

Impact of Pomegranate peel powder (*Punica granatum*) incorporation on growth performance, carcass characteristics and biochemical parameters in Japanese Quails (*Coturnix japonica*)

Impacto de la incorporación de cascara de Granada sobre el crecimiento productivo, características de la canal y parámetros bioquímicos en codornices japonesas

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ABSTRACT

The aim of the assay was to study the effect of pomegranate peel powder (PPP) as an alternative natural additive on growth performance, carcass characteristics, and biochemical parameters of Japanese quails. The experiment involved 208 unsexed Japanese quails' chicks initially fed a standard diet without PPP for the first week. Then, they were weighed and divided into four groups: one control group CTRL and three test groups receiving diets supplemented with 3%, 5%, and 7% PPP. Each group composed of four replicates of 13 quails. The results indicated that during the grower period, the group receiving 7% PPP showed higher feed intake (FI) ($P=0.029$) and feed conversion ratio (FCR) ($P=0.001$). However, body weight (BW) ($P<0.0001$), body weight gain (BWG) ($P=0.001$), and average daily gain (ADG) ($P=0.017$) were decreased. In contrast, during the finisher period, PPP supplementation did not significantly affect the final BW, BWG, or ADG ($P>0.05$). Notably, the groups receiving 5% and 7% PPP experienced a significant reduction in FI ($P=0.001$) and the 7% PPP group showed significant increases in proventriculus weight ($P=0.025$), relative intestine weight ($P=0.017$) and cecum length ($P<0.0001$). Furthermore, this group exhibited a noticeable decrease in albumin levels ($P<0.0001$) and an increase in GOT activity ($P=0.002$). In conclusion, PPP shows promising effects as a nutritional additive and natural growth promoter for Japanese quails. However, it is advisable to incorporate it after the grower period and to be cautious with higher doses due to potential toxicity risks.

Key words: Pomegranate Peel Powder; *Coturnix japonica*; feed formulation; growth performance; biochemical parameters

RESUMEN

El objetivo del estudio es evaluar el efecto del polvo de cáscara de granada (PCG) como un aditivo natural alternativo sobre el rendimiento productivo, las características de la canal y los parámetros bioquímicos en codornices japonesas. El experimento involucró a 208 polluelos de codorniz japonesa no sexados, alimentados inicialmente con una dieta estándar sin PCG durante los primeros siete días. Luego, fueron pesados y divididos en cuatro grupos: un grupo de control (CTRL) y tres grupos de prueba que recibieron dietas suplementadas con 3%, 5% y 7% de PCG. Cada grupo estaba compuesto por cuatro réplicas de 13 codornices cada una. Los resultados indicaron que durante el período de crecimiento, el grupo que recibió 7% de PCG mostró un mayor consumo de alimento (FI) ($P=0,029$) y un mayor índice de conversión alimenticia (FCR) ($P=0,001$). Sin embargo, se encontró una disminución en el peso corporal (BW) ($P<0,0001$), la ganancia de peso corporal (BWG) ($P=0,001$) y la ganancia diaria promedio (ADG) ($P=0,017$). En cambio, durante el período de acabado, la suplementación con PCG no tuvo un impacto significativo en el BW final, BWG o ADG ($P>0,05$). Cabe destacar que los grupos que recibieron 5% y 7% de PCG experimentaron una reducción significativa en el FI ($P=0,001$). Además, el grupo que recibió 7% de PCG mostró aumentos significativos en el peso del proventrículo ($P=0,025$), peso relativo del intestino ($P=0,017$) y longitud del ciego ($P<0,0001$). Asimismo, este grupo exhibió una notable disminución en los niveles de albúmina ($P<0,0001$) y un aumento en la actividad de GOT ($P=0,002$). En conclusión, el PCG muestra efectos prometedores como aditivo nutricional y promotor de crecimiento natural para las codornices japonesas. Sin embargo, se recomienda incorporarlo después del período de crecimiento y ser cauteloso con dosis más altas debido a los posibles riesgos de toxicidad.

Palabras clave: Polvo de cáscara de Granada; *Coturnix japonica*; formulación de alimentos; rendimiento de crecimiento; parámetros bioquímicos

INTRODUCTION

As the global population surges, the search for sustainable sources of animal proteins becomes increasingly critical [1]. The Japanese quail (*Coturnix japonica*), known for its meat and eggs, stands out due to its high nutritional value and economic viability [2]. Consuming two quails daily can meet 40% of human protein needs, because of their rich content of proteins and essential amino acids [3]. This bird species offers multiple advantages: short reproductive cycles, low feed consumption, and a high reproduction capacity. Its adaptability to various farming conditions [4] and lesser environmental impact, make it an ideal choice for agricultural systems aiming to minimize animal breeding ecological footprint, while efficiently meeting market demands [5]. Moreover, quails' robustness against many pathogens presents a significant competitive advantage, since they exhibit better disease resistance than chickens.

Historically, using antibiotics as growth promoters in poultry farming was a widespread practice [6]. This approach has significantly contributed to antibiotic resistance, a critical issue for global public health [7], that led to major revisions in animal farming methods. This includes the ban of these substances in several countries [6]. Thus, the animals farming industry is now turning towards effective and eco-friendly alternatives [8, 9], specifically the valorisation of agricultural by-products into beneficial resources, which stands as an efficient approach to reduce dependence on antibiotics.

Ineffective management of agricultural by-products not only exacerbates environmental problems (such as pollution and waste treatment challenges) but also results in the loss of economic and ecological values [10]. The growing interest in valorising agro-industrial by-products for sustainable resource management and as an effective alternative to antibiotics directs our research towards one of the most abundant yet underutilized raw materials: pomegranate peel (*Punica granatum*).

Global pomegranate production is estimated to be between 2.5 and 3 million tons annually. A significant portion of this output, represented by pomegranate peels, comprises between 30 and 50% of the total fruit weight [11, 12]. According to the FAO, this volume of peels generates approximately 1.3 to 1.5 million tons of industrial wastes each year [13]. Pomegranate peels are valued for their health benefits attributed to their high content of tannins, flavonoids and various phenolic compounds [14]. These bioactive components provide the fruit with its antioxidant and anti-inflammatory properties [1], as well as antimicrobial, antifungal and antiparasitic attributes [12], making it effective in treating several illnesses like various types of cancer for instance [1]. According to Teniente *et al.* [10], polyphenols in pomegranate peels are able to modulate biological mechanisms involved in cervical, breast, and lung cancer. In fact, these substances can neutralise free radicals and inhibit lipid oxidation in fatty foods [1]. Additionally, pomegranate peel boasts a remarkable abundance of other phytochemicals, including vitamins, dietary fibres, essential minerals (potassium, calcium, phosphorus, magnesium, sodium) and complex polysaccharides [15, 16]. Thus, our present study aims to valorise pomegranate peel, a frequently disregarded by-product, by integrating it into the diet of Japanese quails. This approach seeks to replace antibiotics as growth promoters, thereby aiding in the fight against antibiotic resistance.

MATERIALS AND METHODS

Animals and diets

The study was conducted at the experimental farm of the Institute of Veterinary Sciences, University Constantine 1 Frères Mentouri, Constantine, Algeria. Fertile quail eggs were incubated, and the chicks were fed a standard growing diet without pomegranate (*Punica granatum*) peel powder (PPP) for the first week to avoid any potential negative effects from certain anti-nutritional factors. At 7 days (d) old, 208 unsexed Japanese quails were weighed and then randomly divided into four groups: a control group (CTRL) (without supplementation) and three experimental groups that received diets with varying concentrations of PPP (3, 5, and 7%). Each group consisted of 4 replicates of 13 quails. During the experiment, the birds were kept in galvanized cages under uniform conditions with continuous lighting and *ad libitum* access to feed and water.

Pomegranate peels were obtained from local suppliers. They were dried in shade, ground into a fine powder and sieved to achieve a consistent quality. Then, the final product was integrated into carefully designed diets to meet Japanese quails' nutritional needs at their various development stages. Feed formulations were made using the Windows User-Friendly Feed Formulation tool (WUFFDA ver. 1.02, 2004), according to the standards established by the National Research Council NRC [17]. The chemical composition of the PPP is presented in TABLE I and the detailed composition of the diets for each experimental group are shown in TABLE II [18].

TABLE I
Chemical composition of pomegranate peel

Component	Content
Moisture	4.4%
Mineral matter	3.65%
Crude protein	4.8%
Crude fat	5.9%
Crude fiber	10.4%
Ca	1.34%
Phosphore	0.13%
ME ¹	3167.3 Kcal·kg ⁻¹

¹ME: Metabolized Energy

Performance Measurement

Feed intake (FI), body weight (BW), feed conversion ratio (FCR), average daily gain (ADG), body weight gain (BWG), and mortality rates (M%) were recorded throughout the study. FCR, ADG, and BWG were calculated for the grower, finisher, and total rearing periods.

Slaughter and post-mortem analyses

At 42 d of age, 5 males and 5 females were randomly selected from each group and weighed individually using a precision digital scale (Princeton Instruments, model YP601N, accuracy: 0.1 g, maximum capacity: 600 g, USA). These quails were firstly kept separately and subjected to a 12-h fast while allowing them access to water. They were

TABLE II
Ingredients and nutrient composition of diets during grower and finisher periods

Ingredients (g·kg ⁻¹ of feed)	Grower				Finisher			
	CTRL	3%PPP	5%PPP	7%PPP	CTRL	3%PPP	5%PPP	7%PPP
Yellow corn	51.25	49.42	48	46.35	59.50	55	55.35	53.50
Soybean meal 48%	41.13	40.55	40.9	41.05	26.50	30.30	27.4	27.75
Wheat bran	5.87	5.10	4.35	3.85	12.25	9.95	10.50	10
Limestone (%)	0.75	0.75	0.75	0.75	0.75	0.75	0.75	0.75
Dicalcium Phosphate (%)	0.75	0.75	0.75	0.75	0.75	0.75	0.75	0.75
MV premix* (%)	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
PPP %	0	3	5	7	0	3	5	7
Total	100	100	100	100	100	100	100	100
Dry matter (%)	88.1	88.23	88.29	88.36	87.96	88.11	88.15	88.23
ME (kcal·Kg ⁻¹)	2800	2800	2800	2800	2800	2800	2800	2800
Proteins (%)	24	24	24	24	20.01	20	20	20
Fat (%)	2.53	2.44	3.33	3.24	2.89	2.69	2.69	2.61
Crude fiber (%)	3.38	3.53	3.65	3.78	3.69	3.80	3.97	4.09

ME: Metabolized Energy, *MV premix: Mineral-Vitamin supplement provided per kg of diet: Vitamin A: 3750.75 IU, Vitamin D3: 1249.875 IU, Vitamin E: 7.5 mg, Vitamin K3: 0.8 mg, Vitamin B1: 0.6425 mg, Vitamin B2: 0.0175 mg, Vitamin B6: 1.5 mg, Vitamin B12 : 0.004675 mg, Niacin: 17.8 mg, Folic acid: 0.3125 mg, Pantothenic acid: 3.55 mg, Biotin: 0.05 mg, Choline: 150.4125 mg, Betaine: 87.4925 mg, Fe: 12.65 mg, Cu: 3.45 mg, Mn: 25.325 mg, Zn: 16.9 mg, Se: 0.1025 mg, Iode: 0.515 mg, Butylated hydroxyanisole (BHA): 0.05 mg, Ethoxyquin: 0.05 mg, Sepiolite: 7.5075 mg, DL Methionine: 413 mg, L-Lysine: 0.0125%

then euthanized by severing the jugular veins, and subsequently, the feathers, head, viscera, and legs were removed from the carcasses.

The empty carcasses were weighed using an analytical balance (KERN, model PLS, accuracy: 0.0001 g, capacity: 510 g, Germany) and the carcass percentage calculated using the formula established by Brake *et al.* [19]:

$$Dres\ sin\ g\ (\%) = \left(\frac{Empty\ carcass\ weight}{Live\ body\ weight} \right) \times 100$$

Weights of the liver, spleen, intestines, heart, proventriculus, gizzard, ovaries, testes and abdominal fat were measured using the same analytical scale and used to calculate their relative weights as percentages of the total body weight. The entire digestive tract, small intestine, and caeca length measurements were also recorded.

From each bird, blood was collected into a heparinised tube. Then, plasma samples were obtained by centrifugation at 0,805 G for 10 min (Sigma, model 1-6P, maximum speed: 2,837 G, Germany) and stored at -20°C until analysis (ENIEM, model CF 1301, 350 L capacity, Algeria). Later, they were used to measure biochemical parameters, such as blood glucose, cholesterol, triglycerides, total protein, albumin, urea, creatinine, total bilirubin, direct bilirubin, alpha-amylase, calcium, phosphorus, glutamic-oxaloacetic transaminase (GOT), and glutamic-pyruvic transaminase (GPT), using colorimetric methods with commercial kits (Bio Lab®, France; Spinreact®, Spain) and a semi-automatic spectrophotometer (Mindray BA-88A, China).

Statistical analysis

Statistical analysis was performed through SPSS 25 software (SPSS Inc Chicago, IL, USA, 2017). Assessing normality was conducted using the Shapiro-Wilk and the Kolmogorov-Smirnov tests and homogeneity

of variance components between experimental groups by Levene's test. Data that adhered to both normality and variance homogeneity criteria were analysed using one-way ANOVA followed by Tukey's test for post-hoc comparisons. In case of variance heterogeneity, the Welch test and the Games-Howell test were applied. The Kruskal-Wallis and Dunn's tests were utilised for non-normal data for multiple comparisons. Gender differences were evaluated using the independent sample t-test or the Mann-Whitney U test when the data did not meet the normality prerequisite. Results are presented as means ± standard deviation.

RESULTS AND DISCUSSION

Productive performance

The TABLE III presents the Japanese quails' production performances over a 42 d rearing period. During the grower period, quails of the 7% PPP group showed a significantly lower BW than the CTRL and 3% PPP groups ($P < 0.0001$ for both). The 5% PPP group also showed a significantly lower BW than the 3% PPP group ($P = 0.029$). However, the BW in the 3% PPP group, although higher, was not significantly different from the CTRL group. During the grower period, the group fed 7% PPP, had significantly lower WG, BWG and ADG compared to the CTRL ($P = 0.004$) and 3% PPP groups ($P = 0.002$). Additionally, the 7% PPP group had significantly higher FCR and FI compared to the CTRL ($P = 0.017$ and $P = 0.037$ respectively) and 3% PPP groups ($P = 0.002$ and $P = 0.030$ respectively).

In the finisher phase, significant BW differences were noted in the first week, with 5 and 7% PPP groups showing lower BWs compared to the CTRL group ($P = 0.035$ and $P = 0.007$ respectively). The 7% PPP group also had significantly lower BW than the 3% PPP group ($P = 0.018$). No significant disparities in ADG and BWG were

TABLE III
Effect of diets containing different levels of pomegranate peel powder on quail performances

Performances	CTRL	3% PPP	5% PPP	7% PPP	P values	
Grower period	BW at 7 days (g)	26.12 ± 6.11	28.72 ± 4.49	24.51 ± 4.04	23.10 ± 3.93	NS
	BW at 15 days (g)	77.49 ± 20.50 ^a	88.56 ± 11.38 ^b	63.61 ± 8.13 ^c	61.74 ± 9.25 ^c	<0.0001
	BW at 21 days (g)	119.30 ± 17.95 ^{ab}	122.63 ± 15.30 ^{ab}	113.42 ± 15.49 ^{ac}	107.12 ± 15.06 ^c	<0.0001
	ADG (g·d ⁻¹)	6.56 ± 1.07 ^a	6.67 ± 0.90 ^{ab}	6.31 ± 0.97 ^{ab}	5.95 ± 0.91 ^b	0.017
	BWG (g)	91.94 ± 14.99 ^{ab}	93.48 ± 12.60 ^{ab}	88.44 ± 13.65 ^{abc}	83.41 ± 12.77 ^c	0.001
	FI (g·d ⁻¹)	9.42 ± 1.51 ^a	10.01 ± 0.39 ^a	11.23 ± 0.96 ^{ab}	11.41 ± 0.55 ^b	0.029
	FCR	2.19 ± 0.46 ^a	2.24 ± 0.08 ^a	2.68 ± 0.38 ^{ab}	2.87 ± 0.13 ^b	0.001
	M (%)	9.23 ± 8.42	5.76 ± 3.84	5.76 ± 3.84	7.69 ± 0	NS
Finisher period	BW at 28 days (g)	161.43 ± 19.4 ^{ab}	161.74 ± 20.76 ^{ab}	152.13 ± 18.70 ^{bc}	150.27 ± 16.13 ^c	0.001
	BW at 35 days (g)	194.35 ± 25.22	195.20 ± 27.89	187.57 ± 26.21	184.99 ± 19.46	NS
	BW at 42 days (g)	214.31 ± 34.42	215.25 ± 34.38	205.52 ± 29.13	204.72 ± 24.24	NS
	ADG (g·d ⁻¹)	3.77 ± 1.66	3.98 ± 1.53	3.81 ± 1.28	3.88 ± 1.23	NS
	BWG (g)	52.87 ± 23.37	53.50 ± 20.10	53.38 ± 17.97	54.45 ± 17.25	NS
	FI (g·d ⁻¹)	27.88 ± 2.22 ^{ab}	30.47 ± 2.65 ^{ab}	23.95 ± 1.80 ^c	24.89 ± 0.79 ^{ac}	0.001
	FCR	11.09 ± 1.50	12.11 ± 1.65	9.78 ± 1.53	9.41 ± 0.64	NS
	Total FCR	5.45 ± 0.30	5.92 ± 0.52	5.77 ± 0.35	5.29 ± 0.55	NS
M (%)	1.09 ± 2.90	4.00 ± 4.63	0.00 ± 0.00	0.00 ± 0.00	NS	

NS: Not significant. BW: Body weight, ADG: Average daily gain. BWG: Body weight gain. FI: Feed intake. FCR: Feed conversion ratio. M: mortality. Means bearing different superscripts within the same row are significantly different ($P < 0.05$)

observed thereafter, suggesting that the early benefits of 3% PPP do not continue into later stages of grower period. During the finisher stage, a marked sexual dimorphism was observed, with females significantly becoming heavier than males. Throughout this period, no significant dissimilarities in FCR were observed between the groups. On the other hand, FI analysis revealed a significant decrease in the 7% PPP group as compared to the CTRL ($P=0.03$) and 3% PPP groups ($P=0.002$). The 5% PPP group also showed significantly lower FI than the 3% PPP group ($P=0.007$). These results indicate that increasing PPP in the diet can reduce FI. Although, groups with different PPP levels showed reduced mortality rates compared to the CTRL group. These differences were statistically significant throughout neither the grower phase nor the finisher one.

The incorporation of 7% PPP increased FCR and FI while reducing BW, ADG and BWG during the grower period. However, 5 and 7% PPP reduced FI during the finisher period without affecting BW, BWG or ADG. The influence of the birds' age on fibre digestibility and intestinal morphology can explain these observations. For instance, it was reported that crude protein, fat, and energy digestibility decreased in young turkeys on high-fiber diets, but not at older ages [20]. Additionally, soluble fibres have been shown to increase intestinal viscosity, which reduces nutrient absorption and may potentially lower weight gain in poultry [21]. During the finisher phase, quails' metabolism and digestive systems are more stable and adapted to fibres digestion, suggesting that PPP supplementation at post-grower stage minimizes negative impacts on growth performances and feed efficiency. Another study [22] suggests that improved enzymatic activity in the gastrointestinal system enhances nutrient digestion and absorption while reducing FI. Furthermore, antioxidants in PPP, such as flavonoids and phenolic acids, are expected to lower oxidative

stress and intensify energy metabolism that promotes growth even with reduced feed consumption [23, 24]. Additionally, PPP's antibacterial properties can decrease bacterial loads and improve intestinal health and immunity that are essential for poultry development [24].

The findings reported by Sharifian *et al.* [25], describing a decreased FI without affecting growth in broilers fed pomegranate peel extract, corroborate well with our observations. However, other studies showed some disparities. For example, it was found that Japanese quails given low doses (0.5, 1, 1.5 and 2%) of PPP with cadmium chloride did not show any alteration in their FI [26]. Conversely, another study Maqsood *et al.* [27] observed that quails fed 7.5% PPP and raised on the ground, performed worse in terms of growth, but not FI. Differences in rearing conditions could also explain these discrepancies.

Our findings indicate a significant decrease in FI among Japanese quails during the finisher phase with higher PPP levels. Quails receiving 7% PPP had the lowest FI, aligning with the results reported by El-Rayes *et al.* [22]. This reduced consumption may be due to the high concentration of tannins in PPP that affect palatability. Conversely, an increase in FI with 7.5% PPP in 10-week-old quails was noted by Abbas *et al.* [28], possibly due to differences in the quails' initial age and the duration of the experiment. Our study showed no significant effect on FCR, differing from the findings of other research, where an increased FCR but similar final weights to the control group were recorded [29]. We also observed no significant differences in mortality rates, unlike the findings of Hamad *et al.* [30], who reported a reduced mortality with 1 and 1.5% PPP. This discrepancy may be due to our longer study duration, different rearing conditions and drying methods used for PPP. It has been documented that drying methods significantly affect the phytochemical composition and nutritional properties of pomegranate peels, which impact the nutrients quality and their availability [23].

Our study highlights a sexual dimorphism, with females showing higher final BW and BWG than males in all groups. These findings align with those of other research [31], and support the ones presented in another study regarding the leading role of genetic factors in sexual dimorphism expression [32].

Carcass traits

After analysing the carcass traits (TABLE IV), no significant variations were found among the different groups in slaughter weight, carcass weight, carcass yield and absolute and relative weights of organs. However, the 7% PPP group showed a significantly higher proventriculus weight than the CTRL ($P=0.026$) and 3% PPP ($P=0.013$) groups. This last group also had longer caeca ($P<0.0001$) and a higher intestinal proportion ($P=0.017$). Notably, most quails in the 7% PPP group exhibited paler livers. These findings align with those of Kamel *et al.* [33], who found no notable changes with 3%, 6% and 9% PPP levels, and Habibi *et al.* [29], who reported no significant changes with 0.5% and 2%

PPP. However, our study and the one of Kamel *et al.* [33] found no significant change in liver weight, while Habibi *et al.* [29] noted a significant reduction. Methodological differences, particularly in PPP levels, may explain these discrepancies.

A notable finding of our study, is the significant impact of 7% PPP on proventriculus weight and caeca length, that corroborates with the observations of Rezvani *et al.* [34], who noticed increased proventriculus weight in broiler chicks with 2% pomegranate seed extract. Although pomegranate seeds have less tannin than pomegranate peel [16], the digestive system's compensatory response to tannins may explain the increased proventriculus weight. Tannins form complexes with proteins, reducing their digestibility and prompting the digestive system to secrete more enzymes, through potentially enlarged proventriculus [35]. These findings are in contrast with those of another research [29], in which no significant variations in intestinal proportions were recorded.

TABLE IV
Effect of diets containing different levels of PPP on Carcass characteristics

Measurements	CTRL	3% PPP	5% PPP	7% PPP	P values
Slaughter BW (g)	211.26 ± 30.13	220.20 ± 26.82	207.41 ± 23.58	206.55 ± 29.90	NS
Carcass weight (g)	144.02 ± 16.35	153.92 ± 14.12	145.34 ± 16.10	138.56 ± 16.70	NS
Carcass yield (%)	68.57 ± 4.76	70.35 ± 6.05	70.21 ± 4.38	67.43 ± 4.68	NS
Liver weight (g)	4.94 ± 1.62	4.69 ± 1.88	5 ± 1.64	5.11 ± 2	NS
Liver relative weight (%)	2.32 ± 0.63	2.08 ± 0.67	2.39 ± 0.71	2.41 ± 0.66	NS
Heart weight (g)	1.60 ± 0.23	1.65 ± 0.17	1.62 ± 0.24	1.58 ± 0.24	NS
Heart relative weight (%)	0.76 ± .12	0.75 ± 0.08	0.78 ± 0.10	0.76 ± 0.05	NS
Gizzard weight (g)	3.94 ± 0.61	3.71 ± 0.66	3.67 ± 0.69	3.98 ± 0.80	NS
Gizzard relative weight (%)	1.87 ± 0.26	1.68 ± 0.18	1.77 ± 0.29	1.91 ± 0.15	NS
Abdominal fat weight (g)	0.94 ± 0.91	1.25 ± 1.15	1.11 ± 0.47	0.90 ± 0.88	NS
Abdominal fat relative weight (%)	0.42 ± 0.39	0.57 ± 0.50	0.53 ± 0.24	0.42 ± 0.38	NS
Gonads weight (g)	14.46 ± 10.38	12.39 ± 14.48	10.96 ± 6.46	14.61 ± 9.28	NS
Proventriculus weight (g)	1.55 ± 0.44 ^a	1.34 ± 1.04 ^a	1.69 ± 0.59 ^{ab}	1.97 ± 0.42 ^b	0.025
Intestine weight (g)	8.25 ± 3.13	8.80 ± 2.74	9.93 ± 3.76	10.97 ± 3.60	NS
Intestine relative weight (%)	3.84 ± 1.18 ^a	3.92 ± 0.84 ^{ab}	4.69 ± 1.33 ^{ab}	5.23 ± 1.19 ^b	0.017
Intestine length (cm)	65.05 ± 12.56	60.79 ± 7.06	65.80 ± 10.26	69.22 ± 12.01	NS
Caeca length (cm)	9.50 ± 1.99 ^{ab}	10.15 ± 2.10 ^{ab}	11.28 ± 1.67 ^{abc}	13.13 ± 2.16 ^c	<0.0001

Means bearing different superscripts within the same row are significantly different at $P<0.05$.

Biochemical parameters

As shown in Table V, biochemical analysis reveals minimal significant differences among the groups. Markedly, the 7% PPP group had significantly lower albumin levels compared to the CTRL ($P=0.009$), 3% PPP ($P=0.002$) and 5% PPP ($P=0.0001$) groups. No change in albumin levels occurred with 5% PPP, aligning with Elnaggar *et al.* [36], who found no significant disparities in broilers with 0.25 to 1.5% PPP. However, in our study, albumin levels significantly dropped with 7% PPP, corroborating with the results of Kamel *et al.* [33], who observed reduced albumin at 9% PPP in Japanese quails. These findings are in contrast with those of Akuru *et al.* [37] who reported

increased albumin with 6 to 8% PPP in broilers chicks, possibly due to species differences, PPP dosage or other factors not yet investigated.

GOT activity was higher in the 7% PPP group compared to the CTRL ($P=0.038$), 3% PPP ($P=0.012$) and 5% PPP ($P=0.031$) groups. Akuru *et al.* [37] also reported increased GOT levels in quails fed a 7% PPP diet, on the opposite of Ashour *et al.* [38] who stated that broilers fed a mix of medicinal plants including hot red pepper, thyme, rosemary, anise, spearmint, black cumin and garlic showed decreased GOT levels. These plants possess antioxidant properties akin to those of PPP, indicating that the antioxidant effects may differ depending on the plant type and concentration. Additionally, a reduction in

TABLE V
Effect of the incorporation level of PPP on specific biochemical indices in Japanese quails

Parameters	CTRL	3%PPP	5%PPP	7%PPP	P values
Glucose (g·L ⁻¹)	3.42 ± 0.63	3.24 ± 0.50	3.84 ± 0.69	3.74 ± 0.57	NS
Triglycerides (g·L ⁻¹)	2.89 ± 2.28	3.18 ± 1.74	3.21 ± 1.60	2.83 ± 1.81	NS
Albumin (g·L ⁻¹)	16.96 ± 5.1 ^{3a}	20.57 ± 4.84 ^a	19.25 ± 3.39 ^a	12.52 ± 1.73 ^b	<0.0001
Total protein (g·L ⁻¹)	40.65 ± 11.61	33.01 ± 11.27	38.53 ± 12.11	42.36 ± 10.51	NS
GOT (U·L ⁻¹)	43.55 ± 28.78 ^{ab}	33.50 ± 15.07 ^{ab}	65.60 ± 28.16 ^a	93.30 ± 46.46 ^c	0.002
GPT (U·L ⁻¹)	7.46 ± 6.46	7.34 ± 2.51	7.02 ± 4.72	6.97 ± 4.74	NS
Total cholesterol (mg·dL ⁻¹)	4.03 ± 1.49	3.63 ± 0.91	4.03 ± 0.65	4.61 ± 1.18	NS
Urea (g·L ⁻¹)	0.05 ± 0.03	0.04 ± 0.03	0.03 ± 0.03	0.03 ± 0.01	NS
Creatinine (g·L ⁻¹)	3.27 ± 2.05	3.19 ± 0.66	4.38 ± 2.02	4.50 ± 1.73	NS
Total bilirubin (mg·L ⁻¹)	43.39 ± 13.74	44.80 ± 10.19	44.20 ± 9.92	47.62 ± 8.69	NS
Direct bilirubin (mg·L ⁻¹)	36.35 ± 16.64	36.36 ± 15.28	42.18 ± 16.69	43.73 ± 20.55	NS
Alpha amylase (U·L ⁻¹)	2566.66 ± 1919.87	3128.20 ± 1761.55	1543 ± 1262.80	1509.66 ± 1295.15	NS
Calcium (mg·L ⁻¹)	137.73 ± 58.87	110.75 ± 45.75	146.60 ± 51.86	151.86 ± 46.93	NS
Phosphorus (mg·L ⁻¹)	108.64 ± 36.42	114.09 ± 51.16	102.47 ± 23.15	105.32 ± 43.18	NS

Means bearing different superscripts within the same row are significantly different at $P < 0.05$.

GOT levels in broilers consuming PPP has been previously reported by Elnaggar et al. [36]. In our study, serum GOT concentrations, an indicator of hepatocellular injury, suggests no harmful liver effects at up to 5% PPP. However, increased GOT levels at 7% PPP might indicate potential liver toxicity. The lack of GPT variation, another specific liver damage marker, complicates this interpretation [39].

The pale liver seen in quails with 7% PPP might suggest some hepatotoxicity. This observation aligns with the findings reported in the study of Monjur et al. [40], where dietary lead (Pb) caused significant toxic effects in broiler chickens, including pale liver colour, hepatomegaly and elevated GPT levels as definitive indicators of liver damage. In our study, the absence of GPT variations suggests that pomegranate peel may exert less severe toxic effects.

CONCLUSIONS

This study highlights the potential of PPP as a natural additive in poultry diets, offering a promising alternative to synthetic supplements. The findings indicate that while PPP can be incorporated into quail diets without major negative effects at certain levels, attention must be given to higher concentrations due to potential health risks, such as liver toxicity.

The inclusion of PPP aligns with the growing need for more sustainable and environmentally friendly agricultural practices, potentially reducing the reliance on chemical additives in poultry farming. The study underscores the importance of determining optimal dosages to maximize the benefits of PPP, not only in quails but also across a range of poultry species. Overall, PPP presents a viable and nutritious alternative, contributing to a more eco-conscious approach to poultry nutrition.

Conflicts of interest

The authors declare no conflicting interests.

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