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Effect of Replacing Bone Ash with Eggshell Meal on Nutrient Digestibility and Blood Parameters of Broiler Chickens

Research Article

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ABSTRACT

One hundred and eighty 28-days-old Arbor Acres broiler birds were utilized in a 28 day experiment to investigate the effect of replacing bone ash with eggshell meal in the diet of broiler birds on their nutrient digestibility and blood parameters. Five experimental diets tagged T_1, T_2, T_3, T_4 and T_5 were formulated to be isocaloric (2865.53 kcal ME/kg) and isonitrogenous (20% crude protein) such that eggshell meal replaced bone ash at 0, 25, 50, 75 and 100% for treatments T_1, T_2, T_3, T_4 and T_5 respectively. The birds were randomly assigned to the five dietary treatments and replicated twice giving a total of eighteen birds per replicate in a completely randomized design. The study showed that the haematological parameters were not affected ($P>0.05$) by the treatments except for Corpuscular Haemoglobin which were best ($P<0.05$) at T_5 (47.95 pg) and T_3 (45.60 pg) and followed by those fed T_1 (42.85 pg), T_2 (42.20 pg) and T_4 (43.40 pg). Similarly, values obtained for Mean Corpuscular Volume were improved ($P<0.05$) in birds fed T_5 (148.45 fl) followed by those fed T_3 (142.95 fl) and T_4 (140.45 fl). Serum urea, glucose, triglyceride and total protein were not ($P>0.05$) influenced by the treatments. Ten 28-days old birds with similar live body weight were paired and randomly assigned to five dietary treatments tagged T_1, T_2, T_3, T_4 and T_5 which were compounded to contain the same composition as those used in the blood studies. The birds were housed in cages and fed such that each group passed through the five treatments by rotation in a Latin Square Design arrangement. Feed and faecal samples were taken for proximate analysis. The results of the proximate analysis of the oven-dried faeces were used to compute the coefficient of digestibility of the nutrients. The results obtained showed that digestibility of dry matter, crude protein, crude fibre, ether extract, ash and nitrogen-free extract were not affected ($P>0.05$) by the treatments. It is therefore, recommended that since there was no deleterious and adverse effect of including eggshell meal in the diets on the blood parameters and digestibility of the nutrients, farmers can use eggshell meal as a major source of dietary calcium in broiler' diets.

KEY WORDS blood parameters, bone ash, broiler, eggshell meal, nutrient digestibility.

INTRODUCTION

Studies on the evaluation of alternative sources of feed have been more concerned with energy and protein stuffs. Not much has been made on local alternative sources for the major mineral nutrients like calcium and phosphorus (Tumora *et al.* 2004). The main sources of calcium are oyster shells, bone meal, limestone and di-calcium phosphate (Iheukweumere and Emenalon, 2004). Calcium and phosphorus which belong to the group of macro-minerals are the most important elements required by animals as the major components in the bones and teeth development. They also play important role in the muscle growth. It is essential in the formation of shells in eggs and also helps to regulate the

passage of nutrients in and out of the cells (Church and Pond, 1989). Dicalcium phosphate have been found to be major sources of calcium for livestock all over the world but are extremely costly following importation. Organic sources of calcium and phosphorus which are inexpensive and readily available except for the over dependence by the livestock industry include oyster shells and bone meal.

McDonald *et al*. (1992) noted that calcium and phosphorus represent the third most expensive nutrient after energy and protein. Apart from the rinderpest that wiped out thousands of cattle in 1982 - 1983, drought and religious barriers have affected the growth of the ruminant industry and pig production in the northern part of the country respectively.

This has greatly reduced the number of animals produced and consequently the supply of bones for bone meal. Furthermore, the diversion of bones into other uses likes the manufacture of glass, gelatins and fertilizers (Adewoye, 1982) have compounded the problem of organic sources of calcium and phosphorus in livestock industry especially bone ash.

According to Aduku and Olukosi (2000), Nigeria produces about 55.9 million eggs annually; this implies that over 250 metric tones of the shells are produced as by-product. This can be utilized as mineral supplement since eggshell is highly rich in calcium (90%) with little percentage (less than 5%) of phosphorus; it is possible that optimum dietary level of calcium and phosphorus can be met using eggshell meal as a source. The aim of this research is therefore, to evaluate the effect of replacing bone ash with eggshell meal on the blood parameters and nutrient digestibility of broilers finisher birds.

MATERIALS AND METHODS

Study area

The experiment was conducted at the Research and Teaching Farm of the Faculty of Agriculture, Nasarawa State University, Keffi, Shabu, Lafia Campus.

Test ingredient and experimental feeds

The test ingredient (eggshell) was sourced locally within Lafia metropolis sun-dried and crushed into powder to form the eggshell meal and was used to compound the finisher experimental diets.

Five isonitrogenous (20% crude protein) and isocaloric (2865 kcal/kg ME) diets tagged T_1, T_2, T_3, T_4 and T_5 were compounded such that eggshell meal replaced bone ash at 0, 25, 50, 75 and 100% for T_1, T_2, T_3, T_4 and T_5 respectively. All other micro-nutrients were included at the recommended levels to meet the requirement of the birds as prescribed by Oluyemi and Robert (2000).

The percent and calculated chemical composition of the experimental diets for the finisher birds is presented in Table 2.

Experimental design and statistical analysis

One hundred and eighty 28 days old Arbor Acre growing broilers were allotted into five treatment groups in a Completely Randomized Design and replicated twice. Each replicate had 18 birds giving a total of 36 birds per treatment group. The experiment lasted for 28 days. Data obtained were subjected to One Way Analysis of Variance (ANOVA). The separation of means was effected using least significant difference method and tested at probability level of 5% as described by Steel and Torrie (1980). The following statistical model was used:

$$Y_{ij}=U+T_1+€_{ij}$$

Where:
Y_{ij}: individual observation.
U: population mean.
T_1: treatment error.
$€_{ij}$: random error.

Blood collection

At the end of the feeding trial two birds per treatment of similar live body weight were selected for the evaluation of the haematological indices and serum biochemical variables. The birds used were fasted for 12 h over night and bled in the morning between 0700-0800 hours. Fasting was done to avoid temporary evaluation of many metabolites by feeding (Bush, 1975). Blood samples were collected from vein of the birds under the wing using 5ml sterile disposable needles. The collection sites were sterilized with 70% alcohol and xylene to dilate the veins. Blood samples for haematology were separately collected into sample bottles containing 1mg of dipotassium salts of ethylene diamine tetraacetic acid (EDTA-K^2) to 1ml of blood. The haematological indices determined were Packed Cell Volume (PCV), Red Blood Cell count (RBC), White Blood Cell count (WBC) and Haemoglobin Concentration. PCV was determined by using wintrobes micro haematocrit method (Margi, 1997), haemoglobin concentration was determined by the Cyanomethaemoglobin method according to Kelly (1979). The improved Neubar haemocytometer method described by Jain (1986) was used to estimate the red and white blood cells. Mean Corpuscular Volume (MCV), Mean Corpuscular Haemoglobin (MCH) and Mean Corpuscular Haemoglobin Concentration (MCHC) were computed according to Jain (1986). Blood samples were collected in another separate sample bottles without anticoagulant for clotting for serum biochemical analysis. Se-

rum protein, globulin, urea and triglyceride were analyzed using sigma kits while glucose was analyzed according to Schalm *et al.* (1975).

Digestibility trial

Ten birds (28 days old) were paired and randomly assigned to the five dietary treatments. The birds were housed individually in cages measuring 20 cm x 10 cm x 20 cm and fed with a known quantity of feed *ad libitum* to determine the intake throughout the period of the experiment which lasted 5 weeks. Each group of birds were randomly allocated to a treatment and rotated weekly such that each group passed through all the five treatments. A 4-day faecal collection period and a 3-day resting period was adopted. The faeces collection was done using a clean wooden pad placed under the cages. The faeces collected were oven-dried immediately, weighed and pooled per week and per treatment. Samples of the feeds and faeces were taken for proximate analysis according to the methods outlined by AOAC (1990) and the results used to compute the coefficient of digestibility of nutrients.

RESULTS AND DISCUSSION

The mineral analysis of the test ingredient is presented in Table 1.

Table 1 Mineral analysis of the eggshell

Mineral	Percentage
$CaCO_3$	94
Organic matter	4
$MgCO_3$	1
$Ca_3(PO_4)_2$	1

Source: Aduku and Olukosi (2000).

Eggshell contained over 94% calcium in the form of $CaCO_3$ and $Ca_3(PO_4)_2$. Other minerals (MgCO3) and organic matter was about 5%. The energy composition of the experimental diets for broiler finisher birds is presented in Table 3. The moisture content of the diets was within the range of 91.20-93.20%; the crude protein and ME values were isonitrogenous (20% CP) and is ocaloric (2865.53 kcal/kg, ME) respectively. The values for ether extract, crude fibre, ash, NFE, calcium and phosphorus were within the normal ranges as recommended by NRC (1979). The result obtained on the effect of replacing bone ash with eggshell on the haematological parameters and serum biochemical variables of finishing broiler birds are presented in Table 3. The results obtained showed that packed cell volume, white blood cell, red blood cell, mean corpuscular volume, haemoglobin, mean corpuscular haemoglobin, mean corpuscular haemoglobin concentration, neutrophils, and lymphocytes, were not affected (P>0.05) by the replacement of bone ash with eggshell meal in the diets.

Eggshell meal improved (P<0.05) mean corpuscular haemoglobin values (42.85 vs. 42.20 vs. 45.60 vs. 43.40 and 47.95 pg). Birds fed treatment T_5 and T_3 had optimal results followed by those fed treatments T_1, then T_2 and T_4. The study also showed that values obtained for mean corpuscular volume (137.25 vs. 137.95 vs. 142.95 vs. 140.45 and 148.45 fl) for birds in treatment T_5 was best improved (P<0.05) followed by those in T_3 and then T_4. Treatments T_1 and T_2 had the least values. It is concluded that the non-significant difference in the values obtained for most of the haematological indices is an indication of the safety and adequacy of the test ingredient (Alu *et al.* 2009). The values obtained on PCV for all the treatment groups were within the normal range of 24.9-45.2% as reported by Mitruka and Rawnsley (1977).

It is also in agreement with the range (22-26%) as reported by Ameen *et al.* (2007). The range of the result obtained on lymphocyte (96.0-97.5%) is in contrast with the reference normal values of 43.9-81.2% as reported by Mitruka and Rawnsley (1977) and 64-74% as reported by Ameen *et al.* (2007). The result obtained on neutrophils and WBC was below the normal range implying that the birds may not be able to perform their phagocytic functions (Alu *et al*. 2009).

There was no (P>0.05) effect of replacing bone ash with eggshell meal on the urea, glucose, triglyceride and protein. The effect replacing bone ash with eggshell meal on the nutrient digestibility by broiler birds is presented in Table 5.

The results obtained on the coefficient of digestibility of dry matter, crude protein, crude fibre, ether extract, ash and nitrogen-free extract were not affected (P>0.05) by the replacement of bone ash with eggshell meal.

The non significant difference observed on the digestibility of the nutrients agrees with the earlier report of Emmanuel (2008) who investigated the effect of replacing bone ash with rock phosphate at 0, 25, 50, 75 and 100% inclusion levels in the diets of boiler finisher birds and recorded a slight increase with increase in the levels of rock phosphate in the diets.

According to McDonald *et al.* (1992), digestibility is affected by feed composition that is, feed ingredients influence the level of digestion in the animal. Conclusively, the test ingredient used in this study does not have the potential of influencing digestion although Borges (1993) reported that feed intake can be influenced by dietary calcium level which seems to act on the hypothalamus including the release of neuropeniphrine, a neurotransmitter that acts on the central nervous system thereby stimulating feed intake. Similarly, Tumova *et al.* (2004) reported that that non exposure of feed to the digestive action of the enzymes could hinder the digestibility of some feed nutrients.

Table 2 12 Percentage and energy composition of the experimental diets for broiler finisher birds (%)

Feed stuffs	Treatments				
	T_1 (0%)	T_2 (25%)	T_3 (50%)	T_4 (75%)	T_5 (100%)
Groundnut cake	18.50	18.50	18.50	18.50	18.50
Maize	14.20	14.20	14.20	14.20	14.20
Full fat Soybean	13.50	13.50	13.50	13.50	13.50
Cassava peel meal	10.50	10.50	10.50	10.50	10.50
Rice offal	10.05	10.05	10.05	10.05	10.05
Blood meal	2.50	2.50	2.50	2.50	2.50
Bone ash	3.50	2.625	1.75	0.825	-
Eggshell meal	-	2.875	1.75	0.625	3.50
Sodium chloride	0.50	0.50	0.50	0.50	0.50
Methionine	0.25	0.25	0.25	0.25	0.25
Lysine	0.25	0.25	0.25	0.25	0.25
Premix	0.25	0.25	0.25	0.25	0.25
Total	100.00	100.00	100.00	100.00	100.00

The premix (vitamin-mineral) supplied the following per 100 kg of diet: Vitamin A: 1500 I.U; Vitamin D_3: 30000 I.U; Vitamin E: 3000 I.U; Vitamin K: 2.50 mg; Thiamine (B_1): 200 mg; Riboflavin (B_2): 600 mg; Pyridoxine (B_6): 600 mg; Niacin 40 mg; Vitamin (B_{12}): 2 mg; Pantothenic acid: 10 mg; Folic acid: 100 mg; Biotin: 8 mg; Choline chlorine: 50 g; Antioxidant: 12.5 g; Manganese: 96 g; Zinc: 6 g; Iron: 24 g; Copper: 0.6 g; Iodine: 0.14 g; Selenium: 24 mg and Cobalt: 214 mg.

Table 3 Chemical and energy composition of the experimental diets

Nutrient	T_1 (0%)	T_2 (25%)	T_3 (50%)	T_4 (75%)	T_5 (100%)
Dry matter	92.20	93.20	91.80	92.20	91.60
Crude protein	20.20	20.23	20.25	20.19	20.22
Crude fibre	8.50	8.51	8.50	8.41	8.40
Ether extract	5.88	4.10	4.40	3.13	4.44
Ash	15.64	16.91	16.15	16.41	12.45
N.F.E.	5.51	7.27	7.34	11.82	17.75
[b] Energy (kcal/kg ME)	2865.53	2865.53	2865.53	2865.53	2865.53
[a] Calcium (%)	1.06	1.12	1.19	1.21	1.26
[a] Phosphorus(%)	0.77	0.73	0.69	0.64	0.58

a Calculated from NRC (1979).
[b] Calculation from Pauzenga (1985).

Table 4 Effect of replacing bone ash with eggshell meal on haematological parameters and serum biochemical variables of finishing broiler birds

Parameters	T_1 (0%)	T_2 (25%)	T_3 (50%)	T_4 (75%)	T_5 (100%)	SEM	LOS
Haematology							
PCV (%)	32.50	29.50	29.50	28.50	28.00	1.41	NS
Hb (g/dL)	10.10	9.05	9.40	8.85	9.00	0.47	NS
RBC (10^6 µL)	2.36	2.15	2.03	2.04	1.89	0.18	NS
WBC (10^5 µL)	2.88	2.43	2.43	2.46	2.40	0.16	NS
MCH (pg)	42.85^{c}	42.2^{c}	45.6^{ab}	43.4^{c}	47.95^{a}	1.15	*
MCHC (g/dL)	31.20	30.60	31.95	30.90	22.25	0.58	NS
MCV (fl)	137.25^{c}	137.95^{c}	142.95^{b}	140.45^{b}	148.45^{a}	1.85	*
Neutrophils (%)	3.00	4.00	2.50	3.50	4.00	0.85	NS
Lymphocytes (%)	97.00	96.00	97.50	96.50	96.00	0.84	NS
Serum Variables							
Urea (mmol/L)	1.65	1.45	1.40	1.10	1.35	0.13	NS
Glucose (mmol/L)	11.05	11.75	11.35	11.90	12.25	0.83	NS
Triglyceride (mmol/L)	0.75	1.20	0.90	2.05	3.80	1.35	NS
Protein (g/L)	45.00	40.00	27.80	44.55	32.95	12.19	NS

The means within the same row with at least one common letter, do not have significant difference (P>0.05).
* (P<0.05); SEM: standard error of means; LOS: level of significanc and NS: non significant (P>0.05).

Table 5 Effect of replacing bone ash with eggshell meal on nutrient digestibility by broiler finisher birds

Nutrient	T_1 (0%)	T_2 (25%)	T_3 (50%)	T_4 (75%)	T_5 (100%)	SEM	LOS
Dry matter digestibility (%)	59.46	54.05	59.46	62.16	56.75	0.11	NS
Crude protein digestibility (%)	68.57	66.15	72.31	72.31	72.86	0.12	NS
Crude fibre digestibility (%)	69.00	73.33	75.65	71.82	67.53	1.83	NS
Ether extract digestibility (%)	82.00	70.59	89.47	84.61	89.47	1.76	NS
Ash digestibility (%)	81.62	75.13	78.69	76.12	71.41	1.98	NS
NFE digestibility (%)	26.36	32.92	37.49	28.13	27.75	1.94	NS

SEM: standard error of means; LOS: level of significanc and NS: non significant (P>0.05).

The short time spent in the GIT by the feed due to high water intake may also explain the reason for the nonsignificance of the nutrient digestibility.

CONCLUSION

The findings on the effect of replacing bone ash with eggshell meal on the haematological parameters, serum biochemical characteristics and nutrient digestibility of finishing broiler birds indicated the safety of the (eggshell) in the diets and can therefore, be used to completely (100%) replace bone ash without producing negative health status or reduce digestibility of the finishing broilers. It is also recommended that further research should be carried out to test the effect of eggshell meal on mineralization of the bone.

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