Journal of Environmental and Agricultural Studies
ISSN: 2710-140
DOI: 10.32996/jeas
Journal Homepage: www.al-kindipublisher.com/index.php/jeas

Effect of Soil Contamination with Crude Petroleum on Cowpea: An Insight into the Prospects of Crop Production in Nigerian Frontier Basins

Ismail, H. Y[1] ✉ Farouq, A. A[2], Rabah A. B[3], Muhammad A. B[4] and Aliyu R. U[5]
[1]*Department of Microbiology, Faculty of Science, University of Maiduguri, Nigeria*
[23]*Department of Microbiology, Faculty of Chemical and Life Sciences, Usmanu Danfodiyo University Sokoto, Nigeria*
[4]*Department of Applied Chemistry, Faculty of Chemical and Life Sciences, Usmanu Danfodiyo University Sokoto, Nigeria*
[5]*Department of Biochemistry, Faculty of Chemical and Life Sciences, Usmanu Danfodiyo University Sokoto, Nigeria*
✉ **Corresponding Author:** Ismail, H. Y, **E-mail**: yismailh@gmail.com

ARTICLE INFORMATION

Received: 09 September 2021
Accepted: 16 October 2021
Published: 02 November 2021
DOI: 10.32996/jeas.2021.2.2.5

KEYWORDS

Contamination, crude oil, cowpea, environment, soil health, crop

ABSTRACT

Petroleum hydrocarbon contamination remains a major challenge confronting soil health, environmental sustainability, and food security in oil exploration areas. In this study, the effect of Bonny Light crude oil on cowpea (*Vigna unguiculata* L. Walp.) was investigated with a view to assessing its toxicity to plant growth and performance. Pristine soil samples were collected in different pots and contaminated with crude oil to achieve 0.0%, 2.5%, 5.0%, 7.5% and 10.0% v/w contamination levels. Viable seeds of cowpea were planted and monitored for the emergence and subsequent growth for a period of 12 weeks. Results showed that the crude oil extended the period of seed germination and delayed the emergence of sprouts by 2 days at a rate of 96.7%, 80.0%, 50.0%, 96.7%, and 73.3% emergence respectively. The plants' shoots, roots, and leaves lengths were longer in control than in the contaminated soil. Phytotoxicity study showed that shoots, roots and leaves lengths of the plants were significantly reduced by ≥ 50% of the control. The relative plants' weights, chlorophyll, and the number of leaves were worst affected especially in plants grown in higher crude oil concentrations where fewer or absence of leaves was observed at the end of the experiments. No yield parameter was observed in all plants grown in contaminated soil as opposed to the control where flowers, fruits, and seeds were produced. The findings illustrated that the growth rate of *V. unguiculata* was severely affected due to hydrocarbon contamination in a concentration-dependent manner. It further demonstrated the imminent danger to food security especially in frontier basins with impending oil exploration activities. Therefore, there is a need to identify and integrate effective measures that minimize or prevent oil spillage in the course of oil exploration activities with a view to avoiding the repeat of persistent pollution problems disturbing host communities.

1. Introduction

Environmental contamination with petroleum hydrocarbons (PHs) is one of the major threats to soil health and agricultural productivity (Odukoya *et al.*, 2019a). Hydrocarbons in the soil environment originate from petroleum exploration, refining, transportation, and storage. An increase in the demand for petroleum as a source of energy has led to a marked increase in soil contamination especially in developing countries (Kekere *et al.*, 2011). Soil contamination with hydrocarbons is generally due to spillage which is frequently occurring, albeit in small volumes (Fingas *et al.*, 2001). Crude oil spills are linked to technical malfunctions, deliberate human acts, and faults during transportation and storage (Schmidt, 2011). Incessant generation of petroleum wastes and their discharge to the environment in the form of sludge is greatly increasing soil contamination with recalcitrant hydrocarbons and toxic heavy metals (Sangeetha and Thangadurai *et al.*, 2014). The resultant effect on the environment particularly soil is enormous even though little is known about the additive, synergistic, or antagonistic effects of the various components present in the petroleum mixture (Baek *et al*., 2004). Contamination of soil with petroleum hydrocarbons compromises the ability of the soil to filter, buffer and transform inorganic and organic contaminants (FAO, 2018).

Petroleum hydrocarbons exert acute toxicity on plant growth and performance (Terek, *et al*., 2015; Odukoya, 2016; Odukoya *et al*., 2019b). The impact of hydrocarbon contamination on plants is often noticed immediately after spillage or a few days later. Vegetation loss, phytotoxicity, and plant stress which depend upon plant species type and degree of contamination are the major noticeable phenomena (Emengini *et al*., 2013; Mohamadi *et al*., 2016). Petroleum impedes plant growth by reducing the germination rate, soil fertility and reducing the resistance of plants to pests and diseases (Wang *et al*., 2017). It is also associated with a significant decrease in plant biodiversity and richness (Mohamadi *et al*., 2016; Arellano *et al*., 2017). Disturbances of major physiological processes within plants systems are unexceptional and regularly manifest in the form of alterations in morphological, chlorotic, and burnt foliar patterns. Traces of petroleum hydrocarbons are often detected in fruits and vegetables, creating an important health hazard (Paris *et al*., 2018).

Petroleum hydrocarbon spillage is one of the common causes of vegetation loss contributing significantly to deforestation (Duke, 2016). Where farmlands are in the vicinity of an oil spill, crop plants are destroyed and the lands are rendered inoperable (Ani *et al*., 2015). There are reports on environmental degradation of agricultural lands due to negative effects on the farm or fallow acreage and swamps (Akpokodje and Salau, 2015; Ani *et al*., 2015). The magnitude of the obliterations affects the livelihood of communities in form of reduced crop yield which can lead to food insecurity and poverty (Egbe and Thompson, 2010; Osuagwu and Olaifa, 2018). It has been reported that oil spills could lead to a 36% reduction in the ascorbic acid content of vegetables and 40% decrease in crude protein content of cassava and a 60% reduction in household food security (Ordinioha and Brisibe, 2013).

Nigeria is among the leading countries in oil reserves and exploration (Organization of the Petroleum Exporting Countries; OPEC, 2019). Crude oil and natural gas are some of the major drivers of the Nigerian economy, constituting about 10% of gross domestic product, and petroleum exports revenue represents around 86% of total earnings from exports (OPEC, 2021). Although most sedimentary basins in Nigeria are believed to amess petroleum deposits (Eneh, 2005), the Niger Delta basin is the only region with active petroleum production. The region has experienced devastating oil spill scenarios in the last 60 years. Estimations by the Nigerian National Petroleum Corporation (NNPC) showed that there is an average of 300 individual spills in the Niger Delta annually and more than 7,000 oil spill incidents have occurred between 1970 and 2000 (Mahmoud, 2017). With the recurrence of the oil spills (Shell Petroleum Development Company; SPDC, 2019), over 13 million barrels have been spilled in the region, of which the majority of the spilled oil still lingers in the ecosystem (Mahmoud, 2017; Sam *et al*., 2017).

The detrimental effects of oil spillage on agriculture and the socioeconomic activities of people in the Niger Delta region are colossal (Onyema and Sam, 2020). The impacts of oil exploration on agricultural practices in the area and lack of proper commitments towards environmental cleanup have contributed to ineffable hardship which resulted in recurrent socio-economic upheavals by acclaimed freedom fighters which further compound the problem of environmental pollution (Tantua and Kamruzzaman, 2016; Babatunde *et al*., 2017). Different field and laboratory investigations from the region have highlighted the devastating effects of crude oil on farmlands, crop plants, and also the marine environment (Chindah and Braide, 2000; Egbe and Thompson, 2010; Matemilola *et al*., 2018; Onyema and Sam, 2020). Some of the spasmodic experiences include reduction of soil fertility, poor soil aeration, farmland degradation, elevated soil temperature, destruction of soil microbial communities, destruction of soil structure, low productivity, yellowing of crop leaves, stunted growth of the crop, rotting tubers, wilting of the crop, burnt crop leaves and the outbreak of crop diseases (Ani *et al*., 2015). The singular and cumulative effects of the above conditions eventually lead to crop failure or poor yield (Inoni *et al*., 2006; Ukpong and Obok, 2018). For instance, a significant effect of oil spillage on cassava (a common staple tuber crop in the area) production was observed by Ahmadu and Egbodion (2012) to include stunted growth, rotting tubers, crop failure, and poor yield.

In recent years, there is increasing interest to expand petroleum exploration to frontier basins in Nigeria (Adegoke *et al*., 2015). Preliminary investigations have shown that there are petroleum deposits in commercial quantities in Dahomey basin (Osundina *et al*., 2002), Bornu basin (Hamza and Hamidu, 2012, Adekoya *et al*., 2014; Ilozobhie, 2018), Sokoto basin (Obaje *et al*., 2013; Obaje *et al*., 2020) and Mid-Niger basin (Obaje *et al*., 2013; Ojo *et al*., 2020). In October 2019, NNPC announced that it discovered crude oil and gas in the Kolmani River II Well on the Upper Benue Trough, Gongola Basin. It is anticipated that the mining of petroleum resources would be embarked upon within the shortest possible time. However, considering the circumstances following oil exploration in Niger Delta in retrospect, oil spillage in the new mining areas is imminent. Demographically, the socio-economic activities in the areas are not distinctly better than the one in the Niger Delta if not worst.

Damage to the arable lands which form the major source of sustenance in Nigerian frontier basins will lead to unpredictable consequences in the nearest future. This will cause serious havoc to the economy, environment, health, and food security in the regions and the country in general. Consequently, soil damage due to oil pollution would be a great challenge to the ongoing efforts by authorities to revitalize agriculture for food and sustainability. Being cowpea is one of the major food crops cultivated in most of the basins, the present study monitored its growth under hydrocarbon contamination as a model plant. This is with a view to highlighting the negative effects of crude oil contamination particularly on its growth and agricultural productivity in general. This would enable stakeholders to make an informed decision on effective spill prevention protocols to avoid compounding environmental, food, and socioeconomic crises in Nigeria. It would also enable host communities to prepare against the imminent adversities that petroleum exploration may cause to their environment and source of livelihood.

2. Materials and Methods

2.1 Study area

Experiments were conducted at the Botanical Garden of the Usmanu Danfodiyo University Sokoto (UDUS; 13.1246° N, 5.1994° E), in 12 weeks period between September to December 2020. The University is located in Sokoto, the capital of Sokoto State, Northwestern Nigeria. has a tropical continental type of climate dominated by two opposing air masses: the tropical marine from the south and tropical continental from the north. Annual rainfall is about 550 mm with the highest peak in August. Dry season sets in first with the cold harmattan from October to February, and a hot period comes in from March to the end of May when temperatures reach 38ºC during the day with humidity less than 20% and the rain begins in June to September (Sokoto State, 2000).

2.1 Sample collection

Cowpea seeds were collected from the National Animal Production and Research Institute (NAPRI), Ahmadu Bello University, Zaria, Nigeria. Bonny light crude oil was obtained from Kaduna Refinery and Petrochemical Company, Kaduna. Pristine soil samples were collected from the Botanical garden UDU Sokoto

2.3 Study design

The study employed a randomized complete block design involving five sets of plastic pots. The pots were arranged and properly labeled as UR0.0, CR2.5, CR5.0, CR7.5, and CR10. Soil (sandy soil made up of 89.4% sand, 8.6% silt, and 2.0% clay particles; pH 6.8 ± 0.60; 27℃) with no previous history of hydrocarbon contamination was obtained from the botanical garden, UDUS; weighed and placed into the plastic pots.

2.4 Amendment of soil with crude oil

The setup of the experiment prepared earlier was contaminated with different concentrations of Bonny Light crude oil to achieve a contamination level of 0%, 2.5%, 5.0%, 7.5%, and 10% v/w in triplicates. The concentration used in this experiment acknowledged previous investigations that reported contamination beyond 3% concentration has been increasingly deleterious to soil biota and crop growth (Akpoveta, 2011). These pots were allowed to stand for one week in order to acclimatize (Fig. 1).

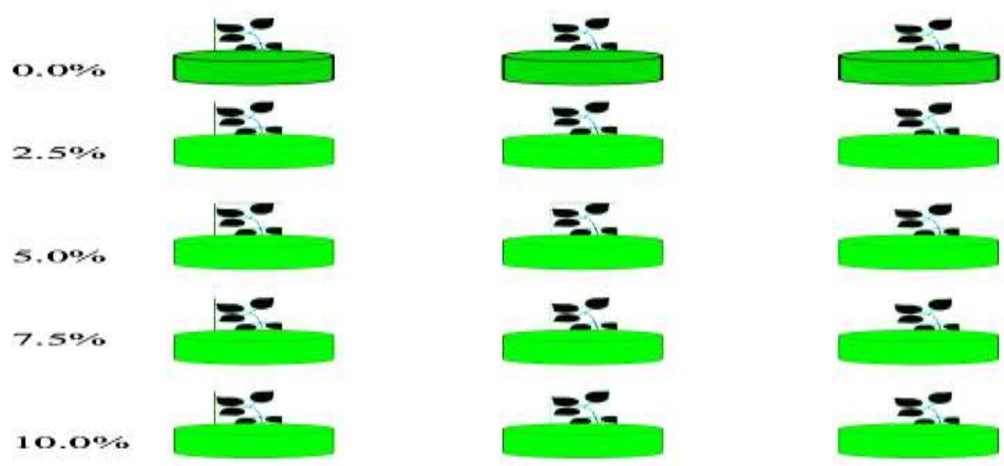

Fig. 1: Experimental design

2.5 Planting of Seeds
The viability of the seeds was tested by the floatation method (Diab, 2008). The seeds were tested by soaking in distilled water for 5 minutes. The floating seeds (non-viable) were removed and the water drained immediately from the seeds that sank (viable seeds). Seeds were sown at the rate of three seeds per hole and were placed at 4 cm deep into the soil (Nasir, 2001, Mullen *et al*., 2003). The pots were irrigated every 48hrs until yielding.

2.6 Phytotoxicity Studies
2.6.1 Determination of plant growth parameters
Plant growth parameters like days for emergence, emergence rate, leaf length, color, stem length, girth, number of flowers, number of leaves, number of seeds, dry weight, wet weight, and chlorophyll content were monitored every two weeks from the day of unequivocal emergence.

2.6.2 Days and rate of emergence
Days for emergence were determined by counting the number of days from the day of sowing to the day of apparent emergence of radicles.
The rate of emergence was determined using this relation:

$$\%\text{Emergence} = \frac{\text{Number of seedlings that spouted}}{\text{Number of seeds sowed}} \times 100 \text{ .... Eqn. (1)}$$

2.6.3 Measurement of plant growth
The girth, lengths of roots, shoots, and leaves of the plants were determined using the meter rule.

2.6.4 Counting of plant parts
The number of flowers, number of leaves, and number of seeds of each plant were counted manually after every two weeks.

2.6.5 Determination of plant weight
In each pot, a plant was harvested after 4 weeks. The plants were washed with distilled water and separated into two parts: shoots (including leaves and stems), and roots. The wet weight (WW) was measured immediately using a weighing balance and the dry weight (DW) was recorded after oven drying of the samples at 70˚C for 5 days.

2.6.6 Determination of chlorophyll content
One gram of leaf sample was cut finely and gently mixed with a clean pestle and mortar. To this homogenized leaf material, 20ml of 80% acetone and 0.5gm $MgCO_3$ powder were added. The materials were further ground gently. The sample was then put into a refrigerator at 4° for 4 hours. Thereafter, the sample was centrifuged at 500 rpm for 5 minutes. The supernatant was transferred to a 100ml volumetric flask. The final volume was made up to 100 ml with the addition of 80% acetone. The color absorbance of the solution was estimated by a spectrophotometer using 645 and 663nm wavelengths against the solvent. Acetone (80%) was used as a blank (APHA, 1989).
The chlorophyll content was determined using the relations:
Chl a = 11.75×A662.6 – 2.35×A645.6 .... Eqn. (2).
Chl b =18.61×A645.6 – 3.96× A662.6 .... Eqn. (3).
Where Chla and Chlb are the chlorophyll a and chlorophyll b respectively, A is absorbance.

2.7 Evaluation of phytotoxicity
The toxicity of hydrocarbon to the plants was determined according to the method of Ibrahim and Nafiu (2017). All the parameters examined above were compared with those in the control and the toxicity evaluated using the following relation:

$$\text{Percentage toxicity (\%)} = \frac{\text{Parameter of control} - \text{Parameter of treatment}}{\text{Parameter of control}} \times 100 \text{ .... Eqn. (4)}$$

2.8 Statistical analysis
Data obtained from this study were subjected to ANOVA using GraphPad Prism version 9.1.0. Significant differences were established by Dunnett multiple comparison test at p = 0.05. Spearman's correlation was used to establish a possible relationship between variables.

3. Results
3.1 Plant growth and performance
The phytotoxic effect of crude oil on cowpea was investigated. The emergence of sprouts was first observed four days after sowing especially in the control and treatments with lower contamination. Unequivocal emergence was recorded on the sixth day across

all the treatments. Results in Table 1 present the emergence rate on the sixth day after sowing, in which the highest rate was observed in uncontaminated or control soil (UR0.0) and soil contaminated with 7.5% v/w (CR7.5) crude oil with 96.7% each. Plants in CR5.0 and CR10 (5.0% v/w and 10% v/w contamination) had lower emergence rate with 50.0% and 73.3 % respectively. Statistical analysis showed that CR5.0 differed significantly from the control and also other treatments at a 95% confidence limit.

Table 1: Emergence (Germination) of *Vigna unguiculta*

	No. of Seeds Emerged/Treatment				
	UR0.0	**CR2.5**	**CR5.0**	**CR7.5**	**CR10**
Mean	9.67	8.0	5.0*	9.67	7.33
SE (±)	0.33	1.53	2.31	0.33	1.20
Emergence rate (%)	**96.7**	**80.0**	**50.0***	**96.7**	**73.3**
Phytotoxicity (%)	**-**	**17.27**	**48.29**	**-**	**24.20**

* Significantly different from others in a raw (p = 0.05; α = 0.0098)

The growth of *Vigna unguiculata* in contaminated soil was monitored against the control for a period of 12 weeks. During the first two weeks after emergence, rapid plant growth was observed in all the treatments. The longest shoots were observed in UR0.0 two weeks after emergence (25.17 ± 1.0 cm) and also throughout the experimental period as shown in Fig. 2. Significant differences were observed in their shoot lengths especially from the 6th through 10th week.

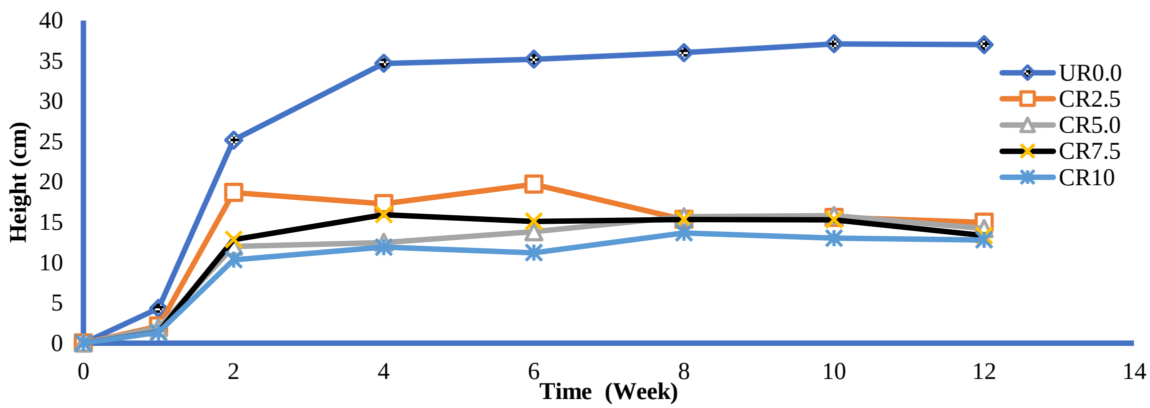

Fig. 2: Shoot length of cowpea plant growing under different hydrocarbon concentrations.

Similarly, plants in UR0.0 had more leaves (9.0 ± 1.0) than those in the other treatments. The least number of leaves were observed in plants CR7.5 (4.33 ± 1.8) and CR10 (5.0 ± 0.1). The plants had also varying numbers of leaves which were significantly different two weeks after emergence (Fig.3). The leaves continued decreasing with increasing hydrocarbon concentration.

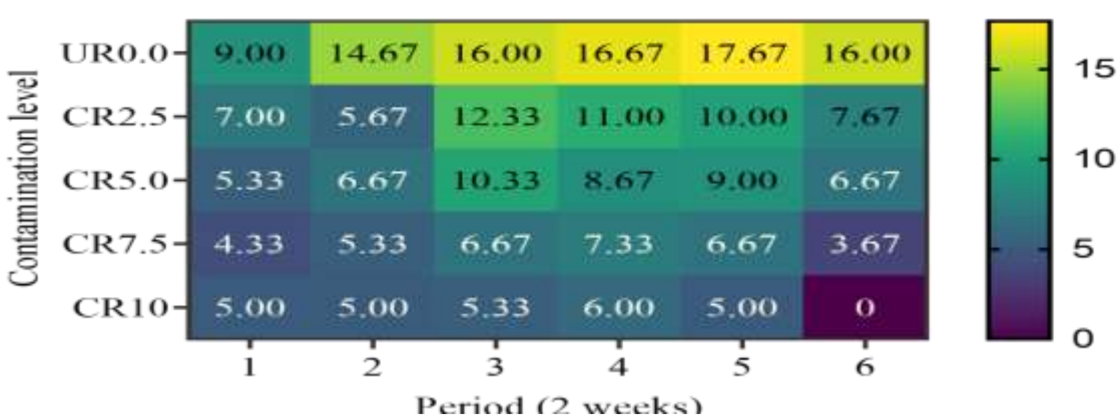

Fig. 3: Distribution of leaves (mean) among cowpea plants grown under different hydrocarbon concentrations

Plants in the control pots (UR0.0) had longer leaves from the beginning till the end of the experiment. A gradual increase in leaf length was observed from 2 weeks (7.5 cm) up to the 10th week (9.2 cm) as against plants in CR2.5, CR5.0, CR7.5, and CR10 where the length kept decreasing gradually until the end of the experiment as shown in Figure 4.

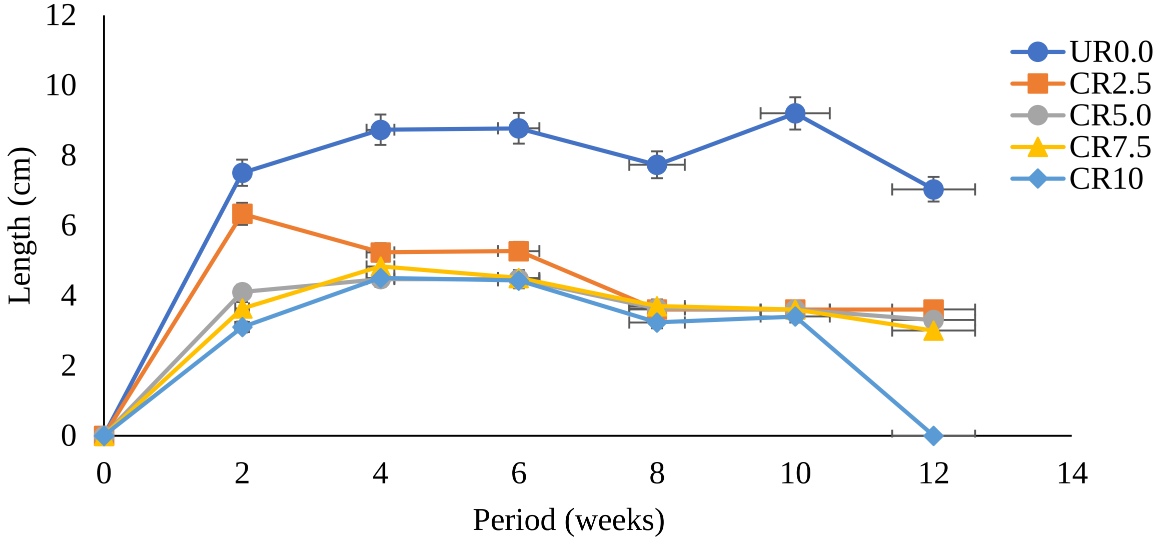

Figure 4: Leaf Length (Mean ± SE) of Cowpea plant grown in hydrocarbon-contaminated soil

More so, the plants in uncontaminated soil were characterized by longer roots than plants in contaminated soil (Fig. 5). The longest roots (19.4 ± 1.2 cm) were observed in UR0.0 after 3 months period. Root length decreased with increasing hydrocarbon concentration from CR2.5 through CR10 which differed significantly from that of the UR0.0 ($p < 0.05$). The shortest roots were observed in CR10 and CR5.0 with 4.2 ± 0.5 cm and 3.5 ± 1.0 cm respectively.

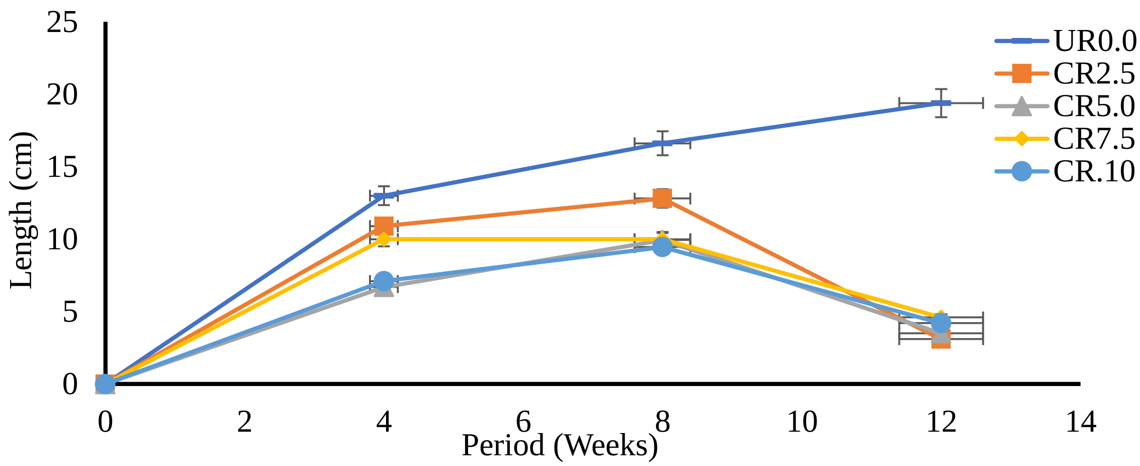

Figure 5: Root length of cowpea plants grown in hydrocarbon-contaminated soil

Figure 6 presents the results of plants' chlorophyll contents. In the early phase of the plant growth, the values for chlorophyll contents were closely related, in which plants in CR5.0 had the highest (41.50 ± 0.1 mg) chlorophyll content. In UR0.0, the chlorophyll was observed to increase over time and reached its peak in the 10th week (51.11 ± 1.1 mg). In CR7.5 and CR10, the lowest chlorophyll contents were recorded especially in the last weeks where the leaves suffered abscission.

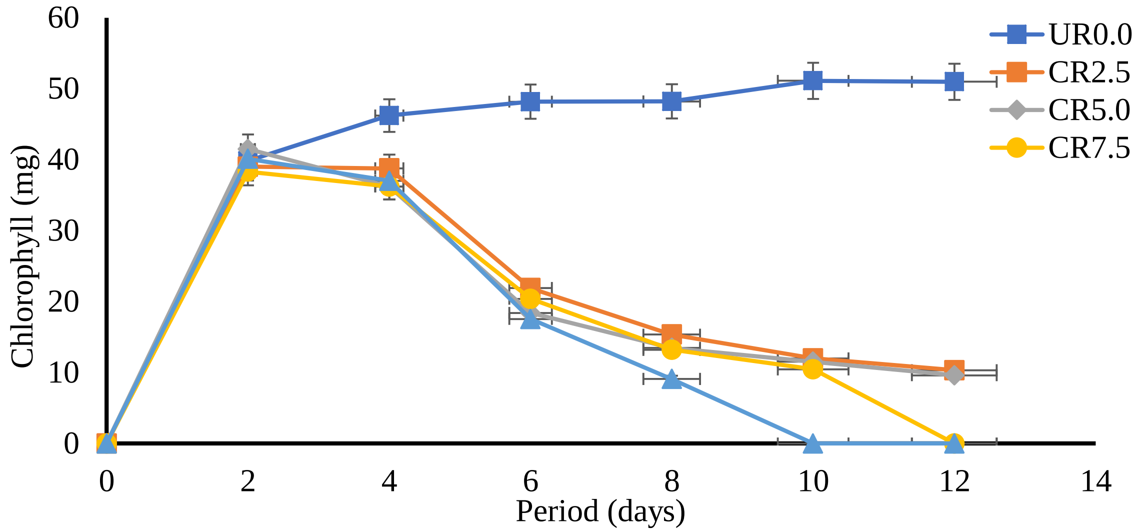

Figure 6: Chlorophyll contents of Cowpea plant grown in hydrocarbon-contaminated soil

Additionally, the weight of plants in UR0.0 was observed to be many folds higher than those in the treatment. The highest wet and dry weights were recorded in UR0.0 after 12 weeks with 32.7g and 13.7g respectively; whereas the least weight was recorded in CR10 in which the wet and dry weights were 8.80g and 1.9g respectively (Fig. 7).

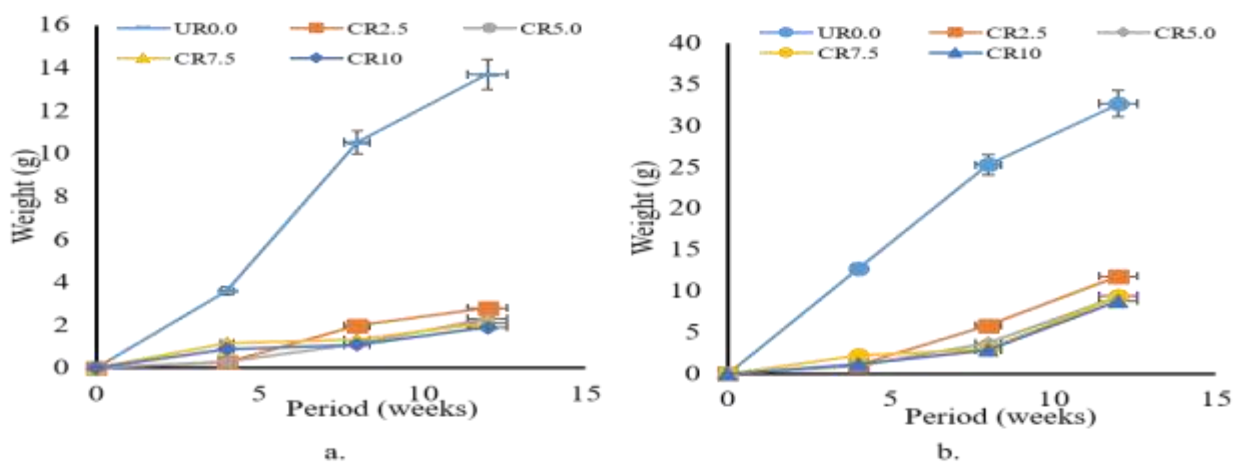

Figure 7: Weight of Cowpea plant grown in hydrocarbon-contaminated soil. a. Wet weight b. Dry Weight

3.2 Phytotoxicity of crude oil on cowpea plants

Results in Table 3 showed that seed germination and the subsequent emergence of sprouts were affected by the hydrocarbon contaminants except in CR7.5. Germination of seeds sown in CR5.0 was reduced by about 50% which represents the highest toxic effect on germination. Germination in CR2.5 and CR10 was also affected by 17.27% and 24.20% less than UR0.0 respectively. The shoot length of plants in CR2.5, CR5.0, CR7.5, and CR10 was reduced by 25.82%, 52.32%, 49.03%, and 58.96%, respectively two weeks after emergence. The highest reduction in shoot length was observed after 12 weeks except in CR10, where the plants were more stunted earlier enough in the 6th week. In terms of the root lengths, the highest toxicity (84.02%) was observed in CR2.5 at the end of the experiment. This was followed by CR10 and CR7.5 with 78.35% and 76.29% reduction in root length during the same period.

The leaf length and number of leaves of the plants were also affected by the hydrocarbon contaminants. The highest toxicity was observed in CR10 from 2 weeks till the end of the experiment where 100% effect (total absence of leaves) was recorded. For chlorophyll contents, the highest toxicity was observed in the last week of the experiment in which 79.63%, 81.13%, 100%, and 100% reduction in chlorophyll contents was observed in plants grown in CR2.5, CR5.0, CR7.5, and CR10 contaminated soil respectively. Correspondingly, the weight of plants was also severely affected by the hydrocarbon contaminants. The highest toxicity was observed one month after emergence, in which ≥ 90% reduction was observed in contaminated soil. Hierarchical clustering (Fig. 7) showed the relative similarity of the plants in various treatments to that of the control based on the induced phytotoxicity.

The yield of the plant shows that only plants in UR0.0 were able to bore flowers, fruits, and seeds. In the contaminated soil, however, neither flower nor fruits were formed as shown in Table 2. Based on the data obtained, plants in CR2.5 and CR7.5 performed better than CR5.0 and CR10 as shown in the hierarchical clustering presented in Figure 8.

Table 2: Yield of cowpea plant grown in different hydrocarbon concentrations

Treatment	Yield			
	Flower (Mean ± SE)	Pod (Mean ± SE)	Pod fresh weight (g)	Number of seeds (pod^{-1})
UR0.0	7.0 ± 1.2*	5.0 ± 1.0*	8.3 ± 0.6*	9.3 ± 2.5*
CR2.5	0.0	0.0	0.0	0.0
CR5.0	0.0	0.0	0.0	0.0
CR7.5	0.0	0.0	0.0	0.0
CR10	0.0	0.0	0.0	0.0

* Extremely significantly different ($p < 0.01$)

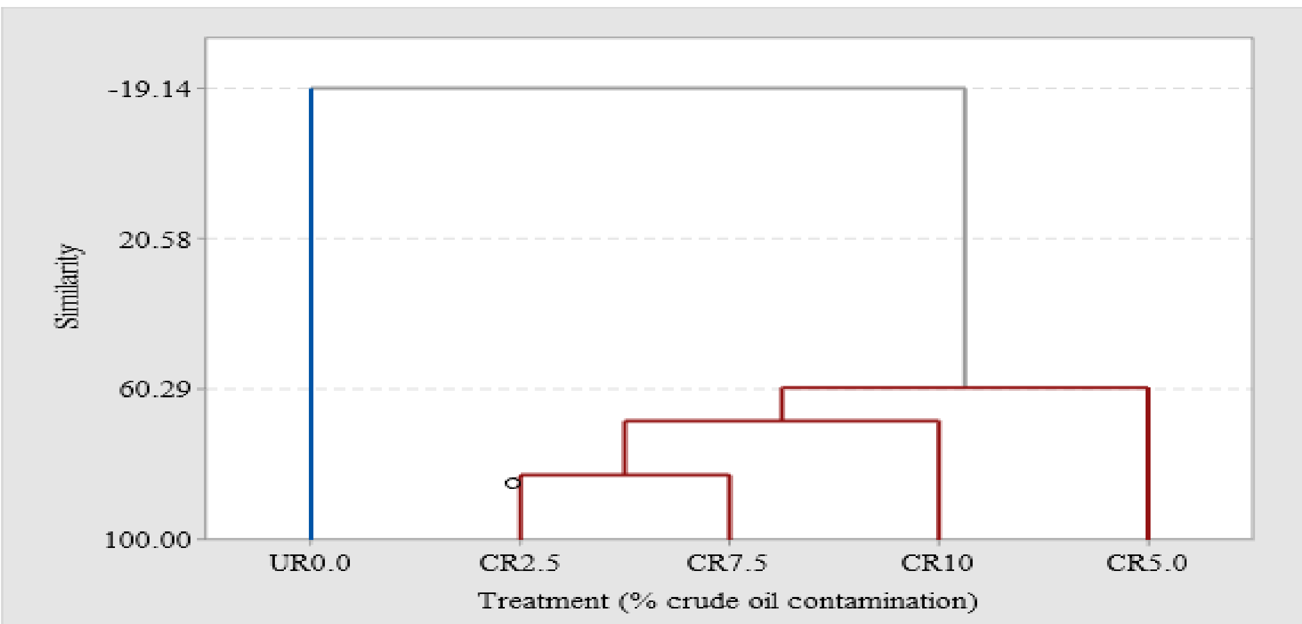

Fig. 8: Hierarchical clustering of plant growth and performance under hydrocarbon stress

Table 3: Relative toxicity of crude oil on the growth of Cowpea

	Toxicity level (%)					
Treatment (% v/w crude oil)	**2 WAP**	**4 WAP**	**6 WAP**	**8 WAP**	**10 WAP**	**12 WAP**
Shoot Length						
CR2.5	25.82	50.19	44.03	57.42	57.95	59.54
CR5.0	52.32	64.03	60.80	56.47	57.41	61.62
CR7.5	49.03	54.05	57.10	57.42	58.76	63.97
CR10	58.96	64.68	68.18	62.03	64.96	65.41
Leaf Length						
CR2.5	15.60	40.09	39.91	53.43	60.87	48.80
CR5.0	45.33	48.80	49.03	53.04	60.87	53.06
CR7.5	51.60	44.67	48.69	52.13	60.87	57.33
CR10	58.67	48.45	49.49	58.21	63.04	100.0

Number of Leaves						
CR2.5	22.22	61.35	22.94	34.01	43.41	52.06
CR5.0	40.78	54.53	35.44	47.99	49.07	58.31
CR7.5	51.89	63.67	58.31	56.03	62.25	77.06
CR10	44.44	65.92	66.69	70.01	71.70	100.0
Chlorophyll Contents						
CR2.5	2.06	16.11	54.54	65.25	76.52	79.63
CR5.0	-	21.64	61.78	69.52	77.42	81.13
CR7.5	3.79	21.64	57.67	70.11	79.53	100.0
CR10	-	19.88	63.55	79.41	100.0	100.0
Root Length						
CR2.5		16.15		22.92		84.02
CR5.0		48.46		37.66		20.03
CR7.5		23.08		39.83		76.29
CR10		45.38		43.08		78.35
Wet Weight						
CR2.5		91.98		76.96		63.91
CR5.0		92.53		85.53		71.25
CR7.5		82.63		88.14		71.25
CR10		90.88		88.70		73.09
Dry Weight						
CR2.5		92.22		81.29		79.56
CR5.0		91.94		89.36		83.21
CR7.5		67.78		87.37		84.67
CR10		75.53		89.55		86.13

WAP: weeks after planting

4. Discussion

The results from this study showed that the rate of seed germination and emergence was reduced by about 17.27% to 50% and linked to hydrocarbon contamination. Plants seeds are seriously damaged by crude oil due to the fact that some of its fractions have the capacity to wet and strongly penetrate into seed coat and embryo, which result in destruction and loss of seed viability (Kathi and Khan, 2011). Phytotoxic effects of hydrocarbons on seeds are correlated with hydrophobic properties of oil that prevent and/or reduce the exchange of water and gases which disrupts the metabolism or causes acute toxicity that destroy the embryo (Amadi *et al*., 1993). Similarly, hydrocarbon contamination leads to a reduction in nutrient release and soil moisture, thus affecting germination. Osuagwu *et al*. (2015) has observed a significant reduction in the emergence rate of *Cajanus cajan, Vigna subterranean,* and *Phaselous vulgaris* when grown in spent engine oil contaminated soil. Ismail *et al*. (2019) reported a 50% and 80% reduction in the germination of bambara nut and cowpea seedlings respectively, using 10% v/w used engine oil. In a comparative study by Baek *et al*. (2004), the germination of cowpea was significantly reduced whereas corn was entirely unable to germinate in 5% (w/w) oil-contaminated soil. Studies by Wang *et al*. (2017) have also reported a number of harmful effects of petroleum-polluted soil including inhibition of seed germination.

The lowest germination rate was recorded in CR5.0, even though there were treatments with higher hydrocarbon concentrations. This might be attributed to the relative abundance of some hydrocarbon components that are known to positively influence plant growth. Earlier studies by Baker (1970) have shown that adequate concentrations of naphthenic acid stimulate plant growth. This is supported by subsequent findings reported by a number of researchers including Achuba (2006), Adieze *et al*. (2012), and Ismail *et al*. (2014). The growth of plants in hydrocarbon-contaminated soil is often reported with discrepancies because concentrations that are supposed to cause significant effects sometimes stimulate plant growth instead. Earlier studies by Malallah *et al.* (1996) have made similar observations using *Vicia faba* as an indicator of hydrocarbon pollution. The work of Bamidele and Agbogidi (2000) also made similar observations in aquatic macrophytes, thus supporting the present findings.

Shoot lengths decreased with increasing hydrocarbon concentration. Shoot lengths were reduced by ≥ 50% relative to the control. This might be associated with the effect of crude on soil physicochemical conditions and the direct effect on plant growth. Petroleum hydrocarbons impede plant growth by reducing the growth rate, soil fertility, and plants' resistance to pests and diseases (Wang *et al*., 2017). Odukoya *et al*. (2019b) have listed direct toxicity, seeds viability, reduced germination, unfavorable soil conditions, inhibition of organic matter decomposition, and preventing nutrient re-mineralization as the possible causes of plant poor growth in crude oil-contaminated soil. Nitrogen deficiency which is occasioned by an increased C:N ratio due to hydrocarbon contamination has been reported to cause significant reduction in plant shoot length and density, biomass, and may lead to the death of the plant (Merkl *et al*., 2004).

The toxic effect of crude oil on roots is enormous as observed in this study. This could be linked to the ability of the oil to coat root surfaces thereby preventing root elongation and exchange of air and nutrients. Crude oil coats the breathing surfaces of roots, stems, and plant seedlings (Zhang *et al*., 2019). The inability of the roots to exchange air, water and nutrients induce abiotic stress to the plant system which manifests as an anatomical and functional disability. Vartapetian and Jackson (1997) indicated that a shortage in the supply of oxygen has a more direct effect on underground organs such as the roots and seeds which ultimately affect the shoot systems as a result of the negative impact of the stress on the root functions upon which the shoots depend. The effect of oil on roots in relation to water and nutrient uptake has been highlighted elsewhere (Kumar *et al*., 2014; Odukoya *et al*., 2019b). There are studies that support the findings of this research on the reduction of plant root length due to crude oil contamination. Root development of corn and red bean was reported to be acutely reduced in soil contaminated with as little as 1% (w/w) crude oil (Baek *et al*., 2004). Terek *et al*. (2015) also observed a modification of roots morphological structure in the form of reduced main root diameter of alfalfa at 5% (w/w) oil contamination. Achuba (2016) has also observed an inhibition of mitotic activity in the root meristems of 4-day-old cowpea seedlings grown in 1% (v/w) crude oil concentrations.

Leaves number and length were also affected respectively. The highest effect was observed in 10% v/w hydrocarbon concentration. The toxicity was in the order of CR2.5 < CR5.0 < CR7.5 < CR10. Hydrocarbon toxicity depends on the degree of contamination and type of plant species (Emengini *et al*., 2013). The plants were also characterized by yellowing of leaves, senescence, and abscission as the experiment approached completion except in the control. Sufficient evidence suggests that plants adapt to crude oil contamination by reducing the amount of auxin (the main growth activator) and stimulating the increase in the amount of abscisic acid as observed in sedge (*Carex hirta L.*) leaves (Terek *et al*., 2015). Disturbances of major physiological processes within plants systems due to hydrocarbon contamination are common; and manifest in the forms of morphological, pigment, chlorotic and necrotic foliar patterns alterations. Previous studies by Kekere *et al*. (2011) observed complete defoliation in cowpea plants grown 4 – 16% oil-in-soil treatments after 4 weeks period. They also observed that crude oil contamination at all intensities resulted in a significant reduction in leaf number when compared with the control treatment in a concentration-dependent manner. Studies by Odukoya *et al*. (2016) noted that crude oil altered stomatal conductance - a measure of the rate of diffusion of carbon dioxide (CO_2) into leaves for photosynthesis, and water loss via transpiration; growth, yield, and composition of the green leafy vegetables. These findings are in accordance with Ali (2019) who observed that hydrocarbons reduce the growth and yield of crops even at low concentrations after an oil spillage.

In this study, a weight loss of > 60.0% was recorded. The highest reduction in the plant weight was observed in CR10 where 73.09% and 86.13% of wet and dry weight were recorded respectively. Previous studies have demonstrated a reduction in plant fresh and dry weight when planted in crude oil-polluted soil. Petukhov *et al*. (2000) have made similar observations in different plants *Avena sativa, Secale cereale,* and *Hordeum vulgare*. The work of Chuku *et al*. (2018) has made a similar observation in *Solanum melongena, Phaseolus vulgaris, and Cucumis sativus* which is in accordance with the present study. Gross reduction in the plant weight could be correlated (r_s = 0.984; p = 0.026) to the low chlorophyll contents observed in the plants' leaves which plays a critical role in plant primary production and biomass generation. This is in accordance with the findings of Han *et al*. (2016) who observed a decrease in photosynthetic functioning due to a sharp decline in chlorophyll contents of *Amorpha fruticosa* in hydrocarbon impacted soil. Baruah *et al*. (2013) also observed a great impact of crude oil contamination on chlorophyll content and weight of experimental *Cyperus brevifolius*. Based on this study, it is established that hydrocarbons exert a toxic effect on the growth and performance of *Vigna unguiculata* in a concentration-dependent manner, and plants in CR2.5 and CR7.5 were more resilient. The low toxicity observed in these two treatments could be attributed to low hydrocarbon concentration and plant density respectively. This agrees with the findings of Hazaimeh *et al*. (2019) who observed that high plant densities mitigate the adverse effects of crude oil contamination on plant growth which eventually promote the phytoremediation of soil contaminated with hydrocarbon pollutants.

In this study, only plants in control were able to produce flowers, pods, and seeds. For plants grown in contaminated soil, the yield parameters were not observed completely irrespective of the contamination level. This finding is not without precedence as some couples of investigations claimed similar results. Studies by Kekere *et al*. (2011) reported that there was no crop yield recorded in cowpea grown in contaminated soil due to plant mortality especially at 8% and 16%w/w oil concentration. Even at lower crude oil concentrations, significantly lower yields were observed compared to those in control. In similar research by Okonokhua *et al*. (2007) poor crop yield was observed in maize grown in engine oil-contaminated soil. Similar findings were also reported by Chuku *et al*. (2018) in beans, cucumber, and garden eggs. There is sufficient evidence that the quality of agricultural produce can be influenced by environmental stress like hydrocarbon contamination which can be linked with the different physiological responses including yield which is the most palpable effect of abiotic environmental stress on agricultural produce (Wang and Frei, 2011).

Reduction in plant growth and performance observed in this study was strongly linked to the effect of hydrocarbons on nutrients bioavailability among other factors. Hydrocarbon compounds limit plants' absorption of essential nutrients for growth and development. It has been established that hydrocarbon compounds react with inorganic nitrogen and phosphorus, limiting the nitrification and removal of phosphoric acid, thereby preventing nitrogen and phosphorus absorption by plants (Liao *et al*., 2015). For effective growth and development, photosynthesis in plants requires an adequate supply of N and P. Earlier studies by Ordinioha and Brisibe (2013) have reported that hydrocarbon contamination caused a 36% reduction in the ascorbic acid content of vegetables and a 40% decrease in crude protein content of cassava and 60% reduction in household food security. Ani *et al*. (2015) have observed poor crop yield as one of the major consequences of crude oil pollution in Delta Central Zone, Delta State, Nigeria. Studies by Mohamadi *et al*. (2016) also observed that hydrocarbon contamination caused significant vegetation loss in Niger Delta, Nigeria.

5. Conclusion

Crude oil contamination greatly affected the germination, growth, and performance of the cowpea plant in this study. Delay in the emergence of sprouts, stunted growth, leaves chlorosis, and abscission was observed especially at higher crude oil concentrations. Plant density has played an important role in alleviating crude oil toxicity to the plant. The cumulative effect of the plants poor growth manifested in low plant biomass and loss of yield which was triggered by reduced leaf number and length, absence of flowers; and wilting. Although plant growth in crude oil polluted soil provides an inconsistent trend, this study was able to elaborate the phytotoxicity using different growth indices at different growth stages, as previous studies were mostly limited to seed germination and stem growth. Considering the crude oil concentrations used in this study and the resultant-induced phytotoxicity, a real oil spill scenario on agricultural land will instigate a food disaster in the affected community. Although climatic and edaphic conditions influence plant growth, spillage of crude oil in large volumes creates an unmanageable condition for plant growth, as evidenced by incidences in places with similar geographical conditions. In order to avoid the repeat of the present situation witnessed in the Niger Delta, adequate preventive and rapid cleanup technologies need to be put in place in the frontier basins. This would immensely prevent imminent adversity and also protect the livelihood of people in the host communities.

Acknowledgments: The authors acknowledge the role of Tertiary Education Trust Fund (TETFUND) Nigeria, for the sponsorship of this research in its 2019-2020 IBR grants.

Conflict of interest: The authors declare that there is no conflicting interest in this research.

References

[1] Achuba, F. I. (2006). The Effect of Sublethal Concentrations of Crude Oil on the Growth and Metabolism of Cowpea (*Vigna unguiculata*) Seedlings. The Environmentalist, 26, 17–20, 2006.

[2] Adegoke, O. S., Ladipo, K. O., Bako, M. D. and Umaru, A. F. M. 2015. Nigeria's Frontier Basins—Unrealized Rift System Hydrocarbon Potential, *Petroleum Systems in Rift Basins, Paul J. Post, James Coleman, Jr., Norman C. Rosen, David E. Brown, Tina Roberts-Ashby, Peter Kahn and Mark Rowan. S*EPM Society for Sedimentary Geology. https://doi.org/10.5724/gcs.15.34.

[3] Adekoya, J., Ola, P. and Olabode, S. (2014) Possible Bornu Basin Hydrocarbon Habitat—A Review. *International Journal of Geosciences*, *5*, 983-996. DOI: 10.4236/ijg.2014.59084.

[4] Adieze, I.E.; Orji, J.C.; Nwabueze, R.N.; Onyeze, G.O.C. Hydrocarbon stress response of four tropical plants in weathered crude oil contaminated soil in microcosms. *Int. J. Environ. Stud. 2012, 69, 490–500*

[5] Ahmadu, J. and Egbodion, J. (2012). Effects of Oil Spillage on Cassava Production in Niger-Delta Region of Nigeria. *American Journal of Experimental Agriculture*, *3*, No. 4.

[6] Akpokodje, J. and Salau, S. (2015). Oil pollution and agricultural productivity in the Niger Delta of Nigeria, Environmental Economics, *6*(4), 68-75.

[7] Akpoveta, O.V.; Egharevba, F. and Medjor, O.W., A pilot study on the biodegradation of hydrocarbon and its kinetics on kerosene simulated soil. International Journal of Environmental Science. *Volume 2* (1). 54-66. December. 2011.

[8] Ali, W. A. (2019). Biodegradation and phytotoxicity of crude oil hydrocarbons in agricultural soil. *Chilean Journal of Agricultural Research*; *79* (2):266–277. doi:10.4067/s0718-58392019000200266.

[9] Amadi, A., Dickson, A.A. and Maate, G.O. (1993) Remediation of oil-polluted soils: Effects of Organic and inorganic nutrient supplements on the performance of maize (Zea mays L.*). Water, Air and Soil pollution,* 66,59-76.

[10] Amarachi P., Kabari S. A review of the threat of oil exploitation to mangrove ecosystem: *Insights from Niger Delta, Nigeria, Global Ecology and Conservation, Volume 22, 2020*, e00961, https://doi.org/10.1016/j.gecco.2020.e00961.

[11] Ani, A.O., Chikaire, J.U., Ogueri, E.I. and Orusha, J.O. (2015) Effects of Oil Spillage (Pollution) on Agricultural Production in Delta Central Agricultural Zone of Delta State Nigeria. *International Journal of Environmental Sciences Vol. 4 No. 2. 2015. Pp. 75-80.*

[12] APHA, 1989. Standard methods for the examination of water and wastewater. *17th Edition. American Public Health Association, Washington, DC.*

[13] Arellano, P., Tansey, K., Balzter, H. and Tellkamp, M. (2017). Plant Family-Specific Impacts of Petroleum Pollution on Biodiversity and Leaf Chlorophyll Content in the Amazon Rainforest of Ecuador. *PLoS ONE; 12(1): e0169867. doi:10.1371/ journal.pone.0169867*.

[14] Babatunde, B.B., Zabbey, N., Vincent-Akpu, I.F., Mekuleyi, G.O., 2017. Bunkering Activities in Nigerian Waters and Their Eco-Economic Consequences. *The Political Ecology of Oil and Gas Activities in the Nigerian Aquatic Ecosystem. Elsevier* Inc. https://doi.org/10.1016/B978-0-12-809399-3.00026-4.

[15] Baek, K.-H., Kim, H.-S., Oh, H.-M., Yoon, B.-D., Kim, J., & Lee, I.-S. (2004). *Effects of Crude Oil, Oil Components, and Bioremediation on Plant Growth. Journal of Environmental Science and Health, Part A, 39(9), 2465–2472.* doi:10.1081/ese-200026309.

[16] Bamidele, J.F. and Agbogidi, O.M.: 2000, 'Toxicity of odidi petroleum oil and its water-soluble fraction on three aquatic macrophytes,' *Nigerian Journal of Science and Environment 2*, 113–121.

[17] Baruah P, Baruah PP, Deka S (2013). Removal of hydrocarbon from crude oil contaminated soil by Cyperus brevifolius Rottb. *Bulletin of Environment Pharmacology and Life Sciences*; *2* (6): 123- 130.

[18] Chindah, A.C. and Braide, A.S. (2000). The Impact of Oil Spills on the Ecology and Economy of the Niger Delta. *Proceedings of the workshop on Sustainable Remediation Development Technology held at the Institute of Pollution Studies, River State University of Science and Technology*, Port-Harcourt, 2000.

[19] Chuku, O.S., Ekweozor, I. K. E. and Jaja, E. T. (2018). Effects of Crude Oil on Fresh and Dry Weights of *solanum melongena, Phaseolus vulgaris,* and *Cucumis sativus* Amended with Leaf Litter and Hydrogen Peroxide. *International Journal of Science and Research; 7(12):1364-1368.*

[20] Diab, E. (2008). Phytoremediation of Oil Contaminated Desert Soil Using the Rhizosphere Effects. *Global Journal of Environmental Research* 2 (2): 66-73.

[21] Duke, N. C. (2016). *Oil spill impacts on mangroves: Recommendations for operational planning and action based on a global review. Marine Pollution Bulletin, 109(2), 700–715.* doi:10.1016/j.marpolbul.2016.06.082.

[22] Egbe, R.E. and Thompson, D. (2010). Environmental Challenges of Oil Spillage for Families in Oil Producing Communities of the Niger-Delta Region*, JHER, 13,* 24-34.

[23] Emengini E.J., Ezeh F.C. Chigbu N. Comparative Analysis of Spectral Responses of Varied Plant Species to Oil Stress. *International Journal of Scientific & Engineering Research. 4* (6), 2013.

[24] Fingas, M. F., Fieldhouse, B., Sigouin, L., Wang, Z., & Mullin, J. V. (2001) "Dispersant effectiveness testing: laboratory studies of fresh and weathered oils". *Proceedings of the 24th Arctic and Marine Oil spill Program Technical Seminar.* 551-566.

[25] Food and Agriculture Organization, FAO (2018). Outcome Document of the Global Symposium on Soil Pollution. *Held on 2 - 4 May 2018 at FAO Headquarters, Rome, Italy*.

[26] Hamza, H. and Hamidu, I. (2012). Hydrocarbon Resource Potential of the Bornu Basin. Northeastern Nigerian. *GLOBAL JOURNAL OF GEOLOGICAL SCIENCES VOL, 10,* NO.1, 2012 71-84.

[27] Han, G., Cui, B. X., Zhang, X. X. and Li, K. R. (2016). The effects of petroleum-contaminated soil on photosynthesis of *Amorpha fruticosa* seedlings. *International. Journal Environmental Science and Technology; 13:*2383–2392. DOI 10.1007/s13762-016-1071-7.

[28] Hazaimeh, M., Almansoory, A. F., Mutalib, S. A. and Kanaan, B. (2019). Effects of plant density on the bioremediation of soils contaminated with polyaromatic hydrocarbons. *Emerging Contaminants*; *5*: 123-127. https://doi.org/10.1016/j.emcon.2019.03.001.

[29] Ibrahim, S. and Nafi'u, S. A. (2017). *In-vitro* Effects of Tannery Effluents on Seed Germination and Growth Performance of Maize (*Zea mays*), Spinach (*Spinacia caudatus*), and Lettuce (*Lactuca sativa*). *International Journal of Applied Biological Research; 8*(1): 160–173.

[30] Ilozobhie, Anthony, Reassessing the Hydrocarbon Potential of Bornu Basin through Electrofacies and Depositional Environment Analysis (2018). *Asian Journal of Applied Science and Technology (AJAST) (Open Access Quarterly International Journal) Volume 2,* Issue 3, Pages 274-283, July-September 2018, Available at SSRN: https://ssrn.com/abstract=3522071 or http://dx.doi.org/10.2139/ssrn.3522071.

[31] Inoni, E.O., Omotor, D.G. and Adun, F.N. (2006). The Effect of Oil Spillage on Crop Yield and Farm Income in Delta State, Nigeria, *Journal of Central European Agriculture*, *7*(1), 41-48.

[32] Ismail, H. Y., Ijah, U. J. J., Riskuwa, M. L. and Ibrahim, A. A. (2014). Biodegradation of Spent Engine Oil by Bacteria Isolated from the Rhizosphere of Legumes Grown in Contaminated Soil. *International Journal of Environment 3(*2):85-97.

[33] Ismail, H. Y., Riskuwa-Shehu, M. L., Allamin, I. A., Farouq, A. A., Abakwak, C. S. (2019). Biostimulation Potentials of Vigna Species (L.) in Hydrocarbon Impacted Soil. American *Journal of Bioscience and Bioengineering; 7*(1): 22-27. doi:10.11648/j.bio.20190701.15.

[34] Kathi S, Khan AB. Toxicity of spent oil contaminants on Dolichos lablab l. and Abelmoschus esculentus. *The Ecoscan* 2011;*4(*1):133-136.

[35] Kekere, O., Ikhajiagbe, B. and Apela, B. R. (2011). Effects of crude petroleum oil on the germination, growth, and yield of *Vigna unguiculata* Walp L. *Journal of Agriculture and Biological Sciences Vol. 2*(6).158 – 165.

[36] Kumar, S.P.; Minhas, P.S.; Govindasamy, V.; Choudhary, R.L. Influence of moisture stress on growth, development, physiological process and quality of fruits and vegetables and its management strategies. *In Approaches to Plant Stress and Their Management; Gaur, R.K., Sharma, P., Eds.; Springer: New Delhi, India, 2014*;. 125–148.

[37] Liao, J. Q., Wang, J. and Huang, Y. (2015). Bacterial community features are shaped by geographic location, physicochemical properties, and oil contamination of soil in the main oil fields of China. *Microbial Ecology*; *70:* 380-389.

[38] Mahmoud, Y. (2017). The state of the Niger delta oil spill. https://borgenproject.org/niger-delta-oil-spill/.

[39] Malallah, G., Afzal, M., Gulsham, S. Abraham, D., Kurian, M. and Dhami, M.S.I.: 1996, 'Vicia faba as a bioindicator of oil pollution, *Environmental Pollution 92*(2) 213–217.

[40] Matemilola, S., Adedeji, O.H., Enoguanbhor, E.C., 2018. Land use/land cover change in petroleum-producing regions of Nigeria. Polit. Ecol. *Oil Gas Activity in Nigerian Aquatic Ecosyst. 257e276.*

[41] Merkl, N.; Schultze-Kraft, R.; Infante, C. Phytoremediation in the tropics—the e_ect of crude oil on the growth of tropical plants. *Bioremediat. J. 2004, 8*, 177–184.

[42] Mullen, C. L., Holland, J. F. and Heuke, L. (2003). Cowpea, Lablab, and Pigeon Pea*. Agfact P4.2.21, First edition* *www.agric.nsw.gov.au*.
[43] Nasir, M. (2001). Production of Lablab in Nigeria. Extension Bulletin No. 206. *Livestock Series No. 23. National Agricultural Extension and Research Liaison Services, Ahmadu Bello University, Zaria.*
[44] Obaje NG, Balogun DO, Idris-Nda A, Goro IA, Ibrahim SI, Musa MK., Dantata SH, Yusuf I, Mamud-Dadi and Kolo IA. (2013). Preliminary integrated hydrocarbon prospective evaluation of the Bida Basin in Northcentral Nigeria. *Petroleum Technology Development Journal.; 3*(2): 36-65.
[45] Obaje, N. G., Aduku, M., and Yusuf, I. (2013). The Sokoto Basin of Northwestern Nigeria: A preliminary assessment of the hydrocarbon prospectivity. *Petroleum Technology Development Journal, 3* (2): 71-86.
[46] Obaje, N. G., Faruq, U. Z., Bomai, A., Moses, S. D., Ali, M., Adamu, S., Essien, A., Lamorde, U., Umar, U. M., Ozoji, T., Okonkwo, P., Adamu, L., and Idris-Nda, A. (2020). A Short Note on the Petroleum Potential of the Sokoto Basin in North-western Nigeria. *Petroleum Science and Engineering*. (*4*):1:34-38. DOI: 10.11648/j.pse.20200401.14.
[47] Odukoya, J., Lambert, R., & Sakrabani, R. (2019). *Understanding the Impacts of Crude Oil and its Induced Abiotic Stresses on Agrifood Production: A Review. Horticulturae, 5(2), 47.* doi:10.3390/horticulturae5020047.
[48] Odukoya, J.; Lambert, R.; Sakrabani, R. Impact of crude oil on yield and phytochemical composition of selected green leafy vegetables. *Int. J. Veg. Sci. 2019a.*
[49] Odukoya, J.O. Influence of Bioremediation on the Chemical and Nutritional Composition of Produce from Crude Oil-Polluted Sites. Ph.D. Thesis, Cranfield University, Cranfield, UK, 2016.
[50] Ojo, O.J., Jimoh, A.Y., Umelo, J.C., *et al.* Organic geochemical and palynological studies of the Maastrichtian source rock intervals in Bida Basin, Nigeria: implications for hydrocarbon prospectivity. *J Petrol Explor Prod Technol* 10, 3191–3206 (2020). https://doi.org/10.1007/s13202-020-00989-z
[51] Okonokhua BO, Ikhajiagbe B, Anoliefo GO, Emede TO. (2007). The Effects of Spent Engine Oil on Soil Properties and Growth of Maize (Zea mays L.). *J. Appl. Sci. Environ. Manage., 11 (3)*:147 – 152.
[52] Ordinioha, B. and Brisibe, S. (2013). The human health implications of crude oil spills in the Niger Delta, Nigeria: An interpretation of published studies. *Nigerian Medical Journal*; 54:10-16.
[53] Organization of the Petroleum Exporting Countries (2019). OPEC Share of World Crude Oil Reserves. *OPEC Annual Statistical Bulletin*. Available at: https://www.opec.org/opec_web/en/76.htm.
[54] Organization of the Petroleum Exporting Countries (2021). Nigeria Facts and Figures. *Annual Statistical Bulletin*. Available at: https://www.opec.org/opec_web/en/about_us/167.htm.
[55] Osuagwu ES, Olaifa E (2018) Effects of oil spills on fish production in the Niger Delta. *PLoS ONE 13(10): e0205114. https://doi.org/10.1371/journal.pone.0205114.*
[56] Osundina, A., Mustapha, A., & Nzewi, T. (2002). Critical Review of the Oil and Tar Sands of the Dahomey *Basin. 25 Annual International Conference and Exhibition of Nigerian Association of Petroleum Explorationists, Nigeria.*
[57] Paris, A., Ledauphin, J., Poinot, P., & Gaillard, J. L. (2018). Polycyclic aromatic hydrocarbons in fruits and vegetables: Origin, analysis, and occurrence. *Environmental pollution (Barking, Essex: 1987)*, *234*, 96–106. https://doi.org/10.1016/j.envpol.2017.11.028
[58] Petukhov V. N., Fomchenkov V. M., Chugunov V. A. & Kholodenko V. P. 2000. Plant biotests of soil and water, polluted with petroleum and petroleum products. *Applied and Environmental Microbiology. 36*(6): 652-655.
[59] Sam, K., Coulon, F., & Prpich, G. (2017). *Management of petroleum hydrocarbon contaminated sites in Nigeria: Current challenges and future direction. Land Use Policy, 64, 133–144.* doi:10.1016/j.landusepol.2017.01.051.
[60] Sangeetha J, Thangadurai D (2014) Effect of biologically treated petroleum sludge on seed germination and seedling growth of *Vigna unguiculata* (L.) Walp. (Fabaceae). *Braz Arch Biol Technol 57*(3):427–433. https://doi.org/10.1590/S1516-89132014005000011
[61] Schmidt, E. Spill occurrences: A world overview. In Oil Spill Science and Technology: Prevention, Response, and Cleanup, 1st ed.; Fingas, M., Ed.; *Gulf Professional Publishing: Amsterdam, The Netherlands, 2011.*
[62] Shell Petroleum Development Company; SPDC, (2019). Oil Spill Data. https://www.shell.com.ng/sustainability/environment/oil-spills.html.
[63] Tantua, B., Kamruzzaman, P., 2016. Revisiting 'militancy': examining Niger delta. Rev. Afr. Polit. Econ. 1e13, 03(05). http://opus.bath.ac.uk/50368/1/Revisiting_Militancy_and_Control_over_Natural_Resources_5_March_2016_TC_accepted_.pdf
[64] Terek, O., Lapshyna, O., Velychko, O., Bunyo, L., Dovgaiuk-Semeniuk, M. (2015). *Crude Oil Contamination and Plants Journal of Central European Green Innovation 3* (2) 175-184.
[65] Ukpong, Inibehe George & Obok, Ekemini, (2018). "Implications Of Crude Oil Extraction On Agriculture And Livelihood In Oil Producing Rural Communities In Nigeria," *Review of Agricultural and Applied Economics (RAAE), 21*(2):71-77. DOI: 10.22004/ag.econ.281199.
[66] Vartapetian, B.B.; Jackson, M.B. Plant adaptations to anaerobic stress. *Ann. Bot-Lond. 1997, 79 (Suppl. A), 3–20.*
[67] Wang, S., Xu, Y., Lin, Z., Zhang, J., Norbu, N., Liu, W. (2017). The Harm of Petroleum-Polluted Soil and its Remediation Research. *AIP Conference Proceedings 1864,* 020222 (2017); https://doi.org/10.1063/1.4993039.
[68] Wang, Y.; Frei, M. Stressed food—the impact of abiotic environmental stresses on crop quality. *Agric. Ecosyst. Environ*. 2011, 141, 271–286.
[69] Zhang, B., Matchinski, E.J., Chen, B., Ye, X., Jing, L., Lee, K. (2019). *Marine Oil Spills9 Oil Pollution, Sources and Effects. World Seas: An Environmental Evaluation*, 391e406. https://doi.org/10.1016/b978-0-12-805052-1.00024.