Effects of dietary *Achyranthes japonica* extract supplementation on the growth performance, total tract digestibility, cecal microflora, excreta noxious gas emission, and meat quality of broiler chickens

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ABSTRACT The present study was investigated the effects of dietary *Achyranthes japonica* extract (**AJE**) supplementation on the growth performance, total tract digestibility, cecal microflora, excreta noxious gas emission, breast meat quality, and organ weight in broiler chickens. In total, 640 Ross × Ross male broiler chickens (1-day-old) were randomly distributed into 4 dietary treatments with 10 replicate cages (16 birds/replicate) per treatment group for 5 wk. The dietary treatments included a control basal diet without AJE, and diets with 0.025, 0.05, or 0.1% AJE. Body weight gain, feed intake, and feed conversion improved linearly with the supplementation of AJE over the experimental period (days 1 to 35) ($P < 0.05$). Dietary AJE supplementation caused a significant increase in the apparent total tract digestibility of dry matter and nitrogen (linear, $P < 0.05$). The cecal *Lactobacillus*, *E. coli*, and *Salmonella* counts were linearly affected with increasing dietary AJE supplementation ($P < 0.05$). With increasing levels of AJE, excreta ammonia gas concentration showed a linear decrease ($P < 0.05$). The breast muscle weight linearly increased, along with a decrease in the abdominal fat weight, in treatment groups fed with AJE ($P < 0.05$). These results indicate that dietary addition with increasing AJE linearly improved growth performance, total tract digestibility, cecal microflora, excreta ammonia gas emission, and abdominal fat weight in broiler chickens.

Key words: abdominal fat, *Achyranthes japonica*, ammonia gas emission, broiler productivity, intestinal microorganism

INTRODUCTION

Medicinal herb plants have been used to treat a variety of human diseases in Korea for centuries, owing to their excellent therapeutic effects and low toxicity. These medicinal herb plants have been used as not only medicines but also as foods, flavors, pigments, and cosmetics for thousands of years in many countries worldwide (Dahanukar et al., 2000; Djeridane et al., 2006). The positive effects of medicinal herb plants suggest the presence of a wide variety of phytochemicals such as phenolics, flavonoids, and tannins, which can play an important role in the prevention and treatment of diseases, owing to their anti-inflammatory, antimicrobial and antioxidant effects (Cho et al., 2003; Choi et al., 2017; Upadhaya and Kim, 2017). In poultry, various medicinal herb plants have contributed to the improvement of productivity, health immunity, nutrient digestibility, excreta noxious gas emission, meat quality, and stabilization of the microflora in the intestinal tract (Lee et al., 2003; Marcinčáková et al., 2011; Gong et al., 2014; Li et al., 2015; Zeng et al., 2015).

Achyranthes japonica is a perennial member of the *Achyranthes* genus in the Amaranthaceae family, which has a wide distribution in East Asian countries including Korea, China, and Japan, where it is mainly used in traditional medicines or folk remedies. The root of *A. japonica* contains multiple active components, including saponin, triterpenoids, phytoecdysteroid, 20-hydroxyecdysone, and inokosterone (Liu et al., 2008; Lee et al., 2012; Al-Mijan et al., 2018). It is used in the traditional medicine of Korea to treat hypertension, rheumatism, osteoarthritis, and analgesic and diuretic. Both in vitro and in vivo experiments have shown that *A. japonica* extracts also have various physiological effects, including anti-allergic, anti-inflammatory, antioxidant, arthritis alleviation, hepato-protective, and anti-cancer properties (Jung et al., 2007; Kim and Park, 2010; Bang et al., 2012; Jang et al., 2012).

Although *A. japonica*-induced pharmacological activities with apparent effect on human or various experimental animals have been widely reported, there has been no extensive study on the effect of *A. japonica* extract (**AJE**) on broiler chickens as a feed additive. Our hypothesis was that *A. japonica* extracts selected according to various physiological effects might

contain multiple active components that could be very effective in maximizing productive performance. Therefore, in this study, we attempted to evaluate the use of *A. japonica* as a natural growth promoter in broiler chicken diets. The evaluation included monitoring of physiological changes in growth performance, total tract digestibility, cecal microflora, excreta noxious gas emission, meat quality, and organ weight.

MATERIALS AND METHODS

The experiment received prior approval from the Animal Protocol Review Committee of Dankook University (Ethics Approval Number: DK-1-1728).

Animals and Experimental Design

In a 35-d trial, a total of 640 1-day-old male Ross 308 broiler chicks were randomly distributed into four experimental diets with similar average body weight (41 ± 0.5 g). The dietary treatments were: 1) CON, basal diet, 2) T1, CON + 0.025% AJE, 3) T2, CON + 0.05% AJE, and 4) T3, CON + 0.1% AJE. There were 10 replicates per treatment, each with 16 birds. All the birds were housed in stainless steel cages (1.75 m × 1.55 m) with 3 floors, and light was provided for 24 h during days 1 to 7, and then for 22 h until day 35. The temperature of the room was maintained at 33°C for the first 5 d, and decreased to 22°C until the end of the experiment. Three diets were used: a starter diet offered from days 1 to 7, a grower diet offered from days 8 to 21, and a finisher diet provided from days 22 to 35. The chicks were given free access to water and mash feed during the entire experiment. All diets were formulated according to the requirements recommended by the NRC (1994) and provided in mashed form (Table 1). AJE was included in the diet by replacing the same amount of corn.

Preparation of AJE

The AJE used in our study was obtained from a commercial company (Synergen Inc., Bucheon, Korea). The manufacturing process of the AJE is briefly described here. *Achyranthes japonica* cultivated in Korea were purchased. The roots of *A. japonica* were washed three times with clean water, and then powdered with a mill (IKAM20; IKA, Staufen, Germany). The dried sample was extracted with distilled water at 80°C, and was then refluxed for 6 h to obtain an initial extract. The residues were extracted with distilled water (1:5) at 80°C for 2 h. The extract solution was filtered under low temperature by a high-velocity centrifugal machine. The useful parts were collected by column and eluted with ethanol. After cooling to room temperature (25°C) and filtering (Whatman No. 2; Whatman Ltd., Kent, UK), the samples were vacuum-dried at a temperature below 40°C. The extracts were completely dried in a

Table 1. Ingredient composition of experimental diets as-fed basis.

Ingredient, %	Starter	Grower	Finisher
Corn	54.19	55.38	56.77
Soybean meal	33.80	26.1	18.23
Canola meal	5.00	10.0	15.0
Soybean oil	2.10	3.62	5.07
MDCP[1]	–	1.28	1.12
DCP[2]	1.70	–	–
Limestone	1.15	1.34	1.22
L-lysine	0.50	0.65	0.81
DL-Methionine	0.46	0.47	0.52
L-Threonine	0.20	0.25	0.32
L-Tryptophan	–	0.01	0.04
NaHCO$_3$	0.10	0.10	0.10
Salt	0.30	0.30	0.30
Vitamin premix[3]	0.20	0.20	0.20
Mineral premix[4]	0.20	0.20	0.20
Choline	0.10	0.10	0.10
Nutrient composition			
ME, kcal/kg	3040	3120	3240
CP, %	22.7	21.4	20.3
Lys, %	1.48	1.39	1.31
Met + Cys, %	1.08	0.99	0.94
AP, %	0.48	0.44	0.41
Ca, %	0.96	0.87	0.81

[1]Monodicalcium phosphate.
[2]Dicalcium phosphate.
[3]Provided per kg of complete diet: 11,025 IU vitamin A; 1,103 IU vitamin D$_3$; 44 IU vitamin E; 4.4 mg vitamin K; 8.3 mg riboflavin; 50 mg niacin; 4 mg thiamine; 29 mg d-pantothenic; 166 mg choline; 33 μg vitaminB$_{12}$.
[4]Provided per kg of complete diet: 12 mg Cu (as CuSO$_4$•5H$_2$O); 85 mg Zn (asZnSO$_4$); 8 mg Mn (as MnO$_2$); 0.28 mg I (as KI); 0.15 mg Se (as Na$_2$SeO$_3$•5H$_2$O).

freeze-drier. AJE contains total flavonoid (1.15 mg/g), total polyphenol (4.26 mg/g) and saponin (0.47 mg/g).

Growth Performance

Body weight (**BW**) and feed intake (**FI**) per cage were recorded on days 7, 21, and 35, and the feed conversion ratio (**FCR**) was calculated using as the FI divided by body weight gain (**BWG**).

Apparent Total Tract Digestibility

At the end of the experiment, 0.2% chromic oxide (Cr_2O_3) was added to the diets as an indigestible marker, for determining the apparent total tract digestibility (**ATTD**). Excreta samples were collected for 3 d, and dried in a 60°C oven for 72 h. The diets and excreta samples were analyzed for gross energy (Parr Instrument, Moline, IL, USA), dry matter (**DM**; methods 934.01), and nitrogen (methods 968.06) using standard procedures of AOAC (2000). Chromium concentration was determined by atomic absorption spectrophotometry (UV-1201, Shimadzu, Kyoto, Japan). The equation for calculating digestibility was as follows: ATTD (%) = $(1-((Nf \times Cd)/(Nd \times Cf))) \times 100$, where Nf = nutrient concentration in feces (% DM), Nd = nutrient concentration in diet (% DM), Cf = chromium concentration in feces (% DM), and Cd = chromium concentration in diet (% DM).

Cecal Microflora Population

One gram of collected sample from the cecum was diluted with 9 mL of sterile peptone water and mixed for 1 min on a vortex stirrer. Samples were serially blended from 10^{-1} to 10^{-6}, and were injected by 50 μL in 3 selective agar media as follows; Lactobacilli MRS agar (Difco Laboratories, Detroit, MI, USA) for *Lactobacillus* spp., coliform bacteria for MacConkey agar (Difco Laboratories, Detroit, MI, USA) and *Salmonella-Shigella* (SS) agar (Difco Laboratories, Detroit, MI, USA) for *Salmonella.* Plates were then incubated aerobically at 37°C for 24 h (MacConkey agar and SS agar) or anaerobically at 37°C for 24 h (Lactobacilli MRS agar). The viable colonies of the respective bacteria were counted and expressed as the log 10 of colony-forming units (cfu) g^{-1} of cecal content.

Excreta Noxious Gas Emission

At the end of the experiment, fresh excreta samples (300 g) were collected from each cage for 4 d for determining ammonia, hydrogen sulfide, and total mercaptan. The subsamples of excreta were taken and stored in 2-L sealed plastic contaniners in duplicate for 5 d at ambient temperature (20 to 24°C). After the fermentation period, a gas sampling pump kit (model GV-100S, Gastec Corp., Tokyo, Japan) was used for gas detection. The concentrations of ammonia, hydrogen sulfide, and total mercaptan were measured by a detector tube within the scope of 0.5 to 78 ppm (No. 3 L, detector tube; Gastec Corp.), 0.1 to 4 ppm (No. 4LT, detector tube; Gastec Corp.), and 0.1 to 8 ppm (No. 70 L, detector tubes; Gastec Corp.), respectively. One hundred milliliters of headspace air was sampled at approximately the upper 2 inches of the excreta surface.

Breast Meat Quality and Relative Organ Weight

The pH values of raw breast meat at 24 h postmortem were measured using a digital pH meter (Testo 205, Lenzkirch, Germany) after blending 10 g of finely homogenized sample with 90 mL of double-distilled water. Color values of the breast meat were measured in three replicates using a Minolta colorimeter (CR-300, Tokyo, Japan) calibrated with a standard white plate and recorded as L*, a*, and b* values for lightness, redness, and yellowness, respectively. To estimate the cooking loss, raw meat samples were packed into Cryovac® Cook-In Bags after weighing, and cooked in a water bath at 100°C for 30 min. Samples were cooled at room temperature for 1 h and reweighed. The water-holding capacity (**WHC**) was determined following the method of Kristensen and Purslow (2001). Five grams of meat sample was heated to 70°C in a water bath for 30 min. The samples were then cooled using ice and subsequently centrifuged at 4°C at $1000 \times g$ for 10 min. WHC (%) was calculated as the ratio of weight loss of the sample during centrifugation, to that of the original liquid. Cooking loss was calculated as the weight difference between the initial raw and final cooked samples. Drip loss (%) was measured for 5 cm × 5 cm cuts of breast meat, which were weighed, hung in a zipper bag, and stored at 4°C. After storage, the moisture on the surface of the meat slices was carefully removed and weighed at days 1, 3, 5, and 7 from the date of sample collection. The initial and final weight of each sample was used to calculate drip loss.

The liver, spleen, and bursa of Fabricius, breast meat, abdominal fat, and gizzard were removed and weighed. Organ weights, breast meat and abdominal fat were expressed as a percentage of live BW.

Statistical Analysis

All data were analyzed as a randomized complete block design using the general liner model procedures of SAS program (SAS Institute Inc., 2014). Linear, quadratic, and cubic polynomial contrasts were performed to determine the effects of different level of AJE on all measurements. Cage was considered as an experimental unit for growth performance, ATTD, and excreta gas concentration. The individual bird was used as the experimental unit for cecal microflora, and meat quality measurements. Variability in data was expressed as the pooled standard error of the mean and a probability less than 0.05 was considered statistically significant.

RESULTS

Growth Performance

Dietary AJE supplementation increased linearly for BWG during days 1 to 7, 22 to 35, and overall (days 1 to 35), as the dietary AJE supplementation increased from 0 to 0.1% ($P < 0.05$). Increasing dietary supplementation of AJE had a positive linear effect on the FI and FCR during days 1 to 7, 8 to 21, and overall ($P < 0.05$) (Table 2).

Apparent Total Tract Digestibility

ATTD of DM and nitrogen increased linearly in broiler chickens fed on diets supplemented with 0 to 0.1% AJE ($P < 0.05$). No treatment effects were observed on the energy retention (Table 3).

Cecal Microbial Count

Lactobacillus counts showed a significant linear increase with increasing dietary AJE supplementation

Table 2. Effect of dietary *Achyranthes japonica* extract (AJE) supplementation on growth performance in broilers.

	AJE (%)					P-value		
Items	0	0.025	0.05	0.1	SEM[4]	Linear	Quadratic	Cubic
D 1 to 7								
BWG[1], g	146[b]	154[a,b]	155[a,b]	159[a]	4.28	0.0184	0.9738	0.5689
FI[2], g	181[a]	177[a,b]	175[b]	175[b]	4.21	0.0055	0.1549	0.9370
FCR[3]	1.240[a]	1.149[b]	1.129[b]	1.101[b]	0.023	0.0005	0.3825	0.6262
D 8 to 21								
BWG, g	678	686	690	693	8.88	0.0628	0.6967	0.9037
FI, g	1036[a]	972[a,b]	972[a,b]	973[b]	9.20	0.0429	0.6737	0.4511
FCR	1.528[a]	1.417[a,b]	1.409[a,b]	1.404[b]	0.017	0.0106	0.5841	0.5889
D 22 to 35								
BWG, g	983[b]	1015[a,b]	1016[a,b]	1020[a]	9.08	0.0297	0.4152	0.5230
FI, g	1698	1697	1698	1696	11.76	0.8896	0.9788	0.9210
FCR	1.727	1.672	1.671	1.663	0.012	0.0727	0.5264	0.5974
D 1 to 35								
BWG, g	1807[b]	1855[a]	1861[a]	1872[a]	12.22	0.0004	0.3566	0.4418
FI, g	2915[a]	2846[a,b]	2845[a,b]	2844[b]	16.25	0.0262	0.5608	0.4844
FCR	1.613[a]	1.534[b]	1.529[b]	1.519[b]	0.015	0.0001	0.2590	0.2850

[1]Body weight gain.
[2]Feed intake.
[3]Feed conversion ratio.
[4]Standard error of means.
[a,b] Means in the same row with different superscripts differ ($P < 0.05$).

Table 3. Effect of dietary *Achyranthes japonica* extract (AJE) supplementation on nutrient digestibility in broilers.

	AJE (%)					P-value		
Items, %	0	0.025	0.05	0.1	SEM[1]	Linear	Quadratic	Cubic
Dry matter	72.7[b]	74.1[a,b]	74.2[a,b]	74.7[a]	0.60	0.0369	0.4524	0.5283
Nitrogen	70.4[b]	72.1[a,b]	72.5[a]	72.6[a]	0.69	0.0307	0.2386	0.7450
Energy	71.5	72.9	73.1	73.1	0.76	0.1371	0.3510	0.7985

[1]Standard error of means.
[a,b]Means in the same row with different superscripts differ ($P < 0.05$).

Table 4. Effect of dietary *Achyranthes japonica* extract (AJE) supplementation on cecal microflora in broilers.

	AJE (%)					P-value		
Items, ($\log_{10}$cfu/g)	0	0.025	0.05	0.1	SEM[1]	Linear	Quadratic	Cubic
Lactobacillus	7.09[a]	7.29[a,b]	8.26[b]	8.23[b]	0.10	0.0070	0.3649	0.6241
E. coil	6.40[a]	6.13[a,b]	5.81[a,b]	5.22[b]	0.14	0.0074	0.7133	0.6015
Salmonella	3.91[a]	3.80[a,b]	3.71[a,b]	2.69[b]	0.17	0.0199	0.5240	0.9003

[1]Standard error of means.
[a,b] Means in the same row with different superscripts differ ($P < 0.05$).

($P < 0.05$). *E. coli* and *Salmonella* counts, on the other hand, decreased linearly with increasing levels of AJE ($P < 0.05$) (Table 4).

Excreta Noxious Gas Emissions

Excreta ammonia emissions decreased as dietary AJE supplementation increased (linear, $P < 0.05$). However, AJE supplementation did not affect the total mercaptan or hydrogen sulfide emissions of broiler chickens fed with different levels of AJE (Table 5).

Meat Quality and Organ Weight

There were no statistically significant differences in pH, color (L^*, a^*, b^*), cooking loss, WHC, or drip loss of breast meat among the 4 treatment groups. Relative weights of most organs (liver, spleen, gizzard, and bursa of Fabricius) were not significantly affected by dietary supplementation of AJE. However, the weights of breast muscle and abdominal fat were significantly different among the four groups. Birds fed with AJE showed significantly higher muscle weights and lower

Table 5. Effect of dietary *Achyranthes japonica* extract (AJE) supplementation on excreta gas emission in broilers.

Items, ppm	AJE (%)				SEM[1]	P-value		
	0	0.025	0.05	0.1		Linear	Quadratic	Cubic
Ammonia	37.8[a]	26.5[b]	26.3[b]	26.2[b]	1.31	0.0019	0.5495	0.9479
Hydrogen sulfide	2.1	1.8	1.9	1.8	0.48	0.1510	0.3451	0.7353
Mercaptan	2.7	2.4	2.3	2.5	0.52	0.1939	0.4656	0.8192

[1]Standard error of means.
[a,b]Means in the same row with different superscripts differ ($P < 0.05$).

Table 6. Effect of dietary *Achyranthes japonica* extract (AJE) supplementation on meat quality and organ weight in broilers.

Items	AJE (%)				SEM[1]	P-value		
	0	0.025	0.05	0.1		Linear	Quadratic	Cubic
pH value	5.35	5.42	5.47	5.48	0.05	0.0575	0.5821	0.8783
Breast muscle color								
Lightness (L[*])	51.17	49.39	49.31	49.31	0.68	0.1910	0.0777	0.4653
Redness (a[*])	11.04	10.92	10.85	10.76	0.28	0.4608	0.9547	0.9570
Yellowness (b[*])	9.61	9.53	9.48	9.50	0.15	0.5897	0.7428	0.9602
WHC[2] %	47.93	48.66	48.51	48.76	1.41	0.7131	0.8626	0.8404
Cooking loss	18.73	18.58	18.56	18.52	0.44	0.7375	0.9093	0.9337
Drip loss, %								
d 1	4.44	4.31	4.29	4.22	0.10	0.1433	0.7812	0.7194
d 3	7.21	7.06	7.11	7.08	0.05	0.1738	0.2297	0.2656
d 5	9.63	9.54	9.50	9.43	0.09	0.1294	0.9475	0.8753
d 7	12.19	12.08	12.04	12.02	0.07	0.0997	0.5699	0.8822
Relative organ weight, %								
Breast muscle	17.56[b]	18.84[a,b]	19.11[a]	19.26[a]	0.20	0.0074	0.6264	0.9751
Liver	2.57	2.65	2.64	2.67	0.05	0.1980	0.5773	0.4614
Bursa of Fabricius	0.14	0.13	0.14	0.13	0.01	0.8506	0.8662	1.0000
Abdominal fat	2.17[a]	1.71[b]	1.60[b]	1.68[b]	0.12	0.0013	0.2107	0.5426
Spleen	0.17	0.17	0.16	0.16	0.01	0.5857	0.8834	0.9130
Gizzard	0.97	0.93	0.92	0.91	0.02	0.0930	0.5306	0.5055

[1]Standard error of means. [2]Water-holding capacity.
[a,b]Means in the same row with different superscripts differ ($P < 0.05$).

abdominal fat weights as the dietary levels of AJE increased (linear, $P < 0.05$) (Table 6).

DISCUSSION

A. japonica has various physiological and biochemical functions in the body, owing to its active components, a variety of antioxidant phytochemicals and bioactive compounds (Liu et al., 2008; Lee et al., 2012). However, there is a lack of data on the efficacy of *A. japonica* in poultry. Therefore, we attempted to evaluate the role of AJE as a natural growth promoter for broiler chickens in this study. The findings of present study demonstrated that AJE enhanced the BWG and FCR, indicating the efficient utilization of feed. The effect of plant extracts on the broilers performance has been investigated in the several studies. In a trial reviewed by Alçiçek et al. (2003), a significant improvement in BWG and FCR was reported when the broilers were fed a diet containing a combination of plant extracts, compared with birds fed a control diet or one containing antibiotics. Previous reports carried out with a plant extract mixture (Petrolli et al., 2012), or a blend (Zhang et al.,

2005) as a substitute for an antibiotic growth promoters also showed similar FCR results as the diet containing antibiotics. Park et al. (2014) indicated that supplementation of broiler diet with medicinal herb plant extracts (*Saposhnikovia divaricate*, *Lonicera japonica*, and *Chelidonium majus*) may potentially improve BWG, blood cell profiles, and meat quality. The mechanism of AJE action in enhancing growth performance of poultry has not been clearly elucidated yet, but our results are in agreement with previous investigations on rats (Tahiliani and Kar, 2000), piglets (Chen et al., 2009), and broilers challenged with *E. coli* (Liu et al., 2018), which demonstrates the ability of *Achyranthes* plants extracts to improve the growth performance. According to these authors, *Achyranthes* plants extracts could exhibit antioxidative properties, antimicrobial activity, and immunostimulating effect, and improve nutrient absorption ultimately improving the animal performance. Therefore, it is likely that the improvement of BWG with improving efficiency of feed utilization in AJE-fed broilers in the present study may be due to the bioactive phytochemicals, including saponin and polyphenol compounds.

In this study, the ATTD of DM and nitrogen increased linearly in broilers fed with AJE diets, indicating that greater digestibility is associated with increasing level of AJE in diet. Previous studies on the digestibility of AJE are limited, however, the results of other studies suggest that feeding broiler with plant extracts can enhance nutrient digestibility by stimulating bile secretion, and increasing digestive enzymes of small intestine. Jang et al. (2007) demonstrated that broilers fed diets containing a high dose of plant extract mixtures (carvacrol, cinnamaldehyde, and capsaicin) showed significantly increased activities of pancreatic trypsin and α-amylase, as well as intestinal maltase, compared with the birds fed control and antibiotic diets. Lee et al. (2003) reported that the ileal digestibility in young broilers receiving essential oils (thymol and carvacol) was greater owing to higher amylase activity, compared with control broilers. Some authors have observed that these additives may have a marked beneficial effect on gut morphology as well (Khalaji et al., 2011). They also found that villus height and crypt depth at the jejunum is greater in broilers fed plant extracts compared with control broilers. Improved villus height or villus height-to-crypt depth ratio are usually associated with efficient nutrient absorption and better performance. The other likely reason for the increased digestibility found in AJE groups may be associated with intestinal microflora. It is possible to change the digestibility by changing intestinal microflora. By inhibiting the growth of harmful microorganisms in the intestines, the plant extract gives beneficial microorganisms a superiority in nutrient competition with harmful microorganisms and can improve the nutrient digestibility of broiler chickens. Therefore, the better digestibility of DM and nitrogen observed in AJE-supplemented diets may be related the digestive enzymes, amelioration of intestinal morphology and increase in the beneficial microorganisms in intestine, which enhanced the growth in broiler chickens.

The cecum plays an important role in preventing colonization of pathogens, the detoxification of harmful substances, the recycling of nitrogen, the synthesis of microorganisms of vitamins, the degradation of some carbohydrates, and the absorption of additional nutrients (Clench and Mathias, 1995). The addition of plant preparations were found to reduce the intestinal pH level, and increasing the number of lactic acid bacteria in the ileum and cecal contents is known to significantly decrease the *E. coli* and *C. perfringens* counts (Dalkilic et al., 2005; Vidanarachchi et al., 2006). It has been also reported that favorable microbial community such as lactobacilli and bifidobacteria promote host protective responses against pathogenic microorganisms. Jung et al. (2008) demonstrated in vitro that 0.5% AJE shows high antibacterial effects by inhibiting the growth of the harmful microorganism, *Clostridium difficile*. The addition of *Achyranthes bidentata* extract is also known to promote the growth of *Lactobacillus* and *Bifidobacterium*, suggesting the prebiotic potential of the plant extract (Xie et al., 2018). In an in vivo study, feeding *Achyranthes bidentata* extract resulted in decreased *E. coli and Enterococcus* in the cecum of broiler chickens challenged with *E. coli* (Liu et al., 2018). Chen et al. (2009) reported that various doses of *Achyranthes bidentata* extract supplementation significantly decreased the diarrhea frequency of weaned piglets, indicating the inhibition of gut pathogens. This study also indicated that broiler chickens fed an AJE-supplemented diet led to higher cecal *Lactobacillus* concentration, and lower *E. coli* and *Samonella* concentration. Therefore, the extracts could promote the development of the normal gut microbiota, suppress bacterial pathogens, and lead to healthy intestinal development.

As for excreta noxious gas emission, the excreta ammonia gas content reduced linearly with increasing dietary levels of AJE. Excreta noxious gas emission of animals is associated with intestinal microflora, particularly harmful intestinal bacteria populations (Ferket et al., 2002). A previous study also suggested that excreta noxious gas content is lowered when the microflora in the intestinal tract of broiler chickens is manipulated (Jeong and Kim, 2014).

Excreta noxious gas emission of animal is closely related to nutrient digestibility (Yan et al., 2011a; Jeong and Kim, 2014), because the increased digestibility allows more complete oxidative dissolution of organic substrate in the intestine, consequently reducing the excreta odor and noxious gas levels. In our study, the AJE-supplemented diet led to a better balance of microbiota ecosystem in the cecum and higher nutrient digestibility than the control diet. Therefore, we speculate that the reason for reduction in excreta ammonia gas content may be the result of increased nitrogen digestibility and *Lactobacillus* populations in broiler chicken ceca. There is still a lack of enough information about the usefulness of AJE in poultry, however, findings from the current study support those from previous studies (Hong et al., 2012; Li et al., 2015; Sharma et al., 2017), showing that dietary supplementation with plant extracts or phytogenic feed additive containing saponin and polyphenol decreased the volatile compounds in broiler litters or excreta ammonia gas emission.

The effects of plant extracts have been studied and reported in a variety of meat types including poultry, pork, beef, lamb, and Equidae (O'Grady et al., 2006; Nieto et al., 2010; Toghyani et al., 2010; Yan et al., 2011b; Rossi et al., 2017). However, the benefits of dietary plant extract feeds on carcass and meat quality enhancement are still ambiguous. Some authors have reported the advantages of plant extract feed supplementation on the chemical composition, physicochemical and sensory properties, and carcass grade (Nasir and Grashorn, 2010; Marcinčáková et al., 2011), whereas others have reported no beneficial effects (Koreleski and Swiatkiewcz, 2007; Kim et al., 2016). Our results suggested that AJE had no direct effect on the improvement of meat quality, and the relative organ weights

(liver, bursa of Fabricius, spleen and gizzard) in broilers. However, our results indicate that AJE decreases the abdominal fat percentage and increases breast meat percentage of broilers. Krishnakumari and Priya (2006) reported that *Achyranthes aspera* supplementation reduced the hyperlipidemic and hypercholesteremic conditions in rats fed high oil diets. *Achyranthes aspera* appeared to ameliorate hypercholesteremia probably by decreasing the exogenous cholesterol absorption and increasing the endogenous cholesterol conversion to bile acid. Latha et al. (2011) also suggested that *Achyranthes aspera* significantly reduced the levels of serum total cholesterol, triglycerides, low-density lipoprotein cholesterol, and very-low-density lipoprotein cholesterol, while significantly increasing the levels of high-density lipoprotein cholesterol, in high-fat induced hyperlipidemic rats. This effect was attributed to the presence of saponins, which could prevent intestinal lipid absorption. There is currently a lack of data regarding the effect of AJE on broilers, and previously reported functions of AJE in other species may not be directly comparable. However, the findings of the current study support those from previous research (Krishnakumari and Priya, 2006; Latha et al., 2011), indicating that *Achyranthes* plants could be used as a lipolytic agent. Therefore, AJE inclusion in broiler diet may result in elevated breast meat yields and reduced abdominal fats.

CONCLUSION

The present study showed that feeding AJE to broilers improved their growth performance, DM, nitrogen digestibility, cecal *Lactobacillus* population, and breast meat production, whereas it deceased the cecal *E. coli* and *Salmonella* population, excreta ammonia emission, and abdominal fat. Therefore, these results confirmed the applicability of AJE as a feed ingredient in broiler diets.

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