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Effect of dietary probiotic supplementation on carcass traits and haematological responses of broiler chickens fed shea butter cake based diets

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ABSTRACT. A 42-day study was conducted to investigate probiotic supplementation on shea butter cake (SBC) based diets of broilers on the carcass traits and haematological indices. A total of 280, 1-day old *Arbor acres* strain broiler chicks were divided into 7 dietary treatments with 4 replicates of 10 birds each in a completely randomized design. Diet 1 (control diet) has no SBC and probiotic supplementation while remaining 6 diets contained 5, 10, and 15% levels of SBC each with (+) or without (-) probiotic supplementation. The results showed that carcass parameters and prime cuts weights increased ($p < 0.05$) in birds fed probiotic-SBC diets compared to control diets. Abdominal fat content was lower ($p < 0.05$) in birds fed probiotic-SBC diets than those on control and non-probiotic SBC diets. Differences in relative organ weights among all treatments were non-significant ($p > 0.05$) except for liver and bursa weight. Packed cell volume and hemoglobin contents were higher ($p < 0.05$) in birds fed probiotic-SBC diets than the group without probiotic supplementation compared to those on control diet. In conclusion, supplementation of multi-strain probiotic improved carcass traits, prime cut-parts and relative organ weights without any adverse effect on the blood parameters up to 15% inclusion level.

Keywords: agro by-product, feed additive, carcass characteristics, blood, chicken.

Efeito da suplementação dietética de probiótico sobre as características de carcaça e respostas hematológicas de frangos de corte alimentados com dietas à base de bolo de manteiga de carité

RESUMO. Este estudo foi realizado com o objetivo de avaliar a suplementação de probiótico em dietas à base de bolo de manteiga de carité - *sheabuttercake* (SBC) sobre as características de carcaça e índices hematológicos de frangos de corte. Foram utilizados 280 frangos de corte da linhagem *Arbor acres*, distribuídos em um delineamento inteiramente casualizado com sete tratamentos, quatro repetições e dez aves por repetição. Foi formulada uma dieta sem SBC nem probiótico e utilizada como dieta controle. As demais dietas experimentais continham 5, 10 e 15% de SBC e com (+) ou sem (-) a suplementação de probiótico. Os parâmetros de carcaça e o peso dos cortes nobres aumentaram ($p < 0.05$) nas aves que receberam as dietas com (+) SBC probiótico quando comparadas com o controle. O teor de gordura abdominal foi menor ($p < 0.05$) nas aves que receberam as dietas com (+) SBC-probiótico do que nas aves alimentas com a dieta controle ou com (-) SBC sem probiótico. Não foram observadas diferenças ($p < 0.05$) para o peso relativo dos órgãos, exceto para o peso do fígado e da bolsa cloacal. A quantidade de eritrócitos e hemoglobina foram maiores ($p < 0.05$) nas aves que receberam as dietas com (+) SBC-probiótico quando comparadas com o tratamento controle. A suplementação com probiótico em até 15% melhorou as características de carcaça e o peso relativo dos cortes nobres e órgãos sem nenhum efeito adverso nos parâmetros sanguíneos.

Palavras-chave: aditivo alimentar, características de carcaça, frango de corte, sangue, subproduto.

Introduction

The rapid growth of human population has intensified the competition between humans and livestock for grains such as maize which is the major source of energy in poultry feeds (Annongu, Ogundu, Joseph, & Awopetu, 2006; Mohammed & Agwunobi, 2009). Consequently, high cost of feeding poultry has necessitated the need to look for alternative energy feed source for poultry in order to reduce cost and limit dependence on maize (Esonu et al., 2006; Wafar, Ademua, Kirfib,

& Shehu, 2016). Shea butter cake (SBC) is an agroforestry by-product obtained from the processing of the nuts of the shea butter tree (*Vitellariaparadoxa*, Gaertn.) for fat with no economic value and its increasing output of late has become an environmental issue (Dei, Rose, Mackenzie, & Amarowicz, 2008; Zanu, Adom, & Appiah-Adu, 2012). Abdul-Mumeen, Zakpaa, and Mills-Robertson (2013) investigated SBC for its proximate quality and reported its overall nutritional value to be high; containing 13.03, 23.38, 4.25, 8.71, 59.37% and 4485.86 kcal ME kg^{-1} of crude protein, crude fat, ash, crude fiber, carbohydrates and metabolizable energy respectively. However, the major nutritional setback of SBC utilization for chicken is poor digestibility possibly due to the presence of anti-nutritional factors like saponins and most particularly tannins (Annongu, Termeulen, Atteh, & Apata, 1996; Agbo & Prah, 2014).

With the advent of biotechnology, the use of dietary certain feed additives such as probiotics are opportunities for economic and efficient utilization of poor quality feed or agro-industrial residues (Kalavathy, Abdullah, Jalaludin, & Ho, 2003; Ezema, Ihedioha, Ihedioha, Okorie-kanu, & Kamalu, 2012; Nawaz, Irshad, Mubarak, & Ahsan-Ul-Haq, 2016). Probiotics are single or mixed cultures of live microbial feed supplements that beneficially affect the host animal by improving its microbial intestinal balance (Fuller, 1989). Dietary probiotics are products have shown to create favorable conditions in the animal's intestine for efficient digestion and absorption of feed as well as improve their health status (Li et al., 2008, Owosibo, Odetola, Odunsi, Adejinmi, & Lawrence-Azua, 2013). Therefore, this study was designed to evaluate the response of broiler chickens to diets containing graded levels of Shea butter cake (SBC) meal with or without multi-strain probiotic supplementation on carcass traits, relative organ weights and haematological indices.

Material and methods

Experimental site and preparation of test ingredient

This study was conducted at the Poultry Unit of Teaching and Research Farm, Federal College of Wildlife Management, New Bussa, Niger State, Nigeria. The SBC used for this study was obtained fresh from the local shea butter processing factories in Karabande, Borgu Local Government Area of Niger state, Nigeria. The fresh SBC was properly sun-dried for 5 days, milled using a hammer mill before incorporation into experimental diets.

Experimental birds and management

Use and care of birds and procedures adopted on this study were approved by the Animal Ethics Committee of the Federal College of Wildlife Management, New bussa before the commencement of the experiment. A total of 280 unsexed day old *Arbor acres* broilers were weighed and allocated to 7 dietary treatments with 4 replications of 10 birds each (5 males and 5 females) in a completely randomized design. The birds were raised in a standard tropical deep litter poultry facility using wood shavings as litter material. The birds were fed*ad libitum* and clean water was provided regularly for a period of 42 days.

Experimental diets

Seven isonitrogenous and isocaloric diets were formulated according to the nutritional recommendations of NRC (1994), such that diet 1 (control diet) has no SBC and probiotic supplementation while remaining 6 diets contained 5, 10, and 15% levels of SBC each with (+) or without (-) probiotic supplementation at both starter and grower phase (Table 1 and 2). A multi-strain commercial probiotic preparation (Biovet- YC®) in a powder form consisting of *Lactobacillus acidophilus, Saccharomyces cerevisiaea* and *Saccharomyces boulardi, was* used at an inclusion rate 0.5 g kg^{-1} according to the manufacturer's recommendation.

Carcass and organ evaluation

At the conclusion of the experiment, two birds whose weights were close to the mean replicate weight were selected from each replicate, fasted overnight, weighed, slaughtered, scalded in 65°C water for 15 seconds, manually defeathered, and eviscerated. Thereafter, the carcasses were dissected; the prime cut-up parts were removed and weighed together with the visceral organs and were expressed as percentage live weight.

Haematological evaluation

3 mL of blood for haematological analysis were collected through jugular veins of two birds of similar weights from each replicate into sterilized glass tubes containing EDTA. Haematological indicators such as white blood cell counts (WBC), red blood cell counts (RBC), packed cell volume (PCV) and haemoglobin were determine using Wintrobe'smicrohaematocrit, improved Neubauer haemocytometer and cynomethaemoglobin method respectively. Mean corpuscular haemoglobin (MCH) and Mean corpuscular haemoglobin concentartio (MCHC) were computed according to Jain (1986).

Table 1. Ingredients composition of experimental broiler starter diets.

Ingredient	0% SBC	5%- SBC	5%+ SBC	10%- SBC	10%+ SBC	15%- SBC	15%+ SBC
Maize	61.50	56.50	56.00	51.50	51.00	46.50	46.00
SBC[1]	0.00	5.00	5.00	10.00	10.00	15.00	15.00
Probiotic	-	-	0.50	-	0.50	-	0.50
Soybean 44%	32.00	32.00	32.00	32.00	32.00	32.00	32.00
Fish meal	3.00	3.00	3.00	3.00	3.00	3.00	3.00
DCP[2]	2.00	2.00	2.00	2.00	2.00	2.00	2.00
Oyster shell	0.50	0.50	0.50	0.50	0.50	0.50	0.50
Premix[3]	0.40	0.40	0.40	0.40	0.40	0.40	0.40
Salt	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Lysine, 78.5%	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Methionine, 99%	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Total	100	100	100	100	100	100	100
Nutrient Analysis							
Crude Protein, %	22.72	22.94	22.93	22.98	22.96	22.96	22.97
Crude Fiber, %	3.82	4.12	4.11	4.23	4.22	4.45	4.42
Ether Extract, %	5.52	6.34	6.33	6.86	6.84	7.12	7.10
Calcium, %	1.22	1.24	1.24	1.25	1.24	1.26	1.25
Phosphorus, %	0.71	0.73	0.72	0.75	0.74	0.78	0.77
Lysine, %`	1.24	1.22	1.22	1.21	1.21	1.20	1.21
Methionine, %	0.53	0.51	0.51	0.49	0.48	0.46	0.45
ME, kcal kg^{-1}	3000.14	3031.39	3013.39	3013.28	3013.28	3050.71	3050.71

[1]SBC: Shea butter cake; [2]DCP: Di Calcium Phosphate. [3]Vitamin mineral premix provided (per kg of diet): Vitamin A, 5000 I.U., Vitamin D_3 1000,000 I.U., Vitamin E 15,000 mg; Vitamin K_3, 100 mg; Vitamin B_1, 1,200 mg; Vitamin B_2, 2,400 mg Biotin, 32 mg; Vitamin B_{12}, 10 mg; Folic acid, 400 mg; Choline chloride, 120,000 mg; Manganese, 40,000 mg; Iron, 20,000 mg; Zinc 18,000 mg; Copper, 800 mg; Iodine, 620 mg; Cobalt, 100 mg; Selenium 40 mg.

Table 2. Ingredients and nutrient composition of experimental finisher broilers diets.

Ingredients	0% SBC	5%- SBC	5%+ SBC	10% -SBC	10%+ SBC	15%- SBC	15%+ SBC
Maize	70.30	65.30	64.80	60.30	59.80	55.30	54.80
SBC[1]	0.00	5.00	5.00	10.00	10.00	15.00	15.00
Probiotic	-	-	0.50	-	0.50	-	0.50
Soybean 44%	23.00	23.00	23.00	23.00	23.00	23.00	23.00
Fish meal	3.00	3.00	3.00	3.00	3.00	3.00	3.00
DCP[2]	2.00	2.00	2.00	2.00	2.00	2.00	2.00
Oyster shell	1.00	1.00	1.00	1.00	1.00	1.00	1.00
Premix[3]	0.30	0.30	0.30	0.30	0.30	0.30	0.30
Lysine, 78.5%	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Methionine, 99%	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Total	100	100	100	100	100	100	100
Nutrient Analysis							
Crude Protein, %	20.22	20.30	20.98	20.20	20.23	20.43	20.42
Crude fiber %	5.42	5.55	5.54	6.13	6.11	6.43	6.41
Ether Extract, %	5.88	6.34	6.33	7.12	7.11	7.65	7.61
Calcium, %	1.22	1.21	1.21	1.20	1.20	1.21	1.20
Phosphorus, %	0.73	0.75	0.74	0.74	0.73	0.73	0.72
Lysine, %	1.12	1.11	1.10	1.09	1.08	.1.09	1.08
ME, kcal kg^{-1}	3069.45	3054.36	3039.87	3085.17	3037.51	3099.92	3064.15

[1]SBC: Shea butter cake; [2]DCP: Di Calcium Phosphate. [3]Vitamin mineral premix provided (per kg of diet): Vitamin A, 5000 I.U., Vitamin D_3 1000,000 I.U., Vitamin E 15,000 mg; Vitamin K_3, 100 mg; Vitamin B_1, 1,200 mg; Vitamin B_2, 2,400 mg Biotin, 32 mg; Vitamin B_{12}, 10 mg; Folic acid, 400 mg; Choline chloride, 120,000 mg; Manganese, 40,000 mg; Iron, 20,000 mg; Zinc 18,000 mg; Copper, 800 mg; Iodine, 620 mg; Cobalt, 100 mg; Selenium 40 mg.

Statistical analysis

Pen means were used as the experimental unit for all analyses. All data collected were subjected to the analysis of variance as a completely randomized design using General Linear Model of SAS software (2006). The means of treatments showing significant difference were separated using Tukey test. All statements of significance were based on the 5% level of probability.

Results and discussion

Table 3 shows the result of the carcass traits and prime-cut weights of the broilers fed SBC based diets with or without probiotic supplementation. The final live weight, carcass weight and carcass yield were higher ($p < 0.05$) in the birds fed control and probiotic SBC diets than those without probiotic addition as the level of SBC increased in the diet. The higher carcass weight and carcass yield observed in the probiotic supplemented groups is attributed to higher body weight in the birds of these groups. These improvements could be as a result of the cumulative effect of the probiotic microbe's action including increased digestive enzyme activity, maintenance of beneficial microbial population and neutralizing the effect of feed toxins in the gut environment for improved digestion and nutrient utilization (Tellez et al., 2001; Shim et al., 2010; Chen, Wang, Yan, & Huang, 2013). These findings are compatible with the reports of previous workers (Adejumo, Onifade, & Afonja, 2004; Nawaz et al., 2016) who observed higher body weight and better carcass yield in birds fed diet containing *Lactobacillus* and *Saccharomyces* species. Birds on non-probiotic SBC diet recorded poor carcass yield at higher levels of SBC and this could be attributed to impairment in utilization of nutrients due to the relatively high concentration of residual tannin in SBC as the level increases in the diets (Annongu et al., 1996, Iji, Khumalo, Slippers, & Gous, 2004). This is as a result of pronounced negative effect of anti-nutritional factors on protein digestibility (Smulikowska et al., 2001), thus leading to poor carcass yield formation. Higher ($p < 0.05$) breast, thigh, drumstick, wing and back meat yield were observed in the birds fed probiotic diets similar to the control group. This is in accordance with findings of Fathi, AL-Mansoor, AL-Homidan, AL-Khalaf, and AL-Damegh (2012) and Mutassim (2013) who reported that supplementation of probiotic increased the carcass prime-cuts yield in broilers. The possible mechanism through which probiotic achieved this improvements are demonstrated by its ability to enhance synthesis and bioavailability of nutrients (Koop-Hoolihan, 2001), accompanying with positive effects on intestine activity and increasing digestive enzymes (Endens, 2003) thereby promoting growth of muscle tissues. Lower ($p < 0.05$) abdominal fat content was recordedin birds fedprobiotic supplemented-SBC diets than those with probiotic addition. This agrees with the findings of previous researchers (Mehr, Shargh, Dastar, Hassani, & Akbari, 2007; Mohammadreza, Alireza, Leila, & Andrés, 2016), who reported that certain microbiota present in gastro-intestinal tract of a bird impaired the absorption of cholesterol and bile acid. Besides, the probiotic microorganisms are able to hydrolyze bile salts or decrease the activity of acetyl-CoA carboxylase, the rate limiting enzyme in fatty acid synthesis, thus causing a lower absorption and deposition of fat content around the abdomen (Kalavathy et al., 2003; Mansoub, 2010).

The effects of SBC based diet supplementation with or without multi-strain probiotics on relative organ weights are presented in Table 4. Significant ($p < 0.05$) differences were observed in the weights of the liver and bursa while all other parameters were not affected ($p > 0.05$) by the dietary treatments. The higher ($p < 0.05$) liver weight observed in birds fed diets containing SBC without probiotic was anticipated due the presence of residual anti-nutritional factors especially tannin. This result is similar with the findings of previous researchers (Akinmutimi & Essien, 2011; Donlaporn & Worapot, 2011) who reported that presence of a toxic factor causes abnormalities on weight of liver and this could be as a result of increase in metabolic rate of the organ in an attempt to reduce the effect of the toxic elements. Moreover, the supplementation of multi-strain probiotic increased ($p < 0.05$) relative weight of bursa of the birds compared to treatments without probiotic. This is in agreement with the findings of Hossain, Begum, and Kim (2015) who reported anincrease in the bursa weight of broilers fed tri-strain probiotics compared to the control group. Besides, Willis, Isikhuemhen, and Ibrahim (2007) also suggested that the bursa was the primary lymphoid organ in broilers and concluded that probiotics could increase the relative weight of bursa to body weight. In contrast, previous authors have reported a non-significant effect of probiotic supplementation on the relative organ weights including liver and bursa of Fabricious, as a proportion of body weight (Naseem, Rahman, Shafee, Sheikh, & Khan, 2012; Nawaz et al., 2016). The observed inconsistency might be attributed to the strains of probiotic, technique of preparation, administration dosage, diet composition and hygienic status (Zhang, Zhou, Ao, & Kim, 2012).

The heamatological evaluation revealed that significant ($p < 0.05$) changes were only observed in packed cell volume (PCV) and haemoglobin concentration of the birds among the treatments (Table 5). The lower ($p < 0.05$) haemoglobin concentration associated with the birds fed non-probiotic diets may be due to the higher concentration of tannin as the inclusion level of SBC increased in the diets. This is in agreement with the reports of Oddoye, Alemawor, Agyente-Badu, and Dzogbefia (2012) and Orogun, Oniye, and Olugbemi 2015). However, multi-strain probiotic supplementation to SBC based dietshowed an increased ($p < 0.05$) PCV and haemoglobin concentration of the birds and this was comparable to those on control diet. These findings is in accordance with the result of Cetin, Güçlü, and Cetin (2005) and Mokhtar (2013), who reported an increase in haemoglobin and PCV values of turkeysand broilers respectivelydue to probiotic supplementation. According to Jin, Ho, Abdullah, and Jalaludin (1997), probiotic tends to increase the haematological profile of poultry either due to its direct effects on haemopoetic organs or the indirect effects on the intestinal micro flora. In contrary, thisresultis not in agreement with the findings of Djouvinov, Boicheva, Simeonova, and Vlaikova (2005), who reported that the probiotic supplementation had no influence on blood constituents comprising haemoglobin concentrations. The differences may be attributed to type and number of species of bacteria present in probiotics (Al-Saad, Abbod, & Abo Yones, 2014). Above all, the haematological values in this present studyare within normal ranges (Mitruka & Rawsley, 1977; Fasuyi & Nonyerem, 2007) and this indicated that the health of the birds was not compromised throughout the study period.

Table 3. Carcass traits and prime-cut parts of broilers fed graded levels of shea butter cake meal based diets with or without multi-strain probiotic supplementation.

Parameters	SBC 0%	SBC 5%-	SBC 5%+	SBC 10%-	SBC 10%+	SBC 15%-	SBC 15%+	SEM
Live weight (kg bird^{-1})	2.44^{a}	2.35^{ab}	2.43^{a}	2.22^{b}	2.41^{a}	1.99^{c}	2.39^{a}	0.07
Carcass weight (kg bird^{-1})	1.89^{a}	1.80^{a}	1.87^{a}	1.66^{b}	1.85^{a}	1.54^{c}	1.81^{a}	0.05
Carcass yield (%)	77.46^{a}	76.60^{ab}	76.95^{ab}	74.77^{c}	76.76^{ab}	74.37^{c}	75.73^{bc}	0.82
Prime-cut weight (% carcass weight)								
Breast %	29.80^{a}	29.01^{a}	29.50^{a}	26.08^{b}	29.31^{a}	25.23^{b}	29.29^{a}	1.23
Thigh %	16.68^{a}	15.70^{b}	16.50^{ab}	14.33^{c}	15.66^{ab}	12.79^{d}	15.23^{bc}	0.47
Drum stick %	13.77^{a}	11.94^{b}	13.47^{a}	10.94^{c}	11.30^{ab}	10.51^{c}	12.11^{ab}	0.36
Wings %	11.77^{a}	11.34^{a}	11.60^{a}	11.24^{a}	11.53^{a}	10.51^{b}	11.05^{b}	0.44
Back %	18.60^{a}	18.02^{ab}	18.52^{a}	15.50^{c}	18.26^{a}	15.12^{c}	16.68^{b}	0.50
Abdominal fat %	3.78^{a}	3.62^{a}	3.46^{a}	3.32^{a}	1.82^{c}	2.85^{b}	1.73^{c}	0.09

^{abc}means in a row with different superscripts differ significantly ($p < 0.05$).

Table 4. Relative organ weights (% live weight) of broilers fed graded levels of Sheabuttercake meal based diets with or without multi-strain probiotic supplementation.

Parameter	SBC 0%	SBC 5%-	SBC 5%+	SBC 10%-	SBC 10%+	SBC 15%-	SBC 15%+	SEM
Liver %	2.15^{a}	2.16^{a}	2.08^{ab}	2.15^{a}	2.03^{b}	2.17^{a}	2.02^{b}	0.04
Kidney %	0.17	0.18	0.19	0.18	0.14	0.15	0.18	0.06
Gizzard %	1.62	1.71	1.84	1.70	1.73	1.74	1.66	1.09
Heart %	0.32	0.23	0.30	0.25	0.22	0.28	0.26	0.10
Bursa %	0.22^{ab}	0.18^{b}	0.24^{a}	0.16^{bc}	0.23^{a}	0.13^{c}	0.22^{ab}	0.02
Intestine %	4.12	3.96	4.10	3.87	4.08	3.66	4.03	0.25

^{abc}means in a row with different superscripts differ significantly ($p < 0.05$).

Table 5. Haematological response of broilers (42-d) fed graded levels of shea butter cake (SBC) meal based diets with or without multi-strain probiotic supplementation.

Parameters	SBC 0%	SBC 5%-	SBC 5%+	SBC 10%-	SBC 10%+	SBC 15%-	SBC 15%+	SEM
PVC (%)	28.25^{a}	25.07^{ab}	27.74^{a}	22.47^{b}	26.59^{a}	19.25^{c}	25.86^{a}	1.60
Hb (g dL-1)	9.43^{a}	8.84^{ab}	9.58^{a}	8.19^{b}	9.21^{a}	6.13^{c}	8.89^{ab}	0.38
WBC (10^{12}mm-^{3})	23.53	22.28	23.55	21.26	22.67	21.69	22.56	1.15
RCB (10^{9}mm-^{3})	3.23	2.97	3.78	2.46	3.66	2.35	3.54	0.74
MCH (ug)	34.95	35.64	33.94	37.88	37.92	33.15	35.43	2.95
MCHC (%)	29.71	25.32	27.35	24.34	31.11	28.34	30.15	2.42

^{abc}means in a row with different superscripts differ significantly ($p < 0.05$).

Conclusion

Based on the findings of this study, higher levels of SBC without probiotic supplementation, acceptability of the diet showed negative influence on carcass parameters as well as PCV and haemoglobinvalues of the birds. Therefore, multi-strain probiotic supplementation of SBC up to 15% level exhibited an improved carcass traits and relative prime cuts equal to the control dietswithout any apparent deleterious health implications.

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