INDIAN PHYTOPATHOLOGICAL SOCIETY

National Symposium & Delhi Chapter Meeting on

Innovative strategies for the management of plant disease under climate change scenario

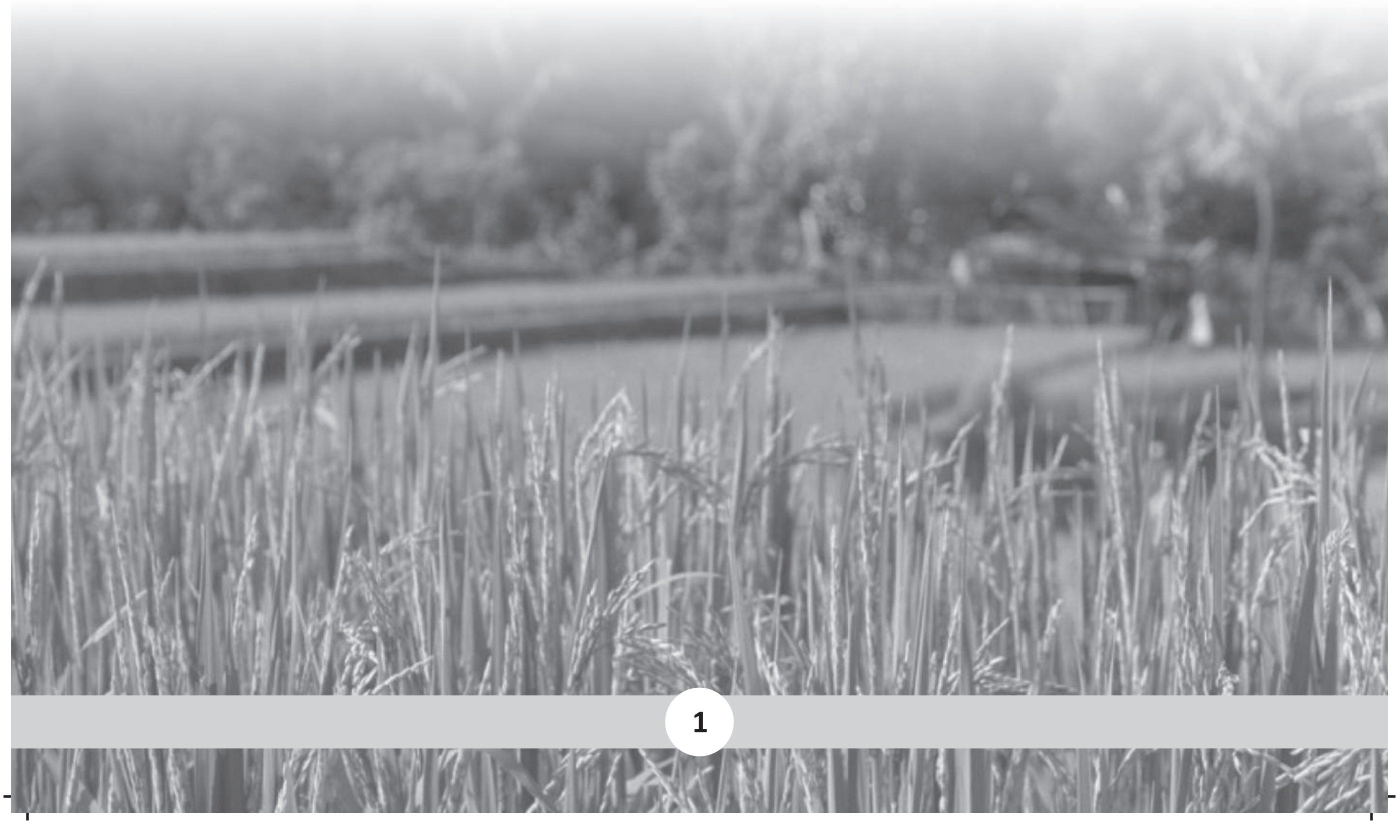
Tuesday, December 19, 2017

Abstract & Souvenir

Venue:-

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

Technical Session 01



Presidential Address

Management of plant diseases under conditions of climate change using combination approach

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Repeated and indiscriminate use of same fungicides often leads to development of fungicide resistance in pathogen. The variety which is resistant today becomes susceptible in course of time due to development of new physiological races of the same pathogen. In India, management of diseases is highly dependent on chemical fungicides and adequate levels of host plant resistance needs to searched out. Therefore advocating suitable fungicides and the manner in which it has to be used by the farmer and at which stage its application, gives maximum benefit is key question to be answered. In my small span of almost 14 years as scientist I have mainly concentrated on working on management of various diseases like spot blotch in wheat, brown spot in paddy and sheath rot in paddy, so that some recommendation can be advocated to the farmers. Ofcourse such kind of experimentation does not lead to high quality publication but satisfaction of farmers is what I consider as ultimate happiness. In my presidential lecture I will be discussing what I have done for management of few plant diseases.

Brown spot of paddy is prevalent in all the rice growing countries of the world and most of the cultivars grown are susceptible to this pathogen. Though the disease is considered to be minor one, it is increasingly posing a serious emerging threat. A field experiment on management of brown spot of paddy was carried out in two consecutive Kharif seasons (2013, 2014) on two aromatic rice varieties Pusa Sugandh-4 and Pusa Sugandh-5 in hot spot area for brown spot disease at IARI Regional Station Pusa (Bihar). Eleven different treatment combinations involving combination of fungicides and bioagent were used in this experiment. The approach of seed treatment, seedling dip and foliar spray alone and in combination was tried against brown spot of paddy. Seed treatment with Carboxin 37.5% and Thiram 37.5% WS @2.5 gm kg-1 seed and Seedling dip in suspension of Pseudomonas flourescens @ 10gm/ litre followed by two sprays of Propiconazole 25% EC @0.1% at 45 days and 60 days after transplanting gave best result. An yield advantage of 12.2 q/ha in Pusa sugandh-4 and 20.7 q/ha in Pusa Sugandh-5 has been observed over control.

Spot blotch of wheat caused by *Bipolaris sorokiniana* (Sacc.) Shoem has been a major disease of wheat grown under humid subtropical climate. The disease has a special significance in eastern Gangetic plains of South Asia that includes India, Nepal and Bangladesh. The average yield losses due to spot blotch in India were reported to be 15.5 per cent and 17 percent, even the grain yield losses ranging from 17.63-20 per cent under favourable condition.

Seed treatment by Carboxin 37.5 percent + Thiram 37.5 percent WS @ 2.5gm kg-1 seed + two foliar sprays of Propiconazole 25 percent EC @ 0.1percent one at boot leaf stage and 20 days after 1st spray gave best result in reducing the spot blotch of wheat as well as increasing the 1000 grain weight and grain yield of wheat. Only two foliar sprays of Propiconazole 25 percent EC @ 0.1 percent one at boot leaf stage and 20 days after 1st spray also gave good result in reducing the spot blotch of wheat. Therefore a combination of seed treatment by Carboxin (37.5%) + Thiram (37.5%WS) @2.5gm kg-1 seed + two foliar sprays of Propiconazole 25 percent EC @0.1percent one at boot leaf stage and another 20 days after 1st spray can be advocated to farmers against spot blotch of wheat based on findings in two years of experimentation.

Sheath rot caused by *Sarocladium oryzae* (Sawada) W. Gams & D. Hawksw is an important seed borne fungal disease of paddy which is becoming a major concern to the rice farmers in India. The combination approach was carried out in kharif 2014, 2015 and 2016 for management of sheath rot at experimental area at IARI but under two different management conditions. Chemical management and Biological management was done separately and it was observed that chemical management is giving better result than biological management.

M. K. Patel Young Scientist Award Lecture

Understanding the complex etiology of an emerging bakanae disease of rice in India

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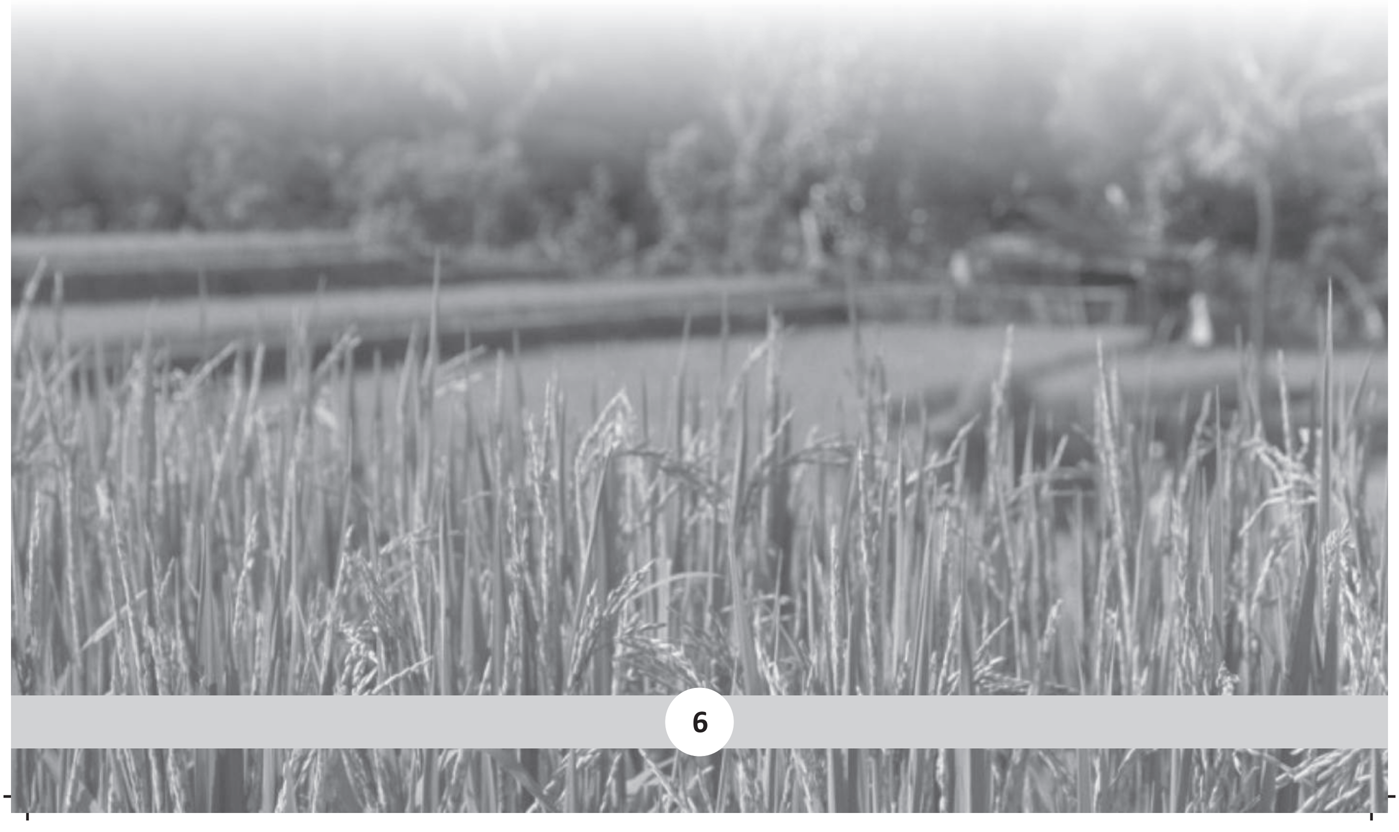
Bakanae caused by *Fusarium fujikuroi (Nirenberg)* is an emerging disease of rice in India. The typical and distinguished symptoms of the disease include elongation and rotting along with stunting in few plants. Among the two other pathogens closely associated with the disease, namely*, F. proliferatum* and *F. verticillioides*, *F. fujikuroi* is considered major incitant of this disease. *F. fujikuroi* isolates were highly virulent and caused both elongation and rotting symptoms of the disease, while *F. proliferatum* and *F. verticillioides* caused only rotting symptoms. Co-inoculations of different pathogenic species showed significant reduction in germination and increased disease severity compared to singly inoculated species. One hundred and twenty-six isolates of *Fusarium* spp. were isolated from symptomatic plants and were characterized morphologically and fort two isolates characterized at the molecular level. Mating type of the population was identified based on MAT-1 and MAT-2 region universal primers for *Gibberella fujikuroi*. Among the 63 isolates, 18 (28.57%) were identified as MAT-1 and 45 (71.42%) as MAT-2. The effective population number for mating type was 89% of the total population. Virulence analysis performed on rice variety Pusa Basmati 1509, categorized the isolates as moderately virulent (37.30%), virulent (34.13%) and highly virulent (28.57%). Morphological identification of the *Fusarium spp. is ambiguous under light microscopy; c*orrect diagnosis and identification of the pathogen is a major key to the successful management of plant diseases, hence, the diagnostics based on molecular tools, i.e., PCR based diagnostic was developed for the detection of disease. The unique sequences of *F. fujikuroi* were obtained from NCBI database and primers were designed. A marker BMFf1900 (Genbank accession no. KY953210) was developed with a sensitivity of 5 pg. To further increase the level of sensitivity of the marker, real-time primers were designed and sensitivity of the same was developed up to 10 fg. For further testing of the marker in field samples, LAMP (loop mediated isothermal amplification) was standardized. Percent transmission of inoculum (seed-seed; seedlings-seed and florets-seed) estimated, revealed maximum transmission (%) of *F. fujikuroi* is through florets followed by seedlings and seed. The disease severity and symptomatology was evaluated under different soil moisture levels, different soil pH levels, different temperatures and under different types of soil. The highest disease severity was observed at 30% soil moisture, soil pH 7.0, 35°C temperature and silty clay loam soil collected from Hapur. GC-MS analysis of inoculated and control plants showed that metabolites like Cycloeicosane were present only in inoculated plants. *F. fujikuroi* isolate "F250" was sequenced with an assembly size of 42.47 Mb providing coverage of 96.89% on reference genome. A total of 13603 protein-coding genes were predicted from genome assembly. Comparative analysis of the isolates of *F. fujikuroi* revealed that they shared a total of 12240 common clusters with F250 showing higher similarity with IMI58289. A total of 1194 secretory proteins were identified in its genome among which there were 356 genes encoding carbohydrate active enzymes (CAZymes) capable for degradation of complex polysaccharides. Besides this, 585 genes essential for the pathogen host interactions were also identified. Selected genes were validated through quantitative real-time PCR analyses in resistant and susceptible genotypes of rice at different hours of inoculation.

Biocontrol agents and fungicides were evaluated for the management of bakanae disease as seed, seedlings and spray treatments. *Talaromyces flavus* application reduced bakanae disease severity and incidence by 70-75%. It decreased gibberellic acid concentration of pathogen by 41-79% depending on the isolates. Further, it also increased aboveground biomass, grains/panicle and yield of rice. Information generated in the present study will be helpful for the better understanding and effective management of the bakanae disease.

Technical Session 02
Invited Lecture



Invited Lecture

Changing Climate, extreme weather events and Crop Diseases

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Climate –variability and -change are posing additional threat on food and nutritional security of developing countries of tropics and subtropics which are already at the threshold to cope with the demand for food for increasing population pressure. Even a small decline in foodgrain production could imbalance the food baskets and thus pushing the vulnerable populations towards hunger deaths. It has been amply demonstrated that changing climate influences the crop growth phenology and thus associated living organisms such as insepct pests and pathogens co-evolved with crops will also have to bear the brunt of changing climate. Literature accumulated so far indicates that climate change and climatic variability could alter stages and rates of development of the pathogen, modify host resistance responses, and result in changes in the physiology of host-pathogen interactions. It is expected that the range of many diseases will expand or change, and new pathogens may emerge when current natural ecosystems respond to altered temperature and precipitation profiles. Although, the research in establishing the impacts of pathogens and their natural enemies is at its infancy, independent studies conducted across the laboratories could be used to draw inferences. Elevated CO_2 levels are known to increase foliar density which in turn will influence the microclimate of the pathogen, altered host morphology which in turn may influence host-pathogen interaction, enhanced sporulation of anthracnose pathogen, and increased dry-root rot under moisture stress conditions. For instance, an increase in temperature may lead to increased host susceptibility, a new/rapid development of the pathogen, more rapid vector development leading to faster spread of the insect-borne viruses, a variable overwintering/over-summering of the pathogen/vector; shift in spread pattern of the pathogens. Initial studies show an increased sporulation and altered biocontrol traits in *Trichoderma*, a known biocontrol agent. Efforts are underway at ICAR-CRIDA to assess climate change impacts on important pathogens and their natural enemies. This effort reviews the interaction of different weather variables with different pathogens and the probable threats for food grain production, availability and quality.

Invited Lecture

Genomics-assisted Genetics: mQTL-seq mapping implicates the role of a transcription regulator gene in *Ascochyta* blight resistance of chickpea

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In this decade, next-generation genomics has drastically influenced the biological research. Application of such technologies in economically important crops will give a boost to the aim of global food security and nutrition. Production of chickpea (*Cicer arietinum*), the world's third most produced legume, is hampered by the devastating necrotrophic fungal pathogen *Ascochyta rabiei*. We applied multiple QTL-sequencing (mQTL-seq) approach to identify the QTLs that provides resistance against *Ascochyta* blight (AB) disease in chickpea. On chickpea LGIV, three robust QTLs (*qABR4.1*, *qABR4.2* and *qABR4.3*) were identified. Among these *qABR4.3* is a novel QTL. In an intra-specific mapping population, we narrowed down in *qABR4.1* region to four genes by classical mapping and progeny testing. The expression analysis of these four transcripts showed that a transcription factor is induced at higher level in resistant accession during AB stress. Dual-luciferase assay in heterologus plant revealed that *cis*-regulatory region polymorphism of this transcription factor is associated with higher expression in AB resistant accessions. The polymorphic marker developed from this *cis*-regulatory region can differentiate between *A. rabiei* pathotype-II resistant and susceptible chickpea accessions. At molecular level, this nuclear localized transcription factor can form a higher order complex with transcription factors of same family and of other families involved in plant-pathogen interaction. This transcription factor is targeted by an *A. rabiei* PEC25 effector in plant nucleus. This effector is delivered by *A. rabiei* in host cells for fungal virulence. Thus, genomics-assisted genetic analysis of AB resistance in chickpea has culminated in the identification of a transcription factor and its polymorphism in *qABR4.1*. The genes and polymorphic markers associated with AB resistance will help in biotechnology and breeding based chickpea improvement programmes. The basic knowledge obtained by functional characterization of such transcription factor-fungal effectors interactions can be extrapolated to other closely associated pathosystems.

Invited Lecture

Innovative strategies for biotic and abiotic stress alleviation in wheat plants

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Spot blotch of wheat caused by Bipolaris sorokiniana is one of the most important diseases of wheat.Serological formats using purified IgG of B. sorokiniana (KM 066949) were developed for screening of resistance of wheat germplasm against the pathogen following PTA-ELISA format and Dot immune binding assay. One of the highly susceptible genotype **(**CWL 6702) was selected for induction of resistance using bioinoculant against B. sorokiniana. Talc based formulation of Bacillus methylotrophicus (NAIMCC-B 01492), a potent PGPR was applied both as seed treatment and foliar application was used as soil application. The bacterium was found to be most effective in disease reduction. Time course accumulation of chitinase (CHT), β-1,3glucanase (GLU) and phenyl alanine ammonia lyase (PAL) increased markedly in treated plants in comparison to healthy control following 12, 24, 48, 72 and 96 h of challenge inoculation with B. sorokiniana. Immunolocalization of chitinase and glucanase in bioinoculant treated and pathogen inoculated leaf tissue was further confirmed bytransmission electron microscopy using PAb of chitinase, glucanase and gold labelled conjugates.Microarray analysis were done to compareup regulated and down regulated gene expression among untreated pathogen inoculated, B. Methylotrophicus treated B. sorokiniana inoculated plants and untreated uninoculated ones. Among the up-regulated genes, most significant ones were those of pathogenesis related protein 10, Phenylalanine ammonia lyase, glucan endo-1,3-beta-D-glucosidase, 5-enolpyruvylshikimate 3-phosphate synthase, β-1,3 glucanase, peroxidase, pathogenesis-related protein 4.Further, relative expressions of defense genes as well as genes involved in phytoalexin biosynthesis were analysed by real-time PCR with cDNA from leaf tissue. Expression of four defense genes encoding chitinase, β-1,3 glucanase, phenyl alanine ammonia lyase and peroxidase as well as genes encoding chalcone synthase (CHS) and chalcone isomerase (CHI) involved in phytoalexin biosynthesis also increased markedly within 48h of inoculation in treated plants than untreated healthy control.

On the other hand, when wheat seeds were primed with either Bacillus safensis and Ochrobactrum pseudogrignonense, showing plant growth promoting activity and one month old young seedlings were subjected to heat treatment for different time period, both the PGPR priming improved heat stress tolerance level of wheat seedlings. However, among the two, Bacillus safensis was more effective for improving thermo tolerance. High temperature induced the accumulation of ROS and was responsible for membrane injury and chloroplast ultrastructure damage. As a defence reaction plants in turn activated antioxidant signalling which increased redox enzyme activity and accumulated osmolytes like proline and glycine betain. PGPR mediated amelioration of heat stress seems to be associated with less ROS production, membrane damage, maintenance of chloroplast structure and photosynthetic efficiency, increased expression of array of redox enzymes and accumulation of osmolytes which improved overall thermo tolerance. Besides, B. safensis treated plants also induced resistance against B. sorokiniana as evidenced by activation of defense genes. This study revealed future perspective of amelioration of biotic and abiotic stresses using microrganisms.

Technical Session 03
MJ Narshimhan
Academic Award Contest

MJ Narshimhan Academic Award Contest

Cross infectivity and comparative genomics of *Bipolaris sorokiniana* and *Bipolaris oryzae* for characterization of pathogenicity related genes in wheat and rice

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Spot blotch of wheat caused by Bipolaris sorokiniana and brown spot of rice caused by *Bipolaris oryzae* have resulted in great losses to food grain yield especially in regions where rice-wheat cropping system is pre-dominant. Here, cross infection study under artificial conditions with most virulent isolates revealed that *when* inoculation of BS-112 on wheat and rice was done, infection index of 89% and 80% was obtained, while BO-1 inoculation showed infection index of 85% and 78% on rice and wheat respectively. Cross infectivity was confirmed by PCR using species specific markers developed in the laboratory. Cross infectivity was also identified under natural condition for both *B. sorokiniana* and *B. oryzae* on rice and wheat at certain places. A high level of similarity between these two closely related pathogens with respect to genes associated with host specificity as well as candidate effectors genes was identified through comparative bioinformatics. Comparative secretomics of *B. sorokiniana* and *B. oryzae* genomes led to identification of 262 and 247 predicted small secreted proteins (SSPs) respectively out of which 34 and 28 SSPs were assigned putative function in B2G analysis. Expression analysis of six selected unique and common SSPs to *Bipolaris* spp. showed that only 2 SSPs (BS _SSP2 and BOBS_SSP2) expressed as a typical fungal candidate effector after inoculation on plant (*in planta*). In addition to this melanin pathway gene SCD1 which encodes scytalone dehydratase, was characterized in *Bipolaris sorokiniana.* Targeted disruption of SCD1 gene showed that this is necessary for melanin biosynthesis in *B. sorokiniana*. SCD1 gene mutant isolates lack melanin synthesis and showed significantly reduced pathogenicity on its own host wheat. Together, our analyses broadly expanded our knowledge and offered insights into the cross infection and pathogenesis of *Bipolaris* spp. in wheat and rice.

MJ Narshimhan Academic Award Contest

Simulation of Cyclic adenosine monophosphate (cAMP) dependent Protein Kinase A (PKA) activity in relation to appresorium formation in Magnaporthe oryzae under temperature influence

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Temperature rise due to climate change is expected to affect pathogenicity of the pathogens. Temperature has a significant influence on growth and sporulation of rice leaf blast pathogen (*M. oryzae*). Various components of rice blast infection process observed maximum at 27°C (optimal temperature) as compared to suboptimal (22°C) and supra-optimal (32°C) i.e., spore germination, appressoria formation, germ tube growth. This is an indirect indication that *M. oryzae* is not capable to cause infection at higher temperature. Temperature has significant influence on cAMP dependent PKA activity. The PKA activity is higher at optimal temperature as compared to both suboptimal and supra-optimal temperatures. The PKA activity increases during appressoria formation and later on it decreases due to dephosphrylation that indicated change in enzymatic activities in the appressoria, leading to change in pathogen aggressiveness. The cAMP dependent PKA activity was fitted to kinetic models. However model could not explain the steady state behaviour that occurs 8 h onwards. May be there for, *M. oryzae* needs additional kinetics parameters in the models.

Total proteins and phosphorylated protein are also found to be higher at optimal temperature. Total protein oscillation is an indication of transfer to fill the need of new protein in particular time. Total phosphorylated protein is highest at the appressoria formation indicated the demand of many biochemical processes where phosphorylation is required. The cAMP dependent PKA activity influence on components of pathogen infection process is an important study to understand pathogen biology that may help in development of new target fungicides.

MJ Narshimhan Academic Award Contest

Molecular characterization of phytoplasma association with four important ornamental plant species in india and identification of natural potential spread sources

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Phytoplasma suspected symptoms of phyllody, witches' broom, leaf yellowing, stunting and little leaf were observed in *Chrysanthemum morifolium, Bougainvillea glabra, Jasminum sambac* and *Callistephus chinensis* during survey of flower nurseries and experimental ornamental fields at Delhi, Maharashtra, Tamil Nadu and Karnataka from 2014 to 2016. Pleomorphic bodies typical to phytoplasma structures were observed in phloem sieve elements of ultrathin sections of all the four symptomatic ornamental plants (stem tissue) in transmission electron microscope. Amplification of 1.8 and 1.2 kb phytoplasma DNA products were observed in all the four test plants in PCR assays using universal primer pairs P1/P7 followed by nested primer pair R16F2n/R16R2, respectively. Pair wise sequence comparison, phylogeny and virtual RFLP analysis of 16S rDNA sequences confirmed the association of two phytoplasma subgroups (16SrI-B and 16SrII-D) in four ornamental plants species. '*Ca.* P. aurantifolia' subgroup D (16SrII-D) was found associated with chrysanthemum phyllody and leaf yellowing at Delhi and Tamil Nadu; bougainvillea little leaf and yellowing at Delhi and Chinese aster phyllody at Bengaluru, Karnataka. However, jasmine little leaf and yellowing at Bengaluru, Karnataka and chrysanthemum stunting at Pune were to be found associated with '*Ca*. P. asteris' subgroup B related strains (16SrI-B). The identification of 16SrII-D subgroup phytoplasma infecting bougainvillea and 16SrI-B subgroup infecting jasmine are the new reports to the world. Besides, weed species, *Cannabis sativa* showing witches' broom in jasmine fields at Bengaluru and *Parthenium hysterophorus* showing witches' broom symptoms in chrysanthemum fields at Delhi were identified to be caused by phytoplasma strains classified under subgroups 16SrI-B and 16SrII-D, respectively by PCR assays and 16Sr DNA sequences comparison analysis. *Empoasca prima* (Distant), *Hishimonus phycitis* (Distant) (HP) and *Amrasca biguttula biguttula* (Ishida) were identified as major leafhopper species feeding on ornamental crops in all the four surveyed states. Out of the three major leafhopper species identified, only *H. phycitis* was identified positive for 16SrI-B and 16SrII-D subgroups of phytoplasma from chrysanthemum fields at Delhi and jasmine fields at Bengaluru, respectively. The identity of similar phytoplasma strains infecting ornamental species in leafhopper, *H. phycitis* (16SrII-D and 16Sr-B) and the weed species (*C. sativa* witches' broom, 16SrI-B; *P. hysterophorus* witches' broom, 16SrII-D) in the present study suggested that *H. phycitis* and weeds may act as potential natural sources for secondary spread of the identified phytoplasmas strains in respective areas of their existence.

MJ Narshimhan Academic Award Contest

Selection of a compatible biocontrol consortium effective against Fusarium wilt of tomato

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Fusarium oxysporum f.sp. *lycopersici* (FOL), an incitant of vascular wilt of tomato, needs to be contained through efficient sustainable management practice to boost the yield and quality of marketable produce. To enhance the consistency and degree of disease control offered by individual antagonists, effective strain mixtures consisting of *Pseudomonas putida* (TEPF), *Bacillus subtilis* (S2BC-1), *Trichoderma harzianum* (S17TH) and *Chaetomium* sp (CG-A) were designed based on growth, antagonism and antifungal enzyme and gene expression assays. In pot experiments, seed treatment and soil applications of suspension cultures of 5 different strain mixtures were assessed for their efficacies in vascular wilt control over individual strains upon challenge inoculation with FOL under polyhouse conditions. In general, the biocontrol strain mixtures greatly reduced vascular wilt incidence besides promoting tomato plant growth in comparison to the pathogenic and untreated controls. However, the biocontrol performance of the consortium housing all the four strains was lesser than the strain mixtures containing three strains. Among the treatments, application of a strain mixture comprising TEPF+CGA+S17TH under challenge inoculated conditions resulted in significantly lower incidence (71.4% reduction) of Fusarium wilt and increased root and shoot length of 135.5% relative to the pathogen control without biocontrol agents. In further studies on elucidating the role of defence genes of tomato plants in vascular wilt suppression by the biocontrol consortia, though strong expressions of *PR3b* and *PR5*, the marker genes for salicyclic acid modulated pathway and *gluB*, the marker gene for ethylene modulated pathway were observed over the pathogenic and untreated controls, in the treatments, S2BC-1+CGA+S17TH and S2BC-1+TEPF+S17TH, a reduction in expression was observed for the best performing strain mixture, TEPF+CGA+S17TH. The reduced expression indicated that novel genes other than the candidate ones might be playing a role in the resistance induction. Further, plant growth promotion by the consortia could be attributed to the suppression of the pathogenic fungi besides other mechanisms that need to be explored in detail. The identified consortium has the potential as a preferred input in integrated disease management systems once formulated and tested under field conditions.

MJ Narshimhan Academic Award Contest

A geminivirus betasatellite encoded βC1 protein interacts with PsbP and subverts PsbP-mediated antiviral defense in plants

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Geminivirus disease complexes potentially interfere with plants physiology and cause disastrous effects on wide range of economically important crops throughout the world. Diverse geminivirus-betasatellite association worsens the epidemic threat for global food security. This study provides the first evidence of chloroplast localization of a DNA virus encoded protein which in turn affects the ultrastructure and function of the chloroplast. *Radish leaf curl betasatellite* (RaLCB) encoded βC1 protein gets localized into the chloroplasts of the infected *N. benthamiana* plants and causes damages to the OEC of PSII. The antiviral response against virus systemic infection is known to be majorly associated with chloroplast function. Conversely, plant viruses primarily target and exploit chloroplasts to establish viral pathogenesis and symptom induction. In this present study, the virus-chloroplast interaction in the scope of viral pathogenesis as well as plant defense response was explored. Interaction between host encoded PsbP protein with RaLCB-βC1 protein was confirmed both in vitro and in vivo. Host PsbP protein binds to geminivirus DNA both in-vivo and in-vitro. Transient silencing of *PsbP* in *N. benthamiana* plants greatly enhances symptom induction and viral DNA accumulation. Over-expression of PsbP impedes disease development during the early phase of infection, suggesting that PsbP generates defense response during geminivirus infection. In addition, βC1-PsbP interaction hampers binding of PsbP to the geminivirus DNA. Taken together, these findings suggest thatbetasatellite encoded βC1 protein accomplishes counter-defense by physical interaction with PsbP and by interfering with ability of PsbP to bind geminivirus genome to ensure the establishment of viral pathogenesis.

MJ Narshimhan Academic Award Contest

Genetic manipulation in the genome of *Soybean yellow mottle mosaic virus* (SYMMV) and infectivity analysis for exploring its potentiality as a transient gene delivery system

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Genetically manipulated plant viruses resulted in the development of vectors that could be utilized for gene expression and silencing studies and for delivery of other genetic constructs into plants. Though many plant viruses have been utilized for such purposes in many crops either transgenically or transiently, but their potentiality in pulse crops still remains unearthed. In the present study, two different mutant amplicons i.e. coat protein deleted mutant and multiple cloning site (MCS)-added full-length mutant of an infectious clone of a pulse infecting carmovirus, *Soybean yellow mottle mosaic virus* (SYMMV) were generated using inverse PCR and overlapping extension PCR methods, respectively. Among these mutants, the coat protein deleted mutant was not stable after cloning in *E.coli*. However, the other mutant, where a MCS with four restriction sites (*Kpn*2I, *Bst*1107I, *Ksp*AI, and *Rru*I) was inserted successfully at the beginning of the coat protein ORF of SYMMV genome, was found to be stable in *E. coli*. This MCS-added full-length mutant successfully reproduced the disease symptoms upon agroinoculation in the different legume species. However, compared to the wild type, the mutant produced milder symptoms and took longer time for symptom expression. The production of mutant virions after agroinoculation and its systemic movement in different agroinoculated legume species was evidenced by electron microscopy, ELISA, and RT-PCR, but its replication is markedly reduced as suggested by real-time estimation of transcript abundance. The approach for inserting an MCS into the infectious construct has been standardized and the same strategy will be helpful for introducing more manipulation in the SYMMV genome in future to enhance the efficacy of the construct so that it could be used as a vector for gene delivery in the plant.

MJ Narshimhan Academic Award Contest

Trichoderma mediate early and enhanced lignifications in chickpea during *Fusarium oxysporum* f. sp. *ciceris* infection

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Lignifications in secondary cell walls are implicated to take part in defensive mechanism inplants during infection by invading pathogens. In the present study, chickpea seeds were treated with 10 different strains of bio-control potential *Trichoderma* strains and subjected tothe wilt pathogen *Fusarium oxysporum* f. sp. *ciceris* (Foc). Subsequently, alterations in lignin content in the chickpea plants was observed through histochemical staining and expression pattern of some lignin biosynthetic pathway genes were studied. Accumulation of lignin wasobserved in transverse sections of shoots near the soil line and expression pattern of thetargeted genes was observed in roots at different time intervals after pathogen inoculation through pholoroglucinol staining and semi quantitative RT-PCR, respectively. Lignin deposition and expression patterns of the target genes were variable in each treatment. Although lignifications took place in all *Fusarium* challenged chickpea plants treated with *Trichoderma* strains, interestingly out of the 10 *Trichoderma* strains 4 strains viz., T41, Mv-41, DFL and RO triggered highest lignifications in chickpea collar region. Time coursestudies showed that effective *Trichoderma* isolates induced lignifications very early compared to the normal process of lignifications and it nearly completes within 6 days of pathogen challenge. The results thus demonstrated that effective *Trichoderma* strains trigger lignifications early in chickpea under Foc challenge and provide better protection to chickpea plants.

Technical Session 04
Poster Presentations

Theme Area: Integrated plant disease management

Gene expression analysis of genes associated with lignin biosynthesis pathway during root wilt pathogen infection in chickpea

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Chickpea is a major pulse crop among all the pulses. It is continuously facing biotic and abiotic challenges, among biotic challenges fusarium wilt incited by *Fusarium oxysporum* f.sp. *ciceris* is a major problem. For this we studied expression pattern of 10 genes associated with lignin biosynthesis during infection and treated with 10 different *Trichoderma* isolates. Lignin is a main component of secondary cell wall which provides strengthening of cell thereby restricts the root invading pathogen by providing physical resistance hence lignin biosynthesis genes were selected in this study. RNA was isolated on 0 day, 6thday and 15th day after inoculaton. PCR analysis showed that lignin biosynthetic pathway genes expression is more on the 6th day after pathogen inoculation compared to 0th and 15th day which showed mostly basal levels of expression of the genes viz., PAL, C3H, C4H, CAD, COMT and F5H. High expression of the genes was recorded with *Trichoderma* isolates T-42, MV-41, RO and DFL which also showed enhanced lignifications. This study is helpful in dual manner one is nonchemical management of root wilt pathogen by triggering the plant strength by using bioagents another is through the antagonistic properties of biocontrol agents itself.

Fluorescent pseudomonads: As potential bio-control agent against corm rot of saffron

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Saffron is known as golden condiment is world's most valuable spice and is derived from stigma of *Crocus sativus*. It possess crocin for the red colour, safranal for its aroma and picro-crocin for bitterness. Saffron crop is majorly grown in Jammu and Kashmir, in recent year the production of the saffron is reducing because of the various biotic and abiotic stresses. Corm rot of saffron is one the major disease which reduces the production and quality of the saffron. Although chemical fungicides were widely advocated and applied for the management of corm rot of saffron but their continuous uses showed hazardous effect on both the soil as well as crop. Therefore biological control gained importance in recent decade by reducing the pathogen population and promoting the growth and yield of the crop. Beneficial bacteria like fluorecscent pseudomonads are reported to be beneficial to the many crops which include all *Pseudomonas* species with the ability to produce fluorescent pigment *viz*., *P. aeruginosa, P. syringae, P. putida and P. fluorescens*. Because of their diverse mechanisms of action, high level of genetic variability and competitiveness in soil, they emerged as effective and economical bio-inoculants for use in integrated disease management. The bio-control mechanism of fluorescent pseudomonads normally involves the production of antibiotics lipopeptide antibiotic viscosinamide, 2,4-diacetylphloroglucinol, secondary metabolites and phytohormones , volatile compounds such as HCN,siderophores and also cell wall hydrolysing enzymes such as protease, chitinase, glucanase. Plant growth-promoting ability of these bacteria is mainly because of the root colonization, production of indole-3-acetic acid,cytokinins, gibberellins and phosphate solubilising activity.

Integrated management of bacterial wilt of tomato caused by *Ralstonia soalanacearum*

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A total of 12 treatments comprising most effective (based on plate/pot culture studies) antibiotics, fungicides, bioagents, organic amendments and phytoextracts were attempted (alone or in combination) for the integrated management of bacterial wilt pathogen (*R. solanacearum)*, by applying sick soil technique. Among the treatments, Streptocycline + *P. fluorescens* + vermicompost were found most effective with significantly highest germination (78.32%), respectively, followed by Streptocycline + Copper oxychloride (73.29 %) and *T. horizanum*+ *P. fluorescens* (68.30 %). All the test amendments recorded significant reduction in least average mortality (PEM and Wilt) over untreated control. However highest least average mortality reduction was recorded with Streptocycline + *P. fluorescens* + vermicompost were found most effective with significantly least reduction (65.73 and 60.27 %), respectively, followed by Streptocycline + Copper oxychloride (59.88 and 54.23 %) and *T. horizanum*+ *P. fluorescens* (52.38 and 47.32 %) respectively.

Role of *Fluorescent pseudomonas* in reduction of the use of chemical pesticides and fungicides in normal and replant sites of apple and pear

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The chemical pesticides and fungicides being used to control the phytopathogenic microbes have the negative effects on the environment and human health. Effective alternative is to employ the pathogen's natural enemies as biocontrol agents, which is less destructive and environmental friendly. PGPR are a group of bacteria that actively colonize plant roots and increase plant growth and yield. Among various biocontrol agents, *Pseudomonas* sp., equipped with multiple mechanisms of biocontrol of phytopathogens is being used widely. Biological control of plant pathogens is through the production of antibiotics, lytic enzyme, siderophore and HCN production. In the present study, the *Pseudomonas* spp. were isolated from the rhizosphere of apple and pear and screened for their biocontrol properties such as antifungal activity, siderophore production and HCN production. Out of 30 isolates, 15 isolates showed antifungal activity against *Fusarium*sp. and *Alternaria* spp. In plate assay method, siderophore production by *Pseudomonas* isolates was recorded in the range of 20 mm to 32 mm in terms of pinnkish/orange zone in Chromeazurol-S agar plates, while in quantitative assay, siderophore production was observed in terms of reduction in blue colour in the range of 20.83 to 68.70 % SU.All the isolates were found to be positive for ammonia and HCN production. Taken together, results suggest that *Pseudomonas* spp. is an efficient approach to replace agrochemicals and pesticides, thereby reducing their negative effects on the environment.

Integrated management of major tomato diseases in the upper Gangetic plains of India

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Tomato is the most important vegetable of India, huge losses of marketable fruit yield and high infestation with different diseases has reduced open field tomato production, causing whole fields to be deserted prior to harvest in the Upper Gangetic Plains of India. Therefore, an integrated approach involving physical, biological and chemical module has been formulated to manage diseases like "early blight", "late blight", "target leaf spot", "*Sclerotium* collar rot", and tomato "leaf curl virus" under control. Results showed that major diseases prevalent in this zone could effectively be controlled in a substantial manner over the years through integration of management practices adopted both in nursery and field by the application of fungicide treatment and neem oil spray [@4g/kg of seed followed by soil application @ 5ml/litter, while filling of plug trays and soil drenching of Seed @5% after seed germination followed by covering of nursery bed with 50-mesh nylon net] and main field [Seedling dip with 0.1 % (Carbendazim 12%+ Mancozeb 63% WP) at the time of transplanting and sequential spraying with Acephate 75% WP @1.5g/l on 10 days after transplanting (DAT)] and [CoC treatment of the soil after transplanting of the seedlings] results better crop helth. Such approach could also be beneficial for prolong tomato production and high economic return. This technology could easily be recommended in areas of the tropics where early *autumn or autumn* season cultivation of tomato is hampered by major diseases.

Appraisal of integrated approaches to management of soil borne diseases in brinjal

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Soil-borne diseases are major constraint in the cultivation of brinjal and cause considerable losses all over the world including India. Present investigation was carried out for two consecutive years during 2014-15 and 2015-16 to evaluate bio-control agents, fungicides and combination of both for their effectiveness against soil-borne disease incidence and also on the growth and yield parameters in brinjal. Our results indicated that under nursery conditions, seed dipping in *Pseudomonas fluorescens* (cfu 10^8/ml) along with soil drenching with carbendazim@ 0.2% exhibited 86 per cent reduction in wilt incidence after 30 DAS as compared to control. Under field condition, integration of root dip with *P. fluorescens* followed by soil drenching with carbendazim twice at seven days interval of transplanting resulted in 82 per cent reduction in wilt incidence and concurrently augmented 45 and 59 per cent in plant height and yield of brinjal crop.

Application of Rhamnolipds biosurfactant for the management of banded leaf and sheath blight (BLSB) disease of maize

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Banded Leaf and Sheath Blight (BLSB) of maize is most prevalent in south and south-east Asian countries. The disease is caused by a soil born fungus *Rhizoctonia solani* f. sp. *sasakii* (Kuhn) Exner [teleomorph: *Corticium sasakii*, syn. *Thanatephoru scucumeris* Frank (Donk)]. The pathogen is one of the most widespread, destructive and versatile pathogen infecting plants belonging to 32 families and 188 genera consisting of cereals, millets, pulses, oilseeds, grasses and weeds. Crop damage is caused by loss of photosynthetic leaf area due to foliar infection and stalk rot, leading to crop lodging. Maximum damage is caused when ears are infected. Present investigation was carried out on the bio-control efficacy of rhamnolipids (RLs) biosurfactant (at 25, 50 and 100 ppm) against the pathogen *in vitro* adopting poison food technique.The biosurfactant inhibited mycelial growth of *R. solani* f. sp. *Sasakii* by 76.7% at 100ppm followed by 71.5% at 50ppm and 70% at 25ppm.Rhamnolipid was further evaluated against BLSB in maize (variety Vivek QPM 9) under field conditions during *kharif* 2017 using four treatments *viz.,* seed treatment @100 mg/lit, foliar spray @ 100 mg/lit, seed treatment with foliar spray @100 mg/lit and Hexaconazole foliar spray @ 1ml/lit. Maximum disease inhibition (16.41%) was found in biosurfactant foliar spray @ 100 mg/lit. Highest grain yield (41.35q/ha) was obtained in the foliar spray of biosurfactant@ 100 mg/lit followed by its seed treatment with foliar spray @100 mg/lit (38.75q/ha). However, the investigation will be further repeated.

Management of Phyllosticta leaf spot (*Phyllosticta zingiberi*) of ginger (*Zinger officialane* Rose.) fungicides

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Ginger (*Zinger officialane* Rose.) belonging to family Zingiberaceae and rhizomatous spice. Ginger is affected by a number of diseases soft rot (Pythium spp.), yellows (Fusarium spp.), rhizome rot complex (Fungi, nematode/ insect interaction), Phyllosticta leaf spot and storage rots (Pythium spp., Fusarium spp. etc.) which lead to reduction in yields . Apart from many leaf spots diseases Phyllosticta leaf spot caused by *Phyllosticta zingiberi* has been observed to be the most destructive. In recent years Phyllosticta leaf spot gaining much importance for the management of the disease. The trail was conducted in the randomized block design with three replications from (2013-14 to 2015-16) at College of Agriculture and Reserach Stataion under AICRP on Spices. The plot size of the experiment is 3m×1m with 30cm x 20cm spacing (50 rhizomes per plot). The recommended dosage of the fertilizers were applied as 120 Kg N: 100 Kg P: 120 Kg K. The local farmer's seed material was used as planting materials. The eight tretamentas follows Foliar spray with Mancozeb (0.3%) first at disease appearance with two times at 20 days of Interval(T_1), Foliar spray with Carbendazim (0.1%) first at a disease appearance with two times at 20 days of Interval(T2).Foliar spray with Carbendazim:Mancozeb (0.1%) first at a disease appearance with two times at 20 days of Interval(T_3), Foliar spray with Blitox (0.3%) first at a disease appearance with two times at 20 days of Interva(T_4) l, Foliar spray with Propiconazole (0.1%) first at adisease appaerence with two times at 20 days of Interval(T_5), Foliar spray with Trycyclazole (0.1%) first at a disease appearance with two times at 20 days of Interval(T_6), Foliar spray with Hexaconazole (0.1%) first at a disease appearance with two times at 20 days of Interval(T_7) and Control(T_8). The result of three years pooled result as Minimum disease intensity 14.52 and maximum yield 5.3 t /ha was found with spray of Carbendazim: Mancozeb (0.1%) first appearance of the disease with two times at 20 days of interval. The next best treatment was Trycyclazole (0.1%) first at disease appearance with two times at 20 days interval and Hexaconazole (0.1%)

Induction of systemic resistance against Fusarium wilt of chickpea by non -convectional chemicals

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Induced systemic resistance in chickpea against wilt disease caused by Fusarium oxysorum f. sp. ciceri (FOC) was studied by treating the seeds with twelve non convectional chemicals viz., Salicylic Acid (SA), Acetyl Salicylic Acid (ASA), DL-β-amino-n-butyric acid (BABA), gamma-amino-n-butyric acid (GABA), Amino-iso- butyric acid(AIBA), Indole -3 –pyruvic acid (IPA), Indole-3 –acetic acid (IAA), Nicotinic acid (NA), Isonicotinic Acid (INA), DL-Norvaline, Benzoic Acid and Cycloheximide,. All the chemicals were effective in reducing mortality of chickpea from Fusarium oxysporum f.sp.ciceri. These non convectional chemicals increased germination of chickpea seed and reduced per cent mortality as compared to control. Individual treatment of the chemicals showed better results than their combinations as plant mortality was reduced and accumulation of SA, BABA and NA increased in their individual treatments. Slight increase in yield was observed with all the treatments All the chemicals showed non-fungi toxic response against Fusarium oxysporum f.sp. ciceri when tested in in vitro by poison food technique. Maximum germination (95%) was observed in pots wherein the seeds were treated with salicylic acid followed by NA (90.6%) and BABA (90.3%). There was non-significant difference between these chemicals. All treatments were significantly superior to check in reducing the wilt incidence.

Management of Alternaria blight and browning diseases of cauliflower under field condition

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Cauliflower is an important vegetable crop grown in India. Browning disease due to deficiency of boron in soil and A*lternaria* blight diseases in cauliflower are the major production constraint in cauliflower in India, particularly in Jharkhand state. High incidences of above diseases in cauliflower are correlated with increase in many parameters involved in cauliflower production system. These include heavy application of nitrogenous fertilizers, use of responsive cultivars, spacing, change in climatic condition, cropping pattern, poor availability of boron in soil etc. associated with modern cauliflower production system. Hybrids and HYVs are more prone to above diseases of cauliflower. Due to these diseases of cauliflower, yield loss was recorded up to 70%. Therefore, the present field trials were conducted at Zonal Research Station (Birsa Agricultural University), Darisai, East Singhbhum, Jharkhand.

Lowest *Alternaria* blight disease intensity of 10.6% and browning disease of 5.8 % were recorded when cauliflower seed was treated with *Trichoderma viride* plus Soil application of Boron @ 10 Kg /ha plus two sprays of Chiraita leaf extract (5%)was applired, this treatment also recorded highest flower yield of cauliflower(520.4q/ha) and increase in flower yield over control of 79.6% and B:C ratio of 1:70. This treatment was at par with Seed treatment with *Trichoderma viride* plus Soil application of Boron @ 10 Kg /ha plus two sprays of tulsi leaf extract (5%) which recorded cauliflower yield of 500.0 q/ha, increase in flower yield over control of 72.5%, *Alternaria* blight disease intensity of 14.4 % and browning disease of 4.2%. Whereas, the control plots recorded *Alternaria* blight of 20.6% and browning disease of 57.5%. and cauliflower yield of 289 q/ha.

Theme Area: Climate change, epidemiology and disease forecasting

Effects of transplanting dates on the incidence of False smut, Sheath rot and Bakanae diseases in different paddy cultivars

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A field experiment was conducted at the experimental farm of IARI, Regional Station, Karnal to evaluate the effect of transplanting time on the disease incidence and seed yield under a uniform fertilizer dose. Assessment of diseases was done on the basis of the data recorded at fortnightly intervals after 15 days of germination till harvest (120 days after sowing). Among three dates of transplanting, the disease incidence varied with the varieties. Highest yield attributing characters were recorded at first date of transplanting (5th July for PB-1121 and PB-6 & 15th July for PS-5 and PB-1509); and growth parameters decreased with further delay in transplanting time probably due to reduction in day length and average temperature. Paddy cv. Pusa Sugandh-5 produced greater quantum of seed than other paddy varieties. The maximum incidence of various diseases was occurred in late transplanted paddy cultivars and it was minimum in paddy cultivars transplanted on 5th of July. The maximum disease incidence of false smut (15.76%) and sheath rot (24.56%) diseases were recorded in paddy cultivar PS–5 transplanted on 25th July and 5th August, respectively. The maximum incidence of bakanae disease (13.92%) was recorded in paddy cv. PB–1121 transplanted on 15th July, 2015. Three years study showed that change in climatic conditions influence the disease incidence.

Etiology of corm rot disease of saffron crop

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Saffron is an important crop having high medicinal value. In India saffron is cultivated in Jammu and Kashmir and is known for its high quality. But the production of the saffron crop is reducing due to various biotic and abiotic stresses. The biotic stress that reduce the production of the saffron is corm rot caused by various plant pathogenic fungi *viz*., *Rhizoctonia crocorum, Phoma crocophila, Fusarium moniliforme* var. *intermedium, Macrophomina phasiolina, F.oxysporium, F. solani, F. pallidoroseum, F.equisseti, Mucor* sp., *Penicillium* sp. and *Sclerotium rolfsii. Fusarium oxysporium* f.sp. *gladioli* was the most predominant pathogen (58%) followed by *F. solani* (28%) associated with corm rot of saffron. Frequency of isolation of *F. moniliforme* was 12 percent, and having 8 per cent of *F. pallidoroseum*. The infected corm show minute black spot s distributed on the corm which later turn into blackish brown irregular lesions, coalescing with each other making the entire corm to dry and reduce the size of the corms as compared to the healthy corm. In the advance stage the entire corm turn into dark powdery mass. As the disease progress the entire affected corm decay and corm appears spongy due to the disintegration of the tissues. With the prevalence of drought conditions, poor corm health and lack of source of resistance against the corm rot are the major factors which aggrevated the diseas in saffron fields of Jammu and Kashmir.

Severity of cercospora leaf spot of greengram in relation to weather parameters and age of the crop

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A field experiment was conducted at RARS, Lam, Guntur, Andhra Pradesh during *kharif* and *rabi* 2015-16 to assess the severity of Cercospora leaf spot on Greengram in relation to weather parameters and phenological stage of the crop. Highest disease severity of 58.2% was observed at flowering stage with maximum mean temperature 37 ^{O}C, minimum temperature 24 ^{O}C, morning RH 92%, evening RH 51 and there is no rainfall during Nov- Dec. Correlation and multiple regression analysis was carried out between (Per cent Disease Index) PDI and weather parameters *viz*., maximum temperature (^{0}C), minimum temperature (^{0}C), morning relative humidity, evening relative humidity, rain fall (mm) and age of the crop. Among the weather parameters, minimum temperature (-0.26) and evening relative humidity (-0.30) were significant and negatively correlated with PDI whereas significant positive correlation observed between PDI and age of the crop (0.85). When the data on PDI was subjected to multiple linear regression (MLR) and correlation with weather variables, it was observed that the coefficient of determination (R^2) for PDI was 0.812 which showed that weather factors were able to cause the variation in PDI to an extent of 81.2%. This study concluded that effective management practices suggest to the farmers based on the disease severity threshold level.

Effect of different abiotic factors on disease severity and symptomatology of bakanae disease of rice incited by *Fusarium fujikuroi*

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The bakanae disease of rice is incited by *Fusarium fujikuroi* Nirenberg (sexual stage: *Gibberella fujikuroi* (Sawada) Wollen worth). The disease is becoming a serious threat to cultivation of rice specially basmati rice in India and other rice growing nations. The disease is characterized by production of various kind of symptoms like crown or foot rotting, lanky, tall slender elongated culms with pale yellow flag leaves and sterile grains. The pathogen is known to produce various secondary metabolites and mycotoxins such as Gibberellic acid, Fusaric acid, Fumonisins, Bikaverin etc which affects the pathogen virulence and type of symptoms produced. The different abiotic factors might play a role in the production of different kind of symptoms. The disease severity and symptomatology was evaluated under different soil moisture levels (fully saturated, 75% of saturation, 50% of saturation and 30% of saturation), different soil pH levels (6.0, 6.5, 7.0, 7.5, 8.0 and control pH of 8.44), different temperatures (25°C, 30°C and 35°C) and under different types of soil (Sandy Loam, Clay, Clay Loam, Silty Clay Loam and Sandy). The parameters like seed germination, disease severity, types of symptoms produced, plant height, root length and plant weight were recorded in all experiments. F250 isolate of *F. fujikuroi* and susceptible rice genotype Pusa Basmati 1121 was used for evaluation of disease under different conditions. The growth and sporulation of pathogen was also observed under different pH and temperature levels. The highest disease severity was observed at 30% soil moisture, soil pH 7.0, 35°C temperature and silty clay loam soil collected from Hapur. The seed germination, plant weight and root length were decreased in inoculated plants as compared to control plants. The maximum growth and sporulation of pathogen under *in vitro* conditions was maximum at pH 7.0 and temperature 25°C and no growth was observed under pH 4.5 and temperature 40°C. To evaluate the combined effect of different abiotic factors on disease severity and symptoms development the combination of soil type (sandy loam soil, silty clay loam soil and clay loam soil), Soil pH (8.44, 6.87, 8.02), soil moisture (saturation, 75% of saturation and 50% of saturation) and temperature (32°C and 35°C). Highest disease severity was observed in combination of 50% soil moisture, silty clay loam soil with pH 6.87 and 35°C temperature. Significant difference in disease severity was observed under different moisture conditions, different soil type with different soil pH and different temperature conditions. Significant variation in disease severity was observed in interaction of moisture and soil pH and soil type and soil type, soil pH and temperature. Non-significant variation in disease severity was observed in interaction of soil moisture and temperature and in interaction of soil moisture, soil pH, soil type and temperature. Elongation kind of symptoms were more prevalent in high moisture conditions, clay loam soil and 30°C temperature, whereas rotting symptoms were more prevalent in low soil moisture, high temperature and sandy loam soil. The regression analysis showed that soil moisture, pH and soil type were significant at 5% level of confidence.

An improved method for rapid isolation and quantification of *Rhizoctonia solani* from mungbean rhizospheric soil

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Rhizoctonia solani causes web blight/wet root rot disease of mungbean and is also the major constraint to mungbean crop production in India. An improved procedure for quantitative estimation of propagules of *Rhizoctonia solani* in soil was developed. Maximum number of colonies were observed in Ko and Hora medium (10) followed by water agar (8) following serial dilution after sieving through 0.150 mm sieve. The highly positive correlation (r = 0.96) was observed between disease incidence (%) and number of cfu/g of soil. In the q-PCR analysis for quantification and monitoring of *R. solani* isolates, the maximum level of inoculum was observed in the rhizospheric soil of mungbean genotype Pusa Vishal at 21 days after sowing. However it was reduced after harvesting in absence of mungbean crop. In conclusion, *R. solani* can be purely isolated and quantified by using Ko and Hora medium serial dilution after sieving through 0.150 mm sieve method and this methods was also found reproducible.

Theme area: Seed Pathology

Impact of Ascochyta blight of chickpea on seed quality

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Ascochyta blight of chickpea caused by *Ascochyta rabiei* L. is one of the most important seed borne disease with yield losses ranging from 10 to 100 % under severe natural epidemics infecting all above ground plant parts. Other than yield loss Ascochyta blight effects quality as well. So, it is important to study different seed quality parameters to study the impact of Ascochyta blight on chickpea seeds quality is important to study based on different seed quality parameter. Keeping in this view germination test, vigour index I, vigour index II and electrical conductivity test was carried out on healthy and disease affected seeds of different varieties of desi and Kabuli varieties of chickpea. The germination, vigour index I, vigour index II and electrical conductivity aspect of infected seed lot of desi chickpea range from 66-78%, 932.7- 1884.2, 13.1-20.59 and 19.87-74.96 respectively. And in case of Kabuli chickpea is Kabuli genotypes, the germination ranged between 62-75%, 607.9-1431.3, 12.32-23.76 and 23.42 to 82.84 respectively. The disease incidence was more in Kabuli genotypes when compared with Desi genotypes. Cultivation of two Desi genotypes namely ICCV 07110 and WR 315 and one Kabuli genotype namely L-550 should be avoided as minimum germination was found in them out of all genotypes evaluated. Similarly cultivation of two Desi genotypes namely WR 315 and JG 11 should be avoided as maximum electrical conductivity was recorded in these two genotypes. In Kabuli genotype ILC 212 and L 550 was recorded with maximum electrical conductivity and therefore farmers should avoid their cultivation.

Effect of heat stress on seed health status in wheat varieties (*Triticum aestivum L.)*

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Wheat is a major cereal crop of the world and one third of the world's population uses it as a staple food. Presently, heat stress is a major factor limiting wheat productivity which causes more than 30% yield loss and influences various processes including physiological, growth, developmental, yield and quality of wheat. The present study was conducted to study the effect of heat stress onseed health status of seeds harvested from both stressed and non-stressed environments. The field experiment was conducted during *rabi*2014-15 and 2015-16 in the field of the Division of Seed Science and Technology, ICAR - Indian Agricultural Research Institute (IARI), New Delhi, India. Eleven representative varieties from different zones were planted on three sowing dates viz. normal sown (mid-November, S_1), late sown (mid-December, S_2) and extremely late sown (early January, S_3) so as to study the comparative changes experienced in heat stress versus non-heat stress environment. The freshly harvested seed from all three sowings was evaluated for the effect on seed health parameters.The incidence of Karnal bunt was recorded as per Aujla *et al.*,1989 and spot blotch incidence as given by Saari and Priscott (1975). The incidence of Karnal bunt was recorded to be 5-10% under S_1 and S_2 whereas only 1-2% incidence was recorded under S_3. Thus, the late sown wheat varieties had lower incidence of the disease. The spot blotch incidence was recorded as 12DD (double digit) under S_1, 24 DD score under $S_2$and 34 DD score under S_3.Thus, higher incidence of spot blotch was recorded under late sown conditions. Hence, heat stress had a significant effect on the seed health status of wheat varieties.

Theme area: Emerging and Re-emerging Plant Diseases

Carbendazim tolerance in *Fusarium fujikuroi* isolates causing an emerging bakanae disease of rice in India

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Fusarium fujikuroi is an emerging disease causes different symptoms in bakanae disease of rice starting from pre-emergence seedling death to grain infection at maturity, elongation or stunted growth and foot rot causing plant death. Benzimidazole fungicides, which have systemic activity against a wide range of fungi, interact with tubulin (especially β-tubulin) and have been widely and extensively used to control bakanae but have generally lost their efficacy because of benzimidazole resistance. Studies have also stated the resistance in *F. fujikuroi* relating to β_2 *tubulin* gene. In this context, the present study was taken to identify the sensitive and resistance isolates of *F. fujikuroi* based upon their sensitivity against carbendazim (methyl-2-benzimidazolecarbamate (MBC) fungicide and conferring a means to molecular mechanism for the detection of the same using β_2 *tubulin* gene. Out of the one hundred and twenty-six isolates of *Fusarium* spp. earlier isolated from the symptomatic plants of basmati growing states of Punjab, Haryana, Uttar Pradesh and Uttrakhand in India, growth of 75 different isolates was checked using different concentrations of the fungicide in PDA (potato dextrose agar) until a concentration was identified where most of the isolates were sensitive while only few were resistant. The growth was not affected up to a concentration of 200µg/µl, but, thereafter, it slowed down till 350 µg/µl and was totally inhibited at 400 and 500µg/µl. Only 5 out of 75 isolates were found resistant. To study the gene expression pattern of carbendazim on resistant and sensitive isolates, qPCR primers were designed and qRT-PCR was performed which showed that the gene expression level decreased on addition of carbendazim fungicide, i.e., the ones isolated from carbendazim amended media gave less expression profiles compared to the sensitive . The study defines the sensitivity of *F. fujikuroi* isolates against cabendazim upto 400 µg/µl, and the presence of β_2 *tubulin* gene responsible for carbendazim tolerance in *F. fujikuroi* isolates.

Theme area: Role of Biotechnology in Managing Diseases Under Climate Change

An Intergeneric comparative secretomics as well as cross infection analysis between fungal phytopathogens *Bipolaris sorokiniana* and *Bipolaris oryzae*

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Spot blotch of wheat caused by *Bipolaris sorokiniana* and brown spot of rice caused by *Bipolaris oryzae* have resulted in great losses to food grain yield in all parts of the country especially in regions where rice-wheat cropping system is pre-dominant. Here, cross infection study under artificial conditions with most virulent isolates revealed that *when* inoculation of BS-112 of *B. sorokiniana* on wheat and rice was done, infection index of 89% and 80% was obtained, while BO-1(*B. oryzae*) inoculation showed infection index of 85% and 78% on rice and wheat respectively. Cross infectivity was confirmed by PCR using species specific markers developed in the laboratory. Synergism between two pathogens revealed infection index of 94% on wheat and 92% on rice, which was more than individual inoculations. Cross infectivity was also identified under natural conditions for both *B. sorokiniana* and *B. oryzae* on rice and wheat at certain places. A high level of similarity between these two closely related pathogens with respect to genes associated with host specificity as well as candidate effectors genes was identified through comparative bioinformatics. Comparative secretomics of *B. sorokiniana* and *B. oryzae* genomes led to identification of 262 and 247 predicted small secreted proteins (SSPs) respectively out of which 34 and 28 SSPs were assigned putative function in B2G analysis, which include alarge and diverse inventory of secretory proteins, transporters and primary and secondary metabolism enzymes. Only 39% of the predicted proteins were distinct between the two secretomes and 61% predicted SSPs were common for both the pathogens. Several of these predicted secretory proteins showed high cysteine content and numerous tandem repeats. Expression analysis of six selected unique and common SSPs to *Bipolaris* spp. showed that only 2 SSPs (BS _SSP2 and BOBS_SSP2) expressed as a typical fungal candidate effectors after inoculation on plant (*in planta*).Together, our analyses broadly expanded our knowledge and offered insights into the cross infection and pathogenesis of *Bipolaris* spp. in wheat and rice.

Arm race between pre-coat protein of geminivirus and RDR1 of *Nicotiana tabacum* for symptom development and recovery

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Leaf curl disease caused by begomoviruses is the major threat for the cultivation of solanaceous crops worldwide. Plants have adopted several mechanisms to fight against viral pathogens. RNA silencing is the most powerful plant machinery against virus infection. In the present study, pathogenesis of *Tomato leaf curl Gujarat virus* (ToLCGV) and *Tomato leaf curl New Delhi virus* (ToLCNDV) was investigated on *N. benthamiana and N. tabacum*. *N. tabacum* (cv. Xanthi) plants inoculated with ToLCGV resulted in symptom remission while infection of ToLCNDV led to severe symptoms on test plants. Hence, we aimed to elucidate the mechanism involved in this virus-specific symptom recovery, and the potential of ToLCNDV in inhibiting host-specific recovery phenomenon. Our result revealed enhanced level of *RNA-dependent RNA polymerase 1* (*NtRDR1*) in the recovered leaves of ToLCGV infected *N. tabacum plants*. Interestingly, *NtRDR1* expression level was reduced in ToLCNDV-infected leaves. Furthermore, the pre-coat protein (AV2) mutants of both the viral strains led to symptom recovery on *Nicotiana benthamiana*. We also found that ToLCNDV-AV2 is a strong suppressor of post transcriptional gene silencing (PTGS). Our results showed that, except AV2 mutant, inoculation with other mutants of ToLCNDV, developed severe symptoms on transgenic *NtRDR1* overexpression line of *N. benthamiana*, while only ToLCGV-inoculation resulted in the symptom remission. Our study unveiled that the pre coat protein of ToLCNDV is a pathogenicity determinant and strong suppressor of PTGS which blocks the recovery process through inhibiting the RDR1-mediated antiviral silencing in tobacco cv. Xanthi.

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