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# A Comparative Assessment of Pig Manure and Poultry Manure in the Biodegradation of Diesel Contaminated Soil

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## Authors' contributions

This work was carried out in collaboration among all authors. Author AI designed the study and wrote the protocol. Author NN performed the statistical analysis and managed the analyses of the study. Author NBO wrote the first draft of the manuscript and managed the literature searches. All authors read and approved the final manuscript.

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## ABSTRACT

**Aim:** This research aims to carry out a comparative assessment on the use of pig manure and poultry manure as a carbon substrate for the degradation of diesel.

**Study Design:** Fifteen experimental pots were used in this analysis and this was carried out for a duration of 44 days. This experiment was done in stages and each stage was for 2 weeks. Experimental pots were labelled appropriately with the right concentrations of soil, diesel oil and manure to avoid cross-contamination. This experiment involved one pollution level of 0.275% of the weight of soil. Two (2) treatment concentrations of 20% and 40% of the weight of soil for pig manure and poultry manure respectively, three temperature levels of 25°C, 30°C, 37°C.

**Place and Duration of Study:** Microbiological laboratory and Environmental Science laboratory of Coventry University/two months.

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**Methodology:** Bioremediation of the diesel contaminated soil involved two concentration levels of 40 g and 80 g. These concentrations (40 g and 80 g) were used for both pig manure and poultry manure. These manures were measured in the fume cupboard using an electronic balance to avoid pollution to air. The right manure concentrations of 40 g and 80 g were transferred into the appropriately labelled pots containing 200 g of soil each. The mixture (soil+ diesel+ pig/poultry manure) was properly homogenised and allowed for biodegradation.

**Results:** Results of TPH analysis showed high percent removal of 84.71%, 90%, 82.35%, 85.29% for soil treated with pig manure at 40 g and 80 g, and soil treated with 40 g, 80 g of poultry manure at 37°C. This study showed that the microbial consortium, nutrient concentration and temperatures played great roles in enhancing bioremediation process.

**Conclusion:** Nutrient addition enhanced the degradation of diesel contaminated soil. It is evident from the results that at 37°C diesel degradation occurred more in all soil samples than at 30°C and 25°C. Therefore, it can be concluded that 37°C is most suitable for diesel degradation with the highest efficiency in soil treated with 80 g of pig manure. However, at 25°C, high percent degradation also occurred in all treated samples with spi-40 g having the highest percent degradation.

*Keywords: Biodegradation; pig manure; poultry manure; bioremediation; diesel.*

## 1. INTRODUCTION

Petroleum based products are major sources of energy in many developed and fast developing economies [1]. In recent decades, however, they have gained notoriety for their contribution to soil and water pollution [2]. Exposure to high concentrations via oil spills, leakages from pipelines, improper waste disposal and management of petroleum wastes and by-products harms the ecosystem and poses health risks [3].

Crude oil is a complex chemical mixture of saturated hydrocarbons, aromatic hydrocarbons, resins and asphaltenes. This causes hydrocarbons to differ in their sensitivity to microbial attack which can be classified as Linear Alkanes > Branched Alkanes > Small Aromatics > Cyclic Alkanes [4]. Hence, the application of microbial consortium to deal with the challenge of metabolising the components of a petroleum mixture [2].

Microorganisms mainly involved in hydrocarbon degradation are; bacteria, yeast and fungi with remediation efficiency of 6% to 82% for soil fungi, 0.13% to 50% for soil bacteria and 0.003% to 100% for marine bacteria [1]. Furthermore, to enhance the efficiency of the bioremediation process, nutrients and inoculants (indigenous or non-indigenous) are added to stimulate the growth of hydrocarbon-degrading microorganisms [5].

Bioaugmentation and/or Biostimulation are methods mainly used in bioremediation. The

process of biostimulation introduces additional nutrients in the form of organic and/or inorganic fertilizers into a contaminated system. A study on the use of fertilizer in enhancing oil biodegradation by naturally occurring microbes showed that fertilizer use has the potential to remove hydrocarbons in a contaminated area [6]. This process has been effective in enhancing microbial activity by supplementing the number of nutrients available in the soil [7]. However, factors such as the composition of inherent biodegradability of petroleum hydrocarbon, temperature, microbial flora and nutrient availability can limit biodegradation. Pig manure and poultry manure have been screened for crude oil-degrading capabilities. Results of the analysis showed that pig manure contained various degrees of crude oil utilizing bacteria in addition to being rich in mineral element [8]. Poultry manure also contained nutritive elements that improve microbial activity for petroleum hydrocarbon degradation [9]. Moreover, pig manure will be further assessed in this research to determine its effectiveness in diesel degradation as compared with poultry manure.

## 2. MATERIALS AND METHODS

### 2.1 Samples Test Materials

This experiment was carried out in the Analytical laboratory, Microbiological laboratory and Environmental Science laboratory of Coventry University. The following materials were used by the researcher to ensure proper preparation of soil samples for analysis; N-hexane,

Thermometer, Rotary evaporator, Soxhlet apparatus, Beakers (50 ml, 100 ml), Graduated cylinders, Electronic balance, Pipettes (200 µl, 1000 µl, 5000 µl), Hach Lange 28800 spectrophotometer, Anhydrous sulphate, Weighing boats, Thimbles, Anti -bumping granules, pH meter, Poultry manure, Pig manure, Conical flask (500 ml), Round bottom flask (250 ml), Whatman filter paper (1-2 mm), Funnels, Water, Distilled water, Diesel (825 g), Spatula, 1/2-pint disposable cups, Ziploc bags, Trays, Experimental pots, Water bath, Orbital shaker, Stearic acid, Hexadecane.

## 2.2 Soil Characterization

Soil sample sieved through a 2 mm mesh size was dried in an oven in order to determine its actual weight. Using an electronic weighing balance, soil samples were measured. Also, the physicochemical parameters of the soil and manure such as soil pH, soil moisture content, soil nutrient content, manure nutrient content were determined. Manure agitated for 2-4 hours before the application has a high concentration of NPK.

## 2.3 Sample Preparation and Pre-treatment

Undesirable substances in soil sample such as sticks and stones were removed and the soil was homogenized using a spatula. Soil was sieved with a 2 mm sieve to obtain fine granules. Sample pre-treatment was done to increase the homogeneity of the soil, increase the extraction of analyte in the soil. This experiment was done in stages.

## 2.4 Soil Measurement

200 g of loamy soil was measured using an electronic weighing balance and transferred into five (5) different experimental pots of 15.5 cm diameter each.

## 2.5 Experimental Design

Fifteen experimental pots were used in this analysis, and this was carried out for a duration of 44 days. This experiment was done in stages, and each stage was for 2 weeks.

Experimental pots were labelled appropriately with the right concentrations of soil, diesel oil and manure to avoid cross-contamination. This

experiment involved one pollution level of 0.275% of the weight of soil. Two (2) treatment concentrations of 20% and 40% of the weight of soil for pig manure and poultry manure respectively, three temperature levels of 25°C, 30°C, 37°C. the experimental layout is shown in Table 1.

## 2.6 Pollution of Soil and Spiking with Manure

Prior to soil pollution, diesel was purchased from Muroco filling station Coventry, United Kingdom. Soil samples used in this experiment were polluted with just one concentration of diesel (0.275%). The total quantity diesel used in each stage of the experiment was 275 g (27.5%).

The measured diesel oil (275 g) was then transferred into a large mixing tray containing 1500 g of soil. This was then mixed using a spatula and then with hands to ensure homogeneity. This mixture (soil + diesel) was transferred to two (2) experimental pots allowed to concentrate for 2 days (48 hours). Before amendment with poultry and pig manure, the soil was still homogenized and 200 g each of mixture (soil+ diesel) was transferred into 5 experimental pots. Soil samples were collected for TPH analysis as described by EPA method 9071B. This was done to determine the initial concentration of TPH in the diesel contaminated soil before treatment.

## 2.7 Remediation Stages

Bioremediation of the diesel contaminated soil involved two concentration levels of 40 g and 80 g. These concentrations (40 g and 80 g) were used for both pig manure and poultry manure. The organic manures used were collected from animal farms in the United Kingdom. Poultry manure was collected from Seven acres poultry farm Coventry while Pig manure was collected from Midland pig producers farm Stafford. These manures were measured in the fume cupboard using an electronic balance to avoid pollution to air. The right manure concentrations of 40 g and 80 g were transferred into the appropriately labelled pots containing 200 g of soil each. The mixture (soil+ diesel+ pig/poultry manure) was properly homogenised and allowed for biodegradation.

### 2.7.1 Remediation set up at 25°C

The first stage of this analysis was done in water bath at 25°C for 2 weeks. The bath was filled

**Table 1. Soil treatments and codes**

S/N	Treatment	Code
1	Control1 (soil + diesel at 25°C)	C1-25A, C1-25B
2	Control 2 (soil + diesel at 30°C)	C2-30A, C2-30B
3	Control 3 (soil +diesel at 37°C)	C3- 37A, C3-37B
4	Soil+diesel+40g pig manure at 25°C	SPI-40-25A, SPI-40-25B
5	Soil + diesel +80g pig manure at 25°C	SPI-80-25A, SPI-80-25B
6	Soil+diesel+40g poultry manure at 25°C	SPO-40-25A, SPO-40-25B
7	Soil+diesel+80g poultry manure at 25°C	SPO-80-25A, SPO-80-25B
8	Soil+diesel+40g pig manure at 30°C	SPI-40-30A, SPI-40-30B
9	Soil+diesel+80g pig manure at 30°C	SPI-80-30A, SPI-80-30B
10	Soil+diesel+40g poultry manure at 30°C	SPO-40-30A, SPO-40-30B
11	Soil+diesel+80g poultry manure at 30°C	SPO-80-30A, SPO-80-30B
12	Soil + diesel +40g pig manure at 37°C	SPI-40-35A, SPI-40-35B
13	Soil + diesel +80g pig manure at 37°C	SPI-80-35A, SPI-80-35B
14	Soil+diesel+40g poultry manure at 37°C	SPO-40-30A, SPO-40-30B
15	Soil+diesel+80g poultry manure at 37°C	SPO-80-35A, SPO-80-35B

with water to the marked point and temperature adjusted to 25°C. Prepared soil samples were transferred from experimental pots after 3days into five (5) 500 ml conical flasks and labelled. 200 ml of distilled water was added into each conical flask and stirred with a spatula. This was to allow for proper agitation. The conical flasks were then placed in the water bath at 25°C. The linear motion was adjusted to 136 strokes per minute. This was to reduce water spillage. The temperature was confirmed using a thermometer to be at the range of 25.0°C – 25.2°C.

### 2.7.2 Remediation set up at 30°C

The second stage of this analysis was done in a similar water bath at 30°C for 2 weeks. The bath was filled with water to the marked point and temperature adjusted to 30°C. Prepared soil samples were transferred from experimental pots after 2 days into five (5) 500 ml conical flasks and labelled. 200 ml of distilled water was added into each conical flask and stirred. This was to allow for proper agitation. The conical flasks were then placed in the water bath at 30°C. The

linear motion was adjusted to 136 strokes per minutes. This was to reduce water spillage. The temperature was confirmed using a thermometer.

### 2.7.3 Remediation set up at 37°C

Prepared soil samples were transferred from experimental pots after 3 days into 500 ml conical flasks and labelled. 200 ml of distilled water was added into each conical flask and stirred with a spatula. This was to allow for proper agitation. The conical flasks were then placed on the orbital shaker at 37°C. The third stage of this experiment was for two weeks.

## 2.8 Analytical Methods

Two analytical methods were used in this experiment. They are the USEPA method 9071B for TPH Extraction and Hach Lange spectrophotometric method for nutrient determination.

## 3. RESULTS AND DISCUSSION

### 3.1 Effect of Remediation Technique on Nutrient Concentration

The chemical properties of the contaminated soil samples analysed using Hach Lange spectrophotometer showed a decrease in the NPK concentration at the end of the analysis temperature 25°C, 30°C and 37°C (Fig. 1 and Fig. 2). Nitrogen, phosphorus and potassium were determined as nitrate-nitrogen ( $\text{NO}_3\text{-N}$ ), orthophosphate ( $\text{PO}_4^{3-}\text{P}$ ) and potassium ( $\text{K}^+$ ). The initial concentration of  $\text{NO}_3\text{-N}$  measured at 345 nm in control, spo-40 g, spo-80 g, spi-40 g and spi-80 g ranged from 0.820 mg/l to 0.956 mg/l for control, 0.950 mg/l to 1.150 mg/l for spo-40 g, 1.460 mg/l to 1.524 mg/l for spo-80 g, 0.981 mg/l to 1.44 mg/l for spi-40 g and 0.992 mg/l to 1.426 mg/l. The initial concentration of  $\text{PO}_4^{3-}\text{P}$  measured at 435nm showed a range of 7.011 mg/l to 12.40 mg/l for control, 14.33 mg/l to 18.5 for spo-40 g, 30.82 mg/l to 43.47 mg/l for spo-80 g, 7.47 mg/l to 10.43mg/l for spi-40 g and 19.0 mg/l to 20.30 mg/l for spi-80 g. Results for  $\text{K}^+$  all showed negatives values, thus was considered insignificant. The reduction in the concentration of  $\text{NO}_3\text{-N}$ ,  $\text{PO}_4^{3-}\text{P}$  in the control and treated soil samples (spo-40 g, spo-80 g and spi-40 g and spi-80 g) as seen in Figs. 1 and 2 suggests the utilization of nutrients during bioremediation process. Organic matter plays a great role in the regulating soil biological activity. In this study,

pig and poultry manure used as soil amendment also served as sources of organic matter thus stimulating microbial activity. Fig. 1 shows that amount of  $\text{NO}_3\text{-N}$  and  $\text{PO}_4^{3-}\text{p}$  utilized in the control was greater compared to amended soil (spo-40 g, spo-80 g, spi-40 g and spi-80 g). This high nutrient utilization in control (unamended soil) demonstrates the importance of biostimulation.

Soil contamination resulted in the reduction of nitrogen, phosphorus and potassium at all temperatures. Fig. 1 shows a significant reduction in the concentration of  $\text{NO}_3\text{-N}$  from 1.44 mg/l to 0.646 mg/l in spi-40 g and 1.37 mg/l to 0.607 mg/l in spi-80 g at 37°C. A significant reduction of  $\text{PO}_4^{3-}\text{p}$  from 7.011 mg/l to 3.5 mg/l in control and 7.47 mg/l to 4.45 mg/l in spi-40 g at 25°C was also observed. This shows a higher utilization of nitrogen in spi-40 g and spi-80 g at 37°C as compared to the other temperatures. At 25°C more phosphorus was used in control and at spi-40 g as compared to other temperature (30°C and 37°C).

Statistical results for potassium ( $\text{K}^+$ ) for control and treated samples at all temperatures showed no available potassium in the soil. Daniel et al [10], explains in a study that in the ecosystem about 90-98% potassium ( $\text{K}^+$ ) in the soil is unavailable for plant use, thus it needs to break down in order to be used. This affirms that if a soil and manure in use for degradation does not contain enough potassium, there is a high tendency of not finding it in the soil during remediation and after remediation.

### 3.2 Effect of pH on Biodegradation

In line with the objectives of this study, to determine the effect of the potential of hydrogen on bioremediation process; the pH of soil, manure, and contaminated soil were determined. Prior to the contamination with diesel and treatment with manure the pH of the soil was 5.30. After contamination with diesel there was an increase in pH value from 5.30 to 7.98 on the sixth day of the bioremediation process. This increase in pH value correlates with the results of Ekperusi and Aigbodion [11] who observed slight pH increase from 0.93-3.90 percent after 90 days of incubation [11]. Wang et al. similarly reported that crude oil contamination increases soil pH up to 8.0 [12]. The further change in soil pH from acidity to alkalinity may also be due to addition of pig manure and poultry manure considering their initial concentrations where high. Statistical

results of the initial pH concentration of pig and poultry manure showed that at 18.8°C the pH Value of pig manure was 9.54 and 9.76 for poultry manure at 19.3°C. At the end of the analysis within the temperatures considered, the pH of control, spo-40 g, spo-80 g, spi-40 g and spi 80 g were within the pH range of 7.71-9.58 at temperature range of 19.0°C – 20°C. Bacteria generally have an optimum growth pH range of 6.5-7.6 and acidic pH is often seen as toxic (Meiying et al. 2014). These condition needs to be sustained for optimal growth. However, results of pH analysis showed a slight increase in pH up to 9.58 (Table 2). These results correlate with that of Das and Chandran [1] which showed that the adequate performance of micro-organisms is within pH 6 – 9. This accounts for the high percentage removal of TPH seen in the control and spi-40 g at 37°C and 25°C.

The pH results of spi-40 g and spi-80 g confirms the results of Yakubu [8] that pig manure has the potential of improving soil quality after used as amendment for remediating contaminated soil. However, the results of this analysis also showed that spo-40 g and spo-80 g had higher pH values ranging from 8.73-9.58 than spi-40 g and spi-80 g which ranged from 8.15 – 8.70 (Table 2). This also implies that spo-40 g and spo-80 g can be used to improve the quality of acidic soils.

### 3.3 Hydrocarbon Degradation Efficiency

The experimental design involved spiking the soil with diesel rather than the use of aged

contaminated soil. Agarry et al. [13] showed that this form of spiking increases the bioavailability of hydrocarbons to inherent micro-organisms. The initial level of TPH in the soil after contamination was obtained on the 2<sup>nd</sup> day. This was to ascertain the TPH concentration at the pollution level of 27.5%.

#### 3.3.1 Initial TPH concentration

The initial concentration of TPH was determined as 173 mg/g at 25°C, 178 mg/g at 30°C and 170 mg/g at 37°C. The difference in initial TPH concentration analysed statistically showed a variance of 0.67, 4.33 and 3.6 respectively.

Statistical results obtained, showed that the mean difference in initial TPH concentrations, control (loam soil +diesel only) and treated soil samples (spo-40 g, spo-80 g, spi-40 g and spi-80 g) was not significant. This shows that at this point no degradation has occurred.

#### 3.3.2 Degradation kinetics/Diesel biodegradation at 25°C, 30°C and 37°C

The biodegradation trend of petroleum hydrocarbon for each nutrient amendment strategies: Soil treated with poultry manure (spo-40 g and spo-80 g), soil treated with pig manure (spi-40 g and spi-80 g) and control (soil +diesel only) showed that biodegradation began on the 5th day of the remediation process. This was observed at all temperatures.

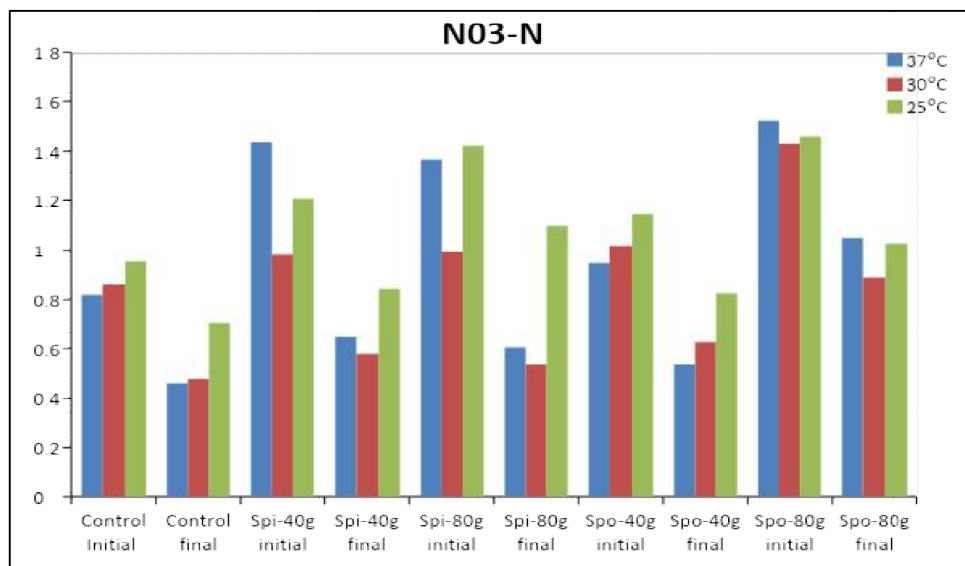


Fig. 1. Nitrogen (NO<sub>3</sub> – N) results at 25°C, 30°C and 37°C

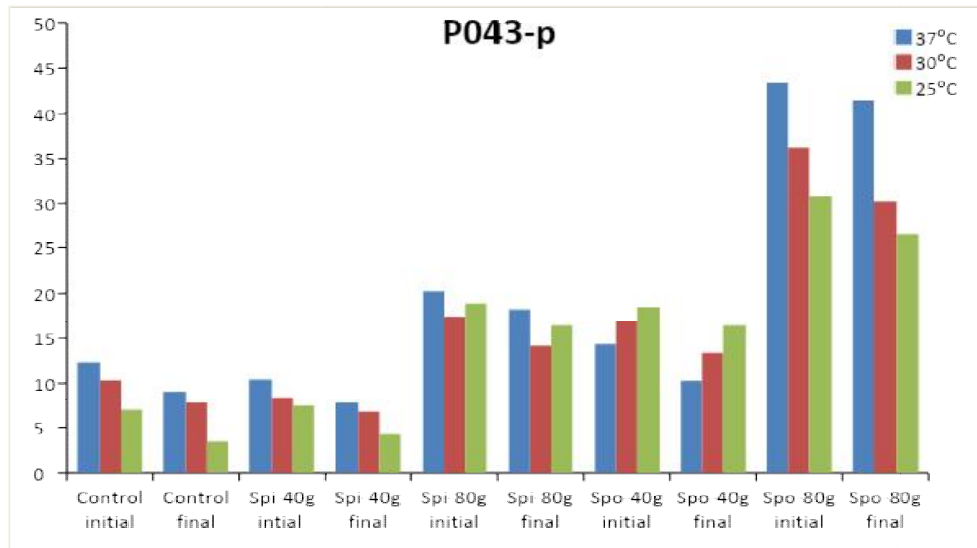


Fig. 2. Phosphorus (Po<sub>4</sub><sup>3</sup>-P) results at 25°C, 30°C and 37°C

Table 2. Initial and final pH results of soil samples at 25°C, 30°C and 37°C

Control	Initial p <sup>H</sup> results			Final p <sup>H</sup> results			Tempt (ATC)
	25°C	30°C	37°C	25°C	30°C	37°C	
	8.25	7.78	7.17	8.84	8.40	7.71	19.5°C-19.7°C
Spi-40g	8.13	8.05	7.68	8.70	8.52	8.15	19.7°C – 20.0°C
Spi-80g	7.57	7.80	7.78	8.36	8.44	8.19	19.4°C -19.8°C
Spo-40g	8.30	8.10	8.23	8.91	8.87	8.73	19.3°C -19.5°C
Spo-80g	7.8	8.82	8.58	8.80	9.27	9.58	19.5°C – 19.8°C

3.3.2.1 Control at 25°C, 30°C and 37°C

TPH percent loss by natural attenuation (control) presented in Fig. 3 showed the smallest percent removal of 3.03% on the 5<sup>th</sup> day at 37°C as compared to that of 30°C and 25°C which was 33% and 57%. The significant difference in TPH percent removal could be due to errors in experimental setup, however natural attenuation is a slow process and could also account for the delay in remediation on the 5<sup>th</sup> day at 37°C (Bambang et al. 2010). On the 10<sup>th</sup>, 15<sup>th</sup> and 17<sup>th</sup> day all the natural attenuated soil at all temperatures showed significant increase in percent TPH removal (Fig. 3).

Fig. 3 shows a 0.9515% reduction in TPH at 30°C on the 15<sup>th</sup> day. This reduction in TPH

removal at 30°C could be due to reduction in available nutrients, water and oxygen etc. Moreover, as the remediation continued to the 17<sup>th</sup> day, the researcher observed that there was an increase in the percent of TPH removal at all temperatures and equivalent decrease in nutrient concentration (Fig. 4). Having observed the degradation trend, there was a spontaneous decrease in concentration of contaminant at 25°C, 37°C.

3.3.2.2 Soil treated with 40 g and 80 g of poultry manure (spo-40 g, spo-80 g)

The additions of poultry manure at concentrations of 40 g and 80 g to the diesel polluted soil enhanced biodegradation. The percent loss as calculated in equation 11 above showed 12.94%, 48% and 72% removal in spo-



40 g at 37°C, 30°C and 25°C while in spo-80 g there was 29.14%, 54% and 68% TPH removal at 37°C, 30°C and 25°C respectively on the 5<sup>th</sup> day (Figs. 5 and 6).

The difference in TPH percent removal between spo-40 g and spo-80 g showed that spo-80 g was twice more on the 5<sup>th</sup> day at 37°C. This implies that an increase in the concentration of stimulated manure may yield an equivalent increase in biodegradation because of sufficient nutrients and micro-organisms. Statistical values of percent degradation also showed that degradation started on 5<sup>th</sup> day in both spo-40g and spo-80 g at 25°C, 30°C and 37°C (A1 -A3). On the 10<sup>th</sup>, 15<sup>th</sup> day of bioremediation, the researcher observed negatives values for spo-40 g and spo-80 g at all temperatures, thus no degradation. This may be due to inhibition caused by competition for limited nutrient; water and reduction in the number of bacteria necessary for degradation. Nwogu et al. [14] stated in a study of enhanced bioremediation of artificially contaminated soil that maintaining nitrogen and phosphorus concentrations during bioremediation is necessary for optimal performance (Nwogu et al. 2015). Though no degradation occurred on day 10 and 15, it was observed that on the 17<sup>th</sup> day degradation occurred. Figs. 7 and 8 showed significant degradation on the 17<sup>th</sup> day in spo-40 g at 25°C, 30°C and 37°C where as in spo-80g percent degradation was observed only at 37°C and

25°C. The high percent degradation of 2.94% (Spo-40 g) and 5.88% (spo-80 g) at 37°C as compared to 1.8% (spo-40 g) and 1% (spo-80 g) at 25°C suggests that the temperature (37°C) in which these samples were cultured also provided a more conducive environment for heterotrophic bacteria to grow thus higher degradation [1]. Also, the increase in percent removal as seen in Fig. 3 implies that nitrogen and phosphorus were utilised during biodegradation of diesel-contaminated soil.

3.3.2.3 Soil treated with 40 g and 80 g of pig manure (spi-40 g, spi-80 g)

Biostimulation of contaminated soil with organic nutrients increases the rate of biodegradation. The effect of pig manure on diesel contaminated soil was investigated at concentrations of 40 g and 80 g respectively. Results obtained showed that the addition of pig manure to the diesel polluted soil enhanced biodegradation. On the 5<sup>th</sup> day of biodegradation process the researcher observed a 72%, 45.1% and 20.59%, TPH removal at 25°C, 30°C and 37°C respectively in spi-40 g and 72%, 65.17% and 73.53% at 25°C, 30°C and 37°C respectively in spi-80 g. These results showed that soil samples treated with pig manure showed greater percent removal than the control on the 5<sup>th</sup> day with some variations in percent removal between spi-40 g and spi-80 g at 25°C, 30°C and 37°C (Figs. 9 and 10).

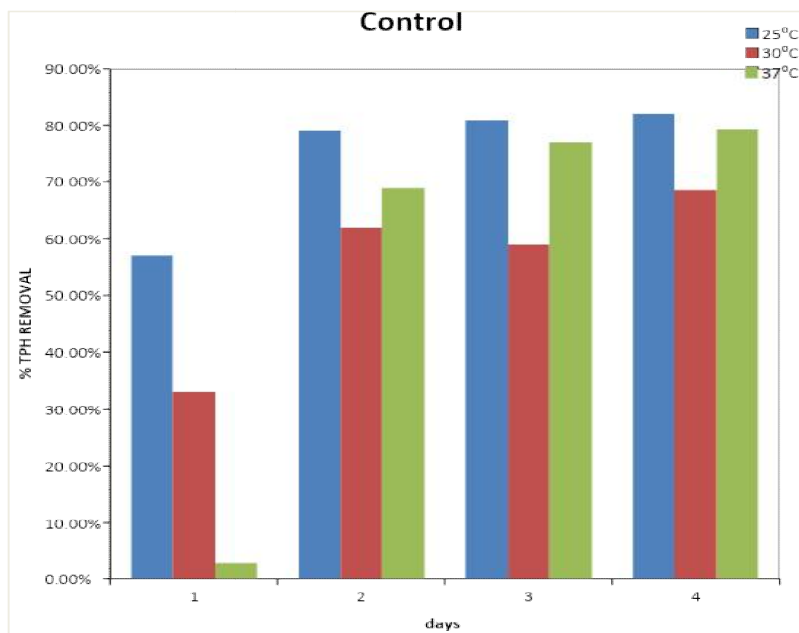


Fig. 3. % TPH removal in control (Natural attenuation)

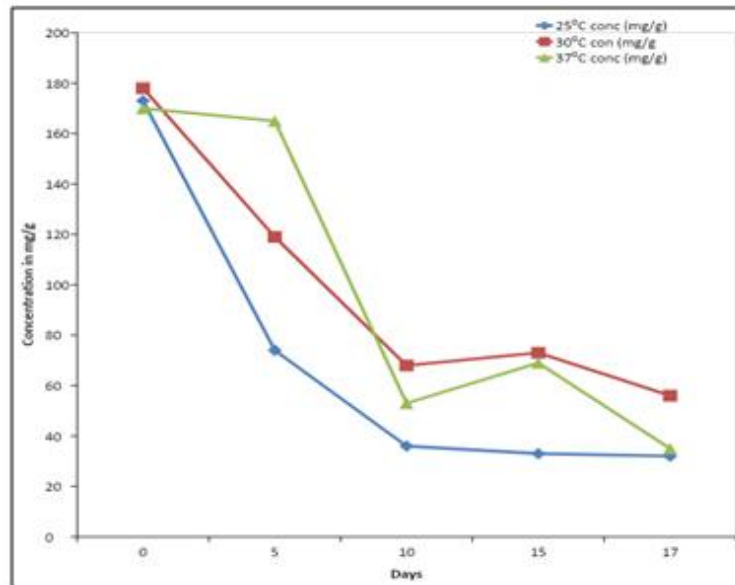


Fig. 4. Trend of degradation in control at 25°C, 30°C and 37°C

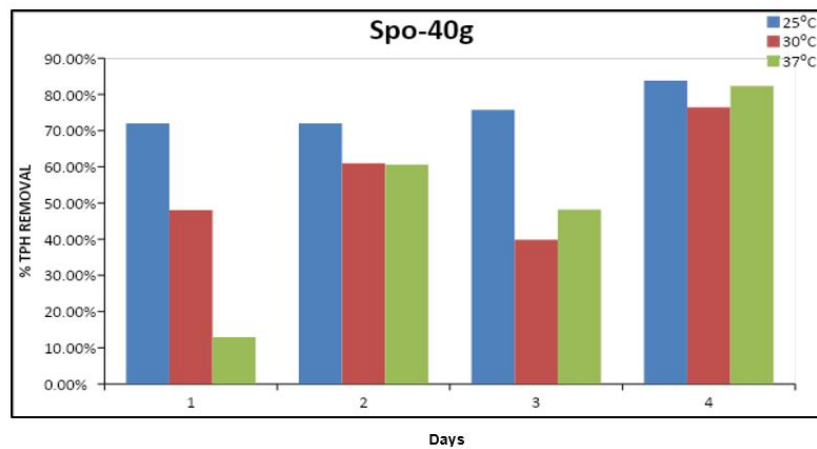


Fig. 5. % TPH removal in spo-40 g

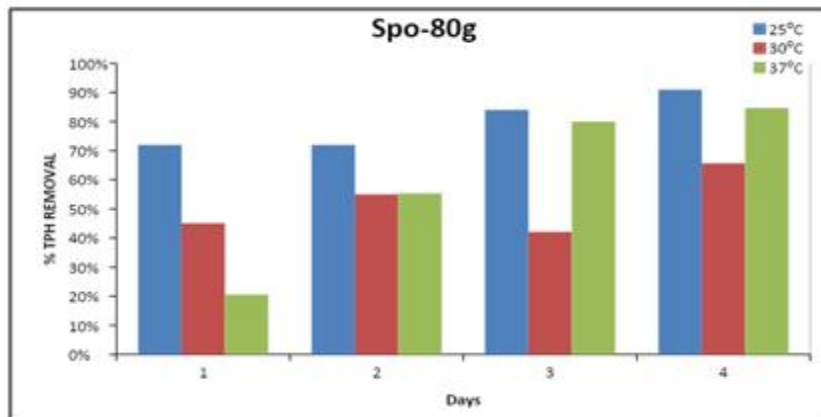


Fig. 6. %TPH removal in spo-80 g

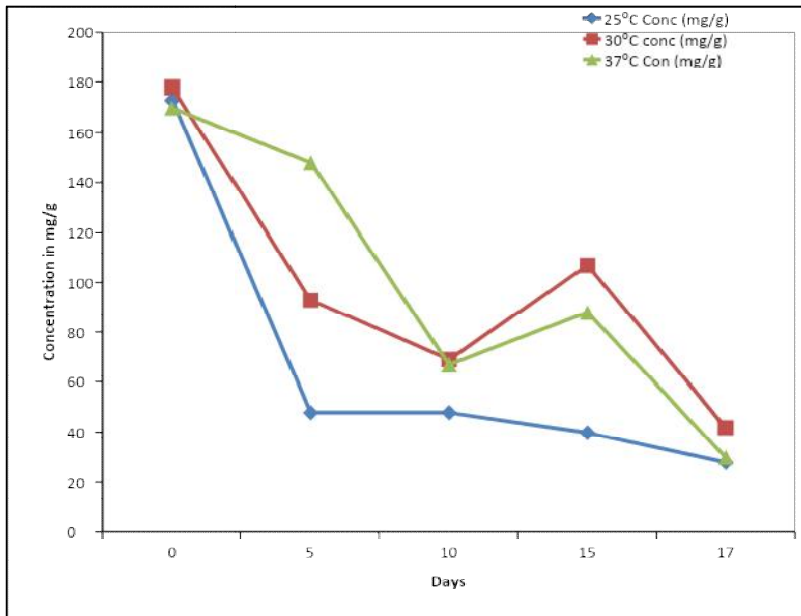


Fig. 7. Trend of degradation in soil treated with 40 g of poultry manure at 25°C, 30°C and 37°C

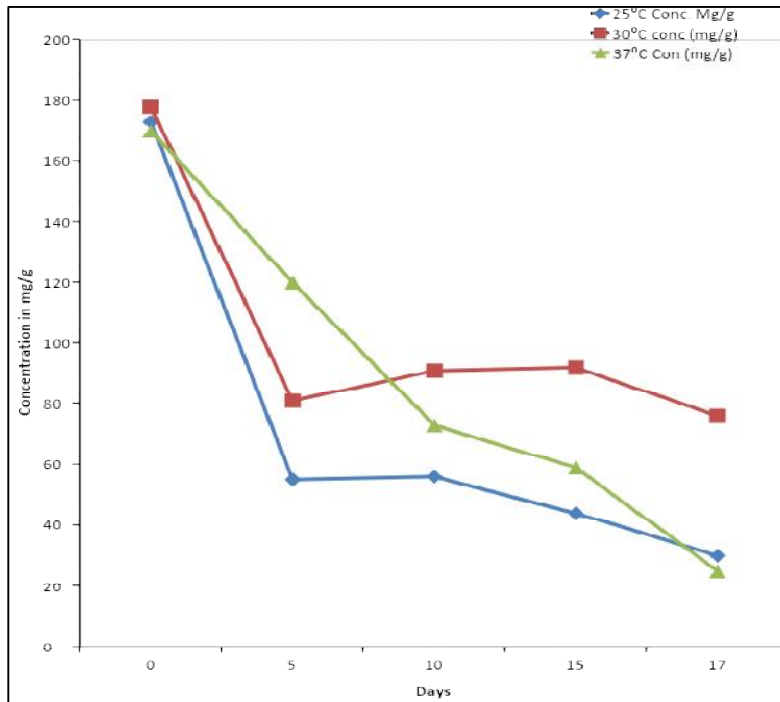


Fig. 8. Trend of degradation in soil treated with 80 g of poultry manure at 25°C, 30°C and 37°C

Figs. 9 and 10 shows high percent removal at 25°C and at 37°C in both spi-40 g and spi-80 g. However, the highest percent removal on the 17<sup>th</sup> day in spi-40 g was at 25°C and spi-80 g was at 37°C.

Statistical results of spi-40 g and spi-80 g at 37°C for percent TPH removal and percent degradation was greater than spo-40 and spo-80 g at the same temperature. However, the percent degradation of spo-80g was higher than spi-40g

at 37°C. Generally, spi-80 g had the highest percent removal of TPH and percent degradation than all treated soil samples including control at 37°C (A1). This is because pig manure used were not tightly bound and particle size were smaller thus could mix properly with the soil. Also, the microbial consortium in pig manure could also account for high percent removal in diesel contaminated soil [13]. The percent removal of TPH in spi-40 g, spi-80 g at 30°C on the 5<sup>th</sup> day was higher than spi-40 g and spi-80 g at 25°C. However, the degradation percent of spi-80 g at 30°C on the 5<sup>th</sup> day was higher than the degradation percent of spi-40 g and spi-80 g

at 25°C. Notwithstanding, on the 17<sup>th</sup> day of remediation process, results of analysis showed 9% and 4% degradation in spi-40 g and spi- 80g respectively at 25°C and -2.77% and - 9.52% in spi-40 g and spi-80 g respectively at 30°C. This shows that no degradation was observed on the 17<sup>th</sup> day in spi-40 g and spi-80 g at 30°C. This suggests that the temperature (30°C) didn't favour the growth of hydrocarbon degrading bacteria or the amount of nitrogen and phosphorus remaining at day 17 was insufficient for microbial activity. Figs. 11 and 12 shows high decrease in concentration of diesel with respect to time on the 17<sup>th</sup> day at 25°C and 37°C.

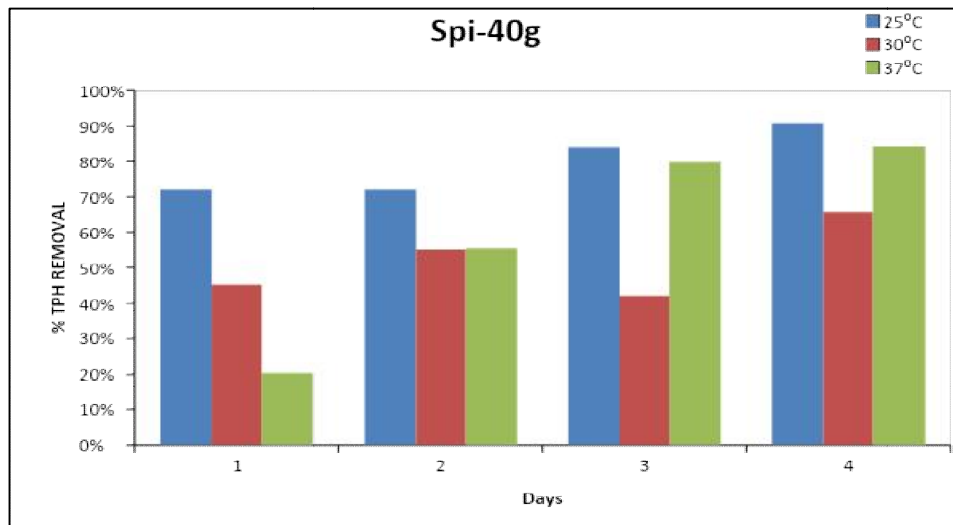


Fig. 9. % TPH removal in spi-40 g

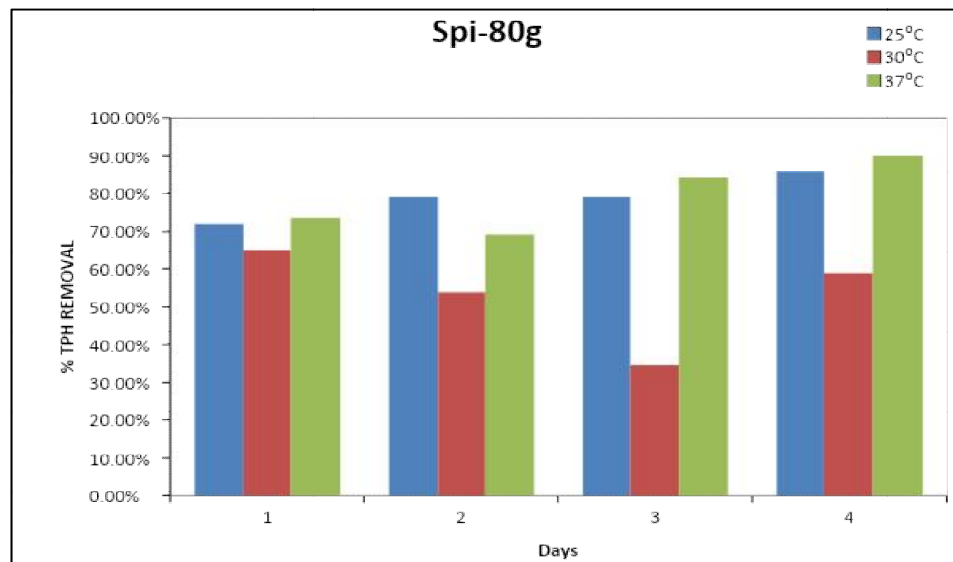


Fig. 10. % TPH removal in spi-80 g

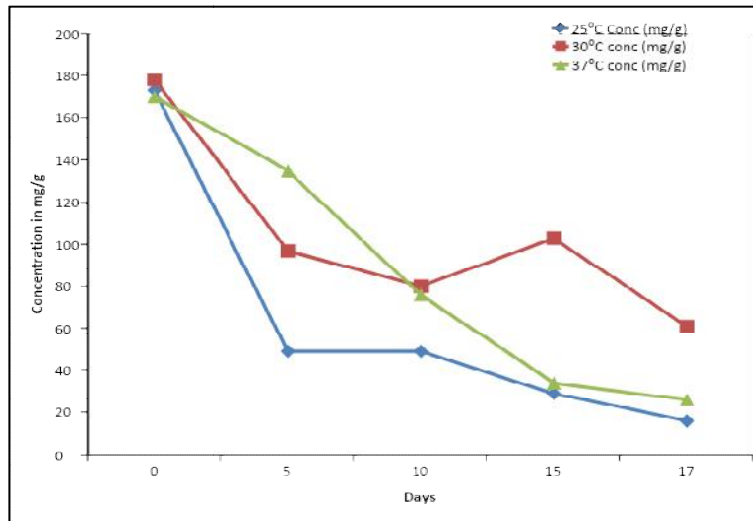


Fig. 11. Trend in degradation in soil treated with 40 g pig manure (spi-40 g) at 25°C, 30°C and 37°C

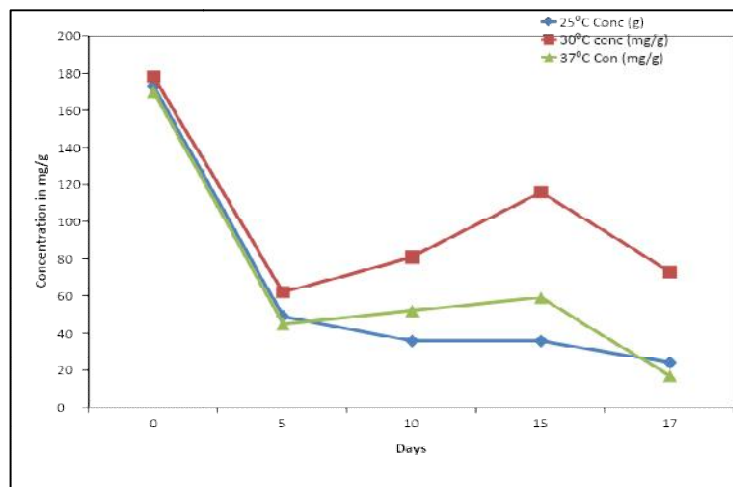


Fig. 12. Trend in degradation in soil treated with 80 g pig manure (spi-40 g) at 25°C, 30°C and 37°C

#### 4. CONCLUSION

The results of this research demonstrated the potential of pig and poultry manure in remediating diesel contaminated soil. Results of the analysis showed high percent degradation at 25°C and 37°C with little or no degradation at 30°C. There was a spontaneous decrease in TPH in control however on the 17<sup>th</sup> day the treated soil samples showed higher % degradation of 2.94%, 5.3% 5.88%, 10.59% for spo-40 g, spi-40 g, spo-80 g and spi-80 g respectively at 37°C on the 17<sup>th</sup> day. At 30°C, percent degradation of 7.9% was observed only in spo-40 g whereas at 25°C there was 1%,

1.8%, 4% and 9% degradation in spo-80 g, spo-40 g, spi-80 g and spi-40 g respectively. Results of pH analysis showed that soil treated with 80g of poultry manure had the highest pH value of 9.27 and 9.58 at 30°C and 37°C respectively. At this pH (9.27 and 9.58) hydrocarbon degrading bacteria cannot thrive, thus slow metabolism of diesel. This explains the relatively low % degradation observed in spo-80g as compared to others at the same temperature. The concentration, state and temperature of the spo-80 g could have also contributed to the low degradation level observed. However, at 37°C, spo-80 g had 5.88% degradation and the lowest activation energy of 0.023 KJ/mol. Results of

TPH analysis showed that soil samples with high percent removal also demonstrated high percent degradation.

Generally, it can be seen from the results that at 37°C diesel degradation occurred more in all soil samples than at 30°C and 25°C. Therefore, it can be concluded that 37°C is most suitable for diesel degradation with highest efficiency in soil treated with 80 g of pig manure. However, at 25°C, high percent degradation also occurred in all treated samples with spi-40 g having the highest percent degradation.

### COMPETING INTERESTS

Authors have declared that no competing interests exist.

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