Kafkas Univ Vet Fak Derg
17 (6): 979-986, 2011
DOI:10.9775/kvfd.2011.4892

RESEARCH ARTICLE

The Effects of Varying Dietary Na/K Ratio and Electrolyte Balance of Diets on Growth, Blood Gases, Hematological Variables, Ionized Calcium and Carcass Traits in Broiler Chickens [1]

Hüseyin NURSOY * Bünyamin SÖGÜT * Türkan TAŞAL**
Reşit ALDEMİR *** Oktay KAPLAN **** Selçuk ALTAÇLI *****

[1] *The present work was supported by the Scientific Research Fund of Yüzüncü Yıl University of Project No: 2007-VF-B14*
* University of Bingöl, Faculty of Agricultural, Department of Zootecnia, TR- 12000 Bingöl-TURKEY
** Van Province Control Laboratory, TR-65100 Van - TURKEY
*** University of Yüzüncü Yıl, Gevaş Vocational College, TR-65700 Van - TURKEY
**** University of Harran, Faculty of Veterinary Medicine, Department of Animal Nutrition and Nutritional Diseases, TR-63300 Şanlıurfa - TURKEY
***** University of Yüzüncü Yıl, Faculty of Veterinary Medicine, Department of Animal Nutrition and Nutritional Diseases, TR-65080 Van - TURKEY

Makale Kodu (Article Code): KVFD-2011-4892

Summary

The effects of 12 different diets which regulated 0.1-0.7 mol Na/K ratio and 176-422 mEq/kg Dietary Electrolyte Balance, DEB, (Na+K–Cl) were investigated on growth, blood gases, hematological variables, biochemical parameters and carcass traits in broiler chickens. Two-day old Ross-308 male chicks (n=180) having individual thirty replicates for 6 groups were used and the study lasted 42 days in the experiment. The S group diet was based on corn and soybean meal and 0.25% salt. This group diet was accomplished by adding 1-0.5% NH_4Cl as A1 and A2 of anionic groups and 0.5-1-1.5% $NaHCO_3$ as C1, C2 and C3 of cationic groups in starter and grower phases, respectively. Increasing Na/K and DEB of the diets had no effect on live weight, body weight gain and feed per gain in 0-42 days (P>0.05). Blood pH quantitatively increased in S, C1 and C2 groups (P<0.05). A1, A2 and S groups of pO_2 levels were higher than those of cationic groups (P<0.05), (R2=0.59). The values of HCO_3, HCO_{3std}, TCO_2, BE_{ecf}, Be(b), Hct and THbc were lower anionic groups than cationic groups (P<0.05), (R2=0.63-0.82). SO_{2c} level was the lowest in C1 group (P<0.05). Anion gaps of the broilers were calculated as 7.97-20.84 mEq/l. Ionized calcium was quantitatively the highest in A1 group. Hot carcass and abdominal fat were not affected by the experimental diets with varying Na/K and DEB (P>0.05). Based on the results of this study, anionic diet such as A1 or 0.1 of Na/K ratio and 212 mEq/kg of DEB could be fed for broilers in case of insufficient Ca. In normal case, it is concluded that C1 and C2 of diets or 0.3-0.5 of Na/K ratios and 259-344 mEq/kg of DEB were appropriate for broilers.

Keywords: *Sodium, Potassium, Na/K, Dietary electrolyte balance, Blood gases, Hematological variables, Anion gap, Ionized calcium, Carcass, Broiler*

Na/K Oranları ve Elektrolit Dengeleri Farklı Olan Rasyonların Etlik Piliçlerde Büyüme, Kan Gazları, Hematolojik Değişkenler, İyonize Kalsiyum ve Karkas Özelliklerine Etkileri

Özet

The effects of 12 different diets which regulated 0.1-0.7 mol Na/K ratio and 176-422 mEq/kg Dietary Electrolyte Balance, DEB, (Na+K–Cl) were investigated on growth, blood gases, hematological variables, biochemical parameters and carcass traits in broiler chickens. Two-day old Ross-308 male chicks (n=180) having individual thirty replicates for 6 groups were used and the study lasted 42 days in the experiment. The S group diet was based on corn and soybean meal and 0.25% salt. This group diet was accomplished by adding 1-0.5% NH_4Cl as A1 and A2 of anionic groups and 0.5-1-1.5% $NaHCO_3$ as C1, C2 and C3 of cationic groups in starter and grower phases, respectively. Increasing Na/K and DEB of the diets had no effect on live weight, body weight gain and feed per gain in 0-42 days (P>0.05). Blood pH quantitatively increased in S, C1 and C2 groups (P<0.05). A1, A2 and S groups of pO_2 levels were higher than those of cationic groups (P<0.05), (R2=0.59). The values of HCO_3, HCO_{3std}, TCO_2, BE_{ecf}, Be(b), Hct and THbc were lower anionic groups than cationic groups (P<0.05), (R2=0.63-0.82). SO_{2c} level was the lowest in C1 group (P<0.05). Anion gaps of the broilers were calculated as 7.97-20.84 mEq/l. Ionized calcium was quantitatively the highest in A1 group. Hot carcass and abdominal fat were not affected by the experimental diets with varying Na/K and DEB (P>0.05). Based on the results of this study, anionic diet such as A1 or 0.1 of Na/K ratio and 212 mEq/kg of DEB could be fed for broilers in case of insufficient Ca. In normal case, it is concluded that C1 and C2 of diets or 0.3-0.5 of Na/K ratios and 259-344 mEq/kg of DEB were appropriate for broilers.

Anahtar sözcükler: *Sodium, Potassium, Na/K, Dietary electrolyte balance, Blood gases, Hematological variables, Anion gap, Ionized calcium, Carcass, Broiler*

İletişim (Correspondence)
+90 426 2160012/1268
nursoymalatya@hotmail.com

INTRODUCTION

Na, K and Cl are important for osmotic regulation and acid-base balance of cells. Glucose which is main energy source for live cells is only penetrated into cell by active transport with Na + K + ATPase pump in cell membranes. Na and K of concentrations into body has opposite effect. If K is more than Na in diet, Na decreases in body because of reabsorbing of Na from kidney, and if Na is more than K in diet, K becomes insufficient [1]. When NaCl or salt added to diet over 0.25-0.50% it results in hypertension [2], and excess KCl is responsible for hypotension in broilers [3].

Dietary Electrolyte Balance (DEB) has been defined as Na+K-Cl and expressed as mEq kg-1. Anionic (-) diets are caused by metabolic acidosis and cationic (+) diets to metabolic alkalosis [4]. Mongin [5] indicated that DEB value of broiler was 250 mEq/kg for optimal growth and acid-base balance. Borges et al.[6] reported that over 360 mEq/kg DEB value is caused by alkalosis in broilers. Ca and 1.25-$(OH)_2D_3$ of absorption in intestine and Ca reabsorbing in kidney are reduced in metabolic alkalosis [4]. Therefore, animals should be fed with anionic diets or lower cation especially phases of excess needed Ca such as growth, lactation, egg-laying [7].

This study aimed to find out the diet in which Na/K ratio and DEB value produce the highest synergetic effect. This research was carried out to determine the effects of 12 different diets groups which regulated 0.1-0.7 mol Na/K ratio and 176-422 mEq/kg DEB on growth, blood gases, hematological variables, biochemical parameters and carcass traits result in high speed growth in broiler.

MATERIAL and METHODS

Bird Management and Diets

This study [approved by Yüzüncü Yıl University Animal Experiments Local Ethic Committee, Date: 02.11.2009, Decision No: 07] was carried out at The Broiler Research Unit of Yüzüncü Yıl University, Van, Turkey. Birds were housed in 1.0x1.5 m for 10 chicks with suspended feeders and drinkers and bedding with wood shavings. The experiment lasted for 42 days. A total of 180 Ross-308 male two-day-old broilers were randomly assigned to 6 dietary treatments with 30 replicates in the experiment. Chicks were purchased from CP Piliç® Hatchery, Erzincan, Turkey. The chicks included in the experiment were weighed on the third day morning. In the beginning of experiment, chicks were attached with "wing banded" and thus each chick has one replication. Poultry house was applied with a light regime of 23L:1D.

Groups of the experiment were formed by anionic groups as A1 and A2, salt group as S and cationic groups as C1, C2 and C3 in starter and grower phases *(Table 1)*. The experiment diets are preferred anionic feeds more less cationic feeds because blood pH, intracellular and extracellular fluids are cationic in most animals. Na/K ratios and DEB values of the experimental diets are made in respect of live body features of chicks such as blood, intracellular and extracellular fluids of pH. S group diet was prepared by using corn and soy bean meal and 0.25% salt. S group diet was accomplished by adding 1-0.5% NH_4Cl as A1 and A2 groups, and 0.5-1-1.5% $NaHCO_3$ as C1, C2 and C3 groups, respectively. The diets were formulated to meet all nutrient requirements of broiler according to NRC [2]. Feed and water were provided ad libitum to the birds.

Chemical Analyses of Feeds and Bloods

Samples of diets of the experiment were analyzed for concentrations of Na, K, Cl, Ca, and P in Ministry of Agricultural, Institute of Central Research of Soil, Fertilizer and Water Sources, Ankara, Turkey. Samples of feeds were burned for all analyses by using 3/1 nitric acid/perchloric acid solution [8]. After supernatants were discharged, remainings were analyzed for total sodium and potassium by Flame Photometric with Jenway PFP 7 (Jenway Ltd., Dunmow, UK); total calcium by Inductively Coupled Plasma-Optical Emission Spectrometry (*ICP-OES*) with Varian 720-ES (Varian Inc., Walnut Creek, CA); total phosphorus by using *vanadomolybdophosphoric yellow colour* procedure with Shimadzu UV-160 Spectrophotometer (Shimadzu Inc., Kyoto, Japan); chlorine by titrimetric with $AgNO_3$ according to Kaçar and İnal [8]. Thus, measured the values were used instead of table values for Na+K-Cl formula.

Feed samples were analyzed according to AOAC [9] for dry matter at 105°C and 16 h (AOAC-967.03) in oven-drying (Nüve Basic Laboratory Equipment, İstanbul, TR); ash for 2 h at 600°C (AOAC 942.05) with furnace (Elektromag, İstanbul, TR); fat by dietilether extraction method (AOAC 920.39) with fat extractor (Velp Scientifica, 148 Solvent Extractor Milan, IT); crude protein by Nx6.25 Kjeldahl method (AOAC 984.13) with protein analyzer (Gerhard Kjeldatherm, Königswinter, DE) in University of Yüzüncü Yıl, Faculty of Veterinary Medicine, Laboratory of Animal Nutrition and Nutritional Disease. Metabolizable energy levels of diets were calculated according to the description of Pauzenga [10].

Blood samples of the experiment were collected from 7 birds of each group into green closure tubes with lithium heparin on slaughter day (42^{nd} day of the experiment), and the samples were immediately taken to Laboratory of Van Medical Park Hospital. The bloods were analyzed on pH, pCO_2, pO_2, pCO_3^{-}, HCO_{3std}, TCO_2, BE_{ecf}, Be(B), SO_2c, Hct, THbc, Ca^{++}, Ca, P, Na, K, Cl, Mg, protein uric acid, glucose, lactate with instrument of blood gases (GEM Premier 3000, Instrumentation Laboratory Inc., Lexington, MA, USA) and auto analyzer (Cobas Integra-800 Analyzer, Roche Diagnostics GmbH., Mannheim, DE).

Table 1. *Ingredients and chemical compositions of diets in the experiment*
Tablo 1. *Deneme rasyonlarının bileşimleri ve kimyasal kompozisyonları*

Ingredients, kg	Starter Phase (0-21 d)						Grower phase (21-42 d)					
	A1	A2	S	C1	C2	C3	A1	A2	S	C1	C2	C3
	Measured Dietary Na/K, mol mol^{-1}/Measured DEB[1], mEq kg^{-1}											
	0.1/212	0.1/230	0.2/254	0.3/319	0.4/344	0.6/422	0.1/176	0.1/220	0.2/217	0.3/259	0.5/311	0.7/356
Corn	512.5	517.5	522.5	517.5	512.5	507.5	608.5	613.5	618.5	613.5	608.5	603.5
Soybean meal, 44% CP	380	380	380	380	380	380	290	290	290	290	290	290
Canola oil	30	30	30	30	30	30	36	36	36	36	36	36
Dicalcium phosphate [2]	20	20	20	20	20	20	20	20	20	20	20	20
Limestone, 38% Ca	39	39	39	39	39	39	27	27	27	27	27	27
Mineral-Vitamin premix [3]	2.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5
Ethoxyquin	1	1	1	1	1	1	1	1	1	1	1	1
DL-Methionine	2	2	2	2	2	2	2	2	2	2	2	2
L-Lysine HCl	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
NaCl [4]	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5
NH_4Cl [5]	10	5	-	-	-	-	10	5	-	-	-	-
$NaHCO_3$ [6]	-	-	-	5	10	15	-	-	-	5	10	15
Total	1000	1000	1000	1000	1000	1000	1000	1000	1000	1000	1000	1000
Analyzed nutrients												
Dry matter (DM), %	91.86	91.89	91.62	91.34	92.18	91.57	91.11	91.68	91.67	91.26	91.46	91.55
Ash, %	7.57	6	6.48	6.36	6.57	7.53	8.78	9.26	8.60	7	9	9.12
Crude protein, %	22	22.1	22	22	22.15	22.05	18.70	18.7	18.5	18.7	18.2	18.3
Ether extract, %	6.20	6.15	6.36	6.42	5.08	4.52	5.09	4.62	4.63	4.21	4.08	4.71
Crude fiber, %	4.19	4.28	4.37	4.35	4.01	4.25	4.2	4.08	4.38	4.3	4.2	4.35
NFE, % [7]	51.9	53.36	52.41	52.19	54.38	53.22	54.34	55.1	55.56	57.05	55.98	55.07
AME, kcal/kg [8]	3164	3215	3195	3192	3166	3075	3037	3026	3036	3062	2994	3017
Ca, %	0.95	0.89	0.76	0.87	0.79	0.94	1.32	1.31	1.42	1.35	1.62	1.86
P, %	0.94	0.77	0.88	0.89	0.89	0.71	0.98	1.05	0.94	1.04	1.04	0.98
Na, % of DM	0.11	0.13	0.15	0.28	0.35	0.54	0.10	0.13	0.15	0.25	0.39	0.50
K, % of DM	0.91	0.92	0.87	0.96	0.95	0.93	0.76	0.76	0.79	0.73	0.75	0.74
Cl, % of DM	0.25	0.22	0.12	0.17	0.18	0.18	0.22	0.11	0.18	0.13	0.18	0.18

[1] DEB, Dietary Electrolyte Balance, mEq/kg DM = (Na + K)-Cl, [2] 24% Ca and 16.2% available P, [3] DSM Rovimix VM 124® in per 2.5 kg: : 15.000.000 IU Vitamin A, 3.000.000 IU Vitamin D_3, 100.000 mg Vitamin E, 5.000 mg Vitamin K_3, 3.000 mg Vitamin B_1, 6.000 mg, Vitamin B_2, 50.000 Niacin, 6.000 mg Vitamin B_6, 20 mg Vitamin B_{12}, 15.000 mg Calcium D-Pantotenate, 50.000 mg Niacin, 1.500 mg Folic acid, 150 mg Biotin, 80.000 mg, Mn, 60.000 mg Fe, 60.000 mg Z, 5.000 mg Cu, 200 mg Co, 1.000 mg I, 150 mg Se, [4] NaCl = 0.40% Na and 0.60% Cl, [5] NH_4Cl = 0.66% Cl, [6] $NaHCO_3$ = 0.27% Na, [7] NFE, Nitrogen Free Extract, % = 100 - (% Moisture + % Crude Protein + % Crude Fiber + % Ether Extract + %Ash), [8] AME, Apparent Metabolizable Energy, kcal/kg = 37 x % Crude Protein + 81.8 x % Ether Extract + 35.5 x % NFE [10]

Growth Parameters and Carcass Traits

All birds of the experiment were weighed and calculated weekly for live weight, feed intake and feed conversion rate (FCR), (feed/gain). On 42nd day of the slaughter day, heads, foots and feathers of birds were separated from those trunks of 7 birds of each group, and organs of digestive system except liver and kidney removed from those trunks as "hot carcass". Dividing hot carcass weight into live weight before slaughtering were calculated "carcass yield". Fats of covers of gizzard, duodenum and intestine were weighed as "abdominal fat".

Statistical Analysis

Data of the experiment were analyzed by GLM procedure and differences of means were considered significant by Duncan Multiple Range test of SAS [11] ($P<0.05$). Regression equations (R^2=over 50%) of variables were determined by linear effect.

RESULTS

Effects of dietary treatments of the experiment were shown on live weight, feed intake and FCR were shown in

Table 2. The highest live weight was quantitatively observed at C2 group in 21-42 d, however, there was no significant differences among the groups in 0-21 and 0-42 days. Feed intake and FCR values of the experiment were not affected by diets at 0-42 days. Blood gases and hematological variables of the experiment are shown in *Table 3*. The pH values of cationic groups were quantitatively higher than anionic groups. Likewise, pCO_2 was quantitatively high in cationic groups (*Table 6*, R^2=0.53). Partial pressures of oxygen of this study were determined between 32.42 to 59.85 mmHg, and pO_2 levels were high A1, A2 and S groups. The values of HCO_3, HCO_{3std}, TCO_2, BE_{ecf}, Be(b), Hct and THbc in this study were observed as lesser anionic groups than those of cationic groups, (P<0.05), (*Table 6*, R^2=0.63-0.82). The SO_{2c} level was the lowest in C1 group (P<0.05) comparing the other groups. The anion gaps of the broilers were calculated as 7.97-20.84 mEq/l. The blood biochemical parameters were presented in *Table 4*. Ionized calcium was quantitatively the highest in A1 group. Ca, Mg and total protein of blood had no effect diets with Na/K ratios and varying DEB. P level was the highest in S group, Na level increased in cationic groups (P<0.05), and K and Cl levels increased quantitatively A1 group. Sodium level of this study was found to be increased, but potassium decreased with increasing Na/K and DEB (*Table 6*, R^2=0.73 and 0.76, respectively). Hot carcass and abdominal fat of

Table 2. *Effects of experimental diets with A1, A2, S, C1, C2 and C3 or Na/K ratio (0.1-0.7 mol/mol) and DEB (176-422 mEq/kg) on live weight, feed intake and FCR*

Tablo 2. *A1, A2, S, C1, C2 ve C3 veya Na/K oranı (0.1-0.7 mol/mol) ve DEB (176-422 mEq/kg) olan deneme rasyonlarının canlı ağırlık, yem tüketimi ve yemden yararlanma üzerine etkileri*

Diets	Live Weight, g			Feed Intake, g			FCR, (feed/gain)		
	Days			Days			Days		
	0 to 21	21 to 42	0 to 42	0 to 21	21 to 42	0 to 42	0 to 21	21 to 42	0 to 42
A1	721	1896^{ab}	2618	1259^{ab}	2846	4105	1.71^{bc}	1.63^{ab}	1.67
A2	693	1894^{ab}	2589	1124^{b}	2858	4082	1.74^{bc}	1.60^{ab}	1.67
S	727	1867^{ab}	2595	1263^{ab}	2849	4109	1.70^{c}	1.70^{a}	1.70
C1	683	1918^{ab}	2602	1280^{a}	2841	4122	1.83^{a}	1.54^{b}	1.68
C2	719	1971^{a}	2691	1287^{a}	2955	4242	1.77^{ab}	1.59^{ab}	1.68
C3	744	1831^{b}	2575	1289^{a}	2855	4144	1.68^{c}	1.69^{a}	1.69
SEM	21.52-	36.08	53.78 -	15.80	80.39 -	84.24 -	0.020	0.049	0.024 -

-: *Different of means of groups in column are non-significant (P>0.05)*
$^{a-c}$ *Means with different superscripts a same column are significantly different (P<0.05)*

Table 3. *Effects of experimental diets with A1, A2, S, C1, C2 and C3 or Na/K (0.1-0.7) and DEB (176-422 mEq/kg) on blood gases and hematological variables*

Tablo 3. *A1, A2, S, C1, C2 ve C3 veya Na/K oranı (0.1-0.7 mol/mol) ve DEB (176-422 mEq/kg) olan deneme rasyonlarının kan gazları ve hematolojik değişkenler üzerine etkileri*

Variables	**A1**	**A2**	**S**	**C1**	**C2**	**C3**	**SEM**
pH	7.14^{c}	7.17^{bc}	7.29^{a}	7.27^{a}	7.30^{a}	7.23^{ab}	0.02
pCO_2, mmHg	30.42^{bc}	31.57^{bc}	26.85^{c}	40.28^{a}	37.00^{ab}	39.28^{a}	2.19
pO_2, mmHg	58.71^{a}	55.57^{a}	59.85^{a}	32.42^{b}	37.57^{b}	40.57^{b}	4.24
HCO_3^-, mmol/l	14.75^{b}	14.02^{b}	20.41^{a}	20.28^{a}	21.32^{a}	21.3^{a}	1.18
HCO_{3std}, mmol/l	14.25^{b}	13.98^{b}	20.22^{a}	19.4^{a}	20.71^{a}	19.88^{a}	1.02
TCO_2, mmol/l	16.08^{b}	15.22^{b}	21.70^{a}	21.61^{a}	22.68^{a}	22.81^{a}	1.24
BE_{ecf}, mmol/l	-14.27^{b}	-14.40^{b}	-6.07^{a}	-6.6^{a}	-5.08^{a}	-6.15^{a}	1.45
Be(b), mmol/l	-13.22^{b}	-13.4^{b}	-5.671^{a}	6.22^{a}	4.84^{a}	-5.84^{a}	1.32
SO_{2c}, %	86.28^{abc}	89.42^{ab}	92.42^{a}	55.28^{d}	71.00^{c}	71.00^{c}	4.90
Hct, %	22.00^{c}	24.14^{bc}	26.14^{bc}	30.42^{a}	37.57^{a}	29.57^{a}	4.24
Thbc, g/dl	6.80^{c}	7.48^{b}	8.11^{b}	9.47^{a}	9.27^{a}	9.17^{a}	0.34
Anion gap, mEq/l	10.79^{b}	20.84^{a}	13.84^{ab}	11.16^{b}	7.97^{b}	13.91^{ab}	2.69

$^{a-d}$ *Means with different superscripts a same row are significantly different (P<0.05)*
pH: *-log10 H^+,* ***pCO_2:*** *carbon dioxide partial pressure,* ***pO_2:*** *partial pressures of oxygen,* ***HCO_3^-:*** *bicarbonate,* ***HCO_{3std}:*** *standard plasma bicarbonate,* ***TCO_2:*** *total CO_2,* ***BE_{ecf}:*** *base excess extra cellular fluid,* ***Be(b):*** *base excess in plasma,* ***SO_{2c}:*** *oxygen saturation of haemoglobin,* ***Hct:*** *hematocrit,* ***Thbc:*** *total haemoglobin,* ***Anion gap:*** *Na^+-(Cl^- + HCO_3^-)*

Table 4. *Effects of experimental diets with A1, A2, S, C1, C2 and C3 or Na/K (0.1-0.7) and DEB (176-422 mEq/kg) on blood and serum parameters*

Tablo 4. *A1, A2, S, C1, C2 ve C3 veya Na/K oranı (0.1-0.7 mol/mol) ve DEB (176-422 mEq/kg) olan deneme rasyonlarının kan ve serum parametrelerine etkileri*

Parameters	A1	A2	S	C1	C2	C3	SEM
Ca^{++}, mmol/l	1.41^{a}	1.36^{ab}	1.28^{c}	1.37^{ab}	1.37^{ab}	1.32^{bc}	0.01
Calcium,(serum),g/dl	12.02	11.57	11.28	11.72	11.17	11.85	0.55 -
Phosphorus, mg/dl	7.15^{b}	6.38^{bc}	8.32^{a}	6.55^{c}	6.04^{c}	6.90^{bc}	0.33
Sodium, mmol/l	141.42^{b}	142.85^{b}	142^{b}	147.85^{a}	146^{a}	147.57^{a}	0.95
Potassium, mmol/l	5.45^{a}	5.01^{ab}	4.80^{b}	2.88^{cd}	2.64^{d}	3.21^{c}	0.16
Chlorine (serum), mmol/L	120.89^{a}	116.9^{bc}	118.7^{ab}	115.0^{bc}	115.1^{bc}	113.14^{c}	1.23
Magnesium, mg/dl	3.25	3.15	3.52	4.13	3.38	3.28	0.41 -
Total protein (serum),g/dl	3.89	2.98	3.36	3.63	3.28	3.48	0.34 -
Uric acid, mg/dl	3.58^{b}	5.65^{a}	4.48^{ab}	4.19^{ab}	3.65^{b}	4.98^{ab}	0.47
Glucose, mg/dl	232^{a}	233^{a}	228^{a}	202^{b}	214^{ab}	230^{a}	7.39
Lactate, mmol/l	5.92^{c}	6.62^{bc}	5.57^{c}	9.62^{a}	8.77^{ab}	8.87^{ab}	0.76

-: *Different of means of groups in rows are non-significant (P>0.05)*
$^{a-c}$ Means with different superscripts a same row are significantly different (P<0.05)

Table 5. *Effects of experimental diets with A1, A2, S, C1, C2 and C3 or Na/K (0.1-0.7) and DEB (176-422 mEq/kg) on some carcass traits*
Tablo 5. *A1, A2, S, C1, C2 ve C3 veya Na/K oranı (0.1-0.7 mol/mol) ve DEB (176-422 mEq/kg) olan deneme rasyonlarının bazı karkas özelliklerine etkileri*

Traits	A1	A2	S	C1	C2	C3	SEM
Hot carcass, g	1705	1817	1825	1775	1863	1790	0.07 -
Carcass yield, %	71.09^{b}	71.55^{b}	75.07^{a}	68.96^{b}	69.30^{b}	71.21^{b}	1.06
Abdominal fat, g	25.71	26.28	26.28	31.14	24.85	32.00	2.72 -

-: *Different of means of groups in rows are non-significant (P>0.05)*
$^{a-b}$ Means with different superscripts a same row are significantly different (P<0.05)

Table 6. *Regression equations (R^2=over 50%) of experimental diets with A1, A2, S, C1, C2 and C3 or Na/K (0.1-0.7) and DEB (176-422 mEq/kg)*

Tablo 6. *A1, A2, S, C1, C2 ve C3 veya Na/K oranı (0.1-0.7 mol/mol) ve DEB (176-422 mEq/kg) olan deneme rasyonlarının regresyon eşitlikleri ((R^2= %50 üzeri)*

Variable (y)	Prediction Equation (regression)*	Mean	R^2
pCO_2, mmHg	y = 2.1149x + 26.831	34.23	0.53
pO_2, mmHg	y = -4.918x + 64.661	47.45	0.59
Hct, %	y = 2.3549x + 20.065	28.31	0.63
HCO_3^-, mmol/l	y = 1.5577x + 13.228	18.68	0.75
HCO_{3std}, mmol/l	y = 1.3577x + 13.321	18.07	0.67
TCO_2, mmol/l	y = 1.5983x + 14.423	20.02	0.76
BE_{ecf}, mmol/l	y = 1.9437x – 15.565	-8.76	0.70
Thbc, g/dl	y = 0.5309x + 6.5253	8.38	0.82
Chlorine (serum), mmol/L	y = -1.3671x + 121.41	116.62	0.82
Sodium, mmol/l	y = 1.3157x + 140.01	144.62	0.73
Potassium, mmol/l	y = -0.578x + 6.0213	4.00	0.78
Lactate, mmol/l	y = 0.7214x + 5.0367	7.56	0.60

** Linear effect, P<0.05*

carcass traits were not affected by the diet with varying Na/K and DEB *(Table 5)*, ($P>0.05$), but carcass yield of S group was higher than the other groups. The regression equations (R^2=over 50%) of the parameters of experiment are presented in *Table 6*.

DISCUSSION

It is required for the diets of broilers to have 0.20% Na, 0.30% K and 0.20% Cl for 0-3 weeks of age and 0.15% Na, 0.30% K and 0.15% Cl for 3-6 weeks of age according to NRC [2]. However, required values of Na, K and Cl are reported apart from those of NRC by researchers [2]. For example, in order to reach optimum body weight, it is emphasized by Mustahq at al.[12] and Murakami et al.[13] that the amount of Na and Cl should be 0.25% and 0.30% for 0-28 days and 0.15% Na and 0.23% Cl for 21-42 days, respectively. Besides, amount of K should be 0.50% and 0.47% for 0-21 d and 21-42 d, respectively in broilers diets [14].

In the present study, the effects of diet supplemented with NH_4Cl or anion groups (A1 and A2), salt group (S) and $NaHCO_3$ or cation groups (C1, C2 and C3) or 0.1-0.7% Na/K and 176-422 mEq/kg DEB on the performances of broilers are shown in *Table 2*. The highest live weight was quantitatively observed in C2 group in 21-42 days ($P<0.05$), however, there were no significant differences among the groups in 0-21 and 0-42 days. Live weight of C2 group (0.4-0.5 ratio of Na/K and 311-344 mEq/kg of DEB) was higher than that of C3 agreed with results of Borges et al.[6] as over 340 mEq/kg of DEB decreased live weight in broiler. Johnson and Karunajeewa [15] was also reported that 0.38 of Na/K and 344 mEq/kg of DEB increased live weight compared to 0.11 of Na/K and 553 mEq/kg of DEB in broilers diets. Our experiment, observing no significant differences among the groups during the trail (0-42 d), ($P>0.05$) are corroborated to Murakami et al.[13] and Ravindran et al.[16] since they have indicated that the existence of 150-330 mEq/kg of DEB has no significant effect on body weight gain. However, Borges et al.[6] reported that body weight increased by increasing DEB from 40 to 240 mEq/kg. Diler et al.[17] observed that increasing Na/K ratio from 0.5 to 1.39 enhanced body weight gain of trout. In this study, using lower Na/K ratio (0.1-0.7) than those of Diler et al.[17] could be the reason for not having body weight gain.

Feed intake and FCR of the experiment were not affected by varying Na/K and DEB during 0-42 days ($P>0.05$) as shown in *Table 2*. Similar results were also reported by Ravindran et al.[16] that over 300 mEq/kg DEB in broiler diet did not influence feed intake, and Johnson and Karunajeewa [15] indicated that 0.38 of equal Na/K ratio and different -29 - 180 mEq/kg of DEB of diets did not affect on FCR.

The effects of varying Na/K and DEB of the diets on blood gases and hematological parameters are presented in *Table 3*. The values of blood pH of our study were agreed with those of 7.10-7.37 in the previous studies [15,18]. In the present experiment, the values of blood pH were found to be quantitatively higher in cationic groups than those of anionic groups, like the result of Olanrewaju et al. [18]. In our opinion, this elevation can stem from the effect of $NaHCO_3$ cationic supplement. However, the other studies [13,15] reported that blood pH was not influenced by Na/K and DEB.

PCO_2 was quantitatively high in cationic groups (*Table 3* and *Table 6*, R^2=0.53). PO_2 of this study were determined between 32.42 to 59.85 mmHg. These findings are lower than those of Murakami et al.[13] reported between 74.63 and 83.9 mmHg. The explanation may be that this study was carried out the experiment at high altitude (1727m) in Van, Turkey, while Murakami et al.[13] carried out his experiment about 932 m altitude in Colombo, Parana, Brazil, about twice lower altitude than Van. Because atmospheric pressure is 760 mmHg at sea level, it becomes 523 mmHg at 3.000 m, and 87 mmHg at 15.000 m. Therefore, amount of oxygen is decreased by soaring in places with high altitude. While partial pressure of oxygen partial pressure was 159 mmHg at sea level, it become 18 mmHg at 15.000 m altitude [20]. Observing higher pO_2 in anionic and S groups of the experiment are stemmed from low level pCO_2, because pO_2 and pCO_2 parameters are opposite effect to each other. Olanrewaju et al.[18] indicated that increasing DEB did not effect on pO_2.

Values of HCO_3, HCO_{3std}, TCO_2, BE_{ecf}, Be(b), Hct and THbc parameters are seen lesser anionic groups than those of cationic groups, ($P<0.05$), (See *Table 6*, R^2=0.63-0.82). The values of parameters confirm results of the other studies [6,13,15,19]. The results were corroborated those of Squires and Julian [19] reported that high bicarbonate ($NaHCO_3$) or DEB of diets increased on pCO_2, HCO_3, Hct and THbc. It is considered that this elevation of values in cationic groups could be due to reabsorption of bicarbonate in kidneys and again converting to CO_2. However, researchers [6,15,18] indicated that increasing DEB did not altered values of pCO_2, HCO_3, HCO_{3std}, TCO_2, BE_{ecf} and Be(b). The percentages of Hct were ranged between 22 and 37.57% in this study. The other studies [6,18] reported that increasing DEB of broiler diet did not affect on Hct. However, Squires and Julian [19] reported that increasing DEB increased Hct. It was estimated in this study that lowering of Hct and THbc of A1, A2 and S1 groups of the experiment is caused by high pO_2 and SO_{2c}. This situation may indicate that oxygen carrying capacity of anionic groups is lower than that of cationic groups.

The SO_{2c} level was the lowest in C1 group ($P<0.05$). Ninety-five percent of oxygen is bonded on haemoglobin. The O_2 saturation of hemoglobin depends on directly pO_2 [21] Murakami et al.[13] reported that SO_{2c} was not affected by increasing DEB or sodium level. The higher level of SO_{2c} in anionic groups may be caused by increasing oxygen consumption of tissue because of an anionic situation such as low pH and pCO_2.

Anion gaps of diets of in the present study were calculated as 7.97-20.84 mEq/l. Anion gap is increased at acidic condition or presence of high Cl. If $NaHCO_3$ or DEB are high in diet, anion gap would be low [18,19]. Normally, low anion gap is preferred. In this study, the lowest anion gap was observed 7.97 mEq/l of C2 group (P<0.05), which may be a sign for the best diet for broilers.

In terms of the blood biochemical parameters (*Table 4)*, the ionized calcium was quantitatively the highest in A1 group. Ionized calcium (Ca^{++}) is 50% of total calcium in plasma [22]. Ionized or free calcium decrease when blood pH rises [23]. Squires and Julian[19] found that high sodium bicarbonate of diet decreased blood ionized calcium. In other words, ionized calcium increases in acidic condition which is parallel with the result of this study.

Effects of diets with Na/K ratios and varying DEB did not on concentrations of Ca, Mg and total protein of blood *(Table 4)*. The highest P was found in S group received diet contains only salt (P<0.05). In general, P decreases in hypokalemia [22]. The P values of plasma in the experiment agree to those of Lima et al.[24]. The Na level of this study increased (P<0.05), (*Table 6*, R^2=0.73), but potassium decreased increasing Na/K and DEB (P<0.05), (*Table 6*, R^2=0.78) because potassium decrease by entering into cell and plunging urine in alkalosis [22]. Increasing Na and lowering K in cationic groups of our study are similar in Squires and Julian's results [19]. However, the other researchers [18,25] elevated DEB value did not affect blood sodium and potassium. Cl was quantitatively the highest in A1 group. Researchers [25] reported that Cl decreases depend on increasing Na/K and DEB. Uric acid, which is high in acidic condition without causing clinical problem, is an end-product of protein metabolism. The highest of uric acid in acidic group (A2) in this study is confirmed by literature [22]. Glucose values of the study agree to those of others studies [6,18,25]. Glucose was not affected by Na/K and DEB value (R^2=<50%, *Table 6*) in the experiment because it is affected by adrenaline, noradrenaline and glucucorticoids [6]. Lactate was quantitatively figured out high in anionic groups, but low in cationic groups in this study (*Table 6*, R^2=0.60). Similar results were corroborated by Raup and Bottje [26] that lesser lactate was produced at low blood pH.

Hot carcass and abdominal fat of carcass traits were not affected by the diet with varying Na/K and DEB (P>0.05) in *Table 5*, but, carcass yield of S group was higher than those of others. No significant effects of DEB with 130-360 mEq/kg were found on carcass yield and abdominal fat by Borges et al.[6]. Parallel results were also reported using of diets with DEB from 210 to 330 mEq/kg by Borgatti et al.[27]. Likewise, the study of Johnson and Karunajeewa [15] found that dressed carcass of broilers were not affected by the diets containing 0.38 of Na/K and between -29 mEq/kg and 553 mEq/kg of DEB. However, decreasing DEB increased abdominal fat, reported by Jianlin et al.[28].

Based on the results of this study, anionic diet such as A1 or 0.1 of Na/K ratio and 212 mEq/kg of DEB could be fed for broilers in case of insufficient Ca. In normal case, it could be said that C1 and C2 of diets or 0.3-0.5 of Na/K ratios and 259-344 mEq/kg of DEB were appropriate for broilers.

Acknowledgments

The authors thank to Dr. Ecevit EYDURAN, Assistant Professor at Iğdır University, for calculating of statistical of the experiment.

REFERENCES

1. Çavusoğlu H: Renkli Fizyoloji Atlası. Nobel/Yüce Yayınevi, İstanbul, 1997.

2. NRC, National Research Council: The Nutrient Requirements of Poultry. 9th Revised ed. National Academy Press, Washington DC, 1994.

3. Hooge DM, Cummings KR, McNaughton JL: Effects of ionophore coccidiostat (monensin or salinomiycin), sodium or potassium bicarbonate, or both bacitracin methylene disaliclate in broiler chicken diets. *Bra J Poult Sci*, 2 (1): 75-83, 2000.

4. Block E: Manipulation of dietary cation-anion difference on nutritionally related productivity, and metabolic responses of dairy cows. *J Dairy Sci,* 77, 1437-1450, 1994.

5. Mongin P: Recent advances in dietary cation-anion balance: Applications in poultry. *Proc Nutr Soc (Camb)*, 40, 285-294, 1981.

6. Borges SA, Fischer da Salvia AV, Ariki J, Hooge DM, Cummings KR: Dietary electrolyte balance for broiler chickens under moderately high ambient temperatures and relative humidities. *Poult Sci*, 82, 301-308, 2003.

7. Leclerc H, Block E: Effects of reducing dietary cation-anion balance for prepartum dairy cows with specific reference to hypocalcemic parturient paresis. *Can J Anim Sci*, 69, 411-423, 1989.

8. Kaçar B, İnal A: Bitki Analizleri. Nobel Yayın No 1241, Ankara, 2008.

9. AOAC: Official Methods of Analysis of the Association of Official Analytical Chemists. 15th ed., Arlington, Virginia, 1990.

10. Pauzenga U: Feeding Parent Stock. Zootech Int, pp. 22-25, 1985.

11. SAS: Statistical Analysis System SAS/STATE User guide. Version 6.12, SAS Institute Inc, Cary, NC, USA, 1995.

12. Mustahq T, Sarwar M, Nawaz H, Aslam MM, Ahmad T: Effect and interactions of dietary sodium and chloride on broiler starter performance (hatching to twenty-eight days of age) under subtropical summer conditions. *Poult Sci*, 84, 1716-1723, 2005.

13. Murakami AE, Oviedo-Rondo EO, Martins EN, Pereira MS, Scapinello C: Sodium and chloride requirements of growing broiler chickens (twenty-one to forty-two days of age) fed corn-soybean diets. *Poult Sci,* 80, 289-294, 2001.

14. Oliveira JE, Albino LFT, Rostango HS, Paez LE, Carvalho DCO: Dietary levels of potassium for broiler chickens. *Rev Bras Ciene Avic* 1, 33-37, 2005.

15. Johnson RJ, Karunajeewa H: The effects of dietary minerals and electrolytes on the growth and physiology of the young chick. *J Nutr,* 115 (12): 1680-1690, 1985.

16. Ravindran V, Cowieson AJ, Selle PH: Influence of dietary electrolyte balance and microbial phytase on growth performance, nutrient utilization, and excreta quality of broiler chickens. *Poult Sci*, 87, 677-688, 2008.

17. Diler İ, Arabacı M, Nursoy H, Göncü M: The effects of diets supplemented with $CaCl_2$ and rock salt on growth, feed efficiency and some serum mineral levels in rainbow trout, *Oncorhynchus mykiss. Kafkas Univ Vet Fak Derg,* 16 (Suppl-B): 271-275, 2010.

18. Olanrewaju HA, Thaxton JP, Dozier III WA, Branton SL: Electrolyte diets, stress, and acid-base balance in broiler chickens. *Poult Sci*, 86, 1363-1371, 2007.

19. Squires EJ, Julian RJ: The effect of dietary chloride and bicarbonate on blood pH, hematological variables, pulmonary hypertension and ascites in broiler chickens. *Brit Poult Sci,* 42 (2): 207-212, 2001.

20. Yalçın M, Kardeşoğlu E, Işılak Z: High altitude and heart. *TAF Prev Med Bull,* 10 (2): 211-222, 2011.

21. Acunaş B: Ventilatördeki Bebeğin İzlemi, http://www.guncelpediatri.com/eng/sayilar/10/ 105-109.pdf, *Accessed: 09.06.2011*.

22. Üstdal M, Karaca L, Testereci H, Kuş S, Paşaoğlu H, Türköz Y: Biyokimya. Pelikan Yayıncılık, Ankara, 2005.

23. Odom TW, Harrison PC, Bottje WG: Effects of thermal-induced respiratory alkalosis on blood ionized calcium levels in the domestic hen. *Poult Sci,* 65 (3): 570-573, 1986.

24. Lima FR, Mendonca JR, CX Alvarez JC, Garzillo JMF, Ghion E, Leal PM: Biological evaluations of commercial dicalcium phosphates as sources of available phosphorus for broiler chicks. *Poult Sci,* 76, 1707-1713, 1997.

25. Borges SA, Fischer da Silva AV, Majorka A, Hooge DM, Cummings KR Physiological responses of broiler chickens to heat stress and dietary electrolyte balance (sodium plus potassium minus chloride, miliequivalents per kilogram). *Poult Sci,* 83, 1551-1558, 2004.

26. Raup TJ, Bottje WG: Effect of carbonated water on arterial pH, p_{co2} and plasma lactate in heat-stressed broilers. *British Poult Sci,* 31 (2): 377-384, 1990.

27. Borgatti LMO, Albuquerque R, Meister NC, Souza LWO, Lima FR, Trindade Neto MA: Performance of broilers fed diets with different dietary electrolyte balance under summer conditions. *Rev Bras Cienc Avic,* 6 (3): 153-157, 2004.

28. Jianlin S, Fritts CA, Burnham DJ, Waldroup PW: Extent to which crude protein may be reduced in corn-soybean meal broiler diets through amino acid supplementation *Int J Poult Sci,* 3 (1): 46-50, 2004.