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Modeling the Biodegradation of Used Engine Oil in Soil

Ejikeme Ugwoha
Department of Civil & Environmental Engineering, University of Port Harcourt, P.M.B. 5323, Nigeria
Corresponding author's email: ugwohaej@gmail.com
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ABSTRACT: The illegal release of used engine oil (UEO) is an environmental hazard with global concerns. UEO contains heavy metals and hydrocarbons that could modify soil properties and gradually destroy soil usefulness. Thus, soil contaminated with UEO requires remediation to restore its usefulness. In this study, the biodegradation of UEO in soil was examined and modeled. Soil samples were contaminated with UEO and inoculated with cultured bacteria isolated from clogged drainage systems for 56 days. Experimental results indicated that *Pseudomonas, Micrococcus, Bacillus cereus, Providencia* and *Acinetobacter* species actively participated in the biodegradation process. The percentage reduction of UEO was statistically highly significant ($p<0.05$) for all five bacterial species and found to be in the following order: *Pseudomonas* (94.67%) > *Micrococcus* (71.45%) > *Bacillus cereus* (31.00%) > *Providencia* (28.37%) > *Acinetobacter* (28.25%). The biodegradation data complied with first-order kinetic model. Accordingly, first-order kinetic models of the biodegradation of UEO in soil for the five active bacteria were developed. The models were used to fit the biodegradation of UEO in soil with coefficient of determination (R^2) range of 0.6757 – 0.9078 and p-value range of 0.1508 – 0.6704 ($p>0.05$), indicating that the developed models can adequately predict the biodegradation of UEO in soil with time by the respective bacteria.
KEYWORDS: Modeling, Biodegradation, Bacteria, Used engine oil, Contaminated soil

I. INTRODUCTION

Contamination of soil by used engine oil is a common occurrence in most developing countries due to increase in the usage of engine oil [1]. The presence of different types of automobiles, machinery and generating sets have resulted in an increase in the use of engine oil. Engine oil is used for lubrication of internal combustion engines with main functions being to reduce friction and wear on moving parts and to clean the engine from sludge and varnish. It also neutralizes acids that originate from fuel, improves sealing of piston rings, and cools the engine by carrying heat away from moving parts [2]. During its use, engine oil picks up a number of additional components from engine wear, including heavy metals, such as lead, chromium, cadmium, and other materials like naphthalene, chlorinated hydrocarbons and Sulphur [3-4]. This affects the viscosity of engine oil and necessitate its replacement after some time. The engine oil that becomes unsuitable after use due to contamination is called used engine oil (UEO) and is required to be properly disposed of. Unfortunately, in many cases it is discharged into an open drain or thrown into the trash where it can contaminate the soil [3].

The illegal release of UEO is an environmental hazard with global consequences [1,4-6]. UEO contains heavy metals and hydrocarbons that could contribute to chronic hazards including mutagenicity and carcinogenicity [7-8]. It has been recommended to consider UEO as a serious environmental problem due to its chemical composition, world-wide dispersion and effects on the environment [9]. UEO in soil are held mostly in soil pores by capillary forces as residual nonaqueous phase liquids [10], and modifies soil properties due to change in the characteristics of soil pore fluid and its interaction with soil particle [3]. Contamination of soil with UEO has been reported to have altered soil engineering properties [3]. Accordingly, the performance of soil as a supporting medium or construction material has been reported to be affected adversely by contamination with UEO [3].

Contaminated soil requires remediation to make it useful again. Remediation of contaminated soils is a practical necessity with respect to environmental and geotechnical considerations [3]. A number of techniques, namely bioremediation, air sparging, excavation and disposal, washing, thermal treatment, electrokinetics and solidification, etc., are now available for the remediation of contaminated soil [11], but the applicability and feasibility of the various options depend on the field conditions [3]. Several studies on bioremediation have concluded that it is a green technology, cost-effective, simple, innovative and attractive approach for the remediation of petroleum contaminated soils [1,12-15]. Bioremediation is the naturally occurring process by which microorganisms transform environmental contaminants into harmless end-products to obtain carbon and energy [6]. Bioremediation could be performed either as biostimulation or bioaugumentation, depending on the pollution situation and type of microorganisms being used. Biostimulation involves the activation of the indigenous microorganisms in the polluted area by addition of nutrients and providing suitable conditions, while

bioaugumentation involves the addition of oxidizing microorganisms isolated from other sites, or addition of genetically engineered microorganisms [16].

Several studies have been conducted on bioremediation of UEO with focus on the identification of suitable microorganisms and extent of bioremediation [3,6,16-20], but just a few had focused on the modeling of the biodegradation of UEO in soil. Therefore, the aim of this study is to model the biodegradation of UEO in soil.

II. MATERIALS AND METHODS

2.1. Contaminant

Used lube oil popularly called used engine oil (UEO)was used in the study. The UEO was obtained from a motor mechanic workshop adjacent to Londa fuel station at Bori. The UEO was first analysed in the laboratory to determine its total petroleum hydrocarbon (TPH) content.

2.2. Bacteria Acquisition

The bacteria used in this study was obtained from a clogged drainage system. The process was fully described by Ugwoha et al. [20]. But briefly, wet sediment samples were collected from a clogged drainage system using a Grab Sampler. The sampling containers were sterilized to maintain interstitial microbial quality in the samples. Collected samples were labelled and stored in a cooler with ice packs prior to analysis in the laboratory.

2.3Bacteria Identification

The identification of the bacteria used in this study was fully described by Ugwoha et al. [20]. The identification involved bacterial culture, isolation of bacteria colonies, and biochemical tests which include catalase test, methyl red test, oxidase test, indole test, and citrate test. Spectrophotometer was used to determine the number of bacterial colonies while Heterotrophic count of bacterial species was carried out to determine the number of bacterial cells. The bacterial growth process in the cultured solution was used to plot the bacterial growth with time.

2.4Collection of Soil Samples

Loamy soil commonly found in most farmland was used for the experiment. Samples of loamy soil were collected from a depth of 0 to 60cm using a standard auger. Collected soil samples were analysed for indigenous bacteria, hydrocarbon content, pH and temperature to establish baseline condition before use in the experiment. Soil samples were heated at 1200^{o}C to destroy the indigenous bacteria before use in the experiment.

2.5Experimental Setup

A 10g soil sample was mixed with 1ml of UEO to provide known quantity of the soil sample and volume of the contaminant as well as the TPH. The contaminated soil sample was thoroughly mixed to ensure that the contaminant was evenly distributed in the soil sample. The experiment was set up in glass beaker. Eleven (11) experiments were set up, one for each bacterium. One (1) control was also setup, given a total of 12 setups for the study. Isolated bacteria solution (9ml) was inoculated into the 11 experimental setups while no inoculation was performed to the control. Exactly 0.5g of nitrogen and 0.5g of phosphorus were mixed in 100ml of distil water and 1ml was transferred into each sample to provide nutrients for the bacteria. The 12 setups were monitored for a period of fifty-six (56) days and samples were taken from each on a weekly basis and analysed for TPH contents.

2.6 Data analysis

Analysis of variance (ANOVA) was performed to determine the significant level of biodegradation by each active bacterium isolates. The percentage of total petroleum hydrocarbon (TPH) biodegraded was computed using Equation (1).

$$\theta_R = \frac{TPH_0 - TPH_t}{TPH_0} \times 100 \qquad (1)$$

where TPH_0 is the initial concentration of UEO in soil and TPH_t is the residual concentration of UEO in soil at time t.

2.7 Model development

The modeling of the biodegradation of UEO in soil followed the concept presented by Ugwoha and Iwuchukwu [21]. The kinetic relationship between the reaction rate and the rate of change of UEO with time in the soil was described using Equation (2). The order of kinetic reaction was determined using Equation (3).

Since the biodegradation of contaminants in soil follows first-order kinetics, the degradation of UEO in the soil with time was expressed as Equations (4) to (7). The half-life of UEO in the soil was computed using Equation (8).

$$r = \frac{\Delta TPH}{\Delta t} \tag{2}$$

where r is the rate of reaction, t is the time in day, ΔTPH is the change in UEO concentrations.

$$r = \frac{dC}{dt} = -kC^n \tag{3}$$

where r is the rate of reaction, C is the concentration of UEO (mg/kg) remaining at any time (t), n is the reaction order, and k is the kinetic rate constant.

Putting n = 1 as appropriate for a first-order kinetic reaction, replacing C with TPH and integrating with initial conditions, t = 0, t = t; C = 0, C = C_t gives:

$$\ln(TPH_t) = \ln(TPH_0) - k_1 t \tag{4}$$

$$\ln\left(\frac{TPH_t}{TPH_0}\right) = -k_1 t \tag{5}$$

$$\frac{TPH_t}{TPH_0} = e^{-k_1 t} \tag{6}$$

$$TPH_t = TPH_0 e^{-k_1 t} \tag{7}$$

where TPH_t is the concentration of UEO (mg/kg) remaining at any time (t), TPH_0 is the initial concentration of UEO (mg/kg), and k_1 is the first-order reaction rate.

$$t_{1/2} = \frac{\ln(2)}{k_1} \tag{8}$$

where k_1 is the first-order reaction rate.

III. RESULTS AND DISCUSSION

3.1 Biodegradation of UEO by Bacterial Species

A total of eleven (11) bacterial species (*Bacillus cereus, Micrococcus, Staphylococcus, Salmonella, Escherichia coli, Proteus, Vibrio cholerae, Shigella, Providencia, Acinetobacter* and *Pseudomonas*) were identified from the clogged drainage wet sediment samples but only five (5) bacterial species (*Bacillus cereus, Micrococcus, Providencia, Acinetobacter* and *Pseudomonas*) actively participated in the bioremediation process (Fig. 1). The existence of these bacterial species in clogged drainage sediment samples have been reported [20,22-23]. The percentage reduction of TPH on day 56 was statistically highly significant ($p < 0.05$) for all five bacterial species and found to be in the following order *Pseudomonas* > *Micrococcus* > *Bacillus cereus* > *Providencia* > *Acinetobacter* (Table 1). Surajudeen [24]) achieved 75% removal of UEO within 70 days of treatment with bacteria.

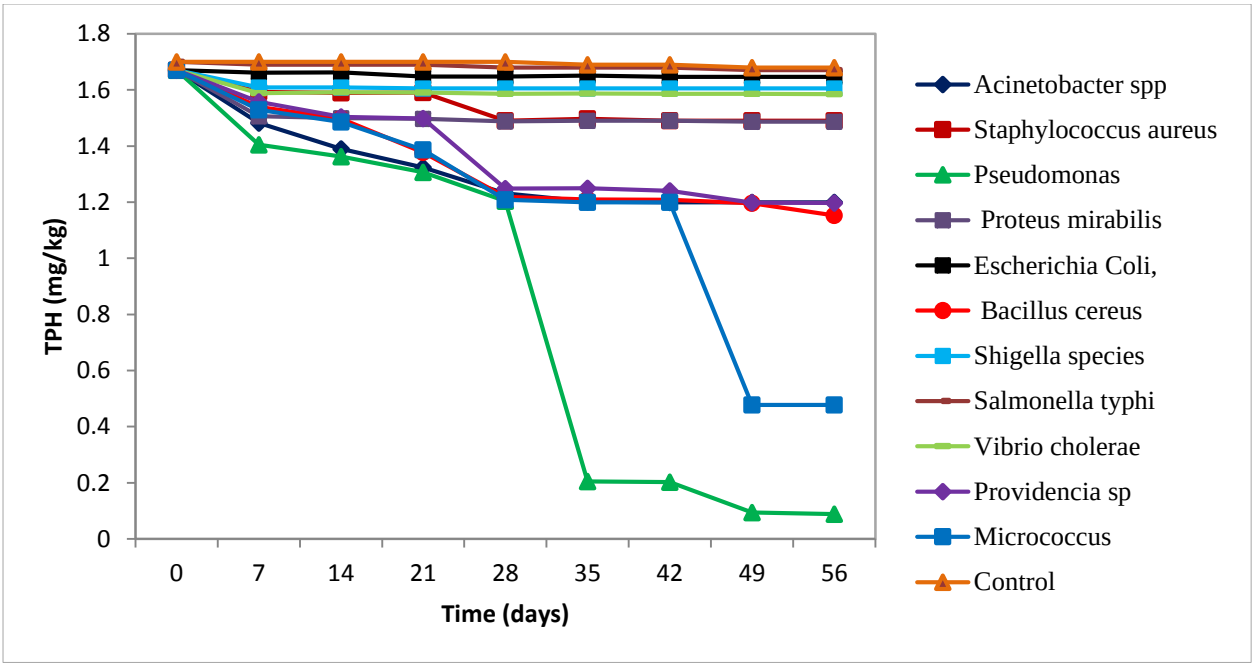

Fig. 1: Biodegradation of UEO in soil by all bacteria isolates

Table 1: Percentage biodegradation of UEO in soil by active bacteria isolates

Bacterial type	% biodegradation	P-value	Level of significance
Pseudomonas	94.67	0.002	Significant
Micrococcus	71.45	0.007	Significant
Bacillus cereus	31.00	$9.3x10^{-5}$	Significant
Providencia	28.37	0.0001	Significant
Acinetobacter	28.25	$1.3x10^{-6}$	Significant

3.2Kinetics of Biodegradation of UEO in Soil

Figures 2 to 6 present linear plots obtained from linear regression, and from these figures coefficient of determination (R^2) values in Table 2 were generated. The linear plots show the first-order kinetic rate constant for each bacterium. The high values of the coefficient of determination ($R^2 > 0.7$) imply the attainment of a good first-order kinetic rate constant for the hydrocarbon utilization bacteria (HUB). The slope of the plot is the first-order kinetic constant (k_1) of Equation (5). This observation agrees with previous studies [25-26]. The values of the reaction rate constant show that *Pseudomonas* degraded the UEO in the soil more efficiently than the other bacteria (*Micrococcus* > *Bacillus cereus* > *Providencia* > *Acinetobacter*).

The biodegradation reaction order (n) for the HUB was obtained as the exponent of the reaction rate constant using Equation (3) and rounded up to a whole number following Yudono et al. [26]. The value of n for all five (5) bacteria rounded up to 1, indicating first-order kinetic. The values of reaction rate constant were substituted into Equation (7) to obtain the first-order kinetic models in Table 3. The UEO biodegradation half-life for each bacterium was computed using Equation (8). The kinetic parameters of the first-order degradation models (Table 3) show that the highest rate of UEO degradation occurred using *Pseudomonas* ($k_1 = 0.0607\ day^{-1}$) with 94.67% removal efficiency and half-life of 11 days while the least occurred using *Acinetobacter* ($k_1 = 0.0056\ day^{-1}$) with 28.25% removal efficiency and half-life of 124 days.

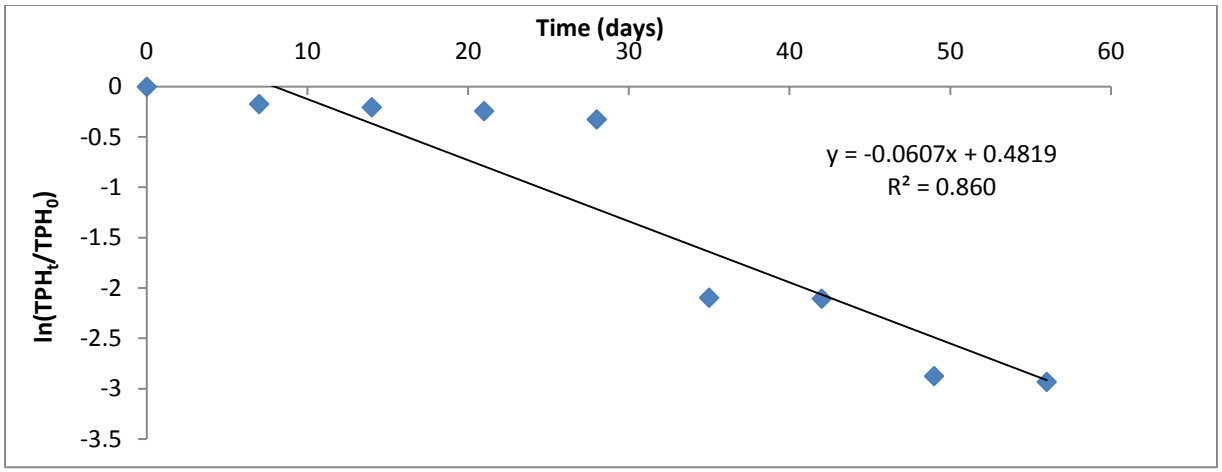

Fig. 2: First-order kinetic rate constant determination for *Pseudomonas* biodegradation of UEO

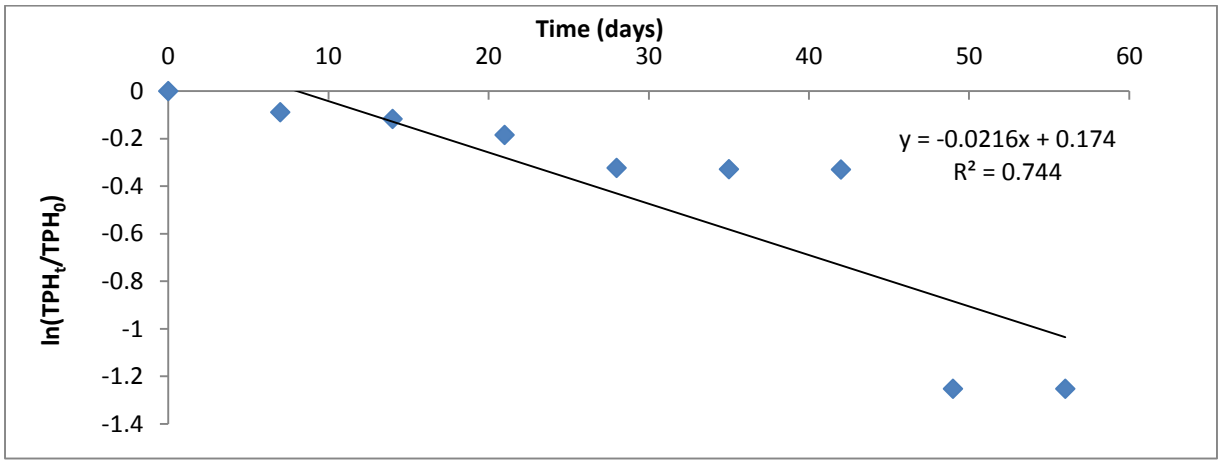

Fig. 3: First-order kinetic rate constant determination for *Micrococcus* biodegradation of UEO

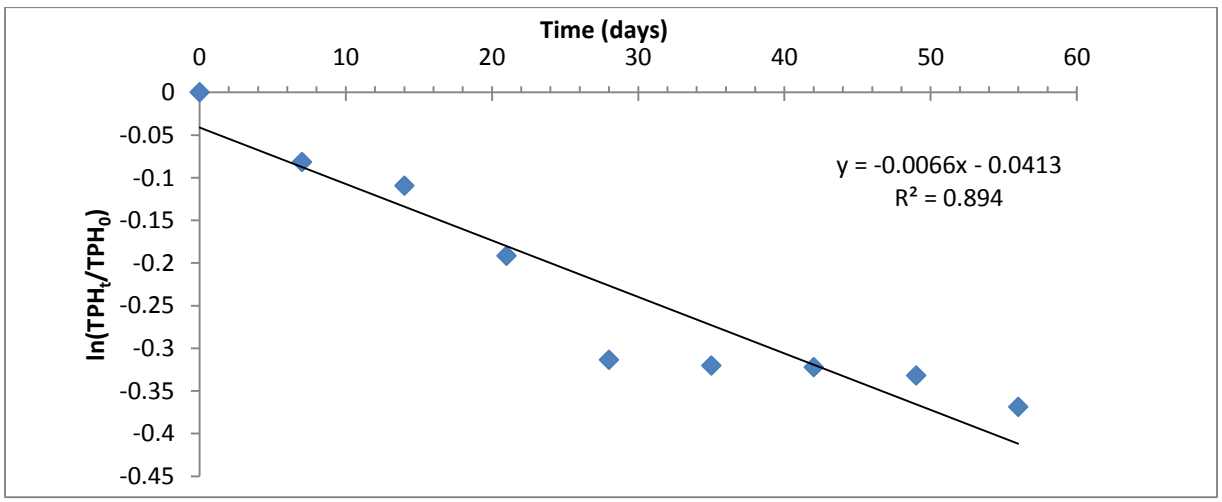

Fig. 4: First-order kinetic rate constant determination for *Bacillus cereus* biodegradation of UEO

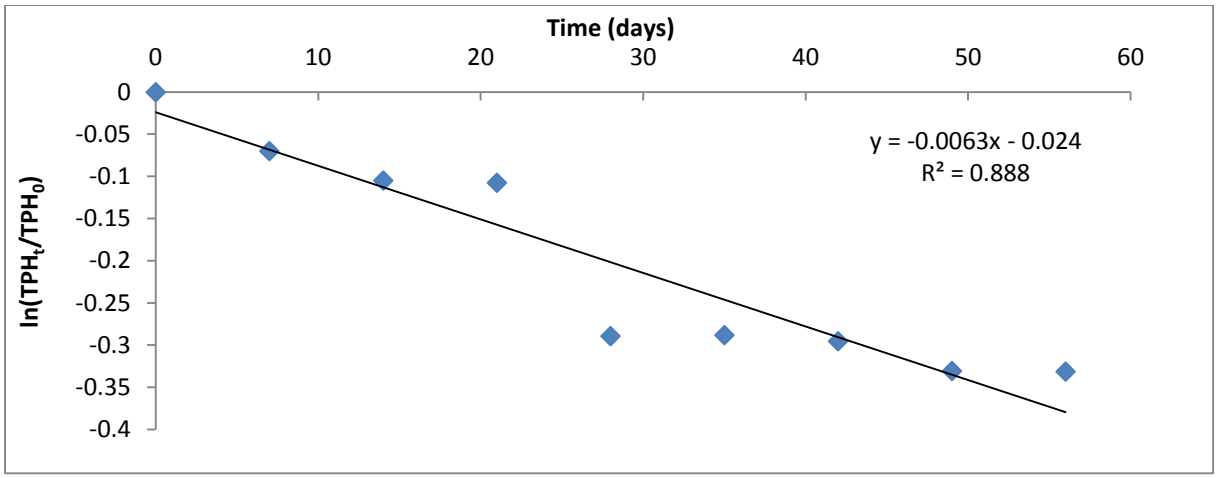

Fig. 5: First-order kinetic rate constant determination for *Providencia* biodegradation of UEO

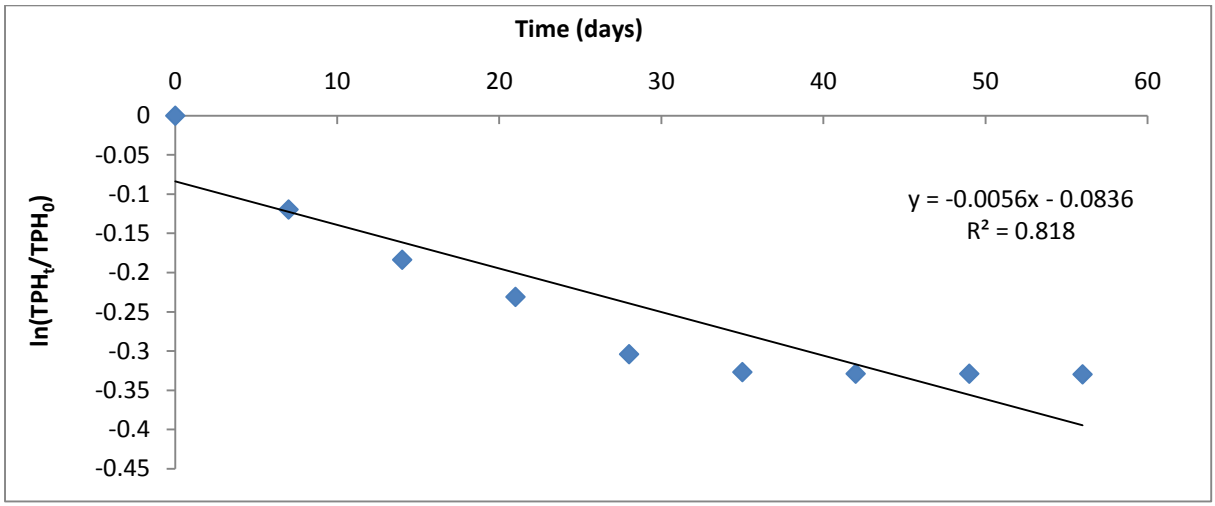

Fig. 6: First-order kinetic rate constant determination for *Acinetobacter* biodegradation of UEO

Table 2: Summary of biodegradation kinetics parameters for UEO

Bacteria type	**Kinetic parameters**		
	Biodegradation reaction order (n)	**k_1 (day^{-1})**	**R^2**
Pseudomonas	1.0626	0.0607	0.860
Micrococcus	1.0218	0.0216	0.744
Bacillus cereus	1.0066	0.0066	0.894
Providencia	1.0063	0.0063	0.888
Acinetobacter	1.0056	0.0056	0.818

Table 3: Summary of biodegradation prediction models and half-life for UEO in soil

Bacteria type	**First-order kinetic models**	**Half-life, $t_{1/2}$ (days)**
Pseudomonas	$C_t = 1.671e^{-0.0607t}$	11
Micrococcus	$C_t = 1.671e^{-0.0216t}$	32
Bacillus cereus	$C_t = 1.671e^{-0.0066t}$	105
Providencia	$C_t = 1.671e^{-0.0063t}$	110
Acinetobacter	$C_t = 1.671e^{-0.0056t}$	124

3.3 Model Validation

The validation of the biodegradation models for individual bacterium using the relationship between the measured and predicted TPH concentrations are presented in Figures 7 to 11. The high coefficient of determinations ($R^2 > 0.6$) obtained in all cases indicate good agreement between the measured and predicted biodegradation of UEO in soil. More so, the p-values ($p > 0.05$) obtained in all cases imply that there is no significant difference between the measured and predicted UEO biodegradation. The accuracy of prediction of biodegradation of UEO in soil by the bacteria was found to be in the following order: *Bacillus cereus* (R^2 = 0.9078) > *Providencia* (R^2 = 0.9010) > *Acinetobacter* (R^2 = 0.8283) > *Micrococcus* (R^2 = 0.7548) > *Pseudomonas* (R^2 = 0.6757).

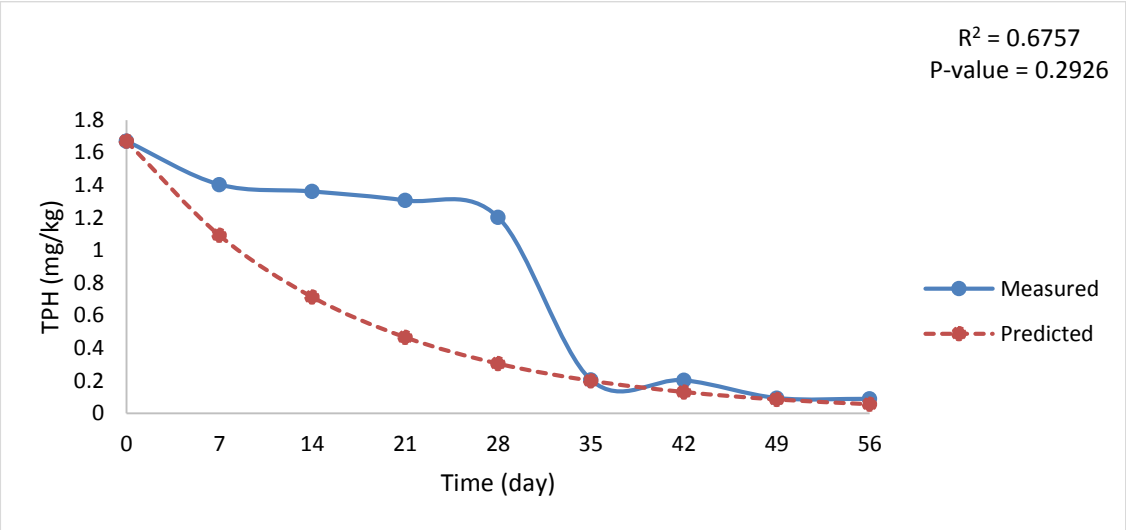

Fig. 7: Measured and predicted biodegradation of UEO by *Pseudomonas*

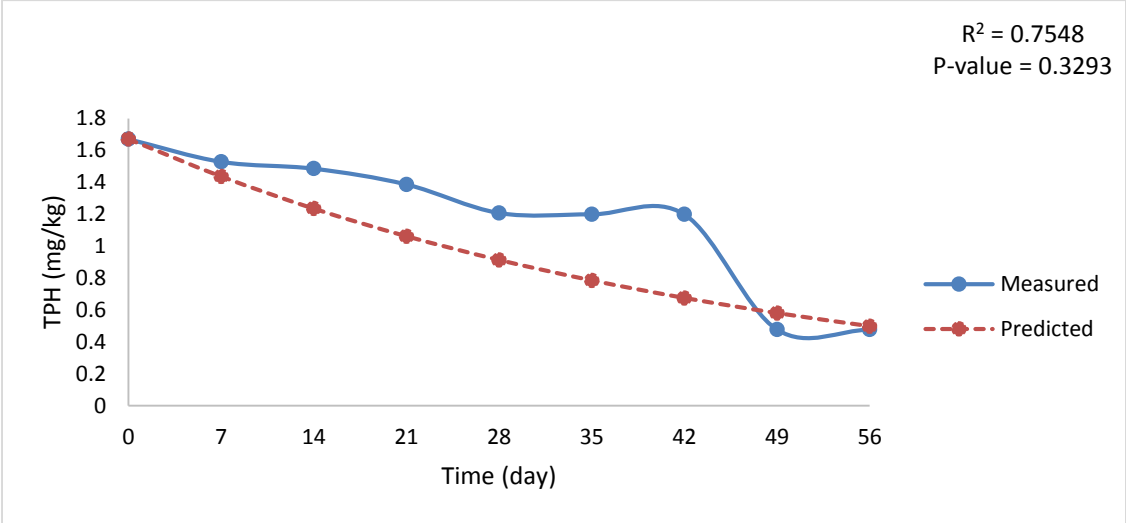

Fig. 8: Measured and predicted biodegradation of UEO by *Micrococcus*

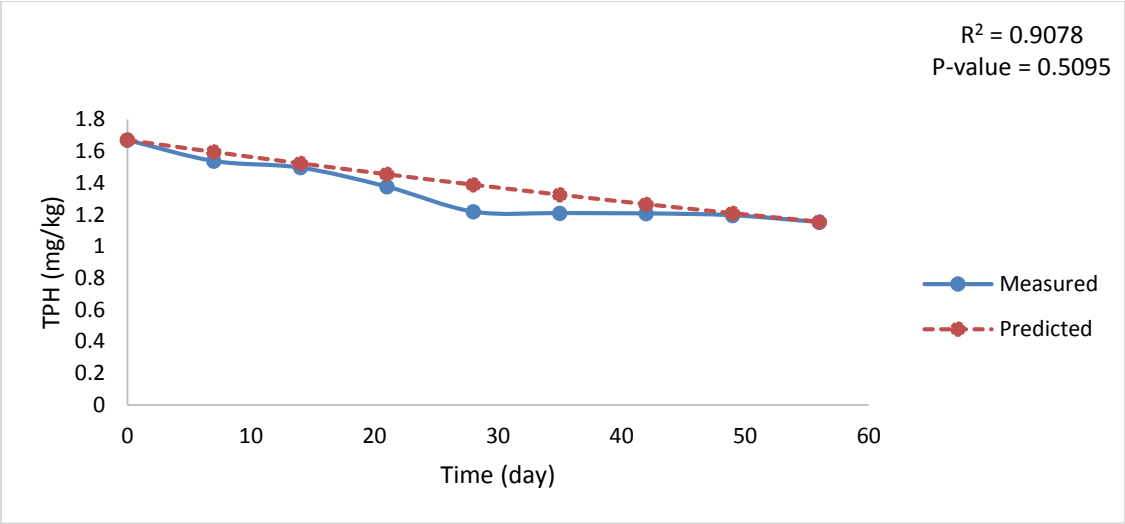

Fig. 9: Measured and predicted biodegradation of UEO by *Bacillus cereus*

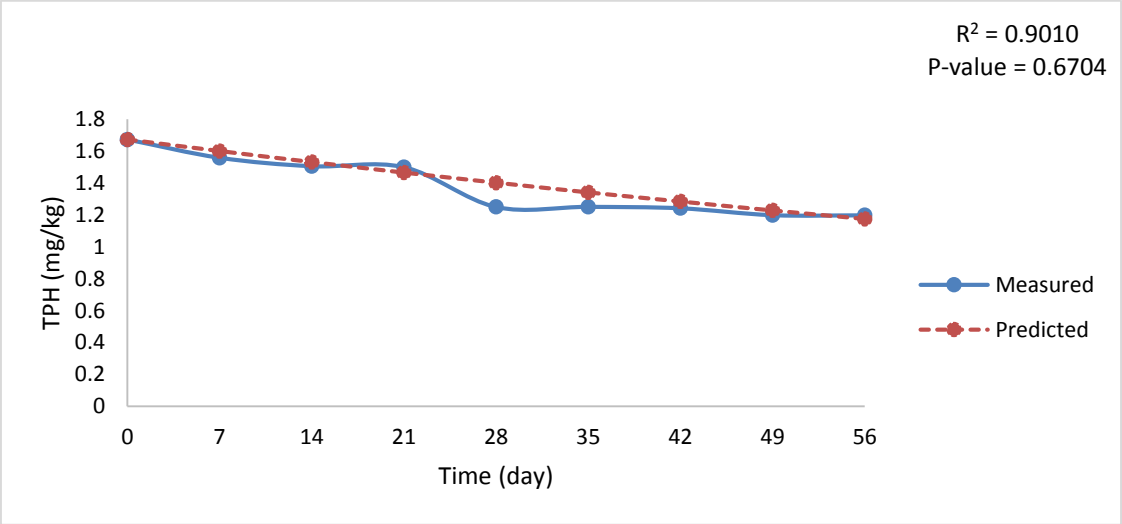

Fig. 10: Measured and predicted biodegradation of UEO by *Providencia*

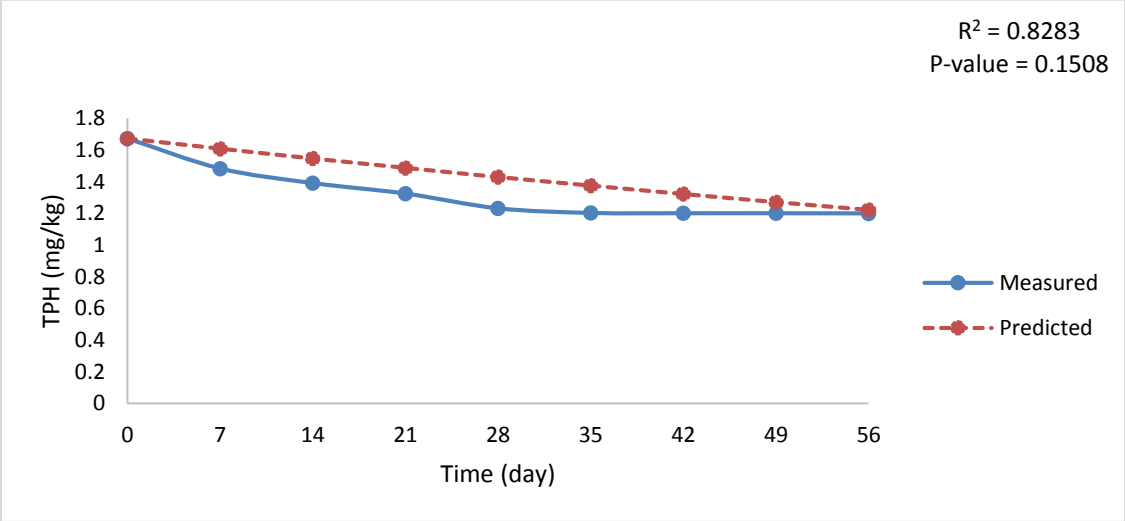

Fig. 11: Measured and predicted biodegradation of UEO by *Acinetobacter*

IV. CONCLUSION

Bacterial species such as *Pseudomonas*, *Micrococcus*, *Bacillus cereus*, *Providencia* and *Acinetobacter* can biodegrade used engine oil (UEO) in soil and achieve the following percentage reduction: 94.67% (*Pseudomonas*), 71.45% (*Micrococcus*), 31.00% (*Bacillus cereus*), 28.37% (*Providencia*) and 28.25% (*Acinetobacter*). The biodegradation of UEO in soil can be predicted by first-order kinetic model developed from measured bacteria-specific data. Such first-order kinetic model can be used to predict the degree of natural attenuation of UEO in soil with any of the studied bacteria.

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