Science World Journal Vol. 18(No 1) 2023
www.scienceworldjournal.org
ISSN: 1597-6343 (Online), ISSN: 2756-391X (Print)
Published by Faculty of Science, Kaduna State University

Full Length Research Article

EFFECTS OF SOIL POLLUTION ON THE GERMINATION, GROWTH, FRUITING AND LEAF ANATOMY OF *ABELMOSCHUS CAILLEI* (A CHEV.) STEVELS MALVACEAE

A. Sagaya[1*], A.A. Abdulrahaman[1], P.O. Oluwanisola[1] and S.B. Tsoho[2]

[1]Applied Plant Anatomy and Wood Technology Laboratory, Department of Plant Biology, Faculty of Life Sciences, University of Ilorin, Ilorin, Nigeria
[2]Department of Biology, Nigerian Army University, Biu, Borno State

*Corresponding Author Email Address: sagaya.aa@unilorin.edu.ng +2348063052975

ABSTRACT
Spent engine oil is one of the most common pollutants in the world. Its Disposal has been persistently problematic and adversely affects plant growth. Effects of different levels (0, 50, 100, 150 and 200 ml respectively in 5 kg of soil) of spent engine oil application on germination, survival, growth and leaf anatomy of *Abelmoschus caillei* (A Chev.) Stevels was investigated using completely randomized design in five replicates. Data collected on germination, seedling survival, growth and fruiting were subjected to the Statistical Package for Social Sciences (SPSS) analyses version 21.0; while leaf epidermal features were determined using standard methods. Spent engine oil at 200 ml significantly ($P<0.05$) delayed and reduced seed germination from 4 days to 2 weeks for all plants and by 85.71%; plant height was reduced by 65.74%; number of leaves by 84.74%; and leaf area by 84.41% compared to the control. Number of stomata on the abaxial and adaxial surfaces was increased by 27.99% and 35.02%, respectively; stomata area on the abaxial and adaxial surfaces was reduced by 97.35%, and 95.59 ($p< 0.05$) respectively. There was no fruiting. Application of high concentration of spent oil in soil affected all the parameters considered in this study. Therefore, soil contamination with spent engine oil should be properly checked in order to ensure sustainable plant productivity.

Keywords: *Abelmoschus caillei,* physiology, anatomy, spent engine oil

INTRODUCTION

Soil is the most valuable and non-renewable component of farming which requires proper maintenance for environmental sustainability. Sustainable use of this natural resource on which agriculture depends is absolutely necessary for agricultural productivity. Soil pollution by crude oil and petroleum products such as fuel oils, spent engine and diesel fuels are presently a menace in Nigeria, particularly in urban areas (Adenipekun *et al.,* 2009, Onwuka *et al*., 2012).

Spent engine oil is a waste lubricating oil collected from automobile workshops, garages and industrial sources like hydraulics oil, turbine oil, process oil and metal working fluids (Osuagwu *et al.,* 2017).

Spent engine oil affects the physical, chemical and biological properties of the soil and hence, it is affecting the growth, development, productivity and yield of plants (Swapna *et al.,* 2021). Continuous increase in the number of cars and automobile users in Nigeria and the ways of disposal of this oil are of major concern to the scientists, agriculturists, environmentalists, and other stake holders. In the recent time, many scientists (Ahamefule *et al.,* 2017; Ikhajiagbe *et al.,* 2017; Oluwanisola and AbdulRahaman 2018) have reported on the implications of the uncontrolled, improper and indiscriminate disposal of spent engine oils on the soil.

Many authors have investigated and reported the effects of petroleum oil and spent engine oil on soil and plant. Ahamefule *et al.* (2015) noted that petroleum oil in soil have deleterious effects on biological, chemical and physical properties of the soil depending on the dose, type of the oil and other factors. Spent auto-engine oil is known to contain increased amounts of heavy metals compared to the unused oil. Oluwanisola and AbdulRahaman (2018) observed that soil contamination with spent engine oil can alter the physico-chemical properties of the soil and degrade its capacity to provide suitable habitat for plants' growth in such soil. *A. esculentus* seed germination was delayed or inhibited due to the presence of spent oil in the soil. Growth, chlorophyll content in leaves, Potassium and sodium content, reproduction and leaf anatomical configuration, were also affected. Ahamefule *et al.* (2017) reported that spent auto engine oil above 1 % v/w soil contamination significantly deteriorated water movement into and within the experimental soil and consequently the growth and yield of soybean was adversely affected.

Abelmoschus caillei (A Chev.) Stevels, commonly referred to as West African okra, originated as an allopolyploid hybrid of *A. esculentus* (L.) Moench and *A. manihot* (L.) Medik. It is cultivated for fresh pods, leaves, stem and seeds. Due to its high yield and hardiness, it has become a major source of okra pods in Nigeria and its cultivation is progressively replacing the conventional type (Kehinde, 1999; Adeniji and Aremu, 2007). Information on the effects of pollutants on plant anatomy is limited. This study, therefore, attempts to establishing the leaf architectural diversity of *A. caillei* under the influence of oil contaminated soil, as a pointer to environmental degradation. The objectives were to assess the effect of spent engine oil polluted soil on the germination, growth and fruiting, to determine the effect of spent oil on the anatomical adaptations of the plant species through their leaf epidermal diversities and to determine whether there are variations in the treatment of the species to the pollutants.

MATERIALS AND METHODS

Study Area and collection of materials

The study was carried out at the Botanical Garden of the University of Ilorin, Ilorin, Nigeria with coordinate of 8° 29'N, 4° 35'E. The top soil sample was collected from the Botanical Garden. Seeds of *A.*

caillei were obtained from the Ministry of Agriculture, Ilorin, Kwara State, while spent engine oil was obtained from Lubcon Oil Ltd, Ilorin.

Experimental design
The experimental design used was Randomized complete Block Design (RCBD) with five replicates of each treatment. The top soil used was thoroughly mixed with the appropriate spent oil level before the poly pots were filled up with 5kg weight of the contaminated soil. The levels of spent engine oil that were used are 0 (control), 50,100, 150 and 200 ml following the method of Oluwanisola and Abdulrahaman (2018).
Seed viability was determined by the floatation method before ten viable seeds were planted into the contaminated and uncontaminated soils in the perforated poly pots and were watered immediately and afterward every day to field capacity. The experimental setup was monitored for 7 weeks after planting, germination and survival patterns were measured from the third day after planting and collated weekly until week three when the seedlings were thinned to two per pot. The growth parameters such as plant height, number of leaves and leaf area were measured fortnightly with effect from the third week after planting. Plant height (cm) was measured with a meter rule from the top soil level to the terminal bud from four weeks after planting (WAP). Numbers of leaves were determined by visually counting the number of leaves per plant per pot per treatment. Leaf area determination was done by measuring the length and breadth of the leaf and correction factor of 0.75 was used to multiply the length and breadth measurements. Yield parameters: number of fruits, fruit length, fresh and dry weights of fruits were determined at weeks 7 and 8 using the method of Ikhajiagbe *et al.* (2017).

Anatomical study
A leaf segment of an area of 1cm square from each specimen was cut and immersed in a concentrated solution of nitric acid in petri dishes for maceration. The samples were then rinsed in several changes of water to rinse off the acid. The upper (adaxial) and lower (abaxial) membranes were separated from the mesophyll using fine forceps and dissecting needles, the membranes were then rinsed in water. The specimens were stained in 1% safranin solution for about five minutes, excess stain was wiped with tissue paper. They were then mounted in glycerine on a microscope glass slide in order to prevent dryness in the course of temporary preservation. Observations of the leaf cuticles were made using a binocular light microscope. The stomatal complex types present were identified following Dilcher (1974) using the field of view at x40. The number of subsidiary cells per stoma was counted. The number of stomata and epidermal cells per millimetre was also determined.

All the qualitative attributes such as types of trichomes and stomatal complexes, frequency of trichomes and stomatal complex types, density of trichomes and stomata, indices of trichomes, stomata and their sizes were determined following methods of Franco (1939), Stace (1965) and Obiremi and Oladele (2001). The terminologies used for description of trichomes and stomatal complexes are according to Dilcher (1974) and Metcalfe and Chalk (1988). Hand drawings of observations were taken. Data generated were analyzed using the Statistical Package for Social Sciences (SPSS) version 21. After testing the homogeneity of variance and normality of data, one way ANOVA at P <0.05 was performed following the description of Steel and Torrie (1981) and significant differences among treatments were separated using Duncan's multiple range tests (DMRT) at P<0.05.

RESULTS
The results for the germination count of the control and treatments are presented in Table 1. The germination count of the control pots was significantly higher (ANOVA, P<0.05) than other treatments from week 1 to week 3. In addition, there were no germinations in the 200 mL pots at week 1, and it recorded the lowest germination counts consecutively till 3 weeks. The mean values at week 2 and 3 showed that there was no significant difference in the number of surviving seedlings among the control, 50 ml and 100 ml plants at *P*< 0.05.

Table 1. Germination count and number of surviving seedlings as affected by different concentrations of spent engine oil

Treatment	1 WAP	2 WAP	3 WAP
Control	1.80 ± 1.64^{a}	5.20 ± 1.10^{a}	4.20 ± 0.84^{a}
50 mL	0.40 ± 0.89^{ab}	3.60 ± 2.70^{ab}	3.00 ± 2.55^{abc}
100 mL	0.80 ± 1.30^{ab}	4.20 ± 1.34^{a}	3.20 ± 2.05^{ab}
150 mL	0.20 ± 0.45^{b}	1.60 ± 2.51^{bc}	1.00 ± 1.73^{bc}
200 mL	0.00 ± 0^{b}	0.40 ± 0.89^{c}	0.60 ± 0.89^{c}

Values with the same letters within column are not significantly different from each other.

The height of the control and 50 ml plants was not significantly different (ANOVA, P<0.05) from week 3 to 7. The lowest recorded heights were in 200 ml throughout, although they were not significantly different from plants in the 150 ml pots at P<0.05. The 100ml plants were not significantly different from any of the other treatments at week 3 (Table 2).

Table 2. Plant heights as affected by different concentrations of spent engine oil

Treatment	3 WAP (cm)	5 WAP (cm)	7 WAP (cm)
Control	10.20 ± 2.28^{a}	15.80 ± 2.59^{a}	21.60 ± 4.10^{a}
50 mL	10.00 ± 1.22^{a}	15.60 ± 1.52^{a}	22.00 ± 2.74^{a}
100 mL	6.80 ± 3.83^{ab}	11.80 ± 6.83^{b}	16.20 ± 9.42^{ab}
150 mL	3.60 ± 4.93^{b}	6.20 ± 8.50^{b}	7.60 ± 10.50^{b}
200 mL	3.20 ± 4.44^{b}	5.60 ± 7.67^{b}	7.40 ± 10.14^{b}

Values with the same letters within column are not significantly different from each other.

The leaf count for the control and 50 ml plants are not significantly different from each other; but were however, significantly higher than 150 ml and 200 ml pots, and the 100 ml plants only differ significantly from the control at week 5 at *P*< 0.05. The 150 ml and 200 ml plants were statistically the same throughout; however, the 200 ml plants had the lowest recorded leaves numbers. The control plants have the highest recorded number of leaves (Table 3).

Table 3. Number of leaves of plants as affected by different

concentrations of spent engine oil

Treatment	3 WAP	5 WAP	7 WAP
Control	5.20±1.10[a]	10.80±1.92[a]	11.80±1.48[a]
50 mL	4.20±0.84[a]	9.60±1.52[ab]	10.60±1.14[a]
100 mL	3.20±2.17[ab]	6.00±3.67[bc]	7.80±4.60[a]
150 mL	1.40±1.95[b]	2.20±3.03[c]	2.80±3.90[b]
200 mL	1.20±1.79[b]	2.20±3.19[c]	1.80±2.68[b]

Values with the same letters within column are not significantly different from each other.

The leaf areas were also observed to decrease progressively as the concentration of pollutant increases from the control to the 200 ml plants, the control plants having the highest leaf area and the 200 ml plants the lowest. The leaf area of the control was significantly different from all the treatment plants at week, The 50 ml plants had no significant difference from, both control and 100 ml plants at week 5, and control plants at week 7 at $P < 0.05$. The leaf area parameters of the control and all treatment concentrations were observed to increase sequentially from week 3 to week 7 (Table 4).

Table 4. Leaf area of plants as affected by different concentrations of spent engine oil

Treatment	3 WAP (cm^2)	5 WAP (cm^2)	7 WAP (cm^2)
Control	8.13±0.61[a]	24.73±1.83[a]	70.86±7.12[a]
50 mL	4.91±0.58[b]	21.08±0.92[ab]	55.40±3.69[a]
100 mL	2.94±1.70[c]	14.34±8.10[b]	34.93±19.94[b]
150 mL	1.06±1.47[d]	5.47±7.50[c]	15.01±20.56[c]
200 mL	0.68±0.93[d]	4.33±5.93[c]	11.05±15.17[c]

Values with the same letters within column are not significantly different from each other.

There was no significant different between the control, 50 ml and 100 ml in the fruit length, fresh weight and dry weight, the fruit number of the control is statistically different from all the treatment pot at $P < 0.05$. In addition, only the 150 ml plants differ from them. The number of fruits decreased progressively as the concentration of the pollutant increased from the control to 200 ml plants, with the control and 200 ml having the statistical highest and no fruit respectively (Table 5).

Table 5. Yield and fruit parameters as affected by different concentrations of spent engine oil

Treatment	Fruit Length (cm)	Fruit Number	Fruit FW (g)	Fruit DW (g)
Control	6.62±3.76[a]	17.80±2.17[a]	5.84±0.99[a]	0.53±0.28[a]
50 mL	4.04±0.65[a]	10.80±2.39[b]	6.20±1.27[a]	0.44±0.12[a]
100 mL	5.30±3.81[a]	5.00±3.16[c]	6.78±3.79[a]	0.55±0.42[a]
150 mL	0.36±0.80[b]	0.20±0.45[d]	0.48±1.07[b]	0.03±0.06[b]
200 mL	0.00±0.00[b]	0.00±0.00[d]	0.00±0.00[b]	0.00±0.00[b]

Values with the same letters within column are not significantly different from each other.

The stomatal complex type observed is paracytic (i.e., stomata that possess one or more pairs of subsidiary cells oriented parallel to the guard cells) (Figure 1). Although variations exist in the epidermal cell shape and anticlinal wall, the basic stomatal appearance was retained and the leaves were observed to be amphistomatic (i.e. having stomata only on the adaxial surface).

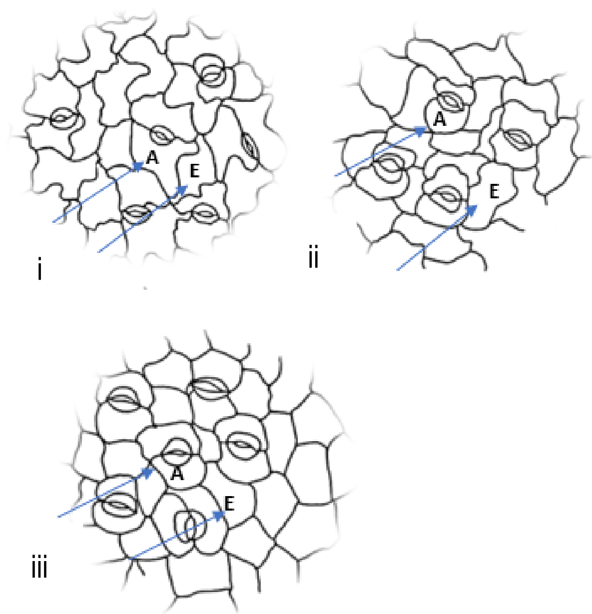

Figure 1. The epidermal features, i: paracytic stomata with irregularly shaped accessory and epidermal cells, ii: paracytic stomata and somewhat elongate epidermal and accessory cells with curved walls, iii: paracytic stomata and accessory and epidermal cells with somewhat straight walls.

The number of stomata and epidermal cells tend to increase with increase in pollutant concentration (47.70 – 64.00 for adaxial and 58.60 – 75.00 for abaxial) while the stomatal index tend to reduce (27.21 -21.75 for adaxial and 27.41 - 23.33 for abaxial) gradually as spent oil concentration increases with variations between abaxial and adaxial surfaces. The stomata number on the abaxial leaf surfaces were always significantly higher than those of the adaxial surface for all treatments at $P < 0.05$ (Table 6).

Table 6: Influence of different concentrations of spent engine oil on stomata and epidermal features

Treatment	Leaf surface	Stomatal number (mm^2)	Epidermal cell number (mm^2)	Stomatal index (%)
Control	Adaxial	47.40±4.16[e]	126.80±6.65[f]	27.21
	Abaxial	58.60±2.70[c]	155.20±14.89[e]	27.41
50 mL	Adaxial	50.00±5.70[e]	135.60±9.07[f]	26.94
	Abaxial	63.60±3.05[b]	175.20±6.14[d]	26.63
100 mL	Adaxial	53.40±2.61[d]	182.80±7.46[d]	22.61
	Abaxial	62.00±2.92[bc]	205.60±7.73[c]	23.17
150 mL	Adaxial	64.60±2.07[b]	252.40±10.14[a]	20.38
	Abaxial	75.60±2.51[a]	259.40±8.38[a]	22.57
200 Ml	Adaxial	64.00±3.39[b]	234.20±15.35[b]	21.75
	Abaxial	75.00±4.64[a]	246.40±22.38[ab]	23.33

Values with the same letters within column are not significantly different from each other.

The stomatal count of the control plants was significantly lower than the 100 ml, 150 ml and 200 ml treatment plants and the 200 ml plants were consistently significantly higher than the control and 50 ml plants. The highest stomatal area observed was 563.11 µm and the lowest was 14.13 µm (Table 7).

Science World Journal Vol. 18(No 1) 2023
www.scienceworldjournal.org
ISSN: 1597-6343 (Online), ISSN: 2756-391X (Print)
Published by Faculty of Science, Kaduna State University

Table 7. Stomatal size as influenced by different concentrations of spent engine oil contamination

Treatment	Leaf surface	Stomatal length (µm)	Stomatal breadth (µm)	Stomatal area (µm²)
Control	Adaxial	26.40	19.70	408.39
	Abaxial	31.30	21.70	533.34
50 mL	Adaxial	24.60	16.50	318.73
	Abaxial	33.20	21.60	563.11
100 mL	Adaxial	23.90	16.50	309.96
	Abaxial	25.70	16.40	330.96
150 mL	Adaxial	9.80	8.10	62.33
	Abaxial	11.70	8.60	79.01
200 mL	Adaxial	4.40	5.20	17.97
	Abaxial	5.00	3.60	14.13

The trichome types observed were simple unicellular, branched unicellular and multicellular trichomes (Figure 2). Walls were observed to be thick and smooth; the trichomes were pointed at the distal end on the adaxial surface of the leaves. The unicellular trichome was observed to be of the highest frequency, density and index followed by the branched unicellular trichome in both the adaxial and abaxial surfaces.

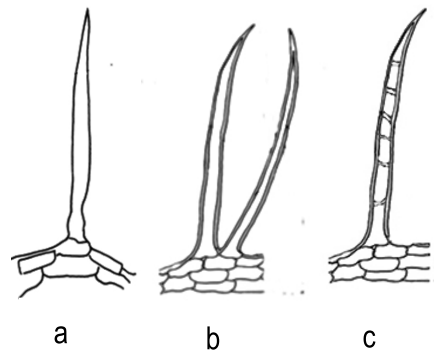

Figure 2. Trichome types observed in the study, a- simple unicellular trichome; b- branched unicellular trichome and c- multicellular trichome

The multicellular trichome had the least average frequency percentage of 14.28 and 20.00 for adaxial and abaxial respectively in the control and 13.33 and 19.05 for adaxial and abaxial respectively in treatment plant while the least average density of 1.56 and 1.90 for adaxial and abaxial respectively in the control and 0.85 and 1.52 for adaxial and abaxial respectively in treatment plants (Table 8).

Table 8. Trichome parameter as affected by different concentrations of spent oil pollution

Treatment	Leaf surface	Trichome type	Trichome frequency (%)	Trichome density (mm²)	Trichome Index (%)
Control	Adaxial	Unicellular	57.14	8	5.97
		Branched unicellular	28.57	4	3.08
		Multicellular	14.28	2	1.56
	Abaxial	Unicellular	60.00	9	5.49
		Branched unicellular	20.00	3	1.90
		Multicellular	20.00	3	1.90
50 mL	Adaxial	Unicellular	60.00	9	6.25
		Branched unicellular	26.67	4	2.88
		Multicellular	13.33	2	1.46
	Abaxial	Unicellular	53.33	8	4.37
		Branched unicellular	26.67	4	2.23
		Multicellular	20.00	3	1.68
100 mL	Adaxial	Unicellular	55.55	10	5.21
		Branched unicellular	27.78	5	2.67
		Multicellular	16.67	3	1.62
	Abaxial	Unicellular	55.00	11	5.09
		Branched unicellular	25.00	5	2.38
		Multicellular	20.00	4	1.91
150 mL	Adaxial	Unicellular	60.00	12	4.55
		Branched unicellular	25.00	5	1.95
		Multicellular	15.00	3	1.18
	Abaxial	Unicellular	52.38	11	4.07
		Branched unicellular	28.57	6	2.27
		Multicellular	19.05	4	1.52
200 mL	Adaxial	Unicellular	60.00	9	3.70
		Branched unicellular	26.67	4	1.68
		Multicellular	13.33	2	0.85
	Abaxial	Unicellular	56.25	9	3.53
		Branched unicellular	18.75	3	1.21
		Multicellular	25.00	4	1.60

DISCUSSION

The result of germination of the African okra seeds used in the experiment varies in oil contaminated soils with spent engine oil. Germination was recorded for all the treatment and there was reduction in the days of germination as the concentration of spent engine oil increased in the soil samples compared to the control and this was statistically significant at P<0.05. The control pots germinated within 4 to 10 days after planting, while seeds in contaminated soils germinated slowly from the 6th day and continued until about 3 weeks after planting. The delay in then contaminated soils might be as a result of the volatile nature of the spent engine oil which melt into the soil and unable the seeds to imbibe enough water for germination. It is obvious that oil in soil could have affected the biological oxygen demand (BOD) level, thereby interfering with normal gaseous exchange which is required for germination and growth performance. Oluwanisola and Abdulrahaman, (2018) reported delayed in two varieties of okra seeds when polluted with spent engine oil. Also, Nwoko *et al*. (2007) observed that spent engine oil soil-plant microenvironment affects normal soil chemistry thereby reducing uptake and release of nutrient. This result was also in tandem with the report of Osuagwu *et al.* (2017) that germination percentage, days to germination and rate of germination of *A. hypogea*, *Z. mays* and *V. unguiculata* in spent engine oil contaminated soil increased the germination percentage, days to germination and rate of germination decreased. There was a significant difference among the treatment compared with the control (P<0.05)

Osawuru and Abioye (2012) also reported delay in the germination of *A. esculentus* in spent engine oil treated soils. Agbogidi and Enujeke (2012) reported a significant oil-dose dependent reduction in the germination percentage of *Arachis hypogaea* seeds planted in spent oil contaminated soil. They also observed delay in seedling emergence in the contaminated soils.

The plant height, number of leaves and leaf area of the plants were affected by the spent engine oil and the results tend toward the same way as the concentrations of the oil increased, the vegetative parameter reduced. Statistical analysis of the vegetative growth showed that there is significant difference in all the vegetative parameter studied. The effect of the spent engine oil was more obvious on the contaminated soil-plant than those of the control. The plants in control soils with their roots undisturbed could have absorbed enough nutrients when compared to the seedlings exposed to higher spent engine oil treatment. These findings support the report of Onwusiri *et al.* (2017) that the mean leaf number, leaf area and plant heights of fluted pumpkin decreased with increasing level of contamination with spent engine oil. They reported that the mean leaf number, leaf area and plant heights of fluted pumpkin decreased with increasing level of contamination with spent engine oil. Also, Osawuru and Abioye (2012) on their work on *A. esculentus* where the control plants grew better than plants sowed in soils treated with spent oil

Agbogidi and Eruotor (2012) reported that spent engine oil contamination on soil significantly affected the heights, number of leaves, collar diameter and leaf area of seedlings with reductions in these parameters being concentration dependent. Nwoko *et al.* (2007) reported that the presence of spent engine oil in the soil-plant microenvironment appeared to have an effect on the normal soil chemistry such that nutrient release and uptake as well as the amount of water could be reduced. Vwioko and Fashemi (2005) among many other authors have reported that reduction in plant height is a visible effect of substances that are toxic, and thus, inhibit plant growth.

The concentration-dependent reduction in leaf area and number of leaves agrees with the reports of Agbogidi *et al.* (2005), Vwioko and Fashemi (2005) and Sharifi *et al*. (2007). According to Kathirvelan and Kalaiselvan (2007) the leaf surface area greatly determines the amount of carbon gained through photosynthesis, the amount of water lost through transpiration and ultimately the crop yield. Therefore, the reduction of the leaf area as was observed in this study implies that there would be a low photosynthetic efficiency of the plant as much of the solar energy emitted by the sun would not be absorbed by plants for photosynthesis which in turn lead to low yields of the plants.

Previous study by Okon and Mbong (2013) showed that there was significant reduction in parameters such as plant height, number of nodes, leaf area of okra sown on engine oil polluted soil, whereas, the parameters were improved in polluted soil which were amended with poultry droppings. The works of Odjegba and Sadiq (2002); Sun *et al.* (2004); Adedokun and Ataga (2007); Njoku *et al.* (2008) and Okon *et al.* (2012) reported observations similar to the findings of this work on growth parameters. Walker *et al*. (2001) reported that availability of nitrogen in the soil directly affects the relative growth rate of plants. Agbogidi *et al*. (2005) also, stressed that petroleum-products are known to reduce nitrogen availability and can have adverse effects on the plant growth parameters with changes in percentage concentration. This work was also in support of the work of Swapna *et al.* (2021) who reported that spent engine oil had a prominent negative impact on the soil characteristics, plant growth and the phytochemical content of *Amaranthus hybridus*. In their work, the most critical soil parameters like pH, organic carbon, bulk density, particle density, etc. were got increased and electrical conductivity, nitrogen, phosphorus, potassium, water holding capacity, and porosity were decreased as the concentration of spent engine oil in the soil increased.

The significant percentage differences observed in the yield parameters such as length, dry weight, fresh weight and number of fruits could be attributed to the high level of oil in soil and hence the uptake of ions and nutrients carried out by the roots becomes difficult (Onwusiri *et al.* 2017). The control and spent oil treatment plants were compared and a concentration dependent reduction in these parameters was observed in the control and the treatment plants except for the 200 ml pots which did not fruit throughout.

The plants in control soils with their roots undisturbed could have absorbed enough nutrients when compared to the those exposed to higher spent engine oil treatment. Agbogidi (2010) reported that a reduction in shoot growth is a direct resultant effect of engine oil pollution in the root growths as roots are input organs for the absorption and translocation of water and mineral nutrients. Furthermore, Agbogidi and Eshegbeyi (2006) noted that hydrocarbons from oil contaminated soils accumulate in the chloroplast of plant leaves. This makes the photosynthetic ability of the leaves become reduced, thereby affecting translocation in affected plants which might be due to obstruction of the xylem and phloem vessels consequently causing reduction in photosynthetic products and dry matter content of the entire plants.

Little is known about the role of the plant cell wall and its binding properties in relation to spent oil tolerance. However, what is reported on has been somewhat controversial. In soil solution, the root cell wall is in direct contact with the metals, so the adsorption of metals onto the cell wall must be low. However, Mehes-Smith *et al*., (2013) reported that a significant amount of metal accumulation occurred between the cell wall and the cell membrane
The influence of pollutant in contaminated soils or concentrations of these elements in plants has been widely studied and it is generally accepted that total oil concentrations in soil do not provide a good indication of the levels potentially available to plants (Ullrich *et al.,* 1999).
The present study showed that stomatal and trichome types observed remain unchanged for all treatments as the basic architectural design did not change under the influence of spent engine oil pollution. This is an indication that they are stable and are of good taxonomic indicator. Alege *et al*. (2013) expressed that such stability is under strong genetic control and may play a significant role in taxonomy. However, the stomatal size, subsidiary and epidermal cell sizes were reduced while the density of stomata and epidermal cell were observed to be increased with increase in pollutant concentration. This significant variation was in agreement with the findings of Oluwanisola and Abdulrahaman, (2018), they observed significant variations in stomatal size, density accessory and ordinary epidermal cell sizes, which were observed to reduce with increase in pollutant concentration. Komolafe *et al.* (2015) reported that higher concentration of pollutants is responsible for the broken and scattered epidermal cells and smaller sizes of the stomata in Guinea corn. These were also observed in both control and treatment plants but were much in the treatment plants. Since the reduction in the amount of water available by the presence of spent oil in the soil was inevitable, it is likely that these features were elaborated to cope with the drought.

Conclusion
The results of this study have demonstrated that soil contamination with spent engine oil reduces the capacity of plant growth in soil. Spent oil pollution delayed or inhibited the germination of seeds sown in the soil; growth and leaf anatomical configuration were also significantly affected. In order to avoid economic loss through low productivity of crops, and prevent alterations in plant anatomic makeup, it is recommended that soil amendment be done on soils polluted with spent engine oil and its associated products before crops such as West African okra are cultivated.

REFERENCES
Adedokun, O. M. and Ataga, A. E. (2007). Effects of amendments and bioaugumentation of soil polluted with crude oil, automotive gasoline oil, and spent engine oil on the growth of cowpea (*Vigna ungiculata* Walp L.). *Scientific Research and Essays*, 2(5): 147-149.
Adeniji, O. T. and Aremu, C. O. (2007). Interrelationships among characters and path analysis for pod yield components in West African okra (*Abelmoschus caillei* (A.Chev) Stev*). Journal of Agronomy*, 6(1): 162-166.
Adenipekun, C. O., Oyetunji, O. J. and Kassim, L. Q. (2009). Screening of *Abelmoschus esculentus* L. Moench for tolerance to spent engine oil. *Journal of Applied Biosciences*, 20: 1131 - 1137
Agbogidi, O. M. and Enujeke, E. C. (2012). Effects of spent motor oil on soil physico-chemical properties and growth of *Arachis hypogaea* L. *Journal of Bioscience and Technology,* 1(1): 71-74.
Agbogidi, O. M. and Eruotor, P. G. (2012). Morphological changes due to spent engine oil contamination and its heavy metal components of *Jatropha curcas* (Linn.) seedlings. International Conference on Bioscience, Biotechnology and Healthcare Sciences (ICBBHS'2012) December 14-15, 2012, Singapore.
Agbogidi, O. M. and Eshegbeyi, O. F. (2006). Performance of *Dacryodes edulis* (Don. G. Lam H.J.) seeds and seedlings in a crude oil contaminated soil. *Journal of Sustainable Forestry,* 22(3/4): 1-13.
Agbogidi, O. M. (2010). Mental contents of *Rhizophora racemosa* (G.F.W. Meyer) seedlings grown in crude oil contaminated soils. *Obeche Journal*, 28(2): 112-118.
Agbogidi, O. M., Nweke, F. U. and Eshegbeyi, O. F. (2005). Effects of soil pollution by crude oil on seedling growth of *Leucaena leucocephala* (Lam.) de Witt. *Global Journal of Pure and Applied Sciences*, 4(2): 453-456.
Ahamefule, H. E., Olaniyan, J. O., Amana, S. M., Eifediyi, E. K., Ihem, E. and Nwokocha, C. C. (2017). Effects of spent engine oil contamination on soybean (Glycine max L. Merril) in an *Ultisol.* Journal of Applied Science and Environmental Management, 21 (3): 421-428
Ahamefule, H. E; Nwokocha, C. C. and Amana, S. M. (2015). Stability and hydrological modifications in a tilled soil under selected organic amendments in south-eastern Nigeria. *Albanian Journal of Agricultural Science*, 14(2): 127 – 136.
Alege, G. O. Okutachi, M. M. and Olubiyo, G. T. (2013). Stability of foliar epidermal attributes in sesame (*Sesamum indicum* L.). *Journal of Biology, Agriculture and Health Sciences,* 2(3): 1-6.
Dilcher, D. L. (1974). Approaches to the identification of angiosperm remains. *The Botanical Review,* 40(1): 1 –157.
Ikhajiagbe,b., Omoregie, G. O., Ekhator, P. O. and Mshelbulla, P. B. (2017). Growth and yield responses of okra (*Abelmoschus esculentus)* in a petroleum hydrocarbon-polluted soil after Substrate amendment. *Ethiopian Journal of Environmental Studies & Management*, 10(7): 866 – 876.
Kathirvelan, P. and Kalaiselvan, P. (2007). Groundnut (*Arachis hypogaea* L.) leaf area estimation using allometric model. *Research Journal of Agricultural Biological Science*, 3(1): 59-61.
Kehinde, O. B. (1999). Floral biology of West African okra. *Nigerian Journal of Genetics*, 14:14-17.
Komolafe, R. J., Akinola, O. M. and Agbolade, O. J. (2015). Effect of Petrol and Spent oil on the Growth of Guinea corn (Sorghum bicolor L.). *International Journal of Plant Biology*, 6(1): 5883
Njoku, K. L., Akinola, M. O. and Oboh, B. O. (2008). Germination, survival and growth of accession of *Glycine max* L. (Merril) (soybean) and *Lycopersicon esculentum* L. (tomato) in crude oil polluted soil. *Research Journal of Environmental Toxicology,* 2(2): 77-84. DOI: 10.3923/rjet.2008.77.84
Nwoko, C. O., Okeke, P. N., Agwu, O. O. and Akpan, I. E. (2007). Performance of *Phaseolus vulgaris* L. in a soil contaminated with spent engine oil. *African Journal of Biotechnology,* 6(16): 1922-1925. DOI**:** 10.5897/AJB2007.000-2291

Obiremi, E. O. and Oladele, F. A. (2001). Water-conserving stomatal systems in selected *Citrus* species. *South African Journal of Botany,* 67(2): 258-260. DOI:10.1016/S0254-6299(15)31127-3

Odjegba, V. J. and Sadiq, A. O. (2002). Effect of spent engine oil on the growth parameters, chlorophyll and protein levels of *Amaranthus hybridus* L. *Environmentalist,* 22(1): 23-28. https://doi.org/10.1023/A:1014515924037

Okon, J. E. and Mbong, E. O. (2013). Effects of nutrient amendments of spent engine oil polluted soil on some growth parameters of *Abelmoschus esculentus* (L.) Moench. in south-south Nigeria*. Bulletin of Environment, Pharmacology and Life Sciences*, 2(5): 75-78.

Okon, J. E., Esenowo, G. J., and Umoh, N. S. (2012). Effect of crude oil pollution of soil and amelioration treatment on the growth of eight varieties of *Manihot esculenta* Crantz. *International Journal of Chemical, Environmental and Pharmaceutical Research,* 3(2): 163-169.

Oluwanisola, P. O. and AbdulRahaman, A. A. (2018). Anatomical and physiological effects of spent-engine oil on two varieties of *Abelmoschus esculentus* (L.) Moench. from Malvaceae. *Notulae Scientia Biologicae,* 10(4):584-596. DOI: 10.25835/nsb10410265

Onwuka, M. I., Chude, V. O. and Ogwuegbu, G. C. (2012). Remediation of spent engine oil polluted soil using two types of organic manure and their effects on maize growth*. Nigerian Journal of Soil Science,* 22(2):245.

Onwusiri, K. C. Aguoru, C. U. and Akomolafe, G. F. (2017). Effect of spent engine oil on germination and growth parameters of fluted Pumpkin (*Telfairia occidentalis* Hook F.) in Makurdi, Benue State, Nigeria. *Journal of Research in Forestry, Wildlife & Environment,* 9(4): 2141 – 1778

Osawaru, M. E. and Abioye, O. (2012). Growth performance of okra [*Abelmoschus esculentus* (L.) Moench] cultivar in soil polluted at different concentrations of spent engine oil. *African Scientist*, 13(1): 11-22.

Osuagwu, A. N., Ndubuisi, P. and Okoro, C. K. (2017). Effect of spent engine oil contaminated soil on *Arachis hypogea* (L.), *Zea mays* (L.) and *Vigna unguiculata* (L.) Walp. *International Journal of Advance Agricultural Research*, 5: 76-81

Sharifi, M., Sadeghi, Y. and Akharpour, M. (2007). Germination and growth of six plant species on contaminated soil with spent oil. *International Journal of Environmental Science and Technology,* 4(4): 463 – 470.

Steel, R. G. D. and Torrie, J. H. (1981). Principles and procedures of statistics: a biometrical approach. *Journal of American Statistics Association,* 76(375) 753-754.

Sun, W. H., Lo, J. B., Robert, F. M., Ray, C. and Tang, C. S. (2004). Phytoremediation of petroleum hydrocarbons in tropical coastal soil: I. selection of promising woody plants. *Environmental Science and Pollution Research,* 11(4): 260-266.

Swapna, A. A., Vijayammal, R. and Radha, D. S. (2021). Effects of spent engine oil on soil characteristics and selected phytochemicals in *Amaranthus hybridus*. Research square: https://doi.org/10.21203/rs.3.rs-939651/v1

Vwioko, D. E. and Fashemi, D. S. (2005). Growth response of *Ricinus cummunis* L. (castor oil) in spent lubricating oil polluted soil. *Journal of Applied Sciences and Environmental Management*, 9(2): 73-79.

Walker, R. L., Burns, I. G. and Moorby, I. (2001). Response of plant growth rate to nitrogen supply: A comparison of relative addition and nitrogen interruption treatments. *Journal of Experimental Botany,* 52(355): 309-317. https://doi.org/10.1093/jexbot/52.355.309