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Physical and Chemical Characteristics of Meats of the Indonesian Indigenous Crossbred Chickens Fed Fermented Mixture of Papaya (*Carica papaya* L.) Leaf and Seed Meal

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ABSTRACT

The objective of this present study was to investigate the effect of feeding fermented mixture of papaya leaf and seed meal (FERM) on the physical and chemical characteristics of meats of the Indonesian indigenous crossbred chicken (IICC). The study was carried out with 300 day-old IICC. The chicks were randomly distributed to five treatment groups, i.e., CONT (control diet based on corn-soybean- diet), FERMA (diet containing 1% FERM), FERMB (2.5% FERM), FERMC (5% FERM) and FERMD (7.5% FERM). Each treatment group consisted of 6 replicates with 10 IICC in each. At week 8, one chick from each replicate was randomly taken and slaughtered. After being de-feathered and eviscerated, samples from breast and thigh meats were obtained. Results showed that the increased levels of FERM was followed by the increased (P<0.05) pH values, moisture and crude protein content of breast meats of the IICC. Dietary incorporation of FERM especially at the level of 2.5% increased (P<0.05) the content of fat in the breast meat of IICC, while further increased levels of FERM did not alter (P>0.05) the fat content of the IICC breast meat. Dietary treatment did not have any effect (P>0.05) on the lightness (L*) values of breast meat of the IICC. The redness (a*) values were higher (P<0.05) in FERMD breast meat than others. FERM diet resulted in lower (P<0.05) yellowness (b*) values in the IICC breast meat. The pH values and moisture content of thigh meat increased (P<0.05) with the increased level of FERM. The WHC decreased (P<0.05) with the elevated levels of FERM in the diets. There was an increase (P<0.05) in crude protein concentration in FERMA as compared to the other meats. Crude fat and ash concentrations in thigh meat were affected (P<0.05) by the treatments. Dietary treatments had no impact (P>0.05) on L* values of thigh meats. Feeding FERM at 7.5% from diets increased (P<0.05) and decreased (P<0.05) the redness and yellowness of meats. In conclusion, dietary inclusion of FERM especially at the level of 7.5% from diets improved the physical and chemical characteristics of the IICC meats.

Key words: Crossbred chicken; Fermented feed; Meat quality

INTRODUCTION

Recently, the Indonesian indigenous crossbred chicken (IICC) has gained more interest from the consumers. Different from modern broiler strains, the IICC produces meat with unique taste and texture. With the intensive rearing system, the crossbred chicken needs only 8 to 10 weeks to reach the commercial weight, which is much shorter compared to the indigenous local chicken in Indonesia (Pramono, 2006; Sugiharto *et al.*, 2018). Similar to modern broilers strains, in-feed antibiotics have commonly been applied in the IICC to maximize the growth and control the outbreak of infectious diseases. Yet, due to the microbial resistance and food safety reasons, therapeutic antibiotics have been withdrawn from diets of the IICC since January 2018.

In response to the antibiotic retraction, many farmers of the IICC are now trying to find the substitutes for in-feed antibiotics (Sugiharto *et al.*, 2018). Phytobiotic, which is plant-based active compound and one the potential antibiotic substitutes, has recently gained a wide popularity among the farmers. Of the phytobiotics, the leaf and seed of papaya (*Carica papaya* L.) have been exploited as phytogenic growth promoter and antibacterial agents in poultry production (Bolu *et al.*, 2009; Onyimonyi and Ernest, 2009; Nideou *et al.*, 2017). Due to their high protein content, papaya leaf and seed have also been employed as dietary protein source for chicken rations (Onyimonyi and Ernest, 2009). Yet, study on the effect of papaya leaf and seed on chicken meat characteristics has never been elucidated

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so far. Note that the chicken meat traits are greatly affected by dietary treatments (Sugiharto *et al*., 2017).

In the present study, papaya leaf and seed meal were firstly mixed and then fermented using the fibrolytic fungus *Chrysonilia crassa* before being incorporated into the chicken rations. Fungal fermentation was actually subjected to decrease the fibre content, while the mixing of papaya leaf and seed was to exert the synergistic effect of both phytogenic materials. The objective of this present study was to investigate the effect of feeding fermented mixture of papaya leaf and seed meal on the physical and chemical characteristics of meats of the IICC.

MATERIALS AND METHODS

The study was initiated by preparing the fermentation starter of the fungus *C. crassa* according to Sugiharto *et al*. (2019). Following this stage, the production of the fermented mixture of papaya leaf and seed meal (FERM) was conducted. In brief, 50% of papaya leaf meal, 45% papaya seed meal and 5% fungal fermentation starter were blended thoroughly. The mixture was added with sterilized water (1:1) and then incubated aerobically at room temperature for 4 days. The FERM was eventually sun-dried before being incorporated into the chicken rations.

The in vivo study was carried out with 300 day-old of the IICC. The chicks were randomly distributed to five treatment groups, i.e., CONT (control diet based on corn-soybean- diet), FERMA (diet containing 1% FERM), FERMB (2.5% FERM), FERMC (5% FERM) and FERMD (7.5% FERM). Each treatment group consisted of 6 replicates with 10 IICC in each. The diets were prepared and formulated in mash form as starter (weeks 1-4) and finisher (weeks 5-8) feeds (Table 1). Throughout the study period, the feeds and water were offered *ad libitum* to all IICC. Vaccination was conducted at day 4 using the commercial Newcastle disease-infectious bursal disease (ND-IBD) vaccines (through eye drop), day 14 using the commercial IBD vaccine (drinking water) and day 22 using the commercial ND vaccine (drinking water). At week 8, one chick from each replicate was randomly taken and slaughtered. After being de-feathered and eviscerated, samples from breast and thigh meats were obtained and stored at freezer until analysis.

Before performing the analysis, the frozen meat was thawed for about 30 min at room temperature. To determine the pH value, 1 g of breast or thigh meats was homogenized in 9 Ml of distilled water, and pH of the resulting filtrate was then measured by means of the digital pH meter (Hanna Instruments, Woonsocket, Rhode Island). The press methods using filter paper (Pla and Apolinar, 2000) was applied to measure the water holding capacity (WHC) of breast and thigh meats, meanwhile the standard proximate analysis (AOAC, 2005) was implemented to determine the chemical composition of the IICC meats. The assessment of colour of the IICC meats was carried out based on Sugiharto *et al*. (2019). The assessment employed a digital colour meter in Mac OS X (set to CIE Lab), and reported as L* (lightness), a* (redness) and b* (yellowness) values.

ANOVA (SAS Inst. Inc., Cary, NC, USA) was employed to analyse the data collected from the present study. If the considerable ($P<0.05$) difference among the treatment groups was found, the data were then subjected to Duncan's multiple-range test.

RESULTS

Table 2 shows the physical and chemical characteristics of breast meat of the IICC. It was apparent that pH values increased ($P<0.05$) with the increased FERM in the IICC diets. Moisture content of breast meat was highest ($P<0.05$) in FERMD than in other breast meats. Crude protein was higher ($P<0.05$) in FERMD compared to that in CONT breast meat. Compared to other breast meat, FERMB had higher ($P<0.05$) fat content. CONT had higher ($P<0.05$) ash content when compared with FERMA and FERB. The a* values were higher ($P<0.05$) in FERMD than in other breast meat groups, while b* values were higher ($P<0.05$) in CONT compared to other breast meats.

Data on the physical and chemical traits of thigh meat of the IICC are presented in Table 3. pH values of thigh meats increased ($P<0.05$) with the increasing levels of FERM in the IICC diets. The WHC was lowest ($P<0.05$) in FERMD compared to other treatment groups. Moisture content was higher ($P<0.05$) in FERMA, FERMB and FERMD compared to that in CONT and FERMC meats. Crude protein was higher ($P<0.05$) in FERMA compared to other meats. Crude fat was lower ($P<0.05$) in FERMA and FERMD compared to other thigh meat groups. Ash content was lower ($P<0.05$) in FERMB than that in other thigh meats. FERMD had the highest ($P<0.05$) a* values and lowest ($P<0.05$) b* values.

DISCUSSION

It was shown in the current work that the increased levels of FERM was followed by the increased values of pH of breast meats of the IICC. In general, the increased pH values were associated with the elevated WHC and moisture content of chicken meat (Kralik *et al*., 2017). In accordance, our current findings documented that the increased pH values were associated with the increased moisture content of the IICC meat. It seemed that the increased pH may avoid the protein denaturation in IICC meat (Kralik *et al*., 2017). The latter condition actually occurred in the breast meat of the IICC as the increased pH values was in line with the higher crude protein content in breast meat of the IICC. In respect particularly to crude protein, feeding FERM was most likely to increase the levels of protein in the breast meat of the IICC. This was supported by Sugiharto *et al*. (2017) showing an increased crude protein content in broiler breast meat with feeding fermented cassava pulp. Also, Marcinčák *et al*. (2018) revealed that feeding fermented bio-product resulted in the increased protein content in the breast meat of broiler chickens. The definite mechanism by which FERM increased the crude protein content of the IICC breast meat is largely unknown, but Sugiharto *et al*. (2017) formerly suggested that the enhancing effect of the probiotic fungus (used as the fermentation starter) on the protein efficiency ratio may be one of the mechanisms. The improved protein efficiency ratio may consequently increase protein production in the muscularity of the IICC. It was shown in this study that dietary incorporation of FERM especially at the level of 2.5% increased the content of fat in the breast meat of IICC, while further increased levels of FERM in the diets did not alter the fat content of the IICC breast meat. In line with our finding, previous study by

Table 1: Ingredients and chemical compositions of experimental diets.

Items (%, unless otherwise noted)	Starter					Finisher				
	CONT	FERMA	FERMB	FERMC	FERMD	CONT	FERMA	FERMB	FERMC	FERMD
Yellow maize	55.0	54.7	54.2	53.5	53.0	60.0	59.8	59.3	58.6	58.0
SBM	35.8	35.2	34.5	33.5	33.0	32.0	31.5	30.7	29.7	28.3
MBM	4.50	4.50	4.15	3.55	2.25	2.65	2.50	2.35	1.65	1.40
Soybean oil	1.50	1.45	1.40	1.25	1.00	2.10	2.00	1.95	1.80	1.57
FERM	-	1.00	2.50	5.00	7.50	-	1.00	2.50	5.00	7.50
DL-methionine	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30
L-Lysine	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Limestone	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50
DCP	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50	1.50
Premix[1]	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50
Salt	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Compositions:										
ME (kcal/kg)	2,900	2,900	2,903	2,904	2,902	3,001	2,999	3,001	3,003	2,999
Crude protein	22.0	22.0	22.0	22.0	22.0	20.0	20.0	20.0	20.0	19.9
Crude fibre	5.60	5.70	5.80	6.10	6.40	5.60	5.70	5.90	6.10	6.40
Ca	1.20	1.20	1.20	1.20	1.20	1.20	1.20	1.20	1.20	1.20
P (available)	0.70	0.70	0.70	0.70	0.70	0.70	0.70	0.70	0.70	0.70
Lysine	1.20	1.20	1.20	1.20	1.20	1.20	1.20	1.20	1.20	1.20
Methionine	0.60	0.60	0.60	0.60	0.60	0.60	0.60	0.60	0.60	0.60

[1]Premix contained (per kg of diet) of vitamin A 7,750 IU, vitamin D3 1,550 IU, vitamin E 1.88 mg, vitamin B1 1.25 mg, vitamin B2 3.13 mg, vitamin B6 1.88 mg, vitamin B12 0.01 mg, vitamin C 25 mg, folic acid 1.50 mg, Ca-d-pantothenate 7.5 mg, niacin 1.88 mg, biotin 0.13 mg, BHT 25 mg, Co 0.20 mg, Cu 4.35 mg, Fe 54 mg, I 0.45 mg, Mn 130 mg, Zn 86.5 mg, Se 0.25 mg, L-lysine 80 mg, Choline chloride 500 mg, DL-methionine 900 mg, $CaCO_3$ 641.5 mg, DCP 1500 mg: CONT: control diet based on corn-soybean- diet, FERMA: diet containing 1% FERM, FERMB: 2.5% FERM, FERMC: 5% FERM, FERMD: 7.5% FERM, SBM: soybean meal, MBM: meat bone meal, FERM: fermented mixture of papaya leaf and seed meal, DCP: dicalcium phosphate, ME: metabolizable energy.

Table 2: Physical and chemical characteristics of breast meat of the IICC fed treatment diets

Variables	Dietary groups					SE	P value
	CONT	FERMA	FERMB	FERMC	FERMD		
pH	6.35^{c}	6.43^{b}	6.44^{b}	6.48^{ab}	6.51^{a}	0.02	<0.01
WHC (%)	37.2	35.8	36.1	36.1	36.4	0.42	0.18
Moisture (%)	74.5^{c}	75.1^{ab}	74.7^{bc}	74.4^{c}	75.3^{a}	0.15	<0.01
Crude protein (%)	22.5^{b}	23.1^{ab}	23^{ab}	23.2^{ab}	23.5^{a}	0.23	0.05
Crude fat (%)	0.54^{b}	0.51^{b}	0.63^{a}	0.48^{b}	0.52^{b}	0.03	<0.01
Ash (%)	0.67^{a}	0.59^{bc}	0.55^{c}	0.65^{ab}	0.63^{ab}	0.02	<0.01
L*	54.6	55.1	56.6	54.3	55.8	0.93	0.41
a*	-1.38^{bc}	-1.23^{b}	-2.22^{c}	-1.43^{bc}	-0.28^{a}	0.33	<0.01
b*	10.6^{a}	8.85^{b}	8.96^{b}	7.44^{b}	8.30^{b}	0.56	<0.01

[a,b,c]Means in the same row with various letters indicate notable differences (P<0.05): CONT: control diet based on corn-soybean- diet, FERMA: diet containing 1% FERM, FERMB: 2.5% FERM, FERMC: 5% FERM, FERMD: 7.5% FERM, SE: standard error of the mean, WHC: water holding capacity, L*: lightness values, a*: redness values, b*: yellowness values.

Table 3: Physical and chemical characteristics of thigh meat of the IICC fed treatment diets.

Variables	Dietary groups					SE	P value
	CONT	FERMA	FERMB	FERMC	FERMD		
pH	6.52^{c}	6.61^{b}	6.63^{b}	6.67^{ab}	6.74^{a}	0.03	<0.01
WHC (%)	36.1^{a}	34.2^{b}	35.3^{a}	34.0^{b}	31.5^{c}	0.3	<0.01
Moisture (%)	74.9^{b}	75.6^{a}	75.8^{a}	75.0^{b}	75.9^{a}	0.13	<0.01
Crude protein (%)	20.4^{b}	21.1^{a}	20.1^{b}	20.1^{b}	20.0^{b}	0.15	<0.01
Crude fat (%)	2.19^{a}	1.62^{b}	2.42^{a}	2.21^{a}	1.63^{b}	0.16	<0.01
Ash (%)	0.80^{a}	0.76^{a}	0.70^{b}	0.77^{a}	0.79^{a}	0.02	<0.01
L*	56.0	59.8	57.5	56.6	54.6	1.51	0.17
a*	1.3^{b}	0.79^{b}	1.68^{b}	1.91^{b}	3.73^{a}	0.64	0.02
b*	10.8^{a}	8.38^{ab}	8.71^{a}	9.39^{a}	6.19^{b}	0.79	<0.01

[a,b,c]Means in the same row with various letters indicate notable differences (P<0.05): CONT: control diet based on corn-soybean- diet, FERMA: diet containing 1% FERM, FERMB: 2.5% FERM, FERMC: 5% FERM, FERMD: 7.5% FERM, SE: standard error of the mean, WHC: water holding capacity, L*: lightness values, a*: redness values, b*: yellowness values.

Zhang *et al.* (2016) documented that the dietary incorporation of 6% fermented feed increased abdominal fat deposition of broiler chickens. In contrary, Nie *et al.* (2015) noticed that feeding fermented cottonseed meal decreased abdominal fat relative weight in broilers. Sugiharto and Ranjitkar (2019) suggested that the differences in the characteristics and proportions of fermented feed in the diets may account for such conflicting data above. Our present finding showed that feeding diets containing FERM particularly at the levels of 1 and 2.5% resulted in lower ash content in the breast meat of the IICC. In line with this present finding, Sugiharto *et al.* (2017) noticed the decreased crude ash content in breast meats when feeding *Acremonium charticola*-fermented feed to broiler chickens. We and also the latter authors could not explain the definite reason for such conditions so far.

Our data showed that dietary treatment did not have any effect on the lightness (L*) values of breast meat of the IICC. Interesting finding was seen in this current study, at which the redness (a*) of the IICC breast meats showed the negative values. According to Kralik *et al*. (2017) and Huang and Ahn (2018), the negative a* values indicate the green appearance of the meats. Lien *et al*. (2012) and Petracci and Cavani (2012) suggested that such green appearance in meat may mainly be due to the excessive wing-flapping or wing exercise in chickens, which may lead to green muscle disease. Interestingly, dietary treatment with FERMD seemed to alleviate the occurrence of the green muscle disease as indicated by the higher values of a* in the respective IICC breast meat. In this study, dietary treatment with FERM resulted in lower b* values in the IICC. Formerly, Allen *et al*. (1997) reported that there was a negative correlation between yellowness versus redness and pH values. Indeed, the higher pH may avoid protein denaturation and hence maintain the redness and yellowness values of breast meats.

In accordance with the breast meat, the pH values and moisture content of thigh meat increased with the increased levels of FERM in the IICC diets. In contrast to breast muscle, the WHC, however, decreased with the elevated levels of FERM in the diets. This finding was in contrast to that of formerly reported by Lee *et al*. (2017) showing the increased WHC of broiler breast meats with feeding fermented soybean hulls and *Pleurotus eryngii* stalk residue with *Aureobasidium pullulans*. Up to now, there is no definite explanation for these conflicting results above. In the present study, there was an increase in crude protein concentration in FERMA as compared to the other meats. The improved protein efficiency ratio leading to the increased protein production in the muscularity of the IICC may be the reason for the latter condition (Sugiharto *et al*., 2017). However, it should be noticed that the inclusion levels of FERM higher than 1% did not exert any impact on the protein content of the IICC thigh meat. With regards to the concentrations of crude fat and ash, the dietary treatment of FERM had a considerable impact on the concentrations of fat and ash in the IICC thigh meats, yet the alteration did not make a specific pattern. This may therefore be difficult to withdraw any inference. Our current finding showed that dietary treatments did not have any impact on the L* values of the IICC thigh meats. However, feeding FERM at the level of 7.5% increased and decreased the redness and yellowness of the IICC meats in the present work. In line with breast meats, the higher pH in the FERMD thigh meat seemed to alleviate the protein denaturation and thus maintain the redness and yellowness values of the IICC thigh meats.

Conclusions

Dietary inclusion of FERM especially at the level of 7.5% from diets improved the physical and chemical characteristics of the IICC meats.

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