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Germination of African oil bean (*Pentaclethra macrophylla,* Benth.) seeds grown in crude oil polluted soil

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Abstract

Background: An assessment of the germination of African oil bean seeds as affected by crude oil pollution of soil was carried out in Asaba, Nigeria in 2008. Six (0.0, 1.4, 2.8, 5.3, 11.2 and 22.4% by weight per 1.6kg of weight of soil sample) crude oil concentrations in soil served as the treatments. A randomized complete block design was adopted with four replications.
Results: The results showed that no significant differences ($P \geq 0.05$) existed between the *Pentaclethra macrophylla* seeds sown in the unpolluted (0.0%) and those planted in the 1.4% level of oil pollution as regards percentage germination, days to germination and rate of germination. Significant differences ($P \leq 0.05$) were however observed in the germination characteristics of *P. macrophylla* seeds sown in soils as the concentration of oil in soils increased.
Conclusion: The study has established that crude oil pollution of soil has a significant effect of reducing the viability of *Pentaclethra macrophylla* seeds. It is also demonstrated in the study that crude oil application to soil significantly delayed and reduced the rate of germination of *Pentaclethra macrophylla* seeds while the minimum (critical) level of crude oil pollution that *Pentaclethra macrophylla* seeds can tolerate with respect to germination is 1.4% wet weight.

Keywords: *Pentaclethra macrophylla,* Viability, Crude oil pollution, Critical level, Tolerance.

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INTRODUCTION

African oil bean (*Pentaclethra macrophylla*, Benth) which is a tropical tree crop found mostly in the Southern and Middle belt regions of West and Central Africa, belongs to the family leguminoseae and the sub-family of mimosoideae (Aseogwu *et al.,* 2006). It is recognized by peasant farmers for its soil improvement properties and as a component of an agro forestry system (Ladipo and Boland, 1995). Enujiugha and Agbede (2000) and Asoegwu and Ohanyere (2006) identified *Pentaclethra macrophylla* as a minor food supplement. The tree crop is recommended for planting to increase honey production, nectar produced by flowers from this tree is very attractive to bees (Latham, 2008).

According to Enujiugha and Agbede (2000) and Akindahunsi (2004), the seed contains 23-28% protein, the twenty (20) essential amino acids and essential fatty acids. The seeds when cooked, processed and fermented can be used for the preparation of many delicious delicacies including African salad, soups and sausages (Aju and Okwulehie, 2005; Envjiugha and Akanbi, 2005). The plant is used as salt substitute, charcoal, carvings, seed craft, dye, mild poison, medicine (against convulsion, abortion, diarrhea, infertility), wound treatment, lactogenicity, ornamental as well as fencing, timber and structural work (Abbiw, 1990; Tico, 2005; Ikhuoria *et al*., 2006).

The tree grows to about 21m in height and up to 6m in girth (Aseogwu *et al*., 2006). It has a characteristic low branching habit and an open crown, which allows substantial light under its canopy hence it, is used in combination with other food crops on farms and particularly in home gardens in South-eastern Nigeria (Idumah *et al*., 2008). It has a crooked bole with low wide buttresses. The bark is grayish to dark reddish-brown, thin flaking off in irregular patches. The leaves with a stout angular common stalk is 20-45cm long and covered with rusty satellite hairs and consists of between 10 and 12 pairs of stout opposite pinnae (Keay *et al*., 1989). The leaflets, in shape of a parallelogram, are practically glabrous. The flowers creamy-yellow or pinkish white and sweet smelling is crowded in narrow spikes (Keay *et al*., 1989). Flowering occurs twice in a year: January-May and July-December (Ladipo and Boland, 1995). The (fruits) pods are persistent, 35-45cm long by 5-10cm broad, widest at the upper end, rounded at the apex, blackish in colour, very hard, woody, splitting open explosively and valves curling up as well as containing between 5 and 8 flat glossy brown edible seeds up to 7cm long. The wood of the plant is reddish-brown and very hard (Keay *et al*., 1989). Nigeria is a major crude oil and natural gas exporter as well as an important agricultural nation in the West African sub-region (Agbogidi and Eshegbeyi, 2006). Crude oil spillage is a common phenomenon in Nigeria not only in the oil producing areas but also in other areas because of pipeline connections and installations (Edema *et al..,* 2007).

Crude oil pollution has been reported to contaminate terrestrial ecosystems resulting to damage in soil properties, destruction of plants and animals and in most cases leading to death. Although research works have be carried out on crop plants and some forest species (Nwadinigwe and Onwumer, 2003; Onweremadu *et al*., 2005; Agbogidi and Ofuoku, 2005), there is paucity of documented data on the response of *Pentaclethra macrophylla* to crude oil. Such data could be used for environmental impact Assessment (EIA) and rehabilitation studies. The occurrence of this multipurpose species in the forest area of the Niger Delta and the great deal of crude oil spillage in the coastal regions prompted the investigation into the germination of African oil bean seeds grown in crude oil polluted soils. Besides, information on the germination of this economically valued forest species is scarce. This study specifically was undertaken to evaluate the viability of *Pentaclethra macrophylla* seeds as affected by various levels of crude oil pollution.

MATERIALS AND METHODS

The experiment was carried out at the nursery site of the Department of Forestry and Wildlife, Faculty of Agriculture, Delta State University, Asaba, Nigeria in 2008. Asaba is located at latitude 06^0 14'N, and longitude 06^0 49'E of the equator. Asaba lies in the tropical rainfall zone. The rainy season is usually between April and October, with an annual rainfall range of 1505 to1849.3 mm. The mean temperature is 28 ± 6^0C. The relative humidity is 69-80%, and the monthly sunshine is 4.8 hours (Asaba Meteorological Station, 2008).

Fruits of *P. macrophylla* were obtained from the parent tree at Ufoma in Ughelli-North Local Government Area of Delta State. Healthy and viable seeds were selected and sorted out by simple floatation test. Soil sample used was obtained from the Gmelina plantation behind the Departmental nursery site. The soil was air-dried and passed through a 2mm sieve. The crude oil with specific gravity of 0.900g/cm^3 was obtained from the Nigerian National Petroleum Corporation (NNPC), Warri, Delta State. Crude oil pollution levels in soil were 0.0, 1.4, 2.8, 5.6, 11.2 and 22.4% per 1.6kg of soil. These concentrations compare with minimum ambient oil pollution in soils at forest species natural habitats. Seeds of *Pentaclethra macrophylla* were sown directly in the polypots (20/40cm) containing the crude oil pollution and the unpolluted (control) soils. The polypots were watered to field capacity immediately after planting and every other day following the procedure of Agbogidi and Dolor (2007). No fertilizer was used. The trial was arranged in a randomized complete block design (RCBD) with four replications in the nursery for subsequent examination. Parameters measured were germination percentage (%), days to germination and rate of germination.

Composite soil samples were collected from 0-20 cm depth prior to treatment application. Also, at harvest, soil samples were collected from each experimental plot at 0-20 cm depth. These samples were used to determine soil physico–chemical properties. The analysis was carried out at the Nigerian Institute for Oil Palm Research (NIFOR) Benin, Edo State, Nigeria. The particle size distribution was determined by the hydrometer method (Bouyoucous, 1951) while bulk density was by core method (Blake and Hartge, 1986). Soil pH was determined in distilled water using a soil: liquid ratio of 1:1, Electrical conductivity was measured by a conductivity bridge (Chandos Conductivity Model A19 Bridge). Phosphate–Phosphorus was measured in soil extracts by the ascorbic acid method (IITA, 1979; Obi, 1990). Total nitrogen was determined by the regular Macro-Kjeldahl digestion technique (Jackson, 1964).

Table 1. Germination characteristics of *Pentaclethra macrophylla* as affected by crude oil in soil.

Oil in soil % (w/w)	% Germination	Days to germination	Rate of germination
0.0	93.33a	8.2d	4.3a
1.4	86.67ab	9.8c	3.9a
2.8	66.67b	15.6b	3.2b
5.6	40.00c	25.5a	1.9c
11.2	20.00d	28.4a	1.0d

Means with the same letters within the same column are not significantly different from each other at P≥0.05 using the Fishers Least Significant Difference (LSD)

Nitrate-nitrogen was determined by the phenoldisulphonic acid method (Esu, 1999), Organic carbon was measured by the wet combustion method (Walkley and Black, 1934) and converted to organic matter by multiplying the values of organic carbon by a factor of 1.724 following Allison (1965). C/N ratio was calculated by dividing % carbon values by that of the total nitrogen. The data obtained were exposed to analysis of variance while the significant means were separated with the Duncan's multiple range tests using SAS (2005).

RESULTS AND DISCUSSION

No significant difference (P≥0.05) existed between the *P. macrophylla* seeds sown in the unpolluted soil (0.0%) and those planted in the 1.4%

Table 2. Physico-chemical properties of Asaba soil before planting.

Parameters	Asaba
Sand (%)	94.5
Silt (%)	2.1
Clay (%)	3.4
Soil pH	5.60
Organic carbon (%)	0.91
Total N (%)	0.06
Available P (mg/kg)	30.00
Ca^{2+}(cmol/kg)	1.31
Mg^{2+} ‘’	0.16
Na^{+} ,,	0.25
K^{+} ,,	0.17
H^{+} (cmol/kg	0.45
Al^{3+} ,,	0.08
ECEC ,,	2.42
Base saturation (%)	78.10

level of oil pollution (Table 1). Significant differences ($P \leq 0.05$) were however observed in the germination percentage of *P. macrophylla* seeds sown in soils as the concentration of crude oil in soils increased (Table 1). For example, while the germination % of *Pentaclethra macrophylla* seeds sown in 0..0 and 1.4%w/w of oil-polluted soils were 93.33and 86.67 respectively, the germination % of Pentaclethra macrophylla seeds planted in soils polluted with 5.6 and 11.2%w/w of the oil recorded 40.00 and 20.00 respectively. Days to germination of *P. macrophylla* seeds grown in crude oil impacted soil increased as the level of oil pollution in soils increased and the values significantly differed ($P \leq 0.05$) when compared with seeds planted in the control soils (Table 1).The rate of germination of *P. macrophylla* seeds as affected by crude oil soils is presented in Table 1.

While more seeds and rate of germination increased in soils without crude oil treatment and those with the lowest oil treatment (1.4). The rate in soils with higher levels of crude oil significantly reduced at the 5% level of probability (Table 1). The physico-chemical property of Asaba soil before addition of oil to soil is shown in Table 2. Table 3 shows the eeffect of different crude oil levels on some soil physical properties at Asaba location. The chemical nutrient elements of Asaba soil as affected by crude oil in soil is presented in Tables 4a and 4b. Effects of different crude oil levels on soil Total Exchangeable Acidity (TEA), Total Exchangeable Bases (TEB), Effective Cation Exchange Capacity (ECEC) and Base Saturation (BS) at Asaba is indicated in Table 5.

Crude oil pollution at 1.4% w/w did not necessarily affect the germination percentage, days to germination and rate of germination. This observation shows that *Pentaclethra macrophylla* can tolerate crude oil pollution to a certain level. As the concentration of the crude oil in the soil increased, stressors increased the critical level at which the plant can no longer withstand the pollution had been reached.

The non-significant ($P \geq 0.05$) effects of crude oil application on soil in both locations physical properties studied may be due to the relatively low C: N content of the crude oil treatment of the soil sample. Low C.N. content of treatment materials may not have much influence on soil particle size distribution, bulk density and porosity since such amendments are rapidly mineralized to release their nutrient components by soil organisms (Nnaji *et al.*, 2005). However, cultivation of land and soil management practices adopted in the process of crop production may tend to lower the total pore space compared (Song and Bartha, 1990; Brady, 1990; Udo and Ogunwale, 1986) to that usually associated with a decease in organic matter content and as a consequence, lowering of granulation.

Similar results of non-significant influence of crude oil on soil texture, bulk density and porosity had been observed by Asuquo *et al.* (2002). The higher the bulk density, the more resistant the soil offers to the root extension and penetration thereby, reducing the areas it can absorb nutrients from the soil. The increase in other cations apart from exchangeable Na^+ and K^+, with increasing levels of oil, collaborate the findings of Amadi *et al.* (1993). The high concentration of exchangeable Ca^{2+} and Mg^{2+} in soil can be attributed to rapid decay and mineralisation of organic and mineral materials in the soils. These processes lead to the release of cations and trace elements (Nnaji *et al.,* 2005). This observation agrees with the report of Udo and Oputa (1984) who stated that the Mg^{2+} and Ca^{2+} are the most predominant cations in soil because of

Table 3. Effect of different crude oil levels on some soil physical properties at Asaba

Crude oil levels (ml)	Particle size distribution			(%) Textural class	Bulkdensity (Mg/m^3)	Particle density (Mg/m^3)
	Clay	Silt	Sand			
0	3.4NS	2.1NS	94.5NS	sand	1.37NS	2.74 NS
1.2	3.1 NS	0.9NS	96.0 NS	sand	1.36NS	2.72 NS
2.4	2.8NS	0.8NS	96.4 NS	sand	1.35NS	2.70 NS
5.6	3.2NS	0.7NS	96.1NS	sand	1.34 NS	2.69 NS
11.2	2.9NS	0.6NS	96.5NS	sand	1.33 NS	2.67 NS

Means in the same column with NS are not significantly different ($P \geq 0.05$), using DMRT.

Table 4a. Effect of different crude oil levels on some chemical soil properties at Asaba

Crude oil level in soil (ml)	pH	EC (µg/cm)	Total C	Organic Carbon	Total N	No_3	P (mg/kg)	C/N ratio
				%				
0	5.60c	142.5a	0.53c	0.91d	0.060b	4.88a	30.00d	8.83d
1.4	5.75c	88.6c	0.58bc	1.00c	0.058b	4.16b	35.45c	10.00d
2.8	5.71c	84.5cd	0.72b	1.24ab	0.050b	3.12c	41.04b	14.40b
5.6	5.79b	82.4cd	0.96a	1.66a	0.044c	3.00d	46.21a	21.82ab
11.2	6.00a	76.8d	1.01a	1.74a	0.043c	2.92d	40.56b	23.49a

Means in the same column with the same letters are not significantly different ($P \geq 0.05$), using DMRT.

their strong absorption and rapid release into the soil through mineral weathering. Isirimal *et al.* (2003) noted that Ca^{2+} and Mg^{+} occur in association with other minerals.

The observed increase in exchangeable Ca^{2+} and Mg^{2+} contents as a result of crude oil application is in line with the findings of Amadi *et al.* (1993) who noted increases in the cations of soils treated with crude oil. All the values of the exchangeable Ca^{2+} still fall below critical limit (4 cmol/kg^{-1}) for fertile soils (FAO, 1976). Thus they are far below the optimum requirements for agricultural productivity. The critical values of Ca^{2+} are 10 cmol/kg^{-1} soils (FAO, 1976). Exchangeable H^{+} was highest in soils treated with 1.4%w/w of crude oil and least in soils that were treated with the highest volume (11.2%w/w) of crude oil. Exchangeable $A1^{3+}$ decreased with increasing level of oil. These may be due to the reduction of leaching as a result of hydrophobic action. Reduction in K^{+} and Na^{+} may be due to nutrient immobilisation consequent on the formation of complexes in the soil after degradation and uptake (Benka-Coker and Ekundayo, 1995).

The observed increase in the phosphorus content of the crude oil contaminated soil may be due to the increase in soil pH resulting from treatment application. This finding supports earlier report by Bielski and Ferguson (1983) who noted that increasing pH increases phosphorus availability up to a pH of about 5.5 – 6.0 thereafter, phosphorus availability starts to decrease. Siddiqui and Adams (2002) had also recorded increased P with increasing concentrations of diesel hydrocarbons up to a stage and then it declined. This increase in soil pH may be attributed to the accumulation of exchangeable bases (Ca^{2+}, K^{+}, Mg^{2+}, Na^{+}) in the crude oil contaminated soils. This finding is consistent with those of Benka-Coker and Ekundayo (1995), Benka -Coker and Ekundayo (1997) and Ekundayo and Obuekwe (1997). Isirimal *et al.* (2003) posited that high pH favours availability of macronutrients but low one (increasing acidity) makes micronutrients more available and consequently, reduce microbial

Table 4b. Chemical nutrient elements of Asaba soil as affected by crude oil in soil

Oil in soil %(w/w)		Ppm	%	%	Meg/100g Soil						
	pH	P	C	N	Na	K	Ca	Mg	H^4	Al^{3+}	ECEC
Control	6.4^{b}	43.59^{a}	1.25^{d}	0.10^{0}	26.15^{a}	0.36^{a}	22.70^{a}	1.68^{a}	0.20^{a}	0.40^{a}	38.67^{a}
1.4	6.4b	41.67^{ab}	1.33^{c}	0.11^{b}	25.88^{ab}	0.19^{a}	9.68^{b}	1.76^{c}	0.20^{a}	0.40^{a}	38.10^{a}
2.8	6.6^{b}	41.02	1.38^{c}	0.13^{b}	25.88^{ab}	0.18^{b}	8.80^{c}	2.24^{c}	0.10^{b}	0.30^{b}	36.83^{b}
5.6	7.1^{b}	37.12^{c}	1.47^{b}	0.15^{ab}	25.88^{ab}	0.18^{b}	8.30	3.92^{b}	0.10^{b}	0.20^{b}	36.80^{b}
11.2	7.2^{a}	33.62^{a}	2.02^{a}	0.20^{a}	25.43^{b}	0.17^{c}	7.68a	4.64^{a}	0.10^{b}	0.10^{b}	32.82^{c}

Means with different superscripts are significantly different at $P \leq 0.05$ using Duncan's multiple range tests activity. The values measured are not detrimental to crops as high agricultural productivity can be obtained in soils with pH up to 6.5 (Russell and Russell, 1960). The values of available phosphorus fall within the range of 20-100 mg/kg of soil indicating optimum levels for growth of crop plants (FEPA, 2002).

The observed increase in the total carbon content of the soil with increasing concentration of the crude oil may be attributed to the high content of carbon in the oil, which was found to contain about 82.8% oxidisable carbon (Table 5). This could have been converted to soil organic carbon of oil polluted soils. Similar findings have been reported (Benka-Coker and Ekundayo, 1995). This observation also agrees with the findings of Ekundayo and Obuekwe (1997) who noted increases in organic carbon content of oil polluted soils in Southern Nigeria. It may also be related to the slow decomposition rate of the oil treatment by soil organisms since contamination of soil with crude oil might have resulted in poor soil aeration. Oil contamination of soil has been shown to limit normal diffusion processes thereby reducing the availability of the level of some nutrients in the soil (Agbogidi and Ejemete, 2005). The soil organic carbon contents are not above the 2.0% critical levels require for plant growth (FAO, 2002).

The decrease in total nitrogen and nitrate nitrogen with an increase in oil levels may be due to immobilisation of this nutrient by microbes resulting which might have increased in population. Jobson *et al.* (1974) had earlier reported that oil spills on land resulted in an imbalance in the carbon: nitrogen ratio which, if greater than 17:1 in soils resulted in net immobilisation of nutrients by microbes leading to loss of soil fertility. Nutrient immobilisation following oil pollution of soil has also been reported by De Jong (1980) for cereals. The resultant increase in the microbe population would demand more nitrogen and thus the total nitrogen and nitrate nitrogen in the soil would decrease. The decrease with time may also be interpreted to be due to high uptake of nitrogen with increased plant growth. Total N content of soils were below (0.2%) the critical value required for optimum agricultural productions (FMANR, 1989). The observed relatively low C/N ratio indicated that the crude oil was highly degradable. The level of C/N ratio ranged from 8.83 – 23.49. This showed that there was a net mineralisation of nitrogen in the soils as reported by Paul and Clark (1989).

The observed change in electrical conductivity indicated that the application of crude oil affected the ionic stability of the soil, which could have contributed to the decreased conductivity with increasing oil levels. An increase in crude oil concentration increased the soils ionic strength thereby increasing nutrients available in the soils. The relatively higher acidity level and lower organic matter content of soils treated with crude oil may have reduced the available phosphorus content of the soils. High soil acidity facilitates fixation of available phosphorus (Esu, 1999) and Jean-Marc *et al.* (1995) while negative correlation existed between soil organic carbon and phosphorus fixation in the soil (Isirimal *et al.,* 2003). Isirimal *et al.* (2003) also noted that low exchangeable phosphorus values

Table 5. Effects of different crude oil levels on soil Total Exchangeable Acidity (TEA), Total Exchangeable Bases (TEB), Effective Cation Exchange Capacity (ECEC) and Base Saturation (BS) at Asaba

Crude oil Levels (ml)	Exchangeable ions									
	Ca^{2+}	Mg^{2+}	Na^{+}	K^{+}	H^{+}	Al^{3+}	TEB	TEA	ECE	BS (%)
0	1.31b	0.16d	0.25b	0.17c	0.45d	0.08a	1.89c	0.53bc	2.42c	78.10b
1.4	1.34b	0.18d	0.10c	0.15c	0.57b	0.07a	1.77c	0.64b	2.41c	73.44c
2.8	1.65a	0.40c	0.31a	0.14c	0.50c	0.06a	2.50a	0.56bc	3.06b	81.70b
5.6	1.74a	0.56ab	0.44a	0.13d	0.45d	0.04b	2.87c	0.49c	3.36b	85.42a
11..2	1.76a	0.64a	0.38a	0.11d	0.42d	0.02c	2.89a	0.44d	3.33b	86.79a

Means in the same column with same letter (s) are not significantly different (P≥0.05), using DMRT.

could stem from low soil organic matter. The values of the total exchangeable bases, total exchangeable acidity and effective cation exchange capacity did not exceed the critical values suitable for optimum crop productions if other environmental factors are favourable (FAO, 1976; Holland *et al.,* 1989). The results of the soil analyses indicated that crude oil application to soil in Asaba had a highly significant effect on soil chemical properties although no significant differences were observed in the physical soil properties as a result of soil treatment with crude oil.

Germination inhibition following crude oil application to soil could be attributed primarily to seed coating, which adversely affected gaseous exchange biological harm on the seeds caused by the characteristics of the crude oil used as well as the physical water-repellent property. The present study where there was a significant difference in all the germination characteristics studied in *Pentaclethra macrophylla* in both the treated and untreated soils agrees with prior reports of Agbogidi and Eshegbeyi (2006) on *Dacryodes edulis*, Agbogidi and Dolor (2007) on *Irvingia gabonensis*, Siddiqui and Adams (2002) had earlier reported that crude oil endangers the life of seed embryo thereby reducing vital metabolic activities.

The absence of germination observed for seeds sown in the11.2%w/w of crude oil pollution could be due to the reduced biochemical activities and metabolic activities. Seeds that germinated died at the emergence of the hypocotyls or roots. This oil treatment could have negatively affected seed germination due to changes in soil oxygen. Similar observation has been reported for maize (Agbogidi *et al*., 2006).

The reports of Gill *et al*., (1992), Siddiqui and Adams, (2002) and Agbogidi and Ejemete (2005) on *Chromolaena odoratus*, perennial ryegrass and *Gambaya albida* respectively lay credence to this finding. Suggestively, this study has established that crude oil pollution of soil has a significant effect of reducing the viability of *Pentaclethra macrophylla* seeds. The study has also demonstrated that crude oil application to soil significantly delayed and reduced the rate of germination of *Pentaclethra macrophylla* seeds while the maximum level critical of crude oil pollution that *Pentaclethra macrophylla* seeds can tolerate as regards germination is 1.4% w/w. Crude oil pollution at 1.4% w/w did not necessarily affect the germination percentage, days to germination and rate of germination. This observation shows that *Pentaclethra macrophylla* can tolerate crude oil pollution to a certain level.

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