



Comparative evaluation of *Schisandra chinensis* and *Viscum album* var. *coloratum* (Korean mistletoe) powders on growth performance, carcass characteristics, serum cholesterol profiles, and meat quality of broilers

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ABSTRACT

Experiments were conducted to compare the effects of *Schisandra chinensis* and *Viscum album* var. *coloratum* (Korean mistletoe [KM]) powders on the growth performance, carcass characteristics, serum cholesterol, and meat quality of broilers. Arbor Acres male broiler chicks (240) were fed diets supplemented with 0% (control), 0.5%, and 1% *S. chinensis*, or 0.5% and 1% KM in a completely randomized design. There were no differences among the groups with respect to growth performance or carcass weight and carcass ratio, but there were differences in mortality and relative weights of organs (liver, spleen, and abdominal fat). Diets supplemented with *S. chinensis* or KM led to reductions in total cholesterol (TC), low-density lipoprotein cholesterol (LDLC), and triglyceride levels and an increase in high-density lipoprotein cholesterol (HDLC) level. However, the *S. chinensis* and KM supplements produced no significant differences in proximate composition. Higher levels of *S. chinensis* and KM supplementation led to significantly better lipid oxidation stability and chicken thigh-meat quality (color), but there was no effect on pH and b*(yellowness). Moreover, the addition of KM had greater antioxidant and immune system effects than those of *S. chinensis*. In conclusion, diets with a 1% KM supplement had the greatest effect on reducing mortality and serum cholesterol levels, and increasing the meat quality of broilers.

Key words: Carcass characteristics, Growth performance, Korean mistletoe, *Schisandra chinensis*, Serum cholesterol

Antibiotics not only improve the growth of food-producing animals but also contribute to improving animal welfare and disease prevention (Chowdhury *et al.* 2009, Allen *et al.* 2014). Resistance to antibiotics has been a major issue in the 21st century, and many countries have banned antibiotics because of potential safety issues. The safety of antibiotic residues in food and persistence of antibiotic resistance are the main causes for concern. Thus, alternatives to feeding of antibiotics to animals are urgently required. It is generally accepted that herbal medicine and plant extracts, because of their natural origin, have the ability to serve as biologically active sources of substances to improve animal growth and welfare (Kim 2014).

Schisandra chinensis grows abundantly in moist, shady, and well-drained areas, and it is widely distributed throughout northeastern China, Korea, and Japan (Panossian and Wikman 2008). *S. chinensis* contains lignans, essential oils, citric acid, and malic acid (Sladkovsky *et al.* 2001, Zhang *et al.* 2002, Hwang 2012, Zhong *et al.* 2016). Lignans, in particular, are known as important bioactive compounds that seem to be responsible for the pharmacological properties of *S. chinensis* (Zhang *et al.* 2018). Previous studies have shown that *S. chinensis* may

be useful in increasing immunity and antioxidant effects in broilers (Ma *et al.* 2006, Ko *et al.* 2012).

Mistletoe is a hemiparasitic plant that grows in temperate and tropical regions worldwide, and it has been utilized as folk medicine or as a source of animal feed for many years (Vicas *et al.* 2011, Hossain *et al.* 2012). Mistletoe has anticancer, antimicrobial, and antioxidant effects besides beneficial effects on the immune system (Hossain *et al.* 2012, Yusuf *et al.* 2013). Mistletoe leaf meal has been reported to be a promising diet for birds, as it has no reported adverse effects (Egbewande *et al.* 2011). The anticancer and antioxidant activities of mistletoe have mainly been attributed to its main compounds, lectins and phenolic acid (Vicas *et al.* 2011, Hossain *et al.* 2012); although digallic and *o*-coumaric acid have also been reported to be associated with its antioxidant activity (Luczkiewicz *et al.* 2001). Most studies have mainly focused on the pharmaceutical effects or biochemical activities of European mistletoe (*Viscum album* L.) rather than on other types of mistletoe (e.g. American mistletoe [*Phoradendron serotinum*], Korean mistletoe [KM; *Viscum album* var. *coloratum*], and African mistletoe [*Tapinanthus bangwensis*]). However, there have been evaluations of the effects of these natural agents on broiler production and meat quality (Hossain *et al.* 2012, Ko *et al.* 2012).

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In order to evaluate the inclusion of *S. chinensis* and KM powders in poultry diets, we investigated the growth performance, carcass characteristics, serum cholesterol profiles, and meat quality of broilers fed *S. chinensis* or KM powder over a 35-day trial period.

MATERIALS AND METHODS

Birds, diets and experimental design: *S. chinensis* fruits, and KM leaves and stems were purchased from a herbal medicine market (Daegu, South Korea). They were then maintained in an oven at 50°C for 8 h before being ground to a fine texture. All procedures were performed in accordance with the guidelines of the animal policy established by the farm of Daegu University (Approval no: DUIACC-2016-015-1012-007). A total of 240 one-day-old male broiler chicks (Arbor Acres) were equally divided into 20 pens, and one of 5 treatments with 4 replicates were performed following a completely randomized design. Twelve birds per experimental treatment were placed in each pen. The treatments used in this experiment were: non-supplemented control diet (control), 0.5% (T1) and 1% (T2) *S. chinensis* powder supplemented, and 0.5% (T3) and 1% (T4) KM powder supplemented. The allowable levels of *S. chinensis* and KM powders in the diets are 0.5% and 1%, respectively, as described by Kim *et al.* (2013) and Kim and Choi (2014). In this study, standard industry broiler diets were formulated as described by Kim (2014), with a grower diet fed from day 1 to day 21 and a finisher diet fed from day 22 to day 35 (Table 1) (Seo 2019). New rice hulls with wood shavings (to a depth of approximately 6 cm) served as pen litter over a concrete floor. Each pen (1.1 m × 1.4 m) was equipped with a single tube feeder and an automatic bell drinker. All birds were kept under a fixed 14:10 h light:dark cycle and were provided with food and water *ad lib.* throughout the experiment. Temperature and ventilation were automatically controlled. For growth performance evaluation, body weights (BW) at chicken ages 1 day and 35 days were recorded in the pens. Feed intake (FI) was determined at each feed change interval. Feed conversion ratio (FCR) was calculated as the ratio of feed weight to weight gain. The mortality rate was calculated as follows: total dead birds throughout the experimental period divided by the total number of birds on day 1.

Collection of meat and blood samples: On day 35, 3 birds per pen were randomly selected for slaughter. After a 6 h fast, the birds were individually weighed and transported to the slaughterhouse where they were stunned electrically, slaughtered by slitting the neck, and exsanguinated. To obtain thigh muscles and evaluate carcass weight as a percentage of live weight (i.e. carcass ratio), the birds were then dissected. Before evaluation for different quality parameters, the skin (along with visible connective tissue and subcutaneous fat) was removed from the thigh muscles. Thigh muscle samples were stored in a sealable plastic bag and held at 4°C until analysis. Eviscerated carcass weight percentages were calculated by dividing eviscerated carcass weight by live weight after fasting and multiplying by 100.

Table 1. Ingredients and chemical composition of the basal diet fed to broilers (as-fed basis)

Ingredient	Starter (1 to 21 d; %)	Finisher (22 to 35 d; %)
Corn	54.624	62.612
Soybean meal CP46%	35.480	30.220
Fish meal	1.000	
Soya oil	4.960	3.620
Dicalcium phosphate	1.940	1.520
Limestone	1.190	1.270
Salt	0.300	0.300
DL-methionine	0.160	0.112
Choline 50%	0.080	0.080
Etoxyquine 30%	0.066	0.066
Vitamin premix ¹	0.100	0.100
Mineral premix ²	0.100	0.100
Total	100.000	100.000
Calculated analysis (%)		
ME (MJ/kg)	12.970	12.970
Crude protein (%)	21.500	19.000
Methionine (%)	0.500	0.380
Lysine (%)	1.100	1.000
Ca (%)	1.000	0.900
Available P (%)	0.450	0.350

¹Vitamin premix provides the following (per kg of diet): Vitamin A, 5,500 IU; Vitamin D₃, 1,100 IU; vitamin E, 10 IU; riboflavin, 4.4 mg; vitamin B₁₂, 12 mg; nicotinic acid, 44 mg; menadione, 1.1 mg; biotin, 0.11 mg; thiamine, 2.2 mg; ethoxyquin, 125 mg. ²Mineral premix provides the following (per kilogram of diet): Mn, 120 mg; Zn, 100 mg; Fe, 60 mg; Cu, 10 mg; Se, 0.17 mg; I, 0.46 mg; Ca, min: 150 mg, max: 180 mg.

Moreover, liver, spleen, and abdominal fat weights were determined after removal from the eviscerated carcass weight.

Blood samples (5 mL) were obtained from 3 birds per pen by inserting a sterile needle into the wing vein at the end of the experiment. Blood was centrifuged at 4000 r/min for 5 min to separate the serum. The collected serum samples were then frozen at -20°C for subsequent serum cholesterol determination. The concentrations of total cholesterol (TC), high-density lipoprotein cholesterol (HDL-C), low-density lipoprotein cholesterol (LDL-C), and triglyceride (TG) levels were determined by enzymatic colorimetric methods incorporated into commercial kits (Boehringer Mannheim, Germany) and an automatic analyzer (Hitachi 747; Hitachi Co., Tokyo, Japan) (Kim 2014).

Analytical procedures: All measurements were performed in triplicate. Moisture, crude protein, crude fat, and crude ash contents of thigh-meat samples were measured according to the methods of AOAC (2005). Thigh meat (10 g) were homogenized with 90 mL of distilled water by using a blender, and the mixture's pH was measured using a digital pH meter (Metrohm, Swiss). Thiobarbituric acid reactive substances (TBARS) were measured using the procedure described by Witte *et al.* (1970), and the results were reported as milligrams of malonaldehyde (MA)

per kilogram of meat. In brief, 20 g of a meat sample was mixed with 50 mL of 20% trichloroacetic acid solution (in 2 M phosphate solution) and homogenized using a blender. The mixtures were blended with 50 mL of distilled water for 2 min and then filtered. Subsequently, 5 mL of the filtered solution was mixed with 5 mL of thiobarbituric acid solution (0.005 M in water) in a test tube and allowed to stand at room temperature for 15 h. The absorbance value of the solution was measured at 532 nm by using a UV-Vis spectrophotometer (Shimadzu, Japan). The surface color values of the thigh-meat samples were obtained by using a Minolta colorimeter, which uses the CIE L* a* b* color system. Calibration was performed with a standard white plate with lightness (L*) = 96.16, redness (a*) = 0.10; and yellowness (b*) = 1.90 values. Thigh muscle surface color was measured three times for each sample.

Statistical analysis: All data were subjected to analysis of variance by using the general linear model procedure of SAS (SAS Institute Inc., 2002) at a 0.05 level of significance. Treatment pen mean values served as experimental units in the statistical analysis. Comparisons among mean values were performed by using Duncan's multiple range test (Duncan 1955).

RESULTS AND DISCUSSION

Growth performance, carcass characteristics, and serum cholesterol levels: The effects of *S. chinensis* and KM treatments on growth performance, carcass characteristics, and serum cholesterol levels are summarized in Table 2. There was no significant difference in growth performance

among the groups ($P > 0.05$). These materials were involved in lipid metabolism without physiological negative effects. This result is consistent with the results reported by Hossain *et al.* (2012) and Ko *et al.* (2012), who detected no significant differences in body weight gain and feed intake for broilers fed with KM (0.5%) or *S. chinensis* (0.2%). Similarly, Kim *et al.* (2013) and Kim & Choi (2014) reported that neither the use of *S. chinensis* (0.5%, 1%, and 2%) nor KM (0.5%, 1%, and 2%) had beneficial effects on growth performance. However, Ma *et al.* (2005) and Kim *et al.* (2007) found that supplementation with 1% *S. chinensis* or 0.5% KM increased the laying performance and the feed conversion ratio of broilers. In addition, Kang *et al.* (2012) and Yeh *et al.* (2011) reported that feed intake of herbal medicines to pigs was high in feed intake and growth rate but not significant.

Natural and herbal plant extracts have digestion-stimulating properties related to their extensive abundance of different molecules with intrinsic bioactivities that can affect animal nutrition and metabolism (Hernández *et al.* 2004). Moreover, an improvement in animal production could be associated with improved intestinal lumen health (Yeggani and Korver 2008). One of the most important observations that influenced broiler performance in this study was the low mortality found in the T4 group (1% KM). On that basis, KM rather than *S. chinensis* could improve the efficacy of immune responses in broilers, possibly because of the presence of active components in KM. Regardless, little has been reported about the benefits of adding *S. chinensis* or KM to the diets of poultry.

Table 2. Effect of feeding *Schisandra chinensis* and Korean mistletoe powders on growth performance, serum cholesterol, and carcass characteristics in broilers

Item	Treatments ¹				
	Control	T1	T2	T3	T4
<i>Growth performance</i>					
Initial body weight (g)	41.25±0.53	41.04±0.21	41.16±0.70	40.94±0.30	41.26±0.14
Final body weight (g)	1868.78±21.38	1866.51±6.63	1867.31±71.31	1863.26±18.52	1863.85±4.37
Weight gain (g)	1827.53±21.91	1825.47±9.84	1826.15±22.10	1822.33±18.22	1822.59±4.23
Feed intake (g)	3138.30±28.55	3137.97±8.52	3134.58±53.36	3143.94±37.27	3139.08±29.75
Feed conversion	1.72±0.04	1.72±0.02	1.72±0.04	1.73±0.04	1.72±0.02
Mortality (%)	3.48±0.08 ^a	2.91±0.06 ^b	1.73±0.09 ^c	1.73±0.07 ^c	1.63±0.04 ^c
<i>Carcass characteristics</i>					
Carcass weight (g)	1343.86±9.26	1349.62±7.37	1342.58±7.49	1347.95±8.20	1343.58±5.14
Carcass ratio (%)	71.50±0.76	71.43±0.09	71.61±0.13	71.65±0.34	71.74±0.06
Liver weight (g)	51.68±2.63 ^a	49.90±0.70 ^a	49.95±0.49 ^a	46.69±1.52 ^b	45.40±1.36 ^b
Spleen weight (g)	2.51±0.07 ^a	2.12±0.14 ^{ab}	2.30±0.08 ^{ab}	2.12±0.37 ^{ab}	1.90±0.39 ^b
Abdominal fat (g)	34.20±1.53 ^a	32.01±1.15 ^{ab}	30.84±1.26 ^b	29.82±2.05 ^{bc}	28.08±1.51 ^c
<i>Serum cholesterol (mL/dL)</i>					
Total cholesterol	169.30±5.65 ^a	162.58±1.00 ^{ab}	154.36±2.43 ^b	154.75±2.58 ^b	147.75±0.94 ^c
HDL-cholesterol	111.64±3.48 ^b	110.31±2.63 ^b	126.42±2.22 ^a	127.01±1.88 ^a	129.29±1.45 ^a
LDL-cholesterol	38.79±3.99 ^a	34.08±2.58 ^b	31.91±1.94 ^b	31.73±0.74 ^b	29.69±0.51 ^b
Triglyceride	120.10±5.16 ^a	118.35±1.31 ^{ab}	112.99±6.42 ^b	114.18±8.23 ^b	111.18±4.61 ^c

Values are expressed as means±SEM. ^{a-c}Mean values within rows with different letters are significantly different ($P < 0.05$). ¹Control, basal diet; T1, 0.5% *S. chinensis* powder; T2, 1% *S. chinensis* powder; T3, 0.5% Korean mistletoe powder; T4, 1% Korean mistletoe powder.

No significant differences in carcass weights or carcass ratios were observed among the groups ($P > 0.05$). A similar pattern was reported for a dietary herbal essential oil mixture used in broiler production, which was shown to have no effect on carcass weights at the 42-day stage (Cabuk *et al.* 2006). The 1% KM-treated (T4) group had the lowest liver, spleen, and abdominal fat weights ($P < 0.05$), and the 0.5% and 1.0% *S. chinensis* groups (T1 and T2, respectively) rather the 0.5% KM group (T3) had higher liver, spleen, and abdominal fat weights. These results are inconsistent with the results of previous studies, in which plant extracts and *S. chinensis* supplementation did not significantly affect the relative weight of digestive organs (Hernández *et al.* 2004, Ko *et al.* 2012). However, Kim *et al.* (2013) obtained results similar to those in the present study by supplementing poultry diets with *S. chinensis* (0.5%, 1%, and 2%). That study reported a decrease in liver and abdominal fat weights. The mechanism underlying the reduction in the relative weights of organs after *S. chinensis* or KM supplementation is undescribed.

Compared to the control group, higher levels of *S. chinensis* and KM supplements significantly decreased TC, LDL-C, and TG levels and increased HDL-C levels at day 35 ($P < 0.05$). These effects were mainly attributed to the presence of lignans, lectins, and phenolic acid, major components in diets supplemented with *S. chinensis* and KM (Luczkiewicz *et al.* 2001, Ko *et al.* 2012); their presence leads to increased antioxidant activities, which inhibits cholesterol absorption, or to decreased synthetic enzyme activity in the intestine. The highest serum TC level was observed in the control group, whereas the lowest serum TC level was measured in the 1% KM supplemented (T4) group. These results suggest that compared to *S. chinensis*, KM has a greater ability to regulate the intestinal environment and, in turn, induce an improvement in antioxidant activity. Our results are consistent with those of Kim *et al.* (2007), Kim *et al.* (2013), and Kim and Choi

(2014), who observed that *S. chinensis* and KM caused a reduction in serum cholesterol levels. Also, Kim (2014) reported that total cholesterol and LDL-cholesterol decreased while HDL-cholesterol tended to increase in the blood composition of broiler chickens fed red ginseng mare and mistletoe powder. In contrast, Hossain *et al.* (2012) observed that a 0.5% water plantain and mistletoe powder supplement did not affect cholesterol levels.

Proximate composition and meat quality: Table 3 summarizes the effects of dietary *S. chinensis* and KM powder supplements on the proximate composition and meat quality of broilers. The Crude protein values of on the proximate composition did not differ among the groups ($P < 0.05$). Crude ash values were lower in the birds that received the 1% KM powder (T4) than in those in the control group and the other three treatment group (T1–T3). For Crude fat values, 0.5% S powder (T1) was higher than the control group and the other three treatment group (T2–T4). For crude ash values, 0.5% KM powder (T3) is approximately equal to the control group and the remaining group is slightly lower than the control group. Overall, the addition of *S. chinensis* or KM to the diet did not have a significant effect on the proximate composition in any of the treatment groups ($P < 0.05$), suggesting that proximate composition after dietary supplementation with *S. chinensis* and KM is not the main factor that affects broiler meat quality. According to Kim *et al.* (2013), increasing the levels of *S. chinensis* (0.5%, 1%, and 2%) had a positive effect on the proximate composition. Consistent with our results, Kim and Choi (2014) reported that KM (but not crude ash) had no beneficial effect on proximate composition. In addition, Kim (2014) reported that Korean mistletoe powder fed broiler chickens tended to be somewhat lower than other treatments in 1% of mistletoe powder ($P < 0.05$).

The pH values of the thigh muscle samples did not differ among the groups ($P < 0.05$), but there were slight differences in TBARS values among the groups. TBARS values tended

Table 3. Effect of feeding *Schisandra chinensis* and Korean mistletoe powders on the proximate composition and meat quality of broiler thigh muscle

Item	Treatments ¹				
	Control	T1	T2	T3	T4
<i>Proximate composition (%)</i>					
Moisture	73.44±0.29	73.54±0.09	73.72±0.11	73.84±0.04	73.67±0.16
Crude protein	22.59±0.12	22.55±0.15	22.54±0.08	22.61±0.19	23.55±0.09
Crude fat	2.67±0.21	2.82±0.22	2.66±0.16	2.46±0.25	2.69±0.05
Crude ash	1.10±0.04	1.09±0.02	1.09±0.02	1.10±0.02	1.09±0.02
<i>Meat quality</i>					
pH	6.20±0.08	6.16±0.08	6.10±0.03	6.10±0.07	6.09±0.02
TBARS (mg MA/kg)	0.47±0.03 ^a	0.45±0.01 ^{ab}	0.42±0.03 ^{ab}	0.41±0.02 ^{ab}	0.39±0.03 ^b
CIE L* (lightness)	57.73±0.88 ^a	57.47±0.91 ^a	55.80±0.16 ^b	55.28±1.03 ^b	54.91±0.85 ^b
CIE a* (redness)	11.09±0.73 ^b	11.18±0.26 ^b	13.00±0.18 ^a	13.13±0.06 ^a	13.40±0.21 ^a
CIE b* (yellowness)	8.29±0.36	8.65±0.27	8.60±0.17	8.57±0.24	8.54±0.20

Values are expressed as means±SEM. ^{a-b}Mean values within rows with different letters are significantly different ($P < 0.05$). ¹Control, basal diet; T1, 0.5% *S. chinensis* powder; T2, 1% *S. chinensis* powder; T3, 0.5% Korean mistletoe powder; T4, 1% Korean mistletoe powder.

to decrease with the addition of *S. chinensis* and KM. In addition, TBARS values were lower in the birds that received the 1% KM powder (T4) than in those in the control group and the other three treatment group (T1–T3). Both *S. chinensis* and KM, rich in lignans, lectins, and phenolic acid, are known for their antioxidant effects. Therefore, an antioxidant effect may be responsible for the decrease in lipid oxidation. Our results are consistent with those reported by Ma *et al.* (2007), who observed that diets with 1% *S. chinensis* could reduce MDA levels in the tissues of chicks. However, Hossain *et al.* (2012) observed no changes in the TBARS levels of breast and thigh meat when broilers were fed 0.5% mistletoe. Overall, our results indicate that KM rather than *S. chinensis* may be more effective as an antioxidant agent. Regardless, the antioxidant effects of *S. chinensis* and KM were not reflected in the pH value of the meat in our study. However, some studies have supported the hypothesis that it is possible to reduce meat pH with antioxidant effectiveness by using plant and herbal agents (Kim *et al.* 2013, Kim and Choi 2014). In addition, the effect of antioxidants on pH has been well described (Aksu 2007).

Meat color, as an indication of meat quality, differed ($P < 0.05$) among the groups that received *S. chinensis* and KM supplements and the control diet. The L^* (lightness) values were decreased and the a^* (redness) values were increased, but b^* (yellowness) did not change with the addition of *S. chinensis* and KM. The 1% KM (T4) group had lower L^* (lightness) values and higher a^* (redness) values than those in the 0.5% and 1% *S. chinensis* (T1 and T2) and 1% KM (T3) groups. It is important to note that KM, which showed a greater antioxidant effect, appeared to be more effective than *S. chinensis*. This is likely to be attributable to differences in the presence of antioxidant compounds in *S. chinensis* and KM. Results similar to those for KM were reported by Fernandez-Lopez *et al.* (2005) who showed a decrease in L^* values and an increase in a^* values when natural extracts were applied to beef meatballs. Moreover, the results are consistent with the findings of Kim *et al.* (2013) who reported that the addition of 0.5%, 1%, and 2% *S. chinensis* to broiler diets improved meat color stability. In contrast to our results, Kim and Choi (2014) reported that broilers in groups fed diets containing KM (0.5%, 1% or 2%) did not exhibit significant improvement in thigh muscle color.

In general, plant extracts have been proven to have antioxidant affects against lipid oxidation, and such effects are responsible for a reduction in the color stability of meat and meat products; however, the antioxidant effects of extracts on meat quality have not been fully described.

On the basis of our results, we concluded that higher levels of KM supplementation reduced mortality, serum cholesterol levels, and the relative weight of organs, as well, it improved meat quality (lipid oxidation and color stability). However, no positive effects on growth performance, carcass characteristics, and proximate composition were observed in the *S. chinensis* and KM treatment groups. In

particular, the results suggest that 0.5% to 1% KM powder could be used as an appropriate dietary supplement for poultry as KM has a potent antioxidant effect, which may help in improving the immune system, providing lipid stability, and reducing serum cholesterol levels. Furthermore, the mechanism underlying the promotion of antioxidant performance by *S. chinensis* and KM should be studied in more detail.

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