



THE UTILIZATION OF MANGO LEAF AND NANO-PARTICLE FOR TOTAL PETROLEUM HYDROCARBON DEGRADATION IN POLLUTED SOIL

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Abstract

The research focused on the degradation of petroleum polluted loamy soil for the removal of Total Petroleum Hydrocarbon (TPH) using Mango Leaf and Nano-particle treatment methods. Mango leaf was prepared by washing with water to remove dirt's and impurities, sun and air oven dried for complete moisture removal prior to its mechanical grinding and sieve to desired micron, while the nanoparticle was achieved by reacting magnesium chloride with sodium hydroxide and the produced nanoparticle was dried and ground to powdered form. The TPH value of loamy soil prior and after pollution was 7.73mg/kg and 15666.7mg/kg respectively. Bacteria growth rate crucial for reduction of TPH was carried out with Mango leaf and Nano-Particle treatment in a batch bio-reactor, which showed a progression across the treatment variations for different sample weight. The TPH percentage degradation in polluted loamy soil for nano-particle treatment ranges between 72.65%, and 99.17% for specific treatment weight, while the TPH percentage degradation for polluted loamy soil using the mango leaf treatment yielded between 70.43%, and 97.21% for different weight samples respectively. The rate constant, predictive model and half-life for loamy soil under mango leaf and nano-particles were also estimated and developed for the control and different weight samples respectively. The model performance indicated that the correlation between the experiment and the expected TPH concentrations by first-order rate kinetics model was 0.9769 and 0.95603 for nano-particles and mango leaf respectively, correlation between the experiment and the Michaelis-Menten equation for both methods were 0.9641 and 0.9971 respectively and correlation between first-order kinetic and the Michaelis-Menten equation yielded 0.9586 and 0.9325 accordingly for nanoparticle and mango leaf. Thus, first-order rate kinetics model predicted TPH degradation with the experimental data more accurately than the Michaelis-Menten model equation. Hence, TPH degradation in polluted soil was achieved by nanoparticle and mango leaf treatment techniques with percentage degradation of 99.17% and 97.21% for nanoparticle and mango leaf respectively at 56th treatment day for 100g sample.

1.0 INTRODUCTION

While exploiting, producing, and processing operations, oil and gas plants produce a lot of oil waste. Odiete asserts that the environment is often impacted in some way by all hydrocarbon production and exploration operations [1]. Petroleum tanks that conveyed crude oil can leak and discharge its content to the surroundings, thereby contributing to environmental damage [2, 3]. Contamination by Crude oil can happen in aquatic and terrestrial settings through spills from oil facilities, or it can

occur in the atmosphere through gas flaring or the vaporization of volatile components. According to research, the most significant contamination from the petroleum sector harm the terrestrial environment, mostly from pipeline, storage tank, and other oil facility leaks [4].

Crude oil impacts plant development and includes radioactive elements and poisonous chemicals that pose major health risks [5, 6]. Due to its low water solubility, petroleum pollution, particularly on soil, holds the potential to hold soil fragments firmly, thereby lowering soil nutrients [7]. The majority of inorganic and organic chemicals are harmful to the soil and can result in low agricultural products yield [8, 35]. Petroleum-entrained soil can be cured through use of various procedures; however, the selection of an approach is based on accessibility, value for money, and elimination of contaminants capability [9]. It is crucial to have an environmentally friendly approach for eliminating petroleum pollutants from the soil and other parts of the environment. Techniques including physical, biological, and chemical procedures could be utilized to clean up the land that has been damaged by oil-based hydrocarbons [10, 11]. These techniques included traditional excavation, removing the contaminated soil to landfills, capping the contaminated sections on the site, stabilizing the soil with cement and certain materials, cremation, and the implementation of organisms. Although the traditional approaches seem to be less expensive and need less experience, they don't completely resolve the issue [11, 12]. Due to its many benefits, the biological technique is typically used to remediate land that entrained by crude hydrocarbons [12, 13]. Accordingly, the availability of remediating agents capable of amending contaminated soil made the biological method one of the most applied techniques for the elimination of hydrocarbon impurities from soil [13]. The biological technique is generally known as bioremediation. Bioremediation is a radiation technique that utilises microorganism or bio-stimulants such as enzymes, spent biomass and fertilizers to remove contaminants from contaminated environment through metabolic process [14]. Bioremediation is economical, adaptable, efficient and eco-friendly to the environment [15].

Bioremediation can be achieved through bio-stimulation, bio-augmentation and phytoremediation, soil washing, thermal treatment, oxidation, natural attenuation etc [16, 17]. Bio-stimulation refers to the use of fertilizer, plant and animal wastes, or any

other agro based material as amendment agents, to arouse the development of microorganisms; while in bio-augmentation bacteria or fungi is directly applied to improve the depletion rate of pollutant [18-20]. In Phytoremediation, a plant is planted in polluted soil to take up the contaminant as it grows. Most often, bioremediation process takes place in a system best described as reactor, which can be implemented in batch or continuous reactors such as the fixed bed, fluidized bed and membrane reactors [21-23]

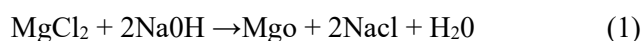
By controlling and monitoring the development of microorganisms required for the breakdown of pollutants, the usage of bioreactors creates the ideal circumstances for the effectiveness of the process [15].

This study adopted the bio-stimulation method implemented in batch reactor for bioremediation of TPH contaminant in entrained loamy soil via the application of Mango Leaf and Nanoparticle (magnesium oxide). This is achieved by characterizing the properties of substrate, chemically and physically, substrate prior and after contaminations with hydrocarbon, examination of the formation rate of bacteria crucial in TPH reduction, analysis of its depletion rate of the TPHs in the samples when mango leaf and nanoparticle (magnesium oxide) at different weights are applied for treatment, development of predictive model equation for batch reactor remediation process, evaluation of degradation rate constant and TPH half-life and performance evaluation of model and empirical analysis

2.0 MATERIALS AND METHODS

2.1 Production of Nanoparticle (Magnesium Oxide)

By use of a process that is exothermic or co-precipitate method, 40g of sodium hydroxide (NaOH) is dissolved into 250ml of water (distilled), and also a process with a reaction that is endothermic of magnesium (II) chloride ($MgCl_2$, 26g) in water (distilled, 160ml), or "wet chemical method," the mixture produced a slightly yellowish substance. To create a dark-colored precipitate, 6g of iron chloride ($FeCl_3$) is gradually dispersed in water (distilled, 25ml) and NaOH (14g). The intended and generated nanoparticle went through drying and then ground into a powder for usage in operations.



2.2 Preparation of Mango Leaf

The mango leaves were plucked and picked from a mango tree and washed to remove dirt's and impurities prior to its cleaning with distilled water. The washed mango leaves were sun dried to remove moisture (water contents), thereby turning the leaves brownish in colour prior to its drying in an air oven for 24 hours at a temperature range between 60°C and 105°C for complete removal of moisture. The dried mango leaves were crushed and ground into a fine powder using mechanical grinder and sieve to achieve uniform particle size prior to its application for treatment of polluted soil. This natural plant contains high organic matter and mineral contents such as potassium, nitrogen, phosphorus, calcium etc that acts as substrate for soil microbes. Also, the presence of porous and bio-active compounds (mangiferin, tanins, flavonoids, terpenoids) in mango leaf creates a favourable condition for TPH degradation due to large surface area after preparation.

2.3 Experimental Procedure

400g slit loamy soil specimens was loaded into 12 different batch reactors (control 1, control 2) and 5g, 25g, 50g, 75g, and 100g samples were weighed in duplicate for the two treatment methods respectively. All samples were analyzed before (control 1) pollution and after (control 2) pollution and each gramme of soil samples was treated using nanoparticle and mango leaf respectively. For a consistent level of petroleum in the soil specimen, two samples (polluted) were thoroughly mixed and allowed to rest undisturbed for three days. Following a period of soil settling, the soil specimens were treated by using 5g, 25g, 50g, 75g, and 100g samples of mango leaf and nanoparticle, respectively. The unpolluted soil was the control one (1) without treatment, and the samples were stirred at three days interval to guarantee consistent level of treatment rates in the reactor, and each soil sample was sent to the laboratory for analysis of physicochemical parameter, TPH, Polycyclic Aromatic Hydrocarbons (PAHs) and Total Heterotrophic Bacteria (THB) count for period of 35 days.

2.4 Determination of Total Petroleum Hydrocarbon

The study of the TPH by use of a Gas Chromatography was carried out using a Flame Ionization Detector Model HP 5890 series II. This analysis was done by Analytical Concept Limited. Total Petroleum Hydrocarbon was obtained using

calibrated graph in the software of the equipment as a reference.

2.5 Soil pH Analysis

Soil pH was determined using pH meter. 20g of 2mm air-dried soil was put into a beaker, weighed and 50ml of distilled water was added and thoroughly swirled for about five minutes with a glass rod before being left for thirty minutes. The glass electrode was calibrated with a pH 4 or pH 9.2 buffer and standardized with a standard buffer of pH 7. By continuous stirring, the electrodes are submerged in the beakers holding the soil water suspension. The pH value was recorded to the closest 0.1 unit after 30 seconds. After recording, the electrodes were taken out of the soil solution and washed with distilled water, and the pH meter was immediately placed in standby mode. After every determination, the electrodes were cleansed and gently wiped dry using filter paper. The glass electrodes were standardized after every 10 readings.

2.6 Total Organic Carbon

Umeda [24] proposed a technique for determining total organic carbon (TOC). Therefore, 1g of earth specimens was placed into a beaker of 250ml, then 10ml of a solution of potassium dichromate was dripped into the bottles by a pipette then slowly spun to fully moisten the sample. After that, concentrated H₂SO₄ of about 20ml was introduced with an automated pipette and carefully spun for 60 seconds to achieve an evenly distributed suspension, and for efficient and more thorough oxidation, prior to allowing it to set for roughly half an hour on sheet of asbestos.

Three to four drops of 0.5 ml diphenylamine indicator were applied after 100 ml of distilled water had settled. 0.5N ferrous sulphate solution was engaged to mix with the solution by titration till the color transitioned to blue from violet and then to vivid green. In order to homogenize the dichromate, the procedure was carried out again using distilled water (blank titration) yet with no dirt. The formula was used in calculating the TOC.

$$TOC = Blank - \frac{\text{volume of soil sample titre} \times 0.195}{\text{weight of soil sample}} \times 100\% \quad (2)$$

2.7 Total Bacterial Count (TBC)

Heterotrophic bacteria and fungus were counted using the pour plating method in a microbiological investigation. This was accomplished by introducing mineral salt agar (MSA) (for degrading of



hydrocarbons), acidified streptomycin (1 mg/100 ml) (fungal), and nutrients agar (bacterial) with 0–1 ml of tenfold serially diluted material. The subsequent composition, expressed in grams per liter of distilled water, is found in the mineral salt medium of Miu et al. as amended by Okpokwasili & Amanchukwu [25]: NaCl (10g), MgSO₄·7H₂O (9.42g), HP04 (1.2g), KH₂P04 (0.83g), KCl (0.29g), NaN02 (0.42g), Agar-Agar (16g), pH 7.2, and 2 mill at gasoline/diesel. Whereas the potato dextrose agar plates were hatched at normal conditions of temperature and measured and reported in cfu/ml, the injected nutrient agar plates were gestated at 37°C for a day.

2.8 Total Petroleum Hydrocarbon (TPH)

The Gas Chromatography-Flame Ionization Detector (GC-FID) was used to analyze the TPH. A soil specimen was transferred into a 1-liter separation funnel as part of the process to determine the TPH. To elevate the inner surface, 50 milliliters of methylene chloride were poured into the container used for holding sample, sealed and shaken for half a minute. After moving the solvent to the separation funnel, the funnel was shaken for two minutes while being periodically vented to relieve any remaining pressure. For at least ten minutes, the organic level was left to differentiate from the aqueous phase. A 250 ml flask was used to extract the methylene chloride. The sample bottle was filled with a second 60ml amount of methylene chloride. Twenty-five milliliters of the solvent were poured into the extract to clean the separating funnel and the separating column. In an Erlenmeyer flask, the extraction process was carried out twice and combined with the other extract. The identical procedure was used for the following extraction. The mixture of extracts was passed down a drying column that contained silica, anhydrous sodium sulfate, and packed cotton wool.

After being collected in a vial, the extract was concentrated by boiling it to 1.0 ml using nitrogen gas. For the TPH analysis, 1-0 ml of the mixture was introduced into the flame ionization detector gas chromatograph after the rest of the extract and 1.0 ml of the solvent had been combined.

The remaining TPH percentage at a specific time was estimated by Equation (3).

$$TPH_R(\%) = \frac{TPH_i - TPH_f}{TPH_i} \times 100\% \quad (3)$$

3.0 MODEL DEVELOPMENT

3.1 First Order Model

One of the most useful models for examining the procedure of bioremediation is the first order kinetic model. Once the speed of decomposition constant has been established, the rate of biodegradation model is utilized to forecast the content of TPH in samples at whatever point throughout the period for bioremediation [26–28]. A mathematical model that describes a system is frequently developed using the conservation of mass balance concept.

$$\left\{ \begin{array}{l} \text{Mass flow} \\ \text{into system} \end{array} \right\} = \left\{ \begin{array}{l} \text{Mass flow} \\ \text{out of the} \\ \text{system} \end{array} \right\} + \left\{ \begin{array}{l} \text{Rate of} \\ \text{depletion by} \\ \text{reactions} \end{array} \right\} + \left\{ \begin{array}{l} \text{Rate of} \\ \text{accumulation} \\ \text{of mass} \end{array} \right\} \quad (4)$$

The different terms in Equation (4) are therefore expressed as

$$Q_o C_{TPH(o)} = Q C_{TPH} - r_{TPH} V + \frac{d(C_{TPH} V)}{dt} \quad (5)$$

Since bioremediation takes place in a batch reactor, with no inflow and outflow terms and constant reactor volume and application of first order kinetics, the rate of degradation upon mathematical analysis yields

$$\ln C_{TPH(t)} = \ln C_{TPH(o)} - k_d t \quad (6)$$

The instantaneous TPH concentration is deduced by Equation (6) to yield

$$C_{TPH(t)} = C_{TPH(o)} e^{-k_d t} \quad (7)$$

Equation (7) is engaged to estimate the TPH concentration remaining in the soil at time t.

3.2 Half Life of TPH Degradation

Half-life is defined as the time needed for a given amount of TPH in soil to degrade to half of its initial value [29]. The half-life was evaluated using Equation (8).

$$t^{1/2} = \frac{\ln(2)}{k_d} = \frac{0.6931}{k_d} \quad (8)$$

3.3 Michaelis-Menten Equation

The rate of depletion of TPH was also studied using Michaelis-Menten Equation, which is expressed as:



$$-r_{TPH} = -\frac{dC_{TPH(t)}}{dt} = \frac{\mu_{max} C_{TPH(t)}}{K_s + C_{TPH(t)}} \quad (9)$$

Equation (9) is linearized to obtain the greatest definite rate constant and the Michaelis-Menten constant according to Equation (10).

$$-\frac{1}{r_{TPH}} = \frac{1}{\mu_{max}} + \frac{K_s}{\mu_{max}} \left(\frac{1}{C_{TPH(t)}} \right) \quad (10)$$

A graph of $-\frac{1}{r_{TPH}}$ against $\frac{1}{C_{TPH(t)}}$ provides a slope of $\frac{K_s}{\mu_{max}}$ with $\frac{1}{\mu_{max}}$ at the intercept.

4.0 RESULTS AND DISCUSSION

4.1 Physicochemical Characteristics of Soil before and after Pollution

The physicochemical characteristics of the specimens prior to and at the end of contamination are revealed in Table 1.

Table 1 illustrates the alterations in the physicochemical attributes of the loamy specimens at 2 different conditions, prior to pollution and then after. The modifications in these properties post-pollution reveal the substantial work of petroleum upon the sample composition. The examination revealed noteworthy reductions in pH, phosphorus (P), nitrogen (N), potassium (K), and Total Bacteria Counts (TBC) in the soil subsequent to crude oil contamination. Conversely, Total Organic Carbon (TOC) in the soil exhibited an increase following the pollution event.

Table 1: Physico-chemical characteristics of soil

Parameters	Loamy Before	After
Temperature (°C)	27	28
pH	6.46	5.3
Microbial Counts (%)	33.37	65.4
Total Organic Content (%)	1.14	150.45
Total Nitrogen (%)	21.2	2.34
Phosphorus Content (mg/kg)	2.23	1.3
Total Petroleum Hydrocarbon (mg/kg)	7.73	15666.7
Total Heterotrophic Bacterial Count (cfu/g)	2.05 x 10 ¹¹	0.217x10 ¹¹
Porosity	0.673	0.241
Bulk Density (g/cm ³)	1.22	8.31

4.2 Total Heterotrophic Bacteria Count

The progression of Total Bacteria Counts (TBC) growth in hydrocarbon polluted loamy soil that underwent different Mango leaf weight treatments over time was depicted in Figure 1. As a result of the pollution, there is reduction in TBC count within the soil. However, upon the introduction of mango leaf treatments, the TBC population in the soil exhibited a notable rise as the treatment weight increases. The TBC within the loamy soil treated with Mango leaves exhibited a positive progression across the treatment variations due to the presence of porous and bio-active compounds (mangiferin, tanins, flavonoids, terpenoids) in mango leaf that creates a favourable condition for the degradation.

Also, the TBC in polluted loamy soil treated with Nano-particle of several weight over time yielded a similar path way as mango leaf treatment as depicted in Figure 2. Thus, Mango leaf treated polluted soil samples exhibited a higher TBC 515.20% (57563cfu/g for control to 296563cfu/g on the 35th day) compared to the Nano-particle 507.26% (58896cfu/g for control to 298754cfu/g on the 35th day) treated soil samples due to the presence of minerals and bio-active compounds that acted as substrate for the microbes in mango leaf treatment, thereby favouring TPH degradation process.



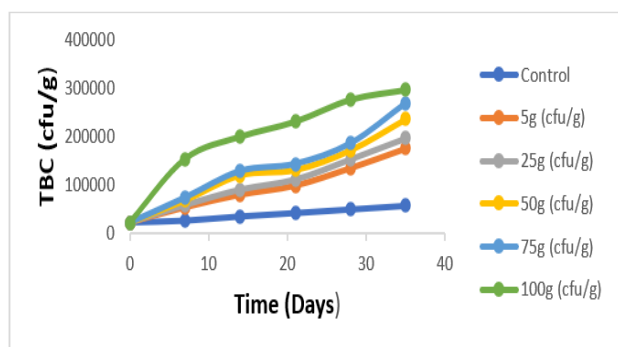


Figure 1: TBC count variation versus time for mango leaf treatment

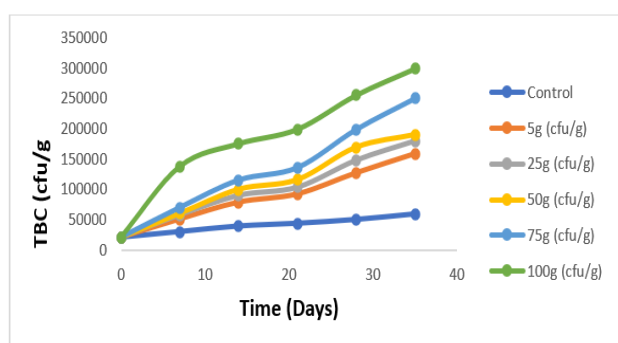


Figure 2: TBC count variation versus time for nano-particle treatment

4.3 Total Petroleum Hydrocarbon Degradation

The degradation trend of TPH in contaminated loamy soil specimens treated with mango leaf and nano-particles in comparison with control samples across various treatment weights and time intervals are depicted in Figure 3 and Figure 4 respectively.

The TPH percentage degradation profiles within the polluted loamy soil reveal that as the experimental process proceeds with time, there is corresponding increase or rise in the percentage of degradation with treatment samples. These variations signify a reduction in the TPH concentration over time. Based on the experimental analysis, it's notable that substantial TPH degradation occurred in 35 days across the various treatment weights, indicating the significant impact of the remediation time on degradation efficiency. Further analysis yielded an optimized degradation percentages of 99.17% and 97.21% for nanoparticle and mango leaf treatment methods respectively after 56 days of degradation time. The results achieved in this research are similar with reported effect of treatment samples on crude oil polluted soil by researchers such as Yu *et al.*, Ogu & Odo and Ukpaka & Pepple [9, 30, 31]. However, Akpe *et al.* observed that elevated levels of crude oil in specimens can negatively affect TPH degradation efficiency, thus 5g sample recorded the least TPH

degradation rate [32]. This implied that treatment contain nutrients that stimulated the hydrocarbon degrading bacteria as posited by Aghalibe *et al.*, Al-Haeash *et al.* and Ere and Amagbo respectively [27, 33, 34]

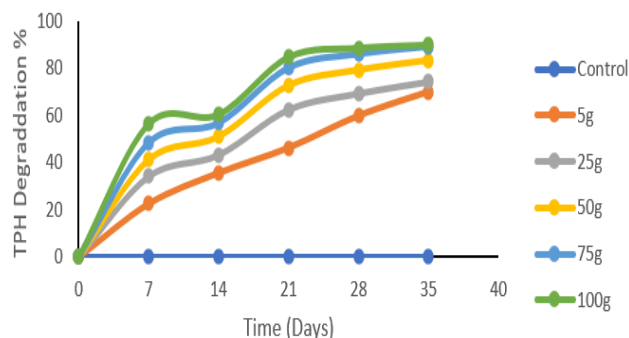


Figure 3: TPH degradation in loamy soil versus time for nano-particle treatment

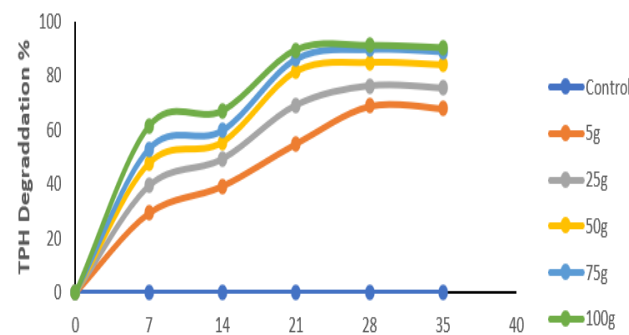


Figure 4: TPH degradation versus time in loamy soil for mango leaf treatment

A similar behavioural trend was monitored in polluted loamy soil inoculated with mango leaf. Therefore, the experimental data was extrapolated for further treatment days of 42nd, 49th and 56th for mango leaf and nanoparticle respectively. The observed TPH percentage degradations are 94.96%, 96.09% and 97.21% for mango leaf while 95.52%, 97.35% and 99.17% for nanoparticle for 42nd, 49th and 56th treatment days respectively. Thus, improved or optimized TPH degradation can be achieved for increased treatment days based on the degradation observed trend.

4.4 Evaluation of Treatment Performance in the Soil

The effectiveness of nanoparticle and mango leaf as bio-stimulants in TPH depletion in petroleum-contaminated loamy soil was contrasted as depicted in Figure 5.

Thus, the comparison of TPH percentage degradation between mango leaf and nanoparticle treatments in polluted loams at 100g treatments amounts were



conducted. As evident from the profiles, the TPH percent degradation in the specimen treated with nanoparticle slightly exceeded that of the sample treated with mango leaf. Also, the degradation rate exhibited a swifter progression with a steeper slope in the mango leaf treated samples in contrast to the nanoparticle treated samples.

LSML – Loamy soil with mango leaves, LSNP – Loamy soil with nano particles

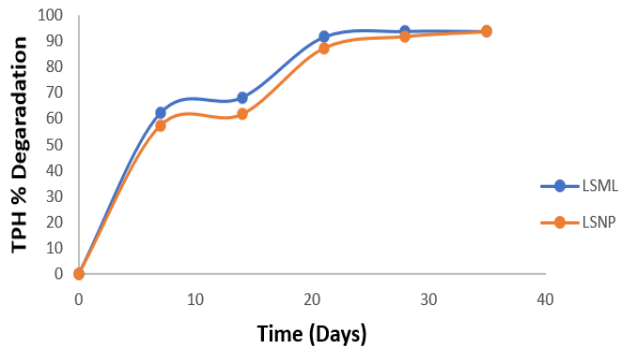


Figure 5: Comparison of TPH removal in the different treatment at 100g.

4.5 Evaluation of TPH Degradation Rate Constants and Half Life

The depletion of TPH in petroleum-entrained specimen was further studied using kinetic model of first order and regression equations for several treatment alternatives. Therefore, utilizing the deduced rate constant, the duration required for the TPHs amounts to decrease to half of its former amounts (half-life) was subsequently assessed.

4.5.1 Evaluation of TPH degradation rate constants and half life using mango leaf

The degradation rate constant (K_d) was ascertained from the linear regression equations in comparison with Equation 7. The identified rate constants along with the initial TPH concentrations was inserted into Equation 8 to yield the final predictive model applied for estimating residual TPH levels within the soil. Thus, the first-order kinetic rate model incorporating the depletion rate constants for the individual remediation options and the projected time (half life) where the TPH levels would decrease by half of its previous amounts for the respective treatment alternatives are depicted in Table 2.

The natural degradation process (control sample) within the polluted loamy soil would necessitate around 770 days to achieve a 50% reduction in TPH concentration. However, the inclusion of 5g mango leaf treatment caused this duration to diminish to approximately 18 days for 50% TPH degradation. The trend of half-life reduction was observed as treatment weight increases to 8 days value for 100g mango leaf treatment weight. The observed trend indicates that augmenting treatment weight leads to a reduction in the needed period for TPH in reducing by half of the former amounts, this is in tandem with Ofeogbu *et al.* (2015) and this deduction is substantiated by the depletion rate constant value that is least in the control specimen but exhibited an increment as procedure level increases.

Table 2: Model equations, rate constants and half life for loamy soil under mango leaf treatments

Weight (g)	Rate Constants (day ⁻¹)	Model Equations	Half-Life (days)
Control	0.0009	$C_{TPH} = 15641e^{-0.0009t}$	770.11
5	0.0378	$C_{TPH} = 14969.90e^{-0.0378t}$	18.33
25	0.0467	$C_{TPH} = 13971.85e^{-0.0467t}$	14.84
50	0.0639	$C_{TPH} = 13714.37e^{-0.0639t}$	10.84
75	0.0789	$C_{TPH} = 13966.26e^{-0.0789t}$	8.78
100	0.0843	$C_{TPH} = 12545.28e^{-0.0843t}$	8.22

4.5.2 Evaluation of TPH degradation rate constants and half life using nano-particle

The degradation rate constant value was deduced from the linear regression equations in tandem with Equation 7. The identified rate constants along with the initial TPH concentrations was inserted into the equation, thereby yielding the predictive model

equation applied for estimating residual TPH levels within the soil. Hence, the first-order kinetic rate model incorporating the degradation rate constants and half life of TPH degradation are shown in Table 3. TPH degradation in polluted loamy soil using nanoparticle showed a similar trend to mango leaf decontamination of TPH in loamy samples with a



reduction in half life from 693days period for control to approximately 9days for nanoparticle 100g weight treatment. Thus, augmenting the treatment weight leads to a reduction in half life needed for TPH to deplete to half of its previous amounts. The depletion of TPH and its depletion rate constant in polluted loamy soil under nanoparticle treatment were least in

the control specimen but rises as the remediation amounts improved. Interestingly, the half-life of polluted loamy soil under both mango leaf and nanoparticle treatments were nearly identical in the control sample.

Table 3: Model equations, rate constants and half life for loamy soil under nanoparticle treatments

Weight (g)	Rate Constants (day ⁻¹)	Model Equations	Half-Life (days)
Control	0.0161	$C_{TPH} = 15615.19e^{-0.001t}$	693.10
5	0.0369	$C_{TPH} = 16015.30e^{-0.0369t}$	18.78
25	0.0431	$C_{TPH} = 14748.54e^{-0.0431t}$	16.08
50	0.059	$C_{TPH} = 14896.77e^{-0.059t}$	11.74
75	0.0767	$C_{TPH} = 15232.70e^{-0.0767t}$	9.03
100	0.081	$C_{TPH} = 13999.82e^{-0.081t}$	8.55

4.6 Evaluation of Michaelis-Menten Constants

An alternative model derived from the Michaelis-Menten equation for determining kinetic rates of substrate concentration in enzyme or microorganism catalysed reactions was employed to further examine the rate of TPH in the loamy soil medium. A Line-Weaver Burke Plot was constructed for mango leaf

and nanoparticle treatments at various options, and with the application of these plots with Equation 9, maximum specific rate constants and Michaelis-Menten constants were deduced as shown in Table 4. These constants were integrated into Equation 10 to predict the dependency of TPH concentration with time.

Table 4: Michaelis-menten constants

Treat. wt. (g)	U _m (mg/l.day)		K _s (mg/l)		R ²	
	LSML	LSNP	LSML	LSNP	LSML	LSNP
Control	3.13E-04	4.00E+02	3.13E+01	1.60E+09	5.60E-03	1.30E-03
5g	1.00E-04	5.00E+03	1.00E+02	1.00E+10	8.72E-02	9.38E-02
25g	1.00E-04	1.00E+04	8.00E+03	9.00E+11	8.42E-02	7.91E-02
50g	1.00E+04	1.00E+04	7.00E+11	8.00E+11	1.16E-01	1.10E-01
75g	1.11E+04	1.00E+04	6.67E+11	7.00E+11	1.38E-01	1.34E-01
100g	1.11E+04	1.11E+04	4.44E+11	5.56E+11	1.28E-01	1.23E-01

LSML – Loamy soil with mango leaves, LSNP – Loamy soil with nano particles

4.7 Comparison of TPH Experimental Results and Model Performance

The residual concentration of TPH obtained from the experimental analysis of different soil treatments with 100g treatment weight using Mango Leaf and

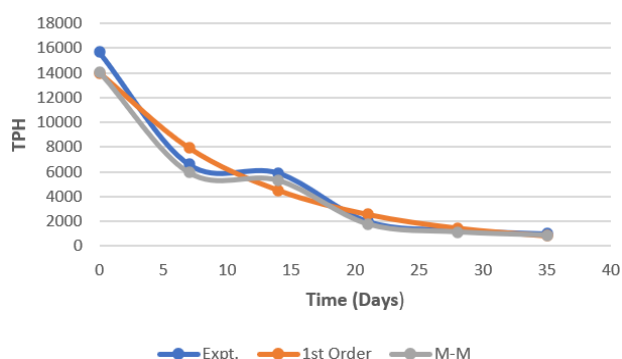
Nanoparticle were compared with the predicted TPH obtained using the first order degradation rate and Michaelis-Menten model equations as shown in Table 5.



Table 5: Comparison of experimental and predicted tph models for 100g treatment

Time (Days)	LSML (ppm)			LSNP (ppm)		
	Expt.	1st Order	M-M	Expt.	1st Order	M-M
0	15666.7	12545.28	13708.36202	15666.7	13999.8	14074.67
7	5871.6	6953.49	5137.649932	6608.2	7941.04	5936.683
14	4914.2	3854.12	4299.924952	5897.9	4504.35	5298.563
21	1318.6	2136.23	1153.774997	1979.7	2554.98	1778.525
28	965.8	1184.05	845.0749982	1273.7	1449.24	1144.268
35	965.8	656.28	845.0749982	987.7	822.04	887.3312

According to the first-order rate kinetics prediction for LSNP, the TPH concentration at stabilization declined from 13999.8ppm to 822.04ppm by the final day, the Michaelis-Menten equation for TPH concentration at stabilization decreased from 14074.67ppm to 887.3312ppm by the final day and the experimental TPH measurements from stabilization stage decline progressively from 15666.7ppm to 965.8ppm on the final day.

**Figure 6:** Comparison of TPH from experiment and model for LSNP

5.0 CONCLUSION

The investigation focused on evaluating the efficacy of Mango leaf and Nano-particle treatments for remediating the contaminated loamy soil specimens. The physicochemical characteristics of the loamy soil underwent notable changes following crude oil pollution, underscoring the significant effect of petroleum on the soil. Specifically, the entrainment by petroleum led to reductions in pH, phosphorus, nitrogen, potassium, and Total Bacteria Counts, while simultaneously elevating electrical conductivity and total organic carbon levels. The examination of bacterial formation under different purification methods unveiled intriguing dynamics. In soils treated with various weights of mango leaf, the Total Bacteria Count (TBC) initially decreased post-pollution, but exhibited substantial population growth as time progressed and remediation level enhanced. Eventually, the rate of expansion of

microbial reached a plateau, while control samples demonstrated sluggish bacterial growth. This disparity underscored the stimulation of bacterial growth by mango leaf treatment. The depletion of TPHs within loamy samples under mango leaf and nano-particle treatments displayed time and weight dependent increments in percentage degradation. Treated soil samples exhibited pronounced degradation rates compared to the slow degradation in control specimens across the polluted soil samples. The TPH degradation percentage trend was further studied for nanoparticle and mango leaf treatments by extrapolating the experimental data for 42nd, 49th and 56th treatment days respectively. The observed TPH percentage degradations are 94.96%, 96.09% and 97.21% for mango leaf while 95.52%, 97.35% and 99.17% for nanoparticle for 42nd, 49th and 56th treatment days respectively. Thus, improved or optimized TPH degradation can be achieved for increased treatment days based on the percentage degradation observed trend. Comparative assessment of diverse treatment options for polluted loamy soil remediation indicated enhanced performance above the 35th day with 100g weight samples demonstrating superior efficacy across the loamy soil. Nano-particle treatment outperformed mango leaf treatment in terms of TPH percentage degradation with 100g nano-particle treatment yielding degradation percentages of 99.17%, compared to 97.21% for mango leaf treatment on the 56th treatment day respectively.

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NOMENCLATURE

Q_o = Inlet volumetric flow rate (kg/day)

Q = Outlet volumetric flow rate (kg/day)

$C_{TPH(o)}$ = Initial concentration of Pollutant (TPH) (mg/kg)

C_{TPH} = Instantaneous concentration of Pollutant (TPH) (mg/kg)

V = Volume of reactor (m^3)

r_{TPH} = Rate of TPH degradation (mg/kg.day)

k_d = TPH degradation rate constant (day^{-1})

t = Time of TPH degradation (day)

$t^{1/2}$ = Half Life

TOC is Total Organic Content

μ_{max} is the maximum specific degradation rate (mg/l.day)

$C_{TPH(t)}$ is TPH concentration (mg/l) with time t , (day)

K_s is Michaelis-Menten constant relating to degradation rate (mg/l).

T = Sample Titration (ml)

B = Blank Titration (ml)

N = Normality of H_2SO_4

S = Sample weight (mg)

TPH_R is the Residual TPH percentage with time

TPH_i is the Initial Concentration of TPH

TPH_f is the Concentration of TPH measured with time.

