PROJECT PROFILE

Title of the Project: **Differential Analysis of Transcript Expression in *Casuarina-Trichosporium* Interaction to Isolate Defense Related Genes**

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Duration: 2003 – 2007

Objectives:

1. Differential expression profiling of transcripts in *C. equisetifolia* during pathogenesis.
2. Identification, confirmation and sequencing of differentially expressed transcripts during infection.
3. Isolation, cloning and characterization of full-length defense related genes.

Funding Agency: Department of Biotechnology, Govt. of India

Summary

- Transcript profiling revealed over expression of 14% pathogen defense- related; 6% other abiotic stress related; 2% symbiotic; 2% cell wall related transcripts and 2% regulatory genes while 70% of transcripts were unknown. Major group of transcripts included chitinase, glucanase, cytochrome oxidase, signal recognition particle, proteasome, arabinogalactan, R gene, heat shock proteins and cyclin dependent kinase involved in all pathogenesis related pathways including HR, PCD and SAR.

- Transcripts like nodulin which are expressed during early nodulation in *Casuarina* was also found to be over expressed when challenged during pathogen elicitation.

- Several transcripts expressed during abiotic stresses like LEA dehydrin and transcripts with similarity to drought stress related ESTs were up-regulated during pathogen elicitation. The up-regulation of an unknown transcript with heavy metal binding domain was documented

during both biotic stress (fungal elicitation) and abiotic stresses (water deficit, salt stress and elevated temperature).

- qRT-PCR analysis revealed 28 fold increase in expression of the glucanase; 13.6 fold increase in expression of chitinase; 16 fold increase of gene coding for cytochrome oxidase; 9 fold increase of gene encoding nodulin and 2.7 fold increase in expression for gene having a heavy metal domain was observed. Transcript coding for signal recognition particle showed 1-fold increase in expression after 48 hours of pathogen elicitation.

- Fifty two EST sequences were submitted to NCBI and is the first set of EST sequences representing this species with accession numbers GR228669 to GR228718 and GR312926 and GR312925.

- Two pathogen defense – related (PR) genes viz. class I chitinase (*CeChi1*) and glucanase (*CeGlu*) were isolated and characterized. This is the first report on isolation of PR genes from this species.

- Complete specifications for joint IFGTB-DBT process patent titled "A simple protocol for isolation of undegraded total RNA from *Eucalyptus* and *Casuarina* and cDNA synthesis from unpurified RNA" was filed with application no. 1927/CHE/2009 dated 13-08-2009. It is a low cost and high recovery protocol for isolation of total RNA from guanidine recalcitrant tissues with high phenolic content using non toxic chemicals. The protocol also describes the down-streaming of total RNA to cDNA without purification.