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Phytase supplementation maintains productive performance, egg quality, and blood biochemical profile in Japanese quails fed phosphorus-reduced diet

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Abstract: This study aimed to investigate the effects of phytase supplementation in phosphorus (P)-reduced diet on the productive performance, egg quality, calcium (Ca) utilisation, and blood profile of Japanese quails. Phytase breaks down phytic acid; thereby, increasing the availability of its bound nutrients, enabling poultry to hydrolyse and utilise these nutrients effectively. A total of 480 female Japanese quails (*Coturnix coturnix japonica*; 28-day-old and average body weight = 94.3 ± 5.7 g) were assigned to four dietary regimens, each consisting of six replicates of 20 birds. The control diet (T0) contained 0.5% nonphytate P. The other experimental diets included: T1 = comprising 0.4% nonphytate P supplemented with 0.1% phytase equivalent to 500 Phytase Unit (FTU)/kg; T2 = comprising 0.3% nonphytate P supplemented with 0.15% phytase equivalent to 750 FTU/kg; and T3 = comprising 0.2% nonphytate P supplemented with 0.2% phytase equivalent to 1 000 FTU/kg. The addition of phytase to P-reduced diets did not affect the overall productive performance in quails. Moreover, yolk weight increased by 3.04% to 10.5% (P = 0.01) and the haugh unit increased by 0.56% to 1.11% (P = 0.04) compared to the control, whereas other quality traits, such as albumen weight, albumen and yolk indices, and eggshell weight, thickness, and breaking strength, remained unaffected following the supplementation of phytase in the P-reduced diet. Additionally, an increase in Ca output in eggshells (5.26% to 15.79%; P = 0.14) and the ratio of Ca in eggshells to Ca intake (5.59% to 16.47%; P = 0.49) correlated with the increase in blood Ca levels in the quails on the P-reduced diet (P = 0.16). In conclusion, the addition of phytase to P-reduced diets has proven effective in maintaining the laying performance, egg quality, and blood biochemical profiles of Japanese quails.

Keywords: coturnix; egg quality; layer; nonphytate; production

Phosphorus (P) is a crucial mineral in poultry nutrition, essential for skeletal development, eggshell formation, and overall productivity. However, in plant-based ingredients, such as maize, wheat, and soybean meal, a significant portion of P is found in the form of phytic acid (Riviere et al. 2021). Phytic acid is a reactive ligand that strongly binds with minerals, such as P, Ca, Mg, Zn, and Fe, small molecules like phosphates, and macromolecules, such as protein and glucose. These chelating properties reduce the bioavailability of P as well as other essential nutrients (Wang and Guo 2021).

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Poultry are monogastric animals that lack the enzyme phytase, which is necessary for hydrolysing phytic acid. This deficiency leads to inefficient nutrient absorption and high P excretion in manure (Englmaierova et al. 2015; Wang and Guo 2021; Eltahan et al. 2023). Consequently, this increases the need for supplemental inorganic P; thereby, raising the production costs, and contributing to environmental concerns, such as soil and water pollution (Demirel et al. 2012; Hirvonen et al. 2019). It is important to note that natural deposits of phosphates are limited. Some phosphate deposits, such as phosphate rocks, may contain cadmium, a toxic heavy metal that can reduce P availability (Santos et al. 2016). To address these challenges, the use of exogenous phytase enzymes has been incorporated into poultry diets. Phytase catalyses the stepwise dephosphorylation of phytic acid, releasing P and other bound nutrients, thereby improving their bioavailability. Commercial phytases, particularly those produced from *Aspergillus niger*, *Klebsiella pneumoniae*, and *Buttiauxella* spp. have shown promising results in enhancing nutrient availability (Qiugang et al. 2019). Notably, phytases derived from *Buttiauxella* spp. are particularly effective in degrading phytic acid from maize and soybean meal into lower inositol phosphates (IP_1–IP_5), which are more easily absorbed in the poultry digestive system (Hirvonen et al. 2019).

By improving phosphorus utilisation, phytase supplementation can reduce reliance on limited phosphate rock resources, thus supporting more sustainable poultry production systems (Qiugang et al. 2019). Studies on quails have reported improvements in performance and blood plasma profiles with phytase supplementation of up to 75 mg/kg diet (Arafa et al. 2019). Furthermore, a diet containing 0.32% available P supplemented with 5 000 Phytase Unit (FTU)/kg was shown to maintain laying performance and egg quality in hens (Saleh et al. 2021).

Japanese quails (*Coturnix coturnix japonica*) are becoming increasingly popular in commercial egg production owing to their rapid growth, high laying capacity, and low input requirements. Thus, optimising their P intake is essential for maintaining performance, health, and sustainability (Ren et al. 2020). Although phytase has been extensively studied in chickens, limited information is available on its impact on Japanese quails, particularly regarding productive performance, egg quality, and blood biochemical parameters under P-reduced dietary conditions. Given the growing role of quails in poultry production, it is essential to assess whether phytase can maintain optimal performance and physiological condition in this species when dietary P levels are reduced. Therefore, this study aimed to evaluate the effects of phytase supplementation in P-reduced diets on productive performance, egg quality, and blood biochemical profiles in Japanese quails. The findings are expected to provide new insights into the nutritional management of quails and the role of phytase in enhancing P efficiency under restricted dietary conditions.

MATERIAL AND METHODS

Quails, experimental design, and diets

The research protocol received approval from the Ethics Committee for Experimental Animal Research (Registration No. 357/UN27.20/PT.01.01/2022). A total of 480 female Japanese quails (28 days old) with an average body weight of 94.3 ± 5.7 g were used in this study. The quails were randomly assigned in a completely randomised experimental design into four dietary treatments, each consisting of six replicates of 20 birds. The diets were as follows: T0 = basal diet (control) containing 0.5% nonphytate P; T1 = 0.4% nonphytate P supplemented with 0.1% phytase (equivalent to 500 FTU/kg); T2 = 0.3% nonphytate P supplemented with 0.15% phytase (equivalent to 750 FTU/kg); T3 = 0.2% nonphytate P supplemented with 0.2% phytase (equivalent to 1 000 FTU/kg). All diets were formulated to be isoprotein and isoenergetic, containing 18.02% crude protein and 11.4 MJ/kg of metabolisable energy. The phytase used was obtained from Natuphos® 500G (BASF, Germany). According to the producer (BASF), Natuphos 500G is 3-phytase produced by *Aspergillus niger* (EC 3.1.3.8). The composition and nutrient content of the experimental diets are presented in Table 1.

Data collection on productive performance and egg quality

The birds were randomly placed in 24 colony battery cages (length = 75, width = 50, and height = 40 cm), with universal maintenance management. The birds were housed under natural temperature conditions,

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Table 1. Composition and nutrient content of the experimental diets

Ingredients	T0	T1	T2	T3
Yellow corn (%)	50.0	50.0	50.0	50.0
Rice bran (%)	11.2	11.2	11.2	11.2
Soybean meal (%)	30.7	30.7	30.7	30.7
Limestone (%)	5.10	5.45	5.83	6.18
Dicalcium phosphate (%)	2.15	1.70	1.20	0.75
DL-methionine (%)	0.08	0.08	0.08	0.08
Premix (%)[1]	0.15	0.15	0.15	0.15
NaCl (%)	0.25	0.25	0.25	0.25
Phytase (%)	0.00	0.10	0.15	0.20
Nutrient content				
Metabolisable energy (MJ/kg)	11.4	11.4	11.4	11.4
Crude protein (%)	18.0	18.0	18.0	18.0
Calcium (%)	3.24	3.24	3.24	3.24
Nonphytate phosphorus (%)	0.50	0.40	0.30	0.20
Lysine (%)	1.01	1.01	1.01	1.01
Methionine (%)	0.40	0.40	0.40	0.40

[1]Supplied per kg diet: calcium 720 mg, phosphorus 22.5 mg, iron 6 mg, manganese 4.1 mg, iodine 0.1 mg, cuprum 0.3 mg, zinc 3.8 mg, vitamin B12 0.001 mg, and vitamin D3 75 IU

T0 = basal diet (Control) contained 0.5% nonphytate P; T1 = diet contained 0.4% nonphytate P supplemented with 0.1% phytase equivalent to 500 FTU/kg; T2 = diet contained 0.3% nonphytate P supplemented with 0.15% phytase equivalent to 750 FTU/kg; T3 = diet contained 0.2% nonphytate P supplemented with 0.2% phytase equivalent to 1 000 FTU/kg

with an average ambient temperature during the experiment at 25.4 °C in the morning, 33.4 °C in the afternoon, and 29.9 °C in the evening. The lighting schedule was 16 h light (5 a.m.–9 p.m.) and 8 h dark (9 p.m.–5 a.m.). The birds were fed a commercial diet until they reached 35 days of age. A basal diet based on corn and soybean meal was administered during the laying period. The dietary treatments were started when egg production reached 10%, indicating that the birds had reached sexual maturity (Ratriyanto and Prastowo 2019).

The treatments lasted for three periods of 28 days (3 × 28 days) with *ad libitum* feed and water supply (Ratriyanto and Prastowo 2019). The feed intake, egg production, and egg weight were recorded daily. The feed conversion ratio was calculated by measuring the feed intake to egg mass ratio (Ratriyanto et al. 2020). Physical egg quality was measured during the last 3 days of each period. In total, 216 eggs were collected from each period, with 9 eggs per replicate, ensuring the egg weight was close to the average, allowing for a determination of egg quality. The eggs were weighed and cracked, and its components were weighed. Eggshell thickness was measured with a digital micrometer (0–25 × 0.001 mm, Digital Outside Micrometer 3 109 – 25A; Insize, P.R. China). Eggshell-breaking strength was measured using a device that recorded breaking strength with 0.01 N accuracy at a constant velocity of 50 mm/min to measure head shift (Kibala et al. 2018). Egg and yolk weights were measured by weighing the eggs and egg yolks. The albumen weight was calculated by subtracting the yolk and eggshell weights from the total egg weight (Ratriyanto et al. 2020). The haugh unit (HU) was calculated using the formula: $100 \times \log (H + 7.57 - 1.7 \times W^{0.37})$, where H is albumen height, and W is egg weight (Zita et al. 2013).

Blood profile analysis

Blood samples were taken from 72 quails (3 quails per replicate) using a vacuum syringe from the right pelvic vein. Blood serum was separated through centrifugation at 3 500 rpm for 10 min (Morishita 2019). The blood serum samples were analysed for chemical content using the photometric method according to DiaSys Diagnostic Systems using Microlab 300 (Vital Scientific BV; Elitech Group

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Company, The Netherlands). Blood Ca concentration was measured using the Calcium AS FS Kit, P concentration was measured using the Phosphate FS Kit, glucose level was measured using the Glucose God FS Kit, and total protein concentration was measured using the Total Protein FS Kit (DiaSys Diagnostic System, Germany).

Calcium utilisation

Calcium (Ca) utilisation was assessed following the feeding treatment period, which was complemented by a five-day excreta collection period involving 48 birds (2 birds per replicate), ensuring their body weight was close to the average. The excreta were then gathered and sun-dried.

The excreta and diet samples were milled through a 0.5 mm mesh screen for analysis. Ca retention (in g/bird) was calculated by subtracting the excreted Ca from the total Ca intake (Walker et al. 2024). The Ca levels in the diet, excreta, and eggshells were analysed using atomic absorption spectrophotometry. Ca utilisation was evaluated as the ratio of Ca in the eggshell to Ca intake (Arpasova et al. 2010).

Data analysis

The data were analysed with variance analysis. The following model was applied:

$$y_{ij} = \mu + \alpha_i + \varepsilon_{ij} \quad (1)$$

where:

μ – the general mean;

α_i – the effect of diets;

ε_{ijk} = experimental error.

The statistically different means were compared using Tukey's test at $P < 0.05$. All statistical analyses were performed using R Statistic program language v4.4.3 (R Core Team 2021).

RESULTS AND DISCUSSION

Performance and egg quality

Reducing the P content in the diet from 0.5% to 0.2% and supplementing with phytase up to 1 000 FTU/kg did not change the laying performance, indicated by similar feed intake, egg production, egg weight, and feed conversion of all treatments (Table 2).

This finding indicates that phytase supplementation in P-reduced diets effectively maintained productive performances. This consistency can be attributed to phytase's ability to increase P availability by hydrolysing phytic acid, which releases bound P and other essential minerals. These results align with previous findings that demonstrate phytase can greatly reduce the requirement for inorganic P supplementation without compromising laying performance (Saleh et al. 2021; Pirzado et al. 2024).

Numerous studies have shown that phytase supplementation improves egg production performance in chickens when nonphytate P levels are low. For instance, hens fed on low nonphytate P diets supplemented with phytase demonstrated egg production and feed efficiency similar to that of hens on high nonphytate P diets (Roque et al. 2023). Eltahan et al. (2023) found that hens fed on diets containing 0.20% nonphytate P supplemented with phytase increased egg production and improved eggshell thickness, whereas egg weight and feed intake remained unchanged. Similar en-

Table 2. Productive performance of quails fed on P-reduced diet supplemented with phytase

Variables	T0	T1	T2	T3	*P*-value
Feed intake (g/day)	18.9 ± 1.67	19.3 ± 1.04	18.8 ± 0.71	18.1 ± 1.86	ns
Egg production (%)	64.5 ± 11.97	64.9 ± 7.37	64.6 ± 6.96	63.0 ± 11.2	ns
Egg weight (g)	9.41 ± 0.30	9.51 ± 0.05	9.48 ± 0.19	9.33 ± 0.19	ns
Feed conversion	3.17 ± 0.41	3.15 ± 0.23	3.09 ± 0.21	3.12 ± 0.29	ns

ns = not significant; T0 = basal diet (Control) contained 0.5% nonphytate P; T1 = diet contained 0.4% nonphytate P supplemented with 0.1% phytase equivalent to 500 FTU/kg; T2 = diet contained 0.3% nonphytate P supplemented with 0.15% phytase equivalent to 750 FTU/kg; T3 = diet contained 0.2% nonphytate P supplemented with 0.2% phytase equivalent to 1 000 FTU/kg

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hancements in laying performance have also been observed in hens fed diets with reduced P and supplemented with phytase (Ren et al. 2020; Roque et al. 2023).

Moreover, the birds fed on a diet containing 0.4% to 0.2% P supplemented with 500–1 000 FTU/kg phytase showed an increase in yolk weight from 3.04% to 10.5% (P = 0.01) and improvements in HU values from 0.56% to 1.11% (P = 0.04) compared to those fed the control diet (P < 0.05). The treatments did not affect other egg quality parameters, such as albumen weight and index, yolk index, eggshell weight, thickness, and breaking strength (Table 3). The increase in yolk weight observed in quail fed on diets supplemented with phytase can be attributed to increased nutrient availability. Phytase enhances the digestibility of essential nutrients, such as P, Ca, Zn, and proteins, by breaking down phytic acid, which in turn enhances gut health and enzyme activity (Ren et al. 2020; Pirzado et al. 2024). These nutrients are critical components of egg yolk, and their improved absorption contributes to the increase in yolk weight. Additionally, phytase facilitates the release and absorption of myo-inositol from phytic acid in the jejunum and ileum. Myo-inositol may then be transferred into the egg yolk, further contributing to its formation and supporting yolk weight.

Phytase supplementation did not negatively affect egg quality traits, including egg weight, specific gravity, and HU (Jing et al. 2021; Kayan et al. 2025). In some studies, it was found that phytase maintains yolk and improves albumen quality (Habibollahi et al. 2019; Kayan et al. 2025). The increased egg weight commonly observed with phytase supplementation may indirectly contribute to better egg quality by improving nutrient efficiency (Englmaierova et al. 2015; Shet et al. 2018). However, other research did not indicate a significant impact on HU or albumen weight (Eltahan et al. 2023; Kayan et al. 2025). This suggests that phytase has limited effects on the freshness of internal egg quality. Nonetheless, parameters, such as yolk and albumen quality, remained stable in low-P diets supplemented with phytase, indicating that internal egg quality can be maintained under these conditions (Englmaierova et al. 2015; Habibollahi et al. 2019; Saleh et al. 2021; Eltahan et al. 2023).

In this study, eggshell weight, thickness, and breaking strength were preserved across treatments, demonstrating that phytase supplementation compensates for dietary P reductions. This effect likely results from the improved Ca and P bioavailability facilitated by phytase activity (Hervo et al. 2023). Comparable findings have been reported that eggshell weight (Saleh et al. 2021) and thickness (Kayan et al. 2025) in quails fed on low digestible-P diets supplemented with phytase did not differ from those fed on a normal diet.

Several studies confirm phytase's role in improving eggshell characteristics. For instance, Englmaierova et al. (2015) and Shet et al. (2018) observed significant increases in eggshell thickness and strength in hens fed on a 1.8 g/kg nonphytate P diet supplemented with 350 FTU/kg phytase. Similarly, phy-

Table 3. Egg quality of quails fed on P-reduced diet supplemented with phytase

Variables	T0	T1	T2	T3	P-value
Yolk weight (g)	2.94 ± 0.09^{c}	3.11 ± 0.13^{a}	3.25 ± 0.15^{a}	3.04 ± 0.19^{b}	0.01
Albumen weight (g)	4.65 ± 0.25	4.61 ± 0.18	4.74 ± 0.21	4.72 ± 0.28	ns
Yolk index	0.51 ± 0.01	0.51 ± 0.01	0.52 ± 0.01	0.50 ± 0.02	0.06
Albumen index	0.08 ± 0.01	0.09 ± 0.01	0.09 ± 0.01	0.09 ± 0.01	ns
Haugh unit	89.9 ± 0.62^{b}	90.6 ± 0.22^{a}	90.9 ± 0.31^{a}	90.4 ± 0.91^{a}	0.04
Eggshell weight (g)	0.79 ± 0.02	0.80 ± 0.02	0.79 ± 0.03	0.79 ± 0.04	ns
Eggshell thickness (mm)	0.16 ± 0.02	0.17 ± 0.01	0.17 ± 0.02	0.18 ± 0.01	ns
Eggshell-breaking strength (kg/cm^2)	0.81 ± 0.01	0.82 ± 0.01	0.83 ± 0.00	0.82 ± 0.00	ns

$^{a–c}$Means in the same row with different superscripts are significantly different (P < 0.05)

ns = not significant; T0 = basal diet (Control) contained 0.5% nonphytate P; T1 = diet contained 0.4% nonphytate P supplemented with 0.1% phytase equivalent to 500 FTU/kg; T2 = diet contained 0.3% nonphytate P supplemented with 0.15% phytase equivalent to 750 FTU/kg; T3 = diet contained 0.2% nonphytate P supplemented with 0.2% phytase equivalent to 1 000 FTU/kg

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tase supplementation enhanced eggshell weight and mineralisation in laying hens (Eltahan et al. 2023). The improvement of eggshell quality is supported by phytase's ability to increase P retention and reduce P excretion, which in turn improves mineral availability for eggshell formation (Habibollahi et al. 2019). P plays a vital role in Ca metabolism, which is essential for eggshell mineralisation. Consequently, phytase's role in P hydrolysis indirectly supports the structural integrity of the eggshell. Furthermore, the addition of phytase improves eggshell quality, which indirectly protects the internal egg components from physical damage during handling and transport (Habibollahi et al. 2019; Kayan et al. 2025).

Calcium utilisation

Phytase supplementation in a P-reduced diet did not affect the Ca intake and retention. However, reducing P in the diet to 0.3% and 0.2% and supplementing with 750 and 1 000 FTU/kg phytase, respectively, increased Ca output in the eggshells ranging from 5.26% to 15.79% (P = 0.14), ultimately increasing the ratio of Ca output in eggshell to Ca intake ranging from 5.59% to 16.47% (P = 0.49; Table 4).

Ca is a vital mineral for the formation of eggshells. Poultry primarily obtains Ca from dietary sources, such as calcium carbonate and Ca bound to phytic acid found in plant-based feed ingredients. Phytic acid is the main storage form of P in plant-derived feedstuffs and has a strong binding affinity for minerals, including Ca, that reduces their bioavailability. Consequently, phytic acid is considered an antinutritional factor that can impair the absorption and utilisation of minerals (Hervo et al. 2023).

Phytase is included in poultry diets because it effectively hydrolyses phytic acid, releasing Ca and P, which enhances their retention and utilisation in the gastrointestinal tract (Krieg et al. 2021). The breakdown of phytic acid causes phytase to reduce its antinutritional effects; thus, facilitating better mineral absorption. This mechanism is essential for maintaining adequate mineral status and supporting optimal egg production. Phytase supplementation has been shown to increase the availability of Ca and P, which work together to develop strong and well-formed eggshells (Eltahan et al. 2023). Ensuring sufficient quantities of both minerals contributes to improved eggshell quality, including increased thickness and strength (Jing et al. 2021; Eltahan et al. 2023). Studies have demonstrated that diets supplemented with phytase, even when formulated with reduced levels of Ca and P, can maintain eggshell quality similar to that found in diets with higher mineral content (Liu et al. 2007; Demirel et al. 2012).

In quail nutrition, reducing dietary P followed by phytase supplementation effectively fulfils Ca requirements. Evidence suggests that the presence of phytase increases overall Ca utilisation. Birds receiving phytase-supplemented diets (T2 and T3) exhibited higher Ca deposition in eggshells and improved ratios of Ca in eggshells relative to intake, indicating enhanced efficiency in Ca utilisation (Arpasova et al. 2010).

Blood biochemical profile

Reducing the P content in the diet from 0.5% to 0.2% and supplementing with phytase up to 1 000 FTU/kg yielded similar levels of blood glucose, protein, P, and Ca. Moreover, reducing the P content from 0.5% to 0.4% and supplementing

Table 4. Ca utilisation of quails fed on P-reduced diet supplemented with phytase

Variables	T0	T1	T2	T3	P-value
Ca intake (g/day)	0.57 ± 0.03	0.57 ± 0.06	0.58 ± 0.03	0.54 ± 0.57	ns
Ca retention (g)	0.43 ± 0.04	0.42 ± 0.06	0.43 ± 0.06	0.41 ± 0.08	ns
Ca output in eggshell (%)	0.19 ± 0.01^{c}	0.20 ± 0.01^{bc}	0.22 ± 0.01^{a}	0.21 ± 0.01^{ab}	0.014
Ca eggshell: Ca intake (%)	34.0 ± 3.07^{b}	35.9 ± 3.42^{ab}	37.6 ± 2.31^{a}	39.6 ± 4.09^{a}	0.049

$^{a–c}$Means in the same row with different superscripts are significantly different (P < 0.05)

T0 = basal diet (Control) contained 0.5% nonphytate P; T1 = diet contained 0.4% nonphytate P supplemented with 0.1% phytase equivalent to 500 FTU/kg; T2 = diet contained 0.3% nonphytate P supplemented with 0.15% phytase equivalent to 750 FTU/kg; T3 = diet contained 0.2% nonphytate P supplemented with 0.2% phytase equivalent to 1 000 FTU/kg

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Table 5. Blood profile of quails fed on P-reduced diet supplemented with phytase

Variables	T0	T1	T2	T3	*P*-value
Glucose (mg/dl)	309.6 ± 28.17	296.9 ± 7.66	289.7 ± 12.39	256.7 ± 112.85	ns
Protein (g/dl)	4.48 ± 0.38	4.99 ± 0.87	5.52 ± 0.77	4.76 ± 0.49	ns
P (mg/dl)	5.03 ± 1.87	5.68 ± 0.93	6.03 ± 1.03	6.17 ± 1.44	ns
Ca (mg/dl)	26.6 ± 3.81^{b}	31.6 ± 4.06^{a}	28.2 ± 2.50^{b}	24.5 ± 3.43^{b}	0.016

^{a,b}Means in the same row with different superscripts are significantly different ($P < 0.05$)

ns = not significant; T0 = basal diet (Control) contained 0.5% nonphytate P; T1= diet contained 0.4% nonphytate P supplemented with 0.1% phytase equivalent to 500 FTU/kg; T2 = diet contained 0.3% nonphytate P supplemented with 0.15% phytase equivalent to 750 FTU/kg; T3 = diet contained 0.2% nonphytate P supplemented with 0.2% phytase equivalent to 1 000 FTU/kg

with 500 FTU/kg phytase increased the Ca levels in the blood by 18.8% ($P = 0.16$; Table 5). Phytase supplementation plays a critical role in maintaining the biochemical balance of blood in poultry, particularly when diets are formulated with reduced levels of nonphytate P. Phytase facilitates the hydrolysis of phytic acid through a dephosphorylation process that sequentially breaks down myo-inositol phosphates, specifically pentakis-, tetrakis-, tris-, bis-, and monophosphate, ultimately yielding myo-inositol and inorganic phosphate ions (Hirvonen et al. 2019; Qiugang et al. 2019). During this enzymatic breakdown, several essential nutrients bound to phytic acid, including P, Ca, N, and amino acid, are released, enhancing their bioavailability (Liu et al. 2007).

The increased mineral liberation helps maintain stable P concentrations in the blood, even in birds fed on low-P diets. Similar to this study, previous observations indicate that phytase supplementation results in blood glucose, total protein, and P levels that are comparable to those observed in control diets, suggesting that nutrient absorption and metabolic functions remain unaffected (Demirel et al. 2012). Additionally, the elevated serum Ca levels observed, even though all treatment groups received diets with equal Ca content, underline the role of phytase in improving Ca availability by releasing it from phytic acid complexes. Evidence supporting phytase's positive impact on mineral status includes findings that layers fed phytase-supplemented low-P diets exhibit higher serum P concentrations than those fed unsupplemented diets (Saleh et al. 2021). Phytase has also been shown to increase the apparent utilisation rate of P while reducing its excretion, indicating more efficient absorption and retention of this vital mineral (Jing et al. 2021; Iqbal et al. 2023).

Beyond its effects on blood mineral levels, phytase also enhances Ca retention in laying hens, particularly in cases of dietary P deficiency (Pirzado et al. 2024; Walker et al. 2024). Additionally, increased tibial Ca content in broilers receiving phytase-supplemented diets has been observed, further indicating improved Ca absorption and utilisation (Iqbal et al. 2023). These outcomes can be attributed to phytase's ability to break down phytic acid, releasing bound minerals and promoting overall mineral balance.

Moreover, phytase appears to affect the expression of genes involved in mineral metabolism. Research suggests that phytase supplementation upregulates genes associated with Ca reabsorption in the kidneys and bones while downregulating genes associated with bone resorption; thus, contributing to Ca homeostasis (Votterl et al. 2020). In addition to its impact on mineral metabolism, phytase has been linked to changes in hormonal and plasma biochemical parameters. Notably, its supplementation has been associated with increased levels of albumin, high-density lipoprotein, P, Ca, estradiol-17β, and luteinising hormone in laying hens (Eltahan et al. 2023). These physiological enhancements may support sustained egg production and improve eggshell quality.

CONCLUSION

Reducing P content in the diet followed by supplementation with phytase can assist in maintaining egg production in quails. This is evident from observations related to feed intake, egg production, egg weight, and feed conversion efficiency. Furthermore, adding phytase to P-reduced diets can improve several egg quality-related traits com-

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pared with a standard diet. Additionally, the improvement in the Ca output in the eggshells and the ratio of Ca in the eggshells to Ca intake might be linked to increased blood Ca concentration. Therefore, this study emphasises the beneficial impacts of phytase supplementation in a P-reduced diet, promoting sustainable poultry production by allowing for lower inorganic P addition.

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Conflict of interest

The authors declare no conflict of interest.

REFERENCES

Arafa MM, El-Gendi GM, El-Garhy OH. Effect of dietary phytase enzyme supplementation on growth performance and some blood parameters of Japanese quails. Ann Agric Sci Moshtohor. 2019 Mar 1;57(1):59-66.

Arpasova H, Halaj M, Halaj P. Eggshell quality and calcium utilisation in feed of hens in repeated laying cycles. Czech J Anim Sci. 2010 Feb 18;55(2):66-74.

Demirel G, Pekel AY, Alp M, Kocabagli N. Effects of dietary supplementation of citric acid, copper, and microbial phytase on growth performance and mineral retention in broiler chickens fed a low available phosphorus diet. J Appl Poult Res. 2012 Jul;21(2):335-47.

Eltahan HM, Cho S, Rana MM, Saleh AA, Elkomy AE, Wadaan MA, Alagawany M, Kim IH. Dietary exogenous phytase improve egg quality, reproductive hormones, and prolongs the lifetime of the aging hy-line brown laying hens fed nonphytate phosphorus. Poult Sci. 2023 Sep; 102(9):102895.

Englmaierova M, Skrivan M, Skrivanova E, Bubancova I, Cermak L, Vlckova J. Effects of a low-phosphorus diet and exogenous phytase on performance, egg quality, and bacterial colonisation and digestibility of minerals in the digestive tract of laying hens. Czech J Anim Sci. 2015 Dec; 60(12):542-9.

Habibollahi M, Abousadi MA, Nakhaee P. The effect of phytase on production performance, egg quality, calcium and phosphorus excretion, and fatty acids and cholesterol concentration in hy-line layers fed diets containing rice bran. J Appl Poult Res. 2019 Sep;28(3):688-98.

Hervo F, Letourneau-Montminy MP, Meme N, Meda B, Duclos MJ, Narcy A. Effect of phytase and limestone particle size on mineral digestibility, performance, eggshell quality, and bone mineralisation in laying hens. Poult Sci. 2023 May;102(5):102613.

Hirvonen J, Liljavirta J, Saarinen MT, Lehtinen MJ, Ahonen I, Nurminen P. Effect of phytase on in vitro hydrolysis of phytate and the formation of myo-inositol phosphate esters in various feed materials. J Agric Food Chem. 2019 Oct 23;67(41):11396-402.

Iqbal W, Yaseen MA, Rahman MA, Bhatti SA, Rahman MS, Yaqoob MU, Ahmad F, Zahid MU, Shoaib M. Effect of phytase supplementation on growth performance, mineral digestibility, and tibia calcium and phosphorous in broilers fed low phosphorus diets. Braz J Poult Sci. 2023 Oct;25(4):eRBCA-2023.

Jing M, Zhao S, Rogiewicz A, Slominski BA, House JD. Effects of phytase supplementation on growth performance, plasma biochemistry, bone mineralisation and phosphorus utilisation in pre-lay pullets fed various levels of phosphorus. Anim Prod Sci. 2021 Feb;61(6):568-76.

Kayan A, Prasongsook S, Poeikhampha T. Diet supplementation with phytase and xylanase on laying performance, egg quality, apparent metabolisable energy, phosphorus and calcium use in laying hens. Vet World. 2025 Jan; 18(1):155-61.

Kibala L, Rozempolska-Rucinska I, Kasperek K, Zieba G, Lukaszewicz M. Eggshell qualities as indicative of eggshell strength for layer selection. Braz J Poult Sci. 2018 Jan-Mar;20(1):99-102.

Krieg J, Borda-Molina D, Siegert W, Sommerfeld V, Chi YP, Taheri HR, Feuerstein D, Camarinha-Silva A, Rodehutscord M. Effects of calcium level and source, formic acid, and phytase on phytate degradation and the microbiota in the digestive tract of broiler chickens. Anim Microbiome. 2021 Dec;3(1):1-8.

Liu N, Liu GH, Li FD, Sands JS, Zhang S, Zheng AJ, Ru YJ. Efficacy of phytases on egg production and nutrient digestibility in layers fed reduced phosphorus diets. Poult Sci. 2007 Nov;86(11):2337-42.

Morishita Y. Poultry blood collection. Columbus, OH: The Ohio State University College of Veterinary Medicine; 2019.

Pirzado SA, Liu G, Purba MA, Cai H. Enhancing the production performance and nutrient utilisation of laying hens by augmenting energy, phosphorus and calcium deficient diets with fungal phytase (Trichoderma reesei) supplementation. Animals (Basel). 2024 Jan;14(3):376.

https://doi.org/10.17221/83/2025-CJAS

Qiugang MA, Rodehutscord M, Novotny M, Li L, Yang L. Phytate and phosphorus utilisation by broiler chickens and laying hens fed maize-based diets. Front Agric Sci Eng. 2019 Dec;6(4):380-7.

R Core Team. R: A language and environment for statistical computing [Internet]. Vienna: R Foundation for Statistical Computing; 2021 [cited 2025 Oct 13]. Available from: https://www.R-project.org/.

Ratriyanto A, Firmanda F, Purwanti H, Murjoko M. Nutrient digestibility, performance, and egg quality traits of quails raised in different stocking densities and ascorbic acid supplementation in a hot, tropical environment. Turk J Vet Anim Sci. 2020;44(2):350-7.

Ratriyanto A, Prastowo S. Floor space and betaine supplementation alter the nutrient digestibility and performance of Japanese quail in a tropical environment. J Therm Biol. 2019 Jul;83:80-6.

Ren Z, Sun W, Cheng X, Liu Y, Han D, Yan J, Pan C, Duan Y, Yang X. The adaptability of Hy-Line Brown laying hens to low-phosphorus diets supplemented with phytase. Poult Sci. 2020 Jul;99(7):3525-31.

Riviere A, Nothof T, Greiner R, Tranchimand S, Noiret N, Robert F, Mireaux M. In vitro assessment of enzymatic phytate dephosphorylation during digestive process of different feeds and feed ingredients. Anim Feed Sci Technol. 2021 Nov;281:115096.

Roque FA, Zorzetto PS, Leite BGS, Granghelli CA, Goncalves JC, Garcia NACR, Wen N, Moraes UR, Araujo CS, Araujo LF. Effects of phytase supplementation to reduced-nutrient diets on performance, egg quality, and economic parameters in commercial layers. Rev Colomb Cienc Pecu. 2023 Jul-Sep;36(3):121-30.

Saleh AA, Elsawee M, Soliman MM, Elkon RY, Alzawqari MH, Shukry M, Abdel-Moneim AM, Eltahan H. Effect of bacterial or fungal phytase supplementation on the performance, egg quality, plasma biochemical parameters, and reproductive morphology of laying hens. Animals (Basel). 2021 Feb;11(2):540.

Santos ID, Rodrigues SL, Siqueira JO, Monte MBM, Dutra AJB. Effect of partial oxidation of organic matter on cadmium leaching from phosphate. Miner Eng. 2016 Dec; 99:67-75.

Shet D, Ghosh J, Ajith S, Awachat VB, Elangovan AV. Efficacy of dietary phytase supplementation on laying performance and expression of osteopontin and calbindin genes in eggshell gland. Anim Nutr. 2018 Mar;4(1):52-8.

Votterl JC, Klinsoda J, Zebeli Q, Hennig-Pauka I, Kandler W, Metzler-Zebeli BU. Dietary phytase and lactic acid-treated cereal grains differently affected calcium and phosphorus homeostasis from intestinal uptake to systemic metabolism in a pig model. Nutrients. 2020 May; 12(5):1542.

Walker H, Vartiainen S, Apajalahti J, Taylor-Pickard J, Nikodinoska I, Moran CA. The effect of including a mixed-enzyme product in broiler diets on performance, metabolisable energy, phosphorus and calcium retention. Animals (Basel). 2024 Jan;14(3):328.

Wang R, Guo S. Phytic acid and its interactions: Contributions to protein functionality, food processing, and safety. Compr Rev Food Sci Food Saf. 2021 Mar;20(2):2081-105.

Zita L, Ledvinka Z, Klesalova L. The effect of the age of Japanese quails on certain egg quality traits and their relationships. Vet Arhiv. 2013 Mar;83(2):223-32.

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