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HYPOTHALAMIC INVOLVEMENT IN THE PITUITARY ADRENO-CORTICAL RESPONSE

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There is now general agreement that the central nervous system, or, more particularly, the hypothalamus, is capable of stimulating the anterior pituitary gland to secrete adrenocorticotrophic hormone (ACTH) whenever the individual is exposed to a stressing agent. This ACTH, by its effect on the adrenal cortical secretion, brings about the adaptation of the body (general adaptation syndrome). On the other hand, there is no agreement concerning the location of the specific region, if any, in the hypothalamus or diencephalon which brings about the secretion of ACTH. Also it is not known whether this response is brought about by direct or indirect activation.

Anand, Raghunath, Dua & Mohindra (1954) localized the hypothalamic centres in the rat which control the nervous phase of ACTH secretion in response to stress. This was done by determining the eosinopenic response to stress stimuli before and after producing small electrolytic lesions in the hypothalamus. These areas are located in the medial part of the anterior hypothalamus and the area just caudal to it, i.e. antero-medial part of the median eminence. de Groot & Harris (1950) and Porter (1953), however, by experiments on rabbits and cats respectively, localized the effective areas in the posterior part of the tuber cinereum and mammillary body. Studies of Hume and Wittenstein, reported by Hume (1952), in dogs, on the other hand, located this area in the anterior portion of the median eminence, but did not localize it more precisely.

The present investigation was undertaken to determine the localization of this area in the cat by stimulation of the hypothalamus with implanted electrodes. The results have tended to confirm the previous observations, made as a result of ablation studies in the rat (Anand *et al.* 1954).

METHODS

Multilead electrodes were implanted in the different hypothalamic regions of fourteen cats, each animal having two sets of electrodes, one on each side. Under pentobarbitone anaesthesia and with aseptic precautions, the head of the animal was fixed in the Horsley-Clarke stereotaxic instrument, and selected points on the skull trephined. Each needle electrode used consisted of four stainless-steel insulated wires, fixed together by Plexiglas. The uninsulated tips of these were spaced 2 mm apart. These needle electrodes were directed to the desired depth in the hypothalamus through the trephine holes, and fixed in position by dental cement and sutures. Covered by polyethylene tubing and passing beneath the skin, the electrodes were brought to the surface through a small opening made at the posterior part of the neck.* The co-ordinates of the stereotaxic instrument for the hypothalamic regions had previously been worked out.

Electrical excitation was begun about a week after the operation, and subsequently at intervals of about a week, in unanaesthetized animals. Stimulation was produced by a square-wave stimulator and in general the parameters used were: 5/sec; 0·2 msec pulse duration; 2 V. Stimulation was continued for 1 hr in each instance.

Eosinophil counts using the method of Randolph were made on freely flowing blood samples from the ear vein; (1) immediately before stimulation, (ii) immediately after 1 hr of stimulation, and (iii) 4 hr after the start of stimulation.

Finally the animals were sacrificed and the exact position of electrodes confirmed histologically.

RESULTS

The direct counting of eosinophils in the peripheral blood has been used as an index of the rate of secretion of adrenal cortical hormone. It has been shown by McDermott, Fry, Brobeck & Long (1950) that the eosinopenic response is at least as satisfactory an index of adrenal cortical secretion as the fall in ascorbic acid content of the adrenal. Although the specificity of eosinopenia as a test for increased adreno-cortical secretion has been questioned (Harris, 1952), there is no more useful pilot method for acute studies in the cat (Porter, 1953). Because of variations in the initial levels of eosinophils, the results are expressed in percentage of change, as it has been shown by McDermott *et al.* (1950) that it is statistically permissible to employ percentage as an accurate index of change.

On the basis of histological studies, the hypothalamic regions stimulated were divided into ten discrete areas (Fig. 1). The average changes in the circulating eosinophil count produced in response to stimulation of each area are shown in Fig. 2. These results clearly show that eosinopenia of an average of 25 % or more is obtained only from stimulation of the medial parts of the anterior and middle hypothalamic regions (C and E, Fig. 1), i.e. the antero-medial part of the median eminence of the tuber cinereum. Stimulation of the other hypothalamic regions did not produce an average eosinopenia of more than 10 %, except the lateral area in the mammillary region (J, Fig. 1) which gave an average of 12 %. Stimulation of the lateral area in the posterior hypothalamic region (posterior part of the median eminence of the tuber cinereum) (H, Fig. 1), on the other hand, produced an increase of eosinophils, averaging about 10–14 %.

* The details of the technique were very kindly taught to one of the authors (B.K.A.) by Dr José M R. Delgado of Yale University School of Medicine, U.S.A.

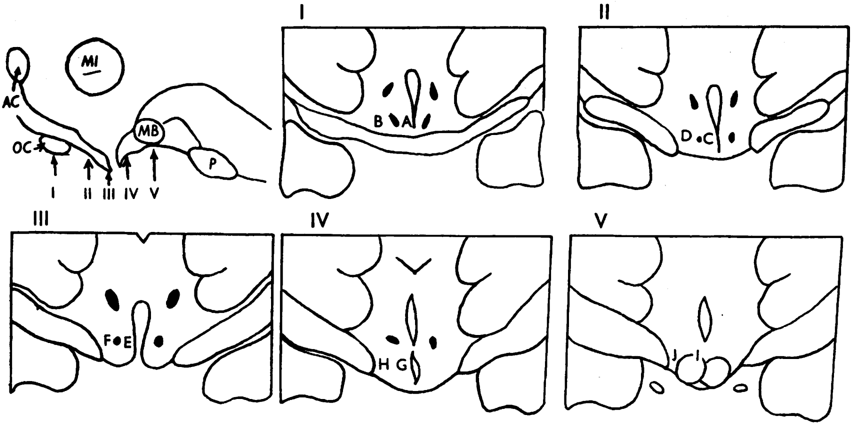

Fig. 1. Mid-sagittal reconstruction of diencephalic region of cat, with transverse sections (I to V) at levels indicated. A to J indicate the ten hypothalamic areas stimulated. *AC*, anterior commissure; *MI*, massa intermedia of thalamus; *OC*, optic chiasma; *MB*, mammillary body; *P*, pons.

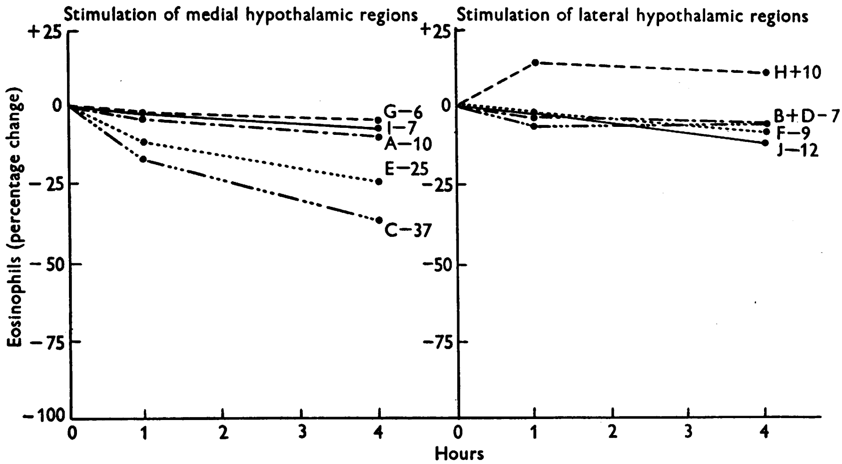

Fig. 2. Averages of changes in the circulating eosinophils, expressed as percentage of the initial levels, obtained by stimulation of the hypothalamic regions (A to J) indicated in Fig. 1.

DISCUSSION

The results of these experiments clearly show that the hypothalamus is intimately concerned with the secretion of ACTH from the anterior pituitary gland, and that the hypothalamic region involved in this is the medial part of the anterior and middle hypothalamic regions (antero-medial regions of the median eminence of tuber cinereum). These data are in agreement with our previously reported findings (Anand *et al.* 1954) that the stress-induced eosinopenia disappears after lesions in this region.

Hume (1952) also reported eosinopenia by remote control stimulation of the anterior hypothalamus in unanaesthetized dogs. These areas were not precisely localized. de Groot & Harris (1950), on the other hand, using remote control stimulation of the hypothalamus of rabbits, obtained lymphopenia from the posterior tuber cinereum and mammillary body. Porter (1953) also obtained eosinopenia by exploratory stimulation of tuberal and mammillary areas in anaesthetized cats, which was greatest when the mammillary bodies were stimulated. Stimulation of the mammillary and posterior tuberal regions in the present study did not produce any marked eosinopenia (Fig. 2). Stimulating the lateral area in the mammillary region did produce eosinopenia which was slightly more pronounced than from some other hypothalamic regions, but much less than that resulting from stimulation of the antero-medial regions. Even in the anterior and middle hypothalamus it is only the medial parts which appear to take part in the control of ACTH secretion and not the lateral parts.

Stimulation of lateral parts of the posterior hypothalamic (posterior tuberal) regions produced an increase in the number of circulating eosinophils, instead of eosinopenia. This may be due to a sympathetic effect causing contractions of the spleen (McDermott *et al.* 1950).

SUMMARY

1. Various hypothalamic regions were stimulated in unanaesthetized cats by means of permanently implanted multilead electrodes, and the number of circulating eosinophils determined.

2. After stimulating the medial part of the anterior and middle hypothalamic regions (antero-medial region of the median eminence of tuber cinereum), the average reduction in eosinophil count was 25% or more of the original level. It is therefore assumed that these hypothalamic regions are concerned with ACTH secretion.

3. Very slight eosinopenia resulted from stimulation of the other hypothalamic regions, including the mammillary regions.

4. Stimulation of the lateral part of the posterior tuberal region produced an increase in the number of circulating eosinophils, probably due to sympathetic effect.

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