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The reproductive performance of female goats treated with melatonin is not improved after introduction to bucks displaying springtime sexual activity if these does are experiencing decreasing body weight/condition score



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#### ABSTRACT

The aim of the present work was to determine whether treatment with melatonin modifies the reproductive response of female goats experiencing increasing or decreasing body weight (BW)/body condition score (BCS) when introduced to bucks displaying springtime sexual activity. During natural anoestrus, 53 does were isolated from bucks for a period of 42 days and distributed into two groups: 1) low BW/low BCS animals (N=24) (LLg group), which were fed 1.9 times their maintenance requirements so they would experience increasing BW and BCS; and 2) high BW/high BCS animals (N = 29) (HHl group), which were fed 0.4 times their maintenance requirements so they would experience decreasing BW and BCS. Half of each group was treated, or not, with melatonin (LLg + Mel N = 12, HHl + Mel N = 15, LLg-Mel N = 12 and HHl-Mel N = 14). On 6th May they were introduced to six males, showing natural sexual activity, fitted with marking harnesses (thus permitting the detection of oestrous activity). The ovulation rate was assessed by transrectal ultrasonography and confirmed via the plasma progesterone concentration (measured twice per week in blood samples). Plasma glucose, IGF-1 and non-esterified fatty acid concentrations were also determined, along with the conception rate, fertility, prolificacy and productivity of the does. LH concentrations and LH pulsatility were also recorded in the hours around introduction to the males. 'Oestrous plus ovulation' was observed only in does treated with melatonin. A higher conception rate and greater fertility and productivity were observed among the LLg + Mel does. These females showed higher glucose and IGF-1 concentrations after the introduction of the males. LH concentrations increased after male introduction independent of all other conditions. In conclusion, the present results show that treatment with melatonin does not enhance reproductive performance in does experiencing decreasing BW/BCS, but can improve it when does are experiencing increasing BW/body fat reserves — even when exposed to males displaying only springtime sexual activity. This might be explained by the higher blood glucose and IGF-1 concentrations of the LLg+Mel females.

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#### 1. Introduction

At Spanish latitudes, natural reproduction in goats is seasonal, with sexual activity occurring during autumnwinter. The photoperiod is the main environmental factor governing the seasonality of goat reproductive activity (Zarazaga et al., 2011a,b,c), and certainly in the Blanca Andaluza breed both the male (Gallego-Calvo et al., 2015a) and female (Gallego-Calvo et al., 2014) show reduced sexual activity during the lengthening days of spring. Information on the photoperiod is conveyed to the neuroendocrine system via the circadian secretion of melatonin from the pineal gland (Bittman et al., 1983). The administration of this hormone during the spring via the use of continuous, slow-release implants has been shown to advance the onset of the breeding season in goats (Chemineau et al., 1992; Williams et al., 1992). Usually, melatonin is provided following the separation of the sexes for 45 days (Zarazaga et al., 2012). When they are later brought back together, the "male effect" optimizes the female reproductive response.

The male effect can be employed to avoid seasonal anoestrus (for details see the review by Delgadillo et al., 2009), and is commonly used in extensive and semi-extensive stockraising systems in the Mediterranean. However, the results of the male effect depend greatly on factors such as breed, the type of previous isolation, the depth of anoestrus, postpartum stage, parity number, nutrition, body condition, and the degree of sexual activity displayed by males in spring (Walkden-Brown et al., 1993; Cerbito et al., 1995; Flores et al., 2000; Urrutia et al., 2003; Veliz et al., 2009; Delgadillo et al., 2009).

The nutritional status of domestic ruminants also affects their reproductive capacity. In Mediterranean extensive and semi-extensive stock-raising systems, food availability in the spring (when melatonin implants are used) can vary widely, and animals may experience an increase or reduction in body weight (BW) and body condition score (BCS) — fluctuations that can modify the reproductive response during male effect-induced spring breeding (Gallego-Calvo et al., 2015b). Variations in food availability may affect metabolic/nutritional factors such as plasma glucose, nonesterified fatty acids (NEFAs) and IGF-1.

Our group has described the interaction between nutrition and exogenous melatonin to affect LH secretion in female goats (Zarazaga et al., 2011b). In the former experiment we observed an increase in LH concentrations in ovariectomized females implanted with melatonin that showed low BW/BCS induced by a lower level of nutrition. In sheep, the ovulation rate is affected too (Robinson et al., 1991; Forcada et al., 1995; Rondón et al., 1996). All the latter authors report the effect of melatonin implantation to increase the ovulation rate more strongly in ewes with a low, rather than a high, feed intake or BCS.

It was hypothesized that, during seasonal anoestrous at Mediterranean latitudes (spring), melatonin administration might reduce the negative effect of a decreasing BW and/or BCS on the reproductive response of does when introduced to males displaying only springtime sexual activity. The present work examines: (1) how the reproductive response of does — with/without a melatonin implant

 to the male effect differs depending on whether female BW and/or BCS is increasing or decreasing, and (2) whether metabolic/nutritional factors explain any of the responses observed.

#### 2. Material and methods

#### 2.1. Study conditions

All procedures were performed by trained personnel in strict accordance with Spanish guidelines for the protection of experimental animals (RD 53/2013), and in agreement with European Union Directive 86/609. The study was conducted at the University of Huelva experimental farm (37° 20′N, 6° 54′ W), which meets the requirements of the European Community Commission for Scientific Procedure Establishments (2010/63).

#### 2.2. Animals and management

Before starting the experiment, the natural BW and BCS of the does was determined over three consecutive weeks to ensure that neither varied in any animal. On March 25th. 53 adult (4 years old), non-pregnant does in anoestrus were divided into two groups depending on their BW and BCS (Mean  $\pm$  SEM): 1) low BW ( $\leq$ 37 kg) (35.2  $\pm$  1.1 kg, range 25-37 kg/low BCS ( $\leq 2.50$ ) ( $2.45 \pm 0.03$ , range 1.75-2.50) (N = 24); this was known as the LowLow-gain (LLg) group since its members were fed 1.9 times their maintenance requirements, ensuring they would experience increasing BW and BCS; and 2) high BW (>38 kg) ( $40.6 \pm 0.5$  kg, range 38-52 kg)/high BCS ( $\geq 2.75$ ) ( $2.84 \pm 0.04$ , range 2.75-3.75) (N = 29); this was known as the HighHigh-loss (HHI) group since these animals were fed 0.4 times their maintenance requirements, ensuring they would experience decreasing BW and BCS. Both nutrition regimens were designed according to INRA standards (Morand-Fehr and Sauvant, 1988) and followed for a total of 98 days.

The feed provided was a commercial concentrate composed of oats (24.7%), maize (23.0%), peas (20.4%), barley (16.3%), lucerne pellets (12.2%) and a mineral-vitamin complement (3.4%). The nutritional values of the concentrate were 0.94 milk fodder units (UFL) and 77 g digestible protein/kg dry matter. This concentrate was offered individually once per day. In addition, barley straw, distributed to each group as a whole, provided 0.37 UFL and 25 g of digestible protein/kg dry matter. All animals had free access to water and mineral/vitamin blocks. The aim of the feeding regimens was that, at the time of introduction to the males, the does of the LLg group would have approximately the BW and BCS originally shown by the does of the HHI group, and that the does of the HHI group would have approximately the BW and BCS originally shown by the does of the LLg group.

At the start of the experiment (March 25th), 12 does from the LLg group (and 15 from the HHI group received a single subcutaneous implant containing 18 mg of melatonin (Melovine®) (CEVA Salud Animal, Barcelona, Spain) at the base of the left ear (LLg + Mel and HHI + Mel subgroups).

To determine their ovarian cyclicity before the onset of the experiment (March 25th), blood samples were col-

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