



HAEMATOLOGICAL AND SERUM BIOCHEMISTRY OF BROILER BIRDS TREATED WITH SCENT LEAF (*Ocimum Gratissimum*).

By

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Abstract

*This study evaluated the haematological and serum biochemical responses of broiler chickens administered aqueous extracts of scent leaf, *Ocimum gratissimum*, through drinking water. A total of 120 day-old broiler chicks were randomly assigned to four treatment groups of 30 birds each, with three replicates of 10 birds per replicate in a completely randomised design (CRD). The treatments consisted of T1 (control; 0 mL), T2 (2 mL), T3 (4 mL), and T4 (6 mL) of *O. gratissimum* extract administered in drinking water. At the end of the feeding trial, data collected were subjected to one-way analysis of variance (ANOVA), and treatment means were separated using Duncan's multiple range test at a 5% probability level. Results were expressed as mean \pm standard error. The haematological indices showed significant ($p < 0.05$) differences among treatment groups. Packed cell volume increased from 23.50% in T1 to 26.33% in T4, haemoglobin concentration ranged from 11.30 to 11.90 g/dL, while red blood cell count increased from $2.66 \times 10^6/\text{mm}^3$ in T1 to $3.11 \times 10^6/\text{mm}^3$ in T4. Lymphocyte values increased from 55.00% in the control group to 62.00% in T4, whereas heterophil counts decreased from 37.50% to 31.33% across the same treatments. Serum biochemical parameters were also significantly ($p < 0.05$) influenced by the treatments. Total protein ranged from 2.60 to 3.36 g/dL, albumin from 1.13 to 1.47 g/dL, and globulin from 1.45 to 1.89 g/dL. Glucose concentrations increased from 191.00 mg/dL in T1 to 267.67 mg/dL in T4, while cholesterol and triglyceride values ranged from 105.39 to 142.18 mg/dL and 99.33 to 116.16 mg/dL, respectively. Electrolyte and mineral parameters remained within physiological ranges, with chloride values ranging from 128.97 to 135.16 mmol/L, sodium from 138.50 to 140.69 mmol/L, calcium from 6.74 to 6.87 mmol/L, phosphorus from 3.36 to 3.60 mmol/L, and potassium from 1.12 to 1.26 mmol/L. The findings indicate that aqueous extracts of *O. gratissimum* improved haematological indices, enhanced immune response, maintained serum biochemical stability, and supported mineral homeostasis in broiler chickens without adverse physiological effects. Therefore, *O. gratissimum* extract may serve as a beneficial natural phyto-genic additive in broiler production.*

Keywords: broiler chickens; *Ocimum gratissimum*; haematology; serum biochemistry; phyto-genic additives

INTRODUCTION

The poultry industry remains one of the fastest-growing sectors of animal agriculture and plays a significant role in meeting the increasing global demand for high-quality animal protein. Broiler chickens, in particular, constitute a major source of affordable meat for both developed and developing countries due to their rapid growth rate, high feed conversion

efficiency, and short production cycle. To maximize productivity and maintain flock health, poultry producers have traditionally relied on antibiotic growth promoters (AGPs) to improve growth performance, enhance feed efficiency, and reduce disease incidence. However, the prolonged and indiscriminate use of antibiotics in poultry production has raised serious public health concerns, particularly regarding antimicrobial resistance, drug residues in animal products, and



environmental contamination. Consequently, the ban or restriction of AGPs in several countries has intensified the search for safe, natural, and sustainable alternatives capable of improving poultry performance without adverse effects on animal or human health.

Among the promising alternatives are phytogetic feed additives derived from medicinal plants and herbs. Phytogetic additives have gained considerable attention due to their antimicrobial, antioxidant, anti-inflammatory, immunomodulatory, and digestive-enhancing properties. These bioactive compounds have been reported to improve nutrient utilization, gut health, growth performance, and disease resistance in poultry birds. One medicinal plant that has attracted increasing scientific interest is *Ocimum gratissimum*, commonly known as scent leaf or African basil.

Ocimum gratissimum is a perennial aromatic herb belonging to the family Lamiaceae and is widely distributed across tropical and subtropical regions, including West Africa and Nigeria. The plant is extensively used in traditional medicine and culinary practices due to its distinctive aroma and therapeutic properties. The leaves contain several biologically active compounds, including eugenol, thymol, flavonoids, alkaloids, tannins, saponins, and essential oils, which exhibit potent antimicrobial, antioxidant, antifungal, and anti-inflammatory activities. These phytochemicals contribute to the medicinal value of the plant and suggest its potential usefulness as a natural growth promoter and health-enhancing additive in poultry nutrition.

Previous studies have demonstrated the beneficial effects of *O. gratissimum* supplementation on the growth performance and health status of broiler chickens. Olumide and Akintola (2020) reported significant improvements in body weight gain and feed intake in broiler chickens fed diets supplemented with varying levels of *O. gratissimum* leaf meal. Similarly, Nte et al. (2024) observed enhanced growth performance and improved carcass traits in broiler chicks administered aqueous extracts of *O. gratissimum*. In another study, Osita et al. (2024) reported positive effects of dietary *O. gratissimum* leaf meal on growth performance and carcass characteristics of broiler chickens. These findings collectively suggest that *O. gratissimum* possesses considerable potential as a phytogetic feed additive in poultry production systems.

Despite the growing body of literature on the growth-promoting and physiological benefits of *O. gratissimum* in broiler production, information regarding its effects on tissue morphology and organ integrity remains limited. Histological evaluation is an important tool for assessing the influence of dietary interventions on cellular architecture, tissue functionality, and organ health, particularly within the gastrointestinal tract where nutrient digestion and absorption occur. Understanding the histological responses of broiler tissues to phytogetic supplementation could provide deeper insights into the mechanisms through which *O. gratissimum* exerts its beneficial effects.

Therefore, this study was designed to evaluate the effects of aqueous extracts of *O. gratissimum* on the haematological and serum biochemical parameters of broiler chickens, with a view to determining its suitability as a natural phytogetic additive for sustainable poultry production.

Materials and Methods

The treatment groups consisted of varying levels of aqueous extract of *Ocimum gratissimum* administered through drinking water as follows:

- i. **T1:** 0 mL aqueous scent leaf extract (control)
- ii. **T2:** 2 mL aqueous scent leaf extract
- iii. **T3:** 4 mL aqueous scent leaf extract
- iv. **T4:** 6 mL aqueous scent leaf extract

Sourcing and Preparation of *Ocimum gratissimum* Extract

Fresh leaves of *Ocimum gratissimum* were harvested from a local farm in Umudike, Abia State, Nigeria. The leaves were thoroughly washed with clean water to remove dirt and contaminants before being ground into a fine pulp using a mechanical blender. The resulting pulp was filtered through a sterile white muslin cloth to obtain the aqueous extract. The extract was collected into sterile containers and refrigerated at 4°C to preserve its bioactive constituents prior to use.

The prepared extract was administered daily through drinking water according to the designated treatment levels throughout the feeding trial.

Experimental Diet

The basal diet fed to the broiler chickens was formulated to meet the nutrient requirements of broilers as recommended by standard poultry nutrition guidelines. The ingredient composition of the diet is presented in Table 1.

Table 1. Composition of Experimental Diet for Broiler Chickens

Ingredient	Composition (%)
Yellow maize	32.72
Groundnut cake	36.71
Palm kernel meal	11.91
Wheat offal	14.87
Bone meal	3.00
L-lysine	0.10
DL-methionine	0.20
Salt	0.25
Vitamin-mineral premix	0.25
Total	100.00

Calculated Nutrient Composition

Nutrient	Value
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Nutrient	Value
Crude protein (%)	22.00
Metabolizable energy (kcal/kg)	2674

The vitamin-mineral premix supplied the following nutrients per 2.5 kg of premix: Vitamin A, 12,500,000 IU; Vitamin D3, 2,500,000 IU; Vitamin E, 40,000 mg; Vitamin K3, 2,000 mg; Vitamin B1, 3,000 mg; Vitamin B2, 5,500 mg; niacin, 55,000 mg; calcium pantothenate, 11,500 mg; Vitamin B6, 5,000 mg; Vitamin B12, 25 mg; choline chloride, 500,000 mg; folic acid, 1,000 mg; biotin, 80 mg; manganese, 120,000 mg; iron, 100,000 mg; zinc, 80,000 mg; copper, 8,500 mg; iodine, 1,500 mg; cobalt, 300 mg; selenium, 120 mg; and antioxidant, 120,000 mg.

Experimental Design

The experiment was arranged in a completely randomized design (CRD) comprising four treatment groups with three replicates per treatment. Each replicate contained 10 birds, resulting in a total of 120 experimental birds. The design enabled unbiased evaluation of the effects of aqueous *O. gratissimum* extract on the haematological and serum biochemical parameters of broiler chickens (Steel and Torrie, 1980).

Blood Sample Collection and Haematological Analysis

At the end of the feeding trial, three birds were randomly selected from each replicate for blood collection. Blood samples were collected via the wing vein using sterile syringes and needles. Samples intended for haematological analysis were transferred into labelled tubes containing ethylenediaminetetraacetic acid (EDTA) as an anticoagulant.

The haematological parameters evaluated included packed cell volume (PCV), haemoglobin concentration (Hb), red blood cell count (RBC), white blood cell count (WBC), lymphocyte count, and heterophil count. Standard laboratory procedures described by Jain (1986) were adopted for the analyses.

Serum Biochemical Analysis

Additional blood samples were collected into plain sample bottles without anticoagulants and allowed to clot. The samples were centrifuged at 3000 rpm for 10 min to obtain serum for biochemical analyses.

Serum total protein, albumin, and globulin concentrations were determined using commercial assay kits obtained from Sigma Chemical Company (St. Louis, Missouri, USA). Serum cholesterol concentration was determined using the enzymatic colorimetric method described by Kohn and Allen (1995), while serum glucose concentration was estimated using the orthotoluidine method. Electrolyte and mineral parameters were analyzed using standard laboratory procedures.

Statistical Analysis

All data generated from the study were subjected to one-way analysis of variance (ANOVA) using IBM SPSS Statistics

version 25.0. Treatment means were separated using Duncan's Multiple Range Test at a 5% level of significance (Steel and Torrie, 1980). Results were presented as mean \pm standard error of mean (SEM).

Results and Discussion

Effect of *Ocimumgratissimum* Supplementation on the Haematological Parameters of Broiler Chickens

The effects of aqueous extract of *Ocimumgratissimum* on the haematological parameters of broiler chickens are presented in Table 1.

Table 1. Effect of *Ocimumgratissimum* Supplementation on the Haematological Parameters of Broiler Chickens

Parameters	T1	T2	T3	T4	SEM
PCV (%)	23.50	24.75	25.00	26.33	0.44
Hb (g/dL)	11.30 ^b	11.65 ^{ab}	11.47 ^b	11.90 ^a	0.07
RBC ($\times 10^6/\text{mm}^3$)	2.66 ^b	2.90 ^{ab}	2.89 ^{ab}	3.11 ^a	0.05
TWBC ($\times 10^3/\text{mm}^3$)	20.98	21.91	22.10	23.15	0.43
MCV (fL)	88.51	85.46	86.59	84.77	1.12
MCH (pg)	42.59 ^a	40.29 ^{ab}	39.73 ^{ab}	38.28 ^b	0.54
MCHC (g/dL)	48.12	47.14	45.91	45.52	0.68

^{ab} Means within the same row bearing different superscripts differ significantly ($p < 0.05$). SEM = standard error of mean; Hb = haemoglobin; PCV = packed cell volume; RBC = red blood cell count; TWBC = total white blood cell count; MCV = mean corpuscular volume; MCH = mean corpuscular haemoglobin; MCHC = mean corpuscular haemoglobin concentration.

The haematological results revealed that dietary supplementation with aqueous *O. gratissimum* extract positively influenced some blood parameters of broiler chickens. Packed cell volume (PCV) values ranged from 23.50% in the control group (T1) to 26.33% in T4, although no significant ($p > 0.05$) differences were observed among the treatments. The PCV values obtained in this study fall within the normal physiological range reported for healthy broiler chickens, indicating that the treatments did not adversely affect erythropoiesis or oxygen transport capacity.

Haemoglobin concentration significantly ($p < 0.05$) increased in birds administered higher levels of scent leaf extract, with T4 recording the highest value (11.90 g/dL) compared to T1

(11.30 g/dL). Similarly, red blood cell (RBC) counts increased significantly ($p < 0.05$) from $2.66 \times 10^6/\text{mm}^3$ in the control group to $3.11 \times 10^6/\text{mm}^3$ in T4. The observed increase in Hb concentration and RBC count may be attributed to the erythropoietic and antioxidant properties of the phytochemicals present in *O. gratissimum*, particularly flavonoids and essential oils, which may enhance blood formation and improve physiological functions.

Total white blood cell (TWBC) counts showed no significant ($p > 0.05$) differences among the treatment groups, although numerical increases were observed with increasing supplementation levels. The absence of significant variation suggests that the aqueous extract did not induce pathological stress or inflammatory responses in the birds. Mean corpuscular volume (MCV) and mean corpuscular haemoglobin concentration (MCHC) were also not significantly affected by treatment, indicating stability in erythrocyte morphology and haemoglobin concentration within the red blood cells.

However, mean corpuscular haemoglobin (MCH) values decreased significantly ($p < 0.05$) as supplementation levels increased, with T1 recording the highest value and T4 the lowest. Despite this reduction, the values remained within physiological limits for broiler chickens, suggesting that the treatments did not compromise erythrocyte functionality.

The findings of the present study agree with previous reports by Bitrus et al. (2024), Ereke et al. (2024), and Osita et al. (2024), who observed that *O. gratissimum* supplementation improved haematological indices and maintained blood parameters within normal physiological ranges in broiler chickens. The results further support the potential of scent leaf as a natural phytochemical additive capable of enhancing blood quality and physiological health in poultry production systems.

Effect of *Ocimumgratissimum* Supplementation on White Blood Cell Differentials of Broiler Chickens

The effects of aqueous *O. gratissimum* extract on white blood cell differentials of broiler chickens are presented in Table 2.

Table 2. Effect of *Ocimumgratissimum* Supplementation on White Blood Cell Differentials of Broiler Chickens

Parameters	T1	T2	T3	T4	SEM
Lymphocytes (%)	55.00 ^b	56.00 ^b	59.67 ^a	62.00 ^a	0.89
Heterophils (%)	37.50 ^a	36.00 ^a	32.67 ^b	31.33 ^b	0.79
Monocytes (%)	5.00	6.00	5.33	5.00	0.19
Eosinophils (%)	2.50	2.00	2.33	1.67	0.19

^{ab} Means within the same row bearing different superscripts differ significantly ($p < 0.05$).

Lymphocyte counts increased significantly ($p < 0.05$) with increasing levels of scent leaf supplementation, ranging from 55.00% in T1 to 62.00% in T4. Birds in T3 and T4 exhibited significantly higher lymphocyte percentages than those in T1 and T2, suggesting enhanced immune responsiveness and improved immunocompetence. Increased lymphocyte production is often associated with stimulation of the immune system and improved disease resistance in poultry birds.

Conversely, heterophil counts decreased significantly ($p < 0.05$) as supplementation levels increased. The reduction in heterophil percentages from 37.50% in T1 to 31.33% in T4 indicates a possible reduction in physiological stress and inflammatory responses in birds receiving higher levels of *O. gratissimum* extract. A lower heterophil-to-lymphocyte ratio is commonly regarded as an indicator of reduced stress and improved welfare in poultry.

Monocyte and eosinophil counts were not significantly ($p > 0.05$) affected by the treatments, indicating that the aqueous extract did not negatively influence phagocytic activity or allergic response mechanisms. The values obtained remained within normal physiological ranges for healthy broiler chickens.

The immunomodulatory effects observed in this study corroborate findings by Bitrus et al. (2024) and Ereke et al. (2024), who reported enhanced lymphocyte counts and reduced stress indicators in broilers supplemented with phytochemical plant extracts. The improvement in immune-related blood parameters may be linked to the antimicrobial and antioxidant activities of the phytochemical constituents of *O. gratissimum*.

Effect of *Ocimumgratissimum* Supplementation on Serum Biochemical Parameters of Broiler Chickens

The effects of aqueous *O. gratissimum* extract on serum biochemical parameters of broiler chickens are presented in Table 3.

Table 3. Effect of *Ocimumgratissimum* Supplementation on Serum Biochemical Parameters of Broiler Chickens

Parameters	T1	T2	T3	T4	SEM
Total Protein (g/dL)	3.15 ^b	2.64 ^b	3.36 ^a	2.60 ^b	0.11
Albumin (g/dL)	1.44	1.13	1.47	1.14	0.06
Globulin (g/dL)	1.71 ^a	1.51 ^b	1.89 ^a	1.45 ^b	0.06
Glucose (mg/dL)	191.00 ^b	240.75 ^{ab}	218.00 ^{ab}	267.67 ^a	9.34
Total Cholesterol (mg/dL)	142.18	105.39	121.40	125.23	5.34



Parameters T1 T2 T3 T4 SEM

Triglycerides (mg/dL) 116.16^a 100.58^{ab} 99.33^b 101.05^{ab} 2.34

^{ab} Means within the same row bearing different superscripts differ significantly ($p < 0.05$).

The serum biochemical profile showed significant ($p < 0.05$) treatment effects on total protein, globulin, glucose, and triglycerides, while albumin and total cholesterol were not significantly affected.

Total protein values ranged from 2.60 to 3.36 g/dL, with birds in T3 recording significantly higher values than other treatment groups. The elevated total protein concentration observed in T3 may indicate improved protein metabolism, enhanced nutrient utilization, and better immune status resulting from moderate supplementation levels of *O. gratissimum* extract.

Albumin values remained statistically similar across treatments, suggesting that liver function and protein synthesis were not adversely affected by the treatments. Similarly, the absence of significant differences in total cholesterol levels indicates that the supplementation did not disrupt lipid metabolism.

Globulin concentration was significantly higher in T1 and T3 than in T2 and T4. Since globulin is associated with immune protein production, the increase observed in T3 may reflect enhanced immune activity stimulated by phytoactive compounds in scent leaf extract.

Serum glucose values increased significantly with increasing supplementation levels, with T4 recording the highest glucose concentration. Although elevated glucose levels may indicate increased metabolic activity and energy mobilization, excessively high concentrations could suggest physiological stress at higher inclusion levels. Therefore, moderate supplementation levels may be more beneficial for maintaining optimal metabolic balance.

Triglyceride values decreased significantly in birds receiving scent leaf supplementation, particularly in T3, indicating improved lipid utilization and possible hypolipidemic effects of the plant extract. These findings are consistent with previous reports by Urom et al. (2018), Eze and Anyanwu (2021), Eze et al. (2022), and Salihu et al. (2023), who reported beneficial effects of *O. gratissimum* on serum protein profile and lipid metabolism in poultry birds.

Effect of *Ocimumgratissimum* Supplementation on Electrolyte and Mineral Balance of Broiler Chickens

The effects of aqueous *O. gratissimum* extract on electrolyte and mineral balance are presented in Table 4.

Table 4. Effect of *Ocimumgratissimum* Supplementation on Electrolyte and Mineral Balance of Broiler Chickens

Parameters (mmol/L)	T1	T2	T3	T4	SEM
Chloride (Cl ⁻)	128.97 ^c	129.52 ^c	131.16 ^b	135.16 ^a	0.76
Sodium (Na ⁺)	138.50	139.26	140.58	140.69	0.36
Calcium (Ca ²⁺)	6.74	6.86	6.82	6.87	0.02
Phosphorus (P)	3.36 ^b	3.52 ^{ab}	3.60 ^a	3.50 ^{ab}	0.03
Potassium (K ⁺)	1.16 ^b	1.12 ^b	1.26 ^a	1.15 ^b	0.02

^{abc} Means within the same row bearing different superscripts differ significantly ($p < 0.05$).

The electrolyte and mineral balance results indicated that chloride, phosphorus, and potassium concentrations were significantly ($p < 0.05$) influenced by treatment, whereas sodium and calcium levels were not significantly affected.

Chloride concentration increased progressively with increasing supplementation levels, with T4 recording the highest value. Despite these differences, the values remained within physiological ranges reported for healthy broiler chickens, indicating that the treatments did not compromise acid-base balance or osmotic regulation.

Sodium and calcium concentrations were statistically similar across treatments, suggesting maintenance of electrolyte stability and mineral homeostasis. Phosphorus and potassium concentrations were significantly higher in T3, which may indicate improved mineral utilization and absorption at moderate supplementation levels.

The stability of electrolyte and mineral profiles observed in this study suggests that aqueous *O. gratissimum* supplementation did not exert adverse effects on renal function or mineral metabolism. Similar observations were reported by Urom et al. (2020), Eze and Anyanwu (2021), and Daramola (2024), who noted that phytoactive supplementation with scent leaf maintained normal electrolyte balance and physiological stability in poultry birds.

Overall, the findings of the present study demonstrate that aqueous extract of *O. gratissimum* positively influenced haematological indices, enhanced immune response, improved selected serum biochemical parameters, and maintained electrolyte balance in broiler chickens without detrimental physiological effects.

Recommendations

1. Long-term feeding trials should be conducted to evaluate the cumulative effects of aqueous *O. gratissimum* supplementation on organ integrity,

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growth performance, and carcass quality in broiler chickens.

- Moderate supplementation levels, particularly the T3 dosage, are recommended due to their beneficial effects on serum protein profile, immune response, and mineral balance without excessive elevation of serum glucose concentration.

APPENDIX A

Image of scent leaf



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