Evaluation of fermented Locust bean meal (*Parkia biglobosa)* as replacement to soybean meal in production performance, blood profile and gut morphology of broiler chicken.

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Abstract

An eight week feeding trial was carried out on the evaluation of fermented locust bean meal (FLBM) *Parkia biglobosa* as replacement to soybean meal (SBM) in broiler production. Ninety day old broilers were allotted randomly to three treatment diets and each replicated thrice. Diets I (control) contained 0% FLBM while diets II and III had 50 and 100% FLBM inclusion as replacement of soybean meal in both starter and finisher phases. At starter phase, results revealed body weight of bird were not significant, the feed intake, feed conversion ratio, and mortality of birds on diet II were lower and significantly ($P<0.05$) different from others. At the finisher level the weight gain of broilers fed diet III were higher significantly ($P<0.05$) while birds fed diet II had the best feed conversion ratio significantly ($P<0.05$) as compared to other diets. The packed cell volume (PCV), haemoglobin, red blood cell and platelets of birds fed diet II were higher significantly ($P<0.05$) as compared to those fed diets I and III and are within standard range for healthy birds. Blood serum showed that the glucose, aspartate transaminase, cholesterol and creatinine of those fed diet III were significantly ($P<0.05$) higher than others. The gut morphology revealed that the liver, lungs, intestine, gizzard and heart of those fed diets II and III were significantly ($P<0.05$) similar and smaller than those on control with no traces of inflammation. Conclusively, FLBM could replace SBM in broiler diet without adverse effect on the production performance, 50% inclusion level of FLBM with SBM is recommended in broiler production.

Keywords: Fermented locust bean meal; soya bean meal; broilers; haematology; production performance

Introduction

The competitive demand for conventional plant protein origin (soybean meal and groundnut cake) has led to high cost of livestock feed in Nigeria. Hence, the need for additional protein supplies to promote sustainable monogastric livestock for least cost formulation and production (Ari and Ayanwale 2012). One of such legume is *Parkia biglobosa*, commonly known as African locust bean, a tropical tree which is native to Africa and widely distributed in the savannah region.

The tree is usually and carefully preserved by inhabitants of the areas where it grows because they are valuable source of reliable food, especially the seeds which serves as source of useful ingredients for consumption as Daddawa in Hausa and Iru in Yoruba (Campbell-Platt, 1980). *Parkia biglobosa* has high protein and better amino acid profile that recommends it for use as a protein substitute for human food and animal feed (Alabi *et al*., 2005).

The utilization of locust bean in monogastric nutrition as replacement for a protein source i.e groundnut cake was reported to improved production performance, nitrogen retention and feed utilization, with varied results (Bridget *et al*., 2004; Alabi *et al*., 2005). Ayanwale and Ari (2002) and Dawodu (2009) found a positive attribute for fermented locust beans as against the unfermented locust bean which was said to inhibit broiler growth due to less protein quality and essential vitamins (Fetuga *et al.,* 1974). However, Anti-nutritional factors (ANFs) such as tannins, oxalate and hydrogen cyanide had been reported to limit the utilization of locust bean as feed ingredient (Apata, 2003). Fermentation has been reported to destroy some natural toxins which may occur in beans, improve the nutritive-value, increase digestibility and enhance growth (Bridget *et al*., 2004).

This study was designed to evaluate the replacement of fermented locust bean meal with soya bean meal in broiler diet in relation to production performance, gut morphology serum and hematological blood profile.

Materials and methods

Location of experiment

The experiment was conducted at the Teaching and Research Farms of Bowen University Iwo, Nigeria. Environmental temperature range of 15- 28^{0}C and mean annual rainfall of 1400mm, subject to climate change.

Preparation of experimental diet

The *p. biglobosa* seeds were sourced from a nearby farm within the town. The *p. biglobosa* seeds were removed from the pods and washed clean off the pulp as described by Ayanwale and Ari (2002). A fermentation process adapted from Fetugal *et al.,* (1974) and local processing industries in Nigeria was used. The seeds were boiled for 12 hours without enzyme or starter culture with large pots in open firewood to soften the seed coats. The seeds were lightly pounded with pestle and mortar to separate the seed coats from the seeds. The decorticated seeds were wrapped in a polyethylene bag, placed inside a basket to drain. The fermentation process lasted until the seeds turn dark brown and softened, which took about 24hours. The seeds were later oven dried at a temperature of 40^0 C for 3days, to prevent denaturation of the protein in locust bean meal and then ground into powdery form to produce fermented locust bean meal (FLBM). The proximate composition of FLBM was analysed using AOAC (1990) and reported in table 1. The other ingredients used for formulating the diets were obtained from a reputable feed meal at Iwo.

Table 1: Proximate compositions of locust beans

Components	**Locust beans**
Dry matter (%)	8.4
Crude protein (%)	6.56
Ether extract (%)	1.8
Crude fibre (%)	11.75
Ash (%)	4.18
Nitrogen free extract (%)	67.31

Experimental procedure and birds' management

Three test diets were formulated, FLBM was used to replace SBM at two inclusion levels. All ingredients were supplied and adjusted to make diets isocaloric and isonitrogenous. The gross composition of the experimental diet of the starter and finisher phase is shown in table 2. Ninety abro acre day old broilers were randomly assigned to three dietary treatments and replicated thrice. Feed and water were provided *ad libitum* and all standard routine management practices, medication and vaccination are strictly observed. Daily feed intake (FI) and weekly body weight (BW) of birds were recorded.

The vaccination programme was planned and strictly followed in accordance with the Immuno-prophylactic and preventive guide for broilers recommended by the veterinary department. Dosages were given according to the specifications of the manufacturers.

Day 1 : @hatchery Marek disease vaccine Subcutaneous
Day 9-10 : Infectious bursal disease Drinking water (Gumboro)
Day14 : Newcastle disease vaccine Drinking water (Lasota)
Day 16-20 : Coccidiostat Drinking water
Day 21 : Infectious Bursal Disease (Gumboro) Drinking water
Day 28 : Newcastle Disease vaccine Drinking water (Lasota)

Table 2: Gross composition of experimental diets

	Starter			**Finisher**		
Ingredients	**I**	**II**	**III**	**I**	**II**	**III**
Maize	57.5	57.5	57.5	70.5	70.5	70.5
Locust beans	-	3.5	7	-	3.	6
Soya bean meal	7	3.5	-	6	3	-
Wheat offal	3	3	3	2	2	2
Fish meal (72%)	9	9	9	3	3	3
palm kernel cake	3	3	3	3	3	3
Groundnut cake	15	15	15	10	10	10
Bone meal	3	3	3	3	3	3
Oyster shell	2	2	2	2	2	2
Salt	0.25	0.25	0.25	0.25	0.25	0.25
Premix	0.25	0.25	0.25	0.25	0.25	0.25
Total (kg)	100	100	100	100	100	100
Lysine	0.1	0.1	0.1	0.1	0.1	0.1
Methionine	0.1	0.1	0.1	0.1	0.1	0.1
Calculated composition						
Metabolizable energy (kcal kg^{-1})	2900.36	2815.66	2730.96	3010.34	3010.24	2865.14
Crude protein (%)	20.17	21.66	21.18	16.18	15.77	15.35
Crude fibre (%)	3.33	3.41	3.48	3.24	3.32	3.39

Premix* To provide the following vitamin/mineral composition: Vitamin B, 6,000,000 IU; Vitamin D3 , 2000000 IU; Vitamin E, 4000mg; Vitamin K3 , 600 mg; Vitamin B1 , 300mg; Vitamin B2 , 3500 mg; Vitamin B6, 800 mg; Vitamin B12, 40 mg; Pantothenate, 1100 mg; nicotinic acid, 15000 mg; folic acid, 250 mg; Biotin, 80 mg; Na2SeO3 , 200mg; FeSo4H2O, 85g; ZnSO4H2O, 90g; CuSO4 H2O, 5g; MnSO4 H2O, 85g.

Blood collection and analysis

Blood samples were collected at the 8thweek of study from experimental birds. Blood sample was collected through the wing web vein punctured method with the use of new syringes. The birds were taken one sample per replicate and allowing free flow of blood into the labelled sterile universal bottles. 5ml of the blood was collected over a labelled sterile sample bottles without anticoagulant and used to determine the biochemical components as described by Tuleun *et al.,* (2009), while another set was collected into sterile bottles with EDTA for haematological analysis.

Gut morphology and carcass analysis

Six (6) birds were randomly selected per treatment (i.e. 2 birds per replicate) for carcass cuts and organ examination at the end of the experiment. Water and feed were withdrawn for 6 hours before the cervical cut of the birds. Each bird was tagged and weighed before and after slaughtering to determine the live weight. Other parameters taken include: defeathered weight, dressed weight, shank, intestine, head, liver, gizzard, lungs, and breast. These weights were expressed as the percentage of the final live weight of the birds.

Statistical analysis

Data obtained were subjected to one way Analysis of variance (ANOVA) software (SAS, 2006). Means with significant difference among the treatments were separated using the Duncan's option of the same software.

Results

The proximate composition of the test ingredient is as shown in table 1. The gross composition of the experimental diet is shown in table 2 for both starter and finisher phases. The feeds were compounded to meet the nutrient requirement as recommended by NRC (1994) for broilers chicken. Crude protein of diet II at starter phase was higher than others, while the metabolizable energy was higher in diet I as compared to other diets at both phases. The crude fibre in diet III was higher as compared to diet I and II at both starter and finisher phases. The composition of the test ingredient compared well with control at both starter and finisher phase. The performance characteristics of broilers fed the experimental diet as shown in table 3, the feed intake (FI) and feed conversion ratio (FCR) were significantly different ($P<0.05$). At finisher phase, the FLBM inclusion diets (Diet II and III) had higher values significantly ($P<0.05$) for body weight gain (BWG). Feed intake and feed conversion ratio (FCR) differed significantly ($P<0.05$) between diets, birds fed FLBM inclusion (Diet II and III) had better FCR and feed efficiency. The result of the blood profile as shown in table 4, showed that all haematological values are significantly different ($P<0.05$) between the treatment diets. Birds fed diet II had higher values for packed cell volume (PCV), red blood cell (RBC), haemoglobin (Hb) and platelets as compared to other dietary treatments. Diet I had the highest value for white blood cells (WBC). Blood serum showed that the glucose, aspartate transaminase and creatinine of those fed diet III were significantly ($P<0.05$) higher than others. Birds fed diet III had highest value for cholesterol (122mg/dl) which is significant ($P<0.05$) as compared to other diets. Total protein was with highest value (4.28mg/dl) in birds fed diet II which differed significantly ($P<0.05$) from other dietary treatment. As shown in table 5, the organ weights of the birds were significantly ($P<0.05$) affected by the dietary treatments. The gut morphology revealed that the liver, intestine, gizzard and heart of those fed diets I (control) and diet II were similar, while those on diet III are smaller as compared to other diets. Live weight value of birds on the control was superior to all others though not significant ($P>0.05$). Birds on diet III had the least eviscerated weight all through. The shank, wing and breast of birds on diet III had the least values. The head, proventiculus and the spleen were similar across the diets and no particular statistical trend was observed.

Table 3: Performance characteristics of broilers fed experimental diets

	Starter phase				Finisher phase			
Parameters	I	II	III	SEM	I	II	III	SEM
Initial BW (kg)	0.04	0.04	0.04	0.001	0.53	0.54	0.53	0.001
Final BW(kg)	0.53	0.54	0.53	0.001	0.89	0.97	1.03	0.40
Total BWG (kg)	0.49	0.50	0.49	0.001	0.36^{c}	0.48^{ab}	0.50^{a}	15.62
Daily FI (g)	247.59^{a}	237.35^{b}	244.13^{a}	3.01	618.20^{a}	599.09^{b}	564.81^{c}	15.26
Total FI (g)	6.93^{a}	6.66^{b}	6.84^{a}	0.13	17.31^{a}	16.77^{b}	15.81^{c}	0.44
FCR	14.14^{a}	13.32^{b}	13.96^{a}	0.25	48.08^{a}	34.00^{b}	32.62^{bc}	4.76
Feed efficiency	0.07	0.08	0.07	0.01	0.02	0.03	0.03	0.00
Mortality	6.00^{a}	3.00^{b}	6.00^{a}	3.33	0.00	0.00	0.00	0.00

^{abc} means along the same row with different superscripts are significantly ($P<0.05$)different using Duncan's test as post hoc analysis.

Table 4: Haematological and serum parameters of the birds fed experimental diets

Treatments	I	II	III	SEM
Packed cell volume (PCV) (%)	22.00^{b}	24.67^{a}	22.33^{b}	1.03
Heamoglobin (g/d)	7.30^{b}	8.23^{a}	6.97^{b}	0.38
Red blood cell (10^{6}ul)	2.04^{b}	2.46^{a}	2.40^{a}	0.20
White blood cell (106ul)	16.58^{a}	14.60^{b}	15.43^{ab}	1.31
Platelet (%)	147.33^{b}	162.70^{a}	123.70^{c}	5.00
Glucose (mg/dl)	207.40^{b}	207.35^{b}	230.32^{a}	7.50
Aspartate transaminase (i.u/l)	124.07^{c}	152.87^{b}	219.88^{a}	16.58
Alanine transaminase (i.u/l)	10.38	9.60	10.09	1.47
Albumin (mg/dl)	1.63	1.66	1.56	0.12
Total protein (mg/dl)	3.71^{c}	4.28^{a}	3.91^{b}	0.19
Cholesterol (mg/dl)	118.40^{b}	110.37^{c}	122.03^{a}	6.28
Uric acid (mg/dl)	2.93	2.97	2.79	0.21
Creatinine (mg)	2.21^{b}	1.87^{b}	4.37^{a}	0.49

^{abc} means along the same row with different superscripts are significantly ($P<0.05$)different using Duncan's test as post hoc analysis.

Table 5: Carcass and organ weight of experimental birds, calculated as percentage of live weight

Parameters	**I**	**II**	**III**	**SEM**
Live weight (kg)	1.60	1.50	1.48	0.02
Shank (g)	17.68^{a}	17.07^{a}	11.44^{b}	0.80
Wing (g)	5.47^{a}	6.21^{b}	5.41^{c}	0.05
Breast (g)	10.55^{a}	10.02^{ab}	8.93^{c}	0.05
Heart (%)	0.43^{a}	0.41^{a}	0.36^{b}	0.01
Head (g)	2.91	3.00	2.99	0.20
Spleen (%)	0.09	0.09	0.08	0.001
Proventiculus (%)	0.47	0.46	0.46	0.01
Gizzard (%)	2.44^{a}	2.41^{ab}	2.31^{c}	0.01
Lung (%)	0.40^{a}	0.40^{a}	0.27^{b}	0.001
Liver (%)	1.47^{a}	1.40^{ab}	1.28^{c}	0.01
Intestine (%)	4.19^{a}	4.09^{ab}	3.91^{c}	0.01

^{abc} means along the same row with different superscripts are significantly ($P<0.05$)different using Duncan's test as post hoc analysis.

Discussion

The increased feed intake of birds on control observed in this study and compared to low feed intake in FLBM diet can be attributed to the odour of locust bean cum reduced feed acceptability by birds. At the level of inclusion 100% the smell of the FLBM masked the smell of other ingredients making the diets unattractive and possibly unpalatable to the birds, while at 50% FLBM inclusion, birds can still tolerate the aroma. This result is similar to the findings of Odunsi (2003) who reported that feed intake increase in animals if the aroma of their diet is acceptable. However, the improved FCR and feed efficiency in FLBM inclusion diets can be attributed to efficiency of nitrogen metabolism by birds for tissue development. This result corrobate the findings of Ari and Ayanwale (2012), fermented locust bean is characterised with high nitrogen retention which enhance protein utilization in animals. Mortality recorded in diet I and III were similar at the starter phase. This could be attributed to varied management practises as reported by Gugolek *et al*., (2007) precisely handling of hygiene, as no adverse effect of feeding test diet was observed during the post-mortem examinations. The result of the blood profile as shown in table 4 can be attributed to efficient feed utilization in the FLBM diets. PCV, red blood cell, white blood cell and haemoglobin content of chicken blood are a factor of their health status and nutrient utilization (Aderemi and Alabi 2013). All haematological values recorded were within normal range for healthy chicken (Mitruska and rawsley, 1977), implying the test diets did not elicit any adverse effect on the bird health. Also, the haematological result suggest that FLBM inclusion diet did not impair dietary iron availability such as could cause anaemia, the nutrients required for haemopoiesis were made available by the feeds and properly absorbed in the gut (Adenkola *et al.*, 2011). The result observed for total protein could probably be attributed to enhance nitrogen utilization in FLBM diets, even though all diets are isonitrogenous during formulation (Ari and Ayanwale 2012). Albumin result corroborates the report of Sokunbi and Egbunike (2000) that albumin levels tend to remain constant throughout life after reaching a maximum at about 3 weeks of age. The cholesterol value was within the normal range for healthy birds suitable for consumption without possible incidence of arteriosclerosis and coronary heart diseases (Alabi *et al.*, 2016). The Aspartate and Alanine transaminase were within normal range for healthy birds, implying the test diets did not elicit any adverse effect on the birds. These enzymes are a mirror for overall body enzymatic and metabolic process in bird and an index of liver disease. Perhaps the probiotic effect of FLBM and interactions with host gastro intestinal microflora was partly responsible for this (Coulibaly *et al* 2011). The glucose content of those fed diet I and II were lowered compared to diet III and no adverse effect on regulation. By implication those on diet III inhibit glycolysis and thus there is adverse effect on regulation of insulin and or blood sugar. This is similar to findings of Aderemi and Alabi (2013). As revealed in table 5, the utilisation of FLBM as alternative protein feedstuff did not suppress the physiological development of the birds. However, the significant ($P<0.05$) decrease in values of dress cuts (shank, wing and breast) from control with increase in FLBM inclusion can be attributed to crude protein content and its utilization in the diets. As reported by Omerovic *et al., (*2016), low protein diets caused differences in carcass yield, because lower protein and amino acids content (lysine, methionine and threonine) influenced weight gain and performance. The percentage of live weight for visceral organs (gizzard, liver, lungs, spleen, heart, and proventiculus) of the broilers on control and diet II were statistically similar. This suggest that boiling and fermentation process may have reduced the antinutritional factors in the locust bean as observed in the

non-inflammation or hypertrophy of the liver. However, the smaller size observed for visceral organs in birds fed diet III can be attributed to growth depression as a result of accumulation in antinutritional factors at higher inclusion rate (100%). As reported by Ismail *et al* (2008) and Soetan and Oyewale (2009), that the presence of trypsin inhibitor in legume seeds used in animal feeds and human foods causes growth depression, and relative smaller size of organ weights to live weight in broilers fed mucuna seed meal.

Conclusion and application

It was concluded that replacement of SBM up to 50% with FLBM has a good potentials for broiler production. As shown with the production performance, haematological / serum indices and gut morphology, the health of the chicken was not compromised with inclusion of locust beans up to 50 %. In view of optimum production performance, feed utilization and blood profile, 50% inclusion of locust bean to replace soybean is hereby recommended in broilers diet. Further research is recommended for increase inclusion level and application in other farm animals.

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