

RESPONSE OF *Vigna unguiculata* L. (COWPEA) AND *Vigna subterranean* (BAMBARA GROUNDNUT) ON SOIL CONTAMINATED WITH SPENT LUBRICATING OIL

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ABSTRACT

Soil contamination by spent lubricating oil from automobiles emerge a growing concern in many countries especially in Nigeria posing an adverse effect on the agricultural land there by affecting the soil physical and chemical properties, increasing heavy metals present in it which can be dangerous to human health. Research on the response of *Vigna unguiculata* (cowpea) and *Vigna subterranean* (bambara groundnut) had been demonstrated to studied how such plants could react to contaminated soil. Viable seeds of *Vigna unguiculata* (cowpea) and *Vigna subterranean* (bambara groundnut) were cultivated on soil contaminated with spent lubricating oil. 4kg of soil each was used for the experiment and treated with 25ml, 50ml, 75ml and 100ml of spent

Introduction:

Contamination of soil by used lubricating oil is rapidly increasing due to global increase in the usage of petroleum products (Mandri and Lin, 2007). Environmental pollution has become a global problem affecting both developed and under developed countries (Suresh and Ravishankar, 2004) and it has assumed global concern as it pose a threat to the well being of all life forms including humans (Pezeshki *et al.*, 2000). In Nigeria, petroleum exploration, exploitation and distribution activities have led to the pollution of land and water ways resulting in less productive agricultural

lubricating oil and control experiment of unpolluted soil and polluted soil were also set up. Germination of plants was observed as the emergence of plumule from the soil surface. Plants height and the number of leaves were measured on the third week after germination occurred and continued every 3 weeks for 15 weeks. The germination results show that the control of *Vigna unguiculata* and *Vigna subterranean* had the highest height of plant and number of leaves followed by the soil treated with low concentration of spent oil. Analysis on the physicochemical properties of soil shows that the presence of spent lubricating oil had some significant effect on the soil. The soil pH is reduced from 6.2 of the unpolluted control soil to 5.4 of polluted soil making the soil more acidic, nitrogen is significantly reduced to 0.32% in the polluted control compared with the 0.60% in the unpolluted control of *Vigna unguiculata* likewise in *Vigna subterranean*, potassium reduces from 83.5ppm in unpolluted soil to 67.5ppm in polluted soil of *Vigna unguiculata* and 90 ppm in unpolluted soil of *Vigna subterranean* to 50ppm in polluted soil, phosphorus reduces from 69.95ppm in unpolluted control soil to 64.85 in polluted soil and also there is increase in the concentration of calcium, magnesium and heavy metals on the soil. However, *Vigna unguiculata* and *Vigna subterranean* were able to grow, germinate and survive well in oil polluted soil this was because they are well tolerant to the soil contaminant and could be considered effective for phytoremediation of pollutant especially at low concentrations.

Keywords: Plumule, phytoremediation, polluted, soil and contaminant

Lands in the affected areas, while the creeks and fishing waters have become toxic to the organisms inhabiting those areas (Dabbs, 1996). Petroleum and petroleum products enters the soil from ruptured crude oil pipelines, leakage from storage tanks, spillage during transport of petroleum products, abandoned manufactured gasoline sites, other unplanned release and current industrial processes (Mishra *et al.*, 2011). Engine oil usually contains chemical additives which include amines, phenols, phosphorus, sulphur and lead (Obidike, 1985). The additives

present in the engine oil have toxic effect to humans and animals when they come in contact with them (Obidike, 1985).

Spent lubricating oil is produced when new mineral-based crank case is subjected to high temperature, high mechanical strain (Agency for Toxic Substances and Disease Registry ATSDR, 1997). It is a mixture of different chemicals (Wang *et al.*, 2000) including petroleum hydrocarbons, chlorinated biphenyls chlorobenzo furans, lubricative additives, decomposition products and heavy metals that come from engine parts as they wear away (ATSDR, 1997).

The physical, chemical and microbiological properties of soil polluted with petroleum hydrocarbons undergo marked changes (Osuagwu and Iwuoha, 2015), the changes in soil due to contamination with petroleum derived substances can lead to water and oxygen deficit as well as shortage of available forms of nitrogen and phosphorus (Wyszokowska and Kucharski, 2000). Contamination of soil with spent lubricating oil lead to significant reduction of soil moisture content which subsequently induces the considerable retardation in plant growth (Anoliefo and Vwioko, 1995). This therefore calls for urgent and cost effective measures for remediation of the hydrocarbon contaminated lands and waterways in the Nigeria environment (Vegter *et al.*, 2002). Different treatment methods have been employed to reclaim contaminated soil.

Phytoremediation, a strategy that uses plant to degrade, stabilize, detoxify or remove soil contaminant (Gerhardt *et al.*, 2009) can be some alternative green technology methods for remediation of hydrocarbon contaminated soil. It offers an environmentally friendly, cost-effective and carbon neutral approach for the remediation of toxic pollutant in the environment (Dowling and Doty, 2009).

Phytoremediation has now emerged as a promising strategy for in-situ removal of many contaminants (Pullford and Watson, 2003). Microbe assisted phytoremediation including rhizoremediation; appears to be particularly effective for the removal and degradation of organic contaminants from contaminated soil (Gerhardt *et al.*, 2009). Thus this research is to determine the response of *Vigna unguiculata*(cowpea) and *Vigna subterranean* (Bambara groundnut) on soil contaminated with spent

lubricating oil. Soil contamination with oil is of great concern because the contaminated soils are not suitable for agricultural, industrial or recreational uses and are possible sources of surface and ground water contamination (Okoh, 2010). Various physical, chemical and thermal processes are already being used to remediate oil polluted site but the enormous costs, adverse effect on the environment and low efficiencies associated with these remediation techniques present limitations to their availability and usage. Thus, the need of biological techniques such as phytoremediation offers an environmentally friendly, cost effective, carbon neutral approach and safety of implementation for the cleanup of toxic pollutants in the environment, hence the choice of these two plant candidates for remediation of simulated oil polluted environment.

MATERIALS AND METHODS

Collection of Samples

The soil sample was collected from the Federal Polytechnic Bida in a polythene bags and transported to the garden. Spent lubricating oil was obtained from a mechanic workshop in Bida, Niger state. Viable seeds of cowpea (*Vigna unguiculata* L) and bambara groundnut (*Vigna subterranean* L) were purchased from old market in Bida, Niger state.

Soil Sampling and Preparation

The sand-loam soil obtained from the Federal Polytechnic Bida was sieved with a mesh of 2mm. This is to separate the non-degradable materials out of the soil in order to keep the soil free of unwanted materials which could disturb the proper functioning of soil microorganisms, proper stretch of the roots and proper organization of the soil.

About 4kg of the sieved soil was weighed into different polythene bags. The bags were perforated at the sides and bottom to avoid water logging and also increase soil aeration. The soil was mixed with different volumes of spent lubricating oil graded from 25ml, 50ml, 75ml and 100ml. The soil was left for a period of 4days without planting. This was done for uniformity of oil, air content, temperature and effective activities of soil microorganisms (Sarkar *et al.*, 2005).

The polythene bags were arranged in a Completely Randomized Design consisting four treatments replicated three times for the two different seeds. Control consisting of a bag of plant without spent lubricating oil and a bag of spent lubricating oil without plant were also set up. The cowpea seeds and bambara groundnut seeds were subjected to viability test using flotation technique. 2 seeds of cowpea and bambara groundnut were planted in each different labeled polythene bag and was consistently watered every day.

Plant height was determined using a meter ruler from the soil level to the tip of the youngest leaf and the number of leaves was counted every 3weeks for the period of 15weeks.

Soil Sample Preparation for Analysis

Soil from each of the bag was taken at a depth of 0-3cm in a randomized manner. The soil samples were taken with sample plate and were given proper labeling. They are spread out to air-dry on a cardboard sheet. Gravels and other metallic substances were removed and large soil aggregate were also broken down and stored in a sachet polythene bag for the laboratory analysis. The soil samples were analyzed for physiochemical parameters.

Physico-chemical analysis

The following physico-chemical analyses were carried out pH, Electrical Conductivity (E.C), Cation Exchanged capacity (CEC) of Ca^{2+} , Mg^{2+} and K^{+} were determined (Science First, 2009), Total Nitrogen was determined using Micro Kjeldahl method. Phosphorus was determined using absorption spectrophotometer. Organic carbon and Organic Matter was determined by Walkley Black chromic acid wet oxidation method.

Heavy Metals analyses

Heavy Metals such as Zn, Iron, Lead and Mn were determined using Atomic Absorption Spectrophotometer (FS240 AAA gilent Technology, USA: Prior to the experiment, the soil samples were also analyzed for the presence of the heavy metals of interest: lead, iron, zinc and manganese.

RESULTS AND DISCUSSION

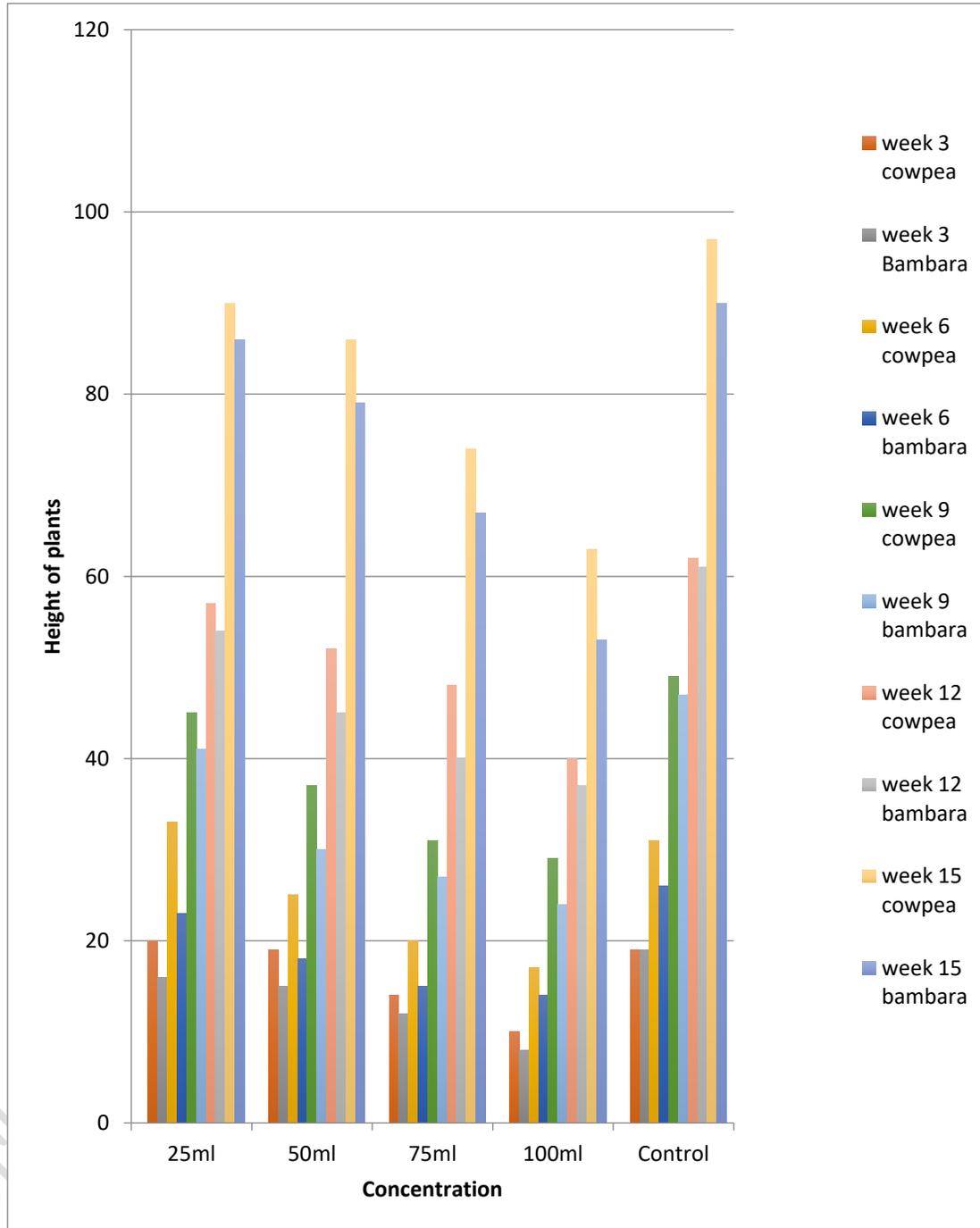


Fig. 1. Graded Concentrations of Spent Lubricating Oil in Relation to Duration on the height of *Vigna unguiculata*(cowpea)and *Vigna subterranean*(bambara groundnut)

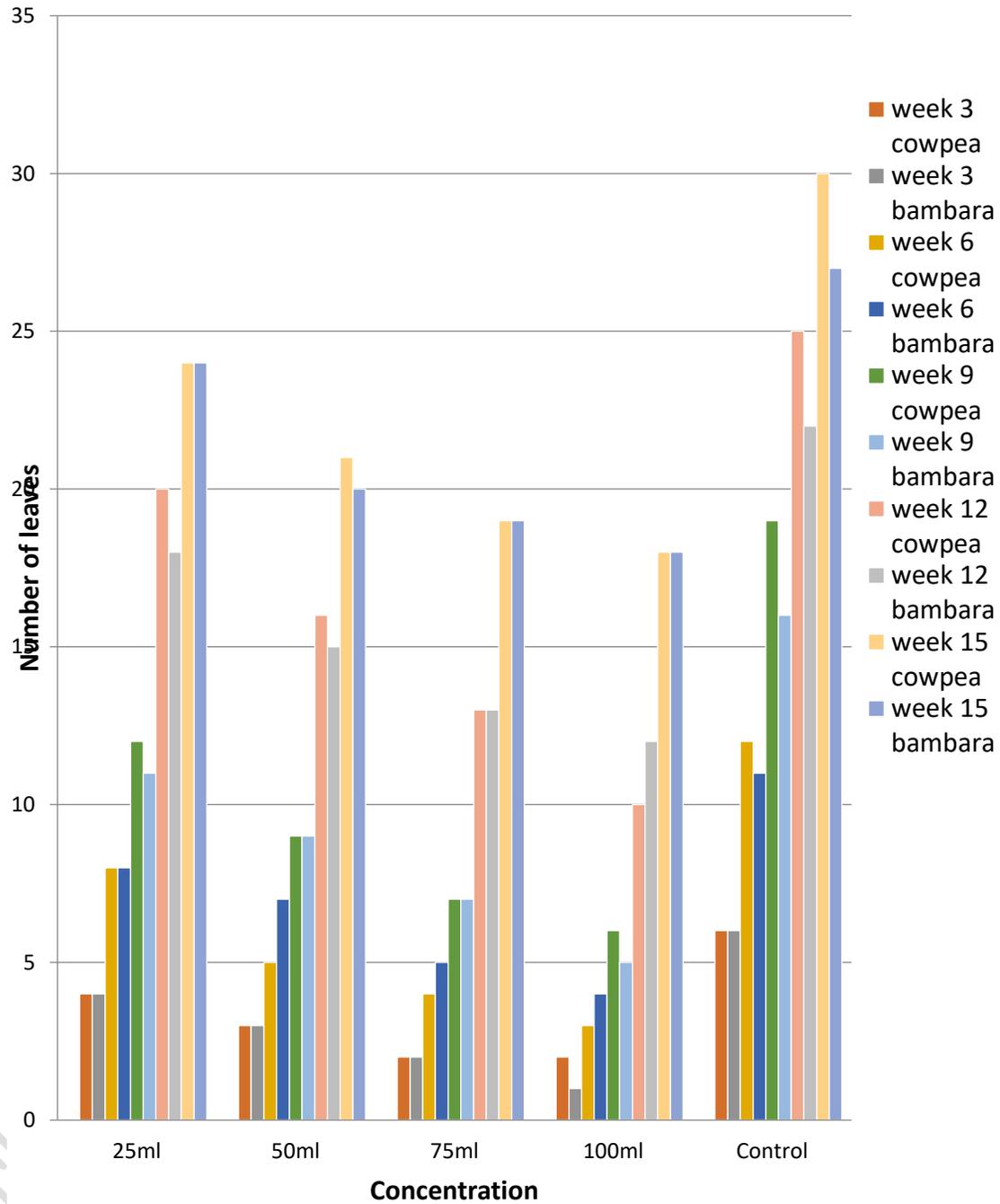


Fig.2. Graded Concentrations of Spent Lubricating Oil in Relation to Duration on the number of leaves of *Vigna unguiculata*(cowpea) and *Vigna subterranean*(bambara groundnut)

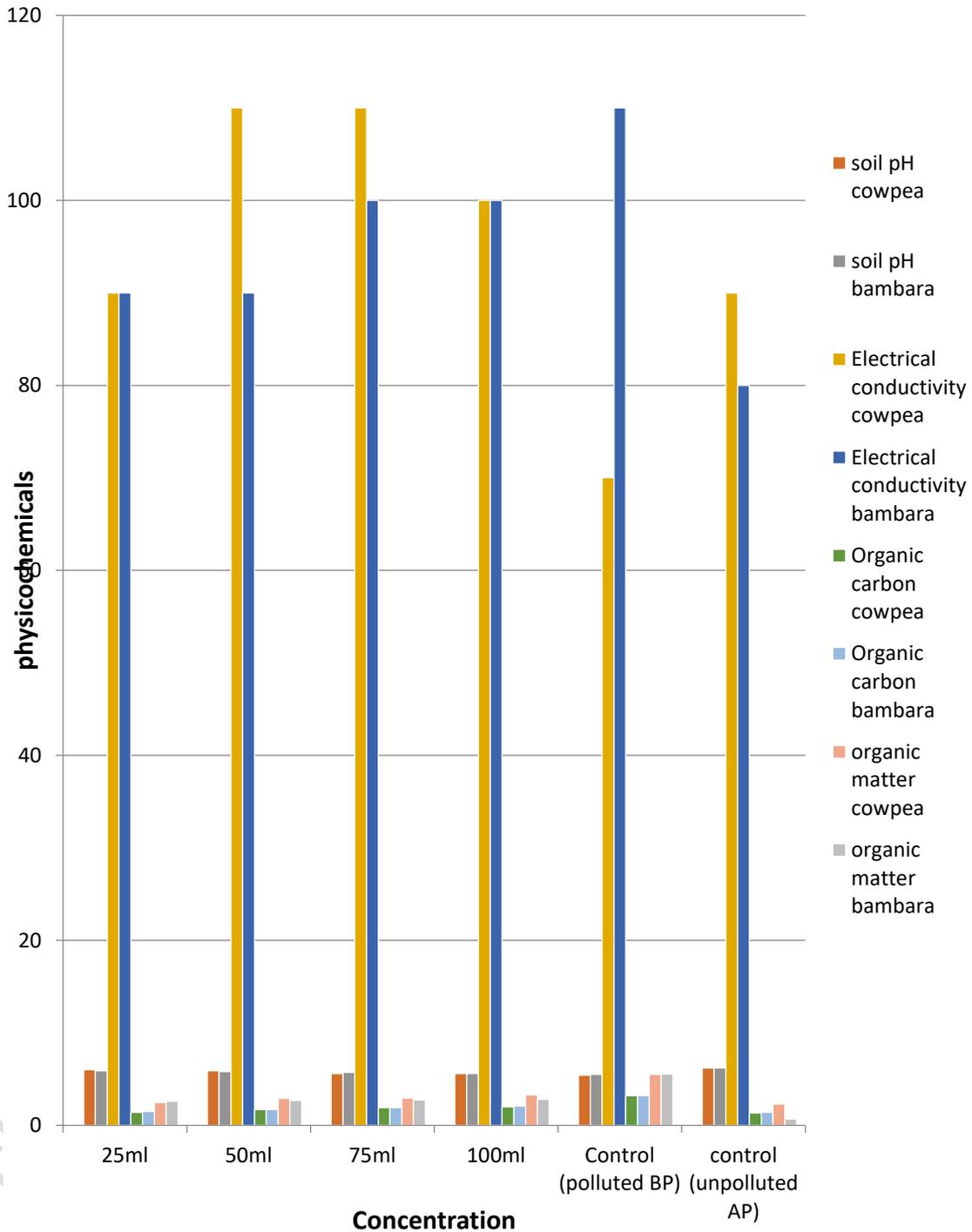


Fig. 3. Effect of Spent Lubricating Oil on the soil physiochemical properties of *Vigna unguiculata* (Cowpea) and *Vigna subterranean* (Bambara groundnut)

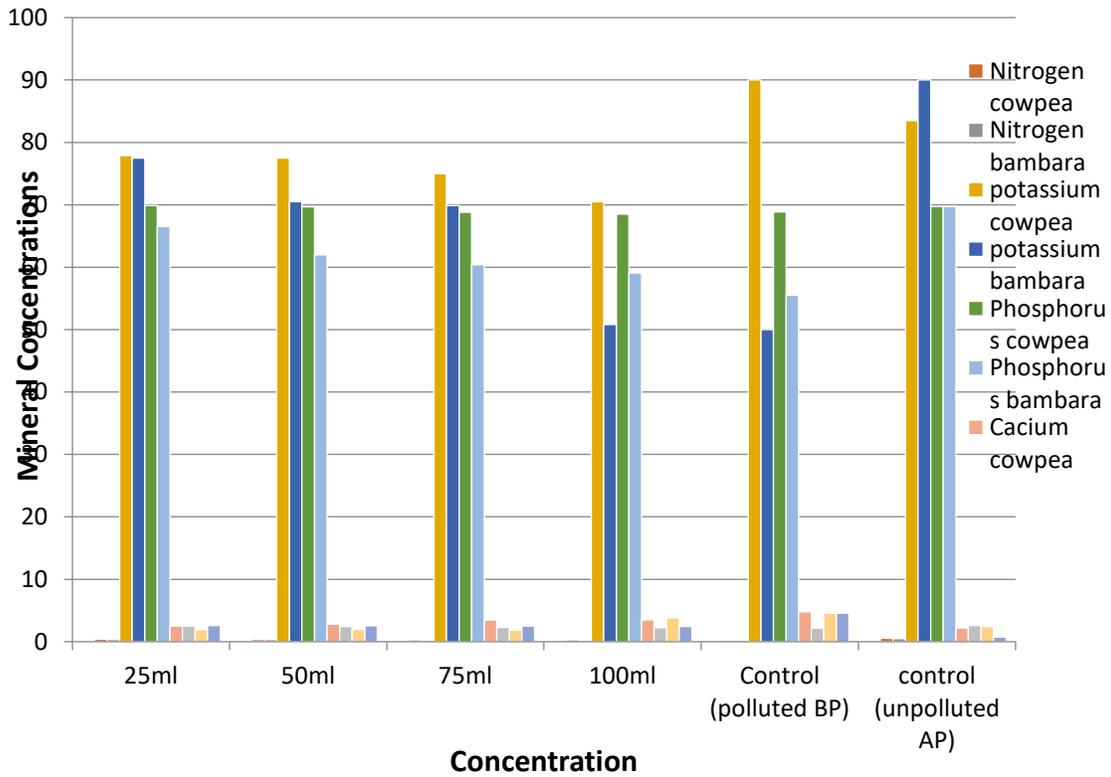


Fig. 4. Effect of Spent Lubricating Oil on the soil physiochemical properties of *Vigna unguiculata* (Cowpea) and *Vigna subterranean* (Bambara groundnut)

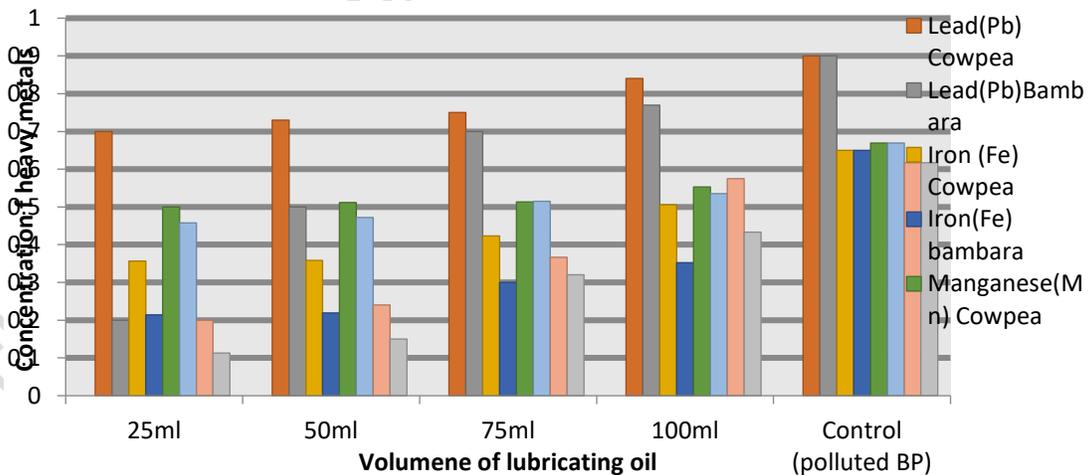


Fig. 5. Degradation of heavy metals in relation to the concentration of spent lubricating oil on *Vigna unguiculata* (Cowpea) and *Vigna subterranean* (Bambara groundnut)

DISCUSSION OF RESULTS

All the test plants used grew successfully in the oil polluted soil, the appearance of the *Vigna unguiculata* (cowpea) and *Vigna subterranean* (bambara groundnut) plants in response to different concentrations of oil was monitored throughout the period of 105 days of the experiment; no plant death was recorded in the treatment of the soil, however some of the plants showed signs of phytotoxicity such as yellowing of leaves, stunted growth compared with control. The signs are in line with the findings of (Vouillamoz and Mike, 2009). Plants in soil contaminated with 75ml and 100ml spent lubricating oil showed high symptoms of phytotoxicity.

During the period of this research work, it has been shown that the presence of spent lubricating oil had a significant and obvious effect on the growth of the plants in terms of height (fig. 2.0). It was discovered that the shortest height of plant occurred with increased amount of lubricating oil (50, 75, 100 ml). This was also observed by Adu *et al.* (2015). The total height of the control of *Vigna unguiculata* and *Vigna subterranean* were significantly greater than that of plants grown in soil polluted with 50, 75, and 100 ml of spent lubricating oil in fig, 2.0. However, there was no much difference in height of control experiment and soil mixed with 25ml of spent lubricating oil. This was in line with (Olayinka and Arinde, 2012) and (Njoku *et al.*, 2008) works.

The reduction in height of the plants could be due to unfavorable soil conditions mainly due to insufficient aeration following a decrease in the air filled pore spaces (Atuanya, 1987), effects on soil microbes (Benka and Ekundayo, 1995), reduced biochemical activities as well as presence of heavy metals (Agbogidi and Egbuchua, 2010) and a disruption in the soil water-plant interrelationship (Agbogidi, 2011). The reduction in height of *Vigna unguiculata* and *Vigna subterranean* due to high level of spent oil could also be attributed to deficiency in availability of nutrients needed to maintain physiological processes involved in plant growth.

These reductions of growth due to high level of pollution are in agreement with findings of Molina *et al.*, (2005) who recorded similar results and inferred that the negative effect could be due to impermeability effect of

petroleum hydrocarbons, or immobilization of nutrients mainly nitrogen or inhibitory effect of some polycyclic aromatic compounds.

There was an increase in height with the plants having lower concentration as time increases from the third weeks, sixth weeks to fifteen weeks which shows that the plant was not so affected with the little amount of spent oil. This was in line with Anoliefo and Vwioko (1995) who reported that low level of soil pollution could be easily degraded by natural rehabilitation of soil. This implies that at low levels of spent lubricating oil, the test crops were able to assessed nutrients due to high microbial activities in such soil which in turn made essential nutrients readily available for canopy development. There was also increase in the plants height with the highest amount of spent oil, but the increase in height is not as healthy as the height of the plant with the lowest concentration of spent lubricating oil.

It was also observed that cowpea (*Vigna unguiculata*) was able to germinate and grow faster compared to bambara groundnut (*Vigna subterranean*) in soil contaminated with spent lubricating oil. The highest height of the *Vigna unguiculata* was observed on the fifteenth week having 25ml of spent lubricating oil with a value of 34.0cm while the lowest height was observed in the third week having 100ml of spent lubricating oil with a value of 3.5cm. The highest height of *Vigna subterranean* was observed on the fifteenth weeks having 25ml of spent lubricating oil with a value of 20.5cm while the lowest height was observed on the third week having 100ml of spent lubricating oil with the value of 3.0cm.

The range of value recorded for the number of leaves experienced fluctuations in relation to concentration and duration of spent lubricating oil (fig. 3.0). This may be attributed to the increased organic matter present in the sand-loam soil which might have induced increased production of leaves in the cowpea and bambara plant (Adewole and Moyinoluwa, 2012). This was in line with the findings of Fernandes and Henriques, (1991) who discovered that some heavy metals at low concentrations are essential micronutrient for plants, but at high concentrations, they may cause metabolic disorders and growth inhibition for most of the plant species.

Therefore it is Important to mention here that plant height and the number of leaves as a plant growth parameter and yield is vital for plants. This is

because the taller a plant is and number of leaves present, the higher the amount of light energy absorbed by such plant and invariably, the higher the amount of light, the higher the rate of photosynthesis and consequently the amount produced by the plants

The results on the physiochemical parameters showed that spent lubricating oil had some significant adverse effect on the chemical properties of the soil. As shown in fig. 4.0, the chemical properties of soil were altered by pollution of the spent lubricating oil. The results of the soil analysis showed that spent lubricating oil reduced the pH value to 5.6 of soil treated with 100ml spent lubricating oil compared with 6.2 of unpolluted control soil thereby making the soil more acidic. The reduction in pH value which resulted in increase in the acid level of the soil was probably due to the addition of carbon present in the spent lubricating oil to the carbon already present in the soil.

Electrical conductivity value recorded for all the soil samples increased to 110mS/m for *Vigna unguiculata* (cowpea) and *Vigna subterranean* (bambara groundnut) in the soil treated with high spent lubricating oil compared to 90 mS/m in the unpolluted control (fig 4.0). This showed that the presence of spent lubricating oil in the soil increase the amount of soluble salts in the soil.

The percentage organic carbon and organic matter were greatly influenced by the presence of spent lubricating oil. In table 4, the percentage organic carbon of unpolluted control was 1.34% and that of the polluted control was 3.19 this was greatly increase due to the influence of spent lubricating oil on it. However the other treatments 25ml, 50ml 75ml and 100ml experience decrease in the percentage organic carbon compared with polluted control this may be due to phytodegradation of the spent petroleum product and its utilization in the soil giving rise to the observed decrease in the organic carbon of the polluted soil. The percentage organic matter was increased in the polluted control (5.50) and reduced in unpolluted control (2.31), percentage organic matter increase in the treatment as the concentration of spent lubricating oil increased.

Fig. 5.0 shows the effect of spent lubricating oil on the concentration of soil nutrients (N, K, P, Ca and Mg). There was a drastic reduction of Nitrogen

in the soil treated with different concentration of spent lubricating oil. This was in line with Kayode *et al.* (2009) work who observed reduced nitrogen in soil treated with spent lubricating oil. Nitrogen is significantly reduced to 0.29% in the treatment of soil containing 100ml of spent lubricating oil as compared with 0.60% in unpolluted control of *Vigna unguiculata*, also from 0.50% of nitrogen in the unpolluted control of *Vigna subterranean* is reduced to 0.20% in the soil treated with 100ml of spent lubricating oil. The reduction of nitrogen is due to the effect of spent oil on the soil. However, *Vigna unguiculata* had increased percentage nitrogen compared to *Vigna subterranean* this is due to the ability of the plant to fix nitrogen to the soil by symbiotic association with Rhizobium (a bacterium).

Potassium had higher values in the treatments that had lower concentration of spent lubricating oil than those that had higher concentration of spent lubricating oil (fig.5.0). Soil treated with 100ml spent lubricating oil of *Vigna unguiculata* had reduced available potassium of 70.5ppm compared with unpolluted control having the value of 83.5ppm, while for *Vigna subterranean* the available phosphorus reduced from 70ppm of the unpolluted control to 59.10ppm of soil treated with the highest spent lubricating oil. The presence of spent lubricating oil on the soil reduced the potassium available for the soil.

Phosphorus content in the soil contaminated with spent lubricating oil had been significantly reduced from 69.95 ppm of the unpolluted control to 68.50ppm of the treatment containing 100ml spent lubricating oil of *Vigna unguiculata* and also from 70 ppm of unpolluted control to 59.10 ppm on the treatment of soil containing 100ml spent lubricating oil of *Vigna subterranean*. The significant decrease of available phosphorus could be attributed to deleterious effect of spent engine oil application on soil. Spent lubricating oil in soil could inhibit microbial transformation of organic matter thus, leading to low mineralization of phosphorus. Furthermore, acidic nature of the soil could have affected phosphorus availability in the soil since it can be fixed by aluminum at low pH. Ogboghodo *et al.* (2004) also noted that available Phosphorus was low in soil treated with spent lubricant oil.

Percentage of calcium and magnesium greatly increase with increase concentration of spent lubricating oil. This shows that the presence of

spent lubricating oil increase the calcium and magnesium content of the soil treated with *Vigna unguiculata* and *Vigna subterranean*

All the chemical properties of soil studied indicated severity as the levels of spent lubricating oil application increased indicating that high disposal of hydrocarbon oil into the soil poses a great danger of soil degradation and low productivity

Fig. 6.0 shows the concentration of heavy metals (Pb, Fe, Mn and Zn) on the soil treated with spent lubricating oil at different concentrations. In this result, lead with the value of 0.90ppm had the highest level of contamination in the control soil followed by Manganese 0.69 ppm, iron 0.650ppm and zinc 0.617ppm in the soil polluted with spent lubricating oil of *Vigna unguiculata* and *Vigna subterranean*. Heavy metals when present in high amount in the soil were found to be toxic and pose a significant effect on the soil physical and chemical properties and also a threat to life of people who come into contact with it. However *Vigna unguiculata* (cowpea) and *Vigna subterranean* (bambara groundnut) had the potential to remediate the level of lead, iron, manganese and zinc that are present in the soil to a lower concentrations (fig. 6.0). This is due to the phytoextraction ability of *Vigna unguiculata* and *Vigna subterranean* on the heavy metals present in the soil.

CONCLUSION AND RECOMMENDATIONS

The study has demonstrated that spent lubricating oil has a significant effect on reducing the germination characteristics and soil nutrients of *Vigna unguiculata* (Cowpea), and *Vigna subterranean* (Bambara groundnut). Comparatively, cowpea and Bambara groundnut show some level of tolerance and respond positively to spent lubricating oil. The different sensitivity of plants to spent lubricating oil toxicity can be exploited in phytoremediation practice by choosing specie that are well tolerant to the contaminant. *Vigna unguiculata*(Cowpea) and *Vigna subterranean* (Bambara groundnut) from the study could be considered for phytoremediation of spent lubricating oil pollution. Therefore they can be used in phytoremediation especially at low concentration and may also grow for food in oil polluted soil

RECOMMENDATIONS

Based on the adverse effect of oil pollution on the agricultural land, it is hereby recommend that

- Government agencies and multinational companies should maintain a consistent effort in reducing petroleum product spill in agricultural lands
- The need to encourage the protection of farmlands and their surroundings against indiscriminate disposal of the oil cannot be overemphasized
- Government should encourage phytoremediation being a low cost and safe biological method of purifying oil polluted soil for agricultural use
- *Vigna unguiculata* and *Vigna subterranean* plant should be considered useful for phytoremediation of polluted land especially at low concentration.
- More research work should be carried out on other plants of possible phytoremediation activity

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