The value of animal manure in the enhancement of bioremediation processes in petroleum hydrocarbon contaminated agricultural soils

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The environmental impact of petroleum is an inevitable consequences of the ever - increasing demand for petroleum products. The impact of oil spill on the environment has therefore raised the interest of researchers in developing techniques for cleaning up of oil in polluted environments. Physical and chemical methods have been applied but are not effective in ameliorating the impacts. Bioremediation offers a more suitable alternative since it is less expensive and can be used to achieve the selective remediation of target contaminants without incurring significant collateral damage to existing fauna and flora. Different bioremediation strategies have been used in both aquatic and terrestrial environment to successfully clean up spilled oil. In many cases the microorganisms involved require bio-stimulation to enable them metabolize the pollutants. Recent studies have shown that animal manure can be used to enhance bioremediation of oil contaminated soils. Micrococcus sp, Bacillus sp, Pseudomonas sp, Enterobacter sp, Proteus kleibsilla, Aspergillus sp, Rhizopus and Penicillium, which are capable of degrading hydrocarbon pollutants have been identified and isolated from animal manure. In a recent study the extent of pollution with spent engine oil in a mechanics village in Nigeria was assessed and the contaminated soils successfully treated with poultry, pig and cattle dung. This paper reviewed the value of animal manure in the enhancement of bioremediation processes in petroleum hydrocarbon contaminated agricultural soils.

Key words: Petroleum hydrocarbon, agricultural soils, bioremediation, animal manure

Introduction

In recent years, considerable attention has been paid to indiscriminate release of petroleum products and their negative repercussions on the environment. Indeed, petroleum is a commodity that must be transported from remote locations, where it is extracted to places where consumption occurs.

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These transportation methods may pollute the environment through operational discharges, which may results in the loss of large quantities of petroleum to the environment.

Studies of the biological effects of petroleum pollutants allow the assessment of environmental qualities (Rodriguez-ortega *et al.*, 2001) or impact of petrochemicals on organisms (Irwin *et al.*, 1997; Brogman, 2000; Rainbow, 2002). Organisms that thrive or survive in petroleum polluted sites, when identified could serve as bioindicators of the health of such sites (NRC, 1992; Udebuani and Ozoh, 2007).

The harmful effects of oil in different environments, has led to the need to develop simple adoptable remediation techniques for petroleum products polluted sites using different simple and affordable methods, which may include physical, chemical and biological processes (Okoh, 2006). Many industrial scale soil and water remediation of process as leading to eventual removal of hydrocarbon from the environment have been extensively documented. The physical methods of incineration or dig and dump in secure landfills (USEPA, 2001; ITOPF, 2006), as well as chemical method which involves the use of thermal and solvent treatment have been extensively reviewed (Rosenberg *et al.*, 1992; Lee and DeMora, 1999; Cohen *et al.*, 2001).

These methods are however expensive when contaminated areas are large (Okoh, 2006) and it may pose possible collateral destruction of the site material or its indigenous flora and fauna (Timmis and Pieper, 1999; Pye and Patrick, 1983). Bioremediation processes that employ the use of microorganisms to degrade environmental contaminants (Atlas and Cerniglia, 1995; McClay *et al.*, 2000; Boopathy, 2001; Bidwell *et al.*, 2002), have also proved effective and could be used to accomplish both effective detoxification and volume reduction. The advantage of this remediation process over physicochemical remediation method is that it is believed to be non invasive and relatively cost effective (April *et al.*, 2000).

Hydrocarbons bind strongly to surfaces including soil, thus, biotransformation and bioremediation of petroleum hydrocarbons continue to represent significant challenge to scientists. The varieties of factors which include physical conditions, the nature, concentration and ratios of various structural classes of hydrocarbon present in a polluted site may limit the biodegradation ability of microbes (Ko and Lebeault, 1999; Suenaga *et al.*, 2001; Venkateswanan and Harayama, 1995; Yuste *et al.*, 2000). Nutrient availability, especially of nitrogen and phosphorus appears to be the most common limiting factors (Pritchard *et al.*, 1992; Rosenberg *et al.*, 1992). Thus, since bioremediation of contaminated soils is adopted principally to improve the bio-physicochemical property of such soils (Bragg *et al.*, 1994).

Bioremediation processes could be enhanced either by addition of commercial microbe cultures (bio-augmentation) (Chhatre *et al.*, 1996; Komukai-Nakamura *et al.*, 1996; Venkateswanan and Harayama, 1995) or by nutrient enrichment (bio-stimulation) of the natural microbial population (Boopathy, 2001; Bidwell *et al.*, 2002).

Several laboratory and field investigation have indicated that addition of nutrients provide certain advantages over addition of microbes, except in cases where pollutant toxicity and appropriate microorganisms are lacking (Lee and Levy, 1991; Okolo *et al.*, 2005). Many published reports have shown that addition of microbes did not significantly enhance the rate of oil biodegradation over that achieved by nutrient enrichment (Fayad *et al.*, 1992; Venosa *et al.*, 1992). This for example was experienced in Exxon Valdez as reported by van Hamme *et al.* (2003) that bioaugmentation was ineffective in petroleum degradation process.

Numerous laboratory studies on the use of fertilizer to enhance oil biodegradation by naturally occurring microbes have concluded that fertilizer use has the potential as a treatment technique for removing hydrocarbon in an impacted area (Lee and Levy, 1991; Pelletier *et al.*, 2004). However, several components of fertilizer are toxic to humans and other organisms even at certain concentration (Lee and Levi, 1991). Secondly nutrient concentration can inhibit the bio-degradation activity (Challaina *et al.*, 2006). Several authors have specifically reported the negative effects of a high NPK level on the biodegradation of hydrocarbons (Oudot *et al.*, 1998; Chaineau *et al.*, 2005). According to Hoff (1991), microbes preferred to utilize organic components of the fertilizer instead of the oil.

Okolo *et al.* (2005) investigated the impact of addition of poultry manure alone to enhance bioremediation process in crude oil contaminated soil, while Ibekwe *et al.* (2006) studied the effect of organic nutrient on microbial utilization of hydrocarbons on crude oil contaminated soil. Ewulo (2005), studied the effect of poultry dung and cattle manure on chemical properties of clay and sandy clay loam soil, exposed to pollutants while Ogboghodo *et al.* (2004) established the effects of application of poultry manure to crude oil polluted soils on maize (*Zea mays*) growth and soil properties. Animal manure has been shown to be nutritionally rich in energy, protein, mineral and vitamins (Abulude *at al.*, 2003), which can help in the improvement of soil properties, especially farmlands, without any potential health risk on living biota.

This paper reviewed the value of animal manure in the enhancement of bioremediation processes in petroleum hydrocarbon contaminated agricultural soils

Remediation of environment of petroleum

The environmental impact of petroleum may be regarded as inevitable consequences of the ever - increasing demand for petroleum products. Majority of oil spills occurring and its catastrophic impact in the environment has attracted a lot of publicity and public attention (Duffy *et al.*, 1980). Colwell and Walker (1977) reported the three major oil spill, which received considerable attention to include, the Torrey Canyon spill (Smith, 1968), Santa Barbara incident (Straughan, 1971) and Florida accident (Blumer *et al.*, 1972; Blumer and Sass, 1972). Nigeria as an oil producing country is not left out. Much of oil that spill on land occur at a moderate scale, however, the primary concern is the serious long-term threat to the environment especially to groundwater quality (Udebuani and Ozoh, 2007). This is because oil spill on land is a potential source of ground water contamination (Duffy *et al.*, 1980).

The impact of oil spill on the environment has raised the interest of researchers in developing techniques used in cleaning up of oil in the polluted environment. Some of the methods developed so far in removing oil include physical methods, such as pumping and the use of heavy mechanical plant/bulldozer to remove oil from a contaminated site. Large quantities of oil could be removed using this method however, the oil removed requires disposal and the mechanical plant/bulldozer may damage the fauna and flora and the integrity of the environment. Absorption is another physical method but it is very labor intensive and generally somewhat limited in its use.

Chemical treatment methods include dispersing, herding, gelling, sinking of oil and burning of oil mass among others (Dewling and McCarthy, 1980). These methods require the use of chemicals, which may be toxic to the environment and expensive. The chemical may not be effective enough to alter or treat oil, while there is also the problem of disposal of the treated oil. Surfactants used in situations of oil contamination have stimulatory inhibitory or neutral effects on bacterial degradation of the oil components (Liu *et al.*, 1995). Due to these technical considerations, physicochemical treatment methods are not considered effective in cleaning up oil at oil-impacted sites.

Fortunately, microorganisms existing in the environment are naturally equipped for degradation or biotransformation of pollutants to avoid their accumulation to a point of being detrimental to life. Bioremediation offers a more suitable alternative to physicochemical treatment methods as it is less expensive and most importantly can be used to achieve the selective remediation of target contaminants without incurring significant collateral damage to existing fauna and flora at the contaminated site. Various workers (Odu, 1978; Ijah and Antai, 1988; Okpokwasili and Okorie, 1988; Pritchard, 1990; Lee and Levy, 1991; Ijah, 2003; Kim *et al.*, 2005; Okolo *et al.*, 2005;

Okoh, 2006) have reported application of microorganisms as effective bioremediation method.

Mechanism of bioremediation

The mechanism of biodegradation is achieved by the primary attack of oxygenase on intact hydrocarbons. This attack requires the presence of free oxygen. In the case of alkanes, monooxygenase attack results in the production of alcohol. Most microorganisms attack alkanes terminally, whereas some perform sub – terminal oxidation (Okoh, 2006). The alcohol produced will be oxidized finally to aldehyde and then to a fatty acid. The fatty acid is degraded further to beta – oxidation (Atlas and Bartha, 1992).

Successful applications of bioremediation methods require knowledge of the characteristics of the site and the parameters that affect the microbial degradation of pollutants (Sabata *et al.*, 2004). However, certain environmental factors limit biodegradation ability of microorganisms. Those limiting factors include petroleum hydrocarbon composition (PHC), physical state, weathering, water potential, temperature, mineral nutrient, reactions and microorganisms (absence and low number) (Bartha, 1986). Studies have shown that the availability of these environmental factors enhanced microbial activities and yielded encouraging result in both *in situ* and *ex situ* experiments of bioremediation (Cooney, 1984; Choi *et al.*, 2002; Pelletier, 2004; Kim *et al.*, 2005; Okolo *et al.*, 2005).

Bioremediation strategies

Bioremediation strategies have always been used in both aquatic and terrestrial environment to successfully clean up oil in an impacted area. This strategy has been promoted over other methods (physical/chemical) for clean up because of the reduced risk of environmental impact (Wrabel and Peckol, 2000; Tsutsumi *et al.*, 2000). These strategies include the use of indigenous microbial populations in the remediation of contaminated sites. This involves the utilization of already existing microorganisms in a given environment to achieve a successful remediation of a contaminated site. It has been established that these indigenous microorganisms are ideal candidates for use in the bioremediation of hydrocarbon pollutants (Kumar *et al.*, 1995). The presence of large number of microorganism is an advantage at the start of the process. Thus, several studies have reported the use of micro-organisms inhabitant in an environment to address the issue of dwindling terrestrial and aquatic environment (Kanaly *et al.*, 2000; Kasai *et al.*, 2001).

Another important strategy of bioremediation is bio-stimulation, which involves supplementing the contaminant soil to change the physical state of the contaminant, thereby converting it to a more bio-available form (Boopathy, 2001). The microorganisms involved may require supplementing the bio-stimulation conditions. This enables them to metabolize the pollutant. The bio-stimulation conditions include oxygen level, temperature, pH, presence of water, soil moisture, number and type of organisms present and the presence of heavy metals and salts (Rubio *et al.*, 1986; Wilderer *et al.*, 1987). Petroleum degradation by microorganisms can occur in an aerobic or anaerobic condition (Zengler *et al.*, 1999). However, the rate of degradation is faster in aerobic than in anaerobic condition.

Temperature is another factor that plays important role in biodegradation of petroleum hydrocarbon, firstly, by its direct effect on the chemistry of the pollutant and secondly, by its effect on the physiology and diversity of microorganisms (Okoh, 2006). Again, nutrient addition in the form of nitrogen and phosphate containing fertilizer and inorganic manure greatly increased the degradation of petroleum contaminants (Choi *et al.*, 2002; Kim *et al.*, 2005; Okolo *et al.*, 2005). However, it has been reported that excessive nutrient concentration can inhibit the biodegradation activity (Challaina *et al.*, 2006). Several researchers have reported the negative effects of a high NPK level on the biodegradation of hydrocarbon (Oudot *et al.*, 1998: Chaineau *et al.*, 2005). Large numbers of microorganisms, especially hydrocarbon degrading organisms in the soil will clearly be an advantage for the degradation process.

Soil pH will also affect both the growth and the solubility of compounds. A slight alkaline pH of seawater seems to be favorable for petroleum hydrocarbon degradation but acidic soil liming to pH 7.8 to 8.0 had a definite stimulatory effect (Okoh, 2006). In certain cases, hydrocarbon contamination may be associated with high level of heavy metals, which may inhibit microbial growth depending on the concentration and type of metals. Several works have been carried out on the effectiveness of the use of these environmental factors to enhance the degradation of contaminated sites (Jobson *et al.*, 1974; Choi *et al.*, 2002; Kim *et al.*, 2005; Okolo *et al.*, 2005) and the results obtained so far are encouraging.

Bio-augmentation is another strategy of the bioremediation process. This involves the addition to the soil of some selected non – indigenous microbial population to speed up degradation (Brodkorb and Legge, 1992; Boopathy, 2001). Bacteria are not only the microorganism used as they can grow under low water condition as well and are present in the soil and water. It has been reported that this technique has the advantage of introducing naturally

developed populations cultured outside the soil. This technique has been shown to enhance the degradation of pentachlorophenol, atrazine and chlorobenzene (Armstrong *et al.*, 1995).

A number of fungi inocular have been used to bio-augment soil contaminated with PCP and this removed 80–90% within four weeks. Fungi species that have been used to bio-augment soil include: Methylosinus trichosporium, and Cladophialophora sp, strain T1 (degrading BTEX) (Venosa *et al.*, 1992). However, some researchers argue that bio-augmentation can only be effective in the laboratory but not in the field. Lee and Levy (1991) reported that addition of microbe did not increase biodegradation because foreign strains of bacteria failed to compete with the indigenous population.

Several investigations have indicated that addition of commercial microbial cultures did not significantly enhance the rate of oil biodegradation over that achieved by nutrient enrichment (Fayad *et al.*, 1992; Prince *et al.*, 1999; Venosa *et al.*, 1992). Some of the possible factors responsible for bio-augmentation failures include the fact that the concentration of contaminants may not be sufficient to support growth; environment may contain substances that inhibit growth, predation by protozoa and that the introduced microbe may not be able to penetrate the soil to reach the contaminant.

More recently, bio-augmentation has had more success using activated soil rather than pure culture. The activated soils are those soils containing indigenous microbial populations recently exposed to the contaminants. This technique has the advantage of introducing naturally developed population not cultured outside the soil.

Phyto-remediation is another strategy for bioremediation. This involves the use of plants to extract or detoxify pollutants through physical, chemical and biological processes (Cunningham and Ow, 1999; Saxena *et al.*, 1999; Wenzel *et al.*, 1999; Udebuani and Ozoh, 2007). The use of plants for bioremediation is a welcome phenomena and it has the advantage of providing aesthetically pleasing ecological options. It has also minimal disruption of the top soil and it can offer the possibility of recovery of metals. Phyto-remediation is inexpensive and very effective with low levels of mixed contaminants.

A plant for phyto-remediation should have the following qualities: It must be able to grow rapidly and produce high amount of biomass. It must be able to tolerate and accumulate high concentrations of pollutants even in the harvestable part of the plant (root, shoot and leaves). Some plants have been reported to be used in phyto-remediation. Such plants include: Dictyledon (*Thlaspi caerulescons, Brassica junica*), Grasses (*Vetiveria zizaniodes*), Fern (*Pteris vittata*) and some aquatic plants (*Azolla pinnata*) (Shrimp *et al.*, 1993; Davis *et al.*, 1998; Schnoor *et al.*, 1995; Erickson *et al.*, 1994) and *Elicine* *Indica* (Udebuani and Ozoh, 2007). Phyto-remediation can be divided into a number of processes, which may include phyto-extraction (phyto-accumulation); the removal of metals from the soil and their storage in the plant. Examples of such plant include vetiver and Bahia grasses.

Phyto-degradation is another strategy that involves the uptake and degradation of organic compound. Phyto-volatilization on the other hand involves the volatilization of pollutant into the atmosphere. Example of such plants includes Indian mustard (*Brassica Juncea*) (Kumar *et al.*, 1995). Phyto-stabilization is the transformation of specie of molecule into less toxic specie (Cr^{6+} or Cr^{3+}) and involves plants such as *Zolium perenne*.

Bioremediation techniques

The different bioremediation methods employed at each point in time will depend on the degree of saturation and aeration of the contaminated area (Figure 1). The techniques involved are *in situ* and *ex situ* bioremediation technique. *In situ* techniques are defined as those that are applied to soil and ground water at the site with minimal disturbance, while *ex situ* technique are those that are treated on or off site, when the contaminated material is excavated or pumped out (Vidali, 2001). The *in situ* and *ex situ* processes are outlined in the figure I to included bioventing, biosparging, biostimulation, phytoremediation.



Fig. 1. Bioremediation Techniques (Source: Vidali, 2001)

In situ method: This involves direct approach to the microbial degradation of pollutants at the site of pollution (soil or ground water).

Addition of adequate quantities of nutrients at the site promotes microbial growth. The growth of the microorganisms and their ability to bring about biodegradation are dependent on the supply of essential nutrients (nitrogen and phosphorus). The *in situ* methods are generally the most desirable options used in the treatment of contaminated soil. They include bioventing, which is the most common *in situ* treatment. It involves supplying air and nutrient through wells to the contaminated soil to stimulate the indigenous bacteria. The increased supply of air will probably increase the rate of natural degradation (Vidali, 2001). Bioventing is one of the methods that have been shown to be effective for removal of simple hydrocarbons and it can also be used where the contamination is deep under the surface.

Another *in situ* method that is also good is biosparging. This process involves the injection of air under pressure below the water table to increase the biological activities of the soil. This method is also very effective and can enhance the rate of biological degradation of contaminants by naturally occurring bacteria (Satyanarayana, 2005).

Land farming is another *in situ* method that involves mixing of the soil by ploughing or some form of mechanical tilling. Ploughing helps to increase the oxygen level in the soil and distributes the contaminants more evenly (Satyanarayana, 2005).

Bioaugmentation/stimulation is the addition of nutrients into contaminated soil, well below the surface to stimulate the indigenous microbial population. This method has been used severally to remove or clean up oil in an impacted area (Pritchard and Costa, 1991; Bragg *et al.*, 1994). This technique is used for the bioremediation of subsurface of soil, buildings and roadways that are polluted.

Ex-situ method: The contaminated material in question could be collected from polluted site and the bioremediation with the requisite microorganisms carried out at a designated place. This process is certainly an improvement over *in situ* bioremediation and has been successfully used at some places (Satyanarayana, 2005). *Ex-situ* is often regarded as a more rapid method of decontaminating the area. The techniques involved in this approach and can be used effectively in land farming (on or offsite), compositing, biopiles and bioreactor (Vidali, 2001).

The two bioremediation techniques (*in situ* and *ex situ*) are very good methods of cleaning up oil, however, *in situ* techniques are generally considered the most desirable option for removing or cleaning oil. This is because of the lower cost and less disturbance to the environment as it provides treatment at the site, avoiding damage to the environment through excavation and transport of contaminants.

Constraints to the biodegradation process

Environmental impacts of oil spill are highly diverse. They may or may not have obvious direct effects on living organisms, but can change the physical environment in such a way as to make conditions less suitable for life or unsuitable for communities present in the ecosystem at that time. Microbial degradation therefore provides an effective and economic means of disposing of such environmental pollutants. Several microorganisms, which can degrade large numbers of pollutant, have been identified in nature (Bossert and Bartha 1984; Britto *et al.*, 2006). However, there are some limitations in the degradation process.

The limitations include the fact that microbial degradation of pollutants is generally a very slow process (Vidali, 2001). No single microorganism can degrade all the xenobiotics present in environmental pollutants. A cocktail or synergetic action of microorganisms is therefore more effective in the degradation process (Adams and Jackson, 1996; Rahman *et al.*, 2002). The pollutant may inhibit the growth of microorganisms, for example, presence of halogen in aromatic compound inhibited microbial degradation. Again, certain xenobiotics get absorbed on the particular matter of soil and become unavailable for microbial degradation (Okoh, 2001).

The need to address all the limitations and carry out ideal process of bioremediation is a step in the right direction. There is therefore the need to develop cheap, simple and adoptable technologies for the remediation of oil impacted sites at local levels. Some attempts have been made using locally available products to enhance bioremediation and results obtained so far have been impressive (Okolo *et al.*, 2005; Ibekwe *et al.*, 2006; Odjegba and Sadiq, 2007). The use of compost, Lime coal fly ash (Kumpiene *et al.*, 2007), organic manure (Okolo *et al.*, 2005), ash (Odjegba, 2007) have also being reported.

Cheap and adoptable bioremediation methods need to be developed in the areas of environmental biotechnology, as these areas will help to open new doors to the approach of bioremediation. Areas, like the examination of microbial communities capable of degrading contaminant without cultivation, using universal primer 16SrRNA gene primer (Borneman *et al.*, 1996) have been explored. In addition, creation and transfer of new strains of microorganisms into another microorganism that can simultaneously degrade pollutants have raised the hopes of researchers in achieving an effective method of bioremediation. These new strains though created are not in regular use and so more research is being conducted to determine the merits and demerits of the use of such strains.

The use of animal manure in remediation of hydrocarbon contaminated soils

Animals used as domestic livestock have helped to improve the human standard of living; however, they generate a lot of waste in the form of manure, which are very useful in their various aspects. Manure as an organic matter contains nitrogen, phosphorus and potassium among many other nutrients. These nutrients are very important for plant growth. Magnesium and sulphur, which are essential nutrients, are also found in animal manure (www.ecochem.com/t_manurefert.html).

Previous studies have reported the characteristics of animal manure to depend on the specie, age, nutrition, production diet of the animal, amount and type of bedding, time of the year and manure storage practice (Kirchmann and Witter, 1992; Van Horn *et al.*, 1994; Wilkson *et al.*, 1992). Nitrogen content in manure varies with the type of animal and feed ratio of such animal.

Poultry dropping, for instance, has the highest rate of ammonium so it is expected to have high nitrogen volatilization rate. Therefore, the total nitrogen content of applied manure is lower because of losses, which occur during storage. Concentrations of nutrients depend on the amount of dry matter present. Research have shown that when manure is fresh, it has around 70 – 85% of moisture content, but when it is air – dried, it could reduce to 9 - 15% of moisture content. It has also been shown that nutrients in dry manure are more concentrated on a weight and volume basis than in that fresh one, due to structural changes in the manure (www.ecochem.com/t_manure_fert.html).

The content of animal manure is a function of the type of animal, digestibility of the feed, microbes and its residues from the digestive system. It is known that feed digestibility increases the nitrogen content of its waste, especially in ruminants. Decrease in plant nitrogen has been reported in dairy cattle manure. Research has shown that pig's diet ensures that its manure has the highest nutritional quality (Hamilton, 1995).

The chemical composition of pig manure is responsible for the odor, which the manure gives off and it all depends on the amount of amino acids containing sulphur (Cysteine, cystine or methionine) (Hamilton, 1995). Animal manure is used for different purposes, it has been found to be effective in maintenance of adequate supply of organic matter in soil (SOM). Animal manure has helped in improving soil physical and chemical conditions and enhanced crop performance (Powel *et al.*, 1998; Ikpe and Powel, 2002).

Addition of animal manure increases soil organic matter (SOM), soil aggregate, stability, water holding capacity, water infiltration and hydraulic condition.

However, animal manure has been reported to help in enriching the soil contaminated with hydrocarbon pollutants. Okoh (2006) reported that the 1945

organic manure binds rapidly to the soil particle, and this facilitates the movement of the pollutants through dirt, when natural events like rain occur. In recent studies, animal manure has been used to enhance biodegradation of contaminated soil. Okolo *et al.* (2005), used poultry manure to enhance crude oil degradation in a sandy loamy soil. Concluded that poultry and cattle manure improved soil chemical properties irrespective of soil texture. Odjegba (2007) also observed that soil amendment with manure and wood ash reduced the bioavailability of cadmium, copper and zinc uptake by *M. hybridus*. Soil amendment actively increased water holding capacity and cation exchange capacity of substrates, providing a slow release of nutrient source, complex toxic metals and boost microbial activity (Tordoff *et al.*, 2000).

Microorganisms capable of degrading hydrocarbon pollutants have been identified and isolated from animal manure. These organisms include; *Micrococcus sp, Bacillus sp, Pseudomonas sp, Enterobacter sp, Proteus kleibsilla, Aspergillus sp, Rhizopus and Penicillium* (Ijah and Antai, 1988). Studies have also shown that because of high concentration of ammonia in the poultry manure, the most common bacteria identified included *Bacillus sp* and *Pseudomonas sp* etc (Tiquia and Tam, 2002). These organisms have been implicated in the degradation of hydrocarbon pollutant.

Value of animal manure in remediation of oil polluted soil

The extent of pollution with spent engine oil in a mechanics village in Nigeria was assessed and an attempt made to solve the problem by treating the contaminated soils with poultry, pig and cattle dung. The study revealed that the mechanics deposited about 1.4 million liters of spent engine oil annually into the immediate vicinity of the study area. Results obtained from the analyses of the spent engine oil polluted soil, showed that the physical and chemical properties of soils were affected when compared unpolluted soil.

Heavy metals such as mercury, lead, zinc, chromium, nickel, cadmium and arsenic, which were highly toxic to human and agricultural soils, were recorded at higher levels in the spent oil polluted soils indicating that they are released into the environment through inappropriate disposal of the spent motor engine oils.

Microbial population of the polluted soils compared to the unpolluted soil was also reduced probably as a result of nutrient imbalance created by spent engine oil pollution. Thus the result of an uncontrolled and unregulated dumping of waste like spent engine oil is excessive pollution of the immediate environment.

To address the immediate problem caused by spent engine oil spillage, efficacy of three animal dung types (Cattle dung, poultry dropping and pig

manure) to enhance biodegradation of spent engine oil polluted soils were tested. These tests proved to be variably effective in the reclamation of the spent oil polluted soils. The nutrient content of the soils, which were severed due to pollution, were restored. This is because the animal dung contains high nutrient composition and so they provided the polluted soil with nutrient element, needed by both the endogenous microbes and those supplied by the different animal dung for their bioremediation activities. This also helped different microbial species found in the soils to proliferate for ultimate utilization of the spent engine oil. The study specifically showed that poultry dropping caused more enhancement than cattle dung and pig manure in the remediation process.

Nutrient supplements in the form of animal dung caused reduction in metals such as Zn, Cd, Ni, Pb, Ar, Cr, and Hg, which were made abundant in the polluted soil. There was also every evidence that cattle dung, pig manure and poultry dropping process mixed culture of petroleum degrading microbes and the addition of these animal dung increase both the population and diversity of both bacteria and fungi isolates in the polluted environment to enhance remediation.

Conclusion

Bioremediation technique employed at any point in time would usually depend on the area or environmental media that is contaminated, the properties of the compound involved, the concentration of the contaminant and the time required to complete the remediation processes. Based on these options there is the need to continue evaluating the best methods that would suit these options in remediation process.

Efforts have been made to characterize microbial community of oil prospecting sites and their response to oil pollutants For example, isolated chicken dropping micro-organisms and studied their value for oil spill remediation. There is therefore the need to isolate potential degraders and the functional genes involved in a particular degradation process (Mesearch *et al.*, 2000). This is because the ability of organisms to degrade oil and its products has been linked to their genes (Okoh, 2006). This is because the genetic information that confers on the host organisms the ability to degrade recalcitrant organic compounds not commonly found in nature exists and the use of these elements to produce new degradation pathway is a possible bio-augmentation process (Ogawa *et al.*, 2004).

Studies on the use of animal dung to remediate petroleum as well as other polluted sites have been conducted. Animal manure has been shown to be nutritionally rich in energy, protein, mineral and vitamins, which can help in 1947

the improvement of soil properties, especially pollution sites, with beneficial results.

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