

Prostaglandin $F_{2\alpha}$ -induced Prolactin Release and Luteolysis in the Goat

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Abstract

Injection of prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$) initiated a significant increase in plasma prolactin levels in all goats except those in anoestrus. Luteolysis occurred in non-pregnant goats during the mid luteal phase when the goats were given $PGF_{2\alpha}$ either with or without the suppression of prolactin release by bromocryptine (CB154). Luteolysis and subsequent parturition also occurred in pregnant goats in mid and late gestation after $PGF_{2\alpha}$ injection, with an associated release of prolactin and decrease in plasma progesterone. Acute prolactin release in response to injection of thyrotrophin releasing factor may have had a transient effect on plasma progesterone levels, but did not appear to be luteolytic in either pregnant or non-pregnant goats.

Introduction

Prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$) has been shown to induce premature parturition, prolactin release and lactation in rats (Deis 1971; Vermouth and Deis 1972) and in the human (Smith *et al.* 1972; Yue *et al.* 1974). It also induces prolactin release in heifers (Louis *et al.* 1974) and in both ovariectomized and intact ewes primed with progesterone and oestradiol (Fulkerson *et al.* 1977). In the goat, as in other ruminants, luteal regression and premature parturition has been demonstrated following infusion of $PGF_{2\alpha}$ into the uterine vein (Currie and Thorburn 1973). However, there are conflicting reports of luteal regression following injection of $PGF_{2\alpha}$ into the corpus luteum (Smith *et al.* 1976; Chamley and O'Shea 1976). The present study in the goat was therefore undertaken to obtain information on prolactin release following injection of $PGF_{2\alpha}$ and on any subsequent influence of acute prolactin release on luteal function.

Materials and Methods

Mature female goats with a known reproductive history were used. The goats were kept in single pens, were used to frequent handling and could be bled from the jugular vein with a minimum of restraint. Samples were collected into heparinized tubes and after centrifugation they were stored at -20°C until analysis.

$PGF_{2\alpha}$ (Upjohn) was injected intramuscularly (10 mg per goat). Thyrotrophin releasing factor (TRF) (Roche) was diluted in 0.9% NaCl (1:1, v/v) and 20 μg was injected intravenously. CB154 (2-bromo- α -ergocryptine, Sandoz) and an equal amount of tartaric acid were dissolved in 70% (v/v) ethanol and 0.9% NaCl solution (40:60, v/v) to give an 8 mg/ml solution for subcutaneous injection of 8 and 4 mg. Pregnant mare serum gonadotrophin (PMSG) (Intervet) was injected subcutaneously (400 i.u. in 0.9% NaCl).

Progesterone Assay

Samples of 0.1–0.5 ml plasma were extracted with 6 ml hexane and the progesterone concentrations determined by radioimmunoassay essentially as described by Aso *et al.* (1975). The sensitivity of the progesterone assay was 1.0 ng/ml plasma.

Prolactin Assay

The caprine prolactin assay adopted in this laboratory was based upon the technique developed by Wallace and Bassett (1970) for the radioimmunoassay of ovine growth hormone in plasma. All assays were performed using a 1:10 dilution of plasma in buffer. Diluted samples (0.2 ml) were incubated with 0.5 ml antiserum (1:10 000) for 4 h at room temperature before the addition of 0.1 ml ^{125}I -labelled prolactin, and then incubated at 4°C overnight. Before the free prolactin was absorbed onto talc (1 ml of a 15% talc suspension in buffer) it was necessary to bring the total plasma content to 0.2 ml in all tubes by the addition of pooled goat plasma. By this method a standard curve for ovine prolactin which exhibited parallelism with diluted caprine plasma to 8.0 ng/0.2 ml was obtained with a sensitivity of 0.2 ng/0.2 ml. Results were calculated by the method of Wallace and Bassett (1970).

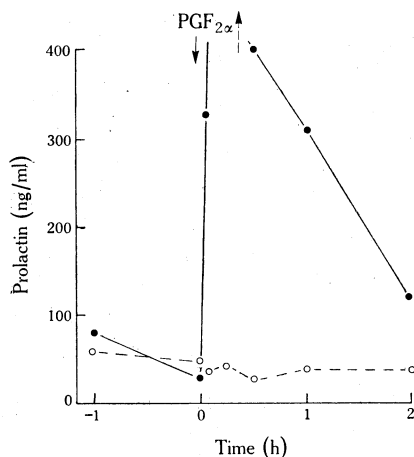


Fig. 1. Changes in mean plasma prolactin levels in response to injection of $\text{PGF}_{2\alpha}$ (unbroken arrow) in three anoestrous goats (\circ) and three goats in oestrus (\bullet).

The antiserum used was raised in turkeys against ovine prolactin (NIH-P-S-II) as caprine prolactin was unavailable; McNeilly and Andrews (1974) have shown that caprine prolactin is immunologically indistinguishable from ovine prolactin in radioimmunoassays. Caprine prolactin levels were expressed in terms of standard ovine prolactin (NIH-P-S-12, 35 i.u./mg). Ovine growth hormone (NIH-GH-S-12) showed a cross reaction of 2% relative to ovine prolactin whilst human prolactin or human growth hormone (Calbiochem) gave negligible cross reaction (<0.1%). The radioactive tracer used was ^{125}I -labelled ovine prolactin (NIH-P-S-12) prepared by the chloramine-T method of Greenwood *et al.* (1963) using ^{125}I from the Radiochemical Centre, Amersham, England.

Results

Prolactin Release in Response to Prostaglandin $F_{2\alpha}$ Injection in Anoestrous Goats and Goats in Oestrus

As shown in Fig. 1 the mean prolactin level in three anoestrous goats fluctuated between 25 and 56 ng/ml plasma and there was no significant increase in prolactin levels when 10 mg $\text{PGF}_{2\alpha}$ was injected intramuscularly. In contrast in three goats in oestrus levels of prolactin rose to ≥ 400 ng/ml within 15 min of $\text{PGF}_{2\alpha}$ injection, and fell to 110 ng/ml 2 h later (see Fig. 1).

Prolactin Release and Luteolysis in Response to Prostaglandin $F_{2\alpha}$ Injection or TRF Injection in Non-pregnant Goats

Five goats were injected with 10 mg $PGF_{2\alpha}$ intramuscularly approximately 9–11 days after oestrus. As shown in Fig. 2a mean levels of prolactin rose to ≥ 400 ng/ml within 15 min of $PGF_{2\alpha}$ injection and were 64 ± 27 ng/ml 2 h later. There was a progressive and complete decline in plasma progesterone levels in all goats from 6.6 ± 1.3 ng/ml (mean \pm s.e.) to negligible levels (< 1 ng/ml) 24 h after injection of $PGF_{2\alpha}$.

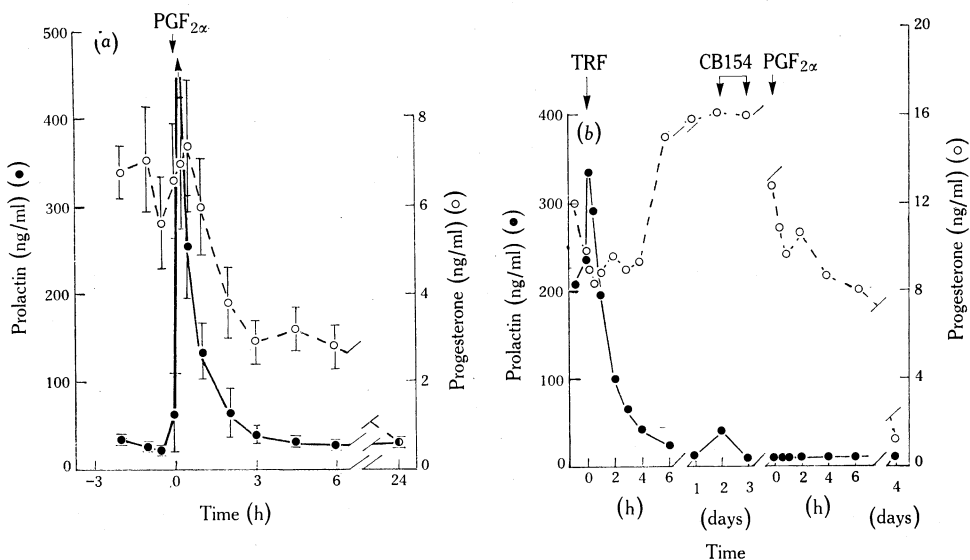


Fig. 2. (a) Changes in mean concentrations (\pm s.e.) of plasma prolactin (●) and progesterone (○) following injection of $PGF_{2\alpha}$ (unbroken arrow) in five non-pregnant goats, 9–11 days after oestrus. (b) Changes in concentrations of plasma prolactin (●) and progesterone (○) in two goats following TRF, and then $PGF_{2\alpha}$ injections with suppression of prolactin release with CB154.

Two other goats were induced to ovulate by injection of 400 i.u. PMSG subcutaneously. Fourteen days later (c. 11 days post-ovulation) 20 μ g TRF was injected intravenously. CB154 was then administered subcutaneously (8 mg then 4 mg) 48 and 72 h after TRF. One hour after the second injection of CB154 10 mg $PGF_{2\alpha}$ was injected intramuscularly. The results are shown in Fig. 2b. Following TRF injections there was an acute release of prolactin, mean plasma levels reaching > 300 ng/ml within 15 min; there was only a transient decline in progesterone levels. In response to $PGF_{2\alpha}$ injection there was a progressive and complete decline in mean progesterone levels from 12.8 to 1.2 ng/ml 24 h after $PGF_{2\alpha}$ even though prolactin release had been suppressed with CB154.

Prolactin Release and Luteolysis in Response to $PGF_{2\alpha}$ or TRF Injection in Pregnant Goats

Two pregnant goats were injected with 10 mg $PGF_{2\alpha}$ intramuscularly and one pregnant goat was injected with 20 μ g TRF intravenously.

Following $\text{PGF}_{2\alpha}$ injection in one pregnant goat of approximately 120 days gestation there was an increase in plasma prolactin from 20 to 190 ng/ml after 60 min, and then a decrease to 115 ng/ml after 3 h. The prolactin level then rose to 258 ng/ml at 33 h before falling to 55 ng/ml at delivery. After a transient increase in progesterone level in the first 90 min following $\text{PGF}_{2\alpha}$, there was a gradual decline from a pre-injection level of 6.4 ng/ml to 1.6 ng/ml 24 h after injection (see Fig. 3a).

Following $\text{PGF}_{2\alpha}$ injection in another pregnant goat of approximately 60 days gestation there was an increase in plasma prolactin from 80 to 200 ng/ml at 15 min, and then a decrease to 38 ng/ml after 3 h. The prolactin level then gradually recovered to 120 ng/ml at delivery. Again there was a transient increase in progesterone in the first 60 min following $\text{PGF}_{2\alpha}$, but plasma progesterone fell from a pre-injection level of 5.1 ng/ml to 0.6 ng/ml after 18 h (see Fig. 3b).

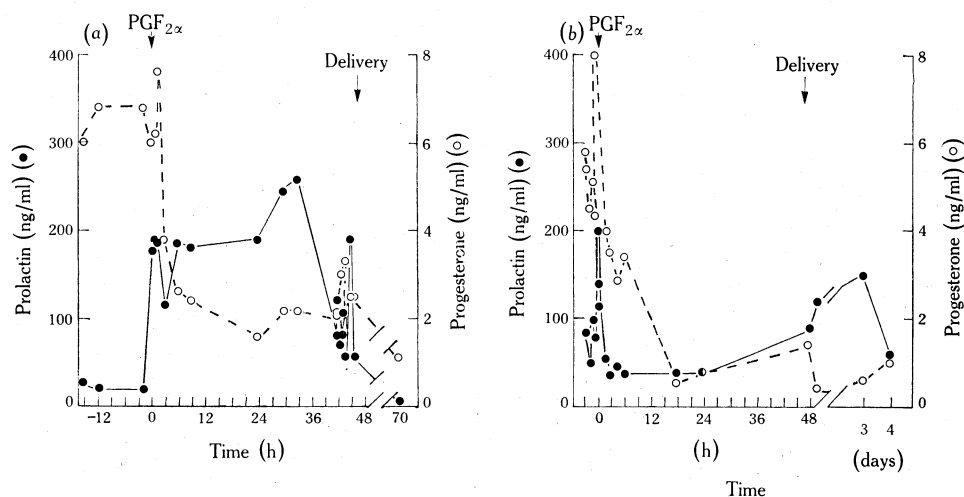


Fig. 3. Changes in concentrations of plasma prolactin (●) and progesterone (○) in (a) one pregnant goat of c. 120 days gestation after injection of $\text{PGF}_{2\alpha}$, and (b) another pregnant goat of c. 60 days gestation after injection of $\text{PGF}_{2\alpha}$.

A third pregnant goat of approximately 120 days gestation was injected with 20 μg TRF intravenously. Plasma prolactin rose from a pre-injection level of 60 ng/ml to >400 ng/ml in 5 min, and remained at this level for 1 h before falling to 270 ng/ml after 2 h and 60 ng/ml after 24 h. Progesterone levels fluctuated between 7.7 and 2.8 ng/ml during this time. This goat continued gestation to term.

Discussion

The results indicate that $\text{PGF}_{2\alpha}$ at a dosage of 10 mg intramuscularly is capable of stimulating an acute release of prolactin in both non-pregnant and pregnant goats, although at the dosage used $\text{PGF}_{2\alpha}$ was unable to elicit increased prolactin release in anoestrous goats. Release of prolactin in female sheep or rats under various experimental conditions was also found to be related to the reproductive state of the animal, being greater during oestrus (Fell *et al.* 1973; Deis and Alonso 1975). This may reflect a change in hypothalamic inhibitory factors in the anoestrous animal,

as $\text{PGF}_{2\alpha}$ may stimulate the secretion of a number of these factors (see Louis *et al.* 1974).

Peak plasma prolactin levels in the goats (> 400 ng/ml) following $\text{PGF}_{2\alpha}$ injections were associated with mean plasma concentrations (\pm s.e.) of $\text{PGF}_{2\alpha}$ and its 13,14 dihydro-15 keto metabolite of 3.0 ± 0.3 ng/ml and 13.6 ± 1.7 ng/ml ($n = 15$) respectively (I. D. Smith and A. H. Clarke, unpublished data). These prostaglandin levels were comparable to those found in the peripheral plasma of parturient ewes (Smith and Clarke 1976). A similar acute release of prolactin in the goat was measured in the peripheral plasma following the administration of $60 \mu\text{g}$ $\text{PGF}_{2\alpha}$ directly into the corpus luteum (see Smith *et al.* 1976) or following the intravenous injection of $20 \mu\text{g}$ TRF to both non-pregnant and pregnant goats. Hart (1973) found similar increases in prolactin levels in the goat at milking.

The major objective of this work was to determine the relationship, if any, of acute hyperprolactinaemia to luteolysis. Since neither blockage of prolactin release by bromocryptine before administered $\text{PGF}_{2\alpha}$ nor induction of hyperprolactinaemia by TRF had a significant effect on corpus luteum function, we did not feel that a study of the response in inert vehicle alone was relevant.

The luteolytic effect of $\text{PGF}_{2\alpha}$ administered directly into the uterine vein has been established (Currie and Thorburn 1973). In this study intramuscular injection of $\text{PGF}_{2\alpha}$ was equally effective. Hart (1973) found that daily injections of CB154 could block prolactin release in milking goats, and thus CB154 was administered to inhibit the prolactin release to $\text{PGF}_{2\alpha}$. It was found that CB154 treatment prior to $\text{PGF}_{2\alpha}$ injection was effective in suppressing prolactin release but did not prevent luteolysis. TRF which is effective in releasing pituitary prolactin in sheep and cattle (Fell *et al.* 1973; Slobodzinski and Wallace 1977) was also effective in releasing prolactin in the goat. As found in this study, however, TRF and the associated release of prolactin produced only a transient effect on progesterone levels. It can be concluded therefore that an acute release of prolactin is not luteolytic in the goat and $\text{PGF}_{2\alpha}$ -induced luteolysis does not require an acute release of prolactin to be effective.

Acknowledgments

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