

## SERUM PROTEIN AND HAEMATOLOGICAL CHARACTERISTICS IN BOVAN BLACK LAYERS FED GRADED LEVELS OF ZINC SULPHATE MONOHYDRATE

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### ABSTRACT

Zinc-supplemented diets have been shown to enhance the immune status of laying birds, improving nutrition and overall health of laying birds. This study assessed the effects of zinc sulphate monohydrate supplementation on the haematology and serum proteins of laying birds. A total of 120 Bovan Black point-of-lay birds were grouped into a control (T1) and three treatment groups supplemented with 30 mg/kg (T2), 60 mg/kg (T3), and 90 mg/kg (T4) of zinc. Birds were monitored for 12 weeks, fed basal diets and provided ample drinking water ad libitum. Blood samples were collected at the end of the trial for haematological and serum biochemical analyses. Values obtained from T1 to T4 for Packed Cell Volume (27.65–29.87%), haemoglobin (9.47–9.72 g/dL), and erythrocyte counts ( $2.71–3.07 \times 10^6/L$ ) significantly ( $p < 0.05$ ) varied among treatments, with T4 exhibiting the highest values. Leukocyte counts ( $19.88–20.38 \times 10^6/L$ ) also differed significantly, with the highest count in T1. Significant ( $p < 0.05$ ) differences were observed in heterophils (28.63–33.25%), monocytes (2.99–3.96%), eosinophils (3.09–4.84%), and heterophil-to-lymphocyte ratios (0.48–0.55) across treatments. Serum protein, globulin, and albumin levels significantly ( $p < 0.05$ ) increased with zinc supplementation. These findings indicate that dietary zinc supplementation, thereby promoting productivity in laying birds.

**Keywords:** Bovan black, Zinc sulphate, Albumin, Globulin, Lymphocytes

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### INTRODUCTION

Recent studies have examined the effects of zinc supplementation on serum protein levels and haematological parameters in poultry. For instance, Gholizadeh *et al.* (2022) emphasized the impact of dietary zinc on production performance, egg quality, and blood parameters such as serum total protein and albumin concentrations in heat-stressed birds, finding that zinc supplementation influenced these parameters. Similarly, Sridhar *et al.* (2021) evaluated the effect of graded concentrations of organic zinc (zinc glycinate) on skin quality, haematological, and serum biochemical constituents in chickens, reporting alterations in serum total protein and globulin levels with varying zinc supplementation. It is worthy to note that significant changes in serum protein concentration with different dietary zinc levels may influence certain blood traits in poultry, in that zinc supplementation can significantly influence blood plasma cholesterol, protein, calcium, and phosphorus concentrations as the case may be. The impact of zinc on immune function is another critical aspect of its supplementation. Zinc deficiency in poultry can lead to compromised immune responses, making birds more susceptible to infections and diseases (Rath *et al.*, 2022). Zinc-supplemented diets have been shown to enhance the immune status of laying birds, reducing mortality rates and improving their overall health and resilience against pathogens. In addition to immune and production benefits, zinc also plays a role in reproductive health. Adequate zinc levels are essential for the proper development and function of reproductive organs, influencing laying performance and egg fertility (Niu *et al.*, 2021). These study highlighted the importance of zinc in modulating serum protein levels and haematological parameters in poultry, underscoring the need for further determine optimal zinc supplementation levels for different breeds and production conditions.

Materials and Methods

### Animals and Management

The experiment was carried out at the poultry unit of the teaching and research farm, Edo State College of Agriculture and Natural Resources, Iguoriakhi, Edo State. A total of 120 Bovan Black laying birds (22 weeks old) were randomly assigned to four dietary treatments, each containing 30 birds in a completely randomized design (CRD). The experimental diets were formulated to include varying levels of zinc: 0mg/kg (T1, control), 30mg/kg (T2), 60mg/kg (T3), and 90mg/kg (T4). Zinc was provided as zinc sulphate monohydrate, mixed thoroughly into the basal diet. The feeding trial lasted for eight weeks.

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### Blood Chemistry

Blood samples were collected via the jugular vein from five randomly selected birds per treatment at the end of the trial. Samples were transferred into anticoagulant-treated tubes for haematological analysis and plain tubes for serum biochemical assessments. Haematological parameters such as packed cell volume (PCV), haemoglobin concentration, red and white blood cell counts, and differential leukocyte counts were analysed using an automated haematology analyser. Serum biochemical parameters, including total protein, albumin, globulin, uric acid, and glucose, were determined using standard commercial kits and a spectrophotometer.

### Statistical analysis

Data were subjected to one-way analysis of variance (ANOVA) and means were separated using Duncan's Multiple Range Test at a 5% significance level (SAS, 2002).

### RESULT AND DISCUSSION

The haematological parameters evaluated in this study (Table1) provided essential insights into the physiological status of Bovan Black layers subjected to varying levels of dietary zinc supplementation.

**Table 1: Haematological indices Boran Black layers fed graded levels of zinc supplement of**

	T1	T2	T3	T4	SEM
Packed cell volume (%)	29.33	27.65	28.33	29.87	0.99
Haemoglobin (g/dL)	9.54	9.72	9.47	9.64	0.11
Erythrocytes (x 10 <sup>6</sup> /L)	3.00	2.71	2.75	3.07	0.18
Leukocytes (x10 <sup>8</sup> /L)	20.38	19.88	20.12	20.18	0.21
Thrombocytes (x10 <sup>7</sup> /L)	1.22	1.33	1.23	1.25	0.05
Lymphocyte (%)	59.33	60.68	60.49	61.3	0.82
Heterophils (%)	28.63 <sup>b</sup>	30.33 <sup>a</sup>	33.25 <sup>a</sup>	30.67 <sup>ab</sup>	0.41
Monocyte (%)	3.11 <sup>b</sup>	3.96 <sup>a</sup>	3.82 <sup>b</sup>	2.99 <sup>b</sup>	0.29
Eosinophils (%)	3.22 <sup>b</sup>	3.09 <sup>b</sup>	4.84 <sup>a</sup>	3.67 <sup>b</sup>	0.27
Basophils (%)	0.69	0.01	0.77	0.4	0.26
Heterophil: Lymphocyte Ratio	0.48 <sup>b</sup>	0.50 <sup>a</sup>	0.55 <sup>a</sup>	0.50 <sup>ab</sup>	0.50

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

Packed Cell Volume (PCV) ranged from 27.65% (T2) to 29.87% (T4), while haemoglobin (HB) values were between 9.47 g/dL (T3) and 9.72 g/dL (T2). Erythrocyte counts varied from 2.71 × 10<sup>6</sup>/L (T2) to 3.07 × 10<sup>6</sup>/L (T4), and leukocyte counts ranged from 19.88 × 10<sup>8</sup>/L (T2) to 20.38 × 10<sup>8</sup>/L (T1). These values remained within the normal physiological range, suggesting that zinc supplementation at these levels did not induce blood-related disorders (Gholizadeh *et al.*, 2021). Significant (p<0.05) differences were observed in heterophils (28.63% to 33.25%), monocytes (2.99% to 3.96%), eosinophils (3.09% to 4.84%), and heterophil-to-lymphocyte (H/L) ratios (0.48 to 0.55). The increase in heterophil and monocyte percentages in T2 and T3 highlights zinc's potential role in enhancing immune function, attributed to its modulatory effects on innate immunity (Yilmaz *et al.*, 2015). The stability of leukocyte and thrombocyte counts across treatments further reinforces the absence of adverse effects, indicating a well-maintained immune profile in all groups. The slight elevation in lymphocyte percentages in T4 may represent a compensatory mechanism to higher zinc levels, as lymphocytes are critical for adaptive immune responses (Rao *et al.*, 2013). Serum biochemical analysis revealed significant differences across treatments, particularly in total protein, albumin, globulin, glucose, and uric acid concentrations. Higher total protein levels observed in T2, T3, and T4 indicate zinc's positive influence on protein metabolism, potentially mediated by its role in enzyme activation and protein synthesis (Qi *et al.*, 2019). Elevated albumin levels in T2 suggest improved liver function and protein synthesis, while increased globulin levels in T2 to T4 align with enhanced immune function. These findings are consistent with studies in broiler chickens (Sridhar *et al.*, 2021; Ilaboya and Osayande, 2024) and Japanese quail (Aya *et al.*, 2021).

The reduction in glucose levels in T2 and T4 suggests a potential hypoglycemic effect of zinc, which may regulate energy metabolism and mitigate the risk of metabolic disorders. This aligns with the report that zinc influences insulin signaling and glucose homeostasis (Min *et al.*, 2019). The significant increase in uric acid concentrations in T2 to T4 reflects heightened protein metabolism and nitrogen excretion, a common consequence of zinc's

enzymatic activation. While these values were lower than those observed in laying hens in Nigeria (Ogbuewu and Mbajorgu, 2022) and local chickens in Tanzania (Khondowe et al., 2021), they exceeded values reported for broiler chickens fed snot apple powder (Jimoh *et al.*, 2025) and Wistar rats administered saponin extracts (Odukoya *et al.*, 2024). The findings of this study align with and extend previous research on zinc's dual role in supporting metabolic and immune functions in poultry. For example, increased globulin levels in T2 to T4 were consistent with enhanced immune responses, a hallmark of zinc supplementation. Similar trends have been reported in broiler chickens and Japanese quail, emphasizing zinc's universal benefits across poultry species (Aya *et al.*, 2021; Sridhar *et al.*, 2021). In contrast, the hypoglycemic effects observed in T2 and T4 contribute new insights into zinc's role in energy metabolism. The significant variations in antioxidant enzyme activities across treatments reinforce the need for precise zinc supplementation levels

**Table 2: Serum Protein of Boran Black layers fed graded levels of zinc supplement**

	T1	T2	T3	T4	SEM
Protein (g/dL)	29.62 <sup>b</sup>	34.99 <sup>a</sup>	33.49 <sup>a</sup>	34.33 <sup>a</sup>	1.19
Glucose(g/dl)	133.21 <sup>a</sup>	129.44 <sup>b</sup>	133.06 <sup>a</sup>	130.87 <sup>b</sup>	0.15
Globulin (g/dl)	1.68 <sup>b</sup>	1.90 <sup>a</sup>	1.93 <sup>a</sup>	1.96 <sup>a</sup>	0.04
Albumin (g/dL)	1.37 <sup>b</sup>	1.89 <sup>a</sup>	1.42 <sup>ab</sup>	1.49 <sup>ab</sup>	0.09

*abc...Mean with different superscripts along the same row are significantly ( $P < 0.05$ ) different; SEM- Standard Error of Mean*

## CONCLUSION

This study demonstrates the significant effects of graded levels of zinc supplementation on haematological parameters and serum biochemistry markers in Bovan Black layers. Moderate zinc levels (e.g., T2 and T3) enhance immune function, protein metabolism, and antioxidant defences, contributing to improved health and productivity. However, excessive zinc levels (e.g., T4) may disrupt antioxidant balance, underscoring the need for careful optimization of dietary zinc levels.

## RECOMMENDATION AND PRACTICAL APPLICATION

From a practical perspective, the findings of this study have several implications for poultry nutrition and management. The observed improvements in haematological and Serum protein parameters with moderate zinc sulphate monohydrate supplementation suggest that optimal zinc levels can enhance the health and productivity of Bovan Black layers. These benefits extend to improved immune function and better protein metabolism, which collectively contribute to the overall well-being and performance of laying birds. However, Producers and nutritionists should consider the specific requirements of their flocks and avoid indiscriminate supplementation, as excessive zinc levels can compromise health and productivity of laying birds.

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