Serotoninergic Mechanism Underlying the Dependence of Some Neurotropic Effects of Lithium Hydroxybutyrate on Circadian Phase of Administration

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Lithium hydroxybutyrate administered in the morning exhibits sedative effect, weakens memory consolidation, accelerates extinction of conditioned active avoidance reaction, and inhibits synthesis and catabolism of serotonin in the brain. When administrated in the evening, this drug apart from its sedative effect, improves learning and memory, prolongs memory retention, accelerates synthesis, and inhibits catabolism of serotonin.

Key Words: *lithium hydroxybutyrate; chronopsychopharmacology; serotoninergic system*

Lithium hydroxybutyrate (LHB) is a highly efficient normothymic preparation with pronounced rhythm-modulating activity [4,5]. This is a beneficial combination in light of the modern chronobiological concept of affective disorders [1]. We have previously demonstrated that effect of LHB on circadian rhythms of spontaneous behavioral activity depends on the time (phase) of administration, initial biorhythms, and illumination regimen [4]. Some neurotropic properties of LHB determined by the presence of bioactive anionic component (hydroxybutyric acid) allows the use of this drug not only in affective disorders. However, phasic rhythm-modulating activity of LHB can affect its psychotropic effects. On the other hand, LHB induces adaptive rearrangements in the cerebral serotoninergic system [10]. Dysfunction of this system affects the formation of circadian rhythms [13]. It is known that circulatory psychoses are characterized by disordered structure of circadian rhythms [1] and impaired function of the serotoninergic system [9,11].

These considerations prompted us to study serotoninergic mechanisms of phasic dependence of sedative and nootropic effects of LHB.

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MATERIALS AND METHODS

Experiments were carried out on male random-bred albino rats (160-180 g) and mice (22-26 g) maintained from January to March under standard illumination regimen (12:12 h light:dark cycle, illumination switched on at 7:00) after a 2-month preadaptation to this regimen (light intensity 120-150 lux). The animals were maintained in individual compartments with free access to food (standard chow) and water. LHB (10 mg/kg) was administered as a single intramuscular injected everyday for intramuscularly for 2 weeks in either the beginning, middle, or end of the illumination period. Control animals were injected with water according to the same scheme. The animals were tested 1 h after the 7th and 14th injection (duration of tests did not exceed 1 h). The following parameters were evaluated: excitability of the nervous system by the integral threshold index (ITI) [12], open field behavior (number of horizontal runs, rearings, and head-dippings), and conditioned reactions. Conditioned active avoidance reaction was assessed by the percentage of positive responses to 10 presentations on the 1st day of conditioning and the number of presentations required for stable conditioned response. Extinction was evaluated by the percentage of positive responses to 10 presentations of the con-

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ditioned stimulus 2 weeks after consolidation of the memory trace.

Metabolism of serotonin in the central nervous system was studied by fluorimetric measurment of the content of its metabolite 5-hydroxyindolacetic acid (5-HIAA) per gram wet tissue [7]. To this end, 4 groups of mice were injected with LHB at 1:00, 8:00, 13:00, and 19:00 for one week. The content of 5-HIAA in the brain (without olfactory bulbs, cerebellum, and medulla oblongata below the inferior colliculi) was measured 1 h after the last injection. Control animals received the same volume of water. The animals were sacrificed by decapitation, and the brain was frozen at -10°C.

The data were processed statistically using the Student *t* test.

RESULTS

The integral threshold index was changed only in rats given morning injections of LHB for 1 week. This rise of ITI attesting to a reduction of excitability of the central nervous system was transient and by the end of the 2nd week ITI did not differ from the initial value (Table 1).

Exploratory head-dipping behavior was suppressed after morning injections of LHB, while horizontal and vertical motor activities were suppressed after 7- and 14-day evening administration, respectively. It should be noted that to the end of the experiment head-dipping and horizontal motor activities returned to normal (Table 1).

Thus, the suppressive effect of LHB on spontaneous behavior, i.e., its sedative effect was phase-dependent, phase sensitivity of different behavioral parameters being unequal. However, a period of maximum sensitivity of the central nervous system preceding the rest and active phases can be determined.

Conditioning (to the first 10 combinations) in rats was considerably promoted only by midday injections of LHB, while memory consolidation was impaired after morning injections and, on the contrary, improved after evening injections (Table 1). This probably explains more rapid extinction of conditioned response in rats injected in the beginning of light phase in comparison with those injected in the middle and end of the light phase. Consequently, similarly to sedative effect of LHB, its nootropic effects was phase-dependent. This dependence was observed after 7-day treatment.

Experiments on mice revealed a clear-cut diurnal dynamics of the content of serotonin and 5-HIAA in the brain: it peaked in the mornings and decreased

TABLE 1. Effect of LHB on Excitability of the Central Nervous System and Conditioned Reactions in Rats as a Function of Time of Injection (*M±m*)

Time of injection and testing, duration of treatment		ITI, V	Head-dipping, abs. units	Horizontal runs, abs. units	Rearing, abs. units	CAAR on day 1, % of positive responses	Formation of 100% CAAR in the group, number of presentations	Extinction, number of positive responses without reinforcement
9.00-10.00 (*n*=10)								
Water	7 days	5.62±0.21	25.8±2.5	19.1±2.7	7.7±2.1	73.3±16.3	—	—
	14 days	5.47±0.09	13.0±1.9	14.8±3.0	12.2±3.2	—	66.8±3.6	69.2±5.5
LHB	7 days	6.62±0.01*	16.2±1.9*	14.3±3.4	9.1±2.5	75.7±8.7	—	—
	14 days	5.77±0.17	12.8±2.1	10.3±2.7	10.4±2.4	—	87.0±5.3*	40.1±3.9*
13.00-14.00 (*n*=12)								
Water	7 days	5.13±0.28	9.7±2.1	10.3±2.6	11.6±1.8	76.3±5.1	—	—
	14 days	6.33±0.28	6.0±2.8	7.75±2.5	4.3±1.5	—	92.4±7.1	72.3±4.8
LHB	7 days	5.66±0.11	15.6±2.3	15.3±3.4	13.1±1.5	92.5±6.2*	—	—
	14 days	4.49±0.28	7.0±3.4	9.8±3.1	7.2±1.9	—	74.2±3.3*	69.5±5.5
18.00-19.00 (*n*=11)								
Water	7 days	5.86±0.22	18.0±3.1	19.4±2.5	8.9±1.5	91.4±6.3	—	—
	14 days	6.59±0.21	10.1±3.1	4.8±1.5	6.0±1.4	—	70.6±2.8	38.3±3.3
LHB	7 days	5.97±0.41	18.6±1.8	7.8±2.3*	7.9±2.4	93.3±6.2	—	—
	14 days	6.33±0.53	3.5±0.53	3.0±1.1	1.7±1.1*	—	44.0±2.4*	28.3±2.1

Note. CAAR: conditioned active avoidance reaction; *n*: number of animals in the group; *$p<0.05$ compared with corresponding control (water).

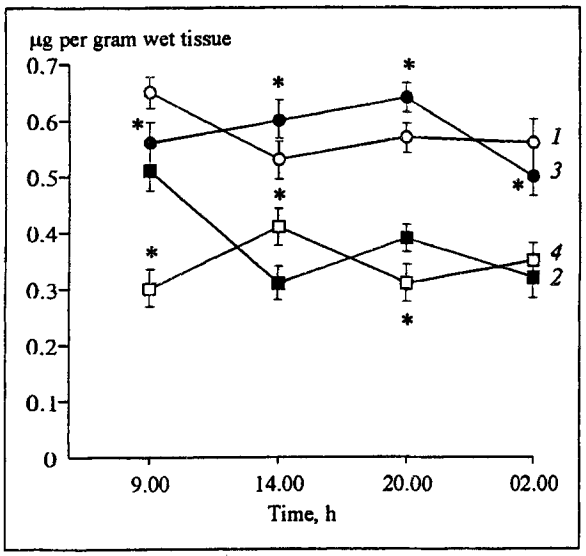

Fig. 1. Effect of lithium hydroxybutyrate on circadian dynamics of the content of serotonin and 5-hydroxyindolacetic acid (5-HIAA) in mouse brain. Serotonin (*1*) and 5-HIAA (2) in control mice; serotonin (*3*) and 5-HIAA (*4*) in mice injected with lithium hydroxybutyrate. *$p<0.05$ compared with the control.

to minimum in the noon (Fig. 1). Morning injections of LHB decreased, while midday injections elevated the content of serotonin and 5-HIAA in the brain; being injected at 20:00 LHB further increased the content of serotonin, but decreased the concentration of 5-HIAA. Hence, the effect of LHB on serotonin metabolism in the brain, a process reflecting functional activity of the central serotoninergic system, depended on the circadian phase of administration: morning injections of the drug stimulated, while midday injections inhibited metabolism of the transmitter; being injected in the evenings, LHB accelerated anabolism of serotonin, while night administration suppressed this process.

Thus, a new rhythm of functional activity imposed by LHB on the serotoninergic system profoundly differ from that in intact animals. This can affect serotonin-regulated behavioral activities. In particular, LHB-induced suppression of the central serotoninergic system can to a certain extent determine the elevation of ITI, since the serotoninergic system restricts activation of the antinociceptive mechanisms in short-term painful stimulation and potentiates it in other influences [2]. The absence of the effect of morning injections of LHB on the level of aggression in isolated animals with destroyed nuclei raphe [6] can also be attributed to suppression of the serotoninergic system due to inhibition of serotonin metabolism. Prolongation of wakefulness in the sleep-wake cycle induced by morning [5] administration of the drug (despite its sedative effect) can also be related to inhibition of the central serotonin metabolism and suppression of the serotoninergic system, which is intimately involved into mechanisms triggering the slow-wave sleep [3].

In contrast to the above-described effects, LHB injected in the evening stimulates serotonin synthesis against the background of its reduced catabolism and exhibites pronounced antiaggressive effect [6]. Enhanced anabolism of serotonin after evening injections of LHB probably underlies its peculiar effect on the sleep structure [5]: prolonged slow-wave sleep, increased latency of the first phase of paradoxical sleep, and shortened duration of the paradoxical sleep [3].

The enhanced metabolism of serotonin after midday administration of the drug (both synthesis and degradation of the transmitter) implies activation of the serotoninergic system. This presumably facilitates learning and consolidation of memory trace, which agrees with published data on stimulation of learning in models with negative reinforcement and long-term memory [8]. Opposite changes in mnestic functions were observed after morning injections of LHB inhibiting synthesis and degradation of the transmitter and suppressing serotoninergic processes. The predominance synthesis over catabolism of serotonin in the brain of rodents receiving evening injections of LHB is probably accompanied by improved consolidation of memory trace [8].

REFERENCES

1. E. B. Arushanyan and V. A. Baturin, *Zh. Nevropatol. Psichiatr.*, No. 3, 85-89 (1995).
2. E. O. Bragin, *Neurochemical Regulation of Nociception* [in Russian], Moscow (1991).
3. A. M. Vein and K. Khekht, *Human Sleep. Physiology and Pathology* [in Russian], Moscow (1989).
4. T. A. Zamoshchina, A. V. Matveenko, V. P. Agarkova, *et al.*, *Farmakol. Toksikol.*, **54**, No. 3, 10-14 (1991).
5. T. A. Zamoshchina, A. V. Matveenko, A. S. Saratikov, *et al.*, *Pat. Fiziol.*, No. 4, 22-24 (1992).
6. T. A. Zamoshchina and A. S. Saratikov, *Eksp. Klin. Farmakol.*, No. 3, 16-19 (1996).
7. B. N. Kogan and N. V. Nechaev, *Lab. Delo*, No. 5, 346-348 (1979).
8. R. I. Kruglikov, *Neurokhemical Mechanisms of Learning and Memory* [in Russian], Moscow (1981).
9. Yu. L. Nuller and A. B. Mikhalenko, *Affective Psychoses* [in Russian], Moscow (1988).
10. A. S. Saratikov, L. L. Fisanova, T. A. Zamoshchina, and S. A. Sakharova, *Byull. Eksp. Biol. Med.*, **108**, No. 7, 54-57 (1989).
11. V. N. Sinitskii, *Depressive States* [in Russian], Kiev (1986).
12. S. V. Speranskii, *Farmakol. Toksikol.*, No. 9, 123-124 (1965).
13. K. Honma, K. Watanabe, and T. Hiroshige, *Brain Res.*, **169**, 531-544 (1979).