PE10.02 The effect of dietary supplementation of the mixture of gallic and linoleic acid on broiler breast meat quality 61.00

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***Abstract*- Polyunsaturated fatty acids (PUFAs) are well known for the element which has the beneficial effects on human health. However, PUFAs are prone to oxidation. In the present study, we studied the effect of mixture of gallic and linoleic acid (MGL) dietary supplement on the breast meat quality of broiler. A total of 90 broilers were assigned to 3 groups with 3 replications. Broilers received 3 dietary treatments (d 22-36): 1) commercial finisher diet (CFD, control diet), 2) CFD + MGL (0.5%/CFD, 1:1, M:M, 0.5% MGL diet), 3) CFD + MGL (1.0%/CFD, 1:1, M:M, 1.0% MGL diet). Lipid oxidation of the breast meat was inhibited by 1.0% MGL diet ($p<0.05$). In the fatty acid composition of the breast meat, arachidonic acid and docosahexaenoic acid was higher in both MGL diets than that of control diet ($p<0.05$). In addition, water holding capacity of the breast meat from 1.0% MGL diet was higher than that of control diet ($p<0.05$). In conclusion, 1.0% MGL diet may improve the quality of the breast meat of broiler.**

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***Index Terms*—Dietary supplementatoin, Gallic acid, Linoleic acid, Meat quality.**

I. INTRODUCTION

Recently there has been an increased interest in food containing high amount of polyunsaturated fatty acids (PUFAs) because PUFAs have shown the beneficial effects on human health such as the prevention of coronary heart disease and other chronic diseases [1]. Linoleic acid (LA) is converted to arachidonic acid and has anti-inflammatory effect by decreasing the secretion of interleukin (IL)-6 and -1β, and the tumor necrosis factor α [2]. Previous study reported that high levels of dietary LA suppressed lymphocyte proliferation in rats [3]. However, PUFAs are prone to oxidation and their oxidation products are leading to deterioration of meat quality such as flavor, color, texture and nutritional value.

The negative outcome of lipid oxidation in chicken meat can be subdued by the use of diet containing antioxidants such as medicinal herb mix and grape pomace, which are natural antioxidant with rich polyphenols. The interest in natural antioxidant was increased because they are usually considered safer than synthetic antioxidant, and have greater application potential for consumer acceptability, palatability, stability and shelf-life of meat products [4]. Gallic acid (GA) is a representative natural polyphenol which shows a strong antioxidant activity and possesses anti-carcinogenic, anti-mutagenic, anti-allergic, and anti-inflammatiry activities [5]. The objective of this study was to evaluate the effect of dietary supplementation of the mixture of GA and LA on the quality and antioxidative potential of the breast meat of broiler.

II. MATERIALS AND METHODS

Animal and experimental design

A total of 90 one-day old male and female broilers (Ross strain) were obtained from a commercial hatchery. Broilers were free accessed to water and diet. After 3 week, broilers were weighed and reassigned according to average weight. Each groups containing 10 broilers, to receive 3 dietary treatments (d 22-36) with 3 replicates of each treatment: 1) commercial finisher diet (CFD, control diet), 2) CFD + M (0.5%/diet, 1:1, M:M, 0.5% MGL diet), 3) CFD + mixture of GA and LA (1.0%/diet, 1:1, M:M, 1.0% MGL diet). At the end of the experimental period (d 36 of age), 21 broilers from each treatment were killed in the stable by the carotid amputation and vacuum packed carcasses were stored by deep freezer at -50° until the use of each analysis.

Measurement of antioxidative potential

Total phenolic content

Each meat sample (3 g) in distilled water (15 mL) was homogenized (T25b, Ika Works (Asia)., Sdn, Bhd, Malaysia) at 1,130 × *g* for 1 min. Chloroform (10 mL) was added to the homogenates and the mixture was shaken vigorously 2 to 3 times to separate the lipids and the aqueous supernatant was separated by centrifuge (Hanil) at 2,090 × *g* for 15 min, which was used for measurement of total phenolic content, 1,1-Diphenyl-2-picrylhydrazyl (DPPH) radical scavenging activity.

Total phenols content in the aqueous supernatant was estimated by the Folin-Ciocalteu method [6]. A 0.1 mL aqueous supernatant was added to the Folin-Ciocalteu reagent (0.2 mL), followed by the addition of 3 mL sodium carbonate solution (5%). The reaction mixture was vortexed and the absorbance was measured with a spectrophotometer (DU 530, Beckman Instruments Inc., Fullerton, CA, USA) at 765 nm after incubation for 1 h at 23℃. Quantification was done based on the standard curve generated with gallic acid and expressed gallic acid equivalent.

DPPH radical-scavenging assay

DPPH radical scavenging activity was estimated according to the method of Blois (1958) with slight modifications. A 200 μL quantity of aqueous supernatant was added to 800 μL distilled water and 1 mL methanolic DPPH solution (0.2 m*M*). The mixture was vortexed and left to stand at room temperature (20-22°) for 30 min. A tube containing 1 mL distilled water and 1 mL methanolic DPPH solution (0.2 m*M*) served as the control. The absorbance of the solution was measured at 517 nm using a spectrophotometer (Beckman). The percentage of DPPH radical scavenging was obtained from the following equation:

Radical scavenging activity = [1 − (absorbance of sample / absorbance of control)] × 100.

2-Thiobarbituric acid-reactive substances (TBARS)

Each meat sample (5 g) in 15 mL distilled water was homogenized (Ika Works) at 1,130 × *g* for 1 min. Sample homogenate (1 mL) was transferred to a test tube and lipid oxidation was determined as the **TBARS** value by using the method described by Ahn et al. (1999). Briefly, 50 μL butylated hydroxyanisol (7.2%) and 2 mL TBA-trichloroacetic acid solution (20 m*M* TBA in 15% trichloroacetic acid) were added to the test tube. Tubes were heated in a boiling water bath for 30 min, cooled, and then centrifuged at 2,090 × *g* for 15 min. Absorbance of the supernatant was measured at 532 nm with a spectrophotometer (Beckman). The increase in absorbance at 532 nm was taken into consideration to calculate the TBARS values. Lipid oxidation development was reported as milligrams of malondialdehyde per kilogram of meat.

Fatty acid composition

Total lipids of samples were extracted by using chloroform-metananol (2:1, v/v) according to the procedure of Folch et al, (1957). The fatty acid methyl esters were prepared from the extracted lipids with BF_3-methanol (Sigma-aldrich). The fatty acid methyl esters were separated on a gas chromatograph (Agillent GC 6890N, Palo Alto, CA, USA) equipped with a mass selective detector (MSD). A split inlet (split ratio, 50:1) was used to inject samples into a HP-5MS capillary column (30 m×0.25 mm×0.25°), and ramped oven temperature was used (150° for 3 min, increased to 180° at 2.5°/min and maintained for 5 min, increased to 220°at 2.5°/min and maintained for 25 min). Inlet temperature was 210°. Helium was the carrier gas at constant flow of 0.7 mL/min. The temperature of the mass spectrometer (MS) source, MS quadrupole, and the transfer line into the MS were 230, 150, and 280° respectively. The fatty acid composition was identified by a mass spectrum database (NIST Library, mass spectral search program, version 5.0, Ringoes, NJ, USA).

Proximate analysis and water holding capacity (WHC)

The moisture, crude fat, crude protein, and crude ash composition of the breast meat was determined according to AOAC (1999) methods. WHC analysis was following to this procedure. One gram of the minced breast meat of broiler was placed on a round filter paper (NO.4, Whatman Ltd. Kent, UK)). The filter paper with meat was into centrifuge tube and then this tube centrifuged (CR 20B2, Hitachi koki Co., Ltd. Fukuoka, Japan) at 6,710 × *g* for 10 min. The released water content was measured and calculated as a percentage of the initial moisture of meat.

Statistical analysis

All experiments were duplicated with three observation numbers adapted for each experiment. Analysis of the variance was performed using the raw data, and the mean values and standard deviation were calculated by the Statistical Analysis System (SAS, 2000). Differences among the mean values were determined by the Duncan's multiple range test with a significance defined at $p<0.05$.

III. RESULTS AND DISCUSSION

Antioxidant activity

The DPPH radical scavenging activity of the breast meat from broiler fed 1% MGL diet was significantly higher than that of the broiler fed control diet during whole storage period (Table 1), whereas no significant difference was found in the breast meat between the control and 0.5 % MGL diet. MGL diet (1.0%) was appeared to delay the lipid oxidation of the breast meat during whole storage period (Table 2), and 0.5% MGL diet inhibited the lipid oxidation of the breast meat until storage d 2. Meat from broiler fed MGL diet produced low levels of TBARS in spite of the fact that MGL diet contained LA, which is PUFA that generated several types of free radicals and then accelerate lipid peroxidation [11]. The breast meat of broiler fed 1.0% MGL diet had significantly higher polyphenol content than that of control (Table 3), but that of 0.5% MGL diet was not significantly different with that of control except for the sample stored 2 days. In the presented study, various analysis of antioxidant activity were conducted to elucidate the potential antioxidant effect of dietary MGL, which is containing GA with LA. Previous study reported that GA is a polyphenyl natural product from gallnut, green tea, and grape and may directly combine with free radicals and lead to inactivate them which may decrease the intracellular concentration of free radicals [12]. Schwarz et al. (2009) reported that reactive oxygen species, including free radicals play a key role in the oxidation process that can damage cells, whereas polyphenols have been shown to scavenge free radicals such as superoxide, peroxyl and hydroxyl radicals, and hence influence on the redox mechanisms that may lead to degenerative diseased conditions such as alzheimer's, atherosclerosis, diabetes, and certain cancers [14].

Fatty acid composition

The concentrations of palmitic acid and oleic acid in the breast meat were significantly lower in the broiler fed 1.0% MGL diet than that of the control diet (Table 4). These results were probably due to the endogenous synthesis in the broiler tissue. The saturated fatty acids (SFAs) in bird tissues rely upon their presence in the diet and their synthesis in the liver and the SFAs synthesis are inhibited in the liver in greater during digestion of unsaturated fats than saturated fats [15]. The increase of PUFAs decreased the synthesis of monounsaturated fatty acids (MUFAs) by inhibiting the activity of the 9-desaturase complex which is the key enzyme need to convert SFAs to MUFAs [16]. Arachidonic acid (AA) in the breast meat was significantly increased by 1.0% MGL diet that could be due to LA in MGL diet, and LA is precursor of AA. Docosahexaenoic acid (DHA) is responsible for hypolipidemic and neuroprotective effect [17]. However, DHA is very sensitive to oxidative compounds that can change their pharmacological properties [18]. In the present study, DHA levels of both MGL diet in the breast meat were significantly higher than that of control diet. These results may be due to predominant antioxidative effect of GA in MGL diet. Results obtained in this experiment showed that PUFAs ratio in the breast meat was increased by both MGL diet compared to that of control diet. It can be attracted to consumers on human nutritional requirements, because high PUFAs ratio can give beneficial effect to human, mainly in the protection against cardiovascular disease [1].

Proximate composition and water holding capacity (WHC)

Crude fat of the breast meat from broiler fed 1.0% MGL was significantly higher than that of control (Table 5). The WHC of the breast meat was significantly improved by dietary 1.0% MGL. Previous study reported that the WHC in the raw meat was especially affected by the content and distribution of intramuscular fat, since the presence of intramuscular fat inhibits moisture diffusivity coefficient [19], and may be correlated to the tenderness of meat [20]. These results suggested that the breast meat of 1.0% MGL diet may be more tender than that of control

IV. CONCLUSION

Lipid oxidation of the breast meat was inhibited by 1.0% MGL diet, although PUFAs ratio of the breast meat was increased by both MGL diets. The composition of AA and DHA in the breast meat was higher in both MGL diets than that of control diet. In addition, WHC of the breast meat from 1.0% MGL diet was higher than that of control diet. These results suggested that 1.0% MGL diet may improve the quality of the breast meat of broiler.

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REFERENCES

[1] Krauss, R. M., Eckel, R. H., Howard, B., Appel, L. J., Daniels, S. R., Deckelbaum, R. J., Erdman, J. W., Kris-Etherton, P., Goldberg, I. J., Kotchen, T. A., Lichtenstein, A. H., Mitch, W. E., Mullis, R., Robinson, K., Wylie-Rossett, J., Jeor, S. S., Suttie, J., Tribble, D. L., & Bazzarre, T. L. (2001). Revision 2000: Statement for healthcare professionals from the nutrition committee of the American Heart Association. The Journal of Nutrition, 131(1), 132-146.

[2] Zhao, G., Etherton, T. D., Martin, K. R., Vanden Heuvel, J. P., Gillies, P. J., West, S. G., & Kris-Etherton, P. (2005). Anti-inflammatory effects of polyunsaturated fatty acids in THP-1 cell. Biochemical and Biophysical Research Communications, 336(3), 909-917.

[3] Yaqoob, P., Newsholme, E. A., & Calder, P. C. (1994). The effect of dietary lipid manipulation on rat lymphocyte subsets and proliferation. Immunology, 82(4), 603-610.

[4] Naveena, B. M., Sen, A. R., Vaithiyanathan, S., Babji, Y., & Kondaiah, N. (2008). Comparative efficacy of pomegranate juice, pomegranate rind powder extract and BHT as antioxidants in cooked chicken patties. Meat Science, 80(4), 1304-1308.

[5] Jo, C., Jeong, I. Y., Lee, N. Y., Kim, K. S., & Byun, M. W. (2006). Synthesis of a novel compound from gallic acid and linoleic acid and its biological functions. Food Science and Biotechnology, 15(2), 317-320.

[6] Subramanian, K. N., Padmanaban, G., and Sarma, S. (1965). Folin-Ciocalteu reagent for the estimation of siderochromes. Analytical Biochemistry, 12(1), 106-112.

[7] Blois, M. S. (1958). Antioxidant determination by the use of a stable free radical. Nature, 181,1199-1200.

[8] Ahn, D. U., Olson, D. G., Jo, C., Love, J., and Jin, S. K. (1999). Volatiles production and lipid oxidation on irradiated cooked sausage as related to packaging and storage. Journal of Food Science, 64(2), 226-229.

[9] Folch, J., Lees, M., & Sloane Stanley, G. H. (1957). A simple method for the isolation and purification of total lipides from animal tissues. The Journal of Biological chemistry, 226: 497–509.

[10] AOAC. (1999). Official methods of analysis. 15th ed. P. Cunniff, ed. Assoc. Off. Anal. Chem., Washington, DC.

[11] Nagendra Prasad, k., Yang, B., Yang, S., Chen, Y., Zhao, M., Ashraf, M., & Jiang, Y. (2009). Identification of phenolic compounds and appraisal of antioxidant and antityrosinase activities from litchi (*Litchi sinensis* Sonn.) seeds. Food Chemistry, 116(1), 1-7.

[12] Priscilla, D. H., & Prince, P. S. M. (2009). Cardioprotective effect of gallic acid on cardiac troponin-T, cardiac marker enzymes, lipid peroxidation products and antioxidants in experimentally induced myocardial infarction in Wistar rats. Chemico-Biological interactions, 179(2-3), 118-124.

[13] Schwarz, M., Rodríguez, M., Martínez, C., Bosquet, V., Guillén, D., & Barroso, C. C. (2009). Antioxidant activity of Brandy de Jerez and other aged distillates, and correlation with their polyphenolic content. Food Chemistry, 116(1), 29-33.

[14] Hogan, S., Zhang, L., Li, J., Zoecklein, B., & Zhou, K. (2009). Antioxidant properties and bioactive components of Norton (*Vitis aestivalis*) and Cabernet Franc (*Vitis vinifera*) wine grapes. LWT-Food Science and Technology, 42(7), 1269-1274.]

[15] Simm, J. S., & Qi, G. H. (1995). Designing poultry products using flaxseed. in Thompson LU, Cunnane S (eds): *Flaxseed* in Human Nutrition, Champaign, American Oil Chemist's Society Press, (pp.315–333).

[16] Pinchasov, Y., & Nir, I. (1992). Effect of dietary polyunsaturated fatty acid concentration on performance, fat deposition, and carcass fatty acid composition in broiler chickens. Poultry Science, 71(9), 1504-1502.

[17] Rodrigues de Turco, E. B., Belayev, L., Liu, Y., Busto, R., Parkins, N., Bazan, N. G., & Ginsberg, M. D. (2002). Systemic fatty acid responses to transient focal cerebral ischemia: influence of neuroprotectant therapy with human albumin. Journal of Neurochemistry, 83(3), 515-524.

[18] Judé, S., Bedut, S., Roger, S., Pinault, M., Champeroux, P., White, E., & Le Guennec, J. Y. (2003). Peroxidation of docosahexaenoic aicd is responsible for its effects on I_{TO} and I_{SS} in rat ventricular myocytes. British Journal of Pharmacology, 139(4), 816-822.

[19] Muriel, E., Ruiz, J., Martín, L., Petron, M. J., & Antequera, T. (2004). Physico-chemical and sensory characteristics of dry-cured loin from different Iberian pig lines. Food Science and Technology international, 10(2), 117-123.

[20] Lee, Y. S., Saha, A., Xiong, R., Owens, C. M., & Meullenet, J. E. (2008). Changes in broiler breast fillet tenderness, water-holding capacity, and color attributes during long-term frozen storage. Journal of Food Science, 73(4), 162-168.

Table 1. DPPH radical scavenging activity of the breast meat from broiler fed mixture of gallic and linoleic acid during storage at 4 °.

Treatment	Storage (day)				
	0	2	4	7	SEM[1]
Control	67.77^{ay}	68.18^{ay}	66.60^{aby}	64.49^{by}	0.716
0.5 %	70.01^{axy}	69.00^{axy}	68.28^{ay}	64.48^{by}	0.718
1 %	72.21^{ax}	70.19^{bx}	70.48^{bx}	67.99^{cx}	0.480
SEM[2]	0.738	0.362	0.564	0.827	

[1]Standard errors of mean (n=12). [2](n=9).
[a-c]Different letters within the same row differ significantly ($p<0.05$).
[x,y]Different letters within the same column differ significantly ($p<0.05$).

Table 2. 2-thiobarbituric acid reactive substances (TBARS) value of the breast meat from broiler fed mixture of gallic and linoleic acid during storage at 4 °

Treatment	Storage (day)				
	0	2	4	7	SEM[1]
Control	0.29^{cx}	0.32^{cx}	0.39^{bx}	0.49^{ax}	0.019
0.5 %	0.23^{by}	0.27^{by}	0.43^{ax}	0.47^{ax}	0.013
1 %	0.23^{cy}	0.26^{by}	0.33^{ay}	0.35^{ay}	0.009
SEM[2]	0.017	0.012	0.014	0.014	

[1]Standard errors of mean (n=12). [2](n=9).
[a-c]Different letters within the same row differ significantly ($p<0.05$).
[x,y]Different letters within the same column differ significantly ($p<0.05$).

Table 3. Total phenolic content (mg GAE/g meat) of the breast meat from broiler fed mixture of gallic and linoleic acid during storage at 4 °.

Treatment	Storage (day)				
	0	2	4	7	SEM[1]
Control	1.48^{by}	1.53^{ay}	1.51^{aby}	1.52^{ay}	0.010
0.5% MGL	1.50^{bxy}	1.58^{ax}	1.58^{axy}	1.54^{aby}	0.019
1.0% MGL	1.54^{bx}	1.60^{ax}	1.60^{ax}	1.62^{ax}	0.014
SEM[2]	0.011	0.007	0.019	0.017	

[1]Standard errors of mean (n=12). [2](n=9).
[a,b]Different letters within the same row differ significantly ($p<0.05$).
[x-z]Different letters within the same column differ significantly ($p<0.05$).

Table 4. Profile of fatty acids (% of total fatty acids) found in the breast meat from broiler fed mixture of gallic and linoleic acid after 35 days

Fatty acids	Breast meat			
	Control	0.5% MGL	1.0% MGL	SEM[1]
C16:0	23.12[a]	22.93[ab]	22.31[b]	0.150
C16:1	2.56	2.11	2.30	0.175
C18:0	15.04	14.88	14.73	0.096
C18:1	33.42[a]	32.66[ab]	32.36[b]	0.261
C18:2	16.46	16.16	16.03	0.123
C18:3	0.51[b]	0.49[b]	0.75[a]	0.029
C20:4	6.88[b]	8.06[a]	8.52[a]	0.138
C22:6	2.00[b]	2.70[a]	2.99[a]	0.081
Saturated	38.16[a]	37.81[ab]	37.04[b]	0.236
Monounsaturated	35.98	34.77	34.66	0.368
Polyunsaturated	25.86[b]	27.42[a]	28.29[a]	0.269
Unsaturated : saturated	1.62[b]	1.64[ab]	1.70[a]	0.014

[1]Standard errors of mean (n=9).
[a,b]Different letters within the same row differ significantly ($p<0.05$).

Table 5. Proximate composition (%) and water holding capacity (%) in the breast meat from broiler fed mixture of gallic and linoleic acid after 36 days

Treatment	Moisture	Crude protein	Crude fat	Crude ash	Water holding capacity
Control	73.23	20.80	1.08[y]	1.04	51.66[y]
0.5% MGL	73.00	20.67	1.30[y]	0.82	54.74[y]
1.0% MGL	73.36	20.77	2.44[x]	0.82	60.40[x]
SEM[1]	0.213	0.085	0.191	0.087	1.194

[1]Standard errors of mean (n=9).
[x,y]Different letters within the same column differ significantly ($p<0.05$).