

## Effects of *Salicornia herbacea* on Growth Performance, Meat Quality, Excreta Microbial Populations, and Noxious Gas Emissions in Broiler Chicks

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The current study was conducted to evaluate the effects of adding *Salicornia herbacea* extracts to the drinking water on the growth performance, meat quality, excreta microbial population, and noxious gas emission in broiler chicks. A total of 544 one-day-old broiler chicks (Ross 308) were used in a 35-d experiment. Broiler chicks were allocated to four treatments with eight replicates, based on a completely randomized design. Diet was the same for all treatments, but a liquid phytogetic supplementation using different quantities of *S. herbacea* was provided in the drinking water as follows: control (CON), with no *S. herbacea*; 1 cc/L *S. herbacea* (SAL1); 5 cc/L *S. herbacea* (SAL2); and 10 cc/L *S. herbacea* (SAL3). During d 22-35, and d 1-35, broilers supplemented with *S. herbacea* extracts had a higher body weight gain (BWG) compared with the broilers in the CON group ( $P < 0.05$ ), but broilers supplemented with *S. herbacea* extracts had a lower feed conversion ratio (FCR) when compared with broilers in the CON group ( $P < 0.05$ ). Supplementation with *S. herbacea* extracts had linear effects on the abdominal fat and the redness ( $a^*$ ) of meat ( $P < 0.05$ ). There were no significant differences between excreta microbial populations and excreta noxious gas emissions in broilers in the CON group, or broilers supplemented with *S. herbacea* extracts. In conclusion, the results of this study demonstrate that *S. herbacea* supplementation positively affected the growth performance and meat quality in broilers, indicating that *S. herbacea* can be safely used to replace antibiotic as a growth promoter, thereby reducing the risk of antibiotic resistance issues.

**Key words:** broiler chicks, growth performance, excreta microbial population, meat quality, *Salicornia herbacea*

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### Introduction

The ban on using antibiotics as feed additives has accelerated research into the use of alternative feed additives in poultry production (Kiczorowska *et al.*, 2016). Phytogetic additives are plant-derived products used in animal feed to improve the performance of agricultural livestock (Windisch *et al.*, 2008; Hashemi and Davoodi, 2010). This class of additives has recently gained increasing interest, especially for use in modern poultry production (Al-Yasiry *et al.*, 2017). Phytogetic supplements can improve the health status of broilers and their production performance, among other things, by stimulating the microbiota of the gastrointestinal tract, or improving the digestibility of nutrients (Abdel-Wareth *et al.*, 2012; Cho *et al.*, 2014). The chemical composition of *Salicornia herbacea* reported by the Korea

National Fisheries Research and Development Institute is as follows: moisture content 90.9%, Fe 84.8 mg, Ca 650 mg, Na 1888.8 mg, Mg 50 mg, K 650 mg, Zn 29.6 mg, and I 70 mg per 100 g dry weight. *S. herbacea* is rich in amino acids (Min *et al.*, 2002); natural minerals (Tikhomirova *et al.*, 2008), and many bioactive substances, such as phytosterols (Zhu and Row, 2010), polysaccharides (Im *et al.*, 2006), and phenolic compounds, including flavonoids (Kim *et al.*, 2011b). Tungtungmadic acid (3-caffeoyl-4-dihydrocaffeoyl quinic acid),  $\beta$ -sitosterol, stigmasterol, uracil, quercetin 3-O- $\beta$ -D-glucopyranoside, and isorhamnetin 3-O- $\beta$ -D-glucopyranoside, were isolated from the methanol extract of *S. herbacea* (Kim and Park, 2004; Lee *et al.*, 2004; Chung *et al.*, 2005; Lee *et al.*, 2005; Kim *et al.*, 2011b). These compounds were recognized as important active ingredients in *S. herbacea*. A number of studies reported that *S. herbacea* possess anti-oxidative, anti-inflammatory, anti-hyperglycemic, and anti-hyperlipidemic characteristics (Im *et al.*, 2003; Seo *et al.*, 2004; Chung *et al.*, 2005; Lee *et al.*, 2006; Kim *et al.*, 2015). Meanwhile, some studies showed that *S. herbacea* had effects on bacteria and fermentations (Seo *et al.*, 2010; Kim *et al.*, 2011a; Kim and Park, 2012; Rad *et al.*,

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2014).

In addition to the many benefits of *S. herbacea* for human health, its effects on animal feeding have been increasingly investigated in recent years. Some studies have shown that *S. herbacea* has positive impacts on the physiological indexes of poultry. Al-Batshan *et al.* (2008) reported that SM (*Salicornia bigelovii* Torr meal) was effective in reducing feed intake and hence body weight gain and final body weight in broilers. Sarker *et al.* (2010) reported that *S. herbacea* probiotics, provided as 0.5% to 1.0% of diets, can be used to replace antibiotics in broiler production. Mohammadi *et al.* (2015) reported that the addition of *Salicornia* extracts to laying hen diets improved egg shell quality, decreased the egg breaking rate, and increased production on commercial farms. Based on these previous studies, we hypothesized that *S. herbacea* might have more positive effects on broiler chickens. The present study was designed to compare the efficacy of different concentrations of *S. herbacea* extracts in drinking water on the growth performance, meat quality, excreta microbial population, and excreta noxious gas emissions in broiler chickens.

### Materials and Methods

The experimental protocols describing the management and care of broilers were reviewed and approved by the Animal Care and Use Committee of Dankook University (No. DKU-1320). The *S. herbacea* products were obtained from a commercial company (Cargill Agri Purina, Inc. South Korea).

#### Experimental Design, Broilers, Housing, and Diets

In a 35-d trial, a total of 544 1-day-old male (Ross 308) broilers with an average initial body weight of  $45.8 \pm 0.3$  g were allotted to four experimental diets according to their initial body weight. There were eight replicate pens per treatment, with 17 broilers per pen. Diet was the same for all treatments, and *S. herbacea* extract was supplemented in drinking water at 0, 1, 5, and 10 cc/L for control (CON), SAL1, SAL2, and SAL3 groups, respectively. The concentrations of *S. herbacea* used in this study were modified from Mohammadi *et al.* (2015), and Sarker *et al.* (2010). *S. herbacea* was extracted with 25% ethanol and water at 70°C (Kim *et al.*, 2007). All broilers were housed in stainless steel cages (1.75 m × 1.55 m) with three floors, and light was provided for 24 h/day during the whole experiment. The initial temperature of the room was set at 32°C, and then the temperature was reduced by 2°C each day for five days, until it reached 24°C. The temperature was set then maintained at 24°C until the end of the experiment. The broiler chicks were given free access to water and mash feed during the entire experiment. All nutrients in the diet were formulated to meet or exceed the NRC (1994) recommendations for broilers (Table 1). The diets were supplied in three phases consisting of a starter phase, from d 0 to 7, grower diets from d 8 to 21, and finisher diets from d 22 to 35.

#### Growth Performance

The broilers were weighed, and feed intake was recorded on d 1, 7, 21, and 35. Body weight gain (BWG), feed intake

(FI), and feed conversion ratio (FCR) were then calculated. Water intake was recorded daily on a pen basis, and recorded daily to 35 days of age by checking the volume of water left in the drinkers at the end of each day and subtracting this from all the water allocated to each drinker in the preceding 24-h period.

#### Meat Quality and Relative Organ Weight

Sixteen broilers per treatment (two broilers/cage) were randomly selected and slaughtered by cervical dislocation. The body organs, including liver, spleen, bursa of fabricius, breast muscle, gizzard, and abdominal fat were collected. Relative organ weight (% of live BW) was calculated. Breast meat samples were collected for meat quality assessment. Meat color (lightness (L\*), redness (a\*) and yellowness (b\*)) was determined using a chromameter (Model CR-410, Minolta Co, Japan) and the standard color plate was L = 94.6, a = 0.3131, b = 0.3194. The pH value was determined using a pH meter (IstekNeoMet 77P, Istek Inc., Korea). The water holding capacity (WHC) was measured in accordance with the methods described by Kauffman *et al.* (1986). Briefly, a 0.2 g sample was pressed at 3,000 psi for 2 minutes on a 125-mm-diameter piece of filter paper. The areas of the pressed sample and the expressed moisture were delineated, and then determined using a digitizing area-line sensor (MT-10S; M.T. Precision Co. Ltd., Tokyo, Japan). The ratio of water/meat area was then calculated, giving a measure of WHC (a smaller ratio indicates a higher WHC). Drip loss was measured using approximately 2 g of meat sample according to the plastic bag method described by Honikel (1998).

#### Excreta Microbial Populations

Excreta samples were collected from four broilers in each cage and pooled. Pooled excreta samples were placed on ice for transportation to the laboratory, where analysis was immediately carried out. One gram of the composite excreta sample from each cage was diluted with 9 ml of 1% peptone broth (Becton, Dickinson and Co., Franklin Lakes, NJ, USA), and then homogenized. Viable counts of bacteria in the excreta samples were then conducted by plating serial 10-fold dilutions (in 1% peptone solution) on to MacConkey agar plates (Difco Laboratories, Detroit, MI, USA) and lactobacilli medium III agar plates (Medium 638, DSMZ, Braunschweig, Germany) to isolate *E. coli* and *Lactobacillus*, respectively. The MacConkey agar plates were incubated for 24 hours at 37°C. The lactobacilli medium III agar plates were then incubated for 48 hours at 39°C under anaerobic conditions. The *E. coli* and *Lactobacillus* colonies were counted immediately after removal from the incubator. The concentration of microflora was finally expressed as log<sup>10</sup> colony forming units per gram of excreta.

#### Excreta Noxious Gas Emission

For analysis of gas contents in excreta, 300 g of fresh excreta samples from each cage were collected in plastic boxes (polyvinyl, 25 × 35 cm) in triplicates at the end of the experiment and fermented in an incubator (28°C) according to the method described by Cho *et al.* (2008). After the fermentation process, a Gastec (Gas Detector, GV-100S;

Table 1. Broiler chicken feed composition<sup>1</sup>

Ingredients (g/kg)	Starter	Grower	Finisher
Corn	211.30	255.80	286.50
Wheat	350.00	350.00	350.00
Soybean meal (440 g crude protein/kg)	277.80	240.00	186.05
Corn gluten meal	50.00	40.00	50.00
Rapeseed meal	25.00	0.00	35.00
DDGS	0.00	11.35	0.00
Tallow	38.84	65.00	55.20
Limestone	16.24	12.90	13.73
Dicalcium phosphate	10.54	7.98	6.63
Sodium chloride	3.20	3.20	3.15
Sodium bicarbonate	1.80	0.56	0.79
Methionine (MHA 840 gr/kg)	2.85	2.13	1.71
L-Lysine-HCl (784 gr/kg)	6.50	6.00	6.03
Threonine (985 gr/kg)	1.20	0.90	0.87
Vitamin premix <sup>2</sup>	1.50	1.50	1.50
Trace mineral premix <sup>3</sup>	2.00	2.00	2.00
Choline chloride (750 gr choline/kg)	0.96	0.40	0.56
Xylanase <sup>4</sup>	0.12	0.13	0.13
Phytase <sup>5</sup>	0.15	0.15	0.15
Analyzed Nutritional Content			
ME, MJ/Kg	12.92	13.72	13.64
CP,%	22.65	21.00	19.13
Lys,%	1.43	1.31	1.18
Met + Cys,%	1.05	0.96	0.92
Thr,%	0.86	0.78	0.70
Ca,%	0.95	0.74	0.75
P,%	0.62	0.53	0.52
Fat, %	5.64	8.48	7.55
Ash, %	5.80	5.12	4.95
Moisture, %	12.00	11.63	11.77
Fiber, %	3.02	2.92	2.77

<sup>1</sup> Starter diets were provided from d 0 to 7, grower diets from d 8–21, and finisher diets from d 22 to 35.

<sup>2</sup> Provided per kg of diet: vitamin A (retinol), 4.5 mg; vitamin D3 (cholecalciferol), 0.094 mg; vitamin E ( $\alpha$ -tocopherol acetate), 55 mg; menadione, 2.55 mg; thiamine, 3 mg; riboflavin, 7.5 mg; pyridoxine, 4.5 mg; cobalamin, 24  $\mu$ g; niacin, 51 mg; folic acid, 1.5 mg; biotin 126 mg; pantothenic acid, 13.5 mg.

<sup>3</sup> Provided per kg of diet: Zn (zinc sulfate), 37.5 mg; Mn (manganese oxide), 137.5 mg; Fe (ferrous sulfate  $\cdot$  7H<sub>2</sub>O), 37.5 mg; I (potassium iodate), 0.83 mg; Se (sodium selenite  $\cdot$  5H<sub>2</sub>O), Cu (copper sulfate), 0.23 mg.

<sup>4</sup> Phytase premix prepared by serial dilution with corn to contain 1000 phytase units/g, provided by Easybiosystem, Seoul Feed Ltd., Seoul, Korea.

<sup>5</sup> Xylanase premix prepared by serial dilution with corn to contain 650 xylanase units/g, provided by Easybiosystem, Seoul Feed Ltd., Seoul, Korea.

Gastec Corp., Kanagawa, Japan) was used to detect gases. Levels of NH<sub>3</sub>, R.SH, H<sub>2</sub>S, and acetic acids were measured within the scope of 5.0–100.0 (No. 3La, detector tube; Gastec Corp.), 2.0–20.0 (4LK, detector tube; Gastec Corp.), 0.5–120.0 (No. 70 L and 70, detector tube; Gastec Corp.), and 2.0–50.0 (No. 81 L, detector tube; Gastec Corp.) ppm. For these measurements, the plastic containers were punctured, and gas was measured by inserting a gas detector tube attached to the detector approximately 2 cm above the feces, at a rate of 100 ml/min. Gas concentrations were obtained based on changes in the color of the gas detector tube.

### Chemical Analyses

All feed samples were analyzed for dry matter (930.15), crude protein (990.03), crude fiber (962.09) crude fat without acid hydrolysis (920.39), ash (940.26), phosphorus (965.17), and calcium (984.01), following AOAC (1995) procedures. Dietary Met and Cys were measured by acid hydrolysis with HCl after an oxidation step for quantification of total sulfur, and Trp was determined using reverse-phase HPLC (Waters 2690; Waters, Milford, MA, USA) after alkaline hydrolysis at 120°C for 16 h. The gross energy was determined using a bomb calorimeter (Mode 1241; Parr Instrument Co., Molin, IL, USA). The lysine content was analyzed using a Sykam

Amino Acid Analyzer (Laserchrom HPLC Laboratories Ltd. Inc., Rochester, UK) after acid hydrolysis for 24 h in 6 mol/L HCl (AOAC 2000).

### Statistical Analyses

The data were statistically analyzed using the GLM procedure of SAS (SAS Institute, 1998), with the pen as the experimental unit. Before conducting statistical analysis of the microbial counts, the value was transformed logarithmically. Orthogonal polynomials were used to assess the linear and quadratic effects of increasing the level of concentrations of *S. herbacea* extracts. Duncan's range test was adopted to compare the means of the treatments. Variability in the data was expressed as the pooled standard error of the mean (SEM), and a probability level of  $P < 0.05$  was considered significant.

## Results and Discussion

### Growth Performance

The supplementation of broiler diets with *S. herbacea* extracts led to a significant linear effect in BWG on d 22–35, and d 1–35 ( $P < 0.05$ ), and the BWG in broilers provided with the *S. herbacea* treatments was greater ( $P < 0.05$ ) than in broilers provided with the CON treatment. In addition, a linear decrease was observed in FCR on d 22–35, and d 1–35 ( $P < 0.05$ ), and the broilers provided with the *S. herbacea* treatment had lower FCR ( $P < 0.05$ ) than broilers provided with the CON treatment. However, there were no significant differences in BWG and FCR among the *S. herbacea* treat-

ments. Furthermore, there were no significant differences in total feed intake (TFI) and water intake among treatments (Table 2). In the current study, the *S. herbacea* supplemented groups had higher BWG and lower FCR compared with the control group. The same results were observed by Al-Batshan *et al.* (2008), who reported that SM (*S. bigelovii* Torr meal) was effective in reducing FI, and hence BWG and final BW in broilers. Mohammadi *et al.* (2015) reported that laying hens receiving 1 cc and 5 cc of *S. herbacea* extracts per liter of drinking water had higher egg production and a lower egg breaking rate, with no significant differences between the two *S. herbacea* treatments. In contrast, Sarker *et al.* (2010) reported that BWG, FCR, and FI of broiler chicks were not significantly affected by *S. herbacea* meal (0.5%, 1.0%) when supplemented in feed. Kim *et al.* (2006) reported that feeding rats normal diets containing 2% enzyme-treated *S. herbacea* extracts had no significant effects on BWG, FI, and food efficiency ratio in rats. The different results between *S. herbacea* meal and *S. herbacea* extracts were presumably due to the presence of some anti-nutritional factors such as saponin in *S. herbacea* meal (Glenn *et al.*, 1992), which can reduce surface tension and inhibit chymotrypsin (Birk and Peri, 1980). In view of the current limited research results, we speculated that adding *S. herbacea* to water might have more positive effects on broilers than adding *S. herbacea* to feed. In the current study, there was no significant difference between *S. herbacea* diets, so, in terms of growth performance, the appro-

Table 2. The effect of *S. herbacea* on growth performance in broiler chicks<sup>1,2</sup>

Items	CON	SAL1	SAL2	SAL3	SEM	P-value	
						Linear	Quadratic
1–7 d							
BWG, g	172.8	177.3	173.5	175.6	3.83	0.79	0.77
FI, g	184.0	180.1	182.8	176.6	6.17	0.46	0.85
FCR	1.070	1.018	1.058	1.010	0.04	0.45	0.96
Water intake ml/bird	314.8	312.5	315.4	315.8	10.73	0.88	0.86
8–21 d							
BWG, g	566.8	580.1	589.0	587.6	8.51	0.07	0.39
FI, g	812.0	795.2	798.6	793.3	17.82	0.51	0.75
FCR	1.434	1.374	1.358	1.352	0.04	0.12	0.47
Water intake ml/bird	1421.0	1416.5	1413.9	1415.7	31.55	0.89	0.92
22–35 d							
BWG, g	863.5 <sup>b</sup>	907.9 <sup>a</sup>	907.0 <sup>a</sup>	908.3 <sup>a</sup>	12.99	0.03	0.11
FI, g	1555.8	1530.2	1529.9	1526.8	20.64	0.35	0.59
FCR	1.803 <sup>a</sup>	1.689 <sup>b</sup>	1.688 <sup>b</sup>	1.687 <sup>b</sup>	0.04	0.04	0.13
Water intake ml/bird	2769.3	2743.6	2748.5	2750.7	37.04	0.76	0.71
1–35 d							
TBWG, g	1602. <sup>b</sup>	1665.2 <sup>a</sup>	1669.4 <sup>a</sup>	1671.5 <sup>a</sup>	11.21	<0.01	0.01
TFI, g	2551.9	2505.4	2511.5	2496.7	27.70	0.21	0.57
TFCR	1.593 <sup>a</sup>	1.505 <sup>b</sup>	1.505 <sup>b</sup>	1.494 <sup>b</sup>	0.02	<0.01	0.07
Water intake ml/bird	4505.1	4472.6	4477.8	4482.2	49.33	0.77	0.71

<sup>1</sup> Abbreviation: CON, basal diet (without *S. herbacea*); SAL1, 1 cc/liter *S. herbacea*; SAL2, 5 cc/liter *S. herbacea*; SAL3, 10 cc/liter *S. herbacea*.

<sup>2</sup> Each mean represents eight replicates with 17 broiler chicks/replicate ( $n = 136$ /treatment).

<sup>a-c</sup> Means in the same row with different superscripts differ ( $P < 0.05$ ).

Table 3. The effect of *S. herbacea* on meat quality and relative organ weight in broiler chicks<sup>1,2</sup>

Items	CON	SAL1	SAL2	SAL3	SEM	P-value	
						Linear	Quadratic
pH value	5.32	5.29	5.28	5.28	0.07	0.37	0.77
Breast muscle color							
Lightness (L*)	57.85	58.04	58.12	58.74	1.18	0.63	0.85
Redness (a*)	15.83	15.44	14.97	13.77	0.57	0.04	0.52
Yellowness (b*)	16.05	15.47	15.75	15.86	0.62	0.94	0.63
WHC, %	56.19	58.56	58.14	58.11	2.66	0.63	0.62
Drip loss, %							
1 d	2.39	2.13	1.98	2.06	0.33	0.60	0.25
3 d	5.77	5.39	5.63	5.25	0.46	0.61	0.32
5 d	9.16	8.77	8.96	8.60	0.47	0.90	0.96
7 d	14.92	14.48	14.33	14.39	0.67	0.06	0.53
Relative organ weight, %							
Liver	4.67	5.21	4.87	4.89	0.30	0.81	0.42
Spleen	0.25	0.26	0.27	0.25	0.02	0.85	0.68
Bursa of Fabricius	0.23	0.23	0.23	0.24	0.04	0.90	0.93
Breast muscle	13.45	13.73	14.57	14.88	1.07	0.30	0.91
Abdominal fat	29.55 <sup>a</sup>	27.20 <sup>ab</sup>	22.09 <sup>bc</sup>	21.29 <sup>c</sup>	1.73	<0.01	0.66
Gizzard	2.08	2.15	2.19	2.40	0.14	0.15	0.61

<sup>1</sup> Abbreviation: CON, basal diet (without *S. herbacea*); SAL1, 1 cc/liter *S. herbacea*; SAL2, 5 cc/liter *S. herbacea*; SAL3, 10 cc/liter *S. herbacea*.

<sup>2</sup> Each mean represents eight replicates with two broiler chicks/replicate ( $n=16/\text{treatment}$ ).

<sup>a-c</sup> Means in the same row with different superscripts differ ( $P<0.05$ ).

appropriate dose of the *S. herbacea* extract might be 1 cc/L. This is similar to results from our previous study (Mohammadi *et al.*, 2015), in which we reported that there were no significant differences between the *S. herbacea* treatments (1 cc/L, 5 cc/L). We believe that the doses of *S. herbacea* may have different effects on different animals, and the effects of adding different amounts of *S. herbacea* may not be significant in poultry. In addition, according to the results of Kim *et al.* (2015), different conditions and different doses of *S. herbacea* extracts affect BW differently in rats. Kim *et al.* (2007) also found that red *S. herbacea* extracts may serve as useful natural anti-oxidants, along with green *S. herbacea* extracts. To summarize, we believe that the effects of *S. herbacea* on growth performance are complex. Different doses of *S. herbacea*, different forms of *S. herbacea*, and different experimental animals produce different results. The effects of *S. herbacea* have not been extensively in birds, but several studies report that *S. herbacea* has positive effects on human health (Kim *et al.*, 2006; Tikhomirova *et al.*, 2008). Therefore, further studies are needed to explore the mechanism of the effects of *S. herbacea* on performance in broilers.

#### Meat Quality and Relative Organ Weight

Supplementing broilers with *S. herbacea* extracts did not significantly affect the organ weights of chicken liver, breast muscle, gizzard, or spleen, and no statistical differences were found in pH value, WHC, and drip loss (Table 3). Redness of breast muscle color and relative abdominal fat weight decreased linearly with increasing *S. herbacea* supplementation ( $P<0.05$ ). No significant differences ( $P>0.05$ ) were observed between the treatments with regards to redness (a\*),

while broilers provided with the SAL3 treatment had the lowest abdominal fat among the treatments. A similar result was found by Bostam *et al.* (2017), who demonstrated that abdominal fat was substantially reduced in fermented *S. herbacea* and *Houttuynia cordata* Thunb. (FSH) supplemented broilers relative to controls ( $P<0.05$ ). A number of studies related to *S. herbacea* have been conducted to explore new compounds from natural sources that may assist in controlling obesity in humans, but these obesity related effects are not well studied in birds. *S. herbacea* contains some anti-obesity components, such as anti-oxidative and matrix metalloproteinase inhibitory activities of glucopyranosides A and B (Kim and Park, 2004; Kong *et al.*, 2008). Kong *et al.* (2012) reported that findings from their study should emphasize the nutraceutical value of *S. herbacea*-derived glucopyranosides as potent anti-obesity agents via alleviation of lipid accumulation. Kim *et al.* (2014) demonstrated that *S. herbacea* water extracts might have an anti-adipogenic effect via enhancement of TNF- $\alpha$  production, which causes de-differentiation and inhibits lipid accumulation in adipocytes. Furthermore, Sarker *et al.* (2010) found that, from the first day till the eighth week, broiler chickens fed a 1.0% SHP (*S. herbacea* probiotics) diet had lower ( $P<0.05$ ) crude fat compared with the broiler chickens fed a control diet, although there were no statistical differences in abdominal fat weights. To date, the mechanism through which *S. herbacea* resulted in a lower level of abdominal fat is not fully explained. We believe it may be due to the fact that *S. herbacea* possesses anti-oxidative, anti-inflammatory, anti-hyperglycemic, and anti-hyperlipidemic characteristics

Table 4. The effect of *S. herbacea* on excreta microflora (log10 cfu/g of wet digesta) in broiler chicks<sup>1,2</sup>

Items log10 cfu/g	CON	SAL1	SAL2	SAL3	SEM	P-value	
						Linear	Quadratic
<i>Lactobacillus</i>	7.58	7.42	7.45	7.44	0.1	0.14	0.19
<i>E. coli</i>	6.44	6.27	6.33	6.20	0.1	0.10	0.80

<sup>1</sup> Abbreviation: CON, basal diet (without *S. herbacea*); SAL1, 1 cc/liter *S. herbacea*; SAL2, 5 cc/liter *S. herbacea*; SAL3, 10 cc/liter *S. herbacea*.

<sup>2</sup> Each mean represents eight replicates with four broiler chicks/replicate ( $n=32$ /treatment).

Table 5. The effect of *S. herbacea* on noxious gas emission in broiler chicks<sup>1,2</sup>

Items	CON	SAL1	SAL2	SAL3	SEM	P-value	
						Linear	Quadratic
NH <sub>3</sub> , mg/h/bird	24.8	22.6	21.1	21.2	1.6	0.14	0.53
R.SH	0.8	0.5	0.3	0.8	0.3	0.88	0.32
H <sub>2</sub> S	1.25	0.50	0.75	1.00	0.4	0.79	0.24
Acetic acid	3.2	2.6	2.4	2.6	0.4	0.30	0.30

<sup>1</sup> Abbreviation: CON, basal diet (without *S. herbacea*); SAL1, 1 cc/liter *S. herbacea*; SAL2, 5 cc/liter *S. herbacea*; SAL3, 10 cc/liter *S. herbacea*.

<sup>2</sup> Each mean represents eight replicates with 17 broiler chicks/replicate ( $n=136$ /treatment).

(Im *et al.*, 2003; Seo *et al.*, 2004; Lee *et al.*, 2006; Kim *et al.*, 2015). More studies are needed to evaluate the exact effect of *S. herbacea* on abdominal fat in broilers. In our study, a linear decrease ( $P<0.05$ ) was observed in redness between the treatments, while there were no significant differences between the treatments with regards to redness of breast meat, yellowness, and lightness. Similar results were reported by Joo and Choi (2014), who found that, as the content of *S. herbacea* powder increased (0, 1, 2, and 3%), L\* and a\* values decreased, while b\* values increased in pork patties. In the current experiment, the broilers provided with *S. herbacea* treatments had slightly paler meat compared with that of the CON broilers. In broilers, pale meat is often associated with low redness values (Qiao *et al.*, 2001). Allen *et al.* (1997) reported that darker broiler breast meat fillets have a shorter shelf-life than lighter breast fillets. The dark fillets had significantly ( $P<0.05$ ) lower lightness values (L\*), higher redness values (a\*), lower yellowness values (b\*), and higher pH values. Similar results were found by Sarker *et al.* (2010), who reported that the thiobarbituric acid (TBA) content of fresh broiler meat was significantly lower in the 0.5% SHP group, and addition of SHP (0.5% and 1.0%) significantly reduced the lipid compared to controls. As such, we believe that the addition of 10 cc/L of *S. herbacea* extracts in drinking water, and 0.5% *S. herbacea* in feed might have enough positive effects on meat storage period extension. Thus, we believe that the addition of *S. herbacea* extracts to drinking water provided to broiler chicks might improve sensory performance, meat quality, as well as lengthen shelf-life.

#### Excreta Microbial Populations and Noxious Gas Emission

The results of excreta microbial analysis are shown in Table 4. There were no significant differences in any measures between treatments. There are very few studies on the effects of *S. herbacea* on *Lactobacillus* and *E. coli*. To the best of our knowledge, very few studies have focused on the antimicrobial activity of *S. herbacea*. Lellau and Liebezeit (2003), reported a high activity of *S. herbacea* against fungi, yeasts, and algae. Essaidi *et al.* (2013) and Rad *et al.* (2014) suggested that the antimicrobial activity of alcohol (methanol and ethanol) extracts of *S. herbacea* is the result of a synergic or additive effect of several compounds present in this plant. They also reported the presence of several phenolic compounds in *S. herbacea*, which could have antimicrobial activity. This activity could be related to fatty acids and osmotic compounds (betaine), tannins, oils, gums, flavonoids, saponins, and essential oil precursors for the synthesis of complex chemical materials (Chandrasekaran *et al.*, 2008; Kim *et al.*, 2010; Viji and Murugesan, 2010; Essaidi *et al.*, 2013; Rad *et al.*, 2013). Kim *et al.* (2011a) suggested that *S. herbacea* did not influence the growth of microorganisms, which is in agreement with the results of the current study. Kim *et al.* (2011a) reported that *S. herbacea* may not be a growth factor for any specific microorganism, and pointed that *S. herbacea* did not influence the *Lactobacillus* and the *Lactobacillus* family. These different results might be due to the different extraction methods, different places of origin, different animals, and different levels of *S. herbacea*. Thus, based on these limited studies, we believe that *S. herbacea* may not have had an influence on *Lactobacillus* and *E. coli* in broilers. Meanwhile, further studies are needed to evaluate

the exact effect of *S. herbacea* on excreta microbial communities in broilers.

Noxious gas concentrations of broilers provided with the experimental treatments are presented in Table 5. No statistical differences were found in R.SH, NH<sub>3</sub>-N, H<sub>2</sub>S and acetic acid concentrations between treatments. To date, few studies have focused on the effect of *S. herbacea* dietary supplementation on the noxious gas emissions of broilers. The current experimental results indicate that *S. herbacea* had no effect on noxious gas emissions in broilers. More studies are needed to explore the effects of *S. herbacea* on noxious gas emission in broilers.

In conclusion, the results demonstrate that the use of *S. herbacea* (5 cc/L, 10 cc/L) led to positive effects on BWG and FCR of broilers and modified the carcass quality by decreasing the rate of abdominal fat. This indicates that supplementing broilers with 5 cc/L of *S. herbacea* would be sufficient to achieve an optimal response in meat quality and growth performance. Thus, we believe that *S. herbacea* can be used as an alternative to antibiotic growth promoters and improve the safety of poultry products. More research is needed to understand the mode of action by which performance is improved, and to clarify the effects of *S. herbacea* on blood metabolites, carcass characteristics, and broiler performances.

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