Advanced Journal of Environmental Sciences
Volume.16, Number 6; June-2025;
ISSN: 2837-3928 | Impact Factor: 9.38
https://zapjournals.com/Journals/index.php/ajes
Published By: Zendo Academic Publishing

EFFECT OF SPENT ENGINE OIL ON SOIL MICRO FLORA

[1]Okoh, Emmanuel Chidera[1], Ugwu, Chioma Victoria[1] and, Okenwa, Emmanuel Chukwuebuka[1]
Email of the corresponding Author: emmachidera9898@gmail.com

Article Info
Keywords: engine oil, soil, microflora, fungi
DOI
10.5281/zenodo.15600048

Abstract
The contamination of soil by spent engine oil presents a significant environmental challenge, particularly due to its adverse effects on soil microflora, which are essential for maintaining soil health and ecosystem functionality. This study investigated the impact of spent engine oil on soil microflora by analyzing the phytochemical parameters and percentage composition of various particles in contaminated soil samples. Soil samples were examined for moisture (0.87%), pH (5.72), organic carbon (4.68%), nitrogen (0.062%), phosphorus (0.54 mg/kg), potassium (286.50 mg/kg), sodium (51.25 mg/kg), calcium (0.69 mg/kg), and magnesium (0.94 mg/kg). The texture analysis revealed sand (96.5%), silt (3.3%), and clay (0.2%). The microbial assessment showed that soil contaminated with 0 ml of engine oil had the highest percentage occurrence of fungal organisms, whereas 10 ml-contaminated soil exhibited the least occurrence. These findings highlight the negative impact of spent engine oil on soil health. Increased oil concentrations suppress fungal populations, reduce nutrient availability, and alter soil physicochemical properties. This study underscores the need for remediation strategies to mitigate the harmful effects of spent engine oil on soil microflora and overall ecosystem health.

INTRODUCTION

Soil is a crucial natural resource, supporting plant growth and maintaining ecosystems by harboring a diverse array of microorganisms. Microorganisms such as bacteria, fungi, and archaea form the foundation of soil health, contributing to nutrient cycling, organic matter decomposition, and the promotion of plant growth (Sharma *et al*., 2019). These microorganisms, collectively known as soil microflora, play an indispensable role in maintaining the ecological balance of terrestrial environments. However, anthropogenic activities such as, industrial waste

[1] Department of Applied Biology and Biotechnology, Faculty of Biological Sciences, Enugu State University of Science and Technology (ESUT).

disposal and the widespread use of petroleum products have, raised concerns regarding soil contamination. One such contaminant of increasing concern is spent engine oil (Ogbo *et al*., 2018).

Spent engine oil is the byproduct of internal combustion engines and typically contains toxic substances, including heavy metals (lead, cadmium, and zinc), polycyclic aromatic hydrocarbons (PAHs), and other hydrocarbons that accumulate over time due to the thermal breakdown of engine components (Ikhajiagbe and Anoliefo, 2018). As vehicles and machinery operate, engine oil becomes contaminated with these hazardous materials, leading to the disposal of spent oil, which has exceeded its useful life. Improper disposal of spent engine oil, such as pouring it on soil surfaces or in open drains, introduces pollutants into the environment, posing serious threats to soil health (Adewuyi and Okeke, 2020).

The contamination of soil by spent engine oil is a global environmental problem. This contamination not only disrupts soil structure and fertility but also has profound effects on soil microflora. Soil microorganisms are highly sensitive to environmental changes, and the introduction of toxic substances, such as spent engine oil, can alter their diversity, abundance, and function (Li *et al*., 2021). Understanding the effects of spent engine oil on soil microflora is essential for developing effective remediation strategies and ensuring sustainable soil management practices.

The contamination of soil by spent engine oil poses significant risks to environmental sustainability, particularly in regions where proper disposal methods are lacking. Soil is a finite resource, and the degradation of soil quality due to pollution has far-reaching consequences for agriculture, food security, biodiversity, and ecosystem services (Adekunle *et al*., 2021). Understanding the effects of spent engine oil on soil microflora is vital for informing soil management practices and developing bioremediation techniques that can restore contaminated soils to their natural state.

Microorganisms are key indicators of soil health because, they respond quickly to environmental stressors such as pollution. By studying the effects of spent engine oil on soil microflora, we can gain insights into the resilience of microbial communities and their ability to adapt to contaminated environments (Nweke and Okpokwasili, 2020). Additionally, microorganisms play a critical role in the degradation of petroleum hydrocarbons, making them essential for the natural attenuation of oil-contaminated soils (Udebuani *et al*., 2018). Identifying the microbial species and functional groups that are most affected by spent engine oil contamination can guide the development of effective bioremediation strategies that leverage the natural capabilities of these organisms.

Furthermore, this study is justified by the increasing reliance on motor vehicles and industrial machinery, which leads to the generation of large quantities of spent engine oil. The improper disposal of this waste product not only contributes to environmental degradation but also poses risks to human health through the contamination of water sources and the food chain (Amadi *et al*., 2018). By investigating the effects of spent engine oil on soil microflora, this research contributes to a broader understanding of the environmental impact of petroleum-based pollution and supports efforts to mitigate its effects.

This study is also timely given, the global emphasis on sustainable development and the need for environmentally sound waste management practices. The findings of this research will provide valuable data for policymakers, environmental agencies, and industries to develop guidelines for the proper handling and disposal of spent engine oil, with the aim of minimizing its impact on soil ecosystems and promoting environmental sustainability (Okoro *et al*., 2019).

Aim of the Study

The primary aim of this study was to investigate the effects of spent engine oil on soil microflora.

Objectives of the Study

The specific objective was to:

- Determine the effect of spent engine oil on soil physiochemical properties.
- Determine the impact of spent engine oil on soil fertility.
- Determine the relationship between spent engine oil usage and the chemical properties of soil degradation.

MATERIALS AND METHODS

Chemical sources

The spent engine oil was collected from a roadside mechanic workshop in, Engr. Cletus Ugwu, Alins Road, Agbani, Enugu State.

Soil Sample Collection

The soil was collected from a biological garden belonging to the Applied Biology Department of Enugu State University of Science and Technology Agbani.

Research Method

About 20 grams of the garden soil was measured out and diluted in 900ml of distilled water, and a 10-fold dilution of the "soil sample" was obtained. 10ml was measured out from the tenfold dilution of the soil sample and transferred into (5) conical flasks. Different volumes of the spent engine oil of 0.1ml, 0.5ml, 1.0ml and 10.0ml was transferred to the conical flasks, leaving out one conical flask without the spent engine oil to serve control. The volumes in the conical flasks were made up to 100ml with the distilled water, resulting in concentrations of 2ml/20kg, 5ml/20kg, and 10ml/20kg of engine oil. The cultures were incubated for 72hrs at room temperature.

After 72hrs the culture sample was inoculated into SDAC (Sabroud Dextrose Agar) medium and incubated for 3 days. The same procedure was performed for the control after 3 days, and the fungal organisms in the culture medium were identified, and a colony count was performed to determine how the different concentrations of engine oil affected the count of the fungal organisms.

Physicochemical Analysis of Contaminated Soil Samples

The Determination of pH

Oil-contaminated soil pH was determined. Approximately 20g of air-dried soil (passed through 2 min sieve) was weighed into a 50ml beaker. 20ml of distilled water was added and allowed to stand for 30min, and stirred occasionally with a glass rod. The pH water was calibrated with a buffer at pH 7.0 before use. The electrode suspension and the reading on the pH water was noted and recorded accordingly.

Determination of Moisture Content

The moisture content of the soil contaminates soil was determined. An empty container was weighed (Na), and 2g of soil contents was added and weighed again (Wi). Samples were then dried in a hot air oven at 105 - 110°C for 24hrs until constant weight was achieved (Na). The container and dried samples were weighed again. The moisture content was calculated as %moisture W2-Wo W1-W2 × 100

Soil particle Determination

The soil particle size was determined. The air-dried soil was weighed. The weighed sample was transferred to a 1k shake mix cup, and 50 ml of 5% sodium hexametaphosphate was added a living with a 100ml of distilled water. The soil suspension was stirred for 15 min. using manual stirrers. The suspension was transferred from the cup to the glass cylinder using a hydrometer. Distilled water was added to the lower blue line of the cylinder. The volume changed to 1130ml and the hydrometer was removed. The top of the cylinder was covered with hand and inverted several times until all the soil was in suspension.

The cylinder was placed on a flat surface and time was noted. The soil hydrometer was immediately placed into the suspension, and the first reading was obtained after the hydrometer was slide slowly in the suspension. The first reading was obtained after 40 seconds (A). Subsequently, the temperature (Ti) was thermometer after the hydrometer was removed. Recorded using a thermometer after the hydrometer was removed.
The suspension was allowed to stand for 3 h, and a second reading was taken for both the hydrometer (H2) and the temperature (T2) respectively. Calculations:
Sand 100. O C + H + 0.3 (T1 - 20) - 2.0)2
Clay 1 + 2 + 0.3 (T2 - 20) -2.00
Soil 100.0- (% Sand + %Clay)

Chemical Analysis of Oil Contaminated Soil Samples

Determination of nitrogen content in soil samples

The percentage nitrogen content in the soil was determined using the following formula:

$$\% \text{ Nitrogen} = \frac{N \times 0.014 \times Vd \times 10}{A \times \text{Wt. of sample}} \times 100$$

Where:
N = acid normality
Vd = digest volume
A = Aliquot of digest

Determination of the Phosphorus

2g of soil, I teaspoon of carbon black and 40ml of the extracting solution, were all put into a 125ml Erlenmeyer flask. The flask was shaken for 30 min. on a mechanical shaker for 30 min. The suspension was filtered through the Whitman No. 40paper. More carbon was added to obtain a clear filtrate, there after 2ml of the clear supernatant was dispensed in a 20ml test tube and 5ml of distilled water plus 2ml of ammonium molybdate was added. The contents were mixed properly and 1ml of dilute stannous chloride solution was added and mixed again. After 5 min, the percentage transmittance of the electro photometer at 660 nm wavelength was measured, and the reading was recorded using the following formula:

$$PCmg/kg = \frac{\text{Reading} \times 0.61 \times \text{dilution factor}}{\text{Atomic weight of Phosphorus}}$$

Determination of K

The flame photometer was set for K by inserting an appropriate filter (usually of 768 wavelengths). The instrument was set to 100% transmittance solutions by feeding 10 ppm K solution and all the standard were running, and a standard curve was prepared by transmittance solution. Readings against concentration plotting of standard K.

$$K = \frac{\text{Milliequivalent per 100g oven dry weight of soil}}{\text{Conc.of K in the extract from the standard curve}}$$

Determination of Na

The flame photometer was set for Na by inserting an appropriate filter (usually 58 wavelengths). The instrument was set to 100% transmittance by feeding 25 ppm Na solution. All the standard solutions were run, and a standard curve was prepared by plotting the transmittance reading against the concentration of the standard Na solution. The soil extract was run again and calculation was made, using the following formula:

$$Na = \frac{\text{Milliequivalent per 100g oven-dry weight of soil}}{}$$

Conc. of Na in the Na+ extract from the standard curve

Determination of Mg

The soil aliquots of the standard solution (100 mg) were pipetted into two station flasks, and distilled water was added to each flask to make a total volume of 100ml. Twenty milliliters of buffer solution was added to obtain a pH of 10. This was followed by the addition of 10 drops of (KON, NH_2) H. Hel, K+ Fe(ON), and triethanolamine. Ten drops of the indicator were added, and the solution was titrated against EDTA from a red to permanent blue color. The blank was then run in mg standard solution. The mg of EDTA was calculated per milliliter as follows:

$$Mg/MIEDTA = \frac{0.1216 \times 5ml\ mg\ standard\ solution}{Net\ ml\ EDTA\ to\ the\ end\ point}$$

Determination of Ca

Two 5ml aliquots of the extract were placed in two titration flasks, and distilled water was added to get a volume of about 150ml. Ten drops of KON, NH_2OH HCI and triethanolamine and 4ml of 10% NaOH were added. The solution was titrated with EDTA to a purple end point. The pH was raised to 12 to precipitate mg as mg(OH)2. The blank was run with 0.5ml of NH4OH solution, and the net mi of EDTA was calculated by subtracting the titer from that needed for the extract from the calibration of EDTA. Ca was calculated as:

Net ml of EDTA-Net ml for Ca.

Fungal Analysis

Fungi were isolated from the soil samples by spread Inoculating 0.1ml of the soil sample suspension (at 10-and 10-6 dilution) on to Sabroud Dextrose Agar (SDA). The plate was incubated at room temperature (28 ± 2°C) for 72hrs (Oyekeke and Manga, 2008). The colonies were examined both macroscopically and for fungi identification.

Isolated fungi were tested for their ability to grow on used engine oil by inoculating 0,1ml of fungal spore's suspension into test tubes and at room temperature for 21 days. After the period of incubation, the growth of the inoculants was determined by visual observation of the turbidity of the soil medium compared with the inoculated control tubes.

RESULTS

Physicochemical Parameters and Percentage Composition of Various Particles in the Contaminated Soil Sample.

The physiochemical parameters and percentage composition of the various particles in the contaminated soil sampled revealed the presence of moisture (0.87%), pH (5.72), Carbon (4.68%), N (0.062%), P (0.54mg/kg), K (286.50mg/kg), Na (51.25mg/kg), Ca (0.69mg/kg), Mg (0.94mg/kg), sand (96.5), silt (3.3), and clay (0.2), as shown in table 1 below:

Table 1: physicochemical Parameters and Percentage Composition of the Various Particles in the Contaminated Soil Sample.

Moisture %	**The pH**	**Carbon** %	**N** %	**P** mg/kg	**K** Mg/kg	**Na** Mg/kg	**Ca** Mg/kg	**Mg** Mg/kg	**Sand**	**Silt**	**Clay**
0.87	5.72	4.68	0.062	0.54	286.50	51.25	0.69	0.94	96.5	3.3	0.2

Qualitative occurrence of fungal organisms in contaminated soil samples

The qualitative occurrence of fungal organisms in the contaminated soil samples revealed the presence of *Aspergillus flavus, Aspergillus niger, Penicillium sp, Rhizopus stolonifera, Fusarium sp, Trichoderma sp* and *Phoma sp* which is shown in Table 2 below:

Table 2: Qualitative occurrence of fungal organisms in contaminated soil samples

Control	*Aspergillus flavus*	*Aspergillus niger*	*Penicillium sp*	*Rhizopus stolonifera*	*Fusarium sp*	*Trichoderma sp*	*Phoma sp*
+++	++	+++	++	+++	+	+++	+

Effects of Different Levels of Spent Engine oil on Percentage Quantitative Occurrence of Fungal Organism.

The results revealed that 0ml control had the highest percentage quantitative occurrence of fungal organisms while 10ml of the soil sample had the least percentage quantitative occurrence of fungal organisms, as shown in table 3 below:

Table 3: Effect of Different Levels of Spent Engine oil on Percentage Quantitative Occurrence of Fungal Organism.

	Aspergillus flavus	*Aspergillus niger*	*Penicillium sp*	*Rhizopus stolonifera*	*Fusarium sp*	*Trichoderma sp*	*Phoma sp*
0ml/control/20kg	84	89	76	78	46	82	54
2ml/20R of soil sample	56	62	56	60	2.7	67	20
5ml/20R of soil sample	32	37	22	42	-	58	12
10ml/20-k soil sample	5.7	4.6	3	29	-	32	-

DISCUSSION, CONCLUSION, AND RECOMMENDATION

Discussion

The findings of this study on the phytochemical parameters and percentage composition of various particles in contaminated soil samples provide insights into the impact of spent engine oil on soil quality and microflora. The results revealed several critical aspects of soil health, including moisture content (0.87%), pH (5.72), carbon content (4.68%), nitrogen (N) levels (0.062%), phosphorus (P) (0.54 mg/kg), potassium (K) (286.50 mg/kg), sodium (Na) (51.25 mg/kg), calcium (Ca) (0.69 mg/kg), magnesium (Mg) (0.94 mg/kg), sand (96.5%), silt (3.3%), and clay (0.2%). The study also demonstrated that the 0 ml control had the highest percentage occurrence of fungal organisms, whereas the 10 ml of the soil sample had the least. Comparing and contrasting these findings with previous studies provides a comprehensive understanding of the broader implications of spent engine oil on soil microflora.

The soil pH observed in this study (5.72) indicates a moderately acidic condition, which is consistent with the findings of **Nwite *et al.* (2018),** who reported a decrease in soil pH in oil-contaminated soils. This reduction in pH is attributed to the breakdown of hydrocarbon compounds in engine oil, which releases acidic metabolites that lower soil pH. In acidic soils, microbial activity, particularly that of bacteria, is reduced because, these organisms typically thrive in neutral to slightly alkaline conditions. In contrast, **Ekundayo and Obuekwe (2018)** found that pH changes vary based on the degree of contamination and soil type, showing higher acidity in sandy soils compared to loamy or clay soils, which buffer against pH shifts more effectively. The pH level in this study indicates a possible impediment to microbial proliferation, particularly for non-hydrocarbon-degrading microbes.

The observed carbon content (4.68%) indicates an increase in organic matter due to the hydrocarbon content of spent engine oil. This finding is consistent with **Amadi *et al.* (2016)**, who observed elevated carbon levels in oil-polluted soils, attributing it to the accumulation of hydrocarbons. However, while carbon content provides a substrate for hydrocarbon-degrading bacteria, it also leads to an imbalance in nutrient cycling, limiting nitrogen availability for other microbial populations.

The nitrogen, phosphorus, and potassium contents also play essential roles in soil fertility. The nitrogen level (0.062%) in this study was markedly lower than that reported for uncontaminated soils, as noted by **Nwaichi and Okpokwasili (2016)**. This is a direct consequence of the oil contamination, as hydrocarbons inhibit the nitrogen-fixing ability of certain soil bacteria. **Nwaichi and Okpokwasili (2016)** also documented reduced nitrogen levels in oil-polluted soils, which were attributed to the suppression of *Rhizobium* and other nitrogen-fixing bacteria.

The phosphorus levels (0.54 mg/kg) in this study, although present, are lower than those found in uncontaminated soils, a trend also reported by **Adekunle *et al*. (2021)**. The decrease in phosphorous availability can be linked to the sequestration of nutrients by oil residues, which limits plant and microbial uptake. Similarly, potassium levels (286.50 mg/kg) align with findings from **Anoliefo and Ikhajiagbe (2018),** who found that potassium availability decreases in heavily contaminated soils due to the disruption of soil structure and microbial communities.

The fungal population in the study was found to decrease with increasing contamination levels, with the control (0 ml) having the highest fungal occurrence and the 10 ml sample showing the least. This result corroborates the findings of **Ikhajiagbe and Anoliefo (2018),** who demonstrated that spent engine oil contamination significantly reduces fungal populations in soil due to hydrocarbon toxicity. Fungal organisms, particularly saprophytic fungi, play crucial roles in decomposing organic matter. However, in oil-contaminated soils, these organisms are often suppressed by the alteration of soil structure and the presence of toxic hydrocarbons.

In contrast, **Li *et al.* (2021)** noted that hydrocarbon-degrading fungi, such as *Aspergillus* and *Penicillium,* could survive in contaminated soils, albeit with reduced populations. These fungi can metabolize hydrocarbons, contributing to the bioremediation process. This study's finding that fungal populations decrease with higher oil concentrations suggests that most fungal species in the sampled soil are sensitive to hydrocarbons. This conclusion aligns with **Ogbo *et al.* (2018),** who also noted the vulnerability of fungal populations to oil contamination in agricultural soils.

The results indicate that spent engine oil negatively affects the quantitative occurrence of fungal organisms, with the highest fungal diversity observed in the uncontaminated control sample. This finding aligns with **Sharma *et al*. (2019),** who reported that spent oil significantly reduces microbial populations in contaminated soils. The toxicity of spent engine oil affects both the diversity and function of soil microorganisms, leading to the loss of essential soil processes such as organic matter decomposition and nutrient cycling. However, **Adekunle *et al*. (2021)** highlighted the resilience of hydrocarbon-degrading microorganisms, which can persist and thrive in oil-contaminated environments, albeit at reduced population densities.

The suppression of microbial populations also corresponds with the findings of **Anoliefo *et al*. (2019),** who noted that the introduction of hydrocarbons into soil ecosystems leads to a decrease in the abundance of bacteria and fungi, particularly those not involved in hydrocarbon degradation. However, the recovery of microbial populations after remediation efforts, such as bio-augmentation or phytoremediation, has been observed by **Ogbo *et al.* (2018),** suggesting that soil microflora can recover once contaminant levels are reduced.

Conclusion

This study provides evidence that spent engine oil contamination significantly affects the chemical composition and abundance of soil microflora, particularly fungal organisms. The pH, moisture content, carbon, nitrogen,

phosphorus, and potassium levels in the contaminated soil indicate that oil contamination alters the physical and chemical properties of the soil, creating an environment that is inhospitable to many soil organisms. The reduction in fungal populations with increasing contamination suggests that spent engine oil has a suppressive effect on soil fungi, limiting their ability to decompose organic matter and cycle nutrients.

The findings are consistent with previous research, which also pointed out the harmful effects of oil contamination on soil health. However, the persistence of hydrocarbon-degrading microorganisms highlights the potential for bioremediation strategies to mitigate the impact of spent engine oil on soil ecosystems.

Recommendations

Based on the findings of this study, the following recommendations are proposed:

- **Implementation of Bioremediation Techniques**: Given the persistence of hydrocarbon-degrading microorganisms in oil-contaminated soils, bioaugnentation and biostimulation techniques should be employed to enhance hydrocarbon degradation and restore soil health. Introducing native hydrocarbon-degrading bacteria or fungi can accelerate the remediation process.
- **Phytoremediation**: The use of plants, such as *Helianthus annuus* (sunflower), which can tolerate oil-contaminated soils and promote microbial activity in the rhizosphere, should be considered. Phytoremediation, in combination with microbial remediation techniques, may offer a more holistic approach to restoring contaminated soils.
- **Long-term Monitoring**: To fully understand the long-term effects of spent engine oil contamination on soil microflora, further studies should be conducted over extended periods. This will provide insights into the persistence of hydrocarbons and heavy metals in soil and their impacts on microbial populations over time.
- **Policy and Regulation**: Stricter regulations should be implemented to control improper disposal of spent engine oil. Public awareness campaigns should be launched to educate the public about the environmental impact of spent engine oil and the importance of proper disposal practices.
- **Further Research**: There is a need for more research on the effects of spent engine oil on other soil microorganisms, such as protozoa, actinomycetes, and archaea, to gain a broader understanding of how oil contamination affects the entire soil ecosystem.

REFERENCES

Adekunle, A. A., Ogunjobi, A. A., and Omole, T. I. (2021). Bioremediation of petroleum hydrocarbons in contaminated soil using organic and inorganic nutrients. *Environmental Science and Pollution Research,* **28**(3): 2395-2403.

Adewuyi, A., and Okeke, O. C. (2020). Bioremediation of spent engine oil-contaminated soil using cow dung and poultry manure. *Journal of Environmental Protection,* **11**(1): 82-91.

Amadi, A., Dickson, A. A., and Maate, J. K. (2018). Impact of crude oil and refined petroleum spills on natural microbial populations in a Nigerian soil. *International Journal of Environmental Science and Technology,* **15**(3): 569-584.

Amadi, A., Nwankwo, C. D., and Nwokorie, C. I. (2016). Effects of spent engine oil on soil physicochemical properties and microbial populations. *Biotechnology Reports,* **12**(4): 87-94.

Anoliefo, G. O.; Ikhajiagbe, B.; and Onianwa, P. C. (2019). Remediation of oil-contaminated soil using organic and inorganic fertilizers. *Journal of Environmental Management,* **235**(5): 524-532.

Anoliefo, G. O., D. E. Vwioko, and B. Ikhajiagbe, (2019). Environmental and biotechnological aspects of spent oil contamination. *African Journal of Biotechnology,* **18**(19): 521-531.

Ekundayo, F. O., and Obuekwe, C. O. (2018). Hydrocarbon-induced changes in the chemical properties of oil-contaminated soils. *Journal of Environmental Science and Health, Part B,* **53**(2): 85-92.

Ikhajiagbe, B., and Anoliefo, G. O. (2018). Fungal population dynamics in petroleum hydrocarbon-contaminated soils under bio-stimulatory conditions. *Environmental Science and Pollution Research*, **25**(1): 21-30.

Ikhajiagbe, B., and Anoliefo, G. O. (2018). Impact of spent engine oil pollution on the growth and yield of *Phaseolus vulgaris* and its remediation using poultry manure. *Environmental Technology,* **39**(3): 432-441.

Ikhajiagbe, B., and Anoliefo, G. O. (2018). Phytotoxicity of spent lubricating oil on the growth of *Zea mays* (corn). *International Journal of Phytoremediation,* **20**(3): 233-242.

Li, J., Han, Z., Zhang, M., and Wang, Z. (2021). Impacts of petroleum hydrocarbons on soil microbial communities and soil fertility in oil-contaminated areas. *Applied Soil Ecology*, **167**(1): 103901.

Li, Q., Zhang, Z., and Cao, Y. (2021). Effects of petroleum contamination on soil microbial diversity and functional genes. *Journal of Soil Science and Plant Nutrition,* **21**(4): 1764-1775.

Li, Y., Zhang, J., Li, X., Wang, T., and Zhao, F. (2021). Degradation of polycyclic aromatic hydrocarbons in spent engine oil-contaminated soil by fungal communities. *Chemosphere,* **274**(5): 129-139.

Nwaichi, E. O., and Okpokwasili, G. C. (2016). Microbial degradation of oil spill sites in the Niger Delta using nutrient amendments. *International Journal of Environmental Research and Public Health,* **13**(9): 423-432.

Nwaichi, E. O., and Okpokwasili, G. C. (2016). The microbial response to oil pollution and its bioremediation potential. *Environmental Monitoring and Assessment,* **188**(1): 44-53.

Nweke, C. O., and Okpokwasili, G. C. (2020). Microbial population and activity in refinery effluent-polluted soil in the Niger Delta region of Nigeria. *Biotechnology and Bioprocess Engineering,* **16**(5): 784-791.

Nwite, J. N., Ezeaku, P. I., and Enyinnaya, D. I. (2018). Effects of engine oil contamination on soil physicochemical properties and maize performance. *Soil and Environment,* **35**(2): 59-64.

Ogbo, E. M., Okereke, C. V., and Nwagu, T. N. (2018). The microbial population in spent oil-contaminated soil is remediated with organic amendments. *Ecotoxicology and Environmental Safety,* **141**(3): 170-177.

Ogbo, E. M., Okhuoya, J. A., and Efeduma, A. (2018). Effects of spent oil on the growth of *Vigna unguiculata* (cowpea) and *Arachis hypogaea* (groundnut). *International Journal of Environmental Science,* **15**(4): 233-240.

Ogbo, E. M., Tanu, R. B., and Mbah, S. C. (2018). Phytoremediation of oil-contaminated soils using *Helianthus annuus* and microbial analysis of the rhizosphere. *Journal of Environmental Biology,* **39**(6): 1011-1017.

Okoro, J., Emedo, N. U., and Udebuani, A. C. (2019). Biodegradation of spent engine oil by bacteria isolated from oil-contaminated soil. *Journal of Applied Science and Environmental Management,* **23**(6): 1105-1112.

Sharma, P., Kumar, V., and Singh, R. P. (2019). The influence of petroleum hydrocarbons on soil microbiological properties: A review. *Environmental Science and Pollution Research,* **26**(13): 13294-13310.

Sharma, S., Agarwal, N., and Verma, P. (2019). Effects of oil spills on soil microbial communities and bioremediation strategies. *Journal of Environmental Management,* **246**(2): 13-20.

Sharma, S., Ahmed, M., and Garg, A. (2019). Impact of petroleum hydrocarbons on soil microbial diversity and bioremediation potential. *Journal of Environmental Management,* **250**(1): 109501.

Udebuani, A. C., Onwuli, D. O., and Akubugwo, E. I. (2018). Bioremediation of spent engine oil-contaminated soil using poultry manure. *International Journal of Scientific and Research Publications,* **8**(2): 535-543.