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OPTIMISATION OF REMAZOL BRILLIANT BLUE R DYE DECOLOURISATION AND LACCASE ENZYME PRODUCTION BY *Marasmius cladophyllus* USING RESPONSE SURFACE METHODOLOGY

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ABSTRACT

The decolourisation of Remazol Brilliant Blue R dye and laccase activity was investigated using pure culture of an endophytic fungus, *Marasmius cladophyllus*. The fungus is found capable of decolourising 99% of the dye after 12 days of incubation in Glucose Minimal (GM) liquid media (pH 5.5) and laccase activity of 285 U/L was recorded. Response surface methodology (RSM) was used to determine and optimise the significant variable(s) in order to obtain the optimum dye decolourisation conditions and laccase production. It was also used to study the interaction effect of the variables on both responses. Box-Behnken Design was used to identify the significant variable(s) whereas the optimisation process was done by using Central Composite Design. It was found that initial dye concentration of 100-300 mg/L, incubation period of 4-20 days and pH of liquid medium of 4-8 significantly influenced the decolourisation of dye and laccase activity. However, only the relationship of the incubation period and pH is significantly affected both the responses. Maximum dye decolourisation of 100% was successfully achieved and the highest laccase activity of 504.53 U/L was recorded after 16 days of incubation period at pH 7 with 259.46 mg/L initial dye concentration.

Key words: Response surface methodology, Remazol Brilliant Blue R, *Marasmius cladophyllus*, decolourisation, laccase

INTRODUCTION

The expanding textile dyeing industry has caused an increase in the volume and complexity of wastewater released to the environment and has created severe water pollution globally (Cervantes & Santos, 2011). During the dyeing processes, there are estimated about 5-50% of unfixed dyes lost in the industrial textile effluent (Maljaei *et al.*, 2009).

Remazol Brilliant Blue R (RBBR) dye is one of the most extensively used dyes in the textile industry. The dye has broadly been used as a model compound in the studies of dye degradation and it is also a derivative of anthracene, which represents an important recalcitrant and toxic organopollutants (Hadibarata *et al.,* 2012). Textile dyes are recalcitrant compounds because they are stable to temperature, microbial attack and light (Rodriguez *et al*., 1999). Thus, the treatment of textile industrial effluent has been one of the most challenging treatments among other industrial effluents (Fu & Viraraghavan, 2001).

Numerous physiochemical systems have been used to treat industrial wastewater (Yeh & Thomas, 1995) but it has many disadvantages (Stolz, 2001) which have resulted in an urge to develop effective biological system to degrade dyes in textile industrial effluent. Lignolytic fungi are known to degrade textile dyes by using laccase enzyme (Stolz, 2001). Due to the aromatic ring structures in the dye that has similar characteristics as the lignin, the white-rot fungi produces extracellular oxidative enzymes such as laccase, lignin peroxidase (LiP) and manganese peroxidase (MnP) (Mohamad Hasnul *et al.,* 2015). The non-specific oxidative enzyme system produced makes the white-rot fungi becomes useful for many types of bio-

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