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Blood leptin, insulin and glucose concentrations in hair sheep raised in a tropical climate

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ABSTRACT

The aims of this study were to determine the effects of body condition score (BCS), breed and dietary supplementation on the concentrations of leptin, insulin and glucose found in the blood obtained from hair sheep during the breeding season. A further aim was to investigate the possible association of fertility and prolificacy with these blood metabolites, BCS and body weight (BW). All ewes were grazed in paddocks with *ad libitum* access to mineral salts and water. A total of 96 ewes were divided into two groups according to breed and treatment: Santa Ines (supplemented or unsupplemented) (24 × 24) and Morada Nova (supplemented or unsupplemented) (24 × 24). Blood samples, and BW and BCS information were collected during the breeding season. The statistical analyses were performed using the program PROC GLM from the SAS software. The leptin concentrations in hair sheep raised in a tropical climate were low. Little effect of breed, treatment or sample collection was found for blood insulin concentrations ($p < 0.05$), although the values were higher in the supplemented groups from both breeds. Significant differences were observed in glucose concentrations between the breeds in the same sample collections, with the higher concentrations being found in the Santa Ines sheep ($p < 0.05$). The BCS for ewes that were not pregnant showed the highest correlation with leptin, insulin and glucose concentrations ($r = 0.53$, 0.52 and 0.43, respectively). In the Morada Nova supplemented sheep (prolificacy: 1.45), there were verified correlations between BCS and BW, BCS and insulin concentration, and also between insulin and leptin concentrations. The present study shows that the Morada Nova breed has a higher reproductive efficiency than the Santa Ines breed. In conclusion, leptin was present in low concentrations in hair sheep and did not influence the reproductive processes in these animals. The dietary supplementation positively affected blood leptin, insulin and glucose concentrations in these breeds of hair sheep, but there was no major effect on the reproductive processes.

1. Introduction

According to data collected by the Brazilian Institute for Geography and Statistics (IBGE) in 2011, there are approximately 9,566,968 sheep in northeastern Brazil. They represent 56.9% of the Brazilian sheep population, and the predominant genetic group is the hair sheep (60–70%). Of the various breeds of hair sheep, the Morada Nova and Santa Ines breeds are the most widespread in northeastern Brazil because of their adaptation to the climatic conditions in this region. Sheep husbandry has traditionally been considered to be of low commercial value in Brazil, and it is generally practiced by the informal economy (is the income generated by economic agents that operate informally)

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(Paiva et al., 2005). In the Northeast and Central West of Brazil, regions characterized by long dry periods, hair sheep represent an important source of protein for the local populations. More recently in these regions, market diversification has increasingly allowed sheep husbandry to be considered as a viable and lucrative occupation.

Despite extensive livestock methods and the semiarid climate, Morada Nova ewes show good reproductive performance and have a better litter size at lambing than other sheep breeds in the Northeast of Brazil (Selaive-Villarroel and Fernandes, 2000). Nevertheless, there have been few studies on hormonal metabolism and the influence of nutrition in the reproductive processes in hair sheep raised in the Northeast of Brazil.

Several studies have related the reproductive performance of flocks to the nutritional status of individuals (Boland and Lonergan, 2003; Kiyma et al., 2004; Kiker et al., 2007), but how this occurs remains unclear. However, it is known that nutrition might influence ovarian function, as it modulates the secretion of hormones that control the reproductive processes in females (Chilliard et al., 2005). Previous studies have shown that the number of follicles and the ovulation rate are directly influenced by nutrition. They can be assessed *via* changes in the concentrations of certain metabolites in the bloodstream, such as insulin, leptin, glucose and growth hormone (GH) (Davies-Morel and Beck, 2003; Ramin and Majdani, 2005; Scaramuzzi et al., 2006).

Insulin is a small globular protein of about 5.7 kDa that differs between species. It is synthesized and secreted from the pancreatic β-cells of the islet of Langerhans in all species (González and Silva, 2006). In ruminants, due to microbial activity in the rumen, little or no dietary carbohydrate is absorbed as hexose sugar in the small intestine (Mcniven, 1984). For this reason, volatile fatty acids (propionate and butyrate) are more potent than glucose for stimulating insulin secretion (Pineda and Dooley, 2003; Brockman, 2005; González and Silva, 2006).

Another hormone, leptin, a 16 kDa protein product of the obese (*ob*) gene is secreted by adipocytes. In normal animals, leptin serves as a metabolic signal to the reproductive system, informing it that sufficient fat stores are available to meet the caloric demands of reproduction (Barash et al., 1996). Thus, leptin expression and secretion are correlated with body condition, physiological status (puberty, pregnancy, lactation), and the age of the animal (Krasnow and Steiner, 2006). Other studies have shown that leptin can be affected by changes in food intake (Williams et al., 2005). Leptin seems to act by modulating the function of several target glands (Moschos et al., 2002).

Glucose, another metabolite widely discussed in metabolism studies, plays a key role in animal metabolism. It is an essential source of energy for the maintenance of many tissues, such as the nervous system, red blood cells, the placenta, the mammary gland (Sastradipradja, 1998) and the ovaries (Rabiee et al., 1997; Leroy et al., 2004; Krasnow and Steiner, 2006). It also serves as a precursor for the biosynthesis of essential cell components, and is therefore equally important as a metabolite for all mammals. In the ruminant, most glucose must be synthesized by gluconeogenesis (Sastradipradja, 1998).

The aims of this study were to verify the effects of body condition score (BCS), breed and dietary supplementation on the blood leptin, insulin and glucose concentrations in hair sheep, and also to study the possible association of fertility and prolificacy with these blood metabolites and BCS and body weight (BW) recorded in ewes during the breeding season.

2. Materials and methods

2.1. Experimental site

The experiment was conducted on the Experimental Farm of the State University of Vale do Acaraú (Sobral, Ceara, Brazil), which is located in a semi-arid region at 3°36′ S, 40°18′ W and is 56 m above sea level. The mean annual temperature in the region is 32 °C and average rainfall is 800 mm per year.

2.2. Animals, experimental diets and management

A total of 96 non-pregnant adult ewes of different ages were used in this study. There were 48 ewes of the Santa Ines breed and 48 ewes of the Morada Nova breed, with average BW and standard deviation of the initial mean of 43.0 ± 5.6 kg and 25.55 ± 3.1 kg, respectively. First the ewes were tagged with an earring for identification. Then they were weighed and were randomly distributed into two groups, unsupplemented and supplemented, for each breed. During the experimental period, all of the ewes were managed under semi-intensive conditions and were grazed in a paddock used for rotational stocking that had been sown with Tifton 85 bermudagrass (Cynodon spp.). All of the animals had free access to water and mineral salt. For statistical analysis, the animals were grouped in a randomized block design (homogenous weights) with four treatments consisting of two supplementation levels (unsupplemented and supplemented) and two breeds (Santa Ines and Morada Nova). The experimental diet fed to the ewes in the supplemented groups was formulated according to the National Research Council (2007) and consisted of ground Tifton 85 hay (70%), 94.67% ground corn, 5.04% soybean meal and 0.29% limestone. The dietary supplementation was provided two weeks before and throughout the breeding season, which lasted for 45 days. The Santa Ines ewes received 250 g of supplementation daily, whereas the Morada Nova ewes received only 150 g because of their smaller size. The supplementation was provided at 9 a.m. each day.

Samples of Tifton 85 forage and concentrates were dried in a forced-air oven at 55 °C for 72 h before being ground in a knife mill with a screen with a 1 mm screen (Wiley mill, Arthur H. Thomas, Philadelphia, PA, USA). The samples were analyzed for their dry matter (DM) content (AOAC, 1990; method number 930.15) and crude protein content (CP; AOAC, 1990; method number 984.13). To analyze the neutral detergent fiber (NDF), the samples were treated with thermostable alpha-amylase without using sodium sulfite. The concentrate consisted of 92% DM, 10% CP, 12% NDF, 0.29% calcium and 0.16% phosphorus.

The breeding season for this study was considered to be 45 days long (starting at the end of June and ending in early August 2008). Five mature rams, two of the Santa Ines breed and three of the Morada Nova breed, with normal reproductive status were used during the breeding season. The rams were fed with the same dietary supplementation that had been formulated for the ewes.

The ewes remained in the paddock during the day and at the end of the day they were taken to the management center. Here they were presented to a teaser ram that had been painted on its sternum with a vegetable dye mixed in a Vaseline base so that the ewes that had been mounted could be identified by the pigment that had been transferred to their rump. After this, the ewes were presented to rams of the same breed for controlled natural mating.

The animals were weighed weekly in order to chart weight gain, and the BCS was measured in all ewes according to Russel et al. (1969). The BCS scale ranges from 1 (thin) to 5 (fat). Blood was sampled from 67 randomly selected ewes. During the trial, samples were collected from each animal by jugular venipuncture in non-anticoagulation gel separator vacuum tubes (Vacuntainer®) to obtain serum and to determine the leptin, insulin and glucose concentrations. Immediately after collection, the samples were centrifuged and the serum was stored at −20 °C until it was assayed.