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Growth pattern of two crop species on bio-remediated hydrocarbon polluted soils

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The effectiveness of agricultural wastes as bioremediation materials is gaining research attention as a better option in mitigating the issue of crude oil effects in soil environment. In the present study, the growth performance of *Telfairia occidentalis* and *Arachis hypogaea linn* in crude oil polluted soil amended with plantain peels (PP) and cocoa pod husks (CPH) were investigated. Eight kilograms (8 kg) each of dried soil samples were collected and weighed into 60 polythene bags. The polythene bags except the pristine control were polluted with 80 ml of crude oil and allowed for 14 days of soil acclimatization. The treatments comprising of CPH and PP were amended after 14 days, using the following concentrations: 0, 100, 150 and 200 g and allowed to acclimatize for 60 days. *T. occidentalis* and *A. hypogaea linn* were cultivated immediately after treatment regimen. The pH of the amended and un-amended soil samples were observed to be at a range recommended for effective bioremediation of hydrocarbon polluted soil. The organic carbon content of the CPH amended soils were significantly reduced as compared to the PP amended soil. The phosphorus, nitrogen, potassium and other essential soil parameters evaluated were significantly high ($P<0.05$) in CPH amended soil than the PP amended soil. Crops grown in the rehabilitated soils possess a high adaptability in CPH amended soil than the PP amended soil. The amendments most preferably cocoa pod husks which tend to be more effective in the reduction of hydrocarbon content of the soil should be utilized in the enhancement of microbial degradation of crude oil product in soils.

Key words: Crude oil, bioremediation, pollution, amendment, cocoa pod husk, plantain, groundnut, fluted pumpkin.

INTRODUCTION

Environmental pollution is a major problem which affects biodiversity, human health, aquatic and terrestrial habitat. The Niger Delta eco-region of Nigeria has been associated with frequent oil spills resulting from oil pipeline vandalization, tanker accidents and accidental rupture of oil pipelines. These mishaps result in the release of crude oil refined petroleum products into terrestrial and aquatic environments. Contamination of soil by crude oil spills is an environmental problem that often requires cleaning up of the contaminated sites

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(Bundy et al., 2007). Crude oil spills affect plants adversely by creating conditions which make essential nutrients like nitrogen and oxygen needed for plant growth unavailable to them. Oil pollution in whatever form is toxic to plant and soil microorganisms, pollution studies have described that oil spills kill agricultural plants or inhibit the growth performance of an entire vegetation cover. Plants have been described as the first victim of an oil spill impacted site. Ogbo et al. (2009) reported that oil in the soil creates unsatisfactory conditions for plant growth probably due to non-sufficient aeration of the soil. Oil readily penetrates the pore space of terrestrial vegetation following any spills with heavier friction which may block the pores and this subsequently impedes photosynthesis and other physiological processes in plant (Ogbo et al., 2009). Adam and Duncan (2002) reported that this effect could be due to the oil which acts as a physical barrier preventing or reducing access of seeds to water and oxygen. One of the environmental challenges posed by oil pollution is the alteration in the physical and chemical nature of the soil. Ekpo et al. (2012) reported that plants like cassava, *Thaumacoccus danielli* and sweet potatoes are tolerant to crude oil pollution. While Agbor et al. (2012) reported that the growth performance of some plants in crude oil polluted soil could be ascertained through effective remediation of the soil to reduce the hydrocarbon content of such environment. Biostimulation has been described as an effective method in the remediation of crude oil polluted soil since it involves the use of economically friendly waste (cocoa pod husks and plantain peels) which are not toxic to soil microorganisms but tend to enhance the proliferation of microorganisms in the soil.

Sequel to this, it is pertinent to reduce the hydrocarbon contents of the soil used for agricultural purposes through remediation. In this present study assessment of the growth performance of *Telfairia occidentalis* and *Arachis hypogaea* linn in plantains peels and cocoa pod husks amended soil were evaluated.

MATERIALS AND METHODS

Collection of materials and study site

Mature seeds of *T. occidentalis* and that of *A. hypogaea linn* were purchased from Marian Market Calabar. The plantain peels and cocoa pod husks were collected from Bashua village in Boki Local Government Area, Cross River State while the crude oil (Bonny light) was obtained from the Nigerian Agip Oil Company (NAOC) Port-Harcourt, Rivers State Nigeria. The study was conducted in Step - B Project Site, University of Calabar.

Sample collection and pollution

Top soil (0-25 cm depth) was collected from four points around Biological Science Experimental Farm (BSEF), bulked to form composite soil sample. The soil was air dried and eight kilograms (8 kg) each was weighed into sixty (60) polythene bags. The soil was then polluted with 80 ml of crude oil and kept for fourteen (14) days. This was to allow indigenous microorganisms present in the soil to be acclimatized with the new condition. After fourteen days, the different amendments were applied as shown in Table 1.

Preparation of cocoa pod husks and plantain peels

The collected cocoa pod husks and plantain peels were air dried for thirty (30) days and crushed to powder using electric blender (Diesel engine R175). The powdered materials were stored in containers.

Application of amendment

The CPH and PP were applied to the various polythene bags as shown in the combination Table 1. However, after the treatment, the soil was allowed for sixty (60) days for biodegradation of crude oil in the soil. During this period 200 ml of water was applied to each of the bags every two (2) day to keep the soil environment moist.

Sowing of seeds

After 60 days of remediation, mature seeds of both plants were used for planting. The seeds were sown into the soil in the polythene bags. Sowing was done to assess the growth performance of the remediated soil.

Determination of physico-chemical parameters of soil

Estimation of physico-chemical parameters of the soil was done using the method of Eno et al. (2009). The following parameters were analyzed: Moisture, organic carbon, particle size, nitrogen, phosphorus, potassium, magnesium, sodium, aluminium, hydrogen, pH, exchangeable cation exchange capacity, base saturation and electrical conductivity.

Data collection

Data were obtained on the following parameters: days to seedling emergence, numbers of leaves, leaf length measured using tape, leaf area measured using graph sheet, plants height measured using tap and leaf width measured using tape.

Statistical analysis

Data collected were subjected to a two-way analysis of variance (ANOVA) while significant means were separated using least significant difference (LSD) tests at 5% probability level.

RESULTS

Growth performance of *T. occidentalis* on crude oil remediated soil using cocoa pod husks and plantain peels

The result obtained shows that cocoa pod husks (CPH) at different concentrations significantly increases ($p < 0.05$) the plant height of *T. occidentialis* than plantain peels. The highest plant height between CPH and PP

Table 1. Treatment combinations of cocoa pod husks and plantain peels**.**

Amendment	Plants	Concentrations	Combinations
CPH	TO	PC	CPH TO PC
		COC	CPH TO COC
		100 g	CPH TO 100 g
		150 g	CPH TO 150 g
		200 g	CPH TO 200 g
	AHL	PC	CPH AHL PC
		COC	CPH AHL COC
		100 g	CPH AHL 100 g
		150 g	CPH AHL 150 g
		200 g	CPH AHL 200 g
PP	TO	PC	PP TO PC
		COC	PP TO COC
		100 g	PP P TO 100 g
		150 g	PP P TO 150 g
		200 g	PP TO 200 g
	AHL	PC	PP AHL PC
		COC	PP AHL COC
		100 g	PP AHL 100 g
		150 g	PP AHL 150 g
		200 g	PP AHL 200 g

Twenty treatment combinations were used and replicated thrice to give sixty plots. CPH, Cocoa pod husks; TO, *Telferia occidentalis;* AHL, *Arachis hypogaea linn;* PP, plantain peels; PC, pristine control; COC, crude oil control.

was observed at concentrations of 150CPH and 150PP (Table 3). The number of leaves, leaf width, leaf length and leaf area was also observed to be significantly high in cocoa pod husks (CPH) remediated soil at different concentrations, this was followed by plantain peels (PP) remediated soil. The improvement in the morphological attributes of the *T. occidentalis* was observed to be dose dependent (Table 1). It was observed that prolonged days to germination of *T. occidentalis* was recorded in crude oil control soil followed by pristine control, 100CPH, 150CPH, 2000CPH and 150PP with no significant difference (P > 0.05) in days to germination while 150PP and 200PP had the shortest days to germination (Table 2).

Growth performance of *A. hypogaea linn* on crude oil remediated soils using cocoa pod husks and plantain peels

Bioremediation is a better tool in achieving optimum growth of plant in hydrocarbon polluted environment. It was observed that the amendment of crude oil polluted soil with 150CPH and 200CPH significantly increase (P < 0.05) the plant height of groundnut plant than the control group. The result also shows that 100CPH, 150PP and 200PP had no significant difference (P > 0.05) in the plant height of groundnut but significantly higher (P < 0.05) than the control group. The crude oil control groups had the shortest plant height which implies that the amendments were effective and significantly reduced the hydrocarbon content of the soil as shown in Table 4. The result also shows that 150CPH and 200CPH had significantly higher (P < 0.05) number of leaves than the control group while the control group, 100CPH, 100PP, 150PP and 200PP had no significant difference (P > 0.05) in the number of leaves but significantly higher than the crude oil control groups, thus, indicating the effect of the oil. The leaf width was significantly increased (P < 0.05) in soil amended with 150CPH and 200CPH than the control group while 100CPH, 150PP and 200PP had no significantly difference (P > 0.05) in leaf width but significantly higher (P < 0.05) than the pristine control group. The soil remediated with 100PP and the pristine control showed no significant difference (P > 0.05) in leaf width which were not significantly higher (P < 0.05) than the crude oil control group (Table 3).

However, the leaf length and leaf area in CPH remediated soil were significantly higher (P < 0.05) than that of PP amended soil. Delay in days to germination was observed in crude oil amended soil while the soil amended with high concentrations of the waste

Table 2. Morphological attributes of fluted pumpkin on remediated soils.

Parameters	PC1	COC1	100CPH	150CPH	200CPH	PC2	COC2	100PP	150PP	2000PP	LSD
Plant height	$34.18^b \pm 0.16$	$20.5^e \pm 0.09$	31.3^c 0.10	$34.18^b \pm 0.18$	$40.97^a \pm 0.11$	$10.3^g \pm 0.11$	$11.22^g \pm 0.08$	$16.12^f \pm 0.05$	$22.48^e \pm 0.12$	$27.05^d \pm 0.06$	2.23
No. leaves	$28.00^d \pm 0.08$	$11.83^g \pm 0.06$	$31.33^c \pm 0.10$	$35.33^b \pm 0.14$	$39.83^a \pm 0.16$	$19.00^f \pm 0.08$	$8.00^h \pm 0.07$	$19.83^f \pm 0.06$	$24.83^e \pm 0.09$	$28.33^d \pm 0.10$	2.23
Leaf width	$10.67^c \pm 0.05$	$8.63^d \pm 0.02$	$13.35^b \pm 0.08$	$14.05^b \pm 0.04$	$15.03^a \pm 0.06$	$5.9^f \pm 0.02$	$4.07^g \pm 0.02$	$5.82^f \pm 0.03$	$6.75^f \pm 0.07$	$7.65^e \pm 0.03$	0.79
Leaf length	$10.58^c \pm 0.06$	$6.37^d \pm 0.03$	$13.00^b \pm 0.06$	$14.5^a \pm 0.04$	$14.88^a \pm 0.05$	$6.07^d \pm 0.06$	$4.03^e \pm 0.04$	$7.47^d \pm 0.03$	$7.92^d \pm 0.04$	$8.25_d \pm 0.05$	1.10
Leaf area	$47.5^d \pm 0.24$	$27.67^h \pm 0.15$	$61.17^c \pm 0.03$	$70.33^b \pm 0.09$	$82.17^a \pm 0.18$	$32.\ 5^g \pm 0.24$	$19.33^i \pm 0.20$	$36.00^f \pm 0.17$	$43.00^e \pm 0.16$	$48.67^d \pm 0.10$	3.22
Days for germination	$7.00^c \pm 0.06$	$13.33^a \pm 0.08$	$8.67^c \pm 0.09$	$8.00^c \pm 0.03$	$7.67^c \pm 0.04$	$7.00^c \pm 0.01$	$11.33^b \pm 0.06$	$7.00^c \pm 0.03$	$6.00^d \pm 0.04$	$5.00^e \pm 0.06$	0.78

Means with the same case letter along the horizontal array indicate no significant difference (P > 0.05). PC1: Pristine control, COC: crude oil control.

Table 3. Morphological attributes of groundnut on crude oil remediated soils.

Parameter	PC1	COC1	100CPH	150CPH	200CPH	PC2	COC2	100PP	150PP	200PP	LSD
Plant height	$29.62^d \pm 0.20$	$16.84^e \pm 0.14$	$28.67^c \pm 0.09$	$31.43^b \pm 0.11$	$34.8^a \pm 0.24$	$25.45^d \pm 0.08$	$13.58_e \pm 0.05$	$17.8^d \pm 0.05$	$21.48^c \pm 0.06$	$23.9^c \pm 0.07$	2.46
No. leaves	$15.69^b \pm 0.11$	$6.67^c \pm 0.04$	$18.83^b \pm 0.08$	$26.17^a \pm 0.10$	$27.17^a \pm 0.12$	$13.33^b \pm 0.06$	$6.67^c \pm 0.04$	$17.00^b \pm 0.05$	$19.5^b \pm 0.07$	$22.00^b \pm 0.11$	3.14
Leaf width	$3.5^c \pm 0.09$	$2.33^d \pm 0.02$	$5.2^b \pm 0.03$	$6.62^a \pm 0.01$	$6.88^a \pm 0.04$	$3.33^c \pm 0.01$	$1.8^d \pm 0.01$	$3.33^c \pm 0.02$	$4.58^b \pm 0.01$	$5.22^b \pm 0.04$	0.82
Leaf length	$10.38^b \pm 0.03$	$8.6^c \pm 0.02$	$9.82^b \pm 0.06$	$11.47^a \pm$ o.10	$11.75^a \pm 0.05$	$4.77^d \pm 0.03$	$2.77^d \pm 0.02$	$3.63^d \pm 0.02$	$3.83^d \pm 0.01$	$3.88^d \pm 0.02$	1.00
Leaf area	$27.5^b \pm 0.16$	$29.67^b \pm 0.11$	$2967^b \pm 0.14$	$31.30^b \pm 0.20$	$34.83^a \pm 0.13$	$12.67^d \pm 0.09$	$8.1^e \pm 0.06$	$15.5^d \pm 0.07$	$19.5^c \pm 0.10$	$22.3^c \pm 0.09$	2.83
Days for germination	$6.00^b \pm 0.02$	$8.67^a \pm 0.04$	$5.00^c \pm 0.03$	$4.33^d \pm 0.03$	$4.00^d \pm 0.02$	$5.33^c \pm 0.04$	$9.00^a \pm 0.03$	$6.00^b \pm 0.07$	$4.33^d \pm 0.02$	$4.00^d \pm 0.04$	0.56

Mean with the same alphabet along the horizontal axis represent no significant difference (P > 0.05).

germinated faster than the control groups (Table 3).

Physico-chemical properties of the amended soil using cocoa pod husks and plantain peels

The moisture content of the soil was observed to be high in 200PP amended soil followed by 200CPH, 100PP, and 150PP amended soil. The soil with 100CPH and the crude oil control had no significant difference (P > 0.05) while the pristine control group produces the lowest moisture content. The pH of the amended soil was significantly high (P < 0.05) in all the treated groups with no significant difference (P > 0.05) in pH range. Among the treated soil it was observed that cocoa pod husks and plantain peels at different concentrations significantly reduce (P < 0.05) the organic carbon content of the soil as compared with the crude oil control soil while the pristine control had the lowest organic carbon content (Table 4). The phosphorus contents of the soil was observed to be significantly high (P < 0.05) in 200CPH amended soil and pristine control groups. However, the different amendments significantly increase (P < 0.05) the phosphorus content of the soil as compared with the crude oil control groups that had the lowest phosphorus content. The result also shows that there were no significant difference (P > 0.05) in the magnesium, potassium, H^+, Al^{3+}, sodium, ECEC and H of the soil (Table 4).

DISCUSSION

Physico-chemical properties of crude oil amended soil using cocoa pod husks and plantain peels

The physical and chemical properties of the soil determine to a large extend the microbial activity of the soil and also the performance of plant. Over the past few decades in oil producing communities,

Table 4. Physico-chemical properties of the soil amended with cocoa pod husks and plantain peels.

Parameters	PC1	COC1	100CPH	150CPH	200CPH	PC_2	COC_2	100PP	150PP	200PP	LSD
Moisture	14.5 ± 0.12	16.8 ± 0.09	16.9 ± 0.05	15.8 ± 0.07	22.6 ± 0.04	14.5 ± 0.12	16.8± 0.09	19.6± 0.05	20.3± 0.06	23.4± 0.08	0.72
pH	5.6 ± 0.03	5.9 ± 0.04	7.2 ± 0.02	7.6 ± 0.03	7.8 ± 0.02	5.6±0.01	5.9 ± 0.04	7.0 ± 0.03	7.2 ± 0.03	7.4 ± 0.04	0.92
Org. c.	1.10 ± 0.02	4.86 ± 0.01	2.7 ± 0.01	2.5 ± 0.02	2.30 ± 0.01	1.10± 0.03	4.86 ±0.01	3.0 ± 0.02	3.0 ± 0.01	2.8 ± 0.01	0.69
Nitrogen (%)	0.18 ± 0.01	0.06 ± 0.01	0.19 ± 0.01	0.25 ± 0.01	0.26 ± 0.01	1.10 ± 0.02	0.06 ± 0.01	0.17 ± 0.01	0.20 ± 0.01	0.22± 0.01	0.05
Phosphorus	0.11±0.01	5.83 ± 0.03	40.0 ± 0.08	42.9 ± 0.12	58.2 ± 0.07	0.18 ± 0.01	5.83 ± 0.03	29.9 ± 0.1	36.6 ± 0.04	45.2± 0.13	1.68
Potassium	0.40 ± 0.01	0.16 ± 0.01	0.13 ± 0.01	0.14 ± 0.01	0.18 ± 0.01	43.8 ± 0.11	0.16 ± 0.01	0.11±0.01	0.14± 0.01	0.16± 0.01	0.67
Magnesium	0.98 ± 0.01	0.84 ± 0.01	2.4 ± 0.02	2.4 ± 0.01	2.8 ± 0.01	0.40 ± 0.01	0.84 ± 0.01	1.2 ±0.01	1.2 ± 0.01	1.5 ± 0.01	0.96
Calcium	1.30 ± 0.02	1.20 ± 0.01	9.6 ± 0.04	11.3 ± 0.03	14.6 ± 0.04	0.98 ± 0.01	1.20 ± 0.01	4.9 ± 0.01	6.4 ± 0.03	8.6 ± 0.03	0.65
H^+	1.09 ± 0.01	1.00 ± 0.01	ND	ND	ND	1.30 ± 0.02	1.00 ± 0.01	ND	ND	ND	NS
Al^{3+}	1.00 ± 0.01	0.78 ± 0.01	ND	ND	ND	1.09 ± 0.01	0.78 ± 0.01	ND	ND	ND	NS
Sodium	0.10 ± 0.01	0.14 ± 0.01	0.09 ± 0.01	0.12 ± 0.01	0.14 ± 0.01	1.00 ± 0.01	0.14 ± 0.01	0.06 ± 0.01	0.09 ± 0.01	0.11± 0.01	NS
Exchangeable CEC	2.78 ± 0.03	2.34 ± 0.02	12.2 ± 0.06	13.96 ± 0.1	17.72 ± 0.1	2.78 ± 0.03	2.34 ± 0.02	6.27 ± 0.03	7.83 ± 0.02	10.37± 0.03	7.31
EC	0.56 ± 0.01	0.30±0.01	0.89 ± 0.01	1.10 ± 0.01	1.20 ± 0.01	0.56 ± 0.01	0.30 ± 0.01	0.45 ± 0.01	0.98 ± 0.01	1.0 ± 0.01	0.42
BS	24.82 ± 0.1	23.93 ± 0.2	100 ± 0.00	100. ± 0.00	100.0.00	24.82 ± 0.2	23.93 ± 0.2	100 ± 0.1	100.0.0	100 ± 0.0	7.93

Means with the same case letter along the horizontal array represent no significant difference (P > 0.05).

farm land deterioration and abandonment have been observed due to oil spills on land. Biostimulation potentials of agro-wastes is beginning to gain research attention as better alternative in enhancing microbial degradation of hydrocarbon in soil and possibly improving the physicochemical properties of the soil. Interestingly, the application of cocoa pod husks and plantain peels in crude oil contaminated soil significantly improved the physicochemical properties of the soil. The extent of degradation of hydrocarbon was significantly high in soil amended with cocoa pod husks and plantain peels. The pH level of soil amended with cocoa pod husks was significantly higher than soil amended with plantain peels. However, the pH level of both amended soil was observed to be alkaline. This implies that the amendments have strong buffering capacity in the soil. The result of Agbor et al. (2013) reported that both cocoa pod husks and plantain peels possess strong buffering effect in the soil. Olabisi et al. (2009) also reported that melon shells have strong buffering effect on soil and observed that melon shells helps in enhancing the microbial degradation of the soil. pH range of a particular soil gives an estimate of the nutritional properties as well as the fertility of the soil. It was observed from the result obtained that the pH of the crude oil control (COC) was acidic. The presence of high amount of acidity in soil, could adversely affect soil conditions including microbial activities in the soil. It has also been observed that increased hydrocarbon concentration in the soil reduces the phosphorus and nitrogen content of the soil as observed in the crude oil control. However, Ekpo et al. (2013) reported that the presence of high concentration of crude oil in soil reduces microbial activity in such environment. Offor and Akomaye (2006) had also reported that the availability of nitrogen and phosphorous in hydrocarbon polluted soil stimulate microbial population in the impacted site. The organic carbon content of the soil amended with cocoa pod husks was observed to significantly reduce (P < 0.05) than the plantain peels amended soil. The productivity of plant in soil with high organic carbon content could adversely be affected.

Assessment of growth performance of *T. occidentalis* and *A. hypogaea linn* on crude oil remediated soil

The improvement in the growth performance of *T. occidentalis* and *A. hypogaea linn* in the biostimulated soil was higher than the pristine soil and crude oil control soil. This could be attributed to the increase in soil nutrient. It was observed that the higher the concentration of the amendment, the greater the growth performance of the plant. Stewart et al. (1974) observed that increased in nitrogen in soil increases vegetative growth of cassava. The significant reduction in growth performance of *T. occidentalis* and *A. hypogaea linn* in the control soil conformed with the report of Venosa et al. (2002) that crude oil polluted soil reduces the growth of plants. Black (1957) maintained that growth and development of plants are adversely affected by crude oil pollution, ranging from wilting, chlorosis, tissue and cell maceration, blotching and the collapses of marginal necrotic spots, which have eventually resulted in the death of plants. The potentials of these agro-wastes in improving the growth performances of these plants in hydrocarbon polluted environment justified “the result of the physicochemical properties of the soil. That indicates the increase in nitrogen and phosphorus content of the soil. In comparison, among the two agro-wastes cocoa pod husks tend to significantly improve the plant height, leaf area, number of leaves, leaf length and width of the *T. occidentalis* and *A. hypogaea linn* than the plantain peels amended soil. However, this result could also imply that the degradation rate of hydrocarbons in the soil amended with cocoa pod husks are good bioremediation agents.

Conclusion

Therefore, this study had revealed that introduction of crude oil into agricultural soil adversely and severely inhibits plant growth and development as observed in the crude oil control. Thus the soil amended with cocoa pod husks and plantain peels tend to reduce the hydrocarbon content of the soil, and significantly improved the growth of *T. occidentalis* and *A. hypogaea linn* in the soil. It can be concluded that cocoa pod husks and plantain peels are good remediating materials that should be used in the cleaning up of crude oil polluted environment.

Conflict of Interest

The authors have not declared any conflict of interest.

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