HISTOLOGICAL MANIFESTATIONS OF EFFECTS OF ELIOZU DUMPSITE LEACHATE ON THE REPRODUCTIVE TRACT OF FEMALE WISTAR RATS

Abstract

Reproduction is an important biological trait to produce new individual organisms and is the fundamental for the life of an individual as well as the survival and development of the system. The response of reproductive organs to toxic substances varies from that of other target organs and may serve as an ideal for the deleterious effects of environmental toxicity on animals and human health. The incidence of infertility and risen maladies are increased in the last five decades. Various human activities have released into the environment plenty toxic substances which could be found in dumpsite leachate. The present study focused on experimental studies of histological effects of Eliozu dumpsite leachate on reproductive tract of female using Wistar rats as model. Twenty five (25) female Wistar rats were divided into five groups of five animals each; the leachate was collected from the dumpsite and water from near-by borehole also collected. Group 1 which served as control group received 1ml of commercial bottle water, group 2 received 1ml of borehole water 1kilometer from the dumpsite, groups 3, 4 and 5 received different concentration of the leachate in 10%, 50% and 100% for thirty days, the animals were sacrificed after being anesthetized with chloroform vapor, the ovaries and uterus were collected and put in 10% formaldehyde, sectioned and stained with haematoxylin and eosin (H&E) for histopathological studies. The slides were placed in a light microscope and analyzed by a consultant Anatomy pathologist. The results revealed that the Eliozu dumpsite leachate does not have any effect on the morphology of the uterus but have deleterious effect on the histomorphology of the ovaries. Eliozu dumpsite leachate has shown deleterious effect on female ovaries, which is an indication of toxicity that may cause infertility to the experimental animals. Therefore, it is recommended that further studies aimed at corroborating this finding be carried out especially on humans and residents around dumpsite should always embark on reproductive tract medical check-up.

Keywords; Eliozu dumpsite leachate, Histological effect, reproductive tract

INTRODUCTION

In developing countries, the dumpsite is the most cost-effective method of disposing of municipal solid garbage (Vaccari, et al, 2012). However, leachate, a liquid effluent, may leak from such a stockpile and contaminate the neighbouring water body (Chatham-Stephens et al, 2014). Uncontrolled overflow, rainfall, and infiltration into the underlying ground water aquifer or surrounding surface water can all expose the environment to dumpsite leachate. Leachate is increasingly recognised as a possible health hazard to both ecosystems and human populations (Al Sabbagh et al, 2012). Heavy metal concentrations were found in high concentrations in leachates at the Delta State Teaching Hospital Oghara dumpsite, according to studies (Nwaka et al, 2018). Characterization and toxicological assessment of leachates from the Air Hitman Sanitary Landfill in Malaysia using Pangasius Sutchis and Clarias

batrachus found that the leachate enclosed significant amounts of ammonia, dissolved organic matter, some semi-volatile organic compounds, and monocyclic aromatic hydrocarbons, all of which are highly toxic to both species of fish, and that these compounds could be used as indicators of leachate contamination in freshwater. Ammonical-nitrogen in the leachate is thought to be the main cause of fish mortality. However, benzene, toluene, and ethyl benzene may have played a role in the toxicity of the leachate (Tsai et al, 2010). Copper, chromium, lead, arsenic, cadmium, manganese, nickel, ammonia, chloride, and phosphate concentrations in the leachates samples of the Eliozu dumpsite were found to be beyond the allowed levels, according to Weleh and coworkers (Weleh et al, 2021). Heavy metals that may be present in leachate have been shown to have haematotoxic potential in rats. (Afolayan et al, 2012). In rats, raw and simulated leachates from the Olusosun municipal solid waste landfill were found to have immunotoxic potential (Subhasini et al, 2009). Subhasini et al. found changes in the biochemical and histopathological profile of the liver in Swiss Mice treated with distillery soil leachate at concentrations of 5%, 10%, and 20%. (Green et al, 2020). Hepatotoxicity and oxidative stress were also detected in rats in a similar survey in Olusosun municipal landfill leachate in Ojota, Lagos State, Nigeria. Neurologic lesions, neurodegenaration of purkinje cells with lose of dendrites, perineural vacoulations of the neuronal cytoplasm (spongiosis) and neuronal necrosis in the brain of Wistar rats exposed to Olusosun and Aba-Eku municipal landfill leachate has also been studied (Weleh *et al*, 2021); these reported brain tissue alterations correlate with a significant decrease in body weight gain and Superoxide dismutase activity but increase in absolute and relative brain weight gain, Malondialdhyde concentration and Catalase activity in both brain and serum in treated rats (Weleh *et al*, 2021)

The Eliozu Dumpsite is one of Rivers State's largest dumpsites, receiving untreated deposits of both domestic and industrial wastes from all over Port Harcourt, despite its potential direct and indirect hazards, given that residents in the dumpsite's vicinity rely heavily on borehole water for drinking. Secondly, despite the fact that infertility remains prevalent in our environment and premium placed on child bearing, there is paucity of literature on the possible effect of leachate on reproduction visa-viz reproductive tract. This study therefore attempts to evaluate the impacts of Eliozu dumpsite leachate on the histology of the reproductive tract using Wistar rats as experimental model. Experimental animals and data from the toxicological studies in Wistar rats is a way of forecasting human risk assessment.

Materials and Methods

Experimental Animals

Twenty-five female Wistar rats (10-12 weeks old, weighing about 200g) were obtained from the Animal House unit of Faculty of Basic Medical Sciences, University of Port Harcourt, Nigeria. The animals were cared for according to the general recommendation and provision of the University Ethical committee. Rats were housed in a climate controlled room (12 hours' dark and light cycle), with free access to drinking water and standard rodent chow (*ad libitum).*

Leachate collection and preparation

Raw leachate sample was collected from leachate well on the dumpsite at Eliozu in Obio/Akpor local government area of River State, Nigeria in clean 5litres plastic containers. The sample was taken to the laboratory of the Department of Chemistry, University of Port

Harcourt where it was filtered and prepared accordingly as described by (Green *et al*, 2020). The different concentrations of the leachate were gotten by serial dilution of leachate with distilled water.

Experimental Design

Following two weeks of acclimatization, the rats were divided into five groups of five Wistar rats each. **Group 1** served as the control and received 1ml of commercial bottled water; **Group 2** received 1ml of water obtained from borehole about 1km from the dumpsite; while **Groups 3, 4** and **5** received 1ml of 10% of leachate concentration, 1ml of 50% of leachate concentration and 1ml of 100% of leachate concentration respectively. All administrations were given once daily using an orogastric cannula for 30 consecutive days. On the 31st day however, the animals were anaesthetized using chloroform vapor and the ovaries and uterus were harvested for histological assay.

Histopathological Examination

Uterus and ovary collected were fixed with 10% formalin, embedded in paraffin, sectioned and stained with hematoxylin-eosin (H&E). Experienced pathologist performed pathological evaluation.

Histology of the Uterus and Ovary

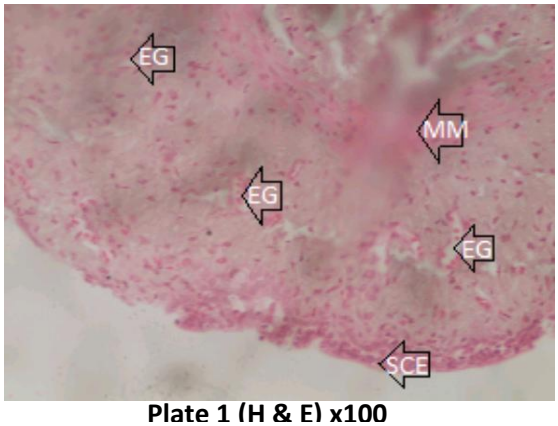

Plate 1 (H & E) x100

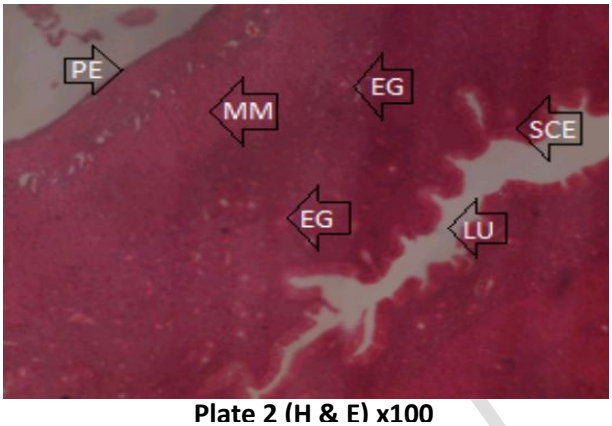

Plate 2 (H & E) x100

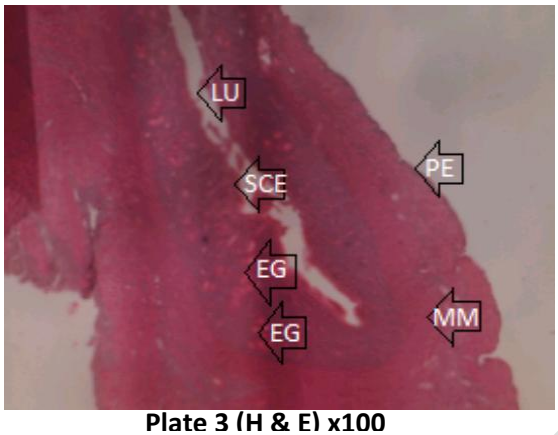

Plate 3 (H & E) x100

Plate 4 (H & E) x100

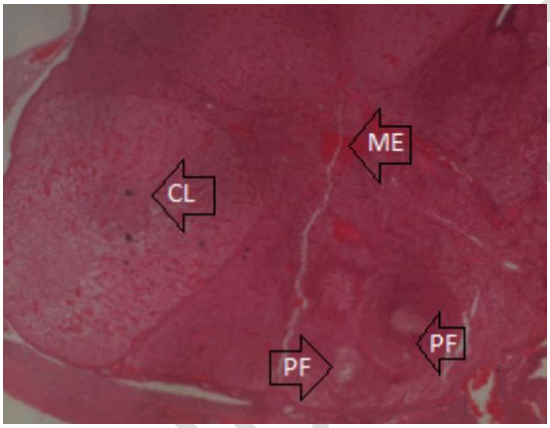

Plate 5 (H & E) x100

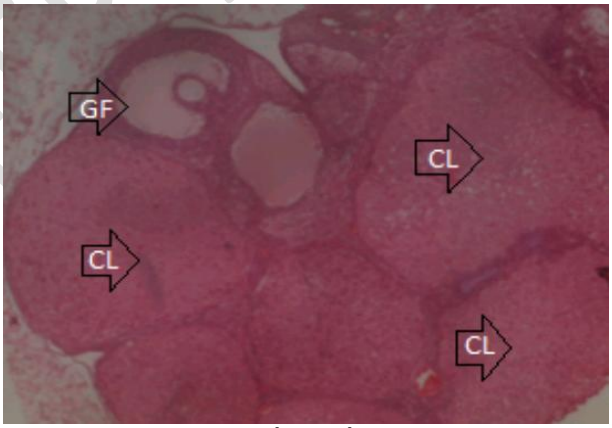

Plate 6 (H & E) x100

Results

The result of photomicrograph of the Wistar rats uterus stained with H&E at the same magnification (X100) displayed in Plates 1 to 3 normal Perimetrium (PM), Myometrium (MM), Endometrium (EM) containing endometrial glands, Simple columnar epithelium lining the endometrium in plate 1 (control). Plate 2 (borehole water) shows the same morphology when compared with the control and plate 3 (100% leachate) also showed the same morphology, without any alteration of the uterus. However, the result of photomicrograph of the Wistar rats ovary stained with H&E at the same magnification (X100) were displayed in Plate 4 to 6. Plate 4 (control) shows normal morphology of the ovary with Medulla (MD), Cortex containing primary follicle (PF), and corpus Luteum (CL), plate 5 also shows the same morphology of the ovary while plate 6 shows distorted ovary with swelling of the Graafian follicles. This alteration in the histology of the ovary examination implies a remarkable alteration of the ovary but not in the uterus by leachate from Eliozu.

Discussion

In developing countries paucity of suitable technological choices could be the reason solid waste are dumped (Vaccari, *et al*, 2012). Exposure to leachate causes health risk. The physicochemical properties of Eliozu dumpsite leachate is higher compared with the accepted values of National Environmental Standard Regulations Enforcement Agency as described by (Weleh *et al,* 2021)

The present study showed that there were no significant morphological changes in the examined uterus; this could be as a result of the short duration of exposure to the dump site leachate. We observed abnormalities in the ovary of the experimental animals that received pure concentration of leachate. There were cellular hypertrophy, degenerative and atrophic changes and swelling of ovarian graffian follicle when compared with the control group. This is in support with earlier report by ((Green *et al,* 2020), on the impact of Eliozu Port Harcourt landfill leachate on some reproductive hormones in female Wistar rats, where it was observed that Eliozu leachate altered female reproductive hormones. This finding indicates that the leachate may have deleterious effects on the oocyte of the ovaries of adult Wistar rats, in extension may contribute to the causes of female infertilities in the human population. It is similar with an earlier report by Eweka and Om'Imaboh's (2011),on the histological studies of effect of Monosodium Glutamate on the ovaries of adult Wistar rats. Cellular swelling is known to occur either directly or indirectly by denaturation of volume-regulating ATPases or indirectly by disruption of the cellular energy transfer pathway required for ionic regulation (Hinton and Laure, 1990). This study further supports an earlier report by Ayandiran *et al.,* 2017). The abnormality observed in the ovary is also similar with an earlier report by (Bakere *et al*, 2013), who reported that raw and simulated solid waste leachate from three dumpsite in Nigeria induce dose dependent increase in frequency of sperms with abnormal morphology in mice. There were similar reports in mice treated with leachates from electronic waste and hospital waste incinerated bottom ash (Akinbola *et al,* 2011). The causative agents of this reproductive abnormality could be lead and mercury as seen in the physicochemical analysis of Eliozu dumpsite leachate (Weleh *et al,* 2021).

Further studies on heavy metals revealed that arsenic, lead, cadmium cause health implications including disturbances in reproduction (Piotr *et.al*, 2015).

In a study, effect of long-term exposure of female Wistar rats to low levels of lead: ovary and uterus histological architecture changes, observed that lead values and the micromorphological structure changes found in the study in the ovaries, the uterus and fallopian tubes demonstrated the deleterious effect of lead, this study is similar with our work on ovary but differs in the morphology of the uterus (Eugenia *et al*, 2015).

Also record shows deleterious effect of the ovaries and uterus exposed to cadmium, lead and mercury on the structure and function of reproductive organs. It further explained that cadmium affects the maturation of follicles, degradation of the corpus luteum (Peter *et al*, 2020).

This observation suggests that leachate from Eliozu dumpsite is capable of inducing damage in germ cells in experimental animals indicating ovarian cyst and data from the toxicological studies in Wistar rats may be a way of forecasting human risk assessment.

Conclusion

The reproductive tracts of Wistar rats exposed to Eliozu dump site leachate has been examined using histological method. The examination showed no abnormality in the morphology of the uterus. However, the ovary of the experimental rats' exhibited morphological abnormalities which is an indication that dumps site leachate may contribute to the causes of female infertility. Therefore, it is recommended that further studies aimed at corroborating this finding be carried out especially on human population and medical check-ups be carried out on regular basis on the reproductive tracts.

Ethical approval

The ethical approval for this study was sought and obtained from the institution research ethics committee on 3rd July, 2018. The approval reference number is UPH/R&D/REC/04.

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