*Indian Journal of Animal Sciences* **91** (1): 58–62, January 2021/Article
https://doi.org/10.56093/ijans.v91i1.113276

# Effect of supplementation of red beetroot powder in quail diets

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Received: 24 September 2020; Accepted: 30 March 2021

ABSTRACT

This study aims to determine the effects of different levels of supplements (0, 0.5, 1.0 and 1.5%) of red beetroot (*B. vulgaris* var. *rubra* L.) powder in the diets of laying quails on productive traits, egg quality, some blood parameters, yolk fatty acid profiles and cholesterol levels. A total of 112, 12-week-old female quails were used in the 4-week study. While the red beetroot powder (RBP) significantly affected egg yield, it was not effective on daily feed intake (DFI), feed conversion ratio (FCR) and egg weight (EW). Among the blood parameters, there were significant differences in terms of the triglyceride (TG), Ca and Na values among the trial groups. In terms of the egg yolk fatty acid profiles, there were significant differences among the palmitic acid and linolenic acid levels of the groups. The results obtained in terms of the egg yolk cholesterol levels showed that the RBP did not significantly affect the cholesterol levels. Consequently, considering its positive effects on some parameters like egg yield, egg yolk colour and palmitic and linolenic acid values, it may be recommended to add RBP to the diets of quails by up to 1%.

**Keywords:** Blood parameters, Egg quality, Fatty acids, Yolk cholesterol

Beetroot is grown in many parts of the world, especially in Asia, North Africa, Europe and North America (Kowalski and Szadziñska 2014). Beetroot is a plant belonging to the Chenopodiaceae family and has many varieties ranging from yellow to red. The dark red root variety is particularly popular because it is widely consumed by people (Singh and Hathan 2014).

In recent years, red beetroot (*Beta vulgaris rubra*) has received an increasing interest in physiological and biological activity due to its effects as a functional food (in terms of health and disease) (Clifford *et al.* 2015). Red beetroots are rich in valuable active compounds such as carotenoids, betalains, polyphenols, flavonoids and saponins (Kaur and Singh 2014). Red beetroots, which contain a significant amount of colour pigments (betalain), contain substances such as antioxidant betanin, isobetanin, vulgaxanthin and various compounds with antimicrobial properties. Red beetroot is also rich in vitamin content. This plant is especially rich in vitamins A, B and C, and it contains Fe, Mg, Ca, K and folic acid. Beetroot, a powerful antioxidant, supports blood production in the body, increases the number of red blood cells and strengthens the immune system. Other important effects of red beetroot are strengthening the stomach and the intestinal system, facilitating digestion and regulating bowel activities.

Red beetroot is consumed in different ways in the human diet and may also be used in poultry diets. For this purpose, although very limited, there are studies about the effects of red beetroot added into animal feeds on the performance and health of the animals. Kopřiva *et al.* (2014) reported that yolk colour was not significantly affected when 1–2% of dried RBP was added to the diets of laying hens. Selim *et al.* (2013) found that red beetroot extract, which was added as a natural antioxidant source to broiler diets in the summer season on levels of 0.5–1.0%, affected FI and FRC, however, lipid and cholesterol levels in the plasma were comparable.

This study investigated the effects of RBP added to quail diets on performance traits, egg quality, blood parameters, yolk fatty acid profile and cholesterol content.

## MATERIALS AND METHODS

*Housing:* The study was conducted in a poultry house of Bingol University, Turkey. The experiment was designed as a factorial treatment arrangement with 4 groups and 4 replications. A total of 112 laying female Japanese quails were used in the experiment, 7 per repetition. The study was conducted for 4 weeks, between January and March 2019. A photoperiod of 16 hrs light to 8 hrs dark was applied for the study. The RBP that was used in the experiment was obtained from a commercial company.

*Diets:* The experimental diets were prepared based on nutrient analysis. In the study, 0.5, 1.0 and 1.5% of RBP was added to the diets of the treatment groups. All the experimental quails were fed *ad lib.* with feeds containing 20% CP and 3,000 kcal/kg ME for 4 weeks (Table 1). According to analysis results of RBP used in the experiment,

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Table 1. Ingredients and chemical composition of diets

| Ingredients (%) | Control | P1 | P2 | P3 |
|---|---|---|---|---|
| Corn | 53.394 | 52.749 | 52.105 | 51.460 |
| Soybean meal | 32.323 | 32.327 | 32.331 | 32.335 |
| RBP | 0 | 0.500 | 1.000 | 1.500 |
| Sunflower oil | 6.736 | 6.874 | 7.012 | 7.150 |
| Limestone | 5.440 | 5.439 | 5.437 | 5.436 |
| DCP | 1.595 | 1.598 | 1.601 | 1.603 |
| Methionine | 0.062 | 0.063 | 0.064 | 0.066 |
| Salt | 0.200 | 0.200 | 0.200 | 0.200 |
| Vit-Min. | 0.250 | 0.250 | 0.250 | 0.250 |
| Total | 100.000 | 100.000 | 100.000 | 100.000 |
| *Calculated values* (%) | | | | |
| Metabolizable energy (kcal/kg) | 3,000 | 3,000 | 3,000 | 3,000 |
| Crude protein | 20.000 | 20.000 | 20.000 | 20.000 |
| Ether extract | 8.650 | 8.770 | 8.890 | 9.010 |
| Cellulose | 2.430 | 2.420 | 2.400 | 2.390 |
| Ash | 10.030 | 10.060 | 10.090 | 10.110 |

P1, 0.5% RBP; P2, 1% RBP; P3, 1.5% RBP.

its dry matter content was 93.75%, crude ash content was 6.66%, organic matter content was 87.09%, crude fat content was 0.94%, sugar content was 6.55%, starch content was 0.5%, crude protein was 10.19%, and 1,546.80 kcal/kg ME.

*Parameters:* In the experiment, DFI, FCR, egg yield and egg quality characteristics were determined. Some blood parameters measured. Fatty acid profiles and cholesterol levels were determined in yolks.

*Egg quality analysis:* An electronic scale was used for egg weighing, and a digital caliper was used for egg width and length measurements. A tripod micrometer was used for yolk and albumen heights, and a digital micrometer was used for shell thickness. The yolk colour of the eggs was determined by using a Digital Yolk Fan. The necessary calculations were made by using the following formulae:

$$\text{Shape index} = \frac{\text{Egg width}}{\text{Egg length}} \times 100$$

Albumen weight = Egg weight – (shell weight + yolk weight)

$$\text{Yolk index} = \frac{\text{Height of yolk (mm)}}{\text{Diameter of yolk (mm)}}$$

$$\text{Albumen index} = \frac{\text{Albumen height (mm)}}{\text{Average of albumen length and width (mm)}} \times 100$$

Haugh Unit=100 log (H+7.57–1.7W0.37) [H=Albumen height, W=Egg weight]

*Blood analysis:* At the end of the experiment, 4 birds from each group were slaughtered and their samples were placed into yellow-cap blood collection tubes with BD Vacutainer gel. The glucose (OSR 6121), TG, total cholesterol (TC), LDL, Ca, P, Mg, K and Na analyses of the serum samples were carried out with an auto-analyzer (Olympus AU400), using commercial kits (Beckman Coulter) by the photometric method. The method described by Folch *et al.* (1957) was used to determine the fatty acid profiles in the yolks. Cholesterol content was determined on the samples described by Keser (2016).

*Statistical analysis:* The data obtained from the study were analyzed using the SAS 9.1.3 program. Duncan's test was used to determine the differences between the mean values which were considered significant.

## RESULTS AND DISCUSSION

The average DFI of the experimental groups varied between 30.61 and 31.57 g in the 1st week and 26.42 and 28.39 g in the 4th week. No significant differences were observed among the groups in terms of the DFI in the quails. This clearly indicated the DFI was not affected by the RBP used on different levels. The results of this study were similar to those reported by Souza *et al.* (2019). The researchers reported that red beet pulp added to the diets of laying hens up to 0.8% did not affect DFI. Similarly, feeding studies with broilers have indicated that addition of beet extract and pulp to chicken diets does not have a negative effect on their feed consumption (Pettersson and Razdan 1993, Selim *et al.* 2013). The FCR of the experimental groups varied between 3.25–3.66 in the 1st week and 3.27–3.82 in the 4th week.

The differences between the groups were found to be insignificant. The results of this study were consistent with those of Souza *et al.* (2019), in which 0.8% red beet pulp was added to the diets of laying hens. The results of both Pettersson and Razdan (1993) and Selim *et al.* (2013) supported our results, whereas the results reported by Kumari *et al.* (2014) differed from those in the present study.

Egg yields of the treatment groups have significant ($P<0.05$) differences except for the 2nd week. In the 1st and 3rd weeks, the 1% supplemented group had higher egg production than the control group. Egg yield of the groups varied between 74.49–86.23% in the 1st week, 74.49–88.27% in the 3rd week, and 71.94–88.78% in the 4th week. The egg yield of the group fed diet with 1.5% beet powder decreased significantly in comparison to the other groups at the 4th week. Adding beet powder to the diet up to 1% did not cause any negative effects on the egg yields. However, the egg yields were negatively affected when the percentage of beet powder added to the diet exceeded 1%. Souza *et al.* (2019) reported that egg yield in laying hens was not affected when 0.8% red beet pulp was added to their diets (Souza *et al.* 2019). The results of this study regarding egg yields were similar to those in the literature.

Adding RBP to the diet up to 1.5% did not cause a significant difference between the EW of the groups. EW of the treatment groups varied between 11.18–11.30 g in the 1st week and 10.32–10.69 g in the 4th week.

The results of this study regarding EW were similar to those of another study in which red beet pulp was added to the diets of laying hens (Souza *et al.* 2019).

The shell weight and yolk index were significantly affected in the 2nd week, and the shape index and yolk colour

Table 2. Exterior and interior quality traits of the groups

| Group | Egg weight (g) | Shape index | Shell weight | Shell thickness (g) | Albumen weight (mm) | Yolk weight (g) | Albumen index (g) | Yolk index | Yolk colour | Haugh unit |
|---|---|---|---|---|---|---|---|---|---|---|
| *2nd wk* | | | | | | | | | | |
| Control | 10.63 | 77.21 | $0.94^{a}$ | 0.22 | 6.42 | 3.27 | 13.15 | 48.21ab | 12.06 | 93.51 |
| P1 | 10.56 | 77.67 | $0.95^{a}$ | 0.22 | 6.36 | 3.25 | 13.27 | 49.50a | 10.75 | 93.31 |
| P2 | 10.44 | 78.40 | $0.92^{ab}$ | 0.22 | 6.22 | 3.29 | 11.96 | 46.96b | 11.62 | 91.51 |
| P3 | 10.48 | 77.69 | $0.86^{b}$ | 0.21 | 6.37 | 3.24 | 12.23 | 46.68b | 11.31 | 91.95 |
| P | 0.72 | 0.65 | 0.04 | 0.37 | 0.46 | 0.94 | 0.20 | 0.04 | 0.10 | 0.24 |
| SEM | 0.53 | 2.67 | 0.09 | 0.01 | 0.37 | 0.28 | 2.08 | 3.17 | 1.50 | 3.33 |
| *4th wk* | | | | | | | | | | |
| Control | 10.34 | $77.91^{ab}$ | 0.91 | 0.20 | 6.27 | 3.16 | 12.99 | 46.09 | 10.94b | 93.24 |
| P1 | 10.57 | $76.52^{b}$ | 0.91 | 0.21 | 6.56 | 3.11 | 12.82 | 48.94 | 11.06b | 93.64 |
| P2 | 10.26 | $76.22^{ab}$ | 0.89 | 0.20 | 6.15 | 3.23 | 13.57 | 47.78 | 12.44a | 93.74 |
| P3 | 10.33 | $79.19^{a}$ | 0.86 | 0.20 | 6.31 | 3.15 | 12.61 | 48.41 | 11.94ab | 92.83 |
| P | 0.37 | 0.02 | 0.24 | 0.51 | 0.17 | 0.85 | 0.40 | 0.10 | 0.04 | 0.77 |
| SEM | 0.52 | 2.45 | 0.08 | 0.01 | 0.52 | 0.39 | 1.67 | 3.38 | 1.71 | 2.72 |

a, b: Differences between means in the same column are significant. P1, 0.5% RBP; P2, 1% RBP; P3, 1.5% RBP.

were significantly affected (Table 2) in the 4th week ($P<0.05$). The other egg quality characteristics were not affected by the RBP. When the shell weight values in the 2nd week were examined, the results obtained in the groups were similar, except for the Group P3. The lowest value in terms of shell weight was measured in the 1.5% supplementation group. The highest values of the yolk index in the 2nd week were obtained from the 0.5% group and control group. The other groups had significantly lower yolk index values (Table 2). In the 4th week, significant differences were observed between the control and treatment groups in terms of the shape index. A significantly ($P<0.05$) higher value in the shape index was measured in the 1.5% group followed by the control and 1% groups. The 0.5% group had a significantly lower shape index ($P<0.05$) than the other groups. In the 4th week, another feature affected by the treatments was yolk colour. The 1% and 1.5% groups had higher yolk color values than the other groups ($P<0.05$).

Ketta and Tùmová (2016) stated that the level of minerals in diet caused structural changes in the egg shell. Similarly, Sawicki *et al.* (2016) reported that beets have a high fibre and mineral content, while Baye *et al.* (2017) found that dietary fibre levels negatively affect mineral absorption. Therefore, increased fibre in diet reduces the absorption of some minerals such as Ca, resulting in a decrease in egg shell weight. In this study, the groups showed statistically significant differences in terms of parameters such as the shape index and egg yolk in the 4th week of the intervention. The colour of the egg yolk is affected by carotenoids, particularly lutein and zeaxanthin, and xanthophyll with their antioxidant effects. *Beta vulgaris* L. ssp. esculenta var. rubra contains, among others, also flavonoids, betanin, quercetin and polyphenols (Kopøiva *et al.* 2014). The results of this study on yolk color were contrary to those reported by Kopøiva *et al.* (2014) and Souza *et al.* (2019).

Significant differences were found between the control and treatment groups in terms of TG, Ca and Na levels (Table 3). The TG level of the control group was significantly lower than those of the treatment groups ($P<0.05$). The highest values of TG were obtained in the 0.5, 1.5 and 1% groups, respectively. In terms of the serum Ca levels, significant differences ($P<0.01$) were found among the groups, and the lowest Ca level was obtained from the control group. The Na levels also showed significant differences ($P<0.05$) among the groups, and they decreased in all treatment groups. High-fibre foods such as beets have been reported to reduce blood cholesterol indirectly (Swain *et al.* 1990; Székely *et al.* 2016). The results of this study regarding serum cholesterol levels differed from those in the literature, suggesting that there was no significant decrease in the serum cholesterol level due to the addition of beets to the diet. Pettersson and Razdan (1993) reported that adding beet pulp to the diet does not generally affect serum cholesterol level. A study in which beet extract was added to broiler rations found no significant differences among the groups in terms of TC, LDL and TG levels (Selim *et al.* 2013). Arslan and Saatci (2003) reported that adding beet pulp to the diet reduced serum cholesterol and TG levels and did not affect glucose levels in geese. The results of this study regarding the blood parameters were supported by the results reported by Pettersson and Razdan (1993), Selim *et al.* (2013) and Arslan and Saatci (2003).

Significant differences ($P<0.05$ and $P<0.01$) were observed only in the PA (Palmitic acid) and ALA ($\alpha$-linoleic acid) levels among the groups (Table 3). It was observed that the PA level was decreased in group P2. In terms of ALA, higher values were obtained in all treatment groups in comparison to the control group. This result differed from those reported by Arslan (2003), who found that adding beet pulp to goose diets was effective on the fatty acid

Table 3. Serum biochemical parameters, fatty acid profile and cholesterol content in yolk of quails at the end of the experimental period

| Parameter | Parameter (mg/dL) | | | | | |
|---|---|---|---|---|---|---|
| | Control | P1 | P2 | P3 | P | SEM |
| Serum glucose | 149.58 | 176.13 | 160.47 | 157.15 | 0.28 | 18.80 |
| Triglyceride | 254.25$^{b}$ | 597.75$^{a}$ | 439.50$^{ab}$ | 492.50$^{ab}$ | 0.04 | 152.60 |
| T. Cholesterol | 137.50 | 145.97 | 158.12 | 153.50 | 0.81 | 32.15 |
| LDL | 18.95 | 11.25 | 15.92 | 18.37 | 0.51 | 7.80 |
| Ca | 10.73$^{c}$ | 16.75$^{a}$ | 15.93$^{a}$ | 13.36$^{b}$ | 0.01 | 1.60 |
| P | 17.35 | 20.96 | 17.95 | 18.05 | 0.54 | 3.74 |
| Mg | 7.14 | 6.39 | 6.19 | 6.29 | 0.24 | 0.69 |
| Na (mmol/L) | 150.25$^{a}$ | 145.00$^{b}$ | 145.75$^{b}$ | 146.25$^{b}$ | 0.04 | 2.59 |
| K ( mmol/L) | 18.33 | 18.87 | 19.07 | 21.22 | 0.70 | 3.69 |
| *Fatty acids* (%) | | | | | | |
| C14:0 | 0.47 | 0.52 | 0.42 | 0.47 | 0.50 | 0.08 |
| C16:0 | 25.30$^{a}$ | 23.70$^{ab}$ | 23.09$^{b}$ | 24.71$^{ab}$ | 0.04 | 1.08 |
| C16:1 | 2.94 | 3.55 | 3.24 | 3.12 | 0.85 | 0.99 |
| C17:0 | 0 | 0.06 | 0.11 | 0.05 | 0.53 | 0.33 |
| C18:0 | 13.77 | 13.25 | 14.34 | 13.53 | 0.88 | 2.00 |
| C18:1 | 29.63 | 29.22 | 29.67 | 30.98 | 0.69 | 2.19 |
| C18:2 | 21.14 | 22.40 | 22.29 | 20.74 | 0.60 | 2.08 |
| C18:3 | 0.98$^{b}$ | 1.46$^{a}$ | 1.35$^{a}$ | 1.37$^{a}$ | 0.01 | 0.16 |
| C20:3 | 0.08 | 0.19 | 0.12 | 0.07 | 0.15 | 0.07 |
| C20:4 | 3.36 | 4.00 | 4.03 | 3.73 | 0.85 | 1.21 |
| C20:5 | 0.92 | 0.00 | 0.11 | 0.09 | 0.38 | 0.81 |
| C22:6 | 1.34 | 1.42 | 1.32 | 1.21 | 0.85 | 0.34 |
| *Cholesterol* (*mg/yolk*) | | | | | | |
| Yolk Cholesterol | 62.75 | 63.85 | 74.50 | 69.45 | 0.34 | 9.82 |

a, b: Differences between means in the same row with different letters are significant (P<0.05).

composition in the tissues, changing the fatty acid composition in general. These changes observed in the tissues were not seen in the egg yolk.

The RBP added to the diets did not significantly affect the egg cholesterol levels. The slight increase in the 1.0% and 1.5% supplemented groups, in comparison to the control group, was not significant.

Beet is a plant with high fibre content (Székely *et al.* 2016). Foods with high fibre content indirectly reduce blood cholesterol levels (Swain *et al.* 1990). The same mechanism was invalid for yolk cholesterol, therefore adding beet powder to the ration did not have a statistically significant effect on the egg yolk cholesterol levels of the groups.

This may have been because of the low level of beet powder addition to the diet in the experiment.

The RBP added up to the level of 1.5% to the diets of quails was not effective on the DFI, FCR and EW values. The RBP was effective on egg yield starting with the 1$^{st}$ week, and a higher yield was obtained from the 1% supplementation group than the other groups. While the values regarding egg yield were similar in the 2$^{nd}$ week of the experiment, the 0.5% and 1% groups provided higher values respectively in the 3$^{rd}$ and 4$^{th}$ weeks. In terms of egg quality, among the characteristics measured in the 2$^{nd}$ week, while there were significant differences in terms of shell weight and the yolk index, the other characteristics were not affected by addition of beetroot powder. In the measurements in the 4$^{th}$ week, the shape index and yolk color differences in the trial groups were found significant. If was determined that the RBP led to a significant darkening especially in the egg yolk. This result was significant in terms of the usability of RBP as a natural food for the colour of egg yolk rather than synthetic colourants. Regarding the blood parameters, there were significant differences in the TG, Ca and Na levels among the groups. While the TG levels were significantly different between the 0.5% supplementation group and the control group, the values of the other groups were similar to the control group. While the Ca levels were higher in the trial groups in comparison to the control group, the highest values were in the 0.5% and 1% supplementation groups. It may be stated that beetroot powder addition affected and lowered the blood Ca levels. According to the results of the analysis conducted in terms of the fatty acid profiles of the egg yolk, there were significant differences among the groups regarding their palmitic acid and linolenic acid levels. The RBP became effective on these two fatty acids, and the control group had a higher value than the 1% supplementation group in terms of palmitic acid. In terms of linolenic acid, the values in the trial groups were higher than those in the control group. It was determined that the RBP significantly affected and increased the linolenic acid levels of the egg yolk. The results showed that the RBP did not affect the cholesterol levels significantly in the egg yolk.

Consequently, it was observed that the RBP added as an additive to the diets of laying quails affected some yield characteristics or some parameters of the quails. Additions made up to the level of 1.5% to the diet did not reveal any negative effects. Additionally, considering its positive effects on some parameters like egg yolk colour and palmitic and linolenic acid levels, it may be recommended to add RBP to the diets of quails by up to 1%.

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