BLOOD PROFILE OF BROILER CHICKENS FED VARYING DIETARY LEVELS OF PEPPERFRUIT (*Dennettia tripetala*) MEAL

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ABSTRACT

An experiment was conducted to assess the blood profile (haematology and cholesterol components) of broiler chickens fed varying dietary levels of Dried Pepperfruit (*Dennettia tripetala) M*eal (DPFM). One hundred and fifty day-old Hubbard broiler chicks were randomly assigned to five treatments containing thirty chicks each. Each treatment was replicated three times, which contained 10 birds each. The experiment was arranged in a completely randomized design. Five diets were formulated to represent the treatments (T1 – T5). Treatment (T1) was the control diet containing no DPFM, while T2, T3, T4 and T5 contained 0.25, 0.50, 0.75 and 1% respectively. The diets were fed to the birds starting from day old for 28 days (starter phase) and 21 days for finisher phase. The birds were allowed access to feed and water ad libitum. At the end of 49 days, results indicated an increase in WBC, RBC and Hb by all the levels of pepper fruit meal. At 0.25% inclusion the neutrophils was reduced. The HDL (good cholesterol), VLDL were increased and LDL (bad cholesterol) was reduced by all levels of DPFM fed (0.25-- 1.0%) while the total cholesterol level was not affected. Inclusion of dried pepper fruit meal up to 1.0% in broiler chicken diets improved their haematology, immunity and good cholesterol levels while bad cholesterol levels was significantly reduced.

Keywords: Broiler chickens, Haematology, Cholesterol components, pepper fruit meal.
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INTRODUCTION

The growth in the poultry industry is having a profound effect on the demand for feed and raw materials for feed production. This is coupled with the fact that the major feed ingredients like maize and soya bean are in high demand for human food. Another pressure which is more profound is the consumer pressure on demand for poultry products free from antibiotic residues (Demir *et al*., 2003). Hence alternative substances and strategies for animal growth performance and disease prevention are being investigated. In this regard, phytogenics have received increased attention and inclusion since they have acquired more acceptability among consumers as natural feed additives (Majid *et al*., 2010). Recently, to show the importance of phytogenics in monogastric animal nutrition, Ndelekwute *et al*. (2017) advocated a comprehensive study of these plant materials which they termed Phytogenicology. The effect of dried pepper fruit meal or powder on haematology and blood cholesterol components of broiler chickens was assessed in this study.

MATERIALS AND METHODS

Experimental Site: The study was carried out at the Teaching and Research Farm of

the Department of Animal Science, University of Uyo, Uyo, Nigeria. Uyo lies between latitude $4^0$31'E and $45^0$31'N and $4^0$45'N and longitude $7^0$31'E and $45^0$351'E. The altitude of the area is 38 m above sea level and a mean rainfall of 2000 mm. The estimated relative humidity during the experiment was 79% and average temperature of 28^0C (Meteorology Station, University of Uyo, Uyo, Nigeria).

Source of Pepper fruit, Processing and Proximate Analysis: The pepper fruits were purchased from a market in Uyo metropolis. The pepper fruits were washed to be free from debris and dried in an oven at a temperature of 60oC to a final residual moisture content of 11%. The dried sample was milled and sieved and thereafter it was stored at room temperature in a plastic container. Proximate analyses of the pepperfruit powder and formulated diets were carried out. The crude protein, crude fibre, ether extract, total ash and nitrogen free extract were determined according to the methods of AOAC (2000).

Experimental Design

150 day-old chicks of Hubbard strain were used. The birds were allotted to five dietary treatment groups (T1, T2, T3, T4 and T5) containing 0.0, 0.25, 0.50, 0.75 and 1.0% respectively of Dried Pepper Fruit Meal (DPFM) or powder in a completely randomized design (CRD). Each treatment group contained 30 birds. Each dietary group was replicated three times and each replicate contained 10 birds.

Experimental Diet

Five experimental diets were formulated at the starter and finisher phases of the experiment. The diets were formulated to provide 23 and 20% crude protein for starter and finisher diets respectively. The metabolizable energy was 2879 kcal/kg (starter diet) and 2926 kcal/kg (finisher diet).Diet 1 was the control which contained no pepper fruit powder. Diets 2, 3, 4 and 5 contained 0.25, 0.50, 0.75 and 1.0% DPFM respectively. The same levels of DPFM was fed at both starter and finisher phases. Starter diet was fed for four weeks and finisher diet for 3 weeks making a total of seven weeks. As shown in Table 1 and 2.

Management of Experimental Birds

The birds were managed as reported by Simeon and Ndelekwute (2020). On arrival to the farm the, day old chicks were weighed as they were allotted to the five dietary treatment groups. Glucose was added to their drinking water. Feed and water were provided *ad libitum* throughout the experiment. For the first three weeks extra heat was provided by using kerosene stoves to keep them warm. The birds were housed in an open sided deep litter floor building. Adequate spacing, ventilation and protection against predators and adverse environmental conditions were ensured. The birds were vaccinated against Newcastle and Infectious Bursal diseases which was carried out by a Veterinary doctor. Other routine management of broiler chicks were also carried out according to Oluyemi and Roberts (2000).

Blood Profile and Serum Biochemical Indices Evaluation

At the end of the experiment, before blood sample were collected, the birds were fasted overnight. Blood samples were collected from 3 birds per treatment that is one per replicate. This was done by puncturing the bronchial vein with syringe and the blood collected into a set of ethylene-diamine-tetra-acetic acid (EDTA) bottles for the determination of haematological parameters (WBC, Lymphocytes, Monocytes, Neutrophils, Eosinophil, Basophils, RBC, HB, PCB, MCH, MCHC, Platelets). Blood samples for determination of lipid profile (Triglycerides, Cholesterol, HDL, LDL and VLDL) did not contain EDTA. The samples were taken to the University of Uyo Teaching Hospital for analysis using full blood count machine/mind ray Auto-Haematology Analyzer.

Table 1 Ingredient and Nutrients Composition of experimental starter broiler diet (%)

Treatments	**T1**	**T2**	**T3**	**T4**	**T5**
Levels of DPFM (%)	**0.00**	**0.25**	**0.50**	**0.75**	**1.00**
Ingredients:					
Maize	52.0	52.0	52.0	52.0	52.0
Soya bean meal	30.0	30.0	30.0	30.0	30.0
Palm kernel cake	10.0	9.75	9.50	9.25	9.0
Fish meal	4.0	4.0	4.0	4.0	4.0
Bone meal	3.0	3.0	3.0	3.0	3.0
Pepper fruit meal	0.00	0.25	0.50	0.75	1.0
Salt	0.25	0.25	0.25	0.25	0.25
Lysine	0.20	0.20	0.20	0.20	0.20
Methionine	0.20	0.20	0.20	0.20	0.20
Premix*	0.35	0.35	0.35	0.35	0.35
Total	**100**	**100**	**100**	**100**	**100**
Determined Nutrient Composition (%):					
Crude protein	22.88	22.88	22.87	22.88	22.87
Ether extract	7.78	7.77	7.78	7.77	7.77
Crude fibre	4.87	4.85	4.82	4.79	4.77
Ash	5.63	5.62	5.63	5.63	5.63
Lysine**	1.22	1.22	1.22	1.22	1.20
Methionine**	0.48	0.48	0.48	0.48	0.47
Calcium**	1.21	1.21	1.21	1.21	1.21
Phosphorus**	0.95	0.95	0.95	0.94	0.94
Energy KcalME/kg**	2879	2879	2888	2879	2879

*1kg of premix contains: Vitamin A (10,000,000iu), vitamin E(16,000mg),vitamin k3 (800mg), vitamins B_{12} (22,000mg), niacin (22,000mg), vitamin B_2 (10mg), folic acid (400mg), biotin (32mg), chlorine chloride (200,000mg) zinc (32,000mg) iodine (600mg), cobalt (120mg), selenium (40mg), antioxidant (48,000mg). **Calculated. DPFM = Dried pepper fruit meal.

Statistical Analysis

The data obtained were subjected to one-way analysis of variance (ANOVA) using SPSS software (IBMSPSS Statistics version 20). Treatment means were separated by Duncan Multiple Range Test of the software.

RESULT AND DISCUSSION

The result of the proximate composition indicated that the Peperfruit meal contained crude protein, 15.76; crude fibre, 14.80%; ether extract, 4.95% and ash, 4.64%. This confirmed earlier report that pepperfruit contains protein, fat and high in fibre (Borokin and Ogunyemi, 2013). In a related report, Simeon and Ndelekwute (2020) maintained that pepper fruit could be potentially useful as feed additive for poultry production because of its nutritional content.

Effect of experimental diets on haematology and cholesterol components of broiler chickens is shown in Table 3. Apart from eosinophil, mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) were not significantly influenced ($P>0.05$) by pepperfruit. All other indices were influenced ($P<0.05$). The values of white blood cells (WBC), red blood cells (RBC), haemoglobin (Hb), packed cell volume (PCV) and platelets were significant in all the groups of birds that consumed diets containing different levels of DPFM. The values of monocytes and basophil components of the white blood cells were also significant at all the levels of inclusion of DPFM. However, above 0.25%

inclusion, the neutrophils was reduced. Increase in the WBC, RBC and Hb is an indication that pepperfruit could support the immunity and blood building of chickens.

Table 2: Ingredients and Nutrients Composition of experimental finisher broiler diet (%)

Treatments	**T1**	**T2**	**T3**	**T4**	**T5**
Levels of DPFM (%)	**0.00**	**0.25**	**0.50**	**0.75**	**1.00**
Ingredients:					
Maize	52.00	52.00	52.00	52.00	52.00
Soya bean meal	28.0	28.0	28.0	28.0	28.0
Palm kernel cake	14.30	14.05	13. 80	13.55	13.30
Fish meal	2.0	2.0	2.0	2.0	2.0
Bone meal	3.0	3.0	3.0	3.0	3.0
Pepper fruit meal	0.00	0.25	0.50	0.75	1.0
Salt	0.25	0.25	0.25	0.25	0.25
Lysine	0.10	0.10	0.10	0.10	0.10
Methionine	0.10	0.10	0.10	0.10	0.10
Premix*	0.25	0.25	0.25	0.25	0.25
Total	**100**	**100**	**100**	**100**	**100**
Determined composition (%):					
Crude protein	20.0	20.0	20.0	20.0	19.95
Crude Fibre	5.64	5.62	5.59	5.57	5.54
Ether Extract	4.70	4.69	4.70	4. 69	4.69
Ash	5.23	5.27	5.27	5.27	5.27
Lysine**	0.95	0.95	0.94	0.93	0.93
Methionine**	0.38	0.38	0.38	0.38	0.38
Calcium**	1.06	1.06	1.06	1.06	1.06
Phosphorus**	0.71	0.71	0.71	0.71	0.71
Energy KcalME/kg**	2926	2926	2926	2926	2926

*1kg premix contained: vitamin A (10,000,000iu), vitamin D3 (1,000,000iu), vitamin E (16,000mg), vitamin k3 (800mg), vitamins B_2 (22,00mg), niacin (22,00mg), vitamin B_{12} (10mg), Folic Acid (400mg) Biotin (32mg), Chlorine chloride (200,00mg), Zinc (32,000mg) iodine (600mg), cobalt (12mg), selenium (40mg), antioxidant (48,000mg). **Calculated; DPFM = Dried pepper fruit meal.

White blood cells are phagocytic in nature. They destroy any invading pathogenic micro-organisms in the body. Adequate and effective circulation of oxygen in the body requires haemoglobin. A study with pepperfruit indicated an increase in WBC, RBC and Hb in rats (Olusola, *et al.* 2015). Contrary to this study, extract of pepper fruit indicated a decrease in WBC in mice (Anaga, *et al.* 2008). Some works using other spices supported this result. Al-kassie (2009) reported that diet supplemented with extract derived from thyme and cinnamon fed to broilers, increased WBC values compared to control diet. On the contrary Majid *et al.* (2010) reported no significant impact of thyme and garlic on WBC of broilers.

However, increase in HDL and VLDL, reduction in LDL and the no significant effect on the total cholesterol is an indication that inclusion of DPFM in broiler chickens diets up to 1.0% level promotes good cholesterol (HDL), decreases LDL (bad cholesterol) and could be a good material for healthy living. High level of cholesterol especially the LDL causes hypertension which leads to arteriosclerosis and heart attack (Kennelly *et al*., 2022). Other reports indicated that high level of serum cholesterol in the blood can damage arteries and are potentially linked to diseases such as those associated

with cardiovascular system (Pearson, *et al*. 2005; Olson, 1998; Haines, 2001; and Pawlina, *et al.,* 2006).

Table 3: Effect of Pepperfruit(%) on haematology and cholesterol components broiler Chickens

Treatments	**T1**	**T2**	**T3**	**T4**	**T5**	**SEM**
Levels of DPFM (%)	**0.00**	**0.25**	**0.50**	**0.75**	**1.00**	
Blood parameters:						
White blood cells ($x10^{10}/1$)	90.30^{b}	112.50^{a}	123.40^{a}	123.40^{a}	123.80^{a}	18.16
Lymphocytes (%)	89.90	86.10	90.40	85.70	87.30	6.18
Monocytes (%)	0.10^{c}	0.30^{b}	0.30^{b}	0.30^{b}	0.50^{a}	0.01
Neutrophils (%)	13.90^{a}	13.20^{a}	10.50^{b}	10.90^{b}	10.70^{b}	3.17
Eosinophil (%)	0.90	1.0	1.0	1.0	1.0	0.20
Basophils(%)	0.20^{c}	0.40^{b}	0.40^{b}	0.40^{b}	0.50^{a}	0.08
Red blood cells ($x10^{10}/1$)	0.64^{c}	1.43^{b}	1.56^{b}	1.45^{b}	2.40^{a}	0.75
Haemoglobin (g/dl)	2.60^{b}	9.50^{a}	9.60^{a}	9.10^{a}	9.30^{a}	3.50
Packed cell volume(%)	8.60^{d}	30.80^{a}	27.30^{a}	28.40^{a}	30.60^{a}	6.80
MCV (fl)	134.40^{a}	126.70^{b}	126.60^{b}	127.60^{b}	127.50^{b}	6.64
MCH (pg/l)	40.60	39.10	42.30	39.0	38.80	3.86
MCHC (g/l)	30.20	30.80	31.0	30.60	30.40	3.77
Platelets ($x10^{3}/\mu l$)	43^{c}	55^{a}	54^{a}	54^{a}	48^{b}	4.12
Triglycerides (mg/100ml)	0.90^{b}	1.09^{b}	1.02^{b}	1.40^{a}	1.21^{a}	0.45
Total Cholesterol (mg/100ml)	2.60	3.0	2.90	2.80	2.80	0.30
HDL (mg/100ml)	1.70^{b}	2.10^{a}	2.00^{a}	2.05^{a}	2.0^{a}	0.35
LDL (mg/100ml)	0.56^{a}	0.43^{b}	0.40^{b}	0.46^{b}	0.30^{c}	0.08
VLDL (mg/100ml)	0.41^{b}	0.50^{a}	0.56^{a}	0.54^{a}	0.55^{a}	0.07

$^{a-d}$Means along the same row with different superscripts are significantly difference ($P<0.05$). SEM = Standard error of means; DPFM = Dried pepper fruit meal; HDL = High density lipoprotein; LDL = Low density lipoprotein; VLDL = Very low density lipoprotein.

CONCLUSION

- Dietary pepper fruit meal up to 1.0% level of inclusion had no adverse effect on the haematological indices and cholesterol components of broiler chickens, but rather improved the red blood cells, white blood cells, immunity, blood building and good cholesterol levels.

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