**Subchronic Oral Administration of Lead Exacerbate Liver and Kidney Toxicity in Male Wistar Rats****J.O. Fatoki^{1*}, T.G. Atere¹, B.J. Dare², O.S. Tokunbo², H.A. Abdulrahim³ and O. Iyapo⁴**¹Department of Medical Biochemistry, Faculty of Basic Medical Sciences, Osun State University, Osogbo, Nigeria.²Department of Anatomy, Faculty of Basic Medical Sciences, Osun State University, Osogbo, Nigeria.³Department of Medical Biochemistry, Faculty of Basic Medical Sciences, University of Ilorin, Ilorin, Nigeria.⁴Department of Morbid Anatomy, Eko University of Medicine and Health Sciences, Ijanikin, Lagos, Nigeria.

ARTICLE INFO

Received: Jan 2022

Accepted: Apr. 2022

Keywords:

Lead, aminotransferase, creatinine, urea, histology

Corresponding author:

Dr. John Olabode Fatoki

john.fatoki@uniosun.edu.ng

ORCID iD: 0000-0003-3246-9172

DOI: 10.55518/fjpas.KNBY5971**Abstract**

Lead is an environmental and occupational toxicant. Compelling experimental data from previous studies have shown that following lead exposure, highest concentration of the absorbed lead is stored in the liver and the kidney, and this is expected to have severe consequences on the two organs. In this study, we investigated the effects of lead on the hepatic and renal structure and functions. Rats ($n=24$) were grouped into four of six animals each. Group 1 (control) received drinking water only. Groups 2, 3 and 4 were orally exposed to 50, 100 and 150 mg/kg body weight of lead respectively for 12 weeks, after which, blood, liver, and kidney were collected from the animals for biochemical and histological studies. One-way analysis of variance followed by Tukey's test was used to analyze the results with ($p < 0.05$) considered significant. Lead exposure caused a general decline in the percentage body weight gain and percentage relative liver weight. Physical trauma was also observed in the lead-treated rats. The lead exposure also significantly ($p < 0.05$) enhanced the plasma activities of aspartate and alanine aminotransferases, gamma-glutamyl transferase, lactate dehydrogenase, and alkaline phosphatase. Furthermore, a significant increase ($p < 0.05$) was observed in the concentration of creatinine and urea in the plasma of the lead-exposed animals. The histological study also revealed several histological alterations in the liver and kidney of lead-exposed animals relative to the control animals. These findings indicated that exposure to lead may have adverse effects on the structure and function of both the liver and the kidney.

1.0 Introduction

Lead is a soft silvery grey heavy metal that is found everywhere in the environment poisoning millions of people [1] and constituting a key public health challenge [2]. Lead is a non-essential trace element and a naturally occurring toxic metalloid that is found in the Earth's crust. It has no specific biological role [3]. Meanwhile, industrially, lead is a very useful metal all over the world because of the chemical characteristics it exhibits such as ductility, malleability, resistance to corrosion and poor conductivity [4], [5], [6].

The major sources of lead exposure include exhaust emission, lead-contaminated soil, food, water, and dust. Individuals working in muffler repair facilities, battery, paint, plumbing, ceramic, and cable manufacturing industries are also occupationally exposed to lead [7], [8], [9]. There is no safe or permissible level of lead exposure in humans, because, even at low doses lead exposure has been found to induce significant health challenges [10]. Once ingested, lead is readily absorbed into the bloodstream. Most of the absorbed lead bonds to the erythrocytes, while

plasma transports the remaining one to the tissues. The half-life of lead in the plasma is about 25 days [11].

Lead exposure had been implicated in neurotoxicity, anemia, cardiovascular diseases (CVD), hypertension, immunotoxicity and infertility [12]. Studies have shown that inhibition of the activity of antioxidants, increased production of free radicals, and induction of lipid peroxidation are some of the mechanisms through which lead exerts its toxicity [13]. Furthermore, lead has been implicated in the modification of xenobiotic metabolic pathway, and perturbation of lipid metabolic homeostasis [14]. Specifically, [15] established a strong link between derangement of lipid metabolism and chronic lead exposure.

[16] had previously identified the liver and kidney as the largest reservoir of lead accumulation in the body after short-time exposure to lead. Although there are some documented data in the literature that attempt to discuss the effect of lead on liver and kidney function, most of these are contradictory [6] and are not well discussed [17]. The present research aimed at closing this knowledge gap. In this study, we investigated the effects of lead on plasma activities of alanine aminotransferase (ALT), aspartate aminotransferase (AST), gamma-glutamyl transferase (γ -GT), alkaline phosphatase (ALP), lactate dehydrogenase (LDH), plasma concentration of urea and creatinine, as well as on liver histoarchitecture.

2.0 Materials and methods

2.1 Chemicals and reagents

Chemicals used for this study include lead acetate and diethyl ether. They were of pure grade and were procured from Sigma-Aldrich, Missouri, USA. Diagnostic kits for AST, ALT, γ GT, ALP, LDH, creatinine, and urea were products of RANDOX® Diagnostic, London, United Kingdom.

2.3 Animals and treatments

Twenty-four (24) healthy male albino rats with body weights between 120 and 200 were used for this study. The rats were kept in a compartmentalized metal cage and were acclimatized for seven (7) days prior to lead

exposure. The animals were allowed free access to a standard pelletized diet and drinking water *ad libitum*. They were randomized into four (4) groups of 6 animals each. Group 1 served as control and received water only. Group 2, 3 and 4 received 50, 100, and 200 mg/kg bwt of lead (II) acetate trihydrate $[(CH_3COO)_2Pb \cdot 3H_2O]$ respectively. The $CH_3COO)_2Pb \cdot 3H_2O$ was prepared in water and administered orally for 12 weeks. Animals were maintained at a temperature of $22 \pm 2^\circ C$ and 12 hours of regular light-dark cycle throughout the study. All measures for animal handling followed protocol of the Committee on Experimental Animals Ethics of Biochemistry Department, LAUTECH, Ogbomoso (LAU/BCH/21/0013).

At the termination of lead administration, blood samples were taken from the rats via cardiac puncture into a heparinized tube, after an overnight fast, under light ether anesthesia. The blood samples were spun at 5000 rpm to separate the plasma and the erythrocytes. All plasma samples were preserved in a freezer at $-20^\circ C$ until analyzed. Liver and kidney were also excised from the animals and were cleansed of blood and attached tissues, rinsed in normal saline, and a part of it was fixed in formalin for histological study.

2.4 Biochemical analyses

2.4.1 Determination of plasma aminotransferases activity

The activities of AST and ALT in the plasma were evaluated using RANDOX diagnostic reagent kits according to the method described by Reitman and Frankel [18]. Similarly, the activity of γ -GT in the plasma was evaluated using the reconstituted diagnostic reagent kit manufactured by RANDOX Diagnostics, following the method described by [19].

2.4.2 Determination of plasma alkaline phosphatase activity

The ALP activity was determined in the plasma using RANDOX diagnostic kit, following the recommendations of the German Society for Clinical Chemistry [20].

2.4.3 Determination of plasma creatinine concentration

The concentration of creatinine in the plasma was determined using RANDOX diagnostic kit following the method of Bartel *et al.* [21].

2.4.4 Determination of plasma urea concentration

Plasma concentration of urea was determined using RANDOX diagnostic kit as described by the Urease-Berthelot method [22].

2.4.5 Determination of plasma activity of lactate dehydrogenase

The activity of LDH in the plasma was assayed with the aid of the RANDOX kit. The assay conditions were largely based on the method described by [23].

2.5 Histopathology

After dehydration, clearing, and embedding, the paraffin block containing the tissue was sectioned by a rotary microtome. The microtome knife was placed at 45°C and the tissue sectioning was done at 4 µm. The sectioned tissues (kidney and liver) cut in ribbons were allowed to float in warm water bath at 40°C and then collected on slide for staining. Histological staining was carried out using Haematoxylin and Eosin as described by [24]. The slides were captured with a binocular microscope (Olympus, New Jersey, USA) linked to a 5.0 MP camera (Amscope Inc, USA).

2.6 Statistical analysis

The data obtained in this study were expressed as mean $\pm$ SD. The levels of homogeneity among the groups were assessed with One Way Analysis of Variance (ANOVA). Where heterogeneity occurred, the groups were separated using Tukey Test. All analyses were done using GraphPad Prism® version 5.

3.0 Results

3.1 Effects of oral administration of lead on body and liver weight, weight change, and relative liver weight of rats

The initial and final body weight of rats, and the liver and relative liver weight of rats exposed to lead is shown in Table 1. The animals in control

group had 44% increase in body weight while the groups that received 50 and 100 mg/kg bwt lead increased only by 25% and 27% respectively. Furthermore, lead intoxication caused 8% reduction in the bodyweight of animals that received 150 mg/kg bwt of lead. Similarly, compared with the control group, lead at the various doses tested caused a significant ($p < 0.05$) reduction in the relative liver weight. The reduction amounted to 3.55% in the control group and 2.96%, 2.78% and 2.58% in the groups that were exposed to 50, 100 and 150 mg/kg bwt lead, respectively.

3.2 Effects of oral administration of lead on alanine aminotransferase, aspartate aminotransferase, and gamma-glutamyl transferase activities in rat

The result from the assessment of hepatic transaminases (ALT, AST, and γ -GT) in the plasma of Wistar rats exposed to lead is presented in Figures 1 – 3. As shown, repeated oral administration of lead resulted in a significant ($p < 0.05$) enhancement of the activities of these enzymes in the plasma of the exposed animals compared with the control group. Lead exposure increased the activity of ALT (Figure 1) from 13.15 ± 2.05 (control) to 16.27 ± 0.82 (50 mg/kg bwt), 20.32 ± 2.57 (100 mg/kg bwt), and 30.76 ± 6.35 (150 mg/kg bwt). Similarly, oral administration of lead enhanced the activity of AST (Figure 2) in the plasma of the animals exposed to lead at 50, 100, and 150 mg/kg bwt by 88%, 19%, and 100% respectively compared with the animals in the control group. Furthermore, the most prominent enhancement of γ -GT (greater than 3-fold) was observed in the group that was exposed to 100 mg/kg bwt (Figure 3). Although to a lesser extent, lead at 50 and 150 mg/kg bwt also increased the activity of the enzyme by about 2-fold and 1.5-fold respectively compare with the control.

3.3 Effects of oral administration of lead on alkaline phosphatase activities in rats

The effect of lead exposure on the activity of ALP in the plasma of animals exposed to lead is presented in Figure 4. As illustrated, treatment with lead caused a dose-dependent, but significant ($p < 0.05$) increase in the activity of ALP in the plasma of the treated animals. Specifically. When

compared with the control, lead caused a 20% (50 mg/kg bwt), 42% (100 mg/kg bwt), and 57% (150 mg/kg bwt) increase in the activity of the enzyme.

3.4 Effects of oral administration of lead on creatinine and urea concentration in rat

Results from the determination of creatinine and urea concentration in the plasma of animals exposed to lead are presented in Figures 5 and 6. As illustrated (Figure 5), lead exposure significantly ($p < 0.05$) increased the plasma creatinine concentration from 5.60 ± 0.55 (control), to 14.86 ± 2.10 (50 mg/kg bwt), 18.54 ± 2.40 (100 mg/kg bwt), and 8.91 ± 1.20 (150 mg/kg bwt). Similarly, lead exposure caused a significant ($p < 0.05$) increase in the plasma urea concentration (Figure 6). The increase in the plasma urea concentration caused by lead at 50, 100, and 150 mg/kg bwt amounted to 2-fold, 2.3-fold, and 1.5-fold respectively when compared with the control.

3.5 Effects of oral administration of lead on lactate dehydrogenase activity in rat

The activity of LDH as affected by lead exposure is presented in Figure 7. Lead exposure significantly ($p < 0.05$) elevated the plasma activity of LDH relative to the control. Specifically, lead at 50, 100, and 150 mg/kg bwt enhanced the plasma LDH activity by 58%, 38%, and 81% respectively.

3.6 Effects of oral administration of lead on hepatic histoarchitecture

Effects of oral administration of lead on the hepatic patho-anatomical profile of rats in the control group (A) and the lead-exposed groups (B, C, and D) are shown in Figure 8. The hepatic histological outline of the animals in the control group was well preserved. The profile was devoid of any cellular alterations. The portal triad (PT) was not occluded, and it was devoid of blood cell deposition. The

hepatocytes appear normal. On the other hand, the outline of the liver from the lead-exposed groups (B, C, and D) displayed several histological alterations. These include distorted morphology of the hepatocytes, marked deposition of blood cells (red asterisk), and occlusion of the portal triad (yellow arrow). This observed histological modification in various treatment groups relative to the control is suggesting a lead-induced alteration of the hepatic functional integrity.

3.7 Effects of oral administration of lead on renal histoarchitecture

As shown in Figure 9, the renal cytoarchitecture of the control appeared normal with no pathological lesion seen, the renal cortex is characterized by normal glomeruli and capsular spaces, proximal and distal convoluted tubules. Also present are lacis cells (slender black arrow) between the afferent and efferent arterioles. Conversely, the morphological presentation of rats exposed to varying doses of lead (groups B, C, and D) showed moderate to severe pathological changes which were dose dependent. Rats in group B treated with 50 mg/kg bwt of lead presented with a mildly normal architecture with attenuated proximal convoluted tubules. On a general outlook, the renal cortex shows normal glomeruli with normal mesangial cells and capsular spaces (yellow arrow), the extraglomerular lacis cells also appear normal (slender black arrow). The histoarchitecture of the kidney of rats exposed to lead at 100 and 150 mg/kg bwt (groups C and D) showed marked changes characterized by few glomeruloscleroses (big black arrow), moderately enlarged capsular space (red arrow), dilatation, and necrosis of the distal convoluted tubule. The presence of interstitial fibrosis (green arrow) and scarring of blood (white arrow) vessels are also observed.

Table 1: Body and liver weight, weight change, and relative liver weight in rats orally exposed to lead for 12 weeks

Group	Initial body weight (g)	Final body weight (g)	Weight change (%)	Liver weight (g)	Relative liver weight (%)
Control	141.60 ± 11.95 ^a	203.40 ± 19.54 ^a	44	7.22 ± 0.89 ^a	3.55 ± 0.26 ^a
50 mg/kg bwt	171.20 ± 17.92 ^b	214.00 ± 23.81 ^a	25	6.32 ± 0.94 ^a	2.96 ± 0.30 ^b
100 mg/kg bwt	166.60 ± 15.44 ^a	211.80 ± 24.60 ^a	27	5.90 ± 0.91 ^a	2.78 ± 0.26 ^c
150 mg/kg bwt	180.20 ± 10.21 ^c	166.40 ± 47.05 ^a	-8	4.78 ± 1.15 ^b	2.58 ± 0.36 ^d

Each value is mean ± SD of six animals in each group. Rows with different alphabets are significantly different at $p < 0.05$ across the groups.

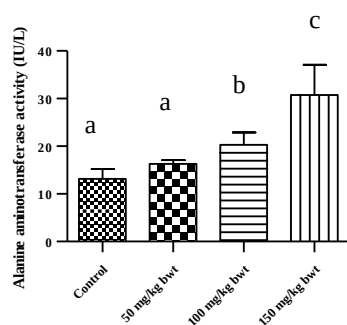


Figure 1: Activity of ALT in the plasma of rat orally exposed to lead. Each value is mean ± SD of 6 rats in the group. Bars with different alphabets are significantly different at $p < 0.05$ across the groups

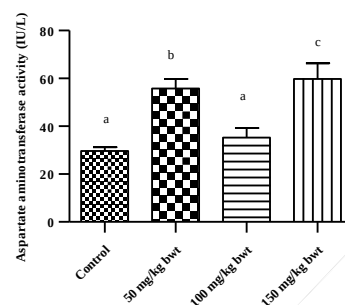


Figure 2: Activity of AST in the plasma of rat orally exposed to lead. Each value is mean ± SD of 6 rats in the group. Bars with different alphabets are significantly different at $p < 0.05$ across the groups

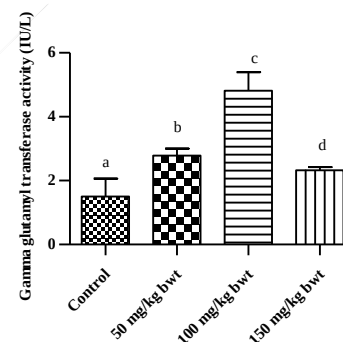


Figure 3: Activity of γ -GT in the plasma of rat orally exposed to lead. Each value is mean ± SD of 6 rats in the group. Bars with different alphabets are significantly different at $p < 0.05$ across the groups

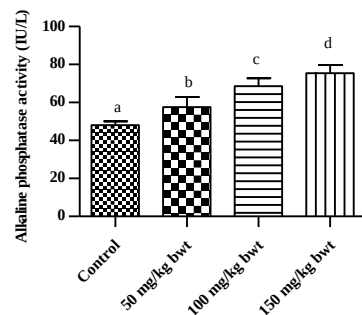


Figure 4: Activity of ALP in the plasma of rat orally exposed to lead. Each value is mean $\pm$ SD of 6 rats in the group. Bars with different alphabets are significantly different at $p < 0.05$ across the groups

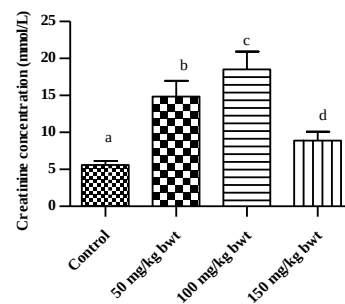


Figure 5: Plasma creatinine concentration in rat orally exposed to lead. Each value is mean $\pm$ SD of 6 rats in the group. Bars with different alphabets are significantly different at $p < 0.05$ across the groups

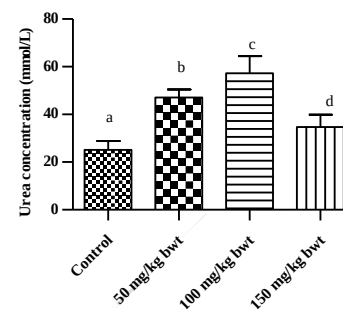


Figure 6: Plasma urea concentration in rat orally exposed to lead. Each value is mean $\pm$ SD of 6 rats in the group. Bars with different alphabets are significantly different at $p < 0.05$ across the groups

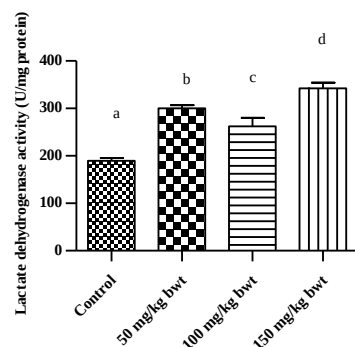


Figure 7: Activity of LDH in the plasma of rat orally exposed to lead. Each value is mean $\pm$ SD of 6 rats in the group. Bars with different alphabets are significantly different at $p < 0.05$ across the groups

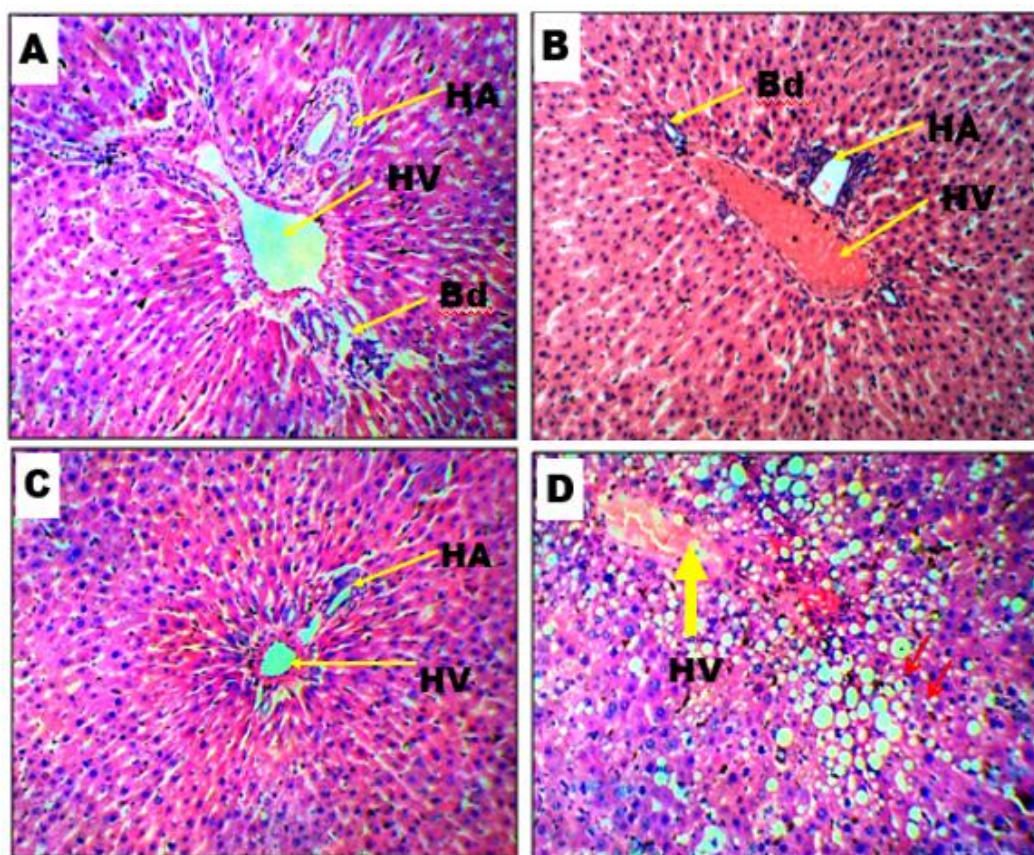


Figure 8: Photomicrograph of the hepatic histoarchitecture of experimental animals (H&E, x40).
Legend: HV- hepatic vein; HA- hepatic artery; BD- Bile duct.

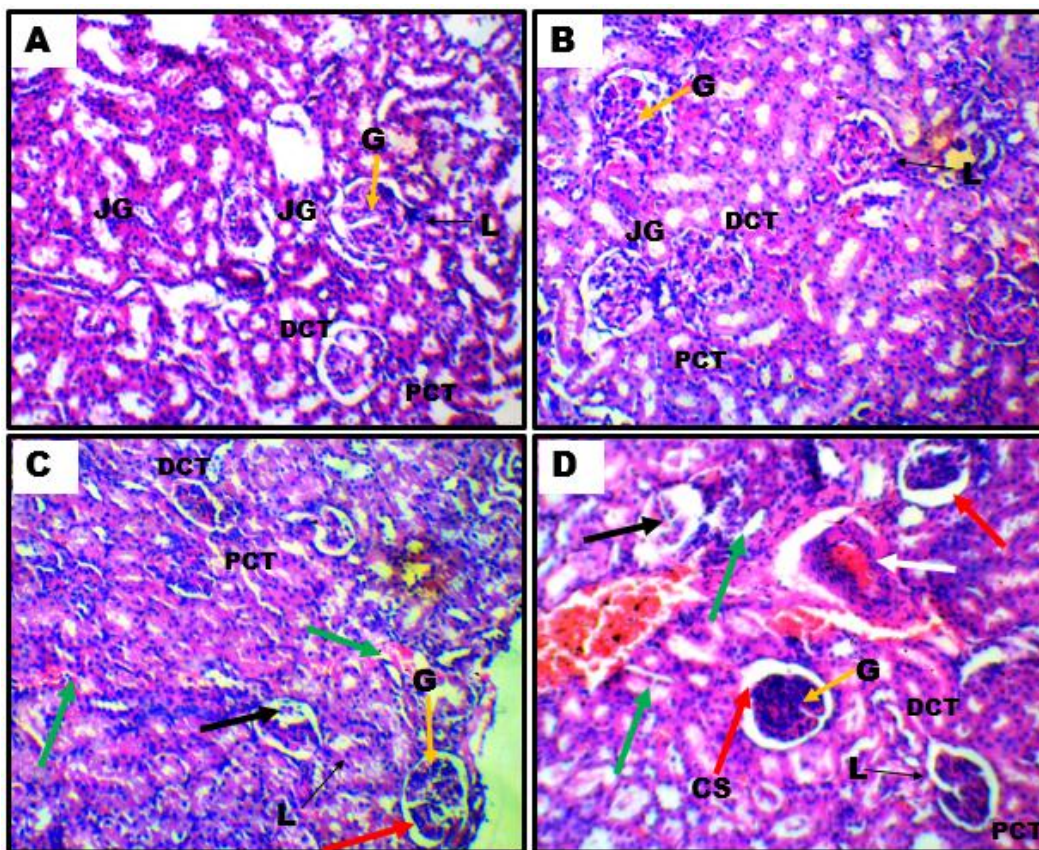


Figure 9: Photomicrograph of the histoarchitecture of the Kidney of experimental animals (H&E, x40)

Legend: G-Glomerulus; PCT- Proximal convoluted tubule; DCT- Distal convoluted tubule; L- Lacis cells (extraglomerular mesangial cells); CS- Capsular space; JG- Juxtaglomerular cells.

4.0 Discussion

Lead is a ubiquitous environmental and industrial toxicant [25]. Moreover, previous studies have shown that following chronic lead exposure, nearly all the lead that is absorbed by the body is stored majorly in the kidney and liver [26]. It will not be out of order, therefore, to hypothesize that exposure to lead might initiate pathogenesis of hepatic and renal diseases. In this study, we beamed our searchlight on the possible effects of sub-chronic lead exposure on hepatic and renal structural and functional integrity.

Data obtained from this study revealed that sub-chronic oral exposure to lead could be linked with hepato- and renal toxicity. This is characterized by a significant decline in body weight gain and relative liver weight, increased activity of ALT, AST, γ -GT, ALP, and LDH in the plasma. Similarly, the

exposure also elevated the plasma concentration of creatinine and urea.

Abnormality in the body weight gain is considered a valuable marker of toxicological index [27]. Similarly, the physiological and pathological state of experimental animals can also be marked by the weight of the organ especially, the liver [28]. The decrease in the weight gain of the experimental animals exposed to lead might be a resultant effect of lead-induced systemic toxicity. This correlate with the findings of [29], where a decline in the bodyweight of experimental animals exposed to cement dust was observed [29]. It is noteworthy that cement dust contains heavy metals including lead. The observed decline in body weight of rats as observed in this study could also be because of anorexia. Although both the control and the groups that were exposed to lead in this study had unlimited access to food and water.

Availability of food notwithstanding, it is very possible that lead exposure might have perturbed the pathway of the neurotransmitter that is involved in the control of food consumption thereby altering control of appetite and energy homeostasis [30]. Similarly, relative organ weight is an important biomarker of organ toxicity, it is equally a very sensitive endpoint of toxicological index [31]. The discrepancies observed in relative liver weight in the lead-exposed animals relative to the control group are not unexpected because the liver is the focal organ that is directly affected by any toxicant including lead [32]. The reduction in relative hepatic weight might have also occurred as a direct resultant effect of the general decrease in the body weight gain as observed in this study.

The liver is the main organ of the body that is responsible for the metabolism of xenobiotics and are therefore the major target of chemical-induced toxicity [33]. Activities of ALT, AST, γ -GT, and ALP in the plasma are important endpoints for the diagnosis of hepatic disease [34]. These enzymes are usually localized in the hepatocytes, although they are also present (but to a lesser extent) in some other organs such as the heart, kidney, and muscle [35]. The increase in the plasma activities of these enzymes in the experimental animals that were exposed to lead relative to the control is suggestive of possible hepatocellular injury [33].

Creatinine and urea are by-products of amino acids metabolism and are released into the circulation from where they are freely filtered by the kidney for excretion through the urine. The plasma concentration of both the creatinine and urea reflect the renal functional integrity as it represents the balance between their production and elimination by the kidney. Kidney's failure to effectively filter and eliminate creatinine and urea due to advanced renal disorders and significant decrease in the glomerular filtration rate is expected to increase the level of these substances in the plasma [33]. Our

findings in this study revealed that lead exposure increased the concentration of urea and creatinine in the plasma relative to the control. This suggests a possible compromise in the ability of the kidney to effectively filter them for urinary elimination. This is an indication of a possible nephrotoxic potential of lead in experimental animals.

Lactate dehydrogenase is a key glycolytic enzyme [36]. Lactate dehydrogenase activity has been a reliable marker for the evaluation stress [37]. It is also an effective marker used for the assessment of toxicity of xenobiotics and in the diagnosis of cellular and tissue damage [38]. Plasma LDH activity increase because of tissue and cellular damage, LDH does not normally accumulate in a healthy cell, when not needed, it is usually destroyed and excreted [39]. The fact that the significant elevation of plasma LDH activity as observed in this study might be a result of lead-induced tissue damage and cannot be ruled out. It is suggested that the observed increase in the activity of plasma LDH in the lead-exposed group relative to the control animals could be a result of lead-induced tissue damage exemplified by possible hepatocellular and renal damage observed in this study.

Histological investigations play a sensitive role in the confirmation of pathological derangements at the tissue level in response to exposure to toxic substances [29]. It provides valuable information as to what extent is the level of damage induced by the exposed toxicant [40]. In this study hepatic and renal histological examinations of the animals in the control group revealed normal renal and hepatic histoarchitecture, demonstrating no pathological lesion. Meanwhile, animals that were exposed to the varying doses of lead (Figures 8 and 9) were presented with morphological perturbations of the renal and hepatic microarchitecture

Conclusion

Our findings in this study revealed that sub-chronic exposure to lead produced different patterns of toxicity. The present study

identified increased activities of AST, ALT, γ -GT, ALP and LDH in the plasma of the lead-exposed animals relative to the control animals. It also increased the plasma concentration of creatinine and urea. Additionally, lead exposure also perturbed the hepatic and renal histoarchitecture of the lead-exposed rats. These might be some of the denominators that mediate the disease endpoint of lead.

Declarations

Conflict of interest

The author declares no conflict of interest.

Funding

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Author contribution

JOF conceived and designed the study, conducted research, provided research materials, and collected and organized data. TGA, BJD, OST, HAA, IO analyzed and interpreted data. JOF, TGA, and BJD wrote the initial and final draft of the article and provided logistic support. All authors have critically reviewed and approved the final draft and are responsible for the content and similarity index of the manuscript.

Acknowledgments

The author acknowledged the technical assistance of the Technical Staff in the Department of Biochemistry, LAUTECH, Ogbomoso, Nigeria.

Reference

[1] Nakhaee, S., Amirabadizadeh, A., Brent, J. and Mehrpour, O. (2019). Impact of chronic lead exposure on liver and kidney function and haematologic parameters. *Basic Clin Pharmacol Toxicol*. 124: 621–628.

[2] World Health Organization (WHO). (2020). Brief guide to analytical methods for measuring lead in blood. Second Edition. Geneva, Switzerland.

[3] Srinivasa-Reddy, Y., Pullakhandam, R., Radha-Krishna, K.V., Uday-Kumar, P. and Dinesh-Kumar, B. (2011). Lead and essential trace element levels in school children: a

cross-sectional study. *Ann Hum Biol*. 38(3):372-377.

[4] Karrari, P., Mehrpour, O. and Abdollahi, M. (2012). A systematic review on status of lead pollution and toxicity in Iran; guidance for preventive measures. *DARU*. 20(1):2.

[5] Ghaemi, K., Ghoreishi, A., Rabiee, N., Alinejad, S., Farzaneh, E., Amirabadi-Zadeh, A., Abdollahi, M. and Mehrpour, O. (2017). Blood Lead Levels in Asymptomatic Opium Addict Patients; a Case Control Study. *Emerg (Tehran)*. 5(1): e69.

[6] Wani, A.L., Ara, A. and Usmani, J.A. (2015). Lead toxicity: a review. *Interdiscip. Toxicol*. 8(2): 55-64.

[7] Amirabadizadeh, A., Brent, J. and Mehrpour, O. (2019). Impact of chronic lead exposure on liver and kidney function and haematologic parameters. *Basic Clin Pharmacol Toxicol*. 124: 621-628.

[8] Nakhaee, S. and Mehrpour, O. (2018). Opium addiction as new source of lead poisoning: an emerging epidemic in Iran. *EXCLI J*. 17:513.

[9] Fathabadi, B., Dehghanifiroozabadi, M., Aaseth, J., Sharifzadeh, G., Nakhaee, S., Rajabpour-Sanati, A., Amirabadizadeh, A. and Mehrpour, O. (2018). Comparison of Blood Lead Levels in Patients with Alzheimer's Disease and Healthy People. *Am J Alzheimers Dis Other Dement*. 2018: 541–547.

[10] Lanphear, B.P., Hornung, R., Khoury, J., Yolton, K., Baghurst, P., Bellinger, D.C., Canfield, R.L., Dietrich, K.N., Bornschein, R. and Greene, T. (2005). Low-level environmental lead exposure and children's intellectual function: An international pooled analysis. *Environ. Health Perspect*. 113: 894.

[11] Adeyomoye, O.I. and Adewumi, N.A. (2019). Lead Exposure Causes Alteration of Haematological Indices in Adult Female Wistar Rats, *AJPRD*. 7(6):30-34.

[12] Nigra, A.E., Ruiz- Hernandez, A., Redon, J., Navas- Acien, A. and Tellez-Plaza, M. (2016) Environmental metals and cardiovascular disease in adults: a systematic

review beyond lead and cadmium. *Curr Environ Health Rep.* 3(4): 416- 433.

[13] Al-Saady, M.S.M., Kamil, H.Z. and Jasim S.H. (2011). Effect of lead acetate in some physiological genetic parameters in white male rat *rattus rattus*. *Fac. J. Educ. Karbala Univ.* 3 (9): 295-301.

[14] Darwish, W.S., Ikenaka, Y. and Ishizuka, M. (2013). Biological responses of xenobiotic metabolizing enzymes to lead exposure in cultured H4IIE rat cells. *Jpn. J. Vet. Res.* 61: S48–S53.

[15] Ademuyiwa, O., Ugbaja, R. N., Idumebor, F. and Adebawo, O. (2005). Plasma lipid profiles and risk of cardiovascular disease in occupational lead exposure in Abeokuta, Nigeria. *Lipids Health Dis.* 4: 19.

[16] Dongre, N.N., Suryakar, A., Patil, A.J. and Rathi, D. (2010). Occupational lead exposure in automobile workers in North Karnataka (India): effect on liver and kidney functions. *Al Ameen J Med Sci.* 3: 284- 292.

[17] Al-Neamy, F.R., Almehti, A.M., Alwash, R., Pasha, M.A., Ibrahim, A. and Bener, A. (2001). Occupational lead exposure and amino acid profiles and liver function tests in industrial workers. *Int Environ Health Res.* 11: 181–188.

[18] Reitman, S. and Frankel, S. (1957). A colorimetric method for the determination of serum glutamic oxalacetic and glutamic pyruvic transaminases. *Am J Clin Pathol.* 28: 56–63.

[19] Szasz, G. A (1969). Kinetic photometric method for serum γ -glutamyl transpeptidase. *Clin Chem.* 15:124-136.

[20] Garman Society of Clinical Chemistry. (1972). Standardisation of methods for the estimation of enzyme activities in biological fluids. Experimental basis for the optimized standard conditions. *J Clin Chem Clin Biochem.* 10(6):281-291.

[21] Bartels, H., Bohmer M. and Heierli, C. (1972). Serum creatinine determination without protein precipitation. *Clin Chim Acta.* 37: 193-197.

[22] Mackay, E.M. and Mackay, L.L. (1927). The Concentration of Urea in the Blood of Normal Individuals. *J. Clin Invest.* 4(2): 295-306.

[23] Wroblewski, F. and LaDue, J.S. (1955). Lactic dehydrogenase activity in blood. *Proc Soc Exp Biol Med.* 90: 210–13.

[24] Fischer, A.H., Jacobson, K.A., Rose, J. and Zeller, R. (2008). Hematoxylin and eosin staining of tissue and cell sections. *Cold Spring Harb Protoc.* 1: 2008.

[25] Ali, S.A., Al-Derawi, K.A. and Al monsour, N.A.A. (2018). Testicular toxic effect of lead acetate on adult male rats and the potential protective role of alcoholic extract of ginseng (histological, histomorphometrical and physiological). *Sci. J. Med. Res.* 2(6): 87-92.

[26] Nakhaee, S., Amirabadizadeh, A., Brent, J. and Mehrpour, O. (2019). Impact of chronic lead exposure on liver and kidney function and haematologic parameters. *Basic Clin Pharmacol Toxicol.* 124: 621–628.

[27] Singh, R., Neetu, G., Anurag, M. and Rajiv, G. (2011). Heavy metals and living systems: an overview, *Indian J. Pharmacol.* 43(3): 246–253.

[28] Dwivedi, K. and Gupta, D.K (2020). Concomitant influence of heavy metal intoxication on size of organs and body weight in albino rats, *IJPSR.* 11(3): 1417–1424.

[29] Owonikoko, M.W., Emikpe, B.O. and Olaleye, S.B. (2021). Standardized experimental model for cement dust exposure; tissue heavy metal bioaccumulation and pulmonary pathological changes in rats. *Toxicol. Rep.* 8: 169–1178.

[30] Miller G.D. (2017). Appetite Regulation: Hormones, Peptides, and Neurotransmitters and Their Role in Obesity. *Am. J. Lifestyle Med.* 13(6): 586–601.

[31] Ying, P., Yumen, L. and Xiaodong, X. (2013). Change trends of organ weight background data in *Sprague dawley* rats at different ages. *J. Toxicol. Pathol.* 26(1): 29–34.

- [32] Tritscher, A., Walker, R. and Younes, M. (2002). Hazard characterization of chemicals in food and diet: dose response, mechanism and extrapolation issues. *Food Chem. Toxicol.* 42: 237–282.
- [33] Fatoki, J. O., Adeleke, G. E., Badmus, J. A., Afolabi, O. K., Adedosu, O. T., Kehinde, S. A. and Adekunle A. S. (2019). Time – Course of Sodium Arsenate Induced Hepatotoxicity and Nephrotoxicity in Male Wistar Rats. *J. Nat. Sci. Res.* 9(4): 78 – 91.
- [34] Fatoki, J. O., Badmus, J. A., Adedosu, O. T., Adegoke, A.A., Lawal, O. R., Ogunmola, I. A. and Omotayo T. M. (2018). Repeated Exposure of Herbal Concoction Imprints Toxicological Implications through Redox Imbalance in Male Wistar Rats. *Int. J. Pharm. Res.* 23(2): 1-18.
- [35] Mauro, P., Renze, B. and Wouter W. (2006). Enzymes. In: Tietz text book of clinical chemistry and molecular diagnostics. Carl, A.B, Edward, R. and David, E.B. 4th ed. Elsevier pp 604-616.
- [36] Shekari, M., Sendi, J.J., Etebari, K., Ziaee, A. and Shadparvar, A. (2008). Effects of *Artemisia annua* L. (Asteracea) on Nutritional Physiology and Enzyme Activities of Elm Leaf Beetle, *Xanthogaleruca luteola* Mull. (Coleoptera: Chrysomellidae). *Pestic Biochem Phys.* 91: 66-74.
- [37] Diamantino, T.C., Almeida, E., Soares, A.M. and Guilhermino, L. (2001). Lactate Dehydrogenase Activity as an Effect Criterion in Toxicity Tests with *Daphnia magna* Straus. *Chemosphere.* 45: 553-560.
- [38] Riseh, N.S., Ghadamyari, M. and Motamediniya, B. (2012). Biochemical Characterization of α - and β -Glucosidases and α - and β -Galactosidases from Red Palm Weevil, *Rhynchophorus ferrugineus* Olivieri (Col.: Curculionidae). *Plant Prot. Sci.* 2: 85-93.
- [39] Marenkov, O., Prychepa, M. and Kovalchuk, J. (2018). The Influence of Heavy Metal Ions on the Viability and Metabolic Enzyme Activity of the Marbled Crayfish *Procambarus virginalis* (Lyko, 2017). *Int. Lett. Nat. Sci.* 70:11-23.
- [40] Lanning, L.L., Creasy, D.M., Chapin, R.E., Mann, P.C., Barlow, N.J., Regan, K.S. and Goodman, D.G. (2002). Recommended approaches for the evaluation of testicular and epididymal toxicity, *Toxicol. Pathol.* 30(2002): 507–520.

<https://fjpas.fuoye.edu.ng>