Theriogenology 123 (2019) 159–166

Contents lists available at ScienceDirect

Theriogenology

journal homepage: www.theriojournal.com

Effect of bST administration on plasma concentrations of IGF-I and follicular dynamics and ovulation during the interovulatory cycle of sheep and goats

J.F. Cox [a, *], F. Navarrete [a], A. Carrasco [a], J. Dorado [b], F. Saravia [a]

[a] Department of Animal Science, Faculty of Veterinary Sciences, Universidad de Concepción, Avda. Vicente Méndez 595, Chillán, Chile
[b] Department of Animal Medicine and Surgery, Faculty of Veterinary Medicine, Universidad de Córdoba, Spain

ARTICLE INFO

Article history:
Received 13 June 2018
Received in revised form
1 October 2018
Accepted 2 October 2018
Available online 3 October 2018

Keywords:
bST
IGF-I
Plasma levels
Follicular dynamics
Ewes
Goats

ABSTRACT

This study used a comparative approach to gather clinical information to assess the effect of bovine somatotropin (bST) on follicular dynamics and ovulation in sheep and goats during an interovulatory cycle. The performance of general markers of ovarian function and specific features of follicular dynamics obtained by daily ultrasonography (US) were used to assess the hypothesis that bST, associated with supraphysiological levels of IGF-I, was able to disrupt the follicular dynamics and ovulation in Highlander ewes and Saanen goats. In Exp 1, 15 ewes and 14 goats were estrous-synchronized (P_4-6 days + PGFα d-6) and then allocated to a bST-treated group (50 and 100 mg, Lactotropin®; n = 5 females each) and to an untreated control group (5 ewes and 4 goats) to assess the activity of bST through plasma IGF-I (RIA). In Exp 2, 12 animals from each species were synchronized. At day 6, they were divided into a bST-group (100 mg in sheep and 50 mg in goats, n = 6 each) and an untreated control group (n = 6 each). Starting at day 6 and up to 22 days after ovulation in sheep and 25 days in goats, each female was subjected to daily US (10 mHz probe) to assess follicular and luteal (CL) dynamics and ovulation. This included assessments of both general ovarian features and specific follicular wave features. Our results showed that bST increased plasma IGF-I by day 3 ($p < 0.01$) when compared to the control group. Moreover, these concentrations were maintained for at least 10 days in sheep and 10 days in goats before returning to pre-treatment concentrations. Increases in IGF-I after bST doses were similar in terms of a daily and total amount ($P > 0.10$). Results from Exp.2 indicate that in sheep, bST administration had a subtle inhibitory effect on follicular function. However, bST in goats had a stronger influence, extending the interovulatory cycle ($P = 0{,}034$), increasing the number of follicular waves during the period ($P = 0.003$), and reducing the functional potential of large follicles as measured by their lower follicular diameter ($P = 0.02$), duration of the follicle waves ($P = 0.02$), and persistence of follicles after reaching their maximum diameters ($P = 0.04$). In addition, untreated sheep and goats shared common patterns of terminal follicular development and ovulations characterized by overlapping between follicular waves and ovulations of follicles from different waves, features not seen in cattle.

1. Introduction

The mechanisms linking energy metabolism and reproduction have been matters of intensive research in ruminants [1,2]. Reproduction is seen as a biological function that requires energy to sustain gestation and lactation [3,4]. However, in spite of its importance in species conservation, reproduction has a secondary priority in nutrient partitioning, and it is suppressed under an energy status that compromises gestation and lactation. Energy status is monitored on an acute basis through endocrine and nutritional signals, which are assessed by central and peripheral specialized cells and tissues. Subsequently, these cells convey this information to regulatory mechanisms controlling the reproductive axis [4,5].

Growth Hormone (GH), insulin, and IGF-I are considered major signals from the metabolic axis. Furthermore, the plasma concentrations of the last two are seen as gateways for sound terminal follicular development, ovulation, and fertility in mammals due to

* Corresponding author.
E-mail address: jfcox56@gmail.com (J.F. Cox).

https://doi.org/10.1016/j.theriogenology.2018.10.003
0093-691X/

their promoting actions and their ability to synergize with gonadotropins on terminal follicular development [6,7]. Both act through their receptors, insulin receptor (INSR) and IGF-I receptor (IGFIR), on granulosa cells to directly promote terminal follicular development by increasing cell proliferation, differentiation, and estradiol production and by reducing apoptosis and increasing the expression of receptors to FSH and LH and the potency of second messenger activation [8–10]. In spite of the limited expression of mRNA for GH receptors (GHR) in granulosa cells in ruminants, there is consensus that their actions at the ovarian level are mediated by IGF-I. In fact, the central role of IGF-I signaling in terminal follicular function was recently highlighted in knockout mice. In this study, knockout mice lacking IGFIR in granulosa cells exhibited infertility and their ovaries lacked late antral follicles [11]. However, the knockout for INSR exhibited no measurable effects on fertility.

Bovine GH (bovine somatotropin, bST) has been used in cattle, sheep, and goats as an approach to improve responses to superovulatory treatments with conflicting results [12]. The administration of bST increases the plasma concentrations of insulin and IGF-I [8,9]. Moreover, collective information indicates that both are mediators of bST influence on terminal follicular development, sharing receptors, and even subcellular signaling pathways due to their molecular similarities in ruminants [6]. Hyperinsulinemia has been shown to affect follicular development and steroidogenesis [13,14], whereas high concentrations of IGF-I have been reported to affect follicular function by down-regulating its receptors [15]. Therefore, variability among experiments could be expected due to bST dosage, but also due to the method of ovarian examination, type and dose of gonadotrophin use for ovarian stimulation, and the number, production status, and body condition of animals used in such studies [9,12].

Follicular dynamics are under tight regulation by the interplay between the reproductive and metabolic axes just described [10,16] and provide functional markers that could lead to information on the relative importance of regulatory factors on terminal follicular function and the manner in which to control them. Thus, assessing the influence of bST on patterns of terminal follicular development can be used as a physiological model to establish metabolic requirements to optimize the efficiency of reproductive protocols [17,18]. This study aimed to assess the influence of the administration of a high dose of bST on follicular dynamics during an interovulatory cycle in sheep and goats, using total IGF-I as a marker of biological activity. Due to the supraphysiological concentrations of IGF-I and insulin, we expected a disruptive influence of bST on follicular dynamics and ovulation in each species.

2. Materials and methods

2.1. Animals and general management

This study used non-lactating and clinically sound Highlander ewes (n = 27) and Saanen goats (n = 26), 3–7 years old. It was conducted at the Faculty of Veterinary Sciences animal facilities, Universidad de Concepción, 36° south latitude, 71° west longitude. Animals were kept in collective pens with adequate space to rest and feed, ventilation, dry bedding, and free access to fresh water. During the day, animals were allowed access to a 4-ha paddock for grazing and exercise. Feeding at the research station was based on lucerne hay, oats grain, calf commercial concentrate, and mineral salts to maintain a BCS of around 3.0. Moreover, animals were subjected to a preventive health program for endemic diseases. Housing and animal procedures followed standards approved by the Ethics Committee of the Faculty of Veterinary Sciences at the Universidad de Concepción.

2.2. Estrous detection

Estrous detection was conducted three times daily (early AM, mid-day, and late PM) by direct observation of tail waving and the attraction exhibited by ewes and goats to males partially separated by a fence that prevented mounting. Hand-controlled teasing was used when required, in order to expose the tolerance of females to be mounted.

2.3. Blood sampling and endocrine measures

Blood samples (3 mL) were collected by jugular venipuncture into heparinized glass tubes and were maintained on ice (less than 2 h) until plasma collection. Plasma was obtained by centrifugation at 5 °C (1500 × g, 20 min), and samples were labeled and stored at −20 °C until assayed. IGF-I concentrations were measured by solid phase RIA using a commercial kit (DIASource, Louvain la Neuve, Belgium) validated for ruminants. The inter- and intra-assay variations were <2.8 and < 5.1%, respectively, and the limit of sensitivity was 3.4 ng/mL.

2.4. Follicular dynamics and ovarian measures

Follicular dynamics were monitored by transrectal ovarian ultrasonography (US) using a 10-MHz linear array probe connected to a B-mode, real-time scanner (Honda 2010 Vet, Toyohashi, Japan). The transducer was fitted to a plastic rod that allowed the transrectal manipulation of the probe. Images were viewed at a magnification of × 2.0 with constant gain and focal point settings. Images of antral follicles and corpora lutea (CLs) were frozen and measured in mm by internal calipers.

Ovarian function was assessed by daily ultrasound that started at 9:00. Antral follicular dynamics were measured during a single interovulatory period. The relative position and dimension of follicles and luteal structures were sketched on ovarian charts. At each examination, the mean diameter and relative location of all follicles ≥3.5 mm in diameter and CLs were registered and mapped to analyze patterns of growth and/or regression.

General ovarian features that were considered in this study included interovulatory intervals, intervals between luteolysis and ovulation and between estrous and ovulation, ovulatory rate, number of ovulatory follicles and diameters of ovulatory follicles, and mature CLs.

2.5. Follicular dynamics definitions

A follicular wave was defined as the follicle or group of follicles that grew beyond ≥3.5 mm up to at least 4.3 mm and 5.0 mm (minimum size recorded for an ovulatory follicle) in sheep and goats, respectively, before atresia or ovulation. We defined recruited follicles as the cohort of follicles ≥3.5 mm that signaled the beginning of a follicular wave. Follicles ≥4.3 and 5.0 mm in diameter were considered pre-ovulatory, and ovulatory follicles were follicles that ovulated as confirmed by assessment of CLs 5–7 days later. The growth phase of large follicles was the interval between emergence and the maximum follicular diameter, whereas the growth velocity was the average mm/day obtained by the large follicle in the growth phase. Follicular persistence was the interval in days between large follicles reached their largest diameter and when they experienced atresia or ovulation, whereas the duration of a follicular wave was defined as the interval in days between the emergence of up to two of the largest follicles and their atresia or ovulation. Atresia was defined as the irreversible reduction in diameter of large follicles to below 4.0 mm and ovulation as the disappearance/collapse of large follicles between two consecutive