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Research Article

Carcass and Meat Characteristics of Broilers Stocked at a High Density and Supplemented with Acidified Turmeric

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Abstract | The objective of this study was to investigate how acidified turmeric affected the meat quality of broilers that were maintained in high-density condition. Two hundred and eighty-five 14 days old Lohmann broiler chicks were randomly allotted to CONT (birds stocked at a density of 9 birds/m^2 and received control/basic feed), HSD (birds stocked at a density of 16 chicks/m^2 and received control feed), HSDT (birds stocked at a density of 16 chicks/m^2 and received feed supplemented with 1% turmeric powder) and HSDAT (birds stocked at a density of 16 chicks/m^2 and received feed supplemented with 1% acidified turmeric powder). Carcass determination and meat sample was collected at day 37. CONT meat had lower ($P < 0.05$) pH than other meats. HSDAT meats had greater ($P < 0.05$) redness (a*), whereas yellowness (b*) values were lower in CONT than other meats. CONT meat had higher ($P < 0.05$) unsaturated and saturated fatty acids (FA) than other meats. CONT had higher ($P < 0.05$) n-3 to n-6 FA ratio than HSDT, but did not differ ($P > 0.05$) from HSD and HSDAT meats. In conclusion, high stocking density adversely affected meat qualities of broilers. Dietary supplementation of acidified turmeric was capable of improving the physical and chemical characteristics of meats of broilers raised in a high density pen.

Keywords | Acidification, Broiler meat quality, High stocking density, Stress, Turmeric

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INTRODUCTION

From an economic standpoint, the efficient broiler farming system is credited with the high stocking density. However, in addition to the harmful effects on health and physiological circumstances, excessive stocking density may have a negative influence on broiler meat quality (Li et al., 2019). Chemical antioxidants have proven to be an effective method for improving broiler meat quality under stressful conditions. Consumers are, however, concerned about the residue of chemical antioxidants in broiler meats, which may cause cancer in humans (Sugiharto, 2020a). As a result, using natural antioxidant sources is favourable for food safety and long-term broiler production.

Turmeric has long been recognised as an antioxidant agent in addition to being a culinary spice. Despite its strong antioxidant ability, the use of turmeric as an antioxidant agent may not always give consistent results (Sugiharto, 2020a). The latter disadvantage is particularly due to turmeric's low bioavailability (Lopresti, 2018). Nutritional and functional characteristics of plant-derived products have been shown to be improved by acidification. According to Bayliak et al. (2016), acid treatment improved the antioxidant potentials of medicinal plants. The solubility and stability of curcumin also increases in acidic solution, which therefore increases its bioavailability to the host (Kharat et al., 2017). The fruit filtrate of *Averrhoa bilimbi* Linn. was used to acidify turmeric powder

in this current work. The *A. bilimbi* fruit is rich in organic acids, particularly citric acid (Sugiharto, 2020b). The use of *A. bilimbi* fruit filtrate to acidify turmeric powder was expected to enhance turmeric's bioavailability, making it a more efficient source of antioxidants for stressed broilers. Fruit filtrate from *A. bilimbi* was chosen since the fruit is available all year and may be harvested for free in some areas because it is an underused fruit (Sugiharto, 2020b).

In a recent work, we incorporated the *A. bilimbi* fruit filtrate-acidified turmeric into broiler feeds and observed that the treatment decreased abdominal fat content of broilers (Sugiharto et al., 2020a). However, no study has yet been done on the use of acidified turmeric on broilers under stressful situations. The objective of this study was to see how acidified turmeric affected the meat quality of broilers that were maintained in high-density condition.

MATERIALS AND METHODS

Preparation of acidified turmeric

The production of acidified turmeric began with the preparation of the filtrate of *A. bilimbi* from the ripe fruit of *A. bilimbi* collected from the garden near the faculty. Turmeric powder was purchased from the local market in Semarang city, Central Java, Indonesia. Acidified turmeric was prepared according to the method described by Sugiharto et al. (2020a). In brief, turmeric powder was mixed evenly with *A. bilimbi* fruit filtrate (1:3; g:mL), anaerobically cultured at room temperature (*ca* 25°C) for 4 days and then sun-dried. Samples of turmeric powder and acidified turmeric were obtained for the measurement of pH, total acid and antioxidant activity. The pH of acidified turmeric was determined using a handheld pH meter (OHAUS ST300). Total acidity was measured on the basis of the titration method. Antioxidant capacity was determined based on the 2, 2-diphenylpicrylhydrazyl (DPPH) free radical scavenging assay, and presented as IC_{50} (13), while the phenolic content was measured according to Folin-Ciocalteu method. The pH of turmeric powder was 5.90, with 2.34% total acidity, 70.9 ppm antioxidant activity, and 3.47% total phenols, whereas the pH of acidified turmeric was 3.70, with 6.41% total acidity, 81.2 ppm antioxidant activity, and 3.35% total phenols.

Chicken trial

The Animal Ethics Committee of the Faculty of Animal and Agricultural Sciences, Universitas Diponegoro approved the chicken trial (No. 57-05/A3/KEP/FPP). Two hundred and eighty-five 14 days old Lohmann broiler chickens (body weight of 370 ± 9.02 g) were randomly distributed to four groups with five replicates. The groups were CONT (birds stocked at a density of 9 birds/m^2 and received control/basic feed), HSD (birds stocked at a density of 16 chicks/m^2 and received control feed), HSDT (birds stocked at a density of 16 chicks/m^2 and received feed supplemented with 1% turmeric powder) and HSDAT (birds stocked at a density of 16 chicks/m^2 and received feed supplemented with 1% acidified turmeric powder). For the course of the study, the birds were raised in an open-sided broiler chicken house with rice husk bedding. Each pen was installed with a hand feeder and a drinker. A continuous lighting regimen was employed for the length of the study. While the birds were fed commercial starter feeds until day 14, the Indonesian National Broiler Feed Standard was used to formulate the basic feed (Table 1) that was given to the chicks from day 14. Turmeric powder or acidified turmeric powder was incorporated into the feed every day from the 14th day until harvest (37th day). In this study, the addition of 1% turmeric or acidified turmeric was not taken into account in the feed formulation, as turmeric was used as a non-nutritive feed additive for broiler chickens. On the fourth and eighteenth days, the chicks were vaccinated against Newcastle disease through the eyes and drinking water. A vaccine for infectious bursal disease was also given by drinking water on day 12.

Table 1: Ingredients and chemical components of feed (days 14-37).

Items	(%, unless otherwise noticed)
Yellow maize	58.7
Palm oil	2.90
Soybean meal	34.7
DL-methionine	0.19
Bentonite	0.75
Limestone	0.75
Monocalcium phosphate	1.20
Premix[1]	0.34
Chlorine chloride	0.07
Salt	0.40
Chemical components	
ME, (kcal/kg)[2]	3,000
Crude protein	20.0
Crude fiber	5.52
Ca	1.00
P (available)	0.56

[1]Provided per kg of feed: 1,100 mg Zn, 1,000 mg Mn, 75 mg Cu, 850 mg Fe, 4 mg Se, 19 mg I, 6 mg Co, 1,225 mg K, 1,225 mg Mg, 1,250,000 IU vit A, 250,000 IU vit D_3, 1,350 g pantothenic acid, 1,875 g vit E, 250 g vit K_3, 250 g vit B_1, 750 g vit B_2, 500 g vit B_6, 2,500 mg vit B_{12}, 5,000 g niacin, 125 g folic acid and 2,500 mg biotin. [2]ME (metabolizable energy) was calculated according to formula: 40.81 (0.87 (crude protein + 2.25 crude fat + nitrogen-free extract) + 2.5).

Two male chicks reflecting the average weight of each cage (10 birds/group) were obtained on the 37th day, and then

slaughtered, de-feathered and dissected. Subsequently, each broiler's carcass and commercial proportions were determined. To determine the physical and chemical indices as well as colour of the meats, samples of breast meat were obtained and frozen at -10°C.

Following the procedure described by Sugiharto et al. (2020b), the frozen meat was first thawed at room temperature for around 30 minutes before being analyzed. Prior to analysis, any water from the melted ice crystals was gently removed using laboratory tissue paper. One gram of breast meat was homogenized with 9 mL of distilled water, and the pH of the resultant filtrate was determined with a electric pH meter (Hanna Instruments, Woonsocket, Rhode Island). The water holding capacity (WHC) of meat was determined using press techniques employing filter paper, and the chemical composition of meat was determined using standard proximate analysis. The meat sample was put in a plastic bag and cooked for 1 hour in boiling water at 80°C to evaluate the cooking loss. The sample was then weighed after it had been allowed to cool to room temperature. Cooking loss was defined as the difference in weight between before and after the cooking. The colour of meat was checked with a digital colour meter on Mac OS X. (set to CIE Lab). The colour was represented using the L* (lightness), a* (redness), and b* (yellowness) values.

Fatty acid contents in breast meats were determined using a conventional gas chromatography technique. By comparing the retention times of each sample to the usual retention times, the presence of fatty acids was determined. Using a lipid conversion factor, the area percentage was normalized and converted to g per 100 g of edible portion for fatty acid measurement. Total saturated fatty acids (SFA) and unsaturated fatty acids (UFA) were calculated by adding each component of SFA and UFA separately.

Statistical analysis

The data were treated using analysis of variance (ANOVA, SPSS 16.0 version). The Duncan multi-range test was employed when dietary treatments showed a significant effect ($P < 0.05$).

RESULTS AND DISCUSSION

The data on carcass indices of broilers are shown in Table 2. Generally, carcass traits and commercial cuts of broilers were not influenced ($P > 0.05$) by the treatments applied. Table 3 displays the physical and chemical indices of meats. CONT had lower pH values ($P < 0.05$) than the other breast meats, with HSDAT having the highest pH values. The WHC in HSDAT was lower ($P < 0.05$) than in CONT, HSD, and HSDT meats. Cooking loss in CONT was higher ($P < 0.05$) than in HSD and HSDT meats, but not different from HSDAT meats. In HSDAT, breast meats had greater a* values ($P < 0.05$) than other broiler meats, whereas b* values in CONT were lower ($P < 0.05$) than other meats. In comparison to other broiler meats, HSDT has lesser crude protein ($p < 0.05$). The HSD meat had less crude fat ($P < 0.05$) than the other meats.

Table 2: Carcass and commercial cuts of broilers.

Items	CONT	HSD	HSDT	HSDAT	SEM	P value
Eviscerated carcass (% live BW)	68.3	67.7	68.8	69.4	0.35	0.34
% eviscerated carcass						
Breast	38.5	37.1	37.5	37.2	0.63	0.87
Wings	10.6	10.8	11.1	10.7	0.19	0.82
Thigh	15.3	15.5	15.1	15.7	0.23	0.83
Drumstick	14.2	14.5	14.5	15.0	0.24	0.74
Back	21.4	22.1	21.9	21.4	0.32	0.86

CONT: birds stocked at a density of 9 birds/m^2 and received control/basic feed, HSD: birds stocked at a density of 16 chicks/m^2 and received control feed, HSDT: birds stocked at a density of 16 chicks/m^2 and received feed supplemented with 1% turmeric powder, HSDAT: birds stocked at a density of 16 chicks/m^2 and received feed supplemented with 1% acidified turmeric powder, BW: body weight, SEM: standard error of the means

Table 3: Physical and chemical variables of broiler breast meats.

Items	CONT	HSD	HSDT	HSDAT	SEM	P value
pH	6.76^{c}	6.83^{b}	6.87^{ab}	6.90^{a}	0.01	<0.01
WHC (%)	40.0^{a}	40.5^{a}	40.5^{a}	33.5^{b}	1.02	0.03
Cooking loss(%)	31.2^{a}	$30.2b^{b}$	30.0^{b}	31.0^{ab}	0.16	0.02
Meat colour						
L*	48.5	48.1	48.7	48.1	0.28	0.82
a*	2.49^{b}	2.87^{b}	3.08^{b}	5.45^{a}	0.30	<0.01
b*	4.99^{c}	7.11^{a}	6.12^{b}	7.08^{a}	0.17	<0.01
Moisture (%)	74.9	74.8	74.8	74.7	0.06	0.61
Crude protein(%)	23.5^{a}	23.2^{a}	22.2^{b}	23.1^{a}	0.10	<0.01
Crude fat (%)	0.78^{a}	0.69^{b}	0.81^{a}	0.78^{a}	0.02	0.04

^{a,b,c}Means in the same row with superscript letters are significantly different ($P<0.05$). CONT: birds stocked at a density of 9 birds/m^2 and received control/basic feed, HSD: birds stocked at a density of 16 chicks/m^2 and received control feed, HSDT: birds stocked at a density of 16 chicks/m^2 and received feed supplemented with 1% turmeric powder, HSDAT: birds stocked at a density of 16 chicks/m^2 and received feed supplemented with 1% acidified turmeric powder, L*: lightness values, a*: redness values, b*: yellowness values, SEM: standard error of the means.

The fat content of CONT meat was higher ($P < 0.05$) than that of other broiler meats in terms of both unsaturated and saturated fat (Table 4). CONT meat had higher ($P <$

0.05) levels of n-6 fatty acids than HSDT and HSDAT meats, but was similar to HSD meat. CONT had more (P < 0.05) n-3 fatty acids than other meats. CONT had a higher (P<0.05) ratio of n-3 to n-6 fatty acids than HSDT, but no difference between HSD and HSDAT meats.

It is widely known that raising broiler chickens at a high density pen resulted in stress conditions that detrimentally affected not only to the physiological conditions, but also the carcass and meat quality of broilers chickens (Sugiharto, 2022). In the current investigation, CONT meat had lower pH values than the other breast meats, with HSDAT having the highest pH values. There are several factors enhancing the pH values of meats post slaughter, one of which is the depletion of stored glycogen in the muscle of broilers. The less glycogen available post-mortem may affect the normal acidification process (postmortem anaerobic glycolysis) and thereby leaving the pH of meat high (Adzitey and Nurul, 2011). When compared to meats from broilers maintained at a normal density, the long-term stress due to high stocking density condition may be related to reduced muscle glycogen storage, resulting in higher pH values of meats post-mortem. Apart from the difference in pH values, all meats tested in this study had pH values more than 6.0. According to Mir et al. (2017), meats with a pH greater than 6.0 experienced less protein denaturation. However, meats with pH levels greater than 6.0 are more likely to develop dark firm dry (DFD) meats (Adzitey and Nurul, 2011), which may reduce customer acceptance. In general, there is a positive link between broiler meat pH and WHC, with greater pH corresponding to increased WHC (Mir et al., 2017). In this study, however, the higher pH values of meats from high-stocked broilers treated with acidified turmeric were accompanied with decreased WHC of the corresponding meats. The lowering of WHC in meats has been linked to the post-mortem denaturation of muscle protein (Adzitey and Nurul, 2011). Given that higher pH levels are associated with less denaturation of muscle protein, the lower WHC of meats obtained from high-stocked broilers treated with acidified turmeric appeared to be incongruent. As mentioned above, the higher meat pH values in this study revealed that the high-stocked broilers fed with acidified turmeric had lower muscle glycogen concentrations. According to El-Senousey et al. (2013), a modest amount of glycogen causes actomyosin linkages to form, thus speeding up the rigor mortis process. Indeed, during rigor mortis, the synthesis of actomyosin causes water molecules to shrink, resulting in fluid secretion in muscles and a drop in WHC. In this investigation, cooking loss was reduced in meats taken from broilers raised under high stocking density induced stress compared to meats collected from broilers reared at a normal density. This finding contradicted the recorded pH and WHC values, as cooking loss is generally inversely proportional to pH and WHC (Mir et al., 2017). The explanation for the reduced cooking loss in broiler meats with high stocking density condition was unknown, however less denaturation of muscle protein (as mentioned above) appeared to explain the lower cooking loss in the corresponding groups' meats.

The L* and a* values of broiler meats did not differ between normal and high stocking density conditions in this investigation. However, when stocking density was high, the b* values of broiler meats increased. This finding differed from that of Goo et al. (2019), who found no effect of stocking density on broiler meat colour (L*, a*, and b* values). Yang and Chen (1993) previously reported that muscle pH has a positive connection with yellowness. The higher b* values in high-stocked broiler meats than in normal-stocked broiler meats might be explained in this investigation because the pH values rose with increasing stocking density. The impact of pre-slaughter stress on rigor mortis may explain the higher pH and yellow colour of high-stocked broiler meats in this aspect (da Silva et al., 2017). Treatment with non-acidified turmeric powder had no effect on the redness values of broiler meat in this study, regardless of the stocking density effect. In line with this finding, Baghban et al. (2017) found that supplementing broiler meats with 0.5% turmeric powder had no effect on the redness of the meats. In contrast, Zhang et al. (2015)

Table 4: Fatty acid compositions of broiler breast meats.

Items	CONT	HSD	HSDT	HSDAT	SEM	P value
Total SFA (g/100 g)	0.53^{a}	0.42^{b}	0.46^{b}	0.45^{b}	0.01	0.01
Total UFA (g/100 g)	0.85^{a}	0.72^{b}	0.59^{c}	0.60^{c}	0.02	<0.01
UFA/SFA	1.64	1.72	1.37	1.37	0.06	006
n-3 PUFA (mg/100 g)	22.2^{a}	8.40^{b}	1.54^{b}	6.05^{b}	2.57	0.02
n-6 PUFA (mg/100 g)	209^{a}	194^{ab}	163^{bc}	148^{c}	6.67	<0.01
n-3/n-6 PUFA	1.06^{a}	0.04^{ab}	0.01^{b}	0.04^{ab}	0.12	0.04

^{a,b,c}Means in the same row with superscript letters are significantly different (P<0.05). CONT: birds stocked at a density of 9 birds/m^2 and received control/basic feed, HSD: birds stocked at a density of 16 chicks/m^2 and received control feed, HSDT: birds stocked at a density of 16 chicks/m^2 and received feed supplemented with 1% turmeric powder, HSDAT: birds stocked at a density of 16 chicks/m^2 and received feed supplemented with 1% acidified turmeric powder, SFA: saturated fatty acids, UFA: unsaturated fatty acids, PUFA: polyunsaturated fatty acids, SEM: standard error of the means.

revealed that curcumin (a bioactive component in turmeric) increased the redness values of broiler meats. Interestingly, acidified turmeric powder evidently increased the redness values of broiler meats in this study. This may suggest that acidification may increase the utilization of curcumin in turmeric powder by broilers. According to Sugiharto et al. (2020a), the acid property of acidified turmeric powder appeared to be beneficial in avoiding myoglobin breakdown in muscle tissues and so boosting the redness of broiler meats. Furthermore, the acid action of acidified turmeric powder improved protein digestion in proventriculus, leading to enhanced muscle protein deposition in broiler chicken meats. The latter condition may thereby increase the redness values of broiler meats. When compared to meats from broilers fed acidified turmeric and a control diet, the treatment with turmeric powder reduced the yellowness values of meats from birds raised in high stocking density conditions. Previous study by Johannah et al. (2018) found no effect of turmeric extract on the yellowness values of broiler meat. The latter authors suggested that the yellowness score of broiler meats was attributed to the curcuminoids absorption from turmeric. For this reason, the lower yellowness values in the HSDT than that of particularly to HSDAT seemed to be associated with the lower absorption of curcuminoids from the turmeric than that from acidified turmeric powder. However, this inference should be taken with caution since there was no substantial difference in the yellowness values between broiler meats from HSDAT and HSD groups.

Regardless of the stocking density effect, adding turmeric powder to the feed reduced the crude protein content of broiler breast meats. This contradicted the findings of Baghban et al. (2017), who reported that feeding turmeric powder had no influence on the crude protein level of broiler meats. The exact cause for turmeric powder's reduced influence on meat's crude protein level is unknown. Turmeric, as previously reported, has been linked to liver damage in a research by Luber et al. (2019). Because the liver is one of the most critical organs for protein production, any abnormality in the lever may reduce protein production. In agreement, Attia et al. (2017) found that supplementing broilers with turmeric (1 and 2 g/kg diets) reduced blood total protein levels. The protein-lowering impact of acidified turmeric powder on broilers, on the other hand, was not observed, implying that acidified turmeric powder can be used safely for broilers. The crude fat content of broiler breast meats was shown to be lower when broiler chicks were kept in high density pens. A previous study in geese found that increased stocking density reduced the intramuscular fat content of breast meat (Wang et al., 2019). The later researchers also claimed that high-stocking density reduced intramuscular fat content, which they believe is attributable in part to the low fat deposit produced by low calorie intake as a result of reduced feed intake under high density.

The total SFA content of meats reduced with higher stocking density, according to the findings of the current study. In contrast, Simsek et al. (2009) found that total SFA levels in broiler breast meats increased as stocking density increased. Reduced feed digestibility resulting in poorer calorie intake were linked to stress conditions (Sugiharto, 2020a). As a result, fat deposition in the broiler's body may be reduced. According to Simsek et al. (2009), there is a positive association between saturated fats and fatness or fat deposition in the body. The levels of UFA and n-3 PUFA in broiler breast meats were shown to be lower when broilers were stocked in high density pens. Simsek et al. (2009) found that increasing stocking density resulted in lower PUFA and n-3 PUFA concentrations in broiler breast meats. The addition of turmeric or acidified turmeric powders to the diet lowered the UFA level of meats much further in this trial. There are conflicting results when it comes to the content of UFA in meats when turmeric or curcumin is fed. Galli et al. (2020) documented that dietary supplementation of curcumin increased PUFA percentage in broiler meats, while Hang et al. (2018) found that feeding curcumin at 20 and 40 mg/kg decreased total n-3 PUFA without affecting the contents of total MUFA and PUFA of chicken breast meats. According to the latter researchers, curcumin may inhibit microsomal 5 and 6 desaturases in the liver, thus reducing the synthesis of longer chain PUFA in breast meats. However, the latter inference should be interpreted with careful as there was no difference in the total n-3 PUFA of meats between high-stocked broilers fed control diet and high-stocked broilers fed diets containing either turmeric or acidified turmeric powders. It was most likely that the decrease in n-6 PUFA contributed to the decreased UFA contents in the breast meats of turmeric or acidified turmeric fed broilers, although other studies found no effect of curcumin treatment on the meat content of n-6 PUFA (Hang et al., 2018). The n-3/n-6 PUFA ratio was significantly reduced in meats from high-stocked broilers fed turmeric powder as compared to control, but the difference was not significant in meats from high-stocked broilers fed acidified turmeric powder. Given that a high n-3/n-6 PUFA ratio is better for consumer health, dietary supplementation with acidified turmeric appeared to be more advantageous in terms of consumer health.

CONCLUSIONS AND RECOMMENDATIONS

High stocking density adversely affected physical and chemical qualities of broiler meats. Acidified turmeric powder increased pH and redness values of meats of broilers at high stocking density pens. Acidified turmeric

powder seemed to be more advantageous than turmeric powder in improving the physical and chemical traits of broiler meats.

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NOVELTY STATEMENT

This is the first study revealing the benefit of acidified turmeric in improving the physical and chemical characteristics of meats of broilers raised under stressful conditions.

AUTHOR'S CONTRIBUTION

MKR conducted experiment and drafted the manuscript, VPB and SK revised the manuscript, SS designed the experiment and revised manuscript.

Conflict of interest

The authors have declared no conflict of interest.

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