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# Meat quality and color of breast during storage in ducks fed diet supplemented with different forms of *Houttuynia cordata* with fermented red koji

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**ABSTRACT.** We evaluated meat quality and color of breast during storage in ducks fed diet supplemented with different forms of *Houtuynia cordata* (HC) with fermented red koji (FRK). In total, 240 one-day-old Pekin ducks (160 males and 80 females) were allocated to four diet treatments (control, 1% HC powder mixed with FRK, pelleted 1% HC with FRK, and coated pellets of 1% HC with FRK). At days 3 and 7 of storage, pH values were slightly influenced (p < 0.05) by different forms of HC with FRK, but was no influence at day 0 of storage day. Diet treatments with different forms of HC and FRK had an effect (p < 0.05) on TBARS values at 0, 3, and 7 days of storage and DPPH radical-scavenging activity at 0 days of storage, except for cooking loss. Overall, in breast meat of ducks, L\* values at day 3 of storage and a\* values at day 0 of storage were significantly influenced (p < 0.05) by treatments with different forms of HC and FRK. In conclusion, using either 1% HC and FRK pellets or pellets coated with HC and FRK at 1% resulted in a decrease in TBARS values and an increase in DPPH radical-scavenging activity during storage.

Keywords: duck breast meat; fermented red koji; Houttuynia cordata; meat color; meat quality.

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## Introduction

Several alternative feed additives have been recently developed for poultry diets and are being used because of antibiotic resistance, which can limit available therapeutic options or potentially leave residues in meat and eggs (Kim, Yi, Cho, & Ku, 2013a). Feeds without antibiotics are increasingly used in poultry nutrition to improve growth performance and meat quality. Consequently, this has led to adaptation in poultry, as new forms of feed additives are the products of modern science (Demir, Sarica, Ozcan, & Suicmez, 2005). The new generation of meat quality enhancers includes botanical additives in feeds, such as appropriate blends of herbs (*Houttuynia cordata*) and fungal species (fermented red koji).

Houttuynia. cordata (HC) is a perennial herb distributed in Korea, China, Japan, and southeast Asia. Its traditional use is medicinal and contains volatile oils, fatty acids, decanoyl acetaldehyde, sterols, flavonoid and alkaloids, which are believed to have a wide range of pharmacological functions, such as antibacterial, antimicrobial, and antioxidant effects (Meng et al., 2009). In a previous study in which HC was used an animal feed, Yan, Meng, and Kim (2011) demonstrated that the inclusion of HC extract powder (1%) in finishing pig diets increased nutrient digestibility and improved growth performance.

Red koji (RK), a product obtained from fermentation of the fungal genus *Monascus*, is the most important agent, which has been used for centuries as a medicinal food or herb in Eastern countries (Kim et al., 2013b). It was gradually considered to be a functional food because of its health-promoting effects (e.g., as an antihypercholesterolemic agent, antioxidant, and hypotensive agent) related to monacolin K, dimerumic acid, and  $\gamma$ -aminobutyric acid (Aniya et al., 2000; Arunachalam & Narmadhapriya, 2011; Su, Wang, Lin, & Pan, 2003). Furthermore, the effects of red koji on animal diets were first reported by Chung and Choi (2016) and Kim, Lee, and Choi (2016). They suggested that using 1% fermented red ginseng marc (FRGM) powder combined with RK in broilers improved growth performance and DPPH radical-scavenging activity (antioxidant effect) in broiler breast meat.

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At present, pelleting of feed is the most common processing method employed by feed manufacturers to improve animal performance and meat quality (Lv et al., 2015). Also, according to several reports, pelleted diets in broiler increased body weight and improved feed conversion compared to mash feeds (Amerah, Ravindran, Lentle, & Thomas, 2008; Chewning, Stark, & Brake, 2012). Apart from the benefits associated with nutritional quality, fermentation is a useful tool for improving overall meat quality and increasing digestibility of the feeds (Cao, Zhang, Yu, Zhao, & Wang, 2012). However, there is limited information on the use of different forms of HC with fermented red koji (FRK) on the quality of duck meat. In this regard, using HC with FRK in pelleting and coating of duck feeds may provide a novel way to improve the quality of duck meat. Therefore, we conducted the present study to evaluate meat quality and color of breast during storage in ducks feed diet supplemented with different forms of HC with FRK.

## Material and methods

The experimental procedures were approved by the Gilhong farm committee (Geochang, South Korea) following animal care guidelines and policies.

### Preparation of feed additives

HC leaves and FRK were purchased from Yusim Co. (Yeongju, Korea). The HC leaves were air-dried (for 12 h) at room temperature, hot-air-dried (50-60°C, for two consecutive days), and ground into a fine powder. The HC powder was thoroughly blended with FRK using a 9:1 ratio of HC:FRK. Pellets of HC mixed with FRK were obtained using a pellet machine (Kum Kang Eng., Daegu, South Korea). Some pellets were coated with canola oil using a small spray pump and dried for 7 d. These various forms of feed additives were placed in a sealed plastic bag and refrigerated until the experiment commenced.

### Animals and diets

After a 14-day brooding period, a total number of 240 one-day-old Pekin ducks (160 males and 80 females) were allocated to four diet treatments based on a completely randomized design. Each of the treatments had four replicates of 15 birds (10 males and 5 females) and lasted 38 d. The control group received the basal diet with no added supplements; Treatment 1, 2, and 3 included 1% HC powder mixed with FRK, pelleted 1% HC with FRK and coated pellets of 1% HC with FRK, respectively. Starter (21% crude protein, 2.5% crude fat, 8% crude fiber, 9% crude ash, 0.40% Ca, and 1.50% P) and finisher diets (17% crude protein, 2.5% crude fat, 8% crude fiber, 9% crude ash, 0.40% Ca, and 1.0% P) were fed to the birds up to 14–21 days and 22–38 days, respectively. During the experimental period, a feeder and a drinker for *ad libitum* access to feed and water in each pen (2.5  $\times$  2 m) was set in a place accessible to the birds. The duck facility was controlled by an automatic ventilation and temperature system to create a favorable environment. The room temperature was 33°C for 7 days and gradually was decreased to 23°C when the ducks were 21 days old, and this lower temperature was maintained until the end of the experimental period.

### Slaughter procedure

On day 38 of the feeding trial, five ducks selected randomly from each pen, were fasted for 12 h and moved to the slaughterhouse. Conventional slaughter house procedures were used to slaughter the ducks (i.e., stunned electrically, slaughtered by cutting the neck, and exsanguinated). Breasts were dissected and separated from each carcass. The skin (subcutaneous fat and visible connective tissues) were removed from the breast muscles before determining meat quality parameters (Kim, Jin, & Yang, 2009). Samples were placed in sealable polyethylene bags, labeled and stored in a refrigerator at 4°C for 0, 3, and 7 d.

### **Analytical procedures**

To determine meat pH, approximately 10 g of minced breast meat was homogenized in 90 mL of distilled water using a blender (HM-3000, Hyundai Electronic Industry Co., Incheon, South Korea). After homogenization, pH values were measured using a digital pH electrode (model 520A, Orion, Beverly, MA) attached to the homogenized solution. Thiobarbituric acid reactive substances (TBARS) values were measured according to the method described by Witte, Krause, and Bailey (1970) and were reported as mg of malondialdehyde (MA) per kg of breast meat. A 0.5 g sample was placed in the test tube and mixed with a combination of 3 mL of 1% trichloroacetic acid solution, 0.3% NaOH solution, 17 mL of 0.25%

trichloroacetic acid solution, and 3.6 mM of HCl solution. These mixtures were heated in a water bath at 98°C for 30 min and cooled for 15 min. After adding 3 mL chloroform, this mixture was centrifuged at 3500 × g for 30 min to separate the layers. Absorbance of the supernatant was measured at a wavelength of 532 nm with an UV-Visible spectrophotometer (UV-24D, Shimadzu, Tokyo, Japan). Free radical-scavenging activity was determined using the 1,1-Diphenyl-2-picrylhydrazyl (DPPH) radical scavenging assay according to the method of Blois (1958) with some modifications. We added 1 mL of extracts to 4 mL of ethanolic DPPH solution (100 μM). The mixture was then vortexed and allowed to stand at room temperature. After 30 min, absorbance was recorded at 517 nm using a UV-Vis spectrophotometer. For calculating losses due to cooking, we subtracted the sample weight after cooking from the sample weight before cooking and multiplied by 100. We then divided this value by the sample weight before cooking. The color of breast meat samples was measured using a colorimeter (Minolta Co. CR 301, Japan) that was calibrated by using a standard white calibration plate (reference number 12633117, Y = 93.5, x = 0.3132 and y = 0.3198). The color was recorded through a Minolta colorimeter, measuring colour coordinates: lightness (L\*), redness (a\*) and yellowness (b\*). All samples were measured in triplicate.

### **Statistical Analysis**

All statistical analyses were carried out using the General Linear Models procedure of the Statistical Analysis System software (SAS, 2004). When differences among treatments were significant, means were compared by Tukey's test. Significance was declared at p < 0.05.

# **Results and discussion**

Table 1 shows the effects of different forms of HC with FRK on meat quality in duck breast meat. First, pH values at 3 and 7 days of storage were slightly influenced (p < 0.05) by different forms of HC with FRK. However, at 0 days of storage, treatments with different forms of HC with FRK showed statistically similar values to those of the control groups (p > 0.05). Compared to the values observed in controls, the addition of different forms of HC with FRK increased or decreased pH values at 3 days of storage and then reduced the pH values at 7 days, except for T1 (1% HC powder with FRK). Possibly, a small reduction in the pH values of HC and FRK (T2 and T3) at 7 days might be explained by the fact that the ability of the antioxidant is dependent on pH values (Xiong, Decker, Robe, & Moody, 1993). Similarly, Kim et al. (2016) showed a minor significant (p < 0.05) effect on pH in broiler breast meats with FRGM with RK as the number of storage days increased.

Dietary treatments with different forms of HC and FRK had an effect (P < 0.05) on TBARS values as the number of storage days increased. At 0 and 3 days of storage, the TBARS values of duck breast meat in the T2 and T3 groups was lower than that of duck breast meat in other groups. This result conforms with the findings of Sarker and Yang (2011), who showed a reduction in TBARS values of broiler meat (breast and thigh) treated with antibiotics and HC with probiotics (HCP, 0.5% and 1%) groups compared with controls. Yan et al. (2011) confirmed that the inclusion of HC extract powder (1%) in finishing pigs increased TBARS values.

Item	day —	$Treatment^1$				- SEM <sup>2</sup>	P - Value
		Con	T1	T2	Т3	SEIVI-	r - value
рН	0	5.90	5.92	5.94	5.92	0.027	0.523
	3	5.91 <sup>b</sup>	5.87 <sup>b</sup>	5.90 <sup>b</sup>	6.07 <sup>a</sup>	0.019	0.002
	7	$6.07^{ab}$	6.10 <sup>a</sup>	$5.94^{\rm b}$	5.99 <sup>ab</sup>	0.048	0.038
TBARS (mg MA kg <sup>-1</sup> )	0	0.76ª	0.72 <sup>b</sup>	0.55 <sup>b</sup>	0.44 <sup>c</sup>	0.032	P < 0.000
	3	$0.89^{b}$	$0.73^{ab}$	$0.47^{a}$	$0.55^{ab}$	0.101	0.023
	7	$0.37^{\rm b}$	$0.47^{a}$	$0.46^{a}$	$0.40^{b}$	0.016	0.001
OPPH radical scavenging (%)	0	72.00 <sup>a</sup>	71.60 <sup>a</sup>	60.60 <sup>b</sup>	57.80 <sup>b</sup>	1.180	P < 0.000
	3	61.50	62.70	63.90	64.50	5.761	0.919
	7	83.10	83.50	85.50	85.60	1.328	0.131
Cooking loss (%)	0	32.00	31.00	30.80	31.20	1.410	0.833
	3	39.50	38.80	40.80	36.30	2.167	0.227

40.50

39.00

0.567

41.00

Sheans in the same rows with no common superscript are significantly different (p < 0.05). Con: basal diet; T1: basal diet + 1% HC powder mixed with FRK; T2: basal diet + pelleted 1% HC with FRK; T3: basal diet + coated pellets of 1% HC with FRK. <sup>2</sup>Values are expressed as standard error means.

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In general, TBARS test is the most commonly used oxidation-marker test for meat and meat products. The presence of oxidized lipids in animal diets and meats resulted in a loss of nutritional and sensory values, as well as the formation of potentially toxic compounds (Sarker & Yang, 2011). Thus, it is reasonable to assume that using HC and FRK reduces TBARS values via antioxidant activity. Additionally, the mechanism of action of this antioxidant effect may be due to one of the following: 1) the presence of bioactive material from a combination of HC (decanoyl acetaldehyde, flavonoid and alkaloids) and FRK (monacolin K, a potent inhibitor of HMG-CoA reductase inhibitor), 2) pelleting and coating feeds improve animal performance and meat quality, and 3) fermentation processes using RK improve the nutritive value of feeds. On the other hand, the TBARS values of duck breast meats in the control samples, in this study, after 7 days of storage were lower compared to those of the groups treated with different forms of HC and FRK; however, the reason for this decrease in controls is far from clear.

Based on the results obtained from duck breast meat, dietary treatments with different forms of HC with FRK had an influence (p < 0.05) on DPPH radical-scavenging activity at 0 days of storage, which was lower than that of the control. Despite the lack of significant differences, DPPH radical-scavenging activity in T1, T2 and T3 at 3 and 7 days of storage, tended to decrease compared to that of the control. In addition, stronger DPPH radical-scavenging activity in duck breast meat was observed in T2 and T3. This phenomenon is similar to observations for red ginseng marc and  $\alpha$ -tocopherol or FRGM powder mixed with RK for broiler breast meat (Kim et al., 2016; Kim, Lee, & Choi, 2014). In fact, the DPPH scavenging ability (antioxidant effect) of HC and FRK supplemental feeds may be primarily the result of essential oils, alkaloids, and flavonoids, or dimerumic acid, which is one of the main components of HC and FRK (Arunachalam & Narmadhapriya, 2011; Fu, Dai, Lin, & Lu, 2013). DPPH scavenging ability and MDA content are indicators of antioxidant effects in meat. This suggests that the antioxidant effects, with respect to HC and FRK, can be expected from the 3 days of storage rather than at 7 days. Although antioxidant activity is dependent on meat pH, improvement in TBARS and DPPH scavenging ability was not associated with the pH of duck breast meat in this study.

During storage, no differences (p > 0.05) were found in cooking losses among all treatments. This result implies that different feed form with combinations of HC and FRK did not affect cooking loss during storage. In some report, cooking loss is well-known as a major factor that affects meat quality, owing to nutrients lost in the exudates through water loss (Park, Kang, Shin, & Shim, 2015). In our study, the detailed reasons for this decrease or increase in the cooking losses of breast meat of ducks fed HC and FRK were unknown.

The effects of different forms of HC with FRK on color of duck breast meat during storage are presented in Table 2. Overall, L\* at 3 days of storage and a\* values at 0 days of storage in breast meat of ducks were significantly influenced (p < 0.05) by treatments with different forms of HC and FRK. There were no differences (p > 0.05) in L\* at 0 and 7 days of storage, a\* at 3 and 7 days of storage, and b\* values at 0-7 days of storage in breast meat of ducks among treatments. In other words, feed form or a combination of HC and FRK did not significantly affect meat color. Similar results and conclusions were documented by Kim et al. (2016), who reported that even though meat color was significantly different among groups fed different forms of red ginseng and red koji, these additives did not have an antioxidant effect on meat color. Also, it has been well documented that the presence of antioxidant compounds in natural products may retard metmyoglobin formation and decrease L\* values or a\* values (Fernandez-Lopez, Zhi, Aleson-Carbonell, Perez-Alvarez, & Kuri, 2005).

Item	day _	Treatment <sup>1</sup>				SEM2	P - Value
		Con	T1	T2	Т3	=	
L (lightness)	0	48.90	50.20	50.30	48.80	0.723	0.340
	3	$49.90^{b}$	$50.40^{\rm b}$	52.50 <sup>a</sup>	$52.50^{a}$	0.529	0.007
	7	53.00	51.10	52.80	48.20	1.198	0.067
a (redness)	0	11.60 <sup>b</sup>	11.40 <sup>b</sup>	11.70 <sup>b</sup>	13.30 <sup>a</sup>	0.568	0.003
	3	15.20	15.90	15.30	15.20	0.307	0.171
	7	14.40	14.10	14.00	15.40	0.564	0.082
b (yellowness)	0	3.45	3.54	3.45	3.84	0.301	0.353
	3	4.68	3.57	3.63	4.21	1.475	0.770
	7	5.47	4.72	4.19	3.76	1.322	0.549

a-bMeans in the same rows with no common superscript are significantly different (p < 0.05). ¹Con: basal diet; T1: basal diet + 1% HC powder mixed with FRK; T2: basal diet + pelleted 1% HC with FRK; T3: basal diet + coated pellets of 1% HC with FRK. ²Values are expressed as standard error means.

However, our observations do not support the findings of Fernandez-Lopez et al. (2005). At present, the exact mechanism causing variation in meat color is not well understood, due to a lack of data in literature on the effects of HC with FRK on meat color and quality.

### Conclusion

Addition of pellets and coated pellets of HC and FRK at a level of 1% in duck diets improved antioxidant effects (TBARS values and DPPH radical-scavenging activity), which were not associated with variation in meat pH. However, this study failed to find an effect on cooking losses and meat color using different forms of HC with FRK.

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