CHAPTER 7

BOTANY

Doctoral Theses

01. ARCHANA KUMARI
Nitric Oxide Mediated Modulation of Glucine Betaine Homeostasis, Aquaporin Expression, Ionic Balance and Proteome in Sunflower (Helianthus annuus L.) Seedlings Subjected to Salt Stress.
Supervisor: Prof. Satish C. Bhatla
Th 24349

Abstract
(Verified)

Sunflower (Helianthus annuus L.) plants are semi-tolerant to salt stress. Glycine Betainesynthesis is linked to onset of environmental stress conditions and its accumulation in plants directly signifies stress tolerance efficiency. Presence of various aquaporins isoforms and theirability to transport small solutes highlights versatility of plants in water regulation under normal growth conditions as well as under stress. Sensitivity or tolerance of plants to salt stress is also depicted through their ability to eliminate excess Na+ from cell cytoplasm. Recent investigation sunder taken in Delhi University has demonstrated prominent roles of melatonin and nitric oxide (NO) in alleviating negative impacts of salt stress on seedlings. Based on several facets of salt stress and roles of melatonin and NO under salt stress examined so far, present work aims todecipher modulation of Glycine Betaine accumulation and Betaine Aldehyde dehydrogenaseactivity and abundance by NO and melatonin in seedlings subjected to salt stress. Further more, possible role of NO in seedling growth and maintenance of water status in seedlings has alsobeen analyzed by determining the impact of NO on relative water content and aquaporins abundance and distribution under salt stress. NO-mediated modulation of sodium, potassium and calcium homeostasis in response to salt stress in seedling roots has also been investigated in order to ascertain the possible way through which NO-modulates ionic balance under salt stress.A comprehensive analysis of NO-modulated responses in seedling cotyledons has been vunder taken through proteomic (LC-MS/MS) approach under pharmacological treatments of NO donor and NO scavenger, to decipher the impact of NO in protein modulation in response to saltstress, and the metabolic tendency of seedling cotyledons to adapt to saline conditions provided. Thus, this work presents new information on NO and melatonin-mediated modulations of accumulation of Glycine Betaine, aquaporin expression, ionic balance and proteome in sunflower seedlings.

Contents

02. ARORA (Shaweta)

Towards Enhanced Production of Eicosapentaenoic Acid: Pathway Engineering in Oleferous Plants, Biochemical Modulation and Mutagenesis Approaches in Monodopsis Subterranea (Eustigmatophyte)

Supervisor: Dr. Girish Mishra

Th 24351

Abstract
(Not Verified)

Eicosapentaenoic acid is nutritionally important due to its cardio-protective, anti-inflamatory and anti-arthritis effects. Wild fishes, the main source of EPA is declining due to over-fishing for fish-meal and fish-oil. Metabolically engineered oil plants or microalgae with improved productivity can be a sustainable and cost effective alternative for EPA production. EPA synthesis pathway was heterologously expressed in A. thaliana using multigene constructs D15D_D6DA, EPA1, EPA2 and EPA3. While D15D-D6DAcylCoA has doubled the amount of ALA as compared to wild type, EPA1, EPA2 and EPA3 transgenic seeds produced 0.1 %, 0.8 % and 0.3 % of EPA respectively. EPA2, containing Δ6-desaturase-acyl-CoA from Ostreococcus tauri has increased the flux toward EPA. B. juncea transformants could not be obtained. Enhancement of EPA productivity was targeted while exploring effect of varying dosage of auxins (IAA, IBA and NAA) and cytokinin 6BAP on M. subterranea at three growth phases. IAA 100 µM exhibited 2.5-fold increase in EPA productivity at LE phase. PCA analysis finds inter- and intra-relation among different fatty acids yield due to dose-dependent treatment effect at three growth phases. PCA analysis depicted that both natural auxins (IAA and IBA) depict similar stimulatory action on biomass and PUFA. Evaluating the effect of varying concentration of GA, MeJA and MI on M. subterranea showed highest EPA productivity with GA 100 µM at ME phase. PCA analysis depicted a strong intra-relation between EPA yield and tetradecanoic acid yield. Strain improvement using UV-C mutagenesis was carried out on M. subterranea. Out of 220 mutants screened EPA and TFA productivities of 15 selected mutant lines were evaluated extensively. Five best lines showing 1.8-2.4-fold fold change in EPA productivity were treated with 100 µM GA as elicitor for maximal EPA productivity. EPA was enhanced to 6.4-fold and 6-fold in mutant line M192 and M220 respectively when compared to control.

Contents

1. Introduction 2. Metabolic engineering of VLC-PUFA pathway geneses in oleiferous crop 3. Biochemical modulation of monosopsis subterranean (eustigmatiophyceae) by auxin and cytolinin enhances enciosapentaenoic acid productivity 4. Effect of gibberellins methyl jasmonate and myo-inositol on eicosapentaenoic and productivity in monodopsis subterranean 5. Strain improvement of M. Subterranea by UV-C mutagenesis to maximize production of eicosapentaenoic acid 6. Summary . Appendices and list of publication and conferences .

03. JATIN KUMAR

Assessment of Genetic Diversity, Population Structure and Generation of Sex-Linked Molecular and Biochemical Markers in Some Multipurpose Dioecious Plants.

Supervisor: Prof. Veena Agrawal

Th 24354

Abstract
(Verified)

For the current investigations, three multipurpose dioecious plants, Simmondsia chinensis, Trichosanthes dioica, and Simarouba amara were selected for sex identification and genetic diversity studies. Genetic diversity and gene flow among different populations were observed to be high in different S. chinensis (Ht and Hs: 0.47 and 0.45, respectively) and T. dioica (Ht and Hs: 0.45 and 0.43,respectively) genotypes/accessions, and low in various S. amara (Ht and Hs: 0.21 and 0.16,respectively) accessions. S. chinensis genotypes of Gujarat region were highly diverse (h: 0.48) and that of Jaipur (h: 0.40) were least diverse. T. dioica populations of Bihar (h: 0.47) were highly diverse and Cuttack and Kolkata (h: 0.33) populations were least diverse. S. amara population of Gujarat (h: 0.20) was highly diverse, while Delhi (h: 0.11) population was least diverse. In S. chinensis and T. dioica, male plants were more genetically diverse as compared to females, while in S. amara, genetic diversity was similar in male, female, and bisexual plants. UPGMA, neighbour-joining, and STRUCTURE analysis divided S. chinensis population into two main clusters, T. dioica population into three main clusters, and S. amara population into two major clusters. For sex identification, one SRAP primer combination 'Em-14/Me-10' amplified a male specific marker of 396 bp in S. chinensis, and in T. dioica, one SRAP primer combination 'Em-6/Me-4' generated two male specific markers of approximately 230 and 290 bp in pooled and all the individual genotypes. In S. chinensis and T. dioica, male plants displayed better protective mechanisms than female counterparts and showed higher levels of enzymatic and non-enzymatic antioxidant activities. In S. amara, significant differences in various qualitative and quantitative characters of leaves/leaflets among male, female, and bisexual trees were observed which can be used for sex identification.

Contents

04. ARORA (Gauri Nee Gauri Joshi)
Comparative Genomics of MIR319 across Brassicaceae, Sequence and Functional Diversification of MIR319 Associated CIS-Elements from Brassica Juncea.
Supervisor: Dr. Sandip Das
<u>Th 24361</u>

Abstract
(Verified)

Polyploidization is a major force, and promotes evolutionary innovations through sub-functionalization, neo-functionalization and pseudogenization. It is known that core Brassicaceae members have experienced three whole genome duplications, with Brassica and Camelina an additional whole genome triplication; subsequent genome fractionations creating three distinct sub-genomes- Least fractionated (LF), Moderately fractionated (MF1) and Most fractionated (MF2).The influence of polyploidy on protein-coding genes is well characterized but not for MIRNAs that exist as gene family. Identical or near-identical mature miRNA sequence of family members also makes identification of source loci difficult based on expression pattern necessitating analysis of associated promoters. MIR319, one of the most conserved miRNA families found across a wide taxonomic spectrum is involved in various developmental processes and stress responses. Brassicaceae comprises of economically important oilseeds, condiments, vegetables and fodder crops. The genus Brassica includes six commercially cultivated species- B. rapa, B. nigra, B. oleracea, B. juncea, B.

napus and B. carinata. The availability of whole genome sequences for Brassicaceae members may allow us to understand the influence of polyploidy on genome architecture especially of MIRNA genes during evolution. In the present study comparative genomics using microsynteny analysis of 100 kb regions harboring MIR319a, MIR319b and MIR319c among different Brassicaceae species revealed differential gene loss/or gain, segmental rearrangements and duplications. Maximum effect of genome fractionation was observed in case of MIR319b block followed by MIR319a and MIR319c. Sequence and functional analysis of MIR319 promoter homologs/homeologs from B. juncea showed functional divergence with distinct spatio-temporal expression patterns. Deletion analysis of promoter sequences led to the identification of region that might play important role in its activity. Differential activities of MIR319 promoter homologs/ homeologs were observed in response to heat, cold, salt and ABA stress which could be attributed to the presence of various cis-motifs present on these promoter homologs/homeologs.

Contents

1. Introduction 2. Review of literature 3. Materials and methods 4. Microsynteny analysis of MIR319 to understand evolution and impact of polyploidization 5. Isolation and functional characterization of MIR319 promoter sequences from B. Juncea 6. In-silico analysis and effect of heat, cold, salt and ABA on various promoter homologs. Summary and conclusion. References. Appendices. List of publication and conference presentations.

05. KUKREJA (Bharti)

Small RNome, Degradome and Metabolome Analysis during SARIUS Oxysporum f. sp. carthami-Carthamus Tinctorius L. (Safflower) Interactions Reveal Molecular Components Involved in Plant Defense.

Supervisor: Prof. Manu Agarwal

Th 24360

Abstract
(Not Verified)

Carthamus tinctorius is an oilseed crop popularly grown in India, US, Mexico, Kazakhstan and Australia. Its demand in global market is growing rapidly owing to the MUFA and PUFA fatty acids present in Safflower oil. Safflower cultivation has seen a serious decline in recent years. Fusarium oxysporum f.sp. carthami (Foc), a soil borne fungus, is a major pathogen which can cause wilting in Safflower and reduce the crop productivity. In this study, Safflower germplasm was screened using hydroponics-based method to identify Foc-resistant and susceptible accessions. The sRNA profiling at 1, 2, 4 and 7 dpi of the Foc-resistant and susceptible Safflower accessions led to prediction of 47 conserved and 72 true novel miRNAs. Degradome sequencing resulted in identification of 204 high confidence targets for the conserved and novel miRNAs. Further, metabolic differences in resistant and susceptible Safflower accessions were studied using 1H-NMR. Out of many metabolites identified in resistant and susceptible accessions, syringic acid, a derivative of benzoic acid, accumulated to higher levels in the infected roots of the resistant as compared to the susceptible accession. Pretreatment of the plants with syringic acid conferred resistance against Foc, in the otherwise susceptible accession. Overall, many novel small RNAs and small metabolites were identified in the study, whose manipulation and exogenous application respectively can ameliorate the resistance of Safflower against F. oxysporum.

Contents

06. KASANA (Shruti)

Taxonomy and Phylogeny of the Saussurea Group (Asteraceae: Cardueae) in India.

Supervisor: Prof. P. L. Uniyal

Th 24357

Abstract
(Verified)

Saussurea DC. (sub-tribe Saussureinae, Cardueae, Asteraceae) includes approximately 493 species in the world. The genus has been split variously based on morphological and molecular preferences and is presently considered as an informal group due to unclear generic boundaries. In the present work, an attempt has been made to understand the systematic position of Indian Saussurea in the global phylogeny with extended sampling and a detailed taxonomic study of the genus in India. More than 300 names have been analyzed and updated their orthography, author citations, protologue information and designated type following the rules of ICN. As a result of the present work, the number of taxa accepted for the various segregate genera in the Saussurea group in India is 54 for Saussurea, three for Dolomiaea, one for Hemisteptia, two for Jurinea and 16 for Lipschitziella. A new taxon, Lipschitziella himachalensis has been described for India from Spiti valley, Himachal Pradesh; a new record to India, Saussurea pseudolingulata from Tungnath, Uttarakhand, and rediscovery of Saussurea andryaloides from Uttarakhand after 125 years and its new report for the flora of Sikkim. The taxonomic significance of achene micromorphology has been highlighted and with the help of molecular phylogenetic analyses, the monophyly of Saussurea and related genera has been ascertained. Four major clades were obtained from the phylogenetic analysis with high statistical support. The results suggested that Saussurea is monophyletic only after the exclusion of S. costus and the genus Himalaiella should be merged within Lipschitziella.

Contents

07. KRISHNAN (Remya)

Studies on Some Aspects of Diversity, Development and Reproduction in Riverweeds (Podostemaceae) of India.

Supervisor: Prof. Rajesh Tondon

Th 24352

Abstract
(Not Verified)

Podostemaceae is one of the most enigmatic aquatic families among angiosperms. In order to understand some of the aspects of diversity, development, and reproduction of riverweeds, a detailed investigation was undertaken by using standard microscopic and molecular techniques. Pollen ontogeny of Polypleurum munnarense revealed that organelle differentiation, number of amylogenesis and

amylolysis cycles and vacuolization pattern varies in accordance to the energy requirement of the developing microspores. The multistoried exine structure helps to prevent immediate desiccation through harmamogathy. Zeylanidium maheshwarii and P. munnarense (Podostemoideae) reproduce through spontaneous autogamy. The proximity of the stigma, the introrse anthers and compound pollen units (dyads) appear to facilitate self-pollination. Further female gametophyte development revealed the occurrence of inverted polarity of embryo sac in P. munnarense. This is intriguing in context of pollen-pistil interaction during final stages of pollen tube attraction. However, the inverted polarity does not affect the pollen tube attraction through micropyle. Phylogenetic analysis of the Indian podostemads using ITS and matK markers did not support the merger of Polypleurum stylosum with Polypleurum wallichii. On the bases of the orbicular and palynological features, the family can be divided into three major groups. Furthermore, morphological and molecular studies of a putative new species collected from river Moovatapuzha, Kerala, established the distinctness of the new species Zeylanidium manasiae sp nov. The reproductive attributes and and genetic diversity of Zeylanidium lichenoides confirms the occurrence of spontaneous autogamy among the selected population across Kerala and Maharashtra. AMOVA results suggest high genetic differentiation between the populations in contrast to low genetic differentiation within the populations.

Contents

1. General Introduction 2. Pollen ontogeny and exine development 3. Reproductive biology and female gametophyte developmet in Zeylanidium maheshwariiand polypleurum munnarense 4. An updated phylogenetic assessment of the Indian podostemacceae 5. To establish the taxonomic identity of new species from Kerala 6. Variation in reproductive attributes and genetics diversity of Zeylanidium lichenoides. Summary and conclusion. Literature cited. Illustrations. Annexures.

08. KEISHAM (Monika)
Biochemical and Proteomic Analysis of Salt Stress-Induces Changes in Sunflower (Helianthus annuus L.) Seedlings.
Supervisor: Prof. Satish C. Bhatla
Th 24358

Abstract
(Verified
)

Sunflower (Helianthus annuus L.) plants are moderately salt-tolerant in nature. Primary effects of salt stress on plants include ion-specific stress that lead to altered K+/Na+ ratios. Calcium ameliorates NaCl-induced Na+ toxicity by inhibiting non-selective cation channels (NSCC). Salt stress enhances ouabain (OU)-sensitive ATPase (putative Na+/K+-ATPase) activity in sunflower. NaCl also rapidly induces activation of phospholipase D (PLD). Past investigations in the author's laboratory have shown an interplay between melatonin and nitric oxide (NO) resulting in salt stress tolerance in sunflower. Melatonin enhances salt stress tolerance and mediates NO signaling and polyamine metabolism pathway. NO regulates ion homeostasis during salt stress tolerance in plants. Sodium nitroprusside (SNP) has so far been the most extensively used source of NO in life science research. However, it also simultaneously releases free iron and cyanide, which limits its usefulness as an NO donor. With this background information in view, present investigations aim at understanding the metabolic and enzymatic modulation of Na+, K+ and Ca2+ homeostasis in response to salt stress and pharmacological treatments of melatonin and NO donors to the seedlings. Furthermore, a probable crosstalk of PLD and NO that could occur in seedlings as a salt stress tolerance mechanism, is being analyzed by investigating the modulation of PLD activity in response to salt stress in seedling cotyledons and roots. A thorough analysis of SNP-modulated growth responses in sunflower seedlings vis-a-vis other established NO donors through pharmacological, proteomic and biochemical approaches has been undertaken to decipher the NO, cyanide and iron-mediated actions of SNP in cotyledons. Thus, the present work presents new information on the modulation of ionic fluxes by melatonin and NO, and signaling role of PLD in salt tolerance in sunflower seedlings. It also provides evidence to demonstrate that SNP action in plants is being mediated by NO, cyanide and iron.

Contents

1. Introduction 2. Review of Literature 3. Materials and methods 4. Results 5. Discussion. Summary. References. Research Publications.

09. PAWAN KUMAR

Consequences of Reciprocal Host-Plant Switches among Garden Nasturtium, Tropaeolum Majus L. and Late Season Cauliflower, Brassica Oleracea L. var. Botrytis on Life History, nutrition and Larval Gut Serine Protease Gene Expression in the Lepidopteran Pest, Pieris Brassicae L.

Supervisor: Prof. Sudeshna Mazumdar-Leighton

Th 24364

Abstract

(Verified)

Pieris brassicae L. is a global pest of crucifer crops. A recent study showed that early season crucifers (especially cabbage and cauliflower) were superior host plants for supporting larval growth as compared to T. majus (Kumar et al., 2015). To investigate use of T. majus by P. brassicae larvae coincident with late season of crucifer crops in north India, host-plant switches between cauliflower and T. majus were examined. Field and lab experiments indicated that larvae feeding continuously on T. majus matured faster than insects feeding on cauliflower suggesting benefits related to early eclosion. Estimation of nutritional indices revealed rapid, adaptive responses of larvae to changes in diets. Levels and properties of gut proteases detected in different instars were influenced by type of diet consumed. Enhanced gut trypsin and chymotrypsin activities were implicated in larval adaptations to a T. majus diet. Larval gut serine proteases were inhibited to varying extent by trypsin inhibitor activities detected in herbivore-induced and un-induced leaves of host plants. This study reports for the first time, identification of a Kunitz trypsin inhibitor from seeds on T. majus (TPTI-S) suggesting a large trypsin inhibitor gene family expressed differently in time and space. At least ten lineages of serine protease genes were identified from gut tissues of P. brassicae (GenBank accessions MK376898-MK376910). Sequence analyses of a large dataset of P. brassicae cDNAs and orthologs from phytophagous Lepidoptera emphasized diversity and complex structure-function relationships of encoded serine proteases. Transcripts encoding diverse trypsins and chymotrypsins were up-regulated in larvae switched to T. majus. No significant differences were discernable in levels of nsp gene transcripts (involved in detoxification of ingested glucosinolates) in larval guts. Gut serine proteases of P. brassicae play major roles in adaptation to host plant switch and utilization of unsuitable hosts like T. majus.

Contents

1. Introduction and review of Literature 2. Materials and methods 3. Results and discussion 4. Results and discussion. Summary and conclusion. References. Annexure I. Annexure II. Publication.

10. SAZDA ABDI

Development of EST-SSR and Intron Length Polymorphic Markers for Genetic Linkage Mapping of Apospory Specific Genomic Region in Cenchrus Ciliaris L. and Assessment of Genetic Variation among Cenchrus and Pennisetum Species and their Allies.

Supervisor: Prof. Vishnu Bhat

Th 24353

Abstract
(Verified)

Apospory specific genomic region in Cenchrus is very large and towards map based cloning of genes controlling apospory, F2 mapping population was developed earlier and to increase recombination events around ASGR, F3 population which segregated for apomixis and sexuality was developed. A total of 90 individuals were characterized for their modes of reproduction which indicated 46 obilgate apomictic, 20 obligate sexual and 24 facultative apomictic types. F3 individuals were screened using already developed seven SCAR markers associated with apomixis and one SCAR associated with sexuality. Male parent-specific SSR alleles were used to further characterize 53 F3 individuals which were used for genetic linkage mapping of ASGR. 116 polymorphic SSR markers derived from transcriptome sequences of C. ciliaris were identified in a test panel and were later used to map 56 F2 and 53 F3 individuals. In addition, one ILP marker was also identified which was closely linked to apomixis. Mapping was attempted only in F3 population. Due to a complex segregation pattern observed among F2 individuals, genetic linkage mapping was attempted for F3 population. While markers grouped into 3 linkage groups, LG1 containing 'Apo' locus was 28.3 cM long with 11 markers out of which seven markers grouped into one Apo locus. Apo locus was flanked by CCSSR_4 and CCSSR_59 at a map distance of 1.9 cM and 8.7 cM respectively. Polymorphic SSR markers were further used to asses genetic variation among 33 accessions belonging to 21 species of Cenchrus-Pennisetum and their allies. Five SSR markers showed 100% cross-species transferability across 21 species. Unique agamic complex specific alleles were also identified for Cenchrus-Pennnisetum and Bothriochloa-Dichanthium complexes. NJ-based clustering showed grouping of accessions based on mode of reproduction both in Cenchrus and Pennisetum species. Phylogenetic analysis using polymorphic SSRs indicated monophyly for Cenchrus-Pennisetum species.

Contents

1. Background 2. Development and characterization on an advance F_3 mapping population in cenchrus ciliaris 3. Development of EST-SSR and Intron Length Polymorphic Markers for Genetic Linkage Mapping of Apospory Specific Genomic Region in F_3 population 4. Assrssment of generic variation among apomictically & sexually reproducing species of cenchrus, pennisetum. Summary. References. List of publication.

11. SHARMA (Bhavana)

Comparative Proteome amd Lipid Profiling of Indian Seabuckthorn for Understanding its Stress Tolerance and Nanobiotechnological Applications

Supervisor: Prof. Renu Deswal

Th 24363

Abstract
(Verified)

Climatic fluctuations due to global climate change affect biodiversity and survival of high-altitude plants. Seabuckthorn (Hippophae), a stress tolerant Himalayan plant is able to withstand extreme environmental conditions. Proteomic analysis to understand its stress tolerance is limited to laboratory grown seedlings. Comparative gel-based and gel-free shotgun proteomics approach was used to dissect stress acclimation strategies in high-altitude (3200-3530 masl) adaptedTrans-Himalayan (H. rhamnoides, H. tibetana) and lower altitude (2500-3111 masl) adapted Sikkim (H.salicifolia) germplasm. A custom-built database (Hippophae+Viridiplantae) was designed for a better annotation of proteins. nanoLCMS/MS analysis identified 1035 protein groups showing differential abundance (DAPs) of metabolic, regulatory and stress responsive proteins in Trans-Himalayan and Sikkim germplasm indicating their probable involvement in differential stress acclimation. KEGG analysis showed their association with metabolic processes, defense responses, redox regulation and secondary metabolite or fatty acid biosynthesis. Validation of downstream metabolic signatures supported proteomic plasticity and justified stress responses. Higher (1.3-2.7 folds) antifreeze activity showed a correlation with higher (1.15-7 folds) abundance of antifreeze proteins (AFPs) in Trans-Himalayan germplasm. Ice-affinity and chitin affinity chromatography allowed homogenous purification of leaf and berry AFPs. Interaction of these with proteins involved in defense and abiotic stress suggested their role in stress acclimation. These AFPs could be utilized as stress tolerance markers for crop improvement. Interestingly, higher (1.2-6 folds) abundances of rate-limiting enzymes of fatty acid and phenylpropanoid pathway complemented higher degree of unsaturation flavonoids in Trans-Himalayan germplasm. Meanwhile, higher (1.5-8 folds) phenolic content, antioxidant potential and unique composition of essential fatty acid (ω-6:ω-3 ratio 4:1 to 1:1) in Sikkim germplasm indicates its better nutraceutical potential. Additionally, gold nanoparticles synthesized from Trans-Himalayan (H. rhamnoides) bioresource allowed efficient remediation of dye-contaminated waste water into non-toxic byproducts with industrial applications. This is first Proteomics analysis dissecting altitudinal gradient associated stress acclimation strategies and nutraceutical variations in naturally growing seabuckthorn.

Contents

12. SHARMA (Shruti)

Characterizing Dioscorea Alata Tuber Proteome for Molecular, Biochemical and Redox Alterations Across different Developmental Stages.

Supervisor: Prof. Renu Deswal

Th 24362

Abstract

(Verified)

Dynamic population growth has led to an ecological disequilibrium resulting in food security concern. Dioscorea commonly called as yams has been explored for its medicinal relevance, while its genomics and proteomics remain relatively unexplored. An integrated proteomics (gel based and gel free) and biochemical approach was adopted to understand the physiological role of dioscorin (major storage protein, 85%) across different developmental stages of the tuber growth. The tuber life cycle was dissected on morphological basis into four distinct stages validated by principal component analysis (PCA). SDS-PAGE and 2-D maps confirmed more than 50% depletion of dioscorin on germination which was further validated using gel free approach. A comprehensive data set was generated using gel free quantification identifying a total of 3,681 proteins. The over-representation analysis using PANTHER and KEGG MAPPER revealed a high abundance of the enzymes of ascorbate-glutathione cycle, carbohydrate metabolism, glycolysis, Tricarboxylic acid (TCA) cycle during tuber degradation and formation. Novel transcription factors associated with oxidative stress tolerance were also identified. An operational Asada- Halliwell cycle was established by analyzing all the component enzyme namely, ascorbate peroxidase, glutathione reductase, dehydroascorbate reductase, mono-dehydroascorbate reductase and end products, ascorbate and glutathione of the cycle. An overall shift in the oxidative conditions during tuber maturation (formation) to reducing condition during its germination (degradation) was established. Higher nitric oxide and S-nitrosothiol content also supported the oxidative conditions during tuber formation. The differential redox conditions also modulated the associated activities of dioscorin suggesting the physiological relevance of these in the tuber. These activities could serve as biomarkers for tuber degradation. In conclusion, the data set comprehensively describes the proteome of Dioscorea tuber that would pave the way for its systematic study that may influence the direction of research for improving the productivity of this under-utilized crop.

Contents

1. Introduction 2. Review of Literature 3. Materials and methods 4. Results and discussion. Summary and conclusion. References. Appendix. List of publications.

13. SINGH (Amrita)
Structural and Functional Geonic Approaches towards Delineating the Mechanism of Phytoplasma Associated Retrograde Metamorphosis during Phyllody.
Supervisor: Prof. Suman Lakhanpaul
<u>Th 24359</u>

Abstract
(Not Verified)

Sesamum indicum (L.) is cultivated for its oil and nutritional seeds. Sesame oil has remarkable antioxidative property. India is the largest producer of sesame seeds in the world. 'Phyllody' is one of the major biotic stresses severely affecting sesame production. It is associated with 'phytoplasma' ,a phytopathogenic bacteria that hijacks host plants by secreting effectors, which modulate plants' development and defense systems. The 'Introduction' part of the thesis provides a compendium of previous research. The first chapter deals with detection of SAP54 ortholog from sesame and analysis of selective forces shaping the evolution of SAP54. The second chapter involves identification of plant proteins that interact with SAP54 ortholog from phytoplasma affected sesame. The third chapter details the studies on epigenetic changes in sesame plants in response to phytoplasma infection. A detailed account of the presence and distribution of putative G-quadruplexes in phytoplasma genomes highlighting their role in pathogenicity is dealt in Chapter 4. The results have been discussed separately for each chapter. The salient findings have been summarized in the 'Summary and Conclusions' part. The study aimed at studying the evolution of the phytoplasma effector in phyllody affected sesame provides an understanding of the functionally important regions of the protein. This knowledge further extended by identification of host proteins that interact with phytoplasma effector protein would provide an insightful platform to dissect the mechanism of action of phytoplasma effectors. The epigenetic study focusing on differential cytosine methylation of genes and transposable elements in response to phytoplasma infection would help in developing mechanisms targeting genes involved in developmental alterations. The information on the predominance of G-rich sequences at specific locations in the genome of phytoplasma and their in vitro potential to form G-quadruplexes can be harnessed to target these structures employing antimicrobial therapy.

Contents

1. Introduction 2. Detection, characterization and evolutionary aspects of SS54Lp (SAP54 like protein) – a phytoplasma effector molecule associated with phyllody development in sesame (Sesamum indicum L.) 3. Identificationb of interacting peartners of Onion yellow phytoplasma (strain OY-M) effector protein S54LP in sesamum indicum (L.) using library scale yeast two hybrid (Y2H) approach 4. Comparative methylome analysis of healthy (H1) and phytoplasma affected (II) sesammum indicum (L.) plants 5. Results 6. Discussion. Summary and conclusion. References. Publications. Presentation in national and international conferences.

14. SINGH (Neeraja)
Assessment of Viru;ence and Genetic Diversity in Fusarium Oxysporum Schlecht. F. Sp. Carthami Klis. & Hous. Isolate in India and Development of a Scar Marker for its Detection in Safflower Seedlings and Soil.
Supervisor: Prof. Rupam Kapoor
Th 24356

Abstract
(Not Verified)

Safflower wilt, caused by Fusarium oxysporum f. sp. carthami (Foc) is a major limiting factor for safflower production worldwide. In India, the disease was diagnosed in 1975 in Maharashtra, since then the disease has been reported from all safflower growing regions. Surveys were conducted in 22 districts of Karnataka and Maharashtra (2015-16 and 2016-17). Among all the districts, Dharwad (Karnataka) exhibited maximum mean disease prevalence (71%), incidence (41.4%), and severity (52.4%). The disease prevalence was less in late sown fields as compared to early sown. Intercropping with chickpea and sorghum together decreased disease prevalence in comparison to monocrop fields. To assess variations among Foc, 90 isolates were collected from four states (Maharashtra, Karnataka, Telangana, and Madhya Pradesh) in India. Isolates showed high variation with respect to macroscopic, microscopic, and biochemical characteristics. Among different fungicides tested, the isolates were most sensitive to propiconazole. Virulence analysis also demonstrated high variation and indicated the existence of six races. Out of 90 RAPD (random amplified polymorphic DNA) and ISSR (inter simple sequence repeats) primers screened, 34 primers revealed a comprehensive picture of genetic diversity. Cluster and principal coordinate analyses distributed isolates from a district in different clusters and quadrants, respectively, indicating high genetic diversity. For specific and sensitive detection of Foc, SCAR (sequence characterized amplified region) marker was developed. The SCAR primer (FocScF/FocScR) specifically amplified a fragment of ≈ 213 bp in all the 75 Foc isolates, but not from non-Foc isolates. The primer set was able to detect 10 pg of Foc genomic DNA using conventional PCR, while SCAR primers when coupled with real-time qPCR demonstrated detection limits of 1 pg for genomic DNA and 1000 conidia/g for soil. The assay enabled reliable diagnosis of Foc DNA in contaminated safflower fields and expedited Foc detection at 72 hours post inoculation in asymptomatic seedlings.

Contents

1. Introduction 2. Review of Literature 3. Survey of fusarium wilt disease of safflower in Maharashtra and Karnataka 4. Assessment of variation among f. oxysporum f. sp. carthami isolate in India 5. Development of as scar marker for unambiguous and sensitive detection and quantification of f. Oxysporum f. sp. Carthami in asymptomatic safflower seedlings and soil. Summary and conclusion. References. Research publications. Posters/ conferences.

15. SHIVANGI

Enhancement of Omega-3 Fatty Acid production in Three Selected Microalgae through Bio-Prospecting, Biochemical Modulation and Transcriptome Studies.

Supervisor: Prof. Dinbandhu Sahoo

Th 24350

Abstract
(Not Verified)

Microalgae are widely sought for their valuable biochemical compounds and potential for sustainable production of essential polyunsaturated fatty acids (PUFAs). Present study aimed at isolating and characterizing omega-3 fatty acid rich microalgal species and to enhance their physiological potential for omega-3 PUFA production under mixotrophic conditions. Two ω-3 fatty acid containing microalgae were isolated from Dal Lake and identified as Tetradesmus sp. DLK-1 and Desmodesmus sp. DLK-2. Analysis of fatty acid methyl esters shows a high content of ω-3 PUFAs accounting for 67.6 % and 65.6 % of total fatty acids in Tetradesmus sp. DLK-1 and Desmodesmus sp. DLK-2 respectively. Effects of carbon supplementation and nitrogen starvation on Desmodesmus sp. DLK-2 under mixotrophic condition were evaluated. A 12.8-fold and 11.4-fold increment in biomass productivity and 22-fold and 26-fold enhancement in lipid productivity were observed in cultures supplemented with glucose and glycerol, respectively, as compared to control. N-starved culture with glucose increased the lipid content by 29.6 % and TFA by 2.9-fold. Carbon supplemented and N-starved cultures contained 68-78 % and 77-91 % of saturated fatty acids and monounsaturated fatty acids respectively. Effects of supplementation with different carbon and nitrogen sources on EPA productivity in Monodopsis subterranea showed highest biomass and EPA productivity which was 49.3-fold and 18.2-fold higher when supplemented with glu 1 % + gly 12 g.L-1 compared to control. Under urea supplementation biomass productivity and EPA productivity increased by 1.8-fold and 2.4-fold when compared to photoautotrophic culture, respectively. A de novo transcriptome sequencing for M. subterranea was carried out, adding to the information of molecular data available for class Eustigmatophyceae. A total of 35,954 transcripts were obtained through final transcriptome assembly. Various fatty acid desaturases and elongases involved in PUFAs biosynthesis were identified along with transcription factors and simple sequence repeats.

Contents

1. Introduction 2. Isolation, identification and biochemical characterization of freshwater green microalgae rich in omega-3 fatty acids 3. Enhancing biomass, lipid content and fatty acid composition of green microalgae desmodesmus sp.DLK-2 under mixtrophic conditions. Summary. Appendices. List of publications and conferences.

16. TAILOR (Aditi)

Polyamine Homeostasis and Salt Stress-Mediated Changes in Proteome, Ionic Balance and Aquaporin Expression in Sunflower (Helianthus Annuus L.) Seedlings.

Supervisors: Prof. Satish C. Bhalta and Prof. Rajesh Tondon

Th 24348

Abstract
(Verified)

Sunflower (Helianthus annuus L.) is a major oilseed crop and the major sunflower producing states in India are affected by salt stress, a major

environmental constraint on world agriculture. In view of all the investigations undertaken so far on various facets of sunflower salt stress physiology in Delhi University (c/o Prof. S. C. Bhatla), present work is aimed at investigating the regulatory roles played by polyamines as yet another important aspect of salt stress tolerance in sunflower. Alterations in the endogenous titres of the three major polyamines (PAs), i.e. putrescine, spermidine and spermine, and modulation of the activity and abundance of PA biosynthetic and catabolic enzymes have been examined in response to salt (120 mM NaCl) stress and nitric oxide (NO) donor treatment to decipher a possible interplay between NO and PA metabolic pathways, particularly with regard to salt stress. Physiological and biochemical changes, including alterations in hydration status and ionic balance, and modulation of expression and accumulation patterns of plasma membrane and tonoplast associated aquaporin isoforms have been analyzed in seedlings in response to salt stress and PA biosynthesis inhibitors. A possible existence of aquaporin isoforms on oil body (OB) membranes and modulation of their accumulation pattern by salt stress and PA inhibitors have been elucidated. LC-MS/MS analysis has been conducted to assess the impact of salt stress and PA depletion on proteome modulation and to decipher probable players in PA-mediated adaptive responses in sunflower seedling cotyledons. Thus, present work provides new information highlighting the significance of PA homeostasis on seedling growth under salt stress, and elucidates a complex interplay between PA homeostasis and other metabolic pathways which, in turn, might be crucial for PA-mediated adaptive responses in sunflower seedling roots upon sensing of salt stress, and in seedling cotyledons, as a long distance signaling response to salt stress.

Contents

17. TOMAR (Urvashi)
Soil Carbon Sequestration, Microbial Acticity and Nutrient Dynamics in Semi-Arid Ridge Forest Ecosystem of Delhi.
Supervisor: Dr. Ratul Baishya
Th 24355

Abstract
(Verified)

Understanding the dynamics of soil carbon (C) is crucial for assessing the soil carbon storage and predicting the potential for mitigating carbon dioxide from atmosphere to biomass and soil. The aim of the present research was to see the seasonal variation in: Soil carbon stock and soil carbon sequestration in the ridge forest ecosystem. Soil respiration, enzyme activity and microbial biomass carbon. Soil nutrient stock and nutrient release dynamics during decomposition. The highest SOC stock was observed during winter (22.81 Mg C ha-1) and lowest during monsoon (2.34 Mg C ha-1) in all the four ridges ($p<0.05$). We observed an increased annual SOC stock and decreased (or same) SIC stock under both the depths. SOC sequestration rate ranged between 0.046 to 0.741 Mg C ha-1yr-1. Significant variations were observed in MBC, soil enzymes and soil respiration with respect to seasons and depth. MBC ranged from 17.08 µg C g-1 to 484.51 µg C g-1. Similarly, β-glucosidase, dehydrogenase and phenol oxidase activity ranged from 11.15 to 212.59 µgPN gDW-1h-1, 0.11 to 16.47 µgTPF gDW-1h-1 and 3914.30 to 10187.55 µmolABTS+ gDW-1min-1, respectively. Soil respiration was observed highest in monsoon (3.39 µmolCO2 m-2sec-1) and lowest in winter (0.69 µmolCO2 m-2sec-1). Soil Total Nitrogen, Available Phosphorus and Exchangeable Potassium ranged from 0.41 to 2.51 Mg ha-1, 12.04 to 24.24 kg ha-1 and 100.98 to 280.36 kg ha-1, respectively. The leaf litter decomposition

rate constant during 3 years of study was estimated as 0.509. The estimates of C sequestration rate in this study provides a realistic image of dynamics of C in the present climatic conditions in semi-arid forests and could be used in generating database and formulating new strategies for carbon dioxide mitigation by enhancing soil C sequestration rate in forests.

Contents

1. Introduction 2. Review of literature 3. Materials and methods 4. Site description 5. Soil carbon stock and carbon sequestration in the ridge forest ecosystem 6. Soil respiration , enzyme ecticity and microbial biomass carbon 7. Soil nutrient stock and nutrient release dynamics during decomposition. Summary and conclusion. References. Publications.

M.Phil. Dissertations

18. ANSHU
Effect of Bryophytes on Soil Physico-Chemical and Biological Properties and their Role in Biomonitoring of Heavy Metals in Temperate Forests of Garhwal Himalaya.
Supervisor: Dr. Archana Kaushik

19. ARYA (Atul)
Ethnobot Anical Survey of Indigenous Medicinal Plants of Palampur, Himachal Pradesh, India.
Supervisor: Dr. Suresh Kumar

20. NEHA
Sequence and Functional Characterization of Regulatory Elements involved in Development and Stress Responses from Brassica.
Supervisor: Dr. Sandip Das

21. POOJA
Phylogenetic Position of Fusarium Oxysporum Schecht f.sp. Carthami Klis. and Hous. In F. Oxysporum Species Complex on the Basis of Conserved Market Genes and Presence of Secreted in Xylem Genes.
Supervisor: Prof. Rupam Kapoor

22. YADAV (Kailash)
Detecting Antifreeze Activity in the Cuticle Proteins of Brassica Juncea, Brassica Nigra. Brassica Rapa & Raphanus Sativus.
Supervisor: Prof. Renu Deswal