

# Utilization of *Solanum nigrum* L. (Black Nightshade) Fruit as a Phytobiotic in Laying Quail

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## Abstract

Black nightshade (*Solanum nigrum* L.) is known to contain bioactive compounds with antioxidant properties. This study aimed to evaluate the effects of black nightshade fruit extract as a phytobiotic on the performance and egg quality of laying quails (*Coturnix-coturnix japonica*). A total of 160 female quails, aged six weeks, were randomly assigned to four treatment groups with four replications, each containing ten birds. The treatments included a control group (T0) and three experimental groups receiving drinking water supplemented with 2 mL/L (T1), 4 mL/L (T2), and 6 mL/L (T3) of black nightshade extract. The parameters observed included feed intake, quail day production, egg production, egg weight, feed conversion ratio (FCR), physical egg quality (egg index, percentage of albumen, yolk and eggshell weight, eggshell thickness, yolk color score, haugh unit, albumen and yolk index) and malondialdehyde (MDA) levels in egg yolk. The cold-press extraction method offers advantages in terms of cost-effectiveness and environmental sustainability. This technique avoids the use of solvents and heat, which helps preserve thermosensitive compounds. However, a notable limitation is that the extracted compounds remain in their complex, non-degraded forms, which may limit the efficiency of isolating certain bioactive constituents—especially those such as phenolics, flavonoids, and alkaloids that typically require solvents for effective extraction. Phytochemical screening conducted in this study revealed low concentrations of several compounds, and notably the extract of *Solanum nigrum* fruit obtained using a cold-press juicer method showed no presence of triterpenoids or steroids. The results showed that black nightshade extract supplementation did not significantly affect quail performance ( $p>0,05$ ). However, significant improvements ( $p<0,05$ ) were observed in physical egg quality parameters, particularly in albumen index, yolk color, and haugh unit at higher doses (T2 and T3). The MDA content in the yolk significantly decreased with increased extract concentration.

**Keyword:** cold-press extraction, egg quality, antioxidant

## 1. Introduction

*Coturnix coturnix japonica* is one of the poultry species widely raised in Indonesia. It has been domesticated and utilized by local communities as a source of both eggs and meat. Quail possess a relatively short reproductive cycle, requiring only about 45 days to mature. Annual egg production per bird can reach approximately 270–300 eggs. The average weight of a quail egg ranges from 11 to 13 grams, which is relatively small compared to chicken eggs and those of other poultry species. Despite their size, quail eggs are rich in nutrients and are considered an excellent dietary source of protein for humans (1).

One promising approach to enhance quail productivity is through the use of local plants as phytobiotics. Phytobiotics are plant-derived bioactive compounds that are environmentally friendly, and capable of boosting both immunity and production performance in poultry (2). Phytobiotics are considered a viable alternative to antibiotic growth promoters (AGPs). The antioxidant components found in phytobiotics help protect body cells from damage caused by free radicals and oxidative stress, and they may also stimulate the expression of endogenous antioxidant enzymes (3). Plants that contain high levels of bioactive compounds have great potential for use as phytobiotics in animal feed.

Several studies have explored the application of phytobiotics in poultry. Krauze (4) reported that supplementing cinnamon oil at 0.25 mL/L in the drinking water of chickens improved performance, metabolism, immune response, and antioxidant activity. Another study demonstrated that administering 2 mL/L of *Bifidobacterium sp.* and *G. ulmifolia* leaf extracts in quail drinking water increased egg mass, Haugh unit, and yolk color intensity (5).

*Solanum nigrum*, commonly known as black nightshade or "leunca" in Indonesia, is a local plant known to contain various bioactive compounds with strong antioxidant activity (6). Given these properties, the use of *Solanum nigrum* fruit extract as a phytobiotic presents significant potential. Therefore, further studies are necessary to evaluate the effects of *S. nigrum* fruit extract as a phytobiotic in laying quail.

## 2. Material and Methods

### 2.1. Experimental Design

The experiment consisted of four treatment groups with four replicates each. Quail rearing was conducted at Arkan Quail Farm, located in Bogor, Indonesia. Evaluation of egg quality was performed at the Poultry Nutrition Laboratory, Department of Nutrition and Feed Technology, Faculty of Animal Science, IPB University. A total of 160 female Japanese quails (*Coturnix coturnix japonica*) were used, reared from 6 to 16 weeks of age. The birds were provided with a commercial feed (New Hope P100, PT New Hope Farm Indonesia), and both feed and drinking water were supplied *ad libitum*. Feeding, watering, and cage cleaning were performed daily. Black nightshade (*Solanum nigrum*) fruit extract was administered every day. Eggs were collected and weighed daily for data recording. Physical egg quality assessment was conducted every two weeks, with three eggs per replication selected using a simple random sampling method. The parameters observed included productivity performance: feed intake (g/bird/day), quail day production (QDP, %), egg weight (g), egg mass (g/bird/day), feed conversion ratio (FCR), and water intake (ml/bird/day). Egg physical quality was evaluated, including yolk, albumen, and shell weight and percentage, egg index, yolk index, albumen index, shell thickness, yolk color, and Haugh unit. The study also measured malondialdehyde (MDA) content (mg/kg) of the eggs.

### 2.2. Black Nightshade Fruit Extraction

The extraction method was a modified procedure based on the technique described by Puspitaningrum (7). The fruits were thoroughly washed and drained prior to extraction. The cleaned fruits were processed using a cold-press juicer to obtain the crude extract, which was subsequently filtered and pasteurized for 10 minutes. The resulting black nightshade fruit extract was stored in refrigerator at 4°C until further use.

## 2.3. Statistical Analyses

Data were analyzed using analysis of variance (ANOVA) with IBM SPSS software (version 25). When significant differences were found ( $p < 0.05$ ), further comparisons between treatment means were conducted using Tukey's post hoc test. The malondialdehyde (MDA) levels in egg yolks were analyzed descriptively. The study included four treatment groups: T0 = just drinking water (control), T1 = 1 L drinking water + 2 mL black nightshade fruit extract, T2 = 1 L drinking water + 4 mL black nightshade fruit extract, and T3 = 1 L drinking water + 6 mL black nightshade fruit extract.

## 3. Results

### 3.1. Phytochemical on Fruit Extraction

The results of the phytochemical screening of black nightshade (*Solanum nigrum*) fruit extract are presented in Table 1. The extract was found to contain alkaloids, saponins, tannins, phenolics, flavonoids, and glycosides. However, triterpenoids and steroids were not detected in the extract.

### 3.2. Production Performance of Laying Quails Aged 6–16 Weeks

The production performance of laying quails aged 6 to 16 weeks supplemented with black nightshade (*Solanum nigrum*) fruit extract in drinking water is summarized in Table 3. The supplementation did not result in significant changes in feed intake, egg production, egg weight, egg mass, feed conversion ratio, or water intake ( $P > 0.05$ ).

### 3.3. Egg Quality of Laying Quails Aged 6–16 Weeks

Egg quality parameters of laying quails from 6 to 16 weeks of age are shown in Table 4. Significant improvements were observed in albumen index, yolk color, and Haugh unit ( $P < 0.05$ ). Among the groups, T3 showed the highest values in these three parameters. No significant differences were found in egg index, yolk index, eggshell percentage, or shell thickness ( $P > 0.05$ ).

### 3.4. Malondialdehyde (MDA) Content in Eggs

The levels of malondialdehyde (MDA) and the percentage reduction across treatment groups are presented in Table 4. The control group (T0) had an MDA level of 1.776 mg/kg. Treatment groups T2 and T3 showed reductions of 23.20%, and 28.38%, respectively, with the T3 group exhibiting the greatest decrease in MDA concentration.

Table 1. Phytochemicals Screening of Black Nightshade Fruit Extraction

Phytochemical	Result <sup>a</sup>
Alkaloids	+
Saponins	+
Tanins	+
Phenolics	+
Flavonoids	+
Glycosides	+
Triterpenoids	-
Steroids	-

<sup>a</sup>Results of laboratory analysis of BSIP TROA, Bogor (2024)

Table 2. Production performance of laying quail (6-16 week)

Variable	Treatment			
	T0	T1	T2	T3
Feed intake (g/bird/day)	23,89 ± 1,16	22,63 ± 1,03	23,55 ± 0,90	23,83 ± 0,67
Quail Day Production (QDP) (%)	83,42 ± 11,95	78,83 ± 9,78	80,45 ± 14,22	84,87 ± 12,63
Egg weight (g/egg)	11,20 ± 0,23	10,85 ± 0,32	11,07 ± 0,22	11,06 ± 0,32
Egg mass (g/bird/day)	9,24 ± 1,44	8,52 ± 1,09	8,93 ± 1,71	9,40 ± 1,50
Feed Conversion Ratio (FCR)	2,63 ± 0,58	2,73 ± 0,67	2,84 ± 1,19	2,63 ± 0,70
Water intake (ml/bird/day)	67,68 ± 11,25	64,28 ± 3,77	64,87 ± 7,03	61,62 ± 9,99

Different letters on the same row indicates statistical difference ( $P \leq 0.05$ ) among treatments.

Table 3. Egg quality of laying quail (6-16 week)

Variable	Treatment			
	T0	T1	T2	T3
Albumen weight (g)	7,76 ± 0,63	7,63 ± 0,60	7,89 ± 0,50	7,92 ± 0,57
Yolk Weight (g)	3,70 ± 0,43	3,64 ± 0,53	3,72 ± 0,35	3,67 ± 0,39
Shell weight (g)	1,53 ± 0,20	1,49 ± 0,15	1,55 ± 0,13	1,55 ± 0,15
Albumen percentage (%)	67,75 ± 2,27	67,75 ± 3,40	67,98 ± 2,05	68,37 ± 2,29
Yolk percentage (%)	32,35 ± 2,27	32,25 ± 3,40	32,02 ± 2,05	31,63 ± 2,29
Shell percentage (%)	13,25 ± 1,17	13,20 ± 1,15	13,31 ± 0,89	13,34 ± 0,90
Egg index	78,16 ± 2,86	78,41 ± 2,83	78,16 ± 2,43	78,50 ± 1,95
Albumen index	11,11 ± 2,82 <sup>a</sup>	11,98 ± 1,74 <sup>ab</sup>	12,46 ± 2,27 <sup>b</sup>	12,29 ± 2,18 <sup>ab</sup>
Yolk index	44,35 ± 2,95	43,63 ± 3,07	43,17 ± 3,46	43,32 ± 2,31
Yolk colour	3,58 ± 0,69 <sup>a</sup>	4,41 ± 0,50 <sup>b</sup>	4,41 ± 0,89 <sup>b</sup>	4,50 ± 0,20 <sup>b</sup>
Haugh Unit (HU)	92,67 ± 1,34 <sup>a</sup>	94,16 ± 0,55 <sup>ab</sup>	95,16 ± 1,50 <sup>b</sup>	95,55 ± 1,03 <sup>b</sup>
Shell thickness (mm)	0,20 ± 0,00	0,19 ± 0,01	0,20 ± 0,00	0,20 ± 0,01

Different letters on the same row indicates statistical difference ( $P \leq 0.05$ ) among treatments.

Table 4. Egg yolk MDA levels of 16 week laying quail

Treatment	MDA (mg/kg)	MDA reduction (%)
T0	1,776	-
T1	1,890	-6,42
T2	1,364	23,20
T3	1,272	28,38

Results of laboratory analysis of Biotech Center IPB University (2024).

#### 4. Discussion

Phytochemical screening conducted in this study revealed low concentrations of several compounds, and notably the extract of *Solanum nigrum* fruit obtained using a cold-press juicer method showed no presence of triterpenoids or steroids. Hydroalcoholic extraction of *Solanum nigrum* fruit from India revealed the absence of alkaloids, while the ethyl acetate method yielded no alkaloids or terpenoids, though the flavonoid content reached 10.46% (8).

The discrepancies between the phytochemical findings of the present study and previous research may be attributed to differences fruit origin and the extraction methods. Different extraction techniques can yield distinct profiles of bioactive compounds. The cold-press extraction method offers advantages in terms of cost-effectiveness and environmental sustainability. This technique avoids the use of solvents and heat, which helps preserve thermosensitive compounds. However, a notable limitation is that the extracted compounds remain in their complex, non-degraded forms, which may limit the efficiency of isolating certain bioactive constituents—especially those such as phenolics, flavonoids, and alkaloids that typically require solvents for effective extraction (9).

The effects of *Solanum nigrum* fruit extract supplementation in quail drinking water on performance during the study are presented in Table 1. The average feed intake ranged from 22.63 to 23.89 g/bird/day. The extract treatments did not negatively affect feed consumption. Analysis of variance showed no significant differences ( $p>0.05$ ), likely due to the use of the same feed composition across all treatments, resulting in uniform energy intake.

Egg production did not differ significantly ( $p>0.05$ ) following black nightshade extract supplementation. Egg weight was also not significantly affected by treatment ( $p>0.05$ ). The average egg weight ranged from 10.85 to 11.20 g/egg. The absence of differences may be attributed to the uniform protein content in the feed provided to all groups. As reported by Ojediran (10), both egg production and egg weight are heavily influenced by protein intake and feed consumption levels. Egg mass was likewise unaffected ( $p>0.05$ ) by *S. nigrum* supplementation. The average egg mass ranged from 8.52 to 9.40 g/bird/day. Egg mass in the T3 group was slightly higher than that of T0. This increase may result from combined effects of protein intake, egg weight, and egg production.

Feed conversion ratio (FCR) showed no significant difference among treatments ( $p>0.05$ ). The FCR ranged from 2.63 to 2.84. A lower FCR indicates higher feed efficiency, meaning quails were more effective in converting feed into eggs (2). Water intake ranged from 61.62 to 67.68 mL/bird/day and was not significantly different between treatments ( $p>0.05$ ). Oxidative stress may influence water consumption, with higher intake possibly indicating elevated stress levels due to non-ideal temperatures (11). During this study, birds were maintained at temperatures ranging from 24 to 36°C, which may not represent optimal conditions.

The physical quality of eggs is shown in Table 2. The weight of albumen, yolk, and shell ranged from 7.63 to 7.92 g, 3.64 to 3.72 g, and 1.49 to 1.55 g, respectively. Egg white weight is generally proportional to total egg weight, which explains the relatively lower yolk weight in heavier eggs (12). There were no significant differences in component percentages ( $p>0.05$ ), with albumen, yolk, and shell percentages ranging from 67.75% to 68.37%, 31.63% to 32.35%, and 13.20% to 13.34%, respectively. Compared to Chimezie (12), the present study yielded higher values for albumen and shell, while yolk proportions were similar.

Haugh unit (HU) values ranged from 92.67 to 95.55 and showed a significant difference ( $p<0.05$ ). These values are consistent with Lovela (5) and higher than those in Chimezie (12). HU is influenced by albumen height and protein quality. Flavonoids present in the extract may contribute positively by enhancing nutrient absorption and improving protein metabolism via proteolytic enzyme activity (13)(14). Egg and yolk indices showed no significant differences ( $p>0.05$ ), while albumen index was significantly

affected ( $p < 0.05$ ). Indices observed were 78.16–78.50 (egg), 11.11–12.46 (albumen), and 43.17–44.35 (yolk). Albumen index correlates with HU and albumen height (15).

Yolk color was significantly affected by treatment ( $p < 0.05$ ), ranging from 3.58 to 4.50, although lower than those reported by Lovela (5). The color enhancement is likely due to carotenoid content in *S. nigrum*, particularly  $\beta$ -carotene (16) (17). Shell thickness showed no significant difference ( $p > 0.05$ ), ranging from 0.19 to 0.20 mm. A thicker shell offers better protection against microbial contamination and water loss (18);(19). Yolk MDA levels ranged from 1,272 to 1,890 mg/kg. MDA is a marker of oxidative stress resulting from lipid peroxidation. T2 and T3 treatments reduced MDA levels by 23.20% and 28.38% respectively compared to T0, suggesting antioxidant effects of *S. nigrum*. The extract contains flavonoids, carotenoids, phenols, tannins, and alkaloids, which may contribute to this reduction(16)(20).

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### Authors' Contribution

Study concept and design : S.S.D; W.H; and S.

Acquisition of data: S.S.D.

Analysis and interpretation of data: S.S.D; W.H; and S.

Drafting of the manuscript: S.S.D.

Statistical analysis: S.S.D.

Administrative, technical, and material support: S.S.D; W.H; and S.

### Ethics

All animal procedures were approved by the Animal Ethics Committee School of Veterinary Medicine and Biomedical Science, IPB University, Bogor, Indonesia (Approval Number : 274/KEH/SKE/XIII/2024)

### Conflict of Interest

The authors declare that they have no conflict of interest

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### Data Availability

The data that support the findings of this study are available on request from the corresponding author

The author acknowledge the use of generative Artificial Intelligence (AI) in the preparation of this manuscript. AI used in the preparation was ChatGPT by OpenAI. AI was used to translating and improving overall language clarity. The AI tool did not contribute to the design of the study, collecting data, data analysis, or interpretation of results, and all content that AI assisted is carefully edited by author to verify appropriateness with the research objective.

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