

**DECOLOURIZATION OF INDUSTRIAL WASTE WATER AND
XYLANASE PRODUCTION BY *ASPERGILLUS LENTULUS***

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CENTRE FOR RURAL DEVELOPMENT AND TECHNOLOGY

INDIAN INSTITUTE OF TECHNOLOGY DELHI

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XYLANASE PRODUCTION BY *ASPERGILLUS LENTULUS***

by

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CERTIFICATE

This is to certify that the thesis entitled **“Decolourization of industrial waste water and xylanase production by *Aspergillus lentulus*”** being submitted by **Ms. Prachi Kaushik** to the Indian Institute of Technology Delhi for the award of the degree of ‘Doctor of Philosophy’ is a record of bonafide research work carried out by her. She has worked under my guidance and supervision and has fulfilled the requirements for the submission of this thesis. To the best of my knowledge the results contained in this thesis have not been submitted in part or full to any other University or Institute for the award of any degree or diploma.

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ABSTRACT

The present study focuses the dual applications of previously isolated fungal strain *Aspergillus lentulus* in decolourization of industrial waste water and xylanase production for pulp and paper industries. To explore the potential of *A. lentulus* for dye removal and to understand the mechanism involved, dye removal studies from dye solutions/mixtures and industrial effluents were performed using growing (i.e. bioaccumulation) as well as resting (i.e. biosorption) fungal biomass. The strain could efficiently (80.6 – 99.4%) remove various anionic and cationic dyes (singularly as well as in mixtures) during its growth, with the former being removed at a relatively faster rate. In depth investigations on the effect of process parameters on widely used Acid Navy Blue dye established that this strain could perform dye removal over a wide range of temperatures, pH, salt and dye concentration. Being alkali, thermo and halo tolerant, *A. lentulus* has immense potential for decolourization of industrial effluents which contain the cocktail of many dyes and are often alkaline, hot and have high salt concentrations.

With a view to minimize the cost of waste water treatment, efforts were made to work out the minimum dose and easy accessibility of nutritional resources. The minimum inputs of conventional carbon (glucose) and nitrogen (yeast extract) sources were worked out and alternative carbon (molasses, jaggery and starch) and nitrogen (urea and NH_4Cl) sources were also established for supporting dye removal. With the help of response surface methodology, process for dye removal was optimized for three variables, glucose, urea and dye concentration. Significant enhancement in dye uptake capacity (from 49.7 mg/g to 97.5 mg/g) was observed when the low cost optimized media (1/10th cost) developed by eliminating yeast extract and reducing the glucose inputs was used for dye removal. Hence, optimization study resulted in lowering the process cost as well as minimization of dye laden sludge.

Biosorption of dyes on pre-cultivated (resting or dead) biomass of *A. lentulus* also resulted in remarkable dye removal (94-97%) for anionic dyes while it remained low for cationic dyes (22-55%). Nevertheless, biosorption performance was significantly influenced

by biomass loading, initial dye concentration, pH, temperature etc. Higher dye removal and dye uptake capacity was recorded in growing (bioaccumulation) mode as compared to the resting (biosorption) mode. Spectroscopic, microscopic and FTIR analysis was used to elucidate the mechanism of dye removal during growth of *A. lentulus*. Bioaccumulation along with biosorption was found to be the principle mechanism involved in dye removal by *A. lentulus*. The applicability of alkali, thermo and halo tolerant *A. lentulus* for decolourization of industrial effluents was further reinforced by demonstrating efficient dye removal from nutrient supplemented textile (65.9% dye removal) as well as pulp and paper (86.4% dye removal) effluents. Results also established that *A. lentulus*, which is much more efficient than native microbes, can very well compete and bring about even higher removal synergistically.

A. lentulus was found to be an effective producer of cellulase free xylanase enzyme and could be easily grown on various agricultural residues yielding high crude enzyme activities. Good xylanase activities were obtained when wheat bran (814.9 U/g) and corn cob (799.4 U/g) were used as the substrates. The spent slurry after enzyme extraction was efficiently utilized for the removal of both cationic as well as anionic dyes and was subsequently routed through vermicomposting process. These observations prove that *A. lentulus* could be sequentially employed, first for xylanase production and then in the form of waste product (spent slurry) for efficient dye removal. A carrier based formulation of *A. lentulus* with a shelf life of six months at ambient conditions and one year at controlled temperatures (4°C and 30°C) was developed and evaluated for dye removal and xylanase production efficacy. Finally, the formulation was developed and optimized into a tabletted product evaluated in terms of biological and physical characteristics. The results provide the first tabletted product for dual purpose of dye removal and xylanase production, an innovation that shall greatly simplify the storage, handling and dosing in the industrial processes, especially for the unskilled masses in small scale industries.

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