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Nutrient Retention and Serum Profile of Broilers Fed Fermented African Locust Beans (*Parkia filicoide*)

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ABSTRACT

Although the African Locust bean (*Parkia filicoide*) tree is widely distributed in Northern Nigeria, its application as an alternative replacement of conventional plant protein source in poultry feeds has not gained significant prominence. This study evaluates the inclusion level of fermented African locust bean seeds in broiler diets. One hundred and twenty Lohman broilers day old chicks were randomly divided into six experimental groups of two replicates each were used in a 9 weeks feeding trial to evaluate percentage of nutrients retained and serum indices of broilers. Dietary treatments were as follows: T1, T2, T3, T4, T5 and T6 representing 0, 25, 50, 75 and 100% replacement of groundnut cake replacement with fermented African locust bean seeds and 100% unfermented African locust beans. Treatment effects was significant ($p<0.05$) in the percentage of nitrogen, lipids, total ash, calcium and phosphorus retained. Crude fibre retention was not significantly ($p>0.05$) affected by experimental treatment. Treatment effect was significant ($p<0.05$) with fermented African locust bean seeds based groups giving better nitrogen, lipids and phosphorus retention with T5 showing best values. Nitrogen retention is positively ($p<0.05$) correlated with total serum protein, cholesterol, Hb/PCV and feed treatment ($p<0.01$); while crude fibre retention is negatively ($p<0.05$) correlated with all serum indices and treatment. Percentage retention of lipids is positively ($p<0.05$) correlated with total serum protein and cholesterol and so also with Hb/PCV and FALB inclusion at ($p<0.01$). Digestible ash is negatively ($p<0.01$) correlated with total serum protein and also negatively ($p<0.01$) correlated other serum indices and fermented African locust bean seeds inclusion at ($p<0.05$). Fermentation of African locust beans improved nutrient retention at both Gastrointestinal Tract (GIT) and portal system and availability for tissue synthesis.

Key words: African locust bean (*Parkia filicoide*), fermentation, nutrient retention, serum

INTRODUCTION

The competitive demand for conventional plant proteins source particularly soybean meal and groundnut cake has led to high cost of animal feed in Nigeria. This has necessitated the search for alternative replacement that have comparative nutritive values to and preferably cheaper than the conventional protein source (Atteh *et al*., 1995).

The potentials of locust beans seed in the nutrition of both human and animals have been assessed. Improved performance traits, nitrogen retention and feed utilization of broilers fed fermented African locust beans as a replacement of ground nut cake have been reported by Kolo (2000), Ayanwale and Ari (2002) and Dawodu (2009). This positive attribute for fermented locust beans (Makanjuola and Ajayi, 2012) as against the unfermented locust beans which was reported not to support growth in broilers by Fetuga *et al.* (1974) can be linked to improvement in the protein quality of the locust beans and the greater supply of thiamine and riboflavin through processes of fermentation.

Digestion and nutrient balance trials as well as serum profiling are good *in vivo* assessors of feed quality and nutrient availability in both monogastrics and ruminant animals (Aletor and Ogunyemi, 1988; MacDowell *et al.*, 1990; Williams *et al.*, 2001). Implications and interactions in the Gastrointestinal Tract (GIT) of single stomach animals with fermented feed products have been reviewed by Williams *et al.* (2001) while Aletor and Ogunyemi (1988), Williams (1995), Nweze *et al.* (2011) and Olajide (2012) observed relationship between nutrients uptake and certain serum metabolites.

This study investigates nutrient retention, serum profile and the relationship between nutrient retention and serum indices of broilers fed fermented African locust beans (*Parkia filicoide*) as a replacement of groundnut cake.

MATERIALS AND METHODS

African locust beans seed collection and fermentation process: African locust beans seeds were obtained from commercial farmers in Shabu, Lafia in Nigeria. The seeds were removed from the pods and washed clean of the yellow pulp as described by Ayanwale and Ari (2002). A natural fermentation technique adopted by local Nigerian women (Campbell-Platt, 1980) in which no enzyme or starter culture were added was adopted in this research. This local fermentation process also described by Fetuga *et al.* (1974) involves boiling the black seeds for about twelve hours with no potash added using large pots and firewood until the seeds are partially softened. The cooked seeds were gently pounded and later washed with clean water to remove the seed coat and the seed coat removal is facilitated by the use of sand as an abrasive agent. The decorticated seeds were further cooked for one hour and ensiled in a nylon bag. The fermentation process last until the seeds turn dark brown and softened, the time frame was twenty four hours. The fermented seeds were then sun-dried to a constant weight and milled using hammer mill to produce Fermented African Locust Beans (FALB) meal. The chemical composition of the FALB sample was analyzed according to AOAC (1990) method as reported by Ayanwale and Ari (2002). This forms the basis of experimental feeds formulation. The groundnut cake GNC was purchased from a commercial producer in Minna, Niger state.

Experimental treatment and diet: One hundred and twenty Lohman broilers day old chicks were used for experiment. The birds were randomly divided into six experimental groups of two replicates each at both starter and finisher phases and designated as thus:

T1 Control: 100% GNC and 0% FALB
T2: 75% GNC and 25% FALB
T3: 50% GNC and 50% FALB
T4: 25%GNC and 75% FALB
T5: 0% GNC and 100% FALB
T6: 0% GNC and 100% UALB

Where, UALB is Unfermented African Locust Beans

The composition of the diet is presented in Table 1. All ingredients were supplied and adjusted to make the diets isocaloric and isonitrogenous at 23% CP and 3000 kcal kg^{-1} for starter and 19% CP and 2800 kcal kg^{-1} finisher diets. All experimental birds were given feed and water *ad libitum* while routine management and vaccination were uniformly undertaken during the nine weeks feeding trial.

Digestibility trail: At 8 weeks, two birds from each replicate were randomly selected from each replicate of the treatment groups and placed in metabolic cage for total faecal collection according to the methods described by Longe (1980). The birds were allowed five days adjustment period and faecal collection was made daily for the last 7 days of the experiment. Each day's collection were bulk for each treatment group, weighed, dried and stored in screw capped bottles and taken for analysis. The chemical composition of the faecal samples was analyzed according to AOAC (1990) methods.

Serum analysis: Blood samples were obtained from the experimental chickens by neck decapitation. The samples from each group were collected into a labeled Ethylene Diamine Tetra Acetic Acid (EDTA) bottles for analysis at General Hospital hematological laboratory, Minna. Packed Cell Volume (PCV), the hemoglobin content and cholesterol were determined by the methods of Green (1976) and Aletor and Ogunyemi (1988).

Statistical analysis: The data collected were subjected to Analysis of Variance (ANOVA), correlation and means were separated where there were significant differences using Duncan's Multiple Range Test using SPSS 16.0.

RESULTS

The compositions of experimental diets are presented (Table 1, 3). The nutrient composition of these feeds is consistent with NRC (1994) recommended feeding standards for broilers. The

Table 1: Composition of diets for broilers (%)

	Starting diets						Finishing diets					
Ingredients	T1	T2	T3	T4	T5	T6	T1	T2	T3	T4	T5	T6
Maize	57.00	53.23	49.50	45.80	42.10	42.10	54.55	52.70	50.80	49.00	47.00	47.00
Groundnut cake	20.45	15.35	10.20	5.10			21.05	15.80	10.55	5.25		
Fermented locust bean		8.90	17.85	26.75	35.65			7.25	14.80	21.75	29.00	
Unfermented locust bean						35.65						29.00
Palm oil	0.55	0.50	0.45	0.25	0.25	0.25	0.50	0.25	0.15			
Rice bran	5.00	5.00	5.00	5.00	5.00	5.00	7.00	7.00	7.00	7.00	7.00	7.00
Fish meal	3.00	3.00	3.00	3.00	3.00	3.00	3.00	3.00	3.00	3.00	3.00	3.00
Blood meal	7.00	7.00	7.00	7.00	7.00	7.00	7.00	7.00	7.00	7.00	7.00	7.00
Bone meal	4.00	4.00	4.00	4.00	4.00	4.00	4.00	4.00	4.00	4.00	4.00	4.00
Oyster shell	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00
Salt	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30
Premix*	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
DL- methionine	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
L-lysine	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20

Premix* To provide the following per KG of diet: Vitamin A: 9,000 IU, Vitamin D3: 2,000 IU, Vitamin E: 18 IU, Vitamin B1: 1.8 mg, Vitamin B2: 6.6 mg, Vitamin B3: 10 mg, Vitamin B5: 30 mg, Vitamin B6: 3.0 mg, Vitamin B9: 1 mg, Vitamin B12: 1.5 mg, Vitamin K3: 2 mg, Vitamin H2: 0.01 mg, Folic acid: 0 .21 m g, Nicotinic acid: 0.65 mg, Biotin: 0.14 mg, Choline chloride: 500 mg, Fe: 50 mg, Mn: 100 mg, Cu: 10 mg, Zn: 85 mg, I: 1 mg, Se: 0.2 mg

Table 2: Composition of GNC, fermented and unfermented African locust bean seeds (%)

Composition (%)	GNC	Unfermented	Fermented
Dry matter	90.24	89.62	88.30
Crude protein	49.33	21.02	34.00
Ether extractable material	11.14	30.64	39.20
Crude fibre	5.02	8.57	7.10
Total ash	5.51	5.30	6.22
Nitrogen free extract	30	34.47	13.48

Source: Excerpted from Ayanwale and Ari (2002)

Table 3: Proximate composition of test diets for broilers (%)

	Starting diets						Finishing diets					
Ingredients	T1	T2	T3	T4	T5	T6	T1	T2	T3	T4	T5	T6
Dry matter	89.60	90.34	90.13	90.60	91.14	90.75	91.00	90.75	90.60	90.40	90.30	90.28
Crude protein	22.40	22.40	22.20	22.70	22.75	22.00	20.25	19.95	20.00	20.10	20.90	21.00
Crude fibre	19.75	21.24	21.25	21.35	21.00	21.95	20.10	22.15	21.75	21.80	21.08	21.80
Ether extract	14.69	14.50	13.00	13.90	12.00	10.66	15.00	13.85	13.45	12.90	13.00	10.95
Total ash	8.45	8.85	9.40	9.20	9.40	5.20	8.21	10.00	10.15	10.75	10.80	5.25
Nitrogen free extract (NFE)	34.71	33.11	33.95	32.85	34.12	40.19	36.44	34.05	34.65	34.65	34.22	41.00
Ca	1.76	2.16	2.62	2.58	2.82	2.29	2.64	2.72	2.78	2.85	3.98	2.97
P	0.85	0.59	0.81	0.62	0.94	1.45	0.63	0.78	0.67	0.82	1.07	0.89

proximate composition of diets were not significantly altered by increase in replacement values of GNC with FALB and UALB. The chemical composition of the FALB, UALB and GNC presented in Table 2 is indicative of the positive effect of fermentation which resulted in the increase in crude protein of FALB from 21.02-34.0%. The crude protein values for the experimental feeds (Table 3) were in the range of 22.00-22.75% and 19.95-20.90% at the starter and finisher diets, respectively. Dietary treatment 5 had best values for CP in the two phases.

Nutrient retention values are presented in Table 4. There were significant ($p<0.05$) differences in the percentage nitrogen, lipids, total ash, Ca and P retention while crude fibre retention were not significantly ($p>0.05$) affected by the replacement of GNC with FALB. Birds in T5 with 100% FALB gave the best values for nitrogen (89.58%), lipids (93.69%), phosphorus (93.00%) while T1 was least in nitrogen, lipids and phosphorous retention with the following values 77.12, 78.21 and 81.00%, respectively. Digestible ash and calcium retention were better with T1 group with 90.38% as digestible ash values for T1 and T3 and 87.12% as calcium retention values for T1. However, addition of FALB in the other treatments groups showed significantly ($p<0.05$) better for nitrogen, lipids and phosphorus retention.

The results of the serum indices are presented in Table 5. There were significant ($p<0.05$) differences in total serum protein, cholesterol and Packed Cell Volume (PCV) expressed as heamoglobin concentration between dietary treatment groups. Birds in T5 with 100% FALB presented the higher values for total serum protein (4.68 g dL^{-1}), cholesterol values were lowest (1.39 mm L^{-1}) with T6 (100% replacement with UALB). Birds in T3 with 50% FALB gave relatively better values for Hb/PCV (10.98 mm L^{-1}). T5 with 100% FALB presented on the average better values for serum indices under investigation.

The results in Table 6 present the correlations matrix of percentage nutrient retention and serum indices of experimental birds. This defines relation between nutrient retention and serum

Table 4: Nutrient retention of broilers fed with fermented African locust beans (*Parkia filicoide)*

	Experimental diets						
Nutrients (%)	T1	T2	T3	T4	T5	T6	SEM
Nitrogen (%)	77.12^{e}	79.87^{d}	83.02^{c}	83.43^{bc}	89.54^{a}	84.07^{b}	±1.16
Crude fibre (%)	83.25^{NS}	81.14^{NS}	79.62^{NS}	82.13^{NS}	72.86^{NS}	79.26^{NS}	±1.58
Lipids (%)	78.21^{f}	83.59^{e}	90.88^{c}	92.20^{b}	93.69^{a}	87.91^{d}	±1.62
Digestible ash (%)	90.38^{a}	90.23^{ab}	90.39^{a}	89.64^{b}	87.16^{c}	89.93^{ab}	±0.35
Ca (%)	91.80^{a}	89.40^{c}	88.92^{c}	87.12^{e}	90.67^{b}	87.92^{d}	±0.43
P (%)	81.00^{e}	83.32^{d}	84.96^{c}	89.53^{b}	93.00^{a}	89.40^{b}	±1.24

Means with different alphabets in the same row were significantly different at p<0.05, NS: Non significantly different at p>0.05, SEM: Standard error of means

Table 5: Serum profile of broilers fed with fermented African locust beans (*Parkia filicoide*)

	Experimental diets						
Serum indices	T1	T2	T3	T4	T5	T6	SEM
Total serum protein (g dL^{-1})	4.28^{bc}	4.38^{b}	4.04^{d}	4.19^{c}	4.68^{a}	4.28^{bc}	±0.60
Cholesterol (mm L^{-1})	1.58^{c}	1.51^{c}	1.78^{a}	1.51^{c}	1.68^{b}	1.39^{d}	±0.38
Hb/PCV (mm L^{-1})	6.56^{e}	10.08^{c}	10.98^{a}	9.99^{c}	10.49^{b}	9.79^{d}	±0.43

Means with different alphabets in the same row were significantly different at p<0.05, NS: Non significantly different at p>0.05, SEM: Standard error of means

Table 6: Correlations matrix of percentage nutrient retention and serum indices

	Nitrogen	Crude fibre	Lipids	Digestible ash	Ca	P	Total serum protein	Cholesterol	Hb/PCV	Treatment
Nitrogen										
Crude Fibre	-0.615*									
Lipids	0.888**	-0.416								
Digestible ash	-0.836**	0.564	-0.594*							
Ca	-0.148	-0.170	-0.460	-0.276						
P	0.942**	-0.498	0.854**	-0.790**	-0.326					
Total serum protein	0.497	-0.484	0.116	-0.786**	0.500	0.465				
Cholesterol	0.215	-0.281	0.285	-0.234	0.417	-0.058	-0.058			
Hb/PCV	0.684*	-0.341	0.830**	-0.314	-0.493	0.579*	0.017	0.251		
Treatment	0.814**	-0.394	0.727**	-0.520	-0.487	0.895**	0.254	-0.276	0.558	

*Correlation is significant at the 0.05 level (2-tailed), **Correlation is significant at the 0.01 level (2-tailed)

indices measured as thus: nitrogen retention is positively (p<0.05) correlated with total serum protein, cholesterol, Hb/PCV and treatment diets (p<0.01), while crude fibre retention is negatively (p<0.05) correlated with all serum indices and dietary treatment. Percentage retention of lipids is positively (p<0.05) correlated with total serum protein and cholesterol and so also with Hb/PCV and dietary treatment at (p<0.01). Digestible ash is negatively (p<0.01) correlated with total serum protein and also negatively (p<0.01) correlated other serum indices and dietary treatment at (p<0.05).

Calcium retention is positively (p<0.05) correlated with total serum protein and cholesterol and negatively correlated with Hb/PCV and dietary treatment. The correlation between phosphorus retention and total serum protein and Hb/PCV is positive (p<0.05), similarly dietary treatment and P retention is positive ($p<0.01$) while a negative correlation ($p<0.05$) is recorded in the relation between P and cholesterol.

DISCUSSION

The proximate composition of diets were not significantly altered by the increase in replacement values of GNC with FALB and UALB due to the fact that the diets were made to be isocaloric and isonitrogenous . The positive effect of fermentation led to the improvement in the crude protein of the FALB which also led to the reduction in the reduction in carbohydrates which are presumed to be used by fermenting microbes. This is in agreement with the findings of Odebunmi *et al.* (2010).

The implication of the results of nutrient retention lies in the fact that it provides close link between performance and nutrient metabolism especially nitrogen retention which can be closely linked to growth and protein metabolism. Therefore, the higher percentage of nitrogen retention values recorded in T5 indicates the efficiency of nitrogen metabolism of birds fed FALB for tissue synthesis. This process of tissue synthesis for growth may have been supported by the higher retention values for lipids and phosphorus for birds fed 100% FALB based diet. This finding is supported by Williams *et al.* (2001), Ayanwale and Ari (2002) and Khosravi *et al.* (2012) who reported that fermentation action products allows for increased utilization of feed by chickens.

The combined effects of thiamine and riboflavin in FALB (Ayanwale and Ari, 2002) is responsible for the high retention of nutrients especially nitrogen. The low nutrient retention of broilers fed UALB and GNC could not be attributed to inadequate intake or quality of nutrients like protein but minerals and vitamin imbalances. The imbalances of micro-nutrients in UALB and GNC is implicated in the poor utilization of feeds and thus reduced nutrient digestibility due to marginal deficiency of the vitamins in these protein sources which are below NRC (1994) recommendation for broilers. The probiotic effects of fermented African locust beans and interactions host GIT microflora provides a positive balance in favour beneficial microorganism through the process of colonization (Williams *et al.*, 2001). This leads to improvement in nutrient utilization by broilers and bioavailability of micro nutrients for metabolic processes (Coulibaly *et al.*, 2011).

Even though crude fibre retention did not differ significantly among treatment diets, the lowest value for crude fibre retention was recorded for birds fed 100% FALB. This suggest that fibrous feedstuff are converted to bacteria protein by GIT microorganism.

Positive correlation between nitrogen retention, total serum protein, cholesterol and dietary treatment indicates that increase in FALB utilization increased nitrogen retained and portal nutrients (protein, Lipids and phosphorus) required for tissue synthesis. This finding is in agreement with Williams (1995) and Nunes *et al.* (1991) who were of the view that portal uptake of protein is related to protein ingested by birds.

The negative correlation between dietary treatments and crude fibre retention indicates that crude fibre retention decreases with increase in FALB in the diet. This is consistent with Piva *et al.* (1996) who observed that as carbohydrates sources (starch, fibre and other fermentable carbohydrates) become depleted due to fermentation in the large intestine of single-stomached animals, carbohydrates sources of the caecum decreases. This further suggests that fibrous feedstuff are converted to bacteria protein by increased interaction between host GIT microorganism and fermentation microbes in fermented feedstuff.

Results obtained in this study indicate the nutritional superiority of fermented oilseeds in providing nutrients required by broilers for tissue synthesis.

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