(105)

Pyrene Degradation Ability of Phyllosphere *Bacillus* Species Bacteria Inhabiting the Urban Areas in Sri Lanka

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Abstract

Polyaromatic Hydrocarbons (PAHs) are a diverse group of ubiquitous hydrophobic organic contaminants which have a wide range of diversity around seventeen polycyclic aromatic hydrocarbons and these compounds are released from the incomplete combustion of organic matter, emissions from automobile exhausts, stationary matter and domestic matter, area source matter such as forest fires and agricultural burning and also in food. Out of many polyaromatic hydrocarbons, pyrene is concerned as a major environment pollutant. Pyrene is composed with three fused benzene rings. Remediation of pyrene from the environment is essential because of their toxic affect to the health. Out of many remediation methods bioremediation is the most eco-friendly and effective method which can be used to convert toxic substances to nontoxic substances. There are many phyllosphere microorganisms which have the capability in pyrene like polyaromatic air pollutant degradation. Therefore this attempt is to isolate, select and identify the efficient pyrene degrading bacteria. Bacteria were isolated using leaf samples of *Ixora chinensis*, *Ervatamia divaricate*, *Plumeria* sp. *Hibiscus rosa-sinensis* and *Amaranthus cruentus* collected from Panchikawatta, Orugodawatta, Pettah, Maradana, Colombo Fort and Sapugaskanda oil refineries like polluted areas in Sri Lanka. Pyrene degradation ability of isolated phyllosphere bacteria was primarily screened using plate assay method. Subsequently, pyrene degradation ability of each bacterial species was further analysed using the UV-Vis spectrophotometer and High-performance liquid chromatography (HPLC).

The selected bacterial isolates were identified up to species level through PCR amplification of a fragment of 16S rRNA gene and sequencing the amplified fragments using the primers 1492R (GGTTACCTTGTTACGACTT) and 27F (AGAGTTTGATCMTGGCTCAG). Four *Bacillus* species *Bacillus* sp. P2B-02, *Bacillus velezensis*, *Bacillus* sp.1 and *Bacillus megaterium* were able to degrade more than 90% of pyrene. Out of these bacteria, *Bacillus* P2B-02 (Accession no: MN190154) and *Bacillus megaterium* (Accession no: MN190174.1) were the most efficient bacterial species which was highly capable in degradation of pyrene with 100% of degradation. The results of the present study clearly clarify the potential use of the phyllosphere *Bacillus* species in remediating environment contaminants such as pyrene. The *Bacillus* species could be beneficial as potential biological agent in bioremediation for polluted environments with pyrene like polyaromatic hydrocarbons.

Keywords: Phyllosphere, Aromatic hydrocarbon, Bioremediation, Pyrene, *Bacillus* species