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Decolourization of the Azo dye (Direct Brilliant Blue) by the Isolated Bacterial Strain

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Abstract—*The present study was carried out to investigate the decolourization efficiency of the bacterial strain isolated from the textile effluent. The decolourization efficiency is largely influenced by the initial dye concentration, pH, incubation time, temperature and nutrient availability. The bacterial strain showed higher decolourization capacity (93%) for the selected azo dye (direct brilliant blue- 50mg/l initial dye concentration) within 18 hours of incubation time at 30º C and pH 7 using composite media. Dye decolourization efficiency increased with increase in the peptone concentration as the sole nutrient source at 50mg/l initial dye concentration. Up to 89% removal was obtained with 2% peptone concentration. This bacterial strain may provide an alternate and economical option for the azo dye decolourization.*

Keywords*: Azo Dye, Direct Brilliant Blue, Bacteria, decolourization.*

1. INTRODUCTION

Waste water discharges from the textile industries are complex solutions which are highly colored and toxic. The textile industries produce effluents that contain several types of chemicals such as dispersants, leveling agents, acids, alkalis, carriers and various dyes. Dyes lost in the effluent during the dying process is very high in concentration (0.6-0.8 g L^{-1} dye)[1] and toxic to the environment. Alterations in the chemical structures of dyes can result in the formation of new xenobiotic compounds which may be more or less toxic than the parental compounds[2]. Azo dyes are the aromatic compound with one or more –N=N– groups and one of the largest synthetic dye used by textile industries. The solution to the environmental problems caused by the textile dyes is being sought by physical, chemical and biological treatment processes. Several physical and chemical techniques have been developed, including physico-chemical flocculation, flotation, electro-flotation, membrane filtration, ion exchange, irradiation, precipitation etc.[3]–[8].Although some of these techniques did prove effective, the disadvantages outweighed the advantages. Secondary waste generated while using these techniques presented many disposal problems, high operating expenses coupled with inefficient removal of azo dyes. Biological methods, which are comparatively inexpensive and easy to use, are being investigated as methods for degrading and decolorizing synthetic dyes[9], [10]. Biological treatment has been shown to be a potential solution to a complete treatment system, with the mineralization of organic dye compounds to inorganic constituents which are non-toxic in nature[11]–[13].

The present study is carried out to decolorize the azo dye using isolated bacteria from the textile effluents. The efficiency of the microbial consortia to degrade azo dye was maximized by optimizing the various parameters such as initial dye concentration, pH, temperature and time. Peptone was tested as the sole nutrient source for bacteria and its decolourization efficacy was observed with different concentration of peptone.

2. MATERIAL AND METHODS:

2.1 Chemicals

Textile dye direct brilliant blue used in this study was obtained from textile industry of sonipat, Haryana. All chemicals used were of analytical grade and were purchased from Hi media, Sigma, SRL, etc.

2.2 Culture Conditions and Biodegradation Assay

One of the bacterial strain (1DD) uses in this study was isolated from textile effluents. The culture was maintained at 4 °C on nutrient agar media (Himedia).The medium for bacterial growth and dye removal study was composed of (in gram per liter): Sucrose (2) K_2HPO_4 (1), $MgSO_4$(0.5), $CaCl_2$(2),

$NaNO_3$(1.5) and NaCl(0.5). Culture was grown at 30 ± 0.2 ºC to inoculate the flask for all the dye decolourization assays. The decolourization assays were performed into 250 ml Erlenmeyer flask containing 100ml composite media. At defined time interval the sample was withdrawn from the flask and supernatant was obtained by centrifugation at 10000 rpm for 10 min. The resulted supernatant was analyzed for dye concentration at absorbance maxima of 615nm using UV-Vis spectrophotometer. All the decolourization assays were performed in triplicates and results was expressed in terms of percentage decolourization using the equation given below:

$$\text{Decolourization}(\%) = \frac{\text{Initial Absorbance} - \text{Absorbance after incubation}}{\text{Initial absorbance}} \times 100$$

2.3 Effect of process parameters

The effect of various process parameters such as initial dye concentration, initial pH, incubation and temperature was studied using the selected isolated bacteria 1DD. The effect of initial direct brilliant blue concentration was studied by inoculating the micro-organism in the sterilized flask containing the composite media and different dye concentration (50, 100, 150 and 200mg/l). Similarly, the effect of initial pH was studied by fixing the pH value of composite media ranging from 5-9. To study the effect of incubation temperature dye removal efficiency was monitored at different temperature (25-45 ºC).

2.4 Effect of nutritional conditions

The effect of nutritional conditions was studied by replacing the composite media with peptone as a sole nutrient source. Dye removal efficiency by micro-organism was investigated at different peptone concentration ranging from 0.5 to 2gm/l.

Dye may be used as the carbon or nitrogen source by the micro-organism. Considering the above fact, the nitrogen and the carbon source were replaced one by one by the dye and decolourization studied was carried out.

3. RESULTS AND DISCUSSION

3.1 Isolation and screening of micro-organism

The bacterial strain was selected on the basis of its ability to form the clear zone on the nutrient agar media containing direct brilliant dye (Fig. 1). By the formation of the clear zones it was conferred that the isolates were able to decolourize the agar media or they were able to degrade the dye in the agar medium.

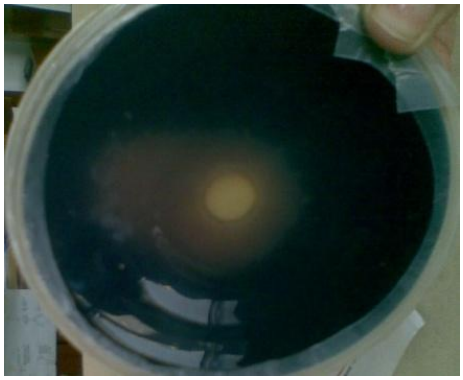
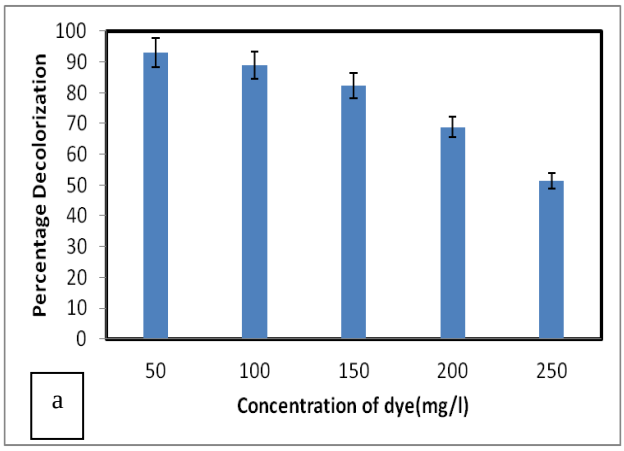
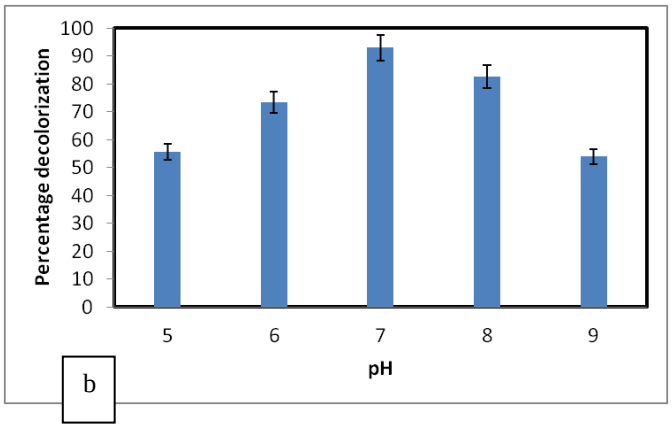


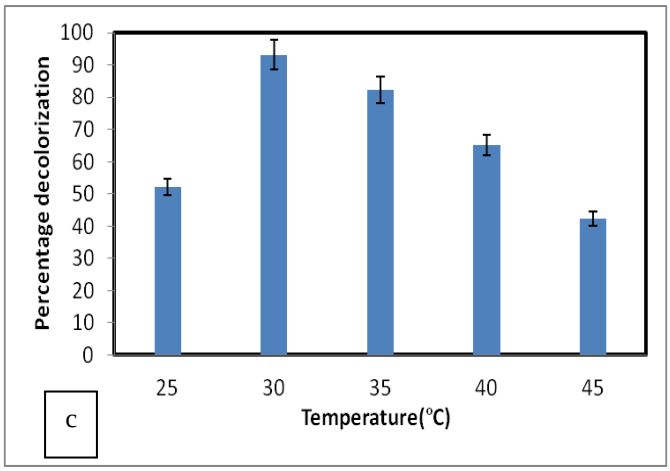
Fig. 1: Formation of clear zone in nutrient agar media by bacterial isolate at 50mg/l initial dye concentration.

3.2 Effect of process parameter on azo dye removal:

Fig. 2(a) indicates the dye colorization by selected bacterial strain 1DD at different initial direct brilliant blue concentrations within 18 hours. Decolourization up to 93% was observed with 50mg/l initial dye concentration .It was observed that with the increase in the initial dye concentration percentage decolourization was decreasing. Similar results were reported to decrease in the dye decolourization with the increase in concentration[10]-[13]. Fig. 2(b) shows the decolourization at different pH with 50 mg/l initial concentration. Maximum decolourization was found to be at pH 7. The pH of the media is important for the decolourization as it is responsible to carry the dye molecule across the cell membrane of the micro-organism and considered as the rate limiting factor in the decolourization process[12]. Fig. 2(c) represents the optimum temperature for decolourization of the azo dye with initial concentration of 50mg/l and it was observed that 30ºC is the optimum temperature for maximum decolourization. Fig. 2(d) indicates that maximum decolourization of the dye was found at 18 hours of incubation with initial concentration of 50mg/l. Increase in the incubation time does not increase the decolourization efficiency of the bacterial strain. Similar results have been reported with the bacterial strains with different azo dye[12], [14]–[16].







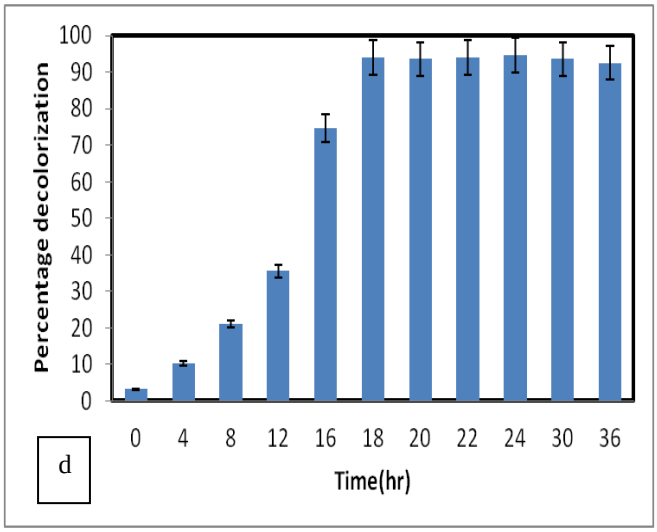


Fig. 2: Effect of process parameters; effect of initial concentration (a), pH (b), incubation temperature (c) and time (d) on decolourization of direct blue by bacterial strain.

3.3 Effect of nutritional conditions

Growth as well as removal efficiency of the particular pollutant by the micro-organism is very much affected by the nutritional conditions. Hence, it is important to investigate the influence of nutrient and optimize its quantity for maximum decolourization. Effect of different concentration of peptone as a sole nutrient source is shown in Fig. 3. Results show that dye decolourization efficiency increases with the increase in the concentration of the peptone. However, significant removal was not observed after 1.5% peptone concentration in the flask. A similar increase in decolourization efficiency of reactive brilliant blue with increase in concentration of peptone have been reported with *Rhodocyclus gelatinosus*[17]. It was also noted that no decolourization was obtained when dye was replaced as the carbon and nitrogen source in the composite media.

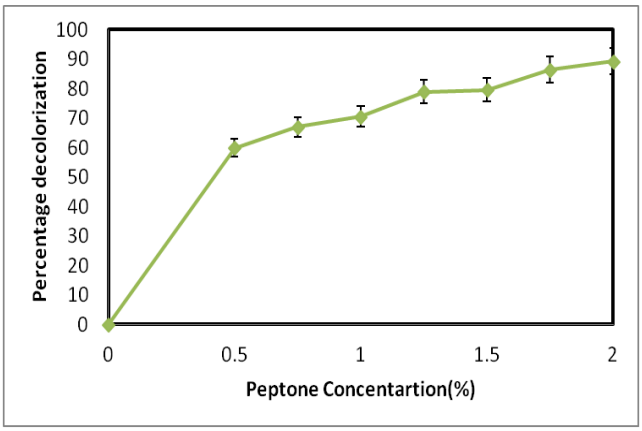


Fig. 3: Effect of different concentration of peptone as the sole nutrient source on decolorization of direct blue by the bacterial strain. (Initial dye concentration: 50mg/l; temperature: 30ºC; pH: 7)

4. CONCLUSION

Results obtained in this study suggest that isolated bacterial strain may provide the promising alternative for azo dye decolourization. However, the decolourization efficacy of bacteria largely depends on the initial dye concentration, pH, temperature and time. The optimal decolourization activity for 50mg/l dye concentration was observed with pH 7 and temperature 30ºC within 18 hours. Composite media shows better decolurization than peptone which was investigated as an alternate nutrition source.

5. ACKNOWLEDGEMENT:

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