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J Anim Sci 1999. 77:2073-2078.

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Facilitation of Sexual Behavior in French-Alpine Goats Treated with Intravaginal Progesterone-Releasing Devices and Estradiol During the Breeding and Nonbreeding Seasons¹

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ABSTRACT: The effectiveness of administering progesterone (P₄) using controlled intravaginal drug release (CIDR) devices on estradiol (E₂)-induced sexual behaviors was examined in ovariectomized (ovx) French-Alpine goats during the fall and spring. Estradiol-induced attractivity and receptivity were facilitated during the spring when P₄-filled CIDR devices were removed 24 or 48 h before injection of 30 µg of E₂. During the fall, attractivity was also facilitated by CIDR removal 24 h prior to E₂ injection, whereas E₂-induced receptivity was unaffected by removal of the CIDR at this interval. Concentrations

of P₄ in circulation during the 3 d of treatment with a CIDR were similar to those during the late luteal phase of the estrous cycle in intact goats. Treatment with P₄-filled CIDR for 3 d, followed by injection with 30 µg of E₂ 24 h after removal, was determined to be a useful model for inducing sexual behavior in a physiologically relevant manner, and it may also be an effective means for facilitating estrus detection due to the high frequency of display of sexual behavior during a predictable time period following steroid treatment.

Key Words: Goats, Sexual Behavior, Progesterone, Seasons

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J. Anim. Sci. 1999. 77:2073–2078

Introduction

A progesterone (P₄)-filled controlled intravaginal drug release (CIDR) device is currently being marketed in Australia and New Zealand for synchronization of estrus in cattle, sheep, goats, and deer. The device has been successfully used to synchronize estrus for AI in goats (Ritar et al., 1989). However, because the efficacy of cervical insemination remains low (Ritar et al., 1990), breeders continue mainly to use natural matings (Jainudeen and Hafez, 1993). It is still beneficial to synchronize estrus under natural mating conditions in order to shorten the interval between kiddings, thus minimizing labor costs. With natural mating, the estrous behavior of does is important for ensuring successful service by the male.

The first ovulation of the breeding season is often not accompanied by overt behavioral estrus and is termed a "silent" estrus (Cole and Miller, 1933). Occurrence of a silent estrus means the does may not be bred until the second ovulation of the breeding season. A method of synchronizing estrus that also increases the likelihood of display of estrous behaviors would be highly desirable.

Goats undergo a breeding season during the fall and winter and a nonbreeding season during the spring and summer (Robinson, 1959). Studies with ovariectomized (ovx) goats have shown that estradiol (E₂) injected 48 h after P₄ can elevate attractivity and receptivity in the spring to levels similar to those observed during the fall (Billings and Katz, 1997). Attractivity refers to the stimulus value of the female toward the male, and receptivity refers to those behaviors performed by the female to permit intravaginal ejaculation (Beach, 1976). An experiment was conducted to determine whether CIDR treatment followed by E₂ injection would facilitate the display of attractivity and receptivity in goats. A threshold dose of E₂ (Billings and Katz, 1998) was used to allow detection of both facilitation and inhibition of E₂-induced sexual behaviors due to P₄ treatment. To limit confounding experimental treatments with variations in individual ovarian steroid secretion, ovx goats were

¹This study was supported by the New Jersey Agric. Exp. Sta., project #06137. The authors would like to thank Harold D. Hafs for advice regarding the use of the progesterone releasing devices.

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Received October 13, 1998.

Accepted March 15, 1999.

used. Injection of P₄ for 3 to 5 d was no more effective in inducing estrous behavior in ovx goats than only 2 d of P₄ injections (Kaplan and Katz, 1994). The duration of treatment with CIDR devices was 3 d in this study.

Materials and Methods

Animals and Housing

Seven ovx French-Alpine goats were housed in a 25-m² pen within a heated barn and had free access to a mixed grass pasture, except on the days of behavior tests, when they were restricted to the indoor pen. The diets of the goats were supplemented according to pasture conditions with grass hay and alfalfa and grain pellets. Mineral supplements and water were available for ad libitum consumption. All experiments were conducted as approved by the Rutgers University Institutional Review Board for the Care and Use of Animals.

Steroid Treatment

The goats were treated in a 5 × 5 Latin square design. In all cases, either a P₄-filled CIDR (Type G; InterAg, Hamilton, New Zealand) or a CIDR without P₄ (**BI-CIDR**) was inserted into the vagina of each goat and removed 72 h later. All E₂ treatments were s.c. injections of 30 μg of E₂ dissolved in 1 mL of 9:1 (vol/vol) corn oil:benzyl benzoate vehicle solution. The treatments were as follows: E₂ injected 48 h after CIDR removal (**48H**); E₂ injected 24 h after CIDR removal (**24H**); E₂ injected 24 h after removal of BI-CIDR (**E2**); vehicle injected 24 h after CIDR removal (**P4**); or vehicle injected 24 h after BI-CIDR removal (**VEH**). Each subsequent series of steroid treatments was begun at least 1 wk after the previous series of treatments to prevent carry-over effects of the treatments on sexual behavior. The experiment was repeated during the fall and spring. In the fall, the experiment was conducted from September 20 to November 21, 1996, when day lengths decreased from 12.3 to 9.8 h of light. The spring experiment was conducted from March 14 to May 15, 1997, when day lengths increased from 11.9 to 14.5 h of light.

Behavioral Observations

At 14 h and again at 17 h following injection with E₂, sexual behavior observations were conducted. These times corresponded to the predicted times when goats would most frequently display sexual behaviors (Billings and Katz, 1997). Two sexually experienced, testosterone-treated wethers were used for the sexual behavior observations. For each test, one wether was placed into the female's home pen for 30 min, and the order in which the two males were used in a given day

was randomized. During that time, the following sexual behaviors were recorded:

- anogenital sniffs: the nose contacts or is near the anus or genitalia of another goat.
- foreleg kicks: a straight-leg kick forward.
- neck-stretches: the male stretches his neck parallel to his back when approaching a female, often accompanied by a "gobble" vocalization.
- mount intention movements: both forelegs are lifted off the ground simultaneously but not high enough to permit mounting.
- mount attempts: forelegs are lifted high enough to permit mounting, but the female either runs away or the mount is interrupted in some way prior to completion, even if the female stands.
- mounts: forelegs and anterior portion of the body are lifted onto another goat.

Statistical Analyses

For each goat, attractivity and receptivity scores were calculated for each 30-min test as previously reported (Billings and Katz, 1997). Receptivity scores were calculated as the number of times each female stood to receive mounts or mount attempts. Attractivity scores were calculated as the number of times that each female received anogenital sniffs, foreleg kicks, neck-stretches, mount intention movements, or mount attempts for which the female did not stand. One goat that was supposed to receive vehicle was inadvertently injected with E₂, so the data for that week's behavior observation for that goat were excluded from analyses, although the goat was present during the observation period. Prior to analysis, attractivity and receptivity scores were transformed using the formula $\log(x + 1)$ to meet the requirements of a normal distribution for parametric tests. Attractivity and receptivity scores were then compared within and across seasons using a GLM procedure (SAS, 1988). The main effects were season, steroid treatment, time of test (14 h or 17 h), and goat. Time of test was not a significant effect, so data were collapsed over time. Significant ($P < .05$) main effects were then compared using Duncan's multiple range test (SAS, 1988). The number of receptive goats for the breeding and nonbreeding seasons was compared for each treatment using a two-sided Fischer's Exact Test (Steel and Torrie, 1980).

Progesterone Profiles

To determine the profile of P₄ in circulation following insertion of CIDR, a separate study was conducted in May 1996. The same seven ovx goats used in the behavior study were treated with either a CIDR (n = 4) or a BI-CIDR (n = 3), and blood samples were collected via jugular venipuncture. Blood collection was done just prior to insertion of the

CIDR, every 4 h for the first 24 h following insertion, and then every 8 h until removal of the CIDR at 72 h. Hourly blood samples were then collected for 10 h after CIDR removal. Serum was harvested and frozen until P₄ concentrations were determined with a RIA. The assay was performed as previously described (Billings and Katz, 1997). The assay sensitivity was .6 ng/mL. The intra- and interassay CV were 9.6 and 5.5%, and 11.1 and 2.2% for serum samples with high and low concentrations of P₄, respectively.

Results

During the fall and spring, both attractivity and receptivity scores were lower (*P* < .05) for goats receiving the P₄ or VEH treatment than their scores with the E₂ treatment (Figure 1). Following the E₂ treatment, goats had higher attractivity and receptivity scores in the fall than in the spring (*P* < .05). When CIDR were removed 24 h prior to E₂ injection (24H), E₂-induced attractivity was facilitated (*P* < .05) during both seasons and was greater in the fall than in the spring (*P* < .05). Receptivity scores were greater when the CIDR was removed 24 h prior to E₂ injection (24H) than when goats received the E₂ treatment (*P* < .05) during the spring, but not the fall; receptivity in the spring was similar to that during the fall for the 24H treatment. When a CIDR was removed 48 h prior to injection with E₂ (48H), both attractivity and receptivity were facilitated during the spring compared with responses to the E₂ treatment (*P* < .05). There was no facilitation of attractivity or receptivity during the fall following treatment with a CIDR removed 48 h prior to E₂ treatment. Again, both attractivity and receptivity in

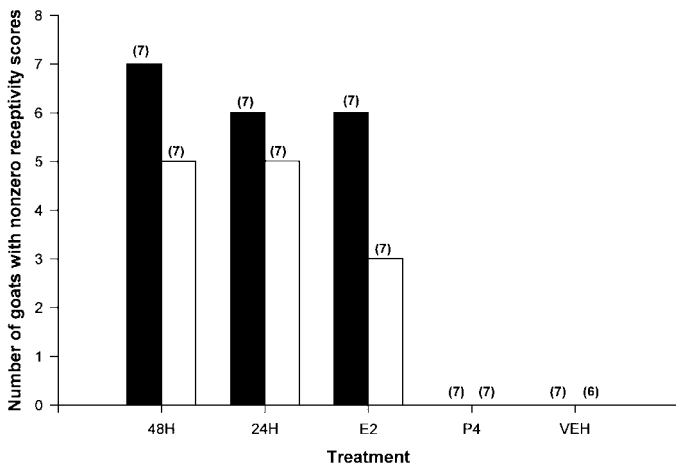


Figure 2. Number of goats for each treatment with nonzero receptivity scores in the fall (black bars) and spring (open bars). Numbers in parentheses indicate the number of goats included in each treatment for each season. The treatments are denoted as in Figure 1.

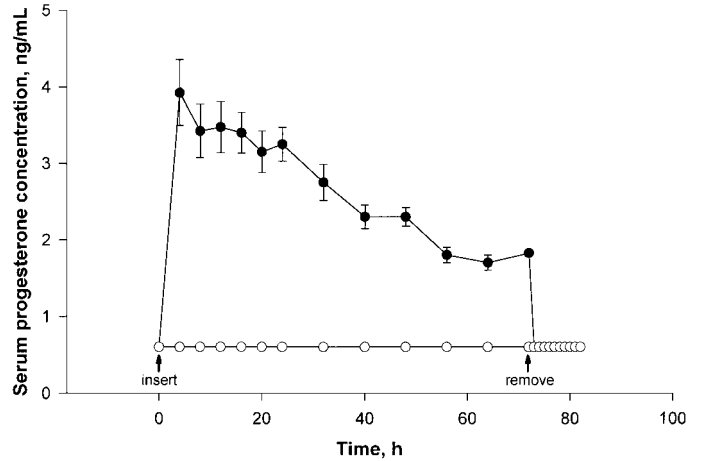


Figure 3. Mean serum progesterone (P₄) concentrations (± SEM) for ovariectomized goats treated with either a P₄-filled controlled intravaginal drug release device (CIDR; ●; n = 4) or a blank CIDR (○; n = 3) for 72 h.

the spring were restored to levels similar to those in the fall when the CIDR devices were removed 48 h prior to E₂ injection. There were no significant differences between the two seasons in the proportion of goats that were receptive for any of the treatments (Figure 2).

The retention rate for the CIDR devices was 100%. Treatment with CIDR resulted in serum P₄ concentrations that had reached a maximum level by 4 h after insertion of the CIDR (Figure 3). Concentrations of P₄ in circulation gradually declined during the 72-h period of treatment but remained > 1 ng/mL. By 1 h following removal of the CIDR, serum P₄ concentrations were undetectable in all goats.

Discussion

The seasonal difference in attractivity and receptivity scores for ovx goats treated with a BI-CIDR followed by E₂ injection is consistent with prior studies of seasonal influences on sexual behavior in goats (Billings and Katz, 1997). This indicates that vagino-cervical stimulation from treatment with CIDR did not alter the seasonal differences in E₂-induced sexual behavior. Even though none of the goats in this study was treated with E₂ in the absence of a CIDR to test the effects of vagino-cervical stimulation directly, both attractivity and receptivity scores for these goats were similar to those previously reported following treatment with E₂ alone (Billings and Katz, 1997). Mating shortens estrus in rats (Blandau et al., 1941). The abbreviation of estrus following mating has also been demonstrated to occur following vagino-cervical stimulation alone in guinea pigs (Goldfoot and Goy, 1970). Because determination of the full sexual

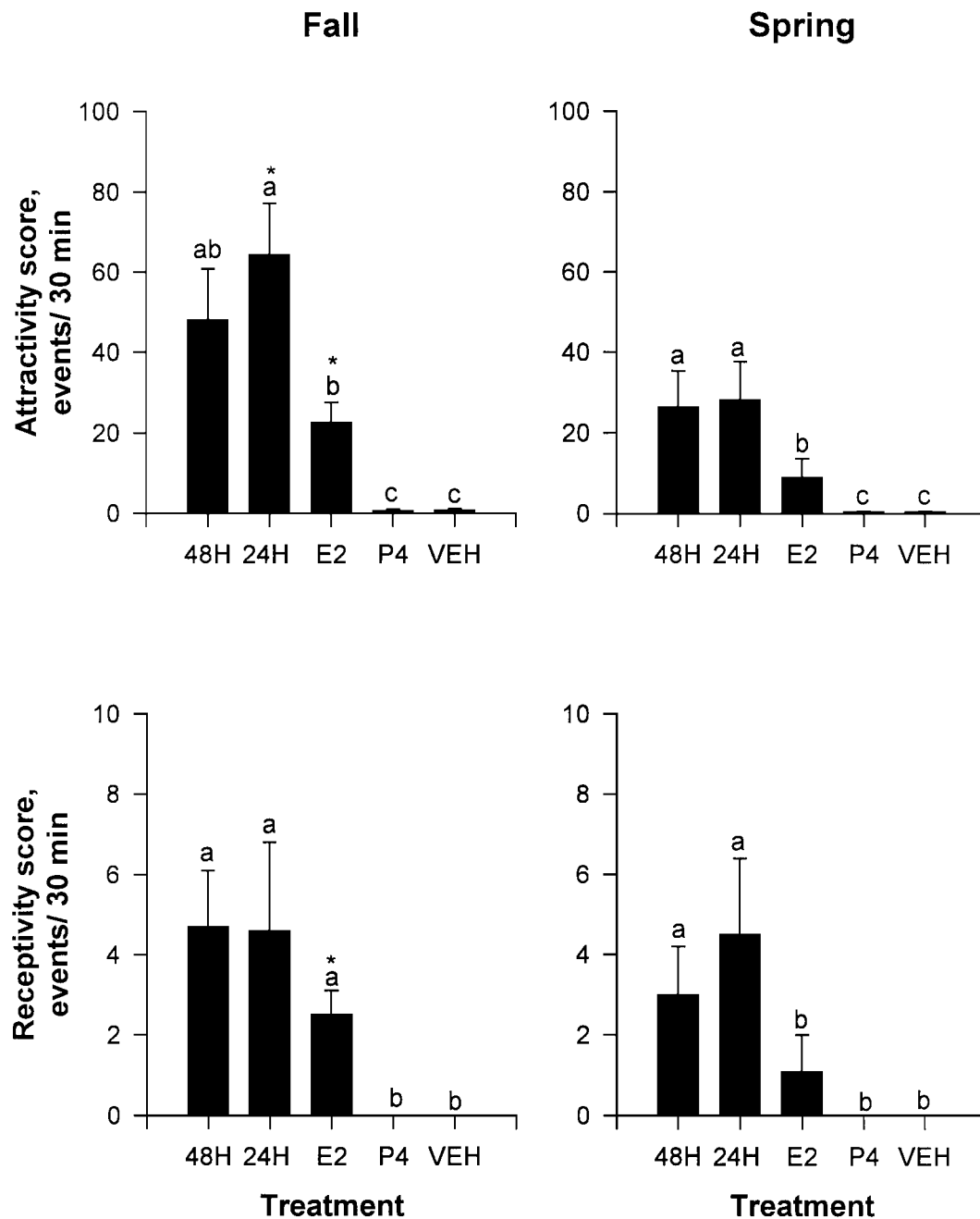


Figure 1. Mean attractivity and receptivity scores (\pm SEM) for ovariectomized goats during the fall and spring. Treatments with different letters differ ($P < .05$) within a season and behavior category. Asterisks indicate seasonal differences ($P < .05$) for a given treatment within a behavior category. Treatments are denoted as follows: 48H, progesterone (P_4)-filled controlled intravaginal drug release device (CIDR) removed 48 h before a 30- μ g estradiol (E_2) injection; 24H, P_4 -filled CIDR removed 24 h before E_2 injection; E2, blank CIDR removed 24 h before E_2 injection; P4, P_4 -filled CIDR removed 24 h before vehicle injection; VEH, blank CIDR removed 24 h before vehicle injection.

behavior repertoire of female goats requires the presence of a male, the potential of mating to abbreviate the duration of display of estrous behaviors was also a consideration. In sheep, infrequent (i.e., every 4 h) teasing with a ram resulted in longer durations of estrus than if the ram remained with the females for the full period of estrus (Parsons and

Hunter, 1967). However, even a single service abbreviated estrus duration in Nubian goats (Romano, 1994). Nonetheless, estrus duration following service was still greater than 24 h (Romano, 1993). Because the male goat was only placed with the females for two 30-min periods beginning 4 h apart on any given test day, it is not likely that estrous behavior would have

been terminated due to the presence of the male prior to the second observation of the day. Such termination of the display of estrous behavior within this time interval has not been observed in previous studies using similar methods (Kaplan and Katz, 1994; Billings and Katz, 1997). One further consideration with respect to using the males as stimulus animals is the potential effect of male fatigue or intermale variation on the behaviors displayed. To minimize these effects, two males were used as stimulus animals in this study. Because the same two males were used over the full time course of the study, and both were used in any given day, this minimized the amount of variation due to the individual males used. To minimize fatigue, one male was used in each of the two tests per day, such that the test conducted 14 h after E₂ injection used a different male than the test conducted 17 h after E₂ injection. Therefore, it is not likely that fatigue of the male goats used would account for any observed differences in sexual behavior in these studies.

Treatment with P₄-filled CIDR for 3 d facilitated both E₂-induced attractivity and receptivity during the spring, whether removed 24 or 48 h prior to E₂ injection. This contradicts previous reports that P₄ injections 24 h prior to E₂ injection inhibited sexual behavior during both the fall and spring (Billings and Katz, 1997). Most likely, this discrepancy is due to differing profiles of P₄ in circulation for the two different routes of administration. When 5 mg of P₄ was injected s.c., circulating concentrations of P₄ reached a maximum 5 h later and did not return to baseline levels until 9 h after injection (Billings and Katz, 1997). In this study, serum P₄ concentrations returned to undetectable levels within the 1st h after the CIDR were removed. This is a benefit of using the CIDR rather than injections, because the interval from removal of P₄ to the onset of estrous behavior can be more precisely determined. The time required for serum P₄ concentrations to return to baseline levels before E₂ was injected was longer when P₄ was administered using CIDR than when a subcutaneous injection of an oil-based solution was given.

Attractivity was also facilitated during the fall when the CIDR devices were removed 24 h prior to injection with E₂. Again, this was not observed when P₄ was injected in previous studies (Billings and Katz, 1997). This facilitation of attractivity during the breeding season by providing a 24-h interval between P₄ and E₂ treatment cannot be accounted for by considering only the interval between clearance of P₄ from circulation and the E₂ injection. It is likely that because the concentrations of P₄ in circulation over time mimicked those occurring during the natural estrous cycle, this resulted in the facilitation of attractivity. In intact goats, circulating P₄ concentrations remain high throughout the luteal phase and decline gradually as the corpus luteum begins to degenerate after approximately d 12 of the estrous

cycle (Harrison, 1948). However, P₄ concentrations do not begin to decline rapidly until approximately 3 d prior to the onset of estrus (Thorburn and Schneider, 1972; Pathak et al., 1990). Circulating concentrations of E₂ reach a maximum approximately 1 d prior to the onset of estrus (Pathak et al., 1990). Therefore, the steadily declining circulating concentrations of P₄ following insertion of the CIDR mimics the profile of circulating P₄ concentrations in intact goats from approximately 4 d prior to the onset of estrus. This, in combination with injection of 30 μg of E₂ 24 h after removing the CIDR, results in steroid hormone profiles very similar to those observed during the estrous cycle of intact goats just prior to and during estrus. Therefore, that this treatment would result in the greatest display of sexual behavior is consistent with the endocrine events leading to the display of estrous behavior in intact goats. In the fall, attractivity scores when CIDR were removed 48 h prior to E₂ were not different from those following treatment with either E₂ alone or when CIDR were removed 24 h prior to E₂ injection. This shows that attractivity decreases as the interval between P₄ and E₂ treatment becomes greater than 24 h.

Even though Hanlon et al. (1996) did not study attractivity, they did report that estradiol benzoate injected 24 h after removal of CIDR increased the number of cattle displaying estrous behavior, and conception rates were similar with or without E₂. This suggests that the use of CIDR may also be a practical means of synchronizing estrus in intact goats while improving estrus detection. Estradiol-induced receptivity was not facilitated by treatment with CIDR during the fall. However, six out of seven goats were receptive following treatment with E₂ alone. There are several possible explanations for this finding. Either the receptivity scores were already approaching maximal levels in response to E₂ alone or the receptivity scores were too low to achieve statistical significance. Also, the number of goats in the study may have been too small to detect differences. Although the number of goats that were receptive for each treatment was always less during the spring than during the fall, this was not a statistically significant difference. Again, this may have been due to the small sample size. This seasonal difference may still be of practical importance when considering the effectiveness of this treatment during the nonbreeding season.

That treatment with P₄ for only 3 d facilitated E₂-induced attractivity during both the fall and spring, and receptivity during the spring further demonstrates the functional significance of an initial silent estrus at the onset of the breeding season (Cole and Miller, 1933), typically followed by a short estrous cycle (Camp et al., 1983). In goats experiencing short estrous cycles, corpora lutea present on d 1 of the estrous cycle were no longer observed by d 5 (Camp et al., 1983). This indicates that induction of sexual behavior following only 3 d of treatment with P₄ is

physiologically relevant to the initial estrous cycle of the breeding season in goats. This similarity between P_4 concentrations following treatment with a CIDR and that during the luteal phase of the initial estrous cycle suggests that this treatment may be effective for increasing the display of sexual behavior accompanying the first ovulation of the breeding season.

Implications

Steroid hormone profiles following treatment with a controlled intravaginal drug release device removed 24 h before injection of 30 μg of estradiol are similar to those occurring naturally at the end of the luteal phase through estrus. Therefore, this treatment can potentially be used both as an ideal model for further study of estrous behavior in goats and as a useful means of enhancing estrus detection during synchronization of estrus for breeding purposes.

Literature Cited

- Beach, F. A. 1976. Sexual attractivity, proceptivity, and receptivity in female mammals. *Horm. Behav.* 7:105–138.
- Billings, H. J., and L. S. Katz. 1997. Progesterone facilitation and inhibition of estradiol-induced sexual behavior in the female goat. *Horm. Behav.* 31:47–53.
- Billings, H. J., and L. S. Katz. 1998. Threshold dose of estradiol for inducing sexual receptivity in ovariectomized French-Alpine goats. *Appl. Anim. Behav. Sci.* 57:109–115.
- Blandau, R. J., J. L. Boling, and W. C. Young. 1941. The length of heat in the albino rat as determined by the copulatory response. *Anat. Rec.* 79:453–463.
- Camp, J. C., D. E. Wildt, P. K. Howard, L. D. Stuart, and P. K. Chakraborty. 1983. Ovarian activity during normal and abnormal length estrous cycles in the goat. *Biol. Reprod.* 28:673–681.
- Cole, H. H., and R. F. Miller. 1933. Artificial induction of ovulation and oestrus in the ewe during anoestrus. *Am. J. Physiol.* 104:165–171.
- Goldfoot, D. A., and R. W. Goy. 1970. Abbreviation of behavioral estrus in guinea pigs by coital and vagino-cervical stimulation. *J. Comp. Physiol. Psychol.* 72:426–434.
- Hanlon, D. W., N. B. Williamson, J. J. Wichtel, I. J. Steffert, A. L. Craigie, and D. U. Pfeiffer. 1996. The effect of estradiol benzoate administration on estrous response and synchronized pregnancy rates in dairy heifers after treatment with exogenous progesterone. *Theriogenology* 45:775–785.
- Harrison, R. J. 1948. The changes occurring in the ovary of the goat during the estrous cycle and in early pregnancy. *J. Anat.* 82:21–48.
- Jainudeen, M. R., and E.S.E. Hafez. 1993. Sheep and goats. In: E.S.E. Hafez (Ed.) *Reproduction in Farm Animals* (6th Ed.), pp 330–342. Lea & Febiger, Philadelphia, PA.
- Kaplan, D. H., and L. S. Katz. 1994. Exposure to constant photoperiod alters serum prolactin concentrations and behavioral response to estradiol in the ovariectomized goat. *J. Anim. Sci.* 72:3088–3097.
- Parsons, S. D., and G. L. Hunter. 1967. Effect of the ram on duration of oestrus in the ewe. *J. Reprod. Fertil.* 14:61–70.
- Pathak, M. M., A. V. Patel, R. S. Jaiswal, and V. M. Mehta. 1990. Circulating levels of progesterone and oestrogen in cyclic goats. *Ind. J. Anim. Sci.* 60:836–837.
- Ritar, A. J., P. D. Ball, and P. J. O'May. 1990. Artificial insemination of Cashmere goats: Effects on fertility and fecundity of intravaginal treatment, method and time of insemination, semen freezing process, number of motile spermatozoa and age of females. *Reprod. Fertil. Dev.* 2:377–384.
- Ritar, A. J., S. Salamon, P. D. Ball, and P. J. O'May. 1989. Ovulation and fertility in goats after intravaginal device-PMSG treatment. *Small Ruminant Res.* 2:323–331.
- Robinson, T. J. 1959. The estrous cycle of the ewe and doe. In: H. H. Cole and P. T. Cupps (Ed.) *Reproduction in Domestic Animals*, Volume 1, pp 291–333. Academic Press, New York.
- Romano, J. E. 1993. Effect of service on estrus duration in dairy goats. *Theriogenology* 40:77–84.
- Romano, J. E. 1994. Effect of service number on estrus duration in dairy goats. *Theriogenology* 41:1273–1277.
- SAS. 1988. *SAS User's Guide*. SAS Inst. Inc., Cary, NC.
- Steel, R.G.D., and J. H. Torrie. 1980. *Principles and Procedures of Statistics: A Biometrical Approach*. McGraw-Hill Publishing Co., New York.
- Thorburn, G. D., and W. Schneider. 1972. The progesterone concentration in the plasma of the goat during the oestrus cycle and pregnancy. *J. Endocrinol.* 52:23–36.

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