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Effects of Qua Iboe (Nigerian) Crude Oil on the Germination and Growth of Okra (*Abelmoschus esculentus*, L) and Fluted Pumpkin (*Telferia occidentalis*, L) in the Tropics

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Abstract: Field studies to ascertain the effects of crude oil spillage on agricultural crops and soil environments are few. A greenhouse experiment was carried out using zero % (control), 2%, 4% and 8% v/w levels of Qua Iboe crude oil treated soils to investigate the response of Okra (*Abelmoschus esculentus*) and fluted pumpkin (*Telferia occidentalis*) to crude oil contamination. The results showed that redox potential, pH, temperature, electrical conductivity, organic carbon, total nitrogen and total hydrocarbon contents of the contaminated soils increased with increase in crude oil concentrations. In contrast, available phosphorus, effective cation exchange capacity, exchangeable bases and acidity decreased with increase in crude oil concentrations. Okra and *Telferia* seeds showed a delayed germination in the 2% and 4% treated soils and total inhibition of germination in the 8% treated soils. The growth of Okra and *Telferia* seedlings decreased significantly ($P < 0.05$) with increase in crude oil concentrations. The amount of heavy metals and THC absorbed by Okra and *Telferia* shoots increased with increase in crude oil concentrations. However, Okra absorbed more Fe, Ni, and Cd while *Telferia* absorbed more Cu, Zn, and THC. This study recommends *Telferia* for a long term bioremediation of oil polluted sites after accidental spillage on land.

Key words: Crude oil, germination, growth, *Abelmoschus esculentus, Telferia occidentalis,* bioremediation.

Introduction

Nigeria is a major crude oil exporter as well as an important agricultural nation in the West African subregion. Economic benefits derivable from oil has no doubt left negative impacts on the Nigerian environment. Such impacts have been reported on farming activities (Odu 1980, Adeniji *et al* 1983), plants (Guthrie & Perry 1975, Odu 1980), man (Guthrie & Perry 1975, Walker 1975), fishes and coastal waters (Odu 1980, Asuquo 1994, Devmankonsult 1998). Since 1976, about 5334 cases of crude oil spillages releasing an estimated 2.8 million barrels of oil into the land, swamp, estuaries and coastal waters have been reported (Odiete 1999).

This study aims at investigating the effects of crude oil on the germination and growth of fluted pumpkin (*Telferia occidentalis*) and Okra (*Abelmoschus esculentus*) in a tropical environment.

Data Collection and Analysis

Location of study

The greenhouse experiment was carried out in the University of Calabar, Nigeria with garden soil samples mixed and treated with varying amounts of crude oil obtained from Qua Iboe Terminal (Ekpe oil well), Eket, Nigeria. The physical and chemical composition of Ekpe oil is given in Table 1.

Sample preparation and soil treatments

In the greenhouse, 4 samples of garden soil were treated with 0 %, 2%, 4% and 8% crude oil treatments according to Nicolotti and Egli (1998). These set up was replicated four times for Okra and *Telferia occidentalis* respectively.

Biotest and sowing of seeds

Seeds of Okra (*Abelmoschus esculentus* (L) *Moench*) and fluted pumpkin (*Telferia occidentalis* Hoof F) were collected from Akwa Ibom State Agricultural Development Project (AKADEP), Uyo, Nigeria. The seeds were soaked in de-ionised water for 4 hours to confirm viability, while non-viable seeds (floating seeds) were discarded. Three seeds of each species were sown in already prepared soils in polythene bags at a planting depth (AKADEP depth) of 10 cm and 5 cm for *Telferia* and *Abelmoschus* respectively.

Physicochemical measurements

Soil pH, e^H, temperature and electrical conductivity were measured insitu using portable pH/e^H meters (WTW 90), and conductivity– temperature meter (HACH Instruments) in soil/water, 1:1 w/ volume measurement. Crop heights were obtained with a metre rule. Total organic carbon, total nitrogen and available phosphorous were carried out according to Feniran and Areola (1978). Exchangeable bases, acidity, Effective Cation Exchange Capacity (ECEC) and Percentage Base Saturation (PBS) were also determined (Feniran and Areola 1978). Heavy metals and total hydrocarbons were determined by AAS and UV spectrophotometry after tissue digestion with 1:3 HNO_3/H_2SO_4 V/V and soxhlet extraction of saponified tissue with Analar grade n-Hexane (Devmankonsult 1998).

Results

Rate of Germination

All Okra seeds germinated on the third day of sowing for the control , 2% and 4% oil treated soils. For the 8% treated soils, there were no germination throughout the test period. *Telferia* seeds germinated after about 6 days of planting. Seeds in the control, 2% and 4% treated soils germinated on the 6^{th}, 9^{th} and 15^{th} days respectively. There was no germination in the 8% oil treated soils.

Seedlings growth

Table 2 shows the plant heights of okra and *Telferia* seedlings. Seedlings growth (cm) was measured at three days interval through out the test period and the mean for the four replicates obtained per treatment. In okra, the shoot lengths decreased from 12.33 cm (control) to 6.80 cm (2% treatment) and then to 0.95 cm (4% treatment). The seedlings on 4% treated soils died on the second day after germination .

In *Telferia* the shoot lengths decreased from 58.83 cm (control) to 27.41 cm (2% treatment) and then to 5.08 cm (4% treatment). The seedlings on 4% treated soils were uprooted after 3 and 12 days of germination for Okra and *Telferia* respectively and preserved for laboratory analysis.

The seedlings height monitored also revealed that while normal growth occurred at the control , the 2% and 4% oil treated soils showed growth retardation. The percentage growth retardation was obtained from the expression:

$$\% \text{ Growth retardation} = \frac{\text{Observed growth}}{\text{Growth at control}} \times 100$$

The Okra seedlings growth were retarded by 55.5% and 13.4% in the 2% and 4% treatments while 45% and 6.2% retardation occurred in 2% and 4% treatments for *Telferia* respectively. This indicates that, *Telferia* seedlings were markedly affected by the crude oil application.

Physicochemistry of treated soils

Tables 3 and 4 show the soil chemistry of the control and treated soils. While Tables 5 and 6 present the levels of heavy metals and THC in the soils and crops after the experiment. No THC and heavy metal measurements were made on the virgin or unpolluted garden soils.

For both crops, redox potentials decreased markedly in the 2% treatment but increased with an increase in oil concentration up to 8% treatment (Fig. 1). In Okra soils, the redox potential reduced from 762 mv in control to 551mv in 2% treatment but increased to 690 mv in 8% treatment. Similarly, in *Telferia* soils, the redox potentials decreased from 772 mv in control to 474 mv in 2% treatment but later increased to 689 mv in 8% treatment.

As shown in (Fig. 1), the pH of the soils are markedly affected by crude oil contamination. The generally low pH of the soils indicate that the soil was acidic. In Okra treated soils, the pH increased from 5.79 in control to 5.92 in 8% treatment. Similarly, in *Telferia* treated soils, the pH increased from 5.63 in control to 6.03 in 8% treatment.

A similar pattern of change as observed with e^H was recorded for soil temperature. Soil temperature for the contaminated and uncontaminated in control soils are typical of soils in the tropics. In Okra raised soils, the temperature decreased from 27.5 °C in control to 27.30 °C in 2% treatment and then to 28.0 °C in 8% treatment. Similarly, in *Telferia* treated soils, temperature decreased from 27.40 °C in control to 27.30 °C in 2% treatment and correspondingly increased to 27.85 °C in 8% treatment (Fig. 2).

Table 1: Characteristics of Ekpe Crude Oil, Nigeria

Parameters	Measured values
Specific gravity	0.826
API gravity (A°)	39.6
Bottom hole Temp. (°C)	101.1
Viscosity (CP)	0.29
Gas Oil ratio	885
Reservoir Depth (m)	2549
Carbon %	83.9 –86.8
Hydrogen %	11.4 – 140
Sulphur %	0.06 - 8.0
Nitrogen %	0.11 – 1.70
Oxygen %	0.5
Trace metals %	0.03

Modified after Asuquo (1994)

Table 2: Seedling heights (cm) measured every 3 days during the study

	Okra				Pumpkin			
DAYS	CONTROL	2%	4%	8%	CONTROL	2%	4%	8%
0	-	-	-	-	-	-	-	
3	7.1	3.71	0.95		-	-	-	-
6	9.25	4.25	-	-	7.5	-	-	-
9	11.8	6.1	-	-	16.8	6.8	-	-
12	12.3	6.25	-	-	31.6	10.15	-	-
15	12.7	6.75	-	-	61.85	19.4	3.1	
18	13.05	7.35	-	-	72.7	25.5	5.4	
21	13.5	7.8	-	-	79.45	39.8	5.8	
24	15.05	8.9	-	-	93.75	54.5	6	
27	16.2	10.1	-	-	107	62.1	-	
Mean	12.33	6.8	0.95	-	58.83	27.41	5.08	-
(sd)	-2.8	-2	(-)		-36.5	-22.6	-1.34	

Table 3: Soil chemistry of Okra treated soils after planting.

	Treatments				
Parameter	control	2%	4%	8%	Critical values in soils (Brady & Weil, 1996)
Total organic carbon (%)	1.53	2.63	3.28	3.6	2.0
Total N (%)	0.11	0.30	0.32	0.34	0.1-0.45
Available P (ppm)	90.30	53.50	49.55	48.60	20-100
Exch. Ca (meq/100 g)	0.4	0.5	0.6	0.8	10
Exch. K (meq/100 g)	0.17	0.13	0.11	0.10	0.2-0.3
Exch. Na (meq/100 g)	0.11	0.10	0.09	0.08	0.6-1.2
Exch. Mg (meq/100 g)	0.8	0.6	0.4	0.3	3-8
Exch. acidity (meq/100 g)	5.0	2.0	2.4	2.7	0.7-2.0
ECEC (meq/100 g)	6.48	3.33	3.6	3.98	3.0
Base sat. (%)	29.6	66.5	50.0	47.4	50-100

Table 4: Soil chemistry of *Telferia* treated soils after planting

Parameter	Treatments				Critical values in soils (Brady & Weil, 1996)
	control	2%	4%	8%	
Total organic carbon (%)	1.50	2.88	3.30	3.57	2.0
Total N (%)	0.12	0.26	0.29	0.31	0.1-0.45
Available P (ppm)	88.75	56.10	50.61	48.50	20-100
Exch. Ca (meq/100 g)	1.2	1.0	0.8	0.4	10
Exch. K (meq/100 g)	0.16	0.13	0.11	0.10	0.2-0.3
Exch. Na (meq/100 g)	0.12	0.11	0.09	0.08	0.6-1.2
Exch. Mg (meq/100 g)	2.8	0.9	0.8	0.4	3-8
Exch. acidity (meq/100 g)	4.8	2.20	2.22	2.6	0.7-2.0
ECEC (meq/100 g)	9.08	4.34	4.02	3.58	3.0
Base sat. (%)	89.17	97.3	81.1	37.7	50-100

The electrical conductivity decreased in Okra soils from 0.084 mS/cm (control) to 0.044 mS/cm (2% treatment) but increased subsequently to 0.066mS/cm (8% treatment). A similar pattern of decrease occurred from 0.066 mS/cm (control) to 0.043mS/cm (2% treatment) and then increased to 0.055 mS/cm (8% treatment) in *Telferia* treated soils (Fig. 1).

Tables 3 and 4 show the data on the chemical properties of Okra and *Telferia* soils respectively. Sandy loamy soil with particle size distribution of sand, 77.6%; clay, 14.7% and silt, 7.75%, was used for the experiment.

The amount of organic matter and total N% increased markedly with increase in crude oil concentrations for both crops; while available P, Exchangeable K, Na and Mg decreased similarly with increase in crude oil concentrations.

The Effective Cation Exchange Capacity (ECEC) decreased from 6.48 meq/100g (control) to 3.33 meq/100g soil (2% treatment) and then increased to 3.98 meq/100g soil (8% treatment). Similarly, the ECEC decreased from 9.08 meq/100g soil (control) to 4.34 meq/100g soil (2% treatment), 4.02 meq/100g soil (4% treatment) and then to 3.58 meq/100g soil (8% treatment) in *Telferia* soils. The high levels of ECEC obtained for *Telferia* soils were associated with high concentrations of magnesium recorded when compared with that of Okra soils.

Table 5 shows the levels of total extractable metals (Fe, Cu, Cd, V, and Zn) in the crop samples after the experiment. Crops from 8% treatment were killed by high concentration of the crude oil. Generally, there was a marked increase of heavy metal concentrations in crops with an increase in crude oil concentrations.

The hydrocarbon contents of the soils and crops after the experiment are shown in Table 6. THC increased with increase in crude oil concentration. In Okra soils, THC increased from 25.0mg/kg (control) to 3762.80 mg/kg (2% treatment), 7102.8 mg/kg (4% treatment) and then to 12582.80 mg/kg (8% treatment). THC increased from 25.0 mg/kg (control) to 4253 mg/kg (2% treatment), 6483.80 mg/kg (4% treatment) and then to 13943.80 mg/kg (8% treatment) in *Telferia* treated soils.

In crops, the THC concentrations increased with an increase in levels of crude oil concentrations. The THC increased from 15.28 mg/kg (control) to 72.76 mg/kg (2% treatment) and then to 257.08 mg/kg (4 % treatment) in Okra. Similarly, the THC increased from 19.92 mg/kg (control) to 520.40 mg/kg (2% treatment) and then decreased to 377.08 mg/kg (4% treatment). However, THC levels in *Telferia* shoots were seven times higher than those of Okra (Table 6).

Discussion:

Growth in control soils represent what is excepted in the crops under normal conditions (Nicolotti and Egli, 1998). The reduction in seedlings growth is attributed to the increase in crude oil concentrations. Okra growth was most affected by the oil than *Telferia* during the period and have been attributed to the permeable nature of the former compared to the impermeable structure of *Telferia* seeds. Through out the test period, *Telferia* showed delayed germination indicating acute effect of the oil on planted seeds. But for the 8% treated soils, there were no observable germination for both Okra and *Telferia* seedlings. It is therefore pertinent to infer that the crude oil concentration was too toxic for the seedlings growth at 8% treatment. The increase in soil temperature is attributed to the interference of crude oil with air circulation in the soil thereby raising the soil temperature slightly. Oil being more viscous than water has a higher specific heat capacity than water and could block air spaces in the soil.

The redox potential values are all indicative of well oxidized soils though treated with varying amounts of crude oil. Such soils are capable of sustaining sufficiently large population of aerobic bacteria that aids in the biodegradation of oil in the contaminated soils (Odu, 1980). The drop in e^H from control to 2% treatment could be attributed to the depletion of oxygen in soils polluted by crude oil as the available oxygen is used up during microbial oxidation. On the other hand, poor aeration of the soil due to crude oil application could contribute to a drop in e^H.

The increase in soil pH is attributed to the accumulation of exchangeable bases Ca^{2+}, K^+, Mg^{2+}, Na^+) in the contaminated soils (Nicolotti and Egli, 1998). The values measured are not detrimental to crops as high agricultural productivity can be obtained in soils of pH up to 6.5. The observed changes in conductivity indicate that the addition of crude oil could have affected the ionic stability of the soil which contributed to the decreased conductivity. This suggest that an increase in crude oil concentration destabilizes the soil ionic strength thereby releasing bound micronutrient into the soils.

The high organic C levels for the contaminated soils can be attributed to the oil treatments of the garden soils. In addition, the slow decomposition by facultative and obligate anaerobes may also contribute to the accumulation of organics in the contaminated soils as oxygen present is limited. The observed values (Table 3) are above the 20% critical levels required for plant growth. The available phosphorus concentration decreased in the oil treated samples for both crops probably due to the conversion of $H_2PO_4^-$ (most available form of phosphorus) to HPO_4^{2-} (less available to plants) and then to PO_4^{3-} as the pH of the soil increases (Brady and Weil, 1996). From this study it is evident that oil interferes with mineral stability and transportation and that pH increases with an increased in oil treatment (Fig. 1). All the values of available phosphorus fall within the range of 20 - 100 ppm indicating optimum levels for growth of crops.

The exchangeable cations (Ca^{2+}, K^+, Mg^{2+}, Na^+) exist in soils in an already oxidized

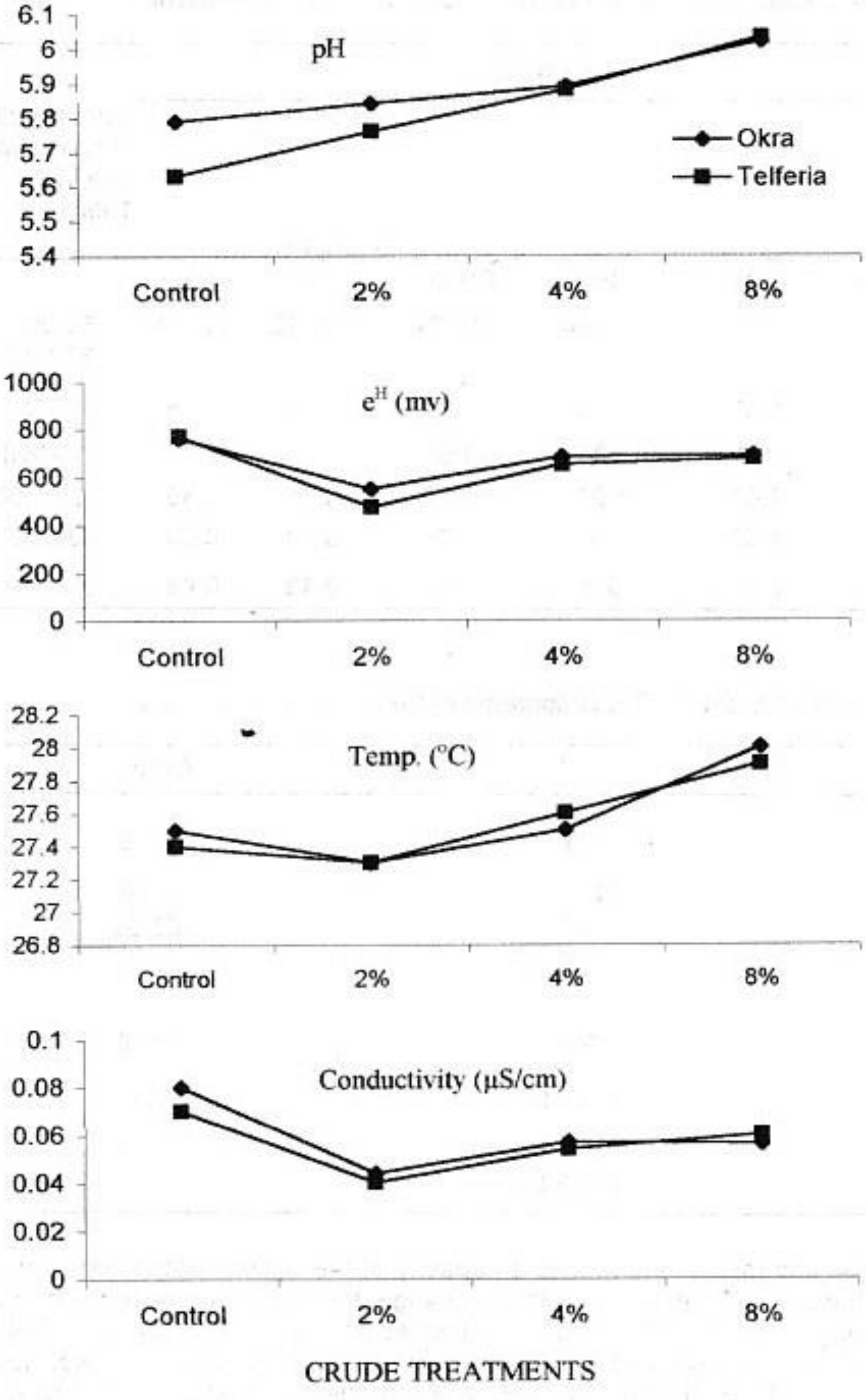

Fig. 1: The variation of physical factors in crude oil treated soils.

Table 5: Total extractable heavy metals in crops after the experiment.

	Treatment						Critical values in soils (Brady and Weil, 1996).
	Okra			Pumpkin			
Heavy metal	Control	2%	4%	Control	2%	8%	
Fe (ppm)	52.74	173.7	194.64	46.74	101.22	139.98	50,000 - 300,000
Cu (ppm)	0.9	1.08	1.08	0.84	1.14	1.15	2-100
Zn (ppm)	1.08	1.2	1.32	1.08	1.26	1.62	10-300
Ni (ppm)	0.84	1.02	1.02	0.12	0.3	0.36	10-1000
Cd (ppm)	1.44	1.56	1.56	0.24	0.54	0.54	0.01-7
V (ppm)	0.36	0.48	0.4	0.12	0.48	0.48	20 - 500

Table 6: Total hydrocarbon (THC) contents of treated soils and crops

Treatments	Soils	Crops
Okra		
Control	25	15.28
2%	3762.8	72.78
4%	7102.8	257.08
8%	12582.8	-
Pumpkin		
Control	25	19.92
2%	4253.8	520.4
4%	6564.8	377.08
8%	13,943.80	-

(stable) state. Therefore, an increase in the moisture capacity of the contaminated soils can enhance their solubility in the medium. Generally all the exchangeable bases decreased with an increase in crude oil indicating an interferes in mineral demobilisation after oil treatments. But the exchangeable calcium in Okra soils showed an increase with oil concentration attributed to the increase in soil pH of the contaminated soils. Low pH values of the soil is associated with a decrease in soil calcium levels (Brady and Weil, 1996).

In *Telferia* soils, the calcium level decreased from 1.2 meq/100g soil (control) to 1.0 meq/100g soil (2% treatment), 0.8 meq/100g soil (4% treatment) and then to 0.4 meq/100g (8 % treatment). The decrease in Ca levels could be due to *Telferia's* ability to absorb more calcium than Okra. Rice *et al*, (1993) opined that fluted pumpkin requires 84mg/g of Calcium for its stalks and gourds development. The values of Ca from uncontaminated and contaminated soils are far below the optimum requirement for agricultural productivity. The critical value of Calcium is 10 meq/100g soil.

In control and treated soils the low value of K is peculiar of soils in the tropics whose soils are low in micas, clay minerals (few exchange sites) and of pH 4.0 – 6.0 (leached by high rainfall), Feniran and Areola (1978). The values of K for contaminated and uncontaminated soils fall below the critical K level (0.2 – 0.3 meq/100g soil) required for optimum plant growth (Brady and Weil, 1996).

Soils of humid and sub-humid regions, in their natural state are generally acid with pH below 5.0. The proportion of the exchangeable acidity which is aluminium and hydrogen ions are absorbed on the soil colloids and move into the soil solution when its acidity is reduced. Aluminium if present in appreciable concentrations in the soil solution is detrimental to plant growth (Brady and Weil, 1996).

Bray and Weil (1996) defined heavy metals as inorganic pollutant particle with densities greater than 5.0 mg/m^3. Heavy metals are micronutrients but may become toxic in excess quantities. Soils rich in scavengers have the capacity to absorb, attract or bind heavy metals thus forming organometallic chelates. These chelates formed are complexed and densed preventing leaching of these micronutrients.

The higher THC in *Telferia* than Okra indicates that *Telferia* absorbed more THC than Okra. It is therefore surprising that absorbed THC concentration for *Telferia* on 4% treatment (377.08 mg/kg) was about 2 fold less than that on 2% treatment (520.40 mg/kg). This is because *Telferia* on 4% treatment was uprooted on the ninth day after germination due to deteriorating condition and preserved in the laboratory for analysis. Thus, the higher treatment delayed the germination process and also affected growth/development of both crops tremendously. Up to 300 mg/kg and 500 mg/kg of THC can be tolerated by Okra and *Telferia* seedlings respectively. Above these levels, the crops die due to chronic toxicity.

During this study, the critical level of THC concentration for optimum plant growth (100.0 mg/kg) in soils were exceeded in the oil treated soils. The results suggest the importance of *Telferia* in the bio-remediation of soils contaminated by crude oil during clean-up operations.

Conclusion

Crude oil contamination affects germination and growth of Okra and *Telferia*. These effects are more pronounced as the crude oil concentration increases. The pH of the contaminated soils were higher than control due to oil treatment. Increased crude oil concentrations in soil depressed soil conductivity and redox potential and increased soil temperature markedly. Apart from its effect on the physical properties of the soil, a number of changes also occurred on the chemical properties. Total organic carbon, total nitrogen, available phosphorus, exchangeable bases and acidity of the contaminated soils were also markedly altered to varying degrees depending on the crude oil concentrations.

The absorption of heavy metals and THC by Okra and *Telferia* increased with an increase in crude oil concentrations. Okra absorbed more of cadmium, nickel and iron whereas *Telferia* absorbed more of copper, zinc and THC. The preferential uptake of THC by *Telferia* suggest the suitability of *Telferia* for bio-remediation of oil contaminated soils.

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