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Effect of Decolourization of Malachite Green by *Aspergillus* and *Fusarium* under Agitation

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Decolourization of the selected dye malachite green by *Aspergillus* sp. and *Fusarium* sp. was observed under agitation. Decolorization of malachite green under agitation was effective with *Fusarium* sp. 71–82% of dye decolourization was obtained at the end of 72 hours by Fusarium and 22–65.4% by *Aspergillus*. The results obtained in this study showed that the decolorization of malachite green under agitation was found to be the effective with *Fusarium*.

Key words: Biodecolourization, *Aspergillus*, *Fusarium*, Malachite green, Congo red, Amylase.

Dyes are chemicals which on binding with a material will give color to the material. Dyes are ionic, aromatic organic compounds with structures including aryl rings which have delocalized electron system. Colored dye wastewater arises as a direct result of the production of the dye and also as a consequence of its use in the textile and other industries (Allen *et al*., 2005). In a textile industry, a number of dyes chemicals and auxiliary chemicals are used to impart desired quality in nature and contains high concentration or BOD, COD, TDS and alkalinity. It can cause environmental problem unless it is properly treated before disposal. Also, the resistance of dyes to degradation in biological treatment systems is of concern.

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Approximately 10,000 dyes and pigments are industrially used and 0.7 million tones of synthetic dyes are produced annually worldwide (Govindwar *et al.*, 2006). As dyes are relatively resistant to biodegradation the elimination of the colored effluents in wastewater is mainly based on physical and chemical methods (Banat *et al.*, 1996). But these procedures generate a significant amount of secondary pollution (Verma *et al*., 2003; Zhang *et al*., 2004).

It is due to these reasons; microbial degradation is preferred and observed. Microbial pollution treatment systems have the advantage if being simple in design and low in cost compared with conventional treatments. Development of efficient dye decolorization requires a suitable strain and its use under favorable conditions to realize the decolorization potential (Bhaskar *et al.*, 2007).

Malachite green is a triphenylmethane dye used both as a dye to color synthetic fibres and silk and to dye paper products. Furthermore the substance is used as veterinary medicinal product for the treatment of ornamental fish. The

potentiality applications of *Aspergillus* sp. for decolorization were investigated. *Fusarium* sp. was not much used in the decolorization process as that of other fungal organisms. Dye removals from wastewater by established Waste Water Treatment Plant are expensive and need careful application. Furthermore, following anaerobic digestion, nitrogen containing dyes are transformed into aromatic amines, that are more toxic and mutagenic than the parent molecules. To overcome these difficulties, fungi are being investigated for their potential to decolorize effluents (Coulibaly *et al*., 2003).

Dyes are removed by fungi by biosorption, biodegradation and enzymatic mineralization. However one or more of these mechanisms could be involved in color removal, depending on the fungus used. It is reported that raw effluent can only partially be decolorized upon fungal treatment. The weak decolorization of these effluents by complete culture was influenced by temperature, pH, salts, inhibitory molecules, carbon and nutrients within these solutions (Coulibaly *et al.*, 2003).This study is focused on the effect of *Aspergillus* sp. and *Fusarium* sp. under agitation to decolorize malachite green.

Methodology

Isolation and identification

The fungal organisms *Aspergillus* sp. and *Fusarium* sp. were isolated from groundnut and potato respectively. The spoiled groundnut and potato were collected and inoculated on the sterile Czapek's dox agar plates aseptically. The plates were then incubated at 27 ºC for 48 hours.

Plating method

The ability of the organism to decolorize the dye was initially tested by plating method. After sterilization, 1.5 gm/100 ml of dye was added to the medium before plating. After solidification, the plates were inoculated with the isolated fungi. The plates were incubated at 27 ºC for 3–4 weeks and observed for decolorization.

Free cell method

The organism isolated were sub cultured in the Czapek's dox broth and incubated at 27 ºC. The culture filtrate was isolated. 5 ml of the culture was added to each concentration of the dye solution (1, 3, 5, 7, 9 mg/100 ml) in the flask. The flasks were incubated at 27 ºC for 72 hours in the shaker at 150 rpm for agitation or in the incubator for static condition.

Decolorization method

Each concentration of the dye was diluted in 100 ml of distilled water. The fungal cultures were added to the dye solution. The flasks were then incubated in the shaker at 150 rpm for agitation. After 72 hours the flasks were observed for decolorization. The concentrations were centrifuged at 2000 rpm for 20 minutes. Supernatant was collected and OD value was taken at 620 nm. (Table 1)

RESULTS AND DISCUSSION

Identification of isolates

Microscopic identification was done to identify *Aspergillus* sp. Black spores was observed at 30–37 ºC on the plate after 48 hours. The reverse mat was found to be yellowish in color. Microscopic identification was done to identify *Fusarium* sp. White cottony layer was observed at 25–30 ºC on the plate after 48–72 hours.

Plating method

The ability of the organism to decolorize the dye was initially tested by plating method.

Table 1. Decolourization percentage of malachite green using *Aspergillus* and *Fusarium* under agitaiton

Dyes used	Concentration of Dye (mg/100 ml)	Percentage of decolourization	
		Aspergillus	*Fusarium*
Malachite green	1	58	82
	3	65.4	77
	5	48	76.4
	7	43	71
	9	22	71

Decolorization was observed in the plates after 5 weeks. This duration was may be due to the solid state condition of the medium or amount of dye added may be a tuff factor for the organism to decolorize. With malachite green, the green color of the medium was decolorized to pale bluish green color.

Decolorization of malachite green by *Fusarium* under agitation

Decolorization of malachite green under agitation was effective with *Fusarium* sp. Each concentration showed decolorized effect from 71-82% at the end of 72 hours under agitation. Effective decolorization was obtained in 1 mg concentration, which showed decolourization up to 82%. The 9 mg/100 ml which is the highest concentration among the variants used was decolourized up to 71%. This is considered to be the lowest decolourized rate in concerned with *Fusarium* (Fig. 1)

Decolorization of malachite green by *Aspergillus* under agitation

Even though *Aspergillus* decolourized malachite green, the result was not up to the effect of Fusarium. The percentage of decolorization ranges from 22–65.4% only. The least decolorization effect obtained was 22% at 9 mg concentration. The first concentration that contains 1 mg concentration was decolorized up to 58% after 72 hours of incubation under agitation. Wafaa *et al.*, (2003) showed that, *Aspergillus* sp. strains were found to be successful in decolorizing textile dyes from liquid medium. (Fig. 1)

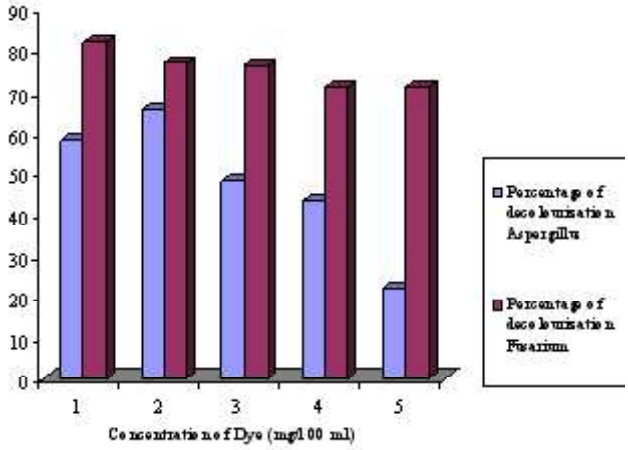

Fig. 1. Decolourization percentage of malachite green under agitation using *Aspergillus* and *Fusarium*

CONCLUSION

This study showed that malachite green dye was decolorized by both *Aspergillus* and *Fusarium*. The effective decolourization was found with Fusarium than Aspergillus in all the five concentrations. In most of the decolorization process, 1 mg concentration was decolorized effectively than the other four concentrations. As the dye concentration increases, the decolourization effect decreases. The results revealed that 5 ml of the culture filtrate could effectively decolorize 1 mg/100 ml of dye concentration. But the other concentrations require more amount of culture for effective decolorization. This may be due to the structure complexity of the malachite green dye. It was viewed obviously that 1 mg of malachite green dye gave dark green color in dilution.

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