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Bioremediation of Azo Dyes Using Fungi

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ABSTRACT

Color is the main attraction of any fabric. No matter how excellent its constitution, if unsuitably colored it is bound to be a failure as a commercial fabric. Use of synthetic dyes has an adverse effect on all forms of life. Presence of sulphur, naphthol, vat dyes, nitrates, acetic acid, soaps, enzymes chromium compounds and heavy metals like copper, arsenic, lead, cadmium, mercury, nickel, and cobalt and certain auxiliary chemicals all collectively make the textile effluent highly toxic. Decolorization of textile dyes using fungi viz., *Aspergillus niger*, *Aspergillus flavus*, *Fusarium sp*, *Mucor sp*, *Trichoderma*, *Aspergillus fumigatus*, *Aspergillus nidulans*, and *Penicillium* were isolated from the soil samples collected from textile industries in Arni. These isolates were tested for decolorization of textile dyes such as Remazol Red and Remazol Black. The results showed that all the eight fungal species were found to be effective in decolorization of textile dyes. Among the isolates *Aspergillus niger* and *Aspergillus flavus* were found to be more effective against decolorization of Remazol Red and Remazol Black respectively. This study concluded that the bioremediation process is ideal to reduce dyes toxicity with low-cost and environmentally friendly.

KEYWORDS: *Azo Dye, Ultraviolet-visible spectroscopy, Aspergillus niger*

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INTRODUCTION

In Tamilnadu textile industries are of the important industry in several districts known for Tirupur plays a vital role in textile industries, the other areas like Kanchipuram, Arni and some areas take part on it. Ultimately these textile industries release the colored effluents with dyes and other toxic materials in to open environment. Dye is defined as a substrate used to impart color to substrate. It is widely used for coloring diverse materials¹. There are more than 10,000 commercially available dyes and pigments are used in various dyeing and printing industries. And around 7 x 10 tones of dyes produced annually of which 10-15% lost in the effluents during process². There are thousands of synthetic azo dyes used in the textile, pharmaceutical, cosmetics and food industries, of which more than 500 contain potentially carcinogenic aromatic amines in their chemical formulation³. When these compounds enter in to the human body through ingestion of food, dermal contact and inhalation, they are metabolized via azo-reductases to aromatic amines by the intestinal micro flora and in the mammalian liver⁴. A color in waste water is highly visible and affects esthetics, water transparency and gas solubility in water bodies, and especially because many dyes are made from known carcinogens, such as benzidine and other aromatic compounds dye wastewaters have be treated⁵. Biological decolorization can be achieved by the use of a number of naturally occurring microorganisms such as fungi and bacteria. Between this the fungi are the most potent decolorizers. In recent years there has been an intensive research on fungal decolorization of dye wastewater. It is becoming a promising alternative to replace (or) supplement present treatment process living (or) dead cells which are capable of decoloring dye wastewaters⁶, so in order to treat the effluents effectively and to remove the colorants, the micro-organisms capable of specifically degrading the compounds present in the textile effluent are cultured on a large scale and introduced into effluent treatment plants.

MATERIALS AND METHODS

The soil samples were collected near the places where the effluents are discharged around the factories situated in Arni. Soil samples were subjected to serial dilution to get dilutions and inoculated on to the sterile Petri dishes which consist of sterile, cool, molten potato dextrose agar medium and then we followed the spread plate method and kept the plates for incubation at 37°C for 3 days. Fungal colonies were identified using microscope and with the help of standard manuals.

Decolonization Study

The dye samples were collected by the textile industry situated in Arni. The fungal species which are identified as *Aspergillus niger*, *Aspergillus flavus*, *Aspergillus nidulans*, *Aspergillus fumigatus*, *Trichoderma species*, *Fusarium species*, *Penicillium species* and *Mucor species*. The isolated fungi were tested for its ability to decolorize textile dyes, Textile dyes, Remazol Red ($\lambda_m=476\text{ nm}$), Remazol Black ($\lambda_m=574\text{ nm}$) were used.

The 50ml of Ashthana and Hawkens broth was amended separately and 1ml of respective dyes were subsequently inoculated with 0.05ml of fungal spore suspension is added on the flask were kept in the shaker for 21 days at 7 days interval for observation samples were centrifuged at 10000xs for 10 min decolorization was assessed by measuring absorbance of the supernatant with the help of uv-spectrophotometer at wavelength maxima (λ_m) of respective dye, controls were maintained for each dyes.

The percentage decolorization was calculated using following formula

$$\text{Decolorization} = \frac{\text{Initial OD} - \text{Final OD} \times 100}{\text{Initial OD}}$$

After decolorization study the comparative study and fungal biomass was examined.

RESULTS AND DISCUSSION

The microbes isolated from textile soil sample were characterized and identified as *Aspergillus niger*, , *Fusarium sp*, *Aspergillus nidulans*, *Aspergillus fumigatus*, *Aspergillus flavus*, *penicillium sp*, *Mucor species* and *Trichoderma sp* (fig 1. a,b,c,d,e,f,g,h). This showed decolorization of different textile dyes as Remazol Red, Remazol Black in liquid medium (Ashthana and Hawkens medium). The range of decolorization activity was found to be 95.9%, 94.4%, 94.2%, 93.5%, 92.3%, 91.8%, 88.4%, 87.1%, with *Aspergillus niger*, *Aspergillus flavus*, *Aspergillus nidulans*, *Aspergillus fumigatus*, *Penicillium sp*, *Mucor sp*, *Fusarium sp*, *Trichoderma sp* respectively after 21 days of incubation. In Remazol Red the maximum decolorization was given by the fungi *Aspergillus niger* (95.9%) (fig 2). similarly in Remazol Black, the range of decolorization activity was found to be 96.4%, 91.4%, 91.2%, 91.0%, 86.9%, 86.7%, 85.6%, 84.3% with *Aspergillus flavus*, *Fusarium sp*, *Aspergillus niger*, *Penicillium sp*, *Aspergillus fumigatus*, *Mucor sp*, *Aspergillus nidulans*, *Trichoderma sp* respectively after 21 days of incubation. In Remazol Black the maximum decolorization activity was given by the fungi *Aspergillus flavus* (96.4%) (fig.3). the percentage decolorization of Remazol Black is slightly higher than Remazol Red.

As far as *Aspergillus nidulans* and *Aspergillus fumigatus* is concerned there is hardly any literature is available about their use for decolorization of dyes is concerned. In our study it is proved that these fungi are also showed the potential to degrade the dyes which is equal to *Aspergillus niger* and *Aspergillus flavus*. *Trichoderma sp*, *Mucor sp* and *Fusarium sp* are also used in this process this all fungi also degrade (80%) of textile dyes. White rot fungus *Thelespora sp* is a choice organism which was used for decolorization of Azo dyes, as Congo red and amido Black⁷. Then *Phanerochaete chrysosporium* also used for decolorization study and clean up 98% of original textile waste water^{8,9}. In our study *Aspergillus flavus* and *Aspergillus niger* were found to be efficient decolorizer in Remazol Red and Remazol Black. During bioremediation, we are using a special medium (Asthana and Hawkers broth). In this broth the fungi decolorize the dyes frequently at the same time the mass multiplication of fungal spore also increased, this is an original mechanism in our study have been examined. In final day the fungal biomass was calculated by using the formula (weight of mycelium-weight of filter paper) table.1. This study concluded that the bioremediation process is ideal to reduce the dyes toxicity with low cost and environmently friendly.

TABLE 1: Decolorization Percentage of Remazol red with the help of UV -spectrometer.

Isolates	7 th DAY		14 th DAY		21 st DAY	
	pH	D%	pH	D%	PH	D%
<i>Aspergillus niger</i>	4.5	76.5	4.7	77.2	4.8	95.9
<i>Aspergillus Flavus</i>	3.4	50.3	4.2	85.5	4.6	94.4
<i>Pencilium sp</i>	7.3	66.9	7.5	79.7	7.9	92.3
<i>Aspergillus nidulans</i>	4.3	80.2	4.8	82.9	5.3	94.2
<i>Trichoderma sp</i>	6.8	52.9	6.9	76.3	6.9	87.1
<i>Mucor sp</i>	7.4	53.2	7.6	80.5	7.9	91.8
<i>Aspergillus fumigatus</i>	7.0	49.0	7.2	87.7	7.5	93.5
<i>Fusarium sp</i>	6.8	55.6	6.9	84.2	7.3	88.4

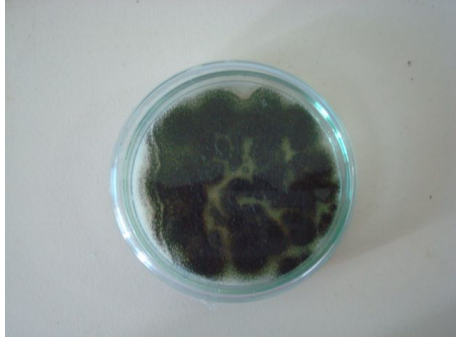
TABLE 2: Decolorization percentage on Remazol black with the help of uv -spectrometer

Isolates	7 th DAY		14 th DAY		21 st DAY	
	pH	D%	pH	D%	PH	D%
<i>Aspergillus niger</i>	4.5	10.7	4.7	85.8	5.1	91.2
<i>Aspergillus flavus</i>	7.1	30.7	7.4	95.5	7.5	96.4
<i>Penicillium sp</i>	6.7	28.2	6.9	89.5	7.2	91.0
<i>Aspergillus nidulans</i>	5.9	58.7	6.3	79.0	6.5	85.6
<i>Trichoderma sp</i>	6.9	45.8	7.1	78.6	7.4	84.3
<i>Mucor sp</i>	7.0	53.7	7.3	75.7	7.6	86.7
<i>Aspergillus fumigatus</i>	7.2	59.1	7.5	70.6	7.7	86.9
<i>Fusarium sp</i>	5.7	51.4	5.9	64.7	6.2	91.4

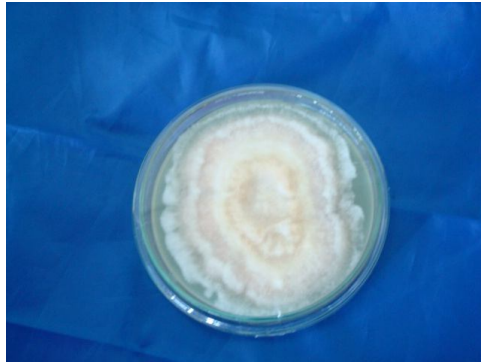
Table 3: Biomass on 21st day :(g)

S.NO	ISOLATES	REMAZOL RED	REMAZOL BLACK
1	<i>Aspergillus niger</i>	1.7	1.1
2	<i>Aspergillus flavus</i>	1.9	1.3
3	<i>Penicillium sp</i>	2.54	1.8
4	<i>Aspergillus nidulans</i>	1.68	2.0
5	<i>Trichoderma sp</i>	2.75	2.4
6	<i>Mucor sp</i>	1.2	1.93
7	<i>Aspergillus fumigatus</i>	1.3	1.8
8	<i>Fusarium sp</i>	2.54	1.1

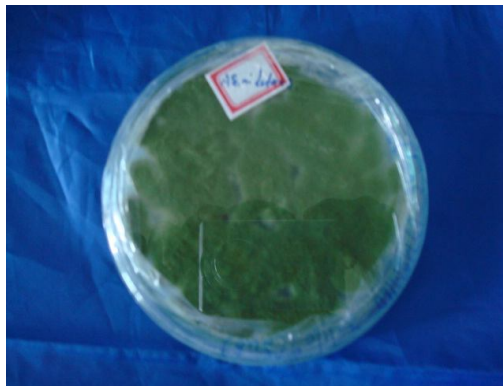
a) *Aspergillus niger*



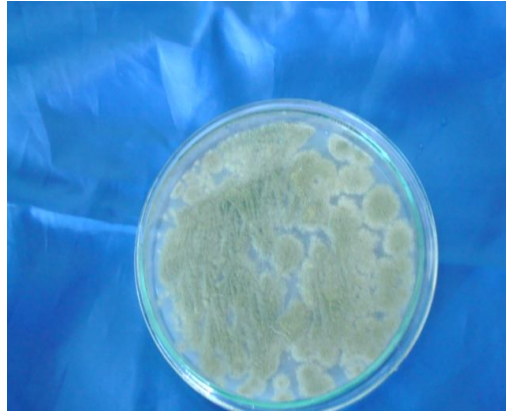
b) *Fusarium species*



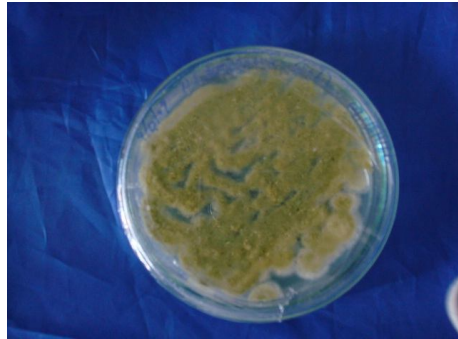
c) *Aspergillus nidulans*



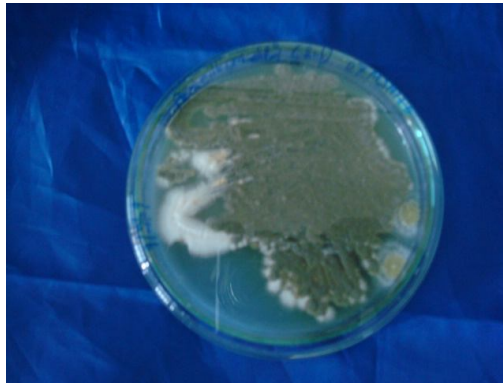
e) *Aspergillus fumigatus*



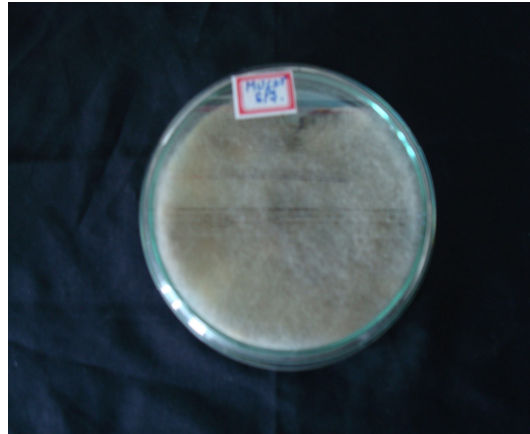
f) *Aspergillus flavus*



g) *Penicillium species*



h) *Mucor species*



i) *Trichoderma species*

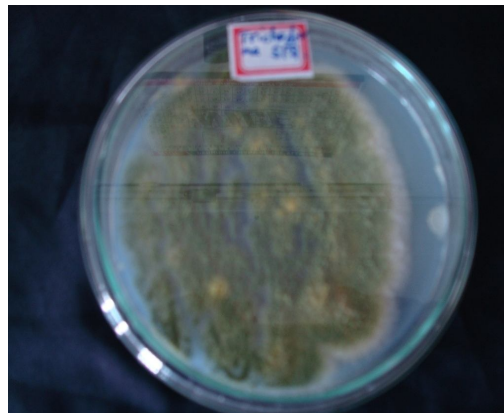


Figure 1. microbes isolated from textile soil sample were characterized and identified.



Figure.2 Remazol Red the maximum decolorization was given by the fungi *Aspergillus niger* (95.9%)



Figure.3 In Remazol Black *Aspergillus flavus* (96.4) shows effective decolorization compare to other fungal species.

CONCLUSIONS

In summary, the decolorization of textile dyes using *Aspergillus niger*, *Aspergillus flavus*, *Fusarium sp*, *Mucor sp*, *Trichoderma*, *Aspergillus fumigatus*, *Aspergillus nidulans*, and *Penicillium* were successfully studied. Among the isolates *Aspergillus niger* and *Aspergillus flavus* were found to be more effective against decolorization of Remazol Red and Remazol Black respectively; This study concluded that the bioremediation process is ideal to reduce dyes toxicity with low-cost and environmentally friendly.

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