

Shalaka Desai

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Background in botany, plant physiology, plant terrestrial ecology, proteomics, molecular biology.

Qualification Summary

- 8+ years of experience in Biological Sciences with specialization in molecular biology and plant physiology
- Experienced molecular biologist with 10 years of professional research, teaching, managing, and technical experience
- Good working knowledge and experience in different methods and technologies in molecular genetics, including tissue culture, cloning, protein expression and purification, differential gene expression, microscopy and scanning microscopy.
- Able to develop novel molecular, biochemical, histological, and other analytical/quantitative assays and technologies, experience in training others in these techniques
- Extensive experience in managing a molecular genetics research lab and supervising students, technicians, and assistants; management of personnel, materials, equipment, and funds in laboratories as well as business/industry setting

Education

PhD– Biology, West Virginia University, Morgantown, WV, US- (Aug'12) G.P.A – 3.82

MS – Botany Major in Plant Physiology, Pune University, Pune, India - (Apr'02)

BS – Botany, Pune University, Pune, India – (Apr'00)

Selected Skills

Molecular Biology

- PCR, RT-PCR, primer construction
- Gel electrophoresis
- 5' and 3' RACE
- Plasmid preparation, cloning
- cDNA synthesis
- polyA+ RNA selection
- DDRT-PCR
- RNA and DNA isolation
- Manual and automated DNA sequencing

Protein Biochemistry

- Protein Isolation
- 2D gel electrophoresis
- HPLC
- Enzyme Assay
- Scanning Electron Microscopy
- AA spectrometry, LI-COR (LI-6400XT)

Bioinformatics

- GenBank searching: BLAST
- Multiple sequence alignments
- Tree building using PHYLIP
- Protein sequence analysis
- SEQUEST

Research Experience

Postdoctoral Appointee, Biosciences Division, Argonne National Laboratory

Oct' 12 – present

Molecular Mechanisms Mediating Environment Sensing and Response

- The Argonne “Environment Sensing and Response” Scientific Focus Area (SFA) program will identify the molecular basis of cellular transport and sensory pathways that mediate the response to environmental nutrients. Our limited understanding of the function and biological role for transporter and sensor domain proteins is a principal impediment to defining nutrient exchange processes and signaling pathways that mediate response to environmental ligands and ecosystem changes. This program will address this knowledge gap by mapping transport and sensor proteins to specific environmental compounds to define their function and biological roles and establish a series of defined connections between the environment and the cell. The knowledge will facilitate the development of system-level models predictive of cellular response to environmental conditions or changes. In addition, the molecular function information derived from these studies will enable synthetic biology approaches that modulate the system response by manipulating components of transporter or proteins containing sensor domains. A key component of the proposal is the re-engineering of specific transport proteins or complexes to modify the system response to phosphorous, nitrogen, or carbon sources in a predictable manner. The increased knowledge of function, regulation, and system response will support the Genomic Sciences Program goal of achieving a genome-based, dynamic systems-level understanding of organism and community functions

Graduate Research Assistant, Department of Biology, West Virginia University

August'06 – Aug'12

Genetic and physiological changes in poplar during mycorrhizal colonization under phosphorus limitation

- Phosphorus (Pi) is a major mineral nutrient, required for plant metabolism. It also forms a major component of sugar

phosphate, phospholipids and nucleic acid. Due to its biochemical property with elements such as calcium, iron and aluminium depending on soil pH, availability of Pi becomes more limiting in forest ecosystem. Plants undergo stress due to this mineral limitation. To overcome this limitation plants form symbiotic association with mycorrhizal fungi.

- In this dissertation physiological, genomics, proteomics, ecophysiological and biochemical approaches were used to understand underlying mechanism under phosphorus limitation during mycorrhizal colonization.

1) To study the effect of phosphorus starvation on the physiology of poplar during ectomycorrhizal colonization

- To understand the physiology of poplar under phosphorus stress during ectomycorrhizal colonization *P.tremuloides* were grown at different Pi levels (0, 1, 5, 10, 25, 50 & 100 μM) colonized with and without ectomycorrhizal fungi i.e *Laccaria bicolor*.
- Completely randomized block design with 7 Pi treatments, 2 mycorrhizal treatment and 10 replicate distributed in 5 blocks were grown for 46 days in control green house conditions.
- Parameters measured were biomass, organic acid exudation, PEPCase activity, antioxidant enzyme activity such as CAT, APX, GDP, SOD and Zn, Cu-SOD and tissues mineral concentration.
- Under Pi limitation growth declined and root: shoot ratio increased. Pi uptake rate was higher in mycorrhizal poplar root and leaf tissue compared to nonmycorrhizal poplar tissue. Antioxidant activity and PEPCase activity was higher in nonmycorrhizal poplar tissue than mycorrhizal poplar tissue under Pi limitation.
- Organic acid exudation in response to Pi limitation was more pronounce in non-mycorrhizal poplar.
- It can be inferred that mycorrhizal poplar have efficient Pi uptake and this maintains plant homeostasis under Pi limitation hence mycorrhizal poplar undergo lower oxidative stress compared to non-mycorrhizal poplar.

2) Investigate the transcriptional responses of poplar during symbiotic interaction with ectomycorrhizal and arbuscular fungi

- *P tremuloides* colonized with ectomycorrhizal fungi i.e *Laccaria bicolor* and *Paxillus involutus* and arbuscular mycorrhizal fungi i.e *Glomus intraradice* where grown under Pi limitation ((5 & 100 μM) to study differentially expressed transcriptome in poplar roots.
- mRNA was isolated and double stranded cDNA was synthesized using reverse transcriptase. Further Cy3 incorporation hybridized to NimbleGen poplar microarray analysis. Image and raw data was produced and quantile normalized.
- Plants roots forming symbiotic associations showed changes in gene expression pattern .Several genes were differentially expressed (up-regulated or down-regulated) due to the transport and/or processing of mineral nutrients and carbon between these symbiotic partners.
- Differences in plant host genes that were differentially expressed due to mycorrhizal colonization by ECM and AM fungi. Difference in colonization and morphological difference in the root architecture were evident in gene expression pattern.
- Phosphorus limitation also changed physiology of the aspen which was reflected in gene expression pattern .Genes related in phosphorus starvation were differential expressed and differ in that of mycorrhizal aspen.

3) To study differentially expressed protein profiles in poplar colonized by mycorrhizal fungi

- *P tremuloides* colonized with ectomycorrhizal fungi i.e *Laccaria bicolor* and *Paxillus involutus* and arbuscular mycorrhizal fungi i.e *Glomus intraradice* where grown under control green house condition to study differentially expressed protein profiles in poplar roots.
- Total protein was isolated from poplar roots and separated using 2-D gel electrophoresis, images analysis was done using PDquest and proteins differentially expressed were excised and digested using trypsin. Subjected to mass spectrometry and peptide sequence were identified using SEQUEST.
- The greatest change in protein accumulation was in the area of energy production and carbon metabolism (39.0%). Mycorrhizal also changed the abundances of proteins involved in metabolism and defense. Both ECM species showed similar changes in functional groups, but differences in specific proteins.
- Protein expression in *P. tremuloides* host plant differed in both ecto and arbuscular mycorrhizal colonization.

4) To investigate changes in photosynthesis, growth, and carbon biochemistry of poplar ectomycorrhizal association under phosphorus limitation

- *P tremuloides* colonized with *Laccaria bicolor* and *Paxillus involutus* where grown under two Pi levels (5 & 100 μM) to investigate changes in photosynthesis, growth and carbon biochemistry.
- This was completely randomized block design with 2 Pi treatments, 3 mycorrhizal treatment and 10 replicate distributed in blocks grown for 46 days in control green house conditions. In this experiment parameters measured were biomass, photosynthetic measurement using LICOR LI-6400XT Portable Photosynthesis System, carbohydrates using enzyme assay, secondary metabolites using HPLC and tissues Pi concentration.
- Photosynthetic parameters (gas exchange and biochemistry) were affected due to Pi limitation. Soluble sugars and carbohydrates were reduced due to Pi stress. Condensed tannins and total phenols and other glycosides increased due to Pi limitation, with changes most evident in non-mycorrhizal poplar.
- Mycorrhizal fungi had affects on photosynthetic parameters, carbon allocation, and reducing the effects of Pi limitation.

- Ectomycorrhizal species behave differently during symbiotic association with same host plant.

Research Assistant, National Chemical Laboratory, Division of Plant Molecular Biology, India May '02 –Nov'04

Resolve plant species complex using molecular markers

- *Trigonella emodi* complex and other related complex of genus *Trigonella* were considered for resolving species complexes. Morphological similarities within these species have been of considerable interest and debate. The genus has gained importance due to its medicinal importance and endangered species status.
- To solve this query Random Amplified Polymorphism DNA markers (RAPD) and Inter Simple Sequence Repeat markers (ISSR) were used. But the gene tree does not necessarily represent the true species tree because of random sorting of polymorphic alleles in different lineages. Cleaved Amplified Polymorphic Sequence (CAP's) method was used. Discovery of underlying phylogenetic relationship of organisms inhabiting these areas can lead to an increased understanding of their evolution and diversification. Locus specific primers such as chloroplast (five chloroplast genes), mitochondrial and Rbc's genes primers were used. For most species fresh tissue could not be obtained so herbarium specimen was used. Taxa considered were 22. Variation in band size was observed in amplified product of locus specific primers. Further work is to be done.

Genetic Diversity within the species *T.foenum-graceum* and *T.caerulae* using ISSR and RAPD markers

- Genetic diversity was studied using ISSR, RAPD and ISSR + RAPD approaches.
- Parameters like average number of alleles per polymorphic loci, percent polymorphism, average heterozygosity and marker index were calculated.
- Total 200 accessions of different geographical location were procured from USDA seed bank.
- *T.foenum-graceum* and *T.caerulae* showed that the accessions from Turkey exhibited maximum diversity falling into different clusters in spite of being geographically very close to each other.
- It can be inferred that Turkey could be the Center of Origin and / or the Center of Diversity for these leafy legumes.

Research Assistant, Dept of Botany, University of Pune, India Oct '00 –Apr'02

Resolution of Chlorophyll –Protein Complexes using a low Ionic strength Native Green Gel System

- Primary goal was to develop solubilization and fractionation conditions to purify each pigment –protein in its native state, i.e. without loss of its non-covalently bound pigments.
- Secondary goal was to isolate the pigment proteins in their more natural photo chemically active multi-protein complexes
- A combination of native gel and fully denaturing PAGE was used to determine number of pigment protein and their polypeptide composition as well as their association in multi-protein complexes.
- Surfactant extraction was successfully applied to the photosynthetic membrane and resolved by electrophoresis

Research Intern, Bhaba Atomic Research Center, Division of Molecular Biology, India Jun'01-Sept'01

Active site study of Phosphoenolpyruvate Carboxylase using site-directed mutagenesis

- Four –primer three PCR approach was used for generating site-directed mutants. Mutagenic primers were synthesized for introducing the H639P, H639Q, R764T, and R 763/764T in *pepc* gene in reverse direction and their corresponding primers in forward directions.
- Two stages PCR were carried out using flanking forward primer and reverse primers versions of mutagenic primers. The third round of PCR was carried out using DNA fragments obtained from the above two PCR reactions as template and flanking non-mutagenic primers.
- Five sets of mutants were isolated and transformed in *E.coli* DH5 α . Recombinants were scored by α -complementation. The plasmids were PCR amplified, which showed 2 kb PCR amplified products. These PCR products were ligated into pET21b+, an expression vector and recombinant clones were obtained. The full-length PCR product obtained were taken for sequencing purposes (Manual sequencing).
- PEP-carboxylase enzyme was purified from *Amaranthus hypochondriacus* using Ion exchange chromatographic techniques.

Research Assistant, Bachelor of Science, Fergusson College, University of Pune, India Jun'97-May'00

In vitro* vegetative propagation of *Mimusops hexandra

- *Mimusops hexandra* is used as a rootstock for *Achras sapota*. *Achras sapota* fruits are common all over Asia. For fruiting *Achras sapota* requires 2-3 years from graft as compared to 10 years from seed. But *Mimusops hexandra* habitat is restricted to certain geographical locations (Madhya Pradesh) and climatic conditions.
- *In vitro* vegetative propagation was conducted on different basal medium, with different concentration of growth regulator.

The subculture could be maintained for 4 weeks, but failed to survive due large amount of exudes (alkaloids).

Mutagenic effect of Bruffin and Quinine on *Lycopersicum esculentum*, *Solanum melangena* and *Capsicum annum*

- Seeds of *Lycopersicum esculentum*, *Solanum melangena* and *Capsicum annum* were treated with different concentration of bruffin and quinine. Morphological, physiological and cytological effects were studied.
- Cytological parameters such as chromosomal numbers, length and deformities were studied.
- Morphological parameters such as growth rate, leaf size, height of plant, flowering period and fruit setting were noted.
- Physiological parameters such as enzyme activity and chlorophyll content were estimated.
- It was observed that plants undergo alteration which indicated that bruffin and quinine may have some mutagenic effect on plants chromosomes and physiological growth.

Physiological studies in *Thespesia populnea* on application of liquid fertilizers

- Studied the effect of different ratios of NPK on growth of *Thespesia populnea*.
- NPK ratios of 6:12:6, 12:6:6, 10:10:5, 10:5:10 and 8:8:8 were given to seedlings and later at regular intervals to the plants grown in controled green house conditions.
- Growth parameters like height of plant, leaf area, number of leaf per plant, and fresh and dry weight were recorded.
- NPK ratio of 10:10:5 and 12:6:6 showed increased growth in *Thespesia populnea*.

Relevant Coursework

Proteomics, Molecular Ecology, Basic Concepts of Modern Genetics, Plant physiology, Plant Growth and Development, Biometry, Trees and the Environment, Water/Nutrient relationship in plants

Computer Skills

MS Word, MS Excel, Powerpoint, UPGMA analysis, WINBoot-JACCARD, Jalview, ClustalW, Primer 3, JMP-7, Sigma Plot 10, Delta2D, PDQuest, MapMan

Techniques

Tissue Culture: plant organ culture, fungal culture, *in-vitro* symbiotic culture of aspen and *Laccaria bicolor*, isolation of DNA, RNA plant and fungi, Total root and leaf proteins from poplar, electrophoresis (Agarose gels, polyacrylamide gels, sodium dodecyl sulphate polyacrylamide gel) 2-D gel, cloning, manual DNA sequencing, real time polymerase chain reaction and enzyme assay, high pressure liquid chromatography, working in clean room, scanning electron microscopy, DDRT-PCR

Teaching Experience

Graduate Teaching Assistant: Aug '06 – Aug'10

- Biology 115 laboratory (Principles of Biology)
- Biology 117 laboratory (The functional diversity of organisms)
- Invited Guest Lecture at Allegheny College, PA '09
- Invited Guest Lecture at University of Pune, India'10

Poster Presentation

- **Shalaka Desai**, Dhiraj Naik, Jonathan R. Cumming. Poster entitled "Effects of mycorrhizal colonization and phosphorus limitation on carbon allocation in poplar", presented at 95th Annual Meeting Ecological Society of America, 2010
- Dhiraj D. Naik, **Shalaka Desai**, Ernest W. Smith, and Jonathan R. Cumming Poster entitled "Ectomycorrhizal fungi Paxillus involutus prime the poplar seedling against aluminum stress by modulating ascorbate glutathione and phospholipase pathway: A transcriptome", presented at 95th Annual Meeting Ecological Society of America, 2010
- **Shalaka Desai**, Dhiraj Naik, Ernest W. Smith, Jonathan R. Cumming. Poster entitled "Phosphorous availability and influence of ectomycorrhizal fungi *Laccaria bicolor* on growth, nutrient acquisition and rhizospheric carbon flux of *Populus tremuloides*", presented at the Ecological Society of America, Pennsylvania State University, 2009
- **Shalaka Desai**, Dhiraj Naik, Ernest W. Smith, Jonathan R. Cumming. Poster entitled "Phosphorus starvation alters exudation and antioxidant enzyme activity of mycorrhizal and non-mycorrhizal *Populus tremuloides*, at the Graduate Recruitment week in the Department of Biology, West Virginia University, November 2008

Awards

- West Virginia University, Summer Research Grant Award 2008/09/10
- West Virginia University, Travel grant award 2009, 2010
- 17th Penn State Plant Biology Symposium, Pennsylvania State University, Travel grant award 2009
- West Virginia University, Eberly college of Arts and Sciences Travel grant award 2010
- Dissertation Fellowship 2010

Visa Status

Permanent Resident of United States of America