A Conspectus Review Efficacy of Locally Sourced Organic Biostimulants on Enhanced Biodegradation of Hydrocarbon-Contaminated Soil

Bright Fegenyie Erenee[a], Ruhuoma Wosu Kinika[a], Chinedu Azubuike Uzor[b], Aloysius Emeka Okah[b] and Leera Solomon[b]

[a]Department of Microbiology, Faculty of Science, University of Port Harcourt, East-West Road, P.M.B. 5323, Choba, 500004 Port Harcourt, Nigeria.
[b]Department of Science Laboratory Technology, School of Science and Technology, Port Harcourt Polytechnic, Rumuola, P.M.B. 5936, Port Harcourt, Nigeria.
fegenyie@yahoo.com

Abstract: A conspectus review on the efficacy of locally sourced organic biostimulants on enhanced biodegradation of hydrocarbons in contaminated soil was undertaken. Due to the consequences of environmental pollution, the remediation of the environment becomes indispensable; this can be achieved through bioremediation by biostimulation. Biostimulation is a viable option since it is cost effective, accessible and ecologically friendly. Hence, there is need for a suitable material that can be used to provide a pool of nutrients for optimum microbial growth, consequently, leading to the removal of hydrocarbon contaminants from the environment. This review focuses on the efficacy of commonly sourced organic biostimulants that has been used in the bioremediation of hydrocarbon contaminated soil. This is also aimed at developing appropriate strategy and efforts directed towards eventual manipulation of the processes of remediation, all geared towards making bioremediation technically and economically viable for comprehensive treatment of petroleum hydrocarbon contaminated soils. It was found out that most of the bioremediation studies carried out were *ex-situ* studies. Also, the removal of the contaminant was not very effective when a single organic material is used. Therefore, it is recommended that the use of any organic material as an amendment to contaminated soil should be done in combination with other organic materials for its effectiveness. Also, Sewage should be treated before it application as amendment in any *in-situ* bioremediation study due to it concomitant health and environmental concern.
[Erenee, B. F., R. Wosu Kinika, C. A. Uzor, A.E. Okah and L. Solomon. **A Conspectus Review on Efficacy of Locally Sourced Organic Biostimulants on Enhanced Biodegradation of Hydrocarbon Contaminated Soil.** *Rep Opinion* 2017;9(4):62-69]. ISSN 1553-9873 (print); ISSN 2375-7205 (online). http://www.sciencepub.net/report. 7. doi:10.7537/marsroj090417.07.

Key words: Environmental pollution, biostimulation, bioremediation, organic biostimulants.

1. Introduction

The environment is constantly witnessing varying proportions of perturbations due to pollution arising from oil industry operations, bunkering, pipeline vandalization and other human activities. In Nigeria, it is a major challenge as most Niger Delta environments especially Ogoniland has been grossly polluted over the years and had been found to contain certain mutagenic and carcinogenic pollutants including benzene that has been reported to be 900 times above the World Health Organization (WHO) standards (UNEP, 2011). These contaminants have been reported to have adverse or lethal effects on the entire ecosystem if proper mitigation measures are not put in place (Solomon *et al.*, 2016). Hydrocarbon pollution has negative impact on microbial diversity and abundance (Wosu-Kinika and Odokuma 2016). The adverse effects of environmental pollution on vital components of the ecosystems could be mitigated by a number of options which includes physical, chemical, biological and/or a combination of methods (Solomon *et al.*, 2016b).

1.1 Bioremediation

Bioremediation is a biological method involving the use of microorganisms (bacteria and fungi) and their metabolic processes (biodegradation, bioaccumulation, biotransformation, bioabsorption, bioadsorption) to remove pollutants from the environment. Biodegradation is a natural process in which organic compounds or contaminants are broken down, over time, by microorganisms that then produce simple substances such as carbon dioxide and water, thereby growing in the process producing cell material (biomass). Fig. 1 shows a diagrammatic presentation of biodegradation of petroleum hydrocarbons.

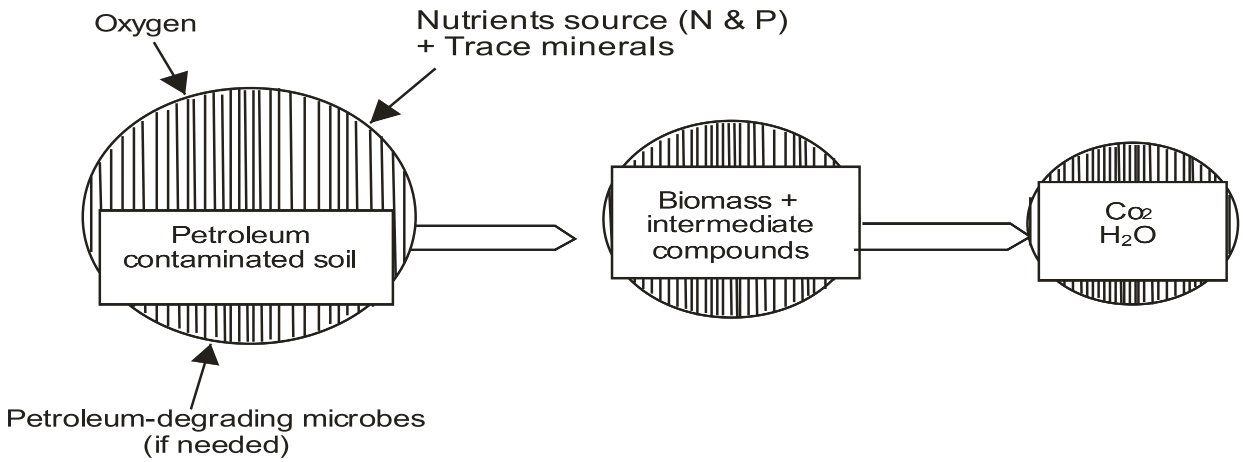

Fig. 1: A schematic diagram of enhanced biodegradation of petroleum hydrocarbons

Enhanced biodegradation ultimately result in bioremediation, as the petroleum hydrocarbons are broken down (degraded) to carbon dioxide and water (Okpokwasili, 2006). The advantages of bioremediation options include the use of inexpensive materials, environmentally friendly nature of the process and simplicity (Odokuma, 2012).

Bioremediation technologies may be classified as *in-situ* or *ex-situ*. *In-situ* bioremediation involves treating the contaminated material at the site while *ex-situ* involves the removal of the contaminated material to be treated elsewhere. Bioremediation can occur on its own (natural attenuation or intrinsic bioremediation) or can be initiated or promoted (bioaugumentation, biostimulation, biofermentation, bioventing and composting). In order to promote bioremediation, there is need for the addition of suitable organic or inorganic material that will serve as a source of nutrient for optimum microbial growth.

1.2 Biostimulation

The role of microorganisms in biodegradation of hydrocarbon is vital, as such requires nutritional support for their growth which will eventually lead to pollutant removal in the polluted soil. Biostimulation involves the use of organic materials to increase the availability of nutrients necessary for optimum microbial growth and contaminants or pollutant removal in the environment. Biodegradation processes may not occur at a desired rate due to certain limiting factors such as low oxygen level or nutrient limitation. It is however important to determine these limiting factors and supply the needed materials or modify the environment for bioremediation (Odokuma, 2015).

It mainly involves the introduction of organic or inorganic components to enhance indigenous microbial growth that directly degrades the contaminants. It has been reported that biostimulation enhances the removal of total petroleum hydrocarbons from contaminated soil (Okpokwasili, 2006; Odokuma, 2012). The accelerating effect of amendments is stringer when nutrient availability is a limiting factor.

Stimulation of microorganisms by the addition of nutrients brought large quantities of carbon sources which tend to result in a rapid depletion of the available pools of major inorganic nutrients such as nitrogen and phosphorus (Sang-Hwan *et al.,* 2007). Substrates such as wood chips, wheat straw, peat, corn cobs, sawdust, a nutrient-fortified mixture of grain and sawdust, bark, rice, annual plant stems, wood, fish oil, alfalfa, spent mushroom compost, sugarcane bagasse, coffee pulp, sugar beet pulp, okra, canola meal, cyclodextrins and surfactants can be used in inoculum production both off-site or on-site, or as mixed with contaminated soils to improve the processes of degradation (Rhodes, 2014).

There is the need for suitable organic components that will be mostly cost effective, accessible and ecologically friendly and that will necessarily serve as nutrients for optimum microbial growth for microorganisms in various polluted environments. Hence, a wide range of organic materials has been applied in the bioremediation of hydrocarbon contaminated soil by different researchers within the tropics (Okerentugba *et al.,* 2015). Although there are various organic materials that can be used in a bioremediation study, only a few of these organic materials were found to be more effective during their application. For this reason, there is need for a conspectus review on the efficacy of these organic materials used in the bioremediation of contaminated environment.

This is important because of their future application in a more tactical bioremediation process.

This review is also crucial in order to promote the utilization of suitable and readily used organic wastes in soil bioremediation.

The full application of these organic wastes will surely reduce the amount of organic waste sent to landfill, thus reducing the emission of landfill gases and also provide a cheap source of organic additive for the remediation purpose. Below are the various categories of common materials of plant and animal origin that have been applied in the bioremediation of hydrocarbons contaminated soil.

1.3 Animal wastes

1.3.1 Cow dung

Gurpreet and Jagdev (2011) reported that utilizing freely available cow dung as slurry or after composting in rural areas is a cheap and effective measure to remediate the harmful pollutants such as pharmaceuticals, pesticides and petrochemicals. Cow dung slurry maintained in the ratio of 1:10 or 1:25 is able to degrade the rural, urban and hospital wastes, including oil spillage to five basic elements (Orji *et al.*, 2012).

The study further highlights the importance of cow dung isolates, both bacterial and fungal, for reducing total petroleum hydrocarbons to 0% in polluted mangrove environment. Cow dung contains diverse group of microorganisms such as *Acinetobacter*, *Bacillus*, *Pseudomonas*, *Serratia* and *Alcaligenes* spp. which makes them suitable for microbial degradation of pollutants (Adebusoye *et al.*, 2007; Akinde and Obire, 2008; Umanu *et al.*, 2013).

Cow dung amended-soil was found to have improved soil physiochemical characteristics that enabled speedy adaptation by the microorganisms to the contaminated soil (Agamuthu, 2013). In another development, cow dung has been used in combination with other organic materials such as sewage sludge, other animal wastes (poultry droppings and goat manure) and inorganic fertilizer to yield a more significant result (Orji *et al.*, 2012).

In a similar study, it was reported that contaminated soil amended with goat manure, poultry droppings, cow dung and the control sample showed 87.1%, 78.6%, 70.7% and 32.1% loss in total petroleum hydrocarbons respectively. However, contaminated soil amended with goat manure showed the highest percentage of total petroleum hydrocarbon loss. From that study, it was concluded that the ratio of biodegradation depends majorly on soil nutrient availability (Obiakalaije *et al.,* 2015).

1.3.2 Poultry droppings

The effect of poultry droppings on bioremediation of crude oil-polluted soil has been evaluated by various researchers (Williams *et al.*, 1999; Ugochukwu *et al.*, 2016). In a particular study, it was reported that poultry droppings can serve as a good remediation material in the reclamation of a crude oil-polluted lithosphere and it is also a potential source of nutrients for microbial activity and it habours microbes capable of utilizing hydrocarbons as source of carbon and energy (Ugochukwu *et al.*, 2016).

Ecotoxicity assay involving seeds of bean seed (Viciafaba) were used to evaluate the extent of contaminant removal in the amended soil with poultry droppings and a germination index of 95% was observed in the 50 % amended option only. In a similar study, the remediation of soil contaminated with petroleum compounds was found to be significantly ($P< 0.05$) enhanced when supplemented with poultry litter (pelleted or nonpelleted) in concentrations of 10% soil volume (Williams *et al.*, 1999).

According to Hamid *et al.* (2005), the addition of chicken manure as a nitrogen source may be necessary to increase microorganism populations at a hydrocarbon contaminated site. An examination of chicken-dropping for oil spill remediation was carried out (Ijah and Antai, 2003) and the results indicated that chicken droppings enhanced degradation of the crude oil in the soil environment.

The amendment raised the acidity (pH 5.7) of the crude oil-polluted soil to alkaline (pH 7.2) within 16 days. The study showed that bacteria in chicken manure ware able to break down 50 percent more crude oil than soil lacking the amedment (Bello *et al.*, 2009).

1.3.3 Rabbit manure

Although there is paucity of information on the application of rabbit droppings in the bioremediation of hydrocarbon contaminated soils, rabbit manure is found to contain dexterous bacterial community capable of degrading various xenobiotics such as polychlorinated biphenyls (PCBs), pharmaceutical compounds and other hydrocarbon derivatives (Tharakan *et al*., 2006; Zeng *et al*., 2015).

Solomon *et al.* (2015) had earlier demonstrated the positive effect of rabbit droppings in enhanced biodegradation of the heavy fractions of total petroleum hydrocarbons in a weathered crude oil-contaminated soil environment.

1.3.4 Goat manure

A study conducted on the effect of combined nutrient sources such as NPK fertilizer and goat droppings on crude oil polluted soil from Afam in Oyigbo Local Government Area of Rivers State, Nigeria when compared with known regression model, shows no significant difference ($p<0.05$) in rates of biodegradation of concentration of total hydrocarbon on the use of fertilizer (NPK) and goat droppings.

Fertilizer application encouraged plant growth better than goat dropping in uncontaminated soil, whereas goat dropping encouraged plant growth better than fertilizer in contaminated soil. It was predicted that goat droppings have a higher organic content than fertilizers and are able to sustain nutrient level of a longer period than fertilizer (Amadi and Ukpaka, 2016).

In a comparative study using biostimulating agents (cow dung, goat manure and spent fruit residues) for the bioremediation of soil artificially and naturally contaminated with fresh unused motor oil and spent motor oil, respectively. The result obtained shows that cow dung showed highest bioremediation potential and removed 73.47% of total petroleum hydrocarbons from artificially contaminated soil while spent fruit residues showed greater reduction (74.91%) of TPH from naturally contaminated soil.

First order kinetic studies revealed that cow dung and spent fruit residues was best biostimulating agent with biodegradation rate constant of 0.026 day^{-1} and 0.027 day^{-1}, respectively. The result obtained demonstrated its potential for spent motor oil bioremediation in the order of spent fruit residues >cow dung >goat manure and for the fresh unused motor oil. The microbial counts were higher in all naturally contaminated soil amended with biostimulating agent ranging between 45×10^5 cfu/g and 89×10^6 cfu/g than artificially contaminated soil amended with sorbent which ranged from 1.2×10^5 cfu/g to 31.9×10^5cfu/g (Sarang *et al*., 2013).

The potentials of *Eichhornia crassipes* (water hyacinth) and goat droppings in enhancing biodegradation of hydrocarbons in soil were investigated (Solomon *et al.*, 2015). The biostimulative effect was observed with the increase in microbial population count in contaminated soil amended with goat droppings than in contaminated soil amended with water hyacinth. This study confirms that water hyacinth and goat droppings have the potential to stimulate bioremediation of hydrocarbon polluted soil.

In general, there was a significant reduction in total petroleum hydrocarbons in the amended set-up (Funmilayo *et al*., 2016). The effect of goat droppings in a bioremediation study involving sawdust, yam peel and a mixture of cow dung, goat dung and poultry dung (alone or in combinations) was statistically not significant ($p < 0.05$), implying that goat droppings had a similar effect as other bulking agents.

The system proposed here takes advantage of the organic wastes bulking properties as well as the autochthonous microorganism and their metabolic activity to efficiently degrade petroleum hydrocarbons (Samuel and Lukuman, 2013).

1.3.5 Sewage sludge

The application of sewage and cow dung as amendments in a petroleum hydrocarbons contaminated soil yielded positive results, as the biodegradation of used lubricant in the soil were much higher than that of the control set-ups (Agamuthu *et al*., 2013). The biostimulatory effect of processed sewage sludge on engine oil contaminated soils was significant (Kamaludden *et al*., 2016).

Cow dung amended set-ups showed 94% biodegradation while sewage sludge amendment gave 82%, as compared to the control set-up (56%). The study showed that both organic matters proved to enhance the multiplication of indigenous microbes thus enabling rapid biodegradation of the contaminant in the soil. Sewage sludge is also generally rich in nitrogen, which is essential for the growth of microorganisms.

Hence, increased microbial density can accelerate contaminant removal, as microbes are the primary agents for biodegradation (Chua and Isa, 2006). In a bioremediation study involving the use of sewage sludge, soybean meal and wheat straw on oil degradation in a contaminated desert soil, the addition of these agents have been shown to enrich soils with nutrients, such as phosphorous and nitrogen, whose limitation is known to slow down biodegradation processes (Sumaiya and Abed, 2016).

1.4 Organic nutrients of plant origin

1.4.1 Spent mushroom compost

It was reported earlier by Buswell (1994) that spent mushroom compost has decontamination potential for land sites used for disposal of hazardous wastes. Similar studies but with diverse category of chemicals were conducted by Fermor *et al.* (2000) and reported a significant ($p<0.05$) potential of phase II compost in remediation of lands contaminated with xenobiotic pollutants like polychlorophenols, polycyclic aromatic hydrocarbons and aromatic monomers (Ahlawat and Sagar, 2007).

Spent mushroom compost has also been applied in treating organo-pollutant contaminated sites (Trejo-Hernandez *et al.,* 2001). Addition of spent mushroom compost resulted in enhanced PAH-degradation (82%) as compared to the removal by sorption on immobilized spent mushroom compost (46%). It was observed that addition of spent mushroom compost to the contaminated medium reduced the toxicity, added enzymes, microbes, and nutrients for the microorganisms involved in degradation of PAHs (Lau *et al.,* 2003). The proper understanding of limiting nutrients and mineral elements composition of spent mushroom compost is required to adjust the nutrient deficiencies of petroleum hydrocarbon polluted soil in order to drive bioremediation (Okerentugba *et al*., 2015).

1.4.2 Rice husk

Identification of total petroleum hydrocarbons and trace of heavy metals, namely; zinc, cadmium, and cobalt in crude oil contaminated soil and effect of rice husk and chicken manure in bioremediation of contaminated soil were studied. The results indicated that rice husk ultimately removed more petroleum hydrocarbons compared to chicken manure and their combinations. It was also found that the combination of rice husk and chicken manure (RC-3:1) reduced 75.8% of concentration, CR-3:1 reduced 89.6% of zinc while CM reduced 65.5% of cadmium (Adams *et al*., 2017).

1.4.3 Sawdust

Sawdust is one of the most common agro-wastes which have been successfully used for the immobilization of bacterial cells and as a stimulant for optimum microbial growth during biodegradation studies. *Arthrobacter* sp. immobilized on sawdust did not lose their enzymatic activity after 6 weeks of storage (at 25°C and 45°C) and was still able to degrade similar quantities of crude oil within reasonable time frame (Obuekwe and Al-Muttawa, 2001).

Sawdust possesses a labyrinthine structure providing very high surface area for cellular attachment. High hydrophilicity of this carrier may hamper the adsorption of oil-degrading microorganisms on the carrier. However, this difficulty may be overcome by non-toxic hydrophobic coating of sawdust (Podorozhko *et al*., 2008).

Hazaimeh *et al*. (2014) during studies on degradation of oil by a bacterial immobilized consortium, demonstrated that immobilization significantly ($p<0.05$) increased the production of biosurfactants by bacteria. This was to increase the solubility and thus the bioavailability of hydrophobic hydrocarbons.

The addition of bulking agents (e.g sawdust) to contaminated soil increased oxygen diffusion and mineral nutrient availability as well as carbon source quality and physical support surface for bacterial adsorption and improves soil physicochemical characteristics as to speed up microbial adaptation and selection (Molina-Barahona *et al*., 2004).

High dose nutrient amendment can accelerate the initial oil degradation rate and may shorten the period to clean up contaminated environments and combination of two animal dung wastes and as well as combination of animal dung wastes and plant sawdust residue, organic wastes has a relative higher biostimulation efficiency in the biodegradation of petroleum hydrocarbons (Ahmed, 2015).

1.4.4 Brewery spent grain

Brewery spent grain (BSG) is a common type of industrial by-product from brewery processes all over the world in both developed and developing countries. Other by-products of the brewery process including spent hops, sludge and yeast are all regarded as wastes (Levic *et al*., 2006). BSG is the most abundant brewery by-product, corresponding to about 85% of the total by-products produced from brewing industries (Tang *et al*., 2009). Due to its high nutritional value for the growth of microorganisms, it has been used as amendment for most *ex-situ* and *in-situ* bioremediation studies for hydrocarbon contaminated soil. *Ex-situ* bioremediation of soil contaminated with 5% and 15% (w/w) used lubricating oil and amended with 10% brewery spent grain (BSG), banana skin (BS), and spent mushroom compost (SMC) was studied for a period of 84 days, under laboratory conditions.

Results obtained from first-order kinetic model that was used to determine the rate of biodegradation of used lubricating oil revealed that BSG amended soil recorded the highest rate of oil biodegradation (0.4361 day^{-1}) in 5% oil pollution, while BS amended soil recorded highest rate of biodegradation (0.0556 day^{-1}) in 15% oil pollution (Abioye *et al*., 2012).

1.4.5 Nipa palm (*Nypa fruticans*) Ash

The efficacy of Nipa palm ash (NPA) on enhanced *ex-situ* bioremediation of a crude oil contaminated soil (COCS) in Ogoniland, Nigeria has been investigated (Solomon *et al.,* 2016b). Result obtained showed that NPA significantly ($p<0.05$) enhanced total petroleum hydrocarbon (TPH) removal from crude oil-polluted soil, indicating its potential as a viable organic biostimulants for enhanced bioremediation.

1.5 Summary of findings

The locally sourced organic substrates discussed in this review included those of animal wastes such as cow dung, sewage, goat manure and rabbit manure, poultry droppings and those of plant origin which include spent mushroom compost, rice husk, sawdust, and brewery spent grain and *Nypa fruticans* Ash.

These organic substrates are reported to be significantly viable during their application, as they provide a pool of nutrients for optimum microbial growth and subsequent biodegradation of the hydrocarbon contaminants. This can also be attributed to their bulking properties as well as the autochthonous microorganism metabolic activity to efficiently degrade petroleum hydrocarbons. Also, the following were noted in course of this review; the removal of the contaminant in soil was observed from a reduction in TPH or through ecotoxicity (Phytotoxicity test); removal of the contaminant was very effective when a combination of the organic substrates was used and most of the bioremediation studies carried out were *ex-situ* studies.

The application of sewage sludge as a nutrient source in any *in-situ* bioremediation process requires meticulous approach due to the concomitant risk of spreading a high load of pathogenic microorganisms.

In essence, safety, public health and environmental concerns limit the discharge of sewage sludge into oceans or landfills and related legislations are anticipated to become more stringent over time. Hence it's subjected to treatment before it eventual usage.

1.6 Conclusion

Enhanced bioremediation seeks to develop and apply a planned strategy that removes, destroys or otherwise reduce the availability of contaminants in the environment and their adverse effects to plants and animals within a short period of time. Hence the introduction of eco-friendly biostimulants of plants and animal origin will ultimately facilitate the bioremediation of hydrocarbons process at a reduced cost.

1.7 Recommendations

Based on the findings reported in this review, it is recommended that, mitigation actions such as bioremediation by biostimulation be adopted in order to effectively remove hydrocarbons contaminants from polluted media and as well ameliorate the negative impacts of oil industry operations on the ecosystem.

The use of organic materials as amendment to enhanced biodegradation of petroleum hydrocarbons-contaminated media should be done in combination with other organic materials to further boost its effectiveness. There is need for a more robust *in-situ* bioremediation study using these organic materials. Sewage should be treated first before being applied for *in-situ* bioremediation study due to it concomitant health and environmental concern.

References

1. Abioye, O. P., P. Agamuthu and A. R. Abdul Aziz (2012). Biodegradation of Used Motor Oil in Soil Using Organic Waste Amendments. *Biotechnology Research International,* 2012(2012):1-8.
2. Adams, F.V., A. Niyomugabo and P.O. Syvester (2017). Bioremediation of Crude Oil Contaminated Soil Using Agricultural Wastes. *Procedia Manufacturing*, 7: 459-464.
3. Adebusoye, S. A., M. O. Ilori, O.O. Amund, O.D. Teniola and S.O. Olatope (2007). Microbial degradation of petroleum hydrocarbons in a polluted tropical stream. *World Journal of Microbiology Biotech.,* 23:1149–1159.
4. Agamuthu, P., Y. S. Tan and S. H. Fauziah (2013). Bioremediation of hydrocarbon contaminated soil using selected organic wastes. *In: Proceedings of environmental science and technology*, *International Symposium on Environmental Science and Technology,* 18: 694 – 702.
5. Ahlawat, O.P. and M.P. Sagar (2007). Management of Spent Mushroom Substrate. *Technical Bulletin, National Research Centre for Mushroom (Indian Council of Agricultural Research*, Chambaghat, Solan - 173 213 (HP).
6. Ahmed, A. B., N. A. Al-Mudaffar and K. H. Mahdi (2015). *Ex situ* bioremediation of soil contaminated with crude oil by use of actinomycetes consortia for process bioaugmentation. *European J. Exp. Biology*, 5(5):24-30.
7. Akinde, S. B. and O. Obire (2008). Aerobic heterotrophic bacteria and petroleum-utilizing bacteria from cow dung and poultry manure. *World J. Microbiology Biotech.* 24:1999-2002.
8. Amadi, S. A. and C. P. Ukpaka (2016). The Effect of Organic and Inorganic Fertilizer on Bioremediation of Crude Oil Polluted Land. *International Journal of Scientific Research Engineering Technology,* 2(2): 84-93.
9. Bello, M. Y., M. A. Huiwen and C. Y. Zhang (2009). Biodegradation of crude oil in soil using chicken manure**.** *International Journal of Environment and Pollution*, 36, 400-410.
10. Buswell, J. A. (1994). Potential of spent mushroom substrate for bioremediation purposes. *Compost - Science and Utilization,* 2(3): 31-36.
11. Chua, C. L. and M. H. Isa (2006). Bioremediation of oil sludge contaminated soil by co-composting with sewage sludge. *Journal of Scientific & Industrial Research*, 65: 364-369.
12. Femor, T., N. Wastls, T. Duncombe, R. Brooke, A. McCarthy, K. Semple and B. Reid (2000). Bioremediation: Use of composts and composting technologies. *Mushroom Science,* XV(II): 833-839.
13. Funmilayo, D., I. Aneyo, N. shobowale (2016). Stimulated biodegradation of waste lubicating oil in soil, using water hyacinth and goat droppings. *Journal of environmental and occupational science,* 5(2): 47-52.
14. Gurpreet, K. R. and S. K. Jagdev (2011). Bioremediation of Pharmaceuticals, Pesticides and Petrochemicals with Gomeya/Cow Dung. ISRN Pharmacology, 362459.
15. Hamid, B., D. Wiltcher and S. Diehl (2005). Bioremediation of Polychlorinated Biphenyl and Petroleum Contaminated Soil. *In: Proceedings of environmental Sci. and Technol.* II: 1-6.
16. Hazaimeh, M., S. A. Mutalib, P. S. Abdullah, W.K. Kee and S. Surif (2014). Enhanced crude oil hydrocarbon degradation by self-immobilized bacterial consortium culture on sawdust and oil palm empty fruit bunch. *Annal of Microbiology*, 64:1769–1777.
17. Ijah, U.J.J. and S.P. Antai (2003). The Potential Use of Cshicken-Drop Micro-Organisms for Oil

Spill Remediation. *The Environmentalist*, 23(1): 89-95.
18. Kamaluddeen, K. Y., B. B. Mohammed, T. Rimi, D. Yusuf (2016). Biostimulatory Effect of Processed Sewage Sludge *In:* Bioremediation Of Engine Oil Contaminated Soils. *International Journal of Scientific & Technology Research,* 5(02): 203-207.
19. Lau, K. L., Y.Y. Tsang and S. W. Chiu (2003). Use of spent mushroom compost to bioremediate PAH-contaminated samples. *Chem.* 52 (9): 1539-1546.
20. Molina-Barahona, L., R. Rodr´iguez-Vazquez, M. Hernandez-Velasco, C. Vega-Jarquin, O. Zapata-Perez, A. Mendoza-Cantu and A. Albores (2004). *Appl. Soil Ecol.* 27: 165 - 175.
21. Obiakalaije, U. M., O. A. Makinde and E. R. Amakoromo (2015). Bioremediation of Crude Oil Polluted Soil Using Animal Waste. International Journal of Environmental Science and Biodegradation, 3:3:79-85.
22. Obuekwe, C.O. and E.M. Al-Muttawa (2001). Self-immobilized bacterial cultures with potential for application as ready-to-use seeds for petroleum bioremediation. *Biotechnological Letters*, 23:1025–1032.
23. Odokuma, L. O. (2012). The Genius in the Microbes: An indispensable Tool for the Management of Xenobiotic Mediated Environmental Flux. University of Port Harcourt inaugural lecture, No. 87.
24. Odokuma, L. O., B. F. Erenee and L. Solomon (2015). Biodegradability of lubricating fluids. Paper presented at the 38th Annual General Meeting and Scientific Conference of Nigerian Society for Microbiology (NSM), Multiple Hall, University of Lagos. 1st – 5th September, (BR-O7) p219.
25. Okpokwasili, G.C. (2006). Microbes and the Environmental Challenge. Inaugural Lecture Series No.53. University of Port Harcourt Press. Port Harcourt, 31-36.
26. Orji, F. A, Ibiene A. A. and Dike E. N (2012). Laboratory scale bioremediation of petroleum hydrocarbon polluted mangrove swamp in the Niger Delta using cow dung. *Malays Journal of Microbiology* 8:219–228.
27. Okerentugba, P.O., Orji, F.A., Ibiene, A.A., and Elemo, G.N (2015). Spent mushroom compost for bioremediation of petroleum hydrocarbon polluted soil: A review. *Global Advanced Research Journal of Environmental Science and Toxicology*, 4(1): 001-007.
28. Podorozhko, E.A., V. I. Lozinsky, I.B. Ivshina, M.S. Kuyukina, A. B. Krivorutchko, J. C. Philp (2008). Hydrophobised sawdust as a carrier for immobilisation of the hydrocarbon-oxidizing bacterium Rhodococcus ruber. *Bioresource Technology,* 99:2001–2008.
29. Rhodes, C. J. (2014). Mycoremediation (bioremediation with fungi)–growing mushrooms to clean the earth. *Chemical Speciation & Bioavail.* 26:3, 196-198.
30. Levic, J., S. Sredanovic and L. Levic (2006). New feeds from brewery by-product. *Romanian Society of Biological Sciences*, 11(2):2611-2618.
31. Samuel, E. A. and L. A. Jimoda (2013). Application of Carbon-Nitrogen Supplementation from Plant and Animal Sources in In-situ Soil Bioremediation of Diesel Oil: Experimental Analysis and Kinetic Modelling. *J. Environment and Earth Science*, 3(7): 51-63.
32. Sang-Hwan, L., L. Seokho, K. Dae-Yeon and K. Jeong-gyu (2007). Degradation characteristics of waste lubricants under different nutrient conditions. *Journal of Hazardous Materials*; 143: 65 – 72.
33. Sarang, B., R. Kalia and R. Chavan (2013). Comparative Study of Bioremediation of Hydrocarbon Fuels. *International Journal of Biotechnology and Bioengineering Res.* 4(7): 677-686.
34. Solomon, L., V. Daminabo and C. A. Uzor (2016a). A Synoptic Review on Ecological Toxicology and Environmental Sustainability. *Researcher,* 8(12):6-10.
35. Solomon, L., C. Mbakwen-Aniebo and C. J. Ogugbue (2016b). Efficacy of Nipa palm (*Nypa fruticans*) ash on enhanced *ex situ* bioremediation of crude oil-contaminated soil. *In: Proceedings of the 4th ASUP/PortPoly Annual Conference*, held at Port Harcourt Polytechnic Conference Hall, Rumuola, Port Harcourt, 13th -15th September, 158-167.
36. Sumaiya, A. and M. M. R. Abed (2016). Effect of Biostimulation Using Sewage Sludge, Soybean Meal, and Wheat Straw on Oil Degradation and Bacterial Community Composition in a Contaminated Desert Soil. *Frontiers in Microbiology, 7:* 240.
37. Tang, D., G. Yin, Y. He, S. Hu, B. Li, L. Li, H. Liang and D. Borthakur (2009). Recovery of protein from brewery's spent grain by ultrafiltration. *Biochem. Engineer. Journal*, 48:1-15.
38. Tharakan, J.T., A. Addagaba and A. Shafagati (2006). Biotransformation of PCBs in Contaminated Sludge: Potential for Novel Biological Technologies. *Engineering in Life Sci.* 6(1): 43-50.
39. Trejo-Hernandez, M.R., LA. R. Opez-Munguia and Q. Ramirez (2001). Residual compost of

Agaricus bisporus as a source of crude laccase for enzymatic oxidation of phenolic compounds. *Process Biochemistry*, 36: 635–639.
40. Ugochukwu, C. O., U. O. Michael S. N. Amechi, G. A. Chikaodili, C. O. Samuel, J. A. Etim, E. O. Ifeanyi and C. A. Kingsley (2016). Effect of Chicken droppings amendment on bioremediation of crude oil-polluted soil. *European Journal of Experimental Biology*, 6(4):62-68.
41. Umanu, G., S. C. U. Nwachukwu and O. K. Olasode (2013). Effects of cow dung on microbial degradation of motor oil in lagoon water. GJBB 2:542–548.
42. UNEP (United Nations Environmental Programme) (2011). Environmental Assessment of Ogoniland, 1:65-66. www.unep.org/nigeria.
43. Williams, C. M., J. L. Grimes and R. L. Mikkelsen (1999). The Use of Poultry Litter as Co-Substrate and Source of Inorganic Nutrients and Microorganisms for the *Ex Situ* Biodegradation of Petroleum Compounds. *Poultry Science,* 78:956–964.
44. Wosu-Kinika, R. and L.O. Odokuma. (2016). Seasonal Influences on the physicochemistry and microbiology of soils in industrial areas in Port Harcourt area. *Nature and Science*, 14(1):30-39.
45. Zeng, B., S. Han, P. Wang, B. Wen, W. Jian, W. Guo, Z. Yu, D. Du, X. Fu, F. Kong, M. Yang, X. Si, J. Zhao and Y. Li (2015). The bacterial communities associated with fecal types and body weight of Rex Rabbits. *Scientific Reports,* 5: 9342.

4/22/2017