A Preliminary Study on Oil Biodegradation Using Microbes Isolated From Various Ecological Sites

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Abstract – In and around Navi Mumbai, hydrocarbon-degrading microorganisms were isolated from ten oil-polluted regions. As a specific enrichment medium with engine oil as a single source of carbon and electricity, Bushnell and Hass broth was used and 19 distinct isolates were collected. Using the Emulsification Index and Gravimetric study, the oil degradation ability of these isolates was evaluated. The GC-MS analysis demonstrated that the conversion of Heneicosane to hexadecane suggests that the organism partly degrades the oil. Biochemical analyses and 16s RNA sequencing verified that the isolate is Bacillus cereus.

Keywords: Degradation of Fuel Oil, Bacillus Cereus.

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INTRODUCTION

Large oil accidents draw the public and the media's interest. This focus has generated a worldwide consciousness in recent years of the dangers of oil pollution and the environmental harm they are causing. In our modern world, though, oil is a must and a significant supporter of our lifestyle. Any, of the energy used in Canada and the United States is for transportation that operates on oil and petroleum products. India is ranked fourth worldwide for oil consumption. This is not expected to decline far in the future, according to patterns in energy consumption. Oil and petroleum derivatives are used by industry to generate such essential goods as chemicals, fertilizers and chemical feedstock's that would also be required in the future? In reality, the worldwide output and usage of oil and petroleum products is growing and the level of oil emissions is correspondingly rising. As many as 10 to 15 movements from many various types of transport, including tankers, tanks, railcars and tank trucks, include the flow of fuel from the oil fields to the market. Oil is processed along the path at distribution points and at ports and refineries. At each of these transportation phases or storage places, accidents can and do happen. Obviously, therefore, ensuring that there are as few leaks as practicable is an vital aspect of preserving the atmosphere, and if they do occur, their disposal must be successful. For the past 40 years, Chang and Lin (2006) have studied 242 incidents related to storage tanks in manufacturing facilities. 74 percent of incidents happened in petroleum refineries, oil ports, or storage facilities, they said. Microbial biodegradation seems to be a

promising instrument for managing petroleum emissions in these ecosystems in order to remediate petroleum pollutants, and it is not shocking that much research is ongoing to examine the biochemistry and genetics involved in this operation. One of the processes by which hydrocarbons are assimilated by microorganisms is the formation of metabolites or particular agents that allow liquid hydrocarbons to be spread as hydrocarbons in water emulsions, micro-droplets or miscellaneous agents that are subsequently transported into the cell through arc. There are arc metabolites or biosurfactants. Arc biosurfactants formed or extracellularly excreted on microbial cell surfaces and include both hydrophilic and hydrophobic moieties that decrease surface tension and interfacial tension between individual surface and interface molecules. These compounds can be used, like chemical surfactants, in several processes including emulsification, foaming and detergency, globalisation and dispersion or solubilization (Desai and Banat 1997; Karanth et. al., 1999; Bodour et al , 2003). Compared to traditional surfactants, biosurfactants have many advantages: lower toxicity, greater biodegradability, improved environmental compatibility, higher foaming, greater selective and precise extreme temperature operation, pH and salinity, and the ability to synthesise from sustainable feedstock (Desai and Banat 1997; Kim et al., 2000; Kumar et. al., 2006). Research on the development of biosurfactants by microorganisms has recently increased. Kim et. al. (2000) defined how Nocardia sp. generates two forms of biosurfactants. L-417 as a carbon source while developed on hexane. Similarly, Bodour et. al.

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(2003) documented the composition and characteristics of a new genus of Flavobacterium sp. biosurfactant-flavolipds. To MTNII. Ilori et. al. (2005) identified factors influencing oil-degrading biosurfactant development by Aeromonas sp. The active usage of biosurfactants to promote oxidation of organic compounds in soil and dispersion of oil from oil spills has also been shown in many studies (Oberbremer et. al., 1990; Banat et. al., 1991; Banat et. al., 2000). Rather recently, in the presence of a biosurfactant generating bacterium, Kumar et. al. (2006) examined biodegradation of oil by hydrocarbon degrading Pseudomonas putida. Several authors stated that in aqueous and soil matrices relative to the individual culture, the co-culture exhibited enhanced degradation capabilities in a reproducible manner.

Crude Oil Constituents Hydrocarbon

Hydrocarbon compound mixtures varying from tiny, volatile compounds to very big, non-volatile compounds are crude oils. This combination of compounds differs based on the geographical formation of the region in which the oil is contained, and the characteristics of the oil are highly influenced. Crude oils that compose mostly of broad molecules, for instance, are viscous and thick. Petroleum goods, such as petrol or diesel fuel, are mixtures with less substances and are hence more specific and less volatile in their properties. Compounds of hydrocarbons consist of hydrogen and carbon, and are also the core components of oils. Various quantities of sulphur, phosphorus, oxygen and occasionally mineral salts are often found in oils, as well as heavy metals such as nickel, vanadium, and chromium. In general, their composition characterises the hydrocarbons present in oils. The structures of hydrocarbons present in oil aresaturates, olefins, aromatics and polar compounds, some of which are shown in Figure 1.

Figure 1 Chemical compound in oils

The saturated part category in oils comprises mainly of alkanes, which are hydrogen and carbon compounds with the highest amount of atoms of hydrogen in each carbon. Thus, since the carbons are filled with hydrogen, the word 'saturate' is used. Cyclo-alkanes, which are substances made up of the same carbon and hydrogen constituents, but with carbon atoms bound to each other in loops or loops, are often found in the saturate group. Larger concentrations of saturate are also referred to as waxes. Olefins, or unsaturated compounds, are another type of compounds having less than the full possible number of hydrogen atoms. Olefins have a requirement of one double

Figure 1 The chemical compound that substitutes two hydrogen atoms in the oilsearbon-to-carbon bond. Only finished goods produce large quantities of olefin. At least one benzene ring with six carbons contains the aromatic compounds. In the loop, three double carbon-to-earbon bonds float and provide stabilisation. Because of this durability, benzene rings may have harmful consequences on the environment and are very durable. Sometimes referred to as BTEX or benzene , toluene, ethyl-benzene, and xylenes are the most important smaller and hazardous compounds present in gasoline. Compounds made of at least two benzene rings are polyaromatic hydrocarbons, or PAHs. PAHs represent approximately 0 and 60 percent of the oil content. As a product of combining with molecules such as sulphur , nitrogen, or oxygen, polar compounds are those that have a large molecular charge. The "polarity" or charge that the molecule bears results in action that is distinct from that of unpolarized molecules in certain conditions. The smallest polar compounds in the petroleum industry are called "resins," which are primarily responsible for oil adhesion. "Asphaltenes" are considered the larger polar compounds because they sometimes make up the greatest proportion of asphalt, which is widely used for road building. Asphaltenes also have very large molecules which have a major impact on the action of the oil if they are abundant in the liquid.

Biodegradation of crude oil

The primary process for decontamination of polluted petroleum and vegetable oil sites is microbial degradation of hydrocarbons. Specific biodegradation information has been extended to the clean-up of toxic land and sealed oil sites. In 1967, after it was permanently moored in Long Beach Harbour, contractors used bioremediation to boost the efficiency of 800,000 gallons of oily waste water left in Queen Mary's bilge tanks. In situ bioremediation is already used by several refineries, tank farms and transfer stations to reclaim soil polluted by unintended oil leaks or other hydrocarbons (Zitrides 1990). As bioremediation applications on land can be carried out in closed or semi-closed environmental environments and can be tightly managed and tracked, bioremediation of
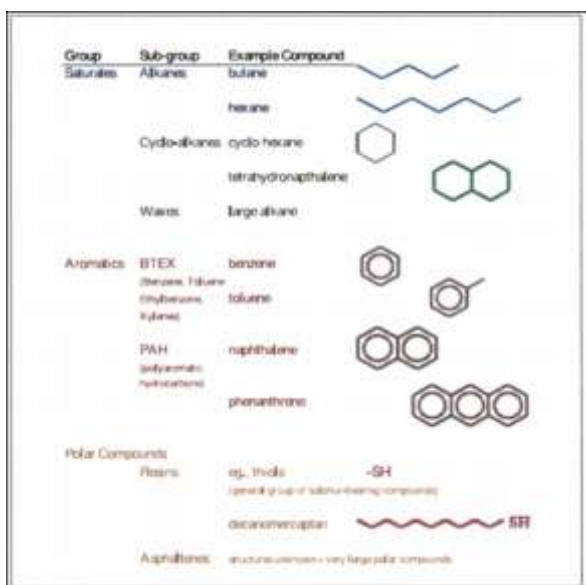
oil-contaminated soil is an increasingly increasing usage of this technology. Bragg et at (1994) documented the efficacy of bioremediation by the fertilisation technique in the Exxon Valdez Spill. On multiple shorelines, this approach has proved useful (Swannell et at., 1996; Swannell et at., 1999; Venosa et at., 1996). It helps in the reduction of pollutants by bulk. It is also a popular way of handling sites polluted with hydrocarbons (Eweis 1998). The reaction to the Exxon Valdez Oil Spill was the biggest campaign ever activated for mitigation and recovery. In 1989, mitigation activities centred on extracting bulk oil from the shorelines and involved manual oil processing with sorbent pads, hot and cold water cleaning on the shore, and bioremediation (Harrison, 1991). Around 20% spilled evaporated and undergone photolysis, 14% Biodegradation is controlled by numerous biotic and abiotic influences.

Abiotic Factors Affecting Biodegradation

Hydrocarbon biodegradation is highly affected by some abiotic influences, such as crude oil quality, oxygen and nutrient supply, pH, salinity and temperature. It is also impaired by the compounds' accessibility to microbial attack. In the event of oil leaks in dust, this relies on the concentration, hydrcphobicity, and adsorbance of soil particles. The maximum concentrations of biodegradation for saturates are accompanied by low molecular weight aromatics. It is considered that resins and asphaltenes are recalcitrant or have an exceptionally low degradation rate. Singer et al (1983), Horowitz et. al. (1975) have demonstrated an efficient degradation rate by microbes of these compounds. Co-oxidation has been due to the microbial breakdown of certain substances. In the presence of another readily degraded hydrocarbon, a hard-to-degrade hydrocarbon can be partially degraded in this process. Biodegradation of hydrocarbons requires oxidation by oxygenases of the substrate, so oxygen is the most essential aerobic biodegradation ingredient. There is also anaerobic biodegradation, but the incidence is quite limited (Frankenberger 1992; McNally 1998). Sediment experiments caused by the Amoco Cadiz spill find that anaerobic degradation is slower than aerobic biodegradation in many orders of magnitude (Ward and Brock, 1978). Under nitrate reducing (Mihelcic and Lutky 1988), sulphate reducing (Reuter et al, 1994; Zhang and Mille 1994), and methanogenic conditions (Karanth et al , 1999), anaerobic biodegradation may occur. Biodegradation and its speed are impaired by nutrients such as nitrogen, phosphorus and iron. There is a nutrient mismatch after major oil spills, with the C: N and C: P ratios shifting and thereby influencing the biodegradation process. In Prince William Sound, Alaska, nitrogen and phosphorous rich fertilisers were applied to increase the rate of biodegradation (Braddock et al , 1997). Both biological processes are influenced by pH and temperature, since these parameters influence the enzymes responsible for them. The optimal biodegradation temperature varies from 18 ° C to 37 ° C, but at temperatures below 5 ° C it has been shown to occur (Mohn et al , 2001; Whyte et al , 1998). The rate of hydrocarbon metabolism by microorganisms decreases at low temperatures. Lighter fractions of petroleum have become less volatile, thereby depressing microbial behaviour, leaving harmful constituents in the atmosphere for a longer period. At low temperatures, oils become more viscous; thus, less distribution exists and fewer surface space is usable for biological attack. The optimal pH is about 6-8 but is strongly alkaline, i.e. greater than 7, for most organisms (Dragun, 1988). The biodegradation of hydrocarbons has been shown to happen at a low pH of 2 by Stapelton et al .(1988). Biodegradation at a lower salinity is safer. Ward and Brock (1978) have demonstrated that while oil and hexadecane oxidation is carried out at salinity levels varying from 3.3% to 28.4%, compound metabolism concentrations decline as salinity rises. However, it has also isolated several marine bacteria capable of destroying hydrocarbons (Iwabuchi et al, 2002).

Biotic Factors that Cause Biodegradation

Hydrocarbon processing is due to bacteria of different kinds. It has been demonstrated that almost all the diverse substances are prone to microbial degradation. Few bacteria have the potential to dissolve multiple materials, while some may only destroy one or two components (Leahy and Colwell, 1990). Almost 40-80 percent of oil spill depletion is compensated for by biodegradation. Microbes with degrading potential for hydrocarbons have been extracted from diverse environments such as aquatic sediments, ground, estuaries, shore, etc. While some fungal genera have been known to dissolve hydrocarbons, such as Candida, Fusarium, Trichoderma, Aspergillus, bacteria are the most effective hydrocarbon breakdown agents. Pseudomonas, Acinetobacter, Bacillus, Alcaligens, Micrococcus (Bartha and Atlas 1977) contain bacterial genera capable of degrading hydrocarbons. Via P-oxidation, biodegradation of alkanes occurs. In general, aromatic hydrocarbon rings are hydroxylated to form diols which form cathccols and then give the TCA cycle intermediates. Tran-sdiols, all of which are carcinogenic, form fungal structures, whereas bacteria nearly exclusively contain cis-diols. Non-toxic final products such as CO_2 and water are formed by the full oxidation of aromatic hydrocarbons. The bacteria's previous exposure to hydrocarbons improves their degradation ability and the population of hydrocarbon degraders in the polluted sites often rises. Hydrocarbon oxidation genes are also coded by plasmids, and bacteria derived from polluted areas have a higher frequency. Many Pseudomonas plasmids have been well examined, such as TOL plasmid for toluene degradation, XYL for xylene degradation, CAM for camphor, and SAL for salicylate. Characterization of

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hydrocarbon-degrading microbial communities in polluted and pristine soils by Margesin et al .(2000) revealed that genotypes comprising gram-negative bacteria genes {Pseudomonas putida alk B, xyl E and ndo B and Acinetobacter alk M) were found to be substantially higher in degraded soils (50-75%) than in pristine soils (0-12.5%). Some scientists also used genetically engineered bacteria for hydrocarbon degradation, such as Chakrabarty (1976). The reason for making such GEMs is that they may be engineered to be more competitive than natural organisms or have the potential to degrade substances that are not degradable by species that exist naturally. Thus, bioremediation, which has barely any harmful consequences and is cost-effective, is evolving as an oil spill cleanup technology.

REVIEW OF LITERATURE

Impact of hydrocarbons on microorganisms A transition happens in the population size and microbial composition of the polluted site if there is an oil spill in the setting. Results of a Cohen (2002) analysis suggest that oxidation of oil was mainly induced by aerobic heterotrophic bacteria. Strains o f sulfate-reducing bacteria and aerobic heterotrophic bacteria o f aliphatic and aromatic hydrocarbons is capable of degrading model compounds o f. In the presence of hydrocarbons, hydrocarbon degradation by microbial organisms relies on the structure of the group and its adaptive reaction (Walker and Colwell 1976).

Effect on community size The hydrocarbon-degrading bacteria account for < 1% of the overall bacterial population in the normal uncontaminated world, according to Atlas (1981). But the proportion of hydrocarbon-degrading bacteria rises exponentially when there is an oil leak (Atlas, 1981). A selective enrichment of hydrocarbon-degrading bacteria from 0.028 percent at the beginning of a bioremediation trial to almost 100 percent two months after the spillage was found by Patel et al .(2003). The process is called adaptation, which stems from the hydrocarbon-oxidizing ability of the population (Spain et al., 1980). Higher oxidation rates and greater reduction of n-alkanes for bacteria grown in mousse were found by Al-Hadhrami et al .(1996) relative to those grown in nutrient broth, suggesting environmental sensitivity of the research bacteria. The Sunderbans Biosphere Reserve coastal water petroleum degrading bacteria survey found the highest amount of bacteria in the waters of Haldia Port and its nearby region, where the water is heavily polluted by discharges from refineries (Roy et al, 2002). During the first month of the trial, studies on the bioremediation of crude oil polluted subantarctic intertidal sediment demonstrated a two-fold magnitude rise in saprophytic and hydrocarbon utilisation by microorganisms (Delille et al , 1998; Delille et al . , 2002). Radwan et al .(2000) stated that the amount of hydrocarbon degraders in the soil

sample rose immediately after crude oil was applied. Song and Bartha (1986) found that, as a consequence of jet fuel exposure, microbial numbers increased by 2-2.5 orders of magnitude in subsurface soil. After exposure by polycyclic aromatic hydrocarbons (PAH), the amount of phenanthrene and chrysene degrading populations increased (Melcher et al , 2002). Soil and water polluted with diesel fuel contained 6xl06 to lOOxlO6 phenantherene damaging bacteria, although there were no observable or moderate amounts of these species in uncontaminated places such as garden soil and reservoir water (Bogardt and Hemmingsen 1992). Geiselbrecht and Staley (1996) recorded that sediments containing 104-107 polycyclic aromatic hydrocarbon degrading bacteria (dry weight) per gramme of sediment from a creosote-contaminated EPA superfund site (Eagle Harbour) produced 104-107 polycyclic aromatic hydrocarbon degrading bacteria (dry weight) per gramme of sediment, while uncontaminated site concentrations ranged from 103-104 per gramme of sediment. After the fifth day, they also noticed a decrease in the amount of bacterial cells.

Impact of Pollution of hydrocarbons on microbial diversity Hydrocarbon exposure impacts the microbial diversity of the polluted site and has been removed from the contaminated site by different microorganisms with the potential to degrade hydrocarbons. Such microorganisms that have the potential to remove the contaminant rule a polluted place. Strained processes are also less diverse than processes that are not strained. During nutrient-enhanced oil spill bioremediation, research by Roy et al .(2002) on the ecology of bacterial communities found that bioremediation treatments lowered the biomass of the bacterial communities. The impact of crude oil on mixed microbial community metabolic behaviour has shown that hydrocarbon involvement raises the proliferation of hydrocarbon-degrading bacteria, but reduces microbial diversity (Nyman 1999). Although several hydrocarbon degradation capable genera have been examined, Pseudomonas, Acinetobacter, Achromobacter, Micrococcus, Alcaligens, Vibrio, Xanthomonas among bacteria and Aspergillus, Penicillium, Saccharomyces, Fusarium, Trichoderma, Candida among fungi are prominent among them (based on the frequency of isolation) (Bartha and Atlas, 1977; Cemiglia 1993; Vivekananda et al , 1999; Walkaromyces, Ridway (1990) recorded that 89.7 percent of the bacteria extracted and classified from a gasoline-contaminated aquifer were Pseudomonas, Alcaligens, Nocardia and Micrococcus. The Pseudomonas and Sphingomonas genera contained nine out of eighteen isolates that were capable of utilising naphthalene and phenanthrene as the sole source of carbon collected by Meyer et al .(1999). Different findings have been reported by Shen et al .(1998). Microbial ecological studies of two districts of Gujarat found that among the hydrocarbon-degrading bacterial genera (Patel and

Ghosh 2003), Pseudomonas was the prevailing genus. Analysis of water and sediment samples from the Cuban hydrocarbon degrading bacteria shelf showed that there were 6 genera. The most prevalent is Pseudomonas, Flavobacterium, Achromobacter, Bacillus, Alcaligenes and Micrococcus (Montero et al , 1996). Isolates generated from the San Diego bay sediments tainted with various polyaromatic hydrocarbons and hexadecane by Melcher et al .(2002) belonged to numerous marine genera such as Vibrio, Marinobacter or Cycloclasticus, Marinomonas, Pseudoalteromonas and Halomonas. Over many (sub) phyla (a, p,-y proteobacteria) and gramme positives, more than 20 genera of marine hydrocarbon degrading bacteria have been defined. Hydrocarbon degrading capabilities have also been identified by Flexibacter-CytophagaBacterioids so far (Engelhard! et al , 2001; Floodgate 1995; Gauthier et al, 1992; Geiselbrecht et al , 1996; Yakimov et al , 1998). In their research on polycyclic aromatic hyrocarbon degradation, Iwabuchi et al .(2002) noticed that the most influential degraders of marine bacteria were representatives of the genus Cycloclasticus. Harayama et al .(1999) stated that when nutrients were augmented, the Alcanivorax community became more prevalent in oil-contaminated sea water. In addition to bacteria, 22 hydrocarbon-degradable fungi and cyanobacteria have been examined (Pelz et al , 1999; Raghukumar et al, 2001). Efficient hydrocarbon degraders have also been shown to be natural and synthetic bacterial consortia (Mohandass et al . 1997; Foght et al . 1990; Vivekanandan et al . 1999; Thouand et al . 1999; Ward et al . 2003).

Petroleum Hydrocarbons Aerobic Biodegradation
Several various forms of organic compounds are present in petroleum hydrocarbons, including alkanes (normal, iso- and cycle-), aromatics, polycyclic aromatics, heterocyclic and asphaltic compounds. In the natural world, each of these substances has a variable degree of biodegradation. The chemical composition of each substance regulates its propensity for biodegradation, but the rate and degree of biodegradation is often influenced by other influences, such as solubility, toxicity, and contact with other molecules (Bartha 1986). Microorganisms that use petroleum hydrocarbons as a supply of cellular carbon and energy are commonly spread in nature (bacteria, yeasts, and fungi). More than 100 bacterial strains that degrade petroleum hydrocarbons (Foght and Westlake 1984) have been described. The following genera belong to several of the more common bacterial species: Pseudomonas, Achromobacter, Arthrobacter, Nocardia, Vibrio, Brevibacterium, Corynebacterium, and Flavobacterium. Bacteria are commonly thought to be mainly responsible for the deterioration of petroleum hydrocarbons in an atmosphere that is not under threat. Compared to yeasts and fungi in an atmosphere polluted by petroleum hydrocarbon products, they are normally in larger quantities. In

conditions precluding bacterial growth, fungi and yeasts perform better in degrading these compounds (Foght and Westlake 1984).

Alkane biodegradation Relative to other hydrocarbons, straight chain n-alkenes are typically more quickly deteriorated, but narrower chain length alkenes (C5 to CIO) are inhibitory to some of the microorganisms that degrade hydrocarbons. Ses molecules serve as a solvent that interrupts the cell's membrane structure. Many microorganisms more readily degrade intermediate chain lengths (CIO to C20) (Bartha 1986). Longer chain length alkanes (>C20), also referred to as' waxes,' are very hydrophobic in nature and are thus quite steadily deteriorated. The alkane chain's branching prevents oxidation. Therefore, microorganisms dissolve n-Cn and n-Cig alkanes quickly, whereas their branched chain equivalents, pristane and phytane, are sluggish to dissolve (Foght and Westlake 1984). The cyclic alkanes degrade very slowly as well. Some monocyclic compounds, such as cyclopentane, cyclohexane, and cycloheptane, have a solvent impact on microbial cell lipid membranes and are thus poisonous to most bacteria that degrade hydrocarbons (Perry 1984). In the presence of oxygen, the primary assault on the hydrocarbon molecule is by the activity of oxygenases. The alkane mono-oxygenase reaction results in an alcohol compound The alcohol compound is then oxidised into an aldehyde and an acid component, respectively. Via beta-oxidation, the acid substance may be further deteriorated. The involvement of divisions interferes with the process of beta-oxidation (Bartha 1986). In the presence of other readily degradable substances (Foght and Westlake 1984), cycloalkanes are vulnerable to degradation by co-metabolism.

Biodegradation of aromatic hydrocarbon compounds Initial aromatic hydrocarbon degradation by prokaryotic microorganisms (bacteria) is carried out by a dioxygenase enzyme mechanism, resulting in cis-hydrodiols which are further oxidised to dihydroxy products. In the case of benzene, catechol is a dihydroxy substance (Bartha 1986). In eukaryotic microorganisms, a mono-oxygenase enzyme initially oxidises the hydrocarbon to a 1 , 2 oxide. This is accompanied by the addition of a water molecule, which is then oxidised to an aromatic hydrocarbon, to yield a dihydroxy-dihydro product. To yield muconic acid or 2-hydroxy muconic semialdehyde, it can be cleaved at the ortho- or meta-position. It is necessary to metabolise these compounds into tricarboxylic acid cycle intermediates (Bartha 1986). Biodegradation of reservoirs or soils polluted with benzene, toluene, ethylbenzene, and o-, m-, and p-xylene (BTEX) compounds, typical components of gasoline, has been studied of several studies (Thomas et al, 1990, Goldsmith and Balderson 1988; Chiang et al, 1989). Either by leaky underwater storage facilities or by unintended leaks, these substances enter the

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atmosphere. At a site in Granger, Indiana, where a fuel spill polluted a shallow aquifer, Thomas et al. (1990) documented an in situ bioremediation system. Core sample results revealed that the subsurface microflora had all eliminated toluene, ethylbenzene, and m-xylene, but o-xylene persisted. The data showed that the biodegradation capacity at the site previously biostimulated remained enhanced 2 years after completion of the in situ biostimulation process. Goldsmith (1988) extracted bacterial strains capable of using gasoline-contaminated benzene, toluene, and p-xylene as primary carbon sources from aged dirt. In microcosm tests utilising soil and groundwater from a petroleum polluted field site in Michigan, Chaing et al. (1989) reported that when the dissolved oxygen (DO) level was greater than 2 mg/L, 80 to 100 percent of die aromatic hydrocarbon (BTX) at levels of 120 to 16,000 ppb was depleted with a half life (T1/2) of 5 to 20 days. When the DO amounts were smaller than 2 mg/L (T1/2, 20 to 60 days), the BTX decay rate slowed down significantly.

Polycyclic aromatic hydrocarbons Polycyclic aromatic hydrocarbons (PAHs) are a category of compounds in a longitudinal, triangular, or cluster structure of two or more fused aromatic rings. Generally, twoto three-ringed PAH compounds degrade at a comparatively quicker rate than PAH compounds containing more than three rings. In certain cases, the degradation rate of higher ringed PAHs is promoted by the existence of other structurally similar two- or three-ringed compounds (Park et al, 1990). Also, higher molecular weight PAHs can be co-metabolized to simpler intermediates in the presence of supplementary carbon streams, viz. biphenyl and succinate. The initial oxidation of a PAH by microbes follows the same route as was stated earlier for aromatic hydrocarbons, i.e., the formation of eis-dihydrodiols by a dioxygenase which then leads to the formation of catechols (Cemiglia 1984). The catechols are substrates for other dioxygenases that cleave the aromatic ring to cis, cis-muconic acid, or 2-hydroxy muconic semialdehyde. Figure 2 demonstrates the mechanism for naphthalene biodegradation by bacteria (Cemiglia 1984). Fungi oxidise PAHs into a mono-oxygenase and epoxide-hydrolase catalysed step to transdihydrodiols (as in mammalian systems), which are further degraded to simpler products (Cemiglia 1984). Payne and Phillips (1985) documented the biodegradation of PAHs in soil ecosystems with additional carbon and energy sources, and pH improvements from 6.1 to 7.5. The implementation of amendments lowered the T1/2 of the recalcitrant PAHs considerably relative to unamended schemes, e.g., the T1/2 for benzo(a)pyrene (B(a)P) decreased from 91 to 69 days. Thus, co-metabolism of the molecules produced a major difference in the biodegradation process. Payne and Phillips (1989) have stated that the inclusion of an acclimated soil microbial community in land fanning would dramatically increase the biodegradation rate of PAH compounds in fossil fuel wastes. A far greater percentage of PAH

elimination was found in the acclimated soils in 22 days relative to unacclimated soils in 40 days. McNally et al. (1998) examined the biodegradation of petrochemical sludges comprising PAH compounds. They picked B(a)P as the target contaminant in the sludge for their analysis, which was carried out in sealed continuous stirred reactors. The B(a)P concentrations ranged from 285 to 3,475 mg/kg of dry solids. The B(a)P and other PAH removals is greater than 90 percent.
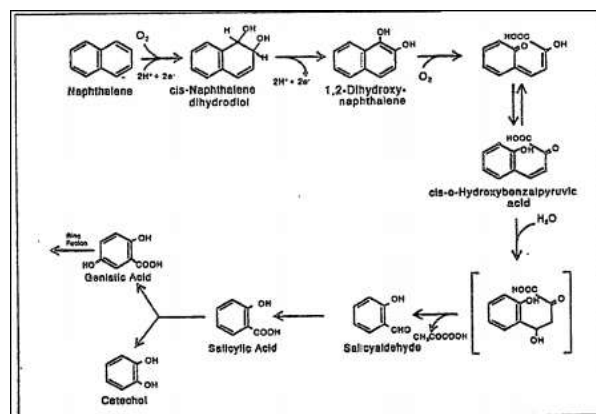

Figure 2. Naphthalene biodegradation pathway (Cernaglia 1984).

Biodegradation kinetics in Park et al .(1990), who observed substantial volatilization (22 to 33 percent) of naphthalene and 1-methylnaphthalene, the two-ringed compounds, recorded unacclimated soils for 14 PAH compounds. The volatilization losses were very minimal for other PAHs (three or more rings) (< 0.1 percent). There was some abiotic loss (1.8 to 17.4 percent) of two- and three-ringed PAHs, but this loss was slight for more than three-ringed compounds. Tic's biodegradation, modified for abiotic failure and volatilization, for two-ringed PAHs was approximately 2 days. The Tic was roughly 59 days for three-ringed PAHs but increased to more than 300 days for more than three-ringed PAHs.

OBJECTIVES OF THE STUDY

1.	Used bacteria extracted from different ecological locations to research oil biodegradation.

CONCLUSION

In conclusion, this analysis offers proof that oil-degrading bacteria may be extracted from environmentally-friendly oil sites. Among the isolates collected, the culture of bacillus cereus expressed the improved capacity of oil emulsification and degradation. This may mean that in the bioremediation of oil emissions, the bacillus cereus strain may be successfully used.

REFERENCES

1. Albalos AA, Pinazo MR, Infante M, Casals FA and Manresa A (2001) "Physicochemical and antimicrobial properties of new rhamnolipids produced by Pseudomonas aeruginosa AT10 from soybean oil refinery wastes." Langmuir 17: pp. 1367-1371.

2. Al-Hadhrami MN, Scott L and Fisher PJ, (1996) "Effects of the addition of organic carbon sources on bacterial respiration and n-alkane biodegradation of Omani crude oil." Mar. Poll. Bull. 32(4): pp. 351-357.

3. Al-TahhanRA, Sandrin, TR, Bodour AA and Maier RM (2000) "Rhamnolipid-induced removal of lipopolysaccharide from Pseudomonas aeruginosa: effect on cell surface properties and interaction with hydrophobic substrates." Appl Environ Microbiol 66: pp. 3362- 3268.

4. Anderson TA, Guthrie EA and Walton BT (1993) "Bioremediation in the rhizosphere: plant roots and associated microbes clean contaminated soil." Environ Sci Technol. 27: pp. 2630-2636.

5. Aprill W and Sims RC (1990) "Evaluation of the use of prairie grasses for stimulating polycyclic aromatic hydrocarbon treatment in soil." Chemasphere 20: pp. 253-265.

6. Atlas RM (1981) "Microbial degradation of petroleum hydrocarbons: an environmental perspective" 45: pp. 180-209.

7. Banat IM, Samarah N, Murad M, Homer and Baneijee S (1991). "Biosurfactant production and use in oil tank clean-up. World J. Microbiol. Biotechnol. 7: pp. 80-84.

8. Banat IM, Makkar RS and Cameotra SS (2000). "Potential commercial applications of microbial surfactants." Applied Microbiology and Biotechnology 53: pp. 495-508.

9. Baneijee S, Howard PH, Rosenberg AM, Dombrowski A E, Sikka H, and Tunis, DL (1984). "Development of a general kinetic model for bio degradation and its application to chlomphenols and related compounds" Environ. Sci. Tech. 18(6): pp. 416-422.

10. Banincasa M, Abalos A, Oliveira I and Manresa A (2004) "Chemical structure, surface properties and biological activities of the biosurfactant produced by Pseudomonas aeruginosa LBI from soapstoek. Antonie Van Leeuwenhoek 85: pp. 1-8.

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