

Effects of Ethanol Seed Extract of *Tetracarpidium Conophorum* in the Treatment of Mercury Chloride-Induced Cardiac and Hepatic Toxicity

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Abstract

Mercury chloride ($HgCl_2$) is a compound of mercury, which is highly toxic and corrosive. Mercury chloride was formerly utilized as a treatment for syphilis; however, it is no longer employed for medical purposes due to its toxicity. A chemical evaluation of *Tetracarpidium conophorum* nuts revealed it can be useful to manage cardiovascular and liver diseases due to the presence of tocopherol. The study investigated the possible medicinal effects of *T. conophorum* seed extract on Mercury chloride-induced cardiac and hepatic toxicity. A total of twenty-four (24) male Wistar rats, weighing between 184-276kg, were used for this study. The rats were randomly divided into six groups (A-F) of four rats each. Group A served as control. Animals in groups B and C received daily oral administration of low dose and high dose 250 and 500mg/kg body weight of *T. conophorum* extracts only respectively. Groups D and E were the mercury chloride induced rats which were treated with low dose and high dose 250 and 500mg/kg body weight of *T. conophorum* extracts. Group F received 1mg/kg body weight solution of Mercuric chloride only. The rats were anesthetized under chloroform, heart and liver harvested and fixed in neutral buffered formalin for hematoxylin and eosin histological staining procedure, and histological slides were examined using light microscope. Histological findings reveal that mercuric chloride is both cardiotoxic and hepatotoxic. It causes severe damage to blood vessels in the heart, leading to ischemic effects on cardiac muscles, and induces significant liver damage, including inflammation and hepatocyte degeneration. *Tetracarpidium conophorum* extract provided some protection against these effects: it helped reduce liver inflammation and damage, and partially shielded the heart from vascular and myocardial damage. However, higher doses of the extract may not offer extra benefits and could potentially have adverse effects.

Keywords: Cardiotoxicity, Hepatotoxicity, Mercuric chloride, *Tetracarpidium conophorum*, Wistar rats

INTRODUCTION

Mercury (Hg) is a heavy metal with a well known toxicity (WHO, 2023). Mercuric chloride ($HgCl_2$) is a highly toxic compound that can cause significant damage to various organs, including the liver (Raeeszadeh *et al.*, 2021). Exposure to mercury chloride, whether through ingestion, inhalation, or skin contact, results in both acute and chronic toxic effects on the heart leading to coronary heart diseases, myocardial infarction, cardiac arrhythmias, carotid

artery obstruction (Bello *et al.*, 2023), also on the liver causing inflammation and cirrhosis (Wu *et al.*, 2024).

The heart is a muscular organ located in the thoracic cavity, consisting of four chambers: two upper atria and two lower ventricles (Rehman *et al.*, 2023). Histologically, the heart has three layers: the endocardium (inner lining), myocardium (thick muscle), and epicardium (outer layer). During embryonic development, the heart arises from the mesoderm and begins forming as a simple tube around the third week of gestation, later dividing into chambers through complex folding and septation processes, developing into the fully functional organ (Buijtenendijk *et al.*, 2020). The liver is the largest gland, located in the upper right abdomen, with two main lobes (right and left) (Vernon *et al.*, 2021). Histologically, it's composed of hexagonal lobules containing hepatocytes arranged in plates around central veins. These lobules are rich in sinusoids—capillaries allowing blood from the portal vein and hepatic artery to mix and filter. The liver originates from the endoderm during embryonic development and forms from the hepatic diverticulum in the foregut (Vernon *et al.*, 2021).

The tropical African walnut, known as *Tetracarpidium conophorum* (*T. conophorum*) belongs to the family Euphorbiaceae (Ayodeji *et al.*, 2018). In Nigeria, Yoruba call it Awusa or Asala, while Igbo calls it Ukpa (Ododo, 2019) it is known as *okhue* or *okwe* among the Bini tribe of Edo State (Chijoke *et al.*, 2015). The seeds are consumed as snacks and refreshments. Several studies on *T. conophorum* indicates that it contains bioactive compounds such as oxalates, phytates, tannins, saponins and alkaloids which partly shows the use of the seeds, leaves and roots in herbal medicine (Ayodeji *et al.*, 2018). A chemical evaluation of *T. conophorum* nuts revealed it can be useful to manage cardiovascular disease due to the presence of tocopherol (Balogun *et al.*, 2023) as well as diseased hepatic conditions (Oriakhi *et al.*, 2018). Therefore, the present study is focused on investigating the possible medicinal effects of *T. conophorum* seed extract on Mercury chloride-induced cardiac and hepatic toxicity.

MATERIALS AND METHOD

Preparation of extract: *Tetracarpidium conophorum* seeds were obtained from New Benin Market and authenticated at the Plant Biology and Biotechnology herbarium, University of Benin, where they were assigned herbarium specimen voucher number UBH-P490. The seeds were cleaned, air-dried at room temperature, and finely ground using a British milling machine (Viking Exclusive Joncod, Type YL112M-2). The powdered seeds were soaked in ethanol for 24 hours, then filtered using filter paper. The filtrate was evaporated at 40°C in a water bath, and the yield was weighed. The residue was refrigerated at the University of Benin's Anatomy Department. For the experiment, 10g of the seed extract was dissolved in 100ml of distilled water. 1g of Mercuric chloride was dissolved in 100ml of distilled water to form Mercuric chloride solution and administered.

Experimental Design: Twenty-four (24) male Wistar rats, weighing between 184-276kg, were used for this study. The rats were randomly divided into six groups (A-F) of four rats each. Group A served as control and received 1ml of distilled water. Animals in groups B and C received daily oral administration of low dose and high dose 250 and 500mg/kg body weight of *T. conophorum* extracts only, respectively. Groups D and E were the Mercuric chloride induced rats which were treated with low dose and high dose 250 and 500mg/kg body weight of *T. conophorum* extracts. Group F received 1mg/kg body weight solution of Mercuric chloride only. Rats were anesthetized under chloroform, heart and liver were harvested and fixed in neutral buffered formalin for hematoxylin and eosin histological staining procedure, and histological slides were examined using light microscope.

Histological Assessment: At the conclusion of the 28-day treatment, the rats were euthanized using chloroform. The hearts were collected and preserved in 10% buffered formal saline for 72 hours, then processed following the hematoxylin and eosin staining method as outlined by Drury and Wallington (1980). The prepared tissue slides were analyzed using a Leica DM750 research microscope equipped with a Leica ICC50 digital camera. Photomicrographs of the tissue sections were captured at 400x magnification.

Full Blood Count Test: The full blood count was performed using an automated hematology analyzer (BC-6000) from Shenzhen Mindray Bio-Medical Electronics Co., Ltd.

Liver Enzymes Tests: Serum was separated from blood samples through centrifugation (3,000 × g for 15 minutes). Liver enzyme and protein levels were then measured using an automated biochemical analyzer (Hitachi 7100, Japan).

Statistical Analysis: Data were analyzed using GraphPad Prism version 8.1. One-way analysis of variance (ANOVA) was performed, and results were expressed as mean ± standard error of the mean (SEM). Post-hoc comparisons were made using the least significant difference (LSD) test. A p-value of <0.05 was considered statistically significant. The statistical results were then displayed as tables.

RESULT

Effect of Treatment on Weight: There were no significant increases ($p > 0.05$) in the final body weights compared to the initial weight in the HgCl₂-only group and the HgCl₂-induced group treated with 250mg/kg extract (Table 1). Also, no significant changes ($p > 0.05$) were observed in heart weight for the HgCl₂-only and extract-treated groups compared to the control. Similarly, there were no significant changes ($p > 0.05$) in liver weight between the HgCl₂-only and extract-treated groups compared to the control. For the organosomatic indices, no significant changes ($p > 0.05$) were found in the organosomatic index for heart weight compared to the control. However, a significant increase ($p < 0.05$) was noted in the HgCl₂-only group compared to the control, but no significant changes ($p > 0.05$) occurred in the extract-treated groups compared to control.

Table 1: Mean values of Body weight, organ weight and organosomatic index of Mercury chloride (HgCl₂) induced toxicity Wistar rats treated with doses of extract.

	Control	250mg/kg <i>T.conophorum</i> only	500mg/kg <i>T.conophorum</i> only	250mg/kg <i>T.conophorum</i> + HgCl ₂	500mg/kg <i>T.conophorum</i> + HgCl ₂	HgCl ₂ only
Initial body weight (g)	238.7 ± 31.67	247.7 ± 14.66	225.0 ± 3.79	222.8 ± 6.5	212.3 ± 12.63	215.0 ± 5.033
Final body weight	278.3 ± 34.67	284.3 ± 13.91	275.3 ± 8.99	244.0 ± 6.89	230.5 ± 18.47	245.0 ± 13.23
Heart weight (g)	0.83 ± 0.42	1.00 ± 0.10	0.90 ± 0.30	0.925 ± 0.13	0.775 ± 0.10	1.10 ± 0.058
Liver weight (g)	9.40 ± 0.61	8.933 ± 0.61	8.87 ± 0.27	9.43 ± 0.78	8.48 ± 0.85	10.17 ± 0.64
Organo-somatic index	0.0039 ± 0.0001	0.0035 ± 0.0002	0.0032 ± 0.001	0.0038 ± 0.0004	0.0034 ± 0.0004	0.0045 ± 0.0004

Values are given as mean ± SEM. *P<0.05 indicates significant difference with control

Effect of Treatment on Liver enzymes: There was a significant increase in the group treated with 500mg/kg of *T. conophorum* extract compared to the control. No significant changes

($p > 0.05$) was noted in other groups. No significant changes ($p > 0.05$) were observed in either HgCl₂-induced or treated groups compared to control. No significant differences ($p > 0.05$) were noted in any of the groups. There were significant decreases ($p < 0.05$) in HgCl₂-induced groups treated with 250mg/kg and 500mg/kg of extract compared to the control, but no significant changes ($p > 0.05$) in the extract-only groups. Significant decreases ($p < 0.05$) were found in both the HgCl₂-induced and treated groups (250mg/kg, 500mg/kg) compared to control. A significant increase ($p < 0.05$) was observed in the 250mg/kg extract-only group compared to the control, but no significant changes ($p > 0.05$) occurred in other groups.

Table 2: Mean values of liver function indices of Mercury chloride (HgCl₂) induced toxicity Wistar rats treated with various doses of extract.

	Control	250mg/kg <i>T.conophorum</i> only	500mg/kg <i>T.conophorum</i> only	250mg/kg <i>T.conophorum</i> + HgCl ₂	500mg/kg <i>T.conophorum</i> + HgCl ₂	HgCl ₂ only
ALP (U/L)	177.7 ± 12.91	168.0 ± 38.40	206.3 ± 21.11	194.8 ± 4.05	295.0 ± 21.71	223.3 ± 43.46
AST (U/L)	88.33 ± 40.34	51.00 ± 35.00	80.67 ± 50.75	60.75 ± 16.27	168.0 ± 31.14	109.3 ± 70.88
ALT (U/L)	82.00 ± 34.77	71.00 ± 45.24	65.33 ± 30.15	83.50 ± 16.68	92.00 ± 14.37	76.33 ± 20.19
total bilirubin	0.30 ± 0.06	0.20 ± 0.06	0.267 ± 0.033	0.25 ± 0.03	0.3000 ± 0.041	0.30 ± 0.00
Total protein	8.200 ± 0.058	8.267 ± 0.12	8.167 ± 0.133	7.3 ± 0.16	7.1 ± 0.168	6.9 ± 0.53
Albumin (mg/dl)	4.17 ± 0.03	3.83 ± 0.03	3.8 ± 0.10	3.350 ± 0.19	3.45 ± 0.095	3.2 ± 0.32
Globulin (mg/dl)	4.03 ± 0.03	4.43 ± 0.09	4.37 ± 0.23	3.95 ± 0.12	3.65 ± 0.18	3.4 ± 0.44

Values are given as mean ±SEM. * $P < 0.05$ indicates significant difference with control

Effect of Treatment on Hematological Indices: For white blood cell count, a significant decrease ($p < 0.05$) was observed in the 250mg/kg extract-only group compared to the control, with no significant changes ($p > 0.05$) in other groups. Lymphocyte Count showed a significant decrease ($p < 0.05$) in the HgCl₂-induced group treated with 250mg/kg extract compared to the control, while no significant changes ($p < 0.05$) occurred in the extract-only groups. MID Count showed significant decreases ($p < 0.05$) in the HgCl₂-induced groups treated with 250mg/kg and 500mg/kg extract compared to the control, with no significant changes ($p > 0.05$) in extract-only groups. Granulocyte Count showed no significant changes ($p > 0.05$) in any of the groups compared to control. For red blood cell count, haemoglobin concentration, and haematocrit level, no significant differences ($p > 0.05$) were seen in any groups compared to control. However, mean cell volume and Mean Corpuscular Haemoglobin showed significant decreases ($p < 0.05$) in the HgCl₂-induced groups treated with 250mg/kg, 500mg/kg, and untreated HgCl₂-induced groups compared to control, with no significant changes ($p > 0.05$) between treated and untreated HgCl₂ groups. Mean Corpuscular Haemoglobin Concentration showed no significant changes ($p > 0.05$) in any groups compared to control.

Table 3: Mean values of haematological indices of Mercury chloride (HgCl₂) induced toxicity Wistar rats treated with doses of extract.

	Control	250mg/kg <i>T.conophorum</i> only	500mg/kg <i>T.conophorum</i> only	250mg/kg <i>T.conophorum</i> + HgCl ₂	500mg/kg <i>T.conophorum</i> + HgCl ₂	HgCl ₂ only
WBC (x10 ⁹ /L)	5.63 ± 1.02	2.17 ± 0.55	4.567 ± 0.75	4.975 ± 0.74	8.875 ± 1.86	8.467 ± 1.09
Lymphocytes (%)	86.77 ± 1.52	88.57 ± 1.24	88.40 ± 1.02	91.83 ± 1.26	91.78 ± 1.28	90.90 ± 1.77
MID (%)	8.63 ± 0.75	5.567 ± 0.90	7.333 ± 0.65	4.825 ± 0.97	5.225 ± 0.91	6.367 ± 1.49
Granulocytes (%)	4.60 ± 0.93	5.867 ± 1.495	4.200 ± 0.40	3.350 ± 0.40	3.000 ± 0.67	2.733 ± 0.29
RBC (x10 ¹² /L)	5.623 ± 0.48	5.460 ± 1.48	6.403 ± 0.24	6.125 ± 0.29	6.148 ± 0.98	6.893 ± 0.49
Haemoglobin (mg/dl)	13.27 ± 0.75	10.83 ± 3.08	13.30 ± 0.46	12.93 ± 0.80	11.98 ± 2.28	14.13 ± 1.014
Haematocrit (%)	39.10 ± 1.47	33.13 ± 8.89	38.80 ± 2.26	38.10 ± 1.45	37.55 ± 6.17	40.70 ± 3.34
MCV (fl)	70.33 ± 3.09	60.93 ± 0.63	60.60 ± 1.21	62.33 ± 0.71	61.03 ± 0.96	59.07 ± 1.59
MCH (pg)	23.70 ± 0.92	19.57 ± 0.50	20.73 ± 0.63	21.03 ± 0.397	19.03 ± 0.998	20.43 ± 0.22
MCHC (g/dl)	33.83 ± 0.79	32.20 ± 0.85	34.40 ± 1.61	33.80 ± 1.02	31.23 ± 1.26	34.73 ± 0.58

Values are given as mean ±SEM. *P<0.05 indicates significant difference with control

Effect of Treatment on Heart Histology: In the control group (Fig. 1A), normal heart architecture was observed, with organized cardiomyocytes and a structurally intact coronary artery. Similarly, the groups administered 250 mg/kg and 500 mg/kg of extract only exhibited normal cardiomyocytes; however, there was noticeable interstitial congestion and vasodilation (Fig. 1B and C). The myocardial fibers appeared normal, but interstitial congestion and vasodilation were present in the group given 250 mg/kg Extract + HgCl₂ (Fig. 1D). Also, the group given 500 mg/kg Extract + HgCl₂ (Fig. 1E) showed signs of myocardial degeneration and coronary vascular stenosis. HgCl₂ only group (Fig. 1F) showed severe coronary vascular stenosis, accompanied by mild inflammatory cell infiltration.

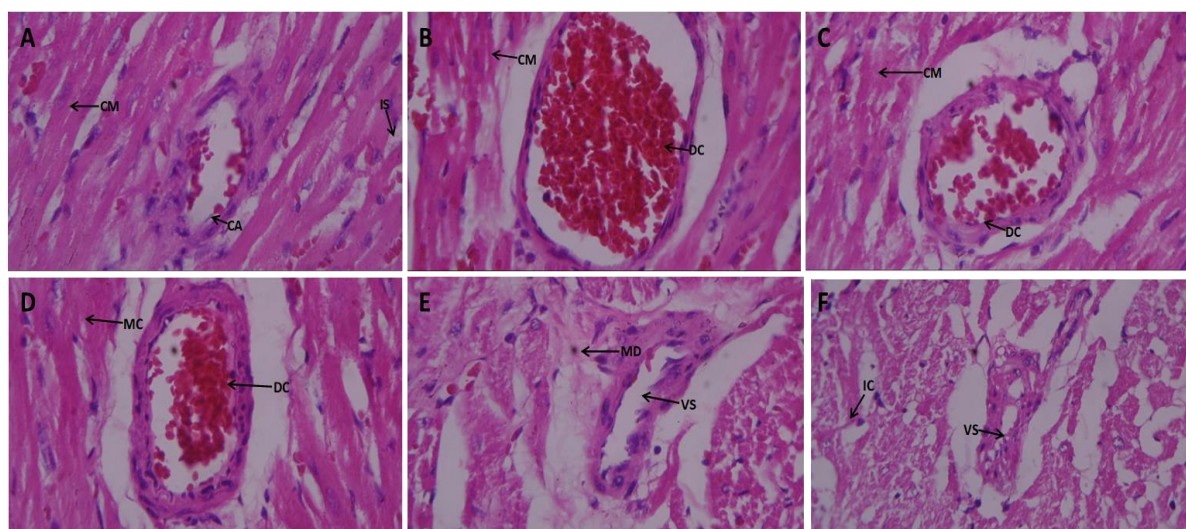


Figure 1: Rat heart. (A) Control group showing normal architecture composed of bundles of cardiomyocytes (CM), interstitial space and (IS) coronary artery (CA), (B) given 250mg/kg Extract only showing: normal bundles of cardiomyocytes (CM) and active interstitial congestion and vasodilatation (DC), (C) given 500mg/kg Extract only showing: normal bundles of cardiomyocytes (CM) and active interstitial congestion and vasodilatation (DC), (D) given 250mg Extract + HgCL₂ showing: normal bundles of myocardial fibres (MC) and active interstitial congestion and vasodilatation (DC), (E) given 500mg/kg Extract + HgCL₂ showing: myocardial degeneration (MD) and coronary vascular stenosis (VS), (F) given HgCL₂ only showing: severe coronary vascular stenosis (VS) and mild inflammatory cell infiltration (IC).

(MD) and coronary vascular stenosis (VS), (F) given HgCl₂ only showing: severe coronary vascular stenosis (VS) and mild interstitial infiltrates of inflammatory cells (IC), H&E 400x.

Effect of Treatment on Liver Histology: In the control group (Fig. 2A), the liver exhibited normal histological features, including healthy hepatocytes, sinusoids, bile ducts, and a properly structured portal vein. The 250 mg/kg Extract only group showed normal hepatocytes with mild vasodilation (Fig. 2B), while the 500 mg/kg Extract only group exhibited normal hepatocytes with Kupffer cell activation (Fig. 2C). Also, the HgCl₂ only group (Fig. 2D) demonstrated periportal hepatocyte degeneration, portal congestion, inflammatory infiltration, and Kupffer cell activation. However, the 250 mg/kg Extract + HgCl₂ group (Fig. 2E) showed normal hepatocytes with signs of vasodilation. Similarly, the group given 500 mg/kg Extract + HgCl₂ (Fig. 2F) showed periportal edema along with inflammatory infiltration.

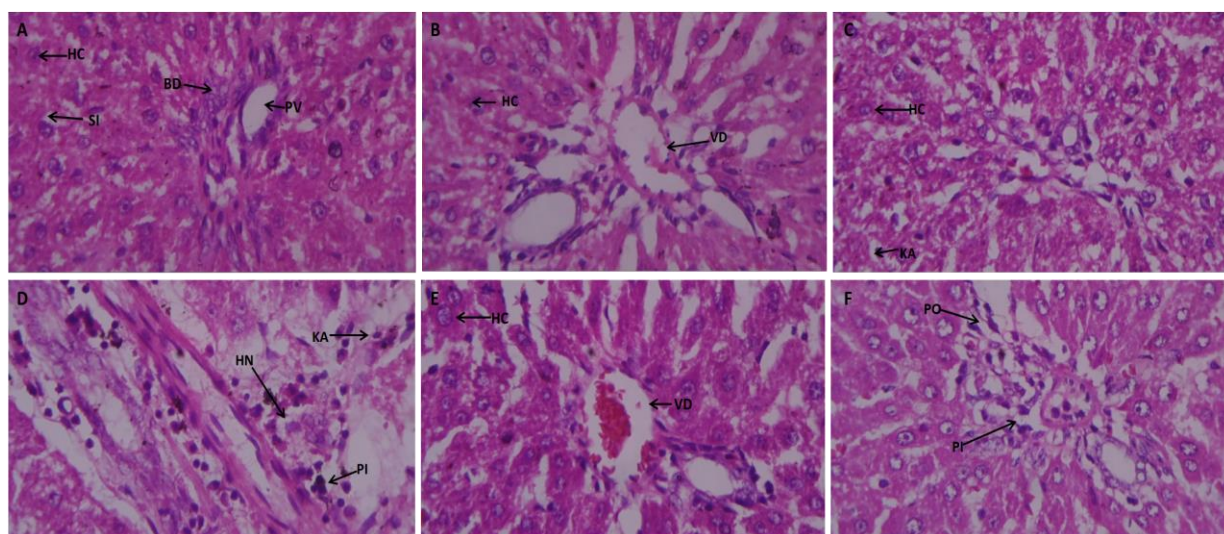


Figure 2: Rat liver, (A) Control. Showing normal architecture composed of: hepatocytes (HC), sinusoids (SI), bile ducts (BD) and portal vein (PV), (B) given 250mg Extract only showing: normal hepatocytes (HC) and vasodilation (VD), (C) given 500mg Extract only showing: normal hepatocytes (HC) and sinusoidal Kupffer cell activation (KA), (D) given HgCl₂ only showing: periportal hepatocyte degeneration (HN), portal congestion (PC), periportal infiltrates of inflammatory cells (PL) and Kupffer cell activation (KA) (E) given 250mg Extract + HgCl₂ showing: normal hepatocytes (HC) and vasodilation (VD), (F) given 500mg Extract + HgCl₂ showing: periportal oedema (PO) and periportal infiltrates of inflammatory cells (PI): H&E 400x

DISCUSSION

Mercury (Hg), a heavy metal with well-documented toxicity is known to cause severe damage to organs including the liver and heart (WHO, 2023). It is highly toxic and affects multiple organs, including the heart, kidneys, liver, and nervous system (Balali-Mood *et al.*, 2021). It induces oxidative stress, leading to cellular damage and apoptosis (Kim *et al.*, 2021). In the heart, it causes cardiotoxicity by disrupting calcium homeostasis, impairing mitochondrial function, and promoting inflammation, which can result in arrhythmias or heart failure (Wu *et al.*, 2024). Mercury chloride is also neurotoxic, damaging the nervous system, and nephrotoxic, harming kidney function (Maria-Francis *et al.*, 2023; Yahyazadeh *et al.*, 2024). Long-term exposure increases the risk of chronic diseases, including cardiovascular problems (Wu *et al.*, 2024). Research shows significant oxidative stress and inflammation across various organs, emphasizing the widespread impact of mercury chloride on human health (Singh *et al.*, 2024). The study investigated the protective effects of *T. conophorum* seed extract against HgCl₂-induced toxicity.

The study showed no significant ($p > 0.05$) changes in total body weight, heart weight, or liver weight among the different experimental groups. Specifically, the HgCl₂-only group and the HgCl₂-induced groups treated with *T. conophorum* extract exhibited similar body and organ weights compared to the control. However, the hepatosomatic index, which reflects the liver weight relative to body weight, was significantly higher in the HgCl₂-only group, indicating potential liver enlargement due to mercury toxicity. This is similar to the findings in a study by Gupta et al. (2017). The extract-treated groups did not show significant changes in the hepatosomatic index compared to the control, suggesting a possible protective effect of *T. conophorum* on liver structure similar to research findings according to Akomolafe et al. (2017). Liver enzymes such as alkaline phosphatase (ALP), aspartate aminotransferase (AST), and alanine aminotransferase (ALT) are crucial indicators of hepatic function and damage. The study observed a significant increase in ALP in the 500mg/kg *T. conophorum* extract-treated group compared to the control, which might indicate liver stress or regeneration as previously noted by Owoeye (2016). No significant changes were observed in AST and ALT levels across the experimental groups, which could suggest that *T. conophorum* extract did not significantly alter the liver's enzyme synthesis in response to HgCl₂. A decrease in total protein and albumin levels were observed in the HgCl₂-induced groups, indicating potential liver dysfunction. A decrease in total protein and albumin levels in HgCl₂-induced groups suggests liver dysfunction because the liver is responsible for producing most of the body's proteins, including albumin (Moman et al., 2022). This disruption in protein synthesis is a key indicator of liver injury, as albumin levels reflect the liver's ability to maintain protein production. Low protein and albumin levels can lead to complications such as fluid imbalance, reduced immune function, and impaired nutrient transport, further indicating hepatic damage in mercury chloride exposure (Balali-Mood et al., 2021). The extract-treated groups showed decreased levels in these parameters, suggesting possible restoration of hepatic function or reduced liver damage.

Hematological assessments revealed significant decreases in white blood cell (WBC) count and lymphocyte count in the 250mg/kg extract-only group compared to control, indicating potential immunomodulatory effects of *T. conophorum*. A reduction in WBCs and lymphocytes typically indicates suppression of the immune response, which could mean the extract is influencing immune cell production or function (Waldman et al., 2020). This effect could be either beneficial or harmful, depending on the context – potentially reducing inflammation or weakening immune defenses. Further research is needed to determine whether these changes in immune cell counts reflect protective or adverse outcomes from the use of *T. conophorum* extract. Significant decreases in MID cell count in HgCl₂-induced groups treated with 250mg/kg and 500mg/kg of the extract, reflected possible modulation of bone marrow activity or immune response. MID cells play crucial roles in immune defense, and their reduction may indicate that the extract affects bone marrow production or influences the body's immune regulation. This could involve suppressing overactive immune reactions or altering bone marrow cell generation, which is essential for producing immune cells (Jiang and Wu, 2019). No significant changes were observed in granulocyte count, RBC count, hemoglobin concentration, or hematocrit levels among the groups, suggesting that *T. conophorum* might not significantly influence these parameters under the given conditions.

Histological examination of the heart and liver tissues revealed that the control group exhibited normal heart architecture, while the HgCl₂-only group showed severe coronary vascular stenosis and mild inflammatory cell infiltration. The 500mg/kg extract + HgCl₂ group displayed myocardial degeneration and coronary vascular stenosis, suggesting that higher doses of the extract might not fully protect against mercury-induced cardiac damage.

This raises important questions about the extract's dose-response relationship. While extracts like *Tetracarpidium conophorum* may have antioxidant or anti-inflammatory properties, higher doses may not always translate into greater protection. In fact, they could interact with mercury's toxic effects in unpredictable ways, either by insufficiently countering oxidative stress or by interfering with the heart's normal repair processes. It's possible that lower doses of the extract might offer more balanced protection or that combining the extract with other treatments could enhance its protective effect.

Histological assessment of the liver in control and extract-only groups showed normal liver architecture. The HgCl₂-only group exhibited severe hepatic damage, including periportal hepatocyte degeneration and inflammatory infiltration. The 250mg/kg extract + HgCl₂ group displayed relatively normal hepatocytes with some vasodilation, while the 500mg/kg extract + HgCl₂ group showed periportal edema and inflammatory infiltration, indicating some level of hepatic protection by the extract but also potential side effects at higher doses. While these findings suggest that the extract still offers some level of hepatic protection, the presence of edema and inflammation indicates that higher doses may introduce unwanted side effects. The periportal edema could be an indication of a compensatory response to damage or toxicity, suggesting that the higher dose of the extract may not fully counteract the effects of HgCl₂ and might even contribute to fluid imbalance or inflammation. These observations raise important considerations regarding the extract's dose-dependent effects. While lower doses appear to provide better protection, higher doses may lead to incomplete protection or additional side effects. The balance between protective and adverse effects at different doses needs to be explored further to optimize the therapeutic use of the extract.

The study highlights significant findings of the effects of *Tetracarpidium conophorum* seed extract on mercury chloride-induced damage. Exposure to mercury chloride caused noticeable damage to liver tissue, including inflammation and congestion. However, rats treated with the seed extract exhibited reduced liver damage, suggesting the extract has protective properties. Mercury exposure can severely impact liver health, leading to conditions like liver fibrosis and potentially cirrhosis, a serious liver disease.

CONCLUSION

The study suggested that *Tetracarpidium conophorum* seed extract helps protect the heart and liver from damage caused by mercury chloride. Its effectiveness depended on lower doses which helped to reduce liver damage, and higher doses did not offer extra benefits and was shown to cause possible harm.

Further investigation into the molecular mechanisms of *T. conophorum*'s action, including its impact on oxidative stress and inflammation pathways, is recommended to fully understand its therapeutic potential.

REFERENCES

- Aguwa, U. S., Owoeye, O., Olu, S. I., and Ukoba, O. (2016). Teratogenic effect of maternal vitamin A consumption on the liver, limbs and other morphological parameters of the pups of wistar rats. *International Journal of Basic, Applied and Innovative Research*, 5(4), 130-137.
- Akomolafe, S. F., Oboh, G., Oyeleye, S. I., and Olasehinde, T. A. (2017). Toxicological effects of aqueous extract from African walnut (*Tetracarpidium conophorum*) leaves in rats. *Journal of Evidence-Based Complementary and Alternative Medicine*, 22(4), 919-925.

- Ayodeji, A. E., and Aliyu, N. (2018). *Tetracarpidium conophorum* (African walnut) Hutch. and Dalziel: Ethnomedicinal uses and its therapeutic activities. *Journal of Medicinal Plants for Economic Development*, **2**(1), 1-10.
- Balali-Mood, M., Naseri, K., Tahergorabi, Z., Khazdair, M. R., and Sadeghi, M. (2021). Toxic mechanisms of five heavy metals: mercury, lead, chromium, cadmium, and arsenic. *Frontiers in Pharmacology*, **12**, 643972.
- Balogun, M. E., Manzuma A. O., Ayinde T. O. and Folami S. O. (2023). *Tetracarpidium Conophorum* (African Walnut): A Review of Pharmaceutical Evidence on Cardiac Toxicity. *Al-Hikmah Journal of Health Sciences*, **3**(1), 36 – 43.
- Bello, K. A., Wilke, M. C. B., Simões, R. P., Landim-Vieira, M., Langa, P., Stefanon, I., Vassallo, D. V., and Fernandes, A. A. (2023). Chronic exposure to mercury increases arrhythmia and mortality post-acute myocardial infarction in rats. *Frontiers in Physiology*, **14**, 1260509.
- Buijendijk, M. F., Barnett, P., and van den Hoff, M. J. (2020, March). Development of the human heart. In *American Journal of Medical Genetics Part C: Seminars in Medical Genetics* (Vol. 184, No. 1, pp. 7-22). Hoboken, USA: John Wiley and Sons, Inc..
- Chijioke, O. C., Anosike, C., and Ani, C. C. (2015). Studies on the phytochemical and nutritional properties of *Tetracarpidium conophorum* (black walnut) seeds. *Journal of Global Bioscience*, **4**(2), 1366-1372.
- Drury, R. A. B., and Wallington, E. A. (1980). Carleton's histological technique. *Archives of Biochemistry and Biophysics*, **82**, 70-77.
- Gupta, N., Gupta, D. K., and Sharma, P. K. (2017). Condition factor and organosomatic indices of parasitized *Rattus rattus* as indicators of host health. *Journal of Parasitic Diseases*, **41**, 21-28.
- Jiang, W., and Xu, J. (2020). Immune modulation by mesenchymal stem cells. *Cell proliferation*, **53**(1), e12712.
- Kim, J. Y., An, M. J., Shin, G. S., Lee, H. M., Kim, M. J., Kim, C. H., and Kim, J. W. (2021). Mercury chloride but not lead acetate causes apoptotic cell death in human lung fibroblast mrc5 cells via regulation of cell cycle progression. *International Journal of Molecular Sciences*, **22**(5), 2494.
- Maria Francis, Y., Karunakaran, B., Ashfaq, F., Yahia Qattan, M., Ahmad, I., Alkhatami, A. G., Khan, M. I., Varadhan, M., Govinda, L., and Ponnusamy Kasirajan, S. (2023). Mercuric chloride induced nephrotoxicity: Ameliorative effect of *Carica papaya* leaves confirmed by histopathology, immunohistochemistry, and gene expression studies. *ACS omega*, **8**(24), 21696-21708.
- Moman, R. N, Gupta N., Varacallo M. Physiology, Albumin. [Updated 2022 Dec 26]. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2024 Jan-. Available from: <https://www.ncbi.nlm.nih.gov/books/NBK459198/>
- Odod0, A. (2024, September 16). Health benefits of walnuts. *The Sun Nigeria (Voice of the Nation)*. <https://thesun.ng/health-benefits-of-walnuts/#:~:text=The%20name%20of%20the%20plant,while%20Igbo%20calls%20it%20Ukpa>.
- Oriakhi, K., Uadia, P. O., and Eze, I. G. (2018). Hepatoprotective potentials of methanol extract of *T. conophorum* seeds of carbon tetrachloride induced liver damage in Wistar rats. *Clinical Phytoscience*, **4**, 1-12.
- Raezadeh, M., Moradi, M., Ayar, P., and Akbari, A. (2021). The antioxidant effect of *Medicago sativa* L.(alfalfa) ethanolic extract against mercury chloride (HgCl₂) toxicity in rat liver and kidney: an in vitro and in vivo study. *Evidence-Based Complementary and Alternative Medicine*, **2021**(1), 8388002.

- Rehman, I., and Rehman, A. (2023, August 28). Anatomy, thorax, heart. In *StatPearls*. StatPearls Publishing. <https://www.ncbi.nlm.nih.gov/books/NBK470256/>
- Singh, S., Dwivedi, S., Khan, A. A., Jain, A., Dwivedi, S., Yadav, K. K., Dubey, I., Trivedi, A., Trivedi, S. P., and Kumar, M. (2024). Oxidative stress, inflammation, and steatosis elucidate the complex dynamics of HgCl₂ induced liver damage in *Channa punctata*. *Scientific Reports*, **14**(1), 9161.
- Vernon, H., Wehrle, C. J., Alia, V. S. K., and Kasi, A. (2018). Anatomy, abdomen and pelvis, liver.
- Waldman, A. D., Fritz, J. M., and Lenardo, M. J. (2020). A guide to cancer immunotherapy: from T cell basic science to clinical practice. *Nature Reviews Immunology*, **20**(11), 651-668.
- World Health Organization. (2023). *Exposure to lead: a major public health concern. Preventing disease through healthy environments*. World Health Organization.
- Wu, Y. S., Osman, A. I., Hosny, M., Elgarahy, A. M., Eltaweil, A. S., Rooney, D. W., Chen, Z., Rahim, N. S., Sekar, M., Gopinath, S. C. B., Rani, N. N. I. M., Batumalaie, K., and Yap, P. S. (2024). The toxicity of mercury and its chemical compounds: molecular mechanisms and environmental and human health implications: a comprehensive review. *Acs Omega*, **9**(5), 5100-5126.
- Yahyazadeh, A., and Gur, F. M. (2024). Promising the potential of β -caryophyllene on mercury chloride-induced alteration in cerebellum and spinal cord of young Wistar albino rats. *Naunyn-Schmiedeberg's Archives of Pharmacology*, 1-15.