



ASSESSMENT OF PHYTOREMEDIATION POTENTIALS OF *Albizia procera* (ROXB.) BENTH: A LEGUMINOUS PLANT SPECIES IN CRUDE OIL-POLLUTED SOIL REHABILITATION



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Received: September 21, 2022 Accepted: November 12, 2022

Abstract

Crude oil-polluted soil was evaluated. The experiment was carried out under controlled environment. The variables assessed were germination percentage, plant heights, stem girths, number of leaves and nodulation with standard procedure. The physiochemical properties including; organic matter, pH, nitrogen, phosphorus, potassium, calcium, magnesium and sodium present in the soil used were determined with standard protocol. Results showed that at different contamination level by crude oil (i.e 0, 25, 50, 75 and 100) ml, *Albizia procera* had germination rate of 30%, 27%, 25%, 21% and 18% respectively, mean height of 45.50cm, 42.20cm, 40.15cm, 37.60cm and 30.80cm respectively in 16 Weeks After Planting (WAP), mean girth of 0.45mm, 0.36mm, 0.35mm, 0.35mm and 0.25mm respectively at 16 WAP, number of nodules of 6, 4, 4, 2 and respectively and number of leaves of 11.00, 11.00, 8.00, 6.00 and 5.00 respectively. The soil physicochemical properties were decreasing as the crude oil contamination level increases. With crude oil contamination level increased, the germination rate, height, girth and number of leaves and nodules decreased suggesting the influence of crude oil contaminants on *Albizia procera*. The ability of the species to survive the different crude oil contamination level suggests that it's a potential leguminous species that is preferred for the remediation and rehabilitation of Niger Delta crude oil contaminated soil.

Keywords:

Crude oil-Pollution-soil, *Albizia procera*, leguminous plant species, Phytoremediation,

Introduction

Phytoremediation being a form of bioremediation strategy using life plants, sometimes with soil amendments and related microorganisms around the plant root zone to remove, degrade, extract and detoxify contaminants (natural or anthropogenic) in soils, groundwater, air, sludge and sediments (White *et al.*, 2006), through absorption, translocation or sequestration of organic contaminants and removal them from the soil (Cunningham *et al.*, 1996). The plant usage in remediating contaminated soil and unrestricted groundwater is interesting for many reasons: among which are, plants use sunlight energy for soil remediation; two, plant covers are beautiful to behold; Three, plant parts can be severed and analyzed in the laboratory for the level of remediation; Four, plants contain the contamination area by taken up water from soil; Five, microbes in the root zone can biodegrade a lot of organic contaminants; and lastly, some plants have structures that allow transportation of oxygen to the root zones (Shimp *et al.*, 1993). However, to develop effective strategies in plant remediation, a good understanding of the physical, chemical and biological components that inform the existence of contaminants in the root zone is required. Phytoremediation is an *in-situ* technology that is not destructive but cost effective and uses plants with their accompanied microbes, to clean contaminated soils and is, now, popularly used in environmental and ecological investigations (Nie *et al.*, 2011; Oyedeji and Kayode, 2019;

Oyedeji and Kayode, 2020). It is now a scientific answer to remediate petroleum hydrocarbon contaminated areas (Frick *et al.*, 1999; Tane and Kinako, 2008; Oyedeji *et al.*, 2021). The plants that grow and develop properly in crude oil-contaminated soil are hypothetically good species for the phytoremediation works (Bamidele and Agbogidi, 2006).

The idea behind phytoremediation is environmentally friendly, sunlight-energy driven, and using "nature to cleanse nature" concept. Crude oil contaminated soil remediation sometimes needs soil additives and microorganisms in the rhizosphere zone of the plants to remove, degrade, extract and detoxify contaminants depending on the soil matrix. Plants growing in polluted soil can be supervised and collected and level of contaminant contained measured.

Plant structures allow for movement of oxygen to the root zones which help remediation of contaminants (Shimp *et al.*, 1993). To this end, plants that thrive properly in contaminated soil have ability to phytoremediate the soil (Bamidele and Agbogidi, 2006). Therefore before plants can be developed for possible utilization in remediation of toxicant, knowledge of the physical, chemical and biological inter-relationships of each contaminant in the root zone is required.

Many scientists have carried out effect of crude oil on some plant species like *Abelmoschus esculentus* (Oyedeji *et al.*, 2012), *Vigna unguiculata* and *Zea mays* (Kayode *et al.*, 2009a), *Capsicum annum* and *Lycopersicon esculentum* (Anoliefo and Vwioko, 1995), *Chromolaena odorata* (Anoliefo *et al.*, 2006), *Machaerium lunatus* (Bamidele and

Agbogidi, 2006). But there is paucity of information about phytoremediation potentials of *Albizia procera* in literature. Therefore the aim of this study is to assess phytoremediation potentials of *Albizia procera* (a leguminous plant species) in crude oil-polluted soil.

Albizia procera commonly called silk tree is mostly fast-growing sub-tropical and tropical tree of the family Fabaceae. *Albizia procera* is an open canopy tree, grows up to 30m high and stem of 35cm in diameter; straight or crooked, up to 9m. Bark is usually smooth with pale grey-green, yellowish-brown, yellowish-green or brown. The underbark is greenish and can change to orange at the surface; inner bark is pink or straw coloured; branches terete, glabrous (Gupta, 1992). The species has good survival and grow rapid early in afforestation trials in saline and alkaline soils, which are largely grown in agroforestry systems. Boiled leaves can be eaten as a vegetable in Philippines. The bark can be grounded with flour and eaten during dart. The protein-rich fodder of *A. procera* makes it suitable to feed cattle, goats, buffaloes, camels and elephants. The wood is suitable for pulp and paper production. The pulp can yield up to 50.3% satisfactory when bleached by the process called 'sulphate'. It's good for writing and printing paper with mean fibre length of 0.9mm, mean fibre diameter of 0.021mm. *A. procera* has a large volume of sapwood which is yellowish-white and non-durable. It has hard and heavy heartwood with dark and light bands. It can be seasoned and polished well. The wood is mostly used in industries such as construction, veneer, furniture, cabinet work, agricultural implements, flooring, moulding, carts, cane crushers, carriages, carvings, oars, oil presses, boats and rice pounders (Parotta and Roshetko, 1997). The bark provides tanning materials used for tanning and dyeing in India. The stem secretes a reddish-brown gum in large volume that is similar to gum arabic chemically. Every plant parts show anti-cancer activity. The root contains alpha-spinasterol and a saponin that possesses spermicidal activity even at 0.008% dilution. *A. procera* is a candidate species in African medicines. The seeds have proceranin A (Parrotta, 1984).

A. procera is commonly cultivated for its good soil-binding ability. It is sometimes grown to provide shade for tea and coffee farms. Also can serve as a wind and firebreak. It is common in the rehabilitation of occasionally dry, eroded and impoverished soils. Its ability to survive on dry, sandy, stony and shallow soils makes it fit for afforestation of degraded sites (Singh, 1982). *A. procera* fixes atmospheric nitrogen. Its root easily forms an association with Rhizobium species, enabling it to thrive in nitrogen-deficient soils. It is used as urban forestry species and amenity planting where trees are grown along avenues and in gardens for aesthetic purposes. The species is good in afforestation programme especially in degraded and marginal lands due to its better adaptability. The drought resistance features of this species made it popular in dry zones. *Albizia procera* is a fast growing species having good soil binding capacity with excellent strength to fix atmospheric nitrogen and so enrich the soil hence its use in saline and alkaline soils experimental planting (Gupta, 1992). It is mostly cultivated in gardens and avenues for ornamental purposes.

Materials and Methods

Study sites

The investigations were conducted in the Screen House, Department of Biological Sciences and Central Research Laboratory, Faculty of Science, Niger Delta University, Wilberforce Island, Nigeria. The physicochemical analysis of soil samples were conducted at the Central Research Laboratory, Federal University of Technology, Akure, Nigeria.

Source of experimental samples

The clay-loamy soil used for the experiment was collected at a depth 0 – 10cm from a 5-year fallowed plot in the Research and Experimental Farm, Niger Delta University, Wilberforce Island, Nigeria. Furthermore, the crude oil (Bonny light crude oil) was collected from Oporoma Flow Station of Shell Petroleum Development Company. The plant (*Albizia procera*) was obtained from National Centre for Genetic and Biotechnology (NAGRAB), Ibadan, Nigeria.

Seed viability test

Floating method previously described by Anoliefo and Vwioko (1995) was employed in this study. About 500 seeds of the plant species were soaked in a beaker containing distilled water for 30 minutes using the procedure described by Oyediji *et al*, 2021.

Laboratory experiment- Germination tests of the tree species

Five medium sized plant bags were filled with clay-loamy soil from the Research and Experimental Farm, Niger Delta University, Wilberforce Island, Nigeria. 3000g plant bags were arranged in the greenhouse. The soil was manually contaminated with varying volumes (0, 25, 50, 75 and 100) ml of crude oil and thoroughly mixed with the soil. The varying volume (0, 25, 50, 75 and 100) ml of the oil in soil represents the treatments (uncontaminated, low, average, high and very high contamination) respectively. Samples of 200 g of the contaminated soil were measured out from each of treatment using DTA Series Electronic Scale FED-3000 (Made in China, 2005) and dissolved in 1000ml measuring cylinder (Technico, England) containing distilled water and left for 72hours to soak. The aqueous extracts were filtered and the filtrates were collected in 500 ml conical flasks (Pyrex, England) and well labeled.

25 Whatman No 1 filter paper double layered petri dishes were divided into five crude oil treatments (0, 25, 50, 75 and 100) ml in soil with 5 replicates. 0ml served as control experiment. Ten seeds of the plant were planted in the petri-dish and watered daily with filtrates of the respective treatment at 0700 within 10 days to complete the seeds germination. Sprouted seeds were counted and recorded daily until 10 days after planting. Percentage germination was estimated, according to Kayode and Oyediji (2012):

$$\text{Germination test Percentage (Gt \%)} = \frac{\text{Number of seedlings that emerged/dish}}{\text{Total number of seeds sown}} \times 100 \dots (1)$$

Early growth response of the selected tree species in crude oil-contaminated soil grown in the Greenhouse

Fifty (50) polythene bags, sized 2000cm³ each were filled with topsoil weighing 3000g and were arranged in the Greenhouse. These were divided into 5 groups of 10 polythene bags each, set up in a row. The groups were mixed with different volumes of oil in soil (0, 25, 50, 75 and 100) ml representing the treatments: control (unpolluted), low, medium, high and very high pollution respectively. The treatments and control were moistened for two weeks in 72 hours at 0700 after which 3 viable seeds were planted in each of the bags two weeks after introduction of pollution (2WAP₀). Seedlings were thinned to one/bag at two weeks. Assessment of the growth parameters such as plant height, girth and number of leaves were taken fortnightly for 16 weeks in to investigate their early growth response in contaminated soil. The seedling girth and height were were calculated according to Oyediji *et al.*, 2021 and average of the girths and heights treatments taken accordingly.

The Relative growth rate (RGR) in the treatments and growth suppression (GS) were analyzed at 16 WAP. The method previously described by Kayode and Tedela (2000) was used to determine RGR.

Analysis of the physical and chemical properties of the soil samples

The soil physicochemical parameters was determined following standard procedures as reported in literatures including bulk density, soil organic carbon, organic matter, soil pH (Ibitoye, 2006), moisture content (Osuji and Onojake, 2004), volume of air in soil, soil water capillarity and porosity (Akinsanmi, 1975), soil nitrogen, calcium and magnesium (Anderson and Ingram, 1996), available phosphorous (Bray and Kutz, 1945).

Results and Discussion

The percentage germination of *Albizia procera* contaminated with crude oil is presented in Figure 1. At varying concentration (0, 25, 50, 75 and 100) ml of crude oil on germination of *Albizia procera* was 30% (24 COV), 27% (20 COV), 25% (18 COV), 21% (15 COV) and 18% (12 COV) respectively. *A. procera* in crude oil-polluted soil showed a decreased in germination potentials. The decrease

is determined by the concentration of oil in the extracts from contaminated soil. Based on the germination rate, the findings in this study supports the reports of Osuji *et al.*, (2005); Kayode *et al.*, (2009); Kayode and Oyediji (2012) that have previously found out that crude oil have adverse effect on soil conditions, microorganisms and plants.

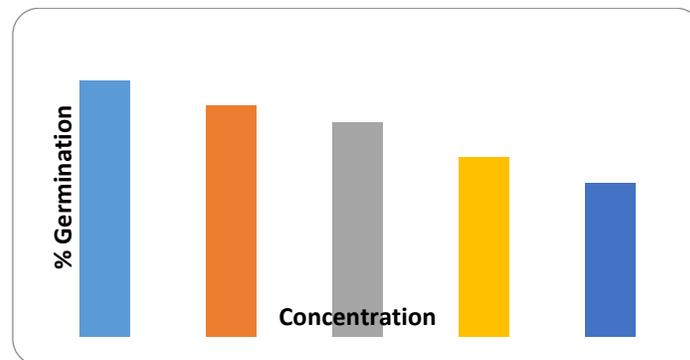


Figure 1: Percentage germination of *A. procera* in crude oil-polluted soil water extracts

Table 1 presents the mean height of *A. procera* grown in crude oil-contaminated soil. Mean height of the species were 2.00±1.65cm, 3.05±1.80cm, 2.80±1.82cm, 2.15±1.72cm and 1.58±2.61cm in the 0, 25, 50, 75 and 100 ml crude oil-contaminated soil respectively at 2 weeks after planting (2 WAP). At 16 WAP, the heights obtained were 45.50±2.71cm, 42.20±2.85cm, 40.15±3.48cm, 37.60±3.80cm and 30.80±3.18cm in 0, 25, 50, 75 and 100 ml crude oil- contaminated soils respectively. The result revealed that the heights obtained are inversely proportional to the soil contamination levels and the percentage growth suppression directly proportional to the soil contamination levels. The height in this findings is in consonant with the works of Anoliefo and Okoloko, (2000) and Kayode *et al.* (2009a, b) that crude oil affects mean height of plants. This study also revealed that the heights obtained inversely related to the soil contamination levels. This suggests that the oil contamination suppressed the growth of the seedlings in the contaminated soil.

Table 1: Mean number of plant height of *A. procera* in crude oil-polluted soil water extracts

Time (WAP)	Plant height (cm)/Crude oil concentration (ml)				
	0	25	50	75	100
2	2.00±1.65	3.05±1.80	2.80±1.82	2.15±1.72	1.58±2.61
4	8.25±2.71	7.83±2.15	6.54±1.78	5.85±1.80	1.72±2.03
6	16.15±3.08	15.60±2.02	12.80±1.43	11.60±2.05	5.50±2.15
8	22.70±1.75	20.70±2.00	17.60±2.17	14.65±2.30	11.50±1.95
10	31.50±2.35	25.80±1.75	23.75±1.80	21.80±1.85	14.55±2.72
12	35.40±1.85	32.50±2.06	30.10±3.08	28.50±1.68	22.50±2.60
14	44.10±2.60	38.20±3.15	35.47±2.50	32.50±2.35	27.00±2.90
16	45.50±2.71	42.20±2.85	40.15±3.48	37.60±3.80	30.80±3.18
$\sum X \pm SD$	205.60±18.70	185.88±17.78	169.21±18.06	154.65±17.55	115.15±20.14
$\Delta H = H_F - H_I$	43.50±1.06	39.15±1.05	37.35±1.66	37.60±2.08	29.22±0.57
RGR	0.22	0.19	0.19	0.20	0.21
GS	0.00	0.073	0.118	0.174	0.323
%GS	0.00	7.30	11.80	17.40	32.30

- RGR = Relative growth rate; GS = growth suppression
- H_I = Initial Height
- H_F = Final Height
- ΔH = Change in height
- \bar{X} = Mean
- (\pm) = Standard deviation

Mean girth of *A. procera* grown in crude oil-contaminated soil is presented Table 2. Seedlings of *A. procera* grown on the contaminated soil had girth values of 0.45±0.04mm, 0.36±0.03mm, 0.35±0.03mm, 0.35±0.03mm and 0.25±0.03mm for 0ml, 25 ml, 50 ml, 75ml and 100 ml respectively at 16 WAP. The findings in this study showed that as concentrations of crude oil in the soil increases, the girth decreased. The biological performance of *A. procera* crude oil-polluted soil observed for leaf numbers and number of nodules are presented in Table 3. At 0, 25, 50, 75

and 100 ml crude oil in the contaminated soil, the number of nodules for *A. procera* were 6, 4, 4, 2 and 0 nodules respectively, while the number of leaves were 11.00, 11.00, 8.00, 6.00 and 5.00 respectively. The decline in number of leaves, nodulation and girth revealed that crude oil has some inhibitory effect on the species grown particularly at higher soil contamination level. Therefore, the action of crude oil in the contaminated soil has hindered some metabolic processes.

Table 2: Mean girth of *A. procera* grown in crude oil-polluted soil

Experimental Time (WAP)	Plant girth (mm)/Crude oil concentration (ml)				
	0	25	50	75	100
2	0.18±0.02	0.13±0.01	0.15±0.02	0.11±0.04	0.11±0.03
4	0.20±0.01	0.14±0.01	0.14±0.01	0.16±0.02	0.10±0.02
6	0.30±0.02	0.20±0.02	0.18±0.01	0.18±0.02	0.14±0.02
8	0.38±0.04	0.30±0.03	0.28±0.03	0.25±0.02	0.18±0.03
10	0.38±0.03	0.31±0.03	0.30±0.02	0.30±0.03	0.21±0.02
12	0.40±0.03	0.31±0.02	0.30±0.03	0.30±0.02	0.25±0.04
14	0.40±0.03	0.35±0.04	0.35±0.03	0.31±0.04	0.25±0.03
16	0.45±0.04	0.36±0.03	0.35±0.03	0.35±0.03	0.25±0.03
$\Delta G = G_F - G_I$	0.27±0.02	0.23±0.02	0.20±0.01	0.24±0.01	0.14±0.00
$\sum X \pm SD$	2.69±0.22	2.10±0.19	2.05±0.18	1.96±0.22	1.49±0.22

Table 3: Mean number of leaf and nodules of *A. procera* grown in crude oil-polluted soil

Mean number	0	25	50	75	100	Mean	Variance	SD
Leaf	11.00	11.00	8.00	6.00	5.00	8.20	7.70	2.77
Nodules	6.00	4.00	4.00	2.00	0.00	4.00	2.67	1.63

The physiochemical characteristics of crude oil contaminated soil that *A. procera* was used for remediation are presented in Table 4. The contaminated soil pH was in the range of 5.02 (at 0 ml) and 4.45 (100ml). But the control soil were in the range of 5.30 – 5.00. The alkalinity and acidity of soils helps in determining the number and kind of soil microbes that change crop residues into useful soil organisms, thereby influencing total soil stability and in turn, the soil aeration and porosity. This observation shows that nitrogen fixing plant such as *A. procera* were slightly more efficient in the elevation of the pH. The result of the soil pH level in *A. procera* showed that contamination levels alter soil pH. The soil pH tends to be acidic in the soil samples which have high level of oil contaminant. However, growing nitrogen fixing plant such as *A. procera* on such soil had a positive influence on it at 16 WAP.

Like pH, the organic matter concentration decreases as the concentration of the crude oil increases i.e 1.56% (0 ml) - 1.49% (100ml), whereas the control had organic matter in the range of 1.83 – 1.61%. Typically, organic matter content of soils is an index of soil fertility. Organic matter is also used as pollution indicator. Organic matter influences nutrients mineralization as carbon content has direct relationship with organic carbon content in the soil; thus bringing oxygen level reduction, which also have effects on microbial metabolism. The decrease in organic matter during soil remediation among the crude oil treated soils as observed in this work shows that the *A. procera* have significant metabolic and absorption abilities with good transport networks that selectively took up the contaminants available in the soil.

The concentration of fertilizer monitoring parameters (nitrogen, phosphorous and potassium) also decreased as the level of the crude oil contamination increased. The concentrations were in the range of 0.39 % (0 ml) – 0.21% (100ml) (nitrogen), 6.75 mg/kg (0 ml) – 5.94 mg/kg (100ml) (phosphorous) and 3.25 mg/kg (0 ml) – 2.78 mg/kg (100ml) (potassium) among the contaminated soil. Basically, macro nutrients measured showed that total nitrogen, phosphorus and potassium were altered in the remediated soil samples at 16 WAP when compared with their controls. The findings showed that *A. procera* can maintain soil nitrogen and available phosphorus balance effectively to a point where bioaccumulation or over reduction that lead to deficiency will not be feasible. Typically, nitrogen fixing plants such as *A. procera* have features in its root systems which produce exudates (energy, carbon, nutrients, enzymes etc) to microorganisms in the root zone (Cunningham *et al*, 1996). The exudates induce and increase population of microorganisms thereby resulted in the degradation of organic contaminants in the root zone. The increased

nitrogen in the contaminated soil would have been used up by the microorganisms in the degradation process. Also, the low level of phosphorus found in the remediated soils could be due to the immobility of phosphorus; it might not have been sufficiently mobilized in the soil to make it available, while the little that was mobilized might have been used up by soil microorganisms rapidly.

Table 4: Physiochemical characteristics of crude oil contaminated soil *A. procera* used for remediation

Parameters	Groups	Crude oil concentration (ml)				
		0	25	50	75	10
						0
pH	Contaminated	5.0	4.8	4.7	4.5	4.4
	Control	5.3	5.1	5.0	5.0	5.0
Organic matter, %	Contaminated	1.5	1.5	1.4	1.3	1.4
	Control	1.8	1.7	1.6	1.6	1.6
% N	Contaminated	0.3	0.3	0.3	0.2	0.2
	Control	0.6	0.5	0.5	0.4	0.4
P, mg/kg	Contaminated	6.7	6.5	6.3	6.1	5.9
	Control	10.	10.	10.	10.	10.
K, mg/kg	Contaminated	3.2	3.0	3.0	2.9	2.7
	Control	3.7	3.5	3.5	3.4	3.2
Na, mg/kg	Contaminated	2.6	2.6	2.4	2.3	2.1
	Control	3.0	2.9	2.8	2.7	2.5
Ca, mg/kg	Contaminated	17.	16.	15.	14.	14.
	Control	26.	25.	24.	23.	23.
Mg, mg/kg	Contaminated	1.8	1.7	1.6	1.6	1.6
	Control	2.8	2.6	2.5	2.4	2.4

The concentration of sodium were in the range of 2.68 mg/kg (0 ml) – 2.17 mg/kg (100ml), calcium were in the

range of 17.60 mg/kg (0 ml) – 14.80 mg/kg (100ml) and magnesium was in between 1.81 mg/kg (0ml) and 1.62 mg/kg (100ml). The results of the soil analyses show that the highest concentrated exchangeable cation was calcium ion, followed by sodium ion and the least cation was magnesium. The result also showed that the *A. procera* reduced the concentrations of the cations, especially the calcium that was impacted most. Soil particles carrying plant nutrients exist within the rhizosphere as ions. The concentrations of the exchangeable cations (calcium, magnesium and sodium) are directly proportional to concentrations of crude oil pollution. Similar trends were observed in past studies. Onyeike *et al.* (2000) established that such exchangeable cations increase in crude oil polluted soil at Ogoni environs. Potassium ion and magnesium ion concentrations observed both in the contaminated and control samples were within the permissible range for low fertility as present in some Nigerian soils. The high calcium ion found in the soil may be occasioned by some anthropogenic activities.

Table 5 shows the effect of physical properties of unpolluted and crude oil-polluted soil used in the experiment. Bulk density of 5.80, 6.40, 6.40, 6.8 and 7.70 g/cm³ were observed in the treatments 0, 25, 50, 75 and 100 ml crude oil-polluted soil respectively. Soil moisture content reduced in the crude oil-polluted soil samples particularly in the 100 ml crude oil-contaminated soil. Similarly, the action of crude oil in the soil samples affects the soil air, 72.50, 38.60, 30.50, 40.40 and 43.60 % were found in 0, 25, 50 75 and 100 ml respectively. Water holding capacity was also reduced in the crude oil-polluted soil.

Table 5: Physical properties of unpolluted and crude oil-polluted soils

Treatment (ml)	Bulk Density(g /cm ³)	Moisture content (%)	Soil air (%)	Water Holding capacity (ml)	Soil porosity(ml)
0	5.8	72	72.5	58.4	86.4
25	6.4	44.5	38.6	50	81.5
50	6.4	40	30.5	34.5	60.4
75	6.8	28.5	40.4	24.1	48.3
100	7.7	18.2	43.6	13.7	32.8

It was observed that the action of crude oil in soil influence the physical properties of such soil. As the contamination levels of the crude oil increased, the soil bulk density, moisture content, soil air, water holding capacity and porosity reduced in the crude oil-polluted soil. This is in consonant with the previous investigation by Ewetola (2013) and Kayode *et al.* (2009b) that reported that the action of crude oil in soil could block pore spaces within the soil and consequently impairs soil aeration, porosity and water infiltration ability which may have deleterious effects on plant growth and productivity.

Conclusion

Oil pollution in soil affects soil physicochemical characteristics like nitrogen, phosphorous, calcium, sodium, magnesium, potassium organic matter adversely. *A. procera* has tolerant ability towards crude oil with regard to germination, height, leaf number and nodulation, which decreased as the concentration of the crude increased. Therefore, *A. procera* is a good candidate plant for phytoremediation of crude oil-contaminated soil in Niger Delta region of Nigeria.

Conflict of Interest: The authors declare that there is no conflict of interests.

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