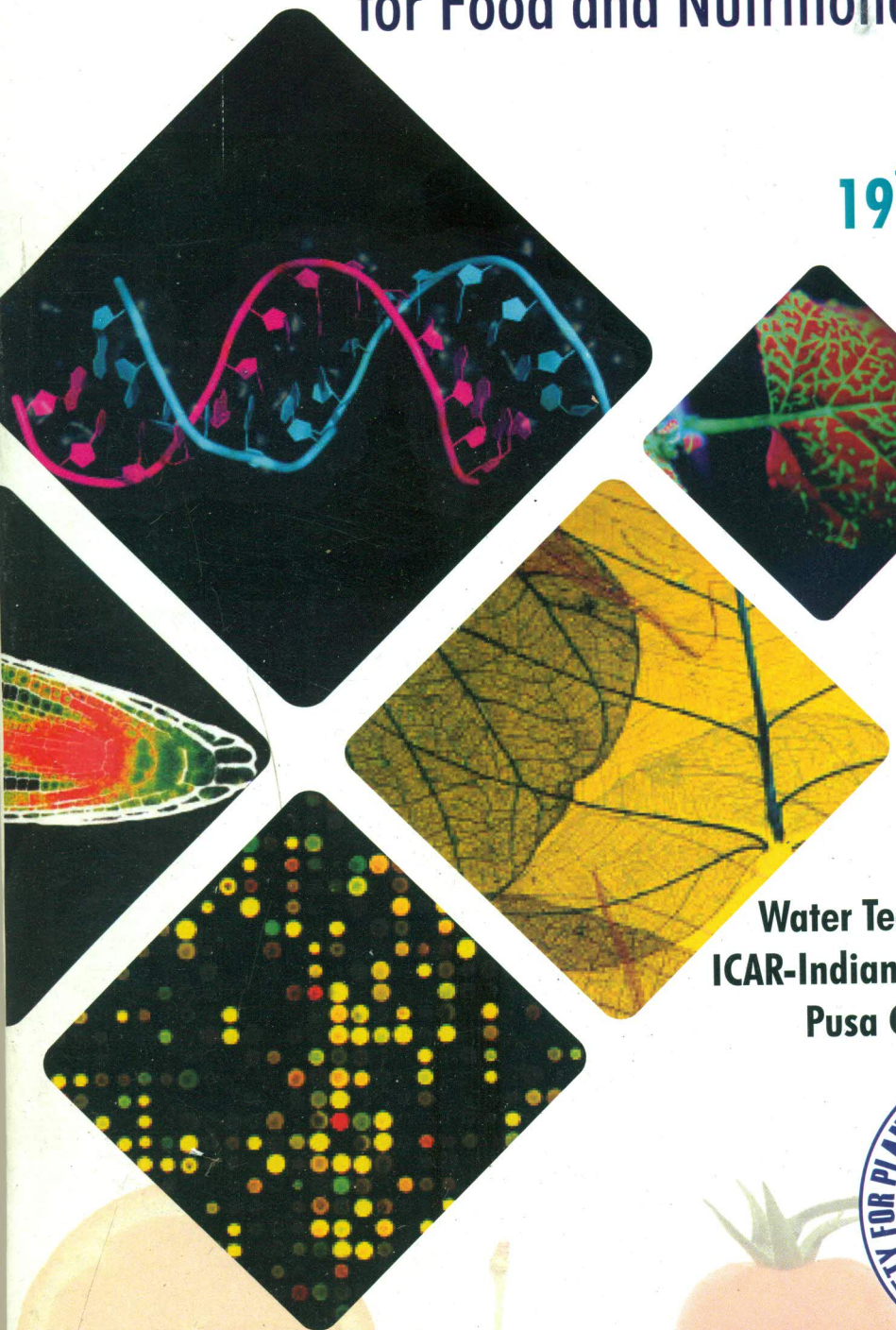
National Symposium on

Connecting Innovations in Plant Biology for Food and Nutritional Security



19th November, 2018

Abstracts & Souvenir

Venue

Water Technology Centre Auditorium
ICAR-Indian Agricultural Research Institute
Pusa Campus, New Delhi, India

Organised by
Society for Plant Biochemistry and Biotechnology
Division of Biochemistry, ICAR-IARI
Pusa Campus, New Delhi

Abstracts & Souvenir

National Symposium

on

Connecting Innovations in Plant Biology for Food and Nutritional Security

November 19th, 2018

Society for Plant Biochemistry and Biotechnology

Division of Biochemistry

ICAR-Indian Agricultural Research Institute

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Compiled and Edited by:

Anil Dahuja

P.K. Jain

Rekha Kansal

Monika Dalal

Veda Krishnan

Naveen Chandra Gupta

Vinutha T

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TECHNICAL PROGRAMME

Time	SESSION-I
10:00-11:30	**N.B. Das Memorial Lecture** **Chairperson:** Prof. V. L. Chopra Former Member, Planning Commission of India & Former-Secretary (DARE) & Director General (ICAR) **Co-chairperson:** Dr. M.L. Lodha Former ICAR-Emeritus Scientist & Ex-Head, Division of Biochemistry, ICAR-IARI, New Delhi **Speaker :** Prof. H.K. Das, Former Dean, JNU, New Delhi **Title : *"A tribute to late Dr. N. B. Das by a former colleague"***
11:30-12:00	**High Tea**
	SESSION-II
12:00-13:30	**Panel Discussion on "Branching Careers in Biochemistry/Biotechnology: Time for Introspection"** **Co-chairs:** Dr. T.R. Sharma, Executive Director, NABI, Mohali (Chandigarh) Dr. R.K. Jain, Dean & JD (Edn.), ICAR-IARI, New Delhi **Convener:** Dr. Ramcharan Bhattacharya, Professor, ICAR-NRCPB, New Delhi **Rapporteur:** Dr. Ranjeet Ranjan Kumar, Senior Scientist, ICAR-IARI, N. Delhi
	Panelists: 1. Dr. Venkateshwarlu Gudipati, ADG (EQ A&R), ICAR 2. Dr. Purnima Sharma, Managing Director, Biotech Consortium India Ltd. (BCIL) 3. Dr. M.V. Rajam, Professor, Department of Genetics., Delhi Univ. 4. Dr. S.K. Khare, Assoc. Dean R&D and Professor of Biochemistry, IIT, Delhi 5. Dr. Deepak Prem, Seed Regulatory Lead, Monsanto, India 6. Dr. Veda Krishnan, Scientist, Division of Biochemistry, ICAR-IARI, New Delhi
13:30-14:30	**Lunch Break**
14:30–15:15	**Annual General Body Meeting**

14:30-16:00 **(15:15-16:00)**	**Poster Session** **(Poster Evaluation)**
	SESSION-III
16:00-17:30	**Lead Lectures**
	Co-chairs : Prof. K.C. Bansal, Area Convener, Nano Biotechnology Centre, (TERI) & Former Director, NBPGR, N. Delhi Prof. Rajat Sandhir, Professor, Department of Biochemistry, Panjab University, Chandigarh **Convener:** Dr. Aruna Tyagi, Professor, ICAR-IARI, New Delhi **Rapporteur:** Dr. Archana Singh, Principal Scientist, ICAR-IARI, New Delhi
	Theme Area 1: Abiotic and Biotic Stress Management
	Dr. C. Viswanathan, ICAR-IARI, New Delhi **Topic:** ABA Receptors: Progress and Prospects for Enhancing Biotic and Abiotic Stress Tolerance of Crops
	Theme Area 2: Nutritional Quality Enhancement
	Dr. Sudesh Kumar Yadav, CIAB, Mohali **Topic**: Biotechnology and bioprocessing for nutritional quality improvement and functional foods
	Theme Area 3: Plant Developmental Biology
	Dr. Aashish Ranjan, NIPGR, New Delhi **Topic**: Optimizing plant developmental features for food security: A case study with photosynthesis
17:30-17:45	**Concluding Remarks and Prize distribution**
17:45-18:00	**High Tea**

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SOCIETY FOR PLANT BIOCHEMISTRY AND BIOTECHNOLOGY (SPBB)

The Society for Plant Biochemistry and Biotechnology was established in May, 1991 with its Headquarters at Division of Biochemistry, Indian Agricultural Research Institute, New Delhi, and registered under the Societies Registration Act of XXI of 1860 on 15th November, 1991. The aims and objectives for which the society was established are as under:

i. To advance Plant Biochemistry, Molecular Biology, Molecular Genetics and Biotechnology research in India.

ii. To provide forum to Plant Biochemists, Molecular Biologists, Molecular Geneticists and Biotechnologists for exchange of scientific knowledge.

iii. To foster high standards in teaching and research in the areas of Plant Biochemistry, Molecular Biology, Molecular Genetics and Biotechnology.

iv. To collect, collate and disseminate information on various aspects of Plant Biochemical, Molecular Biology, Molecular Genetics and Biotechnological research.

v. To work in association and cooperation with other National Societies/ Associations having similar objectives.

vi. To publish a journal covering Plant Biochemistry, Molecular Biology, Molecular Genetics and Biotechnology research.

vii. To do and perform all other acts, matters and things which may assist in, conduce to or be necessary for the fulfillment of the objectives and aims of the Society.

The Society has 831 life Members (upto November, 2018) including Ph.D. scholars, 03 Student/ ordinary members (Year- 2018) and 5 Institutional Members. It has 24 foreign members from 12 different countries. The members are from the disciplines of Biochemistry, Biotechnology and Molecular Biology, Plant Physiology, Genetics, Microbiology, Molecular Genetics, Mycology and Plant Pathology.

The Society had the honour of being led by distinguished scientists/academicians as Presidents : Dr. S. Ramachandran, then the Secretary, DBT (1992-1994); Dr. R.S. Paroda, then the Secretary, DARE & DG, ICAR (1995-2000); Prof. V.L. Chopra, then the Secretary, DARE & DG, ICAR (2000-2005); Dr. S.L. Mehta, former DDG (Education), ICAR and Vice Chancellor, MPUAT, Udaipur.

As its main activity, the Societyis publishing the 'Journal of Plant Biochemistry and Biotechnology' (JPBB) since January 1992. Initially, the Journal was published biannually, but since January 2013, four issues per year are being published due heavy pressure of a large number of MS received from India and abroad. The latest Thompson Reuter 'Impact Factor' 2017 of the Journal is 0.774. It has been our endeavour to timely publish the Journal while maintaining its standard and constantly improving its quality. The members of the Society receive the Journal online, free of cost. Based on the quality/impact factor of the Journal, the Society has been graded and put in category 'A' by the Indian Council of Agricultural Research.

The Journal publishes review articles, research papers, short communications and commentaries in the areas of plant biochemistry, plant molecular biology, microbial and molecular genetics, DNA finger printing, micropropagation, and plant biotechnology including plant genetic engineering, new molecular tools and techniques, genomics and bioinformatics.

The Journal is being Abstracted/Indexed in : Science Citation Index Expanded (SciSearch), Journal Citation Reports/Science Edition, SCOPUS, Chemical Abstracts Service (CAS), Google Scholar, CSA,CAB International, Biological Abstracts, BIOSIS, CAB Abstracts, Elsevier Biobase, Food Science and Technology Abstracts, Global Health, Indian Science Abstracts, OCLC, SCImago, Summon by ProQuest (http://www.springer.com/ life + sciences/journal/13562).

Earlier, the Journal received a rare distinction of being covered in Institute of Scientific Information(ISI)'s Electronic Library Project. From Volume 20 - Issue 1(Jan - June, 2011), the Journal is being co-published by the Society and Springer (India) Private Limited.

Another main activity of the Society has been to organize Symposia/Seminars on topics of current National interest. Since its inception, the Society has organized the following National Symposia/ Seminars and International Conference:

i. The first National Symposium on the "Role of Plant Biochemistry and Biotechnology in Improving Crop Productivity" at the Indian Agricultural Research Institute, New Delhi from November 18-19, 1991.

ii. The second National Symposium on the "Developments in Plant Molecular Biology & Genetic Engineering" at the Tamil Nadu Agricultural University, Coimbatore from December 29-31, 1993.

iii. The third National Symposium on the "Role of Plant Biotechnology in Improving Agriculture-challenges and opportunities, and Physiological & Biochemical Basis of Crop Yield" at the University of Rajasthan, Jaipur from March 23-25, 1995. This Symposium was co-sponsored by the Society for Plant Physiology & Biochemistry.

iv. The fourth National Symposium on "Current Trends in Plant Biochemistry and Biotechnology" was organized at CCS Haryana Agricultural University, Hisar from February 23-25, 1996.

v. The fifth National Symposium on the "Role of Plant Biochemistry and Biotechnology in Improving Crop Productivity" was organized at ICAR Research Complex for NEH Region, Barapani, Meghalaya from March 18-20, 1997.

vi. The sixth National Symposium on the "Role of Plant Biochemistry and Biotechnology in Improving Crop Productivity" was held at University of Agricultural Sciences, Bangalore from March 4-6, 1999.

vii. The seventh National Symposium on "Biotechnology for Sustainability in Agriculture" was held at GB Pant University of Agriculture and Technology, Pantnagar from April 27-29, 2000.

viii. The eighth National Symposium on the "Relevance of Plant Biochemistry and Biotechnology-Modern Trends" was held at The American College, Madurai (TN) from March 1-3, 2001.

ix. The ninth National Symposium on the "New Opportunities and Challenges for Improving Crop Productivity through Biotechnology" was organized at the Department of Biotechnology and Molecular Biology, CCS Haryana Agricultural University, Hisar from February 13-15, 2002.

x. ICAR sponsored Workshop on "Harnessing the Benefits of Biotechnology" was organized in collaboration with the NRC on Plant Biotechnology, Pusa Campus from March 27-29, 2008.

xi. The first "International Conference on Plant Biotechnology for Food Security : New Frontiers" was organized by the Society and the National Research Centre on Plant Biotechnology, in association with the Indian Agricultural Research Institute, New Delhi from Feb 21-24,2012.

xii. The tenth National Conference on "Science of Omics for Agricultural Productivity: Future Perspectives" was organized at GBPUAT during 4-6 March, 2014.

xiii. The eleventh National Symposium on 'Germplasm to Genes: Harnessing Biotechnology for Food Security and Healthwas organized by the Society and the National Research Centre on Plant Biotechnology during 09-11 August, 2015

xiv. The twelfth National Symposium on 'Transgenic Crops in India: Progress and Challenges' was jointly organised at CCSHAU Hisar during 16-17 March, 2016

The Society started 'Dr. N.B. Das Memorial Lecture' in 1993 in the memory of renowned Biochemist late Prof. N.B. Das, the first Head of the Division of Biochemistry, Indian Agricultural Research Institute, New Delhi. The first Memorial Lecture was delivered by Prof. M.S. Naik, then the Head, Division of Biochemistry, Indian Agricultural Research Institute, New Delhi in December, 1993 at the time of second National Symposium. Dr. P.V. Sane, then the Director, National Botanical Research Institute, Lucknow delivered the second Memorial lecture in February, 1996 at the time of the fourth National Symposium.

The Society also started lecture series from this year. The first lecture was delivered by Prof. Dharmendra Saraswat, Purdue University, USA on the topic 'Use of ICT in Food, Energy and Water System' on 09.10.2018 at Division of Biochemistry, ICAR-IARI, New Delhi.

ABOUT THE SYMPOSIUM

A growing world population and dietary shifts associated with economic development are pushing new global demands for food while climate change, depleting soil fertility and water scarcity are threatening our ability to produce food for all at an affordable price. According to the United Nations (UN), global agricultural production will need to be at least 60% higher in 2050 than the levels reported in 2007. This challenge is exacerbated due to continuous drop in the annual rate of crop yield increase resulting from decreasing arable land, emergence of new pathogens and mounting abiotic (drought, salinity, flooding and temperature extremes) stresses.

Since plant-based food forms the major source of human nutrition by providing essential macro- and micro-nutrients, the problem of food insecurity is often intertwined with the more complex challenge of malnutrition. Nearly 825 million people around the globe are undernourished and close to 2 billion are suffering from "hidden hunger" due to inadequate intake of essential micronutrients which has often resulted in the loss of countless lives in many countries. As the development of a country is closely linked with health status of its population, it is becoming increasingly recognized that nutritional security is an enormous challenge with multifaceted social and economic implications; it therefore requires concerted and coordinated efforts to attain nutritional security. There is now a renewed interest in traditional, orphan crops which are rich in micronutrients to correct the nutritional imbalance caused by our overdependence on a few cereal crops. Besides understanding the biochemical and molecular mechanisms governing phytonutrient accumulation in these crops, a new push for breeding improvement of these nutrient-rich crops is needed to make them remunerative to farmers.

There is, thus, a palpable urgency amongst the researchers across the globe to address this seemingly indomitable challenge of food and nutritional security through precise crop optimization with regard to yield, nutrition and plant fitness and performance using modern tools and techniques of molecular biology and biotechnology. The genomics will play a central role in this regard and efforts are underway to enrich the genomic resources of these crops for enhancing the efficiency of plant breeding. The other strategy which holds a great promise for enhancing the crop yield, but has hitherto been overlooked, is the genetic manipulation of certain developmental features of plants such as plant architecture, vasculature architecture and morphological and anatomical features of leaves, which determine the overall crop performance. Optimization of these features may thus have a profound impact on crop yield under normal and challenging conditions. Functional

genomics coupled with phenomics could accelerate discovery of genes and gene regulatory networks underlying these developmental traits of agricultural importance.

Although Biotechnology holds great promise in Indian agriculture and pharma industry, Biotechnology Education seldom finds place in meetings and conferences. Unbridled proliferation of institutes offering Biotechnology degrees at various levels is a matter of concern. Likewise, employment opportunities for new graduates in biochemistry and biotechnology is an aspect that needs wider discussion. We have, therefore, planned to hold a panel discussion on "Biotechnology/ Biochemistry Education" inviting leading academicians, policy makers and Industry representatives.

In view of the above, the Society of Plant Biochemistry and Biotechnology has planned to organize the one-day National Symposium on "Connecting Innovations in Plant Biology for Food and Nutritional Security" on November 19, 2018 at ICAR-Indian Agricultural Research Institute, New Delhi to celebrate its foundation day. The aim of this symposium is to bring together leading scientists and young researchers working in the field of biochemistry & biotechnology and allied sciences to discuss their research findings and share their views about the potential strategies to be adopted to overcome the overriding challenge of food and nutritional security. The symposium will address the following three theme areas:

- Abiotic and Biotic Stress Management
- Nutritional Quality Enhancement
- Plant Developmental Biology

Dr. N.B. Das Memorial Lecture

Dr. N.B. Das
(1908-1971)

Dr. N. B. Das (Nalin Bandhu Das) was born on June 1, 1908 in a family of jurists, educationists and physicians of Chittagong (now in Bangladesh). He graduated in Honours in Chemistry from the Calcutta University. He then joined Bose Institute as researcher and later on served the Bengal Chemical and Pharmaceutical Works for a short term. In 1930s, on being awarded 'The Lady Tata Memorial Scholarship', he proceeded to Germany for higher studies. He was awarded the Doctorate degree for his fundamental research on the biochemistry of enzymes. His Ph.D. research has been quite frequently cited in the text books on enzymology.

He had the privilege of working with two Nobel Laureates. Initially he was with Prof. Hans von Euler at the Biochemical Institute, University of Stockholm, Sweden. Here he worked extensively on the purification and characterisation of enzymes involved in amino acid metabolism in animal system, especially with respect to cofactors/coenzymes, and on the role of ATP in the conversion of DPN to TPN. Later on, as 'The Lady Tata Memorial Scholar', he moved tothe Institute of Medical Chemistry, University of Szeged, Hungary to work with Nobel Laureate Prof. Albert Szent-Gyorgyi, to whom he considered his Guru. He carried out extensive studies on the inhibition of the succinic and lactic-malic dehydrogenases. His research work is widely cited in research papers and books.

After working for nearly 12 years in renowned laboratories abroad, Dr. Das returned back to India on the call given by Prof. P. C. Mahalanobis, the father of modern statistics in India. Patriotic feeling brought Dr Das home to use science and technology for the welfare of people. On his return, he joined the mother campus of Indian Veterinary Research Institute (then Imperial Institute of

Veterinary Research) at Mukteswar to work on cattle vaccines and sera. Subsequently, in the year 1949, he joined the Division of Soil Science and Agricultural Chemistry at IARI as the first Biochemist where he established a Biochemistry Laboratory. This was the beginning of introducing and emphasizing the role of biochemistry in agriculture, especially with respect to the quality of food crops. Dr. Das was the first to study the nutritive quality of crops as influenced by variety, manures and fertilizers. From these studies, the first major recommendation that foliar application of urea and micro-nutrient elements enhances crop yield, was made in early 1950s.

Recognising the importance of Agricultural Biochemistry in improving agriculture, a small Biochemistry Section in the Division of Soil Science and Agricultural Chemistry was transformed into the full-fledged Division of Biochemistry in November 1966, and its founding father Dr. N.B. Das was made the first Head of the Division. Biochemical research and education was tailored to meet the new challenges of Indian agriculture. New Post-Graduate courses were designed to impart skills in new technology that aimed at studying intermediary metabolism, molecular biology, enzymology and nutrition. The first batch of M.Sc. and Ph.D. students was admitted in the academic year 1967-68.

In the formative years, the major emphasis was given on the biochemical and biological evaluation of nutritional value of newly developed strains and varieties of food grains, effect of plant nutrients on nutritional value as well as yield of food grains, biochemical studies in nitrogen fixing bacteria, and utilisation of agro-industrial wastes for the production of lysine as well as enzymes like cellulase and amylase. Under his leadership one of the finest facilities for doing outstanding research in plant biochemistry was built. The technique of PAGE was introduced for the characterisation of soluble proteins and iso-enzymes. Dr. Das guided a number of M.Sc. and Ph.D. students from India and neighboring countries,wholater went on to holdvarious responsible positions within and outside India.

After his superannuation in the year 1970, in recognition of his valuable contributions in the field of biochemistry and his distinct position as a biochemist in the country, he was made Emeritus Scientist by the ICAR.

Dr. N.B. Das was not only a renowned and most respected biochemist, but also an unassuming, quiet and noble gentleman, and his smiling face was characteristic of him. He personified what is best in Indian culture. His commitment to excellence in science was total. This great scientist left for his heavenly abode on 17th November, 1971. The Society of Plant Biochemistry and Biotechnology decided to organise 'Dr N.B. Das Memorial Lecture' to perpetuate his memory.

Session-I

Dr. N.B. Das Memorial Lecture

Speaker: Dr H. K. Das, Former Dean, JNU, New Delhi

Academic Profile of the speaker

Dr. H. K. Das's first love was chemistry. After finishing Intermediate Science in St. Xavier's College, Calcutta, he took admission in Presidency College, Calcutta to study B.Sc. Chemistry Honours in 1950. His next stop was the Department of Applied Chemistry in the University College of Science and Technology of Calcutta University, where he obtained his M.Sc. Tech. degree on his way to fulfil his ambition of starting up of a chemical manufacturing plant. But it was here that he was bitten by the Biochemistry bug. He came under the influence of the late Dr. Sailesh Chandra Roy, who was then a Reader in Biochemistry, which was one of the five sections in the Department of Applied Chemistry. Dr. Das decided to pursue a research career in Biochemistry.

Dr. Das applied for and obtained the Rs. 200 per month Manpower Training Fellowship and initiated his research work. Over the years he studied amino acid metabolism in mitochondria isolated from seedlings of the bean *Vigna sinensis*. He then went on to demonstrate that mitochondria from the same source could synthesize protein. He also presented evidence that these mitochondria contained DNA and some of the mitochondrial proteins were coded for by the mitochondrial DNA.

The Biochemistry section of the Department of Applied Chemistry by then had become a full-fledged Department of Biochemistry of the Calcutta University. Dr. H. K. Das served as an Honorary Lecturer and taught the M.Sc. course of Plant Biochemistry. His Ph.D. degree was in Biochemistry. Later he was also awarded the D.Sc. degree in Biochemistry of the Calcutta University.

In 1964 Dr. Das was awarded the Fulbright fellowship. He went to Stanford University and joined the laboratory of Professor Avram Goldstein as a Research Associate. There he demonstrated that ribosomes in *Escherichia coli* do not wait till transcription is complete, but get attached to the 5′

end of the nascent mRNA soon after coming out of the transcription complex and start synthesizing protein in a coupled manner. In Stanford, Das also elucidated the mechanism by which chloramphenicol inhibited protein synthesis in *E. coli.*

Dr. Das returned to India in 1967 and joined the Scientists' Pool. He developed the Molecular Biology course in the Department of Biochemistry of Calcutta University and taught the course till March, 1969.

In 1969 he was selected as a Senior Biochemist in the Division of Biochemistry of the Indian Agricultural Research Institute. He joined on April 1, 1969 and developed three courses on Molecular Biology, (i) Gene Structure and Replication, (ii) Gene Expression and (iii) Gene Regulation, which were jointly offered by the Division of Biochemistry and the Division of Genetics.

In IARI, Dr. Das demonstrated that lysine deficiency in wheat endosperm proteins resulted not only from decreased synthesis of lysine rich proteins during endosperm development, but also from enhanced degradation of the lysine rich proteins. A proteolytic enzyme, which preferentially degraded lysine rich proteins was partially purified from developed wheat endosperm and characterized.

Das moved to Jawaharlal Nehru University in 1976 and initiated work on nitrogen fixation.

Das's work on nitrogen fixation can be summarized as follows:

(i) Providing physical evidence for the presence of three different pathways of nitrogen fixation by cloning the structural genes of all the three pathways.

(ii) Characterization of a structural gene and a ferredoxin like gene of the vanadium dependent pathway (*vnf*) of nitrogen fixation.

(iii) Cloning and characterization of an operon containing the genes for negative and positive regulation of the *nif* operons (*nifLA* operon).

(iv) Demonstration that DNA gyraseA gene that controls the superhelical status of DNA, is involved in the regulation of the positive regulatory gene *nifA*.

(v) Identification of a positive transcription regulatory element within the coding region of the *nifL* gene.

(vi) Elucidation of upstream activation of the promoters of both *nif* and *vnf* operation.

(vii) Construction of an Azotobacter strain that can replace a substantial amount of urea while sustaining the same wheat yield.

In JNU Dr. Das developed a course on Recombinant DNA Technology and taught it till his retirement in 1998.

Dr. Das contributed to formulation of broad guidelines for teaching Biotechnology in India as a member of the Task Force constituted jointly by the Department of Biotechnology, Government of India and University Grants Commission.

Dr. Das was also served as a member of the Task Force constituted by the Department of Biotechnology, Government of India to advise on the establishment of Bioinformatics network in India.

Dr. Das served as a member of the editorial board of the Indian Journal of Biochemistry and Biophysics during 2005-2008.

In 1978 Dr. Das served as Visiting Professor in the Department of Biology in the San Diego campus of the University of California and in 1979 in the Department of Biochemistry and Biophysics in the San Francisco campus of the same University. In 1989 he served as a Visiting Professor in the Department of Microbiology in the University of Virginia at Charlottesville.

Session-II

Branching Careers in Biochemistry/Biotechnology : Time for Introspection

During the last few decades, the advancements in the fields of biochemistry and biotechnology have contributed tremendously for the growth of various sectors especially the agriculture and medicine and thereby providing major health and economic benefits to the society. The student community was thus fascinated a great deal by these two subjects leading to generation of a large number of graduates, post-graduates and Ph.D. degree holders in the field of biochemistry and biotechnology. The growing interest of the students in this field in turn resulted in the unbridled proliferation of technical institutes, offering degrees in these subjects at various levels, due to social and economic reasons. Till very recently, there were plenty of job opportunities, especially in the academia, for the scholars trained out of these institutes. However, during last few years a sort of plateau seems to have arrived as academic positions are decreasing and research funding is getting tighter. So, there is an urgent need to apprise students about the alternative career paths beyond academia, especially in the industries and to empower and encourage them to pursue these non-traditional paths. Hitherto, our major emphasis had been on developing scientific knowledge and research skills in the students, which enabled them to succeed in academic positions. However, to sustain and thrive in non-traditional career paths, the paradigm needs to be shifted and students must be equipped with additional skill-sets such as scientific communication, networking, artificial intelligence etc. To achieve this objective, we need to start right from the school level- making the desired changes in our course-curriculum at various steps, developing new teaching aids/e-courses/career planning and professional skill developmental programmes etc. Further, the awareness must be generated amongst the students and faculty about the requirements of the private and public sectors and teaching capsules need to be designed accordingly.

Keeping a view of the above, we deemed it appropriate to have a wider discussion on this subject involving all the stakeholders including leading academicians, policy makers and industry representatives for the benefit of the scientific/student community. A panel discussion on the topic "**Branching Careers in Biochemistry/Biotechnology: time for introspection"** has thus been planned in one of the sessions of the current National Symposium on *"Connecting Innovations in Plant Biology for Food and Nutritional Security*", which is being organized by Society for Plant Biochemistry and Biotechnology (SPBB), Division of Biochemistry, ICAR-IARI, New Delhi on 19th November 2018 to commemorate its foundation day.

A. INVITED PANELISTS:

1. Dr. Venkateshwarlu Gudipati, ADG (EQ A&R), ICAR
2. Dr. Purnima Sharma, Managing Director, Biotech Consortium India Ltd. (BCIL)
3. Dr. M.V. Rajam, Professor, Department of Genetics, Delhi University
4. Dr. S.K. Khare, Assoc. Dean R&D and Professor of Biochemistry, IIT, Delhi
5. Dr. Deepak Prem, Seed Regulatory Lead, Monsanto, India
6. Dr. Veda Krishnan, Scientist, Division of Biochemistry, ICAR-IARI, New Delhi

B. POINTS TO PONDER:

- Academic remodeling for broadening career opportunities in the area of Biochemistry and Biotechnology.
- Current initiatives and future Govt. plans/policies for nurturing interest and accelerating growth in the field of Basic Sciences.
- Potential strategies for qualitative and quantitative improvement in the development of comprehensive indigenous teaching resources.
- Bio-entrepreneurship and Bio-enterprise creation in India: a 360 degree SWOT analysis.
- Current requirements and expectations of the biotechnology-based industries from the research community.
- Aspirations of the budding biochemists/biotechnologists in the light of current challenges and opportunities.

Theme Area 1: Abiotic and Biotic Stress Management

ABA Receptors: Prospects for Enhancing Biotic and Abiotic Stress Tolerance of Crops

Santosh Kumar VV, Shashank K. Yadav, Rakesh K. Verma, Sanya Shrivastava, Viswanathan Chinnusamy

Division of Plant Physiology, ICAR-Indian Agricultural Research Institute, New Delhi-110012, India
Email: viswanathan@iari.res.in

Extended Summary

Abscisic acid (ABA) is a key regulator of development and stress responses in plants. Since ABA is the master controller of transpiration and various processes involved in tolerance to abiotic stresses, it is popularly called as the plant stress hormone (Chinnusamy 2008, Cutler et al. 2010, Finkelstein 2013). In addition, ABA also involved in tolerance to biotic stresses as it regulates pathogen-associated molecular pattern (PAMP)-mediated stomatal closure and regulate disease resistance through its synergistic or antagonist interaction with SA, JA, and ethylene (Anderson et al. 2004, Jiang et al. 2010, Xu et al. 2013, Paparella et al. 2014). The enigmatic search for the ABA receptor for more than three decades culminated in identification of ABA receptors (ABARs) in 2009 and elucidation of ABA sinalling pathway in *Arabidopsis* (Ma et al. 2009, Park et al. 2009) and subsequently in many crop plants. The START domain proteins PYRABACTIN RESISTANCE1 (PYR1)/ PYR1-like (PYL)/Regulatory Components of ABA Receptors (RCARs) have been identified as *bona fide* ABA receptors (ABARs) in Arabidopsis (Fujii et al. 2009, Ma et al. 2009, Park et al. 2009). ABAR family in Arabidopsis consists of 14 members. PYR1 and PYL1–PYL3 are homodimers, while PYL4–PYL10 are monomers. The core ABA signaling pathway for stress responsive gene expression and stomatal closure consists of PYR1/PYL/RCAR ABARs, clade A protein Phosphatase 2Cs (PP2Cs) and SnRK2 family ser/thr kinases. The atomic details of mechanisms of ABA–ABAR binding, ABA–ABAR–PP2C binding, and PP2C–SnRK2 binding were elucidated by several crystallographic studies combined with cell biological experiments to validate the biological significance of the observed interactions (Cutler et al. 2009, Fujii et al. 2009, Melcher et al. 2009, Melcher et al. 2010; Peterson et al. 2010; Yin et al. 2010, Yuan et al. 2010; Cao et al. 2013; Okamoto et al. 2013). Under non-stress condition when the ABA concentration is below physiological threshold, PP2Cs are active

and dephosphorylate SnRK2s and block substrate access to the kinase active site by binding to the catalytic cleft of SnRK2. PP2Cs also dephosphorylate and inhibit SnRK1 activity. In stress conditions, ABA accumulates to a higher concentration. ABA binds to the PYL receptors, and the ABA–PYL receptor complex in turn bind to and inhibit PP2Cs. In the absence of PP2C-mediated inhibition, SnRK2s are activated by autophosphorylation. Active SnRK2s phosphorylate and activate/inhibit various enzymes, ion channels and transcription factors and regulate protein activity and transcription necessary for development and stress responses. Upon PYL sequestration of PP2Cs in the presence of ABA, SnRK1 is relieved from repression, and SnRK1 mediates energy signaling leads to altered carbohydrate metabolism, energy conservation, and stress tolerance. The discovery and elucidation of PYR/PYL family of ABARs in *Arabidopsis* triggered work in other plant species and the conservation of PYR/PYL-PP2C-SnRK signalling pathway has been established in diverse plant species.

Crop production is one of the largest consumers of fresh water and uses about 70 % of freshwater worldwide. Enhancing WUE of crops in irrigated agriculture and drought tolerance in rainfed agriculture are major targets for sustaining and enhancing agricultural productivity in the future. Since more than 98 % of water taken up by the plant is lost through transpiration, minimization of transpiration can enhance WUE of agriculture significantly. As ABA is the master regulator of transpiration and drought tolerance, ABA signaling pathway is a potential target for agronomic management of WUE and tolerance to drought and other abiotic stresses. Currently three approaches are being attempted to use the ABA receptors for crop improvement and stress tolerance: 1) Genetic engineering of ABARs (overexpression or knockout), 2) search for cheap agonist and antagonists of ABARs for vaccination of plants, and 3) engineered orthogonal receptors.

Overexpression of ABA Receptors for Enhancement of Stress Tolerance

Overexpression of ABARs is expected to enhance ABA sensitivity, and thus enhanced ABA signalling. Hence this approach is being used by several researches and transgenics were developed in model plants and plants of economic importance. The transgenic *Arabidopsis* overexpressing *PYL5* (*PYL5-OE*) showed reduced water loss and enhanced drought resistance than the wild type (Santiago et al. 2009). The overexpression of *OsPYL/RCAR5* driven by maize ubiquitin promoter in rice also revealed the role of *OsPYL5* in modulating seed germination and early seedling growth in rice (Kim et al. 2012). Interestingly, PYL13, which due to lack of certain conserved residues, does not confirm to function as ABA receptor, when overexpressed in *Arabidopsis* resulted in enhanced

drought tolerance, photosynthetic rate, and water-use efficiency (WUE) in transgenic plants (Zhao et al. 2013). Overexpression of *PYL9* promoted the lateral root elongation through regulation of auxin signalling pathway (Xing et al. 2016) and promoted leaf senescence and drought resistance (Zhao et al. 2016). Overexpression of *Artemisia annua AaPYL9* in *Artemisia* improved drought tolerance by inducing stomatal closure and reducing rates of transpiration and also enhanced secondary metabolite levels (Zhang et al. 2013). Constitutive overexpression of *OsPYL5* resulted in drought and other abiotic stress tolerance of transgenic rice plants, but these transgenic rice plants produced very low yield (Kim et al. 2014).

We identified 12 ABA receptor genes from rice genome, of which two appears to be non-functional as these lack some functional domain. We developed transgenic rice cv. Pusa Sugandh 2/ MTU1010 overexpressing rice ABARs under the transcriptional control of *AtRD29A* promoter. Over expression of rice *ARAR2* enhanced cold tolerance of *Arabidopsis* (Lenka et al. 2018) and rice. OsABAR2 overexpression also enhanced drought tolerance in rice. Similar to *ABAR2*, *ABAR6*, *ABAR9* and *ABAR11* overexpression conferred tolerance to transgenic rice against different abiotic stresses, and play non-redundant roles.

Vaccination of Plants against Biotic and Abiotic Stresses

The discovery of ABARs and the atomic level structural details of ABA receptor–ABA–PP2C complex now enabled the scientists to identify chemical agonists or antagonists of these ABA receptors that can be used for agronomic management of WUE and tolerance to various biotic and abiotic stresses. Timely application of ABA can induce acclimation and tolerance of crops to these stresses. Since ABA is costly, significant efforts are being made to generate synthetic small molecule agonists of ABA receptor for agricultural use. Pyrabactin was used to discover PYR/PYL ABA receptors in *Arabidopsis*. However, our protoplast and *in vitro* ABA signaling pathway reconstitution assays revealed that pyrabactin is an agonist of PYR1 and PYL1, but it antagonizes ABA activation of PYL2. Crystal structures of PYL1– pyrabactin–ABI1 and PYL2–pyrabactin complexes revealed that a single isoleucine (PYR1 and PYL1) versus valine (PYL2) position in the ligand-binding pockets is responsible for pyrabactin agonistic and antagonistic properties. Non-productive binding of pyrabactin occurs without gate closure and prevents receptor activation. This knowledge was used to engineer I137V that converts PYL1 to a pyrabactin-inhibited receptor, A93F in PYL2 that converts PYL2 to a pyrabactin-activated receptor, and identification of pyrabactin-based ABA receptor agonists (Melcher et al. 2010; Peterson et al. 2010; Yuan et al. 2010). Cao et al. (2013) identified

three agonist viz., ABA mimic 1 (AM1), AM2 and AM3. Similar to ABA, treatment with AM1 also inhibited seed germination and minimized excised leaf water loss. The survival rate of DMSO-sprayed plants were only <5 %, while ABA- and AM1-sprayed plants showed 100 and >80 % survival under severe drought stress, respectively (Cao et al. 2013). Further, modification of AM1 with fluorine atoms in the benzyl ring further enhanced the number of hydrogen bonds between the agonist and ABAR ligand-binding pocket. The modified agonist (AMFs) showed significantly higher activity in stomatal closure and stress-responsive gene expression. Foliar application of AMFs conferred drought tolerance to *Arabidopsis* and soybean (Cao et al. 2017).

Okamoto et al. (2013) identified quinabactin as an effective agonist which minimized excised leaf water loss and induced stress-responsive gene expression similar to that of ABA in soybean, barley, and maize. Further, quinabactin foliar spray (50 µM) prior to subjecting the plants to drought stress conferred drought tolerance to *Arabidopsis*, soybean, and barley, but the level of tolerance provided by quinabactin varies in different plant species (Okamoto et al. 2013).

Engineering Orthogonal Receptors

Orthogonal receptor is an engineered receptor that can specifically bind to synthetic ligand (which cannot bind to natural receptor) and then regulate cellular processes, while functionally orthogonal receptor is an engineered receptor, which is not activated by natural level concentrations of the endogenous ligand. The functionally orthogonal receptor is also very useful, as they will be activated by synthetic ligands at very low concentrations, while the natural receptors may require a higher concentration of synthetic ligand for activation. Orthogonal receptors can be used to engineer crop plants, whereas synthetic agonists or antagonists can be used to induce a specific physiological process such as metabolite production, fruit ripening, or tolerance to biotic and abiotic stresses. An orthogonal receptor and ligand pair is highly useful to avoid undesired pleiotropic effect of treatment of crops with antagonists or agonists that activate several receptors and thus general response. Further, these orthogonal receptors can be expressed in transgenic crops by using tissue- or stress-specific or chemical-inducible promoters. In the later case, a combination of chemicals for inducing the expression and activities of orthogonal receptors can yield effective and specific desirable results. Thus orthogonal receptor engineering offers a mean for genetic reprogramming of a specific cellular process or stress response. Melcher et al. (2010) demonstrated that it is possible to design and engineer ABA receptors, which can be activated or inhibited by a synthetic ligand. Pyrabactin activates PYL1 but inhibits PYL2. Site-directed mutagenesis of I137V in PYL1 resulted

in a pyrabactin-inhibited receptor, while A93F and V114I in PYL2 resulted in a pyrabactin-activated receptor (Melcher et al. 2010; Yuan et al. 2010). Further, Mosquna et al. (2011) showed that site-directed mutagenesis can be used to engineer constitutively active receptors, which show high-affinity binding and inhibition of PP2Cs in the absence of ABA. PYL13, which shows very low activity with ABA, could be engineered Q38K-F71L-T135N to convert it into an ABA-activated receptor (Zhao et al. 2013). These studies show the possibilities of engineering ABA receptors, which can be activated by synthetic chemicals (A93F and V114I in PYL2), inhibited by synthetic chemical (I137V in PYL1), and activated by natural ligand (Q38K-F71L-T135N in PYL13) and constitutively active ABA receptors.

Perspectives

Genetic engineering of crops with genes coding for ABA synthesis, catabolism, signaling pathway, and effector genes has demonstrated that genetic modification of ABA pathways can significantly enhance crop yield under abiotic stress conditions. Efforts for identification ABAR agonists and antagonists have also led to enhanced knowledge on ABAR-ligand-binding and identification of novel synthetic molecules that regulate ABARs and drought tolerance. Further, the potential for engineering orthogonal receptors has been demonstrated. These studies on PYL–PP2C–SnRK pathways have laid a strong foundation for genetic engineering and agronomic management of crops for enhanced productivity, quality, and tolerance to biotic and abiotic stresses in the near future.

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Theme Area 2: Nutritional Quality Enhancement

Biotechnology and bioprocessing for nutritional quality improvement and functional foods

Sudesh Kumar Yadav

Center of Innovative and Applied Bioprocessing (CIAB), Sector-81 (Knowledge City), Mohali-140306, Punjab

Email: sudesh@ciab.res.in

Worldwide more than 800 million people are suffering from severe hunger. In developing countries, undernourishment is one the major problem affecting the health of people at large. One of the main reasons for sufficient food availability is poor storage system and supply chain, while for low-nourished is the dependency of larger population on a single, starch-rich crop. Therefore improvement in nutritional quality of commonly used crops for food could be one solution. The inclusion of additional sources of foods to daily diet could be another solution. In this regard, biotechnology or genetic engineering is very effective tool to improve the nutritional quality and quantity of foods. Biotechnology not only improve the scarce nutrients but also the nutrients that are completely absent in the plants. This technique has advantage of speedy improvement compared to the conventional methods. Over last two decades, metabolic engineering has been successfully employed to improve a wide variety of nutritional traits. Recently the innovation in bioprocessing has also been realized to improve the products life, nutritional quality, value to the primary produce, economic growth and ultimately overall societal benefits. Strong attention is needed to apply bioprocessing methods for effective utilization of primary as well as secondary agriculture produce. Bioprocessing is a strong technique for value addition to the fresh produce as well as waste biomass generated by agro-processing industries.

Tea is one of the richest source of antioxidants and therefore contributing towards several health benefits. However, it contains high levels of caffeine and high caffeine intake particularly by women, children and aged people may cause health problems. In view of this, we have developed a selected cultivar of tea with low-caffeine content using RNAi technology. Hence, tea can become a most useful source of beneficial compounds flavonoids, if its caffeine level is decreased to the acceptable level in the plant itself. Flavan-3-ols are the major flavonoids present in tea. Overexpression of

CsF3H cDNA from tea has been found to increase the content of flavan-3-ols and conferred tolerance to biotic and abiotic stress. Transformants have also been observed for increase in primary root length, number of lateral roots, chlorophyll content, and antioxidant system of plants. Similarly, flavonol synthase (FLS) is an important enzyme of flavonoid pathway that catalyzes the formation of flavonols. A novel strategy has been documented for the generation of seedless or less-seeded fruits by downregulation of flavonol biosynthesis through post-transcriptional gene silencing (PTGS) of FLS encoding mRNA. Interestingly, FLS silencing also led to the increase in their nutritional important antioxidants flavan-3-ols such as catechin, epi-catechin and epi-gallocatechin contents. Our comprehensive study has documented the epigenetic regulation of flavonoid biosynthetic and antioxidant pathways during salt-stress exposure of plants and the manipulation of such regulation could be a potential strategy for crop yield enhancement and their nutritional quality improvement.

My group has significantly contributed towards understanding the steviol glycosides biosynthesis at biochemical and molecular levels in Stevia plant. Steviol glycoside and gibberellin biosynthetic routes are known as divergent branches of a common origin in Stevia. We asked a simple fundamental question, as gibberellins are synthesized in other plants then why steviol glycosides, a low-calorie sweetener are restricted in Stevia? To address this, we engineered *Arabidopsis thaliana* for the ectopic over-expression of SrKA13H (ent-kaurenoic acid-13 hydroxylase) cDNA, one of the first enzyme encoding gene that catalyzes the diversion of gibberellins biosynthesis towards steviol glycosides, from *Stevia rebaudiana*. Transgenic *Arabidopsis thaliana* showed significant accumulation of steviol and reductions in endogenous bioactive gibberellins GA1 and GA4. Similarly, a UDP-glycosyltransferase encoded by SrUGT74G1 catalyses the conversion of steviolbioside into stevioside in *Stevia rebaudiana* leaves was overexpressed in transgenic *Arabidopsis thaliana*. Such plants documented significant accumulation of catechinin addition to improved growth and development of plants. Findings have suggested the role of biotechnology, particularly the metabolic engineering in improvement of nutritional quality of crops.

Bioprocessing has recently been recognized as potential tool to impart to the economic gain of farmers and industrial society. In one of our study where liquid whey, a waste generated by milk processing industry was used to produce a very high value rare sugar D-tagatose. We synthesized and characterized a novel hybrid nanoflower of manganese and L-arabinose isomerase and explored its application in synthesis of D-tagatose, a rare sugar of high commercial value. A hierarchical flower-like spherical structure with several nanopetals was self-assembled by using purified recombinant L-arabinose isomerase as the organic component and manganese phosphate as the

inorganic component. Kinetic parameters of L-arabinose isomerase were improved in the hybrid nanoflower. The hybrid nanoflowershave exhibited excellent reusability and reproducibility in the bioprocess.

Rice straw/residues are one of the largest secondary agricultural waste. We are exploring the use of such large agro-waste in the production of value added products through bioprocessing. This biomass mainly contains cellulose, hemicellulose, lignin and silica. Most of the chemical and biochemical processes used for the de-polymerization of structural polymers of lignocellulosic biomass are environment unfriendly and/ or costly. An efficient process based on xylanase hydrolysis of only physically treated rice straw and corn cob has been developed for the production of xylooligosaccharides, a prebiotic useful for gut microflora. Process of xylooligosaccharide production was further improved by developing magnetic-xylanase CLEA. Interestingly, the hydrolysis by our enzymatic forms was found to produce predominantly xylopentose and xylohexose. Hence, the process is environment friendly and the predominant production of xylopentose and xylohexose could find unique prebiotic applications. Additionally, a simple acid-base hydrolytic method has been developed to recover pure silica (SiO_2) nanoparticles and lignin from paddy straw residues, with negligible mineral contaminants. Hence, the recovery of such commercially important molecules would not only provide the possible protection to environmental pollution due to the abundance of paddy straw and its burning but can also contribute to the sustainable economic growth of the country by providing additional income to the farmers.

We have developed green processes for encapsulation or nanoformulation development of bioactives such as quercetin, botulin, catechins, podophyllotoxin, etc to improve their aqueous solubility, stability and efficacy. An innovative approach was adopted where in situ synthesized silver nanoparticles from leaf extract mediated reduction of silver nitrate were simultaneously impregnated into the matrix of cellulose nanocrystals (CNCs) isolated from bamboo, for formation of nanobiocomposites (NCs) in film and ointment forms. NCs possessing water absorption capacity and strong antibacterial activity showed synergistic effect on in vivo skin wound healing and documented faster and significant wound closure in treated mice.

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Theme Area 3: Plant Developmental Biology

Optimizing plant developmental features for food security: A case study with photosynthesis

Aashish Ranjan

Staff Scientist IV & Ramalingaswami Fellow
National Institute of Plant Genome Research, New Delhi

The exponential increase in population and rapid global environmental changes observed in recent years are serious threats to sustainable food production for the planet [1]. Developing crop varieties in order to achieve greater yields has been a major focus of plant biologists and breeders with a view to ensuring food availability for an increasing world population under changing environmental conditions [2-3]. The optimization of plant developmental traits has great potential for sustainable increase in crop yield, as plant performance is strongly associated with, and dependent on, plant development and growth. A number of plant features and traits, such as overall plant architecture, leaf morphological and anatomical traits, vascular architecture and flowering time, are important determinants of photosynthetic efficiency, and hence the overall performance of crop plants [4]. These features can, thus, be considered part of a developmental module that dictates crop performance and yield.

The importance of plant developmental features in increasing crop yield potential became evident during the 'green revolution', when an unprecedented increase in yield was achieved by breeding for semi-dwarf varieties of rice and wheat [5]. Reducing plant height, for example by altering gibberellic acid (GA) biosynthesis and signaling [5-6], had an obvious advantage in reducing lodging as well as increasing the number of tillers. Knowing that leaves are the primary site of photosynthesis, it is logical to imagine that plants can also be engineered to produce leaves with optimal features for more efficient light harvesting, leading to a faster growth rate and increased yield. Indeed, leaf photosynthesis is amenable to genetic manipulation and could thus be targeted to improve photosynthesis and yield [7]. The plant vasculature is another feature that regulates the overall performance of a plant by not only providing mechanical strength but also serving as a channel for the transport of water, minerals and photosynthates [8]. Thus, genetic manipulations that alter these developmental traits in a desirable way may mark a significant step forward in increasing crop yield.

Engineering or breeding for developmental traits with the aim of improving photosynthetic efficiency, and thus yield, requires a thorough understanding of the genetic basis of these traits. The yield potential of rice from improved plant type, semi dwarf and high yielding varieties, has been largely exhausted. Improvement in crop yield due to crop management and genetic gain in harvest index is approaching a plateau over the last decade, and further increase in yield will require increase in photosynthetic capacity. Photosynthesis is a multifaceted process with contributions from leaf developmental features, canopy architecture, biochemical reactions, and source-sink interactions. Therefore, improving photosynthetic efficiency shall include optimizing leaf and canopy developmental features along with improving performance of core photosynthetic enzymes and efficient photoassimilate partitioning [9].

Leaf shape, size and thickness are some of the most important morphological features that can directly affect plant yield. These morphological parameters determine cell number, chlorophyll content and ribulose-1,5-bisphosphate carboxylase/oxygenase (RuBisCO) per unit area exposed to sunlight, thus influencing leaf photosynthetic rate [3]. Leaf morphology is dependent on an intricate network of multiple processes such as cell division, cell expansion, growth axis establishment, and the differentiation and specification of tissues [10]. This network, in turn, is subject to regulation by phytohormones, transcription factors and changes in the mechanical properties of tissues. Leaf anatomical features vary substantially among and within plant species, and are closely related to photosynthetic efficiency under different environmental conditions. Leaf anatomy influences photosynthesis by affecting the distribution of light in the mesophyll, CO_2 diffusion, leaf temperature and leaf water relations [11]. The importance of leaf anatomical features for improved photosynthetic efficiency, and thus the yield, has been evident during the evolution in nature as C_4 plants [12]. The characteristic 'Kranz anatomy' of C_4 plants – a special developmental architecture in which mesophyll cells are surrounded by specialized bundle sheath cells in leaves – helps to increase photosynthetic

efficiency and allow better photoassimilation of atmospheric CO_2. This is also closely associated with an increase in vein density, which is likely to be due to auxin-related modifications, in C_4 plants. The genetic basis of these developmental differences between C_3 and C_4 plants is just beginning to be deciphered [13]. A global consortium based at the International Rice Research Institute (Philippines) aims to convert C_3 rice to C_4 by manipulating the biochemistry and anatomy of the plant [14] (http://c4rice.irri.org/). Besides converting C_3 plants to C_4 plants, there are many other potential ways of changing leaf anatomical features, for example to maintain uniform light

distribution within a leaf and more efficient absorption and usage of light energy, to increase photosynthesis. However, the quantitative effects of these anatomical changes on photosynthesis need to be evaluated. The shape and size of mesophyll cells might also be critical factors for light distribution as well as for CO_2 diffusion to RuBisCO, a key CO_2-fixing enzyme [12, 15]. There is potential to increase photosynthetic efficiency by <"20% by reducing resistance to CO_2 diffusion and optimizing the shape and size of mesophyll cells [3, 15]. Although much progress has been made in understanding the genetic regulatory mechanisms that control leaf growth, the molecular mechanisms regulating leaf anatomical traits that influence photosynthesis are still largely unknown. To allow the engineering of a specific leaf anatomy that would achieve a more homogeneous internal light distribution, more efficient CO_2 delivery and improved water transport capacity, more effort is required to study the genetic mechanisms underlying different leaf structures.

The natural genetic variation in leaf photosynthesis, and underlying developmental, biochemical, and genetic basis is an overlooked and untapped resource. The genus *Oryza*, which includes cultivated rice (*Oryza sativa* and *Oryza glaberrima*) and more than 20 wild relatives, offers tremendous genetic variability to explore photosynthetic differences and underlying biochemical and developmental differences. We observed higher net leaf photosynthesis in some wild relatives of rice compared to cultivated varieties. Leaf photosynthesis per unit area was strongly correlated with leaf mesophyll and vascular features. Wild species with higher leaf photosynthesis per unit area had specific developmental features, such as larger mesophyll cells with more chloroplasts, distribution of chloroplasts along the mesophyll cell wall, larger and closer veins, and smaller number of mesophyll cells between two consecutive veins. The wild species with higher photosynthesis also exhibited striking differences in leaf shape and size, as will as differences in Shoot Apical Meristem (SAM) size and leaf initiation rate. We are, currently, investigating the genetic basis of leaf morphological and anatomical differences that could be attributed to differences in photosynthesis, using transcriptomics and kinematics approaches.

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PA-01

Exogenously-sourced ethylene modulates tolerance mechanisms in Indian mustard under metal stresses

M. Iqbal R. Khan[1,2] and Nafees A. Khan[1]

[1]Plant Physiology and Biochemistry Laboratory, Department of Botany, Aligarh Muslim University, Aligarh-202002, Uttar Pradesh

[2]Department of Botany, Jamia Hamdard, New Delhi-110062

Email: iqbal.khan@jamiahamdard.ac.in

Heavy metal (HM) contamination of agricultural soil is primarily related to the abrupt anthropogenic activities. Plants frequently exposed to high concentration of HMs shows adverse effects. In the present study, ethylene significance was studied in mustard (*Brassica juncea*) plants exposed to nickel (Ni) and zinc (Zn) stress. Application of Ni and Zn at the rate of 200 mg kg^{-1} soil induced increased ethylene formation and oxidative stress and reduced photosynthesis, relative growth rate and dry mass of mustard plants. Exogenous ethylene, as ethephon increased reduced glutathione (GSH) through increased sulfur (S) assimilation and maintained cellular homeostasis. Concomitantly, it also increased proline metabolism and maintained water relations of plants. Taken together, photosynthesis and growth was protected with application of ethephon under Ni and Zn stress through increase S-assimilation and proline. The result was also confirmed by using ethylene action inhibitor, norbornadiene (NBD), which significantly inhibited both S assimilation and proline metabolism resulting in less protection of photosynthesis and growth of plants. The results of the present study could be exploited in situation where plants are grown under higher level of Ni and Zn conditions.

PA-02

In vitro selection against drought in soybean (*Glycine max* (L.)

Nishi Mishra, M.K. Tripathi, Sushma Tiwari and Shikha Upadhyay

Department of Plant Molecular Biology and Biotechnology, College of Agriculture, RVSKVV, Gwalior, Madhya Pradesh

Email: sushma2540@gmail.com, drmanojtripathi64@gmail.com

Soybean [*Glycine max* (L.) Merrill] is one of the major legume crops in the world, providing an abundant source of oil, protein, macronutrients and minerals. Drought is a significant limiting factor for productivity inhibiting plant growth due to reduced water absorption and nutrient uptake. Drought stress, which usually occurs at pod filling stages, may cause significant yield losses, up to 40% in a year, and it deteriorates the seed quality of soybean. *In vitro* selection offers an immense potential for the quick and comprehensive generation of useful somaclones or mutants for resistance against various biotic and abiotic factors. These plants may serve as an excellent donor of the resistance gene(s) in breeding programmes. During present investigation drought tolerance selection of soybean lines generated from somatic embryogenesis using different concentrations of osmotic stress simulation of poly-ethylene glycol (PEG). Two drought susceptible soybean genotypes *namely*: JS335 and RVS 2001-4 were used in this study. *In vitro* selection was in embryogenic callus level using media containing PEG (0, 5, 10, 15, and 20%) was done with both genotypes. The results showed that after 3-4 months in the selection medium, RVS 2001-4 genotype showed higher percentage of fresh callus (surviving callus) and number of embryogenic callus as compared to JS335. PEG in higher concentration decreased the percentage of fresh callus and number of embryogenic callus in both genotypes. RVS 2001-4 showed more survivability and produced more cotyledonary-stage embryos after transferring in MS regeneration medium. Ten somatic embryos of genotype RVS 2001-4 and 4 of genotype JS335 from selection with PEG successfully germinated and regenerated into plantlet(s) as putative drought-tolerant somaclone candidates. This trend showed possibility for development of tolerant lines against drought in soybean.

PA-03

Siderophore induced antioxidant enzyme in hydroponically grown tomato and wheat plant

Abiraami TV[1], Mayur Naitam[1], Bhupinder Singh[2] and Archna Suman[1]

[1]Divisions of Microbiology, [2]CESCRA, ICAR-Indian Agricultural Research Institute, New Delhi-110012

Email: abiraamiabi6363@gmail.com

Iron is one of the major limiting factors and essential nutrients of plant life. Plants have evolved several strategies to enhance Fe acquisition, but increasing evidence has shown that the intrinsic plant-based strategies alone are insufficient to avoid Fe deficiency in Fe-limited soils. Soil micro-organisms also play a critical role in plant Fe acquisition by the production of siderophore that chelates ferric iron with high affinity. Iron homeostasis in plants is important as iron is cofactor of many antioxidant enzymes like catalase, guaiacol peroxidase etc., however excess iron present participate in the Fenton's reaction producing reactive oxygen species that damage the cell. The present study addressed the role of three chemotyped siderophore producing bacterial isolates (BC) and their siderophore preparation (Sid) namely catecholate – *Pantoea agglomerans* (BC1 and Sid1), hydroxamate – *Pseudomonas plecoglossida* (BC2 and Sid2) and carboxylate- *Lactococcus lactis* (BC3 and Sid3) effect on antioxidant stress enzyme of wheat and tomato grown under Fe sufficient (100 µM EDTA) and Fe deficient (1 µM EDTA) condition in hydroponic system. In wheat plant, catalase upon BC1 improved 35.5% and 30% than Fe+ and Fe- control plants. Guaiacol peroxidase upon Sid1 improved 14.7% and 11% than Fe+ and Fe- control plants. In tomato, catalase BC1 and Sid3 influenced maximum under both conditions and the effect was at par with each other. The average enzyme production of both catalase and guaiacol peroxidase was four fold higher in wheat than tomato. The guaiacol peroxidase exhibited higher activity than catalase irrespective of crop and treatment. Catecholate *Pantoea agglomerans* was found to be best in escalating the enzyme activity than other chemotyped siderophore. No significant variation occurs in other treatments but found to influence better than Fe^{-} control. Thus siderophore producing bacterial isolates and their siderophore preparation influenced the antioxidant enzymes underlying siderophore mechanisms in the promotion of plant Fe acquisition.

PA-04

Starch accumulation in rice grains subjected to drought during grain filling stage

Prathap V, Kishwar Ali, Archana Singh, Veda Krishnan, Shelly Praveen, Aruna Tyagi

Division of Biochemistry, ICAR-Indian Agricultural Research Institute, New Delhi-110012

Email: prathuv1994@gmail.com

Starch is a major component of rice grains. The starch biosynthesis pathway mainly take place in chloroplast of photosynthetic tissue, while in non-photosynthetic tissue, amyloplast is major site for starch biosynthesis. The starch synthase (SS) and ADP-glucose pyrophosphorylase (AGPase) are major rate limiting enzymes involved in starch biosynthesis pathway. Drought is a major environmental constraint that affects crop yield by affecting the physiological, biochemical and molecular responses at cellular level. The water is a critical element for rice during the grain filling stage. The drought stress during grain filling stage severely affects the quantity and quality parameters of starch components and also greatly reduces crop yield. This study thus has focused on the estimation of biochemical parameters such as starch, amylose, amylopectin, resistant starch, sucrose, starch synthase and AGPase activity and expression analysis of genes encoding SS and AGPase in two contrasting rice genotypes i.e. N22 (drought tolerant) and IR64 (drought susceptible) after 21 days of drought stress in leaves, roots and grains. It was found that, starch, amylopectin, and sucrose content in immature grains were found to be increased under the drought, while amylose content decreased in both the varieties. However, the starch and sucrose content of leaves and roots were found to be decreased under drought stress in both varieties. The study also showed better yield performance of drought tolerant N22 when compared to drought susceptible IR64. Another key aspect of this study was that SS and AGPase activity was found to be increased in immature grains in both the rice cultivars under drought stress. The SS activity decreased in leaves while AGPse activity was found to be increased under drought stress in both varieties. Quantitative real time PCR was done for the six genes encoding for SS isozymes and six genes encoding multi-subunit protein AGPase. The study revealed spatial and temporal differential expression of genes under the drought stress in both the varieties.

PA-05

Identification of metal stress related gene regulatory network in model plant *Arabidopsis* using fRMA vector

Kuntal Kumar Dey[1], Lochna Patar[1], Debashis Panda[1], Jagjjit Sahu[1], Priyabrata Sen[2], Mahendra Kumar Modi[1,2]

[1]Distributed Information Centre, [2]Department of Agricultural Biotechnology, Assam Agricultural University, Jorhat-785013, Assam

Email: mkmodi@gmail.com, mkmodi@aau.ac.in

Plants become susceptible to toxic metal ions under reduced Potassium (K) and/or Iron (Fe) concentration in rhizosphere. Plants have developed their own response mechanism to overcome such situations. In this study an *Arabidopsis* abiotic stress-related Frozen Robust Multiarray Analysis (fRMA) vector was developed based upon microarray data versatility. This vector was used to analyze microarray data of total RNA expressed in Arabidopsis plants during Fe and K stress separately. The Protein-Protein Interaction (PPI) network of co-expressed genes during metal stress was developed based on Pearson's co-relation coefficient. From this, two sub clusters were extracted that appeared in both the stress situations. Among these, cluster one was found to be highly significant containing 282 interactive proteins. The enrichment analysis of cluster one showed that a large number of these proteins participate in various metabolic activities, such as secondary metabolite production, hormone synthesis, activation of transporter molecules and chelators, which are modulated during exposure to metal ion stress. Our study shows that large scale gene expression data can be efficiently used for identification of metal stress tolerant genes module in plant systems biology through the fRMA vector method.

PA-06

NO defense network in wheat: trigger the biochemical traits associated with stress-tolerance and nutritional quality of wheat grains under heat stress

Ranjeet R. Kumar[1], Sumedha Hasija[1], Suneha Goswami[1], Jyoti P. Singh[1], Ansif Ali[1], Shahnoor Alam[1], Bhargav DV[1], Abhishek Chitranshi[1], Shivdhar Singh[2], Gyanendra P. Singh[3], Chinnusamy Viswanathan[4] and Shelly Praveen[1]

[1]Division of Biochemistry, [2]CESCRA, [4]Division of Plant Physiology, ICAR-Indian Agricultural Research Institute, New Delhi-110012; [3]Indian Institute of Wheat and Barley Research, Karnal, Haryana

Email: ranjeetranjaniari@gmail.com; shellypraveen@hotmail.com

Heat stress drastically reduces the yield and quality of wheat grains. High temperature during critical stages (pollination and grain-filling) causes improper fertilization, and formation of defragmented granules and shriveled seeds. Hormones and signaling molecules modulates the tolerance potential of the plants under stress. Nitric oxide (NO) is an important signaling molecule involved in triggering diverse physiological and biochemical processes under adverse conditions. Here, we studied the effect of sodium nitroprusside (SNP) (150 µM as decided based on pilot experiment) on heat stress-tolerance and grain quality related traits of two contrasting wheat *cvs*. Raj3765 and HD2932 under differential HS (T_1 - 30ºC, 1 h; T_2 - 38ºC, 1 h), as compared to control (22±2ºC). The expression of important stress-associated genes (as identified through Transcriptome sequencing) was observed upregulated in response to NO and HS; small *HSP17* showed maximum fold increase in the expression. Similarly, the network of antioxidant enzymes (SOD, CAT, and GPX) were observed triggered in response to NO, HS and NO+HS at different stages of growth; maximum activities were observed in Raj3765, as compared to HD2932. NO was observed to enhance the accumulation of proline and free amino acid in cytoplasm under HS; accumulation was observed higher in Raj3765, as compared to HD2932 during different stages of growth. Grain quality related traits like carbohydrate (starch) and proteins (gliadin) were observed increase in response to NO under HS; maximum accumulation was observed in Raj3765. Heat stress was observed to increase the activities of α/β amylases in developing grains involved in degrading the starch quality. NO was observed to decrease the amylolytic activity in both the *cvs.*; very low amylolytic activity was observed in thermotolerant, as compared to thermosusceptible *cvs*. Exogenous application of NO (150 µM) at different stages can be used as cheap technology for mitigating the problem of terminal HS in wheat – a farmer friendly approach for maintaining the quality of grains under present threat of global climate change.

PA-07

Unravelling the expression of novel manganese superoxide dismutase (Ta-MnSOD) – a potential biochemical marker for thermotolerance in wheat (*Triticum aestivum*)

Ranjeet R. Kumar[1], Sumedha Hasija[1], Suneha Goswami[1], Jyoti P. Singh[1], Ansif Ali[1], Shahnoor Alam[1], Bhargav DV[1], Abhishek Chitranshi[1], Shivdhar Singh[2], Gyanendra P. Singh[3], Chinnusamy Viswanathan[4], Shelly Praveen[1]

[1]Division of Biochemistry, [2]CESCRA, [4]Division of Plant Physiology, ICAR-Indian Agricultural Research Institute, New Delhi-110012

[3]Indian Institute of Wheat and Barley Research, Karnal, Haryana

Email: ranjeetranjaniari@gmail.com; shellypraveen@hotmail.com

Heat stress is one of the major problems in cereals affecting the growth and development of grains. HS causes oxidative bursts damaging the organelles and nascent proteins. Plant has inherited network of antioxidant defence system to neutralize the effect of ROS. Superoxide dismutase (SOD) provides the first line of defense against the HS by regulating the accumulation of peroxide radicals and H_2O_2 inside the cells. Here, we report identification and cloning of putative Mn-SOD gene from wheat *cv.* HD2985 through de novo assembly. The gene was characterised to have 500 aa with 5'UTR of 29 and 3'UTR of 18 aa. Spatio-temporal expression analysis of *MnSOD*II showed many fold upregulation in thermotolerant wheat cvs. at different stages of growth and development and under differential HS. The abundance of transcript was observed in cytosol and chloroplast. The purified protein of MnSODII expressed through heterologous system showed very high activity (7.5-fold), as compared to SOD extracted from wheat. Kinetics showed the MnSOD to have maximum activity in response to temperature of 36ºC, pH of 9.0 and had K_m value of 1.75 with pyrogallol as substrate. Homologous accumulation of MnSOD protein was observed through immunoblotting in leaves and endospermic tissues. MnSOD, being active at pivotal position, need to be further characterized and used for manipulating the abundance of ROS in wheat in order to cope up with the problem of terminal HS.

PA-08

iTRAQ based identification of stress-associated proteins and characterizing their role in defense and nutrition associated pathways in wheat under heat stress

Ranjeet R. Kumar[1], Suneha Goswami[1], Shahnoor Alam[1], Jyoti P. Singh[1], Sumedha Ahuja[1], Bhargav D.V.[1], Ansheef Ali[1], Abhishek Chitranshi[1], Gyanendra K. Rai[2], Bhupinder Singh[3], Gyanendra P. Singh[4], Himanshu Pathak[5], Viswanathan Chinnusamy[3] and Shelly Praveen[1]

[1]Division of Biochemistry, [3]Division of Plant Physiology,
ICAR-Indian Agricultural Research Institute, New Delhi-110012
[2]Sher-E-Kashmir University of Agriculture Science and Technology, Chatta, Jammu
[4]Indian Institute of Wheat and Barley Research, Karnal, Haryana
[5]Central Rice Research Institute, Cuttack, Orissa
Email: ranjeetranjaniari@gmail.com

Terminal heat stress has detrimental effect on the growth and yield of wheat. Very limited information's is available on heat stress-associated active proteins (SAAPs) in wheat. Here, we have identified 159 proteins groups with 4271 SAAPs in control (22±3ºC) and HS-treated (38ºC, 2 h) wheat *cvs*. HD2985 and HD2329 using iTRAQ. We identified 3600 proteins to be upregulated and 5825 proteins to be downregulated in both the wheat *cvs*. under HS. We observed 60.3% of the common SAAPs showing upregulation in HD2985 (thermotolerant) and downregulation in HD2329 (thermosusceptible) under HS. GO analysis showed proton transport (molecular), photosynthesis (biological) and ATP binding (cellular) to be most altered under HS. Most of the SAAPs identified were observed to be chloroplast localized and involved in photosynthesis. Carboxylase enzyme was observed most abundant active enzymes in wheat under HS. An increase in the degradative isoenzymes (α/β-amylases) was observed, as compared to biosynthesis enzymes (ADP-glucophosphorylase, soluble starch synthase, etc.) under HS. Transcript profiling showed very high relative fold expression of *HSP17*, *CDPK*, *Cu/Zn SOD*, whereas downregulation of *AGPase*, *SSS* under HS. The identified SAAPs can be used for targeted protein based precision wheat breeding program for the development of 'climate-smart' wheat.

PA-09

Evaluating the potential effect of seed priming technique in improving drought tolerance of rice

Mahesh Kumar Samota[1], Monika Awana[1], Sunil Warwate[1], Veda Krishnan[1], Suresh Kumar[1], Aruna Tyagi[1], Shelly Praveen[1], S.V. Amitha Mithra[2], Rakesh Pandey[3] and Archana Singh[1]

[1]Division of Biochemistry, [3]Division of Plant Physiology,
ICAR-Indian Agricultural Research Institute, New Delhi-110012
[2]ICAR-National Research Centre on Plant Biotechnology, Pusa Campus, New Delhi
Email: sarchana19@gmail.com

Rice (*Oryza sativa* L.) is the staple food and its requirement has been increased by more than two folds during the last five decades. Rice being drought sensitive crop is exhibiting serious deleterious effects due to oxidative burst when exposed to water deficit condition especially at reproductive stage. Till the date no strategic details available to mitigate drought stress in agronomically important food crops. Potential of seed priming for drought stress management is a challenging task. Experiments were conducted at reproductive stage of contrasting rice genotypes (Nagina-22, drought-tolerant and Pusa Sugandh 5, drought-sensitive) raised from seeds primed with elicitor-methyl jasmonate (MJ), micronutrients-iron (Fe), Zinc (Zn) and with their combinations. A progressive increase was observed in relative water content (RWC), proline accumulation (~2 fold), chlorophyll content (~0.6 fold), carotenoids, (1.5-2.5 folds) and activity of antioxidant enzymes (catalase, CAT; ascorbate peroxidase, APX; superoxide dismutase, SOD) in the primed samples under drought condition. CAT and SOD activities were found more in Fe alone and MJ+Fe combination while APX activity was observed higher in the MJ+Zn combination. Although in both the cases these were comparable with MJ+Fe+Zn combination.CAT and APX activities were found 1.4 and 4.0 folds higher in the sensitive genotype than the tolerant respectively. Overall, the seed priming with MJ+Fe+Zn combination was found to be most effective in mitigating drought stress induced oxidative burst for improved tolerance in both the genotypes. Thus, seed priming technique being user friendly as well cost effective could be of use to improve drought tolerance in rice for increased yield under dry land and rain-fed areas.

PA-10

Mitogen-activated protein kinase: A master regulator of network pathway associated with heat stress-signalling in wheat

Shahnoor Alam[1], Ranjeet R. Kumar[1], Suneha Goswami[1], Jyoti P. Singh[1], Sumedha Ahuja[1], Bhargav D.V.[1], Ansheef Ali[1], Abhishek Chitranshi[1], Gyanendra K. Rai[2], Bhupinder Singh[3], Gyanendra P. Singh[4], Himanshu Pathak[5], Viswanathan Chinnusamy[3] and Shelly Praveen[1]

[1]Division of Biochemistry, [3]Division of Plant Physiology, ICAR-Indian Agricultural Research Institute, New Delhi-110012

[2]Sher-E-Kashmir University of Agriculture Science and Technology, Chatta, Jammu

[4]Indian Institute of Wheat and Barley Research, Karnal, Haryana

[5]Central Rice Research Institute, Cuttack, Orissa

Email: ranjeetranjaniari@gmail.com; shellypraveen@hotmail.com

Wheat is staple food all over the world, providing 12% of the carbohydrate and 40% of the energy in the diet. Wheat is highly sensitive to terminal HS reducing the quantity and quality of grains. Mitogen-activated protein kinase has been characterised to be the master switch regulating the cross-talk between the biotic and abiotic stresses; acts as signalling molecule in activating the defence mechanism of crops under stress. Here, we have identified 64 novel transcripts showing homology with MAPK through *de novo* transcriptomic approach. Based on the digital fold expression, transcript_1494 was targeted for the cloning. RT-PCR amplification showed amplicon of ~1.3 kb which was further cloned, sequenced by Sangers di-deoxy method and submitted in NCBI GenBank (acc. no. KT835664.1). BLASTn search showed maximum homology with transcript of *Hordeum vulgare* (acc. No. AK353742). BLASTp analysis showed maximum homology with MAPK protein in *Zea mays* (accession. no. NP_001167676.1). ORF analysis showed the presence of 314 aa. Conserved domain search showed the presence of Serine/Threonine type kinase (STK) domain in the sequence. Expression analysis of MAPK showed many fold increase in the expression in response to HS. Stage specific expression analysis showed abundance of transcript during mealy-ripe, as compared to milky-ripe stage under HS. Expression was observed maximum in thermotolerant *cvs.* (HD3059 and Halna) compared with thermosensitive (HD2329 and PBW550) under differential HS. There is, however, need to explore novel MAPKs from contrasting wheat cultivars in order to characterize their role in signalling and thermotolerance of wheat. Over-expression of candidate MAPK in desirable lines of wheat will pave the way for the development of 'climate-smart' wheat.

PA-11

Identification and characterization of microRNAs from flowers in desi and kabuli type chickpea by high-throughput deep sequencing

Lalbahadur Singh[1], Deshika Kohli[1], Bharadwaj Chellapilla[2] and Pradeep K. Jain[1]

[1] ICAR-NRC on Plant Biotechnology, Pusa Campus, New Delhi-110012

[2]Division of Genetics, ICAR-Indian Agricultural Research Institute, New Delhi-110012

Email: jainpmb@gmail.com

Chickpea (*Cicer arietinum*) is the most important legume crop in the Indian subcontinent. It is very important to understand complex gene regulatory mechanisms to improve desired traits in plant(s) of choice. With identification of different small RNAs governing important/key traits in plants, the understanding of gene regulation has become all the more interesting but complicated. MicroRNAs (miRNAs) are essential components of complex gene regulatory networks that are involved in plant development and response to various environmental stresses. In the present study, two small RNA libraries from flower tissues of ICC4958 (desi) and Pusa1053 (kabuli) type chickpea were constructed. Computational analysis of small RNA sequences led to identification of 198 total miRNAs, consisting of 112 conserved miRNAs distributed in 25 different families and 86 novel miRNAs in desi and kabuli flower tissues. Poly(A)-based qRT-PCR (Quantitative real-time PCR) revealed nine conserved and four novel miRNAs to be differentially expressed between desi and kabuli flower tissues. Both, miR159 and miR172, were up-regulated in desi flower and are known to target different APETALA genes which are repressors of flowering, thereby suggesting involvement of miRNAs in flowering. The target genes of all the identified miRNAs were predicted by psRNATarget web server using chickpea transcriptome and genome database. The predicted target genes were extensively involved in different biological processes involving a large number of gene families. The present study has identified different conserved and novel miRNAs along with their expression analysis thereby laying the foundation for studies on gene regulation in response to chickpea flower development.

PA-12

Genome-wide identification of AP2/ERF transcription factors in chickpea (*Cicer arietinum* L.) and their expression profiling under abiotic stress conditions

Pawan Mainkar, Deshika Kohli, Parameswaran Chidambaranathan, J. Prasanth Tej Kumar, Nimmy M.S., Ramamurthy Srinivasan and Pradeep K. Jain

ICAR-NRC on Plant Biotechnology, Pusa Campus, New Delhi-110012
Email: jainpmb@gmail.com

Drought is one of the major constraints for productivity in chickpea, a legume. The role of several classes of genes including certain transcription factor (TFs) has been well established in imparting drought tolerance or susceptibility in plants. One such family of TFs is APETALA2/ethylene responsive element binding factor (AP2/ERF) superfamily. In this study we identified putative 143 ERF genes named as CaERF1-143 (Ca- *Cicer arietinum*). Phylogenetic analysis categorized 141 (excluding two truncated genes) ERF gene family into two different sub-families, DREB and ERF. Among 141 genes, 137 belonged to ERF/AP2 subfamily and only two soloist genes were identified. Some conserved motifs were also identified that are specific to each subgroup. For better understanding of the significance of each subgroup of ERF and DREB subfamily, one representative gene from each subgroup was selected for the quantitative real-time PCR (qRT-PCR) based expression analysis in two chickpea cultivars, namely, ICC4958 (drought tolerant) and ICC1882 (drought susceptible) in response to drought, ABA and ethephon treatments. CaERF29, belonging to group B4 (ERF subfamily) was significantly up-regulated (about 190-fold) under drought stress in both the cultivars (ICC4958 and ICC1882). CaERF51 and CaERF70 genes showed contrasting expression behavior in two cultivars. The results in this study support the hypothesis that ERF family members are involved in regulating environmental stress. In summary, the genome-wide characterization, evolutionary analysis, and expression pattern of CaERF genes in chickpea provides valuable information for characterizing the functions of these genes and utilizing them to impart stress tolerance in plants.

PA-13

Osmotic stress induced root growth and regulation of root development related genes in progenitors of wheat

Sneha Tiwari[1], Pramod Awakale[1], Vinod[2], Monika Dalal[1]

[1] ICAR-NRC on Plant Biotechnology, Pusa Campus, New Delhi-110012

[2]Division of Genetics, ICAR-Indian Agricultural Research Institute, New Delhi-110012

Email: monikadalal@hotmail.com

Maintenance of root growth during water deficit can significantly contribute to yield stability by enhancing the water and nutrient mining from the soil. In this study, three accessions each of *Triticum monococcum, Aegilops speltoides* and *Aegilops tauschii* were analysed for root traits under two moderate levels of osmotic stress viz. -0.5 (S1) and -1.48 bars (S2), for 10 days at seedling stage. Analysis of root traits revealed significant differences in total root length, seminal root number and lateral roots among the diploid accessions. Two of *T. monococcum* accessions (A1 and A3) showed significantly higher total root length, primary root length and seminal root number under control conditions. *T. monococcum* accession (A2) showed highest increase in total root growth under both S1 and S2 stress. Primary root length was mostly unaffected or reduced in all accessions at S1 or S2 stress, except in *T. monococcum* A2 and *Ae. speltoides* B3 accession which showed 17% and 34% increase in primary root length. Stress mediated regulation of orthologs of five root related genes was analysed in diploid accessions. The expression levels of *BREVIS RADIX*, a transcription factor modulating root length was significantly up regulated in *T. monococcum* A2 accession which showed significant increase in total root length and primary root length under osmotic stress. Osmotic stress up regulated the expression of *MIZU-KUSSEI1*, *Crown rootless 1* and *NAM/ATAF/CUC 1* (*NAC1*) in three of the accessions. Detailed analysis of stress mediated regulation of these genes would help understand the molecular mechanisms of stress induced root growth in wheat.

PA-14

Role of superoxide content in determination of seed vigour of Indian quality mustard genotypes

Nipa Biswas[1], Sangita Yadav[1], Navinder Saini[2], Shiv K. Yadav[1], Anil Dahuja[3], Sujata Vasudev[2] and D.K. Yadava[1]

[1]Division of Seed Science and Technology, [2]Division of Genetics, [3]Division of Biochemistry, ICAR-Indian Agricultural Research Institute, New Delhi-110 012

Email: sangitaydv@gmail.com

Reactive oxygen species (ROS) are natural products of metabolism. They originate from either the incomplete or partial reduction of oxygen, which ultimately leads to the formation of superoxide anion and hydrogen peroxide. Superoxide is the first most ROS which give rise to other ROS molecules. The involvement of superoxide anion to regulate cell proliferation and differentiation during growth of radicals has been documented in literature by many workers. The changes in content of superoxide anion with advance of germination was studied and the possible role of superoxide anion in increasing radicle length during the germination was explored which could substantiate the vigour differences in various Indian quality mustard genotypes. Among the different times of germination taken for observation, significant increase in radical length was observed at 36 hours. At this time of germination, the single zero and conventional genotypes of Indian mustard were having higher content of superoxide anion when radicle length was more than 2 mm whereas in case of double zero type of Indian mustard both radicle length and superoxide content was significantly lower. The result also showed a positive correlation between superoxide anion and radicle length which in turn was positively correlated with seed vigour index-I. Therefore, it could be concluded that lower seed vigour in double zero genotypes is due to the lower content of superoxide content resulting in nonsignificant increase in radicle length than the conventional and single zero genotypes of Indian mustard.

PA-15

Cloning and overexpression of an annexin gene in *Solanum tuberosum*

Ankita Sharma[1], Anupama Singh[1], Hemant B. Kardile[2], Nirmal Kant Sharma[2], A.K. Nath[1] and Vinay Bhardwaj[2]

[1]Dr. Y.S. Parmar University of Horticulture and Forestry, Nauni, Solan, H.P.
[2]Central Potato Research Institute, Shimla-171001, HP
Email: ankitasharma8777@gmail.com

Annexins are calcium dependent, membrane-associated proteins involved in wide range of cellular and developmental processes. The role of these protein families have been investigated in several crop species in relation to many biotic and abiotic stresses. Among the abiotic stresses heat stress is very common and many crop species are prone to the severe effects of heat stress leading to the significant yield loss. Heat stress leads to adverse effect on Potato cultivation affecting the tuberization and the bulking process. The tuberization signal travels from the leaf to underground stems and process involves many interconnected metabolic pathways. Being a signaling molecule, it is hypothesized that annexins might play important role in tuberization under heat stress condition. Keeping this in view an annexin gene with potential role in abiotic stress tolerance was cloned from potato heat tolerant cultivar Kufri Surya. For this appropriate primers were designed based on the available sequence from PGSC database. Using cDNA from Kufri Surya, fragment of approximately 950bp was cloned in TA vector and sequenced. The sequence of the cloned fragment was found to be 99% similar to that of potato annexin p34. This was subcloned in binary vector PRI101. The binary vector PRI101 containing the annexin 1 gene construct was mobilized in to Agrobacterium and was used to transform heat sensitive variety Kufri Chandramukhi. Transformed shoots were selected on medium supplemented with Kanamycin and gene integration was confirmed by PCR. Physiological validation of transgenic plants for heat stress is under progress.

PA-16

Overexpression of wheat heat shock transcription factor (HSFA2h) in *Arabidopsis* confers tolerance to high temperature stress

Suneha Goswami[1], Ranjeet R. Kumar[1], Shahnoor Alam[1], Jyoti P. Singh[1], Sumedha Ahuja[1], Ansheef Ali[1], Bhargav D.V.[1], Abhishek Chitranshi[1], Himanshu Pathak[3], Viswanathan Chinnusamy[2] and Shelly Praveen[1]

[1]Division of Biochemistry, [2]Division of Plant Physiology,
ICAR-Indian Agricultural Research Institute, New Delhi-110012
[3]Central Rice Research Institute, Cuttack, Orissa
Email: suneha08@gmail.com; ranjeetranjaniari@gmail.com

Heat shock transcription factors play a key role in thermotolerance acclimation. Here we identified and cloned a putative HSF gene (HSFA2h) from wheat *cv.*, HD2985 using *de novo* transcriptomic approach. Sanger's sequencing showed the presence of 1218 nucleotide. Full length sequence was submitted to NCBI GenBank with accession no. KP257297.1. Transcript profiling of HSFA2h in endospermic tissues of wheat was observed maximum, followed by leaf and stem. Bioinformatics analysis showed small HSPs such as HSP18.2 as the probable target of cloned HSFA2h transcription factor. The HSFA2h and its target gene transcripts was observed more in wheat *cv.*, HD2985 (thermotolerant), as compare to HD2329 (thermosusceptible). We observed positive correlation between the expression of HSFA2h and sHSP17 under HS. The HSFA2h-pRI101 binary construct was mobilized in Arabidopsis, screening of T2 plants showed better tolerance at 38°C, as compare to control. The expression of HSFA2h was found more than 2.9 fold higher in transgenic Arabidopsis line in response to heat stress (HS) treatment as compared to control. All the biochemical parameters associated with thermotolerance was observed high in HS treated transgenic Arabidopsis plant as compared to wild plants.

PA-17

Abiotic stress induced PUB E3 ligase interacts with co-expressed UBC-E2 in *Oryza sativa*

Taru Aggarwal, Tapan Kumar Mondal and Harmeet Kaur

ICAR-NRC on Plant Biotechnology, Pusa Campus, New Delhi-110012

Email: harmeet.nrcpb@gmail.com

Ubiquitination is a major post translational modification pathway which involves three different types of catalytic enzymes i.e. ubiquitin-activating enzyme (E1), ubiquitin-conjugating enzyme (UBC-E2), and ubiquitin ligase (E3). E3 Ubiquitin ligase, either itself or together with E2 determine substrate specificity in the ubiquitination system. U-box proteins are the most recent class of E3 ligases and in plants these are referred as Plant U-Box (PUB) proteins. According to publicly available microarray dataset analysis, several PUB genes present in *Oryza sativa* display a differential response towards several abiotic stresses. Similarly, we identified a PUB protein (LOC_Os10g40490) upregulated in various abiotic stresses in *Oryza sativa*. Interestingly, this PUB gene was found to be significantly co-expressed with another ubiquitination pathway gene which codes for a ubiquitin conjugating enzyme, E2 (LOC_Os10g39120). Therefore, to elucidate the possible interaction between this PUB ligase and E2, we performed protein-protein docking studies using ClusPro and Haddock. For this, we first retrieved the amino acid sequence for both the genes from Uniprot. These sequences were utilized to compute 3D-protein structure using MODWEB. The sequence-structure compatibility of the models was evaluated on Z-score using the SAVES v5.0 web server. The best models were selected on the basis of overall assessment. These models were further used for docking studies. ClusPro analysis revealed that bonding between PUB67 and UBC-E2 may be balanced by hydrophobic or electrostatic interactions. Haddock analysis supported ClusPro results for predicted active and passive sites using cport (an algorithm for the prediction of protein-protein interface residues). These results strongly suggest that PUB67 and the identified E2 may be co-expressed and interact with each other, which can be further validated experimentally.

PA-18

Analysis of carbon dynamics under elevated carbon dioxide and high night temperature

Rajeev Nayan Bahuguna, Ashish K. Chaturvedi, Saurabh Karwa and Madan Pal

Division of Plant Physiology, ICAR-Indian Agricultural Research Institute, New Delhi-110012

Email: madanpal@yahoo.com

IPCC (2013) report documents evidences of increasing carbon-dioxide concentrations (e[CO_2]) and other greenhouse gases leading to occurrence of extreme climate events such as heat waves and drought events. (e[CO_2]) and temperature are two major factors of climate change. While high night temperature (HNT) is documented to adversely affect vegetative growth, reproductive success and grain filling in rice, e[CO_2] could help in increasing growth and productivity by providing more substrate to photosynthesis. Although, e[CO_2] has been reported to mitigate high day temperature effect on yield in rice, a systemic approach is required to investigate response of rice genotypes under HNT-e[CO_2] interaction. In earlier study we phenotyped 230 rice accessions for high CO_2 responsiveness using low planting genometry technique as a surrogate method and identified twenty three genotypes and validated them by using free air CO_2 enrichment facility in the field for two consecutive years. A contrasting set of rice genotypes was exposed to e[CO_2]-HNT interaction at reproductive and grain filling stage. Elevated [CO_2] has significantly increased leaf chlorophyll (17%-38%), photosynthesis (8% to 47%), biomass (22% to 68%), productive tillers (26% to 59%) and grain weight (11% to 39%). Conversely, high night temperature decreased photosynthesis up to 32% in sensitive genotypes and increased night respiration (17% to 72%) at reproductive and grain filling stage. Genotypes showing higher phenotypic plasticity under e[CO_2], recorded lower tissue respiration and better carbon balance at grain filling stage. Higher biomass and grain weight was maintained by e[CO_2] responsive genotypes when tested under e[CO_2]-HNT interaction due to improved sink strength higher photosynthesis. This study provides important information about e[CO_2] impact on carbon balance dynamics under HNT which could help in identifying novel routes to develop rice cultivars able to maintain higher yield under HNT by efficient utilization of elevated e[CO_2] and other recourses.

PA-19

Identification of the candidate gene(s) for grain yield under drought by fine mapping of a major QTL on wheat chromosome 7B and its transfer into popular high yielding varieties of wheat

Sonia Goel[1], Archana Chaudhary[1], Leena Bhardwaj[1] and Nagendra Kumar Singh[2]

[1]SGT (Shree Guru Gobind Singh Tricentenary) University, Gurugram, Haryana
[2]ICAR-NRC on Plant Biotechnology, Pusa Campus, New Delhi-11012
Email: soniabansal2609@gmail.com

Water deficit is a major environmental factor limiting wheat productivity around the world. Moreover, drought conditions are expected to worsen because of diminishing irigation water availability and foreseen global climate change. Wheat is grown under rain-fed conditions in the central and peninsular regions of India, which accounts for 30% of the total production with little access to irrigation water. A recombinant inbred line (RIL) mapping population comprising of 206 RILs was developed from Wheat cultivars WL711 and C306 thought to be drought susceptible and tolerant, respectively in an earlier study involving *Triticum aestivum, T. durum* and *Triticale* varieties. Grain yield and its componenet traits were evaluated for the WL711/C306 RIL population under irrigated (IRR) and water deficit stress (WDS) treatment during grain filling stage under field conditions at three locations and five seasons (Kadam *et al.,* 2012, Shukla *et al.,* 2015). A novel consitent QTL for grain yield under water deficit stress was mapped in a novel genomic region, on chromosomes 7BL, *qGYWD.7B.1*. This region on chromosome 7BL is very important for influencing grain yield under water deficit stress in wheat. F1 population have been developed by intercrossing contrasting RILs and 526 F1s were developed to further produce 2,836 F2 lines. DNA have been isolated from the F2 plants to find the recombinants for the QTL region. A new range of 35 markers have been developed from the QTL flanking region to narrow down the region. Now further plan is to fine map and introgress the QTL into popular high yielding variety HD2967.

PA-20

Genome wide association analysis for submergence tolerance in *Oryza rufipogon* species complex

Balwant Singh, Kabita Tripathy, Vandna Rai and N.K Singh

ICAR-NRC on Plant Biotechnology, Pusa Campus, New Delhi-110012

Email: nksingh4@gmail.com

Submergence is one of the major abiotic stress which affects millions of hectares of rice cultivation across Asia. Rice plants die within a week of submergence due to lack of oxygen and light. A major QTL for submergence tolerance was first reported in FR13A an 'Aus' type Indian rice cultivar, that provides tolerance for up to two weeks of complete submergence. In this study, we aimed to identify novel sources of flooding tolerance among *Oryza rufipogon* species complex (*ORSC)* Indian wild rice accessions. In order to perform genome wide association studies, accessions were phenotyped at NRCPB phenotyping facility, New Delhi in three independent years. Total 222 accessions were screened by submergence for 18 days. Genotyping of the accessions was conducted using 50K SNP chip (OsSNPnks). A compressed mixed liner model (MLM) GWAS analysis revealed 10 SNPs significantly associated with submergence tolerance ($P < 9.5747 \times 10^{-4}$), with percent variation explained ranging from 7.6% to 11.6%. Of the ten associated SNPs, two each were on chromosomes 1, 2 and 6; one each on chromosomes 3, 4, 5 and 12. None of the SNP was closed to the known Sub1 QTL on chromosome 9 suggesting novel sources of flooding tolerance in the wild rice accessions. The analysis identified two genes involved in glucose metabolism and oxidative stress tolerance associated with the submergence tolerance trait in the wild rice accessions.

PA-21

ABA priming enhances plant architecture, seed development and shows transgenerational response under high temperature stress in Indian bread wheat *(Triticum aestivum* L.)

Praful Jaiswal and Sharmistha Barthakur

ICAR-NRC on Plant Biotechnology, Pusa Campus, New Delhi-110012

Email: jaiswalpraful1987@gmail.com; sharmistha.barthakur@icar.gov.in

Terminal heat stress is a major stressor of wheat and combating this has become crucial to fulfill the demand of agricultural production, productivity and feed the world in coming decades. Priming with various chemicals had been used over the years to improve germination, early seedling establishment, stress tolerance and ultimately enhance the yield. Our lab has been analyzing various phytohormone priming along with molecular analyses of unfolded protein response (UPS) involved in plant hormone signaling as well as stress response. ABA regulates various physiological processes and provides adaptation to many abiotic stresses and unfavorable environment. The SKP1 proteins are subunits of the SCF complex E3 ligases an essential component of UPS, modulating protein turnover. Here, seeds of popular wheat variety HD2967 were treated with 50 ppm of ABA for specific period and exposed to heat stress during seed germination as well as during anthesis stages in a customized heat trap in field conditions in two consecutive seasons. Various morphological analyses pertaining to plant architecture and phenotyping through physiological and biochemical parameters were carried out in the primed plants along with regular ambient grown plants. Results have shown that chlorophyll content and several plant architectural traits were enhanced under priming along with positive effects on grain architecture and 1000-grain weight. Differential modulation of Skp1 transcripts was observed in treated and control plants when analyzed in flag leaf and post anthesis developing caryopses. The potential beneficial effect of ABA priming and functional elucidation is being further investigated.

PA-22

RNAseq analysis to identify the heat stress responsive genes from *Z. nummularia*

Kishor Panzade, Alim Junaid, Harinder Vishwakarma and Jasdeep Chatrath Padaria

ICAR-NRC on Plant Biotechnology, Pusa Campus, New Delhi-110012

Email: jasdeep_kaur64@yahoo.co.in

Ziziphus nummularia (Burm.f.) Wight & Arn., a perennial fruit crop of arid and a semi arid region, indigenous to Indian subcontinent is highly tolerant to heat stress. Therefore it is a rich genomic resource for understanding the molecular mechanism of heat stress tolerance. Unfortunately, limited studies have targeted this highly neglected crop for studies on heat stress tolerance. In this study, RNA-sequencing was carried out for one month old seedlings of *Z. nummularia* accessions (heat tolerant -National Identity ICO598427 and heat sensitive- CIAH-Godhra) after subjecting to heat stress at temperature of 42°C for 2 h. Total RNA was isolated from collected leaf samples and RNA sequencing was performed using Illumina HiSeq 4000 (paired-end sequencing with 250x2bp). A total of 18,00,43,656 raw sequence reads were generated, and upon adapter trimming- quality filtration 13,95,70,909 reads were assembled *de novo* to 1,64,177 unigenes. Further characterization of unigenes indicated that 62.76% had significant hits in the protein database. Kyoto encyclopedia of genes and genomes database (KEGG) identified many metabolic pathways from the obtained unigenes. Based upon Log2 fold change, 10 genes were validated for differential expression under heat stress by qRT-PCR. Of these, the most promising gene (*Zn*ClpB1) was cloned and characterized. Functional validation of *ZnClbp1* in *Nicotiana tabacum* for heat stress tolerance is underway.

PA-23

Understanding the molecular mechanism for high temperature stress response in pearl millet

Albert Maibam[1,2], Jaskirat Singh[1] and Jasdeep Chatrath Padaria[1]

[1]ICAR-NRC on Plant Biotechnology, Pusa Campus, New Delhi-110012

[2]ICAR-Indian Agricultural Research Institute, New Delhi-110012

Pearl millet is a widely grown crop in Africa and Indian subcontinent where high temperature and water scarcity are prevalent. However, there are few studies on response to high temperature in pearl millet, and our comprehension is further limited by in limited database. Two contrasting genotypes of pearl millet {841-B (high temperature tolerant) and PPMI-69 (high temperature sensitive)} were subjected to a temperature of 42°C for 6 hours at flowering stage. Flag leaves from the plants subjected to high temperature stress as well as those not subjected to heat stress were used as sample for physiological and biochemical analysis to assess the plant stress level. Further, the flag leaves were used for comprehensive transcriptome profile by RNA-Sequencing on Hi-seq-2500 platform to identify and characterize ESTs that show differential expression in response to high temperature stress. The identified ESTs/genes are validated by qRT-PCR. Two genes (Pg GST, and Pg_ZnF_UCP) that showed distinct differential expression in response to high temperature stress are cloned and characterized. These genes are being functionally validated in *Arabidopsis thaliana* for their role in imparting thermotolerance.

PA-24

Overexpression of abscisic acid receptor *OsABAR11* enhances cold and drought tolerance in rice

Shashank K. Yadav, Santosh Kumar VV, Pragya Yadav, Rakesh Verma and Viswanathan Chinnusamy

Division of Plant Physiology, ICAR-Indian Agricultural Research Institute, New Delhi-110012
Email: viswanathan@iari.res.in

Abiotic stresses adversely affect rice growth and development. Abscisic acid (ABA), the plant stress hormone**,** plays versatile functions in regulating many developmental processes and adaptive stress responses of plants. Upon perception of ABA by ABA receptors (ABARs), a series of signal cascade is initiated which regulates the protein activity and transcription. Rice genome encodes at least 10 potentially functional ABA receptors. The molecular functions of these receptors in development and stress responses are being studied intensively world-wide. This study was conducted to elucidate the role of *OsABAR11* gene in rice. *OsABAR11* was overexpressed from a stress responsive RD29A promoter in rice cv. MTU1010. Transgenic lines overexpressing *OsABAR11* (T2) and non-transgenic (NT) lines were subjected to low temperature (12°C) and moisture-deficit stress. Physiological and biochemical analysis showed that *OsABAR11* confers higher survival rate under these stresses compared to NT plants. Transgenic plants showed higher Membrane stability index (MSI), Chlorophyll content, Lower ROS accumulation and elevated expression of ABA responsive genes. These results showed that *OsABAR11* is a key component of abiotic stress tolerance in rice, and is useful in engineering abiotic stress tolerance in rice.

PA-25

RNA guided genome editing of Indica rice mega variety MTU1010 using CRISPR-Cas9 system

Santosh Kumar VV and Viswanathan Chinnusamy

Division of Plant Physiology, ICAR-Indian Agricultural Research Institute, New Delhi

Email: viswanathan@iari.res.in

CRISPR-Cas9 based genome editing is precise mutation strategy for quicker improvement of crops with enhanced quality, yield and climate resilience. Hence, standardization of CRISPR-Cas9 system for genome editing of *Indica* rice mega varieties is important to utilize this method for correction of genetic errors to further improve yield, quality, resource use efficiency, and biotic and abiotic stress tolerance. Towards this goal, rice mega variety of MTU1010 was selected. Since rice crops uses more than 50% of the water used in Indian agriculture, water saving in cultivation of mega rice varieties is critical for sustainable rice production in India. To improve the water use efficiency and drought tolerance, *DROUGHT AND SALT TOLERANCE* (*DST*) gene was selected for genome editing. To edit *OsDST* gene, SgRNAs were designed using CRISPR-PLANT (https://www.genome.arizona.edu/crispr2/). Two SgRNAs were designed to mutate *DST* at two different target sites. These SgRNAs were cloned individually in pSgR-Cas9-Os vector and then pyramided in pCAMBIA1300 vector. Mature embryogenic calli derived from MTU1010 was transformed with *Agrobacterium* mediated genetic transformation. Hygromycin resistant T0 transgenic lines were confirmed by PCR using Cas9 specific primers. The target region of *DST* was PCR amplified from T0 plants, sequenced and sequences with degenerate chromatograms were analyzed by using DSDecode (http://dsdecode.scgene.com/). T0 lines with single base substitutions, one to two base pairs deletions and heterozygous and biallelic mutations of DST were obtained. Homozygous mutants were obtained in T1 generation which will be further characterized phenotyped for drought tolerance.

PA-26

Waterlogging tolerance in black gram [(*Vigna mungo* (L.) Hepper] is associated with chlorophyll content and membrane integrity

Ruchi Bansal, KuldeepTripathi, Gayacharan and Ashok Kumar

ICAR-National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110012

Email: ruchi.bansal@icar.gov.in

Black gram is waterlogging sensitive legume crop, which is sown during kharif season. During its life cycle, crop frequently faces waterlogging, which may result due to excess rainfall, poorly leveled land or poor drainage. We studied the effect of waterlogging stress on membrane stability index (MSI), lipid peroxidation, superoxide dismutase (SOD) activity, chlorophyll content and chlorophyll fluorescence in four black gram genotypes namely (Uttara, T-44 IC530491, IC519330). Genotypes IC530491 and IC519330 were found waterlogging tolerant in preliminary screening of 350 black gram lines in a pot experiment. In the present study, stress was imposed for 10 days at vegetative stage (30 days after sowing). Thereafter, excess water was drained to allow recovery in stressed plants. Waterlogging treatment significantly increased lipid peroxidation and SOD activity in all the genotypes, which showed the oxidative injury posed by stress conditions. Chlorophyll content and fluorescence reduced under stress conditions. SOD activity, MSI and chlorophyll content was more in IC530491 and IC519330, T44 as compared to Uttara. Lipid peroxidation was high in Uttara. Though chlorophyll fluorescence reduced in all the genotypes under waterlogging, genotypic differences were non-significant. More efficient antioxidative scavenging and chlorophyll content in black gram was found to be associated with tolerance to waterlogging.

PA-27

Genome-wide analysis, characterization and differential expression of Chitinase gene family in *Brassicae juncea* and *Camelina sativa*

Zahoor Ahmad Mir[1], Sajad Ali[2], Prashant Yadav[1], Apekshita Singh[3], Anita Grover[1]

[1]ICAR-NRC on Plant Biotechnology, Pusa Campus, New Delhi-110012
[2]Centre of Research for Development, University of Kashmir, Srinagar
[3]Amity Institute of Biotechnology, Amity University Noida
Email: anitagrover@hotmail.com

Chitinase gene family is widely distributed across diverse biological systems and play's important role in plant defense against vast group of microorganisms especially fungi. Chitinases hydrolyze chitin which is present in the main component of cell wall of various groups of fungi. However, little is known about the chitinase gene family in *Brassica juncea* and *Camelina sativa* an important oilseed crop. In the present study, we identified chitinase genes in *B. juncea* and *C. sativa* by genome-wide investigation, their sequence alignment and expression analysis by RT PCR (Real-time PCR). Phylogenetic analysis of *B. juncea* and *C. sativa* chitinase genes with known chitinases of *Arabidopsis thaliana* revealed five groups of chitinase genes in *B. juncea* and *C. sativa* that were distributed on all the chromosomes. Amongst all the five groups of chitinases were classified on the basis of protein domain containing glyco-hydro 18 and glyco_hydro 19. The number of chitinase genes in *C. sativa* was greater than *B. juncea* and *A. thaliana*. In the present study, we identified Chitinase genes in *Brassica juncea* and *Camelina sativa* by genome-wide analysis, their structure, function and expression pattern by pangenome exploration.

PA-28

Improving groundnut for foliar disease and oleic acid content using SSR and SNP markers in groundnut (*Arachis Hypogea* L.)

Sushma Tiwari, M.K. Tripathi, Anushree Pramanik, Sushmita Mandloi, Vinod Kumar Sahu, Punamchand Bhawar, Tinee Adlak, Ekta Joshi and R.S. Sikarwar

Department of Plant Molecular Biology and Biotechnology, College of Agriculture, RVSKVV, Gwalior-474006, M.P.

Email: sushma2540@gmail.com

Cultivable groundnut (*Arachis hypogea* L.), is an important oilseed crop with an allotetraploid genome (AABB, 2n=4x=40), which is being cultivated and practiced widely. In current study, screening for foliar diseases and high oleic acid content was done in 125 groundnut set using 12 yield related morpho-physiological traits, Simple Sequence Repeat (SSR) markers and allele specific markers. In this study genetic distance for morphological traits was measured by NTSYS2.0 to show diversity. Gene based SSR markers were used for screening of oleic acid and two foliar fungal diseases i.e., rust and late leaf spot of groundnut. Other than SSR markers SNP genotyping using10 plex SNP markers for screening of rust and LLS was also done. Total 35 germplasms were identified having foliar disease resistance and 42 germplasms have been identified having high oleic acid content using gene based markers. To find out linked marker for these two traits 125 highly polymorphic markers were used for screening of these lines. They represented genetic diversity in the range of 52% to 83% and polymorphic information content (PIC) ranging from 0.46 to 0.81 with mean value of 0.42, indicating high genetic diversity in the studied plant material. Analysis of molecular variance (AMOVA) represented 8% variation among various populations, 30% variation within individuals and 62% variation among individual based on allelic variation SSR markers. Principle Co-ordinate Analysis (PCoA) based on origin formed three major population groups. The genetic analysis determined by STRUCTURE divided all the germplasm lines into 10 populations which were similar to the dendrogram based on genetic distance and the clustering of principal component analysis. The clustering pattern shows diverse geographic origin with a few admixtures. The results could be used by breeding programs to assess the genetic diversity and broaden the genetic base using morphological and molecular marker approaches.

PA-29

Biochemical composition of essential oils from common north-western Himalayan weeds and variation in their fungitoxicity against pathogenic fungi

Rajan Katoch[1], Neelam Thakur[1] and Ankur Tripathi[2]

[1]Biochemistry Laboratory, Department of Crop Improvement, CSKHPKV, Palampur-176062, HP

[2]Department of Zoology, PAU, Ludhiana

Email: rajankatoch@yahoo.com

Essential oils, the complex mixture of volatile phytochemicals, have been investigated from a variety of plants including weeds to be used as antifungal agent. Fungal pathogens are major threats to many food crops worldwide as they cause considerable losses in yield. Traditionally, the application of synthetic agrochemicals is considered as an effective way of controlling fungal diseases; however, due to upsurge in the emergence of resistant strains as well as growing concern for human safety and environment protection, research efforts have been proliferated on exploiting the potential of botanicals in crop protection. *Ageratum conyzoides* L., *Lantana camara* L. and *Chromolaena adenophorum* Spreng. are the most prevalent weeds in North-West Himalayas. The compositional analysis of essential oil from these weeds revealed the dominance of sesquiterpenes. Nerolidol was identified as principle constituent of *Ageratum* (28.84%) and *Lantana* essential oil (22.02%). In *Chromolaena* essential oil, 1-phellandrene (11.73%) was the major constituents. The variations in fungitoxic potential of essential oils from these weeds were studied against six phytopathogenic fungi. *Ageratum* essential oil exhibited highest suppressive effect on the mycelial growth of *Sclerotium rolfsii* (130.52%) followed by *Rhizoctonia solani* (23.57%), *Fusarium* oxysporum (19.18%), *Fusarium solani* (11.11%) at 10% (*v/v*) minimum inhibitory concentration (MIC). The growth of *Alternaria brassicae* (55.97%) and *Phoma medicaginis* (25.82%) was inhibited at 25% MIC. At 10% (*v/v*) MIC, *Chromolaena* essential oil suppressed the growth of *Alternaria brassicae* (17.47%) and *Fusarium oxysporum* (17.36%) while *Rhizoctonia solani*, *Sclerotium rolfsii* and *Phoma medicaginis* were inhibited at 50% (*v/v*) MIC value. *Lantana* essential oil showed fungitoxic activity against *Rhizoctonia solani* (15.08 %) at 25% (*v/v*) MIC. On the basis of present study it was evident that the essential oils of these weeds possessed potent fungitoxic activity and can form an integral part of integrated pest management system. Further, the utilization of these weeds for extraction of bioactive principals could keep their escalating population under control.

PA-30

Antimicrobial activity of *Phaseolus vulgaris* L. cultivar Baspa trypsin inhibitor protein

Yamini Thakur, Ankita Sharma, Subhash Chand Parmar and Amarjit K Nath

Department of Biotechnology, College of Horticulture, University of Horticulture and Forestry, Nauni, Solan-173230, Himachal Pradesh

Email: nathamarjit60@gmail.com

The present study aimed to determine antimicrobial activity associated with partially purified trypsin inhibitor from seed flour of local bean (*Phaseolus vulgaris* L.) cultivar Baspa Antimicrobial activity was tested against plant pathogenic fungal and bacterial strains. Maximum per cent (88.12%) inhibition of growth was observed against fungal strain *Cercospora punicae* in medium supplemented with 300 µg ml^{-1} of inhibitor protein. While 84.20%, 81.00% and 79.41% inhibition in growth was found against *Fusarium oxysporum*, *Colletotricum gloeosporioides* and *Alternaria solani*, respectively. The inhibitor protein was highly effective against bacterial strain *Xanthomonas campestris* and less effective against *Ralstonia solanacearum*. With increase in the amount of inhibitor protein in well, the zone of growth inhibition increased. The zone of growth inhibition was 8, 10, 11, 16 and 21 mm in *Xanthomonas campestris*, 6, 8, 12, 13 and 16 mm in *Agrobacterium tumefaciens* and 4, 8, 16, 17 and 19mm in *Ralstonia solanacearum* in presence of 26, 52, 104, 208 and 416 µg/ml of inhibitor protein loaded into the well. No bacterial growth was observed when clear zone was streaked in control media indicating death of bacterial cells. The studies indicated partially purified inhibitor protein to be effective against plant pathogenic bacterial and fungal strains with varying efficiencies. These inhibitor proteins produced from natural products, emerge as a potential antimicrobial agents and can be applied in agricultural sector for development of transgenic resistant to plant diseases caused by pathogenic bacteria and fungus.

PA-31

ROS, catalase and JA-induced PR genes are involved in resistance of Brassica cenospecies to *Alternaria brassicae*

Prashant Yadav[1,2], Zahoor Ahmed Mir[1], Sajad Ali[1,3], Pradeep K Paplao[4], Anita Grover[1]

[1]ICAR-National Research Centre on Plant Biotechnology, New Delhi-110012
[2]ICAR-Directorate of Rapeseed Mustard Research, Bharatpur, Rajasthan
[3]Centre of Research for Development, University of Kashmir, Jammu and Kashmir
[4]Division of Nematology, Indian Agricultural Research Institute, New Delhi-110012
Email: anitagrover@hotmail.com

Alternaria blight is one of the most devastating diseases of rapeseed-mustard caused by a necrotrophic fungus *Alternaria brassicae*. Resistance to *Alternaria* pathogen has not been reported in the primary gene pool of *Brassica juncea*, however, some members of Brassica cenospecies possess a degree of resistance against *A. brassicae*. Studying the factors behind the spectrum of resistance is essential to develop durable resistance in rapeseed-mustard cultivars. In this study, we selected *Brassica juncea* as susceptible, *Sinapis alba* as moderately tolerant and *Camelina sativa* as a highly resistant source for understanding resistance mechanisms. Histopathological studies revealed that the onset of necrosis is faster in *B. juncea* followed by *S. alba* and *C. sativa* and H_2O_2 production was observed at a very early stages in *C. sativa* while later in *S. alba* and *B. juncea* (though it is prolonged in later two species). Biochemical studies indicated that the changes in catalase and peroxidase activity might be associated with disease resistance but peroxidase does not have much role in providing resistance. The role of important PR proteins including PR-1, PR-2, PR-3 and PR-12 were investigated at the transcriptional level. Gene expression studies revealed that the over expression of salicylic acid responsive genes (PR-1, PR-2) in *B. juncea* leads to susceptibility whereas over expression of jasmonic acid responsive genes (PR-3, PR-12) in *C. sativa* leads to resistance against the pathogen. The histopathological, biochemical, and molecular studies show that *C. sativa* is capable of early detection and early response to the pathogen and shows high degree of resistance compared to *S. alba* and *B. juncea* in response to *A. brassicae*.

PA-32

Quinalphos induced biochemical alterations in the vital organs of female albino rats

Neelam Thakur[1], K.S. Khera[2] and Rajan Katoch[2]

[1]Department of Zoology, PAU, Ludhiana-141004, Punjab
[2]Biochemistry Laboratory, Department of Crop Improvement, CSKHPKV, Palampur-176062, HP
Email: neelampalam@yahoo.com

Crop losses due to pest infestation are one of the major contributing factors for declining agricultural productivity in the world. To curtail the insect pest menace, pesticides are being extensively used by the farmers to bring significant improvement in crop production. In recent past, the pesticide usage has surpassed the tolerance level and resulted in hazardous effects on human health and environment. Quinalphos (an organophosphate) is the most preferred insecticide for protecting agricultural crops from the variety of unwanted pests. The present study was undertaken to study effect of chronic toxicity of quinalphos on biochemical parameters of vital organs of the female albino rats. Two groups of albino rats (T-1 and T-2) were orally intoxicated with quinalphos @ 2 and 4mg/kg bodyweight/day corresponding to 1/50 and 1/25 of the LD_{50} for six weeks period. The study revealed degenerative effects in the vital organs of intoxicated rats which were characterized by altered level of different biomarkers enzymes. Qunialphos intoxication revealed dose dependent increase in the activities of acid phosphatase (30.12% and 35.57%) and alkaline phosphatase (10.15% and 15.39%) in the ovaries of T-1 and T-2 group of rats. Significant decrease in the activity of lactate dehydrogenase (LDH) was also observed in both groups (17.68%, 50.34%, respectively). The degenerative changes in ovary of quinalphos intoxicated rats were also evidenced by reduced levels of proteins, lipids, phospholipids and cholesterol. A significant increase in the secretion of proteins and 17â-estradiol was observed from granulosa cells of treated rats. In the study, the activities of acid phosphatase (27.80%, 65.12%), alkaline phosphatase (20.64%, 36.23%) and lactate dehydrogenase (22.22%, 30.95%) were significantly reduced in T-1 and T-2 group of rats which denotes excessive tissue damage and oxidative stress in liver as a result of quinalphos toxicity. Decreased level of proteins revealed stressed metabolic activities in the liver of quinalphos intoxicated rats. From the study it is evident that exposure to pesticides at higher concentrations have toxic effects on vital organs of the body, therefore, indiscriminative use of quinalphos should be avoided to protect non-target organisms from its obnoxious effects.

PA-33

Host Delivered RNAi of effector genes in *Arabidopsis thaliana* facilitates resistance against root-knot nematode, *Meloidogyne incognita*

Ila Joshi[1,2], Anil Kumar[1], Ashish K. Singh[3], Ashok Chaudhury[2], Anil Sirohi[3], Pradeep K. Jain[1]

[1]ICAR-NRC on Plant Biotechnology, Pusa Campus, New Delhi-110012

[2]Department of Bio and Nano Technology, Guru Jambheshwar University of Science and Technology, Hisar-125001, Haryana

[3]Division of Nematology, ICAR-Indian Agricultural Research Institute, New Delhi

Email: jainpmb@gmail.com

Root-knot nematodes (RKNs) are considered as one of the most menacing sedentary endoparasites. With a broad host range, dynamic distribution and inconspicuous above ground symptoms they amount to crop losses of 173 billion dollar annually to world agriculture. The conventional control strategies include growing resistant cultivars, nematicides and crop rotation, but each with its own limitations. Plant parasitic nematodes have effector genes which facilitate the nematodes to successfully parasitize the host and complete their life cycle. These genes have been successfully targeted for effective nematode resistance via host delivered-RNAi (HD-RNAi). In the present study we targeted two *Meloidogyne incognita* effector genes, namely, *Mi-msp3* and *Mi-msp5*, coding for effector protein expressed in sub-ventral esophageal gland and dorsal esophageal gland, respectively. The differential expression analysis of *Mi-msp3* and *Mi-msp5* was carried out using qRT-PCR in different developmental stages viz., eggs, J2s and adult females. The expression of both the genes was found to be more in adult females suggesting their involvement at adult stage. Three independent *Arabidopsis thaliana* transgenic lines, each expressing the dsRNA for aforementioned genes, were evaluated and reduction in infection (in terms of number of galls) was observed upto 89% and 78% for *Mi-msp3* and *Mi-msp5,* respectively in comparison to control plants. Interestingly, the qRT-PCR analysis revealed nearly 30% and as significant as 86% reduction in mRNA levels of *Mi-msp3* and *Mi-msp5*, respectively, in the females feeding on transgenic plants. Therefore, silencing of both *Mi-msp3* and *Mi-msp5* genes in the nematodes, through HD-RNAi, significantly reduces the nematode infection and highlights its importance.

PA-34

Unravelling the role of microRNAs in chickpea-*Fusarium oxysporum* interaction

Parichita Priyadarshini[1], Deshika Kohli[1], Pradeep K. Jain[1] and Bharadwaj Chellapila[2]

[1]ICAR-NRC on Plant Biotechnology, Pusa campus, New Delhi-110012
[2]Division of Genetics, ICAR-Indian Agricultural Research Institute, New Delhi-110012
Email: jainpmb@gmail.com

Chickpea is the most important pulse crop in India. Its productivity is adversely affected by various environmental stresses. Among biotic stresses, Fusarium wilt caused by the fungus *Fusarium oxysporum* f.sp. *ciceris* is the major concern. Recently, microRNAs have emerged as key regulators in plant defense responses. This study is an attempt to understand the role of microRNAs in chickpea under Fusarium wilt stress. Expression profiling of two microRNAs, miR2118 and miR530 in response to wilt stress in wilt-resistant (WR 315, Pusa Green 112) and wilt-susceptible (BG 256, FLIP 90-166) genotypes was analysed by poly (A)- based quantitative real-time PCR (qRT-PCR). miR2118 was upregulated in wilt-susceptible genotypes and downregulated in wilt-resistant genotypes in response to wilt stress. miR530 expression was opposite to miR2118. TIR-NBS-LRR, a disease resistance gene targeted by miR2118 was significantly downregulated in wilt-susceptible genotypes in response to wilt stress suggesting role of miR2118 in development of susceptibility in genotypes. miR530 targets gene encoding microtubule associated protein. This gene is a negative regulator of plant defense and was downregulated in wilt-resistant genotypes suggesting a role of miR530 in imparting resistance to plants. These findings further encouraged us to analyze the role of TIR-NBS-LRR in plant immunity. Using bioinformatics tools we have co-localized sixteen TIR-NBS-LRR genes of chickpea with five of the previously reported QTLs linked to fusarium wilt resistance indicating their role in plant defense. The findings of this study provide a platform to understand the role of microRNAs and their target genes in wilt stress response in chickpea.

PA-35

Identification of promising chickpea genetic resources for various important diseases

Gayacharan[1], Sarvjeet Singh[2], Upasana Rani[2], Anita Babbar[3], Ashwani K Basandrai[4], Neeta Singh[1], G.P. Dixit[6], J.C. Rana[7], Sushil Pandey[1] and Ashok Kumar[1]

[1]ICAR-National Bureau of Plant Genetic Resources, New Delhi-110 012
[2]Department of Plant Breeding & Genetics, Punjab Agricultural University, Ludhiana
[3]College of Agriculture, JNKVV, Jabalpur-482004, Madhya Pradesh
[4]CSK HPKV, Palampur-176047, H.P. ; [5]CSK HPKV, HAREC, Dhaulakuan-173001, H.P.
[6]ICAR-Indian Institute of Pulses Research, Kanpur-208024, Uttar Pradesh
[7]Bioversity International, India Office, NAASC, New Delhi-110012
Email: gayacharan@icar.gov.in

Chickpea (*Cicer arietinum* L.) is the second most important food legume crop of the world. The crop plays a critical role in providing nutritional security in rural areas particularly in developing countries. The chickpea grain is well known to have high quality protein (17-24 %) and the crop also improves soil fertility by fixing atmospheric nitrogen around 60-150 kg/ha. However, the crop is facing several devastating diseases like *Ascochyta* blight (*Ascochyta rabiei*), Botrytis Grey Mold (*Botrytis cinerea*), *Fusarium* wilt (*Fusarium oxysporum* f. sp. *ciceris*), collar rot (*Sclerotium rolfsii*) and dry root rot (*Rhizoctonia bataticola*) which affects its yield and pose risk of irreversible genetic erosion particularly from niche environment of chickpea cultivation. However, the chickpea genepool also harbors a significant variability which is poorly explored against these diseases. Therefore, the chickpea germplasm screening was done against the above mentioned diseases at their respective hot spot locations. During the last four *Rabi* seasons (2014-15, 2015-16, 2016-17 and 2017-18) total 1450 accessions were screened at the nationally identified hotspot locations *viz*. Dhaulakuan (Himachal Pradesh) and Ludhiana (Punjab) for *Ascochyta* blight, Jabalpur (Madhya Pradesh) for *Fusarium* wilt and collar rot, Durgapura (Rajasthan) for dry root rot. These screenings were done following standard procedures in augmented block design. To avoid any scape in *Ascochyta* blight and Botrytis Grey Mold, artificial inoculation and disease spread procedures were followed. Similarly for wilt and dry root rot, established sick plots were used. This study resulted in identification of some resistant promising accessions for *Ascochyta* blight (disease severity score <3) like IC275447, IC117744, EC267301, ICC4330, ICC3625, ICC4381, IC272461 and IC248147. For *Fusarium* wilt IC267112, IC271922, IC327563, IC486759, IC487359 and IC512061 were found with less than 10% mortality during the *Rabi* 2015-16 and 2016-17. For Botrytis Grey Mold, IC244185, IC251727, ICC6881, IC244267, IC244584, IC251816, EC223490 and IC261223 were identified with severity score less than 3. Similarly promising accessions for collar rot and dry root rot were also identified which may serve as a potential donors in chickpea improvement programs.

PA-36

Gene expression study of potential RNAi targets in *L. erysimi* across different developmental stages

Deepa Bhatt[1], Murali Krishna Koramutla[1], Deepa Dhatwalia[1], Sushma Tamta[2] and Ramcharan Bhattacharya[1]

[1]ICAR-NRC on Plant Biotechnology, Pusa Campus, New Delhi-110012
[2]Department of Biotechnology, Kumaun University, Nainital, Uttarakhand
Email: bhattacharyarc@rediffmail.com

Mustard aphid, *Lipaphis erysimi* is a major insect-pest of Indian mustard. *Lipaphis erysimi* reproduce parthenogenetically and its unusual behavior of reproduction and short generation time makes them major devastating pest of mustard crop. The economic importance of this insect-pest has prompted many fundamental researches related to gene knock-down studies. In an attempt to identify key genes regulating parthenogenetic reproduction in this insect-pest, gene-expression pattern of five developmental genes have been studied across three developmental stages namely, nymph, third instar and adult stage. Interestingly, gene expression study revealed subtle regulation in transcript abundance of these genes in different developmental stages of aphid. Highest Expression of *LePG1, LePG3* and *LePG4* were the most during nymphal stage followed by their expression in third instar and reaching at the lowest expression level in adult. In contrast *LePG2* and *LePG5* showed highest expression in adult followed by the third instar and lowest expression in nymphs. Four of them, were selected for targeted RNAi by feeding dsRNA. dsRNA of the four selected genes were supplemented in diet up to 72h. Interestingly, some of the genes showed significant inhibitory effect on fecundity and mortality within 72h. Furthermore, down regulation of the targeted genes were also validated by qRT-PCR. The artificial bioassay and the expression analysis, results demonstrated *LePG1, LePG2,* and *LePG3* of *L. erysimi* are potential targets for developing host-based RNAi mediated resistance against aphid.

PA-37

An analysis of biochemical traits affecting quality and storage behaviour of quality Indian mustard seeds

Sangita Yadav[1], Sunil Swami[1], Nipa Biswas[1], Shiv K. Yadav[1], Manisha Saini[1], Anil Dahuja[3], Atul Kumar[1], P.B. Singh[4], Navinder Saini[2], Sujata Vasudev[2] and D.K. Yadava[1]

[1]Division of Seed Science and Technology, [2]Division of Genetics, [3]Division of Biochemistry, ICAR-Indian Agricultural Research Institute, New Delhi-110012

[4]IARI, Regional Station, Karnal, Haryana

Email: sangitaydv@gmail.com

Studies were undertaken to investigate the seed quality traits in quality Indian mustard genotypes and understand their association with biochemical composition, seed vigour and storage behaviour. Observations were recorded on seed quality parameters, membrane integrity and biochemical activities related to seed vigour. Imbibitional behaviour showed that that the rate were significantly associated with the seed colour as well as quality of the genotypes. Double zero genotypes (having <2% erucic acid and<30µmole/g defatted meal) imbibed more water followed by single zero genotypes (having <2% erucic acid) and than by conventional which influenced their storage behaviour. Seed quality differences among Indian mustard genotypes could be associated with various biochemical parameters. Significant differences were found in superoxide anion, hydrogen peroxide content, superoxide dismutase (SOD), peroxidase (POD), catalase (CAT), ascorbate and total tocopherol contents among different genotypes of Indian mustard seeds. Studies on incidences of storage fungi during storage showed incidence of storage fungi i.e. *Aspergillus* sp., while insect species involved in mustard seed damage were found to be *Corcyra cephalonica* and *Tribolium* spp. especially in double zero genotypes. Our results support the hypothesis that during ageing there is significant increase in reactive oxygen species (ROS) along with loss of free radical scavenging enzymes (*i.e.*, SOD and CAT) and molecules. This has direct manifestation with the germination potential and vigour of Indian mustard seeds. High quality/vigour seed has efficient antioxidant system either in terms of antioxidant enzymes or antioxidant molecules and low content of ROS.

PA-38

Meta-analysis of QTLs and transcriptome profiling to pinpoint the genes regulating important fruit yield and quality related traits in tomato

Pankaj Singla[1], Geetika[1], Praveen Khatri[1], Shweta Singh[1], Pritam Singh[1], Juhi Chaudhary[2], Humira Sonah[1] and Rupesh Deshmukh[1]

[1]National Agri-Food Biotechnology Institute (NABI), Mohali
[2]College of Agricultural & Life Sciences, University of Florida, Gainesville, Florida, USA
Email: rupesh0deshmukh@gmail.com

Tomato, one of the most important crop worldwide has great significance in the human diet. Tomatoes are the primary dietary source of antioxidants, vitamin C, potassium, folate and vitamin K, which has been linked to many health benefits. Most of these traits are regulated by quantitative trait loci (QTLs) which spans several million bases of a chromosomal region. Longer and inconsistent QTLs make it difficult to use for breeding application as well as for the cloning of regulatory genes. In the present study, information of over a thousand QTLs reported in different studies for fruit related traits was compiled. Chromosomal localization of these QTLs has identified chromosomal hotspots consistently reported in different studies conducted under varying conditions. The QTLs were systematically imposed on consensus genetic map and meta-QTLs were identified. The meta-QTL analysis also estimated a more reliable probability for the association of the loci with the trait and the phenotypic effect governed by the loci. The narrowed intervals of meta-QTLs were considered to perform candidate gene identification. For the candidate gene prioritization, meta-data from the publicly available RNA-seq transcriptome profiling will be used. In addition, tightly linked molecular markers from the meta-QTL region will be developed by exploring SNPs and SSRs predicted with genomic sequence resources. Information provided in the present study will be helpful for map-based cloning of candidate genes and for the development of markers useful for the tomato breeding programs.

PA-39

Atriplex Hortensis an underutilized vegetable of Kashmir valley with potent nutraceutical activity

Omi Laila and Imtiyaz Murtaza

Biochemistry and Molecular Biotechnology Laboratory, Division of Basic Sciences and Humanities, SKUAST-K, Jammu

Email: imz_murtaza@hotmail.com

Atriplex Hortensis (commonly known as Wasta Hakh) is an underutilized vegetable in the Kashmir region. This vegetable is believed to be highly nutritional and quite health beneficial; but the crop is highly unexplored. Therefore, the aim of the current investigation was to profile different *Atriplex Hortensis* ecotypes collected from different districts of Kashmir valley for their nutrients and nutraceutical potential. It was observed that the *Atriplex Hortensis* ecotypes found in Kashmir region are quite rich nutritionally, good source of essential minerals (Ca, Mg, Fe, Zn, Mn, Cu, S, P) as well as phytochemicals (kaemferol, quercetin, dopa, dopamine sitosterol, quercetin). The collected ecotypes further exhibited a much higher phenolic linked antioxidant potential as well as potent antihyperglycemic activities (via inhibiting the key enzymes of carbohydrate metabolism including α- amylase, α-glucosidase and invertase) under *in vitro* conditions. The present study reveals a clear link between nutraceutical content and the *in vitro* antidiabetic potential of *Atriplex Hortensis* ecotypes, however, further preclinical and clinical research is needed before recommending it as a potent antidiabetic nutraceutical food.

PA-40

Nutritional and antidiabetic potential of an underutilized vegetable Nunner (*Portulaca oleracea*) from Kashmir Valley

Imtiyaz Murtaza and Omi Laila

Biochemistry and Molecular Biotechnology Laboratory, Division of Basic Sciences and Humanities, SKUAST-K, Jammu

Email: omilailabiotech2007@gmail.com

Portulaca oleracea (commonly known as Nunner in Kashmir) is an ignored and underutilized vegetable in the Kashmir region. This vegetable though believed to be highly nutritional and quite health beneficial, but the crop has been highly unexplored in Kashmir valley. Therefore, the aim of the current investigation was to profile different *Portulaca oleracea* ecotypes collected from different districts of Kashmir for their nutrients, phytochemicals, and nutraceutical potential. It was observed that the *Portulaca oleracea* ecotypes found in Kashmir region are quite rich nutritionally and a good source of essential minerals (Ca, Se Mg, Fe, Zn, Mn, Cu, S, P), nutraceuticals (Kaemferol, Quercetin, Dopa, Dopamine Sitosterol, Quercetin) and essential amino acids (Arg, Leu, Phe, Lys, Val. Further, these ecotypes exhibit a much higher phenolic linked antioxidant potential as well as potent antihyperglycemic activities (via inhibiting the key enzymes of carbohydrate metabolism including á- amylase, á-glucosidase and invertase) under *in vitro* conditions. The present study reveals a clear link between the presence of nutraceutical content and the *in vitro* antidiabetic potential of *Portulaca oleracea* ecotypes, however, further research is needed before recommending its usage for diabetes management.

PA-41

Seaweed extracts, their importance and uses in fruit cultivation

Rachna Arora and Parampreet Kaur

School of organic farming, PAU, Ludhiana

Email: rachnaarora@pau.edu

Seaweeds are the marine algae found free floating in the marine ecosystem. Their extracts are the biological products of great importance for organic fruit growers because of bio stimulant action and multiple beneficial effects. The extracts contain major nutrient elements like N, P, K, Ca, Mg, S, P and trace elements like B, Zn, Fe, Cu and Mn and the varying concentration of these elements is found in different type of commercial products available in the market. The extracts available in powder as well as liquid form have relevancies as manures and fertilizers in horticulture. Use of seaweed extracts has been evaluated by workers across the globe for physiological effects and plant responses. The extracts diluted to varying concentrations are applied to promote tree growth, flowering, fruit set, yield, quality characteristics and resistance to insect pests and diseases. The uptake of inorganic elements from soil is known to improve with its soil application. Its use has also been found to enhance tolerance of the plants to abiotic stresses such as salinity, temperature extremities and drought. The application of extracts on mango and apple has been reported to reduce the alternate bearing significantly. The literature is therefore reviewed here to understand the significance, methods of preparation, time and method of application, working mechanisms, amount and type of plant growth regulatory substances present and effect on fruit crops. The study may be useful for the researchers to have an idea whether seaweed extracts can be an alternative to the inorganic NPK fertilization while formulating future strategies for organic fruit growers regarding application on individual fruit crops for sustainable chemical free production or for planning integrated nutrition in fruit growing.

PA-42

Role of peroxidase and polyphenol oxidase in pearl millet flour shelf life

Ansheef Ali[1], Suneha Goswami[1], Bhargav DV[1], Abhishek Chitranshi[1], Jyoti P. Singh[1], Shahnoor Alam[1], Sumedha Ahuja[1], P. Padma Priya[1], Ranjeet R. Kumar[1], Sumer P. Singh[2], C. Tara Satyavathi[3] and Shelly Praveen[1]

[1]Division of Biochemistry, [2]Division of Genetics, ICAR-Indian Agricultural Research Institute, New Delhi-110012

[3]All India Co-ordinated Research Project, Jodhpur

Email: suneha08@gmail.com

Pearl millet [*Pennisetum glaucum* (L.)] is an important crop with respect to food and nutritional security because of its sustainability at adverse agro-climatic conditions. India is the largest producer with a production of 8.96 MT (DES2016). Being a nutricereals, there has been stimulation in the demand for pearl millet flour with longer shelf-life and lesser cooking time. Its flour is highly susceptible to oxidation of phenolics and lipid hydrolysis during storage, and hence its flour is prone to rancidity within a few days of storage (17 days). Polyphenol oxidase (PPO) and Peroxidases (POX) are enzymes that are well known to be involved in the enzymatic browning reaction with diverse substrate specificity. We have selected six genotypes of pearl millet (411B, ICMR07333, 86M01, Proagro 9444, HHB197, Pusa 1201). PPO and POX enzyme activity at different time intervals from zero days to ten days interval and upto 30 days was observed. In pearl millet, POX convert C-glycosyl flavone into phenolic aglycone, which is responsible for mousy odour development and PPO leads to browning reaction and change the flour colour to more dark. Yet only limited information is available about activities of these enzymes in pearl millet flour. Here we studied the specific activity of both Peroxidase and Polyphenol oxidase after the flour preparation in different days of interval. We observed a decrease in the specific activity of both these enzymes from fresh to 30 days old flour. POX activity from fresh flour was observed maximum in PUSA 1201 (16.43U/ mg protein) and minimum in HHB197 (11.25U/mg protein) at zero days, and 7.15U/mg (PUSA 1201) and 4.88U/mg protein (HHB197) respectively at thirty days. Similarly, PPO activity was observed maximum in ICMR07333 (6.68 U/mg protein) and minimum in Pro-agro 9444 (3.U/mg protein) at zero days, and 1.35 U/mg and 0.73 U/mg protein respectively at thirty days.

PA-43

Physiological and biochemical changes in guava fruit during low temperature storage

Sushma Yadav, Renu Bhardwaj and Sunil Pareek

Department of Agriculture and Environmental Sciences, National Institute of Food Technology Entrepreneurship and Management, Kundli, Sonepat-131028, Haryana

Guava (*Psidium guajava* L.) is one of the important tropical and subtropical fruit. The short storage life even at low temperatures are serious problem and causes huge losses. Further reduction in temperature can be utilized to extend the shelf life but chilling injury is a problem. Therefore, as a first step changes during CI temperature is find out and based on this downregulation and upregulation of compounds and enzymes a full mechanism can be demonstarted. These changes will be helpful in further studies by utlizing some chemical and physical treatments to alleviate CI and increase the shelf life of this important fruit. The CI symptoms include surface pitting, skin and flesh browning that makes it inedible. Introducing chilling tolerance in guava fruit is one of the mechanisms that can enhance its shelf life at a lower temperature. Studying the changes in the fruit at low-temperature storage is essential to develop chilling tolerance. Therefore, the present study was done in fresh guava fruits at two temperature levels (5°C±1 and 25°C±1) for a 15-day storage. Fruits were sampled at an interval of three days. Various physiochemical parameters like firmness, colour, decay rate, chilling injury index, total phenolic content, reactive oxygen species (ROS) production rate, and antioxidant enzymes activity were studied. The results showed that fruits stored at low temperature have a slow decay rate, firmness loss, weight loss and decrease in ascorbic acid and citric acid content in comparison to ambient temperature fruits. However, CI symptoms are problem at low temperature, which makes unfit for comsumption. The activities of antioxidant enzymes were also higher in cold stored fruits in comparison to ambient temperature fruits. CI induces higher production of ROS that leads to cellular damage and cell death. Firmness loss causes shrinkage in membrane showing surface pitting symptoms and loss in biochemical component decrease flavour and taste of the fruit. These findings throw light on the mechanism on chilling injury in this fruit and pave the way to develop cold stress tolerance in guava fruit.

PA-44

Effect of natural safe rock minerals on nutrient quality parameters of rice *(Oryza sativa)* in rice-wheat cropping system

Santosh Ranva[1,2], Yudhveer Singh[2] and Neelam Jain[1]

[1]Amity Institute of Biotechnology, Amity University Rajasthan, Jaipur
[2]ICAR-Indian Agricultural Research Institute, New Delhi-110012
Email: njain1@jpr.amity.edu

Rice–wheat cropping system has contributed enormously in providing food and nutritional security and livelihood not only in India but around the globe. Rice and Wheat being the world's two most important cereal crops contributes 45% of the digestible energy and 30% of total protein in human diet. Looking into significance of Rice–Wheat cropping system, an alternative nutrient management practice for sustainable crop production was conducted at the research farm of ICAR-Indian Agricultural Research Institute, New Delhi to study the influence of natural safe rock minerals on nutrient parameters of Rice-Wheat cropping system. Treatments involved two rice *(Oryza sativa)* establishment method (aerobic rice and transplanted rice) and crop nutrition levels on Rice-Wheat sequence in randomized block design (RBD) with six treatments replicated thrice. On average transplanted Rice recorded higher results in terms of quality parameters compare to aerobic Rice. Concentration of N, P and K in grain and straw of Rice also increased significantly due to the integrated application of Safe rock mineral (SRM) with or without the application of 250 kg SRM + 100% RDF over SRM alone application. Total NPK uptake showed similar trends as shown in N, P and K uptake in grain and straw. Nutrient management with SRM application also increased protein content significantly. The results demonstrated different nutritional level significantly improved the productivity and nutritional quality of Rice and succeeding Wheat crop in both the years, thus ensuring nutritional security and enhancing nutrient quality.

PA-45

Designing, construction and validation of CRISPR/Cas9 - *GmIPK2* (Inositol polyphosphate 6-/3-/5-kinase) cassettes in soybean

Joshna Jose, Smrutirekha Sahu, Monica Jolly, Veda Krishnan, Vinutha T., Anil Dahuja, Shelly Praveen, Archana Sachdev

Division of Biochemistry, ICAR-Indian Agricultural Research Institute, New Delhi
Email: arcs_bio@yahoo.com

The alarming after math of anti-nutritional attributes and environmental implications of phytic acid has instigated numerous studies aimed at its reduction in soybean seeds. However, the vital contribution of inositol phosphates in the growth, development and stress responses of seedlings makes it essential to strike a balance in the phytic acid levels of soybean seeds such that optimal seedling growth is supported without aggravating nutritional deficiencies in its consumers or promoting troubles in the environment. The present study was designed to carry out CRISPR/ Cas9-mediated targeted genome editing of the *IPK2* (Ins (1,4,5) P3 6/3/5-kinase) gene, which encodes for an enzyme placed at the later part of phytic acid biosynthesis. The work was kick-started by identifying potential targets in the *GmIPK2* gene using relevant web-tools and designing a Cas9/gRNA-*GmIPK2* construct which consisted of a 35S promoter for Cas9 expression, NOS terminator, guide RNA cassette driven by U6 promoter, a kanamycin resistance gene for bacterial selection and bar gene as selectable marker for transgenic plant selection. This construct was transformed into soybean by cotyledonary node method to generate T_0 plants which were analysed for mutation in the *IPK2* gene. Concurrently, a simple and efficient protocol for transformation of soybean leaf discs for transient expression assay, AGRODATE was also developed and standardized on the basis of different parameters contributing to increased transformation efficiency.

PA-46

Lipoxygenase: Fe-containing enzyme showing critical role in rancidity of pearl millet flour compromising the shelf life

Abhishek Chitranshi[1], Suneha Goswami[1], Ansheef Ali[1], Bhargav DV[1], P. Padma Priya[1], Vinutha T[1], Ranjeet R. Kumar[1], Sumer P. Singh[2], C. Tara Satyavathi[3] and Shelly Praveen[1]

[1]Division of Biochemistry, [2]Division of Genetics, ICAR-Indian Agricultural Research Institute, New Delhi-110012

[3]All India Co-ordinated Research Project, Jodhpur

Email: suneha08@gmail.com

Pearl millet (*Pennisetum glaucum*. L) is a C4 cereal crop having genome size of 1.79 GB, belongs to the family Poaceae. It is cross-pollinated crop usually grown under the most adverse agro-climatic conditions. It has excellent nutritional composition with 65% of carbohydrates, 1.2% of fiber, and 11.6% of protein, 5% of fats as well as other micronutrients such as iron (Fe), zinc (Zn), vitamin A and folic acid. The major problem with pearl millet is its flour short shelf life. There are different pathways leads to flour rancidity such as enzymatic rancidity, oxidative rancidity and oxidation of phenolic compound. Lipoxygenase (LOX) (EC 1.13.11.12) is an iron-containing dioxygenase enzyme. Lipoxygenase enzyme plays role in both enzymatic rancidity and also in oxidative rancidity by producing free radicals in the presence of oxygen and light. LOX catalyses the oxidation of polyunsaturated fatty acids and esters into the corresponding hydro peroxides. LOX action lead to loss of nutritional quality due to the destruction of the essential fatty acids like linoleic, linolenic, arachidonic acids as a direct consequence of the reaction and indirect degradation of vitamins and proteins by hydro peroxide and the free radical intermediates; and hence development of off-flavor and loss of color due to the degradation of chlorophyll and carotenes by the hydro peroxide and the free radicals. Lox activity increased by Ca^{2+} and Mn^{2+} but strongly inhibited by Na^{+}, Zn^{2+} and K^{+}. We have taken 6 different varieties of pearl millet namely 411B, ICMR07333, 86M01, Proagro 9444, HHB197, Pusa 1201 to estimate the LOX activity at different days interval after milling pearl millet grain. We have observed maximum LOX specific activity in Pusa 1201 (50.12 U/mg) and lowest in Proagro 9444 (30.85 U/mg) at zero days and further the LOX activity was decreased with respect to 30 days interval of storage in both the genotypes (11.85 U/mg and 8.18 U/mg) respectively.

PA-47

"Oil Type and Time of Addition" - Critical factors controlling starch digestibility

Monika Awana[1], Madhupreet Kaur[2], Veda Krishnan[1], Archana Singh[1], Haritha Bollinedi[3], A.K. Singh[3] and Shelly Praveen[1]

[1]Division of Biochemistry, [3]Division of Genetics, ICAR-Indian Agricultural Research Institute, New Delhi-110012

[2]Department of Chemistry, Delhi University, New Delhi

Email: shellypraveen@hotmail.com

Rice is feeding for over half of the world's population, but due to its high carbaholic nature leading to type II diabetes is creating a starch dilemma and is slowly vanishing from our diet. Nutrition scientists are striving hard to reduce the starch digestibility and often neglect the scientific role behind certain ingenious methods of cooking. Recent studies unraveled the role of Starch–lipid complex formation towards its low digestibility in terms of resistant starch type V, but limiting on how different oil types and time of addition play a role in the processing. This study aimed to assess the starch digestibility of white rice (PB1509) cooked with four oil types: ghee (saturated fat), vegetable oil (unsaturated fat), Coconut oil (medium chain saturated fat) and Rice bran oil (poly unsaturated fat) added at 2 different time points of cooking process ("before" and "during"). Digestibility was affected by these oil types and a maximum decrease (~ 4%) of rapidly digestible starch (RDS) was observed using rice bran oil. Interestingly, maximum increase (~3%) in slowly digestible starch (SDS) fraction was observed using ghee. Study highlights the fact that addition of ghee ("during") showed potential for attenuating the post prandial glycemic response and increasing SDS content. It also suggests that adding fat in cooking process at certain time points may be useful in lowering glycemia. The encouraging results of the present study would be justified by an *in vivo* investigation to determine the effect of type and time of addition of fat altering the starch digestibility and blood glucose homeostasis.

PA-48

Targeted engineering of phytate transporter- MRPABCC5 gene using CRISPR/Cas9 in soybean

Smrutirekha Sahu, Joshna Jose, Veda Krishnan, Monica Jolly, Vinutha T., Anil Dahuja, Shelly Praveen and Archana Sachdev

Division of Biochemistry, ICAR-Indian Agricultural Research Institute, New Delhi
Email: sahusmruti2012@gmail.com

The detrimental effects of phytic acid have sparked an interest in generating low phytate crops. An efficient, targeted approach such as CRISPR/Cas9 technique has addressed the shortfalls with the prior methods and gained wide consumer acceptability. In the present study, we employed this methodology to specifically knockdown the phytate transporter gene *(MRPABCC5)* in soybean. The workflow initiated with 23-nucleotide protospacer selection using Stupar's web-tool considering on and off-target effects, which yielded three potential targets. *In silico* characterisation was carried out using web tools like: RNA fold server, OligoAnalyzer and sgRNA scorer2.0 that predicted the target efficiency on the basis of secondary structures, minimum free energy, GC content, off-target effects. Based on the theoretical and *in silico* analysis, target 2 (MFE: -2.05 kCal/ mol, GC content: 47.82%) was considered as most effective. To validate this, constructs were designed with *Bbs*I overhang targets that were cloned into pBlu/gRNA vector, further verified through restriction with target-specific unique restriction enzymes and sequencing. These guide RNA cassettes containing the desirable targets (gRNA-MRPABCC5) were further cloned into the destination vector: Cas9 MDC123. The confirmation for Cas9/gRNA-MRPABCC5 constructs were carried out by PCR using gRNA cassette primers, target specific unique restriction enzymes and sequencing. *In-vivo* validation could be further carried out by developing transient mutants through AGRODATE and by *Agrobacterium* mediated cotyledonary node method for stable mutants. This approach could set forth the creation of 'transgene-clean' low phytate soybean mutants.

PA-49

Lipase: Culprit behind the low storability of pearl millet flour

Bhargav DV[1], Suneha Goswami[1], Ansheef Ali[1], Abhishek Chitranshi[1], Jyoti P. Singh[1], Shahnoor Alam[1], Sumedha Ahuja[1], P. Padma Priya[1], Ranjeet R. Kumar[1], Sumer P. Singh[2], C. Tara Satyavathi[3] and Shelly Praveen[1]

[1]Division of Biochemistry, [2]Division of Genetics, ICAR-Indian Agricultural Research Institute, New Delhi-110012

[3]All India Co-ordinated Research Project, Jodhpur

Email: shellypraveen@hotmail.com

Pearl millet is a C_4 cereal crop, belongs to the family Poaceae. It is usually grown under the most adverse agro-climatic conditions, where, other crops like rice, wheat fail to produce economic yield. It has an excellent nutritional composition providing 361 kcal/100 g of energy as well as other micronutrients such as iron (Fe), zinc (Zn). Despite nutritionally rich and high medicinal value, the full potential of pearl millet is limited due to lipid oxidation and off-odor during storage of 10-17 days. This is mainly due to the high fat content along with highly active lipases enzyme activity, causing hydrolysis of fats into free fatty acids, is responsible for off-odor development. Due to this reason, pearl millet flour can only be stored for short period of span which is the main drawback for its utilization**.** Hence, it is necessary to control lipase activity to reduce its contribution towards rancidity. To check the lipase enzyme activity and its correlation to off odour development, we have selected six different pearl millet genotypes namely, 411B, ICMR07333, 86M01, Proagro 9444, HHB197, Pusa 1201. Lipase enzyme activity was measured at 0, 10, 20 and 30 days after milling. Specific activity of lipase was observed minimum in 86M01 (5.49 nM/min/mg of protein) at zero day and (14.19 nM/min/mg protein) at 20 days. The lipase activity was observed maximum in pearl millet hybrid HHB197 (12.29 nM/min/mg of protein) at zero day and (30.98 nM/min/mg protein) at 20 days after milling. We observed decrease in the lipase activity after 30 days of milling which may be due to the complete utilization of the substrate. Acid value (rancidity measurement test) was observed maximum in HHB197 and minimum in 86M01; a positive correlation was observed between the lipase enzyme activity and rancidity. There is a need to identify a donor having low lipase activity in the flour in order to use it in the back cross breeding program in order to develop a hybrid with high flour storability.

PA-50

Characterization of isoflavone conjugate hydrolyzing â-glucosidase (ICHG) from soybean and probiotic microbes for improving soy-isoflavone bioavailability

Sandeep Kumar, Minnu Sasi, Monika Awana, Khushboo Kumari, A.P. Rajarani, Anil Dahuja

Division of Biochemistry, ICAR-Indian Agricultural Research Institute, New Delhi-110012
Email: ad_bio@yahoo.com

Soybean consumption is increasingly being viewed as potential solution to combat the intractable problem of protein malnutrition in predominantly vegetarian Indian population as its seeds contain abundant amounts of high quality protein (38%). Besides, soybean contains many phytochemicals which provide numerous additional health benefits to humans. Amongst various phytochemicals, soy-isoflavones are of particular interest due to their ability to prevent cancer, osteoporosis and cardiovascular diseases. These isoflavones are primarily stored as glucose conjugates, which need to be converting into their aglycone (deconjugated) forms by the enzyme β-glucosidase produced by Gut microbiota because these forms are efficiently absorbed and are thus more bioavailable. In the present study we have, therefore, attempted to characterize this enzyme in detail. A complete cDNA sequence of *GmICHG* from *Glycine max* was cloned (1545bp), sequenced and submitted to GenBank (Accession # MH190223). The computational tools like SWISS-MODEL, MEME, RAMPAGE, Phyre and PrDOS were employed to elucidate the various structural attributes of the protein, which helped in understanding its structural and functional characteristics. Its comparison with the reference sequence showed 100% coverage and 99% sequence identity (1540/1545). Conserved domain search revealed a glycosyl hydrolase family 1 N terminal signature sequence (49-63) and active site (416-424) in the translated sequence. In addition, six aerobic probiotic bacterial strains were screened for â-glucosidase activity using para-nitrophenyl- β-D-glucoside as substrate for identification of a catalytically efficient isoform-the *Lactobacillus rhamnosus* was found to be having the highest activity i.e. 8.421 units/mg of protein, which can, thus, be deployed for enhancing the isoflavone bioavailability in soy products.

PA-50

Transcriptome data of cultivated tetraploid and hexaploid wheat variety during grain development

Pranab Kumar Mandal[1], Shubham Rai[1], Megha Kaushik[1], Subodh Kumar Sinha[1], Rajesh Kumar Gupta[1] and Anju Mahendru[2]

[1]ICAR-National Research Centre on Plant Biotechnology, LBS Building, Pusa Campus, New Delhi-110012

[2]Division of Genetics, ICAR-Indian Agriculture Research Institute, New Delhi-110012

Email: pranabkumarmandal@gmail.com

Wheat is a major food crop and an important component of human diet particularly in developing countries. Wheat grain composed of all the nutrients that involves in seed maturation and directly affects wheat yield and quality. In order to identify differentially expressed genes playing vital role during grain development, transcriptional comparison of developing grains was carried out between tetraploid durum wheat (AABB:PDW233) and hexaploid bread wheat (AABBDD:PBW343) using RNA-seq. Developing grain samples were collected at 2, 3, 4 and 5 weeks after anthesis (WAA). RNA was isolated from all the samples and then pooled at equimolar concentration. Approximately 96.56 million raw reads were obtained from both libraries After quality check for read quality and removal of contaminations, a huge proportion (88.2 %), of high quality reads (with Phred Score < 30), in which approximately 76.96% reads of hexaploid wheat and 90.79% reads of tetraploid durum wheat were aligned to reference genome. A total of 2324 up-regulated and 522 down-regulated genes were identified as differentially expressed between PDW233 *vs* PBW343. Gene ontology annotation and enrichment analysis gave further information about differentially expressed genes between durum and bread wheat. This information will help in understanding process grain reserve in tetraploid and hexaploid wheat in relation to their nutritional quality.

PA-52

Identification and characterization of noncoding RNAs (lncRNAs and miRNAs) and their strong interaction with mRNA during flowering in wild relatives of pigeonpea (*Cajanus scarabaeoides*)

Antara Das, Swati Saxena, Kuldeep Kumar, Kishor U. Tribhuwan, N.K. Singh, Kishor Gaikwad
ICAR-National Research Centre Plant Biotechnology, Pusa Campus, New Delhi-110012
Email: kish2012@nrcpb.org

Flower development is a result of crosstalk between many factors like photoperiod, vernalization, hormone concentration, epigenetic modification etc. In the present study, we are reporting the involvement of lncRNAs and miRNAs in flower development of *Cajanus scarabaeoides.* The transcriptome data from floral and leaf tissues were assembled and analyzed for noncoding RNA revealing a total of 1,672 high fidelity lncRNAs and 57 miRNAs being expressed in the leaf and bud tissues during flower development. Target prediction analysis of all the identified lncRNAs showed that 1,593 lncRNAs were targeting 3,420 mRNAs. Among these 98 were transcription factors belonging to 48 groups. All the identified 57 miRNAs belonged to 35 miRNA families and were novel. Prediction of the secondary structure of lncRNAs and miRNAs followed by interaction analysis revealed that 199 lncRNAs could interact with 47 miRNAs. Gene Ontology of lncRNAs, lncRNAs targeted mRNAs and miRNAs targeted mRNAs showed the potential role of lncRNAs and miRNAs in the flower development of *C. scarabaeoides*. Among the identified interactions, 17 lncRNAs have eTMs for flowering-related transcription factors. Expression analysis of identified transcription factor reveals a high level of Csa-lncRNA expression in the bud sequesters Csa-miRNA leading to higher expression of a flower-specific transcription factor. Our work revealed the role of lncRNAs in the regulated expression pattern of genes during flower development and will be helpful and pay a way to other researchers to decipher the role of these non-coding RNAs in other developmental processes.

PA-53

Genome-wide identification of novel miRNAs and their targets regulating galactomannan biosynthesis in clusterbean (*Cyamopsis tetragonoloba*)

Anshika Tyagi[1], Deepti Nigam[1], Amitha Mithra S.V.[1], Amolkumar U. Solanke[1], Nagendra Kumar Singh[1], Tilak Raj Sharma[1,2] and Kishor Gaikwad[1]

[1]ICAR-National Research Centre on Plant Biotechnology, New Delhi-110012
[2]National Agri-Food Biotechnology Institute, Mohali, Punjab
Email: kish2012@nrcpb.org

Clusterbean (*Cyamopsis tetragonoloba*) is an economically important legume crop containing 80% soluble dietary fibre with high protein content and high value gum. Due to the accumulation of 90% galactomannans in the seed endosperm, it can be used as model crop to understand galactomannan biosynthesis and its regulation. In order to understand the role of clusterbean miRNAs during galactomannan biosynthesis regulation, small RNA libraries were prepared and sequenced from five tissues i.e., root, stem, leaf, bud and seed of clusterbean genotype, RGC-936. The means of the libraries fragment size distributions of clusterbean tissue were 149 bp, 158 bp, 154 bp, 148 bpand 156 bp, respectively. A total of 187 known and 171 novel miRNAs were found to be differentially expressed from 116 million raw reads of a total size of 3.17 GB using DSAP and miRanalyzer programs. ManS (mannosyltransferase/mannan synthase) and UGE (UDP- D-glucose 4-epimerase) were annotated as novel unigenes and validated as targets for three novel miRNAs, that is Ct-miR3130, Ct-miR3135 and Ct-miR3157. This is the first comprehensive report on the identification and characterization of miRNAs and their potential targets postulated to act in the galactomannan biosynthesis pathway of clusterbean. The present study could enhance our understanding of the miRNAregulatory tissue-specific metabolic pathways in clusterbean.

PA-54

Extensive expression analyses highlighted role of aquaporins during fruit development and drought in watermelon (*Citrullus lanatus*)

Praveen Khatri[1], Shweta Singh[1], Geetika[1], Pankaj Singla[1], Juhi Chaudhary[2], Humira Sonah[1] and Rupesh Deshmukh[1]

[1]National Agri-Food Biotechnology Institute (NABI), Mohali
[2]College of Agricultural & Life Sciences, University of Florida, Gainesville, Florida, USA
Email: rupesh0deshmukh@gmail.com

Watermelon, a plant with fleshy watery fruit has great adaptability to warm and semi-arid conditions. In this regards, understanding regulation of aquaporins, a water channel proteins, is important particularly during the fruit development and under extreme water regimes. In the present study, genome wide identification and characterization of aquaporin genes in watermelon was performed. Based on homology with known genes, phylogenetic distribution and presence of conserved motifs, a total of 35 watermelon aquaporins were identified and further categorized into five subfamilies as 16 PIPs, eight TIPs, eight NIPs, two SIPs and one XIP. Comparative genomics of watermelon aquaporins with rice, arabidopsis and soybean aquaporins revealed several interesting facts. For instance, unlike other plant species, the entire XIP1subgroup was found to be absent in watermelon. Solute specificity predication of aquaporins was performed based on the evaluation of protein tertiary structure, subcellular localization, conserved motifs and pore morphology. Temporal expression changes analyzed using RNA-seq data revealed higher expression of PIP2 aquaporins only during the early developmental stages. During later stage of fruit rind development majorly NIPs and TIPs were found to be expressed higher than the PIPs. Similarly, PIPs were observed as highly expressed class of aquaporins during drought stress. Comparison made between the drought tolerance (M20) and susceptible (Y34) cultivars, identified several significantly differential expressed aquaporins. A ClaPIP1-1was found to be four-fold upregulated only in tolerant cultivar under drought conditions. Furthermore, co-expression network build for aquaporins showed ClaNIP2-2 and ClaTIP1-3 as the two major regulatory hotspots for the entire aquaporin family. The genome-wide characterization of aquaporins provided in the present study provides insight to elucidate their physiological and developmental roles in watermelon.

PA-55

Understanding the role of aquaporins in developing petal spur through genomics and transcriptomic analysis in *Aquilegia coerulea*

Shweta Singh, Praveen Khatri, Geetika, Pankaj Singla, Rupesh Deshmukh, Humira Sonah

National Agri-Food Biotechnology Institute (NABI), Mohali, Punjab

Email: biohuma@gmail.com

Aquaporins (AQPs) the major water conduit present in plants plays a pivotal role in cellular transport systems by fetching several small solutes across the membrane. In the present study, genome-wide identification and characterization of AQPs were performed in *Aquilegia coerulea* (Colorado blue columbine). Classification and intergeneric phylogenetic analysis with the known aquaporins from rice, Arabidopsis and poplar depicted five subgroups of aquaporins representing nine NOD26-like intrinsic proteins (NIPs), ten tonoplast intrinsic proteins (TIPs), four plasma membrane intrinsic proteins (PIPs), two small basic intrinsic proteins (SIPs) and four uncharacterized intrinsic proteins (XIPs). Loss of entire XIP2s subgroup only in case of *A. coerulea* was one of the interesting key features to note. The protein structure analysis of AQPs revealed the structural variation in pore morphology across the subgroups, as SIPs being more hydrophilic compared to other AQPs. The RNA-seq analysis in petal spur deciphered the elevated expression of TIPs and XIPs in the developing spur suggesting its role in the cell division and elongation. The present study is the first comprehensive analysis of *A. coerulea* providing insights of AQP's evolution, distribution, expression dynamics and their probable role in various physiological processes.

PA-56

RNA editing events in mitochondrial genes: a comparison between male sterile and fertile pigeonpea

Tanvi Kaila[1,2], Swati Saxena[1], G. Ramakrishna[1], Anshika Tyagi[1], Kishor U Tribhuvan[1], Sandhya[1], Ashok Chaudhury[2], Nagendra Kumar Singh[1] and Kishor Gaikwad[1]

[1]ICAR-NRC on Plant Biotechnology, Pusa Campus, New Delhi-110012

[2]Department of Bio & Nanotechnology, Guru Jambheshwar University of Science & Technology, Hisar, Haryana

Email: kish2012@nrcpb.org

RNA editing is a post-transcriptional process which leads to the alteration of the nucleotide sequence of a RNA molecule different from that encoded by the particular gene. The majority of the editing processes involve C to U conversion. Any defect in the RNA editing process may lead to altered proteins and sometimes may lead to development of Cytoplasmic Male Sterile (CMS) phenotype. Although, the editing status of various mitochondrial genomes has been reported,but the editing status of pigeonpea mitochondrial genes is unknown. We have explored the editing status of 20 major mitochondrial transcripts in both male sterile (AKCMS11) and male fertile (AKPR303) pigeonpea. With the help of in silico tools, 167 editing sites were identified in male sterile genotype and 166 in male fertile genotype. The predicted editing sites were validated by mapping RNA-seq reads onto the amplified mitochondrial genes, and 165 and 159 editing sites were validated in male sterile and fertile plant respectively. Among the amino acid alteration, the most frequent one was the conversion of hydrophilic amino acid to hydrophobic, which accounted for 48.7% of conversion in AKCMS11 and 49.3% of conversions in AKPR303. Also the most frequent amino acid change was serine to leucine in both the genotypes. In *atp6*, RNA editing resulted in alteration of a codon coding for glutamine to a stop codon, in both the genotypes. Although differences in editing status of mitochondrial genes were seen among both the genotypes but no significant differences were seen among them, that could account for CMS or restorer phenotype.

PA-57

Identification of genetic and genomic resources for early flowering and maturity traits in Linseed (*L. usitatissimum*)

Ankit Saroha, Vikender Kaur, S. Rajkumar, Rajesh Kumar, J. Aravind, J. Radhamani, Manju, B. L. Meena and Dhammaprakash P. Wankhede

Division of Genomics Resources, ICAR-National Bureau of Plant Genetic Resources, New Delhi-110012
Email: d.wankhede@icar.gov.in

Linseed (Flaxseed) (*Linum usitatissimum* L.) is second most important *Rabi* oil seed crops in India. Linseed is mainly grown for its fiber and seeds oil, however, it has caught interest as nutraceutical as well as functional food ingredient owing to rich contents of α-linolenic acid (omega-3 fatty acid) and lignans. As functional food, flaxseed provides nutrition as well as various health benefits such as reducing risks of cancer, diabetes etc. Average flaxseed productivity in India is substantially lower than the global average. Early maturity is considered an important trait as crop escapes from severity of biotic and abiotic stresses. In this direction, we have evaluated 221 diverse linseed germplasm accessions along with three check varieties for flowering, maturity and yield related traits in augmented block design at NBPGR research farm during 2017-18. Observations were recorded on seventeen qualitative and seven quantitative traits. Multivariate analysis of the variables showed that more than 83% variation is contributed by first five principal components. The study helped identification of germplasm accessions, namely, IC0523807 and IC0525939 as early flowering (days to 95% flowering); IC0525939, IC0525915 and IC0525917 as early maturing compared to the check varieties Kartika, Shekhar and T-397. The germplasm accessions were grouped into very early, early, medium, late and very late categories and are being used for identifying genetic basis of variation in flowering time and maturity in linseed using candidate gene approach.

PA-58

Development of novel chloroplast microsatellite markers in pigeonpea (*Cajanus cajan* L.) and their transferability to wild *Cajanus* species

Swati Saxena[1], Tanvi Kaila[1], Archana Singh[2], N.K. Singh[1] and Kishor Gaikwad[1]

[1]ICAR-National Research Centre on Plant Biotechnology, Pusa Campus, New Delhi-110012
[2]Division of Biochemistry, ICAR-Indian Agricultural Research Institute, New Delhi-110012
Email: kish2012@nrcpb.org

Chloroplast microsatellites (cpSSRs) are important genetic markers useful in population genetics and evolutionary studies in plants. Pigeon pea (*Cajanus cajan* L.) is an important legume crop of the rain-fed agriculture. We report on the development of 39 new chloroplast microsatellite markers by analyzing the pigeonpea chloroplast genome sequence. Sequences containing mononucleotides, dinucleotides, tetranucleotides, and compound repeat motifs were used for designing of cpSSR markers. The newly developed chloroplast microsatellite markers were further evaluated for their cross-species transferability in six wild *Cajanus* relatives. In total, 17 cpSSRs showed polymorphism among the wild *Cajanus* species. The average polymorphism information content (PIC) value detected was 0.40 pointing towards their competence in detecting genetic relatedness among the species in this genus. Two major distinct groups (I and II) were revealed by the neighbor-joining method with all the wild species grouped together except for one *C. platycarpus* that was most diverse and an outlier. These cpSSRs are of great significance to study diversity analysis and phylogenetic relatedness within genus *Cajanus*.

PA-59

Genome-wide analysis of DNA methylation and transcriptional trends in fertile hybrid and inbred lines of pigeonpea

Alim Junaid and Kishor Gaikwad

National Research Centre on Plant Biotechnology, LBS Centre, Pusa campus, New Delhi-110012

Email: kish2012@nrcpb.org

DNA methylation is an important heritable landmark that confers epigenetic changes in hybrids and has fascinated biologists and plant-breeders over the years. Although occurrence of epigenetic changes have been documented in rice and maize hybrids, such investigations have not been reported in Pigeonpea. Here we present genome-wide CG, CHG and CHH methylation profiles of pigeonpea-inbred lines (11A and 303R) and their F1 hybrid at single base resolution. We found that methylation levels in sterile line (11A) were higher than fertile hybrid and fertile parent (303R). Furthermore, identification of differentially methylated regions (DMRs) and analysis of their methylation levels revealed remarkable differences between methylation patterns in sterile parent and fertile hybrid. Identification of differentially methylated re-gions (DMRs) in the sterile and fertile parent revealed remarkable differences between their methylation patterns. Investigation of methylation status of parental DMRs in hybrid revealed non-additive methylation patterns resulting from trans-chromosomal methylation and trans-chromosomal demethylation events. By comparing whole transcriptome data with complete genome bisulfite sequencing data, we identified several DMRs associated with differentially expressed genes that are known to play a role in fertility restoration. Further, inbred parents and F1 hybrid displayed a negative correlation between gene expression and CG methylation. Over all, our results provide an understanding of epigenetic changes and their role in regulating gene expression in Pigeonpea hybrids from genome-wide point of view.

PA-60

Identification of candidate genes for seed yield by meta-QTL analysis in soybean (*Glycine max* L.)

Geetika[1], Praveen Khatri[1], Pankaj Singla[1], Shweta Singh[1], Giriraj Kumawat[2], Rupesh Deshmukh[1] and Humira Sonah[1]

[1]National Agri-Food Biotechnology Institute (NABI), Mohali, Punjab
[2] Indian Institute of Soybean Research, Khandwa Road, Indore-452001, M.P.
Email: biohuma@gmail.com

Soybean is one of the most economically important crop worldwide, grown extensively for seed oil and protein-rich meal. Seed yield improvement is the prime goal for most of the soybean improvement programs. However, seed yield is a complex inherited trait largely affected by environmental conditions and governed by many small effect Quantitative Trait Loci (QTL). The utilization of the QTL for map-based gene cloning and breeding applications is difficult because of inconsistent association, small effect, and larger genomic span. To overcome the limitations, a meta-analysis of a substantial amount of QTL related to seed yield identified over the past decade looks promising. In the present study, 116 QTLs related to the seed yield identified using different populations under diverse climatic conditions were analysed. The set of QTLs was projected on the consensus genetic map of soybean and subjected for meta-QTL analysis. A total of five significant meta-QTLs with larger phenotypic effects were identified. The most significant meta-QTL MqSY6-1 comprising 16 QTLs was identified on chromosome 6. Subsequently, ten candidate genes were identified in the reduced confidence interval of MqSY6-1 using similarity-based function annotation, expression profiling and homology with known genes identified in other crop species. The information of meta-QTL and candidate genes presented here will be useful for the development of functional markers for breeding applications and map-based cloning of seed yield-related genes in soybean.

PA-61

Ectopic expression of Cre- recombianse under different germ-line specific promoters in transgenic Indian mustard (*B. juncea*)

Deepa Dhatwalia[1], Veena Pande[2] and Ramcharan Bhattacharya[1]

[1]ICAR-NRC on Plant Biotechnology, Pusa Campus, New Delhi-110012

[2]Department of Biotechnology, Kumaun University, Nainital, Uttarakhand

Email: bhattacharyarc@rediffmail.com

Site specific recombination (SSR) system *Cre-LoxP*, have been extensively used for removal of selectable marker genes or short spacer sequences from the transgenic plants. For ectopic expression of the Cre-recombinase tissue specific expression is preferred compared to expression driven by constitutive promoter. For conferring appropriate tissue specificity to recombinase expression for the purpose of marker rescue germline promoters will be most suitable. However, it is likely that germline specific promoters from heterologous sources will vary in activity when deployed in Indian mustard. In the present study, gene expression of recombinase under two germline specific promoters TA29 and CLV3 promoter have been assessed in transgenic *B. juncea* lines expressing recombinase enzyme. We have compared specificity and relative promoter strength of these two promoters along with a constitutive promoter CaMV35S in anthers of *B. juncea*. Results and experimental data on ectopic activity of these promoters in *B. juncea* for driving anther specific expression of Cre-recombinase will be presented.

PA-62

Gamma irradiation: An economical technology to manipulate the nutritional quality of wheat grains by modulating starch metabolism pathway

Sumedha Ahuja, Bhargav DV, Ansheef Ali, Abhishek Chitranshi, Jyoti P. Singh, Shahnoor Alam, Suneha Goswami, Ranjeet R. Kumar and Shelly Praveen

Division of Biochemistry, Indian Agricultural Research Institute, New Delhi-110012
Email: ranjeetranjaniari@gmail.com

An experiment was carried out to determine the effect of heat stress and gamma irradiation on starch metabolism of wheat (*Triticum aestivum*). Dry seeds of wheat varieties HD3086 and HD 3118 were subjected to pre-sowing treatment of gamma radiation with doses of 0.20, 0.25, and 0.30 Kilogray (KGy) from a cobalt source (^{60}Co) continuously for three days. The plants germinated from the pre-treated seeds were exposed to HS during pollination and grain-filling stages of growth and development. Plants pre-treated with gamma irradiation showed very high activity of á/â-amylases in endospermic tissue under HS; effect was observed more pronounced in wheat *cv.* HD3086, as compared to HD3118. Similarly, gamma irradiation was observed to stabilize the activities of enzymes associated with starch biosynthesis pathway (AGPase and SSS) under HS. Gamma irradiation was observed to have positive effect on the accumulation of starch and amylose in grains under HS; the quality was observed better in HD3086 compared with HD3118. The present findings provide cheap and user friendly technology to mitigate the effect of terminal HS in wheat without compromising with the quality of the grains.

PA-63

Genome-wide comparative analysis of transposable elements highlight diversity and evolution of legume plants

Pawan Kumar Jayaswal[1,2], Asheesh Shanker[2] and Nagendra Kumar Singh[1]

[1]ICAR-NRC on Plant Biotechnology, Pusa Campus, New Delhi-110012

[2]Banasthali University, Rajasthan-228371

Email: nksingh4@gmail.com

Retrotransposons (Res) or mobile genetic elements in plant and animal sharing some of the characteristic features of retroviruses, have played an important role in the evolution and expansion of genome. Availability of genome sequence of different legume genome has provided an opportunity to characterize the abundance, distribution, evolution and divergence analysis of Class I intact long terminal retrotransposons (LTR-RTs) in seven different legume species. Among eight legume species *Arachis ipaensis (Aip)* showed the highest number of full length intact LTR element (3,325 elements) followed by *Glycine max (Gma, 2,328* elements*), Vigna angularis (Van, 1,625,* elements*), Arachis durensis* (Adu*, 1,348,* elements*), Lotus japonica* (Lja*, 1,294,* elements*), Medicago truncatula* (Mtr*, 788,* elements*)* and *Circer arietinum* (Car, 124 elements). All identified elements were having clearly defined motif and TSDs (Target site duplications) structure and 5′-TG..CA-3′ motif was predominant among seven species except in Lja and Mtr which were also observed 5′-TA..CT-3′ and 5′-TA..GA-3′, 5′-TG..TA-3′ type of motif respectively. Further comprehensive analysis of Gypsy and Copia superfamily of the LTR revealed that the amplification timeframe of intact element was dramatically varies in different families, and the average insertion time of Copia-LTR among seven species was varied from 0.51 Mya (Million years ago, Van sps.) to 1.37 Mya (Adu and Aip) while Mtr Gypsy element was younger (0.22 Mya) among other species Gypsy elements. Phylogenetic analysis of reverse transcriptase (RT) domain containing Copia and Gypsy elements showed the clear speciation of different LTR super-families as well as their respective lineages. The results enabled understanding of full-length intact LTR-REs structure and will be a valuable resource for further study of impact of transposable elements on gene structure and expression in legume species.

PA-64

Comparative analysis of mutagenesis approaches for breeding resource development and functional genomics in tomato

Juhi Chaudhary[1], Alisha[2], Praveen Khatri[2], Shweta Singh[2], Geetika[2], Pankaj Singla[2], Humira Sonah[2] and Rupesh Deshmukh[2]

[1]College of Agricultural & Life Sciences, University of Florida, Gainesville, Florida, USA
[2]National Agri-Food Biotechnology Institute (NABI), Mohali, India
Email: rupesh0deshmukh@gmail.com

In the present study, different mutagenesis approaches are being used to develop a mutant resource in tomato aiming for the improved traits and to facilitate functional genomics. As a part of the study, a systematic review has been made to understand the highly explored approach of mutagenesis in tomato. A database of over a hundred genes characterized using the mutagenesis approaches is compiled and will be hosted on an online database for tomato genomics. The literature survey revealed EMS based mutagenesis as the highly explored option in tomato mutation breeding. In this regards, mutagenesis with other options including fast neutron (FN) mutagenesis, magnetofectionof nanoparticles, and N-Ethyl-N-Nitrosourea (ENU), are being used. Usually FN mutagenesis produces large size chromosomal deletion which provides several advantages over the point mutations usually observed with EMS mutagenesis. For instance, the larger deletions by FN (>1 kb) are easy to detect with next generation sequencing approach. Similarly, combined use of ENU and EMS anticipated to produce more frequent mutations. Here we are also trying to use iron nanoparticles to make mutagenic impact under high pressure of magnetic field. The comparative study being performed here will be helpful to generate diversity, novel phenotypes and lines useful for the functional genomics. The mutant lines will be extensively evaluated for the nutritional quality traits in tomato.

PA-65

Application of plant tissue culture in improvement of horticultural crop plants

M.K. Tripathi, Sushma Tiwari, Nishi Mishra, Sonali Singh and Ashok Ahuja

Department of Plant Molecular Biology and Biotechnology, College of Agriculture, RVSKVV, Gwalior
Email: sushma2540@gmail.com, drmanojtripathi64@gmail.com

Plant cell tissue and organ culture is being used as a tool for rapid multiplication of virus free quality planting material by development of micropropagation protocols in most of the horticultural crop plants. Micropropogation technique is economical in time and space affords greater output and provides viral disease free and elite propagules. Storage and conservation of germplasm may be possible through tissue culture. Micropropagation of almost all of the horticultural crops is possible now. Embryo rescue is proved helpful to rescue distant crosses of vegetable crops. Haploid technology is being used to obtain complete homozygosity of the offspring that helps in phenotype selection for quantitatively and qualitatively inherited characters. Somatic hybridization and cybridization has been very useful in transferring cytoplasmic male sterility for obtaining hybrid vigour through mitochondrial recombination and for genetic transformation. *In vitro* selection makes possible to save the time required for developing biotic and abiotic stress tolerant lines. Mutants obtained from somaclonal variants can be effectively selected for tailoring tolerance/ resistant lines against different stresses. In this presentation, micropropogation protocols of different fruit crops *viz*: grape, aonla, *Citrus spp* and pomegranate; ornamental and flowering plants like :gerbera, amaryllis, tuberose and gladiolus, medicinal and aromatic plants: sarpagandha safed musli, ashwagandha, liquorice, *Aloe barbadensis*, sandalwood and *Plumbago zeylanica* developed in our lab will be discussed Besides production of secondary metabolites production in sarpgandha, chitrak and *Tinospora cordifolia* and will also be presented.

PA-66

GFP tagging based method to analyze the genome editing efficiency of CRISPR/Cas9-gRNAs through transient expression in *N. benthamiana*

Swapnil S. Thakare[1], Vinutha, T[1] Navita Bansal[1], Vanchinathan S[1], Rama Prashat G[2], Veda Krishnan[1], Archana Sachdev[1] and Shelly Praveen[1]

[1]Division of Biochemistry, [2]Division of Genetics, ICAR-Indian Agricultural Research Institute, New Delhi-110012

Email:shellypraveen@hotmail.com

The CRISPR/Cas9 (Clustered regularly interspaced short palindromic repeats/CRISPR – associated proteins 9) is simple and highly efficient technology applied to functional studies of genes and genetic crop improvement. Several approaches are available to detect the genome editing efficiency of gRNAs are either expensive or time and labor consuming. In this study, we have demonstrated the utility of green fluorescent protein (*GFP*) marker to detect the targeting efficiency of gRNAs. As a proof of concept, *Glycine max* De-Etiolated 1 (*GmDET1*) gene was chosen and tagged with GFP to rapidly analyze genome editing efficiency of gRNAs. Results showed weaker GFP fluorescence signal in the *N. benthamiana* leaves co-infiltrated with *GmDET1-GFP* overexpression (OE) + *DET1* gRNA1 constructs as compared to the stronger GFP florescence signal in the leaves co-infiltrated with *DET1* gRNA2 and gRNA3 constructs, indicating the highest and lowest targeting efficiency of *DET1* gRNA1 and *DET1* gRNA2/gRNA3 respectively. These results were further confirmed by the detection of the mutation frequencies through T7 endonuclease (T7E1) assay and sequencing; the highest mutation rate of 38.46% in *GmDET1* targeted by DET1 gRNA1 to that of DET1 gRNA2 (7.69%) and gRNA3 (15.38%) was observed. Thus our studies showed "GFP tagging" as the most reliable and rapid method-one can apply to minimize the generation of non-edited transgenic plants resulting from inefficient gRNAs.

PA-67

Development of an efficient regeneration protocol for *Sterculia urens* Roxb., an endangered tree species of India

Mamta Dhiman, Abhijeet Singh and Madan Mohan Sharma

Department of Biosciences, Manipal University Jaipur, Dehmi Kalan, Near JVK Toll Plaza, Jaipur-Ajmer expressway, Jaipur-303007, Rajasthan

Email: madanmohan.sharma@jaipur.manipal.edu

Sterculia urens Roxb., a deciduous tree species belongs to the family *Sterculiaceae*, is native to India. The plant is commonly known as 'Gum Karaya' and is popular for its high value gum used in food, pharmaceutical, cosmetic, textile, paper *etc.* industries and enormous ethnobotanical importance in tribal populations of India. *Sterculia* gum is one of the important non timber forest products of India. Over the high commercial value and demands, establishment of plants in nature is quite challenging (due to physical and physiological factors) and use of non-scientific tapping for gum extraction further causes plant death. To overcome the low abundance, an efficient regeneration protocol is the prerequisite of mass multiplication using *in-vitro* propagation methods. Mother explant for the study was obtained from *in-vitro* grown seedlings, as the collection of the explants for *in-vitro* micropropagation is one of the challenging tasks. Different explants such as hypocotyl, cotyledonary node, inter-nodal segment, epicotyl, leaf and thin cell layers from saplings were studied for shoot multiplication. Optimized explants were further implemented for maximum shoot induction using different growth regulators (both individually as well as in combinations). Thidiazuron (TDZ) at lower concentrations was most effective in maximum shoot induction as compared to other PGRs. Following successful shoot regeneration, trials for root induction were implemented using IBA, NAA, IAA, and 2,4-D. IBA has resulted in root induction at a concentration range of 4-10 µM with callogenesis. Subsequent trials are in progress for successfull rooting by using adjuvants, two phase root induction, modifications in macro & micronutrient concentrations and their effects on root induction.

PA-68

An efficient *in vitro* regeneration protocol of *M. oleifera* for enhanced biosynthesis of nutraceuticals

Swati Gupta[1], S.L. Kothari[2], Sumita Kachhwaha[3] and Rohit Jain[1]

[1]Department of Biosciences, Manipal University Jaipur, Rajasthan
[2]Amity Institute of Biotechnology, Amity University Rajasthan, Jaipur, Rajasthan
[3]Department of Botany, University of Rajasthan, Jaipur, Rajasthan
Email: rohit.jain@jaipur.manipal.edu

Moringa oleifera Lam., popularly known as sehajan is a mineral packed, nutrient rich medicinal tree species of family Moringaceae. Different parts of this tree have been used as remedy for various diseases such as asthma, cancer, Alzheimer's and AIDS. Apart from being an exceptionally rich source of pharmaceuticals and nutraceuticals, seeds and gum exudates of this tree are used in bioremediation, waste water treatment, drug delivery systems and food industries. These multidimensional utilities of sehajan may cause overexploitation of this tree, posing danger to the existing natural variability in the near future. Therefore, there is a need for conservation of the species for ethnobotanical, pharmacological and nutraceutical purposes. In this study, an efficient *in vitro* regeneration protocol for mass multiplication of *M. oleifera* with enhanced nutraceutical value has been developed. The decoated seeds were pre-treated with 1% (w/v) Bavistin and 0.33% (w/v) streptocycline prior to sterilization with 0.1% (w/v) $HgCl_2$. Decapitation of the seeds induced adventitious shoots ranging from 2-10 on MS medium containing auxins or cytokinins. Culture of nodal segments on MS medium along with BA (3 mg L^{-1}) resulted in maximum shoot proliferation with ~20 shoots per explant. Histological studies revealed direct organogenesis from the nodal explants. Shoots (>3cm) were transferred to rooting media and 100% rooting efficiency was attained with half strength MS medium supplemented with IAA and IBA. All the rooted plants were successfully established in greenhouse for further acclimatization. The protocol thus optimized will aid in mass production of disease-free plants of *M. oleifera*.

PA-69

In vitro regeneration of hybrid *Vigna* variety through embryo rescue technology

Ritupriya Singh and Susmita Shukla

Applied Plant Biotechnology Research Lab, Centre for Plant and Environmental Biotechnology, Amity Institute of Biotechnology, Amity University, Noida (U.P.)

Email: sshukla3@amity.edu

Legumes (Mungbean) belonging to Fabaceae have their significance as human sustenance crops. These pulse crops are utilized because of their capacity to fix nitrogen which positively affects soil fruitfulness and consequent harvest efficiency. In this manner, the generation and utilization of these pulse crops in human dietary regimens could help in the decrease of worldwide global warming, eutrophication, fermentation, and land degradation. Legumes gives noteworthy measures of micronutrients, including iron, zinc, calcium, and nutrients. Mungbean is developed generally in developing nations, which has deferred principal genome explore. Stepwise utilization of primary, secondary and tertiary gene pools of this crop can result in tremendous improvement in yield. However, incompatibility between different *Vigna* species hampers the transfer of desired genes and limits the potential for improving the productivity. There exist multivariate biotechnological techniques for crop improvement one of which is embryo rescue technology which aims at rescuing an early stage embryo and culturing it in-vitro for plantlet regeneration. This strategy helps to beat the obstruction amid and post fertilization which is a cumbersome task to produce hybrids in *Vigna* species. Another elective strategy is protoplast fusion for interspecific hybrid generation which could be used however the achievement rate is impressively low. The production of improved variety is essential so that it could be consumed widely and suffices the nutritional demand. The critical parameters are taken into contemplations to encourage the pod setting during cross hybridization including the time of pollination, bud stage, and parent determination. Our study portrays the varietal improvement in *Vigna* species by conventional breeding technique and actualizing the tissue culture (embryo rescue technology) to effectively raise the interspecific hybrids.

PA-70

Superoxide dismutase: Regulate the antioxidant network in stable mutants of wheat developed for thermotolerance

Raja Husain[1], Mohd. Tasleem[1], Sumedha Hasija[1], Kirti Arora[1], Shahnoor Alam[1], Jyoti P. Singh[1], Suman Bakshi[2], Sanjay Jambhulkar[2], Suneha Goswami[1], Ranjeet R. Kumar[1] and Shelly Praveen[1]

[1]Division of Biochemistry, ICAR-Indian Agricultural Research Institute, New Delhi
[2]Department of Atomic Energy, Bhabha Atomic Research Center, Mumbai

Email: ranjeetranjaniari@gmail.com; shellypraveen@hotmail.com

An experiment was conducted to characterize the antioxidant defense network in parents (P1, P2, P3) and stable mutants (SM2 & SM3 at M7 stage) of wheat developed for thermotolerance through gamma-irradiation at BARC, Mumbai. During pollination stage, there was a significant increase in the activity of superoxide dismutase (SOD) in SM2 (12.02 U/mg proteins) after HS while, the minimum activity was in P_2 at control condition (3.02 U/mg proteins). During milky stage, the maximum SOD activity (5.82 U/mg proteins) was observed in SM_3 after HS and minimum (1.48 U/mg proteins) in P2 under control condition. Maximum SOD activity was observed during mealy stage in stable mutants under HS. We estimated the level of H2O2 (ROS) in parent and stable mutants, since SOD acts on the peroxide radicals and the effect is more evident during HS. The level of hydrogen peroxide at pollination stage was found maximum (3.84 µM/g FW) in SM2 after HS and minimum (1.35 µM/g fresh weight) in SM3 during control condition. During milky-ripe stage, the level of hydrogen peroxide was found maximum (3.30 µM/g FW) in P1 after HS and minimum (0.95 µM/g FW) in SM3 under control condition. At mealy-ripe stage, the level of H_2O_2 was found maximum (5.52 µM/g fresh weight) in SM3 after HS and minimum (3.02 µM/g fresh weight) in SM3 under control condition. Maximum accumulation of H_2O_2 was observed during mealy-ripe stage in SM3 under HS. Total antioxidant potential acts as potential biochemical marker for evaluating the plasticity of any cells or tissues. The level of TAC during pollination stage was observed maximum (12.26 µM Fe/g FW) in HS-treated P2 and minimum (5.97µM Fe/g FW) in SM2under control condition. Similar pattern was observed during milky- and mealy-ripe stages in both the parents and stable mutants. We established positive correlation of SOD with TAC of stable wheat mutants under HS at different stages of growth and development. SOD plays crucial role in regulating the accumulation of H_2O_2 in cells, and at same time balance the total antioxidant potential; the effect was more pronounced in stable mutants 3, as compared to parents and SM2. There is a need to further characterize the functional role of SOD in antioxidant defense network of wheat mutants using the advanced tools of proteome and metabolome – the present findings will provide easy and use-friendly technology to develop 'climate-smart' wheat using the gamma irradiation treatment.

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