

Bioremediation of Hydrocarbon Polluted Soil in the Lowland Forest Ecosystem in the Niger Delta through Enhanced Natural Attenuation Process (ENAP)

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Abstract

The Niger Delta region has witnessed environmental pollution arising from oil activities. Soil fertility in the Lowland Forest Ecological Zone was investigated to establish the consequences of oil pollution on the soil and remedial actions to resuscitate the soil. A suitable cost effective and environmentally friendly technology to handle the pollutions in the Niger Delta region can be found in Remediation by Enhanced Natural Attenuation Process (ENAP) which facilitates the activities of microorganisms to biodegrade the hydrocarbon impacted soil. The study compared the bio-physicochemical parameters of the oil spill polluted soil with particular reference to areas of oil production of the Lowland Forest Ecological Systems with those of the unaffected soil as well as their response to bioremediation interventions using ENAP. The results demonstrated significant decrease in the values of the key indicator parameter, the Total Petroleum Hydrocarbons (TPH). But for the unenhanced process, the result showed low level of reduction of TPH values for the polluted soils. A degradation trend was demonstrated with time leading to significant TPH reductions and improved key soil fertility indices. The result showed that the level of the nutrient status of soil in the region can be improved through the natural attenuation process.

Key Words:bioremediation, hydrocarbon, soil, lowland, ecosystem, natural, attenuation

1. Introduction

The incidence of oil spillage constitutes serious soil degradation in the Niger Delta. The area currently faces series of ecosystem depletion as most soil flora and fauna are destroyed. Oil spills from the activities in the oil industry in the region affect the environment in the operational areas, right of ways (ROW) and third party areas. These result from equipment failures, leaks from corroded equipment and vandalisation (sabotage). The spilled crude oil from the source, through a plausible transport mechanism and exposure pathway, gets to the receptors - soil, vegetation, surface and ground water, marine environment, animals and humans - and pollute the environmental media. Soil fertility, measured by physical, chemical and biological parameters, is adversely affected.

The impacts include loss in the productive capacity of soil, with implications on living organisms and economically on the people in the polluted area, and consequently high poverty rate and unemployment. This study examined the application of enhanced natural attenuation process in the remediation of oil polluted soil in the Lowland Forest Ecosystem in the Niger Delta. It examined the effect of oil pollution on soil fertility in the region, examined the relationship between soil temperature and oil pollutants in the polluted soil in the study area, determined the effect of enhanced natural process of bioremediation on soil fertility improvement and compared the differences between the enhanced and unenhanced remediation in the area.

2. Literature Review

A number of researches have been carried out on hydrocarbon polluted soil. The studies include Abii and Nwosu (2009) study on the effects of oil spillage on soil and Aghalino (2000) on the negative impact of oil activities on wild life, soil, air, water and the ecosystem of communities.

The ecological effects include brownish vegetation and soil erosion, diminishing resources of the natural ecosystem, fertile land turned barren and adverse effect on the life, health and economy of the people. In Amadi and Ue Bari (1992) study in the rainforest ecosystem in Nigeria, soil and microbiological properties were evaluated 17 years after oil spillage to assess the effects of oil and interrelationship between the hydrocarbon utilizing and nitrifying micro-organisms. The study showed that organic carbon, total nitrogen, carbon/nitrogen ratio, available phosphate and exchangeable potassium were high at moderate and high impacted zones. Also the distribution of aerobic petroleum hydrocarbon utilizing fungi and bacteria showed a lesser condition at the moderately impacted zone than at the highly impacted zones.

The effect of crude oil pollution on soil fertility and the growth of plants and uptake of nutrients were investigated by Agbogidi, Eruotor and Akparabi (2007) by growing corn on a soil polluted by crude oil. The soil was analyzed for organic carbon, total and available nitrogen, extractable phosphate, and exchangeable potassium, calcium, iron and manganese after each cropping. It was observed that germination and yields were drastically reduced as the level of pollution increased. At 4.2 percent crude oil pollution level, the average reductions were 50 percent in germination and 92 percent in yield. The amount of organic carbon, total nitrogen, extractable phosphate, and exchangeable potassium, iron and manganese increased in the soil with level of crude oil addition, while extractable phosphate and exchangeable calcium were reduced. The poor growth was attributed to suffocation of plants caused by exclusion of air by oil and exhaustion of oxygen by increased microbial activity, interference with plant-soil-water relationships and toxicity from sulfides and excess manganese produced during the decomposition of the hydrocarbons.

Wokocha, Emeodu and Ihenko (2011) examined the impact of crude oil spillage on soil properties and food production in Ogba/Egbema/Ndoni Area in Rivers State, Nigeria. The results showed that the pH status of soil in heavily contaminated and moderately contaminated zones varied from acidic (pH 4.0) to neutral (pH 6.0). The chemical properties of soil indicated that percentage organic matter increased from 1.34 to 2.62, available phosphorus decreased from 15 ppm in control to between 7.34 and 5.42 in soil polluted with high level of crude oil. The result was in line with Amadi and Ue Bari (1994), and Ogboghodo, Osemwota, Iruaga and Chikor (2000). Andrade, Cavelo, Vega and Marcet (2004) in an experiment on the effect of prestige oil on salt marsh soils in the coast of Galicia (Northern Spain) revealed that oil pollution altered both physical and chemical soil properties, lowered porosity, and increased resistance to penetration and hydrophobicity. The crude oil spillage affected the physical, chemical and biological properties of soil, resulting in low food production by reducing the nutrients availability in the soils through increased soil acidity and toxicity of crude oil fractions. The experiment on the effect of poultry manure on maize planted on crude oil polluted soils showed that percentage growth rate in plant height and yield decreased with increase in crude oil contamination (Ogboghodo, 2004).

Crude oil spillage also suppresses seed germination, regeneration and caused cellular and stomata abnormalities (Gill and Sandota, 1976). Ekundayo, Emede and Osayande (2001) confirmed that in crude oil polluted soils, possibility of grain yield is significantly reduced by 95 percent compared with the control. In a study of agricultural land in an oil producing area around Qua Iboe River in the Eastern Niger Delta, the fouled loamy soil samples polluted by crude oil were treated using chemical degreasers and detergents (Essien, et al 2010). The result of the treatments showed a significant effect on soil properties and crop growth parameters; however recovery level was significantly higher than the level of degradation, except in infiltration rate. Soil pH increased by 26% in fouled soil, attributed to bacterial biodegradation of crude oil under the anaerobic conditions present in the soil macro and micro-pores, and indicated the tendency of crude oil spills to buffer acidic soil to neutral. Hydraulic conductivity with 45 – 67% reduction from 82.24 cm/day in the control soil to 39.6 cm/day in polluted soil confirmed the blockage of polluted soils micropores by oil films. Crop growth, indicated by root elongation, diminished to 7.4 ± 0.64 cm in polluted soil compared to 13.47 ± 6.40 cm in the control soil.

Oil in soil reduces essential nutrients available for plant and crop utilization (Abii and Nwosu, 2009). In Obire and Akinde (2006) the effect of various concentrations of crude oil on fungal populations of soil was investigated for a period of 18 weeks using standard methods.

Total fungal counts ranged from 26 to 143×10^2 cfu/g soil and counts of petroleum-utilizing fungi ranged from 2 to 102×10^2 cfu/g soil. Analysis of variance and randomized complete block design of counts of total fungi and petroleum-utilizers showed high significant difference between the control and the oil treated soils. Counts of petroleum-utilizers expressed as a percentage of the corresponding total fungal count of the soils ranged from 4.7 % to 58.8 %.

The result showed that an addition of crude oil concentrations > 3 % to soils resulted in a selective increase in fungal populations and a reduction of species diversity by total elimination of certain species. Since microorganisms play an essential role in biogeochemical cycling, interference with microbial metabolic activities by pollutants in the environment can have far reaching ecological consequences.

Daniel-Kalioet al(2006) studied the effect of Bonny Light Crude oil pollution on soil and successive plantings in the same soil at 4 – week intervals on the growth of dayflower (*Commelinabenghalensis*L.). The factorial sets of treatments were two levels of oil pollution (0 and 50 mg/g) and 5 successive plantings. Characteristics assessed were mean plant height, leaf area per plant and mean dry matter weight. At each of the 5 cropping, mean plant characters assessed were significantly higher at 0 mg/g oil pollution than at the 50 mg oil/g soil pollution level. The result of the investigation confirmed the effects of oil polluted soil on plant growth. Gill and Sandota (1976) examined the effect of crude oil on the growth and anatomical features of *Chromolaenaodorata*. In the investigations the effects of Forcados Blend crude oil on crops, three concentrations (25.0 cm^3 , 50.0 cm^3 and 75 cm^3) applied to soil were found to suppress seed germination and regeneration; and caused cellular and stomatal abnormalities on the leaves of *Chromolaenaodorata*. The result showed that oil pollution affected the rate of plant germination and growth.

Oil pollution, whether acute or chronic, has deleterious effects on agricultural lands and hence significant effects on plant growth (Agbogidiet al., 2005, 2006, 2007). Benka-Coker and Ekundayo (1995) reported that oil pollution tends to change the physical, biological and chemical properties of soil, thus affecting plant growth. Oil pollution has also been reported to create conditions in soils which make some essential mineral nutrients unavailable to plants and some non essential minerals to appear and rise to a toxic level (Siddiqui and Adams 2002).

In the study of the bioremediation of soil sediment contaminated by light Arabian crude oil, D'el Arco and Franca (1999) found a clear evidence of improved bioremediation performance in soil with indigenous bacteria and soil with added land farming bacteria. Straube et al., (1999) carried out a bench scale study to optimize bio-augmentation strategies for treatment of soils contaminated with high molecular weight polyaromatics. Trindade et al., (2004) compared bioremediation between a weathered and recently oil contaminated soil in Brazil. Although there have been reports of investigations on the use of organic nutrients such as cow dung and poultry droppings or inorganic fertilizers in bioremediation of oil polluted sites (Amadi and Ue-Bari, 1992; Obire and Akinde, 2006), there is limited investigation on the bioremediation of oil polluted soils, especially in the Niger Delta Region, using natural methods and the effects of oil pollutions on soil fertility and by implications, effect on soil productivity.

3. Study Area

The study was carried out in Goi community in Ogoniland, a community with oil exploration and production activities, in the Lowland forest ecological zone of the Niger Delta. The soil in the study area was heavily polluted by hydrocarbon arising from pipeline vandalism. The distribution of soil in the area below 2m is clayey. Typical bulk density of sandy soil in the area is $1.5 - 1.7 \text{ g/cm}^3$ with a porosity of between 0.43 and 0.36. Porosity of topsoil typically decreases as grain size increases. The soils in the areas are also largely defined by their acidity (pH 3.9 – 6.13). The acidity is normal and characteristic of the soils of a typical Niger Delta area. Generally, the nutritional status (organic matter content) of the soils was polluted by oil which affected its capacity for agricultural use and the composition, structure and density of the vegetation. Presently there exist patches of distinct secondary forests around the area namely: mixed tropical lowland rainforest, bush fallow vegetation, pipeline line vegetation and farmland/plantation vegetations. The major oil companies operating in the area are Shell Petroleum Development Company, Chevron, Agip, Finale Elf, etc. The major negative impact of their activities is oil pollution with consequences on the environment.

4. Methodology

Source of data is mainly primary source. Soil sample was collected from some selected sites in Goicommunity in Ogoni land. Secondary data were from Shell Petroleum Development Company and Agip Oil Company in Port Harcourt. Samples were collected at a site located at 3km from the centre of Goi community with coordinates 050 26' 30.7" North and 0060 48' 35.6" East: It has sandy topsoil: 10 - 20cm sandy silt, and above 20cm sandy clay. The sampling area ($20 \times 10 \text{m}^2$) was divided into 25 grid plots, each measuring $2 \times 4 \text{m}^2$. Five composite samples were obtained from each section at random at a depth of 0-15cm (topsoil) and 15-30cm (sub-soil).

The same area was measured out for the control. In each treatment plot, the soil was tilled and nutrient amendment, sodium-phosphate-potassium organic fertilizer application was done manually by sprinkling. These soils were further tilled by harrowing and then spiked with water uniformly to soften them and allow the water penetrate the soil matrix. They were tilled in a week after they were spiked. Composite samples were collected and sent to the laboratory for physiochemical and microbial evaluation. The soils were tilled again and homogenized a week after the initial tilling to uniformly distribute the petroleum contaminants and break up the soil lumps to fine particles thereby increasing the surface area. Windrows/ridges were constructed after the secondary tilling of the test site. The ridges measured about 2 feet high and 4 feet wide. The windrows were made to achieve better aeration and optimize the efficiency of the attenuated processes in action, which exposes the microorganisms to oxygen, and aids in the biodegradation process of the petroleum hydrocarbon. The windrows were broken down after standing for between 3 and 4 weeks, after construction. Water was added to the sandy soil to enhance the biodegradation of the petroleum hydrocarbons by the microorganisms when it penetrates the soil.

The samples were analyzed, for the parameters of concern - temperature, Total Petroleum Hydrocarbon (TPH), Electrical Conductivity (EC), Heterotrophic Bacterial Count, Hydrocarbon Utilising Bacteria, pH, and soil Nutrients parameters (nitrate and phosphate). The parameters were engineered and monitored in a way to create a favourable and enabling environment for the microbes to multiply and biodegrade the hydrocarbon impacted soil naturally. The process involved a stepwise monitoring and controlling of the parameters for a period of 24 weeks. Soil temperature was measured *insitu* at the site with the use of a thermometer. The pH was measured *insitu* at site with the use of a pH meter. The nitrate content was determined using titrimetric method; while Phosphate was determined by Vanodomolybdo Phosphoric acid Colorimetric method. Total Petroleum Hydrocarbon (TPH) was measured using Gas Chromatographic (GC) Analysis. Electrical Conductivity was determined electrometrically with a calibrated electrical conductivity meter. Total Heterotrophic Bacterial (THB) Count was determined using pour plate method and Total Hydrocarbon Utilising Bacteria (THUB) was determined using spread plate method. Multiple Linear Regression (MLR) was used for data analysis and test of hypothesis. MLR was used to test the relationship between temperature and soil fertility parameters.

5. Data Analysis and Discussion

Parameters that served as indicators of the levels of pollution and remediation were analysed. Temperature regime in the enhanced process varied between 26°C to 32°C , with an average temperature of 28.25°C and percentage increase of 18.75%. Temperature gradually increased from Week 1 (26°C) to Week 8 (28°C) and then to Week 24 (32°C) (Fig. 2a.). But the unenhanced process showed a variation between 33°C and 25°C , with a mean value of 28.42 ± 3.29 (Fig. 2b). Temperature difference was found to play a key role relative to the parameters that affect bioremediation and soil fertility.

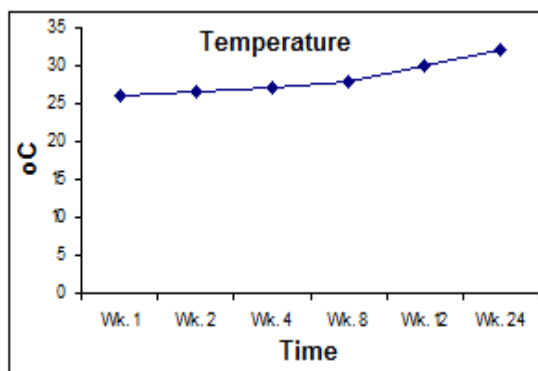


Fig. 2a: Temporal variation in Temperature in Enhanced Process

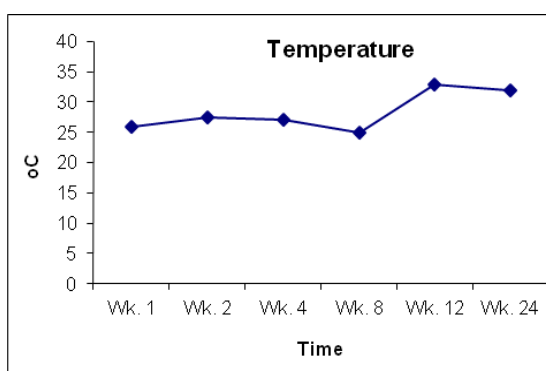


Fig. 2b: Temporal variation in Temperature in Unenhanced Process

pH in the enhanced process as observed from start of treatment in Week 1 to finish in Week 24 showed slightly acidic (6.9) in Week 1 and slightly basic (7.3) in Week 24. The mean pH value was 7.06 ± 0.21 and the percentage variation was 6.84%, while in the unenhanced process, pH varied from weak basic (7.3) to weak acidic (6.8) with percentage variation of 6.85%. Biodegradation process which involved the breaking down of hydrocarbon chains was accompanied by the release of carbon dioxide and biomass as a by-product.

The released CO_2 contributed to the alkalinity in the treatment medium. From the correlation analysis, changes in pH in the polluted soil were highly significant with a correlation variable score of 0.982. While the oil may have had some direct impact in lowering the pH, it is also possible that microbial actions through metabolic process contributed to changes in pH (Manahan, 1994).

Electrical conductivity (EC) increased from 26S/cm in Week 1 to 1.263S/cm in Week 2 in the enhanced process. Thereafter, there was a sharp decrease to 8.3S/cm in Week 4 before gradually increasing to 166S/cm in Week 24. The mean value was 337.06 ± 522.78 S/cm and the percentage decrease was 99.34%. The unenhanced process showed a peak EC at 1,263S/cm in Week 2 and minimum value at 8.3S/cm in Week 4 with percentage variation of 99.34%. EC measures the ionic concentration in the soils and is therefore related to dissolved solutes. The correlation between pH and EC was 99.905. Clustering of EC with other parameters demonstrated an affinity with an average node at 35.3%.

It was obvious that the process of tilling the soil, creation of aeration and moisturing helped in the release of dissolved solutes and hence increase in EC at start of treatment. Previous studies on EC revealed that bulk EC of sediments during microbial mineralization of diesel or crude oil (investigated in a mesoscale laboratory experiment consisting of biotic contaminated and uncontaminated columns) demonstrated variance with level of contamination (Atekwana, et al., 2004a). The numbers of degrading microorganisms increased with a clear pattern of depth zonation within the polluted column. Microbial community structure determined from ribosomal DNA intergenic spacer analysis showed a highly specialized microbial community in the unpolluted column. The polluted column showed temporal increases in bulk conductivity, dissolved inorganic carbon and calcium, suggesting that the high bulk conductivity was due to enhanced mineral weathering from microbial activity. The greatest change in bulk conductivity occurred in sediments above the water table saturated with crude oil. Variations in EC magnitude and microbial populations and their depth distribution in the polluted column are similar to field observations (Atekwana, et al., 2004b).

Nutrients were in high demand from start to end of treatment because it is a buster of the microbes which in turns eats up the hydrocarbon at a speedy rate. In the enhanced process, the maximum value for nitrate (82.4mg/l) was observed in Week 1 and the minimum (27.7mg/l) in Week 4. However, nitrate showed a pattern of increase and decrease due to supplementation and depletion by microbes. Between Week 1 and 2 there was an increase in concentration to 64.2mg/l before decreasing to 34.8mg/l in Week 4. Between Week 4 and 12, the value increased to 82.4mg/l before declining to 18.7mg/l in Week 24. The mean value was 48.88 ± 24.94 mg/l and the percentage decrease was 77.31% (9a). But the unenhanced process had nitrate variation from 82.4mg/l to 18.7mg/l with a mean value of 45.35 ± 23.93 mg/l. High nutrient removal efficiency was observed for both nitrate and phosphate across the treatment media. The high reduction in concentration of nitrate and phosphate can be attributed to uptake by the large population of microbes, adsorption by suspended solids and the adsorptive nutrient precipitation of biological de-calcinations (Lakatset al., 1997; Lau et al., 1995). However, to replenish the depleted nutrients by the microbial population in the treatment medium, organic nutrients were added. The regression analysis of the data revealed that Nitrate and Phosphate were significant. Phosphate was 99.99 % significant and Nitrate was 99.85 % significant respectively.

For the enhanced process, phosphate concentration was not significant. The maximum concentration (3.8mg/l) was observed in Week 1 and the minimum concentration (0.96 mg/l) observed in Week 4. However, from Week 4, the concentration slightly increased before decreasing to 2.9mg/l in Week 24. The mean value was 2.42 ± 1.10 mg/l and the percentage decrease was 74.74% (Fig. 3a) while for the unenhanced process, it varied from 3.8mg/l to 0.96mg/l. The mean value was 2.64 ± 1.12 (Fig 3b).

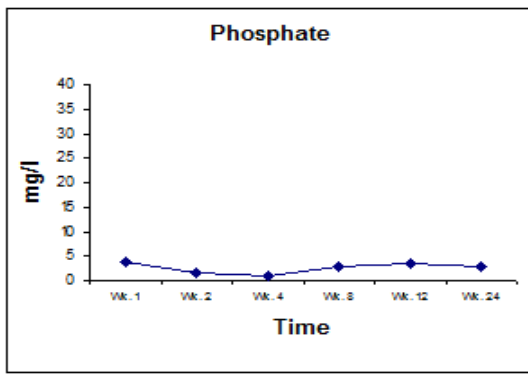


Fig.3a: Temporal variation in Phosphate Concentration in Enhanced Process

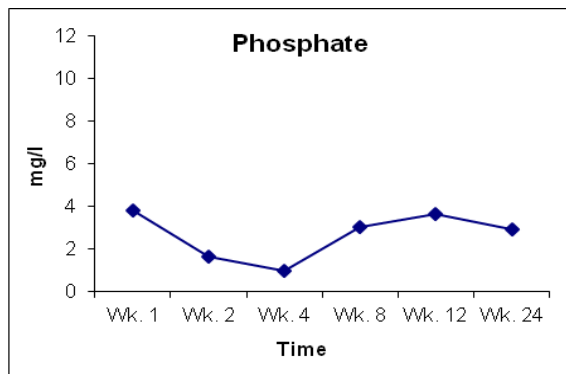


Fig. 3b: Temporal variation in Phosphate Concentration in Unenhanced Process

The TPH value for the enhanced process decreased from 8,565ppm in Week 1 to 191.39ppm in Week 24. The pattern indicated a proportional reduction in TPH value. The mean value was $2,730.1 \pm 2,391.8$ ppm and the percentage reduction was 97.76% (Fig. 4a). The observation in the unenhanced process showed very slow degradation trend between maximum value of 8,565ppm in Week 1 and 6,291 ppm as the minimum in Week 24(Fig. 4b). The percentage degradation was 26.55%. TPH responded to treatment by enhanced natural attenuation. Sharp decrease in TPH concentrations were observed as expected across the waste streams in the polluted soil. The suitability of remediation of hydrocarbon contamination by enhanced natural attenuation technique was demonstrated by the reduction in TPH concentrations in the streams.

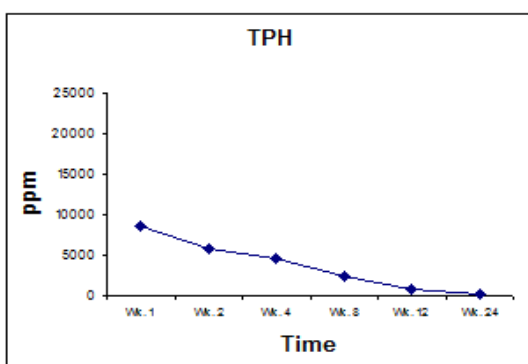


Fig. 4a: Temporal variation in TPH Concentration in Enhanced Process

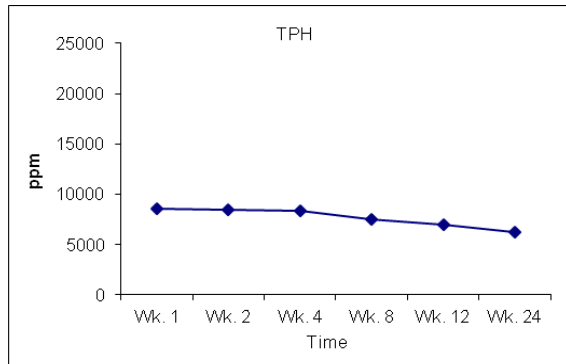


Fig. 4b: Temporal variation in TPH in Unenhanced Process

For the enhanced process, Total Heterotrophic Bacterial Count in Week 1 was 7.3×10^6 Cfug. Decrease in concentration was observed up to Week 4 (5.24×10^6 Cfug). Between Week 4 and Week 8, there was a slight increase in concentration, before decreasing to 1.4×10^6 Cfug in Week 24. The trend showed a gradual reduction from start to end of treatment, an indication of microbial depletion. The mean value was $4.94 \pm 2.06 \times 10^6$ Cfug and the percentage reduction was 80.8% (Fig. 5a). In the unenhanced process the bacterial concentration varied from 7.3×10^6 Cfug in Week 1 and 3.4×10^6 Cfug in Week 24 with percentage change of 53.42% of depletion rate (Fig. 5b). It was observed that Phosphate depletion was higher than for Nitrate. This observation corroborates with Braddock, Ruth, Walworth and McCarthy(1997) which showed that nutrient contents decreased considerably with time at the end of the first few weeks of study. However, this was at variance with the studies by Illmer and Schinner (1995) demonstrating increased Phosphate concentrations than Nitrate concentrations, possibly due to Phosphate remobilization from microbial metabolism.

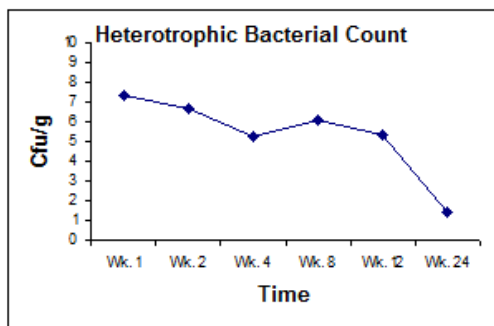


Fig. 5a: Temporal variation in Heterotrophic Bacterial Count in Enhanced Process

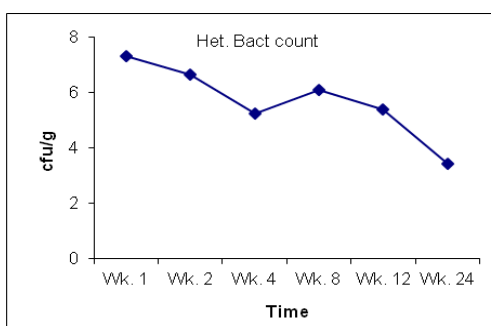


Fig. 6b: Temporal variation in Heterotrophic bacterial Count in Unenhanced Process

This reduction further established that the decrease in the number of hydrocarbon utilising Bacteria (biomass) was a direct consequence of the bioutilization of the hydrocarbons polluting the soil. The hydrocarbon content reduced as the hydrocarbon utilising bacteria decreased in population. This agrees with Braddock et al. (1997). In the enhanced process, Hydrocarbon Utilising Bacteria had concentration reduction from Week 2 (6.87×10^6 Cfu/g) to Week 4 (3.12×10^6 Cfu/g). But there was a gradual increase thereafter to Week 12 due to eating up of the hydrocarbon as substrates by bacteria leading to their multiplication. It finally decreased to 1.64×10^6 Cfu/g in Week 24. The mean value was $3.92 \pm 1.91 \times 10^6$ Cfu/g and the percentage decrease was 76.13% (Fig. 6a). But for the unenhanced process, variation was between 7.3×10^6 Cfu/g maximum value in Week 1 and minimum value of 3.4×10^6 Cfu/g in Week 24. The percentage reduction was 53.43% (Fig. 6b).

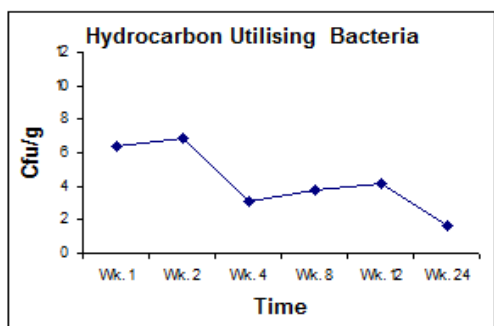


Fig. 6a Temporal variation in Hydrocarbon Utilising Bacteria in Enhanced Process

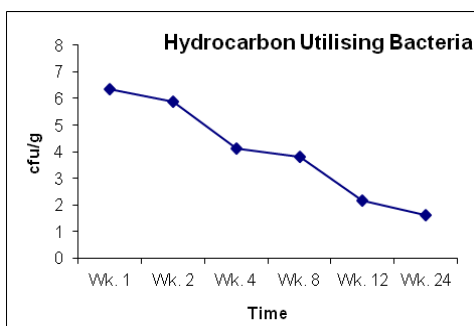


Fig. 6b: Temporal variation in Hydrocarbon Utilising Bacteria in Unenhanced Process

Total Hydrocarbon Utilising Bacteria (THUB) fluctuated from 3.8×10^6 Cfu/g in Week 1 to 5.1×10^6 Cfu/g by Week 2 in the enhanced process. Subsequently, there was gradual decrease to 2.68×10^6 Cfu/g in Week 12, before increasing to 3.24×10^6 Cfu/g in Week 24. The mean value was $3.59 \pm 0.93 \times 10^6$ Cfu/g and the percentage decrease was 47.45%. THUB reduction is an indication of TPH degradation in the enhanced treatment process (Fig. 7a). But for the unenhanced process, the concentration was 3.8×10^6 Cfu/g in Week 1. It reduced to 1.24×10^6 Cfu/g in Week 24 with percentage reduction of 67.36% (Fig. 7b).

MLR and ANOVA were used to test the hypotheses of the study. The regression analysis showed the relationships between soil temperature and oil pollutants. The result shows an R square value of 0.999 and adjusted R square of 0.997. This showed that the various parameters accounted for 99.7 percent of the change in temperature in the polluted soil. This validated the high relationship between soil temperature and oil pollutants in the study area. It presented also the correlation scores for the variables. For instance, the correlation of soil temperature with bacterial count was 0.662 significant at 0.076, soil temperature with nitrate was 0.677 significant at 0.070 and with TPH was 0.658 significant at 0.078. These results indicated high correlation between soil temperature and each of the parameters in the polluted soil. This validated the fact that there was a strong relationship between soil temperature and oil pollutants within the heavily polluted soil in the study area. ANOVA was applied to test the hypothesis which stated that there is no significant difference in the bioremediation rate between the enhanced and un-enhanced attenuation processes of the spillage polluted soil in the Niger Delta. The result is presented in the table 1.

Table 2: ANOVA result for the soil nutrients parameter between the enhanced and unenhanced remediation of polluted soils in Gio community

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	3.844	2	1.922	4.988	.111(a)
	Residual	1.156	3	.385		
	Total	5.000	5			

a Predictors: (Constant), unenhanced polluted soil nutrients, Enhanced polluted soil nutrients

b Dependent Variable: TPH

The result shows the sum of square of 3.844, at a degree of freedom of 2, mean square of 1.922 and F value = 4.988 significant at a value of 0.111. The result indicated that there was a significant increase in the healing in the enhanced process in the polluted soil compared to the unenhanced process. It states that “there is a significant difference in the bioremediation rate between the enhanced and un-enhanced attenuation processes of the spillage polluted soil”.

Other studies revealed that hydrocarbon content of 3,400 – 6,800 ppm represent a high level of hydrocarbon contamination on the site, and a review of existing data on the Niger Delta by (Onifade, 2007). Osuji (2001) and Osuji, Adesiyun and Obute(2004) affirm that such high hydrocarbon levels affect both above-ground and subterranean flora and fauna, which are essential components in the biogeochemical cycle that affects availability of plant nutrients. However, as a result of the intervention through the Enhanced Natural Attenuation Technique, with other processes such as chemical oxidation processes and metabolic activities, the treatment media in Lowland Forest Ecological System were stimulated by simple soil tilling, homogenization and application of nutrient amendment. This process led to high level of hydrocarbon loss during the first 3 weeks of treatment. This high reduction rate was achieved due to microbial population increase by nutrient augmentation.

The correlation of temperature and TPH demonstrated high significance at 99.92. The observed reduction in bacterial load (counts) may be due to exhaustion of the substrate (hydrocarbon), in the waste treatment medium. This reduction was with time, from start of treatment to finish. The other reason for the reduction may be due to reduced nutrient concentration in the treatment medium, which may have affected the microbial population and productivity. According to Oduet al. (1985), hydrocarbon-utilizing microbes such as *Azobacterspp* normally become more abundant while nitrifying bacteria such as *Nitrosomonasspp* become reduced in number. However, the heterotrophic bacterial count and hydrocarbon bacterial count were statistically significant as demonstrated by the regression analysis. The hydrocarbon utilising bacteria in the polluted soil were also statistically significant from the regression analysis with correlation at 99.82 level of significant. Past studies attribute the decrease in microbial counts with time, independent of treatment, to the decreases in the hydrocarbon concentrations. A downward trend for microbial abundance well in advance of the exhaustion of all hydrocarbons has also been noted in other studies (Allard and Neilson, 1997; Harris, Bentham and Birch, 1991; Insam and Domsch, 1988; Turco, Kennedy and. Jawson, 1994).

6. Conclusion

This study demonstrated that oil pollution reduces soil fertility and productivity in the Lowland Rainforest Ecosystem in the Niger Delta Region and that oil polluted sites could be resuscitated by the process of ENAP. Consequently, the high TPH values at the start of the experiment were reduced to low and acceptable threshold values with improved nutrient concentrations and bioavailability.

As oil production activities still continue in the Niger Delta Region for a long time and therefore oil spillages, pollutions due to equipment failures, maintenance, sabotage etc will also continue. These noteworthy results from bioremediation applications confirmed the theoretical information base established by previous studies. The addition of nutrients in form of fertilizer to indigenous micro-organisms has proved to be effective in enhancing biodegradation and environmentally safe. It was also observed that microbes with the capacity to degrade oil are present in lowland environments and environmental parameters besides nutrients affect degradation rates in the field. Thus field applications of nutrients are still to some degree influenced by temperature, water runoff, substrate, and other environmental parameters that are neither fully understood nor easily quantified. However, in the lowland ecosystem of the Niger Delta, environmental conditions are favourable to these parameters. The results further demonstrated that ENAP can assist in achieving the benefits of quick intervention. This study deviated a bit from previous studies and focused on the interrelationship and influence of temperature on bioremediation rates and soil fertility improvement using the enhancement process of bioaugmentation and biostimulation. Based on the findings, it is recommended that remediation of hydrocarbon-polluted soil by enhanced natural attenuation should be encouraged since it is cost-effective and environmentally friendly. Oil companies in the Niger Delta should encourage the use of this technique in remediating hydrocarbon-polluted environment as a way of demonstrating their corporate responsibility of protecting the environment. Further studies should be conducted on how to improve the efficiency of the technique.

7. References

- Abii, T A; Nwosu, P C (2009) The Effect of Oil- Spillage on the Soil of Eleme in Rivers State of the Niger Delta Area of Nigeria. *Research Journal of Environmental Science* 3(3):316 – 320.
- Agbogidi OM, Eruotor PG, Akparabi S O. 2007; Effects of Time of Application of Crude Oil to Soil on the Growth of Maize (*Zea mays* L.) *Research Journal of Environmental Toxicology*.1(3): 116- 123.
- Andrade, M. and Cavelo, Vega, F. A. and Marcel, P. (2004), Technical Reports on Heavy Metals in Environment. Department of Vegetable Biology and Soil Science, AP 874, 36200 Vigo Spain.
- Aghalino, S.O (2000)PetroleumExploration and the Agitation for Compensation by Oil Mineral Producing Communities in Nigeria.*Journal of Environment and Policy Issues*, Vol. 1: II.
- Allard, A. S. and Nelson A.H. (1997). Bioremediation of Organic Waste Sites: a Critical Review of Microbiological Aspects. *Int. Biodeterior, Biodegrad.* 39:253–285.
- Amadi, A. and Ue Bari, Y. (1992) Use of Poultry Manure for Amendment of Oil Polluted Soils - inRelation to Growth of Maize (*Zea mays* L.). *Environment International* 18:521 - 527.
- Amadi, A., Dickson, A.A. and Maate, G.O (1993) Remediation of Oil- polluted Soils: Effect of Organic and Inorganic Nutrient Supplements on the Performance of Maize (*Zea mays* L). *Water, Air and Soil Pollution* 66:59-76.
- Atekwana E. A., Abdel Aal, G. Z., L. D. Slater, and E. A. Atekwana (2004a) Effect of Different Phases of Diesel Biodegradation on Low Frequency Electrical Properties of Unconsolidated Sediments, Paper presented at the Symposium on the *Application of Geophysics to Environmental and Engineering Problems*, Colorado Springs, Colorado.
- Atekwana E. A., Abdel Aal, G. Z., Slater, L. D and Atekwana E.A (2004b) Effects of Microbial Processes on Electrolytic and Interfacial Electrical Properties of Unconsolidated Sediments, *Geophys. Res. Lett.*, 31, L12505, doi:10.1029/2004 GL0200 3 0.
- Benka-Coker, M.O. and J.A Ekundayo, (1995) Effect of an oil spill on soil physic-chemical properties of a spill site in the Niger Delta area of Nigeria. *Environ. Monitor. Assess.*, 36: 93 -104.
- Braddock, J.F. Ruth, M.L..Walworth J.L and MCCarthyK.A (1997) Enhancement and Inhibition of Microbial Activity in Hydrocarbon-Contaminated Arctic Soils: Implications for Nutrient-Amended Bioremediation.*Environmental Science.Technology.*; 31:2078–2084.

- Daniel – Kalio, L A and Pepple S F(2006) .Effect of Bonny Light Crude Oil Pollution of Soil on the Growth of Dayflower in the Niger Delta, Nigerian Journal of Science Environmental Management 10: 111-114
- D’el Arco, J.P. and Franca, F.P., 1999. Biodegradation of crude oil in sandy sediment, International Biodeterioration and Biodegradation, 44: 87–92.
- Ekundayo, E. P, Emede, T. O. and Osayande, D. (2001). Plant foods for human nutrition, University of Benin, Nigeria.
- Essien, O E and John, I A (2010) Impact of Crude-Oil Spillage Pollution and Chemical Remediation on Agricultural Soil Properties and Crop Growth *J. Appl. Sci. Environ. Management*. Vol. 14 (4) 147 – 154 December,
- Gunawan, C., (2007). Bioremediation for Perchlorate-contaminated Groundwater. Michigan State Univ., Microbiology & Molecular Genetics, Course 445. Basic Journal 3: 6-13.
<http://www.taxonomicoutline.org/index.php/mmg445/article/view/220/274>.
- Gill, L.S. & R.M.A. Sandota(1976. Effect of foliarly applied ccc on the growth of Phaseolusaureus Roxb. (mung or green gram). Bangladesh Jour. Biol. Sci. 15:35-40.
- Harris, J.A, Bentham H and Birch P (1991). Soil Microbial Community Provides Index to Progress, Direction of Restoration. *Restoration Manage. Notes*9:133–135
- Illmer, P and Schinner F (1995) Solubilization of Inorganic Calcium Phosphates -Slubilization Mechanisms. *Soil BiolBiochem*. 27:257–263.
- Insam, H. and Domsch K.H.(1988) Relationship between Soil Organic Carbon and Microbial Biomass on Chronosequences of Reclamation Sites. *Microb.Ecology*. 15:177–188.
- Lakatos, G, Kiss M.K and Juhasz P. (1997) Application of Constructed Wetlands for Wastewater Treatment in Hungary. *Water. Sci. Tech.*, 3: 331-336.
- Lau, P. S., N.F.Y. Tam and Y.S. Wong. (1995). Effect of Algal Density on Nutrient Removal from Primary Settled Wastewater. *Environmental Pollution*, 89: 59-66.
- Manahan, S.E (1994). *Environmental Chemistry*. CRC Press, Inc. Florida. pp. 811.
- Obire, O. and Akinde, S.B. (2006). Comparative Study of the Efficiency of Cow Dung and Poultry Manure as Nutrient Alternative Sources in Bioremediation of Oil Polluted Soil. *Niger Delta Biologia* 5(2): 82-91.
- Odu, C.T.I., Nwoboshi L.C and Esuruoso O.F (1985) Environmental Studies (Soils and Vegetation) of the Nigerian Agip Oil Company Operation Areas. In: Proceedings of an International Seminar on the *Petroleum Industry and the Nigerian Environment*, NNPC, Lagos, Nigeria. pp. 274–283.
- Ogboghodo, A. A, Osemwota, I. O. Iruga, E. K and Chokor, J. U (2000) Environmental Monitoring a Assessment. Department of Soil Science, University of Benin.
- Ogboghodo, A. A. et al (2004) *Environment Monitoring and Assessment*. Netherlands Springer.
- Onifade A.K, (2007) Bioremediation of Crude Oil Polluted Soil in the Niger Delta by Enhanced Natural Attenuation. *Research Journal of Applied Sciences* 2(4): 498 – 504.
- Osuji, L.C. (2001). Total Hydrocarbon Content of Soils, Fifteen Months after Eneka and Isiokpo Oil Spills. *Journal. Applied Science and Environmental Management* 5 (2): 35-38.
- Osuji, L.C, Adesiyan S.O and Obute G.C (2004). Post Impact Assessment of Oil Pollution in the Agbada West Plain of Niger Delta Nigeria: Field Reconnaissance and Total Extractable Hydrocarbon Content. *Chem. Biodiv*. 1 (10): 1569-1
- Siddiqui, S. and Adams (2002) The Fate of Diesel Hydrocarbons in Soils and their Effect on the Germination of Perennial Ryegrass. *Environ. Toxicol.*, 17: 49-62
- Straube, W.L., Jones-Meehan, J., Pritchard, P.H. and Jones W.R., 1999, Bench Scale Optimization of Bioaugmentation Strategies for Treatment of Soils Contaminated with High Molecular Weight Polyaromatic Hydrocarbons, *Resources, Conservation and Recycling*, 27: 27–37.
- Trindade, P.V.O, Sobral, L.G, Rizzo, A.C.L, Leite, S.G.F and Soriano, A.U (2005) Bioremediation of Weathered and Recently Oil Contaminated Soils from Brazil: a Comparison Study, *Chemosphere*, 58(4): 515–522.
- Turco, R.F, Kennedy A.C and Jawson M.D (1994) Microbial Indicators of Soil Quality. In J.W. Doran, Coleman D.C., Bezdicsek D.F and Stewart B.A. (ed.) *Defining Soil Quality for a Sustainable Environment*. SSSA Spec. Publ. 35. SSSA, Madison, WI. pp. 73–99.
- Wokocho G. A, Emeodu D and Ihenko S (2011) Impact of Crude Oil Spillage on Soil and Food Production in Rivers State, Nigeria. *Journal of Money, Investment and Banking* ISSN 1450-288X Issue 19