JOURNAL OF BASIC PHARMACOLOGY AND TOXICOLOGY

Original Research Article

Toxicological evaluation of Mojeaga herbal remedy on experimental animals

MacDonald Idu[1*], Monday Ojeaga Alugeh[2], Michael Ojeaga Alugeh[2], Benjamin Ogunma Gabriel[1]

[1]*Phytomedicine Unit, Department of Plant Biology and Biotechnology, PMB 1154, Benin City, Edo State. Nigeria*

[2]*Mojeaga International Ventures Ltd, Flat 75, Block H, Federal Housing Estate, Ikpoba Hill, Benin City, Edo State, Nigeria*

Abstract

This study evaluated the toxicological profile of Mojeaga herbal remedy on male and female animal models. In a time-dependent study, acute and chronic toxicity of Mojeaga herbal remedy in male and female Wistar rats were investigated through thorough examination of mortality rate, body and organ weight changes, hematological indexes, biomarkers of hepatic and renal functions, lipid profile, *in-vivo* antioxidant assay, hormonal assay and histopathological study across all treatment groups using standard protocol. No observable behavioral change with absent lethality at 10 to 10000 mg/kg of Mojeaga herbal remedy. There was no drastic significant change ($p > 0.05$) in the body and organ weight of the male and female animals. In the chronic toxicity studies, Mojeaga herbal remedy indicated no significant difference ($p > 0.05$) in haematological indices, liver function test, kidney function test, lipid profile, antioxidant indexes and hormonal assays with a slight significant increase ($p < 0.05$) in hepatic (ALT, ALP AST) and renal (potassium, sodium and chloride), lipid profile (cholesterol, triglyceride, high density lipoprotein, low density lipoprotein), in-vivo antioxidant assay (MOD, catalase, SOD and glutathione). Also, no defect on male and female hormonal assay (testosterone, estrogen, progesterone, follicle stimulating hormone and luteinizing hormone) activities among treated groups when compared with control. There was no marked significant toxicological effect ($p > 0.05$) on serum total protein (TP), blood urea nitrogen (BUN), albumin (ALB) and creatinine (CREA) urea levels across the whole treated groups at graded doses of 150, 300 and 600 mg/kg. Mojeaga product caused no histopathological variation on vital visceral organs (liver, kidney, heart, stomach, brain, lungs spleen testes and uterus) when compared with control. It is contingent from this study that Mojeaga herbal remedy exhibited bio-protective and hyper-stimulating effects with harmless and protective therapeutic benefits.

Keywords: Toxicological study, Mojeaga herbal remedy, Experimental animals

1 Introduction

Natural source of certain Substances for instance; the use of herbs implicated in management and treatment of human and animal disease since antiquity [1]. Hence, the bioactive component in plants enhances their therapeutic effect with rational therapy against diverse form of diseases [2, 3]. Natural products obtain from plants are of enormous importance due to bioactive phytochemicals in novel drug research and development [4, 5].

Toxicology can be traditionally defined as science of poisons [6]. The understanding of various agents triggering humans and animals' diseases, portray an obvious toxicological definition as "study of adverse effects from physical or chemicals agents in living entities". Adverse effects are of various forms, extending from onset mortality to indirect damage that later actualized [7]. They occur at different stages in the body, ranged from organ, cell type or explicit biochemical. Knowledge of damage done to the body by toxin has advanced to medical information. Possibly, the observable anatomical or physiological changes is as a result of previous unidentifiable variations in various biochemical of the body [8]. Toxic agents are either systemic or organ toxins [9]. A systemic toxin affects the whole body or delicate organs rather than selected pathways. For instance, the case of potassium cyanide being a systemic toxicant affects nearly all cell and organ of the body via stimulating cell's ability to exploit oxygen. Toxicants also affect explicit tissues or organs without any damage done to the body [10]. These selected sites are known as targeted organs or tissues site such as; Benzene and lead has specific organ toxin site that is mainly a blood-forming toxic site for the formal, while central nervous system, kidney, and hematopoietic system for the latter [11]. Toxic sites are targeted tissue [12].

Alchornea cordifolia is a perennial herb belongs to Euphorbiaceae's family. It is known as Dove wood in English. It is known to exhibit a great variety of biological

*Corresponding Author; E-mail: mcdonald@uniben.edu; Tel: +2348050607009

activities from different parts of the plant [13]. It remediates respiratory problems like sore throat, cough and bronchitis [14]. It aids in the treatment of ulcers, dysentery, worms, diarrhea and amoebic. *A. Cordifolia* is implicated in the management of leprosy, also serves as an antidote to snake bite [15]. The root and bark increase sexual recitals in Congo in Africa sub-region [16]. It aids in the treatment of gonorrhea, pain, cough, yaws and rheumatic [14].

Sorghum bicolor leaves is a glasslike with less broad leaves. It belongs to Graminea family [17]. It is locally known as 'oka pupa' in Southern Nigeria. *S. bicolor* is rich in protein with less amount of fatty content, partially responsible for its hemopoietin effect [18]. Report have shown that sorghum serves as anti-abortive, cyano-genetic, emollient, intoxicant, demulcent and diuretic, [19]. Sorghum is potent for the management of cancer, epilepsy, diarrhea, tubercular swellings, flux and stomach ache [20]. Recently, findings showed that it has herbal remedy for anemia by its effect on blood increase concentration, also use as blood tonics. Study from Okonkwo *et al*. [21] validated that accurate laboratory blood parameters determination remain sensitive and consistent for ethical and cogent research, diagnosis, cure and prevention of associated anaemia diseases.

Pennisetum glaucum known as Pearl millet, it belongs to the family Paniceae also called Poaceae. From its folklore report, it showed effect in increasing haemoglobin, zinc (3.1 mg/100 g) and iron (8 mg/100 g) in anemia cases [22]. Report had shown its effect in managing constipation since it contains high fiber content (1.2 g/100 g) and aids in the management of cancer, hence promoting anti-cancer property [23]. Moreover, it aids in the treatment of diarrhea and regulated hyperglycemic state because of its low glycemic index. It is useful as anti-allergic due to the absence of gluten. *P. glaucum* in other studies elicited a well DNA scission via LDL, cholesterol, liposome oxidation and proliferation of HT-29 adenocarcinoma cells reduction and promote nutraceutical content. Pearl millet has a large amount of phosphorus [24].

2 Methodology

2.1 Collection of plant material

Alchornea cordifolia, Sorghum bicolor, Pennisetum glaucum leaves were collected from Mojeaga herbal venture Nigeria LTD Company, Benin City, Edo State, Nigeria and the plants were identified and authenticated by Prof. M. Idu in Herbarium unit of Plant Biology and Biotechnology Department, University of Benin, Benin City, Issued with voucher number UBH-4361.

2.2 Preparation of plant material

The pulverized leaves were weighed separately to attain a uniform weight. The weighed samples were properly mixed together at equal ratio 1:1:1 to form Mojeaga herbal remedy. The mixed samples were further weighted to be 4500 grams and were extracted using maceration technique in aqueous solvent. Filtrate was concentrated using fix-drier into powdered form. It was stored in a sterile container and kept in a refrigerator at 4 ^{o}C.

2.3 Experimental animals

Forty eight (48) healthy male and female Wistar rat weighed 180-250 g and forty five (45) matured Swiss albino mice weighed 25-30 were used. They were bred in Biochemistry animal house, University of Benin, Benin City, in a well-ventilated woody cages under normal laboratory conditions (12 hours light/dark cycle: 23 ± 2 °C) and fed with standard diet. Food and water were given free choice (*ad libitum)*. All selected animals for this experiment were acclimatized for 14 days. They were handled according to standard protocols, all through the experimental period (3 months). The institutional Ethical Review Committee of Life Sciences, University of Benin, approved animal uses for experiment with ethical number of LS20014.

2.4 Acute Toxicity Study

Determination of acute toxicity using modified method by Lorke [25] and class method in adherence to the Organization for Economic Cooperation and Development (OECD) Guideline for Testing Chemicals No. 423 [26]. This was performed in two phases each for Mojeaga product. Phase 1, forty five (45) mice were randomly divided into nine (9) groups of five (5) each. Mojeaga herbal remedy were administered in a single dose of 10, 100 and 1000 mg/kg) orally. Observable toxicological signs were recorded. With absence of toxicity, the second phase was carried out.

Based on the acquired results from phase 1, Phase 2 is scheduled with three (3) mice per group and were administered a single dose (1600, 2900 and 5000, 10000 mg/kg) of Mojeaga herbal remedy orally. The animals in the both phases were kept under same conditions and observed for general behavior changes continuously for 30 minutes, every hour during the first 24 hours and at least once daily for 14 days after administration of the product. Observations were focused on parameters such as piloerection, sensitivity to sound and touch, locomotion, aggressiveness, appearance of feces, salivation, urinating, convulsing, coma and death. The number of survivors was noted after 24 hours. Animals' weights were observed for day 0, day 7 and day 14. The LD_{50} was evaluated and classified according to the Globally Harmonized System (GHS) for the classification of chemicals [26, 27]. The LD_{50} was calculated based on the final results in square root of the product of the lowest lethal dose and the highest non-lethal dose, i.e., the geometric mean of the consecutive doses where 0 and 100 % survival rates were recorded. Where Mojeaga herbal remedy doses above 10000 mg/kg did not cause any visible toxicity or mortality, the extracts were regarded as safe. A last group served as control and was administered 1 ml distilled water orally. Every rat used in the acute toxicity study was observed daily for possible toxicity signs over a period of two weeks.

The LD_{50} was calculated using the formula:

$$LD_{50} = \sqrt{(D_0 \times D_{100})}$$

D_0 = Highest dose that gave no mortality,
D_{100} = Lowest dose that produced mortality.

2.5 Experimental protocol

Male and female albino rats were obtained and randomly divided into four groups (n = 6). The groups received 150, 300, 600 mg/kg of Mojeaga herbal remedy orally. Group A received 150 mg/kg of the Mojeaga herbal remedy orally. Group B received 300 mg/kg of the Mojeaga herbal remedy orally. Group C received 600 mg/kg of the Mojeaga herbal remedy orally. Group D received 0.5 mL/kg of distilled water orally. These substances were given daily to the respective groups throughout the periods of three (3) months. The animals were sacrificed under mild chloroform anesthesia. Blood samples and organs were isolated for analysis.

2.6 Haematological indexes

Haematological analyses were performed on whole blood collected into tubes with ethylenediaminetetraacetic acid (EDTA). White blood cell (WBC), lymphocytes (LYM), mid cells (MID), neutrophils (NEU), red blood cells (RBC), haemoglobin concentration (HB), haematocrit (HCT), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC), red cells distribution weight (RDW) and platelets (PLT) were determined by an automatic analyzer (CELL DYN 1700, Abbott Diagnostics Division, Abbott Laboratories, Abbott Park, IL, USA).

2.7 Kidney function test

Biochemical analyses were performed on serum obtained after centrifugation of whole blood (without anticoagulant) at 3000 rpm for 5 min.
Assay for urea: This test exploits the hydrolysis of urea to ammonia in the presence of urease enzyme. 100 µl of reagent 1 was added to 10µl serum samples in test tubes. 100 µl of reagent 1 was also added to 10µl distilled water to serve as blank. Solutions in the test tubes being thoroughly mixed for incubation at 10 minutes of 37 °C. 2.5 ml of reagents 2 and 3 drops were added in each test tube. Solutions in test tubes were mixed and further incubated for 10 minutes at 37 °C. Absorbances of samples and standard were read against the blank using the wavelength of 546 nm. Urea values were calculated as thus:
Urea concentration (mg/dl) = Absorbance of sample/ Absorbance of standard x 80.
Assay for creatinine: This test exploits the reaction of creatinine present in alkaline mixture using picric acid to produce colored complex. The quantity of complexity formed is directly proportional to creatinine concentration. 0.2 ml sample macro and semi micro were added to the test tube. 0.2 ml standard macro and 0.1 ml semi micro were added to the standard solution test tube. 2 ml of standard macro, 1 ml of semi micro, 2 ml of sample macro and 1 ml of semi micro were added to the working reagent test tube. The solutions in the test tubes were mixed. The absorbances of the samples and standard were measured at 492 nm. Creatinine concentration being calculated as thus:
Creatinine concentration units (mg/dl) = Absorbance of sample/Absorbance of standard x 2.
Assay for electrolytes (Sodium, chloride, bicarbonate and potassium): The assay for bicarbonate exploits the reaction between bicarbonate ions with dilute hydrochloric acid to yield carbon dioxide. The excess acid is titrated with dilute sodium hydroxide using phenol red as indicator. 0.01 N HCl was added to 200 µl of serum sample. The solution was mixed and 1 drop of phenol red indicator added. The solutions were titrated using 0.01 N sodium hydroxide to attain brick red colorations which serves as endpoint [28]. Bicarbonate was calculated as thus:
Bicarbonate (µmol/l) = 50 – Titre
Titre = Endpoint x 100.
The assay for chloride exploits the formation of chloride precipitate in a sample using mercuric nitrate. When chloride is titrated with standard solution of mercuric ion, undissociated but stable mercuric chloride is formed. The excess mercuric chloride nitrate reacts with diphenylcarbazone to produce a violet coloration. 2 ml of deionized water was added to 200 µl serum sample. This solution was mixed thoroughly and 3 drops diphenylcarbazone indicator and 1 drop of nitric acid was added. The mixture was titrated via mercuric nitrate to give a violet endpoint. The same procedure was repeated for chloride standard solution [29]. Chloride calculation such as:
Chloride (µmol/l) = Titre of sample/Title of standard x 100.
The assay method for sodium and potassium entails the injection of solutions containing these elements in flame leaving solid salt, which dissociates to neutral ground state. The atoms become excited in the flame, thus moving to a higher energy state. The excited atoms then fall back to ground state emitting light of characteristic wavelength (590 nm sodium and 770 nm potassium). The light surpasses through viable filter onto photosensitive element while, the quantity of current flows was relatively the amount of potassium and sodium found in serum sample [30].

2.8 Liver function test

A volume of blood from each rat was put into plain bottles and allowed to clot at room temperature for 4 hours before centrifuging using Hettich centrifuge at 4000 rpm for 10 minutes. The sera obtained were used to assay for liver function test endpoints using standard diagnostic kits on automated clinical system in line with manufacturer instructions. Assays were carried out as follows:
Determination of Serum Alanine Aminotransferase (ALT): The serum alanine aminotransferase was determined as described by Reitman and frankel end-point technique [31].
Determination of Serum Aspartate Aminotransferase (AST)*:* The serum aspartate aminotransferase was determined as described by Reitman and Frankel end-point technique [31].

Determination of Serum Alkaline Phosphatase (ALP): The procedure described by Bassey *et al*. [32] as modified by Wright *et al*. [33] using Randox kits was used for the assay. The substrate p-nitrophenyl phosphate is hydrolyzed by alkaline phosphatase from the sample in the presence of magnesium ions, to form nitrophenol which is yellow and can be read at 405 nm. The intensity of color produced is proportional to the activity of alkaline phosphatase.
Determination of Serum Total Bilirubin and Unconjugated Bilirubin: Total bilirubin can be anticipated via acid diazo technique as illustrate by Doumas *et al.* [34] by assay kits (Randox Laboratories Ltd).
Determination of Albumin: The serum albumin of the samples was determined using the method of Doumas *et al.* [34] using randox assay kit.
Determination of total protein: Total protein being colorimetrically evaluated via Randox assay kit as illustrated by Lowry *et al*, [35].

2.9 Determination of body and organ weight

Weights of rats were determined using My Weigh 7001DX Multi-Purpose Digital Scale on day 0 all through 3 months of the studied period. The net change in body weight (difference between final body weight and initial body weight) was determined for all the animals. On day 29, the animals were sacrificed after an overnight fast. The livers were isolated and their weights determined.

2.10 Lipid profile

Total cholesterol was determined using enzymatic method using wet reagents diagnostic kits. This uses a modified method of Trinder [36]. Total triglycerides were determined using enzymatic method with wet reagent diagnostic kits. This is a modification of the method of Tietz [37]. High density lipoprotein was determined using the enzymatic method. A precipitating agent consisting of 0.55 mmol/l phosphotungstic acid and 25 mmol/l magnesium chloride (Friedewald, 1972) were used.
LDL (mg/dl) = Total cholesterol – Triglycerides/5 - HDL [38].

2.11 Determination of anti-oxidant property of the extract

Superoxide Dismutase (SOD) effect was read via methods described by Bagul *et al.* (2005). Principle; Auto-oxidation with heamatoxylin (increases the absorbance at 560nm wavelength) inhibited by SOD effect of the assay at pH 7.8; percentage amount of SOD present within a specific range. The assayed using Bagul *et al.* (2005) method. Test principle: Catalase scavenging hydrogen peroxide, converted into molecular oxygen and water. Was determined using the method Bagul *et al.* (2005). Test principle: The MDA assay is based on the reaction of MDA with thiobarbituric acid (TBA); forming an MDA-TBA2 adduct that absorbs strongly at 532 nm. Glutathione peroxidase activity was measured in terms of the first order rate constant for the decomposition of tetra-butyl hydroperoxide according to Bagul *et al.* (2005).

2.12 Testosterone assay protocol

Secure the desired number of coated wells in the holder. Dispense 10 µl of standards, specimens and controls into appropriate wells. Dispense 100 µl of Testosterone-HRP Conjugate Reagent into each well. Dispense 50 µl of rabbit anti-Testosterone reagent to each well. Thoroughly mix for 30 seconds. It is very important to mix them completely. Incubate at 37°C for 90 minutes. Remove the incubation mixture by flicking plate contents into a waste container. Rinse and flick the microtiter wells 5 times with deionized or distilled water. Do not use tap water. Strike the wells sharply onto absorbent paper or paper towels to remove all residual water droplets. Dispense 100 µl of TMB Reagent into each well. Gently mix for 5 seconds. Incubate at room temperature for 20 minutes. Stop the reaction by adding 100 µl of Stop Solution to each well. Gently mix 30 seconds. It is important to make sure that all the blue color changes to yellow color completely. Read absorbance at 450 nm with a microtiter well reader within 15 minutes (Chen, 1991).

2.13 Progesterone assay protocol

All reagents should be allowed to reach room temperature (18-25C) before use. Pipette 50 ul of standards (ready to use) and diluted samples into appropriate wells within 5 minutes. Add 100 ul of progesterone Enzyme Conjugate Solution to each well (except those set for blanks). Mix well for 30 sec. and incubate for 60 minutes at 37°C. You may use par film to cover the wells or use appropriate zip-lock bag to store the plate during the incubation. Discard the contents of the wells and wash the plate 5 times with Wash Solution (250-300ul) per well. Invert plate, tap firmly against absorbent paper to remove any residual moisture. Add 100 ul (TMB) Substrate solutions to all wells. Remember to follow the pipetting order. Incubate the plate at room temperature (18-28°C) for 10 minutes without shaking. Stop reaction by adding 50 ul of Stopping Solution to wells in the same sequence that the Substrate Solution was added and gently mixed. Read the absorbance at 450 nm with a microwell reader.

2.14 Luteinizing hormonal assay

Secure the desired number of coated wells in the holder. Dispense 50 ul of standards, specimens, and controls into appropriate wells. Dispense 100 ul of Enzyme Conjugate into each well. Mix for 30 seconds. It is very important to have completed mixing at this step. Incubate at room temperature (~37OC) for 2 hours. Remove the incubation mixture by flicking the plate contents into a waste container. Rinse and flick the microtiter wells five (5) times with wash buffer. Strike the wells sharply onto absorbent paper or paper towels to remove all residual water droplets. Dispense 100ul of TMB solution into each well. Gently mix for 10 seconds. Incubate at room temperature for 20 minutes, in the dark. Stop reaction by adding 50 ul (one drop) of 2N HCl to each well. Gently mix for 30 seconds. It is important to observe a color change from blue to yellow. Read optical

density at 450 nm with a micro titer well reader (Wallace, 1988).

2.15 Follicle stimulating hormone

Bring all reagents and samples to room temperature before use. Centrifuge the sample again after thawing before the assay. It is recommended that all samples and standards be assayed in duplicate. Prepare all reagents and samples as directed in the previous sections. Determine the number of wells to be used and put any remaining wells and the desiccant back into the pouch and seal the ziploc, store unused wells at 4°C. Set a Blank well without any solution. Add 50µl of Standard or Sample per well. Standard need test in duplicate. Add 50µl of HRP-conjugate to each well (not to Blank well), then 50µl Antibody to each well. Mix well and then incubate for 60 minutes at 37°C. Aspirate each well and wash, repeating the process two times for a total of three washes. Wash by filling each well with Wash Buffer (200 µl) using a squirt bottle, multi-channel pipette, manifold dispenser, or auto-washer and let it stand for 10 seconds, complete removal of liquid at each step is essential to good performance. After the last wash, remove any remaining Wash Buffer by aspirating or decanting. Invert the plate and blot it against clean paper towels. Add 50 µl of Substrate A and 50 µl of Substrate B to each well, mix well. Incubate for 15 minutes at 37 °C. Keeping the plate away from drafts and other temperature fluctuations in the dark. Add 50 µl of Stop Solution to each well, gently tap the plate to ensure thorough mixing. Determine the optical density of each well within 10 minutes, using a microplate reader set to 450 nm (Wallace, 1988).

2.16 Histopathological study

Liver, spleen, kidney, stomach, heart and lungs were fixed in neutral buffered formalin. The fixed organs were completely dehydrated using absolute ethanol followed by 96% ethanol, 70% ethanol and then rinsed with distilled water. A 4 µm section was prepared in each case and stained using haematoxylin-eosin dye and the stained tissues were viewed using an optical photomicroscope (Leica MC170 HD, Leica Biosystems, Germany) at x 400 magnification (Drury and Wallinton, 2013).

2.17 Statistical analysis

Results are expressed as mean ± SEM (standard error of the mean) using graph prism 6 version. Data for the groups were compared using one-way analysis of variance with Kruskal-Wallis *post hoc* test. Differences between data were considered significant at $p < 0.05$.

3 Results

Acute toxicity study in Table 1 showed absent toxicological effect with less or no adverse effect in phase 1 and 2 of Locke method.

Results in Table 2 showed the hematological indexes with significant increase to normal range across treated groups of 30, 50 and 100 mg/kg Mojeaga product when compared with untreated group, showed no defect in the haematological parameters. No defect was shown in the female hematological indexes at physiological interval of treated groups at 30, 50 and 100 mg/kg Mojeaga product when compared with untreated group showed a normal haematological indexes as exhibited in Table 3.

Results in Table 4 showed the effect of kidney function test with slight decrease yet in normal range across treated groups of 30, 50 and 100 mg/kg Mojeaga product when compared with untreated group rather elicited renoprotective effect. Results in Table 5 showed the effect of female kidney function test with slight decrease yet in normal values in the treated groups of 30, 50 and 100 mg/kg Mojeaga product when compared with untreated group rather elicited renoprotective effect.

Results in Table 6 showed the effect of liver function test with slight decrease yet in normal range in the treated groups at 30, 50 and 100 mg/kg Mojeaga product when compared with untreated group that showed slight increase in hepatic function test hence promote hepatoprotective effect. Results in Table 7 showed the effect female liver function test with slight decrease in across the treated groups (30, 50 and 100 mg/kg) of Mojeaga product when compared with untreated group that showed slight increase in hepatic function test thereby elicited hepatoprotective effect.

Results in Table 8 showed the body weight with slight increase in the treated groups of 30, 50 and 100 mg/kg Mojeaga product when compared with untreated group with slight increase as the treated group. Results in table 9 showed the body weight with slight increase at normal weight increase across treated groups of 30, 50 and 100 mg/kg Mojeaga product when compared with untreated group with slight increase as the treated group.

Results from the organ weight with slight increase in the treated groups specifically at 50 and 100 mg/kg Mojeaga product when compared with untreated group showed that it cause no pathological defect across the studied organs as shown in Table 10

Results in Table 11 showed that the organ weight has slight increase across treated groups of 30, 50 and 100 mg/kg Mojeaga product when compared with untreated control, showed that it causes no pathological defect across the studied organs.

The indexes used to evaluate toxicity study are associated with lipidaemic properties such as Triglycerides, Total Cholesterol, Very Low-Density Lipoproteins, High density Lipoproteins and Low-Density Lipoproteins. Mojeaga herbal remedy at graded doses (150, 300 and 600 mg/kg) maintained TAG, TC and LDL concentrations within normal ranges, with a slight increase in HDL concentration across male and female rats when compared with the control (Figure 1 and 2).

Mojeaga herbal remedy did not cause overproduction of free radicals to the systems because MDA levels is maintained at normal values in the treated groups of male and female animals when compared with the control. The only significant change noticed in MDA across all treated groups.

Mojeaga herbal remedy controlled the level of oxidants, probably due to the presence of natural antioxidant compounds. CAT together with SOD and GPx constitute the primary enzymatic defense, catalyzing decomposition of ROS. Chronic protective effect of Mojeaga herbal remedy increases CAT and SOD activities when compared with the control (Figure 3 and 4).

This study investigated, the male and female animals treated with 150, 300 and 600 mg/kg of Mojeaga herbal remedy were observed to elicit a slight significant increase in the concentration of serum testosterone when compared with the control. Concerning the effect of chronic Mojeaga herbal remedy intake, facilitates the increase in the level of luteinizing hormone and follicle stimulating hormone as shown in Figure 5 and 6.

Result in Figure7 and 8 showed that male and female animals treated with 150, 300 and 600 mg/kg of Mojeaga herbal remedy showed normal architural frame work of the hepatic cells when compared with the control.

The renoprotective effect of the renal cell across the male and female treated groups at 150, 300 and 600 mg/kg of Mojeaga herbal remedy with normal histology of kidney cells when compared with the control as shown in Figure 9 and 10.

Cardiac cell is vital in the body aid in the circulation of blood flow. Hence, result obtained from Figure11 and 12 showed that the male and female animals in the treatment groups exhibited normal function of the heart when compared with the control group.

The protection effect of the lungs across the male and female treated groups at 150, 300 and 600 mg/kg of Mojeaga herbal remedy showed a normal histology of the alveolar cells when compared with the control as shown in Figure 13 and 14.

Spleen is a vital organ in the body aid in stimulating the immune system. Hence, result obtained from Figure15 and 16 showed that the male and female animals in the treatment groups elicited normal function of the heart when compared with the control group.

The brain cells aid in regulating normal functioning of the entire body. Figure 17 and 18 showed that the male and female rats treated with 150, 300 and 600 mg/kg of Mojeaga herbal remedy exhibit a normal histology of the brain when compared.

Several substances interfere with the mucosa lining of the stomach either by inhibiting the release of prostaglandins responsible for the build of the stomach wall. Male and female treated groups at 150, 300 and 600 mg/kg of Mojeaga herbal remedy showed normal architectural structure of the mucosa lining when compared with the control as shown in Figure 19 and 20.

The testes aid normal functioning of the organ during reproduction. Figure 21 showed that the male rats across treatment groups' physiological state of the treated groups when compared with untreated control. Uterus is a stimulating organ that promotes the release of vital hormones during and after child birth. Hence, result obtained from Figure 22 showed that the female animals in the treatment groups elicited normal function of the uterus when compared with the control.

Table 1. Acute toxicological effect of Mojeaga on Swiss mice

Groups	Doses (mg/kg)	No of lethality	% mortality
Control	DW	0/5	0
Mojeaga	10	0/5	0
Mojeaga	100	0/5	0
Mojeaga	1000	0/5	0
Mojeaga	1600	0/5	0
Mojeaga	2900	0/5	0
Mojeaga	5000	0/5	0
Mojeaga	10000	1/5	20

Mean ± SEM (n=5). DW-distilled water

Table 2. Effect of Mojeaga herbal remedy on male Hematological indices

Parameters	Control	150 mg/kg Mojeaga	300 mg/kg Mojeaga	600 mg/kg Mojeaga
WBC x 10^3 / ul	12.13±0.87	18.40±3.28	12.20±1.57	17.20±5.22
LY x 10^3 / ul	9.60±0.78	14.17±2.38	9.30±1.26	14.00±4.62
MO x 10^3 / ul	1.57±0.07	2.57±0.54	1.63±0.07	2.10±0.61
GR x 10^3 / ul	0.97±0.03	1.70±0.42	1.27±0.26	1.10±0.06
LY%	79.13±0.81	77.20±1.45	75.93±0.78	80.30±2.14
MO%	12.90±0.57	13.80±1.05	13.90±1.42	12.30±1.07
GR%	7.97±0.34	9.00±0.74	10.17±0.88	7.40±1.75
RBC x 10^6 / ul	6.64±0.14	7.62±0.03	7.38±0.37	7.54±0.28
Hgb.g/d l	13.63±0.33	14.80±0.27	14.87±0.55	14.47±0.37
HCT%	39.37±1.13	41.50±1.03	41.73±1.81	41.77±1.41
MCV.f l	59.23±0.50	54.47±1.51	56.53±0.61	55.43±0.22
MCH.pg	20.50±0.10	19.43±0.37	19.90±0.10	19.20±0.27
MCHC.g/d l	34.63±0.15	35.67±0.48	35.63±0.24	34.67±0.37
RDWC%	16.87±0.37	17.13±0.54	17.30±0.61	17.07±0.84
RDWS.f l	34.17±0.57	31.57±1.02	34.13±1.33	33.50±1.04
PLT x 10^3 /ul	685.30±21.87	765.00±30.05	733.00±29.51	738.30±40.01
PCT x 10^3 /ul	0.51±0.03	0.57±0.02	0.56±0.02	0.53±0.04
MPV.f l	7.40±0.45	7.43±0.12	7.70±0.25	7.20±0.15
PDW %	15.10±0.82	16.03±0.72	15.57±0.87	15.47±0.61
PLCR %	8.27±1.87	8.83±0.54	10.47±1.37	7.03±0.49

p-value > superscript a= 0.05, b= 0.01 showed the level of significant difference when compared with untreated control, (n=5). DW- distilled water

Table 3. Effect of Mojeaga herbal remedy on female Hematological indices

Parameters	Control	150 mg/kg Mojeaga	300 mg /kg Mojeaga	600 mg/kg Mojeaga
WBC x 10^3 / ul	12.77±0.82	16.90±2.24	13.97±1.25	12.63±0.96
LY x 10^3 / ul	9.37±0.49	11.93±1.34	10.17±0.83	9.17±0.44
MO x 10^3 / ul	1.93±0.30	2.13±0.30	1.73±0.34	2.10±0.32
GR x 10^3 / ul	1.47±0.17	2.83±0.73	2.07±0.81	1.40±0.35
LY%	73.47±2.21	71.07±3.08	73.67±6.50	72.93±4.01
MO%	15.10±1.60	12.63±0.18	12.40±1.91	16.33±1.57
GR%	11.60±0.64	16.30±2.91	13.93±5.11	10.73±2.47
RBC x 10^6 / ul	7.95±0.17	8.09±0.19	8.50±0.29	8.50±0.46
Hgb.g/d l	15.43±0.07	15.70±0.12	16.00±0.27	16.57±0.5
HCT%	45.67±0.47	45.80±0.57	46.97±0.84	45.50±1.46
MCV.f l	53.43±1.48	56.67±0.72	55.43±2.42	53.70±1.41
MCH.pg	19.13±0.35	19.43±0.32	18.87±0.54	19.57±0.44
MCHC.g/dl	35.83±0.38	34.30±0.31	34.07±0.58	36.40±0.27
RDWC%	19.13±1.53	18.90±0.51	18.67±0.41	17.03±0.63
RDWS. f l	33.10±2.14	35.77±1.29	33.90±1.70	30.73±0.62
PLT x 10^3 /ul	686.00±80.31	686.30±39.1	682.70±15.59	700.3±8.33
PCT x 10^3 /ul	0.52±0.07	0.52±0.04	0.522±0.01	0.51±0.01
MPV. f l	7.53±0.20	7.60±0.35	7.67±0.32	7.27±0.09
PDW %	19.13±1.42	16.50±0.32	16.87±0.70	17.10±1.90
PLCR %	9.13±0.99	9.03±2.27	9.47±1.82	7.43±0.88

p-value > superscript a= 0.05, b= 0.01 showed the level of significant difference when compared with untreated control, (n=5). DW- distilled water.

Table 4. Effect of Mojeaga herbal remedy on male kidney function test

Treatment	Doses mg/kg	Urea	Creatinine	Bicarbonate	Sodium Na^+	Potassium K^+	Chlorides Cl^-
Control	DW	25.61±0.02^a	0.37±0.01^a	12.91±0.05^a	94.99±0.24^a	28.40±0.13^a	72.97±0.05^a
Mojeaga	150	24.14±0.02^a	0.36±0.01^a	11.46±0.02^a	98.38±0.04^a	26.08±0.02^a	72.33±0.02^a
Mojeaga	300	30.58±0.02^b	0.47±0.02^b	10.17±0.02^b	103.20±0.91^b	24.79±0.41^b	75.00±0.16^a
Mojeaga	600	28.19±0.21^a	0.41±0.01^a	15.79±0.23^a	99.08±0.02^a	27.78±0.17^a	73.46±0.03^a

p-value > superscript a= 0.05, b= 0.01 showed the level of significant difference when compared with untreated control, (n=5). DW- distilled water.

Table 5. Effect of Mojeaga herbal remedy on female kidney function test

Treatment	Doses mg/kg	Urea	Creatinine	Bicarbonate	Sodium Na^+	Potassium K^+	Chlorides Cl^-
Control	DW	26.67±0.02^a	0.28±0.01^a	12.18±0.04^a	98.51±0.02^a	26.16±0.01^a	73.60±0.03^a
Mojeaga	150	24.05±0.01^a	0.25±0.02^a	13.53±0.01^a	98.46±0.03^a	25.87±0.02^a	72.92±0.05^a
Mojeaga	300	27.24±0.02^b	0.39±0.01^b	11.42±0.02^a	110.80±0.06^b	24.31±0.01^a	76.01±0.08^a
Mojeaga	600	27.13±0.25^a	0.40±0.02^b	13.82±0.01^a	98.79±0.18^a	25.65±0.02^a	74.63±0.02^a

p-value > superscript a= 0.05, b= 0.01 showed the level of significant difference when compared with untreated control, (n=5). DW- distilled water.

Table 6. Effect of Mojeaga herbal remedy on male liver function tests

Treatment	Doses mg/kg	ALT	AST	GGT	Total bilirubin	Conjugated bilirubin	Unconjugated bilirubin	Total protein
Control	DW	28.97±0.30^a	45.99±0.12^a	52.48±0.20^a	0.16±0.01^a	0.08±0.01^a	0.08±0.00^a	12.42±0.05^a
Mojeaga	150	28.33±0.02^a	45.29±0.01^a	52.30±0.02^a	0.16±0.01^a	0.09±0.00^a	0.07±0.01^a	12.55±0.23^a
Mojeaga	300	39.35±0.34^a	50.29±0.02^b	57.59±0.05^a	0.19±0.00^a	0.08±0.00^a	0.11±0.00^a	14.41±0.65^a
Mojeaga	600	31.36±0.04^a	48.66±0.02^a	54.67±0.02^a	0.13±0.01^a	0.09±0.01^a	0.04±0.01^a	12.28±0.05^a

p-value > superscript a= 0.05, b= 0.01 showed the level of significant difference when compared with untreated control, (n=5). DW- distilled water.

Table 7. Effect of Mojeaga herbal remedy on female liver function test

Treatment	Doses mg/kg	ALT	AST	GGT	Total bilirubin	Conjugated bilirubin	Unconjugated bilirubin	Total protein
Control	DW	30.32±0.19[a]	41.83±0.60[a]	49.19±0.06[a]	0.15±0.03[a]	0.07±0.02[a]	0.46±0.19[a]	13.89±0.58[a]
Mojeaga	150	32.41±0.24[a]	49.40±0.01[b]	84.15±0.09[b]	0.18±0.01[a]	0.08±0.02[a]	0.12±0.04[a]	14.11±0.06[a]
Mojeaga	300	34.12±0.07[b]	47.40±0.23[a]	65.29±3.00[a]	0.16±0.01[a]	0.11±0.01[a]	0.05±0.01[b]	12.10±0.06[a]
Mojeaga	600	33.19±0.61[a]	46.57±0.29[a]	64.00±0.58[a]	0.15±0.03[a]	0.09±0.1[a]	0.06±0.01[a]	14.37±0.17[a]

p-value > superscript a= 0.05, b= 0.01 showed the level of significant difference when compared with untreated control, (n=5). DW- distilled water.

Table 8. Effect of Mojeaga herbal remedy on male body mass indexes

Treatment/days	Control	150 mg/kg Mojeaga	300 mg/kg Mojeaga	600 mg/kg Mojeaga
1	218.30±5.24	299.30±10.67	234.30±11.29	271.00±23.76
7	240.7±5.23	331.3±13.97	248.3±15.77	261.70±12.55
14	252.00±2.65	337.70±14.33	256.70±17.38	269.00±12.66
21	259.30±4.41	341.30±15.50	270.70±14.90	272.30±11.79
28	267.30±2.33	350.00±15.95	281.70±15.17	275.00±11.68
35	273.00±2.31	351.00±21.94	288.30±16.83	279.30±11.79
42	277.00±2.31	356.00±34.03	293.00±16.56	283.10±11.72
49	281.70±2.33	359.00±32.70	297.00±17.04	289.00±11.53
56	288.70±1.76	367.70±31.43	288.70±18.56	286.70±12.84
63	292.00±1.53	278.00±29.70	288.70±19.47	284.30±15.68
70	295.00±0.58	276.70±29.23	285.70±18.02	278.20±14.34
77	298.00±1.53	274.00±29.48	279.70±16.70	270.50±12.45
84	303.00±1.73	271.30±29.99	276.30±17.57	261.80±11.14

Mean±SEM (n=5). DW- distilled water.

Table 9. Effect of Mojeaga herbal remedy on female body mass indexes

Treatment/days	Control	150 mg/kg Mojeaga	300 mg/kg Mojeaga	600 mg/kg Mojeaga
1	153.70±2.33	168.30±1.20	184.30±26.83	153.70±2..40
7	191.70±3.84	193.00±6.43	193.30±20.00	184.70±3.76
14	196.00±6.43	198.70±4.98	193.30±20.00	184.70±3.76
21	204.70±5.78	204.70±7.06	201.30±18.41	215.70±15.30
28	201.00±6.11	211.30±7.27	205.30±17.37	226.00±14.01
35	205.50±2.01	217.10±3.21	211.50±11.00	232.40±10.11
42	211.30±4.12	225.20±5.01	216.30±9.98	237.90±11.00
49	216.60±7.06	231.00±4.91	224.70±10.15	243.50±16.04
56	223.80±5.56	239.90±5.55	232.40±8.07	257.10±12.21
63	231.30±1.74	244.20±6.02	239.80±9.69	263.50±8.91
70	237.60±3.14	244.30±3.76	225.10±9.69	253.60±9.34
77	243.10±6.03	241.00±4.18	220.00±6.25	246.20±8.00
84	249.90±4.83	234.30±5.63	213.10±7.01	236.70±3.99

Mean±SEM (n=5). DW- distilled water.

Table 10. Effect of Mojeaga herbal remedy on male percentage organs weight

Treatment/ organs %	Control	150 mg/kg Mojeaga	300 mg/kg Mojeaga	600 mg/kg Mojeaga
Heart	0.33±0.00[a]	0.32±0.03[a]	0.34±0.09[a]	0.37±0.18[a]
Kidney	0.57±0.03[a]	0.69±0.20[a]	0.62±0.21[a]	0.62±0.12[a]
Liver	2.66±0.44[a]	3.78±0.75[a]	3.69±1.00[a]	3.14±0.19[a]
Lungs	0.69±0.12[a]	0.79±0.23[a]	0.87±0.25[a]	0.87±0.03[a]
Stomach	0.59±0.15[a]	0.91±0.41[b]	0.86±0.41[a]	0.59±0.15[a]
Spleen	0.26±0.06[a]	0.38±0.15[a]	0.32±0.13[a]	0.41±0.07[b]
Brain	0.36±0.12[a]	0.57±0.12[a]	0.58±0.15[a]	0.62±0.03[b]
Testes	0.96±0.06[a]	1.13±0.19[a]	1.09±0.12[a]	1.02±0.88[a]

Mean±SEM (n=5). DW- distilled water.

Table 11. Effect of Mojeaga herbal remedy on female percentage organs weight

Treatment/organs %	Control	150 mg/kg Mojeaga	300 mg/kg Mojeaga	600 mg/kg Mojeaga
Heart	0.28±0.06[a]	0.27±0.03[a]	0.32±0.03[a]	0.30±0.06[a]
Kidney	0.55±0.03[a]	0.61±0.09[a]	0.56±0.15[a]	0.51±0.00[a]
Liver	3.16±0.10[a]	2.86±0.36[a]	3.38±1.14[a]	2.62±0.27[a]
Lungs	0.73±0.09[a]	0.84±0.07[a]	0.83±0.09[a]	1.06±0.40[b]
Stomach	0.63±0.15[a]	0.74±0.22[a]	0.70±0.06[a]	0.96±0.41[b]
Spleen	0.31±0.03[a]	0.30±0.06[a]	0.33±0.06[a]	0.33±0.07[a]
Brain	0.56±0.06[a]	0.61±0.09[a]	0.62±0.18[a]	0.52±0.03[a]
Uterus	0.19±0.09[a]	0.13±0.06[a]	0.17±0.03[a]	0.18±0.03[a]

Mean±SEM (n=5). DW- distilled water.

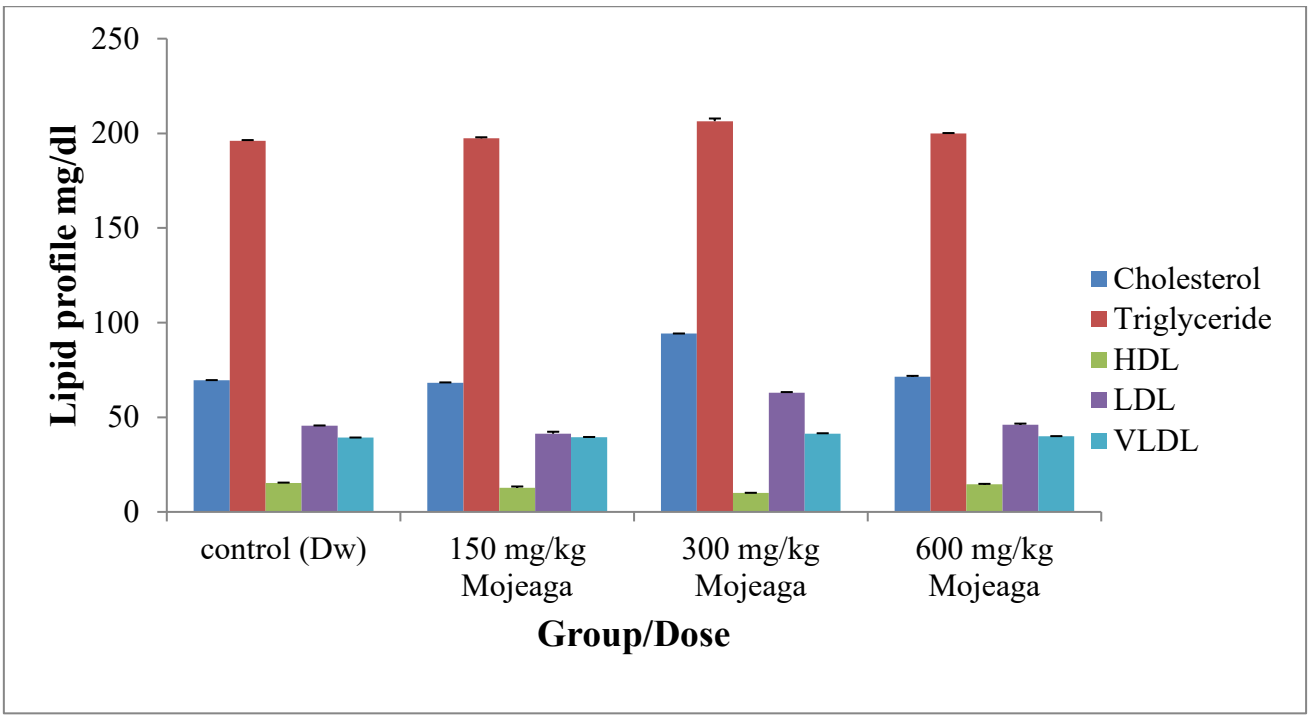

Figure 1. Effect of Mojeaga herbal remedy on Male lipid profile

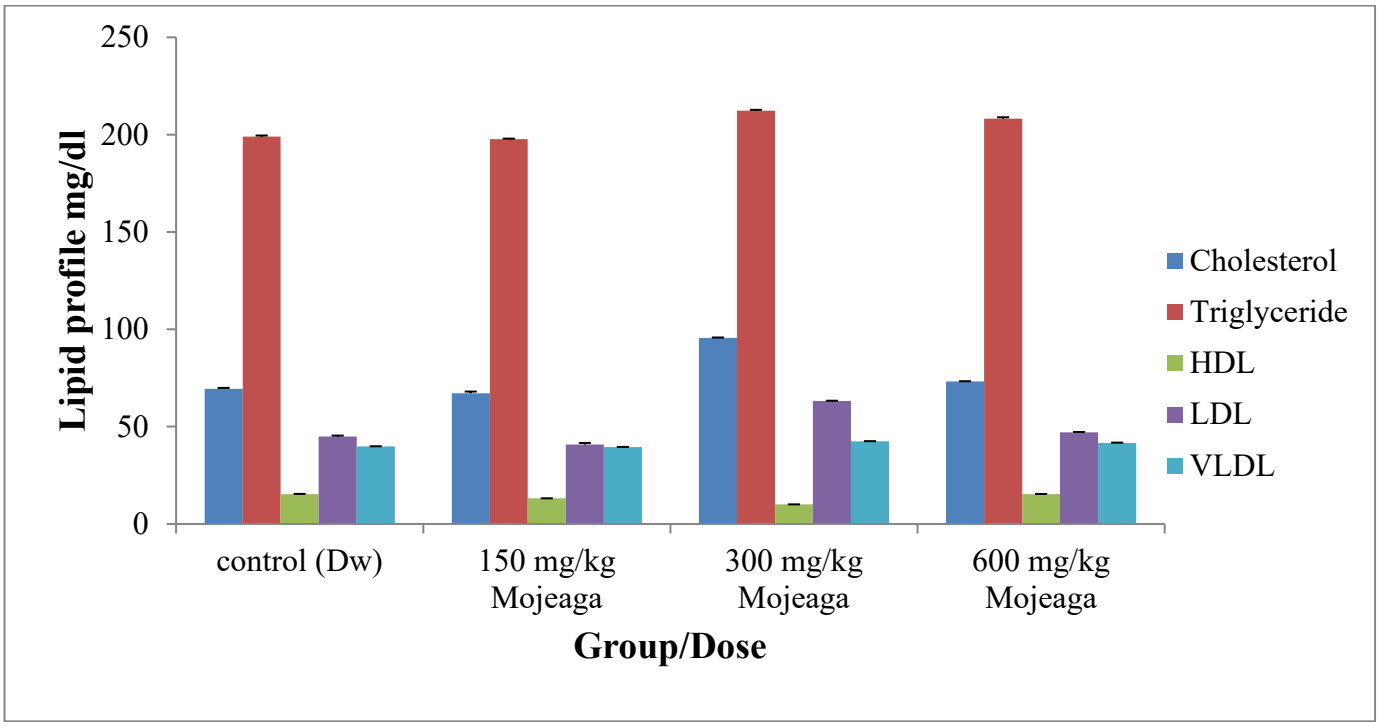

Figure 2. Effect of Mojeaga herbal remedy on Female lipid profile

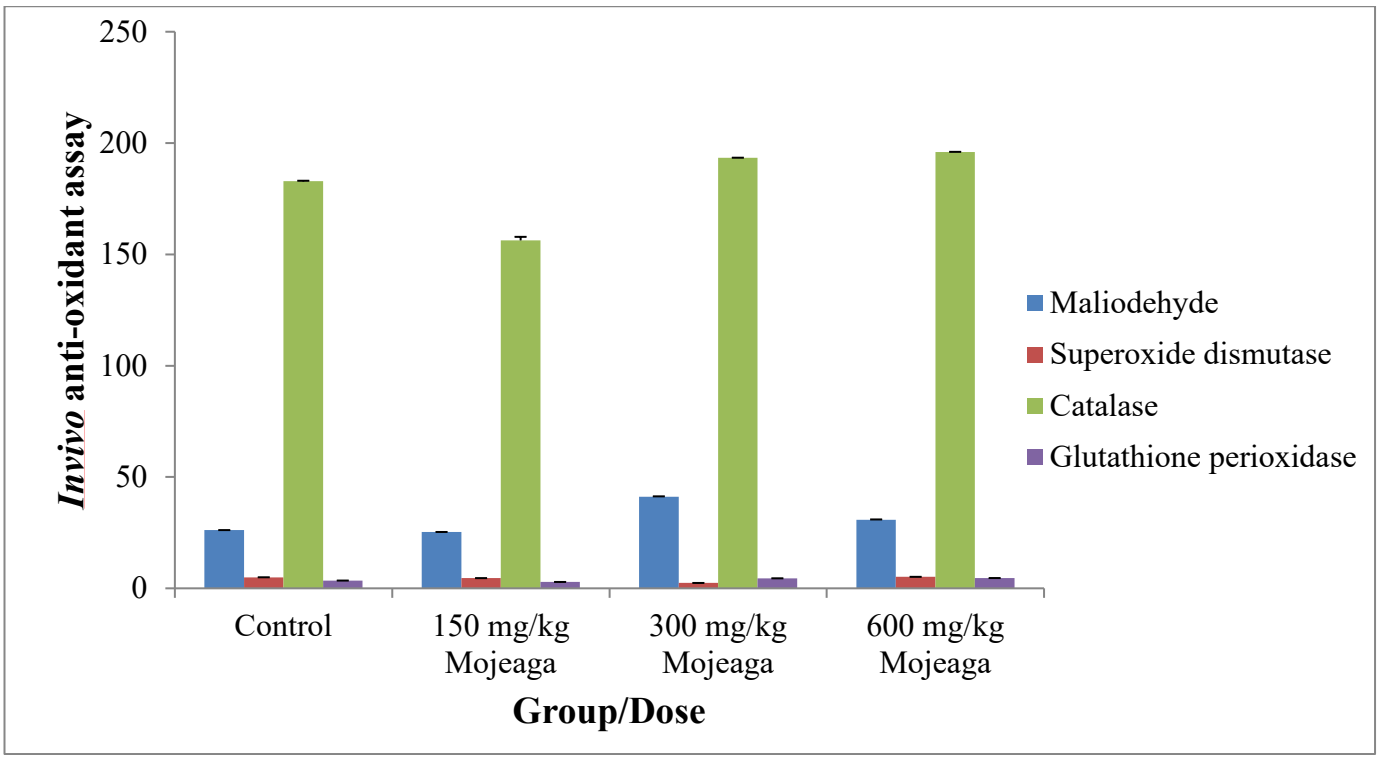

Figure 3. Effect of Mojeaga herbal remedy on male *in-vivo* antioxidant study

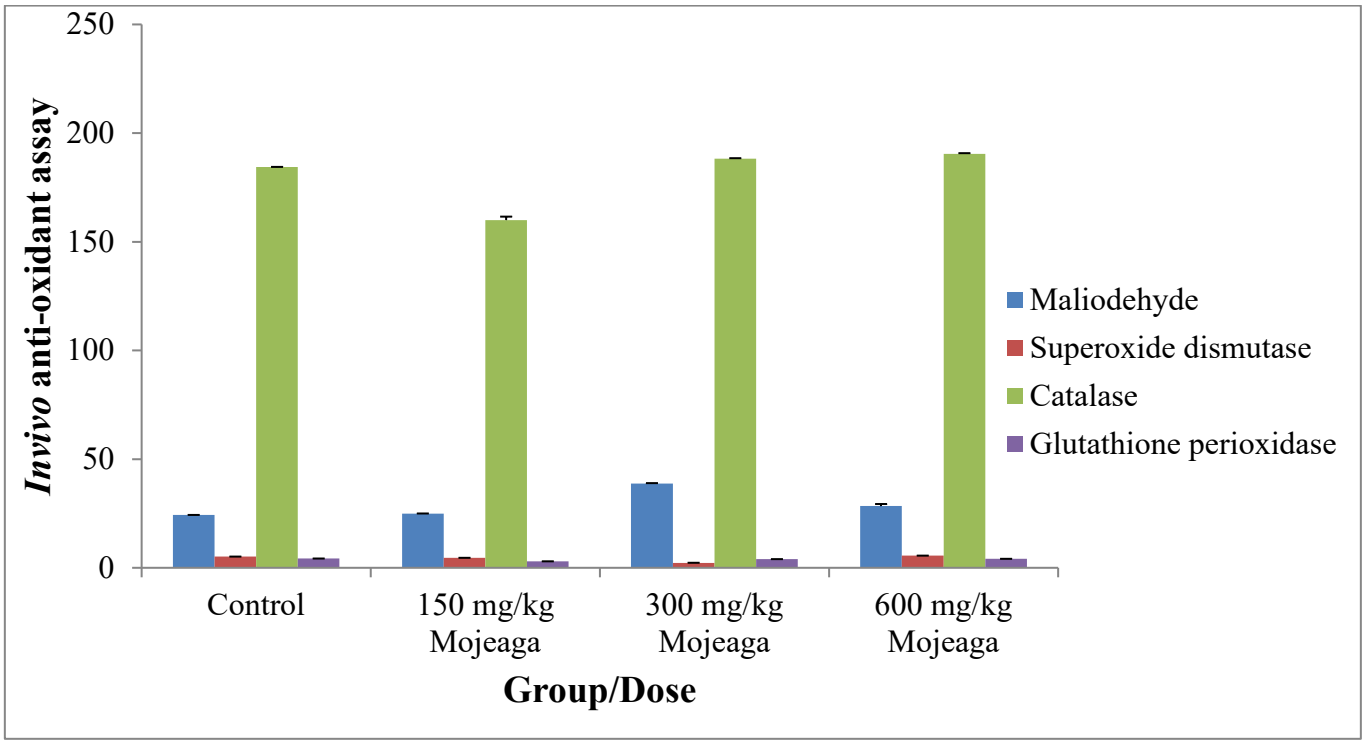

Figure 4. Effect of Mojeaga herbal remedy on female *in-vivo* antioxidant study

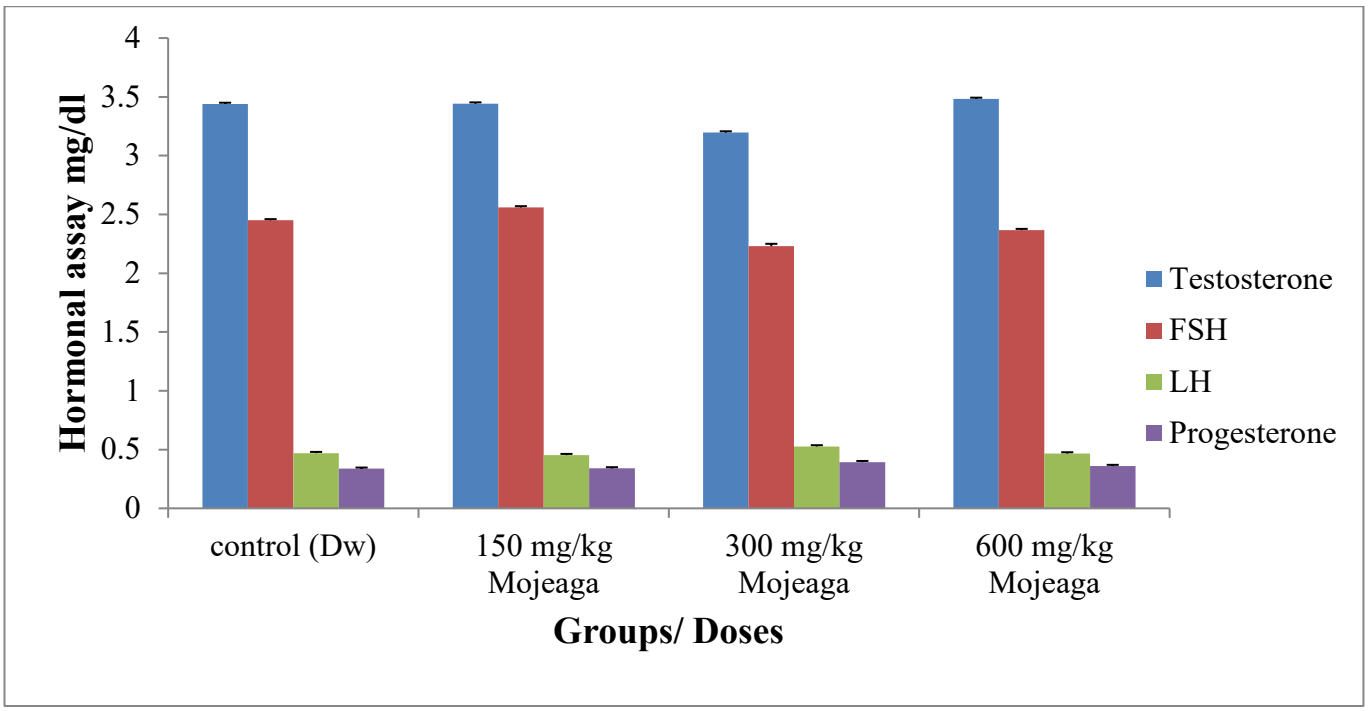

Figure 5. Effect of Mojeaga herbal remedy on male reproductive hormones

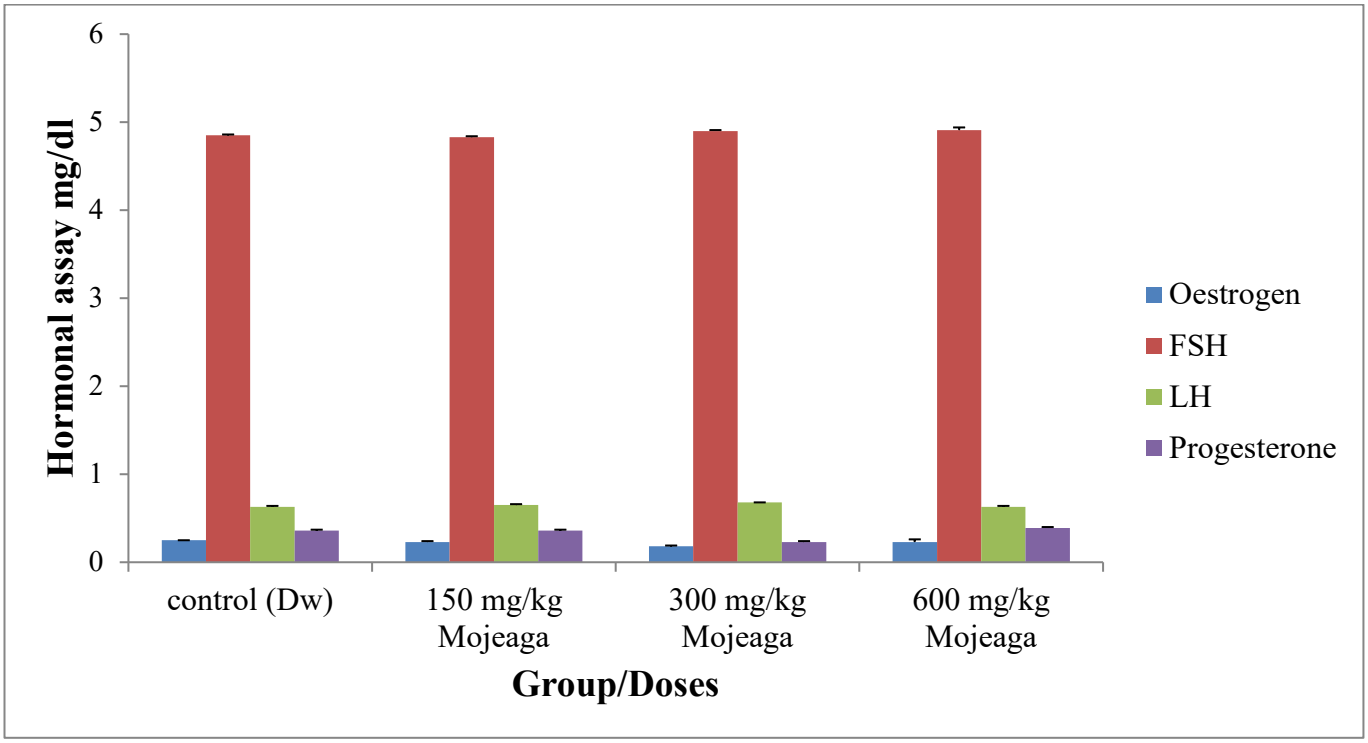

Figure 6. Effect of Mojeaga herbal remedy on female reproductive hormones

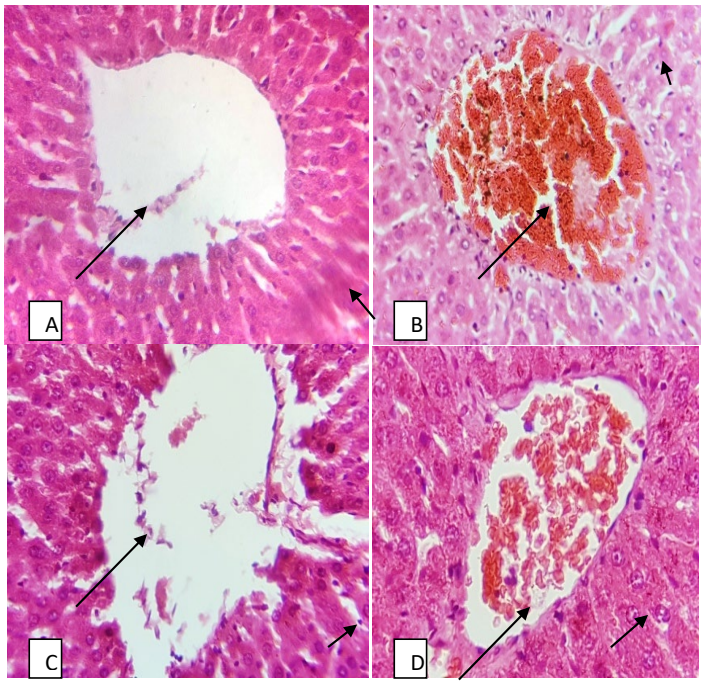

Figure 7. Toxicological effect of Mojeaga herbal remedy on male hepatic cells
A. 150 mg/kg Mojeaga LIVER: reveals visible centriole (long arrow) with the hepatocytes and well fenestrated sinusoidal space (Short arrow).
B. 300 mg/kg Mojeaga LIVER: reveals congested centriole surrounded by mild inflammatory cells (long arrow) with the hepatocytes and well fenestrated sinusoidal space (short arrow).
C. 600 mg/kg Mojeaga LIVER: reveals dilated centriole (long arrow) with the hepatocytes having slighted pyknotic nucleus and well fenestrated sinusoidal space (short arrow).
D. Control LIVER: reveals visible centriole (long arrow) with the hepatocytes having slighted pyknotic nucleus and well fenestrated sinusoidal space (short arrow).

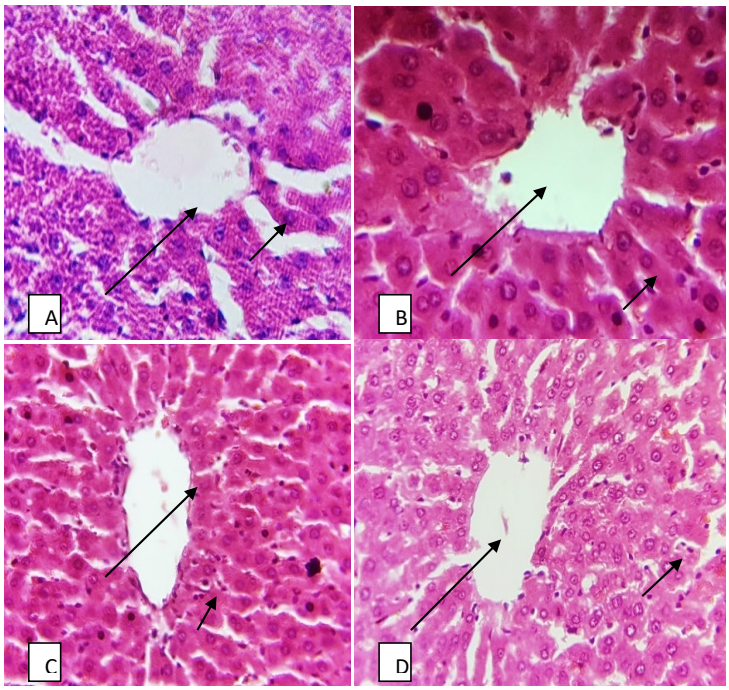

Figure 8. Toxicological effect of Mojeaga herbal remedy on female hepatic cells
A. 150 mg/kg Mojeaga LIVER: reveals distinct centriole (long arrow) with the hepatocytes and well fenestrated sinusoidal space (short arrow).
B. 300 mg/kg Mojeaga LIVER: reveals centriole (long arrow) with the hepatocytes having pyknotic nucleus and well fenestrated sinusoidal space (short arrow).
C. 600 mg/kg Mojeaga LIVER: reveals visible centriole (long arrow) with the hepatocytes having slighted pyknotic nucleus and well fenestrated sinusoidal space (short arrow).
D. Control LIVER: reveals visible centriole (long arrow) with the hepatocytes having slighted pyknotic nucleus and well fenestrated sinusoidal space (short arrow).

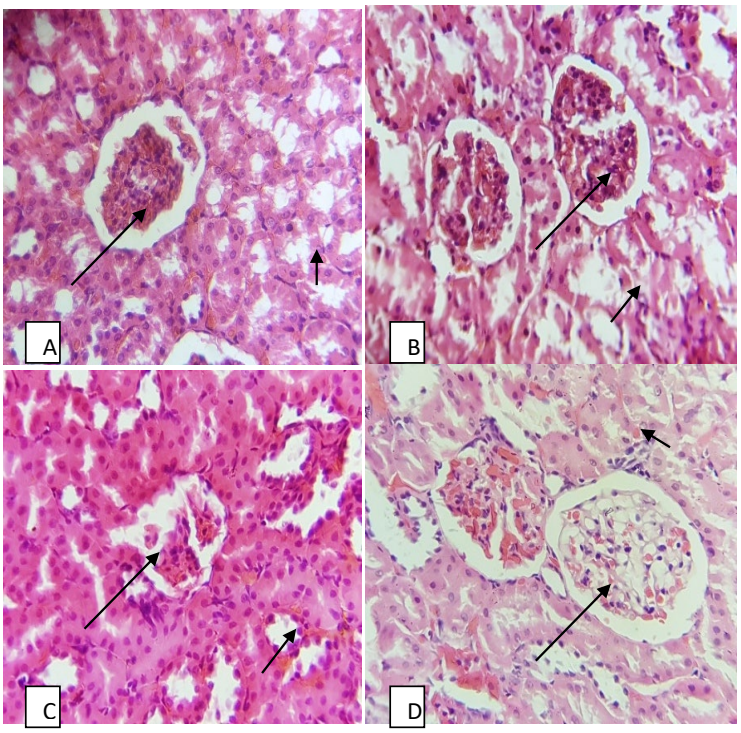

Figure 9. Toxicological effect of Mojeaga herbal remedy on male renal cells.
150 mg/kg Mojeaga KIDNEY: reveals visible renal corpuscle (long arrow) and interstitial space (short arrow) and tubules.
300 mg/kg Mojeaga KIDNEY: reveals visible renal corpuscle (long arrow) and interstitial space (short arrow) and tubules.
600 mg/kg Mojeaga KIDNEY: reveals distorted renal corpuscle with atrophied glomerulus (long arrow) and interstitial space (short arrow) and not so prominent tubules.
Control KIDNEY: reveals enlarged renal corpuscle (long arrow) and interstitial space (short arrow) and not so prominent tubules.

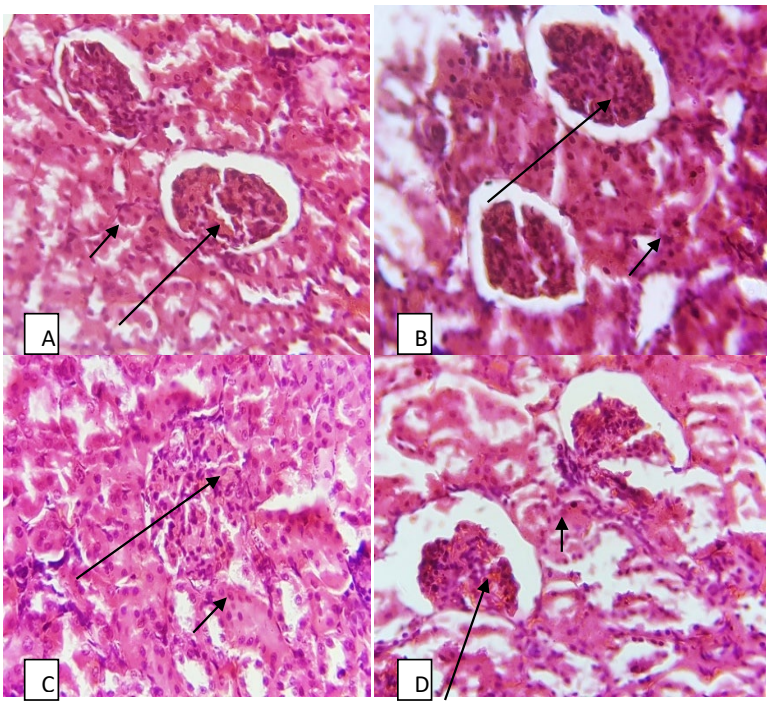

Figure 10. Toxicological effect of Mojeaga herbal remedy on female renal cells
A. 150 mg/kg Mojeaga KIDNEY: reveals visible renal corpuscle (long arrow) and interstitial space (short arrow) and tubules.
B. 300 mg/kg Mojeaga KIDNEY: reveals visible renal corpuscle (long arrow) and interstitial space (short arrow) and tubules.
C. 600 mg/kg Mojeaga KIDNEY: reveals enlarged renal corpuscle (long arrow) and interstitial space (short arrow) and not so prominent tubules.
D. Control KIDNEY: reveals renal corpuscle (long arrow) and interstitial space (short arrow) and visible tubules.

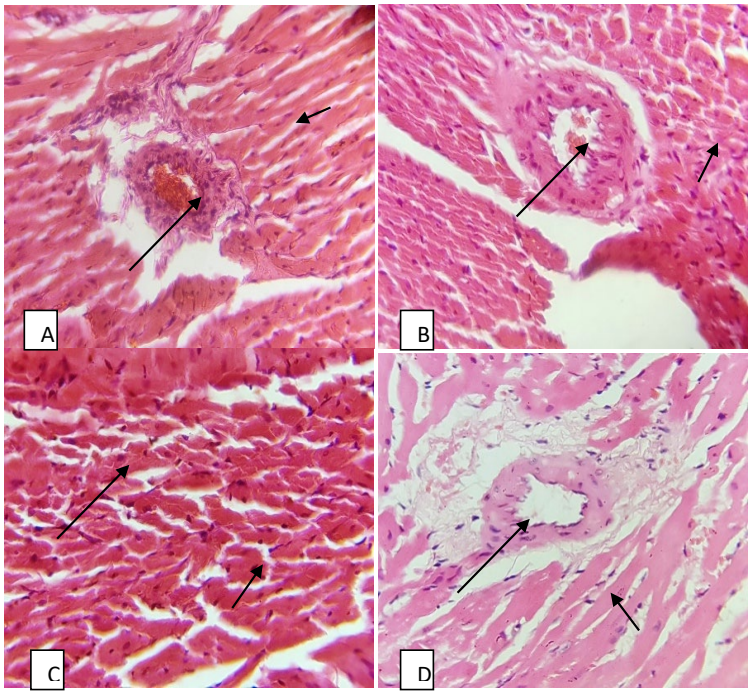

Figure 11. Toxicological effect of Mojeaga herbal remedy on male cardiac cells.
A. 150 mg/kg Mojeaga HEART: composed of bundles of myocardial fibres with visible mononuclear cells (short arrow), interstitial space and visible coronary artery (long arrow).
B. 300 mg/kg Mojeaga HEART: composed of bundles of myocardial fibres (short arrow), interstitial space and coronary artery (long arrow).
C. 600 mg/kg Mojeaga HEART: composed of bundles of myocardial fibres (short arrow), interstitial space.
D. Control HEART: composed of bundles of myocardial fibres (short arrow), interstitial space and prominent coronary artery (long arrow).

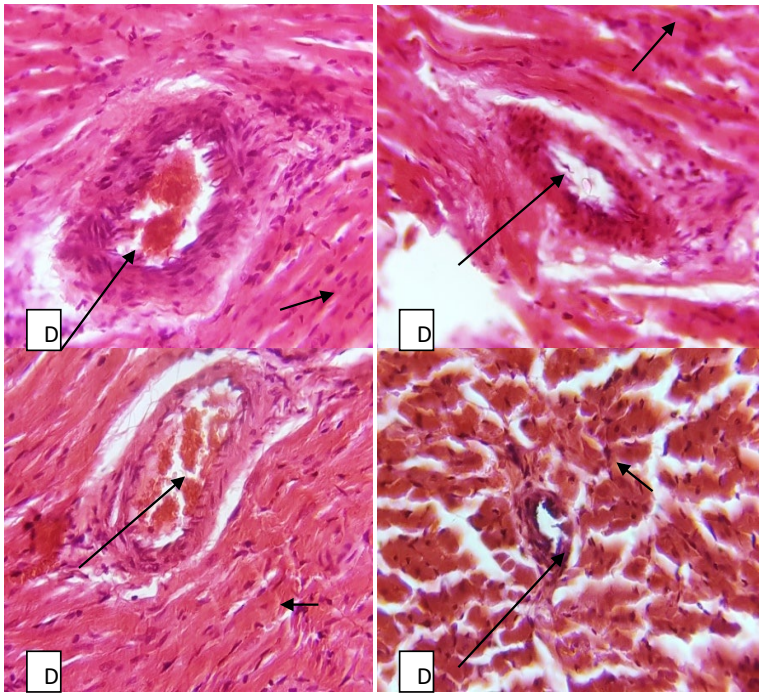

Figure 12. Toxicological effect of Mojeaga herbal remedy on female cardiac cells
A. 150 mg/kg Mojeaga HEART: composed of bundles of myocardial fibres (short arrow), interstitial space and slightly dilated coronary artery (long arrow)
B. 300 mg/kg Mojeaga HEART: composed of bundles of myocardial fibres (short arrow), interstitial space and coronary artery (long arrow).
C. 600 mg/kg Mojeaga HEART: composed of bundles of myocardial fibres (short arrow), interstitial space and enlarged dilated coronary artery (long arrow).
D. Control HEART: composed of bundles of myocardial fibres (short arrow), interstitial space and prominent coronary artery (long arrow).

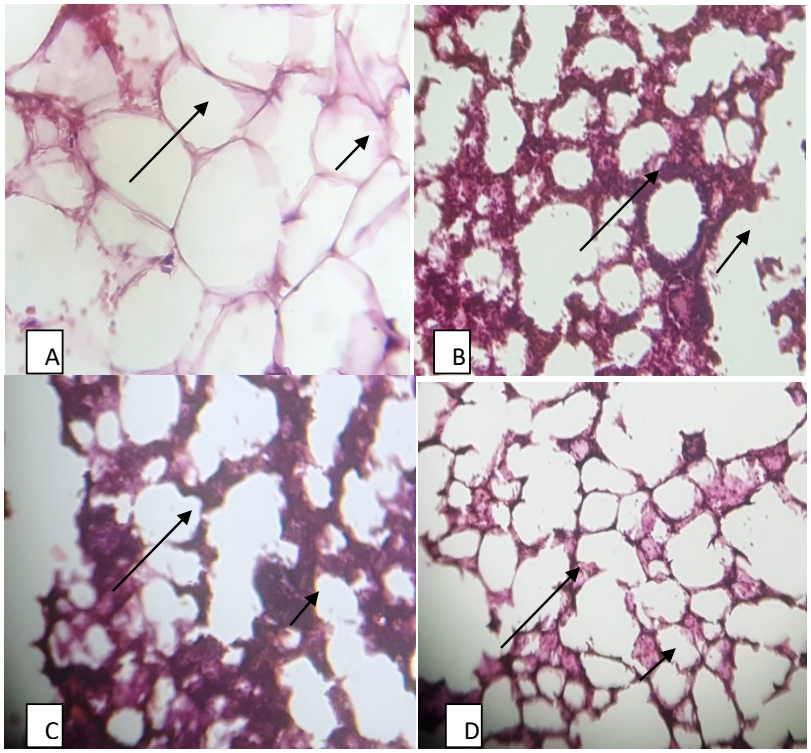

Figure 13. Toxicological effect of Mojeaga herbal remedy on male lungs.
A. 150 mg/kg Mojeaga LUNG: histology with prominent alveolar sac (long arrow) and alveolar ring.
B. 300 mg/kg Mojeaga LUNG: histology with thickened alveolar sac (long arrow) and alveolar ring.
C. 600 mg/kg Mojeaga LUNG: histology with prominent thickened alveolar sac (long arrow) and alveolar ring.
D. Control LUNG: histology with prominent alveolar sac (long arrow) and alveolar ring.

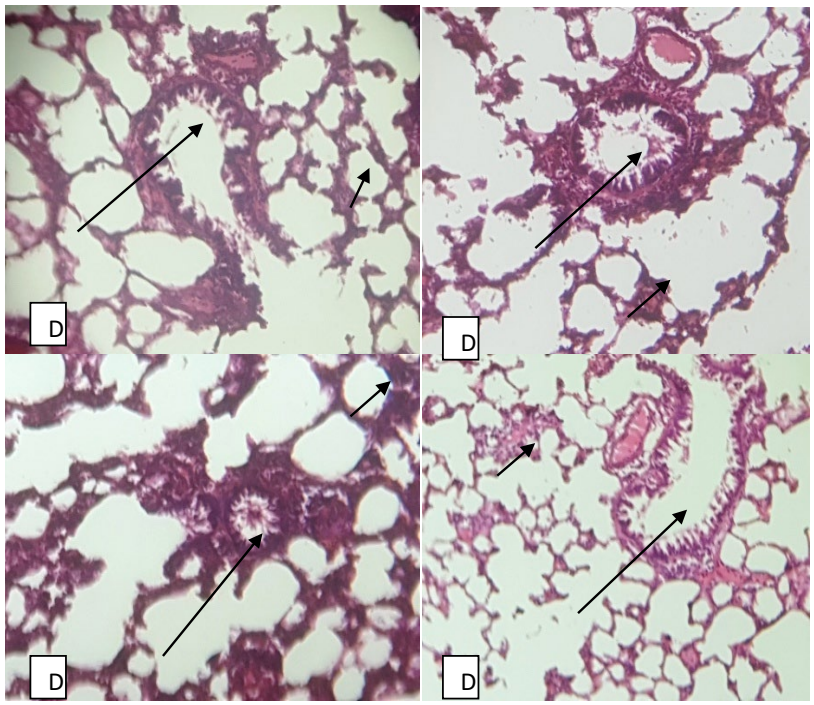

Figure 14. Toxicological effect of Mojeaga herbal remedy on female Lungs
A. 150 mg/kg Mojeaga LUNG: histology with prominent bronchiole with visible alveolar sac (long arrow) and alveolar ring.
B. 300 mg/kg Mojeaga LUNG: histology with prominent bronchiole and slightly thickened alveolar sac (long arrow) and alveolar ring (short arrow).
C. 600 mg/kg Mojeaga LUNG: histology with visible bronchiole and thickened alveolar sac (long arrow) and alveolar ring.
D. Control LUNG: histology with visible bronchiole and prominent alveolar sac (long arrow) and alveolar ring.

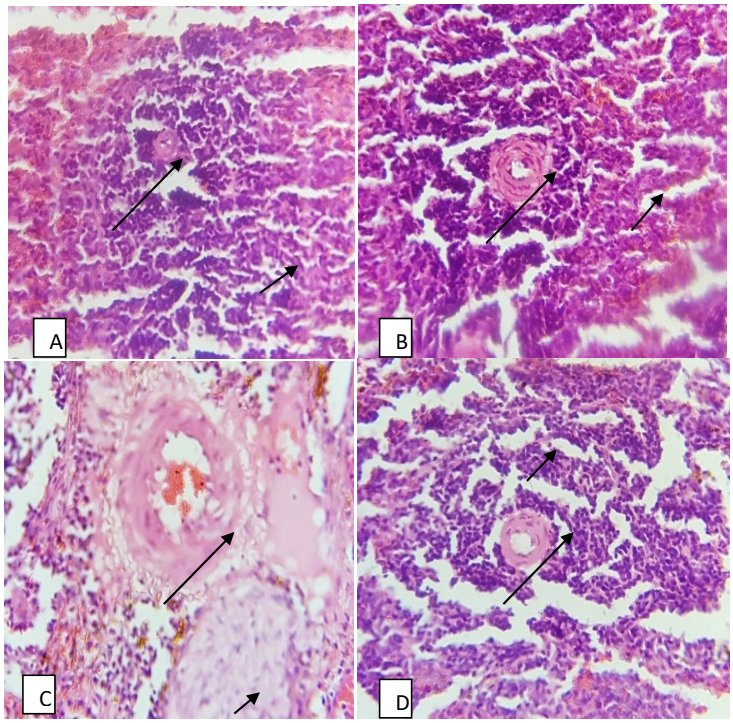

Figure 15. Toxicological effect of Mojeaga herbal remedy on male spleen
A. 150 mg/kg Mojeaga SPLEEN: shows prominent lymphoid follicles with centrally to eccentrically located blood vessels (long arrow). The follicles (white pulp) consist of aggregates of lymphocytes (short arrow). The red pulps are prominent.
B. 300 mg/kg Mojeaga SPLEEN: shows prominent lymphoid follicles with centrally to eccentrically located blood vessels (long arrow). The follicles (white pulp) consist of aggregates of lymphocytes (short arrow). The red pulps are prominent.
C. 600 mg/kg Mojeaga SPLEEN: shows distorted lymphoid follicles with centrally to eccentrically located dilated blood vessels (long arrow). The follicles (white pulp) consist of aggregates of lymphocytes (short arrow). The red pulps appear not so prominent.
D. Control SPLEEN: shows prominent lymphoid follicles with centrally to eccentrically located slightly thickened blood vessels (long arrow). The follicles (white pulp) consist of aggregates of lymphocytes (short arrow). The red pulps are prominent.

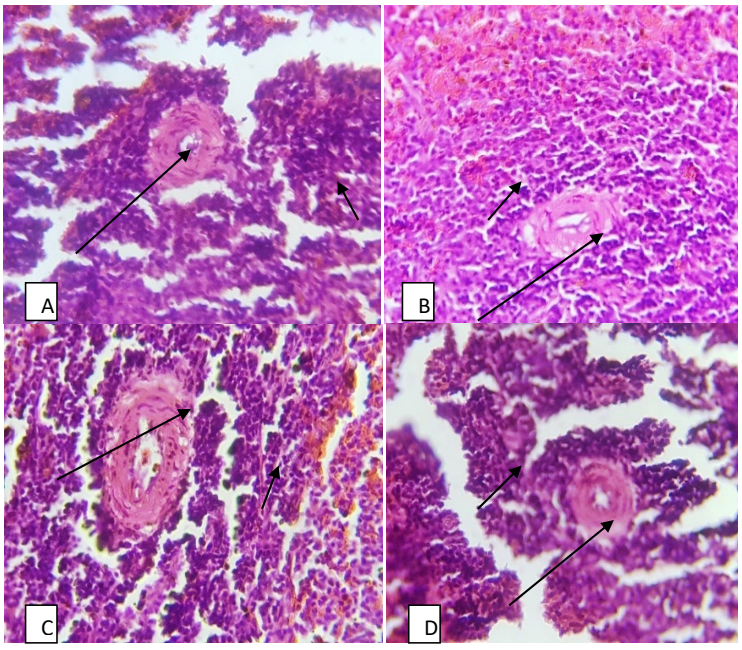

Figure 16. Toxicological effect of Mojeaga herbal remedy on female spleen
A. 150 mg/kg Mojeaga SPLEEN: shows prominent lymphoid follicles with centrally to eccentrically located blood vessels (long arrow). The follicles (white pulp) consist of aggregates of lymphocytes (short arrow). The red pulps are prominent.
B. 300 mg/kg Mojeaga SPLEEN: shows prominent lymphoid follicles with centrally to eccentrically located blood vessels (long arrow). The follicles (white pulp) consist of aggregates of lymphocytes (short arrow). The red pulps are prominent.
C. 600 mg/kg Mojeaga SPLEEN: shows prominent lymphoid follicles with centrally to eccentrically located slightly thickened blood vessels (long arrow). The follicles (white pulp) consist of aggregates of lymphocytes (short arrow). The red pulps are prominent.
D. Control SPLEEN: shows prominent lymphoid follicles with centrally to eccentrically located slightly thickened blood vessels (long arrow). The follicles (white pulp) consist of aggregates of lymphocytes (short arrow). The red pulps are prominent.

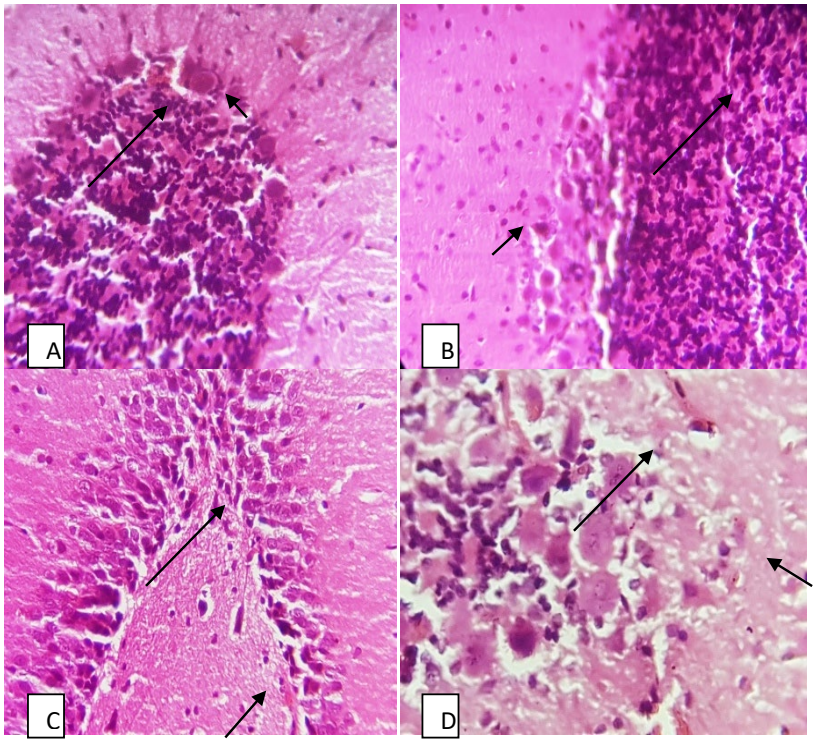

Figure 17. Toxicological effect of Mojeaga herbal remedy on male brain cells
A. 150 mg/kg Mojeaga BRAIN: reveals granule layer (long arrow) bound by prominent Purkinje cell layer. Molecular layer is also seen (short arrow).
B. 300 mg/kg Mojeaga BRAIN: reveals granule layer (long arrow) bound by prominent Purkinje cell layer. Molecular layer is also seen (short arrow).
C. 600 mg/kg Mojeaga BRAIN: reveals granule layer (long arrow) bound by prominent Purkinje cell layer. Molecular layer is also seen (short arrow).
D. Control BRAIN: reveals coarse granule layer (long arrow) bound by basophilic Purkinje cell layer. Molecular layer is also seen (short arrow).

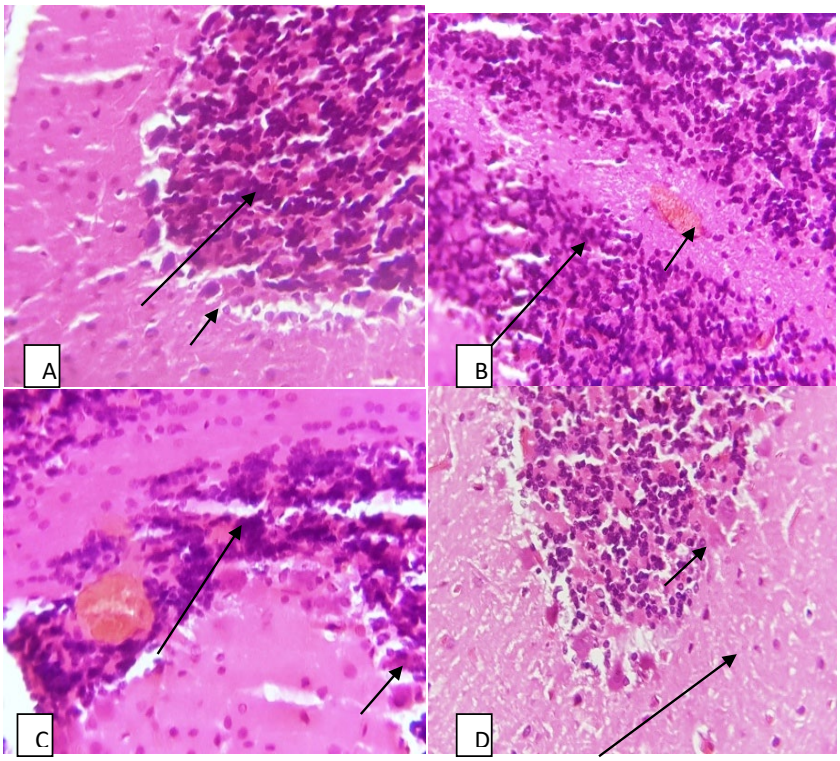

Figure 18. Toxicological effect of Mojeaga herbal remedy on female Brain cells
A. 150 mg/kg Mojeaga BRAIN: reveals granule layer (long arrow) bound by prominent Purkinje cell layer. Molecular layer is also seen (short arrow).
B. 300 mg/kg Mojeaga BRAIN: reveals granule layer (long arrow) bound by prominent Purkinje cell layer. Molecular layer is also seen (short arrow).
C. 600 mg/kg Mojeaga BRAIN: reveals coarse granule layer (long arrow) bound by basophillic Purkinje cell layer. Molecular layer is also seen (short arrow).
D. Control BRAIN: reveals coarse granule layer (long arrow) bound by basophilic Purkinje cell layer. Molecular layer is also seen (short arrow)

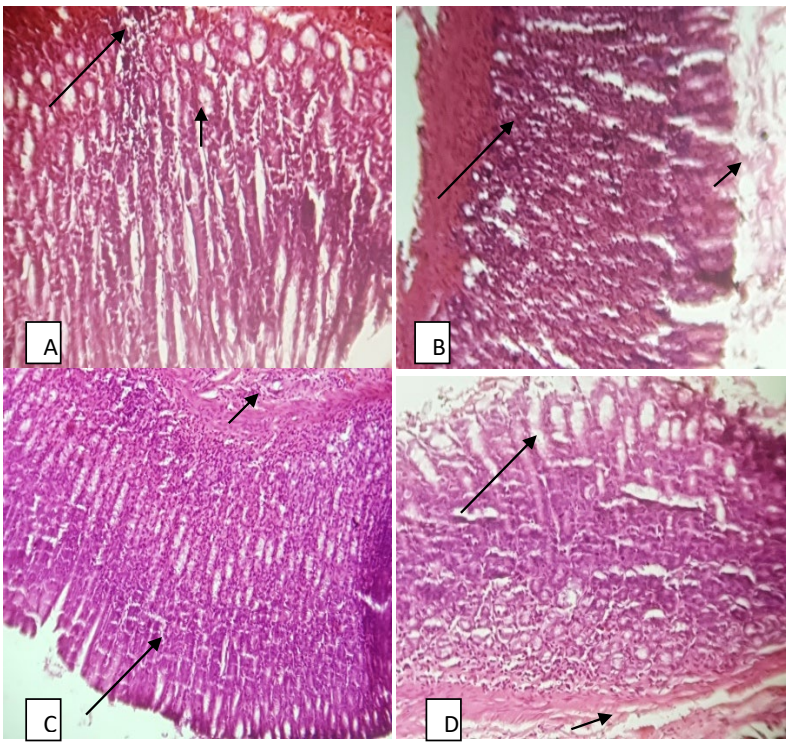

Figure 19. Toxicological effect of Mojeaga herbal remedy on male Stomach
A. 150 mg/kg Mojeaga STOMACH: sections showing histological features, which include the muscularis mucosae (long arrow), sub mucosae (short arrow) at low power magnification with mild neutrophilic and lymphocytic infiltrate (short arrow) infiltrating through the sub mucosa.
B. 300 mg/kg Mojeaga STOMACH: sections showing histological features, which include the muscularis mucosae (long arrow), sub mucosae (short arrow) at l with mild neutrophilic and lymphocytic infiltrates (short arrow) infiltrating through the sub mucosa.
C. 600 mg/kg Mojeaga STOMACH: sections showing histological features, which include the muscularis mucosae (long arrow), sub mucosae (short arrow) with mild neutrophilic and lymphocytic infiltrates (short arrow) infiltrating through the sub mucosa.
D. Control STOMACH: sections showing histological features, which include the muscularis mucosae (long arrow), sub mucosae (short arrow).

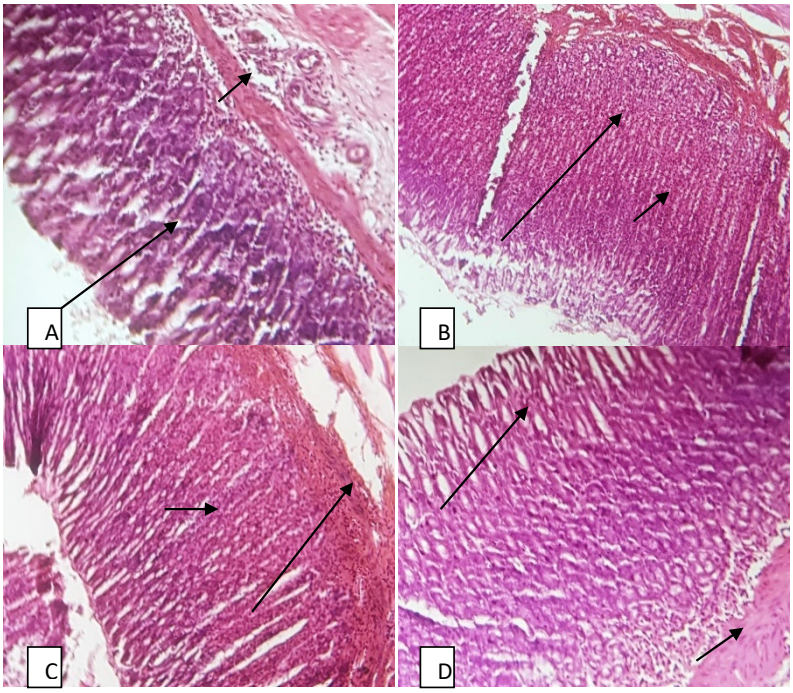

Figure 20. Toxicological effect of Mojeaga herbal remedy on female stomach
A. 150 mg/kg Mojeaga STOMACH: sections showing histological features, which include the muscularis mucosae (long arrow), sub mucosae (short arrow) at low power magnification with mild neutrophilic and lymphocytic infiltrates (short arrow) infiltrating through the sub mucosa.
B. 300 mg/kg Mojeaga STOMACH: sections showing histological features, which include the muscularis mucosae (long arrow), sub mucosae (short arrow) with mild neutrophilic and lymphocytic infiltrates (short arrow) infiltrating through the sub mucosa.
C. 600 mg/kg Mojeaga STOMACH: sections showing histological features, which include the muscularis mucosae (long arrow), sub mucosae (short arrow) with mild neutrophilic and lymphocytic infiltrates (short arrow) infiltrating through the sub mucosa.
D. Control STOMACH: sections showing histological features, which include the muscularis mucosae (long arrow), sub mucosae (short arrow).

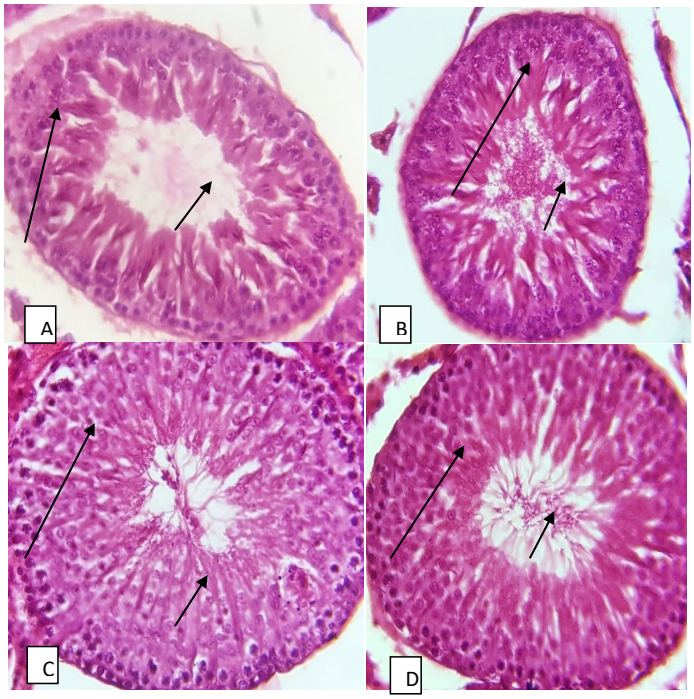

Figure 21. Toxicological effect of Mojeaga herbal remedy on male Testes
A. 150 mg/kg Mojeaga TESTIS: reveals fairly circular Seminoferous tubules [long arrow] containing visible spermatogonia, spermatids and sertoli cells [short arrow].
B. 300 mg/kg Mojeaga TESTIS: reveals fairly circular Seminoferous tubules [long arrow] containing visible spermatogonia, spermatids and sertoli cells [short arrow].
C. 600 mg/kg Mojeaga TESTIS: reveals fairly circular Seminoferous tubules [long arrow] containing visible spermatogonia, spermatids and sertoli cells [short arrow].
D. Control TESTIS: reveals fairly circular Seminoferous tubules [long arrow] containing visible spermatogonia, spermatids and sertoli cells [short arrow].

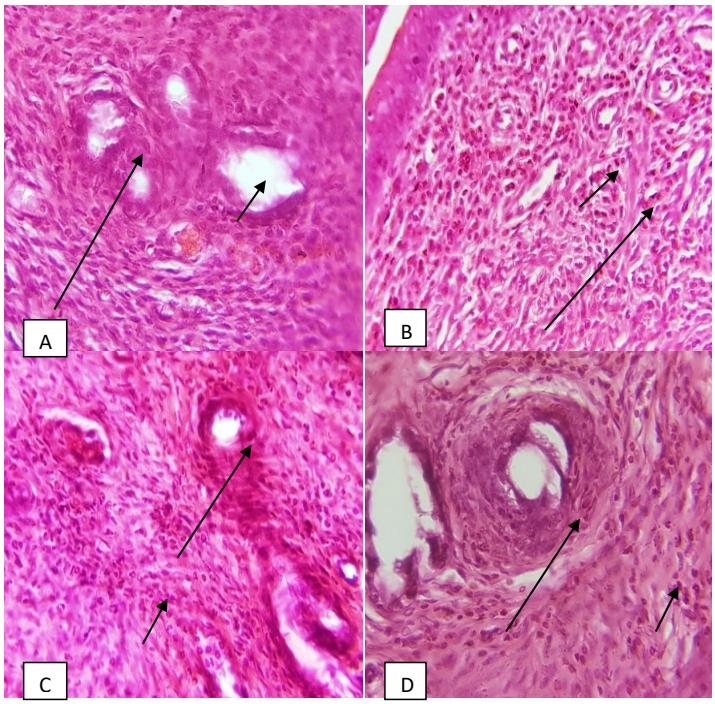

Figure 22. Toxicological effect of Mojeaga herbal remedy on female Uterus
A. 150 mg/kg Mojeaga UTERUS histology reveals not so prominent endometrial glands (long arrow) embedded in the lamina propria (short arrow), there is visible close up of simple columnar epithelium and lamina propria with some areas of haemorrhage.
B. 300 mg/kg Mojeaga UTERUS histology reveals not so prominent endometrial glands (long arrow) embedded in the lamina propria (short arrow), there is visible close up of simple columnar epithelium and lamina propria.
C. 600 mg/kg Mojeaga UTERUS histology reveals not so prominent endometrial glands (long arrow) embedded in the lamina propria (short arrow), there is visible close up of simple columnar epithelium and lamina propria.
D. Control UTERUS histology reveals not so prominent endometrial glands (long arrow) embedded in the lamina propria (short arrow), there is visible close up of simple columnar epithelium and lamina propria

4 Discussion

Since antiquity, traditional medicines and several formulations from herbal plant are known to be safe and effective [39]. Scientific facts about oral toxicity is required not only to help categorize safe doses but to reveal possible clinical signs exhibited by mediators under investigation [40].

Hematological parameters involved blood components that help in determining various degree of toxicity when pre-exposed to toxic substances which could be found in some plant extract. Also, it promote blood functions in plant extracts which possibly can cause degrees of defect [41,42] Normal blood cells differs in their indexes, which can be distorted by overdose intake of toxic plants [43]. Mojeaga herbal remedy involved graded concentration with no alteration to erythrocytes, white blood cells and its differentials, and platelets across the treated groups of male and female animals when compared with the control, showed fall within normal blood range following 3 months chronic study. Slight increase in the level of WBC count and its differentials, red blood cell count, hemoglobin and haematocrite across the treated male and female Wistar rats when compared with the control, indicating that, it promote and improved haematological indexes as shown in table 2 and 3. Thus, this results is in line with Omodamiro and Nwankwo, [44] findings with a marked significant increase in red blood cells total haemoglobin, MCH, PCV and MCV. In regards to the observed hematological parameters, some values were significantly different when compare with the control. It is validated that Mojeaga herbal remedy has the potential to treat blood related diseases [45]. Reductions in WBC, lymphocytes, monocytes, and eosinophils, and platelets counts suggested an Immunoprotective state capable in improving haematinic and immunological effect. Renal function test are biochemical parameters altered by toxin either by triggering over secretion or cause damaged to organs, in which the state of renal function parameters ensured safety. The biosafety properties of this product elicited in this study showed a slight increase ($P < 0.05$) in urea levels across the treated groups in line with standard values when compared with control group in the male and female animals (table 4 and 5). Creatinine levels of mojeaga herbal remedy exhibited slight increased ($P < 0.05$) in graded doses of male and female animals when compared with control. Creatinine and urea concentrations of the product showed nephroprotective property, instituted by urea and creatinine physiological values. This finding agrees with Chukwuma *et al*., [46] reports, indicated that Kidney was implicated in waste removal. Renal function tests ease renal dysfunction, Creatinine and urea are important renal injury signal [47]. Increased in urea levels above normal validated gluconeogenesis uses as substitute source. Increased in Gluconeogenesis is as a result of raised proteolysis that produces glucogenic amino acids undergoing liver deamination of abnormal urea elevation [48]. Effects of Mojeaga herbal remedy proposed renoprotective effect across the treated groups.

Treatment groups of herbal remedy exhibited sustained bicarbonate levels in graded doses. This showed that, it help in preventing a decrease in bicarbonate concentration which could be characterize as kidney diseases under the state of metabolic stress [49]. Hyponatraemia is a defaulted kidney associated diseases. Sodium levels in the control was at normal concentration when compared with treated groups (Mojeaga herbal remedy) with slight significant ($P < 0.05$) increase in sodium level. Symptomatically, sodium level was adjusted with osmotic reduction during diuresis [50]. Potassium levels in treatment groups exhibited significant increase ($P < 0.05$) at 150, 300 and 600 mg/kg of Mojeaga herbal remedy when compared with control among male and female animals, typified renal related hyperkalaemia. Hyperkalaemia with significant values of renal dysfunction is linked with patients prone to hyporeninemic or hypoaldosteronism in nephropathy state [50]. Hyperchloraemia is a feature in control of renal related diseases acting as a pointer to improved renal defects. Chloride ion showed significant increase in Mojeaga herbal remedy at higher dose when compared with control group as shown in table 4 & 5. Frequently, it is confirmed that, the product showed renoprotective effect, this is in line with Chukwuma *et al*., [46]; Osigwe *et al.,* [51] findings that exhibited renoprotective effects of *Citrus paradisci* juice extract.

Liver enzymes includes; AST, ALP and ALT. Certain enzymes action it a functional position of the liver that promotes biochemical markers to hepatic injure [52]. Hepatotoxic drugs stimulate hepatic cell injure with functional enzymes in serum thereby showing a damaging effects [53]. Table 6 & 7 in this present study, an indication of significant decrease in ALT and ALP suggested inhibitory or inactivation enzyme in the treated groups. Nonetheless, ALP and ALT values are in normal ranges [54]. Serum albumin and protein assay were used as a responsive marker in hepatic statutory role to be produced and metabolized via hepatocytes [55]. Total protein reveal nutritional profile that could be screen and aid in the diagnosis of liver disease, kidney disease and several conditions. Mojeaga herbal remedy show no defect in liver functions with insignificant changes in liver enzymes and proteins (table 6 & 7) concentration. This showed sturdy signals of oral protection of Mojeaga herbal remedy in liver function test at graded doses (150, 300 and 600 mg/kg) with insignificant difference to normal ranges ($p<0.05$) in three (3) months chronic study when compared with control, showed no adverse metabolic effects of the pre-exposed rats of the product.

Abnormal weight loss or gain is an indication of toxicity probably as a result of structural proteins breakdown or muscle wastage [56]. This study showed that Mojeaga herbal remedy at graded doses maintain the level of weight gained in relation to toxicity (8 & 9). This result consent with Murunga *et al.* [57] report on grapefruit component to ameliorates weight loss. Similarly, visceral organs such as, spleen, lungs, heart, liver, stomach, brain, kidney, testes and uterus of male and female animals isolated, showed no damage or weight loss across treated when compared with the control as shown in Table 10 & 11 [58].

Hyperlipidemia involved abnormal increase in blood lipids (bad fat) triggered by certain plant phytochemicals. These lipids are cholesterol, triglycerides, lipoprotein and phospholipids [59]. Cholesterol in humans is a metabolic steroidal hormone pioneering the secretion of aldosterone, cortisol, estrogens and androgens that regulate enormous physiologic roles. High cholesterol concentration above 200 mg/dl is a threat to trigger coronary heart disorders. Hence, it activated upsurge artery signs leading to contraction of blood vessels, stimulating high blood pressure or occlusion of vessel causing heart attack. Mojeaga herbal remedy at graded doses (150, 300 and 600 mg/kg) in maintained the level of Triglyceride (TAG), Total Cholesterol (TC) and low density lipoprotein (LDL), with an increased in HDL concentration when compared with the control (Figure 1 & 2). The regulation of TC, TAG and LDL levels at normal values with increase in HDL suggested ameliorative effect of the product in hyperlipidemia, as similarly reported in Murphy et al. [60]; Nissen et al. [61] work. The activities is as a result of increased inhibition of intestinal absorption of cholesterol, intrusion lipoprotein synthesis, increased hepatic expression of LDL receptors and their protective role, leading to an increased in LDL removal from the blood to augment catabolism and cholesterol degradation in the body [62].

Classical uses of bio-markers such as transaminases to evaluate redox state of organs via antioxidant enzymes activity and measurable macromolecule oxidation [63]. Oxidation triggers cellular membranes damage accessed by thiobarbituric acid reactive substances (TBARS) assay. This assay enumerated malondialdehyde (MDA) as a product of lipoperoxidation (LPO). Mojeaga herbal remedy ceased free radicals over secretion in the systems to maintain normal MDA values in the treated groups when compared with the control (Figure 3 and 4). Mojeaga herbal remedy control oxidation, probably in the presence of natural antioxidant components in plant. These compounds are not involved in scavenging radicals as indirect inactivate transcription factors to regulate expressive genes encoded by antioxidant enzymes [64]. Tissue sulfhydryl groups act against reactive oxygen species (ROS) related to toxic tissues caused by oxidative containing phytochemicals in herbs. The principal NPSH, comprised of about 75 % to 90 % total intracellular NPSH reduced glutathione (GSH) [65]. GSH displays an important role in antioxidant defense due to the present of direct radical-scavenging properties, essential component of glutathione peroxidase (GPx) systems, removal of different hydroperoxides [66]. Toxic plants investigated in chronic study deplete GSH content present in visceral tissues and organs due to pro-oxidant effects at higher doses to overcome free-radical stress, GSH is utilized as first line defense against diseases [63] as shown in this study. Non-enzymatic defenses, showed no significance changes in thiols content (NPSH), due to synergistic antioxidant effect between NPSH and SBSB phytochemicals in visceral tissue. Catalase (CAT) and super oxide dismutase (SOD) antioxidant activities were evaluated across treatment groups. CAT together with SOD and GPx constitute enzymatic defense, catalyzing decomposition of ROS against diseases. Mojeaga herbal remedy at graded doses showed an increase in CAT and SOD activities when compared with control (Figure 3 & 4) [67]. Theoretically, SOD and CAT activity rises as compensatory mechanism to scavenge free-radical stress [68].

It is established that an increased in serum testosterone, aid in the regulation of penile erectile magnitude response which stimulates fertility [69]. In this present study, chronic reproductive toxicological investigation of male and female animals treated with graded doses of 150, 300 and 600 mg/kg of Mojeaga herbal remedy, elicited significant increase in serum testosterone when compared with control. This proposed product has the ability to promote testosterone level either by increasing its production or via metabolic impairment. In regards to Mojeaga herbal remedy intake, increase in luteinizing hormone and follicle stimulating hormone in female animals and partially in male were recorded. Findings with similar studies have been reported to clarify potential effect of female sex hormones with a decrease in luteinizing hormone and follicle stimulating hormone impairment due to higher doses of toxic substances, affecting the production and secretion of this hormones [70]. Hence, this study showed that, luteinizing and follicle stimulating hormone showed a significant increase in the treated groups when compared with control (Figure 6). It suggested that the product synthesize and secretion sex hormones with fertility property. Also, prolong intake of Mojeaga herbal remedy rather elevation plasma estrogen concentration in females [71]. This study showed a significant increase in serum estrogen across graded dose 150, 300 and 600 mg/kg of the treatment group (Figure 6). An increased in progesterone concentration in the treated groups to enhance fertility with no adverse effect on reproductive toxicology evaluated.

Histopathological examination of visceral organs like kidney, liver, heart, spleen, stomach, brain and lungs administered with graded doses of 150, 300 and 600 mg/kg of Mojeaga herbal reveals normal renal cell with its functional unit, normal structure of myocardium, pericardium and endocardium muscles, protective stomach mucosa limning, normal bronchi, normal brain cells functioning, alveolar build up and normal histological structure on the alveolar and bronchi muscles and normal spleen architecture when compared with the control [72] (plate 1 - 16). This suggested that the product possess nephroprotective, hepatoprotective, immunoprotective, cardioprotective effect and respiratory effect, similar to the findings recorded in renal and liver function with significant difference ($p < 0.05$). These observations is in line with antioxidant property evaluated from the product, which concurred with the reported of Fuji *et al.* [74] that showed proanthrocyanidins with strongest protective mediating against oxidative stress in visceral organ.

5 Conclusion

In conclusion, Mojeaga herbal remedy showed absent toxicity across the selected doses understudied in chronic oral toxicity studies. The histopathological studies elicited no significant alteration in the visceral organs understudied

in the rats. Furthermore, data from chronic toxicity studies on Mojeaga product affirmed it safety, thus required further therapeutic study.

References

1. Idu M, Omogbai EKI, Aghimien GE, Amaechina F, Timothy O, Omonigho SE. "Preliminary phytochemistry, antimicrobial properties and acute toxicity of *Stachytarpheta jamaicensis* (L.) Vahl. Leaves." *Trends in Medical Res.* 2007;2(4):193–198.
2. Abdel-lateif KS, Maghrabi IA, Eldeab HA. Natural products give maximum defense against diseases leading to human cells damages. *J Bioprocess Biotech*. 2016;6(293):1-7.
3. Seif HAS, Beni-Suef, Univ. bioactive phytochemicals in pharmacology drug research finding and development*, J. Basic Appl. Sci.* 2016;5(2):134-146.
4. Lahlou M. bioactive phytochemicals in pharmacology drug research finding and development*, Pharmacol and Pharm*. 2013;4:17-31.
5. Dhawale PG, Ghyare BP. Bioactive phytochemicals in pharmacology drug research. *J. Nat. Sci. Res*. 2016;6(3):30-35.
6. Orisakwe OE, Afonne OJ, Chude MA, Obi E, Dioka CE. Subchronic toxicity studies of the aqueous extract of Boerhavia diffusa leaves. *J Health Sci*. 2003;49:444-447.
7. Saidu Y, Bilbis LS, Lawal M, Isezuo SA, Hassan SW, Abbas AY. Acute and subchronic toxicity studies of crude aqueous extract of *Albizzia chevalieri*. *Asian J Biochem*. 2007;2: 224-236.
8. Oluwole OB, Elemo GN, Kosoko SB. Nutritional properties and toxicological assessment of high nutrient biscuit developed from blends of some cereals and legume. *J Nutr Disorders Ther*. 2015;5:4.
9. Saidu Y, Nwachukwu FC, Bilbis LS, Farukand UZ, Abbas AY. Toxicity studies of the crude aqueous root extract of Albizzia chevalieri Harms in Albino rats. *Nigerian J Basic Appl Sci*. 2010;18:308-314.
10. Gowda S, Desai PB, Kulkarni SS, Hull VV, Math AA, Vernekar SN. Markers of renal function tests. *N Am J Med Sci*. 2010;2:170-173.
11. Effendy W, Siti-Nurtahirah J, Hussin Z. The side effects of Kacip fatimah extract on liver and kidney of white rats. *J Sustainabil Sci and Manag.* 2006;1:40-46.
12. Thapa BR, Walia A. Liver function tests and their interpretation. *Indian J Pediatr*. 2007;74:663-671.
13. Kwabena J. Combining natural reaction analogues and first derivative spectrophotometer method to enhance the visible spectra of a non-polar crude leaf extract, *Am J Exp Agric*. 2012;2:207-218.
14. Adeshina G, Kunle O, Onaolapo J, Ehinmidu J, Odama L.Phytochemical and Antibacterial Studies of the Hexane Extract of *Alchornea cordifolia* Leaf, Phytochemicals as Nutraceuticals-Global Approaches to Their Role in Nutrition and Health. 2012;65:89-96.
15. Agyare C, Ansah A, Ossei P, Apenteng J, Boakye Y. Wound healing and Anti-infective Properties of *Myrianthus arboreus* and *Alchornea cordifolia*. *Med. Chem*. 2014;4:533-539.
16. Philip CNA, Elijah IO, Amos EK. Performance Evaluation of New Zealand White Rabbits Fed *Alchornea cordifolia* Leaf Meal as Replacement for Soya Bean Meal. *Am J Agric Fores*. 2014;2:51-54.
17. Ademiluyi AO, Oboh G, Agbebi OJ, Boligon AA, Athayde ML. Sorghum [*Sorghum bicolor* (L.) Moench] leaf sheath dye protects against cisplatin-induced hepatotoxicity and oxidative stress in rats. *J Med Food*. 2014;17(12),1332–1338.
18. Ali KS, Elsheikh AM, Ahmed HM, Hassan HA, Hamza NB, Osman MG, Daffalla HM. Phytochemical screening and antibacterial activities of *Sorghum bicolor* leaves derived from *in vitro* culture. *GSC Biol Pharmaceut Sci.* 2020;10(1);65-72.
19. Annor GA, Marcone M, Corredig M, Bertoft E, Seetharaman K. Effects of the amount and type of fatty acids present in millets on their in vitro starch digestibility and expected glycemic index (eGI). *J Cereal Sci*. 2015;64:76–81.
20. Awika JM, Rooney LW. Sorghum Phytochemicals and their potential impact on human health. Phytochem. 2004;65(9):1199-1221.
21. Beta T, Rooney LW, Marovatsanga LT, Taylor JRN. Effect of chemical treatments on polyphenols and malt quality in sorghum. J Cereal Sci. 2003;31(3):295-302. doi:10.1006/jcrs.2000.0310.
22. Andrews DJ, Kumar KA. *Pennisetum glaucum* (L.) R.Br. In: Brink, M. & Belay, G. (Editors). PROTA (Plant Resources of Tropical Africa / Ressources végétales de l'Afrique tropicale), Wageningen, Netherlands. 2006.
23. Burton GW, Wallace AT, Rachie KO. Chemical composition and nutritive value of pearl millet (Pennisetum typhoides) grain. *Crop Sci.* 1972;12:187–188.
24. Hannaway DB, Larson C. Forage fact sheet: pearl millet (*Pennisetum americanum*). Oregon State University, Corvallis, OR. 2004; http://forages.oregonstate.edu/php/fact_sheet_print_grass.php ?SpecID=34&use=Forage (accessed 30 Jul. 2014).
25. Lorke D. A new approach to practical acute toxicity testing. *Arch Toxicol.* 1983;54(1);275-287.
26. Organization for Economic Cooperation and Development. Acute oral toxicity – up and down procedure In: OECD Guidelines for the testing of chemicals No. 425. OECD, Paris, France. 2008a.
27. Organization for Economic Cooperation and Development. Repeated dose oral toxicity test method. In: OECD Guidelines for testing of chemicals, No. 407. OECD, Paris, France. 2008b.
28. Van Skye DD, Neil FM. The determination of gases in blood and other solutions. *J Biol Chem.* 1924;6(6):523.
29. Schales O, Schales SS. A simple and accurate method for the determination of chloride in biological fluids. *J Biol Chem.* 1941;140(5):879-882.
30. Magoshes, Vallee. Flame photometry and spectrophotometry and spectrometry. *J Intern Sci.* 1956;2(1):13-16.
31. Reitman S, Frankel S. A colorimetric method for the determination of serum glutamine oxaloacetic acid and glutamic pyruvic acid transaminase. *J Clin Pathol.* 1957;28:5-12.
32. Bassey OA, Lowry OH, Brock MJ. A Method for the Rapid Determination of Alkaline Phosphates with Five Cubic Millimetres of Serum. *J Biol Chem.*1946;164321-325.
33. Wright PJ, Leatherwood PD, Plummer DT. Enzymes in Rats: Alkaline Phosphatase. *Enzymologia*. 1972;42:317-327.
34. Doumas BT, Perry BW, Sasse EA, Straumfjord JV. Standardization in bilirubin assays: evaluation of selected methods and stability of bilirubin solutions. *J Clin Chem.* 1973;19:984-993.
35. Lowry OH, Rosehmugh NJ, Farr AL, Ran-dall KJ. Protein measurement with the Folin phenol reagent. J Biol Chem. 1951;193:265-275.
36. Trinder P. Determination of glucose in blood using glucose oxidase with an alternative oxygen receptor. *Annals Clin Biochem.* 1969;6:24-27.
37. Tietz NW. Clinical Guide to Laboratory Test. Philadelphia: WB Sanders Company. 3rd edition. 1995;pp.518-519.
38. Friedwald WT, Levy RI, Friedrickson DS. Estimation of the concentration of low density lipoprotein cholesterol in plasma without the use of preparative centrifuge. *Clin chem.* 1972;18:499-502.
39. Schulz V, Hänsel R, Tyler VE. *Rational Phytotherapy. A Physician's Guide to Herbal Medicine*, 4th Ed., Berlin, Springer-Verlag 2001.

40. Pratap B, Singh A. Piscicidal and anti-AChE activity of medicinal plant. *World J Fish Marine Sci.* 2013;5:367-372.
41. Okonkwo JE, Iyadi KC, Effiong CO. Effect of chronic administration of haematological parameters of rats. *Nig J Physiol Sci*. 2004;19(1–2):10–13.
42. Yakubu MT, Akanji MA, Oladiji AT. Hematological evaluation in male albino rats following chronic administration of aqueous extract of *Fadogia agretis* stem. 2007;34(38):0973-1296.
43. Adebayo JO, Adesokan AA, Olatunji LA, Buoro DO, Soladoye AO. Effect of ethanol extract of *Bougainvillea spectabilis* leaves on haematological and serum lipid variables in rats. *Biochem.* 2005;17:45-70.
44. Omodamiro OD, Nwankwo CI. The effect of *Voacanga africana* leaves extract on serum lipid profile and haematological parameters on albino wistar rats. *European J Exp Biol.* 2013;3(3):140-148.
45. Amna OF, Nooraain H, Noriham A, Azizah A, Husna RN. Acute and oral subacute toxicity study of ethanolic extract of Cosmos caudatus leaf in Sprague Dawley rats. *Int J Biosci. Biochem Bioinform.* 2013;3:301–305.
46. Chukwuma OO, Chidozie EA, Ikenna IK, Augusta NC, Jeremiah OS. Anti-diabetic and renal protective effect of the fruit juice of *Citrus X Paradisi* on alloxan induced diabetic male albino wistar rats. *Der Pharmacia Lettre.* 2016;8(19):32-38.
47. Alarcon C, Wicksteed B, Prentki M, Corkey BE, and Rhodes CJ. Succinate Is a Preferential Metabolic Stimulus-Coupling Signal for Glucose-Induced Proinsulin Biosynthesis Translation. *Diabetes*. 2002;51:2496-2504.
48. Abdulazeez MA, Ibrahim K, Bulus K, Babvoshia HB, Abdullahi Y. Effect of Combined Use of *Ocimum gratissimum* and *Vernonia amygdalina* Extract on the Activity of Angiotensin Converting Enzyme, Hypolipidemic and Antioxidant Parameters in Streptozocin-Induced Diabetic Rats. *African J Biochem Res.*2013; **7**:165-173.
49. Wild S, Roglic G, Green ASR, King H. Global Prevalence of Diabetes Estimates for the year 2000 and projections for 2030. *Diabetes Care.* 2004;27(5):1047–1053. PMID: 15111519.
50. Liamis G, Liberopoulos E, Barkas F, Elisaf M. Diabetes mellitus and electrolyte disorders. World J Clin Cases. 2014;2(10):488-496.
51. Osigwe CC. Akah PA, Nworu CS. Biochemical and Haematological Effects of the Leaf Extract of Newbouldia laevis in Alloxan- Induced Diabetic Rats. J Biosci Med. 2017;5:18-36.
52. Moss DW, Ralph HA. Clinical Enzymology. In: Burtis, C.A., Ashword, E.R., (Eds.), Tietz Text book of clinical chemistry, WB Saunders Company, 3rd edition, Philadelphia. 1999;pp 651-683.
53. Kumar AS, Mazumder A, Saravanan VS. Antihyperlipidemic activity of *Camellia sinensis* leaves in Triton WR-1339 induced albino rats. *Phcog Mag.* 2008;4:60-64.
54. Igbe I, Eze GI, Ojameruaye O. Sub-acute toxicity of aqueous fruit pulp extract of Hunteria umbellate. *Niger J Physiol Sci.* 2013;28(1):77-82.
55. Ganong WF. Review of Medical Physiology. 20th ed. New York: Lange Medical Books, McGraw Hill Companies Inc. 2001;pp.500–515.
56. Oyedemi SO, Adewusi EA, Aiyegoro OA, Akinpeanolu DA. Antidiabetic and Hematological Effect of Aqueous Extract of Stem Bark of Afzelia africana (Smith) on Streptozocin-Induced Diabetic Wister Rats. *Asian Pacific J Trop Biomed.* 2011(1):353-358.
57. Murunga AN, Miruka DO, Driver C, Nkomo FS, Cobongela,SZZ, Owira PMO. Grapefruit Derived Flavonoid Naringin Improves Ketoacidosis and Lipid Peroxidation in Type 1 Diabetes Rat Model. *PLoS ONE*, 2016.11(4):e0153241.
58. Witthawaskul P, Ampai P, Kanjanapothi D, Taesothikul N. Acute and Sub-acute toxicities of the Saponin Mixture isolated from Schejjlera leucantha Viguier. *J Ethnopharmacol.* 2003;89:115-121.
59. Iyer D, Patil UK. Effect of *Evolvulus Alsinoides* L. ethanolic extract and its fraction in experimentally induced hyperlipidemia in rats. *Pharmacol online,* 2011;1:573-80.
60. Murphy SA, Cannon CP, Wiviott SD. Effect of intensive lipid lowering therapy on mortality after acute coronary syndrome (a patient– level analysis of the Aggrastat to Zocor and Pravastatin or Atorvastatin Evaluation and Infection Therapy–Thrombolysis in Myocardial Infarction (22 trials). Am J Cardiol. 2007;100(7):1047–1051.
61. Nissen SE, Tuzcu EM, Schoenhagen P. Effect of intensive compared with moderate lipid lowering therapy on progression of coronary atherosclerosis. JAMA. 2011;291(9):1071–1080.
62. Brown MS, Goldstein JL. Lowering plasma cholesterol by raising LDL receptors. *N Eng J Med*. 1981;305:515p.
63. Santosh N, Mohan K, Royana S, Yamini TB. Hepatotoxicity of tuber of Indian Kudzu (*Pueraria tuberosa*) in rats. *Food Chem Toxicol*. 2010;40:1066–1071.
64. Barcelos GR, Angeli JP, Serpeloni JM, Grotto D, Rocha BA, Bastos JK, Knasmüller S, Júnior FB. Quercetin protects human-derived liver cells against mercury-induced DNA-damage and alterations of the redox status. *Mutat Res.* 2011;726:109–115.
65. Moran LK, Gutteridge JMC. Quinlan, G.J. Thiols in cellular redox signaling and control. Curr Med Chem. 2001;8:763–772.
66. Dickinson DA, Forman HJ. Cellular glutathione and thiols metabolism. *Biochem Pharmacol.* 2002;64:1019–1026.
67. Halliwell B, Gutteridge JC. Free Radicals in Biology and Medicine; Oxford University Press: New York, NY, USA. 2007.
68. Bhor VM, Raghuram N, Sivakami S. Oxidative damage and altered antioxidant enzyme activities in the small intestine of streptozotocin-induced diabetic rats. *Int J Biochem Cell Biol.* 2004;36:89–97.
69. Guiliano F, Rampin O, Schirar A, Jardin A, Rousseau JP. Autonomic control of penile erection: modulation by testosterone in the rat. J Neuroendocrinol. 1993;5(6):677-683.
70. Gordon CG, Ahman K, Southern AL, Rubin E, Lieber CS. Effect of alcohol administration on sex hormone metabolism in normal men. New England J Med.1976;295:793-797.
71. Muthusami KR, Chinnaswamy P. Effect of Chronic Alcoholism on male fertility hormones and semen quality. Fertil Steril. 2005;84:919–924.
72. Eleazu CO, Iroaganachi M, Eleazu KC. Ameliorative Potentials of Cocoyam (*Colocasia esculenta* L.) and Unripe Plantain (*Musa paradisiaca* L.) on the Relative Tissue Weights of Streptozotocin-Induced Diabetic Rats. *J Diabetes Res*. 2013;(160964):8.
73. Fuji H, Yokozawa T, Kim YA, Tohga C, Nonaka G. *Mini Reviews in Medicinal Chemistry.* 2006;7:663-6