Performance, haemato-biochemical parameters of broiler chicks administered Rolfe (*Daniellia oliveri*) leaf extract as an antibiotic alternative

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ABSTRACT

The objective of the present study was to investigate the performance, haemato-biochemical parameters of broiler chicks administered *Daniellia oliveri* leaf extract (DOE) as an antibiotic alternative. A total of 250 (Ross 308) one-day old broiler chicks were allotted to five treatments in a completely randomized design and each treatment group was further sub-divided into five replicates containing ten (10) birds each. Treatment 1 (Control) was given 1.20 g/ liter of Neomycin in water, while treatments 2, 3, 4 and 5 were given DOE at 10 ml, 20 ml, 30 ml and 40 ml/liter of water respectively. The experiment lasted for 8 weeks during which performance, hematology and serum parameters were recorded. Average daily gain (DWG) and feed conversion efficiency (FCE) were significantly influenced by the dietary treatment (P<0.05). Average daily feed intake (AFI) and daily water consumption were not significantly different among the treatment (P>0.05). However, increasing the level of DOE from 10 ml to 40 ml/liter tend to decrease the AFI. Highest mortality was recorded in birds fed T1 (10%) followed by T2 (3%), T3 (1%) respectively, none was recorded in T4 and T5. Results on hematology revealed that there were significant differences (P<0.05) in the values of PCV, RBC, Hb, MCV, MCH, MCHC, WBC and its differentials. All results on the serum biochemical parameters were significantly different among the treatment (P<0.05) except for creatinine, total bilirubin, calcium, phosphorus, sodium and chloride ions which were not influenced (P>0.05) by dietary feeding of DOE. It was concluded that feeding birds DOE up to 40 ml/litre enhanced the overall performance and does not have any deleterious effect on the animal.

Keywords: *Daniellia oliveri*; Performance; haematology; serum; broiler chicks

1. INTRODUCTION

Concern about the extensive and indiscriminate use of antibiotics in Nigeria poultry production has led to the launch of a new research project designed to improve the health of the nation's framed chickens using alternative feed additives to antibiotics. One of such alternatives is the use of medicinal plants which are natural, less toxic, residue free and have been found to contain several bioactive chemicals which confers them the ability to perform multiple biological activities (Wang et al., 2008). Among the potential plants is *Daniellia oliveri* which is found to be loaded with minerals, vitamins, amino acid and other secondary metabolites. *Daniellia oliveri* (Rolfe) belongs to the family Fabaceae, commonly known as West Africa copal tree is an evergreen uncultivated copiously available tree, particularly in the savannah zone of Nigeria (Olafadehan and Okunade, 2018). The plant is also found in temperate and tropical regions of the world, Amazon region, South America and Africa (Gentry, 1n evergreen uncultivated copiously available tree, particularly in the savannah zone of Nigeria (Olafadehan and Okunade, 2018). The plant is also found in temperate and tropical regions of the world, Amazon region, South America and Africa (Adubiaro et al., 2016).

According to Osuntokun *et al*. (2016), the leaves of *Daniellia oliveri* produce a liquid oleoresin which consists of large but varying amounts of phytochemicals most especially Oleresins. The Oleresins is traditionally used as an as an anti-inflammatory agent and in the treatment of skin and genito-urinary tract disease (Adubiaro *et al*., 2000). The leaves are also rich in bioactive chemicals such as alkaloids, flavonoids, tannins and saponins which confers it the ability to act as anti-inflammatory, antibacterial, anti-fungal and antioxidant agent (Daniels et al., 2013; Edeoga, 2006).

Although extensive studies have been done on the beneficial effect of some plant extracts in poultry nutrition (Hernandez et al., 2004; Cross et al., 2007). Mohamed et al. (2016) reported that supplementation of garlic extract at 75 mg allicin/kg in the diet of broiler chicken improved feed conversion ratio, protein efficiency ratio, performance index and increased PCV %, Hb %, RBCs and blood serum units. Similar observation was made by Al-Kassie (2009) who showed that the inclusion of 0.5 % dry thyme and cinnamon leaves in broiler diets had significant effects on feed efficiency, growth performance and haematological parameters. Yet there is a dearth of information on feeding *Daniellia oliveri* to broilers, feeding birds with these plants will also help to bridge the gap between production and food safety.

The objective of this study is to evaluate the performance, haemato-biochemical parameters of broiler chicks administered *Daniellia oliveri* leaf extract as an antibiotic alternative.

2. MATERIALS AND METHODS

Area of the study

The experiment was carried out at the University of Abuja Teaching and Research Farm, Animal Science Section, Main Campus, along Airport Road, Gwagwalada, Abuja, Nigeria. Gwagwalada is the headquarters of the Gwagwalada Area Council and is located between latitude $8^0 57'$ and $8^0 5''$N and longitude $7^0 05'$ and $7^0 06'$E. The temperature of Gwagwalada ranges from 28-33 ℃ in the day time and 22-25℃ in the night.

Collection of test material (*Daniellia oliveri* leaf) and preparation of the extract

Fresh and healthy *Daniellia oliveri* leaves were obtained from several strands of the trees at the premises of the University of Abuja, Gwagwalada, Nigeria. The mature leaves were dark green and slightly glossy with a lighter mid veins and undersides. The plant was authenticated at the Herbarium Unit, Department of Biological Sciences, University of Abuja with a voucher specimen number DOS 121 – 2019.

Daniellia oliveri leaf was rinsed thoroughly with running tap water followed by distilled water to remove soil and other bound particles, air dried until a constant weight was obtained and made into meal using a blender. The leaf extract was prepared by putting 250 grams of *Daniellia oliveri* meal into 1000 ml of ethanol (80% BDH) and soaked for 48hrs in an air tight container. The obtained extract was filtered through a normal filter paper using Whatman No.1 filter paper and kept in refrigerator at 4 ℃ for further analysis.

Chemical analysis

The extract was subjected to standard chemical tests for the detection of saponins, flavonoids, phenolics, alkaloids, steroids and glycosides using standard methods described by Harbone (1973); Odebiyi and Sofowora (1978). Tannins (Van-Burden and Robinson, 1981) and flavonoids (Boham and Kocipai-Abyazan, 1974) contents were also determined in the extracts.

Proximate analysis of the leaf was carried out using the method of AOAC (1990). The mineral content of the leaf was determined using Atomic Absorption Spectrophotometer (AAS) model 392 A.

Management of experimental birds

Two hundred and fifty-one-day-old broilers (Ross 308) of mixed sex were obtained from (a commercial hatchery in Ibadan) and transported to University of Abuja Teaching and Research Farm, Abuja. Prior to the commencement of the experiment, the pens were properly disinfected, and the drinkers and feeders were thoroughly washed while wood shavings were spread on the floor as litter material. The chicks were weighed individually at the beginning of the experiment; wing–banded, distributed randomly into 5 treatments of 250 chicks of five replicates each consisting of 10 birds. Electric brooders were used as source of heat. Light was also provided approximately 24 hours in a form of natural light during the day and artificial light during the night, ten bulbs (100 watts) were used for this purpose. The initial brooding temperature was 34°C in the first week of age which was gradually reduced by 2°C per week to 22°C. Birds were kept under similar conditions of management throughout the experimental period. Vaccination was done according to the prevailing disease condition in the environment. Table 1 shows the vaccination schedule of the experimental birds. Water soluble multi-vitamins (Biovite super® at 1 ml to 5 liters of water) was also administered to reduce the vaccination stress on the animal.

Table 1 Vaccination programme for birds

Age / day	Vaccines	Route of administration
7	Lasota (1st dose)	Drinking water
11	Gumboro (1st dose)	Drinking water
15	Lasota (2nd dose)	Drinking water
21	Gumboro (2nd dose)	Drinking water
28	Gumboro (3rd dose)	Drinking water

Table 2 Ingredient composition of the experimental diets

Ingredients	Starters mash (0-4 weeks)	Finishers mash (5-8 weeks)
Maize	52.00	60.00
Wheat offal	2.50	5.00
Soya bean meal	30.00	25.00
Groundnut cake	8.00	4.00
Fish meal (72%)	2.00	2.00
Limestone	1.50	1.50
Bone meal	3.00	3.00
Lysine	0.20	0.20
Methionine	0.20	0.20
*Premix	0.25	0.25
Salt	0.30	0.30
Toxin binder	0.10	0.10
Calculated analysis (% DM)		
Crude protein	23.10	21.40
Crude fibre	4.18	5.01
Ether extract	4.03	4.47
Calcium	1.50	1.60
Phosphorus	0.58	0.66
Energy (Kcal/kg)	2910.3	3200.8

*Premix supplied per kg diet: - Vit A, 13,000 I.U; Vit E, 5mg; Vit D3, 3000I.U, Vit K, 3mg; Vit B2, 5.5mg; Niacin, 25mg; Vit B12, 16mg; Choline chloride, 120mg; Mn, 5.2mg; Zn, 25mg; Cu, 2.6g; Folic acid, 2mg; Fe, 5g; Pantothenic acid, 10mg; Biotin, 30.5g; Antioxidant, 56mg

Experimental diet and design

Birds were fed a basal diet formulated according to NRC (1994). Starter diet (0-4 weeks) containing a crude protein (CP) of 23.23% and metabolizable energy (ME) of 2950.3 kcal/kg and finisher diet (5-8 weeks) containing 21.40 % CP and 3200.8 kcal/kg ME were given. Table 2 shows both the ingredient and calculated composition of the experimental basal diet. There were five treatments in all. Treatment 1 (Control) was given 1.20 g/ liter of Neomycin in water, while treatments 2, 3, 4 and 5 were given *Daniellia oliveri* extracts at 10 ml, 20 ml, 30 ml and 40 ml/liter of water respectively. There were 50 chicks/treatment, and the treatments were randomly assigned to the birds in each experimental group using a completely randomized design (CRD).

Measurements

Feed intake was determined as the difference between feed offered and feed left over. Birds were weighed to determine the weekly weight gain. Feed intake and weight recorded were used to determine feed conversion ratio (FCR).

Blood collection and analysis

At day 56, five birds were randomly from each treatment for hematological and serum biochemical analysis. Selected animals were kept in a stress free environment to prevent oxygenated blood becoming deoxygenated during blood collection. The sampled birds were bled from punctured wing vein to aspire 3mls of blood from each birdand collected into bijou bottle treated with ethylene diamine tetra acetate for hematological assay. Hematological parameters such as Red Blood Cells (RBC), White Blood Cell (WBC), Packed Cell Volume (PCV), Hemoglobin (Hb) and absolute counts of neutrophils and lymphocytes were recorded according to Jain (1986). Values of MCV, MCH and MCHC were calculated using formula described by Jain (1993).

MCV (fl) = PCV/RBC × 100
MCH (pg) = Hb/RBC × 10
MCHC (%) = Hb (100mg blood)/PCV × 100

The second set of bottles was used for serum analysis. Bottles used were free from anticoagulant. The Serum total protein, Blood glucose, Creatinine, Uric acid, Total bilirubin, Albumin, Globulin, Alkaline phosphate,Alanine transaminase, Glutamic Oxaloacetate Transaminase (GOT), Glutamic Phosphate Transaminase (GPT) level were computed according to Scott (1965). Serum cholesterol, Triglycerides, High density lipoprotein, Low density lipoprotein were determined by method outlined by Guache et al. (1991).

3. EXPERMENTAL OUTCOME

Laboratory composition of experimental diet

Table 2 reveals the chemical composition of the experimental diets. The proximate components contained crude protein (23.10 %), crude fibre (4.18 %), ether extract (4.03%), calcium (1.50 %), phosphorus (0.58 %) and metabolizable energy (2910.3 kcal/kg) for starters mash while broiler finisher mash contained crude protein (21.40 %), crude fibre (5.01 %), ether extract (4.47 %), calcium (1.60 %), phosphorus (0.66 %) and metabolizable energy (3200.8 kcal/kg) respectively.

Chemical composition of *Daniellia oliveri* leaf

Table 3 shows the chemical composition of test ingredient. The chemical components of *Daniellia oliveri* leaf meal used for this study is 89.11 %, 18.95 %, 13.11 %, 4.78 %, 6.10 %, 93.9 %, 47.5 %, 28.1 % and 46.17 % for dry matter, crude protein, crude fibre, ether extract, ash, organic matter, neutral detergent fibre, acid detergent fibre and nitrogen free extract respectively.

Table 3 Chemical composition of *Daniellia oliveri* leaf meal

Parameters	% composition
Dry matter	89.11
Crude protein	18.95
Crude fibre	13.11
Ether extract	4.78
Ash	6.10
Neutral detergent fibre	47.50
Acid detergent fibre	28.10
Nitrogen free extract	46.17

Mineral composition of *Daniellia oliveri* leaf meal

The mineral composition of *Daniellia oliveri* leaf meal is presented in Table 4. The sample contained calcium, potassium, phosphorus, sodium, zinc, iron, magnesium, manganese, copper and cobalt at 12.17, 36.88, 42.21, 37.18, 6.41, 1.44, 44.52, 0.78, 0.51 and 0.10 (mg /100 g).

Table 4 Mineral analysis of *Daniellia oliveri* leaf meal

Minerals	Composition (mg/100g)
Macro nutrients	
Calcium	12.17
Potassium	36.88
Phosphorus	42.21
Magnesium	44.52
Sodium	37.18
Micro nutrients	
Iron	1.44
Manganese	0.78
Copper	0.51
Zinc	6.14
Cobalt	0.10

Phytochemical analysis of *Daniellia oliveri* leaf meal

The phytochemical analysis of *Daniellia oliveri* leaf meal is presented in Table 5. The phytochemical components revealed that alkaloids (1.52 mg/100 g), flavonoids (61.11mg/100 g), phenols (56.08 mg/100 g), condensed tannins (1.09 mg/100 g), hydrolysable tannins (2.66 mg/100 g), saponins (4.71mg/100 g), phytate (1.88 mg/100 g), steroids (0.04 mg/100 g) and oxalate (0.01 mg /100 g) respectively. Flavonoids have the highest value while oxalate has the least value.

Table 5 Phytochemical components of *Daniellia oliveri* leaf meal

Parameters	Composition (mg/ 100g)	*Safe recommended level
Alkaloids	1.52	2.13
Flavonoids	61.11	122.1
Phenols	56.08	88.3
Condensed tannins	1.09	2.56
Hydrolysable tannins	2.66	1.88
Saponin	4.71	7.02
Phytate	1.88	23.40
Steroids	0.04	0.10
Oxalate	0.01	0.54

EFSA (2014)

Growth performance of broiler chicks fed different levels of *Daniellia oliveri* leaf extract

The performance characteristics of broiler chicks given different levels of *Daniellia oliveri* leaf extract and neomycin is shown in Table 6. Final live weight (LW), weight gain (WG) and average daily gain (ADG) range between 1847.1 - 2139.3 g, 2099.2 – 1807.1g and 37.49g – 32.27g respectively. They were highest in T4 and T5, intermediate in T2 and T3 and lowest in T1 (P<0.05). Contrary to final LW, WG and ADG, feed conversion ratio (1.81 – 2.16) was lowest for T4 and T5, intermediate for T2 and T3 and highest (P<0.05) for T1. Daily feed and water consumption, which were (69.66 – 67.86 g) and (255.8 – 255.3 ml) respectively, were not (P>0.05) influenced by the treatments. While no mortality was recorded for T4 and T5, mortality was highest in T1 (10 %), intermediate in T2 (3 %) and lowest in T3 (P<0.05).

Table 6 Performance of broiler chicks given varying levels of *Daniellia oliveri* leaf extract

Parameters (g)	T1	T2	T3	T4	T5	SEM
Initial body weight (g)	40.00	40.20	40.00	40.00	40.10	0.01
Final live weight (g)	1847.1^{c}	1996.5^{b}	1980.3^{b}	2102.2^{a}	2139.3^{a}	14.95
Weight gain (g)	1807.1^{c}	1956.3^{b}	1940.3^{b}	2062.2^{a}	2099.2^{a}	48.77
Daily weight gain (g)	32.27^{c}	34.93^{b}	34.65^{b}	36.83^{a}	37.49^{a}	2.71
Feed intake (g)	3900.7	3822.1	3819.1	3800.8	3800.1	71.10
Average feed intake (g)	69.66	68.25	68.20	67.87	67.86	4.22
Feed:gain ratio	2.16^{a}	1.95^{b}	1.96^{b}	1.84^{c}	1.81^{c}	0.40
Mortality (%)	10.0^{a}	3.00^{b}	1.00^{c}	-	-	0.03
Total water intake (ml)	14,301	14,303	14,309	14,321	14,323	2.20
Daily water intake (ml)	255.3	255.4	255.5	255.7	255.8	8.14

Means in the same row with different superscripts are significantly different *(P*<0.05)

FCR, feed conversion ratio; SEM, standard error of mean

Hematological parameters of broiler chicks administered *Daniellia oliveri* leaf extract

Hematological parameters of experimental broiler chicks Table 7. PCV values ranged between (35.04 – 37.79 %), Hb (15.03 – 19.13 g/dl), RBC 2.84 – 3.80 (10^6/µl) and lymphocytes were higher (*P*<0.05) in T5 than in T1 and T2. MCV (105.7 – 112.5 fl), were lower (*P*<0.05) for T1 and T4 than for the other treatments. MCH (42.50 – 50.16 pg), were highest and lowest for T5 and T1 respectively (*P*<0.05). MCHC (43.20 – 48.54%) and basophils (2.10 – 2.31%) were lower (*P*<0.05) for T1 than for other treatments. WBC 29.26 – 34.97 (10^6/µl) and neutrophils (4.13 – 4.48 %) were highest (*P*<0.05) compared to other treatments. Monocytes (1.16 – 1.70 %) were lowest (*P*<0.05) in T1 relative to other treatments. Eosinophils (1.33 – 1.59 %) were lower (*P*<0.05) for T1 and T2 compared to other treatments.

Table 7 Effect of treatments on the hematological traits of broiler chicks

Parameters	T1	T2	T3	T4	T5	SEM
Pack cell volume (%)	35.51^{b}	35.04^{b}	36.51^{a}	36.86^{a}	37.79^{a}	0.06
Hemoglobin (g/dl)	15.46^{c}	15.03^{c}	16.16^{b}	17.01^{b}	19.13^{a}	0.05
RBC $(10)^6$/µl	2.91^{d}	2.84^{d}	3.05^{c}	3.64^{b}	3.80^{a}	0.02
MCV (fl)	105.7^{b}	112.1^{a}	112.5^{a}	109.0^{b}	111.2^{a}	0.54
MCH (pg)	42.50^{c}	47.46^{b}	46.88^{b}	47.69^{b}	50.16^{a}	0.29
MCHC (%)	43.20^{b}	48.54^{a}	46.62^{a}	47.73^{a}	47.75^{a}	0.31
WBC $(10)^6$ /µl	29.26^{c}	29.42^{c}	30.49^{c}	33.63^{b}	34.97^{a}	0.19
Neutrophils (%)	4.48^{c}	4.15^{c}	4.13^{c}	4.23^{b}	4.34^{a}	0.01
Monocytes (%)	1.16^{d}	1.24^{c}	1.60^{b}	1.69^{a}	1.70^{a}	0.94
Lymphocytes (%)	18.36^{d}	18.46^{d}	20.79^{c}	22.47^{b}	27.83^{a}	0.12
Basophils (%)	1.10^{b}	2.13^{a}	2.28^{a}	2.28^{a}	2.31^{a}	0.04
Eosinophils (%)	1.33^{b}	1.35^{b}	1.55^{a}	1.55^{a}	1.59^{a}	0.01

Means in the same row with different superscript are significantly different (*P*<0.05)

RBC, red blood cell; WBC, white blood cell; MCV, mean corpuscular volume; MCHC, mean corpuscular haemoglobin concentration; MCH, mean corpuscular haemoglobin

Serum biochemical parameters of broiler chicks administered *Daniella oliveri* extract

The serum biochemical indices of the experimental birds are presented in Table 8. Whereas total protein (4.75 – 5.06g/dl), globulin (2.61 – 2.92 g/dl) and α1- globulin (1.42 – 1.91g/dl) were lowest (*P*<0.05) for T1, triglyceride (1.33 – 1.63 mg/dl), cholesterol (119.5 – 164.5 mg/dl) and urea (30.40 – 31.74 mg/l) were highest (*P*<0.05) for T1 relative to other treatments. Albumin (2.14 – 2.22 g/dl) was higher (*P*<0.05) for T2 and T4 than for the rest of the treatments. α2-globulin (6.30 – 6.52 g/dl) was lowest (*P*<0.05) for T2 relative to other treatments. β-globulin (2.03 – 2.12 g/dl) and γ-globulin (6.27 – 7.12 g/dl), glucose (403.9 – 416.1mg/dl), conjugated bilirubin (0.49-0.57 µmol/l), total bilirubin (2.94 – 3.08 µmol/l), high density lipoprotein (114.7 – 124.3 u/l), low density lipoprotein (30.88 – 32.28 u/l) and creatinine (0.59 – 0.68 mg/l) were not affected by treatments (*P*>0.05).

Table 8 Effect of treatments on the serum biochemical parameters of broiler chicks

Parameters	T1	T2	T3	T4	T5	SEM
Albumin (g/dl)	2.14^{b}	2.22^{a}	2.12^{b}	2.15^{a}	2.14^{b}	0.01
Globulin (g/dl)	2.61^{c}	2.76^{a}	2.87^{b}	2.88^{b}	2.92^{b}	0.02
Total protein (g/dl)	4.75^{b}	4.98^{a}	5.00^{a}	5.03^{a}	5.06^{a}	0.02
α_{1}- globulin (g/dl)	1.42^{c}	1.66^{b}	1.96^{a}	1.94^{a}	1.91^{a}	0.03
α_{2}- globulin (g/dl)	6.38^{b}	6.30^{c}	6.53^{a}	6.49^{a}	6.52^{a}	0.02
β-globulin (g/dl)	2.07	2.21	2.03	2.03	2.04	0.06
γ-globulin (g/dl)	6.39	6.71	7.12	7.12	6.27	0.15
GLU (mg/dl)	403.9	406.2	409.6	409.8	416.1	1.69
TRY (mg/dl)	1.63^{a}	1.55^{b}	1.49^{c}	1.41^{d}	1.33^{e}	0.02
Cholesterol (mg/dl)	164.5^{a}	154.3^{b}	131.7^{c}	119.5^{d}	113.9^{d}	0.96
CB (μmol/l)	0.49	0.50	0.55	0.51	0.57	0.02
TB (μmol/l)	3.05	3.08	2.99	3.07	2.94	0.22
Creatinine (mg/l)	0.67	0.68	0.67	0.62	0.59	0.01
HDL (u/l)	124.3	116.5	114.7	119.7	116.8	1.41
LDL (u/l)	32.28	31.18	30.88	32.16	31.52	0.20
Urea (mg/dl)	31.74^{a}	31.19^{b}	30.48^{b}	30.40^{b}	30.63^{b}	0.12

Means in the same row with different superscripts are significantly different (P<0.05)

GLU, glucose; TRY, triglycerides; CB, conjugated bilirubin; TB, total bilirubin; HDL, high density lipoprotein; LDL, low density lipoprotein

Effect of *Daniellia oliveri* leaf extract administration on serum enzymes electrolytes of broilers chicks

The serum enzymes and electrolytes as influenced by *Daniellia oliveri* leaf extract are presented in Table 9. Alkaline phosphatase [ALP; 100.0-104.5 (iu/l)], alkaline serum transaminase [AST; 28.70 – 61.88 (iu/l)], serum glutamic pyruvic transaminase [SGPT; 152.12-193.4(iu/l)], serum glutamic oxaloacetate [SGOT; 90.88 – 128.2 (iu/l)) were lowest (P<0.05) in T1 than in other treatments. Serum electrolytes, bicarbonate [18.06 – 18.97(Mmol/l)], Ca [5.57 – 5.90 (mg/dl)], K [1.97 – 2.04 (mg/dl)], Na [140.7 – 147.1 (Mmol/l)] and chloride 70.81 – 72.34 (Mmol/l), were not (P>0.05) affected by the oral administration of *Daniellia oliveri* leaf extract.

Table 9 Effect of dietary treatments on the serum electrolytes and enzymes of broiler chicks

Parameters	T1	T2	T3	T4	T5	SEM
ALP (iu/l)	104.5^{a}	100.4^{b}	101.58^{b}	100.10^{b}	100.0^{b}	0.32
AST (iu/l)	61.88^{a}	56.22^{b}	45.89^{b}	31.44^{b}	28.70^{c}	2.44
SGPT (iu/l)	193.4^{a}	178.3^{b}	165.44^{c}	153.10^{c}	152.12^{c}	1.47
SGOT (iu/l)	128.2^{a}	107.1^{b}	100.8^{b}	99.76^{c}	90.88^{c}	0.55
BC (Mmol/l)	18.06	18.97	18.64	18.52	18.41	0.22
Ca (mg/dl)	5.90	5.89	5.57	5.99	5.86	0.07
P (mg/dl)	1.97	2.02	2.01	2.00	2.00	0.07
Na (Mmol/l)	147.1	140.7	145.1	142.6	144.5	0.04
Chloride (Mmol/l)	71.30	71.22	71.11	70.81	72.34	0.52

Means in the same row with different superscripts differ significantly (P<0.05)

ALP, alanine phosphatase; AST, alanine serum transaminase; ALT, alanine transaminase; SGPT, serum glutamic oxaloacetate; SGOT, serum glutamic oxaloacetate; BC, bicarbonate; P, phosphorus; Na, sodium

4. ANALYSIS

Feed given to the birds contained all necessary nutrients necessary for the growth of animals and it meet up with the stipulation by NRC (1994). Phytochemicals (bioactive components) such as tannins, saponins, alkaloids, flavonoids, phytate, steroids and oxalate were present in *Daniellia oliveri* leaves in amounts that could confer medicinal value. Antibacterial efficacy of phytogenics such as flavonoids and tannins against clinical organisms has been reported (Osuntokun *et al.*, 2016). Ojewuyi *et al.* (2014) also reported that plant bioactive compounds show medicinal activity as well as exhibit physiological activity and anti-inflammatory, antioxidant and membrane stabilizing property (Omale and Okafor, 2008).

The presence of alkaloids in significant amounts in *Daniellia oliveri* implies that it can be as analgesics, anti-malaria and stimulants (Faizi, 2003). Saponins are known to possess both antimicrobial (Soetan *et al*., 2006) and anti-inflammatory activities (Hassan et al., 2012). Studies have also reported the beneficial effects of saponins on blood cholesterol levels and stimulation of the immune system (Cheeke, 2000). Phenols are strong antioxidants which prevent oxidative damage to biomolecules such as DNA, lipids and proteins that play a role in chronic disease. Plant phenols may interfere with all stages of the cancer process, potentially resulting in a reduction of cancer risk (Hollman, 2001). All the phytochemical components of *Daniellia oliveri* were below the lethal levels. This is in conformity with the findings of EFSA (2004) on the safe recommended level of phytochemicals for birds.

The mineral composition of *Daniellia oliveri* leaf confirms the reports of Edeoga *et al.* (2005), that plants store most of their bioactive chemicals in several parts such as the leaves, seeds and roots. Calcium aids digestion and promote good growth and vigour by helping to regulate metabolism (Omale *et al*., 2009). Phosphorus has been reported to be good for bones and teeth formation and also needed for growth, repair of tissues and cells for the production of DNA and RNA (Ajibade and Fagbohun, 2010). Iron is an important component of haemoglobin (Moyo *et al*., 2011) while sodium plays a role in body fluid maintenance in the body tissue (Alinnor and Oze, 2011). Abijabe and Fagbohun (2010) also reported that magnesium is involved in production of cellular energy and the synthesis of nucleic acid and protein.

The improved the final live weight, total weight gain and average daily gain of the treatment birds relative to the control birds could be as result of better feed utilization by the birds. These findings are supported by the enhanced feed conversion ratio (FCR) of the birds when compared with the control birds given a conventional antibiotic (1.25 g/liter neomycin). Similar results were reported by Pascaline *et al.* (2019) who supplemented broiler diets with 2500 g/100 kg of feed with *Petiveriaalliacea* and observed substantial increased weight gain of birds. Among the treatment birds, birds on higher dose of DOE (30 and 40 ml/litre) gained more weight than those on lower dose (10 and 20 ml/litre), implying that higher dose enhanced feed utilization better than the lower dose. The improvement in the final live weight in the present study could be attributed to multiple effects of DOE on the digestive tract which resulted in an increase in gastric and pancreatic secretions, better digestion and absorption of nutrients with improved performance (Burrin and Janeczko, 2008). The improved FCR of treatment birds compared with the control birds indicates better conversion of feed to BW gain. Thus birds on the DOE treatments consumed less feed per unit of the BW gain. The explanation above also holds true for better FCR of birds on higher DOE dose than those on the lower DOE dose. The outcome is in concurrence with Gbore *et al.* (2016), who reported a significant improvement in the FCR of rabbits fed with 2 mg and 4 mg Jumbo cube/kg BW. Feed intake was not affected by the experimental diet; implying oral administration of DOE did not affect feed palatability and acceptability. It can thus be inferred that the concentration or dose of the DOE used in the current study was well tolerated by the birds and were below the threshold level at which feed intake and utilization for BW gain appreciable.

In general, some experiment with broilers (Hernández et al., 2004; Cross et al., 2003) did not find statistical differences in the performance parameters of birds fed infusion of severalphytogenics. Lee et al*.* (2003a) opined that the absence of effect on bird's performance may be related to the composition of the basal diet and/or to the environmental condition of the experiment. Ration containing phytobiotics limit the proliferation of pathogenic organisms in the gut because no substrate is left for bacteria growth, thereby reducing the antimicrobial potential of plant extracts. Similar observation was made when birds were raised under low immune challenge conditions or strict health control.

The absence of mortality in birds on 30 and 40 ml/litre suggests the ability and potency of the bioactive components of DOE to suppress pathogens that causes morbidity and mortality. It appears the higher doses (30 and 40 ml/litre) of DOE improved the concentration, efficacy and potency of the phytogenic substances because at lower doses (10 and 20 ml/litre) of DOE mortality was recorded, though lower than the mortality observed in control birds administered conventional neomycin antibiotic. The reduced mortality of birds given DOE compared with neomycin may be attributed to the bioactive chemicals in DOE. For instance, tannins have multiple biological activities, including antiviral and antibacterial properties attributed mainly to their antioxidant and antiradical activity (Santos-Buelga and Scalbert, 2000) and, in synergistic combination with other secondary metabolites (flavonoids, alkaloids, saponin and phenol),they help to reduce disease pressure on the flock, thereby increasing the opportunity for the flock to achieve target weight gain, better FCR and improved performance (Onibi *et al*., 2009).

The main hematological parameters such as RBC, Hb, PCV, MCV, MCH and MCHC including eosinophils, monocytes, lymphocytes and heterophils were higher in birds fed DOE compared with control (T1). However, basophils did not differ significantly ($P>0.05$) among the treatments. The PCV (22.9 - 41.0 %), Hb (11.60 – 13.68 g/dl) and RBC 4.21 – 4.84 (10^6/µl) reported in this experiment were within the range reported by Talebi *et al*. (2005) for heathy chicken. High RBC level in T5 is an indication of oxygen circulation. RBC are important indicators of anaemia, the present are within the recommended range for birds which removes the fears of imminent hazard that may be associated with the use of DOE (Olafadehan, 2011). Though the animals in T4 and T5 had a higher WBC, lymphocyte, eosinophil, monocytes and neutrophil counts compared with T1, however, the values were within the range reported for broilers (Islam et al., 2004). WBC, lymphocytes, neutrophils and monocytes were found to affect the immune system (Peters *et al*., 2011). Aster (2004) reported that monocytes develop into macrophages that function in the phagocytosis of large particles (fungi and protozoa) and production of antigens. According to Peters et al. (2011) PCV, Hb and MCH are major indices for evaluating circulatory erythrocytes and are significant in the diagnosis of anaemia and also serve as useful indices of the bone marrow capacity to produce red blood cells in mammals (Chineke *et al*., 2006). Hemoglobin plays a major part in oxygen transfer within the body to aid blood circulation in animals. Basophils and eosinophils play a role in regulating allergic and inflammatory processes and host defense responses against parasitic infections like helminthiasis and ectoparasitic infestation (Soetan*etal*, 2009).

Total protein, globulin and albumin of the birds used in this study were higher in groups fed DOE compared with control group. This is an indication that protein levels contained in the diet was enough to support normal protein reserves across the treatments. The similarity in the albumin content could be attributed to the comparable protein intake across the groups. Guache *et al.* (1991) reported albumin content to be specifically influenced by protein shortage. The albumin content and protein observed in this experiment were within the normal ranges reported by Ibrahim Albokhadaim (2012). Protein electrophoresis is an invaluable diagnostic tool to access avian physiological status by determining the relative or total amounts of α-, β- and γ-globulin fractions (Veerapandian *et al*.,2013), variation of these fractions is related to inflammatory processes, age and nutritional status. β- and γ-globulin values increased in T5 compared with the treatment fed neomycin. Veerapandian *et al*. (2013) reported that gamma globulin is formed in the reticuloendothelial system outside the liver or either in plasma cells or lymphocytes or both. Although the albumin, α and β globulin are related in some way to bacterial resistance, it is in the γ- fraction that antibodies are found, the values recorded in this study was in conformity with the normal range reported by Nazifi *et al.* (2011). The increase in blood sugar as the dietary DOE increased is not in agreement with the finding of Nazifi *et al* (2011) who reported that birds maintain a relatively constant blood sugar level even in low feed intake. However, the cholesterol level declines with increase in dietary DOE, this result validates the ability of DOE to reduce cholesterol level in poultry. Similar result was obtained by Offorjindu (2006) when albino rats were fed dried neem leaf at 20mg/kg feed.

Dietary DOE and neomycin did not significantly (P>0.05) affect the total bilirubin, creatinine, LDL, HDL and urea. The values decrease as the oral inclusion of DOE increases, though not at a significant level. However, all the values were within the range reported by Rodriguez et al. (2006). Creatinine is the bye product of protein metabolism (Iyayi and Tewe, 1998). Urea level is also reported to be influenced by dietary protein quality, quantity, bleeding time and is sensitive biomarkers employed in the diagnosis of renal damage (Akandeand Odunsi, 2012). Oladele *et al*. (2005) opined that urea is an indicator of dehydration in chickens rather than being used to detect diseases.

Calcium, sodium and potassium level reduced as the level of DOE increased across the treatment, birds in T5 had the lowest value compared with the control (P>0.05). Alanine transaminase (ALT) and alkaline phosphatase (ALP) were depressed as the level of DOE increased indicating no toxicity. However, age of birds is a factor influencing serum enzymes of birds. Oladele *et al.* (2005) reported that young chickens (8-10 weeks) had significantly greater ALP and ALT values than adult (50-80 weeks) birds. Similarly, Akintomide *et al.* (2018) revealed that AST value above 230 µl is abnormal for poultry. Values recorded in this trial fall within the normal ranges reported by Nazifi *et al.* (2011). SGPT and SGOT values decreased numerically and statistically as the level of DOE increased. T_5 had the least SGPT and SGOT value of 152.12 (iu/l) and 90.88 (iu/l) respectively while T_1 had a high value of 193.4 (iu/l) and 128.2(iu/l) respectively. SGPT and SGOT usually respond to the presence of toxic substances in the diet (Iyayi, 1994). The observed values therefore indicate the ability of the birds to tolerate the anti -nutritional factor (δ-Cadinene and polyalthic acid) identified in *Daniellia oliveri* leaf by Alatoni and Olutunji (2014). The significant effect of the test materials on SGOT and SGPT agrees with the findings of Sokunbi and Egbunike (2000) in rabbits but contrary to the reports of Iheukwuemere et al. (2002) in broiler chickens. The reduction in the concentrations of SGPT and SGOT in birds fed DOE compared with the treatment given neomycin suggests the hepatoprotective effect of *Daniellia oliveri* leaf extract.

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