Executive summary of Minor research Project Entitled "Optimization of Laccase Production from Potential Laccase Producer Isolated from Mixed Microbial Culture of Dyeing Textiles Effluent" PI-Dr. Mrs. S.M.Dharmadhikari Ref No- Letter no. 47-2136/11 dated 8 March 2012

In the present study for isolation of laccase producing microorganisms sample collected from different forest regions, soil samples near from textile and dye industries and textile waste were used. Out of different bacterial, fungal and actinomycetes isolate, one potent Laccase producing fungus namely *RS 3* were selected for further study depending upon its high productivity. Using the gene specific sequencing primers, the purified PCR amplicons was sequenced. The sequences were analyzed using Sequencing Analysis 5.2 software. Blast result and phylogenetic tree analysis clearly indicate that fungal strain RS3 is *A. nidulans* 

For the Laccase assay substrate Guaiacol has been used. The intense brown color development due to oxidation of guaiacol by laccase can be correlated to its activity often read at 450 nm. The Ammonium sulphate was used for purification of produced enzyme and it was found that at 85% saturation of solution maximum enzyme activity was observed in precipitate. The process parameters for enzyme production were optimized viz. Carbon source, Nitrogen source, pH and Time of incubation. It was found that Enzyme activity was maximum in basal medium containing starch (Carbon source), Peptone (Nitrogen source) and at pH 5.5 when incubated for 12h (Time of incubation).

Dye decolourization efficiency of fungi *A. nidulans* was also determined. Initially the activities of Laccase against 12 different synthetic dyes were determined. On the basis of % Decolourization three dyes namely Congo red, Methyl orange and Alizarin red were selected for further study. Decolourization efficiency of free fungal mycelium and crude laccase enzyme was determined and it was found that maximum % degradation was obtained against Congo red (88.48%).Fungal spores were immobilized by using calcium alginate gel entrapment method and activity of immobilized laccase was determined by using Congo red solution of different concentration. Maximum activity was observed when fresh beads of immobilized fungus were used (78.67 %) in 50 ppm dye solution.

The laccase enzyme produced by fungus *A. nidulans* was found to be very useful in degradation of synthetic dyes that are commonly found in textile waste, dye industries and lignocelluloseic wastes when used freely or in immobilized form.