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Research article

Effect of turmeric powder supplementation on physical and chemical egg quality, antioxidant activity, and yolk fatty acid profile

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Abstract

Turmeric powder (TP) supplementation is a widely used strategy to enhance hens' productivity and potentially mitigate egg fat oxidation by increasing antioxidant activity. Therefore, this study aimed to investigate the impact of TP supplementation on egg quality, antioxidant activity, and yolk fatty acid profile. A total of 100 56-week-old Novogen strain laying hens, matched in body weight (1799 ± 124 g), were divided into four treatments (with 5 replications per treatment, and 5 hens per replication). The treatments included control (CON), basal diet + 0.25% TP (T25), basal diet + 0.5% TP (T50), and basal diet + 0.75% TP (T75) administered for 8 weeks. Egg samples were collected on days 28 and 56 for analysis of physical and chemical quality, antioxidant activity, and fatty acid profile. On day 28, TP supplementation at 0.25% increased egg weight and specific gravity ($P < 0.05$), while on day 56, the supplementation at 0.50% and 0.75% reduced yolk cholesterol content ($P < 0.05$). TP supplementation at 0.5% and 0.75% also increased antioxidant activity and decreased malondialdehyde (MDA) content ($P < 0.05$). In conclusion, supplemention of TP enhanced egg quality, and antioxidant activity and also decreased cholesterol.

Keywords: Antioxidant activity, Egg quality, Fatty acid profile, Laying hens, Turmeric powder

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INTRODUCTION

Following the use of antibiotic restrictions, several alternative ingredients were developed to support sustainable poultry farming practices (Haque et al., 2020; Reznichenko et al., 2021; Abd El-Hack et al., 2022; Gómez et al., 2022; Hanim et al., 2023; Ruvalcaba- Trung Thong et al., 2023; Hanif et al., 2024). Among these alternatives, herbal-based feed additives have been extensively investigated for the potential to enhance productivity and health (Arif et al., 2022; Souza and Selvam, 2022; Rubens et al., 2023; Michael, 2024). In this context, turmeric powder (TP) served as an alternative to antibiotics, offering potential benefits in productivity, immunity, liver function, and reproductive hormone regulation in laying hens (Saraswati et al., 2014; Mirbod et al., 2017; Fawaz et al., 2022; Mosayyeb Zadeh et al., 2022; Hanif et al., 2023). Additionally, TP has been shown to improve egg quality (Kujero et al., 2021; Hanif et al., 2023).

Turmeric is well-known for the potent antioxidant properties, mainly attributed to bioactive compounds such as curcumin. This curcumin is a polyphenol present in turmeric and has been extensively studied for antioxidant effects. Furthermore, numerous reviews have shown robust antioxidant activity, such as combating oxidative stress and protecting cells from damage caused by free radicals (Hewlings and Kalman, 2017; Abd El-Hack et al., 2021). Studies have also shown antioxidant potential in various applications. Turmeric extract enriched with curcumin has been shown to enhance antioxidant activity, making it a valuable ingredient in food products (Partio et al., 2023). In addition, turmeric has been introduced into dairy products and soybean oil to increase antioxidant content and quality (Britto et al., 2020; Tinello et al., 2020).

Supplementing layer diets with TP has the potential to enhance egg quality and antioxidant activity. According to Abou-Elkhair et al. (2018), certain phytogenic feed additives supplementation reduced malondialdehyde (MDA) levels in egg yolk. Deniz et al. (2022) also showed that rosemary essential oil supplementation decreased MDA content in egg yolk. Based on the observation of Evans and Omaye (2017), MDA is a by-product of the oxidation of unsaturated fatty acids. There are limited studies on the effect of TP supplementation on antioxidant activity in layer egg. Therefore, this study aimed to investigate the effects of TP supplementation on egg quality, antioxidant activity, and yolk fatty acid profile.

MATERIALS AND METHODS

Ethical clearance and study location

Prior authorization for this experiment was obtained from the Ethics Commission of the Faculty of Veterinary Medicine, Universitas Gadjah Mada, Indonesia, under the number 98/EC-FKH/Eks/2023. The raising laying hens was conducted in the research barn of PT Agromix Lestari Group, situated at Banaran Kidul, Banguncipto, Sentolo, Kulon Progo, Special Region of Yogyakarta, Indonesia and egg quality assessment was conducted at the Faculty of Animal Science, Universitas Gadjah Mada, Sleman, Special Region of Yogyakarta, Indonesia..

Hens, housing, and diets

A total of 100 56-week-old laying hens of Novogen strain, with similar body weights (1799 ± 124 g), were allocated to four treatments, each comprising 5 replicates with 5 hens per replicate, in a completely randomized design. The experiment lasted for 8 weeks, extending until the hens reached 64 weeks of age. An experimental diet was formulated based on the maize–soybean meal, following the Novogen nutritional recommendation (Table 1). The experimental groups were created by supplementing TP as, CON (control, basal diet), T25 (basal diet + 0.25%

TP), T50 (basal diet + 0.5% TP), and T75 (basal diet + 0.75% TP). TP used for the analysis was provided by PT Seribu Cita Bayanaka, Banten, Indonesia. Hens were fed 120 grams per day with a proportion of 48 grams in the morning (07:00) and 72 grams in the afternoon (15:00). These hens were housed in battery cages measuring 22 cm long, 35 cm wide, and 30 cm high, equipped with trough feeders and nipple drinkers. The lighting regimen consisted of 16 hours of continuous light per day, from 06:00 to 22:00.

Table 1 Ingredients and nutrient composition of experimental laying hen diets

Ingredients	Composition (%)
Maize	56.65
Rice bran	3.60
Soybean Meal	24.80
Limestone	10.75
Crude Palm Oil	1.50
Di-Calcium Phosphate	1.00
DL-Methionine	0.25
L-Lysine	0.10
Common Salt	0.25
Mineral Premix[1]	0.20
Vitamin Premix[2]	0.05
Choline Chloride	0.10
Turmeric powder[3]	0.00
Filler[4]	0.75
Total	100
Calculated	
Dry Matter (%)	89.8
Metabolizable Energy (kcal/kg)	2812
Crude Protein (%)	18.3
Extract Eter (%)	3.6
Crude Fiber (%)	2.6
Calcium (%)	4.4
Total Phosphor (%)	0.5
Methionine (%)	0.53
Lysine (%)	0.95

Note: [1]contained per kg: sodium 29.3 g; calcium 261.5 g; iron 17.7 g; potassium 2.4 g; magnesium 2.5 g; phosphor 6.9 g; sulphur 783.6 µg; manganese 900 µg; copper 186.6 µg; zinc 361.6 µg; selenium 0.61 µg; cobalt 5.3 µg. [2]contained per kg: vitamin D3 4.000.000 IU; vitamin A 20.000.000 IU; carnitine 20.000 mg; vitamin K3 7.000 mg; vitamin B2 12.000 mg; vitamin B12 60 mg; vitamin E 15.000 mg; vitamin B6 8.000 mg; Ca-d-panthothenate 20.000 mg; niacin 40.000 mg; and folic acid 500 mg. [3] Each treatment group has a different turmeric powder composition, CON: 0.00%; T25: 0.25%; T50: 0.50%; T75: 0.75%. [4] Each treatment group contains different filler (rice bran) content in the feed, depending on the turmeric powder content, CON: 0.75% filler; T25: 0.50% filler; T50: 0.25% filler and T75: 0.00% filler.

Physical egg quality assessment

A total of two eggs per replication group were collected on days 28 and 56 to assess the physical quality of the eggs. The egg, eggshell, albumen, and egg yolk were weighed using a High-Precision Digital Weight with a capacity of 500 g/0.01 g (PT. Arta Joil Tappa, Indonesia). The egg shape index was calculated by dividing the vertical diameter by the horizontal diameter of the egg using the Egg Form Coefficient Measurement Instrument (FHK, Fujihira Industry Co., Ltd., Japan). Egg volume was determined by using a 1000 ml glass measuring cylinder (PT. Iwaki Glass Indonesia, Indonesia). Egg-specific gravity was established by dividing the egg's weight by its volume. Eggshell straightness was measured using a pressure gauge (FHK, Fujihira Industry Co., Ltd., Japan). The height of the albumen was measured as the distance between the metal plate and electrode placed on top of the thickest part of the egg while the yolk height was measured after the yolk was separated from the albumen. Using a micrometer (FHK, Fujihira Industry Co., Ltd.,

Japan). The albumen index and yolk index are measured by dividing the height by the diameter. The Haugh units were assessed following the equation by (Haugh, 1937) (HU = 100 log (albumen height (mm) + 7.57) - 1.7 * (egg weight (g)) ^ 0.37)). The yolk color was assessed using the Roche yolk color fan scores (RYCF; F. Hoffman-La Roche, Switzerland), which assigned values based on 15 different color samples ranging from 1 (the lightest) to 15 (the darkest).

Chemical egg quality assay

On days 28 and 56, one egg per replicate was collected for chemical quality analysis. Eggs were broken and the yolk was collected for analysis. An examination was undertaken to estimate the moisture, organic matter, crude fat, and ash content of the egg yolk using proximate analysis, following the AOAC guidelines by Hortwitz and Latimer (2005). Yolk cholesterol levels were analysed using the Lieberman-Burchard method, 1 gram of yolk sample was added to 10 ml of acetone and ethanol solvent (1:1) and vortexed for 1 minute. The sample and solvent mixture were immersed in boiling water until the mixture boiled. Afterward, the sample mixture was cooled and centrifuged at 3000 rpm for 10 minutes (Eppendorf 5804R, Germany). The supernatant was taken and immersed in boiling water until the solvent evaporated. The residual fat in the tube was diluted with chloroform solvent to 30 times dilution. 1 ml of the mixture was added to 1 ml of a mixture of sulphuric acid and anhydrous acetic acid (1:30), vortexed for 1 minute, and left for 10 minutes. The absorbance of sample and blank was measured at 515 nm with a UV-Vis spectrophotometer (Thermo Fisher Genesys 10s UV-Vis, USA) at a wavelength of 680 nm.

Antioxidant activity assay

The 2,2-diphenyl-1-picrylhydrazyl (DPPH) method was used to analyze antioxidant activity of egg yolk samples collected on days 28 and 56 (one egg per replicate). For the extraction, 0.2 grams of egg yolk was added to 5 ml of methanol solvent and vortexed for 1 minute. The mixture was then centrifuged at 3000 rpm for 10 minutes (Eppendorf 5804R, Germany). After centrifugation, 0.2 ml of supernatant was added to 2.8 ml of 0.1 mM DPPH reagent and kept in a dark room for 1 hour. The absorbance was then measured at 515 nm with UV-Vis spectrophotometer (Thermo Fisher Genesys 10s UV-Vis, USA) against a blank (methanol) at a wavelength of 515 nm. Scavenging activity was expressed as the percentage inhibition (I%) calculated using the equation, I% = (Abs. blank – Abs. sample/Abs. blank) × 100.

Malondialdehyde assay

Five grams of egg yolk samples collected on days 28 and 56 were mixed with 10 ml of 10% trichloroacetic acid and thoroughly homogenized using a homogenizer (IKA T25, Germany). The mixture was then centrifuged at 3000 rpm for 10 minutes (Eppendorf 5804R, Germany) and filtered using Whatman paper size 1. Subsequently, 2.5 ml of the supernatant was mixed with 0.380% TBA reagent in HCl (2.25 ml /100 ml TBA). The samples and blanks (distilled water) were incubated in a water bath at 90°C for 30 minutes. After cooling, the absorbance was measured using a spectrophotometer (Thermo Fisher Genesys 10s UV-Vis, USA) at a wavelength of 530 nm.

Fatty acid profile assay

One egg per replicate was collected on days 28 and 56 for yolk fatty acid profile analysis. A test tube containing a 5 g sample of yolk was filled with 10 ml of 37% HCl. For three hours, the mixture was heated to 80ºC. The mixture was heated at 80ºC for 3 hours. Following the cooling process, the mixed solution underwent extraction using a blend of 25 ml diethyl ether and petroleum ether in a 1:1 proportion. The mixture was agitated using a vortex, and the top layer (oil) was

removed by evaporating it with a water bath in the presence of N2 gas0.5 mL of oil was added to a tiny test tube and sealed firmly. Afterward, 1.5 mL of a solution containing sodium in methanol was added. The mixed solution was heated at 60°C for 10 minutes with shaking. After cooling, 2 mL of Boron trifluoride methanoate was added and heated at 60°C for 10 minutes and cooled again. The mixture was extracted with 1 mL of Heptana and 1 mL of saturated NaCl. The top layer formed was put into a GC vial as much as 1 µL.

The fatty acid profile was measured using a gas chromatograph (Agilent 7890B autosampler Series, Agilent Technologies, Palo Alto, CA, USA) and an HP-88 capillary column (100 m×0.3 µm×0.2 µm, Agilent Technologies, Palo Alto, CA, USA). The technique employed was gas chromatography-mass spectrometry (GC-MS). The test settings for the temperature program were the following: The program started with a temperature of 100°C for 5 minutes, then climbed to 240°C at a pace of 4°C per minute. The carrier gas used in this experiment was ultrahigh purity helium, which flowed at a rate of 30 mL/min. The temperatures of the injector, interface, and ion source were recorded at 280°C, 260°C, and 240°C, respectively.

Statistical analysis

A two-way ANOVA (4 TP levels × 2 duration treatments) in a completely randomized design was used to analyze the physical and chemical quality of egg and antioxidant activity, while fatty acid profile was analyzed using one-way ANOVA. Data analysis was conducted using the IBM SPSS Statistics 26 statistical program (SPSS Inc., USA). Furthermore, the Duncan Multiple Range Test (DMRT) was used to identify significant differences among treatments ($P < 0.05$).

RESULTS

Physical egg quality

The effect of TP supplementation on physical egg quality was presented in Table 2. Overall, TP inclusion did not affect average egg weight ($P > 0.05$). Intrestingly, TP supplementation at 0.25% increased egg weight and specific gravity on day 28 ($P < 0.05$). Feeding TP increased the average egg shell gravity ($P < 0.05$). Feeding TP increased the average egg shell gravity. In addition, eggs produced on day 56 were smaller than those on day 28. Furthermore, the supplementation at 0.75% showed higher egg-specific gravity compared to other treatments on day 56 ($P < 0.05$). Additionally, TP-supplemented eggs showed lower ESR compared to the control group ($P < 0.05$). However, TP supplementation decreased eggshell ratio on day 56 ($P < 0.05$). Moreover, TP supplementation didn't affect average yolk index. In contrast, the supplementation at 0.50% and 0.75% increased yolk index on day 56 ($P < 0.05$). Egg yolk produced from laying hens fed dietary TP showed higher color than the control group.

Chemical egg quality

All eggs had the same yolk moisture content across treatment groups, as shown in Table 3. There were no significant differences ($P > 0.05$) observed among dietary treatments regarding the organic matter, crude fat, and ash content. Overall, TP supplementation decreased egg yolk cholesterol content ($P < 0.05$). Interestingly, TP supplementation did not affect yolk cholesterol content on day 28, but on day 56, the supplementation at 0.50% and 0.75% reduced yolk cholesterol content ($P < 0.05$).

Table 2 Effects of turmeric powder supplementation on physical quality of eggs

Item	EW	ESI	ESG	ESS	ESR	EST	AI	AR	YI	YR	YAR	YC	HU
Turmeric level (%)													
0.00	60.2	76.7	1.09[b]	0.38	11.1[a]	0.37	0.10	63.1	0.36	25.7	41.0	7.4[b]	90.7
0.25	62.4	74.3	1.15[a]	0.33	10.4[b]	0.36	0.10	63.9	0.36	25.7	40.2	7.8[a]	89.8
0.50	60.8	75.4	1.14[a]	0.34	10.4[b]	0.36	0.10	63.3	0.34	26.2	41.6	8.0[a]	91.3
0.75	61.5	75.0	1.17[a]	0.35	10.2[b]	0.36	0.10	63.7	0.35	26.1	41.0	8.1[a]	92.0
Treatment duration (days)													
28	60.7	75.3	1.15[a]	0.34	10.5	0.36	0.10	63.5	0.35	25.9	41.0	7.7	90.6
56	61.8	75.4	1.12[b]	0.35	10.5	0.37	0.10	63.5	0.35	25.9	40.9	7.9	91.3
SEM													
ANOVA							*P*-value						
Turmeric level	0.34	0.14	<0.01	0.21	0.03	0.69	0.74	0.64	0.67	0.72	0.79	0.00	0.72
Duration treatment	0.21	0.95	0.04	0.54	0.97	0.08	0.27	1.00	0.34	0.99	0.99	0.07	0.61
Turmeric level × Duration treatment	0.25	0.07	0.17	0.58	0.49	0.15	0.60	0.87	0.08	0.87	0.91	0.47	0.74

Note: EW: egg weight (g), ESI: egg shape index, ESG: egg specific gravity (g/ml), ESS: eggshell strength (mPa), ESR: eggshell ratio (%), EST: eggshell thickness (mm), AI: albumen index, AR: albumen ratio (%), YI: yolk index, YR: yolk ratio (%), YAR: yolk albumen ratio (%), YC: yolk color, HU: haugh unit.
[a, b, c] *Means in the same coloum without common letters are different at P<0.05*

Table 3 Effects of turmeric powder supplementation on chemical quality of egg yolk

Item	Moisture (%)	Organic matter (%)	Fat (%)	Cholesterol (mg/g)	Ash (%)
Turmeric level (%)					
0.00	50.4	47.2	29.9	11.4[a]	2.48
0.25	50.4	47.3	29.7	11.0[a]	2.29
0.50	50.3	47.3	29.9	10.3[b]	2.40
0.75	50.4	47.3	29.9	10.2[b]	2.33
Treatment duration (days)					
28	50.2	47.4	30.1	10.6	2.36
56	50.5	47.1	29.7	10.8	2.39
SEM	0.10	0.75	0.67	0.77	0.20
ANOVA			*P*-value		
Turmeric level	1.00	0.98	0.95	<0.01	0.21
Duration treatment	0.14	0.17	0.08	0.31	0.61
Turmeric level × Duration treatment	0.67	0.62	0.78	0.82	0.36

Note: [a, b, c] Means in the same coloum without common letters are different at *P*<0.05

Antioxidant activity

TP supplementation led to increased antioxidant activity in egg yolk, as shown in Table 4. Supplementation at 0.5% and 0.75% increased the percentage of DPPH inhibition in egg yolk (P <0.001). Therefore, TP supplementation decreased MDA content (P <0.001). Dietary TP supplementation at 0.5% and 0.75% reduced MDA levels in egg yolk on both days 28 and 56 (P <0.05). Egg yolk from day 56 showed lower MDA levels than those from day 28 (P < 0.05). However, there were no interactions between TP supplementation level and treatment duration (P > 0.05).

Table 4 Effects of turmeric powder supplementation on antioxidant activity and lipid oxidation of egg yolk

Item	Antioxidant activity (Inhibition %)	Malondialdehyde (nmol/g)
Turmeric level (%)		
0.00	2.31^{b}	42.6^{a}
0.25	2.37^{b}	41.8^{b}
0.50	2.45^{a}	41.3^{bc}
0.75	2.50^{a}	40.9^{c}
Treatment duration (days)		
28	2.41	42.0^{a}
56	2.40	41.4^{b}
SEM	0.02	0.16
ANOVA	*P*-value	
Turmeric level	<0.001	<0.001
Duration treatment	0.57	0.02
Turmeric level × Duration treatment	0.80	0.67

Note: $^{a, b, c}$ Means in the same collum without common letters are different at P<0.05

Fatty acid profile

The effect of TP supplementation on fatty acid profile was presented in Table 5. Feeding TP at 0.75% increased arachidic acid and heneicosanoic acid (P <0.05). However, dietary inclusion of TP did not affect saturated fatty acid, monounsaturated fatty acid, and polyunsaturated fatty acid.

Table 5 Effects of turmeric powder supplementation on fatty acid profile of egg yolk at 56 days of treatment

Variables	CON	T25	T50	T75	SEM	p-value
Butyric acid (%)	0.20	0.14	0.14	0.18	0.02	0.459
Myristic aid (%)	0.27	0.27	0.33	0.36	0.02	0.106
Heptadecanoic acid (%)	2.9	2.7	3.4	3.26	0.23	0.766
Arachidic acid (%)	13.4^{b}	12.4^{b}	13.8^{b}	15.6^{a}	0.37	0.008
Heneicosanoic acid (%)	0.15^{bc}	0.11^{c}	0.20^{b}	0.27^{a}	0.02	0.005
Tricosanoic acid (%)	0.29	0.22	0.14	0.13	0.04	0.443
Palmitoleic acid (%)	28.1	27.9	26.8	27.12	0.25	0.271
Linolaidic acid (%)	9.8	9.3	7.1	7.4	0.69	0.500
Linoleic acid (%)	36.2	39.6	43.2	41.8	1.67	0.548
cis-11-eicosenoic acid (%)	0.13	0.11	0.12	0.11	0.01	0.298
cis-5,8,11,14-eicosatetraenoic acid (%)	4.8	4.4	3.4	2.9	0.36	0.241
cis-5,8,11,14,17-eicosapentaenoic acid (%)	2.4	2.2	2.2	2.2	0.19	0.975
Nervonic acid (%)	0.73	0.61	0.65	0.62	0.13	0.992
cis-4,7,10,13,16,19-docosahexaenoic acid (%)	1.4	1.2	1.0	0.90	0.10	0.271
Saturated Fatty Acid (%)	17.1	14.4	15.1	16.00	1.30	0.927
Monounsaturated Fatty Acid (%)	28.9	28.3	27.7	27.8	0.25	0.342
Polyunsaturated Fatty Acid (%)	54.0	57.2	57.3	56.2	1.40	0.866

Note: CON: control (basal diet), T25: basal diet + 0.25% turmeric powder, T50: basal diet + 0.5% turmeric powder, T75: basal diet + 0.75% turmeric powder

$^{a, b, c}$ Means in the same row without common letters are different at P<0.05

DISCUSSION

This study showed that 0.25% TP supplementation led to an increase in egg weight on day 28. The results were in line with the observation of Fawaz et al. (2022), who indicated that feeding TP at doses of 2.5, 5, and 7.5 g/kg enhanced hens' day egg production, egg weight, egg mass, and feed conversion ratio (FCR). Samia et al. (2018) found that including TP at 6 g/kg increased egg weight and production while reducing FCR compared to diet with 4 g/kg of TP and control

treatments. Mirbod et al. (2017) reported an increase in egg mass and an improvement in FCR with TP supplementation at concentrations of 2.0 and 6.0 g/kg diet. The increase in egg weight might be attributed to the enhanced digestibility of nutrients from the feed into the egg (Fawaz et al., 2022). Moreover, the rise in egg-specific gravity observed in this study could be attributed to the increase in egg weight. Egg-specific gravity was calculated by dividing egg weight by egg volume (Malfatti et al., 2021), and the inclusion of TP improved yolk color. Curcumin, a bioactive compound found in turmeric, has been examined for its potential to enhance yolk color (Gumus et al., 2018).

TP supplementation resulted in reduced cholesterol levels in yolk. Curcumin restricted cholesterol intake in diet and acted as antiatherogenic substance, lowering both blood cholesterol levels and the amount of cholesterol transported to yolk (Suwarta and Suryani, 2019). Another study indicated that TP could reduce cholesterol by increasing cholesterol-7-αhydrolase activity or inhibiting HMG Co-A reductase activity (Malekizadeh et al., 2012). The curcumin compound of turmeric suppressed HMG-CoA activity through transcriptional inhibition and stimulated the conversion of cholesterol into bile acids, facilitating cholesterol removal from the body and increasing cholesterol excretion (Srinivasan and Sambaiah, 1991; Shin et al., 2011; Qinna et al., 2012).

TP supplementation increased antioxidant activity and reduced MDA content in egg yolk. The enhancement of egg yolk antioxidant activity was attributed to the presence of active compounds in turmeric, which functioned as antioxidant (Partio et al., 2023). Curcuminoid compounds found in turmeric, such as curcumin, demethoxycurcumin, and bisdemethoxycurcumin, play an important role as antioxidant (Daneshyar, 2012). The investigation conducted by Novilla et al. (2014) provided insight into the mechanism of phenolic compounds as antioxidant activity in scavenging free radicals through hydrogen and electron donation. Moreover, free radical oxidants engaged in hydrogen abstraction and electron transfer processes. Curcumin can undergo oxidation through electron transfer and hydrogen abstraction at all three active sides (Priyadarsini, 2014). In addition, TP increased antioxidant activity and decreased MDA levels by enhancing antioxidant enzymes (Gowda et al., 2008).

The measurement of MDA content has long served as a lipid peroxidation marker (Morales and Munné-Bosch, 2019). However, egg yolk MDA levels decreased with the inclusion of herbal feed additives in the diet. Both black cumin seed and hot red pepper supplementation at 0.5% reduced egg yolk MDA levels (Abou-Elkhair et al., 2018). Deniz et al. (2022) reported that rosemary essential oil supplementation decreased MDA levels in egg yolk. Additionally, Nunes et al. (2019) investigated the effect of dietary inclusion of dehydrated Bocaiuva pulp on quail egg and observed a linear decrease in MDA content in egg yolk with increased pulp levels. Wang et al. (2022) also found that pomegranate seed oil supplementation significantly diminished MDA content in egg yolk, indicating antioxidant effects from dietary addition. Interestingly, this study showed significant differences in the reduction of egg yolk MDA levels with TP supplementation level and treatment duration. However, there was no interaction between TP level and treatment duration. These results corresponded with Simitzis et al. (2018), who reported that quercetin supplementation reduced MDA levels based on supplementation level and treatment duration without any interaction between quercetin level and treatment duration. A meta-analysis by Jakubczyk et al. (2020) showed the effectiveness of curcumin in reducing MDA levels and the potential to increase antioxidant levels. Therefore, the reduction in oxidative stress was dependent on treatment duration and the dose of curcumin administered.

TP supplementation did not affect fatty acid profile of yolk. Several results might arise when analyzing fatty acid profile under different storage treatments. The addition of phytogenic feed additives showed positive results on fatty acid composition of egg stored under specific conditions. Egg storage temperature

played a crucial role in shaping fatty acid composition of yolk from hens-fed supplements such as lycopene or astaxanthin (Honchar et al., 2022). Additionally, Mierliță (2019) reported that the oxidative stability of egg significantly depended on the levels and types of polyunsaturated fatty acids (PUFAs), α-tocopherol levels, and the duration of egg storage.

CONCLUSIONS

In conclusion, TP supplementation enhanced egg weight, eggshell gravity, yolk color, antioxidant activity, and decreased cholesterol and MDA levels. The recommended dosage for TP supplementation in laying hens' diet was 0.50%. Therefore, future study was needed to explore the effect of TP supplementation and the effect on fatty acid profile of egg stored for different durations.

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AUTHOR CONTRIBUTIONS

Muhammad Fathin Hanif: Collected and analyzed the data and drafted the manuscript. Ali Agus: Designed study concepts and modified the manuscript. Bambang Ariyadi: Supervised the experiment and modified the manuscript. Muhlisin: Supervised the experiment and modified the manuscript. Sesotya Raka Pambuka: Analyzed the data and modified the manuscript All authors read and agreed on the final manuscript.

CONFLICT OF INTEREST

All authors declare that they have no competing interests.

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