

## Effects of Dietary Coffee Ground Supplementation on Carcass Traits, Meat Quality, and Faecal Pathogenic Bacterial Populations in Japanese Quails (*Coturnix coturnix japonica*)

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### ABSTRACT

This study aimed to investigate the effects of dietary coffee ground supplementation on the carcass traits, meat quality, and faecal pathogenic bacteria of Japanese quails. A completely randomised design (CRD) was employed using 180 nine-week-old male quails, which were randomly assigned to three treatment groups: a control diet (T1) and control diets supplemented with 10 g/kg (T2) and 20 g/kg (T3) of coffee grounds. Each treatment consisted of five replicates with 12 birds per replicate, all maintained on an *ad libitum* feeding and watering regimen. At the end of the experiment, samples were taken from each treatment for evaluation of carcass traits and meat quality, and samples of faeces were analysed for bacterial populations. The findings revealed that the dietary treatment of 10 and 20 g/kg of coffee grounds significantly ( $P < 0.05$ ) increased carcass traits. The percentage of commercial cuts and internal organs was favourably enhanced. Dry matter and crude protein content of breast meat were also increased ( $P < 0.05$ ) when the coffee grounds were fed. When fed, the breast meat dry matter and crude protein content were increased, and the yellowness ( $b^*$ ) of the breast meat and skin improved at 0 and 24 hours post-slaughter. Notably, the 10 and 20 g/kg of coffee ground diets caused a significant ( $P < 0.01$ ) decrease of *E. coli* and *Salmonella* spp. in their faeces.

In conclusion, dietary supplementation with 10-20 g/kg of coffee grounds in 15-week-old Japanese quails enhanced carcass yield, improved meat quality, and exerted potent antimicrobial effects by suppressing faecal pathogenic bacteria.

### ARTICLE INFO

#### Article history:

Received: 27 July 2025

Accepted: 22 May 2026

Published: 12 June 2026

DOI: <https://doi.org/10.47836/pjtas.49.3.09>

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**Keywords:** coffee ground, carcass traits, Japanese quails, meat quality, pathogenic bacteria

## INTRODUCTION

As the world's population is growing, the need for greater yield in agriculture and animal husbandry to ensure food supply is increasing. In this regard, the increasing demand for high-quality animal protein has highlighted the importance of developing more environmentally friendly and economically viable poultry production systems, especially for Japanese quails (*Coturnix coturnix japonica*). Japanese quails are very important for their fast growth rates, early sexual maturity, large egg production and high feed efficiency. Moreover, they are easily adaptable to various husbandry conditions; have low production costs; are easy to manage; require very little space; and are suitable for sustainable poultry farming (Batool et al., 2021; Mizutani, 2003; Vali, 2008). All the above benefits notwithstanding, the poultry industry is beset with huge challenges, such as the increasing production costs of animal feeds and the fear of antimicrobial resistance. The universal use of antibiotics as growth promoters has resulted in the emergence of pathogens that are cross-resistant to antibiotics and residues in animal tissues, which present significant health risks to animals and humans. The use of antibiotics as growth promoters has sparked continuous research on natural alternatives to improve animal performance and product quality after the European Union (EU) banned their use as growth promoters in 2006 (Castanon, 2007). At the same time, allocation of agricultural resources for animal feed directly affects the production of human food, hence there is a concern about long-term food security. These factors all call for a need to look at alternative and sustainable feedstuff sources. In recent years, research has been directed to the use of pharmacologically active compounds from medicinal plants.

Research has shown that different herbs, when fed to animals, can enhance animal performance to a great extent (Posan et al., 2023), and combinations of multiple medicinal plants in poultry feed can facilitate growth and strengthen the immune system (Phromnoi et al., 2025). Coffee is a major crop globally and produces several useful products, but it produces a significant amount of waste, which can be an environmental problem. Coffee grounds are a major organic byproduct of the coffee processing that is not used. If not properly handled, it is a source of environmental pollution (Ismail et al., 2014; Nillian et al., 2020). Consequently, it is essential to take environmental protection measures to find alternative applications for such waste materials as a matter of great urgency. Arabica beans are the more delicate flavoured beans, which account for a significant amount of the spent coffee grounds (SCG) produced (Ho et al., 2012; Solange et al., 2011).

Spent coffee grounds (SCG), one of the major byproducts of coffee processing, are a promising and sustainable feed raw material for animal production, especially in terms of environmental and economic benefits. In the global coffee industry, about 70% of the total consumption is *Coffea Arabica*, and 30% is *Coffea Canephora* (Robusta variety), producing millions of tonnes of waste every year (DaMatta et al., 2007; Marcel et al., 2011).

SCG contain various bioactive compounds with potential health benefits for the animal, such as polyphenols, caffeine, vitamins, fibre, and protein, which can improve nutrient utilisation and animal health (Ho et al., 2012; Tajik et al., 2017). Notably, SCG is rich in chlorogenic acids and other active substances with well-established powerful antioxidant and antibacterial activities (Bertrand et al., 2012).

Additionally, coffee contains biologically active fatty acids and triglycerides that have proven anti-inflammatory, anti-bacterial, anti-cancer, anti-diabetic and anti-atherosclerotic properties (Al-Asmari et al., 2020). Incorporation of SCG into animal feed, especially for Japanese quails, can enhance the carcass properties and meat quality, along with gut health. Coffee polyphenols, such as phenolic acids and flavonoids, are known to have beneficial effects on lipid metabolism, antimicrobial activity and antioxidant processes in animal muscle tissues, which positively influence meat quality.

Murwani et al. (2024) also reported that phytochemical compounds, such as those found in SCG, can reduce intestinal inflammation and enhance the productive performance of Japanese quails. In addition to their nutritional benefits, the incorporation of SCG into animal feed contributes to environmental sustainability by valorising agricultural waste materials. This strategy provides a practical and eco-friendly alternative to conventional growth promoters in animal production, while also meeting the growing consumer demand for sustainable poultry products. This study evaluated the effects of dietary coffee ground supplementation on the carcass traits, meat quality, and faecal pathogenic bacteria populations of Japanese quails. We hypothesised that the dietary inclusion of coffee grounds would enhance carcass traits, improve meat quality, and beneficially modulate faecal microbial populations. Ultimately, this research aims to support the utilisation of coffee grounds as a sustainable feed ingredient and an eco-friendly alternative to conventional growth promoters in poultry production.

## **MATERIALS AND METHODS**

### **Code of Conduct for Laboratory Animals**

The current investigation was reviewed by the Institutional Animal Care and Committee, Princess of Naradhiwas University, Thailand, No. PNU.AE-2024/01.009.

### **Coffee Grounds Preparation**

Coffee grounds were collected from Muang Narathiwat district, Narathiwat province. After collection, the samples were sun-dried for two days and subsequently dried in a hot-air oven at 60-65 °C for an additional day. The dried coffee grounds were then finely ground using an electric grinder and passed through a 1-mm mesh sieve to obtain a uniform particle size. The processed coffee ground powder was stored in sealed glass containers and refrigerated at 4°C until use in the experiment.

## Experimental Animal and Management

A 6-week feeding trial was carried out on 180 nine-week-old male Japanese quails (9-15 weeks) in a completely randomised design (CRD). The birds that were selected for uniform starting body weight were randomly assigned to three different dietary treatments. Five replicates were used for each treatment, with 12 quails in each replicate, making a total of 60 quails per treatment group. Throughout the experiment period, quails were provided with *ad libitum* access to a feed and drinking water. All birds were kept in an open system, and good hygiene and ventilation were maintained. In this experiment, the experimental diet was a commercial quail starter feed fed during the first rearing period (1-4 weeks). The guaranteed nutritional composition of this feed was 22% crude protein, at least 3% crude fat and a maximum of 13% moisture. Table 1 shows the complete ration composition and detailed nutritional analysis of this commercial quail feed.

## Carcass Traits

At the end of the experiment (15 weeks of age), carcass traits were evaluated according to the procedures described by Moreng and Avens (1985). Ten quails from each treatment group (two birds per replicate) were randomly selected, weighed, and manually slaughtered after being fasted for 12 hours to determine carcass traits, including dressing, eviscerated weight, carcass, gizzard, liver, heart, spleen, and other total edible parts, which were weighed and calculated for the quail carcass percentage.

Table 1  
*Ration composition and nutritional content of the quail commercial feed*

Composition	Content (%)
Moisture	13
Crude Protein	22
Crude Fat	3
Ash	13
Calcium	3
Phosphorus	0.62

## Meat Quality

Meat quality assessment was performed on the *Pectoralis major* muscle, utilising two boneless, skinless breast fillets collected per replicate. Measurements for pH, colour, and drip loss values were executed in accordance with the methodologies described by Wattanachant (2003) and Wattanachant et al. (2004).

For proximate composition analysis, two breast meat samples per replicate were collected. The percentages of crude protein, crude fat, and dry matter were subsequently determined using the methodology outlined by the Association of Official Analytical Chemists (AOAC) (2000).

pH value analysis: a pH meter (Seven2Go™, Mettler-Toledo, Switzerland) was used to measure the pH of breast meat. The probe METTLER TOLEDO Inlab® 413 IP67 was used and calibrated at 4.0 and 7.0.

For colour analysis, raw, undamaged breast meat fillets (two birds per replicate) were used to measure surface colour using the Complete International Commission on Illumination (CIE) system. Colour parameters measured included lightness ( $L^*$ ), redness ( $a^*$ ), and yellowness ( $b^*$ ). The  $L^*$  value represents whiteness (0) to darkness (100); the  $a^*$  expression ranges from redness (+) to greenness (-); and the  $b^*$  observation denotes yellowness (+) to blueness (-). Meat colour measurements were performed in triplicate on the upper, middle, and lower surfaces of each sample using a Colour Reader CR-13 (Konica Minolta Sensing, Inc., Japan).

To determine water-holding capacity via drip loss, breast meat samples from two quails per replicate were analysed. Fillets were cut into three strips, each measuring 1.5 cm wide, 3.0 cm long, and 0.5 cm thick. Each strip was then individually weighed, placed in a sealed polyethylene bag, and kept at a chilling temperature between 1 and 4 °C for 24 hours. After this period, the samples were reweighed, and the drip loss was calculated as the percentage of weight reduction relative to the initial sample weight.

### Faecal Pathogenic Bacteria

At the end of the experimental period, five faecal samples were collected from quails within each treatment group to quantify pathogenic bacterial populations. The analyses for *Escherichia coli* (*E. coli*) and *Salmonella* spp. were conducted using the methodology prescribed by the United States Food and Drug Administration (FDA) (2020).

In the *Escherichia coli* (*E. coli*) population assay, preparing 25 grams of faecal sample, 0.1% peptone water buffer was added to 225 millilitres. They were then homogenised using a stomacher. After that, the samples were diluted in 0.85% normal saline solution to achieve the proper concentrations (ranging from  $10^{-1}$  to  $10^{-5}$ ). The diluted samples were placed on the surface of the Chrom ID™ Coli agar plates by the spread plate technique. The plates were incubated for 24 hours at 37 °C. After incubation, the plates were evaluated for the *E. coli* population. The typical colony morphology was observed and recorded, and the number of pink-purple colonies was counted.

For the *Salmonella* spp. population assay, preparing 25 grams of faecal sample, 0.1% peptone water buffer was added to 225 millilitres for a pre-enrichment medium. They were then homogenised using a stomacher. Then, samples were diluted in 0.85% normal saline solution to make proper concentrations at dilution levels ranging from  $10^{-1}$  to  $10^{-5}$ . The diluted samples were placed on the surface of the xylose lysine deoxycholate (XLD) agar plates. Spread the sample evenly on the agar surface using the spread plate technique. The plates were incubated for 24 hours at 37 °C. Following incubation, the plates were evaluated for *Salmonella* spp. population, and the colonies were observed and counted. Colonies will appear as round, medium-sized, and red with a black centre due to hydrogen sulphide production. The colonies' surrounding agar will continue to be red.

## Statistical Analysis

All experimental data were subjected to a one-way analysis of variance (ANOVA). When significant differences were detected, treatment means were compared using Duncan's Multiple Range Test (DMRT). Data distribution, normality, and descriptive statistics were verified utilising the UNIVARIATE procedure in SAS software (SAS, 2017).

## RESULT AND DISCUSSION

### Carcass Traits

The present study assessed the effects of dietary supplementation with coffee grounds, at concentrations of 0, 10, and 20 g/kg, on the carcass traits of 15-week-old Japanese quails. The results revealed that supplementing the diet of Japanese quails with coffee grounds, particularly at 10 g/kg (T2) and 20 g/kg (T3), leads to significant quantitative improvements in growth and carcass yield compared to the control group (Table 2). Specifically, the T2 group showed a 7.02% increase in live body weight (171.95 g vs. 160.67 g) and an 8.79% increase in dressing weight (152.84 g vs. 140.49 g). Eviscerated weight also increased by 7.57% in the T2 group. These improvements indicate that the moderate levels of coffee ground supplementation improved overall development of the birds and a significant increase in actual carcass percentage was noted, which being 88.87% in the coffee ground as compared to 87.44% in the control. In addition, the researchers found that there were significant increases in high-value meat amounts and some physiological parameters without any adverse effects on proportions of abdominal fat or internal organs. The percentage of breast and skin weight increased by 9.41% in the T2 group (28.38% vs. 25.94%), while thigh and drumstick weights saw improvements of 14.03% (12.03% vs. 10.55%) and 10.55% (8.28% vs. 7.49%), respectively. Lastly, there were greater weights of testes in quails fed the 10 g/kg and 20 g/kg coffee grounds groups compared to the control group. However, the supplementation did not significantly affect the percentage of the liver, heart and gizzard, suggesting that it does not affect the relative size of metabolic organs, but rather muscle and skeletal growth. The present study shows that feeding coffee grounds in the diet of Japanese quails considerably improves the carcass characteristics. It is noted that dressing and eviscerated weights improved, while the percent of the carcass, breast meat, thigh, drumstick, wing, shank, spleen and testis improved. The improvements are said to be due to the wide range of bioactive compounds found in coffee grounds that have strong antioxidant properties. This finding aligns with some previous research. For instance, Ashour et al. (2020) reported that while dietary coffee grounds had no effect on carcass, dressing, or gilet percentages, different levels of green coffee powder significantly enhanced liver, abdominal fat, intestinal length, and lymphoid organ percentages (spleen and Bursa of Fabricius). Notably, supplementation with 1.25 and 2.50 g/kg of green coffee powder significantly increased ( $P < 0.05$ ) liver percentage.

Table 2

*Effects of coffee ground supplementation in the diet on carcass traits of 15-week-old Japanese quails*

Carcass Traits	Experimental Groups			SEM	P-value
	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>		
Live Body Weight (g)	160.67 <sup>b</sup>	171.95 <sup>a</sup>	170.09 <sup>a</sup>	0.66	0.0013
Dressing Weight	140.49 <sup>b</sup>	152.84 <sup>a</sup>	150.29 <sup>a</sup>	0.64	0.0005
Eviscerated Weight (g) <sup>1</sup>	129.38 <sup>b</sup>	139.18 <sup>a</sup>	138.38 <sup>a</sup>	0.54	0.0005
Carcass Percentage (%) <sup>2</sup>	87.44 <sup>b</sup>	88.87 <sup>a</sup>	88.34 <sup>ab</sup>	0.11	0.0256
Gizzard (%) <sup>3</sup>	1.23	1.31	1.24	0.01	0.3454
Heart (%)	0.98	0.95	1.03	0.01	0.3679
Liver (%)	1.43	1.45	1.44	0.01	0.9543
Spleen (%)	0.08 <sup>b</sup>	0.12 <sup>a</sup>	0.11 <sup>a</sup>	0.003	0.0077
Head and Neck Weight <sup>3</sup> (%)	10.59	11.05	11.17	0.07	0.1908
Breast and Skin Weight <sup>3</sup> (%)	25.94 <sup>b</sup>	28.38 <sup>a</sup>	28.64 <sup>a</sup>	0.09	<0.0001
Thigh Weight <sup>3</sup> (%)	10.55 <sup>b</sup>	12.03 <sup>a</sup>	11.58 <sup>a</sup>	0.07	0.0003
Drumstick Weight <sup>3</sup> (%)	7.49 <sup>b</sup>	8.28 <sup>a</sup>	8.85 <sup>a</sup>	0.07	0.0003
Wing Weight <sup>3</sup> (%)	6.24	6.70	6.68	0.05	0.0838
Shank Weight <sup>3</sup> (%)	2.14	2.23	2.26	0.03	0.5009
Abdominal Fat Weight <sup>3</sup> (%)	1.16	1.06	0.94	0.05	0.6527
Skeleton Weight <sup>3</sup> (%)	20.98	21.34	21.10	0.09	0.7160
Other Organs:					
Testis (%)	2.83 <sup>b</sup>	3.85 <sup>a</sup>	4.02 <sup>a</sup>	0.06	0.0002

Note. T1: Control group (commercial feed without coffee grounds), T2: Commercial feed supplemented with 10 g/kg of coffee grounds, and T3: Commercial feed supplemented with 20 g/kg of coffee grounds

<sup>ab</sup> Means with different letters in the same row show significant differences (P<0.05)

<sup>1</sup>SEM: Standard error of the mean

Live body weight refers to the weight of broiler chickens after a 12-hour fasting period

<sup>1</sup>Carcass weight refers to the weight of the chicken after the removal of internal organs

<sup>2</sup>Calculate as a percentage of live body weight

<sup>3</sup>Calculate as a percentage of carcass weight

Green coffee powder at 2.50 g/kg reduced abdominal fat, likely due to antioxidants decreasing malic enzyme activity, thus depressing abdominal fat deposition.

Furthermore, 1.25 and 2.50 g/kg of green coffee powder resulted in a higher percentage of spleen, and 1.25 g/kg produced a better percentage of Bursa of Fabricius and meat quality for broilers. Likewise, Hosseini-Vashan et al. (2023) confirmed that the addition of 0.9% green coffee powder to broiler diets significantly (P<0.05) improved the carcass traits such as breast, thigh and Bursa of Fabricius. Additionally, the tested levels of green coffee powder, 0.3%, 0.6%, 0.9%, 1.2%, and 1.5%, significantly decreased abdominal fat in broiler chickens compared to the control group. In line with these findings, crude coffee extract was found to significantly (P<0.05) contribute to the increase in carcass percentage,

breast meat, thigh and fillet of broiler chickens, but reduced the abdominal fat compared to control chick (Ghanima et al., 2021). The bioactive compounds present in coffee are responsible for these improvements in meat quality, indicating that the use of coffee extract as a feed additive in poultry production can improve the quality of meat produced. There has been further support for the beneficial effects of coffee-derived compounds on the productive performance of poultry, by Yachai et al. (2022), who found that feeding Thai native chickens with 2.0 g/kg coffee silver skin significantly ( $P < 0.05$ ) enhanced both productive performance and antioxidant content in the breast muscle, as well as improving the morphology of the intestines.

Conversely, other studies like Klompanya et al. (2024) found no significant difference in broiler carcass percentage, breast meat, and thigh weights when the spent coffee grounds were added at 5% and 10% concentration. On the other hand, 5% and 10% of coffee grounds significantly affected liver weight ( $P < 0.05$ ) and 10% significantly decreased abdominal fat and improved wing percentage, which may be attributed to the metabolism of specific ingredients of the coffee ground. The findings of the present study confirm the effectiveness of the use of coffee grounds to improve the carcass properties of the Japanese quail, which probably resulted from its rich and varied active substances. The essential compounds found in coffee are carbohydrates, minerals, caffeine, chlorogenic acid, proteins and lipids, all of which have important pharmacological and biological properties, such as protection against oxidative damage in animal cells, and are known to have antioxidant and free radical scavenging properties (Briandet et al., 1996). The antioxidant activity of phenolic compounds and flavanols has also been identified by other scientists as an important property that can help to prevent damage to cells caused by free radicals (Simoh et al., 2018). In keeping with the above mechanistic explanations, Ko et al. (2012) also found that dried coffee meal had antioxidant activity *in vitro*, and it was a natural source of antioxidants without affecting broiler growth performance. In the small intestine and liver, glutamate thiosemicarbazone is not used as a food source by birds, and therefore, it did not affect the concentrations of antioxidant defence enzymes (glutathione peroxidase, superoxide dismutase, and glutathione S-transferase) or the concentration of the antioxidant defence molecule glutathione. However, in the liver, birds fed 0.5% dry coffee meal showed significantly reduced ( $P < 0.05$ ) lipid peroxidation when compared to the control. Additionally, Vargas-Sánchez et al. (2019) found that the use of natural ingredients such as medicinal herbs, plants, vegetables and spices in Japanese quail diets can improve the quality of the carcass and meat due to their antioxidant effect.

However, this effect is dependent on the concentration, type, and conformation of the molecules present, which can influence the absorption and metabolism of active compounds, enabling or disabling their antioxidant or antibacterial functions. It is also important to consider that high levels of some natural ingredients may adversely affect

quail carcass and meat. Thus, the active ingredients in coffee grounds appear to enhance the immune system response, reduce inflammation, and promote overall animal health, thereby improving the carcass quality of Japanese quails. This study elucidated that coffee grounds can serve as a viable feed ingredient for animal production, with supplementation at 10 and 20 g/kg demonstrating an improvement in the carcass characteristics of 15-week-old Japanese quails.

## **Meat Quality**

### ***Proximate Composition***

Coffee grounds significantly affected ( $P < 0.05$ ) the proximate composition of Japanese quail breast meat, as shown in Table 3. Birds fed either 10 g/kg (T2) or 20 g/kg (T3) of coffee ground showed a significant increase in crude protein and dry matter content when compared with the control birds. The quantitative improvement regarding crude protein was found to be in the range of 21.36 to 22.29% in the T3 group, which was 4.35% higher than in the control group. Similarly, dry matter increased from 24.03% to 24.82% in Treatment T3, which resulted in an increase in nutrient density by 3.29%. At the same time, crude fat content in the breast meat was significantly lowered in the presence of the inclusion of coffee grounds (from 2.23% in the control group to 2.11% in T2 and 2.03% in T3). The results of the study are that the meat quality of quails was improved by the addition of the active ingredient contained in coffee grounds, which led to an increase in crude protein and crude dry matter percentages and a decrease in crude fat percentage in the breast meat.

The present investigation revealed that the crude protein, crude fat, ash and moisture content in coffee grounds were 14.20%, 3.50%, 2.50% and 8.0%, respectively. These results are consistent with the nutritional content of coffee grounds. Arabica spent coffee grounds are rich in various nutritional components such as nitrogen-free extract, protein, fat, fibre, ash, and gross energy, which were reported by Klompanya et al. (2024) as 48%, 12.86%, 3.70%, 17.34%, 1.36%, and 5,184.4 kcal/kg, respectively. Moreover, they reported levels of calcium (0.18%) and phosphorus (0.05%). Likewise, Kourmentza et al. 2018 reported that coffee grounds are 13.5% protein, 2.3% fat and 1.3% ash. The increased proximate composition of the breast meat in Japanese quails may be partly due to these active nutritional constituents present in the grounds of coffee.

## **Meat Quality**

### ***pH and Drip Loss Values***

The effects of coffee ground supplementation in the diet on the meat quality of the breast meat of Japanese quail at 15 weeks of age are presented in Tables 4 and 5. The results indicated that adding the diet with coffee grounds at 10 g/kg (T2) and 20 g/kg (T3) did not result in statistically significant changes to the pH levels or water-holding capacity

(drip loss value) of the breast meat. Immediately after slaughter ( $pH_0$ ), the values remained stable across groups, ranging from 6.21 to 6.26. After 24 hours ( $pH_{24}$ ), pH values slightly decreased to a range of 6.09 to 6.12 but remained consistent between the control and supplemented groups. Similarly, drip loss—a key indicator of meat moisture retention—showed no significant difference, with the control group at 3.15% compared to 3.03% in the T3 group, representing a negligible reduction of approximately 3.8%.

Table 3

*Effects of coffee ground supplementation in the diet on proximate composition in breast meat of Japanese quail at 15 weeks of age*

Experimental groups	Proximate Composition in Breast Meat (%)		
	Crude Protein	Crude Fat	Dry Matter
T1	21.36 <sup>b</sup>	2.23 <sup>a</sup>	24.03 <sup>b</sup>
T2	22.16 <sup>a</sup>	2.11 <sup>b</sup>	24.75 <sup>a</sup>
T3	22.29 <sup>a</sup>	2.03 <sup>b</sup>	24.82 <sup>a</sup>
SEM	0.04	0.01	0.04
P-value	<0.0001	0.0004	0.0007

*Note.* T1: Control group (commercial feed without coffee grounds), T2: Commercial feed supplemented with 10 g/kg of coffee grounds, and T3: Commercial feed supplemented with 20 g/kg of coffee grounds

<sup>a,b</sup> In the same column, means with different letters differ significantly ( $P < 0.05$ )

<sup>1</sup>SEM: Standard error of the mean

Table 4

*Effects of coffee ground supplementation in diet on pH and colour values of Japanese quail breast meat at temperature 0-4 °C for 0 hours ( $pH_0$ ), within 45 minutes after slaughter*

Item	Experimental Groups			SEM	P-value
	T1	T2	T3		
pH Value ( $pH_0$ ) of Breast Meat	6.26	6.21	6.24	0.01	0.5149
Colour Value of Breast Meat:					
Lightness ( $L^*$ )	44.24	44.34	44.50	0.08	0.7567
Redness ( $a^*$ )	11.50	12.21	12.14	0.09	0.2024
Yellowness ( $b^*$ )	10.34 <sup>b</sup>	10.85 <sup>ab</sup>	10.96 <sup>a</sup>	0.06	0.0464
Colour Value of Skin:					
Lightness ( $L^*$ )	45.35	45.39	45.34	0.07	0.9844
Redness ( $a^*$ )	10.32	10.52	10.82	0.07	0.3172
Yellowness ( $b^*$ )	11.05 <sup>b</sup>	11.87 <sup>a</sup>	11.98 <sup>a</sup>	0.08	0.0221

*Note.* T1: Control group (commercial feed without coffee grounds), T2: Commercial feed supplemented with 10 g/kg of coffee grounds, and T3: Commercial feed supplemented with 20 g/kg of coffee grounds

<sup>a,b</sup> Means with different letters in the same row show significant differences ( $P < 0.05$ )

<sup>1</sup>SEM: Standard error of the mean

$pH_0$  is the pH value within 45 minutes after slaughter

Table 5

*Effects of coffee ground supplementation in diet on pH, colour, and drip loss values of Japanese quail breast meat at temperature 0-4 °C for 24 hours (pH<sub>24</sub>) after slaughter*

Item	Experimental Groups				
	T1	T2	T3	SEM	P-value
pH Value (pH <sub>24</sub> ) of Breast Meat	6.09	6.10	6.12	0.01	0.7104
Colour Value of Breast Meat:					
Lightness (L*)	45.34	34.69	45.87	0.07	0.2833
Redness (a*)	12.51	12.83	12.73	0.06	0.5219
Yellowness (b*)	11.79 <sup>b</sup>	12.64 <sup>a</sup>	12.81 <sup>a</sup>	0.08	0.0184
Colour Value of Skin:					
Lightness (L*)	46.60	46.86	46.76	0.07	0.6992
Redness (a*)	11.57	11.90	11.75	0.06	0.4784
Yellowness (b*)	12.10 <sup>b</sup>	12.84 <sup>a</sup>	12.99 <sup>a</sup>	0.05	0.0012
Drip loss (%)	3.15	3.05	3.03	0.01	0.2114

Note: T1: Control group (commercial feed without coffee grounds), T2: Commercial feed supplemented with 10 g/kg of coffee grounds, and T3: Commercial feed supplemented with 20 g/kg of coffee grounds

<sup>a,b</sup> Means with different letters in the same row show significant differences (P<0.05)

<sup>1</sup>SEM: Standard error of the mean

pH<sub>24</sub> is the pH value measured 24 hours after slaughter

## Colour Value

Regarding the colour of breast meat, the current study found that the lightness (L\*) and redness (a\*) of the breast meat were unaffected by the diet; there was a significant quantitative increase in the yellowness (b\*). Within 45 minutes of slaughter, the yellowness value increased from 10.34 in the control (T1) to 10.96 in the T3 group, an improvement of roughly 6%. This trend became more pronounced 24 hours post-slaughter; the T2 (12.64) and T3 (12.81) groups showed significantly higher yellowness compared to the control (11.79). Specifically, the 20 g/kg supplementation (T3) resulted in an 8.6% increase in breast meat yellowness compared to the control group.

For the skin colour of breast meat, the most notable statistical improvements occurred in the colouration of the quail's skin, particularly regarding yellowness. In the initial 45-minute post-slaughter, skin yellowness rose from 11.05 (T1) to 11.98 (T3), a significant increase of approximately 8.4%. At 24 hours post-slaughter, this difference remained significant, with the T3 group reaching a value of 12.99 compared to the control group (12.10). This represents a 7.3% enhancement in skin yellowness for the T3 group. Additionally, the treatment had no significant impact on the lightness (L\*) or redness (a\*) of the meat or skin at any recorded interval, suggesting that coffee grounds specifically influence the yellow pigment profile without darkening the product.

Warriss (2000) reported typical chicken meat pH values of 6.4-7.0 at 0 hours and 5.9-6.0 at 24 hours post-slaughter. Other research indicates that Japanese quail breast meat pH values 24 hours post-slaughter range from 6.1 to 6.3, with breast meat generally having slightly lower values (Genchev et al., 2008). The ultimate pH of meat is primarily determined by muscle glycogen levels, which are significantly influenced by pre-slaughter stressors and locomotor activity (Genchev et al., 2008). As Sanchai (2000) explained, upon an animal's death, oxygen, glucose, and free fatty acid transport to muscles ceases, disrupting oxygen-dependent ATP production. The body then resorts to anaerobic glycolysis, breaking down stored glycogen to produce ATP and lactic acid, which subsequently lowers meat pH from an initial neutral level to a final range of 5.3-5.7 within 24 hours as glycogen is depleted. Genchev et al. (2008) also noted that Japanese quail meat is rich in essential amino acids, with breast meat typically containing 22.23-23.38% protein and 2.21-2.75% fat, and pectoral muscles having 21.68-22.39% free water. The various active ingredients in coffee grounds are responsible for the increase in the yellowness of breast meat and skin in Japanese quails fed coffee grounds. These results are somewhat contradicted by the study by Ashour et al. (2020), who reported that the supplementation of green coffee powder at 2.5 g/kg of broiler feed resulted in a significant increase in the lightness ( $L^*$ ) and redness ( $a^*$ ) of the broiler breast meat, which could be attributed to the reduction of fat and colour oxidation. It is well known that compounds obtained from coffee have beneficial effects. Coffee is also high in chlorogenic acid compounds and has been reported as having significant antioxidant and anti-inflammatory properties and other beneficial effects (Liang & Kitts, 2016). Coffee extract has been shown to possess high in vitro antioxidant activity in previous studies, making it a useful natural source of antioxidants, without negative effects on broiler growth performance. In addition, the relative weight of the thymus was significantly higher ( $P < 0.05$ ), and the blood albumin level was raised in birds fed 0.5% and 1.0% of dried coffee meal, while there was no significant difference in other blood constituents (glucose, triglycerides, and cholesterol) (Ko et al., 2012). Coffee meal (0.5%) did not affect the antioxidant defence system, such as the activities of superoxide dismutase, hepatic glutathione peroxidase, and glutathione S-transferase; however, in birds, there was a significant ( $P < 0.05$ ) decrease in hepatic lipid peroxidation with the addition of 0.5% coffee meal (Ko et al., 2012). The potential and benefits of natural plant extracts with antioxidant and antibacterial properties to preserve meat products (Tandale et al., 2024). Consistent with this, another researcher confirmed that coffee grounds could increase feed efficiency, increase nutrient digestion, promote antioxidant activity, and support immune response due to the presence of total phenolic compounds, such as chlorogenic acid, protocatechuic acid, and flavonoids, all known for their high antioxidant activity (Solange et al., 2011). These compounds likely contribute to the observed improvements in the physical characteristics of Japanese quail breast meat.

## Faecal Pathogenic Bacteria

The study investigated the effects of coffee ground supplementation in the diet on faecal pathogenic bacteria of 15-week-old Japanese quails. The results found that supplementing Japanese quail diets with coffee grounds significantly reduced the populations of *E. coli* and *Salmonella* spp. in their faeces, as shown in Table 6. In the control group (T1), *E. coli* levels were measured at 7.6181 log<sub>10</sub> CFU/g. When quails were fed a diet supplemented with 10 g/kg of coffee grounds (T2), the *E. coli* population dropped to 7.4815 log<sub>10</sub> CFU/g, representing a decrease of approximately 1.79%. Increasing the supplementation to 20 g/kg (T3) resulted in a slightly further reduction to 7.4668 log<sub>10</sub> CFU/g, a 1.99% improvement over the control. These reductions were statistically significant (P<0.0001).

The same trend was observed for *Salmonella* spp., which also showed a downward trend, indicating the antibacterial effects of the coffee ground additive. The log<sub>10</sub> CFU/g of the control group was 7.2392. The addition of 10 g/kg (T2) to the feed lowered this to 7.1353 log<sub>10</sub> CFU/g, which is a quantitative improvement of 1.44%. The higher dose of 20 g/kg (T3) was even more effective, resulting in 7.0708 log<sub>10</sub> CFU/g (a 2.33% reduction from the control group). From the current study, it was concluded that coffee ground supplementation is a highly consistent method for reducing the population of enteric pathogens in 15-week old Japanese quails.

This study confirmed previous research, which showed the antimicrobial activity of coffee by-products. The use of 2.5 g/kg of green coffee powder significantly reduced the levels of *Salmonella* spp., *E. coli*, yeasts and moulds, and *Enterococcus* spp., and it also increased the levels of the beneficial lactic acid bacteria (Ashour et al., 2020). Similarly, Yachai et al. (2022) reported that the cecum of Thai native chickens had a significant reduction in the populations of *Salmonella* sp. and *E. coli* with 2.0g/kg of coffee silver skin (P<0.05).

Table 6

*Effects of coffee ground supplementation in the diet on E. coli and Salmonella spp. populations in the faeces of 15-week-old Japanese quails*

Experimental groups	<i>E. coli</i> (log <sub>10</sub> CFU/g)	<i>Salmonella</i> spp. (log <sub>10</sub> CFU/g)
T1	7.6181 <sup>a</sup>	7.2392 <sup>a</sup>
T2	7.4815 <sup>b</sup>	7.1353 <sup>b</sup>
T3	7.4668 <sup>b</sup>	7.0708 <sup>b</sup>
SEM	0.006	0.007
P-value	<0.0001	<0.0001

*Note.* T1: Control group (commercial feed without coffee grounds), T2: Commercial feed supplemented with 10 g/kg of coffee grounds, and T3: Commercial feed supplemented with 20 g/kg of coffee grounds

<sup>a,b</sup> Means with different letters in the same column show significant differences (P<0.01)

<sup>1</sup>SEM: Standard error of the mean

Additionally, Ashour et al. (2020) reported that the extract of green coffee inhibited the growth of several bacterial strains with MICs of 100, 80, 80, 60, 60 and 50  $\mu\text{g/mL}$  in the case of *Pseudomonas aeruginosa* ATCC 27853, *Klebsiella pneumoniae* ATCC BAA-1705, *Escherichia coli* ATCC 43886, *Staphylococcus aureus* ATCC 25923, *Bacillus cereus* ATCC 14579 and *Bacillus subtilis* ATCC 23857, respectively, while MBCs were 150, 120, 120, 100, 100 and 80  $\mu\text{g/mL}$ , respectively. This could be attributed to the presence of different bioactive compounds of the coffee grounds that had high inhibitory potential on pathogenic bacteria growth. The antibacterial activity of the coffee grounds that was observed is probably due to the abundance of bioactive compounds in the coffee grounds. The coffee grounds are rich in total phenolics ( $55.4 \pm 1.11$  mg GAE  $\text{g}^{-1}$  extract) and flavonoids ( $14.2 \pm 0.42$  mg QE  $\text{g}^{-1}$  extract), which are all effective against microorganisms.

Additionally, coffee grounds are also rich in triglycerides, fatty acids, and antioxidant compounds. These fatty acids exhibit a wide range of biological activities, including anti-inflammatory, anticancer, antibacterial, antidiabetic, and anti-atherosclerotic effects (Al-Asmari et al., 2020). However, the mechanisms through which these phytochemicals act within the gastrointestinal tract remain unclear and require further investigation to identify the specific active compounds and their metabolites. Similarly, Sharma et al. (2022) reported that herbal compounds show strong therapeutic potential against infectious pathogens, particularly bacteria, emphasising the need for additional studies on the diverse phytochemical constituents of plants, their mechanisms of action, and possible synergistic interactions responsible for their antibacterial properties. The consistent reduction of pathogenic bacteria reported in this study demonstrated that coffee grounds could serve as a natural and sustainable feed ingredient for controlling microbial populations in livestock, providing a promising alternative to conventional antibiotics and harmful chemicals used in animal production.

## CONCLUSION

Dietary supplementation of coffee grounds at levels of 10 and 20 g/kg in Japanese quails significantly enhanced live body weight, carcass yield, and the relative proportions of valuable commercial cuts, including the breast, thigh, and drumstick. In addition, positive effects were observed on spleen and testes weights. Coffee ground supplementation also improved breast meat quality by increasing dry matter, crude protein content, and meat yellowness while concurrently reducing faecal populations of *E. coli* and *Salmonella* spp.

These findings indicated that coffee grounds can be effectively incorporated into commercial feed formulations as a cost-efficient and bioactive agro-industrial byproduct to enhance growth performance and meat quality. Furthermore, their antimicrobial properties suggest that coffee grounds may serve as a natural alternative for improving gut health in poultry production. Nevertheless, further studies are needed to determine optimal

supplementation levels, elucidate the underlying physiological mechanisms, and assess the long-term effects on animal health and environmental sustainability.

## CONFLICTS OF INTEREST

This research was conducted independently for non-profit purposes, and the author declares no conflicts of interest.

## ACKNOWLEDGEMENT

The author would like to thank the Faculty of Agriculture, Princess of Naradhiwas University, Thailand, for providing the experimental facilities, necessary equipment, and full support throughout this study.

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