

Bioremediation of Crude Oil-Contaminated Loamy Soil Using Plantain (*Musa paradisiaca*) Stem Waste as a Biostimulant

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Abstract

*Crude oil contamination of soil is a pressing environmental challenge in oil-producing regions, especially in the Niger Delta of Nigeria. It degrades soil fertility, disrupts microbial ecosystems, and poses long-term ecological and health risks. Traditional remediation methods are often cost-prohibitive and environmentally intrusive, prompting the search for sustainable alternatives. This study investigates the potential of plantain (*Musa paradisiaca*) stem waste—a readily available agricultural byproduct—as a biostimulant to enhance the biodegradation of crude oil in loamy soil. The experiment involved treating artificially contaminated loamy soil with 100 ml of crude oil and different dosages (20 g–100 g) of either room-dried or sun-dried plantain stem powder. Each treatment was incubated in a controlled setup for 35 days. Parameters assessed included total petroleum hydrocarbon (TPH) levels, microbial counts (hydrocarbon-utilizing bacteria), soil pH, and nutrient content (nitrogen, phosphorus, and potassium). Analytical techniques such as Gas Chromatography-Flame Ionization Detection (GC-FID), Kjeldahl digestion, and atomic absorption spectrophotometry were employed. The results revealed a marked decrease in TPH concentrations across all treatments, with the highest degradation observed in the 100 g plantain stem-amended soils. Nutrient levels increased, especially in higher concentration treatments, indicating enhanced microbial activity and organic matter decomposition. Microbial counts confirmed a proliferation of hydrocarbon-degrading bacteria, particularly in sun-dried treatments. Kinetic modeling using the Michaelis–Menten equation further validated the efficiency of the bioremediation process. This study concludes that plantain stem waste is an effective, low-cost biostimulant for crude oil remediation in loamy soils. Its use promotes microbial activity, enhances soil fertility, and aligns with sustainable waste management practices. The findings support the feasibility of using agricultural residues in eco-friendly soil restoration strategies, especially in regions burdened by oil pollution. Field-scale trials are recommended to optimize dosage, application methods, and long-term environmental effects.*

Keywords: Bioremediation, biostimulant, crude oil contamination, loamy soil, Nigeria, plantain stem waste, TPH degradation

INTRODUCTION

Crude oil leaks and spills pose serious threats to environmental integrity and public health, often leading to the destruction of soil ecosystems and disruption of natural ecological processes. The frequency and severity of soil contamination incidents have increased since the onset of crude oil exploration and transportation, particularly in oil-producing regions.

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Received Date: January 30, 2026
 Accepted Date: January 31, 2026
 Published Date: February 27, 2026

Citation: Obomanu Cecilia Alex. Bioremediation of Crude Oil-Contaminated Loamy Soil Using Plantain (*Musa paradisiaca*) Stem Waste as a Biostimulant. Emerging Trends in Chemical Engineering. 2026; 13(1): 1–11p.

Human populations are intricately connected to their surrounding environment and rely on essential resources such as air, water, and food. Environmental components, including soil, water, air, and biodiversity, are interconnected, and

degradation often impacts others. According to Haller (2017) [1] and Nellemann and Corcoran (2010) [2], over 60% of the world's ecosystems are adversely affected by pollution, habitat fragmentation, unsustainable resource use, and invasive species. Soil, which is responsible for providing nutrients to approximately 95% of the global food production, plays a fundamental role in sustaining life (FAO, 2015). Numerous studies have documented health hazards associated with exposure to contaminated soils from industrial activities, highlighting their impact on both human and animal populations. These impacts often render the land unsuitable for agricultural and commercial use. Traditional remediation methods, such as excavation and landfilling, are often expensive, environmentally invasive, and fail to fully restore the soil.

Crude oil pollution extends beyond the soil and affects air and water through gas flaring and the release of volatile compounds. Oil spills from pipelines, tanks, and storage facilities are major sources of land contamination. Emerging studies indicate that certain plant species, including *Musa paradisiaca* (plantain), possess the unique ability to tolerate and degrade hydrocarbons, making them promising bioremediation agents. These plants can absorb and metabolize petroleum hydrocarbons through specialized physiological and biochemical mechanisms in their roots and stems.

Additionally, the fibrous porous structure of plantain stems supports microbial colonization, fostering the activity of hydrocarbon-degrading bacteria and fungi. These microorganisms work synergistically with plants to break down complex hydrocarbons into simpler and less toxic compounds, thereby enhancing soil detoxification and nutrient cycling. This natural process, known as bioremediation, employs biological agents such as microbes, enzymes, and organic waste to transform pollutants through metabolic pathways. Compared to traditional methods, bioremediation is more cost-effective, environmentally sustainable, and adaptable [3].

Key bioremediation techniques include bio-augmentation, which introduces specific microorganisms to contaminated sites, and bio-stimulation, which involves the use of organic amendments, such as plant or animal waste, to stimulate indigenous microbial activity. In this context, plant stem waste serves as both a biostimulant and a substrate, combining the benefits of microbial support with phytoremediation potential.

Significant progress has been made in understanding plant-based remediation processes, including pollutant degradation dynamics and optimization strategies. Experimental studies, including laboratory setups, greenhouse trials, and field applications, have evaluated the effectiveness of various plants in different soil types and at different contamination levels. In Nigeria, where crude oil pollution remains a major ecological issue, effective remediation is essential to preserve ecosystem functionality. While chemical and physical remediation approaches are available, they are often limited by economic, logistical, and environmental constraints [4]. The use of agricultural residues, such as plant stem waste, presents a sustainable, low-cost alternative that addresses both environmental and waste management concerns [5, 6].

MATERIALS AND METHODS

Study Area and Sample Collection

Soil samples were collected from the Research and Demonstration Farm of the Rivers State University. Light, bright crude oil was obtained from NNPC Port Harcourt. Plantain stems were collected from an agricultural farm in Igbo Etche, Etche LGA, River State.

Experimental Design

Two sets of treatments were prepared using room-dried and sun-dried plant stem powder. Five treatments (T1–T5) were created for each drying method using 20 g, 40 g, 60 g, 80 g, and 100 g of biostimulant, with 2 kg of loamy soil and 100 mL of crude oil per treatment.

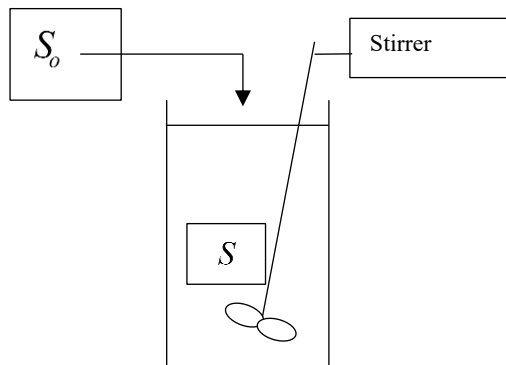


Figure 1. Schematic of the batch reactor.

Each setup was incubated in plastic bioreactors for 35 days. Microbial activity is enhanced by aeration twice a week [7–9].

Analytical Procedures

- *pH*: Measured using a digital pH meter in 1:2.5 soil-water suspensions.
- *Particle size*: Determined using the hydrometer method.
- *Total nitrogen*: Measured using modified Kjeldahl digestion.
- *Phosphorus*: Measured with the ascorbic acid method via spectrophotometry.
- *Potassium*: Analyzed using flame atomic absorption spectrophotometry.
- *Total petroleum hydrocarbon (TPH)*: Determined using GC-FID based on USEPA 8015 and ultrasonic extraction per USEPA 3550C.
- *Hydrocarbon-utilizing bacteria (HUB)* were counted using serial dilution and plate count techniques, and the results were expressed as cfu/g.

Biokinetic Model Application

The Michaelis–Menten equation was used to model the rate of degradation of soil TPH biostimulated with plantain stems. Because the experiment was conducted as a small-scale pilot study using batchwise expression, the rate equation for the batch reactor was first expressed before being merged with an expression for simple exponential decay. Figure 1 shows the batch reactor used to forecast soil TPH concentration at any given moment [10].

The batch model can be expressed using the mass balance Equation (1).

$$\left[\begin{array}{c} \text{Rate of} \\ \text{TPH flow} \\ \text{into} \\ \text{reactor} \end{array} \right] + \left[\begin{array}{c} \text{Rate of} \\ \text{TPH out} \\ \text{flow from} \\ \text{reactor} \end{array} \right] + \left[\begin{array}{c} \text{Rate of TPH} \\ \text{depletion} \\ \text{within} \\ \text{reactor} \end{array} \right] + \left[\begin{array}{c} \text{Rate of TPH} \\ \text{accumulation} \\ \text{with reactor} \end{array} \right] = \quad (1)$$

Let,

$$\text{TPH}_{\text{in}} = F_o S_o \quad (2)$$

$$\text{TPH}_{\text{out}} = FS \quad (3)$$

$$\text{TPH}_{\text{dep}} = -rV \quad (4)$$

$$\text{TPH}_{\text{acc}} = -\frac{dN}{dt} \quad (5)$$

Substituting equations (2–5) into mass balance expression (1) yields

$$FoSo = FS - rV - \frac{dN}{dt} \quad (6)$$

Where,

S_o = Initial concentration of TPH (mg/kg)

S = Instantaneous concentration of TPH (mg/kg)

F_o = Inlet flow rate (L/day)

F = Outlet flow rate (L/day)

V = Volume of reactor (L)

N = Mass of TPH (mg)

t = Time of remediation (day)

For the batch reactor, the inflow and outflow of mass are zero; hence, Equation (6) is reduced to

$$\frac{dN}{dt} = -rV \quad (7)$$

By expressing Equation (7) in terms of concentration, we have:

$$\frac{d\left(\frac{N}{V}\right)}{dt} = \frac{dS}{dt} \quad (8)$$

Thus, Equation (8) becomes:

$$\frac{dS}{dt} = -r_s \quad (9)$$

But the growth rate decay is expressed as:

$$-r = \frac{\mu_{\max} S}{K_s + S} \quad (10)$$

where

r = TPH degradation rate (mg/kg.day)

$\mu_{\max}$ = Maximum specific rate constant (mg/kg.day)

K_s = Constant (mg/kg)

Substituting Equation (9) into (10), we have as follows:

$$-r = \frac{dS}{dt} = \frac{\mu_{\max} S}{K_s + S} \quad (11)$$

To obtain the constants in Equation (11), we take the inverse of both sides.

$$\frac{1}{-r} = \frac{K_s}{\mu_{\max}} \left(\frac{1}{S} \right) + \frac{1}{\mu_{\max}} \quad (12)$$

A plot of $\frac{1}{-r}$ versus $\frac{1}{S}$ gives the slope of the graph as $\frac{K_s}{\mu_{\max}}$, whereas the intercept represents.

$$\frac{1}{\mu_{\max}}$$

Upon determination of the constants, it is then substituted into Equation (12), which can be used to predict the degradation rate of TPH.

RESULTS AND DISCUSSION

Characteristics of Some Materials Used in Setting up the Experiment

Table 1 presents the physicochemical properties of the loamy soil in both its natural (uncontaminated) and contaminated states, as well as those of the crude oil used in the study, identified as Bonny Medium. The analyzed parameters included electrical conductivity ($\mu\text{S}/\text{cm}$), pH, potassium (K, mg/kg), total nitrogen (TN, mg/kg), phosphorus (P, mg/kg), total organic carbon (TOC, mg/kg), and moisture content (%) [11].

The electrical conductivity values recorded were $301.10 \mu\text{S}/\text{cm}$ for the contaminated loamy soil, $10.36 \mu\text{S}/\text{cm}$ for the uncontaminated loamy soil, and $10.24 \mu\text{S}/\text{cm}$ for the crude oil. The pH values were relatively consistent across samples: 6.07 for the contaminated soil, 6.06 for the uncontaminated soil, and 6.27 for the crude oil.

Total nitrogen (TN) levels were $0.275 \text{ mg}/\text{kg}$ in the contaminated soil, $0.096 \text{ mg}/\text{kg}$ in the uncontaminated soil, and $0.208 \text{ mg}/\text{kg}$ in the crude oil sample. Phosphorus levels showed a significant increase in the contaminated soil ($381.07 \text{ mg}/\text{kg}$), compared to the uncontaminated soil ($15.04 \text{ mg}/\text{kg}$) and crude oil ($1.92 \text{ mg}/\text{kg}$). Potassium (K) concentrations were $16.90 \text{ mg}/\text{kg}$ in the contaminated soil, $29.10 \text{ mg}/\text{kg}$ in the uncontaminated soil, and $4.14 \text{ mg}/\text{kg}$ in the crude oil sample [12].

Table 2 details the characteristics of the plantain stem biostimulants. The pH values of the sun-dried and room-dried biostimulants were 6.14 and 5.75, respectively—both within acceptable ranges for soil treatment. Moisture content analysis revealed 12.04% at 25.7°C for the room-dried sample and 10.27% at 26.5°C for the sun-dried sample.

Table 1. Determination of some physicochemical parameters of raw materials used for the investigation.

Parameters	Contaminated loamy soil	Uncontaminated loamy soil	Crude oil
Electrical conductivity ($\mu\text{S}/\text{cm}$)	301.10	10.36 6.06	10.24
Temp.	-	-	-
pH	6.07	6.06	6.27
TN (mg/kg)	0.275	0.096	0.208
P (mg/kg)	381.07	15.04	1.92
K (mg/kg)	16.9044	29.1031	4.1386
TOC (mg/kg)	7.003	3.050	11.011
Sand (%)	N/A	45	N/A
Silt (%)	N/A	40	N/A
Clay (%)	N/A	15	N/A
T. bacteria (cfu/g)	4.02×10^2	3.75×10^2	3.88×10^2
HUB (cfu/g)	120	69	100
Total moisture content (%)	108	73	22

Note: N/A = not applicable

Table 2. Characteristics of sun-dried and room-dried plantain stem substrate.

Parameters	Room-dried substrate	Sun-dried substrate
EC ($\mu\text{S}/\text{cm}$)	661.3	603.7
Temp.	25.7	26.5
pH	6.14	5.75
TN (mg/kg)	2.15	0.64
TP (mg/kg)	62.932	58.316
K (mg/kg)	181.5	163.1
TOC (%)	12.3	10.7
Moisture content (%)	12.04	10.7

In terms of nutrient content, the room-dried biostimulant showed higher values of nitrogen, phosphorus, and potassium (2.15, 62.93, and 181.5 mg/kg, respectively) compared to the sun-dried sample (0.64, 58.32, and 163.1 mg/kg, respectively) [13].

Figure 2 shows the decrease in TPH concentration due to natural attenuation that occurred at 35 days, recorded at 31814.49 ppm against the initial TPH of 54073.08 ppm.

The results in Figure 3 indicate a slight reduction in the TPH concentration using 20 g of room-dried substrate over time. It was observed that 20 g of room-dried plantain stem substrate inoculated into the bioreactor multiplied the microorganisms in the treated loamy soil.

The results obtained in Figure 4 reveal the TPH behavior with an increase in the T5 (100 g) substrate. The regression graph shows a positive correlation between time and TPH levels, indicating a rapid decline in TPH concentration. Seasonal fluctuations in this trend were also observed, which suggests a decrease in microbial activity. This graph demonstrates a clear dose-response relationship.

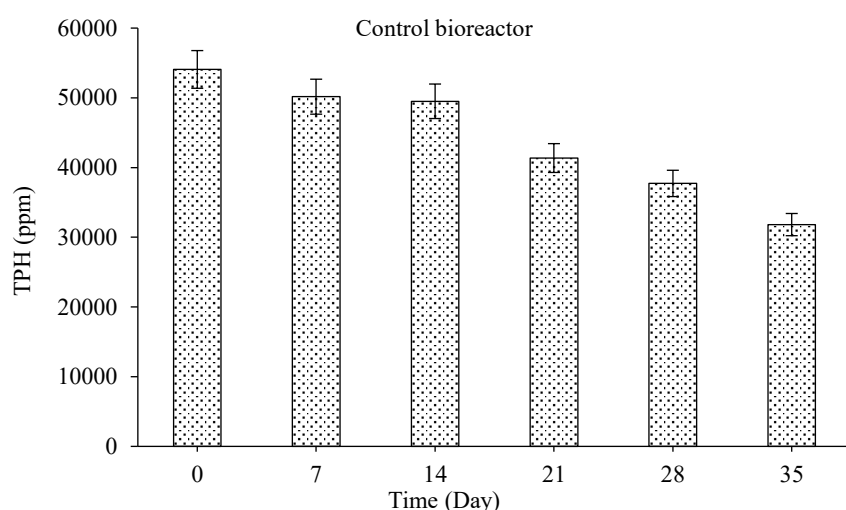


Figure 2. TPH concentration over time for control bioreactors (error bar in percentage).

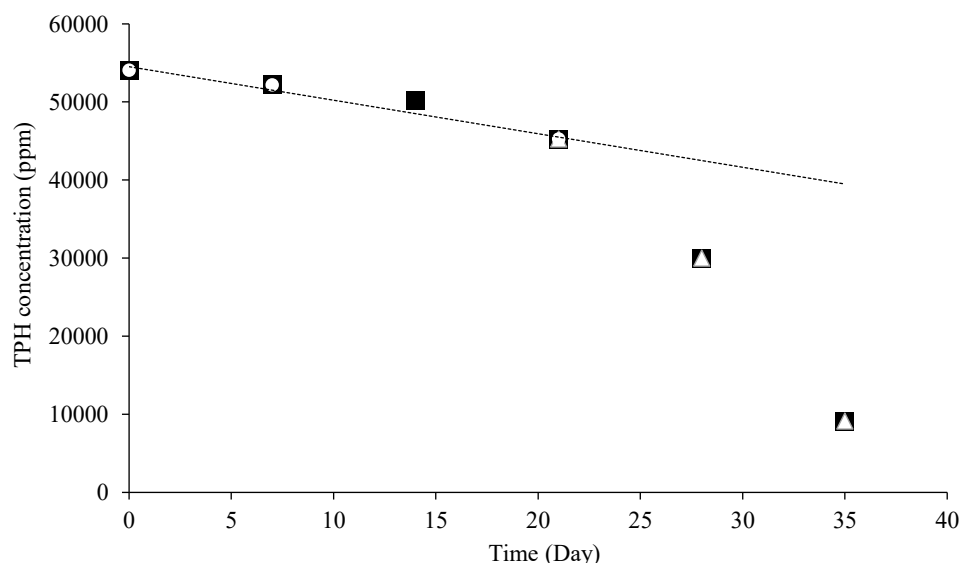


Figure 3. TPH concentration over time for T1 (20 g) room-dried substrate.

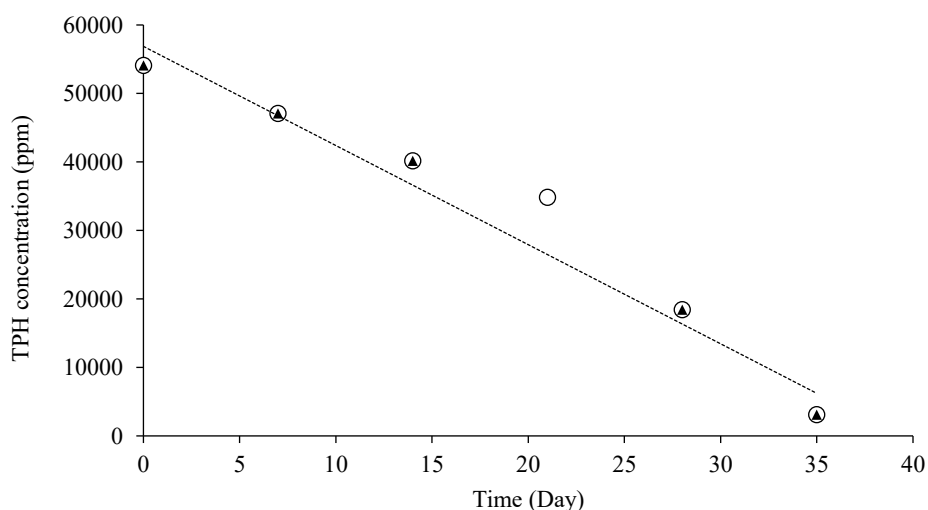


Figure 4. Tph concentration over time for T5 (100 g) room-dried substrate.

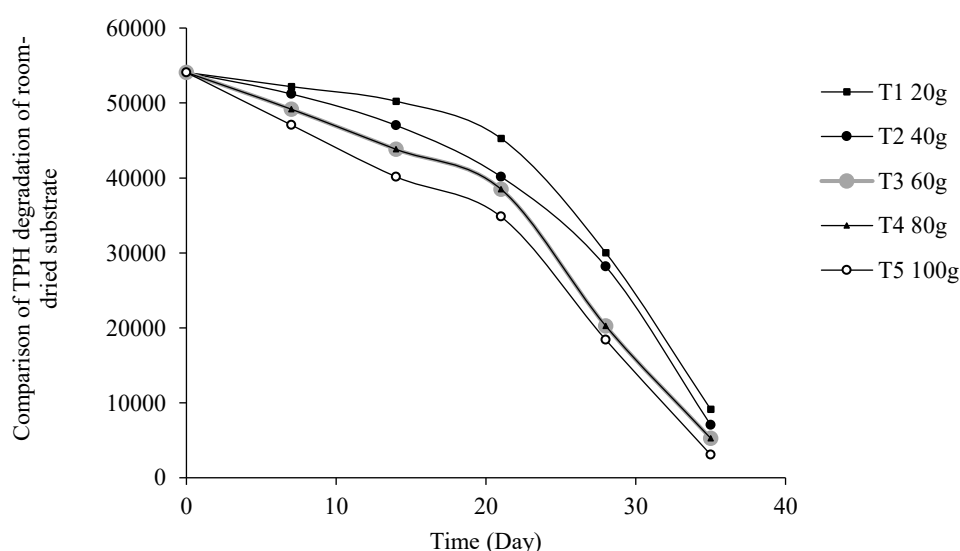


Figure 5. Comparison of TPH concentration among treatment reactors.

This suggests that 98.08%, which is close to one in T5 (100 g), has a strong relationship with the measured data over time. The high reliability of the model for crude oil remediation using plant extract of guava leaf [14].

Figure 5 shows a graphical comparison of TPH degradation of room-dried plantain stem substrates in various treatment reactors monitored for 35 days. The percentage of TPH degradation increased with time across the treatment formulations; however, the rate of degradation differed for each treatment. The increase in biostimulant of plantain stems T1 (20 g), T2 (40 g), T3 (60 g), T4 (80 g), and T5 (100 g) in the bioreactor decreased the TPH concentration over time.

The percentages of TPH degradation in the loamy soil were 98.38%, 96.32%, 99.96%, 98.44%, and 98.08%, respectively. The model has a strong relationship with measured data [15].

Figure 6 shows the correlation value of $R^2 = 0.9964$, expressed as Equation (13)

$$KC_{rd} = 39.041x - 0.0005, \quad (13)$$

Where, KC is the kinetic constant and rd is the room-dried plantain stem substrate for 40 g of plantain stem substrate.

The specific maximum rate constant values (U_m) and Michaelis–Menten constant (K_s) were determined with room-dried plantain stem substrate-treated samples of mass 40 g, using a Lineweaver-Burk plot. The degradation rate of the 40 g plantain stem substrate with contact time obeyed the Lineweaver-Burk plot. It was observed that most TPH degradation occurred with a higher mass of the substrate (100 g) during the 35 d, which significantly affected the removal efficiency [16]. Figure 7 shows the correlation value of $R^2 = 0.9988$, expressed as Equation (14).

$$KC_{rd} = 38.069x - 0.0006, \quad (14)$$

Where, KC is the kinetic constant and rd is the room-dried plantain stem substrate for 60 g of plantain stem substrate.

The specific maximum rate constant values (U_m) and Michaelis–Menten constant (K_s) were determined with room-dried plantain stem substrate-treated samples of mass 60 g, using a Lineweaver-Burk plot. The degradation rate of 100 g plantain stem substrate with contact time obeyed the Lineweaver-Burk plot. It was observed that most TPH degradation occurred with a higher mass of the substrate (100 g) for 35 days, which significantly affected the removal efficiency [17].

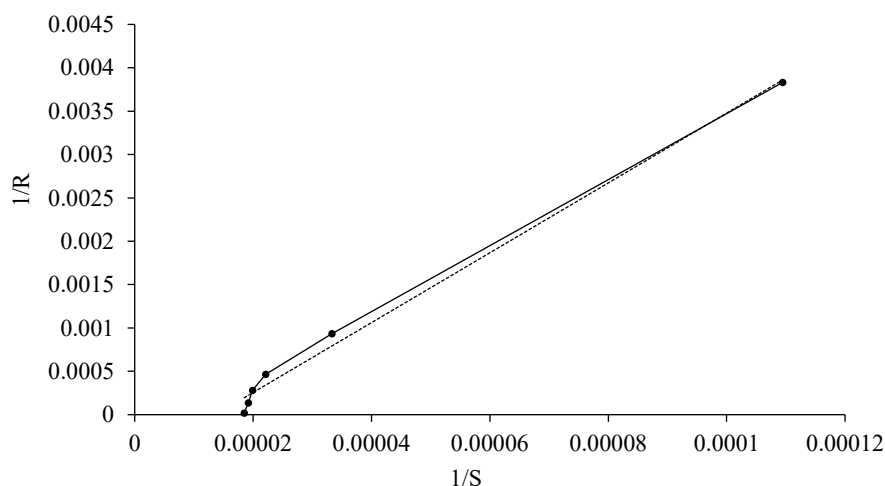


Figure 6. Lineweaver-Burk plot for T1 (20 g) of room-dried plantain stem substrate.

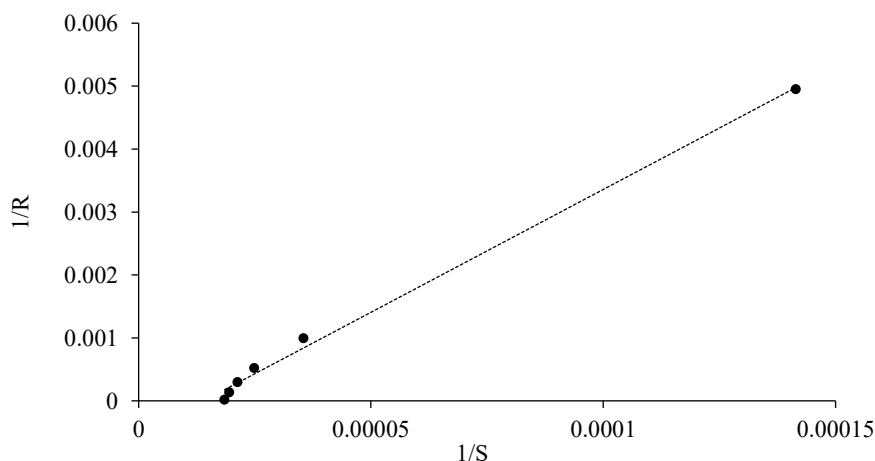


Figure 7. Lineweaver-Burk plot for T2 (40 g) of room-dried plantain stem substrate.

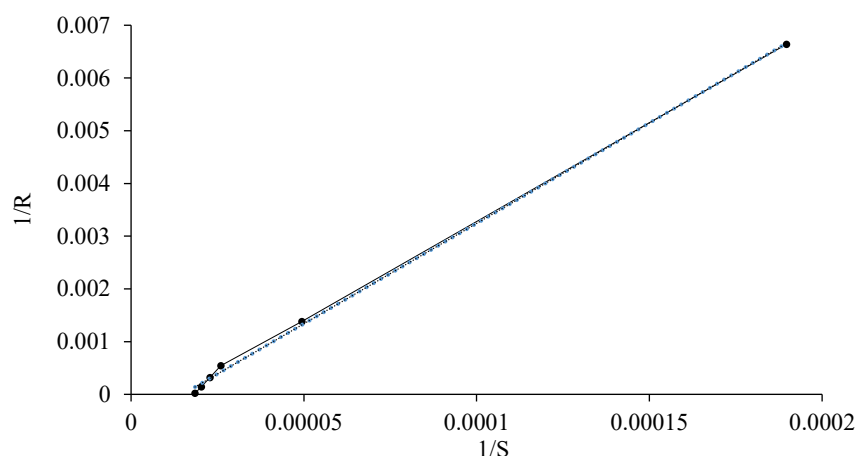


Figure 8. Lineweaver-Burk plot for T3 (60 g) of room-dried plantain stem substrate.

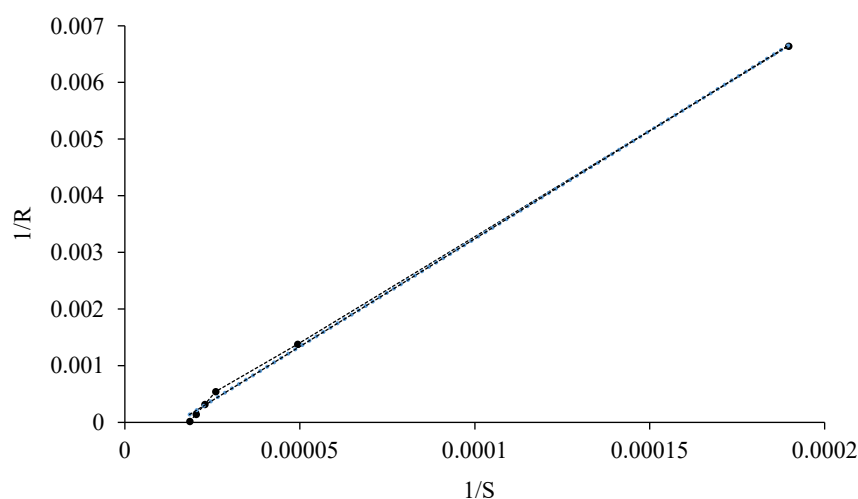


Figure 9. Lineweaver-Burk plot for T4 (80 g) of room-dried plantain stem substrate.

Figure 8 shows the correlation value of $R^2 = 0.9988$, expressed by Equation (15).

$$KC_{rd} = 38.069x - 0.0006, \quad (15)$$

Where, KC is the kinetic constant and rd is the room-dried plantain stem substrate for 80 g plantain stem substrate.

The specific maximum rate constant values (U_m) and Michaelis–Menten constant (K_s) were determined with room-dried plantain stem substrate-treated samples of mass 80 g, using a Lineweaver-Burk plot.

The degradation rate of 60 g plantain stem substrate with contact time obeyed the Lineweaver-Burk plot. It was observed that most TPH degradation occurred with a higher mass of the substrate (100 g) for 35 days, which significantly affected the removal efficiency [18].

The specific maximum rate constant values (U_m) and Michaelis–Menten constant (K_s) were determined with room-dried plantain stem substrate-treated samples of mass T5 (100 g) using the Lineweaver-Burk plot shown in Figure 9. The model shows a correlation value of $R^2 = 0.9997$ in the outcome, as expressed in Equation (16).

$$KC_{rd} = 36.768x - 0.0006 \quad (16)$$

Where, KC is the kinetic constant and rd is the room-dried plantain stem substrate for 100 g of plantain stem substrate.

It was observed that most TPH degradation occurred with a higher mass of the substrate (100 g) for 35 days, which significantly affected the removal efficiency [19].

CONCLUSION

This study evaluated the physicochemical properties of plant stem waste, contaminated and uncontaminated loamy soil, and crude oil. The plantain stem showed high nutrient content, including 2.15 mg/kg of nitrogen, 62.93 mg/kg of phosphorus, and 181.5 mg/kg of potassium, indicating its potential as a biostimulant.

TPH levels decreased significantly in soils amended with plantain stem residue, confirming enhanced microbial activity. Sun-dried biostimulants showed a slightly better degradation performance than room-dried biostimulants, with average correlation coefficients (R^2) of 0.9858 and 0.9824, respectively.

Treatment with sun-dried plantain stems resulted in a more consistent TPH degradation trend over time, whereas room-dried treatments showed more fluctuation. Both the experimental and model-predicted values showed strong agreement, with R^2 values as high as 0.9998 and 0.9997 for room-dried and sun-dried treatments, respectively. The nonlinear regression models demonstrated high precision in predicting TPH degradation over time, validating their effectiveness for future applications.

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