A fall in the levels of glutamic and aspartic acids in the ischemic myocardium was observed previously [4, 9]. The loss of these amino acids was equimolar to the increase in the alanine concentration and took place under the influence of coupled aspartate- and alanine-aminotransferases [10]. The result of degradation of glutamic and aspartic acids is the formation of α-ketoglutarate and oxaloacetate and intensification of substrate phosphorylation in the mitochondria [12, 14]. Thus the use of cardioplegic solutions containing these amino acids ought to reduce their losses during ischemia, facilitate fuller binding of the excess of ammonia and, most important, maintain ATP at a higher level during cardiac arrest. These conclusions are based on the protective effect of these two amino acids during cardioplegia and reperfusion of the heart in animals [3, 7] and in man [11] and they are in agreement with the results of the present investigation.

LITERATURE CITED


1. V. F. Portnoi, A. M. Korostylev, and D. M. Lerner, Grudn. Khir., No. 1, 19 (1983).
2. H. J. Bretschneider, G. Hubner, D. Knoll, et al., J. Cardiovasc. Surg., 16, 241 (1975).
3. R. E. Engelman and W. A. Dobles, Ann. Thorac. Surg., 33, 453 (1982).
4. A. Freminet, Comp. Biochem. Physiol., 70B, 427 (1981).
5. C. J. Hilton, W. Teubl, M. Acker, et al., Ann. Thorac. Surg., 28, 323 (1979).
6. W. Lamprecht and I. Trautschold, Methods of Enzymatic Analysis, ed. by H. U. Bergmeyer, Vol. 4, New York (1974), pp. 2101-2111.
7. H. L. Lazar, G. D. Buckberg, A. J. Mangarano, et al., J. Thorac. Cardiovasc. Surg., 80, 350 (1980).
8. K. J. Peuhkurinen, T. Takala, E. M. Nuutinen, et al., Am. J. Physiol., 244, H281 (1983).
9. O. I. Pisarenko, E. S. Solomatina, I. M. Studneva, et al., J. Mol. Cell. Cardiol., 15, 53 (1983).
10. O. I. Pisarenko, E. S. Solomatina, V. E. Ivanov, et al., Basic Res. Cardiol., 80, 126 (1985).
11. E. R. Rosenkranz, F. Okamoto, G. D. Buckberg, et al., J. Thorac. Cardiovasc. Surg., 91, 428 (1986).
12. T. Sanborn, W. Gavis, S. Berkowitz, et al., Am. J. Physiol., 237, H535 (1979).
13. H. Taegtmeyer, Circ. Res., 43, 808 (1978).
14. T. Takala, J. K. Hiltunen, and I. E. Hassinen, Biochem. J., 192, 285 (1980).


# EMOTIONAL–BEHAVIORAL DISORDERS IN RATS ON CREATION OF A GENERATOR OF PATHOLOGICALLY ENHANCED EXCITATION IN THE BASOMEDIAL NUCLEI OF THE AMYGDALOID BODY


G. N. Kryzhanovskii* and V. I. Rodina



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The theoretical basis of this investigation was the theory of determinant and generator mechanisms of neuropathological syndromes [3, 4], according to which a neuropathological syndrome is the clinical expression of activity of a pathological system arriving under the influence of a hyperactive determinant structure; the working basis of such a structure is a generator of pathologically enhanced excitation (GPEE). By creating such generators in certain parts of the CNS, corresponding neuropathological syndromes can be reproduced. Since the

*Academician of the Academy of Medical Sciences of the USSR.


Laboratory of General Pathology of the Nervous System, Research Institute of General Pathology and Pathological Physiology, Academy of Medical Sciences of the USSR, Moscow.


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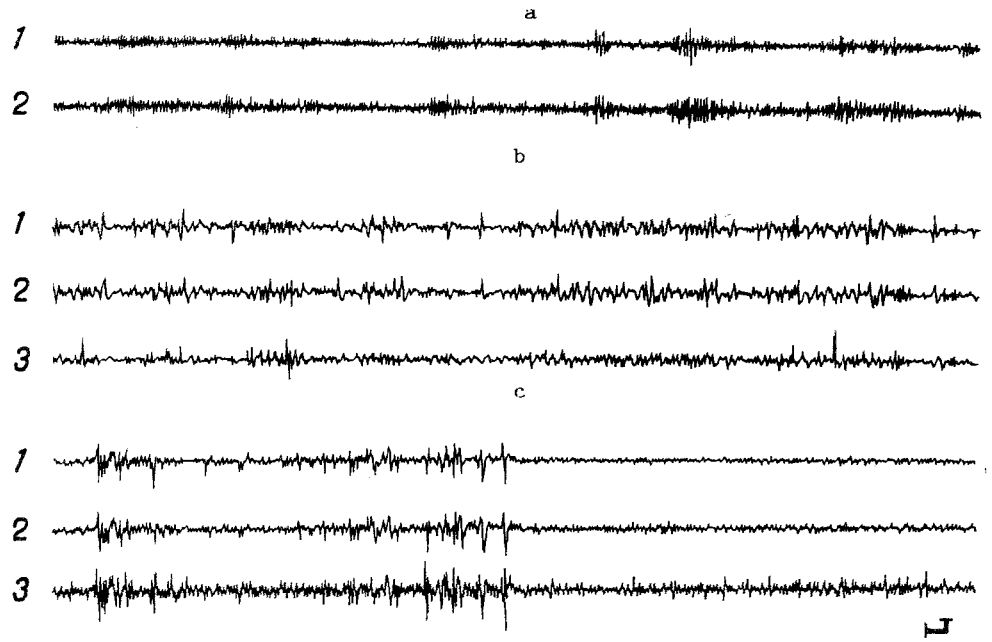


Fig. 1. Electrical activity of basomedial nuclei of right (1) and left (2) amygdaloid body and of dorsal (right) hippocampus (3) of control and experimental rats. a) EA of control animal on 7th day after bilateral injection of 0.3 µl physiological saline into nuclei of amygdaloid body; b) EA of experimental animal on 2nd day after bilateral injection of 10 µg cainic acid into nuclei of amygdaloid body; c) EA in experimental animal on 17th day after injection of cainic acid solution. Calibration: 1 sec, 400 µV.

amygdaloid body plays an essential role in the regulation of an animal's emotional behavior [2, 5], it has been suggested that the creation of a GPEE in that brain region would lead to corresponding disturbances both of behavioral responses and of the emotional state.

## EXPERIMENTAL METHOD

Experiments were carried out on 50 male Wistar rats weighing 250-350 g. Chemical electrodes were inserted under hexobarbital anesthesia (150 mg/kg, intraperitoneally). One part of the chemical electrode was a glass cannula 0.5 mm in diameter, intended for injecting solutions of substances into the brain; the other part consisted of nichrome wire, insulated to the tip, 0.2 mm in diameter, which served as the electrode. A stereotaxic atlas of the rat brain [8] was used. To create the GPEE in both basomedial nuclei of the amygdaloid body (bilaterally) cainic acid and ferrous sulfate — substances with a different mechanism of action but with a similar ultimate effect in the form of hyperactivation of neurons — were used [6, 7, 11]. A 100 mM aqueous solution of ferrous sulfate (12 rats) or a 0.5 mM solution of cainic acid in 5 mM Tris-HCl buffer, pH 7.2 (28 rats), was injected in a volume of 0.3 µl by means of a icrodoser in the course of 2-3 min. Animals of the control group (10 rats) received an injection of physiological saline (0.8% NaCl) or tris-HCl buffer (5 mM, pH 7.2) in the same volume. The reference electrode was inserted into the nasal bones. Electrical activity (EA) was recorded by a monopolar technique on a polygraph (Nihon Kohden, Japan) from an unrestrained animal in a cage, starting from the 2nd day after the operation. The animals' behavioral activity was determined by the open field test. The period of observation was not less than 3 weeks. In the course of the investigation all the animals were kept under standard animal house conditions on a communal diet.

## EXPERIMENTAL RESULTS

Two types of spontaneous EA could be observed on the trace of activity recorded from the basomedial nuclei of the amygdaloid bodies of the control animals throughout the period of observation: low-amplitude (200-250 µV) desynchronized activity, predominating while the animals moved around the cage, and high-amplitude (1-2 mV) spindle-like activity, recorded when the animal was at rest and immobile (Fig. 1a). In experimental animals into which solutions

of cainic acid or ferrous sulfate were injected, the presence of considerable and continuous epileptic activity (EpA) could be observed on the EEG recorded from the basomedial nuclei; it was weaker in the dorsal hippocampus, where single spikes with an amplitude of 1.5-2 mV were recorded (Fig. 1b). After 2-3 days the character of EA of the basomedial nuclei and dorsal hippocampus changed: EpA was exhibited in the form of separate spikes for groups of high amplitude (1.5-2.5 mV), pointed wave bursts. The bursts of EpA were synchronized in all three structures tested, and their appearance was paroxysmal in character: they disappeared for a time after their appearance, and then reappeared. Frequently bursts of EpA in the dorsal hippocampus were stronger than in the basomedial nuclei. No significant differences were found in the character of EpA evoked by injection of cainic acid solution or ferrous ions. EpA was preserved in one form or another and at a certain level of intensity throughout the investigation. Thus injection of both cainic acid and ferrous sulfate into both basomedial nuclei of the amygdaloid body of the animals induced the formation of a GPEE in them.

In the control tests the experimental procedures, namely insertion of electrodes and injection of physiological saline or Tris-HCl buffer into the basomedial nuclei of the amygdaloid body, did not lead to any changes in either the external appearance or the body weight of the animal on days after the operation. The behavioral activity of the control rats did not differ significantly from their preoperative activity.

In the experimental rats which received injections of cainic acid or ferrous sulfate solutions, postural changes were observed: their back was arched, they became humpbacked, and their head was flexed and drawn toward the trunk. These phenomena could be observed virtually immediately after the operation. Sometimes an increase in tone of the tail muscles and extension of the hind limbs were recorded. No movements of the vibrissae or sniffing movements were present. The rhythm of respiration was disturbed and it became discontinuous. The majority of animals showed strong piloerection, many developed ptosis, and some developed swelling of the face. The rats stopped eating and lost much weight: up to 75-80 g in the course of 5-7 days, i.e., about 25% of their initial body weight. After 5-7 days the animals began to eat and put on weight again. The tone of the tail and hind limb muscles gradually decreased. The humpbacked posture and flexion of the head occurred only during long bursts of EpA. Meanwhile trophic disturbances of the hair cover, which appeared on the first days after the operation and took the form of brown discoloration, greasiness, and sometimes falling out of the hair, intensified with the passage of time; constipation was frequent.

During the first postoperative days, when considerable EpA was observed on the EEG (Fig. 1b), the animal's motor activity was sharply reduced. When placed in an "open field" the rat remained immobile, and only when actively stimulated (by repeated taps or by attempts to move it) would it move a few squares and then become fixed again in the humpbacked posture. After 2-3 days, when EpA in the test structures had changed (Fig. 1c) the animals' behavior became different again. Their motor activity became impulsive in character. In the "open field" they ran short distances and then stopped still. Isolated jerks of the head and hind limbs or trunk were observed in some rats. In addition, virtually all the rats developed stereotyped forms of behavior, manifested as frequent repetition of standing on the spot, and peering into the same hole. Some animals exhibited a distinctive habit of standing up in each square of the field through which they passed; stereotyped head shakings, intensified sniffing, and sometimes grunting were observed, while in some cases the rat moved continuously forward in the same direction. The forms of stereotyped disturbances in the same animal could change on different days of the experiment. Meanwhile grooming was depressed and its duration shorter than in the preoperative period in the same animals, and sometimes it was absent altogether. The animals exhibited marked hyperreactivity to various stimuli (touching, blowing, sound or light). A violent response in the form of jumping up or away was exhibited only to the first, unexpected, presentation of the stimulus; it did not recur in response to repeated presentation. This so-called "startle reaction" is interpreted as a manifestation of an emotional alarm response [1]. The animals became irritable, which was reflected in their distinctive psychomotor hyperactivity, in the form of throwing and scattering the sawdust and chewing it. Meanwhile aggressiveness toward the other animals and toward the experimenter was not exhibited. Hyper-reactivity and a state which could be assessed as anxious excitation and irritation, and also stereotypy lasted throughout the period of observation. Some of the animals died at various times, most frequently during the first postoperative days. At autopsy no macroscopic pathological changes could be found.

Three of the experimental animals had fits: one had a fit on the 30th day after an ordinary open field test, while the other two had fits on the 14th and 25th days after the operation, provoked by the cry of rats undergoing painful procedures.

The results of the investigations are thus evidence that the creation of bilateral GPEE in the basomedial nuclei of the amygdaloid body leads to a complex series of emotional–behavioral disturbances; the important factor in this case is not the nature of the substances inducing GPEE formation (cainic acid or ferrous ions), but the activity of the GPEE itself, which plays the role of primary pathological determinant. The hippocampus, where EpA was recorded, also is involved in the process, and in the later stages the EpA in it may actually be stronger than in the amygdaloid body. Hyperactive structures of the hippocampus may perhaps play the role of a secondary pathological determinant. The appearance of extrapyramidal symptoms (rigidity, a humpbacked posture, "freezing" on the spot, compulsive movements, etc.), suggests that the caudate nucleus and globus pallidus are involved. Autonomic disturbances (ptosis, constipation, piloerection, loss of weight, irregular breathing, etc.) are evidence of involvement of the sympathetic and parasympathetic divisions of the autonomic nervous system in the general pathological process. The process evidently also involves the hypothalamo–hypophysio–adrenal system (pigmentation of the hair, swelling of the face, lowering of immune resistance, etc.). A branched pathological system is created, with its components formed by different parts of the brain. Clinically, this system is manifested as a complex polymorphic syndrome for combination of particular syndromes. Meanwhile features of affective disorders are well defined (emotional reactions of anxiety, depression of various motivations, etc.) but in the absence of aggressiveness. The fact that general depression and phenomena of psychomotor inhibition can be replaced by episodes of distinctive psychomotor hyperactivity, when the rat scatters and throws the sawdust around, and chews it, deserves special attention. Hyperactivity, anxious excitation, and increased irritability lasted for a long time. All the facts described above are evidence of involvement of structures of the limbic system and of aminergic mechanism in the pathological process.

It is difficult at present to identify definitely the complex syndrome of emotional–behavioral disorders described above. In many features it closely resembles a depressive syndrome, in which a phase of profound inhibition and general depression is followed, conventionally speaking, by agitation. Depressive states have recently been linked with hyperactivation of certain structures in the limbic system, and they can be terminated by antiepileptic agents [9, 10], evidence of the role of generator mechanisms in the pathogenesis of this affective disorder.

LITERATURE CITED


1. G. Yu. Volynkina and N. F. Suvorov, Neurophysiological Structure of Human Emotional States [in Russian], Leningrad (1981).
2. G. N. Kryzhanovskii, Vestn. Akad. Med. Nauk SSSR, No. 11, 42 (1979).
3. G. N. Kryzhanovskii, Determinant Structures in Pathology of the Nervous System [in Russian], Moscow (1980).
4. R. Yu. Ilyuchenok, M. A. Gilinskii, L. V. Loskutova, et al., The Amygdaloid Body: Connections, Behavior, and Memory [in Russian], Novosibirsk (1981).
5. S. A. Chepurnov and N. E. Chepurnova, The Amygdaloid Body of the Brain [in Russian], Moscow (1981).
6. D. J. King, H. V. Wheal, R. P. Sharma, and G. A. Kerkut, Brain Res., 263, No. 2, 172 (1983).
7. E. W. Lothman and R. C. Collins, Neurology (New York), 30, 386 (1980).
8. G. Paxinos and C. Watson, The Rat Brain in Stereotaxic Coordinates, Sydney (1982).
9. R. M. Post and T. W. Unde, Epilepsia, 24, Suppl. 2, 97 (1983).
10. R. M. Post, D. R. Rubinow, and T. W. Unde, Psychopharmacol. Bull., 20, No. 3, 585 (1984).
11. L. J. Willmore, G. W. Sypert, and J. B. Wunson, Science, 200, 1501 (1978).