ORIGINAL RESEARCH

Influence of sexually inactive bucks subjected to long photoperiod or testosterone on the induction of estrus in anovulatory goats

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Abstract The objective of this study was to evaluate the efficacy of treating sexually inactive bucks with artificial long photoperiod or testosterone on the induction of estrus in anovulatory grazing goats. A total of 91 multiparous mixed-breed anestrous goats were randomly assigned to one of three treatment groups: (1) joining with bucks subjected to 2.5 month of artificial long days (16 h of light/day; n=31), (2) joining with testosterone-treated bucks (n=30), and (3) joining with untreated bucks (control; n=30). There were no differences between the light-treated (100%) and testosterone-treated (93%) bucks in their ability to induce estrus in anovulatory does. On the other hand, none of the goats in contact with control bucks exhibited estrus. The interval from start of mating to estrus was shorter in goats

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J. E. García · M. Mellado Departamento de Nutrición Animal, Universidad Autónoma Agraria Antonio Narro, Saltillo, Mexico with the light-treated bucks (37.9±4.8 h) compared with does in contact with testosterone-treated bucks (58.3 ± 8.7 h). The overall pregnancy rate in goats joined with light-treated, testosterone-treated and control bucks was 84%, 77% and 0%, respectively, with no difference (P > 0.05) between the first two groups. Anogenital sniffing, approaches, mounting attempts, and mounts were highest (P < 0.01) in light-treated bucks and lowest in control bucks. It was concluded that testosterone-treated bucks and long-day-treated bucks were equally effective in synchronizing estrus in anovulatory goats and resulted in similar levels of fertility. Given that lighttreated bucks are unviable in communal production systems of goats raised by resource-poor farmers, the sexual arousal of bucks with testosterone is a practical and reliable method to induce ovulation in anovulatory goats in pastoral goat systems in hot environments.

Keywords Goats \cdot Photoperiod \cdot Ovulation \cdot Estrus \cdot Testosterone \cdot Long days

Abbreviations

BC Body condition

Introduction

In the tropics, goats are continuous breeders, although forage restriction may occasionally causes anoestrous periods (Fatet et al. 2011). Even at latitude 25°N, native breeds are capable to breed in spring (Mellado et al. 2006); however, indiscriminate crossbreeding of native goats with dairy breeds originated in the Alps has led to seasonal anoestrous in crossbred goats at this latitude, although this seasonal anestrous is less marked than that observed in temperate zones (males: Todini et al. 2007; Zarazaga et al. 2009; females: Carcangiu et al. 2009; Arrebola et al. 2010). Induction of fertile estrus for out of season breeding is important for goat producers in many countries in order to overcome the seasonality of the goat milk supply; thus, a considerable research has been carried out to enable yearround kidding of goats (Husein et al. 2005; López-Sebastian et al. 2007). Several methods are available to induce off-season cyclicity and estrous synchronization in does. The most common and widespread method to induce ovulation in anestrous goats is the use of synthetic hormones (Leboeuf et al. 2008), but this technique is out of reach for goat producers in resource-poor systems in subtropical and tropical ecosystems in developing countries. Controlling photoperiod is another technology to induce does and bucks (Ramadan et al. 2009) into sexual activity in spring. However, this technique requires adequate facilities and labor, which, again, is beyond the possibilities of goat producers in resource-poor environments. The buck or "male effect" is an effective management practice to induce ovulation in seasonally anovulatory goats, and because this technique requires a minimal amount of labor and cost, this is the only viable alternative for goat producers in extensive systems. In order for this scheme to be effective, does must present a "shallow" anestrous and bucks must be sexually active (Veliz et al. 2009; Rivas-Muñoz et al. 2010).

Sexual inactive bucks can be brought into full sexual activity by supplementing light during housing and using the natural light as the reduced photoperiod (Pellicer-Rubio et al. 2007). However, the high cost and lack of electricity in goat pens under rangeland conditions make such a practice impossible to implement. Therefore, a need exist for a simpler and effective method to bring bucks into sexual activity, which in turn stimulate the doe flock to become sexually active. The objective of this study was to assess the effectiveness of the application of testosterone or prolonged exposure to a long-day photoperiod to induce sexually inactive bucks to mate in spring, and their ability to induce estrus and impregnate mixed-breed anovulatory goats on rangeland.

Material and methods

The experiment was conducted in a commercial goat farm under extensive conditions in northern Mexico (26°N). Average annual precipitation at the study area is 230 mm, and the highest ambient temperature is 41°C in May and June and the lowest -3° C in December and January. Relative humidity ranged between 26.14% and 60.59% and day length from 13 h 41 min during summer solstice

(June) and 10 h 19 min in the winter solstice (December). This landscape is dominated by the shrubs creosote bush (*Larrea tridentata*) and mesquite (*Prosopis glandulosa*). Goats were grazed on rangeland most of the times, and occasionally on crop residues, mainly corn and cotton.

Buck management

Six sexually experienced mixed-breed adult bucks of proven fertility were used. These animals were kept in a ruffed cement floor pen $(6 \times 6 \text{ m})$ before breeding, where they had free access to water and a mineral mix. Twice daily, bucks were offered alfalfa hay ad libitum consumption. Bucks were randomly allotted into one of three groups (two bucks per group): exposure to long-day artificial photoperiod, injections of testosterone and control (natural photoperiod). Two of the bucks were subjected to a long-day treatment (16 h of light/8 h of darkness) during 2.5 months, starting November 1, 2009 and followed by a natural photoperiod. The second group of bucks received intramuscular injection of testosterone (50 mg, Testosterona 50, Lab Brovel, DF, Mexico) every 3 days during 3 weeks before joining. The control group did not receive supplemental light or testosterone. Bucks were kept permanently in pens; therefore, they were in contact with does from 1900 to 1100 h daily.

Does management and variables recorded

Ninety-one pluriparous lactating mixed-breed (dairy×native) goats of known fertility and kept on rangeland were maintained in isolation from the sight, sound, and smell of bucks before the trial. Mean condition score (1=extremely thin; 5=extremely fat; palpation over lumbar vertebrae, ribs and sternum) was 2.2 ± 0.1 , and goats ranged in weight from 29 to 58 kg. Does had free access to water and a commercial mineral at the pen. All groups of goats grazed separately from 1100 to 1900 h, guided by goat keepers. All groups of goats were treated identically and grazed the same type of vegetation. Before joining with bucks, all does were treated with a single intramuscular injection of 20 mg progesterone (Fort Dodge[®], DF, Mexico) in order to reduce the occurrence of short luteal cycles.

These does were randomly assigned to one of three treatment groups: (1) joining with bucks subjected to artificial long photoperiod (n=31), (2) joining with testosterone-treated bucks (n=30), and (3) joining with untreated bucks (control; n=30). The mating period started on March 22, 2010 and lasted 4 weeks. Goats were not treated against intestinal parasites, because this is not a health problem in this dry environment.

Measurements and recordings

The sexual behavior of bucks was assessed by recording flehmen, anogenital sniffing, nudging, mounting attempts, and mounts. These observations were made during 1 h the first 2 days of joining.

Daily occurrence of estrus (goats showing estrus signs or copulation) was recorded. Estrus was observed for 1 h twice daily (0900 and 1700 h) during the first 15 days of joining. The interval between the onset of joining and occurrence of estrus was also recorded. Short estrous cycles and the interval between these cycles were registered. The length of short estrous cycle was defined as the number of days between two consecutive periods of estrus, when this interval was shorter than 10 days. Transrectal real-time B mode ultrasound scanning (Aloka SSD 500 Echo camera, Overseas Monitor Corp. Ltd., Richmond, Japan) was used for the diagnosis of early pregnancy (45 days post-joining).

Statistical analysis

Percentage of goats in estrus and pregnancy rates (number of pregnant goats/number of mated goats in each group× 100) were analyzed as binomial data with the LOGIT function of the PROC GENMOD of SAS (SAS Inst. Inc., Cary, NC, USA). The model statement contained the effect of treatment (light-treated, testosterone-treated or control bucks). Except for estrus response, data for all other variables were analyzed excluding control bucks, as these animals did not elicit any sexual response in anovulatory does. Interval to estrus was analyzed by the GLM procedure of SAS. Sexual behavior of bucks was compared using the chi-square test.

Results

Figure 1 summarizes the behaviors of bucks during the exposure period to anovulatory does. During the 1-h exposure period during 2 days, the light-treated bucks were more sexually responsive than testosterone-treated and control bucks, which was reflected in a higher (P < 0.01) frequency of flehmen than testosterone-treated bucks and controls. Anogenital sniffing was 3.5 times higher (P < 0.01) in lighttreated bucks than testosterone-treated bucks, with the lowest response in control bucks. Mounts in light-treated bucks were four times higher (P < 0.01) than testosterone-treated bucks, whereas control bucks did not show mounting activity.

Over 90% of eligible goats responded to the stimulus of light-treated bucks or testosterone-treated bucks, whereas none of the goats exposed to control bucks exhibited estrus

% 0 0 2 3 5 6 7 8 10 11 12 13 1 4 9 14 Days after bucks introduction Fig. 1 Percentage of does showing behavioral estrus after the stimulus of bucks previously treated with long photoperiod or testosterone

(Table 1). Pregnancy rate of goats induced into estrus by light-stimulated bucks was 7 percentage points higher compared to goats joined with testosterone-treated bucks, with no difference (P>0.05) between these groups. No pregnancies occurred in goats joined with control bucks. The interval to estrus was shorter (Table 1) and more synchronized (Fig. 2) in goats joined to light-stimulated bucks than testosterone-treated bucks.

Discussion

Behaviors displayed by bucks differed notably among groups of bucks, with light-treated bucks showing the strongest sexual drive in response to anestrous females. In goats, flehmen response is exhibited after a determination of estrus had occurred (Ungerfeld et al. 2006). Therefore, this sexual behavior is apparently used to verify or process information received by the primary olfactory mode. Since the interval to estrus was shorter and more synchronized in goats joined with light-stimulated bucks than testosteronetreated bucks, it appears that the higher levels of sexual performance exhibited by light-treated bucks elicited a stronger sexual stimulus than testosterone-treated bucks, and therefore, estrous does were available sooner for bucks to express their investigatory behavior. It could be that the behavioral response of the light-treated bucks was displayed when the receptive status of the stimulus animal may have already been ascertained, and these behaviors may serve to sustain sexual interest or elicit proceptivity in teased does.

The higher willingness of light-treated bucks compared to testosterone-treated bucks to seek and court the does could be due to a higher serum testosterone concentrations in the former animals, as concentration of this hormone is increased with photoperiodic treatment of males (Bedos et al. 2010), which lead to a increased male odor as well as sexual behavior.



	Control	Long-day treated bucks	Testosterone-treated bucks
Estrus (%)	0 (0/30) ^a	100 (31/31) ^b	93 (28/30) ^b
Estrus day 0 to 5 (%)	_	29/31b	26/30b
Estrus day 6 to 15 (%)	_	19/29 ^a	11/30b
Interval to first estrus (h)	_	37.9 ± 4.8^{a}	58.3 ± 8.7^{b}
Short estrous cycles (%)	_	65 (20/31) ^a	9/28b
Length short estrous cycles (days)	_	$10{\pm}2.2^{a}$	6.2±0.1b
Pregnancy rate (%)*	_	84 (26/31) ^a	77 (23/30) ^a

 Table 1
 Reproductive performance of anestrous mixed-breed goats on rangeland exposed to bucks subjected to prolonged photoperiod or treated with testosterone before joining in March

Nonpercentage data are means \pm SD. Means with different superscripts in rows differ (P<0.05)

*Pregnancy rate at 45 days post-joining

On the other hand, injections of testosterone clearly elicited sexual arousal in sexually inactive bucks, but these animals were subject to an increasing sensitivity to the negative feedback action of testosterone on LH, and possibly, levels of testosterone were not high enough to reach the same levels of male sexual activity displayed by the light-treated bucks.

Does in this study were challenged by a very contrasting buck stimulation (high, medium, and low). Despite these differences in the intensity of sexual stimulus, no difference in estrus response and pregnancy rate was noted in does joined with light-treated and testosterone-treated bucks. This suggests that stimulus by both groups of bucks displayed a similar male-induced pulse and plasma LH secretion pattern, which were sufficient to promote a subsequent sustained increase in plasma LH, necessary for ovulation to occur. These results are not in line with data of



Fig. 2 Sexual behavior components of sexually inactive bucks subjected to artificial long photoperiod or treated with testosterone before joining in March. Number of behavioral events during 1 h in two consecutive events

Vielma et al. (2009) who observed that control bucks were unable to stimulate ovulation in does in spring. The same response has been documented with mixed-breed sexually inactive bucks in this region (Bedos et al. 2010).

Full contact is not necessary for does to respond to males (Rivas-Muñoz et al. 2007; Delgadillo et al. 2009); under the conditions of the present study, the fact that high level of buck stimulation was not required for the does to respond to the buck effect has an important commercial implication. Data suggest a great sensitivity in does to bucks in early spring. This suggests that light-treated bucks had a higher capacity to detect, but not induce, estrus behavior in goats.

As it has been observed in rams (Perkins and Fitzgerald 1992), mounting and flehmen behaviors were positively correlated. In summary, this study provides evidence that sexual behavior of bucks subjected to long photoperiod treatment before mating is much more intense than that exhibited by testosterone-treated buck, but this higher sexual arousal was not reflected in higher pregnancy rates. These results have an attractive practical implications because it demonstrate that a short testosterone treatment (3 weeks) to sexually inactive bucks elicit an effective sexual awakening of buck, which in turn induces ovulation in anestrous goats. This management scheme can fully replace the costly, tedious, and elaborated photoperiodic regimens to induce sexual activity in bucks and therefore can be practically applied in extensive goat production systems.

Conclusions

This experiment provides evidence that mixed-breed bucks stimulated by long photoperiod at latitude 26°N before breeding in spring, or injected with testosterone before exposure to does are equally effective in brining anovulatory does into estrus. Quality of estrous cycle resulting from introducing bucks sexually aroused either with photoperiod treatment or testosterone administration seems to be equal, as no difference in pregnancy rate was detected between groups. These data also show that at this latitude and with mixedbreed goats under rangeland conditions, bucks not sexually active are incapable to induce estrus and ovulation in anovulatory does. Testosterone administration to sexually inactive goat bucks has potential application in extensive goat production systems in subtropical areas, in order for the flock to become sexually active during the short anestrous period experienced in both bucks and does.

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