Sustinere
Journal of Environment and Sustainability
Volume 8 Number 3 (2024) 346-356

Print ISSN: 2549-1245 Online ISSN: 2549-1253
Website: https://sustinerejes.com E-mail: sustinere.jes@uinsaid.ac.id

RESEARCH PAPER

Evaluation of the phytoremediation potential of *T. spathecea* and *P. balforiana* grown in crude oil polluted soils

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Article history:
Received 22 June 2024 | Accepted 30 October 2024 | Available online 30 December 2024

Abstract. The impact of crude oil contamination on the environment, especially on soil quality, has been a major concern in Nigeria and for regulatory bodies worldwide. This study focused on assessing the phytoremediation potential of two plant species *T. spathecea* and *P.balfouriana*, grown on crude oil-contaminated soils. The plant were cultivated in soils polluted with crude oil at concentrations of 0, 50, 100, 150 and 200 mL/kg. The research was laid out in a 5 x 5 factorial experiment with three replicates, in a completely randomized designed to give a total of 75 experimental units. The gas chromatographic flame ionization detector (GC-FID) was used in determining the total petroleum hydrocarbon (TPH), the atomic absorption spectrophotometer (AAS) was used to determine the heavy metals. The relevant analytical techniques of APHA and AOAC (2020) was used to determine the phytochemical contents of the plant species. The results of TPH analysis revealed that the concentration of TPH increased with the rising concentration of crude oil pollutant in the soil. TPH values were 1,120 mg/kg for *P. balfouriana* and 2,762.79 mg/kg for *T. spathecea*. Both plant species showed considerable growth despite increasing crude oil concentrations. The results of heavy metals phytoextraction indicated a trend of increasing metal concentrations with increasing crude oil pollution. Copper concentration ranged from 2.28 mg/kg in *T. spathecea* to 12.64 mg/kg in *R. simplex*. Iron uptake ranged from 11.36 mg/kg in *P. balfouriana* to 22.40 mg/kg *T. spathcea*. The uptake of manganese increased from 5.56 mg/kg in *P. balfouriana* to 10.56 gm/kg in *T. spathecea*, while cobalt concentration decreased from 0.08 mg/kg in *T. spathecea* to 0.04 mg/kg in *P. balfouriana*. The study highlights the potential of *P. balfouriana* and *T. spathecea* for phytoremediation in crude-contaminated soils and advocates for their use as effective tools for soil remediation.

Keywords: Phytoremediation; Total hydrocarbon; Heavy metals; Phytochemicals

1. Introduction

Crude oil is a naturally occurring mixture that consist mainly of hydrocarbons, and the global economy has been highly dependent on it as a source of energy since its discovery (Griffiths et al.,

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DOI: https://doi.org/10.22515/sustinere.jes.v8i3.418

2022). Crude oil can be refined to produce a wide range of petroleum products, exerting a substantial influence on both the industrial and transportation sectors (Htay & Jatta, 2021).

As one of Africa's major oil producers and exporters, Nigeria's economy is characterized by its overwhelming reliance on crude oil revenues to fund public projects and cover operational costs (Kamer, 2023). The substantial contribution of the oil sector to Nigeria's Gross Domestic Product (GDP) underscores the country's dependence on crude oil. However, the exploration, production, and export of crude oil have had profound impact on the economy of Nigeria, which has a substantial effect on several sectors and aspects of the nation's development, particularly through environmental pollution (Chukwuma-ekwueme, 2023). Soil and water pollution from crude oil and its by-products has emerged as a major global environmental concern, particularly in oil producing communities in the Niger Delta region of Nigeria.

The presence of high concentrations of toxic compounds in crude oil makes it a significant environmental pollutant, altering the physio-chemical and biological properties of soil, as recognized by the US Environmental Protection Agency (Omotosho et al., 2018). The degradation rate of crude oil in soil is significantly slower than that of conventional carbon sources, taking substantially longer to break down, especially under severe conditions (Ali et al., 2020).

Traditional fishing in the Niger Delta can no longer serve as a reliable means of sustenance. As a result, rural dwellers are increasingly turning to crop cultivation or supplementing fishing with farming. However, the productive capacity of arable land in the Niger Delta is under constant threat from crude oil pollution, with reports suggesting that the land is rapidly losing its fertility (Oyedeji, et al., 2015). Hectares of farmland in the region lie abandoned because sever degradation of their productive quality (UNEP, 2011). While crude oil pollution in the Niger Delta has been attributed to the criminal negligence of both local and international oil companies, as well as the rising activities of crude oil bunkering and artisanal crude oil operations (Yabrade & Tanee, 2016), there is an urgent need for ecosystem recovery in the region.

To address the environmental implications of crude oil pollution in the Niger Delta, researchers are seeking affordable, efficient, and locally available methods to treat petroleum hydrocarbon-polluted soil, ensuring a safer and more sustainable future. Restoring the region's ecosystem and food security requires both biological and chemical cleanup of contaminated matrices (UNEP, 2011; Zabbey et al., 2017). Although physicochemical methods such as soil washing and thermal desorption have been recommended for petroleum hydrocarbon soil remediation, their widespread adoption is impeded by high costs and restricted local applicability (Dadrasni et al., 2015). Moreover, these techniques typically often fail to ensure complete degradation or removal of contaminants, leaving residual pollutants behind.

Bioremediation, which utilizes microbes and plant-based materials to break down pollutants into less hazardous forms, presents a viable alternative that combines effectiveness with eco-friendliness and cost-efficiency (Nkereuwem et al., 2020). It harnesses the power of natural microbes or plants to degrade or neutralize harmful pollutants, leveraging biostimulation or bioaugmentation to enhance their effectiveness (Adeleye et al., 2019).

For successful soil remediation, bioremediation methods should employ plants or microorganisms with high chemical tolerance and pollutant accumulation capabilities (Oyedeji 2022). Several researchers have reported the benefits of using native plants for phytoextraction and phytostabilization, particular those with a fast growth rate, well-developed root system, large size, and high tolerance to pollutants (Oyedeji, 2022; Adeyemi & Adeyemi, 2020; Onwuna et al., 2022a; Onwuna et al., 2022b).

Plants suitable for the phytoremediation of crude oil-contaminated soil are those that can thrive in polluted conditions (Bamidele & Agbogidi, 2006). Thus, potential remediation plants must exhibit tolerance and robust growth in the presence of toxicants. *T. spathecea* and *P. balfouriana* have been observed proliferating on crude oil-polluted soil within the Ogoni region of the Niger Delta, with no reports of reduced genetic diversity. These plant species are well known

for their fast growth rates, deep root systems, and high biomass production. However, to the best of our knowledge, no studies have reported on their bioremediation potential. This study was therefore conducted to assess the phytoremediation potential of *T. spathecea* and *P. balfouriana* in the decontamination of crude oil-polluted soil.

2. Material and method

2.1. Study location

The experiment was conducted in Environmental Biotechnology Unit of the Department of Genetics and Biotechnology, University of Calabar, Calabar, Cross River State, Nigeria. The unit is well-equipped with facilities for conducting research in environmental biotechnology, making it an appropriate setting for evaluating the bioremediation potential of plant species.

2.2. Identification of plant

Two plant species of interest were obtained from different locations within Cross River State and identified by a taxonomist in the Herbarium unit of the Department of Plant and Ecological Studies, University of Calabar in January, 2024. The species identified were Balfour aralia (*Polyscias balfouriana*) and Boat lily (*Tradescantia spathacea* (Swartz)). The plants were propagated by stem cuttings and grown directly in plastic buckets. All the buckets were watered once daily by spraying to maintain sufficient soil moisture.

2.3. Collection of soil samples and crude oil

Soil sample were collected from three different locations within the University of Calabar, Calabar (Kwa River site, Biological Science, and Staff Quarters). However, these are areas have no history of hydrocarbon pollution. The crude oil used in the study was purchased from the Nigerian Agip Oil Company, Port Harcourt, River State.

2.4. Artificial soil pollution

The collected soil samples were bulked to form a composite soil sample. Five kilograms each of the composite soil were weighed and transferred into labelled plastic buckets with drainage holes at the base. Seventy-five plastic buckets were used in the experiment, which were divided into two experimental groups, each containing five plant species: Group A (*Polyscias balfouriana*), and Group B (*Tradescantia spathacea*). Each group was treated with five different concentrations of crude oil: 0 mL, 50 mL, 100 mL, 150 mL and 200 mL, with three replications for each concentration. After soil contamination, the soil in the various plastic buckets was left for two weeks, during which pre remediation was conducted. Planting was then done, and the plants were allowed to grow for a period of 3 to 4 months, from March to July.

2.5. Experimental design

The experimental design was carried out in a 5 x 5 factorial arrangement in a completely randomized design (CRD). Factor 1 was the plant species, with 5 levels representing different species: *P. balfouriana* (Balfour aralia) and *T. spathacea* (Boat lily). Factor 2 was the crude oil concentration, with 5 levels: 0 mL, 50 mL, 100 mL, 150 mL and 200 mL of crude oil.

2.6. Sample preparation for gas chromatography-flame ionization detection (CG-FID)

Samples were collected from the upper 10 cm of each of the plant material and wrapped in aluminum foil. The samples were stored in an ice chest before being transported to the laboratory for analysis. In the laboratory, the samples were stored in a refrigerator prior to analysis to preserve their integrity. The samples were air-dried for four days, after which they were macerated and sieved through a l mm sieve for total hydrocarbon determination. A 10 g of the homogenized sieved sample was extracted with 100 mL of analytical-grade dichloromethane (CH_2Cl_2). The dichloromethane extracts were cleaned by passing them through a column packed with cotton wool, 1:1 silica gel, and sodium sulfate. The resulting extract was concentrated using

a rotatory evaporator. Hewlett Packed 5890 series II gas chromatographic coupled with a flame ionization detector (FID) was used for the analysis. The hydrocarbon window-defining standard was purchased from AccuStandard, and the calibration curve was prepared by diluting the 500 ug/mL standard solution to concentrations of 300ug/mL, 200 ug/mL, and 100 ug/mL and then run on the GC-FID.

2.6.1 Determination of Total Hydrocarbon Content using gas chromatographic flame Ionization Detector (GC-FID)

The determination of total hydrocarbon (THC) was carried out using a Hewlett Packed 5890 series II gas chromatographic coupled with a flame ionization detector (FID). Separation of THC in the samples was accomplished using a DB-1 fused-silica capillary column (30 m x 0.32 mm).

Helium was used as the carrier gas at a flow rate of 0.45 mL/min, and hydrogen and air were used as the ignition gas. Sample injection was performed in the splitless mode with an injection volume of 1 µL. The GC oven temperature was optimized with an initial temperature of 50°C, held for 2 minutes, and then ramped at 10°C/min to 300°C. This temperature was maintained until the end of the run (47 minutes). The injection port temperature was set at 250°C, and the FID detector was set at 300°C. The oven equilibrium time was 1 minute, the maximum oven temperature was 350°C, and the ambient temperature was 25°C. The hydrogen and air flow rates were set at 40 mL/min and 400 mL/min respectively. Chromatographs were generated for each sample, with the area count plotted on the y- axis and the retention time on the x- axis. The area count represents the area and height of the peaks detected by the GC-FID, with the sample results matched to the calibration standard.

The retention time indicates the time taken for a particular compound in the sample to elute, corresponding to the retention time in the standard calibration curve. The peaks with carbon numbers represent compound present in the calibration standard that were detected in the samples analyzed, while the peaks without carbon numbers represents compound not present in the calibration standard and, therefore, could not be reported on the chromatogram.

The total run time, as inputted in the THC method of GC-FID, was 47 minutes. A calibration equation was automatically generated for each compound in the sample, and the formula for calculating the concentration of the compounds was also generated. The formula was used to calculate the concentrations of the individual compounds in each sample, which were then summed to give the total concentration of hydrocarbon present in each sample. The calculated concentrations are expressed in ng/µg.

2.7. Statistical analysis

The data obtained were subjected to analysis of variance (ANOVA), and significant means were then separated using least significance different (LSD) test. All data were statistically analyzed using statistical package for social science version 20.0 (SPSS Inc., Chicago, IL, USA).

3. Result and discussion

3.1. Results

3.1.1 Soil total hydrocarbon content using GC -FID

An investigation was conducted on the phytoremediation potential of the two test plants on soils contaminated with crude oil, using the soil THC reduction values as an index for measurement. The reduction value was obtained by subtracting the pre-remediation THC values of the soil for each test plant from the remediation result generated automatically using GC-FID. The samples were analyzed for 38 hydrocarbon components in the calibrated curve, and chromatographs were generated along with the results. In the chromatographs, peaks with n-alkanes indicate the presence of hydrocarbons, while the peaks without n-alkanes indicate the absence of hydrocarbons. N-alkane ranged from C2 - C38, along with isoprenoid hydrocarbons, Pristine (Pr), and Phytane (Ph). Some n-alkanes were absent in different plant species at different

concentrations. This variation in the presence and absence of n-alkanes may be due to the differences in the biodegradation processes across the plant species.

From the result analyzed, it was observed that the higher the mean the lower the n-alkanes in the chromatographs, while the lower the mean, the higher the n-alkanes in the chromatographs. The result revealed a significant variation in phytoremediation potential among the two test plants grown on soils of various pollution levels (50 mg, 100 mg, 150 mg and 200 mg), including the control (0 mg). Among the test plants, *T. spathacea* showed significantly higher potentials for hydrocarbon remediation in soils treated with 50 mg, 100 mg and 200 mg at $p < 0.05$. The mean soil THC reduction by *T. spathacea* was 2762.79 ± 32.63 ng/µg, 158.62 ± 0.83 ng/µg, and 1512.07±27.10 ng/µg for soils treated with 50 mg, 100 mg and 200 mg of crude oil, respectively, as shown in Table 1. However, at 150 mL, *P. balfouriana* showed a higher mean of 1120.60 ± 9.25 ng/µg.

Imperatively, the THC contents, it was observed that *T. spathecea* and *P. balfouriana*, which belonging to the grass and legume families, demonstrated the great ability to phytoaccumulate THC. A similar result was documented by Agbor et al., (2012); Ndimele (2010) who reported that grasses and legumes have high potential to remediate THC in a crude oil polluted environment. Thus, the results obtained in our study on the THC accumulation by plants align with the findings of Agbor et al. (2012), who reported that some selected plants bioaccumulate THC. A clear explanation to this could be the presence of extensive fibrous root system with a large root surface area, which can penetrate deep into the soil. The high levels of THC in the plants indicate significant absorption of petroleum hydrocarbons by the plants. The proportion of the THC concentrations in the plants shows that absorption and accumulation of petroleum hydrocarbons in plants depends on the concentration of the pollutant in the environment.

3.1.2 Heavy metal content in soil

The results for the various heavy metal contents in the soil are presented on Table 2. The analysis of heavy metals in the soil shows distinct variations in their concentrations based on plant species and crude oil contaminations levels.

a. *Cadmium*. The result as presented on Table 2 shows that the Cadmium (Cd) concentration in soil grown with *P. balfouriana* at 150 mL of crude oil was the highest. *T. spathacea* at 100 mL crude oil and control group showed no significant difference ($p > 0.05$) in the mean values obtained. The Cd concentration in soil grown with *T. spathacea* had the lowest mean values. For chromium (Cr), the highest concentration was found in the polluted soils grown with *P. balfouriana* at 100 mL of crude oil, followed by Cr concentrations in soils grown with *P. balfouriana* at 200 mL and 150 mL of crude oil. There was no significant difference ($p > 0.05$) in mean values between these concentrations, but were significantly higher ($p < 0.05$) than the Cr content in soil grown with *T. spathacea* (50 mL – 200 mL). The Cr content in soils grown with *P.* balfouriana and *T. spathacea* was the lowest, with no significant difference in mean values (Table 2).

b. *Zinc*. The results for zinc (Zn) show that the zinc content in soils polluted soils polluted with crude oil and grown with *T. spathacea* at 100 mL and 200 mL was the highest, with no significant difference ($p > 0.05$) in the mean values obtained. Soil grown with *P. balfouriana* at 200 mL, 50 mL of crude oil had had the lowest Zn content. The results indicate that the soil grown with *T. spathacea* had the highest Zn content, while the Zn content in soil grown with *P. balfouriana* was the lowest in terms of mean values.

c. *Copper*. The results for copper show that the copper content in soil grown with *T. spathacea* at 50 mL of crude oil-polluted soil significantly higher ($p < 0.05$). This was followed by the Cu in soil grown with *P. balfouriana* at 200 mL of crude oil-polluted soil, which was significantly

Table 1. Heavy metal concentration in soil of plant species grown in hydrocarbon polluted soil

Plant Species (mg/kg)	Cadmium (Cd)	Chromium (Cr)	Zinc (Zn)	Copper (Cu)	Iron (Fe)	Manganese (Mn)
Control	0.40b ± 0.05	0.022c ± 0.002	4.89d ± 0.03	2.46i ± 0.003	24.09c ± 0.64	9.19b ± 0.01
P. balfouriana 50 mL	0.14c ± 0.003	0.14b ± 0.003	3.68e ± 0.023	2.67i ± 0.002	10.36 ± 0.005	7.74b ± 0.001
P. balfouriana 100 mL	0.22c ± 0.04	0.32a ± 0.005	6.28c ± 0.02	3.42h ± 0.003	21.42c ± 0.50	9.86b ± 0.008
P. balfouriana 150 mL	0.40b ± 0.002	0.18b ± 0.002	8.48b ± 0.003	4.82f ± 0.002	11.45 ± 0.30	7.38b ± 0.011
P. balfouriana 200 mL	0.04d ± 0.003	0.24b ± 0.002	4.56d ± 0.006	8.61b ± 0.003	43.98a ± 0.34	8.56b ± 0.002
T. spathacea 50 mL	0.001d ± 0.00	0.0220c ± 0.04	4.98d ± 0.003	9.47a ± 0.00	17.58d ± 0.42	10.50b ± 0.003
T. spathacea 100 mL	0.39b ± 0.01	0.020c ± 0.002	10.14a ± 0.02	5.944e ± 0.01	47.99a ± 0.07	8.64b ± 0.01
T. spathacea 150 mL	0.01d ± 0.000	0.04c ± 0.05	8.68b ± 0.04	2.40i ± 0.17	31.58b ± 0.89	9.30b ± 0.05
T. spathacea 200 mL	0.001d ± 0.00	0.026c ± 0.01	9.86a ± 0.06	3.62h ± 0.01	16.04d ± 1.13	9.26b ± 0.03
LSD	0.04	0.03	0.48	0.36	2.70	0.91

*Means followed by same case superscript along vertical axis signify no significant differences

Table 2. Heavy metal concentration in soil of plant species grown in hydrocarbon polluted soil

Plant species (mg/kg)	Conc.	Cadmium (Cd)	Chromium (Cr)	Zinc (Zn)	Copper (Cu)	Iron (Fe)	Manganese (Mn)
	Control	0.40b ± 0.05	0.022c ± 0.002	4.89d ± 0.03	2.46 ± 0.003	24.09c ± 0.64	9.19b ± 0.01
P. balfouriana	50 mL	0.14c ± 0.003	0.14b ± 0.003	3.68e ± 0.023	2.67i ± 0.002	10.36 ± 0.005	7.74b ± 0.001
	100 mL	0.22c ± 0.04	0.32a ± 0.005	6.28c ± 0.02	3.42h ± 0.003	21.42c ± 0.50	9.86b ± 0.008
	150 mL	0.40b ± 0.002	0.18b ± 0.002	8.48b ± 0.003	4.82f ± 0.002	11.45 ± 0.30	7.38b ± 0.011
	200 mL	0.04d ± 0.003	0.24b ± 0.002	4.56d ± 0.006	8.61b ± 0.003	43.98a ± 0.34	8.56b ± 0.002
T. spathacea	50 mL	0.001d ± 0.00	0.0220c ± 0.04	4.98d ± 0.003	9.47a ± 0.00	17.58d ± 0.42	10.50b ± 0.003
	100 mL	0.39b ± 0.01	0.020c ± 0.002	10.14a ± 0.02	5.944e ± 0.01	47.99a ± 0.07	8.64b ± 0.01
	150 mL	0.01d ± 0.000	0.04c ± 0.05	8.68b ± 0.04	2.40i ± 0.17	31.58b ± 0.89	9.30b ± 0.05
	200 mL	0.001d ± 0.00	0.026c ± 0.01	9.86a ± 0.06	3.62h ± 0.01	16.04d ± 1.13	9.26b ± 0.03
LSD		0.04	0.03	0.48	0.36	2.70	0.91

*Mean with the same superscript along vertical arrays indicates no significant difference ($p > 0.05$).

higher than the Cu content in soil grown with *T. spathacea* at 100 mL, and 200 mL of crude oil-polluted soils, respectively.

d. *Lead*. The result for lead show that the lead content in soil grown with various concentrations of crude oil did not show a significant difference in the mean values obtained. Results for total heavy metal content indicate that the lead content in soil grown with *T. spathcea* was significantly higher ($p < 0.05$) than in soil grown with *P. Balfour Iana*. The iron content in soil grown with *P. balfouriana* (200 mL) and *T. spathacea* (100 mL) of crude oil-polluted soil was higher, but no significant difference ($p > 0.05$) was observed in the mean values obtained. Results for total heavy metal content show that the iron content in soil grown with *T. spathcea* was the highest, while the iron content in soil grown with *P. balfouriana* did not show a significant difference ($p > 0.05$) in the mean values obtained.

e. *Manganese*. The results show that the manganese content in the soil grown with *P. balfouriana* had the lowest Mn content.

f. *Nickel*. The nickel content in soil grown with *P. balfouriana* (100 mL, 200 mL) and *T. spathacea* at 50 mL of crude oil-polluted soil was the highest, with no significant change in the mean values. This was followed by the mean values obtained in the soil grown with *P. balfouriana* at 50 mL and 150 mL. However, a significant reduction in the Ni content of the soil was obtained in soil grown with *T. spathacea* at 100 mL and 200 mL of crude oil-polluted soil, with no significant difference ($p > 0.05$) in the mean values. The Ni content in soil with *T. spathacea* was the lowest, while the Ni content in soils grown with other plants showed no significant difference ($p > 0.05$) in mean values obtained.

3.1.3 Heavy metal in the stem of the plants

The results for the heavy metal contents in the stems of the plant species are presented in Table 3.

a. *Nickel*. The result of the nickel content in the stems of the plants are presented in Table 3. The results reveal that the nickel content in the stem of the control plants was significantly higher ($p < 0.05$) than the values obtained from the stems of the plants exposed to varying concentrations of crude oil. Among the treated groups with crude oil, the stems of *P. balfouriana* (50 mL, 100 mL, 150 mL, 200 mL), and *T. spathacea* at 200 mL had the lowest levels of nickel in the plants.

b. *Copper*. The copper content in the stems of *P. Balfour Iana* (50 mL, 150 mL), *T. spathacea* (200 mL), and the control had significantly high ($p < 0.05$) mean values. The lowest copper content in the stems of the plants was observed in *T. spathacea* (150 mL) of crude oil-polluted soil.

c. *Iron*. The results for iron shows that the stems of *T. spathacea* at 150 mL crude oil pollution had the highest iron content, with no significant difference ($p > 0.05$) compared to the controls. Generally, the iron content was low in the stems of *P. balfouriana* and *T. spathacea* at 200 mL crude oil pollution.

d. *Cobalt, Chromium and Lead*. The cobalt content in the stem of *P. Balfouriana* at 150 mL crude oil-polluted soil was the highest, while the stems of the other plants at varying concentrations and the controls showed no significant difference in the mean values. The chromium content was significantly reduced ($p > 0.05$) in the stem of *T. spathacea* at 100 mL soil pollution, while the stem of the control and the stems of other plants at varying concentrations showed no significant difference ($p > 0.05$) in their mean chromium content.

The lead (Pb) content was observed to be high in the stem of *T. spathacea* at 50 mL soil pollution, while the other groups and controls showed no significant difference ($p > 0.05$) in the mean values. Generally, there was no significant difference ($p > 0.05$) in the mean values of the cobalt, chromium and lead content in the stems of the plants.

Table 3. Heavy metal concentration in the stem of the plant

Plant species (Mg/kg)	Conc	Nickel (Ni)	Copper (Cu)	Iron (Fe)	Cobalt (Co)	Chromium (Cr)	Zinc (Zn)	Manganese (Mn)
	Control	3.86a ± 0.02	4.60a ± 0.05	20.48c ± 1.05	0.140b ± 0.2	0.34a ± 0.003	17.26a ± 0.10	10.28a ± 0.10
P. balfouriana	50 mL	0.14d ± 0.03	4.87a ± 0.02	17.4d ± 0.005	0.04b ± 0.00	0.22a ± 0.03	4.8c ± 0.006	9.72b ± 0.30
	100 mL	0.14d ± 0.01	3.2c ± 0.02	19.2c ± 0.60	0.42b ± 0.03	0.28a ± 0.04	9.18b ± 0.4	10.48a ± 0.40
	150 mL	0.08d ± 0.02	4.48a ± 0.03	16.32d ± 1.1	0.64a ± 0.02	0.36a ± 0.1	5.7c± 0.10	5.56f ± 0.20
	200 mL	0.046d ± 0.01	3.12c ± 0.3	15.28e ± 0.50	0.030b ± 0.01	0.16a ± 0.01	2.68d ± 0.02	10.36a ± 0.10
	200 mL	0.84c ± 0.10	4.2a ± 0.1	22.5b ± 0.06	0.014b ± 0.00	0.16a ± 0.05	6.58c ± 0.01	7.36e ± 0.2
T. spathacea	50 mL	0.36c ± 0.02	3.4c ± 0.04	16.74d ± 0.08	0.014b ± 0.03	0.180a ± 0.02	6.48c ± 0.04	10.56a ± 0.06
	100 mL	0.41c ± 0.02	3.1c ± 0.30	19.06a ± 0.30	0.026b ± 0.003	0.030b ± 0.00	4.5c ± 0.01	7.5e ± 0.60
	150 mL	0.41c ± 0.02	2.28d ± 0.20	22.40b ± 0.06	0.08b ± 0.00	0.32a ± 0.03	7.64c ± 0.03	8.6d ± 0.10
	200 mL	0.084d ± 0.02	4.16a ± 0.2	14.8e ± 0.04	0.042b ± 0.03	0.26a ± 0.02	3.82c ± 0.04	9.2c ± 0.10
LSD		0.12	0.23	0.74	0.01	0.06	0.84	0.38

*Mean with the same superscript along vertical arrays indicates no significant difference ($p > 0.05$).

Table 4. Phytochemical properties of the plants

Plant Species (mg/kg)	Alkaloid	Glycoside	Polyphenols	Reducing Compounds
Control	2.70a ± 0.006	2.54a ± 0.05	20.81b ± 0.006	7.180a ± 0.01
P. balfouriana 50 mL	2.54b ± 0.08	2.54a ± 0.09	18.46b ± 0.009	4.60b ± 0.009
P. balfouriana 100 mL	2.54b ± 0.08	2.44a ± 0.07	18.46b ± 0.08	4.60b ± 0.08
P. balfouriana 150 mL	2.30b ± 0.05	1.58c ± 0.08	12.38c ± 0.08	3.84c ± 0.006
P. balfouriana 200 mL	2.04c ± 0.05	2.04b ± 0.05	6.48d ± 0.009	1.74e ± 0.005
T. spathacea 50 mL	2.46d ± 0.006	2.43a ± 0.006	17.93b ± 0.05	4.58b ± 0.05
T. spathacea 100 mL	2.46d ± 0.006	2.43a ± 0.006	17.93b ± 0.005	4.58b ± 0.006
T. spathacea 150 mL	1.81d ± 0.080	1.57c ± 0.06	7.91d ± 0.009	2.86d ± 0.006
T. spathacea 200 mL	1.60d ± 0.06	1.36d ± 0.006	3.87e ± 0.006	1.60e ± 0.006
LSD	0.12	0.16	0.29	0.13

*Mean with the same superscript along the vertical arrays indicate no significant difference ($p > 0.05$)

e. *Zinc*. The results for zinc show that the control groups had significantly high ($p < 0.05$) zinc content, while a reduction in the zinc content was observed in the stem of *P. balfouriana* (100 mL). This was followed by the zinc content in the stems of *P. balfouriana* (50 mL, 150 mL), and *T. spathacea* (50 mL, 100 mL, 150 mL and 200 mL) with no significant difference ($p > 0.05$) in the mean values obtained.

f. *Manganese*. The manganese content in the control, *P. balfouriana* (100 mL, 200 mL), and *T. spathacea* (50 ml) was significantly high ($p < 0.05$), followed by the manganese content in the stem of *P. balfouriana* (50 mL) with no significant difference ($p > 0.05$) in the mean values obtained. The lowest manganese content was observed in the stem of *P. balfouriana* at 150 mL crude oil soil pollution. Overall, the total heavy metal in the stem of *P. balfouriana* was the highest.

As observed from the heavy metal analysis of the plants, petroleum products had a significant impact on the heavy metal levels in the soil. In this case, the contaminated soil had significantly higher total Cu, Fe, Ni, Mn, and Zn values compared to the control soils ($p = 0.05$). Copper levels in the soil increased from 2.46 mg/kg to 9.47 mg/kg, while lead levels increased from 0.0001 mg/kg to 0.0328 mg/kg, and iron levels increased from 24 mg/kg to 42.09 mg/kg. This study aligns with the findings of Agbogidi et al. (2010), who reported that a significant amount of Mn and Fe accumulated in all plants grown on contaminated soil, while other elements were present in trace amounts.

According to the heavy metal analysis results, the plants demonstrated that petroleum products had a significant impact on the heavy metal levels in the soil. The contaminated soil had significantly higher total Cu, Fe, Ni, Mn, and Zn values compared to the control soils ($p = 0.05$). Copper levels in soil increased from 2.46 mg/kg to 9.47 mg/kg, lead levels increased from 0.0001 mg/kg to 0.0328 mg/kg, and iron levels increased from 24 mg/kg to 42.09 mg/kg. This finding aligns with the study by Agbogidi et al. (2010), who reported that significant amounts of Mn and Fe accumulated in all plants grown on contaminated soil, while other assessed elements were present in trace amounts.

3.1.4 Phytochemical constituents in stem of the plant

a. *Alkaloids***.** The results presented in Table 4 show that significant reductions in the alkaloid content in the leaves of the plants compared to the high values in the control. The alkaloid content in *P. balfouriana* (50 mL, 100 mL, 150 mL of crude oil-polluted soils) was significantly reduced compared to the control but remained significantly higher than the alkaloid obtained in *P. balfouriana* (200 mL). *P. balfouriana* had the highest alkaloid content compared to *T. spathacea*. The reduction in alkaloid content appears to be treatment-dependent, as higher crude oil pollution levels corresponded to lower alkaloid content.

b. *Glycoside***.** The glycoside content in the control, *P. balfouriana* (50 mL, 100 mL), *T. spathacea* (50 mL, 100 mL) was significantly higher ($p > 0.05$) in the leaves compared to other plant species. This was followed by *P. balfouriana* at 200 mL crude oil soil pollution, and subsequently followed by the glycoside content in the leaves of *P. balfouriana* (150 mL). A significant reduction was observed in the glycoside content in the leaves of *T. spathacea* (200 mL). These results imply that the higher the pollution level, the lower the glycoside content in the plants. The findings reveal that the glycoside content in *P. balfouriana* and *T. spathacea* was generally high.

c. *Saponin***.** The result presented on Table 4, shows that the saponin content in the leaves of the plants significantly decreased with increasing concentrations of the crude oil in the polluted

soils compared with the control values. The saponin in *P. balfouriana* at 50 mL and 100 mL of the crude oil-polluted soils was significantly lower than the values obtained in the control.

d. *Flavonoid***.** The flavonoid content in the leaves of the plants significantly reduced with increasing concentrations of crude oil in the soils. The control plants had the highest flavonoid content, while significant reductions were observed in soils polluted with varying concentrations of crude oil. The flavonoid content in the leaves of *P. balfouriana* (50 mL, 100 mL) was significantly reduced, with no significant difference ($p > 0.05$) in the mean values compared to the control. This was followed by the flavonoid content in the leaves of *T. spathacea* (50 mL, 100 mL) in crude oil-polluted soils.

e. *Polyphenol***.** The results show that the polyphenol levels in *P. balfouriana* and *T. spathacea* (50 mL, 100 mL) were significantly reduced compared to the high values recorded in the control. This was followed by the polyphenol content in the leaves of *P. balfouriana* (150 mL), and subsequently *P. balfouriana* (200 mL). The lowest polyphenol values were observed in the leaves of *P. balfouriana* and *T. spathacea* at 200 mL. The results also revealed that the polyphenols levels in the plants decreased with increasing concentrations of crude oil in the soil. However, a comparison of polyphenol content between the plants showed that *P. balfouriana* had the highest polyphenol levels, followed by *T. spathacea*.

The phytochemicals decreased significantly ($p < 0.05$) after phytoremediation, resulting in a reduction in concentrations of the plants. The decrease observed in the soil samples indicated that the test plants were effective in taking up hydrocarbon into its systems. Additionally, there was a significant difference between the contaminated and uncontaminated soils. High levels of PAHs and TPHs has a negative impact on the availability of nutrients needed for plant growth and can lead to death of aquatic and terrestrial organisms. Our findings on phytochemical properties also align with the report of Nwaichi et al. (2021).

4. Conclusion

Plant selection is a prerequisite for successful phytoremediation of contaminated soil, and the two plants used in this study demonstrated high phytoremediation potential. The findings from our study revealed that *T. spathacea* and *P. balfouriana* possess phytoremediation potential.

Hence, the research confirmed that certain plants species, such as *T. spathecea* and *P. balfouriana*, have the ability to take up heavy metals like nickel, lead, and others at different concentrations of crude oil in the soil. The present results also show a corresponding decrease in phytochemical properties in all selected plant species as the concentration of hydrocarbon in the polluted soil increases. With the development of industrialization and urbanization, the abundance of crude oil pollution in the environment has increase enormously, raising significant concerns. To tackle this problem plant-based technology (phytoremediation) is used to clean contaminated soils. Phytoremediation has been proven to be a promising technique for re-vegetation crude oil-polluted soil, as some plants have shown remediation potential. This technique also enjoys good public acceptance and offers several advantages compared with other physicochemical methods.

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