CHANGES IN THE BLOOD SEROTONIN CONCENTRATION IN RABBITS DURING PROLONGED ELECTRICAL STIMULATION OF THE ANTERIOR PART OF THE HYPOTHALAMUS

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UDC 612.129:[612.018-547.757]-06:
612.826.4.014.421

Studies of the quantitative distribution of serotonin in the brain of animals and man have shown that the hypothalamus is one of the places where its concentration is highest [4, 6, 8]. The suggestion has been made that serotonin may perform a mediator function for the parasympathetic centers of the brain [7]. The liberation of serotonin by the higher autonomic center in response to stimulation of the central end of the divided vagus nerve has been demonstrated experimentally [5].

On these grounds the presence of serotoninergic structures has been postulated in the hypothalamic region, and the present investigation was carried out in an attempt to detect them.

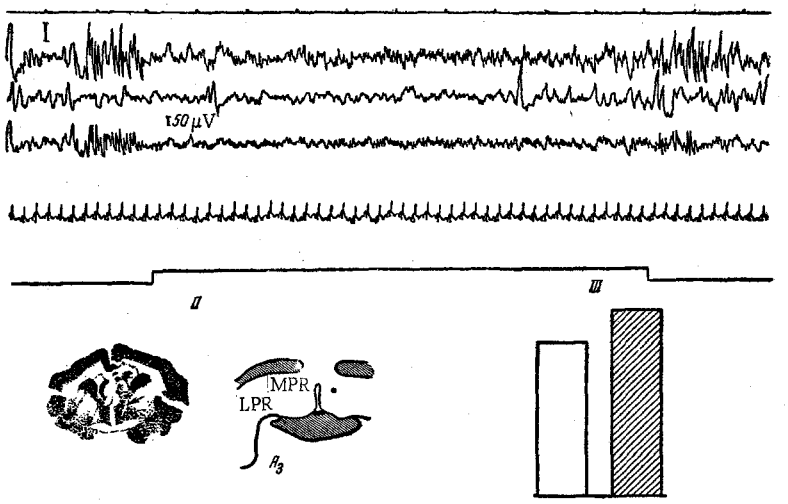
EXPERIMENTAL METHOD

Experiments were carried out on 30 rabbits with electrodes implanted into the brain. The changes in the serotonin concentration in the central and peripheral blood were studied during electrical stimulation of various parts of the anterior hypothalamus and other structures of the diencephalon (optic tract, caudate nucleus, internal capsule, anterior commissure, septum pellucidum). The electrodes were made from Nichrome wire, 0.4 mm thick for the cortical recordings and 0.08 mm thick for the subcortical structures. The cortical electrodes were introduced separately, about 0.8 cm apart. The subcortical electrodes were inserted in pairs, and the vertical distance between them was 0.5 mm. The cortical electrodes were implanted in the cranial bone epidurally above the optic and motor areas of the cerebral cortex. The electrodes were inserted into the hypothalamus by means of Dell's modification of the Horsley-Clark apparatus, in accordance with the coordinates of Sawyer's atlas [11]. The localization of the electrodes in the subcortical structures was verified histologically at the end of the experiments. The electrodes were implanted 7-20 days before the experiments under urethane anesthesia.

The experiments were conducted on unanesthetized animals, in a fasting state, at the same times of day. Bipolar stimulation with rhythmic pulses of electric current (65 cps, 1-2 msec) was applied to the various structures of the diencephalon. The total duration of stimulation was 30 min, in the course of which series of stimuli with a duration of 10-30 sec were applied at intervals of 1-2 min. An electric current of threshold voltage was selected for stimulation, the minimal value causing changes in the electroencephalogram (EEG) and the rhythm of the heart, recorded on a "Galileo" multichannel polyphysiograph. The magnitude of the threshold voltage varied in individual experiments from 1.5 to 5 V. Blood samples for estimation of serotonin were taken before and after stimulation of the hypothalamus and other parts of the diencephalon. Serotonin was estmated in the blood flowing from the brain and in the peripheral blood. The peripheral blood was taken from the marginal vein of the ear. The blood flowing from the brain was taken from the external jugular vein by means of a polythene cannula introduced under local anesthesia into the anterior facial vein is such a way that its end lay at the junction of the anterior and posterior facial veins, the latter receiving blood draining from the brain. Stimulation of the hypothalamus began 1.0-1.5 h after this procedure. Serotonin was determined in acetone extracts of the blood by a biological test [9] on the isolated albino rat's colon, the contractions of which were recorded on a kymograph.

Serotonin creatinine sulfate (GEE Lawson Chemicals) was used as the standard solution. Two standard solutions of serotonin were prepared: I (0.1 μg/ml) and II (1 μg/ml).

Laboratory of Physiological Analysis of Endogenous Neurotropic Substances, Institute of Normal and Pathological Physiology, Academy of Medical Sciences of the USSR, Moscow (Presented by Academician V. V. Parin). Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 63, No. 5, pp. 7-10, May, 1967. Original article submitted March 18, 1966.



Increase in the serotonin concentration in the blood flowing from the brain of rabbit No. 183 following electrical stimulation of the hypothalamus. I) Changes in the EEG and ECG following stimulation of the medial preoptic region of the hypothalamus. From top to bottom: time marker (1 sec); EEG of the right motor, right occipital, and left motor regions of the cortex; EGG, marker of stimulation (an upward deflection of the line corresponds to the period of electrical stimulation of the hypothalamus – 65 cps, 3 msec, 1.5 V); II) localization of electrodes in the preparation and corresponding scheme in accordance with Sawyer's atlas (black spot); transverse section through the brain at a distance of 3 mm anteriorly to the coronal suture; LPR) lateral preoptic region; MPR) medial preoptic region; III) graphic illustration of the serotonin concentration in the central blood before and after stimulation of the hypothalamus. Scale: 1 cm is equivalent to 4 μg serotonin.

EXPERIMENTAL RESULTS

In the rabbits with implanted electrodes, before stimulation of the brain structures mentioned above the mean sertonin concentration in the central blood was 4.63 μg/ml, compared with a mean concentration of 5.42 μg/ml in the peripheral blood. In some experiments this difference was less marked or absent.

Stimulation of the various parts of the anterior hypothalamus, giving an activation reaction on the EEG characteristic of cortical excitation [1-3], was accompanied by changes in the serotonin concentration in both the central and the peripheral blood, differing in character. In 15 of the 21 rabbits with electrodes implanted in the anterior structures of the hypothalamus (the medial preoptic region, the supraoptic nucleus, the anterior hypothalamic region, the ventro- and dorso-medial nuclei) this stimulation caused a mean increase of 48.3% in the serotonin concentration in the blood flowing from the brain. The increase in serotonin was particularly marked following stimulation of the medial preoptic region and the ventro-medial nucleus of the hypothalamus. Only in two animals with electrodes implanted in the ventro- and dorso-medial nuclei of the hypothalamus was electrical stimulation not followed by an appreciable changes in the serotonin cencentration in the blood flowing from the brain. In the remaining 4 rabbits, stimulation of the hypothalamus (anterior part of the ventro-medial nucleus and the medial preoptic region) was accompanied by a decrease in the serotonin concentration in the blood flowing from the brain.

In the peripheral blood the serotonin concentration in most cases fell on the average by 28.7% after stimulation of the anterior structures of the hypothalamus. The exceptions were rabbits with electrodes implanted in the antero-lateral region and the dorso-medial nucleus. Stimulation of these parts of the hypothalamus led to a slight increase in the serotonin concentration in the peripheral blood.

In two cases (electrodes situated in the anterior hypothalamic region and the ventro-medial nucleus) no changes were found in the serotonin concentration in the peripheral blood.

The results of an experiment on rabbit No. 183, in which the medial preoptic region of the hypothalamus was stimulated, are illustrated in the figure.

Stimulation of this part of the hypothalamus, giving rise to an activation reaction on the EEG (see Figure, I), was accompanied by an increase in the serotonin concentration in the blood flowing from the brain (see Figure, III).

Stimulation of certain structures of the anterior hypothalamus (the medial preoptic region, the supraoptic nucleus, the antero-lateral region, the ventro- and dorso-medial nuclei of the hypothalamus) in unanesthetized rabbits by an electric current with the parameters indicated above thus caused an increase in the serotonin concentration in the blood flowing from the brain, whereas in the peripheral blood the changes in serotonin were inconsistent.

Following stimulation of other structures in 9 control rabbits (caudate nucleus, internal capsule, anterior commissure, region of the septum pellucidum) no increase in the serotonin concentration in the central blood was observed in any of the experiments. In two rabbits with electrodes implanted in the optic tract no changes in the serotonin concentration of the blood could be detected after stimulation of this region. In the remaining cases a very slight decrease in the serotonin concentration was observed in the blood flowing from the brain.

Both a decrease and an increase in the serotonin concentration in the peripheral blood was observed in the control animals after stimulation. In two cases no changes were found in the serotonin concentration in the peripheral blood.

In 6 experiments the blood serotonin concentration was investigated in animals receiving reserpine during the previous four days an intramuscular injections of Rausedil in a dose of 0.4 mg/kg ("Gedeon Richter," Hungary). The results of these experiments showed that no serotonin was present in the blood of these animals. Stimulation of the structures of the anterior hypothalamus, usually accompanied by an increase in the serotonin concentration in the blood (antero-lateral region, anterior hypothalamic region, ventro-medial nucleus) in these animals led to the appearance of serotonin in neither the central nor the peripheral blood.

The results described demonstrate that the anterior hypothalamus contains structures, stimulation of which causes an increase in the serotonin concentration in the blood flowing from the brain. The fact that the serotonin concentration in the peripheral blood in these circumstances showed a slight decrease or no change demonstrates that the increase in the concentration of this compound in the blood flowing from the brain was not due to its peripheral formation by the enterochromaffin cells.

The absence of an increase in the serotonin concentration in the central blood under the influence of stimulation of the anterior hypothalamus in rabbits following preliminary administration of reserpine may evidently be attributed to the preliminary removal of the serotonin from the serotoninergic structures of the brain under the influence of reserpine, a phenomenon described by other authors [10, 12].

LITERATURE CITED

1. P. K. Anokhin, The Electroencephalographic Analysis of the Conditioned Reflex [in Russian], Moscow (1958).
2. L. A. Novikova and D. A. Farber, Fiziol. Zh. SSSR, No. 11, 1293 (1959).
3. A. I. Shumilina, Fiziol. Zh. SSSR, No. 10, 1176 (1959).
4. A. H. Amin, T. B. Growford, and J. H. Gaddum, J. Physiol. (Lond.) 126, 596 (1954).
5. G. Benetato, L. Tomusz, et al., Byulleten' nauchnoi Inform. Seriya Med. Nauk, No. 1, 113 (1961).
6. D. E. Bogdansky, H. Wissbach, and J. Udenfriend, J. Neurochem., 1, 272 (1957).
7. B. B. Brodie and P. A. Shore, Ann. N. J. Acad. Sci., 66, 631 (1957).
8. E. Costa and M. H. Aprison, J. Nerv. Ment. Res., 126, 286 (1958).
9. C. E. Dalglish, C. C. Toh, and T. S. Work, J. Physiol. (Lond.), 120, 289 (1953).
10. M. K. Paasonen and M. Vogt, Ibid., 131, 617 (1956).
11. C. H. Sanyer, J. N. Everett, and J. D. Green, J. Comp. Neurol., 101, 801 (1954).
12. P. A. Shore, S. L. Silver, and B. B. Brodie, Sciences, 122, 284 (1955).