Comparison of Alkali-Tolerant Fungus *Myrothecium* Sp. IMER1 and White-Rot Fungi for Decolorization of Textile Dyes and Dye Effluents

Juan Huang[1], **Yun Fu**[2] **and Youxun Liu**[2*]

[*]**Corresponding author:** Youxun Liu, The Laboratory of biochemistry, School of Basic Medical Sciences, Xinxiang Medical University, Jinsui Avenue, Xinxiang, Henan, China, Tel: 86-373-3029127; Fax: 86-373-3831739; E-mail: liuyouxun@126.com

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Abstract

A new isolated nonligninolytic fungus, strain sp. IMER1, was found to decolorize five different synthetic dyes when grown on dye-containing agar plates. The capability of sp. IMER1 for decolorization of Remazol Brilliant Blue R (RBBR) and dye effluents was compared with that of five white-rot fungi. More than 65% RBBR removal by sp. IMER1was observed at various pHs (5-10). About 60-95% of decolorization was observed with these white-rot fungi in the acidic pH range of 5.0-6.0, whereas color removal rate was less than 30% in the basic pH range of 8.0-10.0. sp. IMER1 had a more efficient decolorization of the dye in a broad pH range than white-rot fungi tested. In comparison with color removal performance, sp. IMER1 was approximately 2-5-fold better than white-rot fungi tested in the basic pH range. Additionally, the visual observation and Ultraviolet-Visible (UV-VIS) spectral analysis demonstrated that decolorization of dye by sp. IMER1 was due to biodegradation and biosorption. Biomass production was not affected by changes in the pH range of effluents. Our previous studies showed that the nonligninolytic fungus,

Keywords: Myrothecium sp. IMER1; White-rot fungi; Alkali-tolerant; Decolorization

Introduction

Worldwide, over 10,000 different dyes and pigments are used in tile, cosmetic, printing, drug, and food-processing industries [1]. ost of synthetic textile dyes are mutagenic and/or carcinogenic and ong to the most dangerous pollutants [2-4]. Although conventional emical and physical techniques such as precipitation, adsorption, d ozonation have been employed for the decolorization of dye uents, they possess inherent limitations such as high cost, mation of hazardous by-products, and intensive energy uirements [4-6]. As a feasible alternative, dye decolorization using croorganisms has recently received much attention owing to their t effectiveness [7-11]. Currently, a lot of studies have focused on ite rot fungi that seem to be more prospective organisms [12]. ese fungi are efficient ligninolytic organisms capable of degrading ny xenobiotic compounds including various types of dye such as , anthraquinone, reactive, and triphenylmethane dyes [13-17].

Due to the complexity of the biodegradation mechanism of inolytic system, requirements for some redox mediators and low for the optimum activity of the enzymes will be present in a stewater, which are major disadvantages of bioremediation by white fungi [14,18]. On the other hand, studies of nonligninolytic fungi grading dyes are few [19]. Such studies are very interesting not only m the standpoint of comparative biology but also with the pectation of finding better fungi for use in the treatment of dye

Myrothecium sp. IMER1, can decolorize dyes and dye effluents. Furthermore, unlike white rot fungi, this strain can grow at high pH [20-22].

However, decolorization efficiency of dye/dye effluents by *Myrothecium* sp. IMER1 and white-rot fungi were not systematically compared. Therefore, the major objective of this study is to investigate efficiencies of different white rot fungi strains and *Myrothecium* sp. IMER1 for decolorization of the dye (RBBR) and dye effluents at various pHs. The results would help to evaluate decolorizing performance of these strains tested and select the right strains for an effective decolorization process.

Materials and Methods

Materials

2, 2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) was purchased from Sigma Chemical Company (St. Louis, MO, USA). RBBR (C.I. 61200) was purchased from Sigma (St. Louis, MO, USA), Congo Red (C.I. 22120) and Indigo Carmine (C.I. 73015) from Tianjin Damao Chemical Reagent Factory (Tianjin, China). All other chemicals used were analytically pure. Highly colored effluents (dark red) were collected from a textile dye-producing plant situated in Foshan, Guanzhou, China. The dye effluents contained both acid and

the effluent. It was an alkaline wastewater with a pH value of 10-10.5. Prior to laboratory decolorization treatment, the effluent was centrifuged at 10,000×g for 15 min to remove large suspended particles and then was sterilized. Sample absorbances were measured using a Varian CARY50 UV-Vis (St. California, USA) spectrophotometer; pH values were measured with a pHS-3A pH-meter (Hangzhou Wanda Instrument Factory, China).

Organism and culture conditions

Fungal strains used in this study were: SDK (*Polyporus* sp. SDK), AX3 (*Leutinus* sp. AX3), DS1 (*Schizophyllum* sp. DS1), CD3 (*Irpex* sp. CD3), BP2 (*Pleurotus* sp. BP2), IMER1 (*Myrothecium* sp. IMER1). The strain *Myrothecium* sp. IMER1 was isolated from soil from the suburb of Wuhan, P.R.China. On the basis of the comparison of sequences of the ITS regions and 5.8SrRNA gene with those found in databanks, its morphology, and microscopy observations, it was identified as *Myrothecium* sp. (GenBank accession no. EF458487). These strains were isolated and identified in our laboratory. They were grown in potato dextrose broth medium (PDB), or on potato dextrose agar medium (PDA). Every strain was maintained on PDA medium slant. The slant was inoculated and incubated at 28°C for 5-6 days, and then stored at 4°C and periodically sub-cultured.

Dye decolorization on solid plates

color removal rate was less than 30% in the basic pH range of 8.0-10.0. By contrast, 60-70% color removal was obtained with *Myrothecium* sp. IMER1 in the acidic pH range of 5.0-6.0, and more than 65% of decolorization was observed in the basic pH range of 8.0-10.0. These results showed that *Myrothecium* sp. IMER1 exhibited excellent decolorizing performance at all the pH value tested (5.0 to 10.0). Kapdan *et al*. reported the optimum growth pH of *Coriolus versicolor* as 4.5 [29]. Decolourization of Solar golden yellow R by *Schizophyllum commune* indicated that maximum decolourization efficiency (73%) was observed at pH 4.5 after 6 days [30]. It has been widely reported that for majority of the fungi the optimum pH for dye decolorization is in the acidic range. Unfortunately, such a low pH is not suitable for the wastewater treatment. Therefore, fungal strain showed an appreciable decolorization of dye over a wide range of pH, which is desirable for industrial applications since it can be used for wastewater treatment without a previous pH adjustment stage [31].

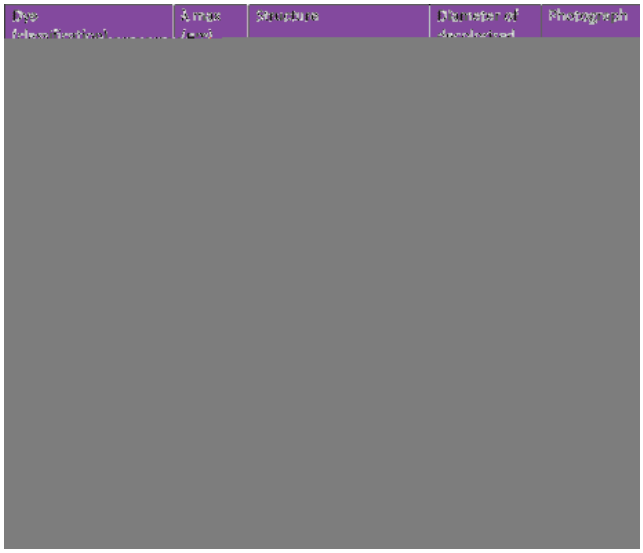

Figure S1: Characteristics, molecular structure and decolorization studies of the selected dyes during solid plate growth of *Myrothecium* sp. IMER1

Decolorization of dyes by most fungi could be due to adsorption to microbial cells or to biodegradation [32,33]. If the dye removal were attributed to biodegradation, either the major UV-VIS light absorbance peak would completely disappear or a new peak would appear. In adsorption, cells of fungus become deeply colored because of adsorbing dyes [34,35]. The absorbance spectra of the dye were scanned before and after decolorization and changes in its absorption spectrum (350-800 nm) were recorded. The absorption peaks of RBBR in the visible region disappeared, suggesting that removal of dye by *Myrothecium* sp. IMER1 should be partly attributed to biodegradation (Figure 1b). In addition, in the decolorization of RBBR by *Myrothecium* sp. IMER1, the cells were stained deeply colored at pHs (5.0-10.0), indicating that the dye was adsorbed on the cell surface, which was partly due to biosorption [Figure 1c]. It suggested that decolorization by *Myrothecium* sp. IMER1 involved biosorption and biodegradation. This result was in agreement with our previous studies [20,21].

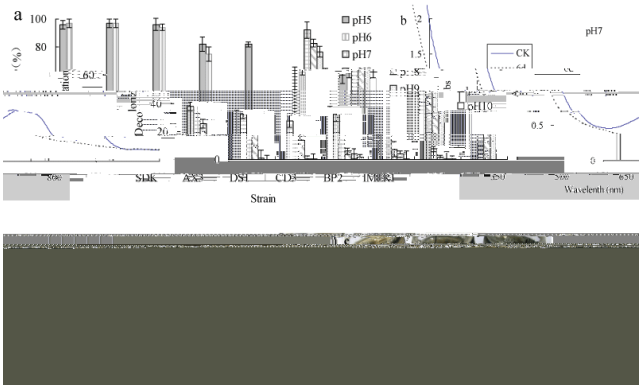

Figure 1: Effect of pH on decolorization of RBBR by *Myrothecium* sp.IMER1/five white-rot fungi (pH5-10). a) Decolorization of RBBR by fungi at various pHs, b) UV-VIS absorbance spectra of decolorization for RBBR by IMER1 and c) Photographs of decolorization for RBBR by IMER1at different pH

To investigate the effectTo

Comparison of *Myrothecium* sp. IMER1 and five white-rot fungi for decolorization of dye effluents

Although a large number of lab-scale studies have been conducted on decolorization of single synthetic dye solutions and simulated dye wastewaters by fungal biosorption/biodegradation, there is a need to generate relative performance data on real dye effluents [37,38]. Decolorization of industrial effluent from dyeing industry is shown in Figure 3. At pH 7.0 and 9.0, decolorization by *Myrothecium* sp.

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