Growth Performance of Broiler Chickens Fed Diets Containing Shea Nut (*Vitellaria paradoxa*, Gaertn.) Meal Fermented with *Aspergillus niger*

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ABSTRACT Shea nut meal is a by-product of the shea fat industry in West Africa. The objective was to determine the effect of shea nut meal fermentation using *Aspergillus niger* on growth performance of broiler chickens. An expeller shea nut meal was fermented in a closed plastic container for 8 d after the addition of 0.25 g of *A. niger* spores per kg of shea nut meal in 2 parts of water. Each of the 2 shea nut meal samples (the unfermented and fermented meals) replaced wheatfeed in a control diet at 100 g/kg and fed to 128 Ross 308 male broiler chickens (22 to 36 d). There were 8 replicates per diet (2 shea nut meal samples and the control wheatfeed diet) and 4 birds per replicate in cages (0.6 m × 0.6 m × 0.9 m). Analysis of variance of data was used to compare the treatment means. The fermentation method reduced the concentrations of total soluble phenolics (21.9%), bound plus soluble proanthocyanidins (34.5%), soluble proanthocyanidins (24.7%), and hydrolysable tannins (52.9%) in the shea nut meal. Broilers fed the fermented meal exhibited higher ($P < 0.001$) growth performance than those fed the unfermented meal. However, the growth performance of broilers fed each of the shea nut meal-based diets was lower ($P < 0.001$) than that of broilers fed the control diet. Mean live weight gain of broilers fed the fermented shea nut meal diet was 82% of that of broilers fed the control diet. The fermentation of shea nut meal using *A. niger* has the potential to improve the nutritive value of shea nut meal for poultry, but requires further development.

Key words: shea nut meal, *Aspergillus niger*, fermentation, broiler

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INTRODUCTION

Shea nut meal is a by-product that is the residue after fat extraction from shea nuts (*Vitellaria paradoxa*, Gaertn.). It is available in large quantities in West Africa. Shea nut meal has similarities in its nutrient composition to wheatfeed (NRC, 1994). Wheat by-products are used extensively in poultry diets in Western Africa. These by-products are often imported and thus can be scarce and expensive. Therefore, the use of nutritionally improved shea nut meal to replace wheatfeed would provide an alternative and available feed resource.

The major nutritional limitation of shea nut meal for poultry is the presence of anti-nutritive factors, particularly tannins that are in the range of 98.7 to 156.4 g/kg (Okai et al., 1995; Annongu et al., 1996). The tannin concentration is similar to or higher than other high tannin-containing feedstuffs such as sorghum grains (*Sorghum vulgare*) and faba beans (*Vicia faba*). The biological significance of tannins in poultry nutrition is related to their characteristic adverse effects on feed intake (Armstrong et al., 1974) and nutrient utilization (Smulikowska et al., 2001). The amounts of tannins in some foods can be reduced by fermentation (i.e., wet incubation of a feedstuff), although the mechanism by which these components are eliminated is not fully understood (Reddy and Pierson, 1994). The fermentation process can create conditions for the growth of microbes (*Bacillus, Corynebacterium, Klebsiella, Aspergillus, Penicillium, Fusarium,* and *Candida*) that break down tannins (Reddy and Pierson, 1994). The microbial degradation of condensed tannins is less than that of hydrolysable tannins in both aerobic and anaerobic environments (Bhat et al., 1998). Reichert et al. (1980) suggest that, during incubation, tannins may react to form higher oligomeric polymers that are not readily soluble in water and, therefore, are less likely to interfere with digestive enzymes or other proteins.

Fermentation is a unique process with great potential for recycling some agro-industrial by-products into useful animal feeds in developing countries. The process does not require the use of chemicals and is easy

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Table 1. Composition (g/kg) of the experimental diets

Ingredient	Amount (g/kg)	
	Control (wheatfeed) diet	Shea nut meal diets
Maize (*Zea mays*)	570.0	570.0
Dehulled soybean meal	220.0	220.0
White fishmeal	50.0	50.0
Soybean oil	25.0	25.0
Wheatfeed	100.0	—
Shea nut meal[1]	—	100.0
Lysine hydrochloride	2.5	2.5
DL-Methionine	2.5	2.5
Threonine	0.5	0.5
Monodicalcium phosphate	8.0	8.0
Limestone	15.0	15.0
Vitamin and trace element premix[2]	2.5	2.5
Salt	3.5	3.5
Calculated composition (g/kg)		
Crude protein	209.3	208.4
Lysine	13.2	13.2
Methionine + cystine	9.5	9.6
Metabolizable energy (kcal/kg)	3,110	3,230

[1]Nutrient compositions of all shea nut meal samples and ME (3,577 kcal/kg) were assumed to be similar.

[2]Vitamin-trace mineral premix for broilers (Ian Hollows Feed Supplement, Whitchurch, UK) added per kilogram of diet: vitamin A, 16,000 IU (as retinol acetate); vitamin D_3, 3,000 IU (as cholecalciferol); vitamin E, 25 mg (as α tocopherol acetate); thiamine, 3 mg (as thiamine hydrochloride); riboflavin, 10 mg; pyridoxine, 3 mg (as pyridoxine hydrochloride); vitamin B_{12}, 0.015 mg (as cyanocobalamin); nicotinamide, 60 mg; pantothenic acid, 15 mg; folic acid, 1.5 mg; biotin, 0.125 mg; choline chloride, 200 mg; iron, 20 mg (as ferrous sulfate); cobalt, 1 mg (as cobalt acetate); manganese, 100 mg (as manganous oxide); copper, 10 mg (as copper sulfate); zinc, 80 mg (as zinc oxide); iodine, 1 mg (as calcium iodate); selenium; 0.2 mg (as sodium selenite), and molybdenum, 0.5 mg (as sodium molybdate).

to manage in on-farm conditions or on an industrial scale. Fermentation processes using aspergilli have been used to improve the nutritive value of some feedstuffs such as soybeans (Chah et al., 1975; Mathivanan et al., 2006), guar meal (Nagra et al., 1998), and koji feed (Yamamoto et al., 2007) for poultry. The desirable characteristics of the fermented products include their acceptability by birds (Nagra et al., 1998) and nutrient availability (Hong et al., 2004). *Aspergillus niger* is a fungus that has the capacity to produce enzymes such as hemicellulases, hydrolases, pectinases, lipases, and tannases (Pinto et al., 2001; Mathivanan et al., 2006). Thus it has been used extensively in the improvement of agricultural by-products through its action on substrates such as nonstarch polysaccharides and proteins (Ong et al., 2007; Aderemi and Nworgu, 2007) or structurally modifying anti-nutritive factors (Hong et al., 2004).

Therefore, this study was undertaken to determine if fermentation of an expeller shea nut meal using *A. niger* could change its nutritional value. The growth performance of broiler chickens was compared relative to the unfermented shea nut meal or wheatfeed when fed at 100 g/kg in nutritionally complete diets.

MATERIALS AND METHODS

Culturing of A. niger

The *A. niger* used in this study was a laboratory strain isolate obtained from the University of Wolverhampton, United Kingdom. It was cultured by an agar plating technique using Sabouraud dextrose agar (Oxoid Ltd., Basingstoke, UK) and incubated at 24°C for 7 d. *Aspergillus niger* spores were harvested by tapping the top of the plate when turned upside down. Spore counts were determined using a hematocytometer according to the Fuchs-Rosenthal technique to be approximately 1.6×10^6 spores, which were equivalent to 0.25 g.

Preparation of Fermented Fungus-Treated Shea Nut Meal Sample

An expeller shea nut meal (Shebu-Loders Croklaan Ltd., Savelugu, Ghana) was used for this study (Dei et al., 2008). It was divided into 2 lots. One lot was untreated (no fermentation), and the other lot was fermented using *A. niger*. The fermentation was carried out by mixing the shea nut meal with water in the ratio 1:2 (1 part shea nut meal to 2 parts water) after the spores of *A. niger* (0.25 g/kg) were premixed with the water. The mixture was packed in a plastic container, gently firmed, and sealed with adhesive film before being kept in a room at ambient temperature (24°C). The sample was fermented for 8 d. Although *A. niger* is an aerobic organism, there would be a production phase under micro-aerobic conditions that existed in the closed container (David et al., 2003). The fermented sample was spread on a polythene sheet in a room at 30 to 40°C, dried for 6 d up to about 90% of the dry matter, and ground.

Table 2. Chemical composition of wheatfeed, unfermented, and fermented shea nut meals (g/kg, dry matter basis)

Component	Wheatfeed	Unfermented shea nut meal	Fermented shea nut meal
Crude protein (CP)	176.0	143.6	165.9
Amino acid[1] (g/kg of CP)			
Methionine	13.0	16.4	17.4
Cystine	19.4	15.7	14.2
Methionine + cystine	32.4	32.1	31.6
Lysine	42.1	33.6	31.6
Threonine	29.2	28.4	30.3
Arginine	60.9	68.7	63.2
Isoleucine	35.0	38.8	33.6
Leucine	59.0	62.7	59.4
Valine	53.1	49.3	45.2
Histidine	28.5	23.9	23.2
Phenylalanine	39.5	31.4	30.3
Glycine	50.5	37.3	38.1
Aspartic acid	64.8	83.6	80.7
Serine	43.4	32.1	34.8
Glutamic acid	174.9	133.6	128.4
Alanine	45.4	42.5	41.9
Tyrosine	29.2	31.4	29.0
Proline	45.4	39.6	39.4
Ether extract	37.5	101.4	130.8
Ash	55.1	52.3	57.7
Neutral detergent fiber	341.5	455.7	415.8
Total soluble phenolics	ND[2]	116.7	91.1
Bound plus soluble proanthocyanidins	ND	253.5	166.0
Soluble proanthocyanidins	ND	18.2	13.7
Hydrolysable tannins	ND	3.4	1.6
Gross energy (kcal/kg)	4,641	5,455	5,550

[1]Tryptophan content was not determined.

[2]ND = not determined.

Broiler Chicken Husbandry

A nutritionally complete broiler chicken feed that contained 100 g/kg of diet of each sample was formulated. Although graded dietary levels of the fermented shea nut meal would have been valuable for assessing its nutritional impact, there was a limitation on the number of experimental units available for this experiment. Sufficient replication of experimental treatments to allow economically important differences to be detected was considered to be essential, so a single level (100 g/kg) of each sample was used at an inclusion that would be viable for use in commercial feedmills. Three experimental feeds were therefore prepared (Table 1). The calculated nutrients in the diets were similar to NRC (1994) specifications for broiler chickens. Ross 308 male broiler chicks were reared in a solid-floored pen and fed a crumbled-pellet broiler starter diet (CP = 235 g/kg, ME = 3,026 kcal/kg) for 22 d. At 22 d of age, 96 broilers of similar body weight were caged (0.6 m × 0.6 m × 0.9 m) in groups of 4 and fed 1 of the 3 mealform experimental diets (8 replicates) to 36 d of age. The experiment was arranged as a randomized complete block design with a pen as the experimental unit. The group body weights of the birds were measured at 36 d. Feed and water were provided ad libitum. Feed intake and feed efficiency data were recorded for the grower phase.

Chemical Analysis

Samples of the shea nut meals, wheatfeed, and experimental diets were ground in a laboratory mill fitted with 1-mm mesh screen. Dry matter content of the samples was determined by drying the samples in an oven at 100°C. The N content of the samples was determined by the combustion method (AOAC, 2000; method 968.06) using Leco (FP-528 N; Leco Corp., St. Joseph, MI) with EDTA as a standard. The crude protein content of the samples was calculated from its nitrogen composition (N × 6.25). The gross energy content of samples was determined by adiabatic bomb calorimeter (model 1261, Parr Instrument Co., Moline, IL) with Analar sucrose used as a standard. Crude fat content of samples was determined by the ether extraction method (AOAC, 2000; method 920.39) using a Soxtec system (Foss UK Ltd., Didcot, UK). Acid hydrolysis (4 *M* hydrochloric acid) of the sample was performed before ether extraction to ensure complete recovery of fat. The ash content of samples was determined by combustion in a muffle furnace for 24 h at 500°C. The neutral detergent fiber content was determined by the method of van Soest (1963) using neutral detergent solution.

The amino acid profiles of all samples were determined by the SCIANTEC Services Ltd., Dalton, United Kingdom. The sample was oxidized with hydrogen

Table 3. Chemical composition of experimental diets (g/kg, dry matter basis)

Component	Control (wheatfeed)	Unfermented shea nut meal	Fermented shea nut meal
Crude protein	223.8	218.5	220.4
Amino acid[1]			
Methionine	6.3	5.4	5.8
Cystine	3.2	3.2	3.2
Methionine + cystine	9.1	8.6	9.0
Lysine	13.4	12.5	12.7
Threonine	8.5	8.1	8.2
Arginine	12.9	12.7	12.7
Isoleucine	7.7	8.7	8.7
Leucine	16.1	16.5	16.7
Valine	10.6	10.3	10.6
Histidine	6.3	5.8	5.8
Phenylalanine	10.0	9.5	9.6
Ether extract	70.5	90.0	92.2
Ash	72.8	73.5	75.0
Neutral detergent fiber	109.1	141.7	122.0
Total soluble phenolics	2.1	6.4	4.0
Bound plus soluble proanthocyanidins	0.9	5.8	2.9
Soluble proanthocyanidins	0.0	6.0	2.5
Hydrolysable tannins	0.0	0.3	0.0
Gross energy (kcal/kg)	4,525	4,715	4,683

[1]Tryptophan content was not determined.

peroxide-formic acid-phenol mixture. Excess oxidation reagent was decomposed with sodium metabisulphite. The oxidized sample was hydrolyzed with 6 *M* hydrochloric acid for 24 h. The hydrolysate was adjusted to pH 2.2, centrifuged, and filtered. The amino acids were separated by ion exchange chromatography using a Biochrom 20 analyzer (Biochrom Ltd., Cambridge, UK) and determined by reaction with ninhydrin using photometric detection at 570 nm (440 nm for proline).

Tannin concentrations of the shea nut meal samples and experimental diets were determined at the Institute of Animal Reproduction and Food Research of the Polish Academy of Sciences, Olsztyn, Poland. The content of total soluble phenolics was determined according to the method described by Naczk and Shahidi (1989) using catechin as a standard. The total soluble phenolics include total free phenolic groups of tannins (hydrolysable and proanthocyanidins) and other compounds that are not tannins. The bound plus soluble proanthocyanidins were assayed colorimetrically by the method of Price et al. (1978) using catechin as a standard. The soluble proanthocyanidins were determined by the acid-butanol reagent method of Porter et al. (1986) as cyanidins using an HPLC (Beckman DU 7500, Fullerton, CA) and detected at a 550-nm wavelength. The soluble proanthocyanidins are not bound to protein or fiber. Hydrolysable tannins were determined as gallic acid after enzymatic hydrolysis by tannase according to the procedure of Karamać et al. (2006) using a Shimadzu HPLC system (Shimadzu Corp., Kyoto, Japan) and detected at a 280-nm wavelength.

Statistical Analysis

Type of experimental diet was considered the treatment factor, while each cage of 3 pens was considered as a blocking factor. A randomized block ANOVA was used to compare the treatment means using GENSTAT 10th version (Lawes Agricultural Trust, 2005).

RESULTS

The unfermented and fermented shea nut meal samples had similar proximate nutrient compositions as well as amino acid profiles (Table 2). However, there were reductions in total soluble phenolics, total proanthocyanidins (soluble and insoluble), soluble proanthocyanidins, and hydrolysable tannins in the fermented

Table 4. Growth performance of broiler chickens (22 to 36 d) fed diets containing 100 g/kg of wheatfeed, unfermented, and fermented shea nut meals

Variable	Control (wheatfeed)	Unfermented shea nut meal	Fermented shea nut meal	Analysis of variance, SED (n = 8)	probability[1]
Feed intake (g/bird per day)	142.0[a]	103.2[b]	136.4[a]	3.57	<0.001
Weight gain (g/bird per day)	81.5[a]	45.3[c]	66.8[b]	3.02	<0.001
Gain-to-feed ratio (g/g)	0.57[a]	0.44[c]	0.49[b]	0.014	<0.001
Live weight at 36 d (kg/b)	2.163[a]	1.661[c]	1.958[b]	0.0419	<0.001

[a–c]Means within a row lacking a common superscript differ ($P < 0.05$).

[1]SED = standard error difference. n = number of replicates (experimental unit = 4 birds).

sample (Table 2). The composition of the wheatfeed was typical of published values for the feedstuff (Sarmiento-Franco et al., 2000). Comparatively, there were similarities in the concentrations of most of the nutrients including amino acids of the wheatfeed and shea nut meal samples (Table 2). However, the wheatfeed had lower neutral detergent fiber content than all shea nut meal samples. Also, its crude fat content was lower than that of the shea nut meal samples. The concentrations of tannins in the wheatfeed were not determined because tannins are not considered as anti-nutritive factors.

All the experimental diets appeared to have approximately similar nutrient compositions with the exception of their tannin contents. The wheatfeed-based control diet had very low concentration of total soluble phenolics. The tannin contents in the fermented shea nut meal diet were lower than in the unfermented shea nut meal diet (Table 3).

The birds fed the fermented shea nut meal diet had higher ($P < 0.001$) feed intakes, weight gains, and feed efficiencies than the birds fed the unfermented shea nut meal diet (Table 4). However, the growth performance of birds fed each of the shea nut meal diets was lower ($P < 0.001$) than that of birds fed the wheatfeed-based control diet.

DISCUSSION

The fermentation of shea nut meal using *A. niger* (Table 2) resulted in low reductions in total soluble phenolics (21.9%), bound plus soluble proanthocyanindins (34.5%), and soluble proanthocyanidins (24.7%), but appreciable reduction in hydrolysable tannins (52.9%). *Aspergillus niger* strains are noted particularly for their tannase synthesis ability (Pinto et al., 2001), that degrades hydrolysable tannins into glucose and gallic acid. All the phenolic compounds, including the total soluble phenolics, may pose nutritional risks when fed to broilers. These include astringency that affects palatability, and reduction in nutrient utilization, particularly protein, that negatively influences growth performance. The decrease in feed intake is usually more pronounced with hydrolysable tannins than proanthocyanidins, whereas the major biological effect of soluble proanthocyanidins is on the efficiency of food utilization (Butler and Rogler, 1992). On the other hand, the lower molecular weight polyphenols that are associated with tannins are more readily absorbed from chick diets than tannins, and these may account for major anti-nutritional effects (Butler and Rogler, 1992).

The *A. niger* fermentation of the shea nut meal (Table 4) resulted in an appreciable increase (24%) in feed intake compared with the unfermented meal. The feed intake of the birds fed the fermented shea nut meal was similar to that of the control diet (wheatfeed-based). This was a clear indication of reduction in the adverse effect of anti-nutritive factors such as tannins in shea nut meal on feed intake of broilers (Annongu et al., 1996). This could be attributed to the lower concentrations of tannins in the fermented meal diet than in the unfermented meal diet (Table 3).

The poor growth performance of birds fed unfermented shea nut meal diet (Table 4) was expected as has been shown in previous studies (Annongu et al., 1996; Olorede et al., 1999). This could be attributed to the relatively high concentrations of tannins in the unfermented shea nut meal diet (Table 3). Broilers fed diet containing the shea nut meal fermented with *A. niger* had an improved growth performance that was 82% of the control birds. Their feed efficiency was 86% of the control. The lowering of growth performance of birds fed the fermented shea nut meal could be due to the effects of residual tannins (Annongu et al., 1996). Although the levels of tannins in the fermented shea nut meal diet were considerably low relative to the unfermented shea nut meal diet (Table 3), they were high enough to depress growth performance of the birds. Many studies have shown that high dietary tannins result in reduced weight gains and poor feed efficiencies in birds (Armstrong et al., 1974; Ahmed et al., 1991; Treviño et al., 1992; Iji et al., 2004). This is due to a pronounced negative effect on protein digestibility (Smulikowska et al., 2001). Nelson et al. (1975) observed that when hybrid sorghums of varying tannin content were fed to chicks, there was a significant ($P < 0.05$) negative correlation between tannin content and metabolisable energy ($r = -0.62$), dry matter digestion ($r = -0.63$), and bioavailability of amino acids ($r = -0.82$). Tannins have been shown to inhibit in vivo activities of trypsin and α-amylase, but increased lipase activity (Longstaff and McNab, 1991), which may affect digestion of proteins and starch. The present study suggests that attempts should be made to further reduce the tannins in the fermented shea nut meal by improving the efficacy of the fermentation process.

Wheat by-products such as wheatfeed and wheatbran are often imported and used extensively in the West African sub-region in poultry diets. Although the fermented shea nut meal in its present form did not match the nutritive value of the wheatfeed used in this experiment, it, however, demonstrated its usefulness as a potential feed ingredient. Also, there is room for refining the fermentation process to further enhance the quality of shea nut meal. This feedstuff at present has no economic value; therefore, further improvement of the fungal fermentation method would make this home-produced material an alternative feed resource for feeding poultry.

It was evident from this study that a solid state fermentation of shea nut meal using *A. niger* had improved substantially (47.5%) the growth performance of broiler chickens, even though it did not match that of wheatfeed (79.9%) with reference to the unfermented shea nut meal. Therefore, this method of improving the nutritive value of shea nut meal requires refinement for it to become more useful in poultry rations.

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