Influence of castration and estrogen replacement on sexual behavior of female-oriented, male-oriented, and asexual rams1,2

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ABSTRACT: An experiment was conducted to determine whether exogenous estradiol-17β (E2) could restore sexual behavior in castrated rams. The protocol consisted of three sequential 6-wk periods during which rams were studied while 1) intact, 2) bilaterally castrated, or 3) implanted s.c. with two 7.6-cm silastic implants each containing 309 µg. Rams (classified as female-oriented [FOR, n = 7], male-oriented [MOR, n = 7], or asexual [n = 7]) were subjected to 30-min sexual behavior tests every 2 wk during the ensuing 18 wk. Rams were observed for mounts and ejaculations using two ovariolectomized, estrous ewes and two intact males secured in stanchions. Behavioral data were analyzed using the signed rank test, but asexual rams showed no sexual behavior and therefore were not evaluated statistically. Jugular blood was collected prior to castration and at the end of the 18-wk period, and testicular venous (n = 21) and arterial (n = 8) bloods were collected immediately prior to castration. Radioimmunoassay was used to quantify systemic levels of estrone (E1), E2, and testosterone (T) and testicular serum concentrations of oxytocin (OT). Mounting behavior of MOR and FOR declined after castration (P < .05 and P < .10, respectively). Castration reduced the number of ejaculations by FOR (P < .05), but not by MOR (P > .10). Mounting behavior of castrated MOR and FOR was not affected by E2 treatment relative to that observed if castrated only (P > .10). Treatment of asexual rams with E2 did not stimulate sexual behavior in these rams. There were no marked differences (P > .10) among ram groups with regard to serum concentrations of E1, E2, or T prior to castration (overall mean ± SE, 12.8 ± 7.6 ± .5, and 2,670 ± 780 pg/mL, respectively) or any difference (P > .10) in systemic concentration of E1 or E2 among ram groups after rams were implanted with E2 (overall mean ± SE, 9.7 ± 7.0 ± .7 pg/mL, respectively). Serum concentrations of E2 after implantation of the steroid did not differ from those present while rams were intact (P > .10). Testicular venous and arterial serum concentrations of OT were low and did not differ within or between rams. These results suggest that restoration of E2 concentrations to physiological levels in castrated adult rams (regardless of sexual orientation) cannot stimulate or reestablish sexual behaviors to levels observed prior to castration.

Key Words: Castration, Estrogens, Oxytocin, Rams, Sexual Behavior

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Introduction

Male-oriented rams (MOR) exclusively seek out rams for sexual partners in the presence of estrous females, whereas asexual rams do not display sexual behavior (Perkins and Fitzgerald, 1992). The MOR have fewer estradiol (E2) receptors in the amygdala (Alexander et al., 1993) and lower systemic levels of E2 and estrone than female-oriented rams (FOR; Resko et al., 1996). Thus, the brains of MOR may not be exposed to the same levels of E2 as those of FOR. If this is true, then E2 therapy might alter their sexual behavior to the point of being more like that of FOR. To test such a premise requires the use of castrated rams, which would also permit assessment of effects of castration on sexual behavior. Castration would be expected to reduce sexual behavior of FOR and MOR, possibly to the same degree as has been reported to occur after castration of guinea pigs exhibiting high to low sexual aggressiveness (Grunt and Young, 1953). Further, injection of pharmacological dosages of E2 have been effective in stimulating or restoring sexual behavior of steers.

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were fed approximately 1.6 kg long stem hay and simulated natural seasonal lighting conditions. Rams were individually isolated in indoor pens until 1 wk prior to behavioral testing. At this time, all rams were housed in outdoor pens in their respective groups until 1 wk prior to behavioral testing. At this time, animals were individually isolated in indoor pens under simulated natural seasonal lighting conditions. Rams were fed approximately 1.6 kg long stem hay and had ad libitum access to a mineral salt block and water. All experimental procedures and surgeries were performed in accordance with the Institutional Animal Care and Use Committee at the U.S. Sheep Experiment Station in Dubois, Idaho.

The investigation period was 18 wk in duration (October 30, 1992) were classified as 1) female-oriented (n = 7), 2) male-oriented (n = 7), or 3) asexual (n = 7). All rams were housed in outdoor pens in their respective groups until 1 wk prior to behavioral testing. At this time, animals were individually isolated in indoor pens under simulated natural seasonal lighting conditions. Rams were fed approximately 1.6 kg long stem hay and had ad libitum access to a mineral salt block and water. All experimental procedures and surgeries were performed in accordance with the Institutional Animal Care and Use Committee at the U.S. Sheep Experiment Station in Dubois, Idaho.

**Materials and Methods**

**Rams.** Twenty-one mature rams previously tested for sexual behavior as described by Perkins and Fitzgerald (1992) were classified as 1) female-oriented (n = 7), 2) male-oriented (n = 7), or 3) asexual (n = 7). All rams were housed in outdoor pens in their respective groups until 1 wk prior to behavioral testing. At this time, animals were individually isolated in indoor pens under simulated natural seasonal lighting conditions. Rams were fed approximately 1.6 kg long stem hay and had ad libitum access to a mineral salt block and water. All experimental procedures and surgeries were performed in accordance with the Institutional Animal Care and Use Committee at the U.S. Sheep Experiment Station in Dubois, Idaho.

**Teaser Animals.** Four mature (two Targhee and two Rambouillet), ovariectomized (OVX) ewes weighing approximately 64 kg each received a vaginal pessary containing 17α-hydroxy-6α-methylpregn-4-en-3-one-17α-acetate (MAP; Tuco Products Limited, Orangeville, Ontario, Canada) that remained in place for 13 d. Pessaries were removed on d 13, and each ewe received an i.m. injection of estradiol benzoate (50 μg in corn oil) to induce behavioral estrus for teasing purposes. Four mature, intact rams (of weight and body size similar to teaser ewes) were also employed as stimulus teasers during sexual behavior tests.

**Experimental Treatments.** The investigation period of this experiment was 18 wk in duration (October 30, 1996, to March 4, 1997) and consisted of three sequential 6 wk periods when rams were 1) intact, 2) bilaterally castrated, and 3) implanted s.c. with estradiol-17β (E2). During the intact period, rams were tested for behavior every 2 wk for a total of three observations per animal beginning on November 12, 1996. One week prior to sexual preference testing, rams were isolated in individual pens to prevent any visual or sexual contact with penmates. Rams were evaluated by group on an alternating basis allowing for each classification to have first access to teasers during successive test sessions. Each behavioral test session consisted of a 30-min observation period during which two test subjects of a particular classification entered one of two private (52 × 78 m) indoor arenas with unrestrained access to two OVX, MAP-primed, E2-treated ewes and two intact rams secured in stanchions. Rams yet to be tested were not able to view the activity in the test arena or other rams in waiting. After completion of the behavior test, each ram was immediately returned to its respective outdoor pen to prevent contact with other rams awaiting evaluation. Observations with regard to sexual behavior (mounts and ejaculations upon intromission) of the test subjects and the sex of the stimulus animal toward which the behavior was directed were recorded by an observer. Other behavioral traits (sniffs, foreleg kicks, flehmen response) have been recorded in some studies of rams. However, for this study, mounting and ejaculation were considered the most important traits measurable that would reflect the ram’s response to treatment.

The second phase of the experiment was conducted after all rams were bilaterally castrated. Prior to castration and while individually isolated, a jugular blood sample was drawn from each ram by venipuncture and collected into a 5-mL vacutainer tube, and the blood was allowed to clot for 2 h at room temperature. All samples were then stored at 4°C overnight, centrifuged at 2,000 × g the following day, and sera were harvested and stored at −20°C to be later extracted and analyzed by radioimmunoassay (RIA) for estrogen and testosterone (T) concentrations. For the castration procedure, which was performed 1 d after sexual behavior testing, rams were anesthetized with an i.v. injection of .5 mL diazepam and 2.25 mL ketamine. Testes were exposed prior to orchidectomy through bilateral proximal scrotal incisions, at which time testicular venous blood, proximal to the pampiniform plexus (n = 21), and arterial blood distal to the plexus in eight rams chosen randomly was collected into 5-mL vacutainer tubes. Samples were allowed to clot for 2 h at room temperature, then stored at 4°C overnight. The following day, samples were centrifuged as described above, and the sera were harvested and stored at −20°C until they were extracted and subjected to RIA for oxytocin (OT). During recovery from surgery, all rams were given 1 g of oral phenylbutazone and an i.m. injection of both 6 mL of LA-200 (200 mg/mL oxytetracycline, wt/vol: 40% 2-pyrrolidone, 5% povidone, 1.8% magnesium oxide, 2% sodium formaldehyde sulfoxylate and monoethanolamine and/or HCl; Pfizer, Exton, PA) and 5 mL of Phytadione (10 mg/mL of vitamin K; Vedco Inc., St. Joseph, MO). Rams were then returned to their respective groups in outdoor pens for further recovery preceding isolation and the first sexual behavior test. Sexual behavioral testing of castrated rams was conducted every 2 wk as previously described (first test December 24, 1996), for a total of three observations per animal, at which time all rams received s.c. E2 implants. Implants were prepared by a modification of procedures described by Karsch et al. (1973).
**Preliminary Estradiol Trial.** In order to determine the quantity of E2 to implant a preliminary trial was conducted in which six mature ewes, having been OVX for a period of 2 wk, were implanted s.c. with a multiple number of silastic medical-grade implants containing either 309 or 178 mg of E2. Implants were soaked overnight in tap water to equalize the amount of hormone in the implant wall, then dried and stored in an airtight container at 4°C until they were used. Two ewes each received the following number of implants, which were left in for 16 d: two large implants (each 309 mg of E2), three small implants (each 178 mg of E2), or five small implants (each 178 mg of E2). On d 16 a jugular blood sample was collected and the resulting serum was analyzed for E2 as described below. It was determined that two large implants (3.12 mm i.d. × 7.6 cm) resulted in a serum concentration of E2 (mean ± SE, 21.5 ± 3.0 pg/mL) that most closely approximated the mean serum levels of E2 in heterosexual rams reported by Resko et al. (1996).

**Estradiol Treatment of Rams.** Rams were implanted with E2 (309 ± 16 mg/implant) in the axillary region of the foreleg following guidelines implemented by Karsch et al. (1980). Following a general cleansing of the region, skin was scrubbed with 70% ethanol and lavaged with a betadine solution. The incision site was liberally injected s.c. with .25-mL increments of lidocaine and an approximately 2- to 3-cm incision was made with surgical scissors. Hemostats were inserted into the incision and opened to create a subcutaneous pouch into which two silastic implants packed with crystalline E2 (pre-soaked for 1 h in 70% ethanol) were placed. Incisions were closed with surgical wound clips and sprayed topically with Furasone. Rams were then returned to their respective groups until they were isolated and tested for sexual behaviors as described above every 2 wk for a total of three observations per animal. At the conclusion of this final 6-wk period while rams were individually isolated (preceding the final behavioral test), jugular blood samples from each ram were collected into 5-mL vacutainer tubes. Blood samples were processed and the sera stored as described above until they were assayed for estrogen concentration.

**Hormone Analyses**

**Oxytocin Radioimmunoassay.** Oxytocin in testicular serum samples (1 mL) was extracted and quantified as described by Orwig et al. (1994) as adapted from Abdelgadir et al. (1987) and Schams (1983) by chromatography using Sep-Pak Plus C18 cartridges (Waters Chromatography Division, Millipore Corp., Milford, MA). Mean extraction efficiency was 53.3 ± 6.0%. All sample values of OT were adjusted for recovery. Radioimmunoassay was performed for OT standards (.12 to 32 pg/tube) or extracted samples (100 μL in duplicate) using OT antibody generously provided by Dieter Schams (Technical Univ. of Munich, Freising-Weihenstephan, Germany) at a final dilution of 1:2,000. Intra- and interassay coefficients of variation were 3.9 and 17.9 ± 8%, respectively. Sensitivity of the OT assay was .12 pg/tube.

**Steroid Radioimmunoassay.** Steroids were extracted from .5 mL (for T) or 1 mL (for estrone [E1] and E2) of serum diluted 1:1 with double-distilled water using 6 to 7 mL of diethyl ether. An additional four to six random samples of serum with 10,000 cpm of [3H]estradiol or T added were included with each set of samples extracted to estimate efficiency of steroid recovery. Samples were centrifuged at 1,000 × g at 4°C for 15 min then snap-frozen in a methanol-Dry Ice bath. Ether extracts were quickly poured into 12- × 75-mm borosilicate tubes and evaporated on a heating block at 37°C under air. Tubes were reconcentrated two times with 200-μL aliquots of diethyl ether and redried as stated above. Extracts were reconstituted with 100 μL hexane:benzene:methanol combination solvent (62:20:13 for estrogens or 85:15:5 for T), then subsequently subjected to chromatographic separation on glass columns (14 × .92 cm i.d.) packed with Sephadex LH-20 (1 g for estrogens; 2.5 g for T; Pharmacia, Piscataway, NJ). After application of the hormone extracts to the columns, the tubes were rinsed with an additional 100 μL of the combination solvent, which was added to the column. Steroids were separated on the columns with the combination solvent mentioned above. Estrone fractions (3 mL collected after elution with 6 mL), E2 fractions (5 mL collected after elution with 10 mL), and T fractions (8.5 mL after elution with 24 mL) were collected. Extracts were evaporated as previously described and reconstituted with 1 mL of ethanol. The E2, E1, and T antisera used for RIA were produced as described by Resko et al. (1973, 1975). Extraction efficiencies for E2, E1, and T were 65, 78.7, and 73.1%, respectively, and were used to adjust sample values accordingly.

Serum concentrations of E2, E1, and T were quantified in a single RIA for each steroid as described by Resko et al. (1980). One hundred microliters of diluted antiserum in .1% gelatin phosphate-buffered saline (GPBS) and 4,500 cpm/100 μL of [3H]-E2 (95.3 Ci/mmol; DuPont NEN, Boston, MA), E1 (85 Ci/mmol), or T (92.4 Ci/mmol) in .1% GPBS were added to the appropriate tubes. Separation of bound from free steroid was achieved by use of dextran-charcoal (2.5 g/L washed neutral norit charcoal [Fisher Scientific, Pittsburgh, PA]; .25 g/L Dextran T-70 [Pharmacia, Uppsala, Sweden]) in phosphate-buffered saline. Intrassay coefficients of variation were .63, 6, and 4.6% for E2, E1, and T, respectively. Sensitivities of the assays for E2, E1, and T were 2.5, 5 and 5 pg/tube, respectively.

**Statistical Analysis**

Because behavioral data for mounts and ejaculations were discrete they were analyzed using the Wilcoxon signed rank test (SAS, 1996); however, for comparative purposes these data are presented as means ± SE. Be-
havioral data for each MOR and FOR collected at 2-wk intervals within each 6-wk treatment period were summed for the analysis by the signed rank test. Respective sums for observed mounts and ejaculations after rams were castrated and when treated with E2 were compared to data collected while respective animals were intact. Similarly, data collected for each ram type after castration were compared to those collected after treatment with E2. Therefore, for MOR and FOR, there were three comparisons made of behavior data: intact vs castrated; intact vs castrated + E2; and castrated vs castrated + E2. Asexual rams consistently showed no sexual behavior and therefore were not evaluated statistically. Differences in testicular venous serum OT and systemic serum concentrations of T among FOR, MOR, and asexual rams were determined by one-way ANOVA (n = 21). Testicular arterial blood samples were collected from each of eight randomly chosen rams only. Therefore, a paired t-test was used to test for significance of testicular veno-arterial differences in serum concentrations of OT in these rams.

Differences among serum concentrations of E1 and E2 in intact and castrated rams implanted with E2 were analyzed by repeated measures ANOVA using the general linear model procedures of SAS (1996). Sources of variation for the statistical analysis of data for each hormone were group (FOR, MOR, asexual), treatment (intact, castrated plus E2), and group × treatment. Group differences among mean sera concentrations of E1 and E2 during the intact period and after castration plus exogenous E2 replacement were determined by the contrast procedure of SAS.

Results

Mounting behavior of the intact rams was consistent with their predetermined classification as originally described by Perkins and Fitzgerald (1992). Mounting behavior declined after castration of MOR (P < .05) and FOR (P < .10; Figure 1). Castration reduced the number of ejaculations by FOR (P < .05), but not by MOR (P > .10; Table 1). Treatment of castrated MOR and FOR with E2 failed to prevent the decline in number of mounts (Figure 1) and ejaculations compared to those observed while rams were intact (mounts: FOR, P < .10; MOR, P < .05; ejaculations: FOR, P < .05) or after castration (mounts and ejaculations for both types of rams, P > .10). Asexual rams consistently displayed no sexual behaviors over the entire duration of the investigation.

Endogenous serum E1, E2, and T concentrations were not significantly different among intact FOR, MOR, and asexual rams (P > .10; Table 2). Serum E1 and E2 concentrations following castration and exogenous E2 replacement, likewise, did not differ significantly among FOR, MOR, and asexual rams (P > .50 and P > .30, respectively; Table 2). Treatment (intact or castrated + E2 replacement) had no significant effect on serum concentrations of E2 among groups before or after castration with exogenous E2 therapy. However, mean serum concentrations of E1 were lower in castrated rams administered exogenous E2 than in the same animals before castration (P < .02; Table 2).

Mean testicular venous sera OT concentrations were not significantly different among FOR, MOR, and asexual rams (117.6 ± 11.7, 128.6 ± 21.1, and 102.9 ± 20.1 pg/mL, respectively; P > .10; n = 21). There was no evidence that the testes of the ram synthesized OT, because the overall mean testicular venous serum OT concentration (96.3 ± 9.4 pg/mL) did not differ significantly from overall mean testicular arterial serum concentration of OT (106.3 ± 17.5 pg/mL, P > .25; n = 8).

Discussion

Typically, following castration an animal’s sexual desire is quelled and a reduction in libido is experienced due to the lack of the stimulatory effects of T at the level of the hypothalamus. In the male, T secreted by the testes is aromatized to E2 in the hypothalamus,

![Image](http://example.com/image)

**Figure 1.** Mean (± SE) mounting behavior per female-oriented (FOR) and male-oriented (MOR) ram observed in intact and castrated rams and castrated rams receiving estradiol (E2) replacement. Asexual rams were consistently inactive (no mounts) and thus no data are depicted. Castrated or castrated plus E2 vs intact (*P < .05; †P < .10).

<table>
<thead>
<tr>
<th>Ram group</th>
<th>Intact (1–6 wk)</th>
<th>Castrated (7–12 wk)</th>
<th>Castrated + E2 (13–18 wk)</th>
</tr>
</thead>
<tbody>
<tr>
<td>FOR*</td>
<td>14.5 ± 1.1</td>
<td>4.3 ± 1.1d</td>
<td>2.8 ± .5d</td>
</tr>
<tr>
<td>MOR†</td>
<td>1.9 ± .8</td>
<td>.14 ± .1</td>
<td>.0 ± .0</td>
</tr>
<tr>
<td>Asexual</td>
<td>.0 ± .0</td>
<td>.0 ± .0</td>
<td>.0 ± .0</td>
</tr>
</tbody>
</table>

*aMeans of three sexual behavior tests per group during each 6-wk period.
*bFemale-oriented.
†Male-oriented.

dMeans differ from means of intact rams (P < .05).
Table 2. Mean (± SE) serum concentrations of estrogens and testosterone in female-oriented (FOR), male-oriented (MOR), and asexual intact rams or castrated rams receiving estradiol (E$_2$) replacement

<table>
<thead>
<tr>
<th>Ram group</th>
<th>Estradiol, pg/mL</th>
<th>Estrone, pg/mL</th>
<th>Testosterone, ng/mL</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Intact</td>
<td>Castrated +E$_2$</td>
<td>Intact</td>
</tr>
<tr>
<td>FOR</td>
<td>8.48 ± .79</td>
<td>10.4 ± 1.30</td>
<td>14.1 ± 1.70</td>
</tr>
<tr>
<td>MOR</td>
<td>7.57 ± .65</td>
<td>8.24 ± .67</td>
<td>12.8 ± 2.00</td>
</tr>
<tr>
<td>Asexual</td>
<td>6.67 ± .77</td>
<td>8.32 ± 1.10</td>
<td>11.6 ± .93</td>
</tr>
</tbody>
</table>

$^a$Differs from means for intact rams (P < .02).

or more specifically the medial preoptic area, which is thought to facilitate the maintenance of sexual behavior in some male mammalian and avian species (Beyer et al., 1976; Balthazart et al., 1992). As an example, Parrott and Baldwin (1984) demonstrated that testosterone propionate (TP) implanted into the medial preoptic area stimulated courtship and mounting behavior in 15-mo-old wethers. When TP implants were augmented with dihydrotestosterone (DHT) propionate injections, an increase in erections was also observed. Hence, it is not surprising that mounting behavior was reduced in MOR after castration and that FOR showed a similar reduction in mounting behavior. Perhaps of more significance is the fact that castration virtually eliminated mounting behavior in MOR while only moderately reducing this activity in FOR. These data are interpreted to suggest that MOR may indeed differ from FOR in some functional aspect of the central nervous system that affects sexual behavior. As discussed below, serum concentrations of gonadal steroids did not differ between intact MOR and FOR. Had the interval between castration and exogenous E$_2$ replacement been extended, the reduction in mounting behavior witnessed during behavioral testing may have been even more significant. It has also been shown that with an increase in the interval between castration and exogenous hormone replacement, there is a reduction in the responsiveness of the system to hormonal restoration (Clark et al., 1995); consequently, it was felt the 6 wk between castration and E$_2$ replacement would be optimal to observe the effects of endogenous hormone removal and also to observe more immediate effects of exogenous E$_2$ on sexual behavior in rams. Pharmacological administration of T, dihydrotestosterone (DHT) in combination with E$_2$, or E$_2$ alone have been effective in stimulating or restoring male sexual behaviors in species such as cattle (Dykeman et al., 1982) and sheep (Clegg et al., 1969; Parrott, 1978, D’Ochio and Brooks, 1980), depending on the stage in life during which the animals were castrated (i.e., castration before or after puberty). Treatment of 2-yr-old castrated rams with implants of T and E$_2$ for 6 wk was sufficient to stimulate mounting of estrous ewes by these males (D’Ochio et al., 1985). D’Ochio and Brooks (1980) found that mounting behavior could be stimulated in rams castrated prepuber-
tally when they were implanted s.c. with pharmacological concentrations of E$_2$. Further, rams castrated before puberty and injected with pharmacological concentrations of either TP or DHT plus E$_2$ responded by exhibiting a complete mating response. These same investigators found that rams castrated after puberty exhibited an increase in mounting behavior when treated with pharmacological injections of E$_2$ alone but failed to respond when administered DHT. With regard to ejaculation, castration of rams in our study was sufficient to drastically reduce the “ejaculatory” capacity of FOR. However, ejaculatory intromissions achieved by MOR did not seem significantly reduced following castration due to the fact that MOR rarely experienced ejaculation prior to castration. This was due, in part, to the fact that the angle and anatomy of MORs’ preferred sexual partner, the teaser ram, was not conducive to intromission, thus precluding successful ejaculatory attempts by MOR the majority of the time. Accordingly, little difference could be observed with regard to MOR ejaculatory intromissions during the intact, castrated, or castrated plus E$_2$ treatment periods.

Systemic concentrations of E$_2$ and T were not significantly different among groups of intact rams. This was unexpected, because Resko et al. (1996) detected a significant reduction in systemic concentrations of these steroids between FOR and MOR (15 vs 8 pg/mL for E$_2$ and 1.6 vs .8 ng/mL for T). In the present study, mean serum concentrations of E$_2$ in MOR were similar to those reported by Resko et al. (1996) but levels of T in rams of the current experiment were greater than those detected by the latter investigators. Similarly, Alexander et al. (1999) also could not detect a difference in systemic serum concentrations of T between MOR and FOR. Failure to detect a difference in systemic serum concentrations of T in MOR and FOR in the present study compared to those previously reported by Resko et al. (1996) may be due to the fact that blood was collected from anesthetized animals in the latter study. Recently, Resko et al. (1999) found that FOR, MOR, and asexual rams did not differ in systemic serum concentrations of T while conscious but that anesthesia resulted in a significant reduction in serum levels of T in MOR and asexual rams compared to those in FOR. The basis for the detected response to anesthesia is
unknown but certainly is an area that warrants further investigation.

Analysis of serum E\textsubscript{2} concentrations before and after castration with E\textsubscript{2} replacement did not result in a significant group \times treatment interaction, suggesting that sufficient E\textsubscript{2} had been administered to mimic the systemic concentrations of E\textsubscript{2} normally present in intact rams. In spite of physiological E\textsubscript{2} replacement in castrated rams, the treatment was not sufficient to restore, or in the case of asexual rams, to stimulate, sexual behavior. It is possible that adequate quantities of E\textsubscript{2} at the cellular level in the brain were not available to facilitate or promote appropriate sexual behavior in the ram. It is conceivable that there are other neurological components involved in promoting expression of sexual behavior by rams besides the response to steroid hormone feedback. This possibility is supported by the fact that castration markedly reduced mounting behavior in MOR but only slightly reduced this behavior in castrated FOR.

Interestingly, systemic E\textsubscript{1} concentrations did not differ significantly among groups while rams were intact or after castration plus E\textsubscript{2} treatment. However, systemic concentrations of E\textsubscript{1} differed significantly within group between treatments (intact vs castrated plus E\textsubscript{2}). Concentrations of this estrogen were reduced in all ram groups after castration with E\textsubscript{2} replacement compared with the same rams while intact. The observed differences in serum concentration of E\textsubscript{1} may be due to removal of the testes, which eliminated endogenous androgens that ordinarily give rise to E\textsubscript{1}.

In males, endogenous OT promotes penile erection, hastens onset of initial ejaculation, regulates number of ejaculations in some species, and may be involved in mating pair-bond formation (Carter, 1992). Oxytocin synthesized within the testes may act locally on myoid cells surrounding the seminiferous tubules to promote sperm transport to the epididymis and stimulate smooth muscle contraction in the male tract (Nicholson and Jenkin, 1995). It has been suggested by Knickerbocker et al. (1988) that the testes of the ram might be a site of OT synthesis. Hence, testicular venous and arterial OT concentrations were evaluated in eight of the animals chosen at random to determine whether the testes of the ram synthesized OT. It was expected that if the testes of the ram synthesized OT, testicular venous serum concentrations of OT would exceed OT concentrations in testicular arterial serum. However, there was no significant difference between OT concentrations in the testicular venous and arterial sera in the rams, thus suggesting that the testes of the ram do not synthesize OT. However, testicular synthesis of OT cannot be completely ruled out because testicular venous and arterial bloods were collected during castration on the day after sexual behavior testing. It is possible that testicular venous and arterial bloods sampled immediately following sexual stimulation might have revealed that OT is synthesized by the testes of the ram as a direct response to sexual stimuli.

In conclusion, these results suggest that restoration of E\textsubscript{2} concentrations in the systemic circulation of adult rams (regardless of sexual orientation) to physiological quantities cannot stimulate or restore sexual behaviors to levels observed prior to castration. Furthermore, this treatment does not seem to affect sexual orientation. Additionally, these data suggest that the testes is not a source of OT and that this nonapeptide does not have a major effect in promoting sexual behavior of rams.

Implications

Approximately 8 to 10% of the ram population is male-oriented or asexual. Male-oriented sexual behavior observed within the sheep population seems to be for reasons other than societal dominance or flock hierarchy. Because there are no readily apparent phenotypic traits that characterize the male-oriented or asexual ram, these animals are frequently selected as flock sires and hence contribute to decreased fertility of ewes and economic loss to the sheep industry. Treatment of these types of rams with estrogens to enhance their sexual behavior or performance may not be practical. Differences in sexual behavior cannot be attributed to differences in testiccular production of hormones as determined by analysis of blood samples collected from conscious rams.

Literature Cited


Behavior of castrated rams


