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Research Article

Decolourisation Capabilities of Ligninolytic Enzymes Produced by *Marasmius cladophyllus* UMAS MS8 on Remazol Brilliant Blue R and Other Azo Dyes

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Marasmius cladophyllus was examined for its ability to degradatively decolourise the recalcitrant dye Remazol Brilliant Blue R (RBBR) and screened for the production of ligninolytic enzymes using specific substrates. Monitoring dye decolourisation by the decrease in absorbance ratio of A_{592}/A_{500} shows that the decolourisation of RBBR dye was associated with the dye degradation. *Marasmius cladophyllus* produces laccase and lignin peroxidase in glucose minimal liquid medium containing RBBR. Both enzyme activities were increased, with laccase activity recorded 70 times higher reaching up to 390 U L^{-1} on day 12. Further in vitro RBBR dye decolourisation using the culture medium shows that laccase activity was correlated with the dye decolourisation. Fresh RBBR dye continuously supplemented into the decolourised culture medium was further decolourised much faster in the subsequent round of the RBBR dye decolourisation. In vitro dye decolourisation using the crude laccase not only decolourised 76% of RBBR dye in just 19 hours but also decolourised 54% of Orange G and 33% of Congo red at the same period of time without the use of any exogenous mediator. This rapid dye decolourisation ability of the enzymes produced by *M. cladophyllus* thus suggested its possible application in the bioremediation of dye containing wastewater.

1. Introduction

Synthetic dyes are widely used throughout the world for various purposes particularly for textile dyeing. Each year, more than 80,000 tons of reactive dyes are produced and consumed for textile dyeing. Given their beneficial characteristic of bright colour, water-fast, and simple application techniques with low energy consumption, reactive dyes have been used extensively to dye more than half of the global production of cotton [1]. However in the process of textile dyeing, 50–60% of these water soluble dyes do not bind to the fiber and are lost into the effluent. In addition to the high volume of water required, each year textile mills generate and discharge billions of liters of wastewater effluent full of colours, salts, and organic chemicals harmful to the environment [1, 2].

Remazol Brilliant Blue R (RBBR) dye, also known as Reactive Blue 19, is a typical reactive dye used in the textile industry for dyeing cellulosic fibers. It is an anthraquinone-based vinylsulphone dye frequently used as the starting material in the production of polymeric dyes. RBBR dye however has relatively low fixation efficiency (75–80%) on cellulose due to the competition between the formation of reactive vinylsulphone and 2-hydroxyethylsulfone in which the latter does not attach to cellulose fiber [3, 4]. When released without adequate treatment, RBBR dye can persist for a long period of time in the environment due to the fused aromatic ring structures of the dye which are difficult to degrade. At a temperature of 25°C and under normal pH of 7, RBBR dye has been reported to possess a half-life as long as 46 years [3]. Moreover, RBBR is an anthracene derivative and represents an important class of toxic and recalcitrant organopollutants [5]. Dye containing wastewater therefore must be treated before their discharge into the environment.

In recent years, interest in using microorganisms to treat dye containing wastewater has increased not only because