**Effect of Different Levels of Crude Oil Contamination on The Germination and Soil Chemical Properties of Soil Grown with *Zea mays L.* and *Abelmoschus esculentus L.***

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**ABSTRACT**

 This study was carried out to determine the effect of different levels of crude oil contamination on the soil grown with Zea mays L. and Abelmoschus esculentus L. in a potted sandy loam soil samples. The experiment was arranged in a Completely Randomised Design (CRD) with four replicates. The factors comprised of four levels of crude oil contamination and two test crops, with each replication containing eight (8) treatments. The soil was treated with different levels of crude oil; 0% , 0.5%, 2% and 5% v/w. The control samples were not contaminated. It was observed that *Zea mays* germinated and grow in crude oil concentrations, 5%v/w, high enough to cause mortality in *Abelmoschus esculentus L*, proving a better candidate for phytoremediation of crude oil polluted soil. Crude oil at a level of 5% increased germination time in *Zea mays* by 24h but at higher dose (5%) germination of  *Abelmoschus esculentus L* seeds was totally inhibited. On the contrary, germination of *Zea mays* seeds occurred at all levels of crude oil pollution although at different rates. The germination time was 5 days in soils with 5% oil concentration. Even though *Zea mays* germinated and grow in all concentrations of crude oil tested, significant (p<0.05) shoot growth retardation still occurred in both cereal and vegetable consequent on crude oil toxicity. The ability of *Zea mays* to grow in crude oil level that was harmful to *Abelmoschus esculentus* make it better suited for growth in oil-contaminated soils and consequently a promising and potential candidate for phytoremediation of such soils than other crops. The results also showed that crude oil contaminations have negative impacts on the soil pH, mineral elements such as nitrogen, phosphorus and potassium contents, organic content, exchangeable bases and acids, and Heavy metals. The observable changes in composition of the mineral elements in the soil could be attributed to the toxic nature of the petroleum products. This study has demonstrated that soil contamination with different crude oil levels has a highly significant effect of retarding the germination of some crops, and mineral composition of soils and creating imbalance in the health status of the soil leading to infertility of the soil.

**Key words:** Zea mays L.; Abelmoschus esculentus L.; Crude Oil; Soil Chemical Properties; Germination; Hydrocarbon

**I. INTRODUCTION**

 Crude oil contamination and its associated products remain a big threat to the soil environment, and as a result a challenging issue in Nigeria as crude oil refining activities improve steadily. This challenging issue manifests particularly in the decrease/loss of fertility of agricultural lands, depletion of ecosystems, biodiversity and death of plants, including crops, in the oil producing areas of Nigeria. A report by Adoki et al,2007 on the same subject matter indicates that it hinders and retards the germination of crops, growth and yield in those areas for varying periods of time. It can be noted that crude oil hydrocarbons have harmful effects on the vegetation, germination, shoot growth and yield of most plant species including seashore plants and field grasses (Bamidele et al, 2011; Debojit et al, 2011). According to Chi Yuan et al, 1995, crude oil and its refining products consisting of aliphatic, oleic, naphthenic, benzene and its substituted cycloalkane rings and polycyclic aromatic hydrocarbons, which can change the physical and chemical characteristics of soil and its structure. These hydrocarbon compounds which are harmfully obnoxious and toxic in nature, are largely responsible for changed fertility of agricultural soils. The affected contaminated soils lose their physical, chemical and biological activities and can require some reasonable amount of periods to recover and reclaim the soil fertility (Wyszkowska et al, 2001).

 It is established that soils contaminated with crude oil harmfully affect seed germination and growth of seedlings (Bamidele, 2010; Lin et al, 2009), and that no two or three plants are expected to show the same measure of resistance or susceptibility to crude oil. What makes the difference is the inherent property of each plant, and seed phytochemistry may be a good pointer to these properties. The two plants used in this research work: *Zea mays* and *Abelmoschus* *esculentus* are staple crops used for food and livestock feeds in different sections of Nigeria. *Zea mays* is one of the major staple cereal crop planted for food, feed and industrial purposes (USAID, 2010; Oyewo et al, 2011)*. Zea mays* is also a key source of carbohydrate and vitamins in human food. A useful quality of vitamins C and A can be derived from white and yellow grains respectively (Agoda et al,2011). For industrial purposes, *Zea mays* grain can be used as livestock feeds and also serve as a rawmaterials for starch, flour and alcohol production (Oyewo et al, 2011; Agoda et al 2011). *Abelmoschus* *esculentus* simply known as ladies finger is one of the main vegetable crops which contain sufficiently good amount of vitamins especially vitamin A and other minerals including folic acids, besides carbohydrates, phosphorus, magnesium and potassium. The research study is to determine the effect of different levels of crude oil contamination on the germination and chemical properties of soil grown with *Zea mays L*. and *Abelmoschus* *esculentus L*.

**II. MATERIALS AND METHODS**

 The experiment was conducted in the Screen house of Department of crop/soil science, Faculty of Agriculture, Rivers State University, Portharcourt, Nigeria**.** The study was a 4 x 2 factorial experiment arranged in a completely randomised design (CRD) with four replicates. The factors comprised of four (4) levels of crude oil contamination (0%, 0.5%, 2% and 5%) and two test crops (Maize and okro), with each replication containing eight (8) experimental units/pots totalling thirty-two pots. The possible combination of the two crops and four different levels of crude oil gave a total of eight (8) treatments. The treatments are C1Ma, C2Ma, C3Ma, C4Ma; C1Ok, C2Ok, C3Ok, C4Ok and the Control. The crude oil was obtained fresh from the production plant of Nigeria National Petroleum Cooperation (NNPC) Port Harcourt Refinery, Alesa-Eleme, Rivers state, Nigeria. Sandy loam soil was obtained from the Teaching and Research farm of Rivers State University, Port-Harcourt. The soil sampled had no previous exposure to crude oil contamination. The viable seeds of maize (*Zea mays*) and okro (*Abelmoschus* *esculentus*) were sourced from the gene bank section of International Institute of Tropical Agriculture (IITA), Ibadan and Rivers State Agricultural Development Programme (ADP) and stored at a room temperature (25-30oC) for 24hrs.Viability tests of the seeds were checked through submerging the seeds in water (by floatation method). The soil samples were air-dried, sieved and dispensed in 5kg weights into thirty-two (32) plastic bucket perforated at the bases. The buckets were grouped into four replications of eight each, and each group was used for the two of the crop plants. Each plastic bucket, containing the soil samples, in a group was contaminated and mixed with one of four different levels of light crude oil ( 0% , 0.5% , 2.0% , and 5% v/w). All control samples were not contaminated and planted. All buckets with soil samples and crude oil were allowed for a week before planting. Thereafter, viable seeds of the crops (maize and Okra) were planted in the buckets and watered every three days by spraying. The experiment lasted for a month and was repeated twice. Data were collected on the following: Germination percentage (%), and Soil PH (Electrode method)**,** Organic carbon (Black,2000), Available phosphorus (Bray II method) (Bray et al, 1985), Total Nitrogen (modified kjeldahl method**)**, Heavy metal content (Atomic Absorption Spectrophotometer), Exch. bases such Ca, Mg, Na, K (titration method and flame photometer) and Exch. acids (titrimetric method). Research data obtained were simply subjected to analysis of variance (ANOVA), while the significant means were separated with least significant difference (LSD) at 5% confidence level (p<0.05).

**III. RESULTS.**

**A. Effect of crude oil on germination of seeds of test crops**

 Crude oil used in this research work had significant effect on the seed germination of the test crops. The oil significantly (p<0.05) suppressed the germination of seeds of the test crops. The effect of crude oil on seed germinations was simply presented in the Table 1 and 2. Crude oil suppressed germination of some seeds especially that of Okro seeds. Germination of okro was totally inhibited at crude oil levels of 5.0% (v/w). In maize, however the seeds germinated in all the levels of contamination (0% to 5% v/w) but at different rates of germination: 4 days for 0 to 2%, 5 days for 5%. In the two test crops especially maize, germination time was increased by a day as from 5% crude oil contamination. One hundred percent (100%) germination was noticed in all the two test crops only at 0 to 0.5% crude oil contamination; in maize, 100% germination occurred in soils that contained up to 0.5% crude oil contamination (Table 2). At 2% crude oil contamination, only 67% of Okro and maize germinated. Even though maize seeds germinated in crude oil level of up to 5%, only 33% of the maize seeds were able to germinate at that level, not the same for okro seed which were unable to germinate (Table 2).

**B. Effect of crude oil contamination on chemical properties of the soil grown with the test crops**

The chemical properties of the soil on the experimental pot before and after contamination are presented in Table 3. Crude oil significantly(p<0.05) affected some of the chemical properties of the soil contaminated, and planted with the test crops. The soil, an acidic sandy loamy soil with no previous hydrocarbon content, had originally pH of (6.17), nitrogen (0.134%), available phosphorus (69.93 mg/kg) and organic carbon (1.84%). On simulation with crude oil, it was observed that the soil pH was not significantly affected, and organic carbon content of the soil increased significantly with increased level of contamination while available phosphorus significantly reduced to 51.12 and 48.99 mg/kg in the 5% soil treatment in maize and okro respectively. Also, the total nitrogen content was slightly increased in 5% polluted soil to 0.17 and 0.22% for maize and okra respectively, 4 weeks after planting.

 The pH of the soil before contamination was 6.17 slightly lower than the pH of the soils at the end of the study for the different treatments. In the 0.5% to 5% treated soils, the pH was slightly increased to 6.26 for maize, 6.25 for okro compared to the pH before contamination.

 The increased levels of crude oil contamination to the soils also increased the percentage organic carbon content of the rhizosphere soils of Maize (4.70) and okro (4.88) compared to the soils before contamination which is 1.84. There was a decrease in available phosphorus with the application of increased crude oil compared to the result of the control for the rhizosphere soil of the test crops. Analysis of variance showed that effect of different treatments on available phosphorus of the rhizosphere soil of the test crops was significantly (P<0.05) different. Available phosphorus of the planted soil also reduced with the application of crude oil to 51.12 in maize, and 48.99 in okro. There were significant reductions on the sodium (Na), potassium(k) and magnesium elements in the planted soil of each crops compared to the uncontaminated soil.

 The details of the heavy metal concentration in both the contaminated and uncontaminated rhizosphere soil of the test crops (maize and okro) were also shown in Table 3. It was observed that crude oil contamination significantly (p<0.05) affected the individual elements of heavy metal concentration in the soil planted with the test crops. The table showed the concentration of lead (Pb) in all the test crop soils. High concentration of both metals were observed in contaminated soil of maize (5.07) while low concentration was observed in polluted soil of okro (4.89). Higher concentration of Fe was observed in polluted maize and okro (1262 and 1397) as compared to unpolluted soil (923.5). Similar trend was observed in cowpea with concentration of (1314). Higher concentration of manganese was observed in contaminated maize (389.7) while the lowest concentration of manganese was observed in uncontaminated soil of the test crops (253.60). The same trend was observed in polluted soil of okro (393.8) when compared with uncontaminated soil. However, higher concentrations of these heavy metals imposed by the crude oil contamination have negative effects on the soil and the crops. Highest concentration of zinc was observed in contaminated soil of maize (85.76) while the lower concentration was observed in uncontaminated soil of the crops (67.55). Similar trend was observed for okro (102.7). Higher concentration of Copper was observed in contaminated soil of maize and okro (24.3, and 24.66).

**IV. DISCUSSION**

**A. Effect of crude oil contamination on the germination of test crops**

 Crude oil contamination at high concentrations as seen in Table 2 had a significant (p≤0.05) negative effect on the germination of test crops. From 5% level of crude oil contamination, there was complete inhibition of germination in Okra. In maize, germination occurred at 5% crude oil level. Maize, however exhibited a remarkable degree of tolerance for the oil by germinating in all the levels of contamination. The inhibition of germination that was observed in okro at 5% level of crude oil is in tandem with the findings of (Malek-Hossein et al, 2007) who studied the effect of crude oil on the germination of *Medicago sativa* (alfalfa) and he observed germination inhibition at high doses of crude oil. His observation also confirmed the report of (Amadi et al, 1996) that high doses of petroleum hydrocarbon can inhibit germination in some crops. The inhibition of germination on some crops occurred as a result of the toxic nature of the inherent components of the hydrocarbon compounds. Hydrocarbon tends to create unhealthy soil conditions for the germination of crops by reducing aeration, infiltration rates of water and depleting microbial characteristic in the soil.

**B. Effect of crude oil on the chemical characteristics of the soil**

Crude oil undoubtedly had significant effects on some chemical properties of the soil. Organic carbon, for example, increased significantly (p<0.05) as the crude oil levels increase. This increase in crude oil levels occurred as a result of the introduction of crude oil into the soil. As the organic carbon increased under normal circumstances, it was expected to reduce the soil fertility and quality. This is because the increase in organic carbon was crude oil-associated it rather reduce the soil quality and fertility. It would be observed that the crude oil sealed up soil pore spaces, and as a result preventing the infiltration and movement of water and air into the soil, reducing the biological activities and disrupting biochemical conversions in the soil necessary to make essential nutrients and some minerals available to the crop plants. Soil pH was not significantly affected by the oil. Reduced soil pH, increases in soil organic carbon and organic matter, sodium (Na), iron (Fe) were observed in crude oil contaminated soils (Chibuike et al, 2013). According to observations made by Obire et al, (2002), soil sodium and iron increased in crude oil-contaminated soils. Crude oil had a dose-dependent inhibitory effect on both the macro and micro elements levels in the soil, with the exclusion of organic carbon which increased as the crude oil was progressively introduced and subsequently increased. There were significant reductions (P<0.05) in the levels of soil Na, K, Mg and Ca with the introduction of crude oil levels, when compared to their levels in the uncontaminated control. Soils contaminated with crude oil is richly endowed with heavy elements such as Fe, Cu, Zn, Mn, Pb and so on (Table 7). A look at the Table 7, high concentration of heavy metals was observed in all contaminated soils. Among all the heavy metals detected in the soil, high concentration of lead (pb) was observed in polluted soil of maize (5.07) while the low concentration of lead was observed in that of okra (4.89) compared to the control. This is a strong prove that maize is a good hyper-extractor which can be used in the process of phytoremediation of crude oil contaminated soil. These heavy elements are characterized by the possession of partially filled d or f-orbitals in any common oxidation state (en. Wikipedia.org/wiki/transition-metal). Based on the report of (Marinescu et al,2010), through these free orbitals contained in heavy elements, they form coordinate bonds and complexes with ligands such as NH3, H2O, NO2 etc or other elements such as K, Ca, Mg etc (Marinescu et al, 2010). When such complexes and bonds happened, the bound ions lost their ionic properties and will be undetectable in solution. Available phosphorus was significantly(p<0.05) reduced as the crude oil levels increased. According to (Okoro et al,2005), it also reported that crude oil contaminations decreased the levels of soil nitrate and phosphorus but the effects on other macro-elements remained uninvestigated. A research work conducted by Wyszkowska et al, (2001) on the crude-oil contaminated soils reported that there was an increase in the levels of nitrogen (N), phosphorus (P) and potassium(K) but in their work the soil was amended with inorganic fertilizer (N.P.K compound fertilizer). Some of these unessential mineral elements such as Ca, Mg and P are required for seed germination and plant growth (Cunningham et al, 1996). However, this ability of maize to withstand the toxicity of hydrocarbon compounds and grow in concentrations that caused death in other crops makes it a possible candidate for the phytoremediation of crude oil contaminated soils. This is because for any crop to be used in the bioremediation process(es), it must possess the ability and capacity to germinate and grow in the hydrocarbon contaminated soil(s). According to Chin-A-woeing(1998), phyto-remediation could be defined as the process by which biological techniques in terms of the use of plants and their associated micro-organisms are used for the purpose to remediate contaminated soil and water.

**V. CONCLUSION**

 According to results of this research work, maize resisted the toxic effects of hydrocarbon compounds (crude oil) more than okra plants. This was observed by its ability to germinate and grow in crude oil concentrations high enough to cause the death of okra. This inherent ability of maize to germinate and grow in crude oil makes it a promising and potential candidate for the phytoremediation of crude oil-contaminated soils since the benefit of any crop in the phytoremediation of a contaminated habitat is determined by its ability to grow in the contaminated habitat. The important findings from this research work include: that all the test crops- maize and okra can germinate and grow in soil contaminated with crude oil at lower levels of contamination. It was observed that higher level of crude oil contaminations has harmful effect on some of the test crops under study. But maize (*Zea mays)* except okro exhibited better growth and germination even at high concentration of crude oil, so the crop(maize) can be used for phytoremediation of crude oil contaminated soils; the results gotten from soil chemical analysis imply that crude oil creates adverse and unfavourable condition to the soil status and composition which made oil contaminated soils unsuitable for cropping. The results also showed that crude oil contaminations have unsuitable impacts on the pH, mineral elements (Nitrogen, phosphorus and potassium contents), organic content, and microbial load of soils contaminated with petroleum. Heavy metal contents of the soil were also affected. Some metals increased in the soil as the crude oil contamination levels increased having harmful effect to the health of the soil and crop plants.

**VI. RECOMMENDATIONS**

 There is need for further research to unravel the factors responsible for the variation observed in degrees of susceptibility to crude oil toxicity between maize and okro; and also, the factors responsible for the depletion of soil health quality in crude oil contaminated / polluted areas.

**TABLE 1. Effect of different levels of crude oil contamination on germination time of the test crops.**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Crop plants |  0% |  0.5% |  2.0% |  5.0% |
| Maize | 4 ± 0.5 | 4 ± 0.5 | 4 ± 0.5 | 5 ± 0.6 |
| Okro  | 4 ± 0.5 | 4 ± 0.5 | 4 ± 0.5 |  X |

 Maize, and Okro LSD0.05 = 4.04,

 Significant at 5% (p<0.05),

values given as mean ± S.E

x = No germination.

**TABLE 2**. **Percentage germination of the plants in sandy loam soil polluted with varying concentration of crude oil.**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Crops | 0% | 0.5% | 2% | 5% |
| Maize | 100 | 100 | 67 | 33 |
| Okro | 100 | 100 | 67 | X |

x =No Germination

**TABLE 3. Chemical properties of non-polluted and polluted rhizosphere soil planted with maize and okro**

|  |  |  |  |
| --- | --- | --- | --- |
| Soil Properties | Before Pollution |  Maize (% v/w)0 0.5 2 5 | Okro (% v/w)0 0.5 2 5 |
| **PH** **Organic Carbon (%)****Total Nitrogen (%)****Available P. (mg/kg)****Exch. Bases (mol/kg)**  C Mg Na K**Exch . Acidity****Heavy Metals** Fe  Cu  Zn Mn Pb | 6.171.840.13469.932.271.200.060.081.56923.518.2067.55253.604.90 | 6.11 6.21 6.23 6.261.58 2.52 2.91 4.700.09 0.16 0.13 0.1767.76 53.52 52.2 51.122.31 2.51 2.72 2.531.16 1.15 1.21 1.110.05 0.04 0.06 0.050.08 0.07 0.06 0.061.52 1.71 1.42 1.611042 1051 1122 1262 20.3 19.36 22.5 24.3 82.2 85.6 82.7 85.76373.7 347.12 371.2389.75.01 5.02 5.01 5.07 | 6.11 6. 15 6.24 6. 251.12 2.91 2.04 4.880.07 0.14 0.12 0.2268.67 53.66 51.66 48.992.35 2.41 2.71 2.491.14 1.08 1.21 1.310.05 0.04 0.05 0.040.08 0.07 0.07 0.061.51 1.64 1.60 1.671085 1022 1126 1397 22.16 24.36 22.3 24.6683.7 100.4 85.36 102.7373.7 332.1 398.1 393.8 5.00 5. 00 4.89 5.04 | Cowpea (%v/w)0 0.5 2 5s | OKro (%v/w)0 0.5 2 5 |

Maize ,Okro ; LSD0.05 for pH= ns, ns ;

Organic Carbon = 2.42 , 0.7 :

Available Phosphorus = 2.20 , 0.93 ;

Total Nitrogen = 0.20 , 0.4

Exchangeable Acids = 1.08,0.53 ;

Iron(Fe) = 4.12, 3.90;

Copper (Cu) = 1.72, 0.52 ;

Lead (Pb) = 0.32, 0.20

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