bowen ratio value for both *Kharif* and *Rabi* seasons was 0.75 and 0.57, respectively.

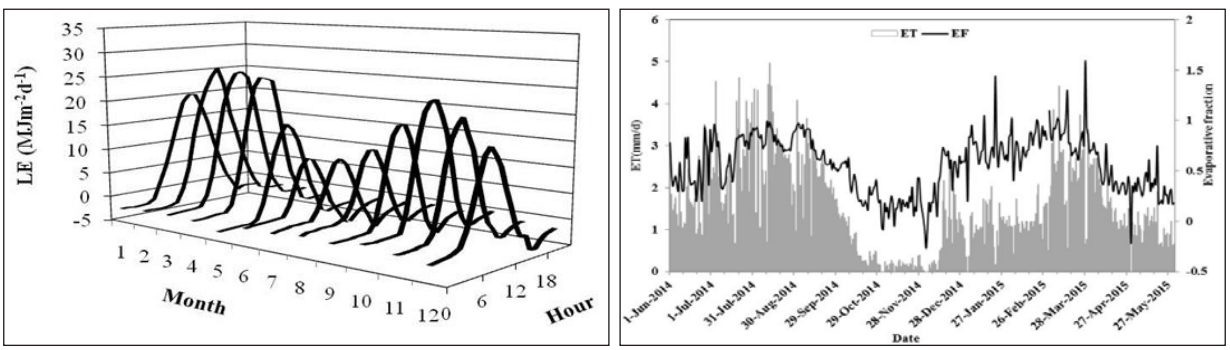

Fig. 3: Graph showing month-wise diurnal variation in latent heat flux over the year and the daily variations in actual transpiration and evaporative fraction.

Affect of high temperature stress on HD 2987 cultivar of wheat under controlled condition (T1-ambient, T2-ambient + 1.5°C, and T3- ambient+3.0°C) using Free Air Temperature Increment Facility and under different dates of sowing (S1-20 Oct, S2- 1 Nov, S3- 15 Nov, S4- 1 Dec and S5- 15 Dec) were studied during *rabi* seasons of 2012-13 and 2013-14. The results showed that temperature stress conditions affected wheat crop growth at various stages by significantly decreasing plant biomass and grain yield. High temperature stress conditions also modified the radiation regime of canopy as reflected by the changes in fIPAR and LAI indicating great variation in leaf area with temperature stress. Changes in biomass partitioning clearly showed decrease in

biomass allocation to storage organs under high temperature stress condition, both under Free Air temperature Stress conditions (T2 and T3) and late sown conditions (S4 and S5). Apart from the significant changes in the above ground phenology of wheat crop, high temperature stress affected soil parameters by decreasing the soil moisture content and increasing soil temperature during the later stages of the wheat crop.

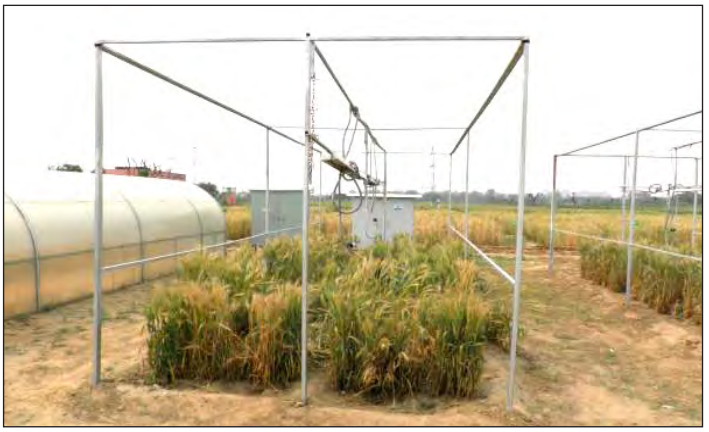

Fig. 4: Free Air Temperature Increment (FATI) facility for inducing high temperature stress on wheat crop at the experimental farm of the Agricultural Physics.

Comprehensive evaluation of three wheat crop models INFOCROP, DSSAT and APSIM for its ability to simulate the effects of high temperature, for wheat in semi-arid region, India was conducted. The models were evaluated for their ability to predict crop development, grain yield, biomass production over time and Leaf Area Index, using two years of data from field experiments. The 2012-13 data were used to calibrate the three crop simulation models, while 2013-14 data were used for evaluating the performance of the model simulations. A high value of R^2 above 0.8, normalized root mean square error below 15% between simulated and observed values for yield showed high accuracy of model predictions for a wide range of wheat treatments. However a low value of R^2 below 0.8, for biomass and crop growth stages clearly showed that crop simulation models have inherent structural deficiencies and need to be suitably modified to simulate the high temperature stress conditions.

An experiment was conducted during summer and rainy season 2012 at research farm of IARI, New Delhi, with an aim to know micro-environment under different colour shade nets and its influence on biophysical parameters of spinach. Spinach (variety Pusa bharti) was sown on 28th April in summer season and on 6th July, 2012 in rainy season respectively. The crop was raised under white, black, red, green colour shade nets and control (without shade net). Results showed that the microenvironment was changed under different colour shade nets in both the season but the difference was more in summer than in rainy season. The air temperature, wind speed, soil temperature, canopy temperature, light intensity, radiation and different accumulated heat indices such as GDD, PTU, HTU was found to be lower under different colour shade as compared to the corresponding value under control. However the relative humidity and soil moisture had higher value under colour shade nets than corresponding value in control. The percentage reflectance as well as value of NDVI, VI and SIPI was found to be more in green followed by red, black, control and white. The heat use efficiency had higher value under green shade nets followed by red, white, black and control. Among the different colour shade used in present study green shade nets was found better followed by red, black and white.

In order to optimize the growth and seed yield in any crop, quantification of Crop-Weather relationships could help in determining proper time for sowing. Evapotranspiration accounts for major water loss from the agricultural fields. So, a field experiment was conducted at research farm of IARI, New Delhi during Rabi 2012 for understanding crop water needs required for irrigation in mustard. Three varieties of mustard viz., Pusa Gold, Pusa Jaikisan and Pusa bold were sown on 14th October, 31st October and 16th November, 2011 for creating different weather condition at different crop stages. The actual crop evapotranspiration were calculated using single crop coefficient, dual crop coefficient and water balance equation. Results showed higher value of biomass, seed yield, water use efficiency and radiation use efficiency in Pusa Jaikisan followed by Pusa Bold and Gold. The value of biomass, seed yield and radiation use efficiency was found to be more in first sowing followed by second and third sowing. The results showed that the value of soil evaporation coefficient was low except during irrigation and precipitation events. The value of water use efficiency calculated using dual crop coefficient were closer to the value of water use efficiency calculated by soil water balance as compared to the

value calculated by single crop coefficient. The crop evapotranspiration calculated from dual crop coefficient was better and more accurate for estimating water needs for mustard crop because water use calculated by dual crop coefficient consider both soil evaporation coefficient and basal crop coefficient. From the above studies it can be concluded that water need requirement in mustard crop can be estimated more accurately by dual crop coefficient approach as compared to single crop coefficient.

Field experiments were conducted during *rabi* season 2009-10 at research farm of IARI, New Delhi, with an aim to study the effect of aphid infestation on growth parameters of mustard under varying weather condition. Remote sensing techniques were also used to study the mustard aphid infestation. Three varieties of mustard viz., Pusa Gold, Pusa Jaikisan and Pusa bold were sown on 19th October, 3rd November and 18th November. The result showed that aphid infested crop had low LAI, chlorophyll concentration, seed yield and percentage oil content as compared to healthy crop. Due to more aphid infestation CATD value was higher in Pusa Gold as compared to other two varieties. The GDD based aphid forewarning model was found to be valid for forewarning of aphids. The crop growth and seed yield were relatively higher in the first sown crop because of more congenial weather conditions. First sown crop had higher value of LAI, biomass, chlorophyll concentration index, chlorophyll concentration, RUE, seed yield and percentage oil content as compared to late sown crop in all three varieties. Delay in sowing time reduces the above parameters significantly. Among the three varieties Pusa Jaikisan produced higher biomass, LAI RUE, seed yield and percentage oil content as compared to Pusa Bold and Pusa Gold in all three dates of sowing. Chlorophyll concentration index and chlorophyll concentration was found higher in Pusa Gold followed by Pusa Jaikisan and Pusa Bold in all three dates of sowing. Aphid infestation can be identified in the visible 550-560 nm, NIR region 750-1250 nm and 1950-2450 nm. Reflectance between 1950-2450 nm can be used for identifying different levels of aphid infestation in mustard crop. The different phenological stages of mustard can be identified using hyperspectral data. NDVI, RVI, SIPI and AI indices can be used for assessing aphid infestation in mustard. Due to aphid infestation crop growth and seed yield decrease significantly in late sown crop. Therefore delay sowing should not be recommended for mustard crop. Hyperspectral remote sensing could be a useful tool for monitoring the effect of aphid infestation in mustard crop.

Field experiments were conducted for two years (2011-12, 2012-13) on wheat cultivar PBW-502 with six treatments (three levels of irrigation and two dates of sowing) to calibrate and validate the AquaCrop simulation model. A high value of R^2 above 0.8, normalized root mean square error below 15% and D-index above 0.8 between simulated and observed values of profile moisture, biomass and yield showed high accuracy of model predictions for a wide range of wheat treatments. Irrigation scenarios varying in number and amount of

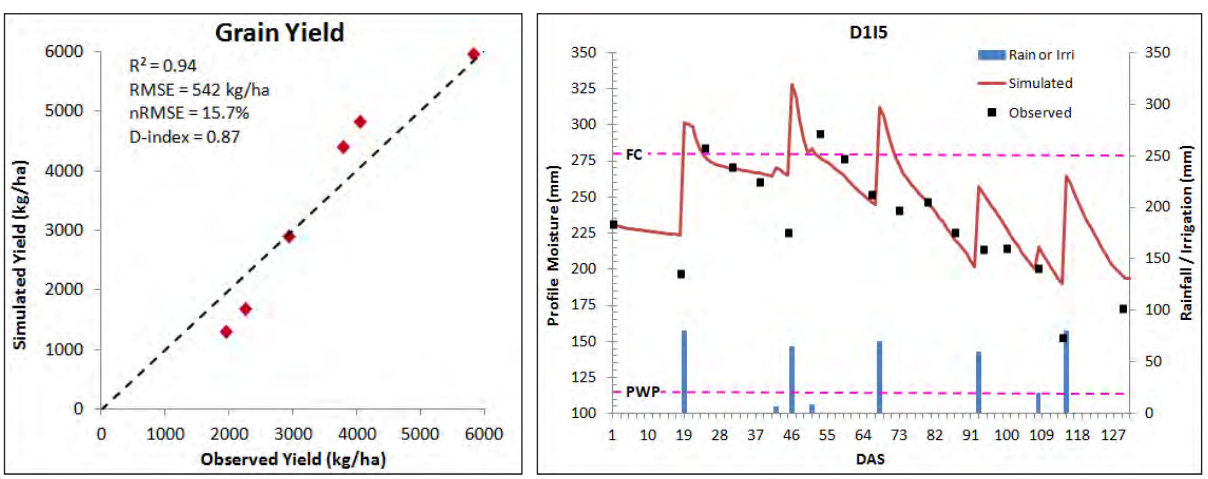

Fig. 5: Comparison of model simulated and observed wheat grain yield and profile moisture content for different treatments.

irrigations for normal sown wheat were evaluated for yield and water productivity by AquaCrop model. The analysis of scenarios showed that normal sown wheat yield can be maximized by applying three irrigations of 80 mm each at 21, 85 and 105 days after sowing (DAS) while its water productivity can be maximized by applying two irrigations of 60 mm each at 21 and 105 DAS with some yield penalty.

The study assessed the on-farm water requirement in wheat crop in future, in semi-arid Indo-Gangetic Plains of India, through field and computer simulations. Field simulation using temperature gradient tunnels shows 18% higher crop evapotranspiration (ETc) and 17% increase in root water extraction at 3.6°C elevated temperature compared to 1.5°C increase over the ambient. A time series model (ARIMA) with long-term (1984–2010) weather data of the experimental site and a global climate model (IPCC-SRES HADCM3) were used to simulate the potential ET (ET0) of wheat for 2020–2021 and 2050–2051 years. The crop coefficient (Kc) values for these years were generated through Kc–CGDD (Cumulative growing-degree-days) relation by using LARS-WG model-derived daily minimum and maximum temperatures. The CWR and NIR (Net Irrigation Requirement) are likely to be less in projected years even though air temperatures increase. Under IPCC-SRES scenarios, the ETc–crop phenophase relation [CGDD–LGP (length of growing period) response] may offset the effect of rising temperature and a net decline in CWR is observed. It may be likely that the effect of temperature increase on CWR is manifested mostly through its relation with crop phenophase (thermal requirement to complete a specific growth stage) and not the temperature effect on ET0 per se. This is certainly a ray of hope in managing the depleting irrigation water resources in the semi-arid wheat-growing regions of the IGP.

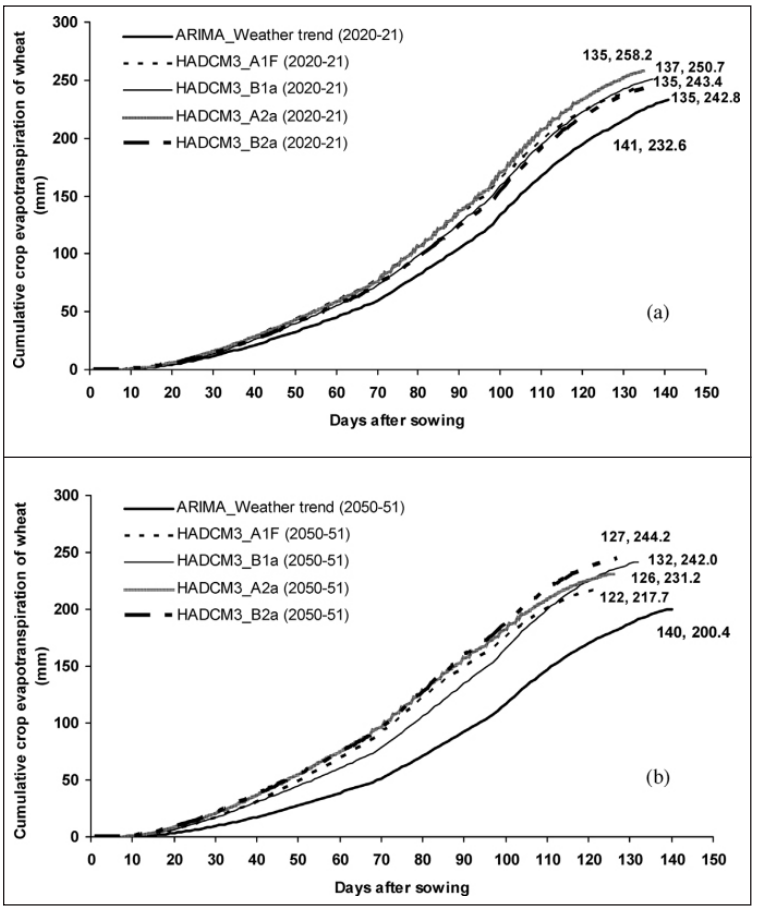

Fig. 6: Progressive crop evapotranspiration in wheat over the growing season under IPCC-SERC and ARIMA-model generated scenarios for 2020–2021 (a) and 2050–2051 (b); data represent duration of crop (days) and total evapotranspiration (mm).

An open top chamber (OTC) study was conducted with two replications to investigate the impact of elevated CO_2 (580 ppm) on canopy radiation interception and its use in relation to yield components of two pigeon pea (*Cajanus cajan* L.) cultivars Pusa-992 and PS-2009. Two OTCs with ambient CO_2 of 380 ppm were used as control. The LAI and above ground biomass were significantly higher during most of the growth stages for plants exposed to higher CO_2 concentration. The canopy radiation extinction coefficient (k) values for both the cultivars were lower for plants exposed to elevated CO_2 than for control plants, indicating a more erect structure of these plants. However, the radiation use efficiency (RUE) was 52.3% higher for plants grown under elevated CO_2 than for plants grown under ambient CO2 for both the cultivars. In cultivar Pusa-992, seed yield increased by 12.0% under elevated CO_2 because of increase in pod numbers and weight. But in this cultivar, the significant increase (40.6%) in biomass under elevated CO_2 did not translate into a corresponding increase in seed yield due to lower harvest index and less numbers of seed per pod. Under elevated CO_2, the other cultivar PS-2009

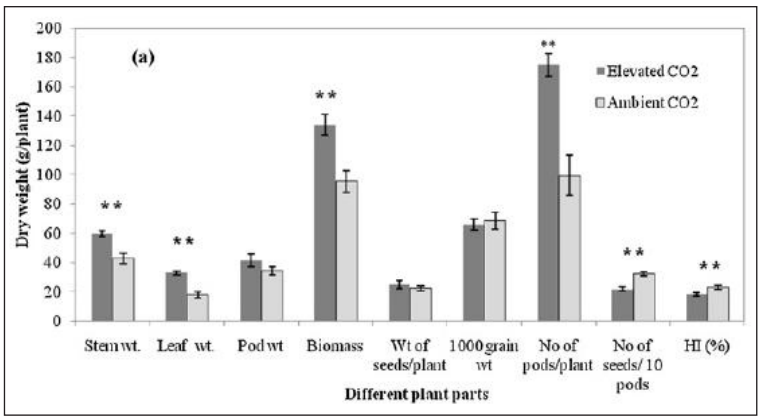

Fig. 7: Yield component at harvest under elevated CO_2 and ambient CO_2 of pigeon pea cultivar Pusa-992.

became indeterminate and did not mature, resulting in undeveloped pods. Hence in PS-2009, elevated CO_2 resulted in poor seed yield, pod numbers and pod weight even though the biomass produced was higher.

A field study was conducted to investigate the impact of elevated CO_2 (580 ± 20 µl l-1) on radiation interception, its use, other related physiological parameters, yield components, soil temperature, soil moisture depletion and its use, root characters, soil physical parameters and different fractions of active carbon for kabuli chickpea in Open Top Chambers (OTCs) and were compared with those grown under ambient levels of CO_2 (approx. 385 µl l-1) in similar OTCs. i.e. control. Though the cumulative radiation interception did not vary between the CO_2 treatments, the radiation extinction coefficient (k) value were lower for plants exposed to elevated CO_2 which indicated a more erect structure for these plants. However, the radiation use efficiency (RUE) was higher by % for elevated CO_2 grown plants than control. LAI was significantly higher during most of the growth stages and canopy temperatures were lower for plants exposed to higher CO_2 concentration compared to control. On per plant basis, seed yield increased by 82% due to elevated CO_2 exposure as the pod number increased significantly though seed test weight was unchanged. Even though elevated CO_2 resulted in increase in biomass, the present increase in seed yield was higher than the present increase in biomass causing increase in harvest index. Decrease in specific leaf area indicated thicker leaves as unit leaf area also increased with wider leaf C: N ratio due to higher C assimilation in leaf under elevated CO_2. The beneficial effect of elevated CO_2 was confined to increase in aboveground biomass production, reduced crop water use and greater nutrient availability in soil. On the other hand increase in labile fractions of soil C pools and decrease in grain quality accounted the negative impact of atmospheric CO_2 enrichment on chickpea.

Impact of drought on *kharif* crop phenology in Rajasthan using meteorological and remote sensing data for 24 years was studied. The length of season of *kharif* crop was derived from multi-temporal NOAA AVHRR PAL dataset. A significant linear relation between anomalies of length of season and seasonal rainfall was obtained in this study showing a shortening of 7 day for every 100 mm deficit in rainfall. Quantification of such relations using ground observations is difficult because of uncertainty in measurement of regionally representative length of crop season parameter.

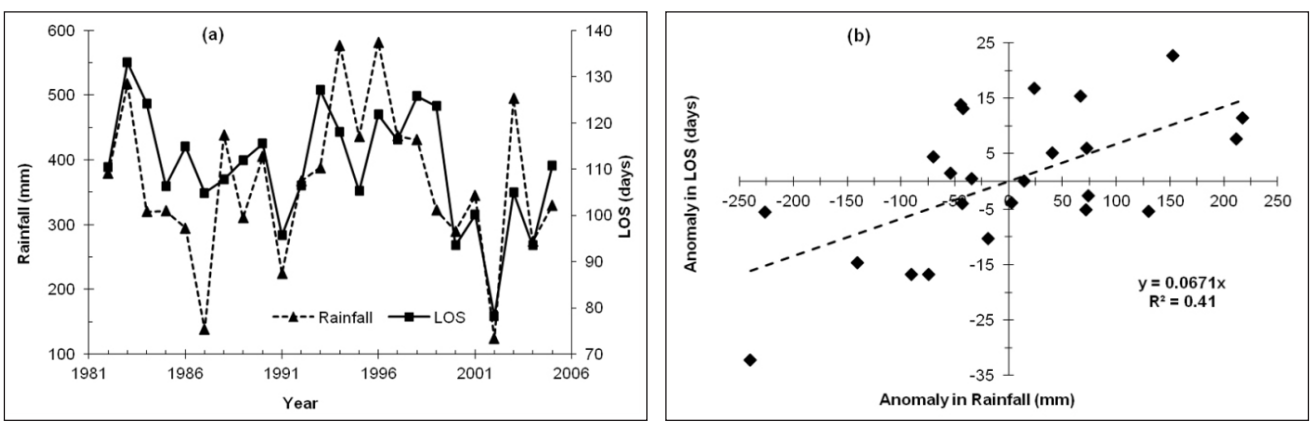

Fig. 8: Relation between seasonal rainfall and length of *kharif* crop season (LOS) in Rajasthan.

Multi stage wheat yield forecast at different growth stage of the crop were done at the district level using weather based statistical and crop simulation model (InfoCrop). Composite weather variables were calculated and model were developed using step-wise multilinear regression equation. These models were used to forecast yield at mid-season (45 days before harvest) and pre harvest stage (25 days before harvest). Yield forecast at different growth stage of the crop were also done by InfoCrop model. For calibration and validation of both models, field experiments were conducted at research farm of the institute during Rabi, 2012-13 and 2013-14. Analogous year approach was used to assimilate runout data from date of forecast to the date of harvest. Studies revealed that the InfoCrop model performs better than weather based statistical models for forecasting yield at different growth stages of the crop during both the years 2012-13 and 2013-14. Also the yield forecast done by both the model at pre harvest stage was better as compared to yield forecast done at mid-season stage. The R^2 for yield forecast done at pre harvest stage was 0.93 and 0.91 and at mid-season stage was 0.83 and 0.82 for InfoCrop and statistical models respectively.

Plant Biophysics

A study to characterize the Crop Status Index (CSI) was performed on wheat crop grown under eight abiotic stress conditions categorized into four levels of stress intensity (No stress, single, double and triple stress). Twenty probable crop abiotic stress indicators were used to select minimum dataset (MDS) responsive of different intensities of abiotic stress conditions. The study identified suitable abiotic stress indicators, the best scoring technique (linear or nonlinear) and appropriate method (additive and weighted) to derive a CSI to characterize abiotic stress condition like moisture, temperature and nitrogen stress alone or in different (single, double or triple) combinations of theses stresses. Twenty Biophysical, Physiological and Biochemical stress indicators comprised the initial dataset, out of which Relative Water Content, Stomatal Conductance, Leaf Temperature, Chlorophyll and Leaf Nitrogen were selected as MDS through PCA. The type of abiotic stress condition and stress intensity affected each indicator in different ways. CSI calculated by means of MDS and nonlinear weighted additive integration showed the best ability to distinguish different intensity of abiotic stress. In general the CSI values were lower under higher intensity of abiotic stress condition, and showed that abiotic stress effect was inclining towards a detrimental effect on crop status for wheat production under semi-arid condition. There was a clear indication that crop quality for wheat production was worsened by moisture and nitrogen stress under normal and high temperature stress conditions. The indexing method used in this study offers a useful, time and cost efficient approach to quantify abiotic stress effects on crop status for wheat production in semi-arid condition.

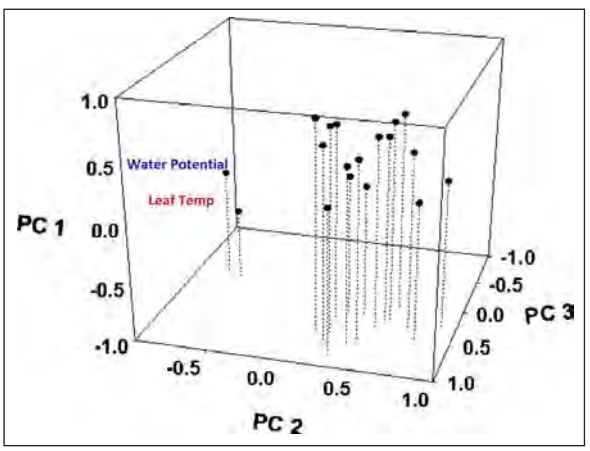

Fig. 9: Principal component (PC) scores of all Stress indicators in the first three PC's used for Crop Status Index (CSI) development.

Experiments were conducted with an aim to study the effect of pre-sowing sunflower (Var. Sunbread-275) seed exposure to standardized static magnetic field on seed water uptake and spin-spin relaxation time during imbibtion, field emergence index, root characteristics, plant growth parameters, radiation use efficiency, water use efficiency and yield parameters. Initial experiments were conducted to find out the optimum dose of magnetic field and duration of exposure for maximum enhancement of seedling characteristics. An exposure of 200 mT for 2h was found best among the other treatments. Seeds exposed to 200 mT static magnetic field for 2h were sown in the field along with untreated control under three irrigations. The seeds exposed to 200mT for 2h showed increased field emergence index, leaf area index, shoot length, number of leaf, chlorophyll content, biomass, 1000 seed weight and seed yield. Magnetic treatment showed significant higher value of total root length, root surface area and root volume as compared to corresponding value in untreated control in all

irrigations under different growth stage. Total root length at different fractional group showed 78 to 86% of total root length, 36 to 55 % of total surface area was found upto 1 mm fractional range, while 86 to 90 % of the total root volume was found above 2 mm range in all treatment. The water uptake and spin–spin relaxation was greater in lag phase II and rapid hydration phase III in magnetically treated seeds than untreated seeds. The magnetic treatment showed higher value of WUE, RUE and %IPAR as compared to untreated control. Hence it may be concluded that exposure of dry seeds to static magnetic field of 200 mT for 2 h improved seed water uptake and spin-spin relaxation time. This resulted in higher shoot and root growth. Improved root system and biomass led to increased seed yield, water use efficiency and radiation use efficiency.

Remote Sensing and GIS

A field experiment was conducted on wheat crop to measure and analyze its bi-directional reflectance anisotropy in relation to sun-target-sensor geometry during different growth stages under two extreme nitrogen treatments (N: 0 & 120 kg/ha). The set of bi-directional reflectance measurements along with correspondingly measured leaf biochemical and canopy biophysical properties were used to validate the canopy radiative transfer model PROSAIL5B in the optical region. The study reconfirms the strong and consistent anisotropic patterns of wheat reflectance in VIS and NIR regions in response to change in sun-target-sensor geometry and the magnitude was highest in the principal plane. The anisotropic pattern extended equally in SWIR wavelength region also. The maximum reflectance was seen in the backward scattering direction for all the treatments and all the dates. The hotspot became broader in wheat with the growth of crop due to increase in LAI, leaf size and planophlic orientation. The PROSAIL model simulated spectra was in good agreement with the observed spectra in the wavelength range of 400-2500 nm for all the view zenith and azimuth angle combinations used in the experiment. The model simulations also showed very good match in the principal plane, the region of highest anisotropy. The model performed best in the NIR region followed by SWIR and maximum relative error was observed for VIS region. Over the whole optical region, model simulations show an average error of 27 percent for wheat and this average error is higher (~33%) in nadir view position than in off-nadir view position.

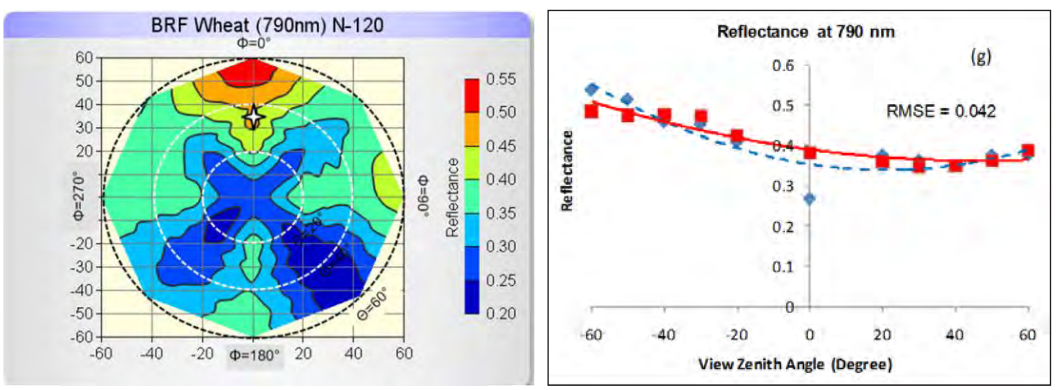

Fig. 10: Bi-directional reflectance factor of wheat and comparison of observed and PROSAIL simulated reflectance at 790 nm at different view zenith angles.

Retrieval of wheat LAI from broadband reflectance data corresponding to Indian Remote Sensing (IRS) LISS-3 (Linear Imaging Self Scanner) sensor by means of canopy radiative transfer model PROSAIL5B was attempted. The PROSAIL5B model was calibrated and validated with ground measurements in experimental fields and farmer's fields for two years. Three inversion techniques to invert PROSAIL5B model tried were: a look up table with best solution (LUT-I), a look up table with best 10% solutions (LUT-II) and an artificial neural network (ANN). All the three techniques could estimate the biophysical variables by capturing variability in

their observed values, though accuracy of estimation varied. Accuracy assessment by target diagram showed the superiority of LUT-II over other two approaches indicating that a set of best 10% solutions is a better strategy while ANN was worst performer showing highest bias. The LUT-II inversion approach of PROSAIL5B model was implemented for IRS LISS-3 image of 5-Feb-2012 for District Sheopur, Madhya Pradesh and retrieved the LAI of wheat crop growing in the district. The satellite image was preprocessed for geometric and radiometric corrections. Software codes in IDL were written for generation of LUT and for performing inversion of the model. The majority of wheat LAI varied between 1.5 and 3.5. The comparison with ground observations showed that the model inversion underestimated LAI with an RMSE of 0.56 though estimated LAI showed a very high R2 of 0.86 ($p<0.01$). The results of study imply that we can generate operational crop biophysical product of LAI from IRS LISS-3 for various applications.

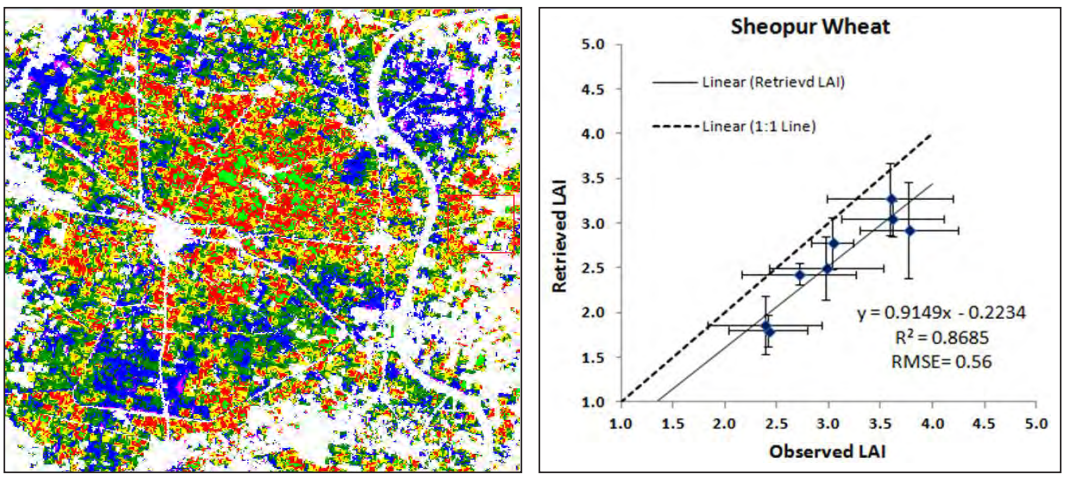

Fig. 11: Wheat LAI map of a part of Sheopur district (Madhya Pradesh) on 5-Feb-0212 and its comparison with ground measurements.

The radiative transfer model PROSAIL was used for retrieval of leaf area index (LAI), chlorophyll (Cab) and equivalent water thickness (EWT, Cw) of wheat crop in Trans-Gangetic Plains through its inversion. The model was calibrated for major parameters such as LAI, Cab, Cw and biomass (Cm) and sensitivity analysis

Table 1: Evaluation of regression approaches and radiative transfer model approach for retrieving canopy parameters.

S.No.	Parameters	Method	Regression equation	R^2 value with measured parameter
1	LAI	Regression with NDVI	$Y=1.8356\ e^{1.5183x}$	0.6759
2	LAI	Regression with EVI	Y=9.4568x+0.3117	0.8098
3	LAI	RT model inversion	Merit function	0.9043
4	Cab	Regression with RI	Y = 4.4544x + 9.4567	0.8515
5	Cab	Regression with GI	Y=9.0607x+0.3124	0.8320
6	Cab	RT model inversion	Merit function	0.9169
7	Cw	Regression with NDWI	Y=0.2264x+0.0167	0.7260
8	Cw	RT model inversion	Merit function	0.8970

was performed. Inversion of PROSAIL model was carried out for LAI, Cab and Cw using look up table (LUT) approach. The merit function was computed and used to best fit the measured data with the simulated one. Results revealed that LAI, Cab and Cw, were very well retrieved with RMSE 0.3892, 4.307 and 0.0063, respectively when compared with measured values. The retrieved products were evaluated with its corresponding regressed products through different vegetation indices. Root mean square error (RMSE) between these regressed estimation and measured parameter values were 0.553, 5.204 and 0.01 for LAI, Cab and Cw, respectively.

A study evaluated the retrieval of leaf area index (LAI), leaf chlorophyll (Cab), canopy chlorophyll (CCC), and leaf wetness (Cw) of soybean from broadband reflectance data of MODIS, TM and LISS-3 sensors through canopy radiative transfer model PROSAIL. Three inversion approaches namely, look up table (LUT), genetic algorithm (GA) and neural network (ANN) were used and their performances were compared. The GA performed similar to the LUT (mean of 10% solution) while ANN was the worst performer. In all the approaches, the order of estimation accuracy was LAI > CCC > Cab > Cw. There was no difference in estimation accuracies between MODIS, TM and IRS LISS-3 reflectance. The look up table (LUT) inversion approach (mean of 10% solutions) of PROSAIL model was applied to MODIS surface reflectance image to retrieve LAI, Cab and Cw of soybean in some districts of Madhya Pradesh.

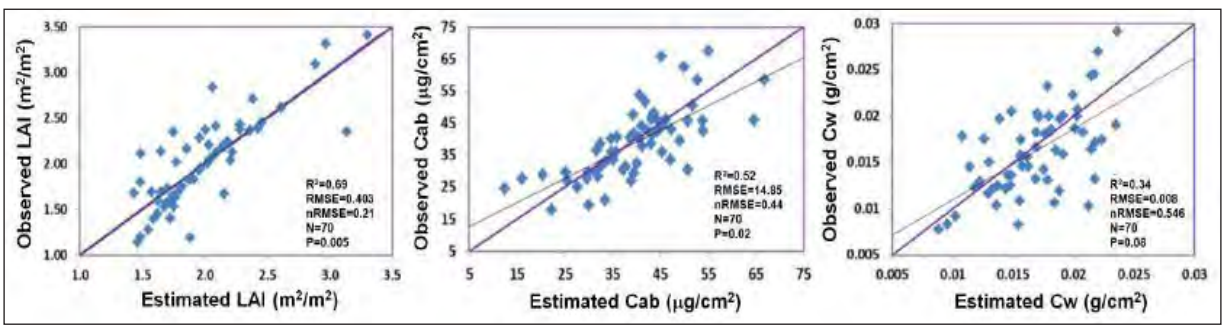

Fig. 12: Regional level validation of PROSAIL model retrieved soybean biophysical parameters with ground measurements.

Principles of infrared thermal imaging were applied to determine the Crop Stress Indices to characterize the changes in growth and yield of wheat crop (cultivar HD 2967) raised under four levels of moisture stress. Periodic observations on biophysical parameters and thermal imaging were made. Three crop Stress Indices namely CWSI, IG and CSI3 were estimated based on the canopy temperature measured through thermal image and compared with those measured with infrared thermometer (IRT). Results showed that the thermal image based stress indices were more consistent than IRT based indices and among the different stress indices tested in this study CWSI showed better correlation with crop canopy based stress parameters like NDVI and LAI. Thermal camera captures the crop canopies of many plants in a plot and thus characterizes the crop canopy more effectively than the spot measurements made using the infrared thermometers. In general under moisture stress condition, the stomatal conductance increases which results in an increase in crop canopy temperature and the stress indices calculated from the thermal image could characterize these changes effectively. Thus the thermal imaging can be used as an efficient technique to characterize the moisture stress in wheat crop.

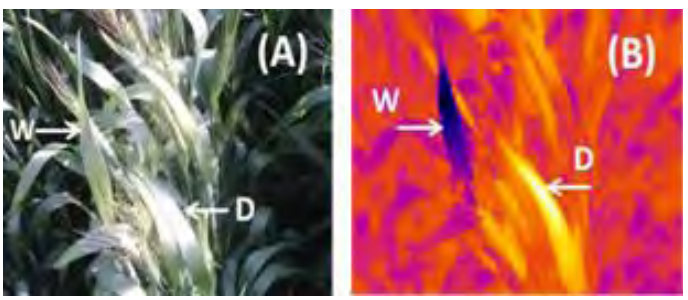

Fig. 13: (A) Digital true color image and (B) thermal image of the wheat crop with dry (D) and wet (W) references for calculation of thermal indices using thermal imaging camera.

Field experiment was conducted three varieties of mustard viz., Pusa Gold, Pusa Jaikisan and Pusa Bold sown on 19th October, 3rd November and 18th November. Pusa Gold had higher aphid infestation as compared to other two varieties. Spectral reflectances of healthy as well as aphid infested mustard were measured in laboratory and field. The aphid infested mustard showed very low reflectance throughout visible region while healthy mustard showed a 10% reflectance at 550nm. In the near infra-red region, the aphid infested mustard showed consistently low reflectance than healthy while this trend reversed in shortwave infrared region (1500-2500nm). At water absorption bands of 1390 and 1900 nm, aphid infested mustard showed very high reflectance than healthy crop. Spectral indices viz. NDVI, RVI, AI and SIPI showed significant correlation with aphid infestation. Hence these indices could be used for identifying aphid infestation in mustard. This study revealed there is scope of using hyperspectral remote sensing for identification of aphid infestation.

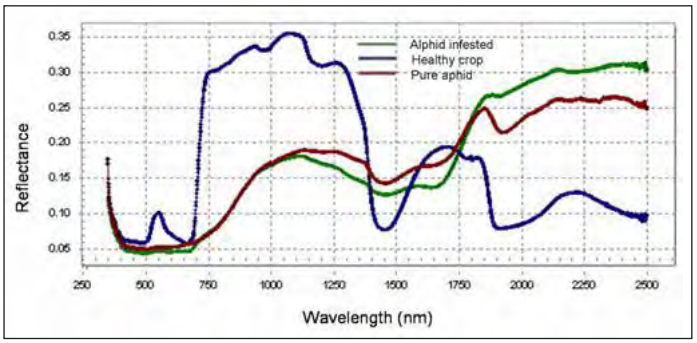

Fig. 14: Spectral reflectance of healthy and aphid infested mustard.

Hyperspectral indices were used to detect, predict and differentiate water and nitrogen stresses in wheat crop. Five models out of 35 for leaf nitrogen content (LNC) based on green normalized difference vegetative index (GNDVI), normalized difference chlorophyll index (NDCI), normalized difference705 (ND705), ratio index-1dB (RI-1dB) and Vogelman index a (VOGa) were found to be the most accurate ones. These are most sensitive indices to quantify nitrogen stress in wheat crop. Relative leaf water content (RLWC) was predicted most accurately by moisture stress index (MSI), normalized difference infrared index (NDII), normalized difference water index1640 (NDWI1640) and normalized multi-band drought index (NMDI). Using these indices, water stress in wheat crop in terms of RLWC can be assessed.

Digital image based LAI estimation in field condition was attempted. Images were acquired using a Canon Powershot G9 (12.1 megapixel, 4000× 3000 resolution, sensor size 25.4 × 43.18mm and focal length 35-210mm with 6x optical zoom) digital camera under natural sunlight at solar noon at 40, 70, 90 and 110 days after sowing of wheat crop from 10 different genotypes with 3 replications. Digital camera was mounted one meter above the canopy and the photos were taken looking vertically downward. The colour images were recorded in jpeg format and colour based different vegetation indices like normalized difference vegetation index (NDVI), excess greenness index (ExG), excess red vegetative index (ExR), ExG-ExR were computed to create tonal images and binary thresholding was done to compute gap fractions from different indices and evaluated with canopy measured LAI. Comparative analysis with measured LAI values revealed that ExG-ExR was best out of the four indices for predicting LAI.

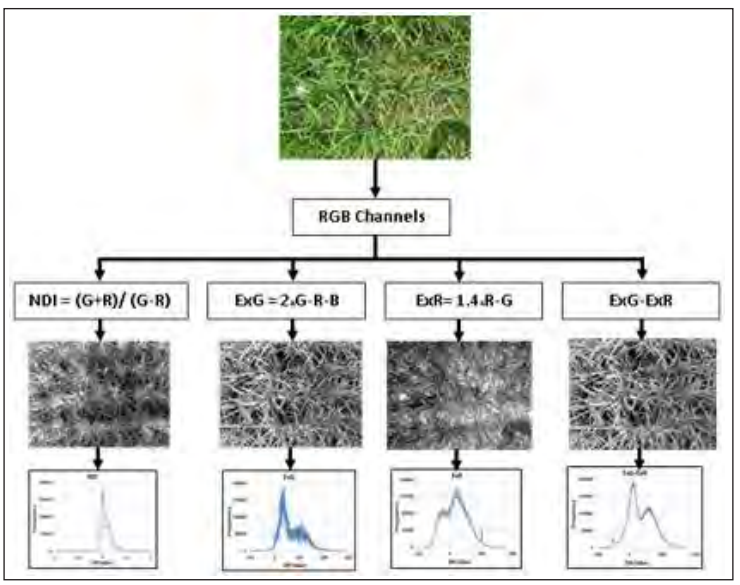

Fig. 15: Methodology of generating tonal images of different indices and histogram for thresholding for generating binary image of vegetation and soil.

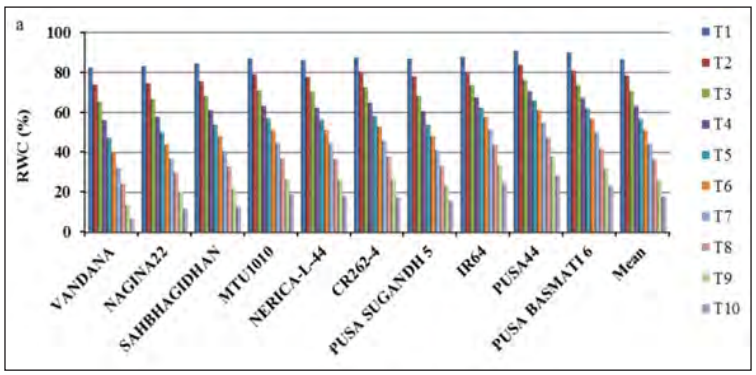

Fig. 16: Spectral index for capturing differential response of rice genotypes to water deficit stress in comparison to conventional approach (using Relative water content, RWC).

Developed a methodology for quantitative estimation of soil fertility from proximal diffuse reflectance data at laboratory and naturally illuminated field conditions. Same methodology was evaluated at regional scale using hyperspectral remote sensing satellite data, i.e. Hyperion sensor of EO-1 for fertility assessment.

Developed non-invasive techniques for estimation of relative water content of rice and wheat plant through hyper spectral remote sensing data and chlorophyll content by digital imaging for screening of cultivars using high throughput phenotyping.

The regional estimation of evapotranspiration and subsequently surface soil moisture from MODIS satellite images during pre-sowing period of rabi season in 2009 and 2010 were carried out in North West part of Uttar Pradesh, India using SEBAL (Surface Energy Balance Algorithms for Land) algorithm and few ground measurements. MODIS and Landsat satellites imageries of the study area were used to determine components of the surface energy balance equation, viz., net radiation, soil heat flux, sensible heat flux and latent heat flux and evaporative fraction. The results indicated that the daily evapotranspiration was in the range of 0.2 to 6.8 mm/day (CV = 19%) in 2009 and 1.3 to 6.9 mm/day (CV = 39%) in 2010 on the date of observation (Nov.6, 2009 & Nov.8, 2010). In 2009, high evaporation between 5.0 to 6.5 mm.day-1, was observed in about 98% of the study area. During 2010, only 78% area exhibited high evaporation rate between 5.0 to 6.5 mm.day-1, whereas 1% area was under low evaporative rate (<2 mm.day-1). The regional surface soil moisture predicted was found to be between 1.5 to 40% (CV= 40%) in 2009 and between 8.5 to 33.4 % (CV=33%) in 2010. The surface soil moisture maps show that in Nov 2009, 60% area was under 20-30% moisture content range and 17% area was under 10-20% moisture content range. In 2010, only 13% area was found to be under 20-30% moisture range and majority of area 77% was in 10-20% range.

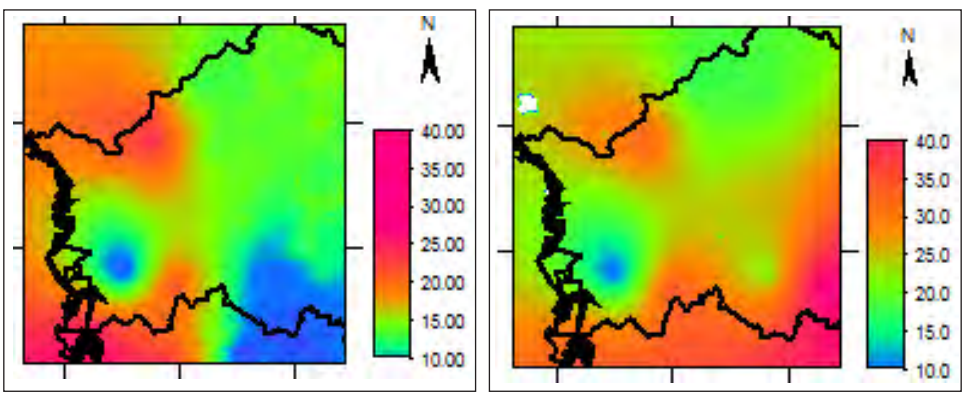

Fig. 17: Surface soil moisture content (% cc/cc) of study region in 2009 and 2010.

A study was conducted for estimation of solar insolation from geostationary satellite data (Kalpana-1 VHRR sensor for the period Nov, 2008 to April, 2009,) for entire Indian territory. Incoming solar radiation on ground under clear and cloudy sky conditions were estimated separately through spectrally integrated radiative transfer models. The methodology proposed the incorporation of sky view factor, atmospheric turbidity and elevation information for each pixel to get better estimates of insolation. The estimated solar radiation was compared

with measured values over a day as well as over fews days. The method worked very well under clear sky conditions for estimation hourly and daily totals.

To separate the effect of weather and technology on crop growth over time in agricultural areas, a modification in "Vegetation Condition Index" (VCI) was proposed and called "Trend Adjusted VCI" (VCITadj). A VCI value of 0.35 and below indicates agricultural drought. The modified index was found better in identification of agricultural drought of 2002 in Rajasthan, as it not only separated the effect of weather and ecology on crops but also accounted for changes in crop production technology over time. The VCITadj was computed for early, mid, late and whole crop seasons for 1981-2006 period by deriving pixel wise crop phenology metrics from filtered NDVI growth profile for Rajasthan. It was found that rainfall anomaly index SPI and VCITadj are linearly related in all the four seasons, the strength of correlation improved with the progress of crop season and these relationships were stronger than between primary variables of rainfall and NDVI.

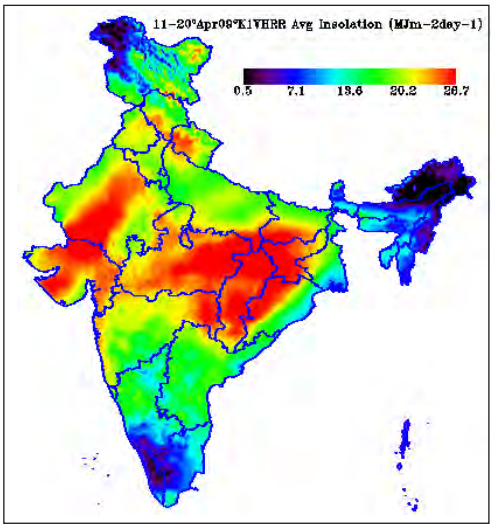

Fig. 18: Solar radiation map in April 2009 derived from Kalpana-1 VHRR.

A new geospatial methodology to assess vulnerability to drought in *kharif* season and its intra-seasonal variability was demonstrated for the state of Rajasthan. Frequency and intensity of Standardized precipitation index (SPI) computed using monthly rainfall data (1951-2006) was used as indicator of hazard probability, frequency and intensity of satellite derived vegetation condition index (VCI) during 1982-2006 and soil water holding capacity were used as indicators of crop sensitivity, whereas percent area under irrigation was used as indicator of adaptive capacity. All the indicators were combined together by computing weights using multi-criteria-analysis to arrive at final composite vulnerability index. The vulnerability was mapped separately for early, mid and late seasons and for whole *kharif* season. Results show that about 11%, 53%, 31%, and 5% of state's net sown area has extreme, high, moderate, and low vulnerability for whole *kharif* season, respectively. The top five districts vulnerable to agricultural drought found were Jaisalmer, Churu, Barmer, Jalore, and Sirohi, whereas Alwar, Sriganganagar, Bundi, Dausa, and Kota had low vulnerability. The district level drought vulnerability statistics showed significant correlation with Human-Development Index (HDI) and foodgrain productivity thus validating the methodology. The proposed methodology can be extended to other States of India for the said purpose.

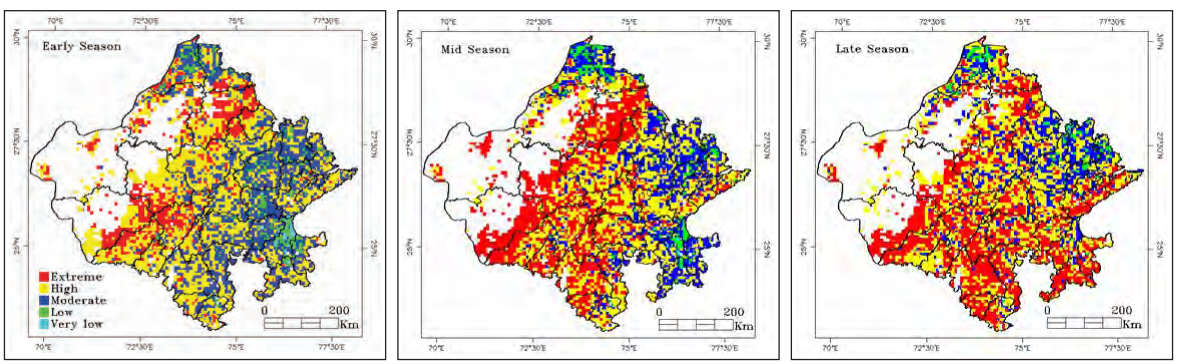

Fig. 19: Agricultural drought vulnerability rating map of Rajasthan during early, mid and late kharif season.

Soil Physics and Soil-Water Conservation

In a field study, the variation of soil penetration resistance (PR) with soil water content (SWC (w/w)) and bulk density (BD) changes under bed and conventionally planted wheat was studied on a sandy loam soil. The

differences in root characteristics of both systems were also compared. The results showed that, in general for all SWC, with increase in BD from 1.2 to 1.6 Mg m^{-3}, PR increased but the increase was more sharp in conventional than in bed planting system. It was further seen that, at all BDs, the critical soil strength of 2 MPa arrived at higher SWC in conventional as compared to bed planting. It thus indicated that root growth continued normally at relative lower SWC in bed planting as compared to conventional planting. Multiple regression analysis of soil penetration resistance as a function of BD, SWC and soil depth showed that SWC alone accounted for 59% variation and along with BD accounted for 93–96% variation in PR. Addition of depth did not improve the correlation. Sensitivity analysis of multiple regression equation predicting penetration as a function of SWC and BD indicated that PR was more sensitive to SWC changes than BD changes. Lower sensitivity indices for both SWC and BD in bed planting than in conventional planting indicated relatively lesser change in PR per unit change in SWC or BD in bed system. Computer aided root length scanning system 'RHIZO' was used to study rooting characteristics. Analysis showed that root characteristics including root length density, average root diameter, surface area density and volume density of roots of wheat up to 30 cm were significantly higher in bed over conventional system. In systems, nearly 70% of root length density and 76–82% of root surface area was confined in upper 30 cm. It was further seen that more than 70% of total root length was in root diameter range below 0.3 mm.

In a study on sandy loam soil of IARI farm, soil hydrothermal environment under bed planted wheat was compared with conventionally flat planted wheat for evaluating the suitability of bed-planting system for the semiarid climate of this region. The soil temperature and water contents of the 0–20 cm soil profile under bed and conventional planting were monitored during drying cycle after 2nd and subsequent irrigations. Results revealed that mostly soil water content under bed planting was lower by 0.5–1% (w/w) than under conventional system mainly due to lower application of irrigation water and because of more porous soil environment. But it was also noticed that by applying irrigation at 22 days interval, soil water under bed remained within the available soil water range. It was observed that under both methods of planting magnitude of the maximum temperature decreased with an increase in soil water content, but for both high and low soil water contents (SWC), the temperatures were higher by 0.5–3 °C at the surface and subsurface (up to 20 cm) under bed than under conventional flat planting. Soil temperature data collected at 2 h interval during the day to monitor the maximum soil temperature showed that magnitudes of the peak at all depths were higher in bed planting and their arrival times were also earlier than in conventional planting. Again, at low SWC, peak soil temperature at the surface was near or higher than aerial temperature and at higher SWC, it was lower than aerial temperature. For similar aerial temperatures (30 °C), with an increase in SWC, both volumetric heat capacity and thermal diffusivity increased and their magnitude on bed was lower than that on conventional planting.

In order to demonstrate a proper procedure for assessing the soil physical health of a farm, an experiment was conducted in a rice–wheat field. Spatial variability analysis of soil physical properties measured on a rectangular grid (30 m×45 m) was carried out by using geostatistical analyst extension of Arc GIS software. Indicators for soil physical health assessment included bulk density (BD), field saturated hydraulic conductivity (Kfs), available water retention capacity (AWRC), organic carbon content (OC) and non capillary porosity (NCP). Rating values of soil physical parameters were different for wheat and rice as the optimum physical environment for both systems were different. Physical rating index (PI) at each sampling point was determined by multiplying the rating values for all five parameters. Results revealed that for BD, Kfs, OC and soil physical health index (PI), major and minor ranges of semivariogram varied between 300–380 m and 55– 90 m, respectively. Whereas for NCP and AWRC, they were relatively short (major range between 114–140 m and minor around 60 m). Results revealed that BD and PI for both surface and subsurface layers showed strong spatial dependence whereas the rest of the parameters showed moderate spatial dependence. Rating maps of mentioned parameters for wheat and rice cultivations were prepared as series of colored contours by using kriging or other appropriate interpolation techniques and suitable semivariogram models. Overall soil physical health of the farm was medium to good for paddy cultivation but was not suitable for succeeding wheat crop mainly because of increased BD and reduced Kfs, NCP and AWRC of the farm during wheat growth. Correlations between PI and grain yield of both wheat

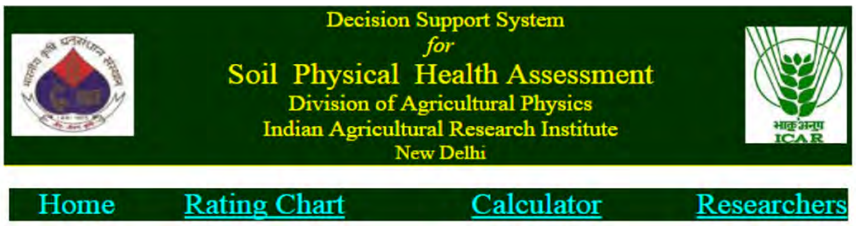

Fig. 20: Web based decision support system (DSS) for soil physical health assessment.

and rice were fairly good (r2=0.67). The results thus supported earlier findings that good soil physical health is essential for optimum sustained crop production.

Developed guideline for evaluating the suitability of any soil conditioner in improving the soil water availability. According to this guideline, if a soil mixed with conditioner has more readily available soil water content (water held between 0.1 - 1bar soil water suction (SWS)) and after wetting takes longer time (more than15-20 days) to reach 1bar SWS, it is effective in improving plant water availability. Under low evaporation, in saturated red sandy loam, alluvial sandy loam, sand and black clay soils with 0.7% hydrogel application, soil water reached at 1 bar in 7, 14, 22 and 4 days, respectively, indicating hydrogel's maximum effectiveness in sandy soil and least in black soil

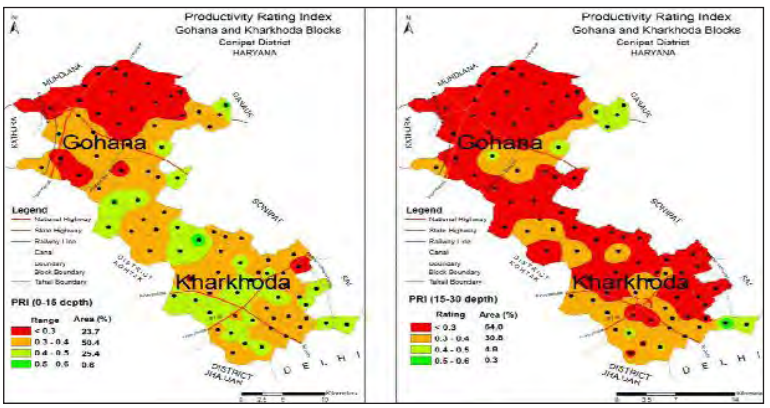

Fig. 21: Prediction map of physical rating index of surface and subsurface soil in Gohana and Kharkhoda blocks of Sonipat (Haryana).

Soil health of Kharkhoda and Gohana blocks of Sonipat district was assessed geospatially. Important parameters namely pH, electrical conductivity (EC), texture, bulk density (BD), saturated hydraulic conductivity (HC), soil organic carbon (OC), available water retention capacity (AWRC) and non capillary pores (NCP) were measured by collecting undisturbed soil samples in nearly 66 villages. Soil physical rating index (PI) method was used to compute PI which was an indicator of soil physical health of that region. Results revealed that in Gohana and Kharkhoda blocks, nearly 90% area had pH 4 dS m^{-1}, which indicated that soils were saline. Prediction maps of soil BD showed that 75% of the total area in 15-30 cm soil layer had BD above >1.6 mg m-3, which indicated the presence of hard pan in subsurface. HC data of subsurface layer also showed that 60% of the area had values10% which indicated adequate water retention capacity of these soils. However 85% of subsurface had poor soil aeration capacity as indicated NCP range < 10 %. Prediction map of PI for subsurface layer showed that majority of area had PI.

The impact of integrated nutrient management practices on the physical properties and structural stability of soil aggregates, and the associated C contents after 18 years of rice–wheat rotation on a sandy loam soil at Modipuram was studied. Treatments included fertilizer nutrients (NPK), NPK with Zn and/or S; and partial (25%) substitution of N with farmyard manure (FYM), sulphitation press mud (SPM), green gram residue (GR) or rice/wheat residue (CR) in various combinations. Soil aggregate properties and its stability, aggregate associated and particulate fractions of C at 0–7.5, 7.5–15 and 15–30 cm depths were studied to document C

sequestration potential of different nutrient management options. The aggregate strength and density were lower with organic substitution ($p < 0.05$) while water retention by aggregates at field capacity was 2–4% higher with organic inputs. Macroaggregates (>0.25 mm) constituted 58–92% of water stable aggregates and varied significantly among treatments and soil depths. Organic material incorporation improved soil aggregation and structural stability and resulted in higher C content in macroaggregates. The strong linear positive response to C additions indicated C sequestration potential in soils, with preferential location in macroaggregates. However, the kind and source of organic inputs strongly influenced both the soil aggregation and C accumulation in aggregates. A combination of GR in rice and FYM in wheat significantly improved C content in macroaggregates, and residue incorporation was beneficial compared to 100% N application through inorganic fertilizer or GR to rice. Coarse particulate organic matter (cPOM, >0.25 mm) accounted most of the increase in C content within macroaggregates and was substantially higher with CR incorporation. A relatively higher C content in microaggregates-within-macroaggregates in organic-amended soil implies potential in bringing higher C stabilization in intensive rice–wheat system through combination of inorganic and organic fertilizers and crop residues.

A study was conducted in farmers' fields of Rai block of Sonepat district, Haryana, India to study the long term impact of two widely adopted resource conservation technologies (RCT's) namely bed planting and zero tillage on structural properties of soils of recent alluvial plains of the river Yamuna. Aggregate mean weight diameter by dry sieving and wet sieving (DS-MWD and WS-MWD) under different RCT's were studied to compare structural condition of the soils under continuous use of these technologies. Other important structural indices such as dispersion ratio (a measure of ease of dispersion; DR), colloid moisture equivalent ratio (a measure of ease of percolation; CMER), erosion ratio (ER), stability index (SI), soil organic carbon (SOC), clay ratio (CR) were also studied to monitor the susceptibility of soil to erosion in the study area. Results revealed that in the surveyed villages under conventional tillage (CT), the mean (of 6 samples) magnitude of DR and ER were 0.58 and 0.82, respectively.

The effects of chiselling on soil water dynamics, root water uptake and water balance components were evaluated using Hydrus-2D model. Hydrus-2D model was applied to evaluate the effect of chiseling in soil water dynamics vis-à-vis crop water uptake. The chisel treatments were RS; Residual (Chiseling on June, 2011), RC; Repeated (Chiseling on June 2011 and again on June 2013), FC; Fresh (Chiseling once in June 2013) and NC; No chiseling (Control). Simulation results showed larger daily actual and cumulative root water uptake in RS and RC compared to the NC treatment and could be attributed to marginally higher LAI and fIPAR in chiseled plots along with greater depth and spread of roots. Model simulated soil water content matched with observed data with good agreement [R^2>0.60, p <0.01; RMSE = 0.04-0.08% and nRMSE = 16.2-28.4%] in the 1st year of rotation. The model was further validated by comparing the simulated and observed values of crop transpiration with R^2=0.88 and 0.83 in soybean wheat crop during 2013-14 (p<0.01). Simulated field water balance components indicated 5-7 cm higher soil water storage in 0-75 soil profile in RC, which had 3-4 cm higher seasonal transpiration by the crops. Chiseled plots (RC and RS) had significantly higher yield and water use in 1st year soybean-wheat rotation, and yield difference was marginal in 2nd year rotation. In view of cost of chiseling and the marginal benefits obtained till 2nd year of crop rotation, chiseling once in 2-3 years may be recommended for the sandy loam to sandy clay loam soils under the semi-arid climate of Delhi.

The root water uptake (RWU) patterns of cotton grown under conservation agriculture (CA) and soil water balance in cotton under a cotton-wheat cropping system were analyzed using the Hydrus-2D model. The treatments were: conventional tillage (CT), zero tillage (ZT), permanent narrow beds (PNB), permanent broad beds (PBB), ZT with residue (ZT+R), PNB with residue (PNB+R) and PBB with residue (PBB+R). Results in the third year of the cotton crop indicated that surface (0-15 cm layer) field saturated hydraulic conductivity in both PNB and PBB plots were similar and were significantly higher than ZT. Computed potential transpiration rates (Tp) under CT were lower than in other treatments, due to less radiation interception and lower leaf area index (LAI). Both PNB and PBB plots had higher Tp and crop yields than CT plots, which were further

improved by residue retention. Predicted soil water content (SWC) patterns during the simulation periods of third and fourth years showed good correlations (r2 = 0.88, the root mean square error (RMSE) = 0.025, and the average relative error (AVE) = 7.5% for the third year and r2 = 0.81, RMSE = 0.021, and AVE = 9% for the fourth year) with the actual field measured SWCs. Cumulative RWU (cm) were in the order: ZT (14.3) < CT (15.7) < PNB (16.3) < ZT+R (17.4) < PBB (18.8) < PNB+R (19.8) < PBB+R (22.6). Thus, PBB+R and PNB+R practices could be adopted for cotton cultivation, as these enhanced root growth and improved radiation interception and LAI. The Hydrus-2D model may be adopted for managing efficient water use, as it can simulate the temporal changes in SWC and actual transpiration rates of a crop/cropping system with reasonable accuracy.

A novel method for computing the lower least limiting water range (LLWR) limit was developed by using a linear function to relate penetration resistance (PR) to gravimetric water content and soil bulk density (BD), rather than volumetric water content. During the third year, the 15–30 cm soil layer beneath the puddled transplanted rice, under conventionally tilled wheat (PTR–CTW) plots had PR values that exceeded 2 MPa, but under direct seeded rice with brown manuring, zero tilled wheat (DSR + BM–ZTW) plots had PR values of less than 1.5MPa throughout the 0–60cm profile. That said DSR + BM–ZTW plots also had significantly higher gain (over the initial soil) in total soil organic C content in the 0–30cm soil layer. The 0–45cm soil layer under permanent broad-beds with residue (PBB + R) had significantly lower PR than permanent narrow-beds with residue (PNB + R) and other plots [PBB; PNB; zero tillage with residue (ZT + R); conventional tillage (CT) and ZT] in the cotton–wheat system. Retaining crop residue resulted in lower BD and PR values in the 0–15cm soil layer than removing them. The PBB + R plots had 12% higher LLWR than CT plots (LLWR= 10.1%) in the 15–30cmlayer. In the 0–15cmsoil layer, ZT + R, PBB + R and PNB + R had nearly 13, 24 and 11% higher mean (n = 24; 3 replications_8 sampling events) LLWR values than ZT, PBB and PNB plots, respectively, confirming that crop residue retention improved LLWR. Sub-surface layers under ZT had significantly lower LLWR values than in the CT plots. Results also reveal that there were no significant relationships between the mean (of two years) grain yields with LLWR for all crops, indicating that LLWR was a poor indicator of crop productivity. Overall, among the treatments, PBB + R and DSR + BM–ZTW were the best management practices for improved soil physical environment under cotton–wheat and rice–wheat systems, respectively, and therefore could be adopted.

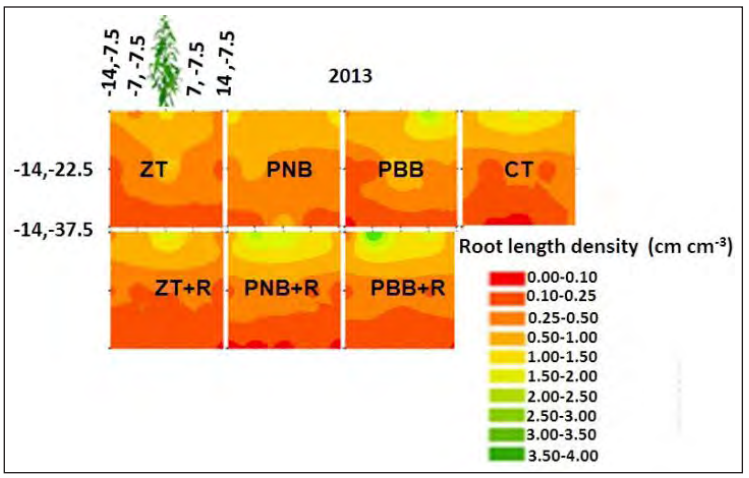

Fig. 22: Spatial distribution of the root length density of the cotton crop at the flowering stage under different soil conservation practices (ZT = Zero tillage; PNB = Permanent narrow bed; PBB = Permanent broad bed; CT = Conventional tillage; ZT+R = Zero Tillage + residue retention; PNB+R = Permanent narrow bed + residue retention; PBB+R = Permanent broad bed + residue retention).

Studies on effects of irrigation, wheat residue mulch and nitrogen interaction on soil structure, soil organic carbon pools, crop productivity and water and nitrogen use efficiency of maize showed that irrigation, crop residue mulching and nitrogen application significantly improve the soil structural properties (MWD, % WSA, WSSI) and soil physical health indices (S index and LLWR). There was improvement in the labile and non labile pools of carbon due to crop residue mulch.There was improvement in the crop yield, water use efficiency and nitrogen use efficiency due to crop residue mulch application. However, there was no significance difference between 75 and 150 kg N/ha with respect to grain yield of maize.So, maize may be grown under need based

Fig. 23: Experimental field photograph showing maize crop during flowering stage under irrigated condition with 150 kg N/ha and wheat residue mulch.

irrigation with 75 kg N/ha and wheat residue mulch (@ 10 t/ha) to achieve higher yield, water and nitrogen use efficiency, better soil structure and soil carbon pools.

Studies on water and nitrogen interaction in wheat showed that there was significant increase in the grain and biomass yield, ET, WUE, TIPAR and N uptake but decrease in nitrogen use efficiency of wheat with the increase in nitrogen dose. InfoCrop model could satisfactorily simulate grain yield, biomass yield, N uptake and Apparent N recovery of wheat but there is scope for improvement in the model for simulation of water balance and radiation balance. So it is recommended that wheat may be grown with 120 kg N/ha and five irrigations at critical growth stages (CRI + Tillering + Jointing + Flowering + Milking) to achieve higher grain yield and water use efficiency in the Inceptisols of upper-gangetic plain region.

Studies on Tillage, residue and nitrogen interaction in wheat showed that the evaporative flux was lower but deep percolation flux was higher under mulching than un-mulch treatment. There was no significant difference between tillage and crop residue mulch with respect to grain and biomass yield of wheat. During low rainfall years, application crop residue mulch could significantly improve the WUE of wheat. The grain yield, biomass yield and WUE increased with increase in the N levels. During high rainfall year (2014-15), the irrigated wheat responded up to 120 kg N/ha but during low rainfall year (2015-16) the crop responded upto180 kg N/ha. There was no significant effect of tillage and residue mulch on N uptake and grain protein content but nitrogen uptake and grain protein content increased significantly with increase in the nitrogen levels. There was no significant difference between tillage and residue management with respect to Partial factor productivity of Nitrogen (PFPN) and Nitrogen Utilization Efficiency (NUtE) by wheat crop but PFPN and NUtE decreased significantly with increase in the nitrogen levels. Crop simulation model DSSAT (ver 4.6) was calibrated and validated for wheat variety HD 2967 for the study region in Indo-Gangetic plain region. Evaluation of the model showed that it could satisfactorily simulate grain yield (R^2 = 0.759), biomass yield at harvest (R^2= 0.728) and seasonal ET (R^2= 0.904) and N uptake in wheat (R^2 = 0.883) under different tillage, residue and nitrogen management practices.

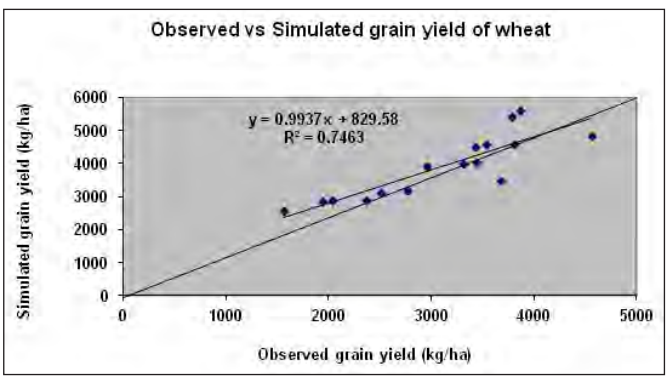

Fig. 24: Observed and InfoCrop model simulated grain yield of wheat.

Summary

- Developed the wheat crop yield loss factors for three dominant cultivars as a result in decrease in incident global radiation for use in models to simulate the affect of air pollutants on crop growth by modification of radiation regime.
- Recommended North-South direction of sowing of mustard in North-West India over East-West and Broadcasting to achieve higher radiation use efficiency, yield and oil content and lower aphid infestation.

- From micro-metrological perspective, recommended conservation tillage practice in Rice-Mustard cropping system.
- Using large aperture scinitillometery (LAS) diurnal and seasonal pattern of surface energy fluxes were studied for irrigated cropland and related to crop growth and soil moisture.
- Using Free Air Temperature Increment facility, quantified the effect of heat stress at different growth stages of wheat cultivar HD-2987.
- Evaluation of crop simulation models like, DSSAT-Wheat, InfoCrop, APSIM were undertaken which showed a satisfactory performance in their ability of simulate growth and biomass of wheat under normal conditions but their performance degraded in simulating growth elevated temperature indicating structural deficiency in models.
- Changes in micro-meteorology of spinach canopy were studied under different colored shade nets. Green colored shade net showed higher heat use efficiency, crop growth and yield.
- Use of hyper-spectral remote sensing indices can be used for detection and quantification of aphid infestation in mustard.
- Using AquaCrop model, three irrigations of 80 mm each at 21, 85 and 105 days after sowing (DAS) can maximize wheat yield in North Plain zones under normal rainfall scenario, while water productivity can be maximized by applying two irrigations of 60 mm each at 21 and 105 DAS with some yield penalty.
- Using IPCC climate change scenario in 2020-21 and 2050-51, study showed that the increase in crop water demand in terms of increase in evapotranspiration may be offset to some extent by the decrease in length of growing period.
- Open top chamber studies were conducted to study the effect of elevated CO_2 on Pigeon pea and Chickpea varieties. Higher CO2 resulted in increase in radiation use efficiency and total biomass in both Pigeon pea and Chickpea but yield increase in biomass did not translate into corresponding increase in seed yield due to lowering of harvest index in Pigeon pea. In Chickpea, the elevated CO_2 resulted in increase in harvest index and hence seed yield.
- The long term changes in crop phenology derived from remote sensing images showed that for 100mm decrease in seasonal rainfall during *kharif* in Rajasthan, decreases crop duration by about 7 days.
- Developed a new Crop Status Index to characterize the intensity of moisture, temperature and nitrogen stress, alone or in combination.
- The pre-sowing sunflower seeds exposure to 200 mT magnetic field for 2 hours resulted in higher initial root growth, more water uptake which led to increase in seed yield, water and radiation use efficiencies.
- Calibration and validation of canopy radiative transfer model PROSAIL was undertaken for wheat and soybean crops in field experiments. Over the whole optical region, model simulations show an average error of 27 percent for wheat and this average error was higher (~33%) in nadir view position than in off-nadir view position.
- Different techniques for inversion of canopy radiative transfer model were validated for retrieval crop biophysical parameters of LAI, leaf chlorophyll and leaf moisture at field level and at regional level using satellite images. Look-up table approach (with mean of 10% best solutions) was found to be best technique for inversion. LAI was retrieved with highest accuracy and leaf moisture with lowest accuracy.
- Established a protocol for quantification of moisture stress in wheat using thermal imaging remote sensing. Hyperspectral indices were also developed and validated to detect, predict and differentiate water and nitrogen stress in wheat crop.
- The experimental results identified the wavelengths and indices that can be used for detection of aphid infestation in mustard.

- Developed non-invasive techniques for rapid estimation of crop LAI and relative water content in rice through hyper-spectral remote sensing and digital imaging for phenotyping application in screening cultivars for drought tolerance.
- Methods were developed for estimation of global radiation from geostationary satellite Kalpana-1 for use in estimating regional scale crop primary productivity. Regional actual evapotranspiration was estimated for crop in a region and its dynamics monitoring by linking remote sensing based SEBAL model with meteorological parameters.
- Proposed improvement in remote sensing derived drought index Vegetation Condition Index (VCI) to separate the effect of technology and weather on crop growth over time and christened "VCI_{Tadj}". The study also showed that rainfall anomaly index SPI and VCI_{Tadj} are linearly related, the strength of correlation improved with the progress of crop season and these relationships were stronger than between primary variables of rainfall and NDVI.
- A methodology was established to map agricultural drought vulnerability which is essential to design area specific drought index, develop crop contingency plan and prioritize drought mitigation plans.
- Recommended the bed-planting system in wheat in terms of lower soil penetration resistance and higher soil water content. Analysis also showed that root characteristics including root length density, average root diameter, surface area density and volume density of roots of wheat up to 30 cm were significantly higher in bed over conventional system.
- Designed and validated a soil physical health index based on soil physical properties under rice-wheat system. The index was used to develop a web-based decision support system for rating soils in terms of its physical health in relation to crop production.
- Developed guideline for evaluating the suitability of any soil conditioner like Hydrogel in improving the soil water availability to crops.
- Studied the impact of integrated nutrient management practices on the physical properties and structural stability of soil aggregates, and the associated C contents after 18 years of rice–wheat rotation on a sandy loam soil. A relatively higher C content in microaggregates within macroaggregates in organic-amended soil implies potential in bringing higher C stabilization in intensive rice–wheat system through combination of inorganic and organic fertilizers and crop residues.
- The study on effects of chiseling on soil water dynamics and its modeling through HYDRUS-2D model revealed significantly higher yield and water use in soybean-wheat rotation.
- HYDRUS-2D model was adopted to study patterns of root water uptake and soil water balance in cotton grown under conservation tillage in cotton-wheat rotation.
- Effects of irrigation, wheat residue mulch and nitrogen interaction on soil structure, soil organic carbon pools, crop productivity and water and nitrogen use efficiency of maize showed that irrigation, crop residue mulching and nitrogen application significantly improve the soil structural properties and soil physical health indices.
- Study on effect of various combinations of conventional, zero-tillage and raised-bed systems on soil aggregation and associated organic C fractions in a rice-wheat rotation revealed that soil C was substantially higher in zero-till systems due to higher proportion of macro-aggregates.
- InfoCrop and DSSAT suit of models were employed to study the growth, yield, water and nitrogen uptake in wheat.

Future Perspective

- Integrating remote sensing and meteorological parameters for regular estimation of actual crop evapotranspiration a regional scale for monitoring crop water use and water stress.

- Developing methodologies to link real-time remote sensing inputs and weather forecasts with crop simulation models to forecast crop yield and aid in agro-advisory to farmers.
- Design and deploy hyperspectral and thermal imaging sensor based protocols for screening of crop varieties for various abiotic stresses as a part of plant phenotyping.
- Development of pest and disease forecasting models and their use using a combination of meteorological, crop and remote sensing parameters.
- Composite indices for drought monitoring, drought forecasting and crop loss for use in crop insurance.
- Land use land cover change dynamics in relation to anthropogenic factors and climate change for developing sustainable land use plans.
- Modeling root zone soil profile moisture content using microwave remote sensing and soil physical properties for quantification of moisture stress.
- Evaluation of soil physical properties and their dynamics under long term conservation tillage system for developing robust soil heath indices at regional scales.
- Regional level simulation of crop yield under different management practices for resource optimization.
- Structural changes in relation to hydro-mechanical behavior of soil across diversified agri-management practices.

Acknowledgement

I wish to thank all the Faculty Members of the Division of Agricultural Physics discipline for extending support and cooperation in preparing the research contributions of M.Sc. and Ph.D. students.

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Agronomy

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Introduction

Agronomy, as a recognized discipline of the Indian Agricultural Research Institute traces its origin to the Crop and Animal Breeding Section started in the Imperial Agricultural Research Institute at Pusa, Bihar in 1905. It assumed the status of a Division in 1923 for the purpose of awarding diploma, providing training and managing the institute farm. In 1936, it was recognized as a Research Division with the shifting of the Institute to New Delhi. A one-year training course for agricultural graduates was started in 1936, which continued till 1945. The Division had five sections, *viz.* soil fertility, vegetable and commercial crops, agronomic trials, farm and dairy, and grassland management in 1940. A two-year Associateship of IARI equivalent to M.Sc. degree was started in 1945, which continued up to 1958-59. Post-Graduate programmes, *viz.* M.Sc. and Ph.D., were started in 1958. Cattle breeding programme was transferred to dairy farm at Karnal in 1952. Several Divisions such as Agricultural Engineering, Agricultural Extension and Microbiology emerged out of the Division of Agronomy in the 1940s and 1950s. Similarly, several Institutes emerged out of the Division namely, National Dairy Research Institute, Karnal; Central Research Institute for Dryland Agriculture, Hyderabad; Indian Agricultural Statistics Research Institute, New Delhi; and the erstwhile Project Directorate for Cropping System Research, Modipuram. The Mandate of the Division is:

1. To provide leadership in crop and cropping system-based agronomic research by developing new concepts and approaches of sustainable crop production suited to farmers,
2. To develop appropriate crop production technologies to enhance the production and productivity of major cropping systems on a sustainable basis and
3. To provide post-graduate teaching and develop human resources.

Agronomy Division provides leadership in agronomic research by developing new concepts and approaches of agronomic technology suited to farmers on a sustainable basis.

Significant Student's Research Achievements

The research achievements presented under this head for Division of Agronomy are the outcome of the post graduate students research sincerely guided by the experienced faculty members and the Professors of this Division. Presently major thrust areas in agronomic are: Nutrient management, Crop diversification/cropping systems research, Conservation agriculture, Dryland agronomy, Weed management, Water management

Nutrient Management

Soil + foliar fertilization of Zn was more effective and economical to improve Zn concentration than soil application alone in both rice and wheat, indicating that it offers a vital solution for enhancing Zn concentration in their edible parts to curtail Zn malnutrition from the humans and animals. In general, zinc sulphate proved a better source than zinc oxide for zinc enrichment of prilled urea at the same level of zinc enrichment. The highest grain yield was obtained with 2.0% ZEU (ZS), which was at par to 2.0% ZEU (ZO), 1.5% ZEU (ZS)

and soil applied zinc sulphate @ 25 kg/ha, and significantly higher to all the other treatments including foliar spray of 0.2% zinc sulphate. All the zinc sources, except 0.5% ZEU (ZO), removed significantly higher amounts of Zn in grain over prilled urea alone. 1.5% zinc enriched urea (zinc sulphate), in general, proved to be the best treatment. The dual-purpose summer legumes such as cowpea or mungbean residue incorporation and application of 2.0% ZEU ($ZnSO_4.7H_2O$) or 2.0% ZEU (ZnO) in the aromatic hybrid rice had proved significantly superior in terms of effect on growth parameters, yield attributes, productivity and quality of aromatic hybrid rice. The dual-purpose summer legumes such as cowpea or mungbean residue incorporation and application of 2.0% ZEU ($ZnSO_4.7H_2O$) or 2.0% ZEU (ZnO) in the aromatic hybrid rice resulted in significantly superior with respects of concentrations and uptakes of macronutrients (N, P, K and S) and micronutrients (Fe, Mn, Zn and Cu) of the aromatic hybrid rice and also N and Zn utilization efficiencies of the aromatic hybrid rice. The inclusion of dual-purpose summer legumes for residue incorporation and application of 2.0% ZEU ($ZnSO_4.7H_2O$) or 2.0% ZEU (ZnO) in aromatic hybrid rice resulted into higher gross returns, net returns and B:C ratios of single crops i.e. aromatic hybrid rice, succeeding wheat and as well as the legume-rice-wheat cropping system under investigation. Incorporation either cowpea or mungbean residue and application of 2.0% ZEU ($ZnSO_4.7H_2O$) in aromatic hybrid rice under legumes-aromatic hybrid rice-wheat cropping system resulted in higher net returns (Rs 1, 13,678 and 1, 37,898) and B:C ratio (2.54 and 3.02).

Application of various fertilizer treatments maintained their superiority in promoting overall growth parameters, yield attributes, yields and quality parameters of sunflower. Between two sources of nitrogen i.e. urea and CAN, CAN was found to be Superior than urea in terms of growth parameters, nutrient concentrations and their uptake by sunflower-mungbean cropping system. Among the different levels of sulphur, the first dose i.e. sulphur @ 25 kg/ha was found to be more effective in terms of growth and yield and positive correlation was recorded up to 30-40 kg S/ha. Boron application @ 0.75 kg/ha and 1.5 kg/ha was very effective and the sunflower crop responded well up to the second dose. It also enhanced the total nutrient concentrations and their uptake by sunflower-mungbean cropping system and maintained a reasonably good available nutrient status of the soil. Quality parameters including protein content and fatty acid composition were influenced more by nitrogen @ 80 kg/ha irrespective of the sources than sulphur and boron. Between the two nitrogen sources, urea proved to be superior than CAN in terms of net returns and benefit:cost ratio. The average economic optimum dose for sulphur remained between 35-40 kg/ha. Nutrients applied to sunflower crop had a significant residual effect on the succeeding crop of mungbean in terms of growth parameters, yield attributes and yields. It also maintained the reasonably good available nutrient status of the soil after the harvest of the sunflower-mungbean cropping system in both the years of the experimentation. If we quantify the influence of variable doses of sulphur and boron on growth, yield and nutrient uptake by sunflower, it can be concluded that the overall effect and influence on all the parameters was higher due to boron application. The efficiency factors were more due to boron application and it also enhanced the conversion efficiency of plant to divert photosynthates to the final yield components and yield.

Groundnut intercropping had no significant effect on growth and yield attributes, yield, nutrient uptake and fibre quality of cotton as compared to sole cotton. Among N management practices of cotton, substitution of 25% RDN through FYM was good as 100% RDN and both recorded significantly higher growth and yield attributes, yield, nutrient uptake and fibre quality of cotton over 50% RDN substitution through FYM and unfertilized control. Intercropping of groundnut in cotton was advantageous as evident from intercropping indices. Intercropped groundnut performed best where cotton growth was poor i.e. under control where on N was applied to cotton. Performance of wheat was markedly superior after cotton + groundnut system. Wheat crop responded to direct application of 100 kg N ha^{-1} in terms of growth and yield attributes, yield and nutrient uptake. Groundnut intercropped in cotton with 50% RDN substitution through FYM and direct application of 100 kg N ha^{-1} to wheat recorded the highest cotton equivalent yield of system and net return. However, B:C ratio was higher with 25% RDN substitution through FYM and direct application of 100 kg N ha^{-1} to wheat. Substitution of 50% RDN through FYM and direct application of 150 kg N ha^{-1} to wheat resulted in higher organic carbon, available N content of soil in cotton-wheat system. Direct and cumulative effect of application

of green manure to rice + 150 per cent RDF recorded higher productivity of rice. Crop residue @ 5 t ha^{-1}, FYM @ 10 t ha^{-1} and control also had higher value at 150 per cent RDF. Residual and cumulative residual effect of green manure application + direct application of 150 per cent RDF or FYM Q 10 t ha^{-1} + 150 per cent RDF recorded higher productivity of wheat. Application of green manure to rice and residual effect to wheat at 150 per cent fertility levels or application of FYM @ 10 t ha^{-1} to rice and residual effect to wheat at 150 per cent recorded higher productivity of cropping system.

Application of 18 kg N + 20 kg P + 30 kg S/ha recorded highest productivity of mungbean. However, the productivity of succeeding mustard was significantly enhanced up to 18 kg N + 20 kg P/ha applied in mungbean. Incorporation of mungbean stover gave a significant boost to the growth and yield of mustard. Direct application of 80 kg N + 17.5 kg P + 30 kg S/ha also significantly increased the growth, seed and oil yield of mustard. Application of 80 kg P_2O_5 ha $^{-1}$ through SSP + PSB or pressmud + PSB recorded higher productivity of pigeonpea. However, APUE, PPEU and APR had higher values with 40 kg P_2O_5/ha^{-1}. Performance of succeeding wheat was markedly superior when pigeonpea received 80 kg P_2O_5 ha $^{-1}$ through SSP + PSB or pressmud + PSB. Direct application of P to wheat up to 80 kg P_2O_5 ha $^{-1}$ recorded higher productivity of wheat. Application of P to pigeonpea at 80 kg P_2O_5 ha $^{-1}$ through SSP+PSB or pressmud + PSB recorded higher productivity of the cropping system. The highest net return was realized with 80 kg P_2O_5 ha $^{-1}$ except when applied through pressmud + PSB to pigeonpea. Further, application of 80 kg P_2O_5 ha $^{-1}$ wheat fetched the highest net return in 2005-06, whereas in 2006-07, the highest net return was obtained with 40 kg P_2O_5 ha $^{-1}$ applied to wheat.

An integrated use of inorganic and organic sources of nutrients along with biofertilizers under FIRB system of planting proved to be better option for sustaining higher production, productivity and profitability of wheat. The conjoint use of recommended dose of fertilizers @ 120 kg N, 60 kg P_2O_5, 60 kg K_2O ha $^{-1}$ or 75% RDF @ 90 kg N, 45 kg K_2O ha $^{-1}$ along with FYM @ 5 t ha $^{-1,}$ biofertilizres and zinc @ 25 kg $ZnSo^{-4}$ registered a higher productivity and profitability of wheat. The FIRB system of planting and combined application of RDF or 75% RDF along with FYM, biofertilizers and zinc indicated a positive effect on soil environment which resulted into better rhizosphere. In the fixed plot experiment, integration of 25% RDN through de-oiled cake of jatropha or castor with 75% RDF produced sunflower seed yield comparable with RDF. The cumulative residual of previous season effect on the next season was recorded at 50% RDN through jatropha cake or castor cake with perceptible increase in seed yield over rest of the treatments. Jatropha and castor cake were found to inhibit the nitrification process compared to RDF. This nitrification inhibition was up to 15.6 percent and 12.19 percent in the 50% RDF + 50% RDN through jatropha cake, respectively. De-oiled cake of jatropha and castor alone or in integrated use with inorganic was found to produce weed population/biomass compared to RDF. Application of RDN through organic sources has improved the soil biological health. On the succeeding maize, residual and cumulative residual effect of treatment was found more pronounced with 50% RDF + 50% RDN through jatropha cake during both the cropping seasons. System productivity was observed maximum due to application of 50% RDF + 50% RDN through jatropha cake followed by castor cake and FYM in the same ration.

PGPR inoculants, i.e. *Azisprillum brasilense* and *Bacillus subtilis* had a significant and favourable effect on soil microbial properties (FDA hydrolysis, dehydrogenase activity and microbial biomass carbon) and root growth over the control in transplanted puddled rice. Application of 0.5 SP along with both PSB and VAM are recommended for improving productivity of soybean-wheat cropping system, although highest benefit cost ratio and better performance of 0.5 RP+PSB+VAM especially in second year of soybean were recorded. 0.5 SP+PSB+VAM provided huge margin of net return than other treatments and it also did not lag behind in benefit cost ratio which was very close to 0.5 RP+PSB+VAM. Integrated use of potassium improves the total bacteria, P solubilizers, actinomycetes, cellulose degrades, K solubilizers, dehydrogenase activity, FDA activity, B-Glucosidase activity, acid phosphatase activity and alkaline phosphatase activity over other treatments applied with fertilizers alone. In maize-wheat cropping system application of 60 kg K ha^{-1} supplemented 30 kg

K through MOP and 30 kg K through FYM in maize followed by 60 kg K ha^{-1} supplemented through MOP alone is found more economical.

Sesbania aculeata green manuring and 2.0% ZEU ($ZnSO_4.H_2O$) proved significantly superior in terms of productivity as well as quality of *Basmati* rice. *Sesbania aculeata* green manuring and application of 2.0% ZEU ($ZnSO_4.H_2O$) Zn fertilization treatments were significantly superior with respects of concentrations and uptakes of N, P, K and S and Fe, Mn, and Cu of the *Basmati* rice. Among the Zn fertilization treatments, foliar spray of 0.2% $ZnSO_4.H_2O$ recorded highest concentration of Zn in *Basmati* rice grain and straw. However, uptake of Zn in grain, straw as well as total (grain + straw) by *Basmati* rice was recorded higher with 2.0% ZEU ($ZnSO_4.H_2O$). Incorporation of *S. aculeata* and application of 2.0% ZEU ($ZnSO_4.H_2O$) in *Basmati* rice had proved significantly superior in terms of residual effect on succeeding wheat with respect to yield attributes, yield and uptake of N and Zn by wheat grain, straw as well as total (grain + straw). Incorporation of *S. aculeata* summer green manuring crop and application of 2.0% ZEU ($ZnSO_4.H_2O$) in *Basmati* rice resulted higher gross and net returns as well as B: C ratio of *Basmati* rice, under *Basmati* rice-wheat cropping sequence. *S. aculeata* summer green manuring crop and application of 2.0% ZEU ($ZnSO_4.H_2O$) in *Basmati* rice under rice-wheat cropping sequence resulted higher grain and straw yields as well as total productivity of *Basmati*-rice wheat cropping system. Residual effect of legume residue incorporated either of cowpea or mungbean resulted in higher value of yield attributes, yields, concentrations and uptake of Zn and N and net returns from the succeeding wheat crop compared to summer fallow. Residual effect of 2.0% ZEU ($ZnSO_4.7H_2O$) applied in the aromatic hybrid rice resulted in better residual effect on succeeding wheat crop and gave higher value of yield attributes, yields, concentration and uptake of Zn and N and net returns of the succeeding wheat during both the years of study over rest of the treatments applied to aromatic hybrid rice.

Paddy control recorded significantly lower weed density, weed dry weight, and nematodes population. However, mulching with wheat straw and *Sesbania aculeate* was essential for increasing water productivity and soil microbial activities. Paddy control was the best in respect to growth, yield and quality of aerobic rice followed by *Sesbania aculeate* mulch. *Sesbania aculeata* resulted in lower cost of cultivation and higher net returns and B: C ratio. Application of 50 kg ha^{-1} $FeSO_4$ + 2 foliar sprays of 2% $FeSO_4$ were beneficial for enhancing the productivity, quality and economics of aerobic rice. Growth parameters of popcorn like plant height, LAI, DMA were the highest at RDF but vermicompost equivalent to 120 kg N/ha application was also equally better to RDF. In potato, the highest growth and yield parameters was recorded on the residual fertility of FYM equivalent to 120 kg N/ha and direct application of FYM equivalent to 90 kg N/ha also found beneficial in potato.

Balanced fertilization maintained higher growth and dry matter accumulation of wheat at all the growth stages. Nitrogen proved to be the most limiting nutrient during both the years and its omission significantly reduced the growth, yield attributes, grain and straw yield of wheat. Phosphorus reduced the growth and yield of wheat significantly, which became more severe during the second year due to increasing soil P deficiency. This highlighted the fast depletion of P resources in the already deficient soil. K omission reduced the grain yield remarkably during second year, though it was statistically at par with optimum fertilization treatment. Sulphur and zinc omissions, however, resulted in non significant reductions in the grain yield possibly because of amply supplies form the indigenous sources. Soil available N, P and K was depleted in the respective omission plots. Balanced fertilization in both cotton and wheat fetched the maximum profits and seems indispensable for maintaining sustainability and profitability of cotton-wheat cropping system. STCR approach to both crops of the system is significantly superior in terms of productivity, profitability and sustainability over recommended dose of NPKZn, which suggests that the recommended dose for the maize-wheat cropping system is sub-optional and establishes STCR based nutrient application as more scientific management of nutrients in the cropping system. Omission of P, K and Zn in the cropping system highlighted the significance of P and k application to both the crops but the effect of K omission is more pronounced as compared to P omission in the system. While Zn may be conveniently omitted from the system due to high DTPA extractable Zn status in soil.

Application of 30 kg S ha^{-1} through gypsum to aerobic rice and 15 kg S ha^{-1} to wheat during initial year of aerobic rice-wheat cropping system and only 30 kg S ha^{-1} through either of sources to aerobic rice during second

year of RWCS should be given for better yields and economic returns. At different time intervals of incubation, single super phosphate (SSP) showed highest amount of released sulphur followed by gypsum, phosphogypsum and elemental sulphur. However, after 30 DAI, no significant differences were found among SSP, gypsum and phosphogypsum with respect to S availability at various intervals of incubation. Application of $CCPU_{1000,}$ in general, proved beneficial over other N sources in aerobic rice-wheat system. Highest gross return, net return and B:C ratio were observed in $CCPU_{1000}$ at 150 kg N ha^{-1} in wheat.

Application of FYM along with inorganic fertilizer and watering during heat stress are good management approaches to reduce adverse effect of heat stress. Under limited irrigation, HD 2987 with the application of two irrigation one at CRI and second at flowering can produce best yield and it is more economical. Cultivar WR 544 was best variety under adverse condition and can be grown under both heat stresses and water stress condition. In molecular study, less expression of HSP70 was recorded under integration of FYM and inorganic fertilizer and watering during heat stress. It indicates that wheat can mitigate heat stress when applied with 50 % RDF + 50% RDN through FYM and water during heat stress.

Leucaena and *Sesbania* application in aerobic rice saved around 40-50% irrigation water compare to TPR. Irrigation and total water productivity of aerobic rice was ~ 2 times higher than TPR. *Leucaena* incorporation and application of 120 kg N/ha + *Azotobacter* resulted in highest yield, net returns and system productivity. Incorporation of paddy straw suppressed the wheat growth initially and resulted in poor development of crop at early stages which resulted in low B: C ratio.

The application of neem coated urea (NCU) @ 150 kg N/ha as basal significantly enhanced growth parameters, yield attributes and that resulted in increased zero-till maize yield and significantly highest BC ratio (3.27) over conventional prilled urea application. However, under no residue condition prilled urea gave the highest net returns and BC ratio. The application of slow release N fertilizers especially NCU and SCU improved nitrogen and water use efficiencies in zero-till maize. Application of residue and nitrogen fertilization improved physico-chemical and biological properties of the soil.

Crop diversification/cropping systems research

DSBR-cabbage-onion followed by DSBR-cabbage-mungbean cropping systems gave higher system productivity, profitability and production efficiency in terms of rice equivalent yield over other cropping systems during both the year of study. DSBR-wheat-mungbean cropping system produced that highest energy output and net energy returns vis-à-vis rice equivalent yield among the test cropping systems. Among the different nutrient sources 50% RDF + 25% RDN-VC + biofertilizers was more effective in increasing the growth, yield attributes, yields and nutrient uptake in all the crops of direct seeded basmati rice-based cropping systems and for most of the parameters remained statistically on a par with 50% RDF + 25% RDN-LC + biofertilizers and 100% RDF. The cropping systems with summer mungbean (DSBR-wheat-mungbean and DSBR-cabbage-mungbean) with the application of integrated nutrient sources (50% RDF + 25% RDN-VC + boifertilizers and 50% RDF + 25% RDN-LC + biofertilizers) were found more suitable, as these systems led to marked improvement in soil physical bulk density (BD) and water stable aggregates (WSA), chemical (NO_3-N, NH_4^+ -N SOC, POC, LFC and available N, P and K status) and biological (MBC, dehydrogenase activity, alkaline phosphatase, ARA, and polysaccharide) soil health after two years of cropping cycle compared with DSBR-wheat-fallow

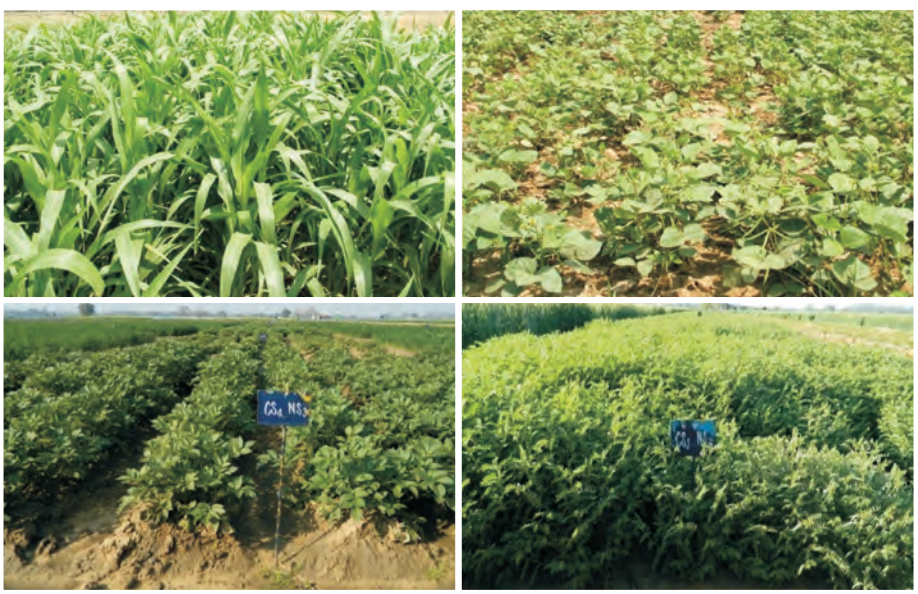
and DSBR-cabbage-onion system.

Among the short-duration summer forage crops: cowpea had significant effects on succeeding aromatic rice growth, yield attributes, yields and nutrients uptake followed by cowpea + maize. Among the different concentrations of gypsum-enriched urea (GEU); 10% GEU recorded significantly higher values with regard to growth, yield attributes, yields, nutrient concentrations and their uptake. Growing of short-duration summer forage crops had positive effect on chemical properties of soil, especially available N, organic carbon and micronutrients availability to the succeeding aromatic rice crop. The maximum net returns and benefit: cost ratio was recorded with maize + cowpea-aromatic rice, cropping sequence among the various cropping sequences studied during the experimentation, whereas 10% GEU resulted in significantly higher net returns and benefit: cost ratio over absolute control, 0 and 5% and remained statistically on par with 15% GEU.

Among the various summer forage crops, sole cowpea had significant effects on succeeding aromatic rice growth, yield attributes, yields and nutrients concentrations and their uptake followed by pearlmillet + cowpea mixture treatment. Among the different concentrations of phosphogypsum-enriched urea (PGEU); 7.5% PGEU recorded significantly higher values with regard to growth, yield attributes, yields, nutrients concentrations and their uptake and quality parameters of aromatic rice. The maximum net returns were recorded with pearlmillet + cowpea mixture – aromatic rice cropping sequence among the various cropping sequences studied during the experimentation, whereas 7.5% PGEU resulted in higher net returns and benefit: cost ration.

Rice-wheat-mungbean cropping system was more sustainable economically and on the basis of soil fertility to the traditional rice-wheat cropping system. Application of vermicompost + crop residue + biofertilizers (BGA + cellulolytic culture + PSB in rice, *Azotobactor* + cellulolytic culture + PSB in wheat, *Rhizobium* + PSB in mungbean) was most productive and FYM + crop residue + biofertilizers was economical for nutrient need of rice-based cropping systems. Both these combinations resulted in higher improvement in grain quality and physical, chemical and biological properties of soil. Pigeonpea-sunflower/wheat system, pigeonpea-sunflower system was found more productive and economical as compared to pigeonpea-wheat system, besides saving of one irrigation

The productivity and profitability of soybean-potato-mungbean system could further be enhanced with the application of best suitable nutrient combination to each crop of the cropping system i.e. 25% RDF + 50% RDN through FYM+biofertilizers to soybean, 100% RDF to potato and 50% RDF + 50% RDN through FYM to mungbean. This combination provided maximum net returns of about Rs. 1,41,400/ha with the saving of



42.5 kg N and 27.9 kg P_2O_5 with additional input of 7.6 kg K_2O/ha/year as compared to 100% RDF. Resource use efficiency especially water and profitability of transplanted Bt cotton is higher than direct sowing. In Bt Cotton-wheat/onion cropping systems 5.0 kg zinc/ha to Bt cotton is sufficient to meet the zinc requirement of both the crops in sequences. Bt cotton-onion system is more productive and profitable than Bt cotton-wheat system.

Conservation agriculture

Soybean and wheat either independently or in sequence can be successfully grown in continuous zero tillage without any yield reduction in comparison to continuous CT. Even in sequential tillage, where the tillage was skipped either in *kharif* to soybean or in *rabi* to wheat, yields and system productivity were similar with average of 7.22–7.43 t/ha and 7.19–7.42 t/ha of wheat equivalent yields in continuous tillage and sequential tillage treatments, respectively. Performance of the system was more visible, where ZT was supplemented with residue application. In flat planting as average net profit of Rs. 56,030/ha was obtained with ZT-F (S)–CT-F (W) and Rs. 54,400/ha was obtained with CT-F (S) –ZT-F (W) in soybean-wheat cropping system. Bed planting was not having edge over flat planting in soybean but in wheat there was saving of irrigation water by 28.6% and improvement of IWUE by 37% ZT brought an average saving of energy in the system by 7.2 and 8.5% with flat and bed planting than the corresponding conventional tillage planting methods.

MBR+DSR-ZTW/M+RR-MB conservation agriculture system was superior to TPR-CTW or other conservation agriculture based rice-wheat or rice-maize systems in productivity and profitability. Transplanted rice produced significantly higher yield (5.37 t ha^{-1}) initial year than DSR. However, in 2011 at par yield with transplanted rice (TPR) was recorded under MBR+DSR-ZTW+RR-MB (5.61 ha^{-1}). Tillage and different residue management practices could not bring out significant difference in wheat productivity in starting 2010-11, but significantly higher yield of wheat (5.33 t ha^{-1}) was recorded with the incorporation of mungbean residue in rice, followed by zero-till wheat with rice residues 9MBR+DSR-ZTW+RR-MB) in 2011-12. Comparatively higher yield of maize was obtained with conventional till maize followed by MBR+DSR-ZTM+RR-MB in both the years. Irrigation water use efficiency (IWUE) of direct seeded rice was 1.85-2.0 times higher than transplanted rice and was positively influenced by residue application. MBR+DSR-ZTW/M+RR-MB system produced highest rice-equivalent yield, gross and net returns and gross output energy while, no-residue showed higher energy-use efficiency in rice-wheat and rice-maize systems. There was an improvement in organic C and available nutrients status. Similarly, the positive beneficial effect of residue management practices on physic-chemical properties of soil was evident. Therefore, a conservation agriculture practice, MBR+DSR-ZTW/M+RR-MB may be recommended for higher productivity, profitability and resource-use efficiency in the rice-wheat as well as in rice-maize cropping system. However, rice-wheat system found superior to rice-maize system with conservation agriculture practice.

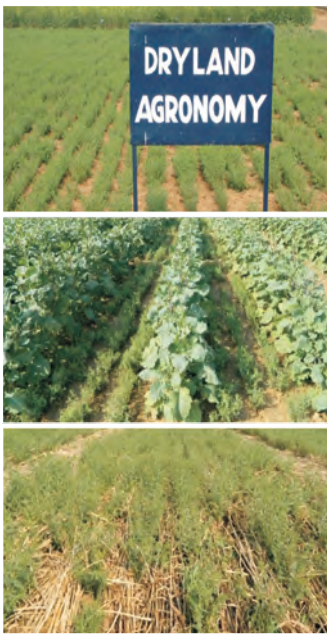
Kharif maize crop sown in conservation tillage based ZT and PB practices under diversified cropping systems resulted higher crop growth, yield attributes, yields, profitability, efficient nutrient use and quality. In long term experimentation the ZT and PB planting of maize under diversified cropping sequences improves the soil physical and chemical properties. Planting of *kharif* maize under ZT flat system resulted in better growth, yield attributes, yields, quality and improved input use efficiencies as well as physical, chemical and biological properties of soil in short term study at fixed site. Nutrient expert based SSNM application in maize showed higher growth, yield attributes & yields and quality of crop with precise nutrient and input management and improved soil health as compared to traditional nutrient management practices.

High density sowing fb alternate row harvesting for fodder at 35 DAS is a suitable practice to get higher forage yield and can be a sustainable alternative of forage shortage under CA. For getting higher yield with more profit under conservation agriculture especially during initial 2-3 years, pearlmillet should be fertilized with 75 N ha^{-1} (125% RDN) as basal. Combination of ZT + residue + 75% N + Green Seeker and sequential

application of pendimethalin fb sulfosulfuron may be recommended for higher productivity, profitability, resource-use efficiency and sustainability of the wheat-greengram sequence in the North-Western Indo-Gangetic plains of India.

Soil physical parameters, viz. mean weight diameter, soil moisture content and soil aggregation were favorable influenced under ZT than CT practices. Residue addition further improved the above parameters including bulk density of soil. Soil pH, EC and available NPK were similar in both zero and conventional tillage flat and bed planting. Residue applied treatments, viz. ZT-flat+R and ZT-bed+R resulted 7.79 and 7.69 % higher SOC than CT-flat and CT-bed, respectively. Microbial and enzymatic activities were significantly higher under beds, ZT and residue application than flats, CT and without residue application, respectively. Lower dose of N improved the microbial and enzymatic activities in soil. The productivity, monetary benefit, nutrient, water and energy-use efficiencies and soil properties were found comparable between ZTB and ZTF but higher than in CTB and CTF treatments. Similarly, residue retention throughout the cropping cycle was better than single season or no residue application. Further, ignoring energy consumption through residues and considering its good effects on soil properties, C-sequestration and abiotic stress moderations, zero tillage with bed or flat establishment techniques and crop residues retention throughout the year may be recommended for the maize-wheat-greengram cropping system in the irrigated North-western Indo-Gangetic Plains.

Dryland agronomy



Broad-bed furrow system gave higher productivity and net profit of pearlmillet, mustard and pearlmillet-mustard cropping sequence. The preceding application of 30 kg N + 20 kg P_2O_5 + 6 t FYM/ha to pearlmillet resulted in marked yield increase of pearlmillet, mustard and system productivity and thus higher net returns over rest of the treatments. The net return was higher with direct application of 60 kg N + 40 kg P_2O_5/ha to mustard in mustard and pearlmillet-mustard cropping system.

Adoption of pearlmillet + pigeonpea with transpiration suppressants and application of 50 kg N + 17.2 kg P/ha would be a better option to sustain the productivity to increase the moisture-use-efficiency and to maintain the soil fertility in limited moisture conditions. Under rainfed conditions of arid and semi-arid areas, pearl millet and mustard crops should be planted under zero tillage with 4 t/ha crop residue and fertilized with 30 kg S/ha for achieving higher productivity, profitability and soil quality.

Adoption of Ethiopian mustard + chickpea intercropping system with moisture conservation practices (FYM @ 5 t/ha + Organic mulch + Kaolin 6% spray) and recommended dose of fertilizer (60 kg P_2O_5/ha + 30 kg S/ha) would be a better option to sustain the productivity and profitability, to increase the moisture-use efficiency and to maintain the soil fertility in limited moisture conditions.

Under limited moisture conditions and in zinc deficient soils of arid and semi-arid areas, for achieving higher productivity and profitability of pearlmillet and chickpea with improved fertility, these crops should be planted either on flat bed with 5.0 t/ha crop residue or narrow bed and furrow with 2.5 t/ha crop residue and fertilizer with 5.0 kg Zn/ha.

Weed management

The major weed flora of the experimental field observed were *Cyperus rotundus, Echinoclo colonum* and *Dactyloctenium oegyptum* among monocots and *Digeria arvensis, Trianthema portulacastrum, Phyllanthusniruri,*

Convolvulus arvensis and *Commelina bengalensis* among dicots. The number of monocot weeds was not influenced by the biofertilizer treatments. The weed free condition and two hand weeding treatments recorded lower number of monocot weeds at all stages of crop growth. The highest number of weeds was observed in weedy control. The clomazone treatment recorded the lowest number of monocot weeds among all the herbicidal treatments.

Increasing *Chenopodium album*/weed infestation level right from 8 *Chenopodium* plants/m^2 resulted in significant reduction in the growth, yield attributing characters and yield of wheat. The lowest plant density of 8 *Chenopodium*/m^2 even reduced wheat yield by 4.6 and 5.7% across the years. This revealed greater competitive ability of *Chenopodium* album even at very low density. This led to the economic threshold of *Chenopodium album* in wheat to be 3.94 and 4.89 plants/m^2 in 2006-07 and 2007-08, respectively. Higher the *Chenopodium* /weed infestation level, greater was the competition from *Chenopodium* /weed. However, the competitive effect was more pronounced when highr density was accompanied by the higher dose of nitrogen. Winter weeds such as, *Chenopodium album*, cannabis sativa, *Chenopodium murale*, *Avena ludoviciana, Coronopus didymus, Cirsium. Arvense, Phalaris minor* and *Melilotus indica* as usual had spectral reflectance and NDVI values varying across them and across their growth stages. This could be of immense use towards identification of these weeds through remote sensing and further application of precision weed management.

Soil solarization fb glyphosate at 1.0 kg/ha proved highly effective towards control of *Cyperus* and other grass and broad-leaved weeds in soybean as well as in wheat. It also concurrently resulted in higher growth parameter (plant height, dry weight accumulation, branch per plant), yield attributes (pods/plant, seeds/plant, 1000-grain weight) and seed and stover yields of soybean and wheat. It, however, proved inferior to other treatments in benefit-cost ratio in soybean and soybean-wheat system mainly because of high cost of polyethylene film. Otherwise, it provided a significant residual effect on weeds in wheat and gave highest benefit-cost ratio. On the contrary, weed control although was not very effective, summer cowpea (fodder) was superior with respect to benefit-cost ratio in soybean and soybean-wheat cropping system mainly because of additional income obtained through cowpea fodder. Therefore, it may be recommended for possible adoption. However, where *Cyperus* management is the main objective in soybean-wheat system, soil solarization fb glyphosate at 1.0 kg/ha may be a superior alternative.

Sequential application of pretilachlor at 750 g/ha as pre-emergence followed by post-emergence application of cyhalofop-butyl at 60 g/ha at 30 DAS, sequential application of butachlor at 750 g/ha as pre-emergence followed by post-emergence application of cyhalofop-butyl at 60 g/ha at 30 DAS and alone application of cyhalofopbutyl at 80 g/ha at 20 DAS were found very effective against mixed weed flora in direct seeded rice and achieved comparable grain yield to weed free environment. Application of cyhalofop-butyl at 60-80 g/ha at 20 DAS was found better than its application at 10 DAS. Performance of pre-emergence application of pretilachlor at 1000 g/ha and butachlor 1000 g/ha was similar in controlling early emerging weeds and increasing grain yield. Herbicidal treatments did not affect the population of bacteria and actinomycetes at harvest. However significant reduction in the fungal population was recorded in herbicide treated plots as compared to untreated plots (weedy check and weed free).

Metribuzin when applied as post-emergence irrespective of dose, volume rate and time of application, did not affect significantly nodule number, but nodule dry weight. These treatments slightly inhibited acetylene reduction, dehydrogenase activity and microbial populations. However, herbicide residue was negligible in soil.

Seed yield was higher with metribuzin 0.5 kg/ha PRE, but metribuzin at 0.1 kg/ha with 200 l/ha applied at 20 DAS was comparable with it. In contrast, metribuzin post-emergence at 0.2 kg/ha with 400 I/ha of water at 30 DAS was the most inferior with yield and other parameters studied. Application of metribuzin as post-emergence increases oxidative stress in soybean plants, but gets mediated by an increase in superoxide and ascorbate peroxidase content. This brings forward the opportunities of using metribuzin as post-emergence in soybean with little or no crop phyto-toxicity.

Among herbicides, penoxsulam @ 25 g/ha applied at 10 days was found most effective in controlling weeds and in augmenting grain and straw yield, followed by its same dose applied at 15 days. The highest yield of rice was obtained in transplanted rice by adopting timely and effective weed control measures. Traditional method of rice culture (transplanting) was found better than direct-seeding. Weed-free condition resulted in the highest grain and straw yield. Residues of penoxsulam in soil at harvest of the crop were found below the detectable level. This indicates that succeeding crop and health will not harm by the use of penoxsulam. Herbicidal treatments brought significant reduction in the population of all three microbes at 10, 30 days and at harvest. Application of penoxsulam at 25 g/ha applied at 10 days or at 15 days crop growth resulted in the highest reduction in the population of all three microbes as compared to untreated soil.

Weed-free treatment recorded the higher increase in seed and stover yield of soybean, followed by two cono weedings at 20 and 40 DAS. Pre-emergence application of diclosulam @ 26g/ha and diclosulam @ 24 g/ha integrated with one cono weeding at 30 DAS showed phytotoxicity to soybean in early stages of crop growth. Among herbicides, pre-emergence application of diclosulam @ 20 g/ha integrated with one cono weeding at 30 DAS was found most effective in reducing weed growth and augmenting weed and stover yield which subsequently resulted in attaining the highest B:C ratio. Herbicide treatment had some adverse impact on the microbial population of bacteria, fungi and actinomycetes as compared to weedy-check, weed-free and two cono weedings treatments. Residues of diclosulam in soil at harvest of crop were found below detectable level.

Results of this study demonstrate the importance of weed control practices in reducing weed seed bank. Chemical control prevented the seed production of weeds and decreased the density of weed seed bank. The dynamics of seed bank is the qualitative and quantitative variation in density and combination of seed bank over time. Study of the variation of seed bank and weed flora and the relations between them is beneficial to predict the population level of weed and to select the best and most effective management system for reducing the density of weed seed bank.

All weed control measures significantly reduced the total weed population as well as dry matter over weedy check. Weed free treatment recorded the highest increase in seed and stover yield of soybean closely followed by sequential application of metribuzin @ 250 g/ha (PE) followed by propaquizafop @ 50 g/ha at 3 weeds after sowing owing to better weed control efficiency which subsequently resulted in attaining the highest B:C ratio. Application of propaquizafop @ 50 g/ha + chlorimuron-ethyl @ 9 g/ha at 3 WAS resulted in the highest reduction in the population of all the three microbes as compared to untreated soil.

All weed control schedules significantly reduced weed population and dry weight over weedy check. Cono weeding thrice at 15, 30 and 45 DAT was found the best treatment with the highest weed control index (88.59%) and lowest weed index (2.96%), which was comparable with one cono weeding at 15 DAT followed by application of bispyribac sodium @ 25 g/ha at 30 DAT with weed control index (86.84%) and weed index of (7.16%). Alone pre-emergence application of pretilachlor @ 1000 g/ha and pyrazosulfuron-ethyl application @ 25 g/ha at 10 DAT were found inferior in reducing the weed growth thus needs to be integrated to achieve higher weed control index. The highest growth and yield attributes of rice crop were registered with weed free treatment closely followed by cono weeding thrice at 15, 30 and 45 DAT, thought the highest B:C ratio was

obtained with later one that proves its economic viability. Economic sustainability of different weed control measures must be well evaluated before recommending them to farmers.

Tank-mix pre-emergence application of pendimethalin 0.5 kg/ha + imazethapyr 0.075 kg/ha having better weed control, higher soybean nutrient uptake, less negative impact on soil micro-flora and microbial activity, and less impact on physiological parameters like leaf chlorophyll and soluble protein, and less APX activity resulted in higher yield, net returns and B:C ratio of soybean comparable with those in weed-free check. All weed control measures significantly reduced the total weed population as well as dry matter over weedy check. Weed-free treatment recorded the highest increase in seed and stover yield of mungbean closely followed by sequential application of pendimethalin @ 1 kg/ha PE + one hand weeding at 30 DAS owing to better weed control efficiency which subsequently resulted in attaining the highest B:C ration. Herbicidal treatment resulted in reduction in the MBC and dehydrogenase activity.

Both flat bed and raised bed were comparable on the reduction in weed density and dry matter, and improving growth, yield attributes and yield of garlic, but the latter was superior to the former. A combination of raised bed and tank-mix pre-emergence application of pendimethalin 0.75 kg/ha $^{+}$ imazethanpyr 0.075 kg/ha, therefore, may be recommended for better weed control and higher garlic yield.

Water management

Mustard crop plants received three irrigation at seedlings, 50% flowering and pod development stages in both the seasons encouraged their initial growth (plant height, DM/plant, LAI) and produced more seed and stalk yields, compared to those received one and two irrigations. The crop irrigated with three irrigations significantly removed higher amount of N and S compared to one and two irrigations. The amount of consumptive water use was increased with increasing number of irrigations up to three irrigations, while maximum CWUE was obtained with two irrigations given at seedling and pod development stages. The maximum gross return, net return (Rs./ha) and B:C ratio was obtained when three irrigations (seedling + 50% flowering + pod development stage) were applied to the crop. Plants fertilized with 90 kg N/ha significantly improved the initial growth (plant height, DM/plant, LAI, number of primary branches), yield attributes (siliquae number per plant, siliqual length, seeds/siliqua and siliquae weight/plant), seed, stalk and oil yields, compared to those fertilized with 30 and 60 kg N/ha. The crop fertilized with 90 kg N/ha contained the maximum amount of N and S in plant and removed significantly higher amount of N and S compared to 30 and 60 kg N/ha. Consumptive water use increased slightly due to N application but water use efficiency increased with increase in successive doses of N up to 90 kg/ha. The crop fertilized with 90 kg/ha gave the highest gross return, net return (Rs./ha) and B:C ratio compared to 30 and 60 kg/ha.

Conventional furrow irrigation, irrigation at 60 mm CPE and an application of 120 kg N (urea) + 30 kg N/ha by FYM, separately resulted in significantly the highest growth parameters, yield attributes, yield and nutrient uptake by potato, and the highest consumptive use of water, rate of water use and moisture extraction from 0-15 cm soil depth. However, available nutrients in soil after potato harvest were significantly the highest, water use efficiency was the highest, nutrient uptake by maize and available nutrient in soil were significantly the higher in alternate furrow irrigation, irrigation at 100 mm CPE and an application of 120 kg N (urea) + 30 kg N/ha by FYM, separately. The combined effect of these treatments also resulted in the highest water-use efficiency.

Irrigation at 60 mm CPE through ridge furrow configuration would be a better option to sustain the productivity of maize, with water economization for *Kharif* and *Rabi* seasons. Alternative irrigation threshold of 0 kPa (complete saturation) could be 10 kPa without any reduction in grain yield of DSR. Likewise, split application of recommended dose of 150 kg N ha^{-1} as ¼ basal + ¼ at 2 week + ¼ at 5 week + ¼ at 9 week gave the highest yield, water use efficiency and economics in DSR. Application of irrigation water at tillering to flowering is very crucial to obtain higher yields in DSR.

Future Perspectives

The future agronomic research should focus on the following aspects:

- Research on the development of profitable and sustainable inter and sequential cropping system
- Development of most effective fertilizer materials for enhancing nutrient use efficiency in order to have better recovery efficiency, productivity and environmental stewardship.
- Development of precision micro-irrigation methods in the major field crops for realization of per drop more crop vision in the country, especially under rainfed conditions.
- Development of effective plant and soil moisture conservation methods to improve productivity of dryland crops
- Harnessing the maximum genotype x environment x management interactions for enhancing farm productivity and profitability
- Enhancing the nutritional quality of the food crop though agronomic intervention, especially agronomic biofortification.

Acknowledgement

We wish to thank all the Faculty Members of the Division of Agronomy for extending support and cooperation in preparing the research contributions of M.Sc. and Ph.D. students made during the period of 2007-16.

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M.Sc. and PhD theses submitted by the students in the Division of Agronomy, ICAR-Indian Agricultural Research Institute, New Delhi 110012 during the period of 2007-2016.

Environmental Science

S. Naresh Kumar and S.D. Singh

Introduction

Environmental pollution and climate change are the two major problems confronting agriculture today. Environmental impacts of agriculture and vice versa have raised serious questions about the sustainability of agricultural production systems because of widespread deterioration of soil, water and air quality. Climate change, caused by the increased concentration of greenhouse gases in the atmosphere, has emerged as one of the most prominent global environmental problems. Together, all these threaten the food security and livelihoods of millions of people in India. To sustain the food and nutritional security of the country, urgent need is to make Indian agriculture more resilient to environmental degradation and climate change. To address these challenges, the Division of Environment Science was established in the year 1993. Subsequently, in January 2012 the division was upgraded to Centre for Environment Science and Climate Resilient Agriculture (CESCRA). The Centre is doing pioneering work on development of sustainable agriculture; protection of environment; developing adaptation and mitigation strategies for climate change; development of simulation models and; providing scientific inputs for environmental policy for the benefit of farming community. The Centre provides inputs on to the India's National Communication to United Nations Convention Framework on Climate Change to the Government of India. Its scientists provide consultancy service to the Government of India with regards to international negotiations on climate change. Centre also provides consultancy services on environmental issues to various stakeholders. The Center collaborates with the public, private and civil society organizations to develop and demonstrate the technologies on-farm and promote them to enhance resilience of Indian agriculture and improve the livelihood of farmers. The mandates of the Centre include.

- To conduct basic and strategic research for environment and climate resilient sustainable agriculture with a special emphasis on rainfed and small-scale farmers.
- To impart post-graduate education and training on agriculture-environment inter-relationships.
- To provide advisory and consultancy services on environment monitoring, assessment and climate change in agriculture.

The Centre offers 23 courses for M.Sc (from 1992) and Ph D (from 1994) programmes in Environmental Sciences discipline. So far, the Centre has produced 81 PhD and 53 MSc students. The Centre offers training on simulation modelling, greenhouse gas emission estimation, climate change studies, environmental pollution assessment, and on resource conservation technologies to the students, scientists, academicians and those involved in agricultural development activities of state and central government. Scientists of the Centre have several externally funded projects, consultancy and contract research projects funded by agencies such as National Agricultural Science Fund, ICAR, DST, MoEF&CC, NTPC, WWF, TERI, etc. Many scientists of the Centre have been bestowed with prestigious awards and recognition of National and International repute.

Significant Student's Research Achievements

Climate Change And Agriculture

Climate change associated with rise in concentration of greenhouse gases (GHGs) is likely to affect crop production. Elevated temperature and carbon dioxide (CO_2) can affect growth and production of rice. Two

separate experiments were conducted during the *kharif* season of year 2013 to study the impacts of elevated CO_2 and temperature on yield and nutrient uptake of rice crop using the Free Air Carbon dioxide Enrichment Facility (FACE) and Temperature Gradient Tunnel (TGT) Facilities, developed by the Centre for Environment Science and Climate Resilient Agriculture (CESCRA) in the IARI farm, New Delhi. In the first experiment two levels of CO_2 (395 ppm and 550 ± 20 ppm) and 4 levels of nitrogen (no N, 75% of recommended dose, 100% of recommended dose and 125% of recommended dose) were used to study the impact of elevated CO_2 and N levels on yield and nutrient uptake. Grain and biomass yield of rice crop significantly increased with elevated CO_2 (550 ppm) compared to ambient CO_2 (395 ppm) level. The plant growth parameters and yield attributes such as plant height, number of tillers, number of panicles per pot, panicle length and number of grains per panicle also increased with elevated CO_2. However, with elevated temperature, the grain and biomass yield of rice crop significantly decreased. In elevated temperature treatment (+3.9 ºC) application of 25% additional dose of N prevented the yield loss of rice by 5.4% as compared to 100% recommended N dose i.e., 120 kg N ha^{-1}. Reduction in rice yield at higher temperature was mainly attributed to reduction in number of tillers, number of panicles per pot, panicle length, number of grains per panicle and harvest index and increase in spikelet sterility. Uptake of major nutrients increased under elevated CO_2 condition while it decreased under elevated temperature condition. Agronomic efficiency of N application in rice crop was higher under elevated CO_2 condition while recovery efficiency was not affected by CO_2 level. At elevated temperature, however, agronomic as well as recovery efficiency of N in rice crop decreased. The study showed that elevated CO_2 could increase yield and nutrient uptake by rice whereas elevated temperature decreased yield and nutrient uptake. Application of additional N over the current recommended dose could help in adaptation to prevent yield loss due to climate change.

Fig. 1: Rice plants grown in elevated CO_2 (in FACE) and temperature (in TGT) conditions

Crops with the C_3 and C_4 photosynthetic pathway like rice and maize respectively are very important to global food supply and are supposed to have impacts from rising atmospheric CO_2. Thus to study interactive effect of CO_2 and nitrogen fertilization, rice (C_3) and maize (C_4) crops were grown in FACE under ambient [CO_2] (380 ppm, AC) and elevated [CO_2] (550 ppm, EC) conditions with five levels of nitrogen (N0=No external supply, N1= 50 kg ha^{-1}, N2=100 kg ha^{-1}, N3=150 kg ha^{-1} and N4=200 kg ha^{-1}) in both AC and EC treatments. Study concludes that, plants responded more at EC under N treatment. Elevated CO_2 enhanced the economic and biological yield of both C3 (rice) and C4 (maize) crops irrespective of nitrogen levels, but the growth and

yield response to CO_2 fertilization was recorded to be higher in C3 crop-rice (12%) compared to C4 crop-maize (7-9%) in all nitrogen levels. However, nitrogen response in respect of growth and yield of both the crops was found to be higher under elevated CO_2 than under ambient CO_2 condition. Increase in economic yield of rice and maize under elevated CO_2 condition was mainly attributed to marked increase in the number of panicles per pot and slight improvement in grains per panicle, while 1000 grain weight was unaffected under same. In maize, however the CO_2 fertilization enhanced the grain yield mainly by improving the number of grains per cob without affecting the number of cobs per plant and 1000 grain weight. Rice being a C_3 crop showed higher response to EC for net photosynthetic rate than C_4 maize at flowering. This suggests that the main challenge is how to enhance sink capacity to utilize the photosynthate concurrently, in order to maximize grain yield under EC. On the other hand, stomatal conductance and transpiration rate was reduced in both the crops. Due to EC, there was slight dilution of nutrient concentration in rice but insignificant in maize. However, total nutrient uptake and their use efficiency in rice and maize was slightly higher due to increase in plant biomass under EC condition. Increased fertilization dose led to nullify the nutrient dilution effect of EC and maintained the quality of crop biomass. Similarly, the concentration of protein reduced in rice and maize grains under EC could partially be mitigated through higher dose of nitrogen fertilization. Thus, in order to harness growth and yield response of EC vis-a- vis to maintain the quality of crop biomass, the nutrient application level needs to be revised under changing climatic scenario in future.

Fig. 2: Rice and maize plants grown in elevated CO_2 (in FACE) with varying levels of nitrogen

Crops with the C_3 and C_4 photosynthetic pathway like rice and maize respectively are very important to global food supply and are supposed to have impacts from rising atmospheric CO_2. Thus to study interactive effect of CO_2 and nitrogen fertilization, rice (C_3) and maize (C_4) crops were grown in FACE under ambient [CO_2] (380 ppm, AC) and elevated [CO_2] (550 ppm, EC) conditions with five levels of nitrogen (N0=No external supply, N1= 50 kg ha $^{-1}$, N2=100 kg ha $^{-1}$, N3=150 kg ha $^{-1}$ and N4=200 kg ha^{-1}) in both AC and EC treatments. Study concludes that, plants responded more at EC under N treatment. Elevated CO_2 enhanced the economic and biological yield of both C3 (rice) and C4 (maize) crops irrespective of nitrogen levels, but the growth and yield response to CO_2 fertilization was recorded to be higher in C3 crop-rice (12%) compared to C4 crop-maize (7-9%) in all nitrogen levels. However, nitrogen response in respect of growth and yield of both the crops was found to be higher under elevated CO_2 than under ambient CO_2 condition. Increase in economic yield of rice and maize under elevated CO_2 condition was mainly attributed to marked increase in the number of panicles per pot and slight improvement in grains per panicle, while 1000 grain weight was unaffected under same. In

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The alarming and unprecedented rise in the concentration of greenhouse gases in the atmosphere, primarily CO_2, under present global climate change scenario, warrants a pressing need and importance to assess the direct and indirect effects of elevated CO_2 on different crops and plant metabolism. Water and nitrogen deficiency are major factors limiting the productivity and these management factors (e.g. irrigation and fertilization) are expected to interact in complex ways to determine the impact of climate change on crop production. Mustard is very sensitive to weather and climatic variables and therefore it is very likely that climate can have significant impact on its production and quality. The present investigation was carried out to study the effect of elevated CO_2 on growth, yield and quality of *Brassica juncea* under different water and nitrogen regimes in Climate Change Research facility at Indian Agricultural Research Institute, New Delhi with twelve treatment combinations of CO_2, water and nitrogen with quadruple replications laid in factorial CRD and as well under ambient (390ppm) and elevated CO_2 (550 ppm). Plants grown under elevated CO_2 condition performed better than ambient CO_2 irrespective of water or nitrogen levels. Although water stress depressed these parameters significantly under both CO_2 levels however, additional N mitigated its negative impact. The magnitude of loss due to moisture stress was lower under elevated CO_2 compared to ambient CO_2. The plants under elevated CO_2 with water stress performed at par with plants under ambient CO_2 with water sufficient condition. CO_2 enrichment increased net photosynthetic rate with simultaneous decrease in stomatal conductance and transpiration rate at all stages resulting in greater carbon fixation and increased water use efficiency. Although additional N supply invariably increased growth and yield parameters, however response under elevated CO_2 was found to be greater whereas plants under ambient CO_2 were N saturated and did not show N response beyond optimum N level. Elevated CO_2 increased carbon content with corresponding increase in carbohydrates thereby decreasing N and protein content and increasing C/N ratio, however additional N supply was found to improve N status. CO_2 fertilization enhanced oil content in seeds but moisture stress depressed oil content significantly which declined further as a result of additional N fertilization. Alterations in fatty acid composition were observed where saturated fatty acids decreased and unsaturated fatty acids increased thereby enhancing oil quality. Among inorganic nutrients (macro- and micronutrients) P, K, S, Ca, Mg, Fe and Zn were adversely affected due to CO_2 enrichment. Moisture stress diminished their status and additional N supply was found to aggravate it further. Contrastingly, increased N supply improved P status. However in seed, while Ca and S were found to increase, K, Mg, Fe and Zn decreased and P remained unaffected.

Different climate change variables like elevated CO_2 and temperature may considerably affect crop productivity and global food security. An experiment was conducted growing maize (variety PEHM 5) and wheat (variety WR 544) in open top chambers (OTCs) in two levels of carbon dioxide i.e., ambient (400 ppm) and elevated (550 ± 20 ppm) and three levels of temperature i.e., ambient, ambient +1.5°C and ambient +3.0°C in a factorial completely randomized design to study the effects of elevated atmospheric CO_2, temperature and their interactions on crop growth, yield, grain quality, soil nutrient availability and microbial diversity in

a maize-wheat cropping system. Maize and wheat crops were grown during *kharif* and *rabi* seasons for two years during 2013-15 at IARI, New Delhi, India. Elevated CO_2 increased grain yield of maize and wheat by 53.69% and 21.36%, respectively compared to ambient CO_2. Similarly, biomass yield, leaf area index (LAI), photosynthetic rate and yield attributes of both the crops also increased with elevated CO_2. However, elevated CO_2 had no effect on days to 50% tasseling and anthesis; and pH, electrical conductivity (EC), organic C content and bacterial diversity of soil but decreased N, P and crude protein contents in grains and available N content of soil. Elevated temperature by 1.5 °C and 3.0 °C decreased LAI, days to 50% tasseling and anthesis, grain yield and yield attributes of both the crops and organic C content but increased N, P, K and crude protein contents of grain and available P content of soil as compared to ambient temperature. Simultaneous elevation of CO_2 and temperature increased LAI and harvest index (HI) of maize but decreased days to 50% tasseling and anthesis, N and crude protein contents in grain and available N, P and K contents of soil. However, pH, EC and organic C contents of soil were not affected due to combined effect of elevated atmospheric CO_2 and temperature. Maximum dehydrogenase activity, bacterial richness, abundance and diversity in soil were observed under elevated CO_2 along with 1.5°C temperature rise. Grain yields increased at elevated CO_2 with 1.5°C temperature rise but further increase in temperature had no impact on grain yield. The study indicated that elevated CO_2 was able to compensate for the negative effect of elevated temperature up to 3.0 °C rise on yields of both crops. It can be concluded that elevated CO_2 and temperature had significant influence on yield and quality of maize and wheat crops; and fertility, microbial activity and bacterial diversity of soil in maize-wheat cropping systems in the north-west India.

Increase in the concentration of atmospheric greenhouse gases, mainly carbon dioxide (CO_2) have significant impact on crop, in terms of increase in photosynthesis rate and growth. The experiment was undertaken during the *kharif* season of year 2014 to study the impacts of elevated CO_2 and cyanonacterial inoculation on growth, yield and N fixation of legumes under different doses of phosphorus (P) using Free Air Carbon dioxide Enrichment Facility (FACE), CESCRA, IARI, New Delhi. Two legume crops viz. mungbean and cowpea were grown in two CO_2 levels, ambient (395 ppm) and elevated (550±20 ppm) in pots with five levels of P (0, 8, 12, 16 & 20 mg P kg^{-1} soil) and 2 levels of *Calothrix* sp. (with & without cyanobacteria) inoculation. Elevated CO_2 and cyanobacteria application increased leaf area, photosynthesis rate and chlorophyll content in both the crops. Elevated CO_2 level increased seed yield by 33.4% in mungbean crop and pod yield by 24.6% in cowpea crop. Cyanobacterial inoculation increased seed yield by 16.4% in mungbean and pod yield by 14.8% in cowpea crop. N fixation increased in elevated CO_2 and higher P doses in both the crop by increasing total nitrogenase activity. P dose of 16 mg kg^{-1} recorded highest N fixation in mungbean while P dose of 20 mg kg^{-1}showed maximum N fixation in cowpea crop. Plant N uptake increased by 26% in mungbean and 16.3 % in cowpea at elevated CO_2 level. Soil N status and other biological properties improved in high CO_2 and cyanobacteria applied treatments. Cyanobacteria improved available P status of soil. P uptake in mungbean and cowpea plant increased by 30.9% and 17.7% respectively at elevated CO_2 level. It can be concluded that elevated CO_2 and cyanobacteria application will enhance productivity and N fixation in legumes, with cowpea being more efficient than mungbean crop and as well supply of P simultaneously will play crucial role in future climate change scenario.

Fig. 3: Legumes grown in elevated CO_2 (in FACE) with varying levels of phosphorus.

Carbon dioxide (CO_2) is the dominant greenhouse gas (GHG) in the atmosphere and its concentration is increasing leading to global warming and climate change. Increase in the concentration of CO_2 has significant

impacts on crops. Rice is the most important food crop, feeding about a half of the world's population. Elevated CO_2 will affect rice productivity along with change in nutrient demand and supply of the crop. The current study was undertaken during the *kharif* season of year 2015 to study the impacts of elevated CO_2 on transplanted and direct seeded rice under varying levels of phosphorus. Rice crop (variety Pusa Basmati 1509) was grown in pots both inside and outside the Free Air Carbon dioxide Enrichment Facility (FACE), in the IARI farm, New Delhi. Carbon dioxide level of 550 ppm was maintained inside the FACE ring whereas the ambient CO_2 concentration was 400 ppm. The crop was grown with four levels of P i.e. control (no P), 75% of recommended dose, 100% of recommended dose and 125% of recommended dose of P. Increased CO_2 level increased leaf area, photosynthesis rate and tiller number in both transplanted and direct seeded rice. Grain yield of transplanted rice increased by 11.2% and of direct seeded rice by 13.8% under elevated CO_2 treatment. Phosphorus application enhanced grain as well as biomass yield of the crop. The P dose upto 27 mg kg^{-1} soil increased yield of rice both under transplanted and direct seeded condition. Although P concentration in rice grains did not change in elevated CO_2 treatment but higher grain yield resulted in higher P uptake of the crop. Grain P uptake increased upto P dose of 27 mg kg^{-1} while total P uptake increased upto P dose of 33 mg kg^{-1} soil. Available phosphorus in soil was higher in transplanted rice than direct seeded rice crop. At flowering stage of the crop soil available P was 12.3% less in direct seeded rice than transplanted rice under ambient condition. But in elevated CO_2 treatment soil available P was 10.8% less in direct seeded rice than transplanted rice at this stage. This shows that availability of soil P improved under high CO_2 condition in direct seeded rice crop. Better root growth and higher activity of both acid and alkaline phosphatase enzymes in elevated CO_2 treatment resulted in increase in availability of soil P. Both agronomic and recovery efficiency of P increased with elevated CO_2 condition.

Two promising rice cultivars viz., Pusa 44 and Pusa Sugandh 2, and wheat varieties namely PBW 343 and UP 2338 were subjected to various thermal treatments such as normal ambient temperature (T0), high day and night temperature (T1) during day and night), high day temperature T2 during daytime only) and high night temperature T3 during night time only) from transplanting to maturity. High day and night temperature (T1) caused maximum growth, yield and quality effect on rice followed by high night (T3) and high day (T2) temperature treatment. High temperature stress enhanced plant height and foliage growth, however, the same reduced the biomass and grain yield of rice varieties significantly. The reduction in grain yield of rice by high thermal stress was attributed to marked decrease in the number of panicles per pot, number of spikelets and grains per panicles, 1000 grain weight and significant increase in spikelet sterility. High temperature induced pollen sterility has led to spikelet sterility, which is a fundamental cause for grain yield reduction. In general, HDNT (T1) affected the growth, yield and grain quality of wheat cultivars to the maximum extent followed by HDT (T2) and HNT (T3). High temperature caused marked increase in the stature of plants by enhancing the length of culm and leaf. High temperature stress manifested significant reduction in grain yield, which was mainly attributed to marked reduction in vegetative and reproductive growth phases, biomass production, number of spikes per pot, number of spikelets and grains per spike and 1000 grain weight. The decrease in biomass per degree increase in temperature is far less than the decrease in grain yield indicating the pronounced effect of high day and night temperature on yield contributing factors rather than on biomass production. High thermal stress showed marked impact on biochemical components of plant tissues. In general, total nitrogen and non-protein nitrogen content in different plant parts increased, while true protein nitrogen reduced drastically by heat stress. Carbohydrates especially starch and hemi-cellulose contents in rice shoots reduced, whereas sugars content in the same increased under hyper thermal stresses. The cell membrane stability of leaves decreased, while cell membrane injury percentage of it increased markedly under high thermal stress. High temperature also caused marked reduction in physical as well as cooking quality of grains. High temperature reduced the head rice percentage, length of cooked rice kernel and elongation ratio, but increased the brown rice (%), total milled rice (%) and gelatinization temperature of rice grains in both the varieties. Importantly, high

temperature treatments reduced the grain starch content and increased grain protein content. Pusa Sugandh 2 though more sensitive to high day and night temperature, out yielded the performance of Pusa 44.

Agricultural crop production in India could be adversely affected by 1.1-6.4 ^{o} C rises in temperature by the end of 21st century as per fourth assessment report of IPCC, 2007. Effects of elevated temperature (TO = Ambient temperature, T1 = TO+1.5-2.5, T2= TO+2.5 – 5.0 and T3 = TO + 5-6.5 oC) on growth, productivity and quality of pulse and oilseed crops were studied in the current investigation during year 2006-2007. In this research pulse crop (moong and chickpea) and oilseed crops (groundnut, mustard and soybean) were grown the green house. Growth parameters (plant height, number of physiological parameters (net photosynthesis rate, stomatal conductance, pollen sterility etc.), nutrients (N,P and K), yield components (pod/plant, num ber of seed /plant, test weight etc.) and seed quality parameters (oil content, protein content and seed density) were analysed. Result show ed reduction in physiological parameters such as net photosynthesis rate, stomatal conductance, yield and quality under elevated temperature treatments (T1, T2 and T3) as compared with ambient temperature whereas growth parameters such as plant height and pollen sterlity were increased with increasing temperatures. Percentage decrease of economic yield / m2 per degree temperature for moong and chickpea was 4.99 to 10.79% and 2.26 to 3.51% under elevated temperature. This indicates that in terms of economic yield, moong was more temperature sensitive under elevated temperature. Percentage decrease of economic yield/m2 per degree temperature for groundnut, mustard and soyabean was 2.0 to 6.0, 2.5 to 3.1 and 3.1 to 8.1% respectively under elevated temperature. The result indicate that in terms of economic yield, among the oilseed crop studied, soybean is most sensitive and mustard is least to elevated temperature. Variation in economic yield under elevated temperature and its percentage decrease per degree temperature rise clearly indicate that in both pulse and oilseed crop, kharif season crop (chickpea and mustard). Groundnut showed intermediate response for decrease in economic yield under elevated temperature.

Fig. 4: Ground nut and mung bean crops grown in temperature gradient tunnel.

Increase in the concentration of atmospheric greenhouse gases, mainly carbon dioxide (CO_2) has significant impact on crop, in terms of increase in photosynthesis rate and growth. It also has effect on the biogeochemical cycle. Legumes get advantage in elevated CO_2 condition than non-legumes by fixing additional nitrogen (N). The following study was undertaken during the *kharif* season of year 2014 to assess the impacts of elevated CO_2 and cyanonacterial inoculation on growth, yield and N fixation of legumes under different doses of phosphorus (P) using Free Air Carbon dioxide Enrichment Facility (FACE), developed by Centre for Environment Science and Climate Resilient Agriculture (CESCRA) in the IARI farm, New Delhi. Two legume crops *i.e.* mungbean and cowpea were grown in two CO_2 levels, ambient (395 ppm) and elevated (550±20 ppm). Crops were grown in pots with five levels of P (0, 8, 12, 16 & 20 mg P kg^{-1} soil) and 2 levels of *calothrix sp*. (with & without cyanobacteria) inoculation. Elevated CO_2 and cyanobacteria application increased leaf area, photosynthesis rate and chlorophyll content of both the crops. Elevated CO_2 level increased seed yield by 33.4% in mungbean crop and pod yield by 24.6% in cowpea crop. Cyanobacterial inoculation increased seed yield by 16.4% in mungbean and pod yield by 14.8% in cowpea crop. N fixation increased in elevated CO_2 and higher P doses in both the crop by increasing total

nitrogenase activity. P dose of 16 mg kg^{-1} recorded highest N fixation in mungbean while P dose of 20 mg kg^{-1}showed maximum N fixation in cowpea crop. Plant N uptake increased by 26% in mungbean and 16.3 % in cowpea at elevated CO_2 level. Soil N status and other biological properties improved in high CO_2 and cyanobacteria applied treatments. Cyanobacteria improved available P status of soil. P uptake in mungbean and cowpea plant increased by 30.9% and 17.7% respectively at elevated CO_2 level. The study revealed that elevated CO_2 and cyanobacteria application will enhance productivity and N fixation in legumes, with cowpea being more efficient than mungbean crop. Supply of P is important for productivity and N fixation of legumes and in present climate change scenario P application will play crucial role.

Tropospheric ozone (O_3) is an important secondary air pollutant, affecting agriculture by reducing yields and degrading the quality of agricultural products. Wheat an important cereal crop has been reported to be sensitive to elevated ozone concentrations. The use of certain antioxidant chemicals can help in mitigating the negative impacts of tropospheric ozone on crop growth and yield. However many of these chemicals may be phytotoxic at higher concentration and hazardous to the environment. So there is a need to search for natural occurring and environment friendly antioxidant chemicals for alleviating the ozone stress in plants. A two year experiment was conducted at the research farms of IARI, New Delhi during the year 2010-11 and 2011-12 growing wheat crop under the following treatments i) Charcoal Filtered Air (CF), ii) 1% Ascorbic acid (AA), iii) 100 ppm Quercetin (Q), iv) 10% Marigold leaves extract (*Tagetes patula* L.) (T) and v) Elevated ozone control (C). 25-35 ppb of additional O_3 over the ambient O_3 levels was maintained in all the open top chambers (OTC) except in CF air treatment. The seasonal average daily mean O_3 concentration in ambient air was 38 ppb in 2010-11 and 29 ppb in 2011-12 during the crop growth period. The Photosynthetic rate (P_n) of the plants treated with antioxidants ascorbic acid, quercetin and marigold leaves extract significantly increased by 22-23%, 17-18% and 10-12% respectively in both the years as compared to control. The yield of wheat in the ascorbic acid treatment was 23-26% higher as compared to control in both the years. The decrease in antioxidant enzymes (superoxide dismutase, catalase and peroxidase) activity varied from 5 to 34% in 2011-12 under the different antioxidant treatments as compared to control at flowering. The Rubisco enzyme activity increased in all the antioxidants treated plants. Significant increase was observed in the belowground carbon (microbial biomass carbon, dissolved organic carbon and root biomass) in different antioxidant treatments as compared to elevated ozone alone. The application of antioxidant chemicals increased the sugar and starch content in both leaves and grains of wheat over elevated ozone alone. The protein content in grains increased under elevated ozone as the grain yield decreased under this treatment. The micronutrients (Cu, Fe, Zn and Mn) content in wheat grains increased under antioxidant treatments, while the macronutrients (Ca, Mg and K) content was lower in these treatments as compared to elevated ozone alone. The antioxidant chemicals, ascorbic acid, quercetin and marigold leaves extract were found effective in providing protection against negative impacts of O_3. Marigold leaves extract may be effectively used as natural protectant for alleviating the tropospheric O_3 stress in wheat.

The atmospheric concentrating of O_3 an air pollutant and CO_2 a major green house gas has been increasing at an alarming rate in the recent past. The changes in the atmospheric ozone and CO_2 concentration are not only affecting the crop productivity and physiological parameters but also altering its nutritional quality. Field experiments were carried out during *rabi* season (Oct-March) 2009-10 & 2010-11 at research farm of Indian Agricultural Research Institute, New Delhi to assess the effects of different ozone treatments alone and along with CO2 interaction on yield and quality of Indian mustard. Mustard crop was grown in open-top chambers with six treatments non-filtered air (NF), charcoal filtered air (CF), elevated ozone (EO), elevated ozone with elevated carbon dioxide ($EO+CO_2$), non filtered air with elevated carbon dioxide ($NF+CO_2$) and chamber less ambient control (AC). All the growth parameters (plant height, root length, leaf area index, specific leaf area and dry matter), seed yield, biological yield, harvest index, quality parameters (oil content, protein content, Fe, Ca, Mg, Zn, S content) and physiological parameters

(photosynthetic rate, stomatal conductance, total chlorophyll and relative water content) were observed to be highest in CF treatment and lowest in EO treatments. Elevated CO_2 increased the growth and yield attributes, photosynthetic rate, total chlorophyll, relative water content and oil content but decreased the stomatal conductance, protein content, macro and micro nutrients in seed of mustard. An elevated carbon dioxide concentration along with elevated ozone was able to counter some of the negative impact of elevated ozone concentrations on plant growth and yield. However, elevated ozone and CO_2 interaction reduced the nutritional quality (protein content, macro and micro nutrients) of mustard seeds.

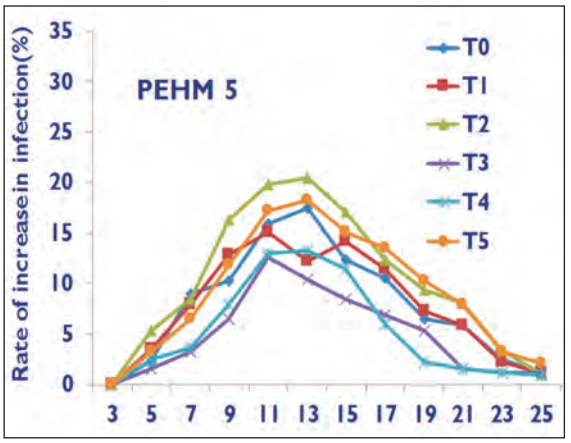

Fig. 5: Effect of elevated temperature and carbon dioxide on maydis leaf blight of maize; T0 = AmT & AmC; T1 = +1.5ºC & AmC; T2 = +3.0ºC & AmC; T3 = AmT & ElC; T4 = +1.5 ºC & ElC; T5 = +3.0 ºC & ElC.

Effect of elevated temperature and CO_2 on maydis leaf blight (MLB) disease development on PEHM-5 and CM-119 maize genotypes and their disease tolerance potential was assessed. Results of the study showed that elevated levels of temperature and CO_2 exposure affected the functional traits (growth, physiology and biochemistry) of maize genotypes as well as MLB disease development. It was observed that between the two maize genotypes effect of temperature and CO_2 treatments was more adverse on CM-119 as compared to PEHM -5 genotype with respect to functional traits as well as MLB disease severity. Among the treatments, at +3°C temperature elevation with ambient CO_2 level effect was most detrimental on functional trait and MLB disease development as compared to control i.e., ambient temperature and CO_2 level. MLB development is more influenced by the combination of temperature and CO_2 than by CO_2 level alone. MLB disease severity was less on plants exposed to elevated CO_2 and more on plants exposed to elevated temperature. Different disease indices also indicate that the tolerance of PEHM-5 was more than CM-119 and it was more adversely affected by temperature stress. Tolerance potential of different growth stages of maize to MLB disease was more at dent stage as compared to vegetative and tasseling stages to elevated CO_2 and temperature treatments.

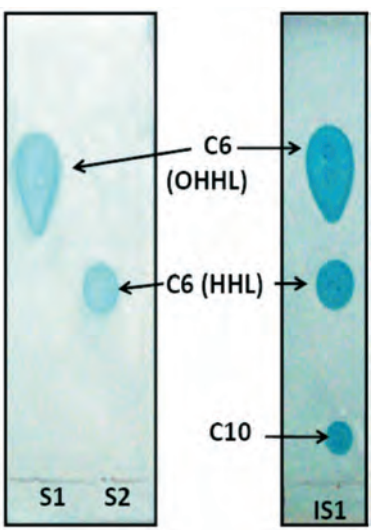

Fig. 6: Novel Signal molecule from *Pectobacterium carotovorum* with retention factor 0.19

In the current climate change scenario and future predictions, the plant pathogenic interaction is expected to become more complex. Plant pathogens cross talk among themselves by producing and secreting certain kind of signal molecules, the mechanism called Quorum Sensing (QS). In *Pectobacterium carotovorum* pv. *carotovorum*, cell wall degrading enzymes (virulent factor) is produced by this kind of mandatory cross communication procedure (QS), which causes maceration of plant tissue. Our aim was to evaluate the impact of climate change variables on QS mediated pathogenesis of soft rot in tomato. From this investigation, one novel and more virulent *Pectobacterium carotovorum* pv. *carotovorum* strain (Gene Bank Accession no.GU590785) was isolated and found to produce C6-OHHL, C6-HHL along with one novel QS active compound namely C10-Undecanoyl Homoserine Lactone (confirmed by LC-MS analysis). C6-OHHL was found to play key role for regulating pathogenesis and the novel C10-Undecanoyl HSL was found to stimulate the function of C6-OHHL. In phytotron, among six different elevated CO_2 and temperature interactions, in T3C2 (600 ppm, 28°C) maximum C6-OHHL, Cellulase and disease severity (73.18%) was noticed. At elevated CO2 and temperature (upto a certain limit) soft rot disease severity increased.

Climate is the primary determinant of agricultural productivity, as crop yields are influenced by many environmental factors viz., temperature and precipitation, which may act either synergistically or antagonistically with other factors. Groundnut is a thermo-sensitive crop and short or prolonged periods of high temperature during reproductive development of peanut are known to cause significant yield losses. The relationships between the C, N, P and S cycling and functional groups of microorganism such as plant growth promoting rhizobacteria (PGPR) and their influences on plant growth are potential indicators in the evaluation of disturbances of the soil environment. The impact of elevated temperature (+2-3°C) at different stages of crop growth development in Groundnut Crop cv variety B-95 grown in small tunnels at IARI farm on some functional bacterial populations in rhizosphere soil was investigated. Rhizosphere soil samples were collected at the vegetative, flowering, pegging and maturity stages of groundnut. Using cultivation-dependent approaches, bacterial population involved in nitrogen-fixing, phosphate-solubilizing, potassium-dissolving and PGPRs were measured in all the treatments. The study revealed that the stage where the temperature was elevated from germination till maturity resulted in significantly higher ($P < 0.001$) growth of nitrogen-fixing (7.0%), phosphate-solubilizing (6.6%) and potassium-dissolving (3.0%) bacteria at 75 days after sowing as compared to the crop grown under ambient temperature. The impact of elevated temperature (+2-3°C) specifically from germination to maturity in Groundnut Crop has resulted in significant increase in functional bacterial population and there seems to be no loss of any PGPR traits amongst the *Pseuodomonas, Azotobacter, Acetobacter* and *Enterobacter* population.

Multiple stresses and crops

In India droughts and floods in a single crop season have recently caused severe crop loss. Therefore understanding crop response to these sequential stresses in a season is important for carrying out large scale-impact assessments and developing adaptation strategies through simulation analysis. Keeping in view of the commercial importance of maize and groundnut a study was undertaken to quantify the response of maize and groundnut to short-term multiple stresses related to floods and drought and stimulate the impact of such short term multiple stresses on groundnut and maize using InfoCrop model. For this, a field study was conducted with maize and groundnut during monsoon season of 2010 where maize and groundnut crops were subjected to flooding and drought alone and in five different combinations. The observed values on phenology, leaf area, dry matter and yield from the above experiment were compared with those of simulated ones using InfoCrop groundnut and maize models. Results indicate that drought conditions hastened the phenology of crops, while excess water delayed flowering and crop maturity. In ground nut drought during early phase caused more reduction in dry matter accumulation than that occurred during seed filling stage. Excess soil moisture seems to increase the vegetative growth and in absence of any other stress increased the crop yield. However, when the crop encountered excess water and drought during the flowering and pod filling stage, it stands to lose the yield. In maize, total biomass was most affected in plants exposed to drought stress during the vegetative growth period and then subjected to excess water during grain filling period. Occurrence of drought during vegetative growth and flowering or pod filling phase, it caused reduction in crop yield. Occurrence of drought during vegetative growth period or during flowering caused more reduction in grain yield. InfoCrop ground nut and maize models could be satisfactorily calibrated to rainfed and irrigated conditions. The ground nut model is good in simulating the phenology, while requires improvement for simulating the dry matter and yield for simulating the crop performance under sequential stresses. In conclsion, the results indicated that the impacts of drought and flood on crop growth and yield are multi-dimensional. Overall results on phenology, leaf area index, biomass accumulation and distribution, yield and yield components indicate that the plants responded differently to sequential stresses depending on the coincidence of the sequential stress. The sensitivity of crops and crop parameters also differed with regards to sequential stresses. Therefore, more experimentation is required to test the performance of models in these situations and also for refining them.

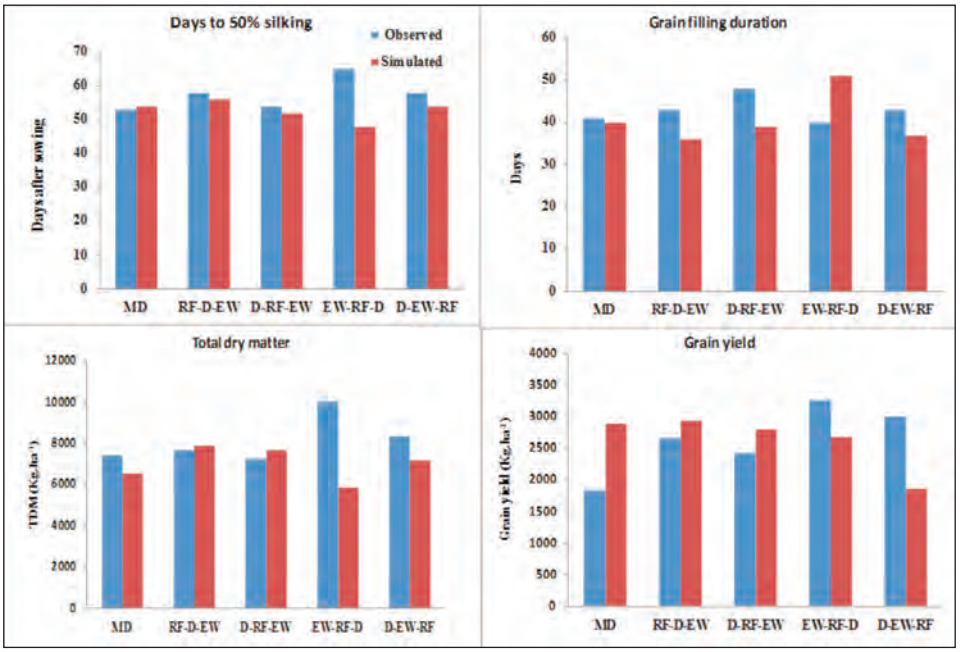

Fig. 7: Effect of sequential stresses on maize crop growth and yield. Comparison of observed and simulated values

In India, floods and droughts accompanied by high temperature in a single crop season have recently caused severe crop loss. Therefore understanding crop response to these sequential stresses in a season is important for carrying out large scale-impact assessments and developing adaptation strategies through simulation analysis. Keeping in view of the commercial importance of soybean, a study was undertaken to i) quantify the response of soybean to short-term multiple stresses related to floods, drought and high temperature and to ii) simulate the impact of short-term multiple stresses of floods, drought and high temperature on soybean using InfoCrop model. For this, a field study was conducted with soybean (var PUSA 1495) during monsoon season of 2011 by imposing flooding (excess water-EW), drought (DR) and high temperature (+2°C with adequate irrigation; HT) stresses in different sequences. Sequential stresses EW-DR-HT, DR-HT-EW and HT-EW-DR were imposed on crop 30 days after sowing (DAS) for 20 days each (30-50; 50-70 and 70-90 DAS) as per the sequence mentioned above. Crop was also maintained in irrigated, rainfed, moderate drought and high temperature conditions. In soybean, drought and high temperature during entire crop growth period or during early growth period hastened the flowering; while excess water delayed it. Even though pod filling duration (PFD) reduced in moderate drought conditions, a combination of factors influenced the duration in sequential stresses. Excess water enhanced the leaf area index, and stem and leaf dry matter particularly in vegetative period; while drought in vegetative phase followed by high temperature during flowering periods reduced them. Dry spell during vegetative growth followed by high temperature coinciding flowering even with adequate soil moisture, and then heavy rains during PFD significantly reduced the biomass and seed yield as also in case moderate drought situation and in high temperature conditions. However, in the event of heavy

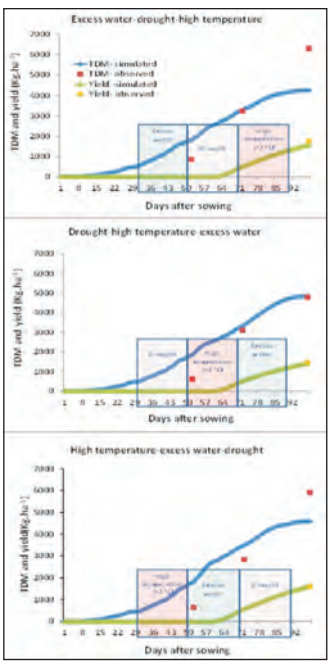

Fig. 8: Time series performance of InfoCrop-soybean model in simulation of TDM and yield under sequential stresses as against observed values

rainfall coinciding vegetative period followed by dry spell during flowering and high temperature situation with adequate soil moisture during pod filling will not affect the soybean yields. InfoCrop-Soybean model satisfactorily simulated the effect of single stress but shown tendency to over-estimate the biomass during vegetative period. Even though the model has over-estimated the biomass while simulating the effect of sequential stresses, the phenology and seed yield is simulated within the acceptable range of error. Results indicate that model can be used for simulating the impacts of climatic variability after minor refinement regarding LAI and total dry matter particularly during vegetative growth phase.

Climate change, crops and cropping systems

Agriculture production and food security are closely linked with the climate conditions of a region. Climate change is projected to increase risks in terms of rise in temperature and enhanced irregularities in rainfall apart from other changes challenging crop production. In order to quantify the possible impacts of climate change and variability on wheat and rice crops, analysis of the climate change data obtained from PRECIS, a Regional Climate Model, field experiments and InfoCrop based simulation studies on impacts of clilmatic variables were carried out. Results indicated that PRECIS had a bias towards higher precipitation along with more intense warm and cold events in the baseline simulation. Analysis indicated that projected increase in temperature is higher for north India, which at present experience low temperatures. Monsoon rainfall and *rabi* season temperature are projected to increase in A2 and B2 2080 scenarios. These climatic changes are likely to reduce the wheat and rice yields in Indo-Gagentic plains of India. Results from field experiments indicated that panicle initiation to flowering period is found to be highly sensitive to high temperature and drought stress. Simulation studies using InfoCrop model also indicated maximum adverse effect on wheat yield mainly due to temperature rise during II fortnight of March in Punjab, I fortnight of March in Haryana, II fortnight of February in Uttar Pradesh, North Rajasthan, and I fortnight of January in North-West Madhya Pradesh and South Rajasthan. These periods generally coincides with flowering time. The yield reduction was found to be higher in late sown crop than the normal one. Drought stress related risk assessment study using crop simulation model showed that a 50% rainfall deficit for 30 days during the second month of crop growth after transplanting (reproductive stage), can cause almost crop failure in Maharashtra, Tamil Nadu and Karnataka, while Uttar Pradesh and MP may register about 35% loss in rice yields. Karnataka and Tamil Nadu are more prone to yield reduction due to variability in rainfall in comparison to Marharashtra, Uttar Pradesh and Madhya Pradesh. The outputs of this study are expected to provide critical inputs in developing climatic-risk-coping strategies for reducing the vulnerability of the large fraction of the resource poor population.

Climate change effects of elevated temperatures and increased occurrences of erratic rainfall patterns and water shortageshas resulted in increased incidences of crop failure, reduced income, and food instability globally and on the Indian-sub continent. The mung bean (*Vigna radiata* (L.) Wilczek)- wheat (*Triticum aestivum* L.) cropping system is relatively new and has been adopted to meet the ever increasing food and dietary needs of the Indian population where other traditional cropping systems have failed. Knowledge of the performance of the cropping system when exposed to elevated temperature and water stress may prove to be critical in terms of crop production in semi-arid regions which are facing climate change challenges. An experiment was conducted on rainfed mung bean variety Pusa Vishal which was then exposed to combined elevated temperature and water stress at 3 growth stages (vegetative, flowering and pod filling) by shedding off using a polyethylene shed while a wheat variety HD2967 at 3 moisture levels i.e. 100% CPE, 60% CPE and 30% CPE irrigation was exposed to combined elevated temperature and water stress at 3 growth stages (tillering, anthesis and milk) using the same polyethylene sheds. The crops were grown in sequence with 2 sowing dates (normal-NS and late- LS) during the Kharif season (June -October) and Rabi season (November to April) of 2014-15 and 2015-16 respectively at the IARI Research farm, New Delhi, India. Elevated temperature (7 - 9°C higher than ambient) and water stress (variable) in both crops was able to reduce yield by 39% and 33.7% (NS and LS crop in 2014 respectively), 42% to 42.6% (NS and LS crop in 2015 respectively) in mung bean while in wheat yield reduction was by

between 18% and 83% (in both NS and LS -2014-15) and 19% to 65% (in both NS and LS -2015-16) with the highest yield reduction obtained in wheat exposed to stress while at 30% CPE irrigation regimes in both NS and LS crops. Similarly length of phenological phases were reduced, TDM, LAI, photosynthesis and most yield attributes were significantly reduced in both crops when exposed to combined elevated temperature and waters stress. However, in wheat at tillering stage, exposure to elevated temperature and water stress resulted in higher photosynthetic rates during the cold winter period though the rates did not translate to increased yield and yield attributes. Crude protein and WUEinst increased in both crops when exposed to stress. In mung bean, the most sensitive growth stages was the anthesis stage followed by the pod filling stage, whereas the mung bean crop was able to recover marginally when exposed to elevated temperature and water stress at vegetative stage. In wheat the most sensitive stages were the anthesis and tillering stages, and wheat plants exposed to stress at tillering stage showed signs of recovery though yield would be lowered due to low number of tillers m^{-2} at tiller formation. The total water footprint (WFproc) from the 2014-15 cropping system was 2969 and 2244m^3 ton^{-1} for NS and LS crop respectively while for the 2015-16 was 3965 and 3908m^3 $ton^{-1}$for NS and LS crop respectively. In both seasons the percentage contribution of mung bean crop to the WFcrop was higher than that of wheat in both sowing dates. It can be concluded that combined elevated temperature and water stress has a momentous influence on growth parameters, yield and yield attributes of mungbean and wheat crops; the water footprint of cropping system varies according to yield and climatic conditions and that the WFproc is higher in mung bean than wheat. There is need for further studies on improving the WUE of the crops and maximizing gaseous exchange parameters in high temperature and limited water conditions so as to increase yield in both crops.

A field experiment was conducted during *kharif* season of 2013 at Experimental Farm of Indian Agricultural Research Institute, New Delhi. The experiment was laid out in factorial RBD with three replications. Treatment consist of three main plot as cropping systems-sole pigeonpea, pigeonpea + green gram (1:1) and pigeonpea + green gram (1:2) with three sub-plot as control, hydrogel and mulching having nine treatment combinations to ascertain effect of cropping system and moisture conservation practices on crop growth, yield and yield attributes, microbial growth, crop physiological parameters, nutrient content, soil moisture studies and economics of pigeon pea and green gram under rainfed conditions. The cropping systems failed to cause any significant variation in growth attributes of pigeonpea and green gram. Similar result was found in case of yield and yield attributes in both crops. Data pertaining to growth attributes of pigeonpea and green gram such as plant height (cm), LAI were positively influenced by moisture conservation practices except that no. of branches/plant. Among the moisture conservation practices, mulching increased plant height and LAI by 8.2% and 11.4% over control whereas the hydrogel treatment remained on par with control. In case of green gram, cropping systems showed non-significant effect on plant height and LAI whereas moisture conservation practices i.e., mulching had significantly increased plant height and LAI by 24.2% and 10.8% over control. The cropping system had no effect on uptake of N, P, K and S but positively influenced by moisture conservation practices in pigeonpea crop. Among the different moisture conservation practices mulching significantly increased N, P, K and S uptake in seed (43.4%, 44.3%, 44.2% and 43.9%, respectively) and stalk (31.8%, 34.2%, 32.5% and 33.8% respectively) over control which was followed by hydrogel teatment. Among moisture conservation practices mulching gave significantly higher net photosynthetic rate as well as instantaneous water use efficiency over control under both pigeonpea and green gram. Bacterial and fungal population increased in mulch treatment by 28.8% and 32.4% respectively over control. Among the moisture conservation practices, mulching recorded significantly higher soil moisture percentage as compared to hydrogel and control. Among the moisture conservation practices mulching gave higher pigeonpea yield which was followed by hydrogel treatment. Green gram yield under mulching treatment was higher as compared to control and hydrogel treatment. Mulching treatments provided a seed yield advantage of 41.9% and 14.3% in pigeonpea and green gram respectively compared to control. Pigeonpea + green gram intercropping system fetched higher net returns (Rs 24289) as well as higher B: C ratio (2.32) over sole pigeonpea due to more combined yield.

In order to prepare ourselves for greater adaptation to climate related problems, we must understand the impact of changing climate on basic food crops. Maize (*Zea mays* L.) is one of the most important cereals of the world and provides more human food than any other cereal. It is an important food and fodder crop of the world in general along with rice and wheat. Maize is widely grown in number of states in India as a *kharif* crop and in some state in *rabi* season as well. The study reported in this thesis, uses field experiments, and simulation models to understand the impact of changing climate on growth and yield of maize plant. In field environment, growth and yield of maize was greatly affected by temperature changes associated with sowing dates. Yield was reduced in late sown crops due to the harmful effect of chilling temperature. Very low minimum temperature experienced by the crop during panicle initiation stage led to death of all maize plants sown on first week of October. Application of irrigation water had positive effect on maize crop growth. Biomass and grain yield as well as other yield attributes were found to be higher in irrigated treatments than the rainfed one. InfoCrop model satisfactorily simulated crop phenology, leaf area index, dry matter production and yield of maize in Delhi region. Characterization of inter-seasonal climatic variability through growth and yield response in different agro-climatic zones can be successfully carried out with InfoCrop model. Sensitivity analysis of maize yield to temperature rise showed reduction in yield with temperature increase during the *kharif* season with percentage reduction more in Patna than in Delhi. In *Rabi* crop the model prediction for the future temperature increase initially showed a positive response up to 2°C.

Adaptation to climatic stresses

India is the second largest producer of wheat (*Triticum aestivum* L.) in the world. High temperature spell towards the end of the crop season i.e., terminal stage is a major determinant of wheat yield, especially under Indian conditions. In this study, field experiments and simulation models were used to understand the impacts of changing climate, particularly increase in temperature on growth and yield of wheat and develop adaptation strategies in the north-west India, the main wheat growing-belt in the country. A field experiment was conducted during *rabi* (November to April) season of 2011-12 and 2012-13 on the farm of ICAR-Indian Agricultural Research Institute, New Delhi to study the response of wheat varieties to terminal heat stress and to identify adaptation options such as identifying suitable varieties, change in sowing date and précised irrigation scheduling for alleviating terminal heat stress. The experiment consisted of (1) three dates of sowing i.e., normal date of sowing during 2nd week of November (D1), late sowing during first week of December (D2) and very late sowing during last week of December (D3); (2) three varieties of wheat i.e., HD 2932, WR 544 and HD 2967and (3) three irrigation schedules i.e., forecast-based irrigation (I1), conventional farmers practice (I2) and light and frequent irrigations (I3) in a split-split plot design. Observations for physiological parameters were taken at tillering, flowering and milking stages of the crop. It was observed that late sowing of wheat induced high temperature stress leading to reduction in membrane stability index (MSI), chlorophyll content, photosynthetic rate, stomatal conductance and canopy temperature depression (CTD) in all the varieties but the impacts were low with variety WR 544. The crop sown at normal date of sowing (D1) showed significantly higher number of tillers m^{-2} (444), plant height (93.2cm),1000-grain weight (37.9 g), grain yield (5.45 t ha^{-1}) and harvest index (0.38) compared to late and very late sown conditions. With delay of sowing, yield loss was about 53, 12 and 62 kg $ha^{-1}day^{-1}$ in varieties HD 2932, WR 544 and HD 2967, respectively. The late-sown crop encountered higher temperature in its terminal stages i.e., flowering, grain filling and maturity resulting in lower yield. Variety WR 544, however, showed less decline in growth and yield parameters compared to other varieties signifying its thermo-tolerant characteristics. Because of well-distributed rainfall in Delhi in the years of study (2011-12 and 2012-13), the irrigation treatments did not make much difference in the growth and yield of crop. InfoCrop-wheat model was used to analyze the impacts of future climate change scenarios. Simulated yield of wheat in terminal heat stress year was less compared to normal year. In terminal heat stress year, yield of wheat can be enhanced using adaptation options such as applying additional doses of

nitrogen fertilizer, preponing date of sowing from mid-November to first week of November and providing an additional irrigation especially at grain filling stage. The best adaptation option for alleviating terminal heat stress in wheat is to use tolerant variety, preponing date of sowing to first week of November with application of 30 kg additional N ha^{-1} and forecast-based irrigation.

Studies on Greenhouse Gas Emission

The Indo-Gangetic Plain (IGP) of India is dominated by rice-wheat cropping system (RWCS) and are crucial for food security in India. However, conventional practice of cultivating rice and wheat in these areas is tillage, water and energy intensive and a potential source of greenhouse gas (GHG) emission. In the present study an attempt has been made to identify low carbon options with high economic benefit for RWCS of the IGP. Initially a survey was conducted of 332 rice-wheat growing farmers in two contrasting regions of the IGP i.e., Trans-IGP (Haryana) and Middle-IGP (Bihar) and data on management practices, inputs and outputs etc. were collected and were used as inputs in the InfoRCT model to simulate GHG emission and global warming potential (GWP) of the two contrasting regions. A field experiment at IARI, New Delhi was also conducted to identify suitable low carbon RWCS for the IGP. Total six RWCS treatments derived from combination of conventional tillage wheat (CTW), zero tillage wheat (ZTW), transplanted puddled rice (TPR), dry Direct seeded rice (DSR), intermittent wetting and drying in TPR (IWD), application of Neem oil coated urea (NOCU) in ZTW and TPR and surface application of rice residue on ZTW were experimented for two consecutive year during 2011-2013 in the alluvial soil of the Indian IGP. The surveyed regions of the IGP significantly differed in nitrogen, water and tillage inputs resulting in differences in emission of CH_4 and N_2O from soil, and CO_2 from farm operation. The simulated average GWP of conventional RWCS in Haryana was significantly higher (2.5 times) than that of Bihar and it ranged from 4268±179 kg CO_2 eq. ha^{-1} in Middle-IGP (Bihar) and 10605±680 kg CO_2 eq. ha^{-1} in the Trans-IGP (Haryana). However, with use of resource conserving technologies (RCTs) such as system of rice intensification (SRI), DSR and ZTW, there was lowering in GWP than conventional puddle transplanted rice and tilled wheat. The continuous flooding in rice, use of electric pump for irrigation and application of high amount of nitrogenous fertilizer were identified as main contributors of GWP. In experimented rice and wheat treatments N_2O emission under ZTW was higher than CTW while application of NOCU and rice residue in ZTW significantly reduced N_2O emission. DSR and IWD significantly reduced CH_4 (> 80% and > 28% respectively) emission however also led to significantly higher N_2O emission (>60%) as compared to TPR. GWP $_{(CH4+N2O)}$ of experimented RWCS treatments ranged from 944 to 1891 kg CO_2 eq. ha^{-1} and 1167 to 2233 kg CO_2 eq. ha^{-1} respectively during first and second year of experiment. Among different RWCS, WR4 (ZTW-DSR) and WR5 (ZTW+RR-DSR) showed lowest GWP with highest B:C ratio while WR1 (CTW-TPR) showed lowest. Result indicated that adoption of DSR followed by ZTW (WR4 and WR5) in IGP in place of conventionally cultivated RWCS can reduce GWP by 55 percent without any penalty on B: C ratio.

Table 1: Effect of neem oil coated urea and zero tillage on the Global Warming Potential of transplanted and direct sown rice

Treatments	GWP (kg CO_2 ha^{-1})		
	CH_4	N_2O	Total
TPR-CTW	1618	320	1938
TPR-ZTW	1487	309	1797
TPR+NOCU - ZTW+NOCU	1408	265	1674
IF - ZTW	1143	443	1586
DSR-ZTW	409	494	903
DSR-ZTW+RR	419	467	886

A field experiment was carried out at the farms of Indian Agricultural Research Institute, New Delhi to quantify the impact of different levels of N fertilizer application and elevated carbon dioxide concentration (500±50 ppm) on nitrous oxide and carbon dioxide emission from soils in maize. The DNDC simulation model was also used to simulate the emission of nitrous oxide and carbon dioxide from maize. The nitrous oxide emission ranged from 181 µg m^{-2} d^{-1} to 1828 µg m^{-2} d^{-1} within the crop growth period. The cumulative emissions of N_2O-N ranged from 521 to 817 g ha^{-1}during the crop growth period and were 18% higher in 160 kg N ha^{-1} treatment over the 120 kg N ha^{-1}. The flux of CO_2C fluctuated between 3.10 kg ha^{-1} d^{-1} to 13.59 kg $ha^{-1}d^{-1}$ during the total crop growing period. Highest cumulative CO_2–C emissions were observed in 160 kg N ha^{-1} treatment at 840 kgCO_2-C ha^{-1}. The cumulative N_2O-N emission was 13% lower under ambient CO_2 as compared to the elevated CO_2. There was no significant increase in maize yield under elevated carbon dioxide. Harvest index under elevated CO_2 was 2.7% lower over ambient control. There was significant increase in carbon efficiency ratio (C fixed/C emitted) in the 160 kg N ha^{-1} treatment. The cumulative N_2O-N emissions simulated using the DNDC models were in good agreement with the observed values, though the temporal values showed variability. The model simulated much higher carbon dioxide emissions as compared to the observed values.

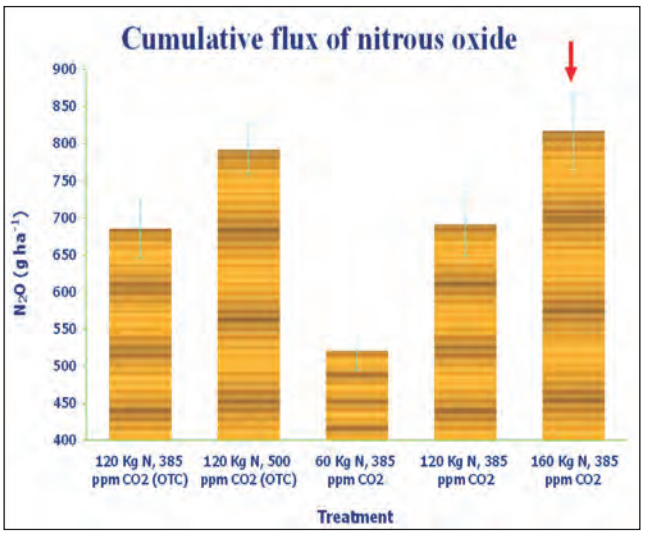

Fig. 9: Cumulative emissions of nitrous oxide in different nitrogen and CO_2 conditions

Vegetable fields are characterized by intensive production and high N application rates controlling nitrous oxide (N_2O) emissions. Thus Potato production can be the major contributor of Greenhouse gases (GHGs) from the crop production sector. Greenhouse gas emissions in crop production, processing and distribution arise from a variety of processes and sources. Therefore, it is important to quantify the GHG emission from the life cycle of potato to determine an energy-effective production and consumption pattern and evaluate the mitigation options to reduce GHGs emissions. A field experiment was conducted growing potato with seven treatments namely recommended dose of fertiliser with N application through urea (2 split), neem oil coated urea (2 split) (NOCU), Urea (3 splits), NOCU (3 splits), integrated nutrient management with 25% N through FYM and 75% N through urea and 25% N through FYM and 75% N through NOCU. Environmental impact of potato cultivation, processing and transportation for distribution to super markets/consumer in terms of greenhouse gas emissions was evaluated using Life Cycle Assessment (LCA) methodology. The selected sites for the study were states of Bihar, Delhi and Punjab. Average seasonal emission of N_2O ranged from 0.36 kg ha^{-1} to 2.11 kg ha^{-1}. The seasonal integrated emissions of CO_2 varied from 1461 kg CO_2 ha^{-1} to 3220 kg CO_2 ha^{-1}. Substitution of 25% of inorganic fertilizer by FYM increased N_2O emission by 8.5%, and CO_2 emission by 29% as compared to urea alone. Application of urea N in three splits reduced the emission of N_2O by 14.2% and

CO_2 by 7% compared to N application in 2 splits. NOCU decreased the N_2O emission by 9.87 -22.6% and CO_2 emission by 7-13%. Application of NOCU and three split application of N lowered the global warming potential (GWP) as compared to the urea treatment. Application of N through in three splits and NOCU increased the yield by 9.7-10.4% and 14.67 – 15.3% respectively as compared to urea N application in two split. Production of 1t of potato chips emits 1.85 t CO_2 eq. t^{-1}. Processing contributes 73% to the total GHG emissions followed by cultivation (24%) and transportation (3%) for production of 1t of potato chips. Soil emissions and farm operation together contribute 55% out of the total GHG emission from potato cultivation. Of the three sites highest emission from potato cultivation is from Punjab and lowest from Bihar. The study revealed that application of N through NOCU in three splits has the potential to reduce the GWP of potato cultivation. A higher emission load from the processing of potatoes indicated the need to reduce the consumption of processed potatoes products.

The amount of residues generated by biogas production has increased dramatically due to worldwide interest in using renewable energy. Biogas slurry (BGS) originate from anaerobic degradation of organic wastes has been proposed as organic fertilisers because of their high content of N, P, K and other valuable macro- and micro-nutrients. The effects of BGS on crop production and on the soil ecosystem and environment urgently need to be investigated before their wider use. Therefore, an on-farm approach was used to investigate impacts of different doses of biogas slurry application to compare with conventional and recommended doses of fertilizer application, in wheat growing field with respect to their (1) ability to provide plants with necessary nutrients for better yield, (2) effects on emissions of the greenhouse gas nitrous oxide (N_2O) and (3) impact on the soil microbial ecosystem. Results from the present study showed that the application of biogas slurry as a nitrogen fertilizer stimulated the growth of wheat yield and biomass. N_2O flux from the treatments showed more or less similar temporal trends with appearance of a peak of N_2O emission three days after urea applications, however, the magnitude of flux differed. The Biolog EcoPlate assay is a robust tool and was sensitive to changes in the short term due to management practices. Overall, a trend is noticed that, C-cycling enzyme potential activities increased with inorganic N availability while those of N-cycling enzymes increased with C availability. The soil microbial biomass, populations of bacteria, fungi and total count as well as enzyme activity increased significantly in the biogas slurry treated soil. In conclusion, as opposed to the conventional and synthetic fertilizer based agricultural practices, BGS cultivation system differ significantly in different aspects like, it reduces nutrient losses, improve soil fertility, and reduce global warming potential while supporting better crop yields in certain contexts.

Rice fields are one of the major sources of methane and nitrous oxide emission, two important greenhouse gases (GHGs) contributing 19% and 5%, respectively to the enhanced greenhouse effect. There is a need to find out options to mitigate global warming potential of continuously flooded rice field without reducing productivity. System of rice intensification (SRI), a new technique of rice cultivation, has potential for GHG mitigation. The GHG emission also varies with rice cultivars due to the difference in their aerenchyma and root exudates. A field experiment was conducted at the research farm of Indian Agricultural Research Institute (IARI), New Delhi in kharif (July to October), 2009 to assess the impact of SRI on GHG emission and growth and yield of rice as compared to conventional transplanted rice (TPR). The treatments consisted of two cultivars of rice i.e., basmati (Pusa 1121) and non-basmati (Pusa 44) under two methods of rice establishment i.e., conventional transplanting with continuous flooding and SRI with shallow irrigation to keep soil moist

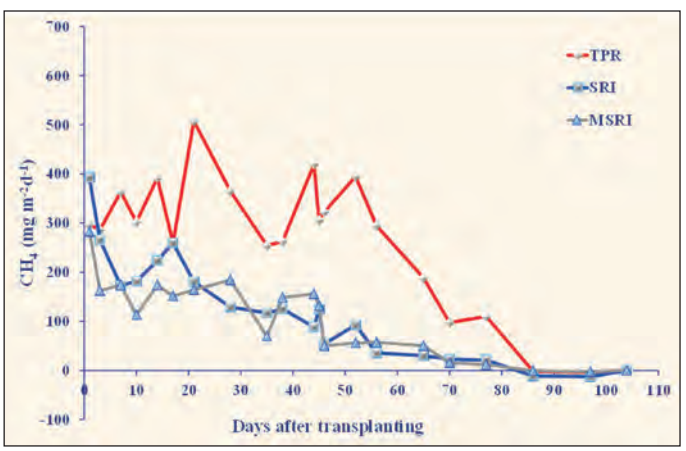

Fig. 11: Temporal emissions of methane from rice fields

and transplanting younger seedling. Under SRI method methane emission reduced by 62.5% but nitrous oxide emission increased by 22.5% over the conventional method (TPR). The global warming potential (GWP) in SRI reduced 28.9% over the conventional method. The SRI saved irrigation water upto 44% but there was no difference in grain yield as compared to the TPR. Methane and nitrous oxide emission from basmati cultivar (Pusa 1121) was 12.7% and 3.5% higher than the non-basmati cultivar (Pusa 44), respectively. The GWP in basmati rice was 7.5% higher than non-basmati rice. The study concludes that the SRI method can reduce GHG emission and saves water without any yield penalty.

Tropospheric ozone (O_3) is a phytotoxic air pollutant and a greenhouse gas and its oncentrations are increasing annually by 0.5±2%. Ozone has detrimental effect on plant productivity and chemistry, which might affect the soil C and N cycle and the emissions of N_2O and CH_4, the two important greenhouse gases from cropped soils. Therefore the present investigation was conducted to assess the impact of surface ozone concentrations on methane and nitrous oxide emissions from rice soil and the growth and yield parameters of rice. CH_4 and N_2O emissions were highest in charcoal filtered treatment and lowest in elevated ozone filtered treatment. Under elevated ozone treatment, cumulative seasonal CH_4 emissions were reduced by 29% over the non filtered control treatment while the emissions increased by 21% in the charcoal filtered treatment over the non filtered control. The cumulative seasonal N_2O emissions in the charcoal filtered treatment were 5% higher than the non filtered control and were 9% lower in the elevated ozone treatment as compared to the non-filtered control. Soil organic C did not change under different treatments whereas dissolved organic C was the lowest under elevated ozone. Grain yield was significantly impacted by different levels of ozone. It reduced by 11.31% under elevated ozone concentration. Filtration of ozone, which led to sub ambient ozone concentration significantly $_{(P=0.05)}$ increased the grain yield and other growth parameters as compared to the non filtered control. The Carbon Efficiency Ratio (CER) was found to be lowest in charcoal filtered treatment and highest in elevated ozone treatment.

Simulation Modeling

Vegetables are common food in human diet. India is the second largest producer of vegetables in the world, next to China. Vegetable crops are grown in 6 Mha of land forming about 3% of total cropped area. A planned development in vegetable production not only will improve the food supply to human being but can also meet the challenge of nutritional requirement for growing population in India. Leafy vegetables are important contributors among all vegetable crops. Nutritionists recommended that the per capita consumption of leafy vegetables should be at least 120 g per day, but only 45 g per person per day is consumed. Indian spinach, a leafy vegetable is highly sensitive to nitrogen and water stress. So, it is important to find optimal management for higher productivity. Simulations models are increasingly being used as decision support systems for agricultural management. But there is no model available for simulating growth, development and yield of Indian spinach. Thus, the present study was carried out with the specific objectives to i) quantify the growth and physiological parameters of Indian spinach under six management conditions and ii) calibrate the crop model to simulate the growth and yield of Indian spinach. To meet

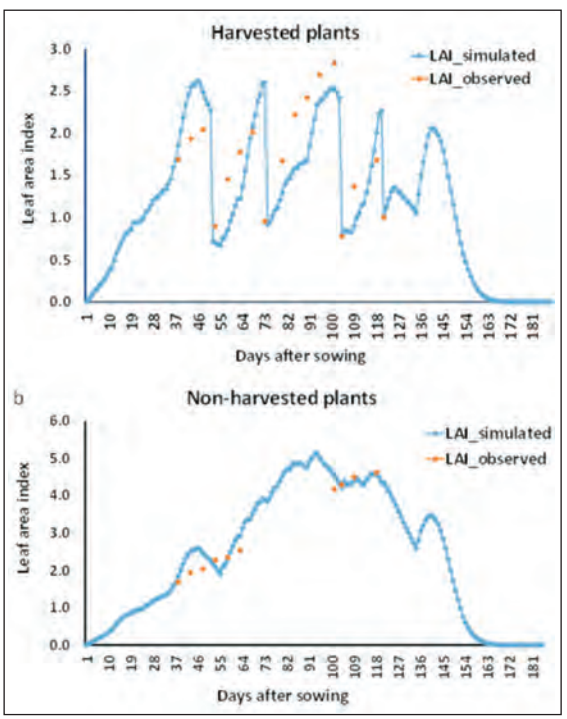

Fig. 12: Observed and simulated LAI in a) harvested and b) non-harvested plants

these objectives, firstly, a field experiment was conducted and the observed data on dry matter partitioning, leaf area index, gas exchange parameters, nitrogen concentration, and leaf and seed yield were collected at specific intervals. These data were used to calibrate and verify the spinach model. In the field experiment, conducted during winter season of 2015-16, Indian spinach variety 'All Green' was subjected to six treatments viz., 30 kg N ha^{-1} with irrigation at 10 days interval, 60 kg N ha^{-1} with irrigation at 10 days interval, 90 kg N ha^{-1} with irrigation at 10 days interval, 30 kg N ha^{-1} with irrigation at 20 days interval, 60 kg N ha^{-1} with irrigation at 20 days interval and 90 kg N ha^{-1} with irrigation at 20 days interval, imposed in four replications. A total of 24 plots with each plot size of $21m^2$ were maintained. Plants from 10 rows were harvested for leaf and rest 5 rows remained as such in each plot. Plants in both the harvested and non-harvested rows showed similar growth behaviour until the 1st harvest at 50 DAS. Harvests at 50 DAS, 71 DAS, 102 DAS and 119 DAS made significant differences in growth of harvested and non-harvested plants. Bolting and flowering occurred earlier in non-harvested plants. However, both plants attained physiological maturity at almost same time. Plants applied with nitrogen 60 kg ha^{-1} and irrigated at 10 days interval had highest TDM, leaf and seed yield as compared to other treatments. In this treatment, maintaining available soil moisture content above 15% (v/v) resulted in higher photosynthetic rate and water use efficiency. High or low dose of nitrogen led to reduction in TDM, leaf and seed yield. Nitrogen application at 90 kg ha^{-1} reduced the leaf yield irrespective of irrigation frequency. However, nitrogen concentration increased with increase in nitrogen dose. Leaves harvested at 50 DAS has least nitrogen concentration while it was more in leaves harvested at 102 and 119 DAS. Though non-harvested plants have more seed yield than harvested ones, differences in seed yield is not much hence, harvesting for leaf yield (3-4 times) and leaving for seed production is a beneficial option for spinach growers. Spinach model could simulate the Indian spinach response to irrigation frequency and nitrogen levels at various stages of growth in terms of phenology, leaf area index, dry matter accumulation and partitioning in to various plant parts, and leaf and seed yield with acceptable levels of MBE, RMSE and AI. The model could satisfactorily simulate the canopy growth behaviour in both harvested and non-harvested plants. This model can be used for application for different studies after more validation.

Table 2: Statistical indicators for evaluation of spinach model performance

Parameter	MBE	RMSE	AI
50% emergence (days)	0.67	0.82	0.92
50% flowering (days)	-0.33	1.00	0.95
Physiological maturity (days)	-0.67	1.00	0.96
TDM ($Mg.ha^{-1}$)	-0.31	1.11	0.99
Total harvested fresh leaf yield (Mg ha^{-1})	-3.09	3.85	0.99
Seed yield (kg ha^{-1})	-186	286	1.00
Total nitrogen in harvested leaf (kg ha^{-1})	21.90	24.7	0.99
Seed N (kg ha^{-1})	-1.20	2.12	0.99

Water scarcity is expected to be a major challenge for most of South Asia due to increased water demand and poor water management leading to water related food scarcity. In view of the projected increase in rainfall variability in future climates, better understanding of effect of dry spells and droughts varying in intensity and duration is essential for optimizing the time of supplemental irrigation under water scarce situation to minimize the yield loss in soybean, a major rainfed crop. Thus, the present study was carried out with the specific objectives i) to study the response of soybean to water deficit stress for identifying suitable agricultural drought index, ii) to simulate soybean growth and yield in response to moisture deficit stress, and iii) to simulate suitable water management strategy for maximizing yield under drought scenarios. A field experiment was conducted with soybean (variety PUSA 2614) subjected to three treatments of

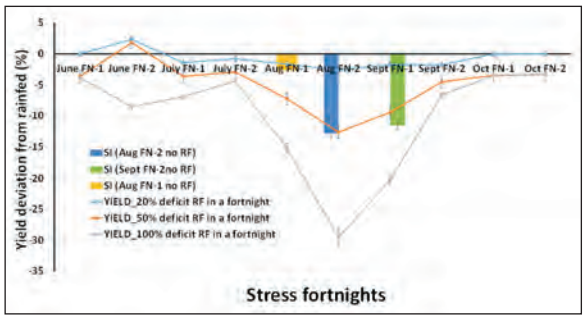

Fig. 13: Per cent deviation in seed yield in different water stress conditions and reduction in yield loss with supplemental irrigation

water stress using rain-out shelter at i) vegetative stage, ii) flowering to pod initiation phase, iii) entire pod filling phase till physiological maturity, in addition to iv) rainfed (since affected with whitefly, the treatment is re-designated as rainfed + biotic stress and v) supplemental irrigation (on 45 and 86 DAS). This experimental data were used to calculate the drought indices and to calibrate and verify the InfoCrop-soybean model before it is applied to simulate the optimal time of irrigation in different drought scenarios. Results indicated that among the treatments plants exposed to water stress during flowing and those exposed during vegetative growth phase had significantly low yield and total dry matter (TDM) as compared to other treatments. Supplemental irrigation led to significant increase in TDM and yield. Quantification of mid-season water stress was done by calculating the aridity anomaly index, moisture adequacy index and water requirement satisfaction index (WRSI). Among these three, WRSI is found to represent the soil moisture and soybean crop performance better. InfoCrop-SOYBEAN could simulate the plant response to water stress at various stages of growth with acceptable levels of MBE, RMSE and AI. This model was further used to simulate the impact of dry spells of different intensity and duration on growth yield of soybean crop. Thereafter the model was used to find optimal time of irrigation in 1620 drought scenarios for New Delhi region using 36 years weather data from 1978 to 2014. Results indicated that in Delhi region, a 20% reduction in rainfall from climatological mean during any fortnight of crop season does not affect the crop yield significantly. However, soybean yields are sensitive to water stress during 1st fortnight of August, 2 fortnight of August and 1st fortnight of September. Yield loss can be minimized with one irrigation during dry spell. From the above results, it can be concluded that, water stress during vegetative growth and flowering affected soybean yield significantly. Dry spells in August and early September significantly reduce soybean yield in Delhi region and supplemental irrigation, in the event of dry spell, can minimize the yield loss.

Appropriate location and operation of the developmental activities across different policy zones of NCR, to facilitate mutually productive "Centre - Periphery" relationship, requires detailed spatio-temporal mapping and long term change detection analysis of the regional resources of the study area. Scanning of literature revealed that this is acutely lacking for the study area. Hence, the present investigation was primarily aimed at a) quantifying the spatio-temporal pattern of the Land Use/ Land Cover Change during last two decades (i.e., between 1989 – 2006), (b) identifying major bio-physical and socio-economic factors governing agriculture production, and (c) assessing the impact of Land Use/ Land Cover Change and major bio-physical and economic factors on the agricultural production in the National Capital Region. In order to achieve these objectives three pairs of multi-temporal classified images for the *Rabi* (January-February) and *Kharif* (August-October) cropping seasons of 1989, 1998 and 2006 were generated through a hybrid unsupervised (ISO data)–supervised (Maximum Likelihood) image classification technique. The so classified images were subjected to a change detection analysis to identify the key bio-physical factors impacting agricultural production of National Capital Region. Other spatial and non-spatial data district-wise time series data of the study area were also acquired from different national data collecting agencies to support the aforementioned analysis.

Geo-spatial analysis of the above data showed that the study area experienced a steep (67.4%) increase in its croplands during 1989 to 1998 but relatively a small (5.7%) increase during 1998 to 2006 period. This was also associated with a similar steep (82.4%) increase in its built-up areas, due a 52.2% increase in its urban population, during the same period. The change detection analysis further showed that 1989-1998, associated with above normal to normal rainfall, was also associated with a steepest (77.9%) increase of the water bodies in the NCR. In contrast to this, due to the below normal total annual rainfalls, 1998-2006

was associated with about 49.5% water body level decline. This was also confirmed by the receding (pre and post-monsoon) ground water levels of NCR, during 1998-2004. It was further observed during 1989-1998 the extent of double cropped areas increased (by about 58.3%) and extended to even the uncultivated (18.6%) lands located within the extensive canal network of the study area. However, due to decreasing total annual rainfalls and increasing urbanization, both double cropped and *kharif* season (single) cropped areas suffered a decline of about 12.4 % and 9.6 %, respectively during 1998 - 2006. During this period about 38.1% of the double cropped areas transformed into the uncultivated lands most of which were centered on the highly urbanized/ industrialized areas in the Policy zones-1 and 2 of the study area. During last one decade the total degraded lands also increased, at a rate of 611.25 Km^2 per annum, from 5428.38 Km^2 (15.87% of NCR area) to 10,929.55 Km^2 (31.95% of NCR area). The impact of decreasing annual rainfalls, receding ground water levels, changing cropping pattern and the study area soil/ topography on the (source independent) available irrigation waters to meet the annual irrigation demand of the study area was also explored to show that the study area is irrigation starved and that this insufficiency is gradually increasing (28.48%), at an annual rate of 0.31 Km^3, over last one decade. The continuously increasing soil salinity/sodicity and declining annual rainfall, surface/ ground water levels and net irrigation water availability appeared to be the major contributing factors for the stagnating/ decreasing crop yields from year 2001 onwards. The decreasing crop yields were also associated with increasing cost of production and stagnating farm harvest prices thereby leading to a general decrease in the crop B/C ratios and hence the agricultural profitability of the study area. A two-decadal food grain security assessment and its optimistic and pessimistic projections for the future 2011 and 2021 years indicated a sharp decline in the study area food grain security since 2001. The analysis suggested a possibility of increased food grain security through a technological breakthrough on the degraded croplands. The study could thus lucidly quantify the major bio-physical factors governing agricultural business – profitability and assess their impact on the overall food grain security of the National Capital Region.

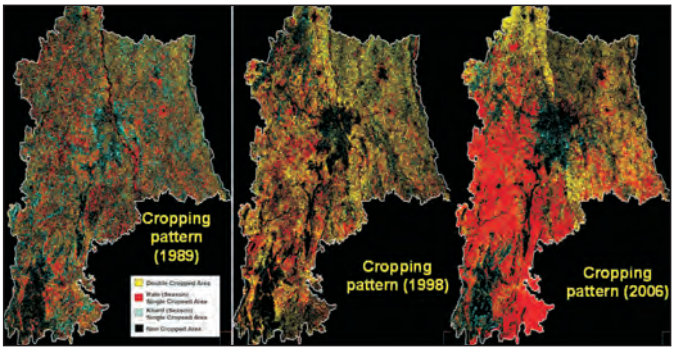

Fig. 14 : Geo-spatial analysis on change in cropping pattern in National Capital Region

Environmental Pollution

Farmers growing rice and wheat in rotation in the Indo-Gangetic Plains and wheat in central India use combine for harvesting their crops, which generates huge amount of residue in the field. Burning of crop residue leads to the emission of various toxic gases, greenhouse gases and particulate matter in the atmosphere. Therefore the present study was conducted to quantify the emission of gaseous air pollutants and particulate matter from burning of rice and wheat crop residues. A field survey was conducted to assess the methods used by farmers for harvesting of wheat (Hamirpur, Jalaun in Uttar Pradesh and Tikamgarh, Chhatarpur in Madhya Pradesh) and rice (Karnal, Kurukshetra in Haryana), amount of crop residues left in the field after harvesting and mode of crop residue disposal. Approximately 42-76% farmers used combines for harvesting, whereas 24-58% harvested manually. The use of combine was maximum in state of Haryana and minimum in Madhya Pradesh. About 60 to 74% of rice residue and 65-91% of wheat residues was burned in the fields after combine harvesting. The combustion efficiency ranged from 82.02 to 84.85%. The concentration of $PM_{2.5}$ and PM_{10} was 401.9 μgm^{-3} and 527.5 μgm^{-3} due to rice residue burning and 342.9 μgm^{-3} and 471.84 μgm^{-3} due to wheat crop residue burning. Organic carbon constituted the major fraction of $PM_{2.5}$. The SEM images of rice $PM_{2.5}$ showed the abundance of soot particles, carbon particles and silica. Whereas, wheat $PM_{2.5}$ showed more soot and carbon particles. Major elements in particulate matter of rice and wheat crop residues were carbon,

Parameters	Rice straw	Wheat straw
CO_2 (ppmv)	680	752
CH_4 (ppmv)	4.22	3.95
N_2O (ppbv)	443	429
CO (ppmv)	106	110
NO (ppbv)	35.6	40.47
NH_3(ppmv)	8.3	9.3
SO_2 (ppbv)	3.4	4.81
VOC (ppmv)	16.4	28.6
$PM_{2.5}$ (mg m^{-3})	401.9	342.9
PM_{10} (mg m^{-3})	527.5	471.8
TPM (mg m^{-3})	1003.8	904.11
Combustion efficiency	**82.02±1.35**	87.85±2.08

oxygen and silica which contributed 78.4% in rice and 84.7 % in wheat. The X-Ray Diffraction of rice and wheat straw ash indicated the presence of silica, potassium chloride, and potassium calcium phosphate. The study revealed that burning of crop residues deteriorated the air quality, which can have negative impacts on environment and human health.

India occupies about one-third of the world acreage under rice, of which basmati occupies a prime position. The country is the largest producer and exporter of basmati rice with about 2/3rd of the production is exported. The country exported 1.5 million ton of basmati rice during 2008-09 at a value of 2,060 million US$. Organically cultivated basmati rice is priced at higher values. Organic farming systems are now increasingly adopted as an alternative agriculture and solution to problems associated with chemicals fertilizers and pesticides. It has caught up because of increased consumer awareness regarding food safety, health issues and environmental concerns. Now, there is a need to increase food production and at same time ensuring quality of produce and environment. A study was carried out to evaluate the environmental impact of organic and conventional cultivations of basmati rice with respect to soil health, ground water quality and greenhouse gas (GHG) emission during *kharif* season (July to October) of 2009 at Kaithal district of Haryana. The study area comprised of 7 organic and 7 conventional fields spread over six villages in Kaithal, where organic farming is practiced for last 9 years. Yield and harvest index of rice cultivated under both the practices were similar. Organically cultivated fields had higher soil organic C, available N and dehydrogenase, acid and alkaline phosphatase activities compared to the conventional cultivation. Quality of water in terms of NO_3 content, sodium adsorption ratio (SAR) and residual sodium carbonate (RSC) were also better in organic fields. In conventional fields soil pH increased from 8.31 to 8.47 and electrical conductivity (EC) from 0.47 to 0.66 dS m^{-1} whereas in organic fields pH and EC decreased. Organic cultivation was better in terms of cost and net return. Estimated GHG emission, however, was higher in organic fields than conventional fields. Organically produced basmati had higher grain elongation, karnel length and karnel length and breadth ratio. Organic basmati exhibited high amylose, reduced crude protein, increased micronutrient (Fe, Mn, Zn and Cu) contents. Lower incidence of key insect pests and diseases with microbial and botanical pesticides in organic fields were recorded.

In excavated field organic carbon percent, SMB and Dehydrogenase activity is lower as compare to unexcavated field. Much of the organic matter content of soil lost along with soil used for brick manufacturing. Removal of

top soil from agriculture field for brick klin industry removes nutrients along with soil. In excavated field 15%, 19%, 26% and 21% of NPKS respectively are lower than Unexcavated field in brick kiln vicinity area. 11.5% of yield reduction is noticed in wheat crop and 13.5% reduction in mustard when it is grown in excavated soil. Every year 72.3 ha of land loses its top 3.5 ft soil in Gautam Budh Nagar district i.e., 1.17 lakh tonne of soil is permanently removed from agriculture land.

N is one of the most essential macronutrient for rice growth and production along with its improved grain quality. Farmers are applying hefty amount of fertilizers to increase crop productivity. Ammonia volatilization is the main process which contributes to nitrogen losses in rice crop. Ammonia emission causes environmental problems along with economic losses due to an increase in the cost of cultivation. Thus this study was conducted to evaluate the effect of high temperature on ammonia volatilization and yield under different N levels in rice crop. The four different temperatures *viz*, ambient temperature (T1) and three Temperature Gradient Tunnel temperatures namely T2, T3 and T4 and four different levels of nitrogen *viz*, 0, 90, 120 and 150 kg ha^{-1} in combinations were taken as the experimentation treatments. Plant height and leaf area significantly increased in TGT as compared to the ambient temperature whereas phenological length of rice crop was decreased. Photosynthetic rate increases as the nitrogen levels increases during all the growth stages and highest photosynthetic rate (14.1%) was observed at N120 compared to N0 at panicle initiation. Maximum photosynthetic rate was recorded during the panicle initiation (26.5 µ mole m^{-2} s^{-1} at N150) followed by grain filling stage (25.5 µ mole m^{-2} s^{-1} at N120). Stomatal conductance was found highest during panicle initiation stage (0.56 dsm^{-1} at N150) followed by grain filling (0.52 dsm^{-1} at N150) and tillering (0.48 dsm^{-1}at N150). Grain weight $plant^{-1}$, total grain weight, total biomass, number of tillers and number of panicles were found maximum in T3 at N120 treatment. The highest harvest index reaches up to 0.32 at N120 treatment T1. N concentration percent in leaves and stem was found highest at N150 whereas N content in grain was higher in N120 treatment. Chlorophyll content was found highest (12.7% increase) at panicle initiation stage in N120 as compared to N0.Urease activity was found maximum at panicle initiation followed by grain filling stage. The NH_3 volatilization was directly dependent on the fertilizer application and was emitted effectively upto the 4-5days from the date of fertilizer application as compared to N0. The rate of NH_3 volatilization increased with increasing temperature. It showed that maximum increase in ammonia volatilization (73%) was in N150 as compared to N0 treatment. The range of NH_3 emission was from 6.1-26.5 kg ha^{-1} $season^{-1}$.

Volatilization is the loss of nitrogen in the form of ammonia (NH_3) and it takes place by the conversion of nitrogenous fertilizer into ammonium gas through hydrolysis. Ammonia volatilization is the major process through which nitrogen losses from soil takes place under increasing temperature scenarios. Large amount of NH_3 emission not only causes environmental problems but also the nutrient losses which lead to economic losses by increasing the cost of production. Wheat is an extensively grown cereal crop in India but excessive use of N fertilizer in the agriculture leads to low nitrogen use efficiency and lower net returns to the farmers. Thus this study was conducted to evaluate the effect of high temperature on ammonia volatilization and yield under different N levels in wheat crop. The four different temperature *viz*. Ambient temperature (T1), Temperature Gradient Tunnel (TGT) Control (T2), plus 3° C (T3) and plus 5° C (T4) and four different levels of nitrogen *viz*. 0, 90, 120 and 150 kg ha^{-1} in combinations were taken as the experimental treatments. Results showed that phenological length was decreased whereas plant height and leaf area significantly increased in TGT as compared to the ambient temperature. As the nitrogen levels increases, photosynthetic rate was also increases during all the growth stages and highest photosynthetic rate was observed at N150 as compared to N120 at anthesis. Stomatal conductance was found maximum during anthesis stage (0.53 dsm^{-1} at N120) followed by grain filling and tillering, respectively and maximum photosynthetic rate was recorded during the anthesis (28.3 µ mole m^{-2} s^{-1} at N150) followed by grain filling stage (25.6 µ mole m^{-2} s^{-1} at N150). Grain weight $plant^{-1}$, total grain weight, total biomass, number of tiller and number of spike were found maximum in T4 with harvest index up to 0.45. In all the treatments, nitrogen levels enhanced the N content in leaves, stem and gain. Chlorophyll content was found highest (60% increase) at anthesis in N150 as compared to N120. Nitrate

reductase activity was found maximum at tillering followed by grain filling whereas glutamine synthetase was highest in N120 at T4 (28.5 µmol glutamylhydroxymate formed mg^{-1} protein h^{-1}) during anthesis. The NH_3 volatilization directly depends on the fertilizer application and was emitted effectively upto the 6th day from the date of fertilizer application. The rate of NH_3 volatilization enhanced with the increase dose of N application and it was observed maximum (72.9%) at N150 as compared to without N level. The ammonia volatilization ranged from 4.2-20.4 kg ha^{-1} $season^{-1}$ and it was highest (20.4 kg ha^{-1} $season^{-1}$) in 150 kg ha^{-1} N application at T4 having 13.6 % applied N loss.

Airborne particulate matter (PM) pollution is a very serious problem in India. Airborne particulate matter represents a complex mixture of organic and inorganic substances of varying sizes. Plants are variably sensitive to abiotic stresses and PM pollutants. Studies on response of growth and yield of crops from PM deposition are lacking, though literature on impact of PM pollution on trees is abundant. Therefore the present study was conducted to quantify the response of growth and yield of rice to PM deposition. The objectives were – (i) quantification of PM load in the air during rice crop growth period and its physicochemical characterization properties (ii) Quantification of growth and yield response of rice genotypes (PB1509 and PS-5) under varying levels of PM deposition. At the experimental site during rice crop growth period PM load in the atmosphere was observed in the range of 162.4 – 660.1 µg/m^3. PM deposition and flux on rice were in the range of 79.4 – 610 µgcm^{-2} respectively. On physico-chemical characterization of atmospheric PM , it was found that they are rich in dolomite, kaolinite, quartz, chlorite and calcite minerals. The contribution of the elements in PM load was in the following order C >Fe >N >Mn>Zn > Ca_2^+ >Cu >Ni >Pb> Na^+> K^+ > Mg^{2+} > SO_4^{2-} >Cr > PO_4^{3-} > NH_4^+ and Cl^- > NO_3^->Co >As and Cd > F^-**.** The enrichment factor indicates that the following elements are of anthropogenic origin C, N, Mn, Ni, Pb, Cd and rest are from natural(crust, sea spray etc) sources. The Air accumulation factor for elements on PS-5 and PB1509 was in the following order- NH_4^+>Cl^-> C > F^-> S > PO_4^{3-} > K^+> SO_4^{2-}> Mg_2^+> N> NO_3^-> Ca_2^+> As > Na^+ > Co > Pb> Fe > Zn > Cu> Mn > Cd. PM deposition adversely affected the growth and yield of both rice genotypes. Among the genotypes PS-5 was adversely affected than PB1509. Among growth and yield parameters radiation interception, canopy area, stomatal size, content of chlorophyll, carotenoids and relative water content, gaseous exchange (photosynthesis, stomatal conductance,) and yield were negatively correlated. Whereas, leaf temperature and transpiration were positively correlated. The reduction in yield in PS-5 was 14.9% and in PB1509 was 11.9%due to natural deposition of PM was observed as compared to control. A regional effect of PM on ecosystems is linked to climate change. Interactions of PM with other pollutants and with components of climate change remain important areas of research in assessment of challenges to agro ecosystem stability.

Environmental contamination by heavy metals is increasing enormously in recent years due to anthropogenic activities, soil being the main terrestrial sink for these pollutants. This is of primary concern in agricultural production due to adverse effects on crop growth and risk of crop and food chain contamination. Various physiological and biochemical processes in plants reported to be affected by heavy metals. Heavy metals, such as copper, cadmium, chromium lead, arsenic and mercury are major pollutants, particularly in areas with high population pressure. A pot experiment was conducted to study the mode of heavy metal contamination, their accumulation and distribution pattern in distinct vegetable crops; and the physiological and biochemical responses induced by these pollutants. Six crops belonging to three categories: leafy vegetables (mustard, spinach), root vegetables (radish, potato) and fruit vegetables (tomato, french bean) were grown on soils contaminated both separately with two levels of metals: nickel (50 and 100 mg kg^{-1}), chromium (50 and 100mg kg^{-1}), lead (100 and 200 mg kg^{-1}), cadmium (5 and 10 mg kg^{-1}) and mercury (25 and 50mg kg^{-1}); and also in combination following the completely randomized design. A set of crops grown on un-contaminated soil served as the control. The study revealed that metal contamination in vegetable crops was mainly contributed through root absorption and a small proportion of contamination was also registered through aerial deposition. Remarkable difference was observed in the accumulation, uptake and distribution pattern of metals between the crop species. Mustard was recorded to be the major accumulator

of Cr, Pb, Cd, Hg; while french bean followed by mustard showed greater accumulation of Ni. However, metal uptake was invariably found higher in mustard plants. The pattern of total metal accumulation in the vegetables followed the order: Pb>Ni>Cr>Cd>Hg in the current study. Variation in metal distribution pattern between different plant parts was also recorded among the crop species. Mustard accumulated more metals in stem, radish in leaves, potato into tubers, tomato in shoot, spinach into leaves and french bean in pods; but in the case of mercury higher accumulation was registered in leaves of most of the crops. The high levels of metal accumulation in plants led to significant reduction in growth and yield of crops, depending upon the level of metal concentration. Physiological responses to heavy metal stress in photosynthetic activity, chlorophyll *a* and *b* contents etc. were also recorded in crops. All parameters showed decrease with increase in metal concentration and responses varied with the type of metal as well. Hg and its combination treatments showed greater adverse effects than Ni, Cr, Pb and Cd in all vegetable crops. In general, heavy metal stress enhanced the accumulation of proline and the activity of anti-oxidant enzymes such as superoxide dismutase, catalase and peroxidase; whereas reduced the content of protein and nitrate reductase activity as compared to control plants. The higher production of proline and anti-oxidant enzymes under metal stress may be thought of as defense mechanisms protecting the crops from further damage by the heavy metals. The responses were observed to follow a concentration dependent pattern between two levels of metals. Among the metals applied, mercury was found to have greater adverse effect on all the crops tested under the investigation.

Food safety is a major public concern worldwide. During the last decades, the increasing demand of food safety has stimulated research regarding the risk associated with consumption of food stuffs contaminated by pesticides, heavy metals and/or toxins. The implication associated with heavy metal contamination is of great concern, particularly in agricultural production systems. These metals can pose a significant health risk to humans, particularly in elevated concentrations above the very low body requirements. Dietary exposure to heavy metals, namely Cadmium (Cd), Lead (Pb), Zinc (Zn), Copper (Cu) has been identified as a risk to human health through the consumption of vegetable crops. Heavy metals are given special attention throughout the globe due to their toxic and mutagenic effects even at very low concentration. The present study was conducted to assess the heavy metals contamination (Zn, Cu, Fe, Pb, Cd, Ni and Cr) in 21 different winter vegetables (Potato, Brinjal, Radish, Chilli, Tomatoes, Shimla Mirch, Onion, Garlic, Mustard, Cabbage, Cauliflower, Carrot, Coriander, Fenugreek, French Bean, Seem, Pea, Chenopodium, Spinach, Aroids, Beet root) and 7 summer (Sponge gourd, Bitter gourd, Cucumber, Kundru, Pointed gourd, Okra) vegetables crops from four different market sources (Okhla market, Inderpuri retail market, Azadpur and IARI) situated in Delhi. Lead contamination was found only in tomato and spinach of Okhla market and Inderpuri retail market samples respectively while remaining vegetables of both source and all vegetables of Azadpur and IARI were Pb free. Seasonal variation reveled that most of the winter vegetables were contaminated with one or more heavy metals while summer vegetables were only contaminated with Ni. Families of vegetables showed variation in accumulation of different heavy metals. Chenopodiaceae had highest mean concentration of Pb (1.54 ppm), Cr (48.50 ppm) and Ni (27.20 ppm) while umbelifereae had highest mean concentration of Fe (876.97 ppm) and Cu (24.62 ppm). Mean concentration of Cd, and Zn was highest in Crucifereae (6.25 ppm) and Solanceae (36.28 ppm) respectively. Among all the vegetables, mustered leaf showed high accumulation of most of the heavy metals mainly Zn (46.48 ppm), Fe (1005.08 ppm), Cu (22.23 ppm) and Cd (4.73 ppm). To evaluate the health risk index due to intake of vegetables of Potato, Onion, Brinjal, Radish, Chilli, Shimla Mirch, Coriander, Spinach, Okra, Sponge gourd, Bitter gourd, Cucumber and Tomato. The mean daily intake was found to be higher in spinach for Fe 0.567 mg/day/person, whereas the minimum intake of Nickel (0.004) was in cucumber. The health risk index of all the heavy metal was found to be less than 1 but in the case of Cr it is found to be more than 1.

Environmental contamination by heavy metals like lead (Pb), mercury (Hg), cadmium (Cd), arsenic (As), nickel (Ni), copper (Cu) and chromium (Cr) is a great threat to human health as these are non biodegradable and persistent. Keeping in view the gravity of such environmental problem, a pot experiment was undertaken

to study the physiological responses and general effect on growth and yield, metal accumulation and distribution pattern in two non-edible, commercial and industrial plants namely castor and cotton grown on heavy metal contaminated soil. Soils were contaminated all separately with two levels of metals: nickel (50 and 100 ppm), chromium (50 and 100 ppm), lead (50 and 100 ppm) and cadmium (50 and 100ppm) and also in combination of all metals at 50 and 100ppm in a completely randomized design experiment. A set of crops grown on soil with no metal amendment served as the control. Higher levels of metal treatments resulted in significant decrease in growth and yield of crops. Plant physiological responses in terms of chlorophyll a and b contents of leaves, photosynthetic activity and water-use efficiency recorded under the investigation showed marked reduction with increasing the level of heavy metal contamination in soil. The biological effects were metal dependent as all metals under study registered varying level of severity with nickel showing least effects compared to other metals. Combination treatments were noted to cause more adverse effects than treatments with single metals. Both crop species showed a remarkable difference in accumulation, uptake and distribution of these metals in their different plant parts. In general, the pattern of total metal accumulation in both crops followed the order Pb>Ni>Cr>Cd. The accumulation was also found to increase proportionately as the concentration of metals in soil increased. Metal uptake was crop specific whereby Ni and Pb uptake was higher in castor while Cr and Cd was higher in cotton. Metal distribution in castor plant parts followed the order Stem>Leaf>Root>Seedshell>Seed; while in cotton it followed the order Stem>bollshell>Leaf>Root>Seed>fibre. Distribution of almost all the metals was found to be maximum in vegetative parts than in reproductive parts for both crops. Within the reproductive parts, pericarp (seed shells and boll shells) recorded maximum distribution as compared to economic parts (seed and fibre). Castor and cotton are both suitable for Phyto-remediation of heavy metal polluted soil. Although both the plant species are hyper-accumulators, but castor could be recommended where Ni and Pb are dominant pollutants and Cotton for Cr and Cd contaminated soil.

Table 3: Emission of gases and particulate matter with burning of rice and wheat straw

Parameters	Rice straw	Wheat straw
CO_2 (ppmv)	680	752
CH_4 (ppmv)	4.22	3.95
N_2O (ppbv)	443	429
CO (ppmv)	106	110
NO (ppbv)	35.6	40.47
NH_3 (ppmv)	8.3	9.3
SO_2 (ppbv)	3.4	4.81
VOC (ppmv)	16.4	28.6
$PM_{2.5}$ (mg m^{-3})	401.9	342.9
PM_{10} (mg m^{-3})	527.5	471.8
TPM (mg m^{-3})	1003.8	904.11
Combustion efficiency	**82.02±1.35**	87.85±2.08

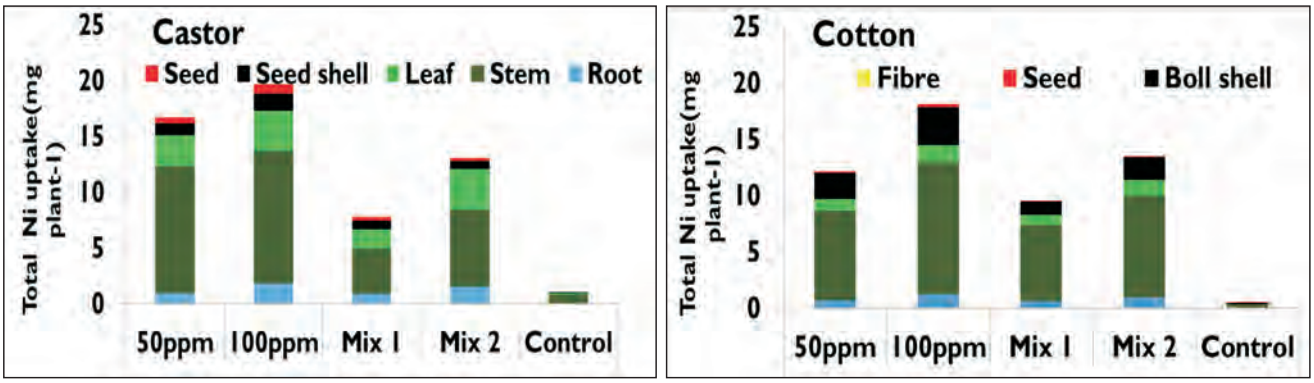

Fig. 15: Nickel extraction by castor and cotton plants and their distribution in plant parts

Environmental contamination by heavy metals is a serious problem throughout the world. Heavy metals are non bio-degradable and persistent and can be differentially toxic to microbes, plants, animals and humans. Contamination of soils by heavy metals such as lead (Pb) is considered being a risk to human health when its concentration exceeds 400–500 mg kg^{-1} soil. The toxicity of Pb contaminated soils can primarily occur from direct ingestion of vegetables and crops. The concentration of Pb in the soil solution is an important factor in all ecological considerations because the plants are expected to take up large amounts from it. Moreover, the mobility of Pb in soils and groundwater depends on its concentration. The fate and transportation of Pb^{2+} ions in the environment are generally controlled by adsorption and complexation reactions. Keeping in view the gravity of such environmental problem, a glasshouse pot culture experiment was conducted to evaluate the potential of various organic (biogas manure and vermicompost) and inorganic (rock phosphate and single super) to stabilize the Pb in contaminated soil. For Pb phyto-availability test, spinach (*Beta vulgaris L.*) crop was grown for 8 weeks. Results show that dry matter yield of spinach is significantly increased in amended soil as compared un-amended Pb contaminated soil. Lead concentration in shoot and root of spinach was significantly reduced in amended soil as compare to un-amended Pb contaminated soil. Adding single super phosphate (SSP), rock phosphate (RP), biogas manure (BGM), vermicompost (VC), and combination of rock phosphate with biogas manure (RP+BGM) and vermicompost (RP+VC) reduced the concentration of DTPA extractable Pb in the contaminated soil by 48%, 22.2%, 24.3%, 40.7%, 42.5 % and 47.1%, respectively. Sequential extraction of Pb in soil indicates that lead concentration in labile pool significantly reduced in amended soil as compare to un-amended soil. Lead content in the labile pool found in the order of Un-amended soil> RP> BGM>VC>RP+BGM>SSP>VC+RP. Therefore, rock-phosphate in combination with vermi-compost and single super phosphate were the most effective amendments in decreasing available Pb as indicated by the lowest phyto-available fraction (labile pool) in soils followed by vermicompost alone, biogas manure alone and rock phospahte. Further, DTPA-extractable Pb in rock phosphate amended showed a significant reduction in relative to the control even though RP contains water soluble P in negligible amount as compared to single super phosphate. In addition, rock phosphate is much cheaper than other single super phosphate and other water soluble P source. From the study it could be concluded that, metal stabilization assisted remediation has potential for success on a field scale reducing Pb entry in the food chain.

Pollution of groundwater due to industrial and municipal wastes is of a rising concern in many cities and industrial clusters in India. Although groundwater is not easily contaminated, once this occurs it is difficult to remediate. Hence it is very important to identify which aquifer systems and settings are most vulnerable to degradation. Detailed information on the farming practices, soils and surface/sub-surface waters of 34 agricultural sites around 30 villages in Faridabad block was generated through actual resource surveys, standard laboratory methods and GIS /remote sensing techniques. This information was used to estimate deep percolation loss and salt / heavy metal leaching under actual land-water management

practices, from each sample site by means of a field scale decision support system – IMPASSE followed by the assessment of the vulnerability of the study area ground waters to a particular pollutant under a specific land-water management practice. These vulnerability values were then compared with the actually observed pollutant concentrations in the ground waters to target the major (natural/ anthropogenic) sources of ground water contamination. The investigation showed that a majority of the study area was affected with soil salinity problem (i.e., EC mean = 4.79 ± 2.76 dS/m, Cl mean = 2043.30 ± 457.26 ppm and F mean = 11.57± 4.83 ppm). These salt affected agricultural lands were found to be 44 irrigated with (good to marginally alkali) canal, (marginally alkali to alkali) drain and (good to alkali) tube well waters. Due to this, the mean EC (2.85 ± 0.85 dS/m), Cl (1227.85 ± 295.93 ppm) and F (6.26 ± 2.12 ppm) concentrations in the deep percolating root zone leachates from the test sites were found to be far beyond their permissible limits of 0.75 dS/m, 1000 ppm and 1.5 ppm, respectively for ground waters. Even the soil root zone water soluble concentrations of some trace metals (viz., Cr: 0.06±0.00 ppm; Ni: 0.02±0.01 ppm, and Pb: 0.05±0.01 ppm) were observed to be of some threat to the deep percolating waters. However the vulnerability assessments indicated that due to the deeper water table depths (about 19.6 meters), the study area ground waters were not vulnerable to any salts/ trace metals in the root zone leachates. A comparison of these salt/ trace metal vulnerabilities with the actually observed ground water quality data for each test site further confirmed that the presence of salts in the study area ground waters was geogenic and that the on-going agricultural practices were not responsible for it.

Phycoremediation is the process of employing algae for removing excess nutrient load from wastewater and subsequently diminish the pollution load. It is an alternative technology of treating sewage wastewater compare to conventional treatment process in economical and sustainable way. Therefore, in the present investigation we had made an effort to phycoremediate the IARI's sewage wastewater with different microalgae *viz*. *Chlorella minutissima*, *Scendesmus spp* & BGA (*Nostoc*) and their consortium. Results showed that these algae were very effective in reduction of BOD_5, COD, NO_3^-, NH_4, $PO4_3$ and TDS in sewage wastewater. Further, it has been observed that *Chlorella* was having best phycoremediation potential as well as manure production among all three microalgae and even better than consortium. Among the potential uses of algal biomass from such systems is its use as a slow release fertilizer. After 20 days microalgae were harvested using muslin cloth and fresh and dry weigh were determined. The maximum biomass was observed in *Scendesmus spp* and *Chlorella minutissima* while percentage of nitrogen and phosphorus was highest in *Chlorella minutissima*. So we could conclude that *Chlorella minutissima* has the best manurial potential. The results of this study suggest that growing algae in nutrient-rich sewage wastewater offers a new option of applying algae to manage the nutrient load and after phycoremediation the biomass itself can be utilized for manure application in agriculture, serving the dual roles of nutrient reduction and valuable manure feedstock production.

Wastewater use for the irrigation purpose have been well documented but still there are loopholes in the study to monitor the effects of sewage water on environment and its suitability for agriculture. Hence, the present investigation was primarily aimed at determination of agriculturally significant physico-chemical parameters of IARI sewage wastewater and industrial wastewater from CETP, Mayapuri and compared their pollution strength, primarily in terms of BOD, COD, TDS, heavy metals and nutrients (N, P, and K). It has been found in the study that the treated wastewater from CETP, Mayapuri has very excellent prospective for irrigation as the most agriculturally significant physico-chemical parameters are under the general standards of FAO guidelines and indicate fairly out of harm's way for the use in irrigation. However, the sewage wastewater of IARI was not safe and sound and required further amelioration before its utilization in irrigation. It has been found from the analysis that the pH, TDS, EC and Pb were on higher side in the wastewater of IARI. Water use for the irrigation must have TDS less than 2000 ppm. Microalgae has been used in past to recycle some of the nutrient within the wastewater sources and also as a step in the wastewater treatment. The outcome from research had suggested the potential of specific

microalgae to reduce the BOD, COD and pollution load of the sewage wastewater of IARI and CETP. In the present investigation *Chlorella minutissima* was found to remove about 98-99% TDS, 70-80% nitrogen, 60-70% phosphorus and 45-50% potassium from the wastewater. The initial TDS value of sewage wastewater was in the range of about 1500 to 2300 ppm and after the phycoremediation by *Chlorella*, it is reduced to the range of 10 to 30 ppm. Hence *Chlorella* could be used for the remediation of sewage wastewater due to potential ability to reduce the TDS and pollution load and can further utilize for the safe irrigation of crops at IARI fields. Inferring from the study it could be safely said that the use of algae for treatment of wastewater can help in reducing pollution load on environmental sector and on the other side of coin recycle the wastewater for the agricultural and others use.

A small scale (batch flow) vertical sub-surface flow treatment wetland of 1570 Lpd capacity was developed for treating municipal wastewaters of IARI. System performance efficiencies were evaluated to develop appropriate design guidelines for treating municipal waters and for in turn augmenting deficit irrigation water supplies of Indian Agricultural Research Institute. The developed wetland system was observed to be having an overall volumetric efficiency of 84% and nutrient/metal removal efficiencies ranging between 40 to 90%. Inter-species differences were also investigated. Typha based systems were in general found to be having far superior nutrient and metal reduction efficiencies than the Pharagmites and Vachh based systems. However volumetric efficiencies were observed to be the highest for the Valchh based systems. For obtaining increased treatment efficiencies and reduced metal/salt retentions or leaching from the contaminated media, gravel (i.e. non reactive media) based wetland system with Typha plantations, was recommended. Ecological footprint and sustainability of the developed wastewater treatment system and a hypothetic sewage treatment plant (STP) were also compared using emergy analysis. Thus, lower cost and higher use of local resources in constructed wetlands make them attractive solutions especially for resource crunched developing countries.

Fig. 16: Treatment cells planted with *Typha latifolia, Phragmites karka, Acorus calamus*

A comparative impact of experimental subsurface flow constructed wetland treated and the raw (untreated) sewage water applications under Paddy-Wheat cropping sequence was evaluated on the historically sewage water contaminated experimental plots of Indian Agricultural Research Institute. Continuous application of treated sewage waters, for 2-years, in place of untreated sewage water applications (i.e. *business-as-usual*) at the project site resulted in significant reduction in soil potassium concentration from an initial (beyond permissible) level of (219.19 ± 12.66 mg/kg) to (125.27 ± 10.57 mg/kg). Soil nitrate and phosphate concentrations also decreased by about 88% and 38%, respectively thereby showing a significant impact of water treatment on soil nutrient load. Soil total and bio-available nickel (Ni), lead (Pb), chromium (Cr) and iron (Fe) concentrations also decreased significantly. However, continuous irrigation of treated sewage waters lead to no depletion of soil micro-nutrient concentrations as well as any adverse effects due to soil electrical conductivity and exchangeable sodium percentage, which remained within safe limits. Wheat and Paddy crops irrigated with treated sewage waters were observed to be associated with larger 1000 seed weights, lower number of unfilled unproductive tillers and the unfilled seeds per panicle and termite and fungal infected tillers. Wheat and paddy grains produced with *Phragmites karka* and *Acorus calamus* treated wastewater were observed to be sequestering significantly lower lead, nickel and cadmium, iron, respectively. Thus food grains produced through *Phragmites karka* treated wastewaters were observed to be associated with 44 to 58% less health hazards. Though, from health point of

view, the agricultural produce from the sewage plot sites was still not suitable for human consumption; especially due to considerable food grain metal viz., lead >> iron > nickel ~ Mn contamination. Yet these risks were far lower than those during previous year, primarily due to continuous application of wetland treated sewage waters and gradual reduction of soil bio-available and total metal concentrations on the historically sewage water irrigated soils. Thus, compared to the untreated sewage waters, the wetland treated sewage waters seemed to be associated with considerably lower environmental foot-print.

Sugar industries play an important role in the economic development of the Indian sub-continent, but the effluents released produce a high degree of organic pollution in both aquatic and terrestrial ecosystems. India has around 300 distilleries with a total installed capacity of 3250 million litres per annum and a current yearly production of 2300 million litres alcohol. Molasses, containing 8 per cent of sugar, serves as a cheap source of raw material for the production of alcohol. For the production of every litre of alcohol, nearly 14 to 15 litres of effluent is discharged, thus every day 1000's of litres of effluent are discharged from the factory. In distillery industry, spent wash (effluent) obtained from yeast fermented molasses after removal of rectified spirit. The RSW is highly acidic and contains easily oxidisable organic matter with very high BOD, COD. Also, spent wash contains highest content of organic nitrogen and nutrients. This effluent along with high organic matter and potassium contents has significant levels of nitrogen which are important ingredient to soil fertility. Along with easily biodegradable organic matter, it also contains plant growth promoters like gibberellic acid (GA) and indol acetic acid (IAA). A study was carried out to assess the long-term application of secondary treated effluent to soil and its consequent impact on nutrient build up and crop yield of wheat. The study area comprised of 14 farmers' fields (7 distilley effluent and 7 ground water irrigated fields) situated in Daurala in the district of Meerut, Uttar Pradesh, India, (Fig. 1) where a molasses-based distillery with the capacity of 300 KL alcohol per day is operating for the last 30 years. Seven farmers were chosen with fields close to each other (PMDE-amended), located near the distillery, where effluent was used as irrigation source for last 10 years at the rate of about 200 m^3 ha^{-1} as pre-sown irrigation. Another set of seven farmers were chosen whose fields were close to each other (unamended-control), located 1 km away from the distillery, where no effluent was applied and irrigation was provided only from groundwater. First the effluent was analysed for its constituents then the two set of fields were analysed for its nutrient content of primary, secondary and micronutrients, and the uptake pattern of micronutrients in wheat grown in the two set of fields. It was observed that Two to three times application of dilute secondary effluent as a source of presown irrigation and subsequent irrigation with good water on a long term basis will sustain the yield level of wheat; along with bringing down the cost of cultivation of farmer by enriching the soil with many nutrients. Dilute doses of effluent application for a long period will gradually build up the soil N, K, organic carbon, Ca, Mg, and with practically no increase in phosphorous content but act as an enhancer of soil micronutrients like Fe, Mn, Zn, which was ultimately reflected in nutrient enrichment of grain.

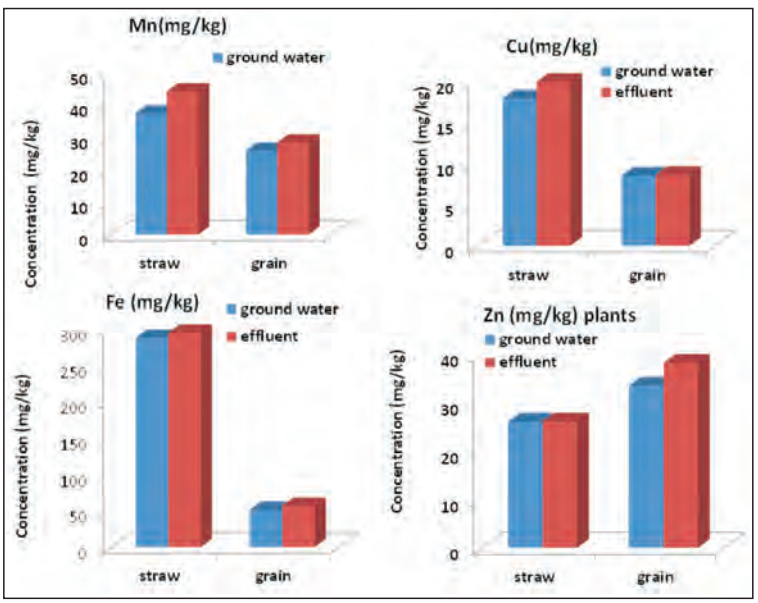

Fig. 17: Effect of industrial effluent on micronutrient content of wheat plant

The Mayapuri CETP of Delhi in Northern India, on an average treats about 4 million liter waste water per day. This waste water is received from Mayapuri industrial

area of Delhi as a mixture of industrial, domestic and commercial waste water. Present study investigated the agriculturally significant physico-chemical, biological and economical characteristics of the treated effluent for its use in agriculture. Samples of the treated effluent were collected from October 2009 to March 2010 at interval of nearly 15 days. The samples were analyzed for pH, EC_w, TDS, major ions (HCO_3^-, CO_3^{2-}, Cl^-, NO_3^-, SO_4^{2-}, Ca^{2+}, Mg^{2+}, Na^+ and K^+), nutrients (N, P and K) selected trace metals (Fe, Cu, Zn, Cd, Pb and Cr), BOD, COD and microbes (*Total coliform*, *Clostridium perfringens* and *Salmonella*). Various indices SAR, Na %, RSC, permeability index (PI), and US salinity diagram were utilized for better understanding of quality of water for irrigation. Cost of waste water treatment was calculated from of operational cost of CETP. Statically, the benefit of use of treated effluent in agriculture was calculated by replacing cost of commercial fertilizer and price for irrigation water. The results of pH, HCO_3^-, NO_3^-, SO_4^{2-}, RSC, Na %, PI, BOD, COD, microbes and trace elements (Cd, Cr, Pb, Zn, and Fe) of samples clearly indicated that this water was under good to slight to moderate quality for agricultural use. However the water quality of treated effluent was characterized by its very high salinity hazard (C4) to medium sodium hazard (S2) which can be considered as unsuitable for agricultural use. Conclusively this treated effluent cannot be used directly for agriculture however appropriate dilution with surface water can make it suitable for use. Result obtained indicated that this treated effluent can irrigate on an average 8 hectare of land per day and can supply 3.69-6.04 kg N, 0.49-1.05 kg P and 5.-8.45 kg K per hectare. Treated effluent can also supply 49.5-115.3 kg Ca, 7.47-35.42 kg Mg, 1.58 – 5.72 kg S, 0.145-0.185 kg Cu, 0.055-0.205 kg Zn and 0.87-2.1 kg Fe per hectare. The treatment of waste water cost Rs 4000 per 5, 00,000 litres of waste water while total benefit (price value for irrigation water + price value for nutrients) of use in agriculture ranged from Rs 335 to 438 per 5, 00,000 litres of treated effluent.

Table 4: Macronutrient content of treated effluent

Parameters	Minimum concentration (mg/L)	Maximum concentration (mg/L)	Average (mg/L)	Std. dev.
Available NO_3^--N	5.98	9.92	8.56	1.38
Total NH_4 - N	1.40	2.60	1.94	0.44
total N	7.38	12.52	10.50	1.84
Total recoverable PO_4-P	0.98	2.10	1.52	0.37
K	10.2	16.90	12.62	2.14
Ca	99	230.60	145.46	43.49
Mg	14.94	70.83	34.75	22.24
SO_4 -S	3.16	11.43	8.6	3.02

At present, a large amount of biogas slurry (BGS) is produced in India. Discharging them unprocessed will lead to environmental pollution, while disposing them as sewage will cost a great deal because of its large pollutant load. Moreover, replacing chemical fertilizers with biogas slurry can not only achieve resource

utilization of biogas slurry, but also reduce the amount of chemical fertilizer. So resource utilization of biogas slurry seems to be potential and necessary. Therefore, the present investigation was done to examine the effectiveness of biogas slurry as a nitrogen source for the production of baby corn and also to determine the suitability of soil for baby corn production. Results showed that 50% BGS along with 50% chemical fertilizer gave 20% more yield in terms of cob as well as biomass yield. Thus a farmer could save the 50% inorganic fertilizer and generate 20% additional revenue with application of biogas slurry. It has been found that combination of BGS and N fertilizer also augment the protein, total sugar, phosphorus and potassium content of the baby corn cob and increase the nutritional value of the produce. Experimental study has found the optimum application rate of BGS and nitrogenous fertilizer to achieve the best economic and biological benefits and to provide an allusion to the resource utilization of biogas slurry in agriculture.

Fig. 18: Design of biogas plant used

Energy-Rich Crops and Biofuels

The main objective of the present investigation was to assess the ecological efficiency of two bio-fuel energy crop plants, jatropha and castor in respect to their growth, biomass, yield, photosynthesis, water loss, nutrient and heavy metal removal under diverse abiotic environmental stresses such as low and high soil fertility, excess and deficit water, soil types, acidity, salinity, alkalinity, low radiation and high temperature so as to explore the possibility of cultivation of these crops on marginal and problematic soils and climatic stresses without competing with agricultural lands and loosing food crop production. The experimental findings revealed that both jatropha and castor showed differential growth, yield and physiological response to such diverse stresses, but castor plants exhibited greater reproductive susceptibility as compared to vegetative ones. Both plants species showed positive growth and yield response to higher level of nutrient fertilization and irrigation, while registered greater degree of ecological sensitivity (susceptibility) to salinity, alkalinity, and low radiation as compared to other stresses like acidity and, heavy metals and heat stresses. Jatropha plants exhibited greater tolerance to water deficit, while castor showed greater susceptibility to the same in respect of their bioproductivity. Some of the foliage characters like specific leaf area and leaf chlorophyll content were enhanced by low radiation, while photosynthetic rate was reduced drastically under low radiation but enhanced under high soil fertility and high temperature in both the plant species. However, other stresses show detrimental effect on the same traits. The phenological characters like days to flowering and fruiting in castor were delayed markedly under water deficit stress, soil acidity, low radiation-L1, while the same was least affected by other stresses. Both the plant species registered higher degree of susceptibility to excess water stress, where both plants species could not survive for a longer time, but relatively castor showed greater degree of sensitivity as compared to jatropha plant. Edaphic stress like salinity and alkalinity hastened temporary wilting compared to normal and other stresses. It was also observed that water loss per plant was highest under high thermal stress in jatropha and light texture soil in castor, while it was lowest by water deficit stress in both the crop plants. Nutrient (NPK) uptake was markedly affected by these stresses in both the plant species, but their uptake was observed to be higher under increased nutrient fertilization treatment (F2). These plant species proved to be heavy metal accumulator under metal contaminated soil. In both the plant species, ecological sensitivity pronounced with the level of stress imposed upon. It is interesting to note that leaf shedding in jatropha was observed to be controlled ecologically depending upon thermal regime during winter season. Thus it is clearly evident

from the findings that both the plant species could be grown successfully on marginal and problem soil under rainfed condition and could also be utilized for phytoremediation of heavy metal polluted soil.

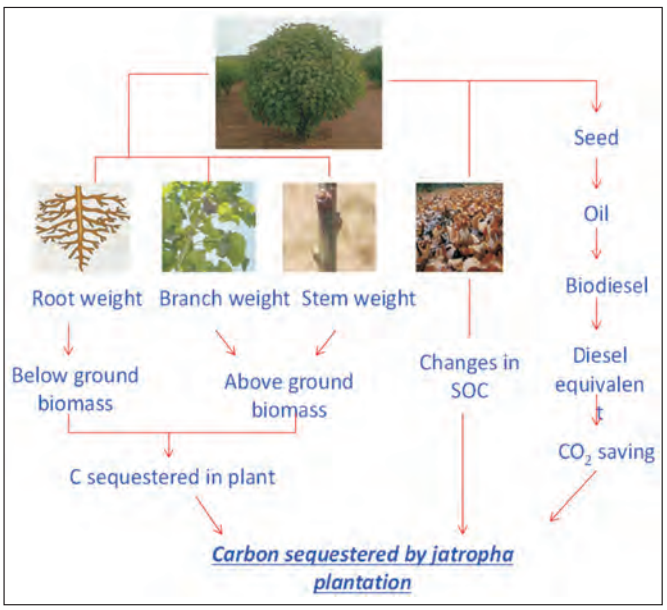

Fig. 19: Steps for calculating carbon sequestration potential of jatropha plantation

The present study was conducted to evaluate the growth, physiology, CO_2 sequestration, allelopathic potentials of jatropha plantation and their effects on soil physical, chemical and biological properties in high (HD), medium (MD) and low (LD) plant population densities. A nearby uncultivated area was taken as the reference site (control). Jatropha cultivation irrespective of plant population density improved soil organic carbon and availability of NPK on surface soil. Jatropha plants grow slowly during leafless period in winter season and faster during the period from April to November corresponding to leafy period and the growth in terms of plant height and stem girths was recorded to be higher in plants grown under low population density. In spite of lesser leaf chlorophyll content in low density plants, net photosynthesis was higher with better nutrient uptake. Nutrients translocated from senescing leaves to the stem during leafless period also enriched the soil surface with CO_2 and nutrients through leaf litter and fruits shedding and their decomposition. Addition of organic matter, retention of soil moisture and moderate soil temperature led to increase in soil microbial activities as supported by increased CO_2 flux and litter decomposition rate, microbial biomass C (MBC) and microbial biomass N (MBN), and urease and dehydrogenase activities. Allelopathic potentials of jatropha on wheat growth were limited to ex-situ bioassay. Biomass and grain yield increased in wheat when soil was amended with different parts of jatropha under pot experiment, and wheat crops flourished when integrated with jatropha in in-situ approach with pruned jatropha plants. However in in-situ approach wheat grain yield and biomass declined under the standing jatropha cultivation with lowest reduction in low plant population density plots mainly due to branches and twigs shading effects on wheat crop. Study concludes that irrespective of plant densities, jatropha could be a potential candidate to grow as a component of agro-forestry system under irrigated condition in North West parts of India and a plant population density of 1666 per hectare would serve the purpose of both agro-forestry and carbon sequestration apart from other ecological services.

The world is presently confronted with crisis of fossil fuel as well as environmental degradation due to increase in carbon dioxide (CO_2) levels in atmosphere. Carbon sequestration through agro forestry is an approach to reduce the CO_2 concentration store atmospheric CO_2 in vegetation, soils, and biomass products in the atmosphere. Potential of the *Jatropha curcas* L. (Euphorbiaceae) has received wide attention in India for production of biodiesel. The Government of India has plans to meet 20 per cent of the country's diesel requirements through jatropha plantation in an area of 11.2 million hectares. Therefore the present study was undertaken to assess the assess carbon sequestration potential of jatropha plantation. Decomposition of jatropha litter was estimated *in-situ* in two different seasons (wet & dry). Rate of litter decomposition during wet season was more at 30 cm of soil depth than 5 cm. During dry season, decomposition was more rapid initially upto 30 days and slow at the later phase. Maximum loss of all the nutrients from decomposing litter occurred during the month of August to October in both the experiments due to high rainfall received during this period. Laboratory experiment was carried out to study the contribution of

litter on sub surface soil to DOC and DON pool of soil with different N source on DOC and DON release. Ammonium nitrate was found to release more DOC and DON as compared to urea and ammonium sulphate. Percent of total litter carbon leached in the form of DOC ranged from 1.8- 2.6% and nitrogen released as DON was 24.7 - 54.7%. Carbon sequestration potential of jatropha plantation was calculated by estimating the C sequestered by plant, soil and amount of CO_2–C emission saved (426-648 kg ha^{-1}) by replacement of conventional diesel with biodiesel. Total carbon sequestered by jatropha plantation was 12.68 and 7.14 t ha^{-1} yr^{-1} for population of 2000 and 1100 plants ha^{-1} respectively. An empirical model "INFOJAT" was developed for determining carbon sequestration potential of jatropha plantation from the empirical equations and co-efficients generated from the above experiments.

Jatropha fruit coat and seed coat powders were evaluated for their adsorption ability towards Cd(II), Pb(II) Cr (III) and Cr (VI) ions from aqueous solution by using batch adsorption technique. The effective factors such as initial pH of aqueous solution contact time, initial metal ion concentration, and adsorbent dosage, and temperature, tec. were investigated upon this technique. Optimum conditions for metal ion adsorption were standardized using batch adsorption experiments. Langmur and Freundlich equilibrium isotherm models and Weber and Morris intra particle diffusion model were used. Kinetic models such as Pseudo-first order (Lagergren), Pseudo-second order and Elovich model were used to understand adsorption mechanism. Thermodynamics of adsorption was also studied. Physiochemical characterization including SEM and FTIR studies were done to understand the surface morphology and chemical property of the adsorbents. Scanning electron microscopy (SEM) showed amorphous and rough surface and small openings on the surface of raw adsorbent which can provide more contact area for adsorption. Fourier Transform Infrared (FTIR) analysis of Jatropha seed coat and fruit hull before and after adsorption showed prominence of hydroxyl and carboxyl groups on adsorbent surface. Jatropha seed coat powder performed better than Jatropha fruit coat powder in mental adsorption capacity. The monolayer sorption capacity ($Q_{m)}$ was found to be 26.53, 22.83, 37.45 and 75.76 mg g-1 respectively for pb (II), Cd (II), Cr(VI) and Cr(III) adsorption on seed coat and 25, 21.97, 37.59 and 24.69 mg g-1 respectively for pb (II), Cd (II), Cr(VI) and Cr(III) adsorption on fruit coat. Metal ion removal using Jatropha seed coat powder and Jatropha fruit coat powder was found to be dependent on contact time, sorbent dose, initial concentration, and pH. Freundlich isotherm provided the best fit for Cr(VI) and Cr (III) adsorption while Langmuir isotherm fitted well with Cd (II) and Pb (II) adsorption. Adsorption process is best described by Elovich kinetic model for both seed coat and fruit coat. The negative values of ΔG at all temperatures indicate spontaneous nature of the adsorption reaction. The positive value of ΔH indicates adsorption process is endothermic. Electrostatic attraction between the charged adsorbent surface and metal ions may be the major mechanism involved in metal ion adsorption. Present investigation shows that Jatropha fruit coat powder and Jatropha seed coat powder, as non-conventional low cost adsorbents, ha potential in metal adsorption.

The increased concern for energy security and the negative impact of fossil fuels on the environment has put pressure on society to find new renewable fuel alternatives. Ethanol produced from agri-residue, is an alternative to fossil fuels. Wheat is the world's most widely grown crop and second largest biomass feedstock in the world after rice. The production of fuel ethanol from low-cost biomass currently suffers from several problems. One of them is the presence of inhibitors in lignocellulosic hydrolysates that are released during pretreatment. Furfural and 5-Hydroxymethyl furfural are the most potent inhibitors to ethanol fermentation by yeast. The present study was done with the aim of quantifying furfural and 5-HMF during pretreatment of wheat straw (variety HD 2932) and its effect on growth and ethanol production by yeast *Pichia stipitis,* NCIM 3498. Pretreatment of wheat straw was carried out in microwave, using 1-4% (v/v) H_2SO_4 at different temperatures ranging from 100° to 220°C. Detoxification of pretreated hydrolysate was carried out by using 5% (w/v) activated charcoal. Pretreated, saccharified and detoxified hydrolysate was fermented by *Pichia stipitis* NCIM 3498. Characterization of the biomass on the basis of fibre, sugar content and other chemical constituents, showed wheat straw as a potential feedstock for ethanol production. The

furfural and 5-HMF production ranged from 0.51-39.17 ppm and 0.70-30.10 ppm, respectively. Higher amount of furfural and 5-HMF from pretreated wheat straw were observed at 180° to 220°C, using 4% (v/v) dilute sulfuric acid. Detoxification of hydrolysate using activated charcoal reduces 73.32 to 84.01% furfural and 28.53 to 76.42% 5-HMF in filtrate. Result showed that up to 30 mM there was no significant inhibition on growth and ethanol yield by *Pichia stipitis* NCIM 3498 on MGYP synthetic media. Fermentation and ethanol yield by *Pichia stipitis* from detoxified hydrolysate was found better as compared to non-detoxified microwave assisted acid pretreated wheat straw hydrolysates. Maximum ethanol yield obtained was 5.29 % (v/v), equivalent to 87.9 % of the theoretical yield. The ethanol yield coefficient was found 0.44 g ethanol g^{-1} sugar utilized.

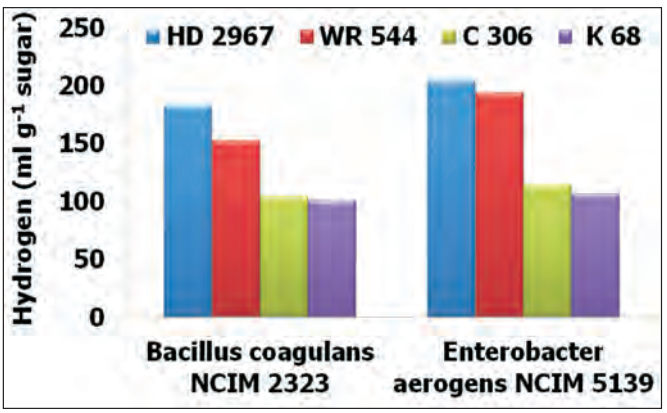

Fig. 20: Hydrogen production potential of wheat straw with different microbial strains

In the last decade, most research has tended to focus on developing an economical biofuels production process. Much emphasis is being given to the production of hydrogen from lignocellulosic biomass. Wheat is the world's most widely grown crop, about 21% of the world's food supply depends on this crop. Among the agricultural crop residues, wheat straw is the second largest biomass production in the world after rice straw. Wheat straw, one of the cheap lignocellulosic biomass, is composed of heterogeneous complex of carbohydrate polymers, especially cellulose and hemicelluloses, which can be readily hydrolyzed into fermentable sugars by pretreatment and enzymatic saccharification. Hydrogen producing microorganisms can convert these hydrolyzed sugars into hydrogen. In the present investigation, wheat varieties C 306, K 68, HD 2967 and WR 544 were collected from the experimental field of IARI, were analyzed and characterized on the basis of chemical constituents. Characterization of the wheat varieties on the basis of fibre, sugar content and other chemical constituents, showed it as a pote ntial feedstock for hydrogen production. The effectiveness of pretreatment using microwave assisted dilute acid to produce total soluble sugar from the straw was also investigated. The results showed that microwave assisted acid pretreatment is a promising approach to enhance fermentable sugar yields. The results showed that the microwave assisted acid pretreatment at 160°C for 10 min was found best for sugar recovery from wheat straw. The samples of microwave assisted acid pretreated straw were processed for enzymatic saccharification with cellulase and β-glycosidase enzymes. The total fermentable sugars from pretreated and saccharified straw were ranged from 46.38 to 67.49%. The saccharified hydrolysates were fermented to produce hydrogen by using promising microbial strains of *Bacillus* and *Enterobacter spp*. The hydrogen yield in terms of mol/mol glucose ranged from 0.23 to 1.49. A maximum hydrogen yield up to 205 mL/g of sugar was obtained from saccharified hydrolysates of wheat straw by *Enterobacter aerogens NCIM* 5139. Maximum hydrogen production potential was observed to be from the wheat varieties HD 2967, followed by WR 544. These findings highlight the potential of wheat straw for the production of hydrogen.

Ethanol is one of the most important renewable fuels that can be utilized for the reduction of negative environmental impacts generated by the worldwide utilization of fossil fuels. The lignocellulosic wastes represent an abundant and potential source of ethanol production. Rice is the third most important grain crop in the world behind wheat and maize in terms of production. According to FAO, India acquires significant position in the global scenario as it is the second largest producer of rice accounting for 26%. Rice straw, one of the cheapest lignocellulosic biomass, is composed of heterogeneous complex of carbohydrate polymers, especially cellulose and hemicelluloses, which can be readily hydrolyzed into fermentable sugars by pretreatment and enzymatic saccharification. Ethanol fermenting microorganisms can convert these hydrolyzed sugars into ethanol. In the present study, straw of rice varieties Pusa 44, PRH 10, IR 36, Pusa Basmati 1121 and Taraori

basmati, collected from the experimental field of IARI, were analyzed and characterized on the basis of chemical constituents. Samples were processed for autoclave and microwave assisted chemical pretreatment, subsequent enzymatic saccharification and ethanol fermentation. The kinetics of ethanol fermentation was carried out in Applikon fermentor. Characterization of the rice varieties on the basis of fibre, sugar content and other chemical constituents, showed it as a potential feedstock for ethanol production. The efficiency of autoclave and microwave assisted chemical pretreatment was compared. Sugar recovery was found maximum by microwave assisted alkali (NaOH) treatment, followed by acid (H2SO4) and minimum in hot water. The results showed microwave assisted alkali pretreatment to be a promising approach to enhance sugar yields. Enzymatic saccharification of pretreated straw gave sugar ranging from 9 to 39%. Taraori Basmati had higher sugar recovery (69.9%), followed by Pusa Basmati 1121 (58.9%), after saccharification and microwave assisted alkali pretreatment. Fermentation of saccharified hydrolysates using *Pichia stipitis* NCIM 3498, gave ethanol yields of 2.1 to 5.16%(v/v), maximum from the rice varieties Taraori basmati, followed by Pusa basmati 1121. The kinetic study of ethanol fermentation from Taraori basmati in fermentor gave 6.63% (v/v) ethanol, equivalent to 92.9% of the theoretical yield. The ethanol yield coefficient was 0.47 g ethanol g-1sugar utilized. The potential of ethanol production from the rice varieties, according to the study, ranged from 141 to 360 L per ton of microwave assisted-NaOH pretreated straw. These findings highlight the enormous potential of rice straw for the production of ethanol.

Corn stover in India with an average productivity of 2414 kg/ha is one of the most promising feedstocks for fuel ethanol production. The present investigation was carried out to maximize fermentable sugars (glucose and xylose) yield for ethanol production by modified pretreatment and subsequent enzymatic hydrolysis of corn stover. The corn stover as a feedstock was pretreatment with hot water for 60 minutes @ 10% total solid loading followed by hydrochloric acid (1%) treatment for 60 minutes, further subjected to trichoderma reesei 1052 treatment and cellulose bydrolysis subsequently for 72h found to be the efficient modified pretreatment for maximum sugar recovery of 92.49% of total fermentable sugar. The xylose and arabinose recovery by modified hot water and hydrochloric acid pretreatment was 82.7, 75.0 percent, respectively. There is an increase in cellulosic conversion from 67.54 to 92.49 per cent after Trichoderma reesei 1052 treatment and enzymatic hydrolysis in the modified pretreatment . The alkali had a comparatively higher percentage increase in sugar recovery than acids among all the modified pretreatments after enzymatic hydrolysis. Pichia stipities NCIM 3499 was the most suitable and efficient fermentative yeast strain over twenty one yeast strains screened for ethanol production from corn stover hydrolysate with 21.6g/1 ethanol concentration, ethanol yield of 0.43g ethanol/g of sugar4 consumed and fermentation efficiency of 86% of theoretical yield. Saccharomyces cerevisiae NCIM 3061 was capable of producing 15.4g/1 ethanol concentration with an ethanol yield of 0.46 g of ethanol/g of hexose and fermentation efficiency of 92 per cent of theoretical yield among Saccharomyces cerevisiae strains. The ethanol from corn stover, being a cheap feedstock compared to molasses, could be economically feasible with the modified pretreatment technology. The biogas and recovery of higher value lignin as co products can reduce the cost of production of cellulosic ethanol from corn stover.

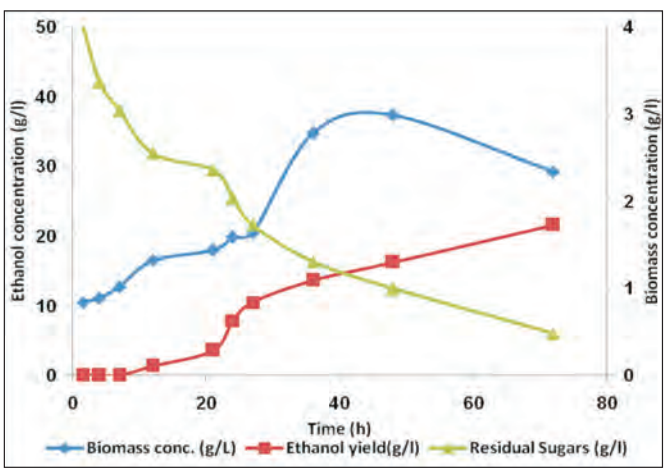

Fig. 21: Fermentation of corn stover hydrolysate from modified pretreatment by *Pichia stipitis* NCIM 3499 on Aplikon bioreactor

Cellulases enzymes currently account for approx. 20% of the world's enzyme market. Till now, the emphasis has been given on fungal cellulases because of large amount of less complex extracellular cellulases, however, recently the shift has been towards the bacterial cellulases, owing of robust bacterial growth and survival in harsh conditions of bioconversion

processes. Six thermophilic cellulases producing bacteria have been isolated from Water samples collected from the hot spring Agnikunda at Bakreshwar, in the West Bengal and also from Vajreshwari, Maharastra in India. From the former three alkaline cellulase producing *Bacillus licheniformis*, *Aneurinibacillus thermoaerophilus* and *Bacillus* sp. have been isolated. For maximum cellulases production, the optimum temperature and incubation period was found to be 60-65^{0}C and 60h respectively. Unlike Barkeshwar hot spring, three acidophilic cellulolytic thermophilic bacteria viz. *Bacillus licheniformis* and two *Bacillus* sp. have been isolated from the Vajreshwari hot spring which showed optimal enzymatic activity at pH 6.0-6.5. Maximum cellulases production was detected after 60h incubation period in wheat and rice straw at 50-55^{0}C.

Biogas is a clean and one of the potent renewable forms of energy which could very well substitute conventional sources of energy (fossil fuels, oil, etc.) especially in the rural sector. The later sources of energy are causing ecological-environmental problems and at the same time depleting at a faster rate. So, there is an urgency to undertake research on biogas generating potential of the biodegradable matters such as agro wastes. The present study dealt with the biogas potentiality of Castor (*Ricinus communis*) alone and in combination with cattle dung in various proportions at 15 percent total solid by anaerobic digestion process as well as evaluated the manurial potential of digested biogas slurry (DBS). Five treatments including admixture in different ratio of CD and COSC, T_1 (CD alone), T_2 (1:2), T_3 (2:1), T_4 (1:1) and T_5 (COSC alone) were taken for experimentation to record the production of biogas and manurial quality of corresponding digested slurry. Treatment T_2 (CD: COSC in 2:1 ratio) generated 284.55 L/kg dm. All the treatments except treatment T_1 have shown the higher biogas production potential than COSC alone. Treatment T_2, T_3 and T_4 produced 335, 285 and 219 L/kg dm of biogas respectively as compared to 145.20 L biogas/kg dm of cattle dung alone (T_1). The result showed that among the treatments admixed with CD and COSC (T_2, T_3 and T_4), the treatment T_2 showed highest content of N (3.6%), P (1.5%) and K (1.05%), while T3 (CD:COSC=2:1) showed lowest content of NPK (2.65, 1.05 and 0.89%) respectively in the manures. The NPK content of all treatment admixed with CW and COSC were higher than that of treatment having CD alone. The comparison with different manure indicated that the N and P content of DBS was higher than that of FYM, rural compost, urban compost, MSW compost and sewage sludge. The N and K content in DBS of CD and COSC admixture as compared to biogas spent sludge (BSS) of another agro material like Jatropha fruit coat and cattle dung admixture was lower by 11-15% and 48-62% respectively, while P content was recorded to be higher by 36-40%.

Summary

- Climate change associated with rise in concentration of greenhouse gases (GHGs) is likely to affect crop production. Elevated temperature and carbon dioxide (CO_2) can affect growth and production of rice. The elevated CO_2 could increase yield and nutrient uptake by rice whereas elevated temperature decreased yield and nutrient uptake. Application of additional N over the current recommended dose could help in adaptation to prevent yield loss due to climate change.
- Availability of soil P improved under high CO_2 condition in direct seeded rice crop. Better root growth and higher activity of both acid and alkaline phosphatase enzymes in elevated CO_2 treatment resulted in increase in availability of soil P. Both agronomic and recovery efficiency of P increased with elevated CO_2 condition.
- High temperature treatments reduced the grain starch content and increased grain protein content in rice. Pusa Sugandh 2 though more sensitive to high day and night temperature, out yielded the performance of Pusa 44.
- Variation in economic yield under elevated temperature and its percentage decrease per degree temperature rise clearly indicate that in both pulse and oilseed crop, *kharif* season crop (chickpea and mustard). Groundnut showed intermediate response for decrease in economic yield under elevated temperature.
- Elevated CO_2 and cyanobacteria application will enhance productivity and N fixation in legumes, with cowpea being more efficient than mungbean crop. Supply of P is important for productivity and N fixation of legumes and in present climate change scenario P application will play crucial role.

- The antioxidant chemicals, ascorbic acid, quercetin and marigold leaves extract were found effective in providing protection against negative impacts of O_3. Marigold leaves extract may be effectively used as natural protectant for alleviating the tropospheric O_3 stress in wheat.
- Elevated carbon dioxide concentration was able to counter some of the negative impact of elevated ozone concentrations on plant growth and yield. However, elevated ozone and CO_2 interaction reduced the nutritional quality (protein content, macro and micro nutrients) of mustard seeds.
- Tolerance potential of different growth stages of maize to MLB disease was more at dent stage as compared to vegetative and tasseling stages to elevated CO_2 and temperature treatments
- The impact of elevated temperature (+2-3°C) specifically from germination to maturity in Groundnut Crop has resulted in significant increase in functional bacterial population and there seems to be no loss of any PGPR traits amongst the *Pseuodomonas, Azotobacter, Acetobacter* and *Enterobacter* population.
- In India, floods and droughts accompanied by high temperature in a single crop season have recently caused severe crop loss. Therefore understanding crop response to these sequential stresses in a season is important for carrying out large scale-impact assessments and developing adaptation strategies through simulation analysis. Dry spell during vegetative growth followed by high temperature coinciding flowering even with adequate soil moisture, and then heavy rains during PFD significantly reduced the biomass and seed yield as also in case moderate drought situation and in high temperature conditions. InfoCrop-Soybean model can be used for simulating the impacts of climatic variability after minor refinement regarding LAI and total dry matter particularly during vegetative growth phase.
- In ground nut drought during early phase caused more reduction in dry matter accumulation than that occurred during seed filling stage. Excess soil moisture seems to increase the vegetative growth and in absence of any other stress increased the crop yield. However, when the crop encountered excess water and drought during the flowering and pod filling stage, it stands to lose the yield. In maize, total biomass was most affected in plants exposed to drought stress during the vegetative growth period and then subjected to excess water during grain filling period. Occurrence of drought during vegetative growth period or during flowering caused more reduction in grain yield. InfoCrop ground nut and maize models could be satisfactorily calibrated to rainfed and irrigated conditions.
- Studies on impacts of increasing climatic risks at different phenological stages on rice and wheat yield indicated that panicle initiation to flowering period is found to be highly sensitive to high temperature and drought stress. The yield reduction was found to be higher in late sown wheat crop than the normal one due to temperature stress. Drought stress related risk assessment study using crop simulation model showed that a 50% rainfall deficit for 30 days during the second month of crop growth after transplanting (reproductive stage), can cause almost crop failure in Maharashtra, Tamil Nadu and Karnataka, while Uttar Pradesh and MP may register about 35% loss in rice yields. Karnataka and Tamil Nadu are more prone to yield reduction due to variability in rainfall in comparison to Marharashtra, Uttar Pradesh and Madhya Pradesh.
- Green gram yield under mulching treatment was higher as compared to control and hydrogel treatment. Mulching treatments provided a seed yield advantage of 41.9% and 14.3% in pigeonpea and green gram respectively compared to control.
- India is the second largest producer of wheat (*Triticum aestivum* L.) in the world. High temperature spell towards the end of the crop season i.e., terminal stage is a major determinant of wheat yield, especially under Indian condition. In terminal heat stress year, yield of wheat can be enhanced using adaptation options such as applying additional doses of nitrogen fertilizer, preponing date of sowing from mid-November to first week of November and providing an additional irrigation especially at grain filling stage. The best adaptation option for alleviating terminal heat stress in wheat is to use tolerant variety, preponing date of sowing to first week of November with application of 30 kg additional N ha^{-1} and forecast-based irrigation.
- Vegetable fields are characterized by intensive production and high N application rates controlling nitrous oxide (N_2O) emissions. Thus Potato production can be the major contributor of Greenhouse gases (GHGs)

from the crop production sector. Application of N through neem oil coated urea in three splits has the potential to reduce the GWP of potato cultivation. A higher emission load from the processing of potatoes indicated the need to reduce the consumption of processed potatoes products.

- The amount of residues generated by biogas production has increased dramatically due to worldwide interest in using renewable energy. Biogas slurry (BGS) originate from anaerobic degradation of organic wastes has been proposed as organic fertilisers because of their high content of N, P, K and other valuable macro- and micro-nutrients. As opposed to the conventional and synthetic fertilizer based agricultural practices, BGS cultivation system differ significantly in different aspects like, it reduces nutrient losses, improve soil fertility, and reduce global warming potential while supporting better crop yields in certain contexts.
- Rice fields are one of the major sources of methane and nitrous oxide emission, two important greenhouse gases (GHGs) contributing 19% and 5%, respectively to the enhanced greenhouse effect. There is a need to find out options to mitigate global warming potential of continuously flooded rice field without reducing productivity. The SRI saved irrigation water upto 44% but there was no difference in grain yield as compared to the transplanted rice. Methane and nitrous oxide emission from basmati cultivar (Pusa 1121) was 12.7% and 3.5% higher than the non-basmati cultivar (Pusa 44), respectively. The GWP in basmati rice was 7.5% higher than non-basmati rice. The study concludes that the SRI method can reduce GHG emission and saves water without any yield penalty.
- Tropospheric ozone (O_3) is a phytotoxic air pollutant and a greenhouse gas and its concentrations are increasing annually by 0.5±2%. Ozone has detrimental effect on plant productivity and chemistry, which might affect the soil C and N cycle and the emissions of N_2O and CH_4, the two important greenhouse gases from cropped soils. Grain yield of rice was significantly impacted by different levels of ozone. It reduced by 11.31% under elevated ozone concentration.
- Leafy vegetables are important contributors among all vegetable crops. Indian spinach plants applied with nitrogen 60 kg ha^{-1} and irrigated at 10 days interval had highest TDM, leaf and seed yield as compared to other treatments. Nitrogen application at 90 kg ha^{-1} reduced the leaf yield irrespective of irrigation frequency. However, nitrogen concentration increased with increase in nitrogen dose. Spinach model could simulate the Indian spinach canopy growth behavior, leaf and seed yield in both harvested and non-harvested plants. This model can be used for application for different studies after more validation.
- Water scarcity is expected to be a major challenge for most of South Asia due to increased water demand and poor water management leading to water related food scarcity. Water stress during vegetative growth and flowering affected soybean yield significantly. Dry spells in August and early September significantly reduce soybean yield in Delhi region and supplemental irrigation, in the event of dry spell, can minimize the yield loss.
- Farmers growing rice and wheat in rotation in the Indo-Gangetic Plains and wheat in central India use combine for harvesting their crops, which generates huge amount of residue in the field. Burning of crop residue leads to the emission of various toxic gases, greenhouse gases and particulate matter in the atmosphere. The concentration of $PM_{2.5}$ and PM_{10} was 401.9 $\mu gm^{-3}$and 527.5 μgm^{-3} due to rice residue burning and 342.9 μ gm^{-3} and 471.84 μgm^{-3} due to wheat crop residue burning. The X-Ray Diffraction of rice and wheat straw ash indicated the presence of silica, potassium chloride, and potassium calcium phosphate.
- India is the largest producer and exporter of basmati rice with about 2/3rd of the production is exported. Organically produced basmati had higher grain elongation, karnel length and karnel length and breadth ratio. Organic basmati exhibited high amylose, reduced crude protein, increased micronutrient (Fe, Mn, Zn and Cu) contents.
- Nitrogen is one of the most essential macronutrient for rice growth and production along with its improved grain quality. Ammonia volatilization is the main process which contributes to nitrogen losses in rice crop. The NH_3 emitted effectively up to the 4-5days from the date of fertilizer application. The rate of NH_3 volatilization increased with increasing temperature.

- Airborne particulate matter (PM) pollution is a very serious problem in India. Plants are variably sensitive to abiotic stresses and PM pollutants. The contribution of the elements in PM load was in the following order C >Fe >N >Mn>Zn > Ca_2^+ >Cu >Ni >Pb> Na^+> K^+ > Mg^{2+} > SO_4^{2-} >Cr > PO_4^{3-} > NH_4^+ and Cl^- > NO_3^->Co >As and Cd > F^-.
- Environmental contamination by heavy metals is increasing enormously in recent years due to anthropogenic activities, soil being the main terrestrial sink for these pollutants. The pattern of total metal accumulation in the vegetables followed the order: Pb>Ni>Cr>Cd>Hg. Dietary exposure to heavy metals, namely Cadmium (Cd), Lead (Pb), Zinc (Zn), Copper (Cu) has been identified as a risk to human health through the consumption of vegetable crops. Seasonal variation reveled that most of the winter vegetables were contaminated with one or more heavy metals while summer vegetables were only contaminated with Ni. Rock-phosphate in combination with vermi-compost and single super phosphate were the most effective amendments in decreasing available Pb for reducing its entry in the food chain.
- *Chlorella minutissima* has the best manurial potential and offers a new option of applying algae to manage the nutrient load and after phycoremediation the biomass itself can be utilized for manure application in agriculture, serving the dual roles of nutrient reduction and valuable manure feedstock production.
- A small scale (batch flow) vertical sub-surface flow treatment wetland of 1570 Lpd capacity was developed for treating municipal wastewaters of IARI.
- Two bio-fuel energy crop plants, jatropha and castor plant species could be grown successfully on marginal and problem soil under rainfed condition and could also be utilized for phytoremediation of heavy metal polluted soil.
- Irrespective of plant densities, jatropha could be a potential candidate to grow as a component of agro-forestry system under irrigated condition in North West parts of India and a plant population density of 1666 per hectare would serve the purpose of both agro-forestry and carbon sequestration apart from other ecological services.
- Fermentation and ethanol yield by *Pichia stipitis* from detoxified hydrolysate was found better as compared to non-detoxified microwave assisted acid pretreated wheat straw hydrolysates. Maximum ethanol yield obtained was 5.29 % (v/v), equivalent to 87.9 % of the theoretical yield. The ethanol yield coefficient was found 0.44 g ethanol g^{-1} sugar utilized.
- A maximum hydrogen yield up to 205 mL/g of sugar was obtained from saccharified hydrolysates of wheat straw by *Enterobacter aerogens NCIM* 5139. These findings highlight the potential of wheat straw for the production of hydrogen.
- The potential of ethanol production from the rice varieties, ranged from 141 to 360 L per ton of microwave assisted-NaOH pretreated straw. These findings highlight the enormous potential of rice straw for the production of ethanol.

Future Perspective

Globally, environmental issues are ever increasing and seen as factors confronting developmental goals. However, considering environmental sustainability while formulating the developmental policies is utmost important for all countries, in general, and for rapidly developing economies such as India, in particular to sustain the economic growth and ecosystems. Like all sectors of economy, Indian agriculture also need to develop at a rapid pace based on the technological innovations. While doing so, there is need to make Indian agriculture resilient to climate change and to environmental stresses while conserving the ecosystem balance. In this scenario, the Environmental Science discipline has a pivotal role to play to analyze, converge by developing and using simulation models, assess and quantify the climate change and its impacts, derive adaptation and GHG mitigation strategies, develop ameliorating strategies for environmental pollution issues and conduct research on current and anticipated environmental issues. The studies should aim to sensitize, generate and provide scientific

information for formulating the eco-friendly developmental policies and developmental initiatives on one hand and to develop the human resources for enabling and strengthening such activities in years to come.

Acknowledgement

We wish to thank all the Faculty Members of the Division of Environmental Science for extending support and cooperation in preparing the research contributions of M.Sc. and Ph.D. students made during the period of 2007-16.

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Microbiology

Sunil Pabbi and Surender Singh

Introduction

The Division of Microbiology was established in 1961 by merging the units of Soil Microbiology of the Division of Soil Science & Agricultural Chemistry and Algology of then Botany Division. The focus of the Division is on basic, applied and strategic research on microorganisms of agricultural importance. Since its inception, the Division has contributed extensively on the isolation and selection of nitrogen fixing- and plant-growth promoting microorganisms for legumes, cereals, millets and oilseed crops. The standard protocols for mass production of bacterial (*Rhizobium*, *Azotobacter*, *Azospirillum* and phosphate& potassium solubilisers), cyanobacterial (blue-green algal bio-fertilizer for rice) and arbuscular mycorrhizal inoculants for various crops have been developed. Effective microorganisms for composting and their quality control parameters for their mass production are available. Mass production of *Rhizobium* inoculants for pulse crops was initiated in the late 1960s. The Division continues to be the prime centre for production and distribution of microbial inoculants of high quality, and for formulation of standards by the Bureau of Indian Standards (BIS) for testing microbial cultures for efficiency and purity of manufactured inoculants.

The Division of Microbiology is the national lead centre for advancing knowledge and understanding of the microbial domains for agricultural application. The blue sky research of the Division includes utilization of microbial gene and metabolite pool for plant growth promotion as well as protection, natural resource management and value addition of biomass through microbial means. More importantly, the Division contributes to the human resource development. The faculty of the Division offers teaching and research guidance for both national and international students for their master's and doctoral degrees. The Centre for Conservation and Utilization of Blue Green Algae (CCUBGA) is affiliated to this Division.The CCUBGA, the National Facility for *Rhizobium* Germplasm Collection and the *Azolla* germplasm collection within the Division are the major provider and storehouse of the national microbial wealth.

The Division of Microbiology serves as a lead centre for development and commercialization of biofertilizers. Liquid bioinoculants for *Azotobacter chroococcum* and phosphate solubilizing bacteria with a shelf-life of 30 months have been developed and filed for patents. The novel strategy of biofilmed biofertilizers aims at using fungal (*Aspergillus awamor*i, *Trichoderma viride* and *Priformospora indica*) and cyanobacterial (*Anabaena torulosa)* matrices for plant growth promotion as well as protection. The Division has developed consortia of hyper lignocellulolytic fungi (*Aspergillus nidulans*, *Trichoderma viride*, *Phanerochaete chrysosporium* and *Aspergillus awamori*) for rapid decomposition of agro-residues. Several biocontrol agents against plant pathogenic fungi and their modes of action have been identified. Gene(s) involved in the fungicidal activity were identified and strategies have been developed for their utilization. The Division has the germplasm of bacteria and cyanobacteria isolated from extreme environments that can tolerate low and high temperature of 4 and 90 °C, 25% NaCl concentration and pH of 3.0. The major research thrust areas of the Division are:

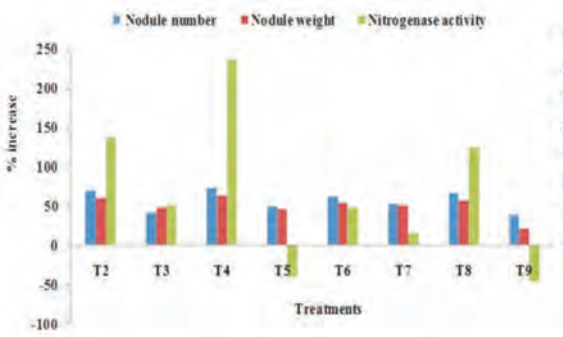
- Plant –microbe interaction for nutrient acquisition and growth promotion as well as protection
- Organic matter decomposition for improved soil fertility and health
- P solubilising and mobilizing microorganisms for enhancing P use efficiency
- K-solubilizing microbes for improving K availability and mechanism of K solubilization

- Formulation of biofilmed biofertilizers for nutrient management and biocontrol measures
- Understanding the mechanism and isolation of microorganisms with biocontrol capabilities
- Diversity analysis of eubacteria and archaebacteria in extreme environments
- Technology development for production of second generation biofuel
- Understanding and application of anaerobic microbial processes of agricultural importance
- Economic utilization of blue green algae and microalgae for value added products

Significant Student's Research Achievements

Plant Microbe Interaction

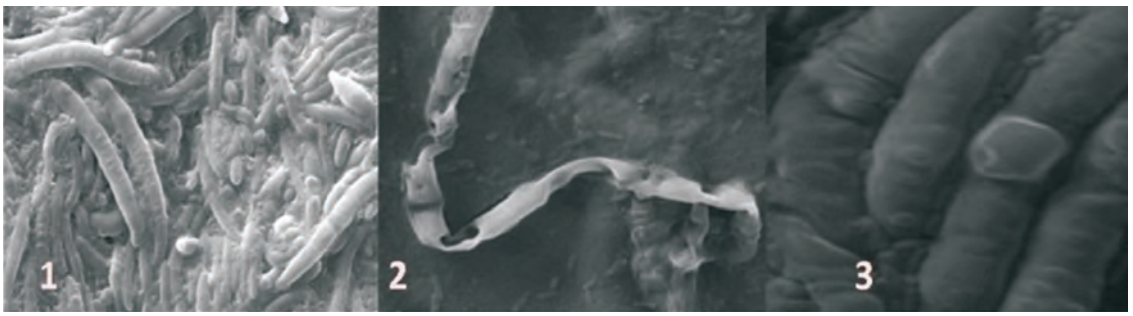
Chickpea (*Cicer arietinum*) is an important cool season legume crop in India. It's economic success depends upon the symbiotic association with root nodulating bacteria (*Mesorhizobium ciceri*). Beside *Mesorhizobium,* other nodule associated bacteria (NAB) also help to stimulate plant growth, nitrogen fixation, phosphate solubilisation, IAA production and induction of resistance to plant pathogens while residing within the plant parts. Hence, the study was undertaken to isolate and identify the most promising nodule associated bacteria from chickpea cultivars. These bacteria were characterized for their plant growth promoting traits as well as associative effect on *Mesorhizobium* inoculation. A total of seventy three NAB were isolated from surface sterilized nodules of different chickpea cultivars using various kinds of media. Screening for the improved seedling growth showed that 91.78% of them enhanced radical length, and 87.67% of them increased plumule length under *in-vitro* conditions. About twenty four isolates were selected from preliminary screening and were subjected to their growth promoting potential under pot culture conditions using selected cultivar (PUSA 372). The promising nodule associated bacteria (NAB 69) showed a significant increase in plant growth regarding shoot dry weight (40.63%) and root dry weight (45.09%) in comparison to control. Its interactive effect was evaluated with *Mesorhizobium ciceri* for growth attributes, nodulation potential and N & P uptake under pot culture conditions. The highest nitrogenase activity (74.12 µmoles C_2H_4/g of fresh weight of nodules/hr) was recorded in the treatment involving NAB 69 along with *Mesorhizobium ciceri* at vegetative stage. Similarly, the plant growth and N & P uptake were also highest under the treatment of NAB 69 along with *Mesorhizobium ciceri* at 50% recommended dose of fertilizers. *In-vitro* screening for the functional potential of NAB showed a positive result with P solubilization and IAA production ability. However, nitrogenase activity, siderophore production, and HCN production were not detected in the selected bacterial isolate. The 16S rDNA sequencing revealed that the bacterial isolate was closely related to *Enterobacter* sp. with 99% maximum identity. This investigation led to identifying the natural associations between the most promising nodule associated bacterium and *Mesorhizobium ciceri* in chickpea and the positive influence of their interactions.



Percent increases or decreases over the control (T_1) are provided. Treatment details are: Treatment details are: T_2-*Mesorhizobium ciceri* alone; T_3-NAB alone; T_4- *Mesorhizobium ciceri*+NAB; T_5-50% RDF (positive control); T_6-*Mesorhizobium* + 50% RDF; T_7-NAB + 50% RDF; T_8-*Mesorhizobium ciceri*+NAB+50% RDF & T_9-100% RDF (positive control)

Nodulation potential and nitrogenase activity in chickpea as influenced by the interactive effect of the promising nodule associated bacterium (NAB) and *M. ciceri*

Among the thirteen chitinolytic rhizobacterial isolates from pigeonpea, two cultures namely *Pseudomonas* sp. strain NS-1 and *Bacillus* sp. strain NS-22 showed good inhibition of *Fusarium udum* growth *in vitro* and also reduced fusarial wilt incidence and promoted better plant germination and plant growth as compared to other cultures. Light and scanning electron micrographs revealed that both the cultures produced changes in hyphal morphology and also carried out lysis of fungal hyphae. Both the cultures possessed multiple biocontrol traits such as siderophore production, production of toxic secondary metabolites such as NH_3 and HCN. Inoculation with these cultures reduced root mortality and improved root and shoot length and yield. Inoculation with these cultures led to higher induction of defense related enzymes such as polyphenol oxidase, peroxidase, phenyl alanine ammonia lyase in the treated plants.



Effect of *Pseudomonas* sp. strain NS-1 and *Bacillus* sp. strain NS-22 on fungal hyphae. 1. Untreated control; 2., 3. Treated fungal hyphae showing lysis and bulging

Soybean is known to be nodulated by two different genera, *Bradyhizobium*(slow growing rhizobia) and *Sinorhizobium* (fast growing) species. The investigation was an attempt to understand the soybean genotype induced alterations on the cell surface of the fast and slow growing root nodulating bacteria of the soybean and how it contributes to the final interaction. Five soybean genotypes viz:DS12-13, DS9712, DS2705, SL979, SL979 were evaluated for their symbiotic potential with two slow (KAS-1, MTCC10753) and two fast growing root nodulating bacteria (DS-1, LSR-8). Genotype DS12-13 nodulated well across the four strains, while DS2705 was poorly nodulated by the four strains. Among the strains, slow growing KAS-1 and fast growing DS-1 strains were effective across the genotypes. The mean ARA of KAS-1 was found to be 30.37 and 26.38 n moles of C4H4 produced /mg/ndw. The generation time of KAS-1 and DS-1 were found to be 12 and 3 hours respectively. These strains confirmed to the biochemical tests of rhizobia by failing to grow on citrate and hofer's alkaline media. Intrinsic antibiotic resistance screening of four strains showed that slow growing strains were resistance to more number of antibiotics compared to fast growing strains. Chemical analysis of the root exudates (RE) of the five genotypes varied in concentration and number of amino acids, organic acids and Isoflavonoides. Isoflavonoides at retention time of 2.9, 4.9 and 7.7 were found common in the root exudates of the all genotypes, however, the ratio of compound with RT of 2.9 to the compound with RT of 7.7 varied among the genotypes. RE influenced alterations were observed through FT-IR on the cell surface of the fast growing strain DS-1. Two proteins of 50 and 35 KDa in fast growing strain DS-1 expressed in SDS PAGE and changes were observed in lipopolysaccharides (LPS) profile when grown with RE of high nodulating genotype DS12-13. Root exudates of the high or low nodulating genotype did not influence the proteins or LPS patterns of slow growing strain KAS-1. The study indicated the role of host RE on Rhizobial associations.

Morphologically diverse 188 endophytic bacteria isolated from two maize varieties PEEHM-5 (Hybrid) and PC-4 (Composite) were screened for P,K, Zn solubilization, production of siderophore, hormones, ACC deaminase, NH_3 and biocontrol of two maize fungal pathogens. Potential 31 PGP endophytes were clustered into 13 phylogenetic groups. Sequencing of amplified 16S rRNA gene of representative isolates from PC-4 identified them as *Bacillus aryabhattai, Pantoea cypripedii, Bacillus licheniformis, Klebsiella* sp. *Pantoea dispersa, Klebsiella variicola, Pantoea* sp. and *Agrobacterium larrymoorei whereas* from PEEHM-5 the endophytic bacterial isolates were identified as *Bacillus* sp., *Bacillus amyloliquefaciens, Lactococcus lactis, Bacillus cereus* and *Staphylococcus*

hominis. In planta evaluation of 6 potential PGP endophytes showed the beneficial impact on plant biometric parameters and the impact of bacterial inoculation at 75% NPK input matched with 100% NPK input; clearly indicating the minimum saving of 25% of NPK.

Plants exhibit differential potential for uptake of nutrients which depends on the genetic makeup of the plant and the availability of nutrients in the soil. This study was carried out to isolate and identify efficient endophytes for wheat to further enhance the genetic potential of low accumulating wheat genotype for uptake and translocation of iron and zinc in plant parts and grains. Of 13 genotypes screened for uptake of these micronutrients, three genotypes (4HPYT-404, GW-07-112, HD-3086) accumulating <20 mg kg^{-1} zinc and 12 genotypes except GW-07-112, accumulating <45 mg kg^{-1} of iron in grains were designated as low accumulator of zinc and iron respectively. A total of 213 endophytes obtained from different genotypes were screened for zinc solubilization and siderophore production and 8 endophytes (6 bacteria and 2 fungi), each efficient for zinc solubilisation or siderophore production were used as inoculants. The percent increase in zinc uptake ranged from15.2-130.4% and iron uptake ranged from 15.16-156.85% respectively, due to inoculation of endophytes. Among the endophytes, isolate DS-178 and DS-179 were more potent for zinc acquisition whereas, endophytes DS-68 and DS-163 were efficient for iron acquisition in grains. All the endophytes showed IAA production, siderophore production, phosphorus solubilisation and ammonia production ability. Only DS-178 exhibited positive test for HCN production. On the basis of 16S rRNA gene sequencing these four endophytes were identified as *Bacillus subtilis* DS-178, *Arthrobacter* sp. DS-179, *Arthrobacter sulfonivorans* DS-68 and *Enterococcus hirae* DS-163.Two siderophore producing (*Arthrobacter sulfonivorans* DS-68 and *Enterococcus hirae* DS-163) and two zinc solubilizing endophytes (*Bacillus subtilis* DS-178 and *Arthrobacter* sp. DS-179) were used to inoculate low and high Fe and Zn accumulating genotypes respectively, in soils sufficient and deficient for Fe-Zn. In general, the amount of Fe and Zn in grains due to inoculation with endophytes was 2 folds higher as compared to un-inoculated control. The low and high Fe or Zn accumulating genotypes responded in almost identical manner to endophyte inoculation irrespective of the soil type. TEM studies revealed that the endodermis, cortical region, root hair extension, xylem and xylem vessels, pericycle and vascular bundles were more pronounced and thicker in inoculated treatments as compared to control. The organic acid profile showed five types of organic acids in root exudates with citric acid being the predominant acid produced. The amount of total organic acids was 5 fold and 8 fold higher due to inoculation with *Arthrobacter sulfonivorans* and *Arthrobacter* sp. respectively as compared to control. The concentration of citric acid, succinic acid and acetic acid increased many fold in root exudates of inoculated treatments. Four *TaZIP* genes were targeted for expression studies using gene specific primers and expression was achieved only for *TaZIP3* and *TaZIP7* genes in wheat genotype 4HPYT-414. The results clearly indicated endophyte mediated over expression of these two genes in roots and shoots.Zinc solubilizing (*Bacillus subtilis* DS-178 and *Arthrobacter* sp. DS-179) and siderophore producing (*Arthrobacter sulfonivorans* DS-68 and *Enterococcus hirae*) endophytes were also evaluated under field conditions for biofortification of wheat grains with Fe and Zn. Both low and high Zn or Fe accumulating wheat genotypes had the significant results for Fe or Zn accumulation in wheat grains under field conditions with respect to endophytes inoculations and phytic acid was significantly decreased in all wheat genotype used. Endophytes inoculation also gave positive feedback to protein concentration and phosphorus concentration in grains.

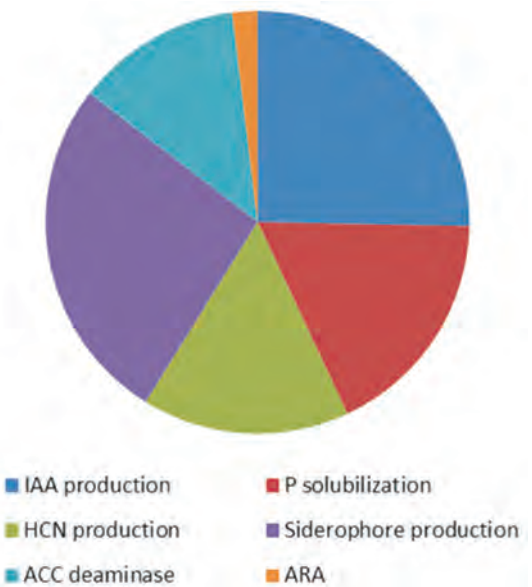
In a pursuit to identify *Rhizobium* cultures that are efficient for nitrogen fixation and can inhibit some of the soil borne fungal pathogens, 150 chickpea specific *Mesorhizobium ciceri* isolates developed through isolation from the root nodules and procured from culture collection, HAU, Hisar were screened against *Fusarium oxysporum* f. sp. *ciceri*, *Ascochyta rabiei*, *Botrytis cinerea*, *Sclerotium rolfsii* and *Macrophomina phaseolina* for their antifungal activity. Among them, 10 isolates showed good antagonistic activity and percent inhibition of fungal mycelia ranged from 5-49 %. Promising cultures also exhibited important anti-fungal and plant growth promoting traits such as production of HCN, ammonia, IAA and phosphate solubilisation. *M. ciceri* isolate A13 followed by CR24 were significantly superior with regards to nodule numbers, nitrogen fixing potential and dry weight of nodules, roots and shoots. In a field trial also, seed treatment with rhizobial isolates A13 and CR24 improved different plant growth parameters and yield. Based on these results,

Rhizobium based biofilms using isolate A13 or isolate CR24 as partners and *Trichoderma viride* as fungal matrix were developed. Jensen's N free medium amended with 1% yeast extract, sequential inoculation of the cultures and incubation under static conditions after inoculation of fungus for 14 days yielded maximum biomass of biofilm. These biofilms showed 9 to 11% increase in antifungal activity towards *Fusarium oxysporum* f. sp. *ciceri* compared to respective rhizobial isolates. These biofilms also exhibited significant enhancement in several plant growth promoting attributes like seed germination, production of ammonia, IAA and phosphate solubilisation, when compared to their individual partners. The effect of selected *M. ciceri* cultures individually and their respective biofilms on growth parameters of chickpea under pathogen challenged soil illustrated that the biofilms performed *at par* with the *M. ciceri* strains for most of the plant biometrical and disease related attributes.

Elicitation of defence related enzymes like L-phenylalanine ammonia lyase (PAL), peroxidase (POX) and polyphenol oxidase (PPO) was found to be higher in *M. ciceri* isolates or their respective biofilms treated plants as compared to uninoculated plants under pathogen challenged soil. Induction of pathogenesis related proteins like chitinase and β 1,3- glucanase was also observed. Profiling of phenolics in root and shoot of *M. ciceri*/biofilm treated plants revealed significant accumulation of tannic, ferulic, salicylic, and t-cinnamic acids as compared to uninoculated plants. Larger accumulation of isoflavonoids like genistein, formononetin and biochanin A and organic acids like malic, succinic, propionic and formic acid was also observed in plants inoculated with rhizobia or their biofilms, which signifies better induction of plant defense mechanisms under pathogen challenge. This study reveals the potential of *Rhizobium-Trichoderma viride* biofilms to be used as inoculant having dual purpose of nitrogen fixation and antifungal activity.

Bacteria isolated from pearl millet belonged to diverse groups such as gram negative bacteria, gram positive bacteria, oligotrophs, diazotrophs and these were found harbouring interiors of pearl millet plant. High population of gram positives and oligotrophs was recorded in all the plant tissues such as roots, stem and leaves for both the drought resistant and susceptible cultivars of pearl millet.

Out of a total of 215 morphotypes obtained, 50 were osmotolerant and possessed multiple plant growth promoting activities. There was a very high incidence of IAA and siderophore producers, P-solubilizers and ACC deaminase producers among these isolates and very low incidence of nitrogen fixers. All the 50 osmotolerant isolates were placed in 16 phylogenetic clusters after RFLP analysis of their 16S rDNA; species of *Bacillus* were the dominant osmotolerant endophytic bacteria colonizing interiors of pearl millet.



Among fourteen osmotolerant isolates screened for water stress alleviation in mustard, *Bacillus* sp. strain NA D7 and *Bacilluscereus* strain MR D 17 performed best for most of the plant parameters. Both the strains possessed multiple plant growth promoting activities such as IAA and GA production, P-solubilization, ACC deaminase activity and exopolysaccharide production. Moderate osmotic stress had a beneficial effect on most of the PGP activities, both under *in vitro* conditions and during association with plants. Inoculation with both the strains significantly improved yield, shoot biomass and root system architecture such as root volume, surface area, diameter and length under both normal and water deficit stress conditions. Inoculation with both the strains significantly improved plant antioxidant status, physiological status and decreased lipid peroxidation. Inoculation with both the strains significantly improved expression of drought stress responsive genes DREB 2, DREB 1-2 and RD 26 under both normal and water deficit stress conditions.

Effect of inoculation on growth of mustard under water stress conditions.
1. No stress uninoculated, 2. Stress uninoculated, 3. Stress inoculated

The osmotolerant bacterial isolates were evaluated for their alleviation of water deficit stress in drought susceptible mustard cultivar Pusa Karishma LES-39. Two promising osmotolerant cultures NA D7 and MR D17 identified as *Bacillus cereus* and *Bacillus* sp. were observed to possess multiple PGP traits such as P solubilization, phytohormone production and ACC deaminase activity. Inoculation had a beneficial effect on plant biomass, productivity, oil content, plant nutrient status and root system architecture under both normal and water stress conditions. Various plant physiological parameters *viz.* amino acid, proline, starch, sugars, phenolics and chlorophyll content were significantly enhanced in inoculated plants under water stress conditions. Inoculation with the osmotolerant bacteria significantly improved membrane stability index and relative water content; and decreased lipid peroxidation in these plants. Activity of antioxidative enzymes such as catalase, ascorbate peroxidase, glutathione reductase and superoxide dismutase also increased significantly when plants exposed to water stress conditions were inoculated with these bacteria. There was also a considerable enhancement in expression of drought responsive genes DREB 2 and DREB 1-2 due to inoculation under water stress conditions.

Salt tolerant *Bacillus* sp. strain SSA61 and *Stenotrophomonas* sp. strain SSM66 possessed multiple plant growth promoting traits viz. P-solubilization, IAA and HCN production. High salt concentration of 10% NaCl did not have any inhibitory effect on the PGP traits of these two cultures.Inoculation with these two cultures improved seed germination, root and shoot dry wt. in maize under saline stress conditions. Inoculation with these two cultures improved plant biochemical status viz. proline, amino acid, total sugars, starch and chlorophyll content. There was also decrease in electrolyte leakage, and increase in relative water content and membrane stability.

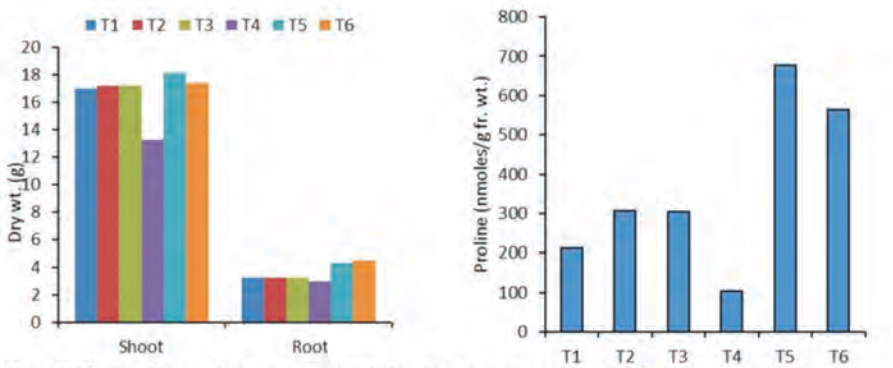
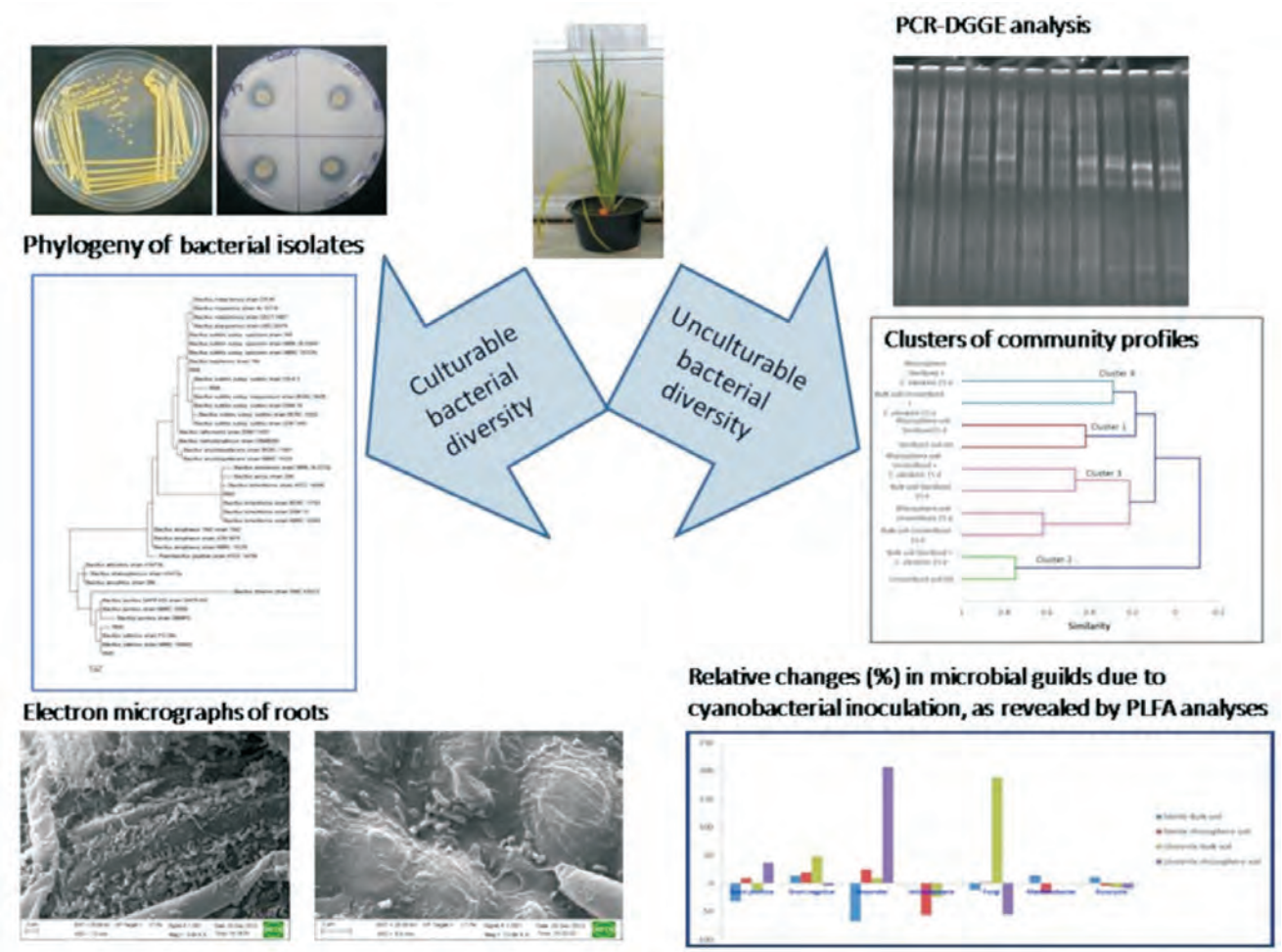
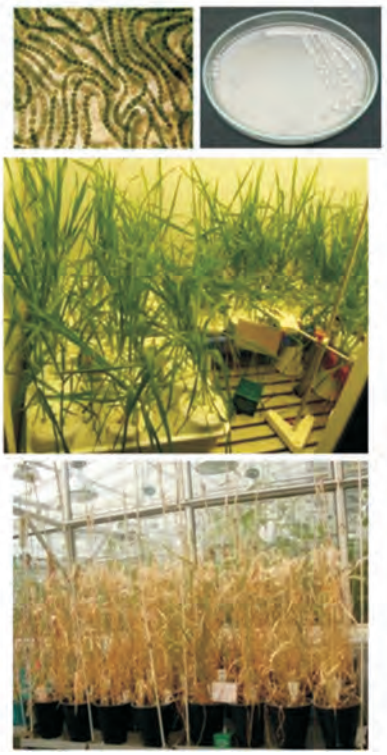


Fig. Effect of inoculation on physiological status and growth of maize plant under saline stress conditions. T1 – No stress uninoculated; T2 – No stress SSA61 T3 – No stress SSM66; T4 – Saline stress uninoculated; T5 – Saline stress SSA61 T6 – Saline stress SSM66

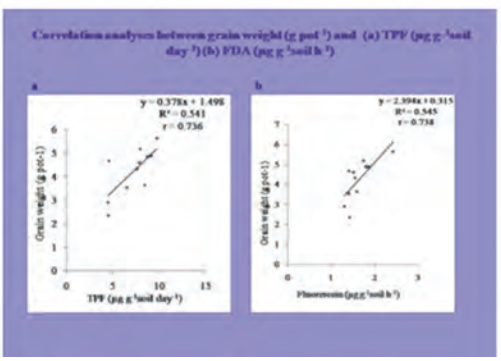
Above ground and below ground interactions between plants and microorganisms represent the main drivers for sustained health and productivity of crops. An investigation was undertaken to analyse the effect of cyanobacterial inoculation (*Calothrix* sp.) on the structure and function of bacterial communities in soil and rice (cv. Pusa 2511) microbiome, using a combination of culturable and culture-independent approaches. Population densities ranging from 10^6-10^9 CFU/ml of culturable microorganisms were recorded in the root and shoot tissues and Fatty acid methyl ester (FAME) analysis, along with 16S rRNA sequencing of the culturable microbiome of rice plants suggested the predominance of the members of *Bacillaceae.* Significant increases in plant growth attributes, nitrogenase activity, and indole acetic acid production, and activities of hydrolytic and defense enzymes were recorded in the plants with cyanobacterial inoculation. Scanning Electron Microscopic examination of roots showed the entry of cyanobacterium. Analyses of the influence of cyanobacterium on the soil microbiome under the controlled conditions of the National Phytotron Facility revealed more than 30 distinct types of phospholipid fatty acids -PLFA(s) and other mixed functional groups. Principal component analysis (PCA) revealed that cyanobacterial inoculation brought about distinct spatial and temporal changes in the concentrations of individual PLFA(s) and in the biomarkers representing different microbial guilds- Gram-positive and Gram-negative bacteria, anaerobes, actinobacteria, fungi and eukaryotes. The denaturing gradient gel electrophoresis analysis of total community DNA supported the PLFA analysis and illustrated the significant influence of cyanobacteria on the diversity and abundance of members belonging to the soil microbiome.It can be concluded that the positive effect of cyanobacterial inoculation on the rice plant growth is probably due to its role in maintaining the "beneficial core microbiome of rice".



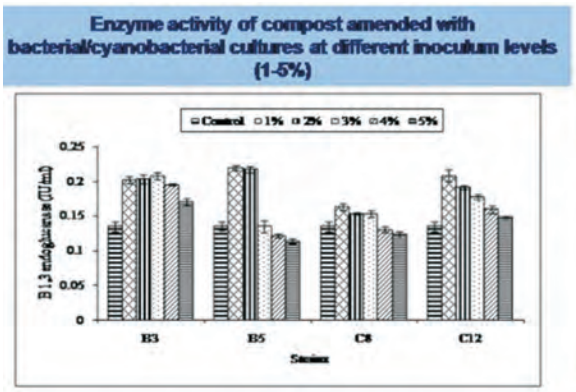
Beneficial plant–microbe interactions in the rhizosphere are considered as the determinants of plant health and soil fertility. An investigation aimed at characterizing the plant growth promoting ability of selected bacteria and cyanobacteria, with emphasis on IAA production was undertaken in wheat. Thin layer chromatographic and HPLC analyses of cell free extracts of bacterial and cyanobacterial strains revealed the presence of indole acetic acid. Hydroponic experiment and pot experiment using unsterile soil with two promising bacterial (WRB4, WRB10-*Providencia* sp. and *Alcaligenes* sp.) and two promising cyanobacterial strains (WRC3, WRC4-*Anabaena oscillarioides* and *Anabaena torulosa*), either singly or in combination resulted in significant enhancement of various soil and plant growth parameters of wheat (PBW 343). Significant correlation between plant biometrical and yield parameters with microbiological activity was recorded. PCR based amplification undertaken to identify gene (s) involved in IAA production revealed the presence of putative amidase in all the four. In two bacterial and two cyanobacterial strains, the presence of indole-3-glycerol phosphate synthase involved in synthesis of indole-3-glycerol phosphate was recorded. This is the first time report on involvement of *Alcaligenes* sp. and *Providencia* sp. in plant growth promotion of wheat crop, and the synergistic effect of bacterial and cyanobacterial strains as evidenced by enhancement of plant growth and soil fertility parameters.

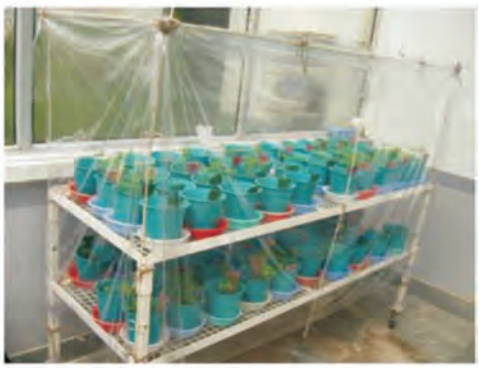


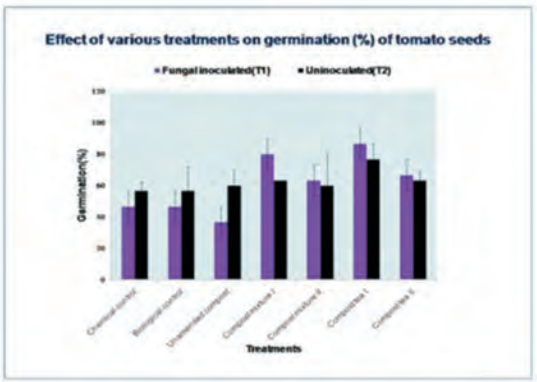
Treatments		Crop biomass (g/pot)	Grain weight (g/pot)	Vigour index
T1	WRC3 + 2/3 N + full dose of P and K	49.037	5.183	6988.8
T2	WRC4 + 2/3 N + full dose of P and K	45.883	4.607	6483.3
T3	WRB4 + 2/3 N+ full dose of P and K	49.297	5.640	7361.2
T4	WRB10+ 2/3 N + full dose of P and K	47.463	4.890	7300.0
T5	WRC3 +WRB4 + 2/3 N+ full dose of P and K	43.317	4.897	7355.6
T6	WRC3+ WRB10+2/3 N+ full dose of P and K	41.927	4.333	7222.2

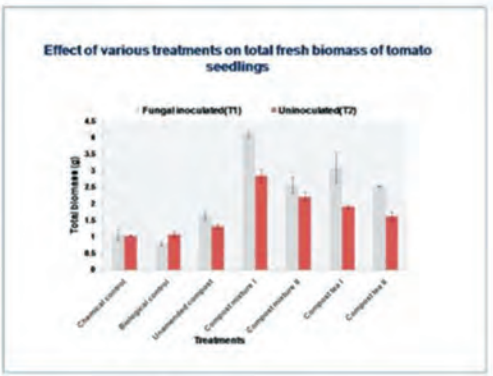


Pre- and post-emergence damping off diseases, caused by fungi bring about serious losses in nursery based horticultural crops. Compost based suppression of a wide range of soil borne diseases has been demonstrated in several studies, but use of composts as carriers for biocontrol agents is not investigated. Amendment of paddy straw compost with bacterial and/ cyanobacterial strains and their characterization in terms of fungicidal activity and activity of hydrolytic enzymes, revealed significantly higher values of microbiological parameters and hydrolytic enzymes at 1-2% level of inoculum. The potential of antagonistic cyanobacteria / bacterial cultures amended compost and the compost tea were evaluated for suppressing the disease caused by the inoculation of a fungal consortium *(Pythium debaryanum, Rhizoctonia solani, Fusarium oxysporum* and *Pythium aphanidermatum*) in tomato. Comparative performance of uninoculated potting mixture, in which the microbe-amended/unamended composts and compost tea were employed, revealed the superiority of the composts/





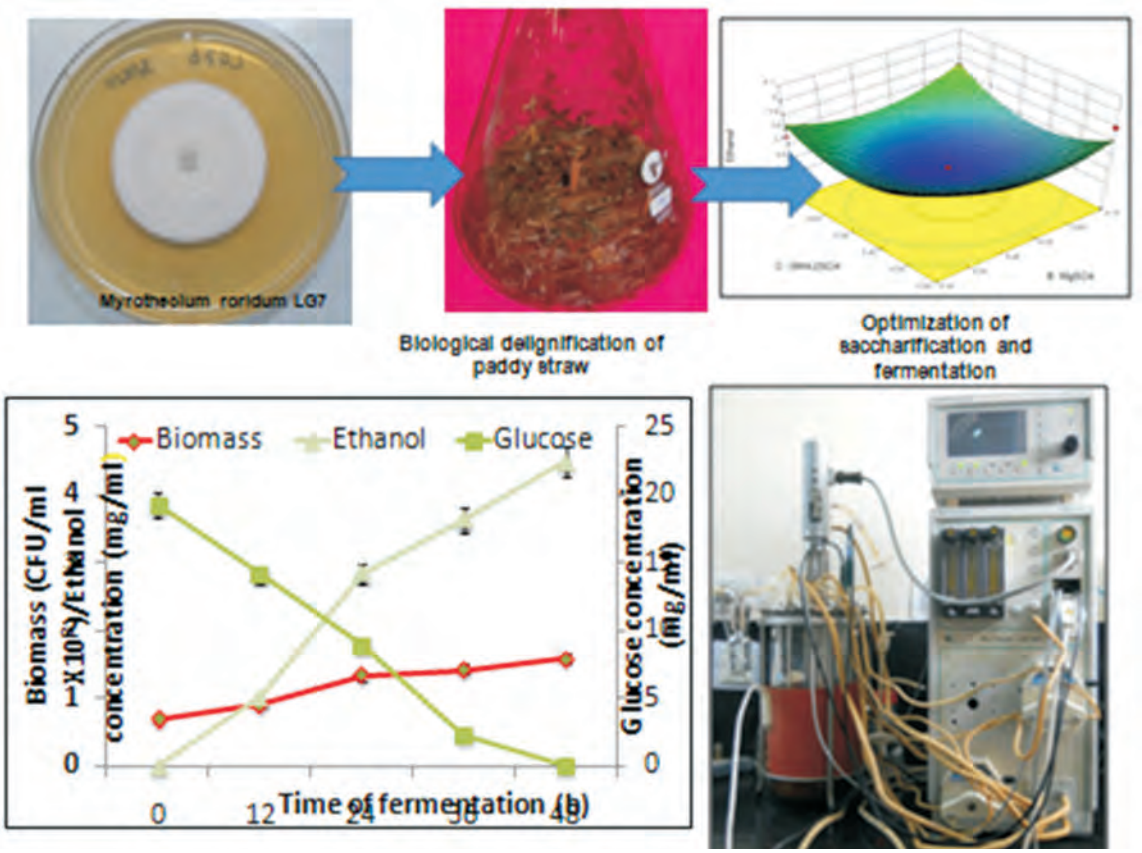




compost teas in enhancing seed germination, seedling length and biomass in the fungi challenged treatments. The most promising compost (amended with *Anabaena oscillarioides* C12 and *Bacillus subtilis* B5) performed significantly better as compared to other treatments in terms of reduction in disease severity, reduction of fungal load and enhancement of plant parameters. Such novel microbe–fortified composts can be useful inputs ion for sustainable and green agriculture.

Agro-Residue Utilization

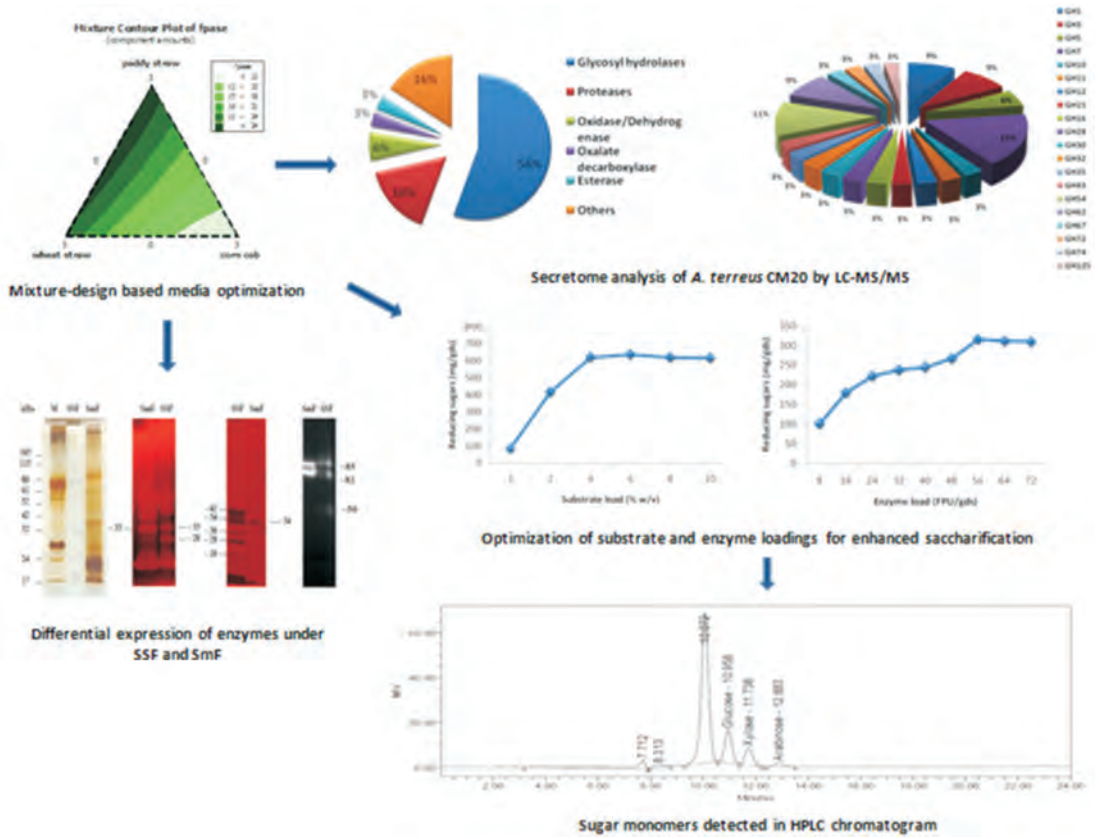
Bioethanol from lignocellulosic biomass is an environmental friendly and sustainable renewable energy source which holds a remarkable potential to meet the current energy demand. During the production of bioethanol, fermentation parameters like temperature, pH and nutrient supplementation play an important role in growth of yeast and to achieve good ethanol yield. By optimization of these parameters, a perfect strategy can be materialized to improve ethanol yield, minimize the total cost of production and maximize substrate consumption. The current study was thus aimed at optimizing the fermentation conditions for efficient ethanol production from biologically pretreated paddy straw using one factor at a time approach. Fifteen yeast strains were screened for their fermentation efficiency on minimal media with glucose as sole carbon source. The strain *Saccharomyces cerevisiae* LN1 showed highest fermentation efficiency and thus, was selected for further fermentation experiments. Effect of pH and temperature on ethanol production by *S. cerevesiae* LN1 was observed by fermentation of minimal media with glucose at different values of pH and temperature. Maximum amount of ethanol was observed at pH 5.0 and temperature 30°C. Paddy straw pretreated with fungus *Myrothecium roridum* LG7 was saccharified with indigenous enzyme cocktail from *Aspergillus niger* SH3 producing total sugar yield of 26.14 mg/ml with 19.23 mg/ml of glucose. Enzymatic hydrolysate was then fermented using *S. cerevesiae* LN1 to observe the effect of nutrient supplementation (Yeast extract, $MgSO_4.7H_2O$ and $(NH_4)_2SO_4$ on ethanol production. Higher ethanol was produced from saccharified material fermented without supplementation of any nutrient source. With the scale up of ethanol production under optimized conditions in 7L bioreactor, 4.46



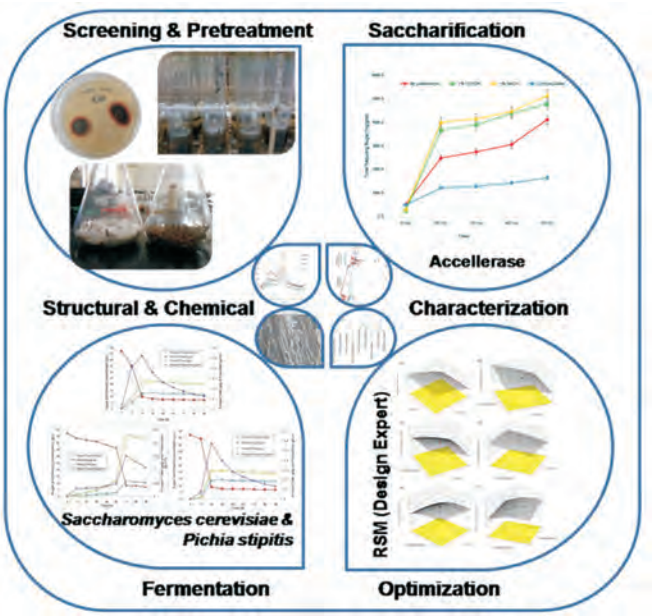
Evaluation of carrot grass (*Parthenium* sp.) for bioethanol production

g/l of ethanol was produced with fermentation efficiency of 47.2%. TLC of enzymatic hydrolysate confirmed the presence of p-coumaric acid, ferulic acid, vanillic acid, gallic acid and many other aromatic compounds and inhibitors in the saccharified material which limits fermentation efficiency of yeast strain. Thus, it can be concluded that optimization of fermentation conditions can lead to development of a cost effective process for efficient ethanol production, exploitation of which also requires removal of aromatic compounds and inhibitors which may hinder the ethanol production efficiency.

Developing an enzyme preparation with defined hydrolytic activities can circumvent the need for supplementing cellulases with accessory enzymes for enhanced hydrolysis. With this objective, mixture design approach was used in the present study to enhance glycoside hydrolase production by a fungal isolate, *Aspergillus terreus* CM20, by determining the proportion of different lignocellulosic components as enzyme inducers in the culture medium. A mixture of paddy straw and wheat straw (1.42:1.58) resulted in 2-4 fold enhancement in cellulase and xylanase activities. The differential expression of enzymes by the fungus under submerged (SmF) and solid-state (SSF) fermentation conditions was also studied. The SSF method was found superior to SmF in terms of 5-8 fold increase in enzyme activities and multiplicity of cellulase and xylanase isoforms observed in zymography. Analysis of the *A. terreus* CM20 secretome, induced using the optimized medium, by liquid chromatography coupled tandem mass spectrometry (LC–MS/MS) identified 63 proteins. Functional classification revealed the hydrolytic system of *A. terreus* CM20 to be composed primarily of glycoside hydrolases (56%), which belonged to 20 GH (glycoside hydrolase), one CE (carbohydrate esterase) and one AA (auxiliary activities) families. The major isoforms of endoglucanase (EGI (33 kDa), EGII (26 kDa)) and β-glucosidase (BGLI (85 kDa), BGLII (82 kDa) expressed by the fungus were electro-eluted and characterized. The isoforms had temperature and pH optima of 50°C and 5.0. Kinetic characterization revealed higher catalytic efficiencies



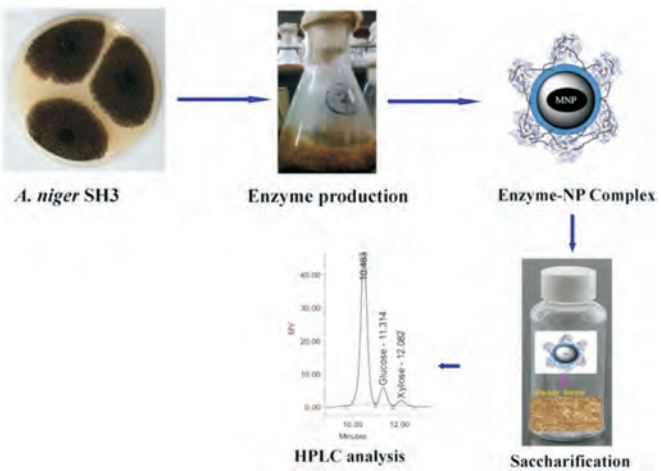
of EGII and BGLI isoforms. The synergistic action of *A. terreus* CM20 enzyme cocktail (CMCase (365.03 18 IU/g), FPase (161.48 IU/g), Avicelase (15.46 IU/g), β-glucosidase (920.92 IU/g) and xylanase (9627.79 IU/g)) was assessed by saccharification of differently pretreated lignocellulosic substrates. Under optimum conditions, saccharification of alkali-pretreated paddy straw released 616.8 mg/gds of sugars, mainly glucose, xylose and arabinose. The study, therefore, revealed the applicability of indigenous cellulase preparations for lignocellulose bioconversion in biorefineries.



The present investigation explores the potential for bioethanol production by using chemical and biological pretreatment methods. Acid and alkali pretreatments were compared with biological pretreatment with lignolytic fungus *Marasmiellus palmivorus* PK-27 isolated from decaying biomass. Changes in structural and chemical properties were studied using Scanning Electron Microscope (SEM), Fourier Transform Infra Red (FTIR) and X-ray diffraction (XRD) analysis after chemical and biological pretreatment. Alkali (1% NaOH) pretreatment showed significantly higher recovery of total reducing sugars (513 ± 41.0 mggds^{-1}) compared to other pretreatments after 48 h of enzymatic hydrolysis with saccharification yield of 76.6%. The enzymatic hydrolysis parameters *viz*. temperature, pH, enzyme loading and substrate loading rate were optimized using response surface methodology (RSM)

with Central Composite Design (CCD) to attain the maximum saccharification yield. The experimental value of saccharification yield (85.80%) was higher than the predicted value (80.08%) proposed by the developed model under optimum conditions (temperature - 50°C, pH - 4.53, enzyme loading - 8% and substrate loading - 2.4%). This demonstrated a significant relation between predicted and experimental response in terms of saccharification yield. Monoculture fermentation of enzymatic hydrolysate with *Saccharomyces cerevisiae* LN1 resulted in ethanol yield of 0.27 g/g within 36 h while *Pichia stipitis* NCIM3498 produced 0.23 g/g of ethanol yield after 72 h. The co-culture fermentation with both yeasts enhanced the ethanol concentration (14.28 ± 0.31 g/L) as well as ethanol yield (0.30 g/g). These findings reveal the promise of *Parthenium* sp. biomass as an attractive feedstock for bioethanol production.

The study was aimed at microbial delignification of paddy straw with three selected organisms, *Trametes hirsuta* MTCC136, a fungal isolate AMI and an actinomycete isolate 38 for enhanced saccharification. High levels of ligninases and low levels of cellulases expressed by these organisms facilitated their selection for solid state fermentation of paddy straw and delignification. High extent of lignin break down by AMI and actinomycete isolate SSR 38 was evident from the high absorbance values at 205 nm exhibited by the alkali extracts of pretreated paddy straw and the partially degraded and chemically modified lignins could be recovered as value-added acid-precipitable polymeric lignin (APPL). Maximum amount of lignin could be recovered as value-added APPL from the paddy straw samples subjected to solid state fermentation with the actinomycete. The high levels of ligninases of *Trametes hirsuta* and presence of lignin fragments in alkali extracts did not correlate, thereby, suggesting a possible mineralisation of lignin by the fungus. Greatest enhancement in glucan content was seen when paddy straw was pretreated for 21 days with fungal isolate AMI and actinomycete isolate 38. Enzymatic saccharification of biologically pretreated paddy straw resulted in much higher levels of reducing sugars as compared to the controls and untreated straw. Paddy straw pretreated with fungal strains yielded higher amounts of sugars, both after 10 and 21 days of pretreatment, when a less potent enzyme mixture was used. With a potent cellulase enzyme, incubation of paddy straw pretreated for 10 days with the actinomycete isolate 38 resulted in an enzymatic saccharification efficiency of almost 100% within 48 hours and nearly 60% saccharification efficiency was obtained when paddy straw was pretreated for 10 days with the fungal isolate AMI. These findings highlight the potential of actinomycete and other fungi for biological pretreatment, which could prove extremely useful in production of bioethanol when used for removal of lignin from lignocellulosic substrate.



Immobilization of indigenous cellulase from the *Aspergillus niger* SH3 and commercial cellulase (Cellic CTec3) on five nanoparticles (Iron oxide, Silicon oxide, Magnesium oxide, Zinc oxide and Silver oxide) was studied by two different methods- physical adsorption and covalent coupling. The enzyme-nanoparticle formulations were screened on the basis of the protein binding, enzymatic activities and immobilization efficiency. Iron oxide-enzyme formulations were found to perform better for both indigenous and commercial enzyme with 60 to 80% immobilization efficiency. Besides better thermo-tolerance, the covalently immobilized showed better catalytic efficiencies. The indigenous immobilized enzyme showed more saccharification yield (375.39 mg/gds) than free enzyme (339.99 mg/gds) under optimized condition (60 ˚C temperature, 5.0 pH and 6% substrate loading). The commercial immobilized enzyme showed less sugar yield (314.77 mg/gds) as compared to the free enzyme (366.6 mg/gds). After 2 cycles of saccharification, 55 and 45 % enzyme activities were recovered for indigenous and commercial enzyme respectively. In conclusion, covalently immobilized magnetic enzyme nanoparticle

complex showed promise for their use in bioethanol production which can help in economical production of bioethanol from lignocellulosic biomass.

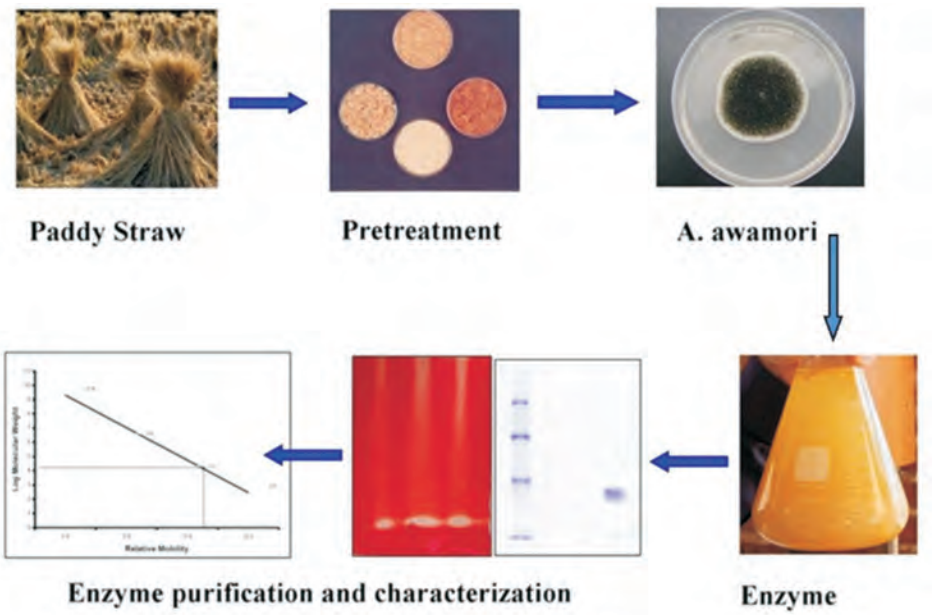
Xanthomonas axonopodis punicae strains (causal organism of pomegranate blight) was explored for the production of cellulase and xylanases for use in saccharification of lignocellulosic biomass.Out of 20 *Xanthomonas axonopodis punicae* strains screened, 10 strains showing better growth on mineral medium containing swollen cellulose, xylan and also producing better zones of hydrolysis on CMC (Carboxy methyl cellulose) agar plates were selected and further evaluated for levels of production of cellulases and xylanases in submerged cultures with CMC as C source. Strain 94 and 96 gave high specific activities of FPase (exo- β-1, 4-glucanase), CMCase (endo-β-1, 4-glucanase), xylanase and cellobiase (β-D-glucosidase). Strain 94 produced high FP activity while higher levels of CMCase and xylanase were produced by strain 96. Optimization for production of cellulolytic and xylanolytic enzymes in both the strains with respect to different nutrient sources and physicochemical factors showed CMC to be the best inducing carbon sources for FP Activity, CMCase and xylanase production while highest production of cellobiase was observed with Starch as carbon source in both the strains.Physicochemical characterization of enzyme activities showed that FP activity, CMCase and cellobiase had temperature optimum around 55^0C in both the strains while xylanase had highest activity at 45^0C and declined as temperature increased. CMCase showed highest pH optimum 8 in both the strains. FPase, xylanase and cellobiase also showed highest activity at pH 5 and 8 in both the strains showing more isoforms. Xylanase and cellobiase were found to be more thermostable than FP activity and CMCase in both the strains. Saccharification studies on three different substrates namely biologically pretreated paddy straw, Filter paper and α-cellulose with substrate loading ranging from 1-10%, showed that sugar yields from filter paper at 1% loading were higher than at 5% loading. Sugar yields increase with time of incubation except in Filter paper with 1% loading which otherwise showed highest yield at 72 hrs. Biologically pretreated paddy straw showed high level of total sugar yields due to synergistic action of both cellulases and xylanases present in the enzyme extract and xylanases released xylose from hemicelluloses content in paddy straw which contributed to sugar yields. The result of this study revealed that *Xanthomonas axonopodis p.v punicae* strains produce higher level of cellulases and xylanases with better characteristics i.e thermal stability, better pH and temperature optima which can be used for supplementing enzyme cocktail for bioethanol production from lignocellulosic biomass.

β-glucosidase producing yeast *Rhodotorula glutinis* was isolated from decaying vegetables. The β-glucosidase enzyme was constitutively expressed on the cell surface. Addition of surfactant to the culture medium or sonication could not release cell-associated β- glucosidase enzyme. While cellulose and glucose induced high levels of β-glucosidase activity, unusual stimulation of β-glucosidase production was observed with cellobiose and soybean meal additive in minimal medium. The enzyme had temperature optimum of 50˚C and pH 6.0-6.5 and showed high glucose tolerance ability as 38.62% activity was retained even at 1.2 M glucose concentration. Culture medium for β-glucosidase production was optimized using Response Surface Methodolgy (RSM) with Box-Behnken design. The optimised predicted values for the three responses: extracellular enzyme activity- 0.034 IU/ml; extracellular specific enzyme activity- 0.319 IU/mg of protein; cell associated enzyme activity- 9.251 IU/ml were obtained. Thus, R. glutinis could be a potential gene source of β-glucosidase with desirable properties to be exploited in biomass hydrolysis.

Xylooligosaccharides (XOs) are oligomers of β-1, 4 linked xylose units and are also considered as prebiotics when consumed as a part of diet. XOs can be produced from corncobs, a low cost lignocellulosic biomass, by enzymatic hydrolysis of xylan. Considering this, a total of eleven xylanolytic fungi were isolated from degraded corncobs collected from different sites and all of them were found to produce xylanase, however, fungal isolate SKF-4, identified as *Aspergillus fumigatus* that produced highest level of β-endoxylanase (143.03 IU/ml) and lowest β-xylosidase (0.01 IU/ml) was selected for optimization of xylanase production. It was found that *Aspergillus fumigates* SKF-4 could produce higher endoxylanase with wheat bran as carbon source, peptone as nitrogen source at pH 5.0 and temperature of 30°C. Response surface methodology with rotatable

central composite design for optimization of parameters could enhance the endoxylanase production by 1.5 times. Physico-chemical characterization of partially purified endoxylanase from *Aspergillus fumigates* SKF-4 showed maximum endoxylanase activity at pH 5.0 and at 45°C. Alkali pretreatment of corncobs was found to be most suitable followed by steam and acid pretreatment for xylooligosaccharides production. Optimization of process parameters for enzymatic hydrolysis of alkali pretreated corncobs led to 4.2 time higher xylobiose production than the unoptimized conditions.

Four microorganisms viz. *Aspergillus awamori, Phanerochaete chrysosporium, Trichoderma reesei* and *Streptomyces aburaviensis* were screened for their potential to produce hydrolytic enzymes especially cellulase on paddy straw and sugarcane bagasse through solid state fermentation (SSF). The cellulase was purified from the crude filtrate and characterized. *Aspergillus awamori* among microorganisms and paddy straw between the two substrates showed significantly higher activities for most of the enzymes studied.



Endoglucanase from *Aspergillus awamori* was purified which showed pH optimum of 4.0 and temperature optimum of 60 °C. The purified monocomponent cellulase produced in the study comparatively has lower Km value, active over broad range of pH (4-9) and is thermostable which indicates its potential for commercial use in detergent and textile industry where monocomponent enzyme formulations of fungal endoglucanases are preferred.

Five *Aspergillus* strains were screened both qualitatively and quantitatively for xylanolytic and cellulolytic activities and hyperxylanolytic strains *A. awamori* F 18 and *A. foetidus* MTCC 508 were selected on the basis of high specific activity (IU/mg protein) of xylanase under submerged and solid state fermentation. Xylanase was produced by these two strains under solid state fermentation using corn cob, paddy straw and mixture of paddy straw and wheat bran (1:1 ratio). *A. awamori* F 18 was most efficient in producing xylanase with specific activity 456.19 IU/mg protein, under solid state fermentation. Xylanase produced by *A. awamori* F 18 was concentrated (11.5 fold) by acetone precipitation method. Xylanase had optimum pH 6.0 and temp. 45 °C. Steam pretreated paddy straw was used as substrate to study the effect of this xylanase supplementation on sugar yields. Supplementation of xylanase to standard cellulases yielded significantly higher amount of sugars than standard cellulases alone.

Soil Microbiology

Exopolysaccharides (EPS) producing bacteria were isolated from the rhizosphere of wheat, maize and mung bean grown under rainfed condition and were screened *in vitro* for moisture stress tolerance in nutrient broth supplemented with different concentrations of polyethylene glycol (PEG 6000). Total 26 isolates (9 isolates from wheat, 7 isolates from maize and 10 isolates from mung bean rhizosphere) were found to tolerate maximum level of stress (45% PEG 6000) in broth culture. These isolates were monitored for the amount of EPS produced under maximum level of polyethylene glycol. The isolate G7, Maize

-5, and Mung-9 were found to be high EPS producing isolates among the wheat, maize and mungbean isolates respectively. These isolates were identified as *Pseudomonas* sp, *Bacillus cereus* and *Microbacterium resistens* respectively on the basis of morphological, biochemical, and 16S rDNA sequence analysis. Exopolysaccharides quantified from *Pseudomonas* sp, *B.cereus* and *M. resistens* was 2.62, 1.93 and 2.30 gm/gm protein biomass respectively. The characterization of EPS by the FT-IR, GC-MS revealed qualitative and quantitative differences in chemical composition. In a pot experiment conducted to elucidate the effect of inoculation with *Bacillus cereus*, *M. resistens* and *Pseudomonas sp.* on the soil physical properties in the rhizosphere of *Triticum aestivum* L. resulted in a significant increase in the root-adhering soil (RAS) dry mass (dm) per root dm (RAS/RT).The increase recorded due to *M. resistens* inoculation at -0.55 MPa hydric level after 20 DAS in RAS/RT was ~ 157 % over the uninoculated control. Intense colonization of the wheat rhizosphere by *Pseudomonas* sp. was also associated with significant increase of mean weight diameter (MWD) and water stable aggregate (>250µm). Scanning electron microscopic studies showed the formation of biofilm of inoculated bacteria on the root surface and this, alongwith a better soil structure, might have protected the plants from the water stress. The study demonstrated that *Pseudomonas* sp. is able to alleviate soils water stress and that EPS-producing bacterial populations play an important role in the rhizosphere through their contribution to soil aggregation.

Rock phosphate is an important natural material traditionally used for production of phosphorus fertilizers. Compared with chemical processing, biological extraction of P from low grade rock phosphate is an alternate environment friendly approach. In this context, sugarcane press mud, citrus fruit peel and paddy straw were evaluated as substrate for fungal mediated solubilisation of Mussoorie, Udaipur and Jabhua rock phosphate under solid state fermentation (SSF). Paddy straw was observed as the best substrate for fungal mediated solubilisation of rock phosphate. Supplementation of paddy straw with nitrogen source in the form of urea, ammonium sulphate or ammonium nitrate exerted an inhibitory effect on release of soluble P from rock phosphate. However, amendment of paddy straw with wheat bran and citrus peel each added separately improved the release of soluble P from Mussoorie and Udaipur Rock Phosphate by > 2 fold compared to un-amended substrate. Subsequent introduction of final fermented product into soil-plant system promoted the wheat plant growth and P acquisition by both straw and grain. The availability of soil P was almost doubled compared to recommend dose of P fertilizer. The study showed that adopting SSF technology for microbially mediated solubilisation of rock phosphate is a promising alternate to obtain soluble P at low cost with minimal environment damage.

Ammonium oxidation is the first rate-limiting step in nitrification. As the ammonia-oxidizing bacteria (AOB) are difficult to culture using the laboratory based culture methods, the molecular characterization of the ammonia-oxidizing microorganisms using the 16S rRNA genes and the functional gene of ammonium monooxygenase (*amoA*) was undertaken. The potential ammonium oxidation activities at the mid-crop stage ranged from 0.08 to 0.15 µg NO_3-N g^{-1} soil h^{-1}. The ammonium oxidation activity was highest at the mid-crop stage in the rhizosphere soils of T2 (RDF) under all three modes of cultivation such as the conventional, system of rice intensification (SRI) and direct seed rice (DSR). The abundances of bacterial *amoA* gene varied from 3.18×10^3 to 1.43×10^5 at the mid-crop stage and from 3.29×10^3 to 5.70×10^5 g^{-1} soil at the harvest stage. The SRI mode of cultivation increased the bacterial ammonium oxidation, evidently from the increases in bacterial *amoA* gene copies, relative to the conventional and direct seeded modes of cultivation. The PCR-DGGE profiling of the ammonia oxidizer communities showed the differential influences of three modes of cultivation and the 'rhizosphere effect' in the mid-crop and harvest stages.

A comparative analysis of two epigeic earthworms *E. foetida* and *P. excavatus* gut bacterial communities and their functions were performed, using culturable and un-culturable approaches. The composition of the cultivable gut communities was found to be dominating by Firmicutes (50–60%), followed by Actinobacteria (26.7–33%), and Alphaproteobacteria (5.6–6.7%). Despite exhibiting similar diversity indices and species

richness, Betaproteobacteria (6.7%) and Gammaproteobacteria (11.1%) were solely present in *E. foetida* and *P. excavatus*, respectively.

Functional characterization revealed that β-glucosidase activity was most prevalent in the culturable microbiome. The analysis of non-cultivable earthworm's gut bacteria was performed by preparation of 16SrDNA clonal libraries and its annotations by MG-RAST, RDP and Metagenassist servers, the results revealed that microbiomes were constituted mainly of Proteobacteria (38-44%), followed by unclassified bacteria (14-18%) and Firmicutes (9.3-11%). Functionally a high abundance of xylan degraders (12.1-24.1%) were present in both the metagenomes. Lignin degradation was detected in 3.7% clones of *E. foetida*, while cellulose degraders represented 1.7%. The mapping of the earthworm gut microbial communities gave insights into a very important micro-ecological niche which can serve as the treasure trove for the researchers to dig for many novel industrially important enzymes. Vermicomposting efficiency of both the earthworms was also evaluated for composting of crop residues (wheat straw and paddy straw) amended with farm yard manure. Mature vermicompost produced during the study had significant increase in total nitrogen (71 -150%), phosphorus (49 %–116%) and potassium (26.3–142%), along with decrease in organic carbon in different experimental vermibeds. Maximum total nitrogen as well as available phosphorous concentration was observed from vermibeds inoculated with *E. foetida*, whereas increased exchangeable potassium was recorded in vermicompost produced by *P. excavatus*. Enhanced hydrolytic enzyme activities of CMCases (1.51 folds), Fpases (2.11 folds) and β-glucosidases (1.38 folds) were recorded in treatments vermicomposted with *P. excavatus*. The mean individual live weight, growth rate (mg wt. worm-1 day-1) of earthworm were higher for *P. excavatus* as compared to *E. foetida*. Overall, *P. excavatus* exhibited better growth and mineralization efficiency, which further support the suitability of the species for large scale vermiculture.

Algology

Genetic diversity of cyanobacteria in organic farming field under rice and wheat cropping system was investigated. Twenty five different cyanobacteria isolated from soil samples taken during rice and wheat crop were analyzed for genetic diversity using Randomly Amplified Polymorphic DNA-Polymerase Chain Reaction (RAPD-PCR). A total of seventeen single oligonucleotide primers were used to generate RAPD patterns for twenty five different cyanobacterial isolates. A total of 519 DNA fragments (DNA bands) were obtained for nine isolates belonging to the genus *Nostoc*. The percentage of polymorphism observed was 92%. The primers CRA-22, D-02 and OPD-20 gave maximum number of polymorphic bands. Similarly, a good polymorphism could be generated in *Anabaena* isolates where 427 polymorphic bands were obtained using seventeen primers. Among all the primers Hip-GC and OPD-20 gave maximum number of polymorphic bands. OPD-20 seems to be a useful primer for molecular analysis of cyanobacteria as it provided maximum polymorphism as compared to other primers. The dendrogram analysis for RAPD-PCR divided the isolates based on their isolation i.e. during rice crop and indicated a high degree of distinctness among isolates belonging to different genus.

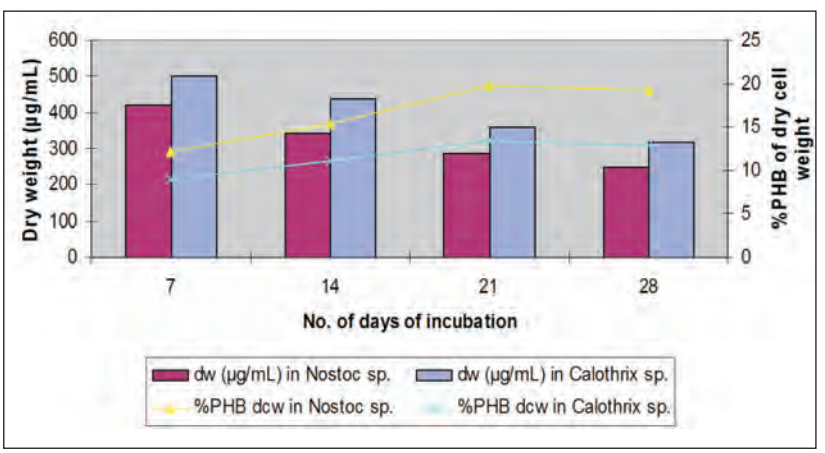
Several cyanobacterial strains ®belonging to *Anabaena* sp. *Nostoc* sp. *Westilopsis* sp. and *Phormidium* sp. were isolated and identified from the soil samples collected from different sites of the biodiversity hotspot Rann of Kutch. Their morphological identity was further confirmed by 16S rDNA analysis and BLAST analysis established homology and close similarity with cultured cyanobacterial species. The higher value ($r = 0.75$) obtained by regression analysis between electrical conductivity (EC) and number of cyanobacteria shows the highest influence of EC on the cyanobacteria. The studies showed that the distribution of cyanobacteria is correlated to salinity and the level of soil salinity is a major factor that determines the distribution and diversity of cyanobacteria in a larger context.

Sixteen cyanobacterial strains from genus *Spirulina* and related genera were procured from culture collection of CCUBGA, IARI, New Delhi-110012. The length of the vegetative cells varied; and a wide range was recorded in chlorophyll content, total soluble proteins and glutamine synthetase activity. The protein profile revealed distinct polymorphism with fourteen polymorphic bands. Some of the strains showed

distinct pattern which could serve as fingerprint for the strains under investigation. The dendrogram based on analysis of combined dataset revealed two major groups. The two *Phormidium* strains and *Lyngbya* strains exhibited 100% similarity on the basis of protein profile dataset. Extracted DNA was subjected to RAPD-PCR and 16S rRNA gene amplification. Out of the total bands scored, 100% distinct polymorphic bands were observed in RAPD reaction with single oligos and the number of polymorphic bands ranged from 9 to 28. A combination of 10 sets of dual primers generated 100% distinct polymorphic bands and the number ranged from 13 to 32. RAPD study revealed unique bands in most of the reactions. Clustering analysis divided strains into two main clusters, with single primers as well as combination of primers. It was interesting to observe that when RAPD data was represented in the form of UPGMA dendrogram using dual primer combinations or developed with single and dual primer combinations together, *Spirulina lonar* and mutant of *S. platensis* showed a very close similarity. In 16S rRNA gene amplification study all the strains produced a single PCR product of about 1500bp. The amplified PCR fragment was digested with different restriction enzymes. Two restriction enzymes namely *Hae*III and *Rsa*I revealed 100% polymorphism when used singly followed by *Mse*I (87.5%) and *Eco*RI (75%) respectively. The number of bands with *Eco*RI and *Mse*I was 4 and 8, with one monomorphic band observed in each case. Restriction enzymes (*Hae*III and *Rsa*I) produced 13 and 11 polymorphic bands. All the sixteen strains of *Spirulina* and related genera were grouped into two main clusters with RFLP analysis of 16S rRNA.The partial 16S rRNA gene sequence of *Spirulina* and other related genera were submitted to the NCBI GenBank and their relationship was determined.

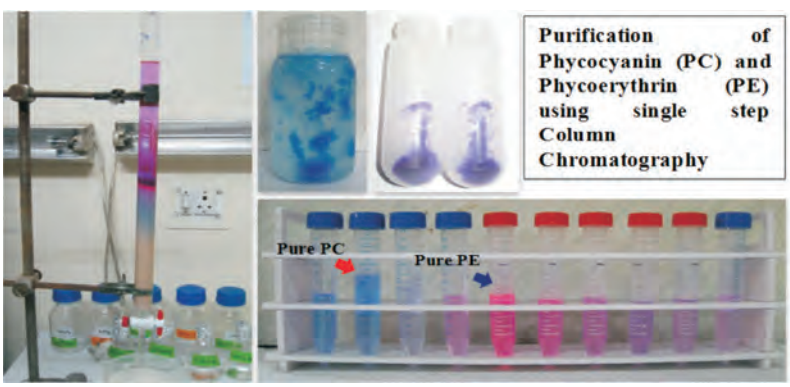
Thirty cyanobacterial strains with optimum growth pH ranging from 4 to 6 were isolated from Eastern, Northeastern and Southern part of India. The low pH tolerance study indicated that *Nostoc* AD1, *Nostoc* AD2 and *Anabaena* AD3 from West Bengal showed a pH tolerance of 4, 4.5 and 5; *Nostoc* ER1, *Anabaena* ER2 and *Nostoc* ER3 from Kerala; and *Nostoc* MK1, *Anabaena* MK2 and *Cylindrospermum* MK3 from Nagaland depicted a pH tolerance of 4.5, 5 and 5.5. These cyanobacterial strains exhibited variation in terms of pH tolerance, growth, pigment profile and other cellular constituents. Of these, four strains belonging to *Nostoc* having better low pH tolerance ability were used for further study under control (pH 7) and acidic pH (4, 4.5) grown conditions. Microscopic observations showed highly mucilaginous mats, formation of cell aggregates, reduced cell size and heterocysts frequency under acidic conditions in comparison to control. The growth rate was also reduced significantly under acidic conditions. There was an enhancement in chlorophyll, carotenoids, phycobilins and cellular constituents under acidic conditions compared to control; however, ammonia excretion was more in control grown cultures. The *in vitro* arginine deiminase (ADI) and anabolic ornithine carbamoyl transferase (OCT) activities were more under acidic as compared to control grown cultures. The amplified *arg*F gene sequences exhibited 88% similarity with reported *arg*F sequence of *Nostoc* PCC 73102. The *arg*F expression was more under acidic than in the control grown conditions, with higher expression depicted by stationary phase cultures. The FAME profile of acid stress tolerant cyanobacteria showed higher percentage of SFA as compared to UFA and MUFA.

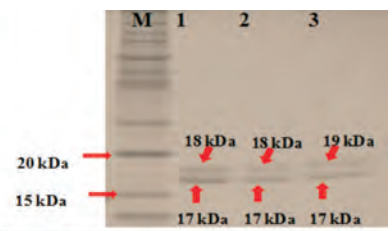
Cyanobacteria were tested for poly-β-hydroxybutyrate (PHB) accumulation. All the strains tested showed varying amounts of PHB accumulation with *Nostoc* sp. (CCC 37) and *Calothrix* sp. (CCC 38) recording maximum accumulation. The heterocystous organisms produced PHB upto 11% and 10% (w/w) of dry cell weight respectively when grown photoautotrophically. The PHB accumulation enhanced to 15-20% or 17-24% in *Nostoc* sp. and 12-19% or 12-16.4% in *Calothrix* sp. after 21 days



in mixotrophy and chemoheterotrophy conditions respectively with varying concentrations (0.1, 0.2, 0.3 and 0.4%) of glucose, fructose, maltose and acetate. Presence of acetate resulted in maximum PHB accumulation in *Nostoc* sp. whereas *Calothrix* sp. recorded maximum PHB in presence of fructose followed closely by acetate. PHB accumulation in both the strains also increased after phosphate starvation.

Three cyanobacteria *viz. Nostoc commune* (CCC391), *Anabaena variabilis* (CCC421) and *Anabaena oryzae* (CCC162) were studied for phycobiliprotein production. Maximum production of total phycobiliprotein (PBP) as well as individual fractions *viz.* phycocyanin (PC), phycoerythrin (PE) and allophycocyanin (APC) in all the three cyanobacteria was recorded at 14 days after inoculation. Extraction using Acetate buffer (pH 5.1) and 65% saturation of $(NH_4)_2SO_4$ for precipitation of PBPs proved superior over other methods. PC from all three cyanobacteria and PE from *N. commune* were purified following a single step anion exchange chromatographic method using DEAE-Cellulose. 46.9%, 36.2% and 41% PC with purity of 3.99, 2.75 and 4.75 was recovered from *N. commune*, *A. variabilis* and *A. oryzae* respectively after chromatographic separation. 41% PE with a final purity of 6.37 was recovered from *N. commune*. Subunits of PC and PE were separated by SDS-PAGE and their identity was confirmed by MALDI-TOF. α and β subunits of PC from *N. commune*, *A. variabilis* and *A. oryzae* were of 17 & 18 kDa, 17 & 18 kDa and 17 & 19 kDa respectively while α and β subunits of PE purified from *N. commune* were having molecular weights of 14 kDa and 18 kDa respectively. PC from all the cyanobacteria showed maximum absorbance at 620 nm while PE had an absorption maximum at 570 nm. A C_5 column based simple HPLC method was also developed in this study for separation of subunits of PC. Studies on relative expression of *cpcB* gene in these cyanobacteria showed that the *cpcB* gene expression decreased at 0.6 ppm and 1.8 ppm iron as compared to control (1.2 ppm Fe from ferric ammonium citrate) irrespective of source. 1.2 ppm iron as ferric ammonium citrate was found to be superior over the other sources of iron and their concentrations with respect to phycocyanin production. Under a light intensity of 65 µmole photons/m^2/s, *cpcB* gene in all the three cyanobacteria was up regulated and phycocyanin production increased significantly in *viz. N. commune* and *A. oryzae* but interestingly in *A. variabilis*, *cpcB* gene was found to be up regulated even under the light intensities of 35 & 45 µmole photons/m^2/s. PC production as well as relative expression of *cpcB* gene increased in *N. commune* and *A. variabilis* when grown with 10 mM NaCl. But both phycocyanin production and *cpcB* gene expression decreased significantly at 50 and 100 mM NaCl.





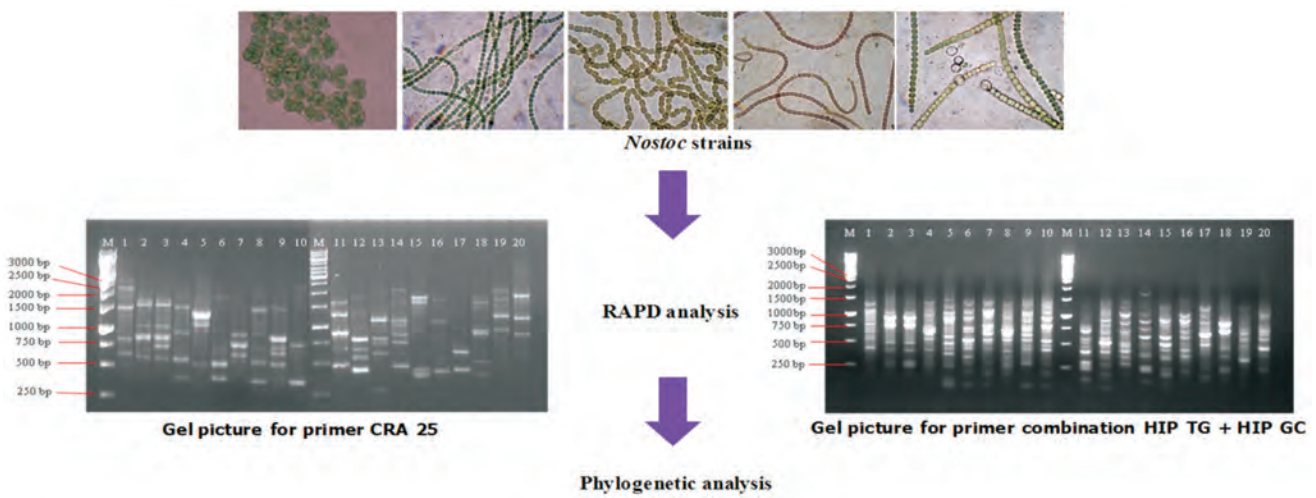
Separation of PC sub-units using SDS-PAGE
M: Protein molecular weight marker; 1: PC purified from CCC391; 2: PC purified from CCC421; 3. PC purified from CCC162

Water samples were collected from Anasagar, Pushkar lake and Sambhar lake located around Ajmer district of Rajasthan, India. The pH of the water samples was alkaline and the Electrical Conductivity of Samhbar lake was highest and that of Anasagar was lowest. The exchangeable Na, organic carbon, available phosphorus and nitrogen of Sambhar lake was maximum. Most of the heavy metals could not be detected in Anasagar and Pushkar lake and Sambhar lake showed the presence of all the heavy metals analyzed. Out of the total twenty three cyanobacterial isolates, twelve were from Anasagar, eight were from Pushkar lake and only three were from the Sambhar lake. Microscopic examination indicated the presence of one colonial, nine heterocystous and the remaining non-heterocystous filamentous forms. When genera wise distribution was recorded, *Nostoc, Phormidium* and *Plectonema* were most abundant followed by *Anabaena* and *Westiellopsis*;and *Microcystis* and

Oscillatoria were least abundant. There was a significant difference in chlorophyll, carotenoids and phycobilins amongst the cyanobacterial isolates. The phycocyanin, allophycocyanin and phycoerythrin differed significantly and phycocyanin/ phycoerythrin ratio was more than one in most of the cases. The nitrate reductase (NR) and glutamine synthetase (GS) activity differed significantly and the nitrogenase activity could be detected only in heterocystous strains.16S rRNA gene amplification depicted the presence of single amplified product of 1500bp. The digestion with restriction enzymes (*Dpn* II, *Mse* I and *Hin* f) exhibited differential restriction profile. Results on the basis of dendrogram developed indicated closeness of genera from the same order in most of the cases with few exceptions. Cyanobacterial strains *Plectonema abornema, Phormidium molle, Phormidium foveolarum* and *Phormidium ceylanicum* clustered together with 100% similarity.

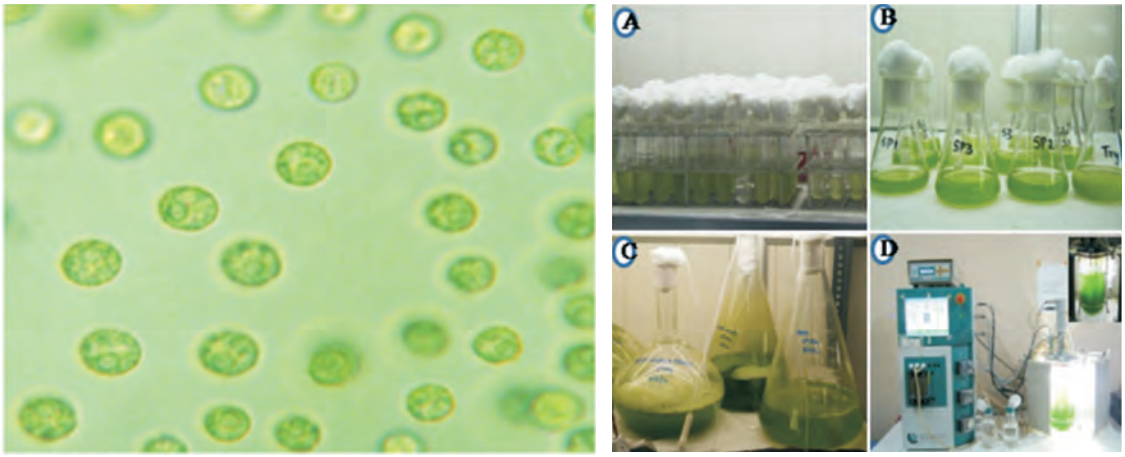
Phylogenetic analysis of *Nostoc* strains using RAPD

In the present study, RAPD was carried out for fingerprinting and phylogenetic analysis of twenty *Nostoc* strains from different geographical origin. Twelve single primers and ten dual primer combinations were used for this purpose. Both the single primers and dual primer combinations produced strain specific unique bands which will be useful for identification of those strains. For sixteen strains, out of twenty, strain specific bands were generated. Cluster analysis revealed a vast heterogeneity among these *Nostoc* strains and no clustering based on geographical origin was found. It was also observed that morphological data do not necessarily correspond to the genetic data in most of the cases. Fitness of the clusters were good in most of the cases while the bootstrap values were not very high except a few clusters which may be due to high level of heterogeneity among the strains tested. From the study it was concluded that a vast heterogeneity existed among the strains of *Nostoc* tested and molecular tools like RAPD is efficient enough for phylogenetic analysis of these organisms. The DNA profiles generated for the *Nostoc* strains and certain species specific unique bands produced by different single and multiplex combinations will be helpful for developing identification tools for those strains.



Role of supplemental calcium in relation to salinity tolerance in the *Anabaena* sp. ccc 441

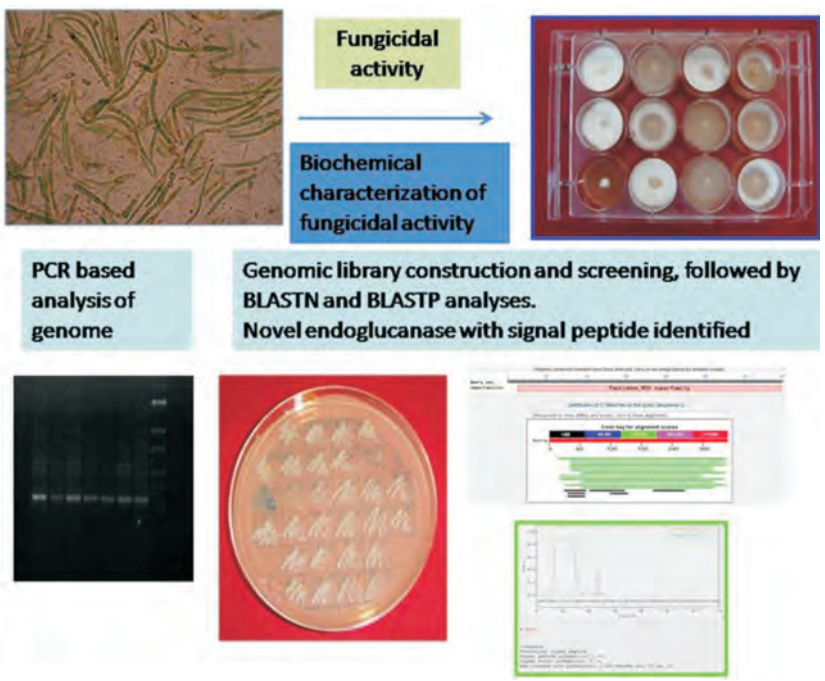
Cyanobacteria contribute to the improvement in the productivity of rice paddy fields but increasing soil salinity restricts their growth. Supplementation of calcium has been reported to alleviate the inhibitory effect due to salinity stress. Therefore in the present study supplementation of calcium in the growth and physiological attributes of the freshwater cyanobacterium *Anabaena* sp. CCC 441was studied. Supplementation of calcium in the salt stressed cyanobacteria improved growth, activity of the enzymes of nitrogen assimilation, antioxidant enzymes and helped in the maintenance of proper Na^+/K^+ ratio. The results suggest that ameliorative potential of calcium on the salt stressed cyanobacterium through the modulation of antioxidants enzymes, ion transport and expression of specific salt stress proteins.



Biochemical engineering of *Chlorella sorokiniana* for enhanced lipid accumulation

The study focused towards screening the modes of nutrition and use of metabolic intermediates along with reducing agents for enhancing the quality and quantity of lipids in *Chlorella sorokiniana*under standard laboratory conditions and its upscaling in a photobioreactor for enhanced lipid productivity. Time course studies using glucose under mixotrophic and heterotrophic conditions vis a vis autotrophy revealed highest values of growth, lipid productivity (29.33% DCW) and lipid content (755. 00 mg/L) under mixotrophic conditions on the 14th day of growth. Upscaling in a photobioreactor with selected substrates (Glucose and tryptophan), along with sodium thiosulphate (ST) as a reducing agent led to lipid productivity of 34.16 and 36.49% after 4th and 8th day respectively in both substrates, vis a vis 11% with glucose alone. FAME analyses revealed a 2% enhancement in total saturated fatty acids (SFA) in tryptophan supplemented culture, which also exhibited an almost ideal fatty acid ratio for use as biodiesel (2.6:4:1 of 16:1.18:1 and14:1). *Chlorella* sp. grown in Glucose + ST produced significant amount of linoleic acid (18:2) and α-linolenic acid (18:3). This strain proved to be a very useful strain with metabolic versatility and multifaceted uses.

Cyanobacteria are commonly used as biofertilizers, but characterization of their potential as biocontrol agents is less investigated. The present study was undertaken for identifying and characterizing gene(s) involved in the production of biocidal compounds in *Calothrix elenkinii* exhibiting fungicidal activity against *Pythium* spp. under *in vitro* and *in vivo* conditions. Chemical analyses of ethyl acetate fraction by LC-MS and NMR revealed the presence of 3-acetyl-2-hydroxy-3-methoxy-4-methyl benzoic acid. PCR based analyses for various cyanotoxins and fungicidal compounds revealed similarities with antibiotic synthetic genes and polyketide synthases which highlight the diversity in the production of metabolites by this strain. The strain was also observed to

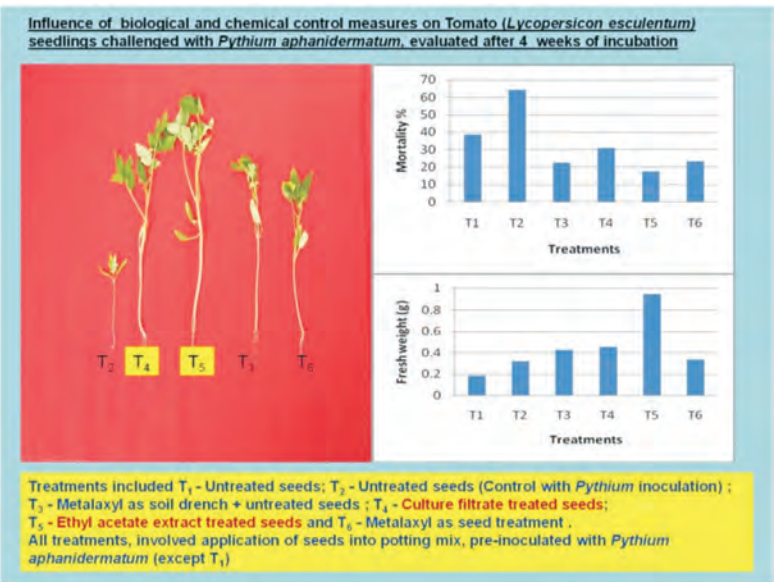


secrete cell wall hydrolases such as chitosanase and endo β-1,4-glucanase. Development of a genomic library, followed by sequencing of the positive clone for both endo β-1, 4-glucanase activity and fungicidal activity against *Pythium aphanidermatum* revealed an ORF of 1.044 kb, encoding 348 amino acid residues (predicted molecular weight of 38 kDa). This showed 97% similarity with glucanase belonging to peptidase M20 family of *Anabaena variabilis* and *Nostoc* sp. PCC7120. BLASTP analyses showed 23-26% similarity with known aminopeptidases or endoglucanases of microbial origin. Putative promoters, ribosomal binding site and a putative signal peptide of 22 amino acid residues were identified in this strain. This is the first report on a novel active endoglucanase and a substituted benzoic acid and its correlation with fungicidal activity in a cyanobacterium.

Different aquatic sampling sites from Dalhousie and Chamba districts of Himachal Pradesh (Northern India) were identified for the collection of samples and isolation of microalgae from the chlorophyceae. Total ten micro-algal strains from five genera namely *Chlamydomonas* ;*Chlorella*; *Chlorococcum* ; *Scenedesmus* and *Ulothrix* were isolated and identified based upon microscopic observations and these were used for further studies. There was a gradual and continuous enhancement in the dry weight with incubation time in days. Mean dry weights varied from a lowest in *Chlamydomonas* to the highest in *Scenedesmus*. The chlorophyll content ranged from the highest in *Scenedesmus* to the lowest in *Ulothrix*. The strains differed significantly with respect to total soluble proteins and *Chlorococcum* showed the highest total soluble proteins and *Ulothrix* showed the lowest content. A lowest of total lipids (5.8%) was recorded in *Ulothrix* and highest (18.7%) was recorded in *Chlorella*.

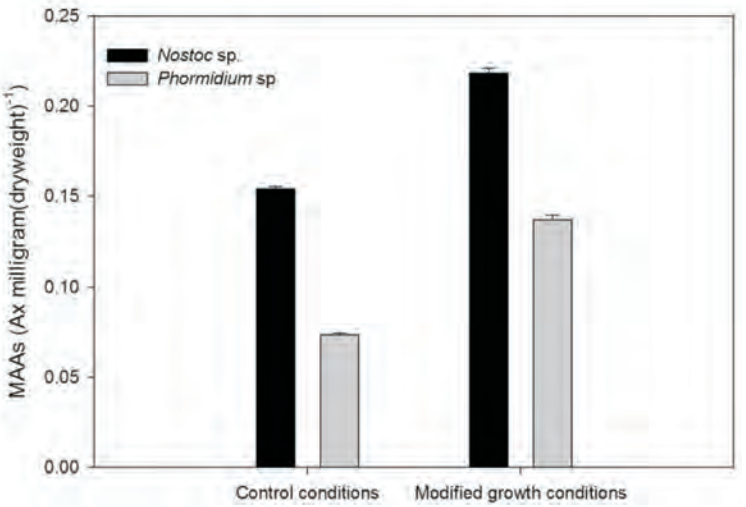
Three microalgal genera namely *Chlamydomonas, Chlorococcum* and *Chlorella* were selected for studies on the optimization of cultural conditions for enhanced lipid production. These were subjected to variable environmental and cultural conditions for a period of 28 days and the differential variables used were temperature , light intensity, pH, sodium chloride and 0.25 g N L^{-1} as $NaNO_3$, NH_4Cl and Urea. Highest lipids were exhibited by *Chlorella* in comparison to *Chlamydomonas*and *Chlorococcum* under different temperatures , light intensities, nitrogen source as well as sodium chloride concentration. The differential effect of environmental variables on total lipids could be recorded. Maximum lipids were recorded at a pH of 7.2 as compared to the pH of 6 and 6.6. The results clearly indicated that the optima for lipids varied in different microalgal cultures at different concentrations of sodium chloride. Fatty acid profile showed the presence of saturated as well as unsaturated fatty acids in three micro-algal strains tested in the present study. Results have clearly indicated that % of total FAME can be obtained from the lipid extract under normal growing conditions. FAME profile was found to be suitable for biodiesel and *Chlorococcum* and *Chlorella* can be considered as promising microalgae to be used as a source of lipids for biodiesel production.

Pythium blight and damping off are among the most common diseases of vegetables & despite the availability of chemical control measures (seed/soil treatments with Metalaxyl, Thiram), the destructive nature of this disease is still significant in terms of economic losses incurred. Investigations were undertaken to evaluate the fungicidal potential of metabolites of cyanobacterium*Calothrix elenkenii* against *Pythiumaphanidermatum* in selected vegetables. Nursery stage evaluation of biocontrol efficacy of culture filtrate and ethyl acetate extract of *Calothrix elenkenii* against *Pythium aphanidermatum* challenged vegetable seeds (Tomato, Chilly, Brinjal) was undertaken at the National



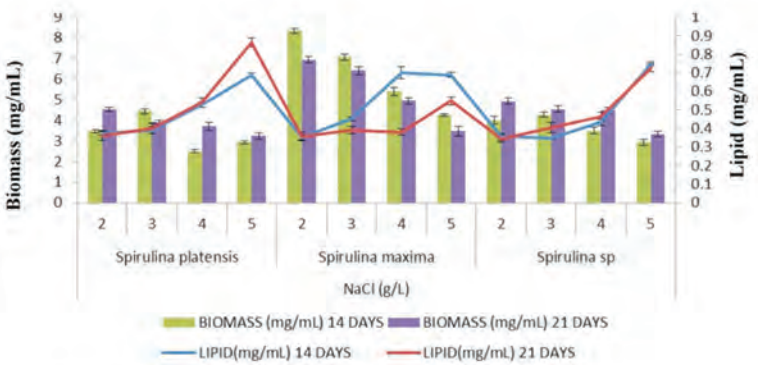
Phytotron Facility, ICAR-IARI, New Delhi. Seeds treated with Ethyl acetate extract of *Calothrix elenkenii* culturefiltratesrecorded significantly higher or statistically at par values with Metalaxyl (chemical control) treated seeds for all the crop parameters. Another interesting conclusion that would be drawn from the observations was that the ethyl acetate extract of *Calothrix elenkenii* was able to not only act as a biocidal agent against *Pythium aphanidermatum,* but was also stimulatory towards growth of the seedlings of tomato, chilly and brinjal. ANOVA revealed that the treatment and crop exhibited significant influence on all the parameters analyzed.

Characterization of cyanobacteria from extreme environment is important for the identification of biotechnologically important compounds. Isolation of soil samples from Rann of Kutch followed by morphological attributes and 16S rDNA analysis led to the identification of *Nostoc* sp (YPR-9) and *Phormidium* sp. (YPR-3). These two species of cyanobacteria were found to efficient producer of mycosporine like amino acids. Mycosporine like amino acid production in these cyanobacteria could be augmented by modifying the growth and culture condition variables such as pH, temperature, salinity and the nitrogen sources.



Liquid biofertilizer could be a possible solution to address the problems of carrier based preparations. Liquid formulations for BGA biofertilizer were prepared for the first time using a composite culture of *Anabaena variabilis (CCC441), Nostoc muscorum (CCC442), Tolypothrix tenuis (CCC443) and Aulosira fertilissima (CCC444),* which have already been used as biofertilizer inoculants in carrier based preparations. Since, BGA biofertilizer is used in paddy that is grown under flooded conditions, the oil based formulations were prepared as oil in water emulsions are easy to disperse into water bases. The formulations remained stable for full period of incubation (nine months) and maintained their physical characteristics of viscosity, pH, suspensability, breaking length and creaming at different storage temperatures of 4°C, 28°C and 40°C. The formulations also showed pseudoplasticity behaviour. The observations on cell count showed that all these formulations maintained very good cell count and maximum viable cells were observed in Formulation 11 and 4. The cultures revived from these formulations also maintained their growth rate and metabolic activity in terms of nitrogen fixation, nitrate reductase and glutamine synthetase enzyme activity which was comparable to control. Overall, the liquid formulation can be a successful option for BGA biofertilizer.

Nutrient limitation in terms of nitrogen and phosphorus increased the lipid accumulation under depleted growth in the *Spirulina* strains. Nitrogen limitation was found more effective than phosphorus in accumulation of lipid in all the three selected strains of *Spirulina*. The fatty acid profile varied differently and palmitic (48.31%), linolenic (20.71%) and linoleic acids (14.63%) were most prevalent. It was interesting that nitrogen limitation was more effective in increasing total lipid content whereas phosphorus limitation had more effect on fatty acid profile. Fatty acid ratios



Growth and Total lipid accumulation in selected *Spirulina* strains grown in Z- medium under salt stress

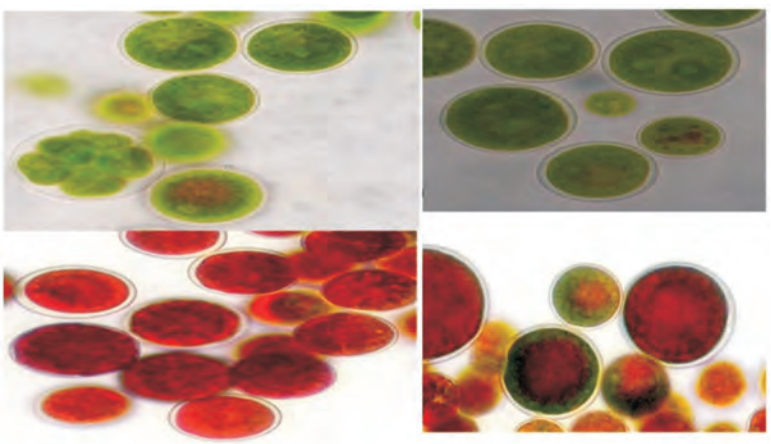
were found to be different under both nutrient limitation conditions in which γ -linolenic acid was accumulated in higher quantities under nitrogen than phosphorus limitation.

Three *Spirulina* strains which were able to withstand salt stress were evaluated for lipid accumulation and fatty acid profile under different salinity levels and incubation period. Presence of salt stress increased the lipid accumulation in selected *Spirulina* strains upto 25.53 %. The fatty acid profile varied differently as palmitic, linolenic, oleic, linoleic and docosahexaenoic acids (in order of abundance) were most prevalent. Fatty acid ratios were found to be different under different salt stress conditions and incubation times in which PUFA and γ-linolenic acid was accumulated in higher quantities at increased salt stress in all *Spirulina* strains.

The cyanobacterial strains were screened for the irability of growth promotion by evaluating the effect of cell free super *natanton* germination and growth of rice (var.Pusa 44) and wheat (var. Pusa 544) seeds. Sixty-six percent strains showed 100% germination in rice whereas 72%strains showed 100% germination in wheat as compared to 70% in control. There was variable response in terms of root growth, shoot growth and fresh weight of seedling in presence of cell free super natants. Preliminary screening led to the selection of 10 cyanobacterial strains, which were characterized for nitrogen fixation, P solubilization, auxin production and side rophore production. Maximum nitrogenase activity was shown by *Hapalosiphon fontinalis* followed closely by *Nostoc commune* whereas *Nostoc* sp. recorded the minimum activity. All the cultures showed P- solubilizing activity with maximum activity shown by *Anabaena* sp. followed by *Nostoc commune*. Indole-3-Acetic acid production was observed in all the strains in presence of varying amount of L-tryptophan. *Nostoc puncti forme showed* the maximum IAA production followed closely by *Nostoc commune*, *Calothrix brevissima* and *Nostoc punctiforme*.The organisms also showed side rophore production with *Nostoc commune* showing maximum production along with *Nostoc* sp. The investigation has revealed the potential of *cyano bacteria* as plant growth regulators and *Nostoc commune* can be considered as potential PGPcyanobacteria and can be evaluated in field trials.

Dry weight, pigment profile, N assimilatory enzymes (nitrogenase, nitrate reductase and glutamine synthetase activity) and photo production of hydrogen differed amongst a set of heterocystous and non-heterocystous cyanobacterial strains procured from CCUBGA, IARI, New Delhi was studied. Nitrogenase activity and hydrogen production enhanced in argon environment as compared to aerobic conditions. Highest nitrogenase activity was recorded in *Nostoc* sp. and lowest was seen in *Plectonema* sp. under anaerobic conditions. *Anabaenadoliolum* showed highest hydrogen production whereas *Phormidium* sp showed the lowest.

Four hydrogen producing, heterocystous nitrogen fixing cyanobacterial strains namely *Anabaena variabilis, Anabaena doliolum, Nostoc* sp and *Nostoc muscorum* were studied for the presence of genes for Hox subunits of bidirectional hydrogenase. Genes coding for larger subunits (*hox*H, *hox*Y) and smaller subunits (*hox*E, *hox*F) of bidirectional hydrogenase were amplified. The results indicated the presence of nickel dependent reversible hydrogenase in all strains taken for the study. Partial *hox*H gene of cyanobacterium *Anabaena variabilis* was cloned and sequenced which indicated that the gene comprised 904 nucleotides. A phylogenetic tree was constructed analyzing the *hox*H nucleotide sequences with the published sequences.

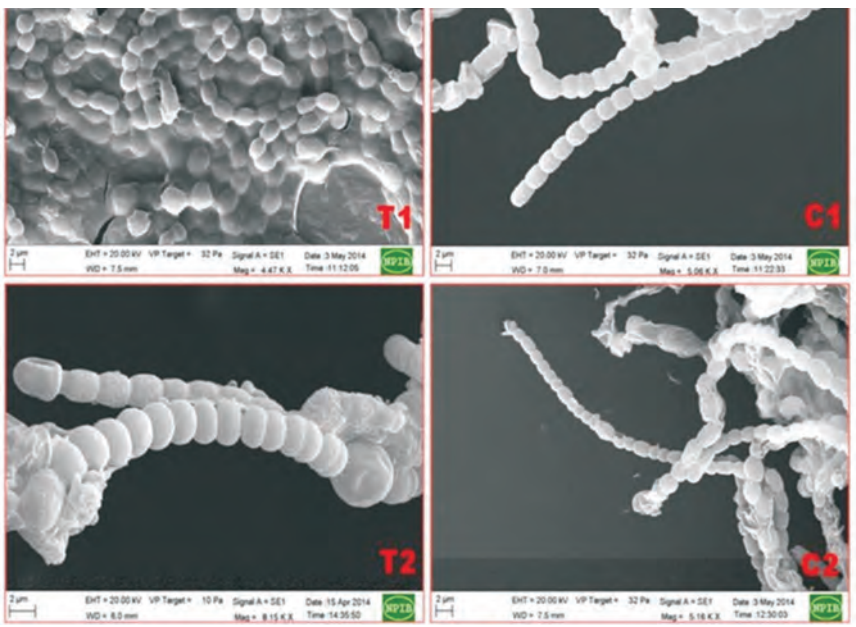


Haematococcus isolate CCCH1 *Haematococcus* isolate CCCH2

Two *Haematococcus* strains were isolated from samples collected from water bodies of Palampur (Himachal Pradesh, India). The isolates were authenticated by microscopic studies and 18s r RNA analysis. Two

Haematococcus standard strains namely *H. pluvialis* UTEX 2505 and *H. lacustris* UTEX 16 were procured from The Culture Collection of Algae at University of Texas, Austin, USA as reference. The astaxanthin accumulating potential of new isolates was studied and the results showed that new isolates accumulated astaxanthin *at par* with the reference strain UTEX 2505. New isolates were exposed to N and P starvation, and to different light intensities to evaluate the influence of these on growth characteristics and pigment formation. There was a significant difference in chlorophyll, carotenoids and astaxanthin contents under N and P experimentation with the highest accumulation of astaxanthin and cellular pigments recorded under optimum N condition than that of N starvation. In P starvation experiments, there was no significant difference between optimum P level and P starved condition. The study showed that the cells were capable of producing astaxanthin even in low phosphate concentrations. High light inhibited growth and pigment yield where as low light yielded high pigment content as that of control. In the experiment to optimize media recipe by addition of ferric citrate and TCA cycle intermediates as carbon source, results showed that ferric citrate and lower concentrations of organic acids like succinic, citric and malic acids showed enhanced astaxanthin accumulation. In order to understand role of plastid terminal oxidase and related proteins in astaxanthin biosynthetic pathway, a study was conducted to determine the presence of *Pgr5* gene which codes for PGR5 protein that associates in cyclic electron flow and helps organism in photo-protection and which is also reported to be present in all the microalgal genomes sequenced so far. Primer designed and product amplified showed nearly 43% similarities with that of *Pgr5* gene of *Arabidopsis thaliana*.

Representative acidic soil samples were collected from three sites namely Alipurduar, Ernakulum and Mokokchung districts of West Bengal, Kerala and Nagaland, India. Thirty cyanobacterial strains were isolated using BG-11 medium with pH ranging from 4 to 6. The low pH tolerance study indicated that *Nostoc* AD1, *Nostoc* AD2 and *Anabaena* AD3 from West Bengal showed a pH tolerance of 4, 4.5 and 5; *Nostoc* ER1, *Anabaena* ER2 and *Nostoc* ER3 from Kerala; and *Nostoc* MK1, *Anabaena* MK2 and *Cylindrospermum* MK3 from Nagaland depicted a pH tolerance of 4.5, 5 and 5.5. These nine cyanobacterial strains exhibited variation in terms of growth, pigment profile and other cellular constituents. Out of nine, four strains belonging to *Nostoc* having better low pH tolerance ability were used for further study under control (pH 7) and acidic pH (4, 4.5) grown conditions. Microscopic observations showed highly mucilaginous mats, formation of cell aggregates, reduced cell size and heterocysts frequency under acidic conditions in comparison to control. The growth rate was also reduced significantly under acidic conditions. There was an enhancement in chlorophyll, carotenoids, phycobilins and cellular constituents under acidic conditions compared to control; however, ammonia excretion was more in control grown cultures. The *in vitro* arginine deiminase (ADI) and anabolic ornithine carbamoyl transferase (OCT) activities were more under acidic as compared to control grown cultures. The activity of these was higher in stationary phase than log phase cultures; OCT activity was more than ADI under both the growth phases. The amplified *arg*F



Scanning electron microphotographs of acid stress tolerant *Nostoc* 1 and Nostoc 2 grown under acidic (T1, T2) and control (C1, C2) conditions

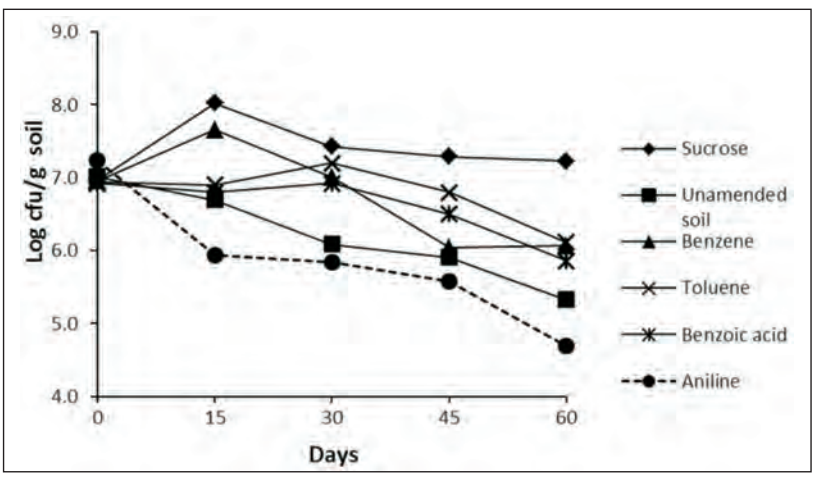
gene sequences exhibited 88% similarity with reported *arg*F sequence of *Nostoc* PCC 73102. The deduced protein sequences from *arg*F gene sequences of two *Nostoc* strains showed 89% and 90% similarity with OCT protein sequence of *Nostoc punctiformis*. The *arg*F expression was more under acidic than in the control grown conditions, with higher expression depicted by stationary phase grown cultures. The FAME profile of acid stress tolerant cyanobacterial strains showed higher percentage of SFA as compared to UFA and MUFA. The protein profile exhibited differential expression of acid stress responsive and acid sensitive proteins at pH 4, 4.5, 5, 5.5 and 7 during log phase as well as stationary phase. These observations and results clearly indicated the usefulness of morphological and /or biochemical attributes in understanding the acid stress tolerance mechanisms in cyanobacteria.

Bioremediation and Other Applications of Microorganisms

Pentachlorophenol biotransforming bacteria (22) from pulp and paper mill effluent irrigated soils of Lal Kuan, Uttrakhand were isolated and identified using molecular tools. The isolated bacteria grew well at 500 ppm of PCP as sole C source in minimal salt medium. However, reduction in their growth was observed at 700 ppm PCP. Out of 22 isolates, 19 isolates belonged to class γ- *Proteobacteria*, matched with sequences of *Pseudomonas* and *Enterobacter.* Two isolates, *Ochrobactrum* sp (LK59) and *Ensifer adhaerens* (LK 4) belonged to α- Proteobacteria. The single isolate of *Firmicutes* phyla was identified as *Lysinibacillus fusiformis (LK156)* which could show significantly higher growth at 300 and 500 ppm of PCP than other isolates. PCP degradation by *Ensifer adhaerens* and *Lysinibacillus fusiformis* is reported for the first time in the present study and these might represent new chlorophenol-degrading taxa.

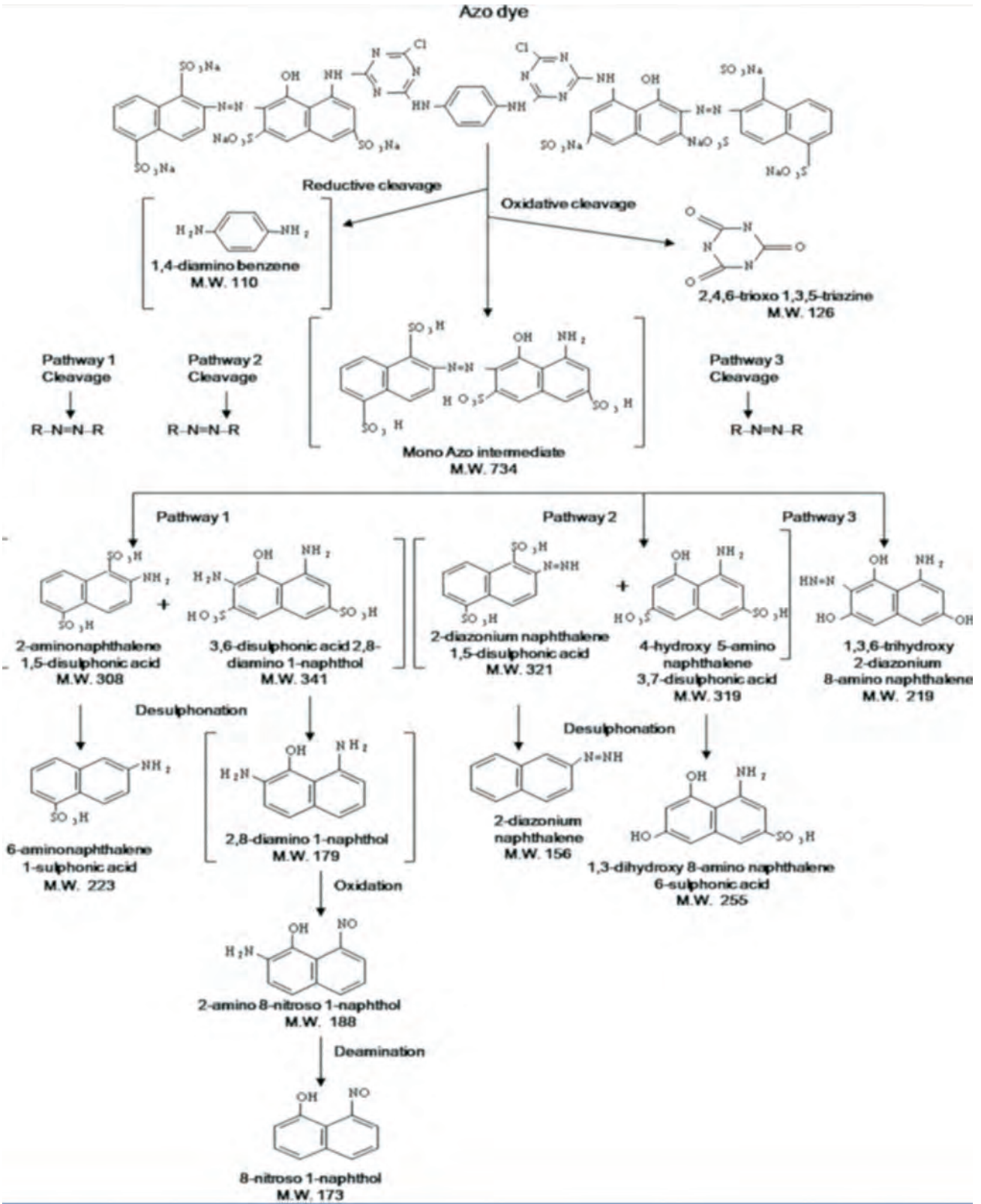
HPLC studies revealed decrease in the residual PCP and formation of intermediates such as tetrachlorohydroquinone (TeCH) and dichlorophenols (DCP) by selected isolates. Removal of PCP from sterile soil by selected isolates, *viz. Ensifer adhaerens, Pseudomonas putida* and *Lysinibacillus fusiformis* , was studied in a 30 d experiment under controlled condition. These isolates were used alone and in combination. PCP removal by mixed inoculum was significantly higher (68.81%) than single inoculation after 30 d of incubation. The potential isolates obtained in the present study can be used for developing consortium for bioremediation of sites contaminated with PCP.

Out of the six morphotypes obtained, which were able to decolourize Red HE7B dye *Bacillus* sp. strain Azo 1 was the most promising. Optimum cultural conditions for maximizing dye decolourization by *Bacillus* sp. strain Azo 1 were static condition, 30°C temperature, pH 7 and dye concentration of 50 mg/ml. Analysis of various oxidative and reductive enzymes showed two fold increase in the activity of azoreductase and four fold higher activity of laccase in induced cell. GCMS analysis of the degradation product showed formation of number of products with varying molecular mass. Some of the products were identified and tentative pathway of degradation was deciphered. Biodegradation of Red HE7B by *Bacillus* sp.Azo1 rendered it nonphytotoxic.



Effect of different aromatic hydrocarbons on survival of *Azotobacter chroococcum* in soil

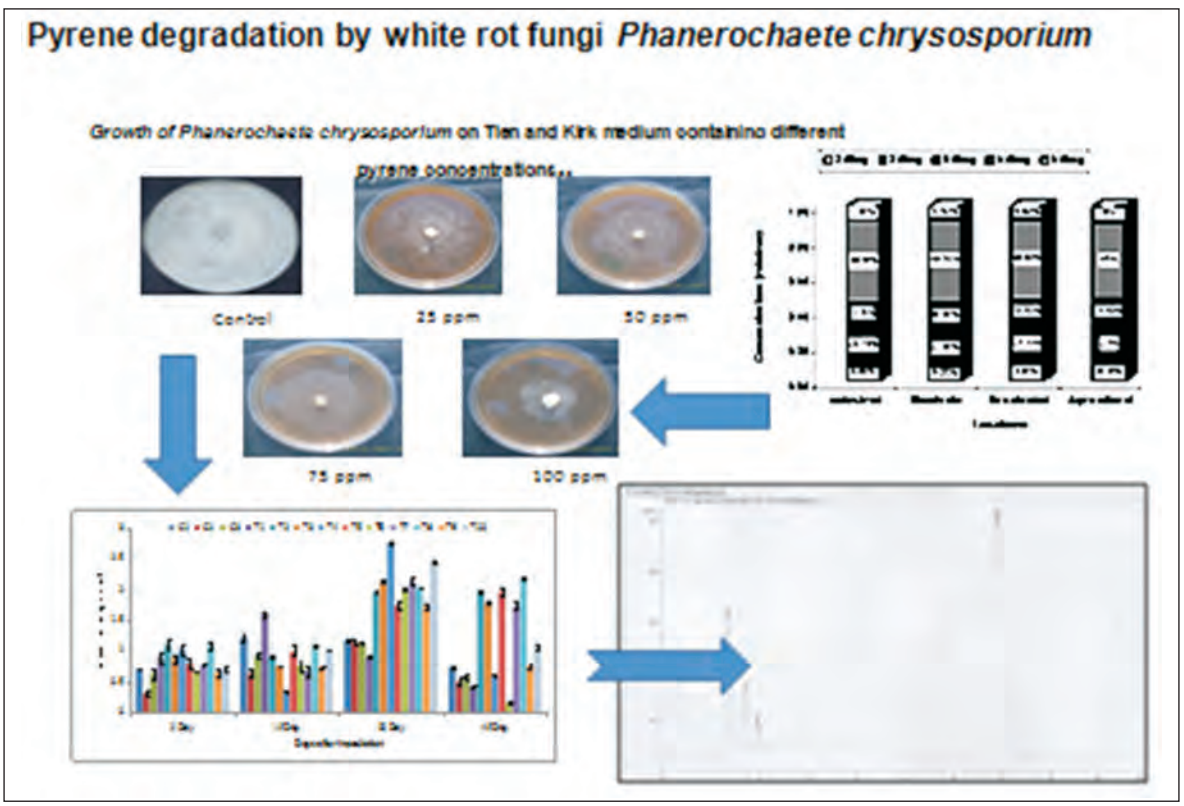
Amongst the different aromatic compounds evaluated, *Azotobacter chroococcum* cultures showed maximum growth on media containing 0.5% aniline and 0.5% benzoic acid.Both the cultures showed highest IAA production and acetylene reduction activity in



Proposed pathways for degradation of Red HE7B by *Bacillus* sp. Azo 1 based on GC/MS analysis

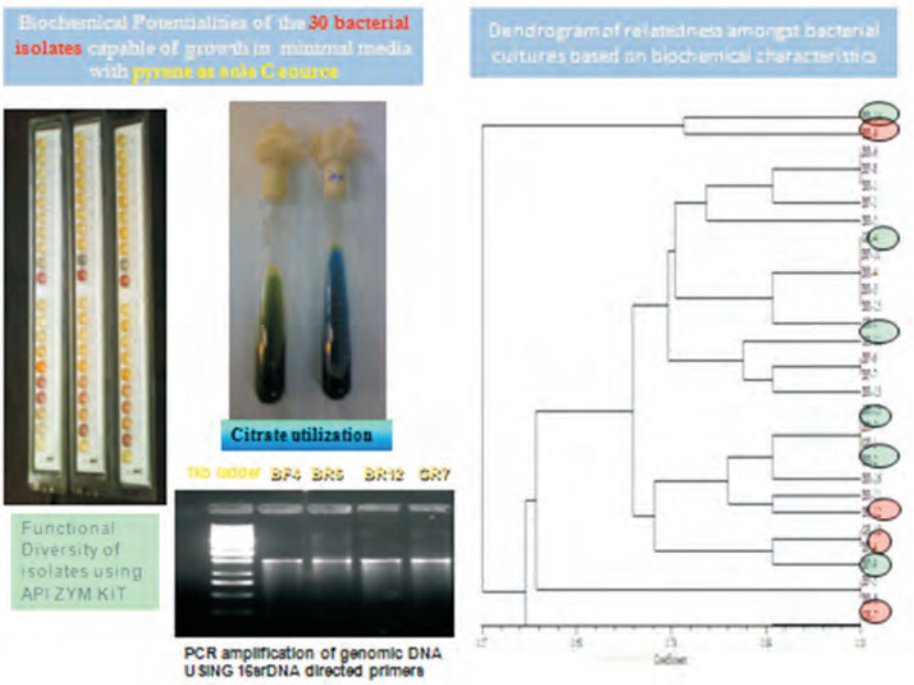
presence of benzene and benzoic acid, respectively. Drastic reduction in population was observed in case of soil amended with 0.5% aniline and it continued to be the least supporter of survival of *Azotobacter* in soil.

Polycyclic aromatic hydrocarbons (PAH) belong to a class of environmentally persistent compounds, which are ubiquitous in aquatic and terrestrial ecosystems. The investigation was undertaken to evaluate pyrene degradation potential of eleven white rot fungal (WRF) strains. After preliminary screening, two strains namely *Phanerochaete chrysosporium* MTCC 787 and *Heterobasidion annosum* MTCC 146 were selected because of their superior mean performances in terms of growth attributes in presence of pyrene as sole source of carbon in minimal media. Upon evaluation of these two strains for their pyrene degradation potential, in presence of different surfactants (Tween-20, Tween-80, Triton-X 100 and PVP) and co-metabolic sources (sucrose and cellulose), it was observed that addition of Tween-80 and Tween-20 at 0.5% w/v significantly improved the amount of pyrene degradation by both the selected cultures. Interestingly, *Heterobasidion annosum* MTCC 146 has shown higher amount of degradation of pyrene in the presence of Triton-X 100 (0.5 % level) while *Phanerochaete chrysosporium* MTCC 787 evoked a poor response. Among all treatments, *Heterobasidion annosum* MTCC 146 was found superior in terms of pyrene degradation in the presence of surfactants as well as in the presence of sucrose (200 ppm) as a cometabolic source. During pyrene degradation under microcosm conditions (soil spiked with 100 ppm pyrene, treatments having agrowastes as amendment recorded higher values of microbial activity. Dehydrogenase activity was found maximum in most of the treatments after 15 days of inoculation in both the selected strains. An increasing trend in FDA hydrolysis activity was observed until 30 days after inoculation in both the selected strains which declined after 45 days of inoculation in all treatments. Synthesis of humus during pyrene degradation by fungi continued till 45 days after inoculation. Maximum humus content (3.5 %) was seen in soil inoculated with *Phanerochaete chrysosporium* MTCC 787 amended with soybean trash. Both the strains of WRF exhibited the same pattern of fungal biomass production which was recorded more after 15 days of inoculation as compared to that after 30 and 45 days of inoculation.



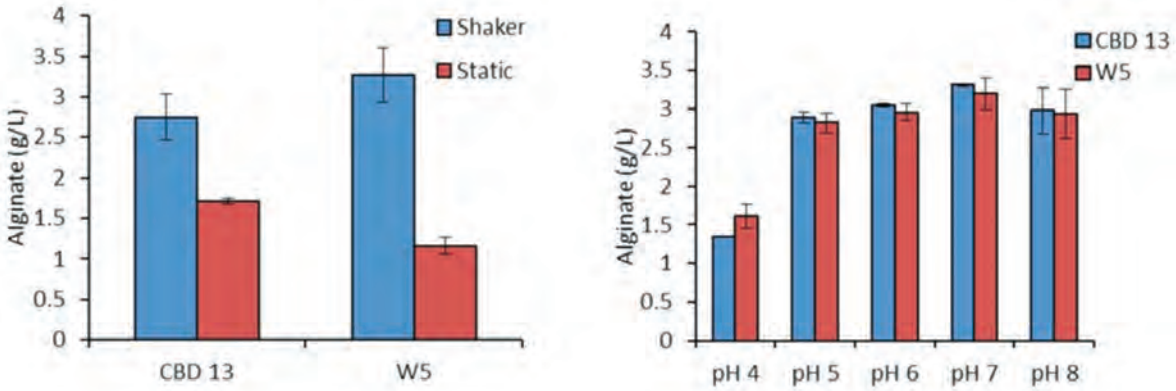
Results of HPLC analysis revealed that amendment with agrowastes enhanced the amount of pyrene degradation by both WRFs. In 100 ppm (100 µg g^{-1} of soil) pyrene spiked soil > 90 % of degradation could be achieved within 45 days when both the white rot fungi were applied individually along with agrowastes (rice straw, wheat straw, maize cob powder, soybean trash) ± (0.5 % Tween 80).

A total of 30 bacterial strains were isolated from two oil refinery sites (Guwahati and Vadodara) on Bushnell Haas medium containing 50 ppm pyrene as sole source of carbon and energy. The bacterial isolates were characterized morphologically and biochemically. A dendrogram was prepared and ten distantly placed isolates were selected based on phylogenetic relationships, biochemical attributes and morphological characteristics. Out of these ten isolates, four isolates were selected for molecular characterization on the basis of their growth potential on pyrene as sole carbon source. The metabolic potential of these four selected isolates was also tested by API-ZYM kit. Genomic DNA of all the four isolates was isolated and PCR-amplification of 16S rRNA gene was done using specific primers. The PCR product was sequenced through ABI-PRISM sequencer using big dye (Chromas Inc.). Phylogenetic tree of genetic relatedness was prepared in Clustal-W for each isolate on the basis of BLAST result. The four selected bacterial isolates namely BF-4, BR-6, BR-12 and GR-7 were found to be the species of *Stenotrophomonas maltophila*, uncultured *Pseudomonas* sp., *Stenotrophomonas* sp. and *Pseudomonas* sp. respectively.



Twenty bacterial isolates were screened for biosurfactant production by emulsification assay and bacterial isolates SL-71 and BR-12 were selected for quantification of biosurafactant by turbidometeric method. The isolate BR-12 produced more biosurfactant as compared to SL-71. The bacterial isolate BR-12 was selected for further biochemical and molecular characterization. 16S rDNA sequencing of the isolate revealed that it was *Stenotrophomonas maltophilia.* This strain was assessed for its ability to grow in Bushnell and Hass medium with pyrene (20, 50 and 100 ppm) as sole carbon source. Estimation of microbial activity parameters such as microbial growth in terms of absorbance, cell pellet protein, total soluble protein, and fluorescein diacetate (FDA) hydrolase activity revealed that culture grows best at 50 ppm pyrene concentrations. The quantification of pyrene degeradation was done by HPLC method by using strain BR-12 and strain SL-71. The isolate BR-12 was found to degrade 87% of the pyrene in period of 20 days, while SL-71 was found to degrade only 68 % pyrene in within the same period. From the experimental data it can be concluded that the biosurfactant production ability of bacteria play major role in determining its ability to degrade pyrene which can be further exploited in bioremediation of contaminated sites.

Among the twenty four bacterial cultures screened, two bacterial cultures *A. chroococcum* strains CBD 13 and W5 had the highest potential for alginate production. Optimum cultural conditions for enhancing alginate production were aeration, pH 7, 30°C temperature, incubation up to 8 days, mannitol and peptone as carbon



and nitrogen source, respectively. *A. chroococcum* strain CBD 13 produced mannuronic acid rich alginate, while strain W5 produced guluronic acid rich polymer. Thereby indicating their potential in pharmaceutical and food industry respectively. Viscosity and gel forming ability of crude bacterial alginate was very similar to that obtained from pure algal alginates. There was a considerable enhancement in the activities of the key enzymes, GDP mannose dehydrogenase and GDP mannose pyrophosphorylase in optimized medium, which resulted in enhanced alginate production.

Future Perspective

- Defining plant and microbial signals for effective colonization in plant and its rhizosphere
- Microbial nitrogen cycling in the rhizosphere
- Study of the functional microbial diversity under the conservation agriculture system
- Role of root exudates in legume-microbe interaction
- Diversity of plant associated endophytes
- Prospecting Wheat/Maize microbiome as influenced by agroclimatic factors
- Interactions between methanogenic archaea and photosynthetic microorganisms in rice rhizosphere
- Prospecting actinomycetes for industrially important enzymes (Proteases, lipases, tannase etc).
- Proteomic and metaproteomics studies to identify novel biocatalysts and biomolecules
- Novel and useful formulations of microorganisms for use as biofertilizers
- Bio-control agents and their impact on rhizospheric microbial communities and their antagonistic potential under different cropping systems
- Evaluation of downstream processing option for preparation of indigenous cellulase enzyme cocktail
- Microbial genomics and bioinformatics tools for mining genes of industrial and agricultural importance from Archaea and Eubacteria
- Evaluation of soil microorganisms in relation to carbon sequestration
- Below and above ground interactions of cyanobacteria with plants
- Cynobacterial biopigments : production and upscaling
- Development of microbe-based technology for imparting tolerance to elevated climatic variabilities and mitigating green house gas emissions from agricultural fields
- Signalling mechanisms in plant- cynobacteria/biofilms and their interactions for improved efficacy as biocontrol/PGP agents
- Role of low temperature tolerant microorganisms for degradation of horticultural waste

- Cold active cellulases for simultaneos saccharification and fermentation of lignocellulosic biomass
- Utilization of lignocellulolytic agro-residues for microbial production of value added products
- Bioprospecting endophytic bacteria/cyanobacteria for useful traits
- Microbial interventions for ameliorating abiotic stresses in crop plants-understanding the basic mechanisms
- Biodegradation of persistent environmental pollutants using efficient microorganisms
- Nutritional quality assessment of cyanobacteria/*Azolla* in relation to its efficient utilization as food and feed supplement.

Acknowledgement

We wish to thank all the Faculty Members of the Division of Microbiology for extending support and cooperation in preparing the research contributions of M.Sc. and Ph.D. students made during the period of 2007-16.

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Soil Science and Agricultural Chemistry

R.D. Singh

Introduction

The history of the Division of Soil Science and Agricultural Chemistry is traced back to the Chemical Section headed by Imperial Agricultural Chemist, which came into existence in 1905 with the establishment of the Imperial Agricultural Research Institute, housed in the Phipps Laboratory, the grand edifice named after the American Philanthropist Mr. Henry Phipps. The Great Bihar Earthquake of 1934 led to the shifting of the Imperial Institute to New Delhi where the building of the erstwhile Chemical Section continues to carry the name of Phipps Laboratory. With the country's independence in 1947 the Chemical Section was renamed as the Division of Soil Science and Agricultural Chemistry.

The main objective of this Division is to carry out research on the fundamental and applied aspects of the physical, chemical and biological properties of soil for restoring and improving soil health; enhancing nutrient use efficiency; fertilizers and manures and their interaction with soil and plant; utilization of organic residues (wastes) as manure; soil test crop response correlation study; advice to the farmers on fertilizer recommendations based on soil tests.

Significant Student's Research Achievements

Carbon Dynamics, Sequestration and Soil Quality under Different Management Practices

Total organic carbon (TOC) and Walkley-Black organic carbon (WBC) were not significantly affected by tillage, whereas there was positive impact of moisture content on TOC and WBC content in soil after completion of seven cropping cycles under rice-wheat system. On an average, puddled soil showed significantly higher labile carbon (LBC) than the non-puddled one, while, negative trend was observed in case of microbial biomass carbon (MBC). With the decrease in the frequency of irrigation, the LBC and MBC content in soil exhibited an increase. Soil treated with FYM and green manure along with mineral fertilizers or organic sources of nutrients, showed significantly higher LBC than the soil receiving only mineral fertilizers. Green manure in the rice-wheat system was more effective in increasing the LBC content of soil as compared to crop residues. Organic sources of N proved to be less efficient in maintaining plant-N content of both the crops as compared to integrated and mineral sources. Results showed that the soil test-based NPK fertilization did not improve the TOC over unfertilized-control, but increased the other SOC fractions significantly in 0-15 and 15-30 cm soil layers. Contribution of WBC, MBC and LBC towards TOC in different treatments was in the range of 17 to 22, 1.0 to 1.8, and 6.7 to 11.8%, respectively. Labile fractions like MBC and permanganate oxidisable C (PmOC) were generally higher in treatments that received FYM, sulphitation pressmud (SPM) or green gram residue (GR) along with fertilizer NPK, whereas these fractions were much lower in NPK+ cereal residue (CR) treatment. Oxidisable organic C fractions measured as per modified Walkley-Black's procedure revealed that active pool (very labile C+ labile C) was much larger in NPK+FYM and NPK+GR+FYM treated plots, whereas passive pool (less labile C+ non-labile C) was larger under control and NPK+CR. The E_4/E_6 ratio of fulvic acid (FA) and humic acid (HA) increased with continuous cropping and integrated use of NPK and organics like FYM, and GR. The INM options, except NPK+CR, also increased mineral N, Olsen-P and

available S contents over sole fertilizer treatments. Highest grain yields of rice (6.3 t ha^{-1}) and wheat (5.8 t ha^{-1}) were obtained in NPK+SPM treatment. Available nutrient contents and active SOC to TOC ratio, especially in top soil (0-7.5 cm), showed highly significant positive relationship with grain yields. The E_4/E_6 ratios of FA and HA were, however, poorly correlated with available nutrients and crop yields.

Distribution of various carbon fractions and enzyme activities was sensitive to the prevalence of dominant land use systems viz. organic farming, soybean-wheat, fodder, undisturbed natural forest and barren land in Almora district of Uttarakhand. The highest values for total organic carbon (TOC) (45.3 g kg^{-1} soil), microbial biomass C (MBC) (783.9 mg kg^{-1} soil), particulate organic C (POC) (10.9 g kg^{-1} soil) and labile C (LBC) (12. 8 g kg^{-1} soil) were found in forest soil and the same were lowest in barren land. Irrespective of the depth and seasonal differences, the undisturbed forest system maintained the highest activities of dehydrogenase (4.68 µg TPF g^{-1} soil hr^{-1}), acid phosphatase (309 µg PNP g^{-1} soil hr^{-1}), alkaline phosphatase (139 µg PNP g^{-1} soil hr^{-1}) and arylsulfatase (184 µg PNP g^{-1} soil hr^{-1}). The carbon fractions were less affected by seasonal changes than the enzymes.

An investigation was carried out in the East Khasi hills of Meghalaya by selecting three major land use systems viz. AF (26 years), agriculture (10 years) and horticulture (15 years) with four major crops or trees under each land uses (Champak, Tree bean, Alder and Khasi pine under AF land use; Maize, Potato, Rice and Turmeric under agriculture; Pear, Peach, Khasi mandarin and Guava under horticulture land use). A fallow land or control plot (without tree or crops) nearby each major land use was selected for comparisons. Amongst the three major land use systems, AF land use recorded maximum value of these attributes followed by horticulture land use and the lowest was under agriculture land use. The results showed that the contents of total organic carbon (TOC), microbial biomass carbon (MBC), labile carbon, non-labile carbon and particulate organic carbon (POC) followed the order: agroforestry > horticulture > agriculture. The present investigation thus concludes that planting agroforestry tree or horticulture fruit trees or agriculture crops cultivation with better management practices in the fallow lands could improve the soil fertility, reduce the soil losses and increase the carbon sequestration in the highly degraded soils.

A field experiment was carried out to study the impact of nutrient management using organic manures and fertilizers on organic carbon fractions, physical properties and enzyme activities in surface (0-15 cm) and sub-surface (15-30 cm) soil depths in a 4-year-old maize-wheat cropping system. Results emanated from the field experiment revealed that total organic C increased by 74.9 and 80.0 per cent under vermicompost and 50% NPK+vermicompost treated plots, respectively over control in surface soil under maize; whereas the corresponding increases were 32.6 and 36.5 per cent over control at sub-surface soil. The Walkley Black C content increased significantly under vermicompost and 50% NPK+vermicompost by 72.0 and 82.6 per cent over control; while these increases were 27.6 and 35.3 per cent, respectively over 100% NPK.

The long-term effect of manuring and fertilization on stability and quality of SOM in high intensity potato-based cropping system in Inceptisol of semi-arid sub-tropical India was studied. Rice-potato-wheat cropping system should be recommended in Inceptisol for long-term C sequestration, while as an alternative cropping system to rice-potato-wheat, maize-potato-onion cropping system should be recommended for higher system productivity in terms of rice equivalent yield. The regression model that has been developed by the ammonium oxalate extractable Fe, Si and Al and total soil C could be used for predicting the stability of carbon in both the potato-based cropping systems in Inceptisol under long-term manuring and fertilization. After 9 years of continuous applications of various organic manures, dramatically enhanced the available N, P, K and contents and the effect was more pronounced in GM + FYM + BF treatment which invariably also showed the highest grain yield of both rice and wheat. From the relationship of estimated return of C (ERC) annually versus annual change in SOC (dCs/dt) over 9 yrs. ($y = 0.2028 x - 0.2239$), the humification rate constant (h) and decay rate constant (r) were estimated as 0.2028 and 0.0079, respectively. This indicates that about 20% of the ERC entered the stable pool of C to enhance the C storage in soil. The annual loss of C from native pool of soil organic matter (SOM) was estimated to 223.9 kg C. The study also revealed that in order to offset the C

loss from the rice-wheat cropping system system in Inceptisol of semiarid subtropical India, about 1104 kg C ha^{-1} $yr^{-1}$is required to be applied to maintain the SOC in equilibrium.

Nutrient Availability and Transformation at the Soil-Root Interface & under Global Warming Scenario

Increase in solution P concentration significantly decreased the oxalic acid exudation from roots; oxalic acid concentration increased and P fixation capacity decreased with increase in the amount of residue added. Studies involving five wheat varieties (HD 2687, HD 2733, HD 2643, HD 2932 and HD 2894) grown with three levels of organic residue (mixture of pressmud, bagasse and rice straw in 1:1:1 ratio) *viz.* R_1 (0 t/ha), R_2 (5 t/ha^{-1}) and R_3 (10 t/ha). Maximum oxalic acid concentration (9.21 mg/kg^{-1}) was observed at 60 days after sowing (DAS) with variety HD 2687 and minimum (1.98 mg/kg^{-1}) was in HD 2932 at 45 DAS. Residue addition significantly increased the P uptake by crop.

AFAS content increased in the rhizosphere, mild extractants extractable SRO dissolution increased due to fertilization and rhizospheric effect. Non- labile carbon showed significant positive correlation with AFAS content in both the clays and allophane content in coarse clay. This suggests the possible role of SRO minerals in carbon sequestration. Significant positive correlation was observed between P fixation and different pools of iron and aluminium. K fixation was significantly positively correlated with AFAS content. This indicates the possible role of SRO minerals in P and K fixation. Therefore, these natural nano materials should be managed for sequestering carbon to mitigate climate change and natural nano material based slow release P and K fertilizer to increase their use efficiency.

Wheat responded positively to elevated CO_2, but negatively to higher temperature. Combined elevation of CO_2 and temperature manifested in a significant ($p< 0.05$) decrease 5% in shoot weight. Ammoniacal-nitrogen (NH_4-N) content remained unaffected, whereas the nitrate-N (NO_3-N) exhibited a significant reduction. Nitrate reductase activity (NRA) and population of denitrifiers were also decreased significantly at the elevated CO_2 level. Elevated temperature impacted positively the potential mineralizable nitrogen, whereas it exerted negative effect on potential nitrification, microbial biomass carbon (MBC), microbial biomass nitrogen (MBN) and dissolve organic carbon (DOC). Elevation of atmospheric CO_2 or temperature did not affect the population of *Nitrosomonas* and *Nitrobacter*, while elevated CO_2 reduced the population of denitrifiers. Higher doses of P fertilization helped in reducing the yield loss caused by combined elevation of CO_2 and temperature, but caused further deterioration in nutritional quality of wheat, suggesting a definite trade-off between yield and nutritional quality of wheat under changing climate. In response to the higher P demand by plant, activities/ concentrations of acid phosphatase, alkaline phosphatase and malic acid in wheat rhizosphere increased by 31.1, 22.9 and 46.3%, respectively under combined elevation of CO_2 and temperature as compared to their ambient combination. Owing to increased phosphatase activity, organic-P in wheat rhizosphere recorded a decline of 21.9%, signifying the increased contribution of organic P mineralization in meeting the higher P requirements of plant under elevated CO_2 and temperature.

Nanotechnological Interventions in Enhancing the Nutrient Use Efficiencies

A series of nanoclay-polymer composites (NCPC) were synthesized based on types of nanoclays (10% by weight) separated from the different soils. The equilibrium water absorbency and nutrient release rate decreased with incorporation of clay into the polymer matrix owing to increase in the cross-linking points and decrease in the mesh size of NCPC as compared to pure polymer. The leaching study showed that the solution concentration and cumulative recoveries of P and mineral N from the NCPC-treated soils were higher. Among the three types of clays, smectite-dominated clay incorporated in NCPC showed better performance in terms of slow release properties of the loaded nutrients. The NCPC-loaded phosphatic fertilizer was superior to conventional P fertilizer in enhancing the Olsen-P, $CaCl_2$-extractable P and mineral-N content in the soil; half

rate of fertilizer application as NCPC was statistically at par with full dose of conventional fertilizer. Decline in the non-labile pools of inorganic P fractions (Al-P, Fe-P and occluded-P) with the application of NCPC suggested transformation into more labile fractions. The effectiveness of metribuzin loaded NCPCs were evaluated under controlled laboratory condition and compared with commercial metribuzin in terms of release both in water and soils (Inceptisol, Ultisol and Vertisol). In both the soil and aqueous system, the release of metribuzin from NCPCs was lower as compared to commercial metribuzin formulation. In all the NCPCs release of metribuzin was slower than commercial metribuzin formulation, so it could be used for control release of metribuzin in soils tailored to different crops/varieties.

Autoclaved nanoparticles drastically reduce the bacterial population. Such reduction in bacterial population after autoclaving was 92% in fullerene followed by nano-ZnO (89%) and nano-Fe_2O_3 (75%) as compared to un-autoclaved nanoparticles. ZnO NPs significantly enhanced the Zn content in wheat and rice straw in both the soil types, whereas Fe2O3 showed significant enhancement of iron content in wheat plants under Inceptisol only. ZnO NPs with Azolla showed significantly higher root biomass; grain yield and harvest index of rice in Vertisol as compared to control, whereas treatments with other NPs did not show any perceptible effects. It was clear that among the NPs, ZnO has a prominent effect to alter different biogeochemical properties of soil ecosystem. Nanoparticles of Fe and Zn oxides at 100 ppm performed better than $FeSO_4$ or $ZnSO_4$ to supply Fe and Zn to rice plant with no adverse impact on microbial activity in soil under ambient and elevated CO_2. Bioaccumulation of n-Fe_2O_3 and n-ZnO in the root cells and toxic effect were evident at higher concentration of these nanoparticles.

Studies were undertaken *viz*., synthesis of nanoclay-polymer composites (NCPCs) loaded with urea and nitrification inhibitors and their characterization and their evaluation for nitrification inhibition activity. A series of poly (acrylic acid-co-acrylamide)/bentonite composites were prepared by partially neutralized acrylic acid and acrylamide in the presence of bentonite nanopowder, using N,N'- methylenebisacrylamide (MBA) as a crosslinker and ammonium persulfate (APS) as an initiator. These NCPCs were swollen in aqueous solution of urea and nitrification inhibitors for nutrient loading to act as a slow release carrier of urea. NCPC functions in delaying the release of nitrogen and nitrification, thereby serving a novel slow release nitrogenous fertilizer. So, this study could provide a convenient method for designing and preparing superabsorbent nanoclay-polymer composites with suitable release nitrogen rates to meet the requirements of the plant, and thus produce economic and environmental benefits. An attempt was made in formulating series of novel zincatednanoclay polymer composites (ZNCPCs), starch grafted zincatednanoclay polymer biocomposites (SZNCPBCs) and chitosan grafted zincatednanoclay polymer biocomposites (CZNCPBCs) with variable percentage of commercial bentonite and nanobentonite (8%, 10 % and 12 % of monomer for each case).

ZNCPCS at two doses (@ 5.0 mg Zn kg^{-1} and 2.5 mg Zn kg^{-1}) were tested in pot experiments alongwith conventional Zn sources ($ZnSO_4$. 7 H_2O) (@ 5.0 mg Zn kg^{-1}) in two Zn deficient soils (TypicHaplustept I and TypicHaplustept II). Observations were taken at different growth stages of rice (maximum tillering stage and panicle initiation stage) and wheat (crown root initiation stage and panicle initiation stage) in rhizosphere (with plants) and non-rhizosphere (without plant) conditions. ZNCPCs increased Olsen-P content in soils as well as P uptake in both soil owing to solubilization of inorganic P through citrate action and organic-P through increase in acid and alkaline phosphatase activity. Overall increase in microbial activity as evidenced by increase in soil dehydrogenease, acid and alkaline phosphatase activity was observed in rhizosphere soil indicated stimulation of microbial activity in rhizosphere soil for both crops and soil types. A series of NCPCs were prepared using nano-bentonite and modified nano-bentonite with varying neutralization degree and cross linker level. Phosphorus was loaded as diammonium phosphate (DAP) after polymerization of NCPCs. Phosphorus loaded NCPCs were evaluated for P release rates in two P deficient soils (red and alluvial) under laboratory incubation experiment and selected the best product (NCPC prepared from modified nano-bentonite at the rate of 60% neutralization and 0.5% cross linker) for pot culture study using wheat as a test crop. Phosphorus uptake increased from 3 mg kg^{-1} in DAP treatment to 9 mg kg^{-1} in NCPC treatment in red soil, the corresponding

values for alluvial soils being 4 to 10 mg kg^{-1}. Soil P fixation percentage was reduced from 60 to 19 in red soil at flowering stage by changing conventional DAP application to P-loaded NCPC application. The corresponding data in case of alluvial soil was from 50 to 0. Fertilizer phosphorus use efficiency increased from 19 to 52 % by changing P application from DAP to P-loaded NCPC in case of red soil and the corresponding increase in efficiency in alluvial soil was from 24 to 58 %. Increase in dehydrogenase as well as phosphatase activity in NCPC treated soil over control indicated stimulation of microbial activity by adding NCPC. Hence use of P loaded, modified clay-NCPC emerged as a promising option not only in increasing P use efficiency but also for stimulating microbial activity.

The yield of wheat grain was higher when oxalic acid loaded NCPC was applied to soil compared to citric acid loaded NCPC in Alfisol and Inceptisol. Significant improvements in yield, P uptake and available P status were observed with PSB inoculation over uninoculated ones. The performance of DAP was slightly better over the RP treatment in case of wheat. But the residual impacts of RP was better over DAP. All the treatments combinations helped to maintain higher saloid-P than that of absolute control. The higher enzyme activity in the PSB inoculated pots were accompanied by a reduction in the organic P content in the soil. The acid phosphatase activity was higher in Alfisol due to the acidic (pH 5.14) while the alkaline phosphatase activity was higher in Inceptisol due to the neutral pH (7.52). The indigenous RP sources along with organic acid loaded nanoclay polymer composites and inoculation with phosphate solubilizing microorganism acted as a promising P source to crops and could be used as an alternative to the costly P-fertilizers like DAP.

Radionuclides in the Soil Environment–Remediation Strategies

Laboratory and green-house studies conducted showed that about 78-96% of ^{134}Cs was adsorbed by the soils; maximum of 96% was adsorbed in Vertisols and minimum of <85% in Aridisols. Higher ^{134}Cs sorption occurred in smectite- and mica-rich soils as compared to kaolinite-dominant soils. Removal of sesquioxides from the clay aggregates significantly reduced the Cs-sorption capacity of the soils. Experiments designed to explore the role of potassium from potassium-rich waste mica in reducing the radio-cesium uptake from the ^{134}Cs-contaminated soils by spinach and lettuce as direct and green gram and soybean as the residual crops showed that the application of mica-waste @ 40 g/kg^{-1} soil was most effective in suppressing the uptake of ^{134}Cs. Effect of application of 10 g waste mica/ kg soil in reducing the ^{134}Cs transfer was comparable to the potassium-fertilizer treatment, thus demonstrating the usefulness of the former in management of the ^{134}Cs-contaminated soils.

Products development for enhancing nutrient use efficiency

Modified phosphatic fertilizers were prepared. Product-A and Product-B were manufactured by reacting NH_3 and H_3PO_4 at a ratio of 2:1 and 1:1, while Product-C and Product-D were prepared by reacting rock phosphate with H_2SO_4 andH_3PO_4, respectively. These products were characterized for their chemical structural composition. Chemical analysis of the four products showed that Product-B had the highest total P (28.8%). Characterization of the manufactured products through FTIR indicated the presence of phosphate (PO_4^{3-}) group. Presence of –NH group was evident in Product-A and Product-B but such –NH stretching was absent in case of both Product-C and Product-D. Among the different coating agents, liquid paraffin coated materials proved to be superior coating agent over polyvinyl alcohol in terms of their yield and nutrients uptake by wheat. Thus, it can be concluded that new modified coated phosphatic fertilizers could be developed by reacting ammonia with phosphoric acid or rock phosphate with mineral acids followed by coating with coating materials like liquid paraffin of polyvinyl alcohol which could commercial DAP fertilizer.

Introduction of microbial cultures along with mica recorded higher yield and uptake of K, N and P than mica alone. Treatment receiving mica along with *B. mucilaginosus* proved to be most effective in terms of yield, uptake and relative efficiencies of K, N and P and K recoveries in all the cuttings of crop. The yield, nutrient uptake and K recoveries as well as water soluble, exchangeable, non-exchangeable and total K were much higher

in Alfisol from Hazaribag than in Alfisol from Bhubaneswar. Mica treated with microbial cultures resulted in more dissolution of K than fresh mica in both the soils. Thus, bio-intervention of waste mica could be an alternative and viable technology to utilize waste mica and could be used successfully as a cheaper source of K-fertilizer.

Incubation and green house studies were conducted to evaluate the effect of partially acidulated phosphate rock (PAPR) on P release pattern in alkaline calcareous soil as well as their agronomic effectiveness on wheat crop. Three acidulants and two levels of acidulation were used to prepare various PAPR products from four sources of PR namely Syrian phosphate rock (SPR), Jordanian phosphate rock (JPR) and two of Indian origin namely Udaipur phosphate rock (UPR) and Purulia phosphate rock (PPR). In the incubation study, the application of PAPR materials increased available P status in the soil over control throughout the incubation period for 90 days, irrespective of sources, acidulants, and levels of acidulation. However, SSP was found to be superior to PAPRs. In pot culture study, the trend of results obtained closely followed the pattern observed in the incubation study. Significant positive responses of wheat crop (dry matter yield, P content, P uptake and relative agronomic effectiveness) to the applied PAPR fertilizers prepared with sulphuric acid or pyrite were observed over control. Effectiveness of phosphate rocks as source of P to wheat followed the order of JPR> SPR=PPR> UPR, while the acidulation agent followed the order of sulphuric acid> pyrite> oxalic acid.

Integrated nutrient management & Customization of fertilizer levels for enhancing nutrient use efficiency

Continuous addition of NPK alone or in combination with FYM significantly decreased soil bulk density and increased porosity and infiltration rate of soil over N, NP, NPK and control. Use of NPK+FYM increased mean weight diameter and saturated hydraulic conductivity of soil as compared to control in the 0-15 cm layer. However, no significant differences were observed in the sub-surface. Bulk density, infiltration rate and porosity significantly and directly influenced the availability of nutrients, whereas the available water content, mean weight diameter (MWD) and saturated hydraulic conductivity (Ksat) indirectly influenced the availability of nutrients by improving the bulk density, porosity and infiltration rate. Infiltration rate contributed largely to yield and uptake of nutrients by the two crops. Other hydro-physical properties, namely mean weight diameter, bulk density, porosity and *K*sat also influenced the yield and uptake of nutrients indirectly through better soil physical environment. Soil organic carbon, available N, P, K, Zn and Fe and all the pool of N, P and K except non-exchangeable K increased significantly in FYM alone and FYM+ NPK treatments over the unfertilized control treatments. Large increase in organic P content in the FYM treated plots suggest due consideration should be given to this pool in deciding the fertilizer P recommendations. Depleting trend was observed in non-exchangeable K in all the treatments compared to the initial values. Long-term application of fertilizers had significant effect on dehydrogenase activity, soil respiration and microbial groups (bacteria, fungi, *Actinomycetes*, *Pseudomonas*, *Azotobacter*, and ammonia oxidising bacteria). Among the five treatments, in most of the cases, balanced application of NPK (*i.e.,* 100% NPK based on initial soil test values) + farmyard manures (FYM) showed highest values of the above-mentioned parameters while control plots invariably showed the lowest. Results showed that the FYM alone and FYM +fertilizer NPK treatments were superior in increasing all the pools of soil organic carbon significantly over fertilizer-NPK-alone and control treatments. Values of MBC and LBC were higher under the integrated (FYM + fertilizer NPK) treatment compared to the rest of the treatments.

Data emanating from the field experiments revealed that integrated sources of nutrients was superior in improving organic carbon fractions, whereas integrated as well as 100% NPK were equally effective in improving the N fractions in soil. Dehydrogenase activity was highest under integrated nutrient management, while urease activity was highest under 100% NPK. Combined application of enriched composts and chemical fertilizers improved available as well as various fractions of P and K significantly after both wheat and soybean than unfertilized control. Significant increase in available and different fractions of P and K in soil was maintained

in treatments receiving enriched compost than ordinary compost after both the crops. Application of enriched compost had a greater effect on P and K forms and accumulation in the surface soil (0–15 cm) than sub-surface soil (15-30 cm). Application of RP enriched composts of rice straw, mustard stover and tree leaves with and without fertilizers significantly improved total organic carbon, Walkley Black carbon, microbial biomass carbon and oxidizable organic carbon fractions. Results also revealed that application of enriched composts treated plots improved significantly higher available as well as various fractions of N, P and S after both the crops. Among the enriched composts the build-up in available N, P and S after wheat were in the order of RP enriched rice straw compost > RP enriched mustard stover compost > RP enriched tree leaves compost. From the data it was found that combined application of enriched composts along with 50% NPK had higher activities of enzymes. It can be concluded from the present study that application of RP enriched compost along with 50% NPK had pronounced impact on improving soil fertility as well as enzyme activities after both the crops indicating which could be followed to improve and maintain soil fertility, increase crop productivity under intensive cropping system.

Using basic data on nutrients requirement and contributions from available soil nutrients, applied fertilizers and farmyard manure to the total nutrients uptake by the crops, fertilizer prescription equations and ready reckoner were developed to prescribe fertilizer nutrients doses for range of soil test values of N, P and S for desired target yield (± 10% of deviation) of hybrid rice and wheat. Multivariate analysis multiple linear regression models were calibrated and validated for the prediction of post harvest soil test values (N, P and S) after each crop and whole rice-wheat cropping sequence using initial soil test values of nutrients, applied fertilizer nutrients through fertilizer and farmyard manure. The spectral algorithms identified in the present study provide possibilities for interpretation of satellite imagery and inclusion of sensitive wavelengths for designing of high resolution satellites for site- specific management of N, P and S in hybrid rice and N, P, K and S in wheat crops.

To investigate the effect of optimal (100% NPK) to super-optimal doses (200% NPK) of mineral fertilizer along with foliar iron ($FeSO_4$) on the yield attributes, nutrient harvest index and uptake pattern in wheat as well as the changes in soil fertility status and biological quality were monitored under a maize-wheat cropping system. Results revealed that foliar Fe spray along with mineral NPK significantly increased the grain Fe concentration by 6-15%; whereas a sharp decrease in the grain Zn concentration is attributed to the antagonistic interaction between phosphorus and zinc. Customized nutrition surely helps the plants to accumulate Fe in the grains and was clearly reflected in the decreased molar ratio of P/Fe, indicating increased bioavailability of Fe in wheat grains; but P/Zn increases under super-optimal application of NPK. Fertility status of soil (available N, P and K) after harvest of two cycles of crop increased with increased application of fertilizers. Fertilizer addition increases the mineral N content (NH_4^+ and NO_3^-).

The present investigation was carried out to study the release kinetics and mineralization of N for synchronizing crop demand from different organic sources with variable C:N ratio and to evaluate their effectiveness for crop productivity and maximizing N use efficiency under wheat crop. The C/N ratio of the organic manures were in the order: FYM (23.8) > sewage sludge (19.1) > rock phosphate enriched compost (18.2). The enriched compost performed better as a source of nutrient which showed 56 per cent increase over control, while treatment receiving FYM+100 mg N kg^{-1} increased grain yield by 48 per cent over control. Results indicated that significant increase in available N (NH_4 -N and NO_3 -N) in soil throughout the crop growth namely, maximum tillering, flowering and after harvest of wheat. Similarly, significant improvements in biological properties like dehydrogenase, acid and alkaline phosphatase activities were observed in soil treated with organic and inorganic sources of N.

In a pot culture experiment, rice and wheat crops were grown in sequence with two types of biochar produced in a low cost pyrolysis kiln viz., rice biochar (RCB), wheat biochar (WHB), four rates of biochar (0, 2.25, 4.50, 6.75 g kg^{-1} soil) and four rates of NPK (0, 50, 75 and 100%) in Alfisol and Inceptisol. In pot culture experiment, the biochar rate of 2.25 g kg^{-1} soil coupled with 100% NPK was found to achieve highest yield of 39.8 g pot^{-1} in both Alfisol and Inceptisol. The application of RCB prepared at 400 °C at the rate of 2.25 g

kg^{-1}(5.0 t ha^{-1}) along with 100%NPK in both Inceptisol and Alfisol emerged as the best treatment combinations showing the highest grain yield as well as apparent N recovery and factor productivity of rice. In general the application of biochar could be beneficial for enrichment of carbon and available nutrients in soil and enhancing the microbial activities in soil.

Management of sludge treated, metal polluted soils and their risk assessment

The properties of sludge obtained from the three sources varied widely but showed a general trend of higher N and P content compared to K. The pH of the sludge was in the acidic range and organic carbon content was high for all the three sources. The concentration of micronutrient (Fe, Zn, Mn, and Cu) was very high. The Fe content ranged from 10.5 to 11.9 and 46.8 to 61.0 g kg-1 during the rainy and winter season, respectively. The only detected heavy metal was lead and its concentration was highly variable. The results demonstrated that sewage sludge could be used as source of plant nutrients for crop production and maintaining soil fertility which could reduce the use of costly chemical fertilizers. A pot experiment was also conducted to evaluate, the effect of sludge application alone and in combination with chemical fertilizers on heavy metal release in Zn and Fe deficient soil with baby corn and used ten different combinations of treatment such as control, 3 treatments of chemical fertilizer and 3 levels of sludge alone and along with chemical fertilizer. Use of sludge did not show any adverse effect as far as metal content in soil and baby corn was concerned. Regarding the changes in biological properties of the soil, microbial population was increased as a result of sludge addition, with higher increase in integrated use of sludge and chemical fertilizers as compared to chemical fertilizers and sludge alone was observed. These results demonstrated that sewage sludge could be used safely as source of plant nutrients for baby corn production in Zn and Fe deficient soil.

A case study was undertaken to assess the risk of sewage-irrigated soils in relation to the transfer of trace elements to rice and wheat grain. For this purpose, peri-urban agricultural lands under Keshopur Effluent Irrigation Scheme of Delhi were selected. Results indicated that sewage effluents contained appreciable amount of plant nutrients, which can significantly curtail the use of chemical fertilizers in the sewage irrigated peri-urban agriculture. Sewage effluents were suitable for using as irrigation water in respect of their toxic trace element contents. The values of hazard quotient (HQ) for intake of trace toxic elements by human through consumption of rice and wheat grain grown on these sewage irrigated soils were well within the safe permissible limit. The variation in Zn, Ni, and Cd content in wheat grain could be explained by solubility-FIAM to the extent of 50.1, 56.8 and 37.2%, respectively. Corresponding values for rice grain were 49.9, 41.2 and 42.7%, respectively. In addition, as high as 36.4% variation in As content in rice grain could be explained by solubility-FIAM model. Toxic limit of extractable Cd and As in soil for rice in relation to soil properties and human health hazard associated with consumption of rice grain by human was established. Similar exercise was also done in respect of Cd in soil for wheat. Risk in terms of hazard quotient (HQ) to human health for intake of metals through consumption of spinach grown on sludge treated soils ranged from 0.05 to 0.23 for Zn, 0.01 to 0.02 for Cu, 0.20 to 0.56 for Fe, 0.07 to 1.18 for Mn, 0.01 to 0.03 for Ni, 0.06 to 0.23 for Cd and 0.04 to 0.12 for Pb. Safe rates of sludge application were worked out as 4.46 and 71.4 g kg^{-1} (10 and 160 t ha^{-1}, respectively) for acid and alkaline soil, respectively as far as transfer of metals from soil to human food chain is concerned.

For risk assessment a study was undertaken to predict the free ion activity of Zn, Cu, Ni, Cd and Pb in metal contaminated soil as a function of pH, extractable metals and soil organic carbon content. Free metal ion activity *viz.* pZn^{2+}, pCu^{2+}, pNi^{2+}, pCd^{2+} and pPb^{2+} as estimated by Baker soil test ranged from 5.44 to 11.7, 8.62 to 15.2, 8.11 to 14.5, 7.04 to 14.6 and 8.12 to 13.5, respectively. Free metal ion activity was predicted by pH-dependent Freundlich equation as a function of pH, organic carbon and extractable metal. Results indicate that solubility model as a function of pH, Walkley and Black carbon and EDTA extractable metals could explain the variation in pZn^{2+}, pCu^{2+}, pNi^{2+}, pCd^{2+} and pPb^{2+} to the extent of 59, 56, 46, 52 and 51%, respectively. Predictability of solubility model based on pH, labile carbon and DTPA or $CaCl_2$ extractable metal was inferior to that based on EDTA.

A pot experiment was conducted to assess the response of soybean to applied Ni (0, 5, 50 and 100 mg kg-1). Subsequently, wheat crop was grown on residual fertility of soil in respect of Ni. Results indicate that there was 16.5 to 26.6% increases in dry biomass yield of soybean to applied Ni @ 5 mg kg-1 over control; the corresponding figures for wheat were 10.7 to 41.1%. Inclusion of pH along with DTPA extractable Ni as predictor variables improved the predictability of Ni content in plant. As high as 27% variability in DTPA extractable Ni could be explained by pH, organic carbon and clay content of soil. Critical limits of deficiency of DTPA extractable Ni in soil were 0.17 and 0.18 mg kg-1 for soybean and wheat, respectively. Whereas, critical concentration of deficiency of plant Ni for soybean and wheat were worked out as 0.20 and 0.37 mg kg-1, respectively. Phytotoxicity limits of DTPA extractable Ni in soil was computed as 3.24 mg kg-1, and a plant content of 12.0 mg kg-1 on dry weight basis can be considered as toxic limit using soybean. Adsorption of Ni was the maximum for clayey and minimum for sandy soils. Freundlich and Langmuir adsorption models were equally effective in describing Ni adsorption in soils. Adsorption equation parameters parameters could explain about 75% of the variability in Ni uptake by soybean from applied Ni. Residual fraction of Ni was the dominant as compared to the other fractions.

Diammonium phosphate (DAP) was most efficient in enhancing biomass yield, frond and root arsenic concentrations and total arsenic removal from soil and reducing the arsenic concentration in soil through enhanced phytoextraction by *Pteris vittata*.. To evaluate the effect of phyto-extraction to the subsequent crop, rice was grown on the ameliorated as well as non-ameliorated soil. The biomass yield of rice, grown after phytoextraction, was higher in soil phytoextracted by *Pteris vittata* with DAP than that in the non-phytoextracted soil. Results showed that the phytoextraction procedure involving *Pteris vittata* for removing arsenic from the contaminated soil is of great use in improving the microbial biomass carbon (MBC), carbon mineralization (C_{min}), dehydrogenase activity (DHA), acid and alkaline phosphatase activity (AcPA, AlPA) and aryl sulphatase activity (ASA) and DAP should be used as a phosphate fertilizer during phytoextraction by *Pteris vittata* for the improvement of MBC, C_{min}, DHA and ASA.

Micronutrients and secondary nutrients management

The interaction effect of lower level of S (40 mg hg^{-1}) and lower level of Si (60 mg hg^{-1}) showed a positive effect on uptake of different nutrients as compared to their individual effect. After harvest, available soil S content significantly increased with addition of S, Si and FYM. Addition of Si and FYM positively influenced availability of P in soil whereas, addition of S and Si showed a negative effect on available Zn content in soil. Continuous submergence led to decrease in pH and Eh of soil. Soil pH increased and Eh decreased due to the application of Si. FYM addition decreased Eh of soil to a great extent under submerged condition. Synergistic interaction between B and S on dry matter and seed yields of mustard and sunflower were found. Similarly, a positive interaction was also observed on oil and protein contents of mustard and sunflower seeds. Release of S and B was relatively more at 400 C than at 250 C. The results further indicate that because of different adsorbing sites of B and S in soil and being non-competing in nature, the site of interaction between B and S is probably in the plant and not in the soil.

Application of lime enhanced the available nitrogen (N), phosphorus (P), calcium (Ca), magnesium (Mg), sulphur (S) and zinc (Zn) content in the boron-deficient acid soil of West Bengal. Sunflower responded very well in terms of increase dry matter yield to B application to the extent of 175 and 188% under 1 and 2 mg kg^{-1} of applied levels of B, respectively over control, while dry matter yield of sunflower reduced to the tune of 29.2 and 42.7% under 2/3rd and 1 LR, respectively over control. Application of lime decreased the readily soluble B content in soil, while oxide bound B was positively influenced by liming. Lime application @ 1/3rd LR with 2 mg kg^{-1} of applied B emerged as an optimum combination in this acid soil.

Sequential fractionation of B revealed that residual B was the dominant fraction contributing, on an average, 84.3 to 94.7% towards total B in different soils. Contribution of other fractions *i.e.* readily soluble,

specifically bound, oxide bound and organically bound B jointly was highest (15.5%) at Delhi, followed by Ranchi (6.9%) and Junagadh (5.3%). Distribution of different B fractions was in the order: readily soluble< specifically adsorbed< oxide bound< organically bound< residual. Continuous use of NPK+ FYM increased readily soluble, specifically adsorbed and organically bound B at all sites, but decreased oxide bound fraction at Ranchi, compared with other treatments. Specifically adsorbed and organically bound B was significantly correlated with available B as also with wheat grain yield and B uptake. Soil organic C and CEC were most important soil characteristics affecting B availability. For B adsorption-desorption study, 12 soils representing Vertisols, Inceptisols and Alfisols were chosen. Across the solution B concentrations, B adsorption followed the order: Vertisol> Inceptisol> Alfisol.

An attempt has been made to enrich two popular vegetable crops, *viz. Palak* (*Beta vulgaris*) and *Chenopodium* (*Chenopodium album*) with Zn, to assess suitability of chemical extractants for assessing availability of Zn in organic-amended soils and to work out hazard quotient (HQ) to human health. Results indicated that both the crops responded positively in terms of dry matter yield to applied Zn (5 mg kg-1) in alkaline soil, while such response was conspicuously absent in acid soil. A significant reduction in Zn content in both the crops was recorded due to application of FYM and sludge particularly at higher levels of applied Zn. A strong depressing effect of applied Zn on iron, manganese and copper content of both these crops was recorded. Computation of HQ for intake of these Zn-enriched leafy vegetables revealed that application of Zn up to the highest level (100 mg kg-1) could produce the values of HQ well below 0.5 in case of alkaline soil, whereas in acid soil, application of Zn even up to 50 mg kg-1 produced HQ Greater than 0.5.

Supplementation of Fe through soil application caused improvement in the DTPA and NH4OAc extractable Fe in soils. The foliar application of Fe (3% FeSO4.7H2O solution, thrice 40, 60 and 75 days after sowing of rice) was more effective and economical in correcting Fe-deficiency, enhancing the yield as well as increasing Fe content of milled rice (grain) grown under aerobic condition as compared to soil application. Ferrous-iron content in rice plants proved to be a better index of Fe nutrition status compared to total plant Fe and chemically extractable soil Fe. The Fe2+ content of $\geq$ 42 mg kg-1 in plants (on dry weight basis) appeared to be an adequate level at 45 days after sowing for direct seeded rice grown under upland aerobic condition. Among the soils, acid soil (without lime) produced highest yield of aerobic rice followed by alkaline, calcareous and lime-treated acid soils.

After 39 years of intensive cultivation, the available, total water soluble, heat soluble and inorganic S fractions of all the 4 soil depths decreased from their initial contents except in 100% NPK+S and 100% NPK+FYM treatments. However, $NaHCO_3$ extractable S (SBES), organic S and total S increased over their initial contents. All the S fractions declined regularly from surface soil (0-15 cm) to lower most soil depth (45-60 cm). The organic S was dominant fraction followed by heat soluble, $NaHCO_3$ extractable, total water soluble, inorganic and available S in all the treatments.

Rice variety Pusa 33 was assessed as an iron responsive and iron deficiency tolerant type while ADT 39 was highly sensitive to iron deficiency results showed that Pusa 33 grain iron increased up to 18.9% and in ADT 39 up to 13.4% under recommended dose of Fe followed by submerged condition. Seed inoculation of rice varieties with siderophore producing micro-organism (*Pseudomonas spp.*) significantly increased biomass produced over control, however, response was more pronounced in Pusa 33 as compared to ADT 39 and was significantly lower than of recommended dose of Fe in the form of $Fe(NO_3)_3.9H_2O$ and spray of 0.5% Fe-EDTA. This study clearly showed that in aerobic rice system grain and straw yield and grain Fe content (Fe biofortification) could be achieved to the level of anaerobically grown irrigated rice system by the selection of suitable iron management practices and iron chlorosis tolerant genotype.

Boron adsorption in soils was affected by pH, soil organic C, free Al- and Fe-oxide and clay content of the soil. For describing B adsorption, Freundlich equation appeared to be the best fit for all soils with both equilibrium times (4 and 23 hr), whereas Langmuir equation could only be fitted to 23 hr equilibrium time. Maximum B retention capacity of soils as evident from Langmuir 'b'/maximum buffering capacity (b×k) or

Freundlich 'a' was positively affected by organic C, clay and free Al- and Fe-oxides. Interestingly B adsorption in soils was found higher both at acidic and alkaline pH ranges as compared to that of near neutral soils.

Application of Zn along with FYM was relatively more effective in enhancing residual available Zn in soil over control as compared to $ZnSO_4$ alone. This was also reflected well on grain Zn content in wheat grown after the harvest of rice. The Fe and P uptake in rice was not affected due to Zn application. Further, daily dietary intake of Zn by human through consumption of rice grain was 3.53 mg day^{-1} in control and 4.38 mg day^{-1} in soil and foliage application of Zn. The corresponding values for wheat were 5.92 mg day^{-1} in control and 6.61 mg day^{-1} in soil treated previously (rice) with $ZnSO_4$and FYM.

Characterization of Soil Resource and Assessment and mapping of soils

Hand held hyper spectral Spectroradiometer was used to characterize the spectral reflectance patterns of four different soil orders. The Mollisol spectral reflectance was the lowest one as compared to all other soil orders throughout the spectral region (350-2500 nm). Spectral reflectance pattern of Vertisol showed higher reflectance pattern than Mollisol but lower than Inceptisol and Alfisol. Few best correlated bands of different spectral parameters were chosen through multiple regression analysis (MRA) and prediction equations were developed taking 50 soil sample data and these were validated with rest 37 soil sample data. The best fitted equations were developed for predicted and measured parameter values and found that they were best fitted with 1:1 line.

Studies on representative pedons along a toposequence belonging to Kumaon Himalaya revealed that the soils on hill top, side slope and broad valley are rich in organic carbon and high to medium in available nitrogen but low in available phosphorus and high to medium in available potassium; whereas soils in the narrow valley are low in organic carbon, medium in available nitrogen, low in available phosphorus and potassium. Available micronutrient cations were sufficient in all soils of the toposequence except Cu in the side slope. Lower cation exchange capacity of the soils along the toposequence is due to the dominance of kaolinite and mica type of clay minerals. As confirmed through the XRD studies, clay transformation in the toposequence followed the route: Mica $\rightarrow$ vermiculite/chlorite/hydroxy interlayered vermiculite $\rightarrow$ kaolinite.

A detailed soil survey was undertaken in the farm of IARI using modern techniques such as satellite image, global positioning system (GPS) and geographic information system (GIS). The spatial variability mapping of soil fertility parameters delineated the spatial extent of low status of soil organic carbon and available N; areas with high to very high level of available P and K besides delineating areas of deficiency in available sulphur. Average contentof soil available Fe, Mn, Zn and Cu within the farm was 12.55, 14.20, 3.30 and 2.24 mg kg^{-1} respectively. Geostatistical analysis revealed that soil available Fe, Mn and Cu concentration had moderate spatial dependency with the spatial range of 404, 801 and 1529 m respectively. Soil available Mn, Zn and Cu concentration was found to be adequate while a few patches of soil showed available iron deficiency in the farm which needs attention for soil management or fertilization with iron for higher crop productivity.

Tillage and crop residue management

Conventional tillage with crop residue was statistically at par with zero tillage without crop residue treatment. Olsen's P and $NaHCO_3$-P (0.5 M $NaHCO_3$, 16 h shaking) of surface soil showed a significant positive relationship with total P uptake of *kharif* crops. A significant positive relationship was observed between biological yield and total P uptake with sub-surface soil $NaHCO_3$-P. Results of this investigation indicate that the soil P management decisions may differ according to the nature of cropping system and tillage as well as crop residue management practices. Tillage and crop residue management caused significant changes in the $NaHCO_3$-P and NaOH-P, and NaOH-P_i fractions of both the rhizosphere and non-rhizosphere soils. Organic phosphorus content of surface soil exhibited increase in the zero-tiled plots whereas inorganic P content tended to show decline. Activity of acid and alkaline phosphatase enzymes was significantly higher in zero-tilled-bed-planted plots which had

received residues of both the crops. It has been observed that soil organic carbon and its different fractions and total N (TN) varied significantly at 0-5 cm and 5-10 cm depths, which may be due to differential application of residues and various tillage options. In comparison to the incorporation of residues of pigeonpea alone (Rip) and wheat alone (Riw), the conjoint application of both the above residues (R2) significantly increased (SOC), particulate organic carbon (POC) under conventional tillage-zero tillage [CT-ZT] and zero tillage-conventional tillage [ZT-CT] combinations at 0-5 cm soil depth. [CT-ZT] and [ZT-CT] also brought significant improvement in mean weight diameter (MWD), geometric mean diameter (GMD), macroaggregate associated carbon (MaTC) and percentage of macro aggregates. Zero tillage (ZT) based main plot treatments usually exhibited increased aggregation and (MaTC) over conventional tillage (CT). The cropping system with ZT promoted macroaggregate formation, which might be favourable to reduce particulate nutrient transport in this soil. The organic carbon fractions were higher in the macroaggregates than microaggregates, irrespective of tillage/nutrient practices under both the cropping systems.

To evaluate different N management options i.e. basal application of 80, 50 and 33% of total fertilizer requirement followed by need-based top dressing as suggested by Green Seeker, and N sources and methods of application on crop yield, N uptake, N use efficiencies (NUE), and temporal changes in soil organic C and mineral-N in maize (*Zea mays* L.)-wheat (*Triticum aestivum* L.) cropping system under conservation agriculture (CA) and conventional tillage (CT) practices were evaluated. Results revealed that maize grain yield was statistically at par under both cultivation practices, whereas grain yield of wheat was significantly higher under CA than that under CT. On an average, N use efficiencies in wheat computed as agronomic efficiency (AE_N), partial factor productivity (PFP_N) and recovery efficiency (RE_N) were 23.2 kg grain kg^{-1} N, 38.4 kg grain kg^{-1} N and 52.5%, respectively under CA; the corresponding values under CT were 15.1 kg grain kg^{-1} N, 26.1 kg grain kg^{-1} N and 37.5%. Among N sources and methods of application band placement of slow release modified urea materials (*i.e.* USG and IFDC-product) resulted in higher yields and NUE compared with urea broadcasting. The benefits associated with CA present a greater potential for its adoption to sustain soil health and crop productivity of maize–wheat cropping system

Soil Quality Index for Soil health

PCA analysis showed, available iron (Fe) and zinc (Zn), hydraulic conductivity (HC), water stable aggregates (WSA), potentially mineralizable N (PMN) and microbial biomass carbon (MBC) emerged as the most sensitive indicators of soil quality after rice-harvest whereas the WSA, soil organic carbon (SOC), available copper (Cu), manganese (Mn) and phosphorus (P), PMN, respiration and dehydrogenase activity (DHA) were sensitive after wheat-harvest. Microbial quotient (MQ) and MBC emerged important for the goal of environmental protection. Soil quality index (SQI), computed following non-linear scoring function (NLSF), and was highest under puddling during rice and no-tillage during wheat; treatments receiving partial or full substitution of N through organics yielded higher SQI. The CF approach was superior to PCA in assessment of soil quality under rice-wheat system.

Nitrification inhibitor

Biological nitrification inhibitors (BNI) extracted from *B. humidicola* with 70 and 40% alcohol were more effective (73.5 and 63.0) as compared to the plant-based (38.0%) and synthetic nitrification (40.48%) inhibitors. Nitrogen-15 aided experiments showed that the total fertilizer N uptake by rice plant increased with increase in the N levels. Nitrogen use efficiency in rice plant was highest under the treatment receiving root extract got with 70% alcohol (41.9%) followed by buffer solution extract (41.3%). The residual fertilizer N uptake by wheat was significantly higher in the salt solution extract followed by neem oil coated one and it was the lowest in control. The nitrogen use efficiency (NUE) in residual wheat biomass significantly increased at 60 mg N kg^{-1} soil over 30 mg N kg^{-1}. The maximum NUE was observed in the salt solution extract (0.77%) followed by the neem oil coated (0.69%), DCD (0.64%) and buffer solution extract (0.61%). Results showed that all the *B.*

humidicola extracts influenced the NH4+-N content in soil. Among the treatments T1 and T6 showed more inhibition nitrification process. BNI and synthetic nitrification inhibitors suppressed significantly nitrification process up to 14 days. However, the inhibition was also observed up to 22 days with reduced extent. It was concluded that increased nitrogen uptake and nitrogen use efficiency in rice and wheat crops when applied with root extracts of *B. humidicola* specially 70 % alcohol extract was demonstrated.

Biological interventions for nutrient use efficiency

Dehydrogenase, alkaline phosphatase and acid phosphatase enzyme activities studied at tasseling (maize), panicle emergence (wheat) and harvest (maize and wheat) showed an increase with microbial inoculation, the effect being more pronounced under AM+PSB treatment. Enzyme activities generally increased at lower P rates over no P, but showed a decrease thereafter with increasing P application rate. Performance of microbial inoculants in affecting soil P fractions, yield and uptake was in the order: AM+PSB>PSB>AM. Phosphorus use efficiency, measured as apparent recovery and agronomic efficiency, was highest in maize (38.3% and 34.6 kg grain kg^{-1} P, respectively) and wheat (45.8% and 84.6 kg grain kg^{-1} P, respectively) when the crops received 50% of recommended P along with AM+PSB inoculation. Results indicated beneficial effect of microbial inoculants, especially AM+PSB, on enhancing crop yields and P use efficiency provided these are used with curtailed P rates. The study suggested for evaluation of interactive effects of fertilizer P and inoculants in different soil types and cropping systems to explore the possibility of curtailing fertilizer P demand.

Acknowledgement

We wish to thank all the Faculty Members of the Division of Soil Science and Agricultural Chemistry for extending support and cooperation in preparing the research contributions of M.Sc. and Ph.D. students made during the period of 2007-16.

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M.Sc. and PhD theses submitted by the students in the Division of Soil Science and Agricultural Chemistry, ICAR-Indian Agricultural Research Institute, New Delhi 110012 during the period of 2007-2016.

Water Science and Technology

Man Singh

Introduction

The Project Directorate of Water Technology Centre has the mandates to conduct basic and applied research on all aspects of water management in agriculture and develop technologies for improved agricultural water management. During 2007-16 the Post Graduate Students of Water Science & Technology conducted research on seven theme areas. These were: (1) Surface and drip fertigation in field and garden crops. (2) Management of water and soil resources. (3) Climate change and water resources planning for irrigation. (4) Surface irrigation design and operation. (5) Diversified use of stored groundwater for aquaculture. (6) Climate Change and Conservation Agriculture and (7) Soil health, poor quality and waste water management. The significant research contributions during the decade are presented theme wise here under:

Significant Student's Research Achievements

Surface and drip fertigation in field and garden crops

Considering future demand of lettuce crop in India a field study was carried out on its cultivation techniques supported by high tech irrigation. The aim of this experiment was to study the optimal crop geometry and estimate water requirement of lettuce under trickle irrigation. There were combinations of seven treatments involving three crop geometry, two different fertigation strategies and two irrigation levels. Based on previous year result like dampness observation, a new treatment of irrigation scheduling for 7 days was added. It was found from experiments that the seasonal crop water requirement for lettuce crop was 162 mm for 2 & 4 days irrigation interval & 183 mm for 7 days irrigation interval. An event of hail storm followed by rainfall of 11.8 mm lead to yield loss of 9%.

An investigation was carried out to evaluate nitrogen fertilizer application strategy, model the nitrogen dynamics under surface fertigation and suggest appropriate design and management guidelines for nitrogen application in surface fertigation. Two of four experiments were carried out under basin irrigation system with and without crop and the other two on non-cropped fields with furrow irrigation as well as raised bed irrigation systems. Three surface fertigation strategies were considered in the investigation, *viz.*, fertigation during entire irrigation duration (EI), fertigation during first half of irrigation duration (FH) and fertigation during second half of irrigation duration (SH). The traditional broadcasting method of fertilizer application was also considered in the study for comparison. WinSRFR and Abbasi models were used to simulate surface irrigation systems and to quantify nitrogen movement in different fertigation strategies, respectively. The results indicated the maximum application efficiency of 89% was found to be in furrow irrigation system. The best fertigation strategy in terms of yield of the crop was "fertigation during the second half of irrigation duration". The model was found to be suitable for simulating fertigation process especially for SH. The study concluded with a recommendation that the discharge rates of 1.5 lps/m and 2 lps/m were appropriate with 0.01% field slope for surface fertigation.

An innovative study was conducted to find out the suitability of drip irrigation for summer *mung* bean production. The treatment consisted of three irrigation methods *i.e.* drip, furrow irrigated raised bed (FIRB)

and check basin(CB) and irrigation regimes *i.e.* 100% ETc, 75% ETc and 60% ETc. Total water applied was 234 mm, 175.5 mm, and 140.4 mm in irrigation at 100% ETc, 75% ETc and 60% ETc, respectively. The study noted that all plant parameters eg. plant height, number of leaves per plant, no of branches per plant, total plant dry weight, leaf area index, crop growth rate at 30 and 45 days after sowing (DAS) and at harvest stage were significantly higher in 100 % Etc treatment that those of 75% ETc and 60% ETc. The yield attributes (number of pods per plant, number of grains per pod, pod length, 1000-grain weight) were recorded highest in drip irrigated treatment with 100% ETc. Interaction effects indicated the combination of irrigations by drip method and irrigation regimes at 100% ETc found to be superior to any other combinations of treatments. The study revealed that that even 60 % ETc with drip recorded significantly higher grain yield (9.17 q ha^{-1}) than that of CB and FIRB.

Regulated deficit irrigation (RDI) and partial root-zone drying (PRD) are potential water saving techniques for garden crops. The response of Kinnow (*Citrus reticulata* Blanco) mandarin to these techniques has not been studied in India. Keeping this in view, the present study was conducted for 2 seasons during 2010 and 2011, with drip-irrigated Kinnow mandarin plants budded on rough lemon (*Citrus Jambhiri* Lush) rootstock. RDI was scheduled with no irrigation and at 50% crop-evapotranspiration (ETc) applied in early fruit growth period (EFGP) and final fruit growth period (FFGP) singly and in combination, whereas PRD was scheduled at 50% ETc and at 75% ETc. Full irrigation (FI: irrigation at 100% ETc) was considered for reference. Irrigation at 50% ETc in EFGP and FFGP ($RDI_{50\text{-}100\text{-}50}$) was observed as the optimum RDI treatment, which saved around 30% irrigation water, producing only 3–6% less yield with better quality fruits compared with FI. Similarly, PRD at 50% ETc (PRD_{50}) proved superior, resulting in 35% improvement in irrigation water use efficiency (IWUE) with the fruits with comparable quality over that in $RDI_{50\text{-}100\text{-}50}$. The results revealed that the PRD (PRD_{50}) technique could impose desirable water stress on Kinnow mandarin plants, thus improving their fruit quality substantially, without affecting the fruit yield significantly.

Limited information is available on microirrigation fertigation and comparison between drip and microsprinkler fertigation. To address that gap field experiments were conducted with *Yamuna safed* variety of garlic to model the nitrogen transport dynamics under microirrigation (drip and microsprinkler) fertigation and to develop design and operation guidelines. Randomized block design was used with four treatments and three replications. The nitrogen level treatments were N0 (0 kg ha^{-1}), N40 (40 kg ha^{-1}), N80 (80 kg ha^{-1}) and N120 (120 kg ha^{-1}). Soil and plant samples were collected periodically along and across the lateral after each dose of fertilizer application (24, 48 and 72 h after fertigation). Observations on yield and quality parameters of garlic were collected after harvesting. HYDRUS-2D model was used for simulation of water and nitrogen dynamics in microirrigation fertigation. The study indicated that all the yield and quality parameters of the garlic were found to be best in the 80 kg N ha^{-1} with drip irrigation treatment. The results of simulation of various scenarios indicated that nitrogen distribution in different textured soils was significantly affected by the emitter discharge rate and fertilizer level. In coarse textured soils like sand, loamy sand, sandy loam and sandy clay loam nitrogen tends to move downward beyond root zone. In fine textured soils like clay, silt clay, silty clay loam, clay loam, silt loam and silt nitrogen moved outward in top two layers (0-15 and 15-30 cm). Data on nitrogen balance suggested that leaching of nitrogen beyond the root zone increased with the level of nitrogen applied in both (drip and microsprinkler) the method of irrigation. In all the soils 80 kg ha^{-1} fertilizer level with 1-2 Lph discharge rate was most suitable considering the water and nitrogen savings. Design and operational guidelines developed in this study are useful for farmers for growing garlic crop under microirrigation fertigation.

An investigation was carried with a goal of developing N and K fertilizer schedule; distribution of ammonium, nitrate and potassium in soil profile; and evaluation of economic returns under varying N and K fertilizer doses through drip fertigation for greenhouse grown sweet pepper of 8 months duration. Field experiments were conducted for two consecutive seasons [each season was October to May] of sweet paper. Eleven N and K fertilizer combinations, including nine combinations of urea and MOP, one ready mix dry liquid fertilizer (DLF) and a control, were considered and fertilizer was applied weekly in 32 equal split doses

through low head low cost drip system while irrigation was scheduled twice a week. In the investigation of nutrient distribution in soil profile, one notable finding was the accumulation of nitrate at the boundary of root zone and accumulation of ammonium concentration in the close proximity of emission points and in top layers. Among all the treatments, nitrogen @ 400 kg ha^{-1} (urea) along with potassium @ 360 kg ha^{-1} (MOP) was found to be the most promising in terms of yield and income. The highest average sweet pepper fruit yield was observed to be 62.2 t ha^{-1} in case of the conventional fertilizer treatment. Field data further, revealed that the use of urea as a nitrogen source retained more nitrate in surface layer making available to the plant absorption than that of DLF. The maximum net returns, benefit cost ratio and payback period for 1000 square meter greenhouse grown sweet pepper were found to be Rs. 171586, 2.2 and 4 years, respectively. Field data suggested that the net returns could be increased by 21 per cent (Rs. 30,000) by enhancing 25 per cent (Rs. 200) nitrogen as compared to the recommended dose of nitrogen (320 kg ha^{-1}). The above fertilizer rate with urea and MOP was found to be the best as far as yield, benefit cost ratio, net income and payback period were concerned. Information obtained from the study will be useful in operation and management of drip fertigation for enhanced production of sweet pepper.

Effect of methods of planting on growth, yield and economics of rice varieties under drip irrigation were evaluated. The treatments included traditional transplanted rice (TTR), direct seeded rice (DSR) and system of rice intensification (SRI) and four varieties Pusa 44, Pusa 834, Pusa 1121 and Pusa 1401. In DSR and SRI methods, water was applied through drip system. Total water applied was 1705 mm, 998 mm, and 984 mm in puddle transplanted rice, DSR, SRI methods, respectively. Growth parameters such as plant height, number of tillers m-2, dry matter accumulation, leaf area index, crop growth rate at 30 days after sowing (DAS) and at harvest stage was significantly higher in traditional method and at 60 DAS in SRI with drip irrigation. The maximum leaf area index, were recorded with the variety Pusa 1401. The yield attributes (number of panicles m-2, length of panicle, weight of panicle, number of filled grains per panicle) was highest under TTR. Similar result was observed in terms of 1000 grain weight and crop yield (grain yield, straw yield and harvest index). However, number of panicle and weight of panicle were maximum in SRI method with drip irrigation. Pusa 1401 gave significantly higher grain yield compared to other three varieties. SRI with drip irrigation gave higher field water use efficiency of 0.45 kg grain/m3, which was higher than that of TTR (0.30 kg grain/m3 of water) and DSR method with drip irrigation (0.27 kg grain m-3). Whereas, irrigation water use efficiency was higher in SRI method with drip irrigation (0.67 kg grain m-3). Interaction effects indicated that variety Pusa 44 along with TTR produced significantly higher yield than the other treatment Indian Agricultural Research Institute 79 combinations. Highest gross return (Rs. 105,640 ha-1), net return (Rs. 77,460 ha-1) and benefit cost ratio (2.7) was observed in TTR method followed by SRI under drip irrigation. Pusa 1401 has given the highest net return (Rs 97,076/ha) and benefit cost ratio (4.1) followed by Pusa 834 (Rs. 49,925.6 ha-1 and 4.1). Interaction effects indicated that Pusa 1401 with TTR method gave maximum net return (Rs. 101,028 ha-1) and benefit cost ratio (4.6).

Summer squash is one of the vegetable crops among the cucurbits which may be grown under the temperature rate of 15-25^{0}C. Plastic low tunnels are considered to be highly suitable and profitable for off season cultivation from December to March, when the open field is not favourable for growing such crop. In the back drop of this a field study was conducted with the aim to mitigate the negative effect of low temperature. The study included drip irrigation scheduling and recommended dose of fertilizer application in Australian green summer squash and aimed to observe the response of growth and yield of off season summer squash under plastic low tunnel. The experiment was laid out in split plot design with three replications. Irrigation treatments consist of three different irrigation water levels in terms of crop water requirement (CWR); (I_1 = 1 CWR, I_2 = 0.8 CWR and I_3 =0.6 CWR) and three N:P:K water soluble fertilization levels in terms of recommended dose of fertilizer (RDF) with the ratio 5:3:5 of (F_1 = 1 RDF kg ha^{-1} ; F_2 =0.8 RDF kg ha^{-1} ; and F_3 = 0.6 RDF kg ha^{-1}). Data showed that the summer squash yield and growth attributes increased with the increase of irrigation water level. However, the I_2 along with F_1 increased yields due to increase in the mean weight of the fruits and fruit number. The results of the study showed that fertigation of 0.8 CWR and 1.0 RDF under plastic low tunnel

produce the highest fruit yield to the tune of 31.2 t ha^{-1} which was significantly higher than that of the two other drip fertigation levels. The total crop water requirement was estimated to be 158 mm. The results of the economic analysis revealed that interactive effect of I_2 and F_1 resulted highest net income, maximum benefit cost ratio and lowest payback period.

Management of Water and Soil Resources

To quantify the surface water resources of the Tauru watershed of Mewat district of Haryana having a geographical area of 4021 ha, satellite imagery of the study area was classified into 5 major classes by supervised classification technique in ENVI and the surface runoff was determined by USDA SCS-Curve Number method. Five year average annual runoff was estimated to be 132 ha-m. The maximum and minimum annual runoff volume was also found to be 546 ha-m and 0.84 ha-m respectively. The average annual groundwater availability was found to be 794 ha-m, out of which 675 ha-m could be safely used for irrigation. To assess the water quality in the area, a linear combination of water quality parameters was formulated to form the Irrigation Water Quality (IWQ) index. This index was used to classify irrigation water with respect to three suitability classes namely; 'safe', 'marginal' and 'unsafe' for irrigation. It was found that groundwater quality of Tauru watershed was between marginal (53%) and safe (47%) for irrigation. A Decision Support System (DSSWST) was also developed to plan the suitable strategies for enhancing the crop productivity under different water availability conditions. The DSSWST enables farmers and end-users in making appropriate decisions to plan their crops in the watershed.

A study was carried out to characterize the soil and water quality and to investigate the economic impact of soil salinization on crop production in the area of Nuh block of district Mewat, Haryana. Gio-spatial maps of pH, EC, SAR and RSC were generated. Analysis demonstrated that 51.8 percent land area was under threat of saline and alkaline soil and only 20.6 percent of the total land was under normal soil suitable for crop production. The study area was also classified into four soil classes i.e. class I (normal soil), class II (medium saline), class III (saline) and class IV (sodic soil). A sample survey was carried out to study crop production and resource use pattern. Bajra and sorghum together, in *kharif* season, occupied 60.22 per cent of the total cropped area while wheat and mustard together, in rabi season, occupied 45.06 per cent of the total cropped area. The productivity and net returns of major crops were worked out which showed a declining trend with increase in the soil class sequence. Production function analysis in case of wheat, mustard and bajra crops showed the major explanatory variables having influence over the crop production. The significant regression coefficients in case of wheat were 'seed cost', 'irrigation numbers', 'irrigation cost' and 'capital cost'. In the case of mustard crop, 'capital cost' was having significant positive regression coefficients but 'machine cost' was having negative coefficient signifying its over use. 'Seed cost', and 'capital cost' were the significant positive variables in case of bajra crop. The study found that Soil class II and Soil Class III, in case of wheat, and Soil Class III in case of mustard were significant variables.

Pearl millet (*Pennisetum glaucum [L.] R. Br.*) forms the staple diet of a majority of the poor smallholders and poor consumers, in the rainfed regions of the country. Limited availability of soil moisture at critical stages of crop growth is major constraint of rainfed agriculture in semi-arid regions. *In-situ* moisture conservation practices minimises loss of valuable top soil, increases water available for plant use, improves soil physical properties and enhances soil fertility. In pursuit of understanding of soil moisture dynamics under different bio-engineering *in-situ* moisture conservation practices and its impact on pearl millet yield, an experiment was conducted in the during *kharif* season (June – October) of 2013 at the Research Farm of Water Technology Centre, Indian Agricultural Research Institute (IARI), New Delhi. The experiment was laid out following randomized complete block design (RCBD) with six treatments and three replications. These treatments were trench-cum-bund (20 cm depth of trench and 20 cm height of bund), bund (30 cm height), ridge and furrow (15 cm height), skip row planting (3:1), basin tillage (45 cm x 45 cm) and control.

It was found that basin tillage moisture conservation practice performed better among different practices with moisture conservation efficiency (88.8%) and highest yield (2.47 tons/ha) in pearl millet. Ridge and furrow also has higher moisture conservation capacity than trench-cum-bund, bund and skip row. Highest harvest index (26.94%) and highest benefit-cost ratio (1.82) in basin tillage poses potential utility under rainfed condition for pearl millet production.

AquaCrop model (v4.0) was used to simulate both pearl millet grain yield and total soil water. Calibration of the model was done using the experimental data of 2011 and validated using the data of 2012 and 2013. Absolute prediction errors of 1.70, 8.74, 14.12, 14.88, 3.27 and 7.25 % in 2012 and 4.45, 3.72, 9.27, 6.53, 8.14 and 21.05% in 2013 for BT, R & F, TCB, bunds, SR and control, respectively were observed when simulation was done for grain yield and compared with the calibrated year of 2011 data set with validated data set of 2012 and 2013. The model predictions were satisfactory for lower rainfall year of <600 mm, whereas for excess rainfall year (2013) model predictions were not very reliable. Similar observations were found for stover yield and soil moisture simulation. The simulation accuracy of crop growth model was better for basin tillage moisture conservation practice compared to others. Overall, the model was overestimating for soil moisture prediction. The validated AquaCrop model can be used for prediction of pearl millet yield and soil moisture with acceptable accuracy under different water conservation practices in a semi arid environment.

Basin tillage method of water conservation practice

Overall view of water conservation practices

Soil Chemical analysis

Based on above results, following conclusions can be quoted at this point as

- Basin tillage has been found best *in-situ* moisture conservation practice among different practices in conserving moisture and ultimately with highest yield in pearl millet.
- Ridge and furrow also has higher moisture conservation capacity than trench-cum-bund, bund and skip row. This conservation practice poses potential utility under rainfed condition for pearl millet production.
- Economic feasibility study suggests that basin tillage has high cost of cultivation than any other conservation practices but high gross and net returns makes it more economically feasible than other *in-situ* moisture conservation practices.
- Evaluation of more *in-situ* conservation practices under drought condition will bring more precision in experimental results and significant differentiation among practices can be established.
- AquaCrop can be a valuable tool for modelling both crop yield of pearl millet and soil water content, particularly considering the fact that the model requires a relatively small number of explicit and mostly intuitive input data.
- Deviation in model performance needs finer calibrations by including more characteristics of the crop under diverse climate and soil conditions may improve the validity of the model.

- Limited specification to different conservation practices brings discrepancy in simulation. This issue must be addressed to develop better options for different field management to include *in-situ* moisture conservation practices.
- Over all, the FAO AquaCrop model predicted pearl millet yield and simulated soil water with acceptable accuracy under various conservation practices.

Climate Change and Water Resources Planning for Irrigation

Impact of climate change on water requirement and crop yield in central Vidarbha Climate variability and its impact on crop water requirement and food production are major concerns in the 21st century. Rise in temperature due to climatic variability is expected to affect the crop water requirement and food production all over the world. It is predicted that crop water requirement in arid and semiarid regions would increase due to rise in temperature. However, overall impact of climate change on crop water requirement and yield is still uncertain. Keeping in view the above concerns, a study was undertaken to evaluate the trend and the variability of rainfall and temperature, to assess their effect on crop water requirement and yield of major crops in the Central Vidarbha region of Maharashtra. Rainfall and temperature were analyzed using parametric and non parametric tests such as regression analysis and Mann Kendall test, Modified Mann Kendall test, Sen's slope estimator. ArcGIS 9.3.1 was used to prepare, the land use map, soil map and spatial variability map of rainfall. Auto Regressive Integrated Moving Average (ARIMA) model was used for forecasting future rainfall and temperature trend. Estimation of crop water requirement was done by standard Penman Monteith method using CROPWAT 8.0 model. Climate change scenarios considered for assessment of climate variability impact on crop water requirement were based on ARIMA, INCCA and IPCC predictions. Whereas, yields were predicted using yield response factor available in CROPWAT. Results indicated that average annual temperature and reference evapotranspiration increased marginally during 1969 to 2009, on the other hand, total annual and monsoonal rainfall decreased during this period. It was observed that there was a shift in the temporal rainfall pattern over the region. Maximum temperature had decreasing trend during pre-monsoon season whereas during monsoon and post-monsoon seasons it exhibited increasing trend. However, minimum temperature showed increasing trend during all seasons except winter. Crop water requirement of various crops grown in the Central Vidarbha region would decrease by 4.6 to 11.2 mm by 2030 when it is determined using all climatic parameters. Study further indicated that, under different prediction scenarios which considered the only rise in temperature, crop water requirement of almost all the crops increased but there was reduction in the yield of different crops from 0.3 to 5 per cent.

The impacts of climate change on water availability and crop production in the Gomti River basin of India were investigated by integrating a widely used hydrological model, Soil and Water Assessment Tool (SWAT) and the output of general circulation model, MIROC3.2 (HiRes). Climate change scenarios were developed for A2, A1B and B1 emission scenarios for three future periods 2020s (2010-2039), 2050s (2040-2069) and 2080s (2070-2099). These climate change scenarios were downscaled to the Gomti basin by delta change approach. The study also tested the trend of the historical climate and its relation with the river discharge of the Gomti basin using Mann-Kendall (MK) test, Sen's slope estimator, and modified MK test.

Climate change scenarios were downscaled to the Gomti basin by delta change approach. Simulation results showed that mean annual rainfall is likely to increase by 10 to 18 %, 15 to 47% and 20 to 30% during the time period 2020s, 2050s and 2080s respectively and the associated mean annual stream flow is projected to increase by 16 to 40 %, 26 to 76 % and 35 to 52 %, respectively for the same period. Projected surface water resources (SWRs) of the basin on annual scale are also likely to increase by 7 to 29%, 17 to 35%, and 35 to 46% during the 2020s, 2050s and 2080s, respectively compared to present day climate. Moreover, relative mean monsoonal changes of rainfall, stream flow and SWRs are approximately consistent with the mean annual changes of the

same. Changes of rainfall, stream flows, and SWRs are greater at upstream than the downstream. SWAT model was also used to simulate rice and wheat production with the changed hydrology of the basin under the climate change. The results showed that mean annual rice yield is likely to increase by 2 to 5%, 12 to 18% and 20 to 28% during the time periods 2020s, 2050s and 2080s, respectively, under the three emission scenarios (A1B, B1, and A2). Mean annual wheat yield is also likely to increase by 9 to 11%, 13 to 17% and 15 to 20% for the same period. This study also examined the historical hydro-climatic trends of the basin. Upstream gauging station showed significant increasing trend of stream flow while mid to downstream gauging station showed decreasing trend in stream flow at annual scale. The results obtained in this study could be useful for planning and managing irrigation water for crop production in the Gomti River basin.

The study involved analysis of long term behaviour of terminal heat and its effect on wheat yield of selected locations (Agra, Bathinda, Hisar, Karnal, Ludhiana and New Delhi) of semi arid region of India for the period spanning from 1984-85 to 2010-11. The daily positive temperature departure by ≥2 °C above long-term normal, for at least continuous five days was taken into consideration for terminal heat stress detection during late vegetative and reproductive stages. Apart from this, a field experiment was also conducted to study the effects of sowing dates and irrigation regimes on growth parameters, yield attributes, yield, water use efficiency (WUE) and quality parameters of wheat. The staggered sowing from 1st November to 16th December and four irrigation regimes, *viz*. 25% maximum allowable depletion (MAD), 50% MAD, 75% MAD and 4 irrigations at critical growth stages were tested.

Long term temperature data analysis revealed that both T max and T min have decreased or increased in the range of -0.02 to 0.49°C 10 yr^{-1} across the selected locations. The average wheat yield reduction was 650 kg ha^{-1} with every unit increase of T mean in a range of 17-19 °C during wheat growing season. The combined effects of positive departure and stress duration revealed that the effect was more pronounced at late vegetative stage (31.6°C days) than reproductive stage (29.1°C days). The T max less than 17°C, 24°C and 30°C during January, February and March, respectively, produced the highest wheat yield at most of the locations.

The results of the field experiment revealed that the significantly highest crop growth, yield attributes and yield (5.04 and 5.36 t ha-1 in the first and second season, respectively) were obtained with 1st November sowing but was at par with 16th November sowing. The grain yield reduction was in the range of 25-30% and 42-45% in 1st December and 16th December, respectively in comparison to 1st November. Each day delay in sowing beyond November 16 led to a yield penalty of about 1%. The grain quality such as test weight, hectoliter weight, grain diameter and flour recovery were decreased with delay in sowing and irrigation frequency. Whereas, protein and dry gluten contents of the grain increased with delay in sowing and irrigation frequency. Study on soil moisture use revealed that maximum soil moisture extraction took place from 0-30 cm soil depth across sowing dates and irrigation regimes. As a result of advanced sowing time from November 16th to November 1st, yield improvement was in the range of 2.4-3.3%. Findings of the present investigation suggest that the sowing during first week of November and irrigations scheduled at 25% MAD of ASW could be helpful to some extent in reducing the risk of terminal heat stress of wheat grown in semi-arid region of India.

Surface Irrigation Design and Operation

A field study was conducted to evaluate the performance of furrow irrigation system for carrot crop in sandy loam and loam soil. The inflow rates of 1, 1.5 and 2 lps were applied to furrows with 20, 25 and 30 m lengths. Nine treatments with three replications each were considered. The depth of water applied and stored in the root zone of the crop was determined by measuring soil moisture content before and after each irrigation event. An inflow cut off criteria for each furrow was based on 80% of the water front advance distance. The time of advance was also observed for each irrigation event at equal distance along the length of furrow. Different design variables were measured such as furrow slope, furrow length, furrow width, furrow depth, inflow rate and the

time of cut off. The estimation of infiltration parameters 'k' and 'a' of Kostiakov's infiltration equation was done by Elliot and Walker two point method. The data were analysed for water application efficiency, water storage efficiency, water distribution efficiency and field water use efficiency and subsequently the performance of furrow irrigation method with different lengths were assessed. The results showed that the furrow lengths of 20, 25 and 30 m gave the best performance with the inflow rates of 1, 1.5 and 2 lps, respectively.

The data generated through field experiment were used for simulation of water distribution using Surface Irrigation Simulation software (ver.4.1.3). The root mean square error (RMSE) and coefficient of variance were used as model performance indicators. Simulation for time of advance was in close agreement with observed values and for water application efficiency was also found satisfactory.

Border/basin irrigation events were monitored under wheat crop to collect data on waterfront advance, inflow rates, topography and time of cutoff (T cutoff). Three irrigation events at three locations at IARI farm were monitored, one each in bare soil, initial crop stage and middle crop growth stage. Three locations at IARI were WTC plot, Mid-block and New area. Infiltration characteristics at these three locations were collected using double ring infiltrometer. The collected data were used for estimating surface irrigation parameters (Kostiakov infiltration parameters and Mannings' roughness coefficient) using inverse methodology in WinSRFR model. These parameters are required for simulation of irrigation hydraulics for developing design and operational guidelines for surface irrigation. For each location at IARI, farm design and operational guidelines were developed for various field sizes and inflow rates. Surface irrigation parameters for infiltration and roughness characteristics for three locations at IARI showed insignificant variations. Operational guidelines for surface irrigation layouts were developed for IARI farm and general operational guidelines were also prepared using simulation module in WinSRFR model. Tcutoff was determined for IARI farm to achieve 75 percent or more application efficiency. Different combinations of field sizes for various inflow rates were also simulated to determine T cutoff for targeted application efficiency of 60 percent and 75 percent with water requirement efficiency of 100 percent. Surface irrigation operational guidelines prepared will be ready reckoner for farmer for adopting best border/basin size/layout for achieving higher application efficiency.

Diversified use of Stored Groundwater for Aquaculture

Water productivity has been defined as the ratio of net benefits of crops, forestry, fishery, livestock and mixed agriculture systems to the amount of water used to produce those benefits. IARI maintains four small ponds to store groundwater and rainwater for interim storage before it is distributed to various farms for irrigation. The hypothesis of this study was to evaluate the feasibility of carps (Fish) production in the existing ponds with IARI groundwater. Water quality is one of the main concerns in aquaculture and any adverse change in the chemical or physical properties of water may cause a native effect on the growth of carps. Major objective of the study was to monitor spatio-temporal groundwater quality for fish production. A field trial was conducted in four ponds located in CPCT and FOSU premise with the stocking density of 0.13 fish/m^3. Growth parameters like weight, length height and width of fish carps were recorded in all the four ponds periodically. Water quality data were also collected and analysed with respect to all four ponds. Results indicated that Dissolved Oxygen (DO), Biological Oxygen Demand (BOD) was found to be suitable for the view point of water quality for fish production. The data further revealed that the pond number 3 located at MB 7 (FOSU) was the most productive.

Fish being harvested in CPCT farm

The average net weight of fishes sampled in pond 3 was found to be maximum of 2.5 kg. The reason for the higher productivity in pond 3 was attributed to fruits dropping from various kinds of trees that existed on the embankment of the pond, excreta from birds and presence of phytoplankton in abundance. The study concluded that the stored groundwater was found to be suitable and technically feasible for different types of carps production during August to March.

Climate Change and Conservation Agriculture

The effect of conservation technology practices on soil moisture balance, the impact of conservation practices and nitrogen levels on yield, and the energy consumption and economics for production of sorghum (Sorghum bicolor (L.) Moench), variety: 'CSH 14' under rainfed conditions were investigated. Two types of tillage (conventional and minimum) and two methods of planting (ridge and furrow and flatbed) were introduced. For each type of tillage and method of planting three levels of nitrogen were applied under Split Plot Design. Soil moisture content was monitored periodically throughout the seasons by both TDR based Field Scout 300 (volumetric basis) at three soil depths (8, 12 and 20 cm), and gravimetric method for the soil layers (0-15, 15-30, 30-45, 45-60 and 0-60 cm) to study the soil moisture retention in each treatment. The results revealed that the ridge and furrow planting method in conventional tillage retained the highest soil moisture content leading to the highest grain and fodder yields amongst the other treatments, which were found to be 2.16 and 1.90 t/ha and 11.21 and 7.65 t/ha with the 100% recommended dose of fertlizer (RDF) for both the growing seasons respectively. Yield attributes were affected by rainfall amount between the two crop growing years. The treatments affected 1000 seeds weight significantly and the plant moisture content was affected by sowing method and tillage type regardless of fertilizer levels. The ranking of treatments in grain and fodder yields were similar to the ranking of retention of soil moisture retention. The total energy used for producing sorghum was found to be 6.94×103 and 7.21×103 MJha-1, the net energy gained 90.27×103 and 77.45×103 MJha- 1, the output-input energy ratio 14.00 and 11.74, the specific energy 4.57 and 6.16 MJ kg-1 and energy productivity be 0.22 and 0.16 kgMJ-1 respectively for the two crop growing seasons. The total expenditure for production was 7.51×103 and 7.92×103 Rsha-1, the gross production value 17.51×103 and 14.49×103 Rsha-1, net return gained 10.00×103 and 6.57×103 Rsha-1, and the benefit-cost ratio 2.33 and 1.83 respectively for the two seasons. The ridge and furrow planting in conventional tillage with 100% RDF, was found to be the best planting system for sorghum production. Although, this combination consumed more energy (106.8%) and expenditure (32.7%), but with higher return of energy (155.3%), money (139.1%) and higher soil moisture retention (32.5%) with higher yield (55%) in comparison to the flatbed planting in minimum tillage with 100% RDF. The study suggested adoption of ridges and furrows method of planting in conventional tillage with 100% recommended dose of fertilizer as the one of the best approach for obtaining higher grain production of sorghum and conservation technology of soil and water resources under rainfed conditions

Resource conservation technologies such as conservation agriculture and efficient irrigation scheduling especially under deficit water availability offer a challenge to examine the suitable combination of both for sustainable wheat production with minimum detrimental to soil health and environment. Therefore, a field study was under taken to study the soil moisture availability under deficit irrigation based conservation agriculture practices and evaluate the response of wheat with/without conservation agriculture practices under deficit irrigation. The conservation agriculture practices such as permanent raised bed + maize residue, minimum tillage + maize residue and conventional tillage without residue along with three irrigation levels viz. 100%, 80% and 60% of ETc were examined.

Results indicated that LAI, dry matter accumulation and crop growth rates were found to be highest under permanent raised beds and irrigations scheduled at 100 % of ETc. Other yield attributing characters such as effective tillers/m^2, spike length, grains per spike were also significantly more under permanent raised bed as compared to the zero tillage with residue and conventional tillage without residue. Amongst irrigation schedules,

irrigations based on 100% of ETc resulted in significantly higher growth and yield attributes than 80% of ETc and 60% of ETc. Similarly, grain yield and biological yields were higher under permanent raised bed (19.22 and 9.07%) followed by zero tillage with residue (8.7 and 17.6%), respectively than conventional tillage without residue. Permanent raised bed with residue showed highest water productivity (1.40 kg/m^3) and it was 8.49 and 18.95% more compared to the zero tillage with residue and conventional tillage without residue, respectively. However, amongst irrigation schedules, 80% of ETc recorded highest water productivity (1.48 kg/m^3) registering 5.4 and 14.86 % more than the 100% and 60 % of Etc respectively. Interaction effect of conservation agriculture technique and irrigation regimes indicated that permanent raised bed with residue retention and 80% of ETc irrigation schedule gave comparable yields and net return to 100% of Etc irrigation schedule but consumed less water and recorded higher water use efficiency. Thus, the findings of the study undertaken suggested that the permanent raised bed with residue retention and irrigations scheduled at 80% of ETc can be regarded as the most inputs effective and profitable method for wheat cultivation.

Intensive cultivation under traditional method of field preparation involving about 6-8 rounds of conventional tillage and indiscriminate application of irrigation have invariably given rise to a host of problems that are threatening the sustainability of crop production system. Therefore, implementation of resource conservation technologies such as conservation agriculture practices along with proper irrigation scheduling, offer a promise for sustainable crop production with minimum environmental degradation. Keeping above facts in view a study was conducted for two consecutive years with the objective to determine the water and energy use efficiency, growth, productivity and economic feasibility of maize-wheat cropping system under different conservation agriculture practices and irrigation scheduling. Treatments comprised of conservation agricultural practices (CAS) i.e. raised bed (RB), zero-tillage (ZT) and conventional tillage (CT) and irrigation regimes viz. irrigation at 40, 50 and 60% of available soil moisture depletion (ASMD) for *kharif* maize (var. HQPM1). During rabi season wheat (var. DBW 17) was grown with same conservation agricultural practices were the same as followed during *kharif.* However, the irrigation schedules were different i.e. critical growth stages, crop water stress index (CWSI) and 60% ASMD along with residue management practices.

Raised bed planting of maize resulted in significantly higher grain yield and net returns followed by zero tillage planting and conventional tillage (CT) flat bed planting. Similarly, energy use efficiency was more under RB while the lowest under CT. The percentage increase in IWUE of maize was about 40-46% and 16-10% under RB and ZT respectively compared to CT. Planting of maize on raised bed improved the overall productivity and profitability in comparison to other systems. Residue retention was beneficial in terms of water productivity and yield of maize than removal of residues. During *rabi* season, wheat grain yield was higher under RB (6-13%) in comparison to ZT and CT. The percentage increase in irrigation water use efficiency was found to be higher in RB (32-40%) followed by ZT (8-12%) and the lowest with CT. The overall productivity of maize-wheat system was significantly higher under raised bed than the other conservation agricultural practices. The yields of both maize and wheat under maize-wheat cropping system and overall system productivity were improved due to retention of residues than removal. The Soil physical properties, viz. hydraulic conductivity, infiltration rate, soil aggregation, organic carbon as well as biological properties were improved with RB and ZT practice compared to conventional tillage practice. To achieve higher and sustainable resource-use efficiency, productivity and profitability from maize – wheat cropping system, study indicated that the conservation agricultural practices like RB and ZT are the essential components irrespective of the irrigation scheduling. Similarly, irrigation scheduling based on soil moisture depletion resulted not only in saving of irrigation water but the higher yield of the system also.

A field study was conducted during *kharif* season in 2015 to study the water productivity and yield of rice under different methods of establishment and nitrogen levels. The treatments consisting of three methods of establishment i.e. traditional puddled transplanting, system of rice intensification (SRI) and SRI with sub-surface drip and four nitrogen levels i.e. 80, 100, 120 and 140Kg N/ha. The irrigation schedule of continuous submergence, 2-3 day drainage period and on the basis of daily Etc was followed for traditional method of transplanting, SRI and SRI with drip irrigation method, respectively.

Rice grown under SRI method of establishment resulted in higher number of tillers per m^2, dry matter accumulation, leaf area index, crop growth rate and relative growth rate at all stages of crop and grain yield followed by traditional transplanting, whereas these parameters were the least under SRI with sub-surface drip method. The significant response of N levels was observed up to 120 kg N/ha thereafter, no significant difference in above mentioned parameters and grain yield was noticed.

Irrigation requirement was highest under traditional puddle transplanting (1105 mm) followed by SRI (775 mm). The least irrigation applied was under sub-surface drip system (452 mm). Saving in irrigation water under SRI and SRI with sub surface drip system was 22.93% and 45.38% than puddle transplanted rice, respectively. Similarly, the total water requirement including effective rainfall of 334 mm was highest under traditional puddle transplanted rice (1439 mm) followed by SRI (1109 mm) and SRI with sub-surface drip irrigation system (786 mm). Though the SRI with sub-surface drip irrigation required less water resulting in higher irrigation (0.96 kg grain/m3 water) and field water use efficiencies (0.55 kg grain/m3 water) but the grain yield was significantly less than the other methods of crop establishment.

Rice cultivation encounters twin challenges of producing more rice per unit area with less water. A shift from puddled transplanting to direct seeding of rice (DSR) supported with conservation agriculture (CA) based crop management practices has been perceived to address these challenges. Again, water budgeting in rice field for irrigation scheduling, and prediction of rice yield by crop growth model are immensely important for planning on water savings and climate change mitigation aspects. Hence, a field experiment consisting of eight treatments, ZT DSR - ZTM: zero till direct seeded rice (ZT DSR) - zero till mustard (ZTM), ZT DSR + BM - ZTM: Zero till DSR + Sesbania brown manuring - ZTM; MR (mustard residue) + ZT DSR - RR (rice residue) + ZTM, MR + ZT DSR + BM - RR + ZTM, ZT DSR - ZTM - ZT SMB (summer mungbean) (without any crop residue), MBR(Mungbean residue) + ZT DSR - RR + ZTM - MR + SMB (with three crops residue), TPR (transplanted puddled rice) - ZTM, and TPR - CTM (conventional till mustard), which was laid-out in a continuing experiment (6^{th} year) at ICAR-IARI, New Delhi during *kharif* 2015 to estimate and analyse various water budgeting parameters which values were taken earlier from assumptions given by FAO, and simulation of crop growth model AquaCrop.

Irrigation scheduling was done following soil moisture depletion method, water budget parameters *viz.* crop water requirement, water to be applied for irrigation, puddling, ponding, nursery raising, runoff, effective rainfall were estimated using FAO water balance equation and amount of irrigation water applied was measured using a star flow meter. All other biophysical parameters including root characteristics were also determined at every stages of crop. The results revealed that in the 0-15 cm soil layer, both DSR and TPR plots exhibited similar soil moisture content (SMC), however, the SMC in MBR+ ZT DSR -RR+ ZTM - MR + SMB treatment was ~12 to 51 % and 2 to 32% more than TPR-ZTM and TPR-CTM treatments plot, respectively in the 15-30 cm

Soil Chemical analysis

Data recording from experimental plot

Water measurement using starflow meter

soil layer at the different crop growth stages, leading less irrigation water application and deep percolation in DSR with CA than TPR. The total amount of irrigation water applied in rice ranged 893.4 - 954.3 mm in DSR plots and 1418.2 - 1464.6 mm in TPR plots. The amount of water used for puddling and ponding in TPR were 135 and 94-98 mm, less than that the FAO assumed values of 200 and 100 mm, respectively. The best treatment (MBR+ ZT DSR- RR + ZTM - MR + SMB) showed deep percolation of 5.8 mm day^{-1} that was 25.6% lower than and the highest deep percolation observed in TPR-CTM treatment (7.8 mm day^{-1}). The different growth, yield parameters and water productivity were recorded highest under MBR+ZT DSR–RR+ ZTM-MR+SMB treatment among other DSR and TPR plots.

Rice under the MBR+ ZT DSR – RR + ZTM - MR + SMB treatment showed higher plant height, leaf area, leaf area index, relative water content, and chlorophyll concentration of the plants. There was slightly increase in organic carbon and available N, P, K in DSR plots compared to transplanted plots at 0-15 cm depth. It was observed that the maximum and minimum error in grain yield prediction during calibration (year 2014) of AquaCrop model in MBR+ZT DSR–RR+ZTM-MR+SMB (best treatment) and TPR-CTM (control) was 3.6 and 0.19 %, whereas during validation (year 2015), 14.88 and 9.83 %, respectively. Overall, it was observed that the yield and water productivity prediction by the AquaCrop model in the experiment were in line with the observed values though there is some over estimation. In summary, DSR with crop residues for three seasons with rice, mustard and mungbean should be retained to have more crop and water productivity, and water savings of 56% compared to puddle transplanted rice.

Based on the above results, following conclusions were drawn:

- MBR+ ZT DSR – RR + ZTM + MR – SMB is the best treatment for conservation agriculture and DSR cultivation
- The cropping system rice-mustard-mungbean may be taken to get more benefit w.r.t water saving, biophysical parameters, soil water and nutrient availability
- DSR is more useful than TPR which saves 56% of water
- Water balance amount is site specific, so estimation of water balance for irrigation scheduling is essential
- The calibrated and validated Aquacrop model can be used for rice grain and biomass yield production under direct seeding rice (DSR) and conservation agriculture (CA)

Rainfed agriculture hinges the food and livelihood security of a large number of people worldwide. However, production of rainfed areas is constrained by the extreme variability in rainfall, poor nutrient status of soils, land degradation through erosion etc. Under such situations, agri-horti and other agro-forestry land use systems are the best option to protect the land from various types of degradation, diversifying the farming system and increasing the soil carbon build up and moisture. These benefits of agro-forestry can be improved by incorporating *in-situ* moisture conservation practices, and legume crops intercropped with fruit trees may provide important source of cash and income. However, less information is available on the effects of legumes in conjunction with trees. Therefore, a field experiment on water balance and legume productivity under different moisture conservation practices in rainfed agri-horti system was carried out at Research Farm, Water Technology Centre, ICAR-Indian Agricultural Research Institute, New Delhi during *kharif* season 2015 on a sandy loam soil in split plot design with four replications under rainfed agri-horti system with three legume crops (soybean, cowpea, and mungbean) grown under *Bael* trees and three different moisture conservation practices of trench, micro-catchment (MC), ring basin (RB) and no moisture conservation (NMC) practices under tree and no tree (NT) as control. Soil moisture content was measured periodically from 0-15, 15-30, and 30-60 cm soil depth and volume water balance approach was used for water budgeting. All water budgeting components, soil properties and biophysical parameters of crops and trees were recorded/analysed using standard procedures.

Higher soil moisture content was observed in trench under soybean and mungbean plots, whereas, micro-catchment in cowpea plots. Soil moisture balance depicts that there were soil moisture storage and deficit

Field experimental plot

Bael harvesting

Visiting of Director and Dean IARI, to the experimental plots

Soil chemical analysis

of 281.8, 269.1 and 274.2 mm; and 246.3, 31.5 and 63.75 mm in soybean, cowpea and mungbean plots, respectively under agri-horti system. Irrespective of the moisture conservation practices, the *Bael* yield was in order of cowpea plots > green gram > no crop plots > soybean plots. This indicates that inclusion of cowpea and green gram enhanced the *Bael* yield. Cowpea + *Bael* agro-forestry was the best yielder in terms of cowpea seed equivalent yield when plots were provided with trench treatment. Under other moisture conservation practices also this system gave higher cowpea seed equivalent yield than green gram + *Bael* and soybean + *Bael* agro-forestry system. Within green gram + *Bael* agro-forestry system, trench and ring-basin yielded higher than micro-catchment and no-moisture conservation practices. In soybean + *Bael* system, trench followed by micro-catchment were better moisture conservation options. Overall, *Bael* tree intercropped with cowpea incorporating with trench moisture conservation practice can fetch more income and enhancing soil fertility under rainfed conditions of arid and semi-arid climate.

Based on the above results, following conclusions can be drawn as follows:

- Water balance amount is site specific, so estimation of water balance for performance evaluation of moisture conservation practices is essential.
- Trench moisture conservation practice is the best moisture conservation practice to enhance soybean and green gram productions, whereas micro-catchment is better to increase cowpea production individually without considering the agroforestry system.
- Similarly trench, micro-catchment and trench moisture conservation practices are produced higher cowpea equivalent yield in soybean, cowpea and green gram, respectively irrespective of agroforestry system.
- *Bael* fruit yield can be enhanced by inclusion of cowpea and green gram.
- In legume based *Bael* agro-forestry system as a whole, trench moisture conservation practice is the best treatment to produce overall cowpea seed equivalent yield.
- Overall cowpea + *Bael* agri-horti system is the best option under legume based alternate land use system.

Soil Health, Poor Quality and Waste Water Management

The present research work aimed at modelling the adsorption and plant uptake of Ni, Cr and Pb in the vertical subsurface flow constructed wetlands (VSSF CWs) of the Indian Agricultural Research Institute. Time series analysis indicated that the plant metal uptake was the highest in summer and the lowest in winter, regardless of plant types and applied metal concentration. The adsorption percentage of Ni on gravels in the VSSF CWs decreased with time but stable for Cr and Pb regardless of their concentration levels. The analysis of auto correlation functions indicated that no seasonality was detected in the data series. The adsorption kinetics of heavy metals on gravels showed that the removal of Ni ranged from 34.8 to 47.2 % with an average

of 40.7 %. Similarly, the adsorption of Cr varied from 42.7 to 54.9 % and Pb from 47.5 to 56.9%, indicating the following and decreasing adsorption order: Pb > Cr > Ni. The maximum adsorption of Ni and Cr was achieved within 12 hours while it was 6 hours for Pb. Freundlich model showed a good fit for Ni and Cr dataset (R2 > 0.9) while Langmuir model fitted better for Pb (R2= 0.7). Equally, pseudo-second-order model showed the best fit that is suitable to simulate the adsorption rates of these metals on gravel.

The gravel material showed good adsorbent capacity (kF> 2 mg/kg) for Ni, Cr and Pb though the adsorption intensities were low (n < 1). It was observed that the coefficient of determination values (R2) of linear models (simplified Freundlich) vary relatively from one model to another from 0.70 to 0.96 irrespective of the process (plant uptake or adsorption), the type of plant, the metal species and the level of metallic concentrations applied. These high values of R2(>0.70) indicated that there is a significant (p-value < 0.001) strong (positive or negative) relationship between metal concentrations in plants or in gravel materials and the metal concentrations in influent. Concerning the mathematical models developed in the STELLA simulation programme, the experimental and simulated data for Ni, Cr and Pb at different concentrations were compared for the adsorption and plant uptake in VSSF CWs using the Wilcoxon Signed Ranks Tests. Irrespective of the processes (adsorption, plant uptake and metal contents in effluent), a good agreement was found between experimental and simulated data indicated by large p values (Asymp Sig. > 0.05). These findings supported the application of different models developed for the effective design and simulation of the dynamics of metals in constructed wetlands

For developing the alternate low cost and eco-friendly sorption technology for wastewater treatment, laboratory studies were conducted at IARI, New Delhi to characterize cellulosic agri-waste and evaluate their potential for removal of heavy metals from municipal wastewater. Keeping in view the generation potential availability, agri-waste chosen for adsorption were: rice husk, rice straw maize cob pith, soybean straw, saw dust and coir pith. The performance of these wastes was compared with activated charcoal. The wastewater initially had high biological oxygen demand and contained Ni, Cr and Pb 0.26, 0.28, 1.03 mg l^{-1} respectively. Metal levels were spiked in the wastewater to achieve 15.0, 20.0 and 6.0 mg l^{-1} of Ni, Cr and Pb. The important functional groups observed in majority of the adsorbents were amides, alkenes, alkyl halides and carboxylic acid. The adsorptions kinetic study showed that 6 hr retention time was sufficient to remove much of the Ni, Cr and Pb from the wastewater, thereafter removal efficiency increased only slightly with increasing time from 6 hr to 24 hr. Activated charcoal performed very well and removed almost all the amount of Ni, Cr and Pb from the wastewater. Amongst different bioadsorbents, coir pith, saw dust and rice husk were found more efficient than maize cob pith and rice straw. Setting the treatment efficiency at 80% level, 100 g of rice husk, saw dust and coir pith packed in the column was able to treat about 2 litre of wastewater containing individual concentration of 15.0, 20.0 and 6.0 mg l^{-1} of Ni, Cr and Pb. But considering higher cost and lesser availability of activated charcoal, low cost locally available cellulosic materials like coir pith, rice husk and sawdust can be exploited for efficient removal of heavy metals from municipal wastewater.

Future Researchable Issues in the stream of Agricultural Water Management

- Surface irrigation hydraulics and their design and operational guidelines.
- Water management of vegetable and pulse crops under varying water supply situation.
- Crop water requirement and sap flow estimation in fruits crops of different ecological zones.
- Development of operational guidelines for surface irrigation using sensors.
- Crop response and nutrient dynamics under drip fertigation using liquid fertilizer.
- Impact of climate change on water availability and crop production in river basin.
- Water conservation and harvesting under rainfed and limited irrigation.
- Water resource planning on watershed basis using RS and GIS.
- Hydrologic modeling

- Safe use of wastewater in agriculture & eco-friendly wastewater treatment.
- Poor quality of water use in agriculture

Acknowledgement

I wish to thank all the Faculty Members of the Division of Water Science and Technology for extending support and cooperation in preparing the research contributions of M.Sc. and Ph.D. students.

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School of Basic Sciences

Biochemistry

Aruna Tyagi

Introduction

The Division of Biochemistry was created in 1966 as a separate Division, with major emphasis on Molecular Biology, Plant Biochemistry and Nutrition. It owes its origin to a section of Plant biochemistry in the Division of Soil Science and Agricultural Chemistry. Since its inception, the Division has acquired the best research, teaching and training facilities, and takes legitimate pride in being recognized as the only Centre of Advanced Faculty Training (CAFT) in Biochemistry in NARS by ICAR ever since 1995. Currently, the Division with limited scientific strength is conducting research on potentially applied aspects in the fields of Plant Biochemistry and Molecular Biology with major emphasis in the areas of abiotic stress tolerance and quality improvement in cereals and oilseed crops. The Biochemistry faculty is actively involved in the teaching curriculum offering 54 credits distributed over 15 courses for M.Sc and Ph.D in the premier PG School, IARI.

The mandates of the division are:

- To undertake fundamental and applied biochemical, molecular biological and nutritional research related to the current problems in agriculture.
- To conduct teaching and training programmes to develop skilled human resources.
- To impart training in advanced techniques in the fields of Plant Biochemistry and Molecular Biology to agricultural scientists of State Agricultural Universities and ICAR institutes.
- To develop instructional aids for teaching in Biochemistry.

Significant Student's Research Achievements

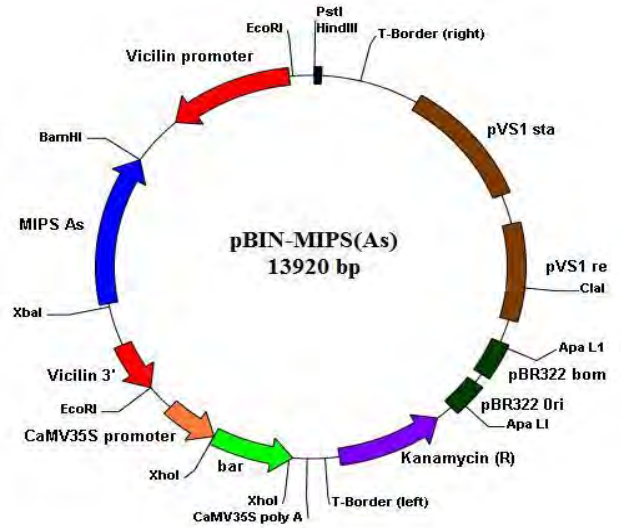
Nutritional Enhancement

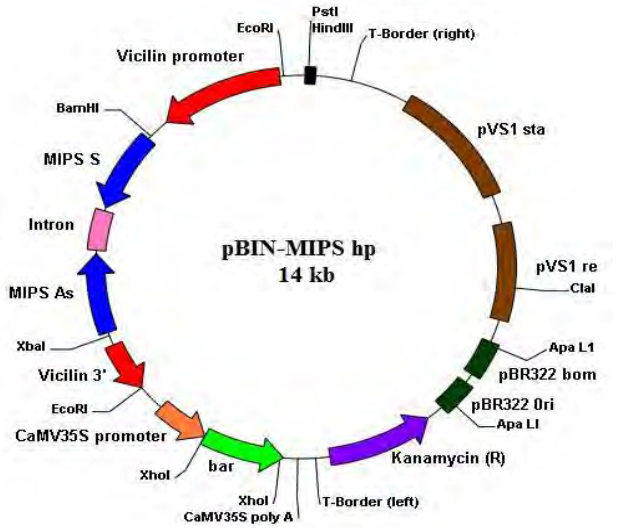
Phytase gene from soybean (*Glycine max* var. Pusa 16) was amplified using long range PCR. Cloning and characterization of the genomic sequence revealed a total length of 4220 bp containing 6 introns interrupting 7 exons. The transcribed sequence of the gene had an expected open reading frame of 1644 bp predicted to encode for 547 amino acids including an N-terminal signal peptide of 28 amino acids confirming its secretory status. Strong homology with the previously reported cDNA *(GmPhy)* as revealed in the BlastN search indicated high degree sequence similarity to purple acid phosphatases including the conserved motifs and the signature metallophosphoesterase domains. mRNA expression for phytase gene as evaluated in the germinating cotyledons by RT-PCR and Northern analysis revealed maximum transcript levels at around 11 days after germination. Single copy of the gene was detected during Southern analysis of the genomic DNA. Developing soybean seeds were analyzed for total phosphorus and phytate-phosphorus contents whereas germinating soybean seeds for endogenous phytase activity. Phytic acid was detected early during embryogenesis in field-grown soybean seeds and accumulated in a linear fashion throughout seed development The initial level of phytate content observed in soybean seeds was 1.17 mg/g on fresh weight basis that reached a maximum of 27.16 mg/g in pods at full maturity thereby constituting 2.7% of the mature seed. Nearly all of the phosphorus translocated to developing seeds was incorporated into phytic acid from fourth week after flowering until physiological maturity, with the sum of non phytic acid phosphorus compounds remaining constant. Phytase activity peaked to maximum at 11^{th} day after germination corresponding well with high transcript levels observed around the same stage by semiquantitative RT-PCR and Northern analysis and thereafter leveled off until 21 days after germination.

A genomic sequence of *MIPS* gene from soybean (*Glycine max* var, Pusa-16) was amplified, cloned and characterized. The sequence revealed a total length of 2608 bps containing 9 introns interrupting 10 exons. The transcribed sequence of the gene had an expected ORF of 1533 bps predicted to encode 510 amino acids with a molecular weight 56472.72 daltons. Nucleotide composition revealed 39.95% G+C and 60.05% A+T content in the *MIPS* sequence. Alignment of amino acid sequence of *MIPS* with other plant species revealed seven conserved regions with identical amino acid sequences. A *MIPS* cDNA was amplified by RT-PCR, characterized and used as a probe for Southern. The Southern analysis suggested that *GmMIPS* was a part of multigene family of at least four similar members. Raffinose family oligosaccharides were estimated in three varieties of soybean where raffinose and stachyose content were highest in var. PK-416 compared to DS-9702 and Pusa-16.

Seed phosphorous and phytate accumulation patterns and *MIPS* expression analysis were conducted in the developing soybean seeds. Phytic acid was detected early during embryogenesis in field grown soybean and it followed a linear accumulation pattern during seed development. A final concentration of 1.9g/100gDW (1.9%) was reached at maturity, which constituted ~80% of total phosphorus in seeds. Inorganic phosphorus was almost constant (0.3g/100gDW) in stems, roots, flowers and developing seeds with a slight decline at seed maturity to a final concentration of 0.25 g/100g DW. *MIPS* cDNA of ~200bp was amplified by reverse transcriptase-PCR using a set *MIPS* specific primer pair, characterized and used as probe during expression studies. Expression analysis by RT-PCR and northern hybridization in developing seeds and vegetative tissues of soybean detected *MIPS* transcripts in early stages of seed development with their level reaching a peak at 6-8mm of seed size followed by a gradual decline. Complete absence of *MIPS* expression was observed in the mature (12-14mm) seeds and other vegetative tissues except leaves. Presence of MIPS expression in leaves and a phytate content of 0.45g/100g DW suggested the presence of a leaf specific MIPS isoform which may be involved in synthesis of phytate in the leaves.

In spite of highly nutritious composition and versatile uses of consumption of soybean is limiting worldwide due to the presence of grassy, beany and rancid flavour. This off flavour is generated due to the presence of inherent lipoxygenase (LOX) activity which acts on polyunsaturated fatty acids of soybean. The amount of antioxidants in soybean are known to control the LOX activity and hence the off flavour generation. It has been suggested that mild gamma irradiation enhances the antioxidants potential of soybean seeds. It is therefore important to find if there is any amelioration of off-flavour generation in soybean after gamma irradiation induced enhancement of its total antioxidant capacity. Presently study was, therefore, aimed to estimate the total antioxidant potential and degree of off-flavour generations, measured by TBA number and carbonyl value, of soybean seeds differing in seed coat colour and the effect of gamma radiation and subsequent storage on the above mentioned parameters. The study revealed that among the seventeen soybean genotype seeds with different seed coat colour, the antioxidant potential was very high in seeds with black and brown seed coat colour as compared to yellow and gree seed coat colour. But the difference in Thiobarbituric acid (TBA) number, carbonyl value and LOX activity of seeds with varying seed coat colour was not proportionate with the difference in their antioxidant capacity. Six varieties with extreme values of these parameter and contrasting colours were selected for gamma irradiation and in the gamma irradiated samples of soybean, the relative enhancement in antioxidant potential was maximum in yellow seed coat coloured genotype wheras it was minimum in black and brown seed coat colour at lower dose of 0.5 Kgy. At higher dose of 1.0 and 2.0 Kgy the total antioxidant and FRSA were decreased in all varieties. The TBA number, carbonyl value and LOX activity decreased at 0.5 Kgy whereas these values increased at 1.0 and 2.0 KGY. In the present study it was observed that total antioxidant capacity of different soybean cultivars effectively control the off-flavour generating compounds and the lower dose of gamma radiation leads to enhancement in the antioxidant potential of soybean seeds, the enhancement being maximum in yellow seed coat coloured genotypes followed by seeds with green, brown and black seed coat. The effect of gamma irradiation on the off-flavour generating potential of soybean seeds was retained only upto 15 days of storage and it subsided sharply after that.





Elucidation of the role of protein oxidation, hydrolysis, and protein-phenolic interactions in off-flavor generation in soybean.

Interest in introducing soybean and soy based food products to the diet has fetched much attention recently owing to the health promoting effects of soy protein and phytochemicals antioxidants present in it. However, the grassy, beany, metallic and bitter flavour associated with soy products restricts the growth in its consumption. Lower doses of gamma radiation are being used in food processing for microbial disinfestations and are also reported to enhance the antioxidant potential in soybean. The present study was done with the objective to explore the effect of three different doses (0.25, 0.5 and 1.0 kGy) of gamma radiations on the levels of individual antioxidant molecules and enzymes in soybean seeds differing w.r.t. their seed coat colour and to determine the effects of changes in antioxidant capacity on the off-flavour generation mediated through oxidation of PUFAs. The study revealed that among the fourteen genotypes of different seed coat colour the varieties with black and brown seed coat were richer in antioxidant molecules mainly because of inherent higher levels of anthocyanin in them. The anthocyanin content increased significantly in dark coloured varieties at all radiation doses but for yellow varieties there was significant decrease at the higher doses of 1.0 kGy. Gamma irradiation resulted in slight enhancement in the level of total phenols at 0.5 kGy but only in dark coloured varieties. The isoflavone content was found to increase at 0.25 kGy and then decreases at higher doses for all varieties dark as well as light coloured. The tocopherol content increased significantly at 0.25 kGy for dark coloured varieties and there after decreases sharply at higher doses while a continuous decrease at all radiation doses was observed for the yellow varieties. The LOX activity decreased significantly for dark varieties at all radiation doses however for yellow varieties activity decreased slightly and non-significantly at 1.0 kGy. TBA number and Carbonyl value decreased for dark varieties at 0.5 and 1.0 kGy but increased significantly for yellow varieties at all doses. Gamma radiation had no significant effect on the activities of antioxidant enzymes. The present study showed that the varieties having dark seed coat colour have higher levels of antioxidant molecules and are more responsive to gamma radiation induced enhancement in the antioxidant potential leading to flavor improvement, as compared to the varieties with yellow seed coat colour.

Soybean is a very nutritive food due to presence of high quality protein, oil and a variety of antioxidant molecules, and is considered as "functional food". But the consumption of soybean is limiting due to the presence of grassy/ beany/ rancid off flavour. The off flavour is generated due to the production of small aldehydes and ketones from polyunsaturated fatty acids by the action of lipoxygenase and hydroperoxidelyase. These aldehydes and ketones also act as the signalling molecule and also reduce storability of soybean seed. Present study was aimed to find the correlation between parameters related with off flavour generation and

storability/germination % of soybean seeds and purification of hydroperoxidelyase from soybean seed. The study revealed that among the 14 genotypes of soybean differing in storability, a significant negative correlation between LOX 2 activity, hydroperoxidelyase activity, TBA No., carbonyl value, total lipid hydroperoxide value with germination % was existed. A significant positive correlation between antioxidant molecules (total phenol and ascorbic acid), antioxidant potential (DPPH and CUPRAC) and lipase activity with germination % was also observed. No significant correlation was found between LOX 1, LOX 3, antioxidant enzymes (catalase, peroxidase and superoxide dismutase), isoflavone and tocopherol with germination %.

Hydroperoxidelyase from soybean seed was purified to 16.5 fold by ammonium sulphate precipitation followed by ion exchange chromatography. The SDS PAGE analysis of purified enzyme revealed it to have a molecular weight of about 53kDa. Kinetic behaviour of purified hydroperoxidelyase was studied and Lineweaver-Burk Plot was drawn from that Vmax and Km value were found to be 275.48×10^{-3} ΔA/min. and 25 µM respectively. The effect of natural antioxidant molecules like α-tocopherol, genistein, glycitein, daidzein and ascorbic acid on the HPL activity showed them all to inhibit the enzyme activity. The most potent inhibitor was found to be genistein then ascorbic acid, α-tocopherol, glycitein, daidzein in the decreasing order.

Indian soybean industries are flourishing at a rapid pace to process soybean for food, feed, pharmaceuticals and industrial applications. However the major bottleneck associated with both traditional soya based food and oil industries is development of grassy-beany or fishy flavour nicknamed as 'soya off flavour', a major deterrent for consumer acceptance of soya based food products. The mature soybean seed contains predominantly three lipoxygenase (lox) isozymes with marked differences in their optimum pH, substrate preference and product profile which culminates into this objectionable flavour composition. The major aim of the present study was to isolate and characterize the Lox isozymes with reference to various biochemical parameters. The Lox isozymes are purified to 107 fold by DEAE sephacel chromatography and then separated into three individual isozymes by denaturing isoelectric focusing gel and confirmed by MALDI-TOF analysis. Lox-2 and Lox-3 displayed remarkable kinetic preference for product generation (HPOD) in low PUFA ratios among the three selected ratio combination with naturally mimicked one. Lox-1 displayed no specific preference. The pure Lox-1 displayed unbiased response towards linoleic and linolenic acids with marginal preference for linoleic acid in its optimum pH at 9.0. The volatile compound profiling in the three selected ratios via GC-MS analysis endorses the direct relationship hypothesis between PR (PUFA Ratios) with hexanal to t-2-hexenal ratio. This kinetic data was functionally validated by determining the off flavour generating potential estimated by TBA values, Carbonyl no. and lipid hydroperoxides level (LHPODs) determined in selected soya cultivars as well as in putative mutants displaying varied PUFA ratios. A significant negative correlation was observed between PUFA ratios with these off flavour estimating parameters. Most significantly the ratio of PI (polyene index) to PR (PUFA Ratios) displayed remarkably high negative correlation with these parameters implying the significance of PUFA ratios in biological lipid peroxidation. Lipoxygenase isozymes (Lox-1, Lox-2, and Lox-3) isolated from defatted beans exhibited inhibition potential by modulating the enzyme to substrate ratio. Lox-2 is the most inhibition prone enzyme. 34 putative soya mutant lines were also characterized for their comparative total protein and oil content, fatty acid composition, lipoxygenase pathway enzyme assay, and total antioxidant potential as well as enzyme assay. The lipoxygenase isozyme 2 (Lox-2) is the predominant enzyme responsible for off flavour generation as it exhibited significant negative correlation with total antioxidant potential and significant positive correlation with off flavour generating potential.

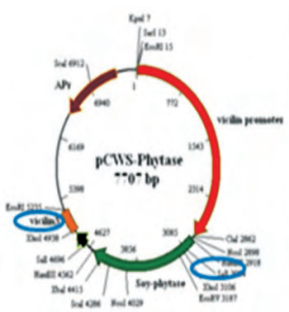
A 2.6 kb full length *lox2* gene (NCBI accession number: JQ929619.1**)** through RT-PCR from developing soybean seeds. The cloned cDNA sequence of *lox 2* gene has the complete Open Reading Frame (ORF) of putative protein having 866 amino acids. Prokaryotic expression was carried out of *lox2* gene by cloning in pET expression system and induction by 1 mM IPTG and 97 kDa recombinant LOX 2 protein was purified. The purified recombinant protein activity was inhibited by soya isoflavones (genistein and diadzein) and antioxidant free radical scavenging vitamins (ascorbate and alpha tocopherol) to exploit their effectiveness. All the natural antioxidants inhibited LOX-2 in non-competitive concentration dependent manner. Alpha tocopherol (Ki=5 µM)

was the most potent inhibitor among all. Among the two isoflavonesgenistein (62 µM) was more potent inhibitor than diadzein (98 µM). High isoflavone and low radical scavenger combination exhibited synergistic mode of action implying the functional interdependence of the isoflavones on primary antioxidants for maintaining steady state. The inhibition kinetics results were further validated by estimating off flavour generating parameters like malonaldehyde, carbonyl compounds and hexanal*in vitro* from seeds using the same inhibitor combination. The present study has helped us to get improved understanding of off flavour generation and its management in soybean. The major findings (ratio of polyene index (PI) to PUFA ratio (PR) can act as biomarker to off flavour generation) will also help significantly to plant breeders for selecting seeds with lower oxidation potential.

A major goal for grain crop improvement is the reduction of its phytic acid (*myo*-inositol hexakisphosphate) content in the seeds to improve micronutrient bioavailability and phosphorus utilization by humans and nonruminant animals as well as reduce environmental pollution. The enzyme *myo*-inositol-1-phosphate synthase (MIPS) catalyses the first step in *myo*-inositol biosynthesis and directs the phytic acid biosynthesis in seeds. The relationship between phytic acid biosynthesis and MIPS make this enzyme an attractive target for reduction of its expression. Therefore, to effectively suppress phytic acid biosynthesis, an interfering RNA (RNAi) and an antisense (AS) construct with *MIPS1* under the control of seed specific vicilin promoter were generated. Soybean transformation was carried out using *Agrobacterium* mediated cotyledonary node method and *bar* gene as a selection marker coupled with glufosinate as a plant selection agent. Putative primary transformants and their progenies were screened by PCR with *bar* and *MIPS* specific primers. The insertion of T-DNA in transformants was confirmed by Southern analysis. RT-PCR and qRT-PCR expression analysis revealed reduced *MIPS1* expression levels in putative transformants. The resulting transgenic seeds (T_1) with AS cassette germinated normally and showed a reduction of ~ 50% in the phytate levels and increase of about 20% in the inorganic phosphate (Pi) levels.

The off-flavour present in soybean limits its consumption and acceptability among people. The immense nutritional benefits of soybean remain underutilized due to this beany flavor. With a view to understand the mechanism of off-flavour generation in soybean and developing methods for its reduction, the inherent levels of off-flavour determining parameters (LOX activity, TBA number and amount of oxidized proteins, trypsin inhibitor activity) andantioxidant molecules (total bound and unbound phenols and tocopherols) were analyzed in fourteen genotypes of soybean seeds that varied with respect to their seed coat colour. The study revealed that out of these fourteen varieties, two varieties namely SL 525(Yellow seed coat colour), EC 109514(black seed coat colour) had the least and the highest off flavor generating capacity, respectively. These cultivars were selected for treatment with 3 biotic elicitors viz. Chitosan (2, 4, and 8 mg/ml, Jasmonic acid(1, 3, and 5 µM) , Salicylic acid (0.1,0.5, and 1.0 mM) for a duration of 8 and 16 days. The treatment of soybean seeds with biological elicitors could alter its inherent antioxidant system which is comprised of non -enzymatic antioxidant molecules (vitamins A, C, E; glutathione, carotenoids, phenolics) and antioxidant enzymes (SOD, Peroxidase and Catalase).

A significant increase in bound and unbound phenols, trypsin inhibitor activity and total tocopherol content and a decrease in Lipoxygenase activity, Thiobarbituric acid (TBA) number and amount of oxidized proteins was observed in seeds treated by all the three elicitors. Both the SL 525 and EC 109514 responded favourably



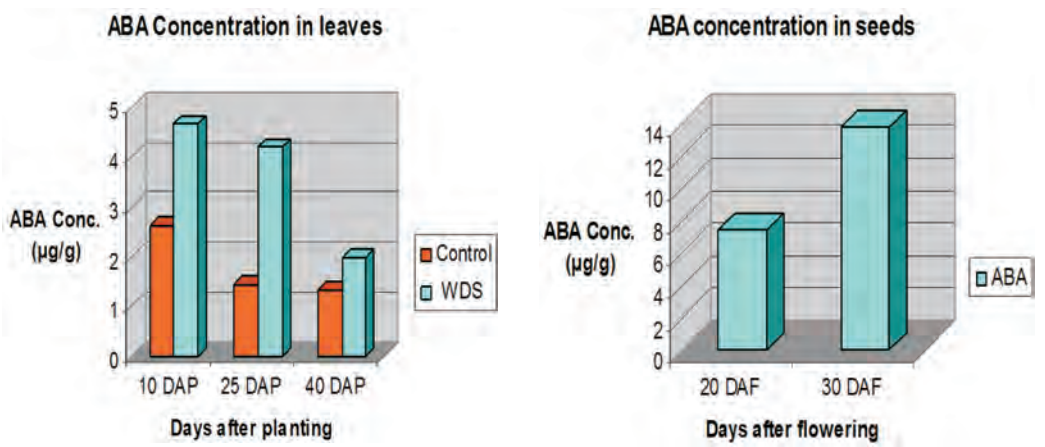



Cultivars	Phytic acid (g/100g)	Percentage reduction of Phytic acid (%)
Non transgenic control	3.875 ± 0.13	
Phytase T4		
PHY 53-33-12-12-13-1	2.19 ± 0.11	43.41
PHY 48-23-29-12-3-2	2.56 ± 0.81	33.85
PHY 23-13-12-2-18-5	2.35 ± 0.06	39.27
PHY 31-23-12-15-13-7	2.72 ± 0.53	29.71
PHY 53-17-22-12-3-8	2.22 ± 0.59	42.63

Reduction in the phytic acid content in the selected transgenic lines (T_4)

to the elicitors with respect to different off flavor controlling parameters. The off-flavour reducing effect of these elicitors is mediated through enhancement in the antioxidant potential of soybean seeds. Furthermore, the ameliorating effects of biotic elicitors seems to be dose and duration dependent as enhancements in the antioxidant potential is more at higher concentrations and at 16 days treatment as compared to 8 days of treatment. EC 109514 responded much better to the elicitors than SL 525 as far as increase in antioxidant potential was concerned and there was a much greater decrease in LOX (Lipoxygenase) activity and TBA (Thiobarbituric acid) number and oxidized proteins in SL525 than in EC 109514.

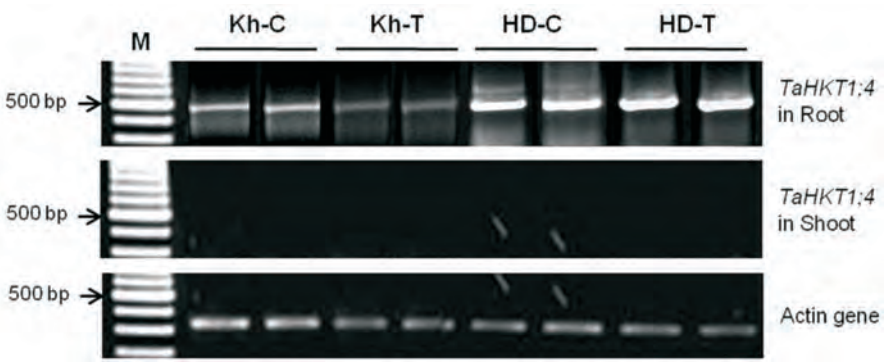
Phytic acid, the principal storage form of phosphorus in soybean seeds is poorly digested by monogastrics including humans. As a polyanion at physiological pH, it has the ability to chelate positively charged cations and also bind to proteins and starch thereby reducing their bioavailability and digestibility as an anti-nutritient. Reduction of seed phytate content being a promising approach for increasing the mineral and protein bioavailability in soybean, characterization of the gene encoding D-*myo*inositol-3-phosphate synthase (MIPS), catalyzing the first and the rate-limiting step in the phytic acid biosynthesis, was taken up as the target gene in the present study. The differential expression profile of four members of the *MIPS* gene family was monitored by qPCR, in the developing seeds and vegetative tissues of soybean. The results indicated both spatial and temporal variations in the expression pattern of *MIPS* gene isoforms. Amongst the four, *MIPS1* exhibited the highest



transcript levels in the soy cotyledons during early developmental stages. A full-length *GmMIPS1* cDNA of 1,791 bp, with an ORF of 1,533 bp, encoding 510 amino acids (HM46196) and showing a striking homology (87-96%) with MIPS of other plant species was cloned and characterized. The predicted protein sequence indicated the absence of a signal peptide. To validate its expression, the *MIPS* (131 to 1556bp) amplified by high fidelity PCR was fused in frame to a 19 aa N-terminal region of 6X His-tag in a prokaryotic expression vector pET28a(+) and transformed into *E. coli.* Optimized induction and expression conditions resulted in the production of a 52 kDa fusion protein confirmed by SDS-PAGE and Western blot analysis. The study has laid a base for the development of low-phytate transgenic soybean by targeting *GmMIPS1* gene using the RNAi approach, which will ultimately result in improved nutrient availability for animals and humans and also in reduced environmental impact of livestock production.

D-*myo*inositol-3-phosphate synthase (EC 5.5.1.4; MIPS) catalyzes the conversion of Dglucose-6-phosphate to D-*myo*-inositol-3-phosphate, the first and rate-limiting step in the phytic acid biosynthesis. The number of *MIPS* genes and their expression differ in the plant genome. In the present study, we monitored the differential expression profile of four, previously reported, members of the *MIPS* gene family in the developing seeds and a vegetative tissue of soybean by quantitative real-time PCR (qRT-PCR) was conducted. The results indicated both spatial and temporal variations in the expression pattern of *MIPS* gene isoforms. Amongst the four isoforms, *MIPS1* exhibited the highest transcript levels in the soy cotyledons in the early developmental stages with the expression showing a peak at around 4 to 6 mm of seed size stage. As the development progressed, the expression decreased to nearly undetectable levels in the seeds of 10 to 12 mm stage size. In comparison, the *MIPS1* transcripts were found in almost negligible concentrations in other organ tissues i.e. seedlings, roots, stems, leaves and flowers. A full-length *GmMIPS1* cDNA (*Glycine max*) of 1,791 bp, containing an open reading frame (ORF) of 1,533 bp, encoding 510 amino acids was cloned and submitted to the NCBI GenBank database under the accession number HM46196. It showed striking homology of 87-96% with MIPS of other plant species. The protein sequence analysis of the predicted *GmMIPS*cDNA indicated the absence of a signal peptide in the N-terminal region. To validate its expression, a fragment containing majority of *MIPS* coding residues (131 to 1556bp) were amplified by high fidelity PCR and was fused in frame to a 19 amino acid N-terminal region of 6X His-tag in a prokaryotic expression vector pET28a(+). The recombinant plasmid was transformed into *Escherichia coli* strain BL21 (DE3). Optimized induction and expression conditions resulted in the production of a 52 kDa fusion protein as inclusion bodies, confirmed by SDS-PAGE and Western blot analysis. The study has laid a foundation for the development of low-phytate transgenic soybean by targeting *GmMIPS1* gene using the dsRNA induced sequence specific RNA degradation mechanism, without affecting the critical aspects of inositol metabolism in other tissues of the plant, which will ultimately result in improved nutrient availability for animals and humans and also in reduced environmental impact of livestock production.

The off-flavor in soybean, generated due to the production of small aldehydes and ketones from polyunsaturated fatty acids by the action of lipoxygenase and hydroperoxidelyase, limits its consumption and acceptability amongst people. The immense nutritional benefits of soybean thus remain underutilized due to this beany flavor. These aldehydes and ketones also reduce the storability of soybean seeds. The inherent levels of antioxidant molecules



Semi-quantitative (RT-PCR) expression analysis of *TaHKT1;4* gene in shoot and root of the contrasting wheat genotypes Kharchia-65 (Kh) and HD-2329 (HD) under control (C) and 200 mMNaCl stress (T) conditions. Actin was used as reference gene. M= Marker, 100bp DNA ladder.

(total bound/unbound phenols, tocopherols) and off flavour determining parameters (lipoxygenase (LOX) activity, Thiobarbituric acid (TBA) number, amount of oxidized proteins, trypsin inhibitor activity) were analyzed in fourteen genotypes of soybean seeds that varied with respect to their seed coat colour. The study revealed two varieties, SL 525 (Yellow seed coat colour), EC 109514 (black seed coat colour) with the least and the highest off flavor generating capacity respectively. The seeds of the selected varieties were treated with 3 different concentrations of 3 biotic elicitors viz. Chitosan (2, 4, and 8 mg/ml, Jasmonic acid (1, 3, and 5 µM) , Salicylic acid (0.1, 0.5, and 1.0 mM) for a duration of 8 and 16 days, as treatment with biological elicitors can alter the inherent antioxidant system which comprises of non enzymatic antioxidant molecules (vitamins A, C, E; glutathione, carotenoids, phenolics) and antioxidant enzymes (SOD, peroxidase and catalase). A significant increase in bound and unbound phenols, trypsin inhibitor activity and total tocopherol content and a decrease in lipoxygenase activity, thiobarbituric acid (TBA) number and amount of oxidized proteins were observed in seeds treated by all the three elicitors. Both the SL 525 and EC 109514 responded favourably to the elicitors with respect to different off flavor controlling parameters at 16 days after treatment compared to 8 days. A much better response was observed at higher concentrations of elicitors at 16 days of treatment than at 8 days. EC 109514 responded better to elicitors than SL 525 as far as increase in antioxidant potential was concerned and a greater decrease in LOX activity, TBA number and oxidized proteins was observed in SL525 than in EC 109514.

Two approaches were carried out to solve the nutritional and environmental problems represented by seed phytic acid ie through screening for low phytate soybean mutants and engineering soybean for ectopically expressing high levels of phytase enzyme in developing seeds. Thirty four mutant populations developed, following the treatment of 17 soybean genotypes with chemical and physical mutagens and also selected for their resistance to yellow mosaic virus (M_2 through M_{10}) were procured from genetics division and analysed for theirphytic acid (PA), protein and mineral levels in the selected M_{10} plants. Five mutant lines showed reduced phytic acid and phytic acid phosphorous contents compared to their parental lines. PA and seed protein were found to be positively and significantly correlated among the mutant populations and negatively and significantly correlated in the putative low phytate mutants. A negative but no significant correlation between PA and divalent cations among the mutant populations and a significant but negative correlation between PA and iron in the low phytate mutant lines identified was observed. For the transgenic approach, to develop a rapid and reliable transformation protocol, *Agrobacterium* mediated stable transformation of Indian soybean genotypes JS 335 and DS 9712, using cotyledonary node and hypocotyl segment methods was attempted and the more efficient cotyledonary node transformation system best standardized for JS335 was further used for developing soybean transgenics. *Phytase* over expressing cassette, with the soybean seedling *phytase* cDNA (*Gmphy)* under the control of a seed specific promoter was introduced into soybean and the putative transgenics selected on glufosinate were confirmed by molecular analysis. Seven independent transgenic soybean lines with single copy integrations were also subjected to *phytase* expression analysis. The phytase activity in the T_2 seeds of the transgenics lines showed upto 2.9 fold increases over the control and the subsequent T_3 seeds revealed a reduction in the phytate levels ranging from 25.7 to 71.7%. The results of the present study reflected the effectiveness of these approaches as potential strategies for reducing phytate content in soybean seeds and can ultimately result in a desired germplasm source for further purposes.

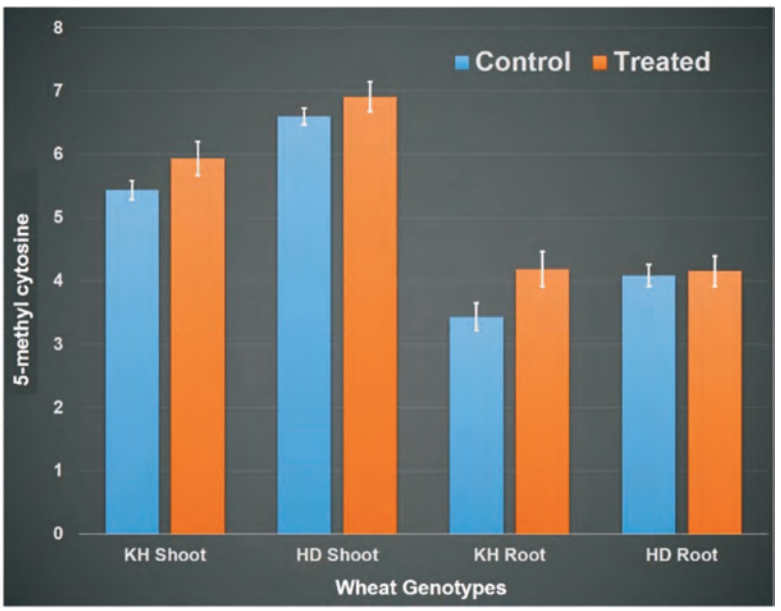
Biochemistry of Stress Responses

ABA was isolated and quantified at different developmental stages of seed and leaves of rice cv N22 in control and water deficit stress conditions. ABA content increases when a plant is exposed to stress. A drought inducible gene encoding WRKY transcription factor was amplified, cloned and sequenced. Sequence analysis predicted that the gene contains an ORF of 186 bp with 5' and 3' UTR of 49 and 43 nucleotides respectively and encodes a protein of 61 aa residues. RTPCR analysis revealed an up regulated expression of gene in ABA treated plants as compared to control. Hence, it can be predicted that the particular WRKY gene shows ABA

induced expression.

A 732 bp cDNA belonging to SHN clade of AP2/ERF transcription factor family associated with water deficit stress was isolated from leaves of *Oryzasativa* L.cv N22. The cDNA showed induced expression during water deficit stress. Epicuticular wax content was compared at varied relative water content in leaves of *Oryzasativa*cv N22. The wax content was found to be negatively correlated with RWC and showed positive correlation with induced expression of gene and WDS.

One of the major problems faced by rice cultivation is the drought stress, as it requires high amount of irrigation water. Drought tolerant cultivars and their phytochemical composition which has a role in providing drought tolerance are gaining importance in this context. In this study, the level of gamma oryzanol which is an antioxidant present in considerable amount in the bran fraction is compared among five different cultivars viz- P1401, Pusa Basmati-1(PB1), N22, APO and PNR381. P1401 and PB1 are known to be drought sensitive cultivars while N22 and APO are drought tolerant cultivars. PNR381 is suitable for both irrigated and upland cultivation. Reverse phase HPLC was done with the crude rice bran oil (RBO) and semi-purified oryzanol obtained from the five cultivars using C18 column as stationary phase and methanol/acetonitrile/dichloromethane/acetic acid (50:44:3:3) as the mobile phase at a flow rate of 2 ml/min. Four peaks were obtained in each sample and their levels were compared. The drought tolerant cultivars showed higher levels of oryzanol compared to drought sensitive cultivars, indicating its role as an antioxidant involved in combating oxidative stress associated with drought. Among the various types of stresses experienced by plants, drought stress is the most important one faced by rice, as it is a crop suitable for semi-aquatic condition. The various antioxidant systems associated with the oxidative stress are gaining importance in this context. In this study, the functional role of gamma oryzanol as an antioxidant is being discussed. The variation in the levels of individual components of oryzanol present in five cultivars of rice viz- P1401, Pusa Basmati-1 (PB1), N22, APO and PNR381 are compared and the effect of its variation on the total antioxidant capacity has been studied. The semi-purified oryzanol obtained from the five cultivars were subjected to CUPRAC (Cupric ion reducing antioxidant capacity) assay. The drought tolerant cultivars N22 and APO showed higher antioxidant capacity compared to the drought sensitive cultivars P1401 and PB1 indicating the role of gamma oryzanol as an antioxidant in combating the oxidative stress along with other antioxidant systems. The four peaks were identified as cycloartanylferulate, 24-Methylene cycloartanylferulate, Campesterylferulate and Sitosterylferulate by comparing their respective retention times with that obtained by Xu and Godber. 24-methylene cycloartanylferulate was the principal component and is thought to be the major source of antioxidant activity.



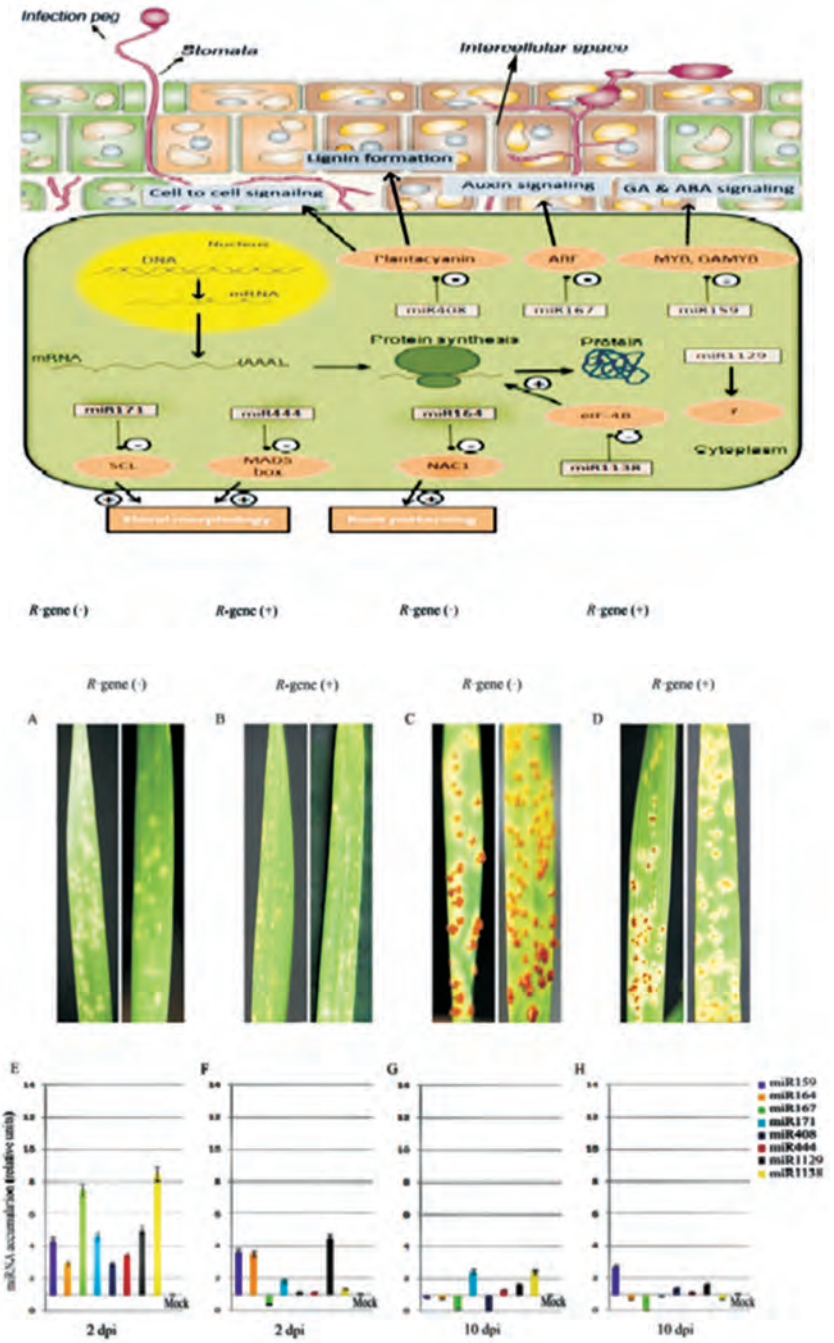
Changes in global-methylation status in shoot and root of contrasting wheat genotypes (HD=HD2329, salt-sensitive and KH=Kharchia-65, salt-tolerant) under control and salt stress (200 mM NaCl, treated) conditions.

An attempt was made to study the activity of α-amylase from different stages of developing seed of bread wheat grown under rainfed and semi-irrigated conditionsto elucidate a relationship between enzyme activity and starch accumulation in the grain. Developmental profiles of α-amylasein developing bread wheat shows

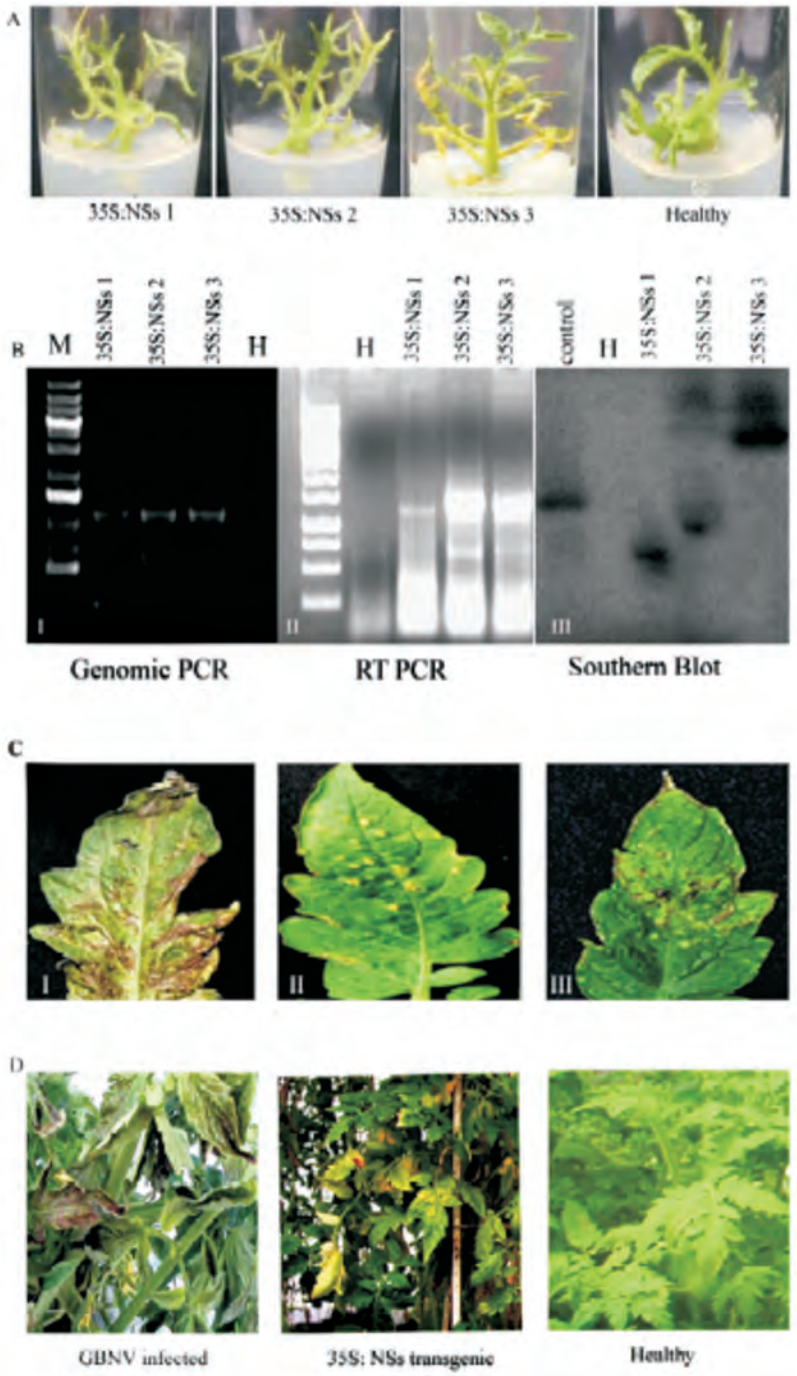
near about twice pre-matured α-amylase activity in varieties grown under rain-fed conditions as compared with that of semi-irrigated conditions. α-Amylase activity in dough stage was lower under both the above indicated conditions (semi-irrigated, rainfed) for all the varieties tested, indicating that the dough stage is the important stage for starch deposition leading to grain filling in wheat grains. A correlation study at maturity stage of α-amylase with plot yield and grain density revealed a very significant negative correlation for the varieties grown in semi-irrigated conditions, which indicated a negative impact of high enzyme activity at mature stage on grain yield. Activity staining and enzymatic assay results of imbibed seeds were almost similar. At least four prominent bands of isozymes were identified after six hour of imbibition in the variety Kundan for both the environmental growing conditions.

Wheat, the second most important crop of India, faces salinization problem which decreases its average yield by more than 50%. The impact of salt stress on root and shoot regions of three contrasting varieties of wheat *viz.*, WH542 (salt susceptible), KRL 1-4 (moderate salt tolerant), KRL 210 (salt tolerant) under differential levels of salt stress (0, 150 and 300 mMNaCl) at different developmental stages (7,14 and 21 days after treatment-DAT) were studied in terms of their membrane stability index (MSI), protein expression profiles, activities of cell-wall degrading enzymes (CWDE) and expression analysis of high affinity potassium transporters (HKT). There was a progressive decline in MSI due to increase in electrolyte leakage at differential levels and stages of salt stress induced cell membrane injury. Salt susceptible wheat genotype (WH 542) was most affected, while salt tolerant wheat genotype (KRL 210) showed maximum resistance to the salt stress. Differential expressions of soluble proteins in these contrasting wheat genotypes revealed their spatial and temporal partitioning during salt stress. The CWDEs were found crucial biochemical markers for resistance against salt stress. Activities of these enzymes were found to be more pronounced in shoot region than root and were significantly high in salt susceptible genotype. A transporter from HKT_1 subfamily-HKT1; 4 (HKT7) was found to be root specific, while HKT2; 1 (HKT1) and HKT2; 3(HKT3) of HKT_2 subfamily were expressed in both root and shoot regions. These findings may help towards deciphering the mechanism of salt-tolerance and evolving wheat varieties with higher salinity tolerance levels.

Plants being sessile in nature have evolved several mechanisms to cope with a variety of environmental stresses. Salt stress is one of the most detrimental factors which limit crop productivity. Wheat faces salinization problem which reduces crop productivity by 50 percent. Kharchia-65 is one of the salt-tolerant wheat genotypes that have been used to understand the mechanisms of salt tolerance. The role of high-affinity potassium transporters (HKTs) in Na^+ compartmentalization in vacuoles has been well accepted to be an important activity in maintaining high K^+/Na^+ ratio in cytosol. Comparative analysis of two important salt-tolerant genotypes (Kharchia-65 and KRL-210) for their tolerance to salt stress on the basis of biochemical and physiological parameters revealed that Kharchia-65 is a more tolerant genotype. Comparative analysis of two elite wheat genotypes (HD-2329 and WH-542) indicated that HD-2329 is more sensitive to salt stress. This pair of contrasting wheat genotypes (Kharchia-65 and HD-2329) was used to understand epigenetic regulation of gene expression under salt stress. DNA methylation due to environmental stresses has been reported to be important regulator of gene expression in higher eukaryotes. Although effect of salt stress on cytosine methylation has been reported in crop plant, its functional consequences are still underexplored. The present study on epigenetic variation in contrasting wheat genotypes due to salt stress imposition revealed that the global methylation level in shoot of both the genotypes was higher (5.4 – 6.6%) compared to that in root (3.4 – 4.0%), and salt stress caused increase in methylation by more than 10% in salt-tolerant genotype. Locus-specific quantitative analysis of cytosine methylation revealed that *HKT2;3* gene in salt-tolerant genotype was hypermethylated with the presence of 93% 5-methylcytosine (5-mC) in both shoot and root tissues compared to only 87% 5-mC in salt-sensitive genotype. Most of the cytosines in CG context were methylated in both the genotypes, however a significant variation in cytosine methylation due to salt stress was observed in CHH context. Expression analysis of *HKT2;3* gene indicated that hyper-methylation of the gene in shoot of salt-tolerant genotype caused 8-fold down-regulation of the gene. With 19% reduction in 5-mC in shoot of salt-sensitive genotype, the gene showed 10-fold up-regulated expression. The hyper-methylated gene was 2.5-fold down-regulated in root of the salt-tolerant genotype. With 15% increase



A. Schematic presentation of spore germination and mycelia growth affecting plant cell by depicting probable targets and cellular functions of eight micro RNAs. B. Phenotyping and miRNA profiling of wheat-stem rust interaction. (A) Emergence of symptoms in C-306 cultivar (R-gene (-)) at 2 dpi (B) Emergence of symptoms in HW-2004 cultivar (R-gene (+)) at 2 dpi (C) Maturation of pustules in C-306 at 10 dpi (D) Maturation of pustules in HW-2004 at 10 dpi (E) Quantitative real-time PCR analysis of 8 selected miRNAs in C-306 cultivar at 2 dpi (F) Quantitative real-time PCR analysis of 8 selected miRNAs in HW-2004 at 2 dpi (G) Quantitative real-time PCR analysis of 8 selected miRNAs in C-306 at 10 dpi (H) Quantitative real-time PCR analysis of 8 selected miRNAs in HW-2004 at 10 dpi. Expression level of each miRNA in mock was set as 1. The relative expression level of all miRNA was calculated using the comparative 2-ddCT method. The results were obtained from two independent experiments and all the data were normalized to U6 snRNA expression level, and considerable error bars might results from the changed culture condition, different host plants, and RNA isolation steps.



Comparision of phenotypic symptoms in GBNV infected and 35S:NSs transgenic tomato plants.

(A) 35S:NSs transgenic lines (35S:NSs1,2 and 3) at shooting stage. Plants show abnormal morphological characteristics such as spiny initial leaves and stunted growth.

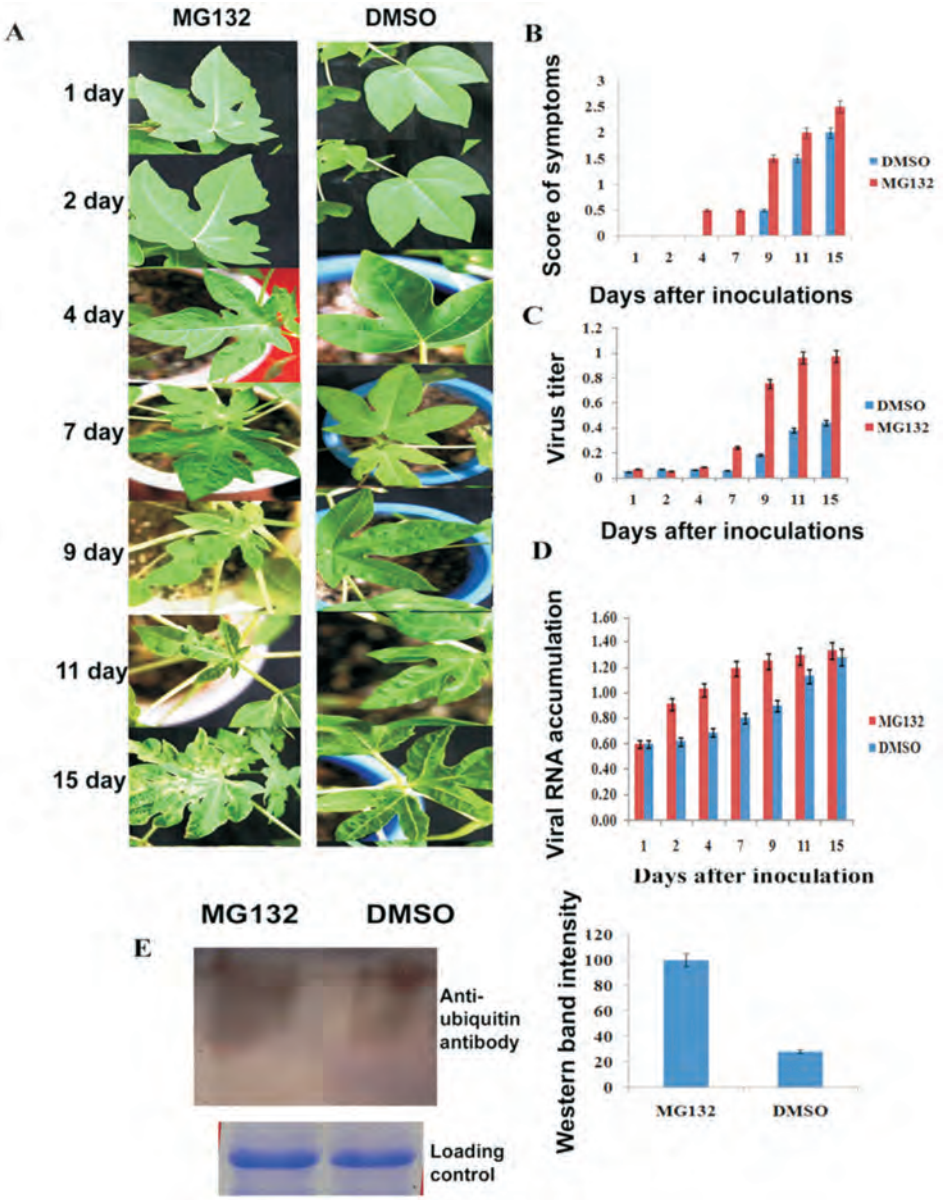
(B) Genomic PCR (I), RT-PCR (II) and Southern blot hybridization (III) of 35S:NSs transgenic lines., M:1 kb DNA ladder, H : healthy, Control: NSs amplicon (C) Close view of the symptoms in GBNV infected and NSs transgenic tomato leaves Necrotic(I) , chlorotic (II) and chlorotic turning to necrotic (III) symptoms appearing in GBNV infected and 35S:NSs transgenic respectively. (D) GBNV infected and 35S:NSs transgenic tomato plants showing necrotic and chlorotic symptoms respectively.

in 5-mC in root tissues of salt-sensitive genotype, the gene showed 1.4-fold down-regulated expression. The findings provide functional correlation between stress-induced DNA methylation and gene expression under salt stress which may prove to be helpful in mitigating the harmful effects of salt stress in crop plants.

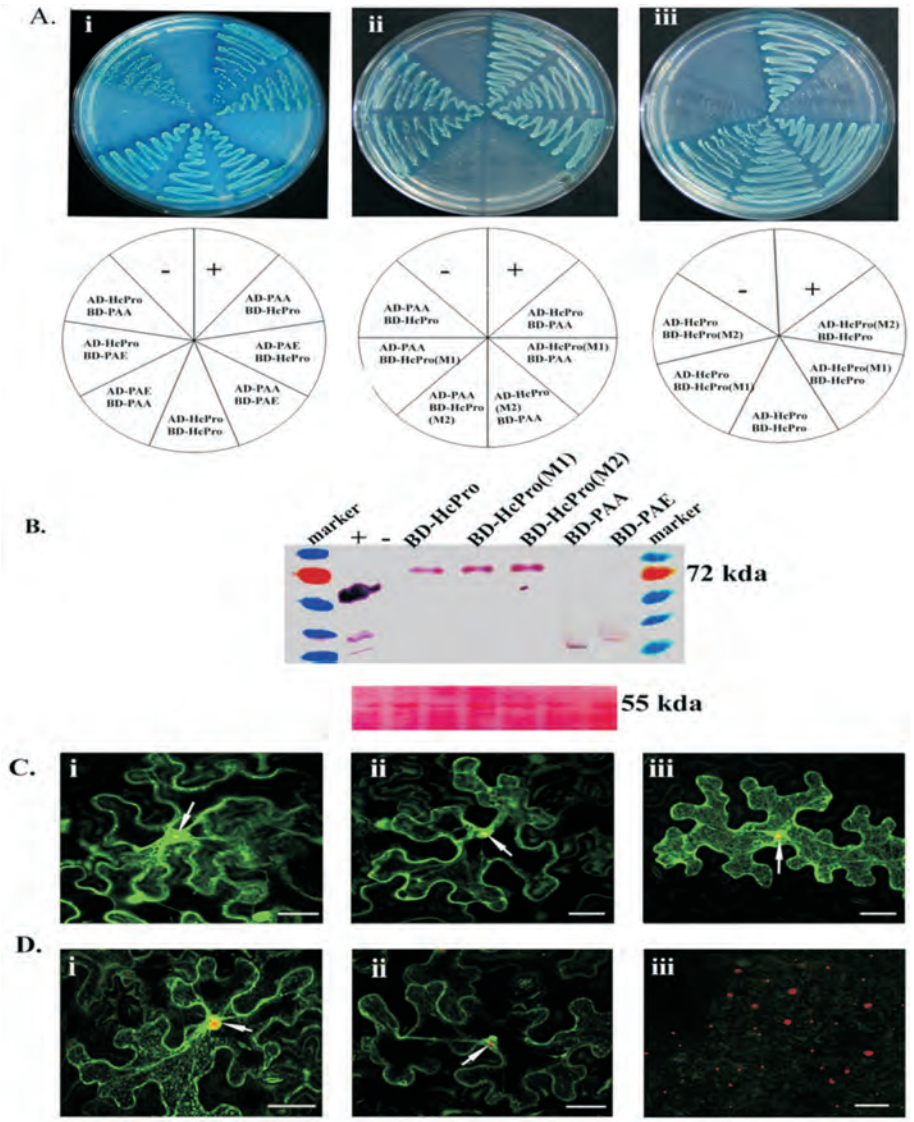
Terminal heat stress (HS) is one of the major problems in wheat growth and yield; it drastically reduces the quantity and quality of the grains. Enzymes associated with starch biosynthesis path way are severely affected by heat stress especially soluble starch synthases (SSSs) which is highly heat labile. Different isoforms of SSS has been reported from rice, maize etc.; limited information is available in wheat. Here, we studied the expression profiling and activity assay of novel SSS (accession no. KM206143)at different sub-stages of grain-filling *i.e.*milky-ripe, mealy-ripe and kernel hardening under control (22°C) and heat stress (42°C, 2 h) conditions in six different genotypes of wheat (HD2985, HD2329, PBW343, PBW 621, Raj3765 and Halna). We observed significant down regulation of novel SSS in response to heat stress during different sub-stages of grain-filling in all the six cultivars; Halnaharbors thermostable SSS as evident from the expression. Similarly, SSS showed decrease in the activity at different sub-stages of grain-filling; percent decrease was observed more in PBW621 and PBW343, as compared to Halna and HD2985. Starch accumulation pattern showed significant increase in the starch content with number of days of growth under normal and HS-treated conditions; maximum was observed in case of thermotolerant cultivars. Scatter plot analysis showed positive correlation between the relative fold expression of novel SSS and starch accumulation under control and HS-treated conditions. To conclude, mealy-ripe stage of grain-filling was observed critical for the starch synthase activity and starch accumulation in wheat. The information generated in present investigation can be used in breeding program for screening wheat lines for thermotolerance and for the development of climate-smart wheat crop.

miRNA are small nucleotide sequence involve in regulating the expression of target genes responsible for different phenotypic characters. Wheat, being the staple crop, is highly sensitive to heat stress and slight variations during critical stages causes-reduction in yield and quality of the grains. We identified 7 novel miRNAs from wheat using de novo assembly and further miR430 was closed from HD2985 cultivar of wheat. The identified miR430 was observed to be localized on Chromosome 4A and predicted to have more than 30 target genes. Most of the candidate miR430 targets identified were associated with the heat shock proteins (HSPs). Validation of miR430 in contrasting wheat cultivars by quantitative real time PCR showed significant variations in the expression under the heat stress. Tissue-specific expression profiling showed relatively low expression in leaves, compared with stem and root. A negative correlation was established between the expression of miR430 and their respective targets under HS in contrasting cultivars. MiR430 can e used as suitable candidate gene for manipulating the mechanism of thermo tolerance in wheat for the development of 'climate smart' crop.

Terminal heat stress causes disintegration of photo system, denaturation of enzymes, defunct pollen, and pseudo seed setting problem in agriculturally important crops. Wheat, being important food grain crop, has been neglected in terms of whole genome sequencing; very limited information is available on stress-associated genes and proteins. The mechanism of thermo tolerance has yet not been elucidated. Transcription factors are protein, which perform their role at transcriptional level by affecting the transcription process of SAGs. Here, we have identified 38 novel transcription factor genes from wheat *cv*. HD2985 using *de novo* assembly. Based on the digital fold expression, we cloned *TaHD97* of ~1.1 kb from wheat *cv.*HD2985. The nucleotide sequence was submitted in NCBI GenBank with accession no.KP259293. CD search revealed the presence of HSF DNA binding domain in the sequence. Expression analysis of cloned TF showed significant variations in the expression in response to heat stress; relative expression was observed very high in HD2985, as compared to HD2329 *cvs*. of wheat. Tissue specific expression profiling showed up regulation of *TaHD97*in leaves, as compared to stem, and endosperm, whereas down regulation was observed in root. Expression analysis of targets of *TaHD97* showed very high relative expression of *HSP17* and *HSP90*under HS; abundance of transcripts was observed more in HD2985, as compared to HD2329. A positive correlation was established between the*TaHD97* and their respective targets under HS. The cloned novel TF can be used as a suitable candidate gene for the breeding



Proteasomal inhibitor MG132 effect on PRSV infection. A. Symptoms produced by PRSV after mechanical inoculation in both MG132 and DMSO treated papaya plants with time. Papaya seedlings were sprayed with MG132 (50µM in 0.02% DMSO) and DMSO (0.02%) by foliar application. The first appearance of flecking symptom on papaya leaves was recorded. B. Symptoms severity kinetics in PRSV-infected papaya leaves treated with DMSO or MG132 in DMSO. Symptom severity was scored by early appearance of flecking followed by mosaic development on leaves, which further resulted in prominent mosaic and chlorosis. Symptom severity score was rated on a 3-point scale: 0 = no symptoms, 0.5 = appearance of mid mosaic, 1 = mild mosaic and chlorosis without leaf deformation, 1.5 = chlorosis with appearance of leaf deformation, 2 = clear mosaic with slight leaf deformation, 2.5 = clear mosaic and chlorosis with slight leaf deformation, 3 = strong mosaic all over the leaflets with leaf deformation, C. Virus accumulation kinetics in PRSV-infected papaya leaves treated with DMSO or MG132 in DMSO. Virus titre was estimated at different time intervals post viral inoculation by DAC-ELISA using PRSV polyclonal antibodies (dilution 1:1000). D. Relative qPCR quantification of PRSV RNA with amplification of 3' end of the genome overlapping CP encoding region in infected plants. Quantitative estimation of viral transcript using SYBR Green in quantitative PCR; amplifying 3' region of viral genome (9389 to 9566 ntd). Error bar represents standard deviation of three repeat assays. E. Western blot of proteins extracted from Nicotianatabacum leaves treated with DMSO or MG132 in DMSO. Total protein was isolated from tobacco leaves after 24 hrs post treatment and western blotting was done by using a rabbit polyclonal anti-ubiquitin antiserum (Sigma; 1: 10,000 dilution). MG132 treatment resulted in acculmulation of proteins; as shown by western blot and by a graph showing relative band intensities.



PRSV HcPro interactions and localization. A. For yeast two-hybrid assays the constructs were transformed in Saccharomyces cerevisiae AH109 cells. The cells were grown on SD/-Ade/-His/-Leu/-Trp + X-α-gal media. pGBKT7-p53/pGADT7-RecT (positive control) symbolized as+, pGBKT7-p53-/pGADT7–lamin (negative control) symbolized as - . Schematic representation of interactions is shown below the plate format where pGADKT7 is denoted as AD and pGBKT7 is denoted as BD. (i). Interaction of PRSV HcPro with the papaya 20S proteasome subunits PAA and PAE, and with itself. HcPro can interact with PAA but not with the PAE subunit of the proteasomal complex and HcProexhibit a strong self-interaction. (ii). Interaction of PRSV HcPro mutants with papaya proteasome subunit PAA. HcPro (M1) can interact with PAA, but HcPro (M2) cannot interact with PAA. (iii) Interaction of PRSV wild type (wt) HcPro with itself and with the HcPro mutants. The wtHcPro can interact with itself and with HcPro (M1), but not with HcPro (M2). B. Western blot analysis of yeast total protein with mouse anti-myc antiserum for expression of all the analyzed HCPro mutants and proteasomal proteins, along with the positive control (+) and negative control (-) mentioned above. C. Subcellular distribution in Nicotianabenthamiana epidermal cells of green fluorescent protein (GFP)-tagged PRSV HcPro and its mutant proteins M1 and M2, expressed transiently by agroinfiltration, as well as of fibrillarin tagged with monomeric red fluorescent protein (Fib-mRFP), as a nucleolar marker. Green fluorescence derived from proteins tagged at their C-terminus with GFP: (i) HcPro-GFP, (ii) HcPro (M1)-GFP, or (iii) HcPro (M2)-GFP. In all three cases, fluorescence was found in both the cytoplasm and nucleus, excluding the nucleolus. Red fluorescence derived from Fib-mRFP was confined to the nucleolus (arrows). Bars in the lower right corners represent 20 µm. D. Bimolecular fluorescence complementation (BiFC) between PRSV HcPro and Carica papaya PAA protein tagged at their N- and C- termini, respectively, with split yellow fluorescent protein halves (sYFP) expressed transiently by agroinfiltration in Nicotianabenthamiana epidermal cells: (i) sYFPN-HcPro plus sYFPC-PAA, (ii) sYFPN-HcPro (M1) plus sYFPC-PAA, and (iii) sYFPN-HcPro (M2) plus sYFPC-PAA. In this latter case, a field of several epidermal cells is shown instead of a single cell, with nucleoli appearing red (arrows) because of the presence of Fib-mRFP. The interactions of HC-Pro and HcPro (M1) with the PAA proteasomal subunit both took place in the cytoplasm, with fluorescence distributing mainly with a reticulate appearance. Bars in lower right corners represent 20 µm in all panels, except in the lower panel to the right, where it represents 50 µm.

program or for manipulating the thermo tolerance of wheat using genetic engineering tool in order to develop climate-smart wheat crop.

To study the differential role RNAi suppressor proteins in plant development viral derived RNAi suppressors from begomo-, tospo-, cucumo and potyviruses were characterized followed by binary construct development of RNAi suppressors; NSs, HcPro, 2b and AC4. In planta expression of these suppressors was conducted to evaluate their role in plant development. Artificial micr RNA based constructs to silence NSs and HcPro were designed and transient expression of amiRNA constructs to evaluate the efficacy of these construct in gene silencing was conducted. Attempts were made to express NSs in prokaryotic systems. Transgenic tomato were developed using amiRNA NSs construct.

MicroRNAs are new class of regulatory molecules that controls gene expression. Besides their roles in developmental biology and maintenance of genome integrity, miRNAs are also integral components of plant responses to adverse environmental condition, including biotic stress. To investigate the molecular mechanism of *R*-gene mediated resistance, miRNA profiling was conducted in wheat during stem rust infection in presence and absence of *R*-gene. The changing profiles of eight miRNAs (5 miRNAs *viz.* miR159, miR164, miR167, miR171, and miR444 involved in regulation of phytohormone and 3 miRNAs *viz.* miR408, miR1138, miR1129 are involved in cellular function homeostasis) during stem rust infection in wheat are reported. Analysis of data revealed that the regulation of miRNA levels differed in early and late infection stages. These miRNA levels were also compared in presence and absence of *R*-gene *Sr24*. The higher accumulation of these miRNAs in absence of *R*-gene provides a basis for initiating study on genetic network of *R*-gene mediated defense responses. Sequence and structural comparison of miRNA backbone (pre-miR159a and pre-miR1123) suggests conserved evolutionary link in the region, specific for formation of miRNA duplexes as well as in sequences required for processing. The results discussed in the present study are the first report of involvement of miRNA driven pathways during stem rust infection in wheat. The finding reported here will help us to better understand the complexities of the miRNA mediated regulatory mechanism during biotic stress.

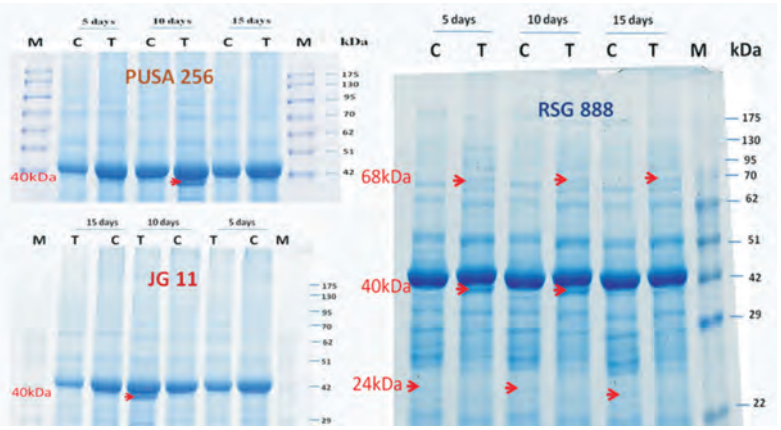
Tospoviruses have unique ability of infecting plants through insect vectors and cuase significant losses in many economically important horticultural crops. They belong to family *Bunyaviridae* and interestingly, besides tospoviruses, four other genera in the family *Bunyaviridae* affect only animals. The genome of tospoviruses consists of three species of single stranded RNA denoted by L, M and S. Phylogenetic survey of recombination events in all the 3 genomic components of GBNV with 8 other tospoviruses and 4 belonging to animal infecting genera of *Bunyaviridae*suggested the molecular evolution of these viruses from animal infecting members of *Bunyaviridae.* Based on recombinational analysis the two important proteins of tospoviruses; residing on the M genome responsible for viral movement (NSm) and glycoproteins (Gn/Gc) involved in insect recognition were found to be evolved by the genome of animal infecting *Bunyaviridae.* Functional significance of Non-structural protein residing on the S genome of the GBNV virus in the host counter defense was studied for the first time and the demonstrate the role of GBNV-NSs in RNAi silencing through epifluorescent bioassay using site-directed mutagenesis. The active protein site responsible for RNAi silencing has been mapped. Leucine rich motifs were found crucial for RNAi suppression. Besides, being a RNAi suppressor, GBNV-NSs was also found to control programmed cell death through miRNA profiling. The study suggested that senescence or programmed cell death, the typical symptom of tospoviruses infection might be triggering through NSs activity. Initiation of senescence might be trigged through three independent pathways involved in regulation of metacaspase and phosphate transporter. These findings will help in the undertaking of host-virus interaction during GBNV infection for developing wide spectrum management strategies.

Viruses exclusively depend on host cell machinery for their propagation and survival; hence they modulate the host gene expression to suit their needs. Thus the range of interactions between various host and viral factors at the host virus interface are interesting to study. These interactions can be fine-tuned in the host by modulating gene expression through triggering defense mechanism and allowing epigenetic modifications. In the viral genomes high rate of mutations add to the complexity of these interactions and help in developing

adaptations to different hosts. In the present study, we analysed host adaptability of *Tomato leaf curl New Delhi virus* by mutation studies in the pathogenicity factor-AC4 during passaging through three different hosts. The N-myristoylation domain responsible for pathogenicity was found to be more prone to mutations. AC4 was also found to play an important role in stress signaling by regulating miRNAs in auxin biosynthesis. RNAi triggered epigenetic modifications in the AC4 gene was found to be prominent host defense pathway. AC4 was also found to be linked with phenotypic expression of symptoms by its role in accumulation of seven studied miRNAs involved in plant developmental processes in three different hosts. Collectively AC4 a pathogenicity factor/RNAi suppressor was found to be crucial for its role in regulating host gene expression, by stress signaling pathway. Host response to infection was studied by RNAi triggered epigenetic modification in this viral gene.

The ubiquitin/26S proteasome system (UPS) plays an essential role not only in maintaining protein turnover, but also in regulating many other plant responses, including plant–microbe interactions. Previous studies highlighted two separate roles of the UPS in plant defense during virus infection, either indirectly through viral suppressor-mediated degradation of Argonaute proteins, affecting the RNA interference pathway, or directly through the RNase activity of the 20S core component of UPS, affecting the levels of viral RNA. The present study was focused on role of protein degradation machinery, the ubiquitin/26S proteasome system (UPS), as one of the mechanism of plant defense. The proteosomal inhibition studies using MG132, a cell permeable proteasomal reversible inhibitor, caused an increase in Papaya ringspot virus (PRSV) accumulation in its natural host papaya (Carica papaya). The increase in the levels of viral transcripts, viral titre as well as phenotypic expression of symptoms were evident during proteosomal inhibition. The increase in the levels of viral transcripts suggested the probable RNAse activity of proteosomes, playing crucial role in regulating RNA viruses. Hence, it was concluded that the 20S core proteosome imposed direct defense against papaya ringspot virus.

Plant viruses invade host cells and utilize the cellular pathways of the host to support various aspects of their infection cycle. To achieve these goals viruses must utilize the multi faceteddefense mechanism of the host to make them susceptible for virus establishment. Hence the functional significance of viral proteins in modulating host defense responses is interesting to study. The main focus in the present study was the functional significance of *Papaya ringspot virus* (PRSV) derived protein Helper component Proteinase (HcPro) during viral infection in host. The present study highlights the essential role of Ubiquitin/26S proteasome system in plant defense responses during PRSV infection in papaya. Also altered expression was observed for microRNAs involved in plant defense and hormonal signaling in the papaya leaf tissues, suggesting their probable role in symptom development.



SDS-Page analysis of HT treated chickpea seedling at different stages of treatment revealed variations in protein profile in seedlings, where as no visible difference were observed in root tissue.

Further the study emphasized the importance of viral protein HcPro in modulating catalytic activities of 20S core of proteasomal complex, in support of virus accumulation through molecular interaction with proteasomal subunit 1 and proteasomal subunit 5. Besides altering 20S proteasomal catalytic activities, HcPro was also found to play an important role in small RNA biogenesis, by its capacity to bind double stranded small RNAs, leading to systemic suppression of RNA silencing. The two well characterized domains at N terminal and the central region of the HcPro protein were found to be associated with these activities respectively. Taken together the present study reveals the critical importance of PRSV-HcPro in modulating plant defense responses like RNAi and proteasomal pathways in favour of virus accumulation.

Micronutrient malnutrition, often called "hidden hunger", is one of the alarming problems in the developing world, with estimated 400 million people including 100-200 million children are at risk of Vitamin A deficiency. Maize is one of the most important cereal crops in the world, however, the traditional yellow maize varieties posses low amounts of provitamin A which is far below the prescribed amount of daily requirement by human. It is therefore important to ascertain the genetic variability for kernel provitamin A content in the available maize germplasm and their potential to utilize inbreeding programmes. The present study revealed that, among the 111 lines analyzed, total carotenoid content ranged from 1.3 to 67.3 µg/g with a mean of 26.6 µg/g, while β carotene content varied from 0.1 to 14.9 µg/g. Among the lines, CM 138 was identified as the best genotype with 14.9 µg/g of β carotene content followed by NAI 125 (14.2 µg/g), CM 136 (12.7 µg/g) and HPLET-03-8 (12.3µg/g). In general, Harvest Plus lines were found to be much superior to the Indian lines in terms of total carotenoid content. The study also showed strong association of kernel colour with that of total carotenoid, but the same was not found to be effective for β carotene content. Allele polymorphism in the 5'UTR of *Y1* gene could not be associated with high/low content of total carotenoid and β carotene content in the selected set of genotypes. Analysis of 3'TE assay in the 3'UTR region of *crtRB1* gene showed that the genotypes lacked the favourable allele, thereby suggesting the need for introgression of the favourable allele from temperate background into tropical/sub-tropical germplasm. Polymorphism analyses using micro satellites markers (SSR) among the selected set of 24 inbred lines revealed 103 alleles with a mean of 4.3 alleles per locus. The average polymorphism information content (PIC) was found to be 0.57, while genetic dissimilarity coefficient varied from 0.13 to 0.85 with an average of 0.36. Cluster analysis grouped 24 genotypes into three major clusters with each comprised of lines having both high and low carotenoid content. The study thus aided in identifying promising inbred lines with phenotypic contrast and genetic divergence which could be utilized in breeding programme for enhancing high carotenoid content in the tropical maize germplasm.

Maize is one of the world's leading cereal crops along with rice and wheat. It is highly popular mainly because of its diverse functionality as a food source for both humans and animals and as an industrial raw material for a number of products such as starch, oil, bio fuels etc. Its kernels can be consumed off the cob, parched, boiled, fried, roasted, ground and fermented for use in breads, porridges, gruel, cakes, and alcoholic beverages. Industrial processing leads to its use as food thickeners, sweeteners, oils and non-consumables. Starch constitutes about 68-72 % of mature kernel on dry weight basis. Sugars range from 1 to 5 % with sucrose as the chief component and maltose, glucose, fructose, and raffinose in trivial amounts. constitutes around 7-13 % of the mature kernel. Fat in the form of oil is the 3rd largest nutritional component of maize kernel, ranging from 3 to 6 % of total kernel weight with an average of around 4.0 %. The maize oil is highly regarded because of its better fatty acid composition and the presence of essential fatty acids in it. Since maize shows wide genetic variability, it is assumed that genetically diverse maize genotypes possess unique fatty acid profile. In the present study different maize genotypes were compared for their oil composition along with nutritional profiles. The available maize lines were categorised according to their pre-established genetic background as normal, high oil, quality protein maize (QPM), and sweet corn. Oil was extracted using soxhlet apparatus, and subjected to GLC for oil profiling. Other nutritional parameters studied were protein, starch, and sugar. The results showed that oil content of high oil maize genotypes (group 2) is significantly higher compared to group 1 (normal maize). The mean value of group 2 for oil concentration is 5.5 % compared to 4.4 % of group 1. Individual fatty acids showed a wide difference between normal and high oil lines. Palmitic acid exhibited a slight elevation from 12.61 % in group 1 to 14.73 % in group 2 genotypes. However group 4 exhibited significantly higher palmitic acid compared to group 1. On the contrary stearic acid demonstrated a significant decline of 56 % in high oil genotypes (group 2) compared to group 1 containing normal maize germplasm. Oleic acid also showed a decline although non-significant from 46.61 (group 1) to 43.31 (group 2), whereas, group 4 showed significantly reduced oleic acid compared to normal and high oil lines. However, a 20 % increase was observed in the linoleic acid in high oil maize (group 2) compared to normal maize (group 1), whereas, significantly increased linoleic acid concentrations were observed in group 3 and 4 compared to group 1. Regarding protein content no significant differences were observed between group 1, group 2 and group 3, whereas the protein content of

group 4 (sweet corn) was significantly higher compared to rest of the groups. High oil maize genotypes (group 2) showed significantly lower starch content compared to normal maize, whereas sweet corn genotypes (group 4) showed drastically lowered starch contents. Although no significant difference was observed in sugar content between group 1, 2 and 3, however, group 4 genotypes showed a significantly higher concentration of sugar as compared to rest of the groups. Present study indicated that oil composition in high oil genotypes (group 2) is better compared to normal maize (group 1) genotypes, however, the increase in oil comes at the cost of starch which is directly proportional to grain yield.

More than 100 maize inbreds for protein quality (protein, lysine and tryptophan) along with micronutrients such as carotenoids, iron, zinc were evaluated. A large variability was observed in the carotenoids among germplasm obtained from Delhi and Bajuara research station. A significant positive correlation ($r = 0.491^{**}$) was observed in kernel color and total carotenoid content, whereas no significant correlation ($r = 0.014$) was observed between kernel color and β-carotene. Normal lines were found possess higher iron concentration as compared to QPM lines. A total of 7 lines {(HKI 164-4(1-3), HKI 34(1+2)-1, HKI 164-4(1-3)-2-2, HKI 193-2-2-2, MRCHY 4840, EC 618225-2 and HKI 1128} were identified in these studies as the most promising lines enriched in all the micronutrients under study.

A set of 100 inbred lines, grouped as 25 lines, each of normal and QPM germplasm obtained from two different locations (Delhi and Bajaura) were analyzed for protein quality and micronutrients such as carotenoids composition as well as iron and zinc concentration. Results revealed that kernel colour intensity and total carotenoids exhibited a highly significant positive correlation, whereas no significant correlation was observed between kernel colour intensity and β-carotene. Nine QPM [HKI-3-4-8-6, HKI 34(1+2)-1, HKI 164-4(1-3)-2-2, HKI 164-4(1-3), HKI 193-2-2-2, NP-06-07R-76-8, NP-06-07R-80-6, LQPM-42 and LQPM-40] and eleven normal lines [EC-618225-2, MRCHY 4840, NZB-1-2-1-3-1, SEED TECH 89-2-3, EC-4400248, BAJIM-08-27, BAJIM-13-1, BAJIM-13-2, HKI 1105, HKI 1128 and HKI 1155] were identified as promising lines in terms of carotenoid composition. Significantly, higher iron concentrations were found in normal as compared to QPM lines. Seven inbreds [HKI 164-4(1-3), HKI 34(1+2)-1, HKI 164-4(1-3)-2-2, HKI 193-2-2-2, MRCHY 4840, EC 618225-2 and HKI 1128], were identified having the highest concentrations of carotenoids along with iron and zinc concentrations. Wide variability was observed in the QPM germplasm with respect to lysine and tryptophan concentrations. Five inbreds [LQPM-43, LQPM-40, NP-06-07R-76-8, HKI 164-4(1-3)-1, CML 153] were found to be having the desired threshold concentrations of protein quality for QPM breeding. CML 168 was identified as the most promising QPM line from Bajuara containing high concentrations of lysine, tryptophan as well as methionine. In general high methionine concentrations were observed in QPMs as compared to normal germplasms. These findings will be useful in future breeding programmes in developing nutritionally improved maize genotypes.

Chickpea is the world's most widely cultivated pulse crop. Checkpea production is significantly affected by the abiotic factors like temperature and drought which are the most important factors affecting quality and quantity. The loss to crop due to temperature stress from 10-40%. An experiment was conducted with three chickpea genotypes Pusa 256 (National Check-North Zone, recommended by late planting), RSG 888 (West Zone, drought tolerant), JG11 (South Zone, recommended for early planting and rainfed cultivation) to study the effect of high temperature stress on physiological parameters (RWC, MSI, chlorophyll, dry matter, leaf area) and biochemical parameters (antioxidant enzymes and protein profile). In all the chickpea genotypes high temperature stress decreased RWC, MSI, Chl 'a' and 'b' content, dry matter and leaf area and increased activity of antioxidant enzymes such as POX, GR and SOD. Correlation coefficient between antioxidant enzymes and various physiological parameters revealed that under high temperature conditions there existed a negative correlation between antioxidant enzymes with RWC and MSI. Significant positive correlation was also observed with dry matter and leaf area. SDS-PAGE analysis of NT and HT treated chickpea seeding at different stages of treatment revealed variations in shoot tissue but no visible differences were observed in root tissue. A polypeptide of approximately 40 kDa was observed in all the three genotypes in response to heat

stress. In addition, two polypeptides of approximately 24 and 68kDa were found to be differentially expressed in response to heat stress in RSG 888 only. RSG 888 possessed better seedling growth parameter under high temperature as compared to Pusa 256 and JG11.

Pigeon pea [*Cajanuscajan* (L.) Millsp], a short-lived perennial shrub, commonly known as 'Arhar' or 'Tuhr' is one of the major grain legume (pulse) crops of the Indian subcontinent. It has a diploid genome with 11 pairs of chromosomes and a genome size estimated to be 858 Mbp. Salinity being a critical factor to sustainable pigeon pea production addresses limitations to meet the demands of resource-poor people where it is grown. Based on percent germination, vigor and salt tolerance, out of total 175 pigeon pea genotypes,12 genotypes were selected for morphological screening. Prior to this, different concentrations (0-300mMNaCl) and durations (0,7, 14, 21 days) of salt stress imposition were also determined. Salt stress with 250mMNaCl for 7 days of stress treatment was found to be optimum for further biochemical and molecular studies. Six most salt responsive pigeon pea genotypes, three from each group of salt-tolerant (ICP6815, ICP7, ICP8860) and salt-susceptible (ICP1071, ICP14155, ICP15185) genotypes were subjected for morphological and biochemical screening to identify one each of most salt-tolerant and salt-susceptible contrasting pigeon pea genotype. Biochemical studies-antioxidant potential (AO), lipid peroxidation (LP) and total phenolic contents (TPC) were carried out in root and shoot tissues of screened 6 genotypes. Based on morphological and biochemical expressions, ICP7 and ICP1071 were found to be most salt-tolerant and salt-susceptible pigeon pea genotypes, respectively. Biochemical expressions were found to more prominent in the shoot as compared to the roots with reference to phenol content and lipid peroxidation but in the case of antioxidant potential it was more or less same. Findings showed that TPC, LP and AO activities are determinant biochemical markers for salt-stress tolerance. A progressive increase in TPC and AO activity were observed during salt-stress imposition in the salt-tolerant genotype than salt-susceptible and LP was found to be in reverse pattern. Partial CDS for pigeon pea stress-responsive genes viz., hybrid proline-rich protein (*CcHyPRP*, 218 bp), cold, drought salt regulatory protein (*CcCDR*, 207 bp) and cyclophilin protein (*CcCYP*, 323 bp) conferring abiotic tolerance were cloned from ICP7 genotype and sequences were submitted to NCBI with the Accession Nos. KP996197, KP996198 and KR262820, respectively. The BLAST search sequence homology characterization revealed their 86-99 % similarity with *Cajanuscajan* and *Glycine max*. The semi-quantitative (RT-PCR) expression analysis of these isolated genes were conducted in root and shoot tissues of contrasting pigeon pea genotype and found to be expressed in both root and shoot tissues. These findings shall go a long way not only in understanding the important molecular and biochemical mechanisms underlying the salinity tolerance of the pigeon but also can be utilized in providing important leads to the breeders to evolve improved varieties of pigeon pea with higher salinity tolerance levels.

Acknowledgement

We wish to thank all the Faculty Members of the Division of Biochemistry for extending support and cooperation in preparing the research contributions of M.Sc. and Ph.D. students made during the period of 2007-16.

References

M.Sc. and PhD theses submitted by the students in the Division of Biochemistry, ICAR-Indian Agricultural Research Institute, New Delhi 110012 during the period of 2007-2016.

Molecular Biology & Biotechnology

R.C. Bhattacharya

Introduction

The teaching discipline of Molecular Biology and Biology is entrusted with the responsibility of developing and utilizing new tools and techniques of plant biotechnology in agricultural research of India. Therefore, the research work undertaken by the post-graduate students consists of basic as well as applied aspects of plant biotechnology for addressing major challenges of Indian agriculture. The salient achievements of research by the post-graduate students during the last ten years have been grouped under a few major subheads. While doing this onerous task of comprehending a vast body of research findings within the limited space of the document, more emphasis was given to ensuring representation of diverse areas of researches that have been pursued by the students in lieu of thoroughly describing the achievements in each of them. Further details on each of the items are available in the annual report of this centre or in the published papers.

Significant Student's Research Achievements

Identification, cloning and characterization of genes/novel alleles involved in agronomically important traits of crop plants

Rice blast caused by *Maganaporthe oryzae* causes significant yield loss every year in rice. Two blast resistant genes *Pi-rh* (an allele of *Pi-k*^{h} gene) and *Pi54* were cloned from blast resistant wild rice species, *Oryza rhizomatis* and indica rice cultivar Tetep respectively (Fig. 1). Functional validation of both these genes were done through transgenic approach. Further in *M. oryzae* two *Avr* candidate genes i.e., *Avr: Pi-k*^{h}*-1* and *Avr: Pi-k*^{h}*-2,* were cloned and characterized. *Avr* gene for pi54 was also identified in *M. oryzae* genome.Mo-01947_9 as the most likely candidate *Avr* gene for *Pi54*. This gene, located on chromosome 4 of *M. oryzae* was cloned and direct physical interaction of its protein with Pi54 protein was established through yeast two-hybrid analysis. For functional validation, the virulent *M. oryzae* strain MG-79 was transformed with Mo-01947_9 gene and challenge inoculated on rice lines carrying *Pi54* gene (Fig. 2). Absence of disease reaction confirmed that the cloned gene is indeed the *AvrPi54* gene. Panicle Blast is also one of the important diseases of rice. So far, only one gene, Panicle blast 1 (Pb1) is cloned and characterised for panicle blast resistance. So *Pb1* allele along with its promoter is amplified and sequenced from 55 Indian rice accessions. Further it was found that this gene expression is more in panicle than leaf and its expression is higher in panicle resistant genotypes.

Another important rice disease is bacterial leaf blight caused by *Xanthomonas oryzae* pv. *oryzae (Xoo).* From six different Xoo isolates 10 different TALe genes were identified from each. Additionally a candidate gene in response to infection by these isolates were identified as sucrose transporter SWEET 11. The induction of SWEET11 by three out of four *Xoo* isolates reiterated the prevalence of cognate TALe pthXo1 even in Indian isolates which fail to find its target in the recessive allele of SWEET11 i.e. *xa*13, hence culminating in resistance. However, one of the isolates compatible with *xa13* facilitated its proliferation in host by binding to another SWEET gene target i.e. SWEET14. These results strongly support the importance of TAL effectors and its targets in determining BLB disease and pave way for development of engineering strategy exploit these determinants to aid in disease control.

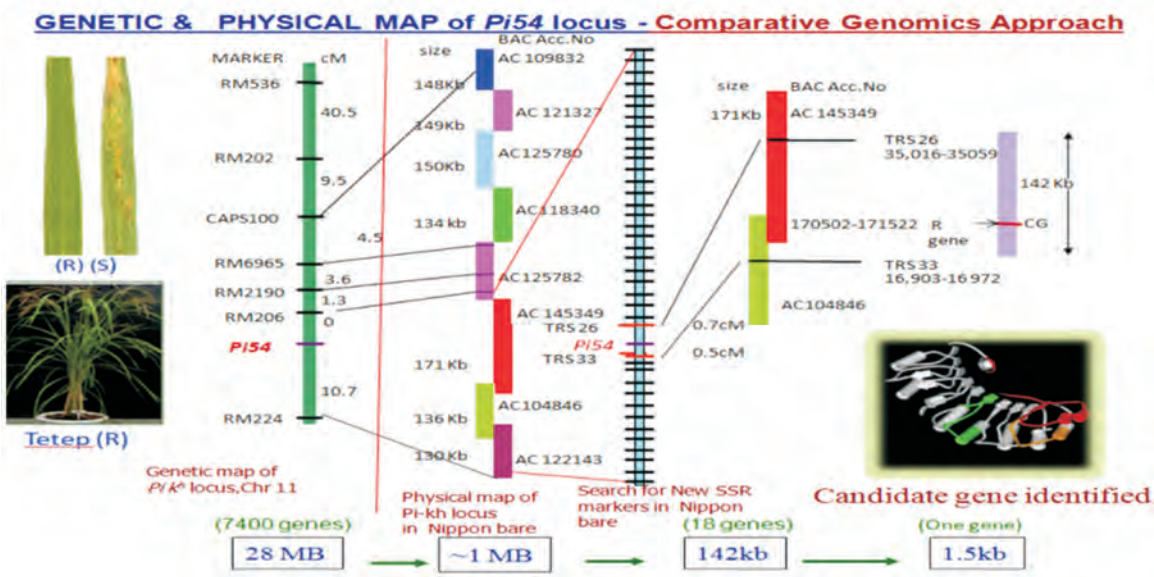


Fig.1: Mapping and cloning of blast resistance gene Pi54.

Fig 2: An example of rice lines containing three blast resistance genes in the background of BPT5204. Blast resistant lines (left) and susceptible lines (right).

Fifty insecticidal crystal proteins of *Bacillus thuringiensis* specific to lepidopteran insects were selected and multiple alignment of the amino acid sequence for the whole toxins was carried out. Cry1Jb and Cry1Ac showed considerable difference in the composition and length of the loops of domain II and III of toxins. Insect bioassays using *E. coli* expressed proteins revealed that the wild type Cry1Ac and Cry1Jb proteins showed differential specificity to *Helicoverpa armigera*, *Spodoptera litura* and *Earias vitella*. Chimeric genes were constructed viz., *Jb-Jb-Ac*, *Jb-Ac-Jb* and *Jb-Ac-Ac* by exchanging the domains between *cry1Ac* and *cry1Jb* and over expressed in BL21 (DE3). JbJbAc showed 7.8 fold higher toxicity followed by Jb-Ac-Jb and Jb-Ac-Ac with 7.5 and 3.6 fold higher toxicity than the Cry1Jb towards *Helicoverpa armigera*, respectively. Bioassay of the parental and chimeric toxins against *Spodoptera litura* revealed no activity even at the discriminative dose of 100 µg/ml but significant growth difference was observed. The LC_{50} of Cry1Ac against *Earias vittella* was 14.8 ng/ml. Chimeric proteins were also found to be effective against *Earias vittella*. No toxicity was observed for Cry1Jb against *Earias vittella* even at a high concentration of 100 µg/ml. *Agrobacterium tumefaciens*-mediated transformation method was used for introduction of the native truncated parental and chimeric toxins in tobacco. ELISA revealed that the expression of native truncated parental and chimeric toxins in the leaf of transgenic tobacco

ranged from 0.001% to 0.007% of TSP. Strong correlation was seen between the levels of *cry* genes expression and the insect control achieved. The results, along with previous domain swapping experiments suggests that protein engineering not only reveals the mechanism by which endotoxins work, but it can generate toxins with enhanced toxicity/introduce a specific activity into a toxin that does not possess it.

Bacillus thuringiensis is a bacterium of great agronomic and scientific interest. Various cry genes were characterized from native Bt isolates collected from diverse origin habitats and locations in India and submitted to NCBI database as well as to *Bacillus thuringiensis* Nomenclature Committee http://www/lifesci.sussex.ac.uk/home/nei_crickmore. *The Bacillus thuringiensis* Nomenclature Committee has provided names *cry*2Ab16 for *cry*2Ab-type gene (Gene Bank Accession No. GQ866914), and name *cry*2Af2 for *cry*2Af-type gene (Gene Bank Accession No. GQ866915) cloned in this study. Efficacy of Cry2Af2 protein from SK-758 native isolate (collected from Jowar Grain dust, Guntur, Andhra Pradesh)was tested against *H. armigera* in laboratory bioassays and was found to have moderate toxicity.

A novel approach for identifying important genes from *B. thuringiensis* using DNA pooling strategy known as Pooling-deconvolution were developed. Furthermore, structural comparative analysis of three *cry* genes *viz* IP7 (NCBI ID: KM053254), IP11 (NCBI ID: KM053252) and IP13 (NCBI ID: KM053253) isolated by this method revealed that the sequence of IP11 protein has >96% homology with Cry4Ca1. this protein was named Cry4Ca2 by *Bacillus thuringiensis* nomenclature committee http://www.lifesci.sussex.ac.uk/Home/Neil_Crickmore/Bt. The expression of *ip13* gene in *Escherichia coli* resulted in a water soluble protein which is toxic against *Helicoverpa armigera.*

The candidate gene related to grain size was mapped on short arms of chromosome 5 which was further fine mapped to an interval of 250kb. Within this region the candidate gene *SBG5* was identified which encodes kinesin 13 family proteins in rice, and has major roles in mediation of microtubule organization during mitosis. The loss of function of *sbg5* gene resulted in reduced rate of grain filling and less amount of dry matter accumulation in the endosperm during the peak grain filling stage of panicle development. Cytological studies of lemma, revealed shorter and the lesser number of cells in the mutant as compared to the wild type contributing to shortening of grain length.

Abiotic stresses like drought, salinity, heat, cold etc. impose major limitation on crop production, and therefore its management is one of the most important challenges that today agriculture is facing. Through bioinformatic approach 24 HSF were identified in pigeon pea out of which 14 were selected for their detail characterization. The phylogenetic relationships, gene duplication and expression profiling of *CcHsf* genes revealed that *CcHsf* genes were distributed into 8 groups, out of a total 13 groups. Among the CcHsf genes, *CcHsfA-1d* and *CcHsfA-2* were prominently expressed during the heat stress condition. In chickpea, out of several HSF an ortholog of Arabidopsis *MBF1C* was found to be upregulated ~30 fold in heat stress. Over expression analysis of *CarMBF1C* in Arabidopsis showed tolerance to heat stress in germination and seedling thermotolerance assays. GUS analysis of 2 Kb promoter region of chickpea *CarMBF1C* showed GUS expression in epicotyls and elongation region of roots only in combination of heat and dehydration stresses. Thus, identification of heat stress responsive genes in pigeon pea and chickpea will help in elucidating thermo-tolerance mechanisms in these pulses.

In wheat based on advanced genomics tools 59 HS (Heat stress) responsive genes including 9 members of Caseinolytic protease (ClpATPase) family, 13 members of Small Heat Shock Protein (HSP20) family and 37 HS responsive genes (excluding HSPs and HSF) were identified in bread wheat. Spatio-temporal, biotic and abiotic stress-specific expression pattern in normalized wheat array data sets revealed constitutive as well as inductive response of *TaClps* and *TaHSPs* in different tissues and developmental stages. Five HS responsive genes namely *TaHSA32*, *TaFAD8*, *TaCPR5*, *TaBI* and *TaDREB2a* having diverse cellular functions were mined in a set of 64 wheat genotypes having contrasting thermotolerance. Seven genotypes namely Stiletto, Warigal, WL 711, Kalgarin, Arena, Kharchiya local and Luan displayed one SNP at 379th position, a A to C transversion

and one dinucleotide insertion CA at 411th position in *TaHSA32* gene. The genotype Wilgoyne has one SNP at 660th position, a T to C transversion in *TaDREB2A* gene. However, this mutation doesn't change the amino acid sequences of TaHSA32 and TaDREB2A. *TaDREB2a* and *TaHSA32* are highly conserved among wheat genotypes, whereas *TaFAD8* and *TaBI-1* displayed higher sequence difference. One SSR derived from *TaCHIP-1* gene displayed polymorphism among the seven genotypes differing in thermotolerance. The data obtained from this study contribute to a better understanding of the complexity of the HS responsive genes in bread wheat, and provide the basis for further studies to dissect the function of these genes during plant growth and development as well as in response to environmental stimuli.

A close relation exist between drought and photosynthetic efficiency of plants. To understand the effect of high temperature and water deficit on photosynthesis, quantitative expression analysis of seven important photosynthetic genes were carried out viz. Rubisco large subunit (*RbcL*), Rubisco small subunit (*RbcS*), Rubisco activase A (*RCA-A*), Rubisco activase B *(RCA-B),* Rubisco Binding Protein (*RBP*), Ferredoxin NADP+ oxidoreductase I (*FNR I*), Ferredoxin NADP+ oxidoreductase II (*FNR II*)) in six wheat genotypes (*Triticum aestivum,* cv.HI1531, HD 2877, Raj 3765, HD 2833, WR 544, and PBW 373) under abiotic stress conditions namely drought, heat and combined heat and drought conditions. Study showed an association between gene expression levels and photosynthesis, and revealed a significant genotypic variation in the expression of key photosynthetic genes under these three abiotic stresses.

Terminal heat stress is one of the limiting factors in wheat production and it is expected to rise under present scenario of climate change. SnRK1 (Sucrose non-fermenting-1-related protein kinase 1) is a key metabolic regulator which plays an important role in plant carbon, nitrogen metabolism and development. SnRK1 expression profiling was done in two cultivars of wheat HD2967 and C306 with contrasting response to abiotic stress. Differential, abundant and positive modulation of SnRK1 was observed in both the cultivars during early as well as terminal heat stress and also under regular growth and development conditions. Contrasting expression was observed with higher expression in early vegetative stages for C-306 and generative stages for HD2967.A full coding region of SnRK1 catalytic subunit was cloned from variety HD2967 and confirmed by various in *silico* analyses (Fig. 3).

In another study on wheat, three progenitor species of wheat; *T. monococcum* (AA genome), *Ae. speltoides* (BB genome) and *Ae. squarrosa* (DD genome) were screened for root traits under osmotic stress and it was found that *T. monococcum* accession A1-81 is the best performing accession for root traits under osmotic stress. Further a root length ortholog gene of Arabidopsis BREVIS RADIX (BRX) was cloned and sequenced (1191 bp) from this *T. monococcum* accessions (A1-81).

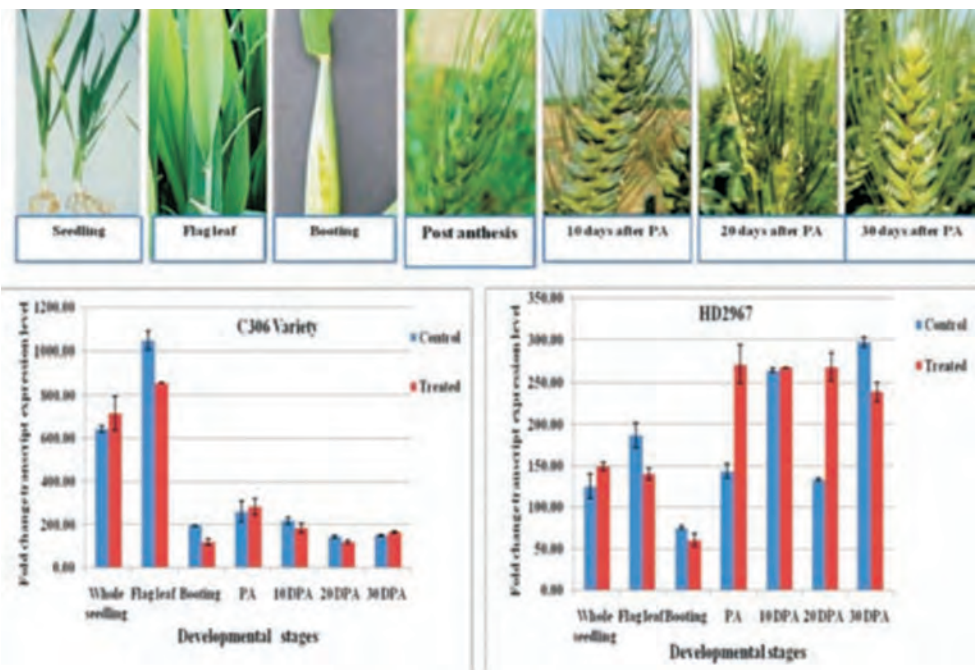


Fig 3. Different developmental stages of wheat cultivar at which samples were collected and expression profiling of TaSnRK1 gene was analysed in wheat genotypes, C306 and HD2967.

In rice allele mining of DREB2A transcription factor was done in 95

different rice accessions with the objective of identifying novel alleles for *DREB2A*, if any, and validating their expression under moisture deficit stress. Eighty-nine genotypes with complete and quality sequences were used for further bio-informatic analysis. A total of 398 unique positions had SNPs/InDels in *DREB2A*. In splice variant 1 and 2 (SV1 and SV2), 39 and 33 SNPs which had effect on amino acid change were found. SNP at position 1644 was found to be associated with panicle length ($p<0.03$) under drought stress (DS). Expression profiling of 10 selected genotypes exhibited significant upregulation in expression level of SV1 variant under DS in seven genotypes. Among all these genotypes, Satti genotype showed highest level of upregulation of SV1 under DS.

Alternaria blight is one of the most economically important diseases of *Brassica juncea* causing up to 45-58% loss in the yield with no proven source of transferable resistance in any of the host genotypes. Expression analysis of important defense genes in response to salicylic acid (SA), jasmonic acid (JA) and *in vitro* fungal infection as three separate treatments were carried out in *B. juncea*. It is observed that chitinase and thionin transcript levels were elevated in mustard leaves upon treatment with JA and during infection with necrotrophic fungal pathogen *Alternaria brassicae*. Conversely, expression levels of *PR1* and *NPR1* were induced exclusively upon SA treatment. They were not induced either by JA or after fungal infection. These results clearly indicated existence of two separate hormone dependent pathways i.e. SA and JA in *Brassica juncea* similar to *Arabidopsis thaliana.*

In this context, studies were conducted on defense gene induction in susceptible *B. juncea*, and moderately resistant and resistant wild relatives, *Sinapis alba* and *Camelina. sativa* to alternaria blight disease caused by *Alternaria brassicae*. Treatments with defense regulators such as salicylic acid (SA) or jasmonic acid (JA) or challenge inoculation with *A. brassicae* resulted in induction of an array of known defense-related genes such as *PR1*, *PDF1.2* and *PR3* at varied levels in all three plant species employed in the present study. Marker genes of SA (such as *PR1* and *PR2)* and JA signaling pathway (such as *PDF1.2* and *PR3*) were induced by SA or JA or *vice versa* in some or all three plant species, suggesting the occurrence of some cross talk among defense signaling pathways in each plant species, deviating from the signaling pathways of the model plant *Arabidopsis thaliana* belonging to the family Brassicaceae. For the first time, induction of four defense-related genes *viz.,PR1, PR3, NPR1* and *PDF1.2* in local as well as systemic leaves of the locally challenged plants of *S. alba* with *A. brassicae* as compared to *B. juncea* was explored. *PDF1.2* and *GST* expressed in local tissues while *PDF1.2*, *PR1, PR3* and *GST* expressed in systemic leaves of *C. sativa* challenged suggesting that both JA and SA pathways play significant roles during the systemic response triggered by *A. brassicae* inoculation. These defense genes induced by *A. brassicae* in *S. alba* and *C. sativa* have not been reported previously and hence they could be attributed to the degree of partial resistance of *S. alba* and resistance of *C. sativa* to *A. brassicae* infection.

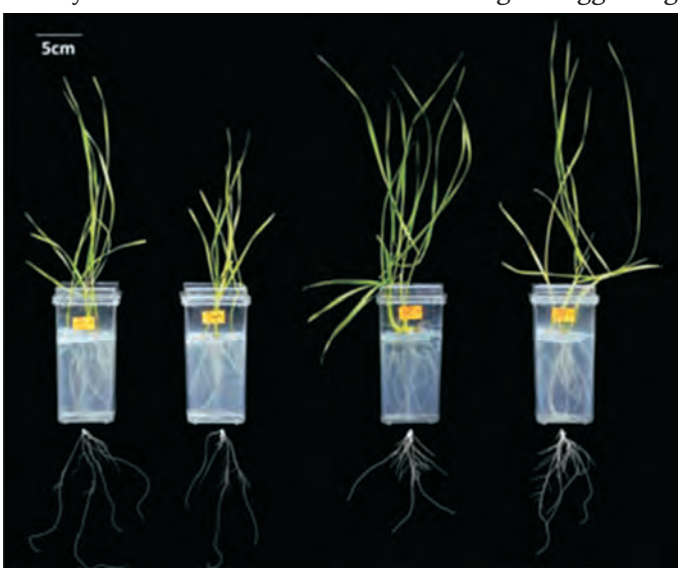


Fig 4. Wheat seedlings of Kalyansona (KS) and NP-890 (NP), growing in N-optimum and N-starved conditions in hydroponics.

Wheat is extensively cultivated worldwide and is the second most important cereal crop in India. Only 40% of the applied nitrogen is used by wheat crop and rest 60% is lost to the environment polluting it one or the other way. Hence, nitrogen use efficiency is a great concern for this important cereal crop (Fig 4). Gene expression of seven enzymes key to nitrogen use efficiency (NUE) were studied in eight wheat genotypes belonging to different group in terms of (NUE) under hydroponics condition with Nitrogen (N)-optimum (4.0 mM) and N-stressed (0.04 mM)

conditions. Soluble protein content, Chlorophyll content, four nitrogen assimilating enzymes namely nitrate reductase (NR), Gulaminesynthetase (GS), Glutamate synthase (GOGAT), Glutamate dehydrogenase (GDH) and three enzymes responsible in proving C-skeleton namely Pyruvate Kinase (PK), Citrate Synthase (CS) and Isocitrate Dehydrogenase (ICDH) were assayed *in vitro* in the leaf samples of 15 days old seedlings. NR activity and its gene expression reduced under N-stressd condition. However, N- assimilating enzymes did not show any NUE specific trend. Soluble protein and Chlorophyll content were reduced to a very high level in case of low NUE genotypes, which could be used along with increased specific activity of ICDH, PK and CS activity could be used as indicator for N-susceptibility and poor NUE of wheat genotypes.

In another study two genotypes HD-2967 and Kharchia showed significant increase in root dry weight under N-stress. RSA parameters showed increase in root length for all the genotypes except Kharchia and HD-2967. Root length, root diameter, the projected area, surface area and root volume showed increase. This study revealed that mainly 1st Order length contributed significantly toward the other significantly different parameters (Fig. 5). When the nutrient is deprived in the solid medium, the root extends in search of nitrogen (in the present study), and the root length increases. However, these parameters of RSA combined with root dry weight (which increased significantly) in case of HD-2967 and Kharchia puts these genotypes under a different root systems. Present study also identified contrasting genotypes WH-147 and VL-40, which could be the basis for future identification of important genetic components of RSA traits under nutrient limiting condition (Fig 6). Gene expression of transporters in diverse set of wheat genotypes at seedling stage seems to be one of firs reports under N-deprived condition. Kharchia showed minimum change in expression, whereas VL-401 and Kalyan Sona were distinctly different from the rest

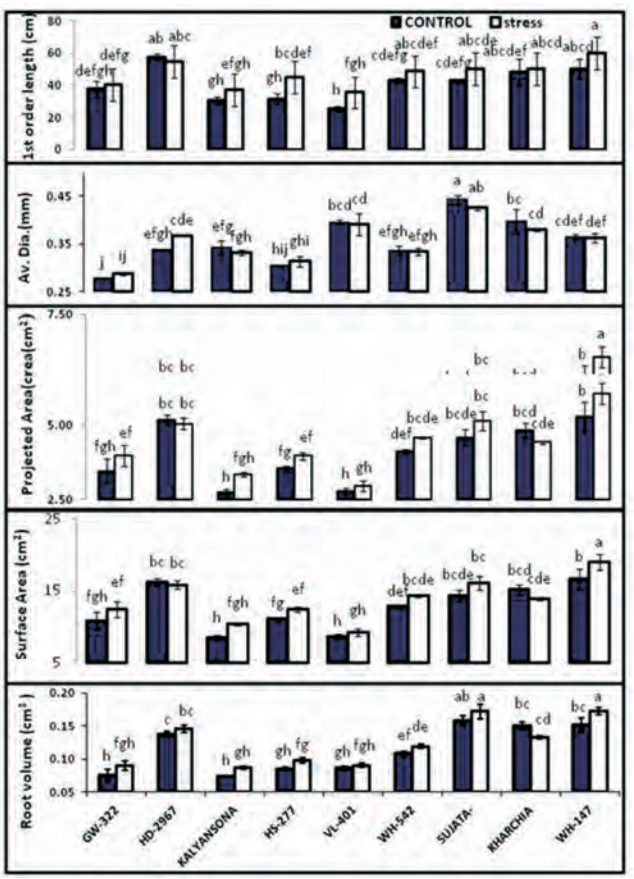


Fig 5. 1st order root length, root diameter, projected area, surface area, and volume of diverse wheat genotypes

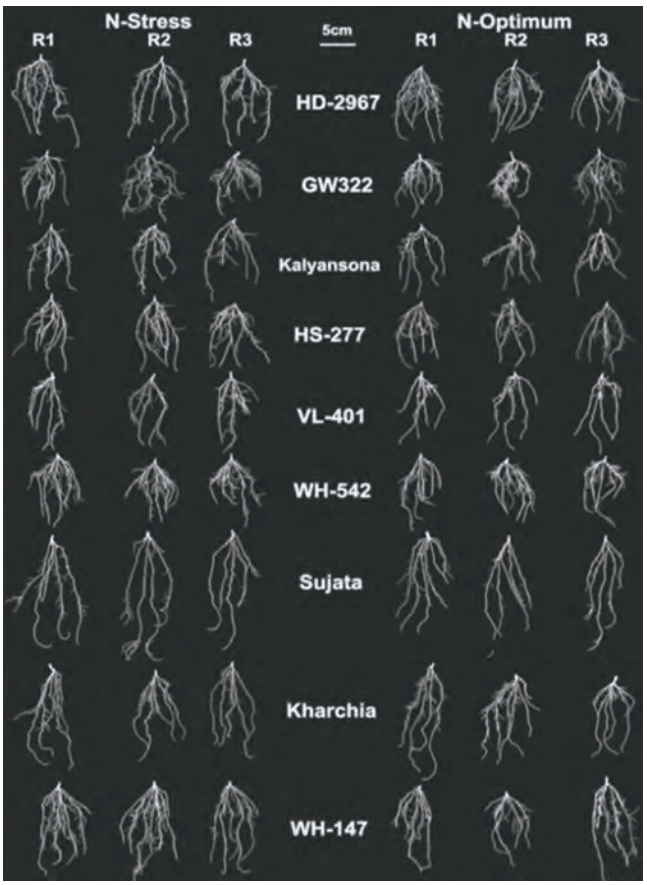


Fig 6. Scanned pictures of RSA for all the genotypes under study with three replications.

of the genotypes for LATs. Kharchia also showed its distinct character by significantly down regulating for HAT under N-stress condition.

Dof1 is a transcription factor, known for regulatory role in C-metabolism, in the TCA cycle. It has been proven that Dof1 also controls the NUE in plant by ensuring optimum C-skeleton supply for N-assimilation. *TaDof1* expression was studied in four diverse wheat genotypes under N-deprived condition which revealed that *TaDof1* expression goes up under N-starvation and decreases gradually with the increase in stress and ultimately down regulated under severe stress. NUE efficient genotype HS-277 showed relatively highest *TaDof1* expression under limited N-stress, mainly during 1st year, however the down regulation was also maximum under severe stress for this genotype. Two of the key enzymes in the N-assimilation process namely GS and GOGAT were assayed during Year 2. GS activity was significantly reduced in low NUE genotype when the N-stress was maximum during StageII. However, GOGAT activity did not show any trend. HS-277 was found different with respect to GS and GOGAT activity also. The *TaDof1* was further studied for their difference in gene sequence. Partial sequence (440bp out of 875 bp) in the coding region, mainly the C-terminal end, showed more than 99% homology among the genotypes, and same was the case when they were converted to amino acid sequences. Studying the complete gene sequence and allele mining of the gene would be further approach towards NUE in wheat.

Transgenic plants for developing resistance to biotic and abiotic stresses

In many of the crop plants lack of resistance source within the primary gene pool necessitates development of transgenics for deploying resistance genes from distant sources. Therefore, many of the research areas of P.G. students have attempted to source diverse transgenes from distant sources and assess their usefulness.

Plant lectins are carbohydrate-binding proteins and have a role in plant defense against insects including aphids. Several transgenic mustard plants cv. Pusa jaikisan carrying pigeonpea lectin (*CCL*) gene driven by *rol*C promoter were developed and characterized for their resistance against mustard aphids (Fig. 7). Lectin genes from pigeonpea and chickpea have also been cloned. Protease inhibitor gene and promoter from black gram (*Vignamungo* L.) were identified and their usefulness in bringing resistance against insect pest have been tested in transgenic tobacco. Insect bioassay showed larval growth of *Spodoptera litura* was severely retarded due to feeding on transgenic leaves as compared to their growth on control plants.

Fig 7. Diagrammatic representation of Chickpea lectingene and rolC promoter in a binary vector pORE4

As an alternate strategy, a novel approach of RNA interference (RNAi)-mediated gene silencing was employed to impart aphid resistance in mustard. For identifying important genes of mustard aphid a cDNA library has been prepared from aphids of all stages in order to identify important *Lipaphis* specific genes. Based on annotation studies finally 112 genes have been identified. 75% of the genes were found to be homologous with pea aphid, *Acyrthosiphon pisum*. In RT-PCR based expression analyses two genes have been identified that are highly expressed in the initial stages of infestation. RNAi mediated down regulation of these genes will lead to reduced survival of aphids. Transgenic Brassica expressing siRNAs specific to these aphid genes are

likely to show resistance to aphids. A plant transformation vector was developed which can express a hair pin loop transcript of aphid serine protease under a phloem specific promoter. This construct is useful to develop transgenic mustard which can deliver through phloem sap specific siRNAs into the aphid gut for silencing aphid serine protease gene.

For imparting effective resistance to biotic stresses quantitative multigenic resistance is important and more desirable compared to single gene. Peptide elicitors emanated as an important group of molecules, which are upstream regulator of the defense response cascade against insect herbivores and pathogens. In *Brassica juncea* two genes ncoding defense elicotor peptides have been identified. The potency of these defense elicitors in transcriptional activation of both JA and SA mediated defense response in *B. juncea* leaves involving activation of several downstream defense genes viz. PIN2, lectin, PR proteins, PGIPs etc. have been demonstrated. Further study clearly established their importance in activating innate defense of *B. juncea* and imparting resistance against biotic stresses.

For developing resistance to gram pod borer in chickpea and pigeonpea two important genes of *Heliothis armigera*, *acetylcholinesterase1* (*Ace1*) and *20-hydroxy ecdysone receptor* (*EcR*), involved in signal transduction and development, respectively, are targeted by host-delivered RNAi (HD-RNAi) for imparting resistance. The inverted repeat gene construct was introduced into tobacco lines which demonstrated moderate to high level resistance against *H. armigera*.

The use of resistance varieties has been considered as one of the best disease management options for rice blast (*Magnaporthe oryzae*). Transgenic lines containing the blast resistance gene *Pi-rh* from *Oryza rhizomatis* has been generated by transferring the gene to a susceptible japonica cultivar Taipei 309 (TP 309).Transformed plants were found to be resistant to *M. oryzae* isolate PLP-1 in T_0 and T_1 generations. These lines have been characterized at the molecular level in present investigation. Using three isolates Deh-1, Hz-1 and PLP-1, progenies of the T_1 plants were studied for blast resistance. The complementation of resistance phenotype was confirmed in all the lines indicating that *Pi-rh* gene was present in the genome of TP309. T_2 plant TP: *Pi-rh* 8.3.45 which showed high degree of resistance to *M. oryza*e was selected and its progenies were screened for resistance. All plants were resistant to Deh-1 isolate in T_3 generation showing that no segregation took place at this locus and indicating the homozygous nature of the gene in TP: *Pi-rh* 8.3.45. PCR and Southern analysis confirmed the presence of the gene.

Alternaria blight caused by *Alternaria brassicae*, one of the most serious and wide spread diseases of Brassica, causes poor yield of the crop accounting for about 57% yield loss. Potential of regulatory genes for developing fungi- and bacteria-resistant transgenic plants has been demonstrated in Arabidopsis and other plants, one such regulatory gene being NPR1. In the study " Development of *Brassica juncea* Transgenic Plants With Defense Regulatory Gene NPR1 For Improving Alternaria Resistance," *Brassica juncea* NPR1 (BjNPR1) gene, cloned in pTriplex was over-expressed in *Brassica juncea* (Fig. 8). Nine out of ten plants analyzed were found to be PCR positive. Moreover, overexpression of the BjNPR1 transcript was shown in 6 out of the total 10 plants analyzed by RT-PCR using NPR1 primers. Beyond this, NPR1 was found to be induced 3-fold by 12 hrs SA-treatment of *B. juncea* plants as compared to control (H_2O-treated) or 12 hrs JA-treated plants.

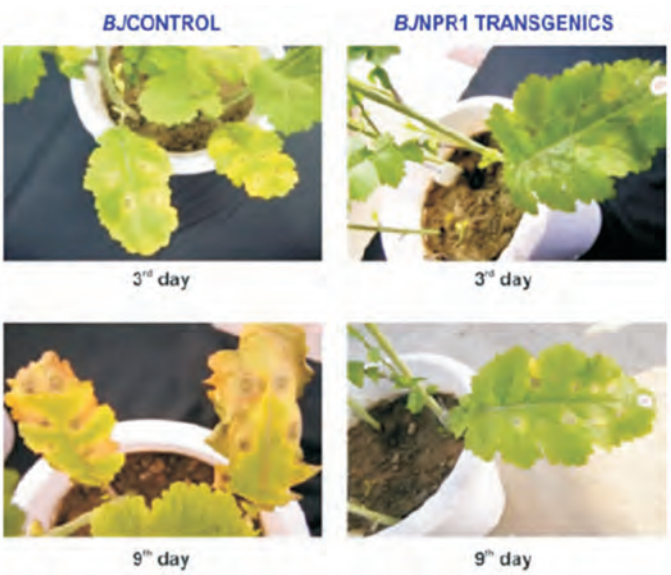


Fig 8. *Brassicajuncea* **non- transgenic (control) and transgenic (with NPR1) plants after infection with** *Alternariabrassicae* **infection**

Drought resistance transgenic wheat was developed by over-expressing ascorbate peroxidase. A thylakoid bound ascorbate peroxidase (TatAPX) cDNA was cloned from a hardy wheat cv. C306 (Accession number EF184291). A total of six gene constructs with three different cDNAs including TatAPX, driven by two different stress inducible promoters were developed. These gene constructs along with pDM803, which contains the selection marker gene BAR, were co-bombarded to obtain marker free transgenic wheat. Three different genotypes PBW343, HDR77 and C306 were used as explants. Four plants were found PCR positive for the gene of interest and selection marker gene BAR. Among these T1 transgenic lines HRCB3#17-37 and PRCB3#22-25 were marker free and showed good level of expression of the gene of interest AtCBF3. The leaf RWC of the T1 transgenics was of 12-15% higher than that of the wild type plants during stress. A yield advantage ranging from 8% to 27% under water deficit conditions was observed in T1 generation of transgenics over the wild type plants.

Transcription factors (TFs) are the major regulators of transcription machinery and are implicated in abiotic stress response. The role of two such TFs encoding genes *BcZF2* and *ZmNF-YB2* were characterized by *Agrobacterium* mediated genetic transformation of *indica* rice *cv*. IR64. Single locus transformants were obtained which segregated in 3:1 ratio. Drought tolerance analysis of *BcZF2* and *ZmNF-YB2* transformants (T_1) conducted by pot culture experiments using physiological parameters such as relative water content (RWC) and membrane stability index (MSI) indicated an enhanced level of drought tolerance in transgenics as compared with the wild type. After 16 days of water-deficit stress, the RWC of wild type plants was around 56% whereas the RWC of transgenic plants was significantly higher with an average of about 81% in *BcZF2* and *ZmNF-YB2* transgenic plants. After 20 days of water-deficit stress, the RWC of wild type plants decreased further from 56% to 44%, whereas the RWC of transgenic plants decreased only marginally to 79%. The transgenic plants thus showed better drought tolerance than the wild type plants confirming the role of *BcZF2* and *ZmNF-YB2* in imparting tolerance to water deficit stress.

A transgenic approach to increase seed/oil yield was explored under the research programme 'Effect of altered expression of key genes involved in fatty acid biosynthesis and seed development on productivity of Indian mustard (*Brassica juncea*)'. Effect of overexpression of key genes involved in oil biosynthesis (DGAT and G3PDH), and seed development (*KLUH* and *SHB1*) on productivity of *B. juncea* was investigated. Gene cassettes for overexpression of these genes under either seed specific or constitutive promoter were synthesized and mobilized into mustard through *Agrobacterium*-mediated transformation (Fig. 9). A total 37 transgenic events were generated for the four gene cassettes. Molecular analysis of transgenics by PCR, RT-PCR, qRT-PCR, and Southern blotting confirmed that the transgenes were stably integrated into the host genome, and were expressed in target tissues (Fig 10). Analyses of seed weight, oil content and other yield traits in T_1 transgenics showed that seed specific overexpression of *G3PDH* and *DGAT* improved the oil content and seed weight significantly. The oil content of some G3PDH transgenics increased by 22.6% whereas one DGAT transgenic gave up to

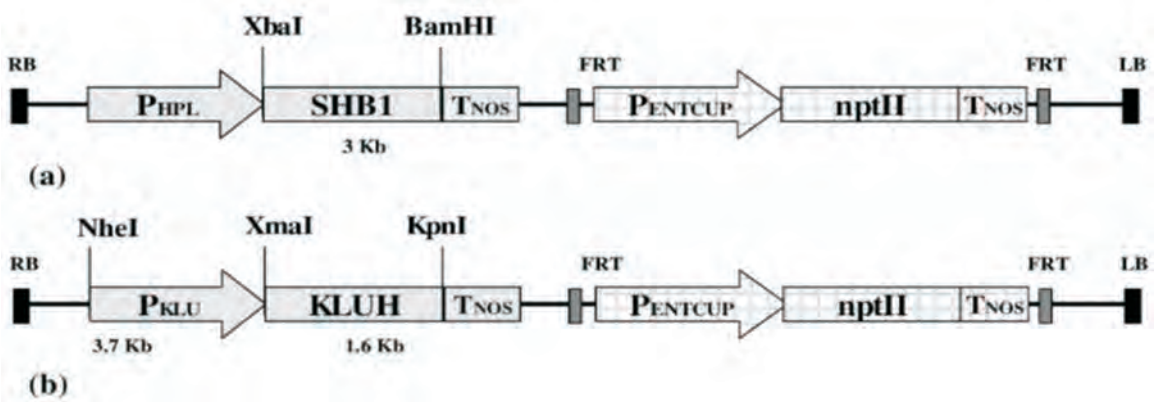


Fig 9. Plant transformation vector constructs

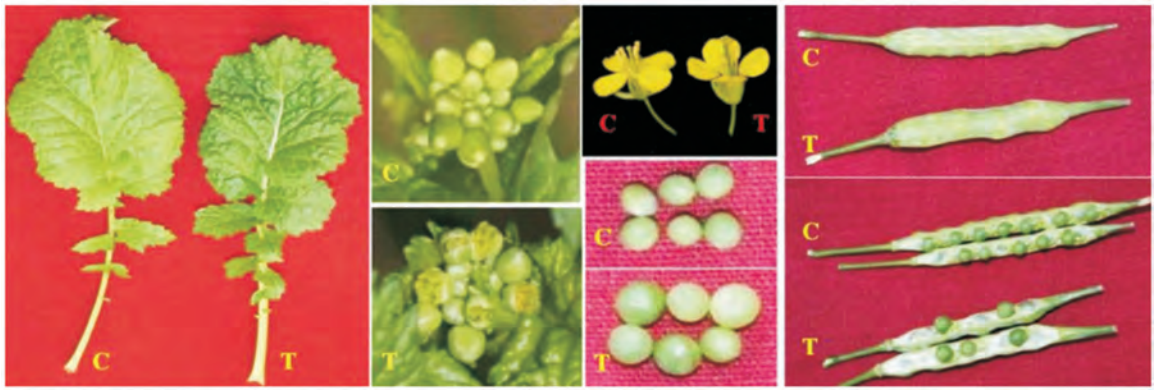


Fig 10. Phenotype of SHB1-2 (T) and the control RLM 198 (C).

67.6% higher seed oil content compared to the untransformed control plants. Likewise analyses of yield traits in KLUH and SHB1 transgenics showed that overexpression of *KLUH* and *SHB1* increased seed weight in mustard. However, the increase in seed weight of KLUH transgenics was not consistent. In case of SHB1 transgenics seed weight increase ranged from 30-40%. In SHB1-2 event seed size was increased significantly compared to control plants. However, SHB1-2 plants showed pleiotropic effects such as crinkled leaves, delayed flowering, mismatch in the lengths of stamen and pistil which resulted in reduced pollination and seed set. Thus the present study showed that careful choice of genes governing seed development and seed oil biosynthesis, the transgenic approach has the potential to break the current yield barriers.

Isolation and characterization of novel plant promoters

Promoters are the DNA sequence located upstream of the coding sequence of a gene. For directing desired expression of the transgene use of appropriate promoter is pivotal. Therefore, it is necessary to isolate and characterize the promoters that can be effectively used in driving the expression of foreign gene in a spatial-temporal pattern.

Two novel promoters, *MYB02* and *MYB04* were cloned from drought tolerant rice cultivar Nagina-22. Both *MYB02* and *MYB04* promoters showed abiotic stress-responsive induction in transgenic *Arabidopsis*. *MYB04* is induced by only drought stress, while *MYB02* promoter showed significant up-regulation under drought, osmotic (mannitol), salt and cold stresses. Both *MYB02* and *MYB04* promoters are induced only in aerial parts of plants but not in roots. Deletion analysis led to the identification of *MYB02* promoter regions responsible for cold, drought and salt stress regulated expression of *MYB02.*

Another novel promoter of the gene *Lateral Organ Junction* gene (*LOJ*) was cloned from *Arabidopsis thaliana* (Fig. 11). Detailed analysis of *LOJ* promoter led to the

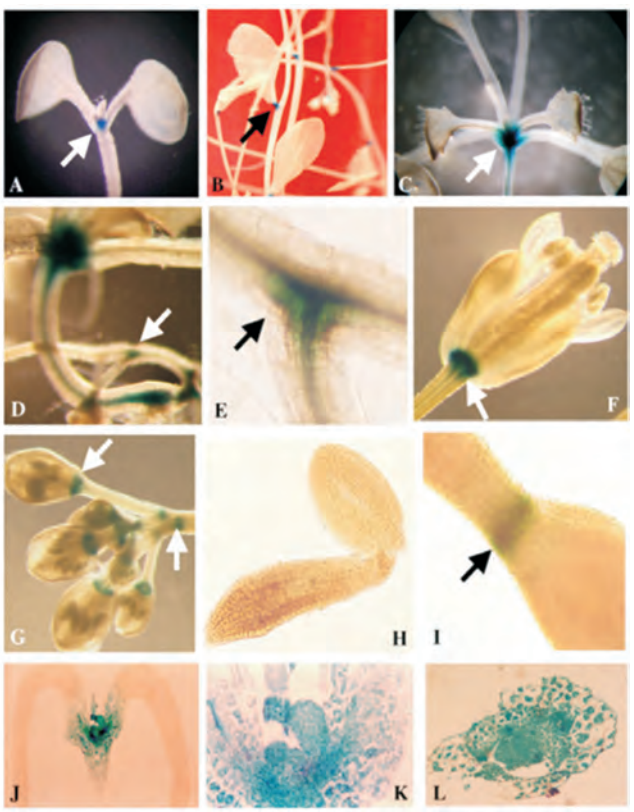


Fig 11. GUS expression pattern detected in lateral organ junction (LOJ) in Arabidopsis promoter trap line 149.

identification of two distinct regulatory domains with spatial and temporal expression patterns. Deletion analysis revealed that the distal regulatory element of the *LOJ* promoter consists of an enhancer element conferring lateral organ junction specificity, while the proximal promoter region consists of regulatory elements specific for expression in anthers and developing seeds of *Arabidopsis thaliana*. The 5' UTR region of the *LOJ* promoter is required for conferring anther and developing seed specific expression. The promoter element also retains its lateral organ junction specificity in a heterologous *Brassica juncea* system.

A Cryptic Root-Specific Promoter (CRS) was isolated from *Arabidopsis thaliana. In silico* characterization of the promoter using various softwares revealed the presence of features typical of the eukaryotic promoter architecture, certain root-specific motifs and also motifs involved in spatial expression and environmental-signal inducibility (Fig. 12). Deletion analysis revealed a 376 bp promoter fragment upstream to the experimental TSS of *SAHH1* gene could confer ovule/ seed-specific expression. The promoter was functional in heterologous system of *Brassica juncea.*

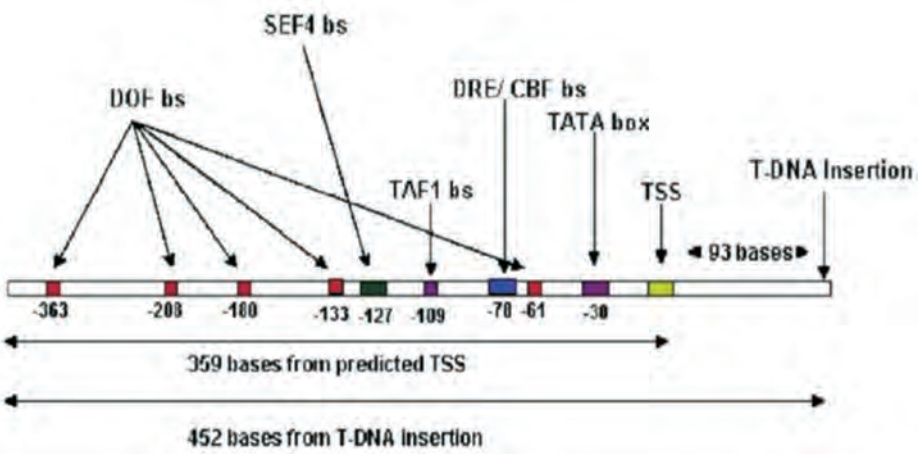


Fig 12. Schematic diagram depicting the structure of the 452 bp fragment upstream of the T-DNA insertion showing different cis-acting regulatory elements

Promoters of the *Arabidopsis thaliana* nuclear gene *Rps14* and *Oryza sativa* nuclear gene *Rps11* coding for mitochondrial ribosomal proteins were analyzed to delineate the promoter region and to assess the promoter strength, respectively. Using transgenic approach with GUS as reporter gene it was shown that promoter region of *Rps14* gene is located between -194 bp upstream of the translation start codon and 347 bp of the coding region. Further, deletion of the coding region nearly abolished GUS expression. Thus *Rps14* gene appears to be unique in having important regulatory sequences embedded in the coding region. Semi-quantitative RT-PCR revealed that *Rps11* gene of rice is driven by a strong, constitutive promoter whose strength is comparable to promoter of *tubulin1* gene. Thus *Rps11* promoter may provide a highly constitutive promoter for transgene expression in monocots.

For combating pathogen attack the wound-inducible promoter can be used to drive expression of transgenes that can provide effective resistance against the invading pathogen. A novel wound responsive promoter was identified by screening T-DNA insertion lines of *A. thalina.* Histochemical and fluorometric analyses of *GUS* activity revealed that the promoter is induced by wounding in internodal region of stem epidermis. Further study identified the promoter of the fatty acyl-*CoA* reductase 6 (*FAR6*) gene. The spatial and temporal regulation of gene expression pattern driven by *FAR6* promoter suggest that the corresponding *FAR6* gene (AT3G56700) may be involved in the defense response of wounding or pathogens that damage the plant mainly through the epidermis (Fig 13). This work provides a promoter element that could be exploited for the genetic engineering of the crop plants.

Similarly, in Chickpea attempts were made to identify novel wound-responsive promoter. The promoters sequence was identified on the basis of homology search with *Arabidopsis thaliana*. TAIR 9 database was used for the identification of genes induced in response to wounding in *Arabidopsis*. Out of the 42 loci matches those genes that were either hypothetical or translated to unknown proteins, were studied for their expression level at 5 min post wounding. Five genes that showed maximum upregulation were verified for rapid wound response using qRT-PCR. The five genes were AT1G72920, AT2G20340, AT2G34810, AT5G47240, and AT5G54170.

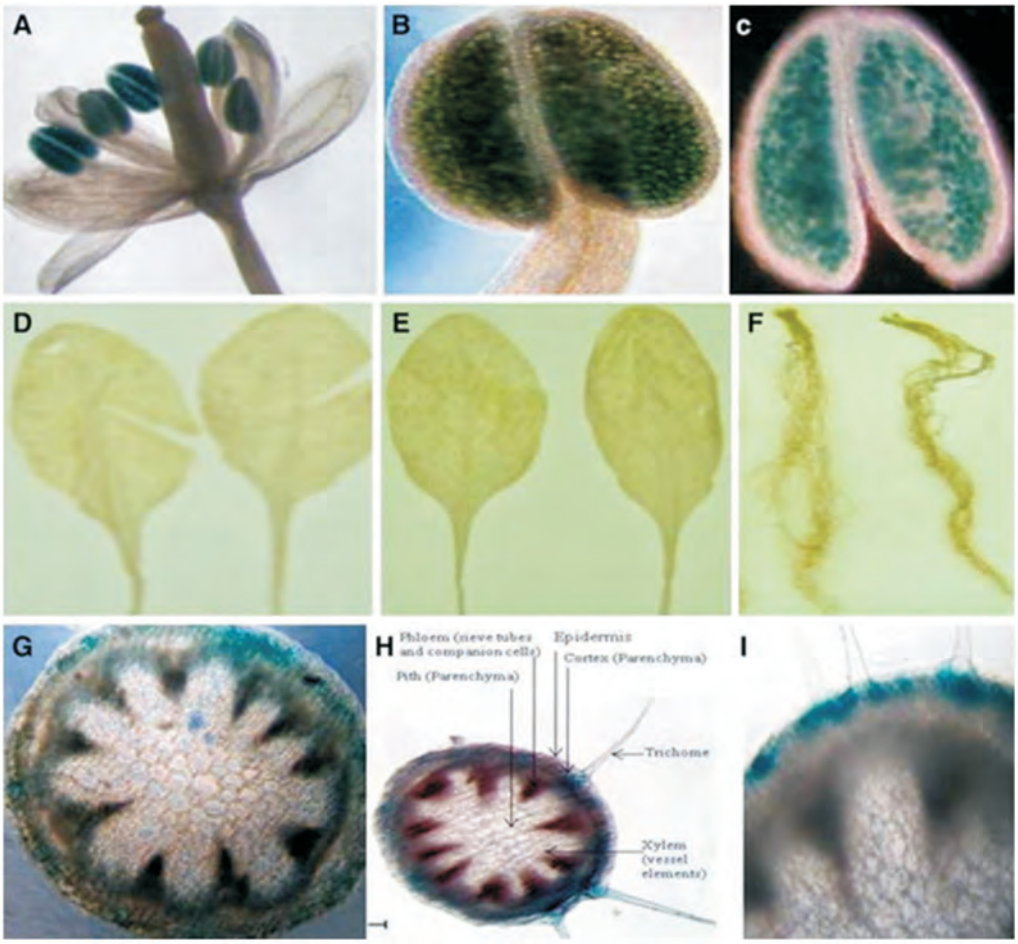


Fig 13. Histochemical analysis of *GUS* expression in different tissues of the mutant line T90.

Out of several wound responsive genes, a START domain containing protein (AT5G54170) was selected, and its corresponding homologue was identified in chickpea, LOC101512743. This homologue in chickpea was also found to respond rapidly to wounding. Further in silico analysis identified an additional TM-START gene, contig2750, along with the chickpea gene contig5716 (LOC101512743) (Fig. 14). Studying these genes in floral development can help us in gathering deep insight into their function in flowering.

Promoters that respond to pathogen attack are vital for driving R genes to elicit resistant reaction from the host. From a set of differentially expressed genes in indica rice cultivar, HR 12 which is susceptible to *Magnaporthe oryzae* a gene CYP76M7 was found to be consistently up regulated from 24 hpi (hours post inoculation) after challenge by *M. oryzae*. The promoter sequence of this gene spanning 520 to 1166 was found to confer *Magnaporthe* inducibility to CYP76M7 promoter. This promoter can be used to develop two component system for development of rice lines with durable blast resistance to *M. oryzae* (Fig 15).

In a similar study, Arabidopsis, *PDF1.2b* (*At2g26020*) was found to be transcriptionally activated by *Alternaria* infection. A systematic deletion analysis of this promoter region was done using *uidA* reporter gene. Histochemical GUS analysis of transgenic Arabidopsis plants carrying various deletion fragments revealed that 660 bp fragment upstream to the coding region was adequate for full expression of *uidA* gene in response to *Alternaria*, jasmonic acid, salicylic acid and wounding. The identified promoter can be deployed to drive R genes for developing Alternaria leaf spot resistant mustard varieties.

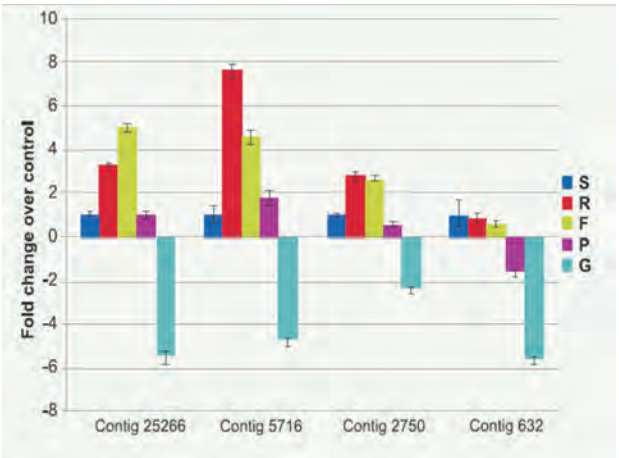


Fig. 14. qPCR analysis of TM-START proteins of chickpea genes done in different tissues. Relative fold change values are given with respect to GADPH2 was used as reference genes. S-Shoot, R-Root, F-Flower, P-Young pod and G-GrainThe expression inshoot was taken as 1 and compared to other tissues.

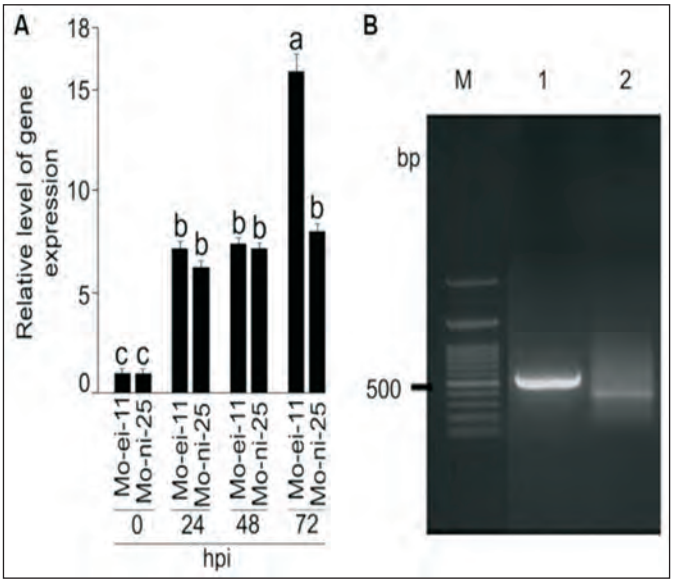


Fig 15. Expression analysis CYP76M7 transcripts and mapping of its Transcription Start Site (TSS).

Expanding Genomic and proteomic resources in crop plants

Omic resources are the backbone of modern plant breeding. Generation of information on genome, transcriptome, proteome and metabolome of major crop plants of India is important for broadening the base of the future crop improvement programs. These resources can be effectively utilised to understand the regulatory network underlying the complex traits that will not only provide a significant insight in plant growth and development but will also be pivotal in designing new methods of crop improvement.

The size and complexity of the genome can be determined from the kinetics of reassociation of its DNA. In pigeon pea based on renaturation kinetics, genomic DNA was classified into highly repetitive (*Cot* 1), moderately repetitive (*Cot* 10) and single/ low-copy (*Cot* 100) sequences that were subsequently cloned to produce *Cot* 1, *Cot* 10 and *Cot* 100 libraries. Homology searches against GenBank nr and EST databases confirmed that *Cot* 1 library was enriched in high-copy number (transposons, rDNA and other dispersed repeats) and the *Cot* 100 library was enriched in low-copy sequences. *Cot* 10 libraries, was found to represent both low copy and high copy DNA sequences.

b) Identification of microRNAs in chickpea

MicroRNAs have emerged as the major regulators of various processes in eukaryotes. Therefore, development of miRNA resources and identifying their target genes is likely to resolve the mechanism of gene regulation in many instances. In chickpea using Illumina Genome analyser microRNAs were analysed and 368 miRNAs were identified. In studying their tissue specific distribution of expression total 24 families consisting of 97 conserved miRNAs and 73 novel miRNAs were identified in root and leaf tissues after extensive sequence analysis. Likewise, 25 families consisting of 112 conserved miRNAs and 86 novel miRNAs were identified in flower tissues of desi and kabuli type. The predicted target genes of all the identified miRNAs were extensively involved in different biological processes involving a large number of gene families (Fig 16).

In chick pea Fusarium wilt cause by fungus *Fusarium oxysporum*f.sp. *ciceris* is a major problem. Possible role of four miRNA viz. miR2118, miR530, miR390 and miR166 in defense against *F. oxysporum,* were studied in

miRNA family	Target	Putative Functions of Predicted Targets
Conserved miRNAs		
mir156_1	TC12891, TC03863, TC05745,TC07318,	Squamosa promoter-binding TF family protein, SCP1-like
	TC03684, TC29077, TC07318, TC15422,	small phosphatase
	TC04572	
mir156_10	TC29077, TC15422, TC12891, TC03863,	SCP1-like small phosphatase, squamosa promoter-
	TC05745, TC03684, TC07318, TC19303,	binding protein, cationic amino acid transporter,
	TC10437, TC05493, TC18211	allantoinase 1-like protein
mir166_1	TC04758, TC15765, TC08004	ClassIII HD-ZIP, REVOLUTA
mir167_4	TC21867, TC03743, TC03697	Monosaccharide transport protein 1, MFS, tubulin-folding
		cofactor E
mir168_1	TC06138, TC16221, TC07642	GTP-binding protein, RNA binding (RRM/RBD/RNP motifs)
mir171_1	TC15816, TC01767, TC07982	HAIRY MERISTEM 3 (HAM3), cdk protein kinase, ClpX3
mir319_6	TC03909	Putative xylogalacturonanxylosyltransferase
mir390_1	TC12049, TC05305, TC19589	Protein kinase, CZF1
mir396_3	TC18749, TC21342, TC02165, TC16760,	RNA-directed DNA polymerase, NAD(P)-binding
	TC09727, TC02085	Rossmann-fold
mir530_1	TC01544, TC20787, TC01795, TC01794	Zinc knuckle protein, expressed protein
mir2118_1	TC01089, TC09480, TC00082, TC21040,	NB-ARC disease resistance protein, expressed protein,
	TC23505	TIR-NBS-LRR
Novel chickpea miRNAs		
car-miRNA008	TC06967, TC05545	RING/U-box superfamily protein, chalcone synthase (CHS)
car-miRNA011	TC02274, TC14659, TC17732, TC08052,	SERPIN family protein, amelogenin, RNA binding
	TC16830, TC06852, TC05883	(RRM/RBD/RNP motifs), LEA, anion channel protein family
car-miRNA015	TC17182, TC10107	Complex 1 protein (LYR family), ribosomal L23/L15e
		family protein
car-miRNA020	TC33381, TC29465, TC00653, TC28744,	TPR-like superfamily protein, ARM superfamily protein,
	TC05383	FAD/NAD(P)-binding oxidoreductase, Protein of unknown
		function (DUF1423)
car-miRNA051	TC11550, TC31151, TC21283	SMG7, HAD superfamily protein, unique electron
		transfer flavoprotein

Fig. 16. Predicted target genes of miRNAs in chickpea.

wilt-resistant (WR 315, Pusa Green 112) and wilt-susceptible (BG 256, FLIP 90-166) genotypes in response to wilt stress. Interestingly, TIR-NBS-LRR disease resistance gene predicted to be targeted by miR2118 was significantly downregulated in wilt-susceptible genotypes in response to wilt stress, suggesting the role of miR2118 in development of susceptibility in genotypes of chickpea. The finding of study provides a significant lead in understanding the role of miRNAs in imparting disease resistance and susceptibility in plants.

From the small RNA libraries generated from deep sequencing of CMS lines and restorer lines of pigeon pea, a total of 260 known miRNAs were identified using the latest version of miRBase. It was found that conserved miRNAs like miR156, miR166 and miR164 were differentially express in bud samples of CMS and restorer line which targets transcriptional factors squamosa promoter binding protein (SBP-box), Class III HD-Zip and NAC transcriptional factors respectively. Another miRNA which was only reported in pulses, miR1510 was found differentially expressed in CMS and restorer bud tissues. This preliminary analysis provides a basis for further investigations on the role of miRNAs in pollen sterility and fertility in pigeonpea.

miRNAs are now emerging as a key player involved in many stress tolerance mechanism in plants. In a study, Nagina22, a drought tolerant popular cultivar and IR64, a drought susceptible mega variety have been used as a model genomic background to unravel the mechanisms of drought tolerance in rice. It was found that 2173 candidate genes expressed exclusively at flowering stage in Nagina 22 under moisture deficit stress. Additionally, 39 miRNAs belonging to 31 families, showing differential expression pattern across the parents and bulks were identified. Six miRNA-target gene pairs from four miRNAs (osa-miR156k, osa-miR396a-3p, osa-miR3979-3p and osa-miR399j) were validated using the genome-wide microarray expression data. Osa-miR3979 was identified as a novel moisture deficit stress responsive miRNA at flowering stage. The superiority of Nagina22, in terms of spikelet fertility under drought stress, was found to be due to its superior alleles for the antioxidant enzymes, namely, superoxide dismutase, SOD (*SOD2, SODCC1, SODA*) and glutathione reductase, GR (*GRCP2*) rather than ascorbate peroxidase, for which IR64 had the superior allele (*APX8*). Additionally, a

high density integrated physical map was made using differentially expressed genes, miRNAs and QTLs specific to Nagina 22 at reproductive stage under drought which could be an excellent resource for the researchers involved in improving drought tolerance in rice.

For understanding the molecular mechanisms involved in fruit development of Brinjal (*Solanum Melongena* L) genome-wide transcriptome and proteome analysis was carried out during different stages of fruit development in brinjal cultivars Pusa Purple Long (PPL) and Killikulam (KKM-1).

In transcriptome analysis, 1395 and 1174 transcripts showed differential expression in brinjal cultivars PPL and KKM-1 at various stages of fruit development, respectively. Microarray analysis showed that genes involved in cell cycle, cell growth, cell wall biosynthesis like *CycB1*, *CycB2*, *CycD3*, tubulin, kinesin, XTR, FLA and EXGT and brasinosteroid synthesis (*HYD1*, *DWF1*) were up-regulated during early fruit development stages. The genes involved in photosynthesis, carbon and energy metabolism and redox responsive genes were up-regulated throughout the fruit development stages. Further, genes involved in ethylene biosynthesis were up-regulated at 20 dpa. Many antioxidant proteins like ascorbate peroxidase, superoxide dismutase and proteins related to glycolytic pathway were up-regulated during fruit development and ripening.

Comparative transcriptome analysis was done between Bt and non-Bt brinjal plants. Transcriptomics of Bt brinjal exhibited very little difference compared with its near isogenic non-Bt variety. 12 genes in fruit tissue and 61 genes in leaf tissue were differentially expressed in Bt brinjal compared with non-Bt variety. The microarray data was validated by quantitative Real-Time PCR analysis. Additionally, in another study a comparative proteome analysis was done between Bt and non-Bt brinjal plants. Proteomics of Bt brinjal exhibited very little difference compared with its near isogenic non-Bt line. Seven proteins in fruit tissue and ten proteins in leaf tissue were differentially expressed in Bt brinjal compared with non-Bt counterpart. These differentially expressed proteins were identified by using MALDI-TOF mass spectrometry. Most of the differentially expressed proteins were related to photosynthetic function. The overall result suggest that the expression of the insect resistance protein does not significantly alter the brinjal fruit and leaf proteomic profiles, contributing to assess the bio safety of insect resistance plant expressing *Bt* protein. Empirical evidence of not having any significant unintended effect of transgenesis will help in advocating public acceptance of the GMO product.

Transcript profiling using the Affymetrix 57 K Rice GeneChip revealed a total of 72 differentially expressed genes at initial interaction stage of 6 hours post inoculation (hpi) with *M. oryzae* isolate Mo-si-63 which were significantly altered after pathogen infection. Majority of the genes of protein degradation and modification, transport, signalling and transcription factors were repressed at the initial stages of interaction whereas hormonal signalling, cell wall defense and transcription factors belonging to the WRKY and Trihelix family genes were upregulated. This comprehensive analysis has identified an inventory of genes with altered expression regulated by *M. oryzae* infection. The data extend the current understanding of rice defense mechanism during compatible interaction with pathogen and serve as a genomic resource for robustly selected, differentially expressed genes for further investigation.

Finger millet (*Eleucine coracana*) or ragi, is a hardy cereal known for its high level of tolerance against drought, salinity and various biotic stresses. For identification of differentially expressed genes under drought stress a cDNA macroarray with 1000 clones was prepared and few drought responsive genes were identified. A drought responsive gene EcDehydrin7 containing dehydrin domain was functionally validated by generating transgenic tobacco plants using *Agrobacterium* mediated plant transformation. Physiological and molecular analysis showed that over-expression of EcDehydrin7 confers drought tolerance in transgenic tobacco (Fig. 17). Generated genomic data and characterized gene information from ragi will enrich the existing meagre genomic resources of this crop.

High temperature stress has profound adverse effects on plant growth and yield. Since the available gene pool is a limiting factor in many crops, it becomes imperative to search for an arsenal of new high temperature tolerant genes from exotic and wild species which can then be transgenically integrated into food crops. Pearl millet (cv. 841 A) has a natural ability to withstand high levels of abiotic stresses. To elucidate the molecular

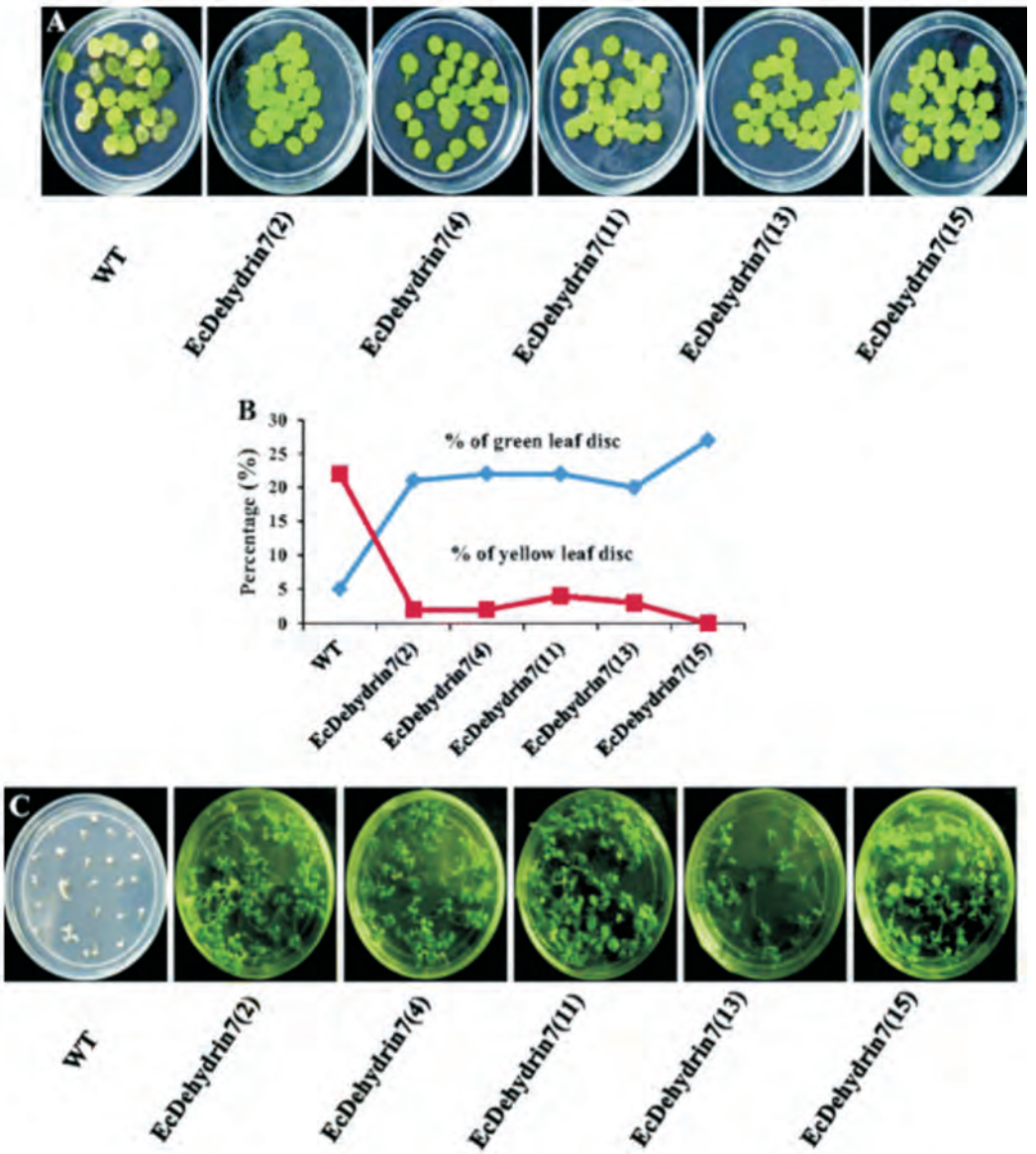


Fig. 17. Physiological analysis of transgenic plants. a: Leaf disc senescence assay of WT and T_0transgenic plants in 200 mM mannitol. b: Bars showing percentage of response to 200 mM mannitol. c: WT and T1 transgenic seed germination on MS media containing 200 mM mannitol and their phenotyping after 20 days

basis of high temperature response, 331 high quality stress responsive ESTs were identified in pearl millet. Similarly, in *Prosopis cineraria* 745 EST clones were obtained under heat stress and 1152 EST clones were identified under drought stress condition. The drought stress induced EST data reported were the first available data for *P. cineraria* and further studies on functional analysis and characterization of unknown genes obtained in the present EST data shall be useful for developing stress tolerant transgenic crops (Fig 18). Also, several ESTs responsive to salt stress were identified in wheat cultivar, Kharchia local.

Photosynthetic fixation of CO2 is more efficient in C4 than in C3 plants. Rice is a C3 plant and a potential target for C4 pathway engineering. It is known that genes encoding C4 specific enzymes are present in C3 plants. *In silico* analysis revealed that 37 members of the five C4 gene families are present in rice. For

detecting the expression of these genes in rice expression level of different C4 specific gene family members [12 of carbonic anhydrase (*ca*), 7 of phosphoenolpyruvate carboxylase (*pepc*), 10 of malate dehydrogenase (*mdh*), 4 of NADP-dependent malic enzyme (*nadp-me*), 2 of NAD-dependent malic enzyme (*nad-me*), and 2 of pyruvate orthophosphate dikinase (*ppdk*)] was quantitatively analysed using real time PCR. All the 37 members belonging to different gene families showed expression and the levels varied among the rice genotypes tested. In another study, the expression of the C4 pathway genes was studied in seven different rice genotypes including three pure line varieties namely, Pusa Basmati-1 (PB-1), Pusa Basmati-6 (PB-6) and Pusa Basmati 1121 (PB1121), and four hybrids, viz. PA6444, PA6129, KRH2, and Pusa Rice Hybrid-10 (PRH-10) during grain filling stage. A short duration high yielding hybrid PA6129 supported C4 gene expression during senescence stage. Correlation analysis revealed a strong relationship between *ppdk* and *ppdk-rp* gene expression in rice leaves during grain filling stage. This study will further facilitate our understanding of the regulation of C4 specific photosynthetic genes in C3 crop plants.

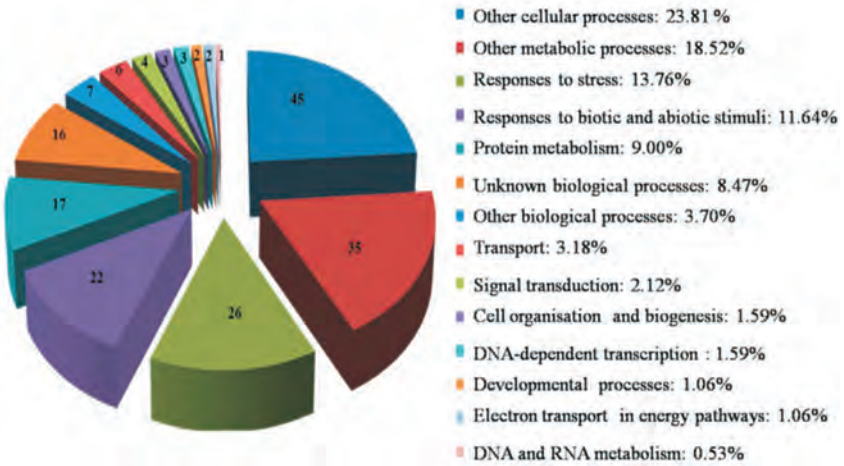


Fig. 18. Fourteen functional genetic ontogeny (GO) categories for genes identified in the heat-stress EST library of P. cineraria annotated using the MIPS functional catalogue in BLASTX against the TAIR database.

The latest tomato genome draft sequence version, release 2.40, was used to analyze anthocyanin genes, annotate gene structures and to determine their exact physical locations in different chromosomes. The position of 5' UTR, 3' UTR, length of ORF and upstream basal promoters were detected for seven anthocyanin pathway genes viz. *CHS, CHI, F3H, F3'5'H, DFR, ANS,* and *UF3GT*. The genes *F3H, DFR, ANS,* and *UF3GT* were of single copy each whereas *CHS, CHI,* and *F3'5'H* were having two copies each. Physically the genes *CHI, F3H, F3'5'H, DFR, ANS,* and *UF3GT* were located in chromosomes 5, 2, 11, 2, 8 and 10, respectively. The two genes *CHS1* and *CHS2* have been identified in chromosomes 5 and 8 respectively. The 1kb upstream region was analyzed by PLACE database to find cis regulatory elements (CRE) motifs. Out of 145 types of CREs found in the seven genes, 93 have been annotated to involve in plant stress responses and 16 in binding sites for MYB and bHLH transcription factors regulating the expression of seven genes for anthocyanin production. RT- PCR and RT-qPCR analysis showed that CHI, DFR and ANS might be the rate limiting enzymes for non-production of anthocyanin in the cultivated tomatoes.

Novel approaches and proof of concepts for developing biological resources

Activation tagging is used to recover and clone dominant gain-of-function alleles and, usually employs a T-DNA vector containing four tandem copies of the CaMV35S enhancer sequence outward, usually at the right border. Constitutive (CaMV35S), inducible (estradiol-inducible XVE) and tissue specific (*TFL1* and *rbcS2B*) promoters have been tested for activation tagging. The exact spatial and temporal expression pattern regulated by *TFL1* and *rbcS2B* promoters were characterized using GUS reporter gene via transgenic approach. *TFL1* promoter drives gene expression mainly in root and inflorescence tissues while *rbcS2B* promoter is green tissue specific. Comparison of promoter strength of cloned *rbcS2B* upstream sequences with the native *rbcS2B* promoter through qRT-PCR revealed thousands fold stronger promoter activity of cloned *rbcS2B* promoter, particularly in leaf. From an initial screening of 500 T1 plants, 33 morphological variants were identified which included seedling mutants (single cotyledon, slow seedling growth, long root, short root), leaf mutants (fused leaves,

excess rosettes, altered shape, hyponastic leaves, bushy rosette, altered leaf polarity), inflorescence mutants (fasciated, inflorescence fused with the petiole of rosette leaf) and flowering time and growth habit mutants (dwarf, tall, miniature, late flowering, sturdy stem). Inducible promoter activation tagged population was screened for salt tolerance, and two mutants were identified in the T2 generation which appeared to be due to T-DNA insertion rather than activation tagging. Analysis of T-DNA integration site in subset of mutants suggested that insertion is more frequent in the intergenic region. Detailed analysis of 35S-84 mutant showed *At5Tase8* gene was upregulated in the mutant leading to longer primary root phenotype. Examination of overexpression line of *At5Tase8* gene revealed that *At5Tase8* gene is indeed involved in modulation of root system architecture.

Cytoplasmic male sterility (CMS) systems involving male sterile, fertility restorer and maintainer stocks form the foundation of hybrid technology in several field crops such as rice maize, sunflower, pearl millet, sorghum etc. Genetic determinants of CMS are present in the mitochondrial genome whereas fertility restorer genes are found in the nuclear genome. Thus, apart from its practical utility in hybrid seed production, CMS serves as an excellent model to study nuclear-mitochondrial gene interactions. In this study, CMSlines of *B. juncea* which are restored by a single fertility restorer line carrying introgression from *Moricandia arvensis* were examined for the mitochondrial genome organization and expression. It was found that in CMS line carrying *Diplotaxis berthautii* mitochondria also, altered expression of *atp*α gene was associated with male sterility (Fig 19). Cloning and comparative analysis of the *atp*α gene and transcripts of the CMS, fertility restored and euplasmic lines revealed some interesting features. A novel *orf108* was found to be associated with male sterility in these CMS lines. The role of *orf108* in causing male sterility was tested in *Arabidopsis thaliana* through transgenic approach. Expression of *orf108* with *A. thaliana* peroxidase promoter induced male sterility (Fig. 20). ORF108 was capable of inducing male sterility when targeted to the mitochondria or present in the cytosol. Thus, *orf108* may be acting gametophytically. This study has identified a common molecular mechanism underlying male sterility and fertility restoration in different CMS lines of Indian mustard.

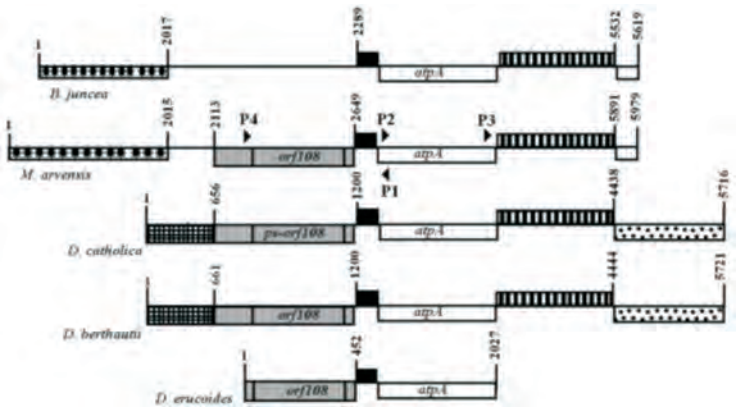


Fig. 19. Comparison of mitochondrial genome sequences encompassing the mitochondrial atpA gene of euplasmic Brassica juncea and CMS Moricandia arvensis, Diplotaxis catholica, *D. berthautii*, and *D. erucoides*.

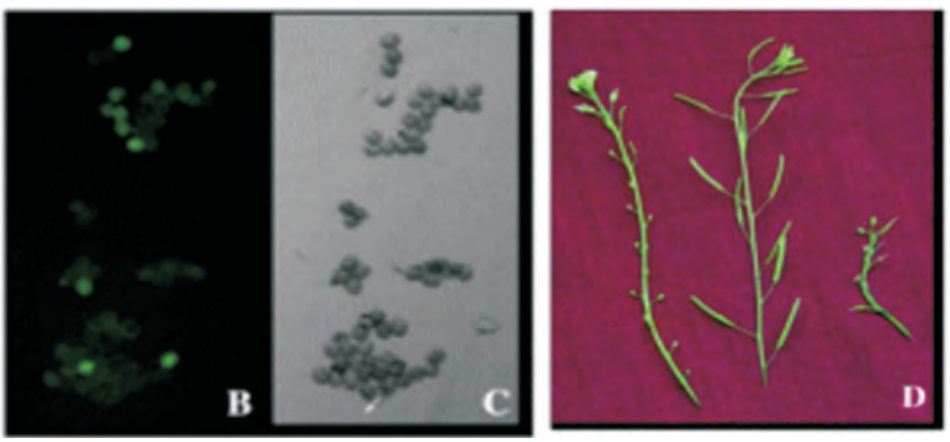


Fig 20.Transgenic expression of orf108 in *Arabidopsisthaliana*

During the course of this investigation, an important discovery was made that mitochondria are biparentally inherited in *B. juncea*. Detailed analysis of progenies of crosses involving different male sterile, fertility restored and normal euplasmic lines confirmed the presence of paternal specific mitochondrial DNA sequences in the F1 progenies. The mitochondrial genome was found to be invariably transmitted from paternal side, albeit at a low level. The paternally inherited mitochondrial genes persisted throughout the life span of the progeny plant and were transcribed. Furthermore, we identified and characterized a rare mitochondrial recombination facilitated by heteroplasmy. Thus this study has for the first time provided unequivocal proof of biparental inheritance of mitochondrial genome in higher plants.

Towards mapping and cloning of male fertility restorergene of CMS (*Moricandia arvensis*) *Brassica juncea*' two pronged approach of molecular mapping and transcriptomics was employed for identification of fertility restorer gene of CMS (*M. arvensis*) *B. juncea*. Towards this end, two BC1F1 populations derived from the cross between CMS and fertility restorer lines were generated. For targeted mapping genome walking was done using sequence information of an earlier identified SCAR3 marker. This suggested that the *Rf* locus is located in the genomics region having synteny with *B. rapa* A09 chromosome. Screening of more than 550 SSR markers designed based on *B. rapa* A09 sequence information identified 12 markers flanking the *Rf* gene and confirmed its location on *B. juncea* A09 linkage group. Two (one EST-SSR and one STS) out of 67 markers designed based on differential gene expression between anther transcriptomes were found to be informative and were linked to the *Rf* gene. The markers were validated in a second BC1F1 population. The closest flanking markers BjESSR01 and BjEST01 were at 0.6 and 1.4 cM from the *Rf* locus. Further, a comparative transcriptome analysis between CMS and fertility restorer line was carried out to identify the candidate fertility restorer gene (Fig 21).

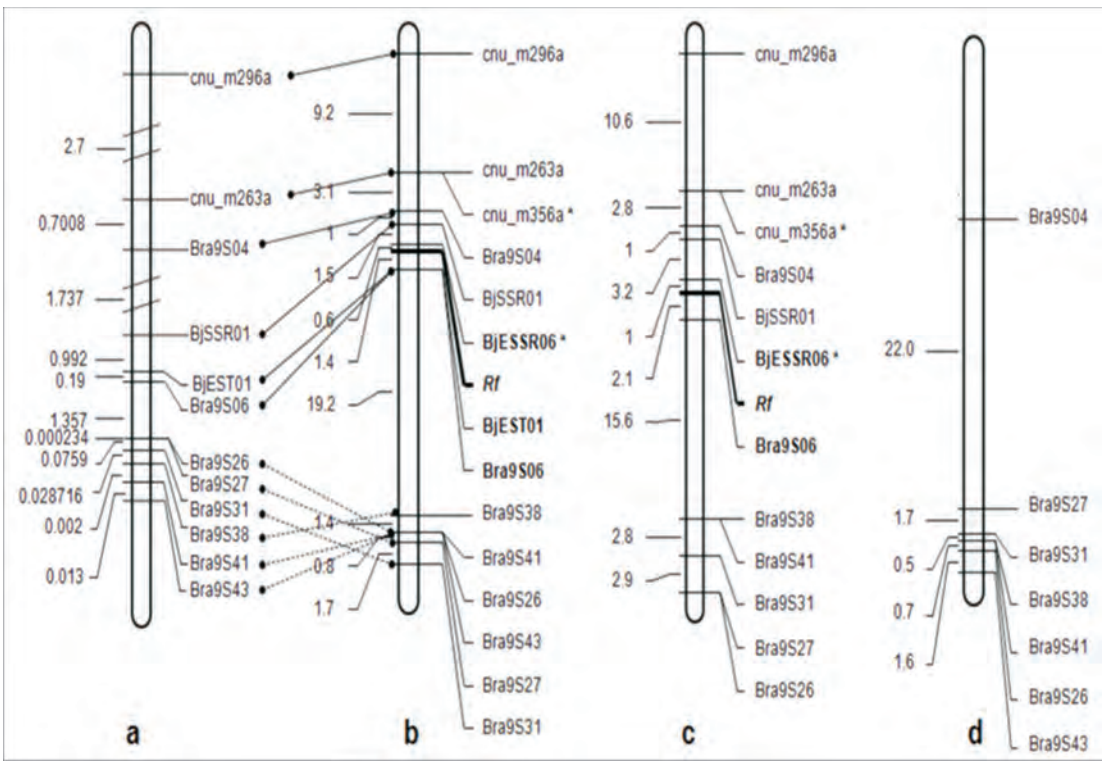


Fig. 21. Molecular linkage map of the *Rf* locus in *B. juncea* and its comparison with the corresponding physical map of *B. rapa*. a Physical map of *B. rapa* chromosome A09 showing location of markers used for tagging of the *Rf* gene. b Genetic map of the *Rf* locus based on Population I. c Genetic map of *Rf* locus based on Population II. d Genetic map of markers in euplasmic *B. juncea*.

Commercial exploitation of heterosis has been possible in rice with the use of wild abortive (WA) cytoplasmic male sterility (CMS). WA cytoplasm based male sterility was studied using rice lines Pusa6A, Pusa6B and Pusa Rice Restorer 78. A longer 3.7 kb transcript in fertile lines Pusa6B, PRR78 and PRH10 but 3.5 kb transcript in the CMS line Pusa6A for the *nadIB* gene. A 1.7 kb transcript for *coxI* gene was absent only in the restorer line PRR78 Sequencing of PCR amplicons for seven of the mt genes revealed 78 SNPs and 28 InDels at an overall frequency of 1/69.7 bp and 1/174.3 bp respectively between Pusa6A and Pusa6B. The frequency was highest in *rps3* gene and lowest in the *atp9* gene. CAPS markers were designed to differentiate the CMS line Pusa6A and its maintainer line Pusa6B in breeding programes. Comparison of DNA sequences of PCR and RT-PCR amplicons detected a total of 38 RNA editing sites, a maximum of 35 of these being present in the mt gene transcripts of the maintainer line Pusa6B, 13 in PRR78 and 11 in Pusa6A. The restorer lines PRR78 differed for four editing events from the CMS line Pusa6A.

Haploids and doubled haploids are invaluable in basic genetic studies and in crop improvement. A study was undertaken to develop haploid inducer (HI) lines of *Brassica juncea* through genetic engineering of the Centromere Histone Protein gene, *CENH3 previously* elaborated in Arabidopsis. Towards this end, the *CENH3* genes and transcripts were cloned from *B. juncea* and five other Brassicaceae species, namely, *B. rapa,B. nigra* and wild allied species *Diplotaxiscatholica*, *D. erucoides* and *Orychophragmus violaceus*. *B. juncea* was found to carry three copies of *CENH3* which gave five different transcripts, of which two resulted from alternative splicing. All other species investigated had one copy of *CENH3*. In *D. erucoides* and *B. rapa*, alternative splicing generated two *CENH3* transcripts, while in all other species a single transcript was found. In *B. juncea, B. rapa*-specific *CENH3* expression level was higher than the *B. nigra* counterpart. For developing HI lines, RNAi approach has been used to knockdown the native *CENH3* genes whereas for rescue of CENH3 silenced cells, a GFP-CENH3-tailswap construct having N terminal GFP fused to H3.3 tail sequences and synthetic CENH3 histone fold domain sequences was devised (Fig. 22). A total 39 transgenic *B. juncea* plants of three kinds of transgenics, namely, those with i) the silencing cassette, ii) the rescue cassette, and iii) both silencing and rescue cassettes were obtained. FACS and cytological analyses of progenies revealed partial or complete elimination of *B. juncea* chromosomes from the co-transformed line thereby giving rise to aneuploids or haploids (Fig. 23). Thus we successfully demonstrated that for the first time that HI lines could be obtained in a polyploid crop species through CENH3 engineering.

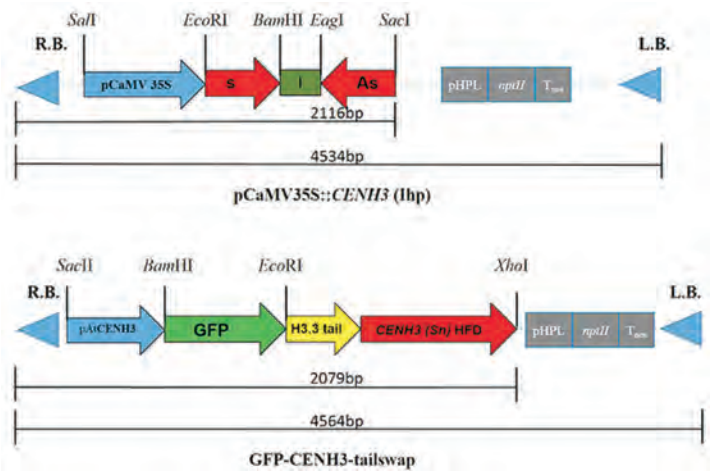


Fig. 22. Schematic representation of T-DNA region of RNAi (pCaMV35S::CENH3) (Ihp) and GFP-synthetic CENH3-tailswap constructs.

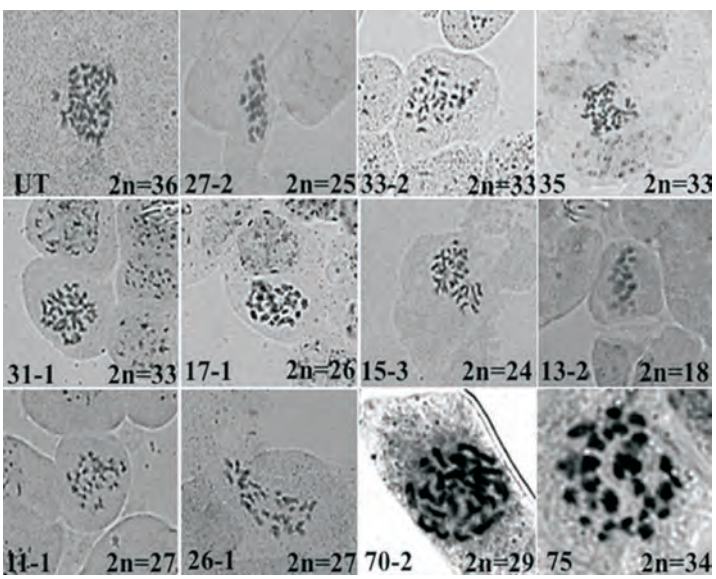


Fig. 23. Cytology of progeny plants derived from the cross between Co-transformed and untransformed or GFP-CENH3 tailswap (event # 38) lines

Competence is one important attribute which is desirable in symbiotic bacteria like Rhizobium. Strains TAL 620, a *Mesorhizobium* strain is proficient in nodule occupancy MC 603 a *comp*⁻mutant was isolated as Tn5 insertional gene mutation. It has a negative comp index of -0.51. The mutation is exclusively due to comp gene inactivation. Because other characters such as growth, nitrogen fixing ability are not affected. Cloning and sequencing of the Tn5 tagged DNA fragment of MC 603, has been identified it as part of the *flh* A gene operon, a gene involved in bacterial flagellar development. The corresponding wild allele of the gene has been isolated from the genomic library of TAL 620. The clone carrying the gene flh A+ is successful in complementing the *comp*⁻mutation of MC 603. Thus it is evident that the mutation of gene flh A+ is responsible for the observed *comp*⁻phenotype of Mc 603.

Productivity gains in several major crops have reached a plateau. Therefore, new approaches to break yield barriers are imperative. A few novel genes have been assessed for their likely potential in improvement of productivity. Low level expression of *ckx3* gene is known to cause cytokinin accumulation in plants' floral meristem thus increasing the number of reproductive organs resulting in enhanced grain yield in rice and mustard. This hypothesis was tested by selecting contrasting mustard varieties Pusa Jaikisan (high yielding) and BEC-144 (low yielding). *Ckx3* gene from *A. thaliana* was cloned for this purpose in pGEMTeasy vector, and bioinformatically analysed with *ckx3* of *Brassica rapa*. Further, showed high yielding Pusa jaikisan has lower *ckx* expression in inflorescences than the stem while inflorescences of low yielding line BEC-144 have higher *ckx* expression in inflorescences. These finding established a negative correlation between yield and *ckx* expression in mustard.

APETALA2 (*AP2*), a member of *AP2*/EREBP class of transcription factors, is a floral homeotic gene and has a broad expression pattern including root, leaf, floral meristem, flower and seed. Mutation or constitutive suppression of *AP2* leads to increase in seed mass but also causes undesirable pleiotropic effects. In order to see whether beneficial effect of *AP2* on seed mass could be used to increase productivity without affecting other traits, *AP2* expression was suppressed in ovules and developing seeds of *Arabidopsis* using tissue-specific promoters. Characterization of these engineered lines suggested that antisense suppression of *AP2* and consequent effect on seed mass could be harnessed for increasing productivity.

Brassinosteroids are essential phytohormones and play key role in plant development. The *DWF4* gene of Arabidopsis codes for an enzyme that mediates multiple 22α-hydroxylation steps in brassinosteroid biosynthesis. *Arabidopsis* plants over-expressing DWF4 protein show increase in inflorescence, number of branches and siliques, thereby give higher seed yield/plant. To test whether overexpression of DWARF4 gene could lead to higher yields in mustard, a 2.9 kb *AtDWF4* gene was PCR amplified, cloned into a binary vector and introduced in mustard cultivar Pusa Jaikisan. Transgenic status was established through PCR and plants were multiplied for further evaluation under contained conditions in phytotron.

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Molecular Markers in Crop Improvement

Progress in plant breeding research in the past 15 years has resulted in development of high yielding crop varieties with superior agronomic potential. This was made possible by efficient utilisation of the vast genetic variation existing in wild relative and land races of the crop plant. Further, molecular mapping of these novel alleles have also significantly accelerated their transfer in cultivated varieties in lesser time.

Rice is one of the most important crops in the world. Grain weight is one of the major yield components of rice. A single InDel in rice QTL, *GW2*, is responsible for the variation in the grain weight. Therefore, to breed rice varieties with higher grain weight attempts were made to identify the allele specific markers for the *GW 2* QTL. Ninety-five rice genotypes including Basmati and non-Basmati *indica* were phenotyped for grain length,

grain width and test weight. Wide range of variation was observed particularly for test weight among these genotypes. Sequencing of the amplicons that corresponded to the functional domain of the protein encoded by the exon 4 of the *GW2* gene was carried out. No functional variation in the functional domain of the *GW2* locus in the *indica* genotypes studied could be found.

For breeding salt tolerant rice varieties A set of 69 rice accessions were evaluated for various salt stress reactions and grouped as tolerant (10), moderately tolerant (30), susceptible (17) and highly susceptible (8) (Fig. 24). SNP genotyping was performed using Illumina 48-Plex Golden Gate assay to analyse population structure and kinship relationships. Thirteen salt responsive candidate genes were re-sequenced from all 69 genotypes and 155 SNPs were discovered. Nucleotide and haplotype diversity was measured across all the genes and only six genes *SOS1*, *DUF6*, *HKT1;1,HKT1;5*(*SKC1*), *CCC* and *SNAC1* were found polymorphic. A candidate gene based association analysis found seven SNP markers in three genes, (*SOS1,DUF6* and *HKT2;4*) were significantly associated with salt tolerance. For *SOS1* gene, three SNPs, (D13, D18, D19) were significantly associated with Na^+ concentration in the shoot. The study identified potential salt tolerant alleles in rice and provided markers for their introgression into elite varieties.

Fig. 24. Response of wild rice accessions after 15 days of 150 mM of salt stress

Discovery and validation of molecular markers is of immense utility in molecular mapping of genes and QTLs. In the recent time, single nucleotide polymorphism (SNP) markers are increasingly becoming the marker system of choice. Therefore, with the objective of identifying SNPs in rice, Basmati370, a traditional Basmati rice cultivar and IRBB60, a non-Basmati *indica* rice variety were sequenced utilising ABI-SOLiD next generation sequencing platform. A total of 384 SNPs were selected covering the whole genome for validation, of which 163 showed polymorphism between Basmati370 and IRBB60. Out of these, 81 were converted to cost effective cleaved amplified polymorphic sequence (CAPS) markers and again 44 of them were validated between the two cultivars by restriction enzyme digestion.

To make advances in crop breeding it is important to identify molecular markers associated with agronomically important phenotypic traits. One of the important aspects of rice grain quality is chalkiness. It lowers the milling yield, gives poor overall appearance and also reduces palatability of cooked rice. To map the QTL for grain chalkiness association studies using diverse germplasm collection was conducted. A set of ninety-six rice genotypes was evaluated for three parameters of rice grain chalkiness viz. percentage of grains with chalkiness (PGWC), mean and median of relative percentage area of chalkiness. Establishment of association between SSR markers and the chalkiness was done by Power Marker software. Six SSR markers distributed on four chromosomes were found to be associated. Two of these linked markers were physically in the vicinity of earlier mapped QTLs. Association between traits and marker in regions that had not been implicated before possibly suggests new QTL.

Rice grain quality comprises aroma, grain dimensions (Length, breadth and shape) and cocking quality. Aroma and grain dimension have a direct effect on marketability or commercial success of improved rice cultivars. These traits are controlled by Quantitative Trait Loci (QTLs), showing continuous phenotypic variation in rice progenies. To map the QTL for grain length a cross between Pusa 1121 (an aromatic rice variety with long grains) and Pusa 1342 (a non-aromatic rice variety with bold grains) was used for the development of RIL population using single seed descent method. Phenotyping for grain length, breadth, shape and aroma was done in F_7 generation. For fine mapping of these QTLs, 184 F_6 RILs were grown and phenotyped in the normal rice growing season

at two different locations. The results of this study clearly indicate that both the parents of the mapping population, Pusa 1121 and Pusa 1342, contain the wild type *GS3* allele (Fig. 25). Besides sensory evaluation for aroma, first time a major compound responsible for aroma (2-Acetyl pyrroline) was chemically estimated using GC-MS (Gas Chromatography-Mass Spectrometry) method. SSR markers were used for fine mapping of the QTLs located on chromosome 1, 2, and 7 for grain dimensions and chromosome 3, 4 and 8 for aroma reported in the earlier study of the laboratory. During this study a new marker NKS_DEL, which is responsible for accurate genotyping of aromatic and non-aromatic rice was also developed through the sequencing of the BAD-2 (Betaine aldehyde dehydrogenase), a major gene present on chromosome-8 and responsible for aroma in rice.

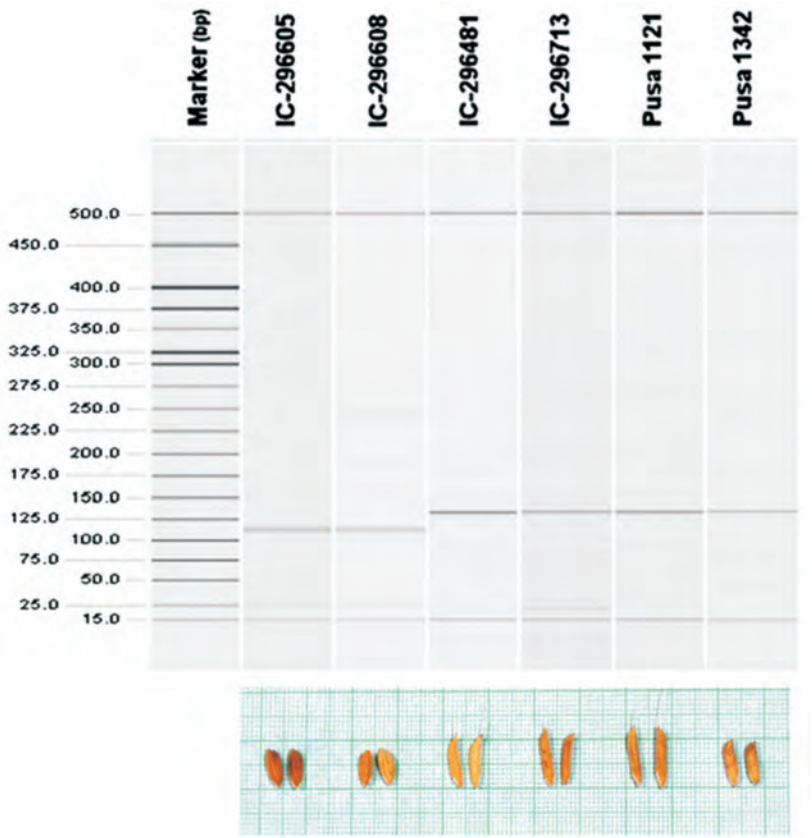

Fig. 25. Validation of the grain length gene *GS3* polymorphism in the parental lines of mapping population, Pusa 1121 and Pusa 1342 using SF28 CAPS marker

Similarly, for mapping grain size QTL in rice, a set of 276 F_7 recombinant inbred lines (RILs) derived from a cross between Basmati370 and IRBB60 were utilized (Fig 26). Seven previously validated CAPS and 45 SSR markers were used to genotype the RILs. A total of 18 markers showed linkage with grain size traits at $P<0.05$ using single marker analysis. Six of these positions were identified to have QTLs on chromosome 1, 6 and 8 explaining phenotypic variation to the extent of 4.8 - 45.4% through composite interval mapping. Fine mapping of the identified QTLs would lead to their cloning and characterization of variation in grain size at molecular level.

To develop a molecular linkage map and to identify QTLs for plant architecture and earliness traits in pigeonpea, a total of 1,439 molecular markers were screened on two parental genotypes, Pusa Dwarf and HDM04-1 (Fig. 27). Out of the selected molecular markers 28 SSRs and 107 SNPs were identified that amplified 136 segregating marker loci in the F_2 population. A framework linkage map was constructed using these 136 loci which were grouped into 12 linkage groups covering a total map distance of 1056.8 cM with an average marker interval of 7.77 cM. Composite interval mapping identified total 12 additive QTLs, 2 to 3 major QTLs for each trait viz., plant height, number of primary branches per plant pods per plant days to flowering and maturity etc. The mapped QTLs will facilitate marker assisted selection (MAS) and thus improve the efficiency of selection for high productivity in pigeonpea breeding.

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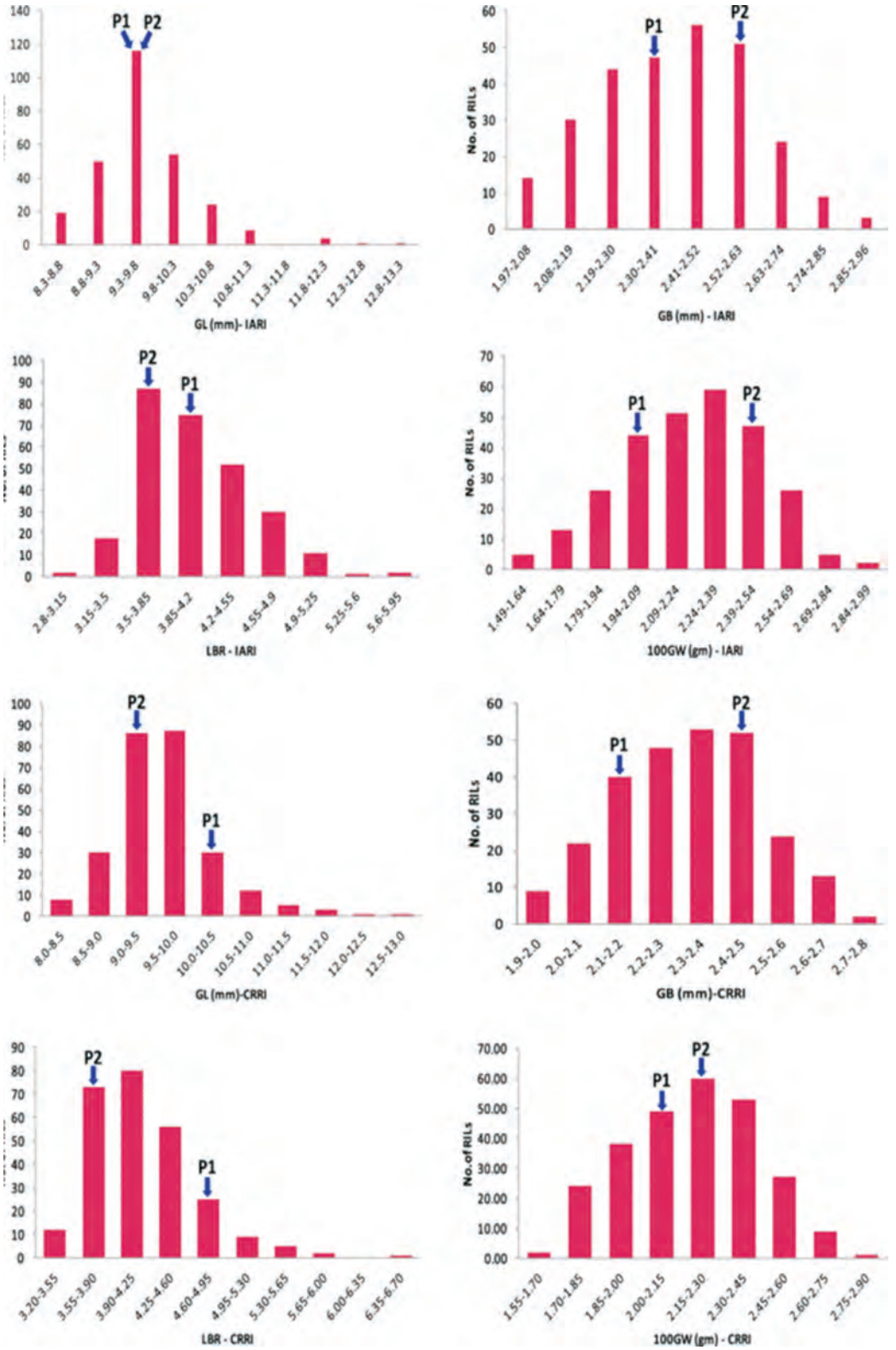

Fig. 26. Phenotypic distribution of the grain traits in Basmati 370/IRBB60 mapping population in IARI and CRRI environments. *P1* Basmati 370, *P2* IRBB60.

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Fig. 27 Parental genotypes Pusa Dwarf and HDM04-1

A research program was undertaken to combine grain quality of Basmati 370 with multiple bacterial blight resistance genes as well as desirable agronomic characteristics from a non-Basmati line IRBB60 using phenotypic and marker assisted selection. A set of 100 recombinant inbred lines (RILs) were selected based on grain length from a total of 1200 RILs derived from the cross of Basmati 370 x IRBB60. These 100 RILs were raised in the field and phenotypic data on disease reaction, agronomic and Basmati quality traits were recorded. Using molecular markers linked to the BLB resistance genes *Xa4*, *xa5, xa13* and*Xa21*, the presence or absence of these genes was ascertained in the selected RILs. RIL44 and RIL24 had aroma among the four-gene resistance lines. RIL44 also combined Basmati cooking quality traits along with superior agronomic traits. 54 polymorphic SSR markers were were used for background analysis of 18 RILs having three or four resistance genes. The extent of similarity with the Basmati parent was 24-69% in the 18 lines analyzed. RIL44 with four resistance genes was most similar to Basmati370 (69%). Moreover, many other RILs were also evaluated for the aroma trait and diseases resistance. The identified RILs can be a promising source for developing disease resistance basmati rice.

Summary

- Two blast resistant genes *Pi-rh* (an allele of *Pi-k*^{h} gene) and *Pi54* were cloned from blast resistant wild rice species, *Oryza rhizomatis* and indica rice cultivar Tetep, respectively.
- Allele mining of *Pb1* along with its promoter was carried out from 55 Indian rice accessions
- Chimeric insecticidal crystal proteins genes were constructed viz., *Jb-Jb-Ac*, *Jb-Ac-Jb* and *Jb-Ac-Ac* by exchanging the domains between *cry1Ac* and *cry1Jb*. Insect bioassay revealed that domain swapping can generate toxins with enhanced toxicity/introduce a specific activity into a toxin that does not possess it.
- *Cry* genes from native Bt isolates collected from diverse habitats and locations in India were characterized and submitted to NCBI database as well as to *Bacillus thuringiensis* Nomenclature Committee.
- The candidate gene related to grain size was mapped on short arms of chromosome 5 which was further fine mapped to an interval of 250kb.
- Through bioinformatic approach 24 HSF were identified in pigeon pea out of which 14 were selected for their detail characterization.
- In wheat based on advanced genomics tools 59 HS (Heat stress) responsive genes including 9 members of Caseinolytic protease (ClpATPase) family, 13 members of Small Heat Shock Protein (HSP20) family and 37 HS responsive genes (excluding HSPs and HSF) were identified in bread wheat.

- Expression profile of TaSnRK1 (Sucrose non-fermenting-1-related protein kinase 1) gene was analysed in wheat genotypes, C306 and HD2967 at different developmental stages.
- Allele mining of DREB2A transcription factor was done in 95 different rice accessions. A total of 398 unique positions having SNPs/InDels in *DREB2A* were identified.
- Defence genes induced by *Alternaria brassicae* were identified in *Sinapis alba* and *Camelina sativa.*They could be attributed to the degree of partial resistance of *S. alba* and*C. sativa* to *A. brassicae* infection.
- Gene expression of seven enzymes key to nitrogen use efficiency (NUE) were studied in eight wheat genotypes belonging to different group in terms of (NUE) under hydroponics condition with Nitrogen (N)-optimum (4.0 mM) and N-stressed (0.04 mM) conditions. Nitrate reductase (NR), glutamine synthatase (GS), and glutamate dehydrogenase (GDH) activity significantly decreased under N-starved condition.
- *TaDof1* expression was studied in four diverse wheat genotypes under N-deprived condition which revealed that *TaDof1* expression goes up under N-starvation and decreases gradually with the increase in stress.
- Transgenic mustard expressing pigeonpea lectin (*CCL*) gene driven by *rol*C promoter were developed and assayed for their insect resistance.
- Plant transformation vector was developed for dsRNA expression of key aphid genes towards introducing RNAi based aphid resistance in mustard.
- Transgenic tobacco plants expressing dsRNA of *acetylcholinesterase1* (*Ace1*) and *20-hydroxy ecdysone receptor* (*EcR)s* showed moderate to high level resistance against *H. armigera*.
- Transgenic lines containing the blast resistance gene *Pi-rh* from *Oryza rhizomatis* were found to be resistant to *M. oryzae* isolate PLP-1 in T_0 and T_1 generations.
- In laboratory based bioassay *Brassicajuncea* transgenic plants constitutively expressing NPR1 were found to be highly resistant to *Alternaria brassicae.*
- Drought resistance transgenic wheat was developed by over-expressing a cDNA encoding ascorbate peroxidase (TatAPX).
- Rice transgenic plants expressing transcription factor *BcZF2* and *ZmNF-YB2* showed better drought tolerance than the wild type plants confirming the role of *BcZF2* and *ZmNF-YB2* in imparting tolerance to water deficit stress.
- Effect of over-expression of key genes involved in oil biosynthesis (DGAT and G3PDH), and seed development (*KLUH* and *SHB1*) on productivity of *B. juncea* was investigated.
- Two novel promoters, *MYB02* and *MYB04* were cloned from drought tolerant rice cultivar Nagina-22. Both *MYB02* and *MYB04* promoters showed abiotic stress-responsive induction in transgenic *Arabidopsis.*
- A novel promoter of the gene *Lateral Organ Junction* gene (*LOJ*) was cloned from *Arabidopsis thaliana*
- A Cryptic Root-Specific Promoter (CRS) was isolated from *Arabidopsis thaliana.*
- Promoters of the *Arabidopsis thaliana* nuclear gene *Rps14* and *Oryza sativa* nuclear gene *Rps11* coding for mitochondrial ribosomal proteins were analyzed.
- The spatial and temporal regulation of gene expression pattern driven by *FAR6* promoter suggest that the corresponding *FAR6* gene (AT3G56700) may be involved in defense response against pathogens that damage the plant mainly through the epidermis.
- *Magnaporthe* inducibile promoter was identified from the rice line, HR 12.
- In chickpea using Illumina Genome analyser 368 miRNAs were identified. Possible role of four miRNA viz. miR2118, miR530, miR390 and miR166 in defense against *F. oxysporum,* were studied in wilt-resistant (WR 315, Pusa Green 112) and wilt-susceptible (BG 256, FLIP 90-166) genotypes in response to wilt stress in chickpea.

- Osa-miR3979 was identified as a novel moisture deficit stress responsive miRNA at flowering stage in Rice.
- For understanding the molecular mechanisms involved in fruit development of Brinjal (*Solanum Melongena* L) genome-wide transcriptome and proteome analysis was carried out during different stages of fruit development in brinjal cultivars Pusa Purple Long (PPL) and Killikulam (KKM-1).
- Comparative proteomics analysis of Bt and non- Bt Brinjal revealed that the expression of the insect resistance protein does not significantly alter the brinjal fruit and leaf proteomic profiles.
- Transcript profiling using the Affymetrix 57 K Rice GeneChip revealed a total of 72 differentially expressed genes at initial interaction stage of 6 hours post inoculation (hpi) with *M. oryzae* isolate Mo-si-63 which were significantly altered after pathogen infection.
- A drought responsive gene EcDehydrin7 containing dehydrin domain was cloned from *Eleucine coracana*. Over-expression of EcDehydrin7 confers drought tolerance in transgenic tobacco.
- EST sequence resource for drought, soil salinity stress and high temperature responsiveness in Pearl millet and *Prosopis cineraria* was developed.
- Cloning and comparative analysis of the *atp*α gene and transcripts of the CMS, fertility restored and euplasmic lines identified the presence of novel*orf108* which is responsible for male sterility in the CMS (*Moricandia arvensis*) *Brassica juncea*.
- Fine mapping of fertility restorer gene for CMS (*Moricandia arvensis*) *Brassica juncea* was done. The closest flanking markers BjESSR01 and BjEST01 were at 0.6 and 1.4 cM, respectively from the *Rf* locus.
- Experimentally, for the first time it was demonstrated that that haploidinducer lines could be obtained in a polyploid crop species through CENH3 engineering.
- For breeding salt tolerant rice varieties nucleotide and haplotype diversity was measured across six genes *SOS1*, *DUF6*, *HKT1; 1,HKT1;5*(*SKC1*), *CCC* and *SNAC1*. A candidate gene based association analysis found seven SNP markers in three genes, (*SOS1,DUF6* and *HKT2;4*) were significantly associated with salt tolerance.
- Grain size QTL in rice was mapped using a RIL population from Basmati × indica cross showing high segregation distortion.
- A new marker NKS_DEL, which is responsible for accurate genotyping of aromatic and non-aromatic rice was also developed through the sequencing of the BAD-2 (Betaine aldehyde dehydrogenase).
- QTLs were identified for plant architecture and earliness in pigeon pea.

Acknowledgment

I wish to thank all the faculty members of this teaching discipline for their relentless contribution in academic curriculum of this discipline.

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Plant Physiology

V. P. Singh and Lekshmy S.

Introduction

The Division of Plant Physiology at IARI was established on 14th November, 1966. Prior to its establishment as an independent Division, discipline of Plant Physiology existed as a section in erstwhile Division of Botany, IARI. Dr. J.J. Chinoy initiated physiological research at IARI way back in the year 1941. In the early fifties and sixties the research was oriented towards problems related with environmental stresses and plant nutrition. Dr. R.D. Asana became the first Head of the Division. The strong foundation for crop physiological research was laid during his tenure, and these studies are of practical relevance even in this era of "omics" research. Dr. G.S. Sirohi realized the importance of basic research in crop improvement and as a result an Advance Center of Plant Physiology was established in the year 1980 under United Nation Development Program. Initiation of Indo-US project under PL-480 scheme on 'Photosynthesis and Crop Productivity' from 1983 to 1993 was another effort towards strengthening the Division of Plant Physiology. Later on research work concentrated on nitrogen metabolism, post-harvest physiology, abiotic stress tolerance and global climate change. Now emphasis is on molecular physiology and hormonal regulation of mineral nutrition, post-harvest physiology and abiotic stress tolerance. Mission of the discipline is to enhance the physiological efficiencies for improving the productivity and production of crop plants.

Significant Student's Research Achievements

Keeping in view of the major aim of enhancing the physiological efficiencies for improving the productivity and production of crop plants, the post graduate research activities were centered around four major themes namely: Crop physiology, Post harvest physiology Mineral Nutrition and Abiotic stress tolerance.

Crop Physiology

To enhance the physiological efficiencies and for higher productivity and production of crop plants, a thorough understanding of crop physiology is warranted. As a part of post graduate research, few studies were conducted to understand the physiology of crop plants.

Barley has an initial advantage over wheat in terms of early emergence and vigour. Genotypic differences were observed in various physiological and biochemical traits, both among and within the species. Further, comparative analysis of vegetative growth in wheat and barley showed that barely had a higher dry matter production as compared to wheat. This was associated with higher rate of production of leaves and shoots or tillers. The number of nodal roots, root dry matter and root length density of upper 0-15 cm soil was higher in barley as compared to wheat. A higher CGR in barley was therefore maintained in comparison to wheat. The higher SLA and leaf growth of barley as compared to wheat was positively associated with the higher H_2O_2 content but negatively with the cell wall ferulic acid content. Overall the total dry matter production was higher in barley as compared to wheat in pots but the difference in dry matter was less in field. Comparative analysis of yield traits showed that barley compensated the lower number of grains per ear from its capacity to produce more tillers. The pattern of seed growth was similar in both wheat and barley in terms of grain

growth rates and grain growth duration; however, in barley the maximum grain growth rates were reached earlier than wheat.

Magnetopriming is a physical seed enhancement technique that causes effective seed invigouration and better plant growth in many crops. Cherry tomato seeds were optimized for seedling vigour by exposing the seeds to pulsed magnetic field of 100 mT for 30 min in 3 min on and off pulse. A maximum two fold increase over the control was recorded in reactive oxygen species (ROS), superoxide anion and hydrogen peroxide (H_2O_2), during germination of magnetoprimed seeds. ROS were involved in oxidative signaling during germination process and their levels were fine tuned to facilitate the enhanced germination characteristics. Superoxide dismutase activity and oxidative reaction of peroxidase resulted in production of increased levels of H_2O_2 during germination of magnetoprimed seeds. Further, ROS homeostasis was maintained by increase in protein carbonyls in primed seeds. The interplay of ROS with hormones showed decreased ABA levels in the germinating magnetoprimed seeds in early stages along with increasing levels of superoxide and H_2O_2. Gibberellic acid (GA) level detected after 8 h in the magnetoprimed seeds followed by a decline suggested that GA signaling might lead to GA biosynthesis towards the completion of germination. A significant 27% increase in yield was observed under controlled conditions indicating that pulsed magnetic field treatment is a remunerative and ecofriendly seed enhancement technology for productivity enhancement of cherry tomatoes.

The biologically active compounds present in smoke are now emerging as potential growth regulators for improving seed germination, vigour and seedling growth. This is due to presence of Karrikins and cyanohydrins in the smoke derived from burning of vegetation. The smoke water solution was prepared by bubbling of smoke into water by burning plant dry matter and filter paper. In the present study, the three seed priming treatments were – imbibition with water, plant derived smoke water (1000x dilution) and filter paper derived smoke water (800x dilution) for 14 hours. It was observed that the germination percentage and seedling growth of maize genotypes, PMH 3 and BIO 9681 was enhanced by smoke water as compared to seeds treated with water. The vigour index of smoke water treated seeds was higher due to better seed imbibition, lower base water potential, seed reserve mobilization and higher respiration. The starch and protein mobilization was higher in smoke water treated seeds. Seedling growth in relation to root length, shoot length and leaf area in genotype BIO 9681 was also enhanced in seeds treated with smoke water. In genotype PMH 3, overall seedling growth was found to be high in smoke water derived from filter paper. Further, smoke water treated seeds have also shown to overcome the effect of water stress, aquaporin inhibitors and high temperature stress. This study therefore showed the physiological aspects of enhanced germination and seedling vigour in maize by priming with smoke water. Thus seed priming with smoke water can be adopted as a cost-effective and simple means of enhancing seedling vigour.

Drought stress at flowering drastically affects yield of maize. Two important physiological traits identified in maize for improving maize yields in drought-prone environments are 1). Short Anthesis-Silking Interval (ASI) and 2). Low kernel abortion. Six advanced generation inbred lines (S_7) originated from drought tolerant population of CIMMYT, Mexico, representing highly tolerant (HT), moderately tolerant (T) and highly susceptible (HS) drought response groups were evaluated under irrigated and managed drought stress conditions. In HT maize inbreds, ASI was within 3 days under drought. Among HS inbreds, ASI was 6 days in MI-H07-103-17 inbred, while in the other two inbreds drought arrested the growth/development and prevented tasseling and silking. Analysis of plant water status clearly showed that, maintenance of high ear leaf relative water content (RWC) and water potential (Ψ) are necessary for short ASI. A RWC of <83% and Ψ of < -2.6 MPa appears to delay ASI, while RWC of <76% and Ψ of < -3.0 MPa prevent tasseling and silking. Analyses of chlorophyll content, leaf rolling and leaf senescence revealed that drought tolerance is associated with high chlorophyll content, less leaf rolling and stay green in maize inbreds. RWC (x) showed significant negative relationship with leaf rolling ($y = -0.041x^2 + 6.45x - 247.03$; $R^2 = 0.96$; $P \leq 0.01$), senescence ($y = -0.424x + 39.581$; $R^2 = 0.81$; $P \leq 0.01$), and significant positive correlation with ear leaf chlorophyll content ($y = 0.063x - 2.708$; $R^2 = 0.76$; $P \leq 0.01$). A study on source-sink carbohydrate levels showed that high level of reducing, non-reducing and total sugar contents in the ear leaf are associated with less kernel abortion in drought tolerant inbreds. Sink tissues such as silk and

ear tip also maintained high reducing, non-reducing and total sugar content in tolerant inbreds as compared to susceptible inbreds under drought stress. High drought induced ABA accumulation in sink tissues was found to be a cause for kernel abortion in maize under drought stress. Gene expression analysis showed that drought stress induced the expression of NAM, ATAF, and CUC (NAC) transcription factor (*NAC1*) gene in drought tolerant maize inbreds, while no expression was found in drought sensitive inbreds in the tassel leaves. In this study, a cDNA with complete coding sequence of *ZmNAC1* was cloned from maize inbreds (Gen Bank acc. No. EU810024 and EU810025). Drought responsive *NAC1* cloned from maize will be very useful in genetic engineering for enhancing abiotic stress tolerance of maize and other crop plants.

Post Harvest Physiology

The post graduate research on the theme post harvest physiology, encompassed studies on regulation of Flower Senescence in ethylene insensitive Gladiolus, ripening in tomato, ripening in banana, nutritional quality of banana, tomato, wheat and soybean.

Application of polyamine, Spermine (5µM) delayed flower senescence in ethylene insensitive flower, gladiolus by 3 days as compared to control. To understand the mechanism of spermine mediated delay in senescence, the impact on antioxidant enzyme superoxide dismutase (SOD) was investigated. SOD activity was significantly higher in treatment over control by 100-150 per cent on all days suggesting a possible role for SOD in delaying flower senescence by spermine. Throughout the observation period the Cu/Zn SOD activity of treated spikes was 50-100 per cent, while Fe SOD activity in treatment was 2-3 times higher than that of control from 1 DAT onwards. The Cu/Zn SOD activity followed the same pattern of total SOD activity in different stages. Cu/Zn SOD activation was higher in the initial stages of flower development; Fe SOD was activated more in the latter stages.

Quantitative analysis of SOD by negative staining on native PAGE using inhibitors revealed two isoforms and three isozymes; one Fe SOD and two Cu/Zn isozymes, Cu/Zn SOD1 and 2. The activity of total Cu/Zn SOD increased in treatment with spermine mainly on account of elevated activity of Cu/Zn SOD 2, which is more responsive to spermine than Cu/Zn SOD1. The amplified partial cDNA sequence of *GgSOD* has 327 bp with no 5'UTR and was partial with respect to 3' end and coded for a cytosolic Cu/Zn SOD.

The mechanism of protein degradation during flower senescence in gladiolus was studied. Twelve endoprotease inhibitors were tested to find out the impact on visible senescence of flowers and vase life and it was concluded that just before visible senescence more than half of total endoprotease activity was apparently due to cysteine proteases followed by serine proteases, with a minor or no role of aspartic and metalloproteases. Treatment of isolated florets with the known specific cysteine protease inhibitors N-ethylmaleimide and E-64 prevented the increase in endoprotease activity and considerably delayed or prevented the normal senescence symptoms. Further, studies on cysteine protease gene expression and proteolytic activity during floral development and senescence in ethylene-insensitive *Gladiolus* flowers revealed that there was up-regulation in the expression of *GgCyP* at the incipient senescent stage of flower development indicating that this gene may encode an important enzyme for the proteolytic process in *Gladiolus*. Regulation of petal senescence vis-à-vis cysteine proteases was also studied by analyzing the expression programme of *GgCyP* in petals as affected by various anti senescent compounds. Results indicated that out of the three antisenescent compounds used (Inositol, salicylic acid and E-64) salicylic acid at 400 µM was most effective and led to a dramatic decrease in *GgCyP* transcript levels within four hour of exposure.

Effect of polyols *viz*., mannitol, sorbitol, inositol and trehalose on delaying senescence of gladiolus was studied. The vase life of flower spikes was significantly increased by treatment with polyols in both the varieties (Snow Princess and Dhanwantari) studied. Spikes kept in vase solution containing inositol (75 mM) also maintained higher membrane stability and total soluble protein content, while lipid peroxidation in terms of thiobarbituric acid reactive substances (TBARS) and lipoxygenase (LOX) activity was reduced in comparison

to control. The activities of various antioxidant enzymes *viz.*, superoxide dismutase (SOD), catalase (CAT), glutathione reductase (GR) and ascorbate peroxidase (AP) were higher in inositol (75 mM) treated florets of both the varieties. These results suggest that inositol acts as a free radical scavenger and hence decreased their interaction with proteins, lipids and nucleic acids, delaying the process of lipid peroxidation and consequently senescence. Programmed cell death (PCD) was delayed significantly in inositol treated gladiolus flowers in terms of DNA fragmentation. Total DNA isolated from I to VII stage showed ladder like DNA banding pattern from IV stage onwards in control and from V stage in inositol treated florets. The expression of senescence associated genes (SAGs) *viz.*, *GgCyP1*, *Gglox1*, *GgERS1a* and *GgERS1b* were downregulated by inositol treatment during the course of flower development.

Flower spikes of *Gladiolus* cv. White prosperity kept in calcium (50mM) solution retained higher fresh weight as compared to the control and extended the vase life by 72 hours. Flower spikes treated with 50 mM Calcium retained higher membrane stability index (MSI) and soluble protein contents, and also for longer duration than the spikes kept in control solution. The level of lipid peroxidation in terms of thiobarbituric acid reactive substances (TBARS) content and lipoxygenase (LOX) activity gradually increased from bud stage to senescence stage in both control and treatment. This increase in TBARS content and lipoxygenase activity was reduced by treatment with calcium (50 mM) in comparison to control. Higher level of calcium was found in petals of calcium treated spikes throughout all the stages of flower development in comparison to control. Overall $CaCl_2$ treatment increased the endogenous level of calcium in the petal. The spikes treated with calcium (50 mM) solution maintained higher activity of SOD, APX and CAT in comparison to control. The expression of senescence associated genes (SAGs) *viz.*, *GgCyP1*, *GgERS1a* and *GgERS1b* increased gradually at initial stages and decreased at later stages in both control and treated florets. The expression of these genes was down regulated by calcium treatment during the course of flower development. The findings of this work will have significant implications for post harvest research. As calcium induced delay in senescence, it can be used in floriculture to prolong the vase life of gladiolus.

The vase life of flower spikes was significantly increased and fresh weight was retained for longer duration by treatment with thidiazuron (TDZ; 100 µM) than control (distilled water). Spikes kept in vase solution containing TDZ (100 µM) maintained higher membrane stability, while lipid peroxidation in terms of thiobarbutric acid reactive substances (TBARS) and lipoxygenase (LOX) activity was reduced in comparison to control. The activities of various antioxidant enzymes *viz.*, superoxide dismutase (SOD), catalase (CAT), total peroxidase (POX) and ascorbate peroxidase (AP) were higher in TDZ treated florets. These results suggest that thidiazuron acts as a free radical scavenger and hence decreased their interaction with proteins, lipids and nucleic acids, delaying the process of lipid peroxidation and consequently senescence. The expression of senescence associated gene *GgCyP1* was downregulated by thidiazuron treatment during the course of flower development and specifically at the terminal stage of flower senescence. However, *GgDAD1, GgERS1a* and *GgERS1b* were upregulated by thidiazuron treatment during the course of flower development and the expression pattern of upregulation was more conspicuous during senescent stages (incipient senescent and fully senescent stages) of flower development. The expression of these genes were more or less static from bud to fully open flower stages and then incipient senescent flower stage onwards the expression increased in both control and treated florets but the treated floret maintained higher expression of these genes than control at terminal senescent stage of gladiolus floret.

The vase life of flower spikes was significantly increased and fresh weight was retained for longer duration by treatment with NO than control (distilled water). Spikes kept in vase solution containing NO (100 ppm) maintained higher membrane stability, while lipid peroxidation in terms of thiobarbituric acid reactive substances (TBARS) and lipoxygenase (LOX) activity was reduced in comparison to control. The activities of various antioxidant enzymes *viz.*, superoxide dismutase (SOD), catalase (CAT), total peroxidase (POX) and ascorbate peroxidase (AP) were higher in NO (100 ppm) treated florets. These results suggest that nitric oxide acts as a free radical scavenger and hence decreased their interaction with proteins, lipids and nucleic acids, delaying the process of lipid peroxidation and consequently senescence. Treatment with NO (100 ppm) solution maintained

lower protease content in comparison to control at all the stages of flower development. The expression of senescence associated genes (SAGs) *viz.*, *GgCyP1*, *GgERS1a* and *GgERS1b* were downregulated by nitric oxide treatment during the course of flower development. The expression of these genes increased gradually during early stages of flower development and start declining from incipient senescent stage onwards in both control and treated florets but the decrease in expression was more prominent in control than NO treated florets. However, *GgDAD1* was upregulated by nitric oxide treatment during the course of flower development. The expression of this gene increased gradually from bud to incipient senescent flower stages and decreased in fully senescent stage in both control and treated florets but the treated floret maintained higher expression of these genes than control at terminal senescent stage of gladiolus floret.

Gladiolus flowers are ethylene insensitive and the signals that start catabolic changes during senescence of gladiolus flower are largely not known. Therefore, experiments were performed to understand the role of abscisic acid (ABA) in floral senescence of gladiolus (*Gladiolus grandiflora* Hort.) flower. It was observed that ABA accumulation increased in attached petals of gladiolus flowers as they senesced. Exogenous application of ABA in vase solution accelerated senescence process in the flowers due to change in various senescence indicators such as enhanced membrane leakage, reduced water uptake, reduced fresh weight and ultimately vase life. ABA (100 µM) also accelerates the senescence process by enhancing the biochemical changes such as lipoxygenase, lipid peroxidation, protease, RNase and DNase activity. Enhancement of *In vivo* ABA level in petals by creating osmotic stress also up regulates the same parameters of flower senescence as those occurring during natural senescence and also akin to exogenous application of ABA. Our finding has provided the initial evidence that, supplementation of gibberellic acid (GA_3) in the vase solution having ABA reversed various senescence related parameters which were down or upregulated by ABA proving role of ABA in senescence of gladiolus flowers. Further, gene expression analysis showed that the expression of *GgCyP1* was up regulated and *DAD1* was down regulated, whereas the ethylene receptor genes (*GgERS1a* and *GgERS1b*) were not significantly influenced in ABA treated flowers as compared to control.

Tomato (*Solanum lycopersicum* L.) fruit is the most important vegetable world over. This fruit is considered as a model system of climacteric fruit ripening but due to its climacteric nature itself, the postharvest losses are huge (up to 25 to 40 % or even more). Being a climacteric fruit, ethylene (a natural plant growth regulator) plays major role in the ripening of tomato. But, besides the ethylene, free radicals also influence the ripening and thereby in literature, especial attention has also been paid to examine the role of these free radicals in the ripening. The regulatory role vitamin C, phenolics, flavonoids and glutathione during fruit ripening was studied in tomato. Based on ripening index (RI %), varieties were grouped into three categories i.e., 1. Fast ripening type (Pusa Ruby and Pusa Sadabahar), 2. Slow ripening type (Pusa Gaurav and Roma) and 3. Intermediate ripening type (Pusa Sheetal, Pusa Uphar, Pusa Early Dwarf, Pusa Selection-8, Pusa-120 and Pusa Rohini). Different pigments and non-enzymatic antioxidants were estimated in the pericarp of tomato fruits at immature and GM stages of plant-harvested fruits and with the progress of ripening of fruits (harvested at GM stage) during storage. With the transition of fruits from immature to GM stage, pigments showed no change but, changes in the levels of antioxidants indicated (i). Increase in the level of oxidative stress and (ii). Tendency of tomato fruit to acclimatise against this change by enhancing the levels of antioxidants. On the other hand, with the progress of ripening the bioactive compounds showed decreasing (chlorophyll *a*, chlorophyll *b* and total chlorophylls, oxidized glutathione and ratio of oxidized to reduced glutathione), increasing (total carotenoids and lycopene, vitamin C, total soluble phenols, total flavonoids, total glutathione, reduced glutathione, glutathione redox ratio and reduced to oxidized glutathione) or static (chlorophyll *a*/*b* and total chlorophylls/total carotenoids) trends. Correlation studies pointed out the need for better protection of tomato fruits from oxidative stress right at the GM stage as this capacity provides the tomato fruit an attribute of delayed ripening during storage. In this context, glutathione appeared to play an important role

The endogenous levels of different mineral nutrients and their changes with the ripening behavior of tomato fruits were analysed. Transition of fruits from immature to green mature stage showed depicted decrease in

contents of Ca, Zn, and Mn in the outer pericarp region of the tomato fruit. This finding is of practical relevance because tomato fruits are generally harvested at this stage for their storage, transportation and marketing purposes. Comparison for the contents of individual nutrient in different varieties at green mature stage revealed varietal variability with respect to K, Mg, Ca, Fe, Cu and Mn. Implications of this observed varietal variability are highlighted on post-harvest physiology and ripening-related changes in tomato fruits.

Twenty cultivars of tomato were screened for higher shelf life treating with various concentration of oxalic acid (OA) and salicylic acid (SA) compounds. Green mature fruits of tomato were dipped in three concentrations of OA (*viz.* 2 mM, 3 mM and 4 mM) and three concentration of SA (*viz*. 0.5 mM, 0.75 mM and 1 mM), and double distilled water (DDW) used as control at 20^0C for 15 min. The most effective concentration was found out from the study was OA (3 mM) and SA (0.75 mM), which prolonged the shelf life of the tomato fruits by 4 and 7 days, respectively. These concentrations effectively delayed and reduced the ethylene production as well as cell wall hydrolyzing enzymes. They also delayed and maintained the synthesis of carotenoids, lycopene, ascorbic acid, total phenolics, free amino acids, γ-aminobutyric acid, total soluble solids, total sugar, titrable acidity and pH of ripened fruits with respect to untreated fruits. Moreover, enzymatic antioxidants activities such as SOD, CAT, GPX, APX and GR increased and oxidative stress reduced effectively in treated fruits as compared to untreated/control. Further, the mineral content such as Ca, Zn and Cu were maintained in treated fruits while it was reduced in untreated/control. Moreover, expression analysis of five ripening regulating genes *viz.* ACC synthase, ACC oxidase, PG, PME and cellulase were studied and it was found out that the expression levels of these genes were low in treated fruits as compared to untreated/control fruits.

Out of various approaches, hyperspectral reflectance is the most important tool for assessment of food quality. A study was therefore taken up with an objective to develop hyperspectral reflectance based indices/ models for non-destructive estimation of nutritional and quality parameters of tomato fruits. Various indices were identified and models were developed, best ones are as follows 1) Ripeness (maturity)/colour; R_{521} (RMSEC = 0.84, RMSEP = 0.86, bias = -0.09), 2) Firmness; $R_{501\ [Ist\ derivative]}$ (RMSEC = 1.20, RMSEP = 1.05, bias = -0.01) and 3) Lycopene; R_{546} (RMSEC = 4.93, RMSEP = 4.62, bias = 0.10). Developed models were found to be valid across varieties, ripening stages, plant harvested (fresh fruits) and stored conditions (aged fruits). The developed indices/models will facilitate the development of cost-effective tools for rapid/automated phenotyping, screening, monitoring and sorting of tomato fruits as desired by researches and tomato based agro-processing industries.

1-MCP treatment (1 µl/l for 6 hrs) to ripening banana caused a significant (68%) extension of shelf life in both the Robusta and Dwarf Cavendish. This delay in ripening caused by 1-MCP was associated with reduction in fresh weight, lower rate of respiration and ethylene evolution, a decreased pulp-peel ratio and TSS % compared to untreated control fruit. In contrast, peel recorded significantly higher activity of antioxidant enzymes studied. 1-MCP treatment significantly increased the activities of antioxidant enzymes viz. Ascorbate peroxidase, Peroxidase and Catalase and significantly reduced the activities of cell wall softening enzymes viz. Polygalacturonase, Pectin methyl esterase, Cellulase and Pectate lyase. 1-MCP also altered the carbon flux by delaying the breakdown of starch into sugars. Expression of Cytochrome P450 and β-1,3 glucanase genes have been associated with a general stress response in plants and are also closely related to fruit ripening. P450 expression was higher in peel than pulp, unlike β-1,3 glucanase, which expressed equally in both peel and pulp. While the expression of P450 was observed only during post-climacteric phase, β-1,3 glucanase expressed both in pre and post climacteric phase. Since exposure to 1-MCP, which is known to inhibit the action of ethylene, suppressed the transcript levels of both P450 and β-1,3 glucanase, it may be concluded that expression of both the genes is ethylene inducible. These genes could be a good choice for the isolation of fruit specific promoters.

Beta carotene is the most potent precursor of vitamin A, found abundantly in fruits like Banana. So, screening the genetic diversity of major Indian banana cultivars that contain various valuable bioactive compounds, such as carotenoids (specifically β-carotene), carbohydrates and protein as well as mineral nutrients in the fruit may offer a potential food source for alleviating vitamin A deficiency and malnutrition. The provitamin A carotenoid

was found to be more in non edible peel than in pulp. Among the cultivars screened Nendran showed the highest beta carotene both in peel and pulp. The estimated protein content also follows the same trend as banana peel contains more protein than in pulp. The various antioxidant enzymes such as SOD, APOX, CAT and GR were present more in peel of plantain 'Monthan'. 'Red banana' contains the highest starch content in peel (82 µg/g peel) and pulp (269 µg/g pulp). Next to Red banana, 'Monthan' contain more starch in pulp (212 µg/g pulp. Thus, banana cultivars like 'Red banana', 'karpuravalli' and plantain cultivars like 'Nendran', 'Monthan' can be exploited for their bioactive compounds. The phytoene synthase (*psy*), a rate limiting enzyme in the carotenoid biosynthetic pathway in banana fruit was isolated and molecularly characterized. The pattern of *MaPsy* expression in peel and pulp revealed that, the expression were 3-4 times higher in non edible peel than the pulp. Moreover, in peel two *MaPsy* transcript variants were present compared to only one in case of pulp.

The effect of gamma radiation and static magnetic field, on the plant growth and yield characteristics of wheat and post harvest storage behavior of tomato and soybean were studied. Gamma irradiation, in general, improved the growth, yield as well as the quality characteristics of wheat. Magnetic field and gamma radiation in combination had antagonistic effect on grain yield of wheat. Low water activity, high seed coat hardness, lower activity of LOX and higher activity of antioxidant enzymes favored the possibility of utilizing the gamma radiation and magnetic energy for prolonging the post harvest storage of soybean seeds under ambient as well as under accelerated aging conditions with minimal effect on the seed vigor and quality. Increased fruit firmness, significantly low ethylene production and high SOD activity together with high K content in post harvest irradiated fruits of tomato suggested that low dose gamma radiation (< 1 kGy) can be used to extend the shelf life of tomato with least effect on the quality of stored fruits.

Mineral Nutrition

Nitrogen and phosphorus (P) plays a pivotal structural and regulatory role in plants at the nexus of photosynthesis, energy conservation and carbon metabolism. But their deficiency is a common phenomenon which seriously limits crop production all over the world. Experiments were conducted to study mechanisms of N and P uptake and assimilation efficiency in different crops with the following objectives:

Organic acid exudation and its relationship with P uptake efficiency in mungbean genotypes grown under low (2 µM) and sufficient (100 µM) P was studied. Forty four mungbean genotypes grown at low and sufficient P were labeled with ^{14}C and total exudation was assessed by counting ^{14}C. The selected 7 genotypes were used to measure root exudation and internal concentration of organic acids (OA). In the rhizosphere under low P mainly oxalic and citric acids were exuded while those in root tissues were oxalic, succinic, lactic and maleic acids. Synthesis of oxalic, succinic and lactic acids were higher under low P in root tissues but they were not released into the rhizosphere. Among the morpho-physiological parameters, the chlorophyll *a*, total chlorophyll and starch concentration were higher in low P compared to sufficient P. The P utilization efficiency was higher in low P plants. Thus, this study shows that exudation of organic acid in the rhizosphere is an important phenomena which in involved in solubilizing the fixed P.

Responses of bread wheat (*Triticum aestivum*), durum wheat (*Triticum durum*) and rye (*Secale cereale*) to the interaction between two P levels (0 and 500 µM) and two CO_2 concentrations (380 and 700 µmol mol^{-1}) in terms of changes at morpho-physiological, biochemical and molecular level were studied. Results revealed that root, shoot and total plant dry matter accumulation and partitioning were positively affected by CO_2 enrichment. Significant difference in root length, surface area and lateral root density in wheat species in response to eCO_2 and P nutrition were observed. The increase in total chl concentration at –P over +P when grown under eCO_2 was 34.3% while P treatment had no significant effect at ambient CO_2. Rye accumulated very high amount of carotenoid under eCO_2 without P. The reducing sugars increased by 91.2% and 67.5%, while non-reducing sugars increased to 2.5 folds and 2.3 folds under aCO_2 and eCO_2, respectively at –P over +P. The shoot P concentration increased by 21.2% at eCO_2 compared to aCO_2 when grown with P. Maximum P use efficiency was recorded in rye, while the lowest was in durum. Among kinetic parameters, I_{max} increased under eCO_2 in

all three species while K_m decreased, except in durum, resulting in rapid uptake of ^{32}P during the initial period of P uptake between 20 to 60 minutes. The expression of high affinity phosphate transporter gene showed that when P-starved seedlings were induced with different concentration of P, the maximum expression was obtained at 5 µM and 20 min. These finding suggests that the plants would be responsive to moderate P deficiency under ever increasing CO_2 level.

Fifty wheat cultivars (*Triticum aestivum* L.) were grown in hydroponics with recommended culture practices in iron sufficient and deficient (100µM and 1µM) nutrient solution culture and greenness index (SPAD readings) was measured every 3 days of transfer to the nutrient solution until 21 days to delineate iron efficient and inefficient cultivars. For further studies, five cultivars from the above were selected and their root characteristics and PS metabolism were studied. Leaf greenness index could be used as a rapid and reliable marker for segregating Fe deficiency tolerant and susceptible cultivars. Activity of SOD, catalase, peroxidase and total antioxidant was higher in case of iron deficient condition as compared to the sufficient condition. Although PS release under Zn, Cu, Mn and multi nutrient deficiency also peaked at 11 day stage, the release was about four times lesser in comparison to iron deficiency. Diurnal rhythm of PS release under various micronutrient deficiency (Fe, Zn, Cu, Mn, multi deficiency) was similar. Limitation at the level of release of the PS was responsible for low Fe use efficiency of the Fe deficiency susceptible cultivars. Multi micronutrient deficiency caused a greater induction of PS synthesis in comparison to iron deficiency but not in the PS release suggesting, that the PS release is determined by a threshold**.**

The role and physiological basis of S mediated regulation of Fe uptake and use efficiency under field (S_0, S_{30} and S_{60}) and solution cuture was studied bread (HD-2967) and durum (HI-8713) wheat genotypes with Fe deficiency tolerance response. Across the wheat genotypes, both Fe and S uptake but not Fe- and/or S-use efficiency were greatly improved at S_{30} and S_{60}. A relatively higher shoot mass of bread wheat over durum wheat, under $^{+}S^{-}Fe$ than $S^{-}Fe^{-}$ treatment was due to better efficiency of bread wheat to cope up with Fe deficiency stress through a higher release of phytosiderophores (PS) in the rhizosphere than durum wheat. Durum wheat released relatively lower PS despite optimum synthesis of root PS. Diurnal variation for the PS release under different treatments of S and Fe availability was similar. Further, the transcript expression profile of S transporter (SULTR1;1) and Fe transporter (YS1) showed that *SULTR1;1* transient expression requires not only low level of S but also a sufficient level of Fe while the reverse was true for *YS1* expression.

To characterise the physiological aspects influencing organic acid exudation under low phosphorus (P) stress, experiments were performed to screen 116 soybean genotypes for total carbon exudation by labelling shoot with $^{14}CO_2$. Among the traits measured at seedling stage, total carbon (^{14}C) exudation, P uptake and total dry weight contributed to maximum genotypic variability. The proportion of organic acids (oxalate, citrate, succinate and fumarate) was highest among root-exuded compounds induced by low P stress. Improved root length, surface area and volume coupled with higher activity of TCA cycle enzymes contributed to enhanced organic acid exudation under low P. Efficient genotypes (EC-232019 and G-2344) exhibited superior growth and P acquisition efficiency under low soil P attributed to its higher root exudation potential aiding in mining fixed P. To understand the molecular mechanism differentially regulating root exudation potential in contrasting genotypes (EC-232019 and EC-113396), root proteome analysis was carried out. Among the total proteins, 32% were differentially expressed between sufficient and low P levels. A total of 14% proteins were down-regulated by > two-fold under low P while 15% were up-regulated by > two-fold at low P. The differential proteins were involved in a myriad of functions including organic acid accumulation, carbohydrate, protein and lipid metabolism under low P stress. Characterisation of 14 proteins with unknown function might reveal roles of novel genes under low P stress. The identified genotypes have potential to be used as donors in crop improvement programs to develop high-yielding P-efficient cultivars.

When plant faces nitrogen stress, it is possible that due to stress in spite of low protein content of tissues, they might be oxidised and carbonylated leading to increased degradation in such plants facing nutrient stress. The interaction between cytokinins gene expression and N was also investigated. The extent of protein

carbonylation was analyzed and an attempt was made to elucidate the pathway of protein degradation. Wheat seedlings (var. PBW 343) were grown under four different N treatments and various parameters of growth, N-assimilation and stress was studied. Result indicated that growth and nitrogen metabolism was enhanced in seedlings under high NO_3^--N when compared to those grown with low and without N. There was increased production of ROS in terms of increase in superoxide radicals and H_2O_2 in NH_4^+-N fed wheat seedlings followed by those grown without N. Increase in protease activity, total free amino acids and TBARS was also observed in these treatments. Under normal conditions, plants contain numerous carbonylated proteins, which are thought to be indicative of oxidative stress damage. Conditions that promote formation of reactive oxygen species (ROS) enhance protein carbonylation, and protein degradation is required to reverse the damage. However, it is not clear how the degradation of carbonylated proteins is controlled *in planta.* Detached wheat leaves rapidly and selectively degrades carbonylated proteins when kept in the dark as compared to the detached leaves incubated in light or in the intact seedlings. The loss of carbonylated proteins corresponded to a loss of soluble protein and accumulation of free amino acids. Degradation of carbonylated proteins was blocked by carbobenzoxyl-leucinyl-leucinyl-leucinal (MG132) in dark detached leaves, but not by 3-methyladenine, suggesting that the 26S proteasome pathway rather than the autophagic pathway was involved in degrading protein carbonyls in dark detached leaves. In light incubation pathway the use of inhibitors revealed that both autophagic and proteasomal pathways were involved in degrading protein carbonyls. The study also indicated the role of cytokinins in preventing the the protein carbonyl degradation. The expression of *IPT3* gene of cytokinin biosynthetic pathway was almost absent in the seedlings grown at zero N and NH4+-N, and in these treatments the degradation of protein carbonyls was high as supported by low level of total soluble proteins and high levels of free amino acids.

Present study was undertaken to understand and characterize the possible role of phytosiderophore in phytoremediation of heavy metals. Several independent experiments were carried out to deduce physiochemical and molecular mechanism behind interaction of phytosiderophore with heavy metals vis-à-vis iron nutrition. Members of crop species from *graminae* family viz wheat (*Triticum aestivum* L., var PBW 343), Barley (*Hordeum vulgare*, var BH-393) and Oat (*Avena sativa*, var Kent) grown under hydroponics system were used for various studies. Root exudates were collected from 10-12 days old iron deficient seedlings of all the three crop species which are known to release different classes of phytosiderophores. Rest of the experiments including molecular studies were carried out with wheat seedlings only, grown in Fe deficient condition. Ten days old wheat seedlings were transferred to nutrient solution containing three different concentrations (2.5, 5.0 and 10.0 mM) of heavy metals (Cd, Pb and Ni) for 72 h and root and shoot tissues and root exudates were collected for various studies. Heavy metals concentration in plants was significantly and positively correlated with heavy metals concentration in the growth solution. Heavy metals treatment led to more accumulation of Fe in both roots and shoots. Root content of Fe was more in case of Cd treatment whereas maximum Fe content in shoots was observed in case of Pb treatment. Phytosiderophore release was also affected by presence of heavy metals in the nutrient solution. The phytosiderophore secretion capacity of wheat seedlings grown in Fe-deficient condition was increased in the presence of heavy metals. It was found that barley secreted highest amount of phytosiderophore followed by oat while phytosiderophore release capacity of wheat was statistically similar to oat. HPLC analysis showed that barley and oat phytosiderophore could mobilise all the heavy metals including Fe while wheat PS could mobilise all but Pb. Resin bound metal mobilization study confirm the above findings. RT-PCR analysis of genes, *nas* and *naat* involved in phytosiderophore biosynthesis revealed that both genes were expressed under Fe-deficiency and in the presence of heavy metals. Multiple sequence alignment of *nas* from wheat revealed 92% sequence similarity with *nas* from barley.

Phosphorus (P) is a non-renewable resource and its interaction with sulphur (S) under elevated [CO_2] influences plant growth at physiological and molecular level. Experiments conducted on wheat (cv. PDW-233 and PBW-396) to study the interactive effects of P (5 µM and 500 µM) and [CO_2] (400 and 700 µL L^{-1}) with different S (10 µM and 2000 µM) levels. Results showed increased biomass, improved root traits, higher exudation of acid phosphatase and total carboxylates in response to low-P and high [CO_2] with a corresponding increase in

activity of enzymes involved in carboxylate synthesis. Interactive effects of high [CO_2] and P stress on membrane lipid composition resulted in reduced amount of phosphatidylglycerol (PG) but sulfoquinovosyl diacylglycerol (SQDG) increased >2-folds. Low S grown plants possessed less SQDG content but the interactive effects of high [CO_2] and low S resulted in 2-fold increase in PG while amount of SQDG remained lower. Similarly, the relative expression of genes (*TaNPC4, TaPLDζ1, TaSQDG1* and *TaSQDG2*) involved in synthesis of membrane lipids was also was highly induced by high [CO_2] and low P or low S. Thus, at higher atmospheric [CO_2] plants respond to P deficiency by altering its membrane lipid composition provided there is sufficient S fertilization.

Nitrogen concentration declines as a result of exposure of the crop plants to the elevated CO_2. Wheat genotypes, DT 144 and PBW 343 were grown in ambient (380 µmol mol^{-1}) and elevated (Free Air CO_2 Enrichment technology, FACE, 550±50µmol mol^{-1}). Both the genotypes showed increased biomass in terms of dry weight per plant, tiller number, leaf area. PBW343 had significantly higher biomass as compared to DT 144 in both AC and EC. Increased yield was due to the increase in the number of structures and not their size. C, C:N ratio, total chlorophyll and carotenoid content increased in CO_2 enriched plants due to significant reduction in N concentration in all the plant parts analysed. It could be due to faster accumulation of biomass and dilution effect. Non reducing sugars increased more than the reducing sugars indicating more mobilization under EC in both the genotypes. Study also indicated that NR activity increased in both genotypes in EC, where as GS activity increased only in PBW 343 and declined in DT 144. The expression of GS 1 also enhanced in the CO_2 enriched plants of PBW 343 during grain filling stages indicating higher N mobilisation. Protein oxidation decreased in the CO_2 enriched plants during reproductive stage and in spite of that lower total soluble proteins were recorded in the plants of both the genotypes grown under EC. The harvested grains of both the genotypes had lower N and higher C:N ratio in the EC grown plants of both the genotypes. Starch content increased in grains of CO_2 enriched plants. HMW glutenins were unaffected in the PBW 343, but the first Ssubunit of the A genome was not detected in DT 144. Lowering of protein content in response to rising CO_2 may affect the baking quality of wheat flour of hexaploid wheats. It may also affect the nutrition as DT 144 is used for making breakfast cereals and may also reduce the quality of straw for animal feed.

Potato is an important food crop and sensitive to high temperature, therefore rising temperature in future may affect productivity of this crop. In the present study, impact of high temperature and CO_2 on growth and yield was studied in high temperature tolerant (*Kufri Surya*) and sensitive (*Kufri Chipsona*-3) potato cultivars. High temperature exposure delayed tuberization, shortened duration of bulking, decreased partitioning of dry matter towards tubers, crop growth rate (CGR), tuber growth rate (TGR) and net assimilation rate (NAR) in both the potato cultivars. Photosynthesis, stomatal conductance, chlorophylls, membrane stability and proline content decreased with increasing the temperature but glycinebetaine (GB) increased and K. Surya maintained higher GB than Chipsona-3. Under high CO_2, the photosynthetic rate, CGR, TGR and partitioning of dry matter increased in both the cultivars. The increase in tuber yield and tuber fresh weight was observed due to enhanced number of tubers per plant. The tuber yield under elevated CO_2 was higher in K. Chipsona compared with K. Surya. Higher glycinebetaine concentration and less reduction in photosynthesis and yield attributes in K. Surya appears to be associated with its high temperature tolerance compared with Chipsona. On the other hand better response of K. Chipsona to elevated CO_2 suggests that elevated CO_2 may mitigate high temperature effects in potato.

The impact of elevated CO_2 and temperature on physiological characteristics of two chickpea genotypes *viz.* Pusa 1103 (*desi*) and Pusa 1105 (*kabuli*) were analysed. Both the chickpea genotypes were raised inside open top chamber (OTC) under ambient (380±30 µmol mol^{-1}) and elevated (570±50 µmol mol^{-1}) CO_2 concentration and for high temperature exposure inside polycover (temperature 5.93±0.22^{0}C above ambient). Elevated CO_2 prolonged the duration to flower and pod formation, while elevated temperature advanced these phenological phases. Elevated CO_2 exposure enhanced seed yield, while temperature enhanced vegetative growth but decreased pod setting and seed yield in both the genotypes. Leaf nitrogen and micronutrient concentration reduced under elevated CO_2. Membrane stability index was not affected significantly and only marginal changes were observed

in both the genotypes. The study also revealed a complex regulation of genes for photorespiration by EC and ET and hence, warrants further studies to understand this critical process under climate change scenario.

A pot study was conducted to quantify the effect of elevated CO_2 and P on growth and physiology associated with biomass accumulation pattern at different growth stages in mungbean (*Vigna radiata* (L.) Wilczek). Seeds of cv. ML-818 and PDM-139 were sown in pots with sufficient and low-P exposed to elevated (EC, 550 ± 50 µmol mol^{-1}) and ambient (AC, 384 µmol mol^{-1}) CO_2 in open top chambers. Biomass accumulation and total leaf area increased under EC compared to AC, maximum accumulation being recorded at anthesis stage. Plants responded to EC even under low-P by accumulating higher whole plant biomass which was comparable to sufficient P grown plant under AC. Rate of photosynthesis was significantly higher under EC with sufficient P. Root length, volume and surface area increased significantly at low-P under EC. Nodule number, weight and nitrogenase activity were significantly higher under EC with sufficient P leading to maximum N concentration and uptake. Partitioning of P in leaf under EC was higher compared to AC grown plants during anthesis. P x CO_2 treatment increased number of pods per plant, 100-seed weight and seed yield while number of seed per pod was not affected by [CO_2]. Though the yield was more under elevated CO_2 in both varieties but the yield stability was higher in PDM-139 as it showed less reduction in yield under elevated CO_2 and low-P compared to sufficient P. These results indicate that higher CO_2 can compensate for P requirement to some extent which may be because of increased nutrient acquisition and utilization efficiency.

The study was conducted to determine the effect of rising CO_2 levels on changes in mechanism of nitrogen (N) assimilation in *Triticum aestivum*, *Triticum durum* and *Secale cereale* grown in ambient CO_2 (AC; 380 ppm) and elevated CO_2 (EC; 550±50 ppm). EC increased biomass, growth, yield in all the genotypes, but resulted in decline in specific leaf weight. The activity of enzyme nitrate reductase, gutamine synthetase (GS) and the expression of NR gene (*NIA2*) and GS1 increased in EC grown plants during reproductive stage but the activity of GS2 declined at dough stage especially in ears, resulting in significant decrease in grain N % and crude protein (%) in EC grown plants. The plant growth under EC was maximum when both ammonium and nitrate were present in the medium. The ammonium nutrition increased the tissue N content of the EC grown wheat plants. Reactive oxygen species generation was lower in EC grown nitrate fed plants, but it was more in ammonium N fed plants, with significant increase in ascorbate peroxidase (APX) activity in these plants. EC though reduced thiobarbituric acid reactive substances (TBARS), irrespective of N source, but could not fully alleviate the ammonium toxicity in wheat seedling and APX might play an important role in alleviating this toxicity. Rising CO_2 might require changes in pattern of fertigation in future as study indicates inhibition of photoassimilation of nitrate as shown by apparent loss of only GS2 expression and activity during reproductive stages in wheat.

The effect of organic and inorganic sources of nutrients on the growth, development and yield of soybean [*Glycine max* (L.) Merr] was studied under field conditions. Treatments, T_1- control; T_2- Farm Yard Manure (FYM); T_3- Vermicompost (VC); T_4- *Rhizobium japonica* (RJ); T_5- FYM+VC+RJ; T_6- 50% (FYM + VC + RJ) + 50% (Urea + Single super phosphate + muriate of potash); T_7- Urea + Single super phosphate + Muriate of potash applied with three replications. The growth and yield parameters of soybean are positively influenced in T_6. This enhancement in the growth parameters may be due to the increased mineral nutrients status of plants which is reflected through increased nutrients status of soybean (N, 0.35%; P, 6.08%; Ca, 11.01%; Mg, 6.17%; S, 26.9%; Cu, 3.03%; Zn, 11.53%; Mn, 2.941%; Mo, 14.8%; B, 8.53%) in seeds of T_6 plants. The reason for increased micronutrients in organic manures applied plants may be due to the ability of these manures to supply the essential nutrients which are seldom supplied by straight fertilizers. There was an increase of phytonutrients, membrane stability in T_5 and T_6 vindicating the beneficial influence of organic manures on soybean. The reason being the essential micronutrients supply met through organic manures. Increased activity of lipoxygenase in the organic manures treated plants indicates the positive role played by manures in enhancing enzyme activity as it involve in plant defence mechanism. The increased ascorbic acid in T_5 and T_6 shows better nutritional status of the plants. Thus we conclude that, organic manures play important roles in enhancing the crop health and the phytonutrients level when compared to inorganic fertilizers in the legume cum oil seed crop soybean.

Abiotic Stress Tolerance

The research theme, abiotic stress tolerance encompassed a major part of post graduate research. The research areas included abiotic stress (Salinity, drought, waterlogging, high temperature) responses in major crops such as, rice, wheat, chick pea, pigeon pea, brassica, mung bean and model plant such as tobacco and *Arabidopsis*.

Abiotic stresses cause depreciation in crop yield up to 70%. Understanding of the molecular basis of abiotic stress tolerance is necessary to develop abiotic stress resistant crops. Putative salt stress responsive T-DNA insertion mutant lines of *Arabidopsis thaliana* (L.) Heynh. ecotype Columbia, were screened to isolate mutants impaired in salt (NaCl) stress response in T3 and were confirmed in T4 generations. Studies on ABA, glucose and oxidative stress responses revealed that *salt stress resistant* (*sst*) mutants isolated in this study are not impaired in ABA signaling pathways and are non-allelic to previously known salt resistant mutants. Genomic location of T-DNA insertion in the mutants was mapped by using TAIL-PCR, and further confirmed by diagnostic PCR. The *salt overly sensitive 101* (*sos101*) mutation is caused by T-DNA insertion in the promoter region of *SOS101* (AT1G64610), which encodes a WD-40 repeat family protein. Salt stress up regulation of *SOS101* in WT plants suggests that SOS101 is a positive regulator of plant salt tolerance. The *sst3* knock-down mutation is caused by T-DNA insertion in the 3' UTR region of SST3 (At2G30210), which encodes a putative laccase (*LACCASE 3, LAC3*). A constitutively low level of *SST3* in the mutants probably contributed to the enhanced tolerance. In *sst4* mutant, T-DNA insertion is at 1087 bases upstream to the initiation codon of *SST4a* (*At3G48770*) and -1189 bases 5' to the transcription start site of *SST4b* (*At3G48780*) genes. *SST4a* encodes an ATP-binding/ DNA binding protein, while *SST4b* encodes a serine C-palmitoyltransferase probably involved in sphingolipid biosynthesis. Thus, three novel loci involved in salt stress response were mapped and cloned from *Arabidopsis*. These genes can be employed for enhancing salt tolerance of crop plants.

Various studies on lipid constitution of membranes revealed the importance of dienoic fatty acids in high temperature tolerance. Post transcriptional gene silencing (PTGS) approach using Virus-induced gene silencing (VIGS) was used to silence FAD7 in order to increase dienoic fatty acids. Endogenous FAD7 gene fragment from *Nicotiana benthamiana* was cloned to construct VIGS vector (pTRV2: FAD7). The *Agrobacterium* strain harboring the pTRV2: FAD7; pTRV2: PDS and pTRV1 were infiltrated into 4 -leaf stage *N. benthamiana* plants. The down regulation of FAD7 gene in different plants were confirmed through semi quantitative RT-PCR and were subjected to high temperature stress. In silenced plants, the content of linolenic acid (18:3) markedly decreased, while linoleic acid (18:2) increased correspondingly and similar changes were observed under high-temperature stress. Under high-temperature stress, silenced plants showed relatively higher level of net photosynthetic rate and chlorophyll fluorescence than that of control and mock plants. However, lower levels of H_2O_2 content, TBARS, electrolyte conductivity and antioxidative enzyme activities were observed in silenced plants as compared to control. The study concludes that FAD7 silenced plants can tolerate high temperature stress through maintaining higher dienoic to trienoic fatty acids ratio and greater membrane stability.

Crop lodging is a serious problem undermining yield under optimum input condition. On the basis of plant height, stem strength and lodging resistance factor index (LRFI) the wheat genotypes were grouped into lodging tolerant and susceptible. The susceptible genotypes had advantage in terms of higher biomass and photosynthetic efficiency; and these traits along with stem strength and lodging resistance factor index (LRFI) are required to be incorporated into tolerant genotypes to enhance productivity potential of wheat genotypes. Under late sown condition plant height was reduced and stem strength increased, which provided stiffness against lodging and increased lodging resistance factor index even in lodging susceptible genotypes. Biochemical components such as potassium, calcium, silica, lignin and hemi-cellulose also increased under late sown conditions, which played vital role in lodging resistance. To understand the mode of action of ethephon application in relation to lodging in three lodging tolerant (HD-2329, Raj-4014 and Raj-3765) and three lodging susceptible (HD-2913, C-306 and RR-3) wheat genotypes were studied. The results indicated that the plant height was reduced significantly in lodging susceptible genotypes, while non-significant differences were observed in case of lodging tolerant

genotypes under ethephon applications. Significant improvement was also observed in the stem strength of lodging susceptible genotypes under ethephon applications. The chain weight recorded positive correlation with lodging resistance factor index, grain yield, grain weight and harvest index, but it was negatively correlated with plant height. Yield and yield components increased in lodging susceptible genotypes under ethephon applications, but not in lodging tolerant genotypes. From the present study it can be concluded that lodging susceptible genotypes should only be sown under late and very late sown condition. Application of ethephon is beneficial only in timely sown crops, in case farmers want to grow tall genotypes with better grain qualities.

Six wheat genotypes viz. Kharchia 65, KRL19, HD2009, HD2687, HD4713, and WL 711 were raised in earthen pots of uniform size and subjected to salinity treatments 100 mM NaCl (S_1), 200mM NaCl (S_2) and water (S_0). Salinity stress reduced relative water content, contents of chlorophyll, and carotenoids, membrane stability index, dry matter (root and shoot), and yield in all genotypes; more so in salinity susceptible genotypes like HD 2687 and WL 711. Salt-stress induced accumulation of reactive oxygen species measured in terms of Lipid peroxidation, Hydrogen peroxide and super oxide radicals were comparatively lower in Kharchia 65 and HD 2009, which was better induction of anti oxidant system consisting of super oxide dismutase, catalase, ascorbate peroxidase and glutathione reductase. Salinity stress stimulated accumulation of compatible solutes like soluble sugars, trehalose, proline and glycine betaine. Tolerent cultivar like Kharchia 65 maintained higher levels of osmolytes by transcriptional activation of corresponding enzymes. Soil salinity significantly reduced the potassium, calcium, nitrogen and iron content in leaf root and seeds of all the genotypes. In contrast, the sodium, manganese, copper and zinc content was increased in all the plant parts under salt treatment. Expression analysis of genes of sodium exclusion mechanism in both roots and leaves of wheat reveals the existence of a more efficient *Salt Overly Sensitive* pathway composed of *SOS1*, *SOS2* ,*SOS3* , vacuolar Na^+/H^+ antiporter and *Vacolar H^+pyro phosphatase* in tolerant genotypes like Kharchia 65 . Kharchia 65 also displayed higher induction of stress signaling genes like MAP Kinase, transcription factors like NAC and bZIP.

Wheat seeds of HD 2967 (salt sensitive) and Kharchia 65 (salt tolerant) magnetoprimed with static magnetic field dose of 50 mT for 2h were subjected to sodium chloride salinity stress from germination to maturity. Enhanced rate of germination and seedling growth under different salinity levels indicated that magnetopriming was effective in alleviating salinity stress at early seedling stage. Transient changes in α-amylase activity correlated well with changes in the rate of starch hydrolysis in germinating magnetoprimed seeds. In Kharchia 65, β- amylase activity contributed to starch metabolism during the early stage of germination. A 2.2 fold increase in the ratio of raffinose to sucrose in the magnetoprimed seeds of HD 2967 under saline conditions during the initial stages of germination suggested its role in seedling establishment. Glucose content could be detected only after 24 h of germination in both the genotypes when the activity of α amylase was high. Under salinity, the Na^+/K^+ ratio was less in plants from magnetoprimed seeds compared to unprimed seeds irrespective of the plant parts. The exclusion of sodium in primed seeds was beneficial in imparting tolerance to wheat genotypes under stress at the whole plant level. Increase in yield of plants from magnetoprimed seeds under non saline and saline condition is an integration of stimulation in growth and tolerance under salinity at all stages of growth. The response of salt sensitive variety HD 2967 was better than Kharchia 65 (salt tolerant) under saline conditions as magnetopriming reduced the adverse effects of salinity. Our results showed that magnetopriming of dry seeds can be effectively used as a pre-sowing treatment for mitigating adverse effects of salinity in wheat genotypes.

Salinity treatment affects various yield attributing characters, which ultimately resulted in severe yield loss in all the seven different genotypes of both *B. juncea* and *B. campestris*. It was observed CS-52 and CS-54 performed better under salt treatment in terms of lesser yield loss. Oxidative stress in terms of H_2O_2, superoxide radical production and lipid peroxidation increased with increasing salinity, however, CS-52 and CS-54 showed less oxidative stress. Activity of SOD, APX, GR. CAT and POX increased under salt treatment, especially in CS-52 and CS-54, which made them relatively tolerant towards salinity stress compared to Varuna, Pusa Jagannath and T-9. It was also evident that CS-52 and CS-54 showed lesser oxidative stress at later stages of its growth mainly due to their more efficient antioxidant defence system. The salinity induced increase in the contents of

total sugars, proline, trehalose and glycine betain was more in CS-52 and CS-54 than Varuna or T-9. Expression pattern of some key genes like *BADH*, *P5CS* and *T6PS*, which regulates the synthesis of these osmolytes was more in CS-52 and CS-54 under salt stress than Varuna or T-9. Soil salinity significantly reduced the potassium and calcium content in leaf, stem and ,root of all the cultivars. In contrast, the sodium content increased in all the plant parts under salt treatment. But, the changes were lesser in tolerant genotypes like, CS-52 and CS-54, which showed higher K/Na and Ca/Na ratio and thus more favourable cellular environment. Gene expression study reveals the existence of a more efficient *Salt Overly Sensitive* pathway composed of *SOS1*, *SOS2* and *SOS3* and vacuolar Na^+/H^+ antiporter in tolerant genotypes like CS-52 and CS-54 compared to Varuna and T-9. Sequence analysis of these partial cDNA also suggests the conserved nature of these genes as well their intra and inter generic relatedness.

Chickpea seeds of Pusa 1053 (kabuli) and Pusa 256 (desi) magnetoprimed with 100 mT 1h static magnetic field were subjected to salinity stress to evaluate the underlying mechanisms behind alterations in physiological parameters, growth and tolerance to salinity. Enhanced rate of germination and seedling growth parameters (root and shoot length and vigour indices) under different salinity levels in magnetoprimed seeds of Pusa 1053 in comparison to Pusa 256 indicated that magnetopriming was more effective in alleviating salinity stress at early seedling stage in Pusa 1053. Dynamics of seed water absorption in magnetoprimed seeds showed increased water uptake in Pusa 1053 under non saline conditions compared to salinity. This could have resulted in faster hydration of enzymes in primed seeds leading to higher rate of germination. At the biochemical level, the activity of amylase, protease and dehydrogenase was higher in primed seeds compared to unprimed seeds under both non saline and saline condition. Production of superoxide radicals was enhanced in germinating seeds of both the genotypes under salinity irrespective of priming. Increased levels of hydrogen peroxide in germinating magnetoprimed seeds, under both the growing conditions, suggested its role as a signaling molecule for the germination of the seed. Physiological traits were significantly improved in plants from magnetoprimed seeds of both the varieties under normal and saline (80 mM NaCl) conditions. Net photosynthetic rate recorded a significant increase in both genotypes under non saline and saline conditions. Stomatal conductance, transpiration rate and total chlorophyll content were also enhanced significantly under priming in leaves of Pusa 1053 compared to Pusa 256 under all treatments. Nodules from plants of magnetoprimed seeds exhibited an increased leghemoglobin content that enhanced nitrogen fixing ability of these plants as observed from enhanced nitrogenase activity. Increase in yield of plants from magnetoprimed seeds under non saline and saline condition is an integration of stimulation in growth and tolerance to salinity at all stages of growth. Our results evinced that magnetopriming of dry seeds can be effectively used as a pre-sowing treatment for mitigating adverse effects of salinity in chickpea genotypes.

Seed gamma irradiation at a very low dose (2.5 Gy) is known to enhance seedling establishment, plant growth and yield of cereals and other crops. The present study is aimed at assessing the interactive effect of pre-sowing treatment of seeds of two genetically diverse varieties of pigeonpea viz., Pusa-991 and Pusa-992 with gamma ray at 0, 0.0025, 0.005, 0.01, 0.02, 0.05 and 0.1 kGy on growth and development of pigeonpea under salt stress, imposed in the soil using 0, 80 and 100 mM NaCl (soil EC equivalent 1.92, 5.86 and 8.02 dSm^{-1} respectively) right from the inception of the experiment. Salt stress had negative effect on plant growth across the varieties and the radiation treatment. Seed gamma irradiation improved seedling establishment, shoot and root mass, leaf area and gas exchange characteristics such as net photosynthetic rate, stomatal conductance and transpiration rate under salt stress when compared with the respective non-irradiated control. Gamma irradiation caused favourable alterations in the source-sink (shoot-root) carbon (^{14}C) dynamics under salt stress in pigeonpea. Seed yield and seed protein and iron content were also positively affected by the low doses gamma irradiation under NaCl stress. An insight into the physiological and biochemical mechanisms underlying the radiation enhanced salt tolerance indicates a multigenic control with active involvement and favourable regulation either increase (+ve) or decrease (-ve) of water activity (+ve) membrane stability index (+ve), protease activity (-ve), shoot and root K^+/Na^+, glycine betaine content (+ve) and activity of antioxidant defence enzymes i.e., superoxide dismutase, catalase and peroxidase (+ve).

Exposure of chickpea genotypes to HT stress (6.01 ^{O}C) from 78 to 115 DAS (37 days) revealed that tolerant genotypes (Pusa 1103 and BGD 72) exhibited a comparatively higher SOD, APX and GR activity along with lower membrane injury index (MII), higher RWC, leaf pigments, Photosystem II yield and electron transport rate, proline and malic acid contents along with higher yield and yield attributes compared to susceptible genotypes (Pusa 256 and Pusa 261). Tolerant genotypes also showed better pollen viability, *in vitro* pollen germination percentage and *in vitro* pollen tube growth rate compared to susceptible genotypes. Under LS condition higher values of growth indices along with early phenology, lesser growing degree days, and better canopy air temperature difference were observed in tolerant Pusa 1103 and BGD 72 compared to susceptible genotypes. Tolerant genotypes also showed higher heat tolerance index and lower heat susceptible index along with better yield and yield attributes under LS condition. Significant increase in oligosaccharides content and decrease in grain protein content was observed under LS condition compared to NS condition. Under HT stress the bold seeded Pusa 256 possessed better seedling parameters compared to Pusa 1103 and BGD 72. Six hour heat stress (35 ^{O}C) led to significant reduction in protein band profile in all the three chickpea genotypes, while Pusa 1103 and BGD 72 showed significantly higher expression of protein bands compared to Pusa 256. It is suggested that selection of genotypes based on simple growth parameters and physiological parameters may help in evolving chickpea genotypes for high temperature stress tolerance with better yield.

Brassica juncea genotypes, subjected to temperature stress by growing the crops at three dates of sowing showed decrease in relative water content (RWC), total chlorophyll content and membrane stability index (MSI) at D2 and D3 sowings compared to the D1. Yield attributes, such as number of primary branches, secondary branches, no of pods per plant and seed yield also showed a similar trend as the level of temperature stress increased at D2 and D3 sowings. It was concluded that parameters like RWC, MSI and chlorophyll contents can be used as simple indices for screening and identifying temperature tolerant genotypes. Increase in the activity of antioxidant enzymes like superoxide dismutase (SOD), ascorbate peroxidase (APX), glutathione reductase (GR) and catalase (CAT) in tolerant genotypes viz., Proagro, CS 52 and NDR 8801 under D3 sowing suggest that the antioxidant defense mechanism plays an important role in the heat stress tolerance. Osmolytes also play major role in temperature stress tolerance in *B. juncea* genotypes. Temperature stress due to late sowing (D2 and D3) resulted in increase in contents of total sugar, proline, glycine-betaine and trehalose contents as compared to normal (D1) sowing in tolerant genotypes. Gene Expression studied by RT-PCR showed temperature stress induced increase in mRNA transcript of Δ-*pyrroline-5-carboxylate synthetase* (*P5CS*), *betaine aldehyde dehydrogenase* (*BADH*) and *trehalose-6 -phosphate synthase* (*T6PS*), which were again higher in Proagro, NDR 8801 and CS 52 compared to Pusa Agrani, EJ 15 and Pusa Tarak. Results suggested that high temperature induced expression of *P5CS*, *BADH* and *T6PS* in Proagro, NDR 8801 and CS 52 resulted in greater accumulation of osmolytes, which could be one of the reasons for their tolerance to temperature stress.

Water stress during wheat developmental stages limits production and its extent vary with stage at which plant encounters water stress. PGRs plays important role in plant response to water stress, cytokinin is important among them, it can induce water stress tolerance by delaying leaf senescence. The present study was conducted to determine the effect of cytokinin treatment under two different water regimes (Control and water stressed) in two contrasting cultivars, water stress tolerant C-306 and water stress susceptible PBW-343. In water stressed plants significant reduction were observed in traits related to photosynthesis which includes RWC, MSI, chlorophyll and carotenoid content, photosynthesis rate, stomatal conductance, photochemical efficiency and starch content. Cytokinin (BAP; 40 µM) treated plants were observed with higher activity of all the parameters studied in relation to photosynthesis and related traits in both the wheat cultivars under moisture deficit condition. Expression of Rubisco SSU was highest at anthesis stage but there was no significant effect due to cytokinin treatment under both water regimes. DHN and COR gene expression was more under water stress condition and expression was found to be increased during later developmental stages with no significant effect of cytokinin treatment. Nitrogen metabolism in plants is severely affected by water stress. There was decrease in activity of enzymes of nitrogen assimilation pathway viz., nitrate reductase and glutamine synthetase which led to decrease in plant total nitrogen and total protein content. However, application of cytokinin was found effective under water

stress to enhance activity of these enzymes and further increased leaf nitrogen and protein content in both the cultivars. Total protease activity was enhanced significantly during water stress condition but it was decreased by application of cytokinin. Under water stress condition there was significant decrease in leaf area and total biomass of plants which increased by treatment with cytokinin in both wheat cultivars. Yield related attributes also showed enhancement due to application of cytokinin in both the cultivars under water stress conditions. However, sensitive cultivars PBW-343 was found more responsive to cytokinin treatment under water stress condition in comparison to drought tolerant cultivar C-306.

India is the second largest producer of wheat in the world. High temperature spells towards the end of the crop season are a major determinant of wheat yield, especially under Indian conditions. Late sowing induced high temperature stress led to the reduction in relative water content (RWC), membrane stability index (MSI), chlorophyll content, carotenoid content and photosynthetic rate in all the varieties. The reduction was higher at anthesis and 10 days after anthesis, when compared to 30 days after sowing and booting. Comparatively higher reductions were observed in susceptible genotype, HD 2687 compared to Halna and DBW 14. High temperature stress induced oxidative stress, measured in terms of lipid peroxidation was found to be significantly higher under high temperature stress treatment especially more so in HD 2687. Crop duration, plant height, plant biomass, number of tillers, number of ears per plant and grain weight per panicle were also negatively affected by high temperature stress. The tolerant genotypes Halna and DBW 14 maintained comparatively high grain weight per ear, 1000 grain wt and grain yield per plant, under high temperature stress. Biochemical analysis of grains revealed that high temperature stress treatment led to significant reduction in starch content and increase in soluble sugar and grain protein content. However, Halna maintained optimal level of starch with little temperature induced increase in soluble sugars and grain protein in comparison with other genotypes. Expression analysis of heat shock transcription factors revealed that tolerant genotypes Halna and DBW 14 were having higher levels of heat stress induced expression of heat shock transcription factors *HSFA2b*, *HSFA4a* and *HSF8*. Halna and DBW 14 also maintained higher levels of expression of small heat shock protein (*sHSP*) like *17.3, 16.9B* and high molecular weight *HSP70*, *HSP80* and *HSP101C*. Sequence comparison of *HSPs* and *HSFs* revealed the homology of nucleotide sequences between *Triticum aestivum, Oryza sativa, Hordeum vulgare and Arabidopsis thaliana,* indicating the conservation of nucleotide sequences of HSPs and HSFs across genera. The amino acid sequences deduced from the nucleotide sequences also showed significant sequence homology across genera. From the study, it can be concluded that the efficient up-regulation of *HSFs* and *HSPs* played a major role in imparting high temperature stress tolerance in wheat.

Increase in temperature due to global warming can cause heat stress: a severe threat to wheat production, particularly when it occurs during reproductive and grain-filling phases. A controlled environment study was conducted with five wheat genotypes *viz.* PBW 550, RAJ 3765, KAUZ, HD 2733 and WH730. Average high temperature stress was created as day/night temperature of 32.4°C/15.5°C; 2.6°C/1.5°C above to ambient at the time of grain filling. This high temperature stress led to the reduction in growth cycle and yield related attributes in all the genotypes. High temperature stress decreased leaf area subsequently at different growth stages, and biomass and grain yield at harvest in all the genotypes. Also GGR and starch were significantly reduced but total sugar increased by high temperature stress during grain filling, irrespective of genotypes. However, under temperature stress KUAZ and WH 730 (relatively thermotolerant) showed lesser decrease in level of starch and a less increase in total sugar content, in comparison to other genotypes. The reduction in activity of soluble starch synthase (SSS) in KAUZ and WH 730 was lesser at 20 DAA when compared to 15 and 25 DAA suggesting higher expression of some isoform during that period of growth in these genotypes. A comparatively higher reduction was observed in susceptible genotype HD 2733 at same stage of grain filling (20 DAA). Expression analysis of *SSS* family members at 20 DAA, revealed that in tolerant genotypes expression of isoforms *SSSII* and *SSSIII* were at higher levels as compared to susceptible genotype. It can be concluded that the efficient up-regulation of different isoform of *SSS* at different stages of grain filling played a major role in imparting high temperature stress tolerance in tolerant genotypes.

Possibilities of foliar application of salicylic acid in improving or maintaining physiological traits in wheat under normal and late sown high temperature condition were explored. It was observed that the exogenous application of SA (0.1mM) efficiently maintained the physiological traits, by enhancing antioxidant enzyme activities and induction of gene expression of Hsps and photosynthesis genes (*TaHSP70*, Ta*psbA* and Ta*psbO*) while decreasing the H_2O_2 and TBARS contents when wheat plants were subjected to heat stress (39±2°C) during vegetative stage. During anthesis under late sown high temperature stress condition some parameters like lipid peroxidation (TBARS content), endogenous level of SA and ABA, RE of NSC and activities of antioxidant enzymes increased, however SA maintained the higher values of all other parameters which were otherwise reduced. Results of gene expression revealed exogenous application of SA plus high temperature enhanced the expression of both transcription factors (*TahsfA2b* and *TahsfA1a*) and large HSPs (Tahsp101, Tahsp16.9 and Tahsp17.8). From present study, it can be concluded that the exogenous application of SA (0.1mM) positively modulated the physiological and biochemical traits and transcription level of Hsfs and large molecular weight HSP analysed two days after foliar application. However, the positive effect on most of physiological and biochemical traits were not consistent as it decreased when analysed ten days after foliar spray. This clearly shows the response of SA is not long lasting or is transient in nature and it is also evident from yield, which was not significantly enhanced.

Different physiological parameters like plant height, plant girth, leaf area index, leaf firing, tassel blasting anthesis-silking-interval etc. were used to identify contrasting genotypes for heat-stress among twenty genotypes. Genotype DTPYC9F119 was found highly tolerant and K64R, highly susceptible to high temperature. Further, different physio-biochemical parameters for these two selected contrasting genotypes were analyzed under 6 days heat treatment (38/28^{0}C) to study the basis of heat stress tolerance. Chlorophyll a and b ratio under heat-stress increased significantly in DTPYC9F119 after 6 days high temperature treatment. Net photosynthetic rate reduced under high temperature in K64R but the reduction rate was relatively lower in case of DTPYC9F119. Activity of anti-oxidant enzymes (viz catalase, peroxidase and superoxide dismutase) and their gene expression increased in both the genotypes under heat condition.

Terminal high temperature stress is one of the major constraints for Chickpea production and may reduce its yield up to 50%. Bioregulators have been reported to play important role in alleviating detrimental effect of high temperature stress. Therefore, a field experiment was conducted using *kabuli* type chickpea varieties Pusa 1108 to test the possibilities of foliar application of abscisic acid (10 ppm), benzyl adenine (40 ppm) and salicylic acid (100 ppm) in improving biochemical parameters, growth, photosynthesis, dry matter partitioning and yield under late sown high temperature condition. High temperature stress was provided by delaying the sowing dates to expose the terminal phase of crop to high temperature. Values of all above parameters were reduced under late sown high temperature condition except lipid peroxidation, proline content, total soluble sugar, ABA content and activities of antioxidant enzymes. Interestingly, application of all bioregulators (ABA, BA and SA) in general maintained the higher values of all these parameters and reduced the lipid peroxidation. Moreover, ABA treatment improved the development of xylem vessels while BA and SA enhanced the development of secondary phloem and reduced ABA content. Present study revealed that bioregulators (ABA, BA and SA) application enhanced chickpea yield under late sown high temperature stress condition by improving growth, biomass partitioning, photosynthetic capacity and water use efficiency. Moreover, their application induced high temperature tolerance by restricting lipid peroxidation and enhancing the level of osmolytes and antioxidant enzyme activities.

High temperature stress at reproductive stage can decrease rice yield due to reduced pollen viability, and increased spikelet sterility. Three signaling molecules *viz*. salicylic acid (SA), calcium chloride (Ca) and brassinosteroid (BR) were exogenously applied at pre-anthesis stage to analyse their role in ameliorating high temperature effects on pollen viability, spikelet fertility, grain yield and quality attributes in two contrasting rice genotypes (Pusa Sugandh-5 and Nerica L-44). Three different concentrations of salicylic acid (0.1, 0.25, 0.5 mM), calcium (10, 50, 100 mM of $CaCl_2$) and brassinosteroid (0.5, 1, 1.5 ppm of 24-Epibrassinolide) were applied

and one set of plants was exposed to high temperature stress (36°C ±1.7°C) inside temperature tunnel. High temperature stress-induced physiological changes led to reduction in grain yield and quality in both the genotypes. Exogenous application of signaling molecules did ameliorate the adverse impacts of high temperature stress on yield and quality of rice, but the magnitude of change varied. In general, SA and Ca at lower concentrations and BR at all concentrations were effective in enhancing the thermo-tolerance. The ameliorative effect of signaling molecule for heat stress was more pronounced in PS-5 compared to Nerica L-44. Pre-treatment with lower concentrations of signaling molecules ameliorated the high temperature effects on pollen viability, spikelet fertility and quality traits and decreased the concentration of peroxidation products in both genotypes, while higher concentrations showed additive stress effects and caused more reduction than high temperature effects. Amelioration effect of SA at lower concentrations can be attributed to increase in the activities of CAT, APX and SOD in PS-5 while, GPX, APX and SOD activity increased in Nerica L-44. The activities of CAT, GPX, APX and SOD increased with Ca pre-treatment in both the genotypes under high temperature. In case of BR pre-treatment higher CAT, APX, GPX and SOD activities at all concentrations were measured in PS-5, but in Nerica L-44, the activity of CAT and SOD decreased. Higher expression of *HSP*101 in both the genotypes under high temperature supported the stress-induced changes in membrane stability. Similarly pre-treatment of signal molecules showed higher gene expression level suggesting the amelioration effects through these molecules. Thus, results of the present study offers a strategic option to minimize the adverse impact of heat stress on rice yield and quality in a changing climate. However, further studies are required to test the role of above signal molecules through pre-treatment of wide range of concentrations.

Ten rice genotypes which differed in their response to either high temperature (HT) or water-deficit stress (WS) at different phenological stages *viz.* Nagina 22, NERICA L 44, Shahbhagi *dhan*, Anjali, Vandana, PUSA Basmati 1, PUSA Sugandh 5, IR 64, Swarna and CR 262-were raised under controlled environment conditions and were subjected to high temperature stress (38°C), water-deficit stress and combined high temperature and water-deficit stress (HT+WS) at anthesis stage till the main tillers completed flowering. HT alone or in combination with WS, led to the reduction in relative water content (RWC), membrane stability index (MSI), panicle length and total spikelets per panicle in all the genotypes. The quantum yield of PS II (Fv/Fm) decreased significantly under HT, WS and HT+WS, accompanied by reduction in grain weight per panicle. Anther dehiscence reduced significantly leading to decrease in spikelet fertility (SF) under HT and HT+WS resulting in significant decrease in panicle harvest index (PHI). Higher reduction was observed in susceptible genotypes, Swarna and PUSA Sugandh 5 compared to Nagina 22 and NERICA L 44. Upland cultivars Anjali and Vandana showed susceptibility to HT and HT+WS. WS led to the reduction in RWC, MSI, panicle length and total spikelets per panicle in all the genotypes, however, reduction in MSI and panicle length was lower under WS compared to HT and HT+WS. Lower anther dehiscence under WS led to a lower SF, but the decrease was less compared to HT and HT+WS, suggesting a dominant influence of high temperature on spikelet fertility. Flowering duration increased in all the genotypes under WS and HT+WS, but significantly reduced under HT, suggesting a probable escape mechanism under HT. WS alone or in combination with HT significantly reduced peduncle elongation, trapping the panicles inside the leaf sheath and thereby exacerbating the effects of heat on trapped spikelets leading to enhanced spikelet sterility under HT+WS. Upland cultivars showed relative tolerance to WS compared to HT. Expression analysis of candidate genes from spikelet fertility QTL *(qtl1.1)* revealed that most of these genes were downregulated under HT, WS and HT+WS conditions, indicating a putative function of these genes working in tandem to regulate the development of reproductive organs. Comparison of sequences of these candidate genes between tolerant (Nagina 22) and susceptible (Swarna and PUSA Sugandh 5) *Indica* cultivars revealed no significant allelic differences in our study.

Role of putrescine in combating abiotic stress in rice was investigated in two contrasting cultivars NL-44 (tolerant) and PS-2 (susceptible). The plants were raised in pots and exposed to low and high temperature salinity and drought stress at panicle initiation and anthesis stages. Expression of *OsAdc1* and *OsAdc2* genes were studied and functional validation of the genes was attempted in *Arabidopsis thaliana.* PS-2 cultivar showed higher oxidative stress in terms of H_2O_2 and lipid peroxidation and lower MSI under all abiotic

stresses. *OsAdc1* and *OsAdc2* gene expression showed variable response under stress. Under salinity stress, there was no change in expression of both the genes but under low and high temperature and drought stress significantly higher expression of *OsAdc1* and *OsAdc2* was observed in both the genotypes. Highest expression of both the genes was found in Nerica L-44 under high temperature stress. Transgenics overexpressing ADC1 and ADC2 proteins were developed for further validation and four weeks old transgenic Arabidopsis plants were subjected to salinity (100mM NaCl) stress, water deficit stress (withholding water for 8 days), low temperature (4°C) and high temperature (35°C) stress. The transgenic lines showed improved putrescine biosynthesis and better tolerance to low and high temperature and drought stress but their performance was poor under salinity stress. The deduced aminoacid sequence of cloned gene and already reported one showed and 99.36% resemblance for ADC1 and ADC2 proteins, respectively. The identity between the two homologs of ADC proteins was found to be 45.96% due to the deletion of sequences from both C and N termini of ADC2. Multiple sequence alignment demonstrated that the deduced ADC protein sequence shared significant identity with ADCs from model organisms including several highly conserved motifs and amino acids. Finally, our results suggest that ADC plays a key role in under low and high temperature and drought stresses tolerance, which may be attributed to ROS detoxification and protection of cellular membranes under stress.

Water deficit stress during wheat developmental stages limits production and its extent vary with stage at which plant encounters water deficit stress. Plant growth regulators (PGR's) plays an important role in plant response to water deficit stress, can induce water stress tolerance by delaying leaf senescence. The effect of PGR's treatment was studied under two different water regimes (control and water deficit stress) in four contrasting wheat cultivars, C 306 and HD 2987 (relatively water deficit stress tolerant) and HD 2888 and HD 2733 (relatively water deficit stress susceptible) screened from twenty various known Indian and Mexican wheat genotypes. In water deficit stressed plants, significant reduction were observed in photosynthetic capacity in term of relative water content (RWC), membrane stability index (MSI), chlorophyll, carotenoid, photosynthesis, stomatal conductance, photochemical efficiency and starch. PGR's {thidiazuron (TDZ); 0.01 µl/L + paclobutrazol (PBZ); 30 µl/L + ascorbic acid (AsA); 100 µl/L} treated plants were observed with higher activity of all the traits studied in relation to photosynthesis and related traits in all the four wheat cultivars under both the water regimes. Rubisco (SSU) expression were studied during anthesis and post anthesis stages of wheat development and higher expression were observed at anthesis and PGR's improved the expression significantly under both the water regimes. PGR's applications down regulate the oxidative stress and improved the various enzymic as well as non- enzymic antioxidants activities. It was also observed that abiotic stress related genes such as DHN, AQP and P5CS expression was also enhanced under water deficit stress condition with PGR's treatment for better tolerance.

Interaction of ethylene and polyamines improves the abiotic stress tolerance in general and drought tolerance in particular. Thirty five wheat core germplasm was evaluated in pot culture and drought treatment was imposed by withholding water at anthesis stage. The fully grown flag leaf was used as the sampling material for all the physiological, biochemical and molecular studies except the phytohormone (ethylene and polyamines) which was studied in ten days old wheat seedlings. The studied genotypes showed significant decline in relative water content (RWC), membrane stability index (MSI), greenness index, photosynthesis, biomass, harvest index and grain yield under water deficit stress as compared to control condition. Three contrasting genotypes were selected on the basis of leaf senescence rate and drought tolerance efficiency from thirty five screened germplasm. Expression analysis studies revealed that in drought tolerant genotype, expression level of *TaSAMDC*, *TaSPDS* and *TaERS1* increased while, decreased level of *TaACO2*, *TaACS1*, *TaCHLase1* and *TaPAO* was observed under water deficit/ osmotic stress condition. Further, we also cloned and characterized the sequence of the genes involved in stay-green trait *viz.* chlorophyll degradation (CHLase), ethylene biosynthesis/ signaling (ACO, ERS), polyamine biosynthesis (SPDS) and their interaction (SAMDC homologs). In conclusion, the tolerant genotype maintained its water relation, antioxidant defense mechanism, stay-green and yield related traits, through upregulation of endogenous polyamine levels which act as positive regulator of drought tolerance.

The evidences proved the hypothesis that genetic manipulation of crop plants with genes encoding enzymes of polyamine biosynthesis pathway may provide better stress tolerance to crop plants under drought stress.

Moisture stress decreased germination percentage, root, shoot and coleoptiles length, vigour index, relative water content (RWC), membrane stability index (MSI), contents of chlorophyll (CHL) and carotenoids (CAR) at all the stages, leaf area at anthesis, and biomass and grain yield at harvest in all the genotypes. Moisture stress induced decrease in RWC, CHL, CAR, MSI, biomass and grain yield were significantly less in C 306 and highest in HD 2733 and PBW 343, while HD 2967, PBW 373, HD 3016 and NI 5439 showed intermediate response. Germination data was found to have good correlation with yield loss under moisture stress under field condition. Studies on the role of osmolytes in imparting moisture stress tolerance revealed that moisture stress induced increase in the contents of total soluble sugars; proline, glycine-betaine and trehalose were significantly greater in tolerant genotypes C 306 and HD 2987 in comparison to susceptible genotypes HD 2733, PBW 343 and HD 2967. Studies on the activity and transcript abundance of enzymes involved in osmolyte biosynthesis showed that moisture stress induced transcription level changes in *trehalose-6-phosphate synthase* (*T6PS*), *betaine aldehyde dehydrogenase* (*BADH*) and *pyrroline-5-carboxylate synthetase* (*P5CS*) resulted in increase in activity of trehalose-6-phosphate synthase, betaine aldehyde dehydrogenase and pyrroline-5-carboxylate synthetase, which ultimately led to increased accumulation of various osmolytes. Grain growth was found to be significantly reduced under moisture stress condition in case of drought susceptible wheat genotypes viz. PBW 343, HD 2733 and HD 2967 as compared to tolerant genotypes C 306 and HD 2987. Under moisture stress total soluble sugars increased and starch content decreased. But the moisture stress induced increase/decrease in sugar/starch contents were more in susceptible genotypes PBW 343, HD 2733 and HD 2967 as compared to tolerant genotypes C 306 and HD 2987. The activity of soluble starch synthase decreased in grain, while, sucrose phosphate synthase activity increased in leaf during moisture stress. The gene expressions of *SPSIIa*, in leaf tissues, and *SSI*, *SSII* and *SSIII* in grains also showed trend similar to the enzyme activities under moisture stress condition. The expression of *sucrose synthase* (*SuSy*) and *ADP-glucose pyrophosphorylase* (*AGPase*) in wheat grains, also decreased under moisture stress, and the moisture stress induced declines were more in susceptible genotypes. The results indicate that under moisture stress condition mobilization of sugars from leaf to developing sink may not be a limiting factor, however, their remobilization in the developing kernel and assimilation in to starch synthesis is greatly affected, resulting in small seed size and loss in yield; and the adverse effect was more in susceptible genotypes as compared to tolerant ones.

Drought is a major constraint in chickpea production. Therefore, present study was undertaken to test the possibility of application of some bioregulators in improving growth, biomass partitioning, yield and to gain insight into the mechanisms of bioregulators induced drought tolerance using two contrasting chickpea varieties viz. desi (Pusa 362) and kabuli (Pusa 1108). Water stress was imposed by withholding water at vegetative and reproductive stages. Just prior to water stress, plants were pre-treated with application of bioregulators (thiourea 1000 ppm, benzyladenine 40 ppm and thidiazuron 10 ppm). Based on preliminary experiment, optimum concentration of thiourea (1000 ppm), BA (40 ppm) and TDZ (10 ppm) were selected. The effect of bioregulators on germination under PEG induced water stress was also studied in lab. Our results showed decrease in RWC, MSI, chlorophyll, carotenoid contents, rate of photosynthesis, stomatal conductance, transpiration, chlorophyll fluorescence parameters, growth parameters, harvest index, drought tolerance efficiency, dry matter stress tolerance index, yield and its attributes and increase in respiration, under imposed water stress condition. Interestingly, application of bioregulators, (thiourea, BA and TDZ) in general, maintained higher values of all these parameters, maintained the integrity of leaf anatomy and chloroplast structure under imposed water stress and also stimulated faster recovery after the water stress termination. Lipid peroxidation and total free amino acids content increased under water stress but bioregulators application minimized their levels in both chickpea varieties. Total soluble sugars, proline content and antioxidant enzyme activities increased under imposed water stress and bioregulators treatment maintained these at a higher level and thus provided osmotic protection. In general, the ABA content gradually increased with imposed water stress and application of cytokinins maintained it at a lower level that in turn indicated the cross talk between ABA and cytokinins and suggested the role of cytokinins & ABA ratio in water stress tolerance.

Cytokinins increased the expression of rubisco, glycolate oxidase and glycine decarboxylase genes under both watered and water stress conditions. From the present study it can be concluded that all the bioregulators (thiourea, BA and TDZ) maintained photosynthetic activity and protected the photosynthetic machinery by enhancing the level of osmolytes, antioxidant enzymes activity, expression of rubisco and photorespiratory enzymes and thereby improving the yield of chickpea under water stress condition.

To understand the role of jasmonic acid in natural and induced senescence, endogenous levels of jasmonic acid was monitored either with or without exogenous application of jasmonic acid in mung bean cv. PS 16. Jasmonic acid levels increased in the leaves with the age of the plant and were found to be highest in senescing leaves at 55 days after sowing (DAS). There was a close association between jasmonic acid levels and ethylene biosynthesis in senescing as well as stressed leaves. To know the significance of elevated jasmonic acid levels an experiment was conducted with the exogenous application of jasmonic acid to the detached leaves. It was observed that jasmonic acid exerts significant influence on ethylene biosynthesis as well as on senescence characteristics of leaves. The influence of jasmonic acid over ethylene biosynthesis was concentration dependent and enzyme specific. In the Jasmonic acid treated leaves, the ACC synthase enzyme activity was increased even with 1 µM concentration. But thereafter increase in the concentration of Jasmonic acid appears to have not much effect on the enzyme activity. The ACC oxidase activity increased with increase in Jasmonic acid concentration. After registering its highest activity at 5µM Jasmonic acid, it showed a sharp decline at 50 µM Jasmonic acid. The senescence characters like loss of chlorophyll, increase in protease activity and decrease in the antioxidant enzyme activity were also observed in stressed and jasmonic acid treated leaves. Increase in endogenous jasmonic acid levels preceded the loss of chlorophyll in both control and stressed plants. Chlorophyll content in the leaves of Jasmonic acid treated plants started declining with an increase in the Jasmonic acid concentration. At higher concentration of jasmonic acid, though ethylene synthesis was not stimulated, chlorophyll loss did occur indicating that jasmonic acid induced chlorophyll loss is ethylene independent. The activity of protease enzyme increased with the severity of the stress as well as in senescing leaves. However in Jasmonic acid treated leaves the protease enzyme though increased at 1µM Jasmonic acid; there was not much increase in the protease activity at higher concentration of Jasmonic acid. There was a spurt in the activity of superoxide dismutase and catalase in response to stress and jasmonic acid treatment, however, the activity declined at higher JA concentrations. The results suggest that externally applied jasmonic acid may be causing stress induced senescence as the senescence characters like loss of chlorophyll, increase in the protease activity and decrease in the antioxidant enzyme activity were all promoted by the jasmonic acid treatment.

Chickpea being grown under rainfed conditions, drought is one of the major constraints in its production. Paclobutrazol (PBZ), has been reported to provide plant protection against drought. Two contrasting chickpea varieties viz. *desi* (Pusa 362) and *kabuli* (Pusa1108) and were subjected to two main treatments (well watered and water deficit condition) and four sub-treatments (PBZ foliar spray (60 ppm), PBZ drenching (120 ppm), water spray and water drenching). The results showed decrease in relative water content, membrane stability index, rate of photosynthesis, stomatal conductance, transpiration rate, Fv/Fm ratio, water use efficiency, chlorophylls, carotenoids, growth parameters, harvest index, drought tolerance efficiency, yield and its attributes and disrupted chloroplast grana under imposed water deficit stress condition. Applications of paclobutrazol maintained higher values of all these parameters and maintained the integrity of chloroplast structure under imposed water stress and also stimulated faster recovery after water deficit stress termination in both chickpea varieties. Biochemical analysis revealed that total soluble sugars, total free amino acids and proline content increased under imposed water deficit stress and paclobutrazol maintained these at higher level and hence has a role in osmo-protection. Paclobutrazol reduced the rate of lipid peroxidation and hence provided reduced membrane leakage. It also enhanced the activity of antioxidant enzymes and increased the level of abscisic acid and so provided stress tolerance ability to both chickpea varieties. In general PBZ drenching in terms of physio-biochemical responses was found better over the foliar application of PBZ. Present findings depict that PBZ application maintained

water relation, membrane stability, and photosynthetic activity and protected the photosynthetic machinery by restricting lipid peroxidation and enhancing antioxidant activities and thereby induced water deficit stress tolerance and enhanced the yield.

Cytokinins (CKs) induced staygreen (SG) trait and its association with drought tolerance was studied in wheat. Thirty five wheat germplasm were phenotyped for staygreen trait by imposing drought for 10 days after anthesis. CHIRYA7 and HW2041 were selected as functional staygreen genotypes (FSG) with low leaf senescence rate (LSR) and DSI and HW2033 and CBW38 were selected as non-staygreen genotypes (NSG). SG genotypes appeared to be drought tolerant than non SG genotypes in terms of maintaining higher chlorophyll content, photosynthesis rate, photochemical efficiency, stomatal conductance, transpiration rate, RWC and MSI. SG genotypes also shows competency to quench ROS due to enhanced activities of antioxidants. Further, the study was performed to learn more about the transcriptional regulation of cytokinin and its association with modulators and effector proteins involved in staygreen trait induced drought tolerance mechanism in wheat under water deficit stress environment. It shed a light on involvement of cytokinin and its signaling cascade-two component system in modulating the cytokinin response on delaying the drought induced leaf senescence and its association with drought tolerance mechanism in wheat. The identified staygreen genes/germplasm have the potential to be used in molecular breeding/ genetic engineering programme for the development of drought tolerant wheat.

Brassinosteroids (BRs) are natural plant growth regulators, which occur at low (nano molar to micro molar) concentration in all plant parts. Studies have shown that besides an essential role in plant growth, development and reproduction, BRs also exert anti-stress effects on plants. We found that maize plants treated with 24-epibrassinoloide (EBR), an active brassinosteroid are more tolerant to reproductive stage drought stress than untreated plants. Foliar application of EBR on drought stressed maize plants one week prior to anthesis significantly enhanced net photosynthetic rate, decreased transpiration rate, prevented chlorophyll and carotenoids degradation and maintained leaf water potential. A marked increase in root biomass was also observed in treated plants, which perhaps maintained plant water status under limited soil moisture conditions. Pre-anthesis application of EBR significantly increased total soluble proteins, glycine-betaine and specific activities of key antioxidant enzymes including superoxide dismutase (SOD), catalase (CAT) and ascobate peroxidase (APX). Our studies suggest that EBR possibly induced drought tolerance in maize by improving photo-assimilate availability to developing silk and preventing pollen from desiccation by maintaining plant water status and improved antioxidant system. EBR induced enhanced accumulation of osmolytes (proline, glycine betaine) perhaps prevented drought induced tassel blasting and increase in anthesis-silking-interval ((ASI).

Waterlogging resulted in rise in ethylene production upto 4 days of waterlogging in green gram genotypes, however, wild and tolerant genotypes showed increase upto 6 days of waterlogging. The rate of ethylene production was very less and slow in susceptible (Pusa Baisakhi and MH 1K-24) genotypes, while the tolerant (T 44 and MH 96-1) and wild (*Vigna luteola* and *Vigna sublobata*) genotypes recorded very high ethylene production. Under 6 days of waterlogging treatment, the roots of *Vigna luteola* alone showed a aerenchyma like structures in their cortical region.The total soluble, non-reducing and reducing sugars declined drastically during waterlogging stress in Pusa Baisakhi and MH 1K-24. The tolerant (T 44 and MH 96-1) and wild (*Vigna luteola* and *Vigna sublobata)* genotypes maintained higher concentrations of total sugars and non-reducing sugars, while the concentration of reducing sugar actually increased in tolerant genotypes upto 4 days of waterlogging treatment. The higher levels of reducing sugars in tolerant and wild genotypes corresponded with high activity of sucrose synthase, while the susceptible genotypes showed less activity of sucrose synthase enzyme during waterlogging stress, which further decreased with duration of treatment. Activity of alcohol dehydrogenase (ADH increased in tolerant and wild genotypes with duration

of waterlogging stress. However, its activity declined in susceptible genotypes, Pusa Baisakhi and MH 1K-24. *Vigna luteola* and T 44 showed greater *non-symbiotic Hb, cNR and NOS* gene expression and nitrate reductase activity than the susceptible genotype Pusa Baisakhi. Similarly *Vigna luteola* and T 44 showed greater NO production even on 8th day of waterlogging, which was much higher than susceptible genotype Pusa Baisakhi. This was also correlated with greater NR activity in these tolerant genotypes. Thus it can be concluded that plants have evolved various mechanisms to survive under adverse environment, and a combination of most of these contribute to their tolerance against waterlogging induced hypoxia/anoxia. The non-symbiotic haemoglobin-NO interaction and their role in regeneration of NAD and continuance of glycolytic cycle could be one of the major physiological factors providing tolerance to T 44 and *Vigna luteola* under waterlogging stress.

Acknowledgement

We wish to thank all the Faculty Members of the Division of Plant Physiology for extending support and cooperation in preparing the research contributions of M.Sc. and Ph.D. students made during the period of 2007-16.

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M.Sc. and PhD theses submitted by the students in the Division of Plant Physiology, ICAR-Indian Agricultural Research Institute, New Delhi 110012 during the period of 2007-2016.

School of Horticulture Sciences

Floriculture and Landscaping

Kanwar Pal Singh, Namita and Sapna Panwar

Introduction

Floriculture is increasingly regarded as a viable diversification from the traditional field crops because of higher returns per unit area. Though the art of growing flowers is not new to India, but large-scale commercial cultivation, protected cultivation is relatively new in India. Enormous genetic diversity, varied agro climatic conditions, versatile human resources offer India a unique scope for diversification in new avenues which were not explored to a greater extent. With the opening up of international market in the WTO regime there is a free movement of floriculture products worldwide. In this context each and every country is having equal opportunity for trade in each other's territory. Globally, more than 140 countries are involved in cultivation of floricultural crops. Among various countries Germany continues to be the highest consumer followed by Japan. India is having a better scope in the future as there is a shift in trend towards flowers and this can be gainfully exploited by country like India with high amount of diversity in indigenous flora. Flowers are intricately entwined in the social fabric of our nation and no function is complete without flowers. The domestic industry is growing at annual rate of 7-8% per annum. The area under flowers has crossed to 2.55 lakh hectares during 2013-14 which is concentrated mostly in Tamil Nadu, Andhra Pradesh, Maharashtra, West Bengal, Karnataka, Kerala, Himachal Pradesh and Uttarakhand.

The Indian Agricultural Research Institute is a pioneer in scientific research on floricultural crops spearheaded by luminaries like Dr. B.P. Pal who laid a foundation for scientific breeding of flower crops in late 50's and early 60's. The work on flower crops was initiated in the Division of Genetics and later in the Division of Horticulture. Ever since its inception as a separate identity in the year 1983, the Division of Floriculture and Landscaping heralded an articulated and comprehensive research programmes on crop improvement using conventional and biotechnology tools, production in open and protected environments, technology dissemination for the benefit of farming fraternity. The Division has following thrust areas:

- Molecular and morphological characterization of flower crops
- Inheritance studies in flower crops for growth and flowering traits
- Improvement of flower crops for qualitative and quantitative traits.
- Profiling of pigments from flower crops.
- *In vitro* techniques for mass multiplication, pigment induction and improvement in ornamental crops.
- Improvement and management of turfgrasses.
- Production technology for open field cultivation of cut and loose flower crops.
- Protected cultivation of flower crops for flower quality and regulation.
- Standardization of production technology in potted plants.
- Enhancing the postharvest life of flowers using novel approaches.

Significant Student's Research Achievements

The research achievements of the Division have been contributed significantly by the students sincerely guided by the experienced faculty members and the Professors of this Division. The research contribution of the students is discussed in theme-wise and presented.

Breeding of Flower Crops

Diversity in 32 genotypes of rose (Fig 1) both at genotypic and phenotypic level were carried out and analysis of variance for all the traits showed highly significant differences among genotypes. Phenotypic coefficient of variation (PCV) for all the characters was higher than the genotypic coefficient of variation (GCV). Moreover, high heritability (> 80 %) were noticed for plant height, internodal length, neck length, flower diameter, flower weight, number of petals per flower and number of flowers per plant (Sapna *et al.* 2012). A significant positive correlation at genotypic and phenotypic level was observed for flower weight with stem girth, leaf area, stalk length, neck length and flower diameter. (Sapna *et al.* 2012).

Fig. 1: Genetic diversity in *Rosa* spp.

The maximum direct contribution to number of flowers per plant was observed from the characters like stem girth, flower diameter, primary branches, days taken to flowering, bud length and number of petals per flower. On the basis of divergence analysis genotypes were grouped in seven clusters with range of 0.25 to 2.88 intra cluster distance. The genetic variability based on RAPD and ISSR markers was analysed among 32 rose genotypes. A total of 307 bands were detected using 20 RAPD and 10 ISSR markers, and number of bands per marker ranged from 6 to 18. The amplification product was in the range from 150bp to 3kb. The average polymorphic information content (PIC), resolution power of primer (RP) and marker index (MI) was 0.29 (± 0.06), 4.31 and 2.88 for RAPD markers and 0.36 (± 0.04), 6.00 and 3.96 for ISSR markers respectively. The similarity coefficient ranged from 0.50 to 0.91 showing the high level of divergence. UPGMA-cluster-analysis based on genetic distance coefficients clearly separated all the genotypes, and showed that Suryodaya and Suryakiran varieties are closely related (Sapna *et al.* 2015). A graphical representation of molecular marker data was done for selection and evaluation of rose genotypes. From the present data it is concluded that cultivated roses display a high level of genetic variability despite the fact that single morphological and physiological characters may be less polymorphic within rose groups. These results will guide new collection activities to establish larger collections and manage the rose genetic resources. The genotypes identified in this study may be directly useful for breeding.

In another study, genetic diversity studies were carried out in rose using morphological and Random Amplified Polymorphic DNA (RAPD) markers among twenty three genotypes (20 species and 3 varieties). For morphological characterization, 16 different morphological traits were evaluated. Jaccard's pair wise similarity coefficient was used to determine genetic diversity and relationship of rose species based on morphological characters. *R. damascena* (Rani Sahiba) and *R. moschata* showed maximum diversity. *R.*×*hybrida*and *R. borboniana*showed the maximum similarity. Dendrogram generated based on (UPGMA) divided the entire genotypes into four major clusters. Random Amplified Polymorphic DNA (RAPD) analysis was employed for the molecular characterization of these genotypes. Out of 50 RAPD decamer primers used, 26 primers produced 169 scorable bands, of which 168 were polymorphic.

Genetic similarity between the rose species was determined on the basis of Jaccard's pairwise similarity coefficient. *R. moschata* and *R. tomentosa* showed highest similarity. *R. nitida*×*R. rugosa* and *R. slancensis* fell in

the same cluster with morphological and molecular characterization (Henuka *et al.* 2015). Characterization by both means revealed considerable diversity in *R. damascena*. *R. damascena* var. Rani Sahiba and *R. damascena* var. Himroz fell in the same cluster and have shown divergence from *R. damascena* var. Jwala in morphological and molecular characterization. In agreement with Rehder's classification, members of Synstylae section; *R. brunonii, R. multiflora* and *R. wichurana* were grouped in the same cluster. The members of the Section Indicae; *R. indica var. odorata, R. indica major, R. chinensisviridiflora* and *R. borboniana* were also grouped together by Molecular characterization (Fig 2). Kakinada rose a local variety of South India has shown closeness with *R. damascena* var. Himroz by morphological and molecular characterization. Both morphological and molecular analysis showed a high degree of variation among analyzed germplasm indicating an important source of genetic diversity that can be used in the crop improvement.

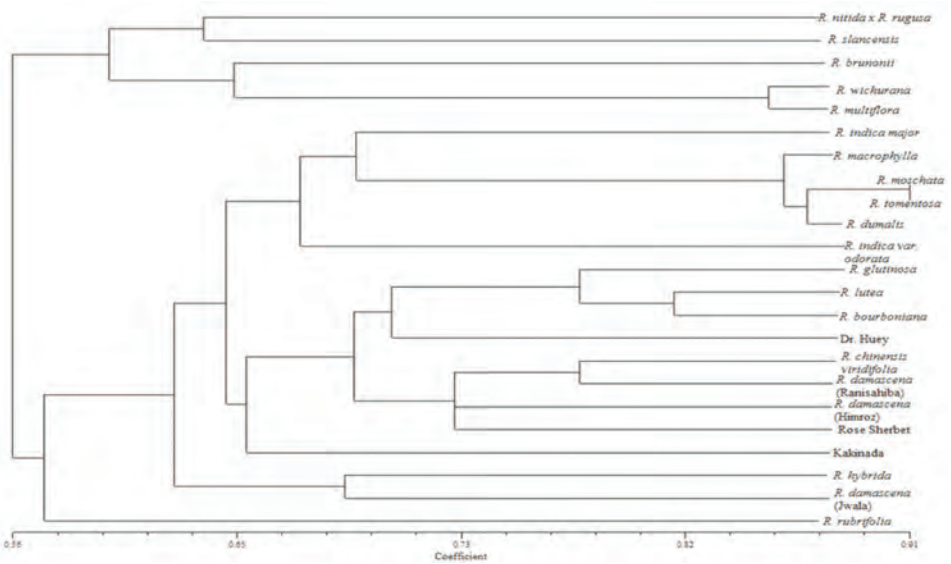

Fig. 2: Dendrogram based on the similarity index values of *Rosa* species using RAPD markers.

The investigations were undertaken to develop protocols for efficient *in vitro* regeneration in two rose cultivars i.e. Pusa Mohit and Raktima and to induce mutants with novel traits through *in vitro* mutagenesis (Fig 3) (Madhu Bala *et al.* 2013). Best culture establishment and shoot proliferation with quality micro shoots were obtained on MS medium supplemented with BAP(3.5 mg/l) +NAA (0.1 mg/l) + GA_3 (0.5 mg/l) in cv. Pusa Mohit and MS medium supplemented with BAP(3.0 MG/L) + NAA (0.1 mg/l) + GA_3 (0.5 mg/l) in cv. Raktima. Better quality longer shoots were obtained on MS medium supplemented with GA_3 (0.5 mg/l) in both cultivars. Half strength MS medium containing NAA (0.5 mg/l) + IBA (0.5 mg/l) was found to be better for root induction. For induction of mutations axillary bud explants of cvs. Pusa Mohit and Raktima were exposed to different gamma ray doses (5 to 80 Gy) and cultured *in vitro* on these standardized medium. In the second approach, *in vitro* proliferated cultures derived from non - irradiated axillary bud explants of these cultivars were treated with 5 to 25 Gy doses of gamma rays. Gamma

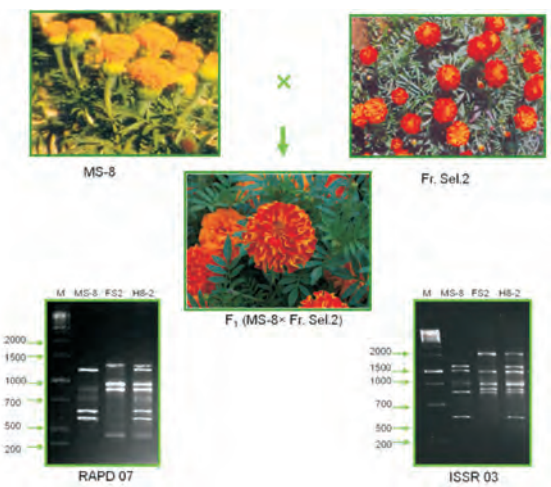

Fig. 3: *Invitro* mutagenesis in *Rosa hybrida*

ray doses from 20 Gy to 55 Gy were found to be effective in inducing the flower colour variants in these cultivars. As a results, ten putative mutants numbered as PM-1, PM-2, PM-3, PM-4, PM-5, PM-6 (from cv. Pusa Mohit) and RK-1, RK-2, RK-3, RK-4(from cv. Raktima) were isolated from plant population derived from irradiated explants/cultures. Higher doses of gamma rays (65, 70 and 80 Gy) proved to be detrimental for the explants resulting in very poor or non survival of explants.

To develop inter-specific F_1 hybrids in marigold (*T. erecta* x *T. patula*), a line x tester analysis was carried out using three male sterile lines of African marigold (*Tagetes erecta*) namely MS-5, MS-7 and MS-8 and eleven pollen parents of French marigold (*Tagetes patula*) numbered as French Selection1 (Fr. Sel.1), Fr.Sel.2, Fr.Sel.3, Fr.Sel.4, Fr.Sel.5, Fr.Sel.6, Fr.Sel.7, Fr.Sel.8, Fr.Sel.9, Fr.Sel.10 and var. Cherry Red to study the combining ability, exploitation of heterosis and to test the hybridity of promising hybrids using molecular markers. Among 33 hybrids developed, six higher yielding hybrids in terms of flower yield were MS-8 x Fr. Sel.1, MS-7 x Fr. Sel.1, MS-8 x Fr. Sel.2, MS-5 x Fr. Sel.1, MS-5 x Fr. Sel.2 and MS-7 x Fr. Sel.2. All the traits except duration of flowering exhibited high heritability in broad sense.

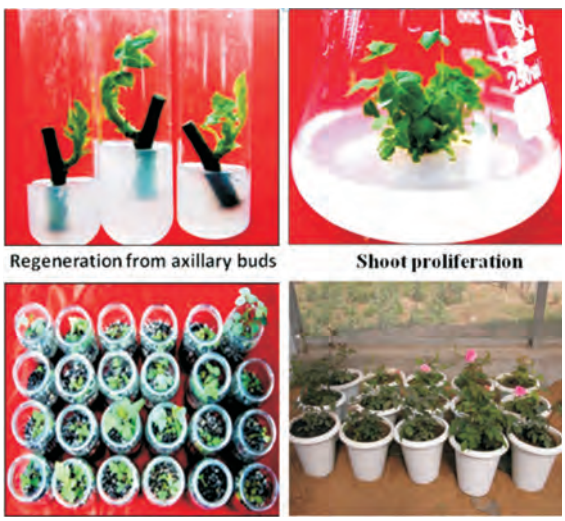

Fig. 4: Hybridity testing using RAPD and ISSR markers

Flower yield, flower weight, number of flowers per plant, stalk length and plant spread possessed greater estimates of genetic advance as per cent of mean coupled with high amount of heritability genotypic coefficient of variation and phenotypic coefficient of variation (Namita *et al.* 2009). Duration of flowering had highly significant positive correlation with flower yield followed by number of flowers per plant, plant spread, harvest index, flower size, flower weight and stalk length. The duration of flowering had highest direct effect (genotypic and phenotypic) on flower yield followed by number of flowers per plant and stalk length. Among male sterile lines, MS-8 and among the pollen parents, Fr. Sel. 1 and Fr Sel.2 were the best general combiners for all the characters (Namita *et al.* 2011). The crosses MS-5 x Fr. Sel. 1, MS-7 x Fr. Sel. 1, MS-7 x Fr. Sel. 2, MS-8 x Fr. Sel. 1 and MS-8 x Fr. Sel. 2 were the best heterotic combinations (in terms of better parent and mid parent heterosis) for flower yield. Genetic diversity was also studied among pollen parents of marigold (Namita et al. 2013). Six hybrids were verified using RAPD and ISSR primers and these were classified into three types (markers specific to male, markers specific to female and markers present in both the parents) according to the presence / absence of bands (Fig 4) (Namita *et al.* 2008).

In order to develop inter-varietal hybrids in marigold, the genetic variability, heritability, genetic advance, character association and path analysis for 16 quantitative characters were carried out in twenty-two genotypes of marigold. High heritability with high genetic advance as per cent of mean was recorded for traits stalk length, flower diameter, fresh weight per flower, flower yield per plant and 1000 seed weight. The correlation coefficient between yield and its components indicated that statistically significant and positive correlation (genotypic and phenotypic) was observed for flowering duration, number of flowers per plant, flower diameter, fresh weight per flower and dry weight per flower with flower yield. Path coefficient analysis revealed that fresh weight per flower had the highest genotypic positive direct effect on flower yield per plant followed by number of flowers per plant, stem diameter, number of primary branches and flower diameter (Sapna *et al.* 2014). Cluster analysis grouped genotypes into seven clusters. These genotypes were further analyzed by RAPD, ISSR and URP marker system. In molecular analysis the degree of polymorphism detected by RAPD, ISSR and URP markers were 98.8%, 92.73 % and 93.08 % respectively. In present study, polymorphism information content (PIC) for RAPD, ISSR and URP was recorded as 0.38, 0.34 and 0.32 respectively. The resolving power (Rp) was obtained as 8.36,

11.20 and 13.75 respectively for RAPD, ISSR and URP markers and the marker index (MI) obtained was 3.44, 2.93 and 3.17 for RAPD, ISSR and URP markers. The individual marker system did not clearly differentiate between male sterile and male fertile lines. When dendrograms generated using RAPD, ISSR, URP and combined data (RAPD, ISSR and URP) were compared, the discrimination among genotypes within these clusters was more effective with the combined analysis. Fifteen genotypes were selected based on molecular diversity and further used in hybridization programme. A line x tester analysis was carried out using molecularly diverse 15 genotypes which includes three male sterile lines namely MS-5, MS-7 and MS-8 and twelve pollen parents of African marigold *(Tageteserecta)* which included 10 selections i.e. African Selection1 (Af.Sel.1), Af.Sel.4, Af.Sel.5, Af.Sel.6, Af.Sel.8, Af.Sel.10, Af.Sel.11, Af.Sel.12, Af.Sel.14, Af.Sel.16 and 2 varieties Pusa Narangi Gainda and Pusa Basanti Gainda to study the combining ability, exploitation of heterosis. Estimates of GCA effects showed that the female parent MS-8 and pollen parents *viz.*, Af.Sel.1, Pusa Narangi Gainda, Af.Sel.5, Af.Sel.4, Af.Sel.8 and Af.Sel.10 were the best general combiners for carotenoids and other flower yield related traits. The female parents MS-7 and MS-8 and pollen parents *viz.*, Pusa Narangi Gainda, Af.Sel.5, Af.Sel.11, Af.Sel.8, Pusa Basanti Gainda and Af.Sel.10 were the best general combiners for flower yield and its related traits. The cross MS-5 x Af.Sel.12, MS-8 x Af.Sel.4 and MS-8 x Pusa Narangi Gainda exhibited high SCA for total carotenoids whereas the cross MS-5 x Af.Sel.10, MS-5 x Pusa Narangi Gainda and MS-5 x Af.Sel.6 exhibited high SCA for flower yield per plant and most of the growth and flower yield related traits. The hybrids MS-8 x Af.Sel.6, MS-8 x Af.Sel.6 and MS-8 x Af.Sel.1(in terms of better parent, mid parent and commercial check) exhibited highest heterosis for total carotenoids and MS-5 x Af.Sel.10, MS-5 x Af.Sel.10 and MS-5 x Pusa Narangi Gainda for flower yield per plant (Fig 5) (Sapna *et al.* 2013).

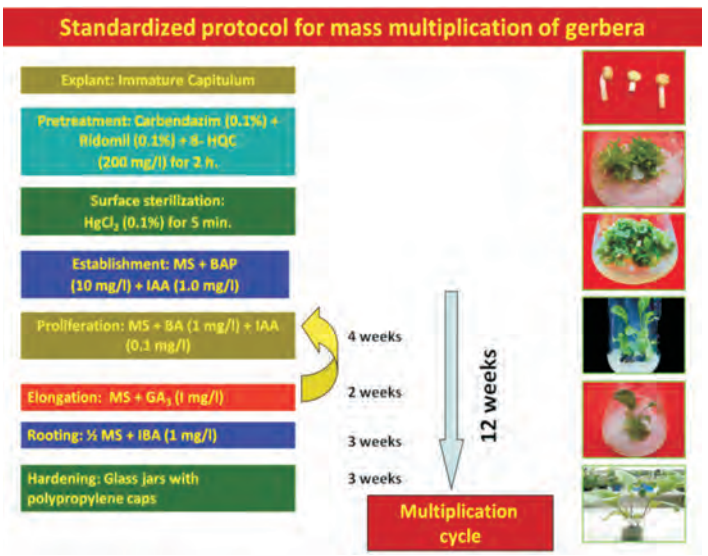

Fig. 5: GMS based Inter varietal hybrids in marigold

Production Technology

A study was conducted to find out the optimum explant source, culture media for efficient regeneration of gerbera *(Gerbera jamesonii)* and to perform genetic analysis of regenerants using DNA markers. Three explants *i.e.* capitulum, leaf and shoot tip of gerbera cv. Cabana were used in the present investigations. Pre-treatment of different explants with Bavistin (0.1 %) + Ridomil (0.1 %) + 8-HQC (200 mg/l) for two hours resulted in minimum culture contamination and maximum explant survival. Surface sterilization of leaf explants with 0.1 per cent mercuric chloride for three minutes and capitulum and shoot tip explants for five minutes gave the maximum number of uncontaminated growing cultures. Best establishment of capitulum explants was obtained on modified MS medium (vitamins increased to ten times) supplemented with 10mg/l BAP + 1mg/l IAA. Among the two stages, capitulum explants cultured at immature stage responded better than the mature stage. In leaf explants, MS medium supplemented with 10mg/l BAP + 2 mg/l NAA was found best for callus induction and MS medium supplemented with 3 mg/l BAP and 0.5 mg/l NAA was found suitable for shoot regeneration from callus. The best culture establishment from shoot tip explants was obtained on MS medium supplemented with 5 mg/l BAP + 0.5 mg/l IAA. MS medium supplemented with 1 mg/l BAP + 0.1 mg/l IAA was found optimum for proliferation of quality shoots. Among the three explants, capitulum explants exhibited the highest proliferation, whereas, shoot tip explants showed the minimum proliferation rate. Half strength MS medium supplemented with 1 mg/l IBA

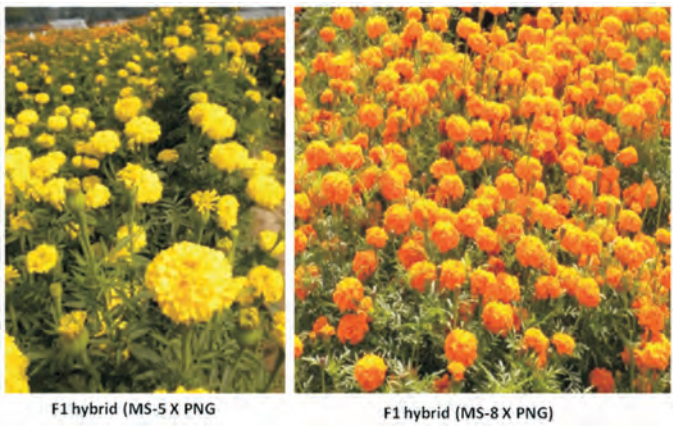

Fig. 6: Protocol for mass multiplication of gerbera

was found best for root induction. Hardening of rooted plantlets was most successful when done in glass jars with polypropylene caps (Fig 6) (Bhatia *et al.* 2012). ISSR markers were more potent than RAPD in detecting variation in the tissue culture raised plantlets. No polymorphism was detected among the mother plant and *in vitro* raised clones derived from capitulum, shoot tip and leaf explants during RAPD analysis, whereas, out of fifteen ISSR primers, twelve primers showed monomorphic banding pattern within *in vitro* raised clones comparable to the mother plant, however, polymorphic bands were detected in leaf derived clones by three ISSR primers (Bhatia *et al.* 2009).

In vitro studies were conducted to explore the response of ray floret explants of *Chrysanthemum morifolium* cv. Pusa Anmol for callus induction and multiplication, treatment combination of Murashige and Skoog' (MS) medium fortified with NAA(1mg/ltr+BAP (1mg/ltr) was found to be the best with respect to days for callus initiation (8.25±0.25) and induction coefficient percentage (94.51±0.40). Sucrose at 204.5 mM in the maximum induction coefficient percentage (79.07±1.10). When the chrysanthemum ray floret callus were cultured on MS medium fortified with different concentrations of the $GA_{3,}$ all the treatments failed to biosynthesize the anthocyanins. Calluses cultured on MS medium fortified with $15mgl^{-1}$ $CoCl_2$ exhibited maximum induction coefficient percentage (85.35± 2.09) and took minimum days (7.50±0.65) for anthocyanin biosynthesis with maximum colour value index (PC=2.48±0.09CV^{g-1}FCW:, PP = 0.74±0.02 CV test tube^{-1}). MS+$NiCl_2$ (10mg/1) medium exhibited maximum induction coefficient percentage (65.42±1.85) with highest colour value index (PC=1.68±±0.11 CV $^{g-1}$FCW:, PP=0.39±0.03 CV test tube^{-1}) but cultures on nickel chloride did not exhibit satisfactory results as compare to $CoCl_{2.}$ Amongst the elicitors tested [salicylic acid (SA), methy1 jasmonate (MeJA), abscisic acid (ABA)], 0.5 µm MeJA were most effective in inducing anthocyanin biosynthesis in calluses. MS media fortified with 0.5 µM MeJA resulted in maximum response coefficient (92.42±1.12) per cent, with highest PC (3.20±0.11 CV $^{g-1}$ FCW) and PP (1.45±0.05 CV test-tube^{-1}). Incorporation of 200 µMSA in MS medium also resulted in good response coefficient percentage (82.84±1.87) with higher colour value index (PC-2.68±0.10CV $^{g-1}$FCW; PP= 1.19±±0.04CV test tube^{-1}).

Study was conducted to explore the response of leaf explant of *Petunia hybrida* cv. Bravo Blue. Among different cultivars selected, Bravo Blue was found to be richest in anthocyanin pigment content (106.15 mg/g fresh weight). Leaf explant of Bravo Blue was found to be the most suitable explant. It gave maximum callusing frequency (73.33 ± 3.33) in minimum number of days (8.88 ± 0.16) with minimum microbial load (19.80 ± 1.57). For callus induction and multiplication, treatment combination of Murashige and Skoog medium (MS; 1962) supplementated with indole-3-butyric acid (IBA) + kinetin (Kin) + adenine sulphate (AdS) {(19.60 + 4.65 µM) + (81.45 mM)} was found to be the best in respect of days to callus initiation, induction coefficient percentage, and growth status of callus. Combination of IBA + Kin+ AdS {(19.60 + 4.65 µM) + (108.6 mM)} was also found good for the maximum gain in fresh- and dry weight with the highest FW/DW ratio (12.651 ± 0.46).Earliest callus induction (9.82 ± 0.39 days) and good callusing percentage (96.79 ± 0.77) was found in the complete darkness in leaf explant. Glucose 5% when used as carbon source was found to have earliest anthocyanin pigment induction with maximum response coefficient with highest pigment content (1.36 ± 0.012 CVg^{-1} FCW). The petunia leaf calli when cultured on MS medium supplemented with GA_3 30µM/1 exhibited maximum response coefficient in minimum number of days with highest colour value index, in respect of PP and PC. Highest response coefficient, earliest anthocyanin induction with maximum colour value index was found on the supplementation of 15 mgl^{-1} cobalt chloride in standard MS media. Cultures on nickel chloride did

not exhibit satisfactory results. The highest response coefficient percentage was recorded when the explants were cultured on FeEDDHA @ 60 µM/l with maximum pigment content and production. Under continuous light conditions, maximum elicitation in pigment content and production was observed. Nutrient stress conditions resulted in improvement of pigment content and pigment production. When the phosphate concentration in MS medium was reduced to quarter strength, highest colour index value was observed with maximum response coefficient (Usha *et al.* 2014).

Production Technology of Flower Crops and Cut Greens

Studies were carried out to find out the reaction time for flower induction in chrysanthemum under greenhouse in IARI developed cultivars. The results revealed that the cultivar having shorter reaction time expressed more antioxidant enzyme activities. Shortest reaction time was observed in cultivar Pusa Sona and Pusa Aditya, while it was longest in Pusa Chitraksha. Gaseous exchange parameters like photosynthesis and internal CO_2had higher values correlated with short reaction time and *vice-versa*. In respect to effect of extended day length extension in growth and flowering behaviour of *Chrysanthemum morifolium* cv. Zembla was examined in growth chamber and greenhouse. Artificial long day (15 hrs light @ 110 µmol sec^{-1} m^{-1}) period was provided for 0 to 18 days based on treatment in growth chamber. Immediately after transplanting control plant was placed under short day condition in polyhouse, and sequentially based on treatment different batch of plants transferred under short day conditions until flowering. Plant treated with photoperiod was found maximum stem length, which was suitable for export owing to good quality cut flowers. Plant growth parameter like crop growth rate, net assimilation rate and relative growth rate show linear growth up to 30 days after planting, and then decrease till flowering. Stem, leaf and flower dry matter accumulation was highest with 18 days photoperiod treatment. For achieving longer stem length and off-season flower forcing with GA_3 gave significant results under greenhouse. In general, it was observed that increase the GA_3 concentration from 50 ppm to 300 ppm, increment of plant growth. Result clearly shows that 300 ppm GA_3show increased maximum plant height (35.09 cm), internodal length (1.37), leaf number (12.3), leaf area (173.41 cm^2), and leaf area index (0.44) compared to control.

Another experiment was conducted on chrysanthemum cv. Zembla in two parts under different photoperiodic treatments of day length extension illuminating from high pressure sodium (HPS) and photosynthetically active radiation from LEDs for 6, 9, 12, 15 days @ 15h/day under growth chamber. In first experiment high pressure sodium bulbs were used whereas in the second experiment, light intensity was fixed by making two different LED panels, one composed of 840 white LEDs and in another a mixture of red (672) and blue (168) LEDs made into a strip backed with power @14.4 W/M, voltage input DC 12V and rating IP22 controlled with an adaptor AC 110~220V, 12V 5Amp, glued together in an area of 1.0 m x 1.0 m panel to achieve an interception of light @ 110-120 µmol $m^{-2}sec^{-1}$ from 80% red 20% blue PAR distributed over the plants in uniformity inside the growth chamber. LEDs light exposure was given for 15 hours as log day (LD) treatment except for control. Every batch after exposure @ 6, 9, 12, 15 days under was transferred to the chamber with white LEDs. Among different growth parameters (plant height, number of leaves, internode length, leaf chlorophyll, leaf area, fresh and dry weight of leaf, net photosynthetic rate, stomatal conductance varied significantly during different stages of growth and found that the increase in photoperiod by 15 days using high pressure sodium (HPS) lamp resulted in a significant increase in stem elongation, leaf number. Increased plant height (46.25 and 42.70 cm), inter-nodal length (2.70 and 2.63 cm) and leaf number (32.75 and 29.75) were observed in cv. Zembla due to long day extension treatments as compared to the control. Total fresh and dry weights of the plants were maximum for plants under 15 day length extension treatment. Total chlorophyll (3.65 and 3.45 mg g^{-1} FW), net assimilation rate, RGR, CGR and LAI values were recorded higher in treated plants compared to control. In another experiments with Long days from LEDs there was an increase in plant height (26.30, 34.30 cm), inter nodal length (2.60, 2.38 cm), number of leaves (24.50, 23.50) and leaf area (197.35, 188.90 cm^2) cv. Zembla maximum (15 days) exposure to LEDs. Early flower response was noticed which was advanced by 6 days as compared with control. Bud and flower diameter (5.78 mm and 4.71 mm) were maximum. Fresh and

dry weight in stems and flower were significant which might have contributed with increased total chlorophyll (4.33 mg g^{-1} fw) content as application of light emitting diodes (LEDs) might resulted in long day effect without disturbing the required minimum dark period for flower induction.

Experiments were conducted on integrated weed management in gladiolus cv. Pusa Srijana with a set of eleven treatments. The results revealed that the number of monocot and dicot weeds, their fresh and dry weight was recorded maximum under control treatment while these were least with the application of atrazine 0.75 kg/ha pre-emergence + carfentrazone @ 0.030 kg/ha post-emergence at 40 days after planting. The maximum vase life of spikes (10.3day) was recorded with the application of pendimethalin 0.75 kg/ha + metribuzin 0.3 kg/ha pre-emergence as compared to control (7.3 day). The yield of corms (81.52 q/ha), cormels (7.96 q/ha) and marketable spikes (1.43 lakh per ha), net profit (Rs 3, 48,694 per ha) and benefit cost ratio (1.99) was received maximum with the application of metribuzin 0.4 kg/ha pre-emergence + residue (dry grass 5.0 tones/ha) over control. The uptake of N P and K through gladiolus were recorded maximum in weed free check i.e. (four hand weeding), while N, P and K uptake through weeds were recorded maximum in weedy check i.e. (control) (Fig 7).

Fig. 7: Influence of herbicides on gladiolus

The research programme was formulated to standardize the shade levels and coloured nets for *Cordyline terminalis* and *Dracaena fragrans*. In the first experiment, plants were raised under green shade net with different shade level *viz.* 35%, 50%, 75%, 90% and control (open condition) while in the second experiment, green, black, red and white coloured net were used along with outdoor environment (Control). Different weather parameters, light intensity, photosynthetically active radiation, transmittance, canopy temperature, gas exchange properties, crop growth parameters were observed during different stages of growth. Shade nets significantly modified the micro environment. The air temperature, light intensity, canopy temperature, photosynthetically active radiation and transmittance were found to be lower under shade nets. The relative humidity increased with shading intensity. The shade levels of 50% were optimal for both the cut greens as they produced plants with highest plant height, number of leaves and petiole length with good harvest index and vase life (Abhay *et al.* 2015). Among coloured shade net, white shade net has been found to be best for *Cordyline terminalis* as plant height, inter-node length, photosynthesis rate, PS 2 efficiency and other important characteristics were superior while same can be said for red coloured net in case of *Dracaena fragrans*. So it can be concluded that red or white coloured net with 50% intensity provide optimal conditions for superior performance of cut greens.

Post Harvest Technology and Value Addition in Flower Crops

In the present investigation gladiolus flowers which are ethylene insensitive were used as a system to investigate the physiological and biochemical changes associated with activity of antioxidants both enzymatic and non enzymatic, protein degradation and protease activity, and lipid peroxidation and lipoxygenase activity during ethylene independent floral senescence. Thirty gladiolus varieties were evaluated for their keeping quality in the field (Field life) and also under laboratory conditions (*in vitro*) during 2010-11 in the research farm of Directorate of Floricultural Research, New Delhi. It was found that the varieties, Big Time Supreme and Jackson Ville Gold recorded maximum keeping quality in terms of field life and vase life in the laboratory (15.67, 13.67 days and 13.67, 12.67 days, respectively). Minimum keeping quality was observed in the varieties Purple Flora (7.33 and 5.67 days) and White Prosperity (8.00 and 6.33 days). The protease activity was reduced with the treatment of protease inhibitors maximum reduction being with the use of 5-SSA. Similarly the protein content was maximum in the treatment with 5-SSA. There was a strong negative correlation between the protease activity and protein content with the use of protease inhibitors. In these four varieties, the effect

of plant growth regulator (5-sulfosalicylic acid, 5-SSA) and protease inhibitors (N-ethylmaleimide, Aprotinin, Leupeptin and E-64 along with a control) on physiological and biochemical changes during senescence of gladiolus flowers was investigated. Superoxide dismutase, catalase, peroxidase, ascorbate peroxidase, and ascorbic acid were increased from stage to stage I still increase thereafter showed sudden and sharp decline in stage IV and stage V. The increase from stage II to stage III was rapid and high in magnitude compared to the increase from stage 1 to stage 2. Lipoxygenase (LOX) and lipid peroxidation showed similar trend of increase from stage 1to stage 3 and remarkable increase in stage 4 and 5. Among all the treatments, 5-SSA was found to significantly increase the activity of all enzymatic antioxidants studied as well as the activity of ascorbic acid .Simultaneously this treatment resulted in decrease of LOX activity and lipid peroxidation compared to other treatments. N-ethylmaleimide was also found to be effective in increasing the activity of antioxidants and in decreasing the activity of LOX and lipid peroxidation. The varieties which showed higher vase life *i.e* Big Time Supreme and Jackson Ville Gold recorded higher activity of antioxidants both enzymatic and non enzymatic compared to low vase life varieties, White Prosperity and Purple Flora. Among the varieties, a reverse trend to that of antioxidants was observed in respect of LOX activity and lipid peroxidation.

A study was conducted to screen out the cultivars of cut tuberose for longer vase life on the basis of Membrane stability index (MSI). Five cultivars of tuberose namely Mexican Single, Phule Rajini, Prajwal, Shringar and Vaibhav were taken for screening. Leakage of ions was measured over five arbitrarily divided stages of the flower development in tuberose. Stage 1, tepals start reflexing during bud opening; Stage 2, half bloom; Stage 3, full bloom; Stage 4, beginning of wilting and Stage 5, complete wilting. There were declining trend in MSI value irrespective of whether they are Single type or Double type cultivars. There was non significant difference in membrane stability index during their first and second stage of flowers development, but showed significant difference at later stages of flower development. Vaibhav had highest MSI value (0.430) throughout the developmental stages of flower as compare to other cultivars used in the experiment. However, Mexican Single showed comparatively lower value of MSI at Stage5 (0.340) which indicate that leakage of ions through membrane is significantly high. There was non significant difference between the cultivar Phule Rajini, Prajwal and Shringar. Further experiment was carried out to study the effect of salicylic acid and 5-Sulphosalicylic acid applied alone or in combination with sucrose in vase solution to study their effect on vase life and other associated parameters in tuberose cultivars Vaibhav and Mexican Single. Treatments used were: Control (distilled water) (T1); Sucrose (4%) (T2); Salicylic acid (50 ppm) (T3); Salicylic acid (100 ppm) (T4); Salicylic acid (100 ppm) + Sucrose (4%) (T5); 5-Sulphosalicylic acid (200 ppm) (T6); 5-Sulphosalicylic acid (100 ppm) (T7); 5-Sulphosalicylic acid (50 ppm) (T8); 5-Sulphosalicylic acid (25 ppm) (T9); 5-Sulphosalicylic acid (100 ppm) + Sucrose (4%) (T10). Among different treatments used in both the cultivar of tuberose, treatment (T5) i.e. Salicylic acid (100 ppm) + Sucrose (4%) followed by treatment (T10) 5-Sulphosalicylic acid (100 ppm) + Sucrose (4%) was best in extending vase life and were able to keep fresh weight of tuberose spikes for longer duration, maximum flower opening, increase in rachis length and spike length as compared to other treatments due to higher uptake of vase solution. Aesthetic look of cut spikes in terms of turgidity and freshness was also better in this treatment.

In another experiment, effect of different polyethylene packaging materials, elicitors, spike length and harvesting stage in tuberose was studied. It was observed that polyethylene packaging comprising of 100µ with 0% ventilation in cv. Mexican Single had more floret weight (0.6g), relative water content (84.0%), membrane stability index (76.9%), total sample weight (178.8g), less PLW (0.6g) and shelf life (5 days). In cv. Prajwal 100µ with 0% ventilation had more floret weight (0.7g), relative water content (82.9%), membrane stability index (77.3%), total sample weight (169.9g) was observed in 75µ with 0% ventilation, less PLW (1.7g) and shelf life (6 days). In cv. Sikkim Selection 100µ with 0% ventilation had more floret weight (0.8g), relative water content (88.8%), membrane stability index (79.8%) was observed in 50µ with 0% ventilation, total sample weight (175.9g), less PLW (2.3g) and shelf life (5 days). Among the elicitors used in cv. Mexican Single, 100ppm nitric oxide had more floret diameter (4.64 mm), relative water content (84.0%) and membrane stability index (76.9%) in 100

ppm boric acid, total sample weight (85.6 g) in 100 ppm salicyclic acid along with maximum shelf life (5days). In cv. Prajwal 200 ppm salicyclic acid had more floret diameter (5.0 mm), relative water content (82.9%) and membrane stability index (77.3%) in 200 ppm boric acid, total sample weight (100.6 g) in 100 ppm salicyclic acid and shelf life (6 days). In cv. Sikkim Selection 100 ppm salicyclic acid had more floret diameter (5.5mm), relative water content (88.8%) in 200 ppm boric acid and membrane stability index (79.8%) 200 ppm nitric oxide, total sample weight (84.9 g) in 100 ppm salicyclic acid and shelf life (5days). Fresh and final weight of the spikes (69.6 and 52.4g, respectively) in Prajwal with 70 cm, RWC (127.5%) in Mexican Single with 60cm, MSI (83.9%) Sikkim Selection with 40cm and vase life (5.2days) in Arka Nirantara with 60cm and in the stages of harvest fresh spike weight (54.0g) was observed in Prajwal with 1-2 florets open, and final weight of the spikes (47.9g) in Shringar with 3-4 florets open, RWC (127.2%) remained highest in Mexican Single with 1-2 florets open MSI (84.2%) Sikkim Selection with 1-2 florets developed colour along with maximum vase life (6 days) in Prajwal harvested with 1-2 florets open.

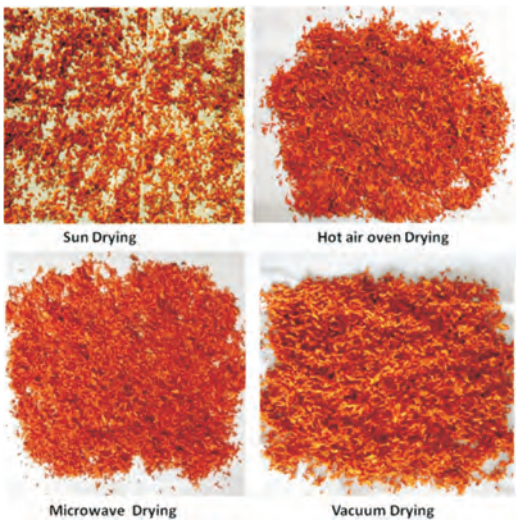

Fig. 8: Drying techniques of marigold for high retention of carotenoids and their anti oxidant activities

Experiments were carried out on estimation of total carotenoids and antioxidant properties, drying of marigold flowers and storage indifferent marigold genotypes. Among the various genotypes studied, selection Af/w-6 had highest total carotenoids on fresh weight basis followed Pusa Narangi Gainda and Pusa Arpita and the selections Af/w-6 , Af/w-4 and Pusa Narangi Gainda had highest antioxidant properties measured by DPPH radical scavenging activity and Ferrous Reducing Antioxidant Power (FRAP) (Akshaya *et al.* 2016). Influence of different drying methods *viz*. sun drying, hot air oven drying, microwave drying and vacuum drying on total carotenoids, bioactive compounds (total phenolic content and total flavonoids) and antioxidant activities [DPPH radical scavenging activity and Ferrous Reducing power (FRAP)] of marigold petals from three varieties *viz.* Pusa Arpita, Pusa Basanti Gainda, and Pusa Narangi Gainda were studied (Fig 8). It was revealed from the studies that vacuum drying was the best drying method with respect to preserving its total carotenoids, bioactive compounds and antioxidant properties followed by microwave drying and hot air oven drying. The components of total carotenoids were quantified by HPLC and it was observed that both lutein and β-carotene content were maximum in vacuum dried marigold petals. The sun drying method had retained lowest carotenoids, bioactive compounds and antioxidant properties. The effect of different storage temperatures and durations on retention of total carotenoids and antioxidant activities of dried marigold flower petals and marigold extract in varieties Pusa Arpita, Pusa Basanti Gainda, and Pusa Narangi Gainda were studied. It was observed that the highest retention of carotenoids and antioxidant properties were recorded at storage temperature of -20°C followed by 4°C and lowest retention was observed at storage at ambient temperature in both marigold flower petals and marigold extract. It was revealed from the studies that the carotenoids, total phenols, total flavonoids and antioxidant properties were found decreased during storage (Akshaya, 2015 and 2016).

In present investigation a total of fifty chrysanthemum genotypes were evaluated, out of which twenty five varieties for total carotenoids (Fig 10) and other twenty five varieties for total anthocyanins and antioxidant activities. Among the various genotypes studied, Red Gold had highest total anthocyanin, total phenol content, highest antioxidant activities (CUPRAC-Cupric reducing antioxidant capacity, FRAP- Ferrous reducing power, DPPH radical scavenging activity) on fresh weight basis followed by Lalpari, Red Stone, Red Spoon, and Jaya. Jubilee had highest total carotenoid, total phenol content, highest antioxidant activities (measured by CUPRAC, FRAP, and DPPH) content on fresh weight basis followed by Star Yellow, Haldighati, Little Orange and Liliput.

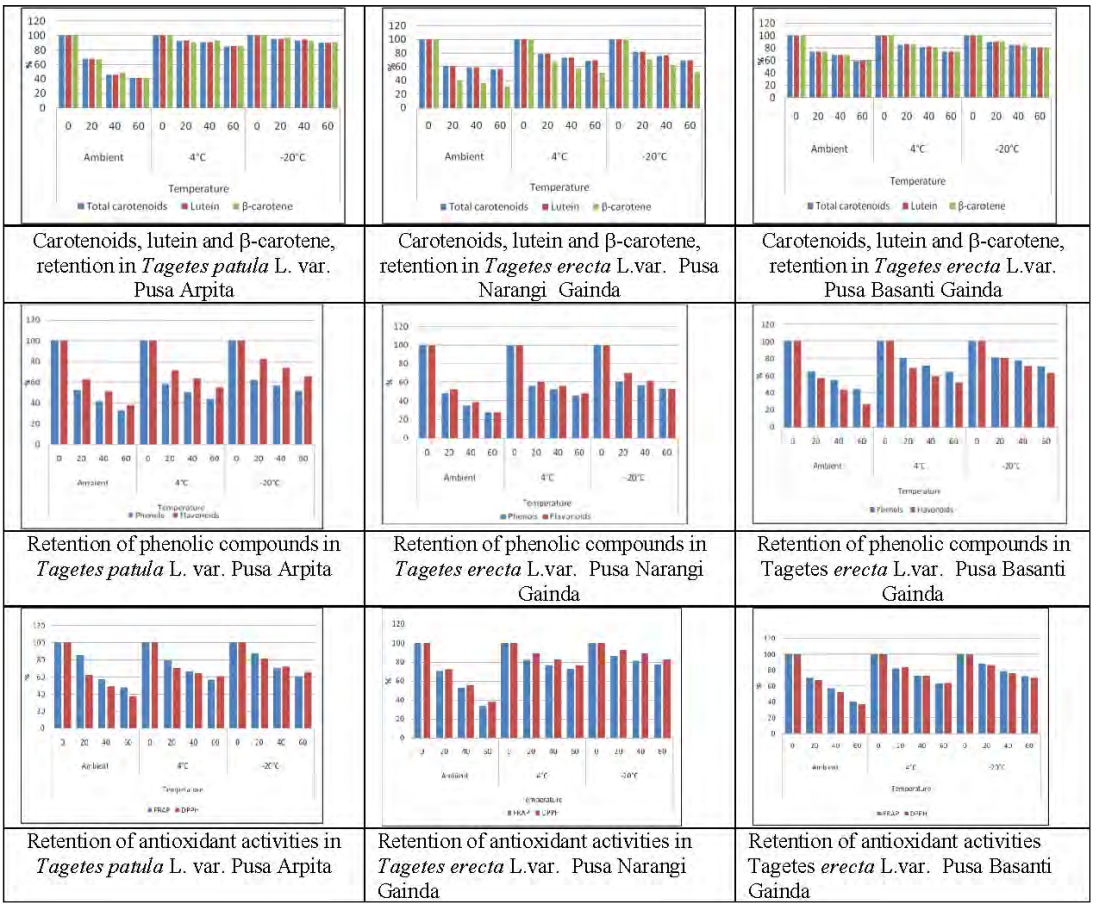

Fig. 9: Retention of carotenoids and antioxidant activities in extract of vacuum dried flowers of African and French marigold varieties during storage

Influence of different drying methods *viz.* air drying, hot air oven drying, and microwave oven drying on total carotenoids, total anthocyanins, total phenolic content and total antioxidant activities (measured by CUPRAC, FRAP and DPPH) of chrysanthemum ray florets from five promising varieties containing high anthocyanin content *viz.*, Red Gold, Lalpari, Red Stone, Red Spoon, and jaya and five promising varieties containing carotenoids *viz.*, Star Yellow, Haldighati, Little Orange and Liliput were studied. It was revealed from the studies that microwave oven drying was the best drying method with respect to preserving its total anthocyanins, total carotenoids, phenol and antioxidant properties followed by air drying and hot air oven drying. The hot air oven drying method had retained lowest anthocyanins, carotenoids, total phenol and antioxidant activities.

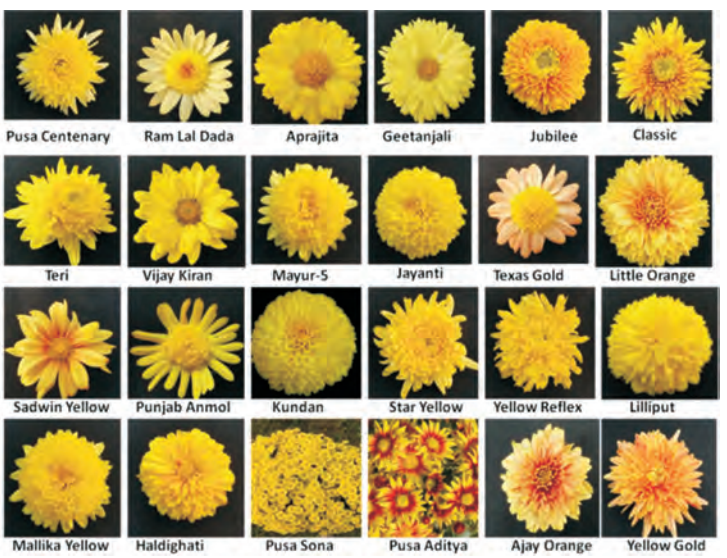

Fig. 10: Genotypes used for determination of total carotenoids

Total anthocyanin, total carotenoids, total phenol, antioxidant activities were highest in fresh ray florets than drying. The components of anthocyanin and carotenoids in promising varieties of fresh ray florets were quantified by HPLC. Among anthocyanins, cyanidin-3-glucoside was the major pigment, Red Gold variety (2123.60µg/g) had highest C-3g followed by Lalpari, Red Stone, Red spoon and Jaya. Among carotenoids, lutein content was high in Jubilee (19.90µg/g) followed by Star yellow, Haldighati, Lilliput and Little Orange. Beta carotene content was highest in Little Orange (5.51µg/g) followed by Haldighati, Jubilee, Liliput and Star Yellow (Fig 11) (Shisa, 2016).

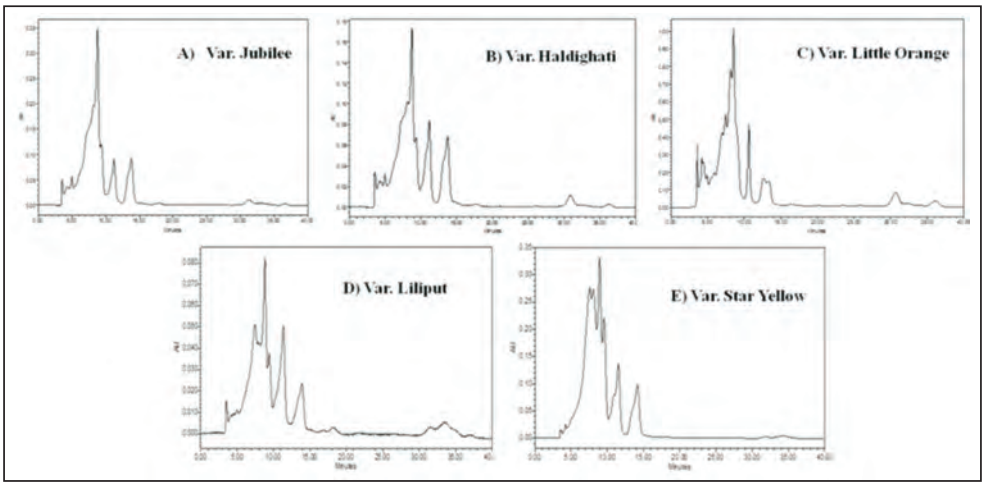

Fig. 11: HPLC chromatogram of carotenoid profile of promising genotypes of *Chrysanthemum* × *morifolium* Ramat. A) Var. Jubilee, B) Var. Haldighati, C) Var. Little Orange, D) Var. Liliput and E) Var. Star Yellow

Turfgrass Management

Turfgrass species *Paspalum notatum* and *Cynodon dactylon* (six cultivars *i.e* Selection 1, Bargusto, Panama Panam, Palma and Tifdwarf 419) were evaluated for various morphological, physiological and biochemical parameters. Tifdwarf 419 emerged as the most promising cultivar for climatic conditions of Delhi among all grasses that were evaluated. Tifdwarf 419 exhibited highest shoot dry weight (1.77 g/ 100 cm^2), shoot fresh weight (5.72 g/ 100 cm^2), root length (17.14 cm), root density (34.17/ 100 cm^3), chlorophyll a (2.67mg/ g fresh weight), total chlorophyll (3.23 mg/ g fresh weight) along with good green colour throughout the growing season than other varieties, whereas shoot density (316.78/100cm^2) was recorded as second highest after selection 1 (384.31/100cm^2).

Tifdwarf 419 was further evaluated under six fertilizer doses [Control (no fertilizer/manure applied), recommended dose (54:18:27 g/m2 N:P:K), ½ of recommended dose, 1½ of recommended dose, FYM @10t/ ha + ¾ recommended dose and FYM @20t/ha + ½ recommended dose], which were applied in three methods, i) full dose of N,P & K as basal dose at the time of planting, ii)½N, full dose of P & K at the time of planting and remaining N after 6 months and iii) 1/3 of N, full dose of P & K at the time of planting, remaining Nin two equal doses after every 4 months. 1½ of recommended dose recorded highest shoot density (562.22/ 100 cm^2), shoot fresh weight (11.35 g/ 100 cm^2), shoot dry weight (3.86 g/ 100 cm2), root density (20.06/ 100 cm3), chlorophyll a (3.87 mg/ g fresh weight), chlorophyll b (0.55 mg/ g fresh weight), total chlorophyll (4.41mg / g fresh weight), leaf nitrogen (2.43 %), leaf phosphorus (0.25 %),soil phosphorus (28.80 kg/ ha), leaf potassium (1.27 %), soil potassium (503.76 kg/ ha) along with high clipping yield (2.53 g/ 100 cm^2) and good root length (30.11 cm). In Tifdwarf 419 highest shoot density (501.17numbers/ 100 cm^2), shoot fresh weight (10.25 g/ 100 cm^2), shoot dry weight (3.98 g/ 100 cm^2), clipping yield (2.30 g/ 100 cm^2), root density (19.31 numbers/ 100

cm3), chlorophyll a (3.84 mg/ g fresh weight), chlorophyll b (0.50 mg/ g fresh weight), total chlorophyll (4.34 mg / g fresh weight), leaf nitrogen (2.43 %), soil nitrogen (225.79 kg/ ha), leaf phosphorus (0.25 %), leaf potassium (1.20 %) and high soil potassium (414.14 kg/ ha) were all recorded with the third method of fertilizer application in Tifdwarf 419, indicating that applying nitrogen in splits of three is more beneficial to turfgrasses than either single application as basal dose or two split applications. Studies revealed that increase in fertilizer rate increased in turf quality parameter and best quality turf was noticed with highest dose of fertilizers i.e. 1½ of recommended dose (81g N, 27g P and 40g K/m^2). Tifdwarf 419 was also studied under four mowing heights of 3 cm, 5 cm, 7cm and 9 cm at three mowing intervals (7 days, 10 days and 14 days). Highest clipping yield (4.72 g/ 100 cm^2), shoot density (441.89 numbers/ 100 cm^2), chlorophyll a (3.18 mg/ g fresh weight), chlorophyll b (0.33 mg/ g fresh weight) and total chlorophyll (3.58 mg/ g fresh weight) along with good root length (26.22 cm) were observed at 3 cm mowing height. Highest clipping yield (4.80 g/ 100 cm^2), shoot density (379.00 numbers/ 100 cm^2), chlorophyll a (3.18 mg of chlorophyll a/ g fresh weight), chlorophyll b (0.36 mg of chlorophyll b/ g fresh weight) and total chlorophyll (3.56 mg/ g fresh weight) were observed with 7 days of mowing intervals. It was evident from this experiment that maintaining Tifdwarf 419 at lower mowing height of 3 cm at mowing intervals of 7 days is suitable for obtaining high quality turfs under climatic conditions of Delhi. It is concluded from present investigations that for climatic conditions of Delhi *Cynadon dactylon* cv. Tifdwarf419 is most suitable which provides high quality turf with fertilizer dose of 81g N, 27g P & 40 g k/m^2 at lower moving height of 3 cm at moving interval of 7 days (Palmsey *et al.* 2015).

Fig. 12: Turfgrass management

Summary

- Cultivated roses and different species display a high level of genetic variability despite the fact that single morphological and physiological characters may be less polymorphic within rose groups. These results will guide new collection activities to establish larger collections and manage the rose genetic resources. The genotypes identified in this study may be directly useful for breeding new cultivars and in development of pre-breeding lines for various traits.
- From molecular analysis, it was concluded that parental lines were genetically more diverse and can be used for heterosis breeding. The hybrid combination, MS-8 x Fr. Sel.1 showed high flower yield among the different combinations.
- Among male sterile lines, MS-8 and among the pollen parents, Fr. Sel. 1 and Fr Sel.2 were the best general combiners for all the characters. Six hybrids were verified using RAPD and ISSR primers and these were classified into three types (markers specific to male, markers specific to female and markers present in both the parents) according to the presence / absence of bands.
- Correlation results based on RAPD and ISSR markers indicate that hybrid performance, heterosis and heterobeltiosis for vegetative and floral characters in marigold increase as the genetic distance between parental lines increases.
- The combine analysis using RAPD, ISSR and URP exhibited the effective variation among marigold genotypes. The female parent MS-8 and pollen parents *viz.*, Af.Sel.1, Pusa Narangi Gainda, Af.Sel.5,

Af.Sel.4, Af.Sel.8 and Af.Sel.10 were the best general combiners for carotenoids and other flower yield related traits. The hybrids MS-8 x Af.Sel.6 exhibited highest heterosis for total carotenoids and MS-5 x Af.Sel.10 for flower yield per plant. These hybrids were further used for breeding of marigold varieties.

- Among the three explants, capitulum explants exhibited the highest proliferation. Modified MS medium (vitamins increased to ten times) supplemented with 10mg/l BAP + 1mg/l IAA showed better establishment. Half strength MS medium supplemented with 1 mg/l IBA was found best for root induction.
- Molecular analysis of *in vitro* raised clones derived from different explants clearly revealed that capitulum and shoot tip gave true-to-the-type plantlets, whereas, leaf derived plantlets are liable to show somaclonal variations.
- Successful implementation of tissue culture technique for the establishment of highly efficient and consistent callus induction, multiplication and subsequent anthocyanin production protocol using ray floret explants of *Chrysanthemum morifolium* cv. Pusa Anmol. Among all the osmoticum (sucrose), salts ($CoCl_2$ and $NiCl_2$) and elicitors (SA, MeJA and ABA); MeJA was found to be the best.
- Bravo Blue was found to be richest in anthocyanin pigment content (106.15 mg/g fresh weight). Leaf explant under *invitro* conditions was found better for induction of anthocyanin pigments in petunia.
- Shortest reaction time was observed in cultivar Pusa Sona and Pusa Aditya, while it was longest in Pusa Chitraksha. The results also revealed that the cultivar having shorter reaction time expressed more antioxidant enzyme activities. Application of atrazine 0.75 kg/ha pre-emergence + carfentrazone @ 0.030 kg/ha post-emergence at 40 days after planting is recommended for weed control and was utilized in controlling weeds in gladiolus fields.
- The shade levels of 50% were optimal for both the cut greens as they produced plants with highest plant height, number of leaves and petiole length with good harvest index and vase life. Among coloured shade net, white shade net has been found to be best for *Cordyline terminalis* and red coloured net in case of *Dracaena fragrans*.
- Big Time Supreme and Jackson Ville Gold recorded maximum keeping quality in terms of field life and vase life in the laboratory (15.67, 13.67 days and 13.67, 12.67 days, respectively). These varieties also recorded higher activity of both enzymatic and non enzymatic antioxidants.
- Salicylic acid (100 ppm) + Sucrose (4%) was best in extending vase life and were able to keep fresh weight of tuberose spikes cv. Vaibhav and Mexican Single for longer duration, maximum flower opening, increase in rachis length and spike length.
- Marigold selection Af/w-6 had highest total carotenoids, high antioxidant activities. Vacuum drying was the best drying method with respect to preserving its total carotenoids, bioactive compounds and antioxidant activities.
- There was highest retention of carotenoids and antioxidant activities in marigold petals stored at storage temperature of -20°C. The standardized drying methods will be useful in better retention of carotenoids and antioxidants in flowers. Farmers will be benefited by getting more income earned by extracting bioactive compounds.
- Red Gold had highest total anthocyanin and Jublee had highest total carotenoid, and total phenol content, highest antioxidant activities on fresh weight basis.
- Microwave oven drying was the best drying method with respect to preserving its total anthocyanins, total carotenoids, phenol and antioxidant activities.
- The synthetic pigments dominate the trade worldwide but they have ill effects on human health and environment. So pigments extracted from natural sources like flowers have great potential for the development of nutraceutical products in the near future. The assessment of antioxidant activities at the basic level will generate information regarding the antioxidant nature of bioactive compounds in chrysanthemum.
- Tifdwarf 419 emerged as the most promising cultivar for climatic conditions of Delhi among all grasses. In variety Tifdwarf 419, application of nitrogen in splits of three is more beneficial to turfgrasses than either single application as basal dose or two split applications.

- Maintaining Tifdwarf 419 at lower mowing height of 3 cm at mowing intervals of 7 days is suitable for obtaining high quality turfs under climatic conditions of Delhi.

Future Perspective

- Identification of new source of resistance to different biotic and abiotic stresses in the commercial flower crops.
- Double haploid production in marigold and other annual flower crops.
- Standardization of protocols for mass multiplication of unexploited flower crops.
- Use of conventional and marker assisted breeding for introgression of desired genes in commercial varieties of flower crops.
- Evaluation of turfgrasses for Indian agro climatic conditions.
- Identification of new genes responsible for pigments in various flowers.
- Exploitation of genetic mechanism like GMS, CMS, antisense RNA technology, etc. for hybrid seed production.
- Association mapping in vegetatively propagated crops.

Acknowledgments

We wish to thank all the Faculty Members of the Division of Floriculture and Landscaping for extending support and cooperation in preparing the research contributions of M.Sc. and Ph.D. students.

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Food Science and Post Harvest Technology

S.K. Jha

Introduction

The research on Food Science & Post Harvest Technology of Perishable Fruits and Vegetables was initiated with PL480 funds in late 1970 in the Division of Fruits and Horticultural Technology, IARI, New Delhi. After the completion of project in early 1980, it was upgraded to AICRP on Post harvest Technology of Horticultural crops during VIth Five Year Plan at eight centres, (four each at ICAR and SAUs). The AICRP on Post Harvest Technology of Horticultural Crops was started in the year 1983 to develop the infrastructure facilities, research promotion and technology generation which can increase the shelf life and quality of fruits, vegetables and flowers for sufficient time to cater the need of the domestic as well as export market. During VIth Five Year Plan the INDO-USAID sub-project on Post Harvest Technology of Fruits and Vegetables was sanctioned by the ICAR with the idea of intensifying research efforts on Post Harvest Technology of seven commercially important fruits and vegetables viz. mango, banana, citrus, guava, potato, onion and tomato. The primary objective of the project was to reduce post harvest losses of fruits and vegetables during harvesting, farm handling, storage, marketing, transportation and processing. The actual implementation of this project was started in October, 1985 and terminated on 3rd March 1991. It remained as a unit of Post Harvest Technology in the Division of Fruits and Horticulture Technology till it became an independent Division of IARI in the year 2002.

The Division of Post Harvest Technology with a multidisciplinary approach was established on 5th February, 2002 in the IX Plan period. A separate teaching discipline leading to M.Sc. and Ph.D degrees was established in the academic year 2004-05. The Division was shifted from its original place of Division of Fruits and Horticulture Technology (Horticulture Building) to the third floor of Nuclear Research Laboratory (NRL) building on 1.7.2010. The name of Division of Post Harvest Technology at IARI was renamed as Division of Food Science and Post Harvest Technology' in November, 2013.

The mandates of the Divisions are

- Integration of production with post harvest management
- Development of appropriate storage protocols
- Valorization through processing
- Design, development and evaluation of packages, low cost post harvest equipment and machinery
- Nutraceuticals and antioxidants in processed food
- Human resource development at Masters and Doctoral level
- Training and Entrepreneurial development programme for transfer of technology
- Advisory Consultancy to both Public and Pvt. Sectors

Significant Student's Research Achievements

Postharvest Management and Shelf life Extension

Kiwi fruits have limited shelf life under ambient conditions. However, it is necessary to extend its life so as to make it available for longer time in the market and to make it commercial fruits in India. Hence, studies

were conducted on the use of 1-Methylcyclopropene (1-MCP), and polyamines. Kiwifruits cv. Allison was treated with different concentrations of 1-Methylcyclopropene (0.5 µL/L, 1 µL/L, 2 µL/L) and un-treated fruits served as control. 1-MCP treatment was given for 24 h at 20°C. After treatments, the fruits were transferred to ambient storage, and observations on different physiological and biochemical parameters were recorded at 3 days interval. Our results indicated that all concentrations of 1-MCP delayed ripening of kiwifruits but 2 µL/L concentrations was the most effective in doing so. Fruits treated with 1-MCP at 2 µL/L started ripening after 12th day of storage whereas untreated fruits started ripening even on 6th day (Jhalegar *et al.*, 2011a). Polygalactouronase (PG) and lipoxygenase (LOX) enzyme activities were lesser in 1-MCP treated fruits than control. 1-MCP treated fruits respired less and evolved lesser ethylene.1-MCP @ 2 µL/L treatment delayed ripening of kiwifruits at room temperature as 1-MCP treated kiwifruits started ripening after 12th day of storage whereas untreated fruits started ripening even on 6th day. Polygalactouronase (PG) and lipoxygenase (LOX) enzyme activities were lesser in 1-MCP treated fruits than control (Jhalegar *et al.*, 2011b).

For studying the effect of 1-MCP on kiwifruits at super market conditions, kiwifruits cv. Allison were treated for 24 h at 20°C with different concentrations of 1-methylcyclopropene (0.5, 1, 2 µl/l) and un-treated fruits served as control. The fruits were transferred to ambient storage (22 ± 4°C; 65-70% RH) after treatments, and observations on different quality parameters were recorded at 3-day interval. The results indicated that all concentrations of 1-MCP influenced PLW, fruit firmness and decay loss in kiwifruit but 2 µl/l concentration was the most effective. PLW in untreated fruits was very high (18.6%) in comparison to 1-MCP treated fruits at the end of 18th day of storage, and 1-MCP (2.0 µl/l) treated fruit were much firmer (31.7 N) than untreated fruit (8.4 N) (Jhalegar *et al.,* 2012a). Fruits treated with 1-MCP (2.0 µl/l) showed the least (8.2%), and untreated fruits the highest fruit decay (22.2%) at the end of storage. TSS in the untreated fruits showed sudden increase on 6th day (12.5%), which increased up to 12th day (16.8%) and declined thereafter (15.7%), whereas in 1-MCP (2 µl/l) treated fruits, it increased sharply from 15th day. 1-MCP (2 µl/l) treated fruit retained ascorbic acid better than other treatments and control. Total acidity and titratable acidity decreased during storage without any significant difference among the treatments. Thus, it could be concluded that 1-MCP (2 µl/l) treated kiwifruits could be stored for 18 days at ambient conditions without much loss in fruit quality (Jhalegar *et al.,* 2012). Similarly, kiwifruits were treated with different concentrations of spermine [(0.5, 1.0 and 1.5) mM] and spermidine [(1.0, 1.5 and 2.0) mM] by the immersion method and stored at ambient conditions for 15 days. Polyamine-treated fruits did not show any amount of ethylene evolution up to the 6th day of storage as compared with those of control, in which it started after the 3rd day of storage. The respiration rate was also the lowest in treated fruit. Similarly, polygalacturonase and lipoxygenase activities rose rapidly in control fruits in comparison with polyamine-treated fruits. The doses of spermine at 1.5 mM and spermidine at 2.0 mM showed the best results in extending the shelf life of kiwifruits when stored in ambient conditions [(22 ± 4) °C, RH: (65 ± 5)%] (Jhalegar *et al.,* 2012b).

Postharvest life of plum is only 3–4 days, and to make it available for longer period in the market, studies were conducted with the use of 1-MCP (1-Methylcyclopropene), ethylene absorbents and ethylene absorbents. Similarly, studies are very important for after storage postharvest removal of plums and their subsequent treatment to make them available for longer time in the market. Fo this different treatments were investigated. In one experiment, 'Santa Rosa' plums i.e. plums were treated at two maturity stages (climacteric and pre-climacteric) with 1-MCP (0.0, 0.5, 1.0, 1.5 µLL^{-1} and ¼ Celfresh tablet) for 24 h at 20°C. After packaging the treated and untreated plums in CFB boxes, these were transported to Delhi, and stored at super market conditions. During storage observations on biochemical attributes and fruit softening enzymes were recorded at 3 day's interval. Our results revealed that all parameters of plums were significantly influenced by maturity stage, and 1-MCP treatment. All 1-MCP treatments have maintained significantly higher levels of phenols, AOX (Antioxidants), anthocyanin content, and delayed fruit ripening and softening by interfering with fruit softening enzymes like PG (Polygalacturonase) and PME (Pectin methyl esterase). In general, fruits of climacteric stage have higher levels of phenolic and anthocyanin contents and AOX activity than plums of pre-climacteric stage. The activities

of fruit softening enzymes like PG and PME were significantly influenced by all concentrations of 1- MCP, but the best inhibition was observed in Celfresh treated plums. Thus, Celfresh tablet can be used for extending the marketability of 'Santa Rosa' plums for about 6 days (Sharma *et al.,* 2012a). At supermarket conditions, untreated plums have very heavy loss during transportation due to compression injury in both stages of maturity (25.0 and 9.0%) being quite higher in climacteric stage than pre-climacteric stage. All 1-MCP treatments had significantly reduced compression injury, being the least (0.0) in 1-MCP (1.5 µl l^{-1}) and Celfresh® tablets. There was significant loss in quality parameters like fruit firmness, AOX capacity, TSS, acidity and ascorbic acid in untreated plums but was maintained quite appreciably by 1-MCP, especially by Celfresh® tablets. Similarly, respiration and ethylene evolution rates were higher in untreated plums than those, which were treated with different concentrations of 1-MCP, being the least in Celfresh® tablet treated fruits days (Sharma *et al.,* 2013). In order to observe the effect of ethylene absorbents, plums were harvested at the pre-climacteric and climacteric stages of maturity and packed with newspaper shreds (control), KMnO4- impregnated chalks, $KMnO_4$-impregnated newspaper shreds or ethylene-absorbent sachets; then they were transported by road to New Delhi. The biochemical and physiological analyses were carried out at 3-day intervals on plums stored in supermarket conditions [(20 ± 1) °C and at (90 ± 2)% RH] for 15 days. Untreated fruits had less firmness than those which were packed with ethylene absorbents. The activities of fruit-softening enzymes such as lipoxygenase (LOX), polygalacturonase (PG) and pectin methylesterase (PME) increased rapidly in the control treatment in comparison with plums packed with various ethylene absorbents, the least being either with ethylene absorbent sachets or with KMnO4-impregnated newspaper shreds. Plums of pre-climacteric and climacteric maturity attained respiration and ethylene evolution peaks on the 9th day and 6th day, respectively. The plums of pre-climacteric and climacteric stages of maturity packed with ethylene-absorbent sachets showed the best results in supermarket conditions in maintaining the firmness, quality and shelf life of plum up to the 12th day and 9th day of storage, respectively (Sharma *et al.,* 2012b).

Nowadays noble molecules are being used extensively in postharvest management of fruits. We also attempted their use in plums. We studied the effect of sodium nitroprusside (SNP) treatment (source of nitric oxide) on chilling injury, electrolyte leakage, malondialdehyde (MDA) content, phenylalanine ammonia lyase (PAL) and pectin methyl esterase (PME) activities during low temperature storage of Japanese plums cv. 'Santa Rosa'. All SNP treatments significantly alleviated CI symptoms during entire period of cold storage. Among different treatments, fruits treated with SNP (0.5 mM) showed 71% lower incidence of CI than control. Minimum electrolyte leakage (49%), PAL (53.6 µmoles of cinnamic acid produced min^{-1} g^{-1}FW) and PME activity (0.36 µmol min^{-1} g^{-1}FW) were observed in 0.5 mM SNP treated plums and the maximum in control. The maximum retention of ascorbic acid (25.6 mg/100 g pulp) and antioxidant capacity (14.7 µmol Trolox g^{-1}) was also exhibited by 0.5 mM SNP treated plums. Lowest MDA content (8.17 nmol g^{-1} FW) and anthocyanin content (592.7 mg/kg FW) were also found in the fruits treated with 0.5 mM SNP. Thus, it can be concluded that SNP is an easy method for application of nitric oxide to maintain fruit quality and extending shelf life by inhibiting fruit softening through interfering with PAL and PME activities during cold storage of 'Santa Rosa' plums (Sharma and Sharma, 2015). Similarly, a study was conducted to study the effect of staggered removal of cold stored (2°C) plums at 7, 14 and 21 days interval and their subsequent treatment with salicylic acid (SA), nitric oxide (NO) and ethylene absorbent (EA) sachets. The fruit were then stored at supermarket conditions (20 ± 1°C and 90 ± 5% RH) with the objective to know whether delayed EA, SA and NO treatments still have significant beneficial effects on the plum fruit quality. The observations on different physiological and biochemical parameters were taken at 2 days interval. The results showed that staggered treatments enhanced postharvest life and maintained fruit quality. We observed that SA-treated plums showed the highest fruit firmness and lowest decay losses when plums were either removed on 7th, 14th or 21stdays of cold storage. Furthermore, SA-treated fruit exhibited lowest rates of respiration and ethylene evolution; phenylalanine ammonia lyase and pectin methyl esterase activities; minimum malondialdehyde content and lowest electrolyte leakage in comparison to those treated either with NO or packed with EA sachets or control fruit. In conclusion, 'Santa Rosa' plum removed after 7th (staggered-I), 14th(staggered-II) and 21st day (staggered-III) from cold storage maintained

a shelf life of 10, 6 and 4 days, respectively at subsequent supermarket storage conditions. The overall results submit that even if the plums are not treated immediately or within few days after harvest and placed as such in cold store, they can be still treated with SA, NO or in-package ethylene absorbent (EA) treatment for beneficial postharvest influences (Sharma and Sharma, 2016).

Kinnow is a very important mandarin. It suffers a lot from several postharvest diseases and for their management, essential oils, plant extracts and edible coatings have been attempted. The essential oils namely lemon grass, eucalyptus, clove and *Neem* on Kinnow mandarin with the objective to combat major post harvest diseases and to prolong its availability for longer time in the season. For this, *in vitro* and *in vivo* studies were conducted. Poisoned food technique was used for in vitro studies, and for in vivo studies, Kinnow fruit were pre-inoculated with pathogens (*Penicillium digitatum* and *P. italicum*), treated with different essential oils and then stored at 5 °C ±1 °C temperature and 85–90 % RH). Our results indicated that all essential oils inhibited the growth (colony diameter) of both pathogens over untreated PDA plates, but the inhibition was the strongest by lemon grass oil. Similarly, under *in vivo* conditions, all essential oils influenced decay incidence, decay loss, lesion diameter, respiration rate, ethylene evolution, overall acceptability and physiological loss in weight but lemon grass was the most effective. And also the incidence of *Penicillium italicum* was more noticed in fruits than *P. digitatum*, however, it was reverse under *in vitro* conditions. The decay rot at all stages of storage was less in Eos treated fruits than untreated fruits, thereby increasing their storage life significantly. Thus, it is evident from our studies that essential oils have the potential to control green and blue mold without causing any injury or harmful effects on Kinnow mandarin, and EOs can be recommended as a safe method for extending its storage life while maintaining fruit quality (Jhalegar *et al.,* 2014; Jhalegar *et al.,* 2015a). Similarly to study the effect of surface coatings such as lac based wax, Citrashine®, P-104® and NiprofreshTM Kinnow fruit were treated with different surface coatings and stored at 5 ± 1°C temperature and 85-90% RH. Our results indicated that all surface coatings maintained a very good profile of quality parameters, but the surface coating of lac based wax was the best followed by Citrashine®, P-104® and NiprofreshTM. All surface coatings influenced all the studied attributes significantly but lac based wax was the most effective in maintaining lowest physiological loss in weight (9.7%), fruit firmness (6.5 N), respiration rate (1.3 ml CO_2 kg^{-1} h^{-1}), and ethylene evolution rate (1.5 µl kg^{-1} h^{-1}). Similarly, all the surface coatings used could retain quality attributes (souble solids content, titratable acidity and ascorbic acid content) of the fruits without any adverse effect. Results suggested that surface coatings have potential to enhance shelf-life of Kinnow mandarin as its fruits could be successfully stored for 60 days under cold storage conditions with highly acceptable sensory quality. The non-coated fruits, on the other hand, maintained postharvest shelf-life of 45 days in cold storage. The surface coatings of indigenous origin (lac-based wax & P-104) can also be recommended owning to their cheaper cost and reasonable effect on storage life (Jhalegar *et al.,* 2015b).

The shelf-life of mango (*Mangifera indica* L.) fruits is only 5 to 6 days under ambient conditions, which can be increased efficiently, if the rates of biological activities and/or changes are reduced by pre and/or post-harvest treatments. Hence, three different concentrations (75, 150 and 200 ppm) of salicylic acid (SA) were applied as pre-harvest treatments to Amrapali mango fruits, one week prior to their commercial harvest. Later the fruits were harvested and stored at ambient conditions (30±5°C and 50±5 % RH). Among various concentrations of SA, the SA (200 ppm) was found to be most effective in delaying the ripening cum senescence processes through suppression of ethylene production rate (0.20 µl C2H4/kg/h) and helped in maintaining the post-harvest quality through better retention of soluble solid concentrates (SSC) (27.72 °B), titratable acidity (0.53 %), ascorbic acid (32.52 mg/100g) and total antioxidant content (11.85 µmol Trolox/g Fresh Weight) etc. The SA treatment was also found to effectively influence the pectin methylesterase activity (0.167 µmol acid/min) as well as the lipid peroxidation (2.26 nmol/g Fresh weight) during storage in order to extend the fruit shelf-life by 3 days compared to the control fruits (Reddy and Sharma, 2016). Mango is a highly perishable and deteriorates at a very fast rate during storage. In contrast to the common practice of postharvest application of growth regulators, we have studied the effect of pre-harvest application of the ethylene biosynthesis inhibitor, viz. salicylic acid (SA)

at three different concentrations (75, 150 and 200 ppm) on 'Amrapali' mango fruits. The results indicate that pre-harvest application of salicylic acid, one week before the commercial harvest could effectively modulate the postharvest behaviour of the mango fruits during storage at ambient conditions (30 ± 5°C and 50 ± 5% RH). Although the mango was responsive to all the treatments, SA (200 ppm) was found to be effective in delaying the ripening cum senescence processes through suppression of respiration rate (105.43 ml CO2 $kg^{-1}h^{-1}$) and ethylene production rates (0.20 µl C_2H_4 kg^{-1} h^{-1}) and retention of high firmness (21.76 N), colour (26.31 ΔE), TSS (27.72°B) and titratable acidity (0.53%) compared to untreated fruits (Reddy *et al.,* 2016). The isolation of high quality RNA from different tissues of mango (*Mangifera indica* L.) is relatively challenging due to the presence of interfering substances such as polysaccharides, polyphenols, and proteins. All these compounds render available isolation protocols useless by reducing the quality (purity and integrity) and quantity of the RNA that can be recovered. Several tissue-specific protocols for the isolation of RNA have been developed specifically for mango, however they are cumbersome, expensive and time-consuming. To overcome these drawbacks, we have developed a comprehensive (CTAB-free, guanidine-free, and LiCl-free) RNA isolation protocol using SDS (sodium dodecyl sulphate) plus phenol which works well for most mango tissues such as leaves, flowers, and fruit, at different stages of development or ripening, as well as fruit peel and seed kernels. This rapid protocol allowed us to process large numbers of samples (12 – 15) simultaneously in a single day. Using this method, we obtained good quantity RNA (16 – 80 µg g^{-1} tissue) from various mango tissues at different stages of development. RNA isolated by this method was pure and amenable to various downstream molecular applications such as RT-PCR and the construction of a cDNA library (Reddy *et al*., 2015). Several postharvest problems affect the sensory quality of mango (*Mangifera indica* L.) fruits which significantly deteriorates the market value of fruits. Among several such problems, lenticel browning (LB) has been reported recently, which affects face value of fruits. Hence, an attempt was made to screen 20 indigenous (Amrapali, Alphonso, Banglora, Bhaduran, Bombay Green, Dushehari, Gulabkhas Green, Langra, Mallika, Neelum, Pusa Arunima, Pusa Pitamber, Pusa Lalima, Pusa Pratibha, Pusa Shrestha, Rataul, Ratna, H-3-2, H-8-11, H-12-1) and 10 exotic (Edward, Eldon, Extrema, Sensation, Irwin, Iturba, Rosari, St. Alexendrina, Tommy Atkins and Zill) varieties/hybrids grown at IARI, New Delhi, to observe the severity of lenticel browning among various varieties/hybrids. Fruits were harvested at full maturity and stored at ambient conditions for 10 days. At the end of 10th day, healthy as well as brown lenticels were counted per square centimetre with the help of a magnifying lens. Our results revealed that among indigenous varieties, Langra had shown the maximum incidence of lenticels browning (100%), followed by Dushehari (52.8%), and among exotic varieties, Sensation had shown the highest (35.9%) incidence of lenticel browning followed by Eldon (28.3%). Interestingly, some hybrids developed by IARI, New Delhi such as H-3-2 (4.3%), H-8-11 (4.5%) Pusa Pratibha (4.8%), Pusa Lalima (5.2%), Pusa Arunima (5.7%), Pusa Shrestha (6.1%) and varieties such as Bhaduran (5.1%) and Alphonso (5.3%) were found to be less prone to LB as they exhibited < 10% incidence of LB. This study revealed that few varieties like Langra and Dushehari were highly susceptible and few hybrids were highly resistant to LB, thus have good scope in International export market (Prasad and Sharma, 2016). Hence to observe the effect of ascorbic acid on lenticels browning (LB) in mango, postharvest dip treatment of ascorbic acid at different concentrations (100, 150 and 200 ppm) was given to four varieties of mango (Indian—'Dashehari', 'Langra'; Exotic— 'Sensation', 'Eldon'). After treatment, fruits were stored at ambient conditions (35 ± 4 °C and 65 ± 5 % RH) for 10 days. At the end of storage period, observations were recorded on several parameters. Our results revealed that fruits of 'Langra' mango exhibited 100 % LB, followed by 'Dashehari' (52.8%), 'Sensation' (42.9%) and 'Eldon' (38.3%). All concentrations of ascorbic acid reduced the LB to greater extent and improved the cosmetic appeal of fruits of all the varieties significantly over untreated fruits but the best results were obtained with 200 ppm concentration for LB (4.8, 1.6, 3.7, 3.2%, respectively) and other attributes, non-significantly followed by 150 ppm concentration. All concentrations of ascorbic also reduced the activities of polyphenol oxidase, peroxidase and lipoxigenase enzymes, and also reduced respiration rate and ethylene evolution rate but without impairing eating quality attributes (soluble solids concentrates and total carotenoids). It was concluded that postharvest dip application of ascorbic acid at 150 ppm reduced LB and improve face value without impairing fruit quality (Prasad *et al.,* 2016).

During mango harvesting, a conspicuous sap exudes out from the plucking point of the fruit stalk and causes brownish-black to black streaks or spots on the skin of mango, which is called sap-burn. The incidence of sap-burn injuries ranges from a low of 5 % to a high of 50%. Lime (calcium hydroxide) has been identified as a potential chemical for de-sapping of mango fruits. It is a strong alkali with a pH 12.4, thus it reduce sap-burn injury by neutralizing the highly acidic sap. Mangoes (with stem attached) are dipped in lime solution (1 %) for 45 – 60 seconds and de-sapped by removing stems while fruits are in submerged condition.

Influence of tree age on postharvest quality and storage behaviour of Amrapali Mango fruits was evaluated (Meena, 2015). It was observed that minimum acidity (0.64%) and highest carotenoids (3.78mg/100g) were found in fruits harvested from 30 years old trees. However, highest fruit peel thickness (1.27mm), ascorbic acid (56.84mg/100g), antioxidant capacity (2.72 µmol trolox.g^{-1}) and lowest respiration rate (37.71mL CO_2 kg^{-1} h^{-1}), polygalacturonase activity (16.26 µg gallic acid.$g^{-1}h^{-1}$), pectin methyl esterase activity (0.023 ΔA_{620} $min^{-1}g^{-1}$ FW) were obtained in fruits harvested from 6 years trees.

The effect of in-package use of ethylene absorbents ($KMnO_4$, Bioconservacion, Al_2O_3, silica gel and $KMnO_4$ + silica gel) and different concentrations of sodium nitropruside was studied in nectarine. The results revealed that among the various ethylene absorbents, $KMnO_4$ sachets was found to be better in extending the shelf life of nectarine fruits up to 21 days at cold storage and for 8 days at super market condition. Among different concentrations of SNP, NO @ 0.5mM was best in reducing PLW, maintaining firmness and retaining higher phenolics, antioxidant activity, exhibiting slower increase in LOX and PME activity and better quality fruits up to 8 days as compared to 4 days of untreated fruits (Jayarajan, 2016).

Salubrious climatic conditions (neither too cool nor too hot) produced nutritionally rich strawberry fruits harvested during March. Besides picking months, varietal difference was also found linked with fruit quality. Higher average fruit weight (>10 g) was recorded during March followed by February and April harvesting months. Highest total sugars and total soluble solids content were recorded with March picking (9.8 to 11.54^{0}brix). Camarosa variety exhibited better adoptability and maximum anthocyanin and total antioxidant content were recorded (Bhatia, 2014) at even higher temperature during March and April.

Generally pomegranate could be stored at 10°C with 95% relative humidity for about 8 weeks but with susceptibility to husk scald development. When pomegranate is stored below 5°C, it develops chilling injury (CI) resulting in a reduction in both internal and external fruit quality. To address these problems pre storage application of polyamine and carnauba wax coating was made. Chilling injury, which is a major bottleneck in low temperature storage, was found to reduce by more than >50% in treated fruits compared to control. Putrescine 2 mM + carnauba wax (1:10 emulsion) treated pomegranate fruits may be stored (at 3 to 5°C and 90 ± 5% RH) in consumer acceptable state for fresh consumption up to 60 days (Bhatia, 2014).

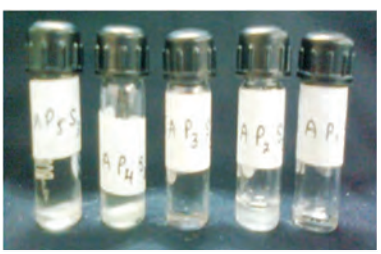
Studies on finding suitable cultivar, packaging material and pre-treatments for minimal processing of pomegranate arils was carried out (Bhatia, 2014). At the end of storage (15th day), minimally processed arils from Mridula cultivar showed the least browning, respiration rate (64.41 ml CO_2 kg^{-1} h^{-1}), weight loss (0.064 %), microbial count (5.4 log cfu g^{-1}) with the highest acceptability

score compared to Kandhari and Bhagwa. Among tested packaging materials, PP bags maintained better quality characteristics of minimally processed arils up to 15 days storage period. It retained better colour (higher anthocyanin retention) and total phenol compared to LDPE and KPA packed arils.

A study was conducted on summer squash to solve postharvest problems of water loss, chilling injury and postharvest decay through the application of suitable surface coating material, anti chilling compound and post harvest disease controlling microbes. The results revealed that 10% gum arabic as a coating material proved to be the best in minimizing water loss (more than 50%), respiration rate (more than 50%) and maintaining higher fruit firmness (7.55 N). Among the tested anti-chilling molecules, salicylic acid @1.0 mM proved the best in minimizing chilling injury score (0.74), PLW (11.73%), retaining higher level of phenolics (420.75 µg/g) and proline (333.87 µg/g) during storage. For increasing shelf life, treatment with ozonated water @ 0.4 ppm retained the best physical traits like fruit firmness (5.84 N) and lower PLW (15.06%), although higher concentration of ozonated water (0.6 ppm) effectively controlled decay incidence (10.83%), bacterial count (3.92 log cfu/g) and yeast-mould count (3.65 log cfu/g). Among bioagents, *Debaryomyces hansenii* proved the best by virtue of higher antagonistic activities against gray mould disease. *Pseudomonas fluorescens* was found to be the best in controlling bacterial soft rot by minimizing lesion diameter (26.67 mm), maximizing zone inhibition (4.33 cm^2) and minimizing fruit weight loss (13.54 %) at the end of storage period of 12 days (Kannaujia, 2016).

Two varieties of garlic, namely, G-1 and G-323 were analyzed for changes in their physical and physico-chemical composition during storage for observing the efficacy of different doses of gamma irradiation (0.075, 0.10 and 0.15 kGy) applied 120 DAH at ambient temperature (18-32°C & 55-65% RH) followed by modified atmosphere packaging of the minimally processed garlic cloves of both the varieties for 28 days at 10°C. Irradiation dose of 0.1 kGy was most successful for checking sprouting, controlling decay by 22%, 28%, PLW by 8.2% and 7.6% in G-1 and G-323 varieties, respectively. Freshness of minimally processed garlic cloves could be suitably retained till 28 days of storage under MA conditions (Madhav, 2013).

Processing of Horticultural Crops



The essential oil from four citrus species namely, *Jatti khatti, Soh sarkar*, grapefruit and pummelo, was extracted both from freshly harvested fruits as well as from fruits stored under ambient and refrigerated conditions. Oil yield decreased initially with increase in storage period but increased at later stage but it was found to be higher from peels of fruits stored at refrigerated condition than fruits stored at ambient condition (Jalgaonkar *et al.,* 2013). Analysis of oil indicated that number of volatile components identified in grapefruit (16 compounds) were more compared to other citrus species such as *Jatti khatti* (14), *Soh sarkar* (11), and pummelo (6). However, the total concentration of volatile compounds were found to be more in *Soh sarkar* (99.95%) followed by *Jatti khatti* (99.82%), pummelo (98.92%) and grapefruit (97.82%). Limonene, one of the major component content was found to be higher in pummelo (96.41%) followed by *Soh sarkar* (96.23%), *Jatti khatti* (95.13%), grapefruit (93.57%). Maximum limonene content was observed in oils extracted from peels of 1.4 mm ground samples in all citrus species (Jalgaonkar *et al.,* 2012).

Extraction of lycopene from freeze dried powder of grapefruit (Cv. Red Blush) and papaya (Cv. P-9-5) using supercritical CO_2. The optimal condition was found to be 70°C, 305.5 bars, 35 g/min and 135 min for grapefruit and 80°C, 225 bar, 25 g/min. and 90 min for papaya. The lycopene recoveries of 92.83% and 93.26% were predicted (Priyadarsini, 2015). Extracted lycopene from all the grapefruit and papaya were stored at different temperatures (3°C, 10°C & 21°C) to study the stability of the same in terms of its half-life. Half-lives of lycopene extracted from grapefruit were 16, 6 and 3 days respectively at 3, 10 and 21°C. The same for papaya were 14, 5 and 2 days respectively. This indicated that the lycopene extracted from grapefruit was more stable compared to those from papaya.

Encapsulation of β-carotene with natural polysaccharides using spray freeze drying technique was carried out. Jackfruit seed starch (JSS) and tamarind seed gum (TSG) were isolated from natural seed sources and used as encapsulants for coating β-carotene in spray freeze drying (SFD) technique. Carrot coagulum extracted from discarded carrots left over agricultural fields using enzyme assisted hydraulic press method was used as source of β-carotene. The optimized process conditions of SFD technique was found as 0.8 mm nozzle size, 30 ml min^{-1} feed flow rate and 0.25 air pressures. This best combination yielded β-carotene 26.37 mg/ 100 g dry matter, total carotene 57.17 mg/100 g dry matter, encapsulation efficiency 76.22%, particle size (D_{90}) 210.37 µm, total colour change (ΔE*) 62.80, antioxidant activity 3.28 mmol TE/100 g dry matter, water solubility index 93.65%, moisture content 9.67% in dry basis and water activity 0.48 (Darshan, 2015).

The watermelon (Sugar Baby variety) juice was extracted and evaluated for quality parameters which showed that it contains 8.00±0.17 mg ascorbic acid/100ml juice, and 5.7±0.13mg lycopene/100g juice respectively. The predominant sugar and organic acid was malic and fructose (3.38 mg malic acid and 31.10 mg fructose/ ml juice), respectively. 1- Dodecyne, 6-Nonenal (Z), Propanoic acid, 2-oxo methyl ester,2-Propanol,1-methoxy, Hexanal and 5-Heptene, 2-one, 6- methyl were the major key flavour compounds identified from watermelon juice during the flavor analysis through GC-MS. Pectin (0.2%) was the most suitable stabilizer for watermelon juice. Pasteurization with thermal treatment of 72°C for 20s, irradiation at 0.5 kGy and microwave pasteurization at 1.56 W/ml juice (280 W/ 2 min) were further studied for the preparation of watermelon drink. The results showed that, microwave pasteurization at 1.5W/ml juice (280W/2min) was comparatively better in preserving the quality especially the colour and flavour of watermelon drink (Shameena, 2016).

Valorization of red capsicum through enzyme assisted processing was carried for developing carotenoid and antioxidant rich extract. Three carbohydrases enzymes namely; viscozyme, pectinase and cellulase were tested for recovering carotenoids and phenolics in the extract. Among the three enzymes (viscozyme, pectinase and cellulase) tested, viscozyme significantly ($p<0.05$) improved the total carotenoid and total phenolic content by 2 fold over untreated extract. Under optimal conditions of incubation temperature (50.98°C), extraction time (32.08 min) and enzyme concentration (0.351%), maximum yield of carotenoids (50.43 mg/100g) was obtained (Nath, 2016). The developed concentrate with high content of carotenoids, phenolics and ascorbic acid was used as functional ingredient in development of yoghurt.

Processing of Cereals, Pulses and Oilseeds

Pearl millet (*Pennisetum glaucum*) is commonly known as bajra. It is one of the most extensively cultivated cereals in the world, after rice, wheat, and sorghum. It is particularly grown in arid to semi-arid regions. It is cultivated in a vast range of environmental conditions including environments with frequent drought and poor soil fertility. It is also less amenable to common diseases and pests. India is the largest producer of pearl millet with 9.1 MT production and 1272 kg/ha yield from the 7.1 million ha area during the period 2014-15. The grain is excellent source of protein, fiber, starch and energy. It constitutes carbohydrate (67.5 g), protein (11.6 g), fat (5 g), fiber (1.2 g), mineral (2.3 g), calcium (42 mg), iron (8 mg), zinc (3.1 mg), vitamins B, especially niacin, B6, folic acid. Also, it provides energy (361 kcal/100g) which is more than commonly consumed cereals such as rice (345 kcal/100g), wheat (346 kcal/100g), sorghum (349 kcal/100g) and maize (125 kcal/100g). It is a gluten free grain and is the only grain that retains its alkaline properties after being cooked which is ideal for people with wheat allergies.

Bioavailability of the nutrients present in pearl millet grain is restricted due to the presence of anti-nutritional factors such as phytic acid, tannins, goitrogens, oxalic acid and trypsin inhibitors. The phytic acid is present in the germ whereas, polyphenols are in peripheral areas of the pearl millet grain. Phytic acid or phytate has a strong ability to chelate multivalent metal ions, especially iron, zinc, calcium and magnesium reducing the absorption of these minerals by humans. Pearl millet flour turns bitter and rancid within a few days of storage

period especially under conditions of moderately high moisture and oxygen exposure. Tiwari *et al.* (2014) studied on the effect of different treatment on quality of pearl millet flour. Pearling, steeping and heat treatment of pearl millet caused significant ($P \leq 0.05$) reduction in phytic acid and total polyphenols. Phytic acid decreased from initial 728 to 591, 396 and 410 mg/100 g, after pearling, steeping and heat treatment, respectively. Heat treatment was inferred as the best pre-milling treatment as it caused non-significant decrease in iron and zinc contents. However, pearling and steeping led to significant reduction in iron and zinc contents. Pearled grain flour had the brightest colour (L^*=69.4) and was followed by heat treated flour (63.6) and steeped (59.7). Pearling and fermentation caused significant reduction in phytic acid of 29.25% and 45.32%, respectively in pearl millet flour. These two pre-treatments also reduced polyphenols, iron and zinc significantly, whereas heat treatment significantly reduced phytic acid by 43.68%, and polyphenol but had no significant reduction in iron and zinc. Tannin content of pearl millet flour was found to be unaffected with any of the pre-milling treatments. The fat acidity and FFA were found to be the minimum in case of heat treated flour. Heat treatment was inferred as the best pre-milling treatment. The phytic acid, fat acidity and FFA increased with increase in storage period. Based on sensory scores and critical levels of phytic acid (600 mg/100 g), fat acidity (30 mg KOH/100 g) and FFA (1%), it was concluded that heat treated pearl millet flour could be stored up to 6 days without undue deterioration in quality at ambient condition.

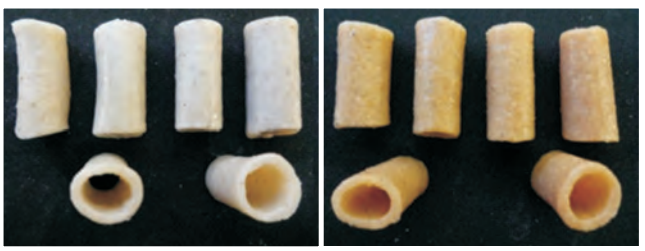
Kirti *et al.* (2016) determined the effect of hydrothermal treatment on storage life of pearl millet flour stored at ambient condition (average maximum and minimum temperatures 33.1±4°C and 21.4±2°C, respectively, and 84.4±2% RH) and controlled condition (30±1°C at 50±2% RH) packed in zip-lip low density polyethylene (LDPE) pouches of 10 µm thickness having permeability 23.13g/m^2 24h.. It was reported that storage life of the flour could be extended up to 60 days. During this period the fat acidity of the flour varied from 23 to 36.5 mg KOH/100g at ambient storage, phytic acid from hydrothermal treatment significantly reduced fat acidity of pearl millet flour by 12%, phytic acidity by 4% and trypsin inhibitor activity by 5%,. It was observed that pearl millet flour can be stored at controlled condition in superior form than ambient condition. Storage period had significant ($P<0.05$) effect on moisture content, fat acidity, phytic acid and trypsin inhibitors activity of pearl millet flour. It was found that hydrothermally treated pearl millet flour can be stored up to 30 days at ambient conditions and 45 days at controlled condition without undue deterioration in quality.

Hydration characteristics of grains are of paramount importance in processing operations like preconditioning, flaking, popping, roasting etc. Hence, hydration characteristics of pearl millet grain were evaluated by Chauhan *et al.* (2014). Water absorption by the grain increased with increase in temperature. The increase in water absorption by the grain was more rapid at temperatures 70°C and above. The initial rates of hydration at temperatures 16, 30, 40, 50, 60, 70, 80, 90 and 100°C were 14.2, 20.5, 24, 32.9, 43.2, 47.6, 94.7, 136.3, and 142.7 g.kg.dm^{-1}.min^{-1}, respectively. The rate of hydration decreased with increase in hydration time and approached very low value (≤0.1 g.water.kg.dm^{-1}.min^{-1}) corresponding to the grain moisture contents 16, 19, 22, 25, 33, 39, 46% (wb), respectively, at 16, 30, 40, 50, 60, 70, 80°C. The maximum moisture content after the hydration of pearl millet grain could be 48%, beyond which bursting of grains was observed. Moisture diffusivity varied from 2.13x10^{-4} m^2.s^{-1} to 30.56x10-3 m^2.s^{-1}, with average value of 9.99x10^{-3} m^2.s^{-1} during hydration. Activation energy for hydration of pearl millet was found to be 55.71 kJ.mol^{-1}. Page model was found to be the best for modelling hydration characteristics of pearl millet.

Chauhan *et al.* (2015) screened thirty eight germplasms/varieties of pearl millet (*Pennisetum glaucum* L. R Br) for popping characteristics at optimum processing conditions. For determining optimum processing condition, popping experiment of pearl millet (PC 443) was initially carried out as per central composite rotatable design in response to surface methodology with three factors (moisture content, tempering time and popping temperature) at five levels. Optimum conditions for maximum popping yield (62%) was determined as 15.92% moisture content, 7.16 h tempering time, and 282°C temperature.

The popped grains were found to have good nutritional value with 11.2% protein, 7.36% fat, 2.96% crude fibre, 3.83% ash, 72.81% carbohydrate, and total antioxidant 15.47 µ mole Trolox /100 g. Popping significantly reduced anti-nutritional factor to a level of 373.82 mg/100g phytic acid. Based on popping characteristics like popping yield, puffing index and size of popped grain, seven germplasms/varieties, viz. IC 283734, IC 283745, IC 283763, IC 283842, IC 283908, 841-B and PPMI 301 were found to have potential popping trait.

Tiwari (2012) developed an expanded ready-to-eat snack for school going children (5-15years) as per ICMR guidelines (ICMR 2010) using pearl millet, QPM and green gram through extrusion processing. The blend composition consisting of 60% pearl millet, 30% QPM and 10% green gram was selected for extrusion processing for snacks preparation as it provided desirable quantities of nutrients (especially protein and iron) at the lowest cost. Response surface methodology was used to study the effects of feed moisture content (10–22%, w.b.), screw speed (200–600 rpm), and barrel temperature of the exit end (80–160°C) on extruder system parameters (product temperature, motor torque, specific mechanical energy) and physical properties (product moisture, WAI, WSI, expansion, bulk density, hardness and crispness) as well as nutritional characteristics (protein, iron, phytic acid, total polyphenols) of a pearl millet flour-based snack. Second-order polynomials fitted well to the extruder responses and product properties as a function of process variables. All three variables affected product and process responses significantly. Desirable expanded product, characterized by high expansion ratio and low bulk density and hardness as well as maximum reduction of anti-nutritional factors, were obtained at low feed moisture of 13%, high screw speed of 470 rpm and barrel temperature of 128°C. The snack produced at this optimized condition had expansion ratio: 4.56, density: 139.88 kg/m^3, WAI: 6.34 g gel/g of dm, WSI: 12.56 g/g, colour (L* value): 65.43, hardness: 1.88 N, phytic acid: 377.31 mg/100 g, total phenols: 302.12 mg/ 100 g, protein: 13.02%, fat: 5.26%, carbohydrate: 73.13%, fibre: 3.07%, ash: 2.59%, iron: 5.24 mg/100 g, and Zn: 3.13 mg/100 g.



Jalgaonkar and Jha (2016) investigated the effect of particle size and blend composition (wheat semolina: pearl millet flour) on quality of pasta. Initially, the pasta was prepared from 100% pearl millet flour of different particle sizes (241e780 mm). Observation indicated that it was not possible to make pasta from 100% pearl millet flour as these disintegrated after cooking. Particle sizes of pearl millet flour showed significant effect on nutritional and cooking quality of pearl millet pasta. Pasta from pearl millet flour of particle size 425 mm had least cooking loss, high protein, iron and zinc contents. Further, with increase in the level of pearl millet flour in the blend composition, protein, ash and cooking loss of pasta increased whereas hardness, cohesiveness, springiness, gumminess and chewiness showed decreasing trend. Blend composition (wheat semolina: pearl millet flour) in the ratio of 70:30 was found to be satisfactory for making pasta with desirable quality characteristics like cooking loss (<8%), protein content (>10%), ash content (<0.7%), colour and texture. However, with the objective of maximum incorporation of pearl millet flour in the final product, a blend composition of 50:50 could be used to make pasta with acceptable quality.

Basmati rice possesses unique cooking, eating and digestive qualities. India produces more than 70% of the world bamsati rice production. Among basmati, PB1121 is widely grown in India and contribute to major share in export of rice from the country. Therefore, drying of PB1121 was carried out with the objective to optimize the drying variables for minimum drying time and maximum head yield. Drying was carried out in 2-passes using a cross-flow dryer. Drying experiment in thin layer was conducted as per central composite rotatable design

in response surface methodology with 3 factors (drying air temperature, tempering time and moisture reduction in first drying pass) at 5 levels. The variables chosen for experiment were drying air temperature (40-70°C), tempering time (60-240 min) and moisture reduction percentage (3-10%) in first pass of drying. Optimization of drying variables was done through the use of a desirability function by combining all responses into one measurement. The desirability functions were minimum drying time, maximum brown rice yield and head rice yield. Total drying time ranged from 35 to 132 min in two stages of drying while in continuous drying it varied from 41 to 204 min. Head rice yield ranged from 39.77 to 71.56%. Optimum conditions for drying of paddy grains were found to be temperature of 58.3°C, 7.13% moisture reduction and 148 min tempering time (Jha *et al.,* 2013). At optimized conditions, total drying time and head rice yield were calculated 53.1 min and 54.53%, respectively.

Patel *et al.* (2013) determined the physical properties of two varieties (PB1 and PB1121) basmati paddy at moisture contents ranging between 22.5 and 9.5% (w.b.). Moisture content was found to have significant effect on the properties of both the varieties. Axial dimension, sphericity and surface area of both varieties decreased with decrease in moisture content. The thousand grain weight, bulk density and angle of repose decreased from 27.83 to 19.72 g, 378 to 322 kg/m^3 and 48.54 to 30.06 degrees, respectively while true density and porosity increased from 1266 to 1298 kg/m^3 and 70.17 to 75.19%, respectively with decrease in moisture content in PB1. In case of PB1121 paddy, the corresponding values were found to be 29.94 to 22.24 g, 464 to 429 kg/m^3, 36.06 to 28.06 degrees, 1366 to 1398 kg/m^3 and 60.03 to 69.31%, respectively. Terminal velocity and coefficient of friction decreased with decrease in moisture content while hardness of paddy increased.

Sakharam *et al.* (2013) evaluated soaking behaviour of paddy (PB1121) at 40, 50, 60, 65, 70, 75 and 80°C soaking temperatures. Soaking curves showed that gradual decrease in soaking time due to increase in soaking temperature. Samples kept at 40°C took 960 min to reach M_e while samples kept at 80°C took 135 min to reach the same. Increase in soaking temperature was accompanied with corresponding increase in the soaking rate due to decrease in grain diffusion resistance. Three distinct soaking rate periods were observed when soaking was carried out at 70, 75 and 80°C while only two soaking periods were observed at 40, 50, 60, 65°C. Gelatinization of paddy at 70, 75 and 80°C caused the second peak in soaking rate and which in turn caused the third soaking rate period. From soaking curves it was observed that during soaking of PB1121, optimum soaking time for soaking at 40, 50, 60, 65, 70, 75 and 80°C was 900, 630, 480, 420, 230, 165 and 105 min respectively. Page model was fitted to soaking data and statistical analysis revealed that MSE and P values increased with increase in the temperatures except 80°C. R^2, P and MSE values of Generalized Page model were 0.550, 163.38 % and 0.01619 indicating good performance of Page model. MLP neural network was used to model soaking data and it was found to be more efficient than Page's model with R^2 0.89 and MSE 0.0061.

Kale *et al.* (2015) determined the qualitative changes in basmati rice (Pusa Basmati 1121, PB1121) during soaking at 40 °C to 80 °C. Soaking temperature had significant effect (α = 0.01) on chemical composition, glycemic index and starch characteristics of rice. Starch content, apparent amylose content, crude protein content and crude fat content in un-soaked rice were found to be 73.24%, 27.26%, 8.79% and 2.56%, respectively, but differences in these traits were observed after soaking. Amylose to amylopectin ratio (Am/Ap) decreased from 0.59 to 0.52 (soaked at 80°C). Crude fibre and crude ash contents increased after soaking. The mineral composition (K, P, S, Ca, Mg, Mn, Fe, Cu and Zn) in soaked rice was found to be 16.46% higher than un-soaked rice at the same degree of polishing. Glycemic index of un-soaked rice was found to be 58.41, but decreased to 54.31 after soaking at 80°C. Pasting properties, scanning electron microscope images, and X-ray diffractograms suggested partial gelatinization of starch in the temperature range of 60°C to 80°C. Based on qualitative changes in rice (apparent amylose content, Am/Ap ratio and crystallinity rate), it was concluded that intermediate soaking temperatures (60 °C to 70 °C) would be useful for soaking of PB1121.

Kaukab (2016) developed reconstituted rice using the broken rice/rice-soy flour through extrusion processing. Optimum range of extrusion process for rice flour based product was found to be 41 to 42°C barrel temperature, 20 to 21.6% (wb) feed moisture content and 26 to 27% of rated screw speed and for rice-soy blend was determined as 67 to 68°C barrel temperature, 19 to 22% (wb) feed moisture content and 45 to 49% rated screw speed. Reconstituted rice with additives (1% soya isolate and 3% $CaCO_3$) had reduced cooking time and solid loss but scored higher (4.98) in sensory analysis.

Development of Breakfast cereal and Snacks

Om (2013) developed a breakfast cereal through extrusion processing using maize (Quality protein maize as well as normal maize), finger millet and carrot powder. Extrusion parameters were standardized as 15.5% feed moisture, 120°C temperature, and 400 rpm screw speed.. The product had 9.6% protein, 1.5% fat, 3.4% fibre, 2 mg/100g β-carotene, 3 mg/100g iron and 122 mg/100g calcium. The bowl life of the product was found to be 4 min. The amino acid profile showed non-essential amino acids (Asparagine, Glutamine, Serine, Glycine, Cysteine, Arginine, Alanine, Tyrosine, Proline) and essential amino acids (Valine, Tryptophan, Phenylalanine, Leucine, Lysine, Threonine). Amino acids were higher in quality protein maize based product than normal maize based product. Two essential amino acids i.e, tryptophan and lysine were 53.7% and 45% higher respectively, in QPM based product than normal maize based product.

Thakur (2015) developed green flakes using Pusa Green 112 variety of chickpea. The optimum moisture and temperature for flaking of grain were found as 30% (w.b.) and 60°C, respectively. Microwave heat treatment of whole grain for better flaking (ie less broken after flaking, and retention of green colour) was established as 18 W/g power for 3 minutes of exposure time was found optimum for flaking. After flaking operation, flaked grains were roasted in a microwave oven at 69.2 W/g power for 1 minute exposure time was found appropriate for maintaining green colour, less brokens, and least trypsin inhibitor activity.

Puineet (2016) standardized hot water soaking at 60°C up to 1 h for retention of green colour of the soybean (var. Hara Soya). Steaming led to decrease in TIA from 27.2 in raw grain to 4.05 mg/g (85% maximum reduction), increase in VER up to 2.11, decrease in bulk density, and also decrease in colour (a*=–1.83 to 2.67). Dry heat treatment using microwave oven was found appropriate at 300 W (10W/g). Soaked grain with this treatment could be converted to soy nuts with natural colour retained, making it soft and crunchy.

Patil (2016) developed innovative gluten free snacks from composite flour comprising of rice, finger millet and horse gram (40:50:10). Optimum processing conditions were: feed moisture: 13.36% wb, die head temperature: 130.58°C and screw speed 329 rpm. The developed snacks had significantly ($p<0.05$) higher total phenolic content (465.23 mg FE/100g dw), antioxidant activity (17.98 µmol TE/g dw) and moderate glycemic index (67.77) than market samples.

Pankaj (2016) optimized processing conditions for extrusion of finger millet as feed moisture: 19% wb, die head temperature: 120°C and screw speed 380 rpm. Inclusion of extruded flour (20g/100g), was found to promote extensibility, work of adhesion and cohesion, in both composite wheat and gluten free dough; mimicking properties of hydrocolloids.

Aabon (2012) standardized process for development of soymilk with reduced LOX activity. Pusa 9814 (yellow variety) and Kalitur (black variety) were selected for the preparation of soymilk. Amongst various pre-treatments, 4 min boiling water blanching of soybeans was found to be best for preparation of good quality soymilk with less LOX activity. Based on organoleptic acceptability, 0.05% citric acid along with 10% mango/ bael pulp and 10% soymilk were found to be the best blends for preparation of soy-fruit beverages. The combined flavor of bael fruit and yellow soymilk was pleasant and bael fruit could effectively mask the 'beany flavour' of soymilk as compared to mango. The samples stored at ambient temperature had a shelf life of 2 months whereas low temperature stored samples were organoleptically acceptable even beyond 3 months.

Darshan (2015) carried studies on microencapsulation of rice bran oil (RBO) with different combinations of starch (Jackfruit seed starch, Tapioca starch) and protein (Whey protein isolate, Soya protein isolate) using spray drying technique. Encapsulation efficiencies varied from 77% to 84.91%; γ-oryzanol contents from 12,390 ppm to 12,843 ppm; and peroxide values from 1.09 meq/kg oil to 1.52 meq/kg oil (well within the acceptable limit of fresh oils <10meq/kg oil). Combination of jackfruit seed starch and whey protein isolate used in a ratio of 2.6:1 having oil concentration of 20% when spray dried using an inlet air temperature of 140°C was found to be the best conditions for encapsulation of rice bran oil without any significant detrimental effect on γ-oryzanol either in its quality or quantity. The samples were found to be stable at ambient condition with the maximum peroxide value of 2.21 meq/kg oil; and minimum γ-oryzanol of 12,088 ppm.

Acknowledgement

We wish to thank all the Faculty Members of the Division of Food Science and Post Harvest Technology for extending support and cooperation in preparing the research contributions of M. Sc. and Ph. D. students made during the period of 2007-16.

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Fruits and Horticultural Technology

O.P. Awasthi and S.K. Singh

Introduction

India has emerged as a major producer of horticultural crops surpassing food grain production. This breakthrough has been achieved though diversified horticulture research and education in the country, particularly Indian Agricultural Research Institute, which has contributed immensely to the changed fruit production scenario in the country. The systematic work on improvement of horticultural crops started at IARI in 1940 with the establishment of small section of Vegetable crops in the erst-while Division of Botany. The Division of Fruits and Horticultural Technology was created in 1970 with the bifurcation of the Division of Horticulture and Fruit Technology, which was in existence at this institute since 1956. The division of Post Harvest Technology was carved out from the Division of Fruits and Horticultural Technology in February 2002 and was established as a separate Division in February 2002. Although the different divisions functioned independently under different heads, the overall charge of the PG activities was under common Professor of Horticulture irrespective of the sub-disciplines up to 2014. With the establishment of School of Horticultural Sciences in the year 2014, an independent professor for each disciplines were entrusted with the responsibility of academic affairs.

Keeping the Division's mandate in focus the teaching, training and research activities at the Division are aimed to strengthen the following objectives with a unique goal oriented approach.

- Undertake basic and applied research related to the current problems in fruit crops.
- Conduct teaching and training programmes to develop skilled human resource.
- Impart training in frontier areas of crop improvement and production technology.
- Develop modern instructional aids for teaching.

The research contributions of the students in the emerging field of basic and applied research have been outstanding and it gives strength to research programmes of the Division in terms of providing basic information, development of superior cultivars/varieties and refinement in production technologies from the point of view of productivity enhancement. During the last four and half decades, the Division has significantly contributed to the cause Horticulture in the country by way of advancing the frontiers of knowledge and by providing practical solutions to some of the most burning problems of the fruit industry.

The faculty of Division of Fruits and Horticultural Technology is actively involved in the teaching curriculum offering 66 credit hours distributed over 14 courses. The discipline has 14 faculty members. During the period 2007-2016, 28 students in M.Sc. and 36 students in Ph.D. graduated from which 63 publications were published in peer reviewed journals. The studies conducted by the students during 2007-2016 are highlighted in the following head/subhead.

Significant Student's Research Achievements

Genetic Diversity and Molecular Characterisation

A total of fifty mango genotypes representing different geographic regions were analyzed using 28 Simple Sequence Repeat markers including EST derived SSRs. DNA profiling of fifty genotypes detected 82 SSR loci. The size of the alleles detected ranged from 90 bp to 450 bp. Polymorphism information content (PIC) of SSR markers ranged from 0.038(RRP) to 0.666 (LMMA 3). The genetic relationships between mango genotypes from 0.45 to 0.88 with an average similarity coefficient of 0.612 indicated sufficient diversity among the genotypes studied. Cultivar specific fingerprints were developed for the genotypes. The probability of identity of molecular profiles of two genotypes taken at random was estimated to be 1.02 X 10-10 indicating high degree of confidence in the fingerprints. The findings of the study can be helpful not only for the varietal identification and maintenance of germplasm, but also for furthering the breeding prospects (Shareefa, 2008). In a study on genetic diversity and varietal identification in mango (*Mangifera indica* L.) using ISSR markers, 63 mango genotypes (*Mangifera indica* L.) was investigated using Inter Simple Sequence Repeat (ISSR) markers. A total of 334 scorable amplification products were detected with 28 ISSR primers of which 331 (99.10%) were polymorphic. Most of the primers (89.29%) exhibited 100% polymorphism. Primers UBC-812 and UBC-891 identified with the highest number of genotypes with unique fingerprints (53). The highest number of different fingerprints (58) was obtained with primer UBC-812. Primers UBC-812, UBC-891, UBC-808 and UBC-836 were found to be of high value for fingerprinting in mango as they were able to resolve 58, 57, 55 and 55 of 63 mango genotypes selected for the study, respectively (Samant *et al*., 2010).

Characterization of Forty eight mango (*Mangifera indica* L.) hybrids mango hybrids using SSR markers were analyzed using 17 simple sequence repeat (SSR) markers, which detected 59 scorable loci, of which 45 were polymorphic. The size of the alleles detected ranged from 100 to 480 bp. SSR markers was highly polymorphic with an average of 3.47 alleles per primer. SSRs gave moderate values of Polymorphism Information Content (PIC) and heterozygosity. The genetic relationship among mango hybrids, based on Jaccard's Similarity Coefficient values ranged from 0.38 (between H-1-13 and H-6-8) to 0.97 (between H-13-4 and H-13-7). The dendrogram, based on UPGMA cluster analysis, grouped the mango hybrids into three major groups. Cluster 'A' comprised of the five most diverse hybrids, namely, H-12-2, H-9-6, Pusa Arunima, H-9-5 and H-1-13. Two unique fingerprints were identified in hybrid H-9-6. The size of unique fingerprints ranged from 90 (MiSHRS-18) to 350 bp (MiSHRS-39). The Principal Coordinate Analysis also exhibited more or less similar distribution of mango hybrids. The tendency of clustering among mango hybrids revealed that they had stronger affinity towards female parent Amrapali (Singh *et al*., 2012).

Fifteen mango genotypes were characterized for their flowering behavior. Dushehari, Langra and Primor de Amoreira showed very early panicle initiation, i.e., before 10th February. However, Erwin and Husnara had very late panicle initiation, i.e., after 3rd March. Totapari Red Small, Pusa Arunima and Janardan Pasand had flowering during 10-20th February. Whereas, Zill, Tommy Atkins, Sensation, Neelum, Mallika, Amrapali and Bhadauran initiated panicles during 21st February to 3rd March. The maximum duration of flowering was noticed in Primor de Amoreira (41.5 days) and minimum in Husnara (13.0 days). Flowering duration was more than 20 days but less than 30 days in Amrapali, Sensation, Bhadauran, Mallika, Totapari Red Small and Janardan Pasand. Total number of flowers per panicle ranged between 133.30 in Bhadauran to 506.10 in Tommy Atkins. In general, per cent hermaphrodite flowers was less in early emerged panicles. The effective period of mango hybridization using diverse parental mango genotypes was found to be from 3rd week of February to mid March under Delhi conditions (Singh *et al*., 2014).

Relationships between different growth parameters with vigour indices of nucellar plants of 16 polyembryonic genotypes *viz.,* 'Bappakai', 'Carabao', 'Cecil', 'Chandrakaran', 'Combodiana', 'Kerala-1', 'Kerla-3', 'Kerala-5', 'Kurakkan', 'Muvandan', 'Mylepelian', 'Olour', 'Peach', 'Saber', 'Turpentine' and 'Vellaikolamban' were studied. Vigour index-I and vigour index-II had linear positive relationship with shoot length (V-I = 2.047 shoot length +

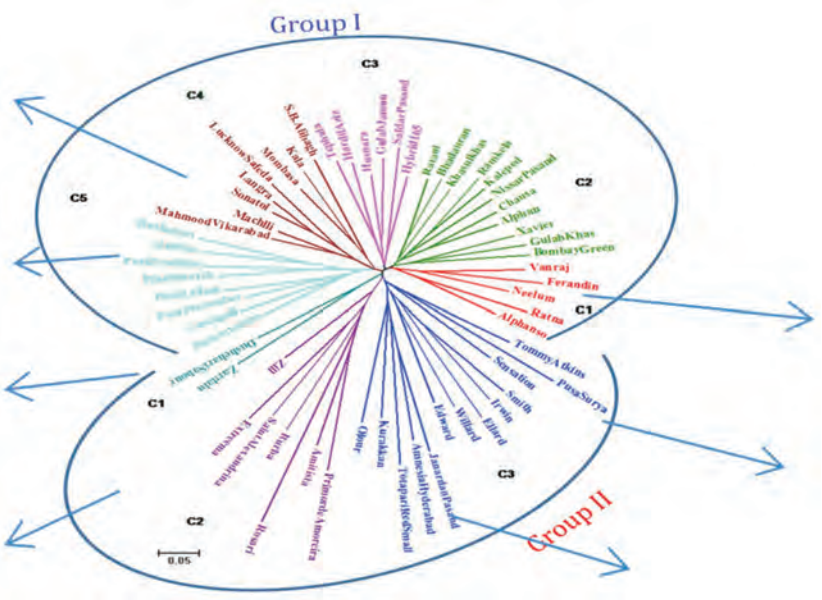
7.336, R^2 = 0.78 and V-II= 0.304 shoot length - 3.340, R^2 = 0.72, respectively) and internodal length (V-I = 6.708 internodal length + 39.45, R^2 = 0.40 and V-II = 1.187 internodal length + 0.727, R^2 = 0.52). The relationship between leaf growth and vigour indices showed very low degree of positive association with vigour indices. Root growth parameters like root length (V-I = 1.462 root length + 11.10, R^2 = 0.90 and V-II = 0.145 root length - 0.100, R^2 = 0.57, respectively) and root diameter (V-I = 9.517 root diameter + 7.927, R^2 = 0.63 and V-II = 1.696 root diameter - 4.919, R^2 = 0.82, respectively) had significantly higher positive regression relationship with vigour indices. The fresh and dry mass production of sixteen polyembryonic mango genotypes showed unique trend and all parameters showed comparatively higher association with vigour index-II than vigour index-I. Moreover, vigour index-II had high degree of positive relation with shoot fresh weight (R^2 = 0.89), shoot dry weight (R^2=96) and root dry weight (R^2=85). The results of the study suggests that growth parameters *viz.* shoot length, internodal length, root length, root diameter and fresh and dry weight of shoot and dry weight root are reliable criteria for determination of vigour in mango at nursery stage (Mahesh Kumar, 2008).

Studies on genetic variability for yield and quality traits in 50 guava (*Psidium guajava* L.) genotypes from Aligarh, Hathras and Muzzafarnagar in Uttar Pradesh and Ranchi and surrounding areas in Jharkhand showed marked variation in terms of vegetative growth, flowering, fruiting and physico-chemical characters. The correlation established among different characters can be utilized for selection of desirable target traits and the quantification of variation for horticultural traits would help in selection of desirable and potential parents for further utilization in improvement programme (Kherwar, 2015).

Genetic diversity analysis of indigenous and exotic apple genotypes using ISSR markers revealed 130 amplification products with 21 ISSR primers of which 130 (100%) were reported to be polymorphic. Primers based on $(AC)_n$ and $(AG)_n$ repeats produced more polymorphic bands. The number of bands generated by 21 ISSR primers ranged from 4 (UBC 840) to 10 (ISSR 3) with a mean of 6.19 bands per primer. The band size ranged from 300-1700bp. The Jaccard's similarity coefficient ranged from 0.435 to 0.822 suggesting that the 46 genotypes used in the study were highly divergent. The highest Polymorphism Information Content (PIC) was recorded to be 3.946 (ISSR 3). The overall Heterozygosity Averages (H_{av}) values of primers were 0.068 and 0.717 respectively for ISSR and UBC series (Kashyap *et al.*, 2010).

Functional Genomics

Sixty mango genotypes were phenotyped for 11 traits and genotyped using 100 leaf transcriptome derived genic SSRs. Out of 100, 87 polymorphic variability was observed for the studied traits and alternate bearing index and a maximum standard deviation and variance was observed for fruit weight (62.29 & 3879.79) and minimum for Hue value (0.10 & 0.01). Highest coefficient of variation was observed for the trait 'a*' (1.07), whereas minimum for Hue value (0.07) and in addition all the traits exhibited heritability (h^2) higher than 80%. The genotypic correlation values were higher than their corresponding phenotypic values for most of the studied traits and at both genotypic and

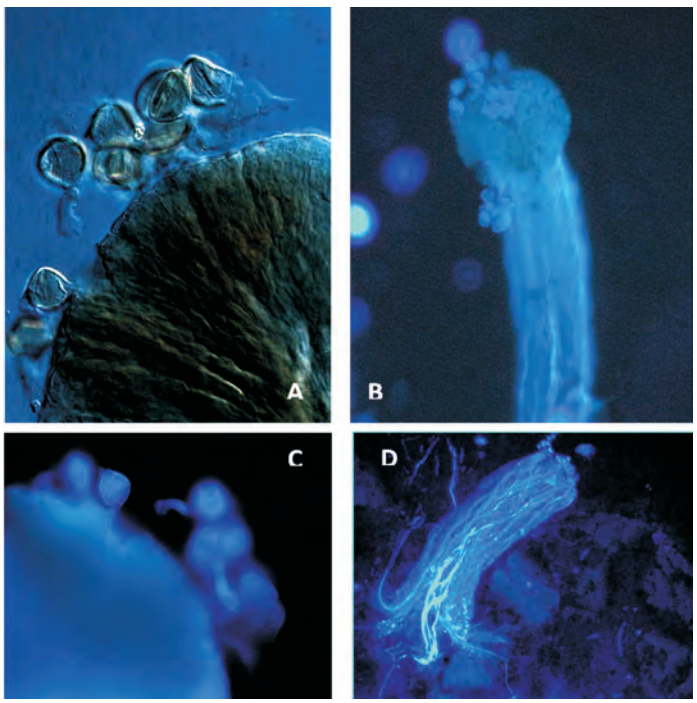


phenotypic level. Fruit peel 'b*' value was found positively and significantly correlated with 'L*' (0.615), 0.607) and a^* (0.481, 0.479), however the fruit length had significant and positive correlation (0.476, 0.466) with fruit weight at both levels. Likewise, fruit diameter also revealed positive and significant association with fruit weight (0.630, 0.626), however, it was negative with fruit length (-0.327, -0.324) at both genotypic and phenotypic levels, respectively. The data indicated that the highest positive direct effect at genotypic level was exhibited via fruit weight (5.74) followed by 'b^*' (1.008), however negative via 'a^*' (-0.013). In PCA analysis, first PC explained 28.9% of total variation and was positively related to fruit weight (0.383) and fruit yield (0.309), however the PC2 accounted to 23.9% of the total variation and the traits with the greatest weight on this component was 'l*' (0.396) and a^* (0.311). In genetic diversity analysis, a total of 263 alleles were amplified with an average of 3.0 alleles per SSR locus. (Shiv Lal, 2016).

Neighbour-Joining (NJ) clustering, Bayesian model-based structure and Principal Coordinate Analysis assigned the genotypes into two major groups from North-West and South East India, Florida, USA and Brazil. A total of six genic-SSRs in Generalized Linea Model (FLM), however five the Mixed Linear Model (MLM) were found to be associated with alternate bearing index and yield traits. Similarly, ten genic-SSRs in GLM and six in MLM were found to be associated with fruit peel colour traits ('l*', 'a^*', 'b^*', Hue value, chroma). For fruit quality traits (fruit length, weight, diameter and fruit shape index), total of 17 genic-SSRs in GLM and 11 in MLM were found to be closely associated. Furthermore, this study identified two genic-SSR loci (MSSR 146 and MSSR 174), which were associated with majority of the traits and will help in elucidating the molecular basis of the alternate bearing, fruit quality, and yield traits. The overall phenotypic variation explained by these loci ranged from 13.92-28.35% and 13.30-20.93% in the FLM and MLM analysis, respectively. (Shiv Lal, 2016).

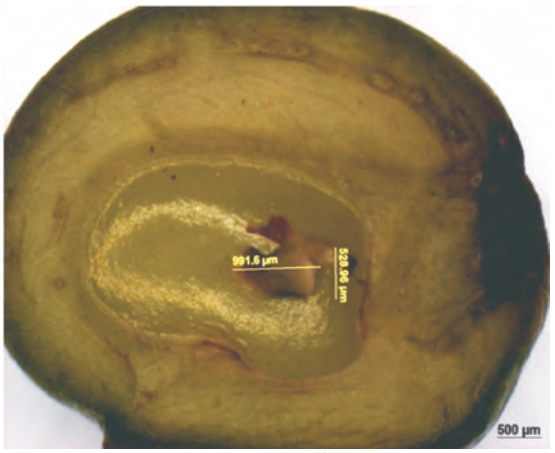
Crop Improvement and Character Association

An attempt was made to study pollen–pistil interactions in four mango cultivars 'Amrapali', 'Mallika', 'Pusa Arunima' and 'Pusa Surya'. Self-pollination resulted in rapid decline in fruit set than open-pollination in 'Amrapali' and 'Mallika' as compared to 'Pusa Arunima' and 'Pusa Surya'. At 48 h after self-pollination, pollen tube growth in the stylar region of 'Amrapali' and 'Mallika' was considerably slower than 'Pusa Arunima' and 'Pusa Surya' and their pollen tube reached up to two-third of the stylar region. However, in case of 'Pusa Arunima' and 'Pusa Surya' pollen tube reached up to the micropylar end. Self-pollination resulted in 75% degenerated ovules in 'Amrapali' and 'Mallika', which dropped within 21 days after pollination (DAP). In contrast, open-pollination resulted in only 20% degenerated ovules in these mango cultivars. The growth of fruitlets and ovules obtained from self- and open-pollination revealed that the fruitlets weight and dimensions of fruitlets and ovules were significantly less in self-pollinated as compared to open-pollinated 'Amrapali', 'Mallika' and 'Pusa Arunima', whereas no significant differences were observed in fruitlet weight and dimension of fruitlets and ovules obtained in self- and open-pollinated 'Pusa Surya'. The present findings bring out clearly that mango cultivars 'Mallika' and 'Amrapali' are self-incompatible, whereas, cultivars 'Pusa Arunima' and 'Pusa Surya' are self-compatible (Dutta *et al.*, 2013).

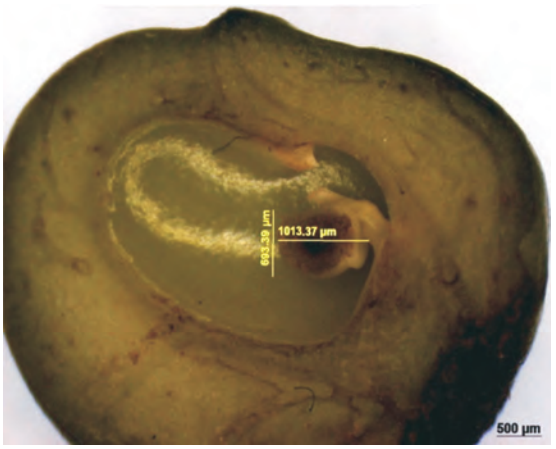


ESTD 9 SSR profile of 13-1 mother tree and multiple seedlings obtained from 13-1 stones. M, 100 bp marker; MT, mother tree; S, seedlings

DNA marker-based differentiation of zygotic and nucellar seedlings and identification of polymorphic microsatellite markers among parental mango genotypes in fruits of polyembryonic rootstocks, namely, Olour, Kurukkan and 13-1 were studied. 42 SSR primers were used, of which 9 primer pairs (LMMA 1, LMMA 2, LMMA 8, LMMA 15, ESTD 1, ESTD 2, ESTD 6, ESTD 9 and ESTD 10) were found informative and 33 primers were monomorphic. These 9 informative primers were used to differentiate nucellar and zygotic seedlings in polyembryonic mango rootstocks. In Olour, primer pairs, *viz*., LMMA1, LMMA2, LMMA8, ESTD 6 and ESTD 10 were found informative and ascertained the zygotic and nucellar origin of seedlings. In Kurukkan rootstock, ESTD1, ESTD2, ESTD6 and ESTD9 were informative and differentiated the zygotic and nucellar seedlings. In 13-1 rootstock, primer pairs LMMA 8, LMMA 15 and ESTD 9 were found to be informative in discriminating the zygotics from nucellar ones. Artificial hybridization was attempted using Amrapali as female and Dushehari, Janardan Pasand, Neelam, Pusa Arunima, Sensation and Tommy Atkins as male donor parents and the recovery of the hybrids was very low. Fourty-two SSR markers were screened for polymorphism among seven parental mango genotypes. Of which 13 primer pairs were polymorphic and 19 were monomorphic. Out of 13 polymorphic primers, only 3 primer pairs (LMMA 11, ESTD 9 and ESTD 10) could confirm the hybridity in different cross combinations (Kamlesh Kumar, 2015).



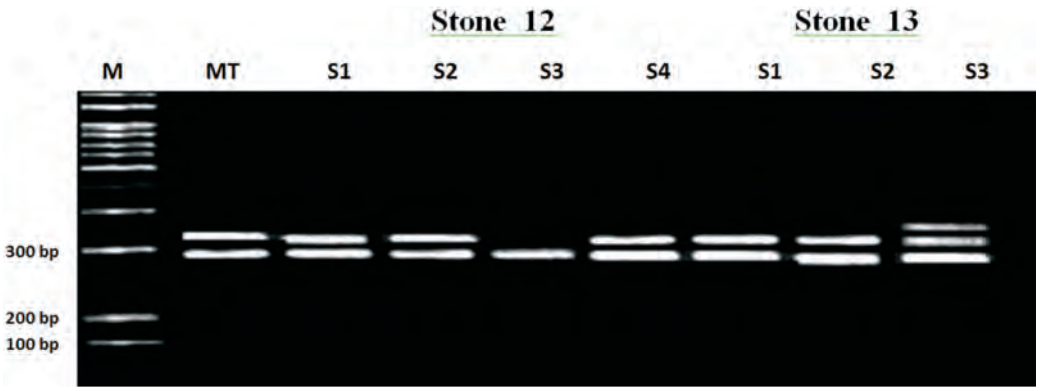
Amrapali



Mallika

Amrapali and Mallika fruitlets showing *in vivo* degeneration of ovules after 14 days of self pollination

For selection of potential parents and their utilization in mango improvement programme, growth, flowering and yield attributes of 50 full-sib mango hybrids belonging to Amrapali x Sensation cross along with their parents were studied. Results revealed significant variation for growth, leaf, flowering and yield characters. Maximum number of leaves were recorded in H-12-8 (48.60), maximum leaf area in H-13-7 (136.82 cm^2) and longest petiole length were recorded in H-13-4 (5.22 cm). Hybrids such as H-1-10, H-13-1



ESTD 9 SSR profile of 13-1 mother tree and multiple seedlings obtained from 13-1 stones. M, 100 bp marker; MT, mother tree; S,

and H-13-4 had the maximum inflorescence pigmentation (7.00 rating). Maximum number of hermaphrodite flowers per panicle was observed in H-1-1 (362.48), whereas male flowers were maximum in H-2-9 (1499.50). However, the maximum (Male: Hermaphrodite) sex ratio was noted in H-3-5 (0.94). The longest flowering duration was obtained in H-1-14 (20.68 days). Maximum fruit weight was recorded in H-1-5 (315.43 g) and yield in H-1-1 (44.25 kg/tree) (Rymbai *et al.*, 2016.)

An attempt was made to develop haploid plants in *Citrus grandis* using gamma irradiated pollens of *Citrus sinensis* and *C. limetta*. Among the different pollen parents used *C. sinensis* was found to be most suitable for the induction of seedlessness in *C. grandis* while *C. limetta* emerged as most appropriate pollen parent for haploid breeding The optimum condition for induction of haploid plants in *C. grandis* through *in situ* parthenogenesis is pollination with irradiated pollen at 300-400 Gy followed by *in vitro* ovule culture at 50 DAP (Kundu *et al.*, 2015).

To understand the mechanism of seedlessness in lemon cv. Kagzi Kalan, flower, pollen structure, pollen viability, *in-vitro* pollen germination and *in-vivo* pollen tube growth in self and cross pollination were studied. The inhibition of pollen tubes at the mid style under self-pollination treatments, and production of seeded fruits under cross pollination treatments indicated presence of self-incompatibility and site of inhibition of pollen tubes. Thus, gametophytic self-incompatibility (SI) caused seedlessness in Kagzi Kalan by blocking fertilization in the ovary (Kakade, 2016).

Hy. ER-R_2P_{36} **Hy. 16/2A-R_1P_2** **Hy. 16/2A-R_3P_{12}** **Hy. ER-R_1P_{19}**

Evaluation of grape genotypes for bioactive compounds were studied. The genotype 'ER-R_2P_{36}' matured earliest (last week of May) and was found most promising in terms of higher TSS (21.25°Brix), vitamin C (2.38 mg/100 g), seedless berries, higher TSS/TA ratio (31.25) and self thinning type loose bunches. Total monomaric anthocyanin content varied significantly from 40.04 ('Flame Seedless') to 791.04 mg C3GE/kg ('Pusa Navrang') fresh weight basis. Genotype 'Tempranillo' and '*Vitis parviflora'* showed significantly higher antioxidant activity in terms of DPPH, CUPRAC, FRAP and ABTS. CUPRAC had positive correlation with TEAC and TP; FRAP with TMA, TEAC, TF and TP; TMA with TEAC and TP; TEAC with TF and TP (Khalil Fitrat, 2015).

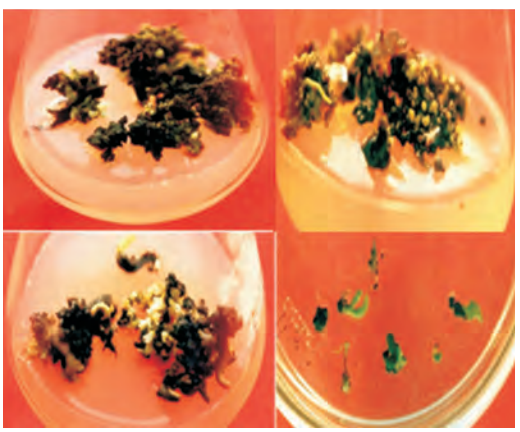
Evaluation of grape hybrids for nutraceutical and antioxidant traits showed that grape hybrid 'ER-R_2P_{36}' matured the earliest (last week of May) just 73 days after full bloom, with minimum growing degree days (1312.77), having maximum TSS content (23.55^{0}Brix), higher TSS:TA ratio (31.25), seedless berries, and self-thinning type loose bunch. The total monomaric anthocyanin content was found higher in hybrids over their parents. Total phenolics and total flavonoids were found in hybrid '16/2A-R_1P_2 and 16/2A-R_3P_{12}. The highest antioxidant activity measured in terms of CUPRAC and FRAP was found in hybrid 'Hy.16/2A-R_1P_{19} (Tanushree Sahoo, 2016).

Efforts were made to identify morphological and physiological parameters related to sex expression and validation of gene-linked molecular markers in papaya (*Carica papya* L.). Among morphological traits, black and brown seed colour was most reliable in predicting female and hermaphrodite plants. The purple stem colour in the seedling predicted hermaphrodite sex forms, though it was observed only in P-9-5 genotype. The lowest total phenols content (29.25 and 39.50 mg/100 g f.wt.) was in hermaphrodite plants of Red Lady and P-9-5. Leaf gas exchange parameters were also indicative of sex expression at seedling stage. The higher stomatal conductance indicated feminism in papaya followed by hermaphroditism. Seed colour as morphological marker was adjudged most stable marker for identifying female and hermaphrodite plants in gynodioecious varieties. Of the two marker systems tested, SCAR markers were most consistent. Markers, namely, T12, CFW+CRV and W11 were most informative to predict 100% sex forms and can be used by the breeders and commercial papaya growers to identify desired seedlings at early stage (Anjali Soni, 2015).

Studies on morphological diversity in papaya genotypes showed highest distinctness between p-7-15 and P-7-2 & P-7-2-V among the gynodiecious genotypes based on Euclidean's distance in a range of 2.16 to 7.39. The two wild species *Vasconcellea cundinamarcensis* and *V. cauliflora* were observed in same cluster No. 'I' along with P-9-25 among the dioeciuos genotypes. The gene diversity range 0.29 to 0.73 with an average of 0.59. The maximum gene diversity (0.73) was observed in primer P3K2981YCO followed by (0.66) in primer CPM1606CC and (0.65) in primer P3K2696CC. Marker P3K2981YCO had produced the highest PIC (0.68) followed by CPM1606CC with 0.58, while marker CPM1774CC had the lowest PIC of 0.25. The maximum observed heterozygosity (0.43) was in two markers, namely, ctg27cc and P3K2696CC. The higher level of heterozygosity (48%) was recorded within the individuals among population. Based on unweighted Pair Group Method of Arithmetic averages (UPGMA) dendroagram, the genotypes were divided into two broad clusters represented by 6 and 17 genotypes, respectively (Kaluram, 2016).

In vitro and mutagenesis studies

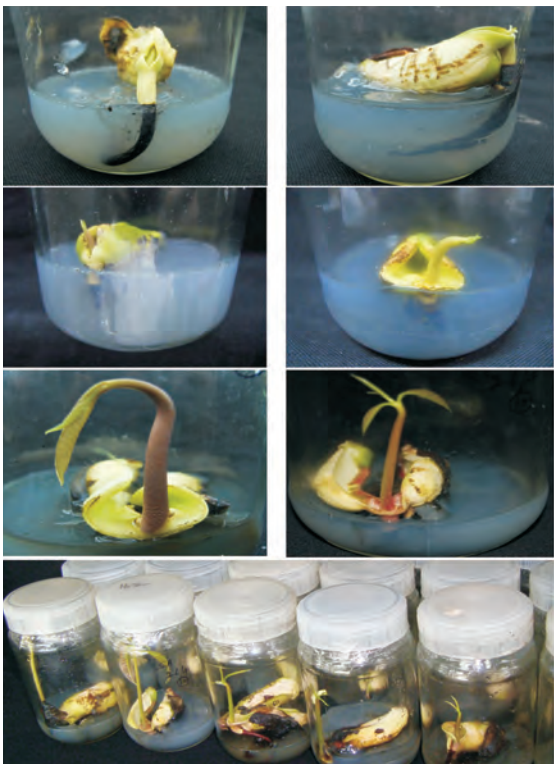
In mango, effort was made to establish *in vitro* cultures from foliar and nucellar tissues in genotypes Amrapali and Dashehari and rootstocks Olur and Kurukkan. Amongst the different explants, the maximum culture establishment was in nodal segment followed by leaf segment. The correlation between polyphenol activity and phenol content with regard to explant survival was found to be 0.73 and 0.91, respectively. When explants from *in vitro* raised nucellar seedlings were cultured there was significant improvement culture establishment. Embryogenic callus induction was noted after 45.8 days on to medium comprising B5 (macro) + MS (micro + organics) supplemented with 400 mg/l L-glutamine, 200 mg/l casein hydrolysate and 20% coconut water (v/v) along with 2.0 mg/l + 0.5 mg/l BAP. Somatic embryos were formed on the hormone-free medium. (Hare Krishna *et al.*, 2008).



Embryo rescue and DNA marker-assisted hybridity analysis in mango (*Mangifera indica* L.) cv. Amraplai revealed that the medium comprising of B_5 (macro) + MS (micro + organics) with sucrose (40 g l^{-1}), casein hydrolysate (200 mg l^{-1}), L-glutamine (400 mg l^{-1}), IAA (0.5 mg l^{-1}) and BAP (2.0 mg l^{-1}) at pH 5.7±0.1 under complete dark condition showed better establishment and maturation of young embryos. Similarly, germination of matured embryos was best on medium comprising of B_5 (macro) + MS (micro + organics) supplemented with sucrose (30 g l^{-1}), casein hydrolysate (200 mg l^{-1}), L-glutamine (200 mg l^{-1}), BAP (1.5 mg l^{-1}), GA_3 (1.0 mg l^{-1}) under 12/12 h light and dark cycle. In order to ascertain the hybridity of progenies using precise system, total 31 genomic and 11 EST derived SSRs were used. Only two SSR primer pairs, *viz.,* ESTSSR 9 and ESTSSR 10 were informative and capable to ascertain the hybridity

of progeny population obtained from crosses Amrapali x Sensation and Amrapali x Tommy Atkins. These SSRs in combination with other SSRs in mango may be used for hybridity analysis in future (Arun Kishor, 2012).

To investigate the impending effect of bacterial bio-agents in controlling *Fusarium mangiferae* in mango, *in-vitro* studies showed the potential of *Bacillus subtilis,* a gram positive bacteria and *Pseudomonas fluorescens,* gram negative bacteria, in reducing the growth of *F. mangiferae*. Both *Bacillus subtilis* and *Pseudomonas fluorescens* inhibited the growth of *F. mangiferae.* However, *in vivo* studies carried out for three consecutive years revealed the potential of only *Bacillus subtilis* in control of mango malformation disease with 58.33% reduction in mango malformation over the untreated mango trees. (Tanushree Debnath, 2014).



Different stages of *in vitro in ovulo* embryo germination (A-G). A, root initiation and root growth; B, initiation of shoot and root elongation; C-F, different stages of *in vitro* plumule growth; G, *in vitro* germinated plants of mango cv. Amrapali.

Protocol was standardized for *in-vitro* propagation in citrus rootstocks, *viz.*, *Jattikhatti, Karnakhatta* and Troyer citrange. Culture establishment was best on MS medium + 3mg/l BAP + 0.5mg/l NAA. Maximum rooting of micro-shoots was recorded in rootstock Troyer citrange (94.8%) followed by *Karna khatta* (91.1%) on the half-strength MS medium supplemented with 2.0half-strength MS medium supplemented with 2.0 mg/l IBA + 200 g/l activated charcoal and 30 g/l sucrose. Rooting performance of different genotypes on MS medium supplemented with IBA (2 mg/l) + AC (200 mg/l). Plantlets of each genotype after hardening were tested for *Citrus Tristeza Virus* using DAC-ELISA and the representative plants were found virus-free. Bio-hardening on *Jatti khatti* and Troyer citrange was better with *Glomus laevis,* while *Karna khatta* responded best with *Glomus gigantia* and Pusa mixed AM strain. Mycorrhizal plantlets showed improved physiological and biochemical status, *viz.* photosynthetic, respiration rate, relative water content, phenols, proline, membrane injury index *etc*. (Wasim H. Raja, 2012).

An efficient protocol for *in vitro* multiplication and bio-hardening of four grape rootstock genotypes, namely, Dogridge, SO4, H-44 and 3309C was developed followed by genetic stability analysis of micropropagated plantlets using RAPD and ISSR molecular markers. Nodal segments pre-treated with Bavistin® (2 g/l) + 8-HQC (200 mg/l) for 2 h resulted in the highest culture establishment (65.44%) on MS medium supplemented with 2.0 mg/l BAP + 0.2 mg/l NAA + 200 mg/l activated charcoal. Glass jars with PP cap filled with coco peat: perlite: vermiculite (2:1:1) was found ideal for *in vitro* hardening (82.0%) in shortest period (45.1 days) for *ex vitro* transfer. The bio-hardening agent, *G. manihotis* exhibited the highest *ex vitro* plantlet survival (83.6%). Genetic uniformity of *in vitro* raised grape populations was examined using 12 RAPD and ten ISSR primers. The relative *in vitro* NaCl tolerance of the different grape rootstocks could be ranked as Dogridge > H-144 > SO4 & 3309C. (Alizadeh *et al*., 2008).

The *in vitro* plantlets of grape were subjected to 0, 5, 15, 20, and 25 Gy doses of gamma rays irrdiation. The highest mean tissue survival was in control (83.78%) followed by irradiation dosage of 5 Gy (80.95%). The dose of 10 Gy gave the survival of 46.66% and 10 Gy dose was ascertained as LD_{50}. The highest shoot bud sprouting (57.59%) was noted in Pusa Navrang followed by Pearl of Csaba (55.59%). Mean shoot abnormalities was significantly the highest (50.29%) in H-76-1 followed by Julesky Muscat (49.38%). Genetic status of the putative mutants was ascertained using molecular analysis, *viz.*, RAPD and SSR markers. Primers OPA01 & OPP04 (RAPD) and VVMD-14 (SSR) were the unique primers. The next higher dose (15 Gy) of gamma rays resulted in development two solid mutants. Two primers RAPD-OPA1 and SSR-VVMD-14 were the most informative for identification of grape mutants. (Rahul Dev *et al.*, 2015).

Five grape genotypes, namely, Dogridge, Salt Creek, 110 Ritcher, St. George & *V. parviflora* were mutated using single-node explant collected from field grown vines and *in vitro* cultures using gamma rays (0-50 Gy & 0-30 Gy, respectively). The LD_{50} dose of gamma rays was 20 and 15 Gy, respectively for the two types of explant. The LD_{50} value for Dogridge (single-node segment) was 30 Gy, while it was 20 Gy for other rootstocks. The single node segments of Dogridge (51.52%) recorded the highest explant survival followed by St. George (41.72%), whereas, the lowest was observed *V. parviflora* (30.81%). The genotypes Dogridge, 1616C and 1103 Paulsen had LD_{50} dose as 20 Gy; Salt Creek and 140 Ruggeri at 15 Gy, whereas, in St. George, *V. parviflora*, 110 R and 1613C it was 10 Gy. The highest explant survival of *in vitro* cultures was observed in 1616 C (68.05%). Screening of parental lines with EST SSRs specific for salinity trait in plant produced amplification in the 15 genotypes and produced 308 reproducible putative alleles with an average of 1.67 alleles/ locus, indicating their efficient transferability. Maximum number of alleles (26 alleles) with 19 EST SSRs were produced in the rootstock 140 Ruggeri and 1103 Paulsen (24 alleles) indicating maximum salinity tolerance. A total of 73 mutants induced from nine rootstock genotypes were screened with EST SSRs, out of which 18 solid mutants having higher salinity tolerance were identified. Maximum mutants were obtained in 20 Gy irradiation dose. (Kalpana *et al.*, 2016).

Modelling Epidemiology of Mango Malformation

A study on prediction model for mango malformation caused by *Fusarium mangiferae* leading to losses upto 60% was developed using multiple linear regression, multiple criteria analysis, geo-statistical, GIS and logistic prediction of spatial distribution of mango malformation at regional scale. The developed model effectively predicted the occurrence of malformation in Uttar Pradesh and Andhra Pradesh (Reddy, 2007).

The potential of using hyperspectral reflectance data to discriminate healthy and malformed mango panicles revealed Normalized Difference Vegetation Index (NDVI) as the best parameter in differentiating the healthy (0.495) and malformed (0.751) mango panicles. The red-edge value (0.0106 and 0.0113) and red edge (692 and 697) position was significantly different for healthy and malformed mango panicles, respectively. Hence, spectral reflectance based indices proved to be a potential tool in the identification of malformed mango panicles spatially through remote sensing data (Nagraja *et al.*, 2014).

Physiological and Biochemical studies

Biochemical studies and phenotyping of 50 mango varieties revealed that they had different degrees of resistance to malformation. Mango cultivars Machali, Mahamood Vikara, and Swarnika were highly susceptible, while Bhadauran and Ellaichi were resistant to malformation. Assessment of mango varieties for different defense responding biochemical compounds revealed significantly higher levels of PPO, LOX, peroxidase and

catalase activity in mango varieties resistant to malformation and can be used as biochemical indicator for screening mango germplasm against malformation disorder (Singh *et al.*, 2012).

An investigation to identify physiological and biochemical markers conferring resistance to mango malformation revealed that out of 44 mango cultivars Bhadauran and Elaichi were resistant (0% malformation) and Rosary, Rataul, Alphan and Ramkela (<10% malformation) were tolerant to mango malformation. The findings of the study suggest that physiological and biochemical parameters in the mango leaves, can be used as marker for identification of mango cultivars resistant or tolerant to malformation. Further, amplification of resistance gene analogues (RGA) in different mango cultivars lead to the conclusion that the sequence can be used for isolating full length R-genes as new technique for identification and isolation of resistance gene in mango against mango malformation (Saboki *et al.*, 2012).

Radio-tracer studies were carried out to study the translocation and distribution of labeled ^{14}C from leaves to fruit and *vice versa* and within the fruit during granulation in Kinnow mandarin and sweet oranges cv. Mosambi during 2007-08. After applying the radioactive $^{14}CO_2$ and sucrose ^{14}C to the fruit/ leaf/rind, the presence of ^{14}C in the different tissues was studied. Survey suggested that the citrus orchards in Sriganganagar were deficient in P, K, Ca, Zn and Cu contents. The granulated citrus fruits had low weight, poor juice, lower TSS, sugars, ascorbic acid and pectinesterase activity with large size, more peel and dry matter. Two sprays of GA_3 30 (ppm) + NAA 400 (ppm) reduced granulation incidence. Radio tracer studies ($^{14}CO_2$) confirmed reduced (15 times) translocation of metabolites to the granulated fruits compared to healthy fruits. Injection of ^{14}C-sucrose in the fruit albedo showed reverse movement of metabolites from fruits to the leaves. (Karuppasamy T., 2009).

In a mtagenesis study, impact of gamma (γ) irradiation and ethyl methanesulfonate (EMS) showed significant reduction in survival rate (%) and leaf area of the Kinnow mutants developed at 20 Gy and 0.5% EMS. The mutants at these doses, however maintained high relative water content (RWC) with lower leaf membrane injury. Maximum chlorophyll breakdown in the mutants were observed due to 20 and 15 Gy doses. The induced biochemical changes revealed enhanced proline accumulation in the leaves at 0.2% EMS, while elevated phenol contents were recorded in the mutants at 20 Gy followed by 15 Gy and 0.5% EMS doses. Mutagenic treatments at 20 and 15 Gy upregulated superoxide dismutase, catalase and peroxidase enzyme activities. The study revealed that both physical and chemical mutagens at higher doses have the potential to generate much needed variation in 'Kinnow' mandarin with respect to physio-biochemical traits including antioxidant enzyme and should thus help in evolving desired mutants for economic traits including seedlessness and subsequent use in future breeding programmes (Mallick *et al.*, 2016).

Plant Bio-regulators and salinity stress

The effects of paclobutrazol (PBZ) on 1-year-old plants of 'Olour' mango subjected to NaCl stress revealed significant reduction in mortality of saline-treated mango plants following the application of PBZ. The treatment with PBZ increased the relative water and chlorophyll contents of mango seedlings, and reduced membrane injury, under salt stress. Furthermore, application of PBZ consistently and significantly lowered the Na^+ and Cl^- ion contents of leaves and roots. The Na+ content of leaves on saline-treated plants was reduced by 1.96- to 2.12-fold, whereas Cl^- ion contents were reduced by 22 – 39% by PBZ treatment compared to salt-treated seedlings without PBZ. The findings of the study suggest a role for PBZ in promoting the avoidance of salt stress in mango by increasing the levels of photosynthetic pigments, water content, K^+ uptake and accumulation, and by reducing defoliation, the membrane injury index, and the uptake and accumulation of harmful Na+ and Cl– ions and hence may be used to improve the yields and quality of mango trees grown in salt-affected areas (Kishor *et al.*, 2009).

Studies on polyembryonic rootstocks cv. Kurukkan and monoembryonic (non-descript) mango seedlings revealed that the application of polyamines improved vegetative growth of mango plants. The effect of spermidine was more pronounced than putrescine in improving vegetative growth of both mango rootstocks. NaCl treatment with or without polyamines showed significant reduction in secondary roots. However, applications of polyamines have increased the number of secondary roots per plant in both rootstocks under salt stress. Salt-treated plants showed very high degree of defoliation while it was significantly reduced by application of spermidine (2.18 fold) and putrescine (1.54 folds decrease). Modulating effect of polyamines on mango plants under salt stress may be attributed to increased photosynthetic pigments, relative water content, proline accumulation, potassium and calcium uptake and accumulation and enhanced activities of antioxidant enzymes and by reducing membrane injury and sodium and chloride uptake and accumulation in mango plants (Kanchan Lata *et al.*, 2011).

Studies on effect of rootstocks and scion combination on modulation of physiological alteration in mango cv. Amrapali grown under NaCl stress suggested that lipid peroxidation in leaves appeared to be scion dependent. Moreover, difference in glutathione reductase (GR) and peroxidase activities in different rootstock scion combinations indicated the significant role in upregulating GR and peroxidase activities. Decrease in transport of Cl^- and Na^+ from the roots of Olour to scion of Amrapali and decreased Na^+ transport from the roots of Kurakkan and non-descript seedling rootstocks to the scion of Amrapali suggested existence of Cl^- exclusion mechanism in Olour rootstock. Hence Olur and Kurakkan can be used as mango rootstocks in areas where irrigation water contains NaCl salts (Dayal *et al.*, 2011).

Biochemical and salt ion uptake responses of seven mango (*Mangifera indica* L.) rootstocks ('Moovandan', 'Bappakai', 'Nekkare', 'Kurukkan', 'Olour', 'Terpentine', and 'Chandrakaran') to NaCl stress revealed enhanced activities of peroxidase (POX), catalase (CAT), and superoxide dismutase (SOD), and the concentrations of malondialdehyde (MDA), proline, and trehalose. The highest anti-oxidant enzyme activities were observed in the rootstocks 'Olour', 'Bappakai' and 'Terpentine' at the highest level of salinity (150 mM NaCl). Leaf proline concentrations increased most in 'Olour', 'Kurukkan', and 'Terpentine' with increasing levels of salinity. The concentration of Na^+ ions in leaf tissues increased to a maximum (123.5%) of the respective (0 mM NaCl) control in 'Chandrakaran', while 'Bappakai' had the highest Na^+ ion concentration in root tissues at the highest level of salinity (77.3%). the maximum increase (109.1%) in leaf Cl^- ion concentration was recorded in 'Moovandan', while 'Chandrakaran' had the highest concentration of Cl^- ions (139.3%) in its root tissues when irrigated with water containing 150 mM NaCl. Our results suggest that the salt-tolerant behaviour of the mango rootstocks 'Olour' and 'Terpentine' may be due to their ability to inhibit the uptake of Cl^- and Na^+ ions, combined with higher accumulations of proline. The findings of the study suggest that Olour and Terpentine may tbe used to improve the yield and quality of commercial mango cultivars grown in salt-affected areas (Pandey *et al.*, 2014).

Studies on effects of exogenous application of polyamines on growth and physiochemical changes in Troyer citrange under NaCl stress revealed the effectiveness of Polyamines such as putrescine (Put), spermidine (Spd) or spermine (Spm) in restricting the accumulation of Na^+ ion. However, Put had more pronounced effect on restricting Cl^- ion accumulation and enhancing the accumulation of K^+ ion in leaf tissues under saline condition. Further, application of Spm was found more effective in improving the activity of SOD and proline content. Overall, polyamines such as putrescine and spermidine was found effective in mitigating salt stress in salt susceptible citrus rootstock, Troyer citrange (Soyimchten, 2010).

Application of paclobutrazol (PBZ) and/or putrescine (Put) in the salt-susceptible citrus rootstock *Karna khatta* under NaCl stress reduced the membrane injury index and increased relative water content, photosynthetic rate, and pigments content under saline conditions. Application of PBZ or Put alone or in combination also improved the activities of SOD and peroxidase and proline content under saline conditions. It also increased K^+ and reduced Na^+ and Cl^- concentrations in leaf tissues. Application of 250 mg l^{-1} paclobutrazol alone or in combination with 50 mgl^{-1} putrescine could improve the tolerance of salt-susceptible *Karna khatta* by regulating absorption and accumulation of ions and improving antioxidant enzyme activities (Sharma *et al.*, 2011).

Effect of NaCl in the irrigation water on growth, anti-oxidant enzyme activities, and nutrient uptake in five citrus rootstocks [sour orange (*Citrus aurantiam*), Attani-2 (*C. rugulosa*), Troyer citrange (*C. sinensis* × *Poncirus trifoliata*), *billikhichlli* (*C. reshni*), and RLC-6 (*C. jambhiri*)] were studied. Results indicated that higher levels of proline accumulation and leaf abscission could be used as indicators for screening citrus rootstocks for resistance to NaCl stress. Sour orange and Attani-2 were able to exclude Cl^- ion, whereas Troyer citrange appeared to exclude Na^+ions at lower levels of NaCl. Overall, salt-tolerance increased in the following order: Troyer < *billikhichlli* < RLC-6 < Attani-2 < sour orange (Patel *et al.*, 2011).

The response given by the photosynthetic pigments, antioxidant enzyme activity and nutrient accumulation of Kinnow (*Citrus nobilis* × *C. deliciosa*) budded on *Citrus jambhiri* Lush rootstocks (var. *Jatti Khatti*) to triazole compounds revealed an increase in the accumulation of K^+ and Ca^{2+} ions and inhibited the accumulation of Na^+ and Cl^-, ions resulting in an improvement in the K^+/Na^+ and K^+/Cl^- ratios. For PBZ-treated plants, the K^+/Na^+ and K^+/Cl^- ratios in leaf tissues increased 2.4- and 2.1-fold compared to plants treated with NaCl alone, while HEX had a more striking effect on Ca^{2+} upregulation and significantly (1.5-fold) increased leaf Ca^{2+} under stress. Thus, Triazole compounds, paclobutrazol (100 mgl^{-1}) and 1,2,3- triazole (20 mg l^{-1}) was found effective in protecting Kinnow plants budded on *Jatti khatti* under NaCl stress (Kakade *et al.*, 2011).

Studies on effect of value-added products of animal farm waste in grapefruit cultivar Marsh seedless revealed that combined application of chemical fertilizers and NADEP compost or farm yard manure could be better combination for getting maximum plant growth, flowering and fruit set in grapefruit cultivar Marsh SL as well as improving soil fertility (Awachare, 2011).

Salinity stress inhibited plant growth, chlorophyll contents, RWC, and restricted the accumulation of beneficial nutrients in all rootstock-scion combinations. Lower accumulation of Cl^- in leaf tissues on RLC-6 or *Attani*-1 and Na^+ on RLC-6, *Karna khatta* and RLC-4 rootstocks suggested that RLC-6 could exclude both Cl^- and Na^+, Attani-1 could only Cl^-, while *Karna khatta* and RLC-4 rootstocks could exclude Na^+ from leaf tissues. Based on the overall performance, RLC-6 can be used as citrus rootstocks in areas where irrigation water contains NaCl salt. Attani-1, *Karna khatta* and RLC-4 may be useful as potential parent to develop both Na^+ and Cl^- excluder rootstock in citrus (Anees, 2013).

In vitro and *in vivo* screening of grape rootstock genotypes for NaCl tolerance were studied by grouping grape rootstock genotypes into three categories based on salt tolerance, namely, (i) Tolerant type (Salt Creek and Dog Ridge A), (ii) Moderately tolerant (SO4, H-144, 1616C, 1613C, Male hybrid and Degrasette), and (iii) Sensitive type (99R, 110R and Dog Ridge B). Rootstocks SO4 and H-144 tolerated *in vitro* salinity up to 40 mM NaCl, Male hybrid up to 60 mM for 50 d, while 1616C tolerated 80 mM NaCl upto 50 d and Salt Creek upto 75 d without significantly affecting tissue mortality. The mechanism of NaCl tolerance in grape rootstock genotypes were found to be (i) due to their ability to exclude Na^+ (99R, 110R, 1613C < Dog Ridge B, Salt Creek, H-144) and/or Cl^- ions (1616C < 1613C, SO4 < 110R < H-144). (Ramajayam *et al.*, 2013).

Biotic Stress tolerance

Forty five diverse grape genotypes including rootstocks, wine making and table purpose types were evaluated under sub-tropical conditions based of eight morphological parameters for their reaction to anthracnose [*Elsinoe ampelina* de Bary) Shear]. Disease severity index during the peak disease incidence, *i.e.* 1st September showed Dog Ridge, Male Hybrid and H-144 to be extremely resistant (DSI = <5.0); St. George, SO4, and 110-R as highly resistant. Rootstock genotypes showed higher resistance (DSI = 5.88) followed by wine making genotypes (DSI = 42.1), whereas, table purpose genotypes were least resistant (DSI = 57.11). Biochemical analysis revealed an increase in activities of defence

related enzymes like peroxidase (PPO), polyphenol oxidase (POD), phenylalanine ammonia lyase (PAL) and malondialdehyde (MDA) content upon disease infection. (Suman Beniwal, 2013).

Further studies on powdery mildew (*Erysiphe necator* (Schw.) Bur. and anthracnose (*Elsinoe ampelina* de Bary) Shear] was undertaken on 32 grape genotypes. Disease severity index (DSI) calculated during the peak anthracnose incidence, *i.e.*, 1st week of September showed that *Vitis parviflora* to be immune (DSI = 0), Male Hybrid as extremely resistant (DSI = 4.15), Pusa Navrang as resistant (DSI = 21.90). Based on the DSI for powdery mildew, the genotype Male Hybrid was found immune (DSI = 0), while *V. parviflora* was rated as highly resistant (DSI = 7.25), Pusa Navrang as resistant (DSI = 19.46). The concentrations of total phenols was highest in resistant genotypes, *V. parviflora* (5.42 mg/g FW), while highest reduction in total and reducing sugars in susceptible genotypes, namely, Hybrid Seedless (7.16, 3.59 mg/g FW), followed by Perlette (7.03, 3.52 mg/g FW) and H-70-56 (7.27, 3.37 mg/g FW) as compared to resistant genotypes. Activity of enzymes like peroxidase, polyphenoloxidase and phenylalanine ammonia-lyase were increased upon disease infection. Correlation between DSI for anthracnose and biochemical parameters showed that total chlorophyll (r = -0.680), total phenols (r = -0.918), total and reducing sugars (r = -0.620, r = -0.773) and activities of PPO (r = -0. 832), POD (r = -0.764) and PAL (r = -0.821), were significantly negatively correlated. Marker VMCNg2f12.1 (7 alleles per locus) was identified as the most informative, while VMC4f3.1 (PIC: 0.75) showed the maximum polymorphism. Two SSR markers, *i.e.*, VMCNg2f12.1 and VMC4A5 produced four unique alleles exclusively in the disease resistance genotypes. These alleles were shown to be flanked to the powdery mildew resistance locus *Run*1. (Pawan S. Gurjar, 2015).

Abiotic stress tolerance

The influence of *Glomus intraradices* colonisation on growth and reactive oxygen metabolism of Cleopatra mandarin (*Citrus reshni* Hort. Ex Tan.) seedlings treated with mycorrhizal helper bacteria containing a mixture of *Bacilus subtilis* and *B. megaterium*, *Azospirillum braisilence* or *Providencia* sp.) showed that mycorrhizal inoculation increased plant growth and nutrient acquisition independent of the water regime. Exposure of plant to drought stress led to generation of superoxide radicals and hydrogen peroxide in leaf tissues, however, their concentrations were lower in seedlings inoculated with *G. intraradices* and phosphate solubilising bacteria. This particular treatment also increased total glutathione content and enhanced antioxidant enzyme activity in plant and microbial activity in soil. (Barman *et al*, 2015).

Canopy Management

Impact of pruning intensities and plant densities on growth, fruitfulness and quality of fruits in Amrapali planted was studied. Shoot bud sprouting and length increased with pruning severity and plant densities. Light penetrance and canopy temperature were the maximum in 60 cm pruning intensity (15.13 k lux; 25.36^{0}C) and 3.0 m × 3.0 m spaced trees. Endogenous hormones (IAA, GA, ABA), phenolics and PPO activity were enhanced with increase in pruning intensity and spacing. Unpruned trees produced the maximum number of malformed

panicles, while in 45 cm pruning intensity it was minimum. The maximum fruit drop (87.96%) was witnessed in 60 cm pruning intensity, while maximum fruit yield (253.34 q/ha) was obtained from 30 cm-pruned trees planted at 2.5 m × 2.5 m. Fruit pulp was maximized in 60 cm pruned trees, while TSS was non-significantly affected. Total sugars, carotenoids and ascorbic acid contents were improved. (Subodh Kumar Singh, 2008).

In another study, three mango cultivars (Amrapali, Mallika and Dashehari), were compared for their pruning response (30, 60 and 90 cm from apex). Cultivar Amrapali registered the highest number of shoot buds, maximum light penetration, lowest shoot length, canopy volume, tree girth, canopy temperature *etc*. The severe pruning led to highest number of shoot buds, smallest shoot length, and canopy volume and highest light penetration and canopy temperature. Minimum number of shoot buds, shoot length, lowest net photosynthetic rate, light penetration and canopy temperature was noticed in un-pruned trees. Dashehari registered the maximum chlorophyll '*a*', proline content, GA-like substances (1.36, 1.31 µg g-1 FW). Cultivar Amrapali had higher P, Ca, S, Cu and Mn contents. The incidence of floral malformation was least (9.40 & 11.21%) in moderately pruned trees. (Sanjay K. Singh, 2008).

Rootstock studies

In a polyembryonic rootstock trial of mango with different scion varieties Olour/Pusa Arunima, Kurakkan/Pusa Surya, Kurakkan/Amrapali, K-5/Amrapali, K-5/Mallika and Kurakkan/Dushehari combinations revealed their excellence over other rootstock/scion combinations with respect to horticultural traits (higher tree vigour, yield, pulp recovery, Total β-carotenoids content and lower acidity). It also shows that metabolism of Kurakkan and/or K-5 is better adapted for N, K, Ca, Mn, Fe and Zn accumulation for most of the mango cultivars studied (Dayal *et al*., 2016).

Ten grape rootstocks, namely, Dogridge A, Dogridge B, Salt Creek, 1613, 1616, St. George, SO4, 1103 P, Teleki 5A and H-144 were evaluated for rooting and other nursery traits. The highest shoot growth rate was registered in Dogridge A. Rootstock SO4 rooted early (30.8 days), while it was most delayed in Dogridge A (44.8 days). Grafting during February at 30 cm height on pencil tick rootstock gave the maximum success. Physiological parameters like, photosynthetic and respiration rates and leaf relative water contents were found to improve with grafting age. Dogridge A was found the most responsive followed by 1613 and Salt Creek. Biochemical parameters like total chlorophylls, phenols, reducing sugars, nitrate reductase activity and polyphenol activity (catecholase and cresolase) were found higher on Dogridge A rootstock. Tissue nutrient (N, P, Ca, Mg, Cu and Fe contents) was higher on Dogridge A compared to other rootstocks. (Verma *et al*., 2012).

In a study on effect of different rootstock on Kinnow mandarin, significant variation were recorded with respect to morphological, physiological and bio-chemical activity of Kinnow. Scion leaves of Kinnow on *Karna khatta* and rough lemon maintained higher relative water content (RWC). *Jatti khatti* rootstock promoted higher photosynthetic rate (*A*) while stomatal conductance (*gs)* and transpiration (*E)* was higher on *Jatti khatti* and rough lemon. Significantly higher superoxide dismutase (SOD), peroxidise (POD), and glutathione reductase (GR) activities were recorded on rough lemon, whereas catalase (CAT) activity and proline accumulation were higher in Kinnow leaves on sour orange rootstock. Furthermore, up regulated polyphenoloxidase (PPO) activity was observed in scion leaves of Kinnow on *Jatti Khatti*. Variation in gas exchange parameters and biochemical parameters clearly indicated that rootstocks had the differential ability to alter the physio-chemical attributes of Kinnow scion. The study suggest that enhanced photosynthetic rate, proline content along with up regulated antioxidant, and PPO activities can be used as a criterion for selecting appropriate rootstock adapting different soil and climatic conditions. (Sunil Kumar, 2015).

Phytophthora inoculation through infected roots tended to show the highest rotting of feeder roots and found best method for the pathogenic studies in citrus rootstocks. The inoculation of *Phytophthora nicotianae* significantly increased the superoxide radical, H_2O_2 level and the activity of enzymes, viz., SOD, GR, CAT, POX, ß1, 3-glucanase and protein content in rootstock seedlings, while morphological parameters and nutrients content were decreased. The order of relative tolerance of rootstocks against *Phytophthora nicotianae* was Troyer citrange > sour orange > Italian rough > RLC 5 > RLC 6 > RLC 7 > Grambhiri > Rough lemon (Kuldeep Singh, 2016).

Integrated Nutrient Management

Studies on effectiveness of AMF on plant growth and physiology in Kinnow budded on *Jatti khatti* rootstock showed that mixed strain gave the maximum root colonization (over 45%). AMF treatments increased plant height, canopy volume, mean leaf area and number of new shoots per plant. Mycorhizal inoculation increased tissue P, K, Mg, Fe and Mn. *Glomus intradices* and *Glomus fasciculatum* were most effective AMF strains for Troyer citrange and *Jatti khatti* seedlings respectively and for budded plants of Kinnow on these rootstocks. There were marked differences in growth, physiological, biochemical and nutritional status of the mycorrhizal plants. *Glomus intraradices* and *Glomus fasciculatum* inoculated Kinnow plants budded on Troyer citrange and *Jatti khatti*, respectively showed the maximum level of phosphorus derived from the applied fertilizers (Nayak, 2007).

The effect arbuscular mycorrhiza fungi (AMF), on physiological and biochemical parameters of plants under well watered (WW) and water deficit stress (WDS) conditions in *Jatti khatti* (*Citrus jambhiri*) seedlings inoculated with two species of AMF, *viz.*, *Glomus fasciculatum* and *Glomus intraradices* revealed that *G. fasciculatum* showed maximum root colonization in *Jatti khatti* seedlings under both WW (49.38%) and WDS (49.10%) conditions thus, improving growth, physiological, biochemical parameters (Dutta Sudip Kumar, 2008).

In 5-year-old Kinnow mandarin plants fertigation with 100 per cent N, P, K (T6) had positive effects in improving photosynthetic rate, transpiration rate, stomata conductance and relative water content. Maximum number of fruits per tree (160) and yield (12.08 t/ha) was also recorded in this treatment. Fertigation had no significant effect on number of seeds per fruit. The maximum juice content (48.59%), TSS (9.46^{0}Brix), acidity (0.94%), ascorbic acid (45.03 mg/100 ml of juice) and total sugars (8.20%) contents were also found under in this treatment. Higher nitrogen (2.85%), phosphorus (0.17%), manganese (61.33 ppm) and copper (14.85 ppm) were recorded in this treatment, while higher potassium (1.84%) and zinc (64.83 ppm) was with 50 per cent N and 100 per cent P, K as soil application. Available nitrogen, phosphorus and potash distribution in soil had shown a decreasing trend with increasing soil depths (0-15 to 30-60 cm) regardless of the treatments. Fertigation resulted in increase in concentrations of both macro- and micro-nutrients near the active root zone (0-15 and 15-30 cm depths), exhibiting a radial decrease with increasing horizontal distance from the point of application, *i.e.* higher at 50 cm and lower at 100 cm distance from the trunk. (Uwisze Marie Grace, 2011).

Studies on co-inoculation of mycorrhiza with helper bacteria on physio-chemical changes in Amrapali mango on recommended rootstock Kurukkan was undertaken. Results revealed that inoculation of different AMF species with PSB and *Azotobacter* were found to decrease soil pH and EC with enhanced organic carbon and nutrient contents in the rhizospheric soil in Kurukkan rootstock seedlings and Amrapali/ Kurukkan composite plants. *Scutellospora hetrogramma* inoculated with PSB and *Azotobacter* was found most effective in enhancement of dehydrogenase (51.74 µg TPF/ g soil/day) and urease (29.32 µg/g soil/h) activities, whereas aryl sulphatase (91.96 µg/g soil) was higher for *Gigaspora gigantia* with helper bacteria. (Sant Ram, 2012).

Summary

On citation of above research review the following conclusions may be drawn:

- For determining vigour in mango at nursery stage, growth parameters *viz.* shoot length, internodal length, root length, root diameter and fresh and dry weight of shoot and dry weight root were found to be reliable criteria.

- Primer UBC-812. Primers UBC-812, UBC-891, UBC-808 and UBC-836 were found to be of high value for fingerprinting in mango.
- The effective period of mango hybridization using diverse parental mango genotypes was found to be from 3rd week of February to mid March under Delhi conditions
- Pollen–pistil interactions in four mango cultivars 'Amrapali', 'Mallika', 'Pusa Arunima' and 'Pusa Surya' showed that 'Mallika' and 'Amrapali' are self-incompatible, whereas, cultivars 'Pusa Arunima' and 'Pusa Surya' are self-compatible.
- In mango, 3 primer pairs (LMMA 11, ESTD 9 and ESTD 10) could confirm the hybridity in different cross combinations
- *In vitro* cultures from foliar and nucellar tissues in mango genotypes, Amrapali and Dashehari and rootstocks Olur and Kurukkan exhibited maximum culture establishment in nodal segment followed by leaf segment.
- Embryo rescue and DNA marker-assisted hybridity analysis in mango (*Mangifera indica* L.) cv. Amraplai revealed ESTSSR 9 and ESTSSR 10 to be informative and capable of ascertaining the hybridity of progeny population obtained from crosses Amrapali x Sensation and Amrapali x Tommy Atkins.
- For *controlling Fusarium mangiferae* in mango, *Bacillus subtilis* a gram positive bacteria was found effective in reducing the growth of *F. mangiferae*.
- Assessment of mango varieties for different defense responding biochemical compounds revealed significantly higher levels of PPO, LOX, peroxidase and catalase activity in mango varieties resistant to malformation when compared to susceptible varieties.
- A disease prediction model was developed for mango malformation using multiple linear regression, geo-statistical & GIS data.
- Studies on polyembryonic rootstocks cv. Kurukkan and monoembryonic (non-descript) mango seedlings revealed that the application of polyamines improved vegetative growth of mango plants.
- Paclobutrazol (PBZ) was found effective in the avoidance of salt stress in mango by increasing the levels of photosynthetic pigments, water content, K^+ uptake and restricted Na^+accumulation.
- Mango variety Amrapalli grafted on polyembryonic rootstock olour was found to be good Cl^- excluder, while non-descript seedling (common mango rootstock) could exclude Na^+ from leaf tissues of scion.
- Polyembryonic rootstock with different scion varieties *viz.,* Olour/Pusa Arunima, Kurakkan/Pusa Surya, Kurakkan/Amrapali, K-5/Amrapali, K-5/Mallika and Kurakkan/Dushehari combinations revealed their excellence with respect to horticultural traits (higher tree vigour, yield, pulp recovery, Total β carotenoids content and lower acidity).
- *C. sinensis* was found to be most suitable for the induction of seedlessness in *C. grandis* while *C. limetta* emerged as most appropriate pollen parent for haploid breeding
- Two solid mutants were developed in grape. Two primers RAPD-OPA1 and SSR-VVMD-14 were the most informative for identification of grape mutants.
- Genotype 'Tempranillo' and '*Vitis parviflora'* in grape showed significantly higher antioxidant activity in terms of DPPH, CUPRAC, FRAP and ABTS.
- Grape hybrid 'ER-R_2P_{36}' was found to be early maturing (last week of May) just 73 days after full bloom, with minimum growing degree days (1312.77), having maximum TSS content (23.55^{0}Brix), higher TSS:TA ratio (31.25), seedless berries, and self-thinning type loose bunch.
- Genetic status of the putative mutants in grape was ascertained using molecular analysis, *viz.*, RAPD and SSR markers. Primers OPA01 & OPP04 (RAPD) and VVMD-14 (SSR) were identified as unique primers.
- In grape 18 solid mutants having higher salinity tolerance were identified.

- Protocol was developed for micro propagation of different grape rootstocks, namely Dogridge A, SO4, ARI-H-144 and 3309C. ISSR was found more potent than RAPD markers for the assessment of clonal fidelity.
- The mechanism of NaCl tolerance in grape rootstock genotypes were found to be due to their ability to exclude Na^+ (99R, 110R, 1613C < Dog Ridge B, Salt Creek, H-144) and/or Cl^- ions (1616C < 1613C, SO4 < 110R < H-144).
- In grape, Defence related enzymes like peroxidase (PPO), polyphenol oxidase (POD), phenylalanine ammonia lyase (PAL) and malondialdehyde (MDA) content showed an increase in reaction to anthracnose.
- Gametophytic self-incompatibility (SI) was identified as a cause for seedlessness in Kagzi Kalan.
- Protocol was standardized for *in-vitro* propagation in citrus rootstocks, *viz.*, *Jattikhatti, Karnakhatta* and Troyer citrange. Culture establishment was best on MS medium + 3mg/l BAP + 0.5mg/l NAA.
- ISSR analysis could identify zygotic and nucellar seedlings in citrus species. A unique ISSR marker was found to identify embryo type seedling at nursery stage.
- In Kinnow mandarin and sweet orange, Radio tracer studies ($^{14}CO_2$) confirmed reduced translocation of metabolites to the granulated fruits compared to healthy fruits.
- Mutagenesis study in Kinnow lead to the conclusion that physical and chemical mutagens at higher doses have the potential to generate much needed variation in 'Kinnow' mandarin and thus can be used to evolve mutants for economic traits including seedlessness.
- In Kinnow, enhanced photosynthetic rate, proline content along with up regulated antioxidant, and PPO activities can be used as a criterion for selecting appropriate rootstock adapting different soil and climatic conditions.
- Studies on effectiveness of AMF on plant growth and physiology in Kinnow budded on *Jatti khatti* rootstock showed that mixed strain gave the maximum root colonization.
- *Phytophthora* inoculation through infected roots tended to show the highest rotting of feeder roots and found best method for the pathogenic studies in citrus rootstocks.
- Plant bio- regulators Paclobutrazol and putrescine was found effective in mitigating the negative effects of NaCl stress in citrus. With the use of these triazole compounds, more area can be brought under citrus cultivation in mild saline prone areas.
- Higher levels of proline accumulation and leaf abscission could be used as indicators for screening citrus rootstocks for resistance to NaCl stress.
- Polyamines such as putrescine (Put), spermidine (Spd) or spermine (Spm) was found effective in mitigating salt stress in salt susceptible citrus rootstock Troyer citrange.
- Triazole compounds, Paclobutrazol (PBZ) 100 mg l^{-1} and triazole (TRZ) 20 mg l^{-1} was found effective in protecting Kinnow plants budded on *Jatti Khatti* under NaCl stress.
- Kinnow mandarin plants fertigated with 100 per cent N, P, K was found effective in improving leaf gas exchange parameters and number of fruits per tree.
- Seed colour as morphological marker was adjudged most stable marker for identifying female and hermaphrodite plants in gynodioecious varieties of papaya. SCAR markers are most consistent.
- Genetic diversity analysis of indigenous and exotic apple genotypes using ISSR markers revealed 130 amplification products with 21 ISSR primers of which 130 (100%) were reported to be polymorphic.

Future Perspectives

The following priority areas have been identified for future postgraduate research in Fruits and Horticultural Technology discipline:

- Phenotyping of mango hybrids and germplasm for different parameters attributing yield and quality.
- Unraveling molecular basis of mango malformation using transcriptomic approaches.
- Identification of QTL(s) for fruit quality trait(s) in mango (*Mangifera indica* L.)
- Mutagenesis studies in mango and morphological and molecular characterization of mutant population.
- Comparative *in vitro* regeneration and genetic transformation studies on mango.
- Molecular characterization of full-sib mango hybrids using DNA markers.
- Induction of parthenogenesis in commercial cultivars of citrus.
- Mutagenesis studies in citrus and morphological and molecular characterization of mutant population.
- Induction of parthenocarpy though Plant Growth Regulators and pollinicide in Kinnow mandarin.
- Mapping of major QTL's in citrus populations for biotic and abiotic stresses.
- Rootstock induced changes in the tree physiology of mango and citrus.
- Seedlessness and *in ovulo* embryo rescue in grape.
- Candidate gene association studies in grape for bioactive compounds and flavonoid content.
- Mechanical behavior of seed hardness and fruit firmness in grapes grown under subtropical conditions.
- Screening and selection of wilt resistant in guava.
- Evaluation and characterization of guava genotypes for wilt tolerance, soft seed and pulp colour.
- Response of papaya genotypes against PRSV infection.
- Sex expression studies in papaya.
- Evaluation of papaya genotypes for cold stress tolerance.
- Shortening breeding cycle using *in-vitro* techniques and vivipary in papaya.
- Response of papaya genotypes against PRSV infection.

Acknowledgements

We wish to thank all the Faculty Members of the Division of Fruits and Horticultural Technology for extending support and cooperation in preparing the research contributions of M.Sc. and Ph.D. students.

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Vegetable Science

Tusar Kanti Behera and Brij Bihari Sharma

Introduction

The inherent richness of vegetable crops for nutrients makes them suitable food material to address the major concerns of hunger and malnutrition in the developing countries. The challenge before India is to meet the requirement of ever increasing population which is expected to surpass 1.4 billion by 2025. The diversified and highly nutritive vegetables are of great importance in alleviating hunger and malnutrition. Although vegetarianism is widely prevalent in India, yet for a population of more than 1.2 billion the consumption of vegetables per caput per day is around 200 g which is below the recommended requirement of 300 g per caput per day. It is, therefore, necessary to increase the present vegetable production of 180 million tonnes (2013-14) to more than 250 million tonnes to meet the dietary requirement of vegetables by 2025.

Improvement work on vegetables started at IARI, New Delhi with the establishment of a small section of vegetables in the erstwhile Division of Botany in the year 1940 which later became a part of the Division of Horticulture after its establishment in 1956 with the main objective to conduct research in all the major horticultural crops including vegetables and to impart post-graduate teaching. The research work was strengthened further with the creation of Division of Vegetable Crops and Floriculture in 1970. In 1982, Floriculture was separated and the Division of Vegetable Crops started functioning independently. The Division was renamed as Vegetable Science in 2004. The mandates of the Division are:

- To conduct applied and strategic research on improvement of vegetable crops
- To impart postgraduate education and training for human resource development
- To disseminate information on recent advances in vegetable crops
- To provide advisory and consultancy services on crop and seed production technology and improvement of vegetable crops.
- To collect, evaluate, purify and preserve the bio-diversity material related to different vegetable crops in collaboration with NBPGR and to utilize the desirable superior genotypes in the breeding programme.

Significant Student's Research Achievements

The research achievements of the Division have been contributed significantly by the students sincerely guided by the experienced faculty members and the Professors of this Division. The research contribution of the students is discussed in head-wise and presented below.

Diversity Analysis

High heritability estimates coupled with high genetic advance were observed in brinjal for fruit weight, fruit index and number of fruits per plant which indicated predominance of additive gene action. A highly significant positive correlation of fruit index, fruit weight and plant height was observed with yield per plant. Fruit diameter had high direct positive effect on yield per plant followed by days to first fruit set. The RAPD analysis generated 69 polymorphic markers by analysing 30 diverse lines with 5.81 bands per primer and the

genetic distance ranged from 0.07 to 0.78 (Rathi, 2008). All the genotypes were grouped into five clusters based on D^2 values, which exhibited no association between geographical and genetic divergence. The maximum distance at intercluster level was between clusters I and clusters IV followed by II and IV which may serve as a potential genotypes for hybridization programme (Rathi *et al*., 2011). In another experiment Premavati *et al*. (2015) found 24 breeding lines of *Solanum* spp., including the 20 eggplant cultivars and four wild species, were grouped into four clusters. The highest intra-cluster distance (5.80) was observed in Cluster IV with eight genotypes and the lowest intra-cluster distance (2.21) was observed in Cluster II also with eight genotypes. The first three principal components (PC1, PC2, and PC3) accounted 73.99% of the total variation among the 24 genotypes. These phenotypic data increase the feasibility of prioritising breeding lines in a crossing programme based on the uniqueness of their desirable morphological traits.

In bitter gourd, 50 indigenous and exotic genotypes were characterized using 12 quantitatively inherited traits and genotypic variation was analysed using RAPD and ISSR markers. Based on the morphological traits, the first female flower appeared at lowest node (9^{th} node) in gynoecious genotypes DBGy201 and DBGy202. Seventeen RAPD markers produced 84 amplicons in 50 accessions, of which 33 (41.34 %) were found polymorphic. Joint comparisons among the 50 accessions using Jaccard's similarity coefficient indicated that genetic distances (GD) ranged from 0.03 to 0.28. Eleven ISSR primers provided a total of 58 amplicons of which 41 (70.0 %) were polymorphic in 50 accessions and the Jaccard's similarity coefficient matrix values ranged from 0.50 to 0.95. The polymorphic information content (PIC), resolving power (RP) and marker index (MI) were 0.17, 1.14 and 0.82, respectively for RAPD markers, whereas ISSR markers showed comparatively high polymorphic information content (0.40), resolving power (1.87), and marker index (2.11). The combined data analysis of RAPD and ISSR markers indicated that the relative polymorphism among accessions was 52.6 % with 2.64 polymorphic amplicons per primer. These data demonstrate a large genetic variability among the Asian bitter gourd genotypes examined, which indicates that they should be considered as a valuable gene pool for bitter gourd breeding programs (Dalamu *et al.,* 2012).

Phylogenetic studies in 6 *Momordica* species (Fig. 1) were made by utilising 21 RAPD and 12 ISSR markers with 35 genotypes and five genotypes of two *Luffa* species. A total of 436 RAPD and 230 ISSR scorable fragments were produced of which 99.8% fragments showed polymorphism among the species and varieties of *Momordica* and *Luffa.* The level of polymorphism detected by the 33 random primers was higher among the species (99.8%) than that estimated among the varieties (61.3%). The varieties belonging to dioecious *Momordica* species (75.6%)

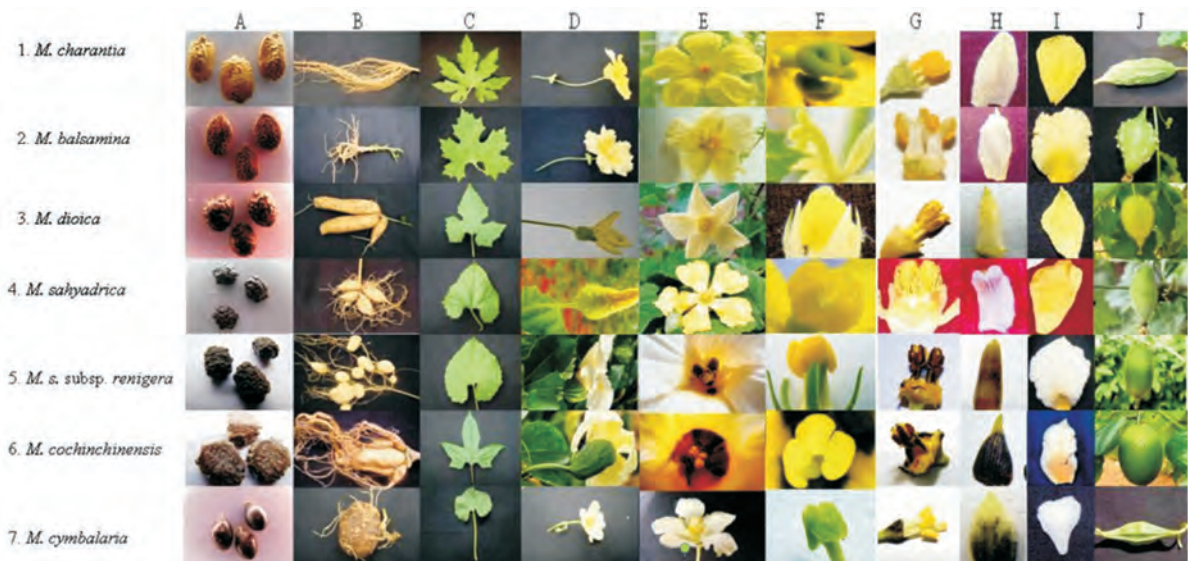

Fig. 1: Genetic diversity in *Momordica* species

showed a higher level of polymorphism as compared to monoecious species (50.3%). A significant level (68.6%) of polymorphism however was detected by the two marker types among the Indian varieties of monoecious *M. charantia* species. Wider divergence of the taxon of controversial identity, *M. cymbalaria* from the other Indian cultivated *Momordica* species and their evolutionary closeness with Luffa species was evident. The clustering pattern obtained among the 40 genotypes belonging to different *Momordica* and *Luffa* species corresponded well with their morphological, cytological and taxonomic classification, which was further supported by high boot-strap values and PCA analysis. Species and genotype-specific fragments detected by the random markers would be useful in introgression breeding for genetic improvement of *Momordica* cultivated in India. (Bharathi *et al.,* 2012).

Significant differences were observed among genotypes in early Indian cauliflower suggesting sufficient variability for yield and quality characters. The genotypes DC-98-4, DC-98-10 and DC-124 were found superior with respect to yield and quality characters. The overall values of PCV were higher than those of GCV. The highest estimate of GCV was observed for vitamin C contents (54.58) followed by duration of curd availability (49.04), while highest heritability was recorded for days to 50% curd formation (0.992). High heritability along with high genetic advance as per cent of mean was estimated for curd compactness, net curd weight and vitamin C content. Total yield had significant positive correlation with net curd weight and harvest index. However, yield was negatively correlated with duration of curd availability and days to 50% curd formation. Path coefficient analysis revealed that net curd weight and curd compactness had the highest positive contribution towards the total yield respectively (Kumar *et al.,* 2011).

The genetic diversity analysis was made using 36 genotypes of cowpea based on 13 yield contributing traits and 5 nutritional traits. The highest phenotypic coefficient of variation (PCV) and Genotypic Coefficient of Variation (GCV) were recorded for number of pods per cluster (50.95 and 45.33), followed by plant height (37.19 and 36.87) and pod yield/ plant (35.73 and 34.92). Broad-sense heritability (h^2b) values were observed with a range of 41.30% to 89.70%. Number of seeds per pod (41.30%) was the least heritable trait while number of pods/plant (89.70%) was the most heritable trait. The strong significant positive correlation of pod yield/ plant manifested with number of pods/plant, number of pods/cluster and pod weight was only due to its direct contribution towards pod yield/ha. Wide range of genetic diversity was observed among 36 genotypes of vegetable cowpea. Based on Mahalanobis' D^2 analysis all genotypes were classified into 8 clusters by considering yield contributing characters. The clustering pattern indicated that geographical distribution need not necessarily be related to the genetic diversity (Sandeep Singh, 2013).

In okra, 30 diverse genotypes were analysed for 19 quality traits. The maximum iron content was recorded in genotype DOV 66 (1.71ppm) and highest calcium content was recorded in Arka Abhay and DOV 23. Genotypes, P 7 (13.33µg/g) and DOV 2 (13.04 µg/g) were found high in ascorbic acid. Based on Mahalanobis D^2 analysis 30 genotypes were grouped into 7 divergent clusters. Cluster VII consisted of maximum (7) genotypes followed by cluster III and V (6 genotypes in each). Maximum inter cluster distance (6.44) was recorded between genotypes of cluster IV and VII. Cluster IV recorded maximum cluster mean for sugar (5.36%), ascorbic acid content (11.42 mg/g), ash content (1.57%), protein content (2.54%) and Mn content (0.29ppm). In principal component analysis of morphological traits, a total of 82.33% variability was contributed by 7 principal components out of 20 principal components (Rahul Kumar *et al.*, 2015).

In garden pea, genetic diversity analysis was made by using 28 genotypes. On the basis of non-hierarchical Euclidean cluster analysis twenty eight genotypes were grouped into 3 clusters. Cluster I had the largest number of 15 genotypes followed by Cluster III which had 8 genotypes and cluster II had 5 genotypes. The maximum inter-cluster distance of 5.613 was found between clusters I and II. These genotypes were also analysed on the basis of RAPD markers. Sixteen RAPD primers produced 79 polymorphic markers which showed 73.15% polymorphism.. The polymorphism information content (PIC) was highest for the RAPD primers OPN14 (0.74) and HU11 (0.64) which reflects that OPN 14 was found to be highly informative for diversity analysis (Arul *et al.*, 2012).

In musk melon, genetic diversity analysis was made using 67 genotypes from 3 horticultural groups on the basis of morphological traits (Fig 2) and molecular marker analysis. The range of variation was about 8-10 times for many fruit quality traits such as total carotenoids, P, K, Mg, Zn, Mg, Cu and Fe content. High phenotypic coefficient of variation (PCV) and genotypic coefficient of variation (GCV) were observed for average fruit weight (49.14, 44.03), yield per plant (43.45, 40.65), and fruit shape index (34.76, 34.38) among yield traits and iron content (56.72, 51.53), zinc content (54.42, 49.57) and total carotenoids (54.77, 51.46) among quality traits which reflected wide variability for traits of economic importance.

Fig. 2: Diversity in muskmelon

Moderate to high heritability coupled with high genetic advance as per cent of mean were found for average fruit weight (80.31, 81.29), yield per plant(87.64, 78.45), total carotenoids (88.29, 99.61), Zn content(82.98,93.02) and high heritability with moderate genetic advance were obtained for TSS(90.47,35.26) and vitamin C(93.16, 31.14). Positive genotypic correlation coefficient between yield per plant and average fruit weight (0.93), flesh thickness (0.60), fruit shape index (0.49), days to first pistillate flower opening (0.33) and total soluble solids (0.16) were found and selection for these traits will be helpful for yield and quality improvement. TSS (24.60), yield per plant (23.25), seed length (14.60) and rind thickness (10.49) were the most important yield traits contributing towards genetic diversity (Beesanakoppa, 2015).

The genetic diversity in 102 genotypes of *Citrullus* species was carried out on the basis of morphological traits and molecular marker analysis. The germplasm was evaluated for resistance against bud necrosis disease under field condition to identify sources of resistance and utilize them in development and characterization of inter-specific hybrids. Five genotypes DWM 152, Sugar Baby, DWM 19, and DWM 171 and DWM 113 can be used directly in the breeding programme for improvement of different yield traits. Principal components analysis suggested that average fruit weight, fruit length, fruit diameter, flesh thickness, fruit yield and number of fruits per plant were the major characters for discrimination of 102 *Citrullus* genotypes which could be classified into two major clusters. Fifty three *Citrullus* germplasm could be classified into two clusters at 48 % similarity based on SSR markers. All 12 wild *Citrullus* genotypes were grouped in cluster I and 41 genotypes from cultivated types were grouped in cluster II. Three *Citrullus* genotypes DWM 30, DWM 55 and DWM 210 (*C. colocynthis*) could be identified as highly resistant to bud necrosis disease under field condition. Inter specific hybrid between Sugar Baby and DWM 210 also exhibited highly resistant reaction against bud necrosis disease which indicates that DWM 210 (*C. colocynthis*) can be utilized as source of resistance for introgression of genes into cultivated varieties in future breeding programmes (Jamatia, 2016).

Genetics and inheritance of traits

In bitter gourd, inheritance pattern of three fruit traits was studied using two diverse parents, namely, Sel-2 (~25 cm long, non-prominent tubercles and curved) crossed with small fruited type Pusa Purvi (DBG-34) (~4 cm long, prominent tubercles and straight) in bitter gourd. F_1 generation obtained was intermediate (avg ~10 cm) for fruit length and in F_2 generation variation ranged from 2.5 to 25 cm. This continuous variation in fruit length of progeny by frequency distribution of nine classes with interval of 2.5 cm indicated its quantitative inheritance and more than 4 genes involved in controlling this trait. Distribution curve for fruit length in F_2 generation skewed towards Pusa Purvi, which suggested that short fruit length is partially dominant over long fruit length. The broad sense heritability for this trait was reported to be 97.40% that is heritable variation and selection for fruit length will be useful for its improvement (Meenu Kumari *et al.,* 2015). The inheritance of tubercles and curviness of fruits in bitter gourd, governed by a single pair of nuclear gene and prominent

tubercles was dominant over non tubercles. In cross Sel-2 × Pusa Purvi indicated that straight fruit is dominant over curved fruit and governed by a single pair of gene. Gene action for yield and its related traits in bitter gourd was studied using six basic generations (P_1, P_2, F_1, F_2, B_1 and B_2). The results revealed the presence of additive, dominance gene effects and epistatic interactions for all the characters studied in cross Sel-2 × Pusa Purvi indicating the importance of both additive and non-additive gene actions in the expression of the characters. Mean generation analysis for fruit length showed that dominance is the major contribution towards small fruits (Meenu Kumari *et al.,* 2015).

In another experiment, phenotypic ratio of 3:1 (monoecious vs gynoecious) in bitter gourd confirmed the monogenic recessive inheritance of gynoecism trait. Further investigation on gene action in the cross DBGy 201 × S-2 and DBGy 201 × Pusa Do Mausami by generation mean analysis revealed duplicate epistasis for most of the traits including node at which first female flower appear, days to first flowering, days to first harvestable maturity, fruit length, fruit diameter, fruit weight and yield suggesting the possibilities of obtaining transgressive segregants in later generations. Moreover, significant additive and non-additive gene effect for traits like node at which first female flower appear, days to first flowering, days to first harvestable maturity and fruit diameter indicates use of reciprocal recurrent selection or bi-parental mating for improvement of these traits. Non-additive gene action was significant for number of fruits per plant, fruit length, weight and yield inferring heterosis breeding as the best option for obtaining higher gain in these characters (Mishra *et al.*, 2015).

The inheritance pattern of gynoecious sex expression in cucumber was studied by utilizing a gynoecious line (GBS-1) and two monoecious lines (Pusa Uday and Punjab Naveen). Crosses were made between gynoecious line (GBS-1) and monoecious lines (Pusa Uday and Punjab Naveen). The F_1 and F_2 population along with parental lines were evaluated to study the inheritance of this trait. All F_1 hybrids showed gynoecious sex in both crosses and in the F_2 generation, the observed distribution of plant phenotypes fitted the expected mendelian ratio of 3 (gynocious plant) : 1 (monoecious plant). The segregation of plant sex types suggested monogenic dominant control of gynoecious sex form in cucumber using genotype GBS-1 (Pati *et al.*, 2015).

Heterosis breeding

In ash gourd, 10 elite inbred lines were selected based on their performance and were crossed in a diallel mating system (without reciprocal crosses) to generate 45 F_1 hybrids. These hybrids along with the parents were grown to study heterosis in 10 yield related characters. The mid and better parent heterosis was observed to be as high as ~165% for yield per vine in DAG-6 × DAG-11. The maximum negative heterosis over the best parent for days to fruit maturity was noticed in cross DAG-2 × DAG-9 (~ -10%) indicating that it can be successfully utilized in breeding for earliness in ash gourd. Two hybrids namely, DAG-1 × DAG-5 (34.33 kg) and DAG-4 × DAG-11 (31.67 kg) recording positive heterosis over the best parent to the extent of 23.5% and 14.0% respectively for yield per vine can be utilized for commercial cultivation (Verma and Behera, 2007).

In Bitter gourd, in the study of exploiting gynoecy for hybrid development, gynoecious parent, DBGy-201 showed maximum GCA effect in desirable direction for node to first female flower, days to first picking, numbers of fruits and yield per plant. The parent, P6 (Priya) exhibited highest GCA effect for fruit length, weight and diameter. These parents were good general combiners for yield and earliness and could be used to improve these traits in bitter gourd breeding programmes for the accumulation of favourable genes (Dey *et al.*, 2010).

In cucumber, 15 fifteen F_1 hybrids developed by using diallel (without reciprocal) matting system involving six parents showed over-dominance and low narrow-sense heritability for earliness, number of fruits per plant and total yield per plant. Components of additive gene actions were significant for fruit weight, fruit length and fruit diameter. The breeding methods like pure line selection for the above characters which have additive gene effects at significant levels and heterosis breeding which having predominant role of non-additive gene action, may be exploited for improvement of these traits (Tiwari *et al.*, 2007). Another study was conducted on a 8 × 8 diallel set of cucumber excluding reciprocals to find out the extent of combining ability for yield and yield

attributing characters. The magnitudes of variance due to general as well as specific combining ability were highly significant indicating the importance of both additive and non-additive gene action. Among parents, gynoecious line GBS-1 showed maximum g.c.a effects in desirable direction for node number of first female flower, days to first female flower anthesis, days to fruit set from opening of first female flower, days to first fruit harvest, number of fruits per plant, and vine length. The parent Pusa Uday exhibited highest positive g.c.a effect for fruit length, fruit diameter, average fruit weight and yield per plant. In order of merit the gynoecious hybrids GBS-1 × Pusa Uday and GBS-1 × Punjab Naveen followed by monoecious hybrid GS-4 × Pusa Uday, which exhibited highest s.c.a effects for number of characters including total fruit yield per plant. In case of yield and other yield contributing characters like node number of first female flower, days to first female flower anthesis, days to fruit set from opening of first female flower, days to first fruit harvest, number of fruits per plant and yield per plant where specific combining ability component of variance is more than general combining ability component of variance, which indicated preponderance of non-additive gene action. Hence, an improvement programme based on heterosis breeding would be appropriate for improving different traits under study (Pati *et al.*, 2015).

In cucumber, the hybrids Pusa Uday × DC-1 followed by Pusa Uday × Kalyanpur Green and Pusa Uday × Punjab Naveen showed maximum heterosis percentage of 86.65, 62.90 and 56.11, respectively over standard parent for total yield per plant. Hence, this hybrid can be exploited commercially for earliness and yield per plant. Parent DC-1 and Pusa Uday was best performing parents for earliness and yield per plant on the basis of gca effects. The F_1 hybrid showed promising results when atleast one of the parental lines exhibiting high gca effect for yield and its component traits were involved in the crosses. Highest sca effect for total yield per plant was shown by hybrid Pusa Uday × DC-1 followed by Pusa Uday × Kalyanpur Green and Punjab Naveen × DC-1. These combinations may be exploited for commercial hybrid production. Generation mean analysis showed the significance of additive, dominance and epistatic interaction in most of the crosses for nine different quantitative characters. Simple mass selection would result in progress for characters governed by additive gene effects, heterosis breeding for characters governed by dominance gene effects and breeding plan based on restricted selection by way of intermating the most desirable segregants followed by selection and/or diallel selective mating system would be most appropriate for characters where duplicate epistasis was predominant. Complementary epistasis was observed for yield per plant, which would increase the heterosis in positive direction. Result pertaining to the inheritance of downy mildew revealed that resistant was controlled by single recessive gene and the resistance can be transferred to horticulturally desirable, susceptible varieties through backcross breeding (Bhutia *et al.*, 2015).

Seven parental lines including two gynoecious lines were used to develop 21 F1 hybrids of cucumber by half diallel mating to determine the magnitude of heterosis for earliness yield and yield related traits during spring summer and rainy season. The mean sum of squares were highly significant for all the characters indicated a wide genetic variation for the characters studied and there is a possibility of genetic improvement using such genetic pools in future breeding programme. The gynoecious parental lines (PPC-2 and GPC-1) and monoecious parent Pusa Uday were observed to be the three top performing parents for yield per plant. Appreciable heterosis was observed over better parent and top parent for most of the characters studied. The F1 hybrids those found to be superior in performance over top parents for various characters were GPC1 × PPC-2 for node number at first female flower, number of fruits per plant, PPC-2 × Pusa Uday for days to fruit set, days to first fruit harvest, yield per plant, Punjab Naveen × Pusa Uday for fruit length, fruit diameter, DC-1 × Pusa Uday for average fruit weight. The best three heterotic hybrids PPC-2 × Pusa Uday, GPC-1 × Pusa Uday and PPC-2 × Punjab Naveen showing 64.51, 55.61, and 54.57% heterosis, respectively, over standard check Pusa Uday for yield per plant, may be exploited for commercial cultivation (Jat *et al.*, 2015).

In sponge gourd, combining ability studies of seven parental lines in 2 Line × 5 Tester design for earliness and yield revealed the predominant role of non-additive gene effect in controlling earliness and increasing yield. Among parents, Pusa Sneha was found the best general combiner for imparting earliness; PSG-9 for

ratio of male and female flowers and NSG-1-11 for total yield per plant. Significant sca effect for earliness was lacking, but five hybrids exhibited desirable sca effects for earliness. DSG-6 × Pusa Sneha was found superior for days to first fruit picking and DSG-7 × NSG-1-11 for number of male and female flowers and total yield per plant. The parent NSG-1-11 and Pusa Sneha were most promising for yield improvement and imparting earliness, therefore, they may be recommended as useful parent in breeding superior varieties. DSG-6 × Pusa Sneha exhibited maximum standard heterosis (%) for earliness and DSG-7 × NSG-1-11 for ratio of male and female flowers and total yield per plant. Therefore, they may be recommended for commercial cultivation in North Indian plains (Islam *et al.,* 2012).In another experiment, 21F_1 hybrids developed by diallel matting design (without reciprocal) along with seven parental lines including two hermaphrodite inbreds were evaluated. The parents DRG-2, Satputia Long and Satputia Small were good combiners for a number of characters, including yield/plant. Ten crosses having hermaphrodite inbred as one of the parents, excelled for high per se performance along with desirable specific combining ability effects. The present findings reveal the breeding value and potentiality of hermaphrodite *Satputia* lines in the genetic improvement for ideal plant type in ridge gourd (Karmakar *et al*., 2014).

Fig. 3: CMS plants developed in Cauliflower background through interspecific hybridization

In cauliflower, cytoplasmic male sterility is being looked upon as potential alternative in the F_1 hybrid seed production, as weak alleles pose a threat of self-seed in SI based hybrid seed production. Morphologically, F_1s were of intermediate type in leaf characteristics, whereas for flower characteristics and bolting behaviour these were like that of sterile source. However, F_1 that originated from the *B. tournefortii* cytoplasm and *B. oleracea* var. *botrytis* had fertile and sterile plants where from sterile plants were selected for backcross (Fig 3). The backcross progeny still segregated for fertility and sterility as confirmed by pollen viability test. From the experimental study and segregation behaviour of sterile and fertile plant, it was revealed that there was fertility restorer gene for the *tournefortii* cytoplasm. The fertility restorer gene may be either from cauliflower or from parental source or due to improper function of sterile cytoplasm with nuclear genes of cauliflower. To achieve complete male sterility, therefore, in this background further backcrossing of the selected male sterile plants to eliminate the restorer gene is essential. The F_1 and BC_1 generations developed using *canariense* sterile cytoplasm and cauliflower as pollen parent were completely sterile with flowers having rudimentary stamen and shriveled anthers and pollen viability test also confirmed sterility (Verma *et al*., 2010).

Eighty crosses derived from line (10) × tester (8) mating design along with their parents were evaluated to study the combing ability and its relationship to gene action and heterosis for eight yield and related traits in early maturity cauliflower. Analysis of genetic component of variance and variance due to specific combining ability (sca) revealed preponderance of dominant variance and non-additive gene action for all of the traits except for days to 50 % curd maturity. In hybrids, contribution of lines was higher over the testers for all the traits. Among the lines cc- 32E, 395aa and 14-4-17 and testers Sl-71, 23000 and Pusa Deepali were identified as promising general combiner for gross plant weight and marketable and net curd weight. However, hybrid cc-32E × Pusa Meghna was earliest (52 days) for curd maturity. For leaf area, plant height, curd compactness and gross plant weight the best combination was cc-32E × 23000. Among the hybrids, identified superior crosses with significantly highest level of heterosis over better parents were 395aa×Sel-7 (68.0 %), cc- 32E × 23000 (48.19 %) and 395aa × Pusa Deepali (34.76 %) for economic trait marketable curd weight. Hence these hybrids can be further tested under different agro-climate for commercial production (Verma and Kalia, 2015).

The general and specific combining ability was studied in early and mid-maturity CMS based lines through line x tester analysis involving two CMS lines in each maturity group along with 6 and 9 testers for early and

mid-maturity group, respectively. The combining ability analysis revealed highly significant differences among genotypes for all the characters studied. The tester 23000 in early and line 8410-22 and tester 754 of mid group showed significantly negative GCA effect for earliness to curd initiation, whereas lines 8498-2 and 8410-22 were best combiners for marketable curd weight in early and mid-group, respectively. Cross combinations 8498-2 × Sl-71 and 8409-309 × SL-1-2 were identified as best specific combinations for earliness amongst early and midseason CMS based lines (Verma and Kalia, 2011). The tomato, hybrids Pusa Sheetal × Pusa Sadabahar had the best performance for early and total yield per plant and it showed significant heterosis over better parent for number of fruits and yield per plant. The SCA effects for most characters especially, days to first fruit set, days to first harvest and total yield per plant was observed highest in Pusa Sheetal × Pusa Sadabahar. Dominance effects as well as non-allelic interaction, dominance × dominance effects were predominant for most of the characters which indicated the importance of heterosis for improvement of various characters (Negi, 2007).

Among the 10 black carrot (IPC 126) combinations with red ones, the range of anthocyanins varied from 30.0 to 61.9 mg/100 g. The highest heterosis for root yield in this set was in IPC 126 × IPC 55 (50.8%) and the highest mean values for total carotenoids, lycopene, β carotene, lutein, iron and anthocyanins were found in IPC 126 × IPC 123 (3.7 mg/100 g), IPC 126 × IPC 116 (1.7 mg/100 g), IPC 126 × IPC 55 (1.8 mg/100 g), IPC 126 × IPC 16 (1.00 mg/100 g), IPC 126 × IPC 122 (16.23 ppm) and IPC 126 × IPC 55 (61.87 mg/ 100 g), respectively. The roots and cross sections of promising hybrids as well as cross sections of 15 parents and their F_1 hybrids are depicted in Figure 4.

Fig. 4: Promising carrot hybrid combinations' roots and cross sections

***In silico* characterization ACS genes for sex expression in cucurbits:** Bitter gourd is a monoecious plant, which is mainly cultivated in tropical and subtropical Asia. Phytohormones have implications on various aspects of reproductive organ development in unisexual as well as bisexual plants. In dicots, higher levels of ethylene and auxins are positively correlated with female sex expression, whereas gibberellin favours differentiation of male sex organs. Therefore, in the present investigation, we characterized monoecious and gynoecious line at early stage by estimation of ethylene and GA_3. However, expression of McACS2 gene which is solely responsible for ethylene action was studied at four different stages of conversion of female to hermaphrodite buds by spray of silver thiosulfate @6mM (Fig. 5).

Fig. 5: Staminal tissue induction by 6 mM Silver thio-sulfate (Scale = 500 µm)

This was further validated by quantification of ethylene, GA_3 and enzymatic activity of Guaiacol peroxidase and antioxidants capacity at all four stages. Reduction in expression of Mc-ACS2 gene was observed after three days of 1st spray and increased during the formation of hermaphrodite buds but its expression was less in modified (hermaphrodite) buds than the female buds. Ethylene was also found to be less in hermaphrodite buds than female buds whereas, GA_3 was found to be more in silver thiosulfate treated buds than female buds. Activity of both enzymes was higher in treated buds with respect to untreated ones. Hence, silver ion action hypothesis can be proposed as: it reduces ethylene perception resulting in less accumulation of McACS2 gene mRNA and endogenous GA_3 cooperation leads to formation of hermaphrodite flowers (Fig. 6) (Meenu

kumari, 2015). The genomic sequences of five ACS genes designated as McACS1, McACS2, McACS3, McACS4 and McACS5 (1942, 2333, 1941, 1481 and 1463 bp, respectively). Coding sequences of McACS1, McACS3, McACS4 and McACS5 consisted only one exon and length of deduced open reading frame (ORF) are 1455, 1455, 1302 and 1446 bp, respectively. In contrast, coding sequences of McACS2 was interrupted by four introns at conserved positions of 132, 147, 159 and 999 bp. In addition, McACS protein sequences had variations in isoelectric point (pI) values (5.80 to 7.12) and molecular weight (ranging from 53.90 to 55.46 kDa). Less variation reported for instability index (43.42 to 47.21) and aliphatic index (79.76 to 82.19) of these ACS proteins. Grand average of hydropathicity (GRAVY) was ranged from -0.202 to -0.307. The deduced amino acid sequences encoded by all five isomers of ACS contain the characteristics seven conserved regions of ACSs as well as the 12 amino acids of the active site of ACS enzymes as depicted. The key residues of conserved protein sequences often describe the biological role of protein. Five most conserved residues in functional domain are Phenylalanine, Proline, Aspartic acid, Lysine and Arginine at 82, 203, 230, 273 and 407 positions, respectively (Meenu kumari, 2015).

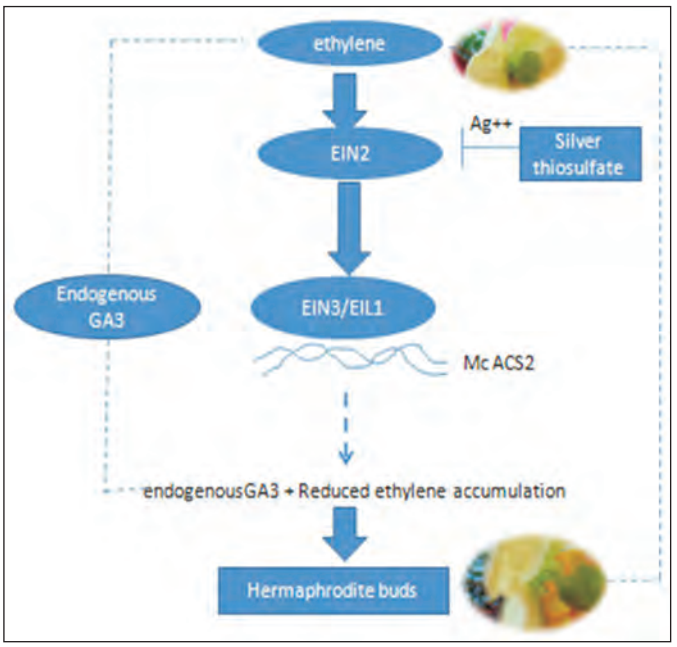

Fig. 6: Proposed model for action of silver thiosulphate in bitter gourd sex expression

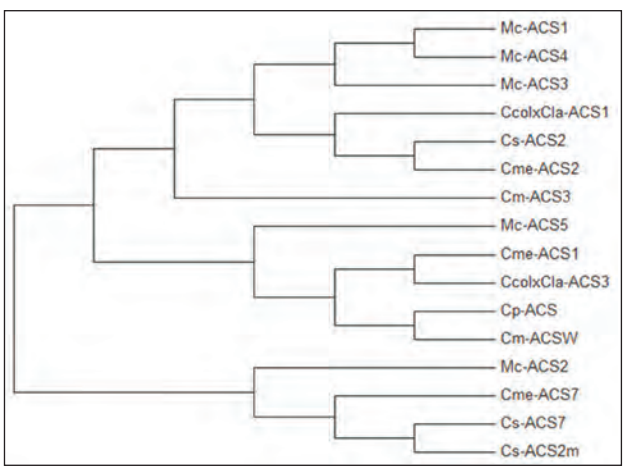

Fig. 7: Phylogenetic tree analysis from deduced amino acids of *McACS* and other cucurbits polypeptides.

The polypeptides encoded by McACSs shows very high identity i.e. upto 99% to one another. McACS1 and McACS4 were most closely related to each other (99% identity) which is followed by McACS2 and McACS5. McACS3 is more closely related to McACS1 and McACS4 than other isomers. However, phylogeny analysis of ACS polypeptides of cucurbits indicated that McACS1, 4 and 3 is most closely related to *Citrullus* spp. Ccol × cla-ACS1 with 93% identity and McACS2 is most closely related to Cme-ACS7 with 72% identity. McACS5 is 84% identical to Cme-ACS1 (Fig. 7).

The homology modelling approach was employed to determine 3D structure of McACS genes and these structures were further used as input to predict protein-ligand interaction active site for Ag+ binding. The created model (Fig. 8a) has shown lower value of DOPE (Discrete Optimized Protein Energy) score (-51,898.2) and PDF (Probability Density Function) energy (18,181.7) with respect to other models. X-ray crystallographic analysis located the hydrophobic pocket where the G31 likely to interact directly with substrate (Ag+ from STS, Fig. 8b) and affect or alter the function of protein which stimulates formation of anthers in pistillate flowers. The overall stereochemical quality of 3D structure of protein was evaluated by using Ramachandran plot which showed maximum residues in the most favoured region followed by allowed region and least in the generously allowed regions (Fig 8c) (Meenu kumari, 2015).

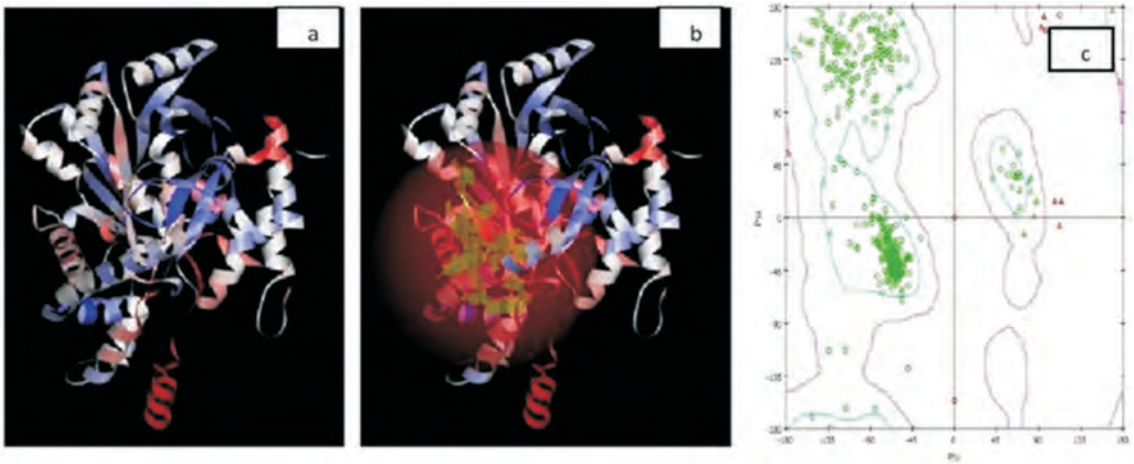

Fig. 8: 3D structure of *McACS2* protein a) Homolgy model predicted by alignment with template (1B8G, PDB database), b) Protein-ligand interaction site and hydrophobic pocket for binding of Ag^{+} ions and c) Ramachandran plot showing the authenticity of predicted model.

Breeding for biotic stress resistance

The significant increase in production as well as productivity of vegetable crops has been possible due utilization of high yielding genetic stocks for development of varieties/hybrids. However, emergence of a large number of biotic stresses has posed a challenge for sustaining productivity in vegetable crops. Major genes for resistance for biotic/abiotic stresses available in cross-compatible wild relatives of vegetable crops have been utilised through inter-specific hybridisation. However, still there are a number of challenge from biotic stresses particularly diseases by plant viruses. These include leaf curl viruses in tomato, chillies, and cucurbitaceous vegetable crops. The sources for resistance to these viral diseases are available in wild relatives which are cross-incompatible to cultivated species, posing challenge in gene transfer.

The genetics of resistance against early blight caused by *Alternaria solani* in tomato (*Lycopersicon esculentum* L. cv. 'Megha'), and to develop molecular markers as an effective strategy to assist in the transfer of resistance. The F_2 segregation pattern of a resistant susceptible ('Megha' 'Pusa Gaurav') cross revealed the trait to be governed by two recessive genes (Rao *et al.*, 2008). Efforts were made to develop RAPD markers to these two recessive resistance genes. Out of 14 random primers that were polymorphic between the parents, two primers (OPG19 and OPE11) produced unique amplicons 1,350 bp and 1,300 bp in length, respectively, that co-segregated in repulsion phase (i.e., were linked to the susceptible disease reaction in F_2 bulks). Single marker analysis also revealed significant differences between the marker genotypes of 75 F_2 individuals, indicating a linkage between the disease reaction and the individual markers $OPG19_{1350}$ and $OPE11_{1300}$. A best-fit ratio of 9:3:3:1 between the two markers, $OPG19_{1350}$ and $OPE11_{1300}$, revealed that they were not linked to each other, suggesting that the markers were linked to two different loci governing the resistance trait (Rao, 2007).

In tomato, the inheritance of resistance to root knot nematode (RKN; *Meloidogyne incognita* race 1) in progeny of cv. Pusa 120 was studied. Twenty-eight diverse genotypes of tomato were screened for resistance against *M. incognita* (race 1). Two susceptible tomato genotypes ('La-Bonita' and 'Selection N-5') used as pollen donors, were crossed with the more resistant 'Pusa 120' as the seed parent to obtain F_1 (Fig 9) and F_2 generations, and two backcross generations. After 60 d of exposure, the numbers of root knots and egg masses per plant root system were counted. A chi-square (χ^2) test of the frequency distribution based on the root-knot index (RKI) of the F_2 progenies of the two crosses (resistant × susceptible) showed the best-fit to a 3(R):1(S), Mendelian ratio. The monogenic dominant model was further confirmed by the 1(R):1(S) ratio found to be the best-fit for the backcross generation (B_2) with the susceptible pollen donor parent. These results confirmed that resistance to

RKN (*M. incognita* race 1) is controlled by a single dominant resistance gene (R-gene) inherited from the resistant parent, 'Pusa 120'. 'Pusa 120' can therefore be used to develop high yielding varieties, or hybrids, of tomato with improved RKN resistance (Shreshtha, 2012).

Fig. 9: Selection N-5 ×Pusa120

Thirty genotypes were screened under natural epiphytotic and artificial inoculation conditions in the rainy season, when the proliferation of whiteflies results in a high incidence of ToLCNDV disease symptoms. Vulnerability index values ranged from 3.33 – 100.0. Disease symptoms started to appear as minute yellow spots on the young leaves of susceptible lines 7 days after inoculation. Four genotypes (DSG-6, DSG-7, DSG-9, and DSG-10) were resistant. Eight genotypes (DSG-12, DSG-23, DSG-25, DSG-43, DSG-48, DSG-52, VRG- 19/33, and U53/V12-157) were moderately resistant. Five genotypes (JSSR-113, JSSR-20, NSG-28, 'Pusa Supriya', and KSG-14) were moderately susceptible, and the remaining 13 genotypes (IN-108, 'Pusa Chikni', JS/RC- 111, ZH/AC-94, SKY 1/26, CHSG-1, CHSG-2, SVS/KPS- 617, 'Pusa Sneha', KG-3/42, KG-3/134, NSG-1-11, and PSG-9) were susceptible (Islam *et al.,* 2011).

Two susceptible genotypes ('Pusa Sneha' and NSG-1-11), both possessing desirable fruit characters, were crossed with the two most promising resistant lines (DSG-6 and DSG-7) and the disease reaction of segregating and backcross generations studied through challenge inoculation with a purified strain of virus under insect-proof greenhouse conditions. A chi-square (v2) test of frequency distribution based on the vulnerability index of the F_2 progenies of the two resistant 9 susceptible crosses revealed monogenic dominant Mendelian ratio 3(R):1(S) to be the best fit in all crosses. This monogenic dominant model was further confirmed by the 1(R):1(S) ratio found to be best fit for the test cross with the susceptible parent. These results reveal that resistance to Tomato leaf curl New Delhi virus associated with yellow mosaic disease of sponge gourd is controlled by a single dominant gene in the genetic background of the resistant parents (DSG-6 and DSG-7) and that these two lines can be effectively utilized for the development of high yielding and yellow mosaic disease-resistant varieties/ hybrids of sponge gourd (Islam *et al.*, 2010)

The screening of cauliflower genotypes for resistance to downy mildew [*Hyaloperonospora parasitica* Constant (Pers.:Fr) Fr.] was performed under pot experiments using artificial inoculation were conducted under a climate favourable to the disease, and the percentage disease index (DI) was measured 10, 20, and 30 d after inoculation (DAI). Of the 190 genotypes screened, seven were highly resistant ($1 < DI \leq 10$), 21 were moderately resistant ($10 < DI \leq 25$), 51 were moderately susceptible ($25 < DI \leq 50$), 95 were susceptible ($50 < DI \leq 75$), and 16 were highly susceptible ($75 < DI \leq 100$). The seven most resistant genotypes were BR-2, CCm, 3-5-1-1, CCm-6, CCm- 5, MGS-2-3, and cc-12. The overall mean DI value increased from 45.2% at 10 DAI to 58.5% at 30 DAI. Among the genotypes screened, the incidence of downy mildew ranged from 4.5 – 84.6% at 10 DAI, 5.5 – 92.0% at 20 DAI, and 6.0 – 98.8% at 30 DAI. Different stages of plant development showed a strong positive correlation for downy mildew reaction in the genotypes tested. Genotypes identified as resistant can be used as donors to develop multiply-resistant varieties of cauliflower (Singh *et al.,* 2013)

The genetics of yield traits and downy mildew resistance in cucumber (*Cucumis sativus* L.) was carried out to identify the source of resistance to downy mildew in the available genotypes of cucumber. A total of 114 genotypes were screened for downy mildew resistance under both field and artificial condition. The eight inbred lines of cucumber were crossed in a 8 × 8 half diallel fashion without reciprocals to develop 28 F_1 hybrids. These eight parents and 28 F_1 hybrids were evaluated for heterosis, combining ability and gene action. The observation were recorded for nine quantitative characters viz. days to first female flower opening, days to

first fruit set, days to first fruit harvest, fruit length, fruit diameter, average fruit weight, number of fruits per plant, total yield per plant and vine length. The results of screening of genotypes for downy mildew resistance revealed that DC-70 (15 and 16) followed by DC-77 (18 and 19.5) showed lowest PDI and were found to be resistance under both the field and artificial condition, respectively whereas Swarna Ageti had highest PDI and was most susceptible in both the conditions (Bhutia., 2015).

Fig. 10: Artificial inoculation of *Xcc*-race 1 culture of *Xanthomonas campestris* pv. campestris

Five genetically diverse Ogura CMS lines of cauliflower *viz,* Ogu 13-01, Ogu 101, Ogu 103, Ogu 119, Ogu 13-85 and seven testers *viz,* Kt-18, Kt-22, DB-1305, DB-187, Lalchowk Maghi, Sel-26 and Suprimax Late were crossed in line x tester mating scheme to obtain 35 F_1 hybrids combinations for studying combing ability and heterosis for fourteen quantitative characters, ascorbic acid and mineral nutrients (Na, K, S, Ca, Mg, Fe, Zn and Mn). The hybrids Ogu 119 × Suprimax Late, Ogu 101 × DB-1305, Ogu 119 × Sel-26, Ogu 101 × DB-187, Ogu 13-01 × Kt-18 and Ogu 119 × Lalchowk Maghi were best heterotic combinations for yield, major yield attributing traits and also had high positive heterosis for ascorbic acid and mineral content**.** The hybrids Ogu 101 × Lalchowk Maghi, Ogu 13-85 × Kt-18, Ogu 13-85 × DB-187, Ogu 119 × Kt-22 and Ogu 119 × Suprimax Late were found to be the most promising for earliness and also exhibited high heterosis for yield and yield attributing traits and various mineral elements. The hybrid Ogu 101 × Lalchowk Maghi was found to be best heterotic combination for K, S and Zn content. The cross combinations Ogu 101 × Sel-26, Ogu 103 × Sel-26, Ogu 119 × Suprimax Late, Ogu 13-85 × DB-187, Ogu 13-01 × DB-187 and Ogu 101 × Kt-22 were the best heterotic hybrids for accumulation of ascorbic acid, Na, Ca, Mg, Fe and Mn. The above promising heterotic hybrids also have excellent external quality traits (Ram *et al.*, 2015).

There is dearth of black rot resistance in *B. oleracea* (C genome), therefore exploration of *Brassica species* (A and B genome) was inevitable as they are potential reservoirs of black rot resistance. Two varieties 'Pusa Vijay' and 'Pusa Bold' of *B. juncea* (AB genome); 'NPC-9' of *B. carinata* ((BC genome, Ethiopian mustard); five genotype/accessions of *B. nigra* (B genome), namely EC289661, 'Sangam', IC247, IC560690, IC56072 and one accession of *B. oxyrrhina* were found resistant after intensive artificial screening in field and laboratory conditions against *Xcc* race 1. The segregating population (F_2) was developed in *B. carinata* by selfing of F_1 plants of cross 'NPC-17' (susceptible) × NPC-9 (resistant) and simultaneously generated backcross generations B_1 and B_2 with 'NPC-17' (P_1) and 'NPC-9' (P_2), respectively. Black rot resistance segregated in 3:1 (resistant: susceptible) ratio in F_2 population, suggesting single dominant gene governing black rot resistance in 'NPC-9'. Segregation analysis in B_1 and B_2 population showed 1:1 and 1:0 ratio, respectively which confirmed its monogenic dominant nature (Sharma, 2016).

In Garden pea, thirty four genotypes screened in two consecutive years in *Fusarium* wilt sick plot revealed significant differences between the genotypes for percent wilt intensity and recorded 6-95%. The genotypes GP-6, GP-55 and GP-942 were found to be highly resistant and GP-17, GP-48, GP-473, GP-941 were resistant as new source for *Fusarium* wilt. It was interesting to record that all the popular cultivated varieties (Arkel, Pusa Pragati, AP-3, VRP-6, VL-7, VL-10 and Arka Ajit) were highly susceptible to wilt. The inheritance of *Fusarium* resistance gene (s) was also studied using F_1, F_2, BC_1 and BC_2 progenies of four crosses *viz.,* Pusa Pragati × GP-55, Pusa Pragati × GP-6 and Arkel × GP-17. Chi-square analysis was performed to determine the Mendelian segregation ratios of resistance and susceptibility among the inoculated progenies which showed F_1 and backcross progenies to the resistant parents segregated in the ratio of 1:0, while the F_2 progenies segregated in the ratio

of 3:1 and backcross progenies to the susceptible parent segregated in the ratio of 1:1. These observed ratios demonstrated a good fit to one gene model and confirmed that the resistance to *Fusarium* wilt in GP-55, GP-6 and GP-17 genotype is governed by monogenic dominant gene. (Shubha, *et al.,* 2016). To identify the molecular marker linked to Fusarium wilt, polymorphic SCAR primer (Y15_999*F*w) and ISSR marker UBC-812 was used for genotyping the F_2 population of cross Arkel (susceptible) and GP-6 (resistant). Observed χ^2 segregation ratio of UBC-812 and Y15_999*F*w fitted well with the expected ratio as it did not differ significantly from a 3:1 confirming its linkage with *Fw* locus. On the basis of phenotypic and molecular marker segregation ratio, it was concluded that *Fw* gene inherited monogenically with dominant nature in genotype GP-6. Further, linkage analysis study revealed that genetic distance of Y 15 SCAR marker and UBC-812 from *Fw* locus were 25.768 cM and 5.011 cM, respectively. Thus, this ISSR marker UBC-812 was identified as new marker found very close to the target resistant locus (*Fw*) and this marker could be used for marker assisted selection to develop *Fusarium* wilt resistant genotypes. (Shubha, 2016)

Breeding for abiotic stress tolerance

Inheritance pattern of salinity tolerance of cucumber was studied by combining ability and gene action analysis of salt-tolerant and salt-sensitive cucumber inbreds (produced through five generation of selfing) and its hybrids at 2 salinity levels, i e control (0.43 dS/m) and 4 dS/m. The higher values of general combining ability than that of specific combining ability, predictability ratio more than 0.5, average degree of dominance in between 0 to 1 and narrow sense heritability in between 50 to 100% indicated the predominance of additive gene action for salt tolerance in cucumber (Tiwari *et al.,* 2011). Increased levels of salt concentration resulted in progressive reduction in germination (78.47, 43.67 and 24.90%), number of leaves (19.26, 10.56 and 6.30), survivability (93.77, 59.79 and 39.25%), vine length (88.99, 49.07 and 28.92 cm) and fruit yield per vine (1.10, 0.62 and 0.29 kg); while increased the affected leaves (28.73, 68.92 and 82.59%) and defoliation (13.39, 59.74 and 74.83%), respectively in control, 2 and 4 dSm-1. Thus, these above phenotypic traits appeared to be promising as selection criteria for salt tolerance at morphological level. As a result, genotypes CRC-8 and CHC-2 observed salt tolerant, whereas DC-1, CH-20 and Pusa Uday were salt sensitive (Tiwari et al., 2013). The genotypes CRC-8, CHC-2 and G-338 showed lower accumulation of sodium, lesser depletion of potassium, lower Na^+–K^+ ratio and higher accumulation of proline, reducing sugars, phenols, better membrane stability and lower yield reduction (%) under salt stress, while CH-20 and DC-1 were sensitive to salt stress. Thus, a combination of traits such as higher membrane stability, lower Na^+– K^+ ratio, higher osmotic concentration and selective uptake of useful ions and prevention of over accumulation of toxic ions contribute to salt stress tolerance in cucumber. These traits would be useful selection criteria during salt stress breeding in cucumber (Tiwari *et al.*, 2010).

In cucumber, drought stress adversely affected the desirable physiological and biochemical parameters, and the adverse effect increased as the soil moisture deficit increased and the yield reduction was observed as high as 51.97% (under 25% of recommended irrigation (Fig 11). The physiological parameters such as proline, reducing sugars, phenol content on dry weight basis were increased significantly (p=0.05) as the drought stress increased from 100% irrigation to 25 % irrigation level. In contrast, the relative water content (RWC), chlorophyll stability, membrane stability index and fruit yield decreased significantly (p=0.05) with the increase in drought stress in all genotypes. Among the genotypes DGC-1, DGC-19 and WBC-13 recorded better RWC, membrane stability, and lower yield reduction (%), while DGC-8, GS-3 and Barsati were sensitive to drought under all irrigation levels (75%, 50% and 25%). These traits would be used as selection criteria for drought stress breeding and could be identified as the marker parameters for explaining the response mechanism of water deficit in cucumber. For yield and related traits (fruit length, diameter and diameter, number of fruits), 3 parents (DGC-19, WBC-13 and DGC-1) out of six parents showed highly significant positive gca effects at both irrigation conditions and were the best general combiners. The best hybrids with maximum heterosis values were DGC-1 x WBC-13 for fruit weight, DGC-13 x DGC-19 and WBC-13 x DGC-8 for fruit numbers and WBC-13 x GS-3 and DGC-19

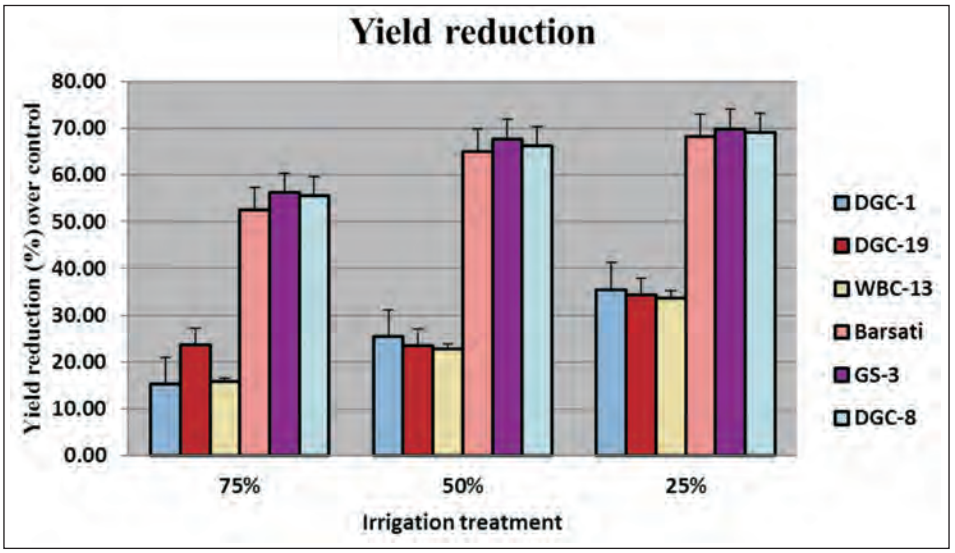

Fig. 11: Effect of irrigation treatment on yield of different genotypes of cucumber

x DGC-8 for yield per plant at 25% irrigation and all these crosses had one of the parents was resistant and another is susceptible (Farag, 2016).

Twenty five tomato genotypes were screened for low temperature tolerance under open field conditions and further selected tolerant and susceptible genotypes were screened under control conditions (phytotron) for confirmation. When grown under field conditions during winter, the membrane stability index was highest (83.27%) in Pusa Sadabahar followed by Pusa Sheetal (81.17%), whereas, it was least in line H-88 (29.67%). The highest number of flowers per truss was observed in the wild species *Solanum pimpinellifolium* and *S. peruvianum* and these lines also had viable pollens, but the number of fruits per truss was negligible in the *S. peruvianum* lines. The cultivar Pusa Sadabahar recorded the highest number of fruits per truss followed by 120-1. *Solanum pimpinellifolium* and S. peruvianum lines, S-699 and DTR-2 showed considerably high shoot dry weight. The physiological parameters like membrane stability index, relative water content, tolerance index and shoot dry weight were reduced in all the genotypes under phytotron in response to low temperature (15/8 and 16/10°C) as compared to plants grown under optimum conditions (26/22°C). Pusa Sadabahar, Pusa Sheetal and DTR-2 showed tolerance to low temperature (15/8 and 16/10°C) with respect to the vegetative characters studied (Seiye *et al.*, 2015). In earlier studies it was reported that cold tolerance in tomato is governed by recessive genes. Pusa Sadabahar x Pusa Rohini exhibited superior performance for fruit diameter, days to first harvest, yield per plant, vitamin C and lycopene content. Pusa Sheetal x Booster showed superior performance for days to 50% flowering, number of fruit set per truss, number of fruits per plant, TSS sand acidity (Droka, 2008).

Twenty one tomato lines/varieties were evaluated to study the morphological, biochemical and molecular differences responsible for their tolerance to heat stress. Pollen germination % and fruit set percentage was recorded maximum in all the tolerant genotypes as compared to heat sensitive genotypes. Relative water content (RWC) and membrane stability index (MSI) under heat stress condition was recorded maximum in Pusa Sadabahar (83% and 86 respectively). Highest proline content was recorded in wild genotypes, like Spm followed by Spr1 and TH-348-4-5-1. Yield per plant had positive and highly significant correlation with pollen germination (%), average fruit weight, fruit set (%), RWC, MSI and lycopene content, but negative correlation with days to 50% flowering (-0.63). High heritability with high genetic advance as per cent over mean was recorded in yield per plant, fruit pericarp thickness, fruit equatorial diameter, chlorophyll a, chlorophyll b and total chlorophyll content. Principal component analysis showed that the first 2 PCs accounting for 77.6 % total variation for morphological traits, while first four PCs accounted for 88.9% of the total variation for biochemical

traits. On over all basis, two varieties, namely Pusa Sadabahar and Balkan and two lines, viz.TH-348-T2 and LP-2 were found most heat tolerant. High value of fruit set (%) and pollen germination, relative water content, membrane stability index, chlorophyll b and proline, and low value of chlorophyll a/b ratio were found most reliable parameters for screening genotypes for heat tolerance (Manish kumar, 2015).

Breeding for Quality

A total of 48 genotypes comprising early, mid-early and mid-late were characterized for 15 morphological and five biochemical traits in cauliflower. In early group, the average marketable curd weight was 0.358 kg which was statistically different from most of the genotypes. The highest GCV (30.89%) and PCV (41.21%) were found for marketable curd weight. The biochemical properties such as curd sinigrin was found to be maximum in DC 41-5 (16.37 µmol $100g^{-1}$ FW) whereas CC 13 had 15.43 µmol $100g^{-1}$ FW of leaf sinigrin. Highest GCV (57.22%) and PCV (57.25%) were recorded for curd sinigrin. Net curd weight (kg) and yield q/ha were recorded highest for Pusa Sharad with 0.310 kg and 103.33 q/ha, respectively for mid-early group. DC 326 attained highest curd sinigrin of 36.93 µmol $100g^{-1}$ FW whereas leaf sinigrin was highest for DC 306 (39.50 µmol $100g^{-1}$ FW). The average net curd weight of mid-late cauliflower was 0.271 kg, where CC 32 and DC 431 had highest net curd weight (0.387 kg). Curd and leaf sinigrin were detected maximum for Pant Gobhi 2 and Selection 1-2 with 16.45, 17.56 µmol $100g^{-1}$ FW, respectively. Mid-early genotypes had maximum glucosinolate (sinigrin). The average antioxidant (CUPRAC and FRAP assay) and total phenol content were highest in early genotypes though mid-early genotypes comprised of highest ascorbic acid content (Vanlalneihi, 2016).

Thirty four onion genotypes were evaluated for their antioxidant activity (AOX) content of phenolics, quercetin and pungency. The AOX in red genotype with highest levels of phenolics is roughly three times higher than commercial white genotype. Pungency levels range from 3.12 to 10.48 µmoles pyruvic acid/g. The mean phenolic and quercetin content was ranged from 60.1 to 1094.8 mg gallic acid equivalents/kg and 22.0 to 890.5 mg/kg, respectively. AOX in red genotype (expressed as µmoles trolox/g) ranges from 1.97 to 5.45 and 3.60 to 6.61 in ferric reducing antioxidant power and cupric ion reducing capacity assays, respectively. Selections, Sel. 383, Sel. 397, Sel. 61, 383, and inbreds I 40, I 80 and Burgundy were found to be potential genotype with high pungency and AOX. An improvement in breeding effort designed to increase the phenolics, quercetin and total AOX in onions (*Allium cepa* L.) provides selected material for analysis. Onion genotype and breeding lines with improved AOX are identified that would help breeders produce onions high in health promoting compounds (Dalamu *et al*. 2008). In another experiment, hybrid A × RO-597 was found superior for phytochemicals and minerals content like Cupric Reducing Antioxidant Capacity (CUPRAC), Ferric Reducing Antioxidant Power (FRAP), anthocyanin, potassium, calcium, magnesium, zinc and iron followed by A × Sel.383. The promising CMS based hybrids for storing bulbs upto five months were A × Sel. 121, and A × Sel. 106 A × Sel.397 with least storage losses. The best combinations may be recommended for commercial cultivation for production bulbs, rich in phytochemicals and minerals, beside yield and good storage capability. (Khade, 2012)

In cabbage, significant differences for antioxidant content and horticultural traits among 36 diverse genotypes revealed the presence of sufficient variability which could be exploited for developing nutritionally and antioxidant rich cultivars. Moderate to high heritability was evident for all the traits and was accompanied with high genetic advance for carotenoids (138.25%), dry matter (62.74%), plant height (49.76%), gross plant weight (65.35%), net head weight (64.71%), stalk length (81.82%), core length (57.72%) and head compactness (76.0%) which showed the presence of additive gene action. On the other hand, low genetic advance observed for ascorbic acid, number of non-wrapper leaves, head shape index, days to 50% maturity and harvest index reflected the predominance of non-additive gene action. (Singh *et al.*, 2011)

In ridge gourd, 28t genotypes, including seven parental lines and 21 crosses, were grown in randomized block design with three replications for fruit sample collection. The highly significant mean squares due to parents, hybrids and parents versus hybrids; and GCA and SCA for yield and antioxidants (ascorbic acid, total

carotenoids, total phenolics, DPPH-RSA, ABTS-RSA and CUPRAC assay) indicate the existence of abundant genetic variation. The per se performance and combining ability of hermaphrodite parents (Satputia Long and Satputia Small) and hybrids of monoecious × hermaphrodite cross were found to be superior for antioxidants along with yield potential. The cross combinations with superior *per se* performance coupled with high SCA estimates and having at least one hermaphrodite parent would be useful for concentrating desirable alleles to improve the antioxidants and yield simultaneously (Karmakar *et al.*, 2014).

The hybrids were found to be superior over their parents for various nutritional traits such as ascorbic acid, total carotenoids and total phenolics content (Fig 12); *in vitro* antioxidant activity in the form of DPPH-RSA, ABTS-RSA and CUPRAC; and P, K, Ca, Na, S, Fe, Zn and Mn content in the fruits. All the nutritional parameters exhibited higher values of dominance variance over the additive genetic variance, more than unity value of average degree of dominance and low narrow-sense heritability (<50 %). The antioxidants and mineral content in the fresh fruits are predominantly attributed by the non-additive genetic component. Hence, the various tools of hybrid breeding would be useful to breed antioxidants- and minerals-rich genotypes of ridge gourd (*Luffa acutangula* Roxb.). (Karmakar *et al.*, 2014)

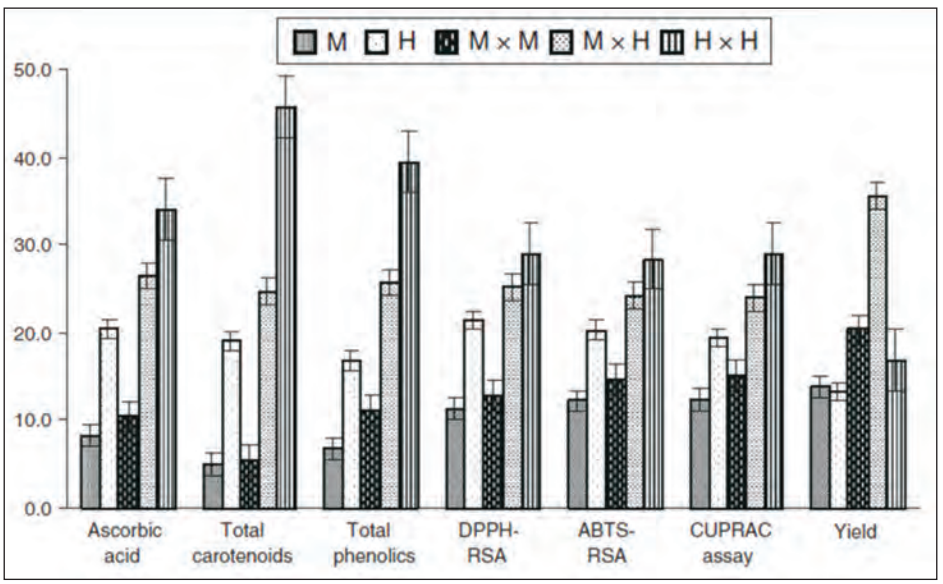

Fig. 12: Relative content and activity of antioxidants and yield potential in monoecious (M) and hermaphrodite (H) parents and their hybrids in ridge gourd (along with standard error bar)

In carrot, IPC 76 (9.47 mg/100 g) had the highest content total carotenoids followed by IPC 25 (9.35 mg/100 g) and IPC 98 (9.17 mg/100 g). The genotype IPC 122 (4.51 mg/100 g) recorded highest lycopene followed by IPC 13 (4.03 mg/100 g) and IPC 34 (3.99 mg/100 g), whereas the genotype IPC 76 (3.06 mg/ 100 g β-carotene, 2.80 mg/100 g lutein) also contained highest β-carotene and lutein followed by IPC 53 (2.89 mg/100 g) and IPC 55 (2.29 mg/100 g) for β-carotene, and IPC 55 (2.55 mg/100 g) and IPC 25 (2.54 mg/100 g) for lutein, respectively. Iron was found to be highest in IPC 98 (16.52 mg/100 g) followed by IPC 13 (15.15 mg/100 g) and IPC 25 (14.53 mg/100 g). This indicates that the genotypes IPC 76, IPC 55, IPC 25, IPC 98, IPC 122, IPC 13 and IPC 53 were invariably among top three ranking genotypes for quality traits. The genotype IPC 126, however, had highest anthocyanin content (144.65 mg/100 g) and root yield of 45.4 t ha-1 which is statistically at par with IPC 76, a third top ranking for root yield. These genotypes, therefore, should be useful for improving root yield and quality attributes of hybrids, simultaneously (Kushlaf and Kalia, 2012).

In carrot, six generations of twelve crosses were subjected to generation mean analysis for inheritance of economic traits. The root epidermal and xylem colour was governed by single dominant gene(s) namely, purple (*P*), orange (*O*), red (*R*) and yellow (*Y*), respectively in different combinations. On the contrary, the phloem colour was controlled by yellow (*Y*), red (*R*), orange (*O*) and purple (*P*), respectively. The phenotyping by HPLC analsysis and RHS based epidermal purple groupings in F_2 population of White Pale × IPC-126, IPC-122 × IPC-126, PM × IPC-126, IPC-126 × White Pale (Fig 13 and 14), IPC-122 × IPC-126 and PM × IPC-126 results revealed high stability of acylated compounds of anthocyanin in RHS-N77A, RHS-59B, RHS-187A and RHS-77B purple groups. Furthermore, peonidin derivatives were identified in RHS-77B groups (Selvakumar, 2016).

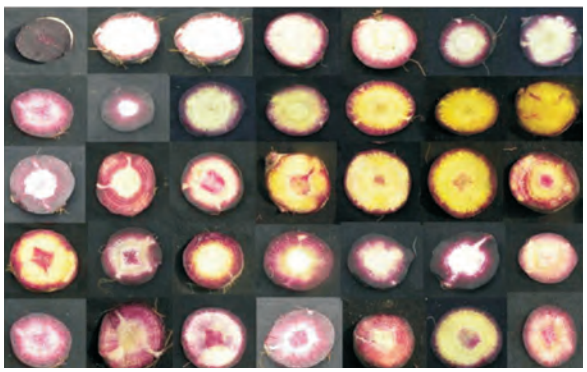

Fig. 13: Yellow with purple segregants in phloem and xylem regions from cross IPC-126 × White Pale

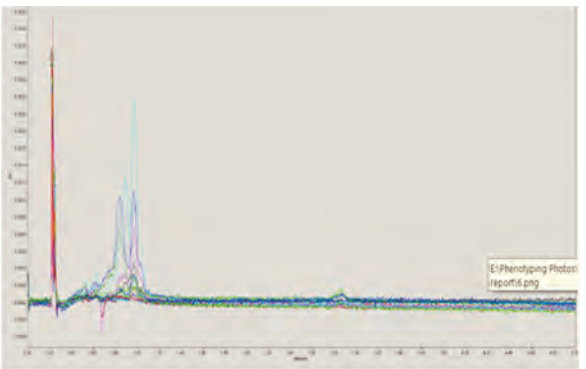

Fig. 14: Carotenoid profile of RHS base yellow with purple groups of F2 population from cross IPC-126 × White Pale

Inter-specific hybridization

A crossing programme involving seven species of *Momordica* and two varieties of *Momordica charantia* was undertaken. Within the sect. *Momordica*, high crossability and pollen fertility was observed in the inter-varietal cross (*M. c.* var. *charantia* × *M. c.* var. *muricata*), whereas low crossability and moderate pollen fertility was observed in the inter-specific cross (*M. charantia* × *M. balsamina*). No crossability barriers were found within the sect. *Cochinchinensis*, except for *M. cochinchinensis* × *M. dioica* and *M. cochinchinensis* × *M. sahyadrica*. *M. dioica* and *M. sahyadrica* showed higher crossability with *M. subangulata* subsp. *renigera* (both directions) and *M. cochinchinensis* (unidirectional). *M. s.* subsp. *renigera* had reproductive compatibility with *M. cochinchinensis* in both directions. *M. sahyadrica* and *M. dioica* showed high crossability in both directions and produced fertile hybrids. Cross between the sect. *Momordica* and *Cochinchinensis* yielded parthenocarpic fruits. *M. cymbalaria* (sect. *Raphanocarpus*) was neither crossable with sect. *Momordica* nor sect. *Cochinchinensis*. The chromosome numbers of the hybrids were as expected from the parental numbers. Based on crossability, a closer relationship was found between two varieties of bitter melon (var. *charantia* and var. *muricata*) and also between *M. charantia* and *M. balsamina*. All dioecious species included in this study appear to be closely related. The result supports that recent taxonomic revision of the genus and the gene pool classification provides a base for improvement of *Momordica* species (Bharathi *et al.,* 2012).

Somatic chromosome number and detailed karyotype analysis were carried out in six Indian *Momordica* species viz. *M. balsamina*, *M. charantia*, *M. cochinchinensis*, *M. dioica*, *M. sahyadrica* and *M. cymbalaria* (syn. *Luffa cymbalaria*; a taxon of controversial taxonomic identity). The somatic chromosome number $2n = 22$ was reconfirmed in monoecious species (*M. balsamina* and *M. charantia*). Out of four dioecious species, the chromosome number was reconfirmed in *M. cochinchinensis* ($2n = 28$), *M. dioica* ($2n = 28$) and *M. subangulata* subsp. *renigera* ($2n = 56$), while in *M. sahyadrica* ($2n = 28$) somatic chromosome number was reported for the first time. A new chromosome number of $2n = 18$ was reported in *M. cymbalaria* against its previous reports of $2n = 16, 22$. The

karyotype analysis of all the species revealed significant numerical and structural variations of chromosomes. It was possible to distinguish chromosomes of *M. cymbalaria* from other *Momordica* species and also between monoecious and dioecious taxa of the genus. Morphology and crossability among the dioecious species was also studied. Evidence from morphology, crossability, pollen viability and chromosome synapsis suggests a segmental allopolyploid origin for *M. subangulata* subsp. *renigera*. The taxonomic status of the controversial taxon *M. cymbalaria* was also discussed using morphological, karyological and crossability data. (Fig 15). (Bharathi *et al.*, 2011)

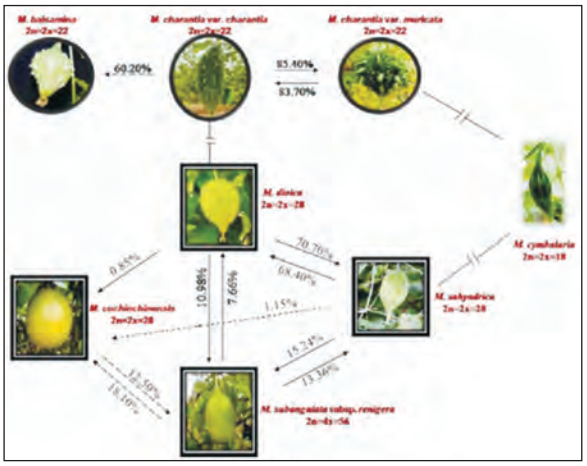

Fig. 15: Crossing relationships among seven species of *Momordica*. → indicates crossing direction from female to male. Double side block (—| |—) indicates that fruit set/fruits set with viable seeds were not obtained. Dashed line (----) indicates partially cross-compatible (< 50% fruits with viable seeds set). Solid line (→) indicates completely cross-compatible (> 50% fruits with viable seeds set). Numbers above the arrows represent percentage of pollen fertility.

In brinjal, interspecific hybridization was carried out involving thirteen cultivated genotypes of eggplant and four wild *Solanum* species (*Solanum incanum*, *S. aethiopicum*, *S. integrifolium* and *S. indicum*) (Fig 16). The cultivated genotypes were used as female parents and wild species as pollen parents in different cross combinations. The crossability relationship of *S. melongena* genoptypes with the wild species was determined by occurrence of fruit set (%), number of seeds per fruit and germination (%) of F1 seed. The mean fruit set was maximum for DBSR-91 (41.25%) followed by DBR-G-190 (37.91%) and NDB-25 (32.08%).

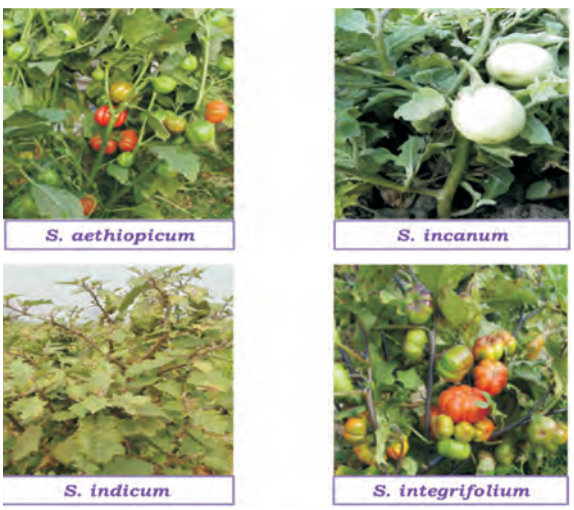

Fig. 16: Wild species used in inter-specific hybridization

The maximum number of seeds per fruit (266.92) was obtained in DBR-G-190 followed by Pusa Uttam (101.42) and Sel-91-2 (92.00). The mean value of highest germination (%) of F_1 seed was observed for DBSR-91 (41.58%) followed by Pusa Uttam (40.25%) and Sel-91-2 (35.50%) when crossed with wild species. The maximum fruit set (80%) was recorded in cross DBSR-91 × S. aethiopicum followed by Pusa Bindu × S. aethiopicum (75%) and NDB-25 × S. aethiopicum (75%). The highest germination of F1 seed (71%) was recorded in three cross combinations, Pusa Uttam × S. aethiopicum, Sel-91-2 × *S. aethiopicum*, DBR-G-190 × *S. aethiopicum*. The highest number of seeds per fruit (754) was recorded in DBR-G-190 × *S. incanum* followed by DBSR-52 × *S. incanum* (226.6), DBR-G-190 × *S. aethiopicum* (206). Among the four wild species, *S. incanum* was found highly crossable with the cultivated S. melongena genotypes. Interspecific crosses reported in this study will be used in transferring desirable traits in different genetic backgrounds of cultivated *S. melongena* (Premabati Devi *et al.,* 2015).

To utilize alien resistance sources, interspecific hybrid and their backcross progeny (B_1) were generated between cauliflower (2n=18, C genome) 'Pusa Sharad' and Ethiopian mustard (2n=34, BC genome) 'NPC-9' employing *in vitro* embryo rescue techniques (Fig 17). All the F_1 Inter-specific plants were found to be resistant to black rot disease on artificial inoculation *Xcc* race 1. Cytology of pollen mother cells (PMCs) of the digenomic F_1 hybrid (BCC) revealed the expected number of chromosomes (2n = 26) at Metaphase I in all the PMCs.

Hybridity confirmation of F_1 inter-specific hybrid was carried out by using a dominant morphological marker (anthocyanin pigmentation on anther tip), one microsatellite co-dominant marker (NI2-C01) and *Brassica* B and C genome-specific (DB and DC) primers. The mean pollen viability of interspecific hybrid was very low (2.77 %) when compared with Pusa Sharad (98.00 %) and 'NPC-9' (93.33 %). Average pollen viability (16.30 %) of BC_1 plants has been increased and observed in a range of 9.00 to 28.00 %. Fertile giant pollen grains (size 45.94 µm) formation was observed in inter-specific F_1 and BC_1 generation. An intron polymorphic marker (At1g70610) from B-7 chromosome of *B. carinata* linked to black rot resistance gene confirmed introgression in BC_1 population. Segregation distortion was observed for black rot resistance including vegetative and floral traits in inter-specific BC_1 generation. This study reports marker assisted introgression of B genome specific *Xcc* resistance from *B. carinata* to *B. oleracea* var. *botrytis* L. employing embryo rescue. This effort will go a long way in pyramiding gene(s) for resistance against black rot in Cole crops especially cauliflower and cabbage for developing durable resistance to minimize dependency on chemical protectants. (Sharma, 2016).

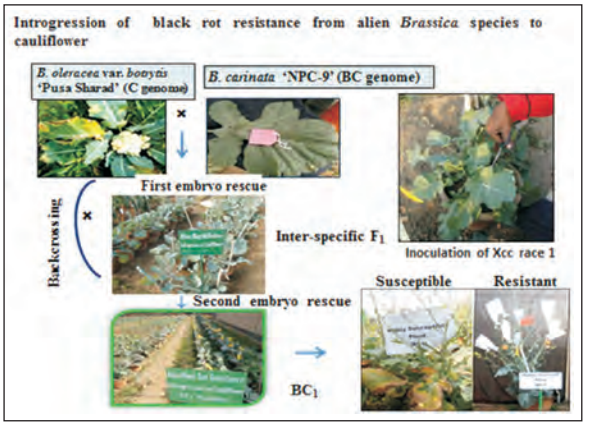

Fig. 17: Introgression of black rot resistance from *Brassica carinata* to *Brassica oleracea* var. *botrytis*

Interspecific hybridisation in okra (*Abelmoschus esculentus* (L.) Moench).' was carried out. The results of crossability studies revealed that DOV-1 × *A. tetraphyllus* showed maximum average fruit set % (52) and seed germination % (90) and DOV-1 × *A. moschatus* showed maximum average fruit set % (60) and seed germination % (45). The morphological studies on interspecific F_1 showed that they were intermediate between both parental species for many characteristics studied as per DUS guidelines. (Xavier, 2016)

Molecular breeding/MAS

Sponge gourd (*Luffa cylindrica* Roem.) is a popular cucurbitaceous vegetable on the Indian sub-continent. The crop is severely affected by *Tomato Leaf Curl New Delhi Virus* (ToLCNDV) a Geminivirus causing a yellow mosaic disease which results in 100% crop loss.

Two sequence-related amplified polymorphism (SRAP) markers were closely-linked to the ToLCNDV-susceptible gene in the susceptible parent and in a susceptible bulk population, while two SRAP markers were closely-linked to the resistance gene in the resistant parent and in a resistant bulk population. The SRAP markers thus identified can be used for early seedling selection in a large number of genotypes and should speed-up the development of *Luffa* cultivars that are resistant to ToLCNDV, thereby decreasing the incidence of this disease in this crop. (Islam *et al.,* 2011)

Black rot is the most devastating disease of cauliflower worldwide causing severe damage to crop. The identification of markers linked to loci that control resistance can facilitate selection of plants for breeding programmes. In the present investigation, F2 population derived from a cross between 'Pusa Himjyoti', a susceptible genotype, and 'BR-161', a resistant genotype, was phenotyped by artificial inoculation using Xcc race 1. Segregation analysis of F2 progeny indicated that a single dominant locus governed resistance to *Xcc race 1* in 'BR-161'. Bulk segregant analysis in resistant and susceptible bulks of F2 progeny revealed seven differentiating polymorphic markers (three RAPD, two ISSR and two SSR) of 102 markers screened. Subsequently, these markers were used to genotype the entire F2 population, and a genetic linkage map covering 74.7 cM distance was developed. The major locus Xca1bo was mapped in 1.6-cM interval flanked by the markers RAPD 04833 and ISSR 11635. The *Xca1bo* locus was located on chromosome 3. The linked markers will be useful for marker-assisted resistance breeding in cauliflower (Saha *et al.,* 2014).

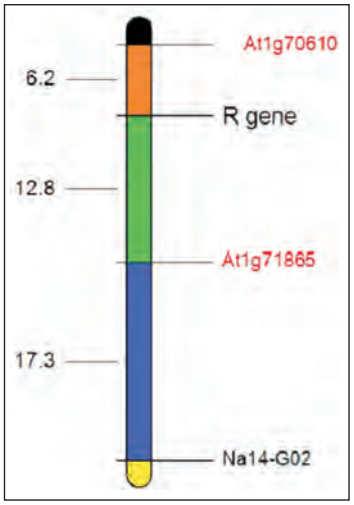

Fig. 18: Linkage map of black rot resistance locus (R Gene) on B-7 chromosome of *Brassica carinata*

Since there are limited sources of resistance to black rot in *B. oleracea* (C genome Brassica), exploration of A and B genomes of *Brassica* was planned as these were thought to be potential reservoirs of black rot resistance gene(s). In our search for new gene(s) for black rot resistance, F_2 mapping population was developed in *Brassica carinata* (BBCC) by crossing NPC-17, a susceptible genotype with NPC-9, a resistant genotype. Out of 364 Intron length polymorphic markers and microsatellite primers used in this study, 41 distinguished the parental lines. However, resistant and susceptible bulks could be distinguished by three markers At1g70610, SSR Na14-G02 and At1g71865 which were used for genotyping of F_2 mapping population. These markers were placed along the resistance gene, according to order, covering a to *Or* gene with in a distance of 0.5 to 0.8 cM were used in our study distance of 36.30 cM. Intron length polymorphic markers At1g70610 and At1g71865 were found to be linked to black rot resistance locus (Xca1bc) at 6.2 and 12.8 cM distance, respectively. This is the first report of identification of markers linked to Xca1bc locus in *Brassica carinata* on B-7 linkage group (Fig 18). Intron length polymorphic markers provided a novel and attractive option for marker assisted selection due to high cross transferability and cost effectiveness for marker assisted alien gene introgression into cauliflower (Sharma *et al.,* 2016).

The investigation was undertaken to introgress β-carotene (pro-vitamin A) enhancing *Or* gene from donor line (1227) into three mid-early maturity group II (November- December) genetic backgrounds of open pollinated variety Pusa Sharad (DC 309) (Fig 19) and the parents of Pusa Hybrid-2, namely CC-35 and DC 18-19. The *Or* gene turned normal white curd colour into orange increasing average β-carotene content in cauliflower from 0.14 to 14.81 µg/g. The three pairs of reported SCAR markers (SA4, SA7, SA9) closely linked for foreground selection. The SCAR marker SA4 was utilized for foreground selection for BC_1F_1 populations of DC 309 × 1227 combination. Newly designed *Or* gene specific STS marker utilized for identifying the homozygous orange inbred line (1227) at 576 bp. Total of 1013 markers (SSR, STS) were used for polymorphic survey, 68 (6.65%) of these were polymorphic between the parents DC 309 and 1227 followed by 6.12% in CC-35 and 1227 and 5.82 % in DC 18-19 and 1227. The background analysis showed maximum recovery of the recurrent parent genome (RPG) to the level of 72%. The β-carotene content of BC_1F_1 dark orange categories were average β-carotene were 14.81 µg/g (DC 18-19 × 1227), 14.00 µg/g (CC-35 × 1227) and 3.56 µg/g (DC 309 × 1227). The combination of high RPG with high level of β-carotene content was recorded in two lines 18-19-1-7-7 (18.60 µg/g) and 18-19-1-3-6 (17.94 µg/g). The selected BC_2F_1 plants advanced to BC_2F_2 and BC_3F_1 generations. The backcross derived line 18-19-P-7 accumulated high level of β-carotene (10.28 µg/g). The promising lines with more than 12 µg/g β-carotene were advanced to BC_2F_2 and BC_3F_1 (Muthukumar, 2016)

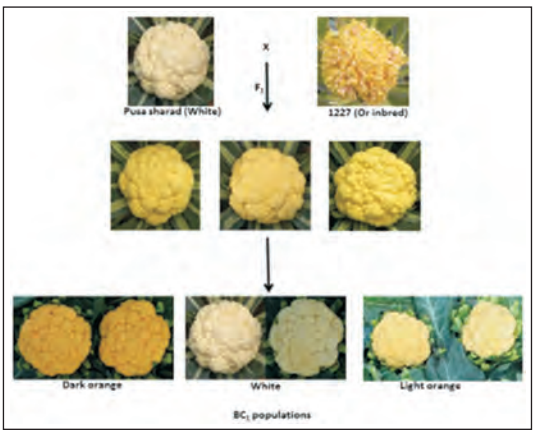

Fig. 19: Curd colour phenotype observed in BC_1 population of cauliflower cross (Pusa Sharad × 1227)

In carrot, the F_2 mapping population developed from the cross White Pale × IPC-126 was evaluated and phenotyped (Fig 20a) and was genotyped using SSR markers to identify markers for anthocyanin content. A linkage map was constructed with 7 SSR markers which were linked chromosome number 3 covering a total length of 10.2cM. Markers of BSSR-43 and GSSR-14 were found to be tightly linked with anthocyanin locus (*P*) at 1.4cM and 4.0cM distance, respectively (Fig 20 b). Therefore, GSSR and BSSR markers will play an important role for precise phenotype selection, cost effectiveness and time saving in carrot marker assisted breeding. (Selvakumar, 2016).

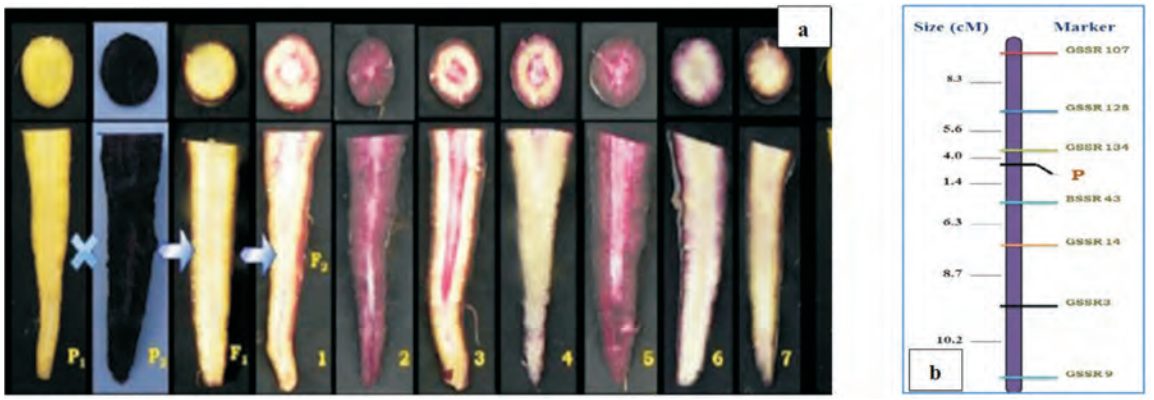

Fig. 20 (a &b): (a) Phenotyping of F_2 mapping populations from cross White Pale × IPC-126 and (b) linkage map of *P* locus corresponds to chromosome 3 of *Daucus carota* L.

In vitro techniques

Micro-propagation can be exploited for maintenance and mass multiplication of gynoecious lines in bitter gourd for hybrid seed production. Therefore, standardization of protocol for rapid *in vitro* multiplication (Fig 21) will also help in development of double haploid (DH) plants in bitter gourd, which may enhance the ability to develop homozygosity in a short period particularly for gynoecious trait. Among the explants, the shoot tip performed better than the nodal explant in all respects. T4 treatment, which is a combination of spray of gentamycin (100 mg/l) + agitation of explants in carbendazim (0.2%) + Mancozeb (0.2%) HQC (200 mg/l) is best for the pretreatment of cultures. Among the stages, 45 days old explants are comparative better than 60 days old. Treatment with mercuric chloride (HgCl2) for 3 minutes duration recorded the maximum *in vitro* survival of the shoot tip of DBGy -201 (57.96%) (Saha and Behera, 2015).

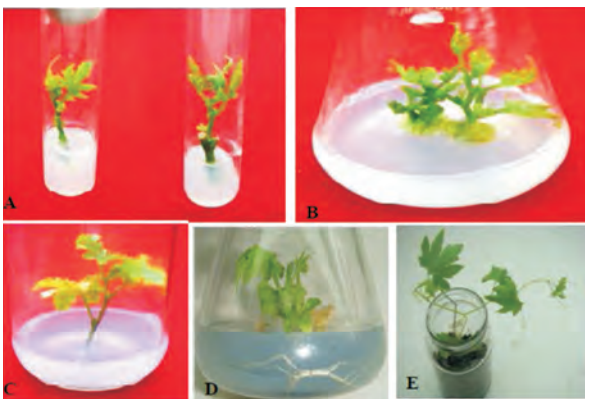

Fig. 21: A. Shoot initiation from apical bud and nodal segment; B. Shoot multiplication through subsequent sub-culturing; C. shoot elongation; D. Rooting and E. Acclimatization of plants in glass jars.

This study was undertaken to standardize an efficient protocol for *in vitro* mass multiplication and maintenance of bitter gourd gynoecious line for its use in hybrid seed production. The apical bud gave better response than nodal segment for culture initiation. Murashige and Skoog (MS) basal medium supplemented with 6 benzyl-aminopurine (2 mg/l) + α-Napthalene acetic acid (0.2 mg/l) was found best for *in vitro* survival (81.3%) in plantlets raised through nodal segment, while it was 77.84% in apical bud. The minimum duration (5.53 days) for bud sprouting was recorded for apical bud. Medium combination MS + BAP (1.0 mg/l) + IBA (0.1 mg/l) + GA3 (0.3 mg/l) was found best earliest shoot proliferation (11.9 &14.62 days for apical bud and nodal segment). The apical bud and nodal segment regenerated 4.77 and 3.56 shoots/ explant, respectively on the same medium. Elongation of microshoots was achieved maximum with MS + GA3 (1 mg/l). The micro-shoots were rooted on full-strength MS medium supplemented with GA3 (1 mg/l) + activated charcoal (100 mg/l). Minimum days were recorded for rooting (10.81) for apical bud and 11.49 days for nodal segment. The percent survival (81.25%) was maximum in glass jar with PP cap in the shoot tip derived plantlets, which was at par with the nodal segment (80.94%). Rooted plants were acclimatized in the greenhouse and subsequently established in soil. The protocol developed for this study led to an alternative for easy maintenance and use in gynoecious inbred development in bitter gourd (Saha *et al.,* 2016).

A successful response in terms of interspecific F_1 hybrid and BC_1 generation development were obtained in cross combination Cauliflower 'Pusa Sharad × Ethiopian Mustard 'NPC-9' using *in vitro* embryo rescue. In this study, researcher tested efficacy of two *in vitro* embryo rescue methods under two different temperature regimes. The success rate of siliqua culture method was slightly higher (2.12 %) at low temperature regime (19.4 ºC/4.3 ºC) than high temperature regime (26.4 ˚C /10.4 ˚C) (1.85%). During low temperature regime (19.4 ºC/4.3ºC), 7-11 DAP stage were identified optimum for *in vitro* siliqua culture producing maximum number (36) of turgid ovules. Two ovules of these could be germinated and ultimately only one grew up as true hybrid. Very slow growth of hybrid siliqua was observed due to low temperature prevalence in natural open conditions. During high temperature regime, 5-7 DAP stage of hybrid siliqua were found optimum for siliqua culture. However, some of the ovaries ceased growing at later stage and their embryos were degenerated. Even single turgid ovule could not be obtained from 15 DAP. Out of 30 turgid ovules, only one germinated and produced hybrid. The success rate of siliqua culture method was slightly higher (2.12 %) at low temperature regime (19.4 ºC/4.3ºC) than high temperature (26.4 ˚C /10.4 ºC) regime (1.85%). In direct ovule culture method, the maximum numbers of turgid ovules (125) were obtained by culturing 240 ovaries. During low temperature regime, 42 turgid ovules were obtained at 15- 19 DAP stage of embryo rescue. Out of these 04 ovules germinated and hybrid seedling could be produced at a 17 DAP stage. In high temperature regime, 52 turgid ovules were obtained at 11-15 DAP and 05 of these germinated, two of which grew as hybrid plants. The success rate direct ovule culture method was higher (4.14 %) at high temperature regime than low temperature regime period (1.75%). The average success rate of direct ovule culture method was higher (3.20 %) than siliqua-ovule culture (1.98) (Fig 22) The direct ovule culture was followed to produce backcross generation (BC_1) (Sharma, B.B, 2016).

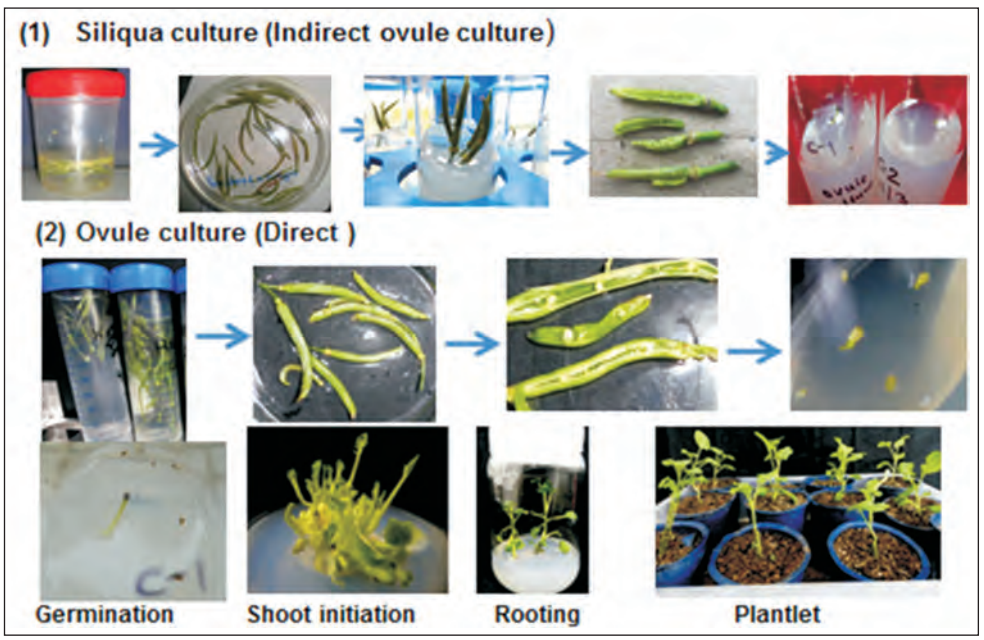

Fig. 22: Standardization of *in vitro* embryo rescue protocol for wide hybridization of Cauliflower × Ethiopian Mustard

Studies on haploidisation in short day onion (*Allium cepa* L.)" was aimed to develop efficient protocol for haploid production in onion variety Pusa Riddhi (Fig 23). The maximum percentage of gynogenesis induction on MS medium, supplemented with 2, 4-D 2mg/l +BAP 2mg/l (HAP30); NAA 2mg/l + 2iP 1mg/l (HAP32); NAA 2 mg/l + 2iP 2mg/l (HAP34) and NAA 0.5 mg/l + BA 1.0 mg/l combinations can be used. Out of all these, HAP30 (2, 4-D 2mg/l +BAP 2mg/l) media resulted in maximum gynogenesis induction. Plantlet obtained from HAP02 (2,4-D 1mg/l + BA 2mg/l) medium which was confirmed cytologically to be haploid

Fig. 23: Gynogenic plantlets emerging from cultured flowers (a to e) normal Plantlets (f) an albino plantlet (as viewed under stereo zoom microscope 20X).

was subjected to flow cytometry analysis. On the basis of nuclear DNA content and its comparison with normal onion as an internal standard, Flow cytometry confirmed the plant to be haploid. (Mathapati, 2016).

Improved production technology

The experiment was conducted to evaluate the performance of hybrids in bitter melon by making crosses on the tissue cultured raised gynoecious line, DBGY- 201 with twelve other inbreds. The twelve different inbreds (Pusa Do Mausami, Pusa Vishesh, Sel 2, Nakhara Local, MC 84, S26, S29, S30, S41, S54, S57 and DBG 34) were crossed with DBGY-201 to produce 12 crosses. The F1 hybrids along with three commercial monecious × monoecious hybrids namely VNR 22, US 33, PH 2 were used as checks to compare the performance of gynoecious × monoecious hybrids. The experiment was laid out in randomised block design with 3 replications. The seedlings were transplanted on both sides of the channel with 2m between channel and 45 cm between plants. The recommended NPK fertilizer doses and cultural practices along with plant protection measures were followed to raise an ideal crop. The bitter gourd is mainly grown in long growing spring summer season (February to May) in north Indian plains. But this experiment was carried out in wet season in order to evaluate the performance of these hybrids in off season. Because of the short period growing season i.e. July-September only two harvests were taken. Fruit length and diameter are also important attributes to determine yield. The maximum fruit length was in DBGy- 201 x S54 with 11.07 cm followed by DBGy- 201 x Pusa Vishesh with 10.87 cm (Table 1). The maximum fruit diameter was registered in DBGy- 201 x Pusa Vishesh (11.87 cm) and DBGy- 201 x Nakhara Local (11.30 cm). The parents with the larger fruits might have contributed to increase the fruit length in hybrids. Among the checks VNR 22 recorded the maximum fruit diameter (12.20 cm). The maximum fruit weight was registered in hybrid Gy x S54 (39.18 g) and was at par with the check VNR 22 (39.43 g). DBGy- 201 x Pusa Vishesh hybrid had also larger fruit weight (38.43 g) while the minimum was in DBGy- 201 x S29 (23.77 g) and the monoecious × monoecious hybrid US 33 produced smallest fruit (15.07 g). Maximum yield per plant (245.86 g) was registered in hybrid DBGy- 201 x Pusa Vishesh with followed by DBGy- 201 x MC 84 (240.21 g). It was found that all the hybrids performed extremely well in comparison to checks in terms of yield (Khan *et al.,* 2011).

Acclimatization of micro propagated plants corresponds to a transition period when roots become adapted to a substrate with less available nutrients, and to an autotrophic condition. To ameliorate this problem in bitter gourd, 30-day-old, in vitro rooted plantlets of bitter gourd cultivars Pusa Do Mausami, Pusa Vishesh and the DBGy 201 were subjected to root inoculation with different arbuscular mycorrhiza fungal (AMF) strains.

All the mycorrhizal treatments showed almost two times higher ex vitro survival than the control plantlets. Mycorrhization plantlet showed increase in vine length in Pusa Vishesh (194.02 cm) in mixed strain, leaf area in Pusa Vishesh (107.91 cm2) in *Acaulospora scorbiculata*, chlorophyll in Pusa Do Mausami (3.29 mg/g FW) in A. scorbiculata) and total phenols content in Pusa Do Mausami (7.84 µg/g FW) in E. columbiana). Photosynthetic rates were enhanced in arbuscular mycorrhizal fungi (AMF) treated plant in Pusa Do Mausami (10.75 µmol CO2/m2 /s1) in mixed strain in comparison to an uninoculated control. Among the AMF species, mixed strain (Nutrilink®) showed good as high as 38% root colonization for all the cultivars. In this experiment the mixed AMF strain has contributed significantly in survival of the plantlets and plant establishment in the field (Saha *et al*., 2013).

The present investigation was carried out under two set of growing conditions viz., insect-proof net house and open-field condition. Highly significant differences in growth and flowering attributes between the seed and pollen parent were observed under insect-proof net house and open field conditions. There was early appearance of female and male flower and significantly higher number of female flower in seed parent and male flower in pollen parent in insect-proof net house. The number of fruits set per plant (2.40) and number of mature fruits per plant (1.90) were significantly superior over the number of fruits set per plant (1.40) and number of mature fruits per plant (1.1) under open-field conditions. Significantly higher fruit weight (1222.70 g/fruit), fruit length (36.07 cm), fruit width (9.19 cm) and flesh thickness (2.14 cm) was recorded when retained two fruit per plant in insect proof net house as compared to open field (590.80 g., 29.97 cm, 7.08 cm and 2.01 cm), respectively .The incidence of virus was very high under open-field, as compared to insect-proof net house.. Seed yield and its contributing characters were significantly higher under insect-proof net house as compared to open field grown seed crop. Seed yield per fruit (38.47 g), seed yield per plant (73.09 g), seed yield hectare (1096.42 kg) was significantly higher under insect-proof net house in comparison to open field (316.88 kg). The investigation highlighted the commercial exploitation insect proof net house for hybrid seed production of summer squash hybrid Pusa Alankar under Northern plains conditions (Aheer, K.B., 2012).

Effect of Silver Nitrate, GA3 and Silver Thiosulfate in regard to induction of hermaphrodite flower in gynoecious line of bitter gourd var. DBGy 201 was evaluated at different concentration. Silver Thiosulfate @ 6M and 3M was found to induce staminal tissue in gynoecious lines of bitter gourd whereas Silver Nitrate @ 200 and 250 ppm was found ineffective and GA3 @ 1000 and 1500 ppm could induce only rudimentary stamen without any fertile pollen and an increased vegetative growth in the present investigation. The induced hermaphrodite flowers exhibited double the size of ovary, stigma, style, stamen, anther, calyx, corolla and petiole of the female flower. Therefore, silver ion applied as Silver Thiosulfate @ 6M is considered the best for inducing hermaphrodite sex form in the gynoecious line of bitter gourd (Mishra, 2012).

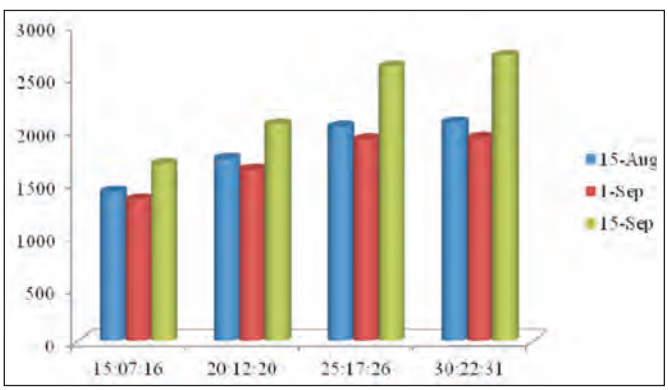

Fig. 24: Effect of date planting and fertilizer dose on fruit yield/ plant (g) of cucumber var. Pusa Seedless Cucumber-6 grown under polyhouse condition

An experiment was conducted to find out the effect of planting time and fertilizer dose on growth, yield and quality of cucumber var. Pusa Seedless Cucumber-6 and bitter gourd var. Pusa Rasdar under two different protected structures. Comparing the effect of protected structures in cucumber yield, it was found that there was significant increase in yield under polyhouse (128.5 q/1000 m^2) compared to net house (111.5 q/1000m2). Under polyhouse condition application of NPK @ 30:22:31 with 15th September planting gave highest yield (180.6 q/1000 m^2) along with higher fruit length, fruit diameter, number of fruits, fruit weight with increased fruit nutrients like nitrogen, phosphorus, potassium, calcium, iron, zinc and manganese. Comparing the effect of protected structures,

the highest yield of bitter gourd was obtained in net house (66.2 q/1000 m^2) compared to polyhouse (64.3 q/1000 m^2). September planting under polyhouse condition can be recommended for successful cultivation of cucumber var. Pusa Seedless Cucumber-6 (Fig 24). While for bitter gourd NPK @ 30:22:31 dose with 15th August planting under net house can be recommended for commercial cultivation of bitter gourd var. Pusa Rasdar. (Maragal, 2016)

Summary

- Phylogenetic studies in 6 *Momordica* species (Fig. 1) were made by utilising 21 RAPD and 12 ISSR markers with 35 genotypes and five genotypes of two *Luffa* species.
- In cucumber, genotypes CRC-8, CHC-2 and G-338 showed lower accumulation of sodium, lesser depletion of potassium, lower Na^+–K^+ ratio and higher accumulation of proline, reducing sugars, phenols, better membrane stability and lower yield reduction (%) under salt stress, while CH-20 and DC-1 were sensitive to salt stress.
- In bitter gourd, tubercles is governed by a single pair of nuclear gene and prominent tubercles was dominant over non tubercles and straight fruit is dominant over curved fruit and governed by a single pair of gene.
- Single dominant gene was found to be resistance to black rot (Xcc race 1) in *Brassica carinata.*
- The gynoecious parental lines (PPC-2 and GPC-1) and monoecious parent Pusa Uday were observed to be the three top performing parents for yield per plant. The best three heterotic hybrids PPC-2 × Pusa Uday, GPC-1 × Pusa Uday and PPC-2 × Punjab Naveen showing 64.51, 55.61, and 54.57% heterosis, respectively, over standard check Pusa Uday for yield per plant, may be exploited for commercial cultivation.
- The hybrids Ogu 119 × Suprimax Late, Ogu 101 × DB-1305, Ogu 119 × Sel-26, Ogu 101 × DB-187, Ogu 13-01 × Kt-18 and Ogu 119 × Lalchowk Maghi were best heterotic combinations for yield, major yield attributing traits and also had high positive heterosis for ascorbic acid and mineral content**.** The hybrids Ogu 101 × Lalchowk Maghi, Ogu 13-85 × Kt-18, Ogu 13-85 × DB-187, Ogu 119 × Kt-22 and Ogu 119 × Suprimax Late were found to be the most promising for earliness and also exhibited high heterosis for yield and yield attributing traits and various mineral elements.
- In okra, 30 diverse genotypes were analysed for 19 quality traits. The maximum iron content was recorded in genotype DOV 66 (1.71ppm) and highest calcium content was recorded in Arka Abhay and DOV 23.1.
- The genetics of resistance against early blight caused by *Alternaria solani* in tomato (*Lycopersicon esculentum* L. cv. 'Megha'), revealed the trait to be governed by two recessive genes.
- The polypeptides encoded by McACSs shows very high identity i.e. upto 99% to one another. McACS1 and McACS4 were most closely related to each other (99% identity) which is followed by McACS2 and McACS5. McACS3 is more closely related to McACS1 and McACS4 than other isomers. However, phylogeny analysis of ACS polypeptides of cucurbits indicated that McACS1, 4 and 3 is most closely related to *Citrullus* spp. Ccol × cla-ACS1 with 93% identity and McACS2 is most closely related to Cme-ACS7 with 72% identity. McACS5 is 84% identical to Cme-ACS1.
- Two varieties 'Pusa Vijay' and 'Pusa Bold' of *B. juncea* (AB genome); 'NPC-9' of *B. carinata* ((BC genome, Ethiopian mustard); five genotype/accessions of *B. nigra* (B genome), namely EC289661, 'Sangam', IC247, IC560690, IC56072 and one accession of *B. oxyrrhina* were found resistant after intensive artificial screening in field and laboratory conditions against *Xcc* race 1.
- The genotypes of cucumber like CRC-8, CHC-2 and G-338 showed lower accumulation of sodium, lesser depletion of potassium, lower Na^+–K^+ ratio and higher accumulation of proline, reducing sugars, phenols, better membrane stability and lower yield reduction (%) under salt stress, while CH-20 and DC-1 were sensitive to salt stress.

- The genotypes of cucumber viz. DGC-1, DGC-19 and WBC-13 recorded better RWC, membrane stability, and lower yield reduction (%), while DGC-8, GS-3 and Barsati were sensitive to drought under all irrigation levels (75%, 50% and 25%). These traits would be used as selection criteria for drought stress breeding and could be identified as the marker parameters for explaining the response mechanism of water deficit in cucumber.
- The cultivar Pusa Sadabahar recorded the highest number of fruits per truss followed by 120-1. *Solanum pimpinellifolium* and S. peruvianum lines, S-699 and DTR-2 showed considerably high shoot dry weight.
- Somatic chromosome number and detailed karyotype analysis were carried out in six Indian *Momordica* species viz. *M. balsamina*, *M. charantia*, *M. cochinchinensis*, *M. dioica*, *M. sahyadrica* and *M. cymbalaria* (syn. *Luffa cymbalaria*; a taxon of controversial taxonomic identity).
- On over all basis, two varieties, namely Pusa Sadabahar and Balkan and two lines, viz.TH-348-T2 and LP-2 were found most heat tolerant. High value of fruit set (%) and pollen germination, relative water content, membrane stability index, chlorophyll b and proline, and low value of chlorophyll a/b ratio were found most reliable parameters for screening genotypes for heat tolerance.
- In cauliflower, the major locus *Xca1bo* was mapped in 1.6-cM interval flanked by the markers RAPD 04833 and ISSR 11635 on chromosome 3.
- The ILPAt1g70610 and At1g71865 were found to be linked to black rot resistance locus (Xca1bc) at 6.2 and 12.8 cM distance, respectively. This is the first report of identification of markers linked to Xca1bc locus in *Brassica carinata* on B-7 linkage group.
- The combination of high RPG with high level of β-carotene content was recorded in two lines 18-19-1-7-7 (18.60 µg/g) and 18-19-1-3-6 (17.94 µg/g). The selected BC_2F_1 plants advanced to BC_2F_2 and BC_3F_1 generations. The backcross derived line 18-19-P-7 accumulated high level of β-carotene (10.28 µg/g).
- The highest number of seeds per fruit (754) was recorded in DBR-G-190 × *S. incanum* followed by DBSR-52 × *S. incanum* (226.6), DBR-G-190 × *S. aethiopicum* (206). Among the four wild species, *S. incanum* was found highly crossable with the cultivated S. melongena genotypes.
- Markers of BSSR-43 and GSSR-14 were found to be tightly linked with anthocyanin locus (*P*) at 1.4cM and 4.0cM distance, respectively in carrot.
- The protocol developed for *in vitro* mass multiplication and maintenance of bitter gourd gynoecious line for its use in hybrid seed production.
- A successful response in terms of interspecific F_1 hybrid and BC_1 generation development were obtained in cross combination Cauliflower 'Pusa Sharad × Ethiopian Mustard 'NPC-9' using *in vitro* embryo rescue. The success rate direct ovule culture method was higher (4.14 %) at high temperature regime than low temperature regime period (1.75%). All the F_1 Inter-specific plants were found to be resistant to black rot disease on artificial inoculation *Xcc* race 1.
- The maximum percentage of gynogenesis induction on MS medium, supplemented with 2, 4-D 2mg/l +BAP 2mg/l (HAP30); NAA 2mg/l + 2iP 1mg/l (HAP32); NAA 2 mg/l + 2iP 2mg/l (HAP34) and NAA 0.5 mg/l + BA 1.0 mg/l combinations can be used. Out of all these, HAP30 (2, 4-D 2mg/l +BAP 2mg/l) media resulted in maximum gynogenesis induction.
- Silver Thiosulfate @ 6M and 3M was found to induce staminal tissue in gynoecious lines of bitter gourd.
- September planting under polyhouse condition can be recommended for successful cultivation of cucumber var. Pusa Seedless Cucumber-6.

Future perspective

- Search for new genes i.e. identification of new source of resistance to different biotic and abiotic stresses in these selected vegetable crops has not been given attention.

- Diversification of sterile cytoplasm using wide hybridization required to be strengthened in different vegetable crops.
- Detail and systematic study on genetics of resistance to different diseases in these selected vegetable crops still remains incomplete.
- Identification of molecular markers and tagging of genomic regions controlling resistance to diseases is very much limited.
- It is not known whether the resistance observed in available resistant sources is against the viruses or to the vector.
- Introgression of desired genes in commercial varieties using both conventional and marker assisted breeding has not been attempted.
- Exploitation of genetic mechanism like CMS, gynoecism and antisense RNA technology etc. for hybrid seed production has not been exploited fully.
- Identifying sources for intrinsic and extrinsic qualities in important vegetable crops has not been given attention in earlier breeding programme.
- Development of varieties/hybrids rich in phyto-nutrients, nutraceuticals and processing traits has not been well focussed earlier.

Acknowledgments

We wish to thank all the Faculty Members of the Division of Vegetable Science for extending support and cooperation in preparing the research contributions of M.Sc. and Ph.D. students.

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School of Social Sciences

Agricultural Economics

Alka Singh

Introduction

The Division of Agricultural Economics, a constituent of School of Social Sciences of IARI, was established in 1960. Since its inception, the Division has been making contributions in basic and applied research with significant implications for agricultural policy. Research focus of the Division has been continuously reoriented to address contemporary development challenges. The division has been focusing on the vital issues facing Indian agriculture in the realms of technology, policy and institutions. The Division gives utmost attention to the newer developments in the subject and tries to apply them in the Indian context.

During the sixties, major research focus of the Division was on farm business analysis, efficient allocation of resources, supply response, input demand analysis and marketing efficiency. To address the challenges and opportunities of the Green Revolution, research programmes during the seventies and eighties were mainly concentrated on capital formation, labour employment, farm mechanization, rural credit needs, yield gap analysis, price policy and subsidy issues, and efficient management of natural resources. In the nineties, issues related to national food and nutrition security, efficiency and sustainability of agricultural production systems, poverty alleviation, research impact assessment and priority setting, export potential of agri-products, etc. were accorded high priority. Besides continuing emphasis on these research programmes, attempts were made in the recent past to assess food demand and supply scenario, impact of WTO regulations, food safety requirements, market information systems, peri-urban agriculture, etc. Currently, the priority areas of research are: Smallholder's productivity, agricultural growth; ecosystem services; inclusiveness and effectiveness of agricultural market and trade reforms; development of agro-industries and impact assessment of agricultural technologies.

Significant Student's Research Achievements

Students' research has been contributing significantly in addressing the prioritized research areas with the help of able guidance of faculty of the division. The significant research contribution is discussed below.

Agricultural Growth, Input use and Farm investments

Many studies have been conducted in the division analysing growth in agriculture sector and drivers of its growth in the changing scenario of economic environment. Summary of findings of such selected studies are presented here.

The status of food processing industry was examined in terms of value addition to agricultural produce and structural changes in input-output relationships therein. Time series data (1981/2009) on gross output, number of factories, input use, number of labourers, emoluments, capital stock and various wholesale and retail level price indices were used for 11 subsectors of the food manufacturing sector at four-digit level of NIC classification. Results discern that food processing sector being more capital intensive could not create sufficient employment opportunity. Capital formation focused primary processing industries viz. sugar. Capital productivity decreased in entire study period with the rate of 9.5 per cent. To tap the potential in food processing sector, call of the

hour is to increase productivity level besides enhancing productive capacity. Quality investment is needed in new technologies, supporting infrastructure and overhead capital. This will lead to the higher productivity, greater economies of scale and will enhance employment.

Table : Decade-wise compound growth rate and average net value added in food processing subsectors in India (at 2004-05 base prices)

	Values in Rs. Billion				CAGR (per cent)			
	TE 1983-84	TE 1992-93	TE 2001-02	TE 2009-10	1981-1990	1991-2000	2001-2009	All Period
Meat	0.34	0.23	0.23	0.22	6.30	3.93	23.61	10.91
Fish	1.15	1.46	1.60	1.47	8.56	-0.50	8.50	4.70
Fruits and Vegetables	0.52	0.74	0.92	0.90	5.61	14.70	5.13	9.58
Oils and fats	9.51	10.37	11.27	10.10	5.51	2.04	14.32	5.25
Dairy	3.29	4.12	3.77	4.87	15.07	14.16	2.67	8.55
Grain milling	7.42	7.88	9.62	9.89	6.36	7.89	14.17	6.69
Starch	0.60	0.64	0.68	0.70	0.90	12.66	13.83	6.93
Animal feed	0.58	0.77	0.89	0.87	10.80	2.58	4.00	9.74
Bakery	1.47	1.78	2.05	2.16	8.31	8.63	21.87	8.69
Sugar	19.54	20.60	18.14	17.98	8.43	5.26	-3.35	3.13
Confectionary	0.21	0.28	0.28	0.36	19.15	10.04	15.31	16.32
Food Industry	86.80	97.95	105.33	103.19	7.92	2.11	10.02	4.18

A study was undertaken mainly to assess the status of mango processing plants and economic feasibility of mango processing plants in the district. The study revealed during the decade of 1980s and 2000 that there was spurt in growth of mango processing firms. Out of the total number of firms about 72 per cent are small scale firms and remaining 28 per cent constitutes medium and large firms. The quantity of mango fruits exported from Chittoor has recorded more than 2 fold increase, whereas, in terms of value it has shown an increase of only 10 per cent. On the other hand the mango pulp export has recorded more than 7 fold increase both in quantity and value terms. The analysis of firm level primary data revealed large variation in investment, ranging from 2 crores (small firms) to 15 crores (large firms) across firm size groups. The ratio analysis revealed that the small firms are more efficient than the other size class of firms. The capital ratios revealed that the large firms are more efficient than the small firms. All the three class of firms are feasible as per both NPV and BC ratio criteria.

Seed is considered as vehicle of technical change. Efficient seed management and its distribution is important to achieve and sustain high productivity of crops. This study examined paddy and wheat seed delivery system and seed management in in two selected districts Karnal and Sonipat of Haryana. The study revealed higher yield of certified seeds over farm saved seeds and availability of appropriate choices for the varieties in the market were the major facilitating factors for higher seed replacement rate. Determinants of farmer decisions for awareness and adoption was analysed using Logit model, and results found that distance from market, farm extension visit, vicinity of model village and level of education were the major determinants for leading the farmers to purchase the seed annually or use their owned seed. The public sector is performing a wide range of roles from the development of a modern variety to the final distribution of seeds to the farmers.

Fertiliser is another critical input for agriculture growth along with seed. While Government of India's interventions and policy support have ensured wide and timely availability of fertilisers at affordable prices to farmers, higher consumption and incentives to the fertiliser industry, certain problems have emerged over time in the form of indiscriminate and imbalanced use of fertilisers, significant regional variability in consumption, and high burden of subsidies. Analysis of fertiliser production capacity in the country revealed that during

the eighties there was a rapid increase in production capacity of nitrogenous and phosphatic fertilisers. Consequently, production of N and P_2O_5 increased rapidly and growth in production was found to be highest during the decade 1981-82 to 1990-91. The production growth was found to be generally higher in case of nitrogenous fertilisers, indicating the tendency of farmers to use large quantities of nitrogenous fertilisers, chiefly urea. In the nineties, efforts were made to induce balanced use of fertilisers among cultivators and during this period growth in consumption of phosphatic fertilisers was highest. A higher degree of inter-year variability in consumption of P_2O_5 and K_2O was observed as compared to N. Inter-regional and inter-state comparisons revealed that total per hectare NPK consumption was highest for the Southern region followed by the Northern region. The same pattern was observed for phosphatic and potassic fertilisers. Across different size categories of farms, per hectare consumption was highest in case of marginal farmers which nullifies the commonly held belief that large farmers use more fertilisers. To ensure wide availability of fertilizers, distribution has been managed through predetermined ECA allocations of different producers to different states under the Fertiliser Movement Control Order.

Investments and innovations are also important for agricultural growth. Patent plays a major role in attracting investments and innovations. A major change in the Indian patent scenario came in 2005, when product patenting was introduced in India. The study evaluated the impact of introduction of new patent regime in agriculture. It found that the patent grants have drastically increased in all the fields, including agriculture after the amendment of Patent Act in 2005. In agricultural science, the highest patents were granted in the field of biocides including pesticides, herbicides and fungicides (67 percent), followed by plant tissue culture and genetic engineering (5.71 percent). It was found that foreign sector participation is increasing in agricultural patenting and it accounts for 75 per cent of total patents granted in India during 2007-2012. Even though new patent regime increased availability of new technology particularly transgenic crops and various pesticidal compositions, there is a growing concern on the increasing cost of protected technologies with the increasing share of private sector in the agricultural research.

Agricultural production, Natural resources and Environment

Studies on economics of different agricultural production systems, natural resource use and associated environmental considerations are presented in this section.

Precision farming is environmental friendly technology that offers higher and quality outputs while reducing levels of resource use. A study was undertaken in Palakkad district of Kerala to assess the determinants and constraints in the adoption of precision farming and to compare input use, productivity, income and efficiency in precision *vis-à-vis* traditional farming in selected crops and market linkages. Logit regression results showed that farm size, level of education and attitude of farmers towards risk orientation were the factors which positively and significantly influence adoption of precision farm technologies. Although, the cost of banana cultivation was found to be higher on precision farms than that of traditional farms, precision farming was found to generate impressively higher returns in comparison to traditional farms. Under the current scheme of subsidy on the establishment of poly-houses, the farmers' investment in the same was found to be economically feasible as NPW, BC ratio and IRR were high.

Agricultural development has taken place at a very fast rate in Punjab State. The study was attempted to see the irrigation utilization pattern in the state of Punjab, its implications on the cropping pattern, falling groundwater table and its impact, water use efficiency in crop production at farmers' level. The source wise irrigated area in the state shows that tube well and wells are the major sources of irrigation during the recent decades. The studies on cropping pattern has shown that because of the profitability and availability of water at shallow depth, rice replaced other kharif crops like maize, ground nut and pulses in the entire state. The area under paddy increased from 7.22 per cent in 1965 to 32.92 per cent in 2005. Similarly the area under wheat cultivation increased from 37.12 per cent to 43.53 per cent in the same period. The mean irrigation efficiency was

57 and 65 per cent in case of paddy production, in the two irrigation systems under study. In wheat production, the mean irrigation efficiency was 61 and 68 per cent respectively in both the systems. It clearly indicates that there is a chance to improve the efficiency of irrigation by 39 and 32 per cent respectively in the sample farms to have technically efficient farms.

In the similar line, a study was conducted on groundwater development and its impact on the cropping intensity and crop productivity in Nalanda district of Bihar under different water market regimes. The growth of tube well irrigation in Bihar has been tremendous (3.59%) in the past twenty years. To assess the water use efficiency in wheat and paddy crops for different water market regimes, production function approach was employed. From the estimated results, it was found that buyers were under-utilizing water resources and water productivity was higher among water buyers in comparison to the other agents of water market. For paddy crop in Bihar, water was over utilized among all categories of players in water market except buyers. On an average, installation cost was higher among the large and medium size holdings since they used high capacity tube well to extract water. Pump sets emerged as the most important component, accounting for 51.1 per cent of total cost of the tube well system.

Groundwater use efficiency and pricing mechanism under different water market regimes in Central Plain Zone of Uttar Pradesh was also under taken. The water use efficiency for different water market regimes, production function approach was used and it was found that buyers and self-user + buyer were underutilizing water resource. On the other hand, self-user and self-user + seller were found to be over utilizing groundwater due to injudicious use of water resource. The estimates used to examine the price equilibrium, showed that out of several factors, water extracted by seller and gross irrigated area of buyer were key determinants of bargaining power and, thus price equilibrium in the study domain, signifying that assured irrigation to buyer category of farmers will help in ameliorating the price mis-match in the groundwater market regime.

Degrading resource base over a period necessitated to think over sustainability of resources and its' conservation. Resource conserving technologies possess such quality of preserving the precious resources, which are being promoted by many government agencies. Among various types of resource conserving technologies, zero tillage and Furrow Irrigated Raised Bed planting was selected, but the study remained confined to former due to very less adoption of later in Punjab and Haryana. The results indicated that the adopter farmers were more literate, operational holding was greater in comparison to non-adopters. Zero tillage improved many bio-physical attributes of soil. Among various factors, source of information, educational level of farmers, operation holding was found highest impact in adoption of zero tillage while, irrigation number adversely affected the adoption. Adopter farms efficiency was greater than the non-adopter farms. Per hectare cost of land preparation was Rs.2011 lower, while 292 kg/ha yield was higher together gave Rs.4670 additional return. Similarly, another study examined adoption of resource conservation strategies adopted by farmers of Haryana. Results indicated that zero tillage and laser levelling has been widely adopted by farmers. The net return in zero tillage (34,027 Rs/ha) was higher than the net return in conventional tillage (26,936 Rs/ha) in wheat cultivation in Haryana. It was observed that the availability of rotavator has greater influence in adoption of conservation agriculture in Haryana. However, high cost of machine coupled with its non-availability and service centres proved to be major constraining factor in adoption of the conservation agricultural practice.

Table: Extent of adoption of different conservation technologies in Haryana

(Area in ha)

S. No.	Year	Zero tillage	Laser levelling	DSR
1.	2009-10	647500	47700	750
2.	2010-11	392250	154649	3000
3.	2011-12	421500	278362	4500
4.	2012-13	482500	550000	7500

Optimal use of water resources and conservation of the same holds the key for future food security. Keeping this in mind, study was conducted to examine economic aspects of decentralized rainwater harvesting structures in two rainfed semi-arid districts namely Karauli and Tonk of Rajasthan for the agricultural year 2014-15. As compared the state government run programs, RKVY was found to give more impetus on growth and development of these structures. The adoption of farm ponds was observed to have significant influence on input and livestock possession, expansion and diversification of cropping pattern, farm income and ecological improvement. Also institutional variables like risk preference, subsidy, training and access to credit along with high farm income and crop diversification were found to significantly influence the adoption. Participation of female gender was found low in various stages of adoption. Seepage and cracking of bases, damage due to uneven heavy rains growing of weeds around structures and high initial cost were ranked as major constraints in adoption.

The concept of ecological economics is attaining greater attention in modern research field to study the interactions between economic systems and ecological systems. The humungous contribution made by the services and biodiversity of the ecosystems to societal welfare is being increasingly recognized in the ecological research. Focusing on these aspects, a study was conducted in one of the fragile ecosystems in India, which has been undergoing serious anthropogenic interventions undertaken for its sustainable development. Kuttanad Wetland Ecosystem of Kerala, which is one of the three "Globally Important Agricultural Heritage Systems (GIAHS)" sites from India was selected for the study. The study observed the degradation occurring in paddy wetlands of Kerala under which around 36000 ha were converted to non-agricultural uses in last three decades. An economic quantification of major selective proposed benefits of the Kuttanad package was done at various discount rates and the present value of benefits lie within the range of Rs.1797-2994 crores. As the Kuttanad package itself had proposed the provision of ration to fishermen BPL during the barrage closure period, an ecological compensation of Rs.21 per fisherman BPU year is proposed based on the results.

In areas where rainfall is erratic and irrigation water availability is limited, resource constrained farmer's livelihood depends on Livestock which is particularly true in case of Rajasthan state. Large part of the state is arid or semi-arid and fall under Thar Desert. A study was undertaken to examine sheep production system in Rajasthan, to examine equity, efficiency and profitability, to assess livelihood security of households and to identify the constraints to the migratory sheep production system. The results indicated that there was a decline in both populations as well as in wool production of the sheep. The temporary migration had declined but total migration had increased over the period. Non-availability of fodder and water due to recurrent draughts was the major factor for the decrease in total population and increase in the total migration. The labour was the major input cost in migratory sheep rearing and it declined with the increase in flock sizes. The share of family labour and hired labour was about two-thirds and more than one-tenth of total input costs, respectively. Large flock owners realized moderate net returns (Rs. 28646 per flock) and small flock owners realized poor net income (Rs. 6608 per flock). The migratory sheep production system generated significant employment opportunities to the flock owners and contributed a considerable share in total income.

Technological and institutional innovations in agriculture and their impact on farm economy

Impact assessment studies of agricultural technologies are important to quantify the benefits of technologies developed. These also forms the base for a sound agricultural policy. Impact assessments provides much needed feedback to technology developers on field performance of technologies and bottlenecks. In this line, division has undertaken impact assessment of various agriculture technologies as well as institutional innovations.

Indian Agricultural Research Institute has made significant strides in vegetable research. The study evaluated the economic impact of the adoption of carrot variety "PUSA RUDHIRA" developed by the Institute, in the Haryana state. It revealed that the total economic surplus accruing from investment in research was found Rs.101 crores. The major constraints in vegetable cultivation was observed to be non-availability and high cost of labour, followed by non-availability of seed, credit and lack of irrigation facility in the study region.

System of Rice intensification (SRI) is an environment well disposed & novel strategy of rice cultivation which incorporates utilizing less seeds, less water and fewer chemicals to obtain higher yield and profitability. A study was undertaken in Bihar to assess the adoption of SRI and its determinants, input use, profitability and constraints in practicing SRIin paddy. The results showed that family labour and number of training attended were the significant factors influencing the adoption of SRI in paddy. Farmers' perception indicated subsidy, higher yield and profit are the important determinants. The results clearly showed that SRI method of rice cultivation gives significantly higher yields in comparison to traditional method and is generating impressively higher returns though input cost is almost the same. Non- availability of skilled labour was found to be the major constraints to adoption of SRI in the study area

Protected cultivation increases productivity, production and quality of the produce and government is also incentivizing its spread by providing subsidy for establishing greenhouses through various programs. A present study was undertaken in Solan district of Himachal Pradesh in the year 2014-15 to find the linkages of various institutions involved in promotion of protected cultivation. Area under greenhouses is currently estimated nearly 27 thousand ha, and floriculture has immerged as a profitable venture in the states like Himachal Pradesh. The benefit-cost ratio, internal rate of return and pay-back period for export-oriented protected cultivation units were estimated 1.60, 117 per cent and 2years respectively. However, for domestic market oriented units, the same were estimated as 1.43, 73 per cent and 3 years, respectively. Protected cultivation has increased income of farmers by 65 per cent and small farmers were found to have spent most of their income on consumption while large farmers on non-farm investment.

Organic farming has shown the potential to provide environmental benefits besides improving the quality of food. A comparative analysis of cost, yield and net return was done among the beneficiaries and non-beneficiaries of organic farming in case of tomato farming in Nasik district of Maharashtra. The study revealed that the organic farming is mainly being adopted by the medium and large farmers. The analysis revealed that under organic farming approach, tomato cost of cultivation was estimated marginally higher (9 per cent), but showed a substantial increase in net return (28 per cent) as compared to the conventional farming. The number of livestock reared, level of education, premium price, market demand and technical guidance influenced the decision of the farmers to adopt organic farming practice. However, the non-availability of labour, high transportation cost, lack of proper technical guidance, problem of certification and non-availability of quality bio-inputs were found to be the major constraints in adoption of organic farming.

It is important to make technology development a demand driven process so that the technologies developed solves pertinent problems in ground situation. Keeping this in mind a study was conducted to know the farmer's varietal preference in case of rice in North Eastern Plain Zone of Uttar Pradesh state. In addition to this, the study explored the costs and benefits of improved rice varieties (tolerant to submergence conditions like Swarna Sub1 and Samba Masuri Sub 1) and other commonly grown rice varieties by the farmers in the study area. Swarna Sub1 varieties are preferred mainly because of their tolerance to submergence conditions prevailing in the area during the rainy season. Sarjoo- 52 is an old variety, but still preferred due to its taste, yield and resistance to bacterial leaf blight and bacterial leaf streak. The farmers growing improved varieties of rice realized a gross return higher as compared to non-adopter. The results also clearly showed that the average per ha yield of improved varieties on adopter farms were estimated higher than that of non-adopter farms. By comparing the means of yield and coefficient of variation of both the groups of varieties, the study clearly showed a significantly higher yield on adopter farms of improved varieties than that of non-adopter farms. The perceptions of the varietal traits, i.e. perceptions of submergence tolerance and yields were found significantly affecting adoption of improved rice varieties. This implies that those farmers who perceived improved varieties have high yield and tolerance to submergence traits are likely to adopt improved varieties more.

Apart from technological innovations, the adequate and timely delivery of inputs is a crucial factor affecting the profitability of farming. The TRIPTI (Targeted Rural Initiative for Poverty Termination and Infrastructure) programme of Orissa uses microfinance as an important means to alleviate poverty by improving the access to

credit, technology transfer and empowerment. The study revealed that the access to credit, education, contact with extension officials was high among the beneficiary households compared to non-beneficiary households. The income of beneficiary households was observed to be 51 per cent higher compared to the non-beneficiary households.

Kisan credit card scheme is an important institutional innovation of the banking sector for promoting agricultural credit and for achieving financial inclusion. Using both secondary and primary data, an attempt was made to analyse the progress and performance of the Kisan Credit Card scheme in India; evaluate the impact of the Scheme on crop productivity, income and employment with a specific focus on Krishnagiri district of Tamil Nadu; The scheme has shown positive growth rate for number of cards issued, and total and per account amounts advanced. The cost and return analysis showed that cost of cultivation and net income per hectare for three crops (paddy, sugarcane and groundnut) were higher for beneficiary farmers than that for the non-beneficiary farmers. It was also found that cost of borrowing credit was higher for non-beneficiary farmers. The constraints in the use of KCC perceived by the farmers for financial institutions was lengthy paper work, delay of payment of loan, and non-availability of loan on time, etc.

A study was conducted to evaluate institutional interventions in terms of schemes and credit to assess the impact of tissue culture banana technology in Gorakhpur district of Uttar Pradesh. The analysis suggested the key role played by various horticultural development schemes such as NHM, SHM, NHB and RKVY in strengthening the horticulture sector. Tissue culture banana cultivation incurred 8.3 per cent higher cost in comparison to sucker propagated one due to higher cost of planting material, fertilizer and labour use. Total cost is highest for semi-medium farmers followed by small farmers. The per unit yield for tissue culture banana is higher than its counterpart in terms of its bunch weight. Cobb–Douglas production function analysis indicated that planting material, human labour, plant protection chemicals, machine and bullock labour significantly affected the production of tissue culture banana, The net income of tissue culture banana adopters is about 17 per cent higher than the non-adopters.

Community action for agricultural development has attained focus of many Government policies. A study was conducted to examine the impact of microfinance mediated through Self Help Groups. It was found that, participation in SHGs is directly related to training received and household employment and inversely related to the farm size of the household. Training is the most important factor identified behind participation in SHGs Transaction cost found least in case of cooperative bank financed SHGs followed by NGO and commercial bank promoted SHGs. Microfinance activities were found instrumental in raising household income and employment. It was observed that age of the SHGs has positive impact on household incremental income. Study reported that average annual income and average annual employment of beneficiary household increased by 27% and 42% respectively. Fluctuation of income and employment is also observed less in case of beneficiary household than income and employment of the non beneficiary household. Effect of microfinance was found more intense in case of non farm activities.

The concept of Participatory Irrigation Management has been recognized all over the world as a tool for improving irrigation management along with sustainability of the system. In India, Water Users Associations (WUAs) are formed to take the responsibility of operation and maintenance of downstream parts of the irrigation system, distribution of water among water users and collection of water rates. A study was undertaken in Thanjavur district of Tamil Nadu to examine the structure and functioning of WUAs, farmers' participation in WUAs, and the determinants and constraints in the participation and to analyze the impact of WUAs on efficiency, equity and reliability in water use. Overall participation of the selected participating farmers was satisfactory. However, purely canal water user farmers have participated in more activities of WUA than canal plus tube well user farmers. The education of the head of the family and operational holding have positive influence and distance from the canal to farm has negative influence on the farmers' effective participation in WUAs. The participating farmers have applied significantly more number of canal irrigations and realized significantly higher yields after participation in WUAs in comparison to non-participating farmers. The

participating farmers have realized higher returns and are technically more efficient than non-participating farms. The irrigation efficiency was also more on participating farms in comparison to non-participating farms. The WUAs were able to address the major concerns of water distribution, timing of release of water and the proper maintenance of channels which reflected in the higher realization of yield and decreased water losses. Corrective measures should be directed to enhance the effective participation of all the stakeholders in WUAs and thereby minimizing the inefficiency and the inequity in water use.

Study on Kudumbashree was also undertaken which is envisaged as approach to poverty alleviation focusing primarily on micro finance and micro-enterprise development, collective farming through Joint Liability Groups (JLGs). Kudumbashree meaning "prosperity to the family", the poverty eradication mission of the state of Kerala is a community based self help initiative involving poor women below poverty line. The poor, especially in market economies need strength through collective action that can offer avenues for their economic, social and political advancement. Neighbourhood Groups (NHGs) in Kudumbashree Mission is a sustainable tool of entire development for the weaker section of society in Kerala if it's all stakeholders gain capability to pay desired attention towards their roles and potentials. The study revealed that the institutional structure of Kudumbashree was well-organized. The number of NHGs eligible for bank linkage programme was found to be very less. The marketing channels, practices and producer's share in consumer's rupee for JLGs closely resemble that of other farmers in the study area. JLGs displayed increase in rice production through higher yield resulting in higher net revenue per hectare. The sustainability of the group farming in Kudumbashree was reinforced as evident by the sound financial ratios. Improved say of women in decision making process in family and other cultural affairs was noted after Kudumbashree initiatives. The borrowing from non-institutional sources has reduced after formation of Kudumbashree groups.

Agricultural price, markets and value chains

Agricultural marketing, value chains for different commodities, export and import of agricultural commodities, prices and their behaviour is also one of the major thrust research are of the division. Summaries of studies conducted within this theme are documented here.

Modern retail is organisations are inducing changes in food markets and purchase behaviour, However, the traditional retail systems continue to hold an important place in the Indian consumers shopping habits. The effect of modern retail on food prices and a comparison between modern retails and traditional retail formats with respect to the pricing of selected commodities was undertaken in the city of Kochi. Significant price differences were found to exist between traditional and modern retail formats in all the commodities studied. The difference was most prominent in rice, attributable to the consumer preference for branded rice, mainly sold through modern retails. The quality of the commodity significantly affected price for potato, apple, rice and coconut oil. The part-worth value obtained from conjoint analysis for price less than traditional retails (1.029) suggest the importance of price competition to attract consumers. The level of excellent quality received low part-worth score (0.726) showing that consumers were willing to compromise on quality for the sake of lower price.

A study on economic analysis of consumer behaviour and impact of organised food retailing was conducted in five zones of National Capital Region (NCR) viz. National Capital Territory (NCI) of Delhi, Ghaziabad, Noida, Gurgaon and Faridabad. The study observed that, the level of education, monthly income of household, ownership of vehicle and working women in the household positively influenced consumer preference for organized food retails. Net returns realized by farm producers in vegetable cultivation were higher by selling them through organized retail consolidation centre. Education, area under vegetable cultivation and own transport facilities positively influenced the sale of vegetables to organized retail consolidation center. The study concludes that organised marketing channel is more efficient than traditional marketing channel as former involves higher producers' share in consumer rupee and lower price spread.

A study to decipher various dimensions of futures trading for their relevance in agricultural commodity markets in India was undertaken. While deciphering the extent and magnitude of the level of market integration, it was observed through co-integration technique that all selected commodities became stationary except in rice, as a prelude for co integration analysis. Notably it was observed that, there has been significant cointegration among the spot and futures price for all commodities except sugar. The spot and futures prices were found to behave in an expected manner that is a converging trend, except in sugar where diverging trend was witnessed. In order to have a smoother functioning of the futures market, the policy options suggested are: thorough review of erstwhile government policies and controls in the current context, staggered MSP policy-giving premium for the post season months till next season, will largely help in mitigating the price instability, Government move to ban cereals and pulses from futures trading does not stem from any empirical basis, as the study clearly sees the convergence in the spot and futures prices hence, the futures trading should be continued in cereals and pulses, the price risk management instruments disseminated amongst farmers and traders in order to mitigate price risks and reduce marketing margins. Hence these should be popularized for greater farmers' participation.

In recent times, horticulture has become the growth driver of India's agriculture with India's emergence as second largest producer of fruits and vegetables in the world after China. In Karnataka, modern retail chains were found to be more efficient with higher producer's share in consumers' rupee for vegetables. In case of arecanut, higher marketing efficiency and lowest price spread was found in marketing channel involving large wholesalers cum commission agents. Price behavior of onion, mango and arecanut indicated the influence of own market price compared to other markets. The study found that, strategic location and size of the market plays significant role in influencing prices in any market. The opinion survey on perceived impact of market reforms in Karnataka state shows farmers and traders are very optimistic about the positive impacts of reform measures like improved quality of produce, reduction in loss of perishables, enhancement of value addition process and creation of market infrastructure. The efficiency in marketing of flowers can be improved by implementing proper auctioning and infrastructure development.

In continuation, a study on changing growth pattern and composition of India's horticultural production and trade, trade competitiveness, and sanitary and phyto-sanitary issues impacting their exports was taken up. Despite being one of the largest producers of fruits and vegetables in the world, India's share in world exports is relatively insignificant. Horticultural products account for one third of the total value of agricultural exports. Among these, exports of spices lead followed by cashew, processed fruits and vegetables. Export of processed fruits and vegetables is growing impressively and has received priority attention too. The commodity-wise analysis showed that India's major importing partners for most horticultural produce are neighbouring countries. Further, the unit price realization from horticultural exports to these countries have been found generally much lower as compared to the European countries, USA and Japan. The study also indicated considerable impact of food safety standards stipulated by the USA and EU on India's horticultural exports. Spices and fresh and processed fruits and vegetables faced higher number of rejections of export consignments and notifications issued, mainly because having filth, pesticide residues, microbial contamination and noncompliance of other mandatory technical parameters.

Production and marketing of jute in West Bengal was studied and the results revealed that Jute acreage was found to be positively influenced by lagged price lagged yield and lagged acreage. The study also revealed the existence of considerable variation in prices received by various intermediaries between various agencies. Among the four types of marketing channels examined in the study, the farmers received higher price in the channel consists of Producers '! businessman '! Jute miller as compared to other channels. Therefore, through this channel mill owners were able to purchase jute from the farmers at remunerative price. Out of various constraints examined, physical infrastructure of market was found to be most important constraint followed by marketing mechanism, suitable agro-climatic situation and technology. Similar study on cotton production and marketing in Karnataka revealed decreasing trend in cotton area and production over the years. The study also revealed the existence of considerable variation in prices received by various intermediaries between various

agencies. Among the three types of marketing channels examined in the study, the farmers received higher price in the channel-I consists of Producer '! Ginning mill as compared to other channels. The price spread in channel-I i.e. Producer'! Ginning was highest, indicating higher marketing efficiency.

A study has been undertaken to examine the trend and seasonality in the prices, as well as extent of integration among selected onion markets. The relationship between arrivals and prices of onion were analyzed through linear regression model and the analysis indicated thatall markets in the study showed a negative relationship between arrivals and prices and positive relationship between price of the onion in a given period and its lagged price. Five channels of onion marketing were identified in Maharashtra. Maximum quantity (64.74%) of onion was passed through channel: Producers '! Wholesaler '! Retailer '! Consumer. Minimum quantity (1.04%) of onion was marketed through the channel: Producer '! Consumer but whose marketing efficiency is very high (83.33). High marketing margins were taken away by the market intermediaries. Lack of proper storage facilities was identified as major problem by the onion farmers. The co-integration analysis revealed that major onion markets in the country are integrated.

Production efficiency and price behaviour of sugar was examined in one of the studies. The study revealed that sugarcane productivity was positively and significantly influenced by the use of human labour, machine, fertilizers, insecticides and farm size. Although sugarcane productivity was found higher in Maharashtra but profitability was estimated higher in Uttar Pradesh. The study estimated that Indian sugar industry was operating at 60.5 per cent overall technical efficiency level in the year 2012-13, implying that 39.5 per cent of input use could be reduced without affecting sugar output. Higher Fair and Remunerative Prices and much higher state advised prices of sugarcane coupled with lower ex-mill sugar prices have resulted in declining profitability and huge cane arrears with sugar mills which in turn has delayed payment to sugarcane growers. The results showed that sugar prices within domestic markets were found to be co-integrated while there is not such transmission of prices from international market to domestic market. Sugar and sugarcane prices were also not found to be co-integrated.

A study on production and marketing of natural rubber in Kerala was conducted. Natural rubber cultivation in India has been traditionally concentrated in Kerala which accounts for 78% area and 90% of production. Natural rubber is one of the important commercial crops of Kerala and rubber plantations have profound influence in economic and social life of people. Around 40% of the area as well as 45% of production of rubber comes from three districts *ie*, Kottayam, Ernakulam and Pathanamthitta. Fluctuations in natural rubber market prices is a major concern for the small rubber producers since it would affect the income stability and has multifaceted effect on the area, productivity and hence the production of the crop. From the present study it is concluded that rubber price are showing considerable instability in both domestic and International price behaviour. It was found that the markets are having significant positive integration. Export competitiveness measured using Nominal Protection Coefficients (NPC) revealed that export of domestic rubber was not competitive during both peak and lean seasons.

A detailed analysis of the composition, diversity, growth and instability of the major agricultural commodities traded within Asia in the post-reforms period was carried out. The results suggested that India earned maximum export revenue from marine products. Oil meals and basmati rice were other major revenue yielding commodities. Among imports, edible oils dominated, amounting to more than 50 per cent of total agricultural imports from Asia. Pulses and cashew were other major imported commodities. Among the various selected commodities, notable growth in exports to Asia was observed for tea, coffee, rice (non-basmati), spices, cashew, meat and preparations and marine products. While fresh fruits and vegetables have grown moderately, the growth in exports of oil meals was insignificant. A country-wise examination of the scenario suggested that Bangladesh, Japan, Malaysia, Saudi Arabia, and UAE are the largest destinations of India's agricultural exports. Among the import sources, Indonesia, Malaysia and Myanmar were the major suppliers of agricultural commodities to India.

Since past two decades, the increasing share of world trade has been taking place through new kind of regional integration initiatives called regional and bilateral trade agreements which is commonly known as Regional Trade Agreements (RTAs). India has also joined the race with the signing of numerous RTAs among which, ASEAN India Free Trade Agreement (AIFTA) is the prominent one The study was carried out in wake of apprehensions of effect on the agricultural sector by the extensive tariff reduction commitment of India under the AIFTA. The study utilized a partial equilibrium analysis model, SMART (Software for Market Analysis and Restrictions on Trade) developed by the World Bank to simulate the impact of tariff reduction scenarios for India and ASEAN countries and to quantify the trade creation and trade diversion effect of FTA. India's agricultural trade intensity with ASEAN was found impressive with intensity values for the period above two reiterating it as a preferred trade partner. However, India's agricultural import intensity was higher than the export intensity as evident from the higher import share of ASEAN. The, trade intensity with less developed countries of ASEAN namely Brunei, Cambodia, Lao were found low during the study period. India has exhibited comparative advantage in cotton, tea, rice, oil meals, and tobacco but faces competition from ASEAN in coffee, spices, and marine products while, India has no comparative advantage in fresh fruit and vegetable export. SMART simulation results revealed that, ASEAN countries will gain higher market share in India due to extensive tariff reduction. India will not gain from ASEAN market from tariff reduction as they have lower base tariff imposed on Indian products which stresses the need to increase the competitiveness of agricultural commodities exported to ASEAN. The study also found that India has the trade potential with countries like Brunei, Lao, Philippines, Singapore, and Thailand. In order to improve the trade relations with ASEAN, trade facilitation, quality standards and infrastructure development should be given considerable importance.

A study was undertaken to analyse the spatial and temporal performance of India's oilseeds economy, examine the dynamics of edible oils imports and analyse the volatility in prices of edible oils. Decadal analysis of the performance of oilseeds sector showed that the last three decades since 1980 witnessed high growth rate in total oilseed production and productivity, while area registered a high compound rate of annual growth during 1980-90 and 2000-10. Only soybean has shown substantially high rates of growth in area, production and productivity during the period. Imports of edible oils have increased substantially after the establishment of the WTO. Palm oil and soybean oil constitute a major part of edible oil imports. High Hirschman Index values in most years indicated that India's imports of edible oils are confined to a few countries. In recent years, Argentina, Brazil, and USA have together accounted for around 90 per cent of India's soybean oil imports. Over 95 per cent of palm oil imports are being made from Indonesia and Malaysia. The volatility and the tendency for volatility to persist for a long time was found to be high for groundnut in Hyderabad and mustard in Delhi, and for groundnut oil in Rajkot and Chennai.

Food safety addresses agents and procedures along the entire food chain, from production and processing to marketing and up to the consumer level. All the members of WTO have set up their own standards to reduce the risk intrinsic in the global trading and also to get access to the markets of importing countries. Considering this, economics of compliance with the sanitary and phytosanitary measures in the supply chain of black pepper in Kerala was studied. At the international level, CAC is the organization that controls and coordinates the setting up of global food safety rules. USFDA fixes standards for black pepper to be sold in USA in consultation with the ASTA. Indian food safety standards on physical, microbiological and chemical parameters revealed some gaps which need to be reviewed. As per a Government of India report, the total magnitude of the transaction cost is about 10 per cent of our export value. The cost of compliance with the quality standards incurred by the Indian black pepper exporters in US, EU and IPC markets were Rs 0.44 per kg, Rs 0.66 per kg and Rs 0.24 per kg, respectively. Export of black pepper to EU market incurs more cost compared to other countries. To meet the raised Codex standards, SPS measures enforced by the importing countries led to incurring of high costs by the exporting countries .The dimensions at farmer's level viz. knowledge about GAP practices and financial infrastructure; at processor's level viz. business strategy and skill and knowledge of processing; and at exporter's level codex knowledge were found influencing SPS compliance in the supply chain.

Production dynamics, Yield gaps and market integration

The study analysed yield gap, productivity trends and sources of productivity growth in major cereal crops of India during the period 1990-91 to 2011-12. Malmiquist index was used to examine total factor productivity growth and sources of productivity growth were decomposed by data envelopment analysis. The results further showed that yield gap in major states like Uttar Pradesh and Punjab in case of wheat and Karnataka and Rajasthan in case of maize have increased considerably in 2000 decade showing serious concern over the extension management and technology dissemination. Maize showed increase in area, yield and production since 2000 due to its increasing demand for industrial usage. In case of bajra, area has declined but production and productivity has seen significant growth during the period. Total Factor Productivity showed decreasing trends in the sub period (2001-11), mainly due to stagnation in efficiency and technology in rice and negative technical growth in case of wheat. The study points out the significant intervention towards the factors that could influence the efficiency along with the factors that result in technical progress especially in case of rice and wheat which are the mainstay of India's food security. It also calls for an effective extension management and technology dissemination strategy to reduce the existing yield gaps.

Pulses are important for ensuring food and nutritional security of the poor households. The stagnation in production and acreage coupled with the widening revenue gap between pulses and its competing crops was observed to be one of the major factors responsible for poor performance of pulses in the country. A study was conducted to estimate the yield gap and extent of integration in Pulse markets. The co-integration analysis of major pulses markets in India indicated that the markets are not perfectly integrated revealing inefficiency in the pulse marketing system. The adoption of different varieties by the Chickpea farmers revealed that the different biological traits along with price emerged as important factors influencing adoption. The study found that Kabuli chickpea variety fetched 31 per cent higher yield over the desi variety. The non-price factors like rainfall have greater influence in farmers' decision with respect to allocation of area under the crop. The yield gap-II was observed to be higher revealing the necessity of effective transfer of technology to enhance the yield and production of pulses in the country.

Climate change and adoption strategies

Agriculture is a source of climate change and a sufferer of adverse effects of it. The divisional research studies on areas of climate change, its perception by farmers and their adoption strategies are documented. Risk management strategies adopted by soybean farmers of Madhya Pradesh in the context of climate change is studied. Soybean is an important oilseed crop cultivated in India, under rainfed conditions. The study was undertaken to analyze the risk in soybean cultivation, risk management strategies adopted by the farmers in cultivation of the crop, and the factors influencing the adoption decision using primary and secondary data. The mean income and variance decomposition analysis showed the positive role of yield in raising the revenue and the larger role of price changes in the revenue variance. The major risks were related to rainfall, drought and biotic factors like diseases and pest. The major adaptation strategies were intercropping, crop insurance, micro-irrigation and varietal diversification. The study revealed the role of risk perception, membership in farmers' organizations, soil quality, irrigation, and non-farm income in adoption of these management practices. The study has revealed large level of economic benefit out of cultivation of improved varieties of soybean (JS 93-05, JS 95-60 and NRC-7). The potential mean annual benefit (economic surplus) was estimated Rs 3161 million, Rs 6291 million and Rs 685 million, respectively for the above varieties. In a drought like situation, the potential reduction in total benefit was much lower for NRC-7 compared to other varieties, pointing to the income stabilizing role of drought resistant varieties.

A study was undertaken to assess the hedging effectiveness of weather based crop insurance in the light of climate change in the state of Tamil Nadu. Vulnerability mapping shows that the districts of North Arcot, Coimbatore, Madurai and Thirunelveli fall under vulnerable category while Tiruchirapalli is found to be most

vulnerable. The farmers in Villupuram and Virudhunagar districts have experienced climate change in terms of decreasing quantity of rainfall, delayed onset of monsoon, increasing temperature, erratic rainfall pattern. The weather based crop insurance product of maize conforms to the principle of no-loss-no-profit to both the insurer and the insured. But in future perspective the product will either benefit the insurer or the insured, but not both simultaneously. On the other hand, the historical payout analysis of weather based crop insurance product of paddy has shown the benefit of insuring is marginally more to the farmers. But in the near future, the benefits of insurance will shift to the insurer at the expense of poor farmers. This calls for redesigning of the crop insurance products by taking into consideration the climate change perspective of future.

Mitigation of Green House Gas (GHG) emissions would help to reduce, delay and avoid impacts, but according to the Assessment Report of the Intergovernmental Panel on Climate Change (IPCC), current mitigation efforts are insufficient, and will result in increased emissions rates and continued warming. The Clean Development Mechanism (CDM) of the Kyoto Protocol offers an opportunity for the country to tap the global market, and to harness associated investment and technology flows. In this light, feasibility of afforestation/ reforestation projects in Haryana, under the ambit of CDM was studied. The feasibility analysis of the project using the discounted measures like NPW, B-C Ratio, and IRR revealed that the project is feasible. It is therefore suggested that such programme need to be promoted so that the small landholders could gain by way of carbon finance. The prominent constraint faced by the farmers is long gestation period, interval of carbon payments are widely dispersed throughout the project period and insecurity of land ownership. It was also revealed that the off-farm income emerged to be dominant factor influencing the participation of the farmers. It is therefore, suggested that efforts need to be made to create awareness and capacity building of the stakeholders so that more and more such programme are framed and implemented.

Acknowledgement

I wish to thank all the Faculty Members of the Division of Agricultural Economics for extending support and cooperation in preparing the research contributions of M.Sc. and Ph.D. students made during the period of 2007-16.

References

M.Sc. and Ph.D. theses submitted by the students in the Division of Agricultural Economics, ICAR-Indian Agricultural Research Institute, New Delhi 110012 during the period of 2007-2016.

Agricultural Extension

R.N. Padaria

Introduction

Established in October 1960, The Division of Agricultural Extension has made significant stride in development of effective paradigm and prototype for research, relevant curriculum and methods of teaching and training, models and framework for information and technology dissemination as well as service provision and generation of successful cases and methodologies of agricultural and rural transformation. For last fifty six years with its vision, action and notable attainments, it has been carrying out a flagship role for extension profession and its centres of excellence in national agricultural research system. Development of innovative extension models, linking farmers to markets and entrepreneurship development, gender empowerment, livelihood and nutritional security, capacity building and rural innovations are the thrust areas of research.

The Division continues to strive for the growth of extension profession with sustained research and quest for advancement in extension education for sustainable livelihoods with following mandates.

- To provide leadership role in carrying out basic and applied research in agricultural extension leading to sustainable development.
- To serve as a centre for academic excellence in the area of post-graduate education in Agricultural Extension
- To provide leadership role in agricultural extension training through integration of experiences, new concepts and models to meet the challenge of globalization .
- To organize seminars, conferences and workshops to integrate experiences of extension professionals resulting in models and strategies for agricultural development.
- To provide advisory and consultancy services in carrying out extension activities for agricultural development.

Significant Student's Research Achievements

The researches carried out by the postgraduate students in the able guidance and mentorship of the faculty as well as the professors of the Division have made significant contributions in advancement of the discipline. The research contributions of the students have been presented theme-wise as below.

Application of Information and Communication Technology (ICT) in Extension

Analysis of Community Radio Station "*Namma Dhwani*" on people's socio-economic and cultural development in Budikote, Karnataka showed that there was participation of 80 per cent of the respondents in the define phase of participatory message development and communication, 50 per cent in design phase, and only 25 per cent in produce phase because of the technicality involved in the message development; while 65 per cent of the respondents participated in the evaluate phase. The different levels of participation indicated that there was high level of participation in the first and the last phase of the message development, medium level of participation in the second phase and low in the third phase of the message development process. The

message effectiveness of the message developed was found to be high among 65 per cent of the respondents. Among the most important problem identified were power break down, internet connectivity, lying down of cable problem, lack of IT literacy, production of need based problem inefficiency in service delivery, hardware and software problem, low efficiency of community resource persons, lack of incentives and more service charges. It led to development of leadership as well as health, education, food habit and family system of the respondents (Rakesh, 2008).

An e-Learning behavior index (ELB Index) was developed and it was found that 68.3 per cent of e-Learners had low ELB Index. The determinants of e-Learning behavior of farmers of Kerala were identified as the years of education and years of experience with computer. It was discovered that, most of the farmers belonged to the category of low e-Learning behavior (68.3 per cent) as the extent of sharing of information through various source was found to be very low. The reason behind this result might be e-Learning is an emerging technology and so it may take time to convince the farming community about the advantage of e-Learning; adoption of any technology is possible only after crossing a well awareness phase. Change in decision quality, gain in knowledge and extent of technology adoption were found to be significant to differentiate between groups of farm women of Kerala using e-learning with groups using traditional learning (Balakrishnan, 2012).

Participatory videos produced and projected by Digital Green showed contextualised content and had great relevance to small and marginal farmers. The videos produced by Digital Green caused significant difference in production and income for all the farmers who had adopted them. The initiatives like Digital Green can be replicated in other parts of country for dissemination of farm technology information and it can be complementary to the conventional extension services (Afroz, 2013).

Measured on a scale of 1 to 5, the overall score obtained in range of 3.62 to 3.78 on perceived usefulness, effectiveness and perceived utility of contents amply showed that the respondents perceived e-learning module on "Climate Change and its Impacts on Indian Agriculture" as an effective tool to improve the knowledge and understanding of extension personnel on subject. E-learning modules can help to learn new concepts, create interest, serve as ready reference material and be a tool for distance education of extension personnel (Bhaurao, 2013).

A majority of the ICT beneficiaries (45.55%) were found to have more favourable attitude towards ICT based extension, while a majority of the ICT users (84.44%) were found to have low level of e-readiness. Mass media exposure, innovativeness, risk orientation and self-confidence were significantly associated with the e-readiness level of the community radio beneficiaries, while mass media exposure, innovativeness, economic motivation, risk orientation, self-confidence and information networking were found to be significantly associated with the e-readiness level of the mobile advisory beneficiaries. The study identified and standardized six indicators namely e-skill, e-awareness, e-ownership, e-willingness, e-frequency of use and e-accessibility to measure the composite e-readiness level of the farmers. Karnataka showed higher e-readiness index value (0.344) compared to Andhra Pradesh (0.331) and Madhya Pradesh (0.323), while Mandya district of the Karnataka had highest e-readiness index value (0.368) compared to other selected districts. The composite e-readiness index of three states in together was found to be 0.332. Adoption of ICT based extension services resulted in increased cost of cultivation but the proportionate increase in the farm income was higher than cost of cultivation. Adoption of community radio services lead to the additional farm income of Rs.9, 967 per anum, while the same was Rs.12, 680 per anum for the mobile advisory beneficiaries. Additional net income of Rs.2713 was achieved by the mobile advisory users compared to the community radio listeners. Lack of training, poor infrastructural development and poor network connectivity were the major barriers of adoption of ICT based extension services. Easy to adopt nature of the advisories, up to date nature of the ICT based extension services, requirement of no expertise, quality content of the services, timeliness of the information and relevancy of the information through ICT based extension were the major drivers for adoption of the ICT based extension services which could act as success determinants of knowledge transfer (Naik, 2014).

A majority of the farmers perceived that mobile Based Agro-Advisory services of TCS called mKRISHI was highly effective in terms of timeliness of information, quality of information, utility of information, satisfaction of farmers and ease of understanding of the information. Information provided by mKRISHI helped in increase in yield, market price and increased farm income. Despite of high level satisfaction, there were some constraints which caused hindrance in the smooth running of mKRISHI. Technological constraints were the most severe in mKRISHI information delivering and followed by economic constraints (Singh, 2014).

The farmers had moderate level of knowledge on Good Agricultural Practices (GAP) for grapes with a mean score of 58.9 per cent. However, majority of the GAP practicing farmers possessed high level of knowledge whereas most of the non-GAP practicing farmers possessed low to medium level of knowledge. There was significant difference in knowledge level between GAP practicing and non-GAP practicing farmers. However, no significant difference was found between Gap practicing and non-GAP practicing farmers in their attitude towards GAP. Assessment of information need revealed that farmers lacked information mostly on post-harvest practices & safety measures and export procedure. Highest information need of the farmers was found for information on certifying agencies and potential economic benefit of GAP, use of chemicals, and sources of credit. Based on the above findings, an e-learning module on GAP for grapes was designed with the help of the software Course Lab 2.7 and was validated by the respondents (Figure-1). The result revealed general acceptability of the module by the users. Overall mean of 4.09 indicated high preference of the farmers for the module. Content & design of the module (mean score 4.14) ranked first based on farmers' opinion while ease of understanding (mean score 4.13), utility (mean score 4.17) and ability to satisfy felt needs (mean score 4.02) ranked second, third and fourth, respectively. Consistent result was found in case of other stakeholders as well with overall mean of 3.91. The opinion of the stakeholders revealed that the module created interest on GAP (mean score 3.96), could be used by extension personnel to disseminate information on GAP (mean score 4.11) and could be used as educational instrument for training on GAP (mean score 3.13). The designed e-learning module is available in CD-ROM format and also at IARI website, URL: http://www.iari.res.in/e-learning_on_GAP_for_grapes/autorun.html (Som, 2015).

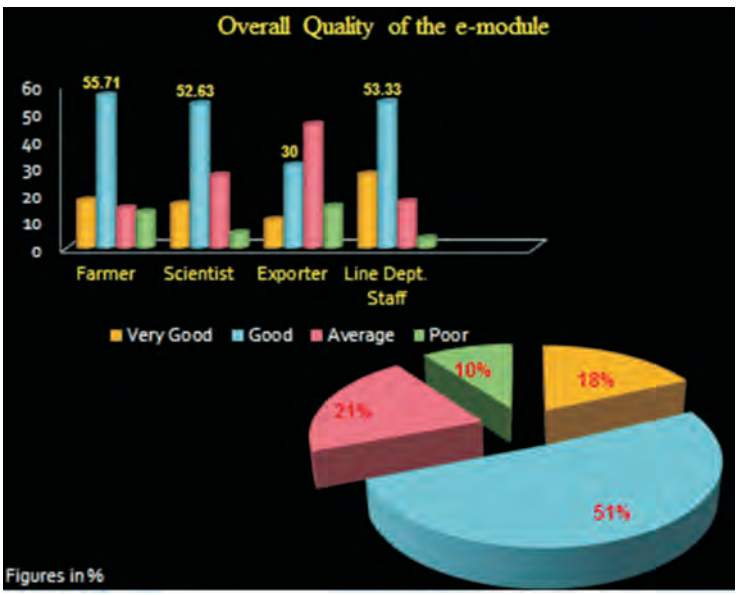


Fig. 1: Validation of e-learning module

The assessment revealed as much as 93 percent farmers had very low level of awareness about WTO. A vast majority of the agricultural scientists (66.67 %) had either very low level or low level of knowledge about issues of WTO while only 10 per cent of scientists had high to very high level of knowledge. A total of 80 per cent of extension personnel had low to very low level of knowledge on WTO issues. Only 3.33 per cent of them had high level of knowledge. The English e-module was designed and developed by using e-learning authoring tool named 'Adobe Captivate 7' (version 7.0.1.237) and the Marathi module was designed using Microsoft Power Point (2010). Thus a total of 56.51per cent knowledge gain was noted after the respondents were exposed to English e-module. Highest knowledge gain of 393.75 per cent was noticed in WTO dimension of "Structure and functions of WTO". Overall 320.51 per cent knowledge gain was observed after exposure to Marathi e-module. In dimension "Evolution and objectives of WTO" knowledge gain of 233.33 per cent was reported. Similarly, for dimensions "Structure and functions of WTO", "Tariffs and quantitative restrictions", "Agreement on Agriculture issues" and "Intellectual property rights issues" observed knowledge gain was 247.83

per cent, 178.13 per cent, 783.33 per cent and 452.17 per cent, respectively. There was significant difference between pre-test and post-test scores of the respondents on all the aspects of WTO for both English and Marathi modules. Thus gain in knowledge was attributed to the learning that has occurred due to exposure to the e-learning modules (Murai, 2015).

Agricultural programmes were the most preferred of all the programmes in Community Radio (CR). The education level and perception about the CR influenced the extent of participation by the respondents in production of programmes. Operational, financial and social sustainability were found to be components of overall sustainability of CR.Specific programmes and action based research are required to highlight the importance of community radio in remote places for the villagers for information about the outside world (Krishna, 2016).

Critical analysis of knowledge management in agriculture under Rice Knowledge Management Portal (RKMP) showed that the farmers needed information on disease management, while the scientists and extension personnel needed information on input and hybrid varieties of rice. Input dealers were found to be major source of information for the farmers. In case of scientists, it was availability of ICT tools and access to internet at office place, whereas in case of extension personnel one additional factor i.e. training in handling ICT tools were influencing factors in using RKMP. Among all the constraints, technological constraint was found to be most limiting factor for farmers, scientists and extension personnel as well. Among the technological constraints, lack of updated information was found most limiting factor. Under social constraints, lack of access to women farmers and lack of institutional supports were most limiting factors for the farmers (Sunil, 2016).

Most of the rural women (80 %) used regularly family as information source followed by Friends/neighbours (62.20 %), television (58.33%), NGOs (45.83), and radio (29.17%). The family members were perceived highly credible source of information by 80 per cent of the rural women, followed by friend/neighbours (73.33%), television (64.16%), and radio (62.50%). NGOs were perceived highly credible source of information by 31.40 per cent women. Twenty per cent of the rural women considered newspaper as highly credible. About 23 per cent of women found computer as high credible information source. Input dealer was perceived by 27.50 per cent respondents as high credible source of information. Personal, technological, cultural and infrastructural barriers were identified as major barriers in adoption and use of ICT among rural women. Technological barriers were perceived as to be the most important barrier followed by cultural barrier among the above barriers (Verma, 2016).

Extension Management and Approaches

Implementation of Agricultural Technology Management Agency (ATMA) ensured wheat farmers in Ahmednagar district of Maharashtra higher per cent of increase in yield per hectare; high returns; and high income. The wheat yield was significantly different between ATMA and non- ATMA farmers. Sugarcane farmers in Ahmednagar district had very low per cent of increase of yield per hectare; very low returns; high income; and medium effectiveness. Sugarcane yield and income were significantly different between ATMA and non-ATMA farmers. Maize farmers in Dahod district had very low per cent of increase of yield per hectare; very low returns; very low income; and low effectiveness. Maize yield and income were significantly different between ATMA and non-ATMA farmers. A majority of the ATMA staff expressed that the farmers had very low level of participation in decision making at various levels of implementation of ATMA. Resource, relations and performance; necessity and adaptability; values; information; and infrastructure were the factors of coordination. Performance of the FIGs in Ahmednagar and Dahod districts was medium. Positive and significant correlation was found among participation, coordination and performance. Participation and coordination contributed significantly for the variation in performance (Lenin, 2009).

Among the motivational factors promoting formation of Rythu Mitra Group (RMG)as perceived by the respondents, inspiration by the friends and relatives was most important one followed by their financial problems,

influence of extension agents and subsidies and other financial benefits. RMG members suggested for reducing the interest rate, increase of loan amount and quick release of loan; while, extension workers suggested for quick release of loan, increase the loan amount and betterment of marketing facilities for improving the performance of RMGs (Manti, 2009).

Deendayal Research Institute (DRI), an NGO established by Shri. Nanaji Deshmukh in 1969 has developed an innovative mechanism for upliftment of the poor and underprivileged in Bundelkhand region. Its Village Self Reliance Campaign (VSRC) successfully used *Samaj Shilpi Dampatis*, a newly wed graduate couples who live and work with villagers for a period of five years, to act as nodal point for all rural development interventions by DRI.The study found that all SSDs possessed a very high level of achievement motivation, organizational commitment, dedication, faith in people, empowerment, and development orientation. The scheme had a great impact on beneficiaries in their raise in income, savings, and increase in employment days. A majority of the respondents, especially marginal and small farmers, adopted either 1.5 or 2.5 acre model that helped ensure food security for their families throughout the year. Adoption of kitchen gardens in their homesteads helped secure nutritional security for the family. Creating social capital had paramount effect in successful mobilization of villagers and sustainability of the scheme even after SSDs left the village. Performance of success varied from 50 to 82 per cent in case of making villages dispute free. The SSD scheme was found to be cost-effective, efficient and worked for the integrated development of villages. This indigenous concept of rural development needs to be considered for upscaling in other parts of the country. The model holds a great hope for government departments, NGOs and voluntary organizations working at grassroots level(Manjunath, 2009).

The study on effectiveness of research-extension and farmers' linkage in agricultural universities showed that the most common place for implementing research work was campus-farm field in India (65%) unlike Non-Govts fields (45%) in Ethiopia. The major constraints for linkage among scientists and farmers in India were lack of proper policy, inadequate fund and administration problem where as in Ethiopia it was work overload of scientists. Action research consisting of suitable interventions to improve linkage may be undertaken. Effectiveness of linkage in agriculture in agriculturally developed state and less developed state should be studied (Helen, 2009).

The effectiveness index score of agri-clinics was high among the beneficiary farmers. About 50percent of the respondents perceived that the paid extension services were moderately effective, while the rest perceived them to be highly effective. Among the correlates; family ties, age, education , and farm size, were positively and significantly associated with effectiveness index score**.** Among the four components i.e. extent of adoption, percent increase in yield, percent increase in profit and farmers' satisfaction, the contribution of extent of adoption was very high in farmer's perceived effectiveness of paid extension services provided by agri-clinic entrepreneurs (Chandrashekhar, 2010).

Rural extension sub-centres with private professionals which provided artificial insemination services and other inputs to farmers who were unreached by formal institutions was found as a successful alternative extension model evolved by *Mitra Niketan*in, Kerala. NGOs can be encouraged to work with Govt. agencies through public-private partnership for ensuring socio economic growth in rural areas. Rural extension sub-centre approach can be scaled by other agencies working for agriculture and rural development (Rejula, 2011).

A study on the effectiveness of Strategic Research and Extension Plan of Agricultural Technology Management Agency in Bihar revealed that the extent of participation of stakeholder was found to be 52.77 per cent in formulation stage and 47.05 per cent in implementation stage of SREP. Crop diversification increased by 17.72 per cent indicating replacement of area under cereals by high value vegetables and oilseeds. The yield of major crops of the project area and the income of the beneficiary farmers also increased significantly. The assessment of effectiveness of training programmes and Research-Extension-Farmer Linkage indicated medium to low level of effectiveness. The overall effectiveness of SREP was found to be only 42.88 per cent

which implies that SREP-ATMA was instrumental in improving socio-economic status of farming community to medium extent only (Dhiraj, 2011).

The level of effectiveness in supply of proper inputs by the private extension support system was medium to high, while for public extension support system, it was medium to low for promoting orange production in Maharashtra. This might be due to the profit motive of private extension support system. The level of effectiveness in provision of advisory services by the private extension support system was reported medium by 41% followed by high (27%) and low (19.5%). The mean effectiveness score was 0.62. The level of effectiveness in provision of advisory services by public extension support system was found medium for 32.5 per cent of the orange growers followed by low (32%) and high (25.5%) level of effectiveness. The t-test clearly revealed that there was significant difference in provision of advisory services by private and public extension support system ($P<0.05$). The level of effectiveness with respect to the timeliness of services was found medium by 39.5 % followed by high (36.5 %) and low (17 %) for the private extension support system. The mean effectiveness score obtained was 0.633.In case of timeliness of services provided found by the public extension support system, it was found that the level of effectiveness was medium for 39 per cent of the respondents followed by low (35%) and high (17%). The mean effectiveness score obtained was 0.514. There was significant difference between the timeliness in provision of services by private and public extension support system ($P<0.05$).The private extension support system had better timeliness in delivery of services as compared to the public extension support system. This might be due to the fact that public extension personnel had huge amount of work and lack of transportation facilities. They had to serve a large number of clients who had different demand for technologies. The 't' test showed that there was no significant difference between the level of effectiveness in fairness and competency of private and public extension support system. This might be due to the poor salaries of government extension personnel and lack of monetary rewards for their work (Gedam, 2012).

An analysis of private extension system was undertaken with a case of Tata Kisan Sansar, the invention of Tata Chemicals, which is working through innovative model called "hub and spoke model". It was found that the private initiative (TKS) provided inputs, services, which was better in accessibility, quality and timeliness to the farmers. The constant advisory support in addition lead to better adoption of technologies which was further leading to increase in yield and income and ultimately satisfaction of the farmers. The six variables *viz.* marketing orientation, education, economic motivation, social participation, family size and extension agency contact was found to influence 74.00 per cent in farmer's membership in TKS. By analysing the constraints through Garrett's ranking it was found that in organisational level job security and in functional level lack of co-operation from existing ground-level organisation got first rank. There is duplication of efforts with multiplicity of agents attending extension work without convergence. As a result, the efficiency is not up to the mark. Therefore, a coordinated attempt to synergise and converge these efforts at the district and below level is required to improve the performance of various stakeholders (Anirban, 2012).

Farmers Field School (FFS) had positive impact. The increase in the yield of paddy among FFS participants was 22.42% whereas it was 11.36% in case of Non- FFS farmers. The average per hectare gross income obtained by FFS farmers during post FFS period was Rs. 29131 which was Rs. 7000 higher than the income (Rs. 21847) gained by non-FFS farmers (Manoj, 2013).

A paradigm shift to the private and voluntary organizations led extension is being witnessed as the public agricultural extension system has been adversely affected by various constraints. ICAR-Indian Agricultural Research Institute (IARI), a premier agricultural research institution took lead to develop linkages with voluntary organizations (VOs) of repute for speedy transfer of agricultural technologies to the ultimate users. Analysis of the effectiveness of IARI linkage with VO namely Young Farmers' Association (YFA) of Punjab state. The advisory services through IARI-Young Farmers' Association (YFA) of Punjab state linkage, higher yield of IARI released varieties; social participation of farmers and their education were the major determinants for the adoption of IARI varieties among the farmers of Patiala district. In case of both rice and wheat, IARI varieties were sown in more than 60% area of the total land holdings of the farmers. Although, the linkage

was perceived highly effective in terms of the delivery of farm technologies, advisory services, farm literature supplied, farmers' fair, front line demonstrations, impact on yield and income of farmers and satisfaction of farmers but non-availability of seeds in desired quantity, higher seed cost and less land for seed multiplication were major inhibiting factors. The study revealed that among the three major stakeholders in delivery of seed, the YFA was found the most effective with the mean percentage score of 73.72 and the farmers were satisfied with the quality as well cost of the seed supplied through YFA. Majority of the farmers reported that the cost of the seed supplied by the private agencies having percentage mean score of 70.66 was very high but were satisfied with the quality of seeds supplied. Farmers responded favourably with the overall seed supply system of state government with average percentage mean score of 65.66 but were less satisfied with the quality of the seed supplied by them. The functional efficacy of linkage may be enhanced through close collaboration of state level government and private agencies for broad based, inclusive and sustainable agricultural development (Gajbhiye, 2013 and Gajbhiye, *et.al.* 2015).

Analysis of functional mechanism and impact assessment of IARI-Post office Linkage Extension Model (Fig. 2) where-in the branch post masters were trained to play the role of community based extension agent for accessing quality seeds and empower fellow farmers and KVK scientists to act as resource partner of IARI by providing technology backstopping to the BPMs and farmers; revealed that half of the beneficiary farmers had highly favourable attitude towards the model due to "high quality seeds", "source credibility" and nearly one-third of the respondents had un-favourable attitude as they did not receive quality seeds. A majority of rice, wheat and mustard farmers were found to have received seeds in time. The economic viability of model was high in comparison to conventional method of seeds delivery. A majority of farmers were satisfied with quality of technology (Mean score=3.21) followed by timeliness (Mean score=2.96), quantity (Mean score=2.29) and technology backstopping (Mean score=1.53). Biasness of Branch Post Master in selection of beneficiaries was the major constraint faced by farmers. Lack of incentive (Mean rank=1.7) was major constraint perceived by BPMs. The model has been scaled up (Fig. 3) can be further up-scaled and out-scaled by other ICAR institutes in technology dissemination (Sahoo, 2016).

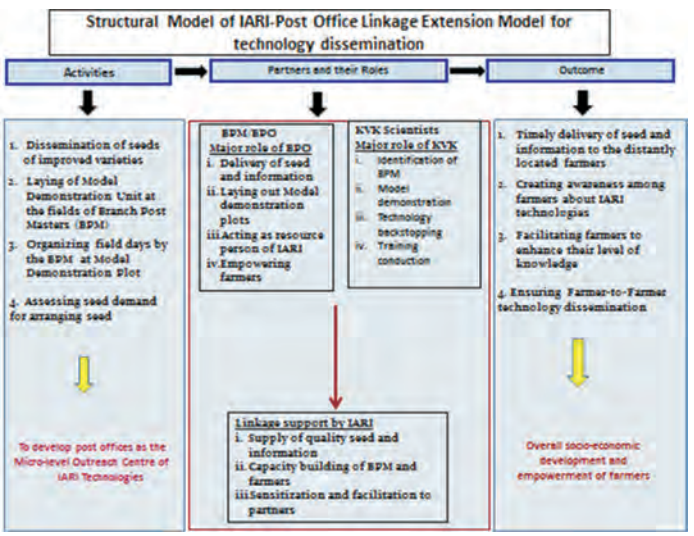


Fig. 2: Structural model of IARI-Post office linkage extension model

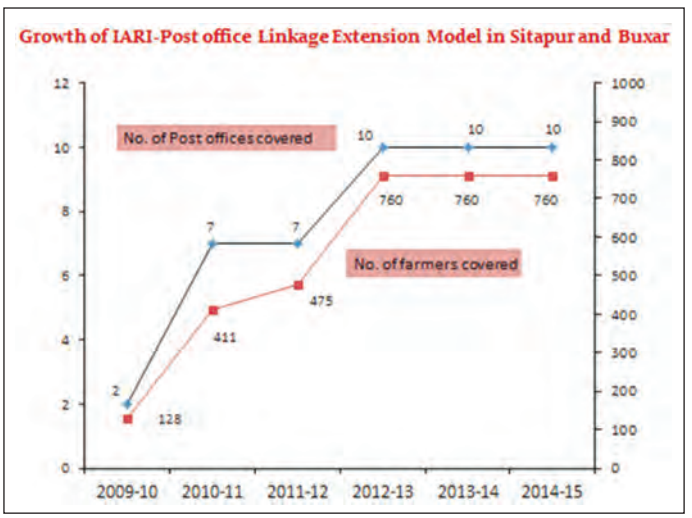


Fig. 3: Growth of coverage under IARI-Post office linkage extension model

The female agricultural officers of Kerala State Department of Agriculture and female extension personnel of Vegetable and Fruit Promotion Council Keralam (VFPCK) were more satisfied with the operating conditions and the nature of work, while female agricultural assistants were more satisfied about the relationship with supervisors, sub-ordinates and clients. Organizational role stress was low among VFPCK

women extension personnel compared to State Department women extension personnel. Analysis for different role stressors indicated role overload was the major one, followed by inter-role distance. Spearman's bivariate correlation indicated a strong and negative correlation between job satisfaction and organizational role stress of women extension personnel. Major factors affecting the job performance of women extension personnel based on factor analysis yielded seven factors in which two organizational factors, one personal, one sociological factor, and three psychological factors were there. For enhancing the job performance of women extension personnel these factors must be taken into consideration, which in turn will help in developing equitable extension system to address wide disparity that exist in gender balance to provide need based services to extension clients accordingly (Anshida, 2016).

Pluralistic regime of extension has raised the issues of complementarity and convergence as well as duplication of efforts. Identification of the extension priorities of the farmers and assessment of the level of convergence and comparative effectiveness of various agencies in addressing those priorities were carried in Akola district of Maharashtra and Bangalore Rural District of Karnataka with the purposively selected initiatives of Convergence of Agricultural Interventions in Maharashtra and Corn Growers Association in Tubagare Hobli of Bangalore Rural District, Karnataka. The most important information, training and support service needs for cotton farmers were related to pest management and timely availability of seeds. Market and price information were important for corn growers. Cotton farmers of Akola District perceived input dealers as the most effective agency followed by State Department of Agriculture and KVK. NGO was found least effective by the farmers. In case of corn growers, highest effectiveness was for State Department of Agriculture, followed by KVK, Input dealers and NGO. The study revealed a high level of convergence for the initiatives (mean index score of 0.58 for CAIM and 0.78 for Corn Growers Association) selected for the study. Further, it was identified that factors namely felt need for convergence, strategic action for convergence, resource investment and structural flexibility are positively related to the extent of convergence. Farmers perceived above average improvement in areas of inputs and services, skills and approaches, economic benefits, and network and gender due to convergence. Further, quantitative assessment focusing on three outcomes (1) Yield, (2) Cost of Cultivation and (3) Net return has also revealed significant impact of convergence initiatives. In case of converging agencies also, impact of convergence was quite visible. Delphi exercise to forecast human resource requirement in extension has figured out optimal number of farmers per extension personnel ratio as 500:1. Three distinct sets of required capabilities of extension personnels identified were 1) managerial capabilities required by extension personnel, 2) Farmers' mobilization, linkage and upliftment using new technologies and 3) Networking and integrating various skills, technologies and knowledge along the value chain (Sajesh, 2016).

The motivational analysis of organizational climate revealed that the overall organizational climate of Krishi Vigyan Kendras was a dependency-affiliation climate profile. Delphi technique used to forecast the future competencies required by the Extensionists in changing agricultural scenario revealed that knowhow skills like international know how, technical knowhow and human know how skill will be important in the coming decade. Knowledge and expertise in ICT, mobilizing and capacity building, creative thinking, conflict resolution, stress management, soft skills etc. are some of the future competencies to be developed by the Extensionists. Training need analysis showed that the "designing and conducting of a training programme" had highest Mean Weighted Discrepancy Score (5.189) followed by "assessing training need of the farmers" (mwds 5.073). Organizational barriers were the most important barriers perceived by the Extensionists in acquiring the desired competencies. The e-learning module developed was validated by the respondents and the results showed a general acceptability of the module in terms of the criteria's taken into consideration. On the basis of theory of planned behavior, subjective norms emerged as the strongest predictor of the Extensionists' intent to use or develop e-module implying that an e-learning environment needs to be promoted in an organization for facilitating capacity building of Extensionists (Rohit, 2016).

Farmers' Innovations and Indigenous Technology

The tribals farmers of Arunachal Pradesh had a rich pool of indigenous technical knowledge. As the local varieties were robust, hardy and tolerant to disease and insect pests, the farmers preferred local verities like Emo, Pyaping, Pyat, Mypya etc. For irrigation purposes they diverted the hill streams through the forest which carried lot of nutrients and organic matter to the agricultural fields. The determinants of usage of ITK were found to be age, education, farming experience, annual income, coordination ability, risk-orientation, self-confidence, etc. There is a need for proper documentation and validation of ITK by in-depth analysis of ITK and sustainable crop biodiversity conservation in several agro ecological systems (Rakesh, 2015).

The socio- psychological factors were the major determinants for the genesis of innovations (Fig. 4). Problem solving nature, creativity in thinking and enterprise diversification were identified as major among the socio-psychological determinants. Identified category wise stimulants of innovations were 'problem faced by self' in crop production and horticulture, 'innovation induces innovation' in farm machinery, 'experimented purposefully with curiosity to start a new venture' in processing and value addition; and animal husbandry innovations. The linkage with organizations was the major means of spread of farmers' innovations. Further Friedman's test also revealed that there was significant difference in different means of spread. Wilcoxon Mann Witney test showed that there was significant difference between innovators and non-innovators. Among the selected categories, it was found that processing and value addition; and horticulture innovations fetched more income to the farmers. Extent of involvement of institutional stakeholders in scaling up of farmer-led innovations was analyzed with Friedman's test to identify the degree of involvement of institutions in each category. It was found that the degree of involvement was higher for documentation and dissemination followed by validation and commercialization. Further prioritization of institutes/organizations for different activities of scaling up was done. It has been inferred that district level institutions for documentation, research institutions for performing the validation, private agencies for commercialization and state agricultural universities for dissemination of farmer-led innovations to play a vital role. It can be concluded from the study findings that marketing constraints for innovators in scaling up, technical constraints for non-innovators in adoption of innovations and administration constraints for officials in scaling-up of farmer-led innovations were most severe constraints (Baliwada, 2016).

Fig. 4: Farmers' innovations: (a) Portable multipurpose food processing device (b) modified paddy transplanter

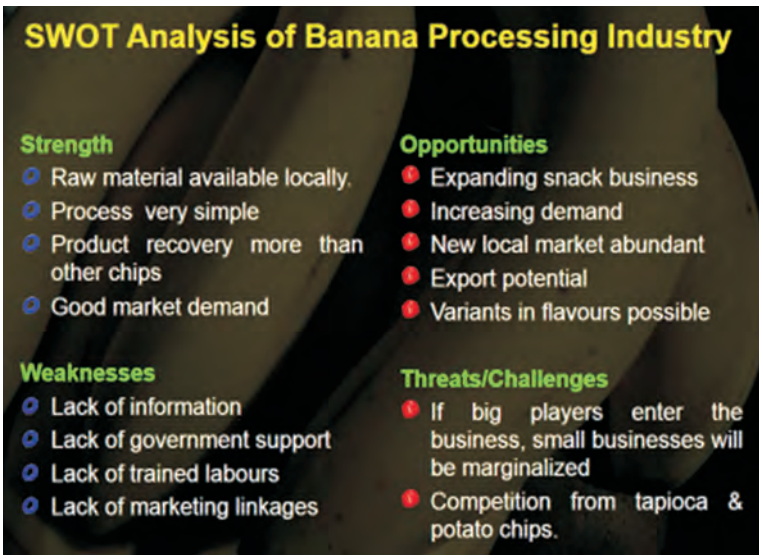
Market-led Production, Value Addition and Agri-Business

A study on theory building in agri-business management behavior of farmers showed that the 9 predictor variables viz., socio-economic status, sources of information utilization, training received, and marketing facilitates, value-orientation, and family income, risk taking ability, and achievement motivation, jointly accounted 75 percent of variables in agri-business management behavior of farmers. Out of nine predictors variables, four variables, viz., sources of information utilization, value orientation, marketing facilities and achievement motivation were more significant and explained to the extent of 71 percent of variation in agri-business management behavior of farmers (Seeralan, 2008).

The contract farming by PepsiCo in Punjab had a positive influence and many companies in different states had adopted contract farming in different crops. Assured price, immediate payment, and input assistance were the important motives of contract farmer. Credit institution must be motivated to provide necessary financial back up for contract farming. There should be provision of legal cell in agriculture department to provide legal advices (Kumar, 2008).

A majority of respondents in Karnataka indicated that gherkin farming was more prestigious compared to other types of crop cultivation. The private-farmer model of gherkin technology is successful, hence it may be scaled up in similar agro ecological situations. Labour saving technology may be developed in order to reduce the labour problem. Safety precautionary measures may be developed for spraying of pesticides (Prasad, 2008).

An analytical study of commercial cultivation of cut flowers in Meerut division of Uttar Pradesh revealed that a majority of the cut flower cultivators were found to have high or medium level of knowledge of technologies and they had a medium level of risk orientation and economic motivation, media exposure was high (Rakesh, 2009).



Processing and vale addition in banana could improve the economic condition of farmers and reduce heavy losses to matured green banana besides reducing the influence and impact of middle men. However, the most severe constraint faced by the banana growers were lack of technical information about vale addition and processing in banana. SWOT analysis of banana cultivation in Maharashtra provided elements of strategies to promote value addition and processing of banana. Availability of raw material locally, simple processing techniques and good market demand were the major strengths; while the lack of information, lack of government support, and lack of trained labours were the major weakness. Entrance of big players in business and competition from tapioca and potato chips were the major threats for the banana processing industry (Bhalerao, 2010).

Principal component analysis showed that five closely related factors like value, ethics, internal dynamics, internal–external relationship and social affiliation were associated in enhancing the development of marketing society. The major factor in facilitation at the initial stage of market society development was financial benefit, later replaced by social belongings and improved social status (Teklehaimanot, 2010).

The organic cardamom production in Kerala is an identifiable example of the positive impact giving factor in the farming sector. The farmers who are following the organic cultivation were earning a desirable margin of the profit by the marketing of the certified brand of the organic cardamom. Aspiration level of the organic and inorganic farmers was not differing significantly. However, the risk taking ability and entrepreneurial competencies of the organic cardamom farmers were much more than that of the inorganic cardamom farmers. The constraints related to certification and marketing aspects had a major role in creating obstacles to promotion of organic farming and its spread in the state (Gills, 2012).

There is need to promote peri-urban agriculture in view of rapid urbanisation. Unavailability of inputs like fertilisers during peak period and unavailability of labour during harvesting with high cost of fertilisers and plant protection chemicals were the prime constraints indicated by the vegetable growing farmers of peri-urban

agriculture of NCR Delhi. IARI emerged as the most sought institution providing technological support in the selected areas. Analysis of production dynamics showed that there was a need to organise training to peri-urban farmers to inculcate entrepreneurial motivation in them (Rohit, 2013).

An analysis on retail markets of vegetables and its implication in peri-urban agricultural system revealed that the farmers' satisfaction towards organized vegetables outlets was high. About two-third of them were satisfied with the overall performance of the collection centers, payment method, and the prices offered by the collection center. As high as 87 per cent farmers were satisfied with the behavior of the employee working in Safal and Reliance fresh collection centers. Similarly, more than two-third of the consumers were satisfied towards vegetable at retail stores of Safal and Reliance fresh found that 69 per cent of the respondent expressed satisfaction with freshness of the vegetables, vegetables quality, hygienic condition of the store, and trustworthiness of staff. Further, in comparison to non-retail farmers, the retail-farmers had increased the cropping intensity, leased in land for cultivation, improved the education of their children, repaid their old loan and increased saving on deposits. Among the constraints ranked by the farmers, high cost of labour ranked the top most constraint followed by high cost of chemical fertilizers and problems of insect-pest infestation (Prasad, 2014).

Market driven factors such as increasing demand for the processed products, assured and remunerative price for the processed product coupled with easier availability of raw material and work force availability were the major facilitators of secondary agriculture while inadequate infrastructure, high upfront capital investment, inadequacy of information and marketing linkages, lack of electricity supply, maintenance of quality standards, *etc.* posed challenges to its growth. Processing activity not only increased the value of the raw material by several times but also brought about desirable socio-economic benefits such as increased employment opportunities and improvement in income and lifestyle. Raw material cost shared significant portion of consumer's rupee (9-34% for banana enterprises; 14-60% for potato enterprises) followed by processing charges (8 to 57%) and processor's margin (24 to 59%).Processor's margin, marketing cost, marketing margin, price spread and producer's share in consumer's rupee varied with the length of marketing channel. Supply chain efficiency was inversely related to the length of marketing channel and marketing costs. Highest proportion of marketing costs was incurred by processor (2-11%) followed by super-stockist (2-4%) or wholesaler (2-3%). Highest percentage of profits were realized by processor (26-59%) followed by retailer (8-17%). Packaging charges (4-11%), transportation costs (2-11%) and sales tax (2-6%) contributed significantly towards increase in consumer's price. Price spread was lesser and producer's share in consumer's rupee was higher for shorter marketing channels with less number of intermediaries and *vice-versa* for lengthier marketing channels (Dabbadi, 2014).

The extension services in respect of backward and forward linkages rendered by North Eastern Regional Agricultural Marketing Corporation Limited (NERAMAC) were found to be medium in effectiveness by majority of the farmers in the region. The member farmers used better mass media and showed greater extension agency contacts, showed better risk and market orientation and higher achievement motivation than the non-member farmers. NERAMAC are efficient in availing inputs to farmers which are better in accessibility, quality and timeliness. From logit analysis it was found that the land holding, monetary profit and market support under fruit crops are the major predictors of motivation to join NERAMAC. As a measure of economic gain, transaction output cost due to sale of harvested produce and the mean transactional input cost per hectare was less for members as compared to non-members. In case of the organizational constraints, the respondents ranked lack of co-ordination among the members as first. The functional constraint like inadequate supply of fertilizer and pesticides at proper time as per Garrett's score ranked at first position. The members ranked timely availability of inputs and market information as the first and foremost major constraints in the order of importance. According to non-members, fluctuating market price and finding appropriate market ranked as the first and foremost major constraint. Therefore a coordinated attempt to promote similar organization and also to synergise and converge these efforts at the district and below level was required to improve the performance of various stakeholders and also for socio economic gain of the farmers (Das, 2014 and Das, *et.al.* 2015).

Analysis of marketing extension system in peri-urban agriculture revealed that the major constraints faced by the farmers were lack of transparency in biddingand lack of price regulation system. Some of the best practices were use of mobiles for enquiring prevalent *mandi* prices, choosing the sale venue based on this information, contract farming, collective farming etc. Some of the strategies were precise marketing intelligence generation; sharing of marketing information; use of modern ICT tools for networking of geographically separated market, sharing of real time market situation among farmers, quick communication, and better connectivity; creation of facilities for primary processing at farm or collective village level; speedier transportation and proper grievance redressal system (Kumar,2014).

The adopters of polyhouse found to have higher education, younger age and higher size of land under polyhouse. The average savings have increased drastically to the tune of more than Rs 60,000 per family per year in Karnataka and Maharashtra states. The analysis of the B: C ratio revealed that that gerbera crops had the highest B: C ratio (3.1), followed by rose (3.0) and carnation (2.46) in Maharashtra whereas it was 2.42, 2.36 and 2.18 respectively for gerbera, rose and carnation in Karnataka. Gerbera has more B: C ratio because of its hardy nature, less price fluctuation in the market and its resistance to pests and diseases. Among the constraints, complete absence of alternative and cheap technologies by public sector has led to the4 monopoly of the private companies for services and inputs (Prabhakar, 2015).

A study on post harvest decision making pattern and marketing behaviour of peri urban farmers in several peri urban areas of northern Indiashowed with contingency analysis using Pearson's Chi-square and significance test using Cramer's V correlation that the access to guaranteed market, access to market information, membership in organizations and education level were highly correlated with the decision to take or not to take post harvest and value addition interventions.All value added commodities other than fish showed statically significant difference in net income when compared with raw form.More than 300 per cent of increase was found in honey.SWOT analysis of different successful cases showed that achievement motivation (100 %) and risk taking ability (93%) were the most important determinants in their success.For highly perishable commodities like fish and flowers, market related constraints were identified as more severe constraint than all other things. In high initial investment needed commodities like seeds and food grains; infrastructure related constraints were identified as most severe one. The study furnished some of the strategies for making farm to fork a reality, which include collective processing and collective marketing, creation of market infrastructure, store houses and cold chain management, creating awareness about the government support policies to farmers and providing market intelligence and guaranteed market (Gills, 2015).

Entrepreneurship Development

A study under taken in three selected agricultural institutes namely IARI, PAU & HAU with total samples of 150 students revealed that most of the students preferred career in non-agriculture sector such as administrative services, police and army officers. It was noticed that greater percentage of the respondents had low level of knowledge and skill in important aspects of managing business operations, required by potential entrepreneurs with respect to entrepreneurial behavioral dimensions It was found that majority of the students had medium level of self confidence, medium and low level of ability to be persistent, medium to low level of usage of feedback for self-development. A large majority percentage of the respondents scored in the range of moderate to high internality with regard to locus of control. Over 59 per cent of the respondents identified security of employment in the work as an important factor in choosing career followed by high starting salary and long-term salary prospect. Whereas majority 62.00, 60.00 and 55.00 per cent of the respondents identified, impact of the work on social life intrinsic interest, value of the work for the community and status attached to the work as fairly important factors respectively. (Gelan, 2007).

The raisin entrepreneurs of Nashik District of Maharashtra scored high on psychological characteristics like risk taking, achievement motivation, innovativeness etc. The raisin makers used marketing channel which

was very long, had high marketing cost and less efficiency. They earned double the profit from raisin than only from grapes. The most severe constraints faced by the raisin entrepreneurs was lack of price policy for raisin by the government, followed by lack of procurement policy of government, and high cost of plant protection chemicals. Development of e-module to give timely marketing information to raisin makers, assured price for raisin by the government, training in packaging, grading and processing were some of the suggestions given to policy makers (Nikam, 2009).

Potential enterprises in both the tribal districts of Rajasthan namely Banswara and Dungarpurwere related with intensifying and expanding existing agriculture and allied enterprises. Some of the new opportunities were off-season cultivation of vegetables (17-30%), seed production of crops and vegetables (8-9%), cultivation of horticulture crops (8-17%), production and sale of vermicompost and getting retailership/dealership of agricultural inputs such as seeds, fertilizers and pesticides. However, the priorities of farmers with respect to prospective enterprises in both the districts differed on account of lack of irrigation facilities in Dungarpur district. Goat keeping (43%), backyard poultry (33%) and dairy enterprise (30%) were the most preferred and sought after enterprises in Dungarpur district whereas farmers preferred dairy enterprise (33%), vegetable cultivation (30%) and seed production (15%) in Banswara district. Majority of the respondents (52 to 68%) exhibited medium level of entrepreneurial orientation irrespective of poor literacy level and very low social participation. Creating opportunities to fully exploit prospective enterprises and capacity building of farmers especially youth would help in improving the livelihood security and sustainability of farming in the region (Dipika, 2009).

An analytical study of best practices and competencies of award winning agripreneurs of Tamil Nadu showed a significant relationship between extent of adoption of best practices and agri-preneurial success. A positive and significant relationship was observed between entrepreneurial competencies and agri-preneurial success. Fluctuating market demand, availability of marketing facilities and high cost of investment were found to be major constraints (Raj, 2010).

A critical analysis of entrepreneurs in protected agriculture in Maharashtra, showed that the entrepreneurs were moderate to high on the entrepreneurial success scale specially developed for the study. The growth of this sector was hindered by many constraints like no proper pricing of the cut flowers, high commission by middlemen, pricing heavily depending on demand and supply, high initial investment, lack of skilled labour, pest and disease management, no proper information regarding pricing of the cut flower to the growers by commission agent and lack of knowledge of cold chain management. High initial investment and high bank interest rate were the main reasons for reluctance on farmer to take up protected cultivation technology (Sopan, 2011).

Regression analysis indicated that 58.80 per cent variation in the entrepreneurial behavior of baby corn farmers of Haryana was attributed to their age, education, family size, occupation, land holding, social participation, family income, farming experience, aspiration, extension orientation and scientific orientation. The study revealed that 78.33 per cent of farmer's family had employment for their male members only but after babycorn cultivation 81.67 percent of farmers got employment for all family members. About 37 per cent of babycorn farmers reported increase in annual income by 3 to 4 lakh rupees. In case of increase in assets possession, half of the baby corn farmers reported an increase of Rs2 to 3 lakhs worth assets as compared to non-baby corn farmers who reported only Rs1 to 2 lakhs worth of assets increase. The fluctuation of market price, unavailability and high cost of quality inputs, and lack of skilled labour were the three major constraints faced by babycorn farmers. A majority of babycorn farmers suggested that quality inputs should be easily and cheaply available to the farmers (Sinha, 2011).

Analysis of microfinance in relation micro-enterprises development in Manipurwith four cases of micro-entrepreneurs engaged in various enterprises like piggery, duckery, poultry and weaving revealed enhanced income generated and enterprise growth over the years, increased spending by families and overall empowering effect for micro-entrepreneurs. Easy to access and availability of service locally and lower interest rate than the

money lender were the most important factors for continuing with the microfinance service. The most severe problem faced by the beneficiary micro entrepreneurs was frequent *bandh* and the blockade. The second most important constraints found was the non-availability of inputs on time. Pearson correlation analysis revealed that the motivation to avail microfinance service was significantly and positively correlated with psychological factor- risk orientation and innovativeness of the respondents (Sharma, 2012).

Content analysis of 20 case studies revealed that innovativeness, social network and risk taking ability were the most common and essential attributes of agripreneurs. Variables such as autonomy, desirability of business and expected utility of business were the motivating factors. The motives also included non-monetary incentives such as professional satisfaction, service to fellow farmers and community and concern for environment. The comparative analysis of agripreneurs and conventional farmers found thatagripreneurs were relatively young with higher education levels, social participation and levels of aspiration. Agripreneurs also owned and cultivated more land and consequently had higher annual incomes. It was found that marketing channel was comparatively short for agripreneurs. Agripreneurs frequently sought new information on agriculture/enterprises from diverse sources including scientists. Education, level of aspiration, manageability and inquisitiveness were found to be correlated with entrepreneurial attitude of farmers. With Mann-Whitney U test achievement motivation ($p<0.01$), innovativeness ($p<0.01$), pro-activeness ($p<0.01$), self-esteem ($p<0.01$) and entrepreneurial attitude index ($p<0.01$) of agripreneurs and conventional farmers were found to be statistically different from one another. Finally, analysis using standardized canonical discriminant function coefficients indicated that aspiration (0.612), pro-activeness (0.351) and information processing behaviour (0.337) were the important predictors of agripreneurship behaviour. The characteristics that differentiate agripreneurs from non-agripreneurs farmers as predictors of agripreneurship behaviour can be used to design agripreneurship development programmes to enhance their effectiveness. Besides resource endowment, technological options, network and linkages, farmer's inner potential and drive are also critical (Hajong, 2015).

A majority of educated youth is shifting towards non-farm sector from farm sector due to lack of profitability, over-dependence on environment and high input requirements. There was no significant difference between farm and non-farm trainees with respect to use of sources of information, social valuation, entrepreneurial intention, entrepreneurial capacity, perceived support and barrier, entrepreneurial needs and entrepreneurial attributes (except self-efficacy, innovativeness and pro-activeness) but there was significant difference between trainees and non-trainees with respect to these characteristics. This indicates that training progarmmes of Rural Development Self-Employment Training Institute RUDSETI (Figure-7) were effective in motivating the youth and equipping them with necessary skills to start their own business. Study also revealed that the youth ventured into entrepreneurship because no other option was left for them to earn their livelihood. The most severe constraints experienced by EDP trainees were lack of financial support and lack of entrepreneurial culture in our society. The study concludes that trainings and skill development by institutes like RUDSETI are important along with Government support in the form of loans which will help in developing first generation entrepreneurs. There is a need to create conducive

Training on Food Processing **Training on Tailoring** **Poultry Trainee of RUDSETI**

Fig. 7: Trainings of RUDSETI

entrepreneurship eco-system and culture across the country to foster and sustain entrepreneurship among youth and create valuable human assets for the country (Sinha, 2016).

Institutional Innovations and Development

A study on Socio-economic assessment of participatory watershed development programme in Maharashtra revealed that most of the respondents had medium level of participation in different watershed activities. Education, income, land holding, occupation, house type, household size were found to be positively correlated with social participation (Athare, 2010).

With a critical analysis of community participation in Watershed Programme of Tarun Bharat Sangh in Rajasthanit was found that the value of agricultural land was found higher and statistically significant in watershed villages because of recharged water table and assured irrigation supply. The productivity of wheat and mustard was found higher and statistically significant in watershed villages4.42 metric tons per hectare and 1.44 metric tons per hectare, respectively), as compared to non-watershed villages(3.88 metric tons and 1.29 metric tons per hectare for wheat and mustard, respectively). The benefit cost ratio (B: C) was found higher and statically significant in watershed villages with B: C ratio of 1.71: 1 and 1.9:1 for wheat and mustard, respectively as compared to 1.19:1 and 1.52:1 for wheat and mustard in non-watershed villages (Raju, 2011).

Tamil Nadu Mango Growers Federation (TAMAFED) fosters functional linkages of producers with different systems and within the system in the form of backward and forward linkages in production process which involve the investments in the form of transactional costs. Farmers' collective action for better production in the form of self-help groups (SHGs), farmers' interest groups (FIGs), marketing associations, producers associations, co-operatives etc. have contributed in maximizing the input-output ratio and ultimately increasing the benefit of producers. The Logit analysis revealed that education, economic benefit and area under mango cultivation were the major determinants of motivation to join producers' federation namely Tamil Nadu Mango Growers Federation (TAMAFED). As a measure of economic gain, transactional output cost due to sale of harvested produce was two and half times less for members compared to non-members. The comparative analysis shows that mean transactional input cost per hectare was found to be significantly lower for members than non-members. The members of TAMAFED gained social and economic benefits and the costs incurred on input and output transactions were quite low ultimately adding to the benefit. Small and marginal farmers, lack the capabilities of investing alone in post-harvest infrastructure and it is hard to meet the requirements of the corporate buyers, who on one hand have certain minimum quantity requirement as well the quality requirements in the form of type of agriculture practices followed. As such, the transactional costs of the farm produce especially the fruits like mango can be reduced through farmers' federations as depicted in the present investigation (Parthiban, 2012 and Prathiban, *et.al*. 2015).

A study of Mahagrapes in India revealed that its members had slightly more income (Rs. 13.32 lakh) than independent farmers (Rs. 12.38). More number of Mahagrapes members were small and marginal famers (15.5 %) than independent farmers. The productivity of grapes at Mahagrapes members field was slightly more (11.78 quintal/acre) than independent farmers. Mahagrapes members were more socially active (mean score 7.3) than independent farmers (mean score 6). In terms of psychological impact also Mahagrapes members had shown more risk taking behaviour, self confidence and achievement motivation. Most of Mahagrapes members were satisfied with extension services of Mahagrapes with overall satisfaction score of 80.87. It was also found that 97.5 per cent members had knowledge about recommended practices and GAP standards, while 98.66 per cent had adopted it. Overall effectiveness index score was found to be 92.34 per cent. Mahagrapes members had backward linkages with scientists from NRC grapes, Pune, Mahagrapes consultant, private input dealers, private nurseries etc. Non-members also had same linkages except consultancy services of Mahagrapes. Members' forward linkages were mostly with Mahagrapes, while the non-members had with private traders and private companies like Mahindra etc. The total input transaction cost of members was Rs. 595 while of non-members was Rs. 816 with difference of Rs. 220. Thus members save 37 per cent of input transaction cost

than non-members. Themembers perceived provision of needed infrastructure like pre cooling, export facility centre etc. as most important determinants of success of Mahagrapes (Nikam, 2013).

The study on farmer organizations in Punjab revealed that the number of members, villages covered, outlets, activities undertaken by the organization were continuously increasing in both the organizations (FAPRO and UAPCL). The days of employment generated and annual turnover (presently, Rs. 200 lakhs for FAPRO and Rs. 70 lakhs for UAPCL) had also steadily risen over the years. The existence of high extent of group processes, with the GPI being 86.12 in FAPRO and 80.84 in UAPCL is an indicator of the long term sustenance of the organizations. Both the organizations portrayed high degree of social equity and environmental accountability. Thus, it can be inferred that the organizations are sustainable and are still continuously growing. It was found that income and employment for the members in both the organizations were significantly higher in comparison to the non-members. The members got employment to the extent of 181.67 days in FAPRO and 151.67 days in UAPCL. A significant difference between the members and non-members was observed in terms of food security and social empowerment (Sahu, 2014).

Risk Perception, Adoption and Impact of Agro-Biotech Crops

The higher Benefit-cost ratio (2.6:1) for Bt. cotton as compared to non-Bt, cotton (1.6:1) showed the profitability of cultivating Bt. Cotton in Ganganagar and Haunmangarh districts of Rajasthan. It was also observed that the average number of pesticide sprays had reduced by about two times. None of the Bt. framers sprayed more than 3 times and about 64 per cent of them sprayed in range of 1-3 times, while 55 percent of non-Bt cotton farmers sprayed for more than five times. The multivariate logistic regression analysis revealed significant and positive relation of innovativeness, social participation and experience in cotton cultivation with adoption. Mean perception score of farmers as 3.34, showed that their perception about Bt. cotton was largely positive. For better adoption of Bt. cotton constraints like high seed cost, unavailability of irrigation in time and in adequate quantity and lack of information dissemination of complete package of technology through public extension system need to be managed. Farmers should be made aware of IPM packages for management of cotton pests through intensive extension programmes. (Leela, 2007).

The average yields of B. cotton obtained by the farmers of Punjab and Karnataka were found to be 11.09 qtl/acre and 10.05 qtl/acre, respectively. A majority of the farmers of Punjab (42.5 per cent) obtained yield in range of 11-13 qtl/ acre while the average yield of 35 per cent of them feel in the yield category of 9-11 qtl/ acre. However, in case of Karnataka, for a majority of them (42.5) average yield was in category of 9-11 qtl/ acre and for 30 per cent of them it was in category of 11-13 qtl/ acre. By cultivation of Bt cotton, Punjab farmer obtained 69.8 per cent extra yield as against traditional cotton while in case of Karnataka, the farmers gained 45 per cent in increase in their average yield. Similarly, the mean difference in yield of Bt. cotton of Punjab and Karnataka was highly significant ($P<.01$). That the Bt. Cotton was beneficial for farmers was revealed by mean perception score above 3.5 on scale of 1 to 5. A majority of farmers expressed agreement to statements like Bt. Cotton hybrids are eco-friendly and high yielder (Mean score:4.46); Seed production and trade of genetically modified crops by public institutions will be more advantageous for farmers (Mean score:4.38); Before the use of Bt. cotton, the environment in crop field remained polluted due to rampant use of pesticide (Mean score: 4.18); Farmers have been benefited immensely by Bt cotton cultivation (Mean score: 3.80); and Pesticidal pollution and health hazards have reduced due to Bt. cotton (Mean score: 3.56). The suggestions related to biosafety included A Block level monitoring Committee needs to be created to ensure better compliance to regulatory measures and to strengthen the existing District level committee; capacity building of extension organizations in transgenic technology and biosafety management needs to be initiated at wider scale; prior consent of farmers and farmer participatory trials of transgenic crops should be stressed upon to address farmers' protest; and development and dissemination of awareness and risk communication literature should be emphasized for proper understanding; were the major recommendations of the study (Surya, 2008).

The study showed that the average yield increase through adoption of Bt cotton was 42 per cent (1240 kg/acre) as compared to conventional variety of cotton (875 kgs/acre) in Gujarat. Total cost increased about 10 per cent in Bt cotton cultivation compared to non-Bt cotton, but at the same time total increment in gross return was about 42 per cent and net profit of about 74 per cent in Bt cotton compared to non-Bt cotton. Overall increase in employment was 14.625 man days. The investigation revealed that the Benefit-Cost Ratio of Bt cotton was 2.541:1 while it was only 1.97:1 to non-Bt cotton. The secondary level impacts due to increased profit level from Bt cotton showed that more number of farmers invested the money in the farm improvement, households material purchase, housing improvements, education, etc. Overall about 41.13 per cent of farmers had awareness with the all aspects of Genetically Modified (GM) crops and the corresponding figure of extension workers was 74.5 per cent. The study revealed that with overall mean attitude score of 3.6, as much as 93.75 per cent of the farmers were found to have favourable attitude towards the GM crops and foods. With the mean attitude score of 3.78, all the (100 %) extension agents had positive attitude towards GM crops and foods. It was found that the 47.5 per cent of the farmers had favourable perception, whereas 45 per cent of the farmers had unfavourable perception towards environmental and health hazards of GM crops. The corresponding figure of extension workers was 53.33 per cent and 33.3 per cent, respectively. It was found that progressiveness, risk orientation, innovativeness, economic motivation and education had positive correlation with the awareness, attitude and perception of the farmers, whereas it was experience, formal training and involvement of advisory to Bt cotton which had positive correlation with the awareness, attitude and perception of the extension workers (Manikanhaiya, 2012).

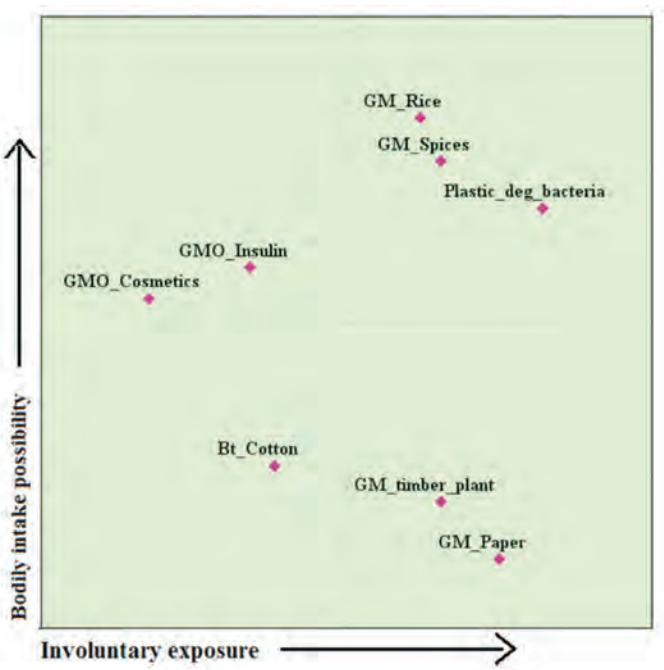


Fig. 8: Multidimensional mapping of various agro-biotech applications

The past experience in the cultivation of agribiotech crops increased the probability of adoption ($\beta = 0.133$, $p = 0.017$)whereas the socio-economic ($\beta = 0.893$, $p < 0.01$) and health risk perception ($\beta = 0.881$, $p = 0.029$) negatively influenced the adoption of agro-biotech crops. The multidimensional scaling revealed that consumers had differential acceptability for various applications of agro-biotechnology even though they are developed using similar technology. It was interpreted that *Possibility of Bodily Intake* and *Extent of Involuntary Exposure* are the major determinants of acceptability of agro-biotechnology applications (Figure 8).

Propensity score matching revealed that as a result of the adoption of transgenic hybrids, the number of pesticide sprays and expenditure on plant protection chemicals had gone down significantly and the cotton productivity increased by 456.75 kg/ha. As a result of which, the net profit from Bt. cotton cultivation increased by 46.13 per cent. It was also observed that as compared to conventional cotton fields, the Bt-cotton cultivating fields witnessed thes ignificantly lower number of health hazard incidences as a result of a reduction in intensity and frequency of pesticide sprays ($t = 6.629$, $p < 0.01$).

Agro-biotech risk communication analysis revealed that print media played limited role in imparting education and creating awareness about agro-biotech crops and portrayed the subjective concerns of non-expert stakeholders. Major emphasis was given to the concerns of well organized but less concerned stakeholders neglecting the experiences and opinions of the farmers and other vulnerable sections.

There was a decrease in income from agriculture and an increase in income from other sources such as industries, business and service sectors and from the foreign countries, especially from the Persian Gulf countries as well as UK and USA. Urban young group was having highest Livelihood Security Index (LSI) value at 58.10, followed by urban old group at 57.50. Rural young and rural old groups had LSI values of 54.74 and 55.98 respectively. Regarding the change in LSI values, there was a positive change in all the four groups over the past period under consideration. This was the maximum in case of urban old and urban young group. The changes in the rural area were not much significant. Dignity was ranked the highest by rural women, which was followed by family, 'social consciousness' 'faithfulness', education' and 'honesty', 'Sacrifice ' and 'spirituality' were the values given lowest ranks by women in all the groups. (Letha, 2007).

A majority of organic and inorganic farmers had medium level of knowledge about organic farming. The variables of organic farmers, namely education, experience in organic farming, livestock possession , social participation, extension orientation mass media exposure, innovativeness, economic motivation, risk orientation, market orientation, decision making behaviour environmental orientation self-confidence and level of aspiration showed significant and positive relationship (Jaganathan, 2009).

Age, social participation, extension contact, and achievement motivation significantly and positively contributed, while the family size satisfaction with life, types of land holding negatively contributed to change in technology use in rice cultivation in Burdwan district of West Bengal (Nag, 2009). Economic factors ('reduction in cost of cultivation', 'enhancement of net income and profit', 'internal dependency on inputs', 'sustain livelihood security' and 'better price for organic produce'); propaganda factors (Krishi Pundit awards', 'promotional schemes and advertisements of the government', 'awareness campaign by government organizations and NGOs'), and 'success stories of organic farmers' were considered to be motivating farmers to practice organic farming. Ecological factors such as 'reduction in pesticide poisoning (nausea) cases', 'diversification of agriculture', and 'increase in productivity of soil and improvement in soil health' were considered to be high motivating factors. The sum of production elasticities or returns to scale in all the three pigeonpea based cropping systems was more than 1 for both organic and conventional farming situations. But it was more in case of organic farming: pigeonpea-sole crop (1.48), pigeonpea+ black gram (1.54) and pigeonpea+ green gram (1.62).Thus it can be concluded that the returns to scale in organic farming of pigeon were increasing gradually from sole crop of pegionpea to intercropped pigeonpea. When the elasticities of production of individual factors were considered, the elasticity of production for human labour, seeds and plant protection were positive and significant in case of organic pigeonpea (sole crop) cultivation. But in case of organic pigeonpea intercropped with black gram or green gram, the elasticities of production for human labour were positive and significant. These results indicate that in organic farming the contribution of human labour is significantly more. In case of conventional pigeonpea cultivation practices only manures and fertilizers were contributing to pigeonpea yields in pigeonpea (sole crop) as well as pigeonpea + black gram cropping systems (Malgatti, 2011).

The technological gap index for pomegranate cultivation in Karnataka was the highest in water management (20.31), followed by weed management (17.43). The disease and pest management during rainy season and hot summers result in low production and low quality produce not suitable for marketing and export, which was found to be at third rank in technological gap with a score of 10.94, followed by 'Nutrient management' with a score of 6.59. The technological gap varied with the land holding of the farmers. In case of land preparation and planting the large farmers had the highest gap with a score of 8.33, followed by small and medium farmers. Whereas in case of nutrient management, medium farmers had the highest gap with a technological gap index of 7.58, followed by small and medium farmers with a technological gap index of 6.41 and 5.77, respectively (Gouda, 2011).

Per hectare saving in total variable cost due to System of Rice Intensification (SRI) wasRs5669 (22%) and Rs 3178 (12%) in Tamil Nadu and Andhra Pradesh, respectively. The major technical constraints were difficulty

in achieving perfect land levelling, problems in developing drainage channels for alternate wetting and drying and unsuitability of SRI in waterlogged condition, low lying areas and problem soils. It is clear that SRI is not suitable to all areas so it should be promoted based on location specific context (Johnson, 2011).

The most crucial constraints to organic farming were lack of reliable package of practices of organic farming, non-availability of organic farming input, lack of awareness of grading and food quality standards among organic producers, high cost and long certification period, loss of yield in initial period and long-time taken for recovery and positive response of ecosystem. Four case studies revealed that NGO and farmer group helped in organising the organic farmers group in providing land holding support in production, quality control and marketing through organic bazar, and eco shops. As all the stakeholders have key role in promoting organic farming, concerted group action need to be taken up by consensus of all stakeholders. Educational and economic support should be provided during initial years of yield loss for neo converts to organic farming (Shinogi, 2011).

Attitude of the farmers in Uttar Pradesh and Rajasthan towards zero tillage technique was found favourable and positive. Favourable attitude among higher proportion of farmers was mainly due to reduction in fuel and machinery use, reduced problem of shortage of agricultural labor, management of the widespread degradation of soil resource base, promotion of climate change adaptation technology as it reduced agricultural- related green house gas emission. Non-adopter also felt that zero tillage technology saved diesel, money and water not only in first irrigation but in subsequent irrigation also. The benefit cost ratio for zero tillage (2.31) and laser land leveling (1.94) was higher than the conventional system. The average technical efficiency of adopter farmers was about 74 per cent and non-adopter was 44 per cent. Garrett Ranking revealed that higher yield, saving of water and saving of monetary input were the major perceived benefits (Garrett score 60) adoption of the CA technologies. CA practice adopters further reported that there were significant farm level, social, economical and ecological changes as a result of adoption of CA practices (Leela, 2012).

The important reasons for low adoption rates of precision farming in Tamil Nadu were high costs of technologies adoption, uncertainty in returns due to adoption, lack of demonstrated effects of technologies on yields and input usage mechanism, small farm size, infrastructure constraints, non-availability of high value inputs, and attack of insect pest and diseases (Krishnan, 2012).

A majority of respondents (45%) in Coral Islands of Lakshyadweep had non-farm based livelihood. The drivers of diversification showed that economic motivation in different livelihoods was not significantly different. The personal constraints followed by economic, promotional and infrastructural constraints were the significant constraints in diversification. There is a need to promote the livelihood based on both farm and non-farm activities for ensuring the sustainability of the livelihood system (Anshida, 2013).

Among the various innovative methods for higher income practiced by farmers in Karnataka, Integrated Farming System (IFS) was found as a common feature among all the cases except for the fact there was variation in the number of enterprises adopted. Small, medium and large farmers integrated six to ten enterprises. The B:C ratio was found to be higher than 1.5 for IFS farms. Highest B:C ratios were obtained from livestock enterprises of sheep and emu rearing. The farmers also expressed that practices such as organic cultivation of crops using locally prepared biomanures and vermicompost led to conserving of natural resources, leading to ecological, economic and social and cultural sustainability. Factor analysis identified six different factors governing progressiveness of farmers namely scientific temperament, perseverance and hard work, cosmopoliteness, achievement motivation, entrepreneurial aptitude and work satisfaction (Rakesh, 2014).

The study conducted in Karnal and Kurukshetra distict of Haryana state on basmati rice growers showed that the awareness and the adoption of the critical GAPs in basmati production system was at lowest ebb whereas the benefits of adoption were well understood by the farmers. The awareness level of farmers about Good Agricultural Practices in basmati rice was found to be 58.33 per cent whereas adoption was only 27.41 per cent which is even less than half of awareness level. Overall preparedness for adoption of GAP was 65.67

per cent which was reported fully prepared for transport (road), telecommunication and electricity. The expected benefits of adoption of GAP expressed in terms of consumer benefits, producer benefits, environmental benefit and national benefit showed farmers awareness regarding the issues. Farmers perceived depletion of ground water, sustainability of soil health and availability of finance as major challenge in adoption of new practices. The strategy for promotion of GAP in basmati rice production system includes the capacity development in technical areas, confidence building, supportive policy environment in term of MSP and premium price, research to reduce cost of cultivation and simplification of certification procedures (Pandit,2015 and Pandit, *et.al*. 2017).

The study on impact assessment of IARI technology was conducted in Ghaziabad and Hapur districts of Western Uttar Pradesh revealed that a majority (70%) of farmers belonged to medium category followed by 20.5 and 9.5 per cent of farmers in low and high category of knowledge about the scientific vegetable cultivation technologies (Figures 9 to 11). The marketing channel of cauliflower (5.49) was found to be more efficient than carrot (4.34) and bottle gourd (2.12). Extension participation, social participation and risk orientation significantly influenced the marketing behavior of respondents. *Pusa Rudhira* had higher Benefit-Cost Ratio (2.58) than the prevailing *Sungrow* variety (1.85). The same trend was found in case of cv.*Pusa Naveen* (2.30) and cv. *Pusa snowball K-I* (2.23) as compared to existing cv. *Kesar* variety (2.19) of bottle gourd and local hybrid of cauliflower (2.03), respectively. The adopters varied significantly from non-adopters in dependent variables *i.e.* knowledge level, marketing behavior and adoption rate. Adoption of IARI technologies led to significant changes in socio-economic conditions and assets of the farmers. Among the various constraints of vegetable

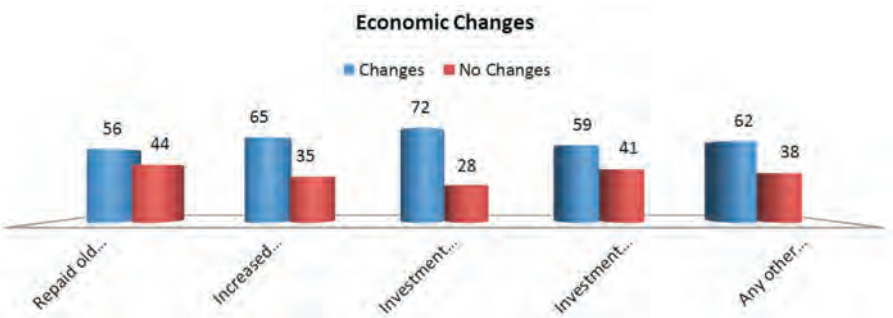


Fig. 9: Economic changes of the respondents before and after adoption of IARI technologies

Fig. 10: Social changes of the respondents before and after adoption of IARI technologies

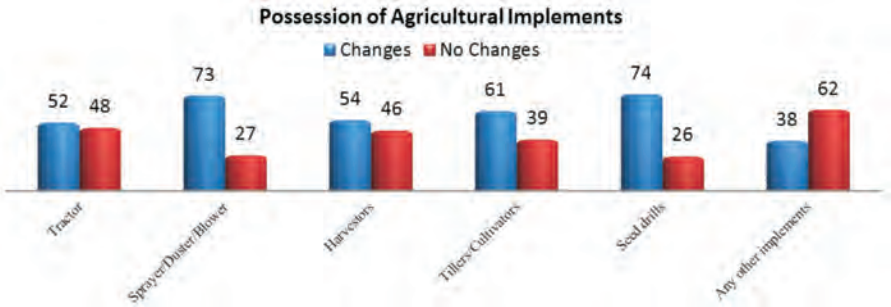


Fig. 11: Changes in possession of agricultural implements of respondents before and after adoption of IARI technologies

cultivation, technical constraints were found to be most severe followed by labour, marketing, economic and environmental constraints. Lack of sufficient amount of IARI seed was the main constraint related to adoption of IARI technologies among farmers. (Barua, 2015).

Empowerment of Women

The study on impact of non-governmental organization led self help groups on empowerment of rural women in Tamil Nadu showed that SHGs led by the Association of Sarva Seva Farms (ASSEFA) of Madurai brought high self-reliance (61.67%), self-esteem (44.44%), credit orientation (92.78%), economic motivation (56.11%), achievement motivation (47.78%) and better empowerment in psychological (76.67%), social (57.22%), economic (76.67%) and political (11.11%) aspects among rural women. SWOT analysis revealed that SHGs had sufficient strengths and opportunities such as savings, awareness, co-operative approach, entrepreneurship, learning etc. The major factors, which were responsible for the success of SHGs were family support, self-satisfaction, accessibility to credit, co-operative approach and social protection. Strict rules and regulations, ambiguity in leadership, difficulty in maintaining accounts, intra group conflicts and sub grouping were the major hindering factors. The study suggests that the focus of the group should be on its quality, capacity-building of its members and NGOs should maintain their standards so that their dictum should not get detaoriated in the long run (Sangeetha, 2009).

High level of achievement motivation, moderate training and social participation were the determinants for SHG micro entrepreneurs. Mobility and decision making were dominant variables heavily contributing for greater empowerment of rural women in India (Narayanan, 2011).

A moderate extent of drudgery was observed in all activities related to women farmers in hills. The congestion and breathing were frequently perceived by a majority (63.33%) of women during cooking. Gender friendly tools &equipment like improved sickle was found most appropriate to the 81.67 per cent of the respondents followed by wheel barrow (70.83%) and okra plucker(30.83%). About 83.00 per cent respondents were found adopters of modern storage structures such as metal *bins*, followed by integrated pest management (73.33 %) and recommended high yielding varieties of crops (56.67%).The major constraints identified were small land holdings, low financial status, and lack of demonstration and high initial cost of technologies in hill agriculture (Singh, 2015).

Climate Change Adaptation and Risk Management

The most important sources of risks in rainfed agriculture were related to weather (87%), market (86%) and financial sources (83%) as their mean scores indicate. These were followed by infrastructural, policy, technological and agronomic practices related sources. The farmers effectively managed risks and uncertainties in rained areas by seeking assistance (from formal and informal sources), managing livestock, economic or employment

strategies (seeking employment, reduction in consumption-expenditure, selling of valuables) and following better agronomic practices in short term period. The farmers in rained areas followed long term strategies like seeking institutional support (infrastructural facilities, demanding relief, irrigation facilities), efficient farming (producing at lowest possible cost, controlling pest and disease attack, conservation farming), diversification of income sources, crop insurance, agro-economic strategies (mixed cropping, intercropping, water harvesting, changing cropping pattern, migration etc.) and livestock management strategies (fodder cultivation, under stocking pasture and hay reserve, preventive measure against diseases) for anticipating and managing risks and uncertainties in agriculture (Kammar, 2008).

With the calculated vulnerability index, a majority of the farmers (about 47 per cent) of Sunderbans of West Bengal were found in highly vulnerable group followed by about 37 per cent in vulnerable group; while about 17 per cent in moderately vulnerable group. Major risks related to forestry and biodiversity included decrease in forest area and reduction in certain plant, animal and bird species. Increase in poverty was the major perceived risk under socioeconomic and cultural life by the respondents. It was observed that people devised different indigenous techniques or practices to overcome the adverse impact of climate change in their farming. The technique like land shaping, ail cultivation, dhibi cultivation etc. were related to soil management and irrigation to solve the increasing problem of soil salinity due to climate change. To protect the crops and plants, people of Sunderban region practiced some innovative technique like application of washed water of Neemplant's leave and seed, application of washed water of earth worm and washing the fog stored in leave of the plant in early morning to protect the crop from different diseases. Farmers had high level of training need in the areas like seri-culture (MS:1.33), fertigation (MS:1.23), organic farming (MS:1.33), multiple cropping (MS:1.35), sustainable farm management (MS:1.38) and entrepreneurship (MS:1.77), etc. Extreme poverty, lack of education and proper knowledge about adaptive practices, lack of information facilities with forecasting and early warning system, lack of awareness about climate change issues, lack of aptitude towards resource conservation and adaptation needs were reported as the major constraints by the farmers in their adaptation endeavours (Sarkar, 2009) .

In the broader framework of disaster management, majority of farmer were aware of during (59.82%) and post disaster(59.46%) measures but were less aware of pre disaster (25.83%) measures. Majority of respondents showed the attitude of dependency on external source of management of problem. Most of the farmers being original inhabitants (58.75%) having mostly local kinshipties(82.5%) showed existence of cohesiveness in the area (Dabbadi 2011).

A majority of the farmers of Adilabad district (highly vulnerable to climate change effects) of Telangana had moderate level of knowledge regarding climate and weather in relation to crop management. The socio-economic and psycho-personal variables like education, occupation, family type, land holding, social participation, economic motivation, innovativeness, scientific orientation and risk orientation showed positive and significant relationship with knowledge level and in regression analysis education, scientific orientation were positive and had significant contribution to the knowledge level of respondents. Television, radio, agriculture officer and progressive farmers were categorized into strong group of information sources usefulness. In order to support farm level decisions and minimize the loses in adverse climatic and weather conditions, farmers' understanding about interaction of climate and agro-ecosystem need to be bridged through inclusion of farmers' communication network (Kumar, 2013).

The level of awareness about climate change was low in women farmers as compared to men farmers in Rajasthan. A majority of women and men farmers also perceived that large scale deforestation, industrialization and heavy use of fossil fuels were the reasons for the present climate change but about 30 % of the men and women farmers were unable to perceive similarly. It was observed that women farmers were more vulnerable than men farmers in the study area. The reasons for high vulnerability in women may be the low level of awareness about climate change, perception about climate change; belief in fatalism and egalitarianism, and high dependency on the common property resources. Adaptation practices like use of soil conservation techniques,

rain water harvesting, use of drought tolerant varieties, intercropping, change in time of farm operations, crop rotation; use of water conservation techniques, *kannabandi*, etc. were used by the men and women farmers. It was observed that the women farmers had less access to institutional interventions as compared to men farmers (Bishnoi, 2013).

Analysis of climate change led vulnerability and adaptive behaviour of farmers in the Himalayan (Himachal Pradesh) and Arid (Rajasthan) revealed that the farmers of had very low awareness and knowledge. The vulnerability index score for Himachal Pradesh was 0.82 and for Rajasthan it was 0.73. The analysis revealed that 38 per cent of the respondents in Himachal Pradesh belonged to highly vulnerable group as against 31 per cent of Rajasthan (Figure-12). The findings on simulation of adaptive behaviorthat majority of the respondents showed highly unfavourable adaptive behavior (35% in Rajasthan and 30% in Himachal Pradesh). Logistic regression analysis was carried out to simulate and identify the predictors of adaptive behavior. The socio-psychological variables like awareness, attitude, stress, pessimism, communication, area, age, education, production orientation and openness were found to be significant explanatory variables of adaptive behaviour. The major documented indigenous knowledge and practices were 'mind' cultivation, 'chal' to harvest water, 'apple paste' to control diseases and 'siddu' to protect from extreme cold in Himachal Pradesh. Similarly, the major documented indigenous knowledge of Rajasthan were–Khadin' farming system to manage drought, 'kanabandi' to manage soil and wind storm, 'tanka' to harvest water, 'jupka' and 'kothi' for storing the grain and feed, etc. Beside these, the people of both the ecosystems observed the movement of insects and animals (butterfly, ant, and termite) to forecast the rainfall and other climatic parameter. Both the ecosystem were highly vulnerable to climate change. Major adaptive strategy included promotion of climate resilient varieties like Tropical Beauty, Michael, Red Chief etc. of apple, short duration varieties of pearl millet (MH-179), cluster bean (Maru), moong bean (S-8, K851) etc; development of community level institution, capacity building programme on climate proof technologies and devising innovative communication strategies for adaptation to climate change. Inclusion of socio-economic factors and personality characteristics of farmers are essential for formulating any development strategy (Sarkar, 2015).

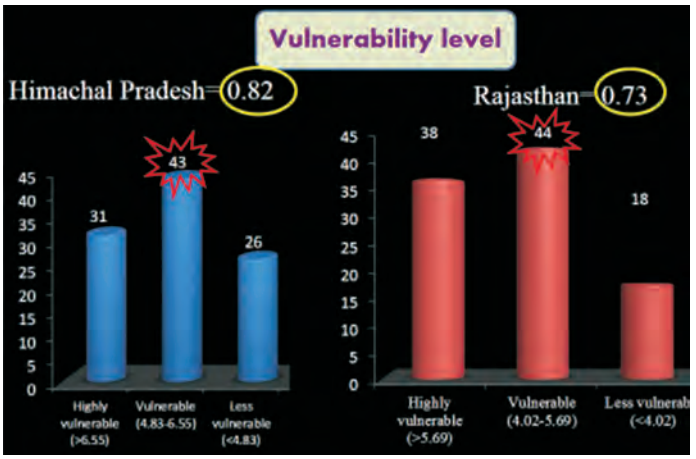


Fig. 12: Comparative vulnerability profile of Himachal Pradesh and Rajasthan

The farmers under NICRA interventions and without interventions in Gumla and Tumkur districts from Jharkhand and Karnataka states, respectively were well aware on aspects of climate change but NICRA farmers had a higher level of awareness on climate resilience (78.83) which was lacked by non-NICRA farmers (23.85). Soil health card, farm ponds, checkdams and its renovation, redgram varieties BRG1and BRG2, ragi-redgram cropping system, preventive vaccination of animals were the most adopted interventions under various categories recommended by NICRA. The most important factors responsible for adoption climate resilient technologies among farmers were experience in farming and leased-in land area. The cropping intensity, crop diversification, knowledge level and resilience were much higher for beneficiaries than non-beneficiaries. A considerable significance in difference of crop diversification, land area irrigated during *kharif, rabi* and summer, *rabi* and summer irrigation frequency achieved between NICRA and non-NICRA farmers. A significant increase in employment, yield of crops and savings of beneficiaries occurred on a longitudinal time basis. KVKs are involved in a variety of activities for adopting adaptive strategies. Input delivery services (7.21) of KVKs were proved to be highly useful. Financial (1.76) and marketing assistance (1.88) were few missing links of NICRA. Trainings on climate resilience recorded highest satisfaction (6.43) among farmers. The officials

evaluated technical constraints, and labour and economic constraints as the most severe, where as farmers did not considered so (Jasna, 2015).

Impact Assessment of Developmental Programmes and Policies

A study of Indira Kranti Patham–Non-Pesticidal Management programme; initiated by the Society for Elimination of Rural Poverty (SERP) in collaboration with of Centre for Sustainable Agriculture (CSA), in Andhra Pradesh for Crops in 12 districts in 2005-06; showed that NPM practices and strategies for pesticide-free cultivation diffused through community mobilization. It involved community managed extension work with active participation of women's self help groups and their federations, complete elimination of chemical pesticides in crop cultivation, sustainable agricultural practices and adequate organizational support with management linkages at village, mandal and district levels. All NPM practices were sustainable with respect to ecological, economic and socio-cultural dimensions of sustainability. Ninety-five per cent of the respondents perceived that the overall impact of NPM practices was high on improvement in crop health, ecological health, human health, and health of animal and birds. Farmers faced constraints in getting seeds of traditional varieties, and found that NPM practices were labour consuming, and the NPM produce does not fetch premium prices (Malgatti, 2008).

A study on Implementation of the National Rural Employment Guarantee Programme in West Bengal- A critical analysis reported that significant changes were found in the level of aspiration, self-confidence, self-reliance and overall empowerment of the respondents after commencement of the program and most of them were found to have shifted from low level to medium level of livelihood security . Late payment of wages and selection of low priority or inappropriate works were some of the problems as perceived by beneficiaries (Roy, 2010**)**.

The livelihood security of tribals of Malbazar and Madarihaat blocks of Jalpaiguri district in West Bengal had changed significantly before and after implementation of the Tribal Sub Plan on Livelihood Security programme. The mean ranks before and after the implementation of the programme were significantly different in areas of food security (Before: After =3.60:4.36), occupational security (2.13:5.14), habitat security (1.19:2.85), educational security (1.11:3.14), and social security (1.00:1.74). In case of health security, no significant change was observed. More men participation as compared to women participation in various activities of TSP was observed and the difference was found to be significant (Female 34.23; male 38.79). A shift in occupation from agriculture and allied sectors to business and other secondary sectors was found to be significant. Increasing tribal migration for easy available work and higher income had become severe. Among the several constraints of successful execution of TSP economic constraint (Mean rank: 464.92) was found to be most severe while, infrastructural (268.85) and social constraint (222.172) were rated as severe, moderately severe and not so severe, respectively (Barua, 2012).

A study on accessibility to quality seeds showed that there was no statistically significant difference ($p>0.05$) between the farmers of AP and Bihar with respect to variables such as proximity to seed source, physical and genetic purity of seed purchased from formal sources and farm saved seed. The major source of purchased seeds in both the states was dealer/retailer at nearby tehsil/mandal/town headquarters. The physical and genetic purity of certified/quality seeds purchased from market in both the states was moderate whereas the quality of farm saved seeds in both the states was very high. The difference between farmers of AP and Bihar was statistically significant with respect to variables such as timeliness in availability of seeds, availability in adequate quantities, credibility of source of seed and price of seeds ($p<0.01$); crop performance, and documentation and record keeping ($p<0.05$). The farmers in AP had significantly higher access to timely availability of seeds in adequate quantities from credible sources. The prices of purchased seeds were significantly higher in AP when compared to Bihar. Overall accessibility to quality seed in both the states was found to be medium for majority of the farmers. The farmers, Agriculture Department Officials (ADOs), Civil Society Organizations (CSOs),

Private Seed Companies (PSCs), researchers and seed dealers strongly demanded for regulation of retail price and trait/royalty fee over seeds by state governments (Manjunath, 2103).

Most of the non–beneficiaries of Bringing Green Revolution to Eastern India (BGREI) programme under RKVY in district Bhagalpur of Bihar and Cooch Behar district of West Bengal belonged to low awareness categories and the main reason for this was assigned to the distance between their home and the agricultural departments. Majority of the beneficiaries had a high level of awareness, since they were in constant touch with the State department of agriculture, KVKs etc. Pearson's coefficient of correlation showed that in case of rice, age and land holding negatively affected the adoption of improved cultivation of rice. In case of wheat, age and family size affected the adoption behavior negatively and in case of maize, age, family size and land holding affected the adoption behavior negatively. It was observed that as the land holding increased, people shifted more towards diversification and the relative proportion of single crop on the land decreased. It was also observed that higher the land holdings, lesser was the adoption of SRI methods due to the fact that it was labor intensive and large farmers were unaware about the result of the technique. Due to BGREI there was 32.70 % increase in the cropping intensity among the beneficiaries, while there was 5.26% increase in the cropping intensity of the non–beneficiaries too. This slight increase may be due to factors like, visit to crop demonstration plots and communication with the beneficiaries. The most common problem faced by the agriculture departments during implementation of the programme was the inability of the farmers to adopt new technologies due to their economic condition. Extension personnel suggested that these constraints could be overcome by increasing the subsidy on the farm implements and need based dissemination of new technologies (Bagdas, 2015).

The area under food crops had decreased during 2001 to 2012 period and among food crops rice recorded maximum reduction in area. Area under food crops showed a comparatively high value of negative growth (2.95%) in area than that of non- food crops (-0.92%). Reduction in area under paddy cultivation showed significant and negative correlation with age, experience in rice cultivation, extension contact, education, crop yield security and input self-sufficiency. Among psychological variables, farming commitment, value orientation and connectedness to nature showed significant and negative correlation with reduction in area under paddy cultivation. Experience in rice cultivation, education, input self -sufficiency, crop yield security and awareness about ecosystem services were identified as the significant predictor variables of reduction in area under paddy cultivation. Underlying reasoning behind implementation of the legislation was the irrational and uncontrolled reclamation and conversion of paddy land and wetland taking place in the state. There was 0.8 per cent reduction in loss in area was as an impact of legislation. Majority of the respondents agreed that the legislation could bring 'more Clarity on regulation of human interventions in wetlands and legislation could bring increased awareness about the importance of wetland conservation. Majority had the opinion that Local Level Monitoring Committee (LLMC) in every village initiated a method of participatory resource conservation. Growth in seafood business of rich people, water shortage during summer for more than 2 months and drought conditions in the region were the most important consequences of changed cropping pattern and land use pattern occurred in three representative regions of Kerala (Rejula, 2015).

Impact Assessment of National Food Security Mission (NFSM) among small and marginal farmers of Uttar Pradesh revealed that eighty per cent of NFSM wheat farmers had high level of adoption, whereas 43.3 per cent of NFSM rice farmers and 56.67 per cent of NFSM pulse farmers had high level of adoption. The variables like education, social participation, communication pattern, economic motivation and scientific orientation were found to have significant correlation with the extent of adoption. Regression analysis showed that farmers' scientific orientation in case of rice and pulse farmers and social participation in case of wheat farmers were found to have significant contribution to the extent of adoption. The scheme had a great impact on beneficiaries in terms of increase in yield, income, savings, expenditure pattern, possession of assets, and increase in employment days, house hold food security, education and housing quality. Constraint analysis with garret ranking showed non-availability of critical inputs in time as a major technological constraint, while a repeated benefit to the same farmer was found to be the major administrative constraint. Some of the

strategies suggested include: planning for timely availability of inputs, decentralization of procurement process to the district level, creation of production units of quality bio- fertilizers and bio-pesticides, establishment of single window input supply centres for cluster of villages under NFSM and strict norms to reduce political interference (Nayak, 2014).

The Karnataka Comprehensive Nutrition Mission (KCNM) was perceived as 'effective' by a majority of beneficiaries as it achieved considerable success through its interventions, innovative strategies and multi-sectoral convergence. Training on establishing Kitchen Garden' was an effective agri-nutri linkage intervention. Farmers were willing to adopt Nutri-Farming System based on opinion of their peer group, relatives, Village Panchayat Chairperson. Since the subjective norms were the most influencing factors for farmers' intention to adopt Nutri-Farming System, a participatory mode in a conducive and favouring social context under multi sectoral approach linking agriculture to nutrition and gender is very important. However, consumption of bio fortified foods was mainly influenced by the Perceived Behaviour Control. It indicates that though the items under dietary basket were influenced by the social norms, the type of food item one wants to consume was a private choice (Motagi, 2016).

Agricultural Research and Education Appraisal

Analysis of work styles, best practices and productivity of agricultural scientist brought forth that the productivity of a majority (88%) of the agricultural scientists from the Low Performing Institutes (LPI) was low to very low and only 5% of them were high in productivity. In case of the High Performing Institutes (HPI), only 6% of the scientists had very low level of productivity and 28% of them had higher level of productivity. Significant correlation between research productivity and different work style variables could be observed. A step-wise multiple regression analysis produced significant regression coefficient for all the work style variables except for supportiveness. The value of adjusted coefficient of determination (R^2) was 0.83. Being creative and innovative, maintenance of deadline, maintaining originality in research, participatory decision making, interactive teaching etc. were among the best practices followed by highly achiever agricultural scientists (Paul, 2012).

Out of total 42 documented best practices for Teaching-Learning in Higher Education in agriculture, 17 practices were perceived as important by the respondents. The factors promoting best practices, revealed through Principal Component Analysis, were leadership, receptivity, pragmatism, pro-activeness, team building skill, communication skill, rationality, and mentoring and dedication for teaching. The motivational analysis of organizational climate revealed that it was achievement-affiliation climate profile for ICAR-IARI; for ICAR-NDRI and UAS it was expert influence-affiliation climate profile; and, for TNAU it was extension-expert influence climate profile. Teachers of all universities, except UAS, agreed that they do lack skill in making use innovative and interesting ways for their students to learn. Forecasting potential training areas for teachers was done using Delphi technique; in which; after two rounds, there was consensus between the experts, except few areas. Some of the important areas which emerged for training were effective communication skills, maintaining professional conduct and ethics in one's profession, classroom management skill etc. An e-learning module was developed and the proto-type module was tested with 60 and validated with 150 respondents. Their responses indicated that the module was well perceived by them. The results showed a general acceptability of the module in terms of the criteria's taken into consideration. The strongest predictor of the teachers' intent to use or develop e-module was subjective norms. It indicates that social pressure played an important role in predicting use or development of e-modules by teachers (Balakrishnan, 2015).

The Experiential Learning programme of ICAR for Entrepreneurial Development has been perceived useful by both students and teachers in imparting entrepreneurial skills and competencies. It allows students to go through all stages of entrepreneurship education. Significant differences of usefulness of the programme as perceived by students from different disciplines were noted. Generally there were no significant differences on perceived utility on the basis of personal characteristics of respondents, except for students' sex and age.

Students had little intention of venturing into self-employment whereas the teachers indicated that none of their graduates had established an enterprise. Even though the programme has been perceived useful in equipping graduates with the necessary entrepreneurial skills and competencies, graduates were not yet motivated to venture into self-employment hence the stages are not being completed as revealed by the perceptions. Extra curricula activities were perceived to have been rarely used by both respondents while significant differences were noted on perceived use of some extra curricula activities by students. No support was provided by institutions to graduates who wished to venture into self-employment. Students perceived experiential learning programme utility to be predicted by administrative factors, teaching methods, caste and role model, while background of teachers and extra curriculum were perceived by teachers. The performance of the programme can therefore be attributed to a diverse curriculum, its inclusion in four year BSc programme, lack of extra curricula activities, institutional support for graduates and lack of trained teachers in the area of entrepreneurship education. The programme therefore need to be reviewed taking into consideration the timing that is when it can best be implemented, academic qualification of the staff involved and the structures that need to be put in place at institutional level (Lekang, 2016 and Lekang, *et.al*. 2016).

Summary

- The message effectiveness of the community radio was effective and led to social and economic changes but lack of incentives and infrastructural facilities were constraints.
- The determinants of e-Learning behavior of farmers of Kerala were identified as the years of education and years of experience with computer
- The videos produced by Digital Green caused significant difference in production and income for all the farmers
- E-learning modules can help to learn new concepts, create interest, serve as ready reference material and be a tool for distance education of extension personnel
- Lack of training, poor infrastructural development and poor network connectivity were the major barriers of adoption of ICT based extension services.
- Implementation of Agricultural Technology Management Agency ensured wheat farmers in Ahmednagar district of Maharashtra higher per cent of increase in yield per hectare; high returns; and high income.
- *Samaj Shilpi Dampatis*, a newly wed graduate couples who live and work with villagers and act as nodal point for all rural development interventions is an innovative and effective extension strategy.
- The paid extension services were found moderately effective by 50 per cent and the rest perceived them to be highly effective.
- Rural extension sub-centres with private professionals evolved by *Mitra Niketan*in, Kerala is a successful alternative extension mode.
- The private extension support system had better timeliness in delivery of services as compared to the public extension support system. There was no significant difference between the level of effectiveness in fairness and competency of private and public extension support system for orange production in Maharashtra.
- Farmers Field School (FFS) had positive impact on enhancement of knowledge and yield.
- IARI-Voluntary Organization partnership based extension was found effective and similar Public-private partnership based could be promoted.
- IARI-Post office Linkage Extension model was found effective in technology dissemination at lesser time lag and quality assurance.
- Major factors affecting the job performance of women extension personnel were related to organizational, personal, sociological, and psychological domains.

- The future competencies required by the Extensionists in changing agricultural scenario would be know-how skills like international know-how, technical know-how and human know-how skill
- The marketing constraints for innovators in scaling up, technical constraints for non-innovators in adoption of innovations and administration constraints for officials in scaling-up of farmer-led innovations were most severe constraints related to farmers' innovations
- There is a need for proper documentation and validation of ITK by in-depth analysis of ITK and sustainable crop biodiversity conservation in several agro ecological systems.
- The constraints related to certification and marketing aspects had a major role in creating obstacles to promotion of organic farming and its spread in the state.
- A majority of farmers were satisfied with vegetable retailers and were also satisfied with the overall performance of the collection centers, payment method, and the prices offered by the collection center.
- Processor's margin, marketing cost, marketing margin, price spread and producer's share in consumer's rupee varied with the length of marketing channel. Price spread was lesser and producer's share in consumer's rupee was higher for shorter marketing channels with less number of intermediaries and *vice-versa* for lengthier marketing channels.
- The average savings have increased drastically to the tune of more than Rs 60,000 per family per year in Karnataka and Maharashtra states due to adoption of polyhouses..
- Access to guaranteed market, access to market information, membership in organizations and education level were highly correlated with the decision to take or not to take post harvest and value addition interventions.
- Content analysis of 20 case studies revealed that innovativeness, social network and risk taking ability were the most common and essential attributes of agripreneurs.
- The benefit cost ratio (B: C) was found higher and statically significant in watershed villages with B: C ratio of 1.71: 1 and 1.9:1 for wheat and mustard, respectively as compared to 1.19:1 and 1.52:1 for wheat and mustard in non-watershed villages.
- The comparative analysis shows that mean transactional input cost per hectare was found to be significantly lower for members than non-members of Tamil Nadu Mango Growers Federation (TAMAFED) and similarly the members of MAHAGRAPE saved 37 per cent of input transaction cost than the non-members.
- The higher Benefit-cost ratio (2.6:1) for Bt. cotton as compared to non-Bt, cotton (1.6:1) showed the profitability of cultivating Bt. Cotton in Rajasthan and the average yield increase through adoption of Bt cotton was 42 per cent in Gujarat and 45 per cent in Karnataka.
- Agro-biotech risk communication analysis revealed that print media played limited role in imparting education and creating awareness about agro-biotech crops and portrayed the subjective concerns of non-expert stakeholders.
- The benefit cost ratio for zero tillage (2.31) and laser land leveling (1.94) was higher than the conventional system. The average technical efficiency of adopter farmers was about 74 per cent and non-adopter was 44 per cent.
- Factor analysis identified six different factors governing progressiveness of farmers namely scientific temperament, perseverance and hard work, cosmopoliteness, achievement motivation, entrepreneurial aptitude and work satisfaction.
- Increase in poverty was the major perceived risk of climate change under socioeconomic and cultural life by the farmers of Sunderbans in west Bengal. It was observed that people devised different indigenous techniques or practices to overcome the adverse impact of climate change in their farming.
- Being creative and innovative, maintenance of deadline, maintaining originality in research, participatory decision making, interactive teaching etc. were among the best practices followed by highly achiever agricultural scientists.

- The factors promoting best practices, revealed through Principal Component Analysis, were leadership, receptivity, pragmatism, pro-activeness, team building skill, communication skill, rationality, and mentoring and dedication for teaching.
- The Experiential Learning programme of ICAR for Entrepreneurial Development has been perceived useful by both students and teachers in imparting entrepreneurial skills and competencies

Future Perspective

- Assessment of innovative extension approaches and e-extension
- Development and validation of e-learning modules on competencies of extension professionals in changing context of agriculture.
- Documentation of farmers' innovation and identification of innovation generation process and framework for upscaling.
- Technology mapping and assessment of extension impact
- Analysis of processes in incubation and uptake of entrepreneurial ventures and start-ups.
- Inventorisation and impact analysis of institutional innovations in extension.
- Analysis of consumer preferences of organic and bio-fortified foods
- Analysis of innovations in training and teaching technologies
- Climate change adaptation and Risk adjustment in agriculture
- Organizational climate of extension and training institutions

Acknowledgement

I sincerely acknowledge the cooperation extended by all the faculty members as well as M.Sc and Ph.D. students of the Division of Agricultural Extension in preparation of the significant contributions of the postgraduate research. Sincere thanks are also due to Shantanu, Alok, Krishna, Rabeesh, Sunil, Shruti and Rimpi.

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Agricultural Statistics

Seema Jaggi and UC Sud

Introduction

ICAR-Indian Agricultural Statistics Research Institute (ICAR-IASRI), an ISO 9001:2008 certified Institute, is mainly responsible for conducting research and education in Agricultural Statistics and Informatics to bridge the gaps in the existing knowledge. The Institute has used the power of Statistics blended judiciously with Informatics and has contributed significantly in improving the quality of Agricultural Research. The Institute has also been providing education/ training in Agricultural Statistics and Informatics to develop trained manpower in the country. The research and education is used in improving the quality and meeting the challenges of agricultural research in newer emerging areas. The Institute has made some outstanding and useful contributions to research in Agricultural Statistics and Informatics.

The functions and activities of the Institute have been re defined from time to time. The basic, applied, adaptive and strategic research in Agricultural Statistics and Informatics is carried out under following broad programmes that encourage interdisciplinary research:

- Development and analysis of experimental designs for agricultural systems research
- Forecasting, modelling and simulation techniques in biological and economic phenomena
- Development of techniques for planning and execution of surveys and statistical applications of GIS and remote sensing in agricultural systems
- Development of statistical techniques for genetics/computational biology and applications of bioinformatics in agricultural research
- Development of informatics in agricultural research

The Institute has been playing the role of a leader in research and education in Agricultural Statistics in the country. The contributions of the Institute towards the education and training in Agricultural Statistics have been tremendous and outstanding. The teaching of post graduate programmes of the Institute [the planning, organization and co-ordination of M.Sc. and Ph.D. (Agricultural Statistics) initiated in 1964-65] is carried out in collaboration with PG School, IARI, New Delhi. The Institute has so far produced 192 Ph.D. and 336 M.Sc. students in Agricultural Statistics. Many of the students occupy prestigious positions in India and abroad. The Institute also conducts Senior Certificate Course, a practical oriented agricultural statistics course with greater application of computer usage for the officials of various government/research organizations. The Institute has been identified by ICAR as Centre for Advanced Faculty Training (CAFT) in Agricultural Statistics and Computer Applications. It is known for its ad-hoc and customized training programmes in these areas for public and private sectors. The entire campus is Wi-Fi enabled with a high speed internet connection.

Significant Student's Research Achievements

The broad areas in which the students undertake research are designing of experiments for different agricultural research situations, sample surveys for estimating the various parameters of the crops, statistical modelling and forecasting, statistical genetics/ genomics, geo-informatics and spatial modelling. During the

period of 2007-16, 64 M.Sc. and 33 Ph.D. students including two foreign students, one each in M.Sc. and Ph.D., have completed their degree in the discipline of Agricultural Statistics. They have undertaken research on the above broad areas and their significant research contributions are as given below.

Designing of Experiments and Analysis of Experimental Data

Experimentation is an essential component of any scientific investigation and designing an experiment is an important part of it. Designs are usually characterised by the nature of grouping of experimental units and the procedure of random allocation of treatments to the experimental units. Designed Experiments are conducted in the field of agricultural and other allied sciences. It is through the data collected from designed experiments that the valid inferences are drawn. The objectives of the experiment or the problems to be solved may be of some specific type of comparisons among the treatments. For example, interest may be in all the possible pair-wise treatment comparisons. In some other situations, comparison has to be made of some new treatments (test treatments) with one particular established treatment (control) and so the tests versus control comparison are of interest. Similarly, when the treatment structure is factorial in nature, some special treatment contrasts known as factorial effects are of interest. The design to be chosen is based on the specific problem. The data generated from a designed experiment is analyzed using analysis of variance. The research carried out by the students in the field of design of experiments is on a wide range of topics such as efficient and robust block designs for making all possible pair-wise treatment comparisons, designs for making test treatment control comparisons, computer aided search of efficient designs, row-column designs, designs for breeding trials, designs in the presence of neighbour effects, response surface designs, change over designs, design and analysis of multi-response experiments, design and analysis of micro array experiments, robustness of designs, etc.

The block design assumes additivity of treatments and bocks. The contributions of block and treatment effects to the response are independent and additive. The computation procedure for testing the assumption of additivity in block design has been developed and illustrated with an experimental data generated using Balanced Incomplete Block (BIB) design (Das, 2011). In Agricultural Field Experiments Information System data base at IASRI, plot wise data relating to 6287 experiments conducted using Randomized Complete Block (RCB) design is available and same has been utilized for (i) testing of assumption of additivity of effects (Tukey's one degree of freedom test) and (ii) testing of assumption of independence of errors (Run test). The assumption of additivity does not hold in 10.85% of experiments whereas in 11.07% experiments, assumption of independence of errors fails. In presence of these disturbances alternate procedure of analysis has been given.

In designed experiments, the assumption of independence of observations gets violated because of various types of dependence among the observations within a block. Also the departures from the basic assumptions may take place in presence of disturbances like outlier(s). Cook-statistic and AP statistic has been developed in general for any t outliers in block designs with correlated errors (Ojha and Bhar, 2014; Ojha and Bhar, 2015) considering Auto-regressive [AR(1)] and equi-correlation structures. Developed statistics are illustrated with real experimental data. A robustness criterion has been proposed for block designs with correlated errors and block designs that are robust by this criterion have been identified.

Outlier is defined to be an observation that is inconsistent with the rest of the data. If the data set contains outliers, then the conclusion drawn from the experiments may be wrong. Outliers may arise in the experimental setup due to pest attack in some particular plot or mistakes may occur during recording of the data. Therefore robust method of estimation of parameters is advocated. The most important robust methods are M-estimation and Least Median of Squares (LMS) estimation. Paul and Bhar (2014) applied LMS technique for robust Analysis of experimental data. In M-estimation, a function of errors in minimized whereas in least squared the error sum of squares in minimized. In the least squares estimation all the observations are given equal weight and that is unity. But if any observation is found to be outlier in the data set then it must get some lesser weight than that of the normal observations. This concept is utilized in the M-estimation. Paul and Bhar (2012) studied

M-estimation in block designs. Different functions of robust estimation are available in the literature that is extensively used in the linear regression model. These functions are made used in the experimental setup also by slight modification. Paul and Bhar (2012) proposed a new objective function for its application in the design of experiments. It was observed that the newly proposed function performs very well in presence of outliers in the sense that it will produce minimum average variance of a set of orthonormal contrasts in most of the cases. Paul *et al.* (2015) performed robust analysis of block designs.

Scientific experimentation requires decision about how to conduct and analyse an experiment. These decisions depend on the purpose of the experiment, but certain choices may be restricted by available resources. Prior information may be available from earlier experiments. The Bayesian approach provides a framework where prior information and uncertainties regarding unknown quantities can be combined to find an experimental design that optimizes the goals of the experiment. Bayesian approach can be used wherein the problem starts with the formulation of a model that is adequate to describe the situation of interest. Then a prior distribution over the unknown parameters of the model is formulated, which is meant to capture the beliefs about the situation before seeing the data. In the literature, there are several distributions available which can be taken as a form of prior information. These are divided into two category namely conjugate and non-conjugate family. Under conjugate family, Normal-Gamma density and Normal-Chi density have been considered for approximating the factors contributing for block design and variance parameter respectively. In case of non-conjugate family, distributions taken are multivariate–t and Gamma for assignable and non-assignable causes respectively. The developed procedures have been applied to real experimental data set pertaining to Long-Term Fertilizer Experiments conducted under AICRP on LTFE (Prasad and Bhar, 2013). It was observed that some of the treatment pairs which were not significant in classical analysis became significant after Bayesian analysis of the same set of data with the use of respective prior information. In some of the cases block effects became significant after applying the Bayesian method.

Experiments in which data on several responses are measured from an experimental unit corresponding to the application of a treatment are known as multi-response experiments. There may be complete multi-response experiments (all the response variables are recorded from each experimental unit) and incomplete multi-response experiments (recording of all the response variables from each experimental unit is not feasible). A stepwise procedure of performing multivariate analysis of variance (MANOVA) of data from complete multi-response experiments conducted in block designs has been developed. The procedure of multivariate treatment contrast analysis has also been developed. To tackle the problem of outlier(s) in multi-response experiments, a test statistic has been developed by Parsad *et al.* (2008) for detection of a single outlier vector in complete multi-response experiments run in a block design. A method of construction of designs for incomplete multi-response experiments is also obtained using a combination of RCB designs and BIB designs. To accommodate more number of response variables, one can use singular group divisible design as response-wise design instead of a BIB design. The designs obtainable from this method are economical from resource point of view. For the experimental situation where it is desired to determine a combination of levels of input factors that simultaneously optimize all the response variables, the procedure for estimation of parameters in incomplete multi-response situation and the procedure of simultaneous optimization of several response variables has been investigated (Nandi *et al.*, 2015). It is very much possible to lose some observations due to some unforeseen causes in case of multi-response experiments. The property of an original design may change due to presence of missing observations. The robustness of block designs against missing data in multi-response experiments has been studied and robust designs have been catalogued (Dasgupta, 2012). Bhar and Ojha (2014) have studied outliers in multi-response experiments.

The objective of a breeding programme is to create variability and to select genotypes for raising or for further breeding purpose. The most common designs used by a geneticist to study genetic parameters and their interpretations are diallel (single cross), triallel (three-way cross) and quadriallel (double cross). Three-way crosses are intermediate between single and double cross hybrids with respect to uniformity, yield, stability and

the relative simplicity of selecting and testing and hence are increasingly gaining popularity among breeders. Considering a full model including general and specific combining ability effects and a reduced model ignoring specific combining ability effects, the information matrices for the estimation of combining ability effects have been derived for designs involving three-way crosses. Four methods of constructing designs for breeding trials involving complete/ partial three-way crosses have been developed (Harun *et al.,* 2016a). Harun *et al.* (2016b) obtained triallel cross designs for comparing a set of test lines with a control line.

There may be experimental situations where several new lines (called the test lines) are developed in the initial stage of an experiment and it is expected that only a few of the new lines may be worthy of further investigation. The new lines may first be compared with one (or more) control line(s) that are already being used by the experimenter in order to screen out the new lines for further investigation. Block designs involving complete/ partial diallel crosses for comparing test lines with control line(s) are suitable when the experimenters are interested to estimate test vs. control comparisons with respect to their gca effects with a minimum variance. Some families of small and efficient block designs for diallel cross experiments for test vs. control comparisons have been constructed (Srivastava *et al.*, 2013). Further, for experimental situations where resources are limited so that a complete diallel is not feasible, but some lines are believed to be superior and are therefore of prime interest, augmented partial diallel cross (APDC) plans are suitable. Some classes of APDC plans have been obtained using various association schemes of partially balanced incomplete block designs (Srivastava *et al.*, 2015).

Neighbour-balanced designs, wherein the allocation of treatments is such that every treatment occurs equally often with every other treatment as neighbours, are used when the treatment applied to one experimental plot may affect the response on neighbouring plots besides the response to which it is applied. These designs ensure that no treatment is unduly disadvantaged by its neighbours and help in estimating the neighbour effects besides the direct effects of treatments. Under the neighbour balanced block (NBB) design setting, it may sometimes be desired to compare a set of test treatments with a control or a set of controls. Some series of NBB (complete and incomplete) designs for comparing a set of test treatments to control(s) have been developed [Abeynayake *et al.* (2011); Abeynayake *et al.* (2012)]. In experimentation using NBB designs, there may arise the possibility that some of the observations could become unavailable for analysis. Abeynayake *et al.* (2011) examined the robustness of NBB designs against missing observation(s) and the efficiency of these designs have been calculated. The NBB designs are found to be robust/fairly robust against missing of observations. Complete NBB designs and incomplete partially NBB designs for two factors have been constructed that are balanced for one factor (Jaggi *et al.*, 2010). The situation where heterogeneity is present in the experimental material in two directions, row-column designs are used. Methods of constructing neighbour balanced row-column designs have been developed and their characterization properties have also been studied (Varghese *et al.*, 2011, Varghese *et al.*, 2014).

In agricultural field experiments, blocks are often formed using adjacent plots within a field. The experimental plots occurring close together within blocks may therefore be correlated. Series of neighbour balanced complete/ incomplete block designs, permitting the estimation of direct and neighbour effects, under non-directional and directional neighbour effects model and dependent observations have been obtained (Tomar and Jaggi, 2007). A catalogue of all the designs giving parameters and efficiencies for direct as well neighbour effects under AR(1) and nearest neighbour (NN) error structures has been prepared for different values of correlation coefficient. Jaggi and Tomar (2005) have studied the robustness of neighbour balanced complete block designs for correlated observations.

Block model with interference effects from the neighbouring units up to distance 2 (or second order) has been studied. Some classes of balanced block designs with neighbour effect upto distance 2 have been obtained and their characterization properties have been studied [Bhowmik *et al.* (2012); Bhowmik *et al.* (2013)]. Strongly balanced block designs with effects from immediate neighbouring units have been identified to be universally optimal for separate estimation of direct effects and interference effects (left and right) of treatments under

the non-additive model (Bhowmik *et al.,* 2015a). Besides interference effects, there may arise situations where trend effects may also affect the plots within block. Block models with interference effects from both adjacent neighbouring units and from neighbouring units upto distance 2 incorporating trend component have been studied [Bhowmik *et al.* (2015b); Bhowmik *et al.* (2015c)].

When the heterogeneity present in the experimental material is from two cross classified sources, then two-dimensional blocking of the experimental units is recommended for control or reduction of experimental error. Some general methods of constructing balanced treatment-control row-column designs in complete/incomplete rows/columns have been developed (Sarkar *et al.*, 2013). Some families of symmetric/asymmetric factorial row-column designs in complete/incomplete rows/columns have been obtained. Most of the row-column designs developed in the literature have only one unit corresponding to the intersection of row and column. However, for the instances when the number of treatments is large with limited experimental resources, Generalized Row-Column (GRC) designs are used where there is more than one unit in each row-column intersection. Some methods of constructing GRC designs have been developed that are structurally complete, i.e. all the cells corresponding to the intersection of row and column receive treatments. Datta *et al.* (2015) developed some series of row-column designs with multiple units per cell. Datta *et al.* (2016) constructed series of incomplete row-column designs with two units per cell. Some series of structurally complete GRC designs with unequal cell sizes and unequal replications of treatments have also been developed. Datta *et al.* (2014) developed some structurally incomplete row-column designs with multiple units per cell. Datta (2016) studied generalized row-colum designs for single and multi-factor experiments. Robustness of different classes of GRC designs against missing of one or more observations has been investigated.

There may arise experimental situations where it is desired to compare treatments belonging to two disjoint sets and the interest is to estimate the contrasts pertaining to treatments from different sets with as high precision as possible. Balanced Bipartite Generalized Row-Column (BBP-GRC) designs have been defined and series of BBP-GRC have been developed in which the contrast of first set versus second set of treatments is estimated more precisely. Generalized confounded row-column (GCRC) designs, generalized partially confounded row-column (GPCRC) designs and fractional GCRC designs have been developed which ensure that all lower order interactions including main effects are estimable.

When heterogeneity present in the experimental material is from three sources, then three-way blocking of the experimental units is recommended for control/ reduction of experimental error. For these situations, experimental designs under three-way blocking structure can be advantageously used. These designs are capable of marking out stony patches and other features that tend to clump in compact areas or similarity that may arise due to same group of housing of animals. Series of designs under three-way blocking structure have been developed for regular and irregular regions (Kumar *et al.,* 2015a). Kumar *et al.* (2015b) have constructed irregular Sudoku-type designs for animal experiments.

Latin hypercube design (LHD) is a way to generate design points that can spread observations evenly over the range of each input variable. LHDs are used where physical experimentation is not feasible. Latin hypercube designs (LHD) are widely used as space-filling designs in the field of computer experiments. Choice of a good LHD closely depends upon space-filling and orthogonality criteria. Space-filling criterion provides maximum coverage to the whole design space and orthogonality criterion helps to estimate effects independently. These two criteria are not dependent upon each another. A good space-filling LHD may not be orthogonal and vice-versa. A method has been developed for construction of LHDs for two factors and any numbers of runs which provides good space-filling property. Methods of constructing orthogonal LHDs for two and three factors have also been developed for any number of runs that helps to estimate effects independently. Mandal *et al.* (2016) obtained orthogonal Latin hypercube design with special reference to four factors.

Designs in which each experimental unit receives some or all of the treatments, one at a time, in successive periods are known as change-over designs (CODs). The distinguishing feature of these designs is that the

treatments applied in a particular period influence the responses of the experimental units not only in that period but also leaves residual effects in the succeeding periods. These designs are of special importance when the experimenter finds it difficult to obtain homogeneous experimental units or when the experimental units are expensive. These designs have been advantageously used in animal nutrition experiments, clinical trials in medical research, long-term fertilizer experiments in agriculture, educational studies, bioequivalence trials, *etc*.

In many experimental situations, it is often required to measure the effect of response from two unrelated factors in the presence of first order residual effects of both the factors. Three series of CODs with two non-interacting factors out of which two are symmetric factorial and one is asymmetric have been proposed (Varghese *et al.*, 2009). To meet experimental situations involving simultaneous application of levels of two factors when one of the factors exhibits residual effects while the other does not, two classes of uniform strongly balanced CODs with two factors balanced for residual effects of levels of one factor have been obtained by Dwivedi, *et al.* (2008). These designs are variance balanced in the sense that the contrasts pertaining to direct effects of treatment combinations consisting of various levels of both the factors are always estimated with a constant variance. Sharma *et al.* (2010) constructed two new classes of minimal strongly balanced CODs assuming the presence of first order residual effects.

Universal optimality of two classes of balanced CODs have been established for the estimation of direct, first and second residuals. Besides, the double-extra-period balanced CODs, obtained by repeating the treatments of the last period in two succeeding periods for the first class and shown to be universally optimal for the estimation of second order residual effects. Lastly, optimal two-treatment CODs are obtained using efficiency factors, when the errors are auto-correlated, considering (i) first order residual effects of treatments and (ii) first and second order residual effects of treatments. Moreover, a class of two-period totally balanced trend-free CODs considering presence of first order residual effects has been developed. A method of construction and analysis of these designs has been given along with an illustration. Universal optimality of these two-period totally balanced trend-free designs has been established for the estimation of contrasts among direct effects and among first order residual effects.

Bioequivalence is the degree to which clinically important outcomes after receiving a new formulation resembles a well-established formulation. Evaluation of veterinary medicinal products is one of the important areas where bioequivalence trials are conducted. Varghese *et al.* (2010) studied repeated measurements designs for bioequivalence trials. In bioequivalence trials, the main interest of the experimenter lies in making test vs. reference formulation comparisons with as much precision as possible and comparisons within test formulations are not of much interest. Hence designs that give estimates of test vs. reference formulations comparisons with maximum precision are a smart choice for bioequivalence trials. Some series of balanced/ partially balanced designs for bioequivalence trials in incomplete sequences have been presented for experimental situations in which the experiment cannot be continued for more number of periods. Further, some series of balanced/ partially balanced designs for bioequivalence trials in complete/ incomplete sequences assuming the presence of residual effects have also been proposed. In experimental situations where a set of test formulations are to be compared with another set of reference formulations, two classes of designs for bioequivalence trials have been constructed assuming residual effects of formulations to be present (Sonawane *et al.*, 2011).

Sensory trials are included as an integral part of food and nutrition experiments involving agricultural/ animal produce to demonstrate some sensory fact. In sensory trials involving large number of products, there are mainly two operational constraints that limit the choice of experimental design. On one hand, the assessor constraint sets a maximum number of products that an assessor can evaluate within a session before onset of sensory fatigue. On the other hand, preparation constraint limits the number of products that can be prepared for a given session without loss of experimental control. Therefore, it is many times necessary to split sensory evaluations into sessions. Methods to construct designs for multi-session sensory trials with/without carry over effects are developed. Series of treatment vs. control designs for multi-session trials have been obtained to deal with such situations (Saurav, 2016).

Computer algorithms have been developed by making modifications in the existing algorithms for searching efficient designs with correlated observations for block designs (Satpati *et al.*, 2007), nested block designs (Satpati *et al.*, 2006) and CODs (Satpati *et al.*, 2012). Designs efficient for correlated observations are position dependent and lack within block randomization. Therefore, robustness aspects of designs that are efficient for zero correlation structure has been studied by performing randomization 5000 times, and for all these randomized layouts, the lower bounds to A- and D-efficiencies have been obtained for a given correlation coefficient and correlation structure. The range of correlation coefficient for different correlation structures when the loss in minimum efficiencies is less than 10% and 1% are obtained and the robust designs are catalogued.

Factorial experiments are widely used in agriculture and several other branches of science. Factorial experiments aim at exploring the effects of the individual factors and their inter-relationship as well. However, quite often in practice, the total number of treatment combinations is too large to allow the use of a complete factorial unless the number of factors and the levels of the factors are too small. Factorial experiment with large number of treatment combinations, apart from being expensive and impracticable in most situations, may not be at all necessary if the interest is in estimating only lower order effects under the assumption of absence of higher order effects. Thus, the experiment that includes only a fraction of the totality of all possible treatment combinations is called fractional factorial experiment. In any factorial experiment, the experimenter is interested in estimating main effects and lower order interactions under the assumption that higher order interactions are negligible. If two fractional factorial designs are of same resolution then the concept of minimum aberration based on wordlength pattern is used to select the best design out of the two designs with same resolution. Multi-level and mixed level minimum aberration fractional factorial plans have been constructed (Jambhulkar and Lal, 2013).

The experimental units in a design of a factorial experiment may exhibit a trend over space or time. Such situations may occur in agricultural experiments when there is a slope in the field and there is sequential application of the treatments to the same experimental unit. This may also happen when the land is irrigated, the nutrients supplied by the fertilizers may not be equally distributed and trend in experimental units may be due to slope. When such land is irrigated, the nutrients supplied by the fertilizers may not be equally distributed and a slope may cause a trend in experimental units. In such type of experiments, the treatments are to be allocated to experimental units in some order to eliminate the effect of such trend. The resulting designs are called as trend-free designs. The interest of the experimenter is to obtain design in which contrasts of main effects and lower order treatment combinations are estimated free from linear trend when experimental material is influenced by trend effect. Sarkar *et al.* (2009) developed an algorithm for construction of trend-free designs for multifactor experimental settings. The algorithms developed are helpful in generating the complete and confounded factorial experiments that are linear trend-free for main effects. Algorithms have been developed to obtain fractional factorial designs that are linear trend-free for main effects and to identify two factor interactions that are linear trend free/ nearly linear trend-free using the criterion of complement foldover.

In designed experiment it is not always true that the error of the generated data follows the normal distribution which is one of the basic assumptions of analysis of variance. 2^k factorial experiment has been considered and the error in the model is assumed to be distributed as generalized logistic distribution (non-normal distribution). Methods have been developed using the modified maximum likelihood estimates in which the maximum likelihood equations are linearized by using the Taylor's expansion and estimates of the parameters are obtained. These estimates are efficient under non-normal error distribution and asymptotic to maximum likelihood estimates (Yadav, 2014). Cook-statistic is developed for detecting outliers in factorial experiments. For developing this statistic, mean-shift model has been considered, *i.e.* mean of each of outlying observation is shifted from the mean of other observations. A general expression of cook-statistic for detecting any t observations has been obtained. The distribution of Cook statistic has also been obtained. A statistic known as outlier sum of squares or Q-statistic has also been developed for the same purpose. These developed statistics were applied to real experimental data. For applying data in Cook-statistic both single and paired observations

have been considered and outlying observation(s) is detected. Analysis has been done on the original data. Then analysis was carried out by replacing the outliers by their missing value estimates. The significant differences in the analysis were noted (Roy, 2014).

Sometimes the experiments have to be conducted to study the effect of two or more factors simultaneously and the heterogeneity in the experimental material is in two directions then the appropriate design is factorial row-column design. In these designs, some of the treatment effects are confounded in rows and some treatment effects are confounded in columns. For the estimation of the effects that are confounded in a single design, it requires more number of row-column designs. Designs for symmetric factorial experiments for nested row-column setting and for asymmetric factorial experiments under row-column settings have been constructed.

In factorial experiments, if the number of factors is large the number of treatment combinations becomes very large. While experimenting with such large number of factors many difficulties are encountered. In such studies complete factorial is not necessary as the interest is generally not in higher order interactions. In such experiments, the experimenter's endeavour is to minimize the number of runs to identify the active factors for efficient utilization of resources and minimization of cost and time. A supersaturated design (SSD) is a fraction of a factorial replication in which the number of factors is greater than the number of experimental runs. These designs are very cost-effective and as such are useful when experimentation is expensive and the number of factors is large. An exchange algorithm has been developed to construct two-level SSDs. Further, an algorithm for the construction of multi-level SSDs has been developed using saturated orthogonal arrays (Kohli, 2007). A new computer aided random generation of multi-level as well as mixed-level SSDs has also been developed. A catalogue of designs generated from these algorithms has been prepared (Gurung, 2009).

Response surface methodology explores the relationship between response variable and several explanatory variables with the motive to obtain an optimal response using a set of designed points. Further in many experimental situations, particularly in field experiments, the observations are correlated through some systematic pattern of environmental variations. Some methods of constructing designs for fitting first order and second order response models with correlated errors (equi-correlation and autocorrelation structure) have been obtained. Also the performance of some existing second order response surface designs viz. central composite and Box-Behnken that are efficient for fitting response surfaces under uncorrelated situation, have been studied under correlated error structure.

In response surface methodology, it is generally assumed that the observations are independent and there is no effect of neighbouring units. But in field experiments, the neighbour effects from the treatments applied to adjacent neighbouring units may arise. Jaggi *et al.* (2010) performed the response surface analysis from a design with factorial treatment structure in which the experimental units exhibit the overlap effects from the neighbouring units. The analysis has been illustrated by fitting first order and second order response surfaces incorporating neighbour effects from adjacent units. The results show that if the neighbour effects are present and are included in the response model, there is a substantial reduction in residual sum of squares and the response is predicted more precisely. Besides, the parameters of the model are estimated with high precision. The procedure for fitting a first order and a second order response surface model incorporating equal neighbour effects from immediate left and right neighbouring units has been derived (Sarika *et al.*, 2009, 2013). Conditions have been obtained for the estimation of the coefficients of the model. First and second order response surface model have also been studied under the assumption of differential neighbour effects from both left and right units. Varghese *et al.* (2016) obtained rotatable response surface designs in the presence of differential neighbour effects from adjoining experimental units. Blocking aspect in response surface designs in the presence of neighbour effects has been studied and the conditions for orthogonal estimation of parameters of first order model have been derived (Varghese and Jaggi, 2011). Some aspects of designs for fitting response surface models with quantitative-cum-qualitative factors have also been studied under neighbour effects/ correlated error structure. Computer aided generation of linear trend-free response surface designs was developed by Sarkar and Lal (2007).

A mixture experiment is an experiment in which the response is assumed to depend on the relative proportions of the ingredients present in the mixture and not on total amount of the mixture. The component proportion in a mixture experiment cannot vary independently since they are constrained to a fixed quantity (say unity). Alam *et al.* (2014) have given construction and analysis of multifactor mixture experiments. Sometimes the response in mixture experiments depends not only on the proportion of mixture components present in the mixture but also on the process conditions. In mixture experiment process variables do not form any portion of the mixture but whose levels when changed could affect the blending properties of the ingredients. When the mixture experiments are conducted with process variables then these experiments are called mixture experiments with process variables. A method of construction of efficient mixture experiments with process variable in minimum number of runs by using the projection matrix has been developed (Pradhan *et al.*, 2016). Also, a methodology has been developed for obtaining the optimum combination of ingredients in mixture experiments with process variables (Pradhan *et al.*, 2012).

Balanced sampling plans excluding adjacent units {BSA (*m*) plans} are useful for sampling from populations in which the nearer units provide similar observations due to natural ordering of the units in time or space. For BSA (*m*) plans, first order inclusion probabilities of all units are same and second order inclusion probabilities for pairs of adjacent units are zero and constant for other pairs of units. Polygonal designs are an important series of incomplete block designs useful for obtaining sampling plans where the contiguous units are similar. Considering the blocks of polygonal designs as samples and the treatments as units, a BSA (*m*) plan can be obtained by assigning equal probability of selection to the blocks. A computer algorithm has been developed to obtain polygonal designs. It is important to note that all polygonal designs are BSA (*m*) plans but vice versa may not be true. BSA (*m*) plans may be obtained by assigning unequal probabilities of selections to the samples in the support. Hence, the computer algorithm developed for generation of polygonal designs has been modified to obtain BSA (*m*) plans directly comparatively in a short time (Mandal *et al.*, 2008a). Mandal *et al.* (2008b) developed IPPS plans excluding adjacent (IPPSEA plans) units for sampling from populations where there is natural ordering of the units along with variability in sizes of the units. IPPSEA plans have been obtained by making use of binary, proper and unequireplicated block designs and linear programming approach. Mandal *et al.* (2009) developed a series of balanced sampling plans. Gopinath (2015) developed two dimensional sampling plan excluding adjacent units. Kumar (2016) obtained polygonal designs for sampling from naturally ordered populations.

The computer algorithm based on linear integer programming approach available for generating efficient binary incomplete block designs has been modified for obtaining polygonal designs for both the circular and linear ordered structure of population units (Mandal, *et al.*, 2011). Two dimensional balanced sampling plans excluding adjacent units (2-BSA(*m*), where *m* denotes the distance between units) have been developed for populations with circular ordering of units Algorithm based on linear programming approach have been developed for construction of 2-BSA(*m*) plans under both adjacency schemes for *m* dH2. Mandal *et al.* (2008c) also studied nested stratified sampling.

Bioassays or biological assays are used in many fields of sciences, especially in life sciences. In many practical situations for conducting experiments on biological assays, the interest of the experimenter lies in comparing several test preparations with a single standard preparation. The main purpose of a bioassay is the estimation of the relative potency of test preparation with respect to standard preparation. Therefore, only some specific contrasts are required to be estimated in bioassays. It is desired that when a block design is used for the assay, these contrasts of interest are estimated with high efficiency. A general method of construction of A-optimal binary block designs for symmetric parallel line assays has been obtained. The proposed method has been extended for multiple parallel line assays as well. Another method of construction of A-optimal binary block designs for asymmetrical parallel line assays has also been proposed. Some methods of construction of block designs for multiple parallel line assays based on BIB design have also been considered (Shekhar and Bhar, 2013; Shekhar *et al.*, 2014). Designs obtained through these methods are highly efficient in the sense that all

the contrasts of interest can be estimated with high efficiency. For the case of multiple slope ratio assays, a general method of construction of efficient block designs has been proposed.

Microarray experiments are conducted to study the expression levels of thousands of genes simultaneously. In these experiments, the treatments are different types of tissues, drug treatments or time points of a biological process, which may be unstructured or have a factorial structure. For single factor microarray experiments, a general method of construction of efficient row-column designs for any number of treatments with two rows has been developed (Sarkar *et al.*, 2010). For multi-factor microarray experiments, based on baseline parameterization a procedure of obtaining efficient block designs for 3-factor mixed level factorial microarray experiments has been given (Sarkar *et al.*, 2007).

In microarray experiments, the four experimental factors are array (*A*), dye (*D*), variety (*V*) and gene (*G*). These four experimental factors give rise to 2^4-1= 15 possible experimental effects excluding the general mean. Out of these 15 possible experimental effects, seven effects viz. array (*A*), dye (*D*), variety (*V*), gene (*G*), array-gene interaction (*AG*), dye-gene interaction (*DG*), variety-gene interaction (*VG*) effects are of main interest to the experimenter. Optimality aspects of designs for microarray experiments, have been studied leaving gene specific effects from the model, *i.e.*, by taking only array, dye and variety effects in the model. Designs that are good under the model containing global effects are also good under the model containing both global and gene specific effects. In 2-colour microarray experiments, only two varieties labelled with two different dyes can be accommodated on one array; therefore, arrays may be considered as blocks of size 2. Further, array effects may be taken as random. To deal with the problem of obtaining efficient designs when array effects are random, the lower bounds to the A- and D-efficiencies of the designs in a given class of designs have been obtained for block designs under mixed effects model. Efficient block designs for 2-colour microarray experiments have been obtained under a restricted model involving array and variety effects. The dye effects have been ignored from the model, since in microarray experiments, the two varieties appearing on the array are to be labeled with two different dyes. The catalogues of all efficient block designs and row-column designs obtained and the best available designs have been prepared along with their lower bounds to A-and D-efficiencies under fixed/ mixed effects models and their robustness status.

Sample Surveys and Spatial Statistics

The population of the country is growing at an enormous speed and therefore, it is of paramount importance to achieve an enhanced rate of production and productivity of crops in order to keep pace with the burgeoning population. This can be achieved through better planning and optimum utilization of the scarce resources. Any planning to be effective should be based on sound facts and credible data collected through scientific means and methods leading to the generation of valid statistics. India has a well-established system for collection of agricultural statistics with scientific methods for collecting, organizing and analyzing agricultural data. This helps in summarizing and presenting the results, as well as drawing valid conclusions for the development of the agricultural sector. This includes quantitative as well as qualitative information on the production, processing, distribution and utilization of agricultural commodities including livestock and fisheries. Survey sampling is one of the most common techniques for collection of reliable information in limited resources i.e. time, human and cost. Institute has one of the main responsibilities for development of survey methodology for collection of agricultural statistics as per the requirement of the country. In this regard, students of PG School, Indian Agricultural Research Institute (IARI), New Delhi have made a significant contribution on emerging issues in the field of survey sampling. Some of important achievements in this regard are as follows:

An alternative sampling methodology has been developed for estimation of average yield of cotton using limited number of pickings (Moury, 2015). The component sampling approach under uni-stage sampling design framework for estimating average yield of cotton at district level has been proposed. The proposed uni-stage component sampling approach has been extended to stratified three stage sampling. Under component sampling

approach, it is proposed to consider the yield of different pickings as the components of total yield. Estimates of average yield of cotton along with percentage standard error have been obtained for two districts namely, Amravati and Aurangabad of Maharashtra State and two districts namely, Warangal and Guntur of Andhra Pradesh State for the year 2012-2013. The estimate obtained using the proposed methodology is reliable and almost at par with the estimates obtained using General Crop Estimation Survey (GCES) methodology and an alternative methodology using double sampling regression procedure under stratified two stage sampling design framework.

The approach of estimation of parameter of interest in the traditional survey sampling theory is popularly called as "design based". Within the design based approach frame work, many different classes of estimators exist and it may not be possible to find an estimator which is best in all the classes of estimators i.e. the non-existence of Uniformly Minimum Variance Unbiased Estimator (UMVUE). An attempt has been made to develop predictor for the finite population mean square under linear model set up when the auxiliary information is assumed unknown for all the units of the population. Accordingly a double sampling based predictor of finite population mean square has been developed. It is shown to be design model unbiased and an expression for the variance has been developed. The efficiency comparison of the proposed predictor has been made vis-à-vis a predictor which utilizes auxiliary information for all the units of the population assuming that cost is incurred to collect information on the auxiliary character under a suitable cost function (Sud *et al.*, 2007).

Development is a multi-dimensional process and its impact cannot be captured fully by any single indicator. An indicator is a quantitative or a qualitative measure derived from a series of observed facts that reveal relative positions in a given area. A composite index is useful in identifying trends and drawing attention to particular issues in the context of policy analysis. An attempt was made to review the methodological issues for the construction of composite index. Agricultural Development Index (ADI) of Bihar State has been constructed using Principal Component Analysis (PCA). The performance of the districts has been compared based on these developed indices. An attempt has also been made to generate thematic maps of Bihar based on the Agricultural Development Index and its sub indices like Input, Output and Infrastructure index using Geographical Information System (GIS). Sensitivity analysis of the developed indices has been carried out using empirical method of variance-based technique (Kumar *et al.*, 2012).

In many surveys, the same population is sampled repeatedly and the same study variable is measured at each occasion so that development over time can be followed. For example, labour force surveys are conducted monthly to estimate the number of employed. Similarly, surveys are conducted for estimation of major livestock products wherein data are collected over seasons. It is a common experience, in sample surveys, to come across units in the sample which do not yield the required information at the first attempt. In such situation repeated visits to the non-respondents is necessary. The situation where some sample units do not provide the necessary information in the context of surveys on two occasions under a two-stage sampling design has been studied (Sud *et al.*, 2012, Aditya *et al.*, 2014). Three different non-response cases have been considered under two different sampling schemes. Different unbiased estimators of population mean along with their variance expressions has been developed to take care of non-response. It has been shown theoretically, that the proposed estimators are more efficient than the estimators obtained from single occasion sampling strategy. The estimation of domain total for unknown domain size in the presence of nonresponse has been studied by Aditya *et al.* (2012).

In many surveys (for example, agriculture, business enterprises, income and expenditure surveys), data are typically outlier contaminated that contain few extreme values and linear model assumptions are questionable. Influential observations may appear in the sample due to imperfections of the survey design that cannot fully account for the dynamic and heterogeneous nature of the population of businesses. An observation may become influential due to a relatively large survey weight, extreme value, or combination of the weight and value. A winsorized approach based estimator of population total with a choice of cutoff points has been proposed that guarantees that the resulting mean squared error is lower than the variance of the original survey weighted estimator (Guha, 2015). The expression for its mean squared error estimator is also obtained. The improved

performance of the proposed estimator is compared to existing estimators using a Monte Carlo simulation study.

Sample surveys are usually planned to produce reliable direct estimates for larger domains or areas and are not appropriate to produce small area statistics due to small sample sizes. Small area estimation (SAE) techniques are used to produce reliable estimates for such areas or domains. Binary data are often of interest in surveys. When small area estimates are required for such binary data, use of standard estimation methods based on linear mixed models becomes problematic. In this case, generalized linear mixed model with logit link function is often used in SAE, and the empirical best predictor (EBP) approach is widely used for the estimation of small area proportions under this model and proven to be efficient as well (Kumar, 2016). Three small area predictors for population proportion have been proposed under generalized linear mixed model with logit link function based on availability of level of auxiliary information. The performance of proposed method of small area estimation was evaluated through simulation studies. The results clearly indicated that the developed method of small area estimation works well for generating the estimates of small area proportions and represents a practical alternative to the EBP when population level auxiliary data in known.

The calibration approach is frequently used to develop precise estimators of important population parameters. A calibration estimator uses calibrated weights, which are as close as possible, according to a given distance measure, to the original sampling design weights while also respecting a set of constraints, the calibration equations. The calibration approach is frequently used for estimation of linear parameters like population total or mean. The calibration approach is used to estimate the parameter of finite population ratio. Different calibrated weights are obtained for different situations of available auxiliary information as well as for different system of weights. The expression for variance and estimated variance of the estimators are developed. The simulation study revealed that the calibrated estimators perform better than an estimator of population ratio which does not make use of auxiliary information. Research has been taken with the specific objectives on non-negative calibrated weight using quadratic programming technique for variance estimation.

Generally, a linear relationship exists between study and auxiliary variable. But in many surveys, the variables are not linearly related; some kind of nonlinear relationship exists between the study and auxiliary variable. The calibration approach is used to develop an estimator to take care of the situation. The variance of the estimator and estimator of variance are also developed. A simulation study has been carried out to demonstrate the improved performance of the proposed calibration approach based estimator over the ratio, regression and Horvitz-Thompson estimator in presence of quadratic relationship between study and auxiliary variable and it is found that the proposed calibration estimator is more precise (Yadav, 2014).

The mail questionnaire method is commonly used to collect data in surveys as the data collection costs following this method are considerably reduced. However, non-response can be serious problem in this method of data collection. The presence of non-response may result in biased estimates, particularly, when the respondents differ from the non-respondents. The calibration technique is widely used to obtain precise estimators of population parameters. The estimators for population total have been proposed through the calibration approach (Raman *et al.*, 2013). Expressions for variance and estimator of variance, to the first order of approximation, of proposed calibrated estimators in presence of non-response have also been developed. The performance of developed calibrated estimators is evaluated through a simulation study where the study population is generated through an assumed model and also by making use of real data. Further, calibration approach based estimators using multiple sets of weights in the context of non response are considered.

The calibration approach is used to estimate finite population ratio under two phase sampling design (Islam, 2015). Different types of calibrated weights are obtained for different situations of available auxiliary information as well as for single and double system of weights. The expression for variance and estimate of variance of the estimators are developed for general sampling design as well as for SRSWOR as a special case. The performance of the different calibrated estimators is studied through a simulation study. The simulation

study revealed that simple ratio estimator which does not make use of auxiliary information perform equally to calibrated estimators when the study variables are highly correlated but the correlation between study and auxiliary variable is very low. But for fixed correlation value between study variable, increase in correlation between study and auxiliary variable efficiency of calibrated estimator gradually increases. Further, the calibrated estimator based on two systems of weights is found to perform better than the other calibrated estimators based on single weight.

In many surveys (for example, agriculture, business enterprises, income and expenditure surveys), data are typically skewed which contain few extreme values and linear model assumptions are questionable. For such data, the relationship between variable of interest and auxiliary variable may not be linear in the original (raw) scale, but can be linear in a transformed scale, e.g. the logarithmic (log) scale. In such cases, it is expected that survey estimation based on a linear model is inefficient and an appropriate technique for estimation of finite population should then be based on a linear model for a transformed version of that variable (Basak *et al.*, 2014). Basak *et al.* (2015) made a prediction of population total for skewed variable under a log transform model. Model calibration approach is a general approach of calibration which is appropriate for estimation of survey data in case of non-linear relationship between target variable and auxiliary variable. A back transformation bias correction in the fitted values, using a second order Taylor series approximation, is used and developed the model-based model calibration estimator of population total for skewed data. The expression for its mean square error estimator is also obtained.

If parameter of interest is geographical in nature, it is always desirable to incorporate the spatial structure into the model for estimation of parameter of interest. Therefore, by incorporating the spatial effects in the model through both simultaneously autoregressive error process and geographically weighted regression approach, it is possible to improve the precision of small area estimates. Sud *et al.* (2015) developed district level crop yield estimation under spatial small area model. Further, it is expected to have more efficient small area estimates using prior knowledge about the parameter of interest i.e. using Bayesian approach in spatial small area models (Gharde *et al.*, 2013). An approach has been developed to incorporate the spatial information in the random area effect present in the unit level small area model (Gharde *et al.*, 2012). This approach is based on geographically weighted regression technique which incorporates the spatial information in the model. Small area estimators were obtained along with MSE using this model. The comparison of proposed estimators with non-spatial estimators have been made using simulated population for 15 different small areas. Rai, *et al.* (2007) studied small area estimation of crop production using spatial models.

In experimental settings where measuring an observation is expensive, but ranking a small subset of observations is relatively easy, Ranked Set Sampling (RSS) can be used to increase the precision of the estimators An attempt was made to develop variance estimation procedures using Jackknife method in RSS under finite population framework (Biswas *et al.*, 2013). Under this study, three different variance estimation procedures have been developed. The efficiency of these developed variance estimation procedures has been compared among themselves through a simulation study. The variance estimation procedures following cycle based approach and strata based approach have performed better than the variance estimation procedure following unit based approach for varying number of cycles as well as for varying ranks.

RSS when applied to spatially-correlated areal population fails to take into account the spatial correlation. Dependent Unit Sequential Technique (DUST) is a sample selection procedure for selection of areal units from spatially correlated population in which spatial correlation among the population units has been incorporated into sample selection procedure. An attempt has been made to propose a sample selection technique named as Spatial Ranked Set Sampling (SRSS) in which desirable features of both RSS and DUST have been incorporated (Kankure and Rai, 2007). SRSS has characteristics of RSS such as randomization technique for better representation of population and additional information about ranking of units within a set in the sample selection process. Also, the proposed SRSS incorporated spatial correlation as in case of DUST in the sample selection process. A spatial simulation study was carried out to empirically test the performance of SRSS with

respect to the traditional sampling techniques. It has been found that SRSS always performs better in terms of efficiency with respect to SRS and there is sufficient gain in efficiency with respect to RSS in case of smaller set size which is generally recommended to avoid ranking errors.

In agricultural surveys, the important parameters of crop production are often spatial in nature, in which observations from neighbouring units tend to share similar statistical properties. In literature, spatial sampling designs were suggested to provide reliable spatial statistics using the spatial dependency among the sampling units. A new efficient approach, viz. Spatial Estimation procedure, for estimation of the mean of spatially correlated finite population units has been developed (Biswas *et al.*, 2015) by incorporating the spatial dependency at the estimation stages of traditional without replacement sampling designs like Simple Random Sampling (SRS) and RSS. In this approach, Spatial Estimators (SE) of population mean were developed following prediction approach in which unobserved population units were predicted based on their distances with observed sampling units following Inverse Distance Weighting (IDW) method. Also, a spatial simulation study was carried out in order to study the performance of proposed SE with respect to the corresponding classical estimators of population mean under both the designs along with its variance estimation following proposed methods. Biswas *et al.* (2015) studied spatial estimation and rescaled spatial bootstrap approach for finite population.

In regression models, when parameter of interest is geographical in nature the regression coefficients do not remain fixed over space. When the regression model is applied to geographical data where each sample observations correspond to a geographical location, space plays no role in the modeling process. To overcome this problem geographically weighted regression was used and the regression model centered at each point p_i could be thought of as a weighted ordinary least square regression. If the concept of spatial dependence was introduced into the weight functions using the spatial variogram models approach then the technique obtained was considered as improved technique over the weights obtained by distance of neighbourhoods. The yield data obtained from crop cutting experiments and the corresponding locations of yield data in terms of latitude and longitude were identified. The spatial statistics was employed to the data of yields and locations to determine the spatial model of best fit along with its parameters. With the help of best fitted spatial model ordinary kriging method was used for spatial prediction at unsampled locations. The average yield and its standard deviation can be obtained for each village.

Measurement errors in explanatory variables of classical regression model makes the estimators of regression coefficients biased and inconsistent. In case, variable of interest is geographical in nature, regression coefficients do not remain fixed over space and usual regression analysis takes no account of spatial location in its analysis. Geographically weighted regression is used in which estimates of regression coefficients are based on local relation instead of global relations among spatial variables of interest. An attempt has been made to propose a Functional Spatial Regression (FSR) model under measurement errors and a Structural Spatial Regression (SSR) model under measurement errors for estimation of regression coefficients in case of spatial variables. Explanatory variables under FSR model are assumed to be fixed while it is random in case of SSR model. Modified estimates of regression coefficients were proposed following Ordinary Least Squares, Generalized Least Squares, Maximum Likelihood Estimation and Method of Moment Estimation approaches in both model structures. It has been shown through spatial simulation that proposed estimators are unbiased, consistent and comparatively more efficient than corresponding usual estimators. Four different approaches were followed to incorporate spatial effects in the proposed models.

Statistical Modeling and Forecasting

Statistical modelling plays a very important role in comprehending underlying relationships among crucial variables in an agricultural system. In any agricultural subsystem, the underlying relationships among variables are generally complex, thereby leading to a nonlinear model. Some of important achievements in statistical modelling and forecasting are as follows:

Modeling growth of an animal is complex in nature. For growth modeling a description of longitudinal measurements about few parameters and the biological interpretations are required. In case of longitudinal growth data, there are within and between individuals variation. The variability between individuals are not included in fixed effect model and the problem can be solved by using non-linear mixed effect models (NLMM). NLMM have flexible covariance structure to handle unbalanced data. Attempt has been made to develop the Von-Bertalanffy mixed model. Logistic, Gompertz and Von-Bertalanffy fixed and mixed models have been explored for growth data of Goat and Pig (Das *et al.*, 2016). Comparison of the models i.e. between fixed and mixed type of the same model and among different fixed and mixed models has been attempted. It has been found that Logistic mixed effect model performed best for the Goat and Pig data. The prediction of this model is also better.

Use of ordinal logistic model has been explored for forecasting wheat yield in Kanpur district of Uttar Pradesh. Weekly weather data (1971-72 to 2009-10) on five weather variables namely maximum temperature, minimum temperature, morning relative humidity, evening relative humidity and Rainfall for 16 weeks of the crop cultivation which include 40th standard meteorological week (SMW) to 52nd SMW of a year and 1st SMW to 3rd SMW of next year have been used in the study. Crop years were divided into two or three groups based on the detrended yield. Crop yield forecast models have been developed using probabilities obtained through ordinal logistic regression along with year as regressors for different weeks starting from 52nd SMW. This approach has been compared with that of discriminant function analysis (Kumari and Kumar, 2014).

In agricultural research, data are usually collected over time called time-series data and one characteristic of such data is that the adjacent observations are dependent. Linear time-series model, like autoregressive integrated moving average (ARIMA), and nonlinear time-series models, like Exponential autoregressive (EXPAR) and Self exciting threshold autoregressive (SETAR) are generally used for modelling and forecasting of cyclical time-series data. Methodologies for combining these by using the constant coefficient regression method as well as the Time-varying coefficient regression method through Kalman Filter (KF) technique has been investigated (Ghosh *et al.*, 2015). As an illustration, these are applied to describe annual Mackerel catch time-series data of Karnataka and it is shown that the latter method has performed better than the former one for the data under consideration. Ghosh *et al.* (2014) have studied KF based modeling and forecasting of stochastic volatility with threshold.

Agricultural price forecasting is one of the challenging areas of time series forecasting. An effort is made to compare the forecasting capabilities of well known Box-Jenkins or ARIMA methods with nonlinear time delay neural network (TDNN) models using data on monthly wholesale price of oilseed crops traded in different markets in India. TDNN models outperform the ARIMA models for six and twelve months ahead forecasting in terms of root mean square error. The sequential combination of ARIMA and TDNN models is adopted to harness the unique strength of individual models for improving the forecast accuracy. The results of the study show that combined models underperform compared to their components' performances which may be due to the failure of the basic assumption of additive relationship between linear and nonlinear components of this approach for the series considered in the experiments.

Several statistical models are developed using Nonparametric Nonlinear Support Vector Machines (SVM) methodology. SVM is a Generalized portrait algorithm proposed for solving problems in classification, function estimation and density estimation (Kumar Mohan, 2014). Basic concepts of linear and nonlinear SVM and their formulation for binary classification problems are discussed. Methodology for extending binary classification problem to multiclass classification problem is considered. Further, optimal hyper-parameters of this model are estimated using Particle Swarm Optimization (PSO) algorithm. As an illustration, this methodology is applied for classification of three varieties of banana based on their morphological characters. Result shows that the methodology has performed the best vis-à-vis other competing methodologies for the data under consideration. Extension of SVM methodology to regression problem called as Support Vector Regression (SVR) is thoroughly studied. PSO technique is employed to estimate its optimal hyper-parameters. Superiority of this methodology

is demonstrated over Artificial neural network and multiple linear regression for maize crop yield data. Various hybrid models are developed to tackle complex time-series data by combining Linear Seasonal Autoregressive Integrated Moving Average (SARIMA) and Nonlinear SVR models for time-series forecasting. Optimal hyper-parameters of these models are estimated using PSO technique. Subsequently, as an illustration, these models are applied to all-India monthly marine products exports time-series data. Gurung *et al.* (2014a) fitted smooth transition autoregressive nonlinear time-series model using PSO technique. Gurung *et al.* (2014b) also applied stochastic volatility model fitting using particle filter.

The ability to accurately forecast the price volatility of agricultural commodities is an important concern among both policy makers and farming community. An effort has been made to evaluate the forecasting performance of dynamic neural networks model with Generalized Autoregressive Conditional Heteroscedastic (GARCH) model for predicting the volatility using monthly price series of edible oils in domestic and international markets in terms of mean square error (MSE) and correct directional change. The neural network based models uniformly outperformed the GARCH model in predicting the direction of price volatility while performance based on MSE provided mixed results. Paul *et al.* (2009) performed GARCH nonlinear time series analysis for modeling and forecasting of India's volatile spices export data. Ghosh *et al.* (2010) and Lama *et al.* (2015) applied the GARCH and Exponential GARCH (EGARCH) nonlinear time-series models for volatile data.

Ghosh *et al.* (2010) studied wavelet frequency domain approach for statistical modeling of rainfall time-series data and Paul *et al.* (2011) used wavelet methodology for estimation of trend in Indian monsoon rainfall time-series data.

Mixture nonlinear time-series modelling have been studied. Three families, viz. Gaussian mixture transition distribution (GMTD), Mixture autoregressive (MAR) and MAR-Autoregressive conditional heteroscedastic (MAR-ARCH) models are studied by considering weekly wholesale onion price data during April, 1998 to March, 2002. Formulae for carrying out out-of-sample forecasting up to three-steps ahead have been obtained theoretically by recursive use of conditional expectation and conditional variance. In respect of out-of-sample data, results derived enable to compute best predictor, prediction error variance, and predictive density. It is concluded that a two-component MAR-ARCH provides best description of the data for modelling as well as forecasting purposes.

Threshold Autoregressive (TAR) and Two-regime Self-exciting TAR (SETAR) family of models are investigated and applied to country's lac export data obtained from annual reports of Shellac Export Promotion Council, Kolkata. Nonlinear Richards growth model has been studied and the methodology for estimation of parameters using Genetic Algorithm (GA) is developed (Iquebal *et al.*, 2009). As an illustration, the same is applied for modelling and forecasting of country's foodgrain production data. Ghosh *et al.* (2008) made a bootstrap study of variance estimation under heteroscedasticity using genetic algorithm. The Self-exciting threshold autoregressive moving average (SETARMA) two-regime model is thoroughly described and used for forecasting of annual mackerel catch time-series data of Karnataka. Iquebal, *et al.* (2010) applied genetic algorithm for fitting self-exciting threshold autoregressive nonlinear time series model. Samanta, *et al.* (2011) used SETARMA nonlinear time-series approach for modelling and forecasting cyclical fish landings. Ghosh *et al.* (2014) fitted SETARMA nonlinear time-series model through Genetic algorithm and developed out-of-sample forecasts.

In general, for analysis of time-series data, linear time-series models are used. But such models cannot capture the asymmetry (when average number of observation in the up cycle is different from that of down cycle) in the data. To deal with such behaviour linear autoregressive time-series model may be extended to nonlinear time-series taking the autoregressive coefficient as a time-varying coefficient i.e. Random Coefficient Autoregressive (RCAR) model and Fourier Autoregressive (F-AR) model. A RCAR model has been fitted by representing the model into state space form followed by estimation of parameters using KF (Ghosh *et al.*, 2010). Fitting of F-AR model is done with minimum number of Fourier coefficients. Finally the two models

are compared with fitted ARMA model. As an illustration purpose, quarterly oil sardine fish catch in Kerala for the period 1985-2008 is considered for building the model and 2009-2010 is used for validation. Ghosh *et al.* (2014) studied Fourier-autoregressive (F-AR) coefficient non-linear time-series model for forecasting asymmetric cyclical data.

The classical time-series theory assumes values of the response variable to be 'crisp' or 'precise', which is quite often violated in reality, and cannot handle 'fuzziness' in the underlying system, a problem which can be solved by fuzzy time-series analysis. A study of fuzzy time-series models in agriculture has been done by Chouwdhary (2013). Attempts have been made to develop a new methodology for fuzzy time-series modelling using non-convex membership functions and perform prediction of out-of-sample data. New methodology is useful in the sense that it removes the difficulty for out-of-sample forecast by difference operator based methodology by obtaining fuzzy forecast of future time point in intermediate step. Singh *et al.* (2008) developed a two-stage fuzzy least squares procedure for fitting linearized von Bertalanffy growth model. Singh *et al.* (2008) gave an application of possibility and necessity measures for fuzzy linear regression analysis.

Artificial neural network and fuzzy regression modelling provide attractive ways to capture nonlinearities present in a complex system. Neuro-fuzzy modeling, which is a judicious integration of merits of these two approaches, has been applied in real data set. A modified fuzzy least-squares approach for estimation of parameters is applied for Pearl Millet crop yield estimation at block level (Singh and Prajneshu, 2008). Multilayered feedforward artificial neural network (MLFANN) is illustrated by considering Maize crop yield data as response variable and Total human labour, Farm power, Fertilizer consumption, and Pesticide consumption as predictors. Adaptive Neuro-fuzzy inference system (ANFIS) model is implemented on Fuzzy Logic Toolbox of MATLAB using ANFIS. As an illustration, the methodology is applied for development of a forecasting model for secondary data of yield of 100 banana plants on the basis of data at six different stages of growth using several biometrical characters, like Plant height, plant girth, and leaf length as predictors (Ghosh *et al.*, 2008).

The performance of resampling based methods with conventional methods for estimation of nonlinear models has been done. For this, productivity of wheat and proportional area under HYVs of maize over time were studied by fitting non-linear growth models viz. Logistic and Gompertz at various locations by employing two estimation procedures; the first one being the conventional Marquardt's nonlinear estimation procedure and the other one is the resampling based Jackknife estimation procedure. Also a simulation study has been done to compute the variances of the parameter estimates. The fitted models have been utilized for forecasting the crop statistics considered for subsequent periods not included in model fitting (Kandala, 2008).

Discriminant function technique has been used for developing the forecast models for wheat crop yield for Kanpur district of Uttar Pradesh using weekly data on the weather variables such as maximum temperature, minimum temperature, rainfall and morning relative humidity. Variables (weather indices) used in the discriminant function analysis were derived through different procedures. In this approach two discriminant scores have been calculated by taking the data of the weather variables of the first week. Then taking these two scores and the weather data of the second week again two scores have been obtained. This procedure is repeated till the last week and finally two scores have been obtained. Then taking these two scores and trend variable as the regressors, forecast regression model has been fitted. This procedure gave minimum root mean square error among all procedures studied (Agrawal *et al.*, 2012).

Classification and prediction in agricultural systems are quite useful for planning purposes. Logistic regression modeling has been employed for classification purposes. The data pertain to the area of agricultural ergonomics with dependent variable as the presence or absence of discomfort for the farm labourers in operating farm machineries along with associated quantitative and qualitative regressors (Bhowmik *et al.*, 2011). Classification of the hold-out datasets revealed that results obtained using logistic regression models were better when compared to those obtained from discriminant function analysis method. The study revealed that logistic regression

modeling can be employed as a viable alternative for classification purposes in the field of agriculture. Sadhu *et al.* (2014) have used decision tree based models for classification in agricultural ergonomics.

Tree based modeling has been employed for classification purposes in the field of agriculture. As the tree based classification method works better for larger datasets, such a dataset was generated by simulation procedure from the available dataset. Classification and Regression Tree (CART) models have been fitted on the different possible predictor variable subsets of this larger dataset. The fitted models have been compared by cross-validation error measures of misclassification rates. All these models were employed for classification of observations not used in models fitting. In addition, an alternative tree-based modeling procedure for classification has been proposed within the CART framework to overcome some of its inherent drawbacks. Classifications using the fitted CART models were found to be better when compared to those obtained for corresponding logistic regression and discriminant function analysis methods.

Ray *et al.* (2014) investigated time series intervention modeling in the domain of agriculture. Such models are employed in situations where it may be known that certain exceptional external events called 'interventions' could affect the time series phenomenon under study. As a case study, Cotton yield of India at all-India level and for two major states viz., Gujarat and Maharashtra have been considered with the intervention being introduction of Bt Cotton variety in 2002. The performance of ARIMA intervention models has also been investigated on data sets simulated under different possible situations taking cotton yield data at all-India level as the baseline data set. When cotton yields were forecasted, the performance of ARIMA intervention models was found to be superior to the conventional ARIMA models.

The behaviour of monthly rainfall data was investigated. There are many exact zeroes in the rainfall recording of a month over a period of years. Probability distributions currently in use are not able to combine the discrete aspect of getting an exact zero reading of rainfall in a month and the aspect of amount of rainfall in the same month. Compound Poisson Tweedie distribution has been used to fit the monthly rainfall data by computing the estimates of its parameters. For the purpose of estimation of parameters of compound Poisson Tweedie distribution, a new algorithm has been developed. The Tweedie family of distributions are good for fitting rainfall months with few or many zero readings (Lall, 2014).

In view of increasing share of energy in the cost of cultivation as well as deregulation of prices of some petroleum products, agricultural commodity prices are vulnerable to the rise in energy prices, particularly of crude oil. An attempt has been made to examine the co-movement between energy and agricultural commodity prices with the help of Johansen co-integration technique using monthly wholesale price indices for the period April 1994 to March 2014. Since the process of deregulation started from April 2002 onward, the entire period was divided in two equal parts, so that before and after period analysis will provide a clear picture of a potential link between prices. The results revealed that energy and selected agricultural commodity prices are integrated in the long-run since 2004 while fruits prices were integrated even before deregulation of petroleum price (Kumar, 2015).

Detrended Yield approach has been used for developing the forecast models for the time series data on the yield of Wheat crop *(Triticum aestivum)* in five districts of Uttar Pradesh namely, Lucknow, Kanpur, Banda, Jhansi and Faizabad, which were subsequently treated as the dependent variable and weekly data on the weather variables (weather indices) were used as the regressors variables in the model. The yield has been detrended by obtaining the parameter estimates of the model and subsequently the detrended yield was used to forecast the yield of the crop using ARIMA model. The proposed approach is better and more suitable as compared to the traditional approach for forecasting the wheat yield (Banerjee, 2016).

Aphid population data on mungbean crop from five different locations of India, viz., Pantnagar, Behrampur, IARI, Mohanpur and Bharatpur farms along with the meteorological data on rainfall, maximum temperature, minimum temperature, maximum relative humidity, minimum relative humidity, sunshine hours, wind speeds etc. for December, January, February, March, and April from 2001 to 2004 have been used to fit various models

appropriate for modeling pest count data. It has been found that results obtained through negative binomial model are the best among other models considered (Ithuteng, 2016).

In statistics, usually data are formatted as single values. However, sometimes the data are represented by lists, intervals, histograms or even distributions. To deal with these kinds of data, the concept of symbolic data was introduced. Among symbolic data, interval-valued data are the most commonly seen in application. Ranganath *et al.* (2014) obtained descriptive statistics for symbolic interval-valued data.

Statistical Genetics and Genomics

The research carried out by students in the field of statistical genetics and genomics is on the diverge topics ranging from estimation of Genetics Parameters, Genotype x Environment Interaction and Crop Yield Stability, Statistical Genomics and Detection of QTLs.

Quantitative traits are the traits controlled by many genes and each of the genes has a small effect on the trait. The loci controlling quantitative traits are referred to as QTLs (Quantitative Trait Loci) and the procedure of finding and locating the QTLs are called QTL mapping. The data of maize crop in drought stress is considered for detecting loci influencing the performance of maize in drought stress Mapping population with 236 RILs (Recombinant Inbred Lines), using Ac7643S_5 (drought tolerant) and Ac7729 (drought susceptible) as male parent, at CIMMYT and evaluated in India under AMBIONET (Asian Maize Biotechnology Network), Maize Genetic Unit, IARI is considered. Phenotypic data on different plant characters have been utilized. Genotypic data on RILs obtained through 138 RFLP markers and a linkage map of a total length of 2250 cM with an average density of 17 cM are used for QTL detection. Genotypic data is obtained through molecular marker assay whereas phenotypic data is derived from evaluation of mapping population of the target traits of interest. It is found that marker MK28 has main-effect on four traits, marker MK28, MK101 and MK34 has main-effect on two traits. Interaction effect has also been found in different marker pairs. For the study the data available on two environments Hyderabad and Karimnagar have been considered, for examing QTL×environment interaction. The results are also obtained using Jackknife procedure and Bayesian methodology and from the results, it is seen that the results obtained by Jackknife and Bayesian methods are very much similar (Jambhulkar, 2007).

There are many characters of economic importance in animal and plant breeding which are polygenic in inheritance but their phenotypic expressions show discontinuities. The characters whose inheritance is multifactorial but exhibit all-or-none or one-or-the-other kind phenotypic expression are called threshold characters. Intraclass correlation coefficient provides a quantitative measure of similarity between individuals within groups. Mastitis or inflammation of the mammary gland, is one of the most common disease of dairy cattle throughout the world which causes heavy economic loss to the farmers. The heritability of mastitis disease in sahiwal breed of cows has been studied by estimating it through intraclass correlation based on different methods. Also heritability of mastitis disease is estimated following beta-binomial method. The estimates of heritability based on both showed similar performance. These heritability estimates have less standard error as compared to other estimates (Behera *et al.*, 2010).

Growth is an important phase in the life of animals which influences the different forms of production such as milk, meat etc. in the later ages. The relationship between body weight and age is important particularly in meat producing animals. Since a series of weight-age data points are analytically difficult to interpret, it is, therefore, desirable to study statistically the growth of animals. Under fitting of nonlinear growth model and choosing best model to describe growth pattern, many works has already been done. Statistical properties of goodness of fit criteria have been studied for selecting best model to describe the growth pattern by using bootstrap technique. Based on these statistical measures the best model is selected to describe growth pattern in given body weight data of Goat and Von bertalanffy model comes out to be the best model (Singh, 2010).

Classification is of broad interest in science because it leads to many scientific studies and also arises in the contexts of many applications. For example in agriculture, crop varieties are classified into different groups which are suitable for different agroclimatic zones of a region. Classification techniques are the multivariate techniques which are based on some assumptions. But in crop data situation, there may be violation of assumptions of the classification techniques. Hence, it was required to investigate how the classification techniques perform when certain assumptions about the data characteristics are violated. Further, occurrence of missing observations in multivariate crop data is a common feature in agricultural experiments due to the non-response of genotypes, disease and pest attack. The classification of genotypes in presence of missing values is a challenging task for breeders. The performance of different classification techniques *viz.* Oblique Axes Method (OAM), *k*-th nearest neighbor (KNN), Linear Discriminant Analysis (LDA) and Quadratic Discriminant Analysis (QDA) are compared based on Apparent classification Error Rate (APER). Further the result shows that, KNN followed by OAM and LDA perform better in skew-normal situations than normal condition and QDA performs better in normal condition. For maximum consistency and accuracy of classification of skew-normal data, KNN is best among the above four classification techniques irrespective to the different levels of missing observations (Das, 2012).

Artificial Neural Network (ANN) methodology has been explored for classification of crop genotypes and to compare it with classical clustering methods based on different distance measures. The secondary data of ten morphological characters consisting of 77 genotypes of maize crop collected from All India Coordinated Maize Improvement Project for the year 2005-06 has been used. The genotypes were clustered using different classical methods such as single linkage, average-between linkage, average-within linkage, Ward's method and non-hierarchical K-means method (Dash *et al.*, 2009, Dash *et al.*, 2012). The five homogeneous groups so obtained from consensus basis were tested for their significant difference using Hotelling's T^2 statistic. Since some of the grouped pairs were non-significant, the data were reclassified by considering four and three homogeneous groups. Taking the mean vector and dispersion matrix of three distinct homogeneous groups the multivariate normal samples of different sample sizes were obtained with prior knowledge of cluster identity. These samples were classified using different classical clustering methods and distance measures as well as ANN method. All the methods were compared by the probability misclassification. From the methods studied, ANN is the best method of classification as it gives minimum probability of misclassification irrespective of distance measures used in classical clustering methods and sample sizes.

Abiotic stress factors severely limit plant growth and development as well as crop yield. Various proteins of the plants are responsible for regulation of these abiotic stresses. A highly accurate prediction method capable of identifying protein function, based on physicochemical properties of protein has been described (Paul *et al.*, 2014). On the basis of 34 features extracted solely from the protein sequence, which are responsible for the regulation of different abiotic stresses *ie* heat, cold, drought and ABA, models were built to predict the functions of different proteins of Poaceae family. Models for classification of cereal proteins have been developed using SVM. Feature selection was performed by stepwise Logistic Regression taking presence of abiotic stress as the response variable. The accuracy of protein function prediction using SVM with different kernel functions ranged from 60% to 100% when classification was performed using physicochemical properties. Prediction accuracy of different models developed using structural composition with different kernel functions was also validated using bootstrap and 10 fold cross validation. Accuracy of prediction of these models ranges from 77% to 100% when classification was done based on structural composition. Bardhan *et al.* (2015) identified suitable clustering method and allocation strategy for core set development in salt stress tolerant rice (Oryzasativa) germplasm.

An attempt has been made to evaluate the principal component based fuzzy c-means algorithm in classifying 518 lentil genotypes based on their numeric agronomic and morphological traits (Mazumder, 2013). The principal component analysis was used for dimensionality reduction or feature extraction which also avoided the ill-effects of collinear variables. Results of the study revealed that genetic divergence is not highly related to geographical origins as exotic and indigenous lentil genotypes were distributed in all the four clusters. Further,

the set of descriptors for the germplasm accessions consists of both numerical and categorical descriptors. This poses a problem for standard principal component analysis which deals with only numeric variables. Hence, nonlinear principal component analysis was used to analyze the descriptors of lentil accessions which can handle mixture of measurement types. The accessions plot based on nonlinear component showed that most of the indigenous genotypes/land races overlapped indicating their narrow genetic base. Most of the outlying accessions belonged to exotic origin or breeding lines derived from crosses with exotic lines. An effort was also made to use first two nonlinear principal components as input to fuzzy c-means algorithm in classifying lentil germplasm collections. The study showed that principal component based fuzzy clustering has a promising potential in agriculture as a tool to evaluate, understand, predict and manage crop production.

Fisher's linear discriminant function (LDF) which is generally used in classification problems assumes multivariate normality and equality of dispersion matrices between different groups. The performance of Fisher linear discriminant function have been studied and compared under multivariate non-normal data with normal data using two sets of live data on three distinct populations of rice and maize consisting of nine and ten morphological characters respectively (Raman *et al.*, 2012; Raman *et al.*, 2015). The performance of LDF is compared on the basis of D^2, discriminating power and probability of misclassification under non-normal situations. It has been observed that D^2 values between groups are significant and probability of misclassification decreases with increase in sample sizes as well as increase in number of characters taken in D^2 statistics in both the rice and maize. It was concluded that the violation of multivariate normality assumption in LDF results in higher probability of misclassification as compared to normal.

Crop genotypes possess a great deal of similarity in respect of qualitative characters, quantitative characters or a mixture of both. Furthermore, quantitative characters' based distinction may not be very accurate because of the influence of environment on expression of these characters. Whereas qualitative characters like the molecular markers reveal this distinction among genotypes, which are not influenced by environments. Many a time, clustering techniques are applied for qualitative data and quantitative data separately in order to classify the genotypes into homogeneous groups. There is a problem when the degree of correspondence between the clusters groups formed based on qualitative traits and quantitative traits do not agree with each other. There was a need to study and identify suitable methods which can classify crop genotypes based on mixture of qualitative and quantitative data. Sarkar *et al.* (2011) made a comparative performance of clustering procedures for mixture of qualitative and quantitative data with an application to black gram. An approach to the development of a core set of germplasm using a mixture of qualitative and quantitative data has been studied by Sarkar *et al.* (2015). Occurrence of missing observations in multivariate genotype data is a common feature in breeding experiments due to non-germination, pest and disease attack *etc.* Thus classification of genotypes based on mixture data in presence of missing observations is of great interest to the breeders. Two methods, namely, INDOMIX and PCAMIX along with k-means clustering were identified as suitable procedure for classification of genotypes based on mixture data as they show lowest weighted average probability of misclassification. The results reveal that the methods are robust up to 10% imputed missing observations (Sarkar *et al.*, 2012).

Identification and proper treatment of multivariate outliers has become increasingly important in breeding data analysis. Four methods of multivariate outlier detection have been considered of which one is a popular and commonly used method and the other three are computer intensive methods (Meher *et al.*, 2010). These methods are compared for their performance based on probability of identified outliers from outlier distribution (correct outliers) as well as probability of outliers from "clean data" distributions (wrong outliers). Also the performance of all the methods is accessed by considering shift outliers alone, scale outliers alone and both shift and scale outliers in the samples generated through simulations. The probabilities are calculated over thousand simulations. The average probabilities of correct outliers and wrong outliers along with their standard errors are also obtained. The method which identifies high average probability of correct outliers and low average probability of wrong outliers has been judged as the best one. The identified best procedure has been applied on real data obtained from multi-location trial multivariate data on maize. Multivariate outliers are then treated

by deletion followed by multiple imputations. The treated data is further analyzed for identifying stable maize genotypes.

Knowledge of germplasm diversity among breeding material is an invaluable aid in crop improvement programs. Genetic diversity refers to variation within the individual gene locus/ among alleles of a gene, or gene combination, between individual plants or between plant populations. With the availability of population of rice germplasm, it will be interesting to identify a core set of germplasm accessions representing the maximum diversity present in the population. These rice germplasm exhibits tolerance to different degree of abiotic stresses like heat, cold, moisture, salinity and submergence. With the advent of Single Nucleotide Polymorphism (SNP) genotyping technologies and availability of phenotypic traits performance of germplasm, the amount of information available on quantitative and qualitative traits is enormous and their number is much larger than the number of germplasm accessions or observations. The application of Random Forest (RF) and Least Absolute Shrinkage and Selection Operator (LASSO) are useful to identify effective SNPs associated with the phenotypic trait performance (Sarkar *et al.*, 2015). Further, a combination of Ward's clustering method with Gower's distance and NY allocation method with at least 25% of sampling intensity is found suitable criterion to develop a maximal diversified core set of rice germplasm for salinity stress tolerance.

Several functional elements like Transcription Factor Binding Sites (TFBS), splice sites, SNPs are present on genome and identification of these elements in wet-lab experiments is quite expensive. Hence, development of computational methods for the prediction of functional elements on genome is essential to understand the underlying complex biological phenomena. Meher *et al.* (2014a) gave a statistical approach for 5' splice site prediction using short sequence motifs and without encoding sequence data. Meher *et al.* (2014b) determined window size and identified suitable method for prediction of donor splice sites in rice (Oryza sativa) genome. One such phenomenon involves the prediction of disease risk of an individual using whole genome level information on SNPs. However, disease risk prediction studies involve imbalanced case-control data, where controls are higher than the cases. Moreover, the number of SNPs remains larger than sample size. For the classification of imbalanced case-control data, a variant of Random Forest - Combined Random Forest (CRF) was proposed and its performance was compared with the existing variants: Balanced RF and Weighted RF. The proposed CRF was found to perform better over existing methodologies in terms of prediction accuracy. For the prediction of splice sites in rice, an encoding approach with SVM, ANN and RF were proposed. A comparison among them reveals that the encoding approach with RF predicts the splice sites with higher accuracy (Meher *et al.*, 2016).

Multi-Location Trials (MLTs) are being conducted for performance testing of the genotypes over environments. However, the perennial crops which exhibit biennial rhythm in fruiting, like mango, may mislead the selection of genotypes unless the bienniality is removed. Also, some of the observations of MLTs data may be missing due to disease attack, adding/dropping of genotypes from year to year, bad weather etc. which leads to incomplete genotype ´ environment data matrix. A moving average approach has been adopted to eliminate bienniality factor in the Additive Main Effects and Multiplicative Interaction (AMMI) model. The correlations between ranks of genotypes assigned based on the performance of yield, stability variance and simultaneous selection indices before and after elimination of bienniality were estimated and found to be deviated from unity due to the effect of bienniality. The results have been verified through simulation and assessed the changes in genotype ranking as well as in the proportion of genotypes selected in the top 50% of the total genotypes under real data situation. Thus, selecting genotypes in MLTs in the presence of bienniality is recommended after eliminating the bienniality from the data. The application of EM-AMMI procedure in the presence of bienniality was found to be more robust than FITCON against increasing rate of missing observations. The results reveal that EM-AMMI can be safely used up to 10 per cent missing observations. Twenty nine *putative* genes responsible for mango bienniality have been identified by *in silico* methods and a database consisting of genic / genomic information has been developed (Choudhary *et al.*, 2016).

Discrimination of coding and non-coding regions of cattle genome has been done based on epigenetic mechanism (Varshney, 2016). The cattle genome is collected from the UCSC Browser. Five different indices namely Deviation of Nucleotide, Deviation of Dinucleotide, Intensity of methylation effect, Triplet avoidance index and tendency of Polypurine and Polypyrimidine that could encode exon and intron sequences have been used. Dispersion matrix was estimated based on the performance of the five indices in the training set. The relative abundance of the five indices has also been worked out in the three categories. The area under the ROC and the PR curve along with their standard error were also computed. The proposed method has been compared with the methods based on LDA and Quadratic Discriminant Analysis (QDA) based on the area under the ROC and the PR curves, whereby the proposed method proved to be better.

Future Perspectives

The future perspectives of research in Agricultural Statistics include robust, optimal and cost effective designs for crop, fisheries and animal experimentation, factorial experiments and response surface designs in minimum number of runs, algorithmic approaches for resource optimization in experimentation, designs for computer experiments, Resampling based approaches for parameters estimation, Small area estimation, Data mining in spatio-temporal domain, Statistical techniques for survival analysis, Technology forecasting, Soft computing techniques for forecasting, Functional data analysis techniques, advance multivariate statistical techniques in high dimensional plant and animal germplasm data with reference to comparative genomics, phenomics and microarray experiments, Computational models for high dimensional data in biological systems.

Acknowledgement

We wish to thank all the Faculty Members of the Division of Agricultural Statistics for extending support and cooperation in preparing the research contributions of M.Sc. and Ph.D. students made during the period of 2007-16.

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Bioinformatics

A.R. Rao and Anil Rai

Introduction

Driven by the recent developments in plant, animal and fish genome projects, bioinformatics is taking an ever increasing role in the agriculture and allied sciences. In a similar way, advances in statistics, information technology and computational methods are driving the mathematical sciences a way forward through its extensive application in bioinformatics. Bioinformatics aims to bring the biologists, statisticians and computer scientists together from the point of view of system biology approach to understand the biological phenomenon. Various factors like, quantitative nature of agricultural traits and exponential growth of crop/animal genetics and genomic data, have resulted in a rapidly increasing demand for individuals and students trained in bioinformatics. Keeping this in view, ICAR- Indian Agricultural Statistics Research Institute (IASRI) has taken an initiative way-back in 2007 to establish Agricultural Bioinformatics Lab (ABL). Later on, Indian Council of Agricultural Research (ICAR) New Delhi has established Centre for Agricultural Bioinformatics (CABin) in the year 2010 with a team of multi-disciplinary research professionals as a division at ICAR-IASRI, New Delhi. The mission of this Centre is to provide computational support to biotechnological research in agriculture with a mandate to undertake research, teaching and training in the field of computational biology and agricultural bioinformatics. The centre has identified the thrust areas: (i) To undertake research in priority areas of computational biology and agricultural bioinformatics (ii) To design, development and implement biological databases and data warehouse (iii) To develop human resources in computational biology and agricultural bioinformatics (iv) To provide analytical and computational support to research in the field of computational biology and agricultural bioinformatics (v) To create and promote inter-disciplinary research groups with focus on computational biology and agricultural bioinformatics.

The CABin, ICAR-IASRI, New Delhi in collaboration with ICAR-Indian Agricultural Research Institute (IARI), New Delhi has initiated M.Sc. degree programme in Bioinformatics in 2011 and Ph.D. programme in 2014. At present, around 25 faculty members are involved in teaching the post graduate students in the discipline of bioinformatics. Currently, 13 students have registered for Ph.D. and 8 students have registered for M.Sc. in Bioinformatics at Post Graduate School, ICAR-IARI, New Delhi. Till date, a total of 16 students have been awarded with M.Sc. degree in Bioinformatics.

Significant Student's Research Achievements

Genomics

Various studies taken up under genomics are development of genome browsers, codon bias studies and change point analysis. Genome browsers are very popular in the field of genome annotation by facilitating the graphical visualization of genomic elements on the genome and comparison of the genome of two or more organisms. Though the cattle genome browser has already been developed, there is a need to develop genome browser for buffalo in the public domain. Choudhary (2016) developed buffalo and cattle genome browsers with an updated version of their whole genome sequences and a synteny browser to map the homologous

contigs of both the species using GBrowse software (Fig 1). The developed browsers are believed to help the research communities to gather the knowledge about cattle and buffalo genome and the homologous genes present between them. The developed browser can help to annotate the unknown genes of buffalo genome which are not well annotated till date. Further, the annotation of buffalo genome will help the breeders to know the genes responsible for agriculturally important trait which in turn will help in the development of trait specific buffalo breeds.

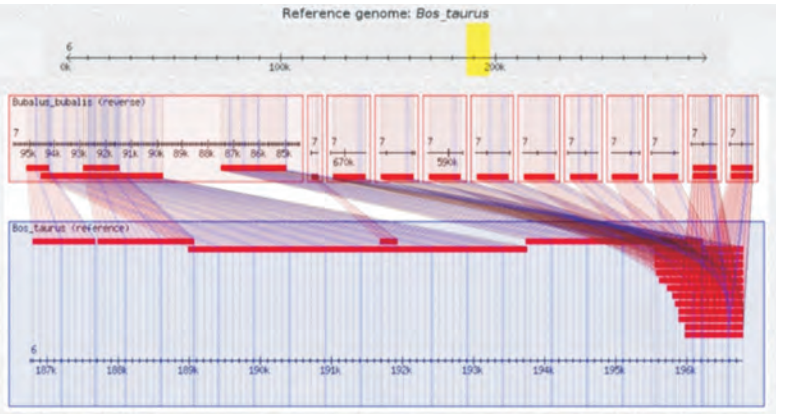

Fig. 1: Synteny between *Bos taurus* to *Bubalus bubalis* (a segment of Chromosome 6 of *Bos taurus* showing synteny with a segment of chromosome 7 of Bubalus *bubalis* (Source: Choudhary, 2016)

The study of codon usage pattern of *Rhizobium* is gaining attention over time. Efforts have been made continuously to understand the codon usage patterns and identify some conserved features of the genus *Rhizobium*. Majumdar *et al.,* (2015) studied the codon usage patterns of the nitrogen fixing genes in order to improve the exogenous gene expression. Three strains of *Rhizobium* namely *Sinorhizobium meliloti* 1021, *Bradyrhizobium japonicum* USDA110 and *Rhizobium tropici* CIAT899 were collected from NCBI (www.ncbi.nlm.nih.gov) for the analysis of codon usage bias. A comparative analysis among these three strains and other six strains of *Rhizobium* was performed. The overall codon usage analysis showed that G and C ending codons are predominant in the *Rhizobium* genome than A and T ending codons. Nc-plot revealed that translational selection along with compositional constraints is the major causes of codon usage bias (CUB) (Fig. 2). The correspondence analysis (COA) showed the first two axes mainly accounted for the maximum variation present in the codon usage data. The Pearson correlation analysis identified significant correlation between the first axes of COA and CAI and other factors of codon usage bias, which indicated that gene expression level played an important role in shaping codon usage pattern of the genus *Rhizobium*. The comparative analysis revealed that the use of cystein codons varied among the different genes in the genus *Rhizobium* and 16 codons that are shared amongst them were identified as a conserved feature of the genus *Rhizobium*.

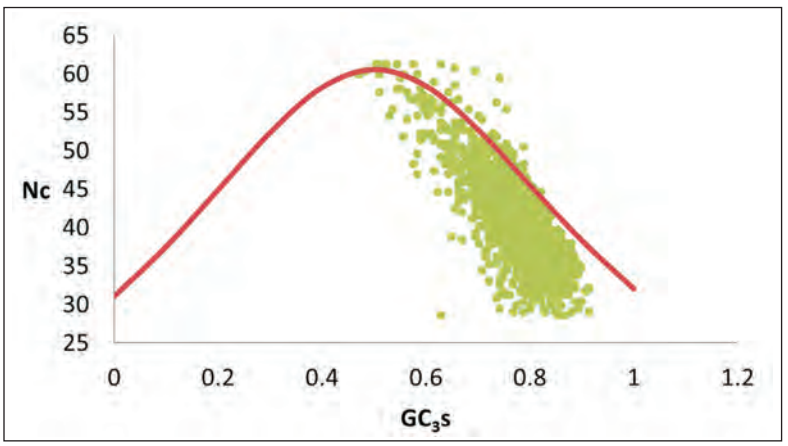

Fig. 2: Nc-plot of *Sinorhizobium meliloti* 1021

Roy (2016) studied change-points in Genomic Sequences. The genomes of complex living organisms sometimes show heterogeneity in relation to a particular biological property or more than one property prevailing throughout the sequence of an organism. These properties may be GC (Guanine-Cytosine) content, copy number variations, Single Nucleotide Polymorphism (SNP) frequency etc. In other words, some areas on the genome are distinct and are termed as segments with "minimum heterogeneity within segments" and "maximum heterogeneity between segments". Change-points describe the boundaries between two dissimilar segments and help to understand biological locations of the start point of heterogeneity within a sequence of genome. A sliding window methodology was developed and used to divide the heterogeneous genome into many segments and identify the change-point intervals. Chromosome 5 of Rice and whole genome of *Escherichia coli*

were considered to detect the regions where significant changes occurred with regard to GC content (Figs. 3a, 3b). It was found that with the increase in sliding window size, the number of significant segments increased. A SAS macro has been developed to execute the said procedure by defining different segment sizes and sliding window sizes.

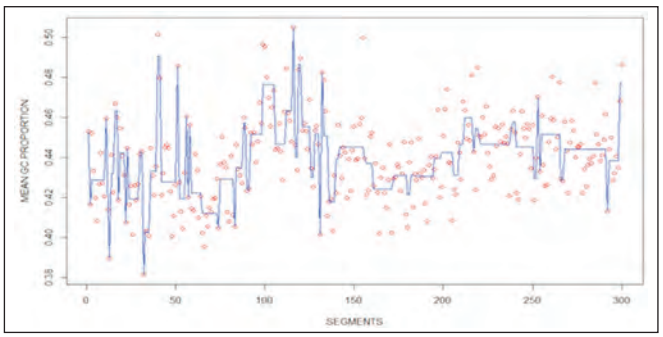

Fig 3a. Segmentation based on mean GC proportion in Rice (Source: Roy, 2016)

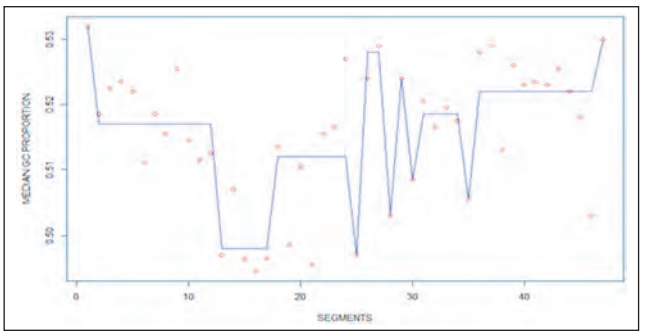

Fig 3b: Segmentation based on median GC proportion in *Escherichia coli* (Source: Roy, 2016)

Transcriptomics

The transcriptomics work done in the discipline involves transcriptome analysis of moisture stress genes, differential gene expression and miRNA identification for late blight of potato. Supriya (2014) identified key genes that play a major role in moisture stress tolerance by *de novo* assembly and annotation of musk melon transcriptome. A total of 47,035,393 and 45,152,235 high quality unique reads for *Cucumis melo* control-sample and stress-sample were assembled into 12859 and 13448 transcripts, respectively using CAP3, DNA STAR and CLC *de novo* assembly programs. Gene ontology distribution reveals 37 functional groups including biological processes, cellular component, and molecular function (Fig 4). Comparison of gene expression levels between control and stress transcriptomes by RNA-Seq mapping revealed 122

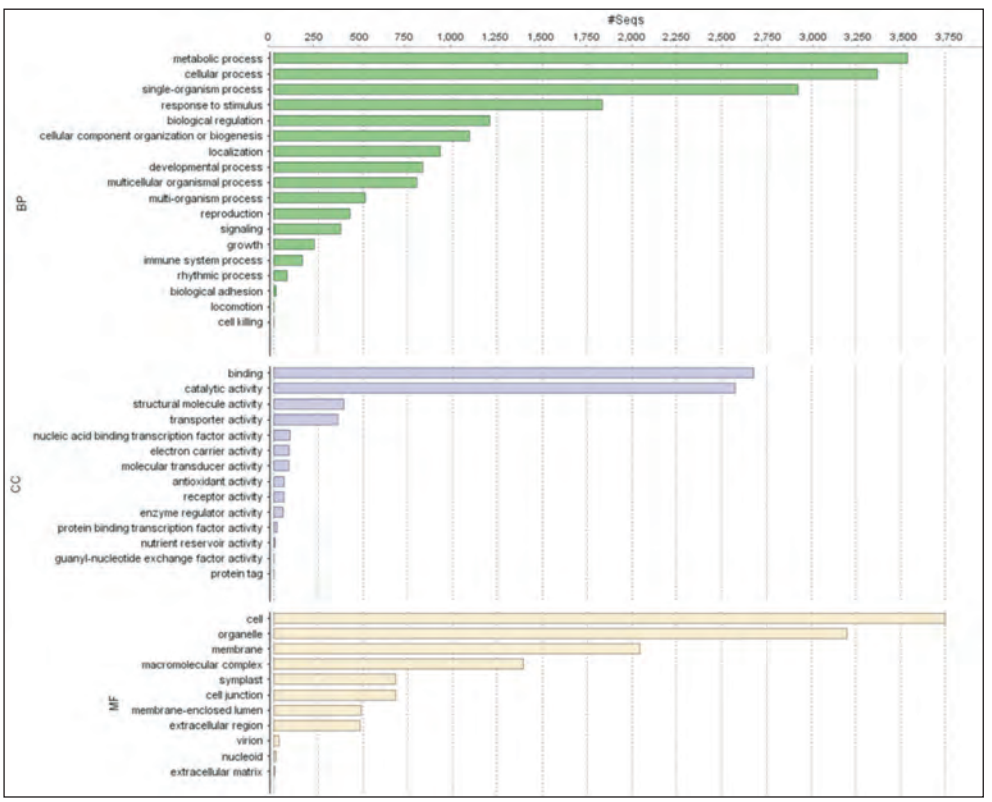

Fig 4. GO Distribution showing various functional groups (Source: Supriya, 2014)

stress responsive genes commonly expressed in both the samples whereas 72 genes highly up regulated in stress sample such as kinases, DREB genes, heat shock proteins, MYB transcription factors, Zinc- finger, and AP2 / ERF domain containing transcription factors. Among these, 4 genes were found to involve in path ways such as plant-hormone signal transduction, protein processing in endoplasmic reticulum and Inositol phosphate metabolism.

It is important to find statistical distributional property of the data to approximate the nature of differential gene expression. Anjum (2015) and Anjum *et al.* (2016) focused on the differential gene expression analysis for sequence data based on mixture distribution model. Comparison was made between mixture/compound distribution and single distribution with respect to the identification of differential expressed genes. Fitting of two-component mixture normal model in case of microarray data is found to be more capable of capturing the variability as compared to single component normal distribution and hence identified differentially expressed genes more accurately. Further, it has been found that in case of RNA-seq data, Negative Binomial as compound Poisson distribution is more appropriate to capture the variability as compared to Poisson distribution. Thus, fitting of appropriate distribution to gene expression data of *Arabidopsis thaliana* provides statistically sound cut-off values for identifying differentially expressed genes (Fig. 5 and Fig. 6).

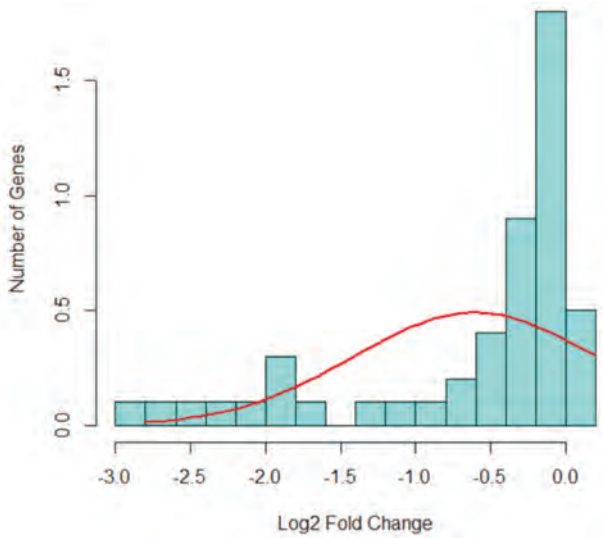

Fig. 5: Normal plot

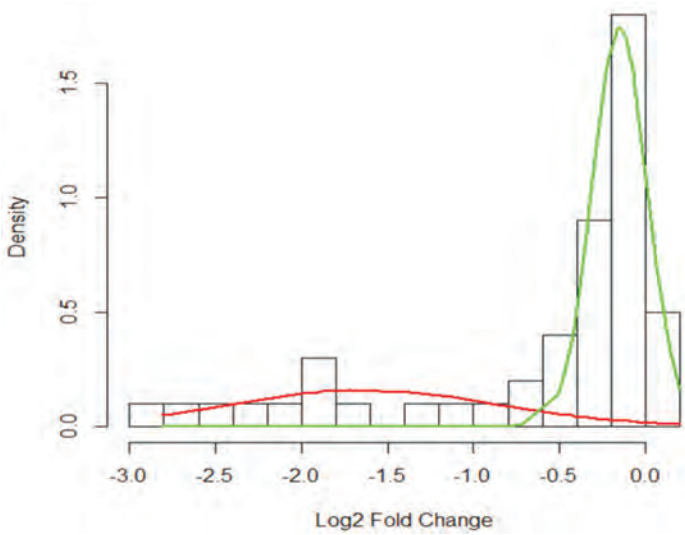

Fig. 6: Two-component mixture normal plot (Source: Anjum, 2015)

Kumar (2015) identified 34 potential miRNA related to 6 different miRNA families *via.* miRNA-miRNA relationship networks, using a total of 23571 non-redundant mature miRNA sequences from miRBase database and 262,006 whole *Solanum tuberosum* EST sequences from NCBI. These miRNA and their targets related to late blight disease of potato canhelp in understanding both the mechanisms involved in development of disease and precise technique for understanding of post transcriptional gene silencing.

Structural Biology

The structural biology studies carried out includes: (i) protein-protein interaction studies in Rice (ii) Effect of mutation on structure of blast resistance gene in rice (iii) Enhanced Disease Susceptibility1 in Brinjal. Sarkar (2013, Ray, *et al.*, 2016) studied the effect of SNPs and InDels on the *pi-54* protein structure and its interaction with Avr-Pi54 protein. The type and numbers of secondary structures were observed to be different in the tertiary protein structure of *Pi54* alleles than the native Pi54 protein derived from rice line Tetep. The stabilization of

the Pi54 protein structure fold was assessed depending on secondary structure, the numbers of H-bonds and the global free minimum energy. The interactions of the Pi54 proteins with Avr-Pi54 protein were also affected by the presence of SNPs and InDels in *Pi54* allele. Out of 74 Pi54-Avr-Pi54 protein combinations, successful interactions were obtained between 52 protein combinations. Besides the SNPs/InDels in LRR region, other regions of the protein also lead to the change in 3D structures of the Pi54 proteins which affects its interaction potential with the Avr-Pi54 protein.

The effect of mutation on proteins of blast resistance gene was analysed by detecting and comparing the natural variations (SNPs and InDels) present in 30 alleles of Pi-ta gene in Oryza sativa (Prabhakar, 2013). Three-dimensional structure of Pi-ta proteins were predicted through the ITASSER software. 3Drefine web server was used to bring the initial model closer to the native structure. Quantitative structural evaluations were performed to find out the stability and disease response of Pi-ta alleles upon mutation. Susceptible indica cultivar C101A51 showed maximum number of InDels events in the gene. The predicted *in silico* protein structure of the Pi-ta alleles was observed to be affected by mutation i.e. SNPs and InDels. Normal mode analysis of modelled proteins showed differences in region of maximum atomic displacements between different alleles of Pi-ta gene. Changes in normalized atomic displacements were observed in susceptible protein ADV02401.1 and resistant protein ADV02400.1. A very less correlation was found between mutations that increase stability and mutations that favour complex formation.

Enhanced Disease Susceptibility1 (EDS1) is a nucleo-cytoplasmic protein, known to be a key regulator of plant basal defense and effector triggered immunity initiated via TIR-NB-LRR class of R proteins. Sharma (2016) extracted detailed information of EDS1 protein in brinjal through in silico approach. SmEDS1 gene (1806 nucleotides) in the contig Sme2.5_09498.1 of its draft genome assembly encoded for 602 amino acid long protein. The comparative analysis of SmEDS1 protein with 46 other species EDS1 supported strong sequence and structural conservation of this protein among plants. The incongruence in sequence and structure based phylogenetic trees was observed that could be attributed due to the influence of difference between global alignment and conservation of sequence signatures or by the fact that sequences are related by phylogeny whereas structures by constraints on their functions and regulations. Further phylogenetic studies based on EDS1 can facilitate its genetic manipulation for agronomic purposes.

Databases and Tools

Kairi (2013) developed an information system on buffalo genome. The developed information system contains 930 complete CDS, 1154 partial CDS, 656 Exon sequences, 237 Intron sequences, 1709 Mitochondrial DNAs, 73 sequences of Promoter Region and 67 sequences of UTR region. Out of 930 buffalo genes, 837 have been found to be mapped onto *Bubalus bubalis* (female Murrah breed). The developed genome browser shows that maximum number of genes are distributed on chromosome 4 followed by chromosome 18. The total number genes present on chromosome 4 and chromosome 18 are 324 and 155 respectively. The results from the study also reveal that 837 out of 930 genes are mapped onto *Bubalus bubalis* genome. Whereas, 561 buffalo genes are mapped onto *Bos taurus* genome. Further, 202 genes are found to be predicted as orthologues between cattle and buffalo genomes. The developed information system provides several user-friendly facilities like search, filter, download, print, visualisation and browsing. The information system on buffalo genome can be accessed at http://cabgrid.res.in:8080/bgis.

Epigenetics is an emerging field which deals with the study of mitotic and meiotic (or both) heritable changes in gene function that cannot be explained by changes in DNA sequence. Majumdar (2014) developed an Epigenetic database in livestock species. The developed information system can be accessed at http.//bioinformatics.iasri.res.in/edil/. The newly developed Livestock Epigenetic Information System contains CpG island information of 8033 genes of *Bos taurus* (Fig 7). Besides, it contains 798 and 105 precursor miRNAs of *Bos taurus* and *Ovis aries* respectively along with their secondary and tertiary structures; 783 and 153 mature miRNA sequence of

Bos taurus and *Ovis aries* respectively; different types of histone protein information of cattle, goat, sheep, camel. The present information system also provides predicted structures of histone proteins along with their pdb files. The structures present in the information system are the refined, validated and stabilized structures. The web-interface developed under the present investigation provides a facility to visualize the genomic positions of buffalo chromosomes which are orthologous to miRNA sequences of *Bos taurus*.

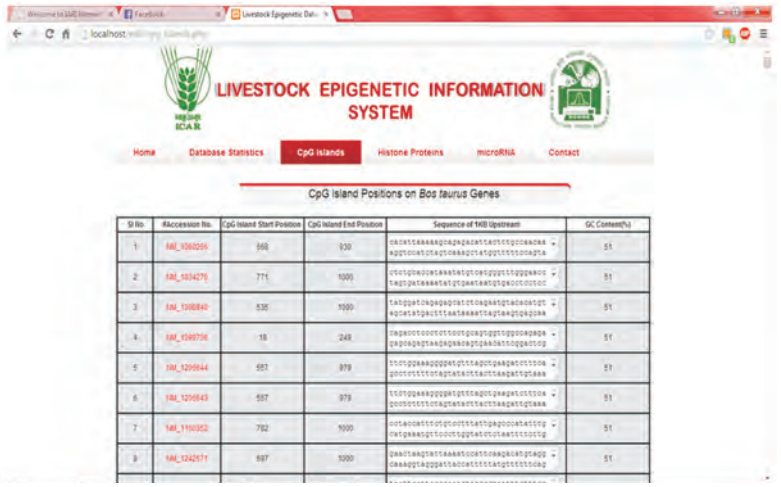

Fig. 7: CpG Island information (Source: Majumdar, 2014)

In silico Prediction of function of an unknown protein is important in understanding the regulation of related biological process of the plant for mitigation of adverse effects of abiotic stress. Budhlakoti (2014) developed a Perl code to compute the physicochemical properties of unknown protein sequences as an alternative to ProtParam tool. The computed physicochemical properties are used as for prediction of function of proteins. A web server for the prediction of proteins has been further developed. It is capable to predict the function of any given protein sequence into five classes of abiotic stresses, *viz*., Heat, Cold, Drought, Salinity, ABA in cereals.

Crop Wild Relative (CWR) is a wild plant species closely related to crops, including wild ancestors. Application of genomic tools in breeding has brought the CWRs in the limelight. Thus it has become possible to transfer useful alleles from wild sources. *Vigna* and *Phaseolus* species are indigenous crop species with high nutritional value but there exists a valuable knowledge gap. Its utilization in crop varietal development programme is limited to non-availability of compiled and collated information of *Vigna* and *Phaseolus* species. Hence, there is a need to develop a single window information system which can facilitate enhanced utilization of valuable germplasm resources along with genomic resources for crop improvement programme. Khan (2015) adopted a three tier architecture in developing CWR resource database for *Vigna* and *Phaseolus* species. The database layer was designed on the basis of ER diagram among entities as taxonomy, germplasm, EST, nucleotide, protein and gene. The genomic data also contained the sequences of each entry, which increased the complexity and need to store in the database. The sequence information was parsed through a Perl script into a fasta file with GI number and made a source directory of corresponding fasta sequence file.

A client side application layer was provided with a graphical user interface to describe the significant connects between genebank and genbank information. One can search either with scientific name, common name, GI, trait or genebank name which would provide all details of taxonomic classification, germplasm conservation and genomic sequences of the *Vigna* and *Phaseolus* species. A facility to download tabular information of germplasm, EST, nucleotide, protein and gene was provided. Also output in MS Excel format was provided. An advanced search to join the genebank with genbank information was provided to add value to germplasm for their enhanced utilization. The developed system to link a specific gene, EST, nucleotide and protein with a functional importance to a germplasm accession may assist molecular breeders in incorporating the germplasm accession in precision breeding programme.

Summary

Keeping in view the aims of bioinformatics, the students at Bioinformatics discipline of Post Graduate School, IARI conducted research work under able-guidance of their respective chairpersons and came out with the following findings:

- GBrowse has been configured to show the synteny between cattle and buffalo genomes. The developed genome browser can help in predicting genes responsible for trait expression in buffalo species.
- Codon usage patterns of nitrogen fixing genes have been studied to improve the exogenous gene expression in three species of Rhizobium.
- Change-points, describing the boundaries between two dissimilar segments, have been studied through statistical distributions to understand biological locations of the start point of heterogeneity within genomes of rice and *Escherichia coli*.
- Comparison of gene expression levels between control and stress transcriptomes of musk melon by RNA-Seq mapping revealed 122 stress responsive genes commonly expressed in both the samples whereas 72 genes highly up regulated in stress sample such as kinases, DREB genes, heat shock proteins, MYB transcription factors, Zinc- finger, and AP2 / ERF domain containing transcription factors.
- Negative Binomial as compound Poisson distribution is more appropriate as compared to Poisson distribution to capture the variability in RNA-seq data of *Arabidopsis thaliana*.
- 34 potential miRNA related to 6 different miRNA families *via.* miRNA-miRNA relationship networks have been identified by using non-redundant mature miRNA sequences from miRBase database and *Solanum tuberosum* EST sequences from NCBI. The identified miRNA and their targets related to late blight disease of potato may help in understanding mechanisms involved in development of disease and post transcriptional gene silencing.
- The effect of SNPs and InDels on the *pi-54* protein structure and its interaction with Avr-Pi54 protein has been studied. It was found that the interaction of *pi54* proteins with Avr-Pi54 protein were affected by the presence of SNPs and InDels in *Pi54* allele of rice line Tetep.
- The effect of mutation on proteins of blast resistance gene was analysed by comparing the natural variations (SNPs and InDels) present in 30 alleles of *Pi-ta* gene in *Oryza sativa*. Normal mode analysis of modelled proteins showed differences in the region of maximum atomic displacements between different alleles of *Pi-ta* gene.
- A comparative analysis of SmEDS1 protein of brinjal with EDS1 gnes from 46 other species supported a strong sequence and structural conservation protein among species.
- A total of 837 genes out of 930 genes of *Bos taurus* available in public domain are mapped onto *Bubalus bubalis* genome. Whereas, 561 buffalo genes are mapped onto *Bos taurus* genome. Further, 202 genes are found to be predicted as orthologues between cattle and buffalo genomes.
- A Livestock Epigenetic Information System was developed, which contains CpG island information of 8033 genes of *Bos taurus*. It also contains 798 and 105 precursor miRNAs of *Bos taurus* and *Ovis aries* respectively along with their secondary and tertiary structures.
- A Perl code was developed to compute the physicochemical properties of unknown protein sequences as an alternative to ProtParam tool. A web server for the prediction of proteins has been further developed. The server is capable to predict the function of any given protein sequence into five classes of abiotic stresses viz., Heat, Cold, Drought, Salinity, ABA in cereals.
- A three-tier architecture was followed to develop Crop Wild Relative (CWR) resource database for *Vigna* and *Phaseolus* species. A client side application layer was provided with a graphical user interface to describe the significant connects between genebank and genbank information to add value to the germplasm for their enhanced utilization.

Acknowledgement

We wish to thank all the Faculty Members of the Division of Bioinformatics for extending support and cooperation in preparing the research contributions of M.Sc. and Ph.D. students made during the period of 2007-16.

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Computer Applications

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Introduction

The present era has seen an exponential growth and diversification in all forms of information, sometimes called, an information explosion. It has been made possible due to the impact of computer or information technology on the modern society. Computerized information systems have influenced nearly all domains. The domain of Agriculture is no exception with its application in various sectors of agriculture. Multiple types of IT based systems have been developed by the M.Sc. students of Discipline of Computer Application of Post Graduate School, IARI over the years to increase their use in the farming and agricultural research community. The research is mainly focused on developing online information systems, decision support systems, expert systems, knowledge base and more recently mobile apps for dissemination of developed technologies, information and knowledge to the farmers or agriculture researchers.

- Crop based Online Information Systems for Farmers
- Online Systems for Transfer of Technologies and Farming Equipment Information
- Online Systems for Analysis of Agriculture Experiments
- Statistical Software for Analysis of Data
- Online Analytical Processing Software
- Online Systems for Water Technology
- Expert Systems & Knowledge Management
- Online Systems for Agriculture Economics
- Software for Data Mining
- Geographical Information System Related Software
- Software for Image Analysis
- Mobile Applications

Significant Student's Research Achievements

Developed software have features like query and report generation for easy retrieval of information. Authorized user access through user name and password. Multiple type of users such as end user and administrator for rights and privilege management. Many software have email alert and FAQs facility also. In the following sections, theme wise various research work undertaken by the M.Sc. students under their respective chairpersons have been discussed.

Crop based Online Information Systems for Farmers

An Information system is a system which captures, stores, retrieves, manipulates and displays information. The specific information provided aims to improve operation, better management and easier decision making. Information Systems can be described as an integration of components which collect,

store and process data to deliver information. It could be composed of both humans and computers which processes information.

Following are the information systems developed for bringing specific interventions

Information System on Post-harvest Management of Citrus Fruits (Kinnow Mandarin) (Nisha, 2007): Online Information Systems have great importance in horticulture as experts are not always available to answer Farmers' query. Information System on Post-harvest Management of Citrus Fruits (Kinnow mandarin) (InPhoCFK) is a Web-based Information System to provide information on post-harvest management of citrus fruits. InPhoCFK has modules to provide information on various aspects of citrus fruits like general information, state wise statistics, harvesting methods, storage practices, packaging, transportation modes, processed products, and processing industry details etc. InPhoCFK is developed using ASP.NET and database part is developed using MS SQL Server.

Information System on Herbicide use in Field Crops (ISHUFC) (Chayana, 2007): Every crop is exposed to severe competition from weeds. So, proper weed management is a pre-requisite for obtaining higher input efficiency. The lack of awareness, information about the availability and the application of herbicides is one of the main reasons for their limited use. Sometimes, the farmers may not know the right dose, time of application and the procedures for the application of herbicide. Because of this farmer doesn't get expected results in spite of its use. Information System on Herbicide use in Field Crops (ISHUFC) is a Web-based Information System to provide information to Extension Personnel, Farmers, Students, Researchers etc. for proper herbicide recommendations in field crops. ISHUFC has a simple query and report generation module to provide the information about crops, weeds, herbicides, herbicide recommendations etc. even in the printable formats. ISHUFC is developed using ASP.NET, an easy and effective tool to develop web based applications. Database part is developed using MS SQL Server.

Online system for Onion Thrips Forewarning System (OTFS) (Subash, 2015): Onion, Allium cepa is important vegetable crop and onion thrips is most damaging pest for this crop. Timely forecast can help in taking remedial measures for the same. Statistical models have been developed at ICAR-IASRI based on weather parameters for forecasting different aspects relating to thrips. The models have been developed for different planting dates for the data taken from ICAR-DOGR. Models have been developed for thirteen dates of sowing for the forewarning of crop age at first appearance of thrips, crop age at peak population of thrips and maximum thrips population. The models have been tested and used by DOGR for forewarning purpose. To disseminate the information in effective manner, online software has been developed based on the weather models. Online system for Onion Thrips Forewarning System (OTFS) has been developed using three-tier architecture. Client layer is implemented using Hypertext Markup Language (HTML), Cascading Style Sheet (CSS) and Java script. In application layer all the programing for models have been done using Java Server Pages (JSP). Database layer for storing the weather data has been implemented in Microsoft SQL Server. User can access the system using web browser. Administrator is another user to whom access rights for data management has been provided. Administrator can upload the weather data in excel sheet to the system. Results have been validated by running the models in SAS software and online software. The developed system will help the farmers, agro-advisors and agricultural extension personnel in forewarning of disease in a timely manner.

Development of Distributed Framework for Crop Protection (Sanober, 2007): Crop protection plays an important role in increasing the yield of crop by identifying the disease, insect and weed infestation and also suggests several treatment and control measures to protect the crop from all these threats. In India, around 30% losses occur in crop due to these pests. Number of information systems and expert systems for different crops are already operational. These information systems and expert systems provide an information on various aspects of crop protection to farming community. The user has to access different expert systems for different crops, so there is a need for building a single window application to provide disease, insect and weed advisory on multiple crops at the same place. For this reliable distributed framework has been developed. A

distributed framework is a framework which captures, stores, retrieves, manipulates and displays information from different sources. The fundamental purpose of a distributed framework is to access data from different sources that is logically related and physically distributed. This thesis presents the design and development of distributed framework for crop protection which provides disease, insect and weed related information from different sources at a single place of wheat and several spices crop. These information will help the farmer in identifying disease, insect and weed infecting the crop and will also suggests control measure to protect the crop from all these threats. The developed system also consist of a form based alert system which provides an online form in which the farmer has to fill some detail. On the basis of temperature, humidity and rainfall value provided by the farmer after filling the form, it gives an alert about the disease, which is going to affect the crop at that particular value.

Online Systems for Transfer of Technologies and Farming Equipment Information

Multiple software have been developed for transferring the technologies to the farmers and other stakeholders

Information System for Agricultural Products and Pricing : India being a large and diverse country with a majority of the population dependent on agriculture and allied activities, the availability of information regarding latest agricultural products, technologies and inputs to the farmers is of vital importance. Information is key to better production and productivity as a large percentage of the farmers in rural areas still follow traditional inefficient products and practices resulting in less remunerative returns. A web based information system will greatly help farmers seeking relevant information regarding products and the traders selling such products. The Indian Council of Agricultural Research (ICAR) has come up with a single window delivery system for services and products of research of agricultural research institutes all over the country called the Agricultural Technology Information Centre (ATIC). They provide farmers with the latest agricultural innovations and products developed at their respective research institutes. But, for getting the information from such centers requires personal visit to these centers which is not always possible by the distant farmers. Information about the various products available at such centers needs to be disseminated to the distant rural areas. The presented web based information system provides information on prices of seeds and planting material of latest varieties, hybrids, and advanced agricultural equipment being developed by ICAR.

Information System for Dissemination of Ready to Adopt Agricultural Technologies (ISDAT) (Sumit, 2007): It is a web based user-friendly, information system for ready to adopt agricultural technologies of ICAR. It is developed as comprehensive information system for ready to adopt agricultural technologies. ISDAT acts as a transfer agent for dissemination of ready to adopt technologies. The information, which is available in the form of books, journals, popular magazines, etc., is beyond the reach to each and every person across the world. Therefore in the electronic era the ISDAT may help progressive farmers, industrialists, researchers, students and extension workers for rapid dissemination of knowledge and benefit humankind. It has a three-layered architecture. Client Side Interface Layer is implemented in HTML and JavaScript. Server Side Application Layer is implemented in Java Server Pages and Java Database Connectivity. Database Layer is implemented in Microsoft SQL Server. The software has one level of authentication i.e. Administrator; which has the privilege to add, modify or delete the information from the database.

Critical Evaluation of Indian Web Portals on Agriculture (Sandip, 2007): Like other fields, web portals have also a major role to play in agricultural field. Agricultural web portals are expected to make farm families more productive, keep farmlands fertile, strengthen rural infrastructure support and help promote a healthy business and social environment through providing the agricultural communities timely and updated information/ services. There are many web portals addressing different sectors of agriculture such as research, development, input supply, protection against pests and diseases, weather, prices and marketing, export/import, farmer associations, government policies, taxation etc., but there are no evaluating tools that could help evaluate the existing agricultural web-portals, guiding the farmers/users in selecting appropriate web portal in their domain

of interest and also to suggest for better designing of web portals in agriculture which are farmers / end users oriented. The research work, "Critical Evaluation of Indian Web Portals on Agriculture", has been carried out with the objective to study and review some popular web portals, followed by identification of the indicators for discrimination among the web portals using Delphi technique and lastly the development of an online evaluation system for evaluating all Indian agro-web portals. Finally, the online evaluation system, named Evaluation System for Agricultural Web Portals (ESAP), that evaluates agricultural web portals and generates automated evaluation reports with proper charts after an interval of seven days, has been implemented using HTML, Java, JSP, PL/SQL and Oracle 9i technologies.

Developing Agents for Bibliographic Data Retrieval System (Murari, 2015): Bibliographic records are used quantitatively by a "Bibliometrician" for analysis and dissemination purpose but with the increasing rate of literature publication in open access journals such as Nucleic Acids Research (NAR), Springer, Oxford Journals etc., it has become difficult to retrieve structured bibliographic information in desired format. A digital bibliographic database contains necessary and structured information about published literature. Bibliographic records of different articles are scattered and resides on different web pages. This thesis presents the retrieval system for bibliographic data of NAR at a single place. For this purpose, parser agents have been developed which access the web pages of NAR and parse the scattered bibliographic data and finally store it into a local bibliographic database. Based on the bibliographic database, "three-tier architecture" has been used to display the bibliographic information in systematized format. Using this system, it would be possible to build the network between different authors and affiliations and also other analytical reports can be generated.

Online Systems for Analysis of Agriculture Experiments

Following systems have been designed using three-layered architecture. Client Side Interface Layer is in HTML and JavaScript, Server Side Application Layer uses Java Server Pages (JSP) or ASP.NET and Java/ ODBC Database Connectivity. Database Layer is implemented using Microsoft SQL Server. Software also have extensive help document with involved statistical concepts, user manual, example data file, example of input files and output files.

Information System for Varietal Experiments (Rakesh, 2009): National Agriculture Research System has number of regional stations in diverse agro-ecological areas to test and evaluate varieties. These stations generate voluminous information on the performance and stability of the varieties. Information System for Varietal Experiments (ISVE) is a Web-based Information System to provide information to extension personnel, students, researchers etc. about zone, centers, variety, trial type, trial series etc. ISVE is developed using ASP. NET and database is developed using SQL Server.

Online Information System for Intercropping Experiments (OISIE) (Ramjilal, 2009): OISIE is an information system for retrieving information related to On Farm and Off Farm activities of Intercropping Experiments. It is developed for information management of all the intercropping experiments conducted in India. OISIE provides information regarding Intercropping Experiments including Experimental site history, Location details, Design details, Objective of the experiment, Treatment details, Soil types and their texture, Season in which the experiment was conducted, Basal condition details which in turn will include Sowing dates, Seed rates, Spacing, Basal manuring, Preparatory cultivation, Planting methods, Irrigation details Date of Harvest for both main crop and inter crop and some General information like Disease and Pest attack, crops condition etc. OISIE provides search facility for centre information, experiment information, treatments applied, main crop and inter crop information's, fertilizer doses, design information, experimental data in case of analyzed experiments and results in case of analyzed experiments.

Development of software for cropping system experiments (Jai, 2011): Cropping system experiments are conducted under the guidance of Project Directorate of Farming System Research for crop diversification and intensification. Also, the use of computers has changed the whole complexion of research. With the development

of Statistical software packages, the statistical analysis of data has become relatively easy. Now it is possible to carry out sophisticated data analysis in no time. With the rapid advancement in the field of information technology, use of web applications is heavily increased. Keeping this in view, Web based Software for Cropping System Experiments has been developed. The software provides the season wise results for analysis that includes Character analyzed, Centre name, Experiment type, Raw and Converted data Season wise, ANOVA, Mean table, Standard Error, and Critical Difference.

Information system on cropping sequence experiments (Anesh, 2007): Large number of agricultural field experiments is conducted in the country for the development of new agro-technologies. The results of experiments are of immense practical value and play crucial role in increasing the productivity of various crops. For increasing the cropping intensities the research is directed to crop sequence and intercropping experiments. Cropping Sequence refers to an arrangement of crops in temporal and spatial dimension on a given piece of land and their interactions with farm resources and available technologies. Sequence cropping comprises the planting of two or more crops in the same plot one after other. Cropping Sequence Experiments Information System (CSEIS) site provides information related to Cropping Sequence experiments, conducted at various agricultural research stations in the country, their data and results. The information has been hyper-linked so that users can view the information in an easy manner. It aims to act as reference material for scientists, research workers and those who are working in the field of agricultural sciences. As improved systems on component technologies will become available, they will replace or be linked to the system. This information system will provide information's regarding following aspects of the cropping sequence experiments: location details, experimental site history, cultural and other practices followed, objectives of the experiments conducted, details about treatments, details about design used, general crop conditions, summary results and / or plot wise observations for each of the crops in the sequence and over all results obtained.

Software package for knowledge extraction from agricultural field experiments (Mrityunjoy, 2011): The software provides the result for reporting the agricultural experiments including Character analyzed, Research centre name, Experiment type, Raw and Converted data Season wise, crop and variety wise, seed rate, spacing, amount of fertilizer and pesticide application for particular crop and the yield of the on-station experiments. The consolidated report for on-station on the basis of experiment-wise, state wise, characters analyzed and Year wise.

Statistical Software for Analysis of Data

Statistics is a science of drawing conclusions on uncertainty and deals with collection, presentation, analysis and interpretation of numerical data. One of the main objectives of statistics is to draw valid inferences about a population from the analysis of a sample drawn from that population. Following software packages have been developed in various sub domains of statistics to help the researchers for analyzing their data:

Software for Fitting of Distributions (Ashutosh, 2009): The theoretical probability distributions provide decision makers with a logical basis for making decisions on the basis of limited information. For example, the distribution of weeds over a unit area in a field crop is fitted by Uniform distribution; but distribution of insects in a field is fitted by Poisson distribution. Similarly, Logistic and Pareto distribution are used to fit population data. Thus, one must be careful while choosing the correct probability distribution as per the situation being faced. The theory of estimation forms the foundation of statistical inference which uses the probability concepts to deal with uncertainty and decision making. The concept of estimation of parameters can't be visualized until and unless the distribution function of population is identified and thus distribution fitting also plays a significant role in finding various statistical constants of the population. Thus an attempt has been made to develop software that can fit certain number of discrete and continuous distributions as per the real life situations and fit the theoretical data using Kolmogorov - Smirnov and Chi-Square tests of goodness of fits, once the parameter have been estimated (broadly using MLE techniques). Leading statistical software including SAS, SPSS, and

Minitab etc. are already available on this aspect yet no indigenous and/or web-enabled software has thus far been developed at IASRI. Keeping this in view, this software has been developed using Visual C# language with ASP.NET 3.5 framework. Data file formats supported by this software are .txt and .xls. In addition to fitting of distribution, the software can also be used interactively to draw Quantile-Quantile plot for continuous distributions; and computing various special functions (including Gamma, Beta, and Incomplete Gamma etc.) at runtime. A user friendly help file with step-by-step method for using the software has also been provided.

Development of statistical package for analysis of cropping system experiments (Ragini, 2009): Statistical Package for Analysis of Cropping System Experiments (SPACSE) is Web based software for the analysis of the data collected from farmers' field trials at various NARP Zone levels and the data collected from On-Station trials. The intent of SPACSE is to provide an easy-to-use statistical analysis program for the novice user. SPACSE supports randomized complete block designs and allows the number of treatments and replications to be variable. SPACSE provides the analysis and consolidated report for grain yield, straw yield, N uptake of grain yield, N uptake of straw yield, P uptake of grain yield, P uptake of straw yield, K uptake of grain yield and K uptake of straw yield for On-farm experiments. It provides the analysis and consolidated report for grain yield and straw yield for On-station experiments. The result for analysis include Character analyzed, Crop-sequence, Year, Type of Experiment, Variety, Treatment details and Raw and Converted data, ANOVA, Mean table, Standard Error and Critical Difference for cropping systems experiment. The consolidated report for On-farm experiments is year-wise and for on-station trials, it is experiment wise. There is no restriction on the number of replications and treatments.

Software for online analysis of split-plot designs (Robin, 2009): Split plot design is often used by the experimenter, where his objective is to compare two or more factors requiring different larger plot sizes for operational convenience. Factor(s) which require larger plot size are applied to the bigger plot called main plot and the one which require smaller plot size are applied to the plot called sub-plot. The experimenter may also have treatments either in the main plot or in sub-plot which are not part of factorial set up and are called control or extra treatments. From the review of software available for data analysis it is seen that no software is readily available to the agricultural experimenter for carrying out the analysis of data generated, under such type of experiments, particularly in situations when there are more than two factors and a control/ extra treatments. This software has the facility to analyze split plot design of upto four factors with or without control treatments. The control/extra treatments are provided in the sub plot. It also provides facility for user to create his own data files for analysis upto split plot design of four factors with or without control/extra treatment. This software is made very simple to use and also a help menu is provided in the package.

Software for stability analysis using AMMI and SREG models (Iti, 2007): Yield stability as a selection trait in plant breeding programmes and evaluation trials is constantly gaining importance over yielding ability. Some of the common techniques as an alternative to additive ANOVA model are Additive Main effects and Multiplicative Interaction (AMMI), Sites Regression (SREG), Factorial Regression, Joint regression etc. Software package for stability analysis is developed using AMMI and SREG models. It has been developed using VC++ and VB. There is no restriction on the number of response variables and observations. This module calculates environment-wise ANOVAs, pooled ANOVA, percentage explained by each Principal Component Analysis (PCA), Gollob test for AMMI analysis and also the biplots of mean vs. First PCA and First PCA vs. Second PCA. It has been provided with an extensive Help document, regarding statistical concepts involved, how to use the software, example data file, example of input files and output files. It has also the options like favorites and search through contents and index.

Some statistical modules for analysis of breeding data (Nongmaithem, 2007): This package has the programs for analysis, namely, Homogeneity testing of variance-covariance matrices by likelihood ratio test, Mahalanobis D2 analysis, Distance analysis by Iterative Minimax and Oblique Axes procedures. Further, two programs viz., calculation of expected responses to recurrent and reciprocal recurrent selections are also included. The package is built using Java Swing to create window based editor and Core Java to develop the statistical modules.

The user is also provided with the information needed to make data and parameter files. This information is available in the help as well as the module's window. The overall architecture of this windows based software can be regarded as two tier with Java Swing as front end and Core Java as back end.

Software for stability analysis using factorial regression (Dibyendu, 2007): Yield stability as a selection trait in plant breeding programmes and evaluation trials is constantly gaining importance over yielding ability. Some of the common techniques as an alternative to additive ANOVA model are Additive Main effects and Multiplicative Interaction (AMMI), Sites Regression (SREG), Factorial Regression, Joint regression etc. Whenever the information on external environmental characteristics such as weather parameters and soil characteristics is available, it may be accommodated in the basic ANOVA model mentioned above in an attempt to interpret the interaction using the technique called Factorial Regression. The software for stability analysis using Factorial Regression has been developed using Visual C++ 6.0 and Visual Basic 6.0 which is more flexible, user-friendly and economic.

Software for multivariate analysis of variance for general block design (Sazia, 2010): Experiments in which data on several responses are measured from an experimental unit corresponding to the application of a treatment are known as multi-response experiments. There is a tendency to analyze data from these experiments as a single response experiment in the sense that each response variable is analyzed separately as if the responses were independent. The response variables are generally correlated. In analyzing data from each response variable separately, the advantage of the correlation structure is lost. Therefore, to take advantage of correlation structure between several response variables in multi-response experiments, the data must be analyzed using multivariate analysis of variance. Keeping in view this, a web enabled "Software for Multivariate Analysis of Variance for General Block Design" has been developed using ASP.NET and C# language, which is more flexible and user-friendly. There is no restriction on the number of variables and observations. The software provides the facility to upload csv or excel data files. It provides MANOVA under the General Block Design set up. It also provides estimated genotypic and phenotypic variance-covariance matrices, heritability coefficient and genetic correlations under an incomplete block design set up. Several useful links have been provided in the home page of the software to grasp the subject matter.

Web Based Software for Estimation of Regression Coefficient (Sarita, 2009): A sample survey is a process for collecting data on a sample of observations which are selected from the population of interest using a probability-based sample design. Statistical software, mostly, assumes that the observations are selected independently and that each observation had the same probability of being selected and does not take in to account few common characteristics of survey data: (i) clustering of observations, (ii) stratification, etc. Some available softwares for survey data analysis are PC CARP, SUDAAN, STATA, WesVarPC, CENVAR, CLUSTER, EPI INFO, VPLX, etc. which are applicable on personal computers. Most of these packages have more extensive features than only estimation for complex survey data and they have some limitations also. Software packages such as SUDAAN, PC Carp and Stata uses the Taylor Series approach for variance estimation. Whereas WesVar and WesVarPC and VPLX packages use replication approaches to variance estimation. Most of these softwares don't provide estimate of the regression coefficient for survey data. With the advent of information technology, a need was felt to develop software for the estimation of regression coefficient for survey data which should be more flexible and user-friendly. Keeping this in mind an attempt is made to develop the web based Software for Estimation of Regression Coefficient (SERC) for survey data. This software can upload the data from MS-excel/ MS-access files. Data files can be opened, deleted and saved as done in other web applications. SERC has the two analyses modules- i) descriptive statistics (mean, variance, and coefficient of variance), ii) Estimation of regression coefficient and estimate of its variance. SERC can analysis the data if it is obtained using simple random sampling and stratified sampling designs. On line help is provided regarding formulae used for various sampling schemes and using SERC. SERC is developed using ASP.NET.

Development of web based software for generation and analysis of partial diallel crosses (Nishikant, 2009): The breeding experiments for plant and animal breeding experiments involve two types of designs, namely

mating designs and environmental design. Mating design is a procedure of producing the progenies, while environmental design is subjecting these progenies to the environmental conditions in a systematic manner. Diallel and partial diallel cross plans are two of the commonly used mating designs. In a complete diallel cross (CDC) plan as the number of lines increases, the number of crosses increases rapidly resulting in difficulty to handle all of them effectively. Hence, it is always desirable to go for a subset or sample of all possible crosses, which is known as partial diallel crosses (PDC). Most of the packages useful in generation and analysis of partial diallel crosses are available either as stand-alone or as a part of standard statistical software package. This software webPDC is created with keeping all these views in mind with this web services are also created in this software so that developers can use developed modules and save application development time and can work further. Keeping this in mind an attempt is made to develop the web based software for generation and analysis Partial Diallel Crosses (webPDC). webPDC provide PDC crossing plans, and analysis of PDC crossing plan is also carried out, with this different association schemes are also developed and web services are created for all these module which are platform independent and easy to consume. This software can upload the data from MS-excel files and ANOVA table with mean table is presented to user and result can be saved in MS-Excel files on users hard disk on desired location. Online help is provided regarding PDC plans, analysis, association schemes and web services. webPDC is developed using ASP.NET and C#.

Software for the Online Analysis of Row-Column Design (Inderjeet, 2012): Experimentation is an integral part of any research endeavor. Designing an experiment is therefore very important so as draw valid inferences from the data generated from the experiment keeping in view the objectives of the study and hypothesis to be tested. Block designs are used when the heterogeneity present in the experimental material is in one-direction. However, when the heterogeneity present in the experimental material is in two directions i.e. rows and columns, then double grouping is done which eliminates from the errors all differences among rows and equally all differences among columns. Designs used for the above situations are termed as Row-Column Designs (RCD) or designs for two-way elimination of heterogeneity. Due to their wide use in agricultural experimentation, a web based software for analysis of the data generated for RCD has been developed. It has been developed on .Net framework using C#. The software gives the analysis of row-column design with matrix method. A statistical engine has been developed in the form of an object oriented C# library for analysis as per the statistical methodology. It also provides the analysis of several character simultaneously. The software has five modules namely ANALYSIS, SAMPLE DATA, CONTACT US, FEEDBACK and HELP accessible to the user through the Home page. Data input is from an Excel file on client side which is analyzed to give the analysis for RCD, p-values, R-square, coefficient of variation, root mean square error and mean of character. It also provides a framework which is easy to use, extend and integrate with other .Net compatible software tools.

Online Analytical Processing Software

In India, the agricultural resources are spread across the nation. Therefore, the first and foremost need for developing a decision support system on agriculture resources is to integrate the scattered historical information into a central data warehouse. Integrated National Agricultural Resources Information System (INARIS), which is developed by IASRI, New Delhi, is an endeavor in developing a central information repository of major agricultural resources. In the following software, the data marts are created from various data sources. The data storage has been converted into a form of multidimensional model, known as cube. These cubes have been designed by using fact and dimension tables and deployed on Internet for on-line analysis, which is called as On-Line Analytical Processing (OLAP). The cubes have been integrated with previously developed data warehouse INARIS. The information in this data warehouse is available to the end-user in the form of decision support system in which all the flexibility of the presentation of the information, such as it's on line analysis is inbuilt into the system. The data in the developed cubes can be viewed in cross tab view as well as graphical views including simple bar graph, pie chart, clustered bar graph, stacked bar graph, multiline graph, three dimensional bar graph etc. Drill downs and roll ups can be performed on the data available in the cubes.

Another important functionality incorporated in these cubes is Drill Through in which user can find interesting trends or anomalies while analyzing data. The advantage of this approach is that the often query-intensive work of ad hoc data analysis is performed using summarized data in the cube.

Design & Development of Data Marts for Household Amenities from Census Data (Maharashtra) (Ramdasi, 2008): In this software, the available data from census of India 2001, regarding the Household Amenities in Maharashtra State have been analyzed to identify possible data marts and the dimensions that can be associated with these data marts. To find out the associated dimensions with the data marts and conformed dimensions, the top-down planning approach called as Data Warehouse Bus Architecture Matrix was used. With the help of this matrix the dimensional models have been designed and subject wise data marts are created.

Design and Development of Data Mart for Consumption Expenditure Survey Data (Debasis, 2010): Consumption Expenditure Survey Data were collected from all over the country by NSSO to fulfil the requirement of data on consumption expenditure (for planners, researchers, development agencies and Government of India) which is required for further studies and evolving realistic strategies for improvement of the standard of living of the people of this country. This collected data have been analyzed to identify possible data marts and the dimensions that can be associated with these data marts. To find out the associated dimensions with the data marts and conformed dimensions, the top-down planning approach called as Data Warehouse Bus Architecture Matrix was used and it was found that a single mart is sufficient enough in order to integrate all the information into a single place. The dimensional models have also been designed and the data mart for consumption expenditure survey data was created.

Online Systems for Water Technology

In the area of water technologies, multiple decision support systems have been developed in collaboration with Water Tecnology Center, IARI

Web Based Decision Support System For Watershed Management (Mukesh, 2007): Web based decision support system for watershed management is web-enabled information system to provide information to farmers, NGO's, teaching community on watershed management. CIS (computer based Information System) has great importance in agriculture an experts are not always available to answer farmers query. In this regard, this DSS is very meaningful to provide the alternate decision scenarios for watershed management .It has five modules for making decision on Soil and Water Conservation Measure, Land Based Activities, Solution to Specific watershed Problems, Crop Selection and Potential Employment Information. The design consideration of Engineering (Structural) and Vegetative (Agronomical) measure are also considered in the DSS. This software has one level of authentication i.e. Administrator. Administrator has the privilege to "Add", "Modify" or "Delete" the information or records from database. Other features such as "Ask Us", "FAQ", "Contact Us", " Useful Links", are available as modules of the of the DSS which makes this software more flexible and interactive. Users can also ask questions regarding the software to the concerned experts by sending an e-mail and can view some important questions in FAQ (frequently Asked Questions). This DSSWM has been developed using recent technologies viz. ASP.NET, SQL Server 2000, HTML, XHTML and Java Script. The developed DSSWM has been successfully validated for the different watersheds such as Datia, Kheda, and Saliyur and other regions in India. Nonetheless, the developed DSS is flexible enough to incorporate activities of other region in India.

Development of Decision Support System For Surface Runoff Estimation (Amreshsing, 2009): Computation of surface runoff is difficult as it depends upon several factors concerned with atmospheric and watershed characteristics. To facilitate this, a Decision Support System for Surface Runoff Estimation (SURE) has been developed which is a Web based software. SURE provides estimate of surface runoff based on Curve Number, Rational, Infiltration, Cooks and Empirical methods. The intent of SURE is to provide an easy-to-use hydrological

program for the novice uses. SURE provides amount of available water through rainfall that help in the design of control structures required to reduce soil erosion. SURE also maintains information about different land use patterns, different treatment patterns, soil type, different zones and stations etc. This will help user for location specific estimation of surface runoff. It will generate reports for zones and stations, curve number and rational method according to user query. The overall design of the system can be regarded as three-layered architecture consisting of Client Side Interface Layer, Server Side Application Layer and Database Layer. There is provision to insert and update the information. On-line help is provided for both administrator and user. SURE will be implemented as a network-based system so that information is available on-line.

Development of Decision Support System for Sprinkler Irrigation System (Mali 2009): The use of sprinkler irrigation is rapidly increasing around the world, and it is expected to be a viable irrigation method for agricultural production in the foreseeable future. It provides many unique agronomic, water and energy conservation benefits that address many of the challenges facing irrigated agriculture. The system needs to be appropriately designed for realizing its full benefits. But the quantitative nature of various factors related to design of sprinkler irrigation is very cumbersome and time consuming. Decision Support System (DSS) may provide for interaction that could be of immense help in solving the typical problems related to design of sprinkler irrigation system. During the course of operation of the DSS, expert advices in the form of default values and help menu provide sound basis for taking appropriate decisions. DSS for Sprinkler Irrigation System Design (SISD) is a Web-based Decision Support System to assist the user in designing sprinkler irrigation system. SISD has online report generation module to provide the layout of area to be irrigated per day, specifications of sprinkler irrigation system etc. even in the printable formats. SISD is developed using ASP.NET.

Development of a Web Enable System for Estimation of Canal Water Availability in a Selected Area (Niranjan, 2015): The Indian farming community is facing a lot of problem in increasing the crop production. Water is one of the major factor that influences the crop cultivation or crop production. Conservation of water is becoming increasingly important. Farmers relies on irrigation and canal irrigation is important component of the same (Rao and Rajput, 2009). Canal irrigation system consists of network of canals which include main canal, distributary canal, minor canal and outlets. The source of water to main canal could be river water or water stand reservoir. From the source water is released to main canal then it goes to distributary canal after that it goes to minor canal then to the field through outlet. Not all the water released in the main canal at river water reaches to the farmers field. A considerable portion of water released in main canal is lost in the form of seepage when it is flowing through the different canal such as main, distributary, minor and outlets. From irrigation point of view, the water that is available at field level is important. Normally the water available at the field level is estimated manually using the various methods to accomplish seepage losses. An online software has been developed to account seepage losses for estimating the water available at the field level. This system has been developed using standard three tier web architecture. The client side interface layer has been implemented using HTML, CSS and JavaScript. The server side application layer has been implemented using JSP technology and the database layer has been implemented using MS SQL Server. This software provides information about the seepage loss for the main canal, distributary canal, minor and outlets. The System also provides how much water available at the field level and the time taken to irrigate the field. This system can serve the Dam manager by providing them information about the seepage loss calculation, water available at field level and time taken to irrigate field.

Web based Information System on Groundwater Resources (Shabana, 2016): Groundwater is a promising but limited resource. According to official reports, our country is facing acute problem regarding over-exploitation of groundwater in Punjab, Haryana and western part of UP whereas in Eastern part of the country underutilization of groundwater is recognized. Information related to groundwater resources are available from different sources and in a different format. Mostly, it is in the form of published reports either in pdf format or printed media. A web-based system of groundwater resources is built to solve the problem of data which is available in heterogeneous format and observation well wise. Observation well are made at different places to record the

groundwater data. The application has been developed using NetBeans IDE with Java programming language. Client side interface is implemented using HTML, CSS and JavaScript. Server side has been implemented using JSP for providing platform independence and dynamic nature to the software. SQL Server 2012 has been used for creating the database. Application will be hosted on Apache Tomcat web server. Some features of the developed software called GIS (Groundwater Information System) includes good security by means of authenticated login, two categories of users (admin and client), comprehensive reporting facilities in the form of district wise, cluster wise, basin wise based on search criteria of year and state. Database maintenance activities like update, delete and insertion can be done by authorized admin. The software has been validated using ground water resources data from Odisha state. In future, software can be upgraded by developing mobile based application and or integrating software with modules for developing clusters.

Expert Systems and Knowledge Management

With the outburst of knowledge it has become necessary to convert unstructured knowledge into structured one. Traditional Knowledge Organization Systems must be reengineered so that they contain domain concepts linked through a rich network of well-defined relationships and a rich set of terms identifying these concepts in order to convert unstructured knowledge to structured one. These systems must also be converted to machine-processable formats based on Web technologies like XML, which tag the vocabularies in a standardized way. Ontology is the latest way of knowledge representation, in any domain as it defines concepts and relationships between them and acts in synergy with agents and Semantic Web Architecture. Building ontologies in different domains of agriculture will be help to convert unstructured knowledge into structured one that can be shared across applications. Following software are developed for building ontologies in the agriculture domain and for developing expert systems by utilizing knowledge stored in the ontologies:

Building and Querying Soil Ontology for Agriculture (Manoranjan, 2010): There are many soil classification systems but USDA Soil Taxonomy is most accepted worldwide. Soil ontology has been designed and created for USDA soil taxonomy through Protégé OWL editor from Order to Sub group level. Using this soil ontology, a web based application, Soil Taxonomy Ontology has been developed. This application has N-tier architecture, application development environment as NetBeans 6.9 editor, Protégé, Web development technology as Java Server Pages (JSP) and SPARQL. Semantic Web Framework layer is implemented using JENA. The search facility provides USDA Soil Taxonomy in details up to Subgroup level of seven orders available in India. Domain experts can see and edit the Soil Ontology or can suggest anything related to the creation of Soil Taxonomy Ontology through Web Protégé. By using Advance Search navigation key one can easily classify a newly found soil up to Subgroup level. This software also facilitates name based search for all soil taxonomic terms.

Building Soil Ontology up to Soil Series (Chandan, 2012): Web based software which use ontology as their knowledge base are gaining importance as they act in synergy with agents and Semantic Web Architecture. Ontologies define domain concepts and the relationships between them, and thus provide a domain language that is meaningful to both humans and machines. For the web, ontology is about the exact description of web information and relationships between web information. Taxonomies describe real world concepts in well-defined hierarchy and exist in standardized form for numerous domains of knowledge. It is imperative that ontologies are built in different domains of agriculture that help to convert unstructured knowledge into structured one which can be shared across applications. Soil Ontology developed for USDA soil taxonomy by Das (2010) and Das et al. (2012) for Soil Orders available in India only up to Sub group level has been used to develop a web based application that now covers all the twelve orders worldwide. The developed soil ontology now is available up to Family and Series level. The developed web based application is having N-tier architecture, developed using NetBeans 6.9 editor, Protégé, Java Server Pages (JSP) and SPARQL. Semantic Web Framework layer is implemented using JENA. Information related to soil taxonomy and newly found soils can be easily classified according to USDA soil taxonomy by mentioning their properties. Domain experts can edit or add any new

information about the soil taxonomy. By using Advance Search navigation key one can easily classify a newly found soil up to series level. This software also facilitates name based search for all soil taxonomic terms. By using the series navigation key one can easily get the details information of state wise series description. Other applications can use its knowledge base as it is in the form of Ontology.

Building and Querying Ontology for Agriculturally Important Microbes (Sreekumar, 2012): Traditional knowledge representation techniques generate unstructured knowledge. The need of converting the unstructured knowledge into structured knowledge is highly felt. Ontologies are the new form of knowledge representation technique that acts in synergy with agents and Semantic Web architecture. Building ontologies in different domains of agriculture will be helpful to convert unstructured knowledge into structured knowledge, which can be shared across different applications. The microorganisms represent a crucial role in the world of agriculture. In this work, an attempt has been made to develop a web based software for the microorganisms, important from agricultural point of view. There are many classification systems for microbes, but the Three Domain System of Microbial Taxonomy is most accepted worldwide. Microbial ontology has been designed and created for Three Domain System of Microbial Taxonomy through Protégé 3.4.6 OWL editor from Domain to Genus level for those bacteria which are important in agriculture. Using this microbial ontology, a web based application, Microbial Taxonomy Ontology has been developed. This application has N-tier architecture, application development environment as NetBeans IDE 7.0.1, Protégé 3.4.6, Web development technology as Java Server Pages (JSP) and SPARQL. Semantic Web Framework layer is implemented using JENA. The search facility provides Three Domain System of microbial taxonomy for agriculturally important bacteria in details up to Genus level of the twenty domains. Domain experts can see and edit the knowledge base (i.e. Microbial Ontology) or can suggest anything related to the creation of Microbial Taxonomy Ontology through a user friendly web interface. By using Advance Search navigation key one can easily classify an unknown microbe up to Genus level. This software also facilitates name based search for all microbial taxonomic terms.

Ontologies based Expert System for Maize (Arijit, 2011): Since time immemorial Indian farmers were growing land races/local cultivars having low productivity. Maize (Zea mays L) is the most versatile crop with wider adaptability in varied agro-ecologies. It has highest genetic yield potential among the food grain crops. In India, maize is the 3rd most important food crops after rice and wheat. Most of the farmers don't know which variety is appropriate for a particular season and also for a particular area. They generally use the same variety for over a long period of time. Beside this pests and diseases are the major causes for the damages in the crops and results in the economic loss to the farmers. The estimate loss per annum due to diseases and pests in a country like India touches to billions of Rupees. Most of the time farmers use over dosages of pesticides and fungicides to save their crop and thus cause environmental hazards. This Ontologies based Expert System for Maize is designed to help farmers to take appropriate decisions and disseminate need based research findings to millions of the farmers at a time, which is neither feasible nor practical by conventional system of extension. In conventional architecture of expert system, knowledge engineers along with domain experts build the knowledgebase manually and these expert systems are known to be working within a narrow domain of knowledge. But for building the expert system in agriculture for a vast and diverse country like India, the conventional approaches fail to meet the need of the farmers. Ontology is the latest knowledge representation technique that allows the domain experts to code their knowledge in a specific domain. Ontology is devised for the Web based systems and provides them with the semantics of the concepts in the specific knowledge domain. They have the potential to be used in a distributed environment like Internet and provide the dynamic and reusable capability to the knowledgebase. The system currently has about 80 maize diseases, 52 insects and 39 varieties of maize. The system works in question-answer mode and allows the farmers to choose options for each of the question asked. At each level the text is supported by pictures. The presented system has a dynamic knowledgebase and acts as a tool for transferring the site and crop specific knowledge of various domain experts to the farmers.

Expert System on Solanaceous Crops under Protected Cultivation (Kamalika, 2013): Protected Cultivation technology is a new technology in agricultural sciences for our country. Most of the farmers don't know about

this new technology of crop cultivation. Solanaceous crops particularly tomato and capsicum are important vegetable crops in India grown under protected condition. Availability of experts on protected cultivation for solanaceous crops is rare. So, there is a need for developing an online expert system to disseminate need based knowledge on solanaceous crops under protected cultivation to the farmers to help them to increase the efficiency for crop management and to increase the yield. Expert systems are identified as a powerful tool with extensive potential in agriculture. It combines the experimental and experiential knowledge with the intuitive reasoning skills of a multitude of specialists to aid farmers in making the best decisions for their crops. Expert System on Solanaceous Crops under Protected Cultivation is designed to help farmers to take appropriate decisions and disseminate need based research findings to millions of the farmers at a time. The presented system uses Ontology as its knowledgebase and provides the user with the result based on the recent knowledge. This framework does not pose any restriction on the domain experts for regular updating and modification of the underlying ontology. The system has been developed with robust JAVA technology and uses MS SQL Server 2008 as database and uses the inferencing capabilities of JENA, a semantic web tool kit. The system currently has information about 17 diseases, 6 insects and 4 types of protective structures of Tomato and Capsicum crops. The system works in question-answer mode and allows the farmers to choose options for each of the question asked. At each level the text is supported by pictures. The presented system has a dynamic knowledgebase and acts as a tool for transferring the site and crop specific knowledge of various domain experts to the farmers.

Expert System for Rapeseed-Mustard Crop (Sahi,2012): Rapeseed-mustard crop is grown in India in diverse agro climatic conditions. Most of the farmers do not know which variety is appropriate for a particular season and also for a particular area. They generally use the same variety for over a long period of time. Besides this, pests and diseases are the major causes for the damages in the crops and result in economic loss to the farmers. Use of over dosages of pesticides and fungicides by farmers to save their crop also causes environmental hazards. This Ontologies based Expert System for Rapeseed-Mustard is designed to disseminate need based research findings to the farmers at a time so that they appropriate decisions. In conventional architecture of expert system, knowledge engineers along with domain experts build the knowledgebase manually and these expert systems are known to be working within a narrow domain of knowledge. But for building the expert system in agriculture for a vast and diverse country like India, the conventional approaches fail to meet the need of the farmers. Ontology is the latest knowledge representation technique that allows the domain experts to code their knowledge in a specific domain. It has the potential to be used in a distributed environment like Internet and provide dynamic and reusable capability to the knowledgebase. The system has been developed with robust JAVA technology and uses MS SQL Server as database. The system currently knowledgebase of 10 diseases, 8 insects and 110 varieties of rapeseed-mustard. The system works in question-answer mode and allows the farmers to choose options for each of the question asked. At each level, the text is supported by pictures. The presented system has a dynamic knowledgebase and acts as a tool for transferring the site and crop specific knowledge of various domain experts to the farmers.

Recommender System for Crop Variety Selection (Sanchita, 2014): With the beginning of the Web 2.0 era, the internet began growing with tremendous speed and developing rapidly regarding both content and users. This has led to an information overload problem in which people are finding it increasingly difficult to locate the right information at the right time. Recommender Systems have come to rescue users from this problem, by guiding users through the big ocean of information. It uses the opinions of members of a community to help individuals in that community to identify the information most likely to be interesting to them or relevant to their needs. These systems use the similarity between the users' choices and the content of the items to form recommendation list for the user. Recommender systems can now be found in many modern applications that expose the user to a huge collection of items. Such systems typically provide the user with a list of recommended items they might prefer, or predict how much they might prefer each item. It also help users to decide on appropriate items, and ease the task of finding preferred items in the collection. But till now Recommender systems have not been exploited to that extent in agricultural domain. Therefore an innovative idea that enables to integrate the recommendation technique with the word-of-mouth information exchange between farmers

has been attempted to model in this thesis. Recommendation technique to suggest crop varieties to farmers is a bit different from the typical recommendation techniques as all the farmers have different socio-economic status which restricts their choices to adopt several newly released varieties directly for cultivation. This thesis presents Recommender System for Crop Variety Selection with a focus on Maize crop. A prototype system, has been developed as a proof of concept. The system creates a personal Farmer Agent for each registered farmer. The farmer agent stores the socio-economic profile of the farmer as well as his experiences of growing a crop variety. Java Agent Development Environment (JADE) has been used to develop these agents as well as to provide the message passing among these agents. A registered farmer can get the variety recommendation through its agent which in turn asks the recommendation from the peer farmer agents having the similar socio-economic profile. All the received recommendations are ranked and the top five are shown to the requesting farmer. The system has been built on modular approach and thus facilitates integration of other complex recommendation algorithms in future. Since, the system has been developed using JADE environment, it also allows to create agents on distributed servers. The cold start problem of recommendation systems has been tackled by registering the farmers selected for Front Line Demonstration for Maize crop as well as scientists who have conducted experimental trials on the released varieties. The presented system has a capability to transfer agricultural technologies using both top-down as well as through peer to peer interaction approaches and will help in faster adoption of new crop varieties/technologies.

Online Systems for Agriculture Economics

Keeping in pace with the development of the internet technology, there is need to develop web based data analysis tools in agriculture research also. Following software are developed in the agricultural economics:

Web based software development for computation of total factor productivity (Samimul, 2010): The rapid advancements in the internet technology front have expanded the potentiality for web based software packages. Web based software allows quick and convenient sharing of methodology among researchers. Productivity growth in agriculture has remained a serious concern for intense research over the last five decades. TFP is an important measure to quantify the productivity growth. Index of Total Factor Productivity (TFP) measures the growth of net output per unit of total factor input. Modules for TFP computation are not available in any statistical software and commonly used econometric packages. Therefore, a need for web based TFP computation software (WBSTFP) was felt and attempt was made to develop the user-friendly online software. The software provides TFP index, output index and input index using Tornqvist index method. Growth curve of each index is also computed and presented with tables and graphs. It is an online software that can be accessed using the default browser of the user's system. The software is completely menu driven and offers user-friendly screens to reduce efforts in understanding the software. Facilities for computing index for single crop and index for aggregate crop have been provided through two separate sub-modules. Facilities for computation of TFP by aggregating data of lower spatial units are also provided. User can register, login, compute TFP and see the results as well as can save result in Excel file. Administrator interface of the software helps in development and maintenance of user database. Software results are validated using a suitable dataset.

Development of Online Software for Computation of Malmquist Index (Sarita, 2013): Productivity growth in agriculture is both necessary and sufficient condition for its development and has remained a serious concern for intense research over the last five decades. Malmquist Index is an important measure to quantify the productivity growth. Malmquist Index is used to measure the total factor productivity change between two data points by calculating the ratio of distances of each data point relative to a common technology (Caves *et al.,* 1982). Modules for Malmquist Index computation are not available in any online software and commonly used econometric packages. After requirement analysis for Malmquist Index Computation software, it was observed that there is a need to develop easily accessible, user friendly and interactive software. Keeping these considerations in mind, in this thesis an attempt is made to design and develop Online Software for Computation of Malmquist Index (MalmSoft). MalmSoft has been designed and developed as per web based three-tier

architecture. Software is developed in Microsoft .NET environment. The User interface layer is implemented using combination of HTML, JavaScript and CSS. ASP.NET 4.0 and C#.NET is used for writing business logic. Database Layer is implemented for user management in MS Access. Being web based, MalmSoft is freely accessible software for Malmquist Index. Software is completely menu driven and offers user-friendly screens to reduce efforts in understanding the software. The software provides functionality for computation of distance function and Malmquist Index for an agricultural firm from one time period to another time period. User can register, login, compute Malmquist index and see the results as well as can save result in excel file. Software results are validated using a suitable dataset whose results have been compared with standard software DEAP. This software is will be useful for agricultural researchers engaged in research in agricultural economics and allied sciences.

Software for Agricultural Productivity Analysis (Surchand, 2015): The rapid advancements in the internet technology front have expanded the potentiality for web based software packages. Web based software allows quick and convenient sharing of methodology among researchers. Keeping in pace with the development of the internet technology, there is need to develop web based data analysis tools in agriculture research also. Productivity growth in agriculture has become a serious issue over the last five decades. Analysis of agricultural productivity can be one cause which can help to improve production in agriculture. It can analyse large agriculture data of different region and help to find factors affecting the growth of productivity. There is wide difference in the agricultural performance within state due to its variation in resource available, climatic, topography etc. so district level study of agricultural data will be a wise step to give a better result. Therefore a need for Software for Agricultural Productivity Analysis (SAPA) is felt. The software is completely menu driven and offers user-friendly screens to reduce efforts in understanding the software. User can register, login, analyse agricultural productivity and see the results as well as can save result in Excel file. Software results are validated using a suitable dataset.

Web based Application for Approtioning Temporal Data at District Level (Saravanakumar, 2015): The rapid advancements in the internet technology front have expanded the potentiality for web based software packages. Web based software allows analysis to be shared among researchers quickly and conveniently. Keeping in pace with the development of the internet technology, there is need to develop web based data analysis tools in agriculture research also. Agriculture performance generally differs widely within state due to varying regional characteristics in terms of resource endowments and climate. Apportioning is an important process to distribute or allocate with proportional obtain from geographical area over three decade of time period (Bhalla & Gurmail Singh, 2001). Methodology for apportioned data for district is not available in any format in any software or in documented form. Therefore, a need for web based application for apportioning temporal data at district level (DAS) was felt and attempt was made to develop the user friendly online software. User can register, login, Apportion district data, and can see the results and save data in excel file for future use. Administrator interface of the software helps in development and maintenance of user database. Software results are validated using real data.

Software for Data Mining

Data Mining is the emerging area in which multiple software have been developed to analyze and find novel pattern hidden in the agriculture datasets

Web Based Fuzzy C-means Clustering Software (Maedeh, 2011): Clustering is an explorative data mining task, which deals with segmenting the data objects into number of clusters such that data items with in a cluster are highly similar whereas data items belonging to different clusters are highly dissimilar. In real life applications there is very often no sharp boundary between clusters. For those cases fuzzy clustering has important role to play. Fuzzy clustering method allows objects to belong to several clusters simultaneously with different degrees of membership. Fuzzy c-means is the well-known fuzzy clustering

algorithm in literature, presented by Dunn and further developed by J.C. Bezdek to carry out fuzzy clustering. Fuzzy clustering has found potential applications in many sector of science including agriculture too. After requirement analysis for fuzzy c-means clustering software, it was observed that there is a need to develop easily accessible, user friendly and interactive software. Keeping these considerations in mind, in this thesis an attempt is made to design and develop web based fuzzy c-means clustering software (wFCM). wFCM has been designed and developed as per web based three-tier architecture. Software is developed in Microsoft .NET environment. The User interface layer is implemented using combination of HTML, jQuery, JavaScript and CSS. ASP.NET 3.5 and C#.NET is used for writing business logic. Database Layer is implemented for user management in SQL Server 2008. Being web based, wFCM is freely accessible software for fuzzy clustering. Software is completely menu driven and presents user-friendly GUI which is developed to minimize efforts in understanding the software. User can upload data to wFCM using three different formats; Excel, CSV and image files. Fuzzy clustering results can be downloaded by the user in excel and PDF formats. User has been given the option to view the clustering results graphically. Result can be downloaded by the user in excel and PDF format. Software results are validated using suitable dataset from machine learning repository. This software will be useful for statisticians, researchers, students and teachers for clustering datasets from agricultural research as well as many diverse areas of other sciences.

Online Rule Generation Software using Decision Tree Classifier (Satma, 2011): The rapid advancements in the internet technology front have expanded the potentiality for web based software packages. Web based software allows analysis to be shared among researchers quickly and conveniently. Keeping in pace with the development of the internet technology, there is need to develop web based data analysis tools in agriculture research also.The handling of enormous amounts of data produced in agricultural research for taking appropriate and logical decisions is of major concern now. E-agriculture is a significantly emerging field focusing on agricultural development through improved information services. It involves the development of production systems like expert systems on various domains like crop cultivation, fertilizer application, agricultural extension etc., which can guide the farmers, agricultural researchers, teachers and students to get an understanding of those domains. For developing these systems, usually experts are generating the input rules manually which is a time consuming process. Therefore, a need for web based rule generation software (GenRule) was felt and attempt was made to develop the user-friendly online software. The software generates the decision rules using the ID3 decision tree classifier. Visualization of the rules is also provided in the form of decision tree. The generated rules are accompanied by various evaluation measures for their validity. GenRule provides the facility to classify future data instances. It is an online software that can be accessed using the default browser of the user's system. The software is completely menu driven and offers user-friendly screens to reduce efforts in understanding the software. In addition, online help is also provided. User can register, login, generate the rules, and can see the results and save in excel, text and XML file for future use.

Development of Software for Back Propagation Neural network with Weight Decay Algorithm (Rakesh, 2012): The rapid advancements in the internet technology front have expanded the potentiality for web based software packages which allow quick and convenient sharing of methodology among researchers. Artificial Neural Networks (ANNs) are non-linear structures used for prediction and classification problems. ANNs can identify and learn correlated patterns between input data sets and corresponding target values. Trained ANN can be used to predict the outcomes of independent variables. Over fitting and under fitting are two major problems that may arise in ANNs. Multi-collinearity is a statistical phenomenon in which two or more predictor variables in a model are highly correlated and provide redundant information about the response. The problem of multi-collinearity leads to overtraining. This problem is handled by using artificial neural networks with weight decay algorithm. Most of the software available for analyzing the data using ANN is either very costly or difficult to use. In this study, a web based software for back propagation neural networks with weight decay algorithm has been developed. Waterfall model has been used for software development process. This software is useful for statistician and researchers working in the area of agriculture.

Web based software for feature selection using rough theory (Rajeshwar, 2013): Rough set theory was proposed by Zdzislaw Pawlak in the early 1982 is now in a state of constant development. It deals with the classificatory analysis of data tables. The main goal of the rough set analysis is to synthesize approximation of concepts from the acquired data. Its methodology is concerned with the classification and analysis of imprecise uncertain or incomplete information. It is considered as one of the first non-statistical approach in data analysis. WEBSFS-RST software is developed on the principle of rough set concept. Rough sets are applied in many domains, such as, for instance in machine learning, knowledge discovery, artificial intelligence, medicine, data mining, expert systems, etc. It helps to find minimal sets of data with the same knowledge as in the original data called the reduct. WBSFS-RST is an online software which is based on the concept of rough set theory so it can perform various operation related to the properties of rough set. The main task is to generate reduct of an information system. Finding reduct is a NP hard problem. WBSFS-RST can generate optimal reduct for an average size dataset containing up to 100 ROWS. No online software has been found to compute reduct so far. WBSFS-RST will be online accessible to user at any time. There is no need of formal training or any programming expertise for using WBSFS-RST. The software has proper security authentication. The system has been tested with different datasets and the results matched with the calculated reducts in the literature.

Online Decision Tree Classification using C4.5 Algorithm (Suvajit, 2013): Data mining is a step of knowledge discovery in databases, performing several tasks such as classification, regression, clustering, association rule mining, outlier detection, etc. Classification is one of the most important and widely carried out tasks of data mining. It is a predictive modelling task which is defined as building a model for the target variable as a function of the explanatory variables. There are many well established techniques for classification out of which, decision tree is a very important and popular technique from the machine learning domain. Decision tree is a decision support tool that uses a tree-like graph or model of decisions and their possible consequences, including chance event outcomes, resource costs and utility. C4.5 is a well known decision tree algorithm used for classifying datasets. The C4.5 algorithm is Quinlan's extension of his own ID3 algorithm for decision tree classification. It induces decision trees and generates rules from datasets, which could contain categorical and/or numerical attributes. The rules could be used to predict categorical values of attributes from new records. C4.5 performs well in classifying the dataset as well as generating useful rules. In this study a web based software for rule generation and decision tree induction using C4.5 algorithm has been developed. The visualization in the form of tree structure enhances the understanding of the generated rules. The software contains the feature to impute the missing values. The enhanced waterfall model has been used for the software development process. This software is useful for academicians, researchers and students working in the area of data mining, agriculture and other fields where huge amount of data is generated.

Software for Image Analysis

Image Analysis is one of the promising area in which large number of applications can be developed for analysing plant phenomenons and for devising new non destructive techniques as compared to the existing lab based time consuming destructive techniques. Students have started working in this area and initial results have started coming

Non-destructive phenotyping of rice plant through image analysis (Tanuj, 2013): Leaf area and chlorophyll content are considered as most important agronomic parameters for studying physiological features related to plant growth, photosynthetic and transpiration process etc. Manual chlorophyll extraction procedure and leaf area measurement with leaf area meter are accurate but are destructive, laborious, time consuming and relatively expensive. High-throughput image analysis for automated phenotyping is used to extract phenotypic parameters related to chlorophyll content and leaf-area of the plant which saves time and effort. With the increase in information availability, there is an immense need for creation of information system for storage, browsing and sharing of the online data generated by phenomics facility and offline data collected by

experimenters and experimental metadata. No freely open source software for high-throughput image analysis is available. There exists a clear need to develop an image analysis tool for phenotyping of plant. Rice crop is selected for phenomics in this work because of its importance as a major food crop. The web based software "NPRIA" for estimation of non-destructive phenotypic parameters for chlorophyll content and leaf area of rice plant is based on the N-tier architecture, developed using NetBeans 7.0.11 IDE, Java Server Pages (JSP), ImageJ API, and MS SQL Server 2008. The two proposed models namely Artificial Neural Networks (ANNs) feed forward back propagation and linear regression model are used to predict chlorophyll content (with RMSE 0.001) and leaf area (with correlation coefficient 0.56) of the rice plant respectively based on image analysis.

Geographical Information Systems Related Software

GIS assisted farm management information System (Kaushik, 2007): GFMIS is a web based user-friendly, integrated solution for the farm management activities, developed in Java Server Pages (JSP) and ArcView software. It is developed as comprehensive farm management software for Indian Agricultural Research Institute (IARI) research farm. Geographic Information System (GIS) is the powerful tool that provides exact geographic information and visualization of any feature. Thus GIS assisted Farm Management Information System helps the farm manager in better and precise decision making for scientific management of the farm. GFMIS provides map search facilities in which one can visualize the location of various features such as the distribution of Irrigation, Road network, location of Residential area and divisions, distribution of soil layers/ series etc. Also provides search facility for plot, crop, variety, soil type, fertilizer, infestation, water source, and year wise information. The software also provides keyword wise searching facility. Users can also view customized reports on various aspects of farm. User can interact with subject specialists through email. On-line help is provided for both administrator and user. The feature of providing information to users through frequently asked questions has been included in this software. Information on various activities being undertaken in various divisions of IARI is also available in GFMIS which contain the General map and Plot map. It has a three-layered architecture. Client Side Interface Layer is implemented in HTML and JavaScript. Server Side Application Layer is implemented in Java Server Pages, ArcView for generation of thematic maps and Java Database Connectivity. Database Layer is implemented in Microsoft Access 2000. GFMIS can be implemented as a network-based system with a server at IASRI so that information is available on-line. GFMIS runs at any node of the Internet through a browser. Security features are provided in such a way that only concerned person can access the database.

Web-based Tool for Interpolation of Climate Data (Ipshita, 2015): In agriculture and environmental planning, climate plays a very important role; it is measure to learn the relationship between biological and environmental phenomenon. Developing countries like India are facing the problem of lack of accurate climate data. This problem affects integrated planning on the environment and agriculture. Agricultural and natural resource management activities require the understanding spatial variation in climatic conditions. Estimates of climatic variables which are spatially distributed are required for use in geographical information systems and models. This means that the effectiveness of agricultural research depends on the techniques which can handle variability in crop and soil, weather parameters, and spatial interpolation, that represents estimation of value of data points at the sites from where no sample is drawn and it is in an area which is already known with sampled points, by using the data obtained from the sampled points. There is necessity of accurate and inexpensive quantitative approaches to interpolate climate data in developing countries. Climate data in India is acquired from the weather stations located at different locations in each state. In India, when required climate data for the locations where no weather stations are located, the average (mean) of the climate data from the surroundings locations is considered. Each state doesn't have enough weather stations to even cover each district. (e.g Uttar Pradesh (UP) has 75 districts and only 47 weather station. Source: IMD). Interpolated climate data is a necessity for better planning, forecasting and research related to global climate change scenario. Around

the world many software have been developed and are being used for interpolating climate data but either they are all stand alone or have been developed for specific locations. Thus a web based tool for interpolation of climate data is needed which can help researchers, students and ecological workers to have an access to the data at unsampled locations for their respective works. The present study was, therefore taken upon to design and develop a Web-based Tool to interpolate the climate data.

Mobile Applications

In last few years, tremendous improvement has been seen in the field of mobile technology. This enhancement has made the smartphones and mobile apps, an integral part of human life. These days, the smartphones and mobile apps have made remarkable changes in all sectors of the Indian economy. The mobile apps are being used everywhere from highly developed cosmopolitan cities to the rural villages. Following software have been developed as mobile apps:

Mobile App for Information Retrieval on Pest and Disease in Crops (Ashraful, 2016): In this study, an attempt has been made to develop a mobile application named 'AGRIdaksh' for the India farmers. This mobile application is aimed to provide timely and valid information to the farmers about the problems related to the crop protection such as diseases, insect-pests, nematodes, weeds and physiological disorder. This app can also help the farmers in selecting proper insect-pests resistant and disease resistant varieties of different crops. The farmers are able to send their queries along with images and videos related to crop protection to experts and the agricultural scientists. This is a two-step process. In the first step, farmers' queries are stored in the database according to the selected domain such as diseases/pest/varieties. In the second step, experts can view the query along with images/videos and then can suggest possible solution of the problems. In this thesis, the 'AGRIdaksh' app has been developed on the android platform with minimum SDK version of API 15: Android 4.03 (Ice Cream Sandwich). The application is based on the n-tier architecture of the software development. The client-side interface is the android application itself which is implemented by the JAVA programming language and XML. The next layer is a web service layer that act as an intermediate interface between the android application and the database in the server. The web service has been developed in JAVA programming language. The server side layer contains core classes like database connection, the connection between other layers. The database layer contains the data of the maize crop related to the crop protection, varieties, farmer queries and user details. Several APIs such as kSOAP2 API, JDBC API has been used for interconnection among various layers. The application has been tested for the maize crop and provided the intended results as specified. The developed android application 'AGRIdaksh' would be of great use to the farmers to deal with crop losses due to the biotic agents and making farming a profitable venture.

Mobile Application for Plant Quarantine Regulations to Import in India (Parvez, 2016): India is a very large country where 69% of total population depends on Agriculture. Agriculture has a great role in Indian economy because it contributes significantly about 14.8% in 2013-14 to the Gross Domestic Product. The percentage share of import and export of plant commodities have been of the order 3.22% and 13.79% of national import and export in 2013-14. There has always been the possibility of moving exotic insect pests, diseases and weeds from other country to India. Exotic pest and diseases introduced into India cause a huge damage to Indian agricultural trade. For these reasons to prevent the introduction of various exotic pests, diseases and weeds from other countries or within states of country, legal restrictions are enforced which is commonly known as Plant Quarantine. Plant Quarantine regulations at national level are known as Domestic Quarantine as well as at international level known as Foreign Quarantine. The implementation of the quarantine measures are assisted by legal approval, called quarantine laws. It acts as an important technique or procedure in to exclude exotic pests from the crop. Efficient implementation of quarantine is extremely emphasized to manage pests, consequently which helps in sustaining the productivity of crops. Now mobile phones are now very much preferable to the common men. Information can easily be shared through the mobile application. At present

Plant Quarantine import regulations are presented in pdf form hence it is a tedious process for the user to find their specific requirement. It is very hard to know for an importer at any situation about this regulations whether any plant commodity is prohibited or allowed to import in India. Thus the need arises to provide the information in searchable manner. Import regulations are enlisted in four schedules with justification, condition, additional declaration on the basis of Commodity, Country of origin of the commodity, Category of the Commodity, Plant Part of the Commodity. This thesis presents the requirement analysis, design, development and testing of an android based mobile application with different features which provides information about import regulations related to Commodity, Country Of Origin, Category, Plant Part which is promulgated by the Directorate of Plant Protection, Quarantine & Storage, Department of Agriculture, Cooperation & Farmer Welfare, Ministry of Agriculture and Farmer Welfare, Government of India to prevent the entry and spread of dangerous pests and pathogens.

Mobile Application for Chlorophyll Content Estimation in Rice (Sonica, 2016): Agriculture plays a vital role in India's economy. As India's economy is dependent on agriculture therefore focus is on increasing the productivity. The abiotic environmental factors affect the crop growth and yield. Chlorophyll content is an important parameter which acts as an indicator of plant growth, deficiency of major nutrients and plant health. Nitrogen (N) is a key element found in chlorophyll molecules. Chlorophyll content is almost proportional to N content of the leaf. Rice is one of the main food crops of India thus ICAR-Indian Agriculture Research Institute (IARI) is conducting different experiments on rice varieties to study the affect of abiotic factors in different conditions. In order to effectively store, analyze and share different experimental data related to phenomics parameters, a system with the name mPhenoDB has been developed at ICAR-Indian Agricultural Statistics Research Institute (IASRI). mPhenoDB system can store data in different formats (image, records, files). This web based system has the provision for storage and analysis of image data for leaf area estimation and chlorophyll content estimation. With the realization of usage of smart phones the idea of development of mobile application was taken up in this study to facilitate non-destructive method based on image analysis for chlorophyll estimation. The mobile based application for estimation of chlorophyll content has been developed using N-tier architecture: Client Side interface Layer (CSIL), Web Service Layer (WSL), Server Side Application Layer (SSAL) and Database Layer (DL). Mobile application has been developed for Android platform. Application is user friendly and users can easily get the quick reference of chlorophyll estimation with click of a button. The application interface provides information and action for capturing and cropping pictures for users. Correspondingly RGB count and Chlorophyll content is estimated. This application is feasible to run on any device with Android platform. Application design is generic which can be extended for other formulae and different crops.

Conclusion

The discipline of computer application have played a key role in development of software in various themes. These researches have resulted in many institute and interinstitutional projects. These systems have been widely used by researchers across the country and as a result the Division of Computer Applications, ICAR-IASRI has become the driving force in inculcating the mindset required for utilizing the benefits from the Information Technology in agriculture sector. Some of the key areas in which the division will focus in near future is use of artificial intelligence, data mining, image analysis, GIS and integration of these technologies through web and mobile technologies to solve the issues related to agriculture or to support the scientific community in carrying out the research more efficiently.

Acknowledgments

I wish to thank all the Faculty Members of the Division of Computer Application for extending support and cooperation in preparing the research contributions of M.Sc. students.

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