EFFECTS OF PROSTAGLANDIN $F_{2\alpha}$ ON PITUITARY LUTEINIZING HORMONE CONTENT OF PREGNANT RATS: A POSSIBLE EXPLANATION FOR THE LUTEOLYTIC EFFECT

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Summary. Prostaglandin $F_{2\alpha}$ induced degeneration of embryos in intact pregnant rats but not in ovariectomized rats maintained on progesterone. The pituitary LH stores in the intact, treated rats were significantly higher than those in the pregnant, pseudopregnant or cyclic controls. The prostaglandin failed to inhibit the ovulatory release of LH in the cyclic rat. It is suggested that the luteolytic effect of $PGF_{2\alpha}$ may be mediated in part by the increased synthesis and secretion of LH.

Prostaglandins (PG) are a group of biologically active compounds, many of which are found in the reproductive tract (Pickles, 1967). Recently, $PGF_{2\alpha}$ has been reported to exert luteolytic effects in pseudopregnant rats (Pharriss & Wyngarden, 1969), hysterectomized guinea-pigs (Blatchley & Donovan, 1969) and pregnant rats, hamsters, rabbits and monkeys (Nutting, 1969; Gutknecht, Cornette & Pharriss, 1969; Kirton, Pharriss & Forbes, 1970). These findings have important implications since the rôle of the uterus in limiting the life-span of corpora lutea (CL) in many species may be mediated through the secretion of PG (for discussion, see Rowson, 1970; Kirton *et al.*, 1970). So far, luteolytic properties of PG have been explained solely on the basis of a local effect (Pharriss & Wyngarden, 1969; Gutknecht *et al.*, 1969). Here, evidence is presented to show that $PGF_{2\alpha}$ may also exert a central effect on the pituitary. It is known that luteinizing hormone (LH) can exert both luteotrophic and luteolytic effects in rats and several other species (for review, see Greenwald & Rothchild, 1968), and it was thought that $PGF_{2\alpha}$ might affect CL by modifying the pituitary LH secretion. This prompted an examination of the effect of the compound on the pituitary LH content of pregnant rats of Alderley Park Strain I.

Racemic $PGF_{2\alpha}$ was dissolved in phosphate buffer at pH 7.4 and 0.5 mg or 1 mg/injection/rat was administered subcutaneously (s.c.) twice daily during pregnancy from Days 4 to 7 inclusive (Day 1 = presence of spermatozoa in the vaginal smear). Other conditions were as previously described (Labhsetwar,

1969). The control animals were injected in parallel with vehicle alone. At autopsy on Day 8 of pregnancy, pituitary glands were collected and subsequently assayed for LH by a modification of the Parlow ovarian ascorbic acid depletion method described elsewhere (Labhsetwar, 1969).

The results obtained with both dose levels of $\text{PGF}_{2\alpha}$ were comparable and therefore have been pooled (Table 1). At these dose levels, no side effects were noted and there was no significant loss in body weight (Table 1). The PG considerably reduced the size of the implants (Table 1). Many implantation sites were haemorrhagic and showed degenerative changes upon histological examination, confirming the results of Nutting & Cammarata (1969). Vaginal smears showed typical pro-oestrous changes on the day of autopsy while those

TABLE 1
EFFECTS OF $\text{PGF}_{2\alpha}$ ON NUMBER AND SIZE OF IMPLANTATION SITES IN RATS

Group	No. of rats	Body weight (g)	No. of corpora lutea	No. of implants	Size of implants (mm)	Ovarian wt (mg)	Vaginal smear
Control	6	230 $\pm$ 5	10.8 $\pm$ 0.9	9.7 $\pm$ 1.6	2.8	65 $\pm$ 3	Leucocytic
$\text{PGF}_{2\alpha}$ *	6	225 $\pm$ 9	11.2 $\pm$ 0.6	7.4 $\pm$ 0.9	$\leq$ 1.0	62 $\pm$ 2	Pro-oestrous

Results expressed as Means $\pm$ S.E.

* 0.5 to 1 mg/injection twice daily during pregnancy from Days 4 to 7 inclusive.

from the control rats were still leucocytic. The ovaries of the treated rats contained healthy follicles. At the doses used, the CL showed no pronounced morphological changes when compared with the controls. However, their capacity to secrete progesterone was reduced since the vaginae of the treated rats showed signs of cornification (Plate 1, Fig. b), while those of the controls were mucified (Plate 1, Fig. a). When $\text{PGF}_{2\alpha}$ (0.5 mg/injection; twice daily) was injected from Days 4 to 7 of pregnancy into rats ($N = 3$) ovariectomized on the morning of Day 5 and maintained on progesterone (4 mg/day, s.c. in 0.2 ml oil, from Days 5 to 10), the number and size of implantation sites at autopsy on Day 11 were comparable to those in the similarly ovariectomized rats treated with progesterone alone. Vaginal smears in the treated rats remained leucocytic throughout the experimental period. These results demonstrated that the effects of $\text{PGF}_{2\alpha}$ on the embryos and vaginae of intact rats were mediated through the ovaries by a decrease in the secretion of progesterone.

The pituitary LH content of rats treated with $\text{PGF}_{2\alpha}$ (0.5 or 1 mg/injection; twice daily) was more than 200% higher than that of pregnant controls (Table 2). The LH stores were also significantly higher than those present in the pseudo-pregnant rats (Day 9) or in the 4-day cyclic rats killed on the morning of pro-oestrus when the rat pituitary gland contains LH in amounts higher than that at any other stage of the oestrous cycle. This suggests that increased levels of LH in rats treated with $\text{PGF}_{2\alpha}$ is not secondary to a mere loss of pregnancy. Similarly, these high LH stores cannot be the consequence of disruption of the negative feedback since the ovaries contained healthy follicles which secreted oestrogen in amounts adequate to induce vaginal changes (Plate 1, Fig. b).

PLATE 1

Sections of vaginae (a) of pregnant control rat and (b) of pregnant rat treated with PGF_{2α} (0.5 mg/injection, twice daily during pregnancy from Days 4 to 7 inclusive). Note vaginal cornification in the treated rat as against mucification in the control. The vaginal smear in this particular rat was nearly cornified. $\times 25$.

(Facing p. 156)

TABLE 2
PITUITARY LH CONTENT OF NON-PREGNANT AND PREGNANT CONTROLS, AND OF PREGNANT RATS TREATED WITH PGF_{2α}

Assay no.	Group	No. of rats	LH† concentration (95% error)	LH content (µg/gland)	Relative potency (95% error)	Index of precision (λ)
1	Oestrous control†	5	0.8 (0.5 to 1.3)	9.0	—	0.253
	Pro-oestrous control†	5	3.2 (1.9 to 5.5)	25.0	—	
2	Pseudopregnant control (Day 9)	8	2.8 (1.9 to 4.2)	29.5	—	0.224
3	Ovariectomized§ control	10	3.5 (2.4 to 5.0)	34.6	—	0.150
4	Pregnant control (Day 8)	3	2.6 (1.3 to 5.2)	23.4	100 258 (118 to 563)*	0.307
	PGF _{2α} (1 mg)	3	6.6 (3.1 to 13.9)	78.3		
5	Pregnant control (Day 8)	3	2.6 (1.5 to 4.6)	23.3	100 307 (179 to 527)* 232 (139 to 389)*	0.230
	PGF _{2α} (0.5 mg)	3	8.6 (4.5 to 11.5)	84.5		
	PGF _{2α} (1 mg)	3	6.4 (3.4 to 11.9)	76.1		

* $P < 0.05$.

† µg equivalents of NIH-LH-S-14/mg wet weight of pituitary.

‡ Data from another study (Labhsetwar, 1970).

§ 14 days after ovariectomy.

Even 2 weeks after bilateral ovariectomy, the total LH content ($\mu\text{g/gland}$) of the pituitary was only half that of the intact rats treated with $\text{PGF}_{2\alpha}$ for 4 days (Table 2). These observations taken together support the view that $\text{PGF}_{2\alpha}$ increased pituitary LH content by acting centrally, either directly on the pituitary gland or indirectly through the hypothalamus. The increased stores, however, could result either from an increased synthesis or a decrease in release of LH. The latter alternative was tested by administering $\text{PGF}_{2\alpha}$ (2 mg/injection, s.c.) to 4-day cyclic rats twice on the day before pro-oestrus, when many inhibitors of gonadotrophic secretion block ovulation (unpublished results) or once on the day of pro-oestrus at 11.00 hours, i.e. before the critical period ($N = 3$ to 5 rats/dose). No interference with the normal progression of vaginal smears was observed and a normal complement of tubal ova was found at autopsy on the day of oestrus. This suggests that $\text{PGF}_{2\alpha}$, as used here, has no detectable inhibitory effects on the ovulatory release of LH from the pituitary gland.

It appears from these results that the luteolytic effect of $\text{PGF}_{2\alpha}$ in pregnant rats is accompanied by increased stores of pituitary LH which are significantly higher than those found in the pregnant or non-pregnant rats (Table 2). The luteolytic effect is defined here as a decrease in the secretion of progesterone rather than the morphological degeneration of CL, which is not pronounced in rats even after hypophysectomy (see Greenwald & Rothchild, 1968). Since no evidence for a decrease in LH output was found, it is suggested that the high LH content results primarily from an increased synthesis and is perhaps associated with an augmented secretion. Preliminary observations using radio-immunoassay for rat LH (Niswender, Midgley, Monroe & Reichert, 1968) indicate that plasma levels of this hormone in the pregnant rats treated with $\text{PGF}_{2\alpha}$ are higher than those in rats in the oestrous stage of the cycle. If this can be further validated by more extensive assays, then increased plasma levels of LH can, at least partly, account for the luteolytic effect of $\text{PGF}_{2\alpha}$. It is already known that injection of LH into pregnant rabbits (Stormshak & Casida, 1965), or into pseudopregnant rats with pituitary autografts beneath the kidney capsule (see Greenwald & Rothchild, 1968), exerts luteolytic effects. It is postulated that the luteolytic effect of $\text{PGF}_{2\alpha}$ may be mediated, in part, by augmented synthesis and release of LH.

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