

## The Effects of Dietary Electrolyte Balance on the Performance and Eggshell Quality in the Early Laying Period

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**Abstract:** This study was designed to evaluate the effects of Dietary Electrolyte Balance (DEB =  $\text{Na}^+ + \text{K}^+ - \text{Cl}^-$ ) on the performance and eggshell quality of layers in their early laying period. Two hundred fifty six Hy-Line strain laying hens, 24-34 weeks of age, were randomly assigned into four treatment. The DEB levels were 0, 120, 240 and 360 mEq  $\text{kg}^{-1}$  and were obtained by addition of NaCl,  $\text{NaHCO}_3$ ,  $\text{KHCO}_3$  and  $\text{NH}_4\text{Cl}$  as needed. Egg production, feed intake, egg mass and Feed Conversion Ratio (FCR) were not significantly affected by any of the treatments, whereas eggshell quality was significantly affected ( $p < 0.05$ ). The shell specific gravity, shell weight, shell thickness, shell ash and shell weight/surface area were increased with increasing DEB from 120-360 mEq  $\text{kg}^{-1}$  relative to the control diet. It was concluded that high rate DEB (360 mEq  $\text{kg}^{-1}$ ) can improve eggshell quality in the early laying period.

**Key words:** Electrolyte balance, eggshell, performance, laying hens

### INTRODUCTION

"Electrolytes" are compounds, which are dissolved and dissociated into positively and negatively ions in a suitable medium. This term, commonly used in animal nutrition primarily refers to sodium ( $\text{Na}_+$ ), potassium ( $\text{K}_+$ ) and chloride ( $\text{Cl}_-$ ) (Hooze, 1995). Dietary Electrolyte Balance (DEB) is often described by simple formulas expressed as mEq  $\text{kg}^{-1}$  of diet such  $[\text{Na}^+ + \text{K}^+ - \text{Cl}^-]$  (Cohen and Hurwitz, 1974). These monovalent electrolytes have important role in maintaining of body acid-base homeostasis (NRC, 1994). These minerals are essential for synthesis of tissue proteins, maintenance of intracellular and extracellular homeostasis and electric potential cell membranes, enzymatic reactions, osmotic pressure and acid-base balance (Borges *et al.*, 2003).

Eggshell quality is of major importance to the egg industry worldwide and may be affected by the DEB (Hughes, 1988). The proper dietary electrolyte balance should not only maintain acid-base homeostasis, but also achieve optimal egg production and feed conversion ratio (Hughes, 1988). In metabolism of poultry, particular in layers, the Na and  $\text{H}_2\text{CO}_3$  have a distinguished importance in egg productivity and shell formation.

The formation of the eggshell in poultry is affected by the acid-base balance in blood because the acid-base rate of the blood is a restrictive factor for the accumulation of  $\text{CaCO}_3$  in eggshell (Mehner and Harlfiel, 1983). Mongin (1968) has noted that the first restrictive factor of the shell formation was the Ca and the second factor was the carbonate ions so the breakage, which was observed in eggshell in hot weather, had been caused by a certain decrease in blood  $\text{CO}_2$  level depending on the increase in respiration speed.

According to Sauveur and Mongin (1978) no effect on shell weight or surface was obtained when electrolyte balance evaluated by  $[\text{Na}^+] + [\text{K}^+] - [\text{Cl}^-]$  was comprised between 160-360 mEq  $\text{kg}^{-1}$ . On the contrary, Hamilton and Thompson (1980) reported no significant alteration of eggshell quality when electrolyte balance was lower than 330 mEq  $\text{kg}^{-1}$  or higher than 620 mEq  $\text{kg}^{-1}$ , but they observed the reduction of the rate of lay and of feed intake. Furthermore, in their experiment, a low electrolyte balance depressed blood pH,  $\text{HCO}_3^-$  concentrations and shell quality. Consequently, we postulate that laying hens need dietary supplementation by alkaline salts through continuous provision of  $\text{NaHCO}_3$  or  $\text{KHCO}_3$  into diets, for compensating lay induced metabolic acidosis and maintaining eggshell quality.

Although, the effects of different Dietary Electrolyte Balance on eggshell quality and laying performance during the peak production period have been investigated intensively in layers, data on its impact during the early laying period are limited. The objective of this study, was to evaluate the effects of DEB various levels on egg production, egg quality parameters of hens during the early laying period.

## MATERIALS AND METHODS

**Hens and dietary treatments:** Two hundred and fifty six 24 weeks old Hy-Line W36 laying hens obtained from a Company, were used in this study. These 256 hens were divided into 16 groups (each group containing 16 laying hens) in completely random design. Treatments were in a completely randomized design. The rations (Table 1) prepared for different experimental groups (n=64) are summarized below:

**Group 1:** Basal diet containing dietary electrolyte balance 0 mEq kg<sup>-1</sup>.

**Group 2:** Basal diet containing dietary electrolyte balance 120 mEq kg<sup>-1</sup>.

**Group 3:** Basal diet containing dietary electrolyte balance 240 mEq kg<sup>-1</sup>.

**Group 4:** Basal diet containing dietary electrolyte balance 360 mEq kg<sup>-1</sup>.

All diets were in the meal form and based on corn and soybean meal. The diets were formulated to be isonitrogenous (14%) and isocaloric (2800 Kcal kg<sup>-1</sup>) According the NRC (1994) as fed basis. Treatment groups were formed according to Dietary Electrolyte Balance (DEB). Dietary Electrolyte Balance was calculated in mEq according to mongin's formula:  $DEB = [Na^+] + [K^+] - [Cl^-]$ . The dietary electrolyte balance differents were obtained with the inclusion of NaCl, NaHCO<sub>3</sub>, KHCO<sub>3</sub> and NH<sub>4</sub>Cl (Table 1). Respective amounts of dietary electrolytes were first blended thoroughly with small amount of dicalcium phosphate then was mixed with a larger amount of the basal diet until the total amount of the respective diets were homogeneously mixed.

Experimental feeds were supplied to the birds after the 24 week old to 34 week of age. They were reared in cages under conditions with access to feed and water ad libitum and with a constant 16 h lighting daily.

Table 1: Composition of laying hen diets (as fed basis)

	DEB			
	0	120	240	360
Ingredient composition (kg t <sup>-1</sup> )				
Corn grain	400	400	400	400
Wheat grain	306.3	306.3	306.3	306.3
Soybean meal	166.5	166.5	166.5	166.5
Oil	14.6	14.6	14.6	14.6
Oyster shell	70.7	70.7	70.7	70.7
Bone meal	17.4	17.4	17.4	17.4
Salt	2.5	2.5	2.5	2.5
Vitamin premix	2.5	2.5	2.5	2.5
Mineral premix	2.5	2.5	2.5	2.5
DL- Methionine (990 g kg <sup>-1</sup> )	.5	.5	.5	.5
Inert	8.4	11.4	6.4	0
NH <sub>4</sub> Cl	8	3	1	0
KHCO <sub>3</sub>	0	0	0	1.7
NaHCO <sub>3</sub>	0	2	9	14.7
Chemical composition (g kg <sup>-1</sup> )				
Metabolizable energy(kcal kg <sup>-1</sup> )	2800	2800		
Crude protein	140	140	140	140
Calcium	32.8	32.8	32.8	32.8
Avail. Phosphorus	3.1	3.1	3.1	3.1
Potassium	5.9	5.9	5.9	5.9
Chloride	7.5	7.5	7.5	7.5
Sodium	1.4	1.4	1.4	1.4
Lysine	6.3	6.3	6.3	6.3
Methionine+ cystine	5.9	5.9	5.9	5.9
Methionine	3.1	3.1	3.1	3.1
Tryptophan	1.8	1.8	1.8	1.8

<sup>1</sup>Dry matter content 900 g kg<sup>-1</sup>. <sup>2</sup>DEB 0 (Dietary Electrolyte Balance = 0), DEB120 (Dietary Electrolyte Balance = 120), DEB 240 (Dietary Electrolyte Balance = 240), DEB360 (Dietary Electrolyte Balance = 360). <sup>3</sup>Premix supplied per kg of diet: 9000 IU vitamin A, 1.78 mg vitamin B<sub>1</sub>, 6.6 mg vitamin B<sub>2</sub>, 30 mg niacin, 10 mg pantothenic acid, 3 mg vitamin B<sub>6</sub>, 0.15 mg biotin, 1500 mg choline, 0.015 mg vitamin B<sub>12</sub>, 2000 IU vitamin D, 18 IU vitamin E, 2 mg vitamin K<sub>3</sub>. <sup>4</sup>Premix supplied per kg of diet: 10 mg Cu, 0.99 mg I, 50 mg Fe, 100 mg Mn. 0.08 mg Se, 100 mg Zn. <sup>5</sup>All values were calculated from NRC values (1994)

**Chemical analysis and measurements of hens performance and eggshell quality:** Prior feeding, laboratory assays were conducted in each diets for sodium, potassium and chloride. The sodium and potassium were determined by Flame spectrophotometer. The Chloride in feed was determined by titration (Lacroix *et al.*, 1970). Assay of these elements determined required values of NaCl, NaHCO<sub>3</sub>, KHCO<sub>3</sub> and NH<sub>4</sub>Cl. Quantities were adjusted to provide the wanted DEB according to assay groups. Analyses of the drinking water revealed very low quantities of sodium, potassium and chloride. Therefore, the intake of these elements via drinking water was not considered in the calculations.

The birds were weighted at the commencement (24 week of age) and the end (34 week of age) of the trial.

Egg production (%hen-day) and egg weight (g) were recorded daily. Daily production was determined on a shelled egg weight basis. Feed intake (g/hen/day) was recorded weekly. Feed Conversion Ratio (FCR) was calculated as gram feed consumption per day per hen divided by gram egg mass per day per hen.

Every 25 d, 4 eggs per replicate (16 eggs per treatment) were individually weighed and the egg specific gravity ( $\text{g mL}^{-1}$ ) was also evaluated. During the 25 day periods, eggs from each treatment group (16 eggs) were taken to determine eggshell quality parameters.

Eggshell weight and shell thickness were determined by randomly collecting 4 eggs from each replicate. After the eggs were broken the shells were washed and dried in room temperature for the determination of shell weight. The shell thickness was measured with a micrometer gauge (Measure, 24 21/1 type) on three part of shell from the eqator of each egg. These measurements were pooled.

The shell weigh/area also recorded and shell ash was determined after drying at room temperature for 3 days.

**Statistical analysis:** Data were analyzed by the General Linear models procedure of SAS Institute 1985. Means for treatments showing significant differences in the analysis of variance were compared using Duncan's multiple range tests. All statements of significance are based on the probability level of 0.05.

## RESULTS AND DISCUSSION

For the total experiment period, hen day egg production, feed intake, egg weight, egg mass and feed conversion ratio were not statistically different between groups (Table 2).

The data of eggshell quality parameters presented in Table 3 show that the specific gravity, eggshell weight, eggshell thickness, eggshell ash and shell weight/surface area were affected in various treatments during the experiment. By contrast, eggshell specific gravity (Table 3) were reduced in group 1 ( $\text{DEB} = 0 \text{ mEq kg}^{-1}$ ) and group 3 ( $\text{DEB} = 240 \text{ mEq kg}^{-1}$ )

( $p < 0.05$ ) but differences between group 2 ( $\text{DEB} = 120 \text{ mEq kg}^{-1}$ ) and group 4 ( $\text{DEB} = 360 \text{ mEq kg}^{-1}$ ) was not statistically significant ( $p > 0.05$ ). The shell weight were reduced in group 1 ( $\text{DEB} = 0 \text{ mEq kg}^{-1}$ ) and group 3 ( $\text{DEB} = 240 \text{ mEq kg}^{-1}$ ) ( $p < 0.05$ ) but differences between group 2 ( $\text{DEB} = 120 \text{ mEq kg}^{-1}$ ) and group 4 ( $\text{DEB} = 360 \text{ mEq kg}^{-1}$ ) were not statistically significant for the total experimental period. The shell thickness were affected in different experimental groups. This trait were reduced in all groupsexcept to group 4 ( $\text{DEB} = 360 \text{ mEq kg}^{-1}$ ) but differences between groups 1, 2 and 3 ( $\text{DEB} = 0 \text{ mEq kg}^{-1}$ ,  $\text{DEB} = 120 \text{ mEq kg}^{-1}$  and  $\text{DEB} = 240 \text{ mEq kg}^{-1}$ ) were not statistically significant. The eggshell ash decreased in groups 1, 2 and 3 ( $\text{DEB} = 0 \text{ mEq kg}^{-1}$  and  $\text{DEB} = 120 \text{ mEq kg}^{-1}$  and  $\text{DEB} = 240 \text{ mEq kg}^{-1}$ ) ( $p > 0.05$ ) but did not reduce in group 4 ( $\text{DEB} = 360 \text{ mEq kg}^{-1}$ ). The shell weight/surface area were reduced in group 1 and 3 ( $\text{DEB} = 0 \text{ mEq kg}^{-1}$  and  $\text{DEB} = 240 \text{ mEq kg}^{-1}$ ) ( $p > 0.05$ ) but differences between group 2 ( $\text{DEB} = 120 \text{ mEq kg}^{-1}$ ) and group 4 ( $\text{DEB} = 360 \text{ mEq kg}^{-1}$ ) were not statistically significant for the total experimental period.

Egg production rate decreases and egg weight increases as age advances (Bustang and Elwinger, 1987; Summers and Lesson, 1983). Egg quality and composition also change in accordance with level of production and age of layer. As age advances, proportion of yolk increases, where as proportions of albumen and shell thickness decrease (Akbar *et al.*, 1983; Fletcher *et al.*, 1983).

The results of this experiment showed that the different level of DEB in diet do not statistically affected on the productivity characteristics in layer. The addition of choloride ammonium to the experimental diets due to decrease of DEB to 0 and 120  $\text{mEq kg}^{-1}$  of diet have not significant effect on egg production. The percentage of egg production in 1-4 different treatments were 87.3,

Table 2: Effects of electrolyte balance on laying performance

Treatments DEB( $\text{mEq kg}^{-1}$ )	Egg production (%)	Feed intake (g/hen/day)	Egg weight (g egg $^{-1}$ )	Egg mass (g/hen/day)	FCR (g feed/egg $^{-1}$ )
0	87.3 <sup>a</sup>	101.28 <sup>a</sup>	56.49 <sup>a</sup>	48 <sup>a</sup>	2.07 <sup>a</sup>
120	85.85 <sup>a</sup>	103.65 <sup>a</sup>	56.46 <sup>a</sup>	48.47 <sup>a</sup>	2.16 <sup>a</sup>
240	89.47 <sup>a</sup>	102.9 <sup>a</sup>	56.64 <sup>a</sup>	50.69 <sup>a</sup>	2.04 <sup>a</sup>
360	84.29 <sup>a</sup>	101 <sup>a</sup>	56.94 <sup>a</sup>	48.08 <sup>a</sup>	2.15 <sup>a</sup>
Pooled mse	$\pm 1.82$	$\pm 1.22$	$\pm 27$	$\pm 1.14$	$\pm .07$

Means within each column with same superscripts are not significantly different ( $p < 0.05$ ) <sup>a</sup>

Table 3: Effects of electrolyte balance on eggshell quality

Treatments DEB( $\text{mEq kg}^{-1}$ )	Specific gravity	Eggshell weight (g)	Eggshell thickness (mm)	Egg shell ash (%)	Shell weight/ surface area( $\text{mg cm}^{-2}$ )
0	1.078 <sup>b</sup>	4.91 <sup>c</sup>	0.262 <sup>c</sup>	79.88 <sup>b</sup>	70.9 <sup>c</sup>
120	1.083 <sup>a</sup>	5.38 <sup>a</sup>	0.276 <sup>ab</sup>	83.56 <sup>ab</sup>	76.8 <sup>a</sup>
240	1.08 <sup>ab</sup>	5.02 <sup>bc</sup>	0.265 <sup>bc</sup>	79.19 <sup>b</sup>	71.71 <sup>bc</sup>
360	1.082 <sup>a</sup>	5.30 <sup>ab</sup>	0.288 <sup>a</sup>	89.36 <sup>a</sup>	75.94 <sup>ab</sup>
Pooled Mse	$\pm .0003$	$\pm .099$	$\pm .0043$	$\pm 2.75$	$\pm 1.89$

Means within each column with different superscripts are significantly different ( $p < 0.05$ ) <sup>a-b</sup>

85.85, 89.47 and 84.29, respectively. Furthermore, feed intake and feed conversion ratio were not significantly different between groups. The average of feed intake were between 101-103.65 in different treatment. Nizamettin *et al.* (2005) formulated different Dietary Electrolyte Balances (DEB, 176, 204, 225 and 242 mEq kg<sup>-1</sup>) and fed to laying hens from 22-30 weeks of age. They reported that egg production, egg shell quality, feed intake, egg mass and feed conversion ratio were not significantly affected by the diets of various DEB.

The results of experiment concerning the effects of electrolyte balance on egg shell quality include egg specific gravity, eggshell weight, eggshell thickness, eggshell ash and shell weight/surface area, were significantly affected by dietary treatments.

Egg specific gravity, eggshell weight, Shell weight/surface area, eggshell thickness, eggshell ash and shell weight/surface area are markedly increased in the group 4 (DEB = 360 mEq kg<sup>-1</sup>). Our findings agree with the obtained results by Gezen (2005) that reported a moderate DEB (256 MEq kg<sup>-1</sup>) can improve eggshell quality. Hughes (1988) and Cohen and Hurwitz (1974) also reported positive results by increasing DEB with its use at the age of 70 week. Chen and Balnave (2001) reported an optimal activity of carbonate anhydrase that plays an important role in eggshell formation in slightly alkaline medium, Moreover an excessive Chloride intake limited calcium transports to shell gland lumen. However, our finding's show that high DEB positively affected the eggshell quality, are consistent with these observations. Using young hens (32 week of age), Hughes (1988) observed curvilinear increase in shell thickness as DEB increase from about 150 mEq kg<sup>-1</sup>.

In contrast, Nizamettin *et al.* (2005) suggested that there was no need for the supplementation of sodium bicarbonate and or potassium carbonate into practical laying hens diets during peak production. Likewise, Austic and Keshavarz (1988) also, reported a weak relationship ( $R^2 = 0/2$ ) between DEB and shell thickness. Sauveur and Mongin (1978) reported no effect of DEB over the range 160-360 mEq kg<sup>-1</sup> on shell weight/surface area was observed. Similarly, Hamilton and Thompson (1980) reported a lack of response to DEB in terms of shell quality in hens of 68 weeks of age. They only found that the rate of lay and feed intake significantly depressed by 330 and 620 mEq kg<sup>-1</sup> which were found to be extreme DEB values and not commonly practiced with the commercial diets.

## CONCLUSION

Increasing DEB to 360 mEq kg<sup>-1</sup> in early laying period can improve eggshell quality of laying.

Consequently, during establishments of layer hen diets, minimum requirements for each mineral given by NRC should be provided in a first attempt, then the dietary electrolyte balance should be adjusted during different production periods.

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