

Biodegradation of Textile Dyes Using Enterococcus Species

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ABSTRACT

Introduction: Environmental pollution is one of the major problem that has affected our lives. One of the main contributing factor to environmental pollution is the use of dyes in textile industries. These industries release a huge volume of dyes in their effluents (Sharma 845-850). The terrestrial and aquatic fauna are greatly affected by the release of these environmental pollutants. The humans when exposed either directly or indirectly to these dyes face serious health problems. The use of eco-friendly methods is major area of interest. The bacteria play important role in degradation of dyes as they possess different enzymatic pathways that convert the complexity of dyes in its simple compounds (Abd El-Rahim269-276).

Objective: To identify the capability of *Enterococcus faecalis* to degrade textile dyes.

Methodology: The biodegradation study was carried out by *Enterococcus faecalis*. The pure culture of *Enterococcus faecalis* was obtained from culture bank of Microbiology Department, Jinnah University for Women. The culture was purified on bile esculin agar and then confirmed by biochemical methods. The molecular characterization was performed using 16S rRNA gene (Santal41-55). The two disperse textile dyes, dark blue HP-LTD and black HP-LTD were collected from a textile industry located in Karachi, Pakistan. The 5% stock solution of both dyes were prepared in distilled water. The effect of biodegradation of textile dyes at different concentrations ranging from 41µg/mL to 650µg/mL was performed. The bacteria was grown in nutrient broth and standardized with 0.5 x10⁵ cells of McFarland Index. The equal volume of nutrient broth, disperse dye and culture was added in each tube (Safitri 183-192). The tubes were incubated in incubator at 37 °C for 24 to 48 hrs. The percentage of dye decolourization was noted with the given formula:

$$\text{Decolourization (\%)} = \frac{\text{initial absorbance} - \text{final absorbance}}{\text{initial absorbance}} \times 100$$

The dye degraded products were centrifuged at 10,000 rpm for 10 minutes. The supernatant obtained was filtered through 0.45 µm What man filter paper. The optical density of dye after degradation was measured by UV –Vis spectrophotometer. The λ_{max} of each dye was measured by UV –Vis spectrophotometer in a range of 200 to 800 nm wavelength and taken as control.

For extraction of metabolites after dye degradation, the equal amount of ethyl acetate was added in supernatant and kept at 40°C for evaporation. Then metabolites in test and control were confirmed using thin layer chromatography.

Result: The bacterial species that we isolated and purified using bile esculin agar were characterized as gram positive cocci. The colonies of *Enterococcus faecalis* on bile esculin were of white to greyish color with black coloration in the media and confirmation was done through carbohydrate fermentation. The DNA was extracted from obtained pure culture for molecular characterization of 16S rRNA gene. The DNA fragments were separated on the basis of their molecular size on 1% agarose gel. The band size 1500bp was observed which was matched with 1 kb ladder.

By UV-Visible spectrophotometer analysis, we observed λ_{max} 610 nm of dark blue HP-LTD and λ_{max} 560 nm of black HP-LTD dye. *Enterococcus faecalis* decolorized both dyes at 37°C in 24 hours. The percentage of degradation of dark blue HP-LTD dye was found as 89% and 90% for dye black HP-LTD. We observed that organism has ability to degrade dye in all concentrations from 41µg/mL to 650µg/mL. The metabolites

were extracted when the degraded dye sample was placed along with equal volume of ethyl acetate at 40°C. Thin layer chromatography of extracted metabolites was performed and observed under ultraviolet light. On alumina plates, blue and black dye showed multiple spots while in test sample after biodegradation the spots were not found. This showed that the dye has been completely degraded.

Conclusion: Biodegradation of textile effluent is an important approach in the bioremediation that can be used on a large scale in textile industries. The utilization of *Enterococcus* species for the biodegradation of dyes is also a promising approach in the bioremediation.

Keywords: Biodegradation, Disperse Dyes, Black HP-LTD, Dark Blue HP-LTD, Enterococcus Species.
