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Bioremediation of Crude Oil Polluted Soil Using Agro-Wastes from Plant Sources

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Abstract

The present research was designed to evaluate the efficacy of various agro-wastes from plant sources in bioremediation of crude oil contaminated soil. The study comprised of eight experimental groups amended with different Agro-wastes namely cassava peels, coconut husk, guinea-corn chaff, different pawpaw parts and pineapple peels. Control group GR1 did not receive any treatment while Control Group GR2 received agricultural soil only. Individual treatment vessels contained 7kg of crude oil impacted soil and the entire remediation period lasted for twelve (12) weeks. TPH reduction in the impacted soil varied between 5.56 and 73.1% after twelve-weeks of applying the various amendments. Values obtained for PAHs within the experimental period revealed percentage reduction ranging from 4.91 to 76.9%. When compared with TPH reduction in the Control Group GR2, the added treatments evidently increased the rate of bioremediation of the petroleum impacted soil. This study revealed that plant parts and wastes used in this study have potentials for sustainable remediation of crude oil contaminated soil.

Index Terms - Agricultural soil, Crude oil, Bioremediation, Agro-waste, Contaminated soil, Soil amendment

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1 Introduction

Crude oil spillage is one of the major environmental pollution caused by exploration and exploitation of crude oil. Crude oil and its hydrocarbon by-products could be discharged into the environment as a result of accident that occur during shipping, onshore/offshore exploration/production and transportation of oil in pipelines. These contaminants affect the chemical composition and physical matrix of soils culminating in loss of soil fertility, change in ecosystem and displacement of communities [1], [2].

Oil spill is a major blow to our ecosystem and affects it drastically when it occurs, spreading spontaneously, destroying crops, livestock and aquacultures and also contaminating groundwater and soil. This has resulted in disappearance of rainforest which covers about 7,400 km^2 [3]. Moreso, crude oil contamination causes health challenges like difficulty in breathing and skin diseases. Sonawdekar [4] reported that the amount of natural oil spillage was estimated to be 600,000 metric tons per year with a range of uncertainty of 200,000 metric tons per year.

Bioremediation deals with biological treatment and recovery of contaminated sites during cleanup processes. It helps in degradation, alteration, removal and depollution of chemicals from nature by means of microorganisms, plants or fungi [5], [6]. During biodegradation, organic matters are broken down into small molecules [7]. Bio stimulation deals with the modification of the environment by introducing special nutrients such as nitrogen and phosphorus, oxygen and other electron acceptors to increase rate of contaminant degradations under indigenous bacteria [8].

Compared with physical and chemical repair technologies, bioremediation has been widely used because of its advantages of good effect, easy operation, low cost, rapid degradation rate, lack of secondary pollution and *in-situ* biodegradation of oil fractions by nutrient amendment [9], [10].

Bioremediation is divided into two types of remediation: *in-situ* and *ex-situ*. *In-situ* remediation techniques include land tillage, microorganism addition, bio-culture, and bio-ventilation. *Ex-situ* remediation technologies include bioreactors and prefabricated beds. The essence of bioremediation technology is the degradation of pollutants through microbial metabolic activities [11], [12].

Agbim [13] and Mbagwu [14] reported on the effectiveness of cassava peels and poultry droppings in enhancing the degradation of crude oil-polluted soil in South-Eastern Nigeria. A greenhouse study aimed at determining the potentials of poultry droppings (PD) and cassava peels (CP) for nutrient enhanced biodegradation of petroleum hydrocarbon (THC) reported that for soils treated with NPK fertilizer, the percentage THC degradation was 40% and 41% for 50 tons NPK and 100 tons NPK fertilizers respectively, while for soils treated with CP and PD, the percentage THC degradation was 26% and 31% for 25 tons CP + 25 tons PD and 50 tons CP + 50 tons PD respectively [15].

Degradation of the soil contaminants have been aided by Biostimulation which provides nutrients and suitable physiological conditions for the growth of indigenous microbial populations, thus increasing metabolic activity. This technique has been widely used in reclaiming oil-polluted soil [16]. Investigation on the population and types of saprophytic and crude oil-degrading fungi from cow dung and poultry droppings were carried out by Obire [17] and it was reported that the addition of cow dung or poultry droppings to polluted soils is highly beneficial since it enhances the proliferation of mycoflora that may be suppressed by addition of crude oil to the soil. The enhancement of microbial utilization of crude oil was further assessed with a suggestion that the addition of organic nutrients (poultry manure) to a contaminated soil causes biodegradation of crude oil-polluted soil [16]. Hwang *et al.* [18] investigated the bioremediation of hydrocarbon-contaminated soil using composting process and found out that mixing of remediated soil with contaminated soil increased the effectiveness of composting [16]. This is because the recycled soils usually have acclimated microorganisms that can significantly affect the degradation rate of contaminants. Also, the use of rice husks for the removal of dyes, heavy metals and some other chemicals has been studied. Their application to absorb lead, copper, cadmium and mercury has also been reported [19], [20], [21].

Soil structure, which is the form of soil particles assemblage, determines the ability of the soil to transmit air, water, and nutrients to the bioactivity zone. Vinas *et al.* [22] attributed a remarkable shift in the composition of bacterial community to both the biodegradation processes and the addition of nutrient. The addition of nutrient to an ecosystem may result in a selective increase in microorganisms capable of utilizing the hydrocarbons [23].

This study exploits the potentials for use plant-derived materials such pawpaw parts - leaves, fruit and root), guinea corn chaff, pineapple peels, coconut husk and cassava peels for treatment of crude oil contaminated soils by enhancing microbial population and nutrient composition of the polluted soil.

2. MATERIALS AND METHODS

Materials and methods

Agricultural soil sample

Agricultural soil (sand – 70%, clay – 9%, silt – 21%, Total Organic Carbon – 3.77g/kg, pH 7.9, and Total Hydrocarbon Content – 122mg/kg) used for bulking was collected from the Garden in the Faculty of Agriculture, University of Port Harcourt. The soil sample was air dried for 7 days and passed through 10mm sieve. The soil samples were passed through 1.7mm sieve. This experiment was performed under a roof cover.

Crude oil impacted soil

For each experimental group, Bonny light crude oil was used to contaminate 7kg of soil sample to achieve 10% pollution level. The contaminated soil sample was allowed to stay one week for microorganisms to acclimatize before it was distributed into groups accordingly. The experiments lasted for 12 weeks. Mixing and watering were repeated every three days.

Preparation of the Agro wastes

Agro wastes were obtained from various locations: Cassava peels and different parts of pawpaw plant were obtained from Alakahia in Port Harcourt, Rivers State, Nigeria; Coconut husk was obtained from Rumokoro market in Port Harcourt, Rivers State, Nigeria; Guinea corn chaff and pineapple peels were obtained from Choba market in Obio/Akpor Local Government Area of Rivers State, Nigeria. The agro wastes were air dried to obtain dry substrates and later milled into semi-fine particles using corn mill Px 2200 China. Coconut husk used was sun dried, cut into bits, and mashed with mortar and pestle to soften them and later ground into semi-fine particles. The ground sample was sieved with a 2mm sieve into a powdered form. Oven drying of agro-wastes were avoided in order to prevent denaturation of some active ingredients that might be present.

Determination of Extractable Total Petroleum Hydrocarbon (ETPH)

Extractable Total Petroleum Hydrocarbon was determined using the USEPA 8015 method described by US EPA [24].

Determination of Extractable Polycyclic Aromatic Hydrocarbons (PAHs)

PAHs were measured using the Microwave extraction – EPA method 3546 described by US EPA [24].

Determination of Heavy Metals Concentration in Soil Samples

Heavy metals were determined by Atomic Absorption Spectrometry method described by Haswell [25].

Determination of Soil pH

pH of experimental samples was determined by the ASTM D4972 method described by ASTM [26].

Determination of Soil Moisture Content

Moisture content of the soil samples was determined by a method described by The Texas Department of Transport (TxDOT Designation: Tex-103-E) [27].

Statistical analysis

Results in this study are expressed as mean± standard deviation. Statistical analysis was carried out using analysis of variance (ANOVA). Data between groups were analyzed using SPSS®: Version 16.0. $P<0.05$ versus respective initial value was taken as significant.

3. RESULTS

The various groups involved in the study are defined as in Table I. Effects of the various amendments on Total Petroleum Hydrocarbon (TPH) are shown in Figure 1. There were significant ($p<0.05$) decreases in TPH in the groups treated with Guinea corn chaff, Pawpaw parts, Pineapple peels as well as the group treated with a combination of cassava peels, coconut husk, guinea corn chaff, pawpaw substrate and pineapple peels at weeks 8 and week 12. Control GR2 that received no form of amendment showed non-significant changes in Total Petroleum Hydrocarbon at the 8th and 12th weeks respectively.

While Figure 1a shows TPH Concentrations in the soil samples at weeks 8 and week 12, Figure 1b shows percentage reduction in TPH in Experimental and control groups. The results showing Polycyclic Aromatic Hydrocarbon (PAH) in Experimental and Control groups are presented in Figure II. Week 8 recorded significant reduction in the PAH concentrations in the groups treated with pawpaw parts, pineapple peels and a combination of all the amendments in this study.

Also, week 12 recorded significant reduction in the PAH concentrations in all the treated groups. Figures 2a & Figure 2b show PAH Concentrations in the soil samples at weeks 8 and week 12, and percentage reduction in PAH respectively. Figure 3 shows pH levels in the treatment and control groups while results showing moisture content in Experimental and Control groups are presented in Table 2.

4. DISCUSSION

The result in Figures 1(a & b) & 2 (a & b) shows TPH and PAH levels in the test soils and the percentage Hydrocarbon degraded respectively, with the highest reduction observed in the group treated with a pineapple peals, followed by the group treated with a combination of plant materials; 73.1% and 72.1% percentage reduction were recorded for both groups respectively. PAHs decreased from week 1 to week 12 in the treated groups; the highest reduction in PAHs was observed in soil samples treated with a combination of agro wastes which recorded 76.9% reduction; this was closely followed by the group treated with pineapple peels in which PAHs were reduced by 70%. These results showed a remarkable significant

Table 1 Experimental Groups in the study

Group	Soil quantity & status	Amendment received
GR1	1kg unpolluted soil	No treatment
GR2	1kg polluted soil	No treatment
GR3	1kg polluted soil	0.1kg of prepared cassava peels.
GR4	1kg polluted soil	0.1kg of prepared coconut husk
GR5	1kg polluted soil	0.1kg of prepared guinea corn chaff
GR6	1kg polluted soil	0.1kg of prepared pawpaw substrate
GR7	1kg polluted soil	0.1kg of prepared pineapple peels
GR8	1kg polluted soil	0.02kg each of plant materials (cassava peels, coconut husk, guinea corn chaff, pawpaw substrate, pineapple peels)

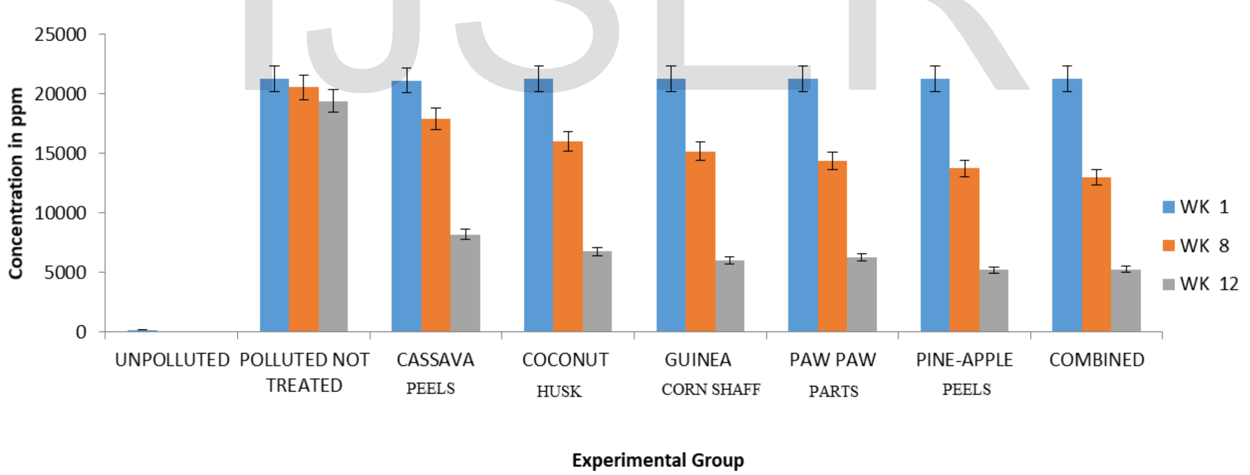


Fig 1a. Total Petroleum Hydrocarbon (TPH) Concentrations of the soil samples

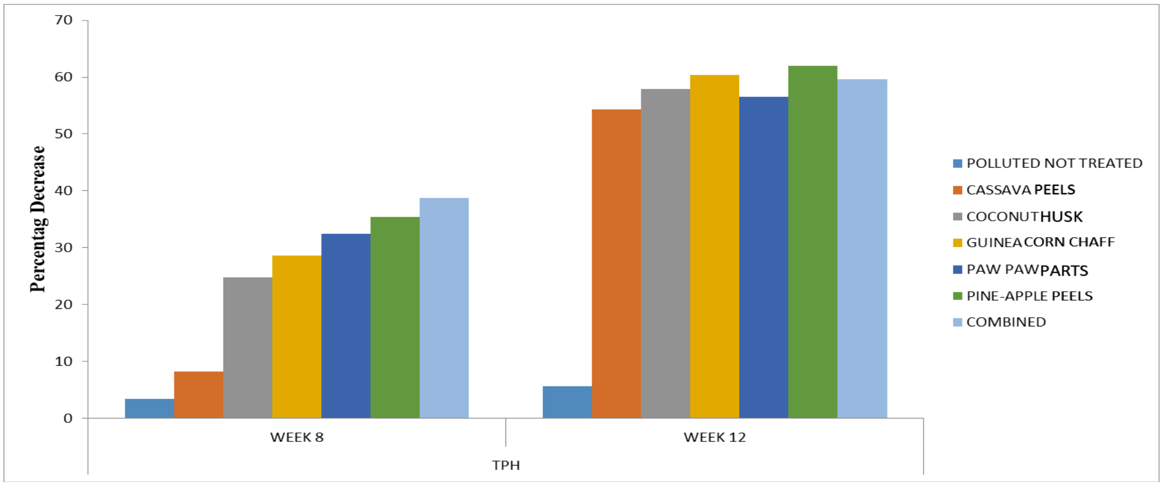


Fig 1b. Percentage reduction of Total Petroleum Hydrocarbon (TPH) Concentrations of the soil samples

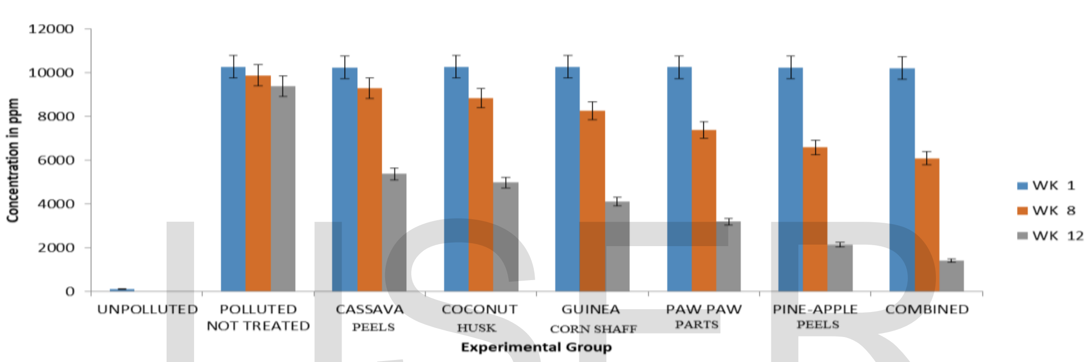
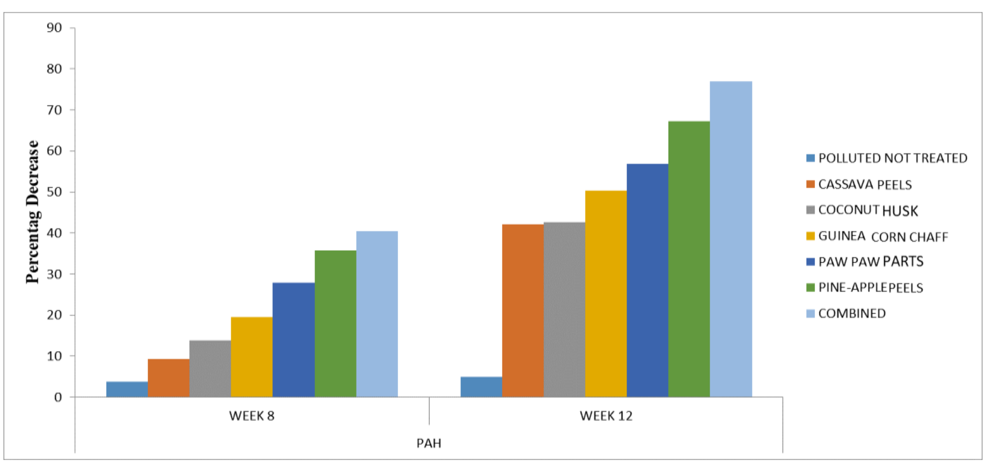


Fig 2b. Percentage reduction of Polycyclic Aromatic Hydrocarbons (PAHS) Concentrations of the soil samples

Fig 2a. Polycyclic Aromatic Hydrocarbons (PAHS) Concentrations of the soil samples



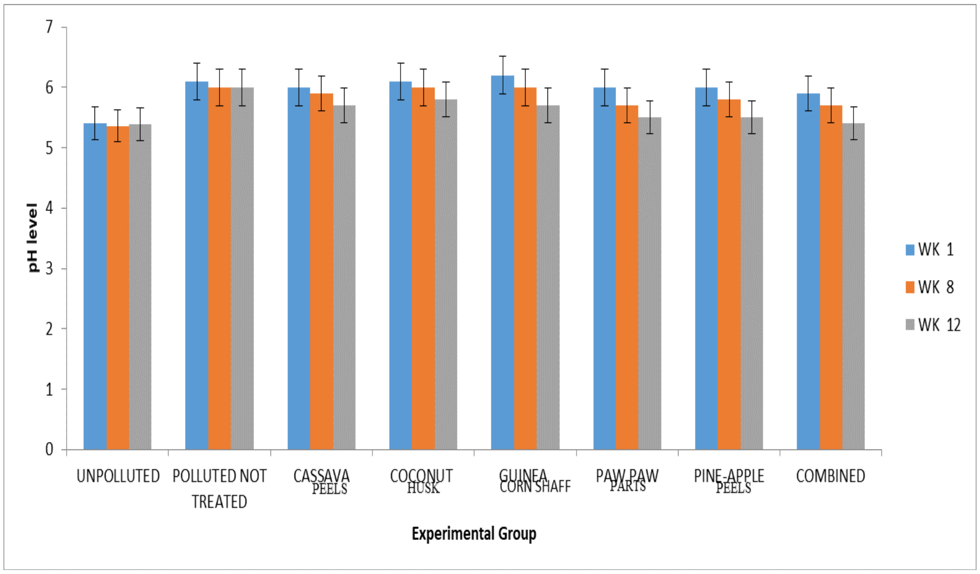


Fig 3 pH level of the soil samples

Table 2 Moisture content of experimental group

Experimental Group	*Moisture Content*		
	WEEK1	WEEK 8	WEEK 12
GR1	16.2	-	-
GR2	17.5	17.1	17.0
GR3	17.2	17.0	16.5
GR4	17.5	17.1	16.6
GR5	17.6	17.2	16.8
GR6	17.2	16.9	16.3
GR7	17.3	17.0	16.5
GR8	17.0	16.3	16.2

decrease in TPH and PAHs content of the treated crude oil polluted soils as compared to the untreated polluted soil. Findings in this study of hydrocarbon loss due to agro waste amendment is in line with previous research carried out by Lee and Trembley [28] and Obasi *et al*. [29] which revealed that agro waste amendments enhance biodegradation by supplying nutrient to the microbial community, thereby increasing the microbial count with increasing degradation over time. The low percentage of crude oil degraded in the control samples indicates the possibility of natural degradation happening though slowly and this corroborates a previous study by Onuoha [30] in which non-treated crude oil polluted soil samples recorded insignificant TPH and PAHs reduction.

In the present study, there were little or no changes in pH and moisture content of both the unpolluted, polluted but not treated and polluted & treated soils. Overall, pH in the experimental group treated with pineapple peels and the group treated with a combination of all the plant materials both increased to 5.2 at week 9. Majority of soil microorganisms thrive best in the pH range of 6 to 8 and adjustment of pH could double the rates of biodegradation. The import of this is that the lower pH observed in the groups treated with cassava peels and pawpaw parts might be responsible for the lower level of TPH reduction recorded for both groups. Physiochemical characteristic of soil is an indication of its health. A healthy soil is required to have normal pH range (close to neutrality), high nutrient (nitrate, potassium, phosphate, etc.) contents that are required for normal plant growth [31].

5. CONCLUSION

Findings from this study indicate that plant parts and wastes have potentials for bioremediation of crude oil polluted soils especially when the various agro materials studied are combined to achieve synergistic effects of bio augmentation and bio stimulation.

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