



## Effect of different male-to-female ratios and testosterone administration upon the male sexual behavior and the out-of-season reproductive response of anestrus goats



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### ABSTRACT

The possible effect of testosterone administration and the male-to-female ratio regarding the male sexual behavior during the resting season and the out-of-season reproductive performance of anestrus goats exposed to the male effect under subtropical conditions (26° N) was evaluated. In the experiment-1 two treatments were considered (1) Testosterone-treated bucks (TTB;  $n = 4$ ; 25 mg, i.m., testosterone, every 3-days  $\times$  3-week), and (2) Non-testosterone treated bucks (NTTB;  $n = 4$ ; i.m. saline every 3-days  $\times$  3-week). Thereafter, both experimental groups were exposed to adult goats over two days (1 h  $\times$  2days) and two sexual behavior tests were performed: appetitive sexual behavior (ASB) and consummatory sexual behavior (CSB). In the experiment-2, multiparous lactating and anoestrous crossbred goats ( $n = 60$ ) were randomly assigned to one of four treatments with different male-to-female ratios (MFR): (1) High MFR goats [HMFR;  $n = 20$ , 1:10 ratio], (2) Low MFR [LMFR;  $n = 10$ , 1:5 ratio] each group exposed to two NTTB, (3) High MFR goats [HMFR + T;  $n = 20$ , 1:10 ratio], and (4) Low MFR [LMFR + T;  $n = 10$ , 1:5 ratio] each group exposed to two TTB. While the TTB displayed higher ASB ( $p < 0.01$ ; 91.9% vs 8.1 %), the NTTB did not express neither CSB ( $p > 0.05$ ; 100% vs 0%) nor sexual behavior irrespectively of the male-to-female load. The HMFR + T depicted a higher ASB ( $p < 0.01$ ; 65% vs 35%) than the LMFR + T, without differences in CSB between HMFR + T and LMFR + T. Also, TTB induced estrus response (86.6%) and pregnancy rate (83.3%) while NTTB did not. Neither estrus response (85 vs 90%) nor pregnancy rate (85 vs 80%) differed between the HMFR + T and LMFR + T groups. Exposing of anestrus goats to testosterone-treated bucks, irrespectively of mating load, was able to successfully invoke neurophysiological pathways to activate ovarian function and to promote a uterine milieu prone to the establishment of pregnancy during the anoestrus season.

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### 1. Introduction

In Northern Mexico, in the Comarca Lagunera 26°N, mixed-breed goats with a high proportion of dairy genes are considered seasonal breeders with female and male goats displaying a period of reproductive inactivity independently of food availability (Delgadillo et al., 1999). Certainly, the bucks breeding season

lasts from May to December (Delgadillo et al., 1999), whereas dams exhibit sexual activity from September to February–March (Duarte et al., 2008; Rivera-Lozano et al., 2011). Therefore, bucks depicting a complete sexual behavior may not be available for breeding at the beginning of the year, a crucial period for goat producers. The last is particularly true under rain fed-pastoralist systems, where kidding during summer is highly desirable, since at this time the rainy season begins. Interestingly, does at this latitude depict a “non-profound anestrus”, making possible to breed them out-of-season, as long as these anestrus goats are exposed to sexually active bucks (Rivas-Muñoz et al., 2007; Véliz et al., 2009; Gonzalez-Bulnes et al., 2011; Luna-Orozco et al., 2012).

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Several technologies have been developed to bring sexually inactive bucks into full sexual activity during the non-breeding season (Gonzalez-Bulnes et al., 2011). One of the most effective and studied methods is the use of lighting regimes, where 'long day photoperiods' stimulate sexual activity in bucks exposed to 16 h-light/8 h-dark, during 2–3 months (Pellicer-Rubio et al., 2007). In bucks, this artificial lighting regime generates increased levels of LH and consequently higher testosterone secretion, which in turn influences the production of pheromones (Okamura et al., 2010) while a complete sexual activity in bucks is observed, with full libido, abundant production of pheromones, increased scrotal circumference and higher semen quality (Delgadillo et al., 1992, 2004).

Yet, the use of this procedure is questionable since controlled lighting requires the availability of a light proof barn to house the bucks. In addition, the building of such infrastructure and the lighting costs added to a complicated and time-consuming herd management is beyond the economic and physical possibilities of most goat-keepers under marginal-range conditions systems. However, while light-treated bucks have displayed an important sexual drive when compared to testosterone-treated bucks (Luna-Orozco et al., 2012), a delay in the doe response to estrus induction varies according to the male-to-female ratios (Carrillo et al., 2007). Building-up on such findings, this study aimed to determine if testosterone administration to non-sexually active mixed-breed bucks can render them sexually active during the spring-natural anestrus season (26°N). Additionally, we aimed to determine if a potential increased sex drive and sexual motivation can be displayed by the testosterone-treated bucks towards the anoestrous does, triggering ovulation and pregnancies, when bucks and does are subjected to different mating loads.

## 2. Materials and methods

### 2.1. Location, environmental conditions, management and experimental design

The experiment was conducted from March to April in a semi-desert area of northern Mexico (Comarca Lagunera, 26° 23' N, 104° 47' W; altitude 1,140 m) dominated by a microphyll desert scrub. Mean annual temperature is 27 °C and average annual precipitation is 230 mm, with 70% falling from June to October. Historically, this communal pasture has been heavily stocked and continuously grazed mainly by large herds of goats and cows, with sheep in lesser extent. Therefore, goats grazed on a deteriorated rangeland showing a low forage production potential. All procedures and methods used in this study were performed by trained technicians and in strict accordance with accepted international guidelines regarding to the use and care of animals (Federation Animal Science Society, 2010).

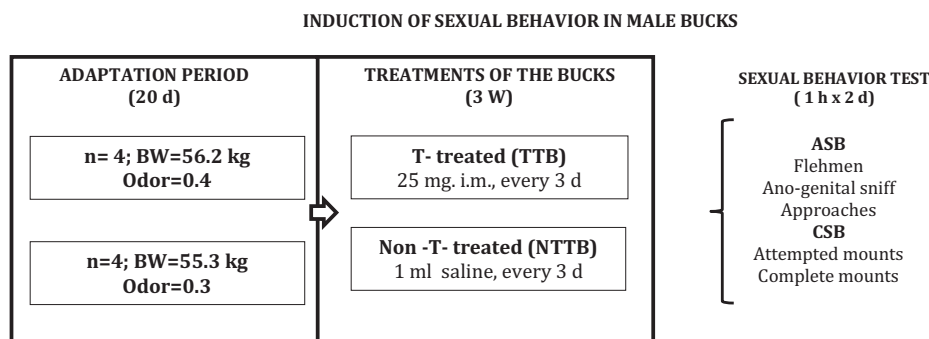
### 2.1.1. First experimental phase

This phase of the study considered the evaluation of the sexual status and induction of sexual behavior in male goats. A total of eight sexually experienced mixed-breed adult bucks of proven fertility and similar genetic background with a high level of dairy breeds were used. Age, mean live weight and body condition score (grading scale 0–5; Santucci and Maestrini 1985) of bucks were 3–4 years old,  $49 \pm 2$  kg and  $2.5 \pm 0.5$  units, respectively. Sexual motivation of bucks was at a low ebb, with a noticeable "female goat odor", therefore identified as sexually inactive. Early on in March, bucks were placed in a roofed dirt pen (10 × 10 m) with free access to water, a mineral–salt mixture and alfalfa hay (17% CP, 1.95 Mcal ME). In addition, each buck received daily 200 g of a commercial concentrate (14% CP and 1.7 Mcal ME). Bucks were randomly allotted into two groups ( $n=4$ , per group). Each group was placed in pens 100 m apart from each other. Before and during this first phase of the experimental period, both groups of bucks were kept under conditions of natural day length (26°N); this adaptation period considered 20 days. Thereafter, bucks were exposed to two treatments: (1) Testosterone-treated bucks (TTB;  $n=4$ ) which received 25 mg i.m., testosterone, every 3-days × 3-weeks (Lab Brovel, DF, Mexico), and (2) Non-testosterone treated bucks (NTTB;  $n=4$ ), which received an i.m. injection of saline every 3-days × 3-weeks. After the 3-week treatment period, a behavioral sexual test was performed to both experimental groups in order to evaluate both appetitive sexual behavior as well as consummatory sexual behavior by exposing each group to a set of adult female goats (1-h × 2-days); response variables within experimental groups were registered and will be pointed-out in the next section. A schematic representation of the main activities carried-out across time between experimental groups during this first experimental phase is depicted in Fig. 1.

### 2.1.2. Second experimental phase.

This second phase of the experimental study was performed in order to evaluate the possible influence of the "male effect" using both male groups (TTB and NTTB) and exposing them to anovulatory adult goats, yet considering two different male-to-female ratios. The response variables included in this second experimental phase considered the sexual behavior of males as well as the sexual behavior and reproductive outcomes of the anoestrous goats.

Healthy multiparous anoestrous crossbred goats ( $n=60$ ) with a mixture of dairy breeds of known fertility from a commercial goat herd were used in this study. Previous to this second phase of the study, goats grazed on an open range driven by a herdsman, 7-h daily (1100–1800 h); goats were maintained in isolation from the sight, sound and smell of bucks prior to the trial. Goats had given birth in September of the previous year and were hand-milked once daily throughout the study. After balancing for weight and body



**Fig. 1.** Experimental schedule for induction of sexual behavior of mixed dairy-breed bucks: Testosterone-treated bucks (TTB,  $n=4$ ) and Non-Testosterone-treated bucks (NTTB,  $n=4$ ) exposed to anovulatory mixed dairy-breed female goats (1 h × 2 days) to perform an appetitive sexual behavior test (ASB) and a consummatory sexual behavior test (CSB) during the traditional male reproductive resting season (Spring) at 26°N.

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