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Biodegradation of dyes in textile wastewater using local fungal isolates

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Abstract Textile wastewater contains dyes and excessive amounts of nitrogen, phosphorus, and metal compounds, as well as organic pollutants that. Wastewater also contains chemical wastes that are not biodegradable that can cause infectious disease. The chemical and biological waste in sewage and water must be broken down before it is deposited to the soil and environment. The extensive use of dyes often causes pollution problems. The presence of very low concentrations of dyes in large water bodies is shown to be highly visible and indisputable and also reduced light penetration and photosynthesis. In addition some dyes either toxic or mutagenic and carcinogenic. In this study, waste water was treated by microbial isolates from Egyptian soil. The potent fungal isolate used for the degradation of the excess dye used in textile industry and waste water. The study investigated the biodegradation process under different growth conditions. Different parameters were involved , dyes concentration, inoculum size incubation time, temperature, and growth medium. The decolorization efficiency for these dyes were investigated. Reactive yellow 145 is used the maximum wave lengthof 475. These isolates were belong to species such as Aspergillus niger, A.ochraceous, Mucor recemosus , Penicilluim notatum and P. chrysogenium. After they screened for optimum efficiency and the condition for temperature and pH which optimized and the effectiveness of biodegradation process. The results showed that the maximum decolorization 65.77% was obtained at pH 6.5, fermentation time of 6 day, dye concentration of 90mg/h, and agitation rate 200 rpm.at $30C^0$.

Keywords: Biodegradation, Fungi, Textile wastewater, Dye, Decolorization

Introduction

One of the biggest environmental problems is the removal of dye from wastewater (Kim *et al.,* 2004; Park *et al.,* 2007). According to Garg *et al.* (2004), dyes are used extensively in a variety of industries, such as textile, leather, paper, printing, plastic, food, etc., to color their products. The microbial degradation of dyes depends on a number of factors, including oxygen, carbon, or energy sources, as well as ideal conditions like pH and temperature. The widespread usage of dyes frequently leads to pollution issues. In addition to being extremely

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obvious and undeniable, the presence of extremely low dye concentrations in big bodies of water also inhibits light penetration and photosynthesis. Furthermore, some dyes are carcinogenic, mutagenic, or poisonous. (Nigam *et al.,* 2000; Birhanli and Oznen, 2005; Degon *et al.,* 2005; Gong *et al.,* 2005).

There is significant environmental degradation as a result of the discharged wastewater contaminating land and water. It may also be poisonous and mutagenic to aquatic plants and animals, change pH and oxygen levels, and prevent light from penetrating the water, disrupting the aquatic ecology. The remaining dyestuff is also linked to a number of negative health effects in humans, such as immune system disorders, respiratory issues, and inflammation. Between 10 and 200 mg/L of dye, along with a variety of other organic and inorganic compounds and additives, can be found in colored wastewater discharged by the textile industry. Indeed, it is believed that up to 90% of these pigments remain chemically unaltered following wastewater treatment and are still released into rivers. The breakdown of dye molecules depends on the complexity of the dye structures (Ali and EL-Mohamedy, 2016).

Novel treatment methods are desperately needed to speed up the slow rate of decomposition of dyes present in wastewater because many dyes and pigments are toxic and hazardous to aquatic life and humans at the concentration at which they are discharged to receiving water bodies. Conventional methods of removing dyes used in the textile industry are resistant to light, oxidizing agents, and biodegradation processes (Salar and Kumar, 2012). The use of certain bio-sources, such as bacteria and natural plant components, is crucial for wastewater treatment. Wastewater can support the growth of certain microorganisms, such as yeast, bacterium and fungi, which used the pollutants as a substrate. The water shortage issue was resolved by indirectly converting the wastewater into more usable forms for certain industrial and agricultural applications (Hassan *et al.,* 2013).

Wool and silk dyeing water waste resulted from fabrics containing reactive colors and acids. Synthetic colors pollute the environment and are poisonous and carcinogenic. Like other industries, the textile sector contributes to environmental pollution through the waste products that are created from the use of chemicals and dyes. In terms of environmental concerns, evaluation of the degradation of chemicals and dyestuffs is crucial (Ali and El- Khatib, 2010) Some studies using various types of microbes sush as *Penicillium* spp. and Pseudomonas aeruginosa for biodegradation of textile dyes from wastewater (Ali and El-Mohamedy, 2012; Mohamed *et al.,* 2014).

The aim of the present study was to investigate the effect of usimg some fungal isolates on the degradation of dye staff remaining on textile industry and

optimization of the decolorization process using the potent selected fungal strains.

Materials and methods

Fungal isolates

The fungal isolates were used as *Aspergillus flavus*, *Aspergillus ochraceous*, *Mucor recemosus*, *Penicilluim notatum* and *P. chrysogenum*. They were isolated from agricultural soil samples from Giza governerate, Egypt.

Fermentation process

The fungal strains was grown in modified DoX medium which consisted of g/l) sucrose 30 g, – dipotassium phosphate 1g, magnesium sulfate 5 g, 0.5 potassium chloride 0.5 g, iron sulfate 0.01g in 1 litre, It was sterilized at 120C^{o}, 1.5 atm for 20 min. Then after cooling to room temperature, it was transferred by the tested fungal strains and the used dye was added then incubated for 6 days on rotatory shaker at 150rpm at 30C^{o}.

Quantification of dye decolonization

The decolourization assay was expressed in the terms of percentage of decolourization using spectrophotometer. The decolourization percentage in dye concentration was calculated for each treatment. It was determined by measuring the absorbance of culture supernatants at absorbance maxima of respective dyes after centrifugation at 10,000 rpm for 15 min (Ali and El-Mohamedy, 2012).
Decolarization % = (initial absorbance − final absorbance)/ initial absorbance × 100

Experiment of dye solution

The dye stock solution was prepared by dissolving accurately weighed dye in distilled water to the concentrations of 500 mg/L. Different concentrations were prepared from the stock solutions of 30, 60, 90, `120, 240 mg/L. The dye used in this study is reactive yellow of 145, the maximum wave length of 47.

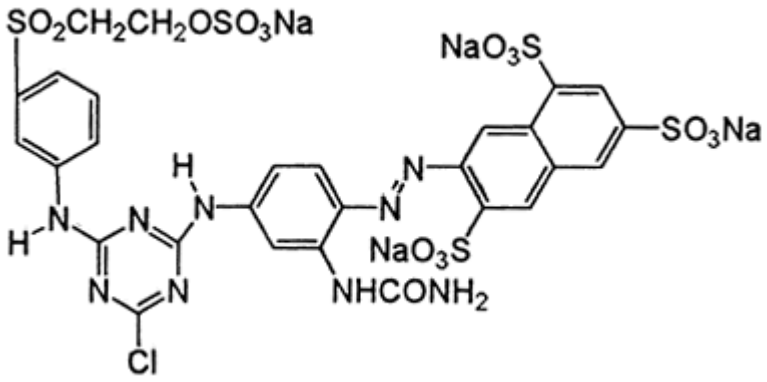

Figure 1. Chemical structure of Reactive yellow 145, the maximum wavelength 475

Results

Estimation of the maximum absorbance of used dye

The results indicated that the maximum wavelength at concentration of 90 mg/L as shown in Table 1 and also the absorbance exhibited the highest value of 0.019 at this concentration of maximum wavelength of 729.5 nm.

Table 1. Estimation of the maximum absorbance of used dye (stander)

Concentrations	Absorbance	Wave length
0 mg/L Control	0.061	414.5
30 mg/L	0.013	635.5
60 mg/L	0.013	662.7
90mg/l	0.019	729.5
120 mg/L	0.016	699.5
240 mg/L	0.016	636.5

Effect of concentration of dye under investigation on the wavelength

Different concentrations were prepared from the stock solution and treated with the fungal isolates under investigation. As the concentration increased s with respect to the treated samples as compared to the untreated until 1.5 mg/L the wavelength increases. The results indicated that the maximum wavelength obtained at concentration of 1.5mg/L as shown in Figure 2.

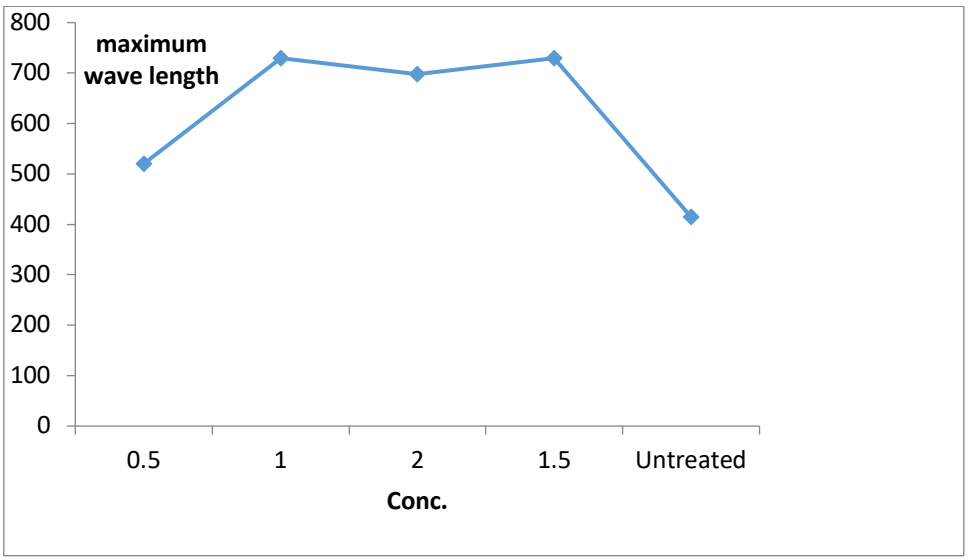

Figure 2. Effect of concentration of dye under investigation on the maximum wavelength

Screening for the potent fungal strain

In this experiment the screening procedure for the best decolozining strain were tested. Result showed that the best fungal strain for the decolorization of the dye used was Mucor recemosus (Table 2). It gave 46.17% as compared to the other fungal strains.

Table 2. The decolorization % and the final absorbance using different fungal strain

Fungal strain	Biomass (g/L)	Final absorbance	Decolorization %
A. flavous	1.77	2.41	23.51
A. ochraceous	2.15	2.55	22.66
M. racemosus	2.34	1.22	46.17
P. notatum	2.10	2.17	37.50
P. chrysogenum	1.89	2.35	33.72

Effect of decolonization time

In the present experiment the decolorization percentwas investigated at different time intervals. The results showed that the maximum decolorization (46.17) was obtained at 6 days (Table 3). At the beginning of growth reduced decolization capacity was shown as 22.87 and 27.64. At the longer time intervals of 7 and 8 days. The decolorization percent were remarkably stable of 46.13 and 45.91 %, respectively as shown in Table 3 and Figures 3-6.

Table 3. Effect of different fermentation time on the bioremediation process

Fermentation time (day)	Biomass (g/l)	Final absorbance	Decolorization %
1	1.45	2.21	22.87
2	1.89	2.22	27.64
3	2.12	1.69	31.98
4	2.26	1.45	34.56
5	2.30	1.33	38.72
6	2.34	1.22	46.17
7	2.15	1.22	46.13
8	1.99	1.21	45.91

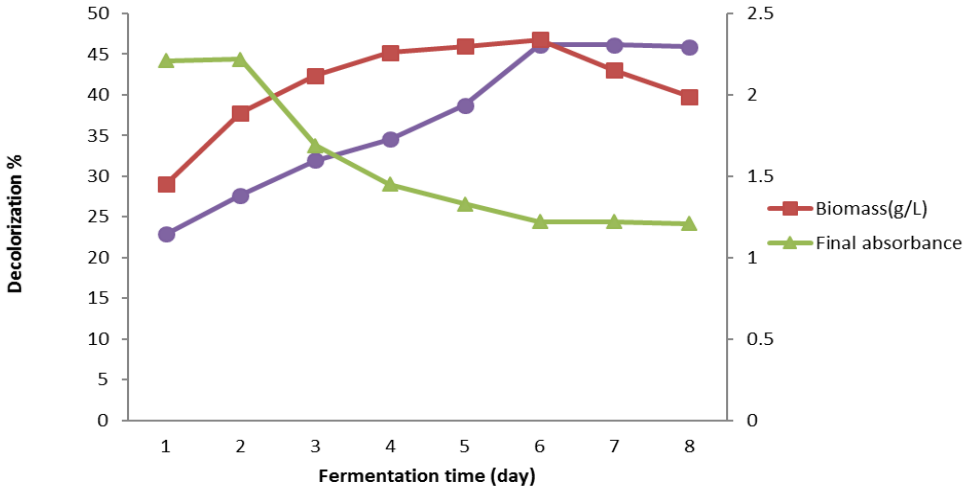

Figure 3. Effect of different fermention time on dye bioremdation

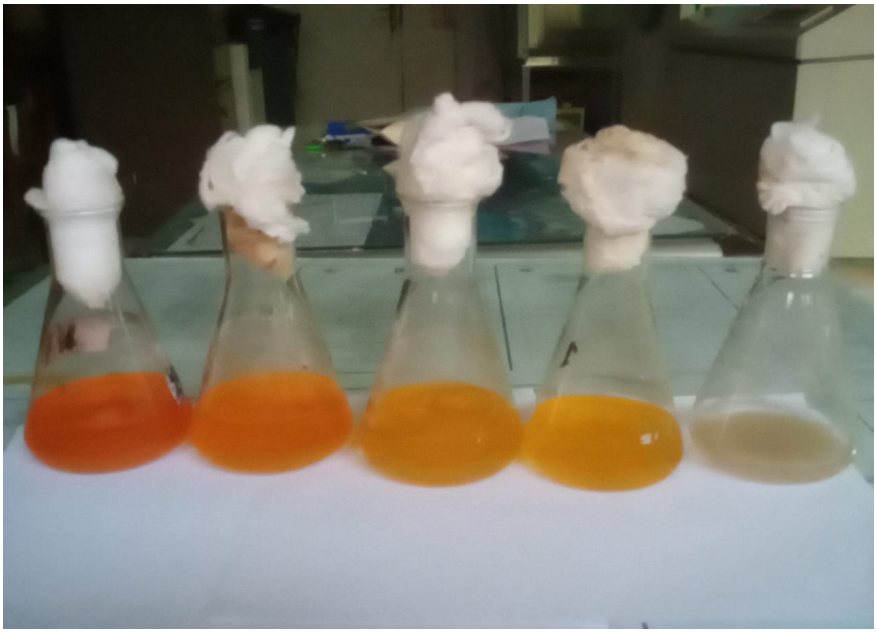

Figure 4. Bioremediation of dye at different time of incubation

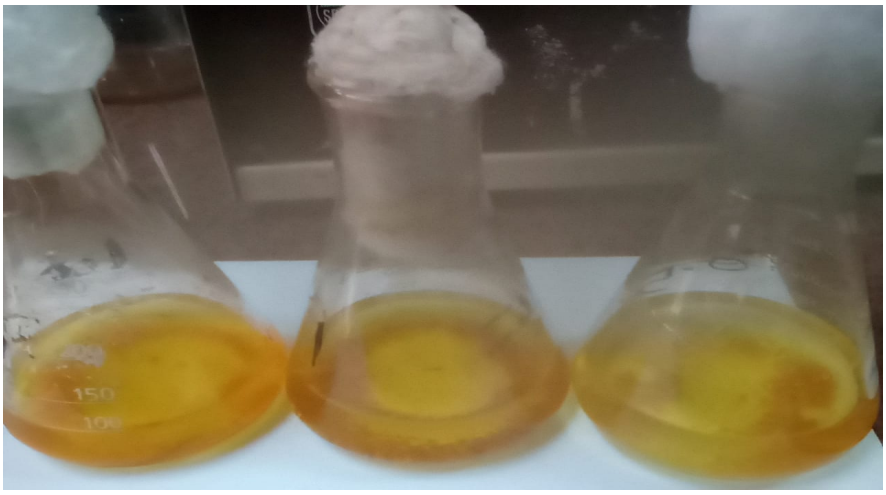

Figure 5. Before treatment of dye using *Mucor racemosus*

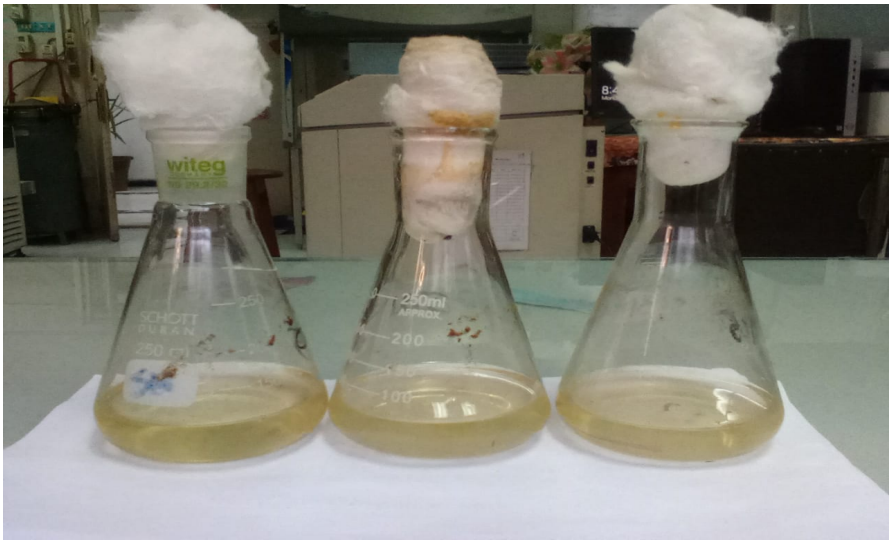

Figure 6. After treatment of dye using *Mucor racemosus*

Effect of pH value of the de colorization percent

The present experiment tested the decolorization efficiency at different pH value of the fermentation medium. Result revealed that the de colorization percent was increased in acidic medium (Figure 7). The best decolorization of 55.92% was obtained at pH 6.5. It was noticed that the basic medium of 88.5 and 9.5 remarkable decreased in de colorization of 31.58 and 33.70 %, respectively.

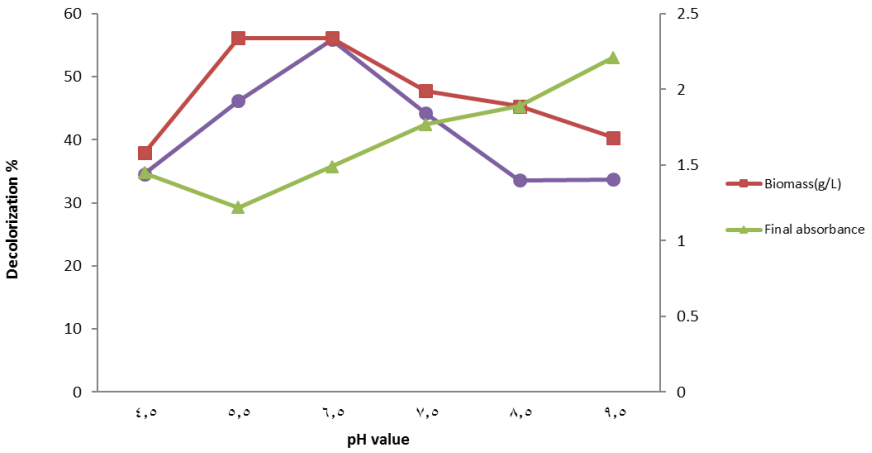

Figure 7. Effect of different pH value of the fermentation medium on dye bioremdation

Effect of dye concentration

The effect of different dye concentration on the decolorization process was tested. It was found that an increasing in the decolorization capacity of tested fungal strain by increasing the dye concentration and reached to the maximum of 65.77% at 90 mg/L. On the other hand, the higher concentration of 120mg/L gave noticeable decreased of 48.40% (Figures 8 and 9).

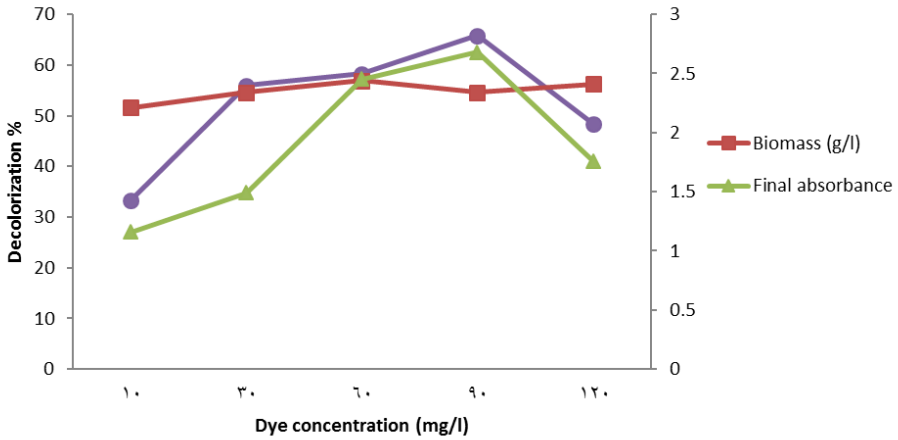

Figure 8. Effect of different dye concentration on the bioremediation

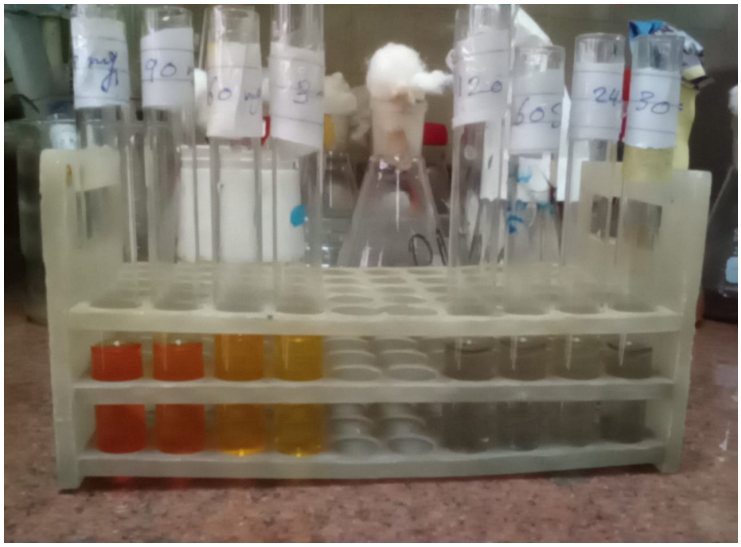

Figure 9. Effect of different dye concentrations

Effect of agitation speed(rpm)

In this experiment the effect of different agitation speed affected on the decolorization efficiency due to their effect on the availability of the dye to the cells. The maximum decolorization of 65.77% as obtained at 200 rpm. At agitation rates of 100 and 150 rpm, the decolorization percent were 44.64 and 51.70 %, respectively. At higher speed of 250 and 300 rpm with low decolization of 61.88 and 60.91 % were noticed (Figure 10).

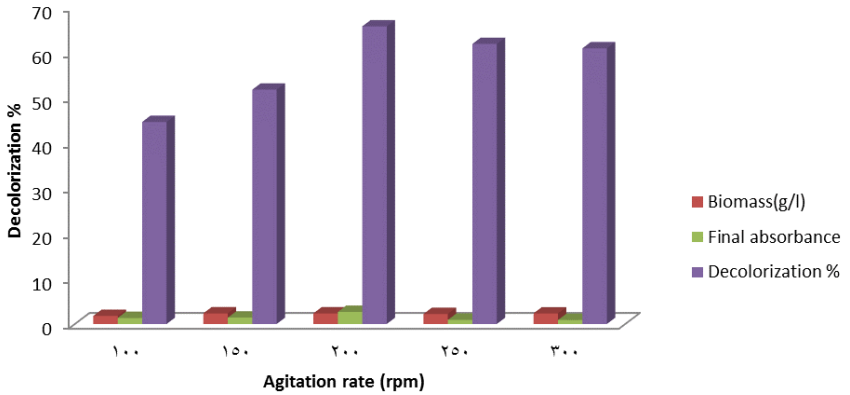

Figure 10. Effect of different agitation rate on dye bioremediation

Discussion

Some fungi possess ligninolytic enzymes which play an important role in the degradation of lignocellulose. These lignin degradation process involved not only in the degradation of lignin in their natural lignocellulosic substrates, but also in the degradation of various xenobiotic compounds, including dyes. Moreover, ligninolytic fungi have been reported to oxidize many recalcitrant substances such as chlorophenols, polycyclic aromatic hydrocarbons (PAHs), organophosphorus compounds, and phenols (Wesenberg *et al.,* 2003). Biosorption of dye occur essentially either through complexation, adsorption by physical forces, precipitation, entrapment in inner spaces of fungal mycelium, ion exchange due to surface ionization, and by formation of hydrogen bonds. Due to an increased cell-to-surface ratio, fungi have a greater physical contact with the environment. The incubation time affects on the degradation process. The results showed that during the beginning of growth a remarkable increase in the decolorization process was noticed. The maximum decolorization was obtained after 6 days incubation .

The pH value of the bioremediation medium are clearly affected on the process. The best decolonization was obtained at pH 6.5. It was noticed that the acidic medium of Mian *et al.,* 2024) stated that enhanced the decoleization by using *M. racemuses*. The dye concentration was found to affect on the decolorization process. The results revealed that the used fungal strain showed great tolerance of dye which showed high bioremediation at 90mg/L, which decreased due to the dye toxicity to fungal cells (Geertanjali *et al.,* 2021). This reflected the tolerance of *Mucor racemosus* against high dye concentrations which supported its application in industrial field. At the higher concentrations of 90 mg/L the decolorization was decreased.

The agitation rate is clearly affected on the availability of the used dye to fungal cell (Kanmani *et al.,* 2011). The best decolorization rate was obtained at 200 rpm. Biosorption of dyes occur essentially either through complexation, adsorption by physical forces, precipitation, entrapment in inner spaces of fungal mycelium, ion exchange due to surface ionization, and by formation of hydrogen bonds. Due to an increased cell-to-surface ratio, fungi have a greater physical contact with the environment. Thus, some fungi have demonstrated better dye adsorption potential exceeding that of activated charcoal. Additionally, it is not unusual for some species to demonstrate both enzyme-mediated degradation and bio sorption in the decolorization of textile dyes (Park *et al.,* 2007). It is thus feasible that in addition to the production of extracellular enzymes, the ability of the soil fungi to decolorize synthetic dyes is coupled also with their bio sorption abilities (Camarero *et al.,* 2005).

The biodegradation of dye is an important process, results in the remove of the hazardous compounds specially the phenolic compounds from the environment. Fungi paly a valuable role in this process specially lignin degrading fungi. The results of our study reveals that the fungus *Mucor racemosus* showed the priority compared to the tested fungi. The maximum decolorization (65.5%) was obtained by using the selected fungus grow on dox medium, at pH 6.5, for 6 days growth, agitation nrate 200rpm at dye concentration 90mg/L.

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