Nigerian Journal of Ecology (2005) 7: 1-5.

ISSN: 1116-753X

EFFECTS OF CRUDE OIL AND ITS WATER SOLUBLE FRACTION ON THE GROWTH OF *Panicum repens* Linn

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(Accepted 10 September 2004)

ABSTRACT: The susceptibility of *Panicum repens* Linn. plants to crude oil and its water-soluble fraction (WSF) was investigated under laboratory conditions. The WSF of the crude oil was prepared to concentration levels of 12.5,25,50 and 100 percent while 10cm^3, 20cm^3, 30cm^3 and 40cm^3 volumes of the crude oil were used. Parameters assessed were growth patterns: change in leaf area, fresh and dry weights, and shoot/root (S/R) ratio. A negative relationship was found to exist between the inhibitory effects and the treatments. At the end of the trial (6 weeks), the growth of this macrophyte exposed to treatments 0.0% and 12.5% (WSF) and 0.0cm^3 and 10cm^3 (crude oil) in terms of leaf area, fresh and dry weights and S/R ratio was consistently more than those of the higher oil/WSF concentration levels and was significantly so at 5% level of probability. Although results obtained from the growth analysis showed the inhibitory effects of crude oil and its WSF on *P. repens*, low amount of the oil (10cm^3) and its WSF (12.5%) stimulated shoot growth. Death of *Panicum* plants was however not recorded even at the highest treatment level used in this study. The plant showed more tolerance to the crude oil than to its WSF. The present study suggests that this plant can serve as bio-indicator of pollution in oil producing areas.

Keywords: crude oil, water-soluble fraction, *Panicum repens*, susceptibility, pollution, and growth stimulation.

Introduction

Nigeria has abundant deposits of oil and natural gas and their exploitation has improved the economy substantially, but with serious adverse effects on the environment (Ogri, 2001). Severe ecological damage has occurred in the Niger Delta where most of the oil industries are based (Benka-Coker and Ekundayo, 1995; Ogri, 2001).

Crude oil is composed of numerous aliphatic, acyclic and aromatic hydrocarbons as well as small amount of oxygen, nitrogen, sulphur-containing compounds including some metal chelating prophyrins (Rosinni, 1960). The water- soluble fraction of crude oil is the fraction of the oil containing components fully or sparingly soluble in water and its constituents include dispersed particulate oil, soluble contaminants such as metallic ions and dissolved hydrocarbons (Kauss *et al.*, 1976).

Crude oil if spilled into water spreads over a wide area forming a slick and the oil in water immediately begins to undergo a variety of physical, chemical and biological changes including evaporation of high volatile fractions, dissolution of the water-soluble fractions, photochemical oxidation, drift, emulsification, microbial degradation and sedimentation (Lee, 1980; Muller, 1987). Oil pollution effects vary according to the type and amount of the oil, its chemical composition, the plant species and its age and the environment (Baker, 1970a).

The role of macrophytes in aquatic systems include primary production, source of food for aquatic fauna, oxygenators, pollution control, habitats diversification, fish capture/attractant and provision of shade (Jamil, 1993; Bamidele and Agbogidi, 2000). Aquatic macrophytes including *Panicum repens* are also being used for the production of materials including paper, roofing and a variety of chemicals (Bamidele and Agbogidi, 2002).*Panicum repens* is a tufted embankment grass but spreads over the water It is useful in the stabilization of sediments. The culms are soft, thick and spongy with branched often prop-rooted from the lower nodes (Mitchell, 1974). It is a common grass of aquatic ecosystems along the Niger Delta region – the hub of oil industry in Nigeria. With the high rate of oil spills in Nigeria shoreline (Baker *et al.*, 1993), this aquatic species stands to be negatively impacted and its role in the consolidation of the soil of the embankment zone among others would not be guaranteed. The objective of this study was to investigate the susceptibility of *P. repens* to crude oil and its WSF.

Materials and Methods

The *Panicum* plants were collected from a fishpond behind Guinness Nigeria Plc Factory after Ramat park along Agbor Road, Benin City, Edo State. They were carefully removed to avoid root damage, packed into a plastic container containing water to avoid excessive loss of water before getting to the laboratory.

The nutrient medium (NM) was prepared according to Hoagland and Arnon (1970). The stock solution was diluted to half strength and $500cm^3$ of the diluted solution was poured in 60 different plastic containers into which the plants were introduced. They were left for four (4) weeks to enable the plant adapt to the nutrient medium. The plants were subsequently observed for deficiency symptoms.

The WSF was prepared following the procedure of Afolabi *et al.* (1985) by slowly mixing $500cm^3$ of crude oil with $1,500cm^3$ of the NM using a Gallenkem orbital magnetic stirrer. The crude oil (light) used had a specific gravity of 0.655g/cc.and was sourced from the Nigerian National Petroleum Corporation (NNPC), Warri and made of Forcados blend. Each crude oil/NM mixture was shaken for 24 hours at room temperature. After shaking, the mixture stood for about 3 hours to obtain clear interphase between the oil and NM. The oily component was decanted and the mixture was poured into a glass stoppered separating funnel to allow for settlement. The WSFs were siphoned into three capped Winchester bottles to make the stock (100% WSF). The stock was diluted with half strength NM serially to give 50%, 25% and 12.5% strength WSF. $500cm^3$ of each of the diluted WSF including the 100% WSF was poured into the different containers into which the plants were transplanted. The NM alone was used as the control. $10cm^3$, $20cm^3$, $30cm^3$ and $40cm^3$ volumes of the crude oil were also introduced into $500cm^3$ NM just in the same way as the WSF. The set-up was left for 6 weeks.

A preliminary range finding test to determine the most appropriate concentration levels of the WSF to employ was carried out hence the following treatment levels were used: 12.5.25.50 and 100 per cent (WSF) and $10cm^3$, $20cm^3$, $30cm^3$ and $40cm^3$ (crude oil).

Description of growth patterns, change in leaf area, and fresh and dry weights were monitored for 6 weeks. The data obtained were used for determining S/R ratio. The leaf area was obtained by tracing out the outline of the leaves on graph sheets of known areas. Total leaf area per plant was calculated by proportion. The fresh and dry weights were determined after 42 days of treatment. The plants were cleaned with white cloth tissue paper to remove water and dust before weighing. The plants were separated into leaves, stems and roots. They were weighed (fresh weights) and later dried in an oven at a temperature of 85° C to a constant weight after which the dry weights were determined using the Mettler E 200 electronic weighing balance. Shoot/Root ratio determination was carried out by dividing the shoot dry weight value of the plant by the root dry weight value.

There were therefore 5 treatments including the control and each consisted of 12 plastic containers replicated 3 times. The experiment was conducted as a complete randomized block design. A total of 360 stands of *P. repens* plants were used for the study (180 for crude, 180 for its WSF).

The effects of crude oil and its WSF on the growth of *P. repens* were compared by ANOVA and the significant means separated using the Duncan's multiple range test (DMRT).

Results and Discussion

Table 1 shows the effect of crude oil and its WSF on the leaf area of *P. repens*. The higher levels of the oil ($30cm^3$ and $40cm^3$) and its WSF (50% and 100%) did not differ significantly from each other in their effects on *P. repens* leaf area. There was growth stimulation in the plants subjected to 10cm oil (100.67) and 12.5% WSF (96.42). Although the growth of *P. repens* plants exposed to higher volumes/concentrations of crude oil ($30cm^3$ and $40cm^3$) and its WSF (50% and 100%) was significantly reduced, no *Panicum* plants was observed death even at the highest treatment level used in this study. *P. repens* showed some level of tolerance even at higher levels of treatment. This observation is consistent with the findings of Kolattukudy (1979) that the effects of petroleum and its products depend on the species of plant. Naegele (1974) also reported that plants respond differently to pollution effects due to an innate genetic response of the plant system as modified by environmental influences. Reduction in the growth parameters of *P. repens* at higher treatment levels of the crude oil and its WSF as observed in the present study corresponds with the findings of Isirimah *et al.* (1989), Asuquo *et al.* (2002) and Agbogidi (2003) that the level of oil or its products present in the environment seems to be exerting the most influence on plants. Furthermore growth reduction in *P. repens* at high concentration levels as observed in the present study could be attributed to reduced photosynthetic activities of this aquatic and since it constitutes part of the primary producers in aquatic food chains, oil pollution therefore can impede aquatic productivity. WSF inhibition of growth has also been reported for *Chlorella vulgaria* (Kauss and Hutchinson, 1975). Similar observations have been made by Idoniboye and Andy (1985) and Powell *et al.* (1985). *Emola et al.* (1983) and Adeola (1996) reported that the much raised alarm on the adverse effects of oil spillage on fishing grounds is indirectly traceable to the deleterious effect of oil on phytoplankton and macrophytes which form the base of aquatic food chain.

Growth stimulation (fertilizer effect) was observed in *P. repens* grown in 10cm^3 oil and 12.5% WSF hence the observed significant difference between them and those grown in the control (unpolluted medium). This shows that little amount of the oil in the growth medium stimulated growth in terms of leaf area, fresh and dry weights. Similar observation was made by Baker (1970b). This may be due to the presence of growth regulating compounds such as naphthenic acids and the effects of essential micronutrients in the oil. These could have led to an increase in cell expansion as well as in total yield since the photosynthetic surface was increased. The effects of crude oil and its WSF on fresh and dry weights of *P. repens* are shown in Tables 2 and 3 respectively. The shoot fresh weight appreciably/significantly decreased while the root fresh weight increased with increasing volume/concentration of oil/WSF. The highest root fresh weight was recorded for plants exposed to 40cm^3 oil (0.77g) and 100% WSF (0.74g) while the lowest shoot weight was obtained for plants subjected to 40cm^3 oil (0.80g) and 100% WSF (0.36g). There was growth enhancement in the shoot fresh weight of *P. repens* plants growth in 10cm^3 oil (4.94g) and 12.5% WSF (3.62g). The dry weights (Table 3) showed the same trend in response to the crude oil and its WSF treatment as that obtained for the fresh weights. Interruption of supply from photosynthesis and translocation of photosynthates stored in the shoot could have led to the non-general increase in the root total growth (Browner, 1968; Saenger, 1982).

Table 4 shows the effect of crude oil and its WSF on the shoot /root ratio of *P. repens*. Although a relationship was found to exit between the inhibitory effect and the treatment volumes / concentration of both the crude oil and its WSF, the S/R ratio of the plants subjected to 10cm^3 oil (43.00) and 12.5% (41.00) showed growth stimulation (significant increase at 5% level of probability). This result indicates that the shoot of the plants was more affected by the oil and its

Table 1. Effect of crude oil (CO) and its water-soluble fraction (WSF) on the leaf area (cm^2) of *P. repens*.

Treatment (Crude Oil) (ml)		Treatment WSF (%)	
0	90.02b	0	90.02b
10	100.67a	12.5	96. 42a
20	70.80c	25	63.. 52c
30	55.51d	50	50.56d
40	50.9d	100	45.41d

Means with the same superscripts are not significantly different at P=0.05 level of significance using DMRT.

Table 2. The effect of crude oil and its WSF on fresh weight (g) of *P. repens*.

Treatment (Crude Oil) (ml)	Plant parts/fresh weight			Treatment (WSF) (%)	Plant parts /fresh weight		
	leaf	stem	root		leaf	stem	root
0	0.56b	3.25b	0.70a	0	0.56b	3.25b	0.70a
10	0.84a	4.10a	0.70a	12.5	0.62a	4.00a	0.71a
20	0.46c	1.76c	0.73a	25	0.39c	1.48b	0.72a
30	0,33c	1.26c	0.75a	50	0.11d	0.45c	0.73a
40	0.30c	0.50d	0.77a	100	0.06d	0.30c	0.74a

Means with the same superscripts are not significantly different at P=0.05 level of significance using DMRT.

Table 3. The effect of crude oil and its WSF on dry weights (g) of *P. repens*

Treatment (Crude Oil) (ml)	Plant parts /dry weights			Treatment (WSF)) (%)	Plant parts /dry weights		
	Leaf	Stem	Root		Leaf	Stem	Root
0	0.03a	0.75a	0.02a	0	0.03a	0.76a	0.02a
10	0.04a	0.77a	0.07a	12.5	0.04a	0.76a	0.02a
20	0.02	0.64b	0.02a	25	0.03a	0.60b	0.02a
30	0.02b	0.40c	0.02a	50	0.02b	0.34c	0.02a
40	0.02b	0.04d	002a	100	0.01b	0.02d	0.02a

Means with the same superscripts are not significantly different at P=0.05 level of significance using DMRT.

Table 4. The effect of crude oil and its WSF on Shoot / Root ratio of *P. repens*.

Treatment (Crude Oil) (ml)		Treatment WSF (%)	
0	39.00	0	39.00
10	40.50	12.5	40.00
20	33.00	25	31.50
30	21.00	50	18.00
40	3.00	100	1.50

Means with the same superscripts are not significantly different at P=0.05 level of significance using DMRT.

WSF than the root. It also shows that little amount of the oil and its WSF could be beneficial of plants. This however may vary among plant species.

It was also generally observed that *P. repens* showed more tolerance to the crude oil than to its WSF. This agrees with the work of Bohon and Clausen (1971) that crude oil contains some essential micronutrients, which may not be present in the WSF.

From this investigation, *P. repens* was observed to be more or less resistant to oil pollution effects. Since dispersal of oil with emulsifiers/ dispersants and other chemicals is not an ecologically acceptable method, *P. repens* could be employed to rehabilitate oil spill sites. Its use for the removal of petroleum hydrocarbons from polluted water bodies will gain a wide support. In addition, above 20cm^3 (crude oil) and 25% (WSF) levels, oil becomes increasingly deleterious to *P. repens* and its growth adversely affected.

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