BRAZILIAN JOURNAL OF MICROBIOLOGY

http://www.bjmicrobiol.com.br/

Environmental Microbiology

Screening of freshwater fungi for decolorizing multiple synthetic dyes

Panpan Yang[a,b], *Wenxiao Shi*[a], *Hongkai Wang*[a,*], *Hongmei Liu*[b]

[a] *Zhejiang University, Biotechnology Institute, State Key Laboratory of Rice Biology, Hangzhou, Zhejiang, China*
[b] *Shandong Agricultural University, College of Life Science, State Key Laboratory of Crop Biology, Tai'an, Shandong, China*

ARTICLE INFO

Article history:
Received 22 May 2015
Accepted 6 March 2016
Available online 4 July 2016

Associate Editor: Cynthia Canêdo da Silva

Keywords:
Freshwater fungi
Biodegradation
Synthetic dyes
Decolorization

ABSTRACT

The biodegradation of synthetic dyes by fungi is emerging as an effective and promising approach. In the present study, freshwater fungal strains isolated from submerged woods were screened for the decolorization of 7 synthetic dyes. Subsequently, 13 isolates with high decolorization capability were assessed in a liquid system; they belonged to 9 different fungal species. Several strains exhibited a highly effective decolorization of multiple types of dyes. New absorbance peaks appeared after the treatment with 3 fungal strains, which suggests that a biotransformation process occurred through fungal biodegradation. These results showed the unexploited and valuable capability of freshwater fungi for the treatment of dye-containing effluents. The ability of certain fungi to decolorize dyes is reported here for the first time.

Introduction

Synthetic dyes are widely used for coloring the products of several industries such as textiles, leather, cosmetics, paper, printing materials, and plastics. It is estimated that 1–2% of dye production is lost, and 5–10% is discharged to the environment when the dyes are used.[1,2] Several dyes and the chemicals used to produce them are often toxic, carcinogenic or even explosive.[3,4] Effluents from industries that use various dyes are considered as pollutants that can cause severe environmental problems as well as medical and esthetic problems.[2] The decolorization of this industrial waste is a challenging task because certain dyes are resistant to degradation.[4]

Physical and chemical decolorizing methods have been developed to remove dyes from wastewater; however, several of them have disadvantages such as high costs and/or limited applicability.[2,5] Studies on the capability of microorganisms to perform dye decolorization has received increasing attention because the use of microorganisms is considered a cost effective and environmentally friendly method for removing organic dyes from wastewater before they are discharged.[6,7] Likewise, the capabilities and mechanisms of decolorization by bacteria have been studied.[8,9] However, the application of bacteria was limited due to the narrow substrate range of various degrading bacteria.[10] Moreover, to complete the degradation of dyes, different groups of bacteria are required in an alternation process from anaerobic to aerobic conditions.[2,7]

Research on the fungal decolorization of dye wastewater has been performed in recent years.[11,12] Several fungi with the capability to decolorize a wide range of dyes have been

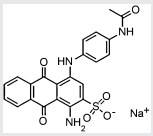
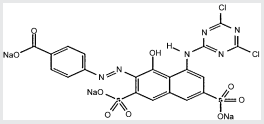
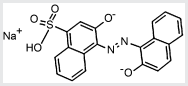
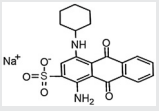
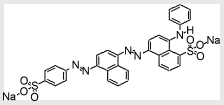
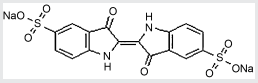
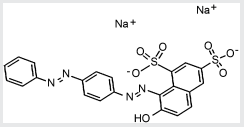
* *Corresponding author.*
E-mail: hkwang@zju.edu.cn (H. Wang).
http://dx.doi.org/10.1016/j.bjm.2016.06.010

reported. For example, the white-rot fungi and brown-rot fungi are well-studied fungi groups with decolorization abilities.[12] Other fungi, such as *Aspergillus niger*, *Rhizopus arrhizus*, and *R. oryzae*, can also decolorize and/or absorb diverse dyes and possess excellent color removal capabilities.[13,14]

The mechanism of fungal decolorization mainly involves two aspects, biodegradation and biosorption.[15] The biodegradation capability of fungi is due to their extracellular, non-specific and non-selective enzyme system.[6] Fungal enzyme production depends on nutrient limitations, and their subsequent dye decolorization ability is achieved depending on the growth conditions.[16] Considering the complex environmental factors involved in the dye wastewater conditions, the screening of more fungi is necessary for use in dye decolorization.

Aquatic fungi are the main decomposers of aquatic ecosystems and play crucial roles in the cycling of nutrients.[17] In addition, a unique characteristic of fungi is their ability to produce several non-specific enzymes.[18] These non-specific extracellular and/or exoenzymes enable the aquatic fungi to attack structurally diverse organic compounds that correspond to different pollutant classes.[19,20] Hence, these fungi may serve as a new resource to treat wastewater. However, little research has been performed on the process of decolorizing wastewater by freshwater fungi,[10] and more fungi with potential for the biodegradation of synthetic dyes need to be explored. In the present study, several freshwater fungi were isolated from streams in the Zhejiang Province, PR China, and their ability of decolorizing multiple synthetic dyes was evaluated.

Table 1 – Structures of the synthetic dyes used in the present study and the wavelength of maximum absorption – λ_{max} (nm).

Dye	Structure	λ_{max} (nm)
Acid Blue 40		620
Reactive Red 11		525
Acid Blue 193		580
Acid Blue 62		590
Acid Blue 113		630
Reactive Blue 74		590
Acid Red 73		500

Materials and methods

Dyes and media

Seven dyes were used for screening the decolorization ability of the fungal isolates. All the dyes were dissolved in distilled water at a concentration of 10 g/L; then, they were filtered and sterilized using a 0.22 μm-diameter bacteria filter before being tested for use. All the dyes were purchased from Sigma–Aldrich. The molecular structures of the dyes are listed in Table 1.

Water agar medium (WA; WA1.5% agar) was used for the fungal isolation; potato-carrot-agar medium (PCA; 5% potato, 5% carrot, 2% agar) was used for the identification of fungal isolates; potato-dextrose-agar medium (PDA; 20% potato, 2% dextrose, 2% agar) was used to grow the fungal cultures and for the DNA extraction; and malt-agar medium (MEA; 2% malt extract, 1.5% agar) was used for screening the decolorization by fungi.

The liquid MEA medium used for the decolorization test was the same as that described above but without agar. The PDA medium was used for colony growth, and the genomic DNA isolation was prepared following the method of Wongsawas et al.[21]

Fungal strain isolation from streams

Submerged wood samples were randomly collected from streams in the Zhejiang Province; then, they were brought back to laboratory and placed separately in snap lock plastic bags with sterile moist paper towels, incubated at room temperature and examined periodically during a three-month period. Single spore isolations were obtained using the methods described by Choi et al.[22,23]

Dye decolorization on solid media

The solid media were prepared with MEA medium and the addition of each dye to a total concentration of 50 mg/L. A mycelium plug derived from the edge of fungal strains grown on WA medium plates for 4 days at 25 °C were transferred to the center of a solid medium plate and inoculated at 25 °C for 1 week. The formation of decolorized zones under or around the developing mycelia was monitored for dye decolorization. All the agar plate assays were performed in triplicate.

Morphological identification of fungal strains with decolorization capability

Pure cultures of fungi with decolorization capabilities were incubated on PCA, WA and PDA media at 25 °C for 2 weeks for their identification. The morphological characters of fungi were examined and recorded in detail under an Olympus BX-51 microscope equipped with an Olympus DP-50 digital camera system.