

Fungal Flora for Degradation of Textile Dye Wastewater

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ABSTRACT: Azo dyes are widely prevalent environmental contaminants that are recalcitrant to biodegradation processes and have detrimental biological effects. Hence, exploring the novel microbial agents and to develop ecofriendly cost-effective process is pertinent for treatment of textile effluents. A fungal consortium comprising three strains *Penicillium oxalicum* SAR-3, *Aspergillus niger* SAR-6 and *Aspergillus flavus* SAB-3 has been developed. The consortium exhibited remarkably high potential to degrade azo dyes (Acid Red 183, Direct Blue 15 and Direct Red 75) at various initial concentrations. The consortium used was found to decolorize all the three dyes almost completely at lower initial concentrations (200–400 mg L⁻¹). The consortium was able to effectively decolorize simulated textile wastewater. UV–Visible and FTIR spectroscopic analysis had denoted the degradation of dyes. Furthermore, toxicity analysis of metabolites generated following degradation had shown significant reduction in the toxicity of dyes. Individual strains of microbial consortium may attack the dye molecules at different positions or may have the capability to utilize metabolites produced by the co-existing strains to promote decomposition, and used consortium of *Trametes versicolor*, *Bjerkandera adusta*, *Bostra fumosa* using straw as solid substrate, to detoxify Poly R-478. Highly efficient fungal mixed culture systems for degradation of azo dyes and textile wastewater treatment have been reported. Fungal consortium has been considered as better treatment for textile wastewater as compared to conventional biological methods or system using single fungal strains. However, up to date, very few studies are available on use of fungal consortium for textile wastewater treatment.

KEYWORDS: textile dye wastewater, fungal strains, consortium, degradation, metabolites, environmental, pollution, toxicity

I. INTRODUCTION

Among pollution causing various industries, textile industry creates a major attention by environmentalists due to consumption of large volume of water, dyes and chemicals for various processing of textiles. Textile effluents contain carcinogenic aromatic amines, dyes, organic and inorganic materials. Removal of colored compounds from textile industry effluents by biological methods is currently available. Recent promising research works on biological decolorization of textile effluent have showed that variety of microorganisms and plants capable of decolorizing wide range of anionic and cationic dyes. [1,2] This review article deals with the dye removing efficiency of fungi through biodegradation and biosorption mechanism. Discharge of colored effluents from dye manufacturing units and textile processing industries was produced by various textile industry process. The production of high amount of effluents mixes into water leading to pollution of aquatic systems and represent major environmental problems. Color was one of the most obvious indicators of water pollution and discharge of highly colored synthetic dye effluents can damage the receiving water bodies. Bioremediation using a variety of microbes for the degradation of xenobiotics seems a green solution to the problem of environmental pollution. Microbes have been gifted by nature with the ability of degrading a wide spectrum of environmental pollutants. Different fungi have the potentials to degrade complex and recalcitrant organic compounds into simpler fragments; sometimes

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achieving complete mineralization. Water pollution through industrial discharges, which is mainly in the form of effluent or wastewater, is one of the biggest problems. These effluents have strong concentrations of chemical oxygen demand (COD), phenol and its derivatives and often contain metals, inorganic nutrients, organic compounds, proteins, cyanides, chlorinated lignin and dyes. Bioremediation of toxic industrial effluents by microorganisms serves as an effective method to substitute the conventional recovery and removal processes. Fungal biomasses have huge capability of treating effluents discharged from various industries. White rot fungi are ubiquitous in nature and their adaptability to extreme conditions makes them good biode-graders. Their enzyme producing activity makes them effective decolorizers; they remove toxic metals by biosorbition ultimately rendering the effluents more ecofriendly. Fungal decolorization of dye effluent is receiving much consideration due to cost effective and less regeneration by microorganisms such as bacteria, fungi, yeast, algae, and plants.[3,4] Recent fundamental research works have revealed the existence of wide variety of microorganisms capable of decolorizing wide range of dyes. The use of microorganisms for the removal of synthetic dyes from industrial effluents offers considerable advantages this process was relatively inexpensive, running costs were low and the end products were completely mineralized with no toxicity. Degradation of dye is a complex process works to detoxify, decolorize, and degrade the dyes are done in lab scale only. Hence the need of effective complete conversion of textile effluent into useful liquid waste by using microbes is required.

Biodegradation has at least three step; a minor change in an organic molecule leaving the main structure still intact, fragmentation of a complex organic molecule in such a way that the fragments could be reassembled to yield the original structure, and complete mineralization i.e. transformation of organic molecules to mineral forms, including carbon dioxide or methane, plus inorganic forms of other elements that might have contained the original structures. It compared the colour removal efficiency of lyophilized fungal culture with lignin peroxidase, both the lyophilized culture and enzyme decoloured the dye. A well-defined lignin-degrading enzyme system, consisting of lignin peroxidase, Mn (II)-dependent peroxidase (MnP) and glyoxal-oxidase, was found to be involved in the dye degradation. Adsorption of dyes to the microbial cell surface is the primary mechanism of decolourization. Absorption was found to be important mechanism that contributes to the decolourization process, possibility to initial transformation of the dye degradation. Microscopic examinations showed that the fungal spores, instead of the fungal hyphae were actually absorbing the dye. A hydrophobic-hydrophilic interaction between the fungus and the dye might contribute to the absorption phenomenon. The dye colour decreased, when either the enzyme or the cell mass concentrations were increased indicating that the dye degradation occurred both by enzymatic action as well as adsorption to the cell mass. The concerted action of both led to greater degradation than single.[5,6]

The water is an essential resource for life on the planet and for human development. The textile industry is one of the anthropogenic activities that most consume water and pollute water bodies. Therefore, the present work aims to undertake a review on the main effects of the release of industrial dyes and the essential bioremediation mechanisms. The textile dyes significantly compromise the aesthetic quality of water bodies, increase biochemical and chemical oxygen demand (BOD and COD), impair photosynthesis, inhibit plant growth, enter the food chain, provide recalcitrance and bioaccumulation, and may promote toxicity, mutagenicity and carcinogenicity. In spite of this, the bioremediation of textile dyes, that is, the transformation or mineralization of these contaminants by the enzymatic action of plant, bacteria, extremophiles and fungi biomasses is fully possible. Another option is the adsorption. Despite some disadvantages, the bioremediation is essentially positive and can be progressively enhanced by modern biotechnological techniques that are related to the generation of more degrading and more resistant engineered organisms. This is a sustainable solution that provides a fundamental and innovative contribution to conventional physicochemical treatments. The resources of environmental biotechnology can, therefore, be used as tangible technological solutions for the treatment of textile dye effluents and are related to the ethical imperative of ensuring the minimum necessary for a quality life for the humankind.[7,8]

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II. DISCUSSION

Many scientist in the field of biology and chemistry have given significant contribution to science and technology by utilizing the natural resources .Rapid industrialization had introduced a lot of chemicals including dyes into the environment polluting the entire ecosystem. Coloration of the natural water bodies is not only undesirable from aesthetic point of view, but it also affects the aquatic flora and fauna by reducing the transmission of sunlight through the water surface. Azo dyes are the largest group of synthetic dyes and pigments with industrial application and over the worldwide production of 100,000 commercially available dyes. These methods cause a significant amount of sludge or may easily cause secondary pollution due to excessive chemical usage. High cost and disposal problems of the chemical and physical methods for treating dye wastewater make them inefficient to cop up pollution caused due to the textile industries . Microbial enzymes, such as laccase, lignin peroxidase, manganese peroxidase and azo reductase are assumed to play an important role in the degradation of lignin in their natural lignocellulosic substrates. As reported , the white-rot fungus degrades dye along with lignin. Most of the xenobiotic compounds and dyes are degraded with ligninolytic system in white-rot fungi. Their enzymes producing activity makes them effective decolorizers and the bio-accessible groups are present in the lignin structure, seem to be access points to the fungus ligninolytic enzymes produced by fungi.[9,10]

In a previous study, Decolorization of six synthetic dyes and two raw textile effluents (A and B) by eight basidiomycetous fungi was investigated. Among eight basidiomycetous fungi, fungal isolate RCK-1 decolorized textile effluent A maximally (42%), while fungal isolate RCK-3 was found to decolorize more of Congo Red (69%), Xylidine Ponceau 2R (100%), Poly R-478 (96%), Indigo Carmine (99%), Lissamine Green B (90%), Toluidine Blue (57%) and textile effluent B (54%), than the rest of fungi. Percentage decolorization of all synthetic dyes and textile effluents by the new fungal isolates RCK-1 and RCK-3 was higher compared to the most widely studied simultaneous lignin degrader, *Phanerochaete chrysosporium* and selective lignin degrader, *Pycnoporus cinnabarinus*, when tested in liquid cultures. A statistically significant positive correlation between laccase production and decolorization of dyes and effluents was obtained as compared to other ligninolytic enzymes (lignin peroxidase and manganese peroxidase) production. This showed the importance of the differential contribution of the different ligninolytic enzymes towards the decolorization of the synthetic dyes and textile effluents. The substantially higher ligninolytic enzyme production by the fungal isolates RCK-1 and RCK-3 also suggested their potential use for textile effluent treatment and other possible biotechnological applications.[11,12]

In another study, 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 24h 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

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strains BWPB 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).

In another investigation, the potential of *Pleurotus eryngii* was studied for decolorization ability of different types of textile dyes which were screened on liquid medium. During decolorization study on seven textile dyes, it was found that *P. eryngii* had the most efficient decolorization on reactive Blue 21 with decolorization percentage (98.4%). On the other hand, the immobilized *P. eryngii* on wheat straw, rice straw, sugar cane waste and wheat bran had biodegradation ability of Blue 21 dye. The immobilization of fungus on sugar cane waste gave more efficient decolorization of Blue 21 with decolorization percentage (83.5%). Also it was found that the decolorization percentage in case of free *P. eryngii* was (50.8%). The fungus has the ability to produce lignin-degrading enzymes as laccase, Mn peroxidase, peroxidase and catechol oxidase enzymes. Immobilized *P. eryngii* on sugar cane waste exhibited the ability to treat industrial textile effluent, 85 to 90% reductions of various nutrients including biological oxygen demand (BOD) and chemical oxygen demand COD were achieved and color removal reached to 96%. So, it was suggested that the immobilized *P. eryngii* might be applicable to textile effluent system. It was found that the reuse of the treated textile effluent with immobilized *P. eryngii* on sugar cane waste for irrigation of Basil plant and good result similar to that obtained by tap water.[13]

Decolorization of dyes was detected using the culture supernatant after centrifugation at 3,000 rpm for 30 min. Control was performed using the same medium but without the tested fungus. Assays were performed by using visible recording spectrophotometer (labomed, Inc) at a specific λ length for each dye. Color removal was reported as: Decolorization (%) = $(A_0 - A_t) / A_0 \times 100$ Where A_0 is the absorbance of initial dye solution, which is constant for each dye and equals 0% decolorization, and A_t is the absorbance at cultivation time, t . Decolorization percentage reading refers to the percentage mean of decolorization percentage of three replicas.[9,10]

Dye decolorization activity of *Pleurotus eryngii* was screened for seven dyes with the final dye concentration of 50 mg l⁻¹, efficiency of *P. eryngii* to decolorize different kinds of dyes: reactive Blue 21, reactive Yellow 160, reactive Orange 122, direct Congo red, direct Yellow 5G, acid Methyl orange and acid Eriochrome black T and the ability to decolorize textile dyes were screened on liquid medium. The dye Blue 21 was decolorized more than the other six dyes where the decolorization percentage of Blue 21, Yellow 160, Orange 122, Congo red, Yellow 5G, Methyl orange and Black T were 98.4%, 96.8%, 97.1%, 97.8%, 76%, 61.1% and 96.9% respectively. These results were correlated with that of Omar (2013) who reported that *Aspergillus niger* isolated from the soil samples around the textile distillery industries was evaluated for its efficiency in decolorization of different textile dyes; reactive red 198, reactive orange 122, reactive yellow 160, reactive blue 21 and reactive blue 19. Fungal biomass walls were composed of macromolecules which contain carboxyl amino, sulphates, and hydroxyl groups which act as metal sorption sites, and they are able to produce an array of enzymes to enable them to grow under a variety of conditions. The decolorization efficiency of fungi can be due the presence of chitin with hydroxyl and amino groups in their cell wall, which make them an efficient adsorbent of dye effluent. *Pleurotus eryngii* has a great potential to decolorize and degrade dyes in the free and immobilized cases, also the fungus has significant effect in treatment industrial textile effluents in immobilized case.[10,11]

III. RESULTS

Industrialization and other human impacts have placed increasing pressure on aquatic environments, with the generation of large quantities of toxic aqueous effluents containing different substances, such as synthetic dyes and

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other organic pollutants. It is estimated that between 10 and 15% of all dyes used in textile processes and other industries are discharged into wastewater, causing extensive aquatic pollution. Biological methods have been employed for the removal of color and toxicity from effluents containing azo dyes. Therefore, biosorption tests were performed in a study, with the dyes Acid Blue 161 e Procion Red MX-5B in simple and binary solutions, whereas biodegradation treatment was performed with the dyes only in binary solution. For biosorption, the dyes were removed by the fungi *Aspergillus niger*, *Aspergillus terreus* and *Rhizopus oligosporus*. The fungal biomass demonstrated good adsorption capacity to these compounds. The elimination of the toxicity of the solution after biosorption demonstrated the effectiveness of the treatment. Intense molecular changes after biodegradation treatment with the *A. terreus* fungus were demonstrated by the FTIR analysis. However, toxicity tests with *Lactuca sativa* seeds and *Artemia salina nauplii* indicated the presence of highly toxic metabolites in the reaction medium at the end of the treatment. Based on the findings, biosorption is more suitable for this type of treatment, since it was also capable of removing the molecules from the medium, with the advantage of impeding the formation of highly toxic by-products.[11,12]

Analyzing the findings, the more efficient treatment was biosorption with the pelletized biomasses of the fungi *A. terreus*, *A. niger* and *R. oligosporus*. The fungal biomass was able to remove the dye from the medium without degrading the molecules, which was very favorable due to the non-formation of highly toxic by-products. In the biosorption treatment, each dye binds to specific active sites on the fungal cell walls, as demonstrated by the greater removal of these molecules in the binary solution, indicating more efficient treatment due to the filling of all active sites. The high rate of decolorization obtained during treatment confirmed dye removal by biosorption in multiple layers. While the biodegradation treatment was quite efficient in achieving decolorization, the formation of by-products with high toxicological potential reduces the usefulness of the method. Biodegradation treatments with longer exposure times may be more efficient, since greater exposure to microorganisms enables the complete degradation and consequent mineralization of these metabolites, which are highly persistent in the environment and cause considerable harm to aquatic fauna and flora.[13]

IV. CONCLUSIONS

White-Rot Fungi used in textile dye wastewater degradation

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| Fast growing fungi | <i>Bjerkandera adusta</i> , <i>Ceriporiopsis rivulosa</i> , <i>C. subvermispora</i> , <i>Diplomitoporus lindbladii</i> , <i>Hyphoderma puberum</i> , <i>P. chrysosporium</i> , <i>P. sordida</i> , <i>P. gigantea</i> , <i>Phlebia concentrica</i> , <i>Ph. subserialis</i> , <i>Ph. tremellosa</i> , <i>Ph. albida</i> , <i>Ph. merismoides</i> , <i>Ph. rufa</i> , <i>Pycnoporus sanguineus</i> , <i>Trametes versicolor</i> , <i>T. villosa</i> , <i>T. hirsuta</i> , <i>Irpex lacteus</i> . |
| Slow growing fungi | <i>Acanthophysium lividocoeruleum</i> , <i>Ganoderma oregonense</i> , <i>Gymnopilus sp.</i> , <i>Heterobasidion annosum</i> , <i>Lentinula edodes</i> , <i>Mycoacia uda</i> , <i>Peniophora sp.</i> , <i>Peniophora pseudo-pini</i> , <i>Perenniporia subacida</i> , <i>Phellinus weirii</i> , <i>Phlebia rufa</i> , <i>Pleurotus ostreatus</i> , <i>Schizophyllum commune</i> , <i>Sistotrema brinkmannii</i> , <i>Stereum hirsutum</i> , <i>Stereum sanguinolentum</i> , <i>Trametes versicolor</i> , <i>Trichaptum abietinum</i> , <i>Trichaptum bifforme</i> , <i>Tyromyces sp.</i> |
| Little growing | <i>Acanthophysium lividocoeruleum</i> , <i>Armillaria mellea</i> , <i>Bondarzewia montana</i> , <i>Cystostereum pini-canadense</i> , |

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fungi *Dichostereum pallescens, Heterobasidion annosum, Pholiota adiposa, Pleuroflammula puberula, Polyporus varius, Schizophyllum commune, Sistotrema octosporum, S. brinkmannii, Stereum gausapatum, Trichaptum biforme.*

White-rot fungi produce several enzymes that have been related to their ability to degrade natural polymers, such as lignin and cellulose, but can also degrade different synthetic chemicals, usually recalcitrant to biodegradation. The promising results obtained with this ligninolytic fungus, lead to the study of the potentialities of other species of ligninolytic basidiomycetes. The role of fungi in the wastewater treatment has been proved them to be suitable and effective for dyes degradation/decolourization and the removal of colourants from the textile effluents. Dyes and pigments are being removed by these fungi either in living or dead form through bio-sorption, bio-degradation, bio-accumulation and enzymatic mineralization, using lignin peroxidase, manganese peroxidase, manganese independent peroxidase and laccase. Fungi has been shifted the research interest for the bio-treatment of wastewater ingredients such as metals, inorganic nutrients and organic compounds. Physico-chemical treatment processes, such as coagulation, precipitation, filtration, adsorption, photolysis and oxidation with hydrogen peroxide or ozone, can generate a large volume of sludge and usually require the addition of other environmental hazardous chemical additives. Biological treatment technologies are attractive alternatives to the traditional physicochemical methods, as they are low-cost, environmental friendly and can selective to provide a complete degradation of organic pollutants without collateral destruction of either the site's flora or fauna. Therefore, as better alternatives, biological processes are getting more and more attention since it does not produce harmful byproducts as well as large quantity of sludge. Fungi are not only used to decolourize/degrade the hazardous dyes (i.e. azo, anthraquinone, heterocyclic, triphenylmethane and polymeric dyes) but also used to have some beneficial products from the wide range of waste materials, by their bio-transformation. Investigations on the topic have been emphasized the biodegradation processes. There should be further advancements in mycoremediation processes targeting the application of fungi in degradation/decolourization of textile dyes and other hazardous effluents.[14]

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