



The Effect of *Abelmoschus Esculentus* (Okro) Crop on Crude Oil Pollution

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Abstract: The effect of *Abelmoschus esculentus* (Okro) crop on crude oil pollution was examined in University of Port Harcourt from July to September, 2016. Microbial enumeration was done by plate count followed by physiochemical parameters. Result shows that the total culturable heterotrophic bacterium (TCHB) in unpolluted of Day 01 shows a count of 2.25 log cfu/g, Day 07 recorded a count of 2.23 log cfu/g, Day 14 recorded a count of 1.70 Log cfu/g while Day 21 count was 1.00 log cfu/g respectively, The Day 01 shows a count of 2.25 log cfu/g, Day 07 recorded a count of 2.28 log cfu/g, Day 14 recorded a count of 2.20 Log cfu/g while Day 21 count was 1.80 log cfu/g in polluted soil. The Hydrocarbon utilizing bacteria (HUB) in unpolluted soil Day 01 count was 1.60 log cfu/g, Day 07 was 1.77 Log cfu/g, Day 14 recorded 1.65 log cfu/g counts while Day 21 count was 1.47 log cfu/g but polluted soil results shows a count of 1.90 log cfu/g for Day 01, Day 07 recorded a count of 2.04 log cfu/g, Day 14 recorded a count of 2.03 log cfu/g while day 21 count was 1.99 log cfu/g. Comparative analysis of TCHB distribution on polluted and unpolluted soil Day 01 of the analysis recorded a uniform count of 2.25 log cfu/g for polluted soil while for Day 21 a count of 1.00 log cfu/g for unpolluted and 1.80 log cfu/g for polluted soil. The moisture content of polluted soil was 22.0 while the non polluted soil had 21.0; temperature of the polluted soil was 29 °C with 25.7 °C for the unpolluted soil. pH for polluted soil was 4.90 while the non polluted soil had a pH of 5.21. Sulphate for polluted soil was 18.0% while unpolluted soil had 34.0% while nitrate concentration for polluted soil was 8.0% and 12.0 % for unpolluted soil. The organisms isolated in this study include; *Bacillus* sp, *Pseudomonas* sp, *Serratia* sp, *Escherichia coli*, and *Enterobacter* sp Due to these exudates, microbial populations and activities are 5 to 100 times greater in the rhizosphere than in bulk soil.

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Key word: *Abelmoschus esculentus*, Crude oil, Hydrocarbon utilizing bacteria, Pollution, Rivers State, Nigeria.

Introduction

Petroleum products are some of the most widely used chemicals which because of their wide usage, can easily spill, leak or be discharged to the environment. The leakage, discharge and spillage of petroleum products lead to the pollution of terrestrial and aquatic environments. It has been known that soil contamination by petroleum products is one of the world's most common environmental problems (Agbogidi *et al.*, 2007). The presence of petroleum products in the environment poses danger to the growth of plants and the wellbeing of animals' resident or dependent on the environment. Several researchers have shown that the individual petroleum products have effects on the growth and performance of plants. Microorganisms and plants have complementary role in rhizoremediation of oil-polluted soil. The plant rhizosphere is recognized as a niche rich in growth substrates in comparison with the surrounding bulk soil. Rhizospheres are dynamic microenvironments in which microbial communities

have access to an elevated supply of carbon, oxygen and energy rich materials from plant roots (Kuiper *et al.*, 2004). Rhizospheres are also stable physically, avoiding the potentially adverse effects of naturally occurring disturbances on microbial community composition or activities (Kuiper *et al.*, 2004).

The success of rhizoremediation of hydrocarbon-contaminated soil is dependent on the plant capacity to enhance the microbial activity in the plant rhizosphere, i.e. the main factor for the remediation of contaminated soil is the rhizosphere microflora of the plant adapted to the contaminant (Muratova, *et al.*, 2003).

Microorganisms on the rhizosphere exert big effect on plants; they lead to a continuous breakdown of organic and mineral compounds, which become available to plants. Moreover, they produce organic and non-organic acids, influence the dissolution of mineral salts and protect the plants against the phytopathogens (Alexander, 1994; Atlas, 1992).

There is the need to reclaim soils polluted with petroleum products. Hitherto remediation of soil polluted with petroleum products has involved physical, chemical and biological processes. The destruction of large areas of arable farmland at different villages in the Niger Delta where oil spillage occurs regularly is of a great concern since majority of people living here are farmers. There is a futuristic tendency of starvation if oil spills continue to spread across unpolluted soils. According to (Udonwa *et al.*, 2014; Udoh *et al.*, 2008). Food shortage and unemployment, increases vulnerability to HIV/AIDS. People affected heavily by oil pollution will involve themselves in any kind of activity that yields money to make ends meet irrespective of the consequences due to the destruction of their means of livelihood.

There is continual contamination of surrounding waters by run-off waters due to rain fall which has adversely affected the aquatic life by depriving them of oxygen. Water bodies stink from suffocation and death of fishes and other living organisms as a result of run-off rain water carrying oil and emptying itself into uncontaminated surrounding water bodies. This also affects the occupation of the fishermen in these areas as fishes are destroyed thus offering massive fishing retrenchment to these fishermen. Oil pollution, whether acute or chronic, has deleterious effects on agricultural lands and hence significant effects on plant growth (Agbogidi *et al.*, 2007). 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). The general 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. However, because of the expensive nature of Physical and chemical methods, the use of biological process for remediation is being evaluated. In the biological process of remediation, plant, animal and/or microbial activities are used. It involves treatment or breakdown of wastes contaminants into non-toxic forms using biological processes. This research article examined the effect of the growth *Abelmoschus esculentus* (Okro) on crude oil polluted soil and isolation of microorganisms that can degrade crude oil with respect to the rhizosphere of *Abelmoschus esculentus* (Okro).

Materials And Methods

Study Location

The research was conducted on soil sample collected from University of Port Harcourt botanical garden located south from Ofrima building complex before the school gate with a link road joining the east

west road and spent with Bonny Light crude oil obtained from a flow station, few kilometers from Port Harcourt Terminal of the Shell Petroleum Development Company of Nigeria.

Sample Collection and set up

The soil samples were collected using sterile bags, 2kg of soil sample was bagged before the seeds were sown (initial) and after the growth of the crops (final). The soil sample for the study was collected from the rhizosphere of *Abelmoschus esculentus* (Okro). Bonny Light crude oil sample was used for the degradation study which was obtained from a flow station, few kilometers from Port Harcourt Terminal of the Shell Petroleum Development Company of Nigeria. Physico-chemical analysis such as water holding capacity of the polluted and non-polluted soil, temperature, moisture content, nitrate, and sulphate content were determined based on the methods of Kennedy (2005).

Enumeration of Microbes by Spread Plate Technique

Each of the prepared media was poured into sterile Petri-dishes in triplicates of three each after autoclaving and cooling down to about 45-50⁰C. The agar media was allowed to solidify and dried in a dryer. After drying a 0.1ml and 1.0ml of each of the last three prepared dilutions (10⁻³ to 10⁻⁵) was transferred into three sterile Petri dishes (triplicate) containing the solidified agar by means of sterile syringes. The triplicate plates were finally incubated at room temperature for 24-48 hours for the potato dextrose agar. For the mineral salt agar, incubation was done for seven days. Filter paper soaked in crude oil was placed on the cover of the Petri dishes containing the mineral salt medium. The hydrocarbons in the crude oil served as sole carbon source to the inoculated microorganisms. The plates containing colonies between the ranges of 30-300 were selected and colony counts taken. The means of triplicates counts was recorded.

Result And Discussion

Microorganisms on the rhizosphere exert big effect on plants; they lead to a continuous breakdown of organic and mineral compounds, which become available to plants, this was investigated with the effect of *Abelmoschus esculentus* (Okro) crop on crude oil pollution in University of Port Harcourt from July to September, 2016. The morphological features of *Abelmoschus esculentus* (Okro) crop on both polluted and unpolluted soil is shown in Table 1. The colour of the polluted soil plant was observed as light green while the unpolluted soil plant colour was dark green. The height in inches on polluted soil was 3” while the unpolluted soil plant measured 1”, the crop grew with

three leaves for both soils and all seeds germinated with stunted growth on polluted soil.

Table 1: Morphological characterization of *Abelmoschus esculentus* (Okro) on polluted and unpolluted soil

S/N	Feature	Polluted Soil	Unpolluted Soil
1.	Colour	light green	Dark green
2.	Height (inches)	1	3
3.	No of leaves	3	3
4.	Germinated seeds	3	3

The total culturable heterotrophic bacterium (TCHB) in polluted and non unpolluted soil is shown in Figure 1. Day 01 shows a count o 2.25 log cfu/g, Day 07 recorded a count of 2.23 log cfu/g, Day 14 recorded a count of 1.70 Log cfu/g while Day 21count was 1.00 log cfu/g respectively. The Day 01 shows a count o 2.25 log cfu/g, Day 07 recorded a count of 2.28log cfu/g, Day 14 recorded a count of 2.20 Log

cfu/g while Day 21count was 1.80 log cfu/g respectively as represented in Figure 2. This was in agreement with the work of Juhanson *et al.*, (2007) they reported that phytoremediation is an emerging alternative with rhyzospherical organisms. The count was however attributed to the high microbial activities in the roots of the crop.

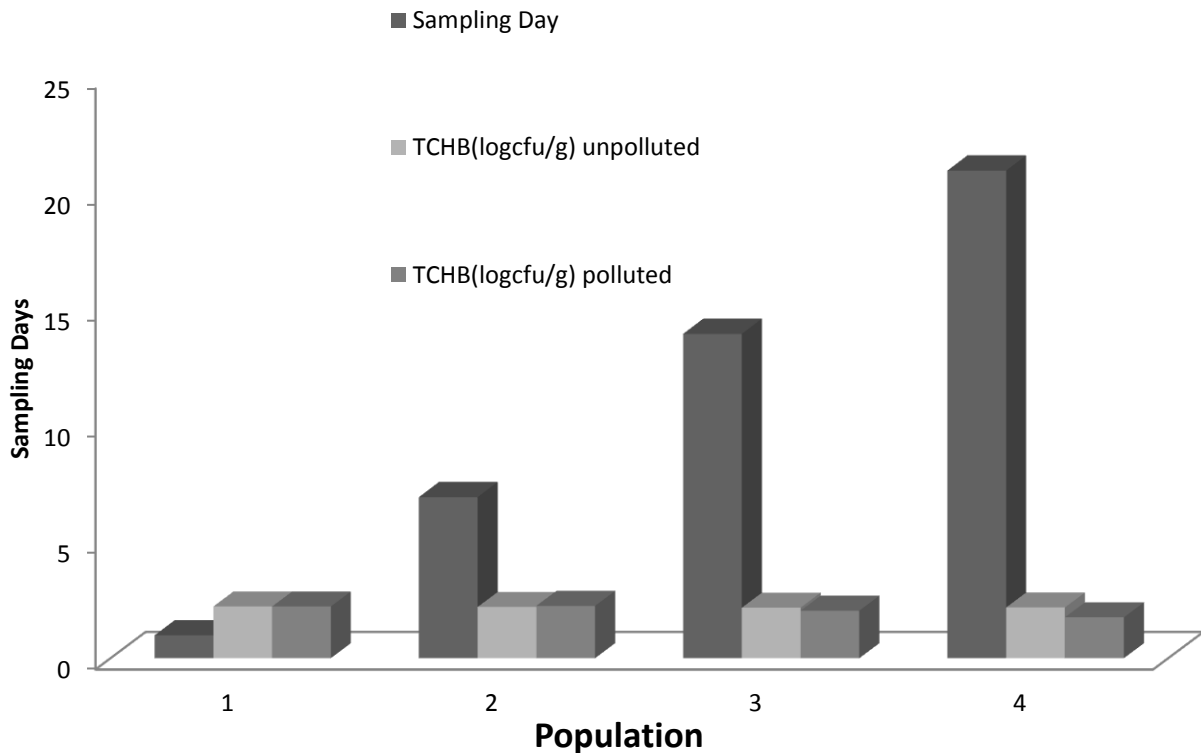


Figure 1: Enumeration of total culturable heterotrophic bacteria (TCHB) in unpolluted soil and polluted soil

The Hydrocarbon utilizing bacteria (HUB) in unpolluted soil count recorded from polluted soil is shown in Figure 4.2. Day 01 count was 1.60 log cfu/g, Day 07 was 1.77 Log cfu/g, Day14 recorded 1.65 log cfu/g counts while Day 21 count was 1.47 log cfu/g. The Hydrocarbon utilizing bacteria (HUB) enumerate from polluted soil I represented in Figure 2. Results

shows a count of 1.90 log cfu/g for Day 01, Day 07 recorded a count of 2.04 log cfu/g, Day 14 recorded a count of 2.03 log cfu/g while day 21 count was 1.99 log cfu/g respectively. These results agrees with the report of Hamamura *et. al.*, (2007) they reported that microbial dynamics is more accumulated in root of plants during rhyzoremediation.

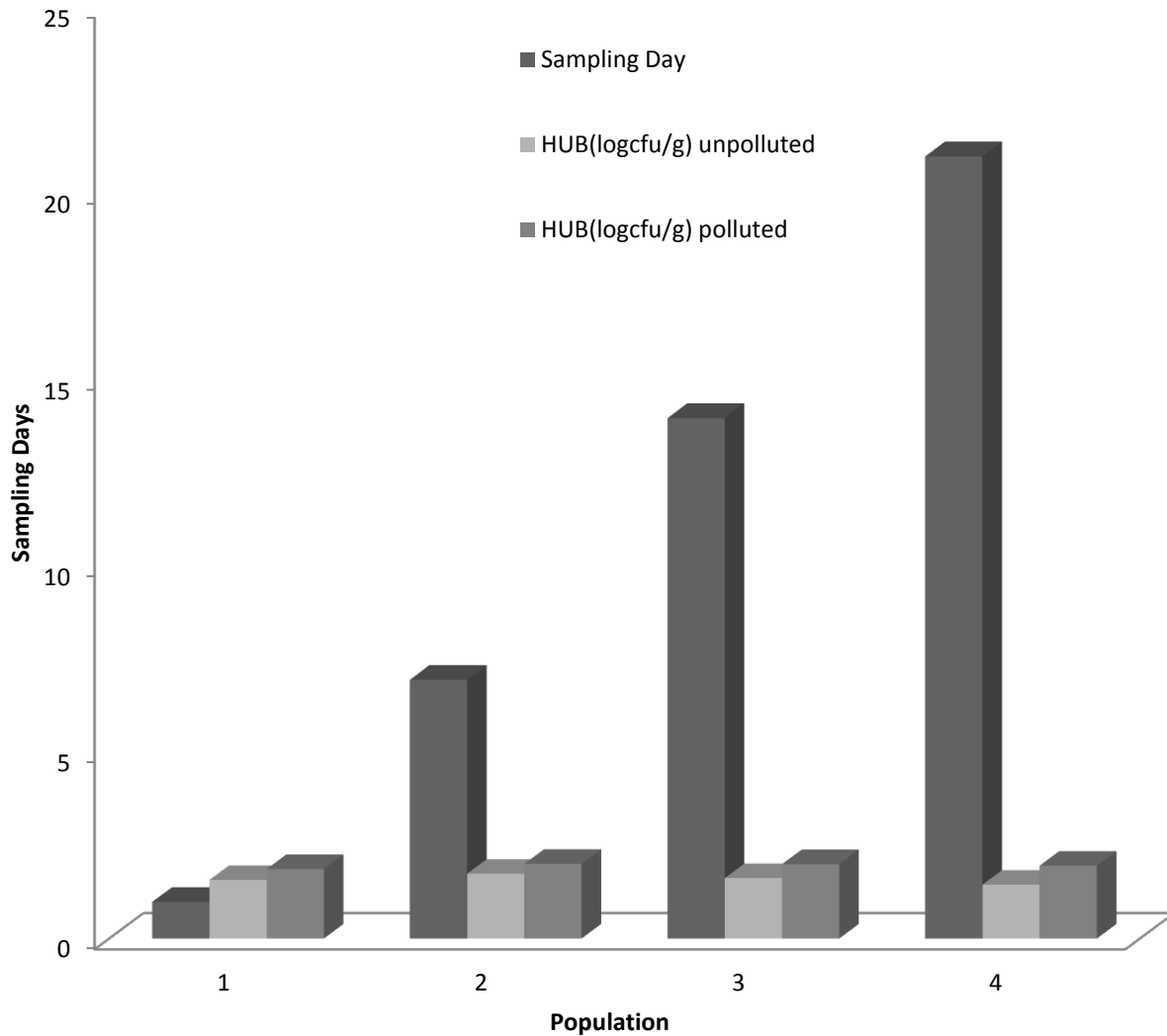


Figure 2: Enumeration of Hydrocarbon utilizing bacteria (HUB) in unpolluted soil and polluted soil

Comparative analysis of TCHB distribution on polluted and unpolluted soil Day 01 of the analysis recorded a uniform count of 2.25 log cfu/g for polluted soil, the Day 07 count was 2.23 log cfu/g for unpolluted and 2.28 log cfu/g for polluted, Day 14 recorded a count of 1.00 log cfu/g for unpolluted and

2.20 log cfu/g for polluted soil while for Day 21 a count of 1.00 log cfu/g for unpolluted and 1.80 log cfu/g for polluted soil respectively. The progressive increase in microbial count agrees with the work of Hamamura *et al.*, (2007), the microbial population dynamics increase during remediation process.

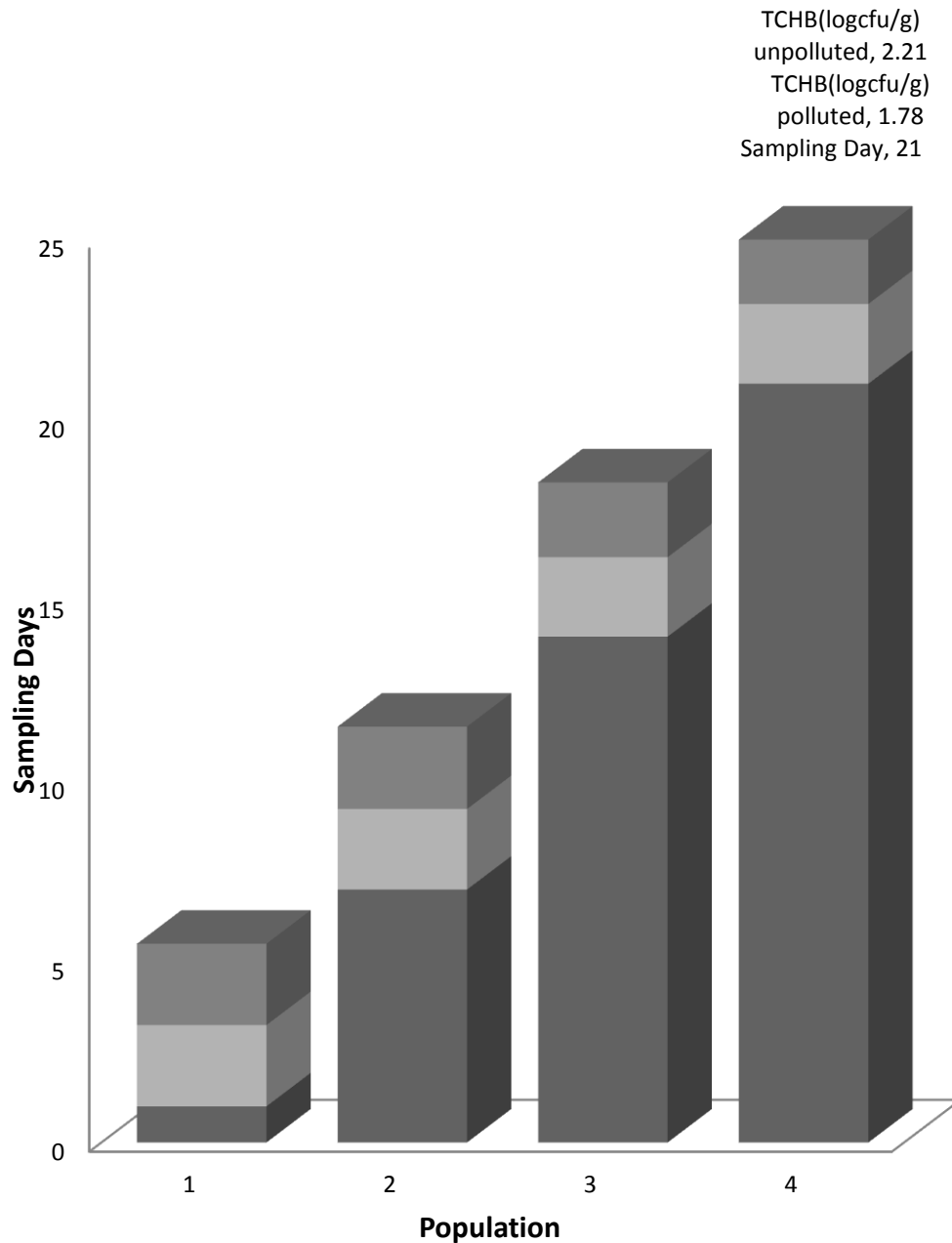


Figure 4: Comparative analysis of TCHB distribution on polluted and unpolluted soil

Table 2 shows the physiochemical parameters for both polluted and unpolluted soil. The moisture content of polluted soil was 22.0 while the non polluted soil had 21.0; temperature of the polluted soil was 29 °C with 25.7 °C for the unpolluted soil. pH for polluted soil was 4.90 while the non polluted soil had a pH of 5.21. The concentration of crude oil used in

polluting the soil was 15 ml for polluted soil, sulphate for polluted soil was 18.0% while unpolluted soil had 34.0% while nitrate concentration for polluted soil was 8.0% and 12.0 % for unpolluted soil respectively. Due to these exudates, microbial populations and activities are 5 to 100 times greater in the rhizosphere than in bulk soil.

Table 2: Physiochemical parameters

S/N Feature	Polluted Soil	Unpolluted Soil
Moisture content	22.0	21.0
Temperature (°C)	29	25.7
pH	4.90	5.21
Concentration (ml)	0.00	15
Sulphate (%)	18.0	34
Nitrate (%)	8.0	12.0

Conclusion

The contamination of crude oil results in an immediate change in the bacterial community structure, an increasing abundance of hydrocarbon-degrading microorganisms and a rapid rate of oil degradation, which suggests the presence of pre-adapted, oil-degrading microbial community and sufficient supply of nutrients.

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