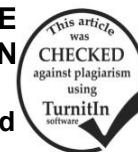


EFFECT OF EARLY HEAT STRESS ON THE PRODUCTIVE PERFORMANCE AND PHYSIOLOGICAL RESPONSE IN SOME LOCAL CHICKEN STRAINS

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ABSTRACT

This experiment was conducted to study the effect of heat stress at early age on heat shock proteins, plasma Triiodothyronine (T_3), thermal reaction, productive performance, egg quality, and immune response of Matrouh and Inshas chicks. Seven hundred and twenty, one day old chicks (360 Matrouh chicks and 360 Inshas chicks) were used in this study. Birds from each strain were divided into three groups. The first was not treated and kept as a control group (T_1), the birds in the second group were exposed to ($42^{\circ}\text{C}-43^{\circ}\text{C} \pm 1^{\circ}\text{C}$) for 4 hrs at three days of age (T_2) while birds of the third group were exposed to the same thermal treatment of the second group but at four weeks of age (T_3). After these treatments birds of (T_1 , T_2 and T_3) were raised under regular conditions. At 18 weeks of age 90 chicks from each strain (30 birds from each group) were subjected to heat stress ($42^{\circ}\text{C}-43^{\circ}\text{C} \pm 1^{\circ}\text{C}$) for 4 hrs. Blood samples were taken from (5 chicks) before and after this heat stress. At 18 wks of age 5 birds from each treatment group were slaughtered. Blood samples were taken just prior and after heat exposure for analysis. Rectal temperature (RT) and respiration rate (RR) were measured before and after exposure to heat stress. Samples of liver (1g) were frozen in liquid nitrogen for further analysis. The second period of experimental was from sexual maturity till 6 months, on the same birds from previous two strains to study the effect of heat stress on egg quality.

The important results obtained were:

- 1- Early heat conditioning birds showed increase on the expression of heat shock proteins (70 and 90).
- 2- Early heat exposure birds had the lowest plasma T_3 content compared to control and (T_3).
- 3- Early heat exposure birds had significant higher antibody titer against SRBC's than control.
- 4- Results show that body weights were the highest in early heat exposed groups only in the Matrouh strain.
- 5- Early heat exposure caused lower rectal temperature and respiration rate in both strains.
- 6- Heat stress caused an insignificant increase in plasma total protein, albumin and globulin than that of control birds.
- 7- Heat stress at early age (3-day) improved in some parameters of egg quality and egg weight. Therefore heat stress at early age can improve thermotolerance at maturity then improve in the performance of bird.

Keyword: Early heat stress, heat shock protein, T_3 hormone, body weights, egg quality.

INTRODUCTION

Poultry production during summer season faces several problems. For instance, high environmental temperature which has an adverse effect on performance of laying hens, such as egg production, egg quality, and shell

quality during summer months (Ahvar, *et al.*, 1981; Dagher 1995; Bollengierleo, *et al.*, 1999).

Heat stress causes serious losses in poultry production because it increases mortality and reduce performance in broilers and laying hens (Teeter *et al.*, 1985). As living organisms, chickens have protective measures against environmental challenges. The heat shock proteins (HSPs) are a set of proteins synthesized, in response to physical, chemical or biological stresses, including heat exposure (McCormick *et al.*, 2003; Gouter *et al.* ;2006).

Heat shock proteins (HSPs) are a group of evolutionarily conserved proteins that are conventionally, classified according to molecular size ranging from 10 to 150 KDa (Benjamin and McMillan, 1998). It has been demonstrated that, HSPs act as molecular chaperones in protein assembly and disassembly (Bukau *et al.* 2000). Heat shock protein (HSPs) play an important role in the protection and repair of cells and tissues. The HSP super family includes a number of different molecular weight class families (HSP 110, HSP 90, HSP 70, HSP 60 and HSP 47) and a group of small HSPs ranging from 16 to 40 kDa (Jonttela and Wissing, (1992), Arrigo and pLandry, 1994). The different families exhibit different functions, among which the activities of the HSP 90 family as considered to be considerably important than the others. HSP 90 can bind steroid hormone receptors, thereby regulating the activity of hormones (Joab *et al.*, 1984). There is an abundance of literature on possible techniques to alleviate the adverse effects of heat stress in broiler chickens. One of the practical approaches that had yielded promising results is altering birds abilities to cope with high ambient temperatures through early stimulation in life. Converging evidence suggests that stressful experiences during the neonatal stage can have considerable impact on various facts of an animal's physiology and behavior. Exposure of 3-day old broiler chicks to elevated temperature improved survivability. Therefore, the aim of this study was to evaluate the effects of early age heat stress on the performance and HSPs of chickens.

MATERIALS AND METHODS

This work was carried out in Inshas Poultry Research farm. Animal Production Research Institute. Dokki, Giza, Egypt.

Birds and experimental design:

The first period: 1. Early age heat exposure:

A total of seven hundred and twenty (720) one day old chicks (360 inshas chicks and 360 Matrouh chicks) were individually weighed to the nearest gram, wing banded and raised under management practice. The brooding temperature was maintained between 34°C and 37°C at the beginning of the growing period and gradually decreased every 2 to 3 days to reach 24°C ± 1 until the end of the growing period.

The birds were raised on the wood shaving litter in floor brooding pens until the end of the experimental. Chicks received starter ration containing 20% protein and 2900 kcal ME /kg diet. Water and food were provided ad-libitum.

The experiment included two strains with three treatments: T₁ (120 chicks were kept at normal temperature , control), T₂ (120 chicks, 3-day-old chicks were exposed to heat stress at (42°C – 43°C ± 1°C) for 4 hrs, then moved to normal temperature, while T₃ (120 chicks, 4 weeks old chicks were treated as T₂ in two strains. The T₂ and T₃ groups were returned to the battery brooder. At 18 wks of age, a total of 90 chickens (30 birds) from each group were subjected to heat stress (42°C - 43°C ± 1°C) for 4 hrs. Five birds were randomly taken from each treatment group and slaughtered. Blood samples were taken in heparinized tube just prior and immediately after heat exposure and centrifuged at 3000 rpm for 10 min. The plasma was stored at - 20°C for future analysis. Rectal temperature (RT) and respiration rate (RR) were measured before and after exposure to heat stress, samples of liver (1g) was frozen in liquid nitrogen and kept at(– 186°C) for further analysis.

The second period:

The period was from sexual maturity till 6 months on the same birds from previous two strains, to study the effects of heat exposure on egg quality. A total of 180 eggs (laid on 3 months in two strains) were taken from experimental groups for egg quality measurements (egg weight, egg length, egg width, yolk height, yolk color, yolk weight, yolk diameter, shell thickness, shell weight, albumen height, albumen weight, and Haugh units, albumen %, yolk%, shell%, egg shape% and egg surface area) according to Stadleman (1977).

1- Liver protein determination and electrophoresis:

The liver samples (1g) for detection of HSPs were taken from all groups, samples were homogenized in 50 ml polypropylene centrifuge tubes, using 10ml lysis buffer (20 mM tris-HCl, pH 7.5; 9g 11 NaCl, 2 mM B-mercaptoethanol). Samples were homogenised, 3 times (30 s) using ultraturax homogeniser at 20000 rpm and ice-bath intervals of 30s. Lysate was centrifuged at 31.000g for 30 min at 4°C. The resulting supernatant was collected into a fresh microcentrifuge tube.

Protein concentrations were calculated using the BCA Protein Assay Kit (Pierce Biotechnology, Rockford, IL) with BSA as the standard. Eight µl of extraction protein were loaded and separated on 10% polyacrylamide gels containing SDS (Laemmli, 1970), using the Mini-protein 11 apparatus (Bio-Rad) at a constant voltage (100 v) for 4 h.

Western blot analysis:

After fraction through SDS-polyacrylamide gels, the proteins were electrophoretically transferred to nitrocellulose membranes using the procedure of Towbin *et al.*, (1979).

2- Immune parameters:

Antibody response against SRBC was measured from 5 chickens per each treatment at 8 and 12 wks of age. Chickens were injected with 0.2 ml of 9% SRBC in 0.9% saline serum. Samples were collected at 2 wk after each injection to determine anti-SRBC primary and secondary antibody titers, respectively. Antibody production was measured by agglutination inhibition test using the microtiter technique (Trout *et al.*, 1996).

3- Triiodothyronine (T3):

Radioimmuno assay (RIA) technique was performed to measure T₃

concentration in previously collected plasma samples with the total triiodothyronine (RIA) kits (Cat, 1699). Immuno T-ECHCA BECKMAN COUNTER COMPANY) according to the manufacture's instructions. Concentration of T3 is expressed as n mol/L.

4- Thermal reactions:

A. Rectal temperature (RT):

Rectal temperature was obtained with a digital thermometers inserted approximately 2 cm into the cloaca for one minute before and after heat stress period (at 18 wks of age) for 5 birds from each treatment.

B- Respiration rate (RR):

Respiration rate per minute was obtained by visual observation before and after heat stress period (18 wks of age) for 5 birds from each treatment (counting the movement of the abdomen in 30 second and then doubled).

5- Blood Metabolites:

Total protein:

Total protein in plasma g/dl was determined colorimetrically by the biuret method (Weichelbaum , 1946).

Albumin:

Determination of albumin was carried out by colorimetric method with Bromocresol green in plasma according to (Doumas, *et al.* 1971).

Globulin:

Plasma globulin was calculated by the difference between total protein and albumin.

6. Productive performance:

Body weight (BW):

Live body weights of chicks were recorded every 2 weeks through the first experimental period of 18 wks.

6. B Weight gain (BG):

Average weight gain was calculated biweekly by subtracting the previous weeks body weight from the following one.

7. Egg quality parameters:

One hundred eighty eggs were taken in three months from each treatment group in two strains to measure interior and exterior egg quality. These measurements involved (egg weight, egg length, egg width, yolk height, yolk color, yolk weight and yolk diameter, and shell thickness, and shell weight, albumen weight, albumen height and Haugh units, albumen %, yolk%, shell %, egg shape index and egg surface area). Egg length and width were measured by using a digital caliper, while egg shell thickness was measured with digital micrometer. The tripod electronic digital micrometer was used to measure the yolk and albumen height. Yolk diameter was measured with the digital caliper (mm) and yolk color was determined by using the Roche color fan. Yolk was weighed using electronic decimal scales. Egg shape index was calculated according to Romanoff and Romanoff (1949). Haugh units were calculated according to the following equation:

$$\text{Haugh unit (HU)} = 100 * \log_{10} (h - 1.7W^{0.37} + 7.6)$$

Where

$$\text{HU} = \text{Haugh unit}$$

h = observed height of the albumen in millimeters (mm).

w = weight of egg in grams (g).

Egg surface area (ESA) was calculated according to Paganell., *et al* (1974) as

$$ESA = 4.835 \times W^{0.662} \text{ Cm}^2$$

Where W = egg weight in grams

8. Immunological parameters:

Antibody response against SRBC was measured from 5 birds in each treatment at 56 and 84 days of age. Birds were injected with 0.2 ml of 9% SRBC in 9% saline. Serum samples were collected at 7 days after each injection to determine anti-SRBC primary or secondary antibody titer, respectively. Antibody production was measured by agglutination inhibition test using the micro titer technique (Trout *et al.*, 1996).

9. Statistical analysis:

Data of Matrouh and Inshas chickens were subjected to analysis of variance using the linear general model of SAS (2003), package software version 9.1. Data were analyzed each breed separately. Data percentages (%) of egg quality were transformed to its arcsin values before the statistical analysis. Significant differences among means were tested by Duncan, (1955).

Statistical models:

Model I: was used to analyze data of body weights, body weight gain and egg quality traits

as follow: $Y_{ijk} = \mu + T_i + S_j + (TS)_{ij} + e_{ijk}$

Where:

μ = general mean, T_i = the fixed effect of i^{th} treatments (1, 2 and 3); S_j = the fixed effect of j^{th} sex; $(TS)_{ij}$; the effect of the interaction between i^{th} treatments and the j^{th} sex and e_{ijk} = the random error.

Model II: was used to analyze data of blood samples, rectal temperature and respiration rate as follow: $Y_{ijk} = \mu + T_i + E_j + (TE)_{ij} + e_{ijk}$

Where:

μ = general mean, T_i = the fixed effect of i^{th} treatments (1, 2 and 3); E_j = the fixed effect of j^{th} time of exposure to heat stress (1 and 2); $(TE)_{ij}$ = the effect of the interaction between i^{th} treatments and the j^{th} time of exposure to heat stress; and e_{ijk} = the random error.

RESULTS AND DISCUSSION

1. Heat shock protein (HSP):

Tables (1-a and 1-b) and Figure (1) shows the SDS electrophoretic pattern of proteins from liver of 18 wks of age Matrouh and Inshas strains subjected to heat stress at 42-43°C for 4 hrs. It is evident from the results that heat stress triggered hyperactivation in the expression of two major HSP families; HSP 90 and 70 KDa as particularly shown by the difference in Table 1-a. In Matrouh chickens under heat stress (T_2) led to increase HSP 90 KDa compared with control group; while, Matrouh chickens under heat stress (T_3) led to decrease HSP 90 KDa compared with control group. Inshas chickens under heat stress (T_2 and T_3) led to the appearance of HSP 90, 70 KDa, but HSP 90 and 70 were not found in control.

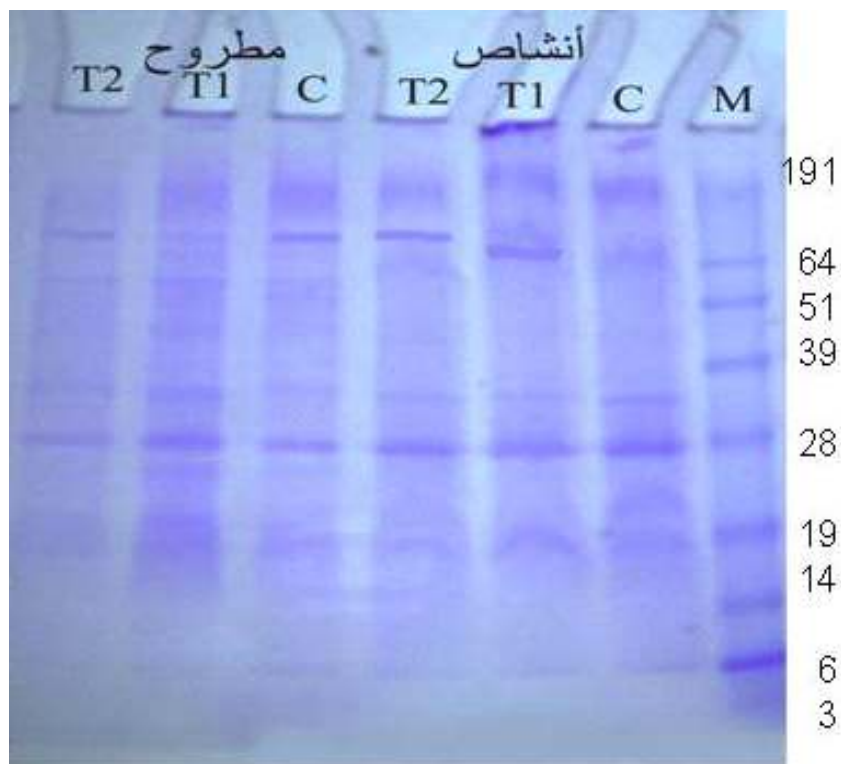


Figure (1): Electrophoretic pattern of protein fraction in liver of chickens (Matrouh and Inshas) at 18vws of age.

These results indicated that chickens under heat stress conditions stimulated HSPs (70 and 90). These results agree with these reported by Hanan, (2006) who reported that exposure to heat stress significantly increased the expression of heat shock protein 70 (HSP 70) compared with control. Jimian *et al.*, (2008) reported that HSP 70 and HSP 90 in the heart tissue of heat-stressed broilers elevated significantly after 2hrs of heat exposure. Wang and Edens (2008) showed that exposure of broiler cockerels to acute heat stress, elevated the synthesis of three HSPs(HSP-90,HSP- 70 and HSP-23) by peripheral blood leukocytes. Lei *et al.*, (2009) showed that the expression of HSP 90 increased in the heart, liver and kidney of broilers after exposure to high temperature for 2 hrs. There is a strong positive correlation between HSP expression and the digestive enzyme activity. The over expression of HSP 70 significantly increases the amylase, lipase and trypsin activities under heat stress. Hence, there is a possibility that the over expression of HSP 70 may improve intestinal digestion and absorption function under acute heat stress (Hao, *et al.* 2012).

Table (1-a): Electrophoretic pattern of protein fraction in liver of Matrouh chicken.

Matrouh							
Marker		T ₁		T ₂		T ₃	
M.W	amount	M.W	amount	M.W	amount	M.W	Amount
191	15.25	129	7.6	135	1.3	132	13.1
64	12.9	90	13.5	90	14.7	90	11.2
51	10.3	59	15.7	58	8.1	43	6.2
39	9.4	47	10.1	46	16.5	38	9.3
28	9.3	39	13.5	40	11.7	35	14.3
19	11.2	30	8.1	29	10.4	28	15.4
14	19.0	21	19.5	21	17.2	22.6	12.2
6	7.2	15	11.2	14	20.8	18	14.7
3	5.2					13	2.7

Marker = standard protein.

T₁ = control; T₂ = heat stress at 3 days of age; T₃ = heat stress at 4 weeks of age.

Table (1-b): Electrophoretic pattern of protein fraction in liver of Inshas chicken.

Inshas							
Marker		T ₁		T ₂		T ₃	
M.W	amount	M.W	amount	M.W	amount	M.W	amount
191	15.25	52	22.7	70.0	8.1	90	11.7
64	12.9	45	23	52	11.1	54	13.4
51	10.3	32	13.9	45	14.6	45	14.6
39	9.4	21	19.7	39	17.3	39	11.9
28	9.3	14.8	20.6	31	15.2	29	17.3
19	11.2			21	8.4	20	10.2
14	19.0			14.5	22.6	16	19.3
6	7.2					11	
3	5.2						

Marker = standard protein.

T₁ = control; T₂ = heat stress at 3 days of age; T₃ = heat stress at 4 weeks of age.

2. Effect of heat stress on productive performance

Body weight and weight gain:

a. Effect of heat stress on body weight of Matrouh chickens:

Table (2) showed that the effect of heat stress on live body weight (BW) of Matrouh strain at 2 and 18 weeks of age. Treatments affected body weights significantly at the early and end of period. There were significant differences due to the sex effect for body weights at 18 wks of age only (table 2). The interaction between and sex had no significant effect for body weight traits studied. Results showed that body weight gain from 2 – 18 wks of age in (G2_18) in Matrouh chickens were affected by heat stress treatments significantly. Sex had significant effect for all body weight gain traits studied. There were no significant effect due to the interaction between treatments and sex for body weight gain traits studied (table 2).

Table (2): Least square means and standard error of factors affecting body weights and body gain of Matrooh chickens.

	BW2	BW18	G218
TR:			
T ₁	100.451 ^a ±1.479	1190.143 ^b ±16.841	1090.932 ^b ±16.896
T ₂	88.213 ^c ±1.498	1274.618 ^a ±16.846	1185.464 ^a ±16.899
T ₃	95.193 ^b ±1.583	1242.329 ^{ab} ±18.146	1147.853 ^{ab} ±18.204
Sex:			
Male (M)	95.008±1.140	1350.463 ^b ±13.905	1256.130 ^a ±13.949
Female (F)	94.229±1.335	1120.931 ^b ±14.324	1026.701 ^b ±18.204
TR x sex:			
T ₁ x M	99.211±1.765	1330.714±22.051	1233.982±22.122
T ₁ x F	101.690±2.374	1049.571±25.463	0947.881±25.544
T ₂ x M	89.383±1.986	1385.469±23.574	1294.204±23.649
T ₂ x F	87.043±2.244	1163.766±24.070	1076.723±24.147
T ₃ x M	96.431±2.154	1335.205±26.424	1240.205±26.508
T ₃ x F	93.955±2.319	1149.455±24.877	1055.500±24.957

^{a, b and c} means within traits having different superscripts differ significantly ($P \leq 0.05$). T₁ = control, T₂ and T₃ = birds exposed to heat stress (42 - 43°C) for 4 hrs at (3 days and 18 weeks) of age.

b. Effect of heat stress on body weight of Inshas chickens:

Table (3) showed that the effect of heat stress on live body weight (BW) of Inshas strain from (2 – 18) wks of age. Treatments affected body weight traits studied significantly at the all period. There were significant differences due to the sex effect for body weights traits at 18 wks of age (table 3). The interaction between treatments and sex had no significant effect for body weight traits studied. Results showed also that body weight gain from 2 – 18 wks of age (G2_18) in Inshas chickens were affected significantly by heat stress treatments. Sex had significant effect for body weight gain. There were no significant effect due to the interaction between treatments and sex for body weight gain traits studied (table 3). Results show that body weight were the highest in early heat exposed groups compared to other groups. It is clearly shown that the early heat exposure for birds led to compensatory villus volume growth. This led to higher digestive capacity of small intestinal tissue. These changes may explain the changes in growth retardations followed by accelerated growth (Uni *et al.*, 2001). The increased villi length suggests an increased absorptive surface area capable of greater absorption of available nutrients (Caspary, 1992). Also, body weight gain was increased in heat stressed group compared to control in the first strain.

3. Total plasma proteins:

Heat stress caused an insignificant increase in plasma total proteins, albumin and globulin than that of control birds (Table 4) in Matrouh and Inshas chickens. These results might refer to the improvement happened in the immunity of chickens exposed to heat stress. These results suggest that early heat exposure influences T3 concentration which in turn alter the intestinal capacity to proliferate, grow and digest nutrients and the liver is site of albumin synthesis but globulin is formed by lymphatic tissue (Uni *et al.*, 2001). These results are in agreement with those of Heller *et al.*, (1979; Gross and Siegel (1983); Mashaly *et al.*, (2004); Ahmed and Nagwa, *et al.*, (2004); and where immunity increased in chickens which pre-conditioning and then exposed to heat stress at later age.

Table (3): Least square means and standard error of factors affecting body weights and body gain of Inshas chickens.

	BW2	BW18	G218
TR:			
T ₁	83.951 ^b ±1.219	1211.554 ^b ±14.828	1127.445 ^b ±14.884
T ₂	90.465 ^a ±1.187	1264.349 ^a ±14.679	1173.654 ^a ±14.734
T ₃	84.998 ^a ±1.297	1261.691 ^a ±15.795	1177.227 ^a ±15.854
Sex:			
Male (M)	85.487 ±0.979	1354.979 ^a ±13.166	1269.589 ^a ±13.215
Female (F)	87.455 ±1.037	1136.749 ^b ±11.447	1049.295 ^b ±11.489
TR x sex:			
T ₁ x M	83.111 ±1.543	1313.191 ±21.081	1229.766 ±21.159
T ₁ x F	84.792 ±1.889	1109.917 ±20.860	1025.125 ±20.938
T ₂ x M	89.259 ±1.719	1380.385 ±23.142	1290.667 ±23.229
T ₂ x F	91.672 ±1.637	1148.313 ±18.065	1056.641 ±18.123
T ₃ x M	84.672 ±1.816	1371.361 ±24.087	1288.333 ±24.177
T ₃ x F	85.900 ±1.852	1152.020 ±20.439	1066.120 ±20.515

^{a, b and c} means within traits having different superscripts differ significantly (P<0.05). T₁ = control, T₂ and T₃ = birds exposed to heat stress (42 - 43°C) for 4 hrs at (3 days and 18 weeks) of age.

Table (4): Least square means ±SE of factors affecting plasma total protein, albumin (Alb.) and globulin (Glob.) in Matrouh and Inshas chickens.

	Matrouh			Inshas		
	Protein	Alb.	Glob.	Protein	Alb.	Glob.
Treatments (T):						
T ₁	4.819 ± 0.294	2.078 ± 0.272	2.741 ± 0.241	5.418 ± 0.431	2.536 ± 0.339	2.883 ± 0.496
T ₂	5.584 ± 0.294	2.608 ± 0.272	2.975 ± 0.241	6.440 ± 0.431	2.685 ± 0.314	3.754 ± 0.459
T ₃	5.224 ± 0.294	2.721 ± 0.272	2.505 ± 0.241	5.606 ± 0.431	2.748 ± 0.314	2.856 ± 0.459
Exposure: (E)						
Before (1)	4.463 ± 0.294	2.289 ± 0.272	2.174 ± 0.197	5.341 ± 0.343	2.305 ± 0.271	3.034 ± 0.395
After (2)	5.955 ± 0.294	2.648 ± 0.272	3.307 ± 0.197	6.302 ± 0.326	3.008 ± 0.257	3.294 ± 0.375
T x E:						
T ₁ x E ₁	4.375 ± 0.416	2.160 ± 0.384	2.215 ± 0.341	4.990 ± 0.651	2.197 ± 0.514	2.793 ± 0.749
T ₁ x E ₂	5.263 ± 0.416	1.995 ± 0.384	2.628 ± 0.341	5.845 ± 0.564	2.875 ± 0.445	2.973 ± 0.649
T ₂ x E ₁	4.545 ± 0.416	2.315 ± 0.384	2.228 ± 0.341	5.800 ± 0.564	2.273 ± 0.445	3.528 ± 0.649
T ₂ x E ₂	6.623 ± 0.416	2.900 ± 0.384	3.723 ± 0.341	7.080 ± 0.564	3.098 ± 0.445	3.980 ± 0.649
T ₃ x E ₁	4.467 ± 0.416	2.393 ± 0.384	2.080 ± 0.341	5.233 ± 0.564	2.445 ± 0.445	2.783 ± 0.649
T ₃ x E ₂	5.980 ± 0.416	3.050 ± 0.384	2.930 ± 0.341	5.980 ± 0.564	3.050 ± 0.445	2.930 ± 0.649

^{a, b and c} means within traits having different superscripts differ significantly (P<0.05).

T₁ = control; T₂ = heat stress at 3 days of age; t₃ = heat stress at 4 weeks of age; Before = before exposure to heat stress; After = after exposure to heat stress.

4. Rectal temperature and respiration rate:

Table (5) showed that thermo respiratory responses (rectal

temperature and respiration rate) decreased significantly in T₂ (early heat exposure) compared with control group (non-heat exposure). Also, there was significant difference between T₂ and T₃ in rectal temperature and respiration rate. Early heat exposure at 3-5 days of age and at 8 wks or at 8 and 16 wks of age may enhance thermo-tolerance of laying hens that would face heat stress in advanced (Yahav and McMurtry, 2001). This assumption might explain the results that T₂, physiologically manipulated to better tolerate heat stress by thermal conditioning.

Table (5): Least square means of factors affecting rectal temperatures (RT) and respiration rate (RR) of Matrouh and Inshas chickens.

Items	Matrouh		Inshas	
	R.T.	R.R.	R.T.	R.R.
Treatments (T):				
T ₁	41.283 ^a ± 0.062	44.417 ^a ± 0.571	41.367 ^a ± 0.059	47.750 ^a ± 0.605
T ₂	40.783 ^b ± 0.062	40.583 ^b ± 0.571	40.833 ^b ± 0.059	43.333 ^b ± 0.605
T ₃	41.233 ^a ± 0.062	44.000 ^a ± 0.571	41.308 ^a ± 0.059	45.417 ^a ± 0.605
Over all means	41.101 ± 0.062	43.000 ± 0.571	41.169 ± 0.059	45.500 ± 0.605
Exposure (E):				
Before (1)	40.400 ^b ± 0.051	33.556 ^b ± 0.466	40.483 ^b ± 0.048	35.722 ^b ± 0.494
After (2)	41.800 ^a ± 0.051	52.444 ^a ± 0.466	41.889 ^a ± 0.048	55.944 ^a ± 0.494
Over all means	41.100 ± 0.051	43.000 ± 0.466	41.188 ± 0.048	45.833 ± 0.494
Treatment × Exposure				
T ₁ × E ₁	40.483 ^c ± 0.088	33.833 ^c ± 0.807	40.583 ^c ± 0.083	36.500 ^c ± 0.855
T ₁ × E ₂	42.083 ^c ± 0.088	55.000 ^c ± 0.807	42.150 ^c ± 0.083	59.000 ^c ± 0.855
T ₂ × E ₁	40.250 ^c ± 0.088	33.000 ^c ± 0.807	40.350 ^c ± 0.083	39.667 ^c ± 0.855
T ₂ × E ₂	41.317 ^a ± 0.088	48.167 ^a ± 0.807	41.417 ^a ± 0.083	52.000 ^a ± 0.855
T ₃ × E ₁	40.467 ^b ± 0.088	33.833 ^b ± 0.807	40.517 ^b ± 0.083	36.000 ^b ± 0.855
T ₃ × E ₂	42.000 ^a ± 0.088	54.167 ^a ± 0.807	42.100 ^a ± 0.083	56.833 ^a ± 0.855
Over all means	41.100 ± 0.088	43.000 ± 0.807	41.186 ± 0.083	45.833 ± 0.855

^{a, b and c} means within traits having different superscripts differ significantly (P<0.05).

T₁ = control, T₂ and T₃ = birds exposed to heat stress (42 - 43°C) for 4 hrs at 3 days and 18 weeks of age.

5. Effect of heat stress on plasma triiodothyronine (T₃):

The effect of heat stress on plasma triiodothyronine (T₃) of chickens at 18 wks of age are presented in Table (6). Plasma (T₃) concentration at 18 wks old chickens subjected early in life (at 3 day and 4 wks) to heat stress was significantly lower than birds in control group. After heat stress at 42°C for 4 hours, plasma T₃ concentration significantly (P≤0.001) decreased in all treatments. These findings proved the inhibitory effect of heat stress on thyroid activity. This is supported by the findings of (Brwen *et al.*, 1984) on the direct relationship between thyroid function and heat tolerance in chickens. The heat adaptation process allowing the adjustment of the metabolic rates in favour of the body heat balance. The overall heat stress depression in T₃ is in agreement with other authors, who reported a decreased thyroid activity in chickens expressed either to acute or chronic type of heat stresses (Yahauv and Plawnik, 1999; Tuo *et al.*, 2006). Mitchell and Carlisle (1992) and Geraert *et al.* (1996) found a dramatic decline of plasma T₃ in broiler chickens reared at 32°C to 35°C environmental temperature, respectively.

Table (6). Least square means ±SE of factors affecting plasma Triiodothyronine(T₃)in Matrouh and Inshas chickens exposed to early heat stress.

		Matrouh	Inshas
Treatments (T):			
T ₁		181.646±7.718 ^a	160.361±7.670 ^a
T ₂		140.546±7.718 ^b	128.773±7.670 ^b
T ₃		162.564±7.718 ^{ab}	136.513±7.670 ^b
Significance		***	***
Exposure (E):			
Before (1)		174.792±6.302 ^a	152.797±6.263 ^a
After (2)		148.379±6.302 ^b	130.967±6.263 ^b
Significance			
T * E			
T ₁	E ₁	184.296±10.915	181.297±10.848
T ₁	E ₂	178.997±10.915	139.429±10.848
T ₂	E ₁	161.082±10.915	133.255±10.848
T ₂	E ₂	120.010±10.915	124.291±10.848
T ₃	E ₁	178.997±10.915	143.840±10.848
T ₃	E ₂	146.311±10.915	129.185±10.848
Significant		n.s	n.s

a, b, c means within the same column with different superscript are significantly different; n.s =non significant; ***= significant (P≤ 0.001); Before = before exposure to heat stress; After = after exposure to heat stress.

It has been reported that thyroid hormone administration stimulate heat production with increased metabolic rate resulting in reduced thermo-tolerance (Bowen and Washar, 1985). Also, early investigations, gave an evidence the thyroid gland of birds decreased in size and activity when birds

become acclimated to high environmental temperatures (Huston and Carm, 1962; Shafic *et al.*, 1979 and Sinurat *et al.*, 1987). Arjona *et al.* (1990) showed that exposure of chickens to high temperature at 42 d of age associated with reduction in plasma T₃ concentration regardless of previous high temperature exposure. In contrast, the present study showed that an early age heat exposure resulted in a significant reduction in plasma T₃ levels. Moreover, the conditioned birds exhibited lower T₃ levels than the control during the high temperature. It appears, therefore, that heat exposure in early age can improve thermo tolerance at maturity improving the ability to reduce T₃ concentration and, consequently reduce heat production.

6. Immunological parameters:

Immune response against (SRBC), data are presented in Table (7), indicated that heat stress caused increased primary and secondary immune response against SRBC than control in Matrouh and Inshas chickens, but the differences were not significant.

Table (7). Least square means ±SE of factors affecting antibody titers against Sheep red blood cells (SRBC) of Matrouh and Inshas chickens exposed to early heat stress.

Item	Matrouh		Inshas	
	Primary antibody titers	Secondary antibody titers	Primary antibody titers	Secondary antibody titers
T ₁	4.200±0.902	4.00±1.254	6.20±0.766	5.80±1.322
T ₂	6.600±0.902	7.60±1.254	7.40±0.766	7.60±1.322
T ₃	5.800±0.902	6.60±1.254	6.40±0.766	7.20±1.322
Significance	n.s	n.s	n.s	n.s

n.s = not significant.

Primary and secondary titers against SRBC were measured at 56 & 84 days of age

Similar results, Al-Bisher (1998) studied the effect of short term heat stress on antibody production of Baladi and leghorn chickens, they found that, heat stress stimulated antibody production during primary immunization and suppressed it during secondary immunization. On the other hand, Guo and Su (2005) found that heat stress significantly inhibited the normal development of lymphoid organs and impaired the immunological competence. Mohamed (2006) and Megahid (2007) found that there were no significant difference of heat stress on antibody production against (SBRC) . Also, Donker *et al.* (1990) found that heat exposure did not reduce antibody production to (SRBC). Heller *et al.* (1979) found a significant increase in antibody titers to (SRBC) following heat stress exposure. The difference in these findings could be associated with age and type of birds used or due to the experimental methodology that applied. Regnier *et al* (1980) suggested that heat induced immunosuppression may depend on breed of bird. While, Kelley (1983) reported that effects on immune responses may depend on the length and intensity of heat exposure. Heat stress was also, reported to cause a reduction in antibody production in young chickens, this reduction could be indirectly due to increase in inflammatory cytokines under stress (Zulkifi *et al.*, 2000); which stimulates the lyothalamic production of corticotrophin releasing factor (Sapolsky *et al.*, 1987). Corticotrophin

releasing factor is known to increase adrenocorticotrophic hormone (ACTH) from the pituitary; (ACTH) then stimulates corticosterone production from adrenal gland-Corticosterone inhibits antibody production (Gross, 1992). Furthermore, heat stress is known to decrease T-helper 2 cyteokines (Wang *et al.*, 2001) which are important for antibody production (Lebman and Coffman, 1988).

7. Egg quality:

Heat stress treatments affected significantly the most egg quality traits studied except for albumen height, shell thickness, the yolk height and Haugh units and egg shape index in Matrouh strain. The effect of the month was significant for most egg quality traits studied except for shell thickness, albumen weight, Haugh units and egg surface area (Table 8-a and 9-a & Table 8-b and 9-b). There were no significant differences due to the interaction between the treatments and the months for all the egg quality traits studied. Also, heat stress treatments had a significant effect for most egg quality traits studied except for shell thickness and yolk color and height in Inshas strain.

Table (8-a): Least square means ±SE of factors affecting egg quality measurements in Matrouh chickens.

	Egg wt (gm)	Alb. wt (gm)	Yolk wt (gm)	Shell wt (gm)	Alb. %	Yolk %	Shell %	Egg length (cm)	Egg width (cm)	Yolk height (mm)
Treatments (T):										
T ₁	42.291 ^c ± 0.447	24.532 ^b ± 0.886	12.929 ^b ± 0.211	5.726 ^b ± 0.114	59.148 ^a ± 0.059	30.769 ^a ± 0.002	12.724 ^a ± 0.011	5.010 ^c ± 0.029	3.877 ^b ± 0.026	15.690 ^a ± 0.766
T ₂	47.052 ^a ± 0.447	27.390 ^a ± 0.886	13.937 ^a ± 0.206	6.202 ^a ± 0.112	58.397 ^a ± 0.059	29.540 ^a ± 0.002	13.126 ^a ± 0.011	5.223 ^a ± 0.029	4.030 ^a ± 0.026	16.640 ^a ± 0.766
T ₃	44.840 ^b ± 0.447	27.835 ^a ± 0.886	13.327 ^a ± 0.222	6.044 ^{ab} ± 0.114	63.569 ^a ± 0.059	29.647 ^a ± 0.002	12.592 ^a ± 0.011	5.097 ^b ± 0.029	3.973 ^a ± 0.026	15.446 ^a ± 0.766
Months (M):										
M ₁	39.947 ^b ± 0.447	25.448 ^a ± 0.886	11.309 ^c ± 0.219	5.014 ^b ± 0.116	65.477 ^a ± 0.059	28.517 ^c ± 0.002	11.024 ^b ± 0.011	4.930 ^b ± 0.029	3.777 ^b ± 0.026	14.043 ^b ± 0.766
M ₂	46.833 ^a ± 0.447	27.284 ^a ± 0.886	13.988 ^b ± 0.209	6.509 ^a ± 0.112	58.400 ^b ± 0.059	29.928 ^b ± 0.002	13.873 ^a ± 0.011	5.173 ^a ± 0.029	4.020 ^a ± 0.026	16.123 ^{ab} ± 0.766
M ₃	47.376 ^a ± 0.447	27.024 ^a ± 0.886	14.897 ^a ± 0.211	6.448 ^a ± 0.112	57.176 ^b ± 0.059	31.531 ^a ± 0.002	13.627 ^a ± 0.011	5.227 ^a ± 0.029	4.083 ^a ± 0.026	17.610 ^a ± 0.766
T × M :										
T ₁ × M ₁	37.909 ± 0.774	24.813 ± 1.535	11.129 ± 0.392	4.659 ± 0.205	67.581 ± 0.177	30.139 ± 0.008	10.209 ± 0.033	4.860 ± 0.051	3.740 ± 0.046	13.000 ± 1.327
T ₁ × M ₂	45.020 ± 0.774	25.214 ± 1.535	13.530 ± 0.351	6.276 ± 0.194	56.000 ± 0.177	30.039 ± 0.006	13.928 ± 0.033	5.080 ± 0.051	3.940 ± 0.046	16.820 ± 1.327
T ₁ × M ₃	43.943 ± 0.774	23.569 ± 1.535	14.131 ± 0.351	6.243 ± 0.195	53.930 ± 0.177	32.144 ± 0.006	14.633 ± 0.033	5.090 ± 0.051	3.950 ± 0.046	17.250 ± 1.327
T ₂ × M ₁	41.648 ± 0.774	24.530 ± 1.535	11.849 ± 0.351	5.269 ± 0.195	58.464 ± 0.177	28.419 ± 0.006	12.633 ± 0.033	4.970 ± 0.051	3.830 ± 0.046	16.390 ± 1.327
T ₂ × M ₂	48.950 ± 0.774	29.436 ± 1.535	14.311 ± 0.369	6.634 ± 0.195	60.464 ± 0.177	29.254 ± 0.007	13.506 ± 0.033	5.350 ± 0.051	3.070 ± 0.046	15.790 ± 1.327
T ₂ × M ₃	50.559 ± 0.774	28.204 ± 1.535	15.652 ± 0.351	6.703 ± 0.195	55.776 ± 0.177	30.964 ± 0.006	13.241 ± 0.033	5.350 ± 0.051	4.190 ± 0.046	17.740 ± 1.327
T ₃ × M ₁	40.366 ± 0.774	27.602 ± 1.535	10.950 ± 0.392	5.116 ± 0.205	69.713 ± 0.177	27.018 ± 0.008	10.304 ± 0.033	4.960 ± 0.051	3.760 ± 0.046	12.740 ± 1.327
T ₃ × M ₂	46.530 ± 0.774	27.204 ± 1.535	14.122 ± 0.369	6.616 ± 0.195	58.720 ± 0.177	30.492 ± 0.007	14.189 ± 0.033	5.090 ± 0.051	4.050 ± 0.046	15.760 ± 1.327
T ₃ × M ₃	47.625 ± 0.774	29.300 ± 1.535	14.908 ± 0.392	6.399 ± 0.195	62.074 ± 0.177	31.486 ± 0.006	13.435 ± 0.033	5.240 ± 0.051	4.110 ± 0.046	17.840 ± 1.327

^{a, b and c} means within traits having different superscripts differ significantly (P<0.05).

T₁ = control, T₂ and T₃ = birds exposed to heat stress (42 - 43°C) for 4 hrs at 3 days and 18 weeks of age.
M = month.

Table (9-a): Least square means \pm SE of factors affecting egg quality measurements in Matrouh chickens.

	Alb. height (mm)	Haugh units	Egg surface area (cm ²)	Shell thickness (mm)	Yolk diameter (mm)	Egg shape %	Yolk color
Treatments (T) :							
T ₁	5.449 ^a \pm 0.204	72.793 ^a \pm 4.208	55.807 ^c \pm 0.416	42.166 \pm 0.709	36.680 ^c \pm 0.036	77.582 ^a \pm 0.009	5.575 ^a \pm 0.153
T ₂	5.798 ^a \pm 0.204	75.026 ^a \pm 4.208	60.185 ^a \pm 0.416	41.500 \pm 0.682	39.770 ^a \pm 0.035	77.274 ^a \pm 0.009	5.133 ^b \pm 0.147
T ₃	5.535 ^a \pm 0.204	72.600 ^a \pm 4.208	58.175 ^b \pm 0.416	40.733 \pm 0.694	38.620 ^b \pm 0.036	78.338 ^a \pm 0.009	5.275 ^{ab} \pm 0.153
Months (M) :							
M ₁	5.384 ^b \pm 0.212	68.147 ^a \pm 4.208	53.650 ^b \pm 0.416	40.829 \pm 0.722	37.320 ^b \pm 0.037	76.756 ^a \pm 0.009	4.883 ^b \pm 0.159
M ₂	6.398 ^a \pm 0.200	79.221 ^a \pm 4.208	60.023 ^a \pm 0.416	41.733 \pm 0.682	38.590 ^a \pm 0.035	77.889 ^a \pm 0.009	5.567 ^a \pm 0.147
M ₃	5.000 ^b \pm 0.196	73.051 ^a \pm 4.208	60.494 ^a \pm 0.416	42.067 \pm 0.682	39.170 ^a \pm 0.034	78.541 ^a \pm 0.009	5.533 ^a \pm 0.147
T \times M :							
T ₁ \times M ₁	5.537 \pm 0.380	63.759 \pm 7.288	51.659 \pm 0.721	41.000 \pm 1.320	35.750 \pm 0.066	77.206 \pm 0.028	5.125 \pm 0.284
T ₁ \times M ₂	5.980 \pm 0.340	81.364 \pm 7.288	58.380 \pm 0.721	41.900 \pm 1.181	36.000 \pm 0.059	77.680 \pm 0.028	5.700 \pm 0.254
T ₁ \times M ₃	4.830 \pm 0.340	73.258 \pm 7.288	57.383 \pm 0.721	43.600 \pm 1.181	38.800 \pm 0.059	77.861 \pm 0.028	5.900 \pm 0.254
T ₂ \times M ₁	5.340 \pm 0.340	78.667 \pm 7.288	55.256 \pm 0.721	40.600 \pm 1.181	39.200 \pm 0.059	77.112 \pm 0.028	5.900 \pm 0.254
T ₂ \times M ₂	6.755 \pm 0.358	72.401 \pm 7.288	61.934 \pm 0.721	42.000 \pm 1.181	40.220 \pm 0.062	76.129 \pm 0.028	5.500 \pm 0.254
T ₂ \times M ₃	6.255 \pm 0.340	74.012 \pm 7.288	63.363 \pm 0.721	41.900 \pm 1.181	39.900 \pm 0.059	78.534 \pm 0.028	5.000 \pm 0.254
T ₃ \times M ₁	5.225 \pm 0.380	62.017 \pm 7.288	54.755 \pm 0.721	40.889 \pm 1.245	37.000 \pm 0.066	75.140 \pm 0.028	4.625 \pm 0.284
T ₃ \times M ₂	6.460 \pm 0.340	83.900 \pm 7.288	59.755 \pm 0.721	41.300 \pm 1.181	39.560 \pm 0.062	79.803 \pm 0.028	5.500 \pm 0.254
T ₃ \times M ₃	4.920 \pm 0.340	71.883 \pm 7.288	60.735 \pm 0.721	40.700 \pm 1.181	39.300 \pm 0.059	79.147 \pm 0.028	5.700 \pm 0.254

^{a, b and c} means within traits having different superscripts differ significantly (P \leq 0.05).

T1 = control, T2 and T3 = birds exposed to heat stress (42 - 43°C) for 4 hrs at 3 days and 18 weeks of age. M = month

There were significant differences due to the effect of month, for most of egg quality traits studied (Table 8-a and 9-a & Table 8-b and 9-b). The interaction between treatments and month had no significant effect for all egg quality traits studied.

There were significant (P \leq 0.001) differences in mean values of egg weigh in two strains (Matrouh and Inshas) under heat stress treatments as compared to control group (Table 8-a and 9-a & Table 8-b and 9-b). Significant differences among treatments were observed in egg surface area (ESA) Table (10). In two strains, the effect of the month was significant for (ESA), but there were no significant differences due to the interaction between the treatments and the month. The results showed that heat stress at early age (T2) improved egg quality and egg weight, this may be due to the

reduction of respiration rate and increased in body weight. In agreement with, Arad *et al.*, (1981) suggested that acclimation of laying hens to high temperatures prevents perturbation in the maintenance of several physiological processes including thermoregulations, acid-base balance and temperature-induced. From pervious results, it can be concluded that, the exposure of chickens to heat stress (42-43°C) for 4 hrs at 3 days and 18 wks of age may alleviate some negative effects of heat stress on the performance of chicks and laying hens during egg production period.

Table (8-b): Least square means ±SE of factors affecting egg quality measurements in Inshas chickens.

	Egg wt (gm)	Alb. wt (gm)	Yolk wt (gm)	Shell wt (gm)	Alb. %	Yolk %	Shell %	Egg length (cm)	Egg width (cm)	Yolk height (mm)
Treatments (T) effect:										
T ₁	39.603 ^c ± 0.497	21.634 ^c ± 0.439	12.561 ^d ± 0.227	5.748 ^d ± 0.621	55.171 ^a ± 0.012	31.304 ^a ± 0.003	14.491 ^a ± 0.001	4.930 ^b ± 0.039	3.763 ^c ± 0.026	16.830 ^a ± 0.355
T ₂	46.319 ^a ± 0.497	26.164 ^a ± 0.439	13.701 ^a ± 0.223	6.455 ^a ± 0.621	56.525 ^a ± 0.012	29.521 ^b ± 0.003	13.925 ^b ± 0.001	5.180 ^a ± 0.039	3.973 ^a ± 0.026	17.620 ^a ± 0.355
T ₃	43.298 ^b ± 0.497	24.594 ^b ± 0.439	12.799 ^b ± 0.227	6.271 ^a ± 0.621	57.564 ^a ± 0.012	29.470 ^b ± 0.003	14.474 ^a ± 0.001	5.123 ^b ± 0.039	3.870 ^b ± 0.026	16.580 ^a ± 0.355
Months (M) effect:										
M ₁	38.633 ^c ± 0.497	22.819 ^b ± 0.439	11.006 ^c ± 0.231	5.515 ^c ± 0.621	59.564 ^a ± 0.012	28.232 ^b ± 0.003	14.293 ^a ± 0.001	4.823 ^c ± 0.039	3.740 ^c ± 0.026	15.816 ^b ± 0.355
M ₂	43.550 ^b ± 0.497	23.925 ^b ± 0.439	13.360 ^b ± 0.223	6.265 ^b ± 0.621	54.763 ^b ± 0.012	30.776 ^a ± 0.003	14.378 ^a ± 0.001	3.110 ^b ± 0.039	3.883 ^b ± 0.026	17.513 ^a ± 0.355
M ₃	47.038 ^a ± 0.497	25.649 ^a ± 0.439	14.696 ^a ± 0.223	6.693 ^a ± 0.621	54.436 ^b ± 0.012	31.304 ^a ± 0.003	14.217 ^a ± 0.001	5.300 ^a ± 0.039	3.983 ^a ± 0.026	17.700 ^a ± 0.355
T × M :										
T ₁ × M ₁	36.000 ± 0.861	21.597 ± 0.761	10.210 ± 0.407	5.214 ± 0.215	60.967 ± 0.003	27.671 ± 0.009	14.474 ± 0.019	4.640 ± 0.068	3.660 ± 0.045	16.220 ± 0.616
T ₁ × M ₂	39.440 ± 0.861	20.520 ± 0.761	13.210 ± 0.386	5.710 ± 0.215	51.940 ± 0.003	33.445 ± 0.009	14.477 ± 0.019	5.000 ± 0.068	3.760 ± 0.045	17.180 ± 0.616
T ₁ × M ₃	43.370 ± 0.861	22.787 ± 0.761	14.264 ± 0.386	6.319 ± 0.215	51.940 ± 0.003	32.889 ± 0.009	14.518 ± 0.019	5.150 ± 0.068	3.870 ± 0.045	17.090 ± 0.616
T ₂ × M ₁	41.030 ± 0.861	23.527 ± 0.761	11.821 ± 0.386	5.682 ± 0.215	57.325 ± 0.003	28.811 ± 0.009	13.482 ± 0.019	4.930 ± 0.068	3.840 ± 0.045	16.780 ± 0.616
T ₂ × M ₂	47.220 ± 0.861	26.732 ± 0.761	13.810 ± 0.386	6.678 ± 0.215	56.587 ± 0.003	29.257 ± 0.008	14.131 ± 0.019	5.200 ± 0.068	4.000 ± 0.045	17.950 ± 0.616
T ₂ × M ₃	50.709 ± 0.861	28.234 ± 0.761	15.471 ± 0.386	7.004 ± 0.215	55.665 ± 0.003	30.500 ± 0.008	13.803 ± 0.019	5.410 ± 0.068	4.080 ± 0.045	18.130 ± 0.616
T ₃ × M ₁	38.870 ± 0.861	23.333 ± 0.761	10.986 ± 0.386	5.650 ± 0.215	60.385 ± 0.003	28.218 ± 0.008	10.101 ± 0.019	4.900 ± 0.068	3.720 ± 0.045	14.450 ± 0.616
T ₃ × M ₂	43.990 ± 0.861	24.523 ± 0.761	13.060 ± 0.386	6.407 ± 0.215	55.752 ± 0.003	29.802 ± 0.008	14.528 ± 0.019	5.130 ± 0.068	3.890 ± 0.045	17.410 ± 0.616
T ₃ × M ₃	47.035 ± 0.861	25.927 ± 0.761	14.353 ± 0.386	6.755 ± 0.215	55.096 ± 0.003	30.534 ± 0.008	14.329 ± 0.019	5.340 ± 0.068	4.000 ± 0.045	17.880 ± 0.616

a, b and c means within traits having different superscripts differ significantly (P<0.05).

T1 = control, T2 and T3 = birds exposed to heat stress (42 - 43°C) for 4 hrs at 3 days and 18 weeks of age. M = month

Table (9-b): Least square means \pm SE of factors affecting egg quality measurements in Inshas chickens.

	Alb. height (mm)	Haugh units	Egg surface area (cm ²)	Shell thickness (mm)	Yolk diameter (mm)	Egg shape %	Yolk color
Treatments (T) :							
T ₁	5.030 ^b \pm 0.212	75.055 ^b \pm 1.831	53.275 ^c \pm 0.470	40.700 \pm 0.621	36.840 ^a \pm 0.0430	76.771 ^a \pm 0.006	5.567 \pm 0.114
T ₂	5.973 ^a \pm 0.212	80.997 ^a \pm 1.831	59.505 ^a \pm 0.470	41.833 \pm 0.621	38.480 ^a \pm 0.0430	76.861 ^a \pm 0.006	5.500 \pm 0.114
T ₃	5.606 ^{ab} \pm 0.212	79.756 ^{ab} \pm 1.831	56.750 ^b \pm 0.470	42.033 \pm 0.621	37.630 ^{ab} \pm 0.0430	75.686 ^a \pm 0.006	5.767 \pm 0.114
Months (M) :							
M ₁	5.206 ^b \pm 0.212	78.994 ^a \pm 1.831	52.358 ^c \pm 0.470	42.500 ^a \pm 0.621	36.290 ^b \pm 0.0430	77.850 ^a \pm 0.006	5.733 \pm 0.114
M ₂	6.206 ^a \pm 0.212	83.181 ^a \pm 1.831	56.986 ^b \pm 0.470	41.467 ^{ab} \pm 0.621	37.930 ^a \pm 0.0430	76.161 ^{ab} \pm 0.006	5.433 \pm 0.114
M ₃	5.196 ^b \pm 0.212	73.632 ^b \pm 1.831	60.186 ^a \pm 0.470	40.600 ^b \pm 0.621	38.730 ^a \pm 0.0430	75.288 ^b \pm 0.006	5.667 \pm 0.114
T \times M :							
T ₁ \times M ₁	5.320 \pm 0.367	80.675 \pm 3.172	49.822 \pm 0.815	41.909 \pm 1.075	34.800 \pm 0.073	79.341 ^a \pm 0.019	5.500 \pm 0.197
T ₁ \times M ₂	5.500 \pm 0.367	79.139 \pm 3.172	53.148 \pm 0.815	40.900 \pm 1.075	37.110 \pm 0.073	75.378 ^{ab} \pm 0.019	5.500 \pm 0.197
T ₁ \times M ₃	4.270 \pm 0.367	65.353 \pm 3.172	56.855 \pm 0.815	39.300 \pm 1.075	38.600 \pm 0.073	78.041 ^b \pm 0.019	5.700 \pm 0.197
T ₂ \times M ₁	5.030 \pm 0.367	76.743 \pm 3.172	54.635 \pm 0.815	42.900 \pm 1.075	37.300 \pm 0.073	71.013 \pm 0.019	5.600 \pm 0.197
T ₂ \times M ₂	6.850 \pm 0.367	86.336 \pm 3.172	60.373 \pm 0.815	41.600 \pm 1.075	38.440 \pm 0.073	75.503 \pm 0.019	5.300 \pm 0.197
T ₂ \times M ₃	6.040 \pm 0.367	79.912 \pm 3.172	63.490 \pm 0.815	41.000 \pm 1.075	39.700 \pm 0.073	76.123 \pm 0.019	5.600 \pm 0.197
T ₃ \times M ₁	5.270 \pm 0.367	79.566 \pm 3.172	52.600 \pm 0.815	42.700 \pm 1.075	36.780 \pm 0.073	75.378 \pm 0.019	5.100 \pm 0.197
T ₃ \times M ₂	6.270 \pm 0.367	84.070 \pm 3.172	57.437 \pm 0.815	41.900 \pm 1.075	38.220 \pm 0.073	75.951 \pm 0.019	5.500 \pm 0.197
T ₃ \times M ₃	5.280 \pm 0.367	75.633 \pm 3.172	60.213 \pm 0.815	41.500 \pm 1.075	37.900 \pm 0.073	74.979 \pm 0.019	5.700 \pm 0.197

^{a, b and c} means within traits having different superscripts differ significantly (P \leq 0.05).

T1 = control, T2 and T3 = birds exposed to heat stress (42 - 43°C) for 4 hrs at 3 days and 18 weeks of age. M = month

Table (10): Least square means \pm SE of factors affecting egg surface area in Matrouh and Inshas chickens.

	Breed 1 (Matrouh)	Breed 2 (Inshas)
Treatments (T) effect:		
T ₁	55.807 ^c \pm 0.416	53.275 ^c \pm 0.470
T ₂	60.185 ^a \pm 0.416	59.505 ^a \pm 0.470
T ₃	58.175 ^b \pm 0.416	56.750 ^b \pm 0.470
Months (M) effect:		
M1	53.650 ^b \pm 0.416	52.358 ^c \pm 0.470
M2	60.023 ^a \pm 0.416	56.986 ^d \pm 0.470
M3	60.494 ^a \pm 0.416	60.186 ^a \pm 0.470
T \times M effect:		
T ₁ \times M1	51.659 \pm 0.721	49.822 \pm 0.815
T ₁ \times M2	58.380 \pm 0.721	53.148 \pm 0.815
T ₁ \times M3	57.383 \pm 0.721	56.855 \pm 0.815
T ₂ \times M1	55.256 \pm 0.721	54.635 \pm 0.815
T ₂ \times M2	61.934 \pm 0.721	60.373 \pm 0.815
T ₂ \times M3	63.363 \pm 0.721	63.490 \pm 0.815
T ₃ \times M1	54.755 \pm 0.721	52.600 \pm 0.815
T ₃ \times M2	59.755 \pm 0.721	57.437 \pm 0.815
T ₃ \times M3	60.735 \pm 0.721	60.213 \pm 0.815

^{a, b and c} means within traits having different superscripts differ significantly ($P \leq 0.05$).

T₁ = control, T₂ and T₃ = birds exposed to heat stress (42 - 43°C) for 4 hrs at 3 days and 18 weeks of age. M = month

CONCLUSION

It is concluded that early age heat conditioning induced a long-term mechanism that acts to reduce hyperthermia during heat challenge and therefore improvement the performance of bird.

REFERENCES

- Al-Bisher, A. A., S. I. Al-Mufarroj, A. K. A. Ali and M. F. Hussein (1998). Effect of short term heat stress on antibody production and blood constituents of Baladi and Leghorn chicken. *J. Appl. Anim. Res.*, 13:119 – 128.
- Ahmed, Nagwa, A., El-Tantawy, S.M.T., Khadr, Amina, F. and El-Badry, A.S.O. (2004). Effect of early heat exposure and feeding system on physiological responses and productive performance of ducting under heat stress conditions. *Egyptian J. Anim. Prod.*, 41, supp. Issue, Nov. 485-499.

- Ahvar, E., Petersen, J., Horst, P. and Thein, H. (1981). Changes in egg quality during the first laying period affected by high ambient temperature. *Archiv Für Gejiügelkunde*, 46: 1-8.
- Arad, Z., Mordr, J. and Soller, M. (1981). Effect of gradual acclimation to temperatures up to 44°C on production performance of the desert Bedouin fowl, the commercial White Leghorn and their reciprocal cross breeds. *Br. Poult. Sci.*, 22 (6): 511-520.
- Arjona, A. A., D. M. Denbow and W. D. Weaver Jr. (1990). Mortality induced thermotolerance physiological responses. *Comp. Biochem. Phsiol.* 95A: 393 – 399.
- Arrigo, A.P. and Plandry, J. (1994). Expression and function of the low-molecular-weight heat shock proteins, in, Morimoter, R.I., Tissieres, A. and Georgopoulos, C. (EDS). *The Biology of heat shock proteins and Molecular chaperones, protein.* 335-373 (New York, Gold Spring Harbor Laboratory press).
- Benjamin, I.J. and McMillan, D.R. (1998). Stress Heat shock proteins molecular chaperones in cardiovascular biology and disease. *Crirc, Res.*, 83: 117-132
- Bollengier-leo, S., Williams, P.E.V. and Whitehead, C.C. (1999). Nutrient requirements of poultry at high temperature. Page 112 in *Poultry Production in Climates* N.J. Dagher, ed. University press, Cambridge, A.M.
- Bowen Sj, and Washar Kw (1985). Thyroid and adrenal response to heat stress in chickens and quail differing heat tokrance. *Poult. Sc.*, 64: 149 – 154
- Brwen, S. J., K. W. Washburn, and M. Husten, (1984). Involvement of the thyroid gland in the response of the young chicken to heat stress. *Poult. Sci.* 63: 66 – 69.
- Bukau B, Deuerling E, Pfund C., raig E A (2000) Getting newly synthesized proteins into shape. *Cell* 10(2): 119 – 122.
- Caspary, W.F. (1992). Physiology and pathophysiology of intestinal absorption. *An. J. Clin. Nutr.* 55: 2995 -3085.
- Dagher, N. J. (1995) *Nutrient requirements of poultry production in hot climates* N. J. Daghired University Press. Ambridge A. M.
- Donker, R. A. Nieuwland, M. G. B. Zipp. A. V. and An – Der (1990). Heat stress influences on antibody production in chickens lines selected for high and low immune responsiveness. *Poult. Sci.*, 69: 599 – 607.
- Doumes, B.T., Watson, W. and Biggs, H.G. (1971). Direct colourimetric method for serum albumin measurements with bromocresol green (BCG) in plasma. *Clin.Chim. Acat.*, 31, 87-96.
- Duncan, D.B., 1955. Multiple ranges and multiple F. tests *Biometrics.* 11:1-42.
- Geraet PA., Padiha Jfc, Guilaumins (1996). Metabolic and endocrine changes induced by chronic heat exposure in broiler chickens.
- Gouter M.T., Ware, L.B., Howard, M., Roux, J., Gartland, B., Mattay, M.A., Fleshner, M. and Pitter, J. (2006). Extracellular heat shock protein 72 is a marker of the stress protein response in acute long injury. *Am. J. Physiol. Lung Molphysiol* 291: L 354- L 361.

- Gross, W.B. and Siegel, H.S. (1983). Evaluation of the heterophil, lymphocyte ratio as a measure of stress in chickens. *Avian Dis.*, 27: 972 – 978.
- Gross, W. B. (1992). Effect of short –term exposure of chickens to corticosterone on resistance to challenge exposure with *Escherichia coli* and antibody response to sheep erythrocytes. *Am. J. Vet. Res.* 53:291 – 293.
- Guo. C.J., and H. X. Su, 2005. Epithelial tissue. Pages 19 – 32 in *Histology and Embryology*. Y. M. Goa and T.B. Song. Ed people, Medical Publishing House. Beijing. China.
- Hanan, S.M. (2006). Some physiological and immunological measurements in local breed of chickens after heat stress. M.Sc., Thesis, Fac., Agric., Cairo, Univ., Giza, Egypt.
- Hao, Y., Gu, X.H. and Wang, L. (2012). Over expression of heat shock protein 70 and its relationship to intestine under acute heat stress in broilers: 1. Intestinal structure and digestive function. *Poult. Sci.*, (91) 781-789.
- Heller, E.D., D.B. Nathan and M. Perek (1979). Short heat stress as an immunostimulant in chicks. *Avian Pathol.* 8: 195 – 203
- Huston, T. M. and D. L. Carmon (1962). The influence of high environmental temperature on thyroid size of domestic fowl, *Poult. Sci.*, 41: 175 – 179.
- Jimian, Y., Endong, B., Jianyan, Y. and Lei, L. (2008). Expression and location of HSPs in the heart and blood vessel of heat stressed broilers. Articles from cell stress and chaperones are provided courtesy of cell stress society international. 13 (3): 327-335.
- Joab, I., Radonyl, C., Renoir, M., Buchou, T., Catell, I.M.G., Birnet, N., Mester, J. and Bauleu, E.E. (1984). Common non-hormone binding component in non-transformed chick oviduct receptors of four steroid hormones. *Nature.*, 308: 850-856.
- Jonttela, M. and Wissing, D. (1992). Emerging role of heat shock proteins in biology and medicine. *Annals of Medicine*, 24: 249-258.
- Kelly, K. W. 1983. immunobiology of domestic animal as affected by hot and cold weather – trans. *An. Soc. Agric.* 26: 834 – 840.
- Laemmli, U.K., (1970). Cleavage of structural proteins during the assembly of the head bacteriophage T4. *Nature* 227: 680-685.
- Lebman, D. A., and R. L. Coffman. 1988. Interleukin 4 causes isotype switching to IgE in T cell – stimulated clonal Bcell cultures, *J. Exp. Med.* 188: 853 – 862.
- Lei, L., Jimian and Endong, B. (2009). Expression of heat shock protein 90 (HSP 90) and transcription of its corresponding mRNA in broilers exposed to high temperature. *Br. Poult. Sci.*, 50: 4, 504-511.
- Mashaly, M.M., Hendricks, G.L., Kalama, M.A., Gehad, A.E., Abass, A.O. and Patterson, P.H. (2004). Effect of heat stress on production parameters and immune responses of commercial laying hens. *Poult. Sci.*, 83: 886-894.
- McCormick P.H., Chen, G., Henrey, S., Kelly, C.J. and Bouchier-Hayes, D.J. (2003). Clinically relevant thermal preconditioning attenuates ischemia-reperfusion injury. *J. Surg Res.*, 109: 24-30.

- Megahid, A. A. (2007). Measurements to heat shock protein and immunity chicks of some local strains of chickens. M. Sc. Thesis. Fac.Agric. Cairo Univ. Giza, Egypt.
- Mitchell, M. A., and A. J. Carlisle, (1992). The effects of chronic exposure to elevated environmental temperature on intestinal morphology and nutrient absorption in the domestic fowl (*Gallus domesticus*). *Comp. Biochem. Physiol.* 101 A : 137 – 142.
- Mohamed, H.S (2006) . Some physiological and immunological measurements in local breed of chickens after heat stress. M. Sc. Thesis, Fac. Agric., Cairo Univ. Giza, Egypt.
- Paganelli, C.V., Olszouka, A. and Ar, A. (1974). " The avian egg " surface area, volume and density – *The condor*, 79: 319-325.
- Regnier. J. A. K., Kelley, and C. T. Gaskins. (1980). Acute thermal stressed and synthesis of antibodies in chickens. *Poult. Sci.* 59: 985 – 990.
- Romanoff, A.L. and Romanoff, A.J. (1949). " The avian egg " John Wiley and Sons, Inc., New York, U.S.A.
- Sapolsky, R. C. Rivier, G. Yamamoto, P. Piotsky, and W. Vale (1987). Interleukin – stimulates the secretion the of hypothalamic corticotropin – releasing factor. *Science* 238: 522 – 524.
- SAS, copyright (2003) version, 9.1, SAS institute Inc. Cary 27513, USA all rights reserved.
- Shafic, M. M., A. M Borady and N. H Abdel Mutael (1979). Acclimation of Fayoumi chickens to constant and varying temperatures. *Egypt. J. Anim. Prod.*, 19: 187 – 193.
- Sinurat, A. P., D. Balance and G. H. McDouell (1987). Growth performance and concentrations of thyroid temperature. *Australian. J. Biol. Sci.*, 40(4) : 443 – 450.
- Stadleman, W.J. (1977). Quality identification of shell egg. In. *Egg Science and Technology*. 2nd Ed. by W.J. Stadleman and O.J. Collierill pup. AVI publisher company. Inc. Connecticut USA.
- Teeter, R.G., Smith, M.O., Owens, F.N. and Arp, S.C. (1985). Chronic heat stress, and respiratory alkalosis: Occurrence and treatment in broiler chicks. *Poult. Sci.*, 64: 1060-1064.
- Towbin, H., T. Stahelin, and J. Cordon. (1979). Electrophoretic transfer of protein from polyacrylamide gels to nitrocellulose sheets: procedure and some application. *Proceedings of the National Academy of Sci. USA*. 76: 4350 – 4354.
- Trout, J. M., Mashaly, M. M. and Siegel, H. S. (1996). Changes in blood and spleen lymphocyte populations following antigen challenge in immune male chickens. *Br. Poult. Sci.*, 37: 819 – 827.
- Tuo, X, Zhang Zy, Dong H. Zhang H, Xin H (2006). Responses of thyroid hormones of market size broilers to thermoneutral constant and Wam cyclic temperatures. *Poult. Sci.*, 85: 1520 – 1528.
- Uni, Z., Gal-Garloer, O., Geyra, A., Shlan, D. and Yahav, S. (2001). Changes in growth and function of chick small intestine epithelium due to early thermal conditioning. *Poult. Sci.* 80: 438-445.

- Wang, S. W. Xu, and Q Cao. (2001). The influence of stress inhibition on the plasma level of LPS, pre inflammatory and Th1/Th2 cytokines in severely scalded rts. Zhanghue Shang Zazhi. 17: 177 – 180.
- Wang, S. and F.W. Edens(2008). Involvement of steroid hormones, corticosterone and testosterone in synthesis of heat shock proteins in broiler chickens. International from Journal of Poultry Science. 2008, 7: 8, 788-797.
- Weichelbaum T. E. C. (1946). Am. J. Clin. Pathol., 7: 40.
- Yahauv, S. and Plawnik, 1 (1999). Effect of early age thermal conditioning and food restriction on performance and thermotolerance of male broiler chickens. Br. Poult. Sci., 40: 120 – 126.
- Yahav, S. and McMurtry, J.P. (2001). Thermotolerance acquisition in broiler chickens by temperature conditioning early in life. The effect of timing and ambient temperature. Poult. Sc., 80: 1662-1666.
- Zulkifi, I. M. T. che Norma, D. n. Israf, and A. R. Omar, (2000). The effect of early age, feed restriction on subsequent response to high environment temperature in female broiler chickens. Poult., Sci. 79: 1401 – 1407.

تأثير التعريض الحراري المبكر على الاداء الانتاجي والاستجابة الفسيولوجية في بعض سلالات الدجاج المحلية

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اجريت هذه الدراسة لمعرفة تأثير التعرض المبكر للاجهاد الحراري على بروتين الصدمة الحرارية وتأثيره على هرمون الغدة الدرقية (T₃) في البلازما وكذلك على بعض القياسات المناعية والفسيولوجية والانتاجية وكذلك على وزن وجودة البيض 3 بعد 3 أشهر من بداية الانتاج في سلالة مطروح وأنشاص تم استخدام 720 ككتوت عمر يوم 360 من سلان مطروح و 360 من سلالة أنشاص (اشتملت التجربة على ثلاثة معاملات كل معاملة 120 ككتوت - الأولى هي معاملة المقارنة) كنترول (وعرضت المعاملة الثانية لاجهاد حراري مبكر على درجة حرارة 43 ± 1 - 42 درجة واحدة مئوية لمدة 4 ساعات وعرضت المعاملة الثالثة لنفس المعاملة الحرارية السابقة ولكن عند عمر 4 اسابيع بعد المعاملات الحرارية أعيدت الكتاكتيت إلى برنامج حراري عادي .عند عمر 18 أسابيع ثم تعريض 90 طائر من كل سلالة إلى درجة حرارة 43 ± 1 - 42 درجة مئوية .ثم أخذ عينات الدم قبل وبعد المعاملة الحرارية مباشرة و بعد الطرد المركزي حفظت للتحليل .وتم قياس درجة حرارة الجسم ومعدل التنفس وأخذت عينات الكبد لقياس بروتينات الاجهاد الحراري وأيضا تم أخذ 90 بيضة لمدة ثلاث شهور لكل سلالة على حده لأخذ قياسات جودة للبيض ووزن البيض .وكانت أهم النتائج هي :

- التعريض للاجهاد الحراري المبكر في كتاكتيت سلالة مطروح وأنشاص أدى الى زيادة التعبير الجيني لبروتينات الصدمة الحرارية في الكبد (70، 90 كيلو دالتون) بالمقارنة بالكنترول .
- أدى التعريض الحراري في عمر 3 أيام إلى إنخفاض مستوى تركيز هرمون T₃ في بلازما الدم بالمقارنة بمجموعة الكنترول والمجموعة الثالثة .
- أدى التعرض الحراري المبكر في عمر 3 أيام و 4 أسابيع من العمر إلى زيادة معنوية في الأجسام المضادة مقارنة بمجموعة الكنترول .
- وزن الجسم تأثر معنوي خاصة في الأسابيع الأولى من النمو في سلالة مطروح فقط في المجاميع التي تعرضت للاجهاد الحراري المبكر .
- وجد نقص ملحوظ في حرارة الجسم ومعدل التنفس في المجاميع التي كانت معرضة للاجهاد الحراري المبكر .
- وجد زيادة غير معنوية في بروتينات البلازما والاليومين والجلومولين في المجاميع التي تعرضت للاجهاد الحراري المبكر .
- الإجهاد الحراري المبكر يؤدي الى تحسن في وزن البيض وأيضا في بعض قياسات جودة البيض .كذلك الإجهاد الحراري المبكر أدى الى التحسن في التحمل الحراري عند البلوغ وبالتالي التحسن في أداء الطائر .