

Anticoccidial effects of methanol extract of *Ocimum gratissimum* (scent leaf) on broilers infected with *Eimeria* oocysts

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ABSTRACT

This study evaluated the anticoccidial property of methanol extract of *Ocimum gratissimum* (OGE) in broiler chicks experimentally infected with sporulated *Eimeria* oocyst. The *O. gratissimum* leaves were collected, authenticated, dried, pulverized and extracted using hydromethanol. Fifty (50) broilers were randomly assigned to 6 groups (1-6) (n = 8). Group 1 was the normal control (uninfected-untreated), while groups 2-6 were infected with *Eimeria* oocyst. Group 2 was untreated, group 3 received amprolium (125 mg/L) and groups 4, 5 and 6 received OGE at 250, 500 and 1000 mg/L of water, respectively, for 7 consecutive days. Thereafter, Faecal oocyst counts (FOC), haematological, biochemical profiles and carcass quality were evaluated. The FOC of OGE (250, 500 and 1000 mg/L) and amprolium (125 mg/L) treated groups were significantly ($p < 0.05$) lower when compared with the FOC of the infected-untreated group on days 13, 15 and 18 post infection. The OGE (250, 500 and 1000 mg/L) and amprolium (125 mg/L) caused a significant ($p < 0.05$) increase in the HB, PCV, and RBC levels of the treated broiler groups when compared with the infected-untreated broiler group. There was no significant difference ($p > 0.05$) in the live weight, dressed weight/carcass weight and the relative weight of thigh and wing weight to live weight of the OGE (250, 500 and 1000 mg/L) and amprolium (125 mg/L) treated broiler groups when compared with the infected-untreated broiler group. This study validates the folkloric use of *Ocimum gratissimum* leaf in treatment of *Eimeria* infections in broilers.

Keywords: Broiler; Coccidiosis; *Eimeria* infection; Herbal medicine; *Ocimum gratissimum*

INTRODUCTION

Coccidiosis is a common parasitic disease affecting poultry and other domestic animals. It is caused by the protozoan parasites belonging to the genus *Eimeria* in most animals, especially poultry or *Isospora* in humans and some animals (Tewari & Maharana, 2011). These parasites invade the intestinal epithelial cells of their hosts, causing cell destruction and leading to clinical signs like diarrhea, weight loss and dehydration. The disease is of significant economic importance in the poultry industry due to its impact on

health, growth and productivity of broilers. Effective management of coccidiosis is essential to ensure economic viability of poultry operations (Ahmad *et al.*, 2023). Traditionally, synthetic anticoccidial drugs have been used to control the disease. However, the overuse of these drugs has led to the emergence of drug resistant *Eimeria* strains, environmental concerns and residual drug accumulation in poultry products, which cause health risks to consumers (Peek & Landman, 2011). As a result, there is increasing interest in exploring natural alternatives with anticoccidial

properties. One promising natural alternative is *Ocimum gratissimum*, commonly known as scent leaf.

Ocimum gratissimum is an aromatic, perennial herb belonging to the family Lamiaceae. It is known as Tanmotsungi-wawagi, Ikeru, Dai doya ta gida, Efinrin ajase and Nchanwu by the Nupe, Ebira, Hausa, Yoruba and Igbo respectively (Chinko & Orlu, 2023). It is commonly found in many geographical regions in the South America and Africa (Uritu *et al.*, 2018). It grows upto 1-3 m tall. The stem is erect, round-quadrangular, much branched, glabrous or pubescent, woody at the base, with the epidermis often peeling in strips. The leaf is simple, opposite and ovate-lanceolate in shape, with a pointed tip and slightly serrated margins. The leaves are typically 5-10 cm in length and 2-5cm in width, though variations may occur depending on environmental conditions (Ashokkumar *et al.*, 2021). Its preliminary phytochemical studies have revealed the presence of bioactive constituents such as alkaloids, flavonoids, tannins and phenols (Alexander *et al.*, 2016). The leaves of *O. gratissimum* is extensively used in the folkloric medicine in the management of several disease conditions (Uritu *et al.*, 2018). The antioxidant, anti-inflammatory, anticancer, hepatoprotective, antidiabetic, antihypertensive, antidiarrhoeal, and antimicrobial properties (Uritu *et al.*, 2018, Chinko & Orlu, 2023). The decoction of the leaves of *O. gratissimum* is used in the traditional management of protozoan such as malaria in human being and trypanosomiasis and *Eimeria* infection in farm animals (Adamu *et al.*, 2009; Pandey *et al.*, 2017). Despite the extensive use the leaves decoction of *O. gratissimum* in the traditional management avian coccidiosis in South-East, Nigeria, there is dearth of information on the scientific literature on the anticoccidial property of the plant harvested in the region. This study aims to evaluate the anticoccidial property of methanol extract of *Ocimum gratissimum* (scent leaf) in broiler chicks experimentally infected with sporulated *Eimeria* oocyst.

MATERIALS AND METHOD

PLANT COLLECTION AND EXTRACTION

The plant material (*Ocimum gratissimum*) leaves were collected from a farmland and authenticated by the botanist. The leaves were washed properly and air dried on laboratory bench. The dried leaves were pulverized with grinding machine to obtain a coarse powder. Methanolic extract was prepared using cold maceration method by soaking the powder in hydromethanol for 48 hours and stirred intermittently. After 48 hours the extract was filtered and concentrated in hot air oven at 40°C. The *Ocimum gratissimum* extract (OGE) was stored in refrigerator throughout the duration of the experiment.

IN-VITRO ANTIOXIDANT ACTIVITIES OF OGE

The free radical scavenging activity of the OGE was evaluated using 2-Diphenyl-1-Picrylhydrazyl (DPPH) and ferric reducing antioxidant power (FRAP) photometric assay as described by Ukwueze *et al.* (2024).

EXPERIMENTAL ANIMAL

A total of 50 day-old chicks were procured from Zartech Broiler Hatchery. They weighed average of 71g and they were housed at the poultry house, Department of Veterinary Animal Production and Biochemistry, Michael Okpara University of Agriculture Umudike, Nigeria. The birds were kept in deep litter poultry system. On arrival, the birds were given glucose in their drinkers and were allowed access to feed and water *ad libitum*. Brooding of the birds occurred immediately on arrival and were provided with a warm temperature with electric bulbs and lamps. At two weeks the birds were randomly divided into 6 (1-6) groups. Vaccination was given according to vaccination schedule for broilers. The birds were reared to maturity.

INFECTION OF THE BIRDS

Sporulated oocysts was collected from Department of Parasitology and Entomology, Michael Okpara University of Agriculture Umudike, Nigeria. About 3 ml containing 3000 sporulated oocysts were inoculated into each bird in group 2-6 at two weeks old. The birds were monitored for clinical signs. The faecal samples were collected from the floor to count oocysts using McMaster technique to confirm the establishment of the infection. Treatment of the birds commenced at 3 weeks of age i.e 1 week post inoculation. The pen of the uninfected-untreated group was cleaned and bedding changed every day to guard against natural infection.

EXPERIMENTAL DESIGN

All the treatments were administered in their drinking water for 7 days. Their weights were recorded weekly, using electronic weighing balance (Made-in-China™, China; 0.001g sensitivity). Immediately after treatment the birds were monitored daily for clinical signs and body weight was taken weekly interval.

At 3 days interval faecal samples were collected to count the *Eimeria* oocysts using McMaster counting chamber which was done by the parasitologist in Department of Veterinary Parasitology and Entomology.

At 5 weeks, the blood samples were collected from the broilers via the jugular vein into EDTA anticoagulant and plain bottles to evaluate the hematological indices and biochemical parameters, respectively.

TABLE I: THE GROUPING AND TREATMENT OF THE BROILERS

Groups	Infection	Treatment
1 (Normal control)	Uninfected	Untreated
2(Negative control)	Infected	Untreated
3 (Positive control)	Infected	Amprolium (120 mg/L)
4 (Low dose)	Infected	OGE, 250 mg/L of H ₂ O
5 (Mid dose)	Infected	OGE, 500 mg/L of H ₂ O
6 (High dose)	Infected	OGE, 1000 mg/L of H ₂ O

DETERMINATION OF OOCYST COUNT

The faecal samples were collected from the floor of each pen using spatula into a plastic container and weighed to obtain 1g. The sample was mixed with saturated sugar solution, the suspension was thoroughly homogenized using a spatula and filtered through a sieve. A measured volume (0.3 ml) of the filtrate was loaded into a McMaster counting chamber using a pipette. The chamber was allowed to settle for 5-10 minutes to ensure oocysts floated into the grid. The slide was examined under a light microscope (10x or 40x objective). Oocysts within the grid area were counted. The total oocyst count per gram of faeces was calculated using the formula:

$$\text{Oocysts per gram (opg)} = \frac{\text{Total oocysts counted}}{\text{Volume of count}}$$

EVALUATION OF HAEMATOLOGICAL PARAMETERS

The manual haemocytometer method was employed in red blood cell (RBC) and total white blood cell (WBC) count; haematocrit method was used in packed cell volume (PCV) estimation while haemoglobin (HB) concentration was estimated with cyanomethaemoglobin method using Drabkin's reagent. The mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) was calculated as described by Brar *et al.* (2000). Thin blood smear was made on clean dry and grease free microscope slide from each of the collected blood sample in EDTA. The slides were air dried and stained with Giemsa stain as described by Brar *et al.* (2000). The slides were later examined with light microscope under oil immersion. The relative number of the neutrophil, lymphocyte, eosinophil, basophils and monocytes were estimated using haemocytometer method. The absolute numbers of the above cell types were calculated from the total WBC count.

$$\text{Absolute number} = (\text{relative number} \times \text{total WBC})/100$$

EVALUATION OF BIOCHEMICAL PARAMETERS

A commercially available reagent kit (Randox Diagnostic Laboratories, United Kingdom) was used to evaluate the serum alanine aminotransferase (ALT), aspartate aminotransferase (AST) and alkaline phosphatase (ALP) activities as well as serum total protein, albumin, total

cholesterol, triglyceride, blood urea nitrogen, and creatinine. The assays was carried out as instructed by the manufacturer.

CARCASS CHARACTERISTIC EVALUATION

At the end of the 7th week of the experiment, three birds were randomly selected from each treatment, weighed and slaughtered for carcass evaluation. The live weight, dressed weight and the weights of the breast, shank and drumstick were determined using electronic scale (Made-in-ChinaTM, China; 0.001g sensitivity).

STATISTICAL ANALYSIS

The data obtained from the study, including oocyst count, weight gain, feed conversion ratio and other parameters were analyzed using statistical package for social science (SPSS 2008), version 22. One-way analysis of variance (ANOVA) coupled with appropriate post-hoc statistics was conducted to compare the differences between treatment groups. Values of $p < 0.05$ were considered significant.

RESULTS

FRAP of OGE

The OGE produced concentration dependent increase in the FRAP values which was significantly ($p < 0.05$) lower when compared with ascorbic acid (Figure I).

DPPH RADICAL SCAVENGING EFFECTS OF

The OGE produced concentration dependent increase in the DPPH radical scavenging potential. At 25, 50, 100, and 200 $\mu\text{g/mL}$ concentration, the percentage antioxidant activity of OGE were significantly ($p < 0.05$) lower when compared with ascorbic acid (Figure II). At 400 $\mu\text{g/mL}$ concentration, the percentage antioxidant activity of OGE was significantly ($p < 0.05$) different when compared with ascorbic acid.

EFFECTS OF OGE ON THE FAECAL OOCYST COUNT (FOC) OF BROILER EXPERIMENTALLY INFECTED WITH *EIMERIA* SPP.

The effect of OGE on FOC is presented in Table II. No oocyst was found in the faece of the uninfected-untreated group throughout the duration of the experiment. The FOC of the infected-untreated group was on a progressive increase throughout the duration of the experiment. The FOC of OGE (250, 500 and 1000 mg/L) and amprolium (125 mg/L) treated groups were on persistent decline from day 10 post infection to the end of the experiment.

The FOC of OGE (250, 500 and 1000 mg/L) and amprolium (125 mg/L) treated groups were significantly ($p < 0.05$) lower when compared with the FOC of the infected-untreated group on days 13, 15 and 18 post infection.

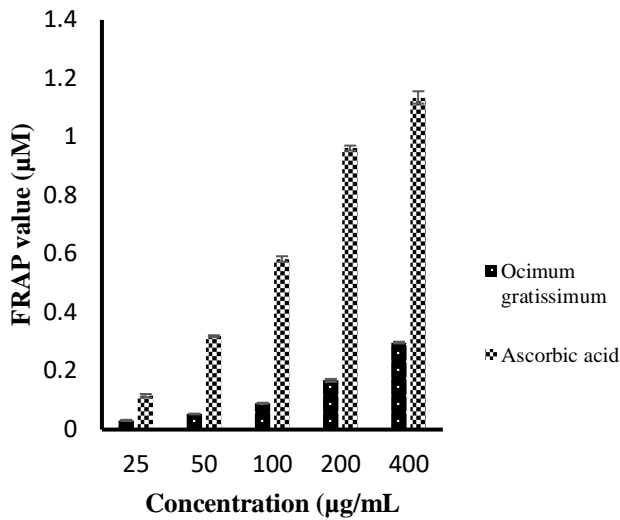


Figure I: The FRAP value of OGE

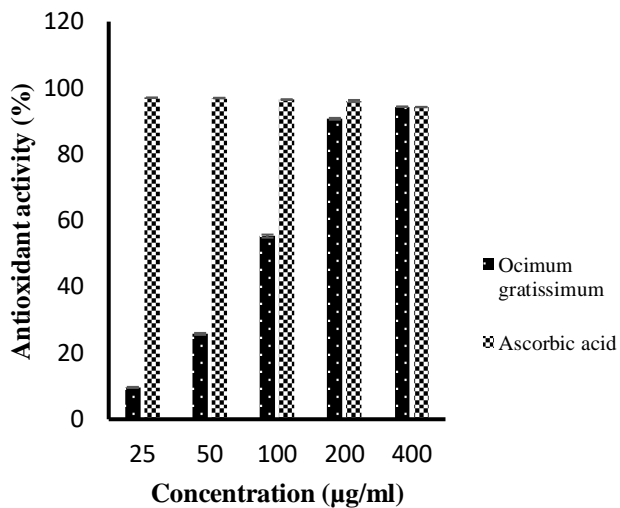


Figure II: DPPH radical scavenging effects of OGE

THE EFFECTS OF OGE ON THE HAEMATOLOGICAL PROFILE OF BROILER EXPERIMENTALLY INFECTED WITH *EIMERIA* SPP

The effects of OGE on the erythrocytic profile of broiler experimentally infected with *Eimeria* spp is presented in Table III. The OGE (250, 500 and 1000 mg/L) and amprolium (125 mg/L) caused a significant ($p < 0.05$) increase in the HB, PCV, and RBC levels of the treated broiler groups when compared with the infected-untreated broiler group.

The OGE (250, 500 and 1000 mg/L) and amprolium (125 mg/L) produced a significant ($p < 0.05$) decrease in the MCH and MCHC levels of the treated broiler groups when compared with the infected-untreated broiler group.

The effects of OGE on the leucocytic profile of broiler experimentally infected with *Eimeria* spp is presented in Table IV.

The OGE (250, 500 and 1000 mg/L) and amprolium (125 mg/L) produced a significant ($p < 0.05$) decrease in the TWBC, relative heterophil, relative eosinophil, absolute heterophil, absolute monocyte and absolute eosinophil levels of the treated broiler groups when compared with the infected-untreated broiler group.

The OGE (250, 500 and 1000 mg/L) and amprolium (125 mg/L) caused a significant ($p < 0.05$) increase in the relative lymphocyte levels of the treated broiler groups when compared with the infected-untreated broiler group.

The OGE (250, 500 and 1000 mg/L) produced a significant ($p < 0.05$) decrease in the absolute lymphocyte levels of the treated broiler groups when compared with the infected-untreated broiler group.

TABLE II: EFFECTS OF OGE ON FAECAL OOCYST COUNT OF BROILER EXPERIMENTALLY INFECTED WITH *EIMERIA* SPP

Treatment	DAY 7 PI	DAY 10 PI	DAY 13 PI	DAY 15 PI	DAY 18 PI
Uninfected-untreated	0.00 ± 0.00	0.00 ± 0.00*	0.00 ± 0.00*	0.00 ± 0.00*	0.00 ± 0.00*
Infected-untreated	7433.33 ± 744.61	8800.00 ± 305.51	9333.33 ± 145.30	9533.33 ± 120.19	10000.00 ± 115.47
Amprolium, 125 mg/L	6200.00 ± 378.60*	4933.33 ± 417.67*	3400.00 ± 550.76*	2266.67 ± 317.98*	1060.00 ± 70.24*
OGE, 250 mg/L	8933.33 ± 202.76	6600.00 ± 346.41*	4766.67 ± 1036.55*	2866.67 ± 669.16*	1433.33 ± 296.27*
OGE, 500 mg/L	10266.67 ± 656.59*	7466.67 ± 611.92	4033.33 ± 497.77*	1800.00 ± 115.47*	980.00 ± 62.45*
OGE, 1000 mg/L	9200.00 ± 624.50*	7100.00 ± 585.95	4066.67 ± 497.77*	1633.33 ± 352.77*	850.00 ± 104.08*

* $p < 0.05$ when compared with the infected-untreated group, OGE = *Ocimum gratissimum* extract

TABLE III: EFFECTS OF OGE ON GENERAL BLOOD PARAMETERS OF BROILER EXPERIMENTALLY INFECTED WITH *EIMERIA* SPP

Treatment	HB (g/dL)	PCV (%)	RBC($\times 10^6/\mu\text{L}$)	MCV (fL)	MCH (pg)	MCHC (g/dL)
Uninfected untreated	13.27 \pm 0.37*	31.33 \pm 2.33*	3.59 \pm 0.25*	87.30 \pm 0.50	37.19 \pm 1.46*	42.62 \pm 1.88*
Infected untreated	11.13 \pm 0.29	22.00 \pm 1.15	2.49 \pm 0.12	88.33 \pm 0.28	44.82 \pm 1.08	50.75 \pm 1.35
Amprolium, 125 mg/L	13.13 \pm 0.29*	31.67 \pm 1.45*	3.59 \pm 0.16*	88.20 \pm 0.05	36.66 \pm 0.87*	41.57 \pm 1.01*
OGE, 250 mg/L	12.87 \pm 0.37	29.33 \pm 2.33*	3.29 \pm 0.26*	89.16 \pm 0.51	39.40 \pm 1.85*	44.20 \pm 2.11*
OGE, 500 mg/L	12.60 \pm 0.31*	28.67 \pm 1.20*	3.29 \pm 0.14*	87.04 \pm 0.04	38.31 \pm 0.66*	44.02 \pm 0.76*
OGE, 1000 mg/L	13.07 \pm 0.07*	30.67 \pm 0.33*	3.46 \pm 0.04*	88.63 \pm 0.16	37.77 \pm 0.36*	42.62 \pm 0.40*

* $p < 0.05$ when compared with infected untreated group, OGE = *Ocimum gratissimum* extract, HB = haemoglobin, PCV = packed cell volume, RBC = red blood cell, MCV = mean corpuscular volume, MCH = mean corpuscular haemoglobin, MCHC = mean corpuscular haemoglobin concentration

THE EFFECTS OF OGE ON THE BIOCHEMICAL PARAMETER OF BROILER EXPERIMENTALLY INFECTED WITH *EIMERIA* SPP

The effects of OGE on the biochemical parameter of broiler experimentally infected with *Eimeria* spp is presented in Table V.

The total protein, albumin, globulin, cholesterol, HDL-C and urea levels as well as ALP, AST and ALT activities of OGE (250, 500 and 1000 mg/L) treated broiler groups were significantly ($p < 0.05$) increased when compared with the infected-untreated broiler group.

The triglyceride and VLDL-C levels of OGE (250, 500 and 1000 mg/L) treated broiler groups were significant ($p < 0.05$) decreased when compared with the infected-untreated broiler group.

EFFECTS OF OGE ON CARCASS QUALITIES OF BROILER EXPERIMENTALLY INFECTED WITH *EIMERIA* SPP

The effects of OGE on live weight and carcass characteristics of broiler experimentally infected with *Eimeria* spp is presented in Table VI.

There was no significant difference ($p > 0.05$) in the live weight, dressed weight/carcass weight and the relative weight of thigh and wing weight to live weight of the OGE (250, 500 and 1000 mg/L) and amprolium (125 mg/L) treated broiler groups when compared with the infected-untreated broiler group.

The dressing percentages and the relative weights of drum stick and thigh to live weight of the OGE (250, 500 and 1000 mg/L) treated broiler groups were significantly ($p < 0.05$) reduced when compared with the infected-untreated broiler group.

DISCUSSION

The study evaluated the anticoccidial property of methanol extract of *Ocimum gratissimum* (scent leaf) in broiler chicks experimentally infected with sporulated *Eimeria* oocyst.

The OGE significantly ($p < 0.05$) reduced the FOC in the treated broilers when compared to the infected untreated group. The OGE treatment ameliorated anaemia and did not manifest hepatotoxic effect, but produced slightly elevated urea levels in the treated broiler. The progressive increase in the FOC of the infected-untreated group reflects the natural progression of the parasitic infection. The amprolium and OGE (1000 mg/L) significantly reduced FOC in the treated groups when compared with the infected-untreated group.

The OGE demonstrated dose-dependent anticoccidial activity. The anticoccidial activity of OGE could be attributed to the presence of some bioactive compounds such as eugenol and thymol. The antiparasitic property of *O. gratissimum* has been linked to eugenol and thymol which disrupt parasite metabolism and replication (Njoku & Asuzu, 1998). The finding of this study corroborated the report of Ogbu & Onuh (2015) on the anticoccidial activity of *O. gratissimum* leaf where the plant product significantly ($p < 0.05$) reduced the FOC.

The high antioxidant content of OGE is of importance in the scavenging of free radicals in the body. Antioxidants can preserve gut integrity and protect gut tissue by lowering oxidative degeneration. Antioxidants also, have anti-inflammatory property which helps in reducing inflammation. Reduced inflammation, enhance gut integrity, gut healing, improve nutrient absorption, and support gut histology (Okusanya *et al.*, 2023).

TABLE V: EFFECTS OF OGE ON THE BIOCHEMICAL PARAMETER OF BROILER EXPERIMENTALLY INFECTED WITH *EIMERIA* SPP

Treatment	Uninfected untreated	Infected untreated	Amprolium, 125 mg/L	OGE, 250 mg/L	OGE, 500 mg/L	OGE, 1000 mg/L
Total protein (g/dl)	2.90± 0.10*	2.74 ± 0.01	2.73 ± 0.08	3.36± 0.02*	3.13 ± 0.02*	3.39 ± 0.04*
Albumin (g/dl)	1.58± 0.02*	1.39 ± 0.04	1.33± 0.03	1.58 ± 0.02*	1.57± 0.03*	1.59± 0.05*
Globulin (g/dl)	1.32 ± 0.10	1.35± 0.04	1.40 ± 0.10	1.78± 0.03*	1.55± 0.02*	1.80 ± 0.03*
ALP (U/L)	113.88± 0.08	113.34 ± 0.23	113.43± 0.25	114.91± 0.25*	114.38 ± 0.25*	114.82± 0.21*
ALT (U/L)	7.20 ± 0.09*	4.88± 0.23	5.52± 0.32	5.66± 0.08	5.92± 0.18*	6.88± 0.83*
AST (U/L)	87.70 ± 0.75	87.05± 2.63	94.85± 6.38	141.00 ± 1.50*	108.50 ± 3.00*	133.20 ± 0.75*
Cholesterol(mg/dl)	112.64 ± 3.31*	97.23± 3.55	89.05 ± 1.20*	116.73 ± 2.96*	102.53 ± 3.01	109.99 ± 3.34*
Triglyceride(mg/dl)	127.30± 3.74*	143.49± 1.27	114.92± 4.48*	123.49± 5.11*	108.25± 1.14*	136.51 ± 3.66
HDL-C(mg/dl)	51.71 ± 1.49*	44.55± 0.65	48.46 ± 0.86	69.92 ± 0.33*	53.98± 0.86*	56.59 ± 0.56*
VLDL-C(mg/dl)	25.46 ± 0.75*	28.70 ± 0.25	22.98 ± 0.90*	24.70 ± 1.02*	21.65 ± 0.23*	27.30± 0.73
LDL-C(mg/dl)	35.47 ± 3.40*	23.98± 3.30	17.61 ± 0.46	22.11 ± 2.28	26.89± 2.71	26.10± 3.80
Urea (mg/dl)	5.63 ± 0.20	4.22± 0.35	5.43 ± 0.60	12.46± 0.53*	13.47 ± 0.88*	13.26 ± 0.35*
Creatinine (mg/dl)	0.44± 0.08	0.48± 0.02	0.97 ± 0.06*	0.54 ± 0.06	0.32± 0.06	0.65 ± 0.03

*p<0.05 when compared with infected untreated group; WBC = white blood cell, OGE = *Ocimum gratissimum* extract

TABLE IV: EFFECTS OF OGE ON THE LEUCOCYTE PROFILE OF BROILER EXPERIMENTALLY INFECTED WITH *EIMERIA* SPP

Treatment	Uninfected-untreated	Infected-untreated	Amprolium, 125 mg/L	OGE, 250 mg/L	OGE, 500 mg/L	OGE, 1000 mg/L
Total WBC (×10 ³ /μL)	20.40 ± 0.53*	32.98 ± 2.04	24.65 ± 1.09*	20.97 ± 1.07*	21.57 ± 0.58*	20.05 ± 0.89*
Relative lymphocyte (%)	61.33 ± 1.45*	40.67 ± 0.88	55.33 ± 1.45*	54.67 ± 0.67*	54.67 ± 0.88*	54.33 ± 0.88*
Relative heterophil (%)	31.00 ± 0.58*	41.00 ± 1.53	34.33 ± 2.33*	36.33 ± 0.33*	37.33 ± 1.33*	37.00 ± 0.58*
Relative monocyte (%)	5.33 ± 0.67	6.00 ± 0.58	4.67 ± 0.33	6.33 ± 0.67	5.33 ± 0.33	6.67 ± 0.33
Relative eosinophil (%)	2.33 ± 0.33*	11.67 ± 0.88	5.67 ± 0.67*	2.67 ± 0.33*	2.67 ± 0.67*	1.67 ± 0.67*
Absolute lymphocyte (×10 ³ /μL)	12.52 ± 0.59	13.40 ± 0.80	13.67 ± 0.96	11.46 ± 0.53*	11.78 ± 0.23*	10.89 ± 0.42*
Absolute heterophil (×10 ³ /μL)	6.32 ± 0.12*	13.51 ± 0.87	8.41 ± 0.22*	7.63 ± 0.46*	8.06 ± 0.43*	7.43 ± 0.45*
Absolute monocyte (×10 ³ /μL)	1.08 ± 0.11*	1.99 ± 0.26	1.16 ± 0.12*	1.33 ± 0.18*	1.15 ± 0.04*	1.34 ± 0.10*
Absolute eosinophil (×10 ³ /μL)	0.48 ± 0.07*	3.86 ± 0.45	1.41 ± 0.23*	0.55 ± 0.05*	0.58 ± 0.15*	0.32 ± 0.12*
Absolute basophil (×10 ³ /μL)	0.00*	0.22 ± 0.11	0.00± 0.00*	0.00 ± 0.00*	0.00 ± 0.00*	0.07 ± 0.07*

*p<0.05 when compared with infected untreated group; WBC = white blood cell, OGE = *Ocimum gratissimum* extract

TABLE VI: EFFECTS OF OGE ON LIVE WEIGHT AND CARCASS CHARACTERISTICS OF BROILER EXPERIMENTALLY INFECTED WITH *EIMERIA* SPP.

TREATMENT	uninfected untreated	Infected untreated	Amprolium, 125 mg/L	OGE, 250 mg/L	OGE, 500 mg/L	OGE, 1000 mg/L
Live weight (g)	1942.50 ± 139.43	2141.50 ± 38.39	2088.50 ± 28.00	2220.50 ± 127.88	1904.50 ± 308.59	2129.50 ± 136.54
Dressed weight (g)	1399.50 ± 62.64	1587.50 ± 41.28	1528.50 ± 63.80	1575.50 ± 110.56	1273.50 ± 230.36	1402.00 ± 53.11
Dressing percentage	72.33 ± 1.98	74.11 ± 0.60	73.13 ± 2.07	70.85 ± 0.90*	66.40 ± 1.40 *	66.06 ± 1.75*
Relative organ weight:						
Wing (%)	6.25 ± 0.09	7.78 ± 0.118*	7.73 ± 0.37*	5.99 ± 0.05	7.29 ± 0.24*	6.28 ± 0.60
Back cut (%)	17.08 ± 0.53	11.78 ± 0.05*	11.24 ± 0.20*	18.09 ± 1.16	9.95 ± 0.31*	18.38 ± 1.76
Breast (%)	28.57 ± 0.89	26.08 ± 0.74*	27.00 ± 2.13	27.16 ± 1.26	24.16 ± 0.68 *	25.58 ± 0.08*
Drum stick (%)	11.97 ± 0.37	11.29 ± 0.27	10.57 ± 0.11*	9.76 ± 0.51*	10.10 ± 0.09*	9.97 ± 0.20*
Thigh (%)	9.93 ± 0.38	12.34 ± 0.68*	13.00 ± 0.21*	8.26 ± 0.15*	12.35 ± 0.46*	8.02 ± 0.25*

*p<0.05 when compared with the infected-untreated group, OGE = *Ocimum gratissimum* extract

Parasitic infections often induce anemia and leukocytosis due to blood loss and immune system activation (Djokic *et al.*, 2021). The infected untreated group had reduced HB, PCV, and RBC levels, indicative of anemia while TWBC levels reflected the body's immune response. The OGE treatment restored these indices (erythropoiesis and immune function) possibly through its antioxidative and anti-inflammatory actions (Abd El-Rahman *et al.*, 2020).

The infection significantly reduced total protein, albumin, and globulin levels, likely due to liver impairment. Treatment with OGE improved these parameters in a dose-dependent manner. The effect may be due to the ability of OGE to mitigate oxidative stress and support liver protein synthesis, as reported by Ojo *et al.* (2013).

Parasitic infections have been associated with dyslipidemia due to altered liver metabolism and function. The infected untreated group exhibited elevated cholesterol, triglycerides, LDL-C, and VLDL-C, along with reduced HDL-C levels (Table IV). The OGE treatment ameliorated the dyslipidemia in a dose-dependent manner, which can be linked to its hepatoprotective and lipid-modulating properties (Yu *et al.*, 2023). The OGE treatment has no hepatotoxic effect because ALT level is within the normal range for the specie. This effect can be attributed to the antioxidant properties of its bioactive compounds which protect hepatocytes from damage caused by oxidative stress and inflammation (Iweala, 2012).

The OGE at 250 mg/L had a positive effect on live and dressed/carcass weights of the treated broiler. The dressing percentage was comparable to the infected untreated group, indicating its potential as an alternative additive for improving growth and performance. At higher concentrations (500 and 1000 mg/L) resulted in reduced dressed/carcass weight and percentages due to toxicity or metabolic stress at higher doses, which negatively impacted meat yield (Nweze & Ekwe, 2012).

In conclusion, this study validates the folkloric use of *Ocimum gratissimum* leaf in treatment of *Eimeria* infections in broilers. The extract produced anticoccidial effect in a dose-dependent manner. *Ocimum gratissimum* leaf extract is a viable alternative to orthodox drugs in the treatments of coccidiosis, enhancing growth and meat yield in poultry. Further study to isolate and characterize the bioactive compound and its mechanism of action should be conducted.

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REFERENCES

- Abd El-Rahman, G. I., Behairy, A., Elseddawy, N. M., Batiha, G. E. S., Hozzein, W. N., Khodeer, D. M. & M. Abd-Elhakim, Y. (2020). *Saussurea lappa* ethanolic extract attenuates triamcinolone acetone-induced pulmonary and splenic tissue damage in rats via modulation of oxidative stress, inflammation, and apoptosis. *Antioxidants*, 9(5), 396.
- Adamu, M., Nwosu, C. O. & Agbede, R. (2009). Anti-trypanosomal effects of aqueous extract of *Ocimum gratissimum* (Lamiaceae) leaf in rats infected with *Trypanosoma brucei brucei*. *African Journal of Traditional, Complementary and Alternative Medicines*, 6(3), 262-267.
- Ahmad, R., Yu, Y. H., Hua, K. F., Chen, W. J., Zaborski, D., Dybus, A. & Cheng, Y. H. (2023). Management and control of coccidiosis in poultry—A review. *Animal Bioscience*, 37(1), 1.
- Alexander, P. (2016). Phytochemical screening and mineral composition of the leaves of *Ocimum gratissimum* (scent leaf). *International Journal of Applied Sciences and Biotechnology*, 4(2), 161-165.
- Ashokkumar, K., Pandian, A., Murugan, M., Dhanya, M. K. & Vellaikumar, S. (2021). Phytochemistry and pharmacological properties of *Ocimum gratissimum* (L.) extracts and essential oil—A critical review. *Journal of Current Opinion in Crop Science*, 2(1), 138-148.
- Brar, R. S., Sandhu, H. S. & Singh, A. (2000). *Veterinary clinical diagnosis by laboratory methods*. (Kalyani Publishers, New Delhi 2000).
- Chinko, B. C., & Orlu, C. N. (2023). Antipyretic and Anti-Inflammatory Effects of *Ocimum gratissimum* in Male Wistar Rats. *Inflammation*, 3, 8.
- Djokic, V., Rocha, S. C. & Parveen, N. (2021). Lessons learned for pathogenesis, immunology, and disease of erythrocytic parasites: Plasmodium and Babesia. *Frontiers in Cellular and Infection Microbiology*, 11, 685239.
- Iweala, E. E. (2012). Analgesic and hepatoprotective activity of methanolic leaf extract of *Ocimum gratissimum* (L.). *Research Journal of Medicinal Plant*, 6(1), 108-115.
- Njoku, C. J. & Asuzu, I. U. (1998). The anthelmintic effects of the leaf extract of *Ocimum gratissimum* (L.). *Phytomedicine*, 5(6), 485-488.
- Nweze, B. O. & Ekwe, O. O. (2012). Growth performance, gut and haemo-microbial study of finishing broilers fed African sweet basil (*Ocimum gratissimum*) leaf extract. *Ozean Journal of Applied Sciences*, 5(2), 185-191.

- Ogbu, C. C. & Onuh, S. S. (2015). Oocyst output, performance and haematological indices of broiler chickens infected with coccidian oocysts and fed *Ocimum gratissimum* leaf extract. *Global Journal Poultry Farming and Vaccination*, 3, 146-153.
- Ojo, A. O., Oloyede, O. I., Olarewaju, O. I. & Ajiboye, B. O. (2013). Evaluation of transaminases activity of aqueous extract of *Ocimum gratissimum* in the liver and kidney of Albino Rats. *International Journal of Biological & Medical Research*, 4(4), 3650-3653.
- Okusanya, P. O., Akinlad, O. O., Jarikre, T. A. & Ockiya, M. A. (2023). Growth Performance, Gut Histology and Anti-Coccidial Effect of Aqueous Blends of Scent Leaf, Ginger and Garlic in Broiler Chicken. *Nigerian Veterinary Journal*, 44(4), 12-22.
- Pandey, A. K., Kumar, P., Singh, P., Tripathi, N. N. & Bajpai, V. K. (2017). Essential oils: Sources of antimicrobials and food preservatives. *Frontiers in Microbiology*, 7, 2161.
- Peek, H. W. & Landman, W. J. M. (2011). Coccidiosis in poultry: anticoccidial products, vaccines and other prevention strategies. *Veterinary Quarterly*, 31(3), 143-161.
- Tewari, A. K. & Maharana, B. (2011). Control of poultry coccidiosis: changing trends. *Journal of Parasitic Diseases*, 35, 10-17.
- Uritu, C. M., Mihai, C. T., Stanciu, G. D., Dodi, G., Alexa-Stratulat, T., Luca, A. & Tamba, B. I. (2018). Medicinal plants of the family Lamiaceae in pain therapy: A review. *Pain Research and Management*, 2018(1), 7801543.
- Yu, W. Q., Wang, X. L., Ji, H. H., Miao, M., Zhang, B. H., Li, H. & Guo, S. D. (2023). CM3-SII polysaccharide obtained from *Cordyceps militaris* ameliorates hyperlipidemia in heterozygous LDLR-deficient hamsters by modulating gut microbiota and NPC1L1 and PPAR α levels. *International Journal of Biological Macromolecules*, 239, 124293.