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Protective Roles of *Adansonia digitata* (African Baobab), *Cucumeropsis mannii* (Melon), and *Abelmoschus esculentus* (Okro) Supplemented Diets against Cadmium-Induced Lipotoxicity, Bone Demineralization, and Cytotoxicity in Rabbits

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Abstract: The present study was undertaken to investigate the protective roles of *Adansonia digitata* (African baobab) *Cucumeropsis mannii* (Melon), *Abelmoschus esculentus* (Okro), on lipid profile, bone health, and selected organs (liver, kidney, brain, and testis) against cadmium-induced toxicity in rabbits. Twenty male rabbits were grouped into five with four rabbits in each group. Group 1 served as positive control and received standard feed and 1.5 mg/kg body weight normal saline, group 2 served as negative control and were exposed to 1.5 mg/kg body weight cadmium chloride, group 3 received 1.5 mg/kg body weight cadmium *Adansonia digitata* supplemented feed, group 4 received 1.5 mg/kg body weight cadmium chloride and *Citrullus lanatus* supplemented feed, while group 5 were given 1.5 mg/kg body weight cadmium chloride and *Abelmoschus esculentus* supplemented feed. All administrations were orally and lasted for 28 days. At the end of the administration, blood, liver, kidney, brain, and testes were harvested from the rabbits for biochemical and histological analysis. One-way analysis of variance followed by Turkey's test was used to analyze the results with $p < 0.05$ considered significant. The results revealed that cadmium exposure caused a significant increase in serum total cholesterol, triacylglycerol, low density lipoprotein cholesterol concentration, and atherogenic index in rabbits exposed to cadmium. Cadmium exposure also reduced bone calcium concentration. *Adansonia digitata* maintained the atherogenic index of plasma at 2.67 compared with the positive control group (2.66). The three vegetables reversed the cadmium-induced up-regulation of total cholesterol and triacylglycerol in the liver, kidney, and brain but not in the testis. All the vegetables also restored bone calcium. *Citrullus lanatus* and *Abelmoschus esculentus* prevented organ damage induced by cadmium exposure. These vegetables however play different protective roles against cadmium exposed rabbits.

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Chemicals have become an essential part of our daily life, they sustain the activities of all living organisms, and they prevent and control the pathogenesis of many disease endpoints. Chemicals endanger human health and reduce life expectancy, they destroy wildlife and change the ecosystem (Tchounwou *et al*., 2012). Cadmium (Cd) is found in the atmosphere, soil, water, and plants. It is also used in batteries, paint pigments (Martelli *et al*., 2006), cosmetics, cigarette smoke (IARC 1993), television screen, and together with zinc to weld seals in lead pipes and galvanizing steel (Angshuman *et al*., 2013). Cadmium toxicity depends majorly on the route of administration and the dose exposed. After exposure, cadmium has been found to induce toxic effects majorly on the various organ of the body system, which includes the lung, liver, kidney, brain, testis, placenta, bones, etc. (Angshuman *et al*., 2013). Similarly, studies have also implicated

cadmium exposure in the onset of pancreas, kidney, and liver dysfunction, dyslipidemia, osteoporosis, and the pathogenesis of cancer of multiple origins (Lin *et al* 2018; Zeng *et al*., 2004). The mechanisms of Cd-induced toxicity have been shown to include induction of oxidative stress leading to oxidative damage of biomolecules, interference with the essential trace elements of metabolism, and mediation of cell apoptosis (Lin *et al* 2018). The Cd-induced oxidative stress is a result of its ability to down-regulate the system's antioxidant defense mechanisms. This antioxidant system includes glutathione-S-transferase (GST), glutathione peroxidase (GPx), superoxide dismutase (SOD), catalase (CAT), glucose-6-phosphate dehydrogenase (G6PD), glutathione reductase (GR), and reduced glutathione (GSH). Cadmium has been shown to form a complex with selenium in GPx to inhibit its activity. Cd also binds sulfhydryl and replaces the Zn^{2+} or Mn^{2+} bound to SOD, reducing its antioxidant activity. Cd also decreases CAT levels which in turn inhibits CAT activity. Furthermore, reports from previous studies also implicated Cd in the depletion of cellular GSH levels (Ferramola *et al*., 2012; Balali-Mood *et al*., 2021). The down-regulation of the cellular antioxidant system is expected to result in the build-up of reactive oxygen species (ROS), or reactive nitrogen species (RNS), damaging the cellular biomolecules (lipid, DNA, and protein). Additionally, Cd alters the mitochondrial membrane and inhibits Ca^{2+}-ATPase to reduce membrane permeability altering energy production via oxidative phosphorylation, this also triggers the release of mitochondrial content into the cytoplasm resulting in the generation of more ROS (Lin *et al* 2018). Cd competes with biologically essential metals, such as calcium, zinc, and magnesium. After entering the cells, Cd mimics Zn^{2+}, $Ca^{2+,}$ and Mg^{2+} to activate or inhibit some enzymes and thus interferes with cell metabolism (Gong *et al*., 2014). Vegetables are normally grown in many countries across all the continents of the world where they serve as a major part of human diets (Rubatzky and Yamaguchi, 2012). Vegetables are natural sources of nutraceuticals. They play several beneficiary roles in the prevention and treatment of several disorders. They are found to be rich in antioxidants and are therefore natural sources of antioxidants (Ramya and Patel, 2019). Additionally, vegetables are also important sources of minerals, vitamins, and dietary fibers (Ülger *et al.,* 2018). As a result of these valuable roles of vegetables, researchers have intensified their research efforts on the potential roles of vegetables in the prevention and treatment of myriads of diseases, since they are considered to be safer and cheaper compared to synthetic drugs. The present research, therefore, beamed its searchlight on the possible roles of some common vegetables, *Abelmoschus esculentus* (Okro), *Cucumeropsis mannii* (Melon), and *Adansonia digitata* (African baobab) orally supplemented to diets against cadmium-induced toxicity in male rabbits.

MATERIALS AND METHODS

Chemicals and reagents: Cadmium chloride salt ($CdCl_2$), disodium hydrogen phosphate (Na_2HPO_4), sodium dihydrogen phosphate (NaH_2PO_4), Nitric acid (HNO_3), isopropyl alcohol are products of British Drug House (BDH) Chemicals Limited, Poole, England. All reagents were of analytical grade. Diagnostic kits for the determination of cholesterol and triacylglycerol, and HCL-C concentration were products of CYPRESS® Diagnostics, Langdorp, Belgium.

Animals: Twenty apparently healthy, male rabbits with an average weight of 1kg were purchased from a commercial breeder in Ogbomoso. The animals were kept in well-ventilated compartmentalized cages at the animal house of the Department of Biochemistry, LAUTECH, Ogbomoso, Nigeria. They were given free access to drinking water and standard feed *ad libitum*. They were allowed to acclimatize for 14 days before the commencement of the study. All efforts were put in place to decrease the suffering of the animals. The rabbits were randomly distributed into five (5) groups of four (4) rabbits each. Group 1 served as positive control and was fed with standard feed and administered a dose of 1.5mg/kg body weight normal saline (vehicle) orally for 28 days. Group 2 rabbits were given standard feed and administered a dose of 1.5mg/kg body weight of cadmium chloride orally for 28 days. Group 3 rabbits were treated with 1.5mg/kg body weight of cadmium chloride and *Adansonia digitata* (Baobab) supplemented feed orally for 28 days. Group 4 rabbits were exposed to 1.5mg/kg body weight of cadmium chloride and *Cucumeropsis mannii* (Melon) supplemented feed for 28 days, while group 5 rabbits were given 1.5mg/kg body weight of cadmium chloride and *Abelmoschus esculentus* (OKRO) supplemented feed orally for 28 days (Sajjad *et al.,* 2014).

Sacrificing of experimental animals and collection of tissues/organs: At the end of the administration, blood was collected from the animals through the cardiac puncture into plain tubes under light ether anesthesia after an overnight fast. The liver, kidney, brain, and testis were excised from the rabbits for biochemical and histological studies. The blood samples were centrifuged at 5000 rpm for 10 min to separate serum from the red blood cells. All samples were stored at −20°C until analyzed.

Table 1: Formulation of the feed

	Standard feed(kg)	Baobab supplemented diet (kg)	Melon supplemented diet (kg)	Okro supplemented diet (kg)
Maize	7.59	5.89	5.89	5.89
Soya bean	1.5	1.5	1.5	1.5
Wheat bran	2.0	2.0	2.0	2.0
Rice bran	5.0	5.0	5.0	5.0
Oyster shell	0.4	0.4	0.4	0.4
Bone meal	0.2	0.2	0.2	0.2
Common salt	0.052	0.052	0.052	0.052
methionine	0.2	0.2	0.2	0.2
Fish meal	0.2	0.2	0.2	0.2
Vegetable supplement	--	1.7	1.7	1.7
Total	**17.14**	**17.14**	**17.14**	**17.14**

10% of the maize was removed and replaced with vegetable supplements

Determination of serum total cholesterol and high density lipoprotein (HDL)-cholesterol concentration: Serum total cholesterol and HDL-C concentration were determined spectrophotometrically according to the method of Meiattini *et al.* (1978), using the Cypress® diagnostic kit manual. Briefly, 1ml of the working reagent (4-aminoantipyrine, cholesterol oxidase, cholesterol esterase peroxidase, phenol, and buffer) was pipetted into each of the blank, standard, and sample test tubes. 10 µL of the standard was then added to the test tube for standard, while 10 µL of the serum was added to the test tubes for the respective samples. 10 µL of distilled water was added to the test tube for blank. The reaction mixture was thoroughly mixed and incubated for 10 minutes in the dark at 37°C. The absorbance (A) of the samples and standard was then measured against the blank at a wavelength of 505 nm. Cholesterol concentration in plasma (mg/dL) was calculated as:

$$ChC\ P = \frac{Absorbance\ of\ sample}{Absorbance\ of\ standard} \times 200$$

Where: ChC P= cholestrerol concentration: 200 = concentration of standard in mg/dL

Determination of serum triacylglycerol concentration: Serum triacylglycerol concentrations were determined spectrophotometrically according to the method of Buccolo and David (1973), using the Cypress® diagnostic kit. Briefly, three test tubes marked blank (B), standard (S), and test (T) were arranged in a test-tube rack. To each of these three tubes was added 1.0ml of the working reagent (50 mmol/l PIPES buffer, pH 6.8; 2 mmol/l p-Chlorophenol, 15 KU/L LPL, 500 U/L GK, 2500 U/L GPO, 0.1 mmol/L ATP, 40 mmol/L Mg^{2+}, 440 U/L Peroxidase, 0.1 mmol/L 4-Aminophenenazone). 10 µL of distilled water, Standard (200 mg/dL glycerol trioleate), and serum were added separately to tubes B, S, and T respectively. The reaction mixtures were incubated at room temperature for 20 min in the dark. The absorbance of the samples and the standard were read at 505 nm against the reagent blank.

Triacylglycerol concentration (mg/dL) was calculated as:

$$TAGC\ \ \frac{Absorbance\ of\ sample}{Absorbance\ of\ standard} \times 200$$

Where : TAGC= triacypglycerol concentration; 200 = concentration of standard in mg/dL

Extraction of lipids from organs: Lipids were extracted from the organs (liver, kidney, brain, and testis) using the method described by Folch *et al.* (1957). A 10% organ homogenate was prepared by homogenizing 0.2 g of the tissue in 1.8 ml of chloroform: methanol (2:1, v/v) mixture with 6 up and down strokes at 1100 rpm using a power-driven, Teflon pestle in a glass homogenizing cup maintained at 4ºC. The homogenate was then centrifuged at 5000 rpm at 4ºC after which the supernatant was taken and 0.1ml of 0.05 M KCl added to it. This was vortexed again and allowed to stand on ice for 5 minutes. The mixture was then centrifuged again and the chloroform (lower) layer was taken into clean Eppendorf tubes and stored at -20ºC for further analysis.

Determination of liver, kidney, brain, and testis cholesterol concentration: One hundred microlitres (100 µL) of the liver, kidney, brain, and testis lipid extract was evaporated to dryness at 60ºC. 20 µL of Triton X-100-chloroform mixture (1:1, v/v) was added, vortexed and again evaporated to dryness. 1ml of cholesterol reagent was added, vortexed, and incubated in the dark at room temperature for 20 minutes. The absorbance was then read at 505 nm (Kriketos *et al*., 2003).

$$ChC = \frac{Abs\ sample}{Abs\ standard} \times 200 \times \frac{TV}{100} \div wet\ wt$$

Where ChC = cholesterol concentration (mg/g); Abs Sample absorbance of sample; Abs Standard = absorbance of standard; TV= total volume of extract in mL; 100 = 100 ml; Wet wt = wet weight of the organ

Determination of liver, kidney, brain, and testis triacylglycerol concentration: An aliquot (100 µL) of the liver, kidney, brain, and testis lipid extract was evaporated to dryness. After cooling, 100 µL of 97% ethanol was added to the dried extract and vortexed. 1ml of triacylglycerol reagent was added and vortexed. The mixture was incubated at room temperature in the dark for 20minutes. The absorbance was then read at 505 nm (Kriketos *et al*., 2003).

$$TAGC = \frac{Absorbance\ of\ sample}{Absorbance\ of\ standard} \times 200 \times \frac{TV}{100} \div wet\ wt$$

Where: TV= total volume of extract in mL; TAGC= triacypglycerol concentration; 200 = concentration of standard in mg/dL; 100 = 100 ml; Wet wt = wet weight of the organ

Low density lipoprotein (LDL) cholesterol concentration was calculated using the FRIELDWALD formula Friedewal *et al*. (1972) as follows

$$LDL - C = TC - \left[\left(\frac{TG}{5}\right) + HDL\right]$$

The atherogenic index (AI) was calculated as follows

$$AI = \frac{LDL}{HDL}$$

Bone mineral determination: The tibia was removed from each rabbit, rinsed in normal saline and all the adjoining tissues were carefully removed. One gram (1 g) of each of the tibia was digested in 10 ml of concentrated nitric acid and heated within the temperature range of 60 and 65°C until fully digested and diluted with 10 ml of distilled water. The digested bones were then analyzed for calcium, magnesium, and phosphorus concentration using atomic absorption spectrophotometer at 422.7 nm for calcium, 285.2 nm for magnesium, and 167.1 nm for phosphorus.

Histological studies: For qualitative analyses of liver, kidney, testis, and brain histology, the tissues samples were fixed for 48 hours in 10 % formalin-saline and dehydrated by passing successfully in a different mixture of ethyl alcohol, water, cleaned in xylene, and embedded in paraffin. Sections of the tissues were prepared by using a rotatory microtome and stained with hematoxylin and eosin dye, which was mounted in a neutral deparaffined xylene medium for microscopic observations. One rabbit was selected in each group for histological examinations.

Statistical analysis: Statistical analysis was conducted with the statistical package for social science (SPSS). Results were expressed as mean±SD. Group differences were tested for statistical significance by using a repeated-measures analysis of variance. The level of significance was set up at $p < 0.05$.

RESULT AND DISCUSSION

Table 2 shows the effects of exposure to cadmium, baobab, melon and okro (groups 2,3,4 and 5 respectively) on serum lipid profile. Exposure to cadmium (group 2) caused elevated serum concentrations of total cholesterol, triglyceride, LDL-C, VLDL-C and atherogenic index. Also, cadmium caused reduced serum concentration of HDL-C. However, Only administration of baobab (group 3) led to appreciable reversal of the toxic effects of cadmium on concentrations of total cholesterol, triglyceride, LDL-C, HDL-C, VLDL-c and atherogenic index. Table 3 shows effects of exposure to cadmium, baobab, melon and okro on concentrations of total cholesterol in liver, kidney, brain and testis of experimental rats.

Exposure to cadmium (group 2) caused elevation in concentration of total cholesterol in liver, kidney, and testis. However, administration of baobab (group 3), melon (group 4), and okro (group 5) led to appreciable reversal of the effect of cadmium on total cholesterol concentration in liver, kidney and brain. Table 4 shows effects of exposure to cadmium, baobab, melon and okro on concentrations of triglyceride in liver, kidney, brain, and testis. Administration of baobab, melon and okro reversed the toxic effects of cadmium on triglyceride levels in kidney, brain and testis of the experimental rats.

Table 2: Effects of vegetable supplemented diets on serum lipid profile.

Lipid parameters	Total cholesterol concentration	Triacylglycerol concentration	HDL-C concentration	LDL-C concentration	VLDL-C concentration	Atherogenic index
Group 1	176 ± 16.98	198.89 ± 23.42	39.91 ± 3.98	105.78 ± 8.39	39.78 ± 4.68	2.66 ± 0.18
Group 2	186.96 ± 21.33	195.75 ± 17.48	*33.27 ± 1.00	114.05 ± 19.12	39.15 ± 3.50	3.45 ± 0.59
Group 3	145.74 ± 18.56	128.66 ± 14.79	33.85 ± 1.57	92.30 ± 24.26	25.73 ± 2.96	2.67 ± 0.79
Group 4	*211.83 ± 15.45	*a152.67 ± 60.51	*32.49 ± 1.45	148.81 ± 12.78	30.54 ± 12.10	*a4.57 ± 0.20
Group 5	183.30 ± 31.69	179.30 ± 37.38	32.37 ± 0.27	143.81 ± 21.69	35.86 ± 7.48	4.43 ± 0.66

Each value is the mean ± SD of four animals in each group

Table 3: Concentrations of total cholesterol in the liver, kidney, brain, and testis of the experimental animals.

Groups	Liver	Kidney	Brain	Testis
Group 1	233.97 ± 48.45	90.96 ± 10.22	64.48 ± 14.35	47.68 ± 3.20
Group 2	272.13 ± 77.55	98.05 ± 66.28	58.37 ± 2.78	82.16 ± 2.84
Group 3	242.44 ± 23.76	75.68 ± 49.35	46.78 ± 12.74	101.04 ± 38.47
Group 4	245.79 ± 37.38	55.37 ± 12.33	50.32 ± 12.94	104.68 v 46.47
Group 5	210. 40 ± 30.00	62.70 ± 16.77	51.41 ± 12.01	108.55 ± 41.38

Each value is mean ± SD of four animals in each group

Table 4: Concentrations of Triglycerides in the liver, kidney, brain, and testis of the experimental animals.

Groups	Liver	Kidney	Brain	Testis
Group 1	110.66 ± 24.80	247.41 ± 11.36	233.97 ± 48.57	194.82 ± 38.99
Group 2	102.32 v 8.31	272.05 ± 43.50	272.14 ± 77.55	395.49 ± 11.04
Group 3	76.87 ± 28.38	75.68 ± 49.35	46.78 ± 12.74	101.04 ± 38.47
Group 4	103.94 ± 7.26	182.73 ± 61.98	245.79 ± 37.38	272.94 ± 14.46
Group 5	120.40 ± 15.25	141.28 ± 11.33	210.40 ± 30.00	150.78 ± 47.65

Each value is mean ± SD of four animals in each group

Table 5 shows the effects of cadmium, baobab, melon and okro on the concentrations of calcium, magnesium and phosphorus in tibia bone of the experimental rabbits. Exposure to cadmium caused reduced concentration of phosphorus in tibia bone of the cadmium-exposed rabbits. Administrationof baobab, melon and okro reversed the adverse effects on calcium and magnesium concentrations in the tibia bone. Evidence from previous studies revealed that cadmium exerts its toxic effects on multiple organs of the body, damaging all these vital organs (Bernhoft, 2013). Humans are exposed to cadmium through several routes some of which include occupational, diet, and other anthropogenic activities. Meanwhile, vegetables contain antioxidants, minerals, vitamins, fibers, and essential bioactive compounds that can ameliorate the damaging effect of cadmium exposure. In the present study, cadmium exposure led to the induction of lipotoxicity in almost all the compartments tested. Meanwhile, the administration of the three types of vegetables used in this study effectively reversed the lipotoxic effects induced by cadmium. Hypercholesterolemia, hypertriglyceridemia, elevated LDL-C levels, and diminished LDL-C are known cardiovascular disease (CVD) risk factors among the human population. Similarly, compelling experimental data from animal studies have implicated high serum total cholesterol (TC) concentration, high LDL-C, and low HDL-C as major risk factors in the onset of CVD (Noh *et al*., 2022; Rizvi, *et al*., 2021). Additionally, studies have shown a strong positive association between elevated TC to HDL-C ratio (TC/HDL-C) and pathogenesis of left ventricular hypertrophy, ischemic stroke, and coronary heart disease (CHD) (Zhang, *et al*., 2019). In this study, cadmium exposure led to a significant elevation of serum TC, LDL-C, VLDL-C, and concomitantly reduced HDL-C compared with the positive control. This observed cadmium-induced hyperlipidemia was reversed by the vegetable supplemented diets, indicating the possible antihyperlipidaemic roles of these three vegetables.

Table 5: Concentration of calcium, magnesium, and phosphorus in the bone of rabbits in the different experimental groups

Groups	Calcium (mg/g)	Magnesium (mg/g)	Phosphorus (mg/g)
Group 1	1.56 ± 0.48	1.33 ± 0.30	0.033 ± 0.01
Group 2	1.21 ± 0.53	0.93 ± 0.30	0.68 ± 0.68
Group 3	2.03 ± 1.37	1.97 ± 1.05	2.03 ± 1.37
Group 4	1.94 ± 0.77	0.68 ± 0.68	1.48 ± 0.50
Group 5	2.60 ± 0.95	1.97 ± 1.05	0.068 ± 0.03

Each value is the mean ± SD of four animals in each group

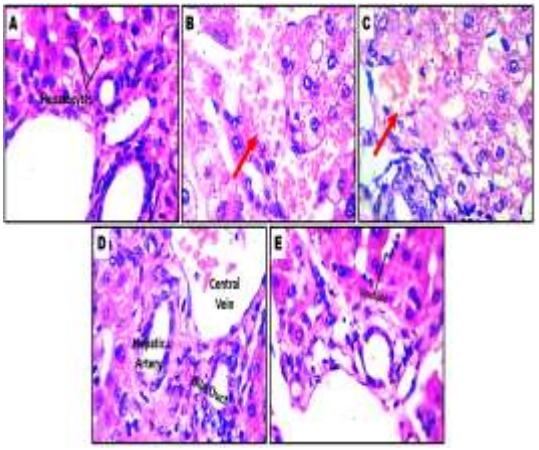

Fig 1: Photomicrograph of the hepatic cytoarchitecture of experimental animals (H&E, x40).

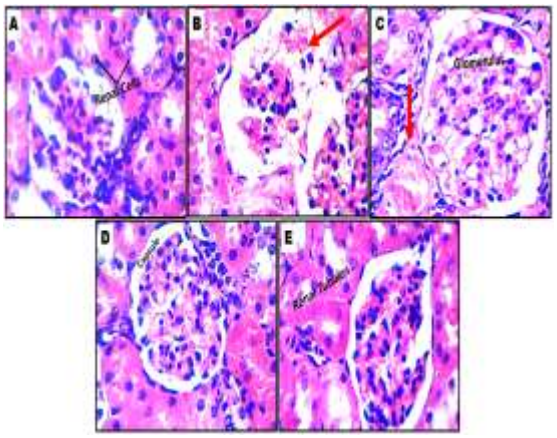

Fig 2: Photomicrograph of the renal cytoarchitecture of experimental animals (H&E, x40)

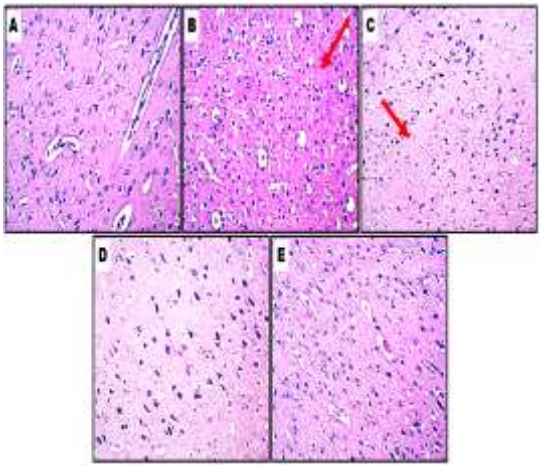

Fig 3: Photomicrograph of the brain cytoarchitecture of experimental animals (H&E, x40)

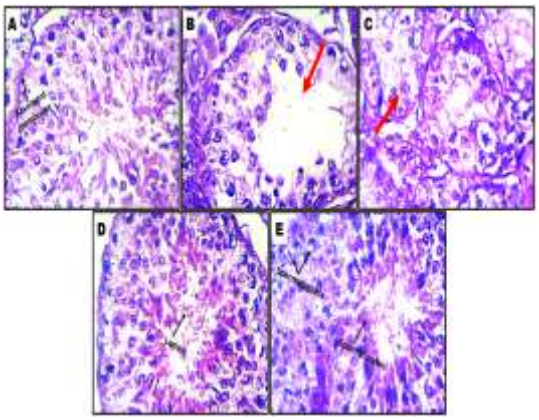

Fig 4: Photomicrograph of the testis cytoarchitecture of experimental animals (H&E, x40)

A high atherogenic index (AI) is a marker of dyslipidemia, atherosclerosis, CVD, and type II diabetes. These conditions arise from increased activity of HMG COA Reductase and damage to lipid and its receptors (Prabu *et al*., 2010). Vegetables are rich in flavonoids (Ogbaga, *et al*., 2017) which prevent the oxidation of LDL-C, enhancing LDL-C and its receptor interaction, thus facilitating the uptake of cholesterol from the blood and inducing the activity of LCAT (Tobar *et al*., 2019; Ochiai *et al*., 2016). Data obtained from this study also revealed that cadmium exposure elevated the cholesterol concentration in the liver, kidney, and testis of the cadmium-exposed animals. This might be due to cadmium-induced enhancement of 3-hydroxyl-3-methylglutaryl Coenzyme A reductase (HMG-CoA reductase) activity. HMG-CoA reductase is the rate-limiting enzyme in cholesterol biosynthesis. It may also be due to cadmium-induced inhibition of cholesterol-7α-hydroxylase activity, thereby limiting biosynthesis of bile acid from cholesterol, a major route of cholesterol elimination from the body (Afolabi, *et al*., 2015). Meanwhile, vegetable supplemented diets used in the present study significantly prevented the cadmium-induced tissue lipotoxicity as evident in the significant decrease in cholesterol concentration in the liver, kidney, brain, and testis of the rabbits that were fed with the three vegetables (*Adansonia digitata* (Baobab), *Cucumeropsis mannii* (Melon), and *Abelmoschus esculentus* (OKRO)) supplemented diet when compared with both positive and negative control. Additionally, the elevated triacylglycerol levels in the organs (liver, kidney, brain, and testis) of cadmium-exposed rabbits were upturned in the organs of the rabbits that were co-treated with vegetables supplemented diet. That is, *Adansonia digitata*, (Baobab) *cucumeropsis mannii* (Melon) and *Abelmoschus esculentus* (Okro) cause a considerable reduction in the amount of total cholesterol and triglycerides in the liver, kidney, brain, and testis. In bone, *Adansonia digitata*, (Baobab) *Cucumeropsis mannii* (Melon), and *Abelmoschus esculentus* (Okro) restore the bone calcium concentration reduced by cadmium in rabbits, possibly by improving the antioxidant defense in osteocytes, directly chelating/inactivation of cadmium, improving kidney function (reabsorption of minerals and activation of vitamin D) or improvement of the parathyroid gland. It may also be via directly replenishing the bone calcium content (Ivanova, 2012). *Adansonia digitata* is an excellent source of calcium, iron, potassium, magnesium, manganese, phosphorus, and zinc (Muthai *et al*., 2017). *Adansonia digitata*, (Baobab) and *Abelmoschus esculentus* (Okro) as shown in this study restore the bone magnesium compared to *Cucumeropsis mannii* (Melon) is perhaps because of the low concentration of magnesium present in *Cucumeropsis mannii* (Melon) compared with *Adansonia digitata*, (Baobab) and *Abelmoschus esculentus* (Okro). *Adansonia digitata*, (Baobab) and *Abelmoschus esculentus* (Okro) improve the bone phosphorous compared with positive control and cadmium treated rabbits. Histological investigations play a sensitive role in the confirmation of pathological derangements at the tissue level in response to exposure to toxic substances (Owonikoko *et al*., 2021). It provides valuable information as to what extent is the level of damage induced by the exposed toxicant (Lanning *et al*., 2002). In this study Liver section of rabbit in the positive control group showed a normal arrangement of hepatocytes and central vain, Cadmium treated group (B) showed periportal inflammation, degeneration of hepatocytes, and necrosis, Cadmium with baobab supplemented diet (C) shows mildly pyknotic, Cadmium with melon supplemented diet (D) showed normal central venules without significant observable congestion, while the group that received cadmium supplemented with okro (E) showed normal

central venules with a mild significant observable congestion. Furthermore, the Kidney section of the rabbit in the positive control group (A) showed normal architecture, the renal cortex showed normal glomeruli with normal mesangial cells and capsular spaces, on the other hand, the renal sections of the animal in the cadmium-treated group (B) revealed severe degenerative changes, characterized by congested renal tubules (proximal and distal convoluted tubules), infiltrated renal parenchyma by red inflammatory cells, dilated renal tubules, signs of glomerulosclerosis and glomerulonephritis, the group that was administered cadmium with baobab supplemented diet (C) showed only slight degenerative changes. Meanwhile, the kidney section of the group that was exposed to Cadmium co-treated with melon (D) revealed normal renal architecture, the renal tubules appear normal, clear, and not congested, while the kidney section of the animals that received cadmium with okro supplemented diet (E) also showed normal renal histoarchitecture, the interstitial spaces also appear normal with a well-defined profile. Additionally, the brain sections of the animals in the positive control as observed in this study (A) showed intact perineural space surrounding, nuclear and cytoplasmic content. , while that od cadmium-exposed group (B) revealed severe conspicuous degenerative changes in the brain characterized by peripheral and central degenerative changes, clustered pyknotic pyramidal neurons that appear with fragmented cytoplasm and condensed nuclei within soma (red arrow). On the other hand, the histoarchitecture of the group that was exposed to cadmium and baobab supplemented diet (C) showed only mild conspicuous degenerative changes in the brain. The brain section of the rabbits that received cadmium (E) with an okro diet showed very mild micromorphological alteration. Testis section of the rabbit in the positive control group (A) is presented with a normal arrangement of spermatocytes, cadmium exposure (B) led to micromorphological alterations characterized by observable interstitial space distortion (widen space), pyknotic interstitial cells, degenerating spermatogia cell nuclei, and diminished basement membrane appear. Cadmium exposure co-treated with baobab diet (B) caused a slight micromorphological alteration, testis of the animals that received cadmium co-treated with melon diet (D) revealed the almost normal appearance of spermatocytes, while administration of cadmium with okro diet (E) showed very mild micromorphological alteration.

Conclusion: In this study, we investigated the protective roles of *Adansonia digitata (*African Baobab), *Cucumeropsis mannii* (Melon), and *Abelmoschus esculentus* (Okro) against the cadmium-induced negative effects in rabbits. The data obtained from the study revealed that the three vegetable supplements possessed the protective potential against cadmium-induced lipotoxicity, bone demineralization, and cytotoxicity.

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