Bromocriptine-induced premature oestrus is associated with changes in the pulsatile secretion pattern of follicle-stimulating hormone in beagle bitches

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The secretory profiles of LH and FSH were investigated before and during the administration of bromocriptine in six beagle bitches. Plasma samples were obtained via jugular venepuncture at 10 min intervals for 6 h every 2 weeks until the next ovulation. Bromocriptine treatment was started 100 days after ovulation. Both before and after bromocriptine treatment, LH and FSH pulses occurred together. The mean duration of the FSH pulse (120 min) was significantly longer than that of the LH pulse (80 min). The interoestrous interval in the bitches treated with bromocriptine was significantly shorter than that of the preceding cycle (160 ± 3 versus 206 ± 24 days). The mean basal plasma FSH concentration (7.4 ± 0.6 versus 6.1 ± 0.7 iu l⁻¹) and the mean area under the curve for FSH (46.6 ± 4.7 versus 40.4 ± 4.4 iu l⁻¹ in 6 h) increased significantly after the start of the bromocriptine treatment. In contrast, the differences in mean basal plasma LH concentration (2.1 ± 0.2 versus 2.0 ± 0.2 µg l⁻¹) and the mean area under the curve for LH (19.0 ± 3.1 versus 19.5 ± 2.5 µg l⁻¹ in 6 h) between the day before and 14 days after the start of the bromocriptine treatment were not significant. Bromocriptine administration also lowered the mean amplitude of the FSH pulse and shortened the mean duration of the FSH pulse, without influencing the LH pulse. In addition to demonstrating the concurrent pulsatile secretion of LH and FSH, the results of the present study demonstrate that the bromocriptine-induced shortening of the interoestrous interval in the bitch is associated with an increase in plasma FSH concentration without a concomitant increase in plasma LH concentration. This finding indicates that treatment with the dopamine agonist bromocriptine increases plasma FSH to a concentration that results in the enhancement of follicle development.

Introduction

The oestrous cycle of the Canidae differs considerably from that of other species. Most studies have been performed in domestic bitches, in which the follicular phase and spontaneous ovulations are followed by a luteal phase with an average duration of about 75 days (Schaefer-Okkens, 1996). A non-seasonal anoestrus of variable duration (2–10 months) follows each oestrous cycle (Bouchard *et al.*, 1991; Concannon, 1993). Endocrine changes leading to termination of anoestrus and the start of a new follicular phase in bitches are poorly understood.

Progression from early to late anoestrus is associated with an increase in the basal plasma FSH concentration, indicating

that, in bitches, an increase in circulating FSH concentrations is a critical event in the initiation of ovarian folliculogenesis (Kooistra *et al.*, 1999). An increasing release of GnRH by the hypothalamus (Tani *et al.*, 1996) and an increase in the sensitivity of the pituitary to GnRH from early to late anoestrus (Van Haaften *et al.*, 1994) have also been observed in this species. In addition, an increase in ovarian responsiveness to gonadotrophins (Jeffcoate, 1993), increased circulating basal LH concentrations at the end of anoestrus (Olson *et al.*, 1982) and a brief period of increased LH pulsatility (Concannon *et al.*, 1986) are important determinants of the initiation of a new follicular phase. It has even been suggested that changes in LH secretion may be more important than changes in FSH secretion in the initiation of a follicular phase leading to ovulation (Concannon, 1993). In line with this suggestion, the

administration of pharmacological doses of LH alone can terminate anoestrus by inducing a follicular phase in bitches (Verstegen *et al.*, 1997).

In addition to these changes in the hypothalamus–pituitary–gonad axis, there is evidence of involvement of dopaminergic influences and prolactin release. Administration of the dopamine 2 receptor agonists, bromocriptine and cabergoline, during both the luteal phase and anoestrus results in shortening of the interoestrous interval (Okkens *et al.*, 1985a; Van Haften *et al.*, 1989; Concannon, 1993; Onclin *et al.*, 1995). The shortened interoestrous interval is the result of a shortening of the duration of both the luteal phase (Okkens *et al.*, 1985a; Okkens *et al.*, 1990) and anoestrus (Okkens *et al.*, 1985a; Concannon, 1993). It has been hypothesized that the dopamine 2 agonist-induced shortening of the interoestrous interval is the result of the suppression of prolactin secretion. Indeed, in humans, it has been demonstrated that high concentrations of prolactin inhibit GnRH pulsatility (Saunders *et al.*, 1984; Yazigi *et al.*, 1997). Studies in several other mammalian species have also demonstrated that hyperprolactinaemia is associated with decreased gonadotrophin secretion (Bever *et al.*, 1983; Park and Selmoff, 1993; Park *et al.*, 1993; Jedlinska *et al.*, 1995). Lowering of the plasma prolactin to basal concentrations is usually associated with the return of gonadotrophic pulsatility (Bever *et al.*, 1983; Saunders *et al.*, 1984).

However, under physiological conditions, no obvious changes in plasma concentrations of prolactin have been observed in bitches during the transition from anoestrus to the follicular phase (Olson *et al.*, 1982). In addition, administration of metergoline, a 5-hydroxytryptamine (serotonin) receptor antagonist, to anoestrous bitches in a dose resulting in plasma prolactin concentrations even lower than those during bromocriptine treatment did not shorten the interoestrous interval (Okkens *et al.*, 1997). These observations indicate that the induction of the follicular phase is not initiated by the suppression of prolactin secretion, but by other direct or indirect dopaminergic effects. The present study investigated the pulsatile patterns of secretion of LH and FSH during bromocriptine-induced transition from anoestrus to the follicular phase in bitches to improve understanding of the interaction between dopaminergic influences and gonadotrophin release.

Materials and Methods

Animals, treatment, and collection of blood samples

Six healthy beagle bitches, 1.5–5.0 years of age, were used in this study. All bitches had been born and raised in the Department of Clinical Sciences of Companion Animals and were accustomed to the laboratory environment and procedures such as collection of blood. They were housed singly or in pairs in indoor–outdoor runs, fed a standard commercial dog food once a day, and given water *ad libitum*.

All bitches were examined three times a week for the presence of swelling of the vulva and a serosanguinous vaginal discharge, which were considered to signify the

onset of pro-oestrus. Plasma concentrations of progesterone were determined three times per week from the start of pro-oestrus until the day at which the plasma progesterone concentration exceeded 16 nmol l^{-1} , which is when ovulation was assumed to occur (Concannon *et al.*, 1977; Wildt *et al.*, 1979; Okkens *et al.*, 1985a).

The bitches were treated with $20 \mu\text{g}$ bromocriptine (Lactafal[®], a gift from Eurovet, Bladel) per kg body weight orally, twice a day, starting 100 ± 2 (mean $\pm$ SD) days after ovulation. Two bitches were treated after the first ovulation, consequently data on the duration of the preceding interoestrous interval were not available. The 6 h secretory profiles of LH and FSH from each bitch were determined the day before (day 0) and every 2 weeks after the start of the treatment with bromocriptine until the next ovulation or for a maximum of six times. Blood samples were collected from the jugular vein between 08:00 and 14:00 h at 10 min intervals, immediately placed in chilled heparin-coated tubes, and centrifuged for 10 min at 2000 g. Plasma was stored at -25°C until assayed.

Hormone determinations

Plasma LH concentrations were determined by a heterologous radioimmunoassay as described by Nett *et al.* (1975). A rabbit antiserum raised against ovine LH (CSU-204; kindly supplied by G. D. Niswender, Colorado State University), radio-iodinated NIAMDD-bLH-4, and canine pituitary standard LER 1685-1 (a gift from L. E. Reichert, Albany Medical College, NY) were used in this assay. The intra-assay and interassay coefficients of variation for values above $0.5 \mu\text{g l}^{-1}$ were 2.3 and 10.5%, respectively. The lowest detectable concentration of LH was $0.3 \mu\text{g l}^{-1}$.

Plasma FSH concentrations were determined by applying a human immunometric sandwich assay (Amerlite, Amersham, Bucks), validated for bitches (Kooistra *et al.*, 1999). Results are expressed in units of a human FSH standard (2nd International Reference Preparation (IRP) 78/549). Values of FSH measured with the Amerlite assay in serum, EDTA plasma, or heparin plasma from bitches were similar. Values of FSH measured with the Amerlite assay after serial dilution of various bitch samples showed good linearity down to a concentration of 0.5 iu l^{-1} , indicating that, in this assay, the immunochemistry of bitch FSH is similar to that of the human FSH standards. The sensitivity of the assay was 0.5 iu l^{-1} . The intra-assay coefficient of variation was $< 5\%$. The interassay coefficients of variation were 5% at a concentration of 35 iu l^{-1} and 9% at a concentration of 3 iu l^{-1} . Recovery of human FSH (assay standard preparation) added to bitch samples was 103%. Possible crossreactivity of bitch LH in the FSH assay was tested by analysing the canine pituitary standard LER 1685-1, purified by L. E. Reichert, in the FSH assay. The purified canine pituitary LH standard showed an apparent crossreactivity in the FSH assay. However, this apparent crossreactivity was probably due to a contamination of the LH preparation with bitch FSH, as judged from the perfect parallelism of the LH preparation in the FSH assay. Even if the crossreactivity is considered genuine, the contribution of an LH peak value to a concomitant FSH peak value was $< 10\%$ of

the FSH value. Plasma from hypophysectomized bitches showed FSH concentrations $< 0.5 \text{ IU l}^{-1}$.

Plasma concentrations of progesterone were determined by a previously validated radioimmunoassay (Dieleman and Schoenmakers, 1979; Okkens *et al.*, 1985b). The intra-assay and interassay coefficients of variation were 11% and 14%, respectively. The lowest detectable concentration was 0.13 nmol l^{-1} .

Data analysis

Plasma concentrations of LH were determined in all samples and plasma concentrations of FSH were determined in plasma samples at 10 min intervals in two bitches and at 20 min intervals in the other four bitches.

The pattern of pulses in the 6 h profiles of FSH and LH was analysed by means of the Pulsar programme developed by Merriam and Wachter (1982). The programme identifies secretory peak values by height and duration from a smoothed baseline, using the assay SD as a scale factor. The cut-off parameters G1–G5 of the Pulsar programme were set at 3.98, 2.40, 1.68, 1.24 and 0.93 times the assay SD as criteria for accepting peaks 1, 2, 3, 4 and 5 points wide, respectively. The smoothing time, a window used to calculate a running mean value omitting peak values, was set at 4 h. The splitting cut-off parameter was set at 2.7 and the weight assigned to peak values was 0.05. The A, B, and C values of the Pulsar programme, used to calculate the variance of the assay, were set at A = 0, B = 10.5 and C = 0 for the LH assay and A = 0, B = 5 and C = 0 for the FSH assay. The values extracted from the Pulsar analyses included the mean of the smoothed baseline, the mean peak amplitude, the pulse frequency, the mean pulse duration, and the area under the curve (AUC). The AUC was calculated above the zero level.

Differences in the duration of LH and FSH pulses and changes in the parameters of the secretory patterns immediately before and 14 days after the start of the bromocriptine treatment were evaluated by unpaired or, if appropriate, paired two-tailed Student's *t* tests. Next, changes in the parameters of the secretory patterns during the first 6 weeks of bromocriptine treatment were evaluated by one-way analysis of variance (ANOVA) or, if appropriate, ANOVA for repeated measures for main effect of duration of bromocriptine treatment. Subsequently, multiple comparisons were performed for data with significant ($P \leq 0.05$) main effect using the Student–Newman–Keuls test or, when day 0 was used as the control group, using the Dunnett test. Differences in the duration of the oestrous intervals were compared using Wilcoxon's signed rank test and differences in pulse frequency were analysed using the Friedman test, with multiple comparisons performed using the Dunnett test. Values are expressed as mean $\pm$ SEM or as mean and range. $P \leq 0.05$ was considered significant.

Ethics of experimentation

This study was approved by the Ethical Committee of the Faculty of Veterinary Medicine, Utrecht University.

Results

The oestrous interval in the bitches treated with bromocriptine was significantly shorter than in their preceding cycle (160 ± 3 versus 206 ± 24 days; $n = 4$). The oestrous interval was also significantly shorter than that in another group of six beagle bitches in the dog colony, in which the oestrous interval was assessed during approximately the same period (225 ± 16 days). In two bitches, ovulation occurred after the fourth sample collection (day 42) for the 6 h secretory profiles of LH and FSH whereas, in two other bitches, it occurred after the fifth sampling period (day 56). In the remaining two bitches, six secretory profiles were determined.

Both LH and FSH were secreted in a pulsatile fashion, although LH and FSH pulses could not be detected in all 6 h profiles (Fig. 1). Thirty-one LH pulses were identified by the Pulsar programme in the 6 h profiles of the six bitches. Each LH pulse coincided with an increase in plasma FSH concentration, but only 28 of the latter were recognized as significant FSH pulses by the Pulsar programme. There were no FSH pulses without a concurrent significant LH pulse. The basal plasma LH concentration was low compared with the maximum peak concentrations, whereas FSH pulses were characterized by relatively low peak values compared with the basal plasma FSH concentration. Both LH and FSH pulses were characterized by a rapid increase and a slow decrease in plasma LH concentration and, even more pronounced, in plasma FSH concentrations (Fig. 1). The mean duration of the FSH pulse (120 ± 5 min) was significantly longer than that of the LH pulse (80 ± 2 min).

In all bitches, both basal plasma FSH concentration and the AUC for FSH increased after the start of the bromocriptine treatment. The mean basal plasma FSH concentrations at 14, 28 and 42 days after the start of the bromocriptine treatment were significantly higher than that at the day before the onset of treatment (Fig. 2). The mean AUC for FSH at 14 days after the start of the bromocriptine treatment was significantly higher than that before treatment. Differences in mean basal plasma FSH concentration and differences in the mean AUC for FSH at 14, 28 and 42 days after the start of the bromocriptine treatment were not significant (Table 1). Differences in mean basal plasma LH concentration and differences in the mean AUC for LH between the day before and 14 days after the start of the bromocriptine treatment were not significant. However, 28 days after the start of the bromocriptine treatment, the mean AUC for LH was significantly higher than that in the other periods, whereas the mean basal plasma LH concentration on day 28 did not differ significantly from the values in the other periods (Table 1).

The frequency of LH and FSH pulses did not differ significantly before and during bromocriptine treatment. However, the mean duration of the FSH pulse before treatment was significantly longer than that at 14, 28 and 42 days after the start of the bromocriptine treatment. In agreement with this observation, the mean amplitude of the FSH pulse before treatment was significantly higher than that at 14 and 28 days after the start of bromocriptine treatment. Differences in the mean duration and the mean amplitude of the LH pulse between the day before and 14, 28

Fig. 1. The secretory profiles of FSH (▼) and LH (●) in a 4-year-old beagle bitch. Blood samples were collected at 10 min intervals for 6 h on the day before (day 0) and every 2 weeks after the start of the treatment with bromocriptine (days 14–70). Ovulation occurred on day 74.

and 42 days after the start of the bromocriptine treatment were not significant (Table 1).

In two bitches, one of the 6 h secretory profiles of LH was characterized by frequent increases of short duration (for example, see day 56 of Fig. 1). However, these increases were often too small to be recognized as significant pulses by the Pulsar programme. In both bitches, this typical 6 h secretory profile of LH was observed a few days before the onset of pro-oestrus and 14 days before the assumed day of ovulation. In two bitches, sampled 56 days after the start of the bromocriptine treatment, the 6 h secretory profiles of LH and FSH were determined during pro-oestrus. In both bitches, these 6 h secretory profiles were characterized by the absence of significant LH and FSH pulses and relatively low basal plasma FSH concentration.

Discussion

As the bromocriptine-induced shortening of the interoestrous interval in bitches can probably not be explained by suppression of prolactin secretion (Okkens *et al.*, 1997), the present study investigated whether the induction of the follicular phase in this species might be initiated by other direct or indirect dopaminergic effects. The effects of administration of bromocriptine on the pulsatile patterns of secretion of LH and FSH were studied, taking into account the essential role of the gonadotrophins in the induction of a follicular phase. The results demonstrate that the bromocriptine-induced shortening of the interoestrous interval in bitches is associated with an increase in both the basal plasma FSH concentration and the AUC for FSH,

Fig. 2. The mean ($\pm$ SEM) plasma (a) LH and (b) FSH concentrations of the smoothed baseline in six beagle bitches the day before and 14, 28 and 42 days after the start of bromocriptine treatment. Asterisks indicate significant difference ($P < 0.05$).

Table 1. Pulse characteristics of the 6 h secretory profiles of FSH and LH in six healthy beagle bitches treated with bromocriptine, starting 100 days after ovulation

	Day 0 (n = 6)	Day 14 (n = 6)	Day 28 (n = 6)	Day 42 (n = 6)	Day 56 (n = 4)	Day 70 (n = 2)
LH peak duration (min)	86 (70–100) (n = 5)	83 (70–90) (n = 3)	78 (70–80) (n = 4)	77 (70–80) (n = 3)	80 (n = 1)	80 (n = 1)
FSH peak duration (min)	150 (120–160) (n = 4)	107 (100–120) (n = 3)	120 (100–140) (n = 4)	117 (100–130) (n = 3)	120 (n = 1)	110 (n = 1)
LH peak amplitude ($\mu\text{g l}^{-1}$)	21.5 $\pm$ 2.3 (n = 5)	17.1 $\pm$ 3.0 (n = 4)	21.7 $\pm$ 4.2 (n = 5)	16.2 $\pm$ 5.5 (n = 4)	22.4 (n = 1)	139 (n = 1)
FSH peak amplitude (iu l ⁻¹)	6.6 $\pm$ 0.3 (n = 5)	4.3 $\pm$ 0.4 (n = 4)	4.3 $\pm$ 0.5 (n = 5)	4.9 $\pm$ 1.8 (n = 3)	5.3 (n = 1)	2.7 (n = 1)
LH pulse frequency (peaks per 6 h)	1.2 (0–2)	1.0 (0–2)	1.5 (1–2)	0.8 (0–2)	0.3 (0–1)	0.5 (0–1)
FSH pulse frequency (peaks per 6 h)	1.2 (0–2)	1.0 (0–2)	1.5 (1–2)	0.5 (0–1)	0.3 (0–1)	0.5 (0–1)
LH basal ($\mu\text{g l}^{-1}$)	2.0 $\pm$ 0.2	2.1 $\pm$ 0.2	2.2 $\pm$ 0.3	2.3 $\pm$ 0.3	2.0 $\pm$ 0.2	2.2 $\pm$ 0.3
FSH basal (iu l ⁻¹)	6.1 $\pm$ 0.7	7.4 $\pm$ 0.6	7.2 $\pm$ 0.5	7.5 $\pm$ 0.4	3.8 $\pm$ 1.4	5.3 $\pm$ 1.1
AUC for LH ($\mu\text{g l}^{-1}$ in 6 h)	19.5 $\pm$ 2.5	19.0 $\pm$ 3.1	25.0 $\pm$ 2.1	18.4 $\pm$ 1.5	16.3 $\pm$ 1.9	14.9 $\pm$ 0.1
AUC for FSH (iu l ⁻¹ in 6 h)	40.4 $\pm$ 4.4	46.6 $\pm$ 4.7	45.0 $\pm$ 3.8	44.6 $\pm$ 1.7	23.0 $\pm$ 9.0	31.4 $\pm$ 6.5

The secretory profiles were determined the day before (day 0) and every 2 weeks after the start of the treatment until the next ovulation. The values are expressed as mean $\pm$ SEM or mean (and range).

AUC: area under curve; n: the number of bitches in which a parameter could be determined.

without a concomitant increase in the basal plasma LH concentration and the AUC for LH. This finding gives further support to the notion that an increase in circulating FSH should be considered to be a critical event required for ovarian folliculogenesis in bitches (Kooistra *et al.*, 1999). In this respect, there are similarities with the situation in primates. Observations during gonadotrophin-induced ovulation in women have emphasized that plasma FSH must exceed a certain concentration before preantral follicles reaching the FSH-dependent stage can progress to maturation (Brown, 1978; Schoemaker *et al.*, 1993). This has been named 'the FSH threshold concept'. Increases in plasma FSH concentrations only 10–30% above the threshold concentration are sufficient to stimulate normal follicular development in women (Brown, 1978). The threshold for FSH may vary among individuals, and each follicle also has its own sensitivity to FSH (Schoemaker *et al.*, 1993). The increase in basal plasma FSH concentration during the progression of anoestrus (Kooistra *et al.*, 1999) and the fact that the bromocriptine-induced shortening of the

interoestrous interval is associated with an increase in plasma FSH concentration indicate that, analogous to the situation in women, an FSH threshold concept can also be postulated in bitches. If this is the case, dopamine 2 receptor agonists directly or indirectly increase plasma FSH concentration above the concentration that results in an enhancement of follicle development.

In the course of the bromocriptine treatment, that is, during progression of follicular development, plasma FSH concentrations decreased again. This was most obvious in two bitches in which the secretory profiles of LH and FSH were determined during pro-oestrus, that is, during the follicular phase. In agreement with earlier observations (Olson *et al.*, 1982; Kooistra *et al.*, 1999), the basal plasma FSH concentrations in these bitches were relatively low during this phase. As has been observed in other species, this finding is probably attributable to negative feedback effects of oestradiol and the ovarian peptides, inhibin and follistatin (Shupnik, 1996). The decreasing FSH release will prevent further development of other follicles in which the threshold

concentration has not yet been surpassed and, consequently, will prevent the development of an excessive number of preovulatory follicles. The largest and foremost follicles may escape the atretic effect of the decreasing plasma FSH concentration because of an increased sensitivity to FSH (Zelezniak and Kubik, 1986).

With regard to the pulsatile secretion pattern of LH, the most remarkable finding was that, in two bitches, one of the 6 h secretory profiles of LH was characterized by frequent increases of short duration. This typical secretory profile of LH has been reported before in bitches and has been associated with the termination of anoestrus (Concannon *et al.*, 1986; Concannon, 1993). Indeed, in both bitches, this LH secretory profile was observed a few days before the onset of pro-oestrus. According to Concannon (1993), the period during which increased LH secretion may be observed is brief, perhaps only 4–8 days, and may not be continuous. The precise role of this increased LH secretion in termination of anoestrus in the bitch remains elusive. There are indications that LH plays an important role in the gonadotrophin-dependent stage of canine follicle development, because administration of pharmacological doses of LH alone can terminate anoestrus by inducing a follicular phase in bitches (Versteegen *et al.*, 1997). Sensitivity to FSH is modulated by regulatory substances of thecal cell origin. Since LH regulates thecal cell function, stimulation by LH may sensitize granulosa cells to FSH indirectly, that is, modulate FSH thresholds (Hillier, 1996).

In agreement with recent observations (Kooistra *et al.*, 1999), FSH and LH pulses were released in concordance, indicating that GnRH is probably the sole hypothalamic releasing hormone for both LH and FSH in this species. Concurrent secretion of LH and FSH has also been reported in humans (Veldhuis *et al.*, 1992; Genazzani *et al.*, 1993; Booth *et al.*, 1996; Lambalk *et al.*, 1997), rats (Urbanski *et al.*, 1988), mares (Irvine and Alexander, 1993; Silvia *et al.*, 1995), deer (Monfort *et al.*, 1993) and cattle (Schallenberger *et al.*, 1985). However, in most of these species, independent FSH and LH pulses are also observed frequently. In these species, FSH and LH synthesis and secretion are regulated differentially by the amplitude and frequency of GnRH pulses. A low frequency of GnRH stimulation increases FSH release preferentially, whereas LH is stimulated optimally by more frequent GnRH pulses (Haisenleder *et al.*, 1991; Shupnik, 1996; Vizcarra *et al.*, 1997). This finding may be explained by the fact that the expression of intrapituitary factors involved in the synthesis of FSH, that is, activin and follistatin, is also dependent on the GnRH pulse frequency (Besecke *et al.*, 1996). Gonadal steroids and gonadal peptides also play a role in the differential or non-parallel secretion of FSH and LH (Shupnik, 1996). In addition, the existence of a specific hypothalamic FSH-releasing factor has been demonstrated in the rat (Yu *et al.*, 1997; Koppa *et al.*, 1998).

Despite the coincidence of pulses of LH and FSH, there are differences in basal concentrations and pulse characteristics, indicating that, in bitches, there is differential regulation of the secretion of these hormones. First, progression of anoestrus in bitches is associated with an increase in the basal concentration of plasma FSH concentration, whereas plasma LH remains at the same basal concentrations

(Kooistra *et al.*, 1999). Second, in contrast to the basal plasma LH concentrations, the basal plasma FSH concentrations are relatively low during the follicular phase of the oestrous cycle (Kooistra *et al.*, 1999). In the present study, administration of bromocriptine was also associated with differential regulation of the secretion patterns of FSH and LH. Fourteen days of bromocriptine administration resulted in a significant increase in basal plasma FSH concentration and AUC for FSH, without a concomitant increase in basal plasma LH concentration and AUC for LH. In addition, bromocriptine treatment resulted in FSH pulses with a lower mean amplitude and a shorter mean duration, without influencing the pulse characteristics of LH. Thus, with regard to the interaction between dopaminergic influences and gonadotrophin release, it may be concluded that the effect of bromocriptine is confined to FSH secretion.

In addition to demonstrating the concurrent pulsatile secretion of LH and FSH, the results of the present study demonstrate that the bromocriptine-induced shortening of the interoestrous interval in bitches is associated with an increase in plasma FSH concentration without a concomitant increase in plasma LH concentration. This finding indicates that treatment with the dopamine agonist, bromocriptine, increases plasma FSH concentration to a concentration that results in the enhancement of follicle development. The FSH threshold concept, originally proposed in primates, seems to hold true also for bitches.

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