

Article

Phytogenic potential of *Bryophyllum pinnatum* (Life plant) blend on haematological, serum and lipid profile of broiler chickens

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Abstract: *Bryophyllum pinnatum* contains different phytogenic substances that have antimicrobial and anti-inflammatory properties capable of enhancing the health and performance of broiler birds. A study was conducted to investigate the influence of *B. pinnatum* on the blood profile of broiler chickens. A total of 200 one- day-old broiler chickens were used for the study. The birds were assigned to 5 treatment groups of 4 replicates, having 10 birds per replicate. *B. pinnatum* blend was administered at a dosage of 10g/L, 15g/L and 20g/L of water for the treatment groups while, treatments 1 and 5 served as Positive and negative control. Data obtained were subjected to one way analysis of variance in a completely randomized design. Result revealed that *B. pinnatum* blend positively influence Packed cell volume, haemoglobin, red blood cell and white blood cell at both starter and finisher phases. Serum indices showed that liver functions were significantly altered by administration of *B. pinnatum* blend. Lowered blood cholesterol, triglycerides and high density lipoprotein was obtained from the treated groups. In conclusion *B. pinnatum* blend could serve as a natural Prophylaxis agent in broiler chickens up to 15g/l as it contains secondary metabolites capable of eliciting and enhancing immune defense in broiler chickens against infections.

Keywords: *Bryophyllum Pinnatum*, broiler, blood profile, health status, phytogenic plants

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1. Introduction

The poultry industries at large have continued to seek different innovative approaches that can enhance production and promote animal health in sustainable ways. Plant extracts contains bioactive substances that are capable of enhancing immune status in poultry birds. They have high beneficial effects on the performance and health condition of poultry birds (Jamroz and Kamel, 2002; Tucker, 2002; Alçiçek et al., 2003). Plants phytochemicals and their derivatives offer promising solution in mitigating the effect associated with the abuse of conventional antimicrobials in poultry farming. Among the phytochemical plants that can be explored as an alternative is *Bryophyllum pinnatum* (Azodo et al., 2021). *B. pinnatum* contains different pharmacological properties, which encompass bioactive compounds like flavonoids, alkaloids and tannins that possesses antimicrobial, antifungal, and anti-parasitic properties (Latif et al., 2019). Other secondary metabolites with remedial value identified in the leaves include glycoside, saponin, triterpenoid and phenolic composite (Fujita, 2000). In addition to its potential as an antimicrobial and anti-parasitic agent, *B. pinnatum* has shown promising characteristics of antibacterial and anti-inflammatory characteristics by modulating or regulating immune responses, which further enhances its role in poultry health. Research findings by Kumar et al. (2019) reported that *B. pinnatum* extracts could enhance immune functions, potentially contributing to lower faecal oocyte counts by bolstering the bird's natural defence mechanisms. *B. pinnatum* leaves have great medicinal value and can be used for treating internal parasites. The leaves possess various properties like haemostatic, refrigerant, emollient, mucilaginous, vulnerary, depurative, disinfectant and tonic (Kamboj and Saluja 2009). They are useful in wound healing, haemorrhoids, menorrhagia, discoloration of the skin, boils, sloughing ulcers, ophthalmic, burns, scalds, corns, diarrhoea, dysentery, headaches (Sofowora, 1993), vomiting, acute inflammations and bronchitis (Kamboj and Saluja 2009). The leaves have also been highly recommended for bleeding disorders and piles. This multiple action of supporting gut health and stimulating immune response positions *B. pinnatum* as a promising natural alternative to synthetic additives in broiler chickens' production by improving both disease resistance and overall health of the birds. However, several studies have established the effect of the plant *in-vitro* and in mice but there is paucity of information on the influence of the plant blend on the blood profile of broiler chickens. Hence, there is a need to explore the potential of *B. pinnatum* on poultry health. Therefore, this study aims to evaluate the influence of *B. pinnatum* on the blood profile of broiler chickens.

2. Materials and Methods

Experimental site

The experiment was carried out at the Directorate of University Farm, Federal University of Agriculture, Abeokuta, Ogun State, Nigeria. The average annual rainfall is 2048mm with minimum and maximum temperatures of 20.66°C and 35.48°C respectively. The vegetation lies in between the tropical rainforest and the derived savannah (Google Earth, 2024).

Sourcing and processing of plant material (test ingredient)

B. pinnatum leaves were freshly harvested from the university environment. After harvesting, the leaves were rinsed thoroughly to eliminate any foreign material that may be attached to them. The leaves were blended at a ratio of 200g of leaves to 400ml of distilled water using a Binatone Electric blender. The resulting mixture was collected and stored in an airtight container till further usage. The blend was then diluted with water per litre as described in the experimental design.

Ethics approval statement

All procedures used in this study adhered with the research ethics and guidelines of Animal Care Committee of the Federal University of Agriculture, Abeokuta, Ogun State, Nigeria.

Experimental Animals and management

A total of 200-day-old *Abor Acre* broiler chickens were purchased from a reputable hatchery. Before the arrival of the chicks, the pens were washed and disinfected. Heat source and light were provided using electric bulbs; the birds were raised in a deep litter system using wood shavings as litter material. Administration of the test ingredient commenced on the 5th day of the experiment and was administered for 5 days every other week throughout the experiment while, the control groups were administered (Enrofloxacin) antibiotics for 5 days at a dosage recommended by the manufacturer. The choice of administration periods for the test ingredients was based on previous studies of Adeyemo et al. (2024) in broiler chickens. The negative control groups were neither given antibiotics nor test ingredient. Routine vaccinations were administered according to standard procedures. The experiment lasted for seven weeks, the birds were fed with commercially produced pelletized broiler starter having 22% crude protein 2800kcal/kg Metabolisable Energy) while the broiler finisher diet had (19% crude Protein, 3000Kcal/ kg Metabolisable Energy). The chickens have access to feed and water ad libitum.

Experimental design

The chicks were randomly allotted to five treatment groups, each group comprising four replicates of 10 chicks per replicate, in a completely randomised design. **Control group:** Conventional antibiotics (Enrofloxacin antibiotic in water), **10g/L group:** 10g/L of *B. pinnatum* blend, **15g/L group:** 15g/L of *B. pinnatum* blend, **20g/L group:** 20g/L of *B. pinnatum* blend while the **Negative control group:** No antibiotics nor the test ingredient (0- VE).

Data Collection

Blood collection

Blood samples were collected at the end of starter phase (28th day) and (49th day) end of finisher phase of the experiment. Eight (8) birds were selected at random from four (4) replicate for haematological, serum and lipid profile analysis. Blood was collected through the wing vein using a 5ml disposable syringe. Blood sample of 3ml was dispensed into labelled sterile anticoagulant bottles for

haematological analysis while, the remaining 2ml of the blood was dispensed into labelled plain bottle for serum biochemical indices.

Determination of haematological Indices

The Packed cell Volume and Haemoglobin were determined using microhemacrit and cytomethaemoglobin methods respectively as outlined by Jain (1986). Red blood cells count was determined using haematocytometry method as outlined by Jain (1986). White blood cell was determined using Neubauer counting chamber according to (Jain 1986). The Mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and the mean corpuscular haemoglobin concentration were calculated using standard formulae (Samour, 2015).

Determination of serum parameters

Serum protein was determined using Biuret method according to the procedures of (Lanzarote et al., 2015). Albumin concentration was carried out using the method described by Buckley et al. (1976) using Biuret method. Alanine and Aspartate Aminotransferase were determined with the aid of spectrophotometer using commercial Randox kit. Serum glucose was determined calorimetrically using Glucose Oxidative Peroxidase GOD/POD method of Barham and Tinder (1972). Creatinine was analyzed using colorimetric method as described Heinegård and Tiderström, (1973). Serum globulin was estimated by subtracting the result of serum albumin from serum total protein.

Determination of blood lipid Profile

High-density lipoprotein concentration was determined using the method described in Albers et al. (1978), low-density lipoprotein (LDL) concentration was determined according to the procedures described by Assman et al. (1984). Total cholesterol concentration was determined according to the method described by Allain et al. (1974), while triglyceride concentration was determined by the method described by Bucalo and David (1973).

Statistical analysis

All data collected were subjected to one-way Analysis of variance (ANOVA), in a completely randomized Design (CRD). Significant ($P < 0.05$) means were separated using Duncan's Multiple Range test as contained in the same statistical package (SAS 2012).

Statistical Model:

$$Y_o = \mu + T_i + \varepsilon_{ij}$$

Where;

Y_o = Dependent variable

μ = Population mean

T_i = Effect of *Bryophyllum pinnatum* on the blood profile

ε_{ij} = Residual error

3. Results

The effect of *B. pinnatum* blend on haematological parameters of broiler Chickens at the starter phase is shown in Table 1. Packed cell volume, Haemoglobin, Red blood cell, white blood cell, Neutrophils, Lymphocytes, Eosinophil, mean corpuscular volume and mean corpuscular haemoglobin concentration were significantly ($p < 0.05$) influenced by the administration of *B. pinnatum* blend. However, there were no significant ($p > 0.05$) differences observed in Basophils, Monocytes and Mean corpuscular haemoglobin.

Table 1: Influence of *Bryophyllum pinnatum* blend on the haematological parameters of broiler chickens at starter phase.

PARAMETERS	0+ve	10g	15g	20g	0-ve
PCV (%)	22.00±0.00 ^a	23.50±0.70 ^a	24.00±1.41 ^a	16.00±1.41 ^b	15.00±0.00 ^b
Hb(g/dl)	7.50±0.00 ^a	8.15±0.07 ^a	8.05±0.63 ^a	5.85±0.07 ^b	5.40±0.14 ^b
RBC(10 ⁶ /L)	2.15±0.07 ^a	2.35±0.07 ^a	2.40±0.28 ^a	1.75±0.07 ^b	1.60±0.14 ^b
WBC(10 ⁹ /L)	13.10±0.84 ^b	14.65±0.63 ^a	13.85±0.07 ^{ab}	11.60±0.14 ^c	8.50±0.70 ^d
NEUT (%)	32.50±0.70 ^b	32.50±0.70 ^b	35.50±0.70 ^a	35.50±0.70 ^a	33.00±1.41 ^b
LYM (%)	66.00±1.41 ^a	63.50±0.70 ^{ab}	60.50±2.12 ^b	62.00±2.82 ^{ab}	66.50±0.70 ^a
EOS (%)	0.00±0.00 ^b	1.00±0.00 ^a	1.50±0.70 ^a	0.00±0.00 ^b	1.00±0.00 ^a
BAS (%)	0.50±0.70	0.00±0.00	0.50±0.70	0.00±0.00	1.00±0.00
MONO (%)	1.00±1.41	2.00±0.00	2.00±1.41	0.50±0.70	0.00±0.00
MCV(fl)	102.38±3.36 ^a	103.90±0.63 ^a	100.35±5.93 ^{ab}	93.95±0.62 ^b	100.10±0.14 ^{ab}
MCH(pg)	34.90±1.14	35.10±0.77	33.62±1.31	33.68±1.27	36.41±1.53
MCHC(g/dl)	34.09±0.00 ^{ab}	34.66±0.69 ^{ab}	33.52±0.67 ^b	34.05±0.92 ^{ab}	35.91±0.82 ^a

^{a,b,c} means on the same row differ significantly ($p < 0.05$); PCV- Packed cell volume, Hb- Haemoglobin, RBC- Red blood cell, WBC- White blood cell, NEUT-Neutrophils, LYM- lymphocytes, EOS- Eosinophils, BAS- Basophils, MONO- Monocytes, MCV- Mean corpuscular volume, MCH- Mean corpuscular haemoglobin, MCHC- Mean corpuscular haemoglobin concentration.

Significantly higher packed cell volume, haemoglobin and red blood cells were obtained from birds in control groups and birds administered 10g/L and 15g/L *B. pinnatum* blend compared to lower mean value obtained for birds on 20g/L and the negative control groups. Significantly higher white blood cell was obtained from birds administered 10g/L of *B. pinnatum* blend compared to lowest count obtained from the negative control groups.

Higher Neutrophils count was obtained from birds administered 15g/L and 20g/L compared to significantly lower neutrophils count obtained from birds in the positive, negative control and birds administered 10g/L of *B. pinnatum*. Significantly higher lymphocytes count was obtained from birds on positive and negative control groups compared to lower count obtained from birds administered 15g/L of *B. pinnatum*.

Higher eosinophils count was obtained from birds on 10g/L, 15g/L and birds in the negative control groups compared to lower eosinophils count obtained from birds in the positive control and birds administered 20g/L of *B. pinnatum*. Significantly comparable mean corpuscular volume (MCV, MCHC) and mean corpuscular haemoglobin concentration was obtained across all treatment groups.

Table 2 shows the effect of *B. pinnatum* blend on the serum biochemistry of broiler chickens at the starter phase. Glucose, Creatinine, Aspartate aminotransferase, and Alanine aminotransferase were significantly ($p < 0.05$) influenced by the administration of *B. pinnatum* blend while other parameters measured were not significantly ($p > 0.05$) influenced by the test ingredient.

Table 2: Influence of *Bryophyllum pinnatum* blend on the serum parameters of broiler chicken at starter phase

PARAMETERS	0+ve	10g	15g	20g	0-ve
T.PROT(g/dL)	5.05±1.48	4.80±0.14	5.00±0.42	5.40±0.14	5.20±0.14
ALB(g/dL)	2.80±0.00	2.45±0.07	3.05±0.07	3.25±0.77	2.65±0.35
GLOB(g/dL)	2.25±1.48	2.25±0.07	1.90±0.28	2.65±0.21	2.50±0.14
GLUC(mg/dL)	125.45±4.03 ^c	183.30±1.69 ^a	154.85 ±18.73 ^b	117.00±2.82 ^c	127.50±2.12 ^c
CREA(mg/dL)	1.99±0.33 ^{ab}	2.30±0.02 ^a	1.51±1.35 ^{ab}	0.70±0.14 ^{ab}	0.44±0.02 ^b
AST(U/L)	120.00 ±4.24 ^c	73.50±2.12 ^e	137.00±1.41 ^a	129.50±0.70 ^b	107.00±2.82 ^d
ALT(U/L)	45.50±9.19 ^{ab}	33.50±2.12 ^b	38.50±0.70 ^b	43.00±1.41 ^{ab}	52.50±3.53 ^a

^{a,b,c} means on the same row differ significantly ($p < 0.05$); T.PROT- Total protein, ALB- Albumin, GLOB- Globulin, GLUC- Glucose, CREA- Creatinine, AST-Aspartate aminotransferase, ALT- Alanine aminotransferase

Significantly ($p < 0.05$) higher glucose level was obtained from birds administered 10g/L of *B. pinnatum* compared to significantly lowered glucose level obtained from birds on positive control, negative control, and birds on 20g/L of *B. pinnatum* blend. Significantly higher creatinine level was obtained from birds administered 10g/L of *B. pinnatum* compared to significantly lower levels obtained from birds in the negative control groups.

Higher Aspartate aminotransferase was obtained from birds on 15g/L of *B. pinnatum* compared to significantly lower level obtained from birds on 10g/L of *B. pinnatum*. Significantly higher Alanine aminotransferase was obtained from birds in the negative control compared to lower mean value obtained for birds on 10g/L and 15g/L of *B. pinnatum*. Table 3 shows the effect of *B. pinnatum* blend on the serum lipid profile of broiler chickens at the starter phase. Significant ($p < 0.05$) differences were observed in all the parameters measured except low-density lipoprotein.

Significantly higher cholesterol level was obtained from birds administered 10g/L of *B. pinnatum* compared to lower levels obtained from birds administered 20g/L of *B. pinnatum*.

Table 3: Influence of *Bryophyllum pinnatum* blend on serum lipid profile of broiler chicken at starter phase

PARAMETERS	0+ve	10g	15g	20g	0-ve
CHOL(mg/dL)	169.45±11.38 ^{ab}	177.30±2.12 ^a	146.40±1.27 ^b	133.30±1.13 ^d	153.00±10.60 ^c
TRIG(mg/dL)	139.15±17.60 ^b	117.35±2.61 ^{bc}	128.45±0.77 ^{bc}	113.75±6.15 ^c	164.65±1.20 ^a
VLDL(mg/dL)	27.80±3.53 ^b	24.15±0.49 ^{bc}	22.75±1.20 ^c	26.55±1.34 ^{bc}	33.65±1.20 ^a
HDL(mg/dL)	82.30±21.35 ^a	77.20±1.97 ^a	79.45±0.35 ^a	57.60±2.54 ^{ab}	51.25±1.20 ^b
LDL(mg/dL)	35.90±3.81	76.90±0.84	55.50±34.22	63.95±2.19	44.30±1.13

^{a,b,c} means on the same row differ significantly ($p < 0.05$); CHOL- Cholesterol, TRIG- Triglycerides, VLDL- Very low-density lipoprotein, HDL- High-density lipoprotein, LDL- Low-density lipoprotein.

Higher triglycerides levels was obtained from birds in the negative control groups compared to lower levels obtained for birds on 20g/L of *B. pinnatum*. Significantly higher very low- density lipoprotein was obtained from the negative control groups compared to significantly lower mean values obtained from birds on 15g/L of *B. pinnatum* blend.

Higher high density lipoprotein was obtained from birds on positive control, birds administered 10g/L and 15g/L compared to lower mean value obtained for birds on negative control groups.

The effect of *B. pinnatum* blend on haematological parameters of broiler chickens at finisher phase is shown in Table 4. Packed cell volume, Haemoglobin, Red blood cell, white blood cell, Lymphocytes, Monocytes, mean corpuscular volume and mean corpuscular haemoglobin were significantly ($p<0.05$) influenced by the administration of *B. pinnatum* blend.

Table 4: Influence of *Bryophyllum pinnatum* blend on the haematological parameters of broiler chicken at finisher phase

PARAMETERS	0+ve	10g	15g	20g	0-ve
PCV (%)	27.00±4.24 ^{ab}	33.50±2.12 ^a	30.00±0.00 ^{ab}	22.00±1.41 ^b	35.00±7.07 ^a
Hb(g/dl)	9.35±1.48 ^{ab}	11.25±0.35 ^a	10.20±0.00 ^a	8.00±0.70 ^b	10.20±0.42 ^a
RBC(10 ¹² /L)	2.50±0.42 ^{ab}	3.20±0.00 ^a	2.25±0.07 ^{ab}	2.00±0.84 ^b	3.00±0.84 ^{ab}
WBC(10 ⁹ /L)	15.25±0.49 ^a	12.75±0.14 ^b	13.45±0.21 ^b	11.60±0.14 ^c	10.60±1.34 ^d
NEUT (%)	32.00±0.00	36.00±1.14	37.00±2.82	38.50±0.70	36.00±5.65
LYM (%)	64.50±0.70 ^{ab}	62.50±0.70 ^b	60.50±0.70 ^b	57.50±0.70 ^b	70.50±6.36 ^a
EOS (%)	1.50±0.70	0.00±0.00	1.00±1.41	1.00±0.00	1.00±0.00
BAS%	0.00±0.00	0.00±0.00	0.50±0.70	0.00±0.00	0.00±0.00
MONO (%)	2.00±0.00 ^{ab}	0.00±0.00 ^b	1.00±1.41 ^b	1.50±0.00 ^b	3.00±1.41 ^a
MCV(fl)	108.12±1.37 ^c	116.23±0.61 ^b	133.39±4.19 ^a	104.50±0.70 ^c	117.96±3.65 ^b
MCH(pg)	37.43 ±0.41 ^b	38.41±0.12 ^b	45.35±1.42 ^a	37.75±0.35 ^b	41.81±5.28 ^{ab}
MCHC(g/dl)	34.62±0.06	33.78±1.30	34.00±0.00	35.60±0.14	36.50±4.94

^{a,b,c} means on the same row differ significantly ($p<0.05$); PCV- Packed cell volume, Hb- Haemoglobin, RBC- Red blood cell, WBC- White blood cell, NEUT-Neutrophils, LYM- Lymphocytes, EOS- Eosinophils, BAS- Basophils, MONO- Monocytes, MCV- Mean corpuscular volume, MCH- Mean corpuscular haemoglobin, MCHC- Mean corpuscular haemoglobin concentration

Significantly higher packed cell volume, haemoglobin and red blood cells was obtained from birds administered 10g/L of *B. pinnatum* and negative control groups compared to significantly lower mean values obtained from birds on 20g/L of *B. pinnatum* blend. Significantly higher white blood cells were obtained from birds in the positive control groups compared to lower mean values obtained from birds on negative control groups. Higher lymphocytes count was obtained from birds in the negative control groups compared to significantly lower lymphocytes count obtained from birds administered 10g/L, 15g/L, and 20g/L of *B. pinnatum* blend.

Monocytes count was significantly higher in birds in the negative control groups compared to lower monocytes count obtained from birds administered *B. pinnatum* blend. Higher mean corpuscular volume was obtained from birds administered 15g/L of *B. pinnatum* blend compared to significantly

lower mean values obtained from birds in the positive control and birds on 20g/L of the blend. The effect of *B. pinnatum* blend on serum biochemical parameters of broiler chickens at finisher phase is shown in Table 5. The administration of *B. pinnatum* blend significantly ($p < 0.05$) influenced total protein, creatinine, and alanine aminotransferase while, other parameters measured were not significantly influenced by the administration of the blend.

Table 5: Influence of *Bryophyllum pinnatum* blend on the serum parameters of broiler chicken at finisher phase.

PARAMETERS	0+ve	10g	15g	20g	0-ve
T.PROT(g/dL)	7.30±2.26 ^{ab}	7.00±0.00 ^{ab}	8.85±0.77 ^a	6.95±0.07 ^{ab}	5.90±0.14 ^b
ALB(g/dL)	3.40±0.00	3.55±0.07	3.40±0.28	3.75±0.35	4.05±0.63
GLOB(g/dL)	3.90±2.26	3.45±0.07	5.45±0.49	3.35±0.07	2.85±0.91
GLUC(mg/dL)	100.10±32.24	87.05±2.19	113.50 ±13.85	106.60±1.55	77.70±2.54
CREA(mg/dL)	1.04±0.63 ^{ab}	0.33±0.02 ^b	1.04±1.30 ^{ab}	0.65±0.02 ^b	2.68±0.70 ^a
AST(U/L)	103.50±14.84	116.00±1.41	130.00±21.21	109.50±0.70	114.50±3.53
ALT(U/L)	32.50±3.53 ^{bc}	32.50±0.70 ^{bc}	39.00±5.65 ^{ab}	29.50±0.70 ^c	46.50±3.53 ^a

^{a,b,c} means on the same row differ significantly ($p < 0.05$); T.PROT- Total protein, ALB- Albumin, GLOB- Globulin, GLUC- Glucose, CREA- Creatinine, AST- Aspartate aminotransferase, ALT- Aalanine aminotransferase

Higher total protein was observed from birds administered 15g/L of *B. pinnatum* compared to lower protein obtained from birds in the negative control groups. Significantly higher creatinine levels were obtained from birds in the negative control groups compared to lower creatinine levels obtained from the birds administered 10g/L and 20g/L of *B. pinnatum* blend. Higher alanine aminotransferase was obtained from birds in the negative control groups compared to lower mean values obtained from birds administered 20g/l of *B. pinnatum* blend.

The effect of *B. pinnatum* blend on serum lipid profile of broiler chickens at finisher phase is shown in Table 6. Administration of *B. pinnatum* blend significantly influenced all serum lipid parameters measured. Significantly higher serum cholesterol was obtained from birds in the negative control groups compared to lower mean values obtained from birds administered 20g/L of *B. pinnatum* blend. Significantly higher triglycerides were obtained from birds in the positive and negative control groups compared to significantly lower mean values obtained 15g from birds administered 20g/L of *B. pinnatum* extract. Higher Very low-density lipoprotein (VLDL) was obtained from birds across all the treatment groups except birds administered 20g/L of *B. pinnatum* blend.

Table 6: Influence of *Bryophyllum pinnatum* blend on the serum parameters (lipid profile) of broiler chicken at finisher phase

PARAMETERS	0+ve	10g	15g	20g	0-ve
CHOL(mg/dL)	128.75±1.76 ^{ab}	121.65±1.62 ^b	114.50±5.37 ^c	106.75±3.18 ^d	131.90±5.09 ^a
TRIG(mg/dL)	197.75±3.18 ^a	156.00±1.41 ^b	162.60±2.68 ^b	91.00±0.70 ^c	188.00±10.60 ^a
VLDL(mg/dL)	37.25±3.88 ^a	32.15±0.49 ^a	36.20±4.66 ^a	17.90±0.56 ^b	37.60±3.32 ^a

HDL(mg/dL)	70.25±7.42 ^{ab}	64.80±0.98 ^b	56.10±2.26 ^b	58.20±1.27 ^b	85.45±14.49 ^a
LDL(mg/dL)	21.40±12.72 ^b	32.40±0.14 ^{ab}	19.85±7.00 ^b	41.15±0.49 ^a	46.90±1.97 ^a

^{a,b,c} means on the same row differ significantly ($p < 0.05$); CHOL- Cholesterol, TRIG- Triglycerides, VLDL- Very low-density lipoprotein, HDL- High-density lipoprotein, LDL- Low-density lipoprotein.

Significantly higher high-density lipoprotein was obtained from birds in the positive control and negative control groups compared to significantly lower higher density lipoprotein obtained from birds on 10g, 15g and 20g/L of *B. pinnatum* blend. Significantly higher low-density lipoprotein was obtained from birds in the negative control groups and birds administered 20g/L of *B. pinnatum* blend compared to significantly lower low-density lipoprotein obtained from birds in the positive control and birds administered 15g/L of *B. pinnatum* blend.

4. Discussion

Significantly higher packed cell volume, haemoglobin and red blood cell obtained at the starter phase from birds administered 10g/L and 15g/L might be attributed to the ability of *B. pinnatum* to enhance Hematopoiesis and improve the oxygen-carrying capacity of the birds during the early growth of the birds. This result corroborates the findings of Uhegbu et al. (2017) who reported significant difference in packed cell volume, haemoglobin and red blood cells of Albino rats administered *B. pinnatum* leaf extract. Also, these findings further corroborate the result of Ezeagu et al. (2017) who reported significant difference and higher packed cell volume, haemoglobin and red blood cell in Albino rats on normal/negative control administered ethanol leaf extract of *B. pinnatum*. Higher white blood cells obtained from birds on 10g/L (though still within the reference range reported by wikivet 2013) might be attributed to the ability of *B. pinnatum* blend to stimulate and improve the immunity status of birds compared to birds on negative control that are at higher risk of been susceptible to infection as a result of lower white blood cells. This result agrees with the findings of Soetan et al. (2013); Etim et al. (2014a) who reported that animals with high white blood cells can generate antibodies and a high degree of disease resistance. Significantly Higher Neutrophils count obtained at higher dosage of administration could be attributed to the immunomodulatory effect of *B. pinnatum* at higher dosages trying to fight some infection within the system of the birds. This result is contrary to the findings of Aprioku and Igbe (2017) who observed that *B. pinnatum* might depress primary immune response and becomes immunosuppressive. Higher lymphocytes obtained from the control groups (Positive and negative) might be an indication that birds are responding to susceptibility to infection as a result of lower white blood cells obtained making the birds to produce higher lymphocytes. This result is contrary to the findings of Aprioku and Igbe (2017) who observed no significant difference in the lymphocytes count of wistar rats administered subcutaneous aqueous *B. pinnatum* extract. Significantly comparable mean corpuscular volume (MCV, MCHC) and mean corpuscular haemoglobin concentration was obtained across all treatment groups, which suggest that the birds didn't suffer from any anaemic condition during the experiment. This suggests that *B. pinnatum* blend

did not negatively interfere with the production of MCV and MCHC. Significantly higher eosinophils count obtained from birds on 10g/L, 15g/L and negative control groups might be attributed to response to gut micro-organism proliferation, allergies or an inflammatory condition compared to lower count obtained at higher dosage of the administration. This result suggests that *B. pinnatum* is dose-dependent as higher inclusion levels reduced eosinophils count to counter effect of parasitism and other inflammatory conditions in the blood streams of the birds. This result further confirms *B. pinnatum* as an antiparasitic (Silvia et al., 1995, 1999) and anti-inflammatory agent (Almeida et al., 2000; Pal et al., 1991) that can be used for poultry birds.

The significant difference obtained at starter phase on the glucose level of the treated birds could be attributed to the presence of phytochemical constituents in the plant capable of lowering blood glucose in a dose dependent manner. This result corroborates the findings of Ezeagu et al. (2017) and Chimere et al. (2016) who observed reduction in the blood glucose of rats on *B. pinnatum* in a dose-dependent manner which was attributed to the phytochemical constituents inherent in the plant. Significantly higher creatinine levels obtained at the starter phase could be attributed to the protective ability of *B. pinnatum* blend on the kidney of the birds that enhance tubular reabsorption for the treated groups compared to the negative control groups. This result corroborates the findings of Uhegbu et al. (2017), who observed significant difference in creatinine levels of albino rats treated with the extract of *B. pinnatum* before exposure to petrol fumes. Higher aspartate aminotransferase obtained from the birds administered 15g/l of *B. pinnatum* could be attributed to the hepatoprotective function of the plant even at higher dosages. Research findings of Latif et al. (2019); Yadav and Dixit (2003) have reported the hepatoprotective activities of the liver thereby defending the liver against damage. Higher Alanine aminotransferase obtained at the starter phase from the negative control groups could be associated with infections or inflammation, thereby leading to increased alanine aminotransferase compared to other treatment groups that had better liver enzyme functions. This result further affirms the hepatoprotective effect of *B. pinnatum* on the liver of poultry birds. Lower serum cholesterol levels obtained at the starter phase from birds administered 20g/l might be attributed to the presence of quercetin and saponins capable of lowering cholesterol, thereby making *B. pinnatum* hypocholestromic plant in a dose- dependent manner. This result agrees with the findings of Ezeagu et al. (2017) who reported significant difference in total cholesterol of rats administered *B. pinnatum* extract. The significantly lower triglycerides obtained from birds administered 20g/l of *B. pinnatum* might be attributed to the presence of flavonoids and saponins capable of lowering blood triglycerides levels. This result is in tandem with the findings of Chattopadhyay and Bandyopadhyay (2005) who observed lower triglycerides levels in streptozotocin-induced diabetic rats administered *Azadirachta indica* leaf extract. Lower mean values obtained on very low-density lipoprotein from the treated groups might be attributed to the presence of bioactive constituents capable of lowering very low-density lipoprotein. This result further gives more credence to *B. pinnatum* blend as a plant that can help in preventing the buildup of fatty deposits in the bloodstream thereby preventing cardiovascular diseases (atherosclerosis). Significantly higher

high-density lipoprotein obtained in this study could be attributed to the ability of the plant extract to get rid of excess cholesterol in the blood stream to the liver where it will be excreted. This result is in tandem with the findings of Ezeagu et al. (2017) who observed increased high density lipoprotein in rat treated with *B. pinnatum* extract compared with the positive control groups

At the finisher phase lower pack cell volume, haemoglobin and red blood cells obtained from birds on 20g/L of *B. pinnatum* blend might be attributed to inhibition of hematopoiesis at higher dosage of administration at the finisher. This result is contrary to the findings of Aprioku and Igbe (2017), who observed no alteration in the pack cell volume of Wister rat administered aqueous *B. pinnatum* subcutaneously. The differences in the result obtained might be due to the route of administration used or differences in the animal species.

Lower mean value for white blood cells obtained from birds in the negative control groups could be attributed to immune suppression and fall in the defensive mechanism of the birds, since a decrease in white blood cell count might reflect a fall in the production of defensive mechanism to combat infection Campbell and Lasley (1975); Eheba et al. (2008). Higher lymphocytes count obtained from the negative control groups might be attributed to a response to immune suppression or proliferation of micro-organism in the gut as the birds were neither administered the test ingredient nor antibiotics, since lymphocytes has been reported to defend the body system when there is infection (Etim et al. 2014). This result is contrary to the findings of Aprioku and Igbe (2017), who observed no significant difference in the lymphocyte count of Wister rats administered aqueous *B. pinnatum* subcutaneously. Lower monocyte count observed from birds administered *B. pinnatum* might be attributed to the antimicrobial and antiparasitic potency of the *B. pinnatum* blend capable of protecting the immune system and preventing the birds from infection, compared to the negative control groups, which might be an indicated that the birds are under some levels of infection. The higher mean corpuscular volume observed at 15g/L of *B. pinnatum* blend showed that the plant leaves did not cause anaemic conditions to the birds. Lower corpuscular volume obtained for positive control and birds on 20g/L of *B. pinnatum* might be attributed to the partial erosion of the gut as a result of coccidiosis, causing partial bleeding.

The result obtained on serum indices at finisher phase showed that higher protein level was obtained from birds administered 15g/L of *B. pinnatum*. This result could be attributed to the availability and utilization of the protein content in the plant leaves. This result is in tandem with the findings of (Ogidi et al. 2019) who reported that *B. pinnatum* leaves contains protein which can be utilized as macroprotein. This result implies that utilization of the plant leaves cannot cause hypoproteinemia in broiler chickens. The significantly lower creatinine levels obtained from birds on 10g/L and 20g/L could be attributed to the nephroprotective mechanism of the *B. pinnatum* on the kidney. This result corroborated the findings of (Latif et al., 2019; Harlalka et al., 2007) who reported nephroprotective activity of the plants leaves. The significantly lower alanine aminotransferase obtained from birds on 20g/L of *B. pinnatum* could be attributed to the hepato protective activity of *B. pinnatum* blend thereby ensuring better liver functions. This result supports the findings of (Yadav and Dixit 2003)

who reported increased regeneration of hepatocytes and ability to defend the liver from hepatic damage proves its hepatoprotective mechanism of the leaves *in vitro*, *in vivo* and during histopathological studies. The significantly lower serum cholesterol obtained from the treatment groups might be attributed to the presence of lipids in the plant blend capable of lowering serum cholesterol in a dose-dependent manner. This result agrees with the findings of Ogidi et al. (2019) who reported that the plant leaves are low in lipid content and therefore can be used in controlling blood cholesterol as low lipid foods are said to reduce the level of cholesterol (Gordon and Kessel 2002). This result also supports the findings of (Adekunle et al., 2016) that reported hypocholesterolemic effect on blood serum when aqueous *B. pinnatum* was administered to rabbits. The significantly lower triglycerides obtained birds administered *B. pinnatum* might be attributed to the presence of quercetin specifically flavonoids that are capable of lowering blood triglycerides. This result support the findings of (Adekunle et al., 2016) that reported *B. pinnatum* leaves lowered serum triglycerides in rabbits.

The significantly lower VLDL obtained from birds on 20g/l of *B. pinnatum* might be attributed to the activity of bioactive substances present in plant leaves. This result is contrary to the findings of (Bassey et al. 2021) who reported no significant difference in the Very low-density lipoprotein of wister rats administered crude aqueous leaves extract of *B. pinnatum*. The significantly lower high-density lipoprotein in birds administered *B. pinnatum* might be attributed to the bioactive compounds present in the plant blend. This result is contrary to the findings of Bassey et al. (2021) who reported no significant difference in serum high density lipoprotein of albino rats administered crude aqueous extract of *B. pinnatum*. Significantly higher low *B. pinnatum* density lipoprotein obtained from negative control and birds administered 20g/L of *B. pinnatum* might be stress-triggered. This result corroborated the findings of (Bassey et al., 2021) who reported significant differences in low density lipoprotein of albino rats administered crude aqueous leaves extract of *B. pinnatum* extract.

5. Conclusions

The administration of *B. pinnatum* blend at the starter phase improved packed cell volume, haemoglobin, red blood cells and reduced eosinophils count at higher dosages thereby regulating inflammatory and reducing allergic reactions. Also, improved liver functions, reduced cholesterol and triglycerides and improved high density lipoprotein was obtained at the starter phase. Administration of *B. pinnatum* blend at moderate dosages (10 and 15g/L) at the finisher improved pack cell volume, haemoglobin and red blood cells. Lowered cholesterol, triglyceride, improved liver functions and better nephro protective effect was obtained at the finisher phase. Therefore, it can be concluded that *B. pinnatum* blend can be administered up to 15g/L in broiler chickens as the aqueous blend is capable of enhancing and improving immune functions of broiler chickens.

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