EFFECT OF LONG-TERM LITHIUM HYDROXYBUTYRATE ADMINISTRATION ON BRAIN LEVELS OF SEROTONIN AND 5-HYDROXYINDOLEACETIC ACID IN RABBITS

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Long-term administration of lithium hydroxybutyrate causes adaptive changes in the serotoninergic system of the brain, which we have recorded 1 h after intramuscular injections of the compound in a dose of 10 mg/kg into rabbits for 7, 15, and 29 days [9]. Meanwhile, for the specific activity of antidepressants and neuroleptics to be exhibited, not only the duration of their administration, but also the time between the last injection and the beginning of the experiment is of essential importance [3, 5, 7]. A rule of this kind, however, is not known for lithium salts.

We investigated the effect of lithium hydroxybutyrate on levels of serotonin (5-HT) and of its principal metabolite, 5-hydroxyindoleacetic acid (5-HIAA), in the cerebral cortex and deep brain structures of rabbits during long-term administration of the compound (for 7, 15, and 29 days). Values of the parameters were recorded 24 h after the last injection of lithium hydroxybutyrate.

EXPERIMENTAL METHOD

Experiments were carried out on male rabbits weighing 2-2.5 kg during January and February. Lithium hydroxybutyrate, in a dose of 10 mg/kg, as a 0.1% aqueous solution, was injected intramuscularly daily at 9 a.m. The control animals were given injections of the corresponding volume of solvent. The animals were decapitated 24 h after the last injection of the compound and weighed samples of individual brain structures were quickly frozen at −10°C. Concentrations of 5-HT and 5-HIAA were determined fluorometrically [4]. The results were subjected to statistical analysis by Student's test.

EXPERIMENTAL RESULTS

Brain levels of 5-HT and 5-HIAA in animals of the control group did not change significantly in the course of the month. The highest 5-HT concentration was found in the corpus striatum and hypothalamus, the highest 5-HIAA level in the midbrain (region of the corpora quadrigemina) and hypothalamus (Tables 1 and 2), in agreement with previous investigations [9].

The 5-HT level in the hypothalamus was lowered by 35% ($p < 0.05$) 24 h after administration of lithium hydroxybutyrate for 7 days, and the 5-HIAA level in the midbrain was lowered by 27% ($p < 0.05$). Only a tendency for concentrations of 5-HT and 5-HIAA to decrease was observed in the other brain structures. At this time the compound evidently inhibited serotonin metabolism in the midbrain and its synthesis in the hypothalamus.

Administration of lithium hydroxybutyrate for 15 days was followed by a decrease in the 5-HT level in the striatum (by 32%, $p < 0.001$) and hypothalamus (by 17%, $p < 0.01$). The 5-HIAA level was lowered in the thalamus and hypothalamus (by 28 and 44% respectively, $p < 0.05$). No significant changes in concentrations of 5-HT or 5-HIAA were found in the other brain formations. After a 2-week course of lithium hydroxybutyrate, synthesis and breakdown of 5-HT in the hypothalamus and striatum are inhibited, as also is 5-HT metabolism in the thalamus.

After a 4-week course of the compound the 5-HT level fell below the control value in the striatum, hippocampus, and amygdala (by 24, 29, and 32% respectively, $p < 0.05$). The 5-HIAA

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TABLE 1. Effect of Lithium Hydroxybutyrate on 5-HT Level (in μg/g wet weight of tissue) in Rabbit Brain (M ± m)

Experimental conditions	Cortex	Striatum	Dorsal hippocampus	Amygdala	Hypothalamus	Thalamus	Midbrain (region of corpora quadrigemina)
Control	0,35±0,01 (10)	1,14±0,05 (10)	0,51±0,02 (10)	0,53±0,02 (9)	0,94±0,04 (10)	0,46±0,08 (10)	0,47±0,03 (10)
Administration of lithium hydroxybutyrate for:							
7 days	0.33±0,02 (5)	1,06±0,08 (5)	0,43±0,05 (5)	0,43±0,04 (5)	0,63±0,07* (5)	0,50±0,06 (5)	0,39±0,03 (5)
15 days	0.32±0,02 (7)	0,78±0,06* (7)	0,42±0,04 (7)	0,53±0,05 (6)	0,77±0,03* (7)	0,52±0,04 (7)	0,49±0,02 (6)
29 days	0,27±0,04 (7)	0,86±0,07* (7)	0,36±0,02*** (7)	0,35±0,01*** (7)	0,74±0,13 (7)	0,42±0,03 (6)	0,40±0,04 (6)

Legend. Here and in Table 2, number of animals shown between parentheses; *$p < 0.05$.

TABLE 2. Effect of Lithium Hydroxybutyrate on 5-HIAA Level (in μg/g wet weight of tissue) in Rabbit Brain (M ± m)

Experimental conditions	Cortex	Striatum	Dorsal hippocampus	Amygdala	Hypothalamus	Thalamus	Midbrain (region of corpora quadrigemina)
Control	0,18±0,03 (7)	0,38±0,04 (8)	0,26±0,03 (7)	0,28±0,03 (8)	0,65±0,03 (8)	0,49±0,03 (8)	0,83±0,05 (6)
Administration of lithium hydroxybutyrate for:							
7 days	0,14±0,01 (5)	0,33±0,02 (5)	0,20±0,01 (5)	0,22±0,03 (5)	0,52±0,08 (5)	0,39±0,06 (5)	0,61±0,08* (5)
15 days	0,13±0,02 (6)	0,31±0,02 (6)	0,24±0,02 (6)	0,25±0,02 (6)	0,37±0,01* (6)	0,35±0,04* (6)	0,72±0,05 (6)
29 days	0,14±0,02 (6)	0,30±0,10 (6)	0,18±0,09 (6)	0,21±0,09 (6)	0,38±0,06* (6)	0,45±0,07 (6)	0,77±0,13 (6)

concentration in the hypothalamus was lowered (by 42%, $p < 0.05$). A tendency for levels of 5-HT and its metabolite to fall also was observed in the remaining brain structures, evidence of the depressant effect of lithium hydroxybutyrate on serotoninergic processes in the brain.

Thus the adaptive changes in the central serotoninergic system observed 1 h after the last of a course of injections of lithium hydroxybutyrate daily for 7, 15, and 29 days in a dose of 10 mg/kg [9], were not found 4 h after the last injection of the compound given for the corresponding periods. The essential feature of these adaptive changes was that, depending on the duration of the course of lithium hydroxybutyrate, processes of 5-HT synthesis predominated in structures of the midbrain and forebrain during the first week of administration of the compound, but later 5-HT metabolism was stimulated and its turnover quickened, whereas after a 4-week course of lithium hydroxybutyrate 5-HT synthesis was inhibited and its metabolism in the brain activated. This phasic nature of the changes, in our opinion, is brought about by the presence of negative feedback between functional activity of serotoninergic terminals and the nuclei raphe [9].

Lithium hydroxybutyrate reduced the 5-HT turnover 24 h after the last of a long course of injections, initially in the midbrain, where the nuclei raphe are located, and later in adjacent structures (the hypothalamus and thalamus), and toward the end of the 4th week, in structures of the forebrain (amygdala, striatum, and hippocampus). During accumulation of lithium and its distribution among the various brain structures, disturbances of metabolism of central 5-HT were evidently becoming increasingly widespread and intensified. With each successive injection of the compound the compensatory powers of the negative feedback were exhausted, and toward the end of a 1-month course of injections 5-HT turnover was observed to be inhibited in both tests (both 1 and 24 h after the last injection) in most brain structures tested, and this inhibition was particularly marked 1 h after a 29-day course of lithium injections. Consequently, adaptive changes in central 5-HT metabolism against the background of long-term lithium hydroxybutyrate administration do not appear until 1 h after the last injection of the compound and stabilize at the level of inhibition of 5-HT synthesis and breakdown observed 1 month after the beginning of the course.

Adaptive changes in central monoaminergic processes under the influence of chronic administration of antidepressants [3, 7] and neuroleptics [5] have been widely discussed in the

literature. It is claimed that experimentally, a specific psychotropic action should be studied only after chronic administration of the preparation, after stabilization of the adaptive changes. It follows from these ideas that the specific activity of lithium hydroxybutyrate ought not to be exhibited until 1 month after its administration, but clinical improvement is known to take place as early as after three daily intravenous injections of the compound in patients with affective mental disorders [1, 6]. The antidepressant action of lithium hydroxybutyrate on a model of reserpine depression is exhibited at about the same time [8]. Lithium hydroxybutyrate is clinically most effective on the 7th-14th days, and the effect does not increase with continued administration [1, 6]. In our experiments it was at these times that the maximal imbalance between synthesis and breakdown of the central 5-HT was observed. Possibly not only stabilization of serotoninergic processes, but also adaptive changes in them may play a definite role in the realization of the psychotropic effect of lithium. It can be tentatively suggested that, by triggering an oscillatory process in the serotoninergic system, lithium itself weakens the pathodynamic structure of the circular psychosis [2], which, in the modern view, is based on fluctuations in the functioning of monoaminergic processes and the balance between them.

LITERATURE CITED

1. G. Ya. Avrutskii, A. A. Neduva, S. N. Mosolov, et al., New Data on the Pharmacology and Clinical Use of Lithium Salts [in Russian], Moscow (1984), pp. 22-30.
2. N. P. Bekhtereva, The Healthy and Sick Human Brain [in Russian], Leningrad (1980).
3. A. V. Val'dman, Neuropharmacology of Antidepressants [in Russian], Moscow (1984), pp. 9-50.
4. B. N. Kogan and N. V. Nechaev, Lab. Delo, No. 5, 346 (1979).
5. M. O. Maimets, "Adaptive changes in dopamine, serotonin, GABA, and benzodiazepine receptors induced by neuroleptics," Author's abstract of dissertation for the degree of Candidate of Medical Sciences, Tartu (1985).
6. S. N. Mosolov, Med. Sestra, No. 4, 36 (1987).
7. D. Yu. Rusakov, "Dynamics of behavior and neurochemical reactivity during chronic administration of antidepressants," Author's abstract of dissertation for the degree of Candidate of Biological Sciences, Moscow (1984).
8. A. S. Saratikov and T. A. Zamoshchina, Zh. Nevropatol. Psikhiat., No. 9, 1362 (1984).
9. A. S. Saratikov, L. L. Fisanova, T. A. Zamoshchina, and S. A. Sakharova, Byull. Éksp. Biol. Med., No. 3, 312 (1986).

ISOLATION AND CHARACTERIZATION OF A PROTEIN C ACTIVATOR FROM *Agkistrodon contortrix contortrix* VENOM

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Protein C (PC) is a vitamin K-dependent blood plasma protein which controls the blood coagulation cascade on the negative feedback principle. Thrombin, bound with thrombomodulin on intact areas of endothelium, converts PC into an active serine protease, which degrades factors V and VIII, which localize the blood clotting process [2, 11]. A congenital or acquired lowering of the PC level leads to the development of thrombosis at an early age [1, 3, 11]. There is evidence of a significant fall in the PC level in patients dying within 10 days of

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