

Demonstration of Percentage Reduction of Petroleum Hydrocarbon Remediation in Soil Using *Ponderosa lemon* Leaf as Bio-Stimulant

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Abstract

The study of the performance of bio-remediant was demonstrated in terms of percentage reduction of petroleum hydrocarbon remediation in soil using *ponderosa lemon* leaf as bio-stimulant. The bio-stimulant was introduced into two environmental conditions of room-dried and sun-dried samples and the impact of the environmental factor was considered in the contributing agent on the variation in the rate of the total hydrocarbon degradation as well as the variation in the percentage of the TPH concentration. Indeed, the investigation revealed that the percentage reduction of the total petroleum hydrocarbon was more with the bio-stimulant subjected into room-dried than the sample subjected into sun-dried; and the reason for such variation was monitored as well as attributed to be the effect of environmental factors. The effect of sunlight induced the nutrient contents of plant extract, especially when they are exposed into such environmental conditions; the study further demonstrates the significance of the effect of sunlight on the total organic content, nitrogen concentration, phosphorus concentration, potassium concentration, electrical conductivity value and pH value. However, the integrity and the potential of plant extract performance for effectiveness in the bioremediation is dependent on the processing conditions in which the raw materials are subjected.

Keywords: Demonstration, percentage, reduction, TPH, remediation, soil, *Ponderosa lemon*

INTRODUCTION

The study and investigation on the degradation rate of crude oil using bitter leaf (*Vernonia amygdalina*) and manure revealed that bioremediants subjected in to room dried was more effective than sun dried sample [1]. Other investigation was made on the usefulness of *Mangifera indica* seed in bioremediation of contaminated soil environment using crude oil as the pollutant, and their research demonstrated that *Mangifera indica* seeds possess the characteristics to enhance restoration of the contaminated soil [2]. However, the application of Grape leaf remediation of contaminated soil polluted with crude oil and the characteristics of the soil after restoration has been studied by various researchers [3, 4].

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Comparison on some biostimulants, which include Mango leaf, palm kernel husk ash as well their combinations with NPK fertilizer as a proven biostimulant for bioremediation were carried out in terms of performance [5, 6]. The study showed that

the agro-wastes performed excellently, though the highest performance was recorded in the combination of the fertilizer and Mango leaf [7]. Similar comparison was reported on the research conducted on poultry manure [8], Mango leaf and NPK fertilizer, with 73 to 85% of TPH removed from the soil by the amendments reported [9]. The researchers reported a degradation efficiency of 70.7, 76.6 and 87.1% TPH degradation in soil amended by Mango leaf, poultry droppings and goat manure [10–13]. They also reported high population increase in microbial bacteria, and in the later study, characterised microbial community in refinery waste and hence, remediated the waste using nitrogen source amendment, with more than 80% of TPH removed after 90 days [14, 15].

Also, in another related study, report shows that cattle dung, goat dung, pig dung and poultry manure used for biodegradation of polycyclic aromatic hydrocarbons (PAHs) in polluted soil after 28 days increased the degradation rate of PAHs with 70.8% removal compared to only 10.1% removed from untreated soil [16–18]. Also, the growth rate of hydrocarbon degrading bacteria was far greater than the untreated soil. In considering the total petroleum hydrocarbon in soil amended by three organic waste composts, which were able to remove 60% of hydrocarbons and metals such as arsenic and chromium, was reported [19]. Equally formulated composts from fresh wastes were used to degrade petroleum hydrocarbon in acid sludge, and removed over 70% of the TPH, as reported [20].

Plant wastes have also gained wide application as treatment agent for polluted soil. One of such plant wastes that have reported as a good amendment for degradation of crude oil in soil is cassava peels [21]. Also, investigation was done on oil palm fibre dried in dark environment for remediation of crude oil polluted loamy soil [22]. Analysis on the dried oil palm fibre showed six different micro-organisms of fungi species that were isolated and identified with a population of 1.2×10^5 cfu/g, while five different bacteria species were isolated and identified with a population of 9.0×10^6 cfu/g. The total petroleum hydrocarbon removed from the loamy soil after 84 days was 76.45%, while the soil physicochemical characteristics improved by the addition of oil palm fibre treatment. Similarly, about 95.38% of the crude oil was removed from polluted soil amended by mango extract using bitter leaf extract in bioremediation of crude oil in polluted soil [23].

In another study, the concept was applied on palm oil mill effluent and goat manure to remove total petroleum hydrocarbons from polluted soil and also to evaluate the rate of CO₂ evolution in the soil [24, 25]. Results obtained showed that the physicochemical properties of the soil with treatment samples amended by goat manure were better restored than palm oil mill effluent [26, 27]. The reported percentage of TPH removed by goat manure and palm oil mill effluent were 58 and 39% respectively, while the rate of CO₂ evolution in contaminated soil treated with goat manure, palm oil mill effluent, and untreated soil were 497, 440, and 296 mg/g, respectively [28–30].

MATERIALS AND METHODS

Materials

In the investigation the following materials were used, which include soil, petroleum, plastic containers, biostimulant leaf, shovel, Gas Chromatograph, hand trowel, etc.

Collection of Sample

The soil sample was collected from Erema Town and then transported to the Department of Chemical/Petrochemical Laboratory. This was further subjected into analysis as well the petroleum sample in Rivers State University Port Harcourt. The soil and the petroleum sample were prepared ready for experiment set up.

Overview of Crude Oil Pollutions

The rate of crude oil pollution in the region of Niger Delta area of Nigeria is increasing day by day as a result of the theft and accidental discharges. So, there is need to employ the use of *Ponderosa lemon* (Grape leaf) for the treatment of contaminated soil. This research is to test the potential of the bio-

stimulant in the remediation of contaminated soil using grape leaf prepared in powder form as well as ascertaining its efficiency of both sun-dried and room dried samples.

Gas Chromatography: An Instrument Used for Crude Oil Analysis

The procedure involves an analytical concept in which samples are measured by the content of various components in it. However, analysis can be performed using gas chromatograph to determine the individual petroleum hydrocarbon concentration. The concept of the application of gas chromatography, makes use of nitrogen as a component of gas carrier in the process. The obtained sample solutions are always injected into the instrument, which allows the entering of gas stream which in turn transports the solution sample into a separation tube called the column with the constituents of Helium or nitrogen used as carrier gas. The different constituents are separated inside the column and the detector measures the quantity of components that exit the column. To measure a sample with unknown concentration, a standard sample with a known concentration is injected into the instrument. The standard sample peak retention time and area was compared to the test sample to calculate the concentration.

RESULTS AND DISCUSSION

The research illustrates the obtained results in Figure 1 as shown below.

Percentage Reduction of TPH under varying Grape Leaf Dosages at Different Time Intervals

Figure 1 shows the percentage degradation of TPH with time in a sandy loam soil treated with sun-dried and room-dried grape leaves at different weights (50 and 100 g), in both treatments. Meanwhile, there was increase in degradation of TPH in the first 7 days after contamination. TPH removed increased almost in all treatments over the period. However, analysis of TPH degradation showed that there was great increase in percentage TPH reduction when 100 g of the sun-dried grape leaves were added to the crude oil-polluted soil at 7 days after application of treatment, next was 100 g of sun-dried (GLSD), followed by 50 g of room-dried (GLRD), 50 g of sun-dried (GLSD) and the least was the control sample as presented in Appendix 1. Trend continued up to the end of the bioremediation period of 42 days as presented in Figure 1. Hence, the percentage TPH degradation in the loamy soil are obtained as 60.62, 66.21, 64.24, 97.08, and 15.61% for 50 g of GLSD, 100 g of GLSD, 50 g of GLRD, 100 g of GLRD and control samples, respectively, at the end of the 42nd day. However, it shows that bioremediation of crude oil-polluted soil using sundried and room-dried grape leaves were quite effective in reducing the level of TPH, it also revealed the bioremediation potential of 100 g of GLRD is better among treatments in the bioremediation of crude-oil polluted soil.

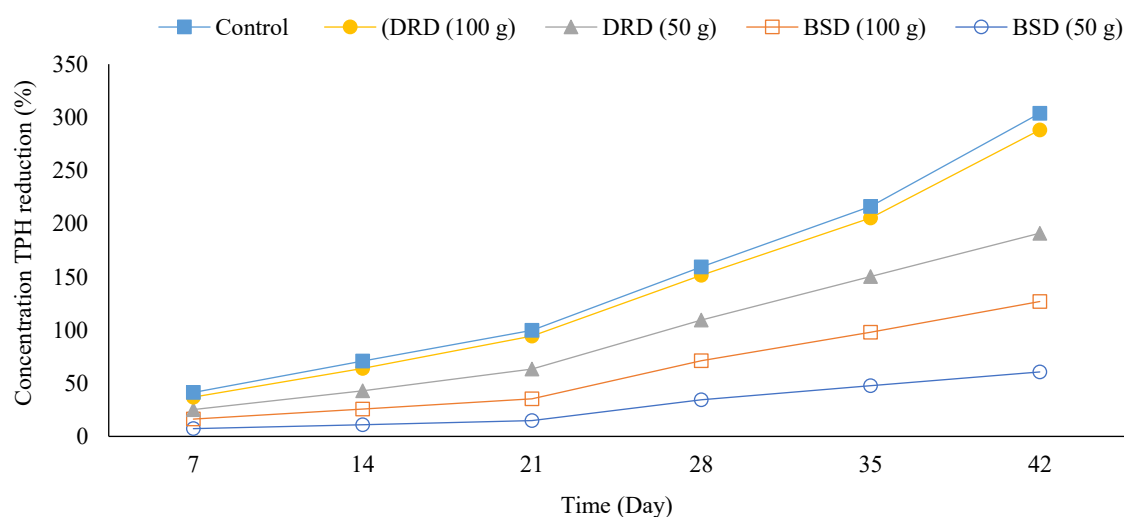


Figure 1. The representation of percentage reduction of TPH on the action of bioremediation dosage with effect of contact time (error represents percentage).

Results of Comparison of the Concentration of TPH Reduction

The degradation of TPH in soil was further studied using first order kinetic model, which has been generally used for evaluation of crude oil degradation rate in soil. The determination of the degradation rate constant within the framework of the first-order rate kinetic model involved a comparison and the regression equations presented in Figures 2–4, encompassing various treatment alternatives. Utilizing the derived rate constant, the duration required for the TPH concentration to decrease to half of its initial concentration (referred to as half-life) was subsequently assessed.

Figure 2 showcases the graphical comparison of the percentage reduction of TPH by the application of biostimulant of 50 g dosage performance compared with the control sample for the remediation of 42 days. The obtained data revealed increase in remediation in terms of percentage reduction with increase in time. The equation of linear line is obtained as: $Y=1.597x-4.312$ with coefficient of determination (R^2)=0.9937 for grape leaf room-dried (GLRD) biostimulant of 50 g; $Y=1.6184x-10.416$ with (R^2)=0.9446 for grape leaf sun-dried (GLSD) biostimulant of 50 g; $Y=0.2887x-1.3953$ with (R^2)=0.8291 for control. The relationship between treatments after 42 days of remediation shows that the correlation value at 50 g of GLRD was higher than the 50 g of GLSD while 50 g of GLSD was higher than the control sample (50 g of GLRD>50 g of GLSD>Control) as shown in Figure 2.

Figure 3 showcases the graphical comparison of the percentage reduction of TPH by the application of bio stimulant of 100 g dosage performance compared with the control sample for the remediation of 42 days. The obtained data revealed increase in remediation in terms of percentage reduction with increase in time. The equation of linear line is obtained as: $Y=2.2044x-11.022$ with $R^2=0.8907$ for GLRD (100 g); $Y=1.6714x-7.94$ with $R^2=0.9641$ for GLSD (100 g); $Y=0.2887x+1.3953$ with $R^2=0.8291$ for control. The relationship between treatments after 42 days of remediation shows that the correlation value at 100 g of GLSD was higher than the 100 g of GLRD while 100 g of GLRD was higher than the control sample (100 g of GLSD>100 g of GLRD>Control) as presented in Figure 3.

Figure 4 showcases the graphical comparison of the percentage reduction of TPH by the application of biostimulant of 50 and 100 g dosages performance compared with the control sample for the remediation of 42 days.

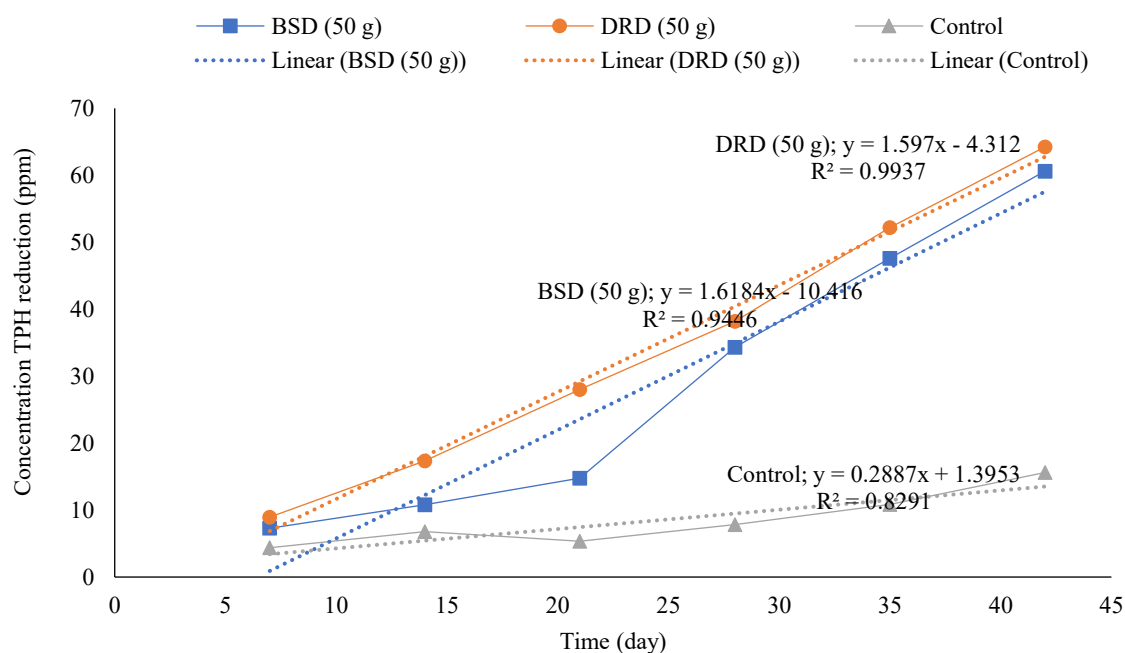


Figure 2. Comparison of percentage reduction of TPH by the action of biostimulant of 50 g dosage for both sun dried and room dried.

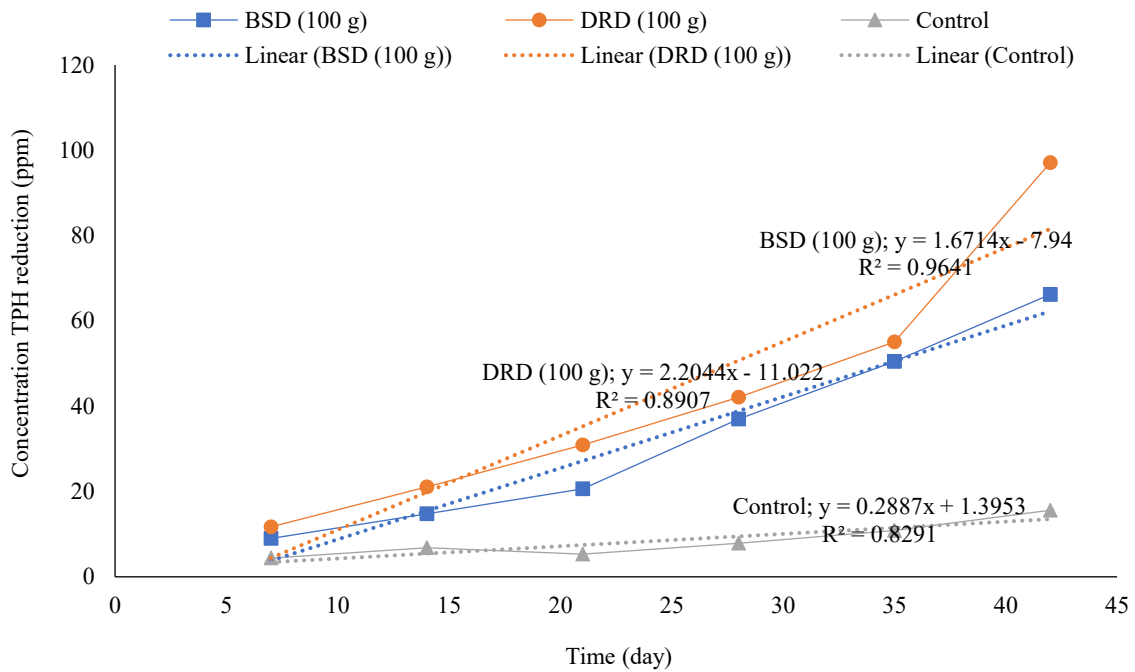


Figure 3. Comparison of percentage reduction of TPH by the action of biostimulant of 100 g dosages for both GLSD, GLRD and control.

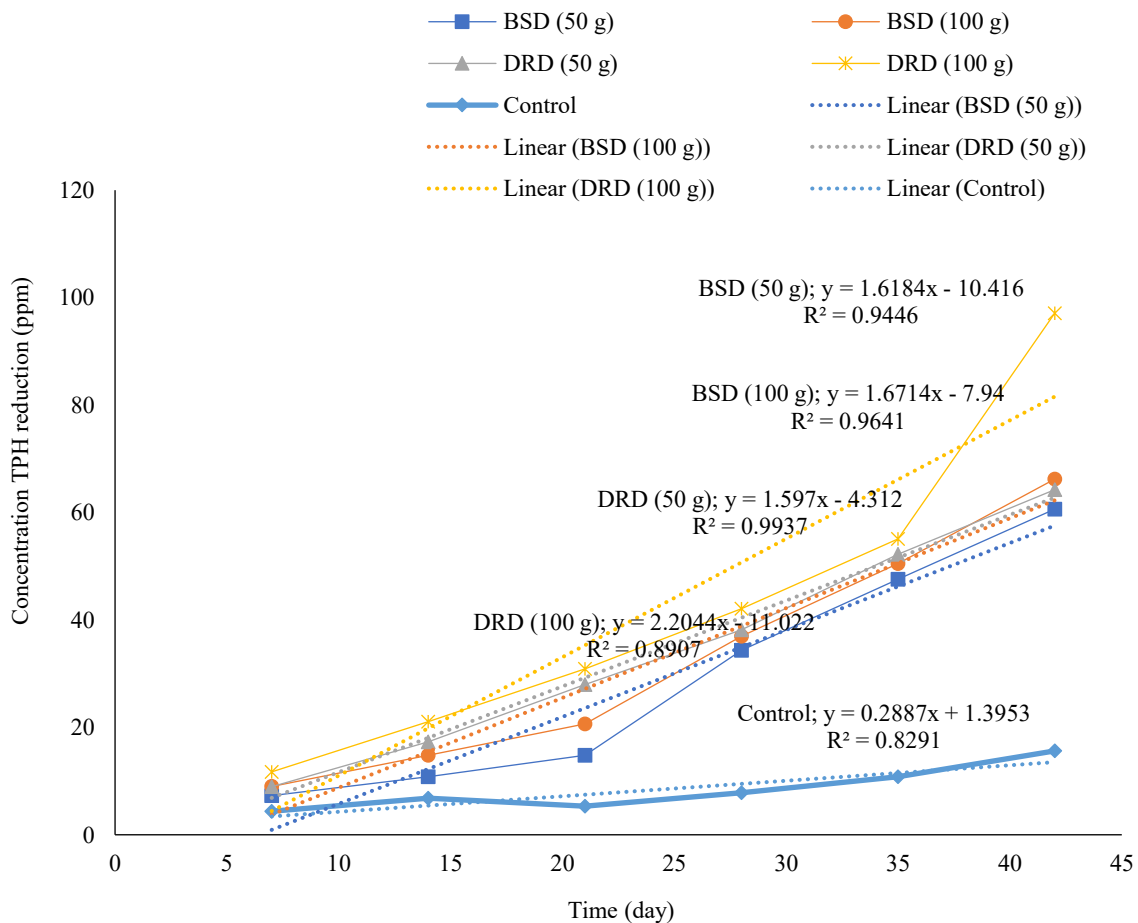


Figure 4. Comparison of percentage reduction of TPH by the action of biostimulant of 50 and 100 g dosages for both GLSD, GLRD and control.

Table 1. Experimental results compared by percentage reduction of TPH in this study.

Time/Days	TPH percentage (%)				
	<i>GLSD bacteria (50 g)</i>	<i>GLRD bacteria (50 g)</i>	<i>GLSD bacteria (100 g)</i>	<i>GLRD bacteria (100 g)</i>	<i>Control</i>
7	10	9.43	11	13.10	13.61
14	13	14.26	11	15.11	14.21
21	18	25.17	14	20.15	14.21
28	22	66.2	18	31.27	15.61
35	60.20	97.08	27	89.07	16.13
42	77.79	96.41	94.46	99.37	82.91

The obtained data revealed increase in remediation in terms of percentage reduction with increase in time. The equation of linear line is obtained as: $Y = 1.597x - 4.312$ with $(R^2) = 0.9937$ for GLRD (50 g); $Y = 1.6184x - 10.416$ with $R^2 = 0.9446$ for GLRD (50 g); $Y = 2.2044x - 11.022$ with $(R^2) = 0.8907$ for GLRD (100 g); $Y = 1.6714x - 7.94$ with $R^2 = 0.9641$ for GLSD (100 g); $Y = 0.2887x + 1.3953$ with $R^2 = 0.8291$ for control. The relationship between treatments after 42 days of remediation shows that the correlation value at 50 g of GLRD was higher than 100 g of GLSD, next was 50 g of GLSD, followed by 100 g of GLRD, which was higher than the control sample (50 g of GLRD > 100 g of GLSD > 50 g of GLSD > 100 g of GLRD > Control) as presented in Figure 4.

Validation of Results

The experimental results in this study were validated. The plots of the sun-dried bacteria and sun-dried fungi were validated against the room dried bacteria and room dried fungi respectively. Figure 4 shows the percentage reduction of TPH on the action of bio-stimulant (Grape leaf) dosage at 50 and 100 g with contact time in loamy soil environments. The results showed reasonable increase of TPH degradation at 100 g of sun-dried Grape leaf after 7 days treatment application. Figures 2–4 show the percentage reduction of TPH by which regression analysis was applied to evaluate the rate constant for Grape leaf treatment in the soil environment. The action of bio-stimulant of 50 and 100 g dosage for both sun-dried and room-dried and control was illustrated. Table 1 shows the comparison of percentage reduction on TPH with different dosages of nutrients. The percentage reduction of TPH was 99.37 and 94.46% for room dried and sun-dried at 100 g dosage, and 96.41% for room-dried and 77.79% sun-dried at 50 g dosage respectively after treatment of 42 days of remediation. The control without treatment had 82.91% of TPH reduction.

Table 1 illustrates the percentage degradation of TPH in soil environment with bio stimulant action. There are two separate dosages of bio stimulants applied in the study. It is deduced from Table 1 that as the time of remediation increased, the percentage reduction of the TPH also increased. It is observed that both 50 and 100 g Room-dried bio stimulant dosage degraded 96.41 and 99.37% while, 50 and 100 g dosage of Sun-dried bio stimulant removed 77.79 and 94.46% respectively. The percentage reduction for Room-dried bio stimulant is more effective in this study.

CONCLUSION

The following are the conclusions of this research:

1. The application of grape leaf as a bio-stimulant to enhance microbial activity in crude oil degradation or remediation yielded a positive outcome and its performance has been established in this study in terms of TPH reduction in relationship to percentage.
2. This study provides useful information to the academia and policy makers on ecofriendly conditions for bio-stimulant preparation for effective performance and this will induce the cost-effective approach for remediation of polluted site.
3. The conceptual parameters have been established in this study to be identified as contributing factors on delay of degradation of TPH in soil environment.

REFERENCES

1. Akpe AR, Ekundayo AO, Aigere SP, Okwu GI. Bacterial degradation of petroleum hydrocarbons in crude oil polluted soil amended with cassava peels. *Am J Res Commun*. 2015; 3(7): 99–118.
2. Al-Hawash AB, Dragh MA, Li S, Alhujaity A, Abbood HA, Zhang X, Ma F. Principles of microbial degradation of petroleum hydrocarbons in the environment. *Egypt J Aquat Res*. 2018; 44: 71–76.
3. Altas RM. Microbial degradation of petroleum hydrocarbon. *Journal of Environmental Perspective Microbiology*. 2015; 45(1): 180–209.
4. Imanian H, Kolahdoozan M, Zarati AR. Vertical dispersion in soil spill fate and transport models. *Journal of Hydrosience and Environment (JHE)*. 2017; 1(2): 21–33.
5. Jiang Y, Brassington KJ, Prpids G, Paton GL, Semple KT, Pollard SJ, Coulon F. Insights into the biodegradation of weathered hydrocarbons in contaminated soils by bioaugmentation and nutrient stimulation. *Chemosphere*. 2016; 161: 300–307.
6. Kang CU, Kim DH, Khan MA, Kumar R, Ji SE, Choi KW, Paeng KJ, Park S, Jeon BH. Pyrolytic remediation of crude oil-contaminated soil. *Sci Total Environ*. 2020; 713: 136498.
7. Murialdo SE, Fenoglio R, Haure PM, Gonzaler JF. Degradation of phenol and chlorophenols by mixed and pure cultures. *Water SA*. 2003; 29(4): 457–463.
8. Neha J, Wati S. Phyto-extraction potential of neem (*Azadirdehta indica*) for cd detoxification. *Climate Change and Environmental Sustainability*. 2018; 6(2): 154–159.
9. Nnaji JC. Nanomaterials for remediation of petroleum contaminated soil and water. *Umudike Journal of Engineering and Technology (UJET)*. 2017; 3(2): 23–29.
10. Obaluagbon FI. A review: aspects of the African oil palm (*Elaeisgluine esisjaeq*) and the Implications of its Bioactives in Human Health. *American Journal of Biochemistry and Molecular Biology*. 2012; 2(3): 106–119.
11. Obiakalaje UM, Makinde OA, Amakoromo ER. Bioremediation of crude oil polluted soil using animal waste. *Int J Environ Bioremediat Biodegrad*. 2015; 3(3): 79–85.
12. Ofoegbu RU, Momoh YOL, Nwoaogazie IL. Bioremediation of Crude Oil Contaminated Soil Using Organic and Inorganic Fertilizers. *J Petrol Environ Biotechnol*. 2015; 6(1): 198–203.
13. Ogu GI, Odo BB. Crude oil bioremediation efficiency of indigenous soil fungal community spiked with cassava peels in Niger Delta region, Nigeria. *International Journal of Science and Technology (IJST)*. 2015; 3(12): 19–26.
14. Ojewumi ME, Emeteri ME, Babatunde DE, Okemyi JO. In-situ bioremediation of crude petroleum oil polluted soil using mathematical experimentation. *Int J Chem Eng*. 2017; 8(4): 55–65.
15. Olajire AA, Abidemi JJ, Lateef A, Benson NU. Adsorptive desulphurization of model oil by Ag nanoparticles – modified activated carbon prepared from Brewer’s spent Grams. *J Environ Chem Eng*. 2017; 5: 147–159.
16. Olu AA. Effectiveness of organic fertilizer as a biostimulating agent for the removal of naphthalene in soil. *Appl J Environ Eng Sci*. 2017; 4(1): 1–12.
17. Salim F, Setiadi Y, Sopandie D, Yani M. Adaptation selection of plants for utilization in phytoremediation of soil contaminated by crude oil. *Hayati J Biosci*. 2020; 27(1): 45–56.
18. Sanni E, Emeteri EM. Mathematical Modelling of Insitu Bioremediation of Crude Oil Polluted Soil. *Science and Engineering Applications*. 2016; 1(4): 27–32.
19. Ugiri AC, Ukpaka CP, Ikenyiri PN. Evaluation of petroleum hydrocarbon remediation on soil environment using *Ponderosa lemon* (grape) seed. *International Journal of Chem-Informatics Research*. 2020; 6(1): 10–17.
20. Ugiri AC, Ukpaka CP, Ikenyiri PN. Evaluation of petroleum hydrocarbon remediation on soil environment using *Ponderosa lemon* (grape) seed. *International Journal of Chemical Informatics Research*. 2020; 6(1): 22–36.
21. Ukpaka CP. Development of model for bioremediation of crude oil using moringa leaf extract. *Chemistry International*. 2016; 2(1): 31–40.
22. Ukpaka CP. Remediation of crude oil in loamy soil: The integration of improved oil palm fiber (*Tekena Species*) dried in dark environment. *Discovery Science*. 2018; 14: 65–73.

23. Ukpaka CP. Comparison on the performance of moringa and neem root extracts on crude oil degradation in soils environment. *Petrol Petrochem Eng J*. 2020; 4(4): 1–9.
24. Ukpaka CP, Okirie FU. Simulation of heat generated on crude oil degradation upon the effect of temperature in bacteria growth kinetics on a bioreactor. *International Journal of Thermodynamics and Chemical Kinetics (IJTCK)*. 2022; 8(2): 39–54.
25. Ukpaka CP, Ugiri AC. Biodegradation kinetics of petroleum hydrocarbon in soil environment using *Mangifera indica* seed biomass. A mathematical approach chemical. *Chemistry International (International Scientific Organization)*. 2022; 8(2): 77–78.
26. Ukpaka CP, Ekperi NJ, Amadi SA. Development of predictive model on corrosion of metals in swampy soil environment. *International Journal of Environmental Chemistry (IJEC)*. 2020; 6(1): 15–29.
27. Vasilyeva G, Kondrashina V, Strijakova E, Ortega-Calvo JJ. Adsorptive bioremediation of soil highly contaminated with crude oil. *Sci Total Environ*. 2020; 706: 135739.
28. Wabuoma L, Ukpaka CP, Ikenyiri PN. Predictive model for petroleum hydrocarbon degradation in aqueous environment. *International Journal of Biochemistry and Biomolecules (IJBB)*. 2020; 6(1): 51–72.
29. Xiangyu H, Lili G, Zhendong C, Jianhua Y. Corrosion and protection of metal in the seawater desalination. *ESMA 2017 IOP publishing: IOP Conf Ser Earth Environ Sci*. 2018; 10(2): 122–130.
30. Yinglun S. (2018 Sep 26). Mango peels hold secret to clean oil pollution: Australian study. [Online]. *Asia & Pacific, News Letter*. Retrieved from www.xinhuanet.com.

APPENDIX

Computation of Percentage Removal

Appendix 1: Comparison of percentage reduction of TPH on the action of biostimulant dosage.

Time/Day	GLSD (50 g)	GLSD (100 g)	GLRD (50 g)	GLRD (100 g)	Control
0	-	-	-	-	-
7	7.29	8.99	8.95	11.71	4.38
14	10.82	14.77	17.33	21.07	6.80
21	14.77	20.66	27.99	30.89	5.35
28	34.32	36.96	38.17	42.08	7.84
35	47.59	50.47	52.21	55.08	10.83
42	60.62	66.21	64.24	97.08	15.61