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BIOREMEDIATION OF TOTAL MONOCYCLIC AROMATIC HYDROCARBONS IN OIL-BASED DRILL CUTTINGS USING POWDERED *Eichhornia crassipes*

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ABSTRACT

Oil-based drill cuttings (OBDCs) contain high concentrations of toxic total monocyclic aromatic hydrocarbons (TMAHs), making them among the most hazardous wastes produced during petroleum exploration. This study evaluated the potential of powdered *Eichhornia crassipes* as a natural organic amendment for enhancing the bioremediation of TMAHs in OBDCs. A completely randomized design (CRD) with five treatment levels was adopted, consisting of a control (0 g) and four amendment levels (30 g, 50 g, 70 g, and 90 g) of dried water hyacinth powder mixed with 1000 g of loamy soil and 500 g of contaminated drill cuttings. Total heterotrophic bacteria (THB), soil pH, and TMAH concentrations were monitored throughout a 50-day aerobic remediation period. Results showed a significant increase in microbial activity in the amended treatments, with THB counts of (3.95×10^3 cfu/g) reaching their peak on Day 30. The highest reduction in TMAH concentration (96.36%) occurred in the 90 g treatment, while the control recorded only 30.48% reduction. The amendment also helped maintain a favourable soil pH for microbial activity. Statistical analysis indicated that amendment dosage and remediation period significantly affected remediation efficiency ($p < 0.05$). These findings demonstrate that *Eichhornia crassipes* can serve as a cost-effective and environmentally sustainable amendment for remediating hydrocarbon-contaminated soils.

Keywords: Bioremediation, Microbial Stimulation, Monocyclic Aromatic Hydrocarbons, Oil-Based Drill Cuttings, Organic Amendment

1.0 INTRODUCTION

The oil-based drill cuttings (OBDCs) production which are made up of rock fragments, drilling fluids, and other chemical additives, has significantly increased as a result of the rapid growth benzene, toluene, ethylbenzene, and xylene (BTEX), which are known for their toxicity, volatility, and environmental perseverance, are among the hazardous substances that are often found in these cuttings (Abioye et al., 2023; Anoliefo and Ikhajiagbe, 2022). These substances provide major ecological and public health hazards for the reason that they can leak into the groundwater and soil when they are not disposed of properly (Okonkwo et al., 2021). The more the crude oil is present in the soil, the more the physicochemical properties of the soil alter, leading to an imbalance in the nature of the soil (Ekemube et al., 2022). Therefore, remediation should be used to restore the soil's originality, according to their findings. In spite of their effectiveness, traditional remediation techniques such as; landfilling, chemical oxidation, and cremation are often costly, energy-intensive, and can produce secondary pollutants (Zhao et al., 2023). By means of using microorganisms' innate ability to degrade hazardous contaminants into non-toxic by-products, bioremediation has become a feasible and affordable substitute (Ite et al., 2022). Though, environmental factors including microbial population dynamics, pH, oxygen, and nutrition availability have a significant influence the effectiveness of bioremediation.

In recent times, there has been a lot of interest in the application of organic amendments to improve bioremediation, commonly known as biostimulation. Additional to providing essential nutrients, organic materials improve soil structure, boost microbial activity, and act as conduits for microbial consortia (Rani and Shara, 2022). The fast-growing aquatic macrophyte *Eichhornia crassipes*, as well known as water hyacinth, has been recognized as a promising amendment among these materials because of its high nutrient content, quick biomass production, and inherent capacity to absorb hydrocarbons and heavy metals (Al-Baldawi et al., 2022; Ezeokoli et al., 2024). Due to its high concentrations of nitrogen, phosphate, potassium, and volatile organic matter, water hyacinth has been used in wastewater treatment and phytoremediation of damaged areas in the past (Chukwuma et al., 2023). These elements aid to activate the natural microbial communities that break down hydrocarbons (Raindran et al., 2021). Furthermore, the fibrous matrix of the plant may hold onto moisture and oxygen, creating an environment that is conducive to aerobic microbial activity. In spite of these benefits, there hasn't been much research done on the use of water hyacinth in oil-based drill cutting clean-up, specifically when it comes to TMAHs. There is a knowledge gap in the remediation of more volatile and mobile monocyclic chemicals since the majority of current research has been on total petroleum hydrocarbons (TPHs) or polycyclic aromatic hydrocarbons (PAHs) (Adebola et al., 2022). Besides, further research is still needed in the areas of application rate and treatment duration optimization.

By filling this knowledge gap, the study provides information on natural, low-cost methods for cleaning up hydrocarbon-polluted areas in oil-producing areas. The aim of this study is to evaluate the effectiveness of powdered *Eichhornia crassipes* as an organic amendment for the bioremediation of TMAHs in oil-polluted drill cuttings. Its specific objective is to ascertain how different concentrations of water hyacinth affect microbial development, pollutant breakdown, and soil property changes throughout a remediation period of fifty days.

2.0 MATERIALS AND METHODS

2.1 Study Materials

Water hyacinth (*Eichhornia crassipes*) was collected from a stagnant pond located in Etegwe community, Bayelsa State, Nigeria. The collected plants were washed thoroughly to remove sediments and debris before being processed. Oil-based drill cuttings were obtained from a drilling waste treatment facility located in Onne, Rivers State, Nigeria, while loamy soil used for the remediation experiment was collected from the Agricultural Research Farm of Rivers State University, Port Harcourt.

2.2 Preparation of Bioremediation Materials

The collected 30 kg of water hyacinth plants were washed thoroughly to remove debris and sediments and then air-dried under shade (**26°C to 28°C)** for seven days in order to preserve their nutrient composition. After drying, the plant material was ground into a fine powder (0.075 mm) using a Model GX-160 mechanical gri2nder (Honda Motor Co., Japan). The soil samples were sieved using a 2-mm stainless steel sieve (Endecotts Ltd., UK) to remove stones and coarse particles, while the drill cuttings were homogenized to ensure uniform contamination prior to mixing.

2.3 Experimental Design and Set Up

A completely randomized design (CRD) consisting of five treatment groups was adopted for the remediation experiment. The treatments included four amendment levels of water hyacinth powder (30 g, 50 g, 70 g, and 90 g) and a control treatment without amendment (0 g). For each treatment, **1000 g of soil and 500 g of drill cuttings** were thoroughly mixed in **5-L plastic containers (diameter: 25 cm; height: 30 cm)**. The respective quantities of powdered water hyacinth were then incorporated into the mixtures. Each treatment was conducted in triplicate. The containers were maintained under aerobic conditions at room temperature (25–35°C) by manually turning the contents every **72 hours for approximately 2–3 minutes** to ensure proper aeration. Moisture content was maintained by adding **250 mL of distilled water every three days** throughout the **50-day remediation period** (Taleat et al., 2026).

2.4 Analytical Procedures

Soil samples were collected at 0, 10, 20, 30, 40, and 50 days during the remediation period. The following parameters were analyzed: Total Monocyclic Aromatic Hydrocarbons (TMAH), Soil pH, Total Nitrogen (TN), Total Organic Carbon (TOC), Volatile Matter, Phosphorus, Potassium, Total Heterotrophic Bacteria (THB), Ash Content.

2.4.1 Total Monocyclic Aromatic Hydrocarbons (TMAH)

TMAH analysis was performed using an **Agilent 7890B Gas Chromatograph coupled with a 5977A Mass Selective Detector (GC-MS) and a 7697A Headspace Autosampler (Agilent Technologies, USA)** following the **USEPA Method 8260C**. The instrument was calibrated using certified BTEX standard solutions (AccuStandard, USA). The GC oven was initially maintained at **35°C for 10 min**, followed by a temperature ramp of **10°C/min to 110°C**, where it was held for **0.5 min**. Helium was used as the carrier gas at a flow rate of **40 mL/min**. Quantification of TMAHs was achieved by comparing the peak areas with those of the calibration standards.

2.4.2 Soil pH

The pH of the soil was measured in a 1:1 soil-to-water mix using a glass electrode pH meter. A glass electrode pH meter and a 1:1 soil to water suspension were used in the lab to determine the pH values of the soil samples (ASTM D4972) with a +0.01 sensitivity.

2.4.3 Total Nitrogen (TN)

Total nitrogen (TN) in the soil samples was determined using the **Kjeldahl digestion and distillation method in accordance with ASTM D1426**. Approximately **1.0 g of the air-dried soil sample** was weighed into a Kjeldahl digestion flask. A volume of **10 mL of concentrated sulphuric acid (H_2SO_4)** was added to the sample and the mixture was gently swirled to ensure proper mixing. To accelerate digestion, **2.5 g of potassium sulphate (K_2SO_4)** catalyst was added to the mixture. The flask was then heated gradually on a digestion block until the solution became clear, indicating complete digestion of organic nitrogen. The digestion temperature was maintained at approximately **380 - 400°C**. After digestion, the solution was allowed to cool and diluted with distilled water before being subjected to **alkaline distillation**. The liberated ammonia was collected in a boric acid solution and titrated with standardized hydrochloric acid to determine the nitrogen concentration. The total nitrogen content was expressed in **mg/kg of dry soil (**Bremner, 1996; ASTM International, 2015).

2.4.4 Total Organic Carbon (TOC)

Total organic carbon (TOC) was determined using the Walkley–Black dichromate oxidation method (ASTM D4129). A 0.5 g air-dried soil sample was weighed into a 250 mL conical flask. Approximately 25 mL of 0.067 M potassium dichromate ($K_2Cr_2O_7$) solution was added, followed by 20 mL of concentrated sulphuric acid (H_2SO_4). The mixture was gently swirled and allowed to stand for 30 minutes to ensure complete oxidation of organic carbon present in the sample. After cooling, 100 mL of distilled water was added to dilute the solution. Subsequently, 5 mL of diphenylamine indicator was introduced, and the excess dichromate was titrated with 0.5 M ferrous ammonium sulphate ($Fe(NH_4)_2(SO_4)_2$) solution until a colour change from violet-blue to green was observed. The amount of organic carbon in the sample was calculated based on the volume of ferrous ammonium sulphate used during titration and expressed as percentage organic carbon (%) (Walkley & Black, 1934; Nelson & Sommers, 1996).

2.4.5 Volatile Matter

Volatile matter content of the water hyacinth sample was determined using the gravimetric method (ASTM D3695). Approximately 2 g of the dried water hyacinth powder was weighed into a porcelain crucible with a fitted lid. The crucible was placed in a muffle furnace (Carbolite ELF 11/6, UK) and heated at a temperature of 550°C for 10 minutes. After heating, the crucible was carefully removed and placed in a desiccator to cool to room temperature. The loss in weight after heating represented the volatile matter released from the sample. The volatile matter content was calculated as a percentage of the original sample weight (Speight, 2015; ASTM International, 2019).

2.4.6 Phosphorus

Available phosphorus in the soil samples was determined using the ascorbic acid molybdenum blue method. Approximately 25 mL of the prepared soil extract was transferred into a 50 mL volumetric flask. To this solution, 5 mL of ammonium molybdate reagent and 2 mL of hydrazine sulphate solution were added. The mixture was thoroughly shaken and diluted to the 50 mL mark with distilled water. The flask was then placed in a boiling water bath at 100°C for 10 minutes to facilitate colour development. After heating, the solution was cooled rapidly using an ice bath. The absorbance of the resulting blue-coloured complex was measured at 830 nm using a UV-Visible spectrophotometer (Shimadzu UV-1800, Japan) with a 1 cm quartz cuvette. The phosphorus concentration was determined by comparison with a standard calibration curve and expressed in mg/kg (Murphy & Riley, 1962; APHA, 2017).

2.4.7 Potassium

Potassium concentration in the soil samples was determined using flame atomic absorption spectrophotometry (AAS) following APHA (1988) Method 3111C. Approximately 0.25 g of the dried soil sample (particle size < 2 mm) was weighed into a digestion vessel and digested using 9 mL of concentrated nitric acid (HNO_3) and 3 mL of hydrofluoric acid (HF). The digestion was carried out in a WX-6000 microwave digestion system (Persee Analytical, China) at a temperature of 95°C for 15 minutes. After digestion, the mixture was filtered and transferred into a 100 mL volumetric flask. The residue was further rinsed using 5 mL hydrochloric acid (HCl) and deionized water before being diluted to the final volume. The potassium concentration in the extract was measured using a UNICAM-969 Atomic Absorption Spectrophotometer (Thermo Scientific, UK) at a wavelength of 766.5 nm. Calibration was performed using standard potassium solutions. The final potassium concentration was expressed in mg/kg (APHA, 1988; Skoog et al., 2018).

2.4.8 Ash Content

Ash content of the sample was determined using the dry ashing method according to AOAC (1999). Approximately 0.5 g of the dried sample was weighed into a pre-weighed porcelain crucible. The crucible was placed in a muffle furnace (Carbolite ELF 11/6, UK) and heated at a temperature of 550°C for 8 hours to ensure complete combustion of organic matter. After ashing, the crucible was removed from the furnace and cooled in a desiccator to prevent moisture absorption. The crucible was then weighed again, and the ash content was calculated as the percentage of the residue remaining after combustion relative to the original sample weight (ASTM International, 2017).

2.5 Percentage Reduction TPH

Based on the starting and ending concentrations of the pollutant (TMAH) in the soil, the percentage decrease was computed using Equation 1 (Ekemube et al., 2025).

$$\%_R = \frac{\mathrm{Ci} - \mathrm{Cf}}{\mathrm{Ci}} \times 100 \qquad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \text{(1)}$$

Where:

$\%_R$ = percentage reduction (%)
Ci = initial concentration of TMAH in the soil sample (mg/kg)
Cf = final concentration of treated TMAH in soil sample (mg/kg)

2.6 Analysis of Statistics

The Microsoft Excel 2021 version was adopted for all statistical analyses in this study. The effects of powdered water hyacinth dosage and treatment period on TMAH degradation were evaluated using two-way analysis of variance (ANOVA) with a 95% significant level ($p \leq 0.05$) (Taleat et al., 2026).

3.0 RESULTS AND DISCUSSION

3.1 Initial Physico-Chemical and Microbial Properties of the Materials

The baseline physicochemical characteristics of the materials used in the remediation experiment are presented in Table 1. The uncontaminated soil exhibited a slightly acidic pH of **6.7** and consisted predominantly of 83.6% sand, 11.6% silt, and 4.8% clay, classifying it as loamy sand soil. Such soil texture is favourable for microbial activity because it enhances aeration and facilitates microbial colonization during aerobic biodegradation processes (Al-Dhabaan, 2022). Water hyacinth powder showed high organic nutrient content, including 12.4 mg/kg nitrogen, 0.58 mg/kg phosphorus, and 2.32 mg/kg potassium, as well as 30.29% volatile matter, indicating its suitability as an organic amendment capable of stimulating microbial activity. These nutrients are essential for microbial metabolism and are known to enhance hydrocarbon biodegradation (Al-Baldawi et al., 2022). In contrast, the drill cuttings exhibited a high TPH concentration of 17,224.71 mg/kg and TMAH concentration of 13.5 mg/kg, exceeding acceptable environmental limits and indicating severe hydrocarbon contamination. These findings are consistent with previous reports on petroleum-contaminated soils (Olajire, 2020; Taleat et al., 2026).

Table 1: Physical-chemical characteristics of clean soil, water hyacinth, and drill cuttings utilized in the cleanup procedure

S/N	Parameters	Uncontaminated Soil	Water Hyacinth	Drill Cuttings
1	pH	6.7	6.5	7.5
2	TPH, mg/kg	43.76	-	17224.71
3	Total organic carbon (%)	0.29	0.22	0.32
4	Volatile Matter (%)	-	30.29	-
5	Total Nitrogen (mg/kg)	15.52	12.4	11.12
6	Ash Content %	18.55	15.23	-
7	Organic Matter (%)	6.9	4.56	-
8	Phosphate (PO4) (mg/kg)	0.87	0.58	-
9	Potassium (mg/kg)	17.07	2.32	-
10	THB ($X10^{-3}$ cfu/g)	0.55	5.89	0.23

3.2 Microbial Response During Remediation

Comparing the water hyacinth application to the control, the THB population increased significantly throughout all altered treatments. Around Day 30, the 30 g amendment reached a high of 3.95×10^3 cfu/g, indicating peak microbial activity, whereas the control treatment plateaued at 1.61×10^3 cfu/g (Table 2 and Figure 1). These results imply that by increasing the carbon content and nutrient availability in the soil matrix, the organic amendment promoted microbial proliferation. This agreed with the finding of Usman et al. (2021).

However, the number of THB start to drop by Day 30, especially at higher dosages. The natural ageing phase in microbial life cycles, the accumulation of toxic intermediates, or nutrient reduction could all be accountable for this decline (Olanrewaju et al., 2023). Other organic-amended remediation experiments have displayed similar patterns, with microbial activity climaxing early and then declining as a result of substrate exhaustion (Njoku et al., 2022).

Table 2: THB count on drill cuttings contaminated soil

S/N	Treatment	3 days	10 days	20 days	30 days	40 days	50 days
		X10^3 cfu/g					
1	0g RM	0.23	0.5	0.91	1.61	0.61	0.42
2	30g RM	0.22	3.1	3.77	3.95	2.1	1.42
3	50g RM	0.23	1.61	2.65	3.01	1.08	0.92
4	70g RM	0.22	1.21	2.31	2.61	1.01	0.75
5	90g RM	0.24	0.92	1.1	1.44	0.91	0.62

RM – Remediation Material

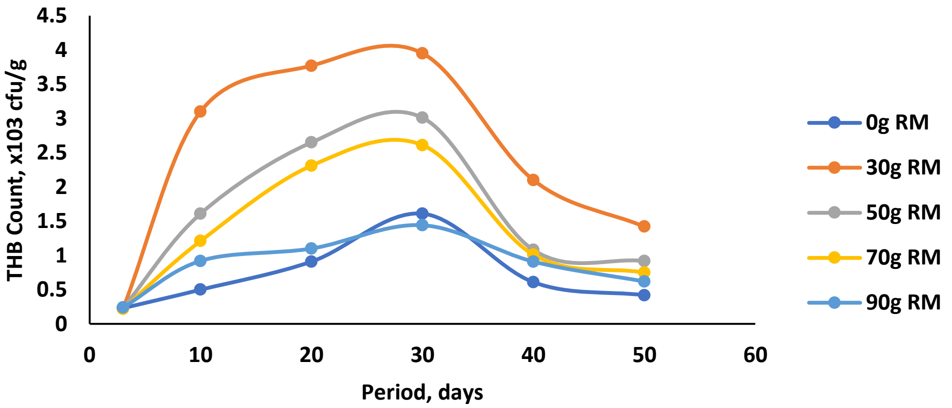

Figure 1: Effect of THB on TPAH

3.3 Reduction of TMAH Concentration

All over the 50-day treatment period, all water hyacinth-amended treatments showed a significant reduction in TMAH concentrations in the contaminated soil (Table 3 and Figure 2). The maximum dosage of powdered water hyacinth (90 g) allowed for a 96.36% reduction in TMAH, but the unaltered control only managed a 30.48% drop (Table 4 and Figure 3). This implies that the efficacy of remediation and amendment dosage are strongly positively correlated. The observed pattern can be clarified by the fact that water hyacinth contains vital nutrients that stimulated the growth of bacteria that break down hydrocarbons, which in turn sped up the degradation of TMAHs. This is in line with the findings of Adebola et al. (2022), who revealed that nutrient-rich organic matter encourages microbial hydrocarbon degradation. Amid Days 10 and 30, the decrease was at its fastest, suggesting a time of increased microbial enzymatic activity. Resulting from this, degradation slowed because of a potential drop in nutrients and bioavailable hydrocarbons. There is a high F-values which demonstrated the validity of the observed trends, and the two-way ANOVA results showed that the treatment dose and duration had statistically significant effects on the decrease of TMAHs ($p < 0.05$) (Table 5). The reputation of water hyacinth dosage and duration in determining cleanup performance is confirmed by this statistical validation (Abioye et al., 2023; Taleat et al., 2026).

Table 3: TMAH Concentration during treatment period

S/N	Treatment	3 days	10 days	20 days	30 days	40 days	50 days
		mg/kg					
1	0g RM	1.05	1.043	0.983	0.878	0.827	0.73
2	30g RM	1.05	0.882	0.699	0.406	0.289	0.177
3	50g RM	1.04	0.632	0.527	0.331	0.238	0.116
4	70g RM	1.02	0.567	0.434	0.238	0.175	0.089
5	90g RM	1.03	0.52	0.316	0.194	0.103	0.0375

RM – Remediation Material

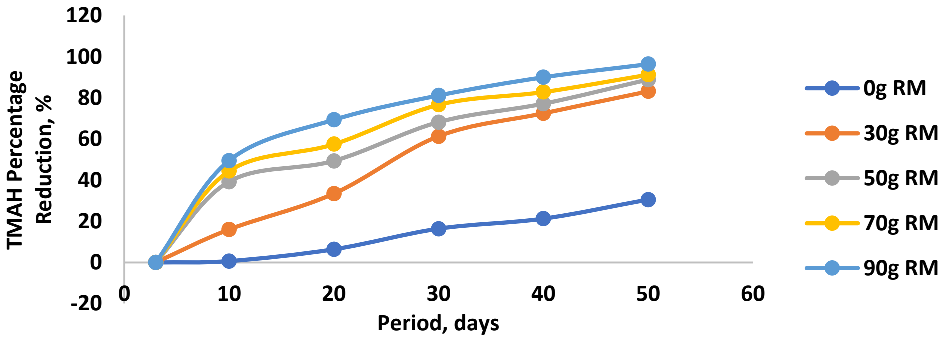

Figure 2: TMAH Concentration during Treatment Period

Table 4: TMAH percentage reduction

S/N	Treatment	3 days	10 days	20 days	30 days	40 days	50 days
		%					
1	0g RM	0	0.667	6.381	16.381	21.238	30.476
2	30g RM	0	16.00	33.429	61.333	72.476	83.143
3	50g RM	0	39.230	49.327	68.173	77.115	88.846
4	70g RM	0	44.412	57.451	76.667	82.843	91.275
5	90g RM	0	49.515	69.320	81.165	90.000	96.359

RM – Remediation Material

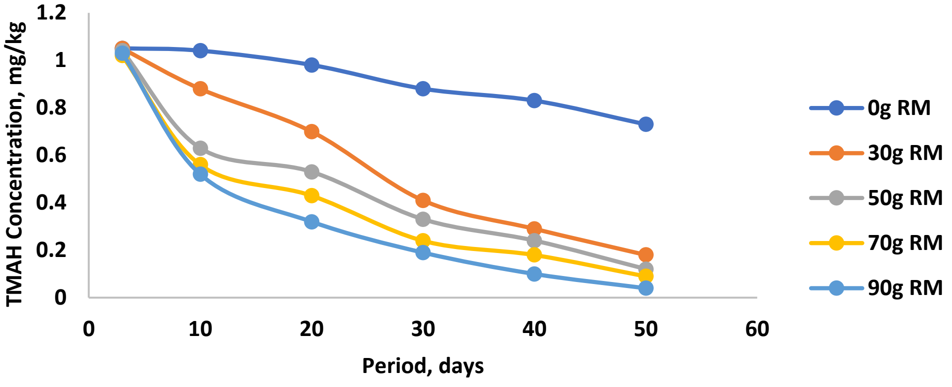

Figure 3: TMAH Concentration during Remediation Period

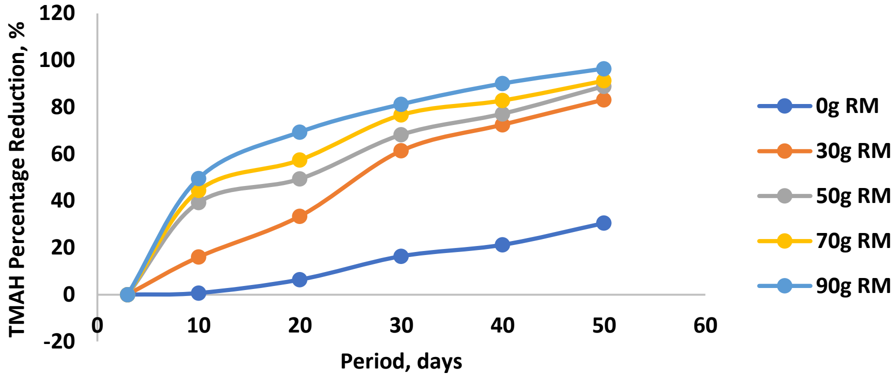

Figure 4: TMAH Percentage Reduction

Table 5 Analysis of variance result

Source of Variation	SS	df	MS	F	P-value	F crit
Rows	1.152077	4	0.288019	20.83013	6.7E-07	2.866081
Columns	2.219958	5	0.443992	32.11036	6.73E-09	2.71089
Error	0.276541	20	0.013827			
Total	1.02E+09	29				

3.4 Changes in Soil pH During Remediation

The whole treatments showed a decreasing trend in soil pH, most likely as a result of organic acids and other by-products of microbial metabolism (Table 6). In difference to the control, whose pH dropped more quickly, the rehabilitated treatments kept their pH rather constant and close to neutral. The 90 g treatment's pH reduced from 6.5 to 5.84 by Day 50, while the control treatment's pH reduced from 6.7 to 4.15. The organic matter and ash content of the water hyacinth may have a buffering effect that aids balance acidity, which could explain the moderate pH in the altered samples (Ezeokoli et al., 2024). Hydrocarbon bioavailability and microbial enzyme activity be subject to maintaining a pH close to neutral. Breakdown efficiency can be hindered by extreme acidity or alkalinity, which can also limit microbial function (Ite et. al (2022). Water hyacinth's use as alleviating supplement in bioremediation systems is further supported by its ability to control soil pH.

Table 6: pH levels of the contaminated soil

S/N	Treatment	3 days	10 days	20 days	30 days	40 days	50 days
1	0g RM	11.95	11.12	10.88	9.29	7.83	7.61
2	30g RM	12.07	10.47	10.09	8.98	6.92	6.72
3	50g RM	12.31	9.62	8.4	7.58	5.88	5.64
4	70g RM	12.36	7.63	6.93	6.04	4.39	4.15
5	90g RM	12.32	5.83	5.73	5.54	4.19	4.09

RM – Remediation Material

4.0 CONCLUSIONS

This study investigated the enhanced bioremediation of total monocyclic aromatic hydrocarbons (TMAHs) in oil-based drill cuttings using powdered *Eichhornia crassipes* as a natural organic amendment. The results demonstrated that the application of water hyacinth significantly improved microbial activity, as evidenced by the increased total heterotrophic bacteria (THB) counts observed during the remediation period. Among the treatment levels evaluated, the **90 g amendment exhibited the highest remediation efficiency**, achieving **96.36% reduction in TMAH concentration after 50 days**. The improved biodegradation performance can be attributed to the nutrient-rich organic matter provided by water hyacinth, which stimulated microbial metabolism and enhanced hydrocarbon degradation. Furthermore, the amendment contributed to maintaining a favourable soil pH environment that supports microbial activity during biodegradation. Statistical analysis confirmed that both amendment dosage and remediation time significantly influenced TMAH degradation. Overall, the results demonstrate that powdered water hyacinth is an effective, sustainable, and low-cost amendment for the remediation of hydrocarbon-contaminated drill cuttings. Optimizing amendment dosage and treatment duration is therefore essential for achieving efficient and economically viable remediation outcomes.

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