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Research Article

Effects of Dietary Supplementation of V*itex doniana* (Black Plum) Leaf Meal on Growth Performance, Blood Parameters, Carcass Traits, and Relative Organ Weights of Broiler Chickens

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ABSTRACT

Introduction: Evaluation of using plants, such as *Vitex doniana* leaf meal as an alternative feed resource in broiler diets decreases the feed costs. The current study examined how air-dried *Vitex doniana,* Black Plum Leaf Meal (BPLM) could affect broiler chicken growth performance, hematological parameters and serum biochemical indices, carcass yield, and relative organ weights.

Materials and methods: In a completely randomized design, 54 Arbor Acre broiler chicks aged 28 days old were allocated to two treatments. Each group included 27 chickens with three replicates (nine chickens per replication). The chickens were fed two diets, including basal diet (control) and basal diet with 5% dry matter BPLM for two weeks.

Results: The results of the nutrient composition of BPLM revealed 17.16% crude protein, 11.10% crude fiber, 1.70% ether extract, 7.10% ash, 39.0% nitrogen-free extract, 2413 metabolizable energy (kcal), 1.34% calcium, and 0.06% phosphorus. As a rich source of phytic acids, BPLM contains (14.69 mg/100g), tannins (3.23 mg/100 g), and oxalates (20 mg/100 g). The results showed that chickens fed a 5% BPLM-supplemented diet had lower feed intake but improved growth performance compared to the control. There was no significant difference in hematological and serum biochemical indices, carcass traits, cut-up parts, and relative organ weights.

Conclusion: Incorporating BPLM into a finisher broiler diet decreases feed intake but improves growth performance. In addition, the results showed no effect on hematological and biochemical parameters or relative organ weights in chickens supplemented with BPLM.

1. Introduction

The increasing expenses associated with traditional feed ingredients have led to elevated prices of processed feed and a significant rise in poultry product costs, ultimately contributing to a shortage of protein. Due to this limitation, investigating alternative ingredients to minimize the cost of chicken production and products has begun. Several multi-purpose tree Leaf Meals such as *Leucaenia leucocephala*[1], *Gmelina arborea*[2], *Azadirata indica* (Neem)[3], *Acacia augustissima*[4], *Ceiba pentadra*[5], *Manifera indica*[6], *Adansonia digitata*[7], *Gliricidia*[8], *Moringa oleifera*[9], *Mistletoe album*[10], and Psidium *guajava*[11] have been added in poultry rations to test their effects on growth performance, hematological, biochemical, carcass, and organs weight of broilers at different inclusion levels.

One alternative ingredient that could also breach the research gap is the *Vitex doniana* plant (black plum leaf). Black Plum Leaf Meal (BPLM) has not received attention as a potential supplement in broiler diets. *Vitex doniana* leaf meals have been reported as rich sources of protein, vitamins, and minerals[12,13]. Plant leaves possess a substantial amount of protein and encompass diverse physiologically active elements that have the potential to enhance growth and overall well-being in broiler chickens[14]. Nevertheless, leaf meals' high fiber content and

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anti-nutritional characteristics may restrict their inclusion in broiler diets[13]. By incorporating enzymes as dietary supplements and implementing various processing techniques such as fermentation, air-drying, boiling, and cooking, it is possible to unlock the potential benefits of bioactive components found in leaf meals without any detrimental effects on broiler production characteristics[3]. This study investigates the impact of Bioactive Plant Leaf Meals (BPLM) on growth performance, hematological and biochemical indicators, and carcass and organ weight parameters in broiler chickens.

2. Material and Methods

2.1. Ethical approval

The chickens were handled following the Nigeria Institute of Animal Science welfare and ethics committee (Act No.26 0f 2007) and with permission of the University ethics and welfare committee (FUW/AN/ET/22/005), Federal University Wukari, Taraba State, Nigeria.

2.2. Location of study

The study was conducted at the Poultry Teaching and Research Farm Unit, Federal University Wukari, Taraba State, Nigeria. Wukari is situated at latitude 7° 51' N and longitude 9° 47'E. The savannah zone dominates the vegetation in the area, characterized by two primary climatic seasons. The wet and rainy season commences around March or April and extends until October, while the dry season prevails from November to March or April[15].

2.3. Collection of Vitex doniana

During the wet season (September), fresh leaves of *Vitex doniana* (black plum) trees were collected from the Federal University Wukari (Nigeria) Protected Forestry and Wildlife field. The leaf samples were identified at the University of Agriculture and Life Sciences Department of Forestry and Wildlife (Nigeria). After 12 days, the freshly gathered leaves were diced to decrease particle size and surface area for speedy air-drying in jute bags in a well-ventilated open shed shielded from rain and direct sunshine. The leaves were then ground in a hammer mill to pass through a 2-mm filter and stored in jute bags for future feed compositions.

2.4. Experimental diets

Two finisher diets were formulated to contain 0% BPLM as a control and 5% BPLM inclusion level (Tables 1).

2.5. Experimental design

Ibadan (Nigeria) supplied fifty-four (54) male 28-day-old Arbor-Acre broiler chick strains. The chickens were raised on commercial feed for four weeks before being randomly assigned to one of two dietary treatments in a completely randomized design, with each treatment having 27 chickens, replicated three times (9 chickens each), and raised in a traditional deep litter poultry house with concrete floor, dwarf walls, and zinc coated metal roofing with wire nettings to improve adequate ventilation and protection. The chickens were fed experimental diets for 14 days and allowed unlimited water access. The chickens were given the Newcastle disease vaccine (Lasota)[16] on days 7 and 19, while the Gumboro vaccine[17] was administered on the 14 and 28 days. Multivitamins, minerals, and glucose were administered in drinking water on arrival. Anti-coccidial drug (Pantacox®, 1 g/l of drinking water)[18] and antibiotic (Ciprofloxacin, 10 mg/kg-1 body weight)[19] were given to the chickens in drinking water on the alternate week.

2.6. Performance

The authors of the current study calculated feed intake (FI), body weight gain (BWG), and feed conversion ratio (FCR) at day 42 of chickens' age. Feed intake was calculated by subtracting the amount of feed supplied from the excess amount[3]. The difference between the beginning and final weights determined the body weight gain[4]. The mean body weight was estimated by taking the average weight of the hens during the experiment. The feed conversion ratio was determined as the ratio of feed intake to chicken live weight growth[5].

2.7. Blood sampling

Blood samples were obtained from one chicken randomly selected from each replication per treatment to

Table 1. Composition of experimental finisher diets of broiler chicken

Ingredients	Dietary inclusion level	
	0%	5%
Maize	55.5	51.5
Maize Offal	5.0	4.0
Black plum leaf meal	0.0	5.0
Fish meal	1.5	1.5
GNC	32.0	32.0
Limestone	2.0	2.0
Bone meal	3.0	3.0
*Premix	0.25	0.25
Methionine	0.25	0.25
lysine	0.25	0.25
Salt	0.25	0.25
Total	100	100
Calculated analysis (% DM basis)		
CP	18.39	18.41
ME (Kcal/kg)	2950	2952
EE	3.90	4.00
CF	4.03	4.50
Ca	1.83	1.84
P	0.78	0.77

*Premix supplied (kg-1 diet): Vitamin A (15,000 I.U); Vitamin D3 (3,000 I.U); Vitamin E (30 I. U): Vitamin K (2.5mg); Thiamin (2mg); Riboflavin (6mg); Pyridoxine (4mg); Niacin (40mg); Cobalamin (0.02mg); Pantothenic acid (910mg); Folic acid (0.06g); Iron (0.024g); Copper (0.006g); Iodine (0.0014g); Selenium (0.24mg); Cobalt (0.024mg); Antioxidant (0.125g); +Anti-mold (Anti-mycotoxin). DM: Dry matter, CP: Crude protein, CF: Crude fiber, EE: Ether Extract, ME: Metabolizable energy, GNC: Ground nut cake

examine hematological and serum parameters at day 42 of chickens' age. A sterile needle was used to pierce the right jugular vein and extract blood into the syringe. Five mL of blood were taken and placed in labeled sterile vials with EDTA (Ethyl diamine tetra acetic acid) powder as an anticoagulant. These samples were utilized in the laboratory (Nigeria) to assess hematological characteristics such as Red Blood Cells (RBC), White Blood Cells (WBC), Packed Cell Volume (PCV), and Haemoglobin Count (Hb)[20].

2.8. Serum biochemical indices

Five mL of blood samples were taken at the end of the experiment (day 42 of chickens' age) per replicate into another labeled sterile bottle without anticoagulant to test serum biochemical elements such as total protein, albumin, globulin, creatinine, triglyceride, cholesterol, aspartate amino transferase (AST), and alanine amino transferase (ALT) by commercial kits according to manufacturer instructions[20].

2.9. Carcass analysis

At the end of the experiment, one broiler chicken from each replicate (three per treatment) was randomly selected and starved for 12 hours before being slaughtered without anesthesia, bled, and de-feathered before being scalded and eviscerated. The live weight, plucked weight, bled weight, dressed weight, cut-up parts, and organs were weighed and measured, and expressed as a percentage of live weight at day 42 of chickens' age.

2.10. Diet assay

The BPLM and experimental diets were analyzed for the proximate composition of moisture, crude protein, crude fiber, ash, and ether extract[21].

2.11. Statistical analysis

Analysis of variance (ANOVA) was used in this study for analyzing the data, and the Least Significant Differences (LSD) was used to separate the means of the different treatments by using computer software IBM SPSS Statistic version 20[22]. The $p < 0.05$ was considered a significant difference between the groups.

3. Results

The experimental finisher diets showed that nutrient compositions for crude protein were iso-nitrogenous with a range value of 19.23-19.40%. The dry matter (93.05%), crude fiber (9.10%), ether extract (11.55%), and ash (6.55%) contents were higher in dietary 5 % BPLM inclusion compared with the control diet (Table 2). The analyzed results of the nutrient composition of BPLM are presented in Table 2 with 95% dry matter (DM), 17.16% crude protein (CP), 11.10% crude fiber (CF), 1.70% ether extract (EE), 7.10% ash, 39.0% Nitrogen free extract (NFE), 2413 metabolizable enegy (ME, kcal), 1.34% Ca**,** and 0.06% P. BPLM is a rich source of phytic acids (14.69 mg/100g), tannins (3.23mg/100g) and oxalates (20mg/100g) presented in Table 3. The performance of finisher broiler chickens fed experimental diets are presented in Table 4. The results showed no significant differences in final live weight, weight gain, and feed conversion ratio ($p > 0.05$). However, chickens on 5% BPLM showed higher body weight, weight gain, and lower feed conversion ratio with decreased feed intake. Results of the percentage carcass, cut-up parts of breast, thigh, wing, back, and organ weights were not significantly influenced by dietary BPLM-supplemented treatments (Table 5, $p > 0.05$). However, the

Table 2. Proximate composition of analyzed black plum leaf meal and experimental finisher diets (% Dm basis)

Sample	DM	CP	CF	EE	Ash	NFE	ME(Kcal)	Ca	P
Control diet	92.35	19.23	7.00	7.75	6.20	59.82	ND	ND	ND
Diet (5% BPLM)	93.05	19.40	9.10	11.55	6.55	53.45	ND	ND	ND
BPLM	95.40	17.16	11.10	1.70	7.10	58.00	24130	1.34	0.06

BPLM: Black plum leaf meal, DM: Dry matter, CP: Crude protein, CF: Crude fiber, EE: Ether Extract, NFE: Nitrogen free extract, ND: Not determined, ME: Metabolizable energy

Table 3. Analyzed anti-nutrients in black plum leaf meal (mg/100g)

Sample	Phytic acid	Tannins	Oxalate
BPLM	14.69	3.23	20.00

BPLM: Black plum leaf meal

Table 4. Performance of broiler chicken (day 42 of chickens' age) fed Black Plum Leaf Meal diets

Parameters	Control	5 % BPLM	SEM
Initial body weight (g)	654.63	676.85	18.27
Final body weight (g)	1941.67	2081.50	35.89
Body weight gain (g)	1287.03	1404.65	48.39
Daily weight gain (g)	36.77	40.19	1.38
Total feed intake/bird (g)	3200.00	3108.00	49.27
Daily feed intake/bird (g)	91.42	88.71	1.41
FCR	2.48	2.21	0.06

BPLM: Black plum leaf meal, SEM: Standard error of mean, FCR: Feed conversion ratio

Table 5. Carcass, cut-up parts and organs characteristic of broiler chickens (day 42 of chickens' age) fed Black Plum Leaf Meal

Parameters	Control	5 % BPLM	SEM
Live weight of birds (g)	1953.33	2099.67	40.03
Carcass (%)	74.64	75.71	1.36
Thigh (%)	14.38	15.76	0.23
Wing (%)	13.33	14.29	0.03
Breast meat (%)	25.13	25.71	1.06
Back (%)	19.49	20.00	0.44
Neck (%)	2.56	2.86	0.03
Head (%)	2.56	2.62	0.02
Shank (%)	2.71	2.84	0.05
Liver (%)	1.83	1.96	0.03
Heart (%)	0.49	0.51	0.12
Kidney (%)	0.20	0.22	0.12
Proventiculus (%)	0.60	0.62	0.02
Pancreas (%)	0.25	0.29	0.02
Gizzard (%)	3.48	4.00	0.04

SEM: Standard error of mean, BPLM: Black plum leaf meal

chickens on a 5 % BPLM-supplemented diet recorded higher numerical percentage values in all the carcass, cut-up parts, and organ weights of the liver, pancreas, kidney, proventriculus, gizzard, and heart. The results of the hematological and biochemical indices showed no significant differences in all the measured parameters ($p > 0.05$, Table 6).

Table 6. Blood constituents of experimental finisher diet (day 42 of chickens' age) fed Black plum leaf meal in broiler chickens

Parameters	Dietary treatments		SEM
	Control (0% BPLM)	5 % BPLM	
Hematological indices			
PCV (%)	31.00	30.00	1.11
RBC (× 10^{12}/L)	4.19	4.56	1.09
Hb (g/dl)	13.32	13.50	0.21
TWBC (× 10^{9}/L)	9.31	10.01	0.23
Neutrophils (%)	56.57	60.73	1.61
Lymphocytes (%)	8.44	9.37	0.23
Eosinophils (%)	1.33	1.80	0.32
Monocytes (%)	2.30	2.56	0.47
Biochemical indices			
Total protein (g/l)	4.23	4.83	0.12
Albumin (u/l)	2.36	2.00	0.48
Urea (mmol/l)	4.54	4.89	0.19
Creatinine (Umol/l)	43.37	45.16	0.44
Triglyceride (mmol/l)	3.22	3.45	0.21
Cholesterol (mmol/l)	4.21	4.26	0.14
Glucose (mmol/l)	8.56	10.98	1.10
Aspartate amino transferase (u/l)	91.39	97.70	2.18
Alanine amino transferase (u/l)	13.63	14.77	0.74

SEM: Standard error of mean, RBC: Red blood cells, TWBC: Total white blood cells, PCV: Packed cell volume, Hb: Haemoglobin Count

4. Discussion

The outcomes of the analyzed experimental diets align with the recommended range for broilers in tropical regions[23]. The crude protein content (17%) and crude fiber content (11.10%) in the analyzed diets exceeded the previously reported value ranges of 8-11% for crude protein and 2.75-7.42% for crude fiber[12, 13, 24]. The NFE (58%) align with earlier reports[12]. In this study, the ash content was 7.10%, which was higher than the 1.63% reported in study[12] but lower than the 9.51% reported in another study[13]. The EE (1.7%) obtained in this study is lower than 2.52-2.92% reported by some authors[12, 13]. The current study's Ca (1.34%) content level was higher than the value of 0.73% obtained in another experiment[12]. Likewise, this study's ME content is 2413 kcal/kg, slightly lower than the 2691 kcal/kg reported by a study[13]. The phytic acid value of 14.96 mg/100g obtained in this study was lower than 96.14 mg/100g reported by some authors[13]. Conversely, the tannin content of 3.23 mg/100g in the present study was higher than that of 1.45 mg/100g reported in previous reports by the same authors. The variations in nutrient compositions, mineral content, anti-nutrients, and ME from the previous study could be attributed to the age of maturity of the leaf, soil type, treatment methods, and analytical procedures.

The improved growth performance of chickens on 5% BPLM supported the finding that green leaves naturally contain various active chemicals such as phenols, flavonoids, alkaloids, saponins, tannins, terpenoids, and steroids that are helpful to both human and animal health[9,13,25,26]. The leaf's phenolic chemicals may serve as antioxidants, antibacterial agents, appetite stimulants, growth promoters, and immune stimulators[27]. Polyphenols in summer-autumn tea leaves also enhanced the oxidative state and immunological competencies of broiler chickens[28]. The improved growth performance of chickens fed 5% BPLM compared to controls was consistent with previous research indicating that leaf meals increased the production and activity of digestive enzymes and improved broiler intestinal morphology (villi development), resulting in improved nutrient digestibility and utilization[29,30]. Similarly, the findings of this study are corroborated by previous studies indicating the improved gut microbial environment, immunological responses, and physiological circumstances of chicks given leaf meals may be attributable to enhanced broiler health performance[30,31].

Improvements in chicken carcass characteristics and growth performance after feeding 5% BPLM also supported research that showed *vitex* leaf to be a rich source of vitamins and minerals that may have helped to boost relative nutrient metabolism[13]. The decrease in feed intake observed in chickens fed 5% BPLM (Black Pepper Leaf Meal) can be attributed to the high fiber content in the diet, as indicated by Table 2.

The percentage carcass values of 74.64 and 75.71% obtained in this study were comparable with the range values of 74.82-77.39% reported for broilers[32]. The higher relative organ weights observed in the gizzard, liver, heart, and kidney of chickens fed BPLM (Black Pepper Leaf Meal) as a dietary supplement may be attributed to changes in live body weight. This is because the chickens' surface area and live weight influence the quantity of feathers and visceral organs required[33,34]. External and internal offal percentages tend to rise as animal slaughter weight rises[35]. These findings support previous findings that organ weights indicate nutritional retention in chickens[36]. Similarly, the greater proportion of gizzard weights in hens

fed 5% BPLM diets accords with previous results that high fiber in diets usually leads to gizzard enlargement[33]. The enhanced performance might be attributed to enough crude fiber levels, which tend to excite the colon, resulting in higher peristaltic movement and enzyme synthesis, leading to better digestion. This result is consistent with previous findings[36,37].

The lack of significant differences observed in the hematological and biochemical indices measured between the treatment groups suggests that the administration of BPLM (Black Pepper Leaf Meal) had no noticeable effects on the health status of broilers. PCV, Hb, RBC, and WBC are known to be positively correlated with protein quality and protein level[38]. The authors reported that decreased red blood cell count is usually associated with low-quality feed and protein deficiency. The PCV, RBC, and Hb values are within the established range[39].

The insignificant variations in serum urea, creatinine, and other measured parameters in this study indicate better protein and balanced amino acid concentration in both diets. The results of the serum biochemical indices and enzyme alanine amino transferase (ALT) and aspartate amino transferase (AST) in this study collaborate with previous reports, which reported similarities among the control and 10% *Vitex* leaf meal inclusion in the diet of cockerel chicks[13]. The plane of nutrition affects animals' ALT and AST values, with high values indicating poor diet or toxic factors.

5. Conclusion

The study concluded that BPLM is a potential feed resource that can be incorporated into the finisher broiler diet. The supplementation of broiler chicken diet with *Vitex* leaf caused less feed intake but improved FCR, and carcass yield without adverse effects on hematological and serum biochemical indices. Further studies on increasing the inclusion level of BPLM in broiler chicken diets are recommended.

Declarations

Competing interests

The authors declare that they have no conflict of interest.

Authors' contributions

Obun Cletus Otu conceived, designed, drafted the article, and revised it critically for important intellectual content; Madaki Sanda Zumunta and Rapheal Wafar have acquired data, analysis, and interpretation. All the authors reviewed the final draft and agreed before submission.

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Availability of data and materials

The authors will provide all necessary data to the editor upon request.

Ethical considerations

All authors have reviewed the ethical problems (including plagiarism, consent to publish, misconduct, data fabrication and falsification, multiple publishing and submission, and redundancy).

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