



Effect of Crude Oil on Germination Percentage of *Arachis hypogea* and Soil Physico-Chemical Parameters

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ABSTRACT: A study on effect of crude oil on percentage germination of *Arachis hypogea* and selected physicochemical properties of soil was carried out at the botanical garden, of Ignatius Ajuru University of Education. 4kg of soil were artificially polluted with 5 ml, 10 ml, 15 ml of crude oil while 0 ml (unpolluted) served as the control. A completely randomized block design was used for the experimental set-up. The experiment lasted for a period of 8 weeks. The polluted soil recorded low germination percentage in the following order: 15 ml (40.8 %), 10 ml (65.2%) and 5 ml (57.5%) while the highest (100%) was recorded in 0 ml (unpolluted soil). The result for physicochemical properties of soil showed increase in soil total hydrocarbon content (THC), 146.875 in polluted against 9.075 recorded in the unpolluted soil. There were also increases pH, conductivity, total organic carbon (TOC) and total nitrogen and heavy metals (Zinc (Zn), copper (Cu) and lead (Pb)) in the polluted soil when compared to that of the control. Available phosphorus and potassium in soil were reduced in polluted soil.

KEYWORDS: pollution, crude oil, soil, growth, intensity, *Arachis hypogea* L

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I. INTRODUCTION

Oil exploration and exploitation in Nigeria has evolved through decades of history (Olujimi *et al.*, 2011) and its lustrous cover-up of financial benefits has created severe environmental pollution in areas where these oil activities carried out (Egbe, 2010). Nigeria having the largest natural gas reserve and second largest oil reserve in Africa has environmental impacts of exploration and exploitation as an issue of serious concern to Nigerian government regulatory agencies and other environmental organizations. Ghosh and Singh (2005) reported oil exploration and exploitation, accidental and process spillage among others, as major factors responsible for the migration of pollutants into the soil and water ecosystem.

Diverse public unrests resulting from degradation of the environment, an effect of oil exploration have occurred in the Niger Delta region of Nigeria (Inoni *et al.*, 2006) since commercial exploration of petroleum started in 1958 (Okoh, 2003). Crude oil has grown to be mainstay of the Nigerian economy. However, the exploration of petroleum has led to the pollution of land and water ways. Oil waste discharges from refineries, factories or shipping into water bodies such as sea, rivers and streams thus causing great impact on the biota of an environment because it contains poisonous compounds poses danger to plants and animals.

Oil spill pollution has tremendous effects on our environment, ecology, economy, and the society as a whole (Chang *et al.*, 2010). Crude oil spills on land and in water, have been a problem the inception of oil as a source of fuel industrial or domestic purposes (Voulvoulis and Georges, 2015). Gibson and Parales, (2000) stated "that poisonous compounds from crude waste pass through the food web of an area and may be eaten by humans causing bioaccumulation stated "that hydrocarbon contamination of the air, soil, freshwater (surface water and groundwater) especially by petroleum aromatic hydrocarbons (PAHs) has drawn public concerns because many PAHs are toxic, mutagenic, and carcinogenic." One maybe right to say that there is a relationship between human health and environmental health. This is because human health is correlated to the observed changes in health with respect to changes in the condition of the environment. (McMichael and Woodruff, 2005). In a nutshell, "human health" might be attributed to "environmental health. Other consequences of crude oil

pollution reported are diseases in human. Oil pollution on land destroys soil fertility, useful microorganisms and impedes proper plant growth. Some farmers in these areas have been forced to abandon their farmlands. This has grave effects on food production and livelihoods of the people (Emmanuel and Gordon, 2006).

(Onwurah, *et al.*, 2007 ; Akujuru, 2014) stated that “a reasonable percentage of oil that spills on dry land between 1978 and 1979 in Nigeria, affected farm- lands in which crops such as rice, maize, yams, cassava plantain were cultivated.” Also, (Essien and John, 2010; Onwurah, *et al.*, 2007) stated that “germination and growth of plants is affected by crude oil.” The quantity and type of oil spilled determines the level of impact on soil fertility. Migration of farmers that depend only on agriculture as a source of living out of their original settlement is caused by severe oil spill. This is because the soil is sterilized by petroleum hydrocarbons thus preventing growth of crop. (Onwurah, *et al.*, 2007) said that “the yield of steroidal sapogenin from tuber tissues of *Dioscorea deltoidea* is adversely affected by some hydrocarbons.” Vegetative cover and the mangrove ecosystem depletion in the Niger Delta region of Nigeria is mainly caused by the negative impact of oil spillage residues. Land contamination by crude oil affects certain soil parameters. Pollution causes changes in plant metabolic processes and soil physico-chemical properties that support plant growth (Odjegba and Sadiq 2002).

II. MATERIALS AND METHODS

Description of experimental site

The experiment was conducted at the Botanic Garden, Ignatius Ajuru University of Education Port Harcourt, Rivers State, Nigeria. An area of 10m x 11m was marked out with measuring tape, cleared to ground level.

Sources of materials

Soil for the experiment was collected randomly with a spade from a fallow patch of land within old Biology Laboratory, Ignatius Ajuru University where no accidental or deliberate oil spill has been recorded. Surface soil (0 to 15cm depth) was collected, bulked and mixed together. The soil was analytically characterized. The buckets used as experimental pots were bought from Mile 3 market, Port Harcourt. The crude oil (bonny light) was obtained from Nigerian National Petroleum Corporation (NNPC), Eleme, Port-Harcourt.

Experimental design

4 kg of soil was weighed into each experimental plastic pot with a 10 cm allowance at the top of the bag for proper watering and artificially polluted with 5ml, 10ml and 15ml of crude oil. The pots were also perforated at base for drainage. A total of 28 pots filled with experimental soil and planted with *Arachis hypogea* L were used for the experiment and arranged using completely randomized design (CRD).

Planting of *Arachis hypogea* L

Seed viability was tested using the germination technique in ten (10) petri dishes. This was done by placing ten (10) *Arachis hypogea* seeds each on filter paper saturated with distilled water and covered in a petri-dish.

Planting was done two weeks after soil pollution. 10 seeds of *Arachis hypogea* L were planted into each plastic pot labeled 0 ml, 5 ml, 10 ml and 15 ml respectively. Number of seeds that germinated in each treatment were counted and percentage germination in each treatment was calculated using the formula.

Percentage germination = $\frac{\text{Number of seeds that germinated}}{\text{Number of seeds placed in pot}} \times 100$

The pots were adequately watered at time of planting and in the morning subsequently throughout the period of the experiment.

Soil Sampling

Soil samples were obtained by composite sampling from replicates using 22cm hand-dug soil auger. The collected soil samples were put in small labeled polyethylene bags.

The soil samples were analyzed to determine its Physico-chemical properties. This procedure was undertaken two times in the course of the study; before pollution and after pollution.

Analysis of Soil Characteristic

Soil's physicochemical parameters determined include Total Hydrocarbon Content (THC), concentrations of soil Nitrogen, Phosphorus, Potassium and Heavy metals (Lead, Zinc and copper). The soil samples were analyzed twice, initial (prior to pollution) and final (after the crops have been harvested).

Soil Analytical Methods used

Soil Texture

Soil particle size analysis was determined by the Bonyoucos (1951), Hydrometer method. 51g of fine textured air dried soil was weighed and placed in the baffled cup. The cup ½ full, was filled with distilled water and 50ml of sodium hexametaphosphate reagent. This was allowed to stand overnight. The cup was placed in stirred until soil aggregates were broken down (6 minutes for sands, 10 minutes for “light” heavy sandy loams and 15 minutes for other soils). The suspension was transferred to a 100ml cylinder and filled to the lower mark with distilled water while the Bouyoucos hydrometer was in suspension. To determine the sand percentage the weight of sand in the sample was obtained by subtracting corrected hydrometer reading from the total weight of the sample. The percentage sand was calculated by dividing the weight of sand by the weight of the sample and multiplied by 100. Similarly to determine percent clay in the sample, the correct hydrometer reading after the silt and sand had settled out suspension represents the grams of clay in the sample. The percent of clay was calculated by dividing this weight by the weight of the weight of the sample and multiplied by 100. Finally, the silt percentage in what was calculated by subtracting the sum of the percentages of sand and clay from 100. Then, the textural class name or texture of the soil was determined with the United States Department of Agriculture (USDA) textural triangle.

Soil pH Determination

Soil pH in water (1:1 soil/water ratio)

20g of air-dried soil was weighed into a 50ml beaker, 20ml of distilled water was added and allowed to stand for 30 minutes and then stirred with a glass rod. The electrodes of the pH meter were inserted into the partly settled suspension and the pH measured.

Soil pH 1m KCL (1:1 soil-to-solution ration)

20g of soil and 20ml of H₂O were equilibrated for 30 minutes with occasional stirring. pH was determined as “soil pH measured in 1m KCL”. The pH of the soil was determined by the electrode pH meter in both distilled water 1:2:5 and 1m KCL (1:1).

Determination of soil conductivity

The soil conductivity was measured using a conductivity meter. Also, the conductivity of the fresh (wet conductivity) and dried (dry conductivity) soil samples were performed. This was also to detect any possible differences. The soil samples were also mixed in a ratio of 1:1 with distilled water before taking the readings. The conductivity was measured in Omhos/cm.

Total Organic Carbon (TOC)

Total organic carbon (TOC) in percentage was determined by the wet combustion method of Walkley and Black (1934). One (1) gramme of finely ground representative sample was weighed in duplicates into beakers. Ten (10ml), milli-liter of potassium dichromate solution was accurately pippered into each beaker and swirled gently to wet the soil sample completely. This was followed by the addition of 20ml to effect complete oxidation and allowed to stand for 10 minutes before diluting with 300ml of distilled water. Twenty five ml of 0.5N ferrous ammonium sulfate was then added and titrated with 12.4N potassium permanganate under strong light. The percentage organic carbon in the soil was calculated.

Using the formula, $\frac{N(T-B)}{W} \times 0.390$; where N= Normality of Kmn₀₄

T=volume of kmn₀₄ used in the titration of soil sample

B = volume of kmn₀₄ used in the titration of the blank samples. W=weight of sample sued.

Total Hydrocarbon Content (THC)

The hydrocarbon content of the soil was determined by shaking 5g of a representative soil sample with 10ml toluene and the oil extracted determined by measuring the absorbance spectrophotometrically at 420nm using a spectronic 20. A standard curve of the absorbance of different known concentrations of hydrocarbon concentration in the soil sample was measured in 0g/g after reference to a standard curve and multiplying by the appropriate multiplication factor (Odu *et al.*, 1985). Nitrate was determined by the Brucine Method. Available phosphorus in the soil was determined by Bray and Kurtz method (1945). Nitrogen was analyzed by the macrokjeldahl method of Bremmer and Mulvaney, (1982).

Phosphate (Available Phosphorus)

Available phosphorus in the soil was determined by Bray and Kurtz method (1945). 2.28g of soil was weighed into an extracting bottle. Twenty (20ml) of Bray No. 1 extracting solution was added (0.025N HCL +0.03N NH₄F) and shaken immediately for one minute. Ten^{ml} (10^{ml}) of filtrate was pitted into 50ml volumetric flask and diluted to about 50ml with distilled H₂O, then 4ml of ascorbic acid solution was added.

This was allowed for at least 30 minutes for full colour development before reading from spectronic 20 at 660nm. Phosphorus concentrations was expressed in glg after reference to a standard curve.

Nitrogen Determination

Nitrogen was analyzed by the macro-kjeldahl method of Bremmer and Mulvaney, (1982).

10g of soil sample (air dried and ground to pass 0.2mm sieve) weighed accurately into a dry 500ml macro kjeldahl flask and 20ml of distilled water was added. The flask was swirled for a few minutes, and allowed to stand for 30 minutes. 1 tablet of mercury catalyst and 10g of K₂SO₄ were added. Then, 30ml of conc. H₂SO₄ was added through an automatic pipette and the flask heated cautiously at low heat on the digestion stand until the water was removed and frothing ceased. The heating was increased until the digest clears. The mixture boiled for 5 hours under regulated heating and the H₂SO₄ condensed about middle of the way up the neck of the flask. The flask was allowed to cool and 100ml of water was slowly added to the flask.

The digest was carefully transferred into another clean macro kjeldahl flask (800ml capacity) with all sand particles in the original digestion flask retained. The sand residue was washed with 50ml of distilled water 4 times and the aliquot transferred into the same flask. Thereafter, 50ml of H₃BO₃ indicator solution and the 800ml kjeldahl solution was poured through the distillation flask by opening the tunnel stopcock with the immediate commencement of distillation. The condenser was kept cool (below 300 by allowing sufficient cold water to flow through, heat was regulated to minimize frothing and prevent such-back 150ml distilled was collected and the distillation stopped. The NH₄N in the distillate was determined by titrating with 0.01m standard HCL with a 25-ml burette graduated at 0.01m intervals. The colour change at the end point is from green to pink.

Calculation

$$\%N = \frac{TXMX14X100}{wt.ofsoilused}$$

Where

T = titre value

M = molarity of HCL.

Determination of Potassium in Soil using the Flame Photometer.

Principle: When atoms of an element are heated in a flame, some of the heat energy is absorbed by a few of the atoms which become excited i.e. there is a transition by one or more electrons from the ground state to higher energy levels. For each elements there are certain permitted shifts giving rise to a series of lines, each series being characteristic of the element. As a liquid containing the metals are introduced into the flame, the water or solvent is vaporized and only dry salt particles are left behind the light emitted by the flame is collected by the reflector, passes through a lens and heat dispersing glasses which protect the head-sensitive photocell. The optical filter isolates the portion of the spectrum appropriate to the element under determination.

The small current generated is passed through amplification circuit to the galvanometer and deflects the mirror. The mirror then turns the light ray according to the magnitude of the electrical energy and this is read on the meters.

Procedure: The instrument supply gas was fully turned on until the burner lighted. The compressed air pump was put on and adjusted until the pressure dial read about 121b/in². The filter plate corresponding to the element to be determined was put in place and the instrument switched on by turning the sensitivity control clockwise until it clicked. The most concentrated of the standard solution (1000m) was used to set the reading at 100 using the sensitivity control. It was then resettled to zero on replacing with distilled water. The standards were read in an increasing order of concentration while setting the meter to zero using distilled water after every successive reading. The samples were then read after the standards and the nebulizer of the burner system was cleaned with distilled water being drawn into the flame and the meter resettled to zero.

Potassium Determination

Stock solution: the stock solution was prepared by using the method discussed under sodium.

$$\begin{aligned} \frac{Kcl}{K} &= \frac{74.56}{39.102} \\ &= 1.9068\text{gm} \div 10 \\ &= 0.1907\text{gm} \end{aligned}$$

0.1907gm of potassium chloride was weighed and dissolved in 1 litre of distilled water to give 100pm of potassium. The working standard was further diluted to produce a range containing 0-10ppm k and made up to volume in a 100ml flask.

Results:

Wt. of sample	-	2gm
Aliquot	-	2ml

Dilution factor (D.F) - 50ml
Solution Volume - 100ml

Determination of Lead (Pb), Copper (Cu) and Zinc (Zn)

Copper (Cu), and Zinc (Zn) were determined after digestion of soil sample with a mixture of hydrofluoric, perchloric and hydrochloric acids.

Procedure

Wet 0.5g sample of the ground soil was placed in 50ml platinum crucible, containing 5ml of Hf and 5ml of HCl₄. The crucible was placed on sand bath and covered with the platinum lid. The crucible was heated to 200-255⁰c to evaporate the content to dryness. The crucible was removed and allowed to cool after which 5ml of 6m Hcl and 5ml of distil water were added. The sample was transferred to a 100ml volumetric flask and the content diluted. Later the concentrations of the elements were analyzed using Atomic Absorption Spectrophotometric procedure (AOAC, 1990).

III. RESULT

The result for percentage germination of *Arachis hypogea* is shown in the figure 1 below. Germination in the control (unpolluted soil) was prompt while that of polluted soil was delayed. Also, germination was observed to be higher in unpolluted soil than in polluted soil.

The result for physico-chemical properties of unpolluted and polluted soil are summarized below in table 1 and table 2 respectively. There was reduction in sand and clay of polluted soil when compared to the control (unpolluted) and increase in silt. Polluted soil was observed to have less moist, poor aeration and compacted. The result showed increase in soil pH, conductivity, total hydrocarbon content (THC), total organic carbon (TOC) and total nitrogen in the polluted soil when compared to that of the control. There were reductions in available phosphorus and potassium while Zinc (Zn), copper (Cu) and lead (Pb) increased.

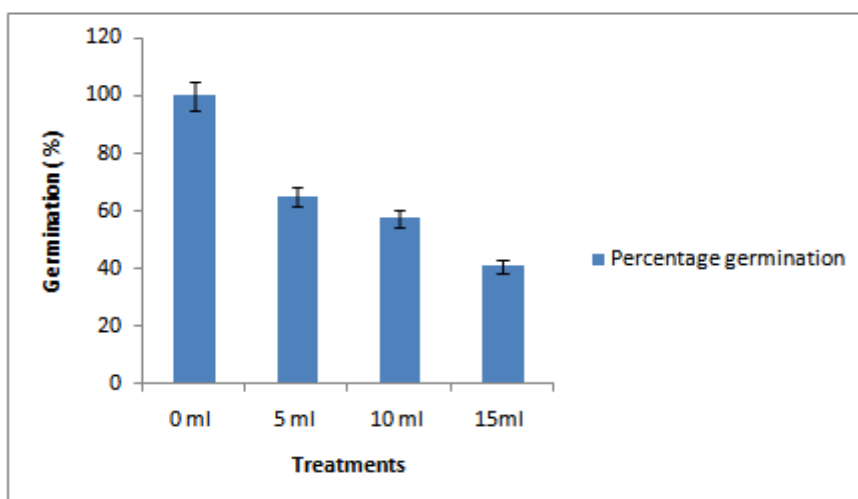


Fig. 1: Effect of different treatments of crude oil on germination of *Arachis hypogea*.

Soil Physio-Chemical characteristics of unpolluted soil

Properties	result	unit
A. Physical		
Texture		
Sand	67.75	%
Clay	14.375	%
Silt	17.875	%
B. Chemical		
PH	5.9625	
Electrical conductivity	415.15	cmol/kg
Organic carbon	13.05	%
Total Nitrogen	9.95	%
Potassium	714	cmol/kg
Total Hydrogen content	9.075	
Phosphorus (P)	123.625	mg/kg
Zinc (Zn)	19.435	mg/kg

Soil Physio-Chemical characteristics of polluted soil

Properties	result	unit
A. Physical		
Texture		
Sand	52.75	%
Clay	11.375	%
Silt	22.875	%
B. Chemical		
PH	6.5625	
Electrical conductivity	666.25	cmol/kg
Organic carbon	23.05	%
Total Nitrogen	12.05	%
Potassium	209	cmol/kg
Total Hydrogen content	146.875	
Phosphorus (P)	93.425	mg/kg
Zinc (Zn)	42.375	mg/kg
Lead (Pb)	0.67	mg/kg
Copper (Cu)	28.5	mg/kg

IV. DISCUSSION

The low germination percentage recorded in the polluted soil may be attributed to the fact that soil physicochemical properties were such as capillarity, aeration, pH, nitrogen and phosphorus contents which are essential for crop germination, growth and development were adversely altered. This result is confirm by the reports of Ogboghodo *et al.*(2004) and Kayode *et al.* (2009). This observation is also in line with other findings that the presence of crude oil in soil reduces seed germination and retards growth of seedlings (Onuh *et al.*, (2008). This is also in agreement with the findings of Ntare *et al.* (2008).

The increase in soil pH and conductivity may have occurred as a result of accumulation of exchangeable bases in the polluted soil which in turn affect stability and availability of nutrients in the soil.

Polluted soil was less moist, with reduced aeration, soil compaction and increased heavy metal accumulation thus the increase in lead, copper and zinc polluted soil. The increase in silt may be attributed to

the compaction of soil particles and soil contamination that adversely affected soil characteristics. This result agrees with the findings in the reported of Kayode *et al.* (2009) and Osuji and Nwonye (2007). This is also in line with the report of Barua *et al.*, 2011; Marinescu *et al.*, 2011 and Abosedede, 2013, that crude oil effects soil physical property by alterations of soil bulk density, soil spore spaces, capillarity and water porosity which in turn lead to impaired soil water infiltration, soil moisture content, water holding capacity and finally contaminated soil becomes oily to feel. Crude oil pollution alters chemical properties of soil. Soil Total Hydrocarbon content higher in the polluted soil, this result confirms that crude oil pollution increases hydrocarbon content of soil. This result is in agreement with the report of Okoro *et al.* (2011) ; Okop and Ekpo (2012) and Oyem and Oyem(2013).The increase in total nitrogen and total organic carbon may be linked with decay (decomposition) of dead plants and animals and the use of carbon by microorganisms during metabolism. Decrease in Phosphorus and Potassium that was observed in polluted soil was also observed and reported by Adefemi and Awokunmi (2009). The decrease in phosphorus may be attributed to the conversion of most available phosphorus to less available forms for easy uptake by plants.

V. CONCLUSION

Petroleum exploration most time is associated with enormous waste generation and oil spillage, causing significant damage on the vegetation of an area. Oil spills have frequently occurred around oil installation, new exploration sites damaging crops and vast area of arable land. As a result, vast arable land is lost when it is polluted with crude oil.Plants perform poorly on polluted soil because in addition to organic materials produced during photosynthesis, plants need a variety of mineral elements. These elements are absorbed from the soil by the root system and transported upward through the xylem and phloem. Plants use these elements to build new parts and to carry on the biochemical activities that take place in their cells. All these elements except nitrogen come from the parent rock, which produces the soil. Some of the mineral elements are regarded as essential and others non-essential element is one without which the plant cannot complete it life cycle, that is, without it normal growth and reproduction cannot occur. It cannot be replaced by another element and the requirement must be direct. For this reason there is changes in plant metabolic processes resulting from changes in soil physico-chemical caused by crude oil pollution. The general conclusion appears to be that germination and growth of seed is adversely affected by pollution of the soil with crude oil. The agricultural and economic implications of environmental degradation resulting from crude oil pollution in the oil – rich Niger Delta cannot be overemphasized as it has left enormous consequences on the people of the area. It has reduced crop yield due to soil infertility and poor farm income. This is a major problem as if affects agricultural production, the main occupation of the Niger Delta people and also threatens food security. Thus, crude oil pollution should be minimally prevented and polluted sites remediated (reclaimed) for cultivation of crops to boost food production.

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