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AMELIORATIVE EFFECT OF THE TIGER NUT (Cyperus Esculuntus) ON THE ALUMINIUM CHLORIDE INDUCED HISTOPATHOLOGICAL CHANGES ON THE SMALL INTESTINE OF ADULT WISTAR RATS

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ABSTRACT

The small intestine is the largest component of the digestive tract and the major site of digestion and absorption. The great abundance of Al increases the risk of exposure and related health issues in humans. Aluminum remains considerable a public health problem, which can cause a number of adverse effects in many body organs such as intestines, testes, kidney, liver, heart etc. which can cause many diseases like Alzheimer's disease, amyotrophic lateral sclerosis, and parkinsonism-dementia. For 4000 years, tiger nut has been used as a healthy plant because of its content of several minerals, energy, and oleic acid. It has a high content of arginine which liberates the hormone that produces insulin, besides its content of carbohydrates with a base of sucrose and starch.

The aim of this study was to evaluate the ameliorative effect of the tiger nut on the aluminum chloride induced histopathological changes on the small intestine of the wistar. The rats were divided into four (4) groups. Group 1 was the control that received normal saline, group 2 received tiger nut (cyperus esculentus), groups 3 and 4 received aluminum chloride and tiger nut (cyperus esculentus) all for 14days. The rats were humanely sacrificed and the proximal part (duodenum) of the small intestine was removed and fixed, processed and stained with haematoxylin and eosin (H&E). The slides were prepared and viewed with the light microscope. The result was obtained and observed. Therefore, from the results it was concluded that the tiger nut extract had ameliorative effect on the aluminum induced histopathological changes.

Keywords: Tigernut, Ameliorative, Aluminum chloride, small intestine, Wistar rats

INTRODUCTION

Tiger nut (Cyperus esculentus Lativum) belongs to Cyperaceae and it is known by other names like chufa, yellow nut sedge, earth almond and ground almond. It is a perennial crop cultivated extensively in Asia,

West Africa, such as Nigeria and parts of Europe particularly Spain as well as in the Arabian Peninsula [1]. Tiger nut is a crop of early domestication, its dry tubers have been found in tombs from predynastic times about 6000 years ago and it was an important food in Egypt where they used their tubers as sweetmeat [2]. Tiger nuts appear to have more prospective usage as nourishment and industrial materials; it can be used to produce beverage, milk or yogurt, flour, nougat, jam, beer, chocolate, a feed source, edible oil and as soaps [3]. Tiger nuts are rich in minerals such as phosphorus, potassium, calcium, magnesium, and iron. It's also rich in vitamins E and C, and a good quantity of vitamin B1 [4]. Tiger nut tubers are beneficial for bones, tissue repair, muscles, the bloodstream and body development due to its richness in phosphorus, potassium, calcium, magnesium and iron necessary [5]. Tiger nut was reported to be high in dietary fiber content, which is effective in the treatment and prevention of many diseases such as colon cancer, coronary health diseases, gastro intestinal disorders, obesity and diabetics [3]. It is used also in the treatment of flatulence, indigestion, diarrhea, dysentery and excessive thirst [6]. It is a rich source of minerals such as iron, magnesium, and carbohydrates more than the cow's milk, in addition to phosphorus, potassium, calcium, unsaturated fats, proteins and some enzymes which help in digestion. The milk is recommended for those who suffer from indigestion, flatulence, and diarrhea because of glucose, unsaturated fats, etc. [3]. Also, tiger nut is thought to have a preventive effect against cardiovascular diseases and cancer because of its content on vitamin E which plays a role on the formation and functioning of the red blood cells. [7]. In Nigeria, Hausa people call it "Aya", Yoruba "mumui", the Igbo "ofio", "aki Hausa" in southern Nigeria [8]. The small intestine (or small bowel) is the part of the gastrointestinal tract following the stomach and followed by the large intestine, and is where much of the digestion and absorption of food takes place. In invertebrates such as worms, the terms "gastrointestinal tract" and "large intestine" are often used to describe the entire intestine. The primary function of the small intestine is the absorption of nutrients and minerals found in food, as well as, completing the digestion of food delivered into it by the stomach [9]. The small intestine is the largest component of the digestive tract and the major site of digestion and absorption. In addition to receiving chyme from the stomach, the initial segment of the small intestine, the duodenum, receives bile from the gall bladder and digestive enzymes from the pancreas. The pancreatic enzymes are produced in an inactive form and only become active in the lumen of the duodenum. The small intestine is divided into three parts, the duodenum (25 cm), the jejunum (2.5 m) and the ileum (3.5 m). The duodenum is a C-shaped or horseshoe-shaped structure that lies in the upper abdomen near the midline. The mucosa of the small intestine is highly modified. The luminal surface is completely covered by a number of finger-like or leaf-like projections called villi, 0.5-1.5 mm in length [9].

Due to its reactivity, Al is naturally found in combination with other elements to form compounds such as Al sulfate and chloride [10]. High consumption of Al-containing products will increase the concentration of this metallic element in the consumers' organs and damage their various tissues (including the small intestine of humans and animals) [11]. Due to its content of carbohydrates with a base of sucrose and starch (without glucose), and its high content of Arginine, which liberates the hormone that produces insulin [12]. Particulate matters distributed by cement -producing factories contain high amount of Aluminum and populations residing in the vicinity are exposed to the pollution [13]. The small intestine is the largest

component of the digestive tract and the major site of digestion and absorption.

Orally-ingested aluminum compounds have been implicated in the development of dialysis encephalopathy, osteomalacic dialysis osteodystrophy and other disorders in both hemodialyzed and nonhemodialyzed patients suffering from chronic renal failure [14]. Studies monitoring aluminum absorption and elimination revealed an average positive balance from 23 to 313 mg of aluminum per day when diets were supplemented with 1 to 3g of aluminum per day [15]. These studies show that a small fraction of the ingested aluminum is absorbed. This absorption presents potential toxic effects to uremic patients whose ability to eliminate aluminum is impaired. In addition,[16] have shown that addition of citric acid to aluminum-supplemented dietary regimens results in blood aluminum levels that are significantly higher than those found in subjects treated with aluminum-supplemented dietary regimens alone. This result suggests that dietary factors may contribute to aluminum absorption.

MATERIAL AND METHODS

Chemicals

Aluminium chloride (AlCl3) was used as solution for oral administration. It was collected from department of biochemistry BUK. About 30g of AlCl3 was used in this research.

Reagents

Hematoxylin and Eosin stains (H&E), 10% formal saline solution (NBF), graded alcohol (absolute ethanol, 90% ethanol, 80% ethanol, 70% ethanol and 50% ethanol), xylene, 1% acid alcohol, chloroform and paraffin wax were obtained from department of human anatomy. Normal saline was purchased from pharmacy and distilled water was obtained from department of pharmacy, and tween eighty was obtained from department of Microbiology.

Instruments and Apparatus

The instruments and apparatus were collected from the department of Human anatomy, Biochemistry, Bayero university Kano, and some were purchased from pharmacy. they include; syringe, canula, small transparent container, small non transparent container, stirrer, digital weighing balance, cup, hand gloves, dissection kit, sacrifice board, cotton wool, masking tape, metallic mould, plastic cassette, microtome, hot air oven, refrigerator, microscope glass slide, water bath, cover slip, microscope, surgical blade, cotton wool, dessicator, refrigerator, and digital microscope imager.

EXPERIMENTAL ANIMALS

Twenty (20) adult wistar rats (combination of males and female) strain of body weight ranging from 60g to 170g were procured from Animal House of Biological science Department, Bayero Universi ty, Kano. The animal were housed in a clean standard polypropylene rat cage with filter tops filled with saw dust bindings. They were maintained and acclimatized to laboratory environment for two weeks at a standard temperature of 20-to-25-degree Celsius with a 12-hour light/12-hour dark cycle. The animal were fed with

grower feed purchased from agro feed mills and water throughout the experiment period. All procedure followed in accordance with the ethical clearance from Bayero University, Kano Animal committee. The rats were randomly selected and used for this study. They were grouped into control group A and treatment groups B, C, and D.

Plant collection and extracts was purchased from the Rimi market Kano. Then it was taken to department of plant biology for identification. The department of pharmacy for extraction. 4L of tiger nut was macerated using ethanol to give the ethanolic tiger nut that was use in this experiment. The weight of the tiger nut after the extraction was 205.31g.

Study Design and Experimental Procedure

After the animals were acclimatized for two weeks, they were divided in to four groups on descending order of weight, each group has five rats. The groups are;

Group 1 - control group, received normal saline.

Group 2 – group, received liquid tiger nut extract (Cyperus esculentus) (C.E) 25% of LD50 daily.

Group 3 – received liquid tiger nut extract 50% of LD50 and aqueous aluminium chloride 500mg/kg daily.

Group 4 - received liquid tiger nut extract 75% of LD50 and aqueous aluminium chloride 500mg/kg daily.

Dose of the substances that was administered for each rat.

Dose of normal saline for group 1

Normal saline has no any case of toxicity; therefore, the selected dose for each rat is same as the stock.

$$\text{Dose} = \frac{\text{Selected dose} \times \text{weight of rat}}{\text{Stock}}$$

∴ The dose of normal saline for each rat in the control group is same as it is body weight in ml

Dose of aluminum chloride for groups 3 and 4

Aluminum chloride has reported to induced toxicity on the small intestine at 500mg [17].

∴ 500mg is our selected dose for each rat in the groups that $AlCl_3$ will be administered.

LD50 = 3470mg/kg

$$\text{Dose} = \frac{\text{Selected dose} \times \text{weight}}{\text{Stock}}$$

Selected dose = 500mg

Stock = 500ml/kg

∴ the dose of $AlCl_3$ for rat is base on its weight in ml.

Dose of tiger nut extract for group 2

Since there is no any case of toxicity associated with tiger nut, the LD50 of tiger nut can be above 5000ml/kg. 5000ml/kg is the LD50 of tiger nut used in this study.

Dose = Selected dose × weight
Stock

Selected dose = 25%LD50 = 1250mg/ml
Stock = 1000ml/kg

Dose of tiger nut extract for group 3
Dose = Selected dose × weight
Stock

Selected dose = 50%LD50 = 2500ml/kg
Stock = 1000ml/kg

3.6.4 Dose of tigernut extract for group 4

Dose = Selected dose × weight
Stock
Selected dose = 75%LD50 = 3750mg/ml
Stock = 1000ml/kg

Animal Sacrifice
Twelve rats three from each group were sacrificed. The rats were sacrificed at the last day of the experiment under chloroform anesthesia. A midline incision was done through the ventral abdominal wall and the small intestine tissue was collected immediately and fixed in 10% formal saline (fixative) for the minimum of 24 hours. The tissue was processed using routine histological techniques and stained with hematoxylin and eosin stains for general tissue architecture.

RESULTS AND DISCUSSION

Histomorphological observation in histology of small intestine
The small intestine tissue (duodenum) photomicrographs were stained with H&E staining process. Group (1) control that received normal saline and showed a normal morphological structure of the small intestine with intact structures of cylindrical villi, mucosa, intestinal crypt, goblet cell, then group (2) received 1250mg of tiger nut extract and also showed normal morphological structures of cylindrical villi, mucosa, goblet cells. But groups 3 and 4 that received 500mg of aluminium chloride each, and 2500mg/kg and 3750mg/kg of tigernut extract respectively, showed no any morphological changes but there is a mild aggregation of lymphocytes in the mucosa of group three (3) and high aggregation of lymphocytes in the mucosa of group four (4) indicating the inflammations in both groups as shown in following respective photomicrographs.

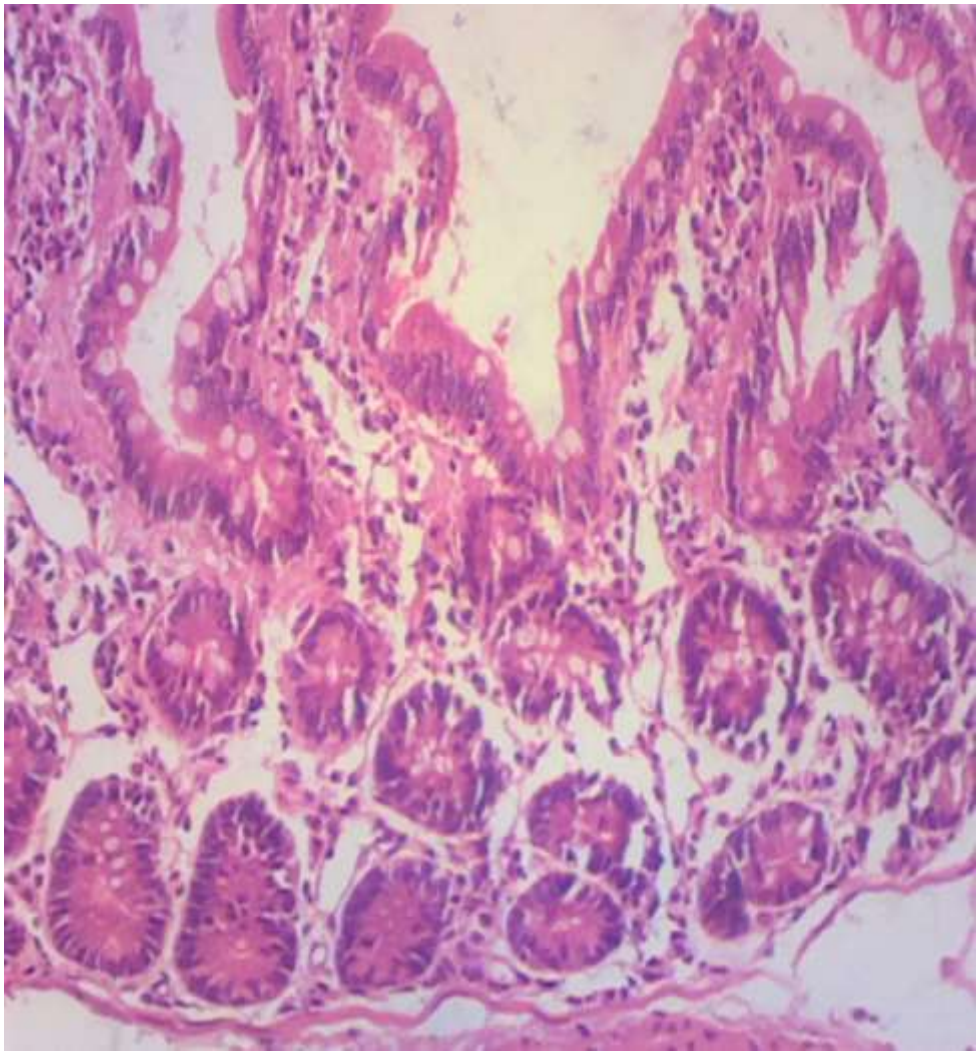

Plate 1: Photomicrograph of the small intestine (duodenum) in the group one (CONTROL) received normal saline showing the normal morphological structures of small intestine with many goblet cells, cylindrical vill, mucosa, and intestinal crypt, (H&E) 100x)

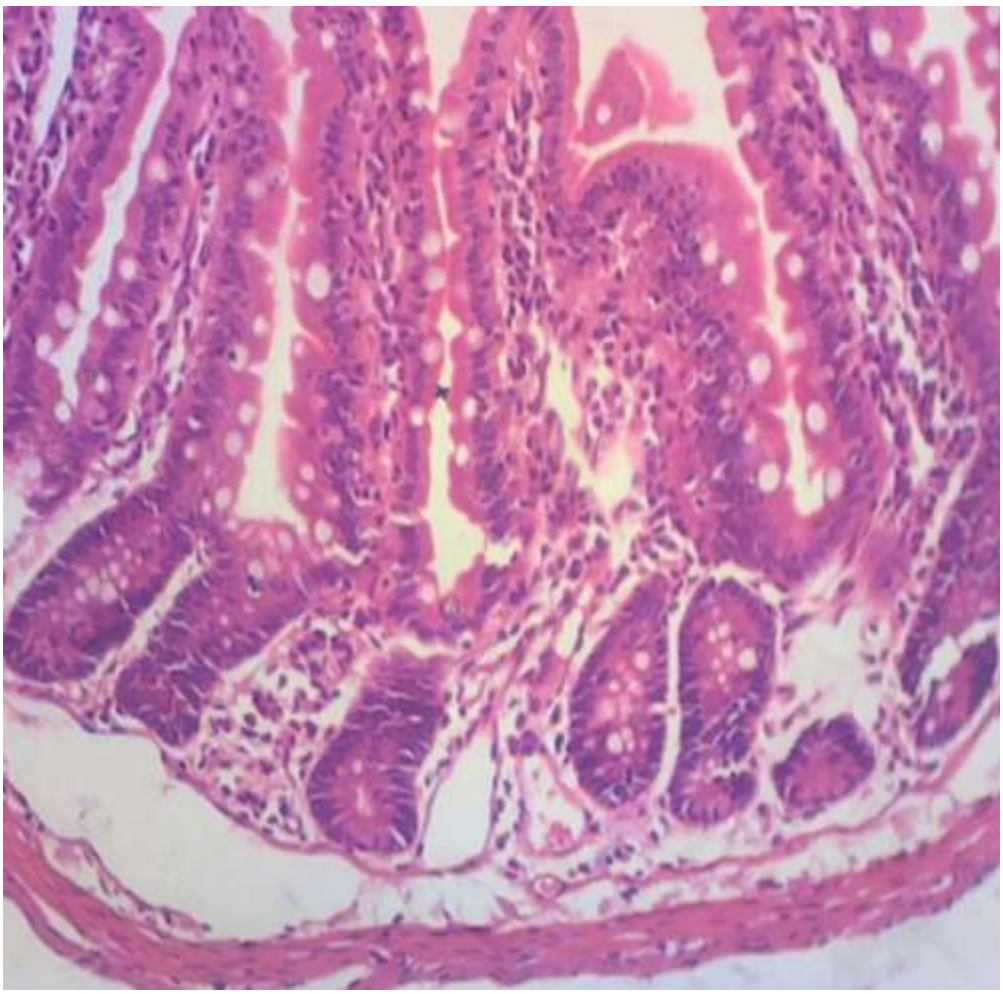

Plate II. Photomicrograph of the small intestine (duodenum) in group (2) received tiger nut extract only showing normal architectures of the small intestine with many goblet cells, cylindrical villi, mucosa and intestinal crypt. (H&E). 100x)

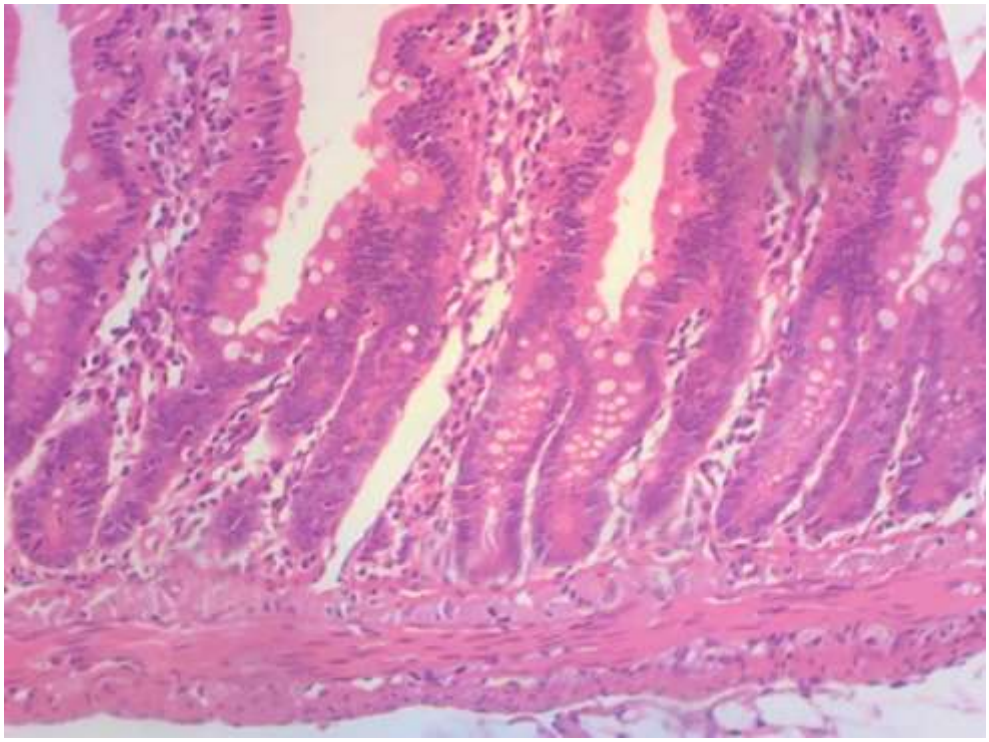

Plate III. Photomicrograph of the small intestine (duodenum) in group 3 received 500mg of aluminium chloride and followed immediately by 2500mg of tiger nut extract showed normal architecture with many goblet cell, cylindrical villi, submucosa, mucosa, intestinal crypt and mild proliferation of lymphocytes. (H&E) 100x)

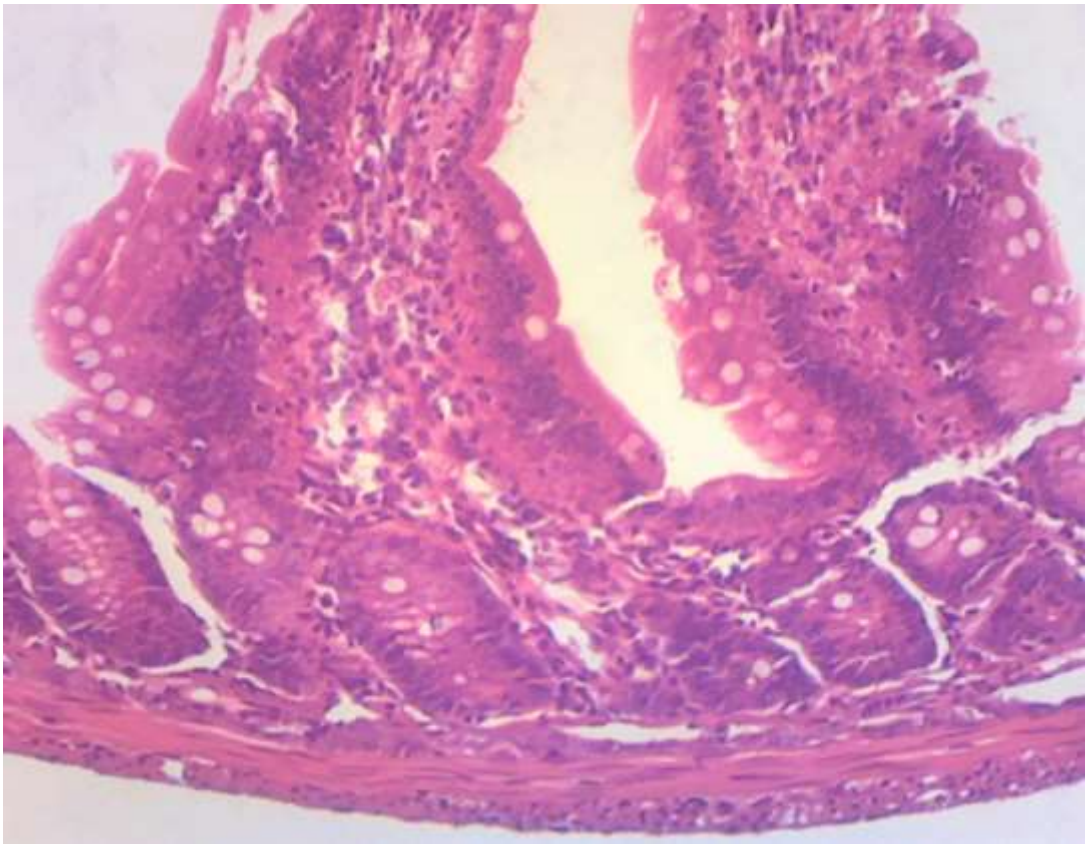

Plate IV. Photomicrograph of small intestine (duodenum) in group 4 received 500mg of aluminium chloride followed immediately by 3750mg of tiger nut extract, showed normal architecture with many goblet cell, cylindrical villi, mucosa, submucosa, intestinal crypt and high intensity of proliferation of lymphocytes (blue spot). (H&E) 100x).

DISCUSSION

According to [17], aluminium in the food supply comes from natural sources including water, food additives and contamination by aluminium utensils and containers. Oral exposure to aluminium is from food, water and pharmaceutical products. Role of Aluminium intoxication in different organs in neurodegenerative diseases has been recently emphasized [18]. There is little indication that aluminium is acutely toxic by oral exposure despite its widespread occurrence in foods, drinking-water, and many antacid preparations [19]. In 1988, a population of about 20,000 individuals in Camelford, England, was exposed for at least 5 days to unknown but increased levels of aluminium accidentally distributed to the population from a water supply facility using aluminium sulfate for treatment. Symptoms including nausea, vomiting, diarrhoea, mouth ulcers, skin ulcers, skin rashes, and arthritic pain were noted. It was concluded that the symptoms were mostly mild and short-lived. No lasting effects on health could be attributed to the known exposures from aluminium in the drinking-water [20].

In the present study the photomicrograph of group 1&2 that received normal saline and tiger nut extract

respectively showed normal architectural structures of the small intestine without any sign of inflammation, and group 3&4 that received aluminium chloride ($AlCl_3$) and tiger nut extract showed a normal architectural structure of wall of the small intestine but there are mild and intensive aggregation of the lymphocytes, which indicate the inflammation. Also indicating the ameliorative effect of the tiger nut induced histopathological changes on the small intestine.

CONCLUSION

Based on the observations from the present study, therefore it can be conclude that aluminium chloride exposure had negative effects on the wall of the small intestine of wistar rats and the tiger nut (Cyperus Esculentus) have the ameliorative effects on negative effects of the aluminium in the small intestine as e showed from the photomicrograph of the result of group 3&4 with aggregation of the lymphocytes In the mucosa of the small intestine.

The study had shown that the administration of the tiger nut over an extended period of time was able to ameliorate the histopathological changes induced by aluminium chloride on the intestine of wistar rat. Hence, tiger nut extract could be used as functional food for natural treatment and prevention of many intestinal diseases and associated abnormalities and may be a useful therapy in aluminium exposed patients.

REFFRENCES

1. Abdelkader H, Ibrahim F, Ahmed M, El-Ghadban E. Effect of Some Soil Additives and Mineral Nitrogen Fertilizer at Different Rates on Vegetative Growth, Tuber Yield and Fixed Oil of Tiger Nut (*Cyperus esculentus* L.) Plants. *Journal of Plant Production.* 2017; 8(1): 39–48.
2. Zohary D. The origin and early spread of agriculture in the Old World. In: Barigozzi C, editor. The origin and domestication of cultivated plants. Amsterdam: *Elsevier*. 1986; 3–20.
3. Achoribo ES, Ong MT. Tiger nut (*Cyperus esculentus*): Source of natural anticancer drug? Brief review of existing literature. *EuroMeditterrean Biomedical Journal*. 2017; 12: 91–94.
4. Maduka N, Ire SF. Tigernut Plant and Useful Application of Tigernut Tubers (*Cyperus esculentus*) - A Review. *Current Journal of Applied Science and Technology*. 2018; 29(3): 1–23.
5. Mohdaly A. Tiger Nut (*Cyperus esculentus* L.) Oil. In: Ramadan MF, editor. Fruit Oils: Chemistry and Functionality. Egypt: *Springer International Publishing*. 2019; 243-269.
6. Adejuyitan JA. Tigernut Processing: Its Food uses and Health Benefits. *American Journal of Food Technology*. 2011; 197-201.
7. Gambo A, Da'u A. Tiger nut (Cyperus esculentus): composition, products, uses and health benefits – a review. *Bayero Journal of Pure and Applied Science*. 2014; 7: 56-61.
8. Omode A, Fatoki O, Olaogun KA. Physico-chemial properties of some under-exploited and non-conventional oil seed. *Journal of Agricultural and Food Chemistry.* 1995; 11:50-53.
9. Sembulingam K, Sembulingam P. Small Intestine. Essentials of Medical Physiology. 5th edition. *Jaypee Brothers Medical Publishers Ltd*. 2010; 248-251.
10. Verstraeten SV, Aimo L, Oteiza PI. Aluminium and lead: molecular mechanisms of brain toxicity.

Archives of Toxicology. 2008; 82(11):789-802.
11. Martinez V. Scientific analysis of effects of tiger nut on heart diseases and related aspects In: Tiger Nut and Health. 2003.
12. Chevallier A. The Encyclopedia of medicinal plants. Dorling Kindersley Press London.1996: 48-51.
13. Shehla KF, Prabhavathi PA, Padmavathi P , Reddy, PP. *Mutation Research.* 2001; 490: 179-186.
14. Alfrey AC, Legendre OR, Kaehny WD. Dialysis encephalopathysyndrome. *North England Journal of Medicine*. 1976; 294:184—188.
15. Gorsky JE, DIetz AA, Spencer A, Osis D. Metabolic balance ofaluminum studied in six men. *Clin Chem*. 1979; 25:1739—1743.
16. Slanina P, Frech W, Ekstrom L, Loo FL. Dietary citric acid enhances absorption of aluminum in antacids. *Gun Chem*. 1986; 32:539—541.
17. Greger JL, Goetz W, Sullivan D. Aluminium levels in foods cooked and stored in aluminium pan, trays and foil. *J. Food Protection*. 1985; 48: 772-777.
18. Somova LI, Missankov A, Khan MS. Chronic Aluminium intoxication in rats: dose dependent morphological changes. *Methods Find Exp. Clin. Pharmacol.* 1997; 19: 599-604.
19. WHO. Aluminum. Geneva. World Health Organisation, International Programme on Chemical Safety. Environmental *Health Criteria*. 1997; 194.
20. Clayton DB. Water pollution at Lowermoore North Cornwall: Report of the Lowermoore incident health advisory committee. Truro, Cornwall District Heal. 1989.