

## RESEARCH ARTICLE

# Quantification of Heavy Metal Bioaccumulation Quotients in Invertebrates from Petroleum Hydrocarbon Impacted Soil

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**Citation:** Ezenwelu Chijioke O, Ilechukwu Cyril C, Chigbo Malachy C, Okeke Chisom M, Oparaji EH (2024) Quantification of Heavy Metal Bioaccumulation Quotients in Invertebrates from Petroleum Hydrocarbon Impacted Soil, J of Ind Pollut Eff and Control 3: 101

## Abstract

The study investigated the level of heavy metals in soil earthworms isolated from a half decade drilling sites located in Rivers state. Earth worm were isolated using standard sampling techniques. Heavy metals of Cd, Cr, Cu, Fe, Ni, Pb and Zn were assayed for in the soil and the isolated soil earthworms using standard analytical spectrophotometric methods. Cd, Cr and Cu were found at the concentrations (mg/kg) of  $0.021 \pm 0.052^c$ ,  $0.137 \pm 0.21^c$ ,  $0.612 \pm 0.12^c$ , respectively in the soil sample; however, Fe, Ni, Pb and Zn were found at the concentrations (mg/kg) of  $18.09 \pm 0.012^c$ ,  $2.45 \pm 0.04^c$ ,  $1.02 \pm 0.12^c$ ,  $3.87 \pm 0.01^a$ , respectively. The estimated metals were significantly different from the control experiment. Soil earthworms analysed for the heavy metals showed the concentrations (mg/kg) of Cd, Cr and Cu at  $0.0085 \pm 0.15^c$ ,  $0.124 \pm 0.1^a$ ,  $0.214 \pm 0.04^c$ , respectively. Quantifications of Fe, Ni, Pb and Zn in the isolated earthworms showed their concentrations (mg/kg) at  $8.65 \pm 0.05^a$ ,  $0.081 \pm 0.01^c$ ,  $0.521 \pm 0.12^c$ ,  $0.911 \pm 0.03^c$ , respectively. Experimentally these heavy metals in the earthworms from the petroleum hydrocarbon impacted soil were significantly high when compared with those from the control experiment. The study has shown the loading index of heavy metals in soil sample with evident history of petroleum hydrocarbon pollution using bioindicator of earthworms as ecological evidence; this will otherwise reveal the toxicological potential of consumables cultivated in the soil sample to human.

**Keywords:** Heavy metals; earthworms, petroleum hydrocarbons

## Study Background

Heavy metals are metals with relatively high atomic mass and thus which reflect in their atomic weights examples includes Arsenic, beryllium, cadmium, chromium, lead, manganese, mercury, nickel, and selenium (Osei). They take part in bio-geo-chemical reactions and are transported between compartments by natural processes, the rate of which are at times greatly altered by human activities [1]. They persist in nature and can cause damage or death in animals, humans, and plants even at very low concentrations (1 or 2 micrograms in some cases) [1].

This is because the metals cannot be metabolized by natural means. Oil spill is a key source of heavy metals (Osuji, Onajeke). The peculiar ability of heavy metals is to accumulate without being noticed to levels of toxicity (Wegwu and Akaninwor). Heavy metals are connected with severe health abnormalities such as nephrotoxicity, neurotoxicity and malignancies of various types (Goyer). Lead (Pb) interferes with haem biosynthesis (USEPA). It inhibits the activity of 2-amino laevulinic acid dehydratase which leads to accumulation of protoporphyrin in the red blood cell (WHO). Cadmium can expediently substitute zinc in several enzymes consequently changing their configuration and inhibiting catalytic function (Wegwu).

Heightened navigational activities in inland and coastal lines and sublines within the hinter sub-regions of Rivers state surrounding the refinery is another environmental health concern and anthropogenic source of refined petroleum pollution to the environment.

Bioindication is a constitutive element of high imperativeness in the assessment of soil quality (USDA-NRCS). Markert *et al.* defined bioindication as qualitative indication of environmental properties, and biomonitoring as quantified bioindication in order to detect trends in time and space. Earthworms are often indicated as bioindicators of soil quality because they are an important part of the soil system and also because they are frequent, easy to collect, and rather simple to identify [2] (USDA-NRCS). The occurrence and the effects of earthworms are generally associated with good soil quality. With intensified farming activities around the southern/eastern coastline, agricultural soil fertility remain the force of breakthrough to every farmer at all cadre within this area and the nation at large. Ecology and toxicological impact assessment using certain biomarkers/indicators such as earthworms, ferns, epiphytes have shown the level of toxicological deterioration of the ecosystem.

Bioaccumulation factor of the earthworms collected from the abandoned drilling sites respectively, revealed the level of toxicants in the soil which will in turn affect the origination of biological activities such as mineralization, distribution of organisms etc within the soil.

The present study stands to show the level of heavy metals in earthworms isolated from a half decade drilling sites located in Rivers state and the physicochemical properties of the surrounding soil respectively.

## Materials and Methods

### Materials

All the reagents, equipment used in the present study were of analytical grade and products of BDh, May and Baker, Sigma Alrich. The equipments are calibrated at each use.

### Methods

#### Collection of the Experimental Samples

## Soil Samples

Petroleum hydrocarbon contaminated soil was collected Agbada oil field (LONG.7° 00",57°.3" E; LAT. 4° 53",70°.04" N) located at Portharcourt, Rivers state, Nigeria as described by Ezenwelu *et al.* Samples were collected at about 6 am stratifiedly from the stated site; 9cm impressions were made at the four perimeters of the marked sites at a distance of 1m from each other from where the soil were collected. The soil was collected in clean sterile sample containers and was taken to the lab for further experiments.

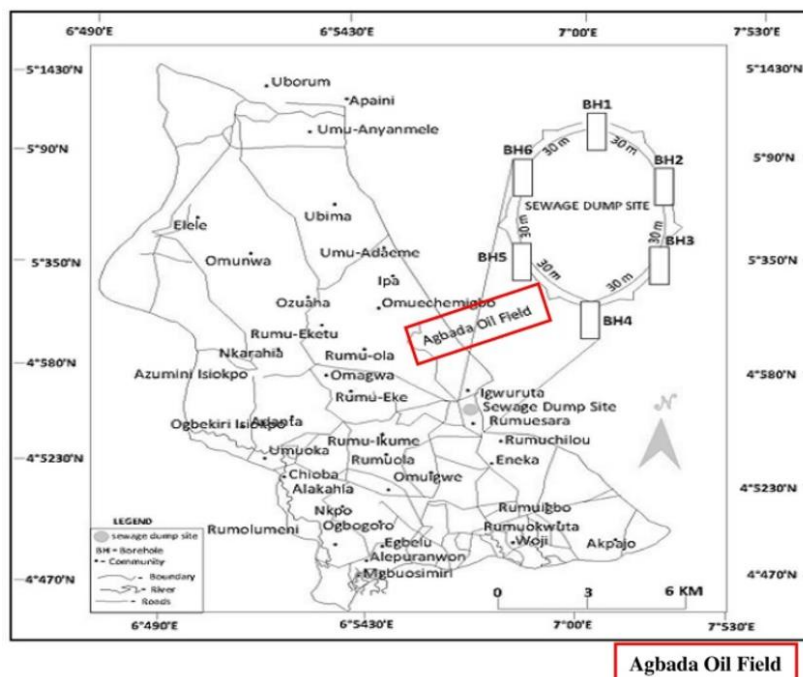


Figure 1: Site area of the collected soil and earthworm samples

## Collection of Earthworms

Earth worms from the Agbada location were collected as described by Vallero. The lower invertebrates were collected after minor excavation of the earth using hand spade. They were homogenously packed inside a transparent moistened container.

## Soil and Earthworms Samples Digestion for Heavy Metal Analysis

Five grams (5g) of the soil and earthworm samples were digested in 250 ml conical flask by adding 30 ml of aqua regia ( $\text{HNO}_3$ ,  $\text{HCl}$  and  $\text{HF}$  in the ratio 3:2:1) and heated on a hot plate until volume remains about 7-12 ml as described by Oyibo *et al.* The digest were filtered using what-man filter paper and the volume was made up to the mark in a 50ml volumetric flask, and will then be stored in a plastic container for atomic absorption spectra (AAS) analysis.

## Results and Discussion

Heavy metal concentration of the soil samples collected from the different sites are presented in table 1. Cd, Cr and Cu were found at the concentrations (mg/kg) of  $0.021 \pm 0.052^c$ ,  $0.137 \pm 0.21^c$ ,  $0.612 \pm 0.12^c$ , respectively in the soil sample; however, Fe, Ni, Pb and Zn were found at the concentrations (mg/kg) of  $18.09 \pm 0.012^c$ ,  $2.45 \pm 0.04^c$ ,  $1.02 \pm 0.12^c$ ,  $3.87 \pm 0.01^a$ , respectively. The result indicates that petroleum hydrocarbon polluted soil show relative high heavy metal contents when compare to the control experiment. Heavy metals such as mercury and arsenic were not detected in both experiments under the measuring concentration limit.

**Table 1:** Soil Heavy Metal Concentrations from the soil samples from Rivers State

Metals (mg/kg)	Control	Polluted soil sample
Cd	BDL	0.021±0.052 <sup>c</sup>
Cr	0.073±0.02 <sup>a</sup>	0.137±0.21 <sup>c</sup>
Cu	1.048±0.21 <sup>c</sup>	0.612±0.12 <sup>c</sup>
Fe	8.11±0.2 <sup>c</sup>	18.09±0.012 <sup>b</sup>
Ni	0.092±0.01 <sup>a</sup>	2.45±0.04 <sup>c</sup>
Pb	0.112±0.25 <sup>a</sup>	1.02±0.12 <sup>c</sup>
Zn	0.216±0.42 <sup>c</sup>	3.87±0.01 <sup>a</sup>

N=3 Mean values with the same superscript letters are not statistical significant at  $p \leq 0.05$ .

Mg/kg= milligram per kilogram. Cd= cadmium, Cr=chromium, Cu=copper, Fe=iron, Ni=nickel, Pb=lead, Zn=zinc.

Evidence of high Fe, Ni and Zn in the polluted soil sample from Rivers state can be attributed to the profile of the soil and the petroleum hydrocarbons. These metals are significantly evident in petroleum hydrocarbon compounds and in certain analysed ferruginous soil. As reported by the ATSDR [3] the most impacted soil contains majorly trace metals of Fe and Cu then lead can be significantly present depending on the activity historical of the plain soil. Alexander et al. in their documentary proceedings on the particulars of claim between the Bodo community, Gokana Local Government Area, Rivers State, Nigeria and The Shell Petroleum Development Company of Nigeria Ltd in the court stated among the ecological damages to their ecosystem a significant presence of Fe, Pb and Ni in the experimented soil.

From the table above ecology significant toxicants (trace metals) like Cd, Cr and Pb were present in the experimented soil; these trace metals as reported by Vallero (2010) are potent poisons to the ecosystem even at minute concentration of part per million (ppm). Their presence in the experimented soil from Rivers state reveal the level of aged influx of noxious petroleum hydrocarbons to the soil and its accruing ecological damage.

Heavy metal concentrations (mg/kg) of earthworms from the collection sites in Rivers state, Nigeria as presented in table 2 below. The result indicates that earthworms from the sites with oil spillage history show relative high heavy metal contents when compare to the control experiment metallic contents. Heavy metals of Fe, Ni, Pb and Zn in the isolated earthworms showed their concentrations (mg/kg) at  $8.65 \pm 0.05^a$ ,  $0.081 \pm 0.01^c$ ,  $0.521 \pm 0.12^c$ ,  $0.911 \pm 0.03^c$ , respectively.

**Table 2:** Soil Earthworms Heavy Metal Concentrations from the respective soil samples in Rivers State

Metals (mg/kg)	Control	Earthworms sample
Cd	BDL	0.0085±0.15 <sup>c</sup>
Cr	0.012±0.025 <sup>c</sup>	0.124±0.1 <sup>a</sup>
Cu	0.123±0.01 <sup>bc</sup>	0.214±0.04 <sup>c</sup>
Fe	1.23±0.01 <sup>b</sup>	8.65±0.05 <sup>a</sup>
Ni	0.094±0.06 <sup>c</sup>	0.081±0.01 <sup>c</sup>

Pb	0.010±0.04 <sup>c</sup>	0.521±0.12 <sup>c</sup>
Zn	0.231±0.012 <sup>b</sup>	0.911±0.03 <sup>c</sup>

N=3 Mean values with the same superscript letters are not statistical significant at  $p \leq 0.05$ .

Mg/kg= milligram per kilogram. Cd= cadmium, Cr=chromium, Cu=copper, Fe=iron, Ni=nickel, Pb=lead, Zn=zinc.

Ecology and toxicological impact assessment using certain biomarkers/indicators such as earthworms, ferns, epiphytes have shown the level of toxicological deterioration of the ecosystem. Bioaccumulation factor of the earthworms collected from the abandoned drilling sites respectively, revealed the level of toxicants in the soil which will in turn affect the origination of biological activities such as mineralization, distribution of organisms etc within the soil. From the table above, heavy metals of Fe, Pb, Zn were significantly high in the isolated earthworm sample from the hydrocarbon impacted soil when compared with those from the control experiment. It was significant from the persistent of Pb and Cr in the isolated earthworms; this correlates with the findings of Vallero; EPA on the bioaccumulation and metabolizing effects of trace metals by inhabitant organisms. They went further to state that generally trace metals are biological unmetabolized *in vivo* as its biochemical pathways are variant inducible in organisms and as well in humans. These to say posed them as potent ecological toxicant to the biological entities generally. The slow difference in the concentration of Pb/Cr in the experimented earthworms from the soil mark them ecological risk recalcitrant. As described by Benard *et al.* *in situ* ecological biomarkers/indicators are foremost ecological signaling entities towards a healthy or compromised environment; these from the result on earthworm samples from Rivers state showed the bioaccumulation quotients of trace metals in earthworm samples. Biochemodynamics of these recalcitrant partitioning within the soil through the organismal entities remain still unexplored. Oparaji [3] stated that their metabolism in the organisms to less toxic end point remain not well expressed.

## Conclusion

The present study has presented the risk index of crude oil impacted polluted soil using certain indices of earthworms. Physico-chemical analysis of the crude oil polluted soils in the present of the control experiment revealed the equivalence of certain hazardous entities in an off threshold numerical. Heavy metal indices of the polluted soils were relatively higher than that of the control experiment. Analysis of heavy metal loading index in the biomarkers (earthworms) showed the presence of health implicated heavy metals of Pb and Cd in a higher concentration. These experimental information will provide the health and risk assessment of inhabitants of concerned community moreso the present study has shown the level of pollution of the community agricultural soil.

## Funding Information

This work was solely funded by the research group of Dr. Ezenwelu Chijioko, O.

## Author's Contributions

**Ezenwelu, C.:** Conceived and designed the experiments, performed the experiment and processed the data, analyzed the data and wrote the manuscript.

**Ilechukwu, Cyril. C:** Co-supervised the research and revised the manuscript.

**Chigbo, Malachy C.:** Analyzed the research design and methodology, interpreted the data.

**Okeke, Chisom, M.:** Analyzed the research design and methodology, interpreted the data.

**Oparaji E. Henry:** Guided the experimental design, supervised the research, performed the experiment interpreted the data, revised the manuscript and processed the data.

## Ethics

Authors declared no ethical issues that may arise after the publication of this manuscript.

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