Eco-Toxicological Implications Of Crude Oil Pollution On *Rhizophora Racemosas* (G.F.W. Meyer)

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Abstract: An experiment was conducted in 2008 in Asaba, Delta State, Nigeria to evaluate the eco-toxicological implications of crude oil pollution on *Rhizophora racemosa* seedlings. Five crude oil levels of crude oil (0.0, 12.0, 18.0, 24.0 and 30.0%) per 1.5kg of flood soils served as the treatments. The experiment was laid out in a randomised complete block design with four replications. The results showed that oil pollution at 18.0, 24.0 and 30.0% significantly affected ($P \geq 0.05$) the seedlings of the test plant in terms of plant height, number of leaves, leaf area, collar diameter and root, growth at the 5% probability level when compared with the seedlings grown in the unpolluted soils and those exposed to 12.0% of the oil. Root growth of the seedlings was significantly reduced ($P \geq 0.05$) with increasing oil levels. At 30.0% oil treatment, root hairs were totally absent. The study has established that *R. racemosa* seedlings tolerated all the crude oil concentrations used. No death was recorded throughout the trail period although significant reductions were noticed with increasing oil levels and this may have implications on the growth and establishment of the red mangrove. Conclusively, *R .racemosa* seedlings conserve as a bio-indicator of pollution and can be recommended for use in area of low levels of pollution for environmental clean-up or bioremediation.

[Agbogidi, O.M. **Eco-Toxicological Implications Of Crude Oil Pollution On *Rhizophora Racemosas* (G.F.W. Meyer).** Nature and Science 2011;9(1):45-49]. (ISSN: 1545-0740). http://www.sciencepub.net.

Key words: Toxic implications, crude oil pollution, *Rhizophora racemosa,* ecosystem

Introduction

Rhizophora racemosa is a wet land tropical tree belongs into the mangrove family Rhizophoracea. *R. racemosa* (Red mangrove) is a_medium tall tree, reaching a height of between 30-50m and between 15 and 35cm in girth (Duke *et a*l., 2006). The tree is mostly multi-stemmed rambling to columnar trees with often distinct above the ground prop roots. The flowers are perfect, containing both male and female parts and flowering period is between August and December in the Southern hemisphere and between February and June in the Northern hemisphere (Duke and James, 2006). The leaves are opposite, simple, bright, green; obvate leathery, margins revolute generally with curve surface. Leaf emergence occurs between November and February and leaf fall occurs chiefly between October and February (Saenger, 2002). The fruits when mature are pear like shaped, elongate, waist constricted and smooth brown surface. *R. racemosa* produces viviparous seeds which are hidden in the fruit (Keyejo et al., 2006). The hypocotyls- the growing seedling from the fruit is narrowly cylindrical, elongated, green, and smooth with irregular small brown lenticels (Saenger, 2002). The hypocotyls fall occurs mainly between November and February (De Lacerda, 2002; Bosire *et al.*, 2005). The seedlings germinate in the mature fruit while the fruit is attached to the parent plant (Bamidele and Agbogidi, 2006; Bamidele *et al.*, 2007).

Rhizophora racemosa is economically important in the following respects:
diversification of wildlife habitat, mulch/organic matter, soil stabilisation, coastal protection, wind break and water quality improvement, timber, fuel, wood production, medicinal uses (the bark and leaves are used for the treatment of angina, boils, fungal infections, diarrhoea, dysentery, fever, malaria, anti-bleeding and leprosy (Bandaranyake, 1999; Koedam, 2000; Aluko *et al.*, 2002; Walters, 2005). *R. racemosa* has been shown to play a vital role in supporting marine food chains; hence De Lacerda (2002) stated that it is used as a stable food and animal fodder.

The importance of wetlands cannot be over emphasised (Bosire *et al*., 2005; Dahdouh- Guedas *et al*., 2005; Bamidele and Agbogidi, 2006; Bamidele *et al.,* 2007). Several mangrove tracts have been destroyed due to susceptibility of some mangroves to oil pollution as a result of leakages of pipelines, flow line, accidents and sabotage in the areas of operations (Agbogidi *et al*., 2005). The spilled oil has a complex mixture of thousands of hydrocarbons and related compounds that are toxic that float as slick buried in the soil or stranded on the shores damage aquatic environment (Azad, 2005; Agbogidi and Bamidele, 2007). The toxic effects of crude oil include acidification of the wetland soil, halt cellular respiration and as such starve the roots of vital oxygen (Merck, 2002). Understanding the critical threshold level at which adverse effect is felt on *R .racemosa* will enhance the life supporting process of plants, especially the coastal areas, discourage global warming, ameliorate desirable habitats for animals, aid water purification as well as the purification of the atmosphere. It is against this background that eco-toxicological implications of crude oil pollution on *R. racemosa* were embanked on. A study as this will contribute to the struggle against environmental degradation and will help to secure the future of the wetland coastal ecosystems (Nwilo *et al.*, 2007).

Materials and Methods

The study was carried out at the nursery site of the Department of Forestry and Wildlife, Delta State University, Asaba Campus (Latitude: $6^{0}14^{1}$N; Longitude: $6^{0}49^{1}$E) (Asaba Meteorological Station, 2008).

R. racemosa seedlings were collected from Warri, in Warri-West Local Government Area, Delta State and the basic nursery care was given for a period of two weeks. Soil samples used were obtained from flooded site close to Anwai River, Delsu Asaba Campus. The crude oil with specific gravity of 0.889g/cm^3 was obtained from the Nigerian National Petroleum Cooperation (NNPC), Warri, Delta State, Nigeria. 0.0, 12.0, 18.0, 24.0, and 30.0% w/w per 1.5kg of soil constituted the treatments. A seedling was planted per poly pot (30/50cm). The experiment was laid out in a randomized complete design with four replications. The trial ran for 12weeks. The seedlings were sown directly into the poly pots (30/50cm in dimension) the poly pots were watered immediately after planting and thereafter, every other day till the end of the trial. The set up was monitored for 12 weeks after transplanting (WAT) while growth characteristics were measured forth nightly. 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 leavers 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 diameter at 2.5cm above the soil level was measured with vennier callipers. Data obtained were subjected to analysis of variance while the significant means were separated using the Fisher's least significant differences (LSD) as recommended by SAS (2005).

Results and Discussion

Rhizophora racemosa seedlings tolerated all the concentrations (0.0, 12.0, 18.0, 24 and 30.0%) used. No death was recorded in the seedlings even at the highest level of crude oil application to soil. The performance of the seedlings in terms of plant height, number of leaves, leaf area, collar diameter significantly decreased ($P \geq 0.05$) with increasing levels of pollution over time (Tables 1-4 respectively). *R. racemosa* seedlings thrived better at 0.0 and 12.0% levels of pollution. At the highest (30.0%) level of pollution, there was leaf fall and yellowing of leaves from deep green to dark brown. The rate of growth at lower oil levels was also observed to be higher when compared with the seedlings grown in soils with higher levels of crude oil over time (Tables 1-4). Root growth of the seedlings was significantly reduced ($P \geq 0.05$) with increasing oil levels (Tables 5 and 6). At 30.0% oil dose, root hairs were altogether absent (Table 6).

The reductions in the growth characteristics of *R. racemosa* seedlings with increasing oil levels with time may be attributed to the various hydrocarbons and related compounds that are toxic to the biological organisms as well as the acidification of crude oil on wetland soils that halt cellular respiration as such, starve roots of vital oxygen hence the reduced root growth at higher concentrations of oil pollution. This observation agrees with the reports of Azad (2005) and Bamidele and Agbogidi (2006) on *Metamysidopsis insularis* and *Machaerium lunatus* respectively. The better performance in the seedlings of *R. racemosa* sown in the unpolluted soil could be attributed to the uninterrupted translocation of nutrients and water to xylem vessels while stress imposed by the crude oil could have accounted for a reduction in the performance of the seedlings exposed to the oil treatment. The study has demonstrated that crude oil application to soil has a negative effect of reducing the growth characteristics of *R. racemosa* seedlings and this may have implication on the growth and establishment of the red mangrove along the coastal region. At low levels (0.0-12.0%), *R. racemosa* seedlings were significantly unaffected in its growth parameters. The present study has established that *R. racemosa* can serve as a bio-indicator of pollution and can be recommended for use in areas of low levels of pollution for environmental clean-up or bioremediation.

Table 1. Plant height (cm) of *R. racemosa* seedlings as influence by different crude oil levels in soil

Oil in soil % (w/w)	Plant height/weeks after transplanting (WAP) 2	4	6	8	10	12
0.00	6.4^{a}	9.4^{a}	11.00^{a}	14.0^{a}	16.0^{a}	18.0^{a}
12.00	5.6^{a}	6.0^{b}	6.1^{b}	7.00^{b}	7.8^{b}	9.4^{b}
18.00	4.5^{ab}	4.9^{bc}	5.6^{bc}	6.3^{b}	6.4^{bc}	6.5^{c}

24.00	4.3[b]	4.5[bc]	5.0[bc]	5.1[bc]	5.2[bc]	5.3[cd]
30.00	2.8[b]	3.1[c]	3.9[c]	4.0[c]	4.1[d]	4.2[d]

Means with different superscripts are significantly different at P≤ 0.05 level of significance using the Fisher's least significant different (LSD).

Table 2. Number of leaves of *R..racemosa* seedlings as influenced by crude oil in soil

Oil in soil % w/w)	Number of leaves/WAP					
	2	4	6	8	10	12
0.00	0.66 [a]	1.33[a]	1.33[a]	2.00 [a]	2.00 [a]	2.66[a]
12.00	0.66 [a]	1.33[a]	1.33[a]	1.33 [ab]	1.33 [ab]	2.00 [ab]
18.00	0.66 [a]	1.33 [a]	1.33 [a]	1.33 [ab]	1.33 [ab]	1.33 [ab]
24.00	0.66 [a]	1.33 [a]	1.33 [a]	1.33 [ab]	1.00 [ab]	1.00[c]
30.00	0.33 [a]	0.66 [a]	0.66 [a]	1.00[b]	0.66[b]	1.66 [c]

Means with different superscripts are significantly different as P≤ 0.05 level of significance using the Fisher's least significant different (LSD)

Table 3. Leaf area of *R. racemosa* seedlings as influenced by crude oil in the soil.

Oil in soil % w/w)	Leaf area/WAP					
	2	4	6	8	10	12
0.00	42.75 [a]	45.03[a]	45.72[a]	52.70 [a]	88.00 [a]	138.00[a]
12.00	30.43 [a]	45.69[ab]	45.15[a]	50.30 [a]	52.70 [b]	85.00 [b]
18.00	15.85 [c]	35.95 [bc]	35.48 [b]	50.20 [a]	49.70 [b]	52.30 [c]
24.00	13.93 [c]	33.50 [c]	35.45 [b]	40.20 [b]	34.98 [c]	36.90[d]
30.00	7.00 [d]	16.59d [a]	34.50 [b]	39.20[b]	32.89[c]	34.60 [d]

Means with different superscripts are significantly different as P≤ 0.05 level of significance using the Fisher's least significant different (LSD)

Table 4. Collar diameter of *Rhizophora racemosa* seedlings as influenced by crude oil in soil.

Oil in soil % w/w)	Collar girth/WAP					
	2	4	6	8	10	12
0.00	4.5 [a]	4.8[a]	4.9[a]	5.0 [a]	50.1 [a]	5.2[a]
12.00	4.5 [a]	4.8[a]	4.9[a]	4.9 [a]	5.0 [a]	5.2[a]
18.00	4.4 [a]	4.6 [a]	4.8 [a]	4.8 [a]	4.8 [ab]	4.9 [ab]
24.00	4.4 [a]	4.6 [a]	4.7[a]	4.8 [a]	4.7 [ab]	4.7[ab]
30.00	4.1 [d]	4.5 [a]	4.6[a]	4.7[b]	4.5[ab]	4.5[ab]

Means with different superscripts are significantly different as P≤ 0.05 level of significance using the Fisher's least significant different (LSD)

Table 5. Root growth (length) of *R. racemosa* seedlings as affected by crude oil in soil

Oil in soil %(w/w)	Root length/WAP					
	2	4	6	8	10	12
0.0	6.7^a	7.8^a	8.6^a	9.4^a	10.2^a	10.8^a
12.0	6.5^a	7.4^a	8.2^b	9.1^a	9.8^a	10.6^a
18.0	6.1^b	6.6^b	6.8^b	7.1^b	7.3^b	7.4^b
24.0	6.0^b	6.2^b	6.3^c	6.2^c	6.2^c	6.2^c
30.0	5.6^c	5.4^c	5.3^d	5.2^d	5.2^d	5.2^d

Means with different superscripts are significantly different as $P \leq 0.05$ level of significance using the Fisher's least significant different (LSD)

Table 6. Root growth (root hairs) of *R. racemosa* seedlings as influenced by crude oil in soil

Oil in soil %(w/w)	Root hairs/WAP					
	2	4	6	8	10	12
0.0	3.4a	5.4a	6.5a	6.8a	7.2a	10.6a
12.0	3.3a	5.3a	6.3a	6.7a	7.0a	10.0a
18.0	3.0b	4.1b	4.6b	4.8b	4.9b	5.0b
24.0	2.6c	2.7c	2.1c	1.4c	0.2c	0.2c
30.0	1.0d	0.6d	0.1d	0.0d	0.0d	0.0d

Means with different superscripts are significantly different as $P \leq 0.05$ level of significance using the Fisher's least significant different (LSD)

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6/9/2009