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Isolation of soil bacteria for bioremediation of hydrocarbon contaminated sites in Sri Lanka

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Soil contamination with hydrocarbons causes extensive damage to local ecosystems. In Sri Lanka, numerous contaminated sites exist as a result of more than 60 years of oil/ petroleum activity. As opposed to chemical agents available to clean up hydrocarbon contaminations, bioremediation provides an environmentally friendly, efficient and cost-effective solution. Although several countries have already used methods including microorganisms for bioremediation of petroleum spills, it has not been previously used in Sri Lanka. The goal of this study was to isolate indigenous bacterial strains from hydrocarbon-contaminated soils and to assess their potential for bioremediation.

Microorganisms that are capable of degrading hydrocarbons were isolated from soils which have been exposed to different petroleum products over a long period of time. The bacteria were cultured with diesel/ engine oil as the sole carbon and energy source in Bushnell Hass (BH) agar. Genomic DNA was extracted from the isolated bacteria and then amplified by polymerase chain reaction (PCR) using 16S rRNA specific primers. The PCR products were sequenced and analyzed. A search of the non-redundant nucleic acid database at National Center for Biotechnology Information (NCBI) with the sequences of the PCR products led to the identification of four bacterial species. They were *Paenibacillus lautus*, *Stenotrophomonas* sp., *Tistrella mobilis*, and *Pseudacidovorax* sp A14. They were then evaluated for their potential to degrade hydrocarbons in artificially contaminated soil/ water media under laboratory conditions by several methods. These methods include, assessing the time taken to show turbidity due to disruption of the film of oil on the surface of the water/ BH media, the increase of water solubility of hydrocarbons due to the degradation by bacteria and by visual observations of the reduction of oil in soil as compared to controls. Their ability to bioremediate the hydrocarbons were also assessed quantitatively using gas chromatography (GC) equipped with a flame ionization detector (FID).

A cocktail made by inoculating all four groups of bacteria showed significant potential in removing/ detoxifying hydrocarbon contamination in soil/ water. GC/ FID analysis revealed the degradation of high molecular weight toxic hydrocarbon compounds in engine oil to low molecular weight less toxic compounds. Work is currently underway to develop a bioproduct useful for soil/ water inoculation to clean up hydrocarbon contaminated sites in Sri Lanka.