HAEMATOLOGY AND SERUM BIOCHEMICAL INDICES OF PULLETS FED ENZYME SUPPLEMENTED BOVINE BLOOD-RUMEN CONTENT MIXED WITH DISCARDED VEGETABLE (*AMARANTHUS* SPP)

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ABSTRACT

Increasing cost of feed and feedstuff in poultry industry request for least-cost diet formulation without compromising the nutritional quality of the diet and health status of birds. A 56-day feeding trial was conducted to investigate the haematology and serum biochemistry of pullet chicks fed enzyme (Rovabio) supplemented Bovine blood-rumen content and discarded vegetable (Amaranthus spp) meal (BBRVM). A total of 720-day-old dominant black strain pullet chicks were assigned to ten (10) dietary treatment of three replicates. BBRVM was used with or without enzyme (20g/100kg diet) to replace soyabean meal at 0, 25, 50, 75 and 100%. Data obtained were arranged in a 2 x 5 experimental layout in a completely randomised design. Significant means were separated (P<0.05) using Duncan's Multiple Range Test as contained in SAS. White blood cell significantly (P<0.05) increased from 28.25cumm^{2}x10^3 (0% BBRVM) to 32.27cumm^{2}x10^3 (100% BBRVM). Lymphocytes decreased (P<0.05) from 79.33% (0% BBRVM) to 74.33% (100% BBRVM). Highest (P<0.05) monocytes (0.13%) and basophil (0.69 %) were obtained for birds fed 100% BBRVM. Serum total protein varied significantly (P<0.05) from 38.70g/l (25% BBRVM) to 47.15g/l (75% BBRVM). Birds fed diet containing enzyme supplemented BBRVM recorded the highest (P<0.05) total protein (43.22g/l) while those fed diet without enzyme supplementation had lowest value (39.54 g/l). This study reveals BBRVM based diets fed pullet chicks had no deleterious effect on their health status.

Key words: ***Biochemistry, bovine, enzyme, haematology, health-status***

INTRODUCTION

The increasing cost of feed and feedstuff has been a threat to the survival of the livestock industry (Sonaiya, 1990) in developing nations, with this situation particularly serious for the monogastric animals (Balogun *et al*., 2000). Moreover, the competition for feedstuff between the increased human population, confectionery industries and livestock industries for ingredients has contributed to the shortage of feedstuff for livestock especially monogastric animals (Adeniji and Oyeleke, 2008). Recycling abattoir wastes in animal nutrition can provide unconventional dietary ingredients as well as reducing disposal and environmental pollution problems (Adeniji and Balogun, 2000). The bovine blood-rumen content meal has been shown to be rich in protein (31% CP) and a good source of B-carotene, but contain crude fibre as high as 14.48% (Adeniji and Oyeleke 2008). The economic and nutritional advantage of the amaranth as a leafy vegetable is accentuated by its agronomic superiority over many plant protein sources (Zraly *et al.,* 2004). Advances in animal nutrition have indicated that exogenous enzyme supplementation can render fibrous polysaccharides utilizable by various classes of monogastric animals (Cowieson *et al.,* 2003). Haematological studies are useful in the diagnosis of many diseases as well as investigation of the extent of damage to the blood. According to Onifade and Babatunde (1993), blood examination is a good way of assessing the health state of an animal and this play an important role in the physiological, nutritional and pathological structure of an organism Therefore, the need to investigate the effect of enzyme supplemented Bovine blood-rumen content-vegetable meal on the blood indices in other establish the replacement level without any deleterious effect on the health of pullet chicks is inevitable.

MATERIALS AND METHODS

Experimental site

The study was carried out at Alao Farms, Tanke-Akata, Ilorin, Kwara State, Nigeria. The site (Ilorin) is on longitude 4° 35'N and latitude 8° 30'E (Google earth, 2023). Mean monthly temperature varies from 25°C to about 29°C. The relative humidity varies from about 70% in the dry months to about 80% in the wet months.

Experimental birds and management

A total of seven hundred and twenty (720) day-old (Dominant Black) pullet chicks were purchased from a reputable commercial hatchery. They were divided into ten (10) treatment groups of Seventy-two (72) birds each. Each group was further divided into three (3) subgroups comprising of 24 birds each serving as replicates. The house and equipment were thoroughly washed and disinfected before the arrival of the chicks. All recommended vaccinations and medications were administered while the birds were intensively managed for period of eight (8) weeks.

Preparation of bovine blood rumen content vegetable meal (BBRVM)

The blood, rumen content and chopped discarded amaranthus were weighed (weight per weight) and mixed in that other using ratio 2:1:1. The mixtures were poured into a drum and boiled with continuous stirring to prevent it from burning for duration of 45 minues until the mixture was semi-dried. The boiled materials were then sun-dried for about 3 days. The resulting Bovine blood rumen content vegetable meal (BBRVM) were bagged and stored before feeding trial.

Experimental diets and design

Diets were formulated (Table 1) using BBRVM as replacement for soyabean meal at 0%, 25%, 50%, 75% and 100%. Enzyme (ROVABIO®) was used to supplement at each level of BBRVM at 0g (without enzyme) and 20g (with enzyme) per 100kg of diet to render fibrous polysaccharides utilizable. A total of seven hundred and twenty (720) day-old pullet chicks were randomly allotted to ten (10) dietary treatment of seventy-two (72) birds each. Each treatment group was sub-divided into three (3) replicates of 24 birds each. They were allotted to the dietary treatment for the period of eight (8) weeks in a 2 x 5 factorial arrangement.

Table 1: Gross composition of experimental chick diets containing bovine blood-rumen content-vegetable mixture

	Unsupplemented diets					Enzyme supplemented diets				
	1	2	3	4	5	6	7	8	9	10
Ingredients (%)	0%	25%	50%	75%	100%	0%	25%	50%	75%	100%
Maize	45.00	45.00	45.00	45.00	45.00	45.00	45.00	45.00	45.00	45.00
Soyabean meal	15.00	11.25	7.50	3.75	-	15.00	11.25	7.50	3.75	-
BBRVM	-	3.75	7.50	11.25	15.00	-	-	-	-	-
BBRVM +	-	-	-	-	-	-+	3.75+	7.50+	11.25+	15.00+
Groundnut cake	15.00	15.00	15.00	15.00	15.00	15.00	15.00	15.00	15.00	15.00
Wheat bran	18.50	18.50	18.50	18.50	18.50	18.50	18.50	18.50	18.50	18.50
Fish meal (72%)	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00
Bone meal	2.50	2.50	2.50	2.50	2.50	2.50	2.50	2.50	2.50	2.50
Oyster shell	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50
Premix***	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Table salt	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Total	**100.00**	**100.00**	**100.00**	**100.00**	**100.00**	**100.00**	**100.00**	**100.00**	**100.00**	**100.00**
Determined Analysis										
Crude Protein (%)	20.17	20.22	20.26	20.31	20.35	20.17	20.22	20.26	20.31	20.35
Crude Fibre (%)	3.89	4.85	4.90	5.82	6.79	3.89	4.85	4.90	5.82	6.79
Fat (%)	4.09	3.97	3.85	3.79	3.62	4.09	3.97	3.85	3.79	3.62
Calcium (%)	3.06	3.05	3.04	3.04	3.03	3.06	3.05	3.04	3.04	3.03
Phosphorus (%)	0.80	0.78	0.75	0.72	0.70	0.80	0.78	0.75	0.72	0.70
*ME(Kcal/kg)	2751.90	2756.40	2760.90	2765.40	2769.90	2751.90	2756.40	2760.90	2765.40	2769.90

*** vitamin A – 12,000,000i.u; vitamin D_3 – 2,750,000i.u; vitamin E – 20,000mg; vitamin K – 2,000mg; Thiamine B_1 – 1,500mg; Riboflavin B_2 – 4,000mg; Niacin – 18,000mg; Pantothenic Acid – 7,000mg; Vitamin B_6 – 2,000mcg; Vitamin B_{12} – 12mg; Folic Acid – 1,000mcg; Biotin – 15i.u; Choline Chloride – 150,000mg; Cobalt – 500mg; Copper – 6,000mg; Iodide – 1,100mg; Iron – 20,000mg; Manganese – 80,000mg; Selenium – 200mg; Zinc – 50,000mg; Antioxidant – 125,000mg.

BBRVM = Bovine blood rumen content vegetable meal

BBRVM += Enzyme supplemented bovine blood rumen content vegetable meal

*ME(Kcal/kg) = Estimated Metabolizable Energy

Blood sample collection and analysis

Blood samples were obtained via the wing vein puncture using needle and syringe. The blood samples were emptied into 2 sets of well labelled bottles; one contained ethylene diamine tetra-acetate (EDTA) as anti-coagulant while the other one contained no anti-coagulant. The samples were analysed in the laboratory for haematological parameters and biochemical.

Data analyses

The data generated from the study were subjected to Analysis of Variance (ANOVA) in a 2x5 factorial arrangement in a completely randomised design. Significant ($P<0.05$) differences among treatment means were separated using Duncan's Multiple Range Test as contained in SAS (2011). Model sums of square were partitioned to test linear, quadratic and cubic trends (Gomez and Gomez, 1983).

RESULTS AND DISCUSSION

The value for WBC significantly ($P<0.05$) increased from 28.25 in the diet containing 0% BBRVM to 32.27($cumm^2x10^3$) in the diet containing 100% BBRVM. Lymphocytes percentage of the chicks significantly ($P<0.05$) decreased with the increased level of BBRVM inclusion. Diet containing 0% BBRVM had the highest value (79.33%) compared to lowest value (74.33%) recorded for birds fed 100% BBRVM. A noticeable statistical ($P<0.05$) variation was observed in the highest percentage (0.13) among 100% BBRVM group for monocytes. Moreover, basophils also elicited significant ($P<0.05$) variation across the treatment groups. Only blood serum total protein was significantly ($P<0.05$) influenced by inclusion levels of BBRVM in the diet, with values ranging from 38.70 g/L for 25% BBRVM group to 47.15 g/L in 75% BBRVM group. Similarly, sole effect of enzyme supplementation significantly ($P<0.05$) affected blood total protein. An elevated value of WBC at 100% BBRVM inclusion is an indication that the birds might be reacting to increased exposure to pathogenic organisms in the feed (Oduguwa, 2006). The birds probably adjusted by producing more antibodies since WBC confer immunity required (Maxwell *et al*., 1990). The serum total protein value reported for the pullet chicks increased as the level of BBRVM increased. This could be as a result of high protein digestibility at this level of BBRVM inclusion. Ahamefule *et al*. (2006) opined that high serum protein is an indication of protein adequacy in the diet of poultry. Hence, enzyme supplementation improved serum protein status of birds.

Table 2: Main effect of levels of BBRVM inclusion and enzyme supplementation on the haematology and serum biochemical indices of pullet chicks (0-8 week)

	Levels of BBRVM inclusion						Enzyme supplementation		
Parameters	**0%**	**25%**	**50%**	**75%**	**100%**	**S.E.M**	**--**	**++**	**S.E.M**
Blood Haematology									
Packed Cell Volume (%)	23.83	23.33	24.33	22.83	20.33	0.99	22.83	23.03	1.03
Haemoglobin (g/dL)	8.48	9.43	9.78	8.83	9.83	0.37	8.27	9.47	0.45
Red Blood Cell($X10^{12}$/L)	2.84	2.10	2.24	2.77	2.44	0.84	2.81	2.77	0.07
White Blood Cell($cumm^2x10^3$)	28.25^{e}	28.92^{d}	29.60^{c}	30.55^{b}	32.27^{a}	0.30	28.27	28.17	0.23
Neutrophils (%)	29.83	28.33	30.31	30.83	31.33	0.24	29.43	30.03	1.73
Lymphocytes (%)	79.33^{a}	78.38^{b}	78.33^{b}	76.33^{c}	74.33^{d}	0.85	76.83	75.23	1.11
Eosinophils (%)	1.57	1.13	1.07	1.13	1.04	0.43	1.08	1.16	0.19
Monocytes (%)	0.00^{b}	0.00^{b}	0.00^{b}	0.00^{b}	0.13^{a}	0.04	0.00	0.00	0.00
Basophils (%)	0.00^{c}	0.13^{b}	0.00^{c}	0.00^{c}	0.69^{a}	0.11	0.00	0.00	0.00
Serum Biochemistry									
Total protein (g/L)	41.85^{c}	38.70^{d}	44.35^{b}	47.15^{a}	44.85^{b}	0.75	39.54^{b}	43.22^{a}	0.91
Albumin (g/L)	22.45	21.55	23.75	24.80	23.95	0.38	23.78	22.82	0.49
Globulin (g/L)	19.40	18.15	20.60	20.05	21.30	0.52	20.44	19.72	0.71
Uric acid (g/L)	3.65	3.35	3.20	3.25	3.75	0.12	3.92	3.12	0.14
Creatinine (g/L)	1.05	1.25	1.15	1.15	1.10	0.38	1.12	1.16	0.06
Glucose (g/L)	156.55	165.95	169.75	168.70	154.40	2.81	156.20	164.34	2.42

[abcde]: Means in the same row with different superscripts differ significantly ($P<0.05$)

[a] Probability for Linear (L), Quadratic (Q) and Cubic (C) trends

BBRVM = Bovine blood rumen content vegetable meal

-- without enzyme supplementation

++ with enzyme supplementation

CONCLUSION

Haematological and biochemical indices reported in this study across the treatment groups fall within the range for normal avian species influenced by the dietary treatments as BBRVM based diets fed to the pullets in this study had no deleterious effect on their health status.

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