



Amelioration of Heat-Stress Conditions of Egyptian Summer Season on Friesian Calves Using Air Condition

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ABSTRACT

Male Friesian calves were used to evaluate cool air condition (AC) in alleviating heat stress (HS) determined by Heat Shock Protein genes expression (HSP), hormonal, biochemical and physiological parameters. The animals were exposed to summer heat stress (HS) under shade for two weeks (control). The maximum temperature humidity index (THI) during summer HS was from 81 to 88. Afterward the animals were exposed to AC, inside a climatic chamber for 6 hours daily for two weeks, where, the THI was from 70 to 71. The results revealed that expression level of the Hsp genes (Hsp72, Hsp70.1, Hsp70 and Hsp47) was lower under air condition treatment than under summer heat stress. Rectal temperature and respiration rate were significantly lower ($p < 0.01$) under air condition treatment than those under heat stress. Total triiodothyronin (T3) level was significantly higher ($P < 0.05$) in AC cooling treatments than in HS, while cortisol level was significantly lower ($P < 0.01$) in AC cooling treatment than in HS calves. Creatinine and Urea -N levels were significantly lower ($P < 0.01$) in AC cooling treatment than in HS calves. Triglycerides, ALT and AST levels were significantly lower ($p < 0.01$), ($P < 0.01$) and ($p < 0.05$), respectively in AC cooling treatment than in HS calves. These results demonstrated that there is a relationship between the molecular weight of HSPs and the level of HSPs gene expression. The higher the molecular weight (HSP 72) the lower is the HSPs gene expression level (0.82 in HS and 0.39 in AC) and vice versa. This holds true in both heat stress and air condition. AC treatment is capable to ameliorate heat stress of Friesian calves under hot summer climate.

Keywords: *Heat stress, Friesian calves, Cooling system, Heat Shock Protein genes, Hormonal, Biochemical and physiological parameters.*

INTRODUCTION

Early and recent investigators concerning the means of alleviating heat stress in cattle have been undertaken to test the benefit of lowering body temperature and respiration rate as well as in correcting the upsets of certain biochemical parameters. These means of cooling animals include different systems such as cool air conditioning, water sprinkling, drinking cool water and shading⁽¹⁻⁶⁾.

However, air condition, though not economical especially under large barns, is thought to be an efficient cooling system as it combines the mean principal items of cooling, i.e. conduction, convection, radiation and evaporation, which satisfy the cooling needs of the animal.

Heat shock proteins are multi gene families that range in molecular size from 10–150 kDa and are found in all major cellular compartments. They are highly conserved proteins present in all the cells of living organisms and are essential for cellular viability⁽⁷⁾. HSPs are present in almost all the subcellular structures of all cell types from prokaryotes to eukaryotes⁽⁸⁾. Heat shock proteins are defined as molecular chaperones that regulate intracellular processes to maintain homeostasis during cell proliferation/differentiation and are essential for the maintenance of normal cell function⁽⁹⁾. The expression of HSPs is increased when the cells are exposed to several types of stressful stimuli e.g., elevated temperature, hypoxia, ischemia, heavy metals, radiation, calcium increase, glucose deprivation, cancer, inflammation, and microbial infection. Furthermore, HSPs play a role in innate and adaptive immune responses⁽⁸⁾. Heat shock proteins can be categorized into several families on the basis of their approximate molecular weight (HSP100, HSP90, HSP70, HSP60, as well as the small HSP family). HSP70 proteins constitute a major group that, even under normal growth⁽¹⁰⁾, can represent up to 1% of total cellular protein content. Among the different forms of the HSP70 family, HSP72 is considered to be the heat-inducible form, as its synthesis is usually restricted to the cell experiencing stress⁽¹¹⁾. However, several studies have shown that, under apparently non-stressful conditions^(12,13), HSP72 is also present at a low concentration in human cells and serum and in the plasma of apparently healthy dairy cows⁽⁴⁾.

This investigation was undertaken to find out the role of air condition in alleviating heat stress in cattle and its effect on heat shock protein level, since the literature is limited in this respect. Furthermore, body functions improvement using air condition treatment, as indicated by hormonal,

biochemical and physiological parameters measurements were investigated.

MATERIALS AND METHODS

Animals, climatic conditions and feeding

The present study was carried out on five male Friesian calves about 300-600 kg body weight. The animals were housed for two weeks in August in semi-open barn (heat stress) at Inshas area. The animals were maintained for two weeks (6 h daily) under air -conditioning (AC), inside a climatic chamber, which belongs to the project of “Bovine Adaptation to the Sahara” of the Egyptian Atomic Energy Authority. Air suction fan was set on during the rest of the day. Ambient temperature (AT) and relative humidity (RH %) were recorded at 11.00 and at 16.00 h daily under each condition (Table 1) using thermo-hygrograph. THI was obtained from the chart axes (AT, °C and RH, %) according to Armstrong⁽¹⁴⁾ as shown in Table (1).

Feeds were offered for the group once daily at 9.00 am, according to animals' body weights at a rate of 2.25 kg per 100 kg B.wt. of a commercial concentrate mixture. Wheat straw was offered at the rate of 1.5 kg per 100 kg B.wt. daily according to NRC⁽¹⁵⁾. Each 100 kg concentrates were supplemented with 0.5 kg sodium chloride, 1 kg di-calcium phosphate and 0.1 kg minerals and vitamins mixture. Animals were allowed to drink fresh water ad libitum.

Rectal temperature and respiration rate

Rectal temperature (RT) °C and respiration rate (RR) breaths per minute were measured three times a week at 16:00 pm during the two periods using a clinical thermometer inserted in rectum and counted flank movements per minute, respectively.

Table (1): Ambient temperature (°C), relative humidity(RH%) and temperature humidity index (THI) at 11.00 -16.00 under Egyptian summer season and air condition treatment.

Condition	Ambirnt temperature (°C)			Relative humidity (RH%)			(THI)		
	11.00	16.00	X ⁻	11.00	16.00	X ⁻	11.00	16.00	X ⁻
Summer									
X±SE	35.34±1.43	43.3±0.12	39.32	33.6±0.1	48.4±1.52	41.50	81.00	95.00	88.00
Aircondition									
X±SE	19.75±28	23.6±0.52	21.68	38.8±0.6	42.5±07	40.70	70.00	71.00	70.50

Blood sampling

Blood samples were withdrawn from the jugular vein at 16:00 pm at the end of each exposure period of summer and air conditioning. Part of the blood samples were kept immediately under ice while transferred to the laboratory in 2 h after collection to be kept under refrigeration for HSP gene expression determination. The other part of blood samples were collected into test tubes containing EDTA as anticoagulant were centrifuged at 3500 rpm for 10 minutes and plasma was stored in a deep freezer at -20°C till blood analysis was undertaken.

Plasma hormones

The determination of plasma total T3 and cortisol was carried out using radioimmunoassay (RIA) technique (Immunotech, A Beckman Coulter Company, France). This assay is based on the competition between each iodine-125 labeled hormone, which was added to the samples, and the respective unlabeled hormone found in the samples for a fixed and limited number of antibody sites. The radioactivity of the amount of bound iodine-125 labeled T3 or cortisol hormone was counted separately in gamma counter. The counts were determined inversely related to the amount of unlabeled T3 or cortisol in the sample.

Blood biochemical parameters

Level of plasma urea and creatinine, triglycerides, ALT and AST was determined by quantitative-colorimetric methods using chemical reagent kits. Urea determination method described by Henry⁽¹⁶⁾. Creatinine determination was carried out according to this method described by Allston⁽¹⁷⁾. Triglycerides was determined colorimetrically with enzyme that produces hydrogen peroxide technique described by Fossati and Prencipe⁽¹⁸⁾. ALT and AST were determined by quantitative enzymatic-colorimetric method based on liver function described by Scherwin⁽¹⁹⁾.

Expression of Heat Shock Proteins (HSP) genes

Extraction of total RNA from blood samples

Total RNA was isolated from 300 to 500 μl of blood samples collected from Friesian calves exposed to heat stress or air condition. The blood samples were centrifuged for 5 min at 2000 g, then the pellet of the cells was used to extract the total RNA by the standard TRIzol extraction method (Invitrogen, Paisley, UK) and recovered in 100 μl of diethyl pyrocarbonate (DEPC)-treated

water. In order to remove any possible genomic DNA contamination, the total RNA samples were pre-treated using DNA-free™ DNase and removal reagents kit (Ambion, Austin, TX, USA) following the manufacturer's protocol.

Synthesis of the first cDNA strand

The complete Poly (A)- RNA isolated from both treatments blood samples were reverse transcribed into cDNA in a total volume of 20 µl using 1 µl aligo (dT) primer. According to Ali et al.⁽²⁰⁾, the composition of the reaction mixture (MM), consisted of 50 mM MgCl₂, 200 U/µl reverse transcriptase (RNase H free), 10x reverse transcription (RT) buffer (50 mM KCl, 10 mM Tris-HCl, pH 8.3), 10 mM of each dNTP (Amersham, Brunswick, Germany), and 50 µM of aligo (dT) primer. The RT reaction was carried out at 25°C for 10 min, followed by 1 h at 42°C, and finished with denaturation step at 99°C for 5 min. Afterwards the reaction tubes containing RT preparations were flash-cooled in an ice chamber until being used for cDNA amplification through polymerase chain reaction (PCR).

Semi-quantitative RT-PCR

The first strand cDNA of calf samples was used as templates for RT-PCR with the specific primers of several HSP genes. The sequences of specific primer of the genes used⁽²¹⁻²³⁾ and product sizes are listed in Table (2). β-Actin was used as a housekeeping gene for normalizing mRNA levels of the target genes. According to Ali *et al.*⁽²⁰⁾, the reaction mixture for RT-PCR was consisted of 10 mM dNTP's, 50 mM MgCl₂, 10x PCR buffer (50 mM KCl, 20 mM Tris-HCl, pH 8.3, Gibco BRL, Eggenstein, Germany), and autoclaved water. RT-PCR amplification with Hsp72, Hsp70.1, Hsp70 and Hsp47 gene-specific primers was performed for 45 (Hsp72), 36 (Hsp70.1), 42 (Hsp70) and 40 (Hsp47) cycles. The PCR cycling parameters were one cycle of 94°C for 5 min, 36-45 cycles of 95°C for 10 s, 55°C to 61°C (Table 2) for 30 s, 72 °C for 40 s, and a final cycle of 72 °C for 7 min. The PCR products were then loaded onto 2.0% agarose gel, with PCR products derived from β-actin of the different fish samples. The PCR reaction was repeated at least five times within each group.

Table (2). Primers sequences used for RT-PCR

Gene	Primer sequence (5 ^f -3 ^f) ^a	Product length (bp)	Annealing T _m (°C)
Hsp72	F: 5'-AACATGAAGAGCGCCGTGGAGG-3' R: 5'-GTTACACACCTGCTCCAGCTCC-3'	169	61
Hsp 70.1	F:AAGGTGCTGGACAAGTGCC AGGAGGTGATT R:ACTTGAAGTAAACAGAAACGGGTGAAAAA	488	59
Hsp70	F: GTCATCAACGACGGAGACAA R: GGTGCTGGACGACAAGGT	555	59.4
Hsp47	F: CCA GGA AAT GGC ACA TGT AT R: TAT AAG CAT GCT GTC GGG TC	290	60
β-Actin	F: TGG GGC AGT ATG GCT TGT ATG R: CTC TGG CAC CCT AAT CAC CTC T	165	55

^a F: forward primer; R: reverse primer. T_m: temperature.

Statistical analysis

The binomial data for semi-quantitative RT-PCR analysis showed normal distribution. Heat shock proteins data were analyzed using the General Liner Models (GLM) procedure of Statistical Analysis System (SAS)⁽²⁴⁾, followed by Scheffé-test to assess significant differences between treatments. All statements of significant were based on probability of $P < 0.05$. The effect of heat stress on various biological parameters in this study was tested statistically by Student “t” test⁽²⁵⁾ using the paired varians, since each calf was used as its control. The percentage changes in previous parameters due cooling treatment were calculated as follows: (treatment-hot) x100/treatment). Improvement index was estimated by subtracting the change percent-age from 100.

RESULTS AND DISCUSSION

Effect of the use of air condition in cooling heat stressed Friesian calves on physiological parameters.

Table (3) shows that treatment of heat stressed Friesian calves using air-conditioning reduced rectal temperature (RT) by 4.2 % ($P < 0.05$) and respiration rate (RR) was by 246 % ($P < 0.01$). Cooling treatment reduced RT and RR in heat stressed Freisian calves under Egyptian summer climate, thus air-conditioning was effective in improving animal's physiological responses.

In Louisiana experiments showed cooling benefits from air movement and wetting the cow's body surface. Collier, *et al.* ⁽²⁶⁾ reported that sprinkling cows before entering a shade reduced respiration rate by 65 to 81% and body temperatures by 46 to 50% over shade alone. Similarly, in Florida researchers reported an 11.6% improvement in milk yield when cows were sprayed for 1.5 min of every 15 min. Cooled cows had sharply reduced respiratory rate (57 vs. 95 breaths/min), and efficiency of production (kg of milk per kg of DMI) was improved for cooled cows. Marcillac-Emberson *et al.* ⁽²⁷⁾ reported that sprinkler system that completely wets the cow by soaking through the hair coat to the skin is more effective than a misting system. Heifers in shaded vs. sprinkler treatments had decreased respiration rates (13%).

Evaporation of water is an effective mean of cooling the body. Although only 1 calorie is needed to elevate the temperature of 1 g of water by 1°C, it takes almost 580 calories to evaporate the same amount of water from the body. At ordinary temperature and humidity, about 25 percent of the heat produced in resting mammals is lost by evaporation of water from the skin and respiratory passages. This insensible water loss, coetaneous and respiratory, is rather constant under basal conditions. An increased flow of blood through the skin causes it to increase somewhat, but the mechanisms of sweating and panting offer much efficient ways to increase the evaporative heat loss ⁽²⁸⁾.

****Table (3): Reducing rectal temperature and respiration rate in heat stressed (HS) Friesian calves using air condition (AC) treatment.**

Parameters (THI)	HS	AC	change	
	81-95	70-71		%
RT $\bar{X} \pm SE$ (°C)	40.5 \pm 0.32	38.8 \pm 0.11	-4.2*	\pm 0.99
RR $\bar{X} \pm SE$ (breath /min.)	78.2 \pm 2.92	22.6 \pm 0.72	-246**	\pm 5.34

* Significant at $p < 0.05$ ** Significant at $p < 0.01$

Gene expression study

Level of expression of heat-shock proteins genes (Hsp72, Hsp70.1, Hsp70 and Hsp47) in the blood samples of Friesian calves exposed to heat stress during hot summer season and then treated by cool air condition is summarized in Table (4) and Figures (1-4). The results revealed that expression level of the Hsp genes (Hsp72, Hsp70.1, Hsp70 and Hsp47) was lower in the cool air condition treatment than in summer heat stress by 52.4, 50.4, 50.4 and

50.0%, respectively (Table 4 and Figs. 1-4). Moreover, the expression level of Hsp72 gene in each of HS and AC treatment was lower than the other Hsp genes. On the contrary, the expression Hsp47 gene level in each of HS and AC treatment was higher than the other Hsp genes.

It is also noted (Table 4) that there is a relationship between the molecular weight of HSPs and the level of HSPs gene expression. The higher the molecular weight (HSP 72) the lower is the HSPs level (0.82 in HS and 0.39 in AC). This holds true in both heat stress and air condition. Welch ⁽¹¹⁾ stated that among the different forms of the HSP70 family, HSP72 is considered to be the heat-inducible form, as its synthesis is usually restricted to the cell experiencing stress.

The expression of Hsp72 gene was significantly higher during summer HS than cool AC treatment (Fig.1). Hsp70.1 and Hsp70 genes expressed in Friesian calves showed that their expression level was significantly higher during summer HS than AC treatment (Figs. 2 and 3). On the other hand, the level of expression was higher in Hsp70 than Hsp70.1 during the both treatments (Fig.2 and 3). In the same trend, the expression level of Hsp47 gene was significantly over expressed during summer HS than cool AC treatment (Fig.4).

The level of Hsp72 expression was lower than the other Hsp genes in Friesian calves exposed to heat stress during summer season or to cool air condition procedures. In agreement with these findings, Kristensen *et al.* ⁽⁴⁾ reported that bovine Hsp72 is absent or expressed at a low level under non stress conditions and is referred to as the inducible form of the Hsps family ⁽¹¹⁾. There are several studies explaining the lower amount of Hsp72 mRNA found in Friesian cow in our study compared to other Hsp genes. These studies performed on bovine or other species evaluated mRNA levels or synthesis of Hsp72 or Hsp70 following short-term exposure to heat-stress conditions. Kamwanja *et al.* ⁽²⁹⁾ found higher cellular resistance to elevated temperatures in lymphocytes from Brahman and Senepol cow with respect to Angus cow. They found a tendency for a lower amount of Hsp70 in the 2 thermo tolerant breeds. Hansen⁽³⁰⁾ speculated that the reduced Hsp70 expression in heat stressed Brahman and Senepol cow might be indicative of reduced protein denaturation (one of the signals for Hsp70 synthesis). Accordingly, others indicated that activation of Hsp genes is primarily related to the defence against cell damage consequent to heat shock⁽³¹⁾, or that Hsp70 expression is positively related with

increase of cell injury score⁽³²⁾. Recently, Kristensen *et al.*⁽⁴⁾ and Lacetera *et al.*⁽²³⁾ suggested that increase of Hsp72 expression might function as a biological indicator for changes in the stress level.

There are no measurements on the expression of HSP70.1, HSP70 and HSP47 mRNA in peripheral blood lymphocyte of Friesian calves exposed to high temperature regimens during the hot summer at Inshas area, which is a semi-arid desert area near the Nile valley under Egyptian summer heat stress (HS), that has not been conducted before. On the other hand, Lindquist⁽³³⁾ indicated that the heat-shock response is accompanied by a reduction in protein synthesis, favoring induction of the heat shock response over the ongoing gene program. These observations are consistent with our findings that the expression of HSP70.1, HSP70 and HSP47 genes were at the highest level during the hot season and may be attributed to low protein level.

It is worth mentioning that the decrease in Hsp genes (Hsp72, Hsp70.1 and Hsp70) was lower under air condition treatment than under summer heat stress by the same level 50% {Table (4) and Figures 1,2,3,4}. This result confirms that treated heat stressed calves with cool air condition is an effective method in lowering genes expression levels of the Hsp and amelioration of heat stress.

Increased levels of HSPs occur in response to environmental stresses, infection, normal physiological processes, and gene transfer. Stress can disrupt the physiology and productive performance of an animal. The increase in body temperature caused by heat stress has direct, adverse consequences on cellular function which can influence the reproductive performance⁽³⁴⁾. The current results suggest that further research is needed to verify whether mechanisms different from Hsp may exist in thermo-tolerance bovine. Hsp mRNA levels are associated with enhanced heat tolerance⁽³⁵⁾. The different steady-state mRNA levels of Hsp genes in the Friesian calves may be depended on transcriptional or post-transcriptional regulation of gene expression. Previous studies conducted indicated that mRNA levels of Hsp72 in cells exposed to stressful stimuli may depend either on transcriptional⁽³¹⁾ or on post-transcriptional regulation⁽³⁶⁾, of gene expression. Additionally, they demonstrated that among the different mechanisms through which translational regulation may occur (pre-mRNA splicing, mRNA transport, mRNA stability), post-transcriptional regulation of Hsp70 gene expression occurs mainly by mRNA stabilization.

Table (4): Heat- induces changes in expression of HSP genes in Friesian calves.

Heat Shock Proteins	HS	AC	Change%
$\bar{X} \pm SE$	Calves	Calves	
Hsp72/B-actin	0.82±0.01	0.39±0.01	-52.44**
Hsp70.1/B-actin	1.23±0.04	0.61±0.04	-50.41**
Hsp70 B-actin	1.43±0.07	0.7±10.08	-50.35**
Hsp47/B-actin	1.52±0.03	0.76±0.03	-50.00**

** Significant at $p < 0.01$

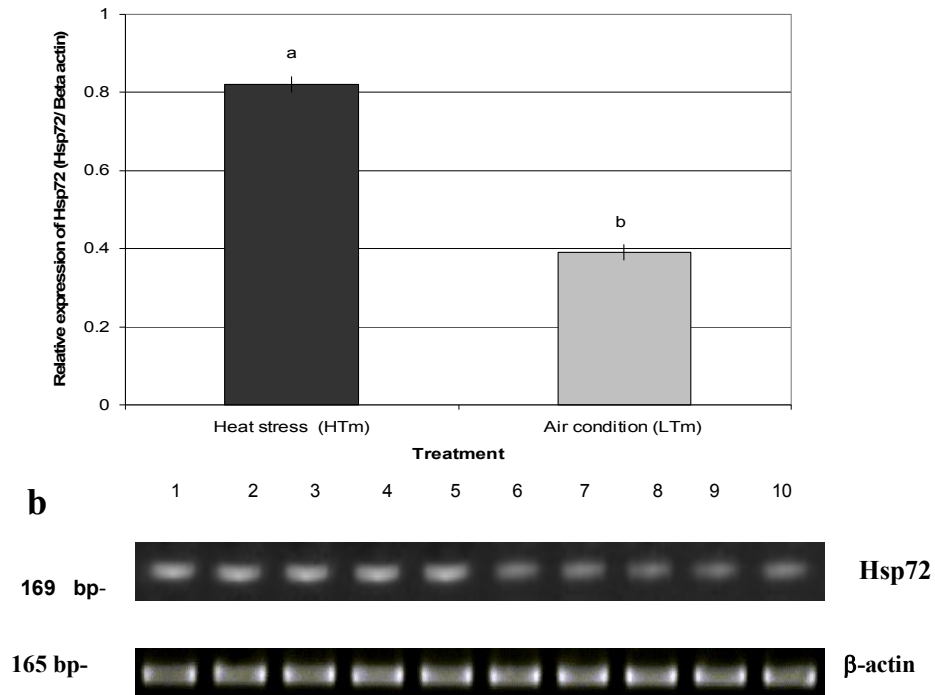


Fig. (1). Expression of Hsp72 gene in the blood of Friesian calves exposed to heat stress (lanes 1-5) and air condition (lanes 6-10) determined by semi-quantitative RT-PCR (a&b). The RNA recovery rate was estimated as the ratio between the intensity of Hsp72 gene and the β -actin gene. Within each column, means superscripts with different letters are significantly different ($P \leq 0.05$).

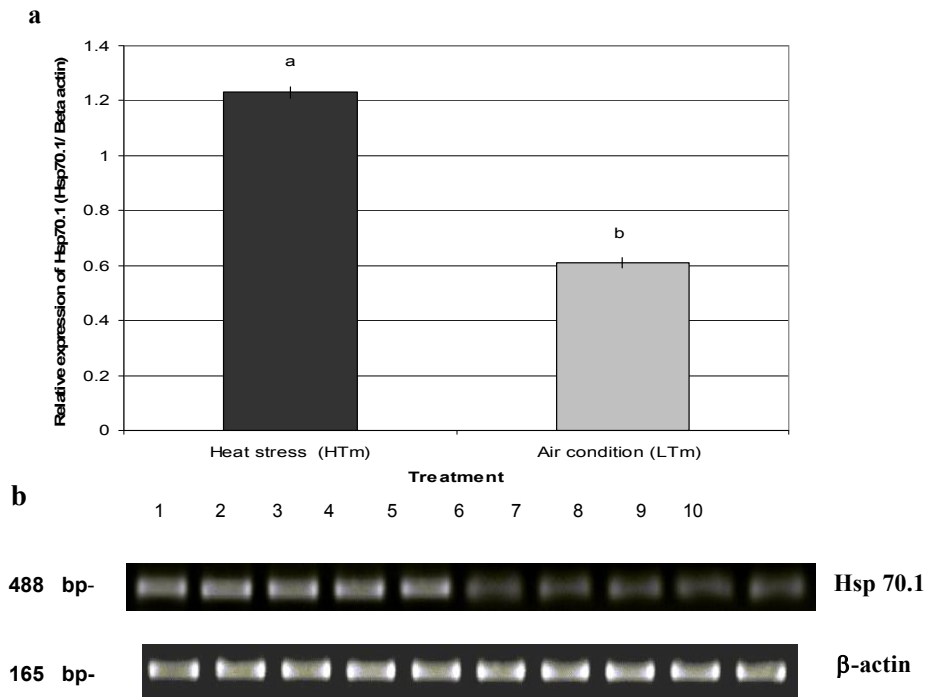
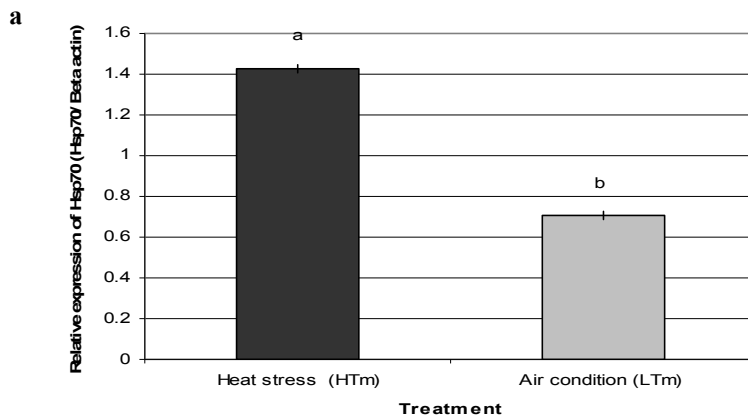


Fig. (2). Expression of Hsp70.1 gene in the blood of Friesian calves exposed to heat stress (lanes 1-5) and air condition (lanes 6-10) determined by semi-quantitative RT-PCR (a&b). The RNA recovery rate was estimated as the ratio between the intensity of Hsp70.1 gene and the β -actin gene. Within each column, means superscripts with different letters are significantly different ($P \leq 0.05$).



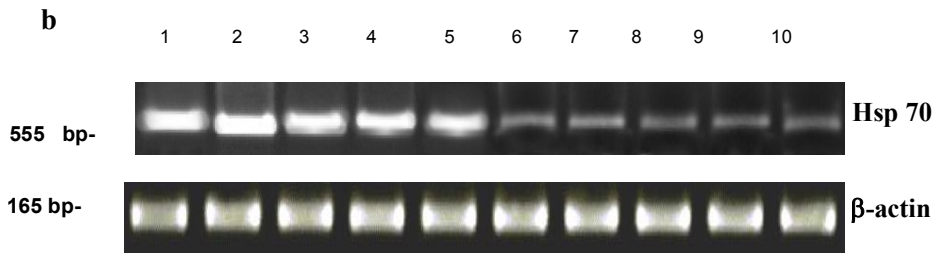


Fig. (3). Expression of Hsp70 gene in the blood of Friesian calves exposed to heat stress (lanes 1-5) and air condition (lanes 6-10) determined by semi-quantitative RT-PCR (a&b). The RNA recovery rate was estimated as the ratio between the intensity of Hsp70 gene and the β -actin gene. Within each column, means superscripts with different letters are significantly different ($P \leq 0.05$).

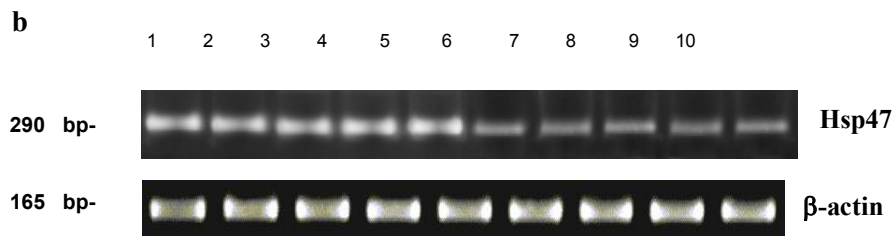
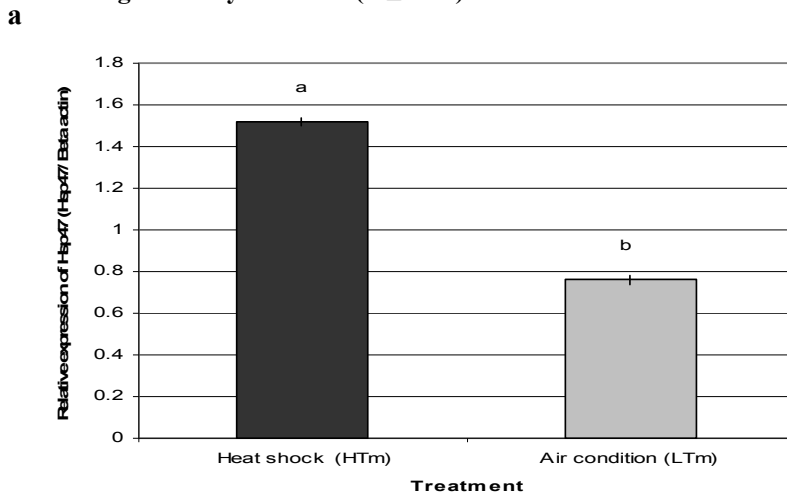


Fig. (4). Expression of Hsp47 gene in the blood of Friesian calves exposed to heat stress (lanes 1-5) and air condition (lanes 6-10) determined by semi-quantitative RT-PCR (a&b). The RNA recovery rate was estimated as the ratio between the intensity of Hsp47 gene and the β -actin gene. Within each column, means superscripts with different letters are significantly different ($P \leq 0.05$).

Effect of the use of air condition in cooling heat stressed Friesian calves on Triiodothyronine and Cortisol plasma levels

Table (5) shows that the treatment of summer heat stressed Friesian calves by air conditioning led to a significant ($P < 0.05$) increase in the average of triiodothyronine plasma level by 48.1%. The same treatment led to a significant ($P < 0.01$) decrease in the average of cortisol levels by 39.2 %. These findings are in agreement with those of *Kamal et al.*,⁽¹⁾ who reported that treated heat stressed Friesian calves with air conditioning increased thyroxin blood concentration from 57.8 to 65.9 ng/ml. These results indicated that the cooling treatment alleviated the thermal hormonal alterations, in T3 and cortisol which took place under a hot summer climate. Consequently, the cooling process by AC may improve production functions as indicated by *Kamal et al.*⁽¹⁾.

Serum cortisol concentration was significantly higher in heat-stressed cows than under air conditioning⁽³⁷⁾. Plasma cortisol concentration of non-shaded cows was higher ($p < 0.05$) than shade cows⁽³⁸⁾. When THI increased from 68 to 78 cortisol concentration was significantly increased while free T3 decreased in crossbred Friesian-Holstein cows⁽³⁹⁾. Serum T3 concentration was significantly Lower in heat-stressed bulls than in bulls under thermoneutral condition⁽⁴⁰⁾.

Baei⁽⁴¹⁾, stated that the effects of HS on DM1 and blood chemistry in dairy cattle can be alleviated by artificial cooling methods designed to counter the effect of high environmental temperature.

Effect of the use of air condition in cooling heat stressed Friesian calves on Creatinine and Urea-N blood levels

Table (4) shows that the treatment of the summer heat stressed Friesian calves by air conditioning led to a significant ($P < 0.01$) decrease in the average of creatinine by 24.8% and level and the average of Urea-N level was decreased significantly by 23.6%, respectively. The decrease in both creatinine and urea-N due to cooling treatment led to less tissue catabolic processes thus improved the body function. Heat stress increased plasma Urea-N levels in lactating Holstein cows from 11.5 mg/dl under thermoneutral condition to 14.8 mg/dl under heat stress in climatic chamber⁽⁴²⁾.

In conclusion, air conditioning cooled the animal's surface by conduction, convection, radiation and evaporation. Thus decreasing the heat load of summer by increasing the heat loss through urine excretion and skin vaporization. This reduction in the heat load improved the appetite of the animal

which caused an increase in feed consumption and consequently in gain and feed efficiency. In addition, the cooling treatment alleviated the thermal hormonal alterations, especially T3 and other factors which depressed the growth under a hot summer climate. Consequently, using the cooling process may be spared for production functions⁽¹⁾. The reduction in the summer heat load due to cooling also aided the animal to reach a steady physiological state as indicated by restored blood parameters level, decreased rectal temperature and respiration rate.

Effect of the use of air condition in cooling heat stressed Friesian calves on Triglycerides, ALT and AST blood levels

Table (4) shows that the treatment of heat stressed Friesian calves by air condition led to a significant ($P<0.01$) decrease in Triglycerides blood level by 17.7% compared with heat stress period. It is noted (Table 4) that the treatment of the summer heat stressed Friesian calves by air condition led to a significant decrease in the average of ALT ($P<0.01$) levels by 16.8 % and in the average of AST ($P<0.05$) levels by 10.2%. These results indicated that cooling treatment caused significant reductions in both transaminase enzymes activities.

Bahga *et al.*⁽⁵⁾ observed that ALT (units/ml) was significantly higher in the hot group (25.17) than in the cooled group (12.85). The hot group had higher AST, alkaline phosphatase, protein and cholesterol. It was concluded that higher values of ALT, alkaline phosphatase and cholesterol in the hot group could be taken as indices of heat stress in this species. Serum ALT and AST concentration was significantly higher in heat-stressed Holstein bulls than bulls under thermonutral condition⁽⁴⁰⁾ and in beef bulls⁽⁴³⁾.

Table (4). Reducing heat-stress (HS) in Friesian calves by using air condition (AC) treatment as indicated by changes in hormonal and biochemical parameters

Parameters $\bar{X} \pm SE$	HS	AC	change%
Total T 3 (ng /dl) $\pm SE$	52.4 \pm 0.3	117.9 \pm 19.2	48.1* \pm 11.4
Cortisol (ng /ml) $\pm SE$	4.4 \pm 0.3	2.7 \pm 0.1	-39.2** \pm 3.1
Creatinine (mg/dl) $\pm SE$	2.0 \pm 0.1	1.5 \pm 0.1	24.8** \pm 1.6
Urea-N (mg/dl) $\pm SE$	19.6 \pm 0.3	15.0 \pm 0.7	23.6** \pm 2.8
Triglycerides (mg/dl) $\pm SE$	139.2 \pm 9.4	114.4 \pm 7.8	-17.7** \pm 1.9
ALT (IU/L) $\pm SE$	12.4 \pm 1.2	10.4 \pm 1.2	-16.8** \pm 1.8
AST (IU/L) $\pm SE$	9.2 \pm 0.3	8.3 \pm 0.4	-10.2* \pm 3.1

* Significant at $p<0.05$

**Significant at $p<0.01$

CONCLUSION

In conclusion, this study demonstrated that there is a relationship between the molecular weight of HSPs and the level of HSPs gene expression. The higher the molecular weight (HSP 72) the lower is the HSPs level (0.82 in HS and 0.39 in AC). This holds true under both heat stress and air conditioning treatments. The Hsp genes studied represent precious biomarkers to detect variations of thermo-tolerance bovine conditions. Further research is needed to verify whether transcriptional or post-transcriptional mechanisms different from Hsp may exist in thermo-tolerance bovine. Air conditioning was an efficient method in improving calf body function. This results may be due to that cool air enter the respiratory system and induce internal cooling besides external cooling (surface cooling). Continued research evaluating methods to improve productive performance and nutritional status of thermally stress animals is needed.

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مجلة البحوث الإشعاعية والعلوم التطبيقية

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تخفيف ظروف العبء الحرارى للصيف فى مصر للعجول الفريزيان باستخدام التكييف

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- اجريت هذه التجربة على خمسة عجول فريزيان يتراوح اوزانهم من 300-600 كجم. خلال شهر اغسطس 2008. حيث تم تعريض العجول للعبء الحرارى تحت الظل صيفا لمدة اسبوعين وكانت مفكرة الحرارة والرطوبة العظمى من 81-88. ثم تم معاملة العجول بالتبريد بالتكييف داخل معمل الأقلمة لمدة ستة ساعات يوميا (10ص-4م) لمدة اسبوعين وكانت مفكرة الحرارة والرطوبة من 70-71.

- تم قياس درجة حرارة الشرج ومعدل التنفس خلال كل مرحلة وتم اخذعينات الدم فى نهاية كل مرحلة. - تم تقدير التعبير الجينى لبروتينات الصدمة الحراريه (70-70.1-72-47) بالدم فى المرحلتين. - تم تقدير مستوى اليوريا والكرياتينين والتراى جلسريدز وانزيمات الكبد بالدم فى المرحلتين. - تم تقدير مستوى هرمون الكورتيزول وهرمون التراى ايودوثيرونين بالدم فى المرحلتين. - تم عمل مقارنة للنتائج بين المرحلتين وذلك لمعرفة تأثير المعاملة بالتبريد بالتكييف على تخفيف العبء الحرارى لعجول الفريزيان صيفا.

اظهرت النتائج ان معاملة عجول الفريزيان المعرضة للعبء الحرارى صيفاً بالتبريد بالتكييف ادت الى: - إنخفاض معنوى (0.01) فى درجة حرارة الشرج ومعدل التنفس. - إنخفاض معنوى (0.05) فى مستوى التعبير الجينى لبروتينات الصدمة الحراريه (70-70.1-72-47) بالدم.

- إنخفاض معنوى (0.01) فى مستوى هرمون الكورتيزول وهرمون التراى ايودوثيرونين بالدم. - إنخفاض معنوى (0.01) فى مستوى اليوريا والكرياتينين والتراى جلسريدز وانزيمات الكبد بالدم.

الخلاصة:

اوضحت نتائج هذه الدراسة وجود علاقة بين الوزن الجزيئى لبروتين الصدمة الحرارية ومستوى التعبير الجينى. الاعلى فى الوزن الجزيئى هو الأقل فى مستوى التعبير الجينى والعكس صحيح. وهذه العلاقة قائمة فى حالة التعرض للعبء الحرارى صيفا وكذلك بالمعاملة بالتبريد بالتكييف.

كما اوضحت ايضاً ان معاملة عجول الفريزيان المعرضة للعبء الحرارى صيفاً بالتبريد بالتكييف ، ادت الى خفض مستوى التعبير الجينى لبروتينات الصدمة الحرارية والى تحسين حالة الحيوان الهرمونية والبيولوجية والفسيولوجية مما ادى الى تخفيف العبء الحرارى الواقع على الحيوان صيفا .

يمكن استخدام التعبير الجينى لبروتينات الصدمة الحراريه فى انتخاب الحيوانات المقاومة للجو الحار. وتشير الدراسة الى الاحتياج الى عمل أبحاث اخرى مستقبلية فى هذا المجال.