RESEARCH PAPER

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Biodegradation of some aromatic hydrocarbons (Toluene and Xylene) by a bacterial strain isolated from petroleum contaminated site in Chennai

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Pseudomonas putida strain was isolated from petroleum polluted soil in Chennai by enrichment culture method to evaluate the biodegradation potential of toluene and xylene. The isolated bacterial strain in this study could grow in Bushnell Haas medium and mineral salt medium containing 1% each of toluene and xylene as the sole source of carbon and energy. The plasmid DNA was isolated by alkali lysis method and separated by agarose gel electrophoresis. The molecular weight of the plasmid was determined as 106Kb. Liquid culture studies were performed to determine the effect of pH and temperature on TX biodegradation. The isolated strain showed maximum degradation at 30^0 C and at pH 7.0. The kinetics of TX biodegradation was investigated at different time intervals (2.5 Hrs. - 12.5 Hrs.) and the biodegradation rate was maximum between 5 Hrs. and 12.5 Hrs. The conclusions that can be drawn from this study indicate that the microorganism present in this study could contribute significantly to bioremediation of TX pollution.

Key words : Biodegradation, Bioremediation, Toluene, Xylene, *Pseudomonas putida*

INTRODUCTION

Toluene is widely used as an organic solvent and starting material for the manufacture of plastics, pesticides and synthetic fibers. Its worldwide production was estimated to be more than 80,000 metric tons per year (Anonymons, 1985). Toluene and other organic solvents such as xylene, benzene and ethyl benzene are ubiquitous pollutants (Gibson and Subramanian, 1984) several environmental protection agencies have declared the removal of these pollutants to be high priority.

Compared to physicochemical methods, bioremediation offers an effective technology for the treatment of oil pollution, because majority of molecules in the crude oil and refined products are biodegradable and oil degrading microorganisms are ubiquitous (Chaineau *et al.,* 2000). Biological treatments for the removal of these organic compounds from contaminated water, soil and reactors are based on the action of degrading microbial communities, so detailed knowledge of the microbiology of petroleum hydrocarbons must be clear and understood to evaluate both the biodegradability of the most petroleum hydrocarbon compounds and the specific degradative activities of the different micro florae (Solano – Serena *et al*., 1999; Cavalca *et al*., 2000).

Contamination of ground water with toluene and xylene (TX) compounds is difficult to remedy because these compounds are relatively soluble in water and diffuse rapidly once introduced into an aquifer. Techniques for *in situ* bioremediation of the TX compounds are used to eliminate or reduce contamination levels in an aquifer. The biodegradability of the TX has been established using some pure bacterial strains or complex micro florae (Mallakin and Ward, 1996, Matteau and Ramsay, 1997); but little is known about the other microbial strains capacity, especially those which are wide-spreading in environment. This work describes the capability of a degrading microbial strain isolated from a polluted site in Chennai.

MATERIALS AND METHODS

Chemicals:

Toluene (99.5% purity), xylene (99% purity) were purchased from Merck Limited, Worli, Mumbai. All other chemicals used were of analytical grade.

Isolation of organism:

Soil samples were collected from petroleum polluted site at Manali, Chennai and were used to isolate

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