Effects of oxytocin and an oxytocin antagonist on testosterone secretion during the oestrous cycle of the goat (*Capra hircus*)

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Summary. Oxytocin at a dose of 100 i.u. injected subcutaneously (s.c.) daily to goats between Days 3 and 6 of the oestrous cycle caused a significant increase in testosterone secretion compared with saline-treated animals. An oxytocin antagonist (0.2 µg/kg) injected intra-arterially between Days 12 and 18 of the oestrous cycle or simultaneously with oxytocin between Days 3 and 6 blocked the increased release of testosterone and occurrence of oestrus. It is suggested that oxytocin-induced oestrus may occur via testosterone secretion.

Keywords: oxytocin; testosterone; oestrous cycle; goat

Introduction

Testosterone secretion has been demonstrated in cyclic ruminants (Shemesh *et al.*, 1975; Kanchev *et al.*, 1976; Homeida & Cooke, 1984). Increased peripheral plasma concentration of testosterone occurs at the start of luteal regression (Homeida, 1986). Oxytocin concentrations in the blood are also raised during the luteal phase and a pulsatile pattern of release is seen at the start of luteolysis (Homeida & Cooke, 1982, 1983). This study was conducted to investigate whether oxytocin will have an effect on testosterone secretion during the oestrous cycle of the goat.

Materials and Methods

Fifteen mature Nubian goats (2–4 years of age and with normal oestrous cycles of 19–20 days) were used. They were housed in individual pens under conditions of natural daylength and temperature. Animals were synchronized with two 5 mg injections of a prostaglandin F-2α analogue (Lutalyse, Upjohn Ltd, Crawley, UK) given 11 days apart; when they exhibited oestrus (determined by a fertile buck) they were randomly divided into the following groups.

In Group A, 3 goats were injected daily with oxytocin given subcutaneously (s.c.) at a dose of 100 i.u. between Days 3 and 6 of the oestrous cycle (oestrus = Day 0). In Group B, 3 goats were treated like those in Group A but were given saline (0.9% w/v NaCl). In Group C, 3 goats were treated like those in Group A but in addition an oxytocin antagonist (1-(B-mercapto-B, B-diethylpropionic acid), 2-O-ethyl tyrosine, 8-ornithino) vasotocin (dEt₂ Tyr (Et)-OVT, Code KB-IV-24 (a gift from Professor M. Manning) was given intraarterially at a dose of 0.2 µg/kg body weight. Oxytocin antagonist or saline was injected as previously described (Homeida & Khalafalla, 1987). Briefly, animals were anaesthetized with sodium thiopentone intravenously and maintained with fluothane and oxygen. At surgery, a femoral artery was catheterized with a polyvinyl catheter. The catheter was fed 20 cm into the vessel so that the tip lay in the abdominal aorta 6–10 cm above the origin of the uterine artery. In Group D, 3 goats were treated with the oxytocin antagonist between Days 10 and 20 of the oestrous cycle. In Group E, 3 goats were treated as in Group D with saline.

All animals were observed for oestrus at least twice daily with the help of a buck. Jugular vein blood (5 ml) was collected 3 times daily by venepuncture using 23-gauge needles. Blood was collected with chilled heparinized tubes, centrifuged at 2000 g and plasma was stored at –20°C until analysed.

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Plasma progesterone and testosterone were measured by radioimmunoassay (RIA) methods, as previously described (Homedia, 1986; *et al.*, 1988). Progesterone antibody (provided by Dr H. Dobson) was raised in rabbits against progesterone-11-succinyl-BSA, and used at a final dilution of 1:7000; cross-reactions were 100% with progesterone and <0.1% with corticosterone, desoxycorticosterone and ketocorticosterone.

The intra- and inter-assay coefficients of variation (CV) for progesterone were 4.6% ($n = 25$) and 11.6% ($n = 20$), respectively for a plasma sample of low progesterone concentration (0.8 ng) and 4.2% ($n = 25$) and 11.6% ($n = 20$), respectively for a plasma sample of high concentration (5 ng). The sensitivity of the assay was 48 pg/tube. Extraction efficiency was 85.1 ± 5 (s.d.)% and the results were corrected for extraction losses. Testosterone cross-reactions obtained were 100% for testosterone, 50% for dehydrotestosterone, 7% for androstanediol and <0.02% for progesterone and oestradiol-17 β .

The intra-assay CV for testosterone was 11.9% ($n = 15$) for a plasma sample of low testosterone (60 pg) and 12.9% ($n = 15$) for a plasma sample of high testosterone concentration (900 pg) and the assay sensitivity was 13 pg/tube. Efficiency of radioactive hormone recovery was 75 ± 2 (s.d.)% and values were corrected for extraction losses. The results were compared by Student's *t* test.

Results

Oestrous behaviour was shown by animals in Group A on Day 6 or 7 ($P < 0.001$), in Group B and C on Day 20 or 21, in Group D on Day 25 ($P < 0.01$) and in Group E on day 20 or 21 of the oestrous cycle. The hormone concentrations in the 5 groups are shown in Tables 1 and 2.

Table 1. Mean ($\pm$ s.d., 3 goats/group) plasma concentrations of progesterone and testosterone in goats treated with oxytocin (Group A), saline (Group B) and oxytocin and oxytocin antagonist (Group C) on Days 3–7 of the oestrous cycle

Day of oestrous cycle	Group A		Group B		Group C	
	Progesterone (ng/ml)	Testosterone (pg/ml)	Progesterone (ng/ml)	Testosterone (pg/ml)	Progesterone (ng/ml)	Testosterone (pg/ml)
3	0.8 $\pm$ 0.2	60 $\pm$ 10	0.7 $\pm$ 0.2	50 $\pm$ 8	0.7 $\pm$ 0.2	60 $\pm$ 10
4	1.1 $\pm$ 0.2	50 $\pm$ 8	1.2 $\pm$ 0.5	56 $\pm$ 10	1.3 $\pm$ 0.2	65 $\pm$ 8
5	0.6 $\pm$ 0.2*	320 $\pm$ 50*	1.9 $\pm$ 0.2	66 $\pm$ 9	2.1 $\pm$ 0.2	57 $\pm$ 10
6	0.5 $\pm$ 0.2*	450 $\pm$ 60*	2.1 $\pm$ 0.3	60 $\pm$ 8	2.6 $\pm$ 0.3	50 $\pm$ 7
7	0.4 $\pm$ 0.2*	480 $\pm$ 80*	2.6 $\pm$ 0.3	50 $\pm$ 10	3.1 $\pm$ 0.3	63 $\pm$ 10

* $P < 0.001$, significantly different from control (saline treated).

Table 2. Mean ($\pm$ s.d., 3 goats/group) plasma concentrations of progesterone and testosterone in goats treated with an oxytocin antagonist (Group D) or saline (Group E) on Days 12–18 of the oestrous cycle

Days of oestrous cycle	Group D		Group E	
	Progesterone (ng/ml)	Testosterone (pg/ml)	Progesterone (ng/ml)	Testosterone (pg/ml)
12	4.7 $\pm$ 0.4	68 $\pm$ 10	4.7 $\pm$ 0.4	68 $\pm$ 16
13	5.1 $\pm$ 0.4	80 $\pm$ 11	5.3 $\pm$ 0.5	90 $\pm$ 14
14	5.2 $\pm$ 0.3	60 $\pm$ 10	5.0 $\pm$ 0.4	660 $\pm$ 110*
15	5.3 $\pm$ 0.4	66 $\pm$ 10	3.2 $\pm$ 0.3	380 $\pm$ 60*
16	4.8 $\pm$ 0.4	50 $\pm$ 11	2.3 $\pm$ 0.2	110 $\pm$ 25
17	4.6 $\pm$ 0.4	55 $\pm$ 9	1.4 $\pm$ 0.2	80 $\pm$ 12
18	4.9 $\pm$ 0.4	50 $\pm$ 10	0.4 $\pm$ 0.2	51 $\pm$ 11

* $P < 0.001$, significantly different from control.

Oxytocin injected between Days 3 and 6 (Group A) caused luteolysis and oestrus as indicated by low progesterone concentration ($P < 0.001$) compared with that in Group B goats. Oxytocin also increased testosterone values on Days 5, 6 and 7 of the oestrous cycle, but not of that of

the control (Group B). Treatment with an oxytocin antagonist (Group C) blocked the effects of oxytocin, and progesterone and testosterone concentrations were similar to those of Group B goats.

Testosterone secretion also increased on Days 13–14 of the oestrous cycle ($P < 0.001$) (Group E) (progesterone concentration started to decrease, reaching 1 ng/ml on Day 18). This effect was blocked by the oxytocin antagonist; in Group D there was no increase in testosterone values and progesterone concentrations were maintained at >4 ng/ml until Day 19 of the oestrous cycle.

Discussion

The present results substantiate earlier reports (Homeida, 1986) and show that a peak of testosterone concentration (Group E) occurred during luteal regression. The origin of this testosterone is believed to be the corpus luteum (Shemesh & Hansel, 1974; Kanchev *et al.*, 1976; Kanchev & Dobson, 1976; Homeida & Cooke, 1984; Homeida, 1986).

Oxytocin injection also caused luteolysis and was associated with increased testosterone concentration (Group A). Such an effect may be a natural one in the steps preceding luteolysis similar to the effects observed during Days 13 and 14 of the oestrous cycle when a peak of testosterone was observed at the start of luteal regression (Homeida, 1986). However, the injection of an oxytocin antagonist maintained progesterone concentrations and blocked the rise in testosterone values, giving further support to the fact that oxytocin given exogenously, as in this study or released endogenously (Homeida & Cooke, 1982), to goats will stimulate the secretion of testosterone.

The oxytocin-induced increase in testosterone level may be a direct one or due to increased release of PGF-2 α (Sharma & Fitzpatrick, 1974; Cooke & Homeida, 1982; Homeida & Cooke, 1982) or to release of LH/FSH (Franchimont & Legross, 1986). Additions of both PGF-2 α and LH to incubations of corpora lutea caused increases of testosterone synthesis (Shemesh *et al.*, 1975). Testosterone and PGF-2 α are released on Days 13 and 14 of the oestrous cycle (Homeida & Cooke, 1982, 1984).

We suggest that oxytocin-induced release of testosterone is most likely to occur via oxytocin release of PGF-2 α . Treatment with an oxytocin antagonist also inhibited the release of PGF-2 α in goats during luteal regression (Homeida & Khalafalla, 1987). However, testosterone, by acting as a precursor in the synthesis of oestrogen (Ainsworth & Kennet, 1966; Massa & Martini, 1974), may assist the secretion of PGF-2 α (Sharma & Fitzpatrick, 1974).

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