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Ameliorative effects of aqueous extract of *Cyperus esculentus* (Tiger Nut) on cadmium-induced liver damage in adult Wistar rats.

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ABSTRACT

Cyperus esculentus (Tiger nut milk) is rich in minerals and vitamins essential for body growth, development and improvement of immune functions. This study was undertaken to investigate the effects of aqueous extract of *Cyperus esculentus* on cadmium induced toxicity in the liver of adult Wistar rats. Thirty (30) rats weighing between 180 – 200g were divided into six groups (A, B, C, D, E and F) of five animals each. **Group A** representing the control was given standard rat chow and water *ad libitum*. However Group B was given cadmium chloride only; while groups C, D, and E were received cadmium chloride and aqueous extract of *Cyperus esculentus* at doses of 150, 300 and 600mg/kg body weight, respectively; Group F was administered cadmium chloride and standard drug, Atorvastatin (10mg/kg). Administration to all the animals in the experimental group was done using orogastric tube for 60 days. At the end of the treatment period, they were anaesthetized using chloroform and blood sample were collected via cardiac puncture for liver function test, while the liver tissue was harvested, and processed for histological analysis. Statistically, the results show no significant difference ($P > 0.05$) with each enzyme and protein in all the groups when compared to control. Histological sections revealed vascular congestion, heavy periportal infiltrates of inflammatory cells in the liver treated with cadmium chloride. The observed histopathological changes are features of liver toxicity induced by cadmium chloride administration. However, administration of *Cyperus esculentus* extract was observed to ameliorate the toxicity in the liver and this compared well with the Atorvastatin.

Keywords: *Cyperus esculentus*, Wistar rats, Liver

INTRODUCTION

Cadmium is a malleable, ductile and toxic metal commonly used in the production of polyvinyl chloride (PVC), pigments and batteries. It is contained in most consumer products like cigarettes, jewelry, and also found in sufficient quantity in sea food. It has been reported that short-term exposure via air to high cadmium levels could lead to coughing, chest pain and nasal irritation, however taking food or water contaminated with cadmium may irritate the stomach and cause vomiting and diarrhea. Long-term exposure could lead to Cancer, Hepatic and Renal failures¹.

The use of plants in most developing countries as a normative basis for the maintenance of good health has been widely observed². They contain various phytochemicals important for up regulating immune function. About 80% of the world's population relies essentially on plants for primary health care³.

Cyperus esculentus commonly known as Tiger nut is a tuber that grows freely in Nigeria, and other parts of West Africa. It contains starch in rich quantity, essential vitamins and minerals⁴. Its phytochemical constituents aid management of cardiovascular diseases and

cancer⁵. Vitamin E and antioxidants present in the plant provide protection against free radicals⁶. The very high fiber content combined with a delicious taste makes it ideal for healthy eating. Report shows that it could prevent and manage urinary tract and bacterial infections⁷. They are thought to be beneficial to diabetics and those seeking to reduce cholesterol or lose weight. Its milk extract has been proven suitable for patients unable to tolerate gluten and lactose. Tiger nuts have been reported to possess aphrodisiac properties, boost fertility and ameliorate testicular toxicity⁸.

The liver is the largest glandular organ in the human body, located in the right hypochondriac region of the abdomen. Responsible for metabolism of biomolecules, therefore its proper functioning is essential in the overall welfare of the human body⁹. Due to its wide range of activities, the liver is prone to damage from oxidative stress induced by heavy metals and environmental toxicants¹⁰, as a result there is a global surge in liver related morbidity due to the preponderance of degenerative diseases¹¹. This study will provide information on the possible ameliorative

effects of aqueous extract of *Cyperus esculentus* on cadmium-induced damage on the liver using adult Wistar rats.

MATERIALS AND METHODS

Purchase of compound and Preparation of Extract:

Cadmium chloride manufactured by LOBA CHEMIE PVT. LTD. With batch No. L231151707, was purchased from Pyrex Pharmaceutical Company. Tiger nuts were purchased from New Benin market, Benin-City, Edo state and identified in the Department of Plant Biology and Biotechnology, university of Benin. 5kg of the air-dried Tiger nut was pulverized using a pestle and mortar. The powdered extract was soaked in 8L of distilled water in a chromatographic jar and stirred vigorously and left undisturbed for 48 hours at room temperature. The mixture was then filtered using a Buchner funnel and Whatman No.1 filter paper. The filtrate was then concentrated to dryness under reduced temperature and pressure using rotary evaporator. The extract was stored in an air-tight container and preserved in the refrigerator at 4°C until use.

Animal care, Grouping and Experimental Design:

Thirty Adult Male Wistar rats (180-200g) purchased and maintained at the Animal house of the Department of Anatomy, University of Benin, were randomly assigned into six groups containing five animals each. Group A served as control given access to feed and

water ad libitum, However Group B animals were administered cadmium chloride only, while animals in Group C, D and E received cadmium chloride and aqueous extract of *Cyperus esculentus* in low (150mg/kg), medium (300mg/kg) and high doses (600mg/kg per body weight) respectively. Group F animals were given cadmium chloride and standard drug, Atorvastatin, (at a dose of 10mg/kg body weight) manufactured by Pfizer Pharmaceuticals with Lot No. R39895 purchased from Chrisilva Pharmacy and Stores, Ovbogie, Benin City, Edo State, Nigeria. The extract was administered via oral route using orogastric tube for a period of sixty days.

Animal Sacrifice and Sample Collection: At the end of the experimental period, animals were anaesthetized using chloroform. The liver tissues were harvested and fixed in 10% buffered formalin for routine haematoxylin and eosin histological processing using the method of Drury *et al*¹². Blood was collected via cardiac puncture and was immediately transferred into plain sample bottles for liver enzyme analysis of Serum Alanine amino Transferase (ALT), Serum Aspartate amino Transferase (AST), Alkaline Phosphatase (ALP), Total Protein (TP), albumin, globulin, and total bilirubin¹³.

All data generated was analyzed using descriptive and inferential statistics. Values were presented as Mean

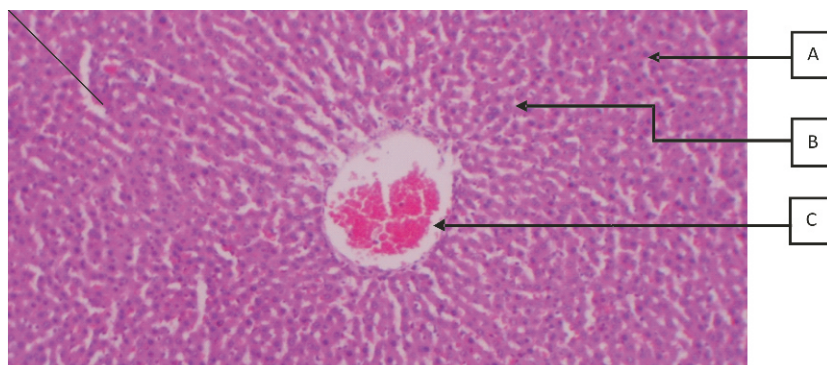


PLATE 1: Control- A: hepatocytes, B: sinusoids, C: central vein (H&E x 100)

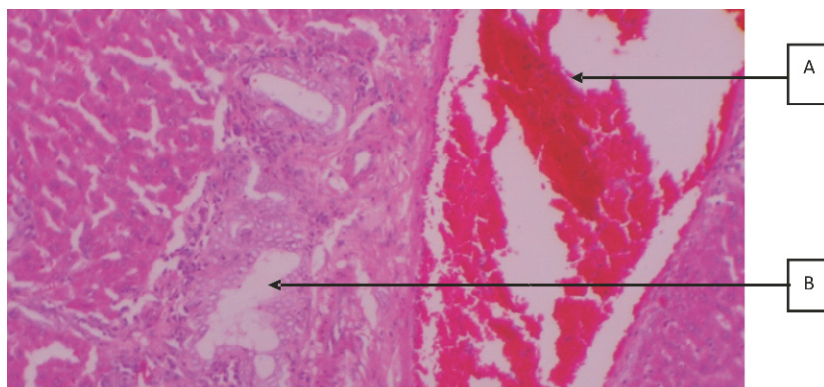


PLATE 2: Cadmium only - A: vascular congestion, B: heavy periportal infiltrates of inflammatory cells (H&E x 100)

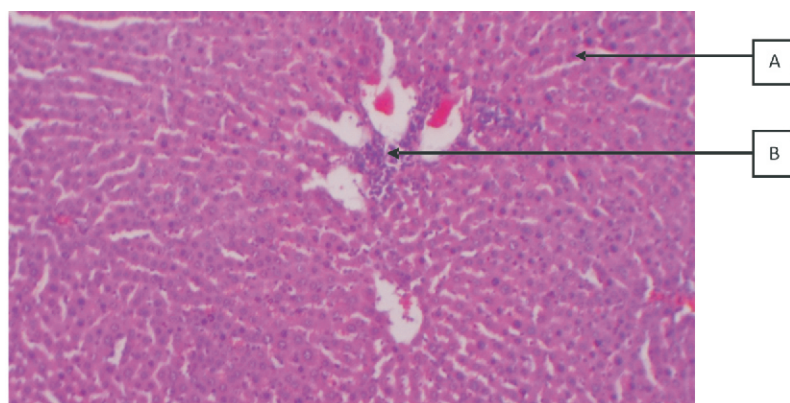


PLATE 3: Cadmium and Low Dose Extract - A: normal hepatocytes and B: mild periportal infiltrates of inflammation (H&E x100)

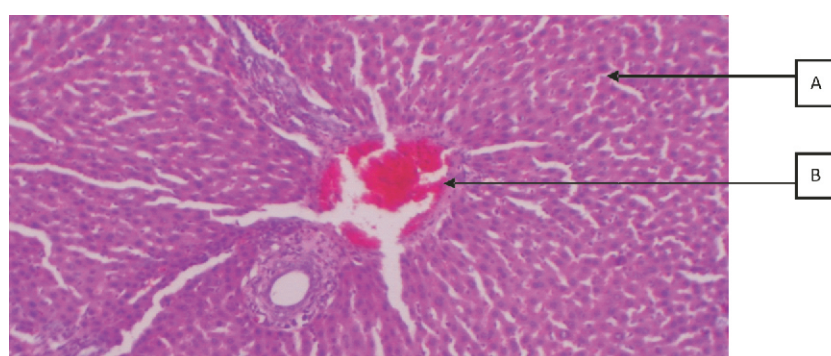


PLATE 4: Cadmium and Moderate Dose Extract - A: normal hepatocytes and B: normal vascular architecture (H&E x100)

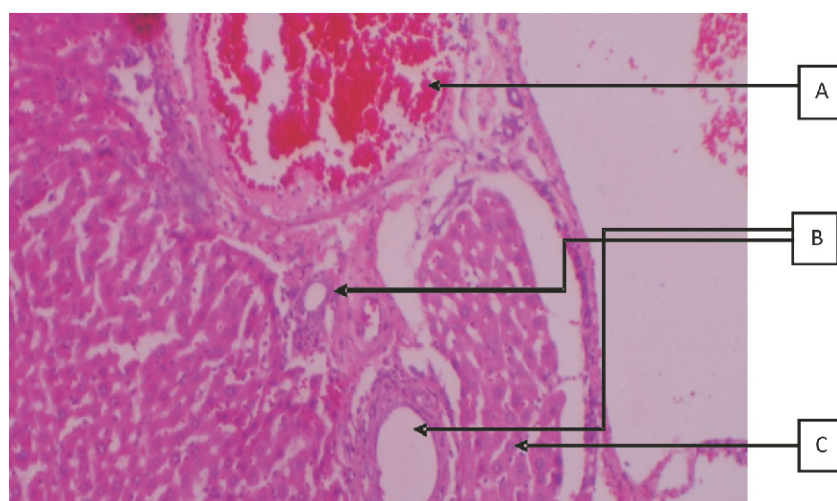


PLATE 5: Cadmium and High Dose Extract - A: normal vascular architecture, B: normal ductal orientation C: normal hepatocyte architecture (H&E x100)

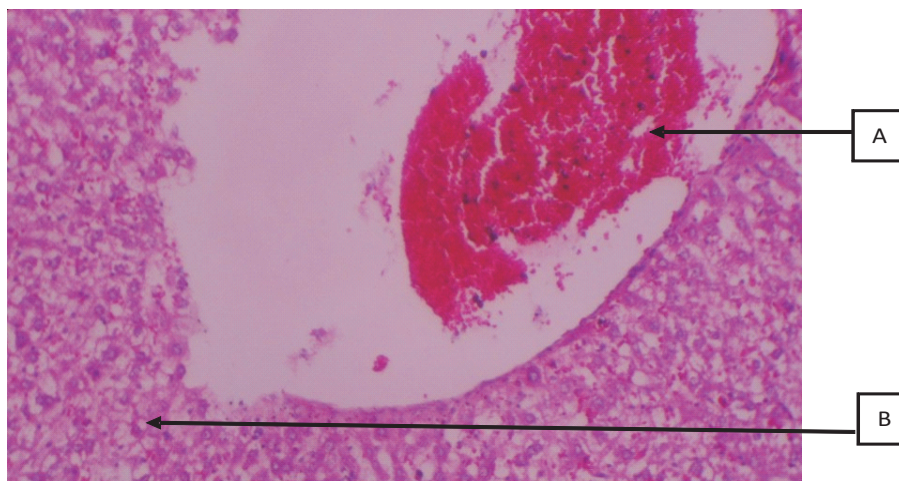


PLATE 6: Cadmium and Standard drug (Atorvastatin) - A: normal vascular architecture B: normal hepatocytes (H&E x 100)

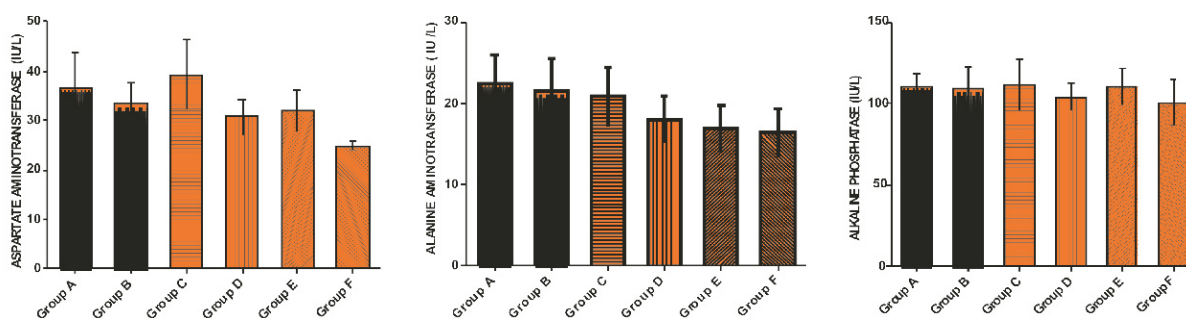


Figure 1: Serum level of liver enzymes following administration of aqueous extract of *Cyperus esculentus* and cadmium across the groups

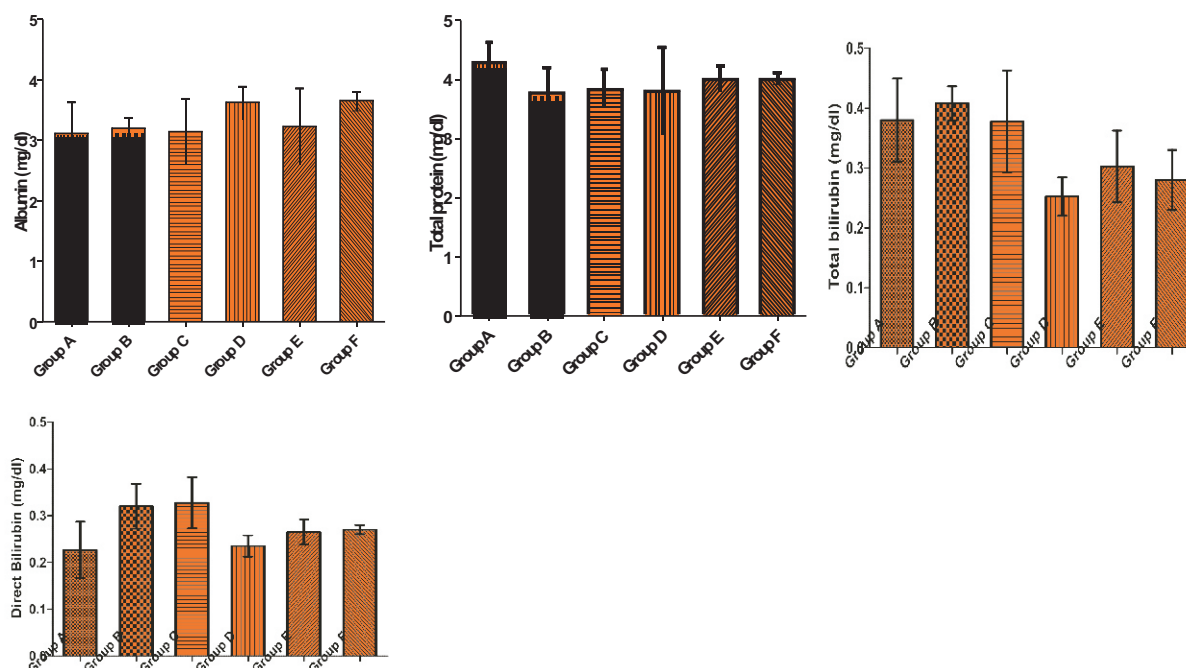


Figure 2: Showing Albumin, total bilirubin and protein following administration of aqueous extract of *Cyperus esculentus* and cadmium at various doses.

DISCUSSION

This study was undertaken to evaluate the possible ameliorative effects of *Cyperus esculentus* on cadmium chloride induced liver toxicity. Photomicrograph of animals in the control group shows normal hepatic architecture appeared with the liver parenchyma consisting of branching and anastomosing cords of hepatocytes radiating from the central vein towards the periphery and separated by slit-like blood sinusoids. These hepatocytes arranged in cords around the central vein have basophilic granular cytoplasm with central, rounded vesicular nuclei (Plate 1).

Cadmium is a toxic environmental chemical with wide range of industrial application¹⁴; it could be dangerous to health as observed in the study, where it was seen to induce severe damage to liver tissues. Cadmium treated group given 0.2mls of the compound, showed partial loss of normal hepatic cells, localized hemorrhage between degenerated cells, dilated and congested portal vein with thick fibrotic wall. Most hepatocytes appeared shrunken with pyknotic nuclei and eosinophilic cytoplasm. Some of them ill-defined and fragmented nuclei (Karyorrhexis). Similar findings were reported by Klassen *et al.*,¹⁵ and Mekawy *et al.*,¹⁶ in fish where the hepatocytes undergo cytoplasmic vacuolation and degeneration after cadmium treatment^{15,16}. More deleterious effects of cadmium chloride have been well established in previous studies^{17,18}.

These degenerative changes might be due to oxidative stress induced by reactive oxygen species; however plants containing substances with antioxidant properties could protect the cells, cushion the effects and possibly reverse the damage¹⁹. Report from the study of Temple *et al.*,²⁰ affirmed that *Cyperus esculentus* is a potent antioxidant rich in flavonoids and other phytochemicals²⁰. At low dose of the extract, there was slight improvement as the cells were mildly infiltrated with reduced inflammation (Plate 3). However at moderate dose, there was presence of normal hepatocyte and vascular architecture with slight congestion of red blood cells within the vessels (Plate 4). At high dose, there was complete reversal of damages induced by cadmium chloride liver, with normal vascular, ductal and hepatic architecture (Plate 5). Group F animals, on administration of standard drug, Atorvastatin, of course had normal hepatocyte configurations (Plate 6). In comparison, of the standard drug to the extract of *Cyperus esculentus*, Group E gave the best result due to the potency of its high dosage of aqueous extract.

There was no statistically significant effect ($P > 0.05$) in the activities of the enzymes Aminotransferases (ALT and AST), ALP as well as proteins, albumin and bilirubin. This result at variance with the findings by Oyedepo and Odoje²¹, who reported marked decline in liver enzymes pretreated with carbon chloride.

CONCLUSION

In conclusion, the aqueous extract of *Cyperus esculentus* demonstrated significant ameliorative potentials against cadmium chloride-induced hepatotoxicity. It was seen to better protect and preserve hepatocellular architecture when compared to standard drug Atorvastatin.

CONFLICT OF INTEREST: Nil

SOURCE OF FUNDING: None

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