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Effect of Biochar from Plantain Trunk Wastes' Remediation on Microbial Community Dynamics of Crude Oil Contaminated Soil

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Abstract

The effect of plantain trunk biochar (PTB) remediation on microbial community dynamics of crude oil-contaminated soil was studied. The microbial load in the experimental soil (both treated and untreated soil) significantly ($p<0.05$) increased as the remediation time increased from week 1 to week 3 after which a significant decrease was recorded for biochar-amended soil at week 4, while control treatment continued increasing. Soil remediated with 100%, Plantain trunk biochar, PTB had significantly ($p<0.05$) high microbial count (108.34±2.39to 207.67±5.34 x10^{-2} Cfu/g) throughout the 4 weeks of the study. The high microbial counts in bio-remediated soil may be the result of the presence of appreciable quantities of nitrogen and phosphorus provided by the biochar which is a necessary nutrient for bacterial biodegradative activities. These higher microbial counts would favor hydrocarbon degradation and this study suggests that soil nutrient enhancement through biochar amendment biostimulates and optimizes bioremediation by increasing microbial biomass activities.

Keyword: Remediation, Microbial Dynamics, Hydrocarbon and Biochar

1. Introduction

Petroleum products are essential to modern society but also represent a significant source of environmental challenges such as pollution, making them one of the most impactful contaminants today (Zhang et al., 2020; Wang et al., 2019). Soil degradation, particularly due to petroleum contamination, is a critical concern as crude oil, diesel spills, and industrial discharges render land unsafe and unproductive. Such contamination introduces severe risks to soil ecosystems, including the inhibition of plant growth, the disruption of soil structure, and the potential contamination of groundwater resources (Li et al., 2021). Addressing these environmental hazards is crucial for preserving soil health and mitigating the long-term impacts of petroleum pollution on ecosystems. A comprehensive understanding of these effects is necessary to develop and implement effective strategies for the remediation and restoration of contaminated environments (Chen et al., 2022).

Recent research confirms that petroleum products play a significant role in modern society but also present considerable environmental hazards, notably as pollutants. Petroleum contamination in soil leads to a range of environmental issues, including impaired plant growth, disruption of soil structure, and contamination of groundwater. These effects highlight the need to address the risks associated with petroleum-contaminated soils (Chettri et al., 2021; Chunyan et al., 2023). The impacts of such contamination extend beyond immediate environmental damage, contributing to broader ecological and human health concerns (Dai et al., 2023).
Efforts to mitigate these effects have emphasized the role of bioremediation as a cost-effective and eco-friendly solution. Biochar, a byproduct of biomass pyrolysis, has gained recognition for its ability to enhance

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soil health due to its large surface area and stability in the soil environment. It provides a robust medium for the bioremediation of petroleum hydrocarbons by supporting microbial communities that facilitate contaminant degradation (Duan et al., 2015; Gidudu et al., 2021). Recent studies underscore biochar's effectiveness in binding pollutants, thereby reducing their bioavailability and assisting in the recovery of contaminated environments (Fawole et al., 2022).

Plantain, scientifically known as Musa paradisiaca and belonging to the family Musaceae, holds a significant position as a fruit crop in tropical and subtropical regions, with cultivation extending over approximately 8.8 million hectares (Mohapatra *et al.*, 2010). Regarded as one of the oldest cultivated crops globally (Kumar *et al*., 2012), plantain trunks, a substantial part of the plant, are often discarded as waste (Mohapatra *et al.*, 2010). For visual reference, Fig.I displays an image of the plantain trunk. This paper reviews recent advancements in using Char from plantain trunks for remediation and the Effects of Temperature on the char.

Fig. 1: Plantain trunks

2. Materials and Methods

Plantain trunks were obtained from Gossa Market, located on Airport Road in Abuja. The soil sample and accompanying crude oil were sourced from the NNPC depot in Umurolu, Port Harcourt, Rivers State, Nigeria. Following proper labeling, these samples were transported to the Biotechnology Advanced Laboratory at the Sheda Science and Technology Complex (SHESTCO) situated on Lokoja Expressway in Abuja for comprehensive analysis.

Soil samples were analysed for their physical and chemical properties before biochar application. After adding plantain trunk biochar, we conducted weekly analyses for four weeks. We assessed the soil samples for their physical and chemical parameters before treatment and at weekly intervals for four weeks, following the protocols outlined in AOAC (2000).

Thermogravimetric analysis (TGA) was conducted to analyze the decomposition of dried and ground plantain trunks. The TGA method revealed that the pyrolysis of the materials can occur between the temperatures of 48.55^{o}C to 553^{o}C for the plantain trunk, with an initial decomposition temperature (IDT) of 250^{o}C and a final decomposition temperature (FDT) of 550^{o}C. The samples were prepared by washing, drying, and grinding, and then weighed into the TGA analyzer. The pyrolysis process was carried out in a controlled nitrogen environment using specific operating conditions. After the pyrolysis, the produced charcoal was collected, sieved, labeled, and stored for further analysis to prevent oxidation reactions.

Five hundred (500) of soil (sieved with 2mm mesh size) was placed in plastic vessels labeled A, B, C, and D. The soil was then polluted with crude petroleum oil at 1:1 and left undisturbed for two days. The soil was assigned to different group treatments as shown below.

Sample A = Served as control with only soil and 10% crude oil

Sample B = 100% Plantain trunk biochar (PTB) + contaminated Soil

Sample C = 70%PTB only + contaminated soil

Sample D = 30% PTB + contaminated soil

The moisture content was adjusted to 60% by adding water, and the soil was tilled for aeration three times per week during the entire experimental period. The plastic vessels were incubated in triplicates at room temperature (30 ± 2oC). The microcosms were maintained at room temperature in the laboratory incubator for 4 weeks. The soil treated with biochar was regularly watered with 100ml of distilled water each week to compensate for evaporation, and mixed every other day for aeration.

The total microbial count in the soil was determined by plating a 1g serially diluted sample of soil on oil agar (OA) and then incubating it at 30°C for 72 hours. The colonies on each plate were counted and recorded as colony-forming units per gram of soil (CFU/g). The oil agar medium contained the following components: 1.8g K_2HPO_4, 4.0g NH_4Cl, 0.2g $MgSO_4.7H_2O$, 1.2g KH_2PO4, 0.01g $FeSO_4$ $7H_2O$, 0.1g NaCl, 20g agar, and 1% (v/v) used lubricating oil in 1L distilled water, with a pH of 7.4 (Zajic and Suplission, 1972).

3. Result and Discussion

3.1 Effect of the plantain trunk biochar remediation on microbial community dynamics of crude oil contaminated soil

The impact of Plantain trunk biochar remediation on the microbial community dynamics of crude oil-contaminated soil is presented in Table 1. The microbial load in the experimental soil, both treated and untreated, exhibited a significant ($p<0.05$) increase with remediation time from week 1 to week 3. Subsequently, a notable decrease was observed in the biochar-amended soil at week 4, while the microbial count in the control treatment continued to rise. Throughout the 4-week study period, the contaminated untreated soil (control) consistently demonstrated the lowest microbial count (18.24±0.67 to 38.35±0.34 $x10^{-2}$ Cfu/g). Among the remediated soil samples, those treated with 100% Plantain trunk biochar (PTB) consistently exhibited the highest microbial count (108.34±2.39 to 207.67±5.34 $x10^{-2}$ Cfu/g) over the same period.

Table 1: Microbial Dynamics of Crude Oil Contaminated Soils Treated with Different Biochar for 4 Weeks

Treatment	Week (10^{-2} Cfu/g soil)				
	0	**1**	**2**	**3**	**4**
A	22.50±0.02	22.36±0.03^{a}	33.34±0.16^{a}	38.35±0.34^{a}	48.24±0.67^{a}
B	22.50±0.02	108.34±2.39^{e}	189.58 ±4.53^{e}	207.67±5.34^{e}	180.23±4.21^{d}
C	22.50±0.02	94.09±0.03^{d}	131.34±3.21^{d}	179.01±3.43^{d}	104.21±3.03^{c}
D	22.50±0.02	68.25±0.37^{c}	106.34±2.83^{c}	132.24±2.32^{c}	100.34±2.44^{c}

Values are Mean ± Standard Error of Mean of replicate determinations. Values with different alphabets along a row are significantly different ($p<0.05$).

Keys: Sample A = contaminated soil only (control); Sample B = contaminated soil +100% Plantain trunk biochar (PTB) only; Sample C = contaminated soil + 70% PTB; Sample D = contaminated soil + 30% PTB

The microbial counts obtained for both uncontaminated and contaminated soil in this study indicate the presence of indigenous hydrocarbon-utilizing bacteria in the soil samples. Recent studies confirm that hydrocarbon-utilizing bacteria such as *Pseudomonas*, *Bacillus*, and *Acinetobacter* continue to dominate in oil-contaminated soils. For example, a 2021 study by Okoro et al. identified similar bacterial species in hydrocarbon-contaminated soils, including *Pseudomonas* and *Bacillus*, as key players in biodegradation. The presence of these bacteria indicates an inherent capacity of soil microbiota to degrade hydrocarbons.

The initial decrease in microbial count in the soil after contamination with crude oil (29 x 10^{-2} cfu/g to 22 x 10^{-2} cfu/g) could be attributed to microbial adaptation to the new environment: The observation of an initial reduction in microbial populations after crude oil contamination, attributed to microbial adaptation, remains consistent with recent findings. A 2022 study by Almansoory et al. described that microbial communities initially decline due to the toxicity of hydrocarbons before acclimating and recovering, especially after amendments such as biochar or nutrients. This adaptation phase is crucial before microbes can actively participate in bioremediation.

There was an initial increase in the hydrocarbon-utilizing microbial population from week 1 to week 3 of the biochar amendment. The role of biochar in enhancing microbial populations and activity is well-documented in contemporary research. A 2020 review by Cai et al. concluded that biochar amendments significantly increase microbial biomass and enzymatic activity, particularly in oil-contaminated soils, by providing a conducive environment for microbial growth. The results of increased microbial counts after the biochar addition align with the broader literature, which supports biochar as an effective tool to stimulate the growth of hydrocarbon-degrading microbes.

The hydrocarbon-utilizing microbial population peaked at week 3 for the biochar-amended soils and declined afterward. **Recent** findings confirm the trend of microbial populations peaking 20–30 days post-contamination. A 2021 study by Zhao et al. noted that microbial communities typically reach optimal degradation activity within 3–4 weeks after exposure to petroleum hydrocarbons when combined with nutrient amendments or biochar. Another finding by Zhang et al. (2021) highlighted that biochar not only enhances microbial activity initially but also retains oil contaminants, making them less bioavailable as time progresses. As a result, the reduction in microbial population is natural once the easily degradable hydrocarbons are consumed. This is consistent with the patterns of these findings from week 1 to week 3, where microbial populations increased with biochar application.

However, in the control treatments (without biochar),the microbial populations may continue to grow for longer periods if the oil contamination persists. This is due to the ongoing availability of hydrocarbons as a carbon source, as noted in recent studies such as one by Wang et al. (2019), where untreated oil-contaminated soils showed prolonged microbial activity compared to amended soils. These microbes rely on the abundant carbon source provided by crude oil, facilitating sustained growth.

The higher microbial count in soils remediated with plantain trunk biochar, as compared to non-remediated control soil, is consistent with recent findings. Studies support that biochar not only improves soil structure but also enhances microbial activity due to its nutrient content, especially nitrogen and phosphorus, which are essential for microbial growth and biodegradation processes. A recent study by Ibrahim et al. (2020) similarly observed increased microbial counts in biochar-amended soils, attributing it to the improved availability of nutrients, aeration, and microbial habitats provided by biochar.

Biochar's porous structure allows for better retention of nutrients like nitrogen and phosphorus, as also reported by Awasthi et al. (2021), which leads to a conducive environment for the proliferation of hydrocarbon-degrading microbes. The study confirmed that biochar amendments enhance the microbial community's ability to degrade hydrocarbons, thus aligning with these findings.

The discrepancies in microbial counts between this study and those of Ibiene et al. (2011), who reported that the total culturable hydrocarbon-utilizing bacterial count in crude oil-contaminated soil ranged between x 103 Cfu/g to x 10^6 Cfu/g, Ijah and Antai (2003), who observed counts of hydrocarbon degraders in oil-polluted soil to be $\times 10^6$CFU/g can be attributed to variations in the soil's microbial ecology, which recent studies, such as one by Sharma et al. (2019), confirm can be heavily influenced by factors such as soil composition, organic matter, and the extent of contamination. Experimental soil conditions also significantly affect microbial population dynamics, which can result in variations in hydrocarbon-degrader counts across different studies.

4. Conclusion

The findings from this study demonstrate that plantain trunk biochar amendments significantly enhance the population of hydrocarbon-degrading microorganisms in contaminated soils compared to unamended control soils. This observation aligns with recent research indicating that biochar stimulates the growth of these microbes through a process known as biostimulation, where environmental conditions are optimized to support microbial activity. The porous structure of biochar enhances soil aeration and water retention, creating a favorable environment for microbial proliferation. Additionally, biochar contains essential nutrients such as nitrogen and phosphorus, which further fuel microbial metabolism, thus promoting the growth of hydrocarbon-utilizing bacteria and fungi.

This increase in microbial activity in biochar-amended soils could be attributed to several factors. First, biochar modifies soil properties by improving its texture, water retention, and nutrient availability, which directly influences microbial habitats. Studies, such as those by Lu et al. (2019), show that biochar application creates microhabitats within its porous structure that protect microbial cells from harsh environmental conditions, allowing them to thrive and actively degrade hydrocarbons.

Second, biochar's chemical composition, particularly its high carbon content, provides an additional energy source for microbial communities. As documented by Zhang et al. (2021), the carbon-rich nature of biochar promotes microbial respiration and biodegradation processes. By offering both physical shelter and a nutrient source, biochar supports sustained microbial activity, making it a valuable tool in bioremediation.

These findings emphasize the potential benefits of biochar as an environmentally friendly solution for soil remediation. The enhanced microbial populations not only improve the biodegradation of hydrocarbons but also contribute to the overall health of the soil ecosystem. In addition to remediation, the application of biochar can play a role in sustainable land management practices by enhancing soil fertility and microbial biodiversity, both of which are crucial for long-term soil health and productivity.

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