

EFFECT OF EARLY HEAT EXPOSURE ON HEAT SHOCK PROTEIN AND PHYSIOLOGICAL RESPONSES OF DUCKS DURING HEAT STRESS

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SUMMARY

One day-old Muscovy and Domyati ducks were used in this study. The objective of the study was to determine the effect of early heat acclimation on thermoregulatory responses of ducks during a subsequent exposure to high environmental temperature later in life. Muscovy and Domyati ducks were divided randomly into three groups (40 birds/group). Group 1 (Control Group), group 2 (Late Heat Stressed Group) and group 3 (Early Heat Acclimated Group). The SDS electrophoretic pattern of proteins in samples liver of Muscovy and Domyati ducks after subjected to heat stress (42-43 °C for 4h) at 12 weeks of age showed that heat stress caused activation in the expression of HSP families (HSP 90, and 70 kDa). However, early heat exposure caused hyperactivation in the expression of the HSP90 (93) than heated birds. While, synthesis rate of HSP70 was lower in early heat acclimated group than late heat stressed group.

Rectal temperature and respiration rate after exposure to heat stress were significantly higher in late heat stressed ducks than control ducks at 12 weeks of age. Rectal temperature was significantly lower in early heat acclimated ducks than late heat stressed ducks at 12 weeks of age. Respiration rate was significantly higher in early heat acclimated ducks than in the other groups. There were no significant differences in body temperature between Muscovy and Domyati ducks. Meanwhile, Muscovy had significantly higher respiration rate than Domyati ducks at all ages.

Heat stress at 12 weeks of age increased significantly plasma AST, ALT and corticosterone in non heat acclimated birds (late heat stressed group) while no significant difference was found between the early heat acclimated and the control groups.

The control group exhibited significant higher T_3 level followed by in late heat stressed and early heat acclimated groups at 12 weeks of age. These parameters were higher in Muscovy than in Domyati at 12 weeks of age.

Hematocrit % decreased in late heat stressed ducks than control birds. While hematocrit % significantly decreased in early heat acclimated ducks compared with late heat stressed birds at 12 weeks of age. This parameter was higher in Muscovy than in Domyati at 12 weeks of age. Values of lymphocytes were significantly decreased in late heat stressed ducks than control ducks. While, early heat acclimated ducks had significantly increased than late heat stressed ducks at 12 weeks of age. While values of heterophils were taken opposite trend under these

treatments the at same age. Values of lymphocytes and hrterophils were significantly increased in Domyati than in Muscovy ducks. While, H/L% was higher in Muscovy than in Domyati at 12 weeks of age.

Keywords: Ducks, Heat Stress, HSPs, T₃, AST, ALT and corticosterone concentration, hematocrit %, immune resposes

INTRODUCTION

Rapid heat shock response involves the expression of the heat shock genes and their encoded protein (heat shock proteins, HSPs) (Sanchez and Lindquist, 1990). The various families of HSPs (HSP 110, 90, 70, 60, 47 and small HSP ranging from 16 to 40 kDa) include proteins that are present prior to heat treatment but whose synthesis becomes detectable only following stress. Among the management procedures adopted to reduce the risk of death by heat stress in broilers is acclimatization.

Acclimatization in broilers occurs in the 3 days of age (Reece *et al.*, 1972). Arjona *et al.* (1988) reported that exposing broilers at just 5 days posthatching to heat significantly increased their ability to cope with heat stress 40 days later. A report by Wang and Edens (1993) showed that conditioning of chickens by daily exposure to high ambient temperature for 1h improved induction of HSP70 mRNA upon further exposure to a heat challenge. While, Yahav *et al.* (1997) showed that the conditioning of 5- days -old chicks at 36 °C improved thermotolerance at 42 days , meanwhile, the concentrations of HSP70 in heart and lung tissues were lower than in controls suggesting that HSP70 is not a part of the long-term mechanism evoked by the early age conditioning. The contradiction between these experiments could stem from the differences in the protocols of conditioning. However, Laszlo (1988) have suggested that there is a relationship between thermotolerance and HSP synthesis rate. Turkey leukocytes collected from animals experiencing heat stress have shown greater resistance to heat stress than cells stressed in vitro (Wang and Edens, 1994).

HSP90, whose family members are abundant at normal temperatures (up to 1% of the total soluble protein in the cytoplasm), interacts with steroid hormone receptors and apparently masks the DNA-binding region of the receptor until the receptor has bound to the appropriate steroid hormone, the receptor can then bind to DNA and activate expression of specific genes. A difference in HSP90/HSP70 ratio could be due to different functions assumed by two HSPs in the cell culture medium and plasma of the animal. HSP70 is involved in protein assembling, folding, and translocation, i.e. a house-keeping role, while HSP90 participates in formation of protein-complexes, such as steroid hormone receptors (Baulieu, 1987). HSP90 complexes with these receptors are closely associated with regulation of hormones, such as aldosterone and corticosterone. Their release is necessary to mount a response to a stressor. Therefore, synthesis of HSPs is so physiological significance in keeping systems in a homeostatic state (Baulieu, 1987).

Certain breed or strain of poultry appears to survive heat stress more successfully than others and birds that are acclimated gradually to elevated temperature are more resilient than those experiencing a sudden heat shock. Wild fowl, as do Bedouin fowl of the Israeli desert survive heat stress more successfully than normal commercial strains of chicken (Marder, 1973). A cross of Leghorn and Bedouin fowl produced

offspring with improved heat tolerance relative to the Leghorn parents (Arad *et al.*, 1975) indicating a genetic component in this tolerance.

It is tempting to speculate that part of the difference in heat tolerance of various breeds could be attributable to different alleles of heat-shock protein-encoding genes, resulting in differing ability to respond rapidly to a heat stress, differing final concentrations of the relevant heat-shock proteins in stressed birds or differing ability of various heat-shock proteins to interact with their normal ligands in the cell (Etches *et al.*, 1995).

Samy *et al.* (2001) found that serum AST and ALT levels decreased in winter than in summer for Muscovy ducks at 10 weeks of age. With respect to the effect of the acclimation to heat stress at early in life on thyroid response, it is noted that the mechanisms associated with the induction of thermotolerance by early-age temperature conditioning may involve the modulation of heat production through reduction in plasma triiodothyronine (T₃) concentration and haematocrit (Yahav and Hurwitz, 1996 and Yahav *et al.*, 1997).

Abdel-Mutaal (2003) reported that the birds exposed to early heat stress at 3 days of age had the lowest H/L value (0.663) followed by chickens conditioned at 5 days of age (0.669). While the control group showed the highest H/L ratio (0.681)

MATERIALS AND METHODS

One day age duckling were weighed, wing banded and divided randomly into three groups in both Muscovy and Domyati ducks (40 birds/group). Group 1 (Control Group) was maintained under normal ambient temperature during the whole period. Group 2 (Late Heat Stressed Group) was exposed to heat stress (42-43 °C) 4hr for 3 days at 12 weeks of age then ducks were maintained under normal ambient temperature.

Group 3 (Early Heat Acclimated Group) was exposed to 37-38 °C for 24h at 3 days of age and then exposed to heat stress (42-43 °C) 4hr for 3 days at 12 weeks of age then ducks were maintained under normal ambient temperature.

Heat Shock Proteins (HSPs) determined according to method of Polyacrylamide Gel Electrophoresis for proteins (SDS-PAGE) Sodium Dodecyl Sulphate (SDS). SDS-PAGE was performed according to the procedure described by Laemmli (1970).

Rectal temperature was measured through the cloacae at the depth of 3 cm by using a clinical thermometer after heat exposure. Respiration rate was measured by counting the body wall movement per minute. The blood samples were collected at 12 weeks of age and were centrifuged at 3000 rpm for 15 min and plasma obtained was stored at -20 °C. Aspartate transaminase (AST) and alanine transaminase (ALT) were determined according to the method of White (1970). Direct radioimmunoassay (R.I.A) technique was performed for plasma triiodothyronine (T₃) according to May (1978). The antiserum is highly specific for T₃ with low crossreactivity to other compounds that might be present in patient samples. Sensitivity: the procedure can detect as little as 7 ng/dL. Plasma corticosterone was estimated in duplicate using a specific radioimmunoassay (Sainio *et al.*, 1988).

Fresh blood samples were collected at the end of treatment (12 weeks of age) where hematocrit was immediately determined. The hematocrit values were expressed as a percent volume of packed cells in whole blood after centrifugation.

Hematocrit values were obtained by applying a column of 75 mm heparinized blood in microhematocrit tubule centrifuged for 20 minutes at 3000 rpm (Hunsaker, 1969). Heterophils \ Lymphocytes ratio (H \ L)

Undiluted heparinized blood were smeared on glass slides, stained with HEMA3 solution stain by dropping slides in each solution for 1 min. then washed by distilled water until pink color appeared in the smear and counted (two slides per bird) differentially using oil immersion lens of the light microcop, H \ L ratio was calculated thereafter. The analysis of the data was carried out by the Computer program SAS (1989), and the statistical model for the experiment was:

$$Y_{ijk} = \mu + T_i + B_j + T_i . B_j + E_{ijk}.$$

Where, μ = overall mean, T_i = Effect of treatment, B_j = Effect of breed, $T_i . B_j$ = Effect of interaction between treatment and breed, E_{ijk} = Random error, Y_{ijk} = Any observation in experiment. Duncan Multiple Range Test was used to test differences among means according to Duncan (1955).

RESULTS AND DISCUSSION

1 Heat Shock Proteins (HSPs).

The SDS electrophoretic pattern of proteins of liver of 12 weeks old Muscovy and Domyati revealed that synthesis rate of HSP90 and 70 Kda were higher in liver of Domyati than in Muscovy ducks in control groups. However, under heat stress the synthesis rate of HSP70 was lower in liver of Domyati than in Muscovy ducks while in early heat acclimated groups HSP90 was higher in Muscovy than in Domyati (Tables 1 and 2). These results indicates that Muscovy having higher efficiency in thermotolerance by early heat acclimated than Domyati ducks. The causes for the observed changes in breed may resulted from their changes in the cellular DNA and RNA (Durairay and Selvarajan, 1992).

The SDS electrophoretic pattern of proteins of liver of Muscovy ducks taken immediately after subjected to heat stress (42-43°C for 4h) is presented in Figure (1_a). It is clearly observed that heat treatment triggered hyperactivation in the expression of three major HSP families: HSP 90 and 70 kDa as particularly shown by the different density (Figure 2) and amount % in Table (1). Results revealed that subjected to heat stress led to increase in the isoforms induction of HSP90 kDa (97) in late heat stressed group compared to the control group. However, the protein bands at molecular weight isoforms of HSP70 kDa (73) and isoforms of 60 kDa (62) were appeared in control and late heat stressed groups but its synthesis rate were higher in late heat stressed group than in the control group (Muscovy ducks). These results were in agreement with Price and Calderwood (1991) and Duncan (1997) who showed clearly that heat induced activation of the HSP 70 gene. Also, these results were in agreement with Gabriel *et al.* (1996) who showed that heat stress induces an increase in HSP70 mRNA and HSP70 in the broiler liver.

Table (1) reveals that in Muscovy ducks EHE enhanced the induction of HSP90 so that the amount was higher than in the control and heat stressed groups. Their enhancement in HSP90 may explain the improve in adaptability to heat stress in EHA due to it regulation of steroid hormones (Baulieu, 1987). However, early heat acclimated had slight effect on HSP70 indicating that the enhancement in heat tolerance by early heat acclimated (Table 3) was due to the effect an enhancement in steroid hormone regulation through HSP90 and not to protein synthesis because no

effect was found in HSP70. HSP70 kDa was higher in early heated acclimated group than the control it was lower than that of the LHS group. These results were in agreement with Wang and Edens (1993) who showed that heat stress conditioning which allows animals to increase their resistance to heat exposure, stimulated expression of HSPs, suggesting a link between HSPs synthesis and development of thermotolerance. While, Yahav *et al.* (1997) reported that early age conditioning by exposure of birds to heat reduces the induction of HSPs. Therefore, the HSP response was not part of the long-term mechanism elicited by the thermal conditioning at an early age.

Table 1. Electrophoretic patterns of protein fraction in liver of Muscovy ducks

Lanes Rows	Marker		Lane 1		Lane 2		Lane 3	
	Mol. Weight	Amount %	Mol. Weight	Amount %	Mol. Weight	Amount %	Mol. Weight	Amount %
1			90	1.31	97	17.36	93	50.90
2			73	8.39	73	46.57	70	18.30
3	66	23.12	60	5.26	57	21.03	57	4.28
4	45	6.44	50	-	42	5.31	44	8.00
5	36	35.46	-	-	33	3.50	33	1.04
6	29	6.36	29	40.18	29	1.40	29	14.66
7			23	14.51	-	-	-	-
8	20	19.70	-	-	-	-	-	-
9	14	8.92	-	-	-	-	-	-
10			10	30.35	-	-	-	-
11			-	-	8	4.83	8	2.82

Marker = Standard protein

Lane 1 = Control

Lane 2 = Late Heat Stress

Lane 3 = Early Heat Acclimated

The SDS electrophoretic pattern of proteins of liver from 12 weeks old Domyati ducks taken immediately after subjected to heat stress (42 °C for 4h).

Heat stress lead to increase HSP90 and 70 kDa and its were higher in late heat stressed group than control group. However, early heat acclimated lead to stimulate the induction of HSP110 kDa and it was appeared only in this group and not in the other two groups. Figures (1_b and 3) and Table (2) reveals that in Domyati ducks (EHA) enhanced the induction of HSP110 and HSP90 kDa so that the amount of HSP90 kDa was higher than in the control and heat stressed groups. However, early heat exposure had slight effect on HSP70 indicating that the enhancement in heat tolerance by early heat exposure (Table 2) was due to the effect an enhancement in steroid hormone regulation through HSP90 and not to protein synthesis because no effect was found in HSP70. HSP70 kDa was higher in early heated acclimated group than the control it was lower than that of the LHS group.

Table 2. Electrophoretic patterns of protein fraction in liver of Domyati ducks

Lanes Rows	Marker		Lane 1		Lane 2		Lane 3	
	Mol. Weight	Amount %	Mol. Weight	Amount %	Mol. Weight	Amount %	Mol. Weight	Amount %
1			-	-	-	-	110	35.94
2			90	14.20	93	18.02	90	34.91
3			70	16.92	70	33.00	70	17.16
4	66	23.12	62	10.31	62	12.61	-	-
5	45	6.44	45	7.12	45	0.73	-	-
6			-	-	42	4.10	42	1.79
7	36	35.46	-	-	33	2.33	32	5.68
8	29	6.36	29	20.64	-	-	-	-
9			23	23.13	22	4.65	-	-
10	20	19.70	-	-	19	13.86	-	-
11	14	8.92	-	-	10	0.65	10	0.93
12			-	-	-	-	9	1.92
13			-	-	8.5	10.05	8.5	1.67
14			8	7.68	-	-	-	-

Marker = Standard protein

Lane 1 = Control

Lane 2 = Late Heat Stress

Lane 3 = Early Heat Acclimated

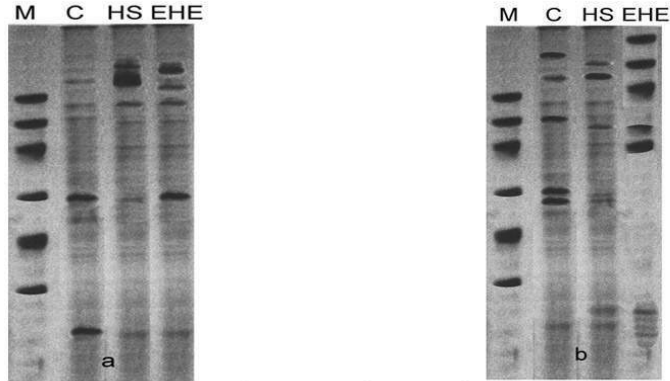
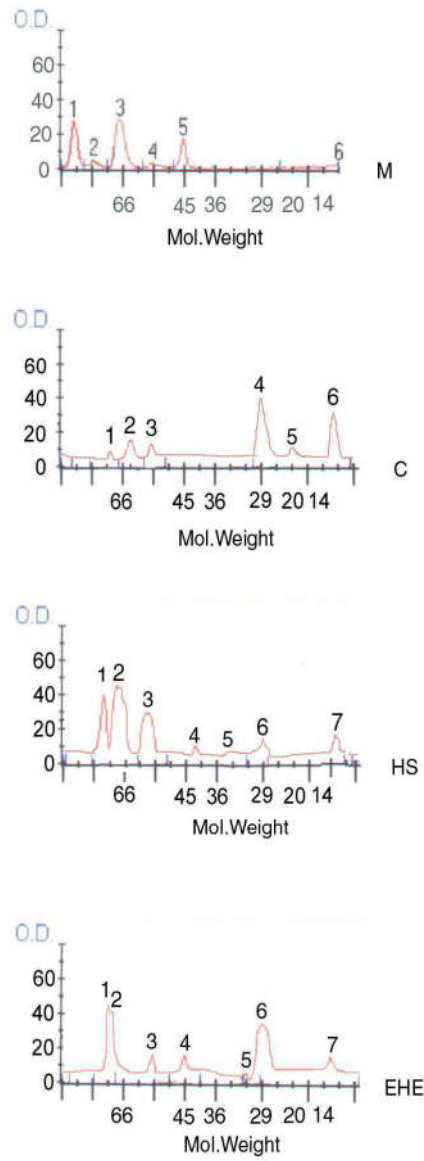
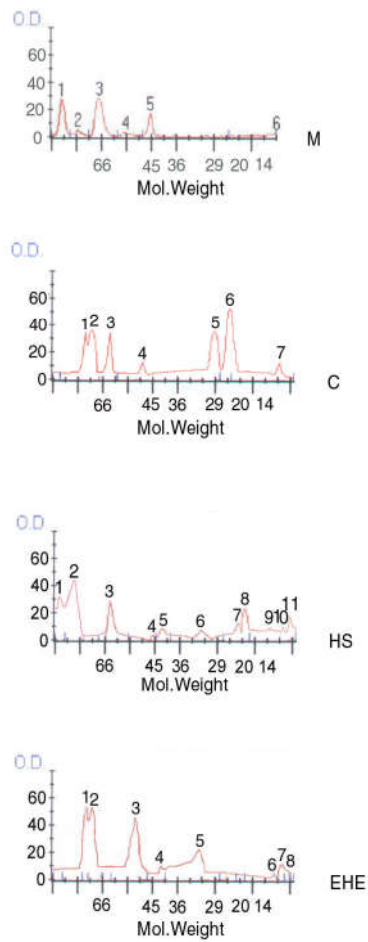


Figure (1) Electrophoretic pattern of protein fractions in liver of Muscovy (a) and Domyati (b) ducks exposed to heat stress at 12 weeks of age, Marker (M), Control (C), Heat Stress (HS) and Early Heat Exposure (EHE).



Figure(2)The densitograms of the protein pattern separation in Liver of Muscovy ducks, Marker(M), Control(C), Heat Stress(HS) and Early Heat Exposure(EHE).



Figure(3)The densitograms of the protein pattern separation in Liver of Domyati ducks, Marker(M), Control(C), Heat Stress(HS) and Early Heat Exposure(EHE).

2 Physiological Responses:

2-1 Thermal reaction:

Rectal temperature after exposure to heat stress was significantly higher in birds exposed to 42 °C at 12 weeks of age than in the control and EHA groups (Table, 3). The average respiration rate was significantly higher in early heat acclimated than in late heat stressed and control groups. Also, it was significantly higher in late heat

stressed group than in the control group (Table, 3) indicating that early heat exposure did not prevent the increase in respiration rate after exposure to heat stress. With respect to the effect of breed on thermal reaction, it is clearly observed that the main effect of breed was significant on respiration rate but not on rectal temperature (Table 3), meanwhile, Muscovy ducks had significantly higher respiration rate than Domyati ducks.

Table 3. Effect of breed and early heat exposure on thermal reaction of ducks at 12 weeks of age

Items	Breed	Treatment			SE	Overall	SE
		C	LHS	EHA			
Rectal temperature(°C)	Muscovy	41.54 ^{cd}	44.18 ^a	42.05 ^{bc}	0.21	42.59	0.13
	Domyati	41.11 ^d	43.93 ^a	42.36 ^b			
	Overall	41.32 ^c	44.05 ^a	42.20 ^b			
Respiration rate breath\min.	Muscovy	48.00 ^d	90.00 ^b	100.50 ^a	1.19	79.50 ^a	1.26
	Domyati	42.00 ^c	68.50 ^c	92.00 ^b			
	Overall	45.00 ^c	79.25 ^b	96.25 ^a			

a,b,c, in the same row are significantly different ($p < 0.05$)

C= Control.

LHS=Late Heat Stress.

EHE= Early Heat Acclimated.

2-2 Liver function:

Results in Table (4) show that, the plasma AST and ALT activity significantly increased in the birds exposed to late heat stress compared to control birds at 12 weeks of age. This result is agreed with Samy *et al.* (2001). Early heat exposure significantly decreased both AST and ALT activity compared with late heat-stressed birds at 12 weeks of age and subsequently the birds showed a degree of acclimation since, their AST and ALT activity were almost similar to the control birds reflecting better liver function. Plasma AST and ALT levels were significantly higher in Muscovy than in Domyati. The significant differences in plasma enzymatic activities (AST and ALT) may be attributed to genetic make-up of each breed (Kaneko, 1989).

2-3 Hormonal changes:

Data presented in Table (4) reveals that the control group having significant higher T_3 level followed by that in late heat-stressed group and early heat acclimated group. These results are in agreement with Osman (1996), Yahav and Hurwitz (1996) and Sabria and Younis (1999) who found that plasma T_3 concentration decreased significantly by exposing to high temperature. They also reported that at high temperature conversion of T_4 into T_3 is reduced to regulate the metabolic rate and then heat production.

Table 4. Effect of breed and early heat exposure on enzymes and hormones concentration of ducks at 12 weeks of age

Items	Breed	Treatment			SE	Overall	SE
		C	LHS	EHA			
AST IU/l	Muscovy	93.44 ^b	116.91 ^a	90.88 ^b	4.01	100.41 ^a	2.43
	Domyati	28.13 ^d	41.26 ^c	31.12 ^{cd}			
	Overall	60.79 ^b	79.08 ^a	60.99 ^b			
ALT IU/l	Muscovy	29.62 ^b	34.54 ^a	30.25 ^b	1.39	31.47 ^a	0.79
	Domyati	18.59 ^c	26.77 ^b	20.25 ^c			
	Overall	24.11 ^b	30.65 ^a	25.25 ^b			
T ₃ ng/100ml	Muscovy	95.08 ^a	65.99 ^{bc}	50.16 ^c	5.09	70.40 ^a	2.95
	Domyati	85.74 ^a	67.05 ^b	55.45 ^{bc}			
	Overall	90.40 ^a	66.51 ^b	52.80 ^c			
Cortico- sterone ng/100ml	Muscovy	13.22 ^c	22.37 ^a	15.24 ^{bc}	0.95	16.94 ^a	0.52
	Domyati	8.28 ^d	16.98 ^b	10.01 ^d			
	Overall	10.75 ^b	19.67 ^a	12.63 ^b			

a,b,c, in the same row are significantly different ($p < 0.05$)

C= Control.

LHS=Late Heat Stress.

EHE= Early Heat Acclimated.

Early heat exposure lowered T₃ level in plasma compared with plasma of late heat stressed birds and control birds. This result is in agreement with Yahav and Hurwitz, (1996); Yahav and Plavnik, (1999) and Uni *et al.* (2001) who reported that early age thermal conditioning resulted in a significant and sustained reduction in plasma T₃ levels. Furthermore, one of the mechanisms that induce thermotolerance involves the modulation of heat production through changes in circulating triiodothyronine (T₃). The ability to reduce plasma T₃ concentration, especially during a thermal challenge, suggests an improvement in thermotolerance (Yahav, 2000). With respect to the effect of breed on plasma T₃ concentration, it was found that plasma T₃ concentration was significantly higher in Muscovy than in Domyati Table (4). No significant breed differences in T₃ concentration was found at 12 weeks of age.

Table (4) indicates that heat stress increased significantly plasma corticosterone concentration, meanwhile early heat exposure ameliorated their effect that plasma corticosterone concentration was slightly in early heat acclimated than in control groups.

The reduction of corticosterone concentration in early heat acclimated birds may be due to that the early heat exposure enhances adaptability of birds to heat stress and the birds previously experiencing a high temperature showed a lower standing-lying frequency during subsequent exposure to high temperature, indicating that they are less "nervous" during subsequent exposure to high temperature (Zhou *et al.*, 1997).

Regarding the effect of breed on plasma corticosterone concentration, results in Table (4) indicate that plasma corticosterone concentration was significantly higher in Muscovy than in Domyati.

2-4 Blood parameters:

Results in Table (5) show that heat stress significantly reduced hematocrit values, as compared to non-heat stressed birds during heat stress at 12 weeks of age. While, early heat acclimated birds having significantly lower hematocrit % compared to late heat stressed birds. These results are in agreement with Yahav and Hurwitz (1996)) who concluded that, blood volume of broilers increased and hematocrit decreased when exposed to cyclic temperature. Sturkie (1986) revealed that the corpuscular volume (Ht) is influenced by age and other factors, which affect cell number. With respect to the effect of breed on hematological parameters, it was found from the results in Table (5) that hematocrit % were significantly higher in Muscovy ducks than in Domyati ducks. Lymphocytes % were significantly decreased in late heat stressed birds than control birds. Early heat acclimated birds had significantly higher lymphocytes % than late heat stressed birds. The mean values of heterophils counts and H / L % significantly increased with exposure to heat stress. Maxwell and Robertson (1998) sustained monitoring of H/L% to heat stress. (Atta, 1996) showed that, heat stress caused suppress in the activity of T and B-lymphocytes and macrophages.

Table 5. Effect of breed and early heat exposure on blood parameters of ducks at 12 weeks of age

Items	Breed	Treatment			SE	Overall	SE
		C	LHS	EHA			
Hematocrit %	Muscovy	35.32 ^a	32.56 ^{ab}	30.84 ^{bc}	1.11	32.90 ^a	0.62
	Domyati	33.32 ^{ab}	30.38 ^{bc}	27.56 ^c			
	Overall	34.32 ^a	31.47 ^b	29.19 ^c			
Lymphocyte %	Muscovy	61.22 ^b	56.62 ^c	60.32 ^b	0.82	59.38 ^b	0.46
	Domyati	64.42 ^a	60.45 ^b	62.56 ^{ab}			
	Overall	62.82 ^a	58.53 ^b	61.43 ^a			
Heterophils %	Muscovy	28.26 ^{bc}	32.14 ^a	25.81 ^d	5.09	28.73	28.06
	Domyati	27.69 ^{cd}	30.38 ^{ab}	26.11 ^{cd}			
	Overall	27.97 ^b	31.26 ^a	25.96 ^c			
H/L %	Muscovy	0.45 ^c	0.56 ^a	0.42 ^d	0.01	0.48 ^a	0.01
	Domyati	0.43 ^d	0.50 ^b	0.41 ^d			
	Overall	0.44 ^b	0.54 ^a	0.42 ^c			

a,b,c, in the same row are significantly different (p< 0.05)

C= Control.

LHS=Late Heat Stress.

EHE= Early Heat Acclimated.

It has been suggested that the ratio could be used to detect the presence of physiological stress for most stressors. There was a significant improvement in H / L ratio sticking with early heat exposure. This confirmed the suggestion that early heat exposure enhanced the ability to heat resistance. With respect to the effect of breed on immune response, Table (5) shows that there were significant differences between breeds for lymphocyte and H / L ratio. These parameters were significantly higher in Domyati than in Muscovy ducks. there was no significant differences

between breeds for heterophils This may indicate that the Domyati breed had higher immunity responsiveness compared with Muscovy ducks.

It can be concluded that early heat exposure in life reduced some of negative effects of heat stress on immune system of ducks when exposed to high environmental temperature at 12 weeks of age by enhancing the ability to express heat shock proteins especially HSP90.

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تأثير التعريض المبكر للحرارة علي بروتينات الصدمة الحرارية والاستجابة الفسيولوجية للبط تحت ظروف الإجهاد الحرارى

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تم تقسيم كل من البط المسكوفى والدمياطى عمر يوم آلى ثلاثة مجا ميع (٤٠ طائر / مجموعة) مجموعة الكنترول ومجموعة الإجهاد الحرارى ومجموعة الأقلمة أجريت هذه التجربة لتحسين الأداء الفسيولوجى والتنظيم الحرارى للبط المربى تحت ظروف الإجهاد الحرارى فى أعمار متأخرة وذلك عن طريق أقلمة الطيور على درجات الحرارة العالية فى أعمار مبكرة .

وكان من أهم النتائج مايلى:

- وجد بالتقريد الكهربى لعينات من الكبد للبط المسكوفى والدمياطى أن التعريض للإجهاد الحرارى سبب زيادة فى التعبير الجينى للبروتينات ٧٠ و ٩٠ كيلودالتون بالمقارنة بالكنترول عند ١٢ أسبوع من العمر بينما الأقلمة المبكرة سببت فى زيادة فى تخليق بروتينات ٩٣ كيلودالتون اعلى بالمقارنة بمجموعة الإجهاد الحرارى عند نفس العمر بينما بروتينات ٧٠ كيلودالتون، ظهرت فى مجموعة الاقلمة ولكن معدل تخليقهم كان اقل مقارنة بمجموعة الإجهاد الحرارى

تحت ظروف البيئة الطبيعية كانت بروتينات ٧٠ و ٩٠ كيلودالتون اعلى فى الدمياطى عن المسكوفى - ارتفعت درجة حرارة الجسم وأيضاً معدل التنفس فى الطيور المعرضة للإجهاد الحرارى بالمقارنة بالكنترول عند ١٢ أسبوع من العمر بينما الأقلمة المبكرة ادت الى انخفاض فى درجة حرارة الجسم عند ١٢ أسبوع من العمر بالمقارنة بالمجموعة المعرضة للإجهاد الحرارى فى حين ان الأقلمة المبكرة ادت الى زيادة فى معدل التنفس عند ١٢ أسبوع من العمر بالمقارنة بالمجاميع الأخرى.

- لا توجد اختلافات معنوية فى درجة حرارة الجسم بين المسكوفى و الدمياطى فى الأعمار المختلفة ولكن كان معدل التنفس عالى فى البط المسكوفى عن البط الدمياطى.

وجد أن نشاط الانزيمات الدالة على وظائف الكبد (ALT, AST) وتركيز هرمون الكورتيكوستيرون تزيد معنويا فى مجموعة الإجهاد الحرارى بالمقارنة بالكنترول عند ١٢ أسبوع من العمر بينما الأقلمة المبكرة سببت انخفاض معنويا فى نشاط هذه الانزيمات بالمقارنة بمجموعة الإجهاد الحرارى عند نفس العمر .

وجد أن نشاط هذه الأنزيمات تزيد معنويا فى البط المسكوفى عن البط الدمياطى فى الأعمار المختلفة.

- أظهرت الطيور بمجموعة الكنترول زيادة معنوية فى تركيز هرمون T₃ بالمقارنة بمجموعة الإجهاد الحرارى عند ١٢ أسبوع من العمر بينما ادت الأقلمة المبكرة الى انخفاض فى تركيز هرمون T₃ للبلازما بالمقارنة بالمجاميع الأخرى.

وجد أن تركيز هرمون T_3 أعلى في البط المسكوفى عن البط الدمياطى .
ادى الإجهاد الحرارى الى انخفاض نسبة المكونات الخلوية بالمقارنة بمجموعة الكنترول عند عمر ١٢ أسبوع
بينما الأقلمة المبكرة سببت انخفاض فى نسبة المكونات الخلوية بالمقارنة بمجموعة الإجهاد الحرارى عند نفس
العمر .

ادى الإجهاد الحرارى الى انخفاض عدد الخلايا الليمفاوية بالمقارنة بمجموعة الكنترول عند عمر ١٢ أسبوع
بينما سببت الأقلمة المبكرة زيادة فى عدد هذه الخلايا بالمقارنة بمجموعة الإجهاد الحرارى عند نفس العمر .
أيضا ادى الإجهاد الحرارى الى زيادة عدد خلايا الهيتروفيل زيادة معنوية بالمقارنة بمجموعة الكنترول عند
عمر ١٢ أسبوع و بينما سببت الأقلمة المبكرة زيادة فى عدد هذه الخلايا . وجد أن عدد الخلايا الليمفاوية و
خلايا الهيتروفيل اعلى فى البط الدمياطى عن البط المسكوفى .