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# The Effect of Dietary Electrolyte Balance on Growth, Tibia Ash and Some Blood Serum Electrolytes in Young Pullets

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Abstract: This study was conducted to evaluate the effect of increasing dietary electrolyte balance [DEB;Na+K-Cl, miliequivalents (mEq) Kg<sup>-1</sup> on growth performance, tibia ash and Ca, Na, K and Cl contents of blood serum in young pullets. A corn-soybean meal based mash diet containing 187 mEq Kg<sup>-1</sup> (Na+K-Cl) was formulated to supply the nutrient requirements of young pullets. DEB of basal diet was increased as much as 43, 64 and 97 mEq Kg<sup>-1</sup> by substitution of sodium bicarbonate, potassium bicarbonate or both of them in part of corn and four experimental diets containing 187, 230, 251 and 284 mEq Kg<sup>-1</sup> (Na+K-Cl) were obtained. Young pullets (n=160) were used from 7 to 35 d of age, in a completely randomized design with four dietary treatments having four replicate cages (10 pullets) each to evaluate different levels of DEB. Weekly feed intake, body weight gain and feed conversion ratio were not significantly affected by dietary treatments. The same results were obtained in growth period of 7 to 35 days. Tibia ash and its calcium content varied between 46.5 to 48.9 and 34.4 to 37.9 percent, respectively and were not significantly affected by different levels of DEB. Among blood serum electrolytes, only K and Cl were significantly affected by DEB (P<0.01). Diet containing 251 mEq Kg<sup>-1</sup> (Na+K-Cl) had the lowest value of blood serum K concentration and concentration of blood serum Cl increased with increasing of DEB.

Key words: Dietary electrolyte balance, tibia ash, serum electrolytes, pullets

#### Introduction

Birds have a minimum requirements for Na, K and CI and this monovalent electrolytes are provided by natural ingredients and electrolyte salts. These monovalent electrolytes have important role in maintaining of body acid-base homeostasis (NRC, 1994; Leeson and Summers, 2001). The lack of proper acid-base balance lead to the metabolic pathways do not work optimally and sources of electrolyte are used in body homeostasis processes instead of growth activities (Mongin, 1981). Mongin (1981) proved that dietary (Na+K-Cl) content could alter body acid-base balance and this physiologically altering effect could be ingested (Na+K-CI)=excreted (anionexpressed as: cation)+endogenous acid(H<sup>+</sup>) + base extracellular fluid(BE<sub>erf</sub>). This term is correct when intake of other minerals are assumed constant. Relatively more papers have been released on the effect of dietary electrolyte balance on broilers performance in past decades. Melliere and Forbes (1966) reported that anion-cation ratio of diet has significant effect on growth and feed consumption of young chickens. Mongin and Sauveur (1977) showed that optimum performance in broiler chickens at 28 d of age could be achieved with 250 mEq Kg<sup>-1</sup> of (Na+K-Cl). Roch et al. (1999) also reported that the best weight gain and homeostasis were obtained in

broiler chickens with diet containing the same value of mEq Kg<sup>-1</sup> of (Na+K-Cl). McNab et al. (1981) studied the effect of four different levels of (Na+K-Cl) ranging from 108 to 402 mEq Kg-1 on the general performance and incidence of tibia dyschondroplasia of broiler chickens from 1 to 28 d of age. Their results did not support the findings of Mongin and Sauveur (1977). Pesti et al. (1997) also studied the effect of four and six levels of (Na+K-CI) on performance of broiler chickens from 0 to 18 and 0 to 42 d of age, respectively and supported the findings of McNab et al. (1981). Recently, Borges et al. (2003a) reported that the best weight gain and feed conversion ratio in broiler chickens under moderately high temperature and relative humidity condition could be achieved when starter and grower diets contained 240 mEq Kg<sup>-1</sup> of (Na+K-Cl). In other study, they also showed that the same value is the best for broiler chickens under thermoneutral condition from 0 to 42 days of age (Borges et al., 2003b). However, pullets are genetically different from broiler chickens and they have also different nutrient requirements. To our best of knowledge there are very limited data (if not) on the effect of dietary electrolyte balance on growth of young pullets, therefore this research was designed to study the effects of dietary electrolyte balance on growth and tibia development of young pullets.

Table 1: Ingredients and chemical composition of the experimental diets

Ingredients %	Dietary cation- anion balance					
	 187	230	251	284		
Corn	66.06	65.83	65.71	65.48		
Soybean meal	21.01	21.01	21.01	21.01		
Wheat bran	4.48	4.48	4.48	4.48		
Fish meal	3	3	3	3		
Alfalfa meal	2	2	2	2		
Limestone	1.1	1.1	1.1	1.1		
Dicalcium phosphate	1.09	1.09	1.09	1.09		
Salt	0.28	0.28	0.28	0.28		
Potassium bicarbonate <sup>1</sup>	0	0.23	0	0.23		
Sodium bicarbonate <sup>1</sup>	0	0	0.35	0.35		
Mineral premix <sup>a</sup>	0.25	0.25	0.25	0.25		
Vitamin premix <sup>b</sup>	0.25	0.025	0.25	0.25		
L-Lysine HCI	0.12	0.12	0.12	0.12		
Total	100	100	100	100		
Calculated Composition						
AME <sub>n</sub> Kcal/kg	2850	2850	2850	2850		
CP %	18	18	18	18		
TSAA <sup>2</sup> %	1.56	1.56	1.56	1.56		
Lys %	0.93	0.93	0.93	0.93		
Ca%	0.9	0.9	0.9	0.9		
$P_{av}$	0.4	0.4	0.4	0.4		
CI	0.24	0.24	0.24	0.24		
K	0.74	0.87	0.74	0.87		
Na	0.15	0.15	0.30	0.30		
Na+K-Cl mEq Kg <sup>-1</sup>	187	230	251	284		
Analyzed Composition						
DM %	92	92	90	92		
CP %	18.2	17.9	17.6	17.6		
Ash	5.6	5.4	6.2	6.2		
Na+K-Cl mEq Kg <sup>-1</sup>	177	217	263	291		

<sup>1</sup>Laboratory grade from Merk Company. <sup>2</sup>Total sulfur amino acids. <sup>8</sup>Supplied per kilogram of diet: 6050  $\mu$ g vitamin A (retinyl acetate +retinyl palmitate), 55  $\mu$ g vitamin D<sub>3</sub>, 22.05  $\mu$ g vitamin E (DL-tocopheryl acetate), 2.0 mg K<sub>3</sub>, 5 mg B<sub>1</sub>, 6 mg B<sub>2</sub>, 60mg B<sub>3</sub>, 4 mg B<sub>6</sub>, 0.02 mg B<sub>12</sub>, 10 mg Panthotenic acid, 6.0 mg Folic acid, .015 mg Biotin and 0.625 mg ethoxyquin. <sup>b</sup>Supplied per kilogram of diet: 500 mg CaCO<sub>3</sub>, 80 mg Fe, 80 mg Zn, 80 mg Mn, 10 mg Cu, 0.8 mg I and 0.3 mg Se.

### **Materials and Methods**

One hundred and sixty young pullets, with the individual initial weight of 60 grams, were housed in 16 cages having 10 pullets in each. Pullets were kept under management and vaccination schedule similar to those used in commercial pullets operation. A corn-soybean meal based mash diet was formulated to supply the nutrient requirements for 0-35 d of age according to NRC (1994). Basal diet contained 187 mEq Kg-1 of (Na+K-Cl) (Table 1). Three other diets containing 230, 251 and 284 mEq Kg<sup>-1</sup> of (Na+K-Cl) were produced by substitution of sodium bicarbonate, potassium bicarbonate or both of them in part of corn in basal diet (Table 1). The diets were subjected to analysis for DM, CP, and ash according to AOAC (1984). Na, K and Cl also were measured according to flame photometry and titration standard methods. The four experimental diets,

in four replicate cages each, were fed ad-libitum from 7 to 35 d of age. Fresh water also provided as free. Feed intake and body weight gain were recorded weekly. By using of sterilized needles, the wing veins of three pullets in each cage were punctured and their venous blood samples collected. All blood serum samples were subjected to analysis for Ca, Na, K and Cl in a reference clinical veterinary lab. Also three other birds from each cage were killed by cervical dislocation and the left tibias were separated to evaluate tibia ash and calcium. The left tibias were cleaned up from attached tissues and cartilage cups were also removed. The tibias were extracted in 90% ethanol for 48 hours and extraction was lasted for more than 48 hours by dried diethyl ether. Fat-free tibia were weighed after drying at 90 degrees of centigrade in oven. The dried fat-free tibias ashed at 590 degrees of centigrade in a furnace for 20 hours.

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Table 2: The effect of different levels of dietary cation -anion balance on weekly performance

	Dietary cation- anion balance							
	187	230	251	284	SE			
	Week- 2							
Body weight (g)	88.9	89.1	88.2	85	1.53			
Feed intake (g)	80.8	93.3	95.2	80.4	6.7			
Weight gain (g)	28.9	29.1	28.2	24.9	6.7			
Feed conversion ratio	2.81	3.21	3.37	3.22	0.18			
	Week- 3							
Body weight (g)	142	137.9	142	139.3	3.16			
Feed intake (g)	137.9	144.1	137.4	143.3	2.91			
Weight gain (g)	53.1	48.4	53	48.4	3.08			
Feed conversion ratio	2.6	2.97	2.6	2.66	0.26			
	Week- 4							
Body weight (g)	199.9	201.3	198.9	197.4	2.84			
Feed intake (g)	183.4	183.6	182.6	179.8	4.14			
Weight gain (g)	57.9	61.6	57.4	58.1	4.28			
Feed conversion ratio	3.17	3.11	3.19	3.10	0.18			
	Week- 5							
Body weight (g)	263.3	267.3	267	263.3	4.87			
Feed intake (g)	216.5	231	229.6	217.5	6.33			
Weight gain (g)	63.4	66	68.1	65.9	3.07			
Feed conversion ratio	3.45	3.52	3.38	3.3	0.13			

Table 3: The effect of different levels of dietary cation –anion balance on the performance from 7 to 35 d of age, tibia ash, tibia ash calcium and concentration of Ca, Na, K and Cl in blood serum of pullets

	Dietary cation- anion balance				
	187	230	251	284	SE
Feed intake (g)	618.5	652.5	644.9	621	15.92
Weight gain (g)	203.3	205.1	206.7	202.0	4.79
Feed conversion ratio	3.04	3.18	3.12	3.06	0.05
Tibia bone					
Tibia ash <sup>*</sup> %	46.67	48.03	48.93	46.54	0.99
Ca %	36.7	34.43	34.67	37.96	0.65
Blood Serum					
Ca (mg/100ml)	12.9	13.3	13.1	12.8	058
Na (mmol/l	142	144.3	141.8	143.1	0.66
K (mmol/l)	4.9 <sup>a</sup>	4.4 <sup>ab</sup>	3.9 <sup>b</sup>	4.4 <sup>ab a</sup>	0.19
Cl (mmol/l)	103.7 <sup>b</sup>	109.8°	111.2°	111.4°	1.35

<sup>\*</sup>Based on dried fat- free bone. <sup>a-b</sup> show significant differences in each row.

Percentage of fat-free tibia ash was obtained by dividing the weight of tibia ash to the weight of dried fat-free tibia (Zhang and Coon, 1977). Calcium content of tibia ash also was determined by titration method. The data were analyzed by method of analysis of variance using GLM procedure of SAS. Comparison of means was made by Duncan's Multiple Range Test (SAS Institute, 1997).

## **Results and Discussion**

Weekly feed intake, body weight gain and feed conversion ratio for each of dietary treatments are presented in Table 2. As shown in the table, no significant effect of increasing DEB, 187 to 284 mEq

Kg<sup>-1</sup>, were found on performance of pullets. Also in growth period of 7 to 35 d of age, the different levels of DEB did not have any significant effect on feed intake, body weight gain and feed conversion ratio (Table 3). We could not find any similar study to compare our results with them. However, findings of the present study on performance of pullets are in agreement with the reports of McNab *et al.* (1981), Hulan *et al.* (1985) and Pesti *et al.* (1999) in broiler chickens. With considering the results of these researchers and the findings of present paper, it seems that the birds are able to adjust their body acid-base balance in a relatively wide range of DEB. However, Mongin and Sauveur (1977), Roch *et al.* 

(1999) and Borges *et al.* (2003a, 2003b) strongly recommended 240 to 250 mEq Kg<sup>-1</sup> of DEB for obtaining the best performance in broiler chickens.

Tibia ash, calcium percentage of tibia ash and calcium concentration of blood serum are major criteria for evaluation of tibia bone mineralization (Soares, 1995). These parameters also were not significantly affected by DEB ranging from 187 to 284 mEq Kg<sup>-1</sup> (Table 3). As presented in Table 3, among monovalents of blood serum, only K and CI were significantly affected (P<0.01) by DEB. The increasing of DEB from 187 to 284 mEq Kg<sup>-1</sup> <sup>1</sup> increased the concentration of CI from 103.7 to 111.2 mmol/l. According to Pitts (1970), increasing of Cl in response to increasing diet cations occurs to maintain blood alkali reserves. This response is necessary to adjust acid-base balance. Low K concentration in diet containing 251 mEq Kg-1 of DEB (with 0.35% NaHCO3 and 0 % KHCO3) may be due to an increase in Na content of this diet. These two monovalent electrolytes are antagonism in intestinal level and can block or decrease absorption of each other (Georgievskii et al., 1982). Borges et al. (2003b) did not find any significant effect of DEB ranging from 40 to 340 mEq Kg<sup>-1</sup> on Na, K and CI concentration of blood in broiler chickens. In the study of Borges et al. (2003b), these monovalent electrolytes had been measured in plasma part of the blood and therefore, the monovalent electrolyte contents of blood cells can be contributed to electrolyte values of blood and make difference with the electrolyte values of pullets blood in the present study, which were measured them in serum part of the blood.

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