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The effect of oxidative stress on the reproductive performance in Hair Goats

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ABSTRACT: Small ruminants make better use of pastures with low grass yield and poor vegetation and therefore contribute significantly to the country's economy. In order to achieve maximum yield from these animals, it is necessary to optimize nutrition and minimize stress factors. Oxidative stress affects the normal functioning of many physiological systems including fertility. This study was carried out to determine oxidative stress with regard to effect of antioxidants on fertility in hair goat in October and May. In this study, oxidative stress index (OSI), total oxidation level (TOS) and total antioxidant (TAS) levels were investigated in 20 male and 100 female hair goats during the breeding season (October) and out-of-season (May). Females between the ages of 2-6 years and male hair goats aged 2-4 years were used in the study. The correlation between TAS, TOS, and OSI and fertility was evaluated according to gender and season. TAS, TOS and OSI values in female hair goats were 1.28 $\mu\text{mol/L}$, 1.34 $\mu\text{mol/L}$; and 11.02 $\mu\text{mol/L}$ in October and 7.13 $\mu\text{mol/L}$; 0.87 $\mu\text{mol/L}$ and 0.53 $\mu\text{mol/L}$ in May, respectively; as for male hair goats 1.32 $\mu\text{mol/L}$, 1.44 $\mu\text{mol/L}$ and 4.24 $\mu\text{mol/L}$ in October and 8.84 $\mu\text{mol/L}$; 0.32 $\mu\text{mol/L}$, and 0.63 $\mu\text{mol/L}$ in May, respectively. Seasonal differences between genders and calving and infertile groups were found ($P=0,001$). As a result, while the increase in TOS level causes a decrease in offspring yield in hair goats, it has been concluded that the increase in TAS level positively affects the offspring's productivity.

Keywords: Small ruminants; Hair goat; oxidative stress; antioxidant; reproductive performance

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INTRODUCTION

Detrimental effects of free radicals develop during the physiological digestion of food. These free radicals are called oxidants, and the damage to tissues is called oxidative stress. The substances that prevent tissue damage by binding these free radicals are called antioxidants (Güntürk, 2021). Oxidative stress has been defined an imbalance between oxidant and antioxidant at the cellular level (Karabulut and Gülay 2016). Normally, the living organism has components and antioxidant enzymes capable of neutralizing harmful free radicals and preventing oxidative stress (tissue damage) (Büyükoğlu, 2018). Some of these antioxidants are ascorbic acid (Vit. C), α -tocopherol (Vit E) and Ceruloplasmin (Dimri et al., 2010). In addition, elements such as zinc, copper, iron and selenium are essential components of certain substances such as hormones and enzymes and are endogenous antioxidants (Aruoma, 1998).

The oxidative stress is reported to induce pathological conditions for the cell and disturbs the comfort of farm animals, therein, affecting production (Mahmood et al., 2020). The high level of total oxidation also negatively affects reproductive performance. Antioxidant supplementation has the effect of reducing the incidence of uterine infection and increasing the pregnancy rate with fewer inseminations (Lopez-Gatius et al., 2006).

Oxidative stress is higher in high-producing dairy cows (Aydilek et al., 2014). Studies report negative effects of oxidative stress on the fertility of these animals (Abuelo et al., 2014; Aydilek et al., 2014; Celi, 2011). Some studies state opposite clear evidence of oxidative stress in fertility (Oyawoye et al., 2003). However, Abuelo (2016) reported a linear relationship between oxidative stress and reproductive performance, consequently, leading to economic losses (Abuelo et al., 2015; Aydilek et al., 2014; Latif et al., 2010). It is stated that oxidative stress index values increase in lactating cows and more inseminations are needed per pregnancy (Mirzad, 2018). Karasahin et al. (2021) reported that the quality of embryos obtained

from donors with high oxidative stress is poor and the pregnancy rate obtained from these embryos is low. Köse et al. (2013) reported that higher lamb yields were obtained from ewes that were given exogenous antioxidants. It is reported that the total oxidation level increases and the TAS level decrease in sheep infected with cyst hydatid (Mahmood et al., 2020) or pneumonia (Yüksek et al., 2018) and the severity of the infection is inversely proportional to the TAS and positively with TOS (Celi, 2010; Merhan et al., 2020).

In this study, the aim was to investigate the relationship between TAS, TOS and OSI values of the hair goat under the same management and nutritional conditions.

MATERIALS AND METHODS

The material of this study composed of 100 females, 2-6 years old and male goats, 2-4 years old, on a family-run business located in Aksaray province. The study was conducted following the approval by Experimental Animals Production and Research Center Ethics Committee, Selcuk University, Faculty of Veterinary Medicine (No: 2020/116; date: 09/12/2020). In the 2nd week of September, 100 females were selected out of 386 animals from a herd. In October, blood samples were taken from the animals 15 days prior to goat addition in order to avoid stress that may affect the yield of the offspring. Blood samples were collected again from the same animals in May before the starting of lactations. Blood samples were taken from jugular veins into anticoagulant-free tubes. The samples were transported to the laboratory in cold chain. Samples were centrifuged at +4°C, 3000 RPM for 10 min and sera were removed into two microtubes and stored at -20°C until further analysis. TAS (Catalog No: RL0017, Lot No: TZ19130A, Rel Assay Diagnostics, Türkiye) and TOS (Catalog No: RL0024, Lot No: TZ19130A, Rel Assay Diagnostics, Türkiye) levels were determined using an ELISA kit by colorimetric method (Table 1). Individual determination of oxidants (malondialdehyde, nitric oxide) and antioxidants concentrations (Vit. E, Se, superoxide dismutase, glutathione peroxidase and catalase) is

Table 1: TAS, TOS and OSI levels in the blood in October and May.

	Female		Male	
	October	May	October	May
	X \pm SH	X \pm SH	X \pm SH	X \pm SH
TAS (mmol/L)	1.28	1.34	1.32	1.44
TOS (μ mol/L)	11.02	7.13	4.24	8.84
OSI	0.87	0.53	0.32	0.63

time-consuming, costly and requires complex techniques. Therefore, the calculation of TAS, TOS and Oxidative stress index ($OSI=TOS/TAS$) reflects the oxidative/antioxidative situation and is more economical (Mahmood et al. 2020). Therefore, this approach was followed in the present study.

Statistical Analysis

Descriptive statistics for continuous variables were as follow; mean, standard deviation, minimum, and maximum values, while categorical variables are expressed as numbers and percentages. One-way Variance analysis was performed by comparing group averages for continuous variables. Duncan's multiple comparison tests was used to determine different groups following variance analysis. Pearson correlation coefficients were calculated in determining the relationship between these variables. A Chi-square test was performed to determine the relationship between groups and categorical variables. The statistical significance level was taken as 5% and the SPSS (ver.21) statistical package program was used for the calculations.

RESULTS

TAS, TOS, and OSI values obtained with regard to gender and season and the importance of the effect of these values on fertility were determined (Tables 2 - 3). Since animals were not hand mated, TAS, TOS and OSI parameters in males could not be linked comparatively to the pregnancy rates in females. The parameters examined were found to be significantly different ($p=0.001$) between the genders within each experimental period (Table 3 and Table 4).

The sex effect was found to be significant between October and May ($p\leq 0.05$) (Table 4 and Table 5). It was also determined that the relationship of TAS, TOS, and OSI values with fertility was also marked ($p\leq 0.05$) (Table 6). It was determined that goats with high TAS values had higher twin births (Table 7) and TAS was found to be important for multiple births ($p\leq 0.05$) (Table 7 and Table 8). In contrast, goats with high TOS values were found to be infertile, and goats with low TOS values had higher twinning rates (Table 8).

Table 2: Descriptive statistics for continuous (TAS, TOS, OSI) variables.

	N	Minimum	Maximum	Mean	Std. Deviation
TAS (mmol/L)	240	0.92	1.76	1.3195	0.2149
TOS (μ mol/L)	240	1.310	21.150	8.57708	3.397453
OSI	240	0.109	1.652	0.65304	0.264120

Table 3: Descriptive and comparative statistics for TAS, TOS and OSI Combinations

	Sex	October				May			
		Mean	Std. dev.	Std.err	p	Mean	Std. Dev.	Std.err	p
TAS (mmol/L)	Male	$\diamond 1.33 \#$	0.10	0.02	0,001	1.44 \diamond	0.21	0.05	0,001
	Female	1.28 #	0.07	0.01		1.34	0.12	0.01	
TOS (μ mol/L)	Male	$\diamond 4.18 \#$	1.58	0.35	0,001	8.84 \diamond	1.86	0.42	0,001
	Female	10.80 #	3.08	0.31		7.18	2.51	0.25	
OSI	Male	$\diamond 0.32 \#$	0.12	0.03	0,001	0.63 \diamond	0.15	0.03	0,001
	Female	0.85 #	0.24	0.02		0.53	0.16	0.02	

Season difference. \diamond Sex difference

Table 4: Descriptive and comparative statistics of TAS, TOS and OSI levels by sex.

		Mean	Std. dev	Min.	Max.	p
TAS (mmol/L)	Male	1.3828	0.17347	0.92	1.76	0.001
	Female	1.3068	0.10422	0.95	1.54	
	Total	1.3195	0.12149	0.92	1.76	
TOS (μ mol/L)	Male	6.51075	2.913405	2.130	11.440	0.001
	Female	8.99035	3.342076	1.310	21.150	
	Total	8.57708	3.397453	1.310	21.150	
OSI	Male	0.47135	0.209592	0.172	0.960	0.001
	Female	0.68938	0.259249	0.109	1.652	
	Total	0.65304	0.264120	0.109	1.652	

Table 5: Descriptive and comparative statistics for sex

		Mean	Std. dev	Min.	Max.	p
TAS (mmol/L)	Male	1.2860	0.07961	1.15	1.47	0.001
	Female	1.3529	0.14507	0.92	1.76	
	Total	1.3195	0.12149	0.92	1.76	
TOS (µmol/L)	Male	9.69850	3.800757	1.310	21.150	0.001
	Female	7.45567	2.490045	3.430	16.440	
	Total	8.57708	3.397453	1.310	21.150	
OSI	Male	0.75823	0.302611	0.109	1.652	0.001
	Female	0.54785	0.161914	0.279	1.089	
	Total	0.65304	0.264120	0.109	1.652	

Table 6: Descriptive and comparison of the effect of TAS, TOS and OSI levels on Birth Type.

		Mean	Std. dev	Min.	Max.	p
TAS (mmol/L)	Infertile	1.3472 ^a	0.13907	0.92	1.76	0.001
	Single	1.2512 ^b	0.05627	1.15	1.38	
	Twin	1.3626 ^a	0.04309	1.32	1.42	
	Total	1.3195	0.12149	0.92	1.76	
TOS (µmol/L)	Infertile	7.29354 ^b	3.219507	2.130	21.150	0.001
	Single	10.51247 ^a	2.888482	1.310	15.300	
	Twin	10.47043 ^a	1.938037	6.670	12.650	
	Total	8.57708	3.397453	1.310	21.150	
OSI	Infertile	0.53992 ^b	0.233254	0.172	1.652	0.001
	Single	0.83949 ^a	0.226975	0.109	1.330	
	Twin	0.76949 ^a	0.149374	0.498	0.958	
	Total	0.65304	0.264120	0.109	1.652	

a,b,c↓: Significance within columns

Table 7: Descriptive and comparative statistics of TAS, TOS and OSI levels for infertile, single and twin births.

		Mean.	Std. Dev.	Min.	Max.	p
TAS (mmol/L)	Single	1.2512 ^{ab}	0.05627	1.15	1.38	0.001
	Twin	1.3626 ^a	0.04309	1.32	1.42	
	Infertile	1.2725 ^b	0.09430	1.15	1.38	
TOS (µmol/L)	Single	10.51247 ^b	2.888482	1.310	15.300	0.001
	Twin	10.47043 ^b	1.938037	6.670	12.650	
	Infertile	18.01250 ^a	3.639454	14.450	21.150	
OSI	Single	0.83949 ^b	0.226975	0.109	1.330	0.001
	Twin	0.76949 ^b	0.149374	0.498	0.958	
	Infertile	1.42056 ^a	0.291571	1.047	1.652	

a,b,c↓: Significance within columns

Table 8: Descriptive and comparative statistics of TAS, TOS and OSI levels for infertile, single, twin and total fertile goats.

		Mean	Std. dev	Min.	Max.	p
TAS (mmol/L)	Total	451.84	73.111	332	568	0.001
	Single	1.2512 ^b	0.05627	1.15	1.38	
	Twin	1.3626 ^a	0.04309	1.32	1.42	
	Infertile	1.2725 ^b	0.09430	1.15	1.38	
	Total	1.2777	0.07195	1.15	1.42	
TOS (µmol/L)	Single	10.51247 ^b	2.888482	1.310	15.300	0.001
	Twin	10.47043 ^b	1.938037	6.670	12.650	
	Infertile	18.01250 ^a	3.639454	14.450	21.150	
	Total	10.80280	3.080898	1.310	21.150	
	Single	0.83949 ^b	0.226975	0.109	1.330	
OSI	Twin	0.76949 ^b	0.149374	0.498	0.958	0.001
	Infertile	1.42056 ^a	0.291571	1.047	1.652	
	Total	0.84663	0.244398	0.109	1.652	

a,b,c↓: Significance within columns

DISCUSSION

It is reported that there is a relationship between nutrition and OSI and that the viability of the embryo and the pregnancy rate are numerically higher in individuals with high total antioxidant levels (Karaşahin et al. 2021). As a matter of fact, in embryo production studies in humans and cows, they reported that higher rates of oocytes, fertilization and embryos were obtained in individuals with high TAS levels (Karaşahin et al. 2021, Oyawoye 2003). It is also determined that higher quality embryos are obtained from donors with high total antioxidant levels and the pregnancy rate is higher (Karaşahin et al. 2021).

This is the first study conducted to determine the TAS, TOS and OSI levels in Hair Goats. In the blood, TAS, TOS and OSI levels vary according to breed, nutrition, environment, and stress status. Mis et al. (2018) report that TAS, TOS and OSI levels are different in Norduz and Morkaraman breed sheep. The TAS rate in goats is higher than in sheep and the TOS rate is low and therefore the OSI rate is lower in goats than in sheep (Mis et al. 2018). Goats experience less stress than sheep and make better use of pasture. For this reason, in Turkey, goat breeders use exogenous antioxidants less than sheep breeders. The birth rate in goats within the same region and the same care/nutrition conditions is higher than in sheep (Yalçın 1990). It is thought that the level of antioxidants in goats is higher than in sheep and may cause a low infertility rate in goats. Therefore, Köse et al. (2013) administered the antioxidant Se + vit. E to sheep and obtained more lamb yield.

It is reported that the level of TOS increases in infected sheep, the level of TAS and fertility are adversely affected (Mahmood et al. 2020, Alaçam 1994). In the case of infection, since fertility (Küplülü 1994) and TAS levels (Mahmood 2020, Merhan 2020) decrease, it is likely that there is a relationship between the level of TAS and the fertility. The increase in offspring productivity in sheep given exogenous antioxidants supports a relationship between TAS level and fertility (Köse et al. 2013). In the present study, it was noted that the offspring yield (single and multiple births) was higher in goats with high TAS levels and the TAS levels of infertile goats were low. The results obtained appear to be in line with the results reported for cows (Elshahawy 2017, Mirzad et al. 2018, Karaşahin et al. 2021) and sheep (Köse et al. 2013, Mis et al. 2018). It is stated that the number of inseminations per pregnancy increases in lactating cows with higher oxidative stress, namely, the infertility rate increases (Mirzad et al. 2018).

The antioxidant ratio in goat's milk was higher than in sheep's milk. These antioxidants are also present in the circulation and can cause the performance of other systems to increase (reproduction) (Akşit et al. 2014, Eğritağ et al. 2018).

It has been reported that the level of TAS in 1st week of postpartum in dairy cows decreases compared to the prenatal period, but increases in the following weeks, which is an advantage for fertility (Elshahawy 2017, Luan et al. 2021). In the present study, it was determined that the offspring yield was high in goats with high TAS levels. No other studies have yet found

the relationship between the level of serum TAS, TOS and OSI and offspring yield in Hair Goats. The data obtained in this study is the first brought to the attention of scientists.

Oxidative stress varies depending on breeds, seasons, and whether animals are housed indoors or outdoors (Mirzad 2018). OSI was lower in May due to the fact that the animals in the present study were almost completely out and they started to go out to new pastures (as of April) and started to consume fresh grass. In October (as of June), they cannot get antioxidant substances with feed because they have been fed with dry grass in pasture for a long time and the level of OSI is higher.

CONCLUSIONS

In conclusion, the fertility of hair goats (single or multiple births) will be higher in goats with a high total antioxidant content suggesting lower oxidative stress, however, it warrants further investigation.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

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