Effects of Estrogen and/or Progesterone on Serum and Pituitary Gonadotropin Levels in Ovariectomized Rats[1] (36942)

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The concept of positive and negative feedback actions of the ovarian steroids on the hypothalamo-hypophyseal axis to alter gonadotropin release is well established (1, 2). Several laboratories have reported changes in pituitary and blood levels of LH, FSH, and prolactin after treatment with estrogen or progesterone in intact or castrated rats. However, the majority of these studies employed bioassay methods for hormone determination, and the results have often been conflicting. This present study was undertaken utilizing the highly sensitive radioimmunoassay (RIA) to evaluate the effects of estrogen, progesterone or the combination of these two steroids on synthesis and release of all three gonadotropins in the same individual ovariectomized animals.

Methods and Materials. Adult virgin Sprague-Dawley (Simonsen) female rats were kept under standard conditions (3) and bilaterally ovariectomized 4–6 weeks prior to experimentation. Rats were primed sc with 50 μg of estradiol benzoate (E), 25 mg of progesterone (P) or both (EP) 72 hr prior to sacrifice by decapitation. At that time body weight was recorded, trunk blood was collected, and the anterior pituitary was removed and weighed. The blood and anterior pituitaries were treated as previously described for subsequent RIA of LH, FSH, and prolactin (3, 4) by the method of Niswender *et al.* (5). All hormones were measured in duplicate, the results averaged and expressed in terms of the *NIAMD* standards. Student's t test was used to analyze the data.

Results. Administration of estrogen either alone or in combination with progesterone markedly increased pituitary weight when compared with non-primed or P-primed animals, significant at the 0.001 level (Table I). This accounted in part for the increased pituitary content of each of the three gonadotropins ($p < 0.02$) in OVX-E and OVX-EP groups when compared with OVX rats (Figs. 1–3). Pituitary concentrations of these hormones were also elevated in E- and EP-primed animals; increases significant at the 0.01 and 0.005 levels, respectively, for LH (Fig. 1) and prolactin (Fig. 3) when compared with the OVX groups. Progesterone administered with E did not alter the pituitary concentration or content of any of the 3 gonadotropins when compared with E-primed rats (Figs. 1–3) nor did it change pituitary LH concentration when administered alone and compared with the non-primed group (Fig. 1). However P itself did cause a significant ($p < 0.01$) elevation in pituitary FSH concentration and content (Fig. 2) and a significant ($p < 0.01$) depression in pituitary prolactin concentration (Fig. 3).

Estrogen caused a moderate depression ($p < 0.05$) in circulating LH levels, which were reduced ($p < 0.001$) even further when P was also administered (Table I). Although E reduced ($p < 0.005$) FSH levels in the blood the combined treatment of E + P did not decrease FSH levels further. In contrast, E and E + P caused a significant elevation in serum prolactin ($p < 0.05$). When administered alone progesterone had no effect on circulating levels of any of the three gonad-

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TABLE I. Effects of Ovarian Steroids on Anterior Pituitary Weight and Serum Gonadotropin Levels in Ovariectomized Rats.

Group[a,b]	Final body wt (g)	Anterior pituitary wt (mg)	Serum hormone (ng/ml)		
			LH	FSH	Prolactin
OVX	328 ± 23[c]	12.7 ± 1.0	554 ± 18	1558 ± 99	15 ± 1
OVX + E	324 ± 19	16.5 ± 0.9	330 ± 88	1028 ± 88	35 ± 7
OVX + P	372 ± 11	12.6 ± 0.8	626 ± 126	1726 ± 171	14 ± 3
OVX + EP	347 ± 11	17.8 ± 0.8	94 ± 18	1102 ± 102	33 ± 7

[a] Rats ovariectomized for 4–6 weeks were injected sc with 50 μg of estradiol benzoate (E), 25 mg of progesterone (P) or both 72 hr prior to decapitation.

[b] Each item represents the mean of 8 rats except that serum LH and prolactin levels were measured in 13 and 4 animals, respectively, and FSH levels in OVX-EP rats were measured in 16 cases.

[c] Mean ± SEM.

otropins.

Discussion. Estrogen stimulates growth of the anterior pituitary by a direct action on pituitary cells (6). In the present study, this growth in size was paralleled by an increase in pituitary stores of all three gonadotropins as well as significant increases in pituitary concentrations of LH and prolactin. Ajika *et al.* (7) have reported that a smaller dose of estrogen (1/10th our dosage) increased the pituitary content but not the concentrations of LH, FSH and prolactin in OVX rats 48 hr after priming. Progesterone was ineffective in altering any of these increments. Pituitary-content data alone provide little information about the dynamic aspects of hormone secretion; an increase in content could reflect 1. increased storage due to diminished release, 2. stimulation of the gland resulting in increased synthesis and release or, 3. reduced *in vivo* destruction of the hormone in the pituitary itself.

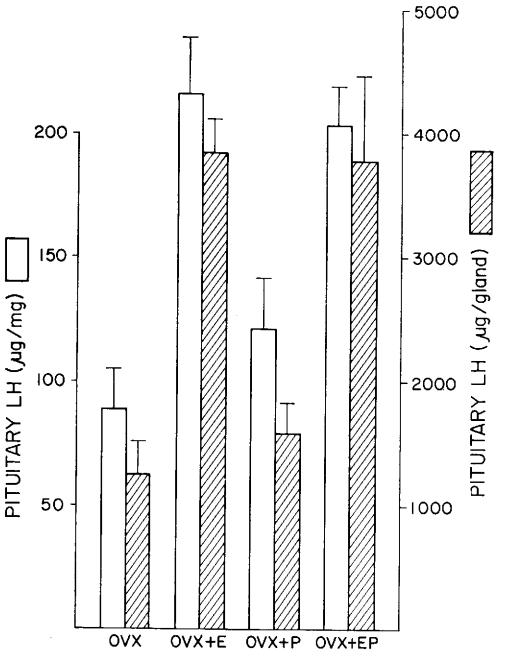

FIG. 1. Pituitary LH concentration (open bars) and total pituitary LH content (shaded bars). LH values are expressed in μg of *NIAMD-Rat LH-RP-1*. Rats ovariectomized for 4–6 weeks were primed with 50 μg of estradiol benzoate (E), 25 mg of progesterone (P) or both 72 hr prior to decapitation. Each bar represents the mean of 4 rats and the lines extending the means represent the SEM.

Prolactin release was enhanced by E with or without P treatment. These findings are in agreement with Kalra *et al.* (8) who reported that E increased circulating prolactin levels. Chen and Meites (9) have similarly reported that chronic E treatment elevates plasma and pituitary prolactin. This effect is probably due to actions at the pituitary level to increase synthesis as well as at the hypothalamic level to depress PIF activity (7, 10) and thus remove inhibition on the synthesis and release mechanisms. However, the interpretation of reduced hypothalamic PIF activity may need to be revised in the light of recent evidence of a prolactin releasing factor in the mammalian hypothalamus (11, 12). Chen and Meites (9) also found that P reduces the E-induced elevation in pituitary

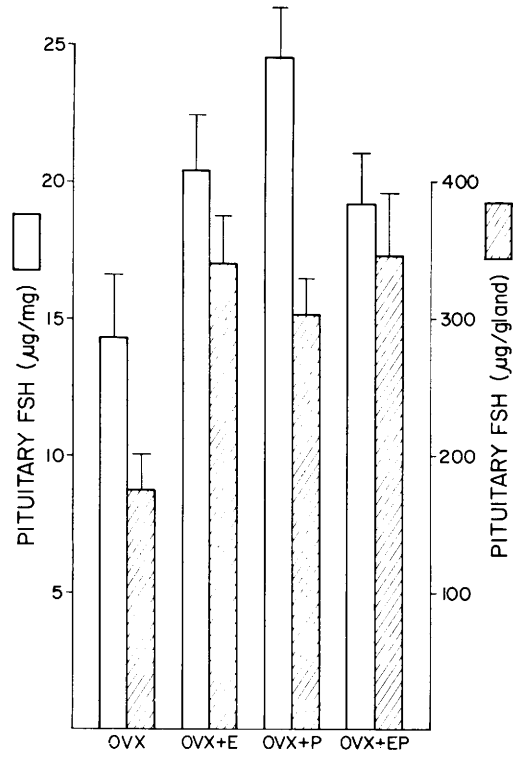

FIG. 2. Pituitary FSH concentration (open bars) and total pituitary FSH content (shaded bars). FSH values are expressed in μg of *NIAMD-Rat FSH-RP-1*. Each bar represents the mean of eight rats except the OVX-P group which represents 7 animals.

prolactin concentration but that this inhibition was masked if the amount of E administered was high as in the present study. They also reported a slight stimulation of pituitary and serum prolactin levels after chronic administration of P, a result not necessarily at variance with the negative findings in our shorter-term study.

In our experiments, the same animals had reduced serum LH levels after treatment with E, and even more dramatic depression in the EP group, even though pituitary LH was elevated. This reflects inhibition of release of LH-releasing hormone (LH-RH) from the hypothalamus, resulting in a build-up of pituitary stores. This interpretation is supported by evidence that exogenous LH-RH releases far more pituitary LH in OVX rats primed with E or EP than in non-primed or P-primed animals (13). The reduction in LH-RH release may be due to depressed hypothalamic content of the releasing hormone following estrogen treatment (7, 14, 15). LH-RH is present in high concentrations in the medial basal hypothalamus (7, 14, 15) and a direct negative feedback action of estrogen on this region has been established (1). It is well known that progesterone synergizes with E in central nervous mechanism (16) and it is quite possible that P synergizes with E to deplete LH-RH stores even further.

LH-RH stimulates release of a minimal amount of FSH *in vivo* (17), but evidence is now mounting that there may be a separate releasing factor for FSH (*e.g.*, 18, 19). Even though estrogen affects pituitary and serum FSH in a manner similar to its action on LH, a dichotomy exists since P is equally as effective as E in raising pituitary FSH but not LH. Additionally, P synergizes with E in reducing serum LH levels but not those of

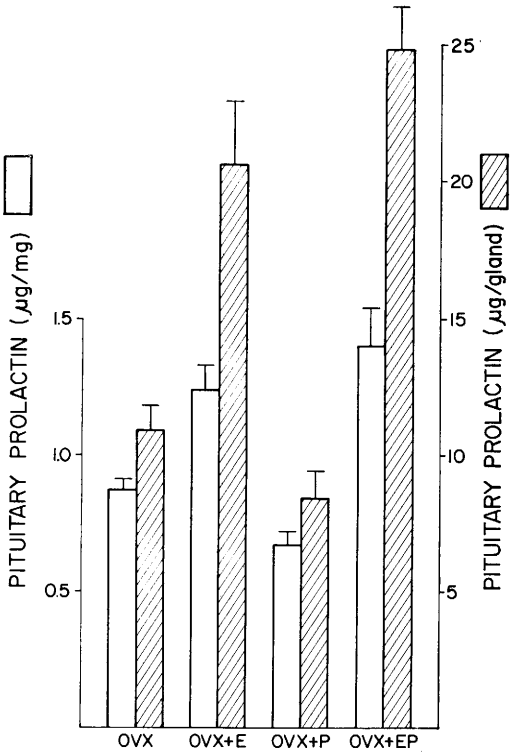

FIG. 3. Pituitary prolactin concentration (open bars) and total pituitary prolactin content (shaded bars). Prolactin values are expressed in μg of *NIAMD-Rat Prolactin-RP-1*. Each bar represents the mean of eight rats.

FSH. It would appear that both steroids act at the pituitary to increase synthesis of FSH and that E but not P has a partial inhibitory action on FSH-RH release from the medial basal hypothalamus. This is supported by evidence that estrogen treatment lowers the hypothalamic stores of FSH-RH (20).

Summary. Pituitary concentration and total content of LH, FSH and prolactin as well as serum levels of the hormones were measured in rats ovariectomized for 4–6 weeks and either untreated or primed with estrogen, progesterone, or both, 72 hr prior to decapitation. Estrogen stimulated synthesis and release of prolactin whereas progesterone did not alter these processes. Estrogen alone or in combination with progesterone caused increased pituitary stores of LH, perhaps by reducing endogenous LH-RH. Either steroid, administered alone or in combination with the other, increased the pituitary content of FSH, and FSH release was only partially inhibited by estrogen or estrogen and progesterone.

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