

Effects of Reserpine and Serotonin on Milk Secretion and Mammary Growth in the Rat.* (25016)

JOSEPH MEITES, C. S. NICOLL AND P. K. TALWALKER†

Department of Physiology and Pharmacology, Michigan State University, East Lansing

Reserpine can induce mammary growth and initiate lactation in the estrogen-primed rabbit(1), and increase prolactin content of rabbit pituitary(2). Reserpine has also been observed occasionally to elicit breast growth and lactation in women(3). Attempts by us to induce mammary growth and milk secretion in rats with reserpine have been unsuccessful, although reserpine prolonged mammary secretion in postpartum rats after litter removal(4, 5). Recently we reported that epinephrine and acetylcholine produced mammary growth and milk secretion in the rat following estrogen administration, but were ineffective when given alone(6). It was of interest, therefore, to see whether reserpine could similarly induce mammary development and secretion in the estrogen-primed rat. The effects of serotonin on mammary function were also determined, since this substance is released from the brain following reserpine administration(7). It was also of interest to compare both these drugs for their ability to retard mammary involution after litter removal in postpartum rats.

Methods. Mature female albino rats (Carrworth) weighing 220-250 g were divided into uniform groups of 5 or 10 each and treated according to regimen in Table I. Virgin rats were injected subcutaneously once daily in 0.1 cc volumes as follows: saline, 0.9% solution; estradiol,‡ 10 µg; reserpine,§ 25 µg; serotonin creatinine sulfate, 0.5 mg serotonin base/kg BW. In postpartum rats the litters were removed on 4th day of lactation, and the

mothers were injected subcutaneously for 10 days with the following: saline, 0.9% solution; reserpine, 10 or 20 µg; serotonin creatinine sulfate, 0.5 mg serotonin base/kg BW. All rats were killed on day after last injection, and the right and left inguinal mammary glands were removed for histological examination. Mammary glands were fixed in Bouin's fluid, sectioned at 6 µ and stained with eosin and hematoxylin. Lactation ratings of 0 to 7 were assigned to each gland, depending on amount of secretion observed microscopically. Percentage of parenchymal as distinct from stromal mammary tissue was estimated from histological sections. From most groups, ovaries, uterus, adrenals, and thymus glands were removed, trimmed, and weighed on Roller-Smith balance.

Results. Table I. The mammary glands of virgin rats given saline had only bare ducts, while estradiol induced limited lobuloalveolar growth which regressed to mostly ducts when followed by saline. No effects were noted with reserpine alone, while estradiol and reserpine together elicited more lobuloalveolar growth than estradiol alone. When estradiol was followed by reserpine, moderate lactation was initiated. Serotonin alone was ineffective, but increased mammary growth when given together with estradiol, and induced marked secretion and mammary enlargement when injected after estrogen. In the non-suckled postpartum rats, reserpine or serotonin partially maintained lactation and retarded mammary involution. Neither drug depressed body growth nor significantly altered weights of ovaries, uterus, adrenals or thymus. Representative mammary sections are shown in Fig. 1-6.

Discussion. These data indicate that estrogen priming is essential for reserpine and serotonin to induce milk secretion and mammary growth in the rat. These drugs were more effective when given after than when given together with estradiol. In virgin rats,

* Published with approval of Mich. Agr. Exp. Sta. as Journal Article no. 2442.

† This work supported in part by NIH grant to senior author.

‡ The estradiol was kindly furnished by Dr. J. O. Reed of Foundation Labs., N. Y. City.

§ We are indebted to Dr. Russel Kraay of Eli Lilly and Co., Indianapolis, Ind., and to Dr. Robert Gaunt of Ciba Pharmaceutical Products, Summit, N. J., for reserpine.

TABLE I. Effects of Reserpine and Serotonin on Mammary Growth and Milk Secretion in 14 Groups of Rats.

Total rats	Treatment*	No. rats with secretion	Avg secretion rating	Avg % mam. parenchyma
Virgin rats				
10	Saline, 15 d	0	0	7
"	Est, 10 d	0	0	18
"	Est, 10 d	0	0	12
	Saline, 5 d			
10	Res, 21 d	0	0	8
5	Est + Res, 10 d	1	<1	35
5	Est, 10 d	4	3	45
	Res, 5 d			
5	Ser, 21 d	0	0	7
5	Castrate + ser, 21 d	0	0	5
5	Est + Ser, 10 d	2	<1	26
10	Est, 10 d	10	>5	57
	Ser, 5 d			
Postpartum rats, litters removed on 4th day				
5	Saline, 10 d	0	0	11
5	Res (10 μ g) 10 d	4	2	21
5	Res (20 μ g) 10 d	4	2	25
5	Ser, 10 d	2	<1	23

* Est = Estradiol. Res = Reserpine. Ser = Serotonin.

estradiol alone induced lobuloalveolar mammary growth, presumably by stimulating ovarian luteal function. Estrogens also increase prolactin content of the rat pituitary, but do not initiate mammary secretion unless ovaries are removed(8). In these experiments the mammary glands regressed following cessation of estradiol injections, but subsequent administration of reserpine or serotonin for 5 days apparently released sufficient amounts of prolactin and perhaps other hormones to initiate milk secretion and maintain the mammary lobuloalveolar system. Apparent increase in mammary parenchymal tissue can be partially attributed to filling of alveoli and ducts with secretion. Ability of these drugs to prolong mammary function in postpartum rats following litter removal is believed to depend on a similar release of prolactin and perhaps other hormones from the pituitary.

The pathways through which reserpine and

serotonin exerted their effects on the mammary gland remain to be determined. Since the hypothalamic portion of brain has been shown to influence secretion of several anterior pituitary hormones(9), it is possible that these drugs activate stimulating or inhibiting centers in the hypothalamus which induce release of pituitary prolactin. There is evidence that the hypothalamus may inhibit secretion of prolactin(10), and since reserpine has been reported to depress certain hypothalamic functions(11), this would remove the inhibition to prolactin secretion. Serotonin may also be involved in this process, since reserpine markedly reduces serotonin levels in the brain (7). There is no evidence that reserpine or serotonin directly influence the anterior pituitary or mammary gland, although this has not been investigated. The effects of these drugs apparently were not mediated through ovaries or adrenals, since dose levels employed did not significantly alter weights of these organs or of uterus and thymus. The possibility cannot be excluded however, that these drugs induced transitory changes in secretion rates by ovaries and adrenals.

Previously only 2 stimuli have been shown capable of initiating lactation and increasing pituitary prolactin secretion, namely certain steroids, particularly estrogens, and the nervous stimulus of suckling or milking(8). We have now demonstrated that 3 other natural body substances, *viz.*, epinephrine, acetylcholine and serotonin, can also initiate mammary secretion. Preliminary evidence suggests that they increase prolactin synthesis and/or release, perhaps through a common mediator in the hypothalamus where they are normally present in high concentrations(9). The lactation-stimulating action of estrogens or suckling may also be mediated through one or more of these neurohormones.

Summary. 1. In mature virgin rats, daily subcutaneous injections of 20 μ g reserpine or .5 mg serotonin creatinine sulfate base/kg BW for 5 days, following daily injections of 10 μ g estradiol for 10 days, initiated milk secretion in 14 out of 15 rats and induced growth of mammary lobuloalveolar tissue. When either drug was injected with estradiol for 10 days, mammary growth was greater

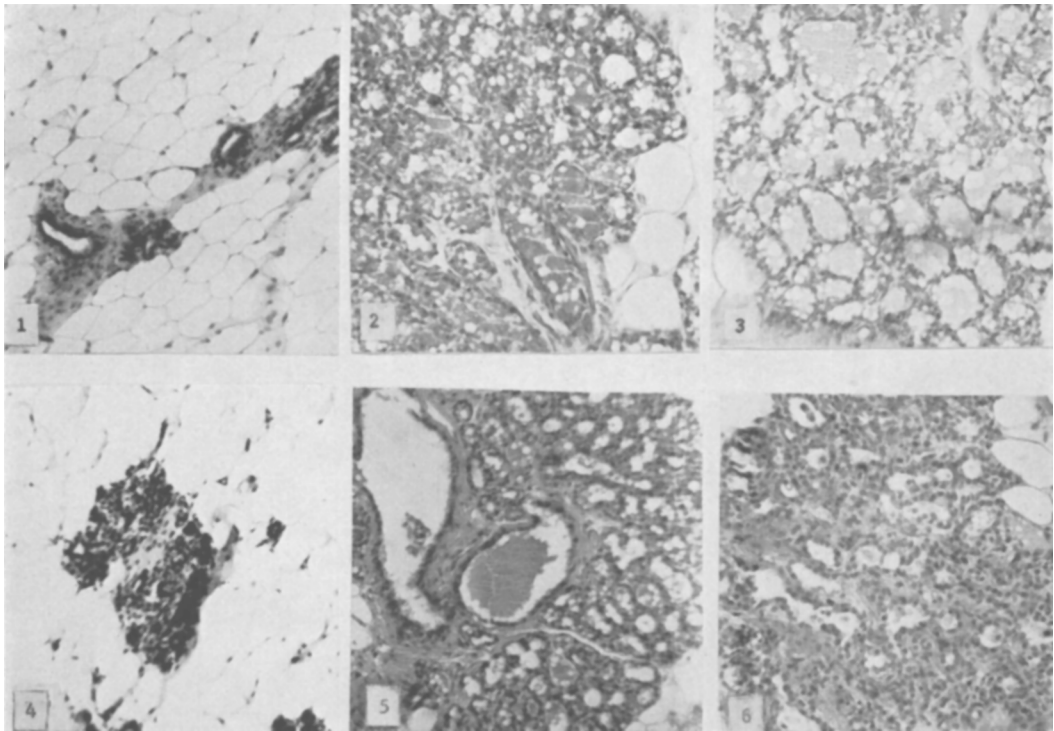


FIG. 1-3. Mammary sections from virgin rats: (1) estradiol 10 d, saline 5 d; (2) estradiol 10 d, reserpine 5 d; (3) estradiol 10 d, serotonin 5 d.

FIG. 4-6. From postpartum rats 10 d after litter removal: (4) saline, (5) reserpine, (6) serotonin. $\times 67$.

than with estradiol alone, but there was little evidence of milk secretion. Injections of reserpine or serotonin alone for 21 days failed to elicit mammary growth or secretion. 2. In rats following litter removal on 4th day after parturition, milk secretion ceased and mammary glands showed pronounced involution 10 days later. Injections of 10 or 20 μ g reserpine or .5 mg serotonin creatinine sulfate base for 10 days following litter removal, retarded mammary involution and maintained secretory function. 3. It is concluded that in estrogen-primed virgin rats or in post-partum rats after litter removal, reserpine and serotonin stimulate secretion of prolactin and perhaps other factors by the pituitary favorable to mammary growth and lactation.

ADDENDUM: Preliminary assays of pituitary prolactin content of rats indicate that epinephrine, acetyl-

choline and probably reserpine and serotonin induce a discharge of prolactin from the pituitary shortly after injection of these drugs.

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Received May 14, 1959. P.S.E.B.M., 1959, v101.