

Telfairia occidentalis seed extract and fractions mitigated doxorubicin-induced cardiotoxicity in rats

Ugochi Q. Nwosu ¹, Kenneth Chidi Opara ², Chinyelu C. Osigwe ³, Ugonma F. Uwaeme ³, Unyime A. Fabian ⁴ and Jude E. Okokon ^{1,*}

¹ Department of Pharmacology and Toxicology, Faculty of Pharmacy, University of Uyo, Uyo, Nigeria.

² Department of Family Medicine, Federal Teaching Hospital, Owerri, Imo State, Nigeria.

³ Department of Pharmacology and Toxicology, Faculty of Pharmacy, Madonna University Nigeria, Elele campus, Rivers State, Nigeria.

⁴ Faculty of Allied Health sciences, Department of Medical Laboratory Sciences University of Uyo, Uyo, Nigeria.

GSC Biological and Pharmaceutical Sciences, 2025, 33(01), 019-030

Publication history: Received on 21 August 2025; revised on 01 October 2025; accepted on 03 October 2025

Article DOI: <https://doi.org/10.30574/gscbps.2025.33.1.0375>

Abstract

Telfairia occidentalis Hook (Cucurbitaceae) seeds are used in the preparation of soups and as medicine traditionally to treat various diseases. This study was designed to investigate the antidotal potentials of the crude extract and fractions of *Telfairia occidentalis* seed against doxorubicin-induced heart toxicity in rats. The seed extract (138-553 mg/kg) and fractions (dichloromethane and aqueous, 276 mg/kg) of the plant were investigated for antioxidative stress and cardioprotective potentials against doxorubicin-induced heart toxicity in rat. Effect of the seed extract and fractions on cardiac marker enzymes, oxidative stress markers, lipid profile indices and heart histology were used as parameters to assess the cardioprotective effect of the extract and fractions. The seed extract and fractions (138-553 mg/kg) significantly ($p < 0.05-0.01$) reduced the serum levels of CK-MB, LDH and troponin I that were elevated by doxorubicin. Also, the levels of GSH, SOD, GPx and CAT in the heart that were decreased by doxorubicin were significantly ($p < 0.01$) elevated and the raised MDA level was reduced by the seed extract and fractions. The seed extract and fractions also reduced significantly ($p < 0.05$) the serum levels of total cholesterol, triglycerides, LDL and VLDL of the treated rats elevated by doxorubicin. Histology sections of hearts of extract/fractions -treated animals showed reductions in the pathological features compared to the organotoxic-treated animals. The chemical pathological changes were consistent with histopathological observations suggesting marked cardioprotective potentials. The anti-toxic effect of this plant may in part be mediated through the chemical constituents of the plant. The seed extract of *Telfairia occidentalis* possesses anti-toxicant properties which can be exploited in the treatment of poisoning.

Keywords: *Telfairia occidentalis*; Anti-Toxicant; Oxidative Stress; Cardioprotective; Antioxidant

1. Introduction

Drug-induced cardiotoxicity is a serious clinical challenge which is associated with so many clinically useful drugs especially cytotoxic drugs one of which is doxorubicin. This often results in cardiac dysfunction and myocardial injury among others which can lead to long term morbidity even after the discontinuation of the drug [1,2]. Doxorubicin (DOX)-induced cardiotoxicity has been suggested to results from free radicals' generation and lipid peroxidation in myocardial cells [3,4]. These undesirable organs toxicities associated with doxorubicin have limited its clinical usefulness and therefore necessitate extensive research into agents that can prevent and or ameliorates these toxic effects.

* Corresponding author: Jude E. Okokon

Telfairia occidentalis Hook is a fluted pumpkin of the Cucurbitaceae family widely consumed as food in Nigeria [5]. It is a popular vegetable all over Nigeria, especially in the Niger-Delta region and the Eastern part of the country; varieties of meals are prepared from the leaves, stem and seeds of the plant [6]. The seeds are very nutritious and are eaten roasted or boiled. The seed has history of being effective in the treatment and prevention of prostrate disorders. The seed extract has been reported to exert antidiabetic [7], cellular antioxidant, immunodulatory, anticancer, antiinflammatory [8], antiplasmodial [5], antioxidant and analgesic [9,10], genotoxic and cytotoxic [11] and in vivo inhibitory effect alpha amylase and alpha glucosidase [12]. while the leaf extract possesses antioxidant, antibacterial [13], hepatoprotective [14] antidiabetic [15], antiplasmodial [5] genotoxic and cytotoxic [11] and antiprostatic [16] activities. Phytochemical studies of the seed extract have shown the presence of alkaloid, flavonoid, tannins, terpenes, saponin, and cardiac glycosides [17]. Okokon et al. [8] reported the presence of compounds such as pentadecanoic acid, hexadecanoic acid; 16-octadecenoic acid methyl ester; 9, 12-octadecadienoyl chloride (Z,Z); 9- octadecadienoic acid (Z)-, 2, 3-dihydroxypropyl ester; octadecanoic acid; hexadecanoic acid, 2,3-is[[trimethylsilyl] oxy] propyl ester, 2,4-heptadien-6-ynal, (E,E); benzoic acid ; dodecanoic acid ; linoleic acid ethyl ester ; hexadecanoic acid, methyl ester ; α -phellandrene; α -campholene aldehyde; terpinen-4-ol; trans- β -ocimene; borneol and stigmastan-3- ol, in the seed extract.

The present study was designed to evaluate the activities of seed extract and fractions of *T. occidentalis* against doxorubicin-induced cardiotoxicity in rats.

2. Materials and Methods

2.1. Plant collection and extraction

Fresh seeds of *Telfairia occidentalis* were purchased from Itam market in Itu L. G. A, Akwa Ibom State, Nigeria, in June, 2023. The seeds were previously identified and authenticated by a taxonomist in the Department of Botany, University of Uyo, Uyo, Nigeria. Herbarium specimens (UUPH 1(b)) were deposited at Department of Pharmacognosy and Natural Medicine Herbarium, University of Uyo.

The fresh seeds of the plant were dried on laboratory table for 2 weeks and reduced to powder. The seeds powder (1 kg) was macerated in 50% ethanol (5000 mL) for 72 hours. The liquid filtrates obtained were concentrated at 40°C and all the ethanol was completely removed. The crude extract (20 g) was dissolved in 500 mL of distilled water and partitioned with equal volume of dichloromethane (DCM, 5 x 500 mL) till no colour change was observed, to obtain DCM and aqueous fractions. The extract and fractions were stored at 4°C in a refrigerator until used for the experiment.

2.2. Animals

In this study, male albino Wistar rats (150-200 g) were used. The animals were sourced from University of Uyo Animal house and sheltered in plastic cages. The rats were fed with pelleted standard Feed (Guinea feed) and given unlimited access to water. The study was approved by College of Health Sciences Animal Ethics Committee, University of Uyo.

2.3. Experimental design

In this study, repeated dose model earlier described by Olorundare et al. [18], which lasted for 14 days was used. Groups I rats which served as the untreated control were orally pretreated with 10 mL/kg/day of distilled water. Group 2 rats were given normal saline (10 mL/kg/day) but equally treated intraperitoneally with 1.66 mg/kg of doxorubicin hydrochloride dissolved in 0.9% normal saline on alternate days for 14 days. Groups 3-5 rats were orally pretreated respectively with 138 mg/kg/day, 276 mg/kg/day, and 553 mg/kg/day of *Telfairia occidentalis* seed extract dissolved in 10% Tween 80, 2 hours before treatment with 1.66 mg/kg of doxorubicin dissolved in 0.9% normal saline administered intraperitoneally on alternate days for 14 days. Groups 6 and 7 were pretreated with 276 mg/kg of DCM and aqueous fractions respectively but administered with 1.66 mg/kg of doxorubicin dissolved in 0.9% normal saline administered intraperitoneally on alternate days for 14 days Group 8 rats which served as the positive control group were equally pretreated with 100 mg/kg/day of silymarin two hours before treatment with 1.66 mg/kg of doxorubicin in 0.9% normal saline administered intraperitoneally on alternate days for 14 days.

2.4. Collection of blood samples and organs

After 14 days of treatment (24 hours after the last administration), the rats were weighed again and sacrificed under light diethyl ether vapour. Blood samples were collected into plain centrifuge tubes and allowed to stand for two hour before being centrifuged at 1500 rpm for 15 mins to separate the serum at room temperature and used for biochemical assays. The hearts of the rats were surgically removed, weighed. Each heart was gently and carefully divided into two

parts; a part was fixed in 10 % formaldehyde for histological processes, while the other part of the heart was briskly rinsed in ice cold 1.15% KCl solution and put in a clean sample bottle. These were stored in ice cold 0.9% NaCl.

2.5. Biochemical Assays

The stored sera samples collected from the rats used in this study were used for the biochemical assays. The clear samples were obtained for assays of the following biochemical parameters: serum cardiac troponin I, lactate dehydrogenase (LDH), and creatine phosphokinase (CK-MB).

2.5.1. Measurement of Cardiac marker Enzymes.

Cardiac enzymes (Creatine phosphokinase –MB (CK-MB), lactic acid dehydrogenase (LDH), and troponin I were measured by using the ELISA kit. All the tests were performed according to the manufacturer's instruction. LDH (LDH kit-Aspen Laboratories Pvt. Ltd., Baddi; India.), Troponin I and CK-MB (Teco Diagnostics, Anaheim, USA).

2.5.2. Assessment of effect of seed extract on lipid profile

The stored sera samples collected from the rats used in this study were used to determine the lipid profile parameters such as serum cholesterol, triglyceride and high-density lipoprotein (HDL) levels of the treated rats using standard colorimetric methods [19]. These lipid parameters determinations were done spectrophotometrically using Fortress Diagnostic Kits® according to standard procedures of manufacturer's protocols. Low and very low-density lipoprotein (LDL and VLDL) were estimated from the formula of Friedwald et al. [20].

2.5.3. Effect of the seed extract on heart oxidative stress markers

The parts of heart samples stored in ice cold normal saline were used in preparation of homogenates for assay of oxidative stress markers. Homogenates were made in a ratio of 1 g of wet tissue to 9 mL of 1.25% KCl by using motor driven Teflon-pestle. The homogenates were centrifuged at 7000 rpm for 10 min at 4°C and the supernatants were used for the assays of superoxide dismutase (SOD) [21], catalase (CAT) [22], glutathione peroxidase (GPx) [23], reduced glutathione (GSH) [24] and malondialdehyde (MDA) content [25]. These oxidative stress markers were used to assess antioxidative stress potentials of the extract.

2.5.4. Histopathological studies

The hearts' parts fixed in 10 % buffered formalin were used for histological processes. They were processed and stained with haematoxylin and eosin (H&E) [26], according to standard procedures at Department of Chemical Pathology, University of Uyo Teaching Hospital, Uyo. Morphological changes were observed and recorded in the excised organs of the sacrificed animals. Histologic pictures were taken as micrographs.

2.6. Statistical analysis

Data collected were analyzed using one way analysis of variance (ANOVA) followed by Tukey Kramer's multiple comparison post-test (Graph pad prism software Inc. La Jolla, CA, USA). Values were expressed as mean \pm SEM and significance relative to control were considered at $p < 0.001$ and $p < 0.05$.

3. Results

3.1. Effect of seed extract and fractions of *T. occidentalis* on body and organs weights of rats with doxorubicin-induced toxicity

Administration of *T. occidentalis* seed extract and fractions to rats with doxorubicin-induced organs toxicities caused considerable improvement of the body weights compared to the organotoxic group. The crude extract (276 mg/kg) and dichloromethane fraction treated groups recorded the highest body weight gains compared to the organotoxic group. However, these were non-dose-dependent and insignificant when compared to the organotoxic group. The heart weights of the group treated with doxorubicin only were found to be increased when compared to those of the normal control group though not statistically significant ($p > 0.05$). However, concomitant treatment of rats with doxorubicin and seed extract and fractions of *T. occidentalis* improved the organs weights though insignificantly ($p > 0.05$) relative to the group treated with doxorubicin only (Table 1).

3.2. Evaluation of effect of seed extract and fractions of *T. occidentalis* on cardiac marker enzymes of doxorubicin-induced heart injuries in rats:

The effect of seed extract and fractions of *T.occidentalis* on cardiac marker enzymes of normal rats and rats with doxorubicin-induced toxicities is as presented in Table. Treatment of rats with doxorubicin (1.66 mg/kg i.p) on alternate days for 14 days caused significant ($p<0.01-0.001$) elevation of cardiac marker enzymes (troponin I, lactate dehydrogenase and CK-MB) activities when compared to normal control. The increases in the levels of lactate dehydrogenase, troponin I and CK-MB were dose-dependently and significantly ($p< 0.05-0.001$) reduced when compared to organotoxic group following concomitant treatment of the rats with seed extract and fractions of *T.occidentalis* (138-553 mg/kg) with the highest dose of the extract (553 mg/kg) and aqueous fraction having the highest effect. Similar effect was observed in the group treated with silymarin. (Figures 1, 2 and 3).

3.3. Effect of *T. occidentalis* seed extract and fraction on heart oxidative stress markers of doxorubicin-induced heart toxicity

Table 2 shows the effect of *T. occidentalis* seed extract/fractions on oxidative stress markers of the heart. Administration of doxorubicin (1.66 mg/kg i.p) on alternate days for 14 days caused decreases of cardiac antioxidant enzymes activities (SOD, GPx, GST, CAT) and GSH levels which were significant ($p<0.05-0.001$) when compared to control except in CAT level. The MDA level was also significantly ($p<0.01$) elevated by doxorubicin treatment relative to control. However, concomitant administration of seed extract/fractions of *T.occidentalis* (138- 553 mg/kg) with doxorubicin for 14 days caused marked elevations of the enzymatic and non-enzymatic endogenous antioxidants in the treated rats' groups when compared to the organotoxic groups. The elevations in the levels of SOD and CAT were dose-dependent and significant (0.05-0.01) in the groups treated with the highest dose (553 mg/kg). DCM fraction treated group had the most significant ($p<0.001$) elevation of SOD, while CAT level was significantly ($p<0.001$) raised by aqueous fraction when compared to control. GPx and GSH levels were also dose-dependently elevated with DCM fraction treated group having the most significant ($p<0.001$) effect when compared to organotoxic group. The treatment with extract/fractions also caused marked non-dose-dependent reduction in the level of MDA of the treated rats which were significant ($p<0.05-0.001$) in the groups treated with the low dose (138 mg/kg), aqueous fraction, DCM fraction and silymarin when compared to organotoxic control with the DCM fraction having the most significant effect (Table 2).

3.4. Effect of seed extract and fraction of *T.occidentalis* on lipid profile of rats with doxorubicin -induced organs toxicities

Administration of doxorubicin (1.66 mg/kg) was observed to caused significant ($p<0.05-0.001$) elevation in levels of total cholesterol, triglyceride, high density lipoprotein, low density lipoprotein, and very low-density lipoprotein. These raised levels of total cholesterol, triglyceride, high density lipoprotein, low density lipoprotein, and very low-density lipoprotein were significantly ($p<0.05-0.001$) reduced when compared to organotoxic group following concomitant treatment with seed extract and fractions of *T.occidentalis* and silymarin. However, the reductions in total cholesterol and low-density lipoprotein were dose-dependent, while non-dose-dependent reductions were observed in triglyceride, high density lipoprotein and very low-density lipoprotein levels (Table 3).

3.4.1. Effect of seed extract and fractions of *T. occidentalis* on histology of rat heart in doxorubicin-induced cardiotoxicity:

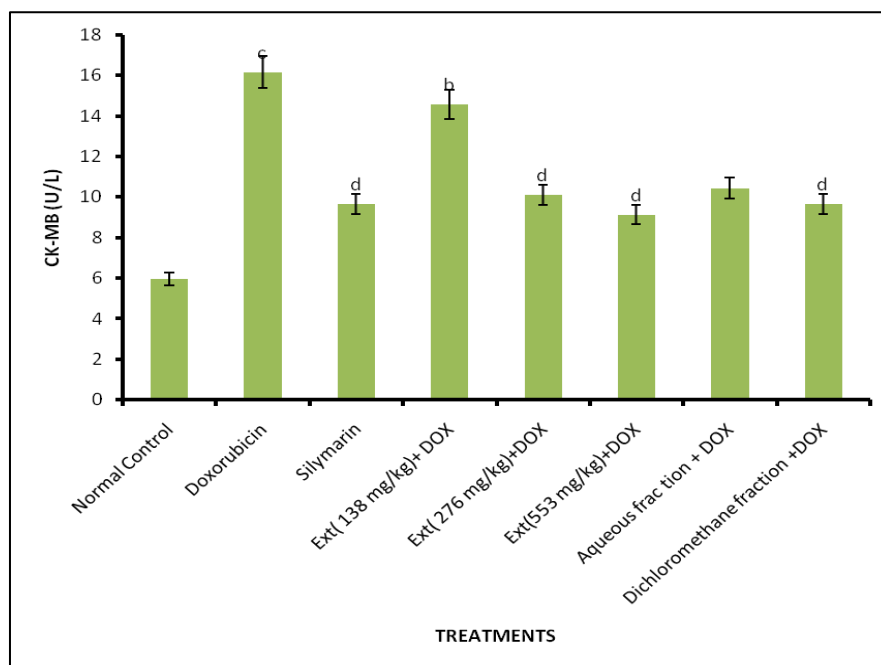
Histologic sections of hearts of rats receiving various treatments at magnification (x100) stained with H&E method revealed that Group 1 (normal control, CONT) treated distilled water (10 mL/kg) had cardiac section showing normal cardiac architecture with the myometrium having well-presented cardiac myocytes, myocyte nuclei, fibrocyte nuclei and endomysium and blood vessels. No pathological change was seen. The organotoxic group (Group 2, T+CONT) treated with doxorubicin (1.66 mg/kg) only revealed heart tissue showing presence of inter-muscular vascular hemorrhage and fibrosis within the cardiac myometrium. Focal hyaline degeneration was also seen. Group 3 (T+STD) treated with 100 mg/kg of silymarin of *T.occidentalis* seed and doxorubicin (1.66 mg/kg) had moderately affected heart tissue showing presence of inter-muscular vascular hemorrhage within the cardiac myometrium. Rats in group 4 (T+LDE) treated with 138 mg/kg of *T.occidentalis* seed extract and doxorubicin (1.66 mg/kg) showed moderately affected cardiac tissue revealing the presence of fibrosis within the cardiac myometrium. Group 5 (T+MDE) treated with 276 mg/kg of *T. occidentalis* seed extract and doxorubicin (1.66 mg/kg) showed heart tissue showing presence of inter-muscular vascular hemorrhage and fibrosis within the cardiac myometrium. Group 6 (T+HDE) treated with 553 mg/kg of *T. occidentalis* seed extract extract and doxorubicin (1.66 mg/kg) showed moderately affected heart tissue showing the presence of inter-muscular vascular hemorrhage and fibrosis within the cardiac myometrium. Group 7 (T+AQE) treated with 276 mg/kg of aqueous fraction of *T. occidentalis* seed and doxorubicin (1.66 mg/kg) showed a moderately affected heart tissue showing the presence of fibrosis within the cardiac myometrium. Rats in group 8 (T+DCME) treated with 276 mg/kg of dichloromethane fraction of *T. occidentalis* seed and doxorubicin (1.66 mg/kg)

moderately affected heart tissue showing the presence of inter-muscular vascular hemorrhage and fibrosis within the cardiac myometrium (Figure 4).

Table 1 Effect of *T. occidentalis* seed extract on body and organs weights of rats with doxorubicin-induced toxicity

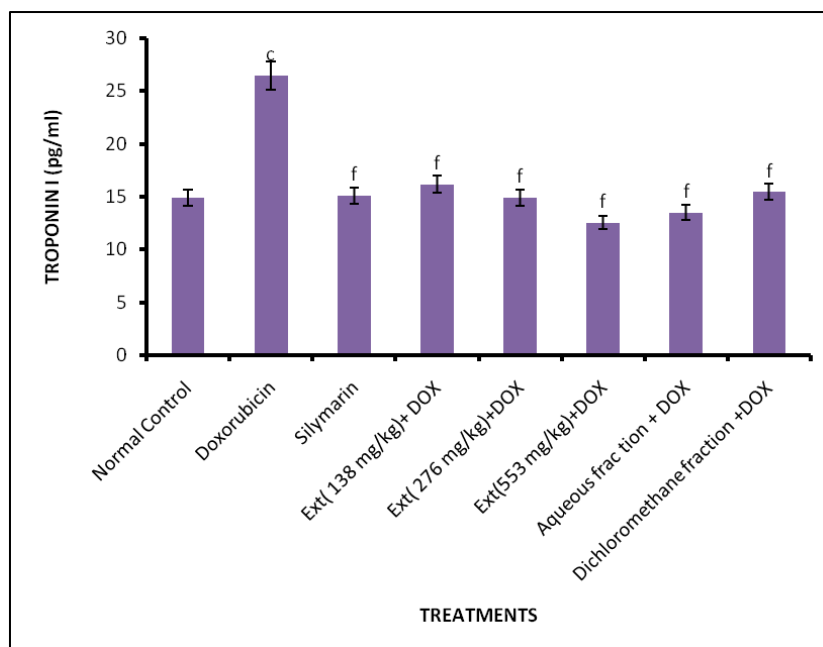
Parameters/ Treatment	Dose mg/kg	Heart	Body weight		
			Before	After	% increase in body weight
Normal control	-	0.60± 0.06	176.28±17.97	198.25± 6.61	12.46
Doxorubicin	1.66	0.65± 0.01	169.66 ± 6.80	181.66±13.24	5.72
Silymarin+DOX	100	0.63±0.03	180.33± 10.86	190.66± 13.24	5.72
Extract+DOX	138	0.65±0.05	176.0± 11.13	191.0± 6.08	8.52
	276	0.58±0.06	167.0± 7.57	185.0 ± 8.73	10.77
	553	0.65±0.03	177.66± 7.42	193.33 ± 5.48	8.82
Aqueous fraction	276	0.69±0.10	187.66± 17.89	195.33± 17.70	4.08
DCM fraction	276	0.52±0.06	162.66± 12.66	180.33± 9.56	10.86

Data are expressed as mean ±SEM. significant at dp<0.001 when compared to normal control; ap< 0.05, bp< 0.01, cp< 0.001 when compared to organotoxic control. n = 5.



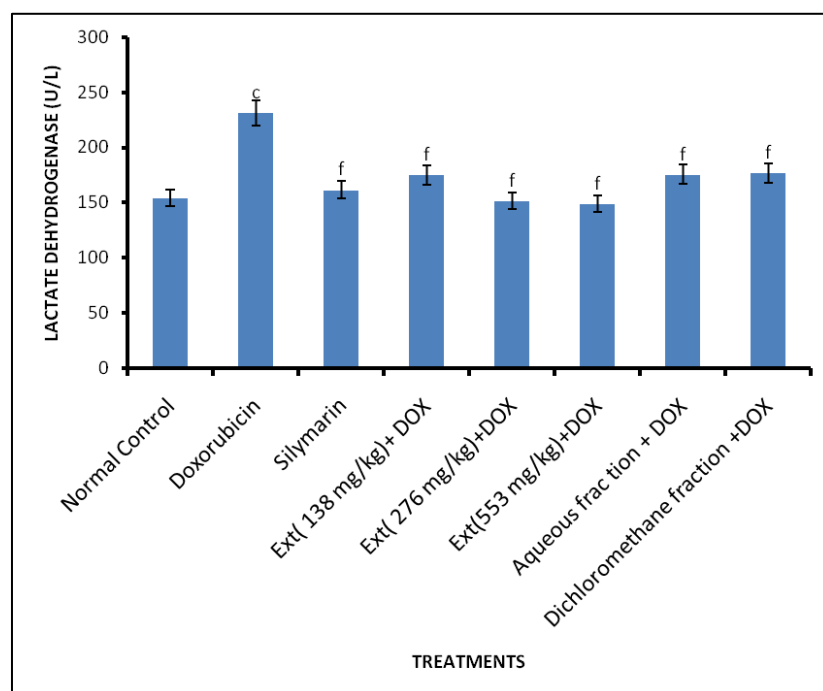
Data are expressed as MEAN ± SEM, Significant at bp<0.01; cp<0.001 when compared to normal control; dp<0.05 when compared to organ toxic control; *p< 0.001 (n=5).

Figure 1 Effect of *T.occidentalis* seed extract and fractions on Creatine phosphokinase-MB enzyme of rats with doxorubicin-induced heart toxicity



Data are expressed as MEAN \pm SEM, Significant at $p < 0.001$ when compared to normal control; $p < 0.001$, when compared to organotoxic control (n=5).

Figure 2 Effect of *T. occidentalis* seed extract and fractions on Troponin I enzyme of rats with doxorubicin-induced heart toxicity



Data are expressed as MEAN \pm SEM, Significant at $p < 0.001$ when compared to normal control; $p < 0.001$, when compared to organotoxic control (n=5).

Figure 3 Effect of *T.occidentalis* seed extract and fractions on lactate dehydrogenase enzyme of rats with doxorubicin-induced heart toxicity

Table 2 Effect of *T. occidentalis* seed extract and fractions on cardiac oxidative stress markers of rats with doxorubicin-induced toxicity

Treatment	Dose mg/kg	SOD (U/ml)	CAT (U/g of protein)	GPX (μg/ml)	GSH (μg/ml)	MDA (μMol/ml)
Control	10	0.29± 0.01	1.86±0.02	0.056±0.003	2.57± 0.01	0.45± 0.01
Doxorubicin	1.66	0.18± 0.02 ^a	1.42±0.08	0.050±0.005 ^a	2.23± 0.01 ^b	0.64± 0.02 ^b
Crude extract	138	0.20±0.03	2.32±0.38	0.050±0.003 ^a	2.49± 0.14	0.47±0.01 ^e
	276	0.22±0.03	2.61±0.17	0.056±0.002 ^d	2.50± 0.13	0.54±0.05
	553	0.28±0.02 ^d	3.42± 0.16 ^{a, f}	0.056±0.002 ^d	2.53± 0.12	0.58±0.02
Aqueous Fraction	276	0.28±0.03 ^d	3.01±0.61 ^e	0.049±0.005 ^b	2.17± 0.01 ^a	0.48± 0.04 ^d
DCM fraction	276	0.35±0.02 ^f	2.36±0.16	0.067±0.008 ^{c, f}	3.06± 0.04 ^{b, f}	0.41± 0.05 ^f
Silymarin	100	0.28±0.02 ^d	1.63±0.27	0.045±0.008 ^c	2.38± 0.03 ^a	0.47±0.01 ^e

Data are expressed as MEAN ± SEM, Significant at ap<0.05, bp<0.01, cp<0.001, when compared to control; Significant at dp<0.05, ep<0.01, fp<0.001 compared to organotoxic group. (n=5)

Table 3 Effect of *T. occidentalis* seed extract and fractions on lipid profile parameters of rats with doxorubicin-induced toxicity

Treatment	Dose mg/kg	Total cholesterol (MMOL/L)	Triglyceride (MMOL/L)	HDL-C (MMOL/L)	LDL-C (MMOL/L)	VLDL (MMOL/L)
Control	10	3.33± 0.20	1.21±0.03	1.30±0.02	2.70± 0.22	0.56± 0.02
Doxorubicin	1.66	4.60± 0.15 ^b	1.55±0.05 ^c	1.60±0.06 ^a	3.70± 0.09 ^c	0.70± 0.02
Crude extract	138	3.80±0.11	1.31± 0.03 ^e	1.44±0.01	2.95± 0.11 ^e	0.60±0.01
	276	3.33±0.23 ^f	1.35±0.03 ^d	1.45±0.04	2.49± 0.20 ^f	0.61±0.01
	553	2.63±0.17 ^f	1.14±0.02 ^f	1.23±0.04 ^e	1.92± 0.13 ^f	0.51±0.01 ^e
Aqueous Fraction	276	2.53±0.17 ^f	1.08±0.07 ^f	1.23±0.06 ^e	1.78± 0.15 ^{a, f}	0.49± 0.03 ^e
DCM fraction	276	2.40±0.17 ^f	1.08±0.02 ^f	1.19±0.07 ^e	1.70± 0.14 ^{a, f}	0.49± 0.04 ^e
Silymarin	100	3.00±0.28 ^f	1.09±0.06 ^f	1.20±0.07 ^e	2.29± 0.24 ^f	0.48± 0.04 ^e

Data are expressed as MEAN ± SEM, Significant at ap<0.05, bp<0.01, cp<0.001, when compared to control; Significant at dp<0.05, ep<0.01, fp<0.001 compared to organotoxic group. (n=5)

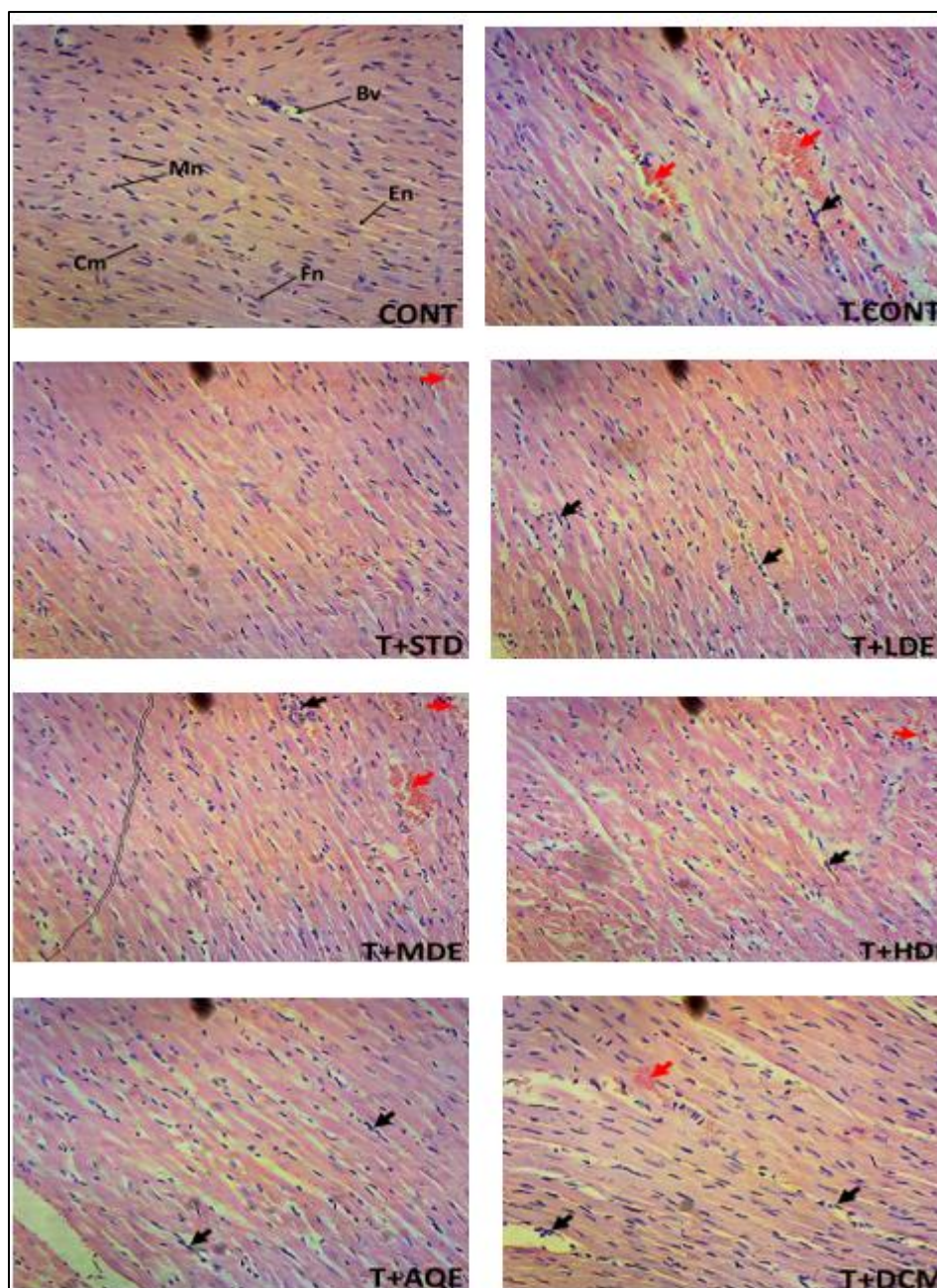


Figure 4 Photomicrographs of sections of hearts of rats treated with distilled water (CONT), doxorubicin only, 1.66 mg/kg (T.CONT), Sylimar, 100 mg/kg and DOX (T+STD), *T.occidentalis*, 138 mg/kg and DOX (T+LDE), *T.occidentalis*, 276 mg/kg and DOX (T+MDE), *T.occidentalis*, 553 mg/kg and DOX (T+HDE), Aqueous fraction, 276 mg/kg and DOX (T+AQE) and DCM fraction, 276 mg/kg and DOX (T+DCME) showing well-presented cardiac myocytes (Cm), myocyte nuclei (Mn), fibrocyte nuclei (Fn) and endomysium (En) and blood vessels (Bv), inter-muscular vascular hemorrhage (red arrow) and fibrosis (black arrow) (H&E x 100)

4. Discussion

Doxorubicin (DOX) is an anthracycline glycoside antibiotic with potent anticancer activity [27], whose clinical usefulness has been marred by irreversible and dose-dependent cardiomyopathy, nephrotoxicity and hepatotoxicity induced by the drug despite having a high therapeutic index [2]. DOX alters structural and functional processes due to its high affinity to phospholipids in the mitochondrial membrane in the cardiac cells [1], thus resulting in nucleic acid damage, sarcomere disruption, and myofibril loss [28]. These undesirable effects have been traced to the activities of its metabolite, DOX-semiquinone, which reacts with O_2 , producing H_2O_2 and O_2^- (superoxide) [3], and inhibits the

activities of endogenous enzymatic and nonenzymatic antioxidants thereby creating oxidative stress with damaging effect on the heart compared with other organs such as the kidney and liver [3,4].

This study was designed to assess the antitoxicant potentials of seed extract and fractions of *Telfairia occidentalis* against DOX-induced cardiotoxicity. The intraperitoneal administration of DOX showed cardiomyopathy manifested by raised levels of CK-MB, troponin I, and LDH and are consistent with earlier studies [29]. It has been observed that the mortality rate in animals treated with DOX was 40% compared with the normal control group before the end of experiment which may be due to accumulation of ascites resulting from abnormality in cardiac functioning [30]. Ascites is thought to developed due to extracellular volume expansion and fluid leakage towards interstitium due to tubular disorder and sodium retention as proposed in previous studies [31]. Co-administration of the seed extract of *T. occidentalis* in this study was observed to reduce the levels of troponin I, LDH and CK-MB, portraying a significant cardioprotective activity. This activity can be attributable to the free radicals scavenging activities of the phytochemical constituents of the seed extract as reported previously [8,9,13].

Reactive oxygen species generation, inflammatory processes and lipid peroxidation have been suggested to be responsible for doxorubicin-induced cardio and hepatotoxicity [32,33].

The findings of this study show that administration of doxorubicin (1.66 mg/kg, i.p) on alternate days for 14 days to rats caused significant decreases ($p < 0.05$) in levels of enzymatic and non-enzymatic endogenous antioxidants (SOD, CAT, GPx and GSH) in the heart and elevated level of MDA when compared to control. Lipid peroxidation is a marker of oxidative stress and elevations in the amount of malondialdehyde (MDA), a lipid peroxidation product, have been reported following Dox treatment [34,35,36]. Similar trend was observed in this study. Coadministration of seed extract and fractions of *T. occidentalis* (138 - 553 mg/kg) with doxorubicin caused significant ($p < 0.05-0.001$) non-dose-dependent elevation in the levels of the antioxidant enzymes (SOD, CAT, GPx) when compared to control. Similarly, GSH level was significantly ($p < 0.001$) elevated following treatment with the extract when compared to control. Similarly, there were significant ($p < 0.05-0.01$) reductions in the level of MDA of the extract-treated rats. It has been documented that DOX inhibits the activities of endogenous enzymatic and nonenzymatic antioxidants as was the case in this study. So, an imbalance between ROS generation and neutralization leads to oxidative stress and injury to the heart [3,4,28]. The seed extract potentials to reduce the level of MDA shows a reduction in lipid peroxidation and generation of free radicals which might have been scavenged by the phytoconstituents present in this extract, hence the protective effect on the heart.

Histological sections of rat hearts treated with doxorubicin alone showed section with degenerating cardiac muscle fibers which is a manifestation of the effect of doxorubicin toxicity due to the effect of free radicals generated by the drug. However, co-administration of *T. occidentalis* seed extract/fractions and doxorubicin reduced the effect of doxorubicin as was seen in mild effect to absent of pathological signs confirming the cardioprotective effect of *Telfairia occidentalis* seed. This effect can be attributed to the antioxidant activities of the phytochemical constituents of the seed extract such as the monoterpenes and polyunsaturated fatty acid (PUFA) previously reported in this seed extract which have been documented to exert antioxidant activities [8].

However, doxorubicin was found to raised significantly the levels of total cholesterol, triglyceride, HDL, LDL and VLDL. This result is consistent with that of Okokon et al. [37]. However, this portrays a serious risk to the heart and also indicates lipolysis promotion activity of the drug which leads to increase in the lipid profile parameters. Increases in lipid profile parameters have been linked to heart diseases [38]. The reduction of the raised levels of total cholesterol, triglycerides, VLDL, LDL and HDL in this study reveals a strong hypolipidemic activity of the seed extract and fractions perhaps due to inhibitory activity on lipolysis which is due to the activities of its phytoconstituents. This further confirms the cardioprotective potentials of the seed extract and fractions

This effect can be attributed to the antioxidant activities of the phytochemical constituents of the seed extract previously reported in this seed extract which have been documented to exert antioxidant activities [8].

5. Conclusion

The findings of this study show that the seed extract and fractions of *Telfairia occidentalis* have the potentials to counteract the injurious effect of doxorubicin on the heart. This activity can be attributed to the antioxidant and antioxidative stress activities of its phytochemical constituents. Thus, the seed can be use to alleviate and/or prevent doxorubicin-induced cardiotoxicity.

Compliance with ethical standards

Acknowledgments

This study was privately funded by the authors and no special financial support was obtained from any funding agency in the public, commercial, or not-for-profit sectors.

Disclosure of conflict of interest

All the authors hereby declare that no conflicts of interest exist related to this article.

Statement of ethical approval

This study was approved and ethically cleared by College of Health Sciences Animal Ethics Committee, University of Uyo (UU/CHS/IHREC/2025/VOL.1/16).

References

- [1] El-Sayed EM, Abd El-azeem A S, Afify AA, Shabana MH, Ahmed H.H. Cardioprotective effects of *Curcuma longa* L. extracts against doxorubicin-induced cardiotoxicity in rats. *Journal of Medicinal Plants Research*. 2011;5(17): 4049–4058.
- [2] Ibrahim MA, Morsy M A, Hafez H M, Gomaa W M, Abdelrahman A M. Effect of selective and non-selective cyclooxygenase inhibitors on doxorubicin-induced cardiotoxicity and nephrotoxicity in rats. *Toxicology Mechanisms and Methods* 2012;22(6): 424–431. <https://doi.org/10.3109/15376516.2012.666658>
- [3] Abdel-Daim MM, kilany OE, Khalifa HA, Ahmed AA M. Allicin ameliorates doxorubicin-induced cardiotoxicity in rats via suppression of oxidative stress, inflammation and apoptosis. *Cancer Chemotherapy and Pharmacology*. 2017;80(4): 745–753. <https://doi.org/10.1007/s00280-017-3413-7>.
- [4] .Abushouk AI, Salem AMA, Saad A. Mesenchymal stem cell therapy for doxorubicin-induced cardiomyopathy: potential mechanisms, governing factors, and implications of the heart stem cell debate. *Frontiers in Pharmacology*, 2019;10: 635 . <https://doi.org/10.3389/fphar.2019.00635>.
- [5] Okokon JE, Ekpo AJ , Eseyin OA. Evaluation of in vivo antimalarial activities of ethanolic leaf and seed extracts of *Telfairia occidentalis*. *Journal of Medicinal Food*, 2009;12(3):649-653. <https://doi.org/10.1089/jmf.2008.0099>.
- [6] Usunobun U, Okpiabhele A. *Telfairia occidentalis* Hook f. mitigates Carbon tetrachloride induced nephrotoxicity in Rat. *Journal of Research in Applied and Basic Medical Sciences* 2023;9 (3) :130-137. <https://doi.org/10.61186/rabms.9.3.130>
- [7] Eseyin OA, Ebong P, Ekpo A, Igboasoyi A, Oforah E. Hypoglycemic effect of the seed extract of *Telfairia occidentalis* in rat. *Pakistan Journal of Biological Sciences* 2007; 10(3): 498- 501.
- [8] Okokon JE, Antia BS, Dar A., Choudhary MI. Immunomodulatory, anticancer and antiinflammatory activities of *Telfairia occidentalis* seed extract and fractions. *International Journal of Food Nutrition and Safety* 2012a;2(2): 72 - 85.
- [9] Osukoya OA, Adegbenro D, Onikanni SA, Ojo OA, Onasanya A. Antinociceptive and antioxidant activities of the methanolic extract of *Telfairia occidentalis* Seeds. *Anc Science of Life*. 2016; 36(2):98-103. https://doi.org/10.4103/asl.ASL_142_16
- [10] Okokon JE, Dar A, Choudhary MI. Chemical constituents and analgesic activity of *Telfairia occidentalis*. *Phytopharmacology* 2012b; 3(2): 359 – 366.
- [11] Magnus SP, Anagboso MO, Johnny II, Ise UP, Okokon JE. Evaluation of genotoxic and cytotoxic activities of leaf and seed extracts of *Telfairia occidentalis*. *Journal of Complementary and Alternative Medicine Research*. 2024; 25(3):7-16. <https://doi.org/10.9734/JOCAMR/2024/v25i3521>
- [12] Enin GN, Okokon JE, Odokwo BO, Antia BS. Preliminary phytochemical screening and in vivo Inhibitory study of *Telfairia occidentalis* Hook f. seeds extract on alpha amylase and alpha glucosidase of rats. *Journal of Science and Technology Research* 2023;5(4) : 26-35. <http://dx.doi.org/10.5281/zenodo.10425980>

- [13] Oboh G, Nwanna E, Elusiyan C. Antioxidant and antimicrobial properties of *Telfairia occidentalis* (Fluted pumpkin) leaf extracts. *Journal of Pharmacology and Toxicology* 2010;5(8): 539-547. <https://doi.org/10.3923/jpt.2006.167.175>
- [14] Nwanna E, Oboh G. Antioxidant and hepatoprotective properties of polyphenol extracts from *Telfairia occidentalis* (Fluted Pumpkin) leaves on acetaminophen induced liver damage. *Pakistan Journal of Biological Sciences* 2007;10(16): 2682-2687. <https://doi.org/10.3923/pjbs.2007.2682.2687>.
- [15] Nwozo SO, Adaramoye OA, Ajaiyeoba EO. Anti-diabetic and hypolipidemic studies of *Telfairia occidentalis* on alloxan induced diabetic rabbits. *Nigerian Journal of Natural Products and Medicine* 2004; 8: 45-47. <https://doi.org/10.4314/njnpm.v8i1.11814>
- [16] Fabian UA, Anagboso MO, Samuel AE, Okokon JE. Effect of seed extract and fractions of *Telfairia occidentalis* on liver and kidney functions and histologies of rats with testosterone-induced benign prostatic hyperplasia in rats. *Journal of Complementary and Alternative Medical Research*. 2025;26(6):1-18. <https://doi.org/10.9734/jocamr/2025/v26i6661>
- [17] Ebong AS, Eseyin OA, Etim EI, Okokon JE. *Telfairia occidentalis* potentiates antiplasmodial activity of artemisinin and amodiaquine combination therapy. *Anti-Infective Agents*. 2020;18(2): 152-159. <https://doi.org/10.2174/2211352517666190206160812>
- [18] Olorundare OE, Adeneye AA, Akinsola AO, Sanni DA, Koketsu M, Mukhtar H. *Clerodendrum volubile* ethanol leaf extract: a potential antidote to doxorubicin-induced cardiotoxicity in rats. *Journal of Toxicology*, 2020; Volume 2020, Article ID 8859716, 17 pages. doi: 10.1155/2020/9535426
- [19] Tietz WW. *Clinical Guide to Laboratory tests*. 2nd edn. Sanders Company. Philadelphia, PA. 1995, Pp. 554-556.
- [20] Friedewald WT, Levy RI, Fredrickson DS. Estimation of the concentration of low density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. *Clin Chem.*, 1972;18(6): 499-502. PMID: 4337382
- [21] Marklund S, Marklund G. Involvement of superoxide anion radical in the autooxidation of pyrogallol and a convenient assay for superoxide dismutase. *European Journal of Biochemistry*, 1974;47: 469 - 474. <https://doi.org/10.1111/j.1432-1033.1974.tb03714.x>
- [22] Sinha KA. Colorimetric assay of catalase. *Analytical Biochemistry*. 1972;47: 389-394. [https://doi.org/10.1016/0003-2697\(72\)90132-7](https://doi.org/10.1016/0003-2697(72)90132-7)
- [23] Lawrence RA, Burk RF. Glutathione peroxidase activity in selenium- deficient rat liver. *Biochemistry and Biophysic Research Communication* 1976;71: 952-958. [http://dx.doi.org/10.1016/0006-291x\(76\)90747-6](http://dx.doi.org/10.1016/0006-291x(76)90747-6)
- [24] Ellman GL. Tissue sulfhydryl groups. *Archives of Biochemistry and Biophysics*. 1959;82: 70-77. [https://doi.org/10.1016/0003-9861\(59\)90090-6](https://doi.org/10.1016/0003-9861(59)90090-6).
- [25] Esterbauer H, Cheeseman KH. Determination of aldehydic lipid peroxidation products: malonaldehyde and 4-hydroxynonenal. *Methods in Enzymology*. 1990;186: 407-421. [http://dx.doi.org/10.1016/0076-6879\(90\)86134-h](http://dx.doi.org/10.1016/0076-6879(90)86134-h).
- [26] Drury RA, Wallington EA. *Carleton's Histological Techniques*. 5th Edition, Oxford University Press, New York, 1980, 195.
- [27] Calabresi P, Chabner BA. Chemotherapy of neoplastic diseases, In: Gilman, AG, Rall TW, Nies AS, Taylor P. (eds.) *The Pharmacological Basis of Therapeutics*. NY: Pergamon Press Inc. 1990, pp. 1203-1263.
- [28] Abushouk AI, Ismail A, Salem AMA, Afifi AM, Abdel-Daim MM. Cardioprotective mechanisms of phytochemicals against doxorubicin-induced cardiotoxicity. *Biomedicine & Pharmacotherapy* 2017;90: 935-946. <https://doi.org/10.1016/j.biopha.2017.04.033>.
- [29] Herman E, Ferrans VJ, Young RS, Hamlin RL. Effect of pretreatment with ICRF-187 on the total cumulative dose of doxorubicin tolerated by beagle dogs. *Cancer Research*, 1988;48 (23): 6918-6925. PMID: 3141049
- [30] Bertinchant JP, Polge A, Juan JM. Evaluation of cardiac troponin I and T levels as markers of myocardial damage in doxorubicin-induced cardiomyopathy rats, and their relationship with echocardiographic and histological findings. *Clinica Chimica Acta*, 2003;329(1-2): 39-51. [https://doi.org/10.1016/s0009-8981\(03\)00013-5](https://doi.org/10.1016/s0009-8981(03)00013-5).
- [31] Deschenes G, Doucet A. Collecting duct (Na⁺/K⁺)-ATPase activity is correlated with urinary sodium excretion in rat nephrotic syndromes. *Journal of the American Society of Nephrology*. 2000;11(4): 604-615. <https://doi.org/10.1681/ASN.V114604>.

- [32] Injac R, Perse M, Cerne M, Potocnik N, Radic N, Govedarica B, Djordjevic A, Cerar A, Strukelj B. (2009). Protective effects of fullerol C₆₀(OH)₂₄ against doxorubicin-induced cardiotoxicity and hepatotoxicity in rats with colorectal cancer. *Biomaterials* 2009; 30: 1184-1196.[https://doi.org/ 10.1016/j.biomaterials.2008.10.060](https://doi.org/10.1016/j.biomaterials.2008.10.060).
- [33] Kalender Y, Yel M, Kalender S. Doxorubicin hepatotoxicity and hepatic free radical metabolism in rats: The effects of vitamin E and catechin. *Toxicology* 2005;209: 39-45. [https://doi.org/ 10.1016/j.tox.2004.12.003](https://doi.org/10.1016/j.tox.2004.12.003)
- [34] Rashid S, Ali N, Nafees S. Alleviation of doxorubicin-induced nephrotoxicity and hepatotoxicity by chrysin in Wistar rats. *Toxicology Mechanisms and Methods*, 2013;23 (5): 337–345.[https://doi.org/ 10.3109/15376516.2012.759306](https://doi.org/10.3109/15376516.2012.759306).
- [35] Rehman MU, Tahir M, Khan AQ, Khan R, Oday OH, Lateef A, Hassan SK, Rashid S, Ali N, Zeeshan M, Sultana S. D-limonene suppresses doxorubicin-induced oxidative stress and inflammation via repression of COX-2, iNOS, and NF- κ B in kidneys of Wistar rats. *Exp. Biol. Med.* (Maywood) 2014; 239:465–476.[https://doi.org/ 10.1177/1535370213520112](https://doi.org/10.1177/1535370213520112).
- [36] Khames A, Khalaf MM, Gad AM, Abd El-Raouf OM, Kandeil MA. Nicorandil combats doxorubicin-induced nephrotoxicity via amendment of TLR4/P38 MAPK/NF- κ B signaling pathway. *Chem. Biol. Interact.*,2019;311: 108777. <https://doi.org/10.1016/j.cbi.2019.108777>
- [37] Okokon JE, Noah K, Ekeleme CM, Udoh IE, Asanga EE, Anagboso MA, Inyang AN. "Integrative investigation on *Hippocratea africana* root: Insights from cardio-protective, anti-oxidative stress activities, isolation, GC-MS, and pharmacological significance profiling. *BMC Complementary Medicine and Therapies*.2025; 25(2025): 200.[https://doi.org/: 10.1186/s12906-025-04941-8](https://doi.org/10.1186/s12906-025-04941-8).
- [38] Abbott RD, Wilson PW, Kannel WB, Castelli WP. High density lipoprotein cholesterol, total cholesterol screening, and myocardial infarction. The Framingham study. *Arteriosclerosis*: 1988; 8(3): 207–211.[https://doi.org/ 10.1161/01.atv.8.3.207](https://doi.org/10.1161/01.atv.8.3.207).