# ROLE OF THE LIMBIC STRUCTURES IN THE MECHANISM OF ACTION OF GLUCOCORTICOIDS AND ESTROGENS ON HYPOTHALAMIC CONTROL OF PITUITARY ADRENOCORTICOTROPIC AND GONADOTROPIC FUNCTIONS

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Glucocorticoids (hydrocortisone, dexamethasone) and ACTH depress the excitability of the amygdala of the dog's brain; this effect is more marked with ACTH. Unlike the glucocorticoids ACTH had no direct effect on excitability of the hypothalamic zones studied. Experiments on rats showed that administration of estradiol monobenzoate 4 weeks after injury to the hippocampus stimulated secretion of follicle-stimulating hormone (FSH) and inhibited the secretion of luteinizing hormone (LH). In control animals the secretion of FSH was reduced and that of LH increased by the doses of estrogen used.

The role of the limbic structure of the brain in the hypothalamic control of ACTH and gonadotropic secretion has been demonstrated [3, 6, 8-10]. In this connection it is interesting to study the role of the limbic structures in the mechanism of the reverse action of hormones of the pituitary-adrenal and pituitary-gonadal systems, for this mechanism lies at the basis of the automatic regulation of neuro-hormonal systems [3, 5].

The object of this investigation was to study the excitability of the limbic structures of the brain (the amygdala and hippocampus) under the influence of glucocorticoids and ACTH and the effect of destruction of these brain structures on the action of estrogen on the hypothalamic control of gonadotropin secretion.

## EXPERIMENTAL METHOD

Chronic experiments were carried out on 8 male dogs (15-18 kg) with electrodes (used also for injecting drugs) implanted into the premamillary region of the posterior hypothalamus, the supraoptic retion of the anterior hypothalamus, the dorsal hippocampus, and the ventro-medial part of the amygdala. Potentials were recorded on a four-channel LZ REMA encephalograph. Excitability of the hypothalamus was judged from changes in the threshold of the activation reaction in the frontal cortex to high-frequency stimulation of the hypothalamus (250 pulses/sec, 0.5 msec, duration 5 sec), amygdala, and hippocampus – by changes in the threshold of paroxysmal afterdischarges recorded during electrical stimulation of these structures and in the corresponding intact opposite structure. Details of the method were described earlier [4]. Water-soluble dexamethasone and hydrocortisone (80-100 $\mu$g) were injected into the brain structures under the stimulating electrode or intravenously (5-15 mg). ACTH was injected in doses of 0.5 unit by microinjection and 20 units intravenously. The volumes of the solutions for the microinjections were 3-5 $\mu$l.

The dorsal hippocampus, including areas $CA_1$-$CA_4$, were destroyed by electrical coagulation (2 mA for 10 sec) in male and female rats (180-200 g). The content of follicle-stimulating hormone (FSH) in the pit-

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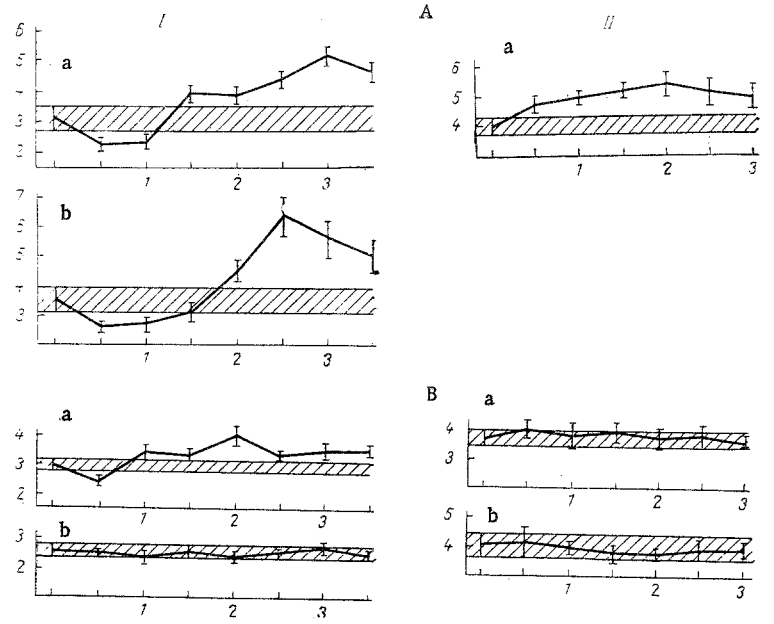


Fig. 1. Changes in excitability of anterior and posterior hypothalamus under the influence of glucocorticoids (A) and ACTH (B): I) premamillary area; II) supraoptic region; a) intravenous injection; b) microinjection. Abscissa, time (in h); ordinate, thresholds of activation reaction (in V).

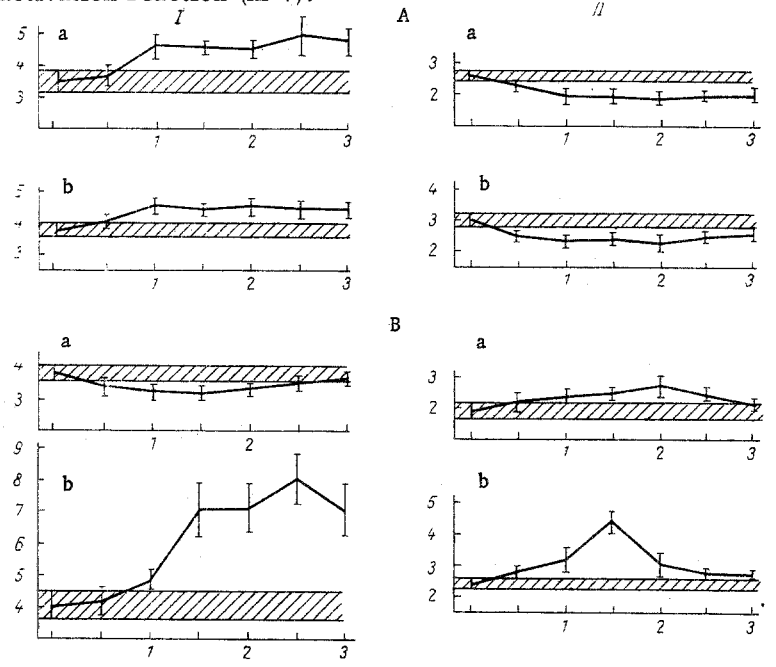


Fig. 2. Changes in excitability of amygdala (I) and hippocampus (II) under the influence of glucocorticoids and ACTH. Ordinate, threshold of paroxysmal afterdischarges (in V). Remainder of legend as in Fig. 1.

uitary was determined by the method of Steelman and Pohly [12] and in the blood plasma after Igarashi and McCann [7]. The content of luteinizing hormone (LH) in the pituitary and plasma was determined by Parlow's method [11]. The results were expressed for LH in micrograms of standard bovine LH (NIHLHB-6) and for FSH in units of the standard preparation (HMG21RP). The animals were used in the experiments 4 weeks after destruction of the brain structures. Estradiol monobenzoate (10 $\mu$g) was injected during the 7 last days from the beginning of brain destruction. The experimental results were subjected to statistical analysis [1].

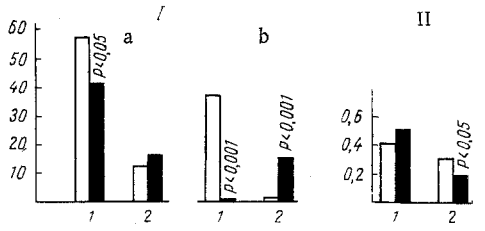


Fig. 3. Content of FSH (I) in pituitary (a) and plasma (b) and of LH (II) in plasma after destruction of hippocampus and injection of estradiol monobenzoate: 1) intact animals; 2) animals with destroyed hippocampus. Unshaded columns – control; shaded columns – after injection of estradiol monobenzoate. Ordinate – level of FSH and LH (in μg/mg pituitary or /ml plasma).

## EXPERIMENTAL RESULTS AND DISCUSSION

The results of the experiments to study the effect of glucocorticoids and ACTH on excitability of the hypothalamus, amygdala, and hippocampus are shown in Figs. 1 and 2. It is clear from Fig. 1 that intravenous injections of dexamethasone, hydrocortisone, and ACTH led to an initial decrease in threshold of the activation reaction in the frontal cortex to high-frequency stimulation of the posterior hypothalamus. During the next few hours, however, this threshold began to increase and was highest 2.5-3 h after the beginning of injection of the drug. The same effect was observed after microinjections of dexamethasone and hydrocortisone into this region of the hypothalamus. Intravenous injection of hydrocortisone led to an increase (without the preliminary decrease) in the threshold of the activation reaction to high-frequency stimulation of the anterior hypothalamus. Microinjections of ACTH neither into the premamillary nor into the supraoptic region of the hypothalamus gave rise to any significant changes in their excitability.

It will be clear from Fig. 2 that intravenous injection or microinjection of hydrocortisone was followed by an increase in the thresholds of the paroxysmal afterdischarges in the amygdala. Hydrocortisone had the opposite action on hippocampal excitability. Injections of ACTH (either intravenously or directly into the structure) led to a marked increase (twofold) in the thresholds of the paroxysmal afterdischarges in the amygdala (Fig. 2), indicating marked depression of the excitability of that structure. Some increase in the thresholds of the paroxysmal discharges was recorded in the hippocampus under the influence of ACTH. In the experiments in which microinjections of bidistilled water (5 μl) were given into the brain structures (control) no appreciable changes in their excitability were observed.

The results of these experiments thus show that hydrocortisone and ACTH considerably modify the excitability of the structures of the limbic system studied – the amygdala and hippocampus. As other workers have shown, electrical stimulation of the amygdala increases the secretion of ACTH, but after excitation of the hippocampus, on the other hand, a decrease in the blood glucocorticoid level and in the response of the pituitary-adrenal system to stimulation is observed [3, 10]. These results, together with those of the present experiments, show that in the reverse (inhibitory) action of glucocorticoids on ACTH, besides their known effect on the hypothalamus, an important role is also played by the changes mentioned above in the state of the limbic structures: lowered excitability of the amygdala and increased excitability of the hippocampus. Depression of the excitability of the amygdala presumably plays a significant role in the negative effect of ACTH (by a short feedback mechanism).

In chronic experiments on rats 4 weeks after electrolytic injury to the dorsal hippocampus, marked inhibition of FSH (experiments on males, Fig. 3) and LH (on females) secretion was observed. As Fig. 3 shows, the decrease in LH secretion after destruction of the dorsal hippocampus in the experiments on the males was very slight. Under the influence of destruction of the dorsal hippocampus the reactivity of the central structures to the reverse action of estrogen was altered. Injections of estradiol monobenzoate (10 μg, 7 days) into animals with a destroyed hippocampus stimulated the liberation of FSH into the blood, unlike in intact animals in which the doses of estradiol monobenzoate used had a negative action. Meanwhile the LH level was slightly increased by the above-mentioned doses of estrogens in the control, but its level in the blood of the animals undergoing operative destruction of the hippocampus was reduced by a statistically significant degree (Fig. 3).

As was pointed out earlier [2], an increase in the secretion of gonadotropins (FSH and LH) by the pituitary was observed in rats after electrical coagulation of the basal nuclei of the amygdala. Meanwhile, the inhibitory action of estradiol monobenzoate on FSH secretion by the pituitary was appreciably weakened after electrical coagulation of the basal amygdala. This effect was accompanied by a decrease in the FSH level in the plasma (very slight compared with the control) and an increase in its level in the pituitary. These results point to a role of the hippocampus and amygdala in the mechanism of the central action of estrogens on FSH and LH secretion by the pituitary.

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