

Bioremediation of crude oil polluted soils using cassava peels and sawdust as biostimulants

Miracle Chika Ogonna¹, Samson Eruke Okoro ^{1,*}, Oghenetega Allen Obewhere ²,
Eka Bassey Essien¹

¹ Department of Biochemistry, [University of Port Harcourt](#), Choba, Rivers State, Nigeria.

Email: eka.essien@uniport.edu.ng

² Department of Chemical and Biomolecular Engineering, [University of Nebraska-Lincoln](#), Lincoln, 68588, Nebraska, United States.

Email: oobewhere2@huskers.unl.edu

* Correspondence: samson.okoro@uniport.edu.ng



ABSTRACT

Petroleum hydrocarbon contamination of soils is a persistent problem in many oil-producing regions, where conventional remediation methods are often too costly or impractical to implement. Biostimulation using locally available organic materials offers a simple and sustainable alternative. In this study, cassava peels and sawdust were evaluated as low-cost agro-waste amendments for the remediation of crude oil-polluted soil. The materials were applied individually and in combination (1:1 ratio) and incubated for 56 days. Soil samples collected on Days 0, 28, and 56 were analyzed for extractable total petroleum hydrocarbons (ETPH), nickel (Ni), cadmium (Cd), and pH. Only minor natural attenuation was observed in the untreated polluted soil, with about 1% ETPH reduction after 56 days. In contrast, cassava peels and sawdust enhanced hydrocarbon degradation, achieving ETPH reductions of 23.1% and 18.7%, respectively. The combined treatment produced the largest reduction (38.6%) and resulted in among the lowest residual Ni and Cd concentrations. Soil pH in amended treatments remained near neutral, creating conditions favorable for microbial activity. Overall, the findings show that agro-waste materials can effectively improve bioremediation performance and provide a practical, low-cost option for managing crude oil-contaminated soils, particularly in resource-limited settings.

Keywords: Bioremediation; ecofriendly; cassava peels; sawdust; total petroleum hydrocarbons; heavy metals; biostimulation; sustainable remediation

INTRODUCTION

Petroleum hydrocarbons are introduced into soil through activities such as crude oil exploration and production, pipeline failures, refining operations, transportation accidents, and improper disposal of petroleum products. In regions with long histories of petroleum extraction, these inputs accumulate over time, resulting in widespread and persistent soil contamination. Once present in soil matrices, petroleum hydrocarbons (PHCs) alter key physicochemical properties, including porosity, nutrient availability, and permeability, thereby reducing soil fertility and impairing ecosystem functions. In addition to these structural impacts, PHCs pose toxicological risks due to the persistence of hazardous fractions such as polycyclic aromatic hydrocarbons (PAHs), which are resistant to natural degradation and can adversely affect terrestrial organisms and human health through prolonged exposure pathways.^{1,2,3} These effects are especially pronounced in oil-producing regions where repeated contamination events and limited remediation capacity allow impacted soils to remain underproductive for extended periods.

A broad range of remediation strategies has been developed to address PHC-contaminated soils, including physical, chemical, thermal, and biological approaches.^{4,5,6} Physical methods, including excavation, soil

washing, and off-site disposal, can provide rapid contaminant removal but often disrupt soil structure and merely transfer contaminants to secondary locations, such as landfills, at considerable economic and environmental cost.^{5,6,7} Chemical remediation techniques, such as chemical oxidation and stabilization, can reduce contaminant concentrations or mobility; however, they require careful site-specific control and may negatively affect indigenous microbial communities that are essential for long-term soil recovery.^{8,9} Thermal methods, including thermal desorption and incineration, are effective for volatile and semi-volatile hydrocarbons but are typically energy-intensive and associated with secondary pollution risks if off-gases are not adequately managed.^{10,11} These limitations are well documented in systematic reviews of remediation technologies for oil-contaminated soils, which consistently highlight the high cost, energy demand, and environmental trade-offs of conventional approaches.⁶

Within this broader remediation landscape, Bioremediation has gained increasing attention because it offers a cost-effective and environmentally compatible approach that supports the recovery of soil function rather than simply removing contaminated material.^{1,3,12} Bioremediation relies on the metabolic capacity of microorganisms to utilize petroleum hydrocarbons as carbon and energy sources, particularly under aerobic conditions where oxidative pathways dominate.¹³ In practice, bioremediation strategies are commonly implemented through natural attenuation, biostimulation, or bioaugmentation.^{13,14} Natural attenuation depends on existing site conditions and typically proceeds slowly, while bioaugmentation introduces specialized microbial strains that may struggle to establish or persist under field conditions. As a result, biostimulation—enhancing indigenous microbial communities through nutrient, organic substrate, oxygen, or moisture additions—is often favored for field-scale applications because it leverages native microorganisms and minimizes ecological disturbance.^{9,15}

Despite its advantages, bioremediation effectiveness is frequently constrained by site-specific limitations, including nutrient deficiencies (particularly nitrogen and phosphorus), insufficient oxygen availability, suboptimal moisture content, and the limited bioavailability of aged or strongly sorbed hydrocarbon fractions.¹⁶ These constraints can significantly slow degradation kinetics even when hydrocarbon-degrading microorganisms are present. Consequently, considerable research effort has focused on identifying low-cost amendment strategies that simultaneously improve soil conditions and stimulate microbial activity. Organic and plant-derived materials, including composts and agricultural residues, have been shown to enhance biodegradation by supplying nutrients, increasing microbial biomass, improving soil structure, and altering contaminant accessibility within the soil matrix.^{1,3,17,18,19}

Agro-waste materials are of particular interest in this context because they are widely available, inexpensive, and compatible with circular economy principles that emphasize waste valorization. Lignocellulosic residues such as sawdust have been reported to enhance petroleum hydrocarbon and PAH degradation by improving soil aeration and providing surfaces for microbial colonization.¹⁹ Similarly, cassava processing residues have been investigated as biostimulants in crude oil-contaminated soils, reflecting their abundance in many oil-producing regions and their capacity to support microbial growth and soil recovery.²⁷ While these materials have shown promise individually, most studies evaluate single amendments in isolation. Fewer investigations directly assess whether combining locally available agro-wastes can produce complementary effects by simultaneously addressing multiple rate-limiting factors, such as nutrient availability and soil physical structure. Field-oriented reviews of remediation technologies consistently emphasize that integrated or combined treatment approaches often outperform single-method strategies by addressing multiple limitations simultaneously and improving overall remediation efficiency.⁶ This perspective is particularly relevant for petroleum hydrocarbon-contaminated soils, where hydrocarbons frequently co-occur with trace metals such as nickel and cadmium, increasing ecological risk and complicating remediation outcomes. Reports in the bioremediation literature indicate that treatment strategies extend beyond petroleum hydrocarbons to include a range of organic and inorganic environmental contaminants, such as heavy metals, chlorinated organics, agricultural pollutants, and other industrial organic compounds (Figure 1).^{1,12,20,21,22,23,24,25,26} Rather than targeting a single contaminant class, these approaches exploit biological processes that can simultaneously influence organic degradation and contaminant mobility by modifying soil properties, including pH, organic matter content, sorption capacity, and microbial activity. The ability to address co-existing organic and inorganic contaminants within

the same soil system underscores the need for integrated remediation strategies when treating crude oil-impacted soils.

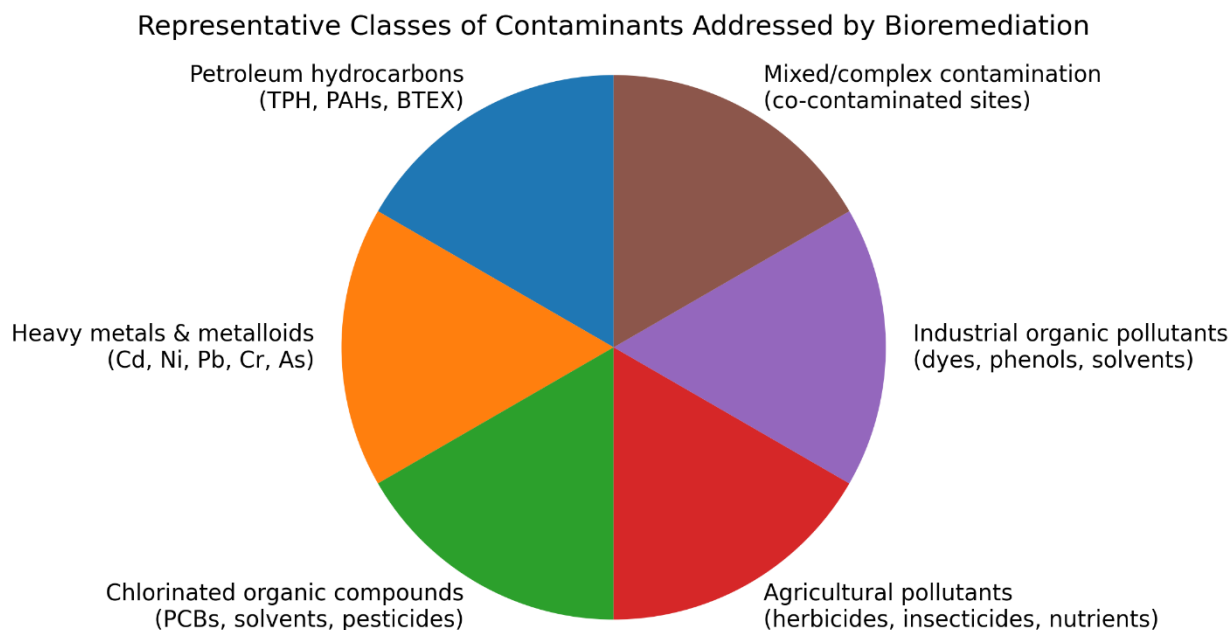


Figure 1. Representative classes of environmental contaminants that can be addressed using bioremediation-based strategies. The figure is illustrative and summarizes major contaminant groups reported in the literature rather than quantitative global proportions.

Therefore, this study evaluates the effectiveness of two widely available agro-wastes—cassava peels and sawdust—as biostimulants for the remediation of crude oil-polluted soil. The materials were applied individually and in combination (1:1 ratio) to stimulate indigenous biodegradation processes under controlled laboratory conditions. Changes in extractable total petroleum hydrocarbons (ETPH), associated heavy metals (nickel and cadmium), and soil pH were monitored over a 56-day treatment period. By directly comparing single-material treatments with a combined amendment approach, this work tests the hypothesis that co-application produces enhanced remediation performance. The findings aim to demonstrate a low-cost, environmentally sustainable alternative to energy-intensive remediation methods, with particular relevance for oil-producing regions such as the Niger Delta, where locally sourced materials can be leveraged to support practical and scalable soil remediation.

MATERIAL AND METHODS

Apparatus and Instrumentation

All glassware used in this study was thoroughly cleaned with detergent, rinsed with distilled water, and oven-dried before use. The major analytical instruments included a gas chromatograph equipped with a flame ionization detector (GC-FID; Agilent 6890, USA) for extractable total petroleum hydrocarbon (ETPH) analysis and a flame atomic absorption spectrophotometer (AAS; Solar Thermo Elemental, Model S4-71096, USA) for heavy metal determination. Additional equipment included an analytical dial spring scale (Hana Scale SP20 kg, China), a digital pH meter with glass electrode (PHS-25, PEC Medical, USA), a mechanical grinder (Px 2200, China), and a stainless-steel soil sieve (10 mm mesh size).

Chemicals and Reagents

All chemicals used were of analytical grade. These included methylene chloride, cyclohexane, acetone, perchloric acid (HClO₄), nitric acid (HNO₃), sulfuric acid (H₂SO₄), and aluminum nitrate solution. Anhydrous sodium sulfate and silica gel were used as drying agents during hydrocarbon extraction. All reagents were

purchased from Sigma Aldrich, Darmstadt, Germany. Distilled water was used throughout for solution preparation and dilution.

Preparation of Agro-Based Biostimulants

Cassava peels were collected from Aluu Community, while sawdust was obtained from a wood-processing mill along the East–West Road, Rumuosi, Obio-Akpor Local Government Area, Rivers State, Nigeria. Both materials were air-dried under ambient laboratory conditions (27–30 °C) for three weeks to reduce moisture content. The dried cassava peels were milled into semi-fine particles using a mechanical grinder, whereas the sawdust was used in its dried form without further size reduction.

Soil Sampling and Pretreatment

Bulk surface soil (0–15 cm depth) was collected from the Faculty of Agriculture garden, University of Port Harcourt. The soil was air-dried for seven days, homogenized, and passed through a 10 mm sieve to remove stones and plant debris. Before contamination, subsamples were analyzed to determine baseline pH, total petroleum hydrocarbon content, and heavy metal concentrations.

Artificial Soil Contamination

To simulate crude oil pollution, 4 kg of the prepared soil were thoroughly mixed with 40 mL of Bonny Light crude oil (light crude) using manual homogenization. The contaminated soil was left undisturbed for one week to allow acclimatization of indigenous microorganisms before treatment application.

Experimental Design

The experiment consisted of five treatment groups, each containing 1 kg of soil, arranged as shown in Table 1. One group served as the uncontaminated control, while another consisted of contaminated soil without amendment. The remaining groups received cassava peels, sawdust, or a combination of both as bio-stimulants. The experiment was conducted over 56 days under laboratory conditions. Soil samples were manually turned twice weekly to improve aeration, and moisture content was maintained by periodic addition of distilled water.

Group	Soil condition	Treatment applied
1	Unpolluted soil (1 kg)	No treatment
2	Polluted soil (1 kg)	No treatment
3	Polluted soil (1 kg)	Cassava peels (100 g)
4	Polluted soil (1 kg)	Sawdust (100 g)
5	Polluted soil (1 kg)	Cassava peels (100 g) + Sawdust (100 g)

Table 1. Experimental design for the different soil treatments.

Sample Collection

Soil samples were collected at predetermined intervals using clean, airtight sampling containers. The samples were stored in insulated coolers with ice packs and transported immediately to the laboratory for analysis. Soil samples for ETPH, Ni, Cd, and pH were collected at Day 0 (baseline), Day 28, and Day 56 of the 56-day experimental period.

Determination of Extractable Total Petroleum Hydrocarbons (ETPH)

Extractable total petroleum hydrocarbons (ETPH) were determined by solvent extraction followed by GC–FID analysis. 10 g of air-dried, homogenized soil was placed in a 1 L separatory funnel and extracted with methylene chloride (20 mL). The extraction was performed three times. Each extraction involved vigorous shaking for 2 min with intermittent venting, followed by phase separation for at least 10 min.

The combined organic extracts were then passed through a drying column containing cotton wool, anhydrous sodium sulfate, and silica gel. The extract was concentrated under a gentle stream of N₂ to 1 mL and diluted with an equal volume of solvent. A 1 µL aliquot was injected into the GC–FID.

Separation was carried out on a capillary column using the instrument's routine temperature program for petroleum hydrocarbon analysis. Quantification was performed using external calibration with hydrocarbon standards. Calibration curves were linear over the working range ($R^2 \geq 0.99$). Reagent blanks and duplicate samples were analyzed with each batch.

ETPH concentration was calculated according to equation 1 as:

$$ETPH \left(\frac{mg}{kg} \right) = \frac{C \times V}{m} \quad (1)$$

Where C is the hydrocarbon concentration obtained from GC calibration (mg/L), V is the final volume of the extract (L), and m is the dry mass of the soil sample (kg).

Determination of Heavy Metals in Soil

Nickel and cadmium were determined after acid digestion followed by flame atomic absorption spectrophotometry (AAS). 1 g of air-dried soil was digested with a mixed acid solution of H₂SO₄:HNO₃:HClO₄ (40:40:20, v/v/v). The mixture was heated on a hot plate until nearly dry, yielding a clear solution. After cooling, the digest was filtered and diluted to 100 mL with distilled water.

Metal concentrations were measured using a flame AAS with element-specific hollow cathode lamps. Calibration was performed using standard solutions prepared from stock standards. Blanks and duplicate samples were included during analysis.

Metal concentration (mg/kg) was calculated (equation 2):

$$Metal \ concentration \left(\frac{mg}{kg} \right) = \frac{A \times V}{m} \quad (2)$$

Where A is the metal concentration measured by AAS (mg/L), V is the final digest volume (L), and m is the dry mass of the soil sample (kg).

Soil pH Determination

Soil pH was measured using a soil–water suspension. 20 g of air-dried soil was mixed with 50 mL of distilled water (1:2.5, w/v). The suspension was stirred and allowed to stand for 30 min, with occasional mixing. pH was measured using a digital pH meter, calibrated with buffer solutions at pH 4.0 and 7.0 prior to measurement.

Calculation of Percentage Reduction

To evaluate the efficiency of Bioremediation, the percentage reduction in total petroleum hydrocarbons and heavy metal concentrations over time was calculated using equation 3:

$$\% \text{ Reduction} = \frac{C_0 - C_t}{C_0} \times 100 \quad (3)$$

Where C₀ is the initial concentration of the contaminant (mg/kg), and C_t is the concentration at time t (mg/kg).

Statistical Analysis

All measurements were conducted in triplicate, and results are reported as mean ± standard deviation (n = 3). One-way analysis of variance (ANOVA) was used to assess overall treatment effects at the p < 0.05 significance level, using SPSS software (version 20.0). Due to the limited sample size, differences among amended treatments were interpreted cautiously and are discussed primarily as trends based on mean values.

RESULTS AND DISCUSSION

Hydrocarbon reduction following amendment application

The results of this study show that agro-waste amendments, specifically cassava peels and sawdust, improved the Bioremediation of crude oil-polluted soil over the 56-day experimental period, with measurements taken at Days 0, 28, and 56. As shown in Figure 2, the untreated polluted control (Group 2) showed only a very small decrease in extractable total petroleum hydrocarbons (ETPH), from $13,000 \pm 30.0$ mg/kg at Day 0 to $12,870 \pm 21.22$ mg/kg at Day 56, representing a reduction of about 1%. This minimal change reflects natural attenuation, which typically occurs slowly in the absence of intervention, as indigenous hydrocarbon-degrading microorganisms may be present but require improved environmental conditions to achieve substantial contaminant removal.

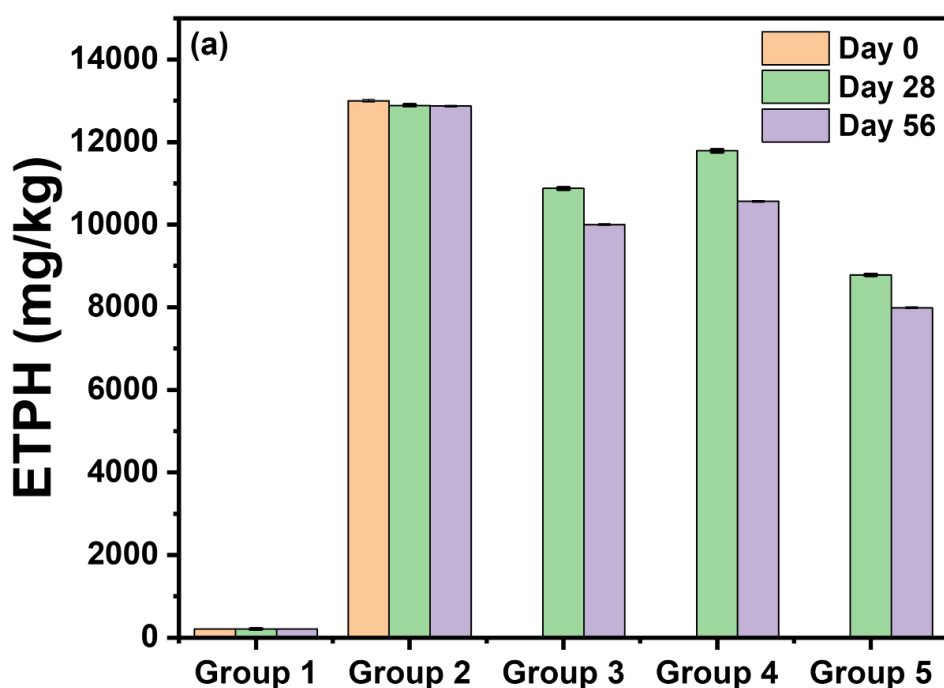


Figure 2. Extractable total petroleum hydrocarbon (ETPH) concentrations in experimental and control groups during the 56-day treatment period. Values are mean \pm standard deviation ($n = 3$). Day 0 represents the contaminated soil baseline before amendment application; amended treatments (Groups 3–5) were applied after contamination and therefore do not have separate Day 0 measurements.

In contrast, cassava peel amendment (Group 3) enhanced hydrocarbon reduction compared with the untreated control (Figure 2). ETPH decreased to $10,877 \pm 24.07$ mg/kg at Day 28 and further to $10,001 \pm 22.33$ mg/kg at Day 56, corresponding to 16.3% and 23.1% reduction, respectively, relative to the polluted baseline at Day 0 ($13,000 \pm 30.0$ mg/kg, Group 2). Sawdust amendment (Group 4) also improved remediation performance, with ETPH values of $11,787 \pm 31.33$ mg/kg at Day 28 and $10,566 \pm 23.23$ mg/kg at Day 56, representing 9.3% and 18.7% reduction. The greatest decrease was observed in the combined cassava peel-sawdust treatment (Group 5), where ETPH declined to $8,787 \pm 16.55$ mg/kg at Day 28 and $7,986 \pm 25.53$ mg/kg at Day 56, equivalent to 32.4% and 38.6% reduction, respectively. Percentage reductions in ETPH, Ni, and Cd were calculated relative to the Day 0 polluted control (Group 2) using equation 3. Overall, the combined treatment showed the largest reductions across the measured parameters. By Day 56, Group 5 had the lowest mean

ETPH concentration among the amended soils, and all amended treatments showed lower ETPH levels than the untreated polluted control, indicating improved hydrocarbon attenuation compared with natural attenuation.

Mechanistically, the enhanced performance of the combined treatment is likely due to the complementary roles of cassava peels and sawdust in improving key factors that limit petroleum biodegradation. Cassava peels can act as a biostimulant by supplying readily degradable organic substrates and nutrients that promote microbial growth and activity, as reported in previous studies on crude-oil degradation.²⁷ Sawdust, in contrast, can improve soil physical properties such as porosity and aeration, provide additional organic matter that supports microbial colonization, and influence contaminant availability through sorption. Together, these effects create a more favorable soil environment for sustained biodegradation.

The improved performance observed in amended soils is therefore consistent with biostimulation processes. Organic amendments can enhance microbial activity, improve soil structure, and modify contaminant bioavailability, thereby contributing to more effective hydrocarbon attenuation. Similar trends have been reported in studies showing that organic amendment-based remediation becomes more effective when multiple limiting factors, such as nutrient availability, oxygen transfer, and soil matrix effects, are addressed simultaneously.^{28,29}

Heavy metals response during treatment (nickel and cadmium)

Crude oil-contaminated soils often contain trace metals such as Ni and Cd, which can contribute additional environmental risk. In this study, nickel concentrations (Figure 3a) decreased the most in the combined amendment treatment. By Day 56, the combined cassava peel-sawdust treatment (Group 5) had the lowest mean nickel concentration among the amended soils and was lower than that of the untreated polluted control (Group 2). Relative to the Day 0 polluted baseline (10.05 ± 0.83 mg/kg), this corresponds to an 11.5% reduction. Reductions observed for the single-amendment treatments were smaller than those for the multiple-amendment treatments. The greater decrease in nickel under the combined treatment may be associated with changes in metal mobility rather than direct removal. The addition of organic materials can increase soil organic matter, enhance sorption, and modify metal bioavailability and partitioning. Such responses are consistent with previous studies showing that organic amendments can influence heavy metal behavior by altering sorption capacity, pH, and microbial activity, even when hydrocarbon degradation is the primary objective.^{28,29}

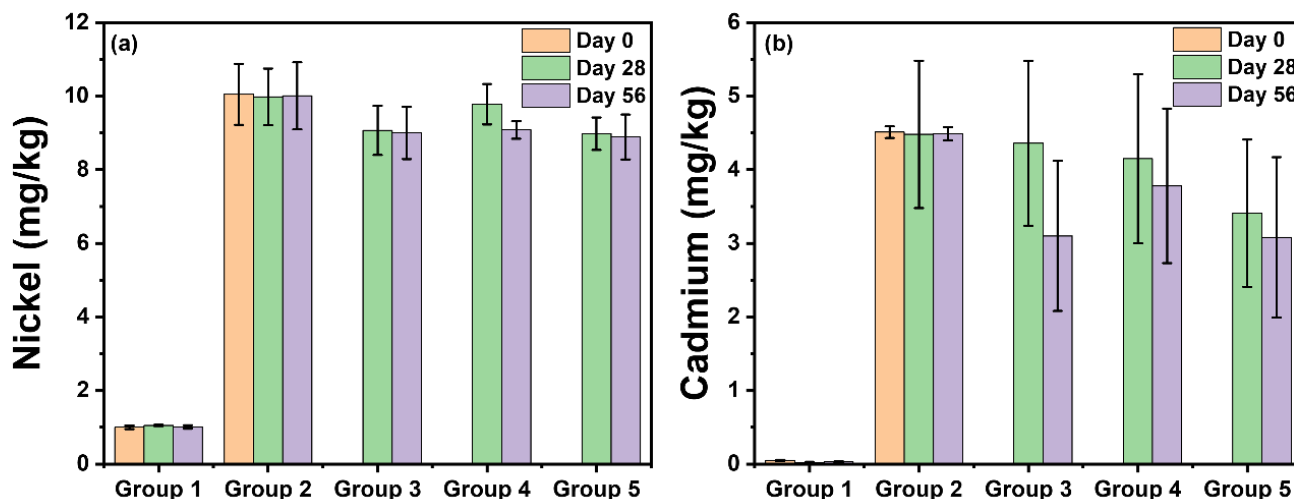


Figure 3. (a) Nickel and (b) cadmium concentrations in experimental and control groups during the 56-day treatment period. Values are mean ± standard deviation (n = 3); small bars reflect low variability.

Cadmium concentrations also decreased under amendment conditions by Day 56. Cassava peel alone reduced Cd to 3.10 ± 1.02 mg/kg, while sawdust alone reduced Cd to 3.78 ± 1.25 mg/kg. Reductions were observed across all amended soils, with the combined cassava peel–sawdust treatment (Group 5) showing the lowest mean Cd concentrations (3.41 ± 0.77 mg/kg at Day 28 and 3.08 ± 0.19 mg/kg at Day 56). Relative to the Day 0 polluted baseline (4.51 ± 0.08 mg/kg, Figure 3b), these values correspond to 24.4% and 31.7% reduction at Day 28 and Day 56, respectively.

The observed decrease in Cd under amended conditions likely reflects changes in metal mobility rather than direct removal. Organic amendments can enhance sorption and modify metal availability by interacting with soil organic matter and altering soil chemistry. Overall, amended soils showed lower Cd concentrations than the untreated control, while differences among amended treatments were relatively small and should be interpreted as trends in mean values given the variability in Cd measurements.

Treatment Group	ETPH reduction (%)*		Ni reduction (%)*		Cd reduction (%)*	
	Day 28	Day 56	Day 28	Day 56	Day 28	Day 56
Group 3 (cassava peels)	16.3	23.1	9.8	10.4	3.3	31.3
Group 4 (sawdust)	9.3	18.7	2.7	9.7	8.0	16.2
Group 5 (combined, 1:1)	32.4	38.6	10.6	11.5	24.4	31.7

*Percentage reductions were calculated relative to the polluted baseline at Day 0 (Group 2). Group 1 (unpolluted control) was excluded from the calculations.

Table 2. Percentage reduction of ETPH, Ni, and Cd in amended soils at Day 28 and Day 56 relative to the polluted baseline (Group 2, Day 0). Values were calculated from mean concentrations (n = 3). Differences among treatments are presented as trends based on mean values.

pH trends and implications for biodegradation performance

Soil pH is an important factor influencing microbial activity and nutrient availability during Bioremediation. In the present study, amended soils showed a gradual shift toward neutral to slightly alkaline conditions by Day 56 (Figure 4), with pH values generally in the range of approximately 7.0–7.5. In contrast, the untreated polluted control remained slightly acidic, with pH values around 6.4–6.5 throughout the study period. Such near-neutral conditions are generally favorable for hydrocarbon-degrading microorganisms and help maintain a stable environment for sustained biodegradation over extended treatment periods. These findings are consistent with previous studies indicating that effective Bioremediation depends on maintaining suitable environmental conditions, including balanced soil chemistry, adequate moisture, and sufficient oxygen availability to support microbial degradation processes.^{30,31}

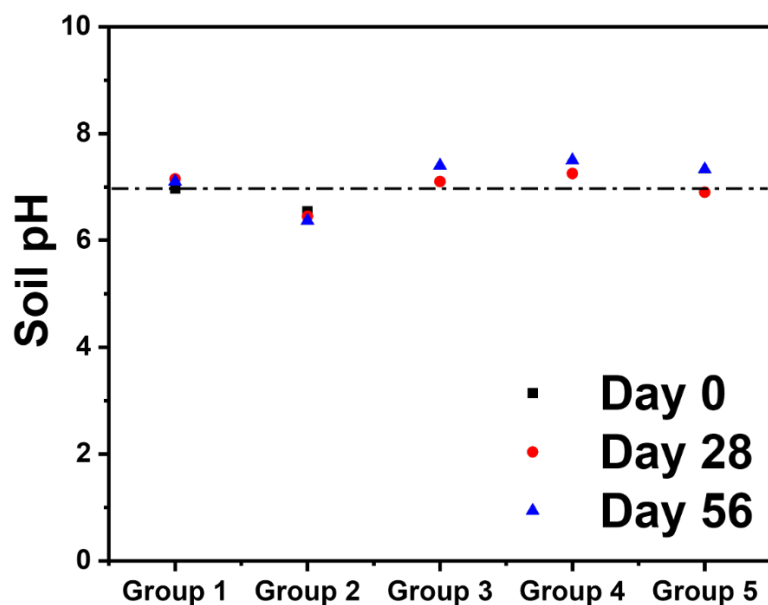


Figure 4. Variation in soil pH across experimental and control groups during the 56-day bioremediation period. Values represent the mean of triplicate measurements; error bars are not shown.

Detailed mean \pm standard deviation values corresponding to Figures 2–4 are included in the Supporting Information (Tables S1–S4).

Why is this approach valuable compared to other remediation strategies?

A central strength of this work is that it leverages materials generally regarded as waste (cassava peels and sawdust) to treat crude oil-polluted soils, supporting both remediation and circular resource use. Compared with thermal and physicochemical methods, amendment-based Bioremediation can reduce secondary environmental burdens. For example, thermal desorption is effective for hydrocarbon-impacted soils, but it generates off-gases requiring air pollution control systems, and secondary pollution concerns are widely noted in technical guidance and peer-reviewed literature.^{10,32} Likewise, systematic reviews of remediation technologies highlight that incineration can be expensive and may lead to secondary pollution.^[6] Chemical oxidation approaches (e.g., ISCO) can provide faster contaminant reduction in some cases, but they require careful oxidant delivery and management and may inhibit microbial communities—making them less aligned with soil ecological recovery in certain contexts.^{20,33} Excavation and landfilling can rapidly remove

contaminated soil from a site, but often transfer the problem elsewhere and add transport/disposal burdens, rather than restoring soil function in place.

In contrast, the approach demonstrated here provides a lower-cost, environmentally favorable pathway that (i) enhances degradation relative to natural attenuation (Group 2), (ii) shows clear improvement with single amendments (Groups 3 and 4), and (iii) shows the strongest overall performance with the combined amendment (Group 5), consistent with an enhanced biostimulation effect (figures 2 and 3). The results therefore, support the practical value of using agro-waste materials as biostimulants—especially in regions where cassava processing and wood milling generate abundant residues and where low-cost remediation approaches are needed.

Practical implications and study scope

Overall, the dataset supports the conclusion that cassava peel and sawdust amendments enhance remediation performance individually, and that their combined application yields the greatest reduction in ETPH and improved heavy metal outcomes over 56 days (Figures 2–4). Future work could strengthen mechanistic attribution by directly tracking microbial abundance/activity, nutrient dynamics (N/P), and oxygen availability across treatments; however, even within the current scope, the clear separation between untreated control and amended soils, and the generally improved outcomes for the combined amendment, underscore the promise of this waste-to-resource remediation strategy.

Limitations of the study

This study used soil artificially contaminated and incubated under controlled laboratory conditions. While this approach ensured consistent starting conditions across treatments, the behavior of hydrocarbons and metals in field soils may differ due to natural heterogeneity, weathering, fluctuating moisture, and varying aeration conditions. Additionally, sampling was performed at three time points (Day 0, 28, and 56). Although these measurements captured short-term trends, the study duration does not allow assessment of long-term stability, potential rebound effects, or the persistence of treatment performance beyond the incubation period. The combined amendment treatment also contained a higher total mass of organic material (200 g/kg soil) compared with the single-amendment treatments (100 g/kg). Therefore, the greater reductions observed in this treatment may reflect differences in amendment dose and associated sorption or dilution effects, and the present design does not allow a clear distinction between dose effects and true interaction between cassava peels and sawdust.

Furthermore, hydrocarbons were reported as bulk extractable total petroleum hydrocarbons (ETPH). Fractionation into specific hydrocarbon groups was not performed, and microbial population dynamics were not evaluated; consequently, the specific degradation pathways and biological mechanisms responsible for the observed changes were not determined because this was beyond the scope of the present study. Similarly, reductions in Ni and Cd are reported as decreases in measured concentrations following digestion and analysis. Metal speciation, fractionation, or mass balance were not assessed; therefore, the results likely reflect changes in metal mobility or immobilization rather than confirmed removal from the soil system.

CONCLUSIONS

The present study demonstrates that agro-waste materials can be effectively repurposed as biostimulants for the Bioremediation of crude oil-polluted soils. The application of sawdust and cassava peels individually enhanced remediation performance relative to natural attenuation, as evidenced by measurable reductions in extractable total petroleum hydrocarbons (ETPH) and associated heavy metals (nickel and cadmium) over a 56-day treatment period. Cassava peels showed particularly efficient hydrocarbon and metal reduction, likely due to their ability to stimulate indigenous microbial activity through the provision of readily available organic substrates. Importantly, the combined application of cassava peels and sawdust at a 1:1 ratio produced the strongest overall remediation performance, yielding the highest reductions in ETPH and low residual concentrations of nickel and cadmium. This improved performance suggests a beneficial combined effect, in which the complementary roles of the two amendments—nutrient stimulation from cassava peels and improved soil structure and aeration from sawdust—collectively enhanced biodegradation processes beyond what was achieved with either amendment alone.

Compared with energy-intensive and environmentally burdensome remediation approaches such as incineration, excavation, or chemical oxidation, the amendment-based bioremediation strategy evaluated in this study offers a low-cost, environmentally sustainable alternative that minimizes secondary pollution while promoting soil recovery. By converting readily available agro-wastes into functional remediation agents, this approach aligns with circular economy principles and provides a practical solution for managing crude oil-contaminated soils, particularly in resource-limited settings. Overall, the findings confirm that cassava peels and sawdust are effective biostimulants for crude oil bioremediation, and that their combined application yields the most robust remediation outcome. This work highlights the potential of locally sourced organic waste as a sustainable tool for environmental remediation and supports further development and field-scale evaluation of integrated agro-waste-based bioremediation strategies.

Supplementary Materials: The results obtained from the analysis carried out on different days to check the effect of cassava and sawdust on the Bioremediation of crude oil polluted garden soil are presented below:

ETPH (mg/kg)

	DAY 0	DAY 28	DAY 56
GROUP 1	210.00±7.00	210.00±5.00	211.00±7.5
GROUP 2	13000±30.00	12888±19.01	12870.00±21.22
GROUP 3	-	10877.00±24.07*	10001.00±22.33*
GROUP 4	-	11787.00±31.33	10566.00±23.23*
GROUP 5	-	8787.00±16.55*	7986.00±25.53*

Values are Mean ± SD. Asterisks* indicate values lower than the Day 0 polluted control (Group 2); statistical comparisons are interpreted cautiously due to n = 3.

Table S1. Total Petroleum Hydrocarbons in Experimental and Control Groups

Nickel (mg/kg)

	DAY 0	DAY 28	DAY 56
GROUP 1	1.00±0.05	1.05±0.02	1.01±0.05
GROUP 2	10.05±0.83	9.98±0.77	10.01±0.91
GROUP 3	-	9.07±0.67*	9.00±0.71
GROUP 4	-	9.78±0.55	9.08±0.24
GROUP 5	-	8.98±0.44*	8.89±0.61*

Values are Mean ± SD. Asterisks* indicate values lower than the Day 0 polluted control (Group 2); statistical comparisons are interpreted cautiously due to n = 3.

Table S2. Soil Nickel Concentration in Experimental and Control Groups**Cadmium (mg/kg)**

	DAY 0	DAY 28	DAY 56
GROUP 1	0.05±0.01	0.02±0.01	0.03±0.01
GROUP 2	4.51±0.08	4.48±1.00	4.49±0.09
GROUP 3	-	4.36±1.02	3.10±1.02*
GROUP 4	-	4.15±0.95	3.78±1.25*
GROUP 5	-	3.41±0.77*	3.08±0.19*

Values are Mean ± SD. Asterisks* indicate values lower than the Day 0 polluted control (Group 2); statistical comparisons are interpreted cautiously due to n = 3.

Table S3. Soil Cadmium Concentration in Experimental and Control Groups**Soil pH**

	DAY 0	DAY 28	DAY 56
GROUP 1	6.97	7.15	7.10
GROUP 2	6.54	6.45	6.37
GROUP 3	-	7.10	7.40
GROUP 4	-	7.25	7.50
GROUP 5	-	6.90	7.33

Values represent the mean of triplicate measurements. Asterisks* indicate values lower than the Day 0 polluted control (Group 2); statistical comparisons are interpreted cautiously due to n = 3.

Table S4. Soil pH in Experimental and Control Groups

Author Contributions: Conceptualization, M.C.O. and S.E.O.; methodology, M.C.O. and S.E.O.; software, S.E.O.; validation, S.E.O. and O.A.O.; formal analysis, M.C.O. and S.E.O.; investigation, M.C.O.; resources, M.C.O., S.E.O., and E.B.E.; data curation, M.C.O.; writing—original draft preparation, M.C.O., S.E.O. and O.A.O.; writing—review and editing, S.E.O., O.A.O and E.B.E.; visualization, O.A.O.; supervision, S.E.O. and E.B.E.; project administration, S.E.O., O.A.O. and E.B.E.; funding acquisition, M.C.O.. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: The data from this study will be made available upon reasonable request from the corresponding author.

Acknowledgments: The authors are grateful for the indirect support that contributed to this work.

Conflicts of Interest: The authors declare no conflict of interest.

REFERENCES

1. E. Koshlaf and A. S. Ball, "Soil bioremediation approaches for petroleum hydrocarbon polluted environments," *AIMS Microbiol.*, vol. 3, no. 1, pp. 25–49, 2017, doi: 10.3934/MICROBIOL.2017.1.25.
2. P. W. Grace Liu, T. C. Chang, L. M. Whang, C. H. Kao, P. T. Pan, and S. S. Cheng, "Bioremediation of

- petroleum hydrocarbon contaminated soil: Effects of strategies and microbial community shift," *Int. Biodeterior. Biodegrad.*, vol. 65, no. 8, pp. 1119–1127, 2011, doi: 10.1016/j.ibiod.2011.09.002.
3. G. Kebede, T. Tafese, E. M. Abda, M. Kamaraj, and F. Assefa, "Factors Influencing the Bacterial Bioremediation of Hydrocarbon Contaminants in the Soil: Mechanisms and Impacts," *J. Chem.*, vol. 2021, 2021, doi: 10.1155/2021/9823362.
 4. C. Lin, N. K. Cheruiyot, X. T. Bui, and H. H. Ngo, "Composting and its application in bioremediation of organic contaminants," *Bioengineered*, vol. 13, no. 1, pp. 1073–1089, 2022, doi: 10.1080/21655979.2021.2017624.
 5. M. Santos, S. Rebola, and D. V. Evtuguin, "Soil Remediation: Current Approaches and Emerging Bio-Based Trends," *Soil Syst.*, vol. 9, no. 2, pp. 1–24, 2025, doi: 10.3390/soilsystems9020035.
 6. U. Michael-Igolima, S. J. Abbey, and A. O. Ifelebuegu, "A systematic review on the effectiveness of remediation methods for oil contaminated soils," *Environ. Adv.*, vol. 9, no. September, p. 100319, 2022, doi: 10.1016/j.envadv.2022.100319.
 7. M. Saqr, R. R. Pant, J. O. Alao, P. K. Chaurasia, B. Abdelkebir, and M. E. Abd-Elmaboud, "Soil remediation through washing and flushing: bibliometric trends, technical review, and future prospects," *Environ. Earth Sci.*, vol. 84, no. 14, 2025, doi: 10.1007/s12665-025-12386-y.
 8. A. S. Correia and M. G. Rasteiro, "A Review of Persistent Soil Contaminants: Assessment and Remediation Strategies," *Environ. - MDPI*, vol. 12, no. 7, pp. 1–31, 2025, doi: 10.3390/environments12070229.
 9. E. O. Nwaichi, I. B. Ahmed, E. Ugwoha, J. N. Ugbebor, and S. B. Arokoyu, "Cost reduction strategies in the remediation of petroleum hydrocarbon contaminated soil," *Open Res. Africa*, vol. 5, pp. 1–18, 2022, doi: 10.12688/openresafrika.13383.1.
 10. C. Zhao, Y. Dong, Y. Feng, Y. Li, and Y. Dong, "Thermal desorption for remediation of contaminated soil: A review," *Chemosphere*, vol. 221, pp. 841–855, 2019, doi: 10.1016/j.chemosphere.2019.01.079.
 11. J. E. Vidonish, K. Zygourakis, C. A. Masiello, G. Sabadell, and P. J. J. Alvarez, "Thermal Treatment of Hydrocarbon-Impacted Soils: A Review of Technology Innovation for Sustainable Remediation," *Engineering*, vol. 2, no. 4, pp. 426–437, 2016, doi: 10.1016/J.ENG.2016.04.005.
 12. B. A. Mekonnen, T. A. Aragaw, and M. B. Genet, "Bioremediation of petroleum hydrocarbon contaminated soil: a review on principles, degradation mechanisms, and advancements," *Front. Environ. Sci.*, vol. 12, no. February, pp. 1–21, 2024, doi: 10.3389/fenvs.2024.1354422.
 13. M. Wu *et al.*, "Bioaugmentation and biostimulation of hydrocarbon degradation and the microbial community in a petroleum-contaminated soil," *Int. Biodeterior. Biodegrad.*, vol. 107, pp. 158–164, 2016, doi: 10.1016/j.ibiod.2015.11.019.
 14. G. Omokhagbor Adams, P. Tawari Fufeyin, S. Eruke Okoro, and I. Ehinomen, "Bioremediation, Biostimulation and Bioaugmentation: A Review," *Int. J. Environ. Bioremediation Biodegrad.*, vol. 3, no. 1, pp. 28–39, 2020, doi: 10.12691/ijebb-3-1-5.
 15. M. Wu, J. Wu, X. Zhang, and X. Ye, "Effect of bioaugmentation and biostimulation on hydrocarbon degradation and microbial community composition in petroleum-contaminated loessal soil," *Chemosphere*, vol. 237, p. 124456, 2019, doi: 10.1016/j.chemosphere.2019.124456.
 16. M. Nocentini, D. Pinelli, and F. Fava, "Bioremediation of a soil contaminated by hydrocarbon mixtures: The residual concentration problem," *Chemosphere*, vol. 41, no. 8, pp. 1115–1123, 2000, doi: 10.1016/S0045-6535(00)00057-6.
 17. C. Lin, N. K. Cheruiyot, X. T. Bui, and H. H. Ngo, "Composting and its application in bioremediation of organic contaminants," *Bioengineered*, vol. 13, no. 1, pp. 1073–1089, 2022, doi:

- 10.1080/21655979.2021.2017624.
18. H. Liu, M. Wu, H. Gao, N. Yi, and X. Duan, "Hydrocarbon transformation pathways and soil organic carbon stability in the biostimulation of oil-contaminated soil: Implications of ^{13}C natural abundance," *Sci. Total Environ.*, vol. 788, p. 147580, 2021, doi: 10.1016/j.scitotenv.2021.147580.
 19. Y. Li, J. Yang, Y. Song, and M. Wei, "Progress in biostimulation-based remediation of TPH-contaminated soils: a comprehensive review," *PeerJ*, vol. 13, pp. 1–45, 2025, doi: 10.7717/peerj.19991.
 20. Interstate Technology & Regulatory Council (ITRC), "Technical and Regulatory Guidelines Technical and Regulatory Guidance for In Situ Chemical Oxidation of Contaminated Soil and Groundwater," Washington, DC, 2005. [Online]. Available: <http://www.itrcweb.org>.
 21. Z. Xiao, W. Jiang, D. Chen, and Y. Xu, "Bioremediation of typical chlorinated hydrocarbons by microbial reductive dechlorination and its key players: A review," *Ecotoxicol. Environ. Saf.*, vol. 202, no. March, p. 110925, 2020, doi: 10.1016/j.ecoenv.2020.110925.
 22. M. Megharaj, B. Ramakrishnan, K. Venkateswarlu, N. Sethunathan, and R. Naidu, "Bioremediation approaches for organic pollutants: A critical perspective," *Environ. Int.*, vol. 37, no. 8, pp. 1362–1375, 2011, doi: 10.1016/j.envint.2011.06.003.
 23. N. Das and P. Chandran, "Microbial Degradation of Petroleum Hydrocarbon Contaminants: An Overview," *Biotechnol. Res. Int.*, vol. 2011, pp. 1–13, 2011, doi: 10.4061/2011/941810.
 24. V. R. Kondakindi, R. Pabbati, P. Erukulla, N. R. Maddela, and R. Prasad, "Bioremediation of heavy metals-contaminated sites by microbial extracellular polymeric substances – A critical view," *Environ. Chem. Ecotoxicol.*, vol. 6, no. April, pp. 408–421, 2024, doi: 10.1016/j.enceco.2024.05.002.
 25. M. Sanjana, R. Prajna, U. S. Katti, and R. V. Kavitha, "Bioremediation - the recent drift towards a sustainable environment," *Environ. Sci. Adv.*, vol. 3, no. 8, pp. 1097–1110, 2024, doi: 10.1039/d3va00358b.
 26. Ibrahim *et al.*, "Bioremediation of soils with emerging organic contaminants using immobilized microorganisms," *Environ. Technol. Innov.*, vol. 40, p. 104345, 2025, doi: 10.1016/j.eti.2025.104345.
 27. K. Jude, F. B. G. Tanee, and A. Ngbaraue, "Use of Cassava Peel as Biostimulant in Bioremediation of Crude Oil-Polluted Soil," *J. Appl. Sci.*, vol. 22, no. 6, pp. 351–361, 2022, doi: 10.3923/jas.2022.351.361.
 28. B. K. Majeed, D. M. S. Shwan, and K. A. Rashid, "A review on environmental contamination of petroleum hydrocarbons, its effects and remediation approaches," *Environ. Sci. Process. Impacts*, vol. 27, no. 3, pp. 526–548, 2025, doi: 10.1039/d4em00548a.
 29. T. G. Ambaye *et al.*, "Remediation of soil polluted with petroleum hydrocarbons and its reuse for agriculture: Recent progress, challenges, and perspectives," *Chemosphere*, vol. 293, p. 133572, 2022, doi: 10.1016/j.chemosphere.2022.133572.
 30. US EPA, L. Land and Emergency Management, and EPA, "How To Evaluate Alternative Cleanup Technologies For Underground Storage Tank Sites," *Epa 510-B-17-003*, no. October, pp. XIII-1-XIII–15, 2017, [Online]. Available: https://www.epa.gov/sites/production/files/2014-03/documents/tum_ch9.pdf.
 31. U.S. Environmental Protection Agency, "For Underground Storage Tank Sites A Guide For Corrective Action Plan Reviewers," no. October, p. EPA 510-B-17-003, 2017, [Online]. Available: https://www.epa.gov/sites/default/files/2014-03/documents/tum_ch10.pdf.
 32. Occupational Safety and Health Administration, "Safety and Health Information Bulletin Remediation Technology Health and Safety Hazards: Thermal Desorption," pp. 1–14, 2003, [Online]. Available: http://www.ertresponse.com/health_safety/index.htm.

33. D. Daâssi and F. Qabil Almaghribi, "Petroleum-contaminated soil: environmental occurrence and remediation strategies," *3 Biotech*, vol. 12, no. 6, pp. 1–17, 2022, doi: 10.1007/s13205-022-03198-z.

Received: January 3, 2025 / **Accepted:** February 26, 2026 / **Published (online):** March 15, 2026 (Europe/Madrid)

Citation. Ogbonna MC, Okoro SE, Obewhere OA, Essien EB. Bioremediation of crude oil polluted soils using cassava peels and sawdust as biostimulants. *BioNatura Journal: Ibero-American Journal of Biotechnology and Life Sciences*. *BioNatura Journal: Ibero-American Journal of Biotechnology and Life Sciences*. 2026;3(1):3. <https://doi.org/10.70099/BJ/2026.03.01.3>

Correspondence should be addressed to: samson.okoro@uniport.edu.ng

Peer Review Information BioNatura Journal thanks the anonymous reviewers for their valuable contribution to the peer-review process. Regional peer-review coordination was conducted under the BioNatura Institutional Publishing Consortium (BIPC), involving:

- Universidad Nacional Autónoma de Honduras (UNAH)
- Universidad de Panamá (UP)
- RELATIC (Panama)

Reviewer selection and assignment were supported via: <https://reviewerlocator.webofscience.com/>

Publisher Information Published by Clinical Biotec S.L. (Madrid, Spain) as the publisher of record under the BioNatura Institutional Publishing Consortium (BIPC). Places of publication: Madrid (Spain); Tegucigalpa (Honduras); Panama City (Panama). Online ISSN: 3020-7886.

Open Access Statement All articles published in BioNatura Journal are freely and permanently available online immediately upon publication, without subscription charges or registration barriers.

Publisher's Note BioNatura Journal remains neutral regarding jurisdictional claims in published maps and institutional affiliations.

Copyright and License © 2026 by the authors. This article is published under the terms of the Creative Commons Attribution (CC BY 4.0) license, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

License details: <https://creativecommons.org/licenses/by/4.0/>

Governance For editorial governance and co-publisher responsibilities, see the BIPC Governance Framework (PDF) at: <https://clinicalbiotec.com/bipc>