

## Phytotoxic Effect of Oilfield Wastewater on the Physiology and Chlorophyll Content of Some Crops

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**ABSTRACT** The phytotoxic effect of various concentrations (0%, 25%, 50%, 75% and 100%) of oilfield wastewater on the physiology of *Zea mays* (Maize), *Abelmoschus esculentus* (Okra), *Arachis hypogea* (peanuts) and *Telfairia occidentalis* (Fluted pumpkin) commonly cultivated in the Niger Delta was investigated. This was achieved by the determination of the plant height and girth, leaf length, width and area, root density, chlorophyll content, crude fibre and total ash using standard scientific methods. Results of the study showed that the girth and height of *Z. mays* were highest at 50% on the 56<sup>th</sup> day after planting (DAP). Generally, the leaf area was highest at 50% concentrations except at the 14<sup>th</sup> DAP. The control plants were consistently taller except at 50% for *A. esculentus*. Leaf area was highest in the control at 14<sup>th</sup> DAP and at the 50% concentration at 56<sup>th</sup> DAP. Leaf width and area of *T. occidentalis* were highest at 50% concentration. Generally, the height and girth of *A. hypogea* were found to decrease with increasing concentration. The leaf length, width and area of *A. hypogea* had the highest value at 56<sup>th</sup> DAP in the control. Except at the 100% concentration, the leaf area was lower in the control than in all other treatment for *Telfairia occidentalis*. Except at the 50% concentration, the leaf area was higher in the control than in all other treatments for *Zea mays*. Generally, except at the 50% concentration, the leaf area was higher in the control than in all other treatment for *Abelmoschus esculentus*. There was a reduction in the size and number of leaves produced by *Arachis hypogea* which became less luxuriant with increasing concentration of oilfield wastewater. Generally, all the physiological parameters of the plant analyzed were higher in the control plants than in plants of the treated soils. The height and girth were found to decrease with increasing concentration of the oilfield wastewater except that there was an increase in girth at the 100% concentration. Statistical analysis using one way ANOVA showed that there was significant difference at  $P = 0.05$  in the physiological characteristics of the various plants. Chlorophyll content was highest for *T. occidentalis* at the 75% concentration, in the control for *A. es-*

*culentus*, and at 50% for *Z. mays* and *A. hypogea*. Fibre content and total ash for *Zea mays* was highest in control and at the 100% concentration respectively. Absorption of total hydrocarbon and other hazardous constituents of the oilfield wastewater by the plants which serve as sources of food for humans pose a serious health hazard. The oilfield wastewater has been shown to have serious deleterious effect on the physiology of the plants which will surely lead to agro production problems that could create an artificial food scarcity due to damage to crops and other vegetation.

*Keywords:* Oilfield wastewater, crops, chlorophyll content, plant height, root density, food scarcity.

## Introduction

Crude oil (including refined petroleum products) is not the only pollutant of the environment from petroleum exploration and productive activities. The oilfield wastewater resulting from crude oil exploration activities is by far the largest volume of by-product or waste stream associated with oil and gas production (Elcock *et al.*, 2002) and is one of the major pollutants of both terrestrial and aquatic environments (Wills, 2000; Obire and Amusan, 2003). Offshore oil production platforms as well as oil-fields discharge the oily wastewater into the environment as part of their normal operations. Accidental or deliberate discharge may also result in the release of oilfield wastewater onto fallow farmland due to faulty construction, deterioration or absence of synthetic impermeable lines. A fault or crack could develop in the wastewater stabilization ponds leading to soil contamination (Ream, 1983). The composition of produced water from one site to another is highly variable, containing different amounts of solids, heavy metals, and other inorganic compounds, as well as a wide spectrum of organic compounds (Bernal *et al.*, 2007). Koons *et al.*, (1977) reported that the relative amount of hydrocarbons contributed to the aquatic environment by oilfield wastewaters is so small that any overall environmental effects from this inputs should be minimal. However, numerous inorganic and organic constituents dissolved in the produced water can be potentially or actually far more hazardous than the crude oil itself (Wardley-Smith, 1979; Scott *et al.*, 2007).

Perhaps the most ecologically damaging types of oilfield related spills are those which involve releases of oil field brine from buried injection lines. Since there is often no petroleum-related sheen associated with spills of these highly saline fluids, they can go unnoticed initially, becoming evident much later when overlying vegetation begins to show signs of stress or dies (Benmoussa and Achouch, 2005). Ajayi and Osibanjo (1981) have shown

the ability of produced water constituents to alter habitat integrity of the natural bodies. Of the major economic sectors, the ones most vulnerable to environment pollution or degradation are agriculture and fisheries, mainly because they are dependent on natural systems and resources with attendant problem of slowing down growth in the world's food output.

The Niger Delta is home to several petroleum and petroleum related exploration activities and petroleum industries. The intense industrial activities have attracted a lot of research interest especially in petroleum hydrocarbons in soil and aquatic environments and in organisms. However, most of these researches are classified information and as a result, there is little or no literature on the impact of oilfield wastewater on the environment and on the physiology of plants that is available to the public. It is therefore necessary to carry out investigation on the impact of oilfield wastewater on the physiology of some plants. Different cells and tissues of a plant are physically and chemically specialized to perform different functions. Chlorophyll is a chemical compound and the primary pigment in plants. The presence and relative abundance of chlorophyll gives plants their green color. Chlorophyll serves as the primary means for plants to intercept light to fuel the process of photosynthesis. The leaf surface area determines in large part the amount of carbon gained through photosynthesis for growth (Watson, 1947) and the amount of water lost through transpiration and ultimately the crop yield. In many crop growth studies leaf area index have been reported to be related to both biological and economical yield. Several methods which include the relationship between the leaf area and the dry matter of either certain plant parts or the whole above ground plant (Aase, 1978) have been used to determine the leaf area in crop plants. Roots function to anchor the plant and acquire minerals in the soil. Leaves trap light in order to manufacture nutrients. For both of these organs to remain living, minerals that the roots acquire must be transported to the leaves, and the nutrients manufactured in the leaves must be transported to the roots.

Stress from the chemical constituent of wastewater can lead to changes in the way a plant functions. The Objectives of this study therefore, were to determine the effect of various concentrations of oilfield wastewater on the physiology of *Arachis hypogea* L (Groundnut), *Abelmoschus esculentus* L (Okra), *Telfairia occidentalis* Hook F (Fluted pumpkin "Ugu") and *Zea mays* L (Maize). These crops were chosen due the fact that they are commonly grown, cultivated and consumed in the Niger Delta region (FPDD, 1989; Akande *et al.*, 2006). They also represent three (3) major plant groups – legumes, vegetables and cereals. This study will therefore help to unravel how these crops respond to conditions and variations in the concentration of oilfield wastewater. This will in turn unravel the phyto-toxic effect of oilfield wastewater on these crops and on agricultural productivity.

## **Materials and Methods**

### *Collection of Oilfield Produced Water Samples*

Freshly treated oilfield produced water samples were collected from the outlet of the separation/treatment plant of an oilfield flow station located at Kwale Community in Ndokwa East Local Government Area of Delta State. Sterile plastic Jerry cans were used to collect the produced water samples. Prior to collection of the produced water, the interior of the nozzle of the outlet valve was flushed by allowing the produced water to flow to waste for 2 to 3 minutes after which the Jerry cans were filled from a gentle stream of the produced water.

### *Collection of Soil Samples*

Soil Samples were collected from a depth of 0 - 15cm with the aid of a brand new shovel sterilized with ethanol and bulked together to form a composite sample. The composite soil was sieved through a 2mm sieve as to remove debris that would otherwise absorb the treatment that will be experimentally performed on the soil and plants. Thereafter, one Kilogramme (1kg) of the sieved soil was weighed out using Analytical weighing balance (AL 2105 Germany) and put into a black polythene bag. A total of 120 polythene bags with sieved soil samples were prepared and kept in a Green house.

### *Collection of Plant Seeds, Viability Test and Preliminary Study on the Plants*

The plant seeds of *Arachis hypogea* L, *Abelmoschus esculentus* L, *Telfairia occidentalis* L and *Zea mays* L were purchased from the Mile 3 market in Diobu, Port Harcourt. The seeds were collected in sterile polythene bags and transported to the laboratory. The viability test as described by Amakiri and Onofeghara (1984) was used to ascertain the viability of the various seeds before they were used for the study. After a preliminary study of the effect of the concentrations of oilfield wastewater on the plants, the 8<sup>th</sup> week (56 days after planting) was taken as the terminal age of plants at which sampling for analysis was carried out.

### *Determination of Water Holding Capacity*

The water-holding capacity of the soil was determined with reference to Bouyoucos (1951). The volume of each concentration of oilfield formation

water used for the treatment was 60% of the water holding capacity of the soil.

### ***Experimental Design Physiological Parameters Analyzed***

The experimental design was the randomized complete block design (RCBD). The five different concentrations of the oilfield wastewater used as treatments for the study were [0% (control), 25%, 50%, 75%, and 100%]. The 0% treatment served as control for the treated plants. All the treatments for each plant seed was replicated four times (i.e.  $5 \times 4 = 20$ ). Each experimental polythene bag was appropriately labeled with the concentration of oilfield wastewater to be used as treatment. Treatment with the different concentrations of oilfield wastewater commenced two (2) weeks after the seeds were sown. This was to allow the seeds to germinate before the treatment. Treatment volumes of 60% water holding capacity were repeated at weekly intervals until after 8 weeks during which the physiological parameters of the plants were analyzed. The physiological parameters determined were plant height (length), plant girth, leaf length, leaf width, leaf area and the root density, chlorophyll content, crude fibre (organic residue) and total ash (inorganic residue).

### **Procedure for Growth Measurement in the Plants**

#### ***Linear measurement of plant height, girth, leaf length, width and calculation of leaf area index***

The procedure as outlined in Akonye and Nwauzoma (2003) for linear measurements of plants was used. Plant height, girth, leaf length and width were obtained by this method. Root density was determined by visual observation after the plants were carefully dug up from the soil. Thereafter, the soil surrounding the roots was carefully washed off to expose the roots. The leaf length and width were obtained by direct measurement. The average of 10 calculated leaf area indices was accepted on the leaf area index of the crop and used for subsequent calculation of the leaf area. The leaf area index factor was also calculated for each plant. Ten leaves of various sizes were collected for the leaf index determination. A 0.02cm transparent linear graph was placed over the leaf and the square spaces counted (1 square space =  $2\text{cm}^2$ ) and totaled on the area of the leaf. The estimated leaf area (A) was expressed in terms of the leaf length (L), leaf width (w) and the leaf area index factor (a):  $A = L \times w \times a$

### *Determination of Total Chlorophyll*

The chlorophyll from the samples was extracted with aqueous acetone and transfused into ether and the optical density measured at 600 and 643nm (Stewart, 1974).

### *Determination of Crude Fibre*

Three gram (3.0g) of sample was weighed into a 1-litre conical flask. Petroleum ether, was added, swirled and left to stand and carefully decanted. This was repeated twice leaving the last quantity of solvent in contact overnight, with a small watchglass over the mouth of the conical. The solvent was carefully decanted to avoid loss of particles of fibre and then warmed gently to remove visible solvent. Further chemical analysis and incineration was performed on the residue and the crude fibre content was calculated (AOAC, 1984).

### *Calculation*

$$\% \text{ fibre} = \frac{\text{Loss in weight from incineration}}{\text{Weight of Sample before defatting}} \times 100$$

### *Interpretation*

This is an empirical method and results depend on adherence to the exact conditions of test.

### *Determination of Total Ash*

The total ash was determined using the method of AOAC (1984). The mechanical convection oven (Gallenkamp), United Kingdom was used.

## **Results**

The mean values of the plant height and girth, the leaf length, width and Area (physiology) of *Telfairia occidentalis* (Fluted pumpkin), *Zea mays* (Maize), *Abelmoschus esculentus* (Okra), and *Arachis hypogea* (peanut) with the various treatments used in this study are shown in Figure 1. The results indicate that, the higher the concentration of the oilfield wastewater the greater the impact on the physiology of the plant. Generally, there was a reduction in root density of all the crops with increasing concentration of oilfield wastewater.

On observation of the impact of the concentrations of oilfield wastewater on the morphology of *Telfairia occidentalis* 56 days after planting showed that the internodes were longer with increasing concentration of oilfield wastewater. The height and girth of *T. occidentalis* were highest at the control treatment and the leaves were very shiny indicating hydrocarbon absorption from treated soils. Except at the 100% concentration, the leaf area was lower in the control than in all other treatment. Statistical analysis using one way ANOVA showed that there was significant difference at  $P = 0.05$  between all the treatments for girth, leaf length and width of *Telfairia occidentalis*.

The plant height and girth of *Zea mays* was highest at 50% concentration (24.2cm and 3.2cm respectively) on 56<sup>th</sup> day after planting (DAP). Except at the 50% concentration, the leaf area was higher in the control than in all other treatments. Statistical analysis using one way ANOVA showed that there was significant difference at  $P = 0.05$  between all the other treatments and at 100% for height, and between all the treatments for girth and width of leaf of *Zea mays*.

The mean value of leaf area of *Abelmoschus esculentus* was highest in the control and in 50% concentration at 14th and 56th DAP respectively. Generally, except at the 50% concentration, the leaf area was higher in the control than in all other treatment. Statistical analysis using one way ANOVA showed that there was significant difference at  $P = 0.05$  between all the treatments for all the physiological parameters of *Abelmoschus esculentus*

There was a reduction in the size and number of leaves produced by *Arachis hypogea* which became less luxuriant with increasing concentration of oilfield wastewater. The greatest impact was observed in the plant from the 75% concentration. Generally, the plant height and girth, leaf length, width and area were higher in the control plants than in plants of the treated soils. The height and girth were found to decrease with increasing concentration of the oilfield wastewater except that there was an increase in girth at the 100% concentration. The leaf length, width and area also decreased with increasing concentration of the oilfield except on the 56<sup>th</sup> DAP. Statistical analysis using one way ANOVA showed that there was significant difference at  $P = 0.05$  between all the other treatments for height, girth, leaf length, width and area of *Arachis hypogea*.

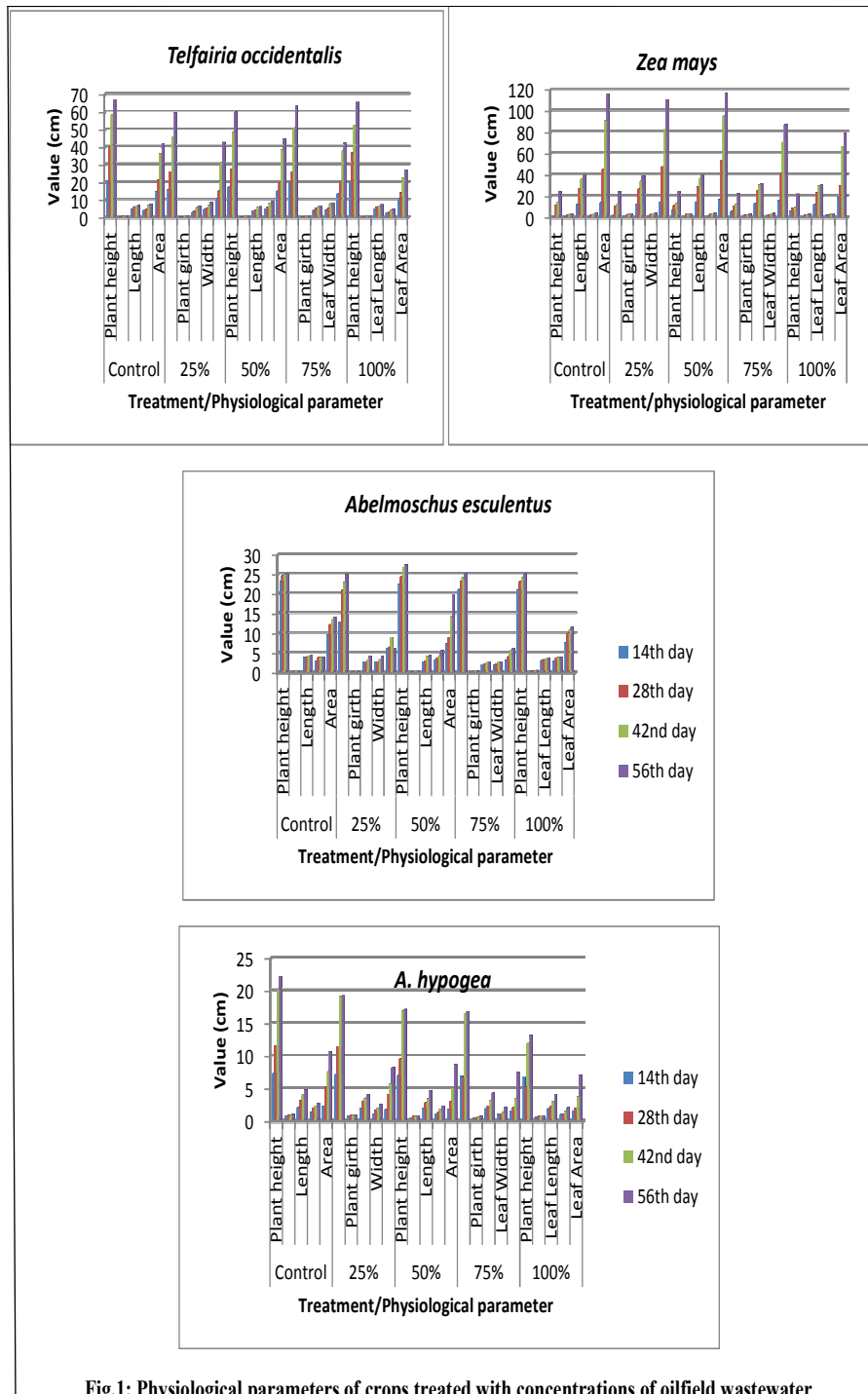


Fig.1: Physiological parameters of crops treated with concentrations of oilfield wastewater



The values of the chlorophyll content, total ash and crude fibre of the various crops with the various treatments are shown in Fig. 2. Chlorophyll content of *Abelmoschus esculentus* generally decreased with increasing concentration of the oilfield wastewater and except at 75% concentration that of *Telfairia occidentalis* decreased with increasing concentration of oilfield wastewater. On the other hand, except at the 75% concentration, the chlorophyll content of *Zea mays* and *Arachis hypogea* increased with increasing concentration. Generally, the crude fibre content was higher in the control of all the plants except in *Telfairia occidentalis* where the reverse was the case. The control recorded the highest and lowest total ash for *Arachis hypogea* and *Zea mays* respectively. On the other hand, except at 75% concentration control recorded the highest total ash for *Telfairia occidentalis* and except at 50% concentration control recorded the lowest total ash for *Abelmoschus esculentus*.

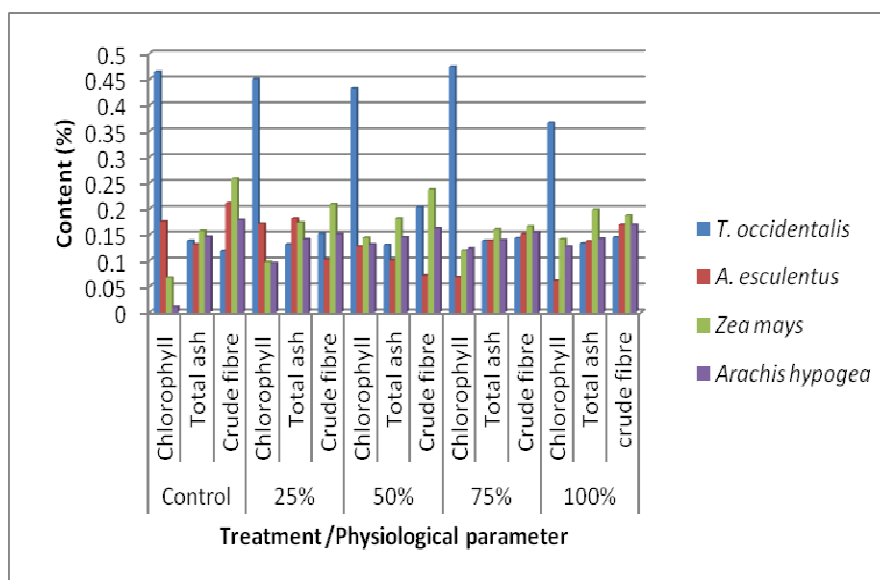


Fig. 2: Chlorophyll content, total ash and crude fibre (%) of crops treated with oilfield wastewater

## Discussion

The present study has unraveled the phytotoxic effect of various concentrations of oilfield wastewater on the physiology of *Zea mays* (Maize), *Abelmoschus esculentus* (Okra), *Arachis hypogea* (peanuts), and *Telfairia occidentalis* (Fluted pumpkin) used in this study. A comparison of the physiological characteristics in the control and treated crops showed that each crop

responded differently to the various concentrations of oilfield wastewater in the soil. The Plant height and girth of *Telfairia occidentalis* with the various treatments showed that the height of the plant was consistently, highest at the control. The leaf length, width and area of *Telfairia occidentalis* with the various treatments showed that leaf width and area was highest at 50% concentration. The Plant height and girth of *Zea mays* with the various treatment showed that the plant got to its highest height at 24.2 on 56<sup>th</sup> day after planting (DAP) at 50% concentration. The girth of the plant was also greatest at the 50% concentration of oilfield wastewater. The leaf length, width and Area of *Zea mays* with the various treatments showed that generally, the leaf area was highest at 50% concentrations except at the 14<sup>th</sup> DAP. The leaf area was best at 50% concentration of oilfield wastewater at 56<sup>th</sup> days after planting.

The plant height and girth of *Abelmoschus esculentus* with the various treatments showed that except at the 50% concentration, the control plants were consistently taller than in other concentrations. The leaf length, width and Area of *Abelmoschus esculentus* with the various treatments showed that, at 14 DAP, the values for leaf area was highest in the control. At 56 DAP; the leaf area was also highest at the 50% concentration of oilfield wastewater.

The plant height and girth of *Arachis hypogea* were generally higher in the control than with other treatment. The height and girth were found to decrease with increasing concentration of the oilfield wastewater. The leaf length, width and area of *Arachis hypogea* showed that the control had the highest values. This is an indication that the oilfield wastewater had an impact on these crops (Biran *et al.*, 2000).

Generally, there was a reduction in root density of all the crops with increasing concentration of oilfield wastewater. An alkaline soil is detrimental to plant growth (Clemente *et al.*, 2003) and root establishment. Odeigah *et al.*, (1997) showed that root lengths decrease with increasing concentration of oilfield wastewater. This agrees with the work of Pezeshki *et al.*, (1987) that increase in salinity as in the case with oilfield wastewater can affect the adsorption proportion of the roots. There was a general variation between the control and soil of the various concentrations in this study. Wardley-Smith (1979) reported that organic constituents dissolved in the produced water can be potentially or actually for more hazardous than the crude oil itself.

The chlorophyll content of the various plants with the various treatments showed that except for the 75% concentration the chlorophyll content of *Telfairia occidentalis* was generally lower in plants from the treated soils than in the control. While the chlorophyll content of *Abelmoschus esculentus* decreased with increasing concentration of the oilfield wastewater. The increased values of the constituents of the oilfield wastewater have been re-

ported to have negative effects on plant growth (Clemente *et al.*, 2003). The soil quality is known to reduce with oil pollution.

On the other hand, the chlorophyll content of *Zea mays* and *Arachis hypogea* were generally higher in plants from the treated soils than the control. This deviation agrees with the works of Amakiri and Onofeghara (1984) which states that sub-lethal doses of pollutant can sometimes elicit remarkable effects. This is an indication that *Zea mays* and *Arachis hypogea* were not stressed in the ability of their leaves to synthesize chlorophyll at the concentrations of oilfield wastewater. This is a proof that different crops or plants can respond differently to different concentrations of a particular pollutant (Akande *et al.*, 2006). This is an indication that the different plants had optimum photosynthetic activities at different concentrations of the oilfield wastewater and also respond differently to various concentrations of the oilfield wastewater. Benmoussa and Achouch (2005) had reported that, different plants had optimum photosynthetic activities at different concentrations and also respond differently to a pollutant. Reduction in chlorophyll results in reduced productivity and yield of a plant.

The crude fibre and total ash of the various plants with the treatments showed that although the crude fibre in *Telfairia occidentalis* was lowest in the control, its total ash was however higher than those of the plants in the treated soils. This was also reported by Amadi *et al.*, (1993). The crude fibre content in *Abelmoschus esculentus* was highest in the control than in plants from the treated soils. Generally, except for the total ash in the plant in 50% treated soil, the total ash of *Abelmoschus esculentus* from treated soils were higher than that of the control. The crude fibre content of *Zea mays* was highest in the control plant as compared to those of the treated plants and the reverse was the case for the total ash content. Total ash was however highest in *Z. mays* at the 100% concentration. The crude fibre content and total ash in *Arachis hypogea* were both highest in the control plant as compared to those of all the other plants in soils treated with the various concentrations of oilfield wastewater.

According to Udo and Fayemi (1975), the higher the fibre content, the hardier the plant that is, the plant can withstand more stress. The results of this investigation have shown that treatment with oilfield wastewater had an impact on the hardiness of the various plants and hence their inability to withstand stress as evidenced in the result of their physiological characteristics.

## Conclusion

The oilfield wastewater used in the study has been proven to have phytotoxic effect on the various crops used in this study. The results indicate that, the higher the concentration of the oilfield wastewater the greater the impact on

the physiology including the chlorophyll content and root density of the plant used in this study. Absorption of total hydrocarbon content by the plants which serve as sources of food for humans poses a serious health hazard. The discharge of oilfield wastewater into the terrestrial environment has been shown to have serious deleterious effect on soil and plants, leading to ecotoxicological and agro soil fertility problems that could create an artificial food scarcity due to damage to vegetation and soil organisms. Obviously, damage to agriculture results in associated health and economic costs (Oboh *et al*, 2009). The large volume of produced water handled in both on-shore and offshore petroleum production operation should be a major concern, especially with the possibility of further reduction in the oil content allowed in the discharged water (offshore operation), as well as the fact that produced water contains a number of undesirable toxic components. Handling this volume of water must be of prime concern to oil companies wherever they operate. The composition of produced water from one site to another is highly variable, containing different amounts of solids, heavy metals, and other inorganic compounds, as well as a wide spectrum of organic compounds. These materials pose operating and environmental problems when produced water is disposed off. It is therefore desirable (or required) to reduce the concentration of some of these contaminants before produced water disposal. Standards for produced water disposal are determined by the State, National and International regulatory bodies. The ultimate goal of produced water management is the protection of the environment in a manner commensurate with public health economic, social and political concerns.

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