

The effects of Pennyroyal (*Mentha pulegium L.*) on performance, carcass and meat quality in Japanese quails (*Coturnix coturnix japonica*)

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Abstract: This study investigated the Pennyroyal in terms of its effects on performance and meat quality (color and pH) in Japanese quails in different levels. A total of one day-old Japanese quail chicks were randomly distributed to a control and two experimental groups containing three different levels of pennyroyal (0.0, 0.1 and 0.2 %). Each experimental group was divided into five repetitive groups with, ten quail chicks in each group. At the end of the experiment, the performance values and meat quality parameters (color and pH) were measured. Significant differences were found between the groups according to the growth performance of the quails ($P<0.05$). At the end of the study, the body weight and body weight gain of the experimental groups in which 0.1% (P1) and 0.2% (P2) pennyroyal supplemented were found to be higher than the control group. It was observed that the feed consumption increased in the P2 group compared to the control group, and the feed conversion rate decreased ($P<0.05$). There was no significant difference was observed in the carcass yield, heart, liver and gizzard weight ($P>0.05$). The brightness (L^*), redness (a^*), yellowness (b^*) and pH values of the breast meat were not affected by the supplementation of pennyroyal ($P>0.05$). According to the results of the study, the supplemented of pennyroyal by 0.2% was more effective on the performance.

Keywords: Meat color, pennyroyal, performance, pH, quail.

Yarpuzun (*Mentha pulegium L.*) Japon bildircinlarında (*Coturnix coturnix japonica*) performans, karkas ve et kalitesi üzerine etkisi

Özet: Bu çalışmada, Yarpuzun (*Mentha pulegium L.*) farklı seviyelerde Japon bildircinlarında performans ve et kalitesi (renk ve pH) üzerine etkisi araştırılmıştır. Toplam 150 adet günlük yaşta Japon bildircin civcivi üç farklı düzeyde yarpuz içeren (%0,0, 0,1 ve 0,2) bir kontrol ve iki deneme grubuna rastgele dağıtıldı. Her deneme grubu içerisinde 10 bildircin bulunan beş tekerrürlü gruba ayrıldı. Deneme sonunda performans değerleri ve et kalite parametreleri (renk ve pH) ölçüldü. Bildircinların büyüme performansı değerlendirildiğinde gruplar arasında önemli farklılıklar bulundu ($P<0,05$). Çalışma sonu itibarıyla %0,1 (P1) ve %0,2 (P2) yarpuz ilave edilen deneme gruplarının canlı ağırlık ve canlı ağırlık artışları kontrol grubuna oranla yüksek bulundu. Yem tüketiminin P2 grubunda kontrol grubuna oranla arttığı, yemden yararlanma oranının ise düştüğü gözlemlendi ($P<0,05$). Karkas randımanı, kalp, karaciğer ve taşlık ağırlıkları açısından anlamlı bir fark olmadığı görüldü ($P>0,05$). Göğüs etinin parlaklık (L^*), kırmızılık (a^*) ve sarılık (b^*) ile pH değerleri yarpuz ilavesinden etkilenmedi ($P>0,05$). Çalışma sonuçlarına göre yarpuzun %0,2 düzeyinde ilavesinin performans üzerine daha etkili olduğu belirlendi.

Anahtar sözcükler: Bildircin, et rengi, performans, pH, yarpuz.

Introduction

Mentha pulegium L., which belongs to the *Mentha* species of the *Labiatae* family, is ubiquitous in Europe, North Africa and Central Asia and known as pennyroyal (11). It has been used as flavorant, expectorant, appetite

regulator or diuretic for many years. In addition, it is widely used in the treatment of diseases such as feed poisoning, bronchitis and tuberculosis (21). In addition to these effects, it has also been reported to have aromatic regulation, antioxidant, anti-inflammatory and

antimicrobial effects due to the phenolic compounds in its structure, such as Linalool, Menthone, p-Menthan, Pulegone, Piperitenone oxide (20, 35, 36). After the prohibition of antibiotics in the breeding of poultry, pennyroyal and aromatic plants are reported to be useful due to its effects on increasing digestibility, balancing the intestinal microbial ecosystem and excreting endogenous digestive enzymes (13, 17, 40). There is a growing awareness in consumers with the development of technology in the field of feed safety, which led to a growing interest in the use of medical aromatic plants that are not harmful to human and animal health as well as their various forms (18, 22, 32). In many studies, it was found that pennyroyal and other *Mentha* species make positive contributions to the efficiency and shelf life of animal products (5, 16, 26).

The main aim of this study was to investigate the Pennyroyal (*Mentha pulegium L.*) in different levels supplemented to quail rations in terms of its effects on growth performance and breast meat quality (color and pH).

Materials and Methods

Animals, experimental design and feed: The experiment was carried out in the Kafkas University Research and Practice Farm. This study was approved by the Kafkas University Animal Experiments Local Ethics Committee (KAÜ-HADYEK/2020-008). A total of 150 a day old Japanese quails (*Coturnix coturnix japonica*) were used as the animal material. The study was performed with one control and two experiment groups, each of which included 50 quail chicks. All groups were divided into five subgroups, each containing 10 quail chicks. Dried and powdered pennyroyal was added to the basic ration of the control and experimental groups by 0.0%, (control) 0.1% (P1) and 0.2% (P2), respectively. The quail chicks were placed in plastic cages of 60×20×100 cm. The study was completed in a total of 35 days, including 7 days of adaptation and 28 days of feeding periods. The animals were given feed and water *ad libitum*. Throughout the study, all animals were kept at 32-33°C for the first three days, which was then fixed to 25°C by a gradual decrease of 1-2°C on a weekly. The cages were kept illuminated for 24 hours/day for 35 days. The rations were prepared in accordance with the NRC (14), and chemical analyses were carried out in accordance with the AOAC (25) (Table 1).

Feed additive: Plants (pennyroyal) were collected from Kars province (40 ° 48'21.2 "N 42 ° 53'37.8" E) in Turkey and harvested in its season. Plants' leaves were dried at room temperature without direct exposure to the sun light. After drying, they were powdered for supplementation to the ration. The essential oil was obtained from Pennyroyal through vapor distillation

method. The main active compositions of the pennyroyal leaves essential oil were determined by GC/MS and contained 13.61% of Linalool, 10.56% of p-Menthone, 6.19% of p-Menthan, 4.45% of Pulegone, 0.11% of Isopulegone, 3.07% of Piperitenone oxide, and 0.49 of Thymol phenolic compounds (35).

Table 1. Ingredients and nutrient composition of diet.

| Ingredients | % |
|--------------------------|-------|
| Corn, yellow | 46.30 |
| Soybean meal (48% CP) | 32.41 |
| Barley meal | 11.00 |
| Corn gluten meal | 4.40 |
| Wheat bran | 2.12 |
| Vegetable oil | 0.60 |
| Limestone | 1.80 |
| Dicalcium phosphate | 1.12 |
| Salt | 0.30 |
| DL-methionine | 0.10 |
| L-lysine sulfate | 0.40 |
| Vit-min mix ¹ | 0.25 |
| Analyzed values | |
| CP (%) | 24.00 |
| ME(kcal/kg) | 2901 |
| Ca (%) | 0.80 |
| Av. P (%) | 0.38 |

CP: Crude protein, ME: Metabolism energy, Ca: Calcium, Av. P: Available phosphorus

¹ Vit-min mix: Vit A; 15.00 IU/kg, Vit D; 3.30 IU/kg, Vit E; 25.00 IU/kg, K; 0.86%, Mg; 0.16%, S; 0.21%, Fe; 146.89 mg/kg, Mn; 121.11 mg/kg, I; 0.50 mg/kg, Zn; 110.03 mg/kg, Cu; 13.05 mg/kg.

Performance: Body weight (BW) and feed consumption (FC) were determined in all groups every week. The increase in body weight gain (BWG) and feed conversion rate (FCR) were calculated (FC/BWG) based on the differences obtained as a result of the weighing processes. At the end of the study, 10 quails were randomly selected from each group to analyze their carcass traits. The animals were not fed for 12 hours. Then, they were weighed individually and slaughtered after determining their slaughtering weight. The blood of the slaughtered animals was drained, and their plume was plucked. Subsequently, internal organs (heart, liver and gizzard) were removed to calculate the carcass and internal organ weights.

Determining the color quality and pH of meat:

Breast meat samples were taken from the slaughtered animals to determine the color intensity and pH. A colorimeter (Minolta-Japan) was used to determine the color intensities of the breast meat samples (L*: brightness, a*: redness, b*: yellowness). The color intensities were determined as per the characteristics specified by the International Commission on Illumination. Average values were determined through the

measurements performed at four different parts of each sample. In order to determine the pH values of the breast meat samples, 10 grams of breast meat was added to 100 ml of purified water. Then it was homogenized and measured using a pH meter (SCHOTT L 6880).

Statistical analysis: The performance and the parameters of breast meat quality (color, pH) were evaluated on the SPSS 20.0 (IBM-USA) statistics software. The difference between the results of the groups was analyzed with one-way variance analysis (ANOVA). The Duncan, multiple comparison test, was used in the bilateral comparisons between the groups. The level of significance was determined to be at $P < 0.05$.

Results

It was observed that pennyroyal had positive effects on performance in the quails ($P < 0.05$). While the BW, BWG and FC values in the groups with pennyroyal supplementations tended to increase compared to the control group, the FCR had a significant decrease in the group with the supplementation of pennyroyal by 0.2% (Table 2). It was determined that pennyroyal did not have any effects on the post-slaughtering carcass yield and weights of the internal organs ($P < 0.05$) (Table 3).

It was observed that pennyroyal did not have any statistical effects on the color of the breast meat (L^* , a^* , b^*) and pH value ($P > 0.05$) (Table 4).

Table 2. The effect of Pennyroyal (*M. pulegium L.*) on performance parameters.

| | BW (g) | | | | |
|------------|--------------------------|---------------------------|--------------------------|----------|-------|
| | Groups | | | P values | |
| | Control | P1 | P2 | L | Q |
| d 7 | 29.20±0.73 | 29.00±0.17 | 28.98±1.39 | 0.982 | 0.939 |
| d 21 | 116.05±0.59 | 117.70±1.46 | 118.70±1.69 | 0.185 | 0.848 |
| d 35 | 180.81±1.97 ^b | 186.05±1.45 ^a | 188.88±1.18 ^a | 0.003 | 0.542 |
| | BWG (g/quails) | | | | |
| | Groups | | | P values | |
| | Control | P1 | P2 | L | Q |
| d 7 to 21 | 86.85±1.01 | 88.69±1.40 | 89.73±2.00 | 0.207 | 0.832 |
| d 21 to 35 | 64.76±1.84 ^b | 68.36±1.07 ^{ab} | 70.17±0.91 ^a | 0.014 | 0.598 |
| d 7 to 35 | 151.61±1.45 ^b | 157.05±1.35 ^a | 159.90±1.78 ^a | 0.003 | 0.506 |
| | FC (g) | | | | |
| | Groups | | | P values | |
| | Control | P1 | P2 | L | Q |
| d 7 to 21 | 276.96±3.23 | 279.23±4.50 | 282.94±3.45 | 0.272 | 0.877 |
| d 21 to 35 | 259.03±4.46 | 265.00±3.00 | 268.35±2.45 | 0.064 | 0.756 |
| d 7 to 35 | 535.99±3.87 ^b | 544.23±3.64 ^{ab} | 551.29±3.26 ^a | 0.004 | 0.895 |
| | FCR (g/g) | | | | |
| | Groups | | | P values | |
| | Control | P1 | P2 | L | Q |
| d 7 to 21 | 3.19±0.03 | 3.15±0.05 | 3.15±0.02 | 0.461 | 0.572 |
| d 21 to 35 | 4.00±0.04 ^a | 3.88±0.06 ^{ab} | 3.82±0.03 ^b | 0.008 | 0.540 |
| d 7 to 35 | 3.53±0.01 ^a | 3.47±0.02 ^{ab} | 3.45±0.01 ^b | 0.001 | 0.172 |

P1: 0.1% Pennyroyal, P2: 0.2% Pennyroyal, BW: Body weight, BWG: Body weight gain, FC: Feed consumption, FCR: Feed conversion rate, L: Linear, Q: Quadratic

^{a,b} Values within a column with different superscripts differ significantly at $P < 0.05$.

Table 3. The effect of Pennyroyal (*M. pulegium L.*) on carcass parameters.

| | Groups | | | P values | |
|-------------------|------------|------------|------------|----------|-------|
| | Control | P1 | P2 | L | Q |
| Carcass yield (%) | 73.16±0.19 | 73.31±0.11 | 73.34±0.12 | 0.380 | 0.730 |
| Heart (g) | 1.60±0.03 | 1.61±0.06 | 1.47±0.06 | 0.839 | 0.124 |
| Liver (g) | 3.31±0.43 | 2.65±0.19 | 3.41±0.41 | 0.130 | 0.542 |
| Gizzard (g) | 2.62±0.20 | 2.26±0.18 | 2.70±0.13 | 0.738 | 0.639 |

P1: 0.1% Pennyroyal, P2: 0.2% Pennyroyal, L: Linear, Q: Quadratic.

Table 4. The effect of Pennyroyal (*M. pulegium L.*) on meat color and pH.

| | Groups | | | P values | |
|----|------------|------------|------------|----------|-------|
| | Control | P1 | P2 | L | Q |
| L* | 45.51±1.23 | 45.96±1.79 | 45.29±1.35 | 0.947 | 0.845 |
| a* | 11.60±0.44 | 11.23±0.41 | 11.41±0.17 | 0.807 | 0.694 |
| b* | 4.85±0.19 | 4.75±0.14 | 4.72±0.17 | 0.725 | 0.911 |
| pH | 5.48±0.07 | 5.50±0.03 | 5.50±0.03 | 0.892 | 0.875 |

P1: 0.1% Pennyroyal, P2: 0.2% Pennyroyal, L *: Brightness, a *: Redness, b *: Yellowness, L: Linear, Q: Quadratic.

Discussion and Conclusion

The supplementation of pennyroyal on quail rations had a positive effect on performance. In the study, it was determined that the supplementation of pennyroyal in increasing levels had a positive effect on the BW, BWG and FC values, compared to the control group. It was determined that the best FCR was obtained in the group (2.86) with supplementation of 0.2% of pennyroyal. The BW and BWG results obtained were in accordance with the results of the study, in which pennyroyal was used as a supporting growth agent (33, 35). The results of the current study also had similarities with the studies on the effects of different *Mentha* species (*M. Longifolia* and *M. Piperita*) on BW and BWG in the poultry (3, 4). In the broiler study, in which the pennyroyal, essential oil, probiotics and antibiotics were used in different levels, the best performance values were obtained in the group with the addition of pennyroyal, thereby affecting the BWG and FCR significantly. The results of the study suggested that pennyroyal could be used as an alternative to antibiotics in poultry (2). It is believed that the phenolic compounds in pennyroyal and other *Mentha* species decreased the presence of pathogenic microorganisms in the intestines of the poultry, thereby showing an antimicrobial effect as well as affecting the growth performance (21, 30). The fact that pennyroyal increases feed consumption due to its appetizing effect has been mentioned in the literature (21). Similarly, there have been studies, in which the *Mentha* species increased feed consumption (3, 4).

Contrary to these studies, there have been studies reporting that the *Mentha* species do not affect the performance values (BW, BWG and FC) in the poultry (15, 19, 34). Erhan et al. (17) found that pennyroyal did not affect the body weight increase in broiler chickens, and it decreased the feed consumption, thereby improving the feed conversion rate. Ghalamkari et al. (20) stated that the addition of antibiotics and pennyroyal did not cause a difference in terms of the performance values of the broiler chickens, Arjomandi et al. (8) found that the pennyroyal mixed with probiotics decreased the feed consumption in laying hens, and Aydin and Bolukbasi (10) reported that the addition of pennyroyal in increasing

levels (32.5, 65 and 130 mg/kg) did not affect the feed consumption in laying hens, which were not in compliance. It is believed that these differences might have stemmed from the region where the plant was grown, the harvest season, the breed of animal used, the dosage used and feeding conditions.

It was determined that the addition of pennyroyal did not affect the weights of carcass and internal organs (heart, liver and gizzard) in quails. The results of the present study were following the results of some other studies (1, 21, 31). Similarly, Nobakht et al. (33) reported that pennyroyal did not have any effects on the carcass yield or the weights of the internal organs. On the contrary, Shamlo et al. (39) stated that the pennyroyal extract (0. 50, 100 and 150 ppm) increased the carcass weight, carcass yield and the weight of the heart; however, it did not have any effects on the weights of the liver and gizzard. Along with this study, there are other studies in the literature stating that the carcass parameters and internal organ weights changed due to the addition of pennyroyal and aromatic plants together or separately (4, 9, 38). It was stated that pennyroyal increased digestive enzymes in the poultry, which had a positive effect on the pancreas, and could positively affect the carcass parameters particularly by the increase of the amino-acid absorption (6). The differences observed between studies might have resulted from the diversity of aromatic plant used, the interactions between additives, dosage, and differences in animals.

The unique color of the product is another significant criterion for consumers to choose any product. This color should not change during the time it remains on the shelf. The oxidation of the myoglobin and hemoglobin pigments is useful in the changing of the natural color of the meat (23). In the study, no pennyroyal-related effects were observed on the color intensities (L*, a*, b*) and pH values of the breast meat of the quails ($P>0.05$). The current study is in accordance with the studies, in which aromatic plants and their essential oil derivatives were used, and had no effects on the color and pH values of the meat (12, 24, 27, 37).

There are also studies determining that pennyroyal and different aromatic plants had effects on the color and pH of the meat (28, 29). It was determined that *Mentha*

piperita affected the a^* and b^* of the meat color intensities; however, it did not affect the L^* and pH in the quails (7). In their study on the effect of thyme and pennyroyal on the broiler meat color and PH value in the broilers, Pirmohammadi et al. (36) found that the pennyroyal affected the L^* value; however, it did not have any effects on the a^* , b^* and pH values compared to the control group. They also stated that thyme and pennyroyal increased the pH when used combined ($P < 0.05$).

As a result, it was concluded that the pennyroyal (*Mentha pulegium L.*) significantly affected the growth performance without changing the carcass, internal organs, or the meat quality (color, pH) in the Japanese quails.

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Ethical Statement

This study was approved by the Kafkas University Animal Experiments Local Ethics Committee (KAÜ-HADYEK/2020-008).

Conflict of Interest

The authors declared that there is no conflict of interest.

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