Sahel Journal of Life Sciences FUDMA (SAJOLS)
September 2024 Vol. 2(3): 83-97
ISSN: 3027-0456 (Print)
ISSN: 1595-5915(Online)
DOI: https://doi.org/10.33003/sajols-2024-0203-12

Research Article

Sub-chronic Toxicological Evaluation of Drinking Water from Internally Displaced Persons (IDP) Camp in Wassa, Federal Capital Territory, Abuja, Nigeria

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ABSTRACT
The number of Internally Displaced Persons (IDPs) camps has increased recently in Nigeria due to insurgency and natural disasters such as floods. This study aimed to evaluate the sub-chronic toxicological effects of the source of the drinking water in Wassa camp, Abuja, Nigeria. Forty-eight (48) mice were randomized into four (4) groups of twelve (12) mice each and orally administered different concentrations (5, 10, and 20 mL/kg body weight respectively) of the water sample from Wassa IDP for fourteen (14) days. Alteration was noticed in the liver and kidney indices of mice by a significant increase ($p<0.05$) in kidney-body weight ratio, kidney, liver and plasma protein concentrations, and considerable decrease ($p<0.05$) in albumin, creatinine, uric acid and total bilirubin concentrations, 14-days post-treatment, particularly at a higher dose. Gamma-glutamyl transaminase in the liver and aspartate transaminase in both liver and kidney significantly ($p<0.05$) increased in mice with a resultant significant decrease ($p<0.05$) in the serum, 14 days post-treatment at 20 mL/kg bwt. There was no significant difference ($p>0.05$) observed in serum triglycerides, very-low-density-lipoprotein cholesterol concentrations at all doses 14-days post-treatment compared with the control but the concentrations of serum low-density-lipoprotein cholesterol and total cholesterol significantly reduced ($p<0.05$) at 20 mL/kg bwt 14-days post-treatment. The drinking water altered some liver-function indices while the cardiac-function indices were less affected. Therefore, prolonged consumption of the water may not be relatively safe. The provision of clean drinking water for the Wassa IDP camp will prevent adverse biochemical reactions from prolonged water consumption.

Keywords: Water; Internally Displaced Persons (IDPs); Toxicity; Liver and kidney function; Lipid profile; Wassa

Citation: Adamu, S. U., Nasir, S., Saliu, O. A., Idowu, A. O., Edogbo, B. E., Yisa, A. G., Sada, S. A., Ebhodaghe, F. E., Eya, C., Adams, M. D., & Tijjani, H. (2024). Sub-chronic Toxicological Evaluation of Drinking Water from Internally Displaced Persons (IDP) Camp in Wassa, Federal Capital Territory, Abuja, Nigeria. *Sahel Journal of Life Sciences FUDMA,* 2(3): 83-97. DOI: https://doi.org/10.33003/sajols-2024-0203-12

INTRODUCTION

Accessible and portable drinking water is crucial to the sustenance of any community (Ongeri, 2023). The need for portable and accessible drinking water is on the increase due to population explosion, shortage of government interventions coupled with the contamination of the aquatic resources with contaminants such as personal care products, pesticides, herbicides, and heavy metals, amongst a myriad of biological contaminants (Whelan *et al*., 2022). Access to clean water is crucial for maintaining health and well-being (Gao *et al*., 2023). Communities with

abundant and safe water sources tend to have better public health outcomes and a higher quality of life, while populations without access to clean and safe water sources face serious health risks from several waterborne diseases caused by poor water quality (Gao *et al*., 2023). Recent study of Adamu *et al*. (2024) reported a high prevailing rate of waterborne diseases in major health facilities in Gwagawalada Area Council of Abuja, Nigeria, due to contamination of water.

The growing number of internally displaced persons (IDP) and camps have been on the increase in recent years in Nigeria. The boomerang effect of internal crises such as flood and security challenges ranging from Boko Haram, Banditry, and Fulani Herdsmen clashes resulted in the displacement of many communities. Many IDP camps in Nigeria including Wassa IDP experiencing various forms of neglection, which is reflected in their lack of portable water, the inadequacy of toilets, among others. IDPs in several states around the nation frequently lament that the federal and state administrations have abandoned them. Water sources in Wassa IDP have been found to pose health threats due to contamination with cadmium, lead and bacteria (Saliu *et al*., 2024).

The availability of portable, affordable and contaminated free water for drinking and domestic activities is a serious limitation in most internally displaced persons (IDPs) camps in Nigeria. The Sustainable Development Goal (SDG) Target 6.1 calls for universal and equitable access to safe and affordable drinking water for all (UNICEF, 2024). The target is tracked with the indicator of "safely managed drinking-water services"- drinking water from an improved water source that is located on-premises, available when needed, and free from faecal and priority chemical contamination. This study aimed to evaluate the sub-chronic toxicological effects of the source of the drinking water in Wassa camp, Federal Capital Territory (FCT), Abuja, Nigeria.

MATERIALS AND METHODS

Reagents and Assay Kits

Assay kits for the determination of cholesterols, triglycerides (TGs), high density lipoprotein cholesterol (HDL)-cholesterol were obtained from Spectrum MDSS GmbH, Schiffgraben, Germany. Low density lipoprotein cholesterol (LDL) was a product from Centronic GmbH, Germany. Albumin, Gamma glutamyl transferase (GGT), bilirubin, uric acid, creatinine, aspartate aminotransferase (GOT) and alkaline phosphatase **(**ALP) were obtained from AGAPPE Diagnostics, GmbH, Switzerland. All other reagents used were of analytical grade.

Experimental Animals

Adult Swiss albino mice (*Mus musculus*) of average weight of 29.70±5.14 g were obtained from the Animal Breeding Unit of the University of Jos, Plateau State, Nigeria. The animals were housed in standard plastic cages and acclimatized for two weeks. They were maintained under standard conditions with free access to feed and water.

Description of the Study Area and Sample Collection

Wassa IDP camp is situated about 5 km away from the city centre behind Apo Village in Abuja Municipal Area Council (AMAC), Abuja, Nigeria (Figure 1). It consists of 7 communities, with a population of about 5121 IDPs (Sampson *et al*., 2023). Wassa IDP camp was purposively selected due to its population and the difficulty faced by the residents in accessing portable drinking water. The study locations are characterized by open defecation and noticeable anthropogenic features such as agricultural activities, washing and swimming. Water samples were collected from the study location in sterile containers and appropriately before usage.

Ethical Clearance

Ethical clearance for the *in vivo* studies was obtained from the National Open University of Nigeria, Abuja, Nigeria.

Animal Groping and Administration of water sample

Adult Swiss mice (48) were randomly distributed into four groups of twelve mice per group and daily administered various doses in mL/kg body weight (bwt) of water samples by the oral route for 14 days as follows.

Group A – Distilled water/Control
Group B – Water Sample at 5 mL/kg bwt
Group C – Water Sample at 10 mL/kg bwt
Group D – Water Sample at 20 mL/kg bwt

Sample Collection and Preparation

At the 24th hour after the first administration period, three mice (each from a group) were slightly anesthetized with diethyl ether, and the neck area quickly cleared of fur and skin to expose the jugular veins. Venous blood was then be collected into EDTA sample. The EDTA blood sample was centrifuged at 1000 rpm for 5 min and the plasma pipetted out. This was stored in a refrigerator for subsequent analysis.

The animals were also quickly dissected; the liver, and kidney were excised from each animal, cleaned of blood, weighed, and then homogenized in ice-cold 0.25M sucrose solution (1:5 w/v). The homogenate were centrifuged at 3000 rpm for 15 min to obtain the supernatant used in the study. The other mice were sacrificed in batch on the 4th, 8th and 14th day respectively.

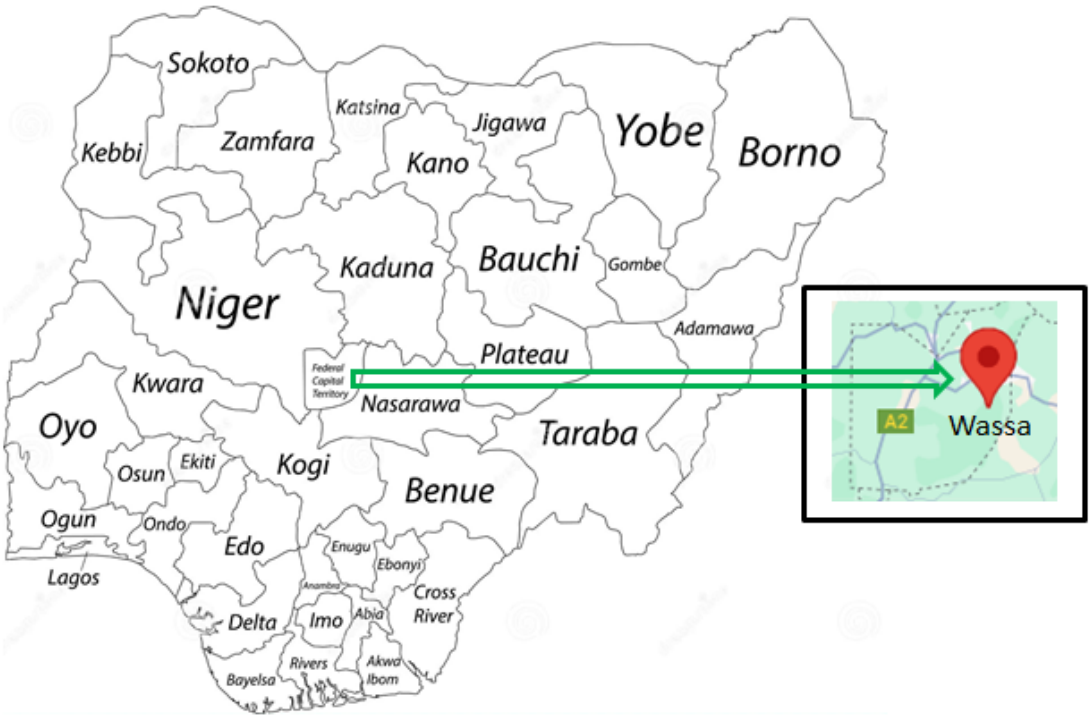

Figure 1: Wassa IDP Camp, Abuja, Nigeria

Organ-Body Weight Ratio

The organ-body weight ratio was calculated using the formula $\frac{weight\ of\ organ}{weight\ of\ whole\ animal} \times 100$

Experimental Assays

Lipid Profile and Cardiovascular Indices

Total cholesterol (Tchol), triglycerides (TGs), and high density lipoprotein cholesterol (HDL-Chol.) were determined according to the method described by Tietz (1995) and Friedwald *et al*. (1972). Very Low Density Lipoprotein Cholesterol (VLDL-chol.) and Low Density Lipoprotein Cholesterol (LDL-chol.) were determined using the formula described by Friedwald *et al.* (1972). Atherogenic index (AI) was calculated using the formula described by Lamarche *et al*. (1996), while cardiac index and coronary artery index were determined according to the formula described by Kang *et al*. (2004).

Estimation of Markers of Liver Function

Total protein concentration was estimated using the method described by Gornall *et al.* (1949). The method described by Reitman and Frankel (1957) was used to determine the activities of aspartate aminotransferase (GOT) in plasma and liver homogenates while the method described by Wright *et al.* (1972) was used to determine alkaline phosphatase **(**ALP) activity in plasma and liver.

Estimation of Markers of Kidney Function

Gamma-glutamyl transferase (GGT), albumin, creatinine, uric acid and bilirubin were estimated using the methods described by Szasz (1969), Doumas *et al*. (1971), Cheesbrough (2004), Fossati *et al*. (1980), and Walter and Gerard (1970) respectively.

Data Analysis

The data was subjected to a one-way analysis of variance to compare the mean differences. Where a significant difference exists, Dunnett's post hoc test was employed to separate the means. Experimental data were expressed as mean ± standard deviation (SD) and plotted using GraphPad Prism 6 software (GraphPad Software, California, USA).

RESULTS

A significant ($p<0.05$) increase in body weight ratio was observed in mice treated with water samples at 5 mL/kg body weight compared with the control group (Figure 2). However, no significant ($p>0.05$) difference between the control group and mice treated with 20 mL/kg body weight. There was dose-dependent increase in liver-body weight ratios on day 8 and day 14 compared with the control group (Figure 3). No significant ($p>0.05$) differences were observed in kidney body weight ratio on day 8 and day 14 consecutive treatments with IDP water sources (Figure 4).

Plasma protein significantly increased ($p<0.05$) on days 2, 4, and 14 at 20 mL/kg body weight compared with control (Table 1). Significant increases ($p<0.05$) were also observed in kidney protein on day 14 at 5, 10 and 20 mL/kg body weight compared with the control, while no significant differences ($p>0.05$) were observed in

liver protein at 10 and 20 mL/kg body weight compared with control on day 8 and 14 (Table 1).

No significant difference ($p>0.05$) in GGT at day 2, 8 and 14 treatments (Figure 5) compared with the control treatments. However, a significant ($p<0.05$) decrease was observed at day 4 treatments with IDP water compared with the control group (Figure 5).

For albumin, no significant ($p>0.05$) decreases were observed at day 8 and 14, significant ($p<0.05$) increase at day 2 post treatments (Figure 6). Creatinine level was significantly ($p<0.05$) decrease at day 4, while no significant ($p>0.05$) difference on day 4 at 10 and 20 mL/kg body weight (Figure 7). No significant ($p>0.05$) difference were observed in uric acid levels (Figure 8), and in ALP at 5 and 20 mL/kg body weight at day 2 (Table 2). For plasma ALP, increase were observed at 5 and 20 mL/kg body weight while in liver ALP a dose dependent increase were observed on day 8, with no significant ($p>0.05$) different on day 14 post treatment with IDP water sample (Table 2).

No significant ($p>0.05$) differences were observed in triglycerides, total cholesterol, VLDL, and LDL in all treatments compared with the control except on day 8 at 10 mL/kg body weight treatment (Figure 9, 10, 11, 12). No significant difference ($p>0.05$) was observed in HDL cholesterol after 14 days of treatment (Figure 13). More so, no significant ($p>0.05$) differences were observed in AI, CI and CAI indices in all doses at day 4, 8, and 14 respectively (Figure 14, 15, 16).

Significant ($p<0.05$) increases were observed in plasma and kidney GOT at 5 and 20 mL/kg bodyweight on day 14, with no corresponding increase in liver GOT at the same period (Table 3). Results for total bilirubin were not significantly ($p>0.05$) different at day 4, 8, and 14 while a significant ($p<0.05$) decrease was observed in direct bilirubin at 5 mL/kg body weight at day 14 (Table 4).

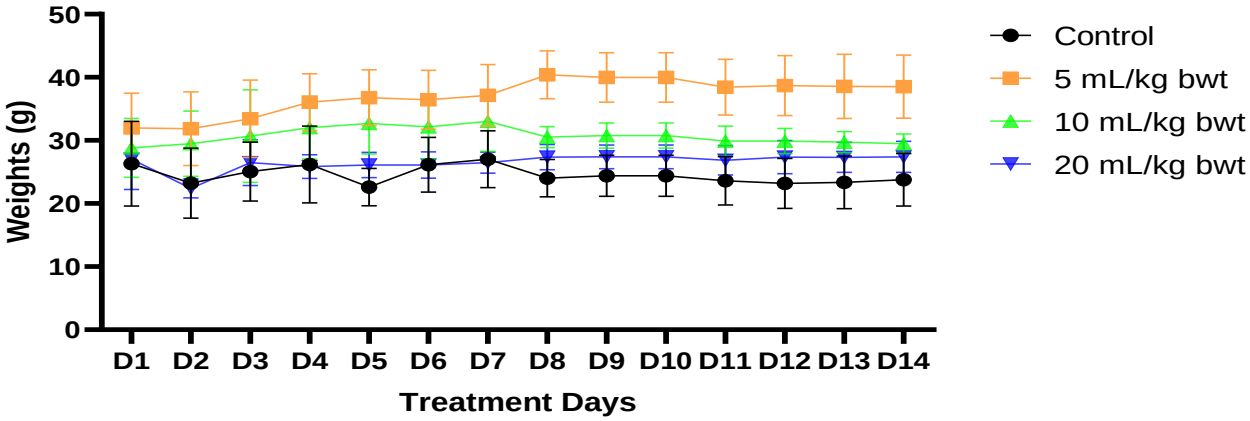

Figure 2. Body weight changes following the administration of IDP water samples to experimental mice. Values are mean ± standard deviation (SD). bwt = body weight

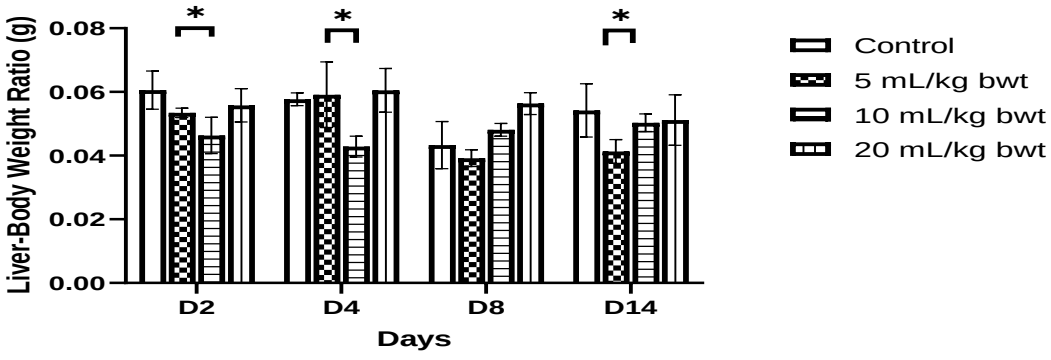

Figure 3. Liver-body weight ratio following the administration of IDP water samples to experimental mice. Values are mean ± standard deviation (SD). bwt = body weight. * Significant difference compared with control ($p<0.05$)

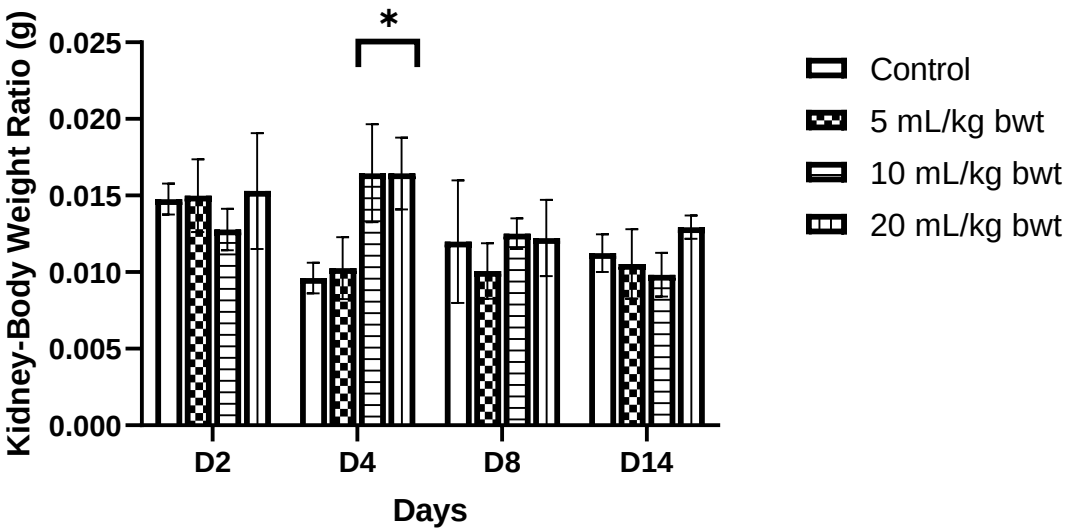

Figure 4. Kidney-body weight ratio following the administration of IDP water samples to experimental mice. Values are mean ± standard deviation (SD). bwt = body weight. * Significant difference compared with control (p<0.05)

Table 1. Total protein concentration following the administration of IDP water samples to experimental mice

Conc. (mL/ bwt)	**Day 2**	**Day 4**	**Day 8**	**Day 14**
pProtein				
Control	450.68±21.21	234.66±14.14	363.64±59.92	241.78±14.14
5	503.09±136.75	397.12±16.18*	347.20±28.28	412.84±48.42*
10	352.63±13.18*	365.76±102.47*	263.81±14.14*	277.46±21.21
20	558.60±63.40	360.25±41.95*	464.66±14.14	548.01±80.00*
kProtein				
Control	923.39±137.23	1010.42±14.14	940.08±54.14	393.73±66.52
5	1158.60±64.08*	1083.98±183.37	661.10±51.98*	921.31±23.51*
10	1406.91±239.03	979.75±164.19	916.36±54.14	760.30±61.07*
20	1370.47±468.88	1262.84±80.09*	515.25±27.07*	678.39±28.28*
LProtein				
Control	646.10±21.21	724.07±21.21	866.19±122.25	742.88±243.89
5	727.20±233.70	1144.28±124.56*	1198.01±154.86*	1192.92±130.55*
10	1412.03±128.69*	769.83±21.21	973.31±28.28	1009.87±22.17
20	812.80±28.76*	1370.04±336.83*	896.27±29.36	1020.89±147.73

pProtein = plasma, lProtein = liver, kProtein = kidney. Values are mean ± standard deviation (SD). bwt = body weight. *Significant difference at p<0.05

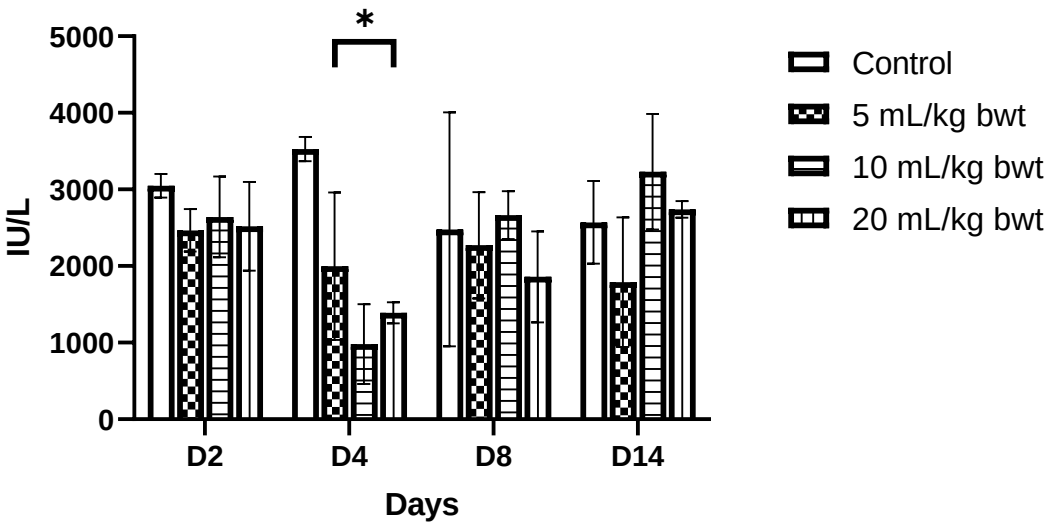

Figure 5. Gamma-glutamyl transferase activities following the administration of IDP water samples to experimental mice. Values are mean ± standard deviation (SD). bwt = body weight. * Significant difference compared with control (p<0.05)

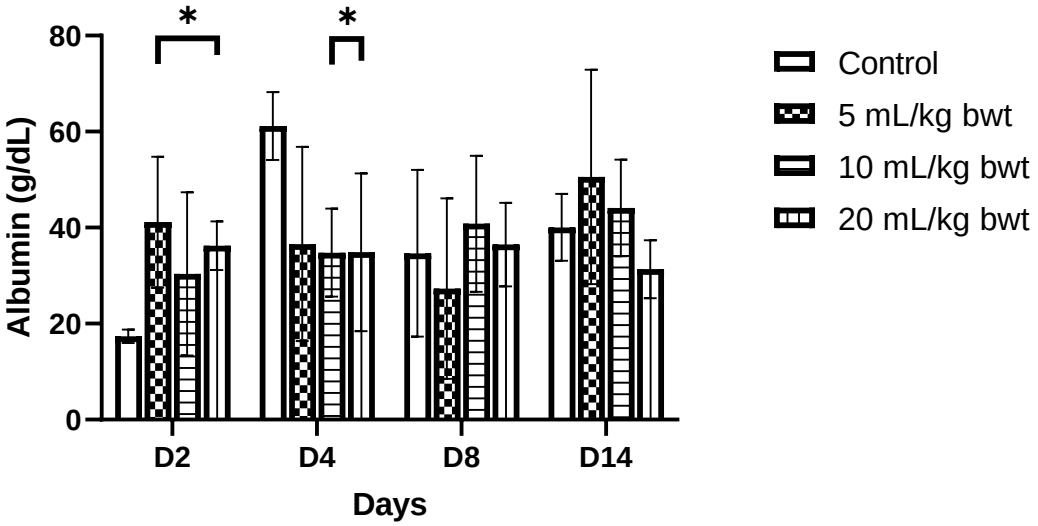

Figure 6. Albumin concentration following the administration of IDP water samples to experimental mice. Values are mean ± standard deviation (SD). bwt = body weight. * Significant difference compared with control (p<0.05)

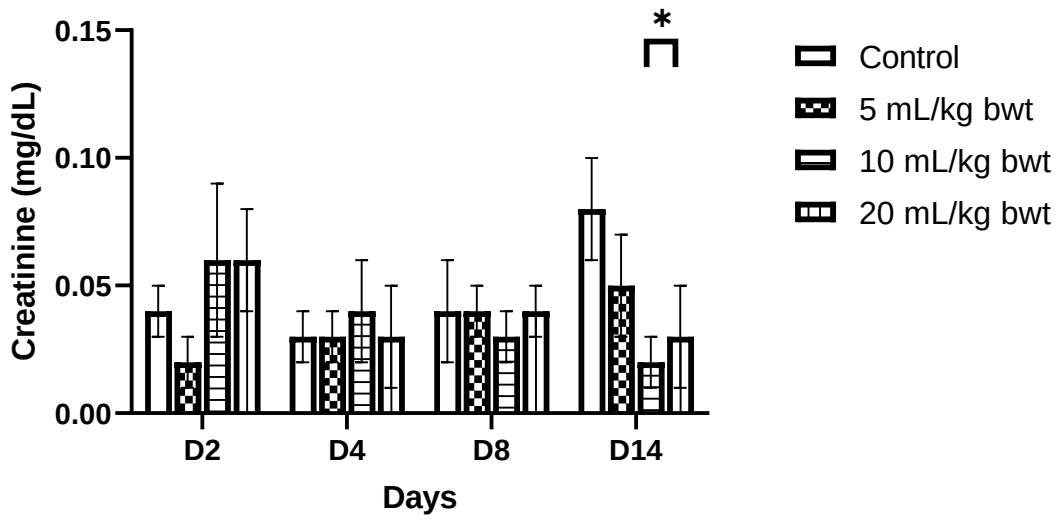

Figure 7. Creatinine concentration following the administration of IDP water samples to experimental mice. Values are mean ± standard deviation (SD). bwt = body weight. * Significant difference compared with control (p<0.05)

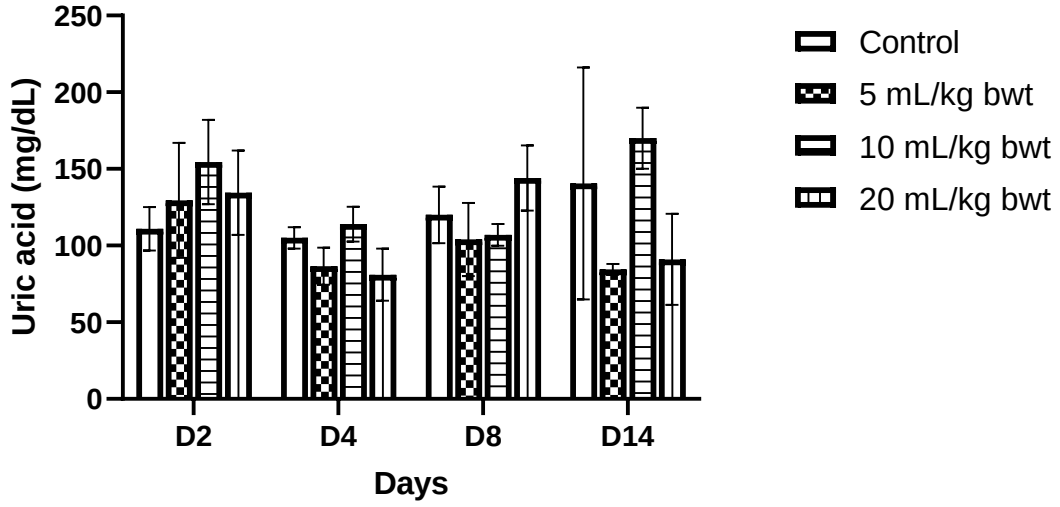

Figure 8. Uric acid concentration following the administration of IDP water samples to experimental mice. Values are mean ± standard deviation (SD). bwt = body weight

Table 2. Alkaline phosphatase activities following the administration of IDP water samples to experimental mice

Conc. (mL/ bwt)	Day 2	Day 4	Day 8	Day 14
Plasma ALP				
Control	168.00±81.21	415.00±114.14	151.50±74.66	138.00±55.56
5	426.50±125.38*	196.50±19.09*	165.00±39.60	344.00±130.93*
10	206.50±19.09	504.00±119.80	520.00±121.21*	123.50±58.69
20	412.50±150.02*	196.50±46.87*	138.00±77.78	467.50±122.96*
Liver ALP				
Control	646.10±21.21	724.07±21.21	866.19±122.25	742.88±243.89
5	727.20±233.70*	1144.28±124.56	1198.01±154.86*	1192.92±130.55
10	1412.03±128.69*	769.83±21.21	973.31±28.28	1009.87±22.17
20	812.80±28.76*	1370.04±336.83*	896.27±29.36	1020.89±147.73

Values are mean ± standard deviation (SD). bwt = body weight. *Significant difference at p<0.05

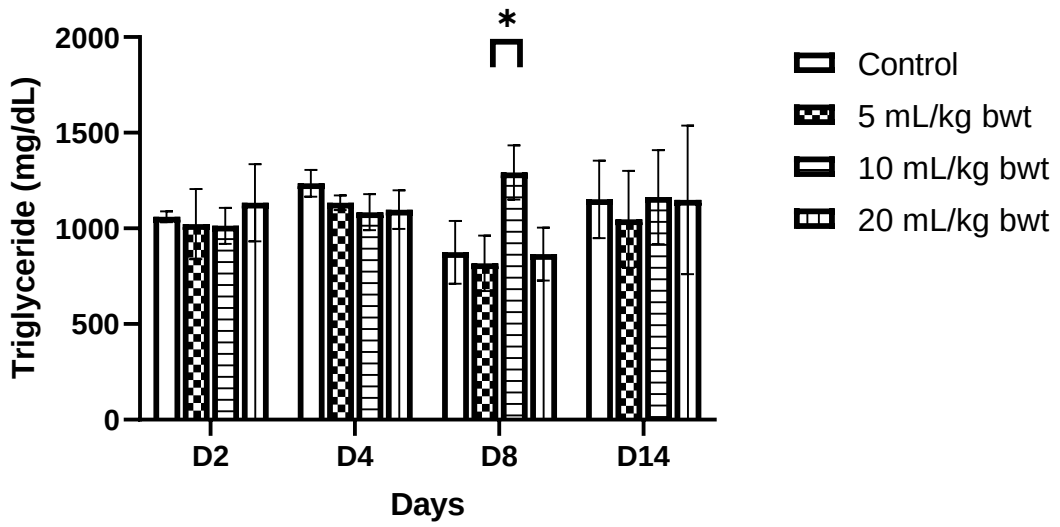

Figure 9. Triglyceride concentration following the administration of IDP water samples to experimental mice. Values are mean ± standard deviation (SD). bwt = body weight. * Significant difference compared with control ($p<0.05$)

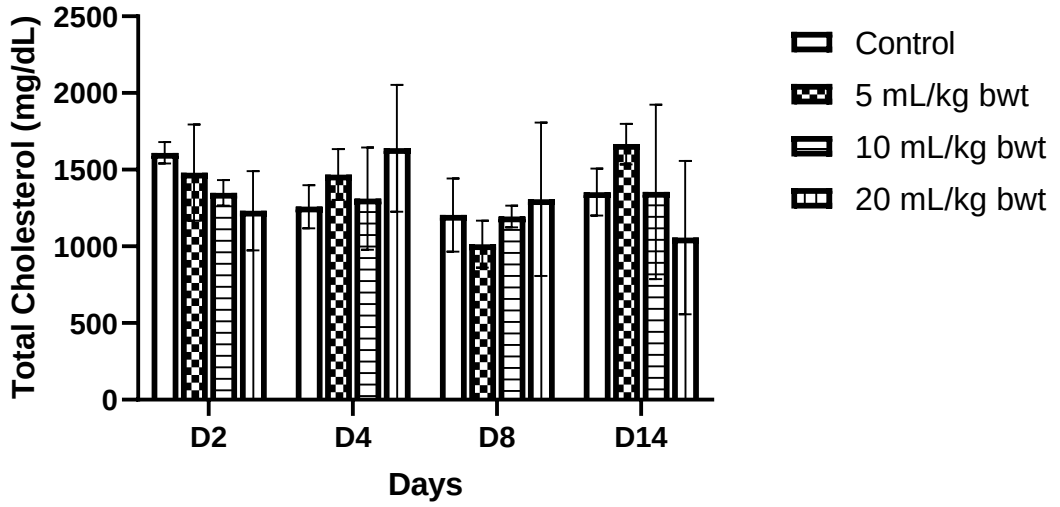

Figure 10. Total cholesterol concentration following the administration of IDP water samples to experimental mice. Values are mean ± standard deviation (SD). bwt = body weight

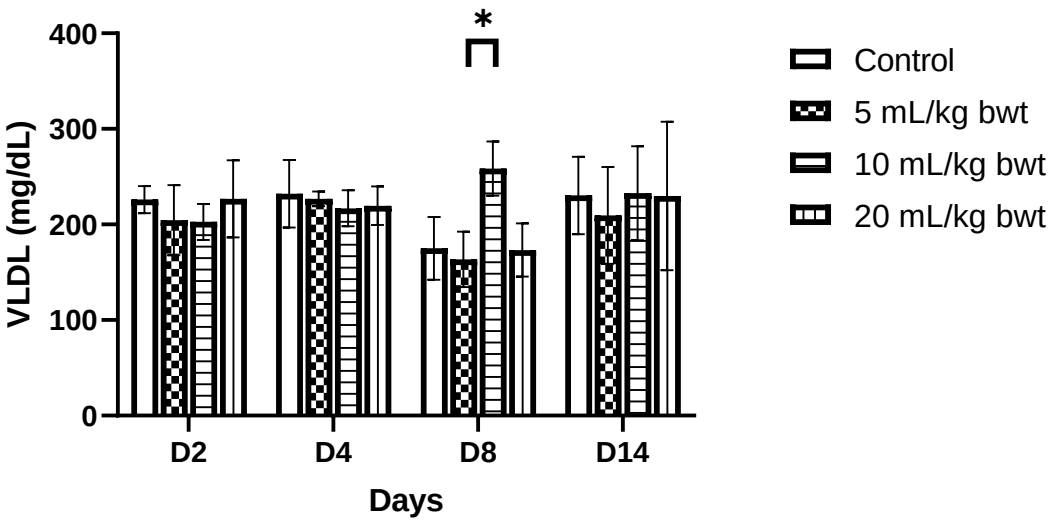

Figure 11. Very low density-lipoprotein concentration following the administration of IDP water samples to experimental mice. Values are mean ± standard deviation (SD). bwt = body weight. * Significant difference compared with control (p<0.05)

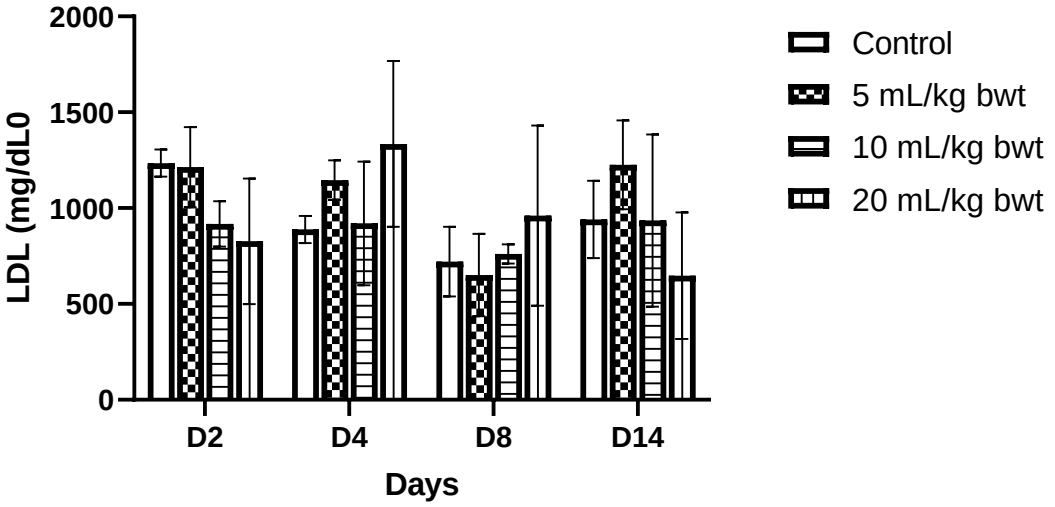

Figure 12. Low density-lipoprotein concentration following the administration of IDP water samples to experimental mice. Values are mean ± standard deviation (SD). bwt = body weight

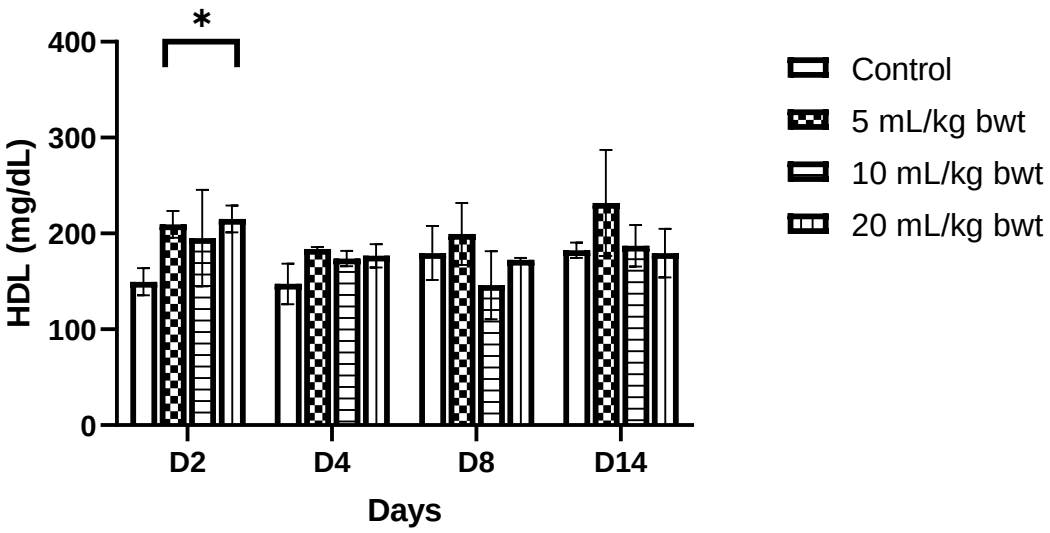

Figure 13. High density-lipoprotein concentration following the administration of IDP water samples to experimental mice. Values are mean ± standard deviation (SD). bwt = body weight. * Significant difference compared with control (p<0.05)

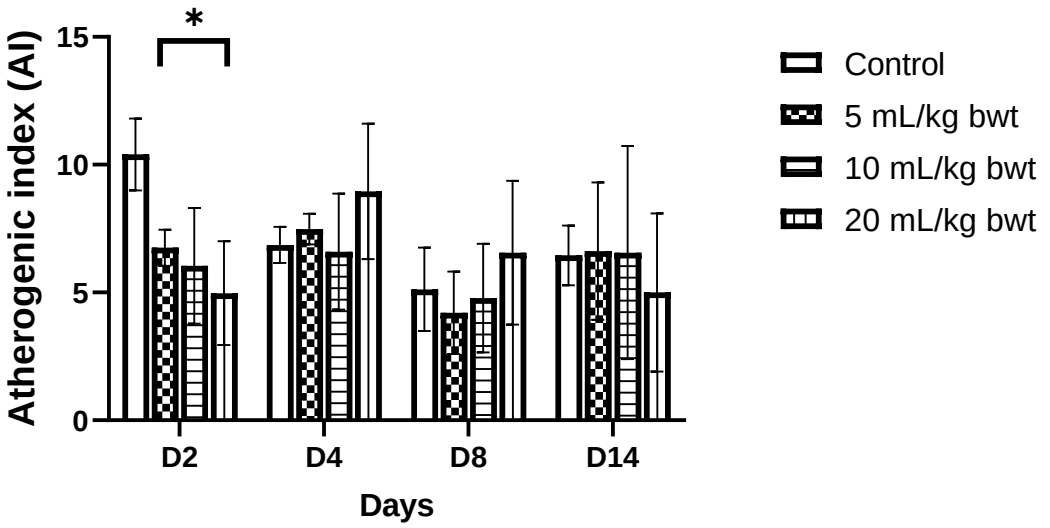

Figure 14. Atherogenic index following the administration of IDP water samples to experimental mice. Values are mean ± standard deviation (SD). bwt = body weight. * Significant difference compared with control (p<0.05)

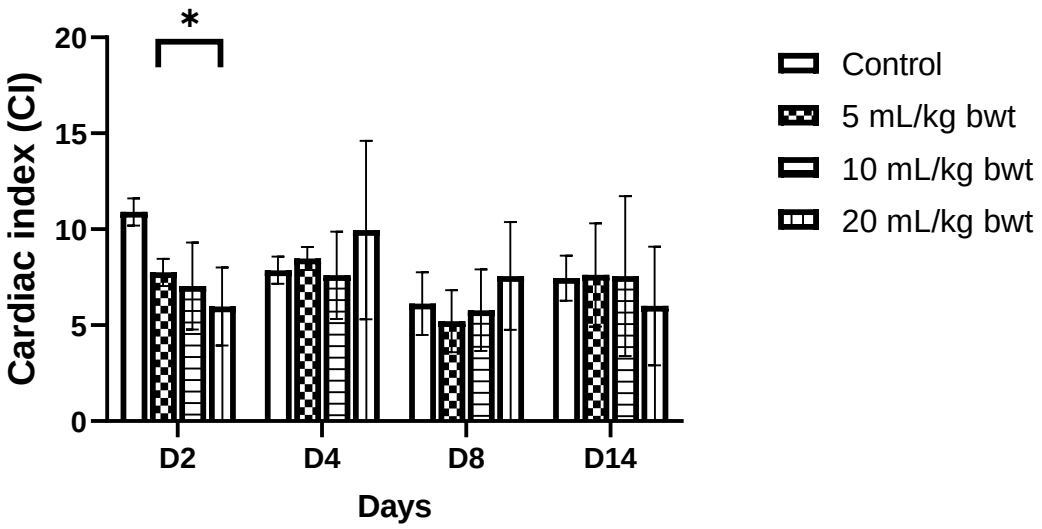

Figure 15. Cardiac index following the administration of IDP water samples to experimental mice. Values are mean ± standard deviation (SD). bwt = body weight. * Significant difference compared with control (p<0.05)

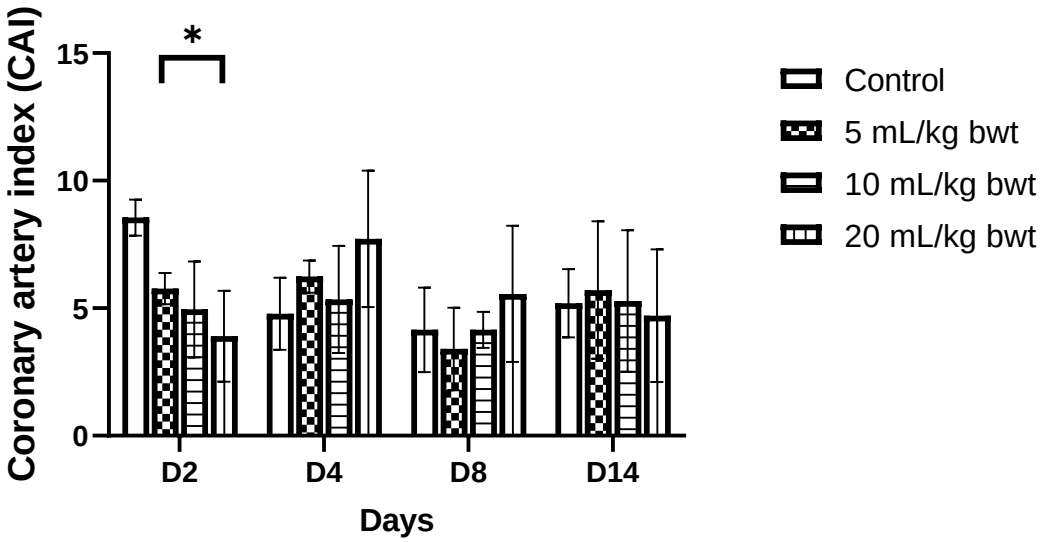

Figure 16. Coronary artery index following the administration of IDP water samples to experimental mice. Values are mean ± standard deviation (SD). bwt = body weight. * Significant difference compared with control (p<0.05)

Table 3. Glutamic oxaloacetic transaminase activities following the administration of IDP water samples to experimental mice

Conc. (mL/ bwt)	Day 2	Day 4	Day 8	Day 14
Pgot				
Control	6451.00±353.55	2144.00±70.71	384.00±73.54	122.00±0.00
5	515.00±234.76	363.00±200.94*	140.00±49.50*	244.00±177.11*
10	1652.00±1448.87	812.50±60.10	125.00±21.21*	145.33±20.21
20	201.00±86.27*	567.00±480.83	314.50±24.75	332.00±177.11*
lGOT				
Control	112.00±14.14	110.00±7.07	105.00±0.00	198.50±89.80
5	78.50±61.52*	977.00±864.08*	1117.00±1258.65*	240.00±102.21
10	78.50±12.02*	131.00±12.73	172.00±28.28	145.67±102.26
20	148.50±61.52	610.50±789.84	235.50±111.02	232.67±89.87
kGOT				
Control	533.00±35.36	30.00±7.07	87.50±24.75	43.50±12.02
5	75.33±24.75*	57.00±31.11	96.00±12.73	159.33±37.90*
10	198.00±98.99*	113.50±12.02*	47.00±7.07*	219.33±133.47*
20	93.00±74.25*	131.00±12.73*	26.00±12.73*	58.33±40.41

GOT = Glutamic oxaloacetic transaminase, pGOT = plasma, lGOT = liver, kGOT = kidney. Values are mean ± standard deviation (SD). bwt = body weight. *Significant difference at $p<0.05$

Table 4. Total and direct bilirubin concentrations following the administration of IDP water samples to experimental mice

Conc. (mL/ bwt)	Day 2	Day 4	Day 8	Day 14
Total Bilirubin				
Control	246.24±15.28	417.63±15.28	178.39±81.28	242.69±91.99
5	159.68±35.37*	325.27±47.25	360.22±128.12	144.62±38.69
10	256.34±22.33	340.75±51.55	151.29±50.00	115.59±34.64
20	126.88±26.07*	423.66±72.73	243.55±72.92	194.62±56.57
Direct Bilirubin				
Control	25.05±1.53	16.71±2.08	16.35±5.52	11.12±1.02
5	12.08±2.17*	17.09±2.74	15.45±1.14	5.20±2.92*
10	14.09±1.21*	6.96±2.75*	14.08±1.00	19.17±4.57*
20	12.85±2.63*	10.03±1.88*	15.32±2.32	13.57±2.95

Values are mean ± standard deviation (SD). bwt = body weight. *Significant difference at $p<0.05$

DISCUSSION

The toxicological evaluation of water samples from the WASSA IDP camp using liver, kidney, and plasma indices in mice has revealed several noteworthy findings. The observed increase in body weight ratio at a lower dose (5 mL/kg body weight) compared to the control group is an intriguing result that warrants further investigation. Low-dose exposure to contaminants may lead to metabolic alterations that are not evident at higher doses (Prins, 2021). The lack of significant body weight change at the higher dose (20 mL/kg) could be attributed to a toxic compensatory mechanism that is yet to be understood.

The dose-dependent increase in liver-body weight ratios on days 8 and 14 suggests hepatomegaly, which is often associated with exposure to hepatic toxins and lipid accumulation (Jiao *et al*., 2020). This finding is consistent with the liver's role as a primary detoxifying organ, responding to xenobiotic stress through hypertrophy and hyperplasia, and the observed hepatomegaly maybe reversed through withdrawal of the toxin source (Mirone *et al*., 2018). In contrast, the absence of significant changes in kidney-body weight ratios indicates that the renal system may not be as directly affected by the toxicants in the water, or that the kidney's compensatory mechanisms are effectively mitigating any weight changes. Drug-induced nephrotoxicity are related with acute renal damage and chronic kidney diseases, which may require other enzyme diagnosis to confirm (Kim and Moon, 2012).

The significant increase in plasma protein levels at the highest dose (20 mL/kg) on multiple days suggests an acute phase response, potentially indicative of inflammation or stress. This may result from pro-

inflammation state, which may how ever be related to chronic or age-related stimulation of macrophages (Papet *et al*., 2003). The concomitant increase in kidney protein on day 14 across all doses could imply renal stress or damage, as proteinuria is a common marker of nephrotoxicity (Fassett *et al*., 2011). However, its limitations as a biomarker of chronic kidney disease progression and response to interventions are limited (Levey *et al*., 2009). Interestingly, liver protein levels did not significantly change, which could suggest that the liver's synthetic capacity is maintained or that the hepatic response is more complex than can't be assessed by total protein levels alone. The significant decrease in GGT at day 4, an enzyme typically associated with hepatocellular damage (Xing *et al*., 2022), could reflect an adaptive enzymatic response to the initial exposure. Higher levels of GGT indicate damage to the liver (Xing *et al*., 2022).

Serum albumin is produced by the liver and is an abundant plasma protein in blood circulation. Plasma albumin is important in the maintenance of plasma colloid osmotic pressure, regulating immune responses, scavenging free radicals, and transporting endogenous compounds (Miller and Jedrzejczak, 2001). The significant increase in plasma albumin on day 2, followed by no significant decreases, suggests an early reaction to the water sample exposure, which may stabilize over time.

The function of the kidney includes the integration of responses to stress, cardiovascular control, electrolytes and water regulations. Prediction of kidney diseases can be determined using renal function status through the flow rate of filtered fluid through the kidneys. Methods used in glomerular filtration rate to assess kidney diseases are time-consuming, invasive and expensive compared with the use of alternative biomarkers such as creatinine, which is found in serum and urine (Gowda *et al*., 2010). The decrease in creatinine levels at day 4 could indicate altered glomerular filtration or muscle catabolism.

Bilirubin is a product of heme catabolism and a fundamental biomolecule with antioxidant and anti-inflammatory functions in serum (Otero Regino *et al*., 2009). Bilirubin plays a critical role in the neutralization of free radicals linked with peroxidation of lipids. Similarly, they protect the cardiovascular, hepatobiliary system and immune systems (Otero Regino *et al*., 2009). The stable total bilirubin levels and the significant decrease in direct bilirubin at 5 mL/kg body weight at day 14 could indicate an alteration in hepatic uptake, conjugation, or secretion of bilirubin.

Furthermore, the study observed increase in plasma ALP at certain doses and a dose-dependent increase in liver ALP on day 8, without a significant difference on day 14. ALP is one of the main groups of phosphatases recommended for use as indicators of hepatobiliary diseases and hepatic damages (Renner and Dällenbach, 1992; Harrison *et al*., 1999). The significant increase in plasma and kidney GOT at 5 and 20 mL/kg body weight on day 14, without a corresponding increase in liver GOT could suggest extracellular release of the enzyme from the kidney rather than the liver.

Low density lipoprotein cholesterol (LDL), triglyceride (TG) and high density lipoprotein cholesterol (HDL) are related markers for cardiovascular diseases. For instance, lowering LDL reduces the risk for cardiovascular events (Linton *et al*., 2019). The stability of lipid profiles and atherogenic indices in most treatments suggests that the water contaminants may not significantly disrupt lipid metabolism or contribute to cardiovascular risk.

CONCLUSION

In conclusion, the water samples from the WASSA IDP camp have variable effects on liver, kidney, and plasma indices in mice, suggesting the presence of contaminants that can induce both organ-specific and systemic physiological responses. These findings highlight the importance of identifying the toxic constituents within the water and addressing the potential health risks for the camp's inhabitants. The studied source of drinking water altered some liver-function indices while the cardiac-function indices were less affected. Therefore, prolong consumption of the water may not be relatively safe. Further studies are required to elucidate the mechanisms underlying the observed toxic effects and to implement appropriate interventions to ensure the provision of safe drinking water.

Acknowledgment: Suwaiba Nasir and Shehu Usman Adamu appreciate the funding from National Open University of Nigeria, Abuja, Nigeria and TetFund Nigeria Institution Based Research (IBR) grant.

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