

**Editorial**

## Molecular Tools in Petroleum Hydrocarbon Degradation: An Overview

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Kerosene, diesel, gasoline and higher alkanes such as octadecane (C<sub>18</sub>), eicosane (C<sub>20</sub>) & docosane (C<sub>22</sub>) are petroleum hydrocarbons. In the environment these hydrocarbons enter through waste disposal, accidental spills, leakage tanks, & losses during transportation [1]. These types of hydrocarbons are hazardous to the plants, animals & human. Especially, higher hydrocarbons are more carcinogenic, mutagenic & potent immune-toxicants. The contamination of these hydrocarbons in the environment creates imbalance in the ecosystem [1,2].

Microorganisms can utilize petroleum hydrocarbon as a sole source of carbon & energy. Biodegradation is the most effective technique to remove the pollutant such as total petroleum hydrocarbon from environment specifically when bacteria are involved. Natural bacteria present in the contaminated site have the potent ability to degrade such hydrocarbons [3,4]. Some usually occurring bacteria in the polluted site are *Rhodococcus*, *Pseudomonas*, *Acinetobacter*, *Burkholderia*, *Gordonia*, *Enterobacteria*, *Novosphingobium*, *sphingobium*, *sphingomonas*, *Bacillus*, and *Psychrobacillus* [2,5].

Every indigenous microorganisms isolated from oil-contaminated soil has certain ability to utilize petroleum hydrocarbons. To evaluate their ability to degrade target pollutants is the most important steps in bioremediation technique. Similarly, time estimation of complete degradation of environmental pollutant from natural sites has another critical role in designing strategy for biodegradation. Monitoring & prediction of biodegradation also has important aspect in the field of cleaning environment from petroleum hydrocarbons [6,7].

### Application of Molecular Techniques

Molecular techniques have revolutionized microbial ecology by developing culture-independent assessment and exploitation of microbial communities present in complex ecosystems like crude-oil/hydrocarbons polluted soil. The combination of PCR-amplification of Metagenomic DNA, microbial community profiling techniques & identification of catabolic genes are ways to elucidate the composition, function & interaction of microbial communities during bioremediation. There are several reports on molecular tools which are used in bioremediation techniques [8,9,12]. PCR-based methods are used in the detection and enumeration of microorganism found in soil and water. This technique is also used to identify catabolic genes involved in biodegradation of hydrocarbons. Amplified ribosomal DNA restriction analysis

(ARDRA), Ribosomal inter genic spacer analysis (RIA), Terminal-restriction fragment length polymorphism (T-RFLP), Fluorescent in-situ hybridization (FISH), DNA microarray technologies, and meta genomic tools are important molecular techniques which have been widely used in the field of microbial degradation of petroleum hydrocarbons [8,12].

Cytochrome p450 alkane hydroxylase, di-iron methane monooxygenase, di-iron alkane hydrolases are important group of enzymes that play essential roles in the biotransformation of many environmental pollutants. The detection of these genes encoding these enzymes is important biomarker that indicates metabolic activity in the biological system. Biomarker provides valuable information to assess whether changes in contaminants concentration are occurring through physical, chemical or biological processes [9]. Similarly, catabolic Genes are promising molecular markers for assessing completion of biodegradation. Identification of monooxygenase gene during the growth of bacteria in hydrocarbon containing medium and oil-contaminated soil serves as the rapid tool for designing biodegradation program [10]. On the other hand, applications of genetically engineered microorganisms (GEMs) in bioremediation have received a great deal of attention to improve the degradation of hazardous wastes under lab conditions [11, 12].

Generally, oil-contaminated soil samples can be processed simultaneously for isolation of bacterial strains and for extraction of metagenomic DNA from soil. The isolated pure colonies of bacteria should be assessed for oil degradation capacity. The extracted soil DNA should be analyzed for detection of catabolic genes. This gives the basic idea to utilize cultural-dependent tools and molecular tools in the field of microbial degradation of hydrocarbons (Figure 1).

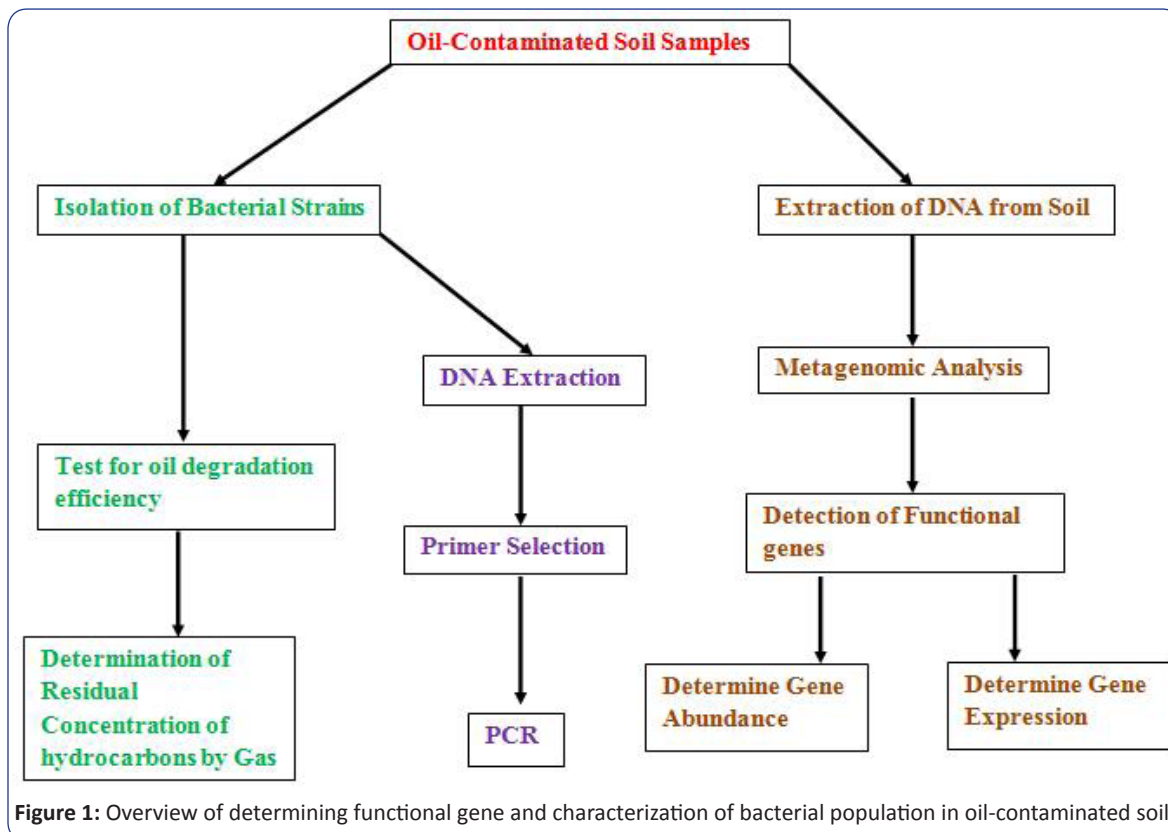
In conclusion, the use of genetically engineered bacteria has been

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applied to bioremediation process monitoring, strain monitoring, stress response, end-point analysis & toxicity assessment. Several species of *Pseudomonas*, *Bacillus* and *Acinetobacter* can be genetically engineered for assessing biodegradation. The precise characterization of bacterial diversity in polluted soil using molecular tools will be fruitful in future application of hydrocarbon degrading bacteria in contaminated sites.

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