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HAEMATOLOGICAL AND BIOCHEMICAL CHARACTERISTICS OF FINISHER BROILER CHICKENS FED ROASTED BAMBARA NUT (*Vigna subterranea* L.)-BASED DIETS

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ABSTRACT

The study was conducted to investigate the haematological and biochemical parameters of finisher broiler chickens fed roasted Bambara nut-based diets. A total of 96-day-old Marshal Strain broiler birds were assigned to four treatments (0, 10, 20 and 30%) replicated four times in a completely randomized design (CRD) comprising of 6 birds in each. Bambara nut seeds were roasted, dried and milled into fine powdery roasted Bambara nut meal. The meal was then incorporated in the diets at 0, 10, 20 and 30% inclusion levels. Samples were then analyzed for proximate composition. At the end of the 8 weeks feeding trial, three birds were selected on the basis of average pen weight from each group and used for the determination of blood indices. Data collected were subjected to analysis of variance (ANOVA) using statistical analysis system (SAS) package. Results revealed no significant ($p>0.05$) differences in most of the parameters measured. However, there were significant ($p<0.05$) differences in red cell distribution width (RDW), and monocytes (MON) levels across the treatments. The result also indicated that RDW% levels decreased with increasing levels of roasted Bambara nut meal (RBM). The result of serum calcium revealed significant ($p<0.05$) differences across the treatments where T4 (30%) presented the highest value (2.43mmol/L). It was concluded that inclusion of Bambara nut meal in the diets of finisher broiler chickens at 30% enhanced the levels of RDW and serum calcium.

Keywords: *Bambara nut; broiler chickens; haematology; biochemical indices*

INTRODUCTION

Feed is an important aspect of poultry production (Adenkola *et al.*, 2013). Adamu and Ubosi (1998) reported that feed is the most top among the factors militating the emergence and development of poultry in Nigeria. The need for feed ingredients, which will reduce the cost of production, is the basis for newest ingredients that are been brought to limelight in livestock feed and production research. This is because man and his livestock are in competition for basic ingredients and such ingredients are not usually produced in sufficient quantities locally (Oluyemi *et al.*, 1978; Omojola and Adesehinwa, 2007). Therefore, availability of feed thus becomes the key factor limiting poultry production. There is a serious shortage of feed ingredients such as wheat, corn, soybeans, etc. these days because of rapidly increasing human population. There is, therefore, an urgent need to close the gap between this explosive population increase and the availability of food. The major locally available plant protein sources commonly used in poultry feed production are soybeans meal and groundnut cake. Soya bean meal has been reported to be scarce and expensive while groundnut cake has been reported to have aflatoxins (McDonald *et al.*, 1998). It is therefore necessary to search for cheaper and locally available replacement alternatives such as Bambara nut. Amaefule and Ironkwe (2007) reported that Nigeria produces approximately 100,000 tonnes of Bambara peanuts per year. Consequently, this crop may be promising in poultry nutrition in Nigeria. Inclusion of feed ingredients at normal or required level will enhance egg and meat production (Chatterjee and Rajkumar, 2015).

Haematological parameters have commonly been used as indicators of physiological conditions and nutritional deficiency in chickens. Onyeyili *et al.* (1991) stressed on the importance of haematological evaluation in poultry as a valuable aid in the diagnosis of many diseases and in determining the exact of damage to blood cells. Dietary components affect the blood profile of healthy birds. It is often very difficult to assess the current health status of animals without detailed examination of blood (Amakiri *et al.*, 2009). Examination of blood provides the opportunity to clinically investigate the presence of several metabolites and other constituents in the body and it plays a vital role in the physiological, nutritional and pathological status of the animal (Doyle and William, 2006). Therefore, whatever affects the blood, either nutrition or drugs will certainly affect

the entire body adversely or moderately in terms of health, growth, maintenance and reproduction. This study was conducted to investigate the haematological and biochemical parameters of finisher broiler chickens fed roasted Bambara nut-based diets.

MATERIALS AND METHODS

Study area

The experiment was conducted at Poultry Unit of Teaching and Research Farm of Animal Science Department, Faculty of Agriculture, Bayero University, Kano, Nigeria. The farm is located at longitude E008°25.420' and latitude N11°59.039' in the semi-arid zone of Nigeria. Kano lies on longitude 9°30' and 12°30' North and latitude 9°30' and 8°42' East on an elevation 468m (NiMet, 2014).

Source and processing of Bambara nut meal

The Bambara nut seed was procured from Dawanau International Grains Market of Kano-State, Nigeria at reasonable price cheaper than other conventional protein sources like soybean. The procured Bambara nut seed was processed in order to get rid of the anti-nutritional factors (protease inhibitor, haemaglutinins, tannins, cyanogenic glycosides and flatulence factors) and to improve its palatability. The stones and other foreign materials were removed and was put inside sack to de-shell manually by using stick then winnowed to remove the chaff in order to have pure Bambara nut grain. The seeds were then washed with tap water and sun-dried, frying pan was placed on a low flamed gas cylinder (which was regulated to approximately 80°C) then the sun-dried Bambara nut was placed in turns for roasting in 30 minutes, adequate turning was ensured at interval to prevent it from burning.

Diet formulation

Diets were formulated according to the Nigeria Institute of Animal Science (NIAS) optimized procedures. The Bambara nut seeds were roasted, dried and milled into fine powdery roasted Bambara nut meal. The experimental birds were fed commercial diets for the first four weeks and the experimental diets were fed to them at the finisher stage (5-8 weeks). The diets were formulated and manufactured to meet the nutrient requirement of the birds. The gross composition of the experimental diets is presented in Table 1.

Birds and experimental design

A total of 96-day-old broiler chicks (Marshal Strain) were purchased from Anadariya Farms in Kano and were used for the experiment. The birds were raised together for four weeks on deep litter pen, after which they were randomly allotted into four dietary treatments. The experiment was laid in a completely randomized design (CRD) with four treatments consisting of 24 birds per treatment and each treatment was further divided into four (4) replicates which comprised of six (6) birds per replicate. The four treatments were coded as T_1, T_2, T_3 and T_4 in which RBM was incorporated at 0, 10, 20 and 30% respectively.

Blood collection and Serum Biochemistry Determination

Blood samples were collected from both the brachial and jugular vein using 5 mL sterilized disposable syringe and needle. In order to minimize the standard error in values, the animals were fasted for twelve hours (12 hrs) prior to blood sampling. The vein was seen after few removals of feathers from the site of collection and the needle at a slight angle was placed, bevel up against the vein on the underside of the wing. The needle was inserted into the vein and blood was slowly aspirated. Blood samples (3 mL) was collected in a labeled sterile universal bottle containing 1.0 mg/mL ethyl diamine tetra acetic acid for haematological analysis and another 2.5 mL was collected in anti-coagulant free bottle for serum biochemical analysis. Serum was separated by centrifugation at 3000 rpm at 4°C for 10 minutes and stored immediately at -20°C until use. Hematological parameters measured were packed cell volume (PCV), hemoglobin (Hb), red blood cells count (RBCs) and total white blood cells (WBCs) and differential leukocyte counts were assessed according to the routine hematological procedures for avian. The biochemical parameters measured were total protein (TP), albumin (Alb), total cholesterol (TC), calcium (Ca^{2a+}) and phosphorous (PO_4). All the parameters were assayed using spectrophotometer and commercial test kits of Randox following manufacturer's instruction.

Statistical analysis

Analysis was done using SAS (2008) statistical package. One-way analysis of variance (ANOVA) was used to analyse the data followed by Tukey's Multiple Comparison Test, to determine any significant differences between the treatments. *P* values <0.05 were considered significant.

Results and Discussion

The proximate composition of RBM is presented in Table 1. All the values obtained for dry matter, crude protein, crude fibre, ash, ether extract, and

nitrogen-free extracts fall within the range for nutrient content in cereals as reported by Olomu (2011). The chemical composition of experimental diets is presented in Table 2. The diets were formulated to meet the nutrient standard for broilers (N R C, 1994).

The result of the effect of Bambara nut on haematological parameters of finisher broiler chickens is presented in Table 3. The result revealed no significant differences ($p>0.05$) in most of the parameters measured. However, there were significant differences ($p<0.05$) in red cell distribution width (RDW), and monocytes (MON) levels across the treatments. The result indicated that RDW% levels decreased with increasing levels of roasted Bambara nut meal (RBM). The mean cell volume (MCV), mean cell haemoglobin (MCH) and mean cell haemoglobin concentration did not show any significant treatment effect. However, the MCHC were within the normal reference values 26.0 – 35.0% (Banerjee, 2013). The MCH were above the values (25 – 27 pg) reported by Swenson (2004). The values of the red blood indices MCV, MCH and MCHC were an indication of a strong immune system. The values of PCV obtained from this study (33.03 – 37.10) were higher than the values 32.66–36.33% and 23.32– 32.33% reported by Okonkwo and Esiegwu (2018) for broiler finishers and were within the normal range (35.9 to 41.00%) (Merck, 2012 and Wikivet, 2013). Demoranville and Best (2013) reported that a reduction in PCV below the normal value was a sign of liver and kidney disease, vitamin B12 and folic acid deficiencies. The values for the PCV showed sign of no toxicity. Adamu *et al*. (2006) observed that nutrition had significant effect on haematological values like PCV, Hb and RBC. The haemoglobin (Hb) did not show any significant treatment effect. Haemoglobin is an important determinant of anaemia (Wikivet, 2013). Low values of RBC and Hb could be a sign of anaemia (Mohammed and Oloyede, 2009). The values for Hb were within the normal reference ranges (7.0 – 13.0) (Banerjee, 2013). The values obtained for Hb (9.57 – 11.77g/dl) were higher than the values (7.10 – 10.10g/dl) reported by Aguihe *et al*. (2014) and were within the normal ranges (11.60–13.68g/dl) as reported by Wikivet (2013). The RBC was significantly decreased at 30% dietary level compared to the control and 10% dietary level and there was no significant difference among the treatments. The white blood cell and lymphocyte did not differ significantly in all the treatments. These indicated the safety of the experimental diets.

The biochemical indices were not ($p>0.05$) influenced by dietary treatments except for calcium (Table 4). The result of serum calcium revealed significant differences ($p<0.05$) across the treatments where T4 (30%) presented the highest value (2.43 mmol/L). Cholesterol is a sterol, a type of lipid molecule, and is biosynthesized by all animal cells as an essential structural component of all animal cell membranes (Hanukoglu, 1992). Total serum cholesterol decreased significantly as levels of RBM-based diets increased. The low serum cholesterol implies that Bambara nut-based diets did not give room for normal fat metabolism and utilization as a result of presence of anti-nutrients. Bambara groundnut may have a hypocholesterolemic action. The reduction in cholesterol suggests that Bambara groundnut meal perhaps possess some anti- cholesterol properties and its action. Bush (1991) and Okonkwo and Esiegwu (2018) reported that low level of cholesterol was an indication of fat malabsorption in the body and in the blood. The result also revealed that the Total protein did not show any significant difference. The total protein levels indicated that both the control and RBM-based diets fed to the birds were unable to efficiently make available the nutrients in the diets to the birds. However, inclusion of RBM in the diets of finisher broiler chickens tended to increase their serum total protein when compared to control suggesting the superiority of the protein quality in RBM. The non-significant effect of the serum albumin in this study may be attributed to the safety of the diets and perhaps good health status of the birds.

Table 1: Proximate composition of roasted Bambara nut meal

	Treatments (% RBM)			
Parameters (%)	**0**	**10**	**20**	**30**
Dry Matter	95.18	95.95	95.80	95.00
Crude Protein	13.13	20.23	22.97	27.89
Ether Extract	1.88	1.88	1.88	2.04
Crude Fiber	11.06	11.10	14.63	21.39
Ash	10.68	8.68	12.35	9.44
NFE	62.77	58.11	48.18	39.25

NFE: Nitrogen Free Extract determined by: NFE=100-(%CP+%CF+%EE+%Ash)

Table 2: Ingredients composition of experimental broiler finisher diets (5-8wks)

	Treatments (% RBM)			
Ingredients	**0**	**10**	**20**	**30**
Maize	56.91	50.59	44.83	38.03
Soybean meal	23.39	19.71	15.97	12.27
Wheat offal	10.00	10.00	9.50	10.00
Fishmeal	5.00	5.00	5.00	5.00
Bambara nut meal	0.00	10.00	20.00	30.00
Bone meal	2.50	2.50	2.50	2.50
Limestone	1.50	1.50	1.50	1.50
Salt	0.25	0.25	0.25	0.25
Premix*	0.25	0.25	0.25	0.25
Methionine	0.10	0.10	0.10	0.10
Lysine	0.10	0.10	0.10	0.10
Total	**100.00**	**100.00**	**100.00**	**100.00**
Calculated Analysis				
ME (kcal/kg)	2821.23	2874.51	2928.40	2981.70
Crude Protein (%)	20.00	20.00	20.00	20.00
Crude Fiber (%)	4.18	4.20	4.12	4.10
Ether Extract (%)	4.03	4.24	4.50	4.66
Calcium (%)	1.87	1.90	1.86	1.87
Phosphorus (%)	1.03	0.98	0.96	0.93
Lysine (%)	1.11	1.12	1.12	1.13
Methionine (%)	0.40	1.40	0.87	0.41

ME = Metabolizable Energy.
*Composition of Premix (Vitamin-mineral mixture): vitamin A: 2 400 000 IU; vitamin D: 1 000 000 IU; vitamin E: 16 000 IU; vitamin K: 800 mg; vitamin B1: 600 mg; vitamin B2: 1 600 mg; vitamin B6: 1 000 mg; vitamin B12: 6 mg; niacin: 8 000 mg; folic acid: 400 mg; pantothenic acid: 3 000 mg; biotin: 40 mg; antioxidant: 3000 mg; cobalt: 80 mg; copper: 2000 mg; iodine: 400; iron: 1 200 mg; manganese: 18 000 mg; selenium: 60 mg; zinc: 14 000 mg

Table 3: Effect of Bambara nut on haematological parameters of finisher broiler chickens

	Treatments				
Parameters	**T1 (0%)**	**T2 (10%)**	**T3 (20%)**	**T4 (30%)**	**SEM**
RDW (%)	7.40^{a}	6.87^{ab}	6.80^{ab}	6.50^{b}	0.163
MCHC (g/dL)	31.77	31.13	30.77	31.73	0.536
MCHP (g/L)	47.10	46.47	43.83	45.97	0.909
MCVF (Li)	150.77	152.07	148.23	147.10	4.046
HC (%)	37.10	29.60	33.03	33.10	2.104
Hb (g/dL)	11.77	9.77	9.57	10.50	0.676
RBC (10^6/HL)	1.46	1.39	1.45	1.22	0.159
MON (%)	12.47^{ab}	12.33^{ab}	10.13^{b}	15.30^{a}	0.823
LYM (%)	79.77	85.13	86.83	85.00	2.613
WBC (10^6/mL)	105.63	106.00	104.63	108.20	1.989

Means within rows with different superscripts are significantly different at $p \leq 0.05$, PCV = Packed Cell Volume, WBC = White Blood Cell, RBC = Red Blood Cell, HGB = Haemoglobin, MCH = Mean Corpuscular Haemoglobin, MCV = Mean Corpuscular Volume, LYM= Lymphocytes, MCHC = Mean Corpuscular Haemoglobin Concentration.

Table 4: Biochemical indices of broiler chickens fed RBM-based diets

	Treatments (%RBM)				
Parameters	T1	T2	T3	T4	SEM
T –cholesterol (mmol/L)	4.52	4.19	3.86	4.20	0.409
Total protein (g/L)	24.00	28.67	26.33	26.00	2.593
Albumin (g/L)	21.00	18.33	19.67	20.00	0.862
Calcium (mmol/L)	2.10^{ab}	1.27^{c}	1.77^{b}	2.43^{a}	0.049
Phosphorus (mmol/L)	1.90	1.77	1.80	1.93	0.113

Means within rows with different superscripts are significantly different at $p < 0.05$.

CONCLUSION

The results of the trial have shown that Bambara nut-based diet can serve as feed ingredient to broiler finisher birds up to 30% inclusion level. It is therefore concluded that broiler finisher chickens can be fed with Bambara nut meal up to 30% inclusion level in their diets as a partial replacement for soybean meal (which is more expensive in the study area) without any detrimental effect on blood indices.

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