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EVALUATION OF AFRICAN BREAD FRUIT (*Treculia africana* Decene) FOR BIOREMEDIATION IN SOILS IMPACTED WITH CRUDE OIL

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ABSTRACT

A Field and Laboratory study was conducted in 2007 to evaluate the efficacy of *Treculia africana* seedlings for bioremediation of crude oil contaminated soil in Asaba, Delta State Nigeria. The experiment was arranged in a completely randomized design with five treatments (0.00, 2.25, 4.45, 6.75 and 9.00% w/w of 1.4kg of soil). and replicated four times; Results showed that crude oil application to soil exerted effects on the physiochemical parameters of soil in Asaba as well as the plant mineral nutrient elements. The result also indicated that plant height, number of leaves, leaf area and collar girth and dry biomass of the test plant were not significantly ($P \geq 0.05$) affected up to 4.56%. Although higher levels of crude oil treatment significantly ($P \leq 0.05$) affected the growth characters, the study has demonstrated that *Treculia africana* has the potential of remediating soils impacted with petroleum hydrocarbons especially at low concentrations.

KEYWORDS: *Treculia africana,* efficacy, remediation.

INTRODUCTION

Phytoremediation, though relatively new, is a technology that uses plants to remove contaminants from the soil and water (Ubalua *et al.*, 2007). It provides a technique in which vegetation can be managed in expensively and efficiently produce biomass for chemical or energy application (Agbogidi and Bamidele, 2007). Agbogidi *et al.* (2007) maintained that the process is easy and simple and in most cases, environmentally friendly. Since plants are often present at contaminated sites, it is desirable to evaluate how they interact with contaminated soils (Asikong *et al.,* 2006; Nwite and Mbah, 2008). Besides, maintaining the balance of ecosystem, vegetation is aesthetically pleasing and can provide information on the health of the affected site. Vegetation makes a desirable habitat for wildlife. Phytoremediation comprises phytoextraction, phytodegradation and rhizofiltration all of which take place in nature (Mcintoch and Manchew, 1993).

Although the application of microbe technology has been successful with petroleum-based constituents, microbial digestion has net limited success for wide spread residual organic and metal pollutants (Mueller *et al*., 1996; Agbogidi and Bamidele, 2007). Incidence of pollution through metals, oil spillage etc have been reported to negatively influence the environment and the general ecosystem (Agbogidi *et al.,* 2007; Ekeke *et al.,* 2008). Vegetation based remediation could be studied to show potential accumulation, immobilization and transformation of a low level of persistent contaminants, which exist in polluted soils. Bioremediation is advantageous over other forms of remediation mainly because it is natural, cheap and environmentally safe. Bioremediation seeks to degrade or decompose toxic pollutants in the environment into less harmful ones using organisms.

Treculia africana is a member of the family *Moraceae.* It is dioeciously and found in the rainforest zone in Nigeria especially in the eastern parts (Irvine, 1981; Keay *et al.*, 1989). The fruit is very large, yellowish in colour, globose or cylindrical and covered with spiky out growths. It is 20-25cm long, 15-30cm wide and weighs between 3 and 8kg (Mcintoch and Manchew, 1993). A single fruit may contain between 100-500 edible seeds and a seed is 2-4 cm long, 1-2cm thick and the inner endosperm becomes very crispy on roasting or boiling (Bennett and Ngozillilo, 1998). *T. africana* is a multipurpose tree crop that provides both timber and non-timber products e.g. food, medicine, clothing materials, construction materials, pulp for paper making, latex, animal feeds for manufacturing industries. Proximate analysis shows that the seeds of *T. africana* contain protein, fat, iron, ash, calcium, phosphorus, carbon, hydrogen, magnesium, iron, sulphur, crude fibre, carbohydrate, and other minerals including zinc, lead, copper, phytate, oxatate and tannin (Nwoboshi, 1985; Nwanna *et al.*, 2008). *T. africana* is a large tree growing up to 25m high and a bole of 4-6m in girth. The plant flowers between October and February (Okafor, 1981; Krauss, 1993; Salami, 2002). *T. africana* is also an important component of traditional agro-forestry system and they are integrated into mixed cropping systems with yams, other root crops, bananas and some cash crops. It has also been recommended as a promising species for use in home gardens and for intercropping system as a tool for conservation (Agbogidi and Ofuoku, 2007; Enibe, 2007).

Several researchers (Jean-Marc, 1995; Burns *et al.*, 1999) have reported on the use of microbes to remediate oil polluted sites. Information on the use of plants especially tree species for phytoremediation is however scarce. The aim of this study was to evaluate the potential of the seedlings of *Treculia africana* for bioremediation in soils impacted with crude oil with a view to planting the species in oil impacted soil and thus contribute to the general quest for environmental friendly processes. The study was carried out in the nursery site of the Department of

Forestry and Wildlife, Delta State University (DELSU), Asaba Campus: Latitude 6^0 14^1N; Longitude 6^0 49^1E, Nigeria (Asaba Meteorological Station, 2007).

MATERIALS AND METHODS

The study was carried out in the nursery site of the Department of Forestry and Wildlife, Delta State University (DELSU), Asaba Campus: Latitude 6^0 14^1N; Longitude 6^0 49^1E, Nigeria (Asaba Meteorological Station, 2007).

The seedlings were procured from the Forestry Research Institute of Nigeria (FRIN) Ibadan, Oyo State. Bottom perforated polypots (25 x 20cm in dimension) were filled with the 1.4kg of top garden soil from the *Gmelina arborea* plantation of DELSU, Asaba Campus. 0.00, 2.25, 4.50, 6.75 and 9.00% w/w of crude oil per l.4kg of soil served as the treatments. The soil was thoroughly mixed in a bowl with appropriate crude oil levels before each of the polypots was filled up with the oil treated soils and the one without oil treatment. The seedlings of *T. africana* in the nursery were then transplanted into the crude oil treated soils and the unpolluted soils. The polypots were watered to field capacity immediately after transplanting and afterwards, every other day until the end of the trial. The experiment was laid out in a completely randomized design (CRD) replicated four times. Ten polypots made up one treatment which implied 50 polypots per treatment per replicate summing up to a total of 200 polypots for the 4 replications. The set-up was monitored for 11 weeks after transplanting while growth parameters were measured forth nightly with effect from the third week after transplanting (WAT). Parameters measured were plant height (cm), number of leaves, leaf area (cm^2) and collar diameter (cm). Plant height was measured with a meter rule at the distance from the soil level to terminal bud. The number of leaves was determined by counting while the leaf area was determined by tracing the leaves on a graph paper and the total leaf area per seedling was obtained by counting the number of 1cm squares. Collar girth at 2.5cm above the soil level was measured with veneer calipers. At the end of experiment, plants were harvested and separated into leaves, stems and roots after which, they were oven-dried at 85^0 for 22 hours following the procedure of Bamidele *et al.* (2007). These were then ground and packaged for analysis at the Nigerian Institute of Oil Research (NIFOR) where soil physico-chemical analyses were carried out. 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). 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 concentration of the elements in the sample was read by means of an atomic absorption spectrophotometer (Perkin–Elmer Model 403). Exchangeable calcium and magnesium were determined on atomic absorption spectrophotometer while sodium and potassium were determined on flame photometer (Udo and Ogunwale, 1986), ammonium acetate extracts of soil samples were used in these exchangeable bases determination. Determination of exchangeable acidity (H^+ and $A1^{3+}$) was by KCl extraction method (McLean, 1965). Total Exchangeable Bases (TEB) was calculated by adding the values of all the exchangeable cations (Ca, Mg, Na and K). Total Exchangeable Acidity (TEA) was calculated as the sum of exchangeable H^+ and $A1^{3+}$ ions. Effective Cation Exchangeable Capacity (ECEC) was calculated by adding the values of the TEB and TEA. The base saturation (BS) was calculated by dividing the values of TEB by the ECEC and multiplying by 100. Data collected were subjected to analysis of variance while significant means were separated with the Duncan's multiple range tests (DMRT) using SAS (2005).

RESULTS AND DISCUSSION

Pollution of soil with crude oil did not have any significant ($P \geq 0.05$) effect on particle size distribution in clay at all levels of crude oil in soil. Significant reductions (($P \geq 0.05$) were however observed for silt particles in the oil treated soils. The sand particle however, significantly ($P \leq 0.05$) increased with increasing oil level in soils (Table 1). The pH values, %C and %N increased significantly ($P \leq 0.05$ increased with increasing oil level while the cations (Na^+, K^+, Ca^{2+} and Mg^{2+}) phosphorous, exchangeable acidity (H^+, Al^{3+}) as well as the effective cation exchange capacity (ECEC) were significantly ($P \geq 0.05$) reduced as the oil levels increased (Table 2). Plant nutrient elements of *Treculia africana* as affected by oil in soil are presented in Table 3. All the nutrients (P, N, Na, K, C and Mg) showed significant ($P \geq 0.05$) reductions with increasing oil levels. The control without the oil treatment had the highest values for all nutrients and were significantly ($P \leq 0.05$) different when compared with seedlings grown in soils treated with oil.

The performance of *T. africana* seedlings in terms of plant height, number of leaves, leaf area and collar girth are shown in Tables 4, 5 6, and 7 respectively. Although there was a progressive increase in the growth characteristics of the seedlings over time, significant ($P \geq 0.05$) reductions were recorded in all the parameters measured with increasing concentrations of the oil in soil with the exception of seedlings grown at 2.25 pollution levels.

The observed changes in soil physico-chemical properties are consistent with the findings of Atuanya (1987) and Agbogidi and Ejemete (2005) who noted that crude oil application to soils influences the physical properties, such as soil structure, chemical and biological properties of the soil. Agbogidi *et al*. (2007) also reported that crude oil in soil exerts a significant effect on the nutrient status as well as plant nutrient. Crude oil pollution of soil has also been reported to cause reduction in the level of some soil nutrients, immobilization of nutrients as well as a rise to a toxic level of certain elements including iron and manganese (Nicololli and Eglis, 1998; Siddiqui and Adams, 2002).

The observed increase in the pH of the soil is not out of place as more cations could have been released into the media. The increase in soil pH with increasing oil level may be attributed to the accumulation of exchangeable bases in the crude oil polluted soil. This finding is consistent with those of Benka-Coker and Ekundayo (1995) and Ekundayo and Obuekwe (1997). Isirimah *et al.* (2003) posited that high pH favours availability of macronutrients but low pH makes micronutrients more readily available and consequently, reduces microbial activity.

The reduction in the plant parameters measured could be related to the increased effects of the crude oil which could have inhibited cell division, enlargement and expansion and nutrient availability as well as the presence of heavy metals. These could have negatively affected the anatomical and physiological structures of the plant and hence growth reduction (Agbogidi and Eshegbeyi, 2006). Agbogidi and Dolor (2007) had reported that oil pollution has a direct herbicidal and phytotoxic property on tree species. Growth reduction in the presence of crude oil has been reported by various researchers including Agbogidi and Nweke (2005) and Bamidele *et al.* (2007).

This study has demonstrated that *Treculia africana* has the potential to remediate soils impacted with petroleum hydrocarbon especially at low concentrations.

TABLE 1. Particle size distribution of Asaba soil as influenced by crude oil

Oil level in soil (% w/w)	Clay	Silt %	Sand
0.00	3.7a	6.6a	89.7b
2,25	3.7a	6.4b	89.9b
4.45	3.7a	5.6b	90.7a
6.75	3.7a	5.4c	90.9a
9.00	3.7a	5.2c	91.1a

Means with different superscripts are significantly different at $P \leq 0.05$ using Duncan's multiple range tests

TABLE 2. Chemical nutrient elements of Asaba soil affected by crude oil

Oil in soil (ww)	pH	Ppm P	% C	% N	Na	K	Ca	Mg	H	Al	ECEC
0.00	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}
2.25	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}
4.50	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}
6.75	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}
9.00	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

TABLE 3. Plant nutrient elements in *Treculia africana* as influenced by crude oil

Plant sample/oil in soil% ww	P	N	Na %	K	C	Mg
0.00	2.43^{a}	1.36^{a}	0.05^{a}	0.68^{a}	1.99^{a}	1.34^{a}
2.25	2.05^{ab}	1.26^{b}	0.05^{a}	0.52^{b}	1.70^{b}	1.24^{a}
4.50	1.63^{c}	1.19^{c}	0.04^{b}	0.49^{c}	1.63^{c}	0.82^{c}
6.75	1.51_{c}	1.12^{c}	0.04^{b}	0.49^{c}	1.57^{c}	0.57^{d}
9.00	1.05^{d}	0.96^{d}	0.04^{b}	0.45^{c}	1.28^{d}	0.57^{d}

Means with different superscripts are significantly different at $P \leq 0.05$ using Duncan's multiple range tests

TABLE 4. Plant height (cm) of *T. africana* seedlings as influenced by various levels of crude oil in soil

Oil in soil % (ww)	3	5	7	9	11
0.00	22.63^{a}	22.67^{a}	22.69^{a}	22.71^{a}	22.76^{a}
2.25	22.58^{a}	22.59^{a}	22.61^{a}	22.63^{a}	22.65^{a}
4.50	22.55^{a}	22.56^{a}	22.57^{a}	22.58^{a}	22.58^{a}
6.75	22.35^{b}	22.36^{b}	22.37_{b}	22.37^{b}	22.36^{b}
9.00	22.31^{b}	22.33^{b}	22.32^{b}	22.32^{b}	22.31^{b}

Means with different superscripts are significantly different at $P \leq 0.05$ using Duncan's multiple range tests

TABLE 5. Leaf area of *Treculia africana* seedlings as influenced by crude oil in soil

Oil in soil % (ww)	3	5	7	9	11
0.00	12.43^{a}	12.57^{a}	12.68^{a}	12.89^{a}	12.92^{a}
2.25	11.83^{a}	11.87^{a}	11.89^{a}	11.91^{a}	11.93^{a}
4.50	11.66^{b}	11.71^{b}	11.72^{b}	11.72^{b}	11.73^{b}
6.75	10.50^{b}	10.54^{b}	10.53^{b}	10.52^{b}	10.52^{b}
9.00	10.41^{c}	10.43^{c}	10.42^{c}	10.42^{c}	10.41^{c}

Means with different superscripts are significantly different at $P \leq 0.05$ level by Duncan's multiple range tests

TABLE 6. Total leaf area of *Treculia africana s*eedlings as influenced by crude oil in soil.

Oil in soil % (ww)	3	5	7	9	11
0.00	154.14^a	156.51^a	157.33^a	160.12^a	162.43^a
2.25	150.07^a	152.71^a	154.42^a	155.31^a	156.24^a
4.50	119.93^b	121.81^b	122.73^b	121.13^b	121.18^b
6.75	115.77^b	117.52^b	118.25^b	116.21^b	115.20^b
9.00	109.55^c	111.22^c	110.73^c	110.69^c	110.55^c

Means with different superscripts are significantly different at $P \leq 0.05$ using Duncan's multiple range tests

TABLE 7. Collar girth (cm) *T. africana* seedlings as influenced by crude oil in soil**.**

Oil in Soil % (ww)	3	5	7	9	11
0.00	2.33^a	2.41^a	2.58^a	2.62^a	2.70^a
2.25	2.30^a	2.32^a	2.34^a	2.35^a	2.36^a
4.50	2.24^b	2.26^a	2.26^a	2.25^a	2.25^b
6.75	2.19^b	2.21^b	2.20^b	2.20^b	2.19^b
9.00	2.15^b	2.18^b	2.17^b	2.16^b	2.16^b

Means with different superscripts are significantly different at $P \leq$ using Duncan's multiple range tests

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