Jurnal Kesehatan
Volume 17, Number 2, August 2026
ISSN 2086-7751 (*Print*), ISSN 2548-5695 (*Online*)
http://ejurnal.poltekkes-tjk.ac.id/index.php/JK

Protective Effects of *Pachyrhizus erosus* Tuber Extract on Oxidative Stress Markers in Doxorubicin-Induced Wistar Rats

Jimmy Hadi Widjaja[1*], Farida Anggraini Soetedjo[2]
[1]Anatomical Pathology Department, Faculty of Medicine, Universitas Wijaya Kusuma Surabaya, Surabaya, Indonesia
[2]Internal Medicine Department, Faculty of Medicine, Universitas Wijaya Kusuma Surabaya, Surabaya, Indonesia

Corresponding author: jimmy.hadiwidjaja@uwks.ac.id

ARTICLE INFO

Article history

Submitted:
19 April 2026

Revised:
18 August 2026

Accepted:
24 September 2026

Keywords:

Anthracycline toxicity;
Reactive oxygen species;
Lipid peroxidation;
Malondialdehyde;
Superoxide dismutase.

ABSTRACT

Doxorubicin is an effective anthracycline chemotherapeutic agent, however its clinical use is limited by oxidative stress–mediated toxicity. This study evaluated the protective effect of *Pachyrhizus erosus* tuber extract on oxidative stress markers in doxorubicin-induced Wistar rats. A controlled laboratory experiment with a post-test-only control group design was conducted using 30 female Wistar rats randomly assigned to five groups: negative control, doxorubicin control, and three extract-treated groups receiving *Pachyrhizus erosus* at doses of 1.25, 2.5, and 5 g/kg body weight. Doxorubicin was administered intraperitoneally at 5 mg/kg body weight on days 1, 5, 9, and 13, whereas the extract was given orally once daily for 14 days. On day 15, serum malondialdehyde and superoxide dismutase were measured using commercial assay kits. Serum malondialdehyde levels were significantly higher in the doxorubicin control group than in the negative control group (18.75 ± 1.60 vs. 5.12 ± 0.45 ng/mL; $p < 0.001$), then decreased to 12.98 ± 1.35, 8.52 ± 0.90, and 6.10 ± 0.72 ng/mL in the low-, medium-, and high-dose groups, respectively (all $p < 0.001$ vs. doxorubicin control). In contrast, superoxide dismutase activity was significantly lower in the doxorubicin control group than in the negative control group (35.1 ± 4.1 vs. 72.5 ± 5.8 U/mg protein; $p < 0.001$) and increased to 48.9 ± 5.2, 59.7 ± 4.5, and 68.3 ± 6.1 U/mg protein after extract treatment (all $p < 0.001$). In conclusion, *Pachyrhizus erosus* tuber extract attenuated doxorubicin-induced oxidative stress with a graded dose-related pattern.

This work is licensed under a Creative Commons Attribution-ShareAlike 4.0 International License.

INTRODUCTION

Cancer remains a major global health challenge, with nearly 20 million new cases and 9.7 million deaths estimated worldwide in 2022 (Belger et al., 2024; Lisna Ardelia et al., 2026). In this setting, anthracycline-based chemotherapy continues to play a central role in the treatment of common malignancies, including breast cancer, leukemia, and lymphoma (Kciuk et al., 2023). Among these agents, doxorubicin (DOX) remains one of the most effective and widely used cytotoxic drugs because of its ability to intercalate DNA, inhibit topoisomerase II, and disrupt nucleic acid synthesis (Schirone et al., 2022). However, the clinical utility of DOX is substantially constrained by dose-dependent toxicity, particularly injury to non-malignant tissues such as the heart and liver, which may necessitate dose reduction or premature discontinuation of therapy (Belger et al., 2024; Linders et al., 2024).

A principal mechanism underlying DOX-induced toxicity is oxidative stress. Through redox cycling and mitochondrial disruption, DOX promotes excessive generation of reactive oxygen species (ROS), overwhelming endogenous antioxidant defenses and triggering oxidative damage to cellular macromolecules (Antonucci et al., 2021; Shi et al., 2023). One of the best-recognized consequences is lipid peroxidation, which compromises membrane integrity and generates reactive aldehydes. Malondialdehyde (MDA), a major end-product of polyunsaturated

fatty acid peroxidation, is therefore widely used as a surrogate marker of oxidative injury. In parallel, superoxide dismutase (SOD), a key first-line enzymatic antioxidant that catalyzes superoxide detoxification, is often depleted or functionally impaired under sustained ROS burden. Accordingly, increased MDA together with reduced SOD activity represents a biologically coherent signature of DOX-induced redox imbalance and has become a useful experimental framework for evaluating antioxidant interventions (Lal-Shahsavar et al., 2025; Soltani Hekmat et al., 2025).

These limitations have stimulated ongoing interest in plant-derived compounds as adjunctive strategies to attenuate chemotherapy-associated oxidative injury. *Pachyrhizus erosus (bengkoang*, *jicama*), an edible tuber widely consumed in tropical regions, has attracted attention because of its antioxidant and nutraceutical potential (Jaiswal et al., 2021). Phytochemical studies indicate that *Pachyrhizus erosus* contains multiple bioactive constituents, including phenolic compounds, flavonoids, triterpenoids, and polysaccharide-rich fractions, several of which have shown free-radical scavenging and redox-modulating activities (Jaiswal et al., 2021; Minh et al., 2021; Xiang et al., 2025). More recent analyses of the tuber and peel have further shown appreciable polyphenol and flavonoid content with measurable antioxidant activity in ABTS, DPPH, and FRAP-based assays (Xiang et al., 2025). In vivo work on yam bean-derived preparations has also suggested favorable effects on oxidative stress markers in metabolic models, supporting the plausibility of a systemic antioxidant effect (Santoso et al., 2022).

Despite this emerging evidence, direct experimental data on the effect of *Pachyrhizus erosus* tuber extract in DOX-induced oxidative injury remain limited. Recent studies using a similar DOX experimental platform evaluated Cosmos caudatus but addressed different biological questions: Lisna Ardelia et al. (2026) assessed serum lipid-profile disturbances using a single extract dose during a longer DOX exposure schedule, whereas Soetedjo et al. (2026) evaluated lung histopathology together with 4-HNE, IL-6, Bax, and angiopoietin-2 at two extract doses. In contrast, the present study evaluates an edible *Pachyrhizus erosus* tuber preparation across three escalating doses and jointly examines serum MDA as a lipid-peroxidation marker and SOD activity as an endogenous enzymatic antioxidant endpoint. Thus, the study extends prior work through a distinct phytochemical source, a graded-dose design, and paired redox outcomes rather than merely substituting the test material. Therefore, the present study aimed to evaluate the effect of *Pachyrhizus erosus* tuber extract on MDA levels and SOD activity in DOX-induced Wistar rats.

METHOD

Study design and experimental animals

This study used a controlled laboratory experiment with a post-test-only control group design. Thirty healthy female Wistar rats (Rattus norvegicus), aged 6–8 weeks and weighing 150–200 g, were included. Following a 7-day acclimatization period, the animals were randomly assigned to five groups (n = 6 per group):

Female rats were selected to maintain a single-sex experimental cohort and reduce between-sex heterogeneity in this exploratory study. This choice should not be interpreted as sex-neutral because experimental evidence indicates sex-dependent responses to DOX, with ovarian hormones and estrogen-linked redox mechanisms contributing to differences in DOX-associated injury (Belger et al., 2024; Su et al., 2024). Accordingly, interpret the findings as specific to young female Wistar rats and confirm them in male animals.

1. Group I, negative control (distilled water, p.o. + normal saline, i.p.);
2. Group II, DOX control (distilled water, p.o. + DOX, i.p.);
3. Group III, low-dose *Pachyrhizus erosus* (1.25 g/kg body weight, p.o. + DOX, i.p.);
4. Group IV, medium-dose *Pachyrhizus erosus* (2.5 g/kg body weight, p.o. + DOX, i.p.);
5. Group V, high-dose *Pachyrhizus erosus* (5 g/kg body weight, p.o. + DOX, i.p.).

The three *Pachyrhizus erosus* doses were prespecified as a two-fold escalating exploratory series (1.25, 2.5, and 5 g/kg body weight) to characterize whether redox markers changed across increasing exposure. These dose levels were not derived from a formal extract-specific NOAEL or LD50 determination. The tuber is the edible portion of *Pachyrhizus erosus* and is chemically

distinct from the rotenone-containing toxic seeds (Jaiswal et al., 2021).

Animals were maintained under standard laboratory conditions at 25 °C with a 12 h light/dark cycle and had free access (*ad libitum*) to a standard pellet diet and water throughout the study. The Ethical Research Committee, Faculty of Medicine, Universitas Wijaya Kusuma Surabaya, Surabaya, Indonesia, approved this study under ethical clearance number 71/SLE/FK/UWKS/2026. All experimental procedures were performed in accordance with institutional regulations for the care and use of laboratory animals.

Plant material, reagents, extraction, and treatment protocol

Fresh Pachyrhizus erosus tubers were purchased from a local market in Surabaya, Indonesia, and taxonomically authenticated by a qualified botanist at the Herbal Laboratory, Materia Medica Batu, East Java Provincial Health Office, Indonesia. Doxorubicin hydrochloride (Kalbe Farma, Jakarta, Indonesia) was used to induce oxidative stress. Ethanol 96% for extraction was obtained from Merck (EMSURE®, Product No. 1.59010, Darmstadt, Germany). Lipid peroxidation for MDA quantification was determined using an Invitrogen™ Thiobarbituric Acid Reactants (TBARS) Colorimetric Assay Kit (Cat. No. EEA021, Thermo Fisher Scientific, Waltham, MA, USA) according to the manufacturer's protocol. Superoxide dismutase (SOD) activity was measured using a commercial SOD Assay Kit (Cat. No. 19160, Sigma-Aldrich, St. Louis, MO, USA) according to the manufacturer's instructions.

Furthermore, fresh *Pachyrhizus erosus* tubers were washed, peeled, and sliced, then oven-dried at 40–50 °C for 48 h before being milled into a coarse powder. The powdered material was macerated in 96% ethanol at room temperature for 3 × 24 h with intermittent agitation and protection from light. The combined filtrates were concentrated under reduced pressure using a rotary evaporator at 50 °C, yielding a viscous ethanolic extract that was stored in an amber container at 4 °C until use. Prior to administration, the extract was freshly reconstituted in distilled water to the desired concentrations (Xiang et al., 2025).

Doxorubicin (DOX) was administered intraperitoneally at 5 mg/kg body weight on days 1, 5, 9, and 13, giving a cumulative dose of 20 mg/kg. The extract was administered orally once daily for 14 consecutive days, beginning 1 day before the first DOX injection. Negative-control animals received the corresponding vehicle orally and intraperitoneally, whereas DOX-control animals received oral vehicle plus DOX according to the same schedule (Soetedjo et al., 2026).

Blood collection and biochemical assays

On day 15, or 24 h after the final DOX injection, rats were terminally anesthetized with ketamine hydrochloride/xylazine hydrochloride solution (Cat. No. K113, Sigma-Aldrich, St. Louis, MO, USA) administered intraperitoneally at ketamine and xylazine doses equivalent to 80 and 10 mg/kg body weight, respectively.

Serum lipid peroxidation was assessed using an Invitrogen™ TBARS Assay Kit (Cat. No. EEA021, Thermo Fisher Scientific, Waltham, MA, USA) according to the manufacturer's instructions. The assay is based on the formation of a pink chromogen when thiobarbituric acid reactive substances react with thiobarbituric acid under acidic, high-temperature conditions. Absorbance was measured at 532 nm using a microplate reader, and the results were expressed as malondialdehyde (MDA) levels in ng/mL. Moreover, serum SOD activity was determined using an SOD Assay Kit (Cat. No. 19160, Sigma-Aldrich, St. Louis, MO, USA) according to the manufacturer's protocol. This method is based on the inhibition of WST reduction by superoxide anions generated in the reaction system. Following incubation at 37 °C for 20 min, absorbance was measured at 450 nm using a microplate reader, and SOD activity was expressed as U/mg protein.

Statistical analysis

Statistical analyses were conducted using IBM SPSS Statistics for Windows, Version 27.0 (IBM Corp., Armonk, NY, USA). Quantitative data are presented as mean ± standard deviation (SD). Data distribution was assessed using the Shapiro-Wilk test, and homogeneity of variances was evaluated using Levene's test. When both assumptions were met, differences among the five groups were analyzed using one-way analysis of variance (ANOVA), followed by Tukey's [u6.1]honestly significant difference (HSD) post hoc test for multiple comparisons. All tests were two-sided, and a p-value < 0.05 was considered statistically significant.

RESULTS

The extraction procedure produced a viscous ethanolic *Pachyrhizus erosus* tuber extract that was reconstituted for oral administration at the three prespecified doses. Quantitative extraction yield was not recorded as a study endpoint. Biochemical data were available for all 30 animals, with six animals in each of the five groups. Descriptive findings for serum MDA and SOD are presented first in Tables 1 and 2.

Table 1. Effect of *Pachyrhizus erosus* tuber extract on serum malondialdehyde (MDA) levels in doxorubicin-induced Wistar rats

Group	Treatment	MDA level (ng/mL)
Group I	Negative control (distilled water, p.o. + normal saline, i.p.)	5.12 ± 0.45
Group II	DOX control (distilled water, p.o. + DOX, i.p.)	18.75 ± 1.60
Group III	Low-dose *Pachyrhizus erosus* (1.25 g/kg body weight, p.o. + DOX, i.p.)	12.98 ± 1.35
Group IV	Medium-dose *Pachyrhizus erosus* (2.5 g/kg body weight, p.o. + DOX, i.p.)	8.52 ± 0.90
Group V	High-dose *Pachyrhizus erosus* (5 g/kg body weight, p.o. + DOX, i.p.)	6.10 ± 0.72

Descriptively, the highest serum MDA level was observed in the DOX control group (Group II), at 18.75 ± 1.60 ng/mL, compared with 5.12 ± 0.45 ng/mL in the negative control group (Group I). MDA levels were lower across the *Pachyrhizus erosus* treatment groups, measuring 12.98 ± 1.35 ng/mL in Group III, 8.52 ± 0.90 ng/mL in Group IV, and 6.10 ± 0.72 ng/mL in Group V. Thus, MDA showed a graded numerical decrease across increasing extract doses; Group V was not significantly different from Group I in the Tukey HSD comparison (adjusted p = 0.535) (Tables 1 and 6).

Table 2. Effect of *Pachyrhizus erosus* tuber extract on serum superoxide dismutase (SOD) activity in doxorubicin-induced Wistar rats

Group	Treatment	SOD activity (U/mg protein)
Group I	Negative control (distilled water, p.o. + normal saline, i.p.)	72.5 ± 5.8
Group II	DOX control (distilled water, p.o. + DOX, i.p.)	35.1 ± 4.1
Group III	Low-dose *Pachyrhizus erosus* (1.25 g/kg body weight, p.o. + DOX, i.p.)	48.9 ± 5.2
Group IV	Medium-dose *Pachyrhizus erosus* (2.5 g/kg body weight, p.o. + DOX, i.p.)	59.7 ± 4.5
Group V	High-dose *Pachyrhizus erosus* (5 g/kg body weight, p.o. + DOX, i.p.)	68.3 ± 6.1

Descriptively, the lowest serum SOD activity was observed in the DOX control group (Group II), at 35.1 ± 4.1 U/mg protein, compared with 72.5 ± 5.8 U/mg protein in the negative control group (Group I). SOD activity increased across the *Pachyrhizus erosus* treatment groups, measuring 48.9 ± 5.2 U/mg protein in Group III, 59.7 ± 4.5 U/mg protein in Group IV, and 68.3 ± 6.1 U/mg protein in Group V. Thus, SOD showed a graded numerical increase across increasing extract doses; Group V was not significantly different from Group I in the Tukey HSD comparison (adjusted p = 0.633) (Tables 2 and 6).

Table 3. Shapiro-Wilk normality test results for serum MDA levels and SOD activity

Variable	Group	df	p-value	Interpretation
MDA	Group I	6	0.522	Normal
	Group II	6	0.084	Normal
	Group III	6	0.525	Normal
	Group IV	6	0.246	Normal
	Group V	6	0.297	Normal
SOD	Group I	6	0.617	Normal
	Group II	6	0.541	Normal
	Group III	6	0.353	Normal
	Group IV	6	0.563	Normal
	Group V	6	0.508	Normal

Shapiro-Wilk testing showed p-values greater than 0.05 in every group for both outcomes. For MDA, p-values ranged from 0.084 to 0.525, while for SOD they ranged from 0.353 to 0.617 (Table 3). Therefore, there was no evidence of departure from normality, and the normality assumption for parametric analysis was considered met.

Table 4. Levene's homogeneity-of-variance test results for serum MDA levels and SOD activity

Variable	Levene statistic	df1	df2	p-value	Conclusion
MDA	1.567	4	25	0.214	Homogeneous
SOD	0.870	4	25	0.496	Homogeneous

Levene's test was non-significant for MDA (Levene statistic = 1.567, p = 0.214) and SOD (Levene statistic = 0.870, p = 0.496) (Table 4). Because both p values were greater than 0.05, the variances across the five groups were considered homogeneous, fulfilling the homogeneity assumption for one-way ANOVA.

Table 5. One-way ANOVA results for serum MDA levels and SOD activity

Variable	F	df1	df2	p value	Interpretation
MDA	160.093	4	25	< 0.001	Significant
SOD	51.152	4	25	< 0.001	Significant

One-way ANOVA demonstrated statistically significant differences among the five groups for MDA, $F(4, 25) = 160.093$, $p < 0.001$, and SOD, $F(4, 25) = 51.152$, $p < 0.001$ (Table 5). These results indicate that at least one group mean differed significantly for each outcome. Because the omnibus tests were significant, Tukey HSD post hoc comparisons were performed to determine which specific groups differed.

Table 6. Tukey HSD pairwise comparisons for serum MDA levels and SOD activity

Variable	Pairwise comparison	Adjusted p-value	Interpretation
MDA	Group I vs. Group II	< 0.001	Significant
	Group I vs. Group III	< 0.001	Significant
	Group I vs. Group IV	< 0.001	Significant
	Group I vs. Group V	0.535	Not significant
	Group II vs. Group III	< 0.001	Significant
	Group II vs. Group IV	< 0.001	Significant
	Group II vs. Group V	< 0.001	Significant
	Group III vs. Group IV	< 0.001	Significant
	Group III vs. Group V	< 0.001	Significant
	Group IV vs. Group V	0.006	Significant
SOD	Group I vs. Group II	< 0.001	Significant
	Group I vs. Group III	< 0.001	Significant
	Group I vs. Group IV	0.002	Significant
	Group I vs. Group V	0.633	Not significant
	Group II vs. Group III	< 0.001	Significant
	Group II vs. Group IV	< 0.001	Significant
	Group II vs. Group V	< 0.001	Significant
	Group III vs. Group IV	0.011	Significant
	Group III vs. Group V	< 0.001	Significant
	Group IV vs. Group V	0.058	Not significant

For MDA, Tukey HSD showed that Group I differed significantly from Group II ($p < 0.001$), Group III ($p < 0.001$), and Group IV ($p < 0.001$), but not from Group V ($p = 0.535$). Group II also differed significantly from Groups III, IV, and V (all $p < 0.001$). Among the extract-treated groups, Groups III and IV ($p < 0.001$) and Groups III and V ($p < 0.001$) differed significantly, as did Groups IV and V ($p = 0.006$). These findings indicate a progressive reduction in MDA across the extract doses, with the highest-dose group not significantly different from the negative control (Table 6).

For SOD, Tukey HSD showed that Group I differed significantly from Group II ($p < 0.001$), Group III ($p < 0.001$), and Group IV ($p = 0.002$), but not from Group V ($p = 0.633$). Group II differed

significantly from Groups III, IV, and V (all $p < 0.001$). Group III also differed significantly from Group IV ($p = 0.011$) and Group V ($p < 0.001$), whereas Groups IV and V did not differ significantly ($p = 0.058$). These findings indicate progressive restoration of SOD activity across the extract doses, with the highest-dose group not significantly different from the negative control and no statistically significant additional increase from the medium to the high dose (Table 6).

DISCUSSION

The present findings show a clear and internally consistent antioxidant effect of *Pachyrhizus erosus* tuber extract in DOX-exposed rats. DOX administration was associated with a marked increase in serum MDA and a parallel decrease in SOD activity, whereas extract treatment shifted both markers in the opposite direction. The pairwise Tukey analysis supports this interpretation: each extract group differed significantly from the DOX control for both outcomes, while the highest-dose group did not differ significantly from the negative control for MDA ($p = 0.535$) or SOD ($p = 0.633$). Across the three administered doses, MDA decreased, and SOD increased in a graded pattern; however, because no formal trend test was conducted and the SOD difference between Groups IV and V was not significant ($p = 0.058$), the findings support a dose-related pattern rather than a statistically proven dose-dependent effect.

The increase in MDA in the DOX control group is consistent with the known redox-active nature of anthracyclines (Vitale et al., 2024). Doxorubicin undergoes intracellular redox cycling and promotes excessive reactive oxygen species formation, which attack polyunsaturated fatty acids in cellular membranes and generate secondary lipid peroxidation products (Sangweni et al., 2022). In experimental models, elevated MDA or TBARS is one of the most reproducible biochemical consequences of this process (Arruda et al., 2024). Within that framework, the significant reduction in MDA after *Pachyrhizus erosus* extract administration indicates attenuation of oxidative membrane damage rather than a nonspecific biochemical fluctuation. This interpretation is further supported by the orderly gradient across the three extract doses, supporting a genuine treatment effect.

The significant reduction in MDA after *Pachyrhizus erosus* extract administration indicates attenuated oxidative membrane damage rather than a nonspecific biochemical fluctuation. This interpretation is further supported by the orderly gradient across the three extract doses, which argues for a genuine treatment effect.

The SOD findings reinforce this interpretation. Doxorubicin reduced SOD activity to less than half of the negative-control value, whereas *Pachyrhizus erosus* extract restored SOD in a graded manner. This result is especially relevant because SOD is a primary enzymatic defense against superoxide radicals, and its depletion reflects impaired antioxidant capacity under sustained oxidant stress (Zheng et al., 2023). The recovery of SOD activity in the treatment groups therefore suggests that the extract did not act only by limiting oxidative end-products, but also by preserving or improving the antioxidant defense system itself. Together, the MDA and SOD results indicate that the extract influenced the redox state in a coordinated manner, a stronger signal of biological protection than movement in either marker alone.

A plausible basis for the decrease in MDA is the phytochemical composition of *Pachyrhizus erosus*. Contemporary profiling of the tuber and peel identified abundant polyphenols and flavonoids; Xiang et al. (2025) characterized 30 compounds in the most active fractions, including 21 flavonoids, six phenolics, two coumarins, and one lignan. They demonstrated measurable DPPH, ABTS, and ferric-reducing antioxidant activity. Phenolic and flavonoid compounds can participate in electron- or hydrogen-donation reactions that neutralize reactive species and interrupt lipid-peroxidation chain propagation. In a DOX model, reducing the radical burden available to attack membrane polyunsaturated fatty acids provides a biologically plausible route to lower formation of secondary lipid-peroxidation products measured as MDA. The graded fall in MDA across the extract groups is therefore consistent with the documented antioxidant capacity of *Pachyrhizus erosus* fractions (Jaiswal et al., 2021; Xiang et al., 2025), while recognizing that the exact composition of the crude ethanolic preparation used here was not quantified.

The increase in SOD activity may reflect preservation of endogenous enzyme function under a lower oxidant burden and, potentially, regulation of antioxidant-response pathways. Some metabolites identified in *Pachyrhizus erosus* have been linked in prior literature to Nrf2-Keap1 and NF-κB signaling, and Nrf2 is a central regulator of antioxidant and cytoprotective gene expression (Ngo & Duennwald, 2022; Xiang et al., 2025). Experimental DOX studies also support the biological relevance of Nrf2 activation for limiting oxidative injury (Zhang et al., 2021). Thus, direct radical scavenging could reduce MDA, while preserving or inducing endogenous antioxidant defenses could contribute to the higher SOD activity observed after extract treatment. Nevertheless, the present study did not measure Nrf2 activation, SOD gene/protein expression, or individual extract constituents; these mechanisms remain biologically plausible explanations rather than demonstrated causal pathways.

Translation of these findings to supportive chemotherapy requires caution because antioxidant co-administration can interact with the antitumor activity of DOX. DOX acts through multiple mechanisms, including topoisomerase II interference, DNA damage, mitochondrial dysfunction, and ROS generation; therefore, altering cellular redox balance may have compound- and context-specific consequences. In xenograft and cancer-cell models, exogenous glutathione reduced DOX anticancer efficacy despite attenuating organ toxicity (Shen et al., 2019), while a systematic review emphasizes that antioxidant supplements may either modify toxicity or interfere with anticancer treatment depending on the agent, dose, timing, and tumor context (Krejbich & Birringer, 2022). Because the present experiment used healthy, non-tumor-bearing rats and did not evaluate tumor response, it cannot determine whether *Pachyrhizus erosus* preserves, enhances, or diminishes DOX anticancer efficacy.

Several limitations should be acknowledged. First, lipid peroxidation was assessed using a TBARS-based assay, which is widely used but is not completely specific for MDA because thiobarbituric acid can react with other aldehydic compounds. Second, the analysis was limited to MDA and SOD and therefore does not capture the broader antioxidant, inflammatory, apoptotic, or histopathological response to DOX. Third, the extract was a crude ethanolic preparation whose active constituents were not individually standardized or quantified, and the selected 1.25-5 g/kg dose series was exploratory rather than derived from an extract-specific NOAEL or LD50 study. Fourth, the study included only female rats; sex-related differences in DOX toxicity and estrogen-linked redox responses limit generalization to males (Belger et al., 2024; Su et al., 2024). Fifth, no standard antioxidant positive-control group was included, preventing direct comparison of the effect size with an established protective agent such as N-acetylcysteine or vitamin E. Sixth, longitudinal body weight, feed and water intake, and systematic mortality/adverse-event data were not available for quantitative analysis, which limits assessment of general condition and potential high-dose extract toxicity. Seventh, the 14-day treatment period was substantially shorter than many clinical anthracycline regimens and does not address chronic or cumulative toxicity. Eighth, the study used healthy rather than tumor-bearing animals, so the potential interaction between *Pachyrhizus erosus* and DOX anticancer efficacy remains unknown. Future studies should incorporate both sexes, standardized phytochemical characterization, dedicated safety testing, a standard antioxidant comparator, longitudinal toxicity monitoring, longer exposure, formal trend analysis, and tumor-bearing models that evaluate both organ protection and anticancer efficacy.

CONCLUSION

Pachyrhizus erosus tuber extract attenuated doxorubicin-induced oxidative stress in female Wistar rats, as shown by reduced serum MDA levels and restored SOD activity. The protective effect showed a graded dose-related pattern, and the highest administered dose was not significantly different from the negative control for MDA (adjusted $p = 0.535$) or SOD (adjusted $p = 0.633$) in Tukey HSD comparisons. These findings support the potential antioxidant effect of *Pachyrhizus erosus* extract against doxorubicin-associated oxidative injury. Further studies are needed to confirm its active constituents, underlying mechanisms, safety, tissue-level protective effects, and whether this protection can be achieved without compromising doxorubicin's antitumor activity.

AUTHOR'S DECLARATION

Authors' contributions and responsibilities

JHW: Conceptualization, methodology, investigation, formal analysis, data curation, visualization, writing original draft preparation; **FAS**: Conceptualization, supervision, validation, methodology, writing (support), review, and editing.

Funding

This research received no specific grant from any funding agency in the public, commercial, or not-for-profit sectors.

Availability of data and materials

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

Competing interests

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

ACKNOWLEDGEMENT

The authors express sincere appreciation to the Laboratory of the Faculty of Medicine, Universitas Wijaya Kusuma Surabaya, for providing the facilities and technical support required for this study.

REFERENCES

Antonucci, S., Di Sante, M., Tonolo, F., Pontarollo, L., Scalcon, V., Alanova, P., Menabò, R., Carpi, A., Bindoli, A., Rigobello, M. P., Giorgio, M., Kaludercic, N., & Di Lisa, F. (2021). The Determining Role of Mitochondrial Reactive Oxygen Species Generation and Monoamine Oxidase Activity in Doxorubicin-Induced Cardiotoxicity. *Antioxidants & Redox Signaling*, *34*(7), 531–550. https://doi.org/10.1089/ars.2019.7929

Arruda, F. S., Tomé, F. D., Milhomem, A. C., Franco, P. I. R., Justino, A. B., Franco, R. R., Campos, E. C., Espindola, F. S., Soave, D. F., & Celes, M. R. N. (2024). Curcumin Attenuates Doxorubicin-Induced Cardiac Oxidative Stress and Increases Survival in Mice. *Pharmaceutics*, *16*(8), 1105. https://doi.org/10.3390/pharmaceutics16081105

Belger, C., Abrahams, C., Imamdin, A., & Lecour, S. (2024). Doxorubicin-induced cardiotoxicity and risk factors. *IJC Heart & Vasculature*, *50*, 101332. https://doi.org/10.1016/j.ijcha.2023.101332

Jaiswal, V., Chauhan, S., & Lee, H.-J. (2021). The Bioactivity and Phytochemicals of Pachyrhizus erosus (L.) Urb.: A Multifunctional Underutilized Crop Plant. *Antioxidants*, *11*(1), 58. https://doi.org/10.3390/antiox11010058

Kciuk, M., Gielecińska, A., Mujwar, S., Kołat, D., Kałuzińska-Kołat, Ż., Celik, I., & Kontek, R. (2023). Doxorubicin—An Agent with Multiple Mechanisms of Anticancer Activity. *Cells*, *12*(4), 659. https://doi.org/10.3390/cells12040659

Krejbich, P., & Birringer, M. (2022). The Self-Administered Use of Complementary and Alternative Medicine (CAM) Supplements and Antioxidants in Cancer Therapy and the Critical Role of Nrf-2—A Systematic Review. *Antioxidants*, *11*(11), 2149. https://doi.org/10.3390/antiox11112149

Lal-Shahsavar, S., Zolbanin, N. M., Jafari, A., & Ghasemnejad-Berenji, M. (2025). Metformin alleviates doxorubicin-induced hepatic damage by modulating oxidative stress: a molecular, biochemical, and histopathological approach in a rat model. *Naunyn-Schmiedeberg's Archives of Pharmacology*, *398*(7), 9099–9107. https://doi.org/10.1007/s00210-024-03688-2

Linders, A. N., Dias, I. B., López Fernández, T., Tocchetti, C. G., Bomer, N., & Van der Meer, P. (2024).

A review of the pathophysiological mechanisms of doxorubicin-induced cardiotoxicity and aging. *Npj Aging*, *10*(1), 9. https://doi.org/10.1038/s41514-024-00135-7

Lisna Ardelia, N., Wardhana, I. G. A. K., Kristijanto, J. A. F., Triadi, A., & Soetedjo, F. A. (2026). Ethanolic Cosmos caudatus Extract Mitigates Doxorubicin-Induced Serum Lipid Profile Derangements in Female Wistar Rats. *Asian Journal of Healthy and Science*, *5*(2), 86–97. https://doi.org/10.58631/ajhs.v5i2.285

Minh, T. T., Toan, H. K., Anh, H. T. L., Huong, T. T., Thao, D. T., & Hoang, V. D. (2021). Chemical Constituents from the Leaves of Pachyrhizus erosus Collected in Vietnam. *Chemical and Pharmaceutical Bulletin*, *69*(11), c21-00614. https://doi.org/10.1248/cpb.c21-00614

Ngo, V., & Duennwald, M. L. (2022). Nrf2 and Oxidative Stress: A General Overview of Mechanisms and Implications in Human Disease. *Antioxidants*, *11*(12), 2345. https://doi.org/10.3390/antiox11122345

Sangweni, N. F., Gabuza, K., Huisamen, B., Mabasa, L., van Vuuren, D., & Johnson, R. (2022). Molecular insights into the pathophysiology of doxorubicin-induced cardiotoxicity: a graphical representation. *Archives of Toxicology*, *96*(6), 1541–1550. https://doi.org/10.1007/s00204-022-03262-w

Santoso, P., Maliza, R., Rahayu, R., Astrina, Y., Syukri, F., & Maharani, S. (2022). Extracted yam bean (Pachyrhizus erosus (L.) Urb.) fiber counteracts adiposity, insulin resistance, and inflammation while modulating gut microbiota composition in mice fed with a high-fat diet. *Research in Pharmaceutical Sciences*, *17*(5), 558–571. https://doi.org/10.4103/1735-5362.355213

Schirone, L., D'Ambrosio, L., Forte, M., Genovese, R., Schiavon, S., Spinosa, G., Iacovone, G., Valenti, V., Frati, G., & Sciarretta, S. (2022). Mitochondria and Doxorubicin-Induced Cardiomyopathy: A Complex Interplay. *Cells*, *11*(13), 2000. https://doi.org/10.3390/cells11132000

Shen, B.-Y., Chen, C., Xu, Y.-F., Shen, J.-J., Guo, H.-M., Li, H.-F., Li, X.-N., Kang, D., Shao, Y.-H., Zhu, Z.-P., Yin, X.-X., Xie, L., Wang, G.-J., & Liang, Y. (2019). Is the combinational administration of doxorubicin and glutathione a reasonable proposal? *Acta Pharmacologica Sinica*, *40*(5), 699–709. https://doi.org/10.1038/s41401-018-0158-8

Shi, S., Chen, Y., Luo, Z., Nie, G., & Dai, Y. (2023). Role of oxidative stress and inflammation-related signaling pathways in doxorubicin-induced cardiomyopathy. *Cell Communication and Signaling*, *21*(1), 61. https://doi.org/10.1186/s12964-023-01077-5

Soetedjo, F. A., Budianto, N. E. W., Khamidah, N., Suhartati, S., Kristijanto, J. A. F., & Nugraha, I. C. (2026). Cosmos caudatus Extract Attenuates Doxorubicin-Associated Lung Injury in Female Wistar Rats: Histopathological and Biomarker Evidence From 4-HNE, IL-6, Bax and Angiopoietin-2. *Open Access Research Journal of Biology and Pharmacy*, *16*(1), 029–040. https://doi.org/10.53022/oarjbp.2026.16.1.0019

Soltani Hekmat, A., Hekmat, M., Ramezanipour, S., & Javanmardi, K. (2025). Protective effects of Alamandine against doxorubicin-induced liver injury in rats. *BMC Pharmacology and Toxicology*, *26*(1), 95. https://doi.org/10.1186/s40360-025-00932-0

Su, Q., Fu, M., Xiao, W., Zhao, H., Yu, Y., Feng, J., Liu, Y., Wan, Y., Yu, Y., & Zhang, C. (2024). Sex Differences in Doxorubicin-Induced Cardiotoxicity: Insights from Transcriptome Analysis. *Advanced Therapeutics*, *7*(9), 2400036. https://doi.org/10.1002/adtp.202400036

Vitale, R., Marzocco, S., & Popolo, A. (2024). Role of Oxidative Stress and Inflammation in Doxorubicin-Induced Cardiotoxicity: A Brief Account. *International Journal of Molecular Sciences*, *25*(13), 7477. https://doi.org/10.3390/ijms25137477

Xiang, J., Huang, S., Wu, X., He, Y., Shen, H., Tang, S., Zhu, F., & Luo, Y. (2025). Phytochemical Profile and Antioxidant Activity of the Tuber and Peel of Pachyrhizus erosus. Antioxidants, 14(4), 416. https://doi.org/10.3390/antiox14040416

Zhang, W.-B., Lai, X., & Guo, X.-F. (2021). Activation of Nrf2 by miR-152 Inhibits Doxorubicin-Induced Cardiotoxicity via Attenuation of Oxidative Stress, Inflammation, and Apoptosis. *Oxidative Medicine and Cellular Longevity*, *2021*(1). https://doi.org/10.1155/2021/8860883

Zheng, M., Liu, Y., Zhang, G., Yang, Z., Xu, W., & Chen, Q. (2023). The Applications and Mechanisms of Superoxide Dismutase in Medicine, Food, and Cosmetics. *Antioxidants*, *12*(9), 1675. https://doi.org/10.3390/antiox12091675