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Short communication

Efficiency of decolorization of different dyes using fungal biomass immobilized on different solid supports

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ABSTRACT

Different technologies may be used for decolorization of wastewater containing dyes. Among them, biological processes are the most promising because they seem to be environmentally safe. The aim of this study was to determine the efficiency of decolorization of two dyes belonging to different classes (azo and triphenylmethane dyes) by immobilized biomass of strains of fungi (*Pleurotus ostreatus* – BWPH, *Gleophyllum odoratum* – DCa and *Polyporus picipes* – RWP17). Different solid supports were tested for biomass immobilization. The best growth of fungal strains was observed on the washer, brush, grid and sawdust supports. Based on the results of dye adsorption, the brush and the washer were selected for further study. These solid supports adsorbed dyes at a negligible level, while the sawdust adsorbed 82.5% of brilliant green and 19.1% of Evans blue. Immobilization of biomass improved dye removal. Almost complete decolorization of diazo dye Evans blue was reached after 24 h in samples of all strains immobilized on the washer. The process was slower when the brush was used for biomass immobilization. Comparable results were reached for brilliant green in samples with biomass of strains BWPH and RWP17. High decolorization effectiveness was reached in samples with dead fungal biomass. Intensive removal of the dyes by biomass immobilized on the washer corresponded to a significant decrease in phytotoxicity and a slight decrease in zootoxicity of the dye solutions. The best decolorization results as well as reduction in toxicity were observed for the strain *P. picipes* (RWP17).

Introduction

Synthetic azo-, triphenylmethane, and anthraquinone dyes are commonly used in the textile, food, cosmetics, papermaking and pharmaceutical industries.[1,2] They are resistant to light operation, moisture and oxidants because most of them have complex aromatic structures. On the one hand, this is a feature desired by the industry, but on the other hand, it is dangerous for the environment. The presence of

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synthetic dyes in water causes a reduction in aquatic biodiversity by blocking the passage of sunlight through the water and creates problems for photosynthetic aquatic plants and algae. Many synthetic dyes are toxic, mutagenic and carcinogenic.[3–6] In addition, dyes may accumulate in sediments, especially in places of wastewater discharge, and affect the ecological balance of the aquatic system. Leaching of contaminants can affect the groundwater system.[7] The most serious problem is associated with effluents from the textile industry, where the dyes used for dyeing and finishing operations vary from day to day and sometimes even several times a day. Imperfection of textile coloration processes may cause losses of applied dyes (even 10–15%).[3–5,8] Because of this, the textile finishing wastewater is characterized by a strong color and large number of suspended solids.[9]

The colored wastewater is mainly cleaned by physical and chemical procedures, such as adsorption, coagulation, flocculation, flotation, precipitation, oxidation and reduction, ozonation and membrane separation. These technologies are very expensive and have drawbacks.[4,9,10] That is why bioremediation by microorganisms is still an environmentally friendly and cost-competitive alternative. Treatment of textile effluent requires an efficient system of color removal. There are many publications confirming the high potential of bacterial, fungal and algae species in dye removal.[4,11–16] Removal of dyes may be achieved through biodegradation/biotransformation and/or adsorption on biomass. Biotransformation of dye may lead to complete mineralization or formation of less toxic products.[4,12,15,16]

Fungi may use an extracellular enzymatic system to transform aromatic substances, such as lignin, polycyclic aromatic hydrocarbons (PAH) or pesticides. Currently much attention is focused on fungal decolorization processes. Fungal biomass is used as a sorbent and/or producer of enzymes involved in biodegradation/biotransformation. The process of biosorption is rapid, efficient and adaptable to diverse types of textile effluents. The results of different experiments emphasize that fungal processes are mostly associated with biotransformation but not biosorption.[8,12,17,18] Biosorption is observed mostly for non-ligninolytic fungi, such as *Aspergillus niger*, of which (dead) biomass may be used as an adsorbent.[13,19] Among these, the most widely researched are white rot fungi, such as *Phanerochaete chrysosporium*, *Bjerkandera* sp., *Trametes versicolor*, *Irpex lacteus*, and *Pleurotus ostreatus*, which produce enzymes, such as lignin peroxidase, manganese peroxidase and laccase. They are able to degrade many aromatic compounds due to their non-specific enzymatic activity.[6,13,17,19–21] As described previously, white-rot fungi are capable of decolorizing dyes significantly, and in most cases, this is due to the activities of lignin peroxidase (LiP)[22] and Mn-dependent peroxidase (MnP).[23] Some studies have demonstrated laccase (Lac)-mediated dye decolorization.[12,24] It was also reported that non-ligninolytic enzymes may play a role in the decomposition of dyes, such as triphenylmethane crystal violet.[17] Process conditions have a significant influence on biotransformation effectiveness. The best results of dye removal by fungi were obtained in more aerated shaken samples. Living biomass of tested strains removed dyes more effectively than dead biomass.[16,17,25,26]

Decolorization effectiveness depends mainly on the strain used in the process as well as on the specific structure of the dye and composition of the dye effluents. It was demonstrated that strains isolated from polluted sites had a greater decolorization potential than others. Additionally, the form and composition of the culture medium play an important role in decolorization processes.[6,25,27–30] The most important factors are the sources and concentrations of carbon and nitrogen, which have a significant influence on production of ligninolytic enzymes. The influence of different carbon sources on decolorization effectiveness has been extensively studied.[19,27,28,31] It should be mentioned that the effectivenes of dye removal depends also on the way biomass is used. Fungal free-cell treatment shows some drawbacks since the mycelium may be more exposed to environmental stresses. Therefore, a good alternative might involve the immobilization of biomass on different supports. Immobilization protects the biomass and improves fungal activity.[32] It has been reported that immobilization of fungal cells may stably maintain the production of various enzymes at levels higher than those achieved with suspended or pellet forms.[33,34] Moreover, the immobilization of fungal biomass increases fungal resistance to environmental stresses, such as the presence of toxic molecules at high concentrations. Immobilization improves decolorization efficiency of biomass due to less dense fiber packing in comparison with the free fungal biomass. This is because the microorganism has a larger surface area available for dye adsorption. The increase in the surface area of fungal biomass tends to reduce the mass transfer limitations, which in turn increases access to pollutant degradation.[32,35–40] Immobilization may allow the use of the system repeatedly, allowing easier liquid–solid separation and avoiding clogging phenomena.[32,35]

Yesilada et al.[5] demonstrated that white rot fungi pellets may be used for effective decolorization of textile dyes. It was also possible to induce dye decolorization activity of *Funalia trogii* by carefully selecting the optimal culture conditions. Pellets could be used several times and still maintain high decolorization activity. Using pellets would allow treatment of effluents with varying dye compositions and in high concentrations of dyes, which are normally toxic at low concentrations.[5] The aim of the present study was to evaluate the influence of the solid support used for biomass immobilization on the decolorization efficacy. Different solid supports were used in the experiment to obtain intense growth of fungal biomass and to assist in the process of dye removal. After the environmental safeties of the solutions after the decolorization processes were assessed, the zoo- and phytotoxicity were evaluated.

Materials and methods

Tested organisms and culture conditions

The fungal strains *P. ostreatus* (BWPH), *Gleophyllum odoratum* (DCa) and *Polyporus picipes* (RWP17) were isolated by the tissue method (MEA medium (Difco)) with fruiting bodies of fungi collected in the woods near Gliwice (southern Poland, Upper