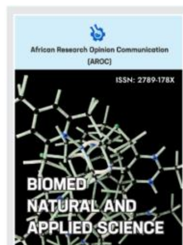
RESEARCH ARTICLE

Bioremediation of soil polluted with spent lubricating oil using bacteria isolated from soil in Abuja Metropolis

Makwin Danladi Makut, Toyosi Michelle Adebayo*, Jibril Egwu Owuna,



[1]Department of Microbiology, Nasarawa State University, P.M.B. 2022, Keffi, Nigeria
Correspondence should be addressed to T.M.A; Email: tma2035@gmail.com

https://doi.org/10.53858/bnas02033340

ABSTRACT

Background: The presence of spent hydrocarbon in soil is a serious problem to the environment hence study on bioremediation of soil polluted with auto-mechanic oil in Abuja Metropolis was carried out. **Methods:** A total of twenty (20) soil samples were collected, bacteria were isolated from the contaminated soil and identified using standard microbiological methods. The spent hydrocarbon utilization was determined using Atomic Adsorption UV Spectrometer. **Results:** The total viable count of the bacteria was 1.07 x 106 from Apo Mechanic village, 1.10 x 106 from Utako Mechanic workshops, 0.40 x 106 from Gwarinpa Mechanic workshops and 2.04 x 106 from Area one Mechanic workshops. The percentage occurrence of bacteria from Apo Mechanic village was Enterobacter species 40.0%, *Pseudomonas synxantha* 60.0%, *Bacillus zanthoxyli* 40.0% and Proteus vulgaris 20.0%. Utako Mechanic workshops were Enterobacter kobei 20.0%, *Pseudomonas synxantha* 40.0%, Bacillus zanthoxyli 20.0% and Proteus vulgaris 40.0%. Gwarinpa Mechanic workshops were *Pseudomonas synxantha* 20.0% and Bacillus zanthoxyli 20.0%. Area one Mechanic workshops were Enterobacter kobei 40.0% and *Pseudomonas synxantha* 40.0%. The effect of days on utilization of spent hydrocarbon showed that *Pseudomonas synxantha* had highest utilized of spent hydrocarbon 19.55mg/ml after 21 days. The effect of pH on utilization of spent hydrocarbon show that at pH 7.5, Enterobacter kobei, Bacillus zanthoxyli and Proteus vulgaris species had the highest utilization of spent hydrocarbon ranging from 5 9.33mg/ml-12.70mg/ml. Effect of temperature on utilization of spent hydrocarbon showed that at 28°C *Enterobacter kobei, Pseudomonas synxantha, Bacillus zanthxyli* and *Proteus vulgaris* had the highest utilization of spent hydrocarbon ranging from 5.51mg/ml- 11.11mg/ml. the bacteria isolated from the contaminated soil have the ability to utilized the hydrocarbon if the soil is amended with some mineral element as shown in this study. **Conclusion:** In conclusion bacteria isolates effectively bioremediated the automechanic oil polluted soil with a reduction of hydrocarbon pollutants.

Keywords: Bioremediation; spent lubricating oi; Abuja Metropolis; auto-mechanic oil

Received: 28 November 2022, revised: 05 December 2022, Published: 12 December 2022

Citation: Makut, M.D., Adebayo, T.M., Owuna, J.E.C(2022). Bioremediation of soil polluted with spent lubricating oil using bacteria isolated from soil in Abuja Metropolis. *BIOMED Natural and Applied Science,* 2(3):33-40, https://doi.org/10.53858/bnas02033340

1.0 Introduction.

Engine oil could simply be defined as a thick mineral liquid applied to a machine or engine to reduce friction between the moving parts of the machine [1]. Oil released into the environment is a well-recognized problem in today's world. Oil spills affect many species of plants and animals in the environment, as well as humans [2]. Spent/Used engine oil is defined as used lubricating oils removed from the crankcase of internal combustion engines [3].

Spent engine oil is a common and toxic environmental contaminant not naturally found in the environment [4], large amount of them are liberated into the environment when the motor oil is changed and disposed into the soil which is a common practice by motor mechanics and generator mechanics including small scale engine oil sellers along the road [5]. Engine oils are altered during use by vehicles, motorbikes, generators and other machinery because of the breakdown of additives, contamination with the products of combustion and the addition of metals from the wear and tear of the engine.

Bioremediation is, therefore, the application of naturally occurring process by which microorganisms transform environmental contaminants into harmless end products [5]. Bioremediation makes use of indigenous oil consuming microorganisms, called petrophiles by enhancing and fertilizing them in their

natural habitats [6]. Petrophiles are very unique organisms that can naturally degrade large hydrocarbons and utilize them as a food source [7]. Microbial remediation of a hydrocarbon contaminated site is accomplished with the help of a diverse group of microorganisms, particularly the indigenous bacteria present in soil. These microorganisms can degrade a wide range of target constituents present in oily sludge [6] there by, can be used in cleaning up contaminated sites.

Many studies have been reported on bioremediation of hydrocarbon pollutants degrading bacteria from auto mechanic workshops' soils but just a few had reported on bioremediation of soil contaminated with used motor oil. Little work has been reported in literature to determine the bioremediation potential of the isolated bacterial consortium by measuring the percentage oil loss and their enzyme activities as a monitoring tool for bioremediation process which is the major focus of this work [7]. Therefore, this study is aimed at bioremediation of soil polluted with auto-mechanic oil using bacteria isolated from soil in Abuja metropolis.

2.0 Materials and Methods

2.1 Chemicals and Reagents

The chemicals and reagents that were used in this study include: England lactophenol cotton blue stain, HCL, NaOH, NH4Cl, KH2PO4, MgSO4,7H2O, ethanol, Xylene solution, Creatinine, Potassium hydroxide and Kovac's reagent, from BDH Chemicals Ltd, England; Ethidium bromide, Iodine solution, EDTA and Glycerol from Sigma chemical Ltd, England; and Agarose gel from Agarose from Schwarz/ Mann Biotech. Nutrient agar and basal medium (minimal salt medium –MSM)

2.2 Study Area

The study area was carried out in Federal Capital Territory Abuja, Nigeria. Abuja is located in the center of the country. Federal Capital Territory is located between latitude 9o4'N of the equator and longitude 7o29'E and situated on an altitude of 360m above sea level.

2.3 Collection of Samples

The study sites were 15 different mechanic workshops situated in different locations in Area One, Utako, Gwarinpa and Apo mechanic village. Apart from visual observation, the attendants at the auto-mechanic workshops were asked questions pertaining to the sites with heavy oil spillage. The sites with the oil spillage had a characteristic black color and the surfaces were hard. They also had no grasses growing on them. Samples were collected at different sites with a shovel to the depth of 0-5cm in triplicates using random sampling technique from each mechanic workshop, augured, and bulked together and put in well labeled black polythene bags and transported to the laboratory for analysis. The samples were subjected to microbiological analyses. Used motor engine oil was also collected from one of the mechanic workshops as the fresh engine oil sample.

2.4 Isolation of bacteria

The isolation of bacteria was carried out using a technique described by Barathi and Vasudvan [9]. The bacteria were isolated using Nutrient agar and basal medium containing 0.3% yeast extract supplemented with 10% V/V oil substrates. The oil substrates were represent equivalent mixture of Gasoline, Engine oil, Diesel oil and spent engine oil. One (1) gram of the hydrocarbon contaminated soil sample was suspension in a test tube containing 9 ml of sterile water to make the soil suspension. Ten-fold serial dilution was made by transferring one ml of the soil suspension to another test tube containing 9 ml of sterile distilled water. These steps were repeated seven times to obtain a dilution of 10-7. From the fourth test tubes, 0.2ml of the aliquot were spread on Nutrient agar and basal medium and incubated at 28oC for 24hours [10].

2.4.1 Total Viable Count

The mean total bacteria present in the samples were determined at weekly interval using spread plate method with Nutrient agar (NA) medium. After tenfold dilution using phosphate buffer dilution saline and 0.1ml of appropriate dilution was plated in triplicates, to be incubated for 24-48 hours at room temperature after which the colonies were counted [11].

2.4.2 Identification of Bacteria isolated from contamination with hydrocarbon soil

Colonies was chosen from each of the cultured plate on the bases of their colonial and morphological similarities. Pure bacterial colonies was identified using Gram staining reaction and biochemical tests according to the methods Cheesbrough.

2.5 Molecular Identification

2.5.1 DNA extraction

Five milliliters of an overnight culture of the bacterial isolate in Luria Bertani (LB) broth were spun at 14000rpm for 3 min. The cells were re-suspended in 500µl of normal saline and heated at 950C for 20 min. The heated bacterial suspension was cooled on ice and spun for 3 min at 14000rpm. The supernatant containing the DNA was transferred to a 1.5ml microcentrifuge tube and stored at -20oC for other downstream reactions.

2.5.2 DNA quantification

The extracted genomic DNA was quantified using the Nanodrop 1000 spectrophotometer. The software of the equipment was lunched by double clicking on the Nanodrop icon. The equipment was initialized with 2µl of sterile distilled water and blanked using normal saline. Two microliters of the extracted DNA were loaded onto the lower pedestal, the upper pedestal was brought down to contact the extracted DNA on the lower pedestal.

2.5.3 16S rRNA Amplification

The 16s rRNA region of the rRNA genes of the isolates were amplified using the 27F: 5'-AGAGTTTGATCMTGGCTCAG-3' and 1492R: 5'-CGGTTACCTTGTTACGACTT-3' primers on ABI 9700 Applied Biosystems thermal cycler at a final volume of 25 microlitres for 35 cycles. The PCR mix included: the X2 Dream taq Master mix supplied by Inqaba, South Africa (Taq polymerase, DNTPs, MgCl), the primers at a concentration of 0.4M and the extracted DNA as template. The PCR conditions were as follows: Initial denaturation, 95°C for 5 minutes; denaturation, 95°C for 30 seconds; annealing, 52°C for 30 seconds; extension, 72ºC for 30 seconds for 35 cycles and final extension, 72°C for 5 minutes [12].

2.5.4 Agarose Gel Electrophoresis of the 16SrRNA gene from the bacteria isolates

The agarose gel electrophoretic separation of digested 16S rRNA gene was carried out as described previously, 8µl of PCR products stained with ethidium bromide was loaded into 1.0% (wt/vol) agarose gel wells with a molecular marker run concurrently at 120 V for 30 min. The DNA bands were visualized and photographed under UV light 595nm.

2.6 Effect of pH on utilization of spent hydrocarbon substrate from contaminated soil

The effect of pH was carried out following a method described by Makut and Majekodunmi, [13]. One hundred (100) ml of the Basal medium substrate amended was transfered into different conical flasks. The pH ranges was adjusted to, 4.5, 5.5 6.5 and 7.5 for degradation media using 1.0 N HCl to adjusting the pH of the media before autoclaving.

2.7 Effect of Temperature on utilization of spent hydrocarbon substrate from contaminated soil

Effect of temperatures was carried out following a method described by previous study. One hundred (100) ml of the degradation media was transfer into different conical flasks and the degradation media was incubated at 28oC, 32oC, 35oC and 38oC

2.8 Total utilization of spent hydrocarbon

Total utilized spent hydrocarbon content of the degradation media samples were determined using UV Visible spectrophotometer methods according to the toluene extraction method Makut and Majekodunmi, [13]. Fifteen miligram (15ml) of each of the sample was transfer into a test tube, and then 1ml of 60ug/ml of 1-chlorooctadecane surrogate standard was added. Then 30 milliliters of dichloromethane (extraction solvent) was added to extract oil in the soil. After shaking vigorously in

 https://doi.org/10.53858/bnas0203340
ISSN: 2789-178X

water bath for 5hrs, the mixture were allowed to stand for 60 minutes and then filtered through Whatman No.1 filter paper fitted with cotton wool and sodium sulphate into a clean beaker washed with methylene chloride. Residue were then washed with 20ml extracting solvent and then filtered through funnel. The extracted oil will be transferred to vial and placed on a UV Visible spectrophotometer chamber for analysis. The amount of waste lubricating oil degraded was calculated by subtracting the weight of residual waste lubricating oil from weight of the initial waste lubricating oil, divided by the weight of the initial waste lubricating oil and then multiplied by 100.

$$HR\ (\%) = \frac{(\text{Weight of oil before remediation}) - (\text{Weight of oil after remediation}) \times 100}{\text{Weight of oil before remediation}}$$

Where HR = Hydrocarbon utilization

3.0 Results

3.1 Total mean viable counts

The total mean viable count of bacteria isolated from contaminated soil of hydrocarbon in Mechanic workshops is as given in Table1. The total viable count of the bacteria was 1.07 x 10^6 from Apo Mechanic village, 1.10 x 10^6 from Utako Mechanic workshops, 0.40 x 10^6 Gwarinpa Mechanic workshops and 2.04 x 10^6 from Area one Mechanic workshops respectively.

Table 1: Total mean viable count of bacteria isolated from contaminated soil of hydrocarbon in Mechanic workshops

Sample location	Total viable count (cfu/g) x10^6
Apo Mechanic village	1.07 x 10^6
Utako	1.10 x 10^6
Gwarinpa	0.40 x 10^6
Area one	2.04 x 10^6

Table 2: Cultural, Morphology and Biochemical characteristics of Bacterial Isolates

Cultural	Grams	Cat	Nit	Coa	Gel	Ure	Glu	Suc	Inference
Brown on MaC and Greenish Colonies on NA	-	+	+	-	+	-	+	-	*Pseudomonas synxantha*
Purple on MaC and yellowish on NA	-	+	+	-	+	-	+	+	*Enterobacter kobei*
Milkfish and dried on NA	+	+	-	-	-	+	-	-	*Bacillus zanthoxyli*
Mucoid and brownish on NA	-	-	+	-	-	-	+	-	*Proteus vulgaris*

KEY: Cat-catalase, Nit- nitrate, Coa-coagulase, Gel-gelatin, Ure- urea, Glu- glucose, Suc-Sucurcose

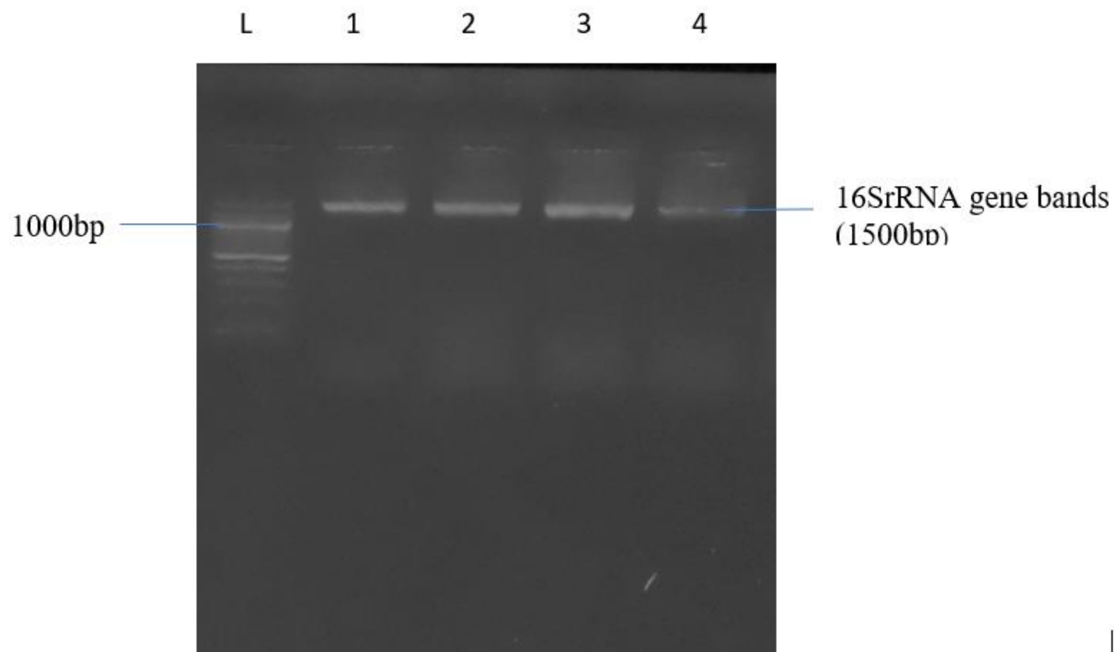


Plate 1: Agarose gel electrophoresis of the 16SrRNA gene of bacteria isolates. L represents the molecular ladder, Lane 1 = *Enterobacter kobei* strain DSM 13645, Lane 2 = *Pseudomonas synxantha*,.strain KENGFT3, Lane 3 = *Bacillus zanthoxyli*, Lane 4 = *Proteus vulgaris* strain FDAARGOS_556.

3.2 The cultural, morphology and biochemical characteristics of the bacteria isolate

The cultural, morphology and biochemical characteristics are as shown in Table 2. The occurrence of bacteria isolated from contaminated soil of hydrocarbon in mechanic workshops in Abuja is as given in Table 3. Out of 5 samples collected from each location. Location A; *Enterobacter kobei* had 40.0%, *Pseudomonas synxantha* 60.0%, *Bacillus zanthoxyli* 40.0% and *Proteus vulgaris* 20.0%. From location B; *Enterobacter kobei* had 20.0%, *Pseudomonas synxantha* 40.0%, *Bacillus zanthoxyli* 20.0% and *Proteus vulgaris* 40.0%. From location C; *Pseudomonas* sp had 20.0%, *Bacillus zanthoxyli* 20.0% and *Proteus vulgaris* 0.00%. From location D; *Enterobacter kobei* had 40.0% and *Pseudomonas synxantha* 40.0%.

Table 3: Occurrence of Bacteria Isolated from contaminated soil of hydrocarbon in mechanic workshops in Abuja in percentages

Bacterial	No samples	A	B	C	D
Enterobacter kobei	5	2(40.0)	1(20.0)	0(00)	2(40.0)
Pseudomonas synxantha	5	3(60.0)	2(40.0)	1(20.0)	2(40.0)
Bacillus zanthoxyli	5	2(40.0)	1(20.0)	1(20.0)	0(00)
Proteus vulgaris	5	1(20.0)	2(40.0)	0(00)	0(00)

Key: location A= Apo Mechanic village, B= Utako, C= Gwarinpa, D= Area one. Data are mean ± SEM of triplicate determination

3.3 Effect of time on microbial on utilization of spent oil

The effect of days on utilization of spent oil is as given in Table 4. After 7days *Enterobacter kobei* were able to utilized 2.20mg/ml of the spent oil, after 14 day it had 5.70mg/ml utilization, 21days it utilized 9.90mg/ml and after 30days it had 5.70mg/ml utilization from the initial concentration of the spent oil. *Pseudomonas synxantha* had 5.10mg/ml utilization after 7days, 9.21mg/ml after 14days, 19.55mg/ml after 21days and 15.41mg/ml after 30 days. *Bacillus zanthoxyli* had 4.11mg/ml after 7days, 7.40mg/ml after 14days, 14.55mg/ml after 21days and 12.01mg/ml after 30days and *Proteus vulgaris* had 1.21mg/ml after 7days, 3.22mg/ml after14days, 11. 00 mg/ml after 21days and 6. 20 mg/ml after 30days. The control showed no reduction.

Table 4 Effect of days on utilization of spent hydrocarbon by bacteria isolated from soil in mechanic workshops

	Concentration (mg/ml) at different days				
Isolates	**Initial**	**7**	**14**	**21**	**30**
Enterobacter kobei	26.85± 0.55	2.20 ± 0.19	5.70±0.28	14.10 ± 0.10	9.90 ± 0.20
Pseudomonas synxantha	26.85± 0.55	5.10±0.18	9.21±1.28	19.55±1.28	15.41±0.48
Bacillus zanthoxyli	26.85± 0.55	4.11±0.08	7.40±2.08	14.55±1.01	12.01±0.66
Proteus vulgaris	26.85± 0.55	1.21±0.05	3.22±0.54	11.00±0.16	6.20±1.28
Control	26.85± 0.55	0.00± 0.00	0.00±0.00	0.00±0.00	0.00±0.00

Data are mean ± SEM of triplicate determination

3.4 Effect of pH on microbial on utilization of spent oil

The effect of pH on utilization of spent oil by bacteria isolated from mechanic workshop is as shown in Table 5. *Enterobacter kobei* showed utilization ability of 2.10mg/ml at pH 4.5, 3.90mg/ml at pH5.5, 5.22mg/ml at pH6.5 and 12.70mg/ml at pH7.5. *Pseudomonas synxantha* had 4.15mg/ml at pH4.5, 5.31mg/ml at pH 5.5, 14.11mg/ml at pH6.5 and 8.11mg/ml at pH7.5. *Bacillus zanthoxyli* utilized 4.05mg/ml at pH4.5, 4.11mg/ml at pH5.5, 6.51mg/ml at pH6.5 and 9.33mg/ml at pH7.5 and *Proteus vulgaris* had 2.20mg/ml at pH4.5, 3.80mg/ml pH5.5, and 4.01 mg/ml at pH6.5 and 7.52mg/ml at pH7.5 respectively

Table 5: Effect of pH on utilization of spent hydrocarbon by bacteria isolated from soil in mechanic workshops

	Concentration (mg/ml) at different pH				
Isolates	Initial	4.5	5.5	6.5	7.5
Enterobacter kobei	26.85± 0.55	2.10 ± 0.11	3.90 ± 0.20	5.22 ± 0.09	12.70±1.48
Pseudomonas synxantha	26.85± 0.55	4.15±1.08	5.31±0.28	14.11±0.18	8.11±0.18
Bacillus zanthoxyli	26.85± 0.55	4.05±0.01	4.11±0.16	6.51±0.32	9.33±2.08
Proteus vulgaris	26.85± 0.55	2.20±0.10	3.80±0.51	4.01±0.25	7.52±0.14
Control	26.85± 0.55	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00

Data are mean ± SEM of triplicate determination

3.5 Effect of temperature on microbial on utilization of spent oil

Table 6 shows the effect of temperature on spent hydrocarbon utilization by some bacteria isolated from mechanic workshops in Abuja. At temperature of 28°C *Enterobacter kobei* utilized 10.12mg/ml of the

https://doi.org/10.53858/bnas0203340
ISSN: 2789-178X

initial concentration of 23.85mg/l of the spent hydrocarbon, at 32°C it utilized 6.10mg/ml, 35°C it utilized 4.30mg/ml and at 38 ^{O}C it had 4.01mg/ml. *Pseudomonas synxantha* utilized 11.11mg/ml at 28 ^{O}C, 7.08mg/ml at 32 ^{O}C, 5.61mg/ml at 35 ^{O}C and 4.15mg/ml at 38 ^{O}C. *Bacillus zanthoxyli* was able to utilized 9.21mg/ml at 28 ^{O}C, 6.63mg/ml at 32 ^{O}C, 4.88mg/ml at 35 ^{O}C and 3.05mg/ml at 38 ^{O}C while *Proteus vulgaris* 5.51mg/ml at 28 ^{O}C, 5.00mg/ml at 32 ^{O}C, 3.00mg/ml at 35 ^{O}C and 2.21mg/ml at 38 ^{O}C respectively.

Table 6: Effect of temperature on utilization of spent hydrocarbon by bacteria isolated from soil in mechanic workshops

	Concentration (mg/ml) at varying temperature				
Isolates	Initial	28°C	32°C	35°C	38°C
Enterobacter kobei	26.85± 0.55	10.12 ± 1.19	6.10±0.18	4.30 ± 0.06	4.01 ± 0.31
Pseudomonas synxantha	26.85± 0.55	11.11±1.11	7.08±0.68	5.61±0.24	4.15±1..18
Bacillus zanthoxyli	26.85± 0.55	9.21±0.02	6.63±0.08	4.88±1.12	3.05±0.21
Proteus vulgaris	26.85± 0.55	5.51±0.25	5.00±0.14	3.00±0.01	2.21±0.50
Control	26.85± 0.55	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00

Data are mean ± SEM of triplicate determination

4.0 Discussions

The indiscriminate disposal of waste oil effects the environment and reduction in oxygen supply to microorganisms [14]. Bioremediation process is a microorganism mediated transformation or degradation of contaminants into non-hazardous or less hazardous substances [14,15]. It is an attractive approach for cleaning up of hydrocarbons from environment because it is a simple technique, easy to maintain [16], applicable over large areas, cost effective and leads to complete destruction of the contaminant. Many physical, chemical, and environmental factors like temperature soil moisture, soil acidity and alkalinity etc affects the process of biodegradation of hydrocarbons [17]. Bacteria and fungi are known to be the principal agents of biodegradation of hydrocarbons. Fungi have a higher tolerance to the toxicity of hydrocarbons due to their physiology and adaptation to such variations in the environment and have the mechanism for the elimination of spilled oil from the environment.

In this study the total viable count observed was high but lower than the one reported by Bada *et al. [18]* and similar to the study reported by Gallego *et al. (*2010), The reasons for the significant low counts of bacteria in the contaminated soil may be due to the non-availability of appreciable quantities of nitrogen, phosphorus and oxygen in the contaminated soil which enhances the reduction of bacteria population.

The bacteria isolated from different mechanic workshops contaminated with spent hydrocarbon were *Enterobacter kobei*, *Pseudomonas synxantha*, *Bacillus zanthoxyli* and *Proteus vulgaris* this is in agreement with the study reported by Onuoha *et al*. [19]. This means that these bacteria exhibited responses and potential to breakdown petroleum hydrocarbon and utilize it as source of energy and carbon in their natural ability. These bacteria species has been implicated in the hydrocarbon degradation by the authors [19]. It has also been observed that some bacteria are more abundant in areas of high concentration of hydrocarbons. These bacteria are actively oxidizing the hydrocarbons, and this is considered as another source of carbon for use in the ecosystem Bada *et al. [18]*

The effect of days on utilization of spent hydrocarbon as observed in this study was indifferent from studies reported by Olubunm et al. [20] who reported high utilization of spent oil at 28days but in this study it was observed that after 28days the amount of the reduction or utilization by the bacteria dropped suggesting decline in the population of the bacteria. Also it was observed that pH plays important role in the degradation or utilization of spent oil by different bacteria as observed in this study which is similar to studies reported by different authors such as Bada *et al. [18]*; Akpoveta *et al*., [21] and Yakubu (2007) who reported high utilization of spent oil at pH of 6.5 and 7.5 which is in agreement with our findings in this study. Soil nutrient decreased after contamination with petroleum products. Akpoveta *et al*. [21] observed significant reduction in pH and available nutrients in soil on simulation of soil with kerosene. Increase in soil acidity as a result of petroleum hydrocarbon in soil is a concern for agricultural soil because low pH values are associated with adverse soil condition which includes reduced availability of nutrients, reduced microbial activity. The effect of temperature on utilization of spent oil from different mechanic workshop as observed in this study showed that high temperature affects the utilization of hydrocarbon. It was observed that at temperature of 28°C were was high utilization of spent hydrocarbon than other temperature tested in-vitro and this is similar to study reported by Schafer *et al.* [22] who reported decrease in population of bacteria in soil and water due to high temperature in area pollution or contaminated with hydrocarbon.

https://doi.org/10.53858/bnas0203340
ISSN: 2789-178X

5.0 Conclusion

From this study it was observed that different bacteria namely *Pseudomonas synxantha*, *Bacillus zanthoxyli*, *Proteus vulgaris* and *Enterobacter kobei* isolated from soil contaminated with in Mechanic workshops were able to show various ability to utilize spent hydrocarbon in varying amounts. This study however, recommended that further studies on the use of bacteria isolates from polluted soils should be carried out to enhance their potential in hydrocarbon utilization

Conflict of Interest: The author declared no conflict of interest exist

Funding: No funding was received

Authors contribution: This work was conducted with the collaboration of all authors: Author TMA conducted the research and wrote the manuscript. Author MDM supervised the work. Author JEO CO-supervised the work. All authors read and approved the final version of the manuscript

Acknowledgement: We acknowledged all the lecturers and the technical staff of the Department of Microbiology, Nasarawa State University

References

1. Akhavan S., Dejban A., Golpasha I., Emami M and Nakhoda, A. M. (2008). Isolation and Characterization of Crude Oil Degrading Bacillus spp. *Iranian Journal of Environmental Health Science and Engineering*.5 (3) 149 –154
2. Nwite, J. N and Alu, M. O. (2015). Effects of different levels of spent engine oil on soil properties, grain yield of maize and its heavy metal uptake in Abakaliki, South Eastern, Nigeria. *Journal of Soil Science and Environmental Management* 5 (4): 44-51
3. Okonokhua, B. O.; Ikhajiagbe, B.; Anoliefo, G. O. and Emede, J. O. (2017). The effect of spent engine oil on soil properties and growth of maize (Zea may L). *Journal of Applied Sciences Environmental Management*. 11 (3): 147 – 152.
4. Dominguez-Rosado R. E and Pichtel D. (2009). Phytoremediation of contaminated with used motor oil. 1. Enhanced microbial activities from laboratory and growth chamber studies. *Environmental* Engineering Science, 2, 157-168
5. Abdulsalam S and Omale A.B (2015) Comparison of biostimulation and bioaugmentation techniques for the remediation of used motor oil contaminated soil, Braz Arch Biol Technol 52: 747-754.
6. Abdulsalam, S., Adefila, S. S., Bugaje, I. M and Ibrahim, S. C. (2012). Bioremediation of soil contaminated with used motor oil in a closed system. *Journal of Bioremediation and Biodegradation* 3(12): 1-7
7. Al-Majed A.A., Adebayo A.R and Hossain M.E. A (2012) sustainable approach to controlling oil spills. *Journal of Applied Sciences Environmental Management*, 113: 213-227
8. Tyagi M, Da Fonseca MMR, De Carvalho CCCR (2011). Bioaugmentation and biostimulation strategies to improve the effectiveness of bioremediation processes. Biodegradation, 22: 231-241
9. Barathi S and Vasudevan N (2010). "Utilization of petroleum hydrocarbons by Pseudomonas fluorescens isolated from a petroleum-contaminated soil," Environment International, 26: (5-6) 413–416
10. Barathi S and Vasudvan N (2016). Utilization of petroleum hydrocarbons by *Pseudomonas florescens* isolated from a petroleum contaminated soil. *Environmental International* 26: 413 -416.
11. Joutey N.T., Bahafid W., Sayel H and Ghachtouli N.E (2013) Biodegradation: Involved microorganisms and genetically engineered microorganisms: Biodegradation-Life of science. Croatia. 289-320
12. Makut, M. D., & Ibrahim, M. Z. (2021). Molecular and phylogenic identifications of potential herbicide degrading microorganisms from contaminated farmland in Keffi, Nasarawa State, Nigeria. *AROC in Pharmaceutical and Biotechnology, 01 (01); 17*, *25*(1.0).
13. Makut, D. M., & Majekodunmi, O. O. (2019). Bioremediation potentials of chicken droppings on crude oil polluted soil from automobile workshop. *GSC Biological and Pharmaceutical Sciences*, *8*(2), 045-052.

14. Makut, M. D., & Ibrahim, M. Z. (2021). Molecular and phylogenic identifications of potential herbicide degrading microorganisms from contaminated farmland in Keffi, Nasarawa State, Nigeria. *AROC in Pharmaceutical and Biotechnology, 01 (01); 17*, *25*(1.0).
15. Alabi, O. H., Obiekezie, S. O., & Ekeleme, K. (2022). Antibiotic resistance profiling and molecular detection of Escherichia coli Pathotypes isolated from irrigated fresh vegetables. *AROC in Pharmaceutical and Biotechnology*, *2*(1), 18-26.
16. Nwankwegu, A. S., Orji, M. U., Onwosi, C. O. (2016). Studies on organic and in-organic biostimulants in bioremediation of diesel-contaminated arable soil. Chemosphere. 162: 148–156.
17. Achal, V., D. Kumari, and. Pan, X. (2011).Bioremediation of chromium contaminated soil by a brown-rot fungus, Gloeophyllumsepiarium. Research Journal of Microbiology. 6:166–171.
18. Bada B. S, Tsobaza I. E., Toyin A. A and Tolulope M. O (2019), Degradation of total petroleum hydrocarbon in petroleum productscontaminated soil using pig dung**,** *West African Journal of Applied Ecology*, 27(2):1-15
19. Onuoha, S.C., Olugbue, V.U., Uraku, J.A and Uchendu, D.O (2011). Biodegradation potentials of hydrocarbon degraders from waste –lubricating oil spilled soils in Ebonyi State Nigeria. *International Journal of Agriculture andBiology,* 13:586-590
20. Olubunm F., Olatunde S., Abiodun A and Adetoyi H (2019). Degradation of spent oil contaminated soil using fungi from organic manure. *International Journal of Research and Developmen*, 4 (10) 36-44
21. Akpoveta, O. V., Egharevba, F., Medjor, O. W., Osaro, K. I. and Enyemike, E. D. (2011). Microbial degradation and its kinetics on crude oil polluted soil. *Resource Journal of Chemical Scences* **1(6)**, 8—14
22. Schafer, A. N., I. Snape and S. D. Siciliano**.** (2009). Influence of Liquid Water and Soil Temperature on Petroleum Hydrocarbon Toxicity in Antarctic Soil. *Environmental Toxicology and Chemistry*, **28**:1409-1415

Submit your article to AROC JOURNALS
-AROC in Pharmaceutical and Biotechnology
-AROC in Agriculture
-AROC in Bioinformatics
-AROC in Food and Nutrition
-AROC in Natural Product Research
-BIOMED Natural and Applied Science
Via https://arocjournal.com/

Copyright © 2022 Makut et al. This is an open access article distributed under the terms and conditions of the Creative Commons Attribution License (CC BY) which allowed unrestricted download, distribution and reused as long as the original authors are properly cited.

 https://doi.org/10.53858/bnas0203340
ISSN: 2789-178X