The Effect of Median Eminence (ME) Lesions on Plasma Levels of FSH, LH, and Prolactin in the Rat (35396)

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It is a well-known fact that the secretion of gonadotropins and prolactin in mammals is regulated by the hypothalamic production and release of specific neurohormones (1). The secretion of follicle-stimulating hormone (FSH) and luteinizing hormone (LH) are normally stimulated by hypothalamic releasing factors (2, 3) termed FSH-releasing factor (FRF) and LH-releasing factor (LRF), whereas the secretion of prolactin is normally restrained by a prolactin-inhibiting factor (PIF) (4, 5). When these hypothalamic influences are removed, the secretion of FSH and LH is suppressed while that of prolactin is stimulated. This pattern of gonadotropin secretion has been experimentally produced in rats by surgical separation of the anterior pituitary from the hypothalamus either by means of hypothalamic lesions or by grafting the pituitary to a site distant from the hypothalamus.

In earlier work with hypothalamic lesions the effect on hormone release was estimated by bioassay or by the use of indirect indices of release such as effects on target glands. In the case of FSH and LH, the effect of hypothalamic lesions on plasma hormone levels could only be evaluated after the release of gonadotropins was elevated by castration. Following median eminence (ME) lesions the plasma levels of both FSH (2) and LH (6) decreased as determined by bioassay. In the case of prolactin, the insensitivity of the available bioassay, the pigeon crop sac assay, precluded actual measurement of plasma levels. Since rats with median eminence lesions developed pseudopregnancy and underwent mammary development progressing to mammary secretion, it was inferred that prolactin release was enhanced (7-10); however, it was still possible that these results were caused by a relatively constant low level of prolactin release. With the availability of sensitive radioimmunoassay methods (RIA) for LH, FSH, and prolactin, we decided to reevaluate the effect of hypothalamic lesions on the secretion of gonadotropins and prolactin.

Materials and Methods. Adult female rats (Holtzman) kept in a constant temperature room, with controlled lighting (lights on 5 a.m. to 7 p.m.) were used. Ovariectomy was performed 4–6 weeks before hypothalamic lesions were made in the ME. An anodic current of 3 mA was passed bilaterally for 15 sec using a Nichrome electrode which was insulated except at the tip. Sham-operated animals in which the electrode was lowered into the hypothalamus stopping just short of the ME were used as controls.

Prior to, and 1 week after the lesions, blood was collected from the jugular vein in a heparinized syringe while the rat was lightly anesthetized with ether. Samples were immediately centrifuged and the plasma was separated and frozen at —20° until assayed by RIA. LH was assayed according to the method of Niswender *et al.* (11);[3] prolactin

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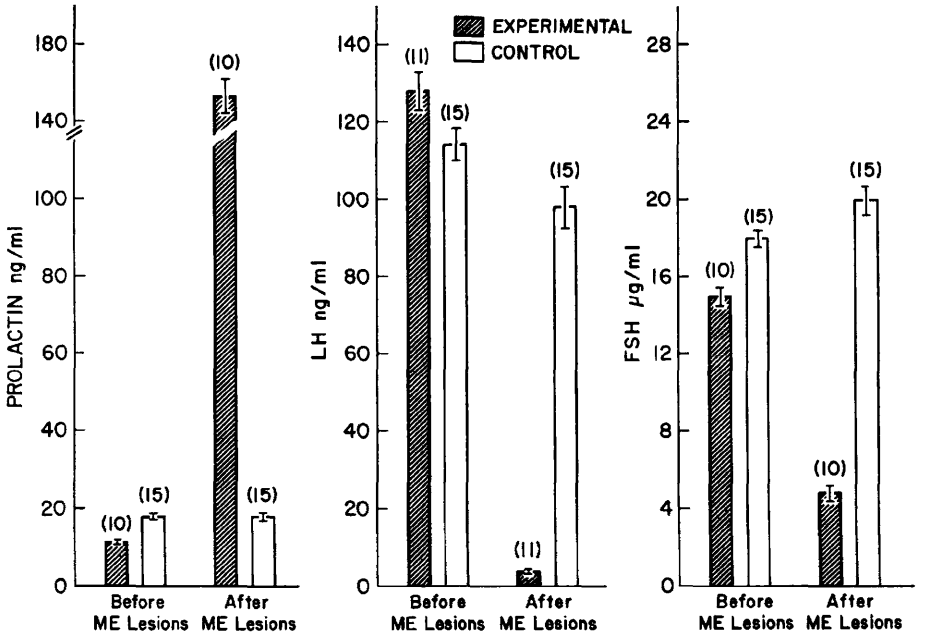

FIG. 1. Plasma prolactin, LH, and FSH in ovariectomized, sham-operated rats (control) and ovariectomized rats with median eminence (ME) lesions (experimental). The height of each column represents the mean value determined just prior to, and 1 week after, placement of lesions. Vertical bars give the standard error of the mean; and number of rats per group is given in parenthesis.

and FSH as recommended in the RIA-NIAMD kits (12).[4]

Results were expressed in terms of NIH LH Sl for LH and the standards supplied in the kits for FSH and Prolactin.

The rats were killed and the brains were fixed in 10% formalin and sectioned at 50 μ in the frontal plane in order to verify the location and extent of the lesions after staining by a Nissl procedure.

Results. As a result of ovariectomy performed 4–6 weeks prior to placement of lesions, all the animals had high initial plasma levels of LH and FSH (Fig. 1). In sham-operated rats, in which the electrode was lowered into the hypothalamus without passage of current, there was a slight but nonsignificant decrease in both FSH and LH on measurement 1 week after operation.

In striking contrast to the lack of effect of sham operation, lesions in the ME resulted in a dramatic, 32-fold, fall in LH levels ($p<.001$) to values which were bearly measurable. In the case of FSH there was also a highly significant ($p<.001$) decline in concentration in rats with lesions, but the values decreased only 3-fold instead of the 32-fold decline observed with LH. Plasma prolactin values were unchanged by sham lesions but increased dramatically ($p<.001$), approximately 15-fold, 1 week after ME lesions.

Examination of the lesions in each rat, both grossly and histologically, revealed that they were quite uniform from animal to animal. The lesions were symmetrical and extended to the base bilaterally, frequently resulting in a hole just lateral to the median eminence that included the arcuate nucleus and part of the ventromedial nucleus. They commenced at the caudal border of the optic chiasm and extended to the premamillary region. The basal tissue between the two lesions in each animal was completely replaced by abnormal tissue containing an

[4] Antirat FSH and prolactin antisera and rat hormones were provided through the NIAMD-National Institutes of Health, Pituitary Hormones Program.

abundance of glial cells. No recognizable median eminence tissue or portal vessels were seen.

Discussion. The present data dramatically reinforce previous evidence for the suppression of FSH (6) and LH release (7) by ME lesions and determine quantitatively the extent of the decrease in plasma levels. They show clearly that such lesions lead to a dramatic enhancement of prolactin release. The degree of suppression of FSH release was less marked than that observed with LH which may suggest that the pituitary can secrete some FSH autonomously in the absence of hypothalamic control. Another possibility would be that the lesions were incomplete. This is unlikely since examination of serial sections through the lesions failed to reveal any normal ME tissue.

The dramatic elevation in prolactin levels removes any doubt that prolactin release is markedly enhanced following removal of hypothalamic inhibitory control. The levels of prolactin observed were nearly as high as those observed during proestrus (13) and following suckling in lactating rats (14) which appear to evoke near maximal prolactin release. Since the completion of this study, a paper has appeared (15) in which lesions in the ME were similarly reported to elevate prolactin levels. The results from the two laboratories are in good agreement.

Summary. Lesions were located in the ME of ovariectomized rats to determine their effect on plasma levels of FSH, LH, and prolactin as determined by RIA. Initial levels of FSH and LH were elevated as a result of removal of steroid feedback from the ovaries. One week after placement of lesions plasma LH levels had fallen, 32-fold, to very low values. There was a lesser, 3-fold, fall in FSH, and a dramatic, 15-fold, rise in prolactin to levels nearly as high as those observed in lactating rats and in females in proestrus. In sham-operated controls, in which the electrodes were lowered into the hypothalamus stopping short of the ME, no significant changes were seen in the plasma levels of all three hormones. The results demonstrate that the release of LH and to a lesser extent FSH is decreased after removal of hypothalamic control, whereas the release of prolactin is greatly enhanced.

1. McCann, S. M., and Porter, J. C., Physiol. Rev. **49**, 240 (1969).

2. Igarashi, M., and McCann, S. M., Endocrinology **74**, 446 (1964).

3. McCann, S. M., Taleisnik, S., and Friedman, H. M., Proc. Soc. Exp. Biol. Med. **104**, 432 (1960).

4. Pasteels, J. L., C. R. Acad. Sci. **253**, 3074, (1961).

5. Talwalker, P. K., Ratner, A., and Meites, J., Amer. J. Physiol. **212**, 157 (1963).

6. Taleisnik, S., and McCann, S. M., Endocrinology **68**, 263 (1961).

7. McCann, S. M., and Friedman, H. M., Endocrinology **67**, 597 (1960).

8. Gale, C. C., Taleisnik, S., Friedman, H. M., and McCann, S. M., J. Endocrinol. **23**, 303 (1961).

9. De Voe, W. F., Ramirez, V. D., and McCann, S. M., Endocrinology **78**, 158 (1966).

10. Nikitovitch-Winer, M. B., Mem. Soc. Endocrinol. **9**, 70 (1960).

11. Niswender, G. D., Midgley, A. R., Monroe, S. E., and Reichert, L. E., Proc. Soc. Exp. Biol. Med. **128**, 807 (1968).

12. Parlow, A. F., Daane, T. A., Schally, A. V., Program 51st Meet. Endocrine Soc. **1969**, 83.

13. Kalra, P. S., Krulich, L., Quijada, M., Kalra, S. P., Fawcett, C. P., and McCann, S. M., Int. Congr. Hormonal Steroids, 3rd, in press.

14. Krulich, L., Kuhn, E., Illner, P., and McCann, S. M., Fed. Proc., Fed. Amer. Soc. Exp. Biol. **29**, 579 (1970).

15. Chen, C. L., Amenomori, Y., Lu, K. A., Voogt, J. L., and Meites, J., Neuroendocrinol. **6**, 220 (1970).

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