

## **Seasonal Variations in Glutathione Peroxidase Activity of Erythrocyte in Sheep Grazed on Pasture in the Region of Kars**

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### **Summary**

Seasonal variations in erythrocyte glutathione peroxidase activity (GSHPx) in 15-day-old lambs were studied in 6 different districts in the around the city of Kars in Turkey. The seasonal variation in erythrocyte GSHPx activity was studied in 800 5-ml blood samples taken by jugular venapuncture from 15-day-old lambs during the lambing periods in this geographical area (spring-summer) and autumn-winter period. The blood samples were analysed for the activity of GSHPx. The overall mean values in autumn-winter ( $123.87 \pm 14.93$  IU/g Hb) were higher than in spring-summer ( $62.26 \pm 6.93$  IU/gHb). In autumn-winter, in none of districts were found with a mean GSHPx activity lower than 60 IU/g Hb (deficient); three district showed mean levels between 60 and 130 IU/g Hb (marginal); and in three of the districts were the mean activities higher than 130 IU/g Hb (adequate). Likewise, in spring-summer two districts had deficient levels, four were marginal and in none of these could the mean GSHPx activities be considered adequate. In two of the districts the mean activities changed from deficient to marginal, and in four of the districts the mean activities changed from marginal to adequate, between the two seasons.

It may be concluded that lambs born in spring-summer in this area are at a higher risk of Selenium (Se)-deficiency related disorders. Programmes designed to prevent Se deficiency should take account of these seasonal variations, increasing the Se intake in the months of greater risk.

**Keywords:** Lamb, Seasonal variation, Pasture, Erythrocyte, Glutathione Peroxidase (GSHPx) activity.

### **Kars ve Yöresi Meralarında Otlayan Koyunların Eritrosit Glutasyon Peroksidaz Aktivitelerinin Mevsimsel Değişimleri**

#### **Özet**

Bu çalışmada, Kars yöresinin 6 farklı bölgesindeki 15 günlük kuzuların eritrosit glutasyon peroksidaz (GSHPx) aktivitelerinin mevsimsel değişimi araştırıldı. Eritrosit GSHPx aktivitelerini belirleyebilmek için sonbahar-kış, ilkbahar-yaz dönemlerinde 800 tane 15 günlük kuzunun kan örnekleri V. jugularis'ten alındı. GSHPx aktiviteleri sonbahar-kış döneminde ( $123.87 \pm 14.93$  IU/g Hb) ilkbahar-yaz dönemine ( $62.26 \pm 6.93$ ) göre daha yüksek bulundu. Sonbahar-kış döneminde belirlenen bölgelerin üçünde GSHPx aktiviteleri 60-130 IU/g Hb (normal düzey) arasında bulunurken, diğer üçünde ise 130 IU/g Hb'den (yeterli düzey) daha fazla tespit edildi. İlkbahar-yaz döneminde ise; GSHPx aktiviteleri bölgelerin ikisinde yetersiz, dördünde normal düzeylerde saptandı. Ancak, bu dönemde hiçbir bölgede GSHPx aktivitesi yeterli düzeylerde belirlenemedi. Belirlenen bu mevsimsel dönemler arasında GSHPx aktiviteleri bölgelerin ikisinde yetersizden normale, dördünde de normalden yeterliye doğru değişiklik gösterdi.

Bu bilgilerden, bölgelerde ilkbahar-yaz döneminde kuzuların selenyum (Se) noksanlığından kaynaklanabilecek hastalıklara karşı daha fazla risk altında olabilecekleri sonucuna varıldı. Bu mevsimsel değişiklikler göz önüne alınarak, Se noksanlığına karşı planlanan koruyucu programlarda riskli bölgelerdeki annelere verilen Se miktarının artırılması gerektiği kanaatine varıldı.

**Anahtar sözcükler:** Kuzu, Mevsimsel değişim, Çayır, Eritrosit, Glutasyon Peroksidaz (GSHPx) aktivitesi.

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## INTRODUCTION

Kars is a city in the north of Eastern Anatolia in Turkey. The climate is continental, characterized by snowy and cold winters and autumns, and by rainy springs and summers. Climatic and pedological features support the development of meadow-pasture hay, which is the basis for sheep production in the region. Over the duration of this study, the highest temperatures were registered during the summer, from June to September, with mean values from 14 to 25°C, and the lowest were recorded in winter, with mean values from -30 to 5°C, when the rainfall was 730 mm. The highest monthly values were 67-86 mm (autumn and spring) and the lowest 34 mm (summer).

Feeding practices in the region of Kars are based on grazing for most of the year, with some cereal grains used in seasons where there is less pasture available. GSHPx (EC 1.11.1.9) has been used as an indicator of selenium (Se) status in animals<sup>1</sup>, due to the high correlation found between dietary Se and the activity of this enzyme in plasma, red blood cells (RBC) and various organs<sup>2</sup>. The aim of the present study was to evaluate blood GSHPx activity in sheep flocks in the region around Kars city and to identify those seasons with the highest risk of Se deficiency, allowing recommendations to be made on the use of supplements containing Se.

## MATERIAL and MET

The study used 6 different locations close to the city of Kars. The seasonal variation in erythrocyte GSHPx activity was studied in 800 5-ml blood samples taken by jugular venapuncture from 15-day-old lambs during the two usual lambing periods in this geographical area (autumn-winter and spring-summer). Anticoagulated blood was separated into plasma and RBC by centrifugation (at 1500g for 15 min). The RBC fractions were washed three times in cold isotonic saline. The RBC samples were hemolyzed with a nine-fold volume of redistilled water. Hemolyzed RBC were stored at -30°C for <3 mo pending measurement of GSHPx activity. The GSHPx activity of the RBC was measured spectrophotometrically at 37°C and 412 nm according to Lawrence and Burk<sup>3</sup>. The hemoglobin concentration in the lysed erythrocytes was determined by the cyanmethemoglobin method<sup>4</sup>.

**Statistical Analysis:** All results were expressed as mean  $\pm$  standard error (SE). Differences between the two seasons were compared using Mann Whitney U test, with the programme SPSS software (version 9.5). The significant level was  $p < 0.05$ .

## RESULTS

The mean ( $\pm$ SE) blood GSHPx activities in sheep from the 6 different locations and the significance level of the statistical comparison between seasons are shown in Table I. The overall mean values in autumn-winter ( $123.87 \pm 14.93$  IU/g Hb) were higher than in spring-summer ( $62.26 \pm 6.93$  IU/g Hb). In autumn-winter, in none of the districts were found to have a mean GSHPx activity lower than 60 IU/g Hb (deficient); three districts showed mean levels between 60 and 130 IU/g Hb (marginal); and in three of the districts were the mean activities higher than 130 IU/g Hb (adequate). Likewise, in spring-summer two districts had deficient levels, four were marginal and in none of these could the mean GSHPx activities be considered adequate. In two of the districts the mean activities changed from deficient to marginal and in four of the districts the mean activities changed from marginal to adequate, between the two seasons.

## DISCUSSION

The dependence of serum Se concentration and blood GSHPx activity upon the Se content of pasture has been reported in sheep and cattle<sup>5,6</sup>. Gardiner<sup>7</sup> described an increase in the number of clinical cases of Se deficiency in spring, attributing this to the high presence of leguminous species in the pasture. A decrease in GSHPx activity in lambs in spring, as a result of a low Se content in pasture, was reported by Paynter et al<sup>8</sup>. Similarly, in our previous study<sup>9</sup>, we observed that a decrease in GSHPx activity in lambs in spring was a result of a low Se content in pasture.

Wheatley and Beck<sup>10</sup>, in studies performed in adult sheep, reported seasonal variations in blood GSHPx activity, with the highest values from March to June and the lowest from August to November, as a result of their experimental animals being housed in winter and maintained on a diet based on concentrates but allowed free grazing in spring. In our study, the seasonal variation in GSHPx activity was examined in 2-week-old lambs. As in adult sheep, GSHPx activity in newborn lambs is directly related to Se availability,

although in this case to its availability to the ewes during pregnancy<sup>11</sup>. The differences between our results and the findings of Wheatley and Beck<sup>10</sup> may be explained by the special features of the climate in the region of Kars city. Rainfall and temperature allow an optimum growth of grass from April to October. Therefore, the lambs born in spring-summer are from pregnancies in months when feeding is based only upon grazing. However, lambs born in autumn-winter come from ewes gestating during the summer when grazing is insufficient to cover the nutritional requirements of the animals and when supplementation with cereal grains is necessary. In addition, summer pasture, although less abundant and digestible, contains more Se than spring pasture, due to the higher content in dry matter and the lower rainfall<sup>7,12</sup>.

Under our husbandry conditions, GSHPx assessment is a useful means of determining flocks with deficient Se levels. In such districts, Se supplementation is essential and is given by treating the ewes with a slow release Se compound<sup>13</sup>, to enrich the diet during the last third of gestation, or by injecting sodium selenite to newborn lambs<sup>14</sup>. However, in districts with low GSHPx activity, the repercussions of absolute Se deficiency were higher than might be expected, with cases of nutritional low myodystrophy being reported. This may be due to low Se levels in the tissues of diseased lambs<sup>15</sup>.

From this experiment, it can be concluded that lambs born in spring-summer in this area are at a higher risk of Se-deficiency related disorders. Programmes designed to prevent Se deficiency should take account of these seasonal variations, increasing the Se intake in the months of greater risk.

## REFERENCES

- 1 **Anderson PH, Berrett S, Patterson DSP:** The biological selenium status of livestock in Britain as indicated by sheep erythrocyte glutathione peroxidase activity. *Vet Rec*, 104, 235-238, 1979.
- 2 **Smith PJ, Tappel AL, Chow CK:** Glutathione peroxidase as a function of dietary selenomethionine. *Nature*, 247, 392-393, 1974.
- 3 **Lawrence RA, Burk RF:** Glutathione-peroxidase activity in selenium-deficient rat liver. *Biochem Biophys Res Comm*, 71, 952, 1976.
- 4 **Canan RK:** Hemoglobin (as cyanmethemoglobin) in blood. *Clin Chem*, 4, 246, 1958.
- 5 **Mee JF, O'Farrell KJ, Rogers PAM:** Baseline survey of blood trace-elements status of 50 dairy herds in the South of Ireland in the spring and autumn of 1991. *Irish Vet J*, 47, 115-122, 1994.
- 6 **Counotte GH, Hartmans J:** Relation between selenium content and glutathione peroxidase activity in blood of cattle. *Vet Q*, 11, 155-160, 1989.
- 7 **Gardiner MR:** Selenium in animal nutrition. *Outl Agricul*, 6, 19-28, 1969.
- 8 **Paynter DI, Anderson JW, McDonald JW:** Glutathione peroxidase and selenium in sheep. II. The relationship between glutathione peroxidase and selenium-response unthriftness in Merino lambs. *Aust J Agricul Res*, 30, 703-709, 1979.
- 9 **Kamiloğlu NN, Beytut E, Gey H:** Relationship between selenium levels in soil and meadow hay and glutathione peroxidase activity in erythrocytes of offsprings of which dams grazed on pasture. *Kafkas Univ Vet Med J*, 11(1): 65-68, 2005.
- 10 **Wheatley LE, Beck NF:** The influence of season and husbandry on the selenium status of sheep in a deficient area. *Br Vet J*, 144, 246-252, 1988.
- 11 **Hanliri A, Olson WG, Johnson DW, Kessabi M:** Evaluation of biochemical evidence of congenital nutritional myopathy in two-week prepartum fetuses from selenium-deficient ewes. *Am J Vet Res*, 51, 1112-1115, 1990
- 12 **Hunter RA, Peter DW, Quinn MP, Siebert BD:** Intake of selenium and other nutrients in relation to selenium status and productivity of grazing sheep. *Aust J Agricul Res*, 33, 637-647, 1982.
- 13 **Andres S, Mane MC, Sanchez J, Barrera R, Zaragoza C, Jimenez A:** Response to barium selenate supplementation in sheep kept at pasture in the Mediterranean area. *Vet Res*, 28: 539-545, 1997c.
- 14 **Andres S, Jimenez A, Mane MC:** Changes in GSHPx and muscle enzyme activities in lambs with nutritional myodegeneration following a single treatment with sodium selenite. *Small Rum Res*, 23, 183-186, 1996.
- 15 **Beytut E, Karataş F, Beytut E:** Lambs with white muscle disease and selenium content of soil and meadow hay in the region of Kars, *Turkey Vet J*, 163, 214-217, 2002.

**Table 1.** Mean values (X±SE) of GSHPx (IU/gHb) in lambs in the two lambing periods and GSHPx status according to the origin of the samples (M: marginal, D: deficient, A: adequate)

**Tablo 1.** GSHPx'in örneklerin orijinine göre durumu ve sonbahar-kış dönemi ile kuzulama dönemindeki kuzuların GSHPx aktivitelerinin (IU/gHb) ortalama değerleri (M: sınırda, D: yetersiz, A: yeterli)

Origin of Samples	Spring-Summer	Autumn-Winter	P
AKYAKA	60.72±9.85 <sup>M</sup>	145.62±24.12 <sup>A**</sup>	0.001
MERKEZ	24.18±8.72 <sup>D</sup>	96.15±14.28 <sup>M**</sup>	0.001
DIGOR	45.13±4.25 <sup>D</sup>	96.70±14.41 <sup>M*</sup>	0.01
ARPACAY	86.14±6.74 <sup>M</sup>	178.95±12.86 <sup>A**</sup>	0.001
SUSUZ	64.23±3.25 <sup>M</sup>	87.55±10.20 <sup>A*</sup>	0.01
SELİM	93.14±9.12 <sup>M</sup>	138.24±12.90 <sup>A*</sup>	0.01
<b>TOTAL</b>	<b>62.26±6.99</b>	<b>123.87±14.93</b>	

\* p<0.01, \*\*p<0.001