

## EFFECTS OF BORIC ACID AND BORAX PENTAHYDRATE ON PERFORMANCE, EGG QUALITY TRAITS AND BONE MINERALIZATION IN LAYING HENS

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### Abstract

*This study evaluated the effects of dietary supplementation with boric acid and borax pentahydrate at two levels (50 and 100 mg/kg) on production performance, egg quality traits, and bone mineralization in laying hens. A total of 315 Atak-S hens were randomly assigned to five groups: a control group and four treatment groups. The inclusion of 100 mg/kg borax pentahydrate in the diet significantly improved egg production, reduced feed intake, and resulted in the most efficient feed conversion ratio ( $P < 0.05$ ). Moreover, boron supplementation significantly increased eggshell thickness, particularly in the borax pentahydrate group ( $P < 0.05$ ), while internal egg quality parameters (Haugh unit and albumen index) remained unaffected ( $P > 0.05$ ). A slight reduction in yolk redness was observed at higher boron levels. However, dietary boron supplementation had no significant effect on bone mineralization parameters ( $P > 0.05$ ). Overall, the findings suggest that dietary boron, especially in the form of borax pentahydrate, can enhance productivity and eggshell quality in laying hens without negatively affecting internal egg characteristics.*

**Key words:** boric acid, borax pentahydrate, egg quality traits, laying hens, productive performance.

### INTRODUCTION

Boron, an essential trace element, plays a significant role in various physiological processes in animals, including bone development, mineral metabolism, and enzymatic functions (Hunt, 1994). In recent years, boron has gained attention in animal nutrition, particularly in poultry production, for its potential benefits, despite not being classified as an essential nutrient (Nielsen, 2014). Research has highlighted its importance in improving growth performance, eggshell quality, and overall mineralization in laying hens (Rossi et al., 2018). Boron supplementation has been associated with enhanced reproductive health, bone strength, and production efficiency in poultry, with studies suggesting its positive effects on calcium metabolism and the utilization of other minerals like magnesium and phosphorus (Kucuk et al., 2003; Wilson & Ruszler, 1997; Hunt, 1994).

The effectiveness of boron supplementation in animal diets largely depends on its form and concentration. Boric acid and borax pentahydrate are two commonly used boron

sources, each with distinct chemical properties and bioavailability (Devirian & Volpe, 2003). Research has demonstrated that dietary boron supplementation can enhance egg production and shell quality in laying hens, but the optimal form and dosage remain unclear (Eren et al., 2004). While studies have primarily focused on boron's effects on eggshell quality and bone mineralization, there is limited information on its impact on internal egg quality parameters, such as albumen and yolk characteristics. Recent findings suggest that boron supplementation in poultry diets may improve egg production, egg quality, and overall health (Eren et al., 2004). For example, Nielsen (2000) highlighted boron's role in enhancing calcium metabolism and bone strength in mammals, suggesting similar benefits for poultry. Eren et al. (2012) further demonstrated that dietary boron positively influences eggshell quality and mechanical properties in laying hens. Additionally, Bozkurt et al. (2015) found that boron supplementation improved eggshell thickness and strength, emphasizing its role in mineral utilization. Similarly, Khattak et al. (2018) reported that dietary boron enhanced bone mineralization and tibia

strength in broilers, indicating its potential to support skeletal integrity in laying hens. These findings underscore the need for further research to optimize boron supplementation strategies for poultry production. Various boron compounds, including boric acid, borax, and sodium tetraborate, have been evaluated in poultry diets. For instance, Kabu and Akosman (2013) found that boric acid supplementation at different levels positively influenced calcium and phosphorus metabolism, contributing to improved eggshell formation. Kurtoglu et al. (2005) further indicated that dietary boron supplementation, regardless of the source, significantly improved bone mineralization and reduced osteoporosis in aged hens. Beyond its effects on bone and eggshell health, boron has been shown to influence immune response and antioxidant enzyme activity. Pradhan et al. (2021) reported that boron supplementation improved immune function by increasing antioxidant capacity and reducing oxidative stress in poultry. Oğuz et al. (2017) also found that dietary boron enhanced the production of immune-related cytokines, strengthening disease resistance. In terms of feed efficiency, Akhavan-Salamat and Ghasemi (2020) explored boron's effects on nutrient absorption and gut health, highlighting its potential to improve feed conversion ratios. Mohammadpour et al. (2016) supported these findings, showing that boron supplementation improved intestinal morphology, nutrient digestibility, and overall performance in broilers. However, there is limited research on the specific effects of boric acid and borax pentahydrate supplementation in laying hens, particularly regarding their impact on yield performance, egg traits, and mineral deposition, underscoring the need for further investigation in this area. Understanding the effects of these boron sources on laying hens is essential for optimizing dietary formulations and enhancing egg production performance. This study aims to evaluate the impact of boric acid and borax pentahydrate on yield performance, egg traits, and mineralization in laying hens. By exploring these parameters, the research will contribute to the broader field of poultry nutrition and provide practical insights for the poultry industry.

## MATERIALS AND METHODS

This research was carried out in compliance with ethical standards and received approval from the Dicle University Experimental Animal Ethics Committee (DÜHADEK) under protocol number 2022/202.

Atak-S hens, at 23 weeks of age, were housed in enriched cage systems (n = 315; 45 cages; 7 hens per cage; floor space of 200 cm<sup>2</sup> per hen). After a 2-week adaptation period, the hens were fed experimental diets for 12 weeks. Throughout the trial, the hens were provided with the same basal diet formulated according to the National Research Council (NRC, 1994) recommendations. While the control group was fed the basal diet, the diets of the other groups were supplemented with boron in different forms (boric acid-sodium pentaborate pentahydrate) and levels (50 mg/kg and 100 mg/kg).

Table. 1 Ingredients, composition of hens diets

Ingredients	%
Sunflower oil	3.70
Maize	55.00
Soybean Meal (44%, CP)	22.00
Sunflower Meal (32%, CP)	8.00
Dicalcium Phosphate <sup>a</sup>	2.00
Limestone	8.50
DL-Methionine	0.20
NaCl	0.35
Vitamin and Mineral Premix <sup>b</sup>	0.25
Items	Analyzed values, %
Dry Matter	90.6
Crude Protein	17.0
Ether extract	5.20
Crude Ash	13.63
Crude Fiber	4.67
Items	Calculated values, %
Metabolizable energy (ME), kcal/kg	2806
Calcium	3.81
Available Phosphorus	0.44
Na	0.17
Cl	0.25
Methionine + Cysteine	0.78
Lysine	0.82
Threonine	0.72
Tryptophan	0.27
Linoleic acid	3.02
Electrolyte Balance (mEq/kg DM)	197

<sup>a</sup>Composition: 240 g Ca and 17.5 g P/kg; <sup>b</sup>Vit+Min. Mineral mixture provides the following nutrients per kg of diet: vitamin A: 12.000.000 IU, vitamin D3: 2.500.000 IU; vitamin E: 30 ppm; vitamin K3: 4.000 ppm, vitamin B1: 3.000 ppm, vitamin B2: 7.000 mg, vitamin B12: 5.000 ppm, vitamin C: 50.000 ppm, Biotin: 45 ppm, folic acid: 1.000 ppm, Fe: 60 ppm, Zn: 60 ppm, Cu: 74 ppm, Se, 150 ppm, Co: 200 ppm, I: 1.000 ppm, Mn: 80.000 ppm.

Lighting was maintained on a 16-hour light and 8-hour dark (16L:8D) schedule throughout the experiment. Feeders were manually replenished daily, and eggs were collected each morning.

At the beginning of the experiment, hens were weighed and placed in cages according to similar live weight and egg yields and treatment groups were formed. Feed intake, egg production and egg weight were measured daily during the trial. Feed conversion ratio and other performance data were calculated weekly. For this purpose, a total of 45 eggs were collected from each group over two consecutive days (in the morning), stored overnight at 4°C, and then subjected to external and internal quality analyses. To determine specific gravity, eggs were weighed after being stored in air for one day, then submerged in water at 22°C and weighed again (Hempe et al., 1988). Yolk color was measured as L\*, a\*, and b\* values using a digital colorimeter (Minolta CR-300). Albumen height was measured with a digital micrometer and calculated using the formula:  $[\text{albumen height (mm)} / ((\text{albumen length (mm)} + \text{albumen width (mm)}) / 2)] \times 100$ . Yolk index was determined by measuring yolk height and diameter with a digital caliper and applying the formula:  $[(\text{yolk height} / \text{yolk diameter}) \times 100]$ . Egg weight (g) and albumen height (mm) were used to calculate Haugh units with the formula:  $100 \times \log (H + 7.57 - 1.7G^{0.37})$ .

At the end of the trial, one hen from each subgroup (replicate) closest to the group average was slaughtered, and a total of six right femur and tibia bones were collected from each group. The femur and tibia bones were cleaned of muscle and fat tissues, wrapped in cling film to prevent air exposure, labelled, and stored in double plastic bags at -20°C for later analysis. Before testing, the bones were dried at 60°C for 24 hours and defatted using an ether solvent. For breaking strength measurements, the frozen bones were thawed at room temperature (28°C) for 16 hours. A Stepless Compression Test Machine was used for 3-point bending tests, with each bone centered under a 10 mm diameter probe moving at 0.5 mm/min. To identify the fracture point, 20 mm sections from the dorsal and distal ends of the femur were excluded, 40 mm was subtracted from the minimum bone length, and the remaining length was divided equally using a calliper.

After strength testing, the bones were dried at 105°C for 24 hours, cooled in a desiccator, and weighed. They were then fat-extracted with ether for 24 hours, reweighed, and ashed at

550°C for 24 hours to determine ash weight. The results were calculated as the percentage of ash relative to the dry, fat-free weight, following the method of Elaroussi et al. (1994). At the end of the trial, bone samples were collected from the femur of one hen per pen per week (n = 45). The samples were dried, ground, and prepared for mineral analysis. Data were subjected to one-way analysis of variance (ANOVA) using SPSS software (version 18.0; IBM Corp., Armonk, NY, USA). When significant differences were detected, treatment means were compared using Tukey's multiple comparison test at a significance level of  $P < 0.05$ .

## RESULTS AND DISCUSSIONS

Table 2 shows the effects of the experimental diets on egg production, egg weight, feed intake and feed conversion rate. Boric acid and borax pentahydrate supplements increased egg production while reducing feed consumption and improving feed conversion ratio. The highest egg production was observed in the borax pentahydrate (100 mg/kg) group (88.6%,  $P = 0.015$ ). The lowest feed consumption was also recorded in the borax pentahydrate (100 mg/kg) group (124.8 g/bird/day,  $P < 0.001$ ), and this was associated with the positive effects of boron supplementation on gut health. The best feed conversion ratio was found in the borax pentahydrate (100 mg/kg) group (2.43,  $P < 0.001$ ), which aligns with findings in the literature indicating that boron supports mineral metabolism and enhances feed utilization (Bozkurt et al., 2015; Rossi et al., 2018; Mohammadpour et al., 2016). Recent studies also support the positive effects of boron supplementation on digestive system health and feed efficiency (Akhavan-Salamat & Ghasemi, 2020; Khattak et al., 2018; Oguz et al., 2017). The highest egg production in the borax pentahydrate (100 mg/kg) group can be explained by boron's ability to improve gut health and enhance feed utilization (Akhavan-Salamat & Ghasemi, 2020). The decrease in feed consumption with increasing boron levels suggests that boron improves intestinal morphology and increases nutrient absorption (Mohammadpour & Kamyab, 2021). The lower feed conversion ratio is attributed to boron's

regulation of calcium and magnesium metabolism, which enhances energy utilization efficiency (Khattak et al., 2018).

The effects of boric acid and borax pentahydrate supplementation on external egg quality traits in hens throughout the study are shown in Table 3.

Boron supplements improved eggshell quality, with the highest shell thickness measured in the borax pentahydrate (100 mg/kg) group (0.379 mm,  $P = 0.026$ ). This result is consistent with studies showing that boron supports calcium metabolism, enhancing shell density and strength (Kabu & Akosman, 2013; Eren et

al., 2012). However, no significant differences were observed among the groups in terms of egg shape index, specific gravity, or shell ratio ( $P > 0.05$ ), which aligns with some studies suggesting that boron supplementation has limited effects on these parameters (Eren et al., 2004). Recent studies also support the positive effects of boron supplementation on eggshell density and quality (Mohammadpour & Kamyab, 2021).

The effects of boric acid and borax pentahydrate supplementation on internal egg quality traits in hens throughout the study are shown in Table 4.

Table 2. Effects of dietary supplementation with boric acid and borax pentahydrate on productive performance in hens throughout the study (25 to 37 wk of age)

Parameters	Control	Boric Acid (mg/kg feed)		Borax Pentahydrate (mg/kg feed)		SEM <sup>1</sup>	<i>P</i> -value
		50	100	50	100		
Egg Production, %	83.4 <sup>b</sup>	85.8 <sup>ab</sup>	86.5 <sup>ab</sup>	86.5 <sup>ab</sup>	88.6 <sup>a</sup>	0.498	0.015
Egg yield, g/hen/day	56.6 <sup>b</sup>	57.8 <sup>ab</sup>	58.8 <sup>a</sup>	57.4 <sup>ab</sup>	59.2 <sup>a</sup>	0.246	0.036
Feed Intake (FI), g/hen/day	138.9 <sup>a</sup>	136.0 <sup>a</sup>	130.8 <sup>ab</sup>	135.5 <sup>a</sup>	124.8 <sup>b</sup>	1.245	<0.001
Egg Weight (EW), g/day	59.1	61.3	60.2	59.8	60.8	0.303	0.156
Feed Conversion Rate (FI/EW)	2.62 <sup>a</sup>	2.56 <sup>ab</sup>	2.48 <sup>bc</sup>	2.54 <sup>abc</sup>	2.43 <sup>c</sup>	0.016	<0.001

<sup>ab</sup>Means within each period with different superscript letters are significantly different ( $P < 0.05$ ).

<sup>1</sup>SEM = pooled standard error of the mean.

Table 3. Effects of Dietary Supplementation with Boric Acid and Borax Pentahydrate on External Egg Quality Traits in Hens throughout the study (25 to 37 wk of age)

Parameters	Control	Boric Acid (mg/kg feed)		Borax Pentahydrate (mg/kg feed)		SEM <sup>1</sup>	<i>P</i> -value
		50	100	50	100		
Egg Shape Index	77.6	76.6	78.1	76.6	77.4	0.287	0.488
Specific Gravity	1.18	1.17	1.18	1.18	1.19	0.0005	0.345
Egg Shell Rate	11.3	11.4	11.5	11.7	11.7	0.078	0.614
Shell Thickness, mm	0.342 <sup>b</sup>	0.344 <sup>b</sup>	0.363 <sup>ab</sup>	0.360 <sup>ab</sup>	0.379 <sup>a</sup>	0.004	0.026

<sup>ab</sup>Means within each period with different superscript letters are significantly different ( $P < 0.05$ ).

<sup>1</sup>SEM = pooled standard error of the mean.

Table 4. Effects of Dietary Supplementation with Boric Acid and Borax Pentahydrate on Internal Egg Quality Traits in Hens throughout the study (25 to 37 wk of age)

Parameters	Control	Boric Acid (mg/kg feed)		Borax Pentahydrate (mg/kg feed)		SEM <sup>1</sup>	<i>P</i> -value
		50	100	50	100		
Haugh Unit	78.2	79.1	80.5	78.7	80.1	0.459	0.456
Albumen Index	8.99	9.16	9.25	8.93	9.12	0.137	0.956
Yolk Index	44.1	45.7	45.9	45.2	45.2	0.281	0.351
L*	55.1	55.5	55.2	54.5	53.8	0.230	0.118
a**	17.8 <sup>a</sup>	16.9 <sup>ab</sup>	17.1 <sup>ab</sup>	16.2 <sup>ab</sup>	15.0 <sup>b</sup>	0.295	0.020
b***	33.8	32.3	34.0	32.5	33.6	0.345	0.183

<sup>ab</sup>Means within each period with different superscript letters are significantly different ( $P < 0.05$ ).

L\*: lightness, a\*\*: redness, b\*\*\*: yellowness.

<sup>1</sup>SEM = pooled standard error of the mean.

The increase in shell thickness at higher boron levels can be explained by boron's support for calcium and phosphorus metabolism, improving shell mineralization (Kabu & Akosman, 2013). The lack of difference in egg shape index and specific gravity may be due to boron's more pronounced effect on external shell structure compared to internal structure. Boron supplementation had a limited impact on internal quality parameters, with no significant differences observed between groups in terms of Haugh unit and albumen index ( $P > 0.05$ ). However, as boron levels increased, the reddish hue of the egg yolk decreased ( $P = 0.020$ ), which may be linked to boron's effects on antioxidant enzyme activity (Oğuz et al., 2017; Pradhan et al., 2021). The limited effect on internal quality parameters suggests that boron's influence on gut health and mineral metabolism may not lead to significant changes in egg composition (Bozkurt et al., 2015; Akhavan-Salamat & Ghasemi, 2020). Recent studies also highlight boron's effects on antioxidant mechanisms, supporting the observed changes in yolk color (Khattak et al., 2018; Mohammadpour & Kamyab, 2021). The lack of significant differences in Haugh unit and albumen index suggests that boron primarily plays a role in mineral metabolism

rather than directly affecting protein structure (Bozkurt et al., 2015). The limited variations in internal quality parameters indicate that boron's metabolic effects are more focused on eggshell formation rather than egg composition (Mohammadpour & Kamyab, 2021). The bone mineralization results in Table 5, although not statistically significant, complement earlier findings in the study regarding improved mineral metabolism with boron supplementation - especially in groups receiving 100 mg/kg of either boric acid or borax pentahydrate. Although improvements in tibia and femur strength and mineral content were not statistically significant, the consistent upward trend - especially in calcium content - aligns with previous research (Khattak et al., 2018; Bozkurt et al., 2015) which noted enhanced bone mineralization from boron supplementation. The lack of significance might be attributed to high inter-individual variability or the duration of the study. Overall, the results suggest a potentially beneficial but subtle role of boron in bone mineralization, with more pronounced effects observed in egg production and shell quality. Longer-term studies or older hen populations might show clearer outcomes in skeletal metrics.

Table 5. Effects of Dietary Supplementation with Boric Acid and Borax Pentahydrate on Bone Mineralization in Hens (at 37 wk of age)

Parameters	Control	Boric Acid (mg/kg feed)		Borax Pentahydrate (mg/kg feed)		SEM <sup>1</sup>	<i>P-value</i>
		50	100	50	100		
Tibia strength (N)	87.1	89.1	92.6	89.4	88.8	0.541	0.089
Tibia ash (%)	48.2	49.1	48.5	49.0	48.8	0.256	0.632
Ca in Tibia (%)	16.9	17.0	17.2	16.9	17.5	0.296	0.442
P in Tibia (%)	7.6	7.4	7.7	7.5	7.8	0.212	0.096
Femur strength (N)	86.1	85.7	85.7	87.1	86.4	0.381	0.401
Femur ash (%)	45.2	46.5	46.7	45.9	46.8	0.130	0.536
Ca in Femur (%)	22.4	23.6	27.5	26.7	27.0	0.301	0.091
P in Femur (%)	9.2	8.9	9.0	9.3	9.2	0.112	0.381

<sup>a,b</sup>Means within each period with different superscript letters are significantly different ( $P < 0.05$ ).

<sup>1</sup>SEM = pooled standard error of the mean.

# CONCLUSIONS

This study explored the effects of boric acid and borax pentahydrate supplementation at different levels (50 and 100 mg/kg) on laying hens. The results showed that 100 mg/kg borax pentahydrate significantly improved egg production, feed efficiency, and eggshell

thickness without negatively affecting internal egg quality. While internal traits remained stable, a slight decrease in yolk redness was observed. Bone mineralization results showed positive trends but were not statistically significant. Overall, boron especially as borax pentahydrate proved beneficial for enhancing egg quality and productivity.

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