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EFFECTS OF AMYGDALOID LESIONS UPON LUTEINIZATION OF OVARIES IN RATS SHOWING SPONTANEOUS PERSISTENT VAGINAL ESTRUS

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Abstract

Bilateral electrolytic lesions were placed in the amygdala in the 15 female Wistar/Tw rats showing spontaneous persistent vaginal estrus. At sacrifice performed three weeks later, 9 out of the 15 rats formed 5–36 corpora lutea in their ovaries, while the other 6 had no corpora lutea. Examination of the lesioned brain revealed no differences in the location of the lesions between rats bearing ovaries with corpora lutea and those without. In any of the 15 rats, lesions were invariably located in an area including the nucleus amygdaloideus corticalis, the nucleus amygdaloideus medialis, and the nucleus amygdaloideus basalis pars lateralis.

These findings seem to suggest that the amygdala, or at least a part of the amygdala, is involved in exerting an inhibitory influence upon the gonadotropin-secreting mechanisms of the hypothalamo-hypophyseal system in the female rat.

Recent experimental findings suggest that the amygdala, an integral part of the rhinencephalic-limbic complex, is involved in the regulation of gonadotropic function of the hypothalamo-hypophyseal system. However, conflicting results have been presented as to the nature of the amygdaloid influence upon the secretion of gonadotropins. In the rat, lesioning of the amygdala or severing of the stria terminalis, a major neuronal connection between the amygdala and the hypothalamus, advances the onset of puberty (Elwers and Critchlow, 1960; 1961), whereas stimulation of the amygdala delays the puberal onset (Bar-Sela and Critchlow, 1966). Synthesis and release of luteinizing hormone (LH) in the adult rat were found to be significantly greater in those bearing lesions of the amygdala than in the non-lesioned animals (Lawton and Sawyer, 1970). All these findings seem to suggest that the amygdala is involved in an inhibitory influence on gonadotropin secretion. By contrast, it has been reported that, in the rats showing persistent vaginal estrus, stimulation of the amygdala enhances the release of LH from the anterior hypophysis and thus causes luteinization in otherwise anovulatory polyfollicular ovaries of the animals (Bunn and

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Everett, 1957; Velasco and Taleisnik, 1969; Arai, 1971). The onset of puberty in the female rat was also delayed by lesioning of the amygdala (Relkin, 1971). These findings support the view that the amygdala exerts a stimulatory effect upon the gonadotropic function of the hypothalamo-hypophyseal system.

The present paper deals with the effects of bilateral placement of electrolytic lesions in the amygdala upon luteinization of ovaries in the rats showing spontaneous persistent vaginal estrus.

Materials and Methods

Female rats of the Wistar/Tw strain from the colony maintained in the animal quarter of the Zoological Institute, Faculty of Science, University of Tokyo were used for these experiments. They were kept on rat chow (CLEA Co., Tokyo) and water *ad libitum* under controlled temperature and lighting (14 hr/day) conditions. Daily examination of vaginal smears was begun when the rats reached 10 months of age and continued until the termination of experiments. In the present colony of the rats of this strain, spontaneous persistent vaginal estrus usually occurs in a majority of female animals when they reach 10—12 months of age. Each rat which had shown spontaneous persistent vaginal estrus for more than three weeks and had no corpora lutea in the ovaries when checked by laparotomy was oriented in a stereotaxic apparatus under ether anesthesia and bilateral electrolytic lesions were placed in the amygdala by means of stainless steel electrodes. The electrodes, 0.35 mm in diameter, were insulated with Epoxylite to within 0.8 mm of the tip. The rat brain atlas of De Groot (1959) was used as a guide for the placement of the lesioning electrode. An anodal direct current of 2.5 mA was delivered through the electrode for 10 seconds, the cathode being placed on the skin incision. Those animals showing spontaneous persistent vaginal cornification but bearing no brain lesions served as controls.

At sacrifice performed three weeks after the placement of brain lesions, ovaries and adrenals were dissected out, weighed and fixed in Bouin's solution. Sections cut in paraffin at 8 μ were stained with Delafield's hematoxylin and eosin. In ovaries of each rat, corpora lutea, if present, were counted and their diameters were measured by the method similar to that described in a previous paper (Machida, 1969). Brains were also fixed in Bouin's solution, cut into serial sections at 10 μ and stained with thionin for verification of the location of lesions.

Results

Bilateral amygdaloid lesions were successfully placed in 15 persistent-estrous rats. Following placement of the brain lesions, 9 of the 15 rats invariably came into vaginal diestrus within a few days and thereafter showed irregular estrous cycles or prolonged diestrus with occasional interruptions by estrous smears. The other 6 animals ran persistent vaginal estrus until the day of sacrifice. In accordance with these findings, the ovaries of the former 9 rats with vaginal diestrus contained 5–36 well-developed

corpora lutea, measuring 0.5–1.75 mm in diameter (Group 2 in Table 1), while the ovaries of the latter 6 animals showing persistent vaginal estrus were largely composed of follicles of varying sizes and the hypertrophied interstitial tissues, being totally devoid of corpora lutea (Group 3 in Table 1).

Table 1 Effects of amygdaloid lesions upon luteinization of ovaries in rats showing spontaneous persistent estrus.

Group	No. of rats	No. of corpora lutea per rat	Body weight (g)	Weight (mg/100 g B.W.) of	
				ovaries	adrenals
1	9	0	239± 9[1)]	15.0±1.3	25.8±2.2
2	9	19.6±3.8	274±15	18.3±2.1	29.3±3.3
3	6	0	273±11	11.0±0.7*	21.0±0.9*

Group 1: control rats bearing no brain lesions, Group 2: rats with brain lesions and corpora lutea in their ovaries, Group 3: rats with brain lesions but having no corpora lutea in their ovaries.

1) mean ± standard error.

* significantly different from the value of Group 2 ($0.01 < p < 0.02$ for the weight of ovaries, and $0.02 < p < 0.05$ for that of adrenals) and Group 1 ($p < 0.05$ for ovaries and adrenals).

Histological examinations of the lesioned brains revealed, however, that there were no appreciable differences in the location of the brain lesions among the 15 rats. As shown in Figure 1, of the 9 rats with corpora lutea in their ovaries (Group 2), 7 had lesions mainly in the nucleus amygdaloideus corticalis (ACO), the nucleus amygdaloideus medialis (AM), and the nucleus amygdaloideus basalis pars lateralis (ABL) (Figure 1 Aa), while the other two had lesions in ACO, ABL, the nucleus amygdaloideus lateralis pars posterior (ALP), and the nucleus amygdaloideus centralis (AC) (Figure 1 Ab). In these two animals, almost regular 4-day estrous cycles occurred following placement of the brain lesions and their ovaries contained 35–36 corpora lutea. In the remaining 6 rats having no corpora lutea (Group 3), lesions were located in an area similar to that of the majority of the Group 2 rats; ACO, AM, and ABL were more or less destructed (Figure 1 B).

The ovaries of the Group 2 rats were significantly heavier than those of the Group 3 animals ($0.01<p<0.02$), probably due to the presence of corpora lutea in the ovaries of the Group 2 rats. The weight of adrenals was also significantly greater in Group 2 rats than in Group 3 animals ($0.02<p<0.05$). The body weight and the weights of ovaries and adrenals were significantly different between Groups 1 and 3 ($p<0.05$). However, there were no significant differences in the body weight and the weights of ovaries and adrenals between Groups 1 and 2 ($p<0.05$).

Discussion

The result of the present experiments showed that the placement of electrolytic lesions in an area including ACO, AM, and ABL was effective in inducing luteinization

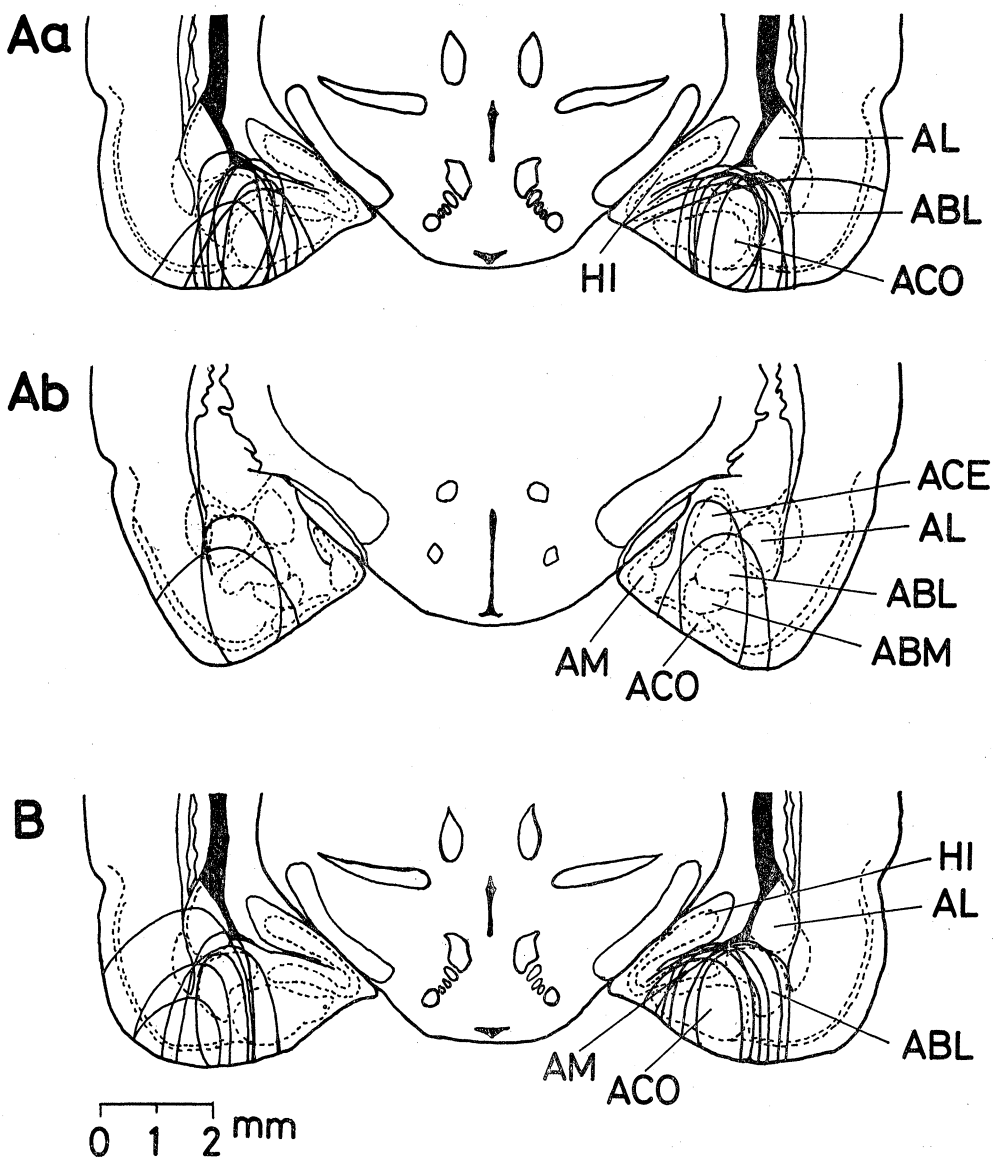

Figure 1. Extent and localization of amygdaloid lesions in rat showing persistent vaginal estrus.

Lesions Aa and Ab caused luteinization in the ovaries and were found in animals of Group 2 in Table 1, while lesion B failed to cause luteinization and were found in Group 3 animals. There were no appreciable differences in the location of lesions among animals in Groups 2 and 3.

ABL: nucleus amygdaloideus basalis pars lateralis, ABM: nucleus amygdaloideus basalis pars medialis, ACE: nucleus amygdaloideus centralis, ACO: nucleus amygdaloideus corticalis, AL: nucleus amygdaloideus lateralis, AM: nucleus amygdaloideus medialis, HI: hippocampus.

in otherwise anovulatory polyfollicular ovaries of the rats showing spontaneous persistent vaginal estrus. These findings seem to suggest that the amygdala, or at least a part of the amygdala, exerts a tonic inhibitory influence upon the gonadotropin-secreting mechanisms of the hypothalamo-hypophyseal system, and therefore the elimination of this structure by electrolytic lesions resulted in an enhanced release of gonadotropins, especially LH, sufficient to cause luteinization in these persistent-estrous rats.

In this connection, it has already been reported that the placement of lesions in the corticomedial portion of the amygdala, *i.e.*, ACO, AM, and ABL, causes the advancement of puberty in the immature female rat (Elwers and Critchlow, 1960), and induces an increase in the synthesis and release of LH in the adult female rat (Lawton and Sawyer, 1970). Lesions in the similar porition of the amygdala likewise enhanced luteinization in the ovaries of the gonadotropin-primed immature rats (Machida, 1972), whereas electrical stimulation of this structure reduced serum LH levels in ovariectomized rats (Ellendorff, Colombo, Blake, Whitmoyer, and Sawyer, 1973). All these findings appear to show that the corticomedial portion of the amygdala is involved in a mechanism normally exerting a tonic inhibitory influence upon the gonadotropin secretion.

On the other hand, several investigators have already presented evidence supporting a stimulatory influence of the amygdala upon the gonadotropin secretion (Bunn and Everett, 1957; Velasco and Taleisnik, 1969, 1971; Kawakami, Terasawa, and Ibuki, 1970; Arai, 1971). These authors generally showed that stimulation of the amygdala enhanced the release of LH and induced ovulation or luteinization in the rat.

In conclusion, it seems that the amygdala has a dual role in the regulation of the gonadotropin secretion in the female rat.

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