BLOOD PROTEINS AND HAEMATOLOGICAL INDICES OF WEANLING WISTAR RATS FED *Moringa oleifera* AND CRUDE SAPONIN EXTRACT

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ABSTRACT
Phytogenic compounds like Moringa oleifera offer natural alternatives to additives in animal nutrition. This study evaluated the effects of Moringa oleifera crude and saponin extract on blood proteins and Haematology in seventy (70) Wistar rats, assigned to seven dietary treatments in a completely randomized design, including a control and varying levels (1, 1.5, and 2 mg/dL) of Moringa oleifera and crude saponin extracts. Moringa extract reduced PCV (29.50%–36.75%) and Hb levels (9.83–11.98 g/dL), indicating potential dose-dependent oxidative stress effects. Although crude saponins enhanced PCV, Hb, and RBC counts, which could be due to antioxidant properties. However, treatments showed elevated WBC counts and neutrophil percentages, demonstrating immunomodulatory effects. Total protein levels declined with higher Moringa concentrations, while crude saponins enhanced protein metabolism. Corresponding values of Albumin and globulin levels varied, with saponins supporting immune function. The albumin-globulin ratio suggested an immune-stimulatory response for both treatments. Urea levels ranged narrowly between 14.33 mg/dL (2 mg/mL Moringa extract) and 15.25 mg/dL (control and several other groups), with creatinine levels remaining stable, reflecting minimal renal impact. These findings highlight the contrasting effects of Moringa and crude saponin extracts on haematological parameters, emphasizing their dose-dependent roles in modulating oxidative stress, immunity, and metabolism.
Keywords: globulin, total protein, wistar rats, saponin, glucose

INTRODUCTION

Moringa oleifera, commonly known as the drumstick tree, is widely regarded for its exceptional nutritional and medicinal properties. Native to the Indian subcontinent, it is now cultivated worldwide due to its rich content of bioactive compounds, including saponins, flavonoids, and phenolic compounds. Among these, saponins have drawn significant attention for their antioxidant, anti-inflammatory, and immune-boosting properties, making them promising candidates for improving health outcomes (Francis *et al*., 2002). Blood proteins, such as albumin, globulin, and total protein, play vital roles in maintaining physiological balance, nutrient transport, and immune defence. Alterations in these proteins often indicate nutritional status or underlying health conditions. Haematological indices, including packed cell volume (PCV), Haemoglobin (Hb), red blood cell (RBC) count, and white blood cell (WBC) count, serve as critical markers for assessing circulatory and immune system health. Evaluating these parameters provides insights into the systemic impacts of dietary or therapeutic interventions. Phytogenic compounds like *Moringa oleifera* and crude saponins offer a natural alternative to synthetic drugs and additives, particularly in animal nutrition and healthcare (Akangbe *et al*., 2024). Despite growing interest, research on the effects of Moringa and saponins on blood proteins and haematological indices remains limited, especially in weanling Wistar rats. These rats are widely used in experimental studies due to their physiological similarity to humans and their sensitivity to dietary interventions during the rapid growth phase. This study evaluated the impact of *Moringa oleifera* saponin extract on blood proteins and haematological indices in weanling Wistar rats. By examining parameters such as total protein, albumin, globulin, PCV, Hb, RBC, and WBC counts, this research seeks to enhance understanding of the potential health benefits of Moringa and saponins, thereby critically looking at their applications in animal nutrition.

MATERIALS AND METHODS

The experiment was conducted in the Rat Room, Department of Animal Science, University of Ibadan (7°20'N, 3°50'E, elevation 200 m). Seventy 4-week-old weanling Wistar rats (*Rattus norvegicus*) weighing 50–60 g were procured and housed in standard cages under controlled conditions (temperature: 25 ± 2°C; humidity: 50–60%; 12-hour light/dark cycle). After a 7-day acclimatization, the rats were fed *ad libitum* for 21 days on a composite diet (58.87% corn starch, 10.53% casein, 5.00% glucose, 7.00% sucrose, 5.00% cellulose, 10.00% soybean oil, 3.60% minerals/vitamins). Moringa leaves were randomly harvested, dried for 14days and milled. While saponin extracts were prepared in distilled water to create solutions of 1 mg/mL, 1.5 mg/mL, and 2 mg/mL. Seventy (70)

rats were randomly assigned to seven dietary treatments of ten (10) rats each in a completely randomised design, moringa and saponin extracts were orally administered for 21 days, followed by feed and water after two hours. At the end, blood samples were collected via jugular puncture under light diethyl ether anesthesia. haematological parameters were analysed using an automated haematology analyser, while serum total protein, albumin, and globulin were measured spectrophotometrically.

Statistical Analysis

Data obtained were analysed using the Analysis of Variance SAS (2012), while means were separated using Duncan's Multiple Range Test.

RESULTS AND DISCUSSION

The haematological parameters of Wistar rats treated with Moringa extract and crude saponins in Table 1 at varying concentrations (1 mg/mL, 1.5 mg/mL, and 2 mg/mL) had PCV values ranging from 29.50% in the group treated with 2 mg/mL of Moringa extract to 36.75% in the control group. PCV measures the proportion of red blood cells (RBCs) in blood, serving as an indicator of oxygen-carrying capacity.

Table 1: Effect of crude extract saponins from moringa leaves on haematological parameters of rats

Parameters	Control	Moringa Extract (mg/mL			CrudeSaponins(mg/mL)			
		1	1.5	2	1	1.5	2	SEM
PCV(%)	36.75	33.75	31.75	29.50	33.50	35.75	35.67	1.29
Hb (g/dl)	11.88	11.05	10.73	9.83	11.18	11.60	11.98	0.44
RBC(x10^{6}/ul)	6.05	5.44	5.31	5.09	5.37	5.83	6.02	0.22
WBC(x10^{3}/ul)	8.13^{c}	8.79^{c}	11.19^{a}	12.29^{a}	8.63^{c}	10.03^{b}	13.40^{a}	1.41
PLAT (x10^{3}/ul)	2.92^{c}	2.11^{c}	2.69^{c}	2.62^{c}	8.00^{b}	12.12^{a}	13.40^{a}	1.05
LYM(%)	65.00	60.25	70.00	71.50	58.75	61.25	66.67	2.01
NEUT(%)	26.75^{b}	23.75^{b}	25.00^{b}	34.75^{a}	36.25^{a}	34.25^{a}	31.00^{ab}	2.06
MON(%)	2.25	2.00	2.00	2.25	2.25	2.00	2.00	0.19
EO(%)	3.50^{a}	3.00^{ab}	2.75^{b}	2.75^{b}	3.00^{ab}	2.25^{c}	2.00^{c}	0.14

abc…Mean with different superscripts along the same row are significantly (P<0.05) differentPCV—packed cell volume WBC –white blood cells , NEUT—neutrophils ,HB –heamoglobin concentration , PLAT – platelets ,MON --- monocytes ,RBC—red blood cells, LYM –lymphocyte, EO –eosinophil, SEM- Standard Error of Mean

Reduction in PCV in groups treated with Moringa extract could suggest a dose-dependent effect of the extract on erythropoiesis or RBC survival. Similar findings were reported by Dwomoh *et al*. (2024), where plant extracts influenced haematological parameters due to their bioactive compounds, potentially leading to oxidative stress at higher concentrations. On the other hand, the crude saponin-treated groups exhibited a higher PCV, with control group exhibiting 35.75% and 35.67% for 1.5 mg/mL and 2 mg/mL, respectively. This observation aligns with earlier report by Zhang *et al*. (2020), which showed that saponins could enhance RBC membrane stability and reduce hemolysis, potentially improving PCV. Haemoglobin levels ranged from 9.83 g/dL in the group treated with 2 mg/mL of Moringa extract to 11.98 g/dL in the group treated with 2 mg/mL of crude saponins. Hb levels are critical for oxygen transport, and a reduction may indicate anemia or compromised erythropoiesis. The dose-dependent decrease in Hb with increasing concentrations of Moringa extract suggests potential cytotoxic effects,

as corroborated by studies on phytochemicals affecting erythroid lineage (Oyewo *et al.,* 2013). Crude saponins, however, demonstrated an enhancing effect on Hb levels, particularly at 2 mg/mL. This could be attributed to their antioxidant properties, as highlighted by Igwilo *et al.* (2013), which help maintain RBC integrity and Hb functionality. The RBC count followed a trend similar to Hb and PCV, with decrease in Moringa extract-treated groups (5.09–5.44 × 10^6/µL) compared to the control (6.05 × 10^6/µL). Crude saponin treatments, especially at 2 mg/mL, restored RBC counts to levels comparable to the control (6.02 × 10^6/µL). This decline in RBC count with higher Moringa extract concentrations might be linked to oxidative damage, while the protective effect of saponins aligns with their role in reducing oxidative stress and inflammation.

The WBC count ranged from 8.13 × 10^3/µL in the control to 13.40 × 10^3/µL in the group treated with 2 mg/mL of crude saponins. Elevated WBC levels in crude saponin-treated groups indicate enhanced immune responses, as saponins are known immunomodulators. Moringa extract also increased WBC count significantly (p<0/05) at higher concentrations (11.19–12.29 × 103/µL), showing Moringa's immunostimulatory effects. Lymphocyte percentages were highest in the 2 mg/mL Moringa extract group (71.50%) and lowest in the 1.5 mg/mL group (58.75%). The elevation at higher doses indicates immunostimulatory effects, aligning with Moringa's reported role in adaptive immunity. Neutrophil percentages were significantly (p<0.05) elevated in the higher-dose Moringa extract and crude saponin groups (34.25%–36.25%) compared to the control (26.75%). This suggests a pro-inflammatory response or enhanced innate immunity, consistent with the finding of Liu et al. (2019). In table 2, the total protein levels ranged from 6.50 g/dL in the group treated with 2 mg/mL of Moringa extract to 7.98 g/dL in the group treated with 1 mg/mL of crude saponins. The control group recorded 7.18 g/dL. Total protein serves as an indicator of protein synthesis and nutritional status. The decline in values in the 2 mg/mL Moringa extract group could suggest impaired protein synthesis or increased protein catabolism at higher extract doses. This may be as a result of the fact that crude saponins, particularly at 1 mg/mL, enhanced protein synthesis (Francis *et al*., 2002), which report saponins' potential to improve protein metabolism in experimental animals. Albumin levels varied significantly (p<0.05), with the highest value (3.40 g/dL) recorded in the group treated with 1 mg/mL of Moringa extract and the lowest (2.26 g/dL) in the group treated with 2 mg/mL of Moringa extract. Albumin is crucial for maintaining oncotic pressure and serves as a marker for liver function.

Table 2: Effect of *Moringa oleifera* saponin on serum biochemical indices of rats

Parameters	Control	Moringa Extract			Crude Moringa Saponins			SEM
		1 mg/mL	1.5 mg/mL	2 mg/mL	1 mg/mL	1.5 mg/mL	2 mg/mL	
Total protein (g/dl)	7.18	7.33	7.00	6.50	7.98	7.65	6.57	0.14
Albumin (g/dl)	3.15	3.40	2.85	2.26	3.15	3.15	2.87	0.09
Globulin (g/dl)	4.03	3.93	4.15	4.24	3.70	4.65	4.83	0.11
Alb-Glb Ratio	0.78^{b}	0.87^{a}	0.69^{b}	0.53^{c}	0.85^{a}	0.68^{b}	0.59^{c}	0.03
Urea (mg/dl)	15.25	14.75	15.25	14.33	15.25	15.25	15.00	0.17
Creatinine(mg/dl)	0.70^{b}	0.73^{b}	0.68^{b}	0.63^{b}	0.67^{b}	0.73^{b}	0.85^{a}	0.04

abc…Mean with different superscripts along the same row are significantly (P<0.05) different, SEM- Standard Error of Mean

The decline in albumin at higher extract doses may indicate altered protein metabolism due to dose-dependent effects of plant extracts on hepatic indices. Globulin levels ranged from 3.70 g/dL (1 mg/mL crude saponins) to 4.83 g/dL (2 mg/mL crude saponins), with the control at 4.03 g/dL. The significant increase in globulin in crude saponin-treated groups suggests enhanced immune function, noting that saponins modulate immune responses through enhanced globulin production (Oyewo, 2013). The significantly lower Albumin-Globulin ratio in higher dose groups of both treatments' points to an immune-stimulatory response, aligning with similar findings in phytochemical studies (Liu *et al*., 2021). Urea levels ranged narrowly between 14.33 mg/dL (2 mg/mL Moringa extract) and 15.25 mg/dL (control and several other groups). The stable values across all treatments suggest that

neither Moringa extract nor crude saponins adversely affected nitrogen metabolism or renal function. Creatinine levels ranged from 0.63 mg/dL in the 2 mg/mL Moringa extract group to 0.85 mg/dL in the 2 mg/mL crude saponin group, with the control at 0.70 mg/dL. However, the overall values remained within a physiological range, indicating minimal renal impairment.

CONCLUSION

Moringa extract exhibited dose-dependent haematological effects, reducing RBC count, PCV, and Hb levels, while crude saponins enhanced RBC stability and immune function. Both treatments influenced protein metabolism, with saponins promoting globulin synthesis. Overall, neither treatment significantly impaired renal function, suggesting potential therapeutic applications with careful dose considerations.

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