Review Article

Biodegradation of petroleum hydrocarbon polluted soil

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Abstract

Crude oil contributes a major percentage to the source of energy used on earth, however, hydrocarbon components have been classified to the family of carcinogens and neurotoxic organic pollutants. The inappropriate drilling, transportation, and usage lead to the increment of soil, air, and water body petroleum hydrocarbon pollution. If this menace is not put in check as a matter of urgency, it can cause an epidemic outbreak in an affected community, shortage in agriculture produces output, threatening of soil useful microbial biome and environmental disaster. Bioremediation as promising modern biotechnology is capable of mineralizing hydrocarbon pollutants into water, carbon dioxide, cell proteins, and an inorganic compound. Indigenous or genetically modified microbes are able to secrete enzymes for the synthesis of biosurfactants that break down organic pollutants into a less toxic form. Bioremediation is not only potent in degrading organic pollutants, but it is also environmentally friendly and cheap compared to other non-natural methods. Therefore, this paper combined and presents a review of empirical researches done on microbial bioremediation of petroleum hydrocarbons pollutants in different countries.

1. Introduction

Petroleum hydrocarbon carbon contamination with environment has been a big threat to human and his natural surroundings. The media that is affected by these pollutants are soil, water and air. It brings change in the environment that resulted in way of introducing harmful effects that altered the quality of human life, animals, plants and microorganisms. To reduce the harms caused by pollution, the three media mention above need to be subjected to physical, chemical, mechanical or biological treatment. Generally in china, according to more than 8% of china’s arable land is severely polluted. This was reaffirmed by saying that one fifth area of China’s arable land is polluted, and the polluted arable land equal the size of Taiwan to a degree that farming should be prohibited there in totality. The United States Environmental Protection Agency(USEPA) most targeted sites of hazardous wastes clean-up programs are made of 600 of 1408 different kinds of hydrocarbons. Soil pollution is considered to be a lifethreatening problem because all the plant consumed by human and the animal is cultivated from the soil, therefore, proper attention most paid to soil pollution to reduce it to the barest minimal if not totally eliminated. Although soil remediation is of a high capital and labour intensive project, but due to man poise for cheaper method for polluted soil remediation, much have been done using microorganisms to bioremediate contaminated soil which proved positive. This method termed to be labour and capital cheap and environmentally friendly. However, not all bacteria have the adaptability to degrade pollutants like petroleum hydrocarbon especially in the soil, according it has been proven that microorganism’s capability and adaptability to degrade pollutants, depend on the individual strains ability to produce suitable biosurfactants and the formation of biodegrading association that is very strong and reliable throughout the degradation period. Also, it depend on the adaptability of the microorganism to carry...
out its physiological functions under natural condition that is characterized with limited water supply, low availability of nutrients and adverse pH condition.

2. Hydrocarbon Contaminated Soil

The present of crude oil in the soil usually by spilling altered its properties. Therefore, define crude oil pollution as the contamination of the environment by crude oil. The products of crude oil (Hydrocarbons, solvents, etc.) used as a source of energy in the oil industry have polluted about 80% of lands. There exist hundreds different combination of polycyclic aromatic hydrocarbons (PAHS), however, 28 compounds was identified to be hazardous by US EPA (United States Environmental Protection Agency) in 2008. Crude oil is made up saturated and unsaturated aliphatic hydrocarbon, monocyclic and polycyclic aromatic hydrocarbon; these components are pollutants that contaminates the soil but aromatic hydrocarbon are the most highly hydrophobic which rendered them difficult to degrade. Low molecular weight PAHs (LMW) and high molecular weight PAHs (HMW) are the classes of PAHs. naphthalene, fluorene, phenanthrene and anthracene with two to three (2-3) aromatic rings are LMW PAHs which are less toxic, while HMW PAHs of 4-7 aromatic rings (chrysene, Triphenylene, Pyrene, pentacene, Corannulene, and coronenes) are highly toxic, carcinogenic to human health and difficult to degrade. Petroleum hydrocarbon pollution in the soil reduces the level of environmental biomass, taxas and the biodiversity. The presence of hydrocarbon in the soil can alter both the physical and chemical properties of the soil, especially the C/N ratio and soluble salt content. The presence of petroleum hydrocarbons in the soil have significant effects on the soil properties, it was noticed in observable differences in data obtained when comparing crude oil-polluted soil and non-polluted soil. Hydrocarbon polluted soils were slightly acidic whereas the control soils set towards neutrality. Crude oil reduced water infiltration into the soil and also causes low permeability. Hydrocarbon pollution increase soil organic content, it reduces the concentration of nitrogen, phosphorus and calcium in the soil. And an increase in hydrocarbon level in the soil caused low fertility of the soil, which negatively affect the agricultural productivity which reduces the source of livelihood of both human and animals in that geographical area.

3. Soil bioremediation and its Mechanism

Bioremediation activities were first noticed among Romans around 600BC, where they used bioremediation to clean their wastewater, though their method was not as developed as today’s biotechnological methods. Bioremediation was officially invented by George Robinson when he used microbes to degrade an oil spill on the coast of Santa Barbara.

California in the late 1960s. Ever since this successful attempt, human has indulged on this natural method to treat contaminated environments. Bioremediation is the process of elimination or reduction of pollutants from the environment. It is an environmental or aesthetics waste detoxification mechanism with the help of microorganisms, plants, or their enzymes. However, when the waste detoxification is done at the contaminated site, it is referred to as In-situ Bioremediation. Contrarily, when the detoxification of waste is done outside the original site of the contamination is called Ex-situ Bioremediation. This process involves relocating the contaminants from the contamination site before treatment is administered in a controlled environment. Also, bioremediation can be in a form of bioattenuation which solely depend of natural process of degradation, biostimulation where indigenous microorganisms are stimulated by addition of nutrients, water, and electron acceptors or electron donors to the soil, and bioaugmentation where microorganisms of high degradation ability are inoculated to the soil to enhance bioremediation. Every bioremediation technology depends on many factors which include; the types, amount, and toxicity of a pollutant chemical species present in the soil site conditions and microbial activity (indigenous or exogenous strains). Bacteria remediation can be anaerobic; in the absence of oxygen as an electron acceptor, bacteria can use organic chemicals or inorganic anions as alternative electrons acceptors. The bacteria involves in this bioremediation do not employ the utilization of oxygen during their metabolic pathway. And bacteria can also be aerobic; the bacteria involve employing oxygen to carry out effective biodegradation of pollutants in the soil. In general, oxygen acts as an electron acceptor by accepting the electrons from oxidizing organic substances (reduction) the organic substance oxidized loses electrons to oxygen (oxidation). For this study, aerobic bioremediation is explained in detail. Mineral oil contents and halogenated products of petrol chemicals are the most important classes of organic pollutants in the soil; therefore, the enzymatic- metabolic activities of aerobic bacteria specifically pertinence for the speedily and complete biodegradation of such organic compound and their various products. The optimal degrading potential of aerobic microorganisms can only be attained if the organic pollutant is accessible to the biodegrading microorganisms. When bacteria pick up organic pollutants as illustrated in Figure 1, the first intracellular remediation approach is enzymatic (oxygenases and peroxidases) oxidative process, cell biomass synthesis particularly from the metabolites in the center precursor, e.g., acetyl-CoA, succinate, pyruvate, and Sugars. These are required for various biosynthesis and growth. They are synthesized by gluconeogenesis.
and finally, the organic pollutants are converted into intermediates of the intermediary metabolism by the surrounding degradation pathways, e.g., the tricarboxylic acid cycle.\textsuperscript{26}

![Bacteria bioremediation procedures](image)

**Fig. 1**: Bacteria bioremediation procedures.\textsuperscript{27}

4. **Influencer factors of biodegradation of Petroleum hydrocarbon**

4.1. **Temperature**

A slight increase in temperature of the soil tends to increase the bioremediation rate while decrease temperature reduces the rate of bioremediation.\textsuperscript{28} There was a significant reduction in the level of total PAH and phenol with an increase in temperature of inoculated treatment in mesocosm studied. There was observable increased in desorption and bioavailability with an increase in temperature.\textsuperscript{29} This could also be a result of the effect of temperature on microbial enzymatic makeup.

4.2. **Nutrients**

Nutrients play a major role in bioremediation; microbes need a commensurate amount of nutrients to carry out their metabolic activities, cell division, and growth. Trace elements that will serve as electron donor and electron acceptor are added into the soil in a form of organic (compost) and inorganic (fertilizer) this not only stimulates but also accelerate bioremediation.\textsuperscript{30} Bioremediation of petroleum hydrocarbons polluted soils can be enhanced with appropriate selection, combination and application of suitable nutrient to mesocosm.\textsuperscript{31}

4.3. **Bioavailability of Contaminant:**

Availability of contaminant has a significant effect on the metabolic component of a cell, this can be view in three contaminant’s availability concentration levels, firstly, at the absent of contaminants, biodegradation will not occur because there will be insufficient energy supply to induce bioremediation. Secondly, at a low contaminant concentration level, bioremediation will occur but at a slow rate, this is a result of no spontaneous increase in the microbial population because there is no availability of energy required for cell division, therefore, the cell will maintain a resting stage. Finally, if there is enough bioavailable contaminant, energy supply will increase thereby inducing biodegrading, at this time it will be in a growing stage, due to cell division and an increase in growth rate.\textsuperscript{32}

4.4. **Moisture content**

Optimum microbial growth can only occur in the presence of water, soil water content regulates oxygen diffusion. From a research work, it was found out that keeping the moisture content of the soil at an optimum value is critically important for a successful bioremediation process. It was also stated by an author that modification of water content of sandy clay soil to 60% of its field capacity, and moisture adjustment is a proper strategy to degrade the contaminated soil.\textsuperscript{28,33}

4.5. **Electron donor, Oxygen content and Redox Potential:**

The potential inhibitory effect of molecular oxygen is negated by excess addition of electron donor. Degradation is inversely proportional to redox potential, increase in oxidation of redox conditions, will result is decrease is bacterial perchlorate degradation. Electron acceptors such as sulfate, manganese oxides and iron oxides have significant effects on redox potential.\textsuperscript{34}

4.6. **PH**

Soil pH alteration by liming process can be of a great important in PAH degradation, as some hydrocarbon feeding bacteria attained maximum potential for degradation in certain level of pH range.\textsuperscript{35} However, in the presence hydrocarbonoclastes 2% Crude, changes in pH have no significant effect on the growth of bacterial strains.\textsuperscript{36}

5. **Bioaugmentation**

In recent years, researchers have shown that aerobic microorganisms have the adaptability to degrade hydrocarbon in the soil but are limited by accessible oxygen, which will act as an electron acceptor to sustain and keep the growing population. For these, researchers developed a means to put this problem in check through bioaugmentation. Bioaugmentation, as the word implies, involves the addition of natural, cultured or genetically modified microorganisms with required metabolic adaptability into treatment which could be contaminated soil, water or sewage for degradation of targeted pollutants or pollutants. When bacteria are exposed to pollutant for a long period it evolved adaptive abilities such as increased in content of cyclopropane fatty acids, Saturation of membrane fatty acids, isomerization of cis unsaturated fatty acids to their appropriate trans isomers, Production of stress proteins, Changes in cell morphology and Toxic pollutants as substrates for the efflux system.\textsuperscript{37}
In hydrocarbon contaminated soil, selected hydrocarbon-degrading microorganisms are inoculated into the soil to speed up the biodegrading capacity of the hydrocarbon in the soil. A single oil-degrading strain inoculates into hydrocarbon culture shows high degrading capacity as it was noticed by a researcher, an isolate G7 (Brevibacillus agri) had high ability to degrade aromatic fraction (61.14%), isolate G3 (Pseudoxanthomonas taewanensis) had high ability to degrade aromatic fraction (38.27%) and resin fraction (29.26%). It was also reported by a researcher that P. aeruginosa BAS-Cr1 was able to degrade oil sludge with more than 80% degradation of TPH at 5% that P. aeruginosa BAS-Cr1 was able to degrade oil sludge with more than 80% degradation of TPH at 5% and 10% concentration within 42 days of treatment. Findings by another researcher show an increase in the degradation rate of 4-chloronitoben-zenes (4CNB) degradation in soil microcosms by inoculation of pure cultured Pseudomonas putida ZWL73. However, studies have shown that the use of consortia with different kinds of aromatic-degrading bacteria has been more efficient in degrading pollutants as compared with using selected single strain. Assessing the degrading mineralize anthracene, phenanthrene and pyrene abilities of a selected microbial consortium of Mycobacterium fortuitum, Bacillus cereus, Microbacterium sp., Gordonia polysoprenivorans, Microbacteriaceae bacterium, and Fusarium oxysporum. Within 70 days, an average degradation of 96% to 99% was recorded in mineralize anthracene, phenanthrene, and pyrene present in the soil, with an initial dose of (250, 500 and 1000 mg kg⁻¹). It was also observed that the PAH in the soil was degraded by 70% by the same consortium within the same incubation period of 70 days. Researches have shown that when a consortium is immobilized, its soil hydrocarbon degradation ability increases. A researcher compared the differences between biostimulation and bioaugmentation treatments on crude oil-contaminated soil; the researcher reported that treatment with bacteria immobilized peanut hull powder has the most effective treatment of hydrocarbon biodegradation in soil. According to, immobilization of microbial cells system for bioremediation possess many advantages with few includes; resist to toxic chemical attack, solvent, heavy metals, temperature, and pH, providing suitable microbial environmental conditions, protection against shear damage, high flow rates of and volumetric productivities, elimination of cell washout problems at high dilution rates, providing cell high biomass. Biocarriers enhanced diffusion of oxygen in the soil, nutrient mass transfer to the bacteria and improved water-retention capacity that served as a limiting factor for bioremediation such contaminants as crude oil.

5.1. Biostimulation for Hydrocarbons Degradation

Biostimulation is a method of remediation of a polluted environment through the addition of stimulants like nutrients, oxygen, and water to the environment of the contaminated site which could be soil, water or sludge, to initiate rapid multiplication and growth of the microbial biome for rapid bioremediation. The stimulation of microorganism’s activities by the addition of substrates, oxygen vitamins, and other microorganisms tolerated compounds to enhance the degradation of pollutants such as petroleum hydrocarbon is termed biostimulation. The depletion of Nitrogen and phosphorus in petroleum-contaminated soil can be responsible for the low degradation of hydrocarbon in contaminated sites. The importance of nutrients for biodegradation of a contaminated site was noticed in the positive correlations of nitrogen and phosphorus with hydrocarbon. Abundant nitrogen and phosphorus in crude oil-contaminated site goes a long way in stabilizing the even distribution of microbial community and the richness if the site which is an essential factors for biodegradation of hydrocarbon.

6. The effect of Biosurfactant on Hydrocarbon Biodegradation

Biosurfactants is amphiphatic configured polymers with prominent hydrophobic and hydrophilic moieties which enable them to mold micelles that gathered at the interface between liquids of different polarities such as water and wax, they are polymers that are partial or total extracellular secreted by bacteria. It modified the surface properties of bacteria cells; increase the bioavailability hydrocarbon and the surface area of hydrophobic water-water insoluble substances like petroleum crude oil. Biosurfactant can function at extreme pH and salinity, as well as variable temperature conditions, less toxic, biodegradable and non-hazardous.

Biosurfactants which are generally derived from the secondary metabolites of microorganisms, are important biomolecules in environmental biotechnology because of its application in oil industries especially oil spill sites bioremediation, recovery of oil, cleaning of oil storage tank from sludge and environmentally friendly characteristics. The physical and chemical properties of microbial biosurfactants such as biodegradability, foaming, and environmental compatibility give them the edge over their equivalent.

Biosurfactant is an effective biostimulant in crude oil hydrocarbon degradation. This was found out in a study of effect of biosurfactant and fertilizer on biodegradation of crude oil by marine isolates of Bacillus megaterium, Corynebacterium kutscheri and Pseudomonas aeruginosa by researchers, according to the research findings, Biosurfactants alone are capable of promoting biodegradation process, if the polluted site have the required nutrients. However, little significant increase in
Table 1: Different countries bioaugmentation with the microbial system and nutrient used

| Countries                 | Microbial system used                                                                 | Nutrients used                                                                                                                                 |
|---------------------------|---------------------------------------------------------------------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------|
| USA                       | Pure or mixed cultures of Bacillus, Clostridium, Pseudomonas, and Gram-negative rods; mixed cultures of hydrocarbon degrading bacteria; mixed cultures of marine source bacteria; spore suspension of Clostridium; indigenous stratal microflora; slime-forming bacteria; ultramicrobacteria | Molasses 2–4%; Molasses and ammonium nitrate addition, addition of Free corn syrup and mineral salts, Maltodextrine and organic phosphate esters (OPE), Salt solution, Sucrose 10% +Peptone 1% + NaCl 0.5–30%, Brine supplemented with nitrogen and phosphorous sources and nitrate, Biodegradable paraffinic fractions + mineral salts, Naturally contain inorganic and organic materials + N, P sources. |
| Russia                    | Pure cultures of C. tyrobutiricum; bacteria mixed cultures; indigenous microflora of water injection and water formation; activated sludge bacteria; naturally occurring microbiota of industrial (food) wastes. | Molasses 2–6% with nitrogen and phosphorous salt addition ; Water injection with nitrogen and phosphorous salt and air addition ; Waste waters with addition of biostimulators and chemical additives ; Industrial wastes with salts addition Dry milk 0.04%. |
| China                     | Mixed enriched bacterial cultures of Bacillus, Pseudomonas, Eurobacterium, Fusobacterium, Bacteroides; Slime-forming bacteria: Xanthomonas campestris, Brevibacterium viscosgenes, Corynebacterium gumiform; Microbial products as biopolymers. | Molasses 4–6%; Molasses 5% + ; Residue sugar 4% +; Crude oil 5% ; Xanthan 3% in waterflooding. |
| Australia                 | Ultra microbacteria with surface activeProperties.                                   | Formulate suitable base media.                                                                                                                                                                           |
| Bulgaria                  | Indigenous oil-oxidizing bacteria from water injection and water formation            | Water containing air C ammonium and phosphate ions ; Molasses 2%                                                                                                                                          |
| China                     | Pure culture of Leuconostoc mesenteroides.                                            | Dry sucrose C sugar beet molasses dissolved in water.                                                                                                                                                   |
| Former Czechoslovakia     | Hydrocarbon oxidizing bacteria(predominant Pseudomonas sp.):sulfate-reducing bacteria | Molasses.                                                                                                                                                                                                |
| England                   | Naturally occurring anaerobic strain, high generator of acids; special starved bacteria, good producers of exopolymers. | Soluble carbohydrate sources ; Suitable growth media (type E and G.                                                                                                                                 |
| Former East Germany       | Mixed cultures of thermophilic Bacillus and Clostridium from indigenous brine microflora | Molasses 2–4% with addition of nitrogen and phosphorous source.                                                                                                                                         |
| Hungary                   | Mixed sewage-sludge bacteria cultures (predominant: Clostridium, Desulfovibrio, and Pseudomonas). | Molasses 2–4% with addition of sugar and nitrogen and phosphorous source.                                                                                                                              |
| Norway                    | Nitrate-reducing bacteria naturally occurring in North Sea water.                    | Nitrate and 1% carbohydrates addition to injected Sea water.                                                                                                                                             |
| Poland                    |Mixed bacteria cultures (Arthrobacter, Clostridium, Mycobacterium, Peptococcus, and Pseudomonas). | Molasses.                                                                                                                                                                                                |
| Romania                   | Adapted mixed enrichment cultures (predominant: Bacillus, Clostridium, Pseudomonas, and other Gram-negativerods). | Molasses 2–4%.                                                                                                                                                                                             |
| Saudi Arabia              | Adequate bacterial inoculum according to requirements of each technology.            | Adequate nutrients for each technology.                                                                                                                                                                  |
| The Netherlands           | Slime-forming bacteria (Betacoccus dextranicus).                                     | Sucrose-molasses 10%.                                                                                                                                                                                   |
| Trinidad-Tobago           | Facultative anaerobic bacteria high producers of gases.                             | Molasses 2–4%.                                                                                                                                                                                             |
| Venezuela                 | Adapted mixed enrichment cultures.                                                   | Molasses.                                                                                                                                                                                                  |

Source 46
Table 2: Biostimulation using difference organic wastes

| Biostimulation contents | Bioremediated Contaminant | TPH % degradation | Bioremediation Duration | Conclusions | References |
|-------------------------|---------------------------|-------------------|-------------------------|-------------|------------|
| Cow dungs and Sewage slug | Used engine oil | 94% and 82% | 98 days | Cow dung and sewage sludge can be an effective organic amendment for the biodegradation of used lubricant contaminated soil. | 4 |
| Tea leaf, soy cake and potato skin | Petroleum Hydrocarbon diesel fuel | 40-89% | 126 days | The study therefore proves the viability of using soy cake amendment in remediating hydrocarbon contaminated soil. | 2 |
| Peanut hull powder (15% w/w, no immobilized cells) | Contaminated soil samples from an oil storage site | 38% | 12 weeks | Oil degradation was enhanced using peanut hull powder as biocarrier. As it enhanced the transferred of biomass, water, oxygen, nutrients and hydrocarbons | 43 |
| Brewery, Spent grains, Banana skin and Spent mushroom compost | Petroleum Hydrocarbons used engine oil | 79% and 92% for 5% oil contamination 36% to 55% for 15% oil contamination | 84 days | There was significant removal Of TPH using the organic nutrient sources. | 51 |
| Non-Sterile poultry wastes | Polluted Mangrove Swamp Soil | 70% | 42 days | Non-sterile poultry waste can effectively and efficiently enhance removal of petroleum from polluted site. | 52 |
| Domestic wastewater sludge | Crude oil-contaminated soil | 98.3% | 30 days | The addition of nutrient and inoculum would be the best option for hydrocarbon biodegradation. | 53 |
| Sugar cane bagasse, empty fruit bunch of Oil palm tree | Petroleum hydrocarbon crude oil | 100%, 97% | 20 days | There was significant increase in the rate of biodegradation of petroleum hydrocarbon using the above supplements which stimulate bacteria growth and metabolism. | 54 |

the rate of biodegradation was observed when compared to the treatments where biosurfactant and fertilizer were combined.61

Biosurfactant secreting bacteria are very efficient in crude oil hydrocarbon degradation, investigating the effect of the Addition of Biosurfactant Produced by Pseudomonas sp. On Biodegradation of Crude Oil by researchers, they find out that growth of the bacterial isolate on crude oil has been associated with the production of biosurfactants, they conclude that the crude oil metabolizing bacterium is able to secrete surfactants which further enhance the hydrocarbon degradation.62 Some examples of biosurfactants of commercial and detoxification importance are; rhamnolipid, a glycolipid type biosurfactant produced by Pseudomonas aeruginosa, lipopeptide biosurfactant commonly known as surfactin produced by Bacillus subtilis, arthrofactin from Pseudomonas species, iturin and lichenysin produced by Bacillus species, mannosylerythritol lipids (MEL) from Candida, emulsan from Acinetobacter species, alasan from Acinetobacter radiosistens, serrawet-tin from Serratia species, viscosin, amphisin, putisolvin, hydrophobin, lokisin and tensin etc.63

7. Genome Sequence Basis for Crude Oil Degradation

Perusal proteomic analyses of strains have been potential tools to examine the relationship between various pathways encoded in the genome.64 This has revealed some potentially crude oil degradation endowed bacteria. These bacteria have a genome with some genes that saves as a genetic base for the production of secondary metabolites from which biosurfactants are derived. Crude oil degradation genes regulates glycolipid, thioesterases and peptide synthetases synthesis,65–67 it is well known that lipopeptides are biosynthesized through the
ribosome-independent pathway with non-ribosomal peptide synthetases (NRPSs) enzymes moderated by genes. A genus Polymorphum SL003B-26A1 endowed with a gene that coded for some vital enzymes such as ketoreductase (RhlG), 3-oxoacyl-(acyl- carrier protein) reductase and acyltransferase, phosphomannomutase (AlgC) that contribute majorly to the synthesis of glycolipids, which equipped this strain for the degradation of both saturated and unsaturated aliphatic hydrocarbon, monocyclic and polycyclic aromatic hydrocarbon, as well enhanced their perpetual adaptation in the crude oil polluted environment. The expression and regulation of these enzymes are coordinated at the transcriptional level of at least two quorum sensing system. The genetic regulated biodurfactant synthentic pathways have not been fully studied, however, according to, there are three major processes involved in biosurfactants biosynthesis which are; biosynthesis of 3-hydroxy-heneicosanoic acid from specific carbon sources through fatty acid, synthesis of hexapeptide by a series of enzymatic condensations from the N-terminal of Leu to the C-terminal of Gly; and the 3- hydroxy-heneicosanoic acid may undergo an enzymatic condensation process, being incorporated at the C- and N- terminals of the hexapeptide to produce a cyclic lipopeptide. Some of the genes that played vital roles in crude oil degradation are illustrated in table3 with their biosurfactants.

Table 3: Genes associated with crude oil degrading biosurtant synthesis

| Microbes                        | Genes                  | Biosurfactant         | References |
|---------------------------------|------------------------|-----------------------|------------|
| Bacillus subtilis               | ituD, ituA, ituB, and ituC. | Iturin                | 73,74      |
| Bacillus licheniformis          | licA, licB and licC   | Lichenyisin           | 75,76      |
| Pseudomonas sp. MIS38           | arfA, arfB, and arfC  | arthrobactin          | 77         |
| Pseudomonas sp                  | rhlA, B, R and I.     | rhamnolipid           | 70         |
| Pseudomonas fluorescens         | ViscAR and ViscBCR    | viscosin              | 78         |
| Pseudomonas syringae            | GacA/GacB and amsY    | Amphisin              | 79         |
| Pseudomonas putida              | dnaK, dnaJ and grpE, psoA, psoB and psoC | Putisolvins | 80         |
| Acinetobacter radioresistens   | AlnA, AlnB and AlnC SrfAA, SrfAB, SrfAC and SrfAD or SrfA-TE, sfp. | Alasan Surfactin | 81,82      |

This work illustrates some methods of crude oil polluted soil bioremediation, effects of PAH on both physical and chemical properties of the soil and some factors that can affect the rate of PAH biodegradation. Environmental PAH pollution has been a threat to man and his immediate surroundings; cleansing of petroleum hydrocarbon pollution has caused many countries the huge amount of capital and financial resources. Understanding the mechanism of some petroleum hydrocarbons degrading bacteria can be very useful when applied to petroleum hydrocarbons contaminated sites like soil, sewage, and water body. Some microorganisms feed on crude oil as a source of carbon and generate energy for their metabolic activities; this is possible because some microbes like some species of bacteria secrete enzymes that can degrade petroleum hydrocarbons. However, most of the tested degradation was carried out in a controlled environment, therefore the probability of recording failure might be high if the degradation is carried out in a non-control environment, because unfavourable climatic factors can alter biodegradation. Moreover, there have been none 100% recorded degradation in the experimental crude oil degradation test. Therefore, further studies need to be carried out on crude oil degradation to understand the degradation mechanisms of some bacteria. A critical study of the genetic characteristics of some already known crude oil degrading bacteria will help researchers to discover new strains with higher degradation capability. More research still needs to be carried out to determine other mechanisms that bacteria make use of to degrade crude oil in their environment.

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None.

10. Conflict of Interest
None.

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