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Antianaemic Effects *Ofmyrianthus Serratusstem* Bark Extract in Phenylhydrazine-Induced Haemolytic Anaemia in Wistar Rats

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ABSTRACT
The stem bark of *Myrianthus serratus* is used in northern Nigerian ethnomedicine to treat anaemia. This study evaluated the effect of its aqueous stem bark extract (AEMS) on haematological, hepatorenal, and oxidative stress markers in phenylhydrazine (PHZ)-induced haemolyticanaemia. A randomised, controlled design was used in 47 adult Wistar rats. Acute oral toxicity was assessed up to 5000 mg/kg by the Lorke method. Anaemia was induced with PHZ (35 mg/kg bw) and confirmed haematologically before treatment. Rats were randomly allocated to five groups (n = 7/group) receiving distilled water, AEMS (200 or 400 mg/kg bw) or vitamin E orally by gavage for 21 days; a non-anaemic group served as normal control. Blood was collected at termination for haematological indices, serum biochemical markers, and antioxidant activities. Data were analysed by one-way ANOVA with Tukey's post-hoc test (SPSS v24.0), with significance at $p < 0.05$. AEMS caused no deaths or signs of toxicity up to 5000 mg/kg. At 400 mg/kg, AEMS raised RBC count and haematocrit relative to untreated anaemic rats, matching vitamin E, but did not raise haemoglobin; 200 mg/kg had no effect on RBC count. Both doses normalised PHZ-elevated red cell indices, leukocyte and lymphocyte counts, AST, ALT, total bilirubin, urea and creatinine ($p < 0.05$), with larger effects at 400 mg/kg. AEMS lowered TBARS by 40–45% and returned serum catalase and GSH towards control values. AEMS is of low acute toxicity and partially corrects PHZ-induced erythrocyte loss while protecting the liver and kidney, plausibly by limiting oxidative haemolysis.

Keywords: Myrianthus Serratus, Phenylhydrazine, Oxidative Stress, Hepatoprotection, Nephroprotection.

INTRODUCTION

Anaemia is a reduction in red blood cell (RBC) number or oxygen-carrying capacity below physiological need. It is diagnosed when haemoglobin falls below 110 g/L in pregnant women and children aged 6–59 months, and below 120 g/L in non-pregnant women of reproductive age (WHO, 2025; Obasohan *et al*., 2022). Its causes include nutritional deficiencies, malaria, obstetric and gynaecological conditions and inherited red cell disorders, all shaped by poverty, gender inequality, low education and unequal access to care (WHO, 2025). Anaemia causes fatigue, dizziness, breathlessness and lost productivity. In pregnancy, it raises the risk of maternal and perinatal death, miscarriage and low birthweight, and impairs the cognitive and motor development of the child (WHO, 2025).

Global progress has stalled. Anaemia affected 29.3% of women aged 15–49 years in 2000 and 30.7% (about 605 million women) in 2023 (WHO, 2025). The WHO African Region carries a 36.5% prevalence, rising to 41.9% in Western Africa and 49.4% among pregnant women there. Only one of 194 countries is on track to halve prevalence by 2030, and 125 are worsening (Onambele*et al*., 2023).

Nigeria lies at the severe end of this distribution. Anaemia affected 57.8% of women of reproductive age in 2018, exceeding 70% in Sokoto, Ebonyi, Zamfara and Katsina (Kareem *et al*., 2022), and 47% in 2024, with pregnant women more affected (50%) than non-pregnant women (46%) (NPC & ICF, 2025). In 2018, 68.1% of children aged 6–59 months were anaemic, and every state exceeded the 40% WHO threshold for a severe public health problem (Obasohan *et al*., 2022). Prevalence is highest among rural, poor, and unschooled households, and malaria is a major driver in children (Kareem *et al*., 2022; Obasohan *et al*., 2022). Persistence despite iron supplementation, malaria control, and deworming points to a need for affordable, locally available adjunct therapies.

Medicinal plants are the first line of care for many rural Nigerians, and several have shown haematinic activity in experimental anaemia (Sanni *et al*., 2005; Gheith & El-Mahmoudy, 2018; Asogwa *et al*., 2024). Phenylhydrazine (PHZ) is the standard agent for modelling haemolyticanaemia. Its autoxidation within erythrocytes generates phenyl and hydroxyl radicals and hydrogen peroxide, which oxidisehaemoglobin, peroxidise membrane lipids and trigger haemolysis and splenic clearance of damaged cells (Beutler, 2001; Pandey *et al*., 2014). Because the haem, iron, and oxidant load of haemolysis also injures the liver and kidney, the model tests both red cell recovery and organ protection (Aloke *et al*., 2021).

Myrianthus is a genus of flowering plants in the family Urticaceae (formerly placed in Cecropiaceae), distributed mainly across tropical Africa, whose members are predominantly dioecious trees, shrubs or lianas bearing simple leaves that are either pinnately veined or palmately structured (Thiombiano et al., 2012). *Myrianthus serratus*, known locally in Nigeria as *faringanye* (Hausa), *apulu* (Jukun) and *uweje* (Idoma), has been documented in ethnobotanical surveys in northern Nigeria as a traditional remedy for anaemia and malarial fever, employing both the stem bark and the leaves (Enegide et al., 2015).

This study therefore evaluated the acute oral toxicity of the aqueous stem bark extract of *M. serratus* (AEMS) and its effects on haematological indices, liver and kidney function, and oxidative stress markers in PHZ-induced haemolyticanaemia in Wistar rats. We hypothesized that AEMS would restore erythrocyte indices and limit PHZ-induced hepatorenal and oxidative injury in a dose-dependent manner.

MATERIALS AND METHODS

Chemicals

Phenylhydrazine hydrochloride and vitamin E (α-tocopherol) were obtained from Sigma Chemical Company, St. Louis, MO, USA. Diagnostic kits for Aspartate aminotransferase (AST), alanine aminotransferase (ALT), bilirubin, total protein, albumin, urea, creatinine, and electrolytes were from Randox. All other reagents were of analytical grade.

Plant Collection

Fresh stem bark of *M. serratus* was collected in April, 2022 from Lugga Mountain, Modire ward, Girei Local Government Area, Adamawa State, Nigeria. The plant was identified by James, Bristone Polaat the Department of Plant Science, Modibbo Adama University, Yola, Adamawa State.

Preparation of The Aqueous Extract

The bark was shade-dried at room temperature for 7 days, pulverized in a mortar, and sieved through a 60-mesh screen. The powdered stem barkweighing 100 g was macerated in 1000 mL distilled water for 72 h with intermittent shaking and filtered through Whatman No. 1 paper (Harborne, 1973). The filtrate was evaporated to dryness in a water bath at 40 °C, giving 100 g of extract (yield 8.23% w/w), which was stored at 4 °C in the refrigerator and reconstituted in distilled water before use.

Experimental Animal

Both sexes of healthy Wistar rats (100–120g; n = 47) were obtained from the National Veterinary Research Institute, Vom, Nigeria. They were housed in ventilated cages at 28–30 °C under a 12 h light/dark cycle, with standard pellet feed and tap water *ad libitum*, and acclimatized for 14 days. The protocol followed the Animal Research: Reporting of In Vivo Experiments (ARRIVE) 2.0 guidelines and the National Institutes of Health (NIH)*Guide for the Care and Use of Laboratory Animals*.

Acute Toxicity

Acute toxicity was assessed by a modified two-phase method of Lorke (1983) in 12 rats. In phase 1, three groups of three rats received AEMS orally at 100, 600, and 1000 mg/kg. In phase 2, three rats received 2000, 3000 or 5000 mg/kg (one rat per dose). Rats were observed closely for the first 4 h and then for 24 h (phase 1) or 72 h (phase 2) for hyperactivity, salivation, paw-licking, writhing, paralysis, respiratory distress, and death.

Induction of Anaemia

Anaemia was induced as described by Sanni et al. (2005), adapted from Harris & Kugler (1971). Rats received a single subcutaneous injection of phenylhydrazine hydrochloride at 35 mg/kg body weight, administered into the dorsal scruff using a sterile 1 mL syringe fitted with a 26-gauge needle. Anaemia was confirmed after 24 h by haemoglobin< 11 g/dL or PCV reduction > 29% from baseline; only confirmed rats entered the study.

Experimental Design

Thirty-five rats were randomly allocated to five groups (n = 7) and treated orally once daily for 21 days:

Table 1: Experimental Design of the Aqueous Stem Bark Extract of *Myrianrhus Serratus*

Group	Treatment (oral, daily × 21 days)
I. Normal control	Distilled water (10 mL/kg)
II. PHZ control	Distilled water (10 mL/kg)
III. PHZ + vitamin E	Vitamin E, 3.5 mg/kg
IV. PHZ + AEMS 200	AEMS, 200 mg/kg
V. PHZ + AEMS 400	AEMS, 400 mg/kg

Sample Collection

Twenty-four hours after the last dose, rats were fasted overnight and anesthetized using diethyl ether and sacrificed. Blood was collected by cardiac puncturewith a surgical blade into Ethylenediaminetetraacetic acid (EDTA) tubes for haematology and into plain tubes for serum. Plain tubes stood for 2 h and were centrifuged at 3000 rpm for 10 min; serum was stored at −20 °C in the refrigerator until required for analysis.

Haematological Analysis

Red Blood Cell (RBC) count, haemoglobin (Hb), haematocrit (HCT), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC), platelet count and total and differential leukocyte counts were measured on an automated analyser (Mindray BC-series, Shenzhen, China).

Liver and Kidney Function

Serum AST and ALT activities were measured by the method of Reitman and Frankel (1957); total and direct bilirubin and total protein following Tietz (1990); albumin by the bromocresol green method (Doumas & Biggs, 1972); urea by the Berthelot reaction (Weatherburn, 1967); creatinine by (Chirmeister*et al*., 1964); sodium following Trinder (1951); and potassium and chloride following Tietz (1990).

Oxidative Stress Markers

Serum catalase activity was measured by the dichromate–acetic acid method of Sinha (1972), reduced glutathione (GSH) with Ellman's reagent (Jollow*et al*., 1974), and lipid peroxidation as thiobarbituric acid-reactive substances (TBARS), expressed as malondialdehyde equivalents (Iqbal *et al*., 1996).

Statistical Analysis

Results were expressed as mean ± standard error of the mean. Statistical data were analyzed using a one-way analysis of variance (ANOVA) (SPSS 24.0). Treatment groups were compared with the control using Dunnett's

tests. For all comparisons, p-values less than 0.05 were considered statistically significant

RESULTS AND DISCUSSION

Acute Oral Toxicity

Aqueous stem bark extract of *Myrianthus serratus* produced no deaths and no behavioural signs of toxicity at any dose up to 5000 mg/kg during 72 h of observation. The oral LD_{50} is therefore above 5000 mg/kg, placing aqueous stem bark extract of *Myrianthus serratus* in the lowest hazard class of the Globally Harmonized System (category 5 or unclassified). Administration of phenylhydrazine significantly (p<.05) decreased RBC, Hb, and HCTcompared to the normal control, as shown in Table 2. Groups treated with 200 and 400 mg/kg bw of aqueous stem extract of *Myrianthus serratus*showed a significant increase in RBC, Hb, and HCT compared to the phenylhydrazine control group.

Table 2: Effect of Aqueous Stem Bark Extract of *Myrianthus Serratus* on Erythrocyte Count, Haemoglobin and Haematocrit in PHZ-Induced Anaemic Rats

Group	RBC ($\times10^{12}$/L)	Hb (g/dL)	HCT (%)
Normal control	7.43 ± 0.06^{a}	13.70 ± 0.12^{a}	38.40 ± 0.12^{b}
PHZ control	4.06 ± 0.54^{c}	9.00 ± 0.58^{c}	28.50 ± 0.23^{d}
PHZ + vitamin E	5.80 ± 0.06^{b}	12.10 ± 0.06^{b}	40.30 ± 0.12^{a}
PHZ + AEMS 200 mg/kg	4.02 ± 0.52^{c}	11.30 ± 0.58^{b}	33.20 ± 0.58^{c}
PHZ + AEMS 400 mg/kg	5.80 ± 0.12^{b}	11.80 ± 0.10^{b}	41.40 ± 0.12^{a}

Values are mean ± SEM (n = 7). Within a column, means with different superscripts differ significantly (p < 0.05; one-way ANOVA, Duncan's test). AEMS, aqueous stem bark extract of *M. serratus*; PHZ, phenylhydrazine.

Table 3shows the effect of aqueous stem bark extract of *Myrianthus serratus* on blood indices. The anaemic rats showed a significant (p<.05) increase in MCV and decrease in MHCH compared to the normal control. MCH insignificantly (p>0.05) increase in Phenylhydrazine group compared to the normal control. Group administered with 200 and 400 mg/kg bw significant (p<0.05) decrease in MCH and MCV indices and increased in MHCH blood indices compared to the normal group.

Table 3: Effect of AEMS On Red Cell Indices and Platelet Count

Group	MCV (fL)	MCH (pg)	MCHC (g/dL)
Normal control	51.68 ± 0.45^{a}	18.44 ± 0.22^{a}	35.68 ± 0.33^{a}
PHZ control	70.20 ± 9.35^{ab}	22.17 ± 3.28^{a}	31.58 ± 2.05^{abc}
PHZ + vitamin E	69.48 ± 0.75^{ab}	20.86 ± 0.24^{a}	30.02 ± 0.17^{bc}
PHZ + AEMS 200 mg/kg	82.59 ± 10.78^{b}	28.11 ± 3.91^{a}	34.04 ± 1.85^{ab}
PHZ + AEMS 400 mg/kg	71.38 ± 1.49^{ab}	20.34 ± 0.45^{a}	28.50 ± 0.26^{c}

Legend as Table 1. MCV, Mean Corpuscular Volume MCH, Mean Corpuscular HaemoglobinMHCH, Mean Corpuscular Haemoglobin Concentration

Groups administered with phenylhydrazine showed a significant (p<0.05) increase in WBC, lymphocytes, and neutrophils compared to the normal control (Table 4). Monocytes showed no significant difference (p>.05) compared with the normal group. Administration of the aqueous stem bark extract of *Myrianthus serratus* at 200 and 400 mg/kg bw showed a significant (p<.05) decrease in the treated group compared to the normal control. Monocyte and neutrophil counts did not differmeaningfully between the groups.

Table 4: Effect of AEMS On Total and Differential Leukocyte Counts

Group	WBC ($\times10^{9}$/L)	Lymphocytes ($\times10^{9}$/L)	Monocytes ($\times10^{9}$/L)	Neutrophils ($\times10^{9}$/L)
Normal control	6.90 ± 0.23^{c}	6.20 ± 0.12^{c}	0.50 ± 0.06^{ab}	0.20 ± 0.06^{ab}
PHZ control	13.40 ± 0.17^{a}	12.50 ± 0.21^{a}	0.50 ± 0.06^{ab}	0.40 ± 0.06^{b}
PHZ + vitamin E	7.10 ± 0.06^{c}	6.40 ± 0.06^{c}	0.43 ± 0.03^{ab}	0.27 ± 0.03^{ab}
PHZ + AEMS 200 mg/kg	11.40 ± 0.17^{b}	10.60 ± 0.17^{b}	0.50 ± 0.06^{ab}	0.30 ± 0.06^{ab}
PHZ + AEMS 400 mg/kg	8.87 ± 0.26^{d}	8.03 ± 0.38^{d}	0.43 ± 0.09^{ab}	0.40 ± 0.02^{a}

Legend as Table 1. WBC, white blood count

PHZ raised AST 4.4-fold, ALT 3-fold, and total bilirubin 2.5-fold (Table 5). AEMS lowered AST dose-dependently (by 21% and 41%) and ALT by 40% and 57%; at 400 mg/kg ALT was not different from normal. Both doses halved total bilirubin, outperforming vitamin E. PHZ also raised total protein and albumin; all treatments lowered them, with albumin returning to near-normal values.

Table 5: Effect of AEMS On Serum Liver Function Markers

Group	AST (U/L)	ALT (U/L)	Total bilirubin (µmol/L)	Direct bilirubin (µmol/L)	Total protein (g/dL)	Albumin (g/dL)
Normal control	8.50 ± 0.87^{d}	6.67 ± 0.67^{d}	6.10 ± 0.12^{d}	3.40 ± 0.06^{abc}	6.52 ± 1.16^{e}	2.33 ± 0.28^{c}
PHZ control	37.67 ± 1.67^{a}	20.33 ± 0.67^{a}	15.50 ± 0.23^{a}	4.60 ± 0.23^{a}	28.94 ± 0.84^{a}	5.63 ± 0.28^{a}
PHZ + vitamin E	9.50 ± 0.50^{d}	8.00 ± 1.15^{d}	9.80 ± 0.12^{b}	3.90 ± 0.06^{ab}	10.44 ± 0.28^{d}	3.00 ± 0.06^{b}
PHZ + AEMS 200 mg/kg	29.67 ± 0.67^{b}	12.17 ± 1.30^{b}	7.67 ± 0.34^{c}	1.60 ± 0.15^{d}	21.72 ± 0.63^{b}	2.80 ± 0.12^{bc}
PHZ + AEMS 400 mg/kg	22.33 ± 1.33^{c}	8.67 ± 0.67^{cd}	7.87 ± 0.22^{c}	3.30 ± 0.85^{bc}	17.33 ± 0.62^{c}	2.90 ± 0.06^{bc}

Legend as Table 1.

PHZ raised urea 9.6-fold and creatinine by 70%, with increases in sodium, potassium and chloride (Table 6). AEMS reduced urea by 34% and 69% and creatinine by 18% and 29% at 200 and 400 mg/kg; at 400 mg/kg creatinine fell slightly below the vitamin E value. Potassium and chloride at 400 mg/kg returned to near-normal values.

Table 6: Effect of AEMS On Serum Kidney Function Markers and Electrolytes

Group	Urea (mmol/L)	Creatinine (µmol/L)	Na^{+} (mmol/L)	K^{+} (mmol/L)	Cl^{-} (mmol/L)
Normal control	2.40 ± 0.12^{e}	54.97 ± 0.12^{e}	82.50 ± 0.23^{e}	4.20 ± 0.06^{d}	96.20 ± 0.12^{c}
PHZ control	23.00 ± 0.23^{a}	93.47 ± 0.17^{a}	154.90 ± 0.52^{a}	17.70 ± 0.06^{a}	106.40 ± 0.23^{a}
PHZ + vitamin E	3.90 ± 0.29^{d}	68.07 ± 0.20^{c}	85.70 ± 0.17^{d}	5.30 ± 0.12^{c}	86.20 ± 0.12^{c}
PHZ + AEMS 200 mg/kg	15.10 ± 0.06^{b}	76.40 ± 0.17^{b}	143.90 ± 0.12^{b}	12.50 ± 0.06^{b}	99.70 ± 0.35^{b}
PHZ + AEMS 400 mg/kg	7.20 ± 0.12^{c}	66.00 ± 0.17^{d}	116.50 ± 0.12^{c}	5.20 ± 0.12^{c}	86.20 ± 0.12^{c}

Legend as Table 1. Na, sodium, K, potassium, Cl, chloride

PHZ raised serum TBARS 7.6-fold, catalase 8.6-fold and GSH 1.8-fold (Table 7). AEMS lowered TBARS by 40% (200 mg/kg) and 45% (400 mg/kg), matching vitamin E, though values stayed above normal. Catalase and GSH fell to normal-range values at both doses; at 400 mg/kg GSH fell below normal.

Table 7: Effect of AEMS On Serum Oxidative Stress Markers

Group	Catalase (U/mL)	GSH (µmol/L)	TBARS (µmol/L MDA eq.)
Normal control	2.43 ± 0.01^{c}	46.34 ± 1.02^{c}	1.53 ± 0.19^{d}
PHZ control	20.87 ± 3.50^{a}	83.69 ± 4.25^{a}	11.68 ± 0.30^{a}
PHZ + vitamin E	4.29 ± 1.49^{c}	49.82 ± 0.53^{bc}	6.51 ± 0.16^{c}
PHZ + AEMS 200 mg/kg	7.36 ± 0.13^{bc}	50.41 ± 1.03^{bc}	7.01 ± 0.17^{b}
PHZ + AEMS 400 mg/kg	5.35 ± 0.71^{c}	38.27 ± 1.66^{d}	6.46 ± 0.16^{c}

Legend as Table 1. GSH, reduced glutathione; MDA, malondialdehyde; TBARS, thiobarbituric acid-reactive substances.

Discussion

Phenylhydrazine (PHZ) is a well-established model of haemolyticanaemia. Its auto-oxidation generates reactive oxygen species (ROS), including superoxide anions, hydrogen peroxide, and phenylhydrazyl free radicals. These oxidizehaemoglobin, promote Heinz body formation, and initiate lipid peroxidation and protein oxidation in the erythrocyte membrane, which ultimately leads to premature erythrocyte destruction (Aloke *et al.*, 2021). Consistent with this mechanism, PHZ administration significantly ($p < 0.05$) reduced red blood cell (RBC) count, haemoglobin (HGB) concentration, and haematocrit (HCT) compared with the normal control (Table 2). These findings agree with previous reports in PHZ-treated rats (Umaru *et al.*, 2018; Kasang*et al.*, 2020; Abiodun *et al.*, 2026).

Treatment with the aqueous stem bark extract of Myrianthus serratus (200 and 400 mg/kg bw) significantly ($p < 0.05$) increased RBC, HGB, and HCT compared with the PHZ control, in a dose-dependent manner. At 400 mg/kg bw, and in the vitamin E group, HCT rose above the normal control value, suggesting robust stimulation of erythropoiesis during recovery. A comparable overshoot of packed cell volume and haemoglobin above healthy control values was reported with *Telfairiaoccidentalis* leaf extract (Abiodun *et al*., 2026). Similar haematinic effects have also been reported for *Tapinanthusglobiferus* (Asogwa *et al*., 2024), *Ficuscapensis* (Njoku-Oji *et al*., 2016), *Ocimumgratissimum*(Dasofunjo*et al*., 2020), and raw and boiled leaves of *Adansoniadigitata* (Hamzah *et al*., 2022). This restoration of RBC, HGB and HCT indicates recovery of blood oxygen-carrying capacity and withdrawal of the cardiovascular compensation imposed by anaemic hypoxia, while the supraphysiological HCT most plausibly reflects rebound reticulocytosis during recovery rather than a sustained increase in red cell mass. The close correspondence between the highest extract dose and vitamin E suggests that polyphenolic constituents act principally by protecting the erythrocyte membrane against phenylhydrazine-induced lipid peroxidation and hemichrome formation, with direct erythropoietic stimulation, provision of haematopoietic substrates and hepatoprotection as additional, non-exclusive contributors. These observations are potentially relevant to human disorders in which oxidative erythrocyte injury shortens red cell survival, notably G6PD deficiency, sickle cell disease and malaria-associated

haemolysis, which are prevalent in the region. Extrapolation remains provisional, however, given the greater erythropoietic reserve of rodents, the self-limiting nature of this model, and the absence of standardisation, pharmacokinetic and toxicity data.

PHZ administration significantly ($p < 0.05$) increased mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) relative to the normal control. The rise in MCV reflects macrocytosis, most likely resulting from compensatory reticulocytosis: large, immature erythrocytes are released into the circulation in response to haemolysis. This pattern has been observed previously in PHZ-treated rats (Abiodun *et al.*, 2026). Treatment with 200 and 400 mg/kg bw of the extract significantly reduced MCV, MCH, and MCHC toward control values. This normalization suggests improved erythrocyte maturation and differentiation, and a shift from a stress-driven to a steady-state erythropoietic profile (Mél*et al.*, 2025). Comparable results have been reported for the aqueous root bark extract of *Psorospermumfebrifugum* (Félicienne *et al.*, 2019) and the aqueous extract of *Cocos nucifera* (Tchogou*et al.*, 2016).

PHZ significantly ($p > 0.05$) increased total white blood cell (WBC), lymphocyte and neutrophil counts compared with the normal control. This indicates an inflammatory and immune response to oxidative erythrocyte injury and the clearance of haemolytic debris. The finding is consistent with Abiodun *et al.* (2026), who reported concurrent elevation of leukocytes and hepatic interleukin-6 in PHZ-treated rats. Both vitamin E and the extract reduced WBC and lymphocyte counts, suggesting that the treatments attenuated the underlying inflammatory stimulus. This immunomodulatory effect may reflect the antioxidant properties of the extract, which could limit ROS-driven immune activation; the constituents responsible remain to be identified. Similar findings have been reported for *Justicia secunda* (Fagbohoun*et al.*, 2022), *Justicia carnea* (Okorie *et al.*, 2025) and *Telfairia occidentalis* (Abiodun *et al.*, 2026).

The liver is central to xenobiotic detoxification, protein synthesis, and bilirubin conjugation and excretion. The elevated serum aspartate aminotransferase (AST) and alanine aminotransferase (ALT) activities in PHZ-treated rats indicate hepatocellular injury. This is plausibly mediated by PHZ-derived ROS and by the hepatic burden of haem and iron released during haemolysis. Similar elevations, accompanied by degenerated and pyknotic hepatocytes, have been reported in this model (Abiodun *et al.*, 2026). The extract significantly reduced both enzymes, indicating a hepatoprotective effect.

PHZ also raised total and direct bilirubin levels, consistent with accelerated haem catabolism and with previous findings in PHZ-induced anaemic rats (Ayoade *et al.*, 2020). Treatment with the extract significantly lowered bilirubin relative to the PHZ control, reflecting reduced haemolysis and improved hepatic handling of bilirubin. Total protein increased significantly after PHZ administration. This contrasts with Zangeneh *et al.* (2019) and Ayoade *et al.* (2020), who reported decreased total protein in PHZ-treated rats. The discrepancy may reflect differences in PHZ dose, exposure duration or hydration status, since haemoconcentration can raise apparent serum protein levels. Administration of the extract restored total protein and albumin toward control values.

Serum urea and creatinine are established markers of renal function. Their elevation in the PHZ group indicates nephrotoxicity, likely arising from oxidative damage and from tubular exposure to haemoglobin and its breakdown products. This agrees with earlier reports (Ayoade *et al.*, 2020; Abiodun *et al.*, 2026). Vitamin E and the extract significantly ($p < 0.05$) reduced urea and creatinine, demonstrating an aMelilaiorative effect against PHZ-induced renal injury comparable to that reported for *Telfairia occidentalis* (Abiodun *et al.*, 2026). PHZ also disturbed serum sodium, potassium, and chloride concentrations, which suggests renal tubular dysfunction; potassium release from lysed erythrocytes may have contributed to the potassium changes. Both vitamin E and the extract significantly corrected these alterations relative to the PHZ control. Potassium and chloride concentrations at 200 and 400 mg/kg bw returned close to normal control values. Similar results have been reported for a combination of *Ficus capensis* and *Cnidoscolusaconitifolius* (Ezeigwe*et al.*, 2020) and for *Alchornea cordifolia* (Ezugwu *et al.*, 2022).

Oxidative stress arises when ROS generation exceeds the capacity of the body's enzymatic and non-enzymatic antioxidant defenses to remove them. Catalase (CAT) and reduced glutathione (GSH) are key components of these defenses, while malondialdehyde (MDA), a by-product of lipid peroxidation, serves as a marker of oxidative membrane damage. PHZ significantly increased MDA, confirming PHZ-induced lipid peroxidation, in agreement with previous studies (Ajibade *et al.*, 2017; Abiodun *et al.*, 2026). CAT activity and GSH levels also increased, contrasting with previous reports of CAT and GSH depletion following PHZ exposure (Luangaram*et al.*, 2007; Ajibade *et al.*, 2017; El-Shehry *et al.*, 2023; Abiodun *et al.*, 2026). The increase may represent an adaptive, compensatory upregulation of antioxidant defenses in response to sustained oxidative challenge. The divergence may reflect differences in PHZ dosing regimen, sampling time, or the tissue analyzed. Time-course studies would be needed to determine whether this response precedes eventual depletion.

Treatment with 200 and 400 mg/kg bw of the extract and with vitamin E returned all three parameters toward normal control values. This indicates a reduced oxidative burden, so that the compensatory antioxidant response was no longer required. This suggests the extract has radical-scavenging activity, possibly against hydroxyl and peroxyl radicals; direct *in vitro* antioxidant assays are needed to confirm this. Antioxidant-mediated protection in PHZ-induced anaemia has been reported for *Beta vulgaris* (Gheith & El-Mahmoudy, 2018), *Mucuna poggei* and *Telfairia occidentalis* (Udeozor *et al.*, 2022). These observations support the view that natural antioxidants stabilize cell membranes by terminating radical chain reactions and limiting oxidative damage (Da Pozzo *et al.*, 2018).

CONCLUSION

The findings indicate that the aqueous stem bark extract of *Myrianthus serratus* reverses PHZ-induced haemolyticanaemia. It restores erythrocyte parameters, normalizes erythrocyte indices, attenuates inflammation, and protects the liver and kidneys from oxidative injury, with effects comparable to those of vitamin E. These results provide pharmacological support for the traditional use of *Myrianthus serratus* in the management of anaemia.

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