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### Determination of Serum Leptin Levels in Cattle, Sheep, Goats and Buffaloes in Burdur Province in Türkiye by ELISA Method

# Bayram Kudret Karaayvaz<sup>1,a</sup>, Reyda Kıyıcı<sup>1,b</sup>, Yahya Öztürk<sup>2,c</sup>, İbrahim Bağcı<sup>1,d</sup>, Taha Gürsoy<sup>3,e</sup>, Durmuş Kahraman<sup>1,f</sup>, Hasan Altan Akkan<sup>4,g</sup>, Nuri Mamak<sup>4,h</sup>, İbrahim Taşal<sup>5,i</sup>, Mehmet Karaca<sup>4,j</sup>

<sup>1</sup>Burdur Food Agriculture and Livestock Vocational School, Burdur Mehmet Akif Ersoy University, Burdur, Türkiye

<sup>2</sup>Department of Zootechni, Faculty of Veterinary Medicine, Burdur Mehmet Akif Ersoy University, Burdur, Türkiye

<sup>3</sup>Rectorate, Burdur Mehmet Akif Ersoy University, Burdur, Türkiye

<sup>4</sup>Department of Internal Medicine, Faculty of Veterinary Medicine, Burdur Mehmet Akif Ersoy University, Burdur, Türkiye

<sup>5</sup>Department of Obstetric and Gynecology, Faculty of Veterinary Medicine, Burdur Mehmet Akif Ersoy University, Burdur, Türkiye

\*Corresponding author

Research Article	ABSTRACT				
History	Leptin is a hormone that provides the central and/or peripheral regulation of food intake, energy expenditure, whole body energy balance, fertility and immune functions by creating a feeling of satiety in animals and humans.				
	This study was carried out to determine serum leptin values for some ruminant species and breeds that were				
Received: 23/08/2022	healthy and had at least one birth. In the study, 15 Simental and 15 Holstein cows, 15 Italian buffalo, 15 Pırlak				
Accepted: 07/10/2022	sheep, 15 Honamlı and 15 Turkish hair goats were used as material. Serum leptin concentrations were measured				
	with the BT LAB Bovine Leptin ELISA kit. Serum leptin levels were determined as 2.78±0.22 ng/ml in Simmental				
	cows, 2.87±0.11 ng/ml in Holstein cows, 3.6±0.48 ng/ml in Pırlak sheep, 3.38±0.76 ng/ml in Honamlı goats,				
	5.48±0.92 ng/ml in Hair goats and 2.50±0.17 ng/ml in Italian buffaloes. As a result, it was determined that serum				
	leptin levels obtained from different ruminant species that were healthy and gave at least one birth in Burdur				
	province were within the reference values reported for ruminants.				
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4.0 International License	Keywords: Honamli goat, Italian buffalo, Leptin, Ruminant				
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#### Introduction

Leptin is a word derived from the Greek word leptos, meaning thin and weak (Comba, 2014). Leptin is a hormone primarily secreted from adipose tissue in mammals. Although the main production site of leptin is adipose tissue, liver, stomach, rumen abomasum and/or duodenum (Younekura et al., 2002) breast tissue (Chelikani et al., 2003) are also reported to be secreted from bone marrow, intestine, ovary, testicles, skeletal muscle and placenta. Leptin was initially defined to be related to satiety and energy balance, but later it was found to be an antiobesity factor with feedback effect from adipocytes to the hypothalamus. Decreased leptin levels in serum and adipose tissue indicate that there is an energy deficit in the brain. It has also been determined that leptin plays an active role in adapting animals to malnutrition. In addition, it has been reported that leptin has both central and peripheral effects on thermogenesis, carbohydrate-fat storage and metabolism, regulation of cardiovascular and immune functions, as well as its effect on feed consumption and energy expenditure (Comba et al., 2015).

It has been reported that leptin is an effective lipostatic factor against excessive fat accumulation and may be the main factor helping animals adapt to periods of malnutrition. Decreased leptin levels in serum and adipose tissue indicate that there is an energy deficit in the brain. Leptin hormone is responsible for lowering the intracellular lipid level in beta cells of skeletal muscles, liver and pancreas by interacting with insulin (Klaus, 2004). It has been suggested that body weight is regulated by the hormone leptin (Baile et al., 2000). Insulin is considered a satiety hormone because it is thought to mediate the synthesis and secretion of leptin hormone (Schoeller et al., 1997). In addition, the leptin plays an important role in the adaptation of animals to malnutrition. In malnourished animals, rapid decrease in plasma leptin levels has been observed (Chilliard et al., 2000; Chilliard et al., 2001).

Between 1994-2001, studies on the physiology of leptin gained momentum in rodents and humans. However, studies have been reported to progress more slowly in ruminant species due to difficulties in developing specific tools to study leptin gene expression and plasma leptin variations in ruminants (Chilliard et al., 2001).

In the following years, Thomas et al. (2002) conducted studies in Angus, Brangus and Brahman bulls, Zieba et al. (2004) Brahman hybrid cattle and Delavaud et al. (2002) determined serum leptin levels in adult Holstein cattle. In a study in Turkey, Guzel et al. (2012) determined serum leptin levels in Brown Swiss (2.35±0.34 ng/ml), Holstein bulls (3.55±0.53 ng/ml), Avesi sheep (2.16±0.29 ng/ml), saanen (2.23±0.46 ng/ml) and hair goats (0.94±0.10ng/ml) by the radio immune assay (RIA) method. In the early 2000s, studies investigating leptin levels serologically were more common, but in the following years more genetic studies began to gain importance (Ferchichi et al., 2018; Kaplan, 2018; Abbas et al., 2019; Avondo et al., 2019; Das et al.,2019; Ibrahim et al., 2020; Mahrous et al., 2020; Sedykh et al., 2020).

The aim of this article is to determine the serum leptin level in various ruminant species and breeds that are healthy and have given birth at least once in Burdur province.

#### **Material and Methods**

Blood from Italian buffaloes was taken from the vena subcutanea abdominalis. Before blood collection, light sedation was provided with xylazine in buffaloes. Blood was drawn from vena jugularis from other species and breeds. After separating the serum samples, they were stored at -200C until analysis. Measurement of leptin hormone levels in serum samples was measured with BT LAB Bovine Leptin ELISA kit in accordance with the procedure in Diagen laboratory (Diagen Biyoteknolojik Sistemler AS, Ankara).

Descriptive statistical analyzes of the data were performed using the Jamovi statistical package program. Study was approved by the Local Ethical Committee on Animal Research of Burdur Mehmet Akif Ersoy University, Turkey (No: 789).

#### Results

Serum leptin levels in the animals used in the study were determined as 2.78±0.22 ng/ml in Simmental, 2.87±0.11 ng/ml in Holstein, 3.6±0.48 ng/ml in Pırlak

sheep,  $3.38\pm0.76$  ng/ml in Honamlı goat,  $5.48\pm0.92$  ng/ml in Hair goat and  $2.50\pm0.17$  ng/ml in Italian buffalo. Data were reported as mean  $\pm$  SEM (Table 1).

Table 1. Serum leptin levels in various ruminant species
by species/breed, age, and body weight.

Spacios / Prood	Leptin	Age	Body weight
species/breed	(ng/ml)	(month)	(kg)
Simmental	2.78±0.22	36-48	500-700
Holstein	2.87±0.11	36-48	500-700
Italian buffalo	2.50±0.17	36-132	550-700
Hair goat	5.48±0.92	96-120	50-70
Honamli goat	3.38±0.76	24-72	65-70
Pirlak sheep	3.6±0.48	24-60	50-70

#### Discussion

In ruminants, leptin level is also effective on nutritional and physiological factors. In a study conducted in different sheep breeds, it was determined that there is a relationship between serum leptin level and lipid profile. In malnourished animals, rapid decrease in plasma leptin level, reproductive cessation and decrease in thyroid activity, energy expenditure and protein synthesis were observed. In ruminants, malnutrition helps metabolic adaptation against malnutrition by decreasing leptin and increasing cortisol. When adequate nutrition is passed, insulin secretion is stimulated, and current high blood cortisol levels stimulate leptin secretion. After reaching the high blood leptin level, blood insulin and cortisol levels return to normal in order to restore the hemostatic balance. Therefore, cortisol-insulin-leptin interactions play an important role in malnutrition and adaptation to the normal feeding process in ruminants. In metabolism, leptin basically causes a decrease in food intake and an increase in energy expenditure. It is stated that leptin treatment with different dose applications in animals leads to loss of food intake, appetite, and body weight, as well as loss of fat stores and improvement in energy metabolism (Comba et al., 2015). In a study conducted in obese mice, it was determined that external administration of leptin helped rapid weight loss and improvement of metabolic abnormalities in these animals (Topal, 2004).

Thomas et al. (2002) determined serum leptin levels as 3.2 ng/ml in Angus cattle, 1.9 ng/ml in Brangus and 1.8 ng/ml in Brahman bulls. Leptin levels determined as 2.0-6.5 ng/ml in Brahman hybrid cows (Zieba et al., 2004), 1.3-2.8 ng/ml in adult Holstein cows (Delavaud et al., 2002), 4.97-5.28 ng/ml in periparturient sheep (McFadin et al., 2002), 3.22-5.2 ng/ml in Saanen goats (Magistrelli et al., 2011). In Turkey, Comba (2014) determined serum leptin levels as 6.5±3.35 ng/ml in Morkaraman sheep, 12.08±0.82 ng/ml in Karagül breed, 5.04±2.61 ng/ml in Norduz and 16.68±6.78 ng/ml in Tahirova breed. Comba (2014) found high levels of serum leptin levels and body weights in Tahirova breed and determined statistical significance. And it has been reported that the high serum leptin level in thin-tailed sheep is not related to the tail fat ratio. Avcı et al. (2013)

investigated plasma leptin levels by adding 250 ppm zinc to the diets of Akkaraman and Merino sheep. In the study, it was determined that the plasma leptin level was higher in the zinc-administered group than in the control group, but there was no statistical significance between the groups. They stated that this situation may be due to the fact that the zinc level in the basal ration is within the normal limits that should be taken daily. In the study, serum leptin levels in Akkaraman and Merino sheep were determined as 4.44±0.48 ng/ml and 4.03±0.64 ng/ml in the control groups, respectively, and 5.68±0.5 ng/ml and 5.84±0.92 ng/ml in the zinc added group.

Comba (2014) determined leptin levels as 12.08±0.82 ng/ml in Karagül breed, 6.5±3.35 in Morkaraman breed, 5.04±2.61 in Norduz breed and 16.68±6.78 ng/ml in Tahirova breed in a study conducted in sheep in Van region.

In this study, serum leptin levels were determined as  $2.78\pm0.22$  ng/ml in Simmental cows,  $2.87\pm0.11$  ng/ml in Holstein cows,  $3.6\pm0.48$  ng/ml in Pırlak sheep,  $3.38\pm0.76$  ng/ml in Honamlı goats,  $5.48\pm0.92$  ng/ml in Hair goats and  $2.50\pm0.17$  ng/ml in Italian buffaloes.

The leptin results obtained in this study were similar to the leptin level determined by Delavaud et al (2002) in the Holstein breed, when compared with the results of the researchers reported above. It is close to the leptin value determined by Thomas et al (2002) in Angus breed but higher than that in Brangus and Brahman breed bulls. In this study, it was determined that the leptin values detected in the thin-tailed Pırlak sheep were lower than the leptin values determined in the sheep by McFadin et al (2002). In addition, it was found to be lower than the leptin levels detected by Avcı et al (2013) in Akkaraman and Merinos and by Comba (2014) in Karagül, Morkaraman, Norduz and Tahirova breeds. On the other hand, it was found to be higher than the leptin level reported by Blache et al (2000) in rams.

The leptin levels in Honamlı goats and Hair goats determined in this study were found to be similar to the levels reported by Magistrelli et al. (2011) in Saanen goats.

Tajik and Nazifi (2011) determined serum leptin level as 3.5±0.40 ng/ml in male buffaloes and 7.11±0.49 ng/ml in female buffaloes in Iran. In this study, serum leptin level in female buffaloes was determined as 2.50±0.17 ng/ml.

#### Conclusion

Considering all these results, it was concluded that the differences in serum leptin levels between species and breeds may vary depending on the analysis method used, geography, feeding regime of animals, breed, age, and gender.

As a result, it was determined that serum leptin levels obtained from different ruminant species that were healthy and gave at least one birth in Burdur province were within the reference values reported for ruminants.

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#### **Ethical Statement**

This study does not present any ethical concerns.

#### **Conflict of interest**

The authors declared that there is no conflict of interest.

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