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Effect of Heat Shock at Early Growth Phase on Glucose and Calcium Regulating Axis in Broiler Chickens

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Abstract: The present study was carried out to investigate the effect of different heat shock programs at early growth phase on glucose and calcium regulating axis in broiler chickens. The Comparison of body weight gain between different experimental groups showed that relative growth rate was significantly reduced in heat shock treated groups at first week of age. After this period, the heat exposed groups gradually showed a compensatory growth and at the end of starter period the relative growth rate was significantly higher in heat shock treated groups compared to control group. The exposure of heat shock at the beginning of rearing period (day 3-6) and at the end of rearing period (day 42) has significantly increased plasma calcium levels. After the heat shock period, no differences in plasma calcium concentrations was found. The plasma glucose level was significantly reduced during temperature conditioning in heat shock treated groups compared with control group. After heat shock conditioning, plasma glucose levels returned to normal situation indicating that no differences in plasma glucose concentrations could be detected. There were no differences in plasma triglycerids concentrations between experimental groups. The results indicate that the glucose and calcium metabolism axes is one of the most important target in pre-heat shock conditioning to reduce mortality in broilers raising. Probably, the exposure of heat shock at early growth phase in broiler chickens can improve the bird's thermotolerance ability to heat shock at higher ages by rapid response of birds to provide glucose via stored resources. In conclusion, exposure of heat shock at early growth phase in spite of improved bird's final performance and survival ability during acute heat shock at later ages, can be used as a suitable model to understand the biological phenomenon related to the mechanisms of thermoregulation.

Key words: Heat stress, thermotolerance, broilers, management

Introduction

In the recent years a considerable development has been occurred in broiler industry as a result of suitable genetic selection strategies. This genetic trends has been resulted broiler birds with higher growth rate, lower feed conversion ratio and finally heavier broiler birds in a relatively shorter growth period for broiler industry. Although, the selection strategy has been increased the size of organs related to blood and respiration systems but in parallel with this increase in relative growth rate, these organs had a slow growth pattern (Yahav and McMurty, 2001). Since, in an unoptimized raising condition birds can not balance the optimal required energy. For example, rearing broiler chicks in an unoptimized climate condition has resulted in lower production and finally lower economic gain in poultry industry (Havenstein *et al.*, 2003). It is known that birds are capable to regulate their body temperature in a limited range of temperature changes. The increases of temperature above the capacity of this regulating range may results a series of unreturn events in temperature

regulating system and finally causes bird's death (Arjona *et al.*, 1988).

For increasing the thermotolerance capacity of the birds and also inhibition of economic losses as a result of heat stress two management models has been suggested; 1) heat shock and 2) adaptation of birds to desired environmental conditions (Horowitz, 1998). These methods have provided some suitable results for broiler industry. The fast response of heat shock can be achieved by application of this technique at early growth phase (Ait *et al.*, 1995; Yahav and Hurwitz, 1996; Yahav *et al.*, 2004). This technique benefits from the unmaturing temperature regulating system in broiler chickens at early age (Modrey and Iceman, 1992). The development of this mechanism can be obtained via the activities of sympathetic nerve system, conduction of thermal information to hypothalamus and finally elevated the potential of thermotolerance in central part of thermal regulation axis (Rothwell, 1992). It has been reported that the power of this mechanism for reducing the heat production is related to thyroid hormone (T_3) activity and

Table 1: Ingredients and calculated nutrient analyses of the experimental diet at different rearing periods.

Ingredients	Starter (%)	Grower (%)	Finisher (%)
Maize	61	62	62
Soyabean meal	28.78	27.50	28
Fish meal	5.3	3.4	2
Fat	0.8	3.5	5
Limestone	0.85	1.2	1.2
Calcium phosphate	1.1	1	1
DL-methionine	0.12	0.02	0.05
L-lysine	0.05	-	-
Vitamin & mineral premix	1.06	1.06	1.06
Salt	0.27	0.27	0.27
Calculated analysis			
Crude protein	21.2	19.2	18.5
ME, Kcal/kg	2977	3123	3200
Calcium	0.96	1	0.8
Phosphorous	0.05	0.48	0.44

also heat loss's via evaporation, radiation and convection (McNabb and King, 1993). It has been shown that short time (24 h) treatment of broiler chicks to heat stress ($36\pm1^\circ\text{C}$) with 70-80 % relative humidity at the first week of life resulted a slow reduction in body weight gain. Immediately after heat stress period a compensatory growth has occurred and at the end of rearing period the body weight in heat shock treated chicks was higher than in control group (Yahav and Plavink, 1999b; Yahav *et al.*, 1995).

Recently, different researcher groups have published several reports on growth induction and also growth inhibitory mechanisms using heat stress model. The application of higher temperature at early age increases the number of satellite cells that are necessary for muscle tissue's hypertrophy (Halevy *et al.*, 2001). The effect of higher temperature at 3 day of age in broiler chicks significantly increased the number of satellite cells in compare to control group. This trend was associated considerably with IGF-I secretion (Halevy *et al.*, 2001). It has been reported that the increase of plasma T3 concentrations which is known as an important growth induction factor have a positive correlation with feed intake in turkey and broiler chicks. It has been reported that this hormone is a key player in growth regulation in broiler chicks under heat stress conditioning (Uni *et al.*, 2001). The fast compensatory growth, which will occur immediately after heat stress induction, was associated with higher plasma T3 concentrations. This pattern of T3 secretion has occurred only at compensatory growth phase since after this period plasma T3 concentrations have been decreased (Yahav and Plavink, 1999b). The plasma T3 levels has an important role in relation to growth

inhibitory effect in normal growth and also in compensatory growth phase in broiler chickens. The maximum live weight in turkey and broiler chicks can be achieved in temperature ranges of $18\text{-}20^\circ\text{C}$ (Yahav, 1999a; Yahav, 2001). It has been reported that the temperature lower than 18°C and higher than 25°C increased and decreased feed intake respectively (Hurwitz *et al.*, 1980; Yahav and Plavink, 1999b). In addition it has been shown that the induction of heat shock at incubation time increased the thermotolerance of post-hatched chicks (Moraes *et al.*, 2003; 2004). Although, the considerable attempts has been done for lowering the mortality rate via heat shock management technique in broiler rearing, but the mechanism of death, as a result of heat stress induction was not clarified well. Recently, it has been shown that feeding of broiler chicks under heat shock conditioning by glucose overcome the negative effect of heat stress on broiler performance (Hayashi *et al.*, 2001).

The present study was carried out to investigate the effect of different heat shock programs at early growth phase on glucose and calcium regulating axes in broiler chickens

Materials and Methods

Chickens, Housing and Management: The details of the experimental procedures have been explained in previous paper (Rahimi *et al.*, 2002). In brief, day-old male broiler chicks (Arian breed) were obtained from a local commercial hatchery. At the age of 1 day birds of extreme weights were discarded leaving 480 male broiler chicks to be distributed into 24 experimental groups (4 treatments six replicates) with 20 birds per replicate. All chicks were vaccinated against infectious bronchitis, Gumboro, and Newcastle disease. Lighting schedule provided 23 h of light per 24 h through the entire experiment. Water and feed were provided *ad libitum* consumption. The ingredients used in the diet and the calculated nutrient analyses are shown in Table 1.

The experiment included four treatments; 1) an unexposed control group (T1); 2) group exposed to higher temperature ($38\pm1^\circ\text{C}$) for 48 h at day 3-4 (T2); 3) chickens exposed to $38\pm1^\circ\text{C}$ at day 4 and 6 days of age for 48 h (T3); 4) group exposed to $38\pm1^\circ\text{C}$ at days 3-5 for 72 h (T4). The temperature treatment programs are presented in Table 2. The relative humidity was ranged between 70-75% throughout the rearing period. For thermotolerance evaluation chickens again were thermally challenged by exposure to $37\pm1^\circ\text{C}$ for 4 hours at 42 day of age.

Measurements: Blood samples were collected from the

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Table 2: Treatment of different heat shock programs for induction of thermotolerance in broiler chickens

Experimental groups	Heat shock programs	Time of heat shock induction
Group 1	T1	Not exposed (control group)
Group 2	T2	Heat induction at day 3
Group 3	T3	Heat induction at days 4 and 6
Group 4	T4	Heat induction at days 3-5

Table 3: The relative growth rate in broiler chicks subjected to different heat shock programs. Values are means \pm SEM

Groups	Day			
	7	17	34	42
T1	146 \pm 4a	101 \pm 1b	85 \pm 2	45 \pm 1
T2	130 \pm 3b	114 \pm 2a	86 \pm 2	50 \pm 2
T3	135 \pm 2b	104 \pm 1b	84 \pm 1	48 \pm 2
T4	131 \pm 1b	115 \pm 2a	87 \pm 2	51 \pm 1

Means with no common superscripts in columns differ significantly ($P < 0.05$)

brachial vein of 12 birds per treatment into heparinized syringes before and at the end of each treatment procedures. The collected blood samples were centrifuged at 3000 rpm for 10 min and the resulted plasma sample was stored at -20°C until their metabolite contents were assayed. The plasma triglyceride, glucose, and calcium concentrations was estimated using a commercial diagnostic kit (Ziestshimi Co., Iran).

Statistical analysis: The performance results of this experiment are documented in an earlier paper (Rahimi *et al.*, 2002). Plasma metabolic data were analyzed by analysis of variance using the General Linear Model procedure (SAS Institute, 1985) with temperature treatments as fix effects. If a significance effect of variables was calculated, means were constructed by Duncan's multiple range test.

Results and Discussion

The details of broiler performance results are presented in previously paper (Rahimi *et al.*, 2001). At the end of first week the comparison of body weight gain showed that the relative growth pattern in control group was significantly ($P < 0.05$) higher than that of heat exposed groups. But after this period the exposed groups showed gradually a compensatory growth and at the end of starter period all exposed groups showed significantly ($P < 0.05$) higher growth in compared to control group (Table 3). The relative growth rate in T2 and T4 groups was greater than that of T3 group. These results indicate that the birds at early ages show a better response to heat shock induction. The higher relative growth rate in parallel with better feed conversion rate as a result of heat shock induction in early growth phase has been reported by other research groups (Yahav and McMurty,

2001; Zhou and Yamoto, 1997). These results are very similar to the results of feed restriction model at early growth phase in broiler chickens (Yahav and Plavink, 1999b). The induction of heat shock has the same effect as quantitative feed restriction in early growth phase on feed consumption. It has been reported that the induction of feed restriction at early age by affecting on heterophil to lymphocyte ratio and heat shock protein concentration increases the bird's thermotolerance at later ages (Zulkili *et al.*, 2003).

The plasma calcium levels at the end of first and second week of ages are presented in Fig. 1. The exposed groups showed higher plasma calcium concentrations at 7-week of age and this was significantly ($P < 0.05$) higher in T3 groups in compared to control one. Since at the second week this difference was not existing it may indicate that one of the targets in heat shock induction can be calcium regulating axis. Induction of heat shock altered the calcium metabolism cycle but elimination of these environmental conditions (heat shock) has returned the calcium metabolism cycle to its normal situation. With this finding it can be stated that part of the positive response to heat stress as an induction of heat shock at early age may be as the result of hormonal response related to calcium homeostasis reactions. Since the metabolism of phosphor is dependent to calcium and interaction of this cycle with others like sodium-potassium pump, the measurement of these parameters can be also very important for understanding of thermotolerance mechanism. The higher sodium to calcium ratio as induction of heat shock has been reported by other researcher (Ait *et al.*, 1995).

The plasma glucose concentration is shown in Fig. 2. The comparison of plasma glucose levels between experimental groups at first week showed that the

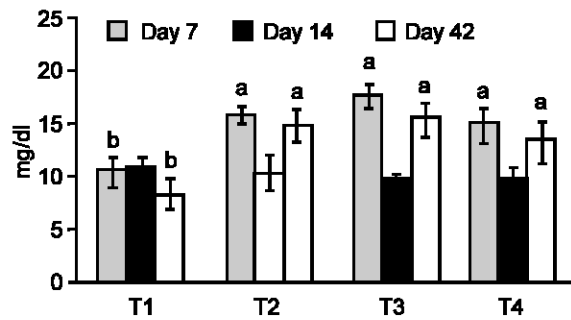


Fig. 1: The effect of heat shock on plasma calcium concentration in broiler chickens. Bars marked with different letters are significantly different ($P < 0.05$).

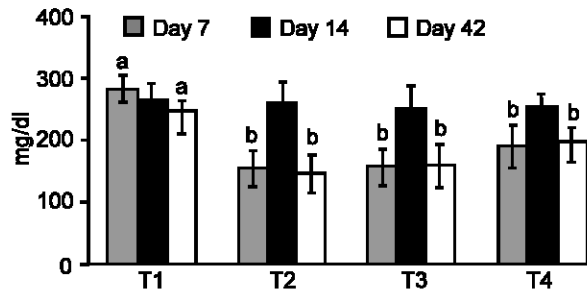


Fig. 2: The effect of heat shock on plasma glucose concentration in broiler chickens. Bars marked with different letters are significantly different ($P < 0.05$).

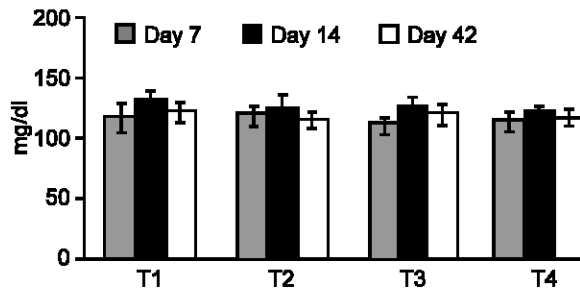


Fig. 3: The effect of heat shock on plasma triglyceride concentration in broiler chickens.

exposed groups had a significantly ($P < 0.05$) lower glucose concentration than unexposed group. But at the second week of age there were no differences in plasma glucose concentrations between experimental groups (Fig. 2). The reductions of plasma glucose concentrations as an induction of heat shock in all exposed groups indicate that the glucose metabolism cycle is one of the other important targets in heat stress procedure. Other research groups have reported the

reduction of plasma glucose concentrations by induction of heat shock. They concluded that the reduction of glucose level as the effect of heat stress might be one of the main factors for bird's death since the feeding of chicks by glucose inhibited birds' mortality (Hayashi *et al.*, 2001).

The need for glucose has increased during heat conditioning therefore, the availability of glucose sources at this time are very important for the birds. Since induction of heat stress reduces the feed consumption, the hepatic storage of glycogen might be the first available sources of energy that should be provided for the birds. There was no significant difference in triglycerides between experimental groups (Fig. 3).

The reduced hepatic glycogen in parallel with elevated corticosterone hormone concentrations in heat-treated chicks has been reported by other research groups (Hayashi *et al.*, 2001). It has been indicated that the main application of the use of heat shock technique in broiler rearing can be achieved via compensatory growth and also reduction of mortality rate (Yahav, 2001; Yahav *et al.*, 2004). It has been reported one of the main targets as an induction of heat shock in regulation of compensatory growth is thyroid and growth hormone axes (Yahav *et al.*, 2001).

Conclusion: The results of this study indicate that, the induction of heat shock at early in life may influence the ability of broiler chickens to withstand high temperatures at market age via fast response of birds to providing glucose from stored glucose resources. In conclusion, the induction of heat shock at early age in spite of improved bird's final performance and survivability during acute heat challenge at later in life can be used as a suitable model to understand the biological phenomenon related to the mechanisms of thermoregulation in birds.

Acknowledgments

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