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Effects of Boron Supplementation Fed with Low Calcium to Diet on Performance and Egg Quality in Molted Laying Hens

Osman Olgun, Yusuf Cufadar and Alp Onder Yildiz
Department of Animal Science,
Faculty of Agriculture, University of Selcuk, 42079, Kampus, Konya, Turkey

Abstract: This study was conducted to different levels of dietary Calcium (Ca) with Boron (B) supplementation effect on performance and egg quality in molted laying hens. Two hundred and eighty, 78 weeks old, White Leghorn LSL laying hens were assigned to 8 groups with 7 replicates. Experiment had a 2×4 factorial arrangement of treatments with 2 levels of Ca diet (4.0% control and 3.5% low Ca) and 4 levels of B (Borax Pentahydrate) preparation (0, 100, 200 and 300 mg kg^{-1}) were used. Laying hens were fed to eight dietary treatments during the 78-90 weeks periods. The different levels of dietary Ca and B as a main factor did not significantly effect on initial body weight, final body weight, egg production, feed conversion ratio, specific gravity, egg shell weight, damaged egg and egg shell breaking strength except for egg weight, feed intake, egg mass, haugh unit and albumen index. There was no interaction effect of Ca and B levels on the parameters except for egg yolk index, egg shape index and egg shell thickness ($p<0.01$; $p<0.05$). It is concluded that molted laying hens to the diet were not added B when the laying hens consumed adequately feed and Ca.

Key words: Molted laying hens, boron, calcium, performance, egg quality, diet

INTRODUCTION

Eggshell quality has always been a problem in the layer industry especially in the last stage of the laying period. Numerous studies have been conducted to solve the problems of poor shell quality. Many of these studies have focused on macro minerals, especially Ca and P and trace minerals. Boron (B) is a trace element that is turning out to be essential for humans and also for animals (WHO, 1998) and it's abundant in nature as boric acid and borate and can be obtained in the diet through the consumption of fruits, vegetables and legumes (Sutherland *et al.*, 1998). Boron is known to influence a variety of metabolic actions, in addition interacts with Calcium (Ca), vitamin D and Magnesium (Mg), which are all important in bone metabolism (Devirian and Volpe, 2003). Boron has been examined as a possible nutritional factor in Ca metabolism and utilisation and thus, as a factor in the development and maintenance of normal bone (NRC, 1984). The NRC (1984) suggested that the trace mineral supplements to chemically defined diets should contain at least 2 ppm B. Hunt *et al.* (1983) in a series of experiments with day-old chicks fed for 30 days, indicated a relationship between B and other nutrients (Ca, Mg and vitamin D3). Rossi *et al.* (1993) shown that B is an important mineral for body weight, feed consumption and reduced mortality rate in broilers. Qin and Klandorf (1991) stated that the effects of dietary B on egg production, egg shell quality and Ca metabolism in broiler breeder. Kurtoglu *et al.* (2002) reported that the effects of dietary boron (0, 50, 100, 150, 200 and 250 mg kg^{-1}) on performance and egg quality are investigated in laying hens. There was no significant difference between the control and the boron supplemented group in the feed intake, feed conversion ratio, egg production, body weight, egg weight and specific gravity except for damaged eggs. Eren *et al.* (2004) investigated that dietary B supplementation (0, 5, 10, 50, 100, 200 and 400 mg kg^{-1}) effect on egg production, interior and exterior egg quality in laying hens. According to results of the study, body weight, feed intake and egg production were reduced in the 400 mg kg^{-1} B supplemented group. Boron changed the interior and exterior quality of eggs. Yesilbag and Eren (2008) reported that the effects of dietary boric acid supplementation on performance, egg shell quality and some serum parameters were examined in 100 layer hens at 60 weeks of age. The study results that feed efficiency and egg production values were not affected significantly by dietary treatments during the experiment. However, layer hens that received diets supplemented with boric

Corresponding Author: Alp Onder Yildiz, Department of Animal Science, Faculty of Agriculture, University of Selcuk, 42079, Kampüs, Konya, Turkey

acid had better feed intake, egg weight, percentage of damaged eggs and egg shell quality (eggshell thickness, eggshell breaking strength) than the control group. According to researchers that dietary B supplementation has positive effect on mineral balance and therefore, reduces the percentage of damaged eggs by improving eggshell thickness and egg shell breaking strength in aged laying hens. The aim of the present study, was to determine the effects of boron supplementation fed with low calcium to diet on performance and egg quality in molted laying hens.

MATERIALS AND METHODS

At 78 week old 168 molted White Leghorn LSL laying hens were fed to 8 dietary treatments during the 78-90 weeks periods. The birds were randomly divided into 8 treatments. Each treatment consisted of 7 replications (5 birds/replicate). The birds were fed a basal diet containing 15% CP and 2800 kcal ME kg^{-1}. Basal diets were shown in Table 1. Eight diets, arranged in a factorial design (2×4) with 2 levels of Ca diet (4.0% control and 3.5% low Ca) and 4 levels of boron (Borax Pentahydrate-$Na_2B_4O_7.5H_2O$) preparation (0, 100, 200 and 300 mg kg^{-1}) were used. In order to have a homogeneous distribution, powdered Borax Pentahydrate was added by premixing. For that calculated amounts of Borax Pentahydrate were mixed with a small volume of the ration and then added to main ration. The house had controlled ventilation and lighting (16 h day^{-1}). All hens were supplied with feed and water *ad libitum*.

Body Weight (BW) was obtained by weighting hens at the beginning and end of the experiment. Feed Intake (FI) and Egg Weight (EW) were recorded biweekly. Egg Production (EP) was recorded daily and Egg Specific Gravity (ESG) was measured monthly. Egg Mass (EM) was calculated from collecting data of EP and EW at biweekly via:

$$EM = \frac{EP \times EW}{\text{Period (days)}}$$

Feed Conversion Ratio (FCR; g of feed g^{-1} of egg) was calculated via:

$$FCR = \frac{\text{FI (g of feed/hen/period)}}{\text{EM (g of egg / hen/period)}}$$

Specific gravity and egg quality characteristics measurements were made on all collected eggs produced during 2 consecutive days of per at the end of 28 days period during the experiment. Specific gravity was determined same day using graded salt solutions ranging from 1.060-1.100 with gradations of 0.005 (Holder and Bradford, 1979). Egg shape index was determined by equipment (Digital calliper, Mitutoyo) that calculated the width: length ratio as a percentage. Egg yolk and albumen height was determined by digital height calliper.

Table 1: Composition of diets

Ingredients	Control diet (4.0% Ca)	Low-Ca diet (3.5% Ca)
Corn	57.000	60.000
Soybean meal (44% CP)[1]	15.500	15.000
Sunflower meal (28% CP)[1]	11.500	11.500
Vegetable oil (7800 kcal kg^{-1} ME)[1]	4.400	3.230
Limestone	9.790	8.460
Dicalcium phosphate	0.900	0.900
Premix[2]	0.250	0.250
Salt	0.400	0.400
Methionine	0.600	0.600
Sand	0.200	0.200
Total	100.000	100.000
Calculated nutrients		
ME (kcal kg^{-1})	2805.000	2802.000
Crude protein (%)	14.990	15.040
Ca (%)	4.000	3.510
Available P (%)	0.290	0.290
Lysine (%)	0.673	0.667
Methionine (%)	0.311	0.314
Methionine + Cystine (%)	0.600	0.605

[1]Analyzed value; [2]Premix provided/kg of diet; Mn: 60 mg; Fe: 30 mg; Zn: 50 mg; Cu: 5 mg; 1, 1.1 mg; Se: 0.1 mg, vitamin A, 8.800 IU; vitamin D_3, 2.200 IU; Vitamin E, 11 mg; Nikotin asit, 44 mg; Cal-D-Pan, 8.8 mg; Riboflavin 4.4 mg; Tiamin 2.5 mg; Vitamin B_{12}, 6.6 mg; folic acid, 1 mg; D-Biotine, 0.11 mg; Coline: 220 mg

$$\text{Yolk index (\%)} = \frac{\text{Yolk height}}{\text{Yolk diameter}} \times 100$$

$$\text{Albumen index (\%)} = \frac{\text{Albumen height}}{\text{Albumen length and width}} \times 100$$

$$\text{Haugh unit} = 100 \times \log (AH + 7.57\text{-}1.7 \times EW^{0.37})$$

Where:

AH = Albumen Height

EW = Egg Weight

Egg quality analyses were completed within 24 h of the eggs being laid

$$\text{Egg shell weight (\%)} = \frac{\text{Egg shell weight (g)}}{\text{Egg weight (g)}} \times 100$$

The eggs were subjected to determine characteristics of egg shell quality parameters (shell thickness and shell breaking strength) on all collected eggs produced during 2 consecutive days of per at the end of 28 days period during the experiment. Egg shell breaking strength was measured using a cantilever system by applying increased pressure to the broad pole of the shell using an instrument (Hisar terazi, Istanbul). Shell thickness was measured at 3 locations on the egg (air cell and any side of equator) using a micrometer (Mitutoyo, 0.01 mm, Japan)

$$\text{Egg shell weight (\%)} = \frac{\text{Egg shell weight (g)}}{\text{Egg weight (g)}} \times 100$$

Data were subjected to ANOVA by using General Linear Model procedure (GLM) in Minitab (2000). Duncan's multiple range tests were applied to separate means (Duncan, 1955). Statements of statistical significance are based on a probability of $p<0.05$.

RESULTS AND DISCUSSION

The different levels of dietary Ca and B as a main factor did not significantly effect on Initial Body Weight (IBW), Final Body Weight (FBW), Egg Production (EP), Feed Conversion Ratio (FCR), Specific Gravity (SG), Egg Shell Weight (ESW), Damaged Eggs (DE) and Egg Shell Breaking Strength (ESBS) except for Egg Weight (EW), Feed Intake (FI), Egg Mass (EM), Haugh Unit (HU) and Albumen Index (AI) (Table 2 and 3). There was no interaction effect of Ca and B levels on the parameters except for Egg Yolk Index (EYI), Egg Shape Index (ESI) and Egg Shell Thickness (EST) shown in Table 2 and 3 ($p< 0.01$; 0.05).

The dietary Ca and B levels and its interactions were not significantly effect on IBW, FBW, EP and FCR. The EW and FI decreased with additional B in diets. But, these parameters were not significantly affected by the dietary Ca level. The EM was significantly fed with the addition of 200 and 300 mg kg^{-1} B in diets lower than 0 mg kg^{-1} B in diet (Table 2). The EM, EW, EM, FI, FCR and egg grades (large-sized eggs and above and medium-sized eggs and below) were not influenced by dietary level of Ca (Keshavarz, 2003). Wilson and Ruszler (1998) reported that B supplementation with 50, 100 and 200 mg kg^{-1} increased the BW means when compared with the control in laying hens. Kurtoglu *et al.* (2002) reported that supplemental B in diets did not significantly effect on BW in laying hens. The dietary Ca and B levels and its interactions did not significantly effect on EP and FCR. The results of the present experiment are generally agreement with results of other experiment (Kurtoglu *et al.*, 2002; Rossi *et al.*, 1990; Wilson and Ruszler, 1995, 1996, 1998; Yesilbag and Eren, 2008). These results contrast with these of Qin and Klandorf (1991) that B supplementation at 100 mg kg^{-1} caused a significant decrease in EP in 1 of 2 experiments. In view of these observations, the toxic effect of B may be associated with age to the physiological status of the hen in addition to the level of dietary Ca. Growing chick and molted laying hens may possess a greater tolerance to B (Qin and Klandorf, 1991). The birds on the 3.34% Ca diet consumed 108.8 g feed/hen/day, which means that a Ca intake of 3.63 g/hen/day was sufficient to support performance and shell quality (Keshavarz, 2003). The EW decreased with additional B in diets ($p<0.01$) in the present study and these results are in agreement with results of some other experiments (Eren *et al.*, 2004; Wilson and Ruszler, 1998; Yesilbag and Eren, 2008), but Wilson and Ruszler (1996) found that 400 mg kg^{-1} dietary B concentration had no effect on EW in 81 weeks aged birds. The FI was depressed by 400 mg kg^{-1} B in laying hens (Wilson and Ruszler, 1996, 1998) and these results are agreement with the present study. Contrary to that B supplementation at level of 250 mg kg^{-1} had no effect on FI in laying hens (Eren *et al.*, 2004).

The SG, ESI, DE and ESBS were not influenced by the dietary treatments (Table 3). Kurtoglu *et al.* (2002) stated

Table 2: Effects of boron supplementation fed with low calcium to diet on performance

Ca (%)	B (ppm)	Initial body weight (g hen^{-1})	Final body weight (g hen^{-1})	Egg production (%)	Egg weight (g egg^{-1})	Feed intake (g/hen/day)	Feed conversion ratio (g feed/g egg)	Egg mass (g/hen/day)
4.0		1648±11.63	1702±16.24	90.86±0.73	67.99±0.47	126.0±1.40	2.05±0.02	61.67±0.59
3.5		1655±15.14	1656±21.09	91.20±1.11	68.18±0.42	126.1±1.60	2.04±0.03	62.18±0.85
	0	1676±21.80	1698±38.19	91.58±1.47	70.93±0.49^{A}	133.3±2.12^{A}	2.06±0.03	64.94±1.01^{A}
	100	1635±21.61	1693±31.00	92.89±1.12	66.69±0.39^{B}	124.6±2.08^{B}	2.01±0.02	61.95±0.80^{AB}
	200	1647±16.00	1662±10.34	89.26±1.62	67.70±0.42^{B}	123.5±1.17^{B}	2.06±0.05	60.44±1.21^{B}
	300	1647±15.79	1663±22.39	90.39±0.85	67.02±0.46^{B}	122.9±1.77^{B}	2.03±0.04	60.56±0.55^{B}
4.0	0	1655±23.90	1743±29.79	91.95±1.33	71.02±0.84	133.5±2.25	2.05±0.03	65.28±0.99
4.0	100	1667±29.47	1731±44.16	93.03±0.76	67.13±0.62	124.1±2.59	1.99±0.02	62.45±0.77
4.0	200	1630±13.52	1663±10.67	88.04±1.86	67.43±0.55	121.9±1.70	2.06±0.02	59.32±0.92
4.0	300	1640±25.98	1672±31.12	90.42±1.22	66.39±0.67	124.5±2.79	2.08±0.06	59.99±0.44
3.5	0	1697±36.69	1653±68.90	91.21±2.74	70.83±0.57	133.1±3.79	2.07±0.05	64.59±1.84
3.5	100	1603±28.45	1655±41.60	92.89±2.21	66.26±0.48	125.0±3.46	2.04±0.04	61.46±1.42
3.5	200	1665±28.73	1661±18.68	89.26±2.73	67.96±0.66	125.1±1.48	2.06±0.01	61.55±2.25
3.5	300	1654±19.68	1654±34.30	90.36±1.29	67.66±0.57	121.2±2.22	1.99±0.04	61.13±1.01

$^{A, B}$Values in columns are statisticaly different; $p<0.01$

Table 3: Effects of boron supplementation fed with low calcium to diet on egg quality characteristics

Ca (%)	B (ppm)	Specific gravity	Haugh unit	Albumen index (%)	Egg yolk index (%)	Egg shape index (%)	Egg shell weight (%)	Egg shell thickness (mm×10^{-2})	Damaged eggs (%)	Egg shell breaking strength (N)
4.0		1.084±0.001	88.32±0.77^{b}	4.68±0.11^{b}	44.32±0.17	75.07±0.19	9.02±0.07	39.5±0.72	2.83±0.45	25.63±1.35
3.5		1.082±0.001	91.30±0.52^{a}	5.09±0.08^{a}	45.44±0.17	74.51±0.29	8.86±0.08	38.2±0.61	2.87±0.62	24.71±1.13
	0	1.082±0.001	91.16±0.96	4.96±0.14	44.69±0.26	74.72±0.37	8.83±0.11	39.6±0.99	1.66±0.42	24.97±1.23
	100	1.083±0.001	90.15±0.85	4.95±0.13	45.04±0.25	75.25±0.32	8.87±0.10	38.0±0.55	3.51±1.12	25.53±1.28
	200	1.084±0.001	89.57±1.14	4.90±0.18	44.78±0.39	75.12±0.33	9.08±0.08	39.4±0.48	3.20±0.70	25.65±1.36
	300	1.083±0.001	88.36±1.05	4.97±0.14	45.16±0.19	74.07±0.33	8.98±0.15	38.5±0.58	3.02±0.62	24.53±1.20
4.0	0	1.084±0.001	91.13±1.62	5.00±0.24	44.52±0.38^{BC}	75.76±0.32^{A}	8.99±0.15	41.3±0.81^{a}	1.99±0.61	26.71±0.96
4.0	100	1.083±0.001	88.97±1.31	4.79±0.20	44.40±0.33^{BC}	74.87±0.47^{AB}	8.88±0.13	38.4±0.53^{bc}	3.21±1.49	25.23±1.24
4.0	200	1.085±0.001	86.74±1.48	4.43±0.22	43.60±0.23^{C}	74.79±0.35^{AB}	9.17±0.10	40.0±0.41^{ab}	2.80±0.37	26.81±1.79
4.0	300	1.083±0.001	86.43±1.48	4.49±0.21	45.04±0.22^{AB}	74.87±0.35^{AB}	9.02±0.21	38.4±0.52^{bc}	3.31±0.88	23.76±1.23
3.5	0	1.081±0.001	91.18±1.16	4.92±0.17	44.86±0.38^{AB}	73.69±0.37^{B}	8.67±0.15	37.8±0.69^{c}	1.33±0.60	23.22±1.16
3.5	100	1.083±0.001	91.32±0.99	5.10±0.14	45.68±0.19^{AB}	75.62±0.44^{A}	8.87±0.15	37.5±0.57^{c}	3.82±1.79	25.83±1.41
3.5	200	1.083±0.001	92.41±0.90	5.37±0.14	45.96±0.36^{A}	75.45±0.56^{A}	8.98±0.12	38.9±0.46^{bc}	3.60±1.39	24.48±0.61
3.5	300	1.082±0.002	90.28±1.14	4.97±0.17	45.27±0.31^{AB}	73.26±0.37^{B}	8.94±0.23	38.5±0.68^{bc}	2.73±0.93	25.29±1.19

$^{A, B}$Values in columns are statisticaly different; $p<0.01$; $^{a, b}$Values in columns are statisticaly different; $p<0.05$

that SG was not affected by B supplementation in laying diets. In some other researches reported that B supplementation to 400 mg kg^{-1} did not cause negative effect on SG in broiler breeder hens and laying hens (Eren *et al.*, 2004; Qin and Klandorf, 1991; Rossi *et al.*, 1993; Wilson and Ruszler, 1996). In the present study, the dietary B levels did not effect on HU and AI, but these parameters were significantly influenced by dietary Ca levels. The HU and AI decreased significantly with control diet (4.0% Ca). Similar result was reported by Eren *et al.* (2004). The EYI was slightly increased by B supplementation and the highest EYI was observed fed with containing 3.5% Ca and 200 mg kg^{-1} B in group. Similar results were reported by and Eren *et al.* (2004) and Qin and Klandorf (1991). Also, same researchers indicated that the values of ESI did not change in all treatments. Similarly, the ESI was not influenced by Ca level (Keshavarz, 2003). Kurtoglu *et al.* (2002) have reported that the DE was influenced by B supplementation on the period of 60-90 days. During the present experiment, despite no significant differences, the DE ratio decreased in all groups when compared with the control group. Obtaining the low DE ratio from the experiments used with laying hens has profitable effect in poultry industry. It is suggested that poor shell quality is associated with a loss in the ability to absorb minerals of aged laying hens. The EST affected by dietary B and Ca levels in the present study. The highest EST was observed fed with containing 4.0% Ca and 0 mg kg^{-1} B in group. Similar results were reported by Eren *et al.* (2004) and Yesilbag and Eren (2008). This positive effect on EST in thought to results from the effects of B as a nutritional factor in Ca metabolism and utilisation in aged laying hens (Nielsen, 1992). In the present study, the ESBS was not influenced by the dietary B and Ca. Eren *et al.* (2004) found that 0-400 mg kg^{-1} B supplementation had no effect on ESBS in laying hens (22 and 28 weeks), but Yesilbag and Eren (2008) stated that ESBS increased by B supplementation in aged laying hens.

CONCLUSION

In this study, the B supplementation was negatively effect on EW, FI and EM but the other performance parameters were not influenced of B supplementation in molted laying hens. The EYI, ESP and EST were affected by B supplementation, however, these effects were not observed clearly. It is concluded that the molted laying hens to the diet were not added B when the laying hens consumed adequately feed and Ca.

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