Published by SET Publisher

Journal of Pharmacy and Nutrition Sciences

ISSN (online): 1927-5951

Protective Effect of Curcuminoids Consumption on Cadmium-Induced Testicular Injury in Albino Rats

Amusan A. Timothy[1,2] and Emokpae A. Mathias[1,*]

[1]*Department of Medical Laboratory Science, School of Basic Medical Science, University of Benin, Benin City, Edo State, Nigeria*

[2]*Veterinary Teaching Hospital, Federal University of Agriculture, Abeokuta, Ogun State, Nigeria*

Article Info:

Keywords:
Cadmium,
curcuminoids,
antioxidant,
semen analysis,
Albino rats.

Timeline:
Received: November 20, 2023
Accepted: December 06, 2023
Published: December 30, 2023

Citation: Amusan TA, Emokpae MA. Protective effect of curcuminoids consumption on cadmium-induced testicular injury in albino rats. J Pharm Nutr Sci 2023; 13: 57-67.

Abstract:

This study evaluated the protective effects of curcuminoids against cadmium-induced testicular injury in Albino rats. Male albino rats were divided into nine groups, group 1 (control), groups 2-4 received 120 mg/kg, 240 mg/kg, and 360 mg/kg curcuminoids daily for 28 days without testicular injury. Group 5 received 20 mg/kg cadmium chloride (CdCl2) solution every other day for 28 days (positive control), while group 6 received 20mg/kg CdCl2 + 240 mg/kg curcuminoids every other day for 28 days. Group 7 received 20mg/kg CdCl2 every other day for 28 days, then treated with 240 mg/kg for 14 days. Group 8 received 20 mg/kg CdCl2 solution every other day for 28 days, then treated with 240 mg curcuminoids for 28 days. Group 9 received 20 mg/kg CdCl2 solution every other day for 28 days and left to recover 28 days. Serum and seminal plasma malondialdehyde, catalase, superoxide dismutase, glutathione peroxidase, and glutathione reductase were assayed using Spectrophotometry technique. Semen analysis was determined microscopically. Sperm motility and count were significantly reduced, acrosome defect and percent abnormal sperm morphology were increased among the positive control group when compared with negative control group ($p<0.05$). Serum GPX, GSH, and SOD, semen GPx , GSH and SOD were significantly reduced ($p<0.001$), while MDA was significantly increased ($p<0.001$) in CdCl2 administered rats than negative control. The supplementation of Curcumins resulted in the improvement of sperm quality indices in a dose-dependent manner. Curcumin supplementation may significantly reverse the adverse effects of cadmium chloride testicular injury.

DOI: https://doi.org/10.29169/1927-5951.2023.13.06

*Corresponding Author
E-mail: mathias.emokpae@uniben.edu

INTRODUCTION

Toxic metal contamination is increasingly being implicated as a reproductive hazard and this toxic metals determination is routinely investigated [1]. Cadmium can act as an endocrine disruptor and is recognized as metallohormone that has opposing actions against steroid hormones and causes adverse effects on testicular function. The declining male fertility in sub-Saharan Africa has been associated, among other factors, with toxic metal exposure [2]. Cadmium (Cd) in particular is a widespread heavy metal and a ubiquitous environmental toxicant. The major sources of Cd exposure in the general population include cigarette smoking, air pollution, eating contaminated foods, and drinking water. However, individuals working in battery manufacturing industries, mines, and paints containing Cd could have occupational exposure. Current evidence indicates an association between lifestyle habits and male fertility, and the potential of nutraceutical supplementation as a therapeutic strategy to prevent and/or ameliorate Cd-induced testicular injury has been emphasized [3]. The testicular injury caused by Cd may be via several specific mechanisms including oxidative stress, inflammation, and apoptosis [4]. Due to the absence of specific therapy for the prevention and treatment of perturbations caused by Cd exposure, the need for novel therapeutic agents is urgently desired. Therefore, a dietary master plan and the use of nutraceuticals rich in antioxidants, grains, vegetables, and certain oils are suggested for the prevention of Cd-induced testicular injury [5,6]. Nutraceuticals are defined as "foods (or part of a food) that provide medical or health benefits, including the prevention and/or treatment of a disease" [7].

Curcuminoids (Curcumins) are a group of active compounds within turmeric. These are polyphenolic compounds and include curcumin, demethoxy-curcumin, and bisdemethoxycurcumin [8]. Out of these, Curcumin has a wide range of pharmacological and biological activities [9] with powerful radical-scavenging antioxidant properties and increases intracellular glutathione [10]. Curcumin can also reduce lipid peroxidation by enhancing the activities of antioxidant enzymes.

The three groups of enzymes that play significant roles as oxidant scavengers are superoxide dismutases, catalase, and glutathione peroxidase [11]. Superoxide dismutases (SOD) are metal-containing enzymes that catalyze the conversion of two superoxides into oxygen and hydrogen peroxide, which is less toxic than superoxide. Catalase, an enzyme found in peroxisomes, degrades hydrogen peroxide to water and oxygen, thereby completing the reaction started by SOD. Glutathione peroxidase also acts to degrade hydrogen peroxide. Other enzymes, such as glutathione transferase, ceruloplasmin, or hemoxygenase may also participate in enzymatic control of oxygen radicals and their products [12].

Research Objectives

This study aimed to evaluate the protective effects of curcuminoids against cadmium-induced testicular injury in Albino rats.

MATERIAL AND METHODS

Animals

Forty-five healthy mature male albino rats weighing 160 ± 10 g were procured from the College of Veterinary Medicine, Federal University of Agriculture, Abeokuta, Ogun state, Nigeria. They were randomly selected into nine groups of five rats per group (n=5). They were housed inside plastic cages containing wood shavings for bedding, and were preserved under regulated conditions: 12 h light, 23 ± 1 °C, and 12 h dark cycle. Rats were fed with standard laboratory pelleted feed and accommodated for 14 days before the commencement of the research. Cadmium chloride monohydrate ($CdCl_2$ H_2O) was applied to induce testicular injury.

Chemicals

Cadmium chloride monohydrate ($CdCl_2$ H_2O) (LOBA CHEMIE PVT. LTD, 107, Wodehouse Rd, near Bank of India, Cuffe Parade, Mumbai, Maharashtra 400005, India) was used in this experiment.

Turmeric Extraction Procedure

Curcumin contents of dry turmeric, 0.2 g of ground dry turmeric were weighed in a 15 mL screw-cap test tube, and 5 mL ethanol was added. The mixture was vortexed every 15 mins for 2 hours and centrifuged at 4,500 rpm/min for 10 mins at room temperature. The clear supernatant was collected in a 25 mL volumetric flask. The extraction was repeated until a pale yellow supernatant deposit solution was observed.

Experimental Design

The experimental rats were randomly divided into nine groups (5 rats per each). The turmeric extract and

cadmium chloride solution were administered orally via an oral gastric tube.

- Group 1 was fed with feeds and water *ad libitum* (negative control).
- Groups 2-4 were administered 120 mg/kg, 240 mg/kg, and 360 mg/kg curcumin daily for 28 days.
- Group 5 was administered with 20 mg/kg cadmium chloride (CdCl2) solution every other day for 28 days (positive control).
- Group 6 was administered with 20 mg/kg CdCl2 and 240 mg/kg curcumin treatment simultaneously every other day for 28 days.
- Group 7 was administered with 20 mg/kg CdCl2 every other day for 28 days, then treated with 240 mg/kg for 14 days.
- Group 8 was administered 20 mg/kg CdCl2 solution every other day for 28 days, then treated with 240 mg/Kg Curcumin for 28 days.
- Group 9 was administered with 20 mg/kg CdCl2 solution every other day for 28 days and left to recover without treatment with Curcumins for another 28 days.

Samples Collection

5mL of whole blood sample was collected from each of the rats aseptically by cardiac puncture into vacutainer plain tubes and EDTA bottles. The blood in the plain bottle was allowed to clot, retracted, and thereafter centrifuged at 4000 rpm for 10 minutes. The serum was separated into a vacutainer plain tube and stored at -40 ˚C for 2 weeks before biochemical analyses were done.

Semen samples were collected by open castration method (Orchidectomy). The rats were sedated using ether and subsequently sacrificed by cervical dislocation. A midline or pre-scrotal incision was created and the testicles were milked out of the incision spot. The testicles and spermatic cord were opened by incising the tunica vaginalis. Semen samples were thereafter collected from the cauda epididymis and analyzed immediately. Testes were harvested and kept in 10% neutral buffered formalin for histomorphological examination.

Evaluation of Sperm Indices

Epididymis from each rat was harvested the caudal portion was gently pressed and seminal content was extracted and diluted with 1 mL of normal saline. Sperm count was done microscopically using the newly improved Neubauer counting chamber. Sperm morphology, sperm viability, and non-viability were evaluated by applying the same volumes of semen and eosin-nigrosin stain (one drop each). A thin film slide was made from the mixture of the semen sample and eosin-nigrosin stain thereafter observed with a light microscope at (40x). Eosin is a differential stain that has an affinity to stain the head of dead sperms with red, while nigrosin stain is applied for background staining. Live and dead sperms were recorded in percentage and sperm abnormalities were also evaluated 13.

Determinations of Biochemical Parameters

Both serum and semen malondialdehyde, glutathione peroxidase, catalase, superoxide dismutase, and glutathione reductase activities were estimated by spectrophotometry method [14-18]

Histopathological Examination

The left and right testes were harvested and preserved in 10% neutral buffered formalin and processed. Histomorphological slides were prepared from sectioned specimens and stained with hematoxylin and eosin (H&E). It was examined using a light microscope [19].

Ethical Consideration

The study was conducted by the regulatory standard for the precaution and usage of laboratory animals of the National Institutes of Health (NIH) and the research procedure was approved by the College of Veterinary Medicine Research Ethics Committee (CREC), Federal University of Agriculture, Abeokuta, Ogun state, Nigeria (Ref. FUNAAB/COLVET/CREC/2022/02/05).

Statistical Analysis

The data obtained was analyzed using Statistical Package for Social Science (SPSS) version 21 software. Simple descriptive statistical analysis was performed to calculate percentages, mean, and standard deviation of data. Independent student t-test, one-way analysis of variance (ANOVA), and Pearson correlation coefficient were used to compare and

correlate measured variables between groups. A p-value of less than 0.05 was considered statistically significant, and the confidence interval for the study was set at 95%.

RESULTS

Table **1** shows the sperm indices of Abino rats administered with CdCl2 only (positive control) and the group not administered with $CdCl_2$ (negative control). The progressive motility and sperm counts were significantly reduced, while acrosome defect and percent abnormal sperm morphology were increased among the positive control group than the negative control group ($p<0.05$).

Table **2** shows the effect of the administration of varying concentrations of Curcumins on Albino rats, It indicates that percent progressive motility, viability, and sperm count increased in a dose-dependent manner, while the percent non-viable sperm cells decreased with increasing concentrations of Curcumins ($p<0.001$).

Table **3** shows Albino rats administered with CdCl2 followed by supplementation with 240 mg/kg Curcumins either simultaneously for 28 days (group 6), 14 days post-injury (group 7), 28 days post-injury (group 8) or left to recover without supplementation with Curcumins for 14 days (group 9). The percent progressive motility, viability, and sperm count were significantly increased, while non-viable sperm cells were significantly decreased after treatment with Curcumins. The observed changes were not dependent on the duration and mode of treatment with Curcumins. There was no change observed in acrosome defect and abnormal morphology. Although the percent motility, viability, and sperm count among the group left to recover without supplementation with Curcumins were significantly higher than the positive control group, the levels were lower than observed among the treatment group.

Table **4** indicates that serum GPX, GSH and SOD and semen GPx , GSH and SOD were significantly reduced ($p<0.001$) in $CdCl_2$ administered rats than negative

Table 1: Comparison of Sperm Indices among Albino Rats Administered with 20 mg/Kg of Cadmium Chloride only and Negative Control Group

Parameters	Group 5 (Positive Control) Mean +S.D	Group 1 (Negative Control) Mean +S.D	P -Values
Progressive Motility (%)	67.0±2.7	77.2±1.6	0.050
Viability (%)	60.6±16.6	65.0±13.0	0.754
Non-Viable (%)	35.0±16.6	17.0±13.0	0.754
Sperm count ($X10^{6/}$mL)	54.6±12.1	92.8±11.8	<0.001
Normal Morphology (%)	90.8±4.5	94.6±2.3	0.751
Acrosome Defect (%)	3.2±2.3	0.00±0.0	<0.001
Abnormal Morphology (%)	1.0±0.0	3.4±0.9	<0.001

Table 2: Effect of Varying Dosage of Curcumins Administration on Sperm Indices among Albino Rats

	Progressive Motility %)	Viability (%)	Non-Viable (%)	Sperm count ($X10^6$/ml)	Normal Morphology (%)	Acrosome Defect (%)	Abnormal Morphology (%)
Group 1 (Control)	77.2±14.6	63.0±13.0	37.0±13.0	92.8±11.8	94.6±2.3	0.00±0.0	3.4±1.1
Group 2	80.6±9.6	77.0±12.0	23.0±12.0	112.0±13.5	91.2±8.6	1.0±0.0	3.2±5.2
Group 3	85.2±6.6	77.4±9.9	22.6±9.9	108.0±22.9	88.6±5.9	0.0±0.00	4.0±3.7
Group 4	92.6±3.4	93.8±4.4	6.2±4.4	144.2±7.0	95.0±4.1	0.0±0.0	4.8±1.9
P^a – Values	0.581	0.033	0.033	0.031	0.297	0.045	0.845
P^b Values	0.120	0.029	0.029	0.084	0.070	1.000	0.217
P^c – Values	0.004	<0.001	<0.001	<0.001	0.902	1.000	0.450

P^a-Values: P values between Group 2 and Control.
P^b- Values: P values between Group 3 and Control.
P^c- Values P values between Group 4 and Control.

Table 3: Effect of Curcumins Supplementation on Cadmium Chloride Administered Albino Rats and a Group Left to Recover Compared with Positive Control Group

	Progressive Motility %)	Viability (%)	Non-Viable (%)	Sperm count ($X10^6$/ml)	Normal Morphology (%)	Acrosome Defect (%)	Abnormal Morphology (%)
Group 5 (PC)	60.0±2.7	60.6±16.6	35.0±16.6	54.6±12.1	90.8±4.5	3.2±2.3	1.0±0.0
Group 6	84.8±4.1	87.6±5.4	12.4±5.4	77.8±8.9	93.0±3.7	1.0±0.0	1.0±0.0
Group 7	83.0±2.8	78.0±7.3	22.0±7.3	77.4±13.0	93.0±4.5	2.0±0.0	1.0±0.0
Group 8	85.4±7.6	84.6±6.6	15.4±6.6	85.0±12.3	91.2±4.4	2.0±0.0	1.2±1.0
Group 9	71.8±5.4	71.8±8.1	30.2±8.1	68.6±14.5	90.0±5.3	2.0±0.0	1.0±00
P^e-Values	0.001	0.001	<0.001	0.002	0.622	0.05	1.000
P^f- Values	0.001	0.023	0.023	0.002	0.622	1.000	1.000
P^g- Values	0.001	0.002	0.002	0.001	0.297	1.000	1.000
P^h- Values	0.05	0.05	0.173	0.018	0.164	1.000	1.000

PC= Positive Control (group 5).
P^e-Values: P values between Group 6 and Control.
P^f- Values: P values between Group 7 and Control.
P^g- Values P values between Group 8 and Control.
P^h- Values P values between Group 9 and Control.

control group, while serum MDA level was significantly increased ($p<0.001$) among $CdCl_2$ treated rats than non-treated group.

Table **5** shows that in the groups that received Curcumins supplementation, the activities of serum CAT, GPx, and SOD increased significantly ($p<0.05$), while MDA significantly decreased ($p<0.001$) in a dose dependent manner. There was no significant different in the level of GSH between rats administered with varying concentrations of Curcumins and control.

Among the groups of rats treated with Curcumins following CdCl2 administration, serum CAT, GPx, GSH and SOD activities increased significantly ($p<0.05$), while MDA concentration decreased ($p<0.05$) irrespective of the duration and mode of treatment with Curcumins. Among the group left to recover without Curcumins supplementation, serum CAT and GPx significantly decreased ($p<0.05$), while the decrease in the activities of GPx and SOD was not significant.

Table **7** shows that the seminal plasma activity of CAT was significantly increased in group 3 (rats given a supplementation of 240 mg/kg Curcumins) but there was no significant increase in group 2 and group 4. The activity of GPx increased in group 3 ($p<0.001$) and group 4 ($p<0.05$), while the activity of SOD increased in group 3 ($p<0.002$) and group 4 ($p<0.05$) compared with control. There was no significant difference in the

Table 4: Effect of $CdCl_2$ Injury on Serum and Seminal Plasma Antioxidant Parameters in Albino Rats

Parameters	Group 5 (Positive Control)	Group 1 (Negative Control)	P -Value
Serum Catalase (U/L)	6.36±0.2	6.78±0.9	0.405
Serum GPx (U/L)	1.14±0.2	2.05±0.1	<0.001
Serum GSH (U/L)	44.15±2.0	58.4±7.7	<0.001
Serum MDA (nmol/L)	2.02±1.0	1.23±0.2	<0.001
Serum SOD (U/L)	0.34±0.1	0.48±0.0	<0.001
Semen Catalase (U/L)	7.13±4.4	8.82±0.1	0.128
Semen GPx (U/L)	1.92±0.1	3.14±0.2	<0.001
Semen GSH (U/L)	90.23±8.4	108.8±10.3	<0.001
Semen MDA (nmol/L^9)	1.64±0.1	0.54±0.0	0.540
Semen SOD (U/L)	0.54±0.1	0.91±0.1	<0.001

Table 5: Effect of Varying Dosages of Curcumins supplementation on Serum antioxidant Parameters among Albino Rats

	Catalase U/L	GPx U/L	GSH U/L	MDA nmol/L	SOD U/L
Control	6.78±0.9	2.05±0.1	58.4±7.7	1.23±0.2	0.48±0.0
Group 2	5.24±1.7	2.27±0.3	57.35±0.9	0.63±0.2	0.57±0.1
Group 3	7.20±0.3	2.52±0.1	57.82±5.4	0.64±0.1	0.63±0.0
Group 4	8.11±1.0	2.64±0.8	58.48±3.4	0.66±0.2	0.69±0.0
P^a – Values	0.003	0.05	0.814	0.001	0.05
P^b –Values	0.001	0.013	0.862	0.001	0.05
P^c – Values	0.001	0.005	0.958	0.001	0.05

P^a – Values: P values between Group 2 and Control.
P^b - Values: P values between Group 3 and Control.
P^c – Values P values between Group 4 and Control.

Table 6: Effect of Curcumins Supplementation on Serum Antioxidant Enzymes and Malondialdehyde in Cadmium Chloride Administered Albino Rats and a Group Left to Recover Compared with Positive Control Group

	Catalase U/L	GPx U/L	GSH U/L	MDA nmol/L	SOD U/L
Positive Control	6.36±0.2	1.14±0.2	44.15±2.0	2.02±1.0	0.34±0.1
Group 6	6.37±0.3	1.60±0.3	50.25±4.4	1.04±0.1	0.44±0.0
Group 7	7.10±0.5	1.63±0.1	49.4±1.4	0.95±0.1	0.40±0.1
Group 8	7.89±0.2	1.66±0.2	45.70±3.1	0.77±0.0	0.41±0.0
Group 9	5.11±0.0	1.26±0.1	39.25±3.4	1.35±0.2	0.29±0.3
P^e – Values	0.99	0.032	0.001	0.05	0.009
P^f- Values	0.05	0.032	0.001	0.05	0.05
P^g- Values	0.03	0.02	0.001	0.03	0.05
P^h –Values	0.05	0.347	0.001	0.553	0.5

P^e-Values: P values between Group 6 and Control.
P^f- Values: P values between Group 7 and Control.
P^g- Values P values between Group 8 and Control.
P^h- Values P values between Group 9 and Control.

Table 7: Effect of Different Concentrations of Curcumins Supplementation on Seminal Plasma Antioxidant Enzymes and Malondialdehyde in Albino Rats

	Catalase U/L	GPx U/L	GSH U/L	MDA nmol/L	SOD U/L
Controls	8.82±0.1	3.14±0.2	108.8±10.3	0.54±0.0	0.91±0.1
Group 2	8.94±0.2	3.18±0.2	101.7±5.5	0.50±0.1	0.90±0.2
Group 3	10.05±0.4	4.32±0.1	104.5±2.1	0.56±0.0	0.89±0.1
Group 4	8.01±2.5	3.84±0.5	111.07±7.5	0.58±0.1	0.76±0.0
P^a – Values	0.883	0.870	0.721	0.240	0.951
P^b - Values	0.05	0.001	0.783	0.605	0.002
P^c – Values	0.459	0.005	0.672	0.601	0.025

P^a – Values: P values between Group 2 and Control.
P^b - Values: P values between Group 3 and Control.
P^c – Values P values between Group 4 and Control.

Table 8: Effect of Curcumins Treatment on Seminal Plasma Antioxidant Enzymes and Malondialdehyde in Cadmium Chloride Administered Albino Rats and a Group Left to Recover Compared with Positive Control Group

	Catalase U/L	GPx U/L	GSH U/L	MDA nmol/L	SOD U/L
Positive Control	8.82±0.1	3.14±0.2	108.8±10.3	1.64±0.0	0.91±0.1
Group 6	9.55±0.6	2.13±0.6	98.98±2.1	1.13±0.4	0.87±0.0
Group 7	9.98±0.6	3.87±0.5	111.04±11.8	0.88±0.0	0.58±0.2
Group 8	9.38±0.2	2.72±0.5	108.2±3.1	0.90±0.0	0.69±0.0
Group 9	7.04±0.4	2.17±0.3	112.6±14.2	1.21±0.2	0.72±0.0
P^e-Values	0.443	0.001	0.101	0.001	0.542
P^f- Values	0.313	0.035	0.677	0.001	0.001
P^g - Values	0.508	0.093	0.901	0.001	0.001
P^h- Values	0.107	0.001	0.484	0.001	0.005

P^e-Values: P values between Group 6 and Control.
P^f- Values: P values between Group 7 and Control.
P^g- Values P values between Group 8 and Control.
P^f- Values P values between Group 9 and Control.

activity of GSH in all the groups compared with control. The activities of CAT, GPx, GSH and SOD as well as MDA level did not change among rats in group 2 compared with control.

The supplementation with curcumins following $CdCl_2$ testicular injury reveals no significant increases in the activities of CAT, GPx and GSH, except for SOD which was significantly increased among rats in groups 7 and 8 ($p<0.05$). Also, seminal plasma MDA was significantly increased among rats in groups 6, 7 and 8 ($p<0.001$). The rats in group 9 that were allowed to recover without Curcumins supplementation had significantly reduced GPx ($p<0.001$), MDA and SOD ($p<0.05$) compared with positive control group. There was no significant change in mean CAT, GPx and GSH levels compared with positive control.

Figures **1-4** are sections from negative control group 1, positive control group 5, cadmium chloride treated rats and supplementation with Curcumins group 6 and the group left to recover without Curcumins supplement 9. The testicular structures indicate that the negative control group shows normal and uniform structure of seminiferous tubule, (Group 1). The cadmium chloride-treated group (Group 5), shows some degenerative alterations such as shrunken seminiferous tubules wall, hypoplasia of spermatogonia and vacuolation of the seminiferous epithelium. Testicular section from the rats administered with 20mg/kg cadmium chloride every other day for 28 days and left to recover without treatment for another 28days shows little recovery from the shrunken seminiferous tubules and vacuolation of the seminiferous epithelium

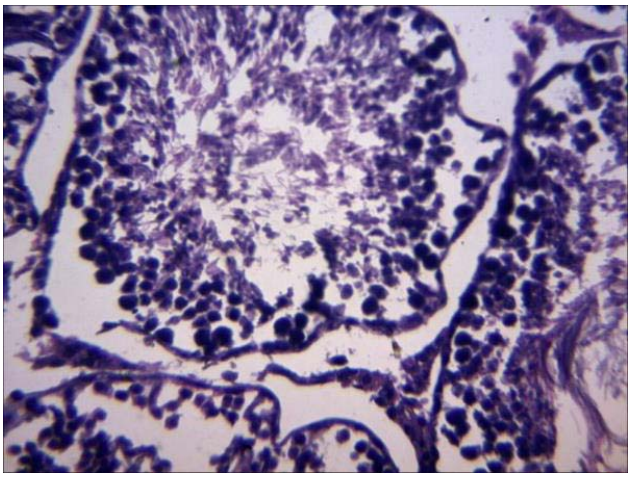

Figure 1: Testis section from negative control rats showing no visible lesions.

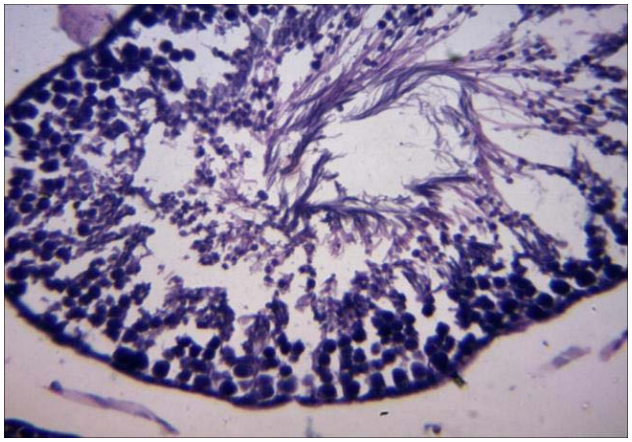

Figure 2: Testis section from Positive control rats administered with 20 mg/kg cadmium chloride solution showing shrunken seminiferous tubules/vacuolation of the seminiferous tubules (Group 5).

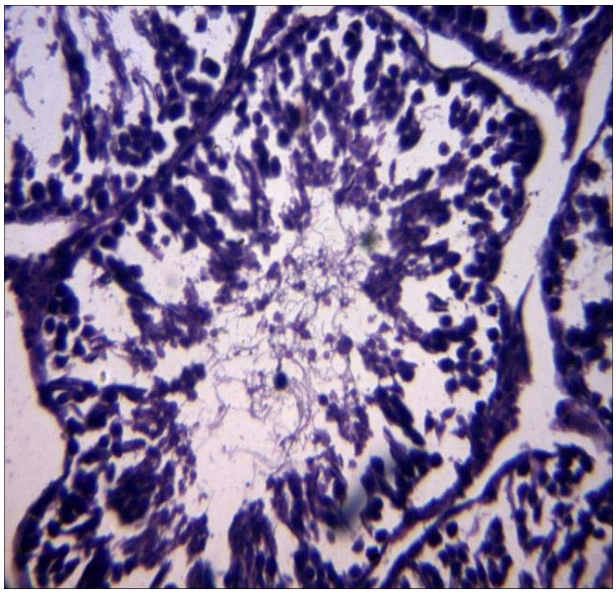

Figure 3: Testis section from rats administered with 20 mg/kg cadmium chloride solution and 120 mg/kg curcumins simultaneously every other day for 28 days showing no visible lesions. There was an amelioration of the vacuolation of the seminiferous tubules (Group 6).

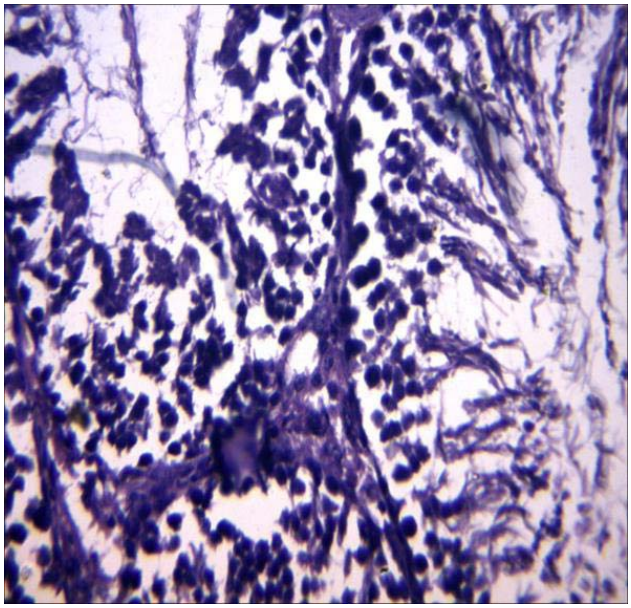

Figure 4: Testis section from rats administered with 20mg/kg cadmium chloride every other day for 28days and left to recover without treatment for another 28days (Group 9) shows little recovery from the shrunken seminiferous tubules and vacuolation of the seminiferous epithelium.

DISCUSSION

Exposure of animals to toxic metals such as Cd often leads to reproductive toxicity and infertility within the testicular tissue of animals [20]. Cadmium is an ecological impurity that can induce the excess generation of free radicals while devastating the antioxidant defense system. Cadmium has been reported to have adverse effects on human and animal reproductive health even at low levels of exposure, and males appear to be more susceptible than females to the adverse effects of occupational or environmental exposures to reproductive toxicants [4]. Conversely, Curcumin is considered a free-radical scavenger and a potent antioxidant and it serves as a protective factor against oxidative stress inducers such as cadmium and may avert the histopathological impairment caused by cadmium chloride testicular injury [21]. It is imperative to study curcumin applications for food and reproductive health promotion among males. This is particularly important because of the declining trend in semen quality and quantity indices that are prevalent in sub-Saharan Africa. Several properties have been attributed to curcumin as having antioxidant, anti-inflammatory, neuroprotective, anticancer, hepatoprotective, and cardioprotective effects [22].

In this study, it was observed that the administration of cadmium chloride to Albino rats resulted in a significant decline ($p<0.001$) in sperm quality parameters and an increase in abnormal morphology when compared with controls. This aligned with a previous study [23], that reported that Cd may cause deleterious structural damage to the seminiferous tubules, Sertoli cells, and blood-testis barrier, which may lead to poor sperm quality and quantity indices. An important finding of this study is the acrosome defect which has not been previously evaluated to the best of our knowledge. Other researchers have shown that Cd can affect the Sertoli cells to cause poor spermatogenesis [23].

The administration of different concentrations of curcuminoid extract to male albino rats causes enhancement in some sperm characteristics. There was significant enhancement in sperm viability, and sperm counts and a significant decrease in non-viable sperm cells ($p<0.001$). This was partly consistent with previous study [24] in which authors investigated the effects of dietary curcumin supplementation in broiler breeder roosters. It was reported that administration of dietary supplementation of curcumin was associated with beneficial effects on semen quality indices and fertility rate in aged broiler breeder roosters in a dose-dependent manner [24]. Curcumin was reported to remedy testicular function and Sperm indices in Male Mice fed with low-carbohydrate-diet-induced metabolic dysfunction. It was suggested that curcumin may improve poor spermatogenesis and sperm indices, and reverse oxidative stress, prevent inflammation and

apoptosis in the testes of low-carbohydrate-diet-fed mice [23,24].

In this study, among animals fed with cadmium and supplemented with varying concentrations of curcumins there was an enhancement in sperm viability and reduction of non-viable sperm cells as well as the improvement of sperm morphology in a dose-dependent manner. This finding was corroborated by an in-vitro study, which showed that the addition of 20 μM curcumin to a freezing medium containing sperm cells from healthy individuals caused increases in progressive and non-progressive motility as well as significant reductions in intracellular reactive oxygen species and DNA fragmentation in frozen-thawed sperm cells [25]. In an earlier study, it was reported that the use of curcumin in preventing the deleterious effects of cadmium on the reproduction of male rats led to improvement in sperm functional characteristics, decreased sperm count, testosterone concentration, and oxidative stress [26]. The present study has shown that the administration of curcumin may confer some protection against the adverse effects of cadmium in male Albino rats.

The mechanisms by which the protection is conferred might be via inhibition of oxidative stress. This is so because the levels of serum and semen antioxidant levels in albino rats administered with 20g/Kg cadmium solution in this study were significantly lower compared to the control. In contrast, serum MDA was higher when compared with negative controls. Upon supplementation with varying concentrations of curcumins, serum antioxidant levels were significantly higher in rats administered with curcumins daily for 28 days in a dose-dependent manner. Curcumin supplementation increased the activity levels of catalase, superoxide dismutase, glutathione reductase, and glutathione peroxidase (GPx) but caused a decrease in malondialdehyde (MDA) and hydrogen peroxide values.

Curcumin may possess the capacity to promote antioxidant activity by scavenging a variety of reactive oxygen species (ROS) as superoxide radicals, hydrogen peroxide, and nitric oxide (NO) radicals and by inhibiting lipid peroxidation [27]. Some authors have suggested that Curcumin can increase the GSH levels by upregulating glutathione transferase and their mRNAs. Curcumin may also prevent ROS-generating enzymes, such as LOX, COX, and xanthine oxidase and curcumin has been considered a chain-breaking antioxidant because of its lipophilic nature, potentially acting as a peroxyl radical scavenger [22].

The histomorphological evaluation of the testis in our study shows that there was a normal and uniform structure of seminiferous tubules in the control group. Whereas, in the cadmium chloride-treated groups, some degenerative alterations were noted such as the seminiferous tubule's wall shrunken, and vacuolation of the seminiferous epithelium was observed. The exposure of rats to 20 mg/kg cadmium chloride for 28 days resulted in ultrastructure changes. In a similar study, rats exposed to a 3 μmol/kg given as a single dose resulted in vacuolation in the Sertoli Cells cytoplasm and damage to sperm DNA [28]. Some authors have shown that exposure to Cd by inhalation for 28 days also caused alterations in mitochondrial integrity in adult mice [29]. In the cadmium chloride treated and curcumin supplemented groups, it appears curcumin conferred some measure of protection against the cadmium-induced histopathological alterations [30-32]. Curcumin is a potent antioxidant, which may recoup the harmful effects of cadmium on lipid peroxidation, potentiated serum and semen antioxidant defense system, and improve some testicular morphology in the testis of cadmium-chloride treated Albino rats [33,34]. This observation suggests that supplementation with curcumin may reverse the toxic effects of cadmium chloride on the mean diameter of seminiferous tubules and seminiferous tubule lumen.

6. CONCLUSION

It can be concluded from this study that curcumins supplementation has protective effect against cadmium-induced testicular injury in a dose dependent manner. This protective effect may be due to the antioxidant property, prevention of lipid peroxidation and oxidative stress by curcumins. Curcumin supplementation may prevent harmful reproductive effects of cadmium chloride in males.

CONFLICT OF INTEREST STATEMENT

The authors declare that there is no conflict of interest in the current research work.

RESEARCH ETHICS COMMITTEE PERMISSION

The research work was approved by the College of Veterinary Medicine Research Ethics Committee (CREC), Federal University of Agriculture, Abeokuta, Ogun state, Nigeria (Ref. FUNAAB/COLVET/CREC/ 2022/02/05).

ACKNOWLEDGEMENT

The authors appreciate the contributions of the medical laboratory scientists and the research assistants for their contributions towards the completion of this study.

FUNDING

This study did not receive any funding or material support.

REFERENCES

[1] Tvrdá E, Benko F, Slanina T, du Plessis SS. The Role of Selected Natural Biomolecules in Sperm Production and Functionality. Molecules 2021; 26(17): 5196. https://doi.org/10.3390/molecules26175196

[2] Emokpae MA, Adobor CA. Association of seminal plasma cadmium levels with semen quality in non-occupationally exposed infertile Nigerian males. J Environ Occupation Health 2015; 4 (1): 40-42.

[3] Marini HR, Micali A, Squadrito G, Puzzolo D, Freni J, Antonuccio P, Minutoli L Nutraceuticals: A New Challenge against Cadmium-Induced Testicular Injury. Nutrients 2022; 14: 663. https://doi.org/10.3390/nu14030663

[4] Piadé J, Jaccard G, Dolka C, Belushkin M, Wajrock S. Differences in cadmium transfer from tobacco to cigarette smoke, compared to arsenic or lead. Toxicol Rep 2014; 2: 12-26. https://doi.org/10.1016/j.toxrep.2014.11.005

[5] Richter P, Faroon O, Pappas RS. Cadmium and Cadmium/Zinc Ratios and Tobacco-Related Morbidities. Int J Environ Res Public Health 2017; 14: 1154. https://doi.org/10.3390/ijerph14101154

[6] Rinaldi M, Micali A, Marini H, Adamo EB, Puzzolo D, Pisani A, Trichilo V, Altavilla D, Squadrito F, Minutoli L. Cadmium, Organ Toxicity and Therapeutic Approaches: A Review on Brain, Kidney and Testis Damage. Curr Med Chem 2017; 24: 3879-3893. https://doi.org/10.2174/0929867324666170801101448

[7] DeFelice SL. The nutraceutical revolution: Its impact on food industry R&D. Trends Food Sci Technol 1995; 6: 59-61. https://doi.org/10.1016/S0924-2244(00)88944-X

[8] Ilbey YO, Ozbek E, Cekmen M, Simsek A, Otunctemur A, Somay A. Protective effect of curcumin in cisplatin-induced oxidative injury in rat testis: mitogen-activated protein kinase and nuclear factor-kappa B signaling pathways. Hum Reprod 2009; 24(7): 1717-1725. https://doi.org/10.1093/humrep/dep058

[9] Chitturi R, Renuka IV, Premalatha P, Madhavi K. Patterns of semen analysis in male partners of infertile couples at a tertiary care hospital. Indian J Pathol Oncol 2009; 4(4): 536-539. https://doi.org/10.18231/2394-6792.2017.0116

[10] El-Wakf AM, Elhabiby EM, El-kholy WM. Use of tumeric and curcumin to alleviate adverse reproductive outcomes of water nitrate pollution in male rats. Nat Scis 2011; 7(7): 229-239. http://www.sciencepub.net/nature

[11] Samarghandian S, Azimi-Nezhad M, Shabestari MM. Effect of chronic exposure to cadmium on serum lipid, lipoprotein and oxidative stress indices in male rats. Interdiscip Toxicol 2015; 8(3): 151-154. https://doi.org/10.1515/intox-2015-0023

[12] Tremellen K. Oxidative stress and male infertility: A clinical perspective. Humums Reprod Update 2008; 14: 243-58. https://doi.org/10.1093/humupd/dmn004

[13] Ikpeme EV, Udensi O, Ekaluo UB, Uyoh EA, Asuquo BO, Udoh FV, Udoh PB. Effect of crude extract of carica papaja seeds on the reproductive efficiency of male albino rats. Global J Pure Applied Sci 2007; 13: 365-368.

[14] Albro PW, Corbett JT, Schroeder JL. Application of the thiobarbiturate assay to the measurement of lipid peroxidation products in microsomes. J Biochem Biophys Methods 1986; 13(3): 185-194. https://doi.org/10.1016/0165-022x(86)90092-8

[15] Rotruck JT, Pope AL, Ganther HE, Swanson AB, Hafeman DG, Hoekstra W. Selenium: biochemical role as a component of glutathione peroxidase. Science 1973; 179(4073): 588-590. https://doi.org/10.1126/science.179.4073.588

[16] Hadwan MH, Abed HN. Data supporting the spectrophotometric method for the estimation of catalase activity. Data in Brief 2016; 6: 194-199. https://doi.org/10.1016/j.dib.2015.12.012

[17] Sun YI, Oberley LW, Li Y. A simple method for clinical assay of superoxide dismutase. Clin Chem Lab Med 1988; 34(3): 497-500. https://doi.org/10.1093/clinchem/34.3.497

[18] Gangarapu V, Gujjala S, Korivi R, Pala I. Combined effect of curcumin and vitamin E against CCl4 induced liver injury in rats. Am J Life Sci Res 2013; 1(3): 117-124. https://doi.org/10.11648/j.ajls.20130103.17

[19] Suvarna KS, Layton C, Bancroft JD. Bancroft's Theory and Practice of Histological Techniques. 7th ed. Oxford, Churchill Livingstone Elsevier 2013; p. 654.

[20] Uadia PO, Emokpae MA. Male infertility in Nigeria: A neglected reproductive health issue requiring attention. J Basic Clin Reprod Scis 2015; 4(2): 45-67. https://doi.org/10.4103/2278-960X.161042

[21] Momeni H, Eskandari N. Curcumin protects the testis against cadmium-induced histopathological damages and oxidative stress in mice. Hum Experimen Toxicol 2020; 39(5): 653-661. https://doi.org/10.1177/0960327119895564

[22] Sharifi-Rad J, Sharifi-Rad M, Salehi B, Iriti M, Roointan A, Mnayer D. *In vitro* and *in vivo* assessment of free radical scavenging and antioxidant activities of Veronica persica Poir. Cell Mole Biol 2018; 64: 57-64.

[23] Zhu K, Shi F, Zhu X, Yang W. The roles of oxygen vacancies in electrocatalytic oxygen evolution reaction. Nano Energy 2020; 73: 104761. https://doi.org/10.1016/j.nanoen.2020.104761

[24] Haffor AS, Abou-Tarboush FM. Testicular cellular toxicity of cadmium: transmission electron microscopy examination. J Environ Biol 2004; 25: 251-258.

[25] de Souza Predes F, Diamante MA, Dolder H. Testis response to low doses of cadmium in Wistar rats. Int J Experimen Pathol 2010; 91: 125-131. https://doi.org/10.1111/j.1365-2613.2009.00692.x

[26] Ibrahim J, Kabiru AY, Abdulrasheed-Adeleke T, Lawal B, Adewuyi AH. Antioxidant and hepatoprotective potentials of curcuminoid isolates from turmeric (Curcuma longa) rhizome on CCl4-induced hepatic damage in Wistar rats. J Taibah Univ Sci 2020; 14(1): 908-915. https://doi.org/10.1080/16583655.2020.1790928

[27] Ak T, Gülçin I. Antioxidant and radical scavenging properties of curcumin. Chem Biol Interact 2008; 10; 174(1): 27-37. https://doi.org/10.1016/j.cbi.2008.05.003

[28] Bizarro P, Acevedo S, Niño-Cabrera G, Mussali-Galante P, Pasos F, Avila-Costa MR, Fortoul TI. Ultrastructural modifications in the mitochondrion of mouse Sertoli cells after inhalation of lead, cadmium or lead-cadmium mixture. Reprod Toxicol 2003; 17(5): 561-566. https://doi.org/10.1016/s0890-6238(03)00096-0

[29] Kazemizadeh A, Zare Shahneh A, Zeinoaldini S, Yousefi AR, Mehrabani Yeganeh H, Ansari Pirsaraei Z, Akhlaghi A. Effects of dietary curcumin supplementation on seminal quality indices and fertility rate in broiler breeder roosters. Br Poult Sci 2019; 60(3): 256-264. https://doi.org/10.1080/00071668.2019.1571165

[30] Priyadarsini KI. Chemical and structural features influencing the biological activity of curcumin. Curr Pharm Des 2013; 19(11): 2093-2100. https://doi.org/10.2174/138161213805289228

[31] Mohajeri M, Rezaee M, Sahebkar A. Cadmiuminduced toxicity is rescued by curcumin: a review. Biofactors 2017; 43(5): 645-661. https://doi.org/10.1002/biof.1376

[32] Marathe SA, Sen M, Dasgupta I, Chakravortty D. Differential modulation of intracellular survival of cytosolic and vacuolar pathogens by curcumin. Antimicrob Agents Chemother 2012; 56(11): 5555-67. https://doi.org/10.1128/AAC.00496-12

[33] Momeni HR, Eskandari N. Effect of curcumin on kidney histopathological changes, lipid peroxidation and total antioxidant capacity of serum in sodium arsenite-treated mice. Exp Toxicol Pathol 2017; 69(2): 93-97. https://doi.org/10.1016/j.etp.2016.08.006

[34] Nair AR, Degheselle O, Smeets K, Van Kerkhove F, Cuypers A. Cadmium induced pathologics: Where is the oxidative balance lost (or not)? Int J Mol Sci 2013; 14: 6116-6143. https://doi.org/10.3390/ijms14036116