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Original Research Article

Histopathological Effects of Spent Oil Based Drilling Mud and Cuttings on the Earthworm, *Aporrectodea longa*

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Abstract

Purpose: To evaluate the histopathological effects of spent oil based drilling mud and cuttings on *Aporrectodea longa*.
Methods: The earthworm, *Aporrectodea longa,* was exposed to different concentrations of suspended particle phase (SPP) of drilling mud and cutting, collected from an abandoned oil drilling site in Mpanak, Niger Delta Region, for 28 days. Toxicological effects on the annelid was evaluated using standard procedure.
Results: At the end of the exposure, bioaccumulation of the toxic chemical constituents of the drilling mud (heavy metals and total hydrocarbons) increased ($p<0.05$) in whole tissue of earthworm with increase drilling mud concentration. Histopathological changes in the crop, gizzard and intestine of earthworms were moderate necrosis in the lower concentrations and severe necrosis in the higher concentrations.
Conclusion: The results of this study have shown that drilling mud and cuttings could cause serious health risk to *Aporrectodea longa*. The discharge of drilling mud into the terrestrial ecosystems should be discouraged or adequate treatment options for detoxification should be employed prior to discharge.

Keywords: Drilling mud; Toxicology; Heavy metals; Hydrocarbons; Histology; Annelid

Indexing: Index Copernicus, African Index Medicus

Introduction

Oil and gas exploration and development are major activities in the Niger Delta Region of Nigeria. These activities have environmental impacts arising from pollution of aquatic and terrestrial systems and toxixity to resident biota. The impact of drilling mud discharges in the Niger Delta ecosystem of Nigeria and the disappearance of the rich biodiversity of the Niger Delta, which is the key resource for sustainable development, is a current environmental concern [1]. Drilling mud/fluid is a term that refers to the lubricant used in oil drilling operations that stabilizes and lubricates the drill bit in the drilling process [2].They are suspension of solid in liquid emulsion and/or dissolved materials with chemical additives which are employed during exploration to remove cuttings. Drilling mud spent during drilling of oil and gas wells serve several important functions including coolling and cleaning the bit, maintaining pressure balance between the geological formation and the borehole, lubricating the bit, reducing friction in the borehole, seal permeability formations, stabilizing the borehole, and carrying cuttings to the surface for disposal. Drill cuttings are particles of crushed rock produced by the grinding action of the drill bit as it penetrates into the earth [3]. On the other hand, swamp cuts are stagnant man-made channel of water near the rig, usually subjected to seasonal flooding or tidal actions.

The effects of waste drilling fluids on swamp ecology may be more serious than on the marine environment when exposed for a pre-determined time [4]. When discharged untreated into the environment, drilling mud and cuttings are capable of interfering with normal functions of organisms such as circulatory system and antioxidant levels [5, 6]. Knowledge of the toxic potential and mechanisms of action of chemicals present in drilling mud and cuttings is critical for determining risk to wellbeing of organisms.

Earthworms have been used as biomarkers for assessing chemical environmental pollution and earthworms contribute to the aeration of the soil [7]. In soils, earthworms constitute 60 – 80% of the animal biomass and play a critical ecological role [8]. For example, it is estimated that under favourable conditions, earthworms can move up to 18 tons of soil per acre, per year, being continuously exposed to the soil through dermal contact and ingestion of soil materials. Earthworms can accumulate drilling mud in their body from soil. Histopathological response in earthworm have also been reported a valuable marker of toxicity in previous studies. These changes have been observed both following exposure to organic and inorganic pollutants: swelling, tissue necrosis, changes in chloragogenous cells and intestinal epithelium [9].The use of Histopathological responses as important biomarkers for the possible effects of toxic chemicals on organism has been reported to be effective [10]. It is imperative that histological biomarkers are the indicators of pollutants in the overall health of the entire population in the ecosystem [11]. The study of histological changes in the organ of animals is an accurate way to assess the effects of xenobiotic compounds in experimental studies [12].

The aim of this study is to assess the histopathological effects of spent oil based drilling mud and cuttings to earthworm, *Aporrectodea longa*.

Experimental

Annelids

One hundred and fifty *Aporrectodea longa* (mean weight 4.31g) were collected from a semi-pristine environment at the Botanical garden of the University of Benin, Benin City, Nigeria. The annelids were kept under standard condition .in the laboratory until used.

Study site description/Collection of samples: Toxicological effects of Spent oil based drilling mud and cuttings were collected from swamp cuts close to an abandoned oil drilling site in Mpanak (point 1: 4°33'7.35"N, 8° 0'19.31"E; point 2: 4°32'41.75"N, 8° 0'1.71"E; point 3: 4°33'3.36"N, 7°59'43.19"E; point 4: 4°32'39.58"N, 7°59'34.99"E), Niger Delta Region in Nigeria.

Treatment of Samples: . The procedure for the exposure was according to the Organisation for Economic Co-operation and Development (OECD) recommended method for testing chemicals No 207 [13]. Test concentrations of the drilling mud (0, 62,500, 125,000, 250,000 and 500,000 ppm of suspended particle phase (SPP) were prepared as earlier reported by Soegianto *et al* [14]. SPP is the unfiltered supernatant extracted from a 1:9 mixture of the drilling mud and water that was allowed to settle for 1 hr.Each test annelid was then exposed to the concentrations of the drilling mud for 28 days.At the end of the settling period, the SPP (1000 000 ppm) was decanted into a container. The different concentrations were made from this stock solution using distilled water as the diluent. Test soil (1kg) was spiked with each concentration of SPP in a glass jar containing 10 annelids in triplicates. The jars were covered with net of very small mesh size to allow aeration and prevent the annelids from wriggling out of the jars for 28 days. The annelids were then removed, cleaned and voided. Samples of each annelid was digested and analysed for toxicological effects as described by Bamgbose *et al* [15] and Booth *et al* [16], presence of heavy metals (bioaccumulation) using atomic absorption spectrophotometer and total hydrocarbons using UV/visible spectrophotometer (Searchtech 720). Histopathological alterations were observed in the organs (crop, gizzard and intestine) after fixing in 10% formal saline for 24 hr using an automatic tissue machine using standard procedure.

Data Analysis

Quantitative data were analyzed using one-way analysis of variance (ANOVA) followed by Duncan's Multi sample range post test using SPSS 15 software (SPSS Inc. Chicago). Statistical significance was considered at $p<0.05$ level of significance.

Results

At the end of the exposure, bioaccumulation of the chemical constituents of the drilling mud (heavy metals and total hydrocarbons) increased ($p<0.05$) with increase drilling mud concentration (Figures 1 and 2). Histopathology evaluation of the organs (Figure 3) revealed moderate and severe necrosis in the crop, gizzard, and intestine at the lower and higher concentrations, respectively.

Discussion

In this study, there was bioaccumulation of the toxic constituents (hydrocarbons and heavy metals) of the spent drilling mud and cuttings on the earthworm, *A. longa* exposed for 28 days. The constituents of drilling mud increased in earthworm tissue with increase in concentration of the drilling mud. This is consistent with the findings of Morgan [17] who studied the accumulation of metals (Cd, Cu, pb and Zn) by two ecological contrasating earthworm species (*Lumbricus rubellus and Aporrectodea calignosa*).

Findings from this study also showed that *Aporrectodea longa* manifested histopathological alterations in the heart, clitellum and blood vessels when exposed to different drilling mud concentrations. The crop, gizzard, and intestine and heart showed moderate necrosis in the lower concentrations and severe necrosis in the higher concentrations. In the clitellum severe necrosis was the only alteration that occurred in the 125,000, 250,000 and 500,000ppm SPP and brownish deposits were observed in the 62,500 and 500,000ppm SPP.

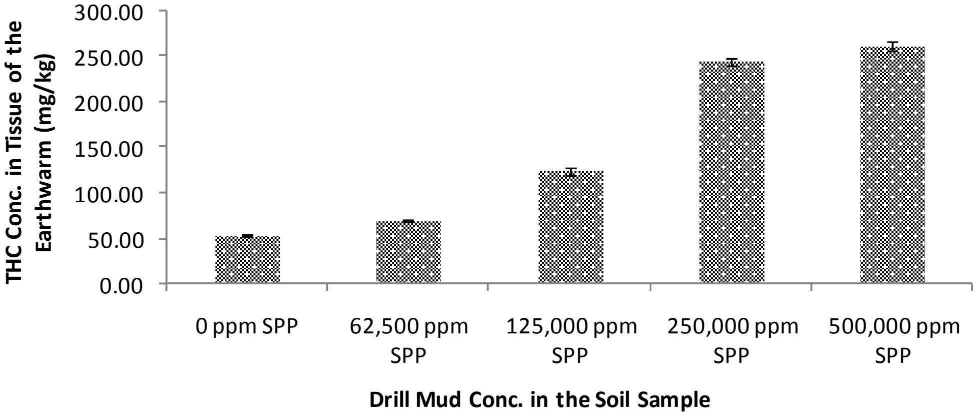

Figure 1: Bioaccumulation of total hydrocarbons in earthworms exposed to spent drilling fluid and cuttings

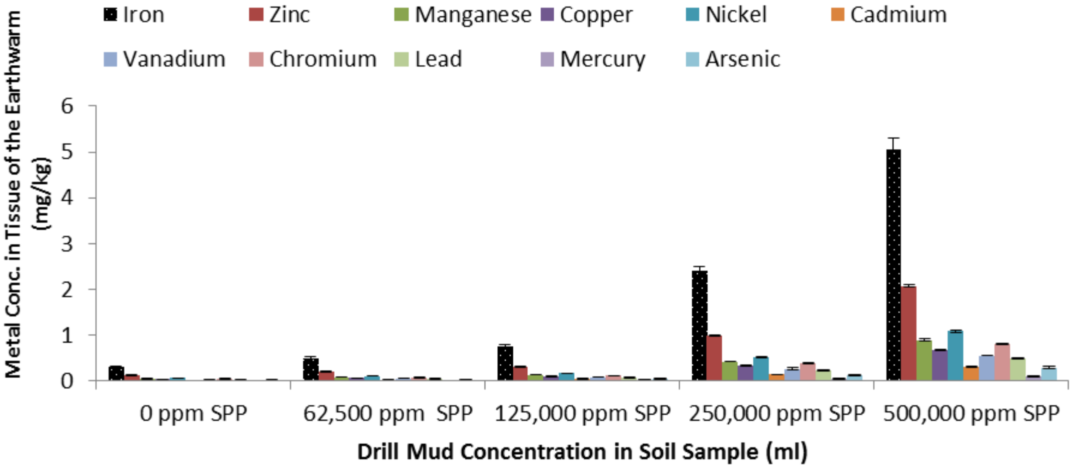

Figure 2: Bioaccumulation of heavy metals in earthworms exposed to spent drilling fluid and cuttings

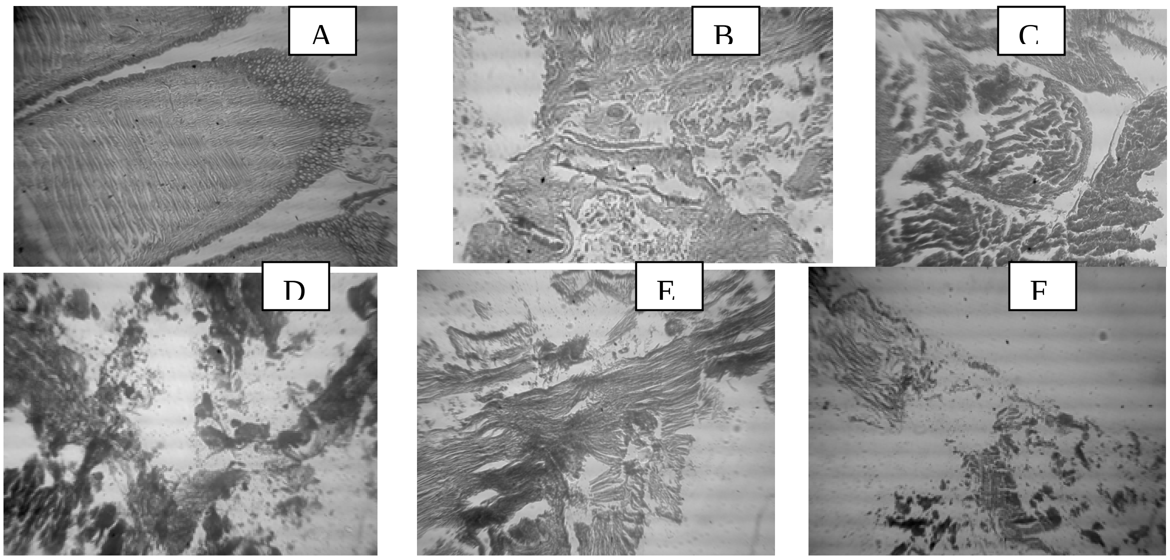

Figure 3: A - Crop of control earthworm showing distinct band of muscle and tissues observed after 28 days of exposure to drilling oil mud (H & E ×100), **B** - Crop of earthworm exposed to 500,000 ppm SPP of drilling mud resulting in severe necrosis after 28 days (H & E ×100), C - Gizzard of control earthworm showing distinct lumen of normal tissues observed after 28 days (H & E ×100), **D** - Gizzard of earthworm exposed to 250,000ppmSPP of drilling mud resulting in severe necrosis after 28 days (H & E ×100), E - Intestine of control earthworm showing distinct lumen of normal tissues observed after 28 days (H & E ×100), **F** - Intestine of earthworm exposed to 250,000ppmSPP of drilling mud resulting in severe necrosis after 28 days (H & E ×100).

Congestion of blood vessels (62,500 and 125,000 ppm SPP) and brownish deposits (250,000 and 500,000 ppm SPP) occurred in the blood vessels.

Ogeleka and Tudararo-Aherobo [18], reported the toxic effects of oil-based drilling mud (drilling waste) on brackish water shrimp (*Palaemonetes africanus*). This showed increase in mortality of the organisms with increased concentration of the drilling mud. Hart and Ulonnam [19], reported the toxicity and histopathological effects of oil-based drilling mud on edible periwinkle (*Tympanotonus fuscatus*). They reported irregular tissue shape, macrophage, inflammatory cells and basophilic spots in the organism. Agoes, *et al* [20] reported the toxicity of drilling waste and its impact on gill structure of post larvae of tiger prawn (*Penaeus monodon*).

The histopathological findings from this study varied with that of Kilic [21] who investigated the biological effects of soil pollution by evaluating the biochemical and histopathological responses of *Lumbricus terrestris* to heavy metals (Fe, Zn, Mn, Cr, Pb, Cu, Ni, Cd, Co, V, Hg and As). He showed that histopathological changes in the intestine of the worm included chloragogenous cells, deformation and fusion of epithelial cell lining, destroyed or distorted muscles and increased intestinal cell lining.

A reoccurring alteration in this study is necrosis. Necrosis means death of cells and when it occurs, it reduces the number of cells in the tissue and subsequently physiological dysfunction. A large number of studies have reported general histopathological changes in earthworm both following exposure to organic and heavy metal pollution [22,23]. Swelling tissue necrosis, changes in chloragogenous cells and brownish deposits in the tissues were the most common observation.

The biological and ecological responses to certain pollutants (organic and inorganic) may vary from changes at the population/community level, organ/tissue and even at the molecular level [24]. Since Histopathological changes produced by pollutants in organs and tissues can occur before they produce irreversible effects on the biota, histological method can be used in conjunction with other parameters and/or ecotoxicological bioindicators as an early warning system for the survival of the species as well as environmental protection [25].

The severity of histopathological changes shown in this study increased with increase in the concentration of the drilling mud. This may be due to direct toxic effects of the constituents of the drilling mud on tissues. Indiscriminate dumping of drilling mud at drilling sites, surroundings and even other environments should be discouraged, as it poses threat on the invertebrates living in soils, especially earthworm.

The accidental and intentional release of drilling mud wastes constitutes is a potential threat to environmental sustainability, soil biota and human health. The chemical compounds in the mud could also bioaccumulate in the organism and cause adverse maladies in the earthworm. There is therefore the need to adequately protect the delicate and rich biodiversity of the Niger Delta ecosystem.

Conclusion

This study has shown that drilling mud in soil is toxic and can cause severe damage to tissues and organs of earthworms. Histopathological alterations were studied in the clitellum, heart and blood vessels of the earthworms. Mild to severe necrosis and brownish deposit were observed which increased in severity with increase in concentration of drilling mud.

The results of this study have shown that drilling mud could inflict hisopathological maladies on *Aporrectodea longa*. Legislations should therefore be made to regulate, enforce and control the disposal of drilling mud spent during exploration, since oil exploration is the mainstay of Nigeria economy.

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Conflict of interest

There is no conflict of interest associated with this work.

Contribution to authorship

We declare that this work was done by the authors, Alex A Enuneku, Lawrence I Ezemonye and Mike Ajieh. Any liability pertaining to claims relating to the content of this article will be borne by the authors. The first two authors jointly conceived and designed the study. The third author collected the drilling mud samples from impacted site while all the authors jointly analysed the data.

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