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Haematological, Serum Biochemical and Oxidative Stress Profiles in Yankasa Sheep with Naturally Acquired *Haemonchus contortus* Infection

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ABSTRACT

Haemonchus contortus remains the primary cause of economic loss in small ruminants production, often manifesting as the severe clinical state known as Parasitic Bottled Jaw Syndrome (PBJS). This study investigated haematological, serum biochemical, and oxidative stress profiles in Yankasa sheep naturally infected with *H. contortus* to better understand the systemic pathogenesis of the disease. A total of Twenty-eight sheep were examined and used for this study. The subjects were categorized into two groups: eighteen sheep clinically diagnosed with PBJS and ten healthy serving as the control group. Clinical assessment of the PBJS group revealed a high prevalence of submandibular osigns oedema (100%), diarrhoea (89%), and pale mucous membranes (72%). Laboratory analysis indicated significant physiological shifts; sheep with PBJS exhibited macrocytic hypochromic anaemia, alongside marked hypoproteinaemia, hypoalbuminaemia, hypoglycaemia. Furthermore, renal function markers were affected evidenced by elevated blood urea nitrogen (BUN) and creatinine concentrations. A critical finding indicates the presence of systemic oxidative stress characterized by a significant increase in malondialdehyde (MDA) levels and a concurrent reduction in glutathione (GSH). These findings underscore that oxidative stress is a primary component in the pathogenesis of PBJS in Yankasa sheep. The results suggest that MDA and GSH may serve as valuable biomarkers for assessing the severity of the condition. Consequently, effective management in endemic areas should transition toward an integrated approach that combines standard parasite control with antioxidant supplementation to improve clinical outcomes.

Keywords: Hypoproteinaemia; Nigeria; Oxidative damage; Ovine, Parasitic bottled jaw syndrome

INTRODUCTION

Sheep production is a cornerstone of food security and rural livelihoods across sub-Saharan Africa, providing essential protein and income for millions of backyard farmers. However, gastrointestinal parasites remain a primary barrier to maximizing the productivity of this flocks. Among these parasites, the abomasal nematode

Haemonchus contortus is particularly devastating due to its voracious blood feeding pattern (Kimeli *et al*., 2025). This parasite causes both acute and chronic blood loss, which characteristically manifests as severe anaemia, hypoproteinaemia and the hallmark clinical sign of submandibular oedema, commonly referred to as "bottled jaw" (Kandil *et al*., 2025).

In Nigeria, the impact of haemonchosis is profound, with over 80% of traditionally managed flocks affected by parasitic infestations. The Yankasa sheep, being the most populous and widely distributed breed in the country, often bears the major brunt of this parasitic burden. While the classical symptoms of anaemia and protein loss are well documented in veterinary literature, the underlying biochemical shifts particularly those involving oxidative stress remain insufficiently explored in naturally infected indigenous African breeds (Alade & Bwala, 2013; Bello . *et al*., 2025).

The pathogenesis of haemonchosis extends beyond simple blood loss. The inflammatory response and subsequent haemolysis triggered by the parasite lead to the excessive generation of reactive oxygen species (ROS). These ROS can overwhelm the host's antioxidant defenses, resulting in the oxidative damage to cellular membranes, exacerbating tissue injury and further impairing the animal's immune response (Orrico *et al*., 2023). While experimental models have provided some insights into oxidative stress during infection, there is a critical lack of data regarding these markers in naturally acquired infection in Yankasa sheep.

Bridging this knowledge gap is essential for improving clinical management. Therefore, this study aimed to evaluate the haematological, serum biochemical, and oxidative stress profiles in Yankasa sheep presenting with naturally acquired Parasitic Bottled Jaw Syndrome (PBJS). By identifying these specific physiological alterations, we seek to uncover novel diagnostic biomarkers and potential therapeutic targets to improve the health and survival of sheep in endemic regions.

MATERIALS AND METHODS

Study Area and Experimental Animals

This study was conducted in March 2024 at a commercial small ruminant farm located in Ilorin, Kwara, State, Nigeria. A total of twenty-eight (n = 28) adult Yankasa sheep were purposely selected for the study. The cohort was divided into two groups: eighteen (n = 18) sheep presenting with clinical Parasitic Bottled Jaw Syndrome (PBJS) and ten (n = 10) clinically healthy sheep which served as the control group. All experimental procedures were conducted in strict accordance with the ethical guidelines provided by the University of Ilorin (Approval No: URE/FVM/2023/019).

Clinical and Parastilogical Examination

PBJS cases were identified through a standardized clinical assessment focusing on pathognomonic signs such as submandibular oedema, pale ocular mucous membranes and poor body condition. To confirm the presence and etiology of the infection, faecal samples were collected directly from the rectum of each animal. These samples were processed using the sodium chloride flotation technique for the identification of helminth ova. *Haemonchus contortus* eggs were morphologically identified based on their characteristic ellipsoidal shape and morulated internal structure (Figure 1).

Figure 1. Verification of *Haemonchus contortus* infection in Yankassa sheep
Caption: Photomicrograph showing standard faecal float slide from a Yankassa sheep presenting with clinical Parasitic Bottled Jaw Syndrome (PBJS). Multiple ellipsoidal, morulated helminth ova are visible morphologically confirmed as *Haemonchus contortus*. The identification of these eggs serve to verify the natural aetiology of the PBJS cases analyzed throughout this study for haematological, serum biochemical and oxidative stress shifts.
Note: Scale bar = 10 µm. Magnification verified at x 400 [40x objective]

Blood Collection and Processing

Approximately 5 mL of whole blood was collected from each sheep via jugular venipuncture. For haematological analysis, 2 mL was dispensed into tubes containing Ethylenediaminetetraacetic acid (EDTA). The remaining 3 mL was collected in plain tubes for serum extraction. The plain tube samples were allowed to clot at room temperature before being centrifuged at 3000 x g for 10 minutes. The resulting serum was aspirated and stored in sterile cryovials at −20°C until biochemical and oxidative stress assays were performed.

Haematological and Serum Biochemical Analyses
Haematological parameters, including complete blood counts, were determined using a Mindray BC-2800 automated analyser (Mindray, China). Serum biochemical analyses were performed using a Roche Cobas c111 analyser. The parameters quantified were: Protein profile (total protein and albumin), Metabolic marker (glucose), Renal function (blood urea nitrogen (BUN) and creatinine) and Liver enzymes (alanine aminotransferase (ALT), alkaline phosphatase (ALP), and gamma-glutamyl transferase (GGT)). All analyses utilized high-quality commercial reagent from Randox Laboratories (Crumlin, UK). .

Oxidative Stress Assays
Oxidative stress status was evaluated through the quantification of lipid peroxidation and antioxidant capacity. Malondialdehyde (MDA) concentrations were measured using the thiobarbituric Acid Reactive Substances (TBARS) method via Elabscience ELISA kits (Houston, USA). Additionally, reduced glutathione (GSH) levels were quantified spectrophotometrically using specialized assay kits from the same manufacturer. All procedures followed the manufacturer's established protocols.

Statistical Analysis
All data were processed and analyzed using GraphPad Prism 9.0 (GraphPad Software, San Diego, CA, USA). Continuous variables (vital signs, haematology, serum biochemistry and oxidative stress markers) are expressed as mean ± standard error of the mean (SEM) and were compared between groups using an unpaired Student's t-test. Categorical clinical signs were expressed as frequencies and percentages and were evaluated using Fisher's exact test. A P value < 0.05 was considered statistically significant.

RESULTS
Clinical Manifestations
The clinical presentation of Yankasa sheep naturally infected *H. contortus* was characterized by a high prevalence of classic haemonchosis symptoms. All subjects in the PBJS group exhibited submandibular oedema (100%) followed by high incidence of diarrhoea (89%) and dehydration (83%). Pale ocular mucous membranes indicative of severe anaemia was observed in 72% of the affected animals. However, despite these severe clinical symptoms, vital signs did not show significantly significant differences between the PBJS and control groups suggesting a state of chronic compensation (Table 1).

Table 1. Clinical presentation and physiological parameters in Yankassa sheep with naturally acquired *Haemonchus contortus* infection (PBJS) compared to healthy controls.

Parameter	Control (n = 10)	*H. contortus* infected (n =18)	*P*-value
Clinical Signs (% affected)			
Submandibular oedema ("bottled jaw")	0 (0%)	18 (100%)	< 0.001*
Pale mucous membrane	0 (0%)	13 (72%)	< 0.001*

Dehydration (skin tent >3)	0 (0%)	15 (83%)	< 0.001*
Diarrhoea	0 (0%)	16 (89%)	< 0.001*
Poor body condition score (≤2/5)	0 (0%)	12 (67%)	< 0.01*
Anorexia	0 (0%)	9 (50%)	< 0.05*
Vital Parameters (Mean ± SEM)			
Rectal Temperature (°C)	36.9±0.1	37.2±0.2	> 0.05*
Heart rate (beats/min)	81.0±1.6	82.8±1.8	> 0.05*
Respiratory rate (cycles/min)	23.1±0.91	23.5±0.9	> 0.05*

Note: Asterisks values indicate statistically significant differences ($P < 0.05$) *

Haematological Alterations

Haematological analysis revealed profound systemic changes in the PBJS group, notably severe macrocytic hypochromic anaemia which was evidenced by significantly lower levels of haemoglobin (Hb) concentration, packed cell volume (PCV), total red blood cell (RBC) counts, and mean corpuscular haemoglobin concentration (MCHC). Conversely, a significant increase in mean corpuscular volume (MCV) was recorded supporting the macrocytic classification of the anaemia (Figure 2, Table 2). Additionally, the PBJS group displayed marked leukocytosis and eosinophilia reflecting the host's robust inflammatory and immunological response to the helminth burden (Figure 3).

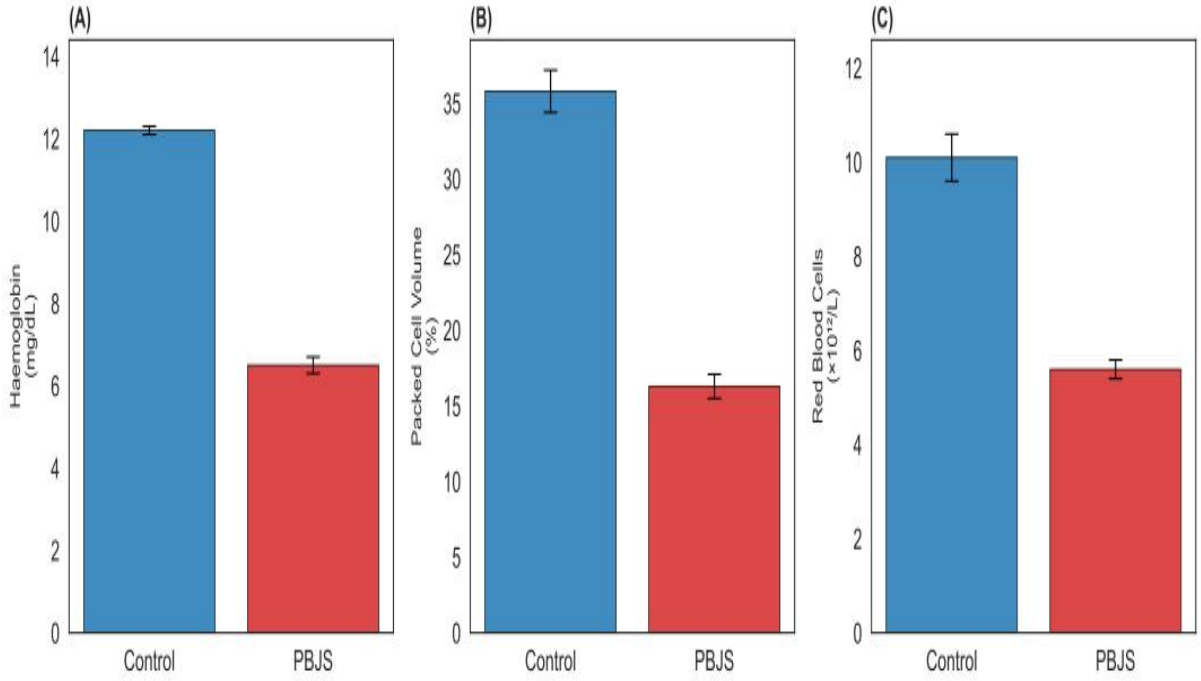

Figure 2. Erythroid parameters of healthy control and *Haemonchus contortus*-infected Yankassa sheep.

Caption: Comparison of erythroid profiles between healthy control sheep (n = 10) and Yankassa sheep with naturally acquired *Haemonchus contortus* infection presenting as Parasitic Bottled Jaw Syndrome (PBJS, n = 18). Values represent mean ± SEM for: (A) haemoglobin, (B) packed cell volume, and (C) red blood cell counts. Note: conventional veterinary units for haemoglobin are g/dL (Panel A). P < 0.001 indicates significant differences from the control group.

Table 2. Haematological indices of Yankasa sheep with naturally acquired *Haemonchus contortus* infection (PBJS) compared to healthy controls.

Parameter	Control (n = 10)	*H. contortus* infected (n =18)	*P*-value
Mean corpuscular volume (fL)	33.4 ± 0.4	40.4 ± 1.1	< 0.01*
Mean corpuscular haemoglobin concentration (g/dL)	32.3 ± 0.5	30.0 ± 0.6	< 0.05*
Neutrophils (x 10^9/L)	3.7 ± 0.3	3.9 ± 0.4	> 0.05*
Lymphocytes (x 10^9/L)	4.8 ± 0.1	5.3 ± 0.7	> 0.05*
Monocytes (x 10^9/L)	0.2 ± 0.1	0.3 ± 0.2	> 0.05*

Note: Asterisks values indicate statistically significant differences (*P* < 0.05) *

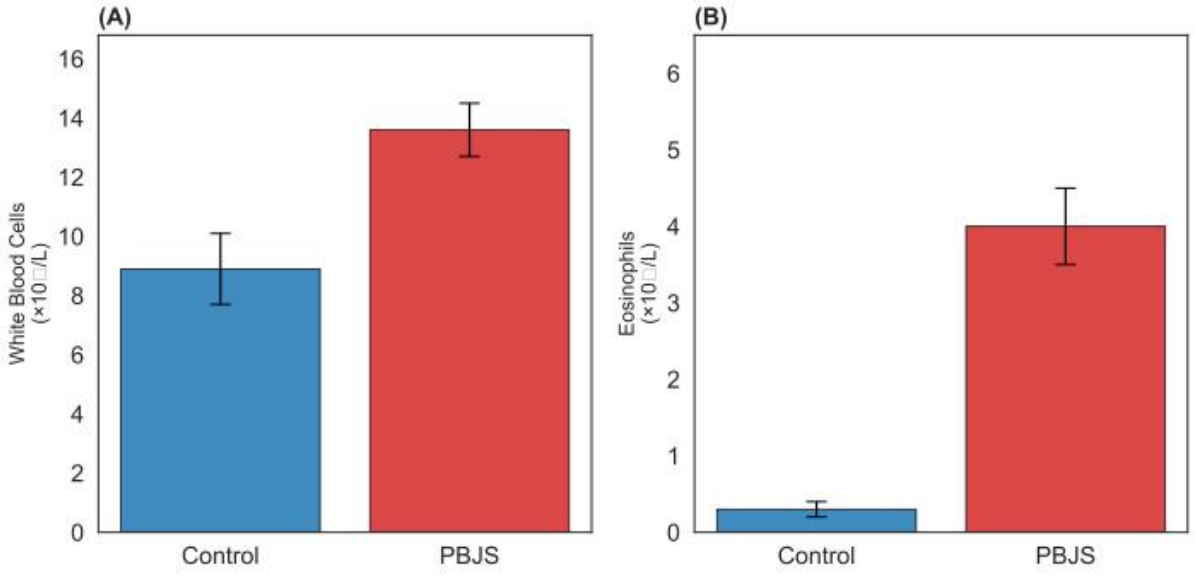

Figure 3. Leukocyte response in control and *Haemonchus contortus*-infected Yankassa sheep.
Caption: Leucogram changes (A) Total leukocyte (TWBC) count and (B) absolute eosinophil count. The graphs compare healthy control sheep (n = 10) and Yankassa sheep presenting with clinical Parasitic Bottled Jaw Syndrome (PBJS) due to

naturally acquired *H. contortus* infection (n = 18). Values represent the mean ± SEM for each parameter. The P-value of < 0.001 indicates statistical significance between the Control and PBJS groups for both leukocyte types.

Serum Biochemical Profiles

Significant metabolic and organ function shifts were observed in serum biochemistry of infected sheep. The PBJS group was characterized by protein imbalance evidenced by hypoproteinaemia and hypoalbuminaemia, energy deficit recorded as marked hypoglycaemia as well as profound renal impairment seen as elevated blood urea nitrogen (BUN) and creatinine. While activities of liver enzymes such as Aspartate aminotransferase (AST), Alanine aminotransferase (ALT) and gamma-glutamyl transferase (GGT) were mildly elevated, these changes did not reach statistical significance (Table 3).

Table 3. Serum Biochemical Profiles of Yankasa sheep with naturally acquired *Haemonchus contortus* infection (PBJS) compared to healthy controls.

Parameter	Control (n = 10)	*H. contortus* infected (n =18)	*P*-value
Total Protein (g/dL)	7.8 ± 0.2	5.5 ± 0.6	< 0.01*
Albumin (g/dL)	3.5 ± 0.1	2.3 ± 0.2	< 0.01*
Globulin (g/dL)	3.4 ± 0.2	3.2 ± 0.3	> 0.05*
Glucose (mg/dL)	64.0 ± 0.4	55.9 ± 1.1	< 0.05*
Blood urea nitrogen (mg/dL)	18.1 ± 1.5	23.9 ± 2.6	< 0.05*
Creatinine (mg/dL)	1.4 ± 0.1	2.6 ± 0.3	< 0.01*
Alanine aminotransferase (U/L)	32. ± 4.4	35.9 ± 5.1	> 0.05*
Alkaline phosphatase (U/L)	135.3 ± 11.4	142.7 ± 10.0	> 0.05*
Gamma-glutamyl transferase (U/L)	47. 9 ± 6.1	50.1 ± 8.5	> 0.05*

Note: Asterisks () values indicate statistically significant differences ($P < 0.05$).

Oxidative Stress Profile

The most critical finding regarding the systemic pathogenesis of PBJS was the confirmation of oxidative imbalance (P<0.001). Infected sheep exhibited significantly higher concentrations of malondialdehyde (MDA), a primary marker of lipid peroxidation. This was accompanied by a significant reduction in reduced glutathione (GSH) levels (Figure 4) confirming that naturally acquired haemonchosis induces severe systemic oxidative stress overwhelming the sheep endogenous antioxidant defense.

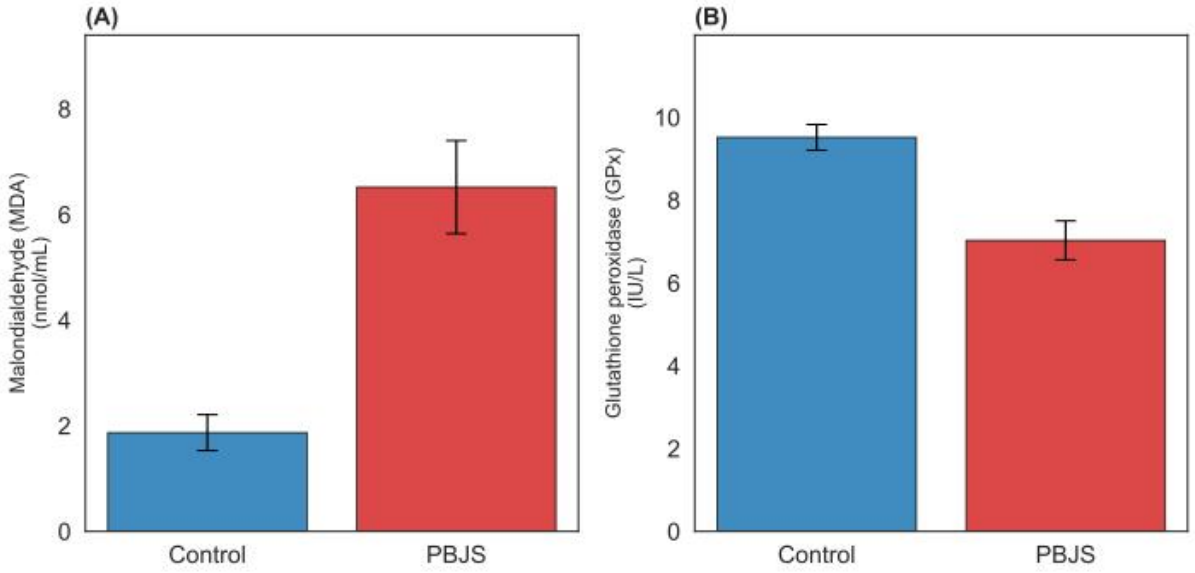

Figure 4. Comparative analysis of systemic oxidative stress markers in healthy and *Haemonchus contortus*-infected Yankassa sheep.
Caption: Indicators of oxidative status in Yankassa sheep. Bar graphs represent the mean ± Standard Error of the Mean (SEM) for (A) Malondialdehyde (MDA) concentration (nmol/mL) and (B) Reduced Glutathione (GSH) concentration (µmol/L). The Control group (n = 10) consists of clinically healthy sheep, while the PBJS group (n = 18) represents sheep with naturally acquired *H. contortus* infection presenting with Parasitic Bottled Jaw Syndrome. Statistical significance between groups was determined using an unpaired Student's t-test. The marked increase in MDA and depletion of GSH confirm a state of systemic oxidative imbalance in the infected subjects. Significance is indicated as: ***P < 0.001.

DISCUSSION

This study reinforces the role of *Haemochus contortus* as the primary etiological agent of Parasitic Bottled Jaw Syndrome (PBJS) in Yankasa sheep. The systemic impact of this infection is multi-faceted and characterized by a triad of severe anaemia, hypoproteinaemia, and significant oxidative stress.
The observed macrocytic hypochromic anemia is likely a consequences of nutrition imbalance secondary to chronic blood-feeding activity of the parasites. This regenerative attempt by the bone marrow coupled with iron and protein depletion underscores the physiological strain on the host. Furthermore, the marked leukocytosis and eosinophilia reflects a robust Th2-mediated immune response which is a hallmark of helminth infections as reported in recent literature (Stucke *et al.*, 2026).
Metabolic disturbances were evidenced by elevated BUN and creatinine levels which strongly suggest the presence of prerenal azotaemia related to dehydration. Additionally, mild increases ALT, AST, and GGT activities point toward subclinical hepatic stress possibly due to the metabolic burden of the infection or in part due to the localized inflammatory response (Allam *et al.*, 2020).
A central finding in this research is the pronounced oxidative imbalance in the infected Yankassa Sheep. Elevated concentration of malondialdehyde (MDA) serve as clear indicator of lipid peroxidation while the concurrent reduction in the reduced glutathione (GSH) levels highlights a depletion of the host's endogenous antioxidant defenses (Birben *et al.*, 2012). This study suggests that chronic inflammation,

haemolysis, hypoxia, and nutrient deficiencies contribute to the excessive production of reactive oxygen species (ROS). These factors likely impair immune modulation and exacerbate the severity of PBJS. The finding of this study aligns with previous report (Allam *et al.*, 2020), and suggest that oxidative stress is critical, yet overlooked component of the disease's pathogenesis.
Given the results, the inclusion of antioxidant supplementation such as Vitamin E, selenium and polyphenols could serve as a valuable adjunct to standard anthelmintic therapy. Such an integrated approach warrants further exploration through controlled clinical trials to improve recovery rates in endemic regions.

CONCLUSION

In Conclusion, Parasitic Bottled Jaw Syndrome in Yankasa sheep is inextricably linked to *H. contortus* infection, which induces a devastating combination of causing severe anaemia, hypoproteinaemia, renal stress, and systemic oxidative imbalance. Monitoring biomarkers such as MDA and GSH may provide clinicians with essential diagnostic and prognostic insights into the severity of the infection. To mitigate the disease's impact on small ruminant productivity, an integrated management strategy combining rigorous parasite control with targeted antioxidant supplementation should be adopted.

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CONFLICTS OF INTERESTS

The Authors declares no conflicts of interest

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