



## Potential of Dried Burdock Leaves as a Feed Additive in Laying Quail (*Coturnix Coturnix Japonica*): An Experimental Study

Kurutulmuş Dulavrat Otu Yapraklarının Yumurtacı Bildırıcınlarda (*Coturnix Coturnix Japonica*) Yem Katkı Maddesi Olarak Kullanım Potansiyeli: Deneysel Bir Çalışma

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**Abstract:** This study aimed to evaluate the effects of dried burdock leaves (*Arctium lappa*) as a feed additive on production performance and egg quality in laying Japanese quails (*Coturnix coturnix japonica*). Ninety female quails were randomly assigned to three groups receiving a control (0.0%) or diets supplemented with 1.0% and 2.0% dried burdock leaves (DBL) for six weeks. Although dietary DBL supplementation did not significantly affect overall performance parameters, a numerical increase in feed intake was observed. However, certain egg quality traits, including relative eggshell weight, yolk color, and malondialdehyde (MDA) concentration, were notably influenced by DBL inclusion. Yolk color intensity significantly decreased in the 1.0% DBL group but increased in the group supplemented with 2.0% DBL. In contrast, eggshell weight and MDA levels were reduced across the DBL-supplemented treatments. Other egg quality parameters, such as shell thickness and internal quality indices, remained unaffected by DBL supplementation. These findings suggest that incorporating 2.0% DBL into laying quail diets may improve antioxidant status and yolk pigmentation without compromising overall laying performance. Nonetheless, the observed reduction in relative eggshell weight indicates that higher DBL inclusion levels might exert a mild negative effect on shell formation. Further research is required to determine the optimal inclusion level and to understand the mechanisms underlying its effects.

**Keywords:** Dried burdock leaves, Performance, Egg quality, Laying quail

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**Öz:** Bu çalışma, kurutulmuş dulavrat otu (*Arctium lappa*) yapraklarının yem katkı maddesi olarak yumurtacı Japon bildırıcınlarında (*Coturnix coturnix japonica*) üretim performansı ve yumurta kalitesi üzerindeki etkilerini değerlendirmeyi amaçlamıştır. Doksan dişi bildırıcın, altı hafta boyunca kontrol (0.0%) veya %1.0 ve %2.0 kurutulmuş dulavrat otu (DBL) içeren rasyonlar ile beslenecek şekilde rastgele üç gruba ayrılmıştır. Rasyona DBL ilavesi genel performans parametrelerini önemli ölçüde etkilememiş olsa da, yem tüketiminde sayısal bir artış gözlemlenmiştir. Bununla birlikte, nispi yumurta kabuğu ağırlığı, yumurta sarısı rengi ve malondialdehit (MDA) konsantrasyonu dahil olmak üzere belirli yumurta kalitesi özellikleri, DBL katılımından belirgin şekilde etkilenmiştir. Yumurta sarısı renk yoğunluğu %1.0 DBL grubunda önemli ölçüde azalmış, ancak %2.0 DBL ilave edilen grupta artmıştır. Buna karşılık, yumurta kabuğu ağırlığı ve MDA seviyeleri DBL ilave edilen uygulamaların tamamında azalmıştır. Kabuk kalınlığı ve iç kalite indeksleri gibi diğer yumurta kalitesi parametreleri, DBL ilavesinden etkilenmeden kalmıştır. Bu bulgular, yumurtlayan bildırıcın rasyonlarına %2.0 DBL dahil edilmesinin, genel yumurtlama performansından ödün vermeden antioksidan durumunu ve yumurta sarısı pigmentasyonunu iyileştirebileceğini göstermektedir. Bununla birlikte, nispi yumurta kabuğu ağırlığında gözlemlenen azalma, daha yüksek DBL dahil etme seviyelerinin kabuk oluşumu üzerinde hafif bir olumsuz etki yaratabileceğine işaret etmektedir. En uygun kullanım düzeyinin belirlenmesi ve etkilerinin altında yatan mekanizmaların anlaşılması için daha fazla araştırmaya ihtiyaç vardır.

**Anahtar Kelimeler:** Kurutulmuş dulavrat otu, Performans, Yumurta kalitesi, Yumurtacı bildırıcın

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## INTRODUCTION

Antibiotics have been included in poultry feed at subclinical levels as feed additives due to their antimicrobial growth-promoting properties (Li et al., 2016). However, the indiscriminate use of antibiotics has led to the accumulation of residues in poultry products and the development of microbial resistance. Moreover, antibiotics are often poorly absorbed in the poultry gut and are usually excreted without being metabolized. These excreted antibiotics accumulate in the environment, enter the human body through the food chain, and cause further residue buildup (Rafiq et al., 2022). Due to these harmful effects, the development of alternative feed additives that can replace antibiotics as growth promoters and performance enhancers is of great importance to the poultry industry (Khaligh et al., 2011). Organic acids, enzymes, probiotics, prebiotics, and phytochemical substances are among the promising alternatives to antibiotics (Lee et al., 2022).

In the Classical Chinese Pharmacopoeia, many formulations of Chinese herbs (CH) have been described for the treatment of diseases and the maintenance of human health (Li et al., 2016). CH are primarily known for their immunostimulant and antioxidant effects, attributed to their phytochemicals such as polyphenols, flavonoids, polysaccharides, and lectins (Li et al., 2016; Liu et al., 2023). Moreover, the use of CH as feed additives in poultry production has recently gained significant attention as a potential alternative to antibiotics (Li et al., 2016; Li et al., 2024). Studies have shown that CH hold promise as feed additives in poultry, enhancing growth performance, boosting immune responses, and modulating gut microbiota. CH such as sweet basil, fennel, and *Astragalus membranaceus* have been associated with improved average daily gain, feed efficiency, antibody production, and increased populations of beneficial gut bacteria (Gong et al., 2014).

Burdock (*Arctium lappa*) is a medicinal and aromatic plant used in traditional Chinese medicine and associated with various biological effects (Itaya et al., 2018). It is rich in bioactive secondary metabolites and serves as an important natural source of flavonoids and lignan compounds (Moro and Clerici, 2021; Pădureț, 2019). Although the roots of the plant are mostly used for nutritional and medicinal purposes, the leaves and seeds of burdock are also utilized (Chan et al., 2011). *In vitro* and *in vivo* studies with burdock have reported that it has antimicrobial, hepatoprotective, antioxidant, antimutagenic and anti-inflammatory effects (Horng et al., 2017; Lou et al., 2010). The burdock leaves contain high levels of total polyphenols, chlorophylls, and carotenoids all of which contribute to their strong antioxidant potential. The antioxidant activity, measured via DPPH radical scavenging, confirmed that these bioactive compounds can effectively combat oxidative stress, highlighting burdock leaves as a potent natural antioxidant source (Pădureț, 2019).

The studies on using burdock plant itself, extract or by-products conducted in poultry are very limited. In a study, the effects of burdock root peel supplementation on the performance, egg quality, and blood biochemical parameters of Hy-Line Brown laying hens was evaluated. The findings demonstrated that dietary supplementation with these extracts, particularly polyphenols, significantly improved laying performance, reduced egg defects, enhanced egg quality (notably the Haugh unit), and favorably altered blood biochemical indices, including reduced ALT, TC, LDL-C, and phosphorus, alongside increased serum calcium levels (Ma et al., 2022). In another study, the researchers indicated that evaluated the prebiotic effects of burdock flour and inulin extract in broiler chickens, using 1% burdock flour in feed for 42 days and 100 mg bird<sup>-1</sup> inulin via gavage for the first three days of life; results showed that while burdock flour compromised intestinal integrity and growth performance, it provided partial protection against *Salmonella kedougou*, whereas inulin extract had no adverse effects but also failed to protect against *Salmonella enteritidis*, highlighting that the form and duration of administration significantly influence the efficacy of burdock-derived additives (Itaya et al., 2018).

Although several studies have investigated the inclusion of CH in laying hen diets, no research on the use of burdock leaves in poultry was found in the literature review. This original research article presents the first known study exploring the effects of dried burdock leaves (*Arctium lappa*) as a feed additive in laying

Japanese quails. The findings offer novel insights into the potential of plant-based supplements to enhance egg quality and oxidative stability in poultry production.

## MATERIAL AND METHOD

### *Animals, Experimental Diets, and Management*

The study was conducted with 90 female 12-week-old Japanese quail (*Coturnix coturnix japonica*) on a quail farm in Eskil District, Aksaray, Türkiye. All birds picked for the study had similar body weights ( $267.88 \pm 21.53$ ). The experimental design involved dividing the quails into three dietary groups, with each group comprising five replicates. Every replicate set contained six quails, and quails were assigned to these groups randomly. The study utilized identical cages measuring 45 cm in width, 30 cm in length, and 20 cm in height. These enclosures were maintained under consistent environmental conditions and featured the necessary equipment to provide unrestricted access to feed and water. The study spanned a period of six weeks, running from October through December 2023. Throughout this duration, the researchers maintained a consistent lighting regimen of 16 hours per day. A commercial food company supplied dried burdock leaves (DBL), which was then finely ground to achieve a 1 mm particle size for incorporation into the experimental diets.

**Table 1.** Chemical composition, total phenolic content, and antioxidant capacity of dried burdock leaves.

*Çizelge 1. Kurutulmuş dulavrat otu yapraklarının kimyasal bileşimi, toplam fenolik madde içeriği ve antioksidan kapasitesi.*

| Nutritional components*                             |       |
|-----------------------------------------------------|-------|
| Dry Matter, %                                       | 93.09 |
| Crude protein, % DM                                 | 15.39 |
| Crude ash, % DM                                     | 15.00 |
| Crude fat, % DM                                     | 0.78  |
| Acid Lignin Fiber, % DM                             | 18.60 |
| Neutral Lignin Fiber, % DM                          | 40.80 |
| Total Carotenoid Composition (mg kg <sup>-1</sup> ) | 0.11  |
| DPPH (% reducing)                                   | 28.65 |
| Total phenolic content (mg GAE kg <sup>-1</sup> )   | 52.18 |

\*DM: Dried Matter, DPPH: 2,2-diphenyl-1-picrylhydrazyl

Prior to its use, the powder underwent proximate analysis in accordance with the methods described by AOAC (2000). The method outlined by Kılınc et al. (2023) was employed to measure the total phenolic content, the antioxidant scavenging ability and inhibitory action. Additionally, the total carotenoid content of the DBL was quantified using the technique detailed by Kara and Baytok (2017). All the collected values were shown in Table 1. The control diet, no DBL included, was designed to meet the nutritional standards for laying quails as recommended by the National Research Council (1994). The experimental diets were developed by incorporating DBL into the control diet in mash form at inclusion levels of 1.0, and 2.0%. All diets were formulated to be isocaloric (2800 kcal ME kg<sup>-1</sup>) and isonitrogenous (20% crude protein), ensuring no variations in energy or protein content among the experimental groups. Identical analytical techniques employed for DBL were utilized to determine the basal diet's chemical composition (Table 2).

### *Determination of Laying Performance Parameters*

To evaluate changes in their mass, the quails were weighed at the beginning and end of the study using a high-precision digital scale with an accuracy of  $\pm 0.01$  g. Feed intake per cage was assessed by calculating the difference between the initial feed quantity and the remaining feed at the end of each experimental period. The average daily feed consumption was determined by dividing the total feed intake by the number of quails and the experiment duration, reported as grams per bird per day. The body weight gain in grams over the course of the study was calculated by subtracting the initial body weight from the final body weight.

Throughout the experiment, eggs were gathered daily at a consistent time (16:00) and recorded to determine egg production, which was expressed as a percentage of the total quails per cage. Separately, damaged or cracked eggs were tallied, and their ratio to overall egg production was computed. In the last three days of the study, individual eggs were weighed on a precision scale ( $\pm 0.01$  g) to evaluate egg weight. The egg production percentage was then multiplied by the mean egg weight to calculate the total egg mass. Feed conversion ratio (FCR) was calculated by dividing feed intake by egg mass.

**Table 2.** Formulation and chemical composition of basal diet.

Çizelge 2. Bazal rasyonun formülasyonu ve kimyasal bileşimi.

| Ingredients                                      | %       |
|--------------------------------------------------|---------|
| Corn                                             | 52.80   |
| Soybean meal (46% CP)                            | 33.90   |
| Sunflower oil                                    | 3.55    |
| Limestone                                        | 7.55    |
| Dicalcium phosphate                              | 1.34    |
| Common salt                                      | 0.40    |
| Premix <sup>1</sup>                              | 0.25    |
| DL-methionine                                    | 0.21    |
| Total                                            | 100.00  |
| Chemical composition                             | %       |
| Dry matter                                       | 90.86   |
| Crude protein                                    | 20.11   |
| Crude ash                                        | 10.65   |
| Crude fat                                        | 4.93    |
| Crude fiber                                      | 4.13    |
| Methionine+Cystine                               | 0.91    |
| Calcium                                          | 3.22    |
| Available phosphorus                             | 0.38    |
| Metabolizable energy (kcal ME kg <sup>-1</sup> ) | 2819.89 |

<sup>1</sup>Per kg, Premix provides 20000 IU vitamin A (trans retinol acetate), 0.30 mg biotin, 0.0275 mg vitamin B<sub>12</sub>, 10 mg riboflavin, 2.5 mg folic acid, 5 mg thiamin, 10000 IU vitamin D<sub>3</sub> (cholecalciferol), 125 mg vitamin E (tocopherol), 5 mg vitamin K<sub>3</sub>, 112.5 mg nicotinic acid, 37.5 pantothenic acid, 3.75 mg pyridoxine, 10 mg copper, 3 mg iodine, 50 mg iron, 60 mg manganese, 50 mg zinc, 0.75 mg selenium.

### Determination of Egg Quality Traits

Upon completion of the experiment, 30 eggs per group (six from each cage) were individually weighed. The eggs were then cracked on a clean, flat glass surface. The eggshells were separated and left to air-dry at ambient temperature for a period of three days before being weighed to calculate their proportion of the total egg weight. A digital caliper (Tresna®, Guilin, China) with 0.01 mm precision was employed to measure eggshell thickness. Following these steps, the eggshells underwent ashing at 550°C in a muffle furnace (Nüve MF 106) to determine their crude ash content.

Internal quality parameters of the cracked eggs were assessed by evaluating both the albumen and yolk characteristics. Albumen and yolk heights were measured with a height gauge, while the length and width were determined using a digital caliper (Tresna®, Guilin, China). The yolk index, albumen index, and Haugh unit were calculated following the equations provided by Gümüş et al. (2021). Yolk color was evaluated using a DSM YolkFan™ (formerly known as Roche Yolk Color Fan).

Additionally, two eggs were collected from each cage (10 eggs per group) to determine the total carotenoid, cholesterol and lipid peroxidation content of the yolks. The total carotenoid concentration was measured using the procedure outlined by Bidura et al. (2020) and expressed as  $\mu\text{g } 100 \text{ g}^{-1}$  of egg yolk. The method outlined by Gümüş et al. (2021) was employed to determine total cholesterol levels. This procedure

involved homogenizing 0.1 g of hard-boiled egg yolk with 5 mL of isopropyl alcohol, followed by thorough vortexing. The homogenate was then subjected to centrifugation at 3000 rpm for 10 minutes. Subsequently, the supernatant was analyzed using an autoanalyzer equipped with a total cholesterol test kit. The cholesterol content was expressed in milligrams per gram of yolk. The assessment of egg yolk lipid peroxidation was conducted using the thiobarbituric acid (TBA) method outlined by Zeb and Ullah (2016). This method involved extracting one gram of yolk with 5 mL of glacial acetic acid and 1 mL 0.01% butylated hydroxytoluene, followed by agitation for 60 min at ambient temperature. Subsequently, 1 mL of the extracted solution was combined with 1 mL of 4 mM TBA and heated to 95°C for 60 min. The absorbance was then measured at 532 nm and compared against a series of malondialdehyde (MDA) standards to quantify lipid peroxidation levels, expressed as micromoles of MDA per gram of yolk.

### Statistical Analysis

The data obtained were subjected to ANOVA using the general linear model (GLM) procedure of SPSS Software (Version 26, SPSS Inc., Chicago, IL, USA). To analyze the data, the Duncan test was employed for calculating means, while a polynomial contrast test was utilized to evaluate egg quality parameters and performance characteristics. The threshold for statistical significance was established at  $Quadratic < 0.05$ .

## RESULTS AND DISCUSSION

The effects of different levels (1%, 2%) of DBL supplementation on initial body weight, final body weight, body weight gain, egg production, egg weight, egg mass, feed intake and feed conversion ratio of laying quails are given in Table 3. The effect of the addition of DBL to the feed on performance parameters of laying quails was found to be statistically insignificant ( $P > 0.05$ ).

**Table 3.** The effect of dried burdock leaves on egg performance in laying quails.

Çizelge 3. Yumurtacı bıldırcınlarda kurutulmuş dulavrat otu yapraklarının yumurta performansı üzerine etkisi.

| Parameters*                                            | Dried Burdock Leaves (%) |        |        | S.E.M.** | Main Effect | P        |           |
|--------------------------------------------------------|--------------------------|--------|--------|----------|-------------|----------|-----------|
|                                                        | 0.0                      | 1.0    | 2.0    |          |             | Contrast |           |
|                                                        |                          |        |        |          |             | Linear   | Quadratic |
| Initial body weight (g)                                | 262.44                   | 263.64 | 263.02 | 0.97     | 0.895       | 0.732    | 0.755     |
| Final body weight (g)                                  | 266.96                   | 277.51 | 266.85 | 2.47     | 0.125       | 0.491    | 0.056     |
| Body weight gain (g)                                   | 4.53                     | 13.87  | 3.83   | 2.50     | 0.193       | 0.611    | 0.085     |
| Egg production (%)                                     | 95.47                    | 95.47  | 92.49  | 1.10     | 0.477       | 0.428    | 0.361     |
| Egg weight (g)                                         | 13.01                    | 12.51  | 13.61  | 0.22     | 0.130       | 0.598    | 0.054     |
| Egg mass (g quail <sup>-1</sup> day <sup>-1</sup> )    | 12.42                    | 11.94  | 12.62  | 0.28     | 0.620       | 0.969    | 0.339     |
| Feed intake (g quail <sup>-1</sup> day <sup>-1</sup> ) | 35.72                    | 39.48  | 39.13  | 0.90     | 0.169       | 0.074    | 0.593     |
| FCR (g quail <sup>-1</sup> g egg <sup>-1</sup> )       | 2.89                     | 3.31   | 3.14   | 0.11     | 0.275       | 0.186    | 0.358     |

\*FCR: Feed conversion ratio; \*\*S.E.M.: Standard error means.

Although polyphenol-rich feed additives are mostly used to improve performance in livestock, they may also negatively affect digestion in some cases. This can occur at higher doses due to their inhibitory effects on digestive enzymes or through the formation of insoluble complexes with proteins and certain minerals in the intestines, which are then excreted without being absorbed (Mahfuz et al., 2021).

In the current study, although feed intake increased, FCR was negatively affected in the groups supplemented with DBL—not statistically, but numerically. It has been demonstrated that the inclusion of polyphenol-rich feed additives in poultry diets can increase feed intake, possibly due to enhanced saliva production and improved feed palatability (Dissa et al., 2023). Similar to our results, several researchers have reported that supplementation with polyphenol-rich plant leaves increases feed intake in hens (Dissa et al., 2023; Melesse et al., 2011) and older laying quails (Gül et al., 2022). Conversely, polyphenolic feed additives at certain levels may also impair nutrient availability in the intestines by inhibiting the activity of digestive enzymes. This could be attributed to the ability of polyphenols to form insoluble complexes with endogenous proteins and feed particles (Abd El-Hack et al., 2023). Our findings are in line with those of

(Hayajneh, 2019), who reported that dietary supplementation with ginger root powder or apple cider vinegar had no significant effect on FCR but showed a slightly negative impact when compared to antibiotic growth promoters in broiler chickens. Furthermore, Lu et al. (2016) found that dietary supplementation with 15% *Moringa oleifera* leaf significantly increased FCR in laying hens, indicating reduced feed efficiency at higher inclusion levels.

The effects of different levels of DBL supplementation in laying quail diets on damaged egg ratio, shell weight, shell thickness, shell ash content, white index, yolk index, Haugh unit, egg cholesterol, total carotenoids, MDA levels, and yolk color are presented in Table 4. The addition of DBL to the diets had a statistically significant effect ( $P < 0.05$ ) on relative eggshell weight, MDA levels, and yolk color. In the present study, DBL supplementation decreased both relative eggshell weight and MDA levels. Furthermore, yolk color was reduced in the group that received the 1.0% DBL-supplemented diet, whereas the 2.0% DBL group exhibited the most favorable results compared to the control. Conversely, the effects on damaged egg ratio, shell thickness, shell ash content, white index, yolk index, Haugh unit, egg cholesterol, and total carotenoid content were not statistically significant ( $P > 0.05$ ).

**Table 4.** Effect of dried burdock leaves on egg quality traits in laying quails.

Çizelge 4. Yumurtacı bıldırcınlarda kurutulmuş dulavrat otu yapraklarının yumurta kalitesi özellikleri üzerine etkisi.

| Parameters*                                 | Dried Burdock Leaves (%) |                    |                    | S.E.M.** | Main Effect | P        |           |
|---------------------------------------------|--------------------------|--------------------|--------------------|----------|-------------|----------|-----------|
|                                             | 0.0                      | 1.0                | 2.0                |          |             | Contrast |           |
|                                             |                          |                    |                    |          |             | Linear   | Quadratic |
| Damaged egg (%)                             | 0.66                     | 0.37               | 0.45               | 0.15     | 0.15        | 0.746    | 0.496     |
| Relative eggshell weight (%)                | 1.11 <sup>a</sup>        | 0.99 <sup>b</sup>  | 1.05 <sup>ab</sup> | 0.02     | 0.02        | 0.011    | 0.018     |
| Eggshell thickness (µm)                     | 0.28                     | 0.26               | 0.26               | 0.00     | 0.00        | 0.202    | 0.098     |
| Eggshell ash (%)                            | 79.92                    | 78.68              | 79.98              | 0.35     | 0.35        | 0.244    | 0.613     |
| Albumen Index                               | 3.11                     | 3.26               | 3.11               | 0.06     | 0.06        | 0.547    | 0.732     |
| Yolk Index                                  | 48.87                    | 50.87              | 50.29              | 0.45     | 0.45        | 0.183    | 0.102     |
| Haugh Unit                                  | 90.09                    | 90.06              | 88.61              | 0.09     | 0.09        | 0.461    | 0.405     |
| Cholesterol (mg yolk <sup>-1</sup> )        | 644.16                   | 657.40             | 662.79             | 6.35     | 6.35        | 0.496    | 0.246     |
| Total Carotenoids (µg 100 g <sup>-1</sup> ) | 23.14                    | 22.27              | 24.98              | 2.53     | 2.53        | 0.915    | 0.873     |
| MDA (µM MDA g yolk <sup>-1</sup> )          | 1.30 <sup>a</sup>        | 1.19 <sup>ab</sup> | 1.06 <sup>b</sup>  | 0.04     | 0.04        | 0.026    | 0.013     |
| Yolk Color (1-15)                           | 11.05 <sup>b</sup>       | 7.40 <sup>c</sup>  | 11.90 <sup>a</sup> | 0.28     | 0.28        | 0.000    | 0.013     |

\*MDA: malondialdehyde; \*\* S.E.M.: Standard error means.

a,b: Means with different superscripts in the same row were significantly different ( $P < 0.05$ ).

Egg quality is determined by various standards that define both internal and external characteristics. Internal quality includes factors such as yolk height, yolk color, albumen viscosity, and Haugh unit, while external quality involves traits like eggshell thickness, egg width, height, and cleanliness (Abebe et al., 2023; Kumar et al., 2022).

The composition and quality of eggs can be influenced by the inclusion of different feed additives in poultry diets (Li et al., 2016). In the present study, a significant decrease in eggshell weight was observed with the inclusion of DBL in laying quail diets. Similarly, Zhu et al., 2020 reported that dietary supplementation with tea polyphenol products in laying hen diets significantly reduced eggshell weight at higher inclusion levels. Moreover, Gümüş et al. (2025) also found that the eggshell ratio in quail eggs decreased with increasing levels of red cabbage waste. On the contrary, several researchers have reported that polyphenol-rich feed additives did not negatively affect eggshell weight in laying hens (Dissa et al., 2023; Kılınç et al., 2023) and in laying quails (Gül et al., 2022; Gümüş and Sevim, 2024). Overall, the impact of dietary polyphenol supplementation on eggshell weight and quality in laying quails and hens could vary with the type and concentration of polyphenols used. The choice of polyphenol and its dosage might be crucial in determining the outcome on eggshell characteristics.

According to our observations, MDA levels decreased linearly with increasing levels of DBL supplementation, with the greatest reduction observed in the 2.0% DBL group. A lower MDA concentration indicates reduced lipid peroxidation, suggesting enhanced oxidative stability. The improved antioxidant status associated with burdock supplementation may be attributed to its phenolic derivatives, which are known for their antioxidant properties (Pădureț, 2019). Similar to our findings, other researchers have reported that polyphenol-rich feed additives reduced MDA levels in laying hens, such as Chinese herbal mixtures (Liu et al., 2023; Yu et al., 2023) and *Moringa oleifera* leaves (Lu et al., 2016).

In the present study, 2.0% DBL supplementation resulted in the most intense yolk color in laying Japanese quails compared to the control group. Similarly, Li et al. (2016) reported that the inclusion of a Chinese herbal mixture in the diets of laying hens enhanced yolk pigmentation. Lu et al. (2016) also found that dietary supplementation with *Moringa oleifera* leaves deepened yolk color in laying hens. Burdock is rich in carotenoids, particularly lutein and zeaxanthin (Pădureț, 2019), which are key contributors to yolk pigmentation, as carotenoids are responsible for the yellow and orange hues (Li et al., 2016). Conversely, the 1.0% DBL group exhibited the lowest yolk color intensity compared to the control. Karunaratne et al. (2025) similarly reported that supplementation with polyphenol-rich sugarcane extract in the drinking water of laying hens led to a significant reduction in yolk color at week 45. However, the contradictory results observed in the current study are not entirely clear. One possible explanation is that polyphenols can exert pro-oxidant effects in animal metabolism, potentially accelerating the oxidation of yolk pigments (Abd El-Hack et al., 2023). Another possibility is that DBL may contain compounds such as tannins that interfere with the absorption of nutrients, including fat-soluble carotenoids. In this case, the 1.0% DBL diet may not have provided enough beneficial pigments to compensate for the inhibitory effects on nutrient absorption.

## CONCLUSION

This study demonstrated that DBL supplementation in laying quail diets had limited impact on overall production performance but positively influenced specific egg quality traits. While egg production and feed efficiency remained statistically unaffected, the inclusion of DBL—particularly at 2.0%—enhanced yolk pigmentation and significantly reduced yolk MDA levels, indicating improved antioxidant status. However, a reduction in eggshell weight and the unexpected decline in yolk color at 1.0% DBL suggest that the effects of DBL were dose-dependent and complex. These findings highlight DBL's potential as a natural antioxidant-rich feed additive, though further research is needed to optimize its use. Future studies should focus on evaluating different dosages and durations, elucidating absorption mechanisms, and exploring its influence on gut health and nutrient interactions to fully understand its efficacy in poultry nutrition.

## CONFLICT OF INTEREST

The authors declare that there is no conflict of interest regarding the publication of this article.

## DECLARATION OF AUTHOR CONTRIBUTION

EG, BS and EK designed the study and supervised the research process. Cİ was responsible for animal care and data collection. EG and BS also conducted laboratory analyses and contributed to data interpretation. OK assisted with the ethical committee approval process. All authors contributed to the writing, reviewed the final manuscript, and approved it for publication.

## DECLARATION OF ETHICS COMMITTEE

The research protocols for this study were approved by the Ethics Committee for Animal Experiments of the Faculty of Veterinary Medicine, Selçuk University (Decision No: 2023/141). The guidelines set forth in the European Animal Protection Policy were rigorously followed throughout the experiment.

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