

HISTOLOGICAL 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 Elioazu dumpsite leachate on reproductive tract of female using Wistar rats as model. Twenty five 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 been anesthetized with chloroform vapor, the ovaries and uterus collected for histopathological studies. The results revealed that the Elioazu dumpsite leachate does not have any effect on the morphology of the uterus but have deleterious effect on the histomorphology of the ovaries. Elioazu 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.

Keywords; Elioazu dumpsite leachate, Histological effect, reproductive tract

INTRODUCTION

Dumpsite is the most economic viable means of municipal solid waste disposal practiced in developing countries [1]. However, liquid effluent known as leachate may drain from such stockpile and contaminate surrounding water body [2]. Exposure of the environment to dumpsite leachate may occur through uncontrolled overflow, rainfall and infiltration into the underlying ground water aquifer or nearby surface water. Leachate is now being recognized as a potential health risk to both surrounding ecosystems and human populations [3]. Studies on leachates from the dumpsite of Delta State Teaching Hospital Oghara reported high concentrations of heavy metals [4]. Also, Characterization and toxicological evaluation of leachates from Air Hitman Sanitary Landfill in Malaysia using *Pangasius Sutchis* and *Clarias batrachus* concluded that leachate contained significant quantities of Ammonia, dissolved organic matters, some semi-volatile organic compounds and monocyclic aromatic hydrocarbons which are very toxic to both species of fishes; and this may be used as indicators of leachates contamination in freshwater. Ammonical-nitrogen present in the

leachate is considered the principal cause of the fish mortality. However, benzene, toluene and ethyl benzene may have contributed to the leachate toxicity [5]. Weleh and co-workers reported concentrations of Copper, Chromium, lead, Arsenic, Cadmium, Manganese, Nickel, ammonia, Chloride and Phosphate in the leachates samples of Elioizu dumpsite above standard permissible limits [6]. The haematotoxic potentials of heavy metals that may be contained in leachate have been reported in rats [7, 8]. Immunotoxic potentials of raw and simulated leachates from Olusosun municipal solid waste landfill in rats has also been reported [9]. Subhasini *et.al* reported alterations in the biochemical and Histopathological profile of the liver in distillery soil leachate treated Swiss Mice at concentrations 5%, 10% and 20% of leachate [10]. A similar study at Olusosun municipal landfill leachate Ojota Lagos State, Nigeria also reported hepatotoxicity and oxidative stress in rats [11]. Neurologic lesions, neurodegeneration of purkinje cells with lose of dendrites, perineural vacuolations 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 [6]; 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, Malondialdehyde concentration and Catalase activity in both brain and serum in treated rats [6].

The Elioizu Dumpsite is one of the largest dumpsite in Rivers State; it receives untreated deposits of both domestic and industrial wastes from all over Port Harcourt and despite its possible direct or indirect hazardous nature, considering the fact that residents around the dumpsite largely depend on borehole water around the dumpsite for consumption. 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 Elioizu 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 (25) 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 Elioizu 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 (10).

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 oro-gastric 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

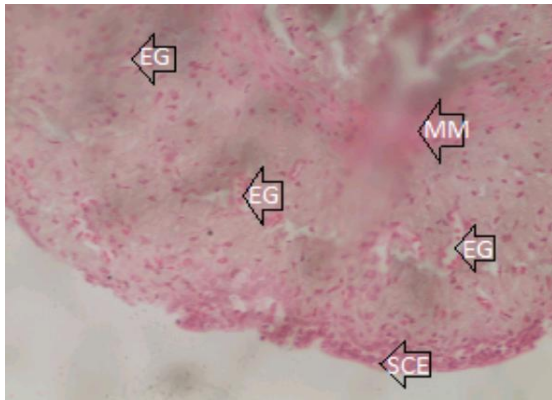


Plate 1 (H & E) x100

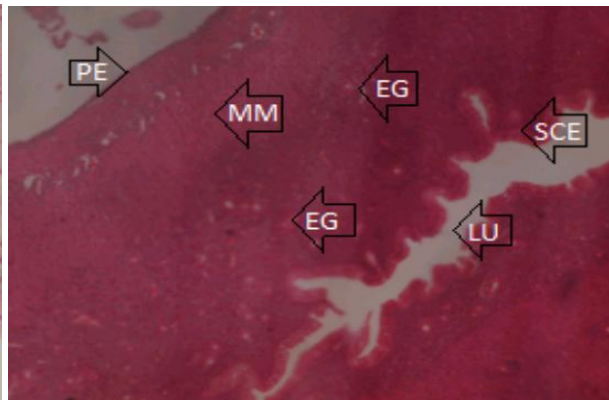


Plate 2 (H & E) x100

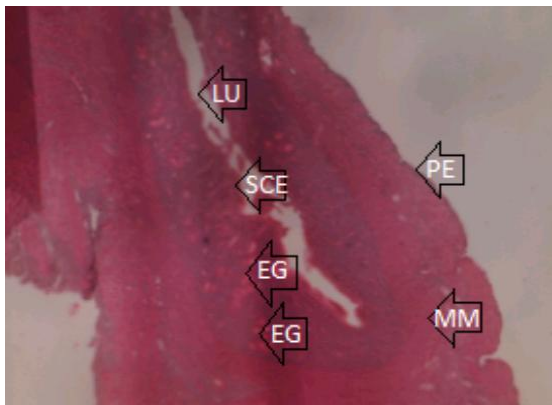


Plate 3 (H & E) x100

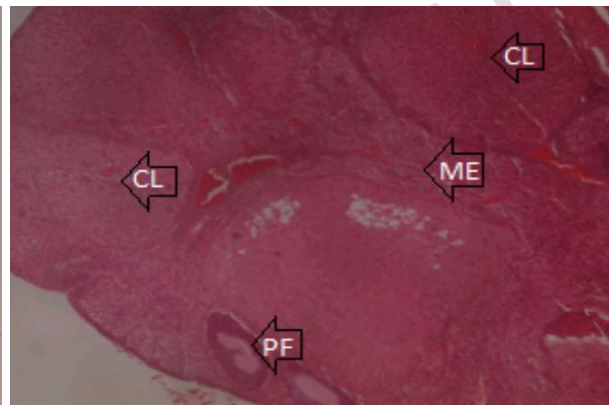


Plate 4 (H & E) x100

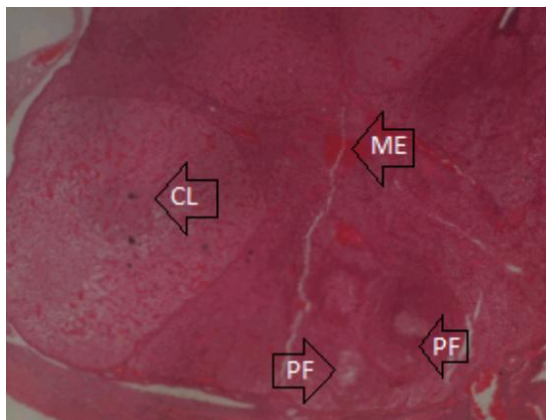


Plate 5 (H & E) x100

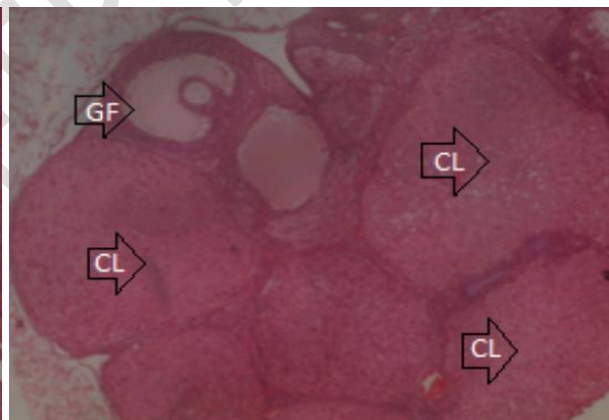


Plate 6 (H & E) x100

Result

The result of photomicrograph of the Wistar rats uterus stained with hematoxylin-eosin (H&E) at the same magnification (X100) displayed in Plate 1 to 3 shows 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) shows the same morphology, without any alteration of the uterus. However, the result of photomicrograph of the Wistar rats ovary stained with hematoxylin-eosin (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 graffian 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 Elioizu.

DISCUSSION

In developing countries paucity of suitable technological choices could be the reason solid waste are dumped (1). Exposure to leachate causes health risk. The physicochemical properties of Elioizu dumpsite leachate is higher compared with the accepted values of National Environmental Standard Regulations Enforcement Agency as described by (12)

The present study shows 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, this is in support with earlier report by (10) on the impact of Elioizu Port Harcourt landfill leachate on some reproductive hormones in female Wistar rats, where it was observed that Elioizu 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 (13) 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 (14). 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 (15), 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 (16). The causative agents of this reproductive abnormality could be lead and mercury as seen in the physicochemical analysis of Elioizu dumpsite leachate (6).

Further studies on heavy metals revealed that arsenic, lead, cadmium cause health implications including disturbances in reproduction (17).

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 histomorphological 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 (18).

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 (19).

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

Conclusion

The reproductive tracts of Wistar rats exposed to Elioza dump site leachate has been examined using histological method, which has been proven as adequate method to determine histopathological changes. The examination shows no abnormality in the morphology of the uterus. The ovary of the experimental rats' shows morphological abnormalities which is an indication that dump 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.

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.

References

- 1 Vaccari M, Torretta V, Collivignarelli C. (2012) Effect of improving environmental sustainability in developing countries by upgrading solid waste management techniques: A case study. *Sustainability*. 4:2852–2861
- 2 Chatham-Stephens K, Caravanos J, Ericson B, Landrigan P, Fuller R. (2014) The pediatric burden of disease from lead exposure at toxic waste sites in low and middle income countries. *Environmental. Research*. 132:379–383.
- 3 Al Sabbagh MK, Velis CA, Wilson DC, Cheeseman CR. (2012) Resource management performance in Bahrain: A systematic analysis of municipal waste management, secondary material flows and organizational aspects. *Waste Management. Research*.;30:813–824

- 4 Nwaka PO, B Anegbe, O Adeniyi, IG. Okunzuwa and A Jidonwo (2018). Impact of leachate on physicochemical properties of soil, within the vicinity of Oghara medical dumpsite, Delta State, Nigeria. *Physical Science International journal* 17 (1): 1-14.
- 5 Tsai CL, Krogmann U, Strom PF. (2010) Effect of forced aeration and mechanical turning on leachate quantity and quality from glass cullet stockpiles. *Journal of Environmental Engineering*.136(8):854-9.
6. Weleh I.I, I. E. Weleh and I. K. Green (2021), Impact of (ELIOZU) Dumpsite Leachates on Some Haematological Parameters in Wistar Rats. *International Journal of Research and Reports in Hematology*, 4(2): 70-78, 2021.
7. Adeyemi O, Oloyede O, Oladiji A (2006). Effect of leachate contaminated groundwater on the growth and blood of albino rats. *The internet journal of Hematology* volume 3, number 2.
8. Afolayan, OS, Ogundele FO and Odewumi SG (2012). Spatial variation in landfills leachate solution in urbanized Area of Lagos State, Nigeria. *American International journal of contemporary Research* 2 (8): 178-184.
9. Subhasini Sharma, Kalpana Sharma, Oivedita Yadav and KP. Sharma (2009) Alterations in Biochemical and Histopathological Profile of Liver in Distillery Soil Leachate Treated Swiss Albino Mice (*Mus Musculus L.*). *Pharmacologyonline* 3: 1047-1053
10. Green, I. Kinikanwo¹, Oriji, K. Vadunume¹ and Weleh, I. Ikechukwu (2020). Impact of Eliozu Port Harcourt Landfill Leachate on Some Reproductive Hormones in Female Wistar Rats, *International Journal of Research and Reports in Gynaecology* 3(1): 20-24.
11. Vaccari, M, Torretta, V, Collivignarelli, C (2012). Effect of improving environmental sustainability in developing countries by upgrading solid waste management techniques: A case study. *Sustainability*, 4, 2852–2861.
12. Weleh I.I, Weleh I. E. and Green I. K. (2021). Impact of (ELIOZU) Dumpsite Leachates on Some Haematological Parameters in Wistar Rats, 4(2): 70-78.
13. Eweka A.O and Om'Imaboh's F.A.E (2011). Histological studies of the effects of monosodium glutamate on the ovaries of adult wistar rats. *Annals of Medical and Health Sciences Research*.1 (1):37-43.
14. Hinton DE and Laure DJ (1990). Liver Structural Alterations Accompanying Chronic Toxicity in Fishes; Potential Biomarkers of Environmental Contamination. In: McCarthy JF, *Biomarkers of Environmental Contamination* Boca Raton: Lewis 17-57.
15. Bakere AA, Alimba CG and Alabi OA (2013). Genotoxicity and mutagenicity of solid leachates: A review. *African Journal of Biotechnology* Volume 12 (27): 4206-4220.
16. Akinbola Temitayo, Adetutu Adeyemi, Olajumoke A. Morenikeji and Adekunle Bakere (2011). Hospital waste incinerator bottom ash leachate induced cytogenotoxicity in *Allum cepa* and reproductive toxicity in mice. *Toxicology and industrial health* 27(6):505-514.

17. Piotr Rzymiski, Katarzyna Tomczyk, Pawel Rzymiski, Barbara Poniedzialek, Tomasz Opala and Maciej Wilczak (2015). Impact of heavy metals on the female reproductive system. *Annals of Agriculture and Environmental medicine* 22 (2): 259-264.

18. Eugenia DUMITRESCU, Victoria CHIURCIU, Florin MUSELIN, Roxana POPESCU, Diana BREZOVAN, Romeo T. CRISTINA (2015). Effects of long-term exposure of female rats to low levels lead: ovary and uterus histological architecture changes. *Turkish journal of Biology* 284-288.

19. Peter Massányi, Martin Massányi, Roberto Madeddu, Robert Stawarz and Norbert Lukác (2020). Effects of Cadmium, Lead, and Mercury on the Structure and Function of Reproductive Organs. *Toxic.* 8, 94;