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INFLUENCE OF INTRAVENOUS INJECTION OF NaCl AND $CaCl_2$ ON PHYSIOLOGICAL RESPONSE OF BROILER CHICKS

A.M. El-Kaiaty

Department of Animal Production, Faculty of Agriculture, University of Cairo , Giza, Egypt

SUMMARY

This experiment was designed to test the hypothesis that broiler chicks which were injected intravenouslly by either NaCl or $CaCl_2$ under hot climate would have greater thermotolerance than sham treated chicks. One hundred and eighty chicks one day of age (Arbor Acres) were obtained and divided into six equal groups as: group 1: operated as sham "control"; group 2: injected by avian physiological saline (0.9 % NaCl); group 3: injected by saline + 2% NaCl; group 4: injected by saline + 4% NaCl; group 5: injected by saline + 2% $CaCl_2$; and group 6: injected by saline + 4% $CaCl_2$. Intravenous injection by 1 ml of each solution in each group was given biweekly (2, 4 and 6). The experiment continued during July and August with natural day light, with an average of ambient temperature of 33.6±2.0°C° and an average of relative humidity of 65±4%.

Body weight, feed intake and feed conversion were not affected by applied treatments. Moreover, no incidence of mortality occurred in the treated groups,although 5 chicks (16%) died in the control group.

NaCl and $CaCl_2$ appeared to regulate the thermostasis differently, the first by increasing ($P<0.05$) the rectal temperature and the second by increasing ($P<0.05$) respiration rate and decreasing rectal temperature.

Intravenous injection by either NaCl or $CaCl_2$ improved the response of chickes to high temperature as observesd from all plasma constituents studied. The result of significant ($P<0.05$) increase in plasma total protein, T3 and T4; and significant ($P<0.05$) decrease of plasma glucose and corticosterone are confirming the above mentioned suggestion. The result also showed significantly ($P<0.05$) high increase in plasma calcium and inorganic phosphate due to the injection of NaCl or $CaCl_2$.

Keywords: Broiler, hot climate, NaCl and $CaCl_2$ injection, thermoergulation

INTRODUCTION

Birds are homeotherms which means that they maintain a relatively constant deep body temperature. Broilers gain body weight very rapidly and are efficient converters of food. This is mainly achieved by controlling the response to environmental conditions in accordance with capacity of metabolic energy. Variation in body temperature is considered as a good indicator for stressed or tolerant birds.

When chicks are confronted with acute hot climate, the physiological emphasis shift from productivity to survival and a target to decline heat load must be achieved. Edens (1976) and Hamdy *et al.* (1992) reported that Na^{+} and Ca^{++} treatments were useful in tolerating hot climate.

Plasma corticosterone concentration has been used as an index of the response to stressors. Reduction of plasma thyroid hormones concentration during stress is related to elevated plasma corticosterone (Klundorf *et al.*, 1981). During times of heat stress plasma concentrations of glucocorticoids increase "Satterlee *et al.*, 1989). Exposure to stressor increased plasma corticosterone (Jones, 1988). High concentration of glucocorticoids cause atrophy and loss in skeletal muscles and thus depress growth (Bartov, 1985).

The present work was carried out to examine the influence of intravenous injection of NaCl or $CaCl_2$ on heat tolerance of Arbor Acres broiler chicks.

MATERIALS AND METHODS

Two hundred broiler chicks (Arbor Acres) obtained from a commercial hatchery at one day of age were placed in one brooding house until fifteen days of age and were provided *ad libitum* water and commercial starter diet of 22.8% crude protein and 2980 K Cal ME/Kg. On day fifteen, the chicks having body weight heavier or lighter than the mean value plus or minus twice the standard deviation, respectively, were discarded and the remaining number, a hundred and eighty chicks were used according to the experimental design.

At the start of experiment, the chicks were divided randomly and assigned to six groups each of thirty chicks. The thirty chicks of each group were subdivided into three replicates each of ten chicks. All chicks were located in one grower battery house with natural day light. Tap water and grower diet (19.6% crude protein, 3090 K Cal ME/Kg, 1.16% calcium, 0.54% phosphorus, 0.16% sodium and 0.28% chloride) were provided *ad. Libitum*. The experiment was conducted during the hotest summer months, July and August, for 4 weeks. Average of ambient temperature was 33.6±2.0°C ranging from 42.7±1.8°C to 24.9±2.3°C..Average relative humidity recorded was 65%±4% ranging from 60% to 69% for min. and max. respectively. All chicks were vaccinated against infectious bronchitis at one day of age and against Newcastle's disease at seven days of age.

The six groups applied were assigned as follows: 1: treated as sham "control"; 2: injected by avian physiological saline (0.9% NaCl); 3: injected by saline + 2% NaCl; 4: injected by saline + 4% NaCl; 5 : injected by saline + 2% $CaCl_2$; and 6: injected by saline + 4% $CaCl_2$. In each treatment one ml of the solution was injected, i.v. at biweekly intervals, at 2,4 and 6 weeks from the start of exposure to hot climate at one day of age. The solutions were made with deionized twice glass distilled water and reagent grade chemicals. After the solutions had been prepared they were filtered through a 0.4 μ millipore filter. These procedures were conducted to insure that all solutions were pyrogen free. Body weight, feed intake and feed conversion (gms feed intake/ gm body gain) were obtained biweekly to the nearest gram. Mortality rate throughout the experimental period was recorded.

Four chicks were randomly choosen from each replicate (18x4=72) to determine rectal temperature at 4 hr after injection by a clinical mercury thermometer inserted into the vent to the depth of 2 cm for one minute. Respiration rate was also recorded,

at the same time, for the same number of chicks by counting the abdomenal movement per minute.

At 2 hrs after injection, blood samples were collected in hepranized tubes from five chicks of each replicate (18x5=90) in pledge for the determination of plasma intended traits. Blood plasma was separated by centrifuging the whole blood at 3000 r.p.m. for 20 minutes. The plasma was immediately stored at -20°C till analysis. Methods used for analysis were, total Ca^{++} (Baginski *et al.*, 1974); inorganic phosphate (Goldenberg and Fernandez, 1966), glucose (Trinder, 1969) and total protein (Weichslboun, 1946).

Plasma concentration of corticosterone, thyroxin (T4) and triiodothyronin (T3) were determined using double-antibody radioimmunoassay kits (Antibodies Incorporated Davis, California USA). The standard curves ranged from 0 to 600 ng/dl, 0 to 240 ng/dl and 20 to2000 ng/ml for T3, T4 and corticosterone, respectively.

Data were statistically analyzed by the analysis of variance with the General Linear Model (GLM) procedure of the SAS Institute (SAS, 1985). All statements of significance are based on the 0.05 level of probability.

RESULTS AND DISCUSSION

1- Productive Characteristics

Data of body weight, feed intake and feed conversion are shown in Table (1). Obtained results indicated that there were no significant effect of intravenous injection treatments on those parameters with only two exceptions of the significant ($P<0.05$) increase in body weight of saline injected chicks. Also, data in Table (1) shows that saline injected chicks consumed significantly ($P<0.05$) more feed than the other groups while no significant effect of treatment was observed in feed conversion. Treatment with the expermintal solution did not cause any mortality in any group, while five chicks out of thirity (16.0%) died from the control group.

2- Physiological Responses

2-1- Rectal temperature and respiration rate:

Data of rectal temperature are presented in Table (2). Birds injected by either saline or saline + NaCl recorded significantly ($P<0.05$) higher rectal temperature than both control and birds iby saline C_2. On the contrary, the latter gtemperaturewill make the chicks less dependent on water evaporation to dissipate excess of heat. Myers (1971) suggested that the ratio between Na_+ and Ca^{++} in the posterior hypothalamus determined set point for rectal temperature. This hypothesis was based upon a series of experiments and confirmed that Na_+ when elevated in the blood, produced a hyperthermic state, while Ca^{++}, on the other hand, produced a profound hypothermia. In the present study, the significant ($P<0.05$) reduction of rectal temperature of Ca^{++} injected chicks is in good agreement with those observed by Edens (1976).

The results of rectal temperature may be concerned in explanation for those of respiration rate. Data in Table (2) show that changes in respiration rate were in an opposite direction with that of rectal temperature. Autonomically, under hot climate birds tried to increase their respiratory evaporative heat dissipation.

Table 1. Means ±SE of body weight, feed intake and feed conversion as affected by i.v. injection of NaCl or $CaCl_2$ to chicks under hot condition.

Item	weeks under hot climate	Chicks injected i.v. with					
		Sham treated chicks	saline	Saline+ 2% NaCl	Saline+ 4% NaCl	Saline+ 2% $CaCl_2$	Saline+ 4% $CaCl_2$
Body weight (g)	2	340±2	335±3	340±5	336±4	333±3	334±3
	4	905±4	920±5	910±8	915±7	926±3	918±5
	6	1585±11^b	1693±13^a	1561±12^b	1598±14^b	1580±13^b	1589±12^b
Feed intake (g)	2-4	1017±.13	998±11	970±14	985±13	1008±16	995±12
	4-6	1293±21^b	1468±23^a	1170±18^b	1299±23^b	1243±21	1274±20
Feed conversion	2-4	1.8±0.1	1.7±0.1	1.7±0.1	1.7±0.1	1.7±0.1	1.7±0.1
	4-6	1.9±0.1	1.9±0.1	1.8±0.1	1.9±0.1	1.9±0.1	1.9±0.1

a,b Means in the same row with no common superscripts are different significantly (P<0.05).

Table 2. Means ±SE of rectal temperature (°C) and respiration rate/min. as affecte by i.v. injection of NaCl or $CaCl_2$ to chicks under hot condition.

Items	weeks under hot climat	Chicks injected i.v. with					
		Sham treated chicks	Saline	Saline+ 2% NaCl	Saline+ 4% NaCl	Saline+ 2% $CaCl_2$	Saline+ 4% $CaCl_2$
Rectal Temp. (°C)	2	39.97±0.01^b	41.14±0.01^a	41.35±0.01^a	41.60±0.01^a	39.41±0.01^c	39.43±0.01^c
	4	41.53±0.01^b	41.80±0.01^a	41.82±0.01^a	41.93±0.01^a	41.16±0.01^c	41.32±0.01^c
	6	41.51±0.01^b	41.79±0.01^a	42.03±0.01^a	41.97±0.01^a	41.32±0.01^c	41.36±0.01^c
Resp. rate/min	2	45.8±1.5^b	47.4±1.3^b	46.4±1.1^b	49.5±1.2^b	59.8±2.1^a	71.8±2.3^a
	4	46.4±2.7^b	46.8±3.0^b	50.1±3.0^b	53.4±2.7^b	61.3±1.5^a	83.3±4.2^a
	6	46.7±3.8^b	44.6±3.6^b	48.6±4.2^b	50.7±3.5^b	70.4±4.8^a	90.2±4.5^a

a,b Means in the same row with no common superscripts are different significantly (P<0.05).

The present study showed that ,the chicks treated with Ca^{++} exhibited sequential behavioral responses, such as, vigorous head shaking, defecation and panting. While chicks treated by Na^{+} did not respond behaviorally different than the control. This agrees with the explanations of Hensel and Schafer (1974) reporting Ca^{++} stimulate peripheral warmth receptors, and depressed peripheral cold receptors and caused panting.

2-2- Plasma constituents

a- Plasma calcium and inorganic phosphate

Table (3) shows that plasma calcium significantly ($P<0.05$) increased in the $CaCl_2$ treated chicks than each of sham and NaCl traeted ones. However, it is very important to decline the plasma calcium level to maintain the cation homeostasis .

Analysis of the data for plasma inorganic phosphate showed that there were significant ($P<0.05$) differences among treatments (Table 3). Increased plasma inorganic phophate by Ca^{++} treatments proved that inorganic phosphate is parrallel with calcium level in chicken plasma as reported previously by Sullivan *et al.* (1992).

b: Plasma glucose

Data of plasma glucose are presented in Table (3). The obtained data show that plasma glucose decreased significantly ($P<0.05$) due to the i.v. of NaCl and $CaCl_2$ treatments. The control chicks became hyperglycemic under hot climate as judged by the simulaneous elevation of the glucose level in plasma for sham treated group. At two weeks of age there were no significant differences among treatments in plasma glucose, while, at late age (6 week) injected chicks recorded significasntly lower values than control. This trend shows that rapid changes in plasma glucose during hot climate. Hamdy (1993) reported that the rapid changes in plasma glucose during hot climate can be modified and produced by increases in plasma epinephrine and norepinephrine and decreases in adrenal catecholamines . This may be due to the effect of cortisol to face the increasing demand to energy needed for biological reactions to cope with heat stress.

c: Plasma total protein

Data on plasma total protein are presented in Table 3. Chicks treated with NaCl or $CaCl_2$ had significantly ($P<0.05$) higher plasma total protein than sham (control) treated chicks. In fact, some of the metabolic changes due to high ambient temperature increase the excretion of catecholamines, therefore, this may lead to protein catabolism and hence to nigative nitrogen balance and correspondingly an increased excretion of N in the urine (Fleck, 1980). Henry (1981) demonstrated that the increased N excretion with the urine in stressed organsim is due to the inhibition of muscle protein synthesis which results in mobilization of amino acids from the muscles increaseing the supply to the liver. The changes in plasma total protein in chicks in hot climate are caused by either excess fluid or fluid retention, increased capillary permeability, increased catabolism or decreased synthesis of protein. The present results are in good agreement with those obtained by Hamdy *et al.* (1995).

Table 3. Means ±SE of plasma constituents studied as affected by i.v. injection of NaCl or $CaCl_2$ to chicks under hot condition .

Item	weeks under hot climat	Chicks injected i.v. with					
		Sham treated chicks	saline	Saline+ 2% NaCl	Saline+ 4% NaCl	Saline+ 2% $CaCl_2$	Saline+ 4% $CaCl_2$
Plasma Ca^{++} (mg/dl)	2	9.5 ± 1.7^{b}	9.6 ± 1.5^{b}	9.4 ± 1.5^{b}	9.5 ± 1.7^{a}	10.3 ± 1.3^{a}	10.4 ± 1.3^{a}
	4	9.6 ± 1.8^{b}	9.6 ± 1.2^{b}	9.5 ± 1.4^{b}	9.7 ± 1.8^{b}	11.7 ± 1.2^{a}	10.8 ± 1.4^{a}
	6	$10.0\pm1..3^{b}$	10.0 ± 1.0^{b}	9.8 ± 1.5^{b}	9.9 ± 1.4^{b}	11.5 ± 1.6^{a}	11.4 ± 1.0^{a}
Plasma inorganic phosphate (mg/ dl)	2	4.8 ± 0.2^{b}	4.9 ± 0.1^{b}	4.6 ± 0.2^{b}	4.7 ± 0.1^{b}	5.3 ± 0.3^{a}	5.9 ± 0.2^{a}
	4	4.7 ± 0.3^{b}	4.9 ± 0.2^{b}	4.7 ± 0.1^{b}	4.7 ± 0.2^{b}	5.6 ± 0.2^{a}	5.8 ± 0.3^{a}
	6	4.8 ± 0.5^{b}	5.1 ± 0.2^{b}	4.8 ± 0.3^{b}	4.9 ± 0.2^{b}	5.8 ± 0.3^{a}	5.9 ± 0.1^{a}
Plasma glucose (mg/ dl)	2	208±11	210±9	201±12	197±13	215±10	199±14
	4	275 ± 14^{a}	238 ± 11^{b}	226 ± 13^{b}	216 ± 11^{b}	230 ± 12^{b}	210 ± 12^{b}
	6	299 ± 13^{a}	246 ± 12^{b}	254 ± 12^{b}	238 ± 11^{b}	260 ± 14^{b}	240 ± 11^{C}
Plasma protein (g/ dl)	2	4.51 ± 0.6^{b}	4.11 ± 0.7^{a}	5.18 ± 0.8^{a}	5.21 ± 0.7^{a}	5.17 ± 0.8^{a}	5.21 ± 0.8^{a}
	4	5.17 ± 0.8^{b}	5.60 ± 0.8^{a}	5.89 ± 0.9^{a}	5.75 ± 0.6^{a}	5.81 ± 1.0^{a}	5.76 ± 0.7^{a}
	6	5.81 ± 0.9^{b}	6.18 ± 0.9^{a}	6.03 ± 1.0^{a}	6.28 ± 0.8^{a}	6.18 ± 0.9^{a}	6.21 ± 0.8^{a}
Plasma T3 (ng/dl)	2	3.9 ± 0.4^{b}	4.8 ± 0.2^{a}	4.7 ± 0.6^{a}	4.6 ± 0.2^{a}	4.8 ± 0.2^{a}	4.3 ± 0.5^{a}
	4	4.4 ± 0.3^{b}	4.9 ± 0.3^{a}	5.2 ± 0.5^{a}	4.8 ± 0.3^{a}	4.9 ± 0.4^{a}	5.0 ± 0.3^{a}
	6	4.5 ± 0.2^{b}	5.2 ± 0.4^{a}	5.0 ± 0.2^{a}	5.3 ± 0.2^{a}	5.3 ± 0.4^{a}	5.2 ± 0.5^{a}
Plasma T4 (ng/ dl)	2	11.3 ± 0.3^{b}	13.4 ± 0.2^{a}	15.8 ± 0.2^{a}	15.2 ± 0.3^{a}	15.3 ± 0.2^{a}	13.8 ± 0.2^{a}
	4	12.1 ± 0.4^{b}	15.6 ± 0.6^{a}	14.9 ± 0.4^{a}	13.8 ± 0.6^{a}	13.8 ± 0.4^{a}	14.4 ± 0.4^{a}
	6	12.4 ± 0.5^{b}	15.7 ± 0.3^{a}	15.4 ± 0.6^{a}	14.9 ± 0.4^{a}	15.2 ± 0.3^{a}	13.9 ± 0.6^{a}
Plasma corticosterone (ng/ ml)	2	11.2 ± 2.1^{a}	11.3 ± 3.0^{a}	9.4 ± 2.1^{a}	8.2 ± 1.9^{b}	9.8 ± 1.8^{a}	9.2 ± 1.7^{a}
	4	13.6 ± 2.0^{a}	14.2 ± 2.1^{a}	10.2 ± 2.0^{b}	8.3 ± 1.8^{c}	10.0 ± 2.2^{b}	10.1 ± 1.9^{b}
	6	16.4 ± 2.8^{a}	16.5 ± 2.2^{a}	11.3 ± 1.5^{b}	9.5 ± 1.8^{c}	12.4 ± 2.7^{b}	12.3 ± 2.1^{b}

a,b Means in the same row with no common superscripts are different significantly ($P<0.05$).

d: Plasma T3 and T4

Data on plasma T3 and T4 concentrations for all treatments are shown in Table 3. Both plasma T3 and T4 concentrations were significantly ($P<0.05$) increased in treated chicks indicating that hyperthyrodism occurred. Similar results were obtained by Hamdy *et al.* (1995). Within certain limits, the function of the thyroid is governed by the concentration of the circulating thyroid hormones and their effects on the hypothalamic controlling pituitary relase of TSH (Mashaly, 1984; Mashaly *et al.*, 1984 and Siam, 1994). A decrease in the amount of circulating thyroid hormones, during hot environments to a level below metabolic requirements prompts the neuroendocrine controlled anterior pituitary to increase the relase of TSH.

e: Plasma corticosterone

The adrenal glands play an improtant role in the response to offensive stimulation. Data in Table (3) reveale that levels of corticosterone significantly ($P<0.05$) increased by countinous exposure to hot climate .This is consistent with previous findings of Freeman and Flack, (1981), that the heat stress significantly increased plasma corticosterone. An increase in the plasma corticosterone concentration invariably accompanies stressful, chronic stress caused adrenal hypertrophy, particularly of the inner gland zone (Harvey *et al.*, 1984). Plasma corticosterone decreased significantly ($P<0.05$) due to i.v. injection by either NaCl or $CaCl_2$

It can be concluded that i.v. injection by either NaCl or $CaCl_2$ may improve the heat tolerance of broiler chicks. However, there were no differences observed between the two concentrations (2% and 4%) used, in relation to heat tolerance.

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تأثير الحقن الوريدى بكلوريد الصوديوم أو كلوريد الكالسيوم على الاستجابة الفسيولوجية لبدارى التسمين

أحمد محمد القياتى محمد

قسم الإنتاج الحيوانى، كلية الزراعة جامعة القاهرة، الجيزة، مصر

أجريت هذه الدراسة بغرض معرفة تأثير الحقن الوريدى بكلوريد الصوديوم أو كلوريد الكالسيوم على التحمل الحرارى لبدارى التسمين. أستخدم مائة وثمانون كتكوت قسمت إلى ستة مجاميع كل مجموعة ثلاثون كذلك كل مجموعة قسمت إلى ثلاثة مكررات كل مكرر به عشرة كتاكيت. وكان توزيع المجاميع كالآتى:

١- المجموعة الأولى: عوملت كمجموعة مقارنة، ٢- المجموعة الثانية: حقنت بمحلول فسيولوجى للطيور (٠,٩٪ ص كل)، ٣- المجموعة الثالثة: حقنت بمحلول فسيولوجى +٢٪ ص كل)، ٤- المجموعة الرابعة: حقنت بمحلول فسيولوجى +٤٪ ص كل)، المجموعة الخامسة: حقنت بمحلول فسيولوجى +٢٪كا كل٢)، والمجموعة السادسة: حقنت بمحلول فسيولوجى +٤٪ كا كل).

وقد تم الحقن بـ ١سم٣ فى الوريد كل أسبوعين عند عمر ٢، ٤، ٦ أسابيع تعرضت فيها الطيور للظروف الطبيعية من ضوء وحرارة (خلال شهرى يوليو وأغسطس) وكان متوسط درجات الحرارة العظمى ٤٢,٧±١,٨°م والصغرى ٢٤,٩±٢,٣°م والرطوبة النسبية ٦٥±٤٪.

وأوضحت النتائج عدم تأثر وزن الجسم، إستهلاك العليقة ومعامل تحويل العلف (معامل التحويل الغذائى) بالحقن. بينما نفقت خمس كتاكيت من مجموعة المقارنة ولم يتم نفوق أى كتاكيت فى باقى المجاميع.

أوضحت النتائج أن كلا من ص كل و كا كل٢ دور فى التنظيم الحرارى لبدارى التسمين حيث أرتفعت درجة حرارة الجسم معنويا لمجاميع ص كل بينما إنخفضت حرارة الجسم معنويا وأرتفع معدل التنفس للمجاميع التى حقنت كا كل٢ .

أوضحت النتائج أن الحقن زاد معنويا من الكالسيوم والفسفور الغير عضوى، البروتين الكلى والهيروكسين والترای أيودوثيرونين فى بلازما الدم. بينما قلل معنوياً من جلوكوز والكورتكوستيرون.