EFFECTS OF DIFFERENT ENVIRONMENTAL TEMPERATURES ON THE PHYSIOLOGICAL PERFORMANCE OF JAPANESE QUAIL

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ABSTRACT:

A total number of five hundred and forty sexed Japanese quail birds were used to study the effect of different environmental temperature on the physiological performance. Birds were assigned randomly into four equal groups, each group containing 135 birds in nine replicates of 15 birds each (5 males +10 females), in laying batteries. The four groups were raised at different temperatures (20, 25, 30 and 35^{o}C). The birds were fed diet containing 19.5% CP and 2852 Kcal ME/Kg diet. Blood samples were collected from 5 birds (2 males and 3 females) at slaughter during the periods of sexual maturity, peak of egg production and at the end of egg production phase. Hemoglobin, packed cell volume, Red blood cells (RBCs), White blood cells **(**WBCs) and lymphocyte were significantly decreased (P<0.05) under heat stress at beginning, peak and end of egg production. Heat stress significantly (P<0.05) increased heterophil cells and H/L at the beginning, peak and end of egg production. Eosinophils, basophil and monocyte were not affected by heat stress. Heat stress resulted in significantly (P< 0.05) decrease total plasma protein, albumin and globulin at the beginning, peak and end of egg production. Serum phosphorus and calcium levels were decreased due to the effect of heat stress. Heat stress resulted in significantly (P< 0.05) increase plasma triiodothyronine (T_3) level, but it decreased the plasma tetra-iodothyronine (T_4) level at the beginning, peak, and end of egg production. Significant effect of the temperature stress was noticed on thyroid gland and adrenal gland of the exposed groups of Japanese quail. The effect was remarkable mainly on size of the organs than the morphological characters.

Key Words: Japanese quail, Heat stress, Blood parameters, Thyroid gland, Adrenal gland.

INTRODUCTION

It is generally recognized that the growth rate of domestic birds exhibit many variation by many factors. The environmental temperature is an influencing factor in this variation. In Japanese quail heat stress, generally, reduced total red blood cells, packed cell volume, hemoglobin and leukocytes count (Nirmalan and Robinson, 1971; Vo *et al.* 1978 and Galal, 1999). Normal percentage of different types of leukocytes in Japanese quail were recorded by Sturkie (1988) for lymphocytes, heterophils, eosinophils, basophils and monocytes which represented 72.6%, 21.3%, 3.4%, 0.3% and 2.4 respectively, of adult mixed-sex Japanese quail. Galal (1999) mentioned that in Japanese quail erythrocytes and leukocytes numbers were significantly lower after heat stress at all ages and in all experimental groups. She also found that lymphocyte and basophil number were significantly lowered after exposure to heat stress at all ages. The heterophil/lymphocyte ratio was significantly higher after heat stress at all ages in Japanese quail (Kassab *et al*. 1992; Osman. 1996 and Galal, 1999). Total plasma proteins were consistently decreased as environmental temperature

increased. (Yahav *et al.*, 1997; Zhou *et al.* 1998 and Galal 1999) Albumin and globulin decreased during heat stress (Vo. *et al.* 1978, Ibraheem, 1987 and Zhou *et al* 1998). Plasma calcium and inorganic phosphorus concentration of Japanese quail was significantly lower under heat stress. (Boulhsan, 1993 and Galal, 1999 Bowen and Washburn, 1985; and Galal 1999) found that plasma corticosterone in Japanese quail was significantly higher after heat stress Bobek *et. al.*, (1980); Bowen and Washburn, (1985) and El-Nabarawy (1997) reported that in Japanese quail heat stress reduced (T_4) levels while (T_3) levels was increased after heat sress. The main olject of this work was to study the physgicalogical performance of Japanese quail under different environmental temperatures.

MATERIALS AND METHODS

The present experiment was carried out during the period from 18th January to August 2002 in the Poultry Research Station, Poultry Department, Faculty of Agriculture, Fayoum, Cairo University. The experiment was designed to study some physiological, productive and reproductive parameters of Japanese quail under heat stress.

Birds and management:

Five hundred and forty sexed Japanese quail birds (seven weeks of age) were assigned randomly into four equal groups, each group containing 135 birds in nine replicates of 15 birds each (5 males +10 females), in laying batteries. The four groups were housed in four different environment temperatures (20, 25, 30 and 35^{o}C). The temperatures were kept constant for each room. The relative humidity was held between 50 and 60 % for all room.

The birds from one day until 42 days of age (growing period) were given diet containing 23.7% CP and 2876 Kcal ME/Kg diet, after which (laying period) the birds were fed diet containing 19.5% CP and 2852 Kcal ME/Kg diet. Diets were formulated according to NRC recommendation (1994). The composition and chemical analysis of the experimental diets are shown in Table (1). Feed and water were provided *ad libitum.*

- Blood hematological parameters:

Blood samples were collected from 5 birds (2 male and 3 female) at slaughter during the periods of sexual maturity, peak of egg production and at the end of egg production. About 3cm^3 of blood from each bird was collected in a heparinized test tube to determine the parameters of blood.

-Blood hemoglobin (Hb %), Packed cell volume (PCV), Red blood cells (RBCs) count (x10^6/mm^3), and White blood cells (WBCs) count (x10^3/mm^3): were counted by using the method of Bauer (1970).

- Differential count of white blood cells: A thin blood film was prepared by using a small drop of blood and two slides (plain and spreader). The blood film was completely dry before staining by using Wright,s stain, the film was stained for three minutes, after that washed in distilled water and dried. The blood film was examined by using the light microscope. Different types of WBCs were counted in several fields (total 100 cell). Numbers of different types were expressed as percentage.

- Heterocyte/Lymphocyte (H/L) ratio: H/L ratio = Number of H/ Number of L

- Chemical parameters:

- Total plasma protein (g/dl) and plasma albumin (g/dl):) were determined as described by Connon, *et al.* (1974) and by Webster (1974) respectively. **- plasma globulin (g/dl)** =Total protein of plasma (g/dl)– plasma Albumin (g/dl)

- **Albumin: globulin (A/G) ratio:** = Albumin (g/dl) / Globulin (g/dl)
- **Inorganic phosphorous (mg/dl):**
- **Calcium ions (mg/dl):** was measured as described by Elveback (1970).
- **Triiodothyronine (T_3) (ng/dl)**, **Thyroxine (T_4) (ng/dl):** were determined by radioimmunoassay procedure (Tietz, 1995).
- **Corticosterone (ng/dl)** was determined by radioimmunoassay procedure (Sainio, 1988).
- **Histological parameters:** Thyroid and Adrenal glands were obtained at the beginning of egg production (6 weeks of age), peak of egg production (13 weeks of age) and at the end of egg production (23 weeks of age) after slaughter and prepared slides according to methods of Culling (1974).
- **Statistical analysis:** Analysis of variance was computed using the general linear model (GLM) procedure of statistical analysis system (SPSS, 1997). Variable means for treatments indicating significant differences in the ANOVA were compared and the differences were indicated using Duncan's multiple range tests (Duncan, 1955).

Table (1): Composition and chemical analysis of experimental diets (growing and laying).

	Growing diet	**Laying diet**
Ingredient	**%**	**%**
Yellow corn, ground	58.00	64.50
Soyabean meal (44%CP)	31.31	20.50
Concentrate (50% CP)	10.00	10.00
Limestone	------	2.00
Dicalcium phosphate	------	2.31
Salt	0.30	0.30
*Vit. and Min. premix	0.30	0.30
DL- methionine	0.09	0.09
Total	100	100.00
Chemical composition %		
Crude protein	23.71	19.50
Ether extract	2.89	3.01
Crude fiber	3.61	2.99
Calcium	0.912	2.33
Total phosphorus	0.691	0.873
Available phosphorus	0.447	0.665
Lysine	1.345	1.042
Methionine	0.586	0.516
ME, Kcal/Kg diet	2876.46	2852.90

- *Each 3 Kg of the Vit and Min. premix contains: Vit A 12000000 IU, Vit.D3 2500000 IU, Vit. E 10g, Vit. K 2.5g, Vit. B1 1.5g, Vit.B2 5g, Vit B6 1.5g, Vit B12 10mg, Choline chloride 1050gm, Biotin 50mg, Folic acid 1gm, Nicotinic acid 30g, Ca pantothenate 10g, Zn. 55g, Cu 10g, Fe 35g, Co 250mg, Se 150mg, I 1g, Mn 60g and antioxidant 10g*

RESULTS AND DISCUSSIONS

1) Blood parameters:

1-1) Hemoglobin (g/100m), Packed cell volume (P.V.C), total red blood corpuscle count (RBCs). The elevation in acute temperature caused significant progressive decreased in the amount of hemoglobin or packed cell volume in blood at all periods of egg production. (Table 2) Similar results was obtained in Japanese quail by Osman (1996), El-Nabarawy (1997) and Galal (1999). Heat

stress leads to decrease of RBC numbers, the mean corpuscular hemoglobin concentration and life span of the red blood cells. This led to decreasing the hemoglobin concentration in blood due to the positive relation between the RBC number and hemoglobin concentration in blood. This decrease in total number of RBCs many be due to the inhibition of heat stress on the life span of the present RBCs as well as inhibition the production of new RBC from the bone narrow.

1-2) Total leukocyte count (W.B.Cs): There is a significant relation between heat stress and the decrease in number of W.B.Cs in exposed quail. The same results were recorded previously in Japanese quail by McFarlane and Curties (1989), Maxwell *et al.,* (1990) and Galal (1999) after exposure to 38 ^{o}C - 42^{o}C heat stress. (Table 2).

1-3) Effect of heat stress on differential leukocyte count :

1-3-1) Heterophil: The highest significant (P< 0.05) value of heterophil cells was recorded in birds exposed to 35^{o}C while the lowest value was observed in birds exposed to 25^{o}C, in comparison with that exposed to 20 ^{0}C at all periods of egg production. Similar results was obtained in Japanese quail by Strukie (1988), Mcfarjane *et al.,* (1989), Al-Murrani *et al.,* (1997) and Galal, (1999). Increase in heterophil cells as a results of heat stress may be due to more release of adrenocorticotrophic (ACTH) hormone, which led to increase heterophil cells. (Table 3).

1-3-2) Lymphocyte: Significant (P< 0.05) decrease in lymphocyte was recorded in birds exposed to 35^{o}C followed by that exposed to 30^{o}C than that of 25^{o}C, in comparison with that exposed to 20^{0}C at beginning, peak and end of egg production. The change in lymphocyte and heterophil percentage due to the release of ACTH through the hypothalamus pituitary axis, after exposing the birds to stress which in turn decrease the lymphocyte percentage, whereas heterophil percentage increase. (Table 3).

1-3-3- Heterophil/Lymphocyte Ratio (H/L): Significant (P< 0.05) increased in H/L was recorded in birds exposed to 35^{o}C followed by that exposed to 30^{o}C than that 25^{o}C, in comparison with that exposed to 20^{0}C at beginning, peak and end of egg production. Similar results was obtained in Japanese quail by McFarlane, *et al., (*1989) and Galal (1999). The decrease in lymphocyte and increase in heterophil percentage was described as a better measure of the chickens perception of stress in its environment than the number of hetyerophil or lymphocyte only (Gross and Siegel, 1983). Heterophil/lymphocyte ratio was found to vary with change of temperature and other stressors (Siegel, 1980; Gross and Siegel, 1983 and Kassab *et al*, 1992). (Table 3).

2) Effects of heat stress on chemical parameters of blood

2-1) Total proteins, Albumin, Globulin Significant (P< 0.05) decrease in total proteins, albumin and globulin were recorded in birds exposed to 35^{o}C followed by that exposed to 30^{o}C, then that exposed to 25^{o}C, in comparison with that exposed to 20^{0}C at beginning, peak and end of egg production season for both male and female birds. Similar results was obtained heat stress by Yahav *et al*., (1997), Zhou *et al.,* (1998) and Galal (1999). The decrease in globulin indicate the decrease in immune response by the increase of temperature. These results was agreed with that previously described by Ibraheem (1987) and Galal (1999). (Table 4).

Table 2

Table 3

Table 4

Table (5) Effect of heat stress on serum Phosphorus and Calcium levels of Japanese quail at End of egg production (23 week of age).

Parameter / Treatment	Phosphorus			Calcium			Ca / P ratio		
	M	F	overall mean	M	F	overall mean	M	F	overall mean
Room Temp	6.650[a] ± 0.05	6.533[a] ± 0.03	6.492[a] ± 0.04	8.200[a] ± 0.11	8.233[a] ± 0.12	8.217[a] ± 0.05	1.233[a] ±0.002	1.26[a] ± 0.01	1.3[a] ±0.01
25ºC	6.150[b] ± 0.07	6.233[b] ± 0.03	6.192[b] ± 0.03	6.150[b] ± 0.05	6.167[b] ± 0.03	6.158[b] ± 0.1	1.00[a] ±0.003	0.99[a] ± 0.02	0.99[a] ± 0.02
30ºC	5.450[c] ± 0.06	5.367[c] ± 0.07	5.408[c] ± 0.04	5.850[c] ± 0.06	5.800[c] ± 0.58	5.83[c] ±0.054	1.08[a] ±0.004	1.08[a] ±0.03	1.08[a] ±0.03
35ºC	5.350[c] ±0.05	5.400[c] ±0.06	5.375[c] ±0.03	5.800[c] ±0.12	5.833[c] ±0.03	5.817[c] ±0.01	1.08[a] ±0.003	1.08[a] ±0.04	1.08[a] ±0.04

(a, b, c, d) means within each column with different superscripts are significantly different at ($p < 0.05$)

2-2) Phosphorus level, Calcium level: Significant ($P<0.05$) decrease in phosphorus level was recorded in birds exposed to 35^{o}C followed by that exposed to 30^{o}C then that of 25^{o}C, in comparison with that exposed to 20 ^{0}C at beginning, peak and end of egg production. Similar results was obtained by McCormick and Garlich, (1982), El-Nabarawy (1997) and Galal (1999). In the same manner, Hocking *et al* (1994) suggested that heat stressed in single comb white leghorn revealed lost of calcium as a consequence of panting, became rapidly alkaloid and this caused the increased ionization of blood protein. As the hydrogen ions left the protein, their absence created a negative charge that attracted and bounded the free calcium in the blood. The free calcium also bound to the lactate molecule in the blood because it had negative charge and lactate accumulated in the blood. (Table 5).

2-3) Calcium/Phosphorus ratio: There is an indirect relation bet ween the degree of temperature used in heat stress and the decrease in calcium/ phosphorus ratio in exposed quail. (Table 5).

3) Effect of heat stress on plasma hormones.

3-1) Triiodothyronine (T_3): Significant ($P<0.05$) increased in Tri-iodothyronine level was recorded in birds exposed to 35^{o}C or 30^{o}C, in comparison with that exposed to 20^{0}C at beginning, peak and end of egg production. This result was agreed with the previous studies of Bowen and Washburn (1985) and May *et al.,* (1986) who showed that heat stress caused a significant reduction in T3 concentration in chicken. The same observations were reported by El-Nabarawy, (1997) in quail where she mentioned that Tri-iodthyronine (T_3) increased under heat stress because tetera-iodothyronine (T_4) was conversion to (T_3). (Table 6).

3-2) Tetraiododthyronine level (T_4): Significant ($P<0.05$) decrease in (T_4) was recorded in birds exposed to 35^{o}C followed by that exposed to 30^{o}C then that exposed to 25^{o}C in comparison with that exposed to 20 ^{0}C for both males and females at beginning, peak and end of egg production. Similar results was obtained in Japanese quail at 38 ^{o}C -42^{o}C heat stress by El-Nabarawy (1997) who showed that significant decrease in thyroxin (T_4) levels while tri iodothyronine (T_3) levels was increased as a result of heat stress. The heat stress stimulates the hypothalamus to lower level of thyroid releasing hormone (TRH) secretion and affect on thyroid gland to decrease thyroid secretion. (Sturkei, 1988).

Table 6

3-3) T3/T4 ratio Data in (Tables 5) cleared that serum T_3/T_4 was higher at 35°C than that at 30°C or at 25°C and 20°C. These higher value was significant (P<0.05) along the egg production period (beginning, peak and end). Similar results was obtained in Japanese quail at by Bowen and Washburn (1985) and El-Nabarawy (1997). They mentioned that reduced response of the thyroid activity in Japanese quail was associated with heat stress. The inverse relationship between thyroid function and heat stress survival time in chicken may be due to a number of factors including body fat, the nervous system, thermo-genesis and metabolic rate as mentioned by Bowen, *et al., (*1984).

3-4) Corticosterone level: Significant (P<0.05) increase in corticosterone was recorded in birds exposed to 35°C followed by that exposed to 30°C then that exposed to 25°C in comparison with that exposed to 20 °C at beginning, peak and end of egg production season.

The level of corticosterone inversely related with thyroxin level, where the decreased T4 level stimulate indirectly the increase secretion of cortisone. This also may be due to increase in heterophile and H/L ratio. The level of corticosterone increased with the increased T3. (Table 6).

Table (7) Effect of heat stress on the size of thyroid gland and adrenal gland (mm).

Time of sample collection	Tested groups	Thyroid gland		Adrenal gland	
		Mean of length	*Mean of breadth*	*Mean of length*	*Mean of breadth*
Beginning of production	***1–G*** **(20 ºC temperature)**	2.3 ±0.22^{a}	1.1 ± 0.33^{a}	2.8 ±0.3^{a}	1.0 ± 0.2^{a}
	2–G **(25 ºC temperature)**	2.1 ± 0.20^{b}	1.1 ± 0.22^{a}	2.6 ± 0.3^{b}	1.0 ± 0.3^{a}
	3–G **(30 ºC temperature)**	2.0 ± 0.33^{b}	0.9 ± 0.35^{b}	2.4 ± 0.33^{c}	0.9 ± 0.4^{b}
	4–G **(35 ºC temperature)**	1.9± 0.33^{c}	0. 9± 0.33^{b}	2.2± 0.33^{a}	0. 9± 0.4^{b}
Peak of production	***1–G*** **(20 ºC temperature)**	2.8±0.33^{a}	1.4 ±0.4^{a}	4.0 ±0.3^{a}	1.2 ± 0.2^{a}
	2–G **(25 ºC temperature)**	2.5 ± 0.30^{b}	1.3 ±0.32^{a}	3.8 ± 0.4^{b}	1.1 ± 0.4^{b}
	3–G **(30 ºC temperature)**	2.4 ± 0.4^{c}	1.0 ±0.33^{b}	3.2 ± 0.33^{c}	1.0 ± 0.3^{c}
	4–G **(35 ºC temperature)**	2.4 ±0.22^{c}	1.0 ±0.25^{b}	3.2± 0.33^{c}	1.0± 0.4^{c}
End of production	***1–G*** **(20 ºC temperature)**	2.6±0.30^{a}	1.2 ±0.33^{a}	3.8 ±0.4^{a}	1.2 ± 0.3^{a}
	G - 2 **(25 ºC temperature)**	2.2 ± 0.5^{b}	1.1 ±0.22^{a}	3.4 ± 0.5^{b}	1.1 ± 0.3^{a}
	G - 3 **(30 ºC temperature)**	2.1 ± 0.30^{c}	1.0 ±0.35^{a}	3.1 ± 0.35^{c}	1.0 ± 0.5^{b}
	G - 4 **(35 ºC temperature)**	2.0 ±0.5^{d}	1.0 ±0.22^{a}	3.1± 0.33^{c}	1.0± 0.33^{b}

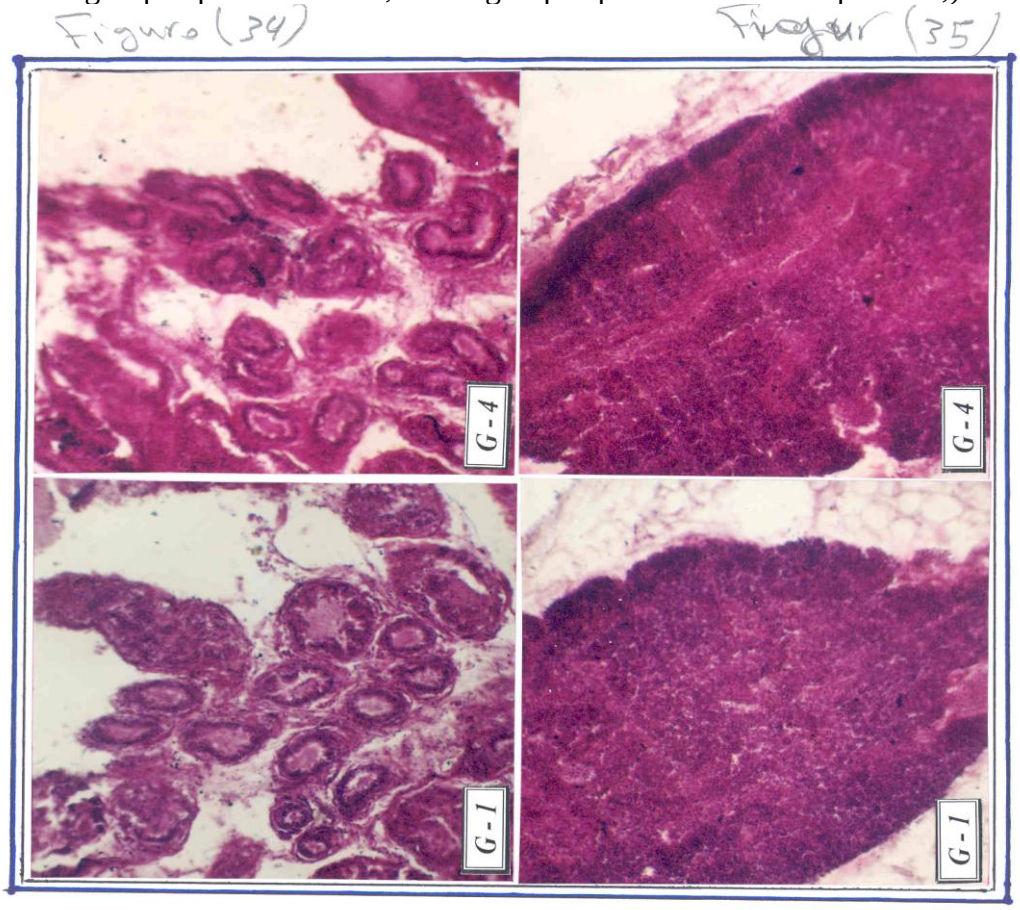
(a, b, c, d) means within each column with different superscripts are significantly different at ($p < 0.05$)

4) Effects of heat stress on histological structure of thyroid and adrenal glands :-

The results of histological sections and that of measurements revealed marked significant effect of the temperature stress on Thyroid gland, Adrenal gland and gonads of the exposed groups of Japanese quail. The effect was remarkable on size of the organs mainly (Table, 7) than in the morphological characters under light microscpe as in the illustrated figuers. The significant effect (P> 0.05) of temperature was recorded between the control group and that exposed to highr temperature degre (35ºC). This came in agreement with Snedecor (1968) who mentioned that the size of thyroid gland in chickens was increased in could conditions than in worm one. While Bhattacharyya *et al*. (1967) mentioned that adrenal glands increased in size in cold climat. They also, mentioned that the adrenal of birds are encapsulated with a thin connective tissue with no defined cortex and medulla, nor obvious zonation of cortical tissue into three distinct layers as in mammals.

Figure (1) Histological section in thyroid gland and adrenal gland of heat treated groups at peak of egg production. (X = 200)

(G-1 = group exposed to 35 ^{o}C, G-4 = group exposed to room temperature,)



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تأثير درجات الحرارة المختلفة على الآداء الفسيولوجى لطيور السمان

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** معهد بحوث الإنتاج الحيوانى – الدقى – مركز البحوث الزراعية – الجيزة - مصر.

لدراسة تأثير درجات الحرارة المختلفة على الآداء الفسيولوجى لطيور السمان تم استخدام ٥٤٠ طائر سمان قسمت هذه الطيور الى ٤ مجاميع متساوية العدد (١٣٥ طائر) كل مجموعه تتكون من ٩ مكررات (٥ ذكور + ١٠ إناث) ربيت فى بطاريات مختلفة درجات الحرارة (٢٠ و ٢٥ و ٣٠ و ٣٥ °م).

- تم تغذية الطيور على عليقة تحتوى على ١٩.٥% بروتين ٢٨٥٢ كيلو كالورى.
- تم الحصول على عينات دم من ٥ طيور (ذكرين + ٣ إناث) وذلك بالذبح عند أعمار النضج الجنسى، قمة إنتاج البيض ونهاية فترة الإنتاج.
- تم استئصال غدة الدرقية وكذلك غدة الأدرينال بعد الذبح وتم تجهيز شرائح منها.
- تسبب التعرض لدرجات الحرارة المرتفعة الى نقص معنوى ($P<0.05$) للهيموجلوبين وعدد كرات الدم الحمراء وعدد كرات الدم البيضاء.
- خلايا الهتروفيل وكذلك نسبة H/L كانت أعلى فى الطيور المرباه تحت ظروف حرارة عالية وقد تسبب الإجهاد الحرارى الى خفض معنوى فى مستويات البروتين الكلى والألبومين والجلوبيولين وكذلك لمستويات فوسفور وكالسيوم السيرم.

أما مستوى هرمون ترای أيودثرينين فقد ارتفع تحت تأثير الحرارة المرتفعة بعكس هرمون تترأيودوثريونين الذى انخفض مستواه فى الدم.

- كما اثر الإجهاد الحرارى تأثيراً معنوياً على الغدة الدرقية وغدة الأدريتال وخاصة على الحجم وليس على الناحية المورفولوجية.