

Evaluation Of Justicia Carnea-Enriched Zobo Drink On Haematological And Histological Parameters Of Iron Deficiency Anaemia-Induced Albino Rats

Orisa, Catherine A, Anyika-Elekeh J.U, Ezenwa Happiness C, Josiah-Orisa, Princess-Christabel T

Department Of Home Science And Management, Rivers State University, Port Harcourt, Nigeria.

Department Of Human Nutrition And Dietetics, Michael Okpara University Of Agriculture, Umudike

Department Of Obstetrics And Gynaecology, Rivers State University Teaching Hospital, Port Harcourt

Abstract

Iron deficiency anaemia (IDA) remains a major public health concern particularly in low- and middle-income countries. This study evaluated the haematological and histological effects of *Justicia carnea*-enriched zobo drink (*Hibiscus sabdariffa*) on phenylhydrazine-induced anaemia in male albino rats. Six blend ratios (A: 100% zobo; B: 63% zobo/37% *J. carnea*; C: 58/42%; D: 55/45%; E: 42/58%; F: 46/54%) were formulated using the Pearson Square method. The proximate composition, physicochemical properties, mineral content, iron bio-accessibility, sensory characteristics, serum mineral levels, haematological parameters, lipid profiles, and organ histopathology (liver, kidney, stomach) were determined. Results showed that fortification significantly increased protein, carbohydrate and iron content in a dose-dependent manner. Iron bio-accessibility improved from 42.5% (control) to 74.6% (Group E). Phenylhydrazine induction caused marked reductions in haemoglobin (Hb), packed cell volume (PCV) and red blood cell (RBC) counts, and elevated white blood cell (WBC) counts. Treatment with fortified drinks restored haematological indices in a dose-dependent manner, with Groups E and F achieving near-complete recovery (Hb: 13.8 g/dL; PCV: 41.0%; RBC: $7.3 \times 10^6/\mu\text{L}$). Lipid profiles improved significantly, with reductions in total cholesterol and triglycerides alongside increased HDL-C, particularly in higher-fortified groups. Histopathological scores of liver, kidney, and stomach confirmed severe damage post-induction, which was dose-dependently ameliorated following treatment. Sensory evaluation indicated good consumer acceptability, particularly for sample C. These findings demonstrate the nutraceutical potential of *J. carnea*-fortified zobo as a functional, culturally acceptable dietary strategy for managing IDA and associated organ damage.

Keywords: *Justicia carnea*, zobo drink, iron deficiency anaemia, haematology, histopathology

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I. Introduction

Iron deficiency anaemia (IDA) is a widespread nutritional disorder characterised by a decrease in haemoglobin concentration and red blood cell count due to insufficient iron stores. It affects millions of individuals globally, with particularly high prevalence in developing countries where access to nutrient-rich diets is limited (Kassebaum, 2016; WHO, 2021). In Nigeria, IDA is attributed to inadequate dietary iron intake, poor absorption, increased iron requirements during pregnancy and childhood, and chronic blood loss (Kumar *et al.*, 2022; Khalafallah, 2017). The consequences of IDA are far-reaching, manifesting as fatigue, impaired cognitive function, reduced work productivity, and increased susceptibility to infections (Camaschella, 2019). In children and pregnant women, it contributes to developmental impairment, preterm birth, and maternal mortality.

Fortification of commonly consumed food and beverages has emerged as an effective public health strategy to combat micronutrient deficiencies. Zobo drink, derived from the dried calyces of *Hibiscus sabdariffa*, is a popular traditional Nigerian beverage rich in anthocyanins, vitamin C, and antioxidants. These constituents may enhance non-haem iron absorption and have demonstrated erythropoietic potential in experimental models (Chukwu *et al.*, 2018; Umoren *et al.*, 2020). However, the iron content of unfortified zobo drink does not meet the recommended daily allowance, necessitating nutritional enhancement strategies.

Justicia carnea (blood leaf) is a perennial shrub indigenous to tropical Africa, traditionally used in Nigeria as a blood tonic for managing anaemia. Empirical studies have confirmed its haematinic potential, attributing its bioactivity to iron, folates, flavonoids, alkaloids, and antioxidant compounds (Ezeokonkwo *et al.*, 2020; Andrew *et al.*, 2024). Aqueous leaf extract of *J. carnea* has also demonstrated hepatoprotective, nephroprotective, anti-inflammatory, and antioxidant properties in preclinical models (Chukwu *et al.*, 2020; Okonkwo and Ezeonu, 2021). Despite these promising findings, the combined effect of *J. carnea* enrichment of

zobo drink on haematological parameters and organ histology in an experimental anaemia model remains largely unexplored.

This study therefore aimed to evaluate the effects of *J. carnea*-enriched zobo drink on proximate composition, physicochemical properties, mineral content, iron bio-accessibility, sensory properties, haematological indices, lipid profiles, and organ histopathology of phenylhydrazine-induced anaemic male albino rats. The findings are expected to provide scientific validation for the use of this indigenous functional beverage as a dietary intervention for managing IDA in resource-limited settings.

II. Materials And Methods

Sample Collection and Preparation

Dark-red *Hibiscus sabdariffa* calyces were obtained from Mile 3 Market, Port Harcourt, while fresh *Justicia carnea* leaves were harvested from the Rivers State University School Farm, Port Harcourt, Rivers State, Nigeria. Fresh *J. carnea* leaves (220 g) were sorted, washed in running water, boiled in 3.7 L of water for 30 minutes at 100°C, then filtered using muslin cloth and stored at 4°C. Dried roselle calyces (300 g) were rinsed, boiled in 1,600 mL of water at 80-90°C for 10-15 minutes, cooled, filtered, and refrigerated at 4°C.

Formulation of Blends

Blends were formulated using the Pearson Square method (Nnam *et al.*, 2023) to meet the recommended daily allowance for iron (7-15 mg; NIH, 2023). Six samples were prepared: A (100% zobo, control), B (63% zobo/37% *J. carnea*), C (58/42%), D (55/45%), E (42/58%), and F (46/54%). All formulations maintained a consistent total volume of 1,600 mL.

Chemical Analyses

Proximate composition (moisture, protein, fat, ash, and carbohydrate) was determined using standard AOAC (2023) methods. The Anthrone reagent method (2022) was used for carbohydrate determination. Physicochemical properties including total soluble solids (°Brix), total solids, and pH were measured by standard procedures (AOAC, 2023). Mineral content (Mg, Zn, Na, K, Ca, Fe, I) was determined by atomic absorption spectrophotometry (AAS) using the Buck Scientific Model 210 after dry ashing at 500°C. Iron bio-accessibility was assessed by an *in vitro* simulated gastrointestinal digestion method (Sulaiman *et al.*, 2022) involving gastric (pepsin, pH 2.0) and intestinal (pancreatin-bile) digestion stages.

Sensory Evaluation

Sensory properties (taste, colour, flavour, mouthfeel, and overall acceptability) were assessed by 20 semi-trained panellists using a 9-point hedonic scale, where 1 = disliked extremely and 9 = liked extremely (ISO 8586:2023; Mehle *et al.*, 2020).

Animal Studies

Thirty male albino rats (58-109 g) from the Animal House, Rivers State University, were acclimatised for 14 days under standard housing conditions. Anaemia was induced by intraperitoneal administration of phenylhydrazine (PHZ, 0.1 mL/kg body weight, 5% dilution in distilled water) on two consecutive days with a three-day interval. Rats were assigned to six groups (n=5/group, Groups A-F) and administered 1.5 mL per 100 g body weight of the respective blend daily for 14 days. All procedures complied with institutional animal care protocols (American Veterinary Medical Association, 2020). Haematological parameters [haemoglobin (Hb), packed cell volume (PCV), red blood cell (RBC), white blood cell (WBC) counts] were determined at baseline, after induction, and at the end of treatment using standard haematological methods (Cheesbrough, 2019). Serum mineral elements (Fe, Zn, Ca, Na, K) were quantified by AAS and flame photometry. Lipid profiles (total cholesterol, triglycerides, HDL-C) were assessed using standard enzymatic colorimetric methods.

Histopathological Evaluation

Liver, kidney, and stomach tissues were excised post-sacrifice, fixed in 10% neutral buffered formalin, processed by standard paraffin-embedding techniques, sectioned at 2 microns, and stained with haematoxylin and eosin (H&E) (Bancroft and Gamble, 2019). Histopathological scoring was conducted on a 0-3 scale (0 = normal; 3 = severe damage).

Statistical Analysis

Data were expressed as mean \pm standard deviation (SD). One-way ANOVA was performed using Minitab version 18.0, and Fisher's Least Significant Difference (LSD) post hoc test was applied to separate means. Statistical significance was set at $p < 0.05$.

III. Results And Discussion

Proximate Composition

The proximate composition of the zobo/*Justicia carnea* blends is presented in Table 1. Moisture content ranged from 77.82% (sample C) to 84.43% (sample A), with higher *J. carnea* inclusion reducing moisture content ($p < 0.05$). These values align with the 81.61-86.95% reported by Adelekan *et al.* (2014) for fruit-enhanced zobo drinks. Protein content ranged from 0.26% to 0.85%, with Sample B recording the highest value, indicating that moderate *J. carnea* inclusion augments protein content. Fat was consistently low (0.06-0.54%), consistent with the low-lipid nature of herbal beverages (Okoye *et al.*, 2020). Ash content showed no significant differences among samples ($p > 0.05$), ranging from 0.50 to 0.69%. Carbohydrate content ranged from 14.14% to 21.26%, with Sample C recording the highest value, suggesting that *J. carnea* contributed additional soluble solids to the blends.

Table 1: Proximate Composition of Zobo/*Justicia carnea* Blends (%)

Nutrients (%)	A	B	C	D	E	F
Moisture	84.43 ^a ±0.32	83.92 ^{ab} ±0.27	77.82 ^c ±0.03	80.94 ^{cd} ±0.31	81.65 ^c ±0.01	82.66 ^b ±0.08
Protein	0.31 ^d ±0.01	0.85 ^a ±0.02	0.26 ^d ±0.01	0.55 ^c ±0.02	0.57 ^{bc} ±0.01	0.62 ^b ±0.01
Fat	0.27 ^a ±0.01	0.54 ^c ±0.00	0.06 ^d ±0.00	0.16 ^b ±0.00	0.12 ^c ±0.00	0.15 ^b ±0.01
Ash	0.69 ^a ±0.00	0.55 ^a ±0.06	0.60 ^a ±0.01	0.58 ^a ±0.10	0.50 ^a ±0.00	0.60 ^a ±0.00
Carbohydrate	14.30 ^{ab} ±0.02	14.14 ^c ±0.03	21.26 ^a ±0.04	17.77 ^d ±0.02	17.16 ^d ±0.01	15.97 ^a ±0.05

Values are means ± SD. Means in the same row with different superscripts are significantly different ($p < 0.05$). A=100% zobo; B=63%/37%; C=58%/42%; D=55%/45%; E=42%/58%; F=46%/54% (zobo/*J. carnea*).

Physicochemical Properties

Total soluble solids ranged from 14.10 °Brix (sample D) to 16.75 °Brix (sample E), with higher *J. carnea* inclusion increasing soluble solid content due to bioactive phytochemical contributions (Okudu *et al.*, 2024). Total solids increased from 15.58 mg/L (control) to 22.18 mg/L (sample C). pH values ranged from 3.07 to 3.28, showing a gradual increase with *J. carnea* inclusion, consistent with its buffering effect (Ajayi and Oyerinde, 2020). The pH range remained acidic enough to ensure short-term microbial stability while reducing the tartness of the base drink.

Table 2: Physicochemical Properties of Zobo/*Justicia carnea* Blends

Physicochemical Property	A	B	C	D	E	F
Total Soluble Solids (°Brix)	14.95 ^b ±0.07	14.85 ^b ±0.07	15.15 ^b ±0.07	14.10 ^a ±0.14	16.75 ^a ±0.07	14.95 ^b ±0.07
Total Solids (mg/L)	15.58 ^c ±0.32	19.39 ^b ±0.27	22.18 ^a ±0.03	19.06 ^{bc} ±0.31	18.35 ^a ±0.01	17.34 ^d ±0.08
pH	3.07 ^c ±0.03	3.12 ^b ±0.02	3.15 ^b ±0.01	3.16 ^b ±0.01	3.24 ^a ±0.01	3.28 ^a ±0.02

Values are means ± SD. Means in the same row with different superscripts are significantly different ($p < 0.05$). Sample keys as in Table 1.

Mineral Composition

Mineral composition varied significantly ($p < 0.05$) across samples (Table 3). Iron content increased progressively from 2.32 mg/100g (control) to 7.77 mg/100g (sample E), confirming the iron-rich nature of *J. carnea* decoction. Similar enhancements have been reported when *J. carnea* was incorporated into food products (Alade *et al.*, 2023; Lawal *et al.*, 2022). Potassium increased with higher *J. carnea* inclusion (5.97-7.11 mg/100g), supporting cardiovascular health. Calcium and sodium decreased with higher *J. carnea* inclusion, possibly due to competitive divalent cation interactions (Olagunju *et al.*, 2021). Magnesium and zinc showed variable trends, with moderate inclusion (sample D) yielding the highest values for both minerals.

Table 3: Mineral Composition of Zobo/*Justicia carnea* Blends (mg/100g)

Minerals	A	B	C	D	E	F
Magnesium(mg/100g)	21.38 ^a ±0.01	19.05 ^{ab} ±0.01	21.48 ^a ±0.01	23.52 ^a ±2.82	15.11 ^b ±0.01	19.00 ^{ab} ±0.00
Zinc (mg/100g)	0.83 ^b ±0.01	0.12 ^c ±0.00	0.07 ^c ±0.00	0.92 ^a ±0.01	0.71 ^c ±0.01	0.63 ^d ±0.01
Sodium (mg/100g)	24.41 ^a ±0.01	24.00 ^a ±0.01	24.14 ^b ±0.01	24.39 ^a ±0.01	20.00 ^c ±0.01	19.87 ^d ±0.01
Potassium (mg/100g)	5.97 ^a ±0.01	6.11 ^c ±0.01	6.07 ^d ±0.01	6.01 ^d ±0.01	7.11 ^a ±0.01	7.09 ^b ±0.01
Calcium (mg/100g)	2.81 ^a ±0.01	2.11 ^f ±0.01	2.30 ^e ±0.01	2.74 ^b ±0.01	2.14 ^e ±0.01	2.16 ^d ±0.01
Iron (mg/100g)	2.32 ^c ±0.01	3.18 ^d ±0.01	3.33 ^c ±0.01	5.10 ^b ±0.01	7.77 ^a ±0.01	5.12 ^b ±0.01

Values are means ± SD. Means in the same row with different superscripts are significantly different ($p < 0.05$). Sample keys as in Table 1.

Sensory Properties

Taste ratings ranged from 6.20 to 8.50, with Sample C scoring highest. Colour scores were comparable across samples (7.00-7.90), indicating that *J. carnea* addition did not significantly alter the characteristic red pigmentation of zobo (Eze *et al.*, 2021). Flavour improved with *J. carnea* inclusion (5.80-7.67), and overall

acceptability ranged from 6.20 to 7.80. Samples C, E, and F exceeded the 7.0 hedonic threshold considered satisfactory in consumer studies (Grafiati, 2021), indicating that the blends would be well accepted in consumer markets.

Table 4: Sensory Evaluation of Zobo/*Justicia carnea* Blends (9-point hedonic scale)

Sensory Parameter	A	B	C	D	E	F
Taste	6.20 ^b ±2.15	7.20 ^{ab} ±1.69	8.50 ^a ±0.71	7.50 ^{ab} ±1.27	7.60 ^{ab} ±1.17	8.10 ^{ab} ±1.73
Colour	7.44 ^a ±2.19	7.00 ^a ±1.70	7.90 ^a ±1.19	7.20 ^a ±1.62	7.20 ^a ±1.32	7.10 ^a ±1.37
Flavour	5.80 ^a ±1.55	6.60 ^a ±1.51	7.60 ^a ±1.07	7.20 ^a ±1.23	7.40 ^a ±1.71	7.67 ^a ±1.58
Mouthfeel	7.10 ^a ±1.45	7.10 ^a ±1.66	7.90 ^a ±1.20	6.60 ^a ±1.90	7.00 ^a ±1.56	7.30 ^a ±1.64
Overall Acceptability	6.40 ^a ±2.41	6.20 ^a ±2.15	7.80 ^a ±1.23	6.90 ^a ±1.29	7.60 ^a ±1.43	7.70 ^a ±1.64

Values are means ± SD. Means in the same row with different superscripts are significantly different (p<0.05). Sample keys as in Table 1.

Serum Mineral Levels in Experimental Rats

PHZ induction caused a sharp decline in serum Fe (baseline: 124-127 mg/dL; post-induction: 47-49 mg/dL), Zn (90-93 to 61-63 mg/dL), and Ca (9.1-9.4 to 6.0-6.2 mg/dL), confirming successful anaemia induction (Table 5). Post-treatment, recovery was dose-dependent. Groups E and F (highest *J. carnea*) achieved near-baseline Fe (118-120 mg/dL), Zn (90 mg/dL), and Ca (9.0-9.1 mg/dL) levels. The ascorbic acid and organic acids in zobo are known to enhance non-haem iron absorption (Kim *et al.*, 2011), and the synergistic action of both plants likely protected minerals from forming insoluble complexes during digestion.

Table 5: Serum Iron (Fe), Zinc (Zn), and Calcium (Ca) Levels in Male Albino Rats (mg/dL)

Group	Stage	Fe (mg/dL)	Zn (mg/dL)	Ca (mg/dL)
A	Baseline	125 ± 4 ^a	92 ± 3 ^a	9.2 ± 0.3 ^a
	After Induction	48 ± 3 ^c	62 ± 2 ^c	6.1 ± 0.2 ^c
	End of Treatment	95 ± 4 ^b	82 ± 3 ^b	8.1 ± 0.3 ^b
B	Baseline	126 ± 3 ^a	91 ± 4 ^a	9.3 ± 0.2 ^a
	After Induction	47 ± 2 ^c	61 ± 3 ^c	6.0 ± 0.2 ^c
	End of Treatment	105 ± 3 ^b	85 ± 3 ^b	8.5 ± 0.2 ^b
C	Baseline	124 ± 4 ^a	90 ± 3 ^a	9.1 ± 0.3 ^a
	After Induction	49 ± 3 ^c	63 ± 2 ^c	6.2 ± 0.3 ^c
	End of Treatment	110 ± 4 ^b	87 ± 3 ^b	8.7 ± 0.3 ^b
D	Baseline	127 ± 4 ^a	93 ± 3 ^a	9.4 ± 0.3 ^a
	After Induction	48 ± 2 ^c	62 ± 3 ^c	6.1 ± 0.2 ^c
	End of Treatment	116 ± 3 ^b	89 ± 2 ^b	8.9 ± 0.2 ^b
E	Baseline	126 ± 3 ^a	92 ± 4 ^a	9.3 ± 0.2 ^a
	After Induction	47 ± 3 ^c	61 ± 3 ^c	6.0 ± 0.3 ^c
	End of Treatment	120 ± 4 ^b	90 ± 3 ^b	9.1 ± 0.3 ^b
F	Baseline	125 ± 4 ^a	91 ± 3 ^a	9.2 ± 0.3 ^a
	After Induction	48 ± 2 ^c	62 ± 2 ^c	6.1 ± 0.2 ^c
	End of Treatment	118 ± 3 ^b	90 ± 2 ^b	9.0 ± 0.2 ^b

Values are means ± SD. Means in the same group with different superscripts are significantly different (p<0.01). Group keys as in Table 1.

Iron Bio-Accessibility

In vitro iron bio-accessibility increased progressively from 42.5% (control) to a peak of 74.6% (Group E), then slightly declined in Group F (72.8%) (Table 6). The lower bio-accessibility of the control is consistent with the inhibitory effects of polyphenols and tannins in *H. sabdariffa* on non-haem iron absorption (Abioye *et al.*, 2017; Afify *et al.*, 2021). The close parallel between *in vitro* bio-accessibility and *in vivo* mineral recovery validates the formulation strategy, consistent with the Bioavailability and Bioaccessibility Theory (Grasso *et al.*, 2023).

Table 6: In Vitro Bio-Accessibility of Iron in Fortified Zobo Drinks

Group	Bio-Accessible Iron (%)
A (100% zobo, control)	42.5 ± 1.5 ^f
B (63% zobo + 37% <i>J. carnea</i>)	55.3 ± 1.8 ^c
C (58% zobo + 42% <i>J. carnea</i>)	61.4 ± 2.0 ^d
D (55% zobo + 45% <i>J. carnea</i>)	68.2 ± 1.9 ^c
E (42% zobo + 58% <i>J. carnea</i>)	74.6 ± 2.1 ^a
F (46% zobo + 54% <i>J. carnea</i>)	72.8 ± 2.0 ^{ab}

Values are means ± SD. Means with different superscripts are significantly different (p<0.01). Group keys as in Table 1.

Haematological Parameters

PHZ administration caused approximately 51-54% reduction in Hb (14.1-14.5 to 6.6-6.9 g/dL), ~50% reduction in PCV (42-43.5 to 21.0-22.0%), and ~58-62% decline in RBC counts (7.4-7.7 to 2.9-3.1 x10⁶/μL), confirming haemolytic anaemia induction (Table 7). WBC counts doubled (6.2-6.6 to 12.7-13.1 x10³/μL), reflecting PHZ-induced inflammatory responses. Following treatment, haematological recovery was dose-dependent. Groups E and F achieved near-baseline Hb (13.8 and 13.6 g/dL), PCV (41.0 and 40.5%), and RBC counts (7.3 and 7.2 x10⁶/μL), while WBC normalised across all groups. These findings corroborate the erythropoietic potential of *J. carnea* reported by Ezeokonkwo *et al.* (2020) and Okonkwo and Ezeonu (2021). The anti-inflammatory properties of *H. sabdariffa* and *J. carnea* likely contributed to WBC normalisation (Montalvo-González *et al.*, 2022).

Table 7: Haematological Parameters of Male Albino Rats (Mean ± SD)

Group	Stage	Hb (g/dL)	PCV (%)	RBC (x10 ⁶ /μL)	WBC (x10 ³ /μL)
A	Baseline	14.2 ± 0.5 ^a	42.0 ± 1.5 ^a	7.5 ± 0.3 ^a	6.5 ± 0.4 ^b
	After Induction	6.8 ± 0.4 ^c	22.0 ± 1.2 ^c	3.0 ± 0.2 ^c	12.8 ± 0.6 ^a
	End of Treatment	11.5 ± 0.5 ^b	36.0 ± 1.3 ^b	6.2 ± 0.3 ^b	7.8 ± 0.4 ^b
B	Baseline	14.3 ± 0.4 ^a	43.0 ± 1.4 ^a	7.6 ± 0.3 ^a	6.4 ± 0.5 ^b
	After Induction	6.7 ± 0.3 ^c	21.5 ± 1.1 ^c	2.9 ± 0.2 ^c	12.7 ± 0.7 ^a
	End of Treatment	12.0 ± 0.4 ^b	37.0 ± 1.2 ^b	6.5 ± 0.2 ^b	7.2 ± 0.4 ^b
C	Baseline	14.1 ± 0.6 ^a	42.5 ± 1.3 ^a	7.4 ± 0.3 ^a	6.6 ± 0.5 ^b
	After Induction	6.9 ± 0.4 ^c	22.0 ± 1.3 ^c	3.1 ± 0.3 ^c	13.0 ± 0.5 ^a
	End of Treatment	12.5 ± 0.5 ^b	38.0 ± 1.2 ^b	6.7 ± 0.2 ^b	7.0 ± 0.3 ^b
D	Baseline	14.4 ± 0.5 ^a	43.2 ± 1.5 ^a	7.6 ± 0.3 ^a	6.3 ± 0.4 ^b
	After Induction	6.6 ± 0.3 ^c	21.0 ± 1.2 ^c	3.0 ± 0.2 ^c	12.9 ± 0.5 ^a
	End of Treatment	13.2 ± 0.4 ^b	39.5 ± 1.3 ^b	7.0 ± 0.3 ^b	6.8 ± 0.3 ^b
E	Baseline	14.5 ± 0.4 ^a	43.5 ± 1.3 ^a	7.7 ± 0.3 ^a	6.2 ± 0.4 ^b
	After Induction	6.8 ± 0.3 ^c	21.8 ± 1.1 ^c	3.1 ± 0.2 ^c	13.1 ± 0.6 ^a
	End of Treatment	13.8 ± 0.5 ^b	41.0 ± 1.4 ^b	7.3 ± 0.2 ^b	6.5 ± 0.3 ^b
F	Baseline	14.3 ± 0.5 ^a	43.0 ± 1.4 ^a	7.6 ± 0.3 ^a	6.4 ± 0.5 ^b
	After Induction	6.7 ± 0.4 ^c	21.5 ± 1.2 ^c	3.0 ± 0.2 ^c	12.8 ± 0.6 ^a
	End of Treatment	13.6 ± 0.4 ^b	40.5 ± 1.3 ^b	7.2 ± 0.2 ^b	6.6 ± 0.3 ^b

Values are means ± SD. Means in the same group with different superscripts are significantly different (p<0.01). Group keys as in Table 1.

IV. Conclusion

This study demonstrated that fortification of zobo drink with *Justicia carnea* decoction significantly improved its nutritional, functional, and therapeutic potential in a phenylhydrazine-induced anaemia model. Fortification enhanced protein, carbohydrate, and iron content in a dose-dependent manner, with iron bio-accessibility reaching 74.6% in Group E. Haematological parameters, including Hb, PCV, and RBC counts, were markedly restored following treatment, with Groups E and F achieving near-complete recovery. Overall, *J. carnea*-enriched zobo drink represents a promising, affordable, and culturally acceptable nutraceutical intervention for managing iron deficiency anaemia and associated metabolic and organ complications in resource-limited settings.

Conflict Of Interest

The authors declare no conflict of interest.

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