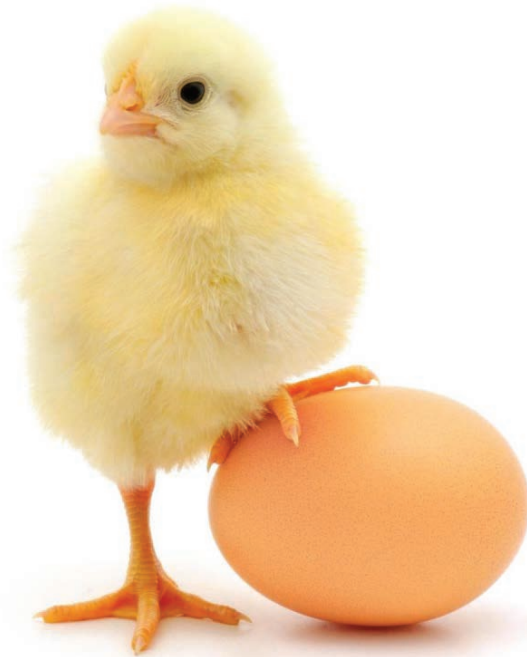


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Research Article

Effect of Stabilized Products of Sorghum Enriched with Lactobacilli (SPSL) on Growth Performance, Haematological Parameters and Ileal Microflora of Guinea Fowl Broilers (*Numida meleagris*)

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Abstract

Objective: This experiment was conducted to investigate the effects of Stabilized products of sorghum enriched with lactobacilli (SPSL) on growth performance, haematological parameters and ileal microflora of Guinea fowl. **Materials and Methods:** A total of 520 one day old guinea fowls with average body weight of 33.02g were assigned to 4 treatments with 5 replicates (26 birds/replicate). The 4 treatments were: (1) Only basal diet (T⁻), (2) Basal diet supplemented with antibiotics in water (T⁺), (3) Basal diet supplemented with the SPSL at the dose of 1.5% (T_{1.5}), (4) Basal diet supplemented with SPSL at the dose of 3% (T₃). At 12 week of age, blood samples were collected from 40 birds per treatment for haematological analysis. The birds were also slaughtered and ileal contents were harvested for microbiological analysis. **Results:** The results showed that there were no significant differences in the feed intake, feed conversion ratio and body weights of the birds across the treatments. Weight and length of intestine, caeca length and abdominal fat of the birds in T₃ were higher (p<0.05) than those of the other treatment groups. The lymphocyte in T_{1.5} group was higher than those of T⁻ group (p <0.05). Total coliforms bacteria was higher in the birds of T⁺ and T⁻ treatment groups than those of T_{1.5} and T₃. The level of *Escherichia coli* was lower (p<0.05) in the birds of T₃ group compared to other treatment groups. Total coliforms in T_{1.5} and T₃ birds were lower than those of T⁻ and T⁺. **Conclusion:** It was concluded that the SPSL significantly improved the intestinal parameters and reduced the potential pathogen bacteria.

Key words: Guinea fowl broilers, growth, antibiotic, ileal microflora, sorghum, coliforms bacteria

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

The inappropriate use of antibiotics to prevent disease and improve animal performance is a common practice in poultry production. This practice has resulted in various problems, such as drug residues in poultry meat and eggs¹, development of drug-resistant bacteria² and imbalance in normal gut microflora³. Thus, it has become necessary to develop alternatives using non-therapeutic substitutes (prebiotics, probiotics and symbiotic). Among these alternatives, prebiotic and probiotic has been given more attention. Prebiotics are non-digestible feed components that are potentially beneficial to host health because of their fermentable properties that stimulate the growth and/or activity of bacteria such as *Lactobacillus*, *Enterococcus*, *Pediococcus* and *Bacillus* in the ileum and caecum⁴. It generally consists of short chain polysaccharides and oligosaccharides.

Several prebiotics are generated from yeast cell walls and fermentation products. Prebiotics are not digestible by the host but commensal intestinal bacteria (*Lactobacillus*, *Bifidobacterium*, *Leuconostoc*, *Enterococcus*, *Lactococcus*, *Bacillus*, *Saccharomyces*, *Aspergillus* and *Pediococcus*) can metabolize them to produce short chain fatty acids like propionate, acetate and butyrate⁵. These prebiotic constituents have positive effects on poultry productivity and contribute to a healthy intestinal tract and can be a good alternative to antibiotics⁶. When ingested, prebiotics alter the caecal microbial composition, resulting in changes in the proteobacteria and changes in the genus and family of bacteria which causes change in growth⁷.

Probiotics are live microorganisms that, when administered in adequate amounts, confer a health benefit on the host⁸. It has been reported that probiotics stimulated the immune system and increased defense activity against pathogenic bacteria in broiler chickens⁹. The purpose of feeding probiotics is to stabilise beneficial microbes, to prevent the accumulation of pathogenic gastrointestinal bacteria and subsequently, to help maintain animal health¹⁰. Probiotics alter the intestinal microbial population and maintain its natural microbial flora by stimulating the growth and proliferation of useful bacteria. Other studies have reported an improvement in performance of chickens when they were fed with probiotic^{5,11-14}.

Generally, guinea fowls (*Numida meleagris*) are kept by smallholder farmers for meat, eggs and cash. However, mortality of young guinea fowls (keets) is high due to microbial infestations¹⁵. Probiotics can provide the same protection as a naturally developed commensal

gastrointestinal tract (GIT) microflora^{16,17}. Stabilized products of sorghum enriched with lactobacilli (SPSL) is a stabilized product of sorghum flour and sorghum malt enriched with lactobacillus (*L. casei*, *L. fermentatum*, *L. acidophilus* and *Enterococcus faecium*). Gnikpo *et al.*¹⁸ demonstrated that feeding SPSL to rabbits in a diet, increased body weights and improved health status. To our knowledge, there are scarcity of reports regarding the use of SPSL in poultry diets. Therefore, this study aimed to investigate the effect of SPSL on growth performance, haematological parameters and ileal microflora of "Galor" guinea fowl broilers.

MATERIALS AND METHODS

Experimental design: A total of 520 one-day-old guinea fowls broilers with average body weight of 33.02 g were assigned to 4 treatments with 5 replicates (26 birds/replicate). The treatments were: (1) Only basal diet (negative control) (T⁻), (2) basal diet supplemented with antibiotics treatments in water (T⁺), (3) Basal diet supplemented with the SPSL at the dose of 1.5% (T_{1.5}), (4) Basal diet supplemented with SPSL at the dose of 3% (T₃). The birds were reared for 12 weeks partitioned into starter (0-4 weeks), grower (4-8 weeks) and finisher (8-12 weeks) phases. The composition and nutritive values of each diet are presented in Table 1. The birds were reared on a floor pen with litter at stocking density of 20 birds m⁻² and photoperiod of 23 h of light during the first 4 weeks of age. From 5 weeks of age onward, the stocking density was

Table 1: Composition of the basal diet

Feedstuff	Dry matter (%)		
	Starter mash	Grower mash	Finisher mash
Maize	57.00	54.60	64.50
Wheat bran	4.00	13.50	8.00
Fish meal	2.00	5.00	2.00
Soya seed	15.00	10.00	18.00
Concentrate	5.00	5.00	2.00
Oyster shell	1.00	2.50	2.00
Total	100.00	100.00	100.00
Calculated nutritive values			
Crude protein (%)	21.27	18.20	17.74
Lysine (%)	0.50	0.92	0.20
Methionine (%)	0.50	0.60	0.30
Methionine+Cysteine (%)	1.21	0.60	0.77
Calcium (%)	0.92	1.57	1.08
Phosphorus (%)	0.62	0.56	0.45
Fiber (%)	5.34	5.30	5.34
Metabolizable Energy (kcal kg ⁻¹)	2970.00	2786.00	3002.00

¹Supplied per kilogram of diet; vitamin A: 15,000 IU, Vitamin D3: 5 000 IU, Vitamin E: 100 mg, Vitamin K: 5 mg, Thiamin: 5 mg, Riboflavin: 8 mg, Pyridoxine: 7 mg, Vitamin B12: 0.02 mg, Niacin: 100 mg, Folic acid: 3 mg, Biotin: 0.3 mg, Calcium pantothenate: 25 mg, Choline: 550 mg, Manganese: 80 mg, Zinc: 90 mg, Iron: 50 mg, Copper: 20 mg, Iodine: 2 mg, Selenium: 0.2 mg, Cobalt: 0.6 mg, Butylated hydroxytoluene: 125 mg

10 birds m⁻² and light was reduced gradually until natural photoperiod of 12L/12D at the end of 6 weeks of age. Feed and water were supplied *ad libitum* to the birds throughout the experiment. The antibiotic used was TetracolivitND produced by Laprovect (Address: 7 Rue du Tertreau, 37390 Notre-Dame-d'Oé, France) and contained tetracycline, colistin and some vitamins. Body weights and feed intake were recorded weekly. Mortality was recorded according to treatment during the 12 weeks of rearing.

Process of SPSL preparation:

- **Microbial sources for SPSL production:** Four strains of lactic acid bacteria (LAB) i.e. *Lactobacillus casei*, *Lactobacillus fermentatum*, *Lactobacillus acidophilus* and *Enterococcus faecium* isolated from kpètè-kpètè¹⁹ and previously characterized for their probiotic potential²⁰ were used in this study
- **SPSL production from sorghum and bacteria:** SPSL was produced and stabilized according to the modified methods of Chabi *et al.*²¹ (Fig. 1). Sterilized sorghum flour (75% of dehulled sorghum grains and 25% of sorghum malt) was mixed with distilled water to obtain a dough with a water content of 45%. The dough was aseptically inoculated with the four LABs (*L. casei*, *L. fermentatum*, *L. acidophilus* and *Enterococcus faecium*) in order to obtain 10⁶ UFC g⁻¹ of each strain and then kneaded into dough and allowed to ferment in a plastic bucket with lid for 36 h. The fermented dough samples were dried in a ventilated oven drier (Venticell, Fisher, Bioblock Scientific, MMM, Medcenter) at optimal conditions (42°C, 24 h) ensuring the best functionality of the product

Data collection

Growth performance: The birds were weighed weekly until 12 weeks of age. Feed intake was recorded weekly and was

used to calculate average feed intake per bird. Body weights and feed intake were used to calculate average body weight gain and feed conversion ratio as shown below:

$$\text{Feed conversion ratio} = \frac{\text{Feed intake}}{\text{weight gain}}$$

Haematological analysis: At week 12, blood samples were collected through the wing veins of 40 birds per treatment for the determination of haematological parameters. Blood samples were collected into anticoagulant (heparin) bottles and analysed for red blood cells, haemoglobin, packed cell volume, white blood cells using automatic analyzer: ABX Micros 60 from Sysmex Corporation International Company as described by Nakul-Aquaronne *et al.*²².

Organs weights and carcass yield percentages: At the end of week 12 of age and after weighing, 40 birds from each treatment were slaughtered to determine the weights of carcass, heart, liver, gizzard, pancreas, spleen, empty intestine, empty caeca and abdominal fat. These weights were used to determine the carcass yield and the organ weight/body weight ratios as:

$$\text{Carcass yield or ratio organ weight to body weight} = \frac{100 \times (\text{carcass weight or organ weight})}{\text{Body weight}}$$

The length of the intestine and caeca was also measured.

Gut digesta pH and microbiological profile: The pH of the digesta in the different parts of the digestive tract (crop, proventriculus, gizzard, duodenum, jejunum and ileum) was measured using a HANNA instruments pH-meter "pH H10838" and 10 cm segments of ileum was dissected and approximately 1 g of ileum contents was aseptically collected into petri dish (Thermo Fisher Scientific Inc., Seoul,

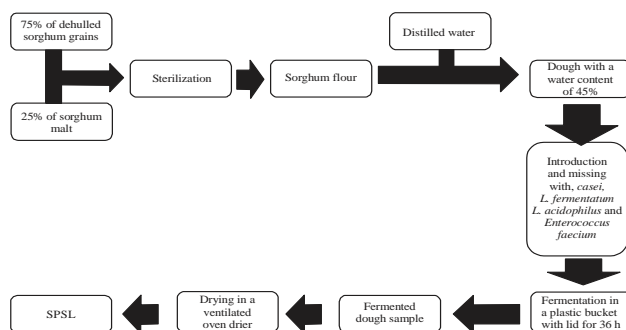


Fig. 1: Process of SPSL preparation

South Korea) for microbiological analysis. The method of Foo *et al.*²³ was used to determine: Total aerobic bacteria, *Escherichia coli*, Total coliforms bacteria and *Salmonella* spp. Microbial enumeration was performed as follows: 1 g of each sample was crushed in 9 mL tryptone salt in aseptic conditions. Serial dilutions from 10⁻¹ to 10⁻⁵ were prepared from these suspensions. One millilitre of each dilution was used for cell enumeration. Total aerobic bacteria were determined by plate count agar after 72 h incubation at 30°C. *Escherichia coli* were enumerated on "Brilliance *E. coli*" after 24 h incubation at 44°C and coliform total bacteria were determined on violet red bile lactose after 24 h incubation at 30°C. For *Salmonella* spp., buffered peptone water was used for pre-enrichment at 37°C for 24 h. Thereafter, enrichment at 37°C for 24 h was made with rappaport Vassiliadis soya broth prior for isolation and counting on Hektoen and SS agar at 37°C (24 h). Bacteria types were identified with the Api 20E system (Apparatus and Identification Procedures, La Balmeles-Grottes, Cedex 2 France). All bacteria were counted and expressed as total colony forming unit (CFU) per g of the digesta and results were presented as log₁₀-transformed data.

Statistical analysis: The data were analyzed using one-way analysis of variance (ANOVA) followed by Tukey test to compare differences between treatment groups using the Graph Pad software. Results are presented as means±the standard error of the mean (M±SEM). Difference of p<0.05 were considered statistically significant.

RESULTS

Effect of SPSL on production parameters: Figure 2 shows the weekly body weights according to the dietary treatments. Overall, body weight increased with age. The data show that there was no significant difference (p>0.05) in body weights across the treatment groups during the experimental period. Table 2 shows the effect of SPSL on the growth performance of guinea fowl broilers. There was no significant difference (p>0.05) in the feed intake, body weight gain and feed conversion of the birds.

Table 2: Effect of SPSL on the growth performance of guinea fowl broilers

Parameters	Groups				p-value
	T ⁻	T ⁺	T _{1.5}	T ₃	
Feed intake (g)	57.45±8.62	57.22±9.39	59.31±8.97	66.14±10.17	0.895
Body weight gain (g)	13.97±1.88	13.83±1.47	14.02±1.68	14.34±1.87	0.835
Feed conversion ratio	4.32±0.55	4.13±0.45	4.21±0.39	4.65±0.55	0.885

T⁻: Only basal diet, T⁺: Basal diet supplemented antibiotics treatments in water, T_{1.5}: Basal diet supplemented with the SPSL at the dose of 1.5%, T₃: Basal diet supplemented with SPSL at the dose of 3%

Effect of SPSL on relative organ weights and intestinal parameters:

Table 3 shows the effects of SPSL on relative organs weights and intestinal parameters of guinea fowl broilers. The intestinal length of guinea fowls of the birds of T₃ was significantly higher (p<0.05) than those of the other treatments whose values were comparable (p>0.05). A similar trend was observed in the in the intestinal weights and Caecal length of the birds. The caecal weights of the birds in T₃ and T⁺ were similar to those of T⁻ but significantly higher (p<0.05) than those of T_{1.5}. Liver weights of the guinea fowls in T⁺ group were similar to those of T⁻ and T₃ but higher (p<0.05) than those of T_{1.5}. The liver weights of the birds in T_{1.5}, T⁻ and T₃ were comparable. The relative weights of pancreas of the birds in T_{1.5} was similar to that of T⁺ but significantly higher (p<0.05) than those of T⁻ and T₃. Abdominal fat of T₃ guinea fowl was higher than those of the other treatments whose weights were alike.

Effect of SPSL on haematological parameters:

Table 4 shows the effect of SPSL on haematological parameters of guinea fowl broilers. The white blood cells of T₃ birds was lower (p<0.05) than that of T⁺ but not different from T⁻ and T_{1.5}. The lymphocyte was significantly higher (p<0.05) in the bird in T_{1.5} group than that of T⁻ but comparable to those of the other treatments. The granulocyte of T_{1.5} and T₃ birds was similar but

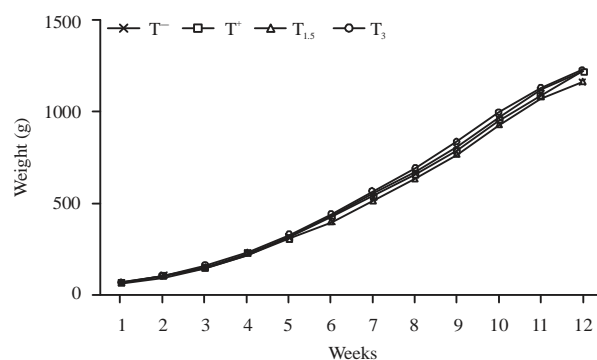


Fig. 2: Effect of SPSL on body weight

T⁻: Only basal diet, T⁺: Basal diet supplemented antibiotics treatments in water, T_{1.5}: Basal diet supplemented with the SPSL at the dose of 1.5%, T₃: Basal diet supplemented with SPSL at the dose of 3%

Table 3: Effect of SPSL on relative organ weights and intestinal parameters of guinea fowl broilers

Parameters	Groups				p-value
	T ⁻	T ⁺	T _{1.5}	T ₃	
Intestine length (cm)	168.00±2.72 ^b	168.60±2.04 ^b	169.90±2.51 ^b	174.20±2.63 ^a	0.0292
Intestine weight (%)	2.98±1.04 ^b	2.48±1.31 ^b	2.98±0.87 ^b	3.89±0.70 ^a	0.0001
Caeca length (cm)	32.88±0.57 ^b	33.44±0.69 ^b	34.44±0.56 ^b	36.11±0.49 ^a	0.0013
Caeca weight (%)	0.36±0.04 ^{ab}	0.37±0.02 ^a	0.33±0.02 ^b	0.37±0.15 ^a	0.0028
Gizzard(%)	2.19±0.10	2.20±0.10	2.06±0.07	1.90±0.06	0.0719
Liver (%)	1.78±0.07 ^{ab}	1.85±0.05 ^a	1.59±0.04 ^b	1.63±0.03 ^{ab}	0.0023
Heart (%)	0.38±0.01	0.42±0.01	0.42±0.02	0.40±0.01	0.1040
Pancreas (%)	0.17±0.01 ^b	0.16±0.007 ^{ab}	0.14±0.005 ^a	0.17±0.005 ^b	0.0140
Spleen (%)	0.05±0.003	0.05±0.002	0.05±0.002	0.05±0.004	0.0980
Carcass (%)	71.81±0.77	71.20±1.13	73.31±2.41	74.41±0.24	0.2865
Abdominal fat (%)	0.84±1.12 ^b	0.92±0.84 ^b	1.02±0.95 ^b	1.81±1.58 ^a	0.0031

^{a,b}Means in a row followed by different subscripts differ significantly (p<0.05). T: Only basal diet, T⁺: Basal diet supplemented antibiotics treatments in water, T_{1.5}: Basal diet supplemented with the SPSL at the dose of 1.5%, T₃: Basal diet supplemented with SPSL at the dose of 3%

Table 4: Effect of SPSL on haematological parameters

Parameters	Groups				p-value
	T ⁻	T ⁺	T _{1.5}	T ₃	
White blood cells (× 10 ⁹ L ⁻¹)	195.00±2.53 ^{ab}	202.50±2.05 ^a	200.80±2.21 ^{ab}	194.00±1.84 ^b	0.0229
Lymphocyte (× 10 ⁹ L ⁻¹)	33.47±2.39 ^b	38.88±1.66 ^{ab}	43.86±3.14 ^a	38.98±2.62 ^{ab}	0.0437
Granulocyte (× 10 ⁹ L ⁻¹)	86.36±2.78 ^a	84.86±1.75 ^a	79.20±2.75 ^b	78.29±2.34 ^b	0.0489
Red blood cells (× 10 ¹² L ⁻¹)	2.59±0.04 ^{ab}	2.67±0.04 ^a	2.56±0.04 ^{ab}	2.43±0.03 ^b	0.0009
Haemoglobin (g dL ⁻¹)	13.52±0.20 ^{ab}	13.88±0.14 ^a	13.59±0.18 ^{ab}	13.02±0.14 ^b	0.0075
Haematocrit (%)	42.74±0.68 ^{ab}	44.29±0.47 ^a	42.16±0.52 ^{ab}	41.23±0.58 ^b	0.0030
Mean Cell Volume (fl)	164.80±1.48	166.20±1.42	165.20±1.78	169.70±1.24	0.0850
Mean cell haemoglobin concentration (g dL ⁻¹)	31.61±0.25 ^{ab}	31.30±0.15 ^b	32.20±0.23 ^a	31.82±0.22 ^{ab}	0.0436
Platelet (× 10 ⁹ L ⁻¹)	26.33±2.16 ^b	26.11±1.7 ^b	23.88±2.13 ^b	42.00±6.07 ^a	0.0030

^{a,b}Means in a row followed by different subscripts differ significantly (p<0.05). T: only basal diet, T⁺: Basal diet supplemented antibiotics treatments in water, T_{1.5}: Basal diet supplemented with the SPSL at the dose of 1.5%, T₃: Basal diet supplemented with SPSL at the dose of 3%

Table 5: Effect of SPSL on the pH of digestive tract contents

Segments	Groups				p-value
	T ⁻	T ⁺	T _{1.5}	T ₃	
Crop	5.32±0.18 ^a	5.27±0.14 ^a	4.68±0.08 ^b	4.97±0.09 ^{ab}	0.0028
Proventriculus	5.44±0.07 ^a	5.51±0.006 ^a	5.06±0.08 ^b	5.49±0.09 ^a	0.0005
Duodenum	6.23±0.06 ^a	6.35±0.02 ^a	6.22±0.08 ^a	5.90±0.12 ^b	0.0030
Jejunum	6.60±0.08 ^a	6.38±0.06 ^a	6.10±0.09 ^b	6.35±0.05 ^a	0.0003
Ileum	7.08±0.03 ^a	6.85±0.04 ^b	6.87±0.08 ^{ab}	6.79±0.0 ^{ab}	0.0042
Caeca	6.82±0.09	6.83±0.08	6.88±0.07	6.93±0.07	0.7000

^{a,b}Means in a row followed by different subscripts differ significantly (p<0.05). T: Only basal diet; T⁺: Basal diet supplemented antibiotics treatments in water, T_{1.5}: Basal diet supplemented with the SPSL at the dose of 1.5%, T₃: Basal diet supplemented with SPSL at the dose of 3%

significantly lower (p<0.05) than those of T⁻ and T⁺. The red blood cells, haemoglobin and haematocrit of T₃ guinea fowl were lower (p<0.05) than those of T⁺ but similar to those of the birds in the other treatment groups. However, the level of Mean cell haemoglobin concentration was higher (p<0.05) in the birds of T_{1.5} group than that of T⁺. The platelet level was significantly higher (p<0.05) in the birds of T₃ group than those of the other groups. In addition, the level of Mean cell volume did not show any significant difference across the treatments.

Effect of SPSL on pH of digestive tract contents: Table 5 shows the pH of the digesta in the different parts of the digestive tract of guinea fowl broilers. The crop digesta pH was significantly lower (p<0.05) in T_{1.5} birds than those of T⁻ and T⁺ but similar to that of T₃ treatment group. The proventriculus digesta pH was lower in the birds of T_{1.5} group than those of the other treatment groups whose pH was similar. The pH in the duodenum digesta was significantly lower (p<0.05) in the T₃ group than those of the other treatment groups. Also, the jejunum digesta pH was comparable in T⁻, T⁺ and T₃ but

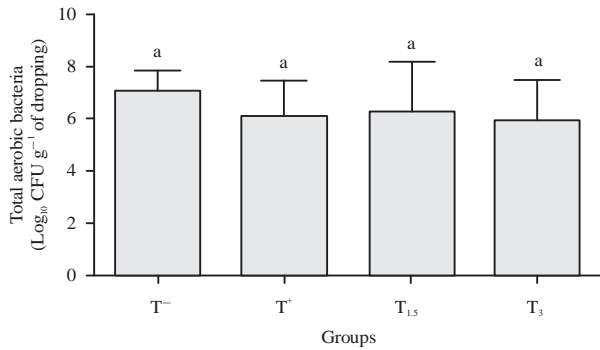


Fig. 3: Effect of SPSL on Total aerobic bacteria

T: Only basal diet, T⁺: Basal diet supplemented antibiotics treatments in water, T_{1.5}: Basal diet supplemented with the SPSL at the dose of 1.5%, T₃: Basal diet supplemented with SPSL at the dose of 3%

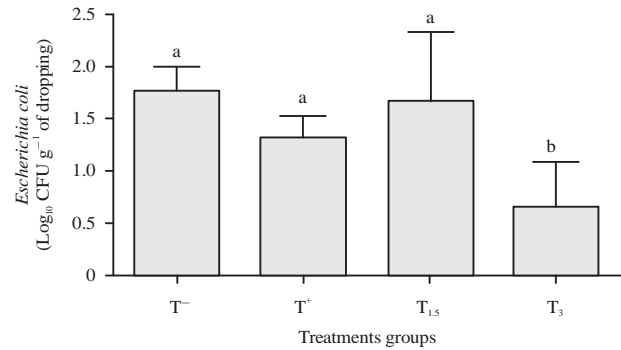


Fig. 5: Effect of the SPSL on *Escherichia coli*

^{a,b}Means in a row followed by different subscripts differ significantly (p<0.05). T: Only basal diet, T⁺: Basal diet supplemented antibiotics treatments in water, T_{1.5}: Basal diet supplemented with the SPSL at the dose of 1.5%, T₃: Basal diet supplemented with SPSL at the dose of 3%

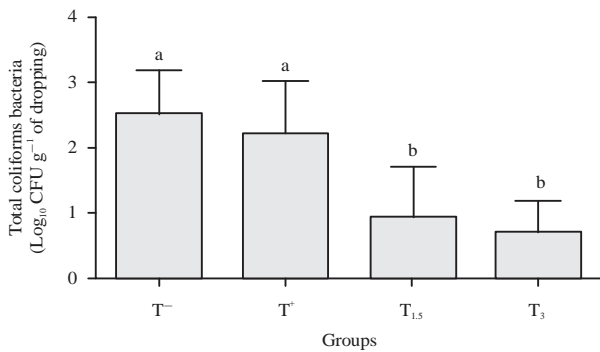


Fig. 4: Effect of the SPSL on the total coliforms bacteria.

^{a,b}Means in a row followed by different subscripts differ significantly (p<0.05). T: Only basal diet, T⁺: Basal diet supplemented antibiotics treatments in water, T_{1.5}: Basal diet supplemented with the SPSL at the dose of 1.5%, T₃: Basal diet supplemented with SPSL at the dose of 3%

significantly higher (p<0.05) than that of T_{1.5}. In addition, the pH in the ileum digesta was significant lower (p<0.05) in the birds of T⁺ group than those of T⁻ group but similar to those of T_{1.5} and T₃. The pH of caeca did not show any significant difference across the treatments.

Effect of SPSL on Ileal microflora: Figure 3, 4 and 5 show the effect of the SPSL on the intestinal microflora identified in digesta collected from ileum part of the intestine. There was no significant difference (p>0.05) in total aerobic bacteria (Fig. 3). However, the level of total coliforms bacteria was lower (p<0.05) in the birds of T_{1.5} and T₃ groups than those of T⁺, T⁻ groups (Fig. 4). The level of *Escherichia coli* was lower (p<0.05) in the birds of T₃ compared to the other groups (Fig. 5). No *salmonella spp* was found in all the treatments.

DISCUSSION

This study has demonstrated that inclusion of sorghum enriched with lactobacilli (*Lactobacillus casei*, *Lactobacillus fermentatum*, *Lactobacillus acidophilus* and *Enterococcus faecium*) in the guinea fowl broilers diet did not significantly improve the weight gain, feed intake and feed conversion ratio of the birds. The similarity in the weight gain of the birds in the present study partially agrees with the observation of Huang *et al.*²⁴ who reported that there was no difference in the performance parameters of broiler chickens supplemented with either *lactobacillus casei* or *lactobacillus acidophilus* with or without cobalt. There have also been several studies with no positive results when broilers were fed with probiotic supplements^{25,26}. Moreover, Vale *et al.*²⁷ and Öztürk *et al.*²⁸ reported that the supplementation of chicken diet with organic acids, probiotics or antibiotics did not have significant effect on weight gain. The findings in the present trial is, however, at variance with the observation of Sarfo *et al.*¹⁵ who reported an increase in the weights of guinea fowl fed 1.5%DFM[®] commercial probiotic composed of *Lactobacilli* (1×10⁸ CFU g⁻¹), *Bacillus* (1×10¹² CFU g⁻¹) and *Saccharomyces cerevisiae* (yeast, 1×10⁵ CFU g⁻¹). Other studies also reported that there was higher weight gains when probiotics were fed to birds²⁹⁻³¹.

In agreement with the finding in the present study, Kalavathy *et al.*³² reported that probiotic supplementation of *Lactobacillus* did not affect feed intake of hens during the rearing period. The similarity in the feed intake of the birds in this study corroborates the report of Gnikpo *et al.*¹⁸ who indicated that the SPSL did not contain any toxic substance that may limit feed intake.

The birds in T_{1,5} had lower liver weights than those of the control group (receiving antibiotic). This difference could be attributed to the fact that the liver of the birds in this treatment was less stressed during the process of digestion and absorption of nutrients. The relative weights of gizzard, heart, spleen and carcass yield percentage of the birds in this study were not affected by the treatments. There is a paucity of information on the effects of probiotic on relative organ weights of guinea fowl. Similar to our findings, Wang and Gu³³ did not observe any significant effects of probiotic supplementation on relative weights of gizzard, heart and spleen, spleen of broiler chickens. However, Awad *et al.*³⁴ reported that probiotic supplementation increased the carcass yield percentage during the rearing period. For abdominal fat, the highest fat level was recorded in the birds administered 3% of the SPSL. This result may be due to the excess energy provided by the diet in this treatment group which was converted into fat by the birds. In terms of length and weight of the intestine, the birds offered 3% of SPSL showed the best performance compared to the other groups. This may be ascribed to the probiotic bacteria contained in the SPSL, which enhanced the digestion of the birds by promoting degradation and food absorption.

With regard to haematological parameters, the level of lymphocytes in the birds of T_{1,5} was higher than those of the control group T⁻. This result may be due to the activation of lymphocytes by dendritic cells following the penetration of antigens into the body. In terms of mean cell volume, the values were similar in all the treatments, corroborating the results of Sarfo *et al.*¹⁵ who reported that DFM[®] probiotic did not affect mean cell volume. These results are in agreement with those of Bhatti *et al.*³⁵ who studied the influence of probiotic on blood parameters and reported that there was a stimulation of the hematopoietic system with an increase in erythrocytes without an increase in haemoglobin, associated with an increase in the average (corpuscular) volume (MCV). There was no salmonella found in the intestinal microflora of the birds. These results confirm the observation of Ramdane and Guitarni³⁶ who did not observe any salmonella due to the effect of probiotics on three intestinal flora germs in broiler chickens. The current study demonstrates that total coliforms bacteria were low in the ileum of guinea fowl broilers. The SPSL reduced the population of total coliforms bacteria in the ileum compared to the control groups T⁻ and T⁺. This is in agreement with the findings of Mulder *et al.*³⁷ who reported that inoculation with a probiotic strain of *L. reuteri* significantly reduced the number of total coliforms bacteria in broiler chickens. A similar finding was reported by Lan *et al.*³⁸ with a mixture of *L. acidophilus/gallinarum*, *Lactobacillus agilis*, *L. salivarius* and *Lactobacillus* spp. Probiotics, such as

L. crispatus, *L. salivarius* and *L. johnsonii*, have antimicrobial activities against total coliforms bacteria^{16,39-41}. Cao *et al.*⁴² reported that broiler chickens fed diets supplemented with *Lactobacilli* spp. were more resistant to the pathogenic effects of *E. coli*. The antimicrobial effects of probiotics are due to the volatile fatty acids (VFA), other organic acids such as lactate and succinate⁴³, production of bacteriocins and phage-displayed peptides⁴⁴⁻⁴⁶.

As regards *Escherichia coli*, the level of the bacteria was lower in the birds of T₃ than those of T_{1,5} and control groups (T⁻, T⁺). This result is consistent with the findings of Ramdane and Guitarni³⁶ who reported that *Escherichia coli* was lower in the broilers fed probiotic throughout the rearing period. This observed variation may be due to lactobacilli contained in the SPSL. *Lactobacillus reuteri* produces reuterin, an intermediate metabolite with antimicrobial activity and also acting against *salmonella*, *Escherichia coli* and *compylobacter*⁴⁷.

CONCLUSION

The supplementation of SPSL up to 3% did not significantly improve the weight gain, feed intake and feed conversion ratio of guinea fowl broilers. However, The SPSL at 3% improved the intestine length, intestine weight, caeca length, abdominal fat and reduced the level of total coliforms bacteria and *Escherichia coli*.

SIGNIFICANCE STATEMENT

This study discovered that inclusion of sorghum enriched with lactobacilli (*Lactobacillus casei*, *Lactobacillus fermentatum*, *Lactobacillus acidophilus* and *Enterococcus faecium*) in the diets of guinea fowl broilers improved carcass yield, the intestine length, intestine weight, caeca length, abdominal fat and reduced the level of total coliforms bacteria and *Escherichia coli*. Supplementation of SPSL in the diet can improve carcass yield and beneficial microbiological profile of guinea fowl broiler without the use of antibiotics as growth promoters. This study will help the researcher to uncover the critical area of the effect of SPSL on growth performance, as well as the microbiological profile of guinea fowl broilers which many researchers have not been able to explore. Thus, a new theory on the use of SPSL in the diet of guinea fowl broiler can be achieved.

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