EFFECT OF DIETARY SHEA NUT CAKE AND RICE HUSK FERMENTED WITH *RHIZOPUS OLIGOSPORUS* ON GROWTH PERFORMANCE AND SERUM ELECTROLYTES OF WEANER PIGS

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Abstract

The high cost of conventional feed ingredients has led to the search for alternative feed sources for pig production. This study investigated the effect of dietary fermented shea nut cake (FSNC) and fermented rice husk (FRH) with *Rhizopus oligosporus* on the growth performance and serum electrolytes of weaner pigs. Solid-state fermentation using *R. oligosporus* CBS 534 was carried out on rice husk and shea nut cake for 10 days. 36 Duroc x Large White weaned pigs (18 males and 18 females), with an average age and weight of 8 weeks and 10 ± 2 kg were randomly assigned to dietary treatments in a 3 x 3 x 2 factorial arrangement. The factors included FSNC (0, 5, or 10%) x FRH (0, 5, or 10%) in diets x sex (male or female) in a completely randomised design with 18 treatments and 2 replicates over a period of 5 weeks. Results showed that the feed intake and weight gain were not significantly ($P > 0.05$) influenced by dietary FSNC and FRH. Feed conversion ratio was significantly ($P < 0.05$) affected by dietary FSNC, FRH and sex. The serum electrolyte concentrations showed that pigs fed FSNC had a significant ($P < 0.05$) effect on HCO_3, FRH significantly ($P < 0.05$) influenced Na and Cl, and sex had a significant ($P < 0.05$) effect on Na. Pigs fed FSNC showed reduced HCO_3 concentration with increasing FSNC inclusion. The findings suggest that up to 10% FSNC and FRH could be incorporated into weaner pig diets without adverse effect on growth performance and electrolyte balance.

Keywords: shea nut cake, rice husk, *Rhizopus oligosporus*, weaned pigs, serum electrolytes

Introduction

A major problem for the expansion of the pig industry is the short supply of conventional feed ingredients, which are expensive, compelling the use of alternative feedstuffs (Atteh and Apata, 2024). Shea nut cake (SNC) and rice husk (RH) are major agro-industrial by-products that are readily available in large quantities at a low cost. RH consists of 39-42% crude fibre, 2.9-3.6% crude protein, 8-12% ether extract, and 15-22% ash and is limited in usage for monogastric animals due to its high fibre content (Aderolu et al., 2007). SNC is an agroforestry by-product obtained from the processing of nuts of the shea butter tree (*Vitellaria paradoxa*) that explored its fat with no economic value (Dei et al., 2008). Abdul-Mumeen et al. (2013) reported that its overall nutritional value contained 13.03, 23.38, 4.25, 8.71, 59.37% and 4485.86 kcal ME kg^{-1} of crude protein, crude fat, ash, crude fibre, carbohydrates, and metabolisable energy, respectively. SNC has been used for dietary maize replacement in poultry rations (Aguihe et al., 2017). However, the presence of anti-nutritional factors like saponins, alkaloids, phytic acids and tannins resulted in its poor digestibility possibly (Annongu et al., 1996; Oddoye et al., 2012). RH included at 10-20% replacement of maize led to decreased growth performance and increased the rate of mortality among weaner pigs (Ejiofor et al., 2024); high fibre in RH resulted in poor digestibility in pigs (Omotoso and Arilekolasi, 2019).

Fermentation is a distinctive process with great potential for re-recycling some agro-industrial by-products into useful animal feeds. The process does not require the use of chemicals and is easy to manage in on-farm settings

and/or on an industrial scale. Fermentation processes using *Rhizopus* species have been used to improve the nutritive value of some feeds, such as soybean pulp for pigs (Vong et al., 2018). Kehinde et al. (2020) observed that solid-state fermentation improved the nutritive value of SNC. Mafimidiwo (2022) reported that solid-state fermented rice husk could replace up to 75% wheat offal in the diet of growing pigs.

The blood biochemical profile, including the electrolytes, is an index for the health of livestock. The normal blood biochemical indices have been found to be influenced by sex, diet, and feeding method (Adeyemo et al., 2018). Electrolytes are important components in livestock, such as Na, K, and Cl concentrations in blood, which reflect the changes of electrolyte balance, and their concentrations within the normal range are important in maintaining physiological function (Mushtaq et al., 2005; Gonzalez-Esquerra et al., 2006). Potassium plays a vital role in regulating the osmotic pressure of cells, as well as maintaining the acid–base and electrolyte balances of body fluids (Wei et al., 2023). RH is acknowledged for its high fibre and low protein content, along with antinutritional factors such as tannins, saponins, alkaloids, and phytates in SNC, which hinder its application as feed for pig production. The necessity of enhancing the nutritional value and minimising the harmful constituents in these materials has led to the cost-effective application of solid-state fermentation (SSF), a fungal biotechnology that transforms these wastes into beneficial feed for pigs. This study investigated the growth performance and blood electrolytes of weaned pigs fed shea nut cake and rice husk fermented with *Rhizopus oligosporus*

Materials and methods

Experimental site

The research was carried out at the piggery unit of the Teaching and Research Farm, Kwara State University, Malete, Nigeria, which is located in the Southern Guinea Savanna Vegetation Ecological zone on longitude 11^0 71''N and latitude 40^0'44''E 671m above sea level.

Preparation of fermented rice husk and shea nut cake

RH and SNC were obtained from Ilesha-Baruba, Baruteen Local Government Area, Kwara State. Samples (500 kg) each of the RH and SNC were milled and then subjected separately in batches to the SSF process. The fungus used for fermentation was *Rhizopus oligosporus* CBS 534, and the stock was obtained from the Microbiology Laboratory, Kwara State University, Malete.

In preparing suspension, *R. oligosporus* CBS 534 was cultivated on potato dextrose agar (PDA) at 28±1°C for 7 days, after which it was subcultured to obtain a pure suspension containing 5 x 104 spores ml^{-1} following the procedure outlined by Hesseltine (1983). A batch (1 kg) each of the RH and SNC were separately autoclaved at 121°C for 15 min. Autoclaved meals were cooled to room temperature for 1 h, hydrated with distilled water at a ratio of 1:1.5 (w/v), and inoculated with 20 ml of spore suspension of *R. oligosporus* CBS 534 per kg of wet meal. The meal was thoroughly mixed and fermented in an airtight plastic bucket for 10 days. The fresh fermented samples were then re-autoclaved at 121°C for 15 min to destroy R. oligosporus and sun-dried for 5 days. The dried products were ground in the mill, packed in a polythene sack, sealed and stored at ambient temperature until mixed into diets described below.

Pigs, diets, management and experimental design.

Thirty-six Duroc x Large White weaned pigs (18 males and 18 females) with an average age and weight of 8 weeks and 10 ± 2 kg were randomly assigned to dietary treatments in a 3 x 3 x 2 factorial arrangement. The factors

included FSNC (0, 5, or 10%) x FRH (0, 5, or 10%) in diets x sex (male or female) in a completely randomised design with 18 treatments and 2 replicates over a period of 35 days. The dietary formulation is presented in Table 1.

The weaner pigs' initial weight was recorded at the beginning of the feeding trial and thereafter weekly using a weighing balance. The final weight was also recorded at the end of the feeding trial. The weight gain was computed as the difference between the final weight and the initial weight measured. The mean weight gain was then computed. The feed intake per pig per day was estimated by subtracting the feed offered from the feed left over.

Electrolyte Determination

At the end of the experimental period, one weaner pig was randomly selected from each replicate, and blood samples were collected into clean, dry tubes for serum electrolyte analysis. Serum samples were separated from whole blood by centrifugation at 3000 rpm for 10 min and stored at -4℃. All the parameters viz. sodium (Na), potassium (K), chloride (Cl), and bicarbonate (HCO_3) were analysed by using the Acculyte-3P electrolyte analyser (Shakkarpude et al., 2023).

Statistical analysis

Data were analysed using a three-way analysis of variance (ANOVA) and SPSS version 25. Duncan's multiple range test was used to determine significant differences at the 0.05

Table 1 Formulation (gkg^{-1}) of weaner pig diets

	Diets								
	1	2	3	4	5	6	7	8	9
Ingredients									
Maize	430.0	430.0	430.0	360.0	350.0	330.0	330.0	330.0	330.0
Fermented shea nut cake (FSNC)	0	50.0	100.0	0	50.0	100.0	0	50.0	100.0
Fermented rice husk (FRH)	0	0	0	50.0	50.0	50.0	100.0	100.0	100.0
Groundnut cake	354.0	255.0	95.0	330.0	250.0	50.0	285.0	200.0	50.0
Soybean meal	40.0	70.0	150.0	63.0	65.0	195.0	100.0	115.0	175.0
Cassava powder meal	30.0	20.0	50.0	30.0	30.0	30.0	40.0	30.0	50.0
Maize offal	97.0	100.0	50.0	100.0	100.0	100.0	50.0	50.0	20.0
Vegetable oil	3.0	10.0	30.0	20.0	30.0	50.0	40.0	50.0	60.0
Blood meal	1.0	20.0	50.0	2.0	30.0	50.0	10.0	30.0	70.0

Limestone	25.0	25.0	25.0	25.0	25.0	25.0	25.0	25.0	25.0
Bone meal	10.0	10.0	10.0	10.0	10.0	10.0	10.0	10.0	10.0
	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5
Vitamin/mineral premix[a]									
Lysine	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5
Methionine	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5
Salt	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5
Nutrient content[b] (gkg^{-1})									
Crude protein	209.7	208.1	206.9	206.7	203.7	206.7	207.2	206.5	207.4
Crude fibre	41.6	62.1	57.1	38.1	64.8	55.5	42.2	64.8	55.9
Ether extract	55.8	84.5	76.4	62.4	86.3	71.0	54.9	83.5	68.4
Calcium	13.9	14.1	13.7	14.1	13.8	13.8	13.6	14.1	13.9
Phosphorus	6.3	4.5	5.2	5.1	5.2	5.7	5.9	5.6	6.1
Methionine	5.4	5.0	5.0	5.1	5.0	5.1	5.2	5.2	5.3
Lysine	13.2	14.3	13.7	13.6	13.3	13.3	13.1	13.5	13.2
Metabolizable energy (MJ kg^{-1})	12.0	11.7	11.8	11.4	11.8	11.9	11.9	11.9	11.8

[a] Provided 1.5 mg retinol, 25 mg cholecalciferol, 16 mg α-tocopherol, 1mg menadione, 0.8 mg thiamine, 2.4 mg riboflavin, 14 mg nicotinic acid, 4 mg calcium D-pantothenate, 1.4 mg pyridoxine, 10 mg cyanocobalamin, 0.4 mg folic acid, 0.02 mg biotin, 120 mg choline chloride, 0.62 mg zinc bacitracin, 36 mg avaten, 40 mg Mn, 18 mg Zn, 0.8 mg Cu, 0.09 mg Co, 20 mg Fe, and 0.04 mg Se kg−1 diet.

[b] Determined values except for metabolisable energy (ME), lysine, methionine, calcium and phosphorus which were calculated from the published (NRC, 2024) compositions of the ingredients used and the ME of FSNC and FRH was estimated from their analytical data (Pauzenga, 1985).

Results and Discussion

The effect of dietary *Rhizopus oligosporus* FSNC and FRH at varying inclusion rates on the weaner pigs' growth performance is shown in Table 2. The FSNC and FRH did not significantly affect the feed intake and weight gain ($P > 0.05$). However, there was a trend of increase in the feed intake and weight gain with increasing dietary FSNC. Feed conversion ratio (FCR) was significantly affected by dietary FSNC and FRH and sex. There was no interaction

between FSNC x FRH and FSNC x Sex on feed intake and weight gain, but there was an interaction among FSNC x FRH x Sex on FCR. The slight increase in the observed feed intake and weight gain in the weaner pigs with increasing FSNC inclusion compared to those fed the control diet could be ascribed to the fermentation process which improved the nutritive value of SNC. This is in agreement with Dei et al. (2008), who reported improved nutritive value of SNC for poultry, consequent upon reduction in the adverse effect of anti-nutritive factors such as tannins and saponins in SNC (Annongu et al., 1996). FCR improved significantly in female weaner pigs fed both FSNC and FRH compared to their male counterparts. This could be potentially related to hormonal or metabolic differences between males and females. Weaner pigs fed 10% FRH had an increase in FCR values, indicating inefficient conversion of the feed material at that level. This could be partly attributed to decreased nutrient digestibility and lower energy utilisation linked to high fibre in FRH. This corroborates earlier reports of Omotoso and Arilekolasi (2019) and Hamid et al. (2020).

Table 2: The effect of *Rhizopus oligosporus* fermented sheanut cake and rice husk at different inclusion rate on the growth performance of weaner pigs

Treatment	Initial body weight (kg)	Final body weight (kg)	Feed intake (kg per pig day^{-1})	Weight gain (kg per pig day^{-1})	FCR
FSNC (%)					
0	10.42 ± 0.41	23.20 ± 0.81	1.20	0.37	3.24^{b}
5	10.12 ± 0.41	23.88 ± 0.67	1.28	0.39	3.28^{b}
10	10.57 ± 0.47	24.63 ± 0.87	1.33	0.40	3.33^{b}
FRH (%)					
0	10.38 ± 0.32	24.13 ± 0.43	1.27	0.39	3.25^{b}
5	9.68 ± 0.31	23.03 ± 0.73	1.25	0.38	3.28^{b}
10	11.03 ± 0.53	24.55 ± 1.06	1.30	0.38	3.42^{c}
Sex					
Female	10.23 ± 0.33	23.46 ± 0.56	1.20	0.38	3.15^{a}
Male	10.50 ± 0.37	24.36 ± 0.71	1.38	0.39	3.54^{d}
Interaction					
FSNC x FRH	ns	Ns	ns	ns	ns
FSNC x Sex	ns	Ns	ns	ns	ns
FRH x Sex	ns	Ns	ns	ns	ns
FSNC x FRH x Sex	ns	Ns	ns	ns	*

Means values with superscript [abcd] within the same factor and within the same column are significantly different ($p < 0.05$), FCR= Feed Conversion Ratio, FSNC = fermented shea nut cake, FRH = fermented rice husk, ns = not significant. *= significant

Table 3: Effect of dietary shea nut cake and rice husk fermented with *Rhizopus oligosporus* on serum electrolytes of weaner pigs

Treatment	K^+ (mmol/L)	Na^+ (mmol/L)	Cl^- (mmol/L)	HCO_3^- (mmol/L)
FSNC (%)				
0	5.53	139.17	97.33	22.58[a]
5	5.21	140.17	97.92	21.42[ab]
10	5.84	139.33	98.42	19.08[b]
FRH (%)				
0	5.31	139.58[ab]	97.58[b]	21.58
5	5.78	141.17[a]	99.25[a]	20.92
10	5.49	137.92[b]	96.83[b]	20.58
Sex				
Female	5.46	138.72[b]	97.89	21.00
Male	5.60	140.39[a]	97.89	21.06
Interactions				
FSNC x FRH	ns	ns	Ns	*
FSNC x Sex	ns	ns	Ns	*
FRH x Sex	ns	ns	ns	ns
FSNC x FRH x Sex	ns	ns	ns	ns

Means values with superscript [ab] within the same factor and within the same column are significantly different ($p < 0.05$), FRH= Fermented Rice Husk, FSNC = Fermented Shea nut cake, K^+ = Potassium, Na^+ = Sodium, Cl^- = Chloride, HCO_3^- = Bicarbonate

The serum electrolyte concentrations of weaner pigs fed FSNC and FRH are presented in Table 3. The results revealed that pigs fed FSNC had a significant ($P<0.05$) effect on bicarbonate (HCO_3), the FRH significantly ($P<0.05$) influenced Na and Cl, and sex had a significant ($P<0.05$) effect on Na. Pigs fed FSNC showed reduced HCO_3 concentration with increasing FSNC inclusion. There was no main effect of the three factors or interaction on K concentration in weaner pigs. In the present study, the serum values obtained for most of the electrolytes are within the normal range for pigs (Yeom et al., 2012), which are important in maintaining physiological functions (Mushtaq et al., 2005; Gonzalez-Esquerra et al., 2006). The serum electrolyte concentrations of weaner pigs fed FSNC and FRH revealed significant effects on some electrolytes. However, the decrease in HCO_3 in weaner pigs with increasing FSNC inclusion could affect the acid-base balance in the pigs. Similar findings have been reported by Lin et al. (2024), who found decreased pH and HCO_3 in piglet blood fed low-protein diets. Generally, HCO_3 are waste products of mitochondrial energy production that play a role in pH buffering throughout the body. As a result, they neutralise bases (urea from the catabolism of proteins) and acid produced in the blood through other metabolic processes (Kumari et al. 2014).

The Na and Cl balance is one part of the internal environment homeostasis that is necessary to maintain the health of the small intestine of pigs (Zhang et al., 2022). In the present investigation, the significant influence of FRH on Na and Cl concentrations observed suggests that FRH may have a specific effect on the regulation of these electrolytes.

The sex-related difference in Na concentration warrants further investigation, potentially related to hormonal or metabolic differences between males and females. In conclusion, the findings demonstrate that up to 10% FSNC and FRH could be incorporated into weaner pig diets without adverse effects on growth performance and electrolyte balance.

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