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Evaluation of heavy metals status and microbiological properties of some oil exposed soils in Obama Flow Station in Bayelsa State, Nigeria

Namson Amadu Tsaku[1,2]✉
Funmilayo Alfa Yusufu[3]
Mary Ngumalen Bar-Anyam[4]
Eche Onah Otakpa[5]
Akwashiki Ombugadu[6]


(✉ Corresponding Author)

[1]*Department of Agronomy, Nasarawa State University, Shabu-Lafia Campus, Nigeria.*
[2]*Department of Soil Science, College of Agronomy, Joseph Sarwuan Tarka University, Makurdi, Benue State, Nigeria.*
[1,2]*Email: ntsaku225@gmail.com*
[3]*Department of Geography College of Education, Akwanga, Nasarawa State, Nigeria.*
[3]*Email: funmialfa@gmail.com*
[4]*Department of Agronomy, Faculty of Agriculture, Federal University of Lafia, Nasarawa State, Nigeria.*
[4]*Email: maryanyam94@gmail.com*
[5]*Department of Zoology, Faculty of Natural Sciences, University of Jos, Jos, Plateau State, Nigeria.*
[5]*Email: otakpaeche@gmail.com*
[6]*Department of Zoology, Faculty of Life Sciences, Federal University of Lafia, Lafia, Nasarawa State, Nigeria.*
[6]*Email: akwash24@gmail.com*

Abstract

Crude oil exploration has been associated with disruptions in soil chemical and microbial balance in oil-producing regions, raising environmental concerns. This study assessed heavy metal contamination and microbiological properties of soils from the Obama Flow Station, Bayelsa State, Nigeria, during the wet and dry seasons of 2022. Soil samples were collected from surface (0–15 cm) and subsurface (15–30 cm) depths across thirteen oil-exposed sites and one control site and analyzed for selected heavy metals and microbial populations. Chromium, cadmium, lead, and nickel were below detection limits (<0.001 mg kg^{-1}) in all samples. Iron was the most abundant metal, with higher concentrations recorded in subsurface soils (705.20 mg kg^{-1}) compared with surface soils (473.04 mg kg^{-1}), with seasonal variation between wet and dry periods. Zinc concentrations remained low, ranging from 0.19 to 0.22 mg kg^{-1} across depths and seasons. Total heterotrophic bacterial counts were generally higher in surface soils during the wet season (1.80×10^5–2.90×10^5 cfu g^{-1}), while fungal counts ranged from 1.40×10^4 to 2.86×10^4 cfu g^{-1}. Overall microbial populations were higher in surface soils during the wet season, reflecting seasonal influences. Comparable microbial values in control soils suggest limited alteration of microbial communities due to oil exposure. Overall, the soils showed no significant heavy metal contamination, though continued monitoring is recommended to prevent future accumulation.

Keywords: Bayelsa State, Heavy metals, Hydrocarbon-utilizing bacteria, Niger Delta, Oil-exposed soils, Seasonality, Soil microbiology.

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Contents

Contribution of this paper to the literature

This study provides the first integrated seasonal (wet/dry) and depth-resolved assessment of heavy metals and microbial communities, including hydrocarbon-utilizing and faecal coliform bacteria, in oil-exposed soils at Obama Flow Station, establishing baseline data and linking microbial functional groups with oil exposure in the Obama Flow Station, Bayelsa State.

1. Introduction

Since the discovery of crude oil in the mid-19th century, the sector has emerged as a significant source of revenue in Africa, particularly through trade and investment. Globally, Africa alone is home to five out of the thirty oil-producing countries [1, 2], with the notable oil-producing countries in Africa being Nigeria, Angola, Algeria, Libya, and Egypt. In the global crude oil supply, these five top producers of crude oil in Africa produced approximately 8 million barrels of crude oil per day in 2019, which was estimated at 10% of the global oil output [3].

Despite the gains associated with crude oil exploration, severe environmental hazards have been associated with the process, particularly in Africa for instance, most of the challenges associated with crude oil exploration results from waste (atmospheric emission, oil spills, drilling fluids, oily drill cuttings, gas-flares, well treatment fluids, and deck drainage, etc.) discharge on land and water bodies [4, 5]. Oil spillage during petroleum exploration and refining poses serious harm to the soil and aquatic communities, often resulting in a disruption in microbial processes and functions. The contaminants released into the environment could indirectly be absorbed by plants, thereby influencing the food chain/web negatively, with a resultant cascading impact on the entire ecosystem [6].

Nigeria has a long history of crude oil exploration and exploitation. However, with the growing human population, there has been increased pressure on oil products globally, causing an increase in oil exploration activities in the region to meet the growing demands, and consequently an increase in oil pollution and contamination of the environment resulting from the oil exploration activities. Thus impacting the lives of the terrestrial and aquatic communities [7, 8]. This has raised a very serious concern, particularly due to the health and ecological threats they pose. Crude oil spillage on soil disrupts the physical and chemical properties of soils, causing a buildup of petroleum hydrocarbons in the soil, raising the acidity of the soil, and increasing the concentration of heavy metals such as zinc (Zn), lead (Pb), cadmium (Cd), and chromium [9-11].

Heavy metals are a major threat to the ecosystem and general public health. They are persistent in the environment and non-biodegradable and have the tendency to bioaccumulate in nature, causing them to be very toxic [12]. They can affect the respiratory and nervous systems, and also cause heart, liver, and kidney dysfunction as well as some forms of cancer. For instance, exposure to high doses of chromium (Cr), arsenic (As), nickel (Ni), and cadmium (Cd) has been associated with cancer [13-15]. Exposure to some of these heavy metals may result when they dissolve in water bodies, from where they are transported by the underlying hydraulic conducting system, where they seep into other water sources [16, 17], from where they are taken up by both humans and animals.

The wastes derived from crude oil exploration, as well as spilled oil, alter the structure of the soil and vegetation in an area through the introduction of total petroleum hydrocarbons (TPH) and toxic metals. This causes a rise in the hydrocarbon-utilizing fungi (HUF) and the hydrocarbon-utilizing bacteria (HUB) of the place, thereby affecting the nutritional value of the plants growing in the area [18-20]. Among the bacteria associated with petroleum pollution, the commonest ones include: *Staphylococcus*, *Pseudomonas aeruginosa*, *Mycobacterium*, *Acinetobacter*, *Streptococcus*, and *Rhodococcus*. Among the notable fungal species with a remarkable ability to feed on petroleum products are *Penicillium notatum* and *Aspergillus niger* [21, 22].

Although several studies have documented the crude oil contamination level in oil exploration areas, particularly in the Niger Delta area of Nigeria, little is known about the heavy metal contamination of these soils resulting from crude oil exploration activities, and how these affect the soil microbial component of the area. In the Nigerian State of Bayelsa, located in the Niger Delta Region of the country, the Nigerian Agip Oil Company (NAOC) has been involved in oil mining with several flow stations located in the hinterland with its attendant consequences on plant, animal, and human health as well as the environment. This study, therefore, seeks to assess the heavy metal status of these oil-exposed soils and as well, to assess the microbiological properties of the soils at the Obama flow station in Bayelsa State, Nigeria.

2. Materials and Methods

2.1. Study Area

Laboratory and field studies were carried out at Obama flow stations, located in Bayelsa State, Nigeria. The facilities are owned by Nigeria, Agip Oil Company (NAOC), as shown in Figure 1.

2.2. Soil Sample Collection

Collection of soil samples was done using a stainless-steel auger at two depths, i.e., surface (0-15 cm) and sub-surface (15-30 cm) at the experimental locations (Figure 1) and stored in labeled polythene bags [23]. This study was conducted in the wet and dry seasons of 2022. Fourteen (14) sampling points were used for the study. Thirteen (13) were oil-exposed soils within the Obama flow station, while one (1), which served as the control (non-oil exposed soil), was a point away from the Obama flow station. From each sampling point, two soil samples were taken. One at the surface (0 - 15 cm) and the other at the sub-surface (15 - 30 cm), bringing the total number of collected samples for the two seasons to fifty-six (56). These depths were selected because hydrocarbon contaminants are usually concentrated within the top 30 cm of soil due to limited vertical infiltration, as reported in previous studies [24].

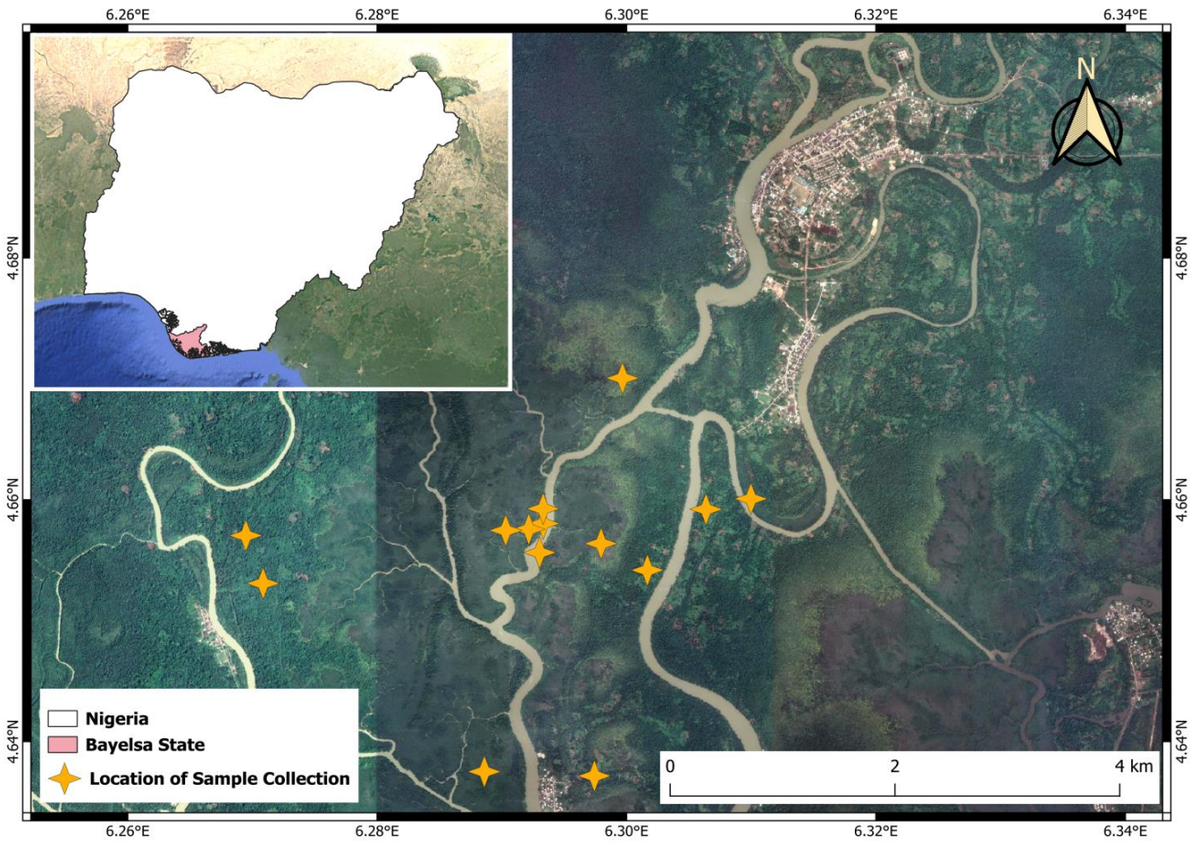

Figure 1. Soil collection sites in Obama oil flow stations in Bayelsa State, Nigeria (Generated using QGIS version 3.44.0-Solothurn).

2.3. Sample Preparation and Digestion (Aqua Regia Method)

Before evaluating the soil samples for the presence of heavy metals, a portion of the sample was air-dried in the lab, then sieved using a 2 mm mesh. 1 g of the dried sample was weighed using an electronic balance and digested using 10 mL of Aqua regia ($HCL:HNO_3$ = 3:1). The mixture was heated to 96 °C for 2 hours in a fume hood. On cooling, the mixture was filtered, and the volume of the filtrate was made up to 50.0 mL using deionized water [25].

2.4. Detection of Heavy Metals Presence

The detection of heavy metals in the collected samples was done using an Agilent Technologies 240 AA Atomic Absorption Spectrophotometer (AAS) equipped with a deuterium background corrector and an air-acetylene flame as described by Anyanwu [12]. For each metal, hollow cathode lamps were used. The analysis was done at specific wavelengths, lamp current, and slit width with an air-acetylene flame as the energy source [26].

Iron was analyzed at 248 nm, 0.2 nm slit, and 5mA. While chromium was detected at 358 nm with a 0.9 nm slit and 7 mA. Lead was analyzed at 283.5 nm with a 0.7 nm slit and 10 mA lamp current. The detection of zinc was at 214 nm, 1.0 nm slit, and 5 mA. Copper was determined at 324.9 nm, 0.5 nm slit and 3 mA, while Cadmium was measured at 229.1 nm, 0.7 nm slit and 8 mA current. Calibration curves were prepared using multi-element standard solutions at 0.1, 0.5, 1.0, 2.0, and 5.0 mg/L [27]. Blanks and certified reference materials were analyzed to ensure quality assurance [28].

2.5. Laboratory Preparation for Microbiology Load Assessment

All microbiological assays were done in aseptic conditions. Sterilization of glass wares including beakers, pipettes as well as Petri dishes, etc. wrapped in aluminum foil was done for 1 hour in a Carbolite PF 60 oven at 200°C for 1 hour in Carbolite PF 60 oven. Except where stated otherwise, all media, quarter strength Ringer's solution (diluent), Whatman No. 1 filter paper (carefully wrapped in aluminum foil), and membrane filtration apparatus were sterilized in the Schoeller- Bleckmann-Steels (SBS 20) sterilizer and Tuttnauer 13200EN autoclave- steam sterilizer at 121°C and 15 psi pressure for 15 minutes.

The room temperature was taken with a clinical thermometer clamped on a retort stand and maintained at 25°C throughout the duration of the study. Work bench top, inoculating hood, and anaerobic chamber were sanitized with 95% ethanol. The media used for the study (in both Petri dishes and test tubes) were decontaminated in an autoclave before being discharged into the environment.

2.6. Determination of Total Heterotrophic Bacteria and Fungi

The determination of the total heterotrophic bacteria (THB) and total heterotrophic fungi (THF) in the soil samples was done by the heterotrophic plate count method in accordance with ASTM D 5465-93 and APHA 9215. For each soil sample collected, 1g was weighed into 9ml sterile Ringer's solution. This was further diluted following the protocol, and 0.1ml aliquot of 103 and 1ml of 101 dilutions were then spread plated in duplicates on marine agar (MA) and potato dextrose agar (PDA) plates, respectively. Chloramphenicol (500mg/litre) was added to the PDA medium to prevent bacterial growth.

The plates were then inverted and incubated at 37°C for 24-48 hours for THB samples and at room temperature for 3-7 day for the Samples for THF. After incubation, plates that had about 30-300 colonies were counted and reported as colony-forming units per ml (cfu/ml) of the sample analyzed. Pure cultures of THB and

THF were made by sub-culturing the bacterial isolates on fresh MA and PDA, and thereafter transferring to nutrient agar (NA) and PDA slants. The samples were stored at 4°C before identification and characterization.

2.7. Determination of Hydrocarbon Utilizing Bacteria and Fungi

The detection of hydrocarbon-utilizing bacteria (HUB) and hydrocarbon-utilizing fungi (HUF) in soil samples was done by the heterotrophic plate count method in accordance with ASTM D 5465-93 and APHA 9215 using Minimal Salts Medium of Mills, et al. [29]. For each soil sample collected, 1g was weighed into 9ml sterile Ringer's solution. This was further diluted following the protocol, and 1ml aliquot of 101 dilutions was then spread plated in duplicates on HUB and HUF minimal salts medium agar plates, respectively, and chloramphenicol (500mg/litre) was added to prevent fungal and bacterial growth on HUB and HUF plates, respectively.

The plates were inverted and incubated for 7-10 days at room temperature. Thereafter, plates with 30-300 colonies or fewer were counted and reported as colony-forming units per ml (cfu/ml) of the sample analyzed. To obtain a pure culture, the isolates were subcultured on nutrient agar plates and then on nutrient agar slants. The samples were then stored at 4°C before identification and characterization.

Total Hydrocarbon (THC) Infra-red spectrophotometer, as described in American Society for Testing Materials method [30], was used to determine the THC in soil/water samples.

3. Results

3.1. Selected Heavy Metals Status of the Experimental Soils for the Wet Season of 2022

Table 1 contains the results of the heavy metals detected in the soil samples during the wet season of 2022. Cadmium (Cd), Chromium (Cr), Nickel (Ni) and Lead (Pb) were below detection limits (<0.001 mg kg^{-1}) in both surface (0–15 cm) and subsurface (15–30 cm) soils. Iron (Fe) recorded mean values of 434.97 ± 433.93 mg kg^{-1} in the surface soil and 705.20 ± 646.55 mg kg^{-1} in the subsurface soil. Zinc (Zn) recorded mean values of 0.22 ± 0.11 mg kg^{-1} and 0.20 ± 0.08 mg kg^{-1} in the surface and subsurface soils, respectively.

3.2. Selected Heavy Metals Status of the Experimental Soils for the Dry Season of 2022

During the dry season of 2022 (Table 2), the results also indicated that Cadmium (Cd), Chromium (Cr), Nickel (Ni), and Lead (Pb) were below the detection limits (<0.001 mg kg^{-1}) in both soil depths. Iron (Fe) recorded mean values of 473.04 ± 453.78 mg kg^{-1} in the surface soil and 466.62 ± 458.03 mg kg^{-1} in the subsurface soil. Zinc (Zn) recorded mean values of 0.21 ± 0.11 mg kg^{-1} in the surface soil and 0.19 ± 0.08 mg kg^{-1} in the subsurface soil.

Table 1. Heavy metal properties of surface and sub-surface soils at Obama during the wet season.

Point/ Depth (cm)	Cd		Heavy Metals (Mg/Kg soil)						Pb		Zn	
			Cr		Fe		Ni					
	0-15	15-30	0-15	15-30	0-15	15-30	0-15	15-30	0-15	15-30	0-15	15-30
SS_1	<0.001	<0.001	<0.001	<0.001	214.527	391.616	0.036	0.012	<0.001	<0.001	0.1132	0.0513
SS_2	<0.001	<0.001	<0.001	<0.001	447.208	817.227	0.018	0.004	<0.001	<0.001	0.0886	0.1325
SS_3	<0.001	<0.001	<0.001	<0.001	411.835	836.522	0.006	<0.001	<0.001	<0.001	0.3285	0.2112
SS_4	<0.001	<0.001	<0.001	<0.001	104.380	163.537	0.008	<0.001	<0.001	<0.001	0.2352	0.0956
SS_5	<0.001	<0.001	<0.001	<0.001	188.731	370.416	<0.001	<0.001	<0.001	<0.001	0.5349	0.3628
SS_6	<0.001	<0.001	<0.001	<0.001	427.146	609.288	<0.001	<0.001	<0.001	<0.001	0.2551	0.1668
SS_7	<0.001	<0.001	<0.001	<0.001	239.633	310.968	0.018	0.011	<0.001	<0.001	0.1821	0.1376
SS_8	<0.001	<0.001	<0.001	<0.001	336.102	416.139	0.014	0.002	<0.001	<0.001	0.2118	0.1831
SS_9	<0.001	<0.001	<0.001	<0.001	205.324	327.190	<0.001	0.064	<0.001	<0.001	0.2698	0.2697
SS_{10}	<0.001	<0.001	<0.001	<0.001	288.147	326.911	<0.001	<0.001	<0.001	<0.001	0.2093	0.2119
SS_{11}	<0.001	<0.001	<0.001	<0.001	412.883	738.421	<0.001	<0.001	<0.001	<0.001	0.1974	0.2248
SS_{12}	<0.001	<0.001	<0.001	<0.001	1713.214	2104.818	0.013	0.060	<0.001	<0.001	0.1581	0.2113
$SS\ CTL_1$	<0.001	<0.001	<0.001	<0.001	1022.151	2193.262	0.035	0.084	<0.001	<0.001	0.1234	0.1815
$SS\ CTL_2$	<0.001	<0.001	<0.001	<0.001	78.291	266.719	0.028	0.051	<0.001	<0.001	0.1415	0.2066
Mean	<0.001	<0.001	<0.001	<0.001	434.97	705.20	-	-	<0.001	<0.001	0.22	0.20
Std. Values					433.93	646.55					0.11	0.08

Table 2. Heavy metals properties of surface and sub-surface soils at Obama during the dry season.

	Heavy Metals (Mg/Kg Soil)											
Point	Cd		Cr		Fe		Ni		Pb		Zn	
Depth (cm)	0-15	15-30	0-15	15-30	0-15	15-30	0-15	15-30	0-15	15-30	0-15	15-30
SS_1	<0.001	<0.001	<0.001	<0.001	210.516	211.420	0.030	0.032	<0.001	<0.001	0.1112	0.1130
SS_2	<0.001	<0.001	<0.001	<0.001	247.208	246.108	0.010	0.013	<0.001	<0.001	0.0867	0.0856
SS_3	<0.001	<0.001	<0.001	<0.001	407.830	403.420	0.008	0.006	<0.001	<0.001	0.3240	0.3260
SS_4	<0.001	<0.001	<0.001	<0.001	102.260	101.160	<0.001	<0.001	<0.001	<0.001	0.2340	0.162
SS_5	<0.001	<0.001	<0.001	<0.001	168.720	178.730	<0.001	<0.002	<0.001	<0.001	0.5140	0.3630
SS_6	<0.001	<0.001	<0.001	<0.001	320.140	330.140	0.013	0.0016	<0.001	<0.001	0.2461	0.2370
SS_7	<0.001	<0.001	<0.001	<0.001	226.160	236.160	0.0015	0.009	<0.001	<0.001	0.1720	0.1636
SS_8	<0.001	<0.001	<0.001	<0.001	332.003	234.006	<0.001	<0.001	<0.001	<0.001	0.2108	0.2010
SS_9	<0.001	<0.001	<0.001	<0.001	206.123	208.146	<0.001	<0.001	<0.001	<0.001	0.2367	0.2460
SS_{10}	<0.001	<0.001	<0.001	<0.001	267.06	236.07	0.006	0.008	<0.001	<0.001	0.2060	0.2016
SS_{11}	<0.001	<0.001	<0.001	<0.001	410.863	411.862	<0.001	<0.001	<0.001	<0.001	0.1968	0.1920
SS_{12}	<0.001	<0.001	<0.001	<0.001	1706.116	1712.120	0.010	0.013	<0.001	<0.001	0.1564	0.1484
$SS\ CTL_1$	<0.001	<0.001	<0.001	<0.001	1012.162	1010.134	0.032	0.035	<0.001	<0.001	0.1220	0.1210
$SS\ CTL_2$	<0.001	<0.001	<0.001	<0.001	1011.361	1012.206	0.023	0.020	<0.001	<0.001	0.1410	0.1408
Mean	-	-	-	-	473.04	466.62	-	-	-	-	0.21	0.19
Std. Values					453.78	458.03					0.11	0.08

3.3. Selected Microbiological Properties of the Obama Flow Stations During the Wet Season of 2022

The selected microbiological properties of the soils during the wet season are presented in Table 3. The mean Heterotrophic Bacteria Count (HBC) was slightly higher in the surface (2.42×10^5 cfu g^{-1}) compared to the subsurface soil sample (2.02×10^5 cfu g^{-1}). A similar result was also observed for the Heterotrophic Fungal Count (HFC) with a higher mean value in the surface (2.28×10^4 cfu g^{-1}) than the subsurface (1.90×10^4 cfu g^{-1}). Hydrocarbon-utilizing bacteria (HUB) were also much higher in the surface soils (7.74×10^3 cfu g^{-1}) compared to the subsurface soils (5.86×10^3 cfu g^{-1}). Similar results were recorded for Hydrocarbon Utilizing Fungi (HUF), with a higher value in the surface (2.07×10^2 cfu g^{-1}) compared to the subsurface sample (1.73×10^2 cfu g^{-1}). Faecal Coliform Count (FCC) ranged from 1.9x 103 to 8.1x 103 MPN/100 g in the surface soil and 1.2x 103 to 6.0x 103 MPN/100 g in the subsurface soil, with mean values of 3.64×10^3 MPN/100 g and 2.60×10^3 MPN/100 g, respectively.

3.4. Selected Microbiological Properties of the Obama Flow Stations During the Dry Season of 2022

The selected microbiological properties of the soils during the wet season are presented in Table 4. Similar to the wet season, all values in the surface soils were higher compared to the subsurface. The mean Heterotrophic Bacteria Count (HBC) was 2.14×10^5 cfu g^{-1} while at the sub-surface, it was 1.83×10^5 cfu g^{-1}. Heterotrophic Fungal Count (HFC) at the surface was 2.08×10^4 cfu g^{-1}, and 1.76×10^4 cfu g^{-1} at the subsurface. Hydrocarbon-utilizing bacteria (HUB) were also much higher in the surface soils (7.36×10^3 cfu g^{-1}) compared to the subsurface soils (5.74×10^3 cfu g^{-1}). Hydrocarbon-utilizing fungi (HUF) were slightly at the surface ($1.97 \times 10^2 \times 10^2$ cfu g^{-1}) compared to the subsurface (1.68×10^2 cfu g^{-1}). Faecal Coliform Count (FCC) ranged from 1.7x 103 to 7.5x 103 MPN/100 g in the surface soil and 1.4x 103 to 5.3x 103 MPN/100 g in the subsurface soil, with mean values of 3.47×10^3 MPN/100 g and 2.38×10^3 MPN/100 g, respectively.

Table 3. Microbiological properties of surface and sub-surface soils at Obama during the wet season of 2022.

Microbiological Property										
Point	**HBC (Cfu/g)**		**HFC (Cfu/g)**		**HUB (Cfu/g)**		**HUF (Cfu/g)**		**FCC (MPN/100g)**	
Depth (cm)	**0-15**	**15-30**	**0-15**	**15-30**	**0-15**	**15-30**	**0-15**	**15-30**	**0-15**	**15-30**
SS $_1$	2.70 x 10^5	2.40 x 10^5	2.86 x 10^4	2.58 x 10^4	9.20 x 10^3	7.60 x 10^3	2.20 x 10^2	2.00 x 10^2	4.3 x10^3	3.7x 10^3
SS $_2$	2.10 x 10^5	1.80 x 10^5	2.30 x 10^4	1.70 x 10^4	6.80 x 10^3	4.60 x 10^3	1.90 x 10^2	1.60 x 10^2	2.1x 10^3	1.6x 10^3
SS $_3$	2.60 x 10^5	2.20 x 10^5	2.40 x 10^4	1.90 x 10^4	8.20 x 10^3	6.40 x 10^3	2.10 x 10^2	1.50 x 10^2	3.6x 10^3	2.4x 10^3
SS $_4$	2.36 x 10^5	1.90 x 10^5	2.10 x 10^4	1.60 x 10^4	5.20 x 10^3	4.40 x 10^3	1.70 x 10^2	1.40 x 10^2	3.3x 10^3	2.7x 10^3
SS $_5$	2.10 x 10^5	1.60 x 10^5	2.00 x 10^4	1.80 x 10^4	7.20 x 10^3	5.80 x 10^3	2.10 x 10^2	1.66 x 10^2	3.1x 10^3	1.8x 10^3
SS $_6$	2.58 x 10^5	2.20 x 10^5	2.38 x 10^4	1.96 x 10^4	8.50 x 10^3	6.20 x 10^3	2.28 x 10^2	1.84 x 10^2	2.5x 10^3	1.6x 10^3
SS $_7$	2.80 x 10^5	2.16 x 10^5	2.60 x 10^4	2.25 x 10^4	9.60 x 10^3	6.40 x 10^3	2.50 x 10^2	2.20 x 10^2	7.2x 10^3	5.4x 10^3
SS $_8$	2.36 x 10^5	1.78 x 10^5	2.10 x 10^4	1.64 x 10^4	7.80 x 10^3	4.40 x 10^3	1.96 x 10^2	1.70 x 10^2	2.2x 10^3	1.4x 10^3
SS $_9$	1.80 x 10^5	1.50 x 10^5	1.56 x 10^4	1.30 x 10^4	5.60 x 10^3	3.80 x 10^3	1.40 x 10^2	1.18 x 10^2	1.9x 10^3	1.2x 10^3
SS $_{10}$	2.90 x 10^5	2.76 x 10^5	2.80 x 10^4	2.52 x 10^4	9.80 x 10^3	8.20 x 10^3	2.60 x 10^2	2.30 x 10^2	8.1x 10^3	6.0x 10^3
SS $_{11}$	2.52 x 10^5	2.28 x 10^5	2.30 x 10^4	2.00 x 10^4	7.80 x 10^3	6.60 x 10^3	2.28 x 10^2	1.86 x 10^2	3.6x 10^3	2.4x 10^3
SS $_{12}$	2.40 x 10^5	1.86 x 10^5	2.18 x 10^4	1.70 x 10^4	7.40 x 10^3	5.80 x 10^3	2.00 x 10^2	1.64 x 10^2	3.0x 10^3	2.1x 10^3
SS CTL$_1$	2.60 x 10^5	2.30 x 10^5	2.46 x 10^4	2.20 x 10^4	8.40 x 10^3	7.20 x 10^3	2.30 x 10^2	2.10 x 10^2	3.4x 10^3	2.2x 10^3
SS CTL$_2$	2.10 x 10^5	1.60 x 10^5	1.90 x 10^4	1.40 x 10^4	6.80 x 10^3	4.60 x 10^3	1.60 x 10^2	1.30 x 10^2	2.7x 10^3	1.9x 10^3
Mean	2.42 × 10^5	2.02 × 10^5	2.28 × 10^4	1.90 × 10^4	7.74 × 10^3	5.86 × 10^3	2.07 × 10^2	1.73 × 10^2	3.64×10^3	2.60 × 10^3
Std. Values	3.10 × 10^4	3.61 × 10^4	3.49 × 10^3	3.85 × 10^3	1.37 × 10^3	1.34 × 10^3	3.37 × 10^1	3.36 × 10^1	1.83×10^3	1.46 × 10^3

Table 4. Microbiological properties in surface and sub-surface (15-30) soils at Obama during the dry season 2022.

Microbiological Property										
Point	**HBC (Cfu/g)**		**HFC (Cfu/g)**		**HUB (Cfu/g)**		**HUF (Cfu/g)**		**FCC (MPN/100g)**	
Depth (cm)	**0-15**	**15-30**	**0-15**	**15-30**	**0-15**	**15-30**	**0-15**	**15-30**	**0-15**	**15-30**
SS $_1$	2.58 x 10^5	2.30 x 10^5	2.60 x 10^4	2.30 x 10^4	8.80 x 10^3	6.40 x 10^3	2.44 x 10^2	2.10 x 10^2	4.2x 10^3	3.5x 10^3
SS $_2$	1.90 x 10^5	1.76 x 10^5	2.00 x 10^4	1.50 x 10^4	5.60 x 10^3	4.00 x 10^3	1.80 x 10^2	1.46 x 10^2	1.7x 10^3	1.5x 10^3
SS $_3$	2.40 x 10^5	2.00 x 10^5	2.30 x 10^4	1.70 x 10^4	8.40 x 10^3	7.60 x 10^3	2.00 x 10^2	1.60 x 10^2	3.4x 10^3	2.9x 10^3
SS $_4$	1.80 x 10^5	1.66 x 10^5	1.98 x 10^4	1.66 x 10^4	6.00 x 10^3	5.20 x 10^3	1.70 x 10^2	1.50 x 10^2	2.2x 10^3	1.5x 10^3
SS $_5$	1.90 x 10^5	1.50 x 10^5	1.80 x 10^4	1.58 x 10^4	7.00 x 10^3	4.60 x 10^3	2.08 x 10^2	1.84 x 10^2	2.5x 10^3	1.4x 10^3
SS $_6$	2.40 x 10^5	2.00 x 10^5	2.00 x 10^4	1.70 x 10^4	8.60 x 10^3	6.40 x 10^3	2.10 x 10^2	1.98 x 10^2	4.0x 10^3	2.7x 10^3
SS $_7$	2.60 x 10^5	2.30 x 10^5	2.40 x 10^4	2.18 x 10^4	9.00 x 10^3	8.20 x 10^3	2.40 x 10^2	2.00 x 10^2	6.4x 10^3	4.0x 10^3
SS $_8$	2.10 x 10^5	1.60 x 10^5	1.92 x 10^4	1.54 x 10^4	6.80 x 10^3	4.00 x 10^3	1.80 x 10^2	1.60 x 10^2	2.7x 10^3	1.5x 10^3
SS $_9$	1.74 x 10^5	1.36 x 10^5	1.86 x 10^4	1.50 x 10^4	5.80 x 10^3	3.40 x 10^3	1.76 x 10^2	1.40 x 10^2	2.2x 10^3	1.4x 10^3
SS $_{10}$	2.70 x 10^5	2.50 x 10^5	2.60 x 10^4	2.32 x 10^4	9.20 x 10^3	8.60 x 10^3	2.40 x 10^2	2.18 x 10^2	7.5x 10^3	5.3x 10^3
SS $_{11}$	2.20 x 10^5	1.96 x 10^5	2.46 x 10^4	2.20 x 10^4	8.00 x 10^3	7.40 x 10^3	2.00 x 10^2	1.70 x 10^2	3.6x 10^3	2.4x 10^3
SS $_{12}$	2.00 x 10^5	1.70 x 10^5	2.00 x 10^4	1.60 x 10^4	6.80 x 10^3	5.60 x 10^3	1.82 x 10^2	1.50 x 10^2	2.9x 10^3	2.3x 10^3
SS CTL$_1$	1.80 x 10^5	1.50 x 10^5	1.80 x 10^4	1.66 x 10^4	6.00 x 10^3	5.20 x 10^3	1.94 x 10^2	1.62 x 10^2	2.6x 10^3	1.5x 10^3
SS CTL $_2$	1.90 x 10^5	1.50 x 10^5	1.40 x 10^4	1.26 x 10^4	7.00 x 10^3	3.80 x 10^3	1.30 x 10^2	1.10 x 10^2	2.7x 10^3	1.4x 10^3
Mean	2.14 × 10^5	1.83 × 10^5	2.08 × 10^4	1.76 × 10^4	7.36 × 10^3	5.74 × 10^3	1.97 × 10^2	1.68 × 10^2	3.47× 10^3	2.38× 10^3
Std. Values	3.33 × 10^4	3.53 × 10^4	3.46 × 10^3	3.39 × 10^3	1.28 × 10^3	1.72 × 10^3	3.12 × 10^1	3.02 × 10^1	1.65× 10^3	1.20× 10^3

4. Discussion

The contamination of soils by crude oil and other associated heavy metals is a well-documented environmental issue in the Niger Delta region of Nigeria, arising from decades of exploration, spills, and poor waste management [7, 11]. Heavy metals found in the environment can persist in the environment and as well bio-accumulate in several tissues, and because of their toxic nature, they can pose long-term risks to ecosystem structure and function [9]. This study assessed the heavy metal load and microbiological properties of soils exposed to oil at the Obama flow station in Bayelsa State during the 2022 wet and dry seasons.

In the current study, only Iron (Fe) and Zinc (Zn) occurred in detectable quantities. Other metals such as Chromium (Cr), Cadmium (Cd), Nickel (Ni) and Lead (Pb) were consistently below detection limits (<0.001 mg kg^{-1}) in both surface (0–15 cm) and subsurface (15–30 cm) soils. The occurrence of iron was slightly higher in concentration in the surface compared to the subsurface soils, both in the wet and dry seasons. Other studies have equally reported the dominance of iron compared to other metals. This observation could be due to both natural geochemical processes and inputs from petroleum activities in the area [11, 31]. Similar findings were also reported by Douglas, et al. [32] and Ayobami [33], who also found a higher concentration of iron compared to other heavy metals studied. Contrary to what was found in this study, Onwugbuta, et al. [34] found moderate contamination from other metals, including Cd, Cr, Zn, Mn, Ni, and Pb. The absence of detectable Cr, Ni, Cd, and Pb in this study could be due to these metals either being naturally low in the area's parent materials or to historical contamination not having led to their accumulation at levels above the detection threshold of the analytical method used. Similarly, Afolabi and Adesope [35] also found low concentrations of Cd and Pb in their study, despite evidence of oil contamination, which suggests that heavy metal profiles can vary significantly across sites depending on spill history, soil properties, and hydrology. Of note is the fact that the concentration of iron detected in the current study exceeds typical regulatory baseline levels for agricultural soils (often ~100 mg kg^{-1}) although values up to 1,000 mg kg^{-1} have been reported previously [36]. Elevated iron alone, while not directly the most toxic heavy metal, can indicate overall disturbance of soil geochemistry and warrants periodic monitoring, especially where soil is used for crop production or livestock grazing.

Apart from metals, the microbial profile of a soil is a sensitive indicator of soil health and organic contamination [37]. Heterotrophic bacteria (HBC), heterotrophic fungi (HFC), hydrocarbon-utilizing bacteria (HUB), and hydrocarbon-utilizing fungi (HUF) were consistently present in both seasons, with higher microbial counts generally observed in surface soils compared to subsurface soils in both seasons studied. This is likely because surface soils contain greater organic matter, oxygen, and moisture contents compared to the sub surface, and these factors are needed for microbial activities in the soil [38]. The seasonal variation observed also in the soil microbial content is consistent with other studies where moisture availability in wet seasons enhances microbial metabolic activity and growth [39]. In contrast, reduced moisture and temperature variation in the dry season can limit nutrient diffusion and microbial growth, resulting in lower counts [40].

In this study, hydrocarbon-utilizing bacteria (HUB) and hydrocarbon-utilizing fungi (HUF) were detected across both depths and seasons, which is suggestive of the ecological adaptation and enrichment of microbial communities capable of degrading petroleum hydrocarbons. Soils contaminated by hydrocarbons exert selective pressure that favors indigenous microorganisms able to metabolize petroleum compounds as carbon and energy sources [38]. Thus, such environments often harbor diverse populations of bacteria and fungi involved in natural attenuation and bioremediation processes. It has been reported previously in the Niger Delta areas where petroleum contamination is common that indigenous microorganisms such as Bacillus, Pseudomonas, Micrococcus, and Aspergillus species utilize the crude oil and related hydrocarbons effectively [41, 42]. The hydrocarbon is utilized by these organisms as a growth substrate, resulting in elevated counts of hydrocarbon-degrading microbes compared with uncontaminated environments. Furthermore, studies in the same region also showed that indigenous bacterial communities possess the metabolic capacity to degrade various petroleum fractions, indicating their potential role in natural attenuation and bioremediation of hydrocarbon-impacted environments [43, 44].

In this study, however, hydrocarbon-degrading microorganisms were detected in both the control and exposed sites, which may suggest that there is widespread low-level hydrocarbon exposure, which is characteristic of many ecosystems in the Niger Delta area. Naturally, the hydrocarbon-utilizing microorganisms occur in soil and aquatic environments. Several microbial taxa are known to possess metabolic pathways capable of degrading petroleum-derived compounds [45]. However, decades of petroleum exploration, transport, and spill events have resulted in chronic environmental contamination in parts of the Niger Delta, which could have influenced the structure and functional capacity of indigenous microbial communities. As a result, these organisms may persist even in areas without any apparent record of pollution due to long-term ecological adaptation and background hydrocarbon inputs.

Unlike hydrocarbon-degrading microorganisms, faecal coliform counts (FCC) in this study ranged between 1.2 $\times 10^3$ and 8.1 $\times 10^3$ MPN / 100 g. This observation suggests that there is an introduction of faecal material into the soil environment. Faecal coliform bacteria are widely used as indicators of contamination from the intestinal wastes of humans and warm-blooded animals and commonly occur in environments influenced by livestock activities, sewage discharge, or surface runoff [46, 47]. Their occurrence, therefore, is suggestive of the influence of surrounding land-use practices and anthropogenic inputs rather than petroleum contamination.

5. Conclusion

The current study assessed the level of heavy metal contamination and microbiological properties of soil samples around the Obama Flow Station in Bayelsa State, Nigeria. The result indicated that iron was the most abundant heavy metal detected, occurring more in subsurface soils during the wet season, while zinc occurred at very low concentrations across both depths sampled in both seasons. The level of contamination with other heavy metals such as cadmium, chromium, nickel, and lead was, however, negligible. Microbial analysis showed relatively high populations of heterotrophic bacteria and fungi, particularly in surface soils during the wet season. However, the current study revealed that the general microbial structure of the soil remains largely unaffected. This study therefore indicates that the soils around the Obama Flow Station are not heavily impacted by petroleum-associated heavy metals. However, continued environmental monitoring is necessary to ensure early detection of any future accumulation of contaminants associated with oil exploration activities.

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