Original Research Article

Ameliorative Effect of Aqueous Crude Extract of the Aerial parts of *Leonurus cardiaca* on Doxorubicin-Induced Cardiovascular Damage in Wistar Rats

ABSTRACT

This study investigated the ameliorative effect of aqueous crude extract of the aerial parts of *Leonurus cardiaca* on doxorubicin-induced cardiovascular damage in Wistar rats. Aqueous crude extract of *Leonurus cardiaca* was prepared based on standard Laboratory condition. A total of 55 male and female rats weighing between 170 ± 15 and 180±15g were used for this study. The rats were grouped into 11 groups of five rats per group. Group 1 and 2 served as normal and negative control rats respectively. Rats in group 3-11 were intraperitoneally administered 18mg/kg of doxorubicin once and were treated with the crude extract at 166, 250, and 500mg/kg for 7, 14, and 21 days. All biochemical, hematological, and histological analyses were carried out based on standard methods. The mean plasma troponin T and I, IL-6 and C-reactive protein concentrations of the negative control were 357.93±0.01 pg/mL, 241.51±0.00 pg/mL, 63.94±0.01 pg/dl, and 41.03±0.01 mg/ml respectively. The plasma troponin T and I, IL-6 and C-reactive protein concentrations treated for 21 days were 270.32±0.01 pg/mL, 217.84±0.01 pg/mL, 42.75±0.01 pg/dl, and 18.03±0.01 mg/ml respectively. The mean MDA, GSH concentrations, GPx, CAT and SOD activities in doxorubicin-induced cardiovascular damage rats, treated with the extract at 500mg/kg were 5.01±0.01mmol/l, 68.46±0.02µg/mg.protein, 70.21±0.01mg/pro.min, 141.62±1.80IU/g, and 19.05±0.02mg/g respectively and were significantly different from those of group 2. Aqueous crude extract of *Leonurus cardiaca* at 500mg/kg ameliorated the damage induced by doxorubicin on the heart tissues, hence could serve as a novel herbal agent for the treatment of cardiovascular damage.

Keywords: Cardiovascular damage, doxorubicin, heart tissue, *Leonurus cardiaca*, oxidative stress, and Wistar rats.

1. INTRODUCTION

"Anthracyclines are widely adopted antineoplastic regimen, either alone or alongside other cytotoxic agents" [1]. "Among the anthracyclines, doxorubicin and epirubicin are the widely applied agents in the treatment of cancers of different types and childhood solid tumours, soft tissue sarcomas, and aggressive lymphoblastic or myeloblastic leukemia" [1].

“Doxorubicin is reported to elicit ventricular systolic function or heart failure in 7–26% of the patients (0.9–11.4%)” [2]. “Doxorubicin-induced cardiotoxicity may be manifested as arrhythmias, ischaemia, systolic dysfunction, and heart failure, and the major reasons for these changes are cardiac cell death and necrosis” [3].

“Doxorubicin elicit cardiotoxicity through several mechanism. These mechanisms include: oxidative stress induced by reactive oxygen species (ROS), topoisomerase II inhibition, and deoxyribonucleic acid (DNA) double strand break leading to transcriptional alteration of the genes and apoptosis, activation of apoptotic pathway by the impairment of mitochondrial function, and intracellular calcium dysregulation” [4, 5]. Amidst all these mechanisms, oxidative stress is the most common cause of doxorubicin-induced cardiotoxicity [6], which mainly precipitated by imbalance between the production of reactive oxygen species and the intrinsic antioxidant mechanism in the cardiomyocytes. “The mitochondrion participate in doxorubicin-induced cardiotoxicity as anionic cardiolipin present in the inner mitochondrial membrane has high affinity to bind with cationic drug doxorubicin” [4].

Leonurus cardiaca plant has been reported engender dramatic pharmaceutical values [7]. “The medicinal plant commonly identified as motherwort, which is a member of Lamiaceae family usually consumed in Asian countries as a traditional remedy against nervous and functional cardiac disorders” [8, 9]. “Phytochemical and essential oil characterization of the aerial parts of *Leonurus cardiaca* revealed the presence of seven different terpenoids whose total concentration were 26.19×10^{-1} mg/100 g, nine different phenolic acids (506.33 mg/100 g), twelve different saponin (62.33 mg/100 g), seven different cyanogenic glycosides (118.03 mg/100 g), thirteen different glycosides (16.17 mg/100 g), five (5) different anthocyanins (56.53 mg/100 g), twenty six different alkaloids (1.31 mg/100 g), six different flavonoids (7.31 mg/100 g), seven different

sterol (5.91 mg/100 g), tannins (426.49 mg/100 g), and phytate (69.12 mg/100 g) with fourty one different essential oils (100. 00 %) as reported" by Wellington *et al.* [10]. "Acute, sub-acute and chronic toxicity evaluation of aqueous extract of the aerial parts of *Leonurus cardiaca* in normal non-pregnant female Wistar albino rats per OECD 425 TG indicated that aqueous extract of the aerial parts of *Leonurus cardiaca* at 166, 250, and 500mg/kg engendered non-toxic effect on the haematological, liver, kidney, and heart biomarkers as well as on the liver, heart, and kidney architecture" [11]. Proximate, mineral, vitamin and amino acid analysis of *Leonurus cardiaca* indicated that mean and standard deviation of the moisture content was 16.51±0.12%, crude protein (22.00±0.11%), crude fat (12.61±0. 12%), crude fibre (9.11±0. 06%), ash content (4.12±0. 06%), carbohydrate (35.65±.06%), vitamin A concentration was 22.66±0.02mg/100 g, vitamin E (16.07±0.01 mg/100g), Vitamin C (51.22±0.02 mg/100g), vitamin A (16.07±0.01 mg/100g), niacin (B3) (32.47±0.01 mg/100g), thiamine (B1) (15.53±0.02 mg/100g), vitamin B5 (31.91±0.02 mg/100g), sodium concentration was 38.68±0.31 mg/100g, potassium (598.10±0.04 mg/100g), magnesium (496.74±0.19 mg/100g), calcium (263.89±0.03 mg/100g), copper (1.32±0.11 mg/100g), iron (8.62±0.16 mg/100g), zinc (4.41±0.27 mg/100g), manganese (2.12±0.41 mg/100g), selenium (1.83±1.06 mg/100g), and phosphorus (104.09±3.02 mg/100g) [12]. Studies on the effect of aqueous extract of the aerial parts of *Leonurus cardiaca* on cisplatin-induced hepato-renal damage in Wistar albino rats revealed that *Leonurus cardiaca* crude extract at 166, 250, and 500mg/kg evoked hepato-renal curative potential after treatment for 7, 14, and 21 days as reported by Wellington *et al*. [13]. As the major mechanism of doxorubicin-induced cardiovascular damage is also the oxidative stress, the objective of this study was to investigate ameliorative effect of aqueous crude extract of the aerial parts of *Leonurus cardiaca* on doxorubicin-induced cardiovascular damage in Wistar rats

2. MATERIALS AND METHODS

2.1 Preparation of Plant Extract

The aerial parts of *Leonurus cardiaca* were harvested from Idema Community in Abureni Clan of Ogbia Local Government Area, Bayelsa State. Botanical identity was determined by the Department of Plant Science and Biotechnology (PSB), University of Africa Toru-Orua. The sample was registered with Voucher Number UAT/P/211 and sample was deposited at the Herbarium.

Plant extracts were prepared according to a method that was optimized and modified previously in the Biochemistry laboratory University of Africa Toru-Orua. The aerial parts of *Leonurus cardiaca* were dried at 40°C until a constant weight was reached and coarsely grounded. Grounded plant material (24.00 g) was dissolved in 500mL of distilled water and refluxed for 4 hrs. The mixture was strained and the final volume was adjusted to 350 mL and freeze-dried.

2.2 Experimental Animals and Design

Total of 55 healthy male and female Wistar rats, weighing between 170 ± 15 and 180±15g, were purchased from the animal house of the Biochemistry Research Institute, University of Port Harcourt, to carry out the experiment. The rats were housed in a well-ventilated animal house at the Faculty of Basic and Applied Sciences, University of Africa Toru-Orua. The rats were maintained on a standard laboratory diet of rat pellets and water *ad libitum*. Rats were acclimatized to the environment (temperature, 23 ± 2°C; relative humidity, 50 ± 5%; and 12-hour light-dark cycle) for one week prior to experimental use. All protocols used in this study were

approved by the Ethical Review Committee of the Faculty of Medicine, University of Africa Toru-Orua, guided by the CIOMS international guiding principles of biomedical research involving animals. The rats were grouped into eleven groups of five rats per group treated as follows:

Group 1: Received rat feed + H_2O only, serving as normal control

Group 2: Received rat feed + 18mg/kg Dox + H_2O only, serving as negative control

Group 3: Received rat+ 18mg/kg Dox.+ 166mg/kg + H_2O for 7 days

Group 4: Received rat+ 18mg/kg Dox.+ 166mg/kg + H_2O for 14 days

Group 5: Received rat+ 18mg/kg Dox.+ 166mg/kg+ H_2O for 21 days

Group 6: Received rat+ 18mg/kg Dox.+ 250mg/kg + H_2O for 7 days

Group 7: Received rat+ 18mg/kg Dox.+ 250mg/kg + H_2O for 14 days

Group 8: Received rat+ 18mg/kg Dox.+ 250mg/kg + H_2O for 21 days

Group 9: Received rat+ 18mg/kg Dox.+ 500mg/kg + H_2O for 7 days

Group 10: Received rat+ 18mg/kg Dox.+ 500mg/kg + H_2O for 14 days

Group 11: Received rat+ 18mg/kg Dox.+ 500mg/kg + H_2O for 21 days

Note: Dox= Doxorubicin, which was administered intraperitoneally once while the crude extract at 166, 250, and 500mg/kg was orally administered. Rats were sacrificed at seven days interval. Blood sample was collected for biochemical analyses (cardiac troponin I, T, IL-6, C-reactive protein, LDH, AST, ALT, ALP, creatine kinase, MDA, CAT, SOD, GPx, lipid profile, and

haematological parameters) while heart tissue was harvested for homogenate and histological examination.

2.3 Assessment of Blood Parameters

The collected blood was centrifuged and plasma was separated. C-reactive protein, cTnI and cTnT levels were estimated using commercially available enzyme-linked immunosorbent assay (ELISA) kits purchased from Elabscience Biotechnology Co., Ltd, China. AST, ALP, and ALP activity were measured by using a commercially available colourimetric enzyme assay kit purchased from Biorex Diagnostic, United Kingdom. Commercially available spectrophotometric enzyme assay kit purchased from Biorex Diagnostic, United Kingdom, was used to measure LDH, IL-6, and CK, activity in the plasma. MDA level was estimated by using commercially available ELISA kit purchased from DRG International Inc., United States of America (USA).

2.4 Assessment of Antioxidant Parameters and Lipid Peroxidation in Homogenate of Heart Tissue

The right halves of the rat hearts were collected into ice-cold PBS buffer (pH 7.4) and they were used to prepare the homogenate. Heart tissue was weighed into a homogenizer tube and ice-cold PBS buffer was added (the ratio of tissue weight to homogenization buffer was 1 : 10) to homogenize the heart tissue. The supernatant was collected into a prechilled fresh microcentrifuge tube to assess antioxidant parameters and lipid peroxidation. The MDA and GSH levels were estimated using commercially available spectrophotometric assay kits purchased from Biorex Diagnostic, United Kingdom. CAT activity was measured using a

commercially available test kit purchased from antibodies-online.com, USA. SOD and GPx activity and lipid peroxidation were estimated using commercially available colourimetric assay kits purchased from Sigma Aldrich, USA.

2.5 Histological Assessment of Cardiovascular Damage

Myocardial tissue was sampled from the left half of the heart of all animals for histological assessment and fixed in 10% formal saline. They were processed, sectioned in 3 μm thickness, and stained with haematoxylin and eosin. The sections were examined under the light microscope and necrotic changes were scored. Scoring of necrotic changes was performed according to a grading system developed by the authors as follows: 0, no cells with necrotic changes; 1, up to 10 cells with necrotic changes; 2, 10–50 cells with necrotic changes; 3, 50–100 cells with necrotic changes; 4, >100 cells with necrotic changes. Myocytes with nuclear pyknosis or karrheorhexis or karyolysis with hypereosinophilic cytoplasm and striation were identified as necrotic myocytes. Necrotic myocyte density was assessed separately in peripheral and subendocardial regions of the myocardium.

2.6 Statistical Analysis

Data were expressed as the mean ± standard deviation (SD). One-way analysis of variance followed by Dunnett's multiple comparisons test was used to analyze the statistical difference between different treatment groups using SPSS 22.0 software. Differences were considered statistically significant at $P \leq 0.05$.

3. RESULTS AND DISCUSSION

Table 1 shows the effect of aqueous extract of the aerial parts of *Leonurus cardiaca* on the plasma cardiac bio-markers on doxorubicin-Induced cardiovascular damage in Wistar albino

rats. The mean plasma troponin T and I, IL-6 and C-reactive protein (CRP) concentrations of normal control were 284.45±0.01pg/mL, 168.32±0.01 pg/mL, 31.02±0.01 pg/dl, and 8.16±0.01 mg/ml respectively (Table 1). The mean plasma troponin T and I, IL-6 and C-reactive protein (CRP) concentrations of the negative control were 357.93±0.01 pg/mL, 241.51±0.00 pg/mL, 63.94±0.01 pg/dl, and 41.03±0.01 mg/ml respectively (Table 1). The mean plasma troponin T and I, IL-6 and C-reactive protein (CRP) concentrations of the doxorubicin-induced cardiovascular damage rats, treated with the aqueous extract at 166 mg/kg b.wt for 7 days were 357.24±0.01 pg/mL, 237.04±0.01 pg/mL, 63.83±0.01 pg/dl, and 39.84±0.01 mg/ml respectively. The mean plasma troponin T and I, IL-6 and C-reactive protein (CRP) concentrations of the doxorubicin-induced cardiovascular damage rats, treated with the aqueous extract at 166 mg/kg for 14 days were 358.44±1.80 pg/mL, 236.87±0.01 pg/mL, 63.64±0.01pg/dl, 39.62±0.00 mg/ml respectively while those treated for 21 days were 356.21±0.00 pg/mL, 234.23±0.01 pg/mL, 63.46±0.01 pg/dl, and 39.47±0.01mg/ml respectively (Table 1). The plasma troponin T and I, IL-6 and C-reactive protein (CRP) concentrations of doxorubicin-induced cardiovascular damage rats, treated with 250 mg/kg were for 7 days were 331.53±0.01 pg/mL, 225.95±0.01 pg/mL, 58.83±0.01 pg/dl, and 32.05±0.01 mg/ml respectively. The plasma troponin T and I, IL-6 and C-reactive protein (CRP) concentrations of doxorubicin-induced cardiovascular damage rats, treated with 250 mg/kg were for 14 days were 331.04±0.02 pg/mL, 225.05±0.02 pg/mL, 58.73±0.01pg/dl, and 31.82±0.01 mg/ml respectively while those treated for 21 days were 329.85±0.01 pg/mL, 254.56±0.00 pg/mL, 58.56±0.01 pg/dl, and 27.94±0.01 mg/ml respectively (Table 1). Meanwhile, the plasma troponin T and I, IL-6 and C-reactive protein (CRP) concentrations of doxorubicin-induced cardiovascular damage rats, treated with 500 mg/kg of the extract for 7 days were 283.13±0.01 pg/mL, 218.44±0.01 pg/mL, 45.05±0.01 pg/dl, and

18.66±0.05 mg/ml respectively. The plasma troponin T and I, IL-6 and C-reactive protein (CRP) concentrations of doxorubicin-induced cardiovascular damage rats, treated with 500 mg/kg b.w of the extract for 14 days were 275.72±0.01 pg/mL, 218.01±0.00 pg/mL, 43.21±0.01 pg/dl, and 18.34±0.01 mg/kg respectively while those treated for 21 days were 270.32±0.01 pg/mL, 217.84±0.01 pg/mL, 42.75±0.01 pg/dl, and 18.03±0.01 mg/ml respectively (Table 1). Concurrently, the mean plasma troponin T and I, IL-6 and C-reactive protein (CRP) concentrations of the negative control were significantly different from the normal control group. The mean plasma troponin T and I, IL-6 and C-reactive protein (CRP) concentrations treated with 166, 250, and 500 mg/kg for seven, 14, and 21 days, in doxorubicin-induced cardiovascular damage rats were significantly different from the negative control.

Plate 1 shows the photomicrograph of heart tissue of normal control Wistar albino rats. The photomicrograph of normal control Wistar albino rats showed normal myocytes and normal heart architecture. Plate 2 shows the photomicrograph of heart tissues of negative control, indicating fatty inflammation of myocytes and disintegration of myocytes with loss of nuclei, intracellular accumulation and apoptosis.

Plate 3 shows the photomicrograph of heart tissues on doxorubicin-induced cardiovascular damage rats, treated with aqueous extract of *Leonurus cardiaca* at 166 mg/kg for 7 days, indicating fatty inflammation of cardiac muscle cells, disintegration of myocytes, intracellular fatty accumulation and loss of cardiac nuclei in comparison to the negative control heart tissue (Plate 2). Plate 4 shows the photomicrograph of heart tissue on heart tissue of doxorubicin-induced cardiovascular damage rats, treated with 166 mg/kg b.w of aqueous extract of the aerial parts of *Leonurus cardiaca* for 14 days, showing enlarged myocytes and nuclei (red arrows), in comparison to the negative control heart tissue (Plate 2). Plate 6 shows photomicrograph of heart

tissue on doxorubicin-induced cardiovascular damage rats, treated with 166 mg/kg b.w of aqueous extract of the aerial parts of *Leonurus cardiaca* for 21 days, showing enlarged myocytes and nuclei (blue arrows), in comparison to the negative control heart tissue (Plate 2). Treatment with the extract at 166 mg/kg reduced fatty inflammation of cardiac muscle cells, mild disintegration of myocytes, and loss of cardiac nuclei in comparison to the negative control heart tissue (Plate 2). The ameliorative effect of aqueous extract of *Leonurus cardiaca* on the mean plasma cardiac biomarkers (CTn I and T) is similar to the claim of Abdel-Daim *et al*. [14] on Allicin ameliorates doxorubicin-induced cardiotoxicity in rats via suppression of oxidative stress, inflammation and apoptosis and Asmaa *et al.* [15] on the ameliorating effect of Lycopene and N-Acetylcysteine against doxorubicin-induced cardiac injury in rats.

Table 2 shows the effect of aqueous extract of the aerial parts of *Leonurus cardiaca* on biomarkers of cardiac homogenate enzymes activities on doxorubicin-induced cardiovascular damage in Wistar albino rats. The mean cardiac homogenate creatine kinase (CK), ALT, ALP, AST and LDH activities of the normal control were 182.66±0.01U/L, 71.42±0.01 U/L, 78.14±0.01 U/L, 171.65±0.01 U/L, and 182.25±0.01 U/L respectively (Table 2). The mean cardiac homogenate creatine kinase (CK), ALT, ALP, AST and LDH activities of the negative control were 240.51±0.01 U/L, 94.78±0.05 U/L, 100.33±0.01 U/L, 200.03±0.01 U/L and 221.67±0.01 U/L respectively (Table 2). The mean cardiac homogenate creatine kinase (CK), ALT, ALP, AST and LDH activities of the doxorubicin-induced cardiovascular damage rats, treated with the aqueous extract at 166 mg/kg for 7 days were 237.04±0.01 U/L, 91.63±0.01 U/L, 97.45±0.01 U/L, 197.22±0.01 U/L, and 204.35±0.01 U/L respectively. The mean cardiac homogenate creatine kinase (CK), ALT, ALP, AST and LDH activities of the doxorubicin-induced cardiovascular damage rats, treated with the aqueous extract at 166 mg/kg for 14 days

were 234.63±0.04 U/L, 86.26±0.01 U/L, 95.74±0.01 U/L, 193.04±0.01 U/L, and 202.85±0.01 respectively, while those treated for 21 days were 227.45±0.01 U/L, 81.05±0.01 U/L, 92.16±0.01 U/L, 187.26±0.01 U/L, and 196.72±0.01 U/L respectively (Table 2). The mean plasma cardiac homogenate creatine kinase (CK), ALT, ALP, AST and LDH activities of the doxorubicin-induced cardiovascular damage rats, treated with the aqueous extract at 250 mg/kg for 7 days were 201.56±0.01 U/L, 74.83±0.01 U/L, 87.25±0.01 U/L, 181.35±0.01 U/L, and 191.25±0.01 U/L respectively (Table 2). The mean plasma cardiac homogenate creatine kinase (CK), ALT, ALP, AST and LDH activities of the doxorubicin-induced cardiovascular damage rats, treated with the aqueous extract at 250 mg/kg for 14 days were 195.64±0.01 U/L, 72.23±0.01 U/L, 83.70±0.02 U/L, 176.83±0.01 U/L, and 186.03±0.01 U/L respectively while those treated for 21 days were 184.54±0.01 U/L, 68.48±0.01 U/L, 78.03±0.01 U/L, 173.01±0.01 U/L, and 182.73±0.01 U/L respectively (Table 2).

More so, the mean creatine kinase (C-kinase), alanine amino transferase (ALT), alkaline phosphatase (ALP), aspartate transaminase (AST) and lactate dehydrogenase (LDH) activities of normal rats orally administered with the extract at 500 mg/kg were 102.85±0.01 U/L, 73.08±0.01 U/L, 45.76±0.01 U/L, 83.03±0.02 U/L, and 97.02±0.02 U/L respectively for 7 days, 97.24±0.01 U/L, 63.66±0.01 U/L, 41.25±0.01 U/L, 83.03±0.02 U/L, and 92.76±0.01 U/L respectively for 14 days, and 94.05±0.03 U/L, 61.76±0.01 U/L, 35.96±0.01 U/L, 74.04±0.01 U/L, and 85.66±0.01 U/L respectively for 21 days while those treated with the positive were 74.40±0.02 U/L, 42.72±0.02 U/L, 31.64±0.01 U/L, 69.05±0.02 U/L, and 68.24±0.01 U/L respectively (Table 2).

Concurrently, administration of 18mg/kg of doxorubicin to the negative control caused significant increases on the cardiac homogenate CK, ALT, ALP, AST, and LDH activities when compared to the normal control (Table 2). The mean cardiac homogenate CK, ALT, ALP, AST,

and LDH activities treated with the aqueous extract at 166 mg/kg for 7, 14, and 21 days on doxorubicin-induced cardiovascular damage rats resulted in significant decreases on the mean cardiac homogenate CK, ALT, ALP, AST, and LDH activities in comparison to the negative control. Meantime, the mean cardiac homogenate CK, ALT, ALP, AST, and LDH activities in doxorubicin-induced cardiovascular damage rats, treated with the extract at 250 and 500 mg/kg for 7, 14, and 21 days, significantly decreased the cardiac homogenate CK, ALT, ALP, AST, and LDH activities when compared to the negative control.

Meanwhile, plate 7 shows the photomicrograph of heart tissue on doxorubicin-induced cardiovascular damage rats, treated with 250 mg/kg of aqueous extract of the aerial parts of *Leonurus cardiaca* for 7days, indicating myocyte and nuclei regeneration, in comparison to the negative control heart tissue (Plate 2). Plate 8 shows the photomicrograph of heart tissue on doxorubicin-induced cardiovascular damage rats, treated with 250 mg/kg of aqueous extract of the aerial parts of *Leonurus cardiaca* for 14 days, showing formation new myocytes and small nuclei, in comparison to the negative control heart tissue (Plate 2). Plate 9 shows the photomicrograph of heart tissue on doxorubicin-induced cardiovascular damage rats, treated with 250 mg/kg of aqueous extract of the aerial parts of *Leonurus cardiaca* for 21 days showing formation new myocytes and small nuclei, in comparison to the negative control heart tissue (Plate 2). Treatment with the extract at 250 mg/kg for 7, 14, and 21 days, caused moderate regeneration of muscle cells, moderate fatty inflammation of cardiac muscle cells, moderate interstitial oedema and moderate cardiac nuclei in comparison to the negative control heart tissue (Plate 2). These findings are supported by the work of Gunes *et al.* (2016) on cardioprotective effect of selenium against cyclophosphamide-induced cardiotoxicity in rats.

Cardiac-specific troponins (cTn) and markers such as troponin T (cTnT), troponin C (cTnC), troponin I (cTnI), c-reative protein and IL-6 are diagnostic biomarkers of myocardial infarction [16]. Both cTnT and cTnI are released into the circulation following cardiac damage and are the preferred cardiac biomarkers when myocardial infarction (MI) is suspected [17]. Elevated cardiac troponin T (cTnT), cardiac troponin I (cTnI), sodium, potassium, chloride electrolytes, urea and bicarbonate in the blood of a patient are primary diagnosis for heart failure, coronary embolism, glomerulosclerosis and chronic kidney damage [18]. In this present research, intraperitoneal administration of 18mg/kg of doxorubicin resulted in significant increases on mean plasma cTnT, cTnI, IL-6 concentrations, cardiac homogenate creatine kinase, ALT, ALP, AST, and LDH activities in the negative control when compared to the normal control (Table 1 and 2). The significant increases on the mean plasma cTnT, cTnI, c-reactive protein and IL-6 level and on the mean cardiac homogenate creatine kinase, ALT, ALP, AST, and LDH activities observed in the negative control in comparison to the normal control (Table 1and 2). However, treatment with aqueous extract of the aerial parts of *Leonurus cardiaca* at, 166, 250 and 500 mg/kg for 7, 14 and 21 days, significantly caused decreases on the plasma cTnT, cTnI, c-reactive protein and IL-6 concentrations as well on cardiac homogenate creatine kinase, ALT, ALP, AST, and LDH activities when compared to the negative and normal control values (Table 1 and 2). The significant decreases on the plasma cTnT, cTnI, c-reactive protein and IL-6 concentrations as well as on the cardiac homogenate creatine kinase, ALT, ALP, AST, and LDH activities are suggestive of the ameliorative potential of the extract against cisplatin-induced cardiovascular damage in Wistar albino rats, which justifies the use of formulation from the leaves of *Leonurus cardiaca* as cardiovascular tonic in Latin as reported by Rauwald *et al*. [19] which is similar to the report of Bekalu *et al.* [20] on the plasma troponin levels rats in their study on

cardioprotective effect of crude extract and solvent fractions of *Urtica simensis* leaves on cyclophosphamide-induced myocardial injury in rats, Jayassinqhe *et al.* [21] on cardioprotective potential of *Murraya koenigii* (L.) Spreng leaf extract against doxorubicin-induced cardiotoxicity in rats as well as the report of Mahammed [22] on Cardioprotective properties of Artemisia herba alba nanoparticles against heart attack in rats: A study of the antioxidant and hypolipidemic activities.

Table 1 Effects of aqueous extract of the aerial parts of *L. cardiaca* on the plasma cardiac bio-marker on doxorubicin-induced cardiovascular damage in Wistar rats

Treatments	CTn- T (pg/Ml	CTn- I (pg/mL)	IL-6 Level (pg/dl)	CRP (mg/ml)
N/Control	284.45±0.01	168.32±0.01	31.02±0.01	8.16±0.01
Ne/Control	357.93±0.01^{a}	241.51±0.00 ^{a}	63.94±0.01 ^{a}	41.03±0.01 ^{a}
166mg/kg+DX7	357.24±0.01^{ab}	237.04±0.01^{b}	63.83±0.01^{ab}	39.84±0.01^{b}
166mg/kg+DX14	358.44±1.80^{ab}	236.87±0.01^{b}	63.64±0.01^{ab}	39.62±0.00^{b}
166mg/kg+DX21	356.21±0.00^{ab}	234.23±0.01^{b}	58.73±0.01^{b}	39.47±0.01^{b}
250 mg/kg+DX 7	329.85±0.01^{bc}	225.95±0.01^{b}	63.46±0.01	32.05±0.01^{b}
250mg/kg+DX14	331.04±0.02^{bc}	225.05±0.02^{b}	58.83±0.01^{b}	31.82±0.01^{b}
250mg/kg+DX21	329.85±0.01^{bc}	254.56±0.00^{b}	58.56±0.01^{b}	27.94±0.01^{b}
500 mg/kg+DX 7	283.13±0.01^{b}	218.44±0.01^{b}	45.05±0.01^{b}	18.66±0.05^{b}
500mg/kg+DX14	275.72±0.01^{b}	218.01±0.00^{b}	43.21±0.01^{b}	18.34±0.01^{b}

500mg/kg+DX21	270.32±0.01[b]	217.84±0.01[b]	42.75±0.01[b]	18.03±0.01[b]

DX= doxorubicin. Values are reported as mean ± standard error of mean (M±SEM) (n =5). Values bearing superscript ("ab") were not significantly different($p \leq 0.05$) from the negative control down the group. Values bearing superscript ("bc") were significantly different ($p \leq 0.05$) from the negative control down the group. Values bearing superscript ("b") were significantly different($p<0.05$) from the negative and normal control down the groups.

Table 2 Effect of aqueous extract of the aerial parts of *Leonurus cardiaca* on Cardiac Homogenate activities on doxorubicin-induced cardiovascular damage in Wistar rats

Treatment	CK (U/L)	ALT (U/L)	ALP (U/L)	AST (U/L)	LDH (U/L)
N/Control	182.66±0.0	71.42±0.01	78.14±0.01	171.65±0.01	182.25±0.01
Ne/Control	240.51±0.01[ab]	94.78±0.05[ab]	100.33±0.0[ab]	200.03±0.0[ab]	221.67±0.01[ab]
166mg/kg+DX7	237.04±0.0[b]	91.63±0.01[b]	97.45±0.01[b]	197.22±0.01[b]	85.66±0.01[b]
166mg/kg+DX14	234.63±0.0[b]	86.26±0.01[b]	95.74±0.01[b]	193.04±0.01[b]	204.35±0.01[b]
166mg/kg+DX21	227.45±0.1[b]	81.05±0.01[b]	92.16±0.01[b]	187.26±0.01[b]	202.85±0.01[b]
250mg/kg+DX7	201.56±0.0[b]	74.83±0.01[b]	87.25±0.01[b]	181.35±0.01[b]	196.72±0.01[a]
250mg/kg+DX14	146.31±0.0[b]	72.23±0.01[b]	83.70±0.02[b]	176.83±0.01[b]	191.25±0.01[b]
250mg/kg+DX21	184.54±0.1[b]	68.48±0.01[b]	78.03±0.01[b]	173.01±0.01[b]	186.03±0.01[b]
500mg/kg+DX7	171.35±0.0[bc]	55.62±0.01[b]	73.22±0.01[bc]	163.73±0.01[bc]	182.73±0.0[b]
500mg/kg+DX14	167.02±0.1[b]	51.83±0.01[b]	70.04±0.01[b]	158.25±0.01[b]	168.04±0.02[b]
500mg/kg+DX21	147.14±0.0[b]	46.94±0.01[b]	64.23±0.01[b]	137.72±0.01[b]	163.25±0.02[b]

DX= doxorubicin.

Values are reported as mean ± standard error of mean (M±SEM) (n =5). Values bearing superscript ("ab") were significantly different ($p \leq 0.05$) from the normal. Values with superscript ("c") were significantly ($p \leq 0.05$) different from the negative control down the groups. Values with superscript ("b") were significantly different from the negative and normal control.

Table 3 shows the effect of aqueous extract of the aerial parts of *Leonurus cardiaca* on the plasma lipid profile of doxorubicin-induced cardiovascular damage in Wistar albino rats. The mean plasma total cholesterol, HDL, LDL, VLDL and triglyceride concentrations of the normal control were 94.22±0.01 mg/dl, 56.34±0.01 mg/dl, 60.21±0.00 mg/dl, 9.32±0.02 mg/dl, 48.13±0.02 mg/dl respectively, those the negative control were 37.13±0.02 mg/dl, 27.76±0.01 mg/dl, 84.26±0.01 mg/dl, 4.27±0.01 mg/dl, and 25.64±0.01 mg/dl respectively (Table 3). The mean plasma total cholesterol, HDL, LDL, VLDL and triglyceride concentrations in doxorubicin-induced cardiovascular damage rats treated with 166 mg/kg were 37.45±0.01 mg/dl, 27.95±0.01 mg/dl, 84.04±0.01 mg/dl, 4.34±0.01 mg/dl, and 25.93±0.01 mg/dl respectively for 7 days, 38.06±0.01 mg/dl, 29.15±0.01 mg/dl, 83.86±0.01 mg/dl, 4.75±0.01 mg/dl, 4.75±0.01 mg/dl and 26.05±0.01 mg/dl respectively for 14 days, and 38.76±0.01 mg/dl, 31.42±0.01 mg/dl, 83.46±0.01 mg/dl, 4.94±0.01 mg/dl, and 26.76±0.02 mg/dl respectively (Table 3). The mean plasma total cholesterol, HDL, LDL, VLDL and triglyceride concentrations in doxorubicin-induced cardiovascular damage rats, treated with 250 mg/kg were 41.05±0.02 mg/dl, 31.64±0.00 mg/dl, 83.22±0.01 mg/dl, 5.06±0.00 mg/dl, and 27.15±0.01 mg/dl respectively for 7 days, 41.05±0.02 mg/dl, 31.94±0.01 mg/dl, 83.02±0.01 mg/dl, 5.23±0.01 mg/dl, and 27.15±0.01 mg/dl respectively for 14 days, and 41.66±0.01 mg/dl, 34.22±0.00 mg/dl, 82.89±0.00 mg/dl, 5.46±0.00 mg/dl, and 27.65±0.01 mg/dl respectively for 21 days (Table 3).

More so, plate 10 shows the shows the photomicrograph of heart tissue on doxorubicin-induced cardiovascular renal damage rats, treated with 500 mg/kg of aqueous extract of the aerial parts of *Leonurus cardiaca* for 7 days, indicating few fatty inflammation of cardiac muscle cells, interstitial oedema and loss of cardiac nuclei, in comparison to the negative control heart tissue (Plate 2). Plate 11 shows the photomicrograph of heart tissue on doxorubicin-induced

cardiovascular damage rats, treated with 500 mg/kg of aqueous extract of the aerial parts of *Leonurus cardiaca* for 14 days, showing almost normal myocytes and nuclei, in comparison to the negative control heart tissue (Plate 2). Plate 12 shows the photomicrograph of heart tissue on doxorubicin-induced cardiovascular damage rats, treated with 500 mg/kg of aqueous extract of the aerial parts of *Leonurus cardiaca* for 21 days, pointing to almost normal myocytes and nuclei, in comparison to the negative control heart tissue (Plate 2). Treatment with the extract at 500 mg/kg b.w for 7, 14, and 21 days, resulted in the regeneration of cardiac muscle cells, appearance of normal myocytes and regeneration of new cardiac nuclei in comparison to negative heart tissue (Plate 2).

Meanwhile, the mean plasma total cholesterol, HDL, LDL, VLDL and triglyceride concentrations in doxorubicin-induced cardiovascular damage rats, treated with 500 mg/kg were 56.14±0.01 mg/dl, 52.45±0.01 mg/dl, 62.02±0.01 mg/dl, 6.24±0.01 mg/dl, and 31.66±0.01 mg/dl respectively for 7 days, 68.34±0.01 mg/dl, 61.03±0.01 mg/dl, 46.24±0.01 mg/dl, 6.89±0.01 mg/dl, and 36.15±0.01 mg/dl respectively for 14 days, and 76.24±0.01 mg/dl, 75.05±0.02 mg/dl, 37.75±0.01 mg/dl, 7.32±0.01 mg/dl, and 43.08±0.02 mg/dl respectively for 21 days (Table 3). The intraperitoneal administration of 18mg/kg of doxorubicin to the negative control caused significant decreases on the plasma total cholesterol, HDL, VLDL and triglyceride and an increased LDL concentrations when compared with the normal control group. Treatment with aqueous extract of *Leonurus cardiaca* at 166, 250, and 500mg/kg to doxorubicin-induced cardiovascular damage rats for 7, 14, and 21 days resulted in significant decreases on the mean plasma LDL-cholesterol of the negative control group. Also, significant increases on the mean plasma , HDL, VLDL and triglyceride levels were observed at 166, 250, and 500mg/kg for 7, 14, and 21 days. More so, a mean more significant effect on the mean plasma total cholesterol, HLD,

VLDL and triglyceride as well as on LDL concentrations occurred with treatment at 500 mg/kg of the extract, followed by 250 mg/kg while the least was observed with 166 mg/kg (Table 3).

More so, plate 10 shows the shows the photomicrograph of heart tissue on doxorubicin-induced cardiovascular renal damage rats, treated with 500 mg/kg of aqueous extract of the aerial parts of *Leonurus cardiaca* for 7 days, indicating few fatty inflammation of cardiac muscle cells, interstitial oedema and loss of cardiac nuclei, in comparison to the negative control heart tissue (Plate 2). Plate 11 shows the photomicrograph of heart tissue on doxorubicin-induced cardiovascular damage rats, treated with 500 mg/kg of aqueous extract of the aerial parts of *Leonurus cardiaca* for 14 days, showing almost normal myocytes and nuclei, in comparison to the negative control heart tissue (Plate 2). Plate 12 shows the photomicrograph of heart tissue on doxorubicin-induced cardiovascular damage rats, treated with 500 mg/kg of aqueous extract of the aerial parts of *Leonurus cardiaca* for 21 days, pointing to almost normal myocytes and nuclei, in comparison to the negative control heart tissue (Plate 2). Treatment with the extract at 500 mg/kg b.w for 7, 14, and 21 days, resulted in the regeneration of cardiac muscle cells, appearance of normal myocytes and regeneration of new cardiac nuclei in comparison to negative heart tissue (Plate 2).

The lipid profile indices are useful in monitoring health status of the cardiovascular system. Although the rat total cholesterol level was not affected by Fijk herbal mixture administration, elevated levels of TAG and LDL-cholesterol may predispose to cardiovascular related disorders [23]. Increased level of LDL-cholesterol has been associated with higher risk of atherosclerosis while elevated level of HDL-C is linked to reduced occurrences of cardiovascular disorder [24]. In this study, the intraperitoneal administration of 18mg/kg of doxorubicin (negative control) caused significant decreases on the mean plasma total cholesterol, HDL-cholesterol VLDL-

cholesterol and triglyceride and an increased LDL-cholesterol concentrations when compared with the positive control group (Table 3). The lipid profile of the normal and negative control are similar to those of Ejoba *et al*. [25] on Effect of *Saccharum barberi* extract on lipid profile level in albino Wistar rats. The significant decreases observed on the mean plasma total cholesterol, HDL-cholesterol, VLDL-cholesterol and triglyceride and an increased LDL-cholesterol concentrations when compared to the negative control group is suggestive of toxicity induced by doxorubicin exposure. Treatment with aqueous extract of the aerial parts of *Leonurus cardiaca* at 166, 250 and 500 mg/kg for 7, 14, and 21 days resulted in significant increases on the mean plasma total cholesterol, HLD-C, VLDL-C and triglyceride level with decreases in LDL-cholesterol concentration (Table 3). The significant increases observed on the mean plasma total cholesterol, HDL-cholesterol, VLDL-cholesterol and triglyceride and decrease LDL concentrations when compared with the negative control values are suggestive of the ability of the extract to enhance metabolism of total cholesterol, HDL-cholesterol, VLDL-cholesterol and triglyceride levels against cisplatin induced toxicity in the rats. Meanwhile, Feng *et al.* [26] on ameliorative effects of Nacetylcysteine on fluoride-induced oxidative stress and DNA damage in male rats' testis, reported similar claims. This finding is agree with lipid parameters is in line with the publication of Maruthappan and Sakthi (2010) on hypolipidemic activity of Haritaki (*Terminalia Chebula*) in atherogenic diet induced hyperlipidemic rats.

Table 3 Effect of aqueous extract of the aerial parts on the plasma lipid profile on doxorubicin-induced damage in Wistar rats

Treatment	Total-CHIL (mg/dl)	HDL-CHOL (mg/dl)	LDL-CHO (mg/dl)	VLDL-CHOL (mg/dl)	TG (mg/dl)
N/Control	94.22±0.01	56.34±0.01	60.21±0.00	9.32±0.02	48.13±0.02
Ne/Control	37.13±0.02^{ab}	27.76±0.01^{ab}	84.26±0.01^{ab}	4.27±0.01^{ab}	25.64±0.01^{ab}
166mg/kg+DX 7 D	37.45±0.01^{b}	27.95±0.01^{ab}	84.04±0.01^{ab}	4.34±0.01^{ab}	25.93±0.01^{ab}

166mg/kg+DX14D	38.06±0.01^{b}	29.15±0.01^{b}	83.86±0.01^{ab}	4.75±0.01^{ab}	26.05±0.01^{ab}
166mg/kg+DX21D	73.25±0.01^{b}	31.42±0.01^{b}	83.46±0.01^{ab}	9.15±0.02^{b}	26.76±0.02^{b}
250 mg/kg+DX7D	41.05±0.02^{b}	31.64±0.00^{b}	83.22±0.01^{b}	5.06±0.00^{b}	27.15±0.01^{b}
250mg/kg+DX14D	41.66±0.01^{b}	31.94±0.01^{b}	83.02±0.01^{b}	5.46±0.00^{b}	27.65±0.01^{b}
250mg/kg+DX21D	41.66±0.01^{b}	34.22±0.00^{b}	61.44±0.02^{b}	5.23±0.01^{b}	27.65±0.01^{b}
500 mg/kg+DX7D	68.34±0.01^{b}	52.45±0.01^{b}	62.02±0.01^{b}	6.24±0.01^{b}	31.66±0.01^{b}
500mg/kg+DX14D	68.34±0.01^{b}	61.03±0.01^{b}	46.24±0.01^{b}	6.89±0.01^{b}	36.15±0.01^{b}
500mg/kg+DX21D	76.24±0.01^{b}	75.05±0.02^{b}	37.75±0.01^{b}	7.32±0.01^{b}	43.08±0.02^{b}

DX=doxorubicin. Values are reported as mean ± standard error of mean (M±SEM) (n =5). Values bearing superscript ("ab") were significantly different ($p \leq 0.05$) from the normal control down the group. Values bearing superscript ("b") were significantly different ($p \leq 0.05$) from the negative control down the group.

Table 4 shows the effect of aqueous of extract of the aerial parts of *L. cardiaca* on the oxidative stress biomarkers of heart homogenate on and doxorubicin-induced cardiovascular damage Wistar albino rats. The mean MDA, GSH concentrations, GPx, CAT and SOD activities of the normal control were 3.27±0.01 mmol/l, 78.40±0.03µg/mg protein, 92.10±0.02 mg/pro.min, 151.40±0.03 IU/g, and 28.82±0.03 mg/g respectively, those of the negative control were 15.47±0.02 mmol/l, 19.84±0.01µg/mg protein, 30.64±0.03 mg/pro.min, 73.16±0.01IU/g, and 7.35±0.02 mg/g respectively (Table 4). However, the mean plasma MDA, GSH concentrations, GPx, CAT and SOD activities in doxorubicin-induced cardiovascular damage rats, treated with aqueous extract of the aerial parts of *L. cardiaca* at 166 mg/kg b.w were 15.01±0.01, 21.32±0.02 mmol/l, 33.11±0.02 µg/mg protein, 79.87±0.02 mg/pro.min, and 7.85±0.02 mg/g respectively for 7 days, 13.73±0.01 mmol/l, 27.79±0.03 µg/mg protein, 48.07±10.02 mg/pro.min, 84.25±0.01 IU/g, and 2.15±0.01 mg/g respectively for 14 days, and 12.89±0.01 mmol/l, 31.02±0.01 µg/mg protein, 43.22±0.02 mg/pro.min, 95.32±0.01 IU/g, and 2.87±0.02 mg/g respectively for 21 days (Table 4). The mean MDA, GSH concentrations, GPx, CAT and SOD activities in doxorubicin-induced cardiovascular damage rats, treated with aqueous extract of the aerial parts of *L.*

cardiaca at 250 mg/kg were 10.56±0.01 mmol/l, 42.64±0.02 µg/mg protein, 52.73±0.01 mg/pro.min, 108.19±0.02 IU/g, and 8.65±0.02 mg/g respectively for 7 days, 10.06±0.02 mmol/l, 43.11±0.01 µg/mg protein, 54.23±0.02 mg/pro.min, 111.60±0.03 IU/g, and 8.95±0.01 mg/g respectively for 14 days, and 9.62±0.01 mmol/l, 43.94±0.01 µg/mg protein, 54.94±0.01 mg/pro.min, 112.06±0.01 IU/g, and 9.36±0.02 mg/g respectively for 21 days as presented in Table 4. Meanwhile, the mean MDA, GSH concentrations, GPx, CAT and SOD activities in doxorubicin-induced cardiovascular damage rats, treated with aqueous extract of the aerial parts of *L. cardiaca* at 500 mg/kg were 5.65±0.02 mmol/l, 61.48±0.01 µg/mg protein, 67.14±0.02 mg/pro.min, 142.73±0.01 IU/g, and 18.63±0.01 mg/g respectively for 7 days, 5.11±0.01, 61.95±0.01, 67.84±0.01, 143.01±0.01, and 18.95±0.02 respectively for 14 days, and 5.01±0.01mmol/l, 68.46±0.02 µg/mg protein, 70.21±0.01 mg/pro.min, 141.62±1.80 IU/g, and 19.05±0.02 mg/g respectively (Table 4). More so, administration of 18mg/kg of doxorubicin to the negative control resulted in a significant increase in the mean MDA and GSH concentrations which also significantly increased the mean GPx, CAT, and SOD activities in comparison to the normal control group. Concurrently, Treatment with the extract at 250 mg/kg b.w in doxorubicin-induced cardiovascular damage rats, caused significant decrease in the mean MDA and GSH levels as well as yielded significant increases on the mean GPx, CAT and SOD activities for 7, 14, and 21 days, in comparison to the negative control group and a more significant improvement were observed in 500 mg/kg treatment (Table 4).

Antioxidant enzymes such as glutathione peroxidase, glutathione S-transferase, and phospholipid hydroperoxide glutathione peroxidase, decompose lipid hydroperoxides to alcohols, and glutathione peroxidase and catalase also reduce hydrogen peroxide to nontoxic substances [27]. Lipid peroxidation generates a wide range of products such as malondialdehyde (MDA) which is

a biomarker of oxidative stress [28]. In this study, administration of 18mg/kg of doxorubicin to the negative control group significantly increased and decreased the mean MDA and GSH concentration respectively, with significant decreases in the GPx, CAT and SOD activities in comparison the positive control values (Table 4). The significant increased and decreased the mean MDA and GSH concentration respectively which indicative of lipid peroxidation (Table 4). Treatment with the extract at 250 and 5000mg/kg significantly caused decreased and increased MDA and GSH levels respectively as well as increases on the mean plasma GSH, GPx, CAT and SOD activities which is reflective of the ability of the extract to mop up free radicals. Wang *et al.* [29] demonstrated similar reports in their study on Lycopene's protective effect on oxidative damage of L02 cells and its mechanism. This findings is also in line with the report of Feng *et al*. [26] on the ameliorative effects of Nacetylcysteine on fluoride-induced oxidative stress and DNA damage in male rats' testis and Wang *et al.* [29] on Lycopene's protective effect on oxidative damage of L02 cells and its mechanism.

Intraperitoneal administration of 18mg/kg of doxorubicin resulted in increased fatty inflammation of myocytes, and disintegration of myocytes with loss of nuclei, intracellular accumulation and apoptosis, glomerular and tubular congestion, atrophic glomerulus in deep cortex, increase in inflammatory cells when compared to the positive control histology are suggestive of severe cardiotoxicity and renal damage. Treatment with aqueous extract of the aerial parts of *Leonurus cardiaca* at particulary at 250, and 500 mg/k b.wt for 7, 14 and 21 days resulted in regeneration of glomerular cell, improved glomeruli and tubules, capsule, changes incardiac, perivascular inflammation and normal myocytes, andrenalmorphology when compared to the positive and negative control histology. The significant regeneration of heart

tissue observed is suggestive of the ameliorative potential of crude aqueous extract of the aerial part of *Leonurus cardiaca*.

Table 4 Effect of aqueous extract of the aerial parts of *L. cardiaca* on the oxidative stress biomarkers of heart homogenate on doxorubicin-induced cardiovascular damage Wistar rats

Treatment	MDA (mmol/l)	GSH (µg/mg protein)	GPx (IU/g)	CAT (mg/pro.min)	SOD (mg/g)
N/Control	3.27±0.01^{a}	78.40±0.03^{a}	92.10±0.02^{a}	151.40±0.03^{a}	28.82±0.03^{a}
Ne/Control	15.47±0.02^{a,b}	19.84±0.01^{a,b}	30.64±0.03^{a,b}	73.16±0.01^{a,b}	7.35±0.02^{a,b}
166mg/kg+DX7	15.01±0.01^{b}	21.32±0.02^{b}	33.11±0.02^{b}	79.87±0.02^{b}	7.85±0.02^{b}
166mg/kg+X14	13.73±0.01^{b}	27.79±0.03^{b}	48.07±10.02^{b}	84.25±0.01^{b}	2.15±0.01^{b}
166mg/kg+DX21	12.89±0.01^{b}	31.02±0.01^{b}	43.22±0.02^{b}	95.32±0.01^{b}	2.87±0.02^{b}
250 mg/kg+DX7	10.56±0.01^{b}	42.64±0.02^{b}	52.73±0.01^{b}	108.19±0.02^{b}	8.65±0.02^{b}
250mg/kg+DX14	10.06±0.02^{b}	43.11±0.01^{b}	54.23±0.02^{b}	111.60±0.03^{b}	8.95±0.01^{b}
250mg/kg+DX21	9.62±0.01^{b}	43.94±0.01^{b}	54.94±0.01^{b}	112.06±0.01^{b}	9.36±0.02^{b}
500 mg/kg+DX7	5.65±0.02^{b}	61.48±0.01^{b}	67.14±0.02^{b}	142.73±0.01^{b}	18.63±0.01^{b}
500mg/kg+DX14	5.11±0.01^{b}	61.95±0.01^{b}	67.84±0.01^{b}	143.01±0.01^{b}	18.95±0.02^{b}
500mg/kg+DX21	5.01±0.01^{b}	68.46±0.02^{b}	70.21±0.01^{b}	141.62±1.80^{b}	19.05±0.02^{b}

DX=doxorubicin. Values are reported as mean ± standard error of mean (M±SEM) (n =5). Values bearing superscript ("ab") were significantly different ($p \leq 0.05$) from the normal control down the group. Values bearing superscript ("b") were significantly different ($p \leq 0.05$) from the negative control down the group.

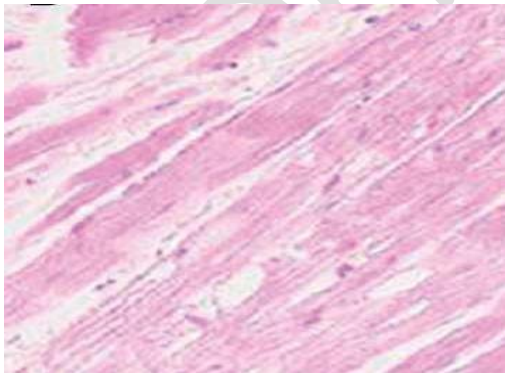

Plate 1:N/C

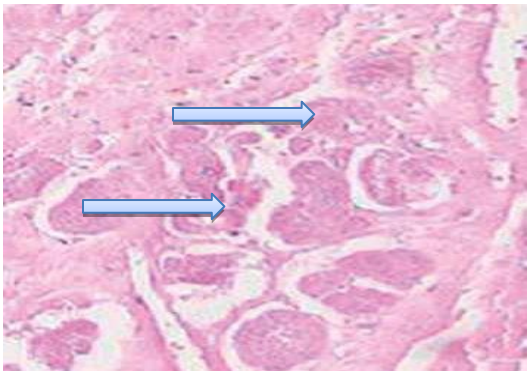

Plate 2: Ne/C

Plate 3: Positive control

Plates 1-3: Photomicrograph heart tissue of the normal, negative and positive control (H & E staining) x 400. Normal control (N/C) was orally administered with distilled water and rat feed, Negative control (Ne/C) was intraperitoneally administered 18mg/kg b.w of doxorubicin once and observed for 21 days, showing large area with myocytes with hypereosinophilic cytoplasm, striation, and nuclear changes in cell death, pyknosis, karrheorhexis, and karyolysis.

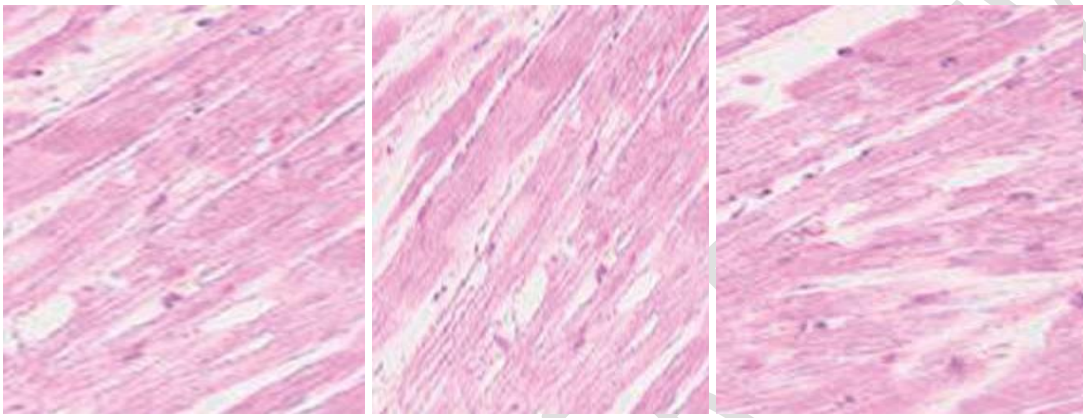

Plate 4 Plate 5 Plate 6

Plates 4-6: Photomicrograph heart tissue of the normal, negative and positive control (H & E staining) x 400. Plate 4 was treated with crude extract of *Leonurus cardiaca* at 166mg/kg for 7 days. Plate 5 was treated with crude extract of *Leonurus cardiaca* at 166mg/kg for 14 days while Plate 6 was treated with crude extract of *Leonurus cardiaca* at 166mg/kg for 21 days, showing almost normal heart tissues.

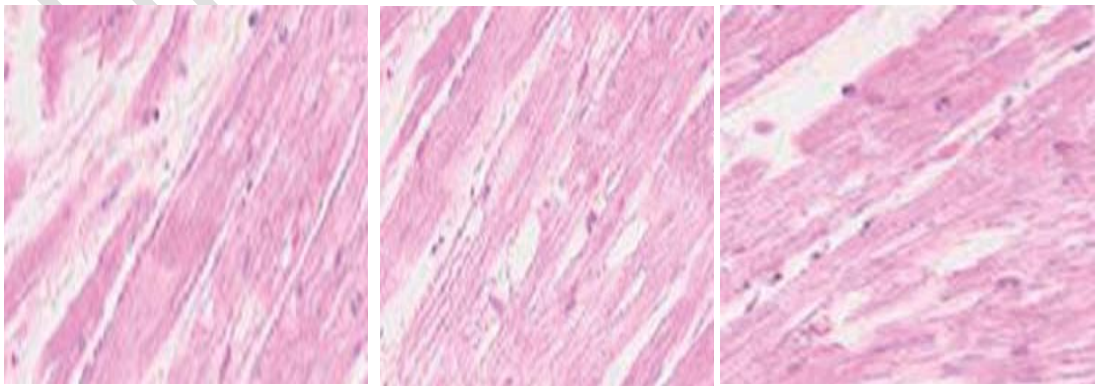

Plate 7 Plate 8 Plate 9

Plates 7-9: Photomicrograph of heart tissues on doxorubicin-induced cardio-renal damage rats, treated with 250 mg/kg b.w of aqueous extract of the aerial parts of *Leonurus cardiaca* for 7, 14, and 21 days respectively (H&S staining) x 400. Plate 7 was treated with crude extract of *Leonurus cardiaca* for at 250mg/kg 7 days. Plate 5 was treated with crude extract of *Leonurus cardiaca* at 250mg/kg for 14 days while Plate 6 was treated with crude extract of *Leonurus cardiaca* at 250mg/kg for 21 days, showing almost heart normal heart histology

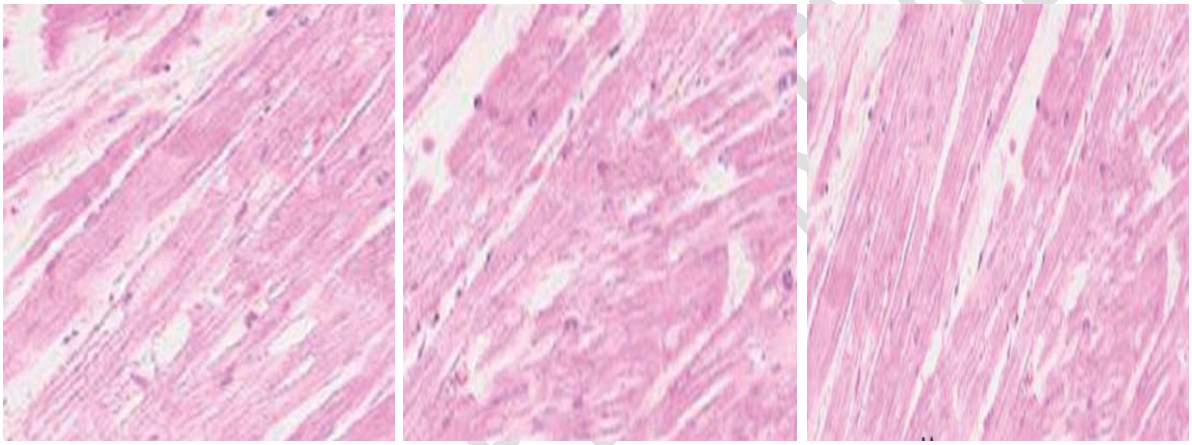

Plate 10 Plate 11 Plate 12

Plates 10-12: Photomicrograph of heart tissues on doxorubicin-induced cardio-renal damage rats, treated with 500 mg/kg b.w of aqueous extract of the aerial parts of *Leonurus cardiaca* for 7, 14, and 21 days respectively (H&S staining) x 400. Plate 10 was treated with crude extract of *Leonurus cardiaca* for at 500mg/kg 7 days. Plate 11 was treated with crude extract of *Leonurus cardiaca* at 500mg/kg for 14 days while Plate 12 was treated with crude extract of *Leonurus cardiaca* at 500mg/kg for 21 days, indicating normal heart tissues.

4. CONCLUSION

The consequence of cardiovascular damage facilitated by a known chemotherapeutic drug intends that medicinal plant with ameliorative potential signifies a hopeful avenue for management. Plans for the management and preclusion of cardiovascular dysfunction requires considerate of mechanism by which the prophylactic agents may possibly preclude the lethal effects. *Leonurus cardiaca* may be preferable for doxorubicin-induced cardiovascular damage by hampering oxidative trauma. However, this warrants further investigations to confirm the mechanism of action and develop approaches against doxorubicin-induced cardiovascular damage.

NOTE

This study highlighted the effectiveness of "traditional medicine" which is an ancient tradition practiced in some parts of India. This ancient concept should be carefully investigated in the light of modern clinical science and can be adopted partially if considered appropriate.

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