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Original Research Paper

Decolorization and Degradation of Textile Azo Dye Golden Yellow HE2R by Adapted Bacteria

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ABSTRACT

Degradation and decolorization of azo dye Golden Yellow HE2R was carried out by soil bacterial isolates. Soil samples from various places were collected and acclimatized for decolorization of azo dye Golden Yellow HE2R. The acclimatized culture GY-B could decolorize 90% of the dye with 10,000 mg/L concentration. Eight promising isolates were able to decolorize the dye solution more than 83.43% averagely in less than 24 hours. Percent decolorization was studied by spectrophotometry. Hyper-interactions between carbon source like 1% glucose, 1% starch and 1% yeast extract and bacteria to decolorize the dye was investigated and modelled. In percent COD reduction studies, it was observed that more than 75% COD was reduced after decolorization process. The decolorization of the dye in half strength nutrient broth was also studied. The products formed after the degradation process were analysed by GC-MS technique.

INTRODUCTION

Largest and the most widely used important dyes, which are applied in textile industry, are azo dyes. Azo dyes are largest group of man-made chemicals used in textile, paper, printing, pharmaceuticals, leather, food industries, etc. These dyes are usually recalcitrant to conventional wastewater treatment (Kodam et al. 2004) and also mutagenic and carcinogenic which require proper degradation and safer disposal (Vyas et al. 1995). Number of physicochemical methods such as adsorption, coagulation, precipitation, filtration and oxidation are used for the treatment of dye stuff effluent, but these methods have many limitations like cost effectiveness, residue (sludge) formation, etc. and, hence it is important to develop a cost effective and efficient method for decolorization and degradation of dyes. Biotransformation of azo dyes can be used for their degradation and decolorization. It includes the use of microorganisms, especially bacteria. This treatment is cheaper, effective and environment friendly. The degradation generally involves two steps; first, decolorization of the azo dye by its reduction and second, degradation of the azo dye metabolites or aromatic compounds (Pandey et al. 2007).

The study deals with decolorization and degradation of an azo dye Golden Yellow HE2R by using acclimatized cultures which were isolated from soil. The ability of isolates to degrade the dye was observed in various conditions like in nutrient broth, half strength nutrient broth, in presence of co-substrates like 1% glucose, 1% starch and 1% yeast extract. The decolorization observed was determined by spectrophotometer. Percent COD reduction after the decolorization was determined by COD analysis. Also effect of cell-free extract on decolorization and degradation of the dye was studied. The degradation of the dye was determined by GC-MS analysis.

MATERIALS AND METHODS

Acclimatization of bacteria: Soil samples from nearby area of waste disposal site of textile industries, sewage, sludge, ETP along with compost were collected and homogenized properly. The microflora from the samples were acclimatized in the dye Golden yellow HE2R (1%) for the period of one month. One gram of acclimatized soil was inoculated in the nutrient broth, after incubation isolation was carried-out on nutrient agar incorporated with the same dye concentration. The colonies showing decolorization were selected for the further studies

Decolorization of dye in nutrient broth: The selected microorganisms were inoculated in 10 mL of nutrient broth containing 1 mL dye solution. These tubes were then incubated at ambient temperature for 24 hrs and observed for decolorization of the dye.

Decolorization of dye in half strength nutrient broth: Isolated cultures were then inoculated in 10 mL of half strength nutrient broth containing 1 mL of dye solution. These tubes were incubated at ambient temperature for 24 hrs and decolorization pattern was studied.

Effect of co-substrate on decolorization of dye: Co-substrates like 1% glucose, 1% starch and 1% yeast extract were used. These substrates were added to the nutrient broth (1% w/v) containing 1 mL of dye solution. The decolorization was observed after 24 hrs of incubation.

Cell-free extract studies on decolorization of dye: The cells grown in nutrient broth were lysed by using lysozyme and centrifuged in cooling centrifuge (Remi 412-LAG) at 7000 rpm for 10 min. The 10 mL supernatant was added with 1 mL of dye solution in nutrient broth and incubated at ambient temperature.

Table 1: % decolorization of golden yellow HE2R in nutrient broth after 24 hours at 412 nm (λ max).

Sr. No.	Culture code	% Decolorization
1.	GY-B	90.20
2.	GY-C	88.20
3.	GY-E	80.10
4.	GY-F	80.50
5.	GY-G	78.02
6.	GY-J	77.37
7.	GY-K	82.13
8.	GY-L	91.16

The percent decolorization studies were carried out by using spectrophotometer (Systronics-106 model). Percent COD reduction values were calculated by COD analysis. Degradation of the dye was confirmed by GC-MS analysis after extraction of the degraded products by di-chloro methane (DCM).

RESULTS AND DISCUSSION

The cultures showing decolorization of the dye Golden Yellow HE2R on agar medium were selected as the promising isolates and used for further study. In all, total 8 promising isolates *viz.*, GY-B, GY-C, GY-E, GY-F, GY-G, GY-J, GY-K and GY-L, were isolated on nutrient agar with dye decolorizing ability. These isolates were studied for their gram nature, motility and biochemical characters. Out of all bacterial isolates, GY-B showed more than 90% decolorization. The results are given in Table 1. The percent decolorization was determined by spectrophotometer (Systronics-106 model). The use of half strength nutrient broth showed no significant change in percent decolorization when compared with normal nutrient broth. The results are given

Table 2: % decolorization of golden yellow HE2R in half strength nutrient broth after 24 hours at 412 nm (λ max).

Sr. No.	Culture code	% Decolorization
1.	GY- B	90.00
2.	GY- C	87.15
3.	GY- E	80.00
4.	GY- F	80.12
5.	GY- G	78.00
6.	GY- J	77.03
7.	GY- K	81.94
8.	GY- L	90.05

Table 3: % decolourization of golden yellow HE2R in nutrient broth + (co- substrates) 1% glucose, starch and yeast extract after 18 hours at 412 nm (λ max).

Sr. no.	Culture Code	% Decolorization at 412 nm.		
		1% Glucose	1% Starch	1% Yeast Extract
1.	GY-B	90.24	90.20	90.85
2.	GY-C	88.20	88.20	88.03
3.	GY-E	80.19	80.10	80.36
4.	GY-F	80.52	80.50	80.00
5.	GY-G	78.15	78.02	78.43
6.	GY-J	77.45	77.37	78.00
7.	GY-K	82.50	82.13	82.19
8.	GY-L	91.36	91.16	91.67

in Table 2.

The effect of co-substrate on percent decolorization of the dye by these 8 isolates were studied and showed no significant change, but the time required for the decolorization of the dye was decreased by 6 hours. The results are given in Table 3.

The percent decolorization of the dye Golden Yellow HE2R was studied by cell-free extract of the two bacterial isolates GY-B and GY-F, which showed the decolorization of dye same as in nutrient broth. The isolates GY-B and GY-F showed percent decolorization of 90.00% and 80.12% respectively. The results are given in Table 4.

Table 4: % decolorization obtained by cell free extract at 412 nm (λ max)

Sr. No.	Culture code	λ max	% Decolorization
1.	GY-B	412 nm	90.00
2.	GY-F	412 nm	80.12

Table 5: % COD reduction of golden yellow HE2R after 24 hours at 412 nm (λ max).

Sr. No.	Culture code	% COD Reduction
1.	GY-B	75.37
2.	GY-C	69.18
3.	GY-E	66.10
4.	GY-F	66.83
5.	GY-G	51.08
6.	GY-J	51.95
7.	GY-K	66.75

The COD reduction values in nutrient broth were calculated and it was observed that the GY-B strain was able to reduce the COD by more than 75%. The results are given in Table 5. The promising isolate GY-B was then selected for the degradation studies. The GC-MS reports (Fig. 1) showed that the azo dye Golden Yellow HE2R was degraded in its smaller products having molecular weights 99 d, 154 d, 194 d and 244 d.

The isolate GY-B showed good decolorization ability. The degradation of the dye was confirmed by GC-MS analysis (Kodam 2005). The study of co-substrate showed no significant increase in the percent decolorization, but in turn decreased the time required for decolorization by 6 hours. Kothari (2005) observed same results with co-substrates in dye degradation capacity. The results obtained were similar with results given by Chinwetkitvanich (2000), but they used *Tapioca* as substrate. McMullan (2005) showed that microorganisms are able to degrade the textile dyes.

CONCLUSION

The acclimatized cultures showed on an average 97.00% decolorization of the dye Golden Yellow HE2R. After addition of the co-substrate, not much change was observed, however, the time required for the decolorization of the dye was decreased. From the percent COD reduction values, it can be

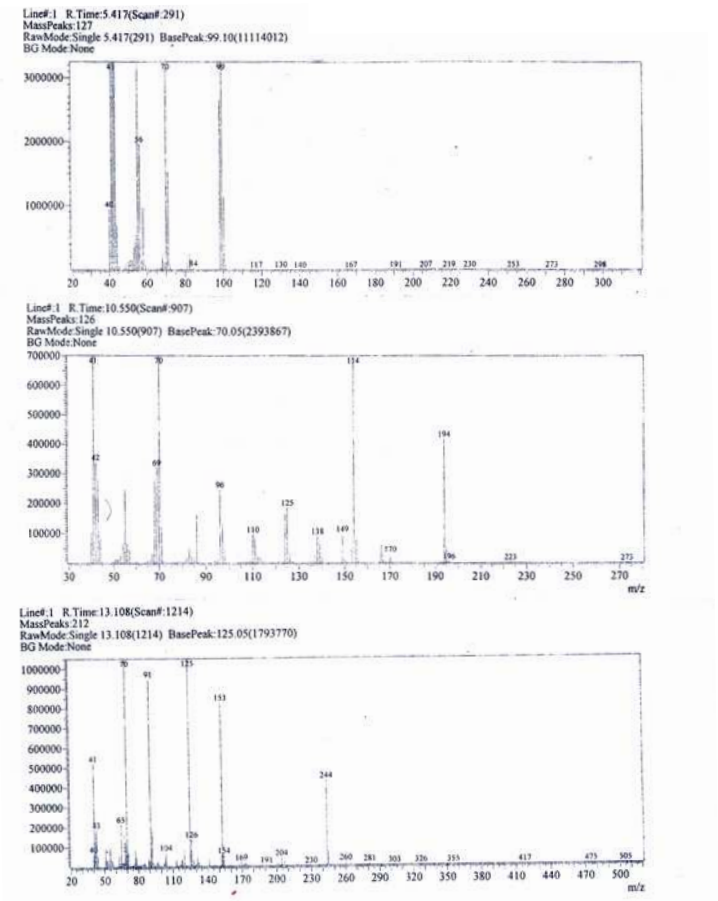

Fig. 1: GCMS analysis report of the degraded product by GY-B.

concluded that dyes were degraded by the isolates. Cell free extract studies showed good decolorization by the promising isolates like GY-B and GY-F within 24 hours. The promising isolate GY-B degraded the dye and products were analysed by GC-MS.

REFERENCES

Chinwetkitvanich, S., Tuntoolvest, M. and Panswad, T. 2000. Anaerobic decolorization of reactive dye bath effluents by a 2 stage UASB system with *Tapioca* as co-substrate. Water Research, 34: 2223-2232.

Kodam, K.M., Soojhawon, I., Lokhande and Gawai, K.R. 2005. Microbial degradation of reactive azo dye under aerobic conditions. World Journal of Microbiology and Biotechnology, 21: 367-370.

Kothari, R.K. 2005. Degradation Biotechnology. 1st Ed., Discovery Publication House.

McMullan, G.C., Meehan, C., Conneely, A., Kirby, N., Robinson, T., Nigam, P., Banat, I.M., Marchant, R. and Smyth, W.F. 2000. Microbial decolorization and degradation of textile dyes. J. Appl. Microbiol. Biotechno. School of Environmental Studies, University of Ulster, Coleraine, London. 56(1-2): 81-87.

Pandey, A., Singh, P. and Iyengar, L. 2007. Bacterial decolorization and degradation of azo dyes. Int. Biodeter. Biodegr., 59: 73-84.

Vyas, B.R.M. and Molitoris, H.P. 1995. Involvement of an extracellular H_2O_2 dependent lignolytic activity of white rot fungus *Pleurotus ostreatus* in decolorization of Remazol Brillant Blue R. Appl. Environ. Microbiol., 61(11): 3919- 3927.