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Original Article
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Cardioprotective Effect of *Peperomia pellucida* against Doxorubicin-Induced Cardiotoxicity in Wistar Rats via Modulation of Electrocardiographic and Cardiac Biomarkers

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Objectives: This study assessed the electrocardiographic pattern and cardiac inflammatory response of doxorubicin-induced myocardial injury in Wistar rats treated with *Peperomia pellucida* ethanol extract.
Methods: Female Wistar rats (190-200 g) were assigned into five groups of seven rats each. The Group 1 (Control group) was given rat chow and drinking water while the Group 2 (doxorubicin group) received intraperitoneal administration of doxorubicin (2 mg/kg) once weekly for three weeks. The Group 3 (*Peperomia pellucida* group) received 200 mg/kg of ethanolic extract of *Peperomia pellucida* daily. Group 4 (Doxorubicin + *P. pellucida* group) received doxorubicin in addition to *Peperomia pellucida.* Group 5 (Captopril (50 mg/kg) was administered to another group in addition to *P. pellucida* while the doxorubicin + captopril group was administered captopril in addition to doxorubicin. Electrical recording and cardiac markers were evaluated.
Results: The results revealed a significant ($p < 0.01$) elevation of T-wave and altered electrocardiographic parameters in the doxorubicin group than the control, *P. pellucida*, and other experimental groups. The heart rate, cardiac troponin level, lactate dehydrogenase, creatine kinase, angiotensin-converting enzyme activities, and inflammatory biomarkers were significantly ($p < 0.01$) higher while nitric oxide level was significantly ($p < 0.05$) reduced in the doxorubicin-only group compared to the control. Cardiac cell hypertrophy and inflammatory cell infiltration were observed due to doxorubicin administration. Treatment with *P. pellucida* extract and captopril reversed these trends and improved the antioxidants and inflammatory activities.
Conclusion: *Peperomia pellucida* extract improves electrocardiographic pattern, has cardioprotective ability, and prevents doxorubicin-induced myocardial injury probably due to its phytochemical constituents and anti-inflammatory properties.

Keywords: captopril, cardiac troponin, cardiotoxicity, electrocardiograph, inflammation, *Peperomia pellucida*

INTRODUCTION

Coronary heart disease is one of the leading causes of morbidity and mortality worldwide, with myocardial infarction (MI) as its most common manifestation [1]. MI, a life-threatening condition, is marked by sudden cardiac death [2]. The annual mortality rate due to MI is estimated to exceed 15% [3]. MI results from decreased or interrupted blood flow to parts of the heart, leading to damage to the cardiac muscle [4]. Early symptoms include chest pain, nausea, shortness of breath, fatigue,

and abnormalities in heart rate and blood pressure [5]. Major risk factors for MI include advanced age, hypertension, diabetes mellitus, elevated total cholesterol, and high low-density lipoprotein (LDL) levels [6].

In MI assessment, electrocardiography (ECG) and cardiac inflammatory biomarkers are routinely used to determine severity. Heart injury typically results in ECG pattern alterations and increased plasma levels of cardiac troponin (cTn), which indicate myocardial damage [7, 8]. Cardiac troponin has three isoforms—cTnC, cTnT, and cTnI—each involved in muscle contraction regulation within the thin filament. Troponin is released into the bloodstream due to cell turnover, myocyte apoptosis, necrosis, reversible injury, increased cell membrane permeability, and the release of degradation products [8, 9].

C-reactive protein (CRP), an inflammation marker, is a valuable prognostic indicator in patients with acute coronary syndrome, where elevated levels predict outcomes such as cardiac death, MI, and congestive heart failure [10]. While multiple factors contribute to MI, drug-induced MIis particularly notable for its detrimental effects on cardiac function. Doxorubicin (DOX), a widely used chemotherapeutic drug, has been reported to cause adverse cardiac reactions, including toxicity [11]. This toxicity is dose-dependent, with higher doses increasing the risk of myocardial injury and congestive heart failure [12]. Mechanisms of DOX-induced cardiotoxicity include increased oxidative stress, gene alteration, and myocardial cell apoptosis [13].

In many developing nations, plants are commonly used as medicinal sources to manage various ailments. *Peperomia pellucida*, a member of the *Piperaceae* family, is rich in phytochemicals and widely recognized for its therapeutic properties. Known locally as Pepper elder, silverbush, rat ear, little heart, and *Ewe rinrin* by Nigeria's Yoruba tribe, this plant is used to treat ailments such as colds, fever, infections, and rheumatic pain [14]. It is also used in managing cataracts and conjunctivitis across Africa, America, and Asia, further highlighting its global medicinal relevance [15]. The plant is abundant in phytochemicals, including saponins, alkaloids, flavonoids, triterpenoids, tannins, cardiac glycosides, phytosterols, phenolics, and essential oils, which exhibit pharmacological and biological activities [14]. Additionally, *P. pellucida* has anti-inflammatory, antioxidant, hypotensive, and antidiabetic properties [16-18]. However, its use in mitigating DOX-induced myocardial injury remains undocumented. This study aimed to evaluate the potential cardioprotective effects of *P. pellucida* ethanolic extract on the electrical activity of the heart and inflammatory markers in a DOX-induced myocardial injury model using Wistar rats.

MATERIALS AND METHODS

1. Chemicals and diagnostic kits

Captopril was obtained from Crescent Pharma Limited (Overton, Hampshire, UK), while DOX was purchased from Naprod Life Sciences Pvt. Ltd. (Boisar, Thane, India). Ketamine was sourced from Sigma-Aldrich (St. Louis, MO, USA). The reagent kits, including cardiac troponin T, angiotensin-converting enzyme (ACE), and lactate dehydrogenase (LDH) ELISA kits, were obtained from Cusabio Biotech Co. (China). Captopril and DOX solutions were freshly prepared each day by dissolving the drugs in distilled water before administration to the animals.

2. Plant material and extraction

The entire *P. pellucida* plant was harvested from the University of Calabar farm and identified by a botanist in the Department of Plant and Ecological Studies. After washing and drying, the plant was ground into a powder for extraction. Using the maceration method, the powdered plant material was combined with 80% ethanol as a solvent and left at room temperature for 48 hours with periodic agitation [19]. The mixture was then filtered into a clean beaker, evaporated using a rotary evaporator, and dried in an oven at 45℃. The dried crude plant extract was stored in a sealed, clean container until use.

3. Phytochemical analysis using gas chromatography-mass spectrometry (GC–MS)

Phytochemical analysis was conducted on the plant extract using GC–MS on a Varian 3900, Saturn 2100T model. The sample was vaporized in the gas chromatograph, and a capillary column with a stationary liquid phase separated its components. The compounds were ionized and fragmented upon exiting the column, with ions separated by their mass-to-charge (m/z) ratios. The resulting peaks, corresponding to each compound's concentration, generated unique mass spectra for compound identification.

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4. Experimental animal handling and design

Female Wistar rats weighing 190-200 g were obtained from the animal facility at the Faculty of Basic Medical Sciences, University of Calabar, Nigeria. Rats were housed under standard laboratory conditions (temperature 28 ± 1℃, 12-hour light/dark cycle) and provided with a standard rat pellet diet and water. The rats were randomly divided into five groups, each consisting of seven rats:

- Group 1 (Control): Only standard chow and water are given.
- Group 2 (DOX-only): Received an intraperitoneal dose of DOX (2 mg/kg) once weekly for three weeks as per established methods [20].
- Group 3 (PP-only): Received *P. pellucida* ethanolic extract (200 mg/kg) daily for 21 days [21].
- Group 4 (DOX + PP): Treated with both DOX and *P. pellucida* as in Groups 2 and 3.
- Group 5 (DOX + CAPT): Captopril (50 mg/kg) was administered orally once daily for three weeks [22], alongside DOX, as in Group 2.

Doses of *P. pellucida* and captopril were selected based on prior studies demonstrating their beneficial effects at these levels [21, 22]. Ethical approval for this study was granted by the Faculty of Basic Medical Sciences Animal Research Ethics Committee, University of Calabar, with approval number 272PHY2324.

5. Electrocardiograph (ECG) recording

Heart electrical activity was recorded 24 hours after the final administration of the plant extract or captopril for each rat in all groups. Each animal was anesthetized with ketamine (80 mg/kg) and positioned on a prepared board. Electrodes (Gold cup electrodes; Grass Instrument Company) were attached to the forelimbs and left hind limb, forming an "Einthoven Triangle" configuration. The electrodes were connected to an ECG monitoring device (Heart and Brain SpikerBox; Backyard Brains, Ann Arbor, MI) and linked via USB to Spike Recorder software. ECG signals were captured from limb lead II, with data logged as sound (wave) files, and each recording session lasted 1-2 minutes.

6. Animal sacrifice and sample collection

Following the 21-day treatment period, rats were anesthetized with ketamine (80 mg/kg). A midline incision exposed the heart, allowing for blood collection via cardiac puncture. Blood samples were left to clot, then centrifuged at 200 g for 10 minutes, with serum separated and stored at −20℃ for later biochemical analysis. The heart was carefully excised, cleared of connective tissue, and homogenized in ice-cold phosphate-buffered saline (pH 7.4) to create a 10% homogenate. The homogenate was centrifuged at 15,000 rpm for 10 minutes, and the supernatant was stored at −20℃ until needed.

7. Biochemical parameter analysis

Cardiac troponin T (cTnT) levels were measured using an enzyme-linked immunosorbent assay (ELISA) Cusabio test kit following the manufacturer's instructions. ACE activity and lactate dehydrogenase levels were assessed using respective ELISA kits, and CRP levels were measured via a competitive enzyme immunoassay (Biosystems S.A., Spain). Nitric oxide (NO) levels were analyzed using the Griess reagent colorimetric assay kit. Tumor necrosis factor-alpha (TNF-α) and interleukin-6 levels were determined with Cusabio ELISA kits according to the manufacturer's protocol.

8. Heart histopathology

Heart tissue was dissected, fixed in 10% buffered formalin, and processed by standard protocols. Tissues were dehydrated in graded ethanol solutions (70-100%), cleared in xylene, and embedded in paraffin wax. Thin sections (5 mm thick) were cut, stained with hematoxylin and eosin (H&E), and examined under a light microscope. Images were captured using a Leica DM750 photomicroscope (Leica Microsystems, Heerbrugg, Switzerland).

9. Statistical analysis

Data were presented as mean ± standard error of the mean (SEM) and analyzed using one-way analysis of variance (ANOVA) with GraphPad Prism software (version 9.02, GraphPad Software, San Diego, CA, USA). Tukey's post-test was applied for group comparisons, with statistical significance set at $p < 0.05$.

RESULTS

1. GC–MS analysis of *P. pellucida*

The GC–MS analysis identified and quantified the active compounds in *Peperomia pellucida*. Fig. 1 displays the GC–MS profile of the ethanol extract, while Table 1 lists the active compounds present. Peaks 1 to 4 in Fig. 1 represent butylated hydroxytoluene, an antioxidant from the phenol class, with an abundance of 20.92%. Peaks 5 to 6 indicate hexadecanoic acid methyl and ethyl esters, while peaks 7 to 10, 12, 15, 20, 21, 23, and 27 to 30 correspond to 9,12-Octadecadienoic acid methyl esters, which are fatty acid esters. Hexadecanoic acid methyl ester and 9,12-Octadecadienoic acid methyl ester (Z, Z) accounted for 2.90% and 34.85%, respectively, comprising a combined abundance of 37.75%. Peaks 13, 14, and 24 to 26 indicate gamma-sitosterol (35.85%) and beta-sitosterol (1.15%). Cyclopropane octanal, with a relative abundance of 2.24%, appears in peaks 11, 17, 18, and 19. Additional compounds include oleic acid (peaks 22 and 28), dodecatrienol (peak 16), and 7-pentadecyne (peak 31), contributing to a total abundance of 2.11%.

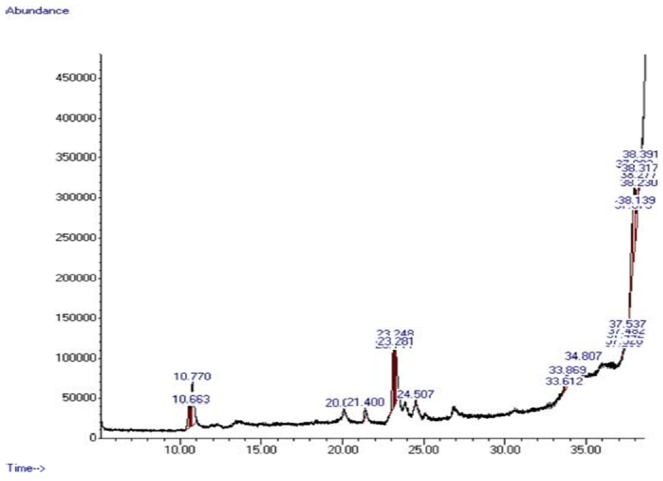

Figure 1. GC-MS analysis of *Peperomia pellucida* ethanolic extract.

2. ECG tracings and parameters

Fig. 2 shows ECG readings for the control group (Group 1), DOX-only group (Group 2), PP-only group (Group 3), DOX + PP group (Group 4), and DOX + CAPT group (Group 5). The ECG results reveal a normal, regular pattern in the control group, with distinct alterations observed in the DOX-treated groups. Treatment with *P. pellucida* and captopril appeared to restore ECG tracing closer to normal. In the DOX-only group, abnormal R and T waves and an elevated ST segment were noted, contrasting with the control pattern.

The heart rate in the DOX group (440 ± 20 beats/min) was significantly elevated ($p < 0.05$) compared to the control group (340 ± 20 beats/min), PP group (350 ± 3.3 beats/min), DOX + PP group (357 ± 5 beats/min), and DOX + CAPT group (344 ± 13 beats/min). Heart rate values in the PP, DOX + PP, and DOX + CAPT groups were comparable to the control group (Fig. 3A).

The duration of the ST segment was significantly increased in the DOX group (24.4 ± 1.4 msec) relative to the control group (11.1 ± 1.2 msec) and PP-only group (13 ± 1 msec). However, treatment with *P. pellucida* significantly reduced the ST segment duration in the DOX + PP group (16 ± 2 msec) and DOX + CAPT group (14 ± 2 msec) compared to the DOX-only group, with these values approximating control levels (Fig. 3B).

A significant ST-segment elevation (1.3 ± 0.01 mV) was observed in the DOX-only group, compared to the control (0.5 ± 0.02 mV), PP (0.4 ± 0.03 mV), DOX + PP (0.7 ± 0.03 mV), and DOX + CAPT (0.6 ± 0.03 mV) groups. However, treatment with *P. pellucida* or captopril significantly reduced ST segment

Table 1. Phytochemical analysis of *Peperomia pellucida* ethanol extract

S/N	Compound	Molecular formula and weight	Composition (%)
1	Butylated hydroxytoluene	$C_{15}H_{24}O$; 220.35	20.92
2	Hexadecenoic acid, methyl ester	$C_{17}H_{34}O_2$; 270.4507	2.90
3	9,12-Octadecadienoic acid (Z, Z), methyl ester	$C_{19}H_{34}O_2$; 294.4721	34.85
4	Beta sitosterol; gamma-sitosterol	$C_{29}H_{50}O$; 414.7	1.15
5	Gamma-sitostenone	$C_{29}H_{48}O$; 412.69	35.85
6	Cyclopropane octanal	$C_{19}H_{36}O$; 280.49	2.24
7	Oleic acids Dodecatrienol 7-pentadecyne	$C_{18}H_{34}O_2$; 282.5 $C_{12}H_{20}O$; 180.29 $C_{15}H_{28}$; 208.38	2.11

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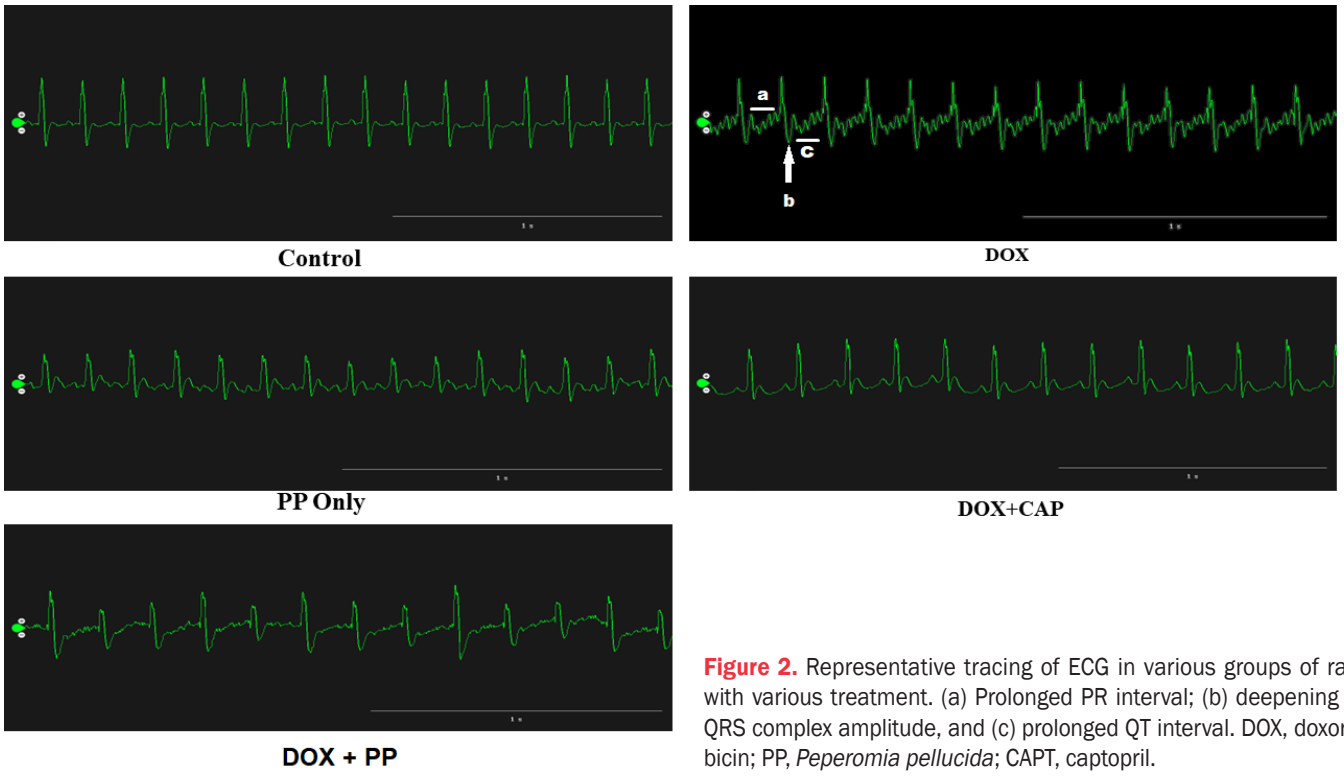

Figure 2. Representative tracing of ECG in various groups of rats with various treatment. (a) Prolonged PR interval; (b) deepening of QRS complex amplitude, and (c) prolonged QT interval. DOX, doxorubicin; PP, *Peperomia pellucida*; CAPT, captopril.

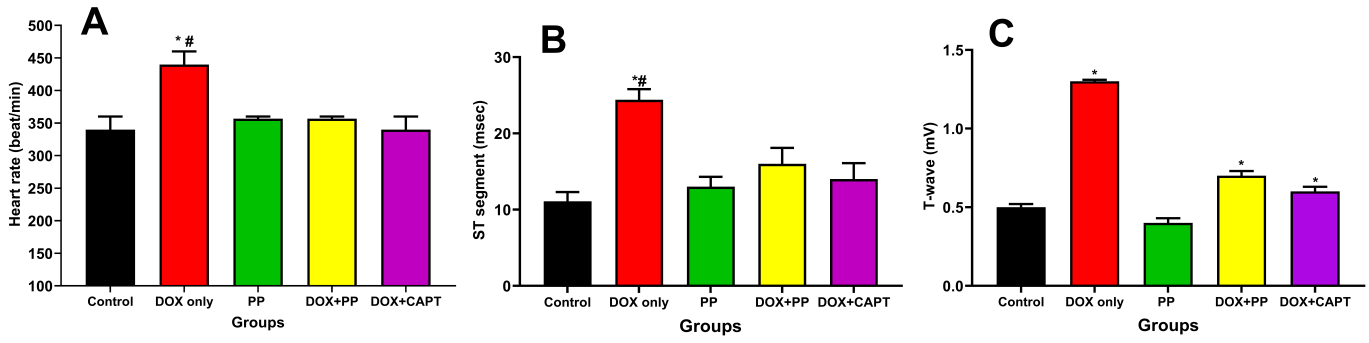

Figure 3. ECG parameters in control and myocardial infarction-induced rats treated with *Peperomia pellucida* extract and captopril. (A) Heart rate, (B) ST segment, (C) T-wave. *p < 0.05 compare with the control group, #p < 0.05 compared with DOX + PP and DOX + CAPT groups. DOX, doxorubicin; PP, *Peperomia pellucida*; CAPT, captopril.

elevation compared to the DOX-only group, yielding values comparable to the control group (Fig. 3C).

Table 2 presents the P-R interval for each experimental group, indicating the time required for impulse conduction from the atria through the atrioventricular node and Bundle of His to the ventricles. The DOX-only group exhibited a significant increase in the P-R interval (66 ± 1.1 msec) compared to the control (50 ± 0.43 msec), PP (53 ± 2.6 msec), DOX + PP (56 ± 2.6 msec), and DOX + CAPT (54 ± 3.2 msec) groups. Administration of *P. pellucida* or captopril significantly reduced the P-R interval relative to the DOX-only group, though these values remained slightly elevated compared to the control.

For the QRS complex, values in the DOX-only group (39 ± 2.2 msec) were comparable to the control (42 ± 1.4 msec) and PP (37 ± 1.7 msec) groups. In the DOX + PP (51 ± 3.4 msec) and DOX + CAPT (51 ± 4.8 msec) groups, the QRS complex was significantly increased compared to the DOX-only group. The QT interval was notably prolonged in the DOX-only group (170 ± 2 msec) compared to the control (110 ± 4 msec), PP (113 ± 5 msec), DOX + PP (118 ± 4 msec), and DOX + CAPT (115 ±

Table 2. Effect of *Peperomia pellucida* extract on ECG parameters in doxorubicin-induced cardiac toxicity

Parameters	Control	DOX	PP	DOX + PP	DOX + CAPT
P-R interval (ms)	50 ± 0.43	66 ± 1.1*,#	53 ± 2.6	56 ± 2.6	54 ± 3.2
QRS complex (ms)	42 ± 1.4	38 ± 2.2*,#	44 ± 1.7	51 ± 3.4	51 ± 4.8
QT-interval (ms)	110 ± 4	170 ± 2*,#	113 ± 5	118 ± 4	115 ± 3
R-R interval (ms)	0.15 ± 0.01	0.13 ± 0.01*,#	0.16 ± 0.02	0.16 ± 0.03	0.15 ± 0.05

*p < 0.05 compared with control; #p < 0.05 compared with DOX only group.
CAPT, captopril; DOX, doxorubicin; PP, *Peperomia pellucida*.

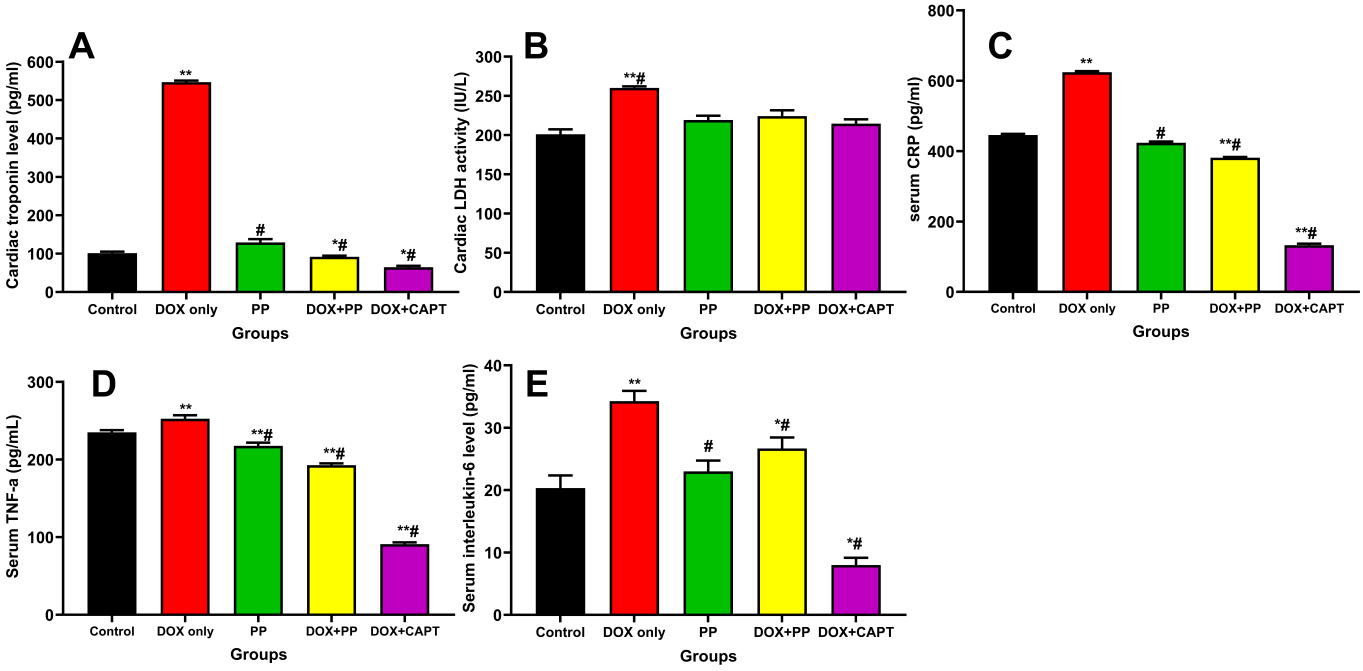

Figure 4. Cardiac inflammatory biomarkers in control and cardiotoxic groups treated with *Peperomia Pellucida* extract and captopril. (A) Cardiac troponin, (B) Cardiac lactate dehydrogenase, (C) C-Reactive protein, (D) Tumor necrosis factor-alpha, (E) Interleukin-6. *p < 0.05 vs. control group; **p < 0.01 compared with control; #p < 0.05 compared with DOX + CAPT. DOX, doxorubicin; CAPT, captopril.

3 msec) groups, with captopril treatment significantly reducing the QT interval relative to the DOX + PP group. The R-R interval (msec) in the DOX-only group (0.13 ± 0.01) was significantly decreased compared to the control (0.15 ± 0.01), PP (0.16 ± 0.02), DOX + PP (0.16 ± 0.03), and DOX + CAPT (0.15 ± 0.05) groups, though these values were comparable to the control.

3. Cardiac inflammatory biomarkers

The cTnT level (pg/mL) was significantly increased (p < 0.05) in the DOX-only group (547 ± 3.8) relative to the control (101 ± 4), PP (129 ± 9), DOX + PP (92 ± 3), and DOX + CAPT (65 ± 4) groups. Treatment with *P. pellucida* extract or captopril significantly reduced cTnT levels to control levels (Fig. 4A). The LDH enzyme activity (IU/L) was significantly elevated (p < 0.01) in the DOX-only group (250 ± 8) compared to the control (200 ± 17), PP (218 ± 5), DOX + PP (223 ± 13), and DOX + CAPT (212 ± 9) groups. However, in groups treated with *P. pellucida* or captopril, LDH activity was comparable to the control group (Fig. 4B).

Serum CRP levels (pg/mL) were significantly elevated in the DOX-only group (624 ± 3.2) compared to the control (446 ± 3), PP (424 ± 3.5), DOX + PP (381 ± 1.8), and DOX + CAPT (133 ± 4) groups. *P. pellucida* extract and captopril administration significantly reduced CRP levels (Fig. 4C).

The tumor necrosis factor-alpha (TNF-α) level (pg/mL) was significantly elevated (p < 0.01) in the DOX-only group (253 ± 4.3) relative to the control (242 ± 5.9), PP (253 ± 4.3), DOX +

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PP (193 ± 2.3), and DOX + CAPT (91 ± 2.3) groups. Treatment with *P. pellucida* or captopril significantly reduced TNF-α levels in the experimental groups (Fig. 4D).

Interleukin-6 (IL-6) levels (pg/mL) in the DOX-only group (34 ± 1.7) were significantly elevated compared to the control (14 ± 2.9). In the PP (24 ± 3.8), DOX + PP (27 ± 1.8), and DOX + CAPT (8 ± 1.2) groups, captopril administration significantly reduced IL-6 levels compared to the DOX + PP group, with further reductions noted in the *Peperomia pellucida*-treated groups (Fig. 4E).

4. Hypertension markers

The mean NO levels (ng/mL) are presented in Fig. 5A. NO levels in the DOX-only (49 ± 3) and PP-only (71 ± 2.9) groups were significantly decreased compared to the control group. However, NO levels in the DOX + PP (106 ± 2.3) and DOX + CAPT (128 ± 2.9) groups were significantly elevated relative

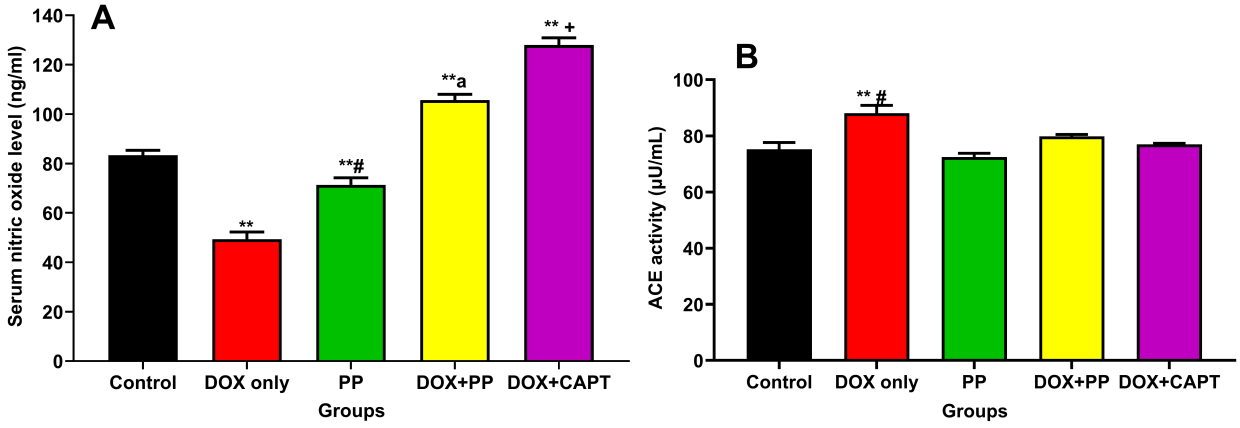

Figure 5. Hypertension biomarkers in control and cardiotoxic groups treated with *Peperomia Pellucida* extract and captopril. (A) Nitric oxide, (B) Angiotensin Converting Enzyme. **p < 0.01 compared with control group; #p < 0.01 with DOX + PP and DOX + CAPT groups; †p < 0.05 vs. DOX + PP; ªp < 0.05 vs. PP group. DOX, doxorubicin; PP, *Peperomia pellucida*; CAPT, captopril.

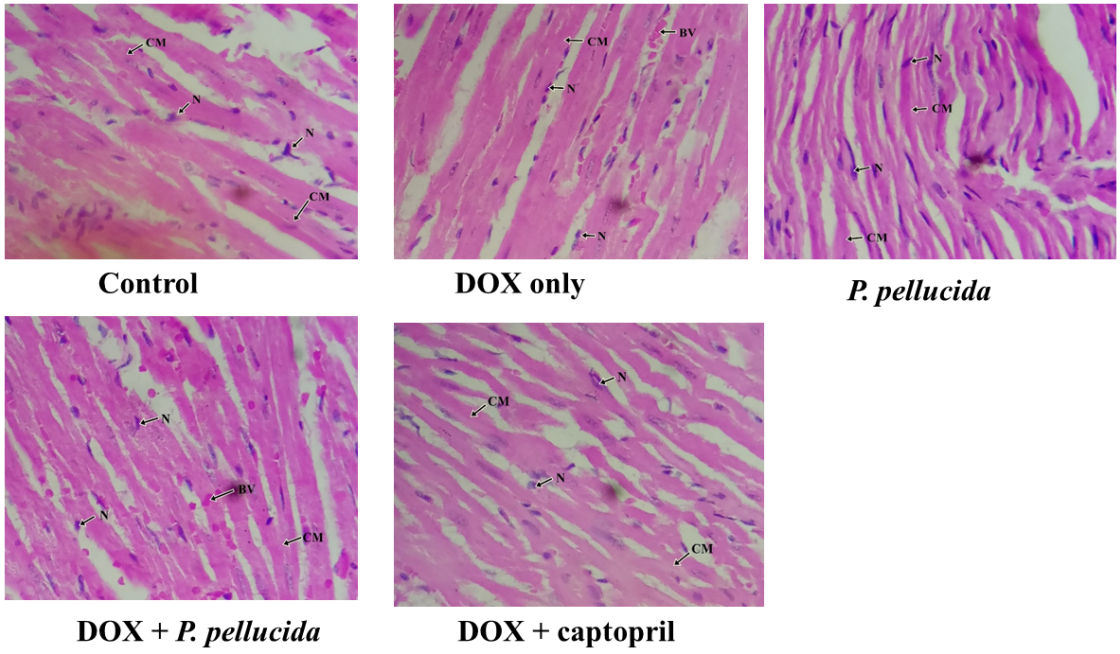

Figure 6. Photomicrographs of a cross-section of the cardiac muscles treated with *Peperomia pellucida* extract and captopril in doxorubicin-induced cardiac toxicity. H&E stain, ×400 magnification. CM, cardiac muscles fibers; N, nuclei; BV, blood vessels.

to the control, with the DOX-only group showing the lowest levels among all groups. ACE activity (IU/L) was significantly increased ($p < 0.01$) in the DOX-only group (88 ± 2.8) relative to the control (75 ± 2.4), PP (73 ± 1.3), DOX + PP (80 ± 0.6), and DOX + CAPT (77 ± 0.4) groups (Fig. 5B). However, ACE activity in the DOX + PP and DOX + CAPT groups was similar to control levels.

5. Histopathological changes in heart tissue

Histopathological analysis of DOX-treated animals revealed cardiac muscle cell hypertrophy with widespread interstitial fibrosis around the myocardium (Fig. 6). In contrast, treatment with *P. pellucida* or captopril reduced cardiac cell hypertrophy reversed myofibril disorientation, and decreased inflammatory cell presence, restoring cardiac muscle fiber structure without focal necrosis.

DISCUSSION

This study demonstrates that *P. pellucida* has cardioprotective and anti-inflammatory effects against DOX-induced myocardial injury, with effects comparable to those of captopril. The phytochemical analysis revealed active compounds within *Peperomia pellucida*, highlighting its rich content of phenolic compounds such as butylated hydroxytoluene, which has known antioxidant properties [23], and essential oils like hexadecanoic acid and 9,12-octadecadienoic acid, documented for their anti-inflammatory effects [24]. Additionally, beneficial compounds like testosterone, stigmasterol, gamma sitosterol, and oleic acid, which may play a role in cardiovascular health, were present. These findings suggest that the compounds within *P. pellucida* may contribute to mitigating the cardiotoxic effects associated with DOX toxicity.

This study examined multiple biomarkers to assess the impact of *P. pellucida* on DOX-induced cardiotoxicity. Indicators of myocardial injury, including alterations in cardiac electrical activity and biomarkers, were evaluated in this study. The results demonstrated that *P. pellucida* ethanolic extract alleviated markers of myocardial injury. Notably, myocardial injury is often characterized by disrupted electrical activity within the heart. The findings revealed that DOX administration led to ECG pattern alterations, such as irregularities in P-R, QT, QRS, and R-R intervals, elevated T waves, and prolonged ST segments—hallmarks of MI. These observations align with prior studies showing similar ECG changes in DOX-induced MI [25]. However, pretreatment with *P. pellucida* extract and captopril markedly reduced ST-segment elevation, suggesting a protective effect on cardiac muscle cells.

Additionally, a significant increase in heart rate was observed in the DOX-only group compared to the control, with *P. pellucida* and captopril treatments leading to heart rate reductions. This reduction aligns with previous findings where *P. pellucida* reduced heart rate in normotensive blood pressure models [18, 26]. This effect may stem from a vasodilatory mechanism associated with NO, a potent vasodilator that modulates blood pressure and heart rate, thus supporting cardiovascular health [27]. NO's blood pressure and heart rate reduction mechanisms include direct vasodilation and inhibition of sympathetic tone [28]. In the present study, *P. pellucida*-treated rats showed significantly increased NO concentrations compared to the DOX-only group, implying that DOX reduces NO, while *P. pellucida* reverses this effect. Increased NO levels may relate to the plant's phytochemicals, with previous studies showing flavonoids, a major constituent of *P. pellucida*, stimulate NO production [29]. Further, NO reduction may contribute to tachycardia through the modulation of autonomic control and direct effects on sinoatrial node function.

A range of cardiac biomarkers was examined in this study, including cardiac troponin, CRP, and LDH, which are commonly altered in MI. As reported in previous DOX administration studies, elevated cardiac troponin levels are a common indicator of acute MI [30]. Cardiovascular troponin levels were significantly reduced after interventions with *P. pellucida* and captopril. The ameliorative effect on cardiac troponin may stem from phytochemicals within *P. pellucida*, such as hexadecanoic acid methyl esters, which can relax vascular tone and regulate signaling pathways integral to cardiac health [31]. Additionally, beta-sitosterol, another compound in *P. pellucida*, is known to reduce troponin levels during MI [32]. The decline in troponin levels suggests reduced cardiac damage and restricted troponin release, as indicated by the observed reductions in the *P. pellucida* and captopril groups.

The level of CRP, an inflammatory biomarker, was elevated in the MI group relative to the control and other experimental groups, indicating an inflammatory response. However, both *P. pellucida* and captopril reduced CRP levels, implying anti-inflammatory effects. The CRP-lowering effect of *P. pellucida* may result from its phytochemicals, such as isoflavone genistein, which has been reported to inhibit CRP's inflammatory action

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in the heart [33]. As a marker of inflammation, elevated CRP can exacerbate inflammation and reduce NO levels, impairing blood flow and increasing inflammatory compounds [34]. This phenomenon could explain the decreased NO observed in the DOX-only group in this study.

LDH, an enzyme converting lactate to pyruvate, is another biomarker linked to acute MI, with increased activity indicating cardiac cell damage. In line with previous studies, the DOX-only group exhibited elevated LDH activity compared to the control [35]. In contrast, the groups treated with *P. pellucida* or captopril showed no significant difference in LDH activity relative to the control, suggesting protective effects against cardiac cell damage.

ACE, essential in the renin-angiotensin system, converts angiotensin I to angiotensin II, a potent vasoconstrictor. Our results indicated increased ACE activity in the DOX-only group, with reduced activity following *P. pellucida* and captopril treatments. This finding aligns with previous studies reporting elevated ACE activity in MI [36]. The reduction in ACE activity following *P. pellucida* and captopril treatment may be attributable to flavonoids and phenolic compounds within *P. pellucida* that inhibit ACE [37]. Additionally, *P. pellucida* is rich in octadecadienoic acid (Z, Z), a methyl ester reported to inhibit ACE activity, thus protecting the heart [38].

These results align with earlier research, indicating that *P. pellucida* may reduce inflammatory responses and protect cardiac function through antioxidant mechanisms, similar to captopril, a standard drug for myocardial injury. Captopril's cardioprotective effects in DOX-induced cardiotoxicity have been well-documented, including its ability to reduce biochemical parameters linked to cardiotoxicity [39]. This finding aligns with prior research by Saputri et al. [40], who reported that *P. pellucida* normalized cardiac function similarly to captopril by inhibiting ACE activity and reducing blood pressure. Such results suggest that *P. pellucida* may share a mechanism of action with captopril.

This study has several limitations. Only a single dose of the plant extract was used to measure its effect on myocardial toxicity. However, prior research has shown that this specific dose effectively lowers blood pressure in normotensive rats [18]. Future research should consider using a range of doses to better understand the dose-response relationship. Additionally, as this study utilized whole plant extract containing multiple active ingredients, it is challenging to attribute the cardioprotective effects to specific components. However, these ingredients likely act synergistically to provide observed cardiac benefits, indicating a need for future studies to investigate individual components. Molecular docking studies are also recommended to fully elucidate the mechanisms of action.

CONCLUSION

This study demonstrated that *P. pellucida* ethanol extract has significant cardioprotective potential in DOX-induced myocardial injury. The extract effectively improved ECG parameters, reduced cardiac inflammation, and moderated key biochemical markers associated with myocardial damage, showing effects comparable to captopril. The antioxidant and anti-inflammatory phytochemicals within *P. pellucida*, such as flavonoids and phenolics, likely contribute to these protective effects.

These findings support the ethnobotanical use of *P. pellucida* in cardiovascular management and suggest its potential as an adjunct therapy to reduce chemotherapy-induced cardiotoxicity. However, further studies are necessary to clarify its specific mechanisms and investigate its efficacy and safety in different dosing regimens.

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AUTHORS' CONTRIBUTIONS

Conceptualization: Daniel U. Owu; Methodology: Efiok A. Archibong, Investigation: Efiok A. Archibong, Glory A. Ikum, Stella C. Anaba; Supervision: Justin A. Beshel; Formal analysis: Idara A. Okon; Writing – Original draft: Efiok A. Archibong; Writing – Review & Editing: Idara A. Okon, Daniel U. Owu.

ETHICAL APPROVAL

This research was approved by the institutional animal care and use committee, Faculty of Basic Medical Sciences Animal Research Ethics Committee, University of Calabar, Calabar (272PHY2324, approval date 06/02/2024).

DATA AVAILABILITY

The data that support the findings of this study are available from the corresponding author upon reasonable request.

CONFLICTS OF INTEREST

The authors declare that they have no conflicts of interest.

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