

## EFFECT OF ATMOSPHERIC TEMPERATURE ON BLOOD CHANGES OF TOTAL OXIDATIVE STRESS INDEX, (OSI) AND LIPID PROFILE IN PERIPARTUM EWES

GHADA A.E. MOHAMED<sup>1</sup> and HAYAM M. A. MONZALY<sup>2</sup>

<sup>1</sup> Biochemistry and Nutritional Deficiency Diseases Unit., Animal Health Research Institute, Assiut Branch, Agriculture Research Center, Egypt

<sup>2</sup> Animal Production Research Institute- Sheep and Goat Research Department.

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### ABSTRACT

Oxidative stress index (OSI) is recently used as a valid tool for providing an in depth picture about redox status and oxidative stress. The study was conducted on Fourteen multiparous, singleton bearing Osimi ewes (1.5-2 years, 35-40 kg) belonging to agriculture research station, Assiut district, A.R. Egypt during spring (as controls, N=7) and during summer (as heat stressed, N=7). Ewes were blood sampled weekly throughout the last three weeks prepartum and weekly throughout the first three weeks post-lambing for determination of blood serum lipids and oxidative stress biomarkers. Results showed increased blood serum total cholesterol, triglycerides, high density lipoprotein (HDL) and low density lipoprotein LDL ( $p<0.05$ ) before lambing and then decreased ( $p<0.05$ ) after lambing. Total Cholesterol, triglycerides and HDL was lower ( $p<0.05$ ) in summer compared to spring. In spring and summer, total peroxide (TPX) increased at the first week ( $p<0.05$ ) before and after lambing in addition to summer values were higher ( $p<0.05$ ) than spring values. In spring, total antioxidant capacity (TAC) increased at the second week ( $p<0.05$ ) after lambing, whereas a reverse trend was noticed in summer season, so that summer TAC values were lower ( $p<0.05$ ) than spring values. Pearson's Product Moment Correlation between TPX and TAC did not reveal significant indication. However, values of oxidative stress index ( $OSI = TPX / TAC \times 100$ ) progressively increased ( $p<0.05$ ) in summer and reached its maximal value at the first week after lambing, then steadily decreased until reached the basal data at the third week after lambing. Summer TPX values were higher ( $p<0.05$ ) than spring values. This study is the first to evaluate the degree of OS in HS ewes during the peripartum period by detection of TPX, TAC and OSI values. Our data clarified that the maximal oxidative stress occurred at the first week after parturition that was associated with an increase of pro-oxidants rather than reduction of antioxidants during spring. Instead, both the increase in pro-oxidants and the reduction of antioxidants are sharing in the oxidative stress in summer.

**Key words:** Ewes-Lactation-pregnancy-heat stress- lipid profile.

### INTRODUCTION

High environmental temperatures in tropical and subtropical areas challenge the animal's ability to maintain the balance of its body (Silanikove, 2000). An imbalance between metabolic heat production inside the animal body and its dissipation to the surroundings results to heat stress (McKinley, *et al.*, 2017). Heat stress (HS) reduces the metabolic rates and alters post-absorptive metabolism, regardless of the decreased feed intake (Baumgard and Rhoads, 2013; Sejian *et al.*, 2018). It also suppresses the immune and endocrine system, thereby enhances susceptibility of an animal to various diseases and health problems (Das *et al.*, 2016). Therefore, determining the energetic of

heat-stressed animals would presumably provide evidence on how to better treat animals suffering from heat-related illnesses (Johnson *et al.*, 2015, Lees *et al.*, 2017).

Recently, it has been shown that heat stress disturbs the steady state concentrations of free radicals, resulting in both cellular and mitochondrial oxidative damage (Pandey *et al.*, 2012). Belhadj *et al.* (2014) qualified heat stress as cytotoxic, as it alters biological molecules, disturbs cell functions, modulates metabolic reactions, induces oxidative cell damage and activates both of apoptosis and necrosis pathways. The drastic effects of HS depend on its duration and severity (Belhadj *et al.*, 2016).

Lipids especially polyunsaturated fatty acids are sensitive to oxidation forming a complex series of compounds, leading to the term lipid peroxidation (Halliwell & Gutteridge, 2017). Recent studies showed that the use of oxidative stress markers as a

Corresponding author: Dr. GHADA A.E. MOHAMED  
E-mail address: dr\_kada2012@yahoo.com  
Present address: Biochemistry and Nutritional Deficiency Diseases Unit., Animal Health Research Institute, Assiut Branch, Agriculture Research Center, Egypt

measure of oxidant-antioxidant balance could provide complementary information about the homeostasis of the animal than conventional metabolic parameters alone (Gutteridge & Halliwell, 2018).

Since the measurement of different oxidant molecules separately is not practical and their oxidant effects are additive, the total oxidant status of a sample is measured and this is named total peroxide (TPX) (Erel, 2005).

Serum concentrations of antioxidants can be measured separately in the laboratory, but these measurements are time-consuming, labor intensive and costly. Since the effects of the antioxidant components in serum are additive, measurement of the (TAC) accurately reflects the redox status of the serum (Halliwell & Gutteridge, 2017; Gutteridge & Halliwell, 2018).

Abuelo, *et al.* (2013) introduced the term Oxidative Stress Index (OSI), based on the ratio between TPX and serum TAC, that is, (TPX/ TAC), comparing the information given by this parameter with those given by TPX and TAC separately. Under the conditions of oxidative stress, the OSI provides an objective assessment of the relationship between oxidants and antioxidants, not seen by the determination of both components separately. An increase in TPX/ TAC ratio indicates a higher risk for OS because of an increase in ROS production, defensive antioxidant consumption, or both.

Pregnancy is a physiological process characterized by a drastic increase in energetic and oxygen demands, to ensure an adequate fetal development and growth, thus, both mother and fetus are likely to experience oxidative stress, during pregnancy (Mutinati *et al.*, 2013). The transition peripartum period is a critical phase and particularly important for health of dairy animals (Ingvarsen and Andersen, 2000). The high metabolic demand in late pregnancy and early lactation period may also induce oxidative stress (Fassah *et al.*, 2015).

There is a lack of information concerning the interaction between oxidative and antioxidant status in periparturient ewes especially under Upper Egypt conditions. In addition, determination of serum TPX and TAC with calculation of OSI value have not been used in ewes, especially under different production and reproduction phase. The nature of this mechanism still indistinct and the significance of their levels remain unclear. Therefore, the goal of this study was to document the effect of HS on the changes in lipid profile as a biomarker of energy metabolism, in addition to evaluation of redox balance. The redox included TPX as a biomarker of oxidants and TAC as a biomarker of antioxidant status in the peripartum ewes reared in winter and

summer seasons. Further, to evaluate the oxidative stress by the estimation of OSI based on the ratio between TPX and the TAC during the late pregnancy and early lactation periods in ewes.

## MATERIALS AND METHODS

The study was carried out at Agriculture research station, Assiut district, A.R. Egypt. And done during March and April (representing spring, the thermoneutral zone) and July and August (representing mid-Summer; the hot period). Weather data were obtained from the local weather station. Briefly, in summer, this area is characterized by intensified air temperature, intensified sun radiation, long sunshine duration and low relative humidity, while in spring it is relatively thermoneutral Rainfall is almost negligible. The recorded arithmetic means of ambient air temperature and relative humidity during spring was 24°C, 36% respectively, whereas during mid-Summer they were 44.13°C, 27%, respectively.

### Animals:

Fourteen multiparous, singleton bearing Osimi ewes (1.5-2 years, 35-40 kg). Clinically healthy were used during spring (as controls, N=7) and during summer (as heat stressed, N=7). During spring, ewes allowed to graze on Egyptian clover pasture (*Trifolium Alexanderinum*). During summer, clover hay and silage were offered in addition to grazing on medium-to-low quality forages including crop stubbles and green fodder. During the peripartum period, in addition, concentrate mixture was offered (250 gm/ewe) consisting of (24% yellow corn, 38% cotton- seed meal, 34% wheat bran, 3% molasses and 1% salt). The selected animals were subjected to standard methods of clinical examinations to prove their fitness.

### Blood sampling:

Blood was sampled by jugular vein puncture into 10-ml clean test tubes without anticoagulant. Sampling was carried out weekly throughout the last three weeks pre- partum and weekly throughout the first three weeks post-lambing. The accurate estimation of the day of pre partum sampling was assessed in relation to the lambing time. Blood samples were centrifuged at 3000 rpm for 15 minutes to obtain serum, which was kept at -20°C until be used for biochemical assay.

### Biochemical analysis:

#### Blood serum lipid profile:

Total cholesterol (TC), Triglycerides (Trig) and High- density lipoprotein cholesterol (HDL) were measured using the standardized kit (Biodiagnostic, Dokki- Giza - Egypt), after the methods described by Richmond (1973), Fossati & Prencipe (1982) and Burstein *et al.* (1970), respectively. Low-density lipoprotein cholesterol (LDL) was calculated by

using the Friedewald formula (Friedewald *et al.* 1972) as following:  $LDL-C = TC - (HDL-C + triglycerides/5)$ .

#### **Blood serum Total Antioxidant Capacity (TAC) concentration:**

Total antioxidant capacity was measured using the standardized kit (Biodiagnostic, Dokki- Giza - Egypt), based on the reaction of antioxidants in the sample with a defined amount of exogenously provide hydrogen peroxide ( $H_2O_2$ ). The antioxidants in the sample eliminate a certain amount of the provided hydrogen peroxide. The residual  $H_2O_2$  is determined colorimetrically by an enzymatic reaction which involves the conversion of 3,5, dichloro-2-hydroxy benzensulphonate (ABTS) to a colored product. This has a relatively stable blue-green colour which is measured at 600 nm. For accuracy and reproducibility control, total antioxidant control was used after the method described by Koracevic *et al.* (2001).

#### **Blood serum Total Peroxide (TPX) concentration:**

Blood serum TPX concentration was measured as a  $H_2O_2$  equivalent after the method described by (Erel, 2005). This method based on the oxidation of ferrous iron (as a catalyst) to ferric iron by the various types of plasma peroxides in the presence of xylenol orange in an acidic medium, which binds with iron(III) and forms a colored complex whose absorbance can be measured at 560 nm.

#### **Oxidative stress index (OSI):**

To perform the calculation, the OSI value was calculated using the following formula:  $OSI = [(TOS, \mu mol/L) / (TAC, mmol equivalent/L) \times 100]$  according to Erel (2005).

#### **Statistical analysis:**

The results were analyzed by analysis of variance (ANOVA) followed by student "t" test for the comparison between seasons. F test followed by pair-wise comparisons (Duncan's new multiple range test) were used for the comparison between periods within the peripartum period of each season. Pearson Product Moment Correlation (PPMC) was performed on paired data obtained by individual cases for TPX and TAC. Data correlation (r), linear regression analysis (R<sup>2</sup>) and significance level (P-value) were estimated. The computer packaged software program SPSS was used for analysis according to (SPSS, 2002). Significance level was set at  $P < 0.05$ .

## **RESULTS**

The recorded mean ambient temperature and relative humidity in spring was 24°C, 36% respectively, whereas during mid-Summer they were 44.13°C, 27%, respectively. The mean value ( $\pm SE$ ) of rectal

temperature °C of ewes was 38.91 $\pm$ 0.08 and 39.47 $\pm$ 0.07 in spring and mid-summer respectively. The values significantly differ at ( $P < 0.05$ ).

Blood serum total cholesterol concentration in spring was significantly increased at the first week ( $p < 0.05$ ) before lambing, then significantly decreased ( $p < 0.05$ ) at the first week ( $p < 0.05$ ) after lambing and returned to the base line level in the second and third weeks. In summer, it was significantly decreased at the first week ( $p < 0.05$ ) before lambing, and at the second week ( $p < 0.05$ ) after lambing. The values were significantly lower in summer at the first week prepartum and the second week postpartum when compared with the spring values (Tab. 1, Fig. 1).

In spring, Blood serum triglyceride concentration was significantly increased at the first week ( $p < 0.05$ ) before lambing, then significantly decreased ( $p < 0.05$ ) at the first week ( $p < 0.05$ ) after lambing and returned to the base line level in the second and third weeks. In summer, it was significantly decreased at the second week ( $p < 0.05$ ) before lambing, and at the first week ( $p < 0.05$ ) after lambing. The values were significantly lower at the first and second weeks prepartum and the first week postpartum when compared with the spring values (Tab. 1, Fig. 1).

Blood serum HDL concentrations in spring were significantly increased at the first week ( $p < 0.05$ ) before lambing, and then significantly decreased ( $p < 0.05$ ) at the third week ( $p < 0.05$ ) after lambing. In summer, HDL concentration was significantly decreased at the third week ( $p < 0.05$ ) after lambing. The values were significantly lower in summer at the first week prepartum and the first week postpartum when compared with the spring values (Tab. 2, Fig. 2).

In spring, LDL concentration was significantly decreased at the second week ( $p < 0.05$ ) after lambing and significantly increase first week before lambing. In summer, LDL concentration was significantly increased at the first week ( $p < 0.05$ ) before lambing, and decrease at the second and third weeks ( $p < 0.05$ ) after lambing. Season had no significant effect on LDL concentrations (Tab. 2, Fig. 2).

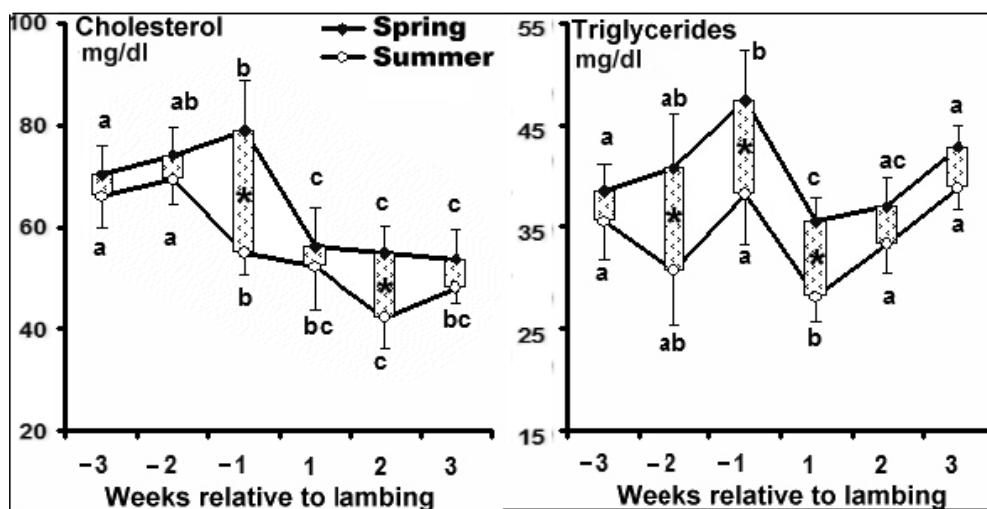
In spring and summer, blood serum TPX concentration was significantly increased at the first week ( $p < 0.05$ ) before and after lambing and continued at a high level ( $p < 0.05$ ) during the second week ( $p < 0.05$ ) after lambing then decreased to the baseline values at the remaining period. However, summer values were significantly higher ( $p < 0.05$ ) than spring values during the first week ( $p < 0.05$ ) before as well as the first week after lambing (Tab. 3, Fig. 3).

Blood serum Concentrations of TAC in spring increased at the second week ( $p < 0.05$ ) after lambing. A reverse trend was noticed in summer season, where the values were decreased at the first week before lambing and continued at low level during the first and second weeks after lambing when compared with the base line data. Summer values were significantly lower ( $p < 0.05$ ) than spring values during the first week ( $p < 0.05$ ) before as well as the first week after lambing (Tab. 3, Fig. 3).

Values of OSI in ewes progressively increased ( $p < 0.05$ ) during the peripartum period at summer and reached its maximal value at the first week after lambing, then steadily decreased till reached the basal data at the third week after lambing. Summer

values were significantly higher ( $p < 0.05$ ) than spring values during the first week ( $p < 0.05$ ) before as well as the first and second week after lambing (Tab. 3, Fig. 4).

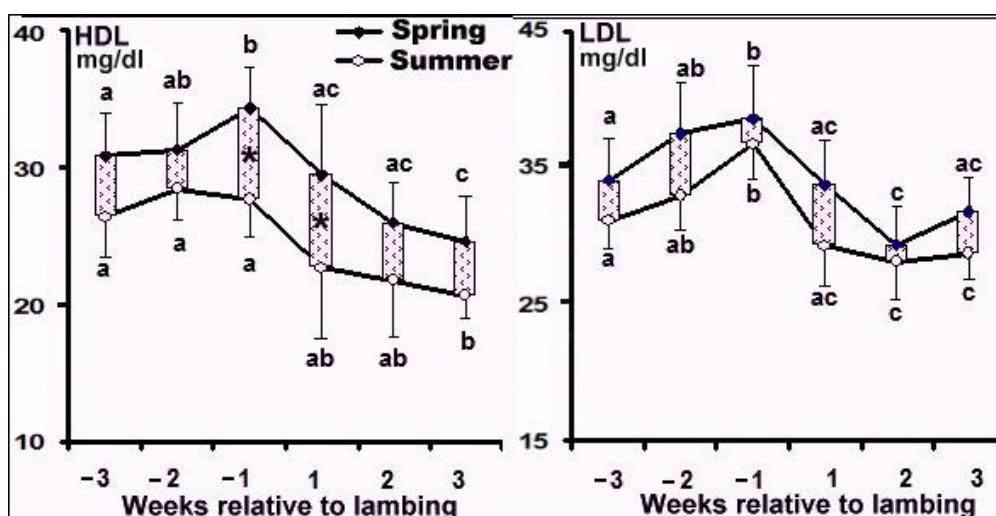
Pearson's correlation and regression analysis of TPX (as a representative value of the by-product of oxidative stress) with TAC (as a representative value of defense antioxidants) is presented in Fig. (4A). Linear regression analysis ( $R^2$ ) and Pearson's correlation ( $r$ ) of the paired data obtained by the individual cases did not reveal significant correlation between TPX and TAC ( $r = - 0.09$ ,  $R^2 = 0.017$  and  $P = 0.601$ ).



**Fig. 1:** Mean value of blood serum Total cholesterol and Triglycerides in ewes during the peripartum period at spring and summer (N=7).

a, b, c Duncan's multiple row test: different letters within the same season indicate significantly different results ( $p < 0.05$ )

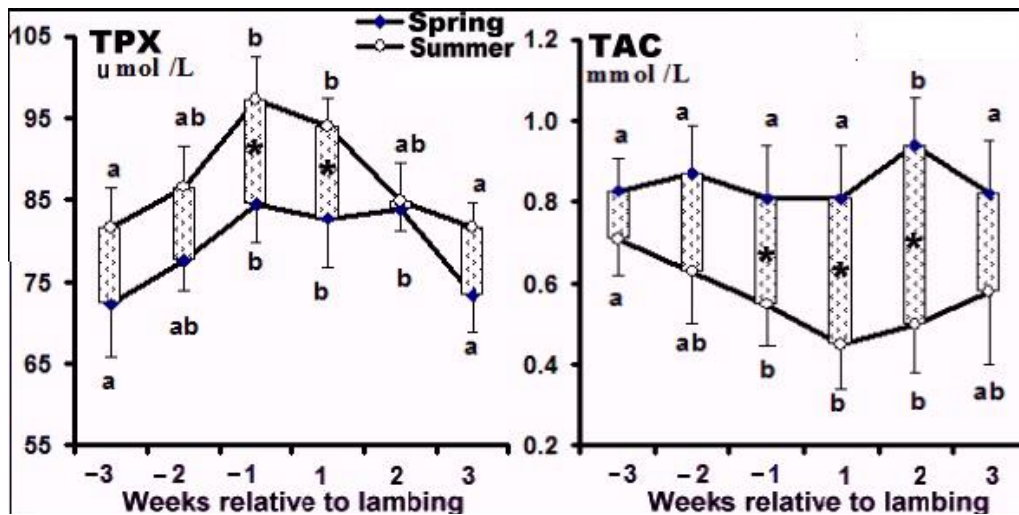
\* Within up down bars indicate significant different values between seasons



**Fig. 2:** Mean value of blood serum HDL and LDL in ewes during the peripartum period at spring and summer (N=7).

a, b, c Duncan's multiple row test: different letters within the same season indicate significantly different results ( $p < 0.05$ )

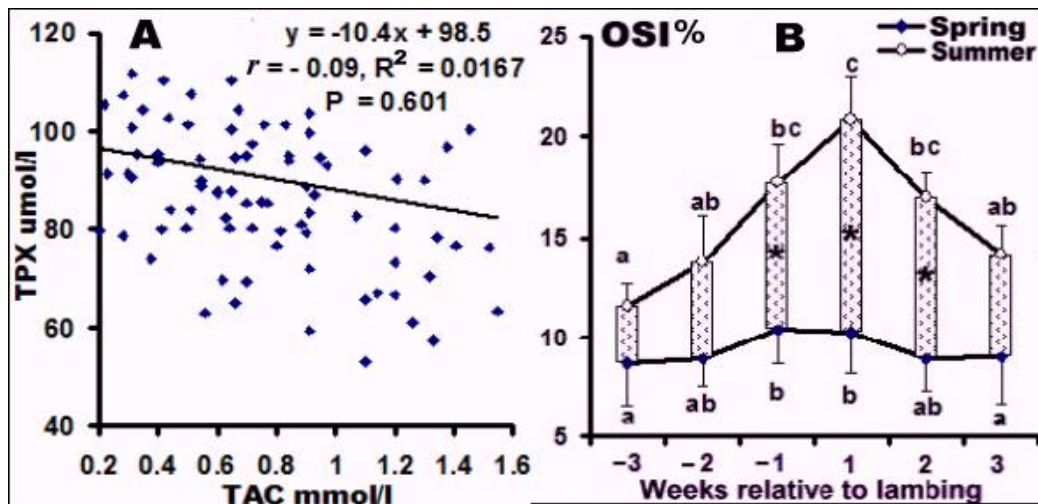
\* Within up down bars indicate significant different values between seasons



**Fig. 3:** Mean value of blood serum TPX (umol/L) and TAC (mmol/L) in ewes during the peripartum period at spring and summer (N=7).

<sup>a, b</sup>: Duncan's multiple row test: different letters within the same season indicate significantly different results (p<0:05)

\* Within up down bars indicate significant different values between seasons.



**Fig. 4. A:** Pearson's correlation (*r*), Linear regression (*R*<sup>2</sup>) analysis (of paired data obtained from individual ewes in both seasons, (N = 84) of the enhanced TPX and TAC concentrations.

**B:** Mean value of OSI in ewes during the peripartum period at spring and summer. <sup>a, b, c</sup> Duncan's multiple row test: different letters within the same season indicate significantly different results (p<0.05). \* Within up down bars indicate significant different values between seasons

**Table 1:** Mean value (±SE) of blood serum total cholesterol (mg/dl) and triglycerides (mg/dl) in ewes during the peripartum period at spring (N=7) and summer (N=7).

Parameter	Season	Weeks before lambing			Weeks after lambing		
		-3	-2	-1	1	2	3
Total Cholesterol	Spring	69.49±5.8 <sup>a</sup>	73.35±5.1 <sup>ab</sup>	77.60±13.5 <sup>b*</sup>	55.59±7.7 <sup>c</sup>	55.21±5.1 <sup>c*</sup>	53.66±5.4 <sup>c</sup>
	Summer	65.25±5.9 <sup>a</sup>	68.72±4.9 <sup>a</sup>	54.82±4.3 <sup>b</sup>	52.12±8.5 <sup>bc</sup>	42.47±6.2 <sup>c</sup>	47.87±3.1 <sup>bc</sup>
Triglycerides	Spring	38.94±3.3 <sup>a</sup>	40.71±4.4 <sup>ab*</sup>	46.02±4.2 <sup>b*</sup>	36.29±1.8 <sup>c*</sup>	37.17±2.5 <sup>ac</sup>	42.48±1.7 <sup>a</sup>
	Summer	36.29±3.5 <sup>a</sup>	30.98±4.2 <sup>ab</sup>	37.17±4.1 <sup>a</sup>	27.44±1.7 <sup>b</sup>	32.75±2.6 <sup>a</sup>	37.17±1.6 <sup>a</sup>

<sup>a, b, c</sup> Duncan's multiple row test: different letters within the same season indicate significantly different results (p<0:05)

\* Indicate significant different values between seasons

**Table 2:** Mean value ( $\pm$ SE) of blood serum HDL (mg/dl) and LDL (mg/dl) in ewes during the peripartum period at spring (N=7) and summer (N=7).

Parameter	Season	Weeks before lambing			Weeks after lambing		
		-3	-2	-1	1	2	3
HDL	Spring	30.50 $\pm$ 2.7 <sup>a</sup>	30.89 $\pm$ 3.1 <sup>ab</sup>	33.59 $\pm$ 2.7 <sup>b*</sup>	28.96 $\pm$ 4.6 <sup>ac*</sup>	25.87 $\pm$ 2.7 <sup>ac</sup>	24.71 $\pm$ 3.1 <sup>c</sup>
	Summer	26.25 $\pm$ 2.8 <sup>a</sup>	28.19 $\pm$ 1.9 <sup>a</sup>	27.41 $\pm$ 2.3 <sup>a</sup>	23.17 $\pm$ 5.8 <sup>ab</sup>	22.39 $\pm$ 3.8 <sup>ab</sup>	21.24 $\pm$ 1.5 <sup>b</sup>
LDL	Spring	35.14 $\pm$ 3.4 <sup>a</sup>	37.45 $\pm$ 3.8 <sup>ab</sup>	45.95 $\pm$ 3.8 <sup>b</sup>	33.98 $\pm$ 3.4 <sup>ac</sup>	29.73 $\pm$ 2.7 <sup>c</sup>	32.05 $\pm$ 2.7 <sup>ac</sup>
	Summer	31.66 $\pm$ 3.6 <sup>a</sup>	34.36 $\pm$ 3.4 <sup>ab</sup>	38.22 $\pm$ 2.7 <sup>b</sup>	30.50 $\pm$ 2.9 <sup>ac</sup>	29.34 $\pm$ 3.9 <sup>c</sup>	29.73 $\pm$ 1.9 <sup>c</sup>

<sup>a, b, c</sup> Duncan's multiple row test: different letters within the same season indicate significantly different results ( $p < 0.05$ )

\* Indicate significant different values between seasons

**Table 3:** Mean value ( $\pm$ SE) of blood serum TPX (umol/L) and TAC (mmol/L) in ewes during the peripartum period at spring (N=7) and summer (N=7).

Parameter	Season	Weeks before lambing			Weeks after lambing		
		-3	-2	-1	1	2	3
TPX	Spring	72.2 $\pm$ 6.3 <sup>a</sup>	77.7 $\pm$ 3.7 <sup>ab</sup>	84.5 $\pm$ 4.5 <sup>b*</sup>	82.8 $\pm$ 4.0 <sup>b*</sup>	84.2 $\pm$ 2.8 <sup>b</sup>	73.4 $\pm$ 4.6 <sup>a</sup>
	Summer	81.6 $\pm$ 7.7 <sup>a</sup>	86.6 $\pm$ 8.0 <sup>ab</sup>	97.4 $\pm$ 5.1 <sup>b</sup>	94.01 $\pm$ 3.4 <sup>b</sup>	84.8 $\pm$ 4.1 <sup>ab</sup>	81.7 $\pm$ 2.9 <sup>a</sup>
TAC	Spring	0.83 $\pm$ 0.08 <sup>a</sup>	0.87 $\pm$ 0.12 <sup>a</sup>	0.81 $\pm$ 0.13 <sup>a*</sup>	0.81 $\pm$ 0.13 <sup>a*</sup>	0.94 $\pm$ 0.12 <sup>b*</sup>	0.82 $\pm$ 0.13 <sup>a</sup>
	Summer	0.71 $\pm$ 0.09 <sup>a</sup>	0.63 $\pm$ 0.08 <sup>ab</sup>	0.55 $\pm$ 0.14 <sup>b</sup>	0.45 $\pm$ 0.11 <sup>b</sup>	0.50 $\pm$ 0.12 <sup>b</sup>	0.58 $\pm$ 0.18 <sup>ab</sup>
OSI	Spring	8.70 $\pm$ 1.2 <sup>a</sup>	8.93 $\pm$ 2.3 <sup>ab</sup>	10.38 $\pm$ 1.9 <sup>b*</sup>	10.22 $\pm$ 2.2 <sup>b*</sup>	8.93 $\pm$ 2.9 <sup>ab*</sup>	8.95 $\pm$ 1.5 <sup>a</sup>
	Summer	11.49 $\pm$ 2.1 <sup>a</sup>	13.75 $\pm$ 1.3 <sup>ab</sup>	17.70 $\pm$ 1.7 <sup>b</sup>	20.89 $\pm$ 2.0 <sup>b</sup>	16.96 $\pm$ 1.6 <sup>ab</sup>	14.09 $\pm$ 2.3 <sup>a</sup>

<sup>a, b, c</sup> Duncan's multiple row test: different letters within the same season indicate significantly different results ( $p < 0.05$ )

\* Indicate significant different values between seasons

## DISCUSSION

The study was designed to evaluate serum oxidant-antioxidant status and degree of oxidative stress index (OSI) in ewes during the peripartum period under two different ambient temperatures. To the best of our knowledge, there is a limited study on determination of the oxidative status and degree of oxidative stress using by TOS and OSI in ruminant (Abuelo *et al.*, 2013; Abdel-Ghani *et al.*, 2016).

The significant increase in rectal temperature may be attributed to the increased ambient temperature in the summer group ewes (Srikandakumar *et al.*, 2003).

Heat stressed mammals undergo changes in thermoregulatory mechanisms, which ultimately affect metabolic and hormonal responses as well as alter substrate utilization (Lees *et al.*, 2017). Therefore, a better understanding of how environmentally induced heat stress affects post-absorptive metabolism is a main condition to developing improvement strategies targeting the physiological and metabolic ramifications of heat-related illnesses (Baumgard and Rhoads, 2013; Sejian *et al.*, 2018).

In the current work, blood serum concentrations of total cholesterol, triglycerides, HDL and LDL were significantly increased before lambing, and then significantly decreased after lambing. These results are in accordance with those reported previously by Gradinski-Urbanc *et al.* (1986) and Nazifi *et al.* (2002). Gradinski-Urbanc *et al.* (1986) reported that during pregnancy and at the start of lactation, the serum triglyceride content was high, but fell during lactation. However, these results contradict those reported by Turk *et al.* (2015). This increase of cholesterol, triglycerides and HDL during late pregnancy may be linked to the diminished responsiveness of the target tissue to insulin during late pregnancy, predisposing the ewes to an increase of cholesterol, triglyceride and lipoprotein concentrations (Schlumbohm *et al.*, 1997). On the other hand, the significant decrease in serum lipids in the post-partum period could be related to the effect of increased lipolysis, which is hormonally regulated, and not an expression of energy deficiency (Holtenius and Hjort 1990).

In the current study, season had a significant effect on blood serum lipid profile. Blood serum concentrations of total cholesterol, triglycerides and

HDL were significantly decreased in summer when compared with those in spring. Turk *et al.* (2015) obtained similar results. Belhadj *et al.* (2016) demonstrated that chronic HS has a direct effect on lipid metabolism and liver enzymatic activities in ruminants. These changes are typical patterns during negative energy balance where dietary and tissue-derived nutrients are allocated to the mammary gland to support milk production (Turk *et al.*, 2008). In addition, these changes could be a consequence of the activity of maternal reproductive glands for steroid hormone synthesis (Turk 2009). This could indicate that animals during summer have mobilized more energy from body reserve to support milk production due to a high environmental temperature exposure (Das *et al.*, 2016).

In the last few years, oxidative stress markers have involved in the mechanisms of metabolic disorders, especially important in dairy animals, in which peripartum period imposes great demands on the body's homeostatic mechanisms (Gaál *et al.*, 2006; Ribeiro *et al.*, 2018). Concentration of TAC considers the cumulative action of all the antioxidants, rather than simply the sum of measurable antioxidants and accurately reflects the redox status (Ghiselli *et al.*, 2000).

In the current study, TPX concentration was increased at the first week before lambing and continued at a high level during the remaining period in spring and summer when compared with the baseline values. Whereas, concentrations of TAC in spring increased at the second week after lambing. A reverse trend was noticed in summer season, where the values were decreased at the first week before lambing and continued at low level during the first and second weeks after lambing. The increase of oxidants and decreased antioxidants around partum in this study lies with the results obtained previously by Turk *et al.* (2013; 2015). During the peripartum negative energy balance, a large amount of ROS is being generated in the course of NEFA oxidation in the liver resulting in raised lipoperoxidative processes and lowered antioxidant status (Bionaz *et al.*, 2007; Turk *et al.*, 2008).

Heat stress is suggested to be responsible of enhances ROS production and peroxide generation in livestock animals and induces oxidative stress, which can lead to cytotoxicity (Bernabucci *et al.*, 2002; Ganaie *et al.*, 2013; Nizar *et al.*, 2013). In the current study, concentrations of TPX are more enhanced during summer and the values were significantly higher than spring values during the first week before and after lambing. A reverse trend was noticed for TAC where summer values were significantly lower than spring values from the second week before lambing and continued during the remaining period. Our results agree with those previously reported for ruminants by (Ganaie *et al.*,

2013; Nizar *et al.*, 2013; Turk *et al.*, 2015). Di Trana *et al.* (2006) reported that seasonal rather than nutritional factors have a more pronounced effect on oxidative status markers. This suggested that the summer group had more prolonged negative energy balance and oxidative stress than the spring group (Marai *et al.*, 2007; Belhadj *et al.*, 2016).

The presence of OS could either be a consequence of an excessive production of TPX and/or a decrease in the body TAC, and therefore these parameters are strictly interdependent. In the current work, Pearson's correlation and regression analysis of TPX (as a representative value of the by-product of oxidative stress) with TAC (as a representative value of defense antioxidants) did not reveal significant correlation. The estimation of OSI provides an objective assessment of the relationship between oxidants and antioxidants, not seen by the determination of both components separately. An increase in TPX/ TAC ratio indicates a higher risk for OS because of an increase in ROS production, defensive antioxidant consumption, or both. The biomarker OSI was used in human medicine to evaluate oxidative stress in passive smoker infants and their mothers (Aycicek *et al.*, 2005). It has been used as a new tool to assess redox status in dairy cattle during the transition period (Abuelo *et al.*, 2013), and to investigate the changes in profile of the oxidant and antioxidant indicators throughout pregnancy in goats (Abdel-Ghani *et al.*, 2016). It used also as an oxidative stress marker of redox status in sheep as reference intervals for adult sheep (Oikonomidis *et al.*, 2017). Also, it used for estimation of redox status as a valid tool for providing a detailed picture about the oxidative status of healthy sheep and goats and to show any possible correlation of oxidative stress with daily milk yield or milk components (Cecchini *et al.*, 2018). Here in the present study, the OSI was used to assess the redox status during the peripartum period in sheep reared under the effect of HS under Upper Egypt conditions. When the oxidative stress index is studied in our work, statistical differences between parturition stages were found. Our data suggests that these ewes experienced their maximal oxidative challenge at the first week after parturition. In fact, there are no closed results from other studies to compare our results. However, our data clarify that the maximal oxidative stress occurred at the first week after parturition that was associated with an increase of pro-oxidants rather than reduction of antioxidants during spring. Instead, both the increase in pro-oxidants and the reduction of antioxidants are sharing in the oxidative stress in summer.

Can be concluded that, this is a trial study to evaluate the degree of OS in HS ewes during the peripartum period by detection of TPX, TAC levels and OSI value. These ewes experienced their

maximal oxidative challenge at the first week postpartum. Further studies to ascertain precisely how the changes in temperature affect the blood biochemistry of animals under different production and reproduction phase.

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## تأثير درجة حرارة الجو على التغيرات الدموية لمؤشر الإجهاد التأكسدي الكلي وصورة الدهون في النعاج حول الولادة

غاده عبد العظيم محمد ، هيام محمد المصطفى منزلي

E-mail: [dr\\_kada2012@yahoo.com](mailto:dr_kada2012@yahoo.com) Assiut University web-site: [www.aun.edu.eg](http://www.aun.edu.eg)

استخدم مؤشر الإجهاد التأكسدي (OSI) حديثاً كأداة لتقديم صورة واضحة عن حالة الأوكسدة والاختزال التأكسدي . تم الدراسة على ١٤ نعجة أوسيمي متعددة الولادات (١,٥-٢ سنة ، ٣٥-٤٠ كجم) في محطة البحوث الزراعية بأسبوط خلال موسم الربيع (مجموعة ضابطة N=7) وخلال الصيف (مجموعة الاجهاد الحراري N=7) . أخذت عينات دم من النعاج أسبوعياً على مدار الأسابيع الثلاثة الأخيرة قبل الولادة وأسبوعياً خلال الأسابيع الثلاثة الأولى بعد الولادة لفحص دهون المصل والمؤشرات الحيوية للإجهاد التأكسدي. أظهرت النتائج زيادة الكوليسترول الكلي والدهون الثلاثية والبروتينات الدهنية عالية ومنخفضة الكثافة ( $P < 0.05$ ) قبل الولادة ثم نقصت بعد الولادة ( $p < 0.05$ ). كان الكوليسترول والدهون الثلاثية والبروتينات الدهنية عالية الكثافة (HDL) أقل في الصيف مقارنة بالربيع ( $p < 0.05$ ). في فصلي الربيع والصيف ازداد إجمالي البيروكسيد الكلي (TPX) في الأسبوع الأول قبل وبعد الولادة ( $p < 0.05$ ) بالإضافة الى ان قيم الصيف كانت أعلى من قيم الربيع ( $p < 0.05$ ). في الربيع ازدادت السعة الكلية لمضادات الأوكسدة (TAC) في الأسبوع الثاني بعد الولادة ( $p < 0.05$ ) ، في حين لوحظ اتجاه عكسي في فصل الصيف حيث كانت قيم TAC الصيفية أقل من قيم الربيع ( $P < 0.05$ ). معامل بيرسون للارتباط والانحدار بين TPX و TAC لم يظهر اي دلالات معنوية . بينما ارتفعت قيم مؤشر الإجهاد التأكسدي ( $OSI = TPX / TAC$ ) x100 تدريجياً ( $p < 0.05$ ) في الصيف ووصلت إلى القيمة القصوى في الأسبوع الأول بعد الولادة ، ثم انخفضت بشكل مطرد في الأسبوع الثالث بعد الولادة . كانت قيم TPX الصيفية أعلى ( $p < 0.05$ ) من قيم الربيع . واخيرا ، هذه الدراسة هي الأولى لتقييم الاجهاد التأكسدي في النعاج المجهدة حرارياً خلال فترة الحمل عن طريق الكشف عن قيم TPX و TAC و OSI . كما أوضحت البيانات أن اقصى إجهاد التأكسدي حدث في الأسبوع الأول بعد الولادة والذي كان مرتبطاً بزيادة البيروكسيد عن نقص مضادات الأوكسدة خلال الربيع . ولكن، فإن كلا من زيادة البيروكسيد ونقص مضادات الأوكسدة تقاسمت السبب في الإجهاد التأكسدي في الصيف.