

The central oxytocin pulse generator: a pacemaker for the ovarian cycle

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Abstract. During luteolysis in sheep, episodic pulses of oxytocin (OT), contributed by the neurohypophysis and the corpus luteum (CL), stimulate uterine luteolytic pulses of prostaglandin (PG) F₂ α *via* endometrial OT receptors. To distinguish relative contributions of neurohypophysial and luteal OT, ovariectomized sheep were given estradiol-17 β (E) and progesterone (P) to simulate levels during the cycle. In intact sheep, luteectomy was performed to exclude the CL as a source of OT and to initiate P withdrawal. In ovariectomized sheep, E (1 μ g/h for 12 to 36 h) superimposed on basal E (0.05 μ g/h), caused a series of 4 to 6 episodes of high frequency pulses of OT, each episode lasting 1 to 2 h at intervals of 3 h, and commencing at 24 h. Withdrawal of P (500 μ g/h), superimposed on basal E in ovariectomized sheep, or luteectomy in intact sheep, evoked similar episodes of high frequency pulses of OT beginning at 24 h. We conclude that (1) an increase in E levels, or the return of E action following P withdrawal, causes intermittent increases in the frequency of the central OT pulse generator. (2) high frequency pulses of OT initiate subluteolytic levels of uterine PGF₂ α which trigger a supplemental release of luteal OT; (3) luteal OT amplifies the secretion of uterine PGF₂ α which initiates luteolysis and causes more luteal OT to be secreted; and (4) in addition to the established hypothalamic-anterior pituitary-gonadal axis for initiating the ovarian cycle (*via* the gonadotrophins), there is now evidence for a hypothalamic-posterior pituitary-gonadal axis for terminating the ovarian cycle (*via* OT).

Key words: oxytocin, neurohypophysis, corpus luteum, pulse generator, hormone pulsatility, pacemaker, ovarian cycle, luteolysis

Mini-review

INTRODUCTION

Identification of prostaglandin F_{2α} as the luteolytic hormone

The corpus luteum (CL) is formed from the cells of the follicle wall after ovulation and secretes the hormone progesterone which is responsible for the maintenance of pregnancy in mammals. In addition to gestational effects on the uterus, an important action of progesterone is to suppress gonadotrophin secretion from the anterior pituitary and thus prevent further ovulatory activity. In species which exhibit regular ovarian cyclicity, when conception does not occur, the CL has to be "removed" from the ovary (luteolysis or CL regression) so that a new ovarian cycle can be initiated. The importance of the uterus in the control of CL life span was recognized many years ago by Loeb (1923) who showed that the corpora lutea of guinea pigs failed to regress after hysterectomy. Subsequently, a similar effect of hysterectomy on luteal life span was demonstrated in several other non-primate species including the sheep (Anderson et al. 1969). However, it was not until 1972 that the hormone-like substance, prostaglandin F_{2α} (PGF_{2α}) was identified by GC/MS as the uterine luteolytic hormone in sheep (McCracken et al. 1972). Evidence has since accumulated that PGF_{2α} performs a similar role in several other non-primate species while in primates, in which the life span of the CL is unaffected by hysterectomy, the source of PGF_{2α} for luteolysis may include the ovary itself (see McCracken and Schramm 1988). During luteolysis in sheep, PGF_{2α} is secreted by the uterus into the utero-ovarian vein and, to some extent, into the lymphatics and reaches the ovary directly *via* a counter current transfer mechanism in the ovarian vascular pedicle. Such a mechanism permits a small amount of uterine PGF_{2α} (about 1%) to diffuse into the ovarian artery and to reach the ovary directly without passing through the systemic circulation where it is rapidly metabolized to its inactive metabolite (PGFM), particularly in the pulmonary vascular bed (McCracken et al. 1972).

Hormonal regulation of uterine prostaglandin F_{2α} synthesis

Early studies in sheep indicated that PGF_{2α} synthesis in the endometrium was influenced by the ovarian steroids estradiol-17β (E) and progesterone (P) (Caldwell et al. 1972, Wilson et al. 1972, Barcikowski et al. 1974, Louis et al. 1977). It was found that E stimulated endometrial PGF_{2α} synthesis, but that E-induced PGF_{2α} production was markedly enhanced by a prior exposure to P. Similar effects of E and P on uterine PGF_{2α} synthesis were obtained in other species such as the guinea pig (Blatchley et al. 1972), and the rat (Castracane and Jordan 1975). Subsequently, it became apparent in the ovine species that E and P indirectly controlled uterine PGF_{2α} synthesis *via* the regulation of receptors for oxytocin (OT) in the endometrium. This was based on the finding that mechanical stimulation of the ovine uterus evoked the secretion of PGF_{2α} only early and late in the cycle, while no effect was seen during the mid-luteal phase of the cycle (Wilson et al. 1974). Since mechanical stimulation of the female reproductive tract causes an elevation of OT in the peripheral blood of sheep and goats (Roberts and Share 1969, Blank and DeBias 1977) *via* the centrally acting Ferguson reflex (Ferguson 1941), we considered that OT released by mechanical stimulation of the uterus might be responsible for the observed stimulatory effect on PGF_{2α} secretion. This seemed plausible since exogenous OT had previously been shown to shorten the estrous cycle of the cow when administered early in the cycle (Armstrong and Hansel 1959), an effect which had been postulated to be due to OT-stimulated uterine PGF_{2α} synthesis (McCracken 1972). We subsequently demonstrated that OT infused into the arterial supply of the ovine uterus mimicked the cyclical variation in the effects of mechanical stimulation on PGF_{2α} secretion from the uterus (Wilson et al. 1974, Roberts et al. 1975, Roberts and McCracken 1976). Therefore, it seemed likely that the cyclical variation in the ability of OT to stimulate the synthesis of endometrial PGF_{2α} was due to a cyclical variation in the

concentration of receptors for OT in the endometrium. This proposal was supported by reports that target sites for OT, such as the mammary gland and the oviduct, had been shown to bind OT with high affinity (Soloff et al. 1975) and that E enhanced the binding of OT by the uterus and oviduct in the rat. Moreover OT-induced secretion of $\text{PGF}_2\alpha$ by the uterus of anestrus sheep was enhanced by pre-treatment with E (Sharma and Fitzpatrick 1974). Subsequently, we showed that OT-stimulation of $\text{PGF}_2\alpha$ from ovine endometrium *in vitro* was positively correlated with the relative abundance of OT receptors in this tissue (Roberts et al. 1976). A model for the hormonal regulation of endometrial $\text{PGF}_2\alpha$ synthesis in the sheep is depicted in Fig. 1. It is proposed that E enhances the formation of OT receptors in the endometrium and that during the luteal phase P, by blocking the action of E, reduces the concentration of OT receptors. However, P eventually catalyzes the destruction of its own receptor so that towards the end of the luteal phase E action is no longer suppressed and thus induces the formation of OT receptors. The greatly enhanced synthesis of endometrial $\text{PGF}_2\alpha$ by OT at the end of the

luteal phase most likely results from the priming effect of P on lipid precursors in the endometrium during the luteal phase (McCracken 1980).

Pulsatile secretion of uterine prostaglandin $\text{F}_2\alpha$

Following the identification of $\text{PGF}_2\alpha$ as a luteolytic hormone in sheep, more frequent sampling of uterine venous blood revealed that $\text{PGF}_2\alpha$ was secreted by the uterus in a pulsatile pattern during luteolysis (McCracken et al. 1973, Thorburn et al. 1973, Barcikowski et al. 1974, Baird et al. 1976). As shown in Fig. 2, each pulse of $\text{PGF}_2\alpha$ lasts about one hour and occurs at intervals of about six to nine hours. The discovery of the pulsatile pattern of $\text{PGF}_2\alpha$ secretion was subsequently supported by the detection of a series of intermittent peaks of the primary metabolite of $\text{PGF}_2\alpha$, 15-keto-13;14-dihydro- $\text{PGF}_2\alpha$ (PGFM), in the peripheral blood of sheep during luteolysis (Kindahl et al. 1976, Peterson et al. 1976). Pulsatile secretion of uterine $\text{PGF}_2\alpha$ during luteolysis is also observed in the cow, sow, and mare (see McCracken and Schramm 1988). The secretion of $\text{PGF}_2\alpha$ in a pulsatile pattern appears to play an important role in the induction of luteolysis. In sheep it was shown that four separate hour-long infusions of $\text{PGF}_2\alpha$, given at intervals of 6 hours (4 pulses in 19 h) into the arterial supply of the ovary on cycle day 12, caused permanent regression of the CL in only one of four subjects. The addition of a fifth hour-long infusion (5 pulses in 25 h) caused permanent CL regression in four out of four sheep. A single hour-long infusion of $\text{PGF}_2\alpha$ (0.1 $\mu\text{g}/\text{h}$) given daily for four consecutive days caused a temporary fall in P after each infusion followed by recovery. Permanent CL regression did not occur with this protracted regimen, suggesting that pulses of $\text{PGF}_2\alpha$ occurring at a relatively short pulse interval over a period of about 24 h is a necessary condition for physiological regression of the CL in the sheep. Moreover, the minimal effective dose of $\text{PGF}_2\alpha$, when given as five separate hour-long pulses over 24 h, was 1/40th of the amount of $\text{PGF}_2\alpha$ (2.5 $\mu\text{g}/\text{h}$ for 6 h) which was required to cause luteolysis when

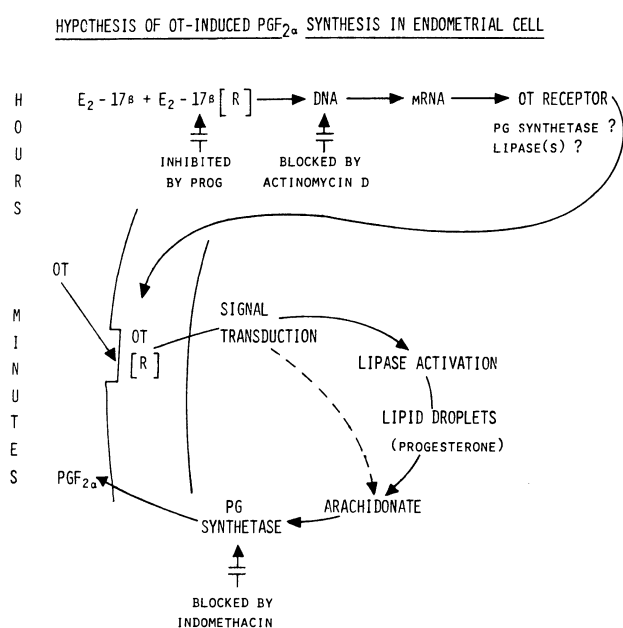


Fig. 1. Model for the regulation of the ovine endometrial OT receptor by E and P. (From: McCracken et al. 1995).

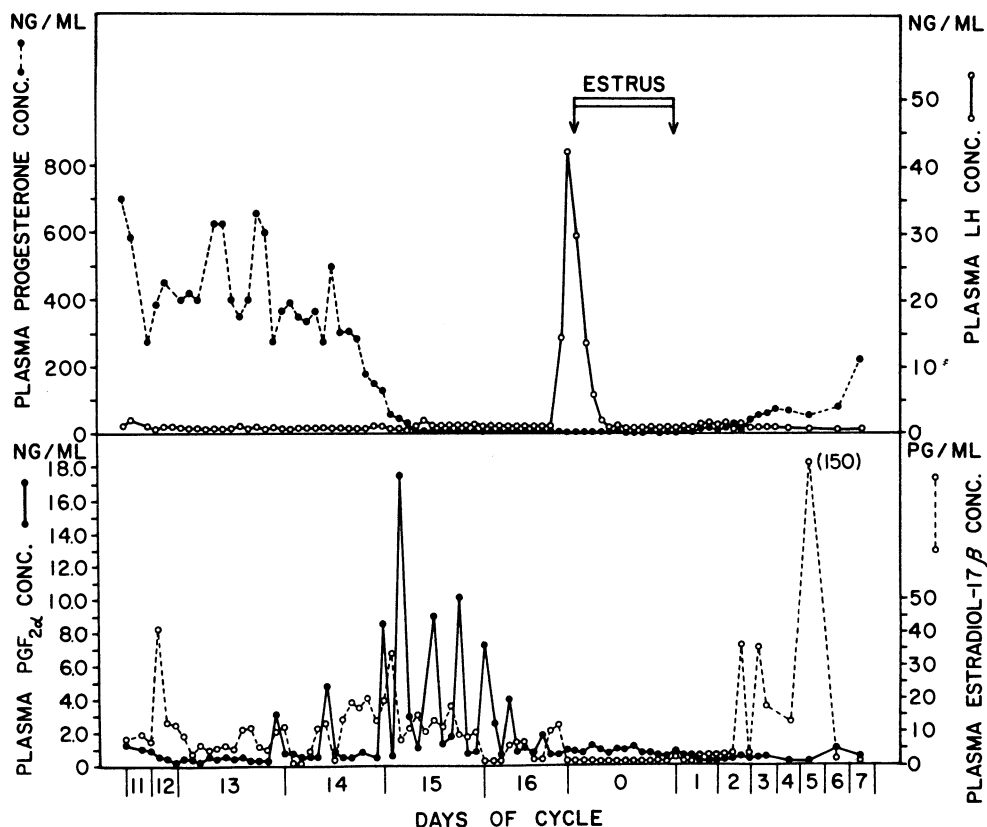


Fig. 2. Concentration of $\text{PGF}_{2\alpha}$, estradiol-17 β , progesterone and LH in utero-ovarian vein plasma samples collected every 2 h from sheep with utero-ovarian autotransplant (From: Barcikowski et al. 1974).

given as a continuous infusion (Goding et al. 1972, McCracken et al. 1973). Thus, the pulsatile infusion of $\text{PGF}_{2\alpha}$ is more efficient in causing luteolysis than a constant infusion, suggesting that a specific pulsatile pattern of $\text{PGF}_{2\alpha}$ secretion by the uterus is advantageous for causing luteolysis.

METHODS

Regulation of the pulsatile secretion of prostaglandin $\text{F}_{2\alpha}$ by the central oxytocin pulse generator

Although it was established that E and P indirectly controlled $\text{PGF}_{2\alpha}$ synthesis in the endometrium by regulating the formation of OT receptors (see Fig. 1), the role of circulating levels of OT was unclear. Preliminary evidence indicated that, in addition to controlling endometrial OT receptors, E and P might also regulate endogenous circulating

levels of OT (McCracken 1980). Indeed, that same year it was reported that peaks of neurophysin I/II carrier proteins, co-secreted with OT, were observed synchronously with peaks of PGFM in the peripheral blood of sheep during luteolysis (Fairclough et al. 1980). This finding was subsequently confirmed by the report that peaks of OT occurred synchronously with peaks of PGFM in the peripheral blood of sheep during luteolysis (Flint and Sheldrick 1983).

To assess the role of circulating levels of OT in the regulation of the estrous cycle in sheep, we initially developed a biometric method to measure oxytocic activity in the circulation of conscious sheep in various experimental states (Schramm and McCracken 1982, McCracken et al. 1984a,b). Pressure sensitive probes were inserted within the myometrium of both uterine horns and the connecting catheters were attached to pressure transducers, the outputs of which were recorded on a polygraph.

Since OT causes uterine contractions, this biometric method formed the basis of our early studies on OT bioactivity in sheep. Such methodology was later combined with the measurement of low levels of OT in jugular plasma by a sensitive RIA method (Amico et al. 1981). In ovariectomized sheep we found that small pulses of intramyometrial pressure (IMP) occurred synchronously in both uterine horns with a mean duration of 5.9 min and a pulse interval of 14.2 min (Schramm and McCracken 1982). We also found that low basal levels of E (0.05 $\mu\text{g/h}$) were required to maintain the 20 min frequency of

the small pulses of IMP. Subsequently we established that the small pulses of IMP were caused by intermittent small pulses of OT secreted by the neurohypophysis (McCracken et al. 1984b). This conclusion was based on the finding that the infusion of 0.01 mU OT given over one minute into one uterine artery produced an ectopic pulse of IMP only in the adjacent horn, while a one minute infusion of 2.0 mU OT into a jugular vein elicited an ectopic pulse of IMP in both uterine horns. In addition, the infusion of a potent antagonist of OT (dET₂ Tyr(ET)OVT; Bankowski et al. 1980) in-

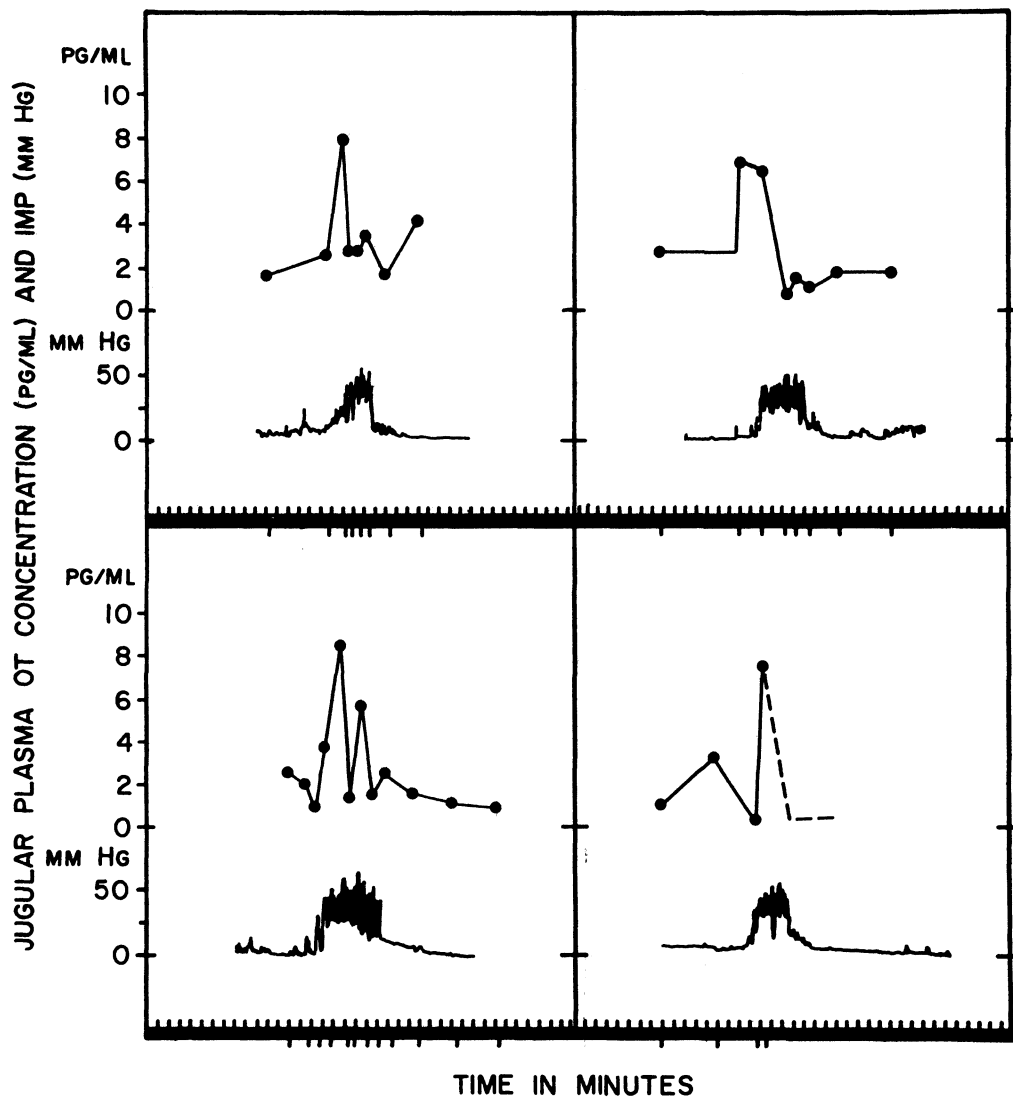


Fig. 3. Oxytocin (OT) concentration in jugular venous plasma sampled at 1 min intervals from conscious intact sheep before, during, and after several endogenous small pulses of intramyometrial pressure (IMP) (From: McCracken et al. 1995).

fused for 30 min into one uterine artery suppressed endogenous pulses of IMP only in the infused horn. Lastly, peaks of OT in jugular plasma (~ 10 pg/ml) were observed to occur synchronously with pulses of IMP (see Fig. 3). In the cyclic sheep, the pulse amplitude and frequency of IMP was high around the time of estrus, but these relatively small pulses of IMP gradually declined during the luteal phase and began to increase again towards the end of the luteal phase around day 13 of the cycle. However, as shown in Fig. 4, in addition to the small 20 min pulses of IMP, large hour-long bursts of IMP were observed to occur at intervals of about 6 h over the period of luteolysis (McCracken et al. 1984a). Measurement of OT in jugular plasma revealed that plasma levels of OT were markedly increased during each of these hour-long bursts of IMP, whereas

plasma levels of vasopressin remained unchanged (Fig. 5). While the concentration of OT during the first large burst of IMP reached about 200 pg/ml of plasma, the peak concentration of OT declined by about 50% during each subsequent burst of IMP. These results suggested that these large intermittent releases of OT, interacting with rising levels of endometrial OT receptors, caused the episodic pulses of uterine $\text{PGF}_2\alpha$ which mediate luteolysis in this species. However, because of the unexpected discovery that the ovine CL contained large amounts of OT (Wathes and Swann 1982) and that luteal OT could be discharged by the systemic administration of an analog of $\text{PGF}_2\alpha$ (Flint and Sheldrick 1982), it was unclear what proportion of the elevated levels of OT, observed during luteolysis in sheep, emanated from the posterior pituitary *versus* the CL.

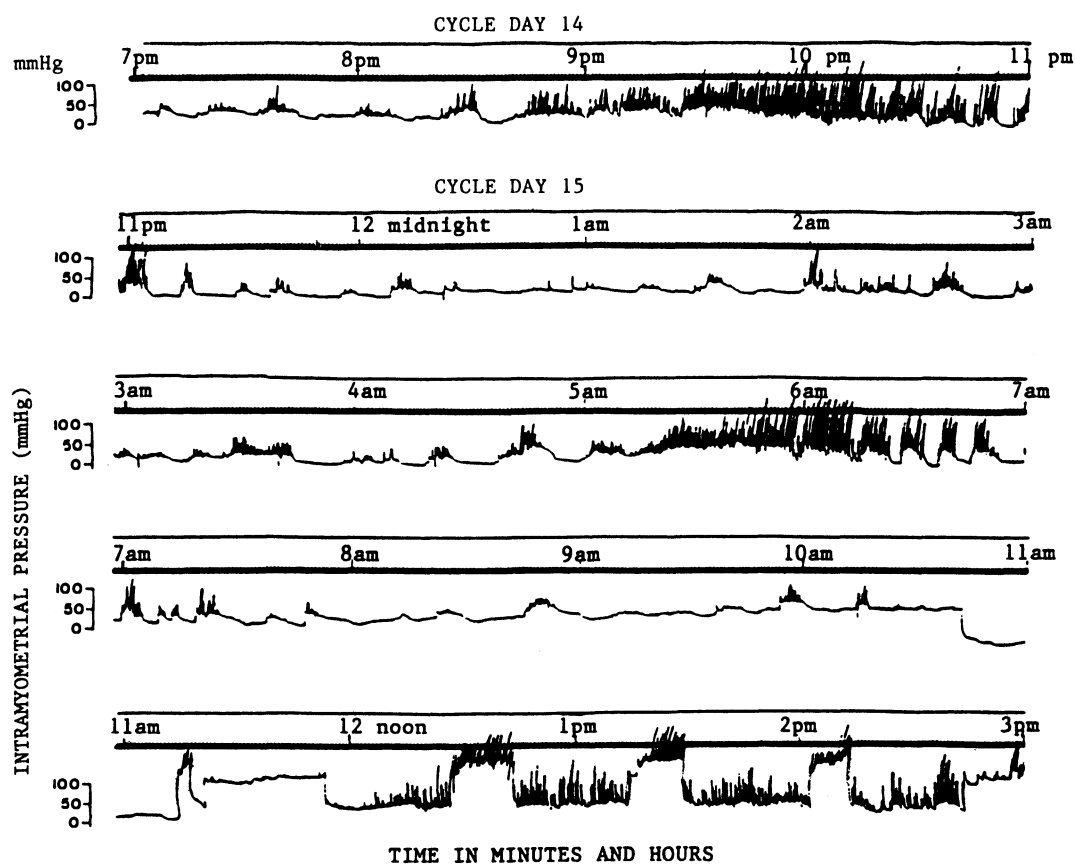


Fig. 4. Intramyometrial pressure changes in the uterus of a conscious sheep on days 14 and 15 of a cycle (recorded continuously over 20 h). In addition to small episodic pulses, three periods of high intensity of contractions occurred at intervals of 5.5 to 6.5 h. (From: McCracken et al. 1984a).

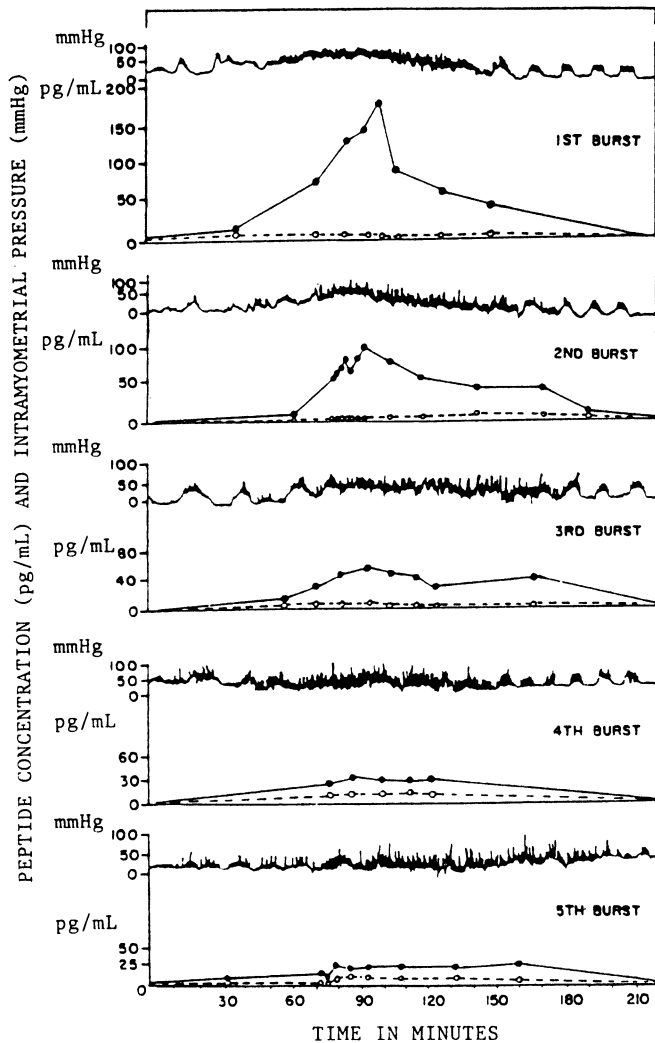


Fig. 5. Peripheral plasma concentration of oxytocin (•) and vasopressin (o) (pg/mL) during successive bursts of intramyometrial pressure (mmHg) observed during luteolysis in sheep. Peak levels of oxytocin declined as luteolysis progressed. (From: McCracken et al. 1995).

RELATIVE CONTRIBUTION OF THE NEUROHYPOPHYSIS TO CIRCULATING LEVELS OF OT AND THE REGULATION OF ITS PULSATILE SECRETION BY E AND P

To distinguish between the relative contributions of the neurohypophysis and the CL, two model systems were employed to exclude the CL as a source of OT. First, in ovariectomized sheep maintained on low E (0.05 µg/h) to preserve the basal frequency of the central OT pulse generator, high E (1.0 µg/h) or P (500 µg/h) were infused to determine their effects on the pattern of OT release

from the neurohypophysis in the absence of the CL (McCracken et al. 1991). Second, in intact cycling sheep, the CL was removed surgically (luteectomy) during the luteal phase of the cycle to exclude the contribution of OT from the CL and, at the same time, subject the animal to a premature withdrawal of endogenous P (McCracken et al. 1995). In ovariectomized sheep, the infusion of high E (1.0 µg/h) for 12 to 36 h, superimposed on low E (0.05 µg/h), elicited a series of 4 to 6 large bursts of IMP, each lasting 1 to 2 h. These large bursts of IMP occurred simultaneously in both uterine horns at intervals of about 3 h, commencing approximately 24 h after beginning the infusion of high E (Table I). The measurement of OT in jugular plasma during these bursts of IMP revealed a series of high frequency small pulses of OT, the largest of which reached 13.4 pg/mL (Fig. 6). The withdrawal of 10 day infusions of P (500 µg/h) superimposed on low E (0.05 µg/h) also evoked a similar series of large bursts of IMP, each lasting 1 to 2 h and beginning about 24 h after P withdrawal (Fig. 7). In intact cycling sheep, the CL (or in one case, twin CLs) was surgically removed on days 6, 8, or 10 of the cycle. In all luteectomized animals, a series of large bursts of IMP was observed similar to those seen following the withdrawal of a 10 day infusion of P in the ovariectomized animal maintained on low E (Table I).

We conclude that an increase in circulating levels of E in the ovariectomized sheep causes the central OT pulse generator to alter its frequency intermittently, thus producing a series of 4 to 6 episodes of rapid small pulses of OT. Similar changes in the frequency of the OT pulse generator is evoked, either by the withdrawal of P superimposed on low E in the ovariectomized animal, or by the withdrawal of endogenous levels of P by surgical removal of the CL during the luteal phase in the intact cycling animal. It appears, therefore, that either increasing circulating levels of E in the ovariectomized animal maintained on low E, or the return of E action after the withdrawal of P, evokes several intermittent episodes of high frequency activity of the neurohypophyseal pulse generator. It should be noted that the interval between the large bursts of

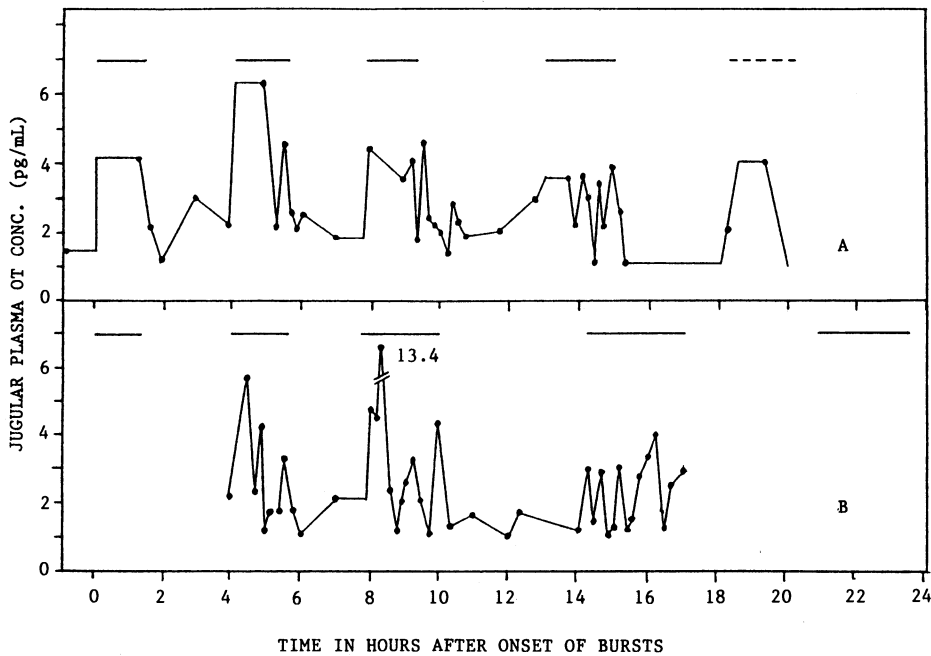


Fig. 6. Jugular concentration of oxytocin in two ovariectomized sheep maintained on low concentration E ($0.05 \mu\text{g/h}$) after the infusion of high E ($1.0 \mu\text{g/h}$) for 12 h (A) or 36 h (B). Horizontal lines mark bursts of intramyometrial pressure. (From: McCracken et al. 1995).

IMP observed in animals subjected to hormonal manipulation is about 3 h (Table I), whereas during natural luteolysis the interval between episodes is about 6 to 9 h (Fig. 4). We explain this difference

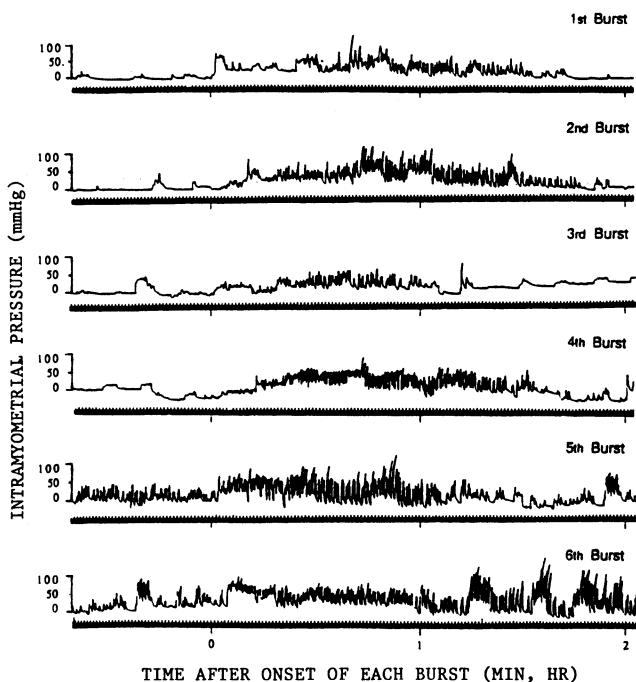


Fig. 7. Large bursts of intramyometrial pressure in ovariectomized sheep maintained on low E ($0.05 \mu\text{g/h}$) following withdrawal of 10 days of P ($500 \mu\text{g/h}$). (From: McCracken et al. 1995).

by the fact that animals undergoing experimental treatments were subjected to very acute changes in E and P levels, whereas during natural luteolysis, endogenous changes in E and P levels would occur more gradually. Thus the slower rate of change in E and P levels in the intact cycling animal may account for the longer intervals observed between the large bursts of IMP. In intact animals following luteectomy (Table I), the mean number of bursts (7.0) was greater than the number seen following the withdrawal of P in the ovariectomized animal maintained on low E (5.3). This may be due to the fact that endogenous levels of E would be expected to increase following luteectomy and, thus, optimize the number of large bursts of IMP.

The concentration of OT in peripheral plasma during the first luteolytic pulse of $\text{PGF}_{2\alpha}$ in the intact cycling animal reaches about 200 pg/ml (Fig. 5) while the levels of OT observed during the bursts of IMP induced by hormonal treatment in the ovariectomized animal is $< 20 \text{ pg/ml}$ (Fig. 6). Thus, it would appear that, at the onset of luteolysis in the intact cycling sheep, the contribution of the neurohypophysis to circulating levels of OT is about 10%, whereas the supplemental contribution from the CL amounts to 90% of the circulating blood levels of OT. However, since the magnitude of the

TABLE I

Effect of infusions of E ($n=3$) or P ($n=3$) in ovariectomized sheep maintained on low E ($0.05 \mu\text{g/h}$) or luteectomy in intact cycling sheep ($n=3$) on large bursts of IMP. (Based on data from McCracken et al. 1995)

| Dose of P ($\mu\text{g/h}$) | Duration (days) | Dose of High E ($\mu\text{g/h}$) | Duration (h) | Mean # Large Bursts IMP | Mean Duration (h) | Mean Interval (h) | Time to 1st Burst of IMP (h) |
|----------------------------------|--------------------|---------------------------------------|-----------------|----------------------------|----------------------|----------------------|---------------------------------|
| | | | | | | | From (+) E or (-) P |
| - | - | 1.0 | 12-36 | 3.7 | 1.8 | 3.2 | 22.8 |
| 500 | 7.5-10 | - | - | 5.3 | 1.6 | 3.6 | 23.6 |
| | | | | | | | From Luteectomy |
| Luteectomy | D. 6-10 | - | - | 7.0 | 2.1 | 3.3 | 23.9 |

large elevations in plasma OT observed during luteolysis declines by about 50% with each successive episode of OT (See Fig. 5), the relative contribution of OT from the neurohypophysis will be proportionately larger as luteolysis progresses.

REGULATION OF THE SUPPLEMENTAL SECRETION OF OT FROM THE CL

Several studies indicated that $\text{PGF}_2\alpha$ could evoke the secretion of OT from the ovine CL. This conclusion was based on the observation that releases of OT from the CL usually occurred synchronously with pulses of $\text{PGF}_2\alpha$ from the uterus (Flint and Sheldrick 1983, Hooper et al. 1986, Moore et al. 1986). In addition, a $\text{PGF}_2\alpha$ analog given systemically caused the secretion of ovarian OT (Flint and Sheldrick 1982). Also the systemic administration of high levels of $\text{PGF}_2\alpha$ in intact ewes increased peripheral levels of OT-associated neurophysin, but not in ovariectomized ewes (Watkins and Moore 1987). Thus, there was evidence that $\text{PGF}_2\alpha$ could stimulate luteal OT secretion. To examine more closely the effect of $\text{PGF}_2\alpha$ on luteal OT secretion, we gave brief infusions of very low levels of $\text{PGF}_2\alpha$ (5 to 100 pg/min) intra-arterially into the ovary of conscious sheep. We found that these low levels of $\text{PGF}_2\alpha$ evoked the secretion of luteal OT without any effect on P secretion (Lamsa et al. 1989). When these subluteolytic levels of $\text{PGF}_2\alpha$ were given continuously, luteal OT was secreted only for one hour followed by desensitization and recovery of the re-

sponse after 6 to 9 h (Lamsa et al. 1992). In subsequent studies, we found that immediately after desensitizing the high affinity $\text{PGF}_2\alpha$ receptor with a subluteolytic infusion of $\text{PGF}_2\alpha$ (100 pg/min for 2 h), a luteolytic level of $\text{PGF}_2\alpha$ (2,500 pg/min for 2 h), not only evoked an additional hour-long secretion of luteal OT, but also now caused P secretion to decline (Custer et al. 1995a,b, see Fig. 8). We concluded that this dual sensitivity of the CL to $\text{PGF}_2\alpha$ was due to the existence of high and low affinity states of the $\text{PGF}_2\alpha$ receptor in the CL. Moreover, the small increases in circulating levels of OT caused by the periodic increase in the frequency of the OT pulse generator would be expected to stimulate the low levels of uterine $\text{PGF}_2\alpha$ secretion which we have demonstrated will initiate the large supplemental release of luteal OT.

DEVELOPMENT OF A MODEL FOR THE ROLE OF THE OT PULSE GENERATOR AS A PACEMAKER FOR THE OVARIAN CYCLE

Based on the foregoing observations, we have developed a model (Fig. 9) to illustrate how the central OT pulse generator acts as a pacemaker for luteolysis and hence, controls the length of the ovarian cycle (see numbers in diagram).

1. Loss of P action occurs both in the hypothalamus and the endometrium resulting in the return of E action in these tissues.

2. Returning E action will (a) stimulate the hypothalamic OT pulse generator to secrete high fre-

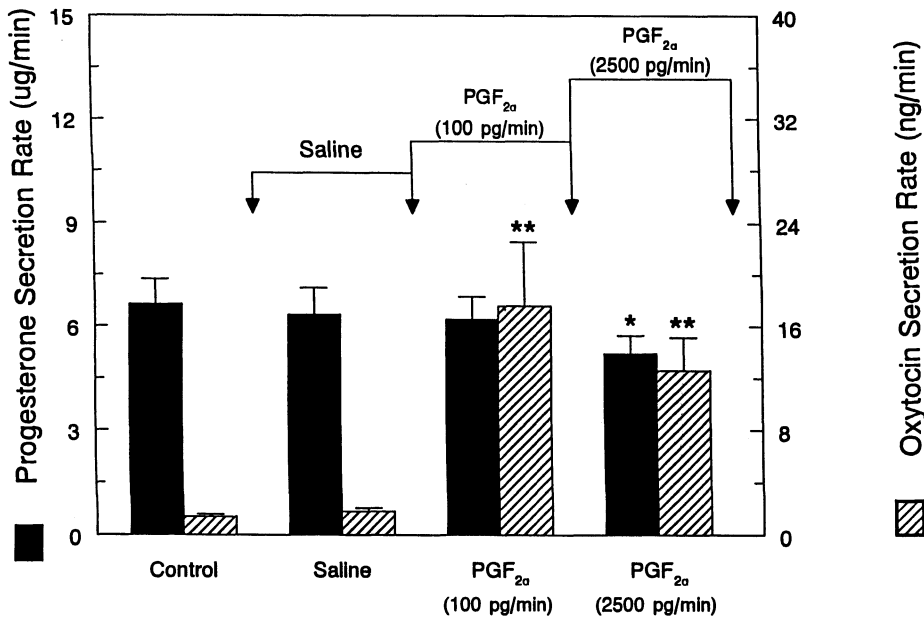


Fig. 8. Secretion rate of OT and P (mean \pm SE, $n=7$ sheep) by the CL of the autotransplanted ovary during the control period and during sequential 2 h intra-arterial infusions of 0.9% saline, 100 and 2,500 pg/min of PGF_{2α}. *Treatment effects on P secretion rate ($P<0.05$). **Treatment effects on OT secretion rate ($P<0.01$). (From: Custer et al. 1995a).

quency bursts of low level OT and (b) simultaneously up-regulate endometrial OT receptors in the uterus.

3. Low level PGF_{2α} (subluteolytic) will be released from the uterus due to the interaction of posterior pituitary OT and endometrial OT receptors.

4. Low level uterine PGF_{2α} will act locally on the ovary *via* the high affinity PGF_{2α} receptors (HFPR) on the large OT-containing cells of the CL to initiate a supplemental release of luteal OT.

5. Such a supplemental release of luteal OT will now amplify the synthesis of endometrial PGF_{2α} to a luteolytic level.

6. The luteolytic level of PGF_{2α} from the uterus will activate the low affinity PGF_{2α} receptor (LFPR) and will now (a) inhibit P secretion (functional luteolysis) and (b) promote the secretion of additional luteal OT, hence reinforcing uterine PGF_{2α} secretion. Such a closed loop system will continue until both affinity states of the PGF_{2α} receptor are desensitized, thus curtailing each supplemental release of luteal OT and terminating the production of each luteolytic pulse of PGF_{2α} from the uterus.

The subsequent release of the next luteolytic pulse of PGF_{2α} will depend on three factors: (1) The next high frequency burst of low level OT from the posterior pituitary *via* the central OT pulse generator. (2) The recovery of the high/low affinity PGF_{2α} receptors in the CL in 6 to 9 h. (3) The recovery of the endometrial OT receptors. The latter may be down-regulated by the supplemental releases of luteal OT, at least in the early stages of luteolysis when the luteal contribution of OT is maximal.

In some species, which do not synthesize large amounts of OT in the CL such as the sow and the mare, uterine PGF_{2α} is also secreted in a pulsatile

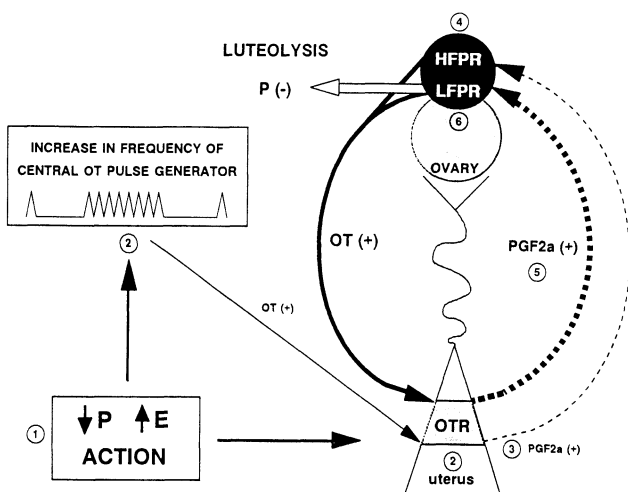


Fig. 9. Model for regulation of uterine luteolytic pulses of PGF_{2α} in sheep. See text for explanation. (From: McCracken et al. 1995).

pattern which may be controlled solely by the central OT pulse generator. Thus, the uterus appears to act as a transducer which converts neural signals (OT pulse generator) into uterine $\text{PGF}_2\alpha$ pulses which are required for luteolysis. In the sheep and other ruminants, luteal OT appears to act as a supplemental source of OT which amplifies these neural signals (OT pulse generator) and hence increases the magnitude of luteolytic pulses of uterine $\text{PGF}_2\alpha$.

DISCUSSION

The regulation of the central OT pulse generator by E and P described above has remarkable similarities to the regulation of endometrial OT receptors by E and P (McCracken 1980, McCracken et al. 1984a). At the end of the cycle, P influence on the endometrium wanes due to the catalytic loss of P receptors by P (Millgrom et al. 1973, Vui Hai 1977). The loss of P action and the consequent return of E action, then up-regulates endometrial OT receptors which, when interacted with OT, will evoke $\text{PGF}_2\alpha$ secretion (McCracken et al. 1995, see also Fig. 1). Both E and P receptors are present in the hypothalamus of most species including rodents (Blaustein et al. 1995), sheep (Lehman et al. 1993), and cats (Bayliss et al. 1991) and P has been shown to down-regulate its own receptor in the hypothalamus (Blaustein and Feder 1979, Moguilewsky and Raynaud 1979). Thus, the general components of the E and P receptor system and their interaction in the uterus are also present in the hypothalamus. Therefore, we propose that as in the uterus, a similar down-regulation of P receptors by P secreted during the luteal phase, will occur in the hypothalamus. Such loss of P action will up-regulate E receptors in critical neurones and, hence, allow circulating levels of E to increase the frequency of the hypothalamic OT pulse generator intermittently. In addition to excitation of hypothalamic neurones (Akaishi and Sakuma 1985), gonadal steroids have been shown to up-regulate OT and vasopressin gene expression in the hypothalamus (Caldwell et al. 1989, Amico et al. 1995), thus potentially amplifying steroid

regulatory effects on the OT pulse generator. Because of the proposed role for the central OT pulse generator as a pacemaker for luteolysis, it might have been expected that section of the pituitary stalk would prevent or delay luteolysis. Rather surprisingly, section of the pituitary stalk in the sheep does not prevent normal cyclic regression of the CL (Denamur et al. 1966, Mallory et al. 1986). However, the above observation is explained by the discovery that within one or two days of stalk section, posterior pituitary hormone secretion returns to normal from the axons proximal to the site of section (Guyton 1991, Makara et al. 1995).

Several other studies support the view that E and P can act simultaneously at the level of the hypothalamus and the uterus. For example, the injection of a luteolytic level of E during the luteal phase of the cycle in sheep causes a premature up-regulation of OT receptors, the appearance of premature $\text{PGF}_2\alpha$ pulses (presumably initiated by the effect of E on the central OT pulse generator) and the premature regression of the CL (Hixon and Flint 1987). Further evidence is seen from a study in which ovariectomized sheep were treated with a regimen of E and P to simulate the levels during the cycle. Pulses of PGFM ($\text{PGF}_2\alpha$ metabolite) were observed in peripheral blood at about the same time and frequency as those observed in intact cycling sheep, but the pulses of PGFM were reduced by about 75% compared to the intact sheep (Silvia and Raw 1993). The appearance of pulses of PGFM in the ovariectomized animals most likely reflects stimulation of uterine $\text{PGF}_2\alpha$ secretion solely by OT derived from the OT pulse generator in these animals. The observed reduction in the magnitude of the pulses of PGFM can be explained by the absence of a supplemental secretion of OT from the CL which, of course, was not present in the ovariectomized animals. It is not known whether these reduced pulses of $\text{PGF}_2\alpha$ (PGFM) would have been adequate to initiate luteolysis had the CL been present. It has been shown in the cow that a 60 to 75% reduction of OT in the CL does not prevent luteolysis in this species (Kotwica and Skarzynski 1993). However, the remaining OT in the CL could have contributed to the

magnitude of the pulses of PGF₂α which presumably occurred in these animals.

CONCLUSIONS

We conclude that termination of the ovarian cycle *via* the luteolytic action of uterine PGF₂α secretion is mediated by the action of the ovarian steroid hormones E and P, not only by up-regulating endometrial OT receptors, but also by regulating the central OT pulse generator. Thus, in addition to the well established interdependence of ovarian steroid hormones and the anterior pituitary gonadotrophins (FSH, LH and prolactin) required to initiate follicular growth, ovulation, and CL function, a second interdependence also exists between ovarian steroid hormones and the posterior pituitary hormone OT which is required to terminate the cycle. Thus, for both the initiation and termination of the reproductive cycle, there is now good evidence for a close interaction between the ovary and the brain.

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