



EFFECT OF *Macrophomina phaseolina*, ORGANIC MANURE AND SPENT ENGINE OIL ON *Luffa aegyptica* (Mill)

*Ani, E.,^{1,2} Adekunle, A. A.¹ Kadiri, A. B.¹ and Njoku, K. L.³

¹Department of Botany, University of Lagos, Akoka, Lagos. Nigeria.

²Department of Biological Sciences, Yaba College of Technology, Yaba, Lagos. Nigeria.

³Department of Cell Biology and Genetics, University of Lagos. Nigeria

*Corresponding author: emson4j@yahoo.com, 07037816642

ABSTRACT

There is an increasing environmental concern over improper disposal of spent engine oil (SEO) from serviced engines. These concerns are the results of increasing pollution of the environment by SEO with its negative consequences on plants and other forms of life. The present study assessed the effect of *Macrophomina phaseolina* and nutritional amendment on *Luffa aegyptiaca* grown on spent engine oil polluted soil in a screen house. The effect of SEO and nutritional amendment on *L. aegyptiaca* was assessed on germination, plant height, leaf area, stem girth and number of leaves. Two concentrations of SEO (100 and 200ml) in four kilogram of soil were used in four replications each. Results show that SEO adversely affected the germination and growth of *L. aegyptiaca* as delayed germination, reduction in heights, leaf area and number of leaves were observed in the crop planted in SEO polluted soil. Germination (emergence) of *L. aegyptiaca* seeds planted in SEO polluted soil began after four days compared to two days for plants in untreated soil. Germination efficiency for seeds planted in unpolluted soil was up to 100% but reduced to 20% when the soil was polluted with SEO. However, nutritional amendment and *M. phaseolina* increased seed germination in SEO polluted soil from 20% to 60%. Nutritional amendment and *M. phaseolina* were thus shown to enhance the germination and growth of *L. aegyptiaca* in SEO polluted soil.

Keywords: *Macrophomina phaseolina*, *Luffa aegyptiaca*, Germination, Effect, Spent engine oil

INTRODUCTION

Luffa aegyptiaca belong to the family cucurbitacea and is commonly called sponge gourd, loofa, vegetable sponge or bath sponge (Partap, 2012). The leaf is 13cm and 30cm in length and width respectively and has the acute-end lobe. It is hairless and has serrated edges. The flowers of *L. aegyptiaca* is yellow and blooms from August- September (Mazali *et al.*, 2005). Its fruit is green and has a large cylinder like shape. Generally, *L. aegyptiaca* can be used in virtually all areas. Factors such as high surface area per volume, strong and durable structure, low specific gravity (which makes it light) and reasonable cost are characteristics of loofa making it a suitable alternative as a packing medium in an attached growth system (Mazali and Alves, 2005) Young fruits are edible and matured fibers are generally used as bathroom sponge, in washing ships and decks, manufacturing slippers or baskets, as shoes, mats, (Partap, 2012). In traditional African medicine, *L. aegyptiacac* or its extracts are use in the treatment of constipation, as a diuretic, nose cancer, as an abortifacient, to promote wound healing, oedemas, for the treatment of malaria, *enterobiasis*, *filariasis*, whooping cough, stomach-ache, to facilitate childbirth and as a purgative. Young fruits are used as vegetable. (Sangh *et al.*, 2012).

Engine oil or motor oil is used around the world in cars and other engines like power generating sets. The contamination of soil by used engine oil is on the increase due to increased use of engines that use petroleum products (Mandri. and Lin 2007). In Nigeria, environmental pollution caused by improper

disposal of spent engine oil has reached alarming proportion. Plants especially legumes have been identified to play important role in remediating oil polluted soil. (Mojiri *et al.*, 2012, Parrish *et al.*, 2005). A number of researches have been carried out on the effect of pollutants on plants. Abii *et al.*, 2009 studied the effect of crude oil spillage on soil fertility and plant growth. Odjegba *et al.*, 2002 and Agbogidi *et al.*, 2010; showed the effect of spent engine oil on plants. Some of the most commonly observed symptoms of oil pollution on plants include; deformation of chlorophyll, alteration in the stomata mechanisms and reduction in photosynthesis and respiration, increase in the production of stress related phytohormones (Larcher *et al.*, 2003), accumulation of toxic substances or their by product in vegetal tissue, decrease in size and less production of biomass (Adenipekun *et al.*, 2008). *Macrophomina phaseolina*, is a soil and seed-borne polyphagous pathogen. It causes diseases of more than 500 crop and non-crop species, including economically important hosts such as soybean, common bean, corn, sorghum, cowpea, peanut and cotton (Dhingra and Sinclair, 1977; Ndiaye *et al.*, 2010). The fungus has a worldwide distribution, but is regarded as economically more important in subtropical and tropical countries with semi-arid climates (Wrather *et al.*, 2001). *Macrophomina phaseolina* is known to induce diseases on a range of crops, ranging from seedling blight, root and stem rot, wilt, and pre- to post-emergent damping off, which result in decreased stem height, girth, root and head weight, or death, of affected plants (Wrather and Koenning 2010).

However, the potentials of *Macrophomina phaseolina* to enhance the germination and growth of plants in polluted environment has not been explored. The present study investigates the influence of *Macrophomina phaseolina* and nutritional amendment on *Luffa aegyptiaca* grown on spent engine oil polluted soil.

MATERIALS AND METHODS

Sample collection

Mature and dried seeds of *Luffa aegyptiaca* were collected at Somolu, Lagos State (5°33'N, 3°20'E), spent engine oil was collected at local Auto Mechanic workshop, Sandy loam soil was collected from Yaba College of Technology staff quarters (6° 31'N, 3° 40'E) while Processed organic manure was collected from Lagos state Waste Management Authority, LAWMA, Ikorodu, Lagos. (6° 35' N, 3°22'E).

Isolation of fungi from rhizosphere of *L. aegyptiaca*

Macrophomina phaseolina was isolated from the rhizosphere of *L. aegyptiaca* following the method described by Reyes and Mitchell (1962).

Physio chemical analysis of soil and spent engine oil

Physico-chemical characteristics of the experimental soil and spent engine oil used were analyzed using 250 pH/Ion/Conductivity Meter. The particle size distribution were determined, soil pH were determined in distilled water using soil liquid ratio of 1:1, electrical conductivity were measured using conductivity bridge. Phosphate- phosphorous, total nitrogen were also determined, Nitrate-nitrogen was determined by the phenol-disulphonic, Organic carbon were measured using wet combustion method and converted to organic matter by multiplying the values of organic carbon by a factor of 1.722. Exchangeable cation was determined on atomic absorption spectrophotometer (Adekunle *et al.*, 2015).

RESULTS

The physico-chemical properties of the soil and spent engine oil used for the experiment are presented in table I below.

Table 1: Physiochemical properties of the soil and spent engine oil used

Properties of the soil and Spent engine oil	Soil	Spent engine oil
PH	6.30	6.04
Total organic carbon (TOC %)	1.59	ND
Moisture content (%)	ND	13.92
Temperature (C ⁰)	ND	29.80
Organic matter %	2.74	ND
Density g/cm ³	ND	0.94
Viscosity	ND	24.00
Clay %	30.00	ND
Silt %	47.00	ND
Flash point	ND	122.00
Sand %	23.00	ND
Sulphur (mg/kg)	1.01	0.97
Exchangeable cation (cmo/kg ⁻¹)	187.28	ND
Available Phosphate (mgkg ⁻¹)	10.20	ND
Available sulphate (mgkg ⁻¹)	119.70	ND
Iron (mg/kg)	180	26.92
NO ₃ (mgkg ⁻¹)	48.00	ND

Key: ND = Not determined

Experimental design

The research work was conducted in Botanical Garden of Yaba College of Technology, Yaba, Lagos, Nigeria (latitude 6° 31'N, longitude 3° 40'E). Four kilogram(4kg) of sieved and dried sandy-loam soil was measured into an experimental bucket (7L) with a measuring cylinder. Spent engine oil (100ml and 200ml) were each introduced independently into some of the buckets and mixed thoroughly. Viable seeds of *L. aegyptiaca* were planted 24hrs after introduction of the SEO at a depth of 3cm. Viability of the seeds was tested by floatation method as described by Jephris *et al.*, 2015 before planting. Processed organic manure was introduced seven days after planting into some of the buckets. *M. phaseolina* was introduced 24hrs after organic manure was introduced. Trays were placed under each treated buckets with SEO to retain the SEO that might have wash down from the soil through the perforated buckets during watering and are poured back into the bucket. Each experiment was set up in four replications using Randomized complete block Design.

Data collection

The effect of spent engine oil on Leaf area, stem girth, plant height and number of leaves of *L. aegyptiaca* were measured at two weeks' interval for 24weeks while the effect on germination was measured daily. Leaf area was calculated using the method described by Okon and Mbong (2013). The germination percentage was calculated according to Oyedeji *et al.*, 2012.

$$\text{Percentage germination} = \frac{\text{No of germinated seeds} \times 100}{\text{No of seeds sown}}$$

Statistical Analysis

Data were analyzed statistically using IBM SPSS statistics (version 24.0) at (P<0.05)

Effect of *M. phaseolina*, spent engine oil and nutritional amendment on germination of *L. aegyptiaca* seeds

Results of the effect of SEO and *M. phaseolina* on germination of seeds of *L. aegyptiaca* are presented on table 2. Germination (emergence) of *L. aegyptiaca* seeds in Spent engine oil polluted soil began after four days compared to two days for plants in

untreated soil (control) and three days for seeds planted in soils treated with SEO, nutritional amendment and *M. phaseolina* (fungus). Germination efficiency for seeds planted an unpolluted soil was up to 100% but reduced to 20% when the soil was polluted with SEO. However, nutritional amendment and *M. phaseolina* increased seed germination in SEO polluted soil from 20% to 60%.

Table 2: Effect of *M. phaseolina*, spent engine oil and organic manure on germination of *L. aegyptiaca* seeds

Treatments	No of seeds planted	No of seeds germinated	Germination (%)
Plant only (T ₁)	10	9	90
Plant + Organic manure (T ₂)	10	8	80
Plant + fungus (T ₃)	10	8	80
Plant + fungus + Org. M (T ₄)	10	10	100
Plant + SEO (T ₅)	10	2	20
Plant + SEO + fungus (T ₆)	10	4	40
Plant + SEO + Org. M(T ₇)	10	4	40
Plant + SEO + fungus + org. M(T ₈)	10	6	60

Effect of *M. phaseolina*, spent engine oil and nutritional amendment on the Growth of *L. aegyptiaca*

The effect of SEO, *M. phaseolina* and nutritional amendment on the height, leaf area, number of leaves and stem girth of *L. aegyptiaca* are presented on table 3. Statistical analyses show that plant grown in non-oil contaminated soil performed better than those in Spent Engine Oil-polluted soil. Some of the

observed effects of SEO on the growth of *L. aegyptiaca* include reduced heights, reduction in number and leaf areas in addition to chlorosis on the leaves. The effect of spent engine oil on *L. aegyptiaca* was shown to increase with increased concentration. The introduction of *M. phaseolina* and nutritional amendment on the polluted soil however, improved these agronomic parameters (table 3).

Table 3: Effect of SEO on, plant height, no of leaf, leaf area and stem girth of *L. aegyptiaca*

Treatments	Plant height (Inch)	No of Leaf	Leaf Area (Cm ²)	Stem girth (Cm)
P only	46.17 ± 15.53 ^a	20.50 ± 3.89 ^{abcd}	9.60 ± 1.92 ^a	0.35 ± 0.02 ^{ab}
P + F	50.83 ± 13.73 ^a	30.17 ± 6.25 ^{cd}	11.87 ± 1.45 ^a	0.40 ± 0.00 ^{ab}
P + OM	46.17 ± 13.98 ^a	26.17 ± 5.47 ^{bcd}	9.30 ± 1.24 ^a	0.37 ± 0.02 ^{ab}
P + SEO (100ML)	22.33 ± 9.02 ^a	13.33 ± 2.69 ^{ab}	6.17 ± 1.60 ^a	0.18 ± 0.03 ^a
P + SEO (200ML)	9.00 ± 2.22 ^a	7.00 ± 0.86 ^a	3.47 ± 0.56 ^a	0.12 ± 0.02 ^a
P + SEO (100 ML) + F	40.50 ± 12.74 ^a	22.00 ± 4.91 ^{abcd}	7.87 ± 2.14 ^a	0.13 ± 0.02 ^a
P + SEO (200 ML) + F	16.00 ± 4.53 ^a	16.33 ± 2.95 ^{abcd}	4.73 ± 0.63 ^a	0.10 ± 0.00 ^a
P + SEO (100 ML) + OM	35.83 ± 10.82 ^a	17.50 ± 3.47 ^{abcd}	8.40 ± 1.79 ^a	0.78 ± 0.08 ^{bc}
P + SEO (200 ML) + OM	29.00 ± 8.61 ^a	15.33 ± 3.26 ^{abc}	7.53 ± 1.78 ^a	1.07 ± 0.39 ^c
P + SEO (100 ML) + F + OM	49.67 ± 15.93 ^a	31.50 ± 8.35 ^d	9.15 ± 1.92 ^a	0.76 ± 0.16 ^{bc}
P + SEO (200 ML) + F + OM	32.67 ± 8.85 ^a	29.17 ± 6.52 ^{bcd}	8.13 ± 2.44 ^a	0.64 ± 0.12 ^b
F – Statistics	F _{10,55} = 1.537 p = 0.151	F _{10,55} = 2.582; p = 0.012	F _{10,55} = 1.949; p = 0.058	F _{10,55} = 5.634; p < 0.001

NB: Means with the same superscripts are not significant at 5% level of significance

Key: P= Plant, F=Fungi, OM = Organic Manure, SEO = Spent Engine Oil,

DISCUSSION

Pollution of the environment by SEO from serviced automobiles and machines is constituting a major challenge to agricultural productivity in Nigeria. The negative influence of oil pollution to agricultural crops and soil micro-organisms has been highlighted by some authors. (Odjegba *et al.*, 2002; Agbogidi *et al.*, 2010; Anoliefo, and vivioko, 2006.). This study demonstrated that used motor oil has significant effect on the germination and growth of *L. aegyptiaca*. Germination of *Luffa aegyptiaca* seeds was delayed in SEO polluted soil similar to the findings of (Adenipekun *et al.*, 2009, Nwite and Alu

2015). The effect of used motor oil on *L. aegyptiaca* was observed in reduction in the number and leaf area, reduced height, internodes and stem girth similar to the findings of Agbogidi *et al.*, 2006, Igbal *et al.*, 2016 who showed that crude oil application to soil significantly reduced the growth of okra and that there was reduction in number of leaves for *Parkinsonia aculeate* grown in an oil polluted soil. The response of seeds and seedlings of *L. aegyptiaca* in SEO polluted soil shows that the negative effect of SEO on the growth parameters measured increased with increased concentration of SEO similar to the findings of Gamal, (2005).

However, treatment of soil polluted with used motor oil with *M. phaseolina* and nutritional amendment significantly ($p < 0.05$) improved the agronomic parameters compared with polluted soil without *M. phaseolina* and nutritional amendment. *Macrophomina phaseolina* and nutritional amendment is shown by the result to enhanced the ability of *L. aegyptiaca* to thrive in SEO polluted soil similar to the finding of Adu (2015). Fungi have been known to improve the performance of plant under environmental stress through modifying root architecture (Hooker and Atkinson, 1996), improving membrane function (Graham *et al.*, 1981) and by enhancing production of oxidative enzymes (Salzer *et al.*, 1999). The ability of fungi to increase the supply of mineral nutrients to plant, stimulate plant growth through the production of phytohormones have been highlighted by (Prakash and Sheela 2016). This study shows that nutritional amendment and *M. phaseolina* improved the germination and growth of *L. aegyptiaca* in SEO polluted soil

CONCLUSION

The role of *M. phaseolina* nutritional amendment in germination, survival and growth *Luffa aegyptiaca* SEO polluted soil is highlighted in this study the ability

REFERENCES

- Abii, T.A. and Nwosu, P.C. (2009): The effects of oil spillage on the soil of Eleme in Rivers state of Nigeria. *Research Journal of Environmental Toxicology*, **1**(3): 116-123
- Adekunle, A. A., Ani, E. and Kanife, U. C. (2015). Biodegradation of petroleum oils by fungi isolated from oil palm fruit and mechanic village, *Ife Journal of Science*, **17**(3), 613-625
- Adenipekun, C.O., Oyetunji, O.J. and Kassim, L.Q. (2009): "Screening of *Abelmoschus esculentus* L. Moench for Tolerance to Spent Engine," *Journal of Applied Biosciences*, **20**:1131-1137
- Adenipekun, C.O., Oyetunji, O.J. and Kassim, L.S. (2008): Effect of spent engine oil on the growth parameters and chlorophyll content of *Cocchorus olitorus* Linn. *Environmentalist*, **28**: 446-450.
- Adu, A. A.; Aderinola, O. J. and Kusemiju, V. (2015): Comparative effects of spent engine oil and unused engine oil on the growth and yield of *Vigna unguiculata* (Cowpea). *International Journal of Science and Technology*, **4**(3):105-118.
- Agbogidi O.M, Onosode A.T, Okonta B.C. (2006): Susceptibility of *Dennettia tripetala* (Bak.) F. seeds to crude oil. *Journal of Food, Agriculture and Environment*, **4**(2): 350–352.
- Agbogidi, O.M. and Egbuchua, C.O. (2010): Heavy metal concentrations of soil contaminated with spent engine oil in Asaba, Delta State. *Acta Agronomica Nigeriana*, **9**(2): 1-6.
- of rhizospheric fungus (*M. phaseolina*) to enhance the germination and growth of *L. aegyptiaca* in SEO contaminated soil has also been highlighted. Germination of *L. aegyptiaca* was delayed in SEO polluted soil. Some of the observed effects of SEO on the growth of *L. aegyptiaca* include reduced heights, reduction in number and leaf areas in addition to chlorosis on the leaves. The effect of spent engine oil on *L. aegyptiaca* was shown to increase with increased concentration. A better understanding of the function and diversity of rhizospheric fungi is needed to enhance optimal performance of plants in an oil polluted environment.

Author's contributions:

1. Ani, E. Participated actively in design, experimentation and drafting of manuscript
2. Adekunle, A. A. Participated actively in design and supervision of the research
3. Kadiri, A. B. Actively participated in design and statistical analysis
4. Njoku, K. L. Actively participated in experimentation and manuscript drafting.

Conflict of interest:

The authors declare that there is no conflict of interests

- Mandri T. and Lin J (2007): Isolation and characterization of engine oil degrading indigenous microorganisms in Kwazulu-Natal. *African Journal of Biotechnology*, **6**: 23-27.
- Mazali, I.O. and Alves, O.L. (2005): Morphosynthesis: high fidelity inorganic replica of the fibrous network of loofa sponge (*Luffa Cylindrica*). *An Academy Base Journal*, **77**(1): 25-31.
- Mojiri, A., Abdul Aziz, H., Zaman, N.Q. and Azi, S.Q. (2012): A Review on Anaerobic Digestion, Bio-reactor and Nitrogen Removal from Wastewater and Landfill Leachate by Bio-reactor. *Advances in Environmental Biology*, **6**(7): 2143-2150,
- Ndiaye M., Termorshuizen, A.J. and van Bruggen, A.H. (2010). Effects of compost amendment and the biocontrol agent *Clonostachys rosea* on the development of charcoal rot (*Macrophomina phaseolina*) on cowpea. *Journal of Plant Pathology*, **92**: 173–180
- Nwite, J. N. and Alu, M. O. (2015): Effect of different levels of spent engine oil on soil properties, grain yield of maize and its heavy metal uptake in Abakaliki, Southeastern Nigeria. *Journal of Soil Science and Environmental Management*, **5**(4):44-51
- Odejgba, V.J. and Sadiq, A.O. (2002): Effect of spent engine oil on the growth parameters, chlorophyll and protein levels of *Amaranthus hybridus* L. *Journal of Environment*, **22**: 23-28
- 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 Science*, **2** (5): 75- 78
- Oyedeki, A. A., Adebisi, A. O. Omotoyinbo, M. O. and Ogunkunle. C. O. (2012): Effect of Crude Oil-Contaminated Soil on Germination and Growth Performance of *Abelmoschus esculentus* L. Moench—A Widely Cultivated Vegetable Crop in Nigeria. *American Journal of Plant Sciences*, **3**:1451-1454
- Parrish, B. D., Luzadis, V.A. and Bentley, W.R. (2005): What Tanzania's Coffee Farmers Can Teach the World: A Performance-Based Look at the Fair Trade-Free Trade Debate. *Journal of Sustainable Development*, **13**:177–189
- Partap, S., Kumar, A., Sharma, N. K. and Jha, K. K. (2012): *Luffa Cylindrica*: An important medicinal plant. *Journal of National Products of Plant Resources*, **2**: 127-134.
- Prakash Deshmukh, Sheela Shinde (2016): Beneficial Role of Rhizosphere Mycoflora in the Field of Agriculture: An Overview. *International Journal of Science and Research*, **5**(8):529-533
- Reyes, A.A. and Mitchell, J. E. (1962): Growth response of several isolates of *Fusarium* in rhizosphere of host and non-host plants. *Phytopathology*, **52**: 1196-1200.
- Salzer, P., Corbiere, H. and Boller, T. (1999): Hydrogen peroxide accumulation in *Medicago truncatula* roots colonized by the arbuscular mycorrhiza-forming fungus *Glomus intradices*. *Planta*, **208**: 319-325.
- Sangh, P., Amit, K., Neeraj, K.S., Jha K.K. (2012): *Luffa Cylindrica*: An important medicinal plant. *International Journal of Agricultural Science*, **2**:127-134.
- Wrather J.A., Anderson, T.R., Arsyad, D.M., Tan, Y., Ploper, L.D., Porta-Puglia, A., Ram, H.H. and Yorinori, V. (2001). Soybean disease loss estimates for the top 10 soybean producing countries in 1998. *Canadian Journal of Plant Pathology*, **23**: 115–221.
- Wrather J.A. and S.R. Koenning, (2010). Soybean Disease Loss Estimates for the United States, 1996– 2009. Delta Research Center: Agriculture Experiment Station. University of Missouri, College of Agriculture, Food and Natural Resources, USA.