Effect of $MgSO_4$, L-DOPA, and Haloperidol on Delayed Stereotyped Behavior Caused by Hyperactivation of the Sensorimotor Cortex in Rats

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Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 128, No. 9, pp. 294-298, September, 1999
Original article submitted July, 1998

Repeated rhythmical electrostimulation of the right sensorimotor cortex resulted in persistent psychoaffective disorders and locomotor stereotypy in rats. Intraperitoneal or intrastriatal administration of $MgSO_4$ did not prevent stereotyped behavior. The animals with developed syndrome exhibited sharply enhanced behavioral responses to L-DOPA and weakened cataleptic reactions to haloperidol and showed psychoaffective disorders in response to $MgSO_4$ administration. It is suggested that dopaminergic hyperactivity and NMDA glutamate receptors are involved in the realization of delayed stereotyped behavior.

Key Words: *cortical seizure activity; excitotoxicity; NMDA receptors; delayed behavioral stereotypy*

Rhythmic electrical stimulation (RES) of the right sensorimotor cortex (SMC) of the rat brain, accompanied by intense and long-term seizure after discharges [1], results in the development of sustained psychoaffective disorders and locomotor stereotypy [1,3]. These disturbances are probably due to poststimulation changes in glutamatergic and/or dopaminergic activity of the central nervous system resulting from the excitotoxic damage to striatal interneurons. Seizure activity in SMC [15] and direct RES of glutamatergic pathways [18] can damage subcortical neurons receiving corticofugal glutamatergic inputs. Intracerebral administration of NMDA and non-NMDA glutamate receptor agonists exerts a neurotoxic effect similar to that caused by glutamatergic neuron hyperactivity [6,7]. The excitotoxic damage to striatal neurons with glutamate receptor agonists modifies function of other neurotransmitter systems. With respect to the dopaminergic system, it is manifested in enhanced behavioral responses to dopamine agonists [10] and weakened cataleptogenic effect of haloperidol [5].

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In the present study we attempted to elucidate the involvement of NMDA receptors in RES-induced delayed stereotyped activities using NMDA receptor antagonist Mg^{2+} [14]. The role of the dopaminergic system in the formation of RES-induced symptoms was assessed by analyzing behavioral and postural reactions to systemic administration of haloperidol and L-DOPA. The contribution of poststimulus changes in NMDA receptor-mediated synaptic transimission manifested itself in animal responses to $MgSO_4$ administration.

MATERIALS AND METHODS

Experiments were carried out on outbred male rats weighing 250-300 g. Experimental procedure, electrode positioning, and parameters of RES were described in detail previously [1,2]. The thresholds of direct and transcallosal responses (DR and TCR, respectively) were determined before and after 20 RES. In experiments with $MgSO_4$ (150 mg/kg in 0.5 ml distilled water), the thresholds were determined before and 30 min after its intraperitoneal injection, then RES was applied.

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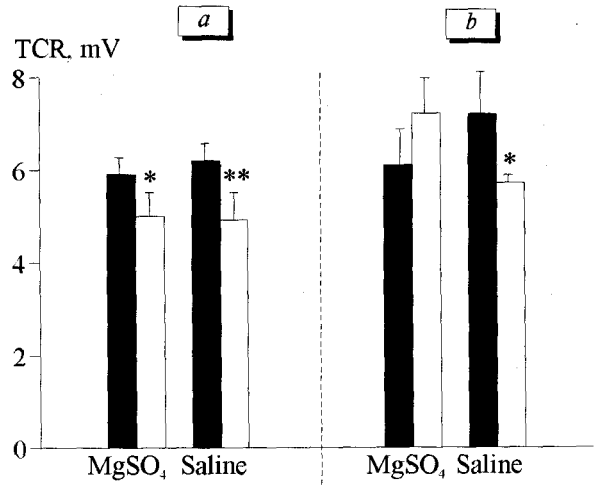

Fig. 1. Effect of $MgSO_4$ on poststimulus changes in transcallosal response (TCR) thresholds before (filled bars) and after (open bars) 20 series of rhythmic electrical stimulation (RES). *a*) intraperitoneal injection of $MgSO_4$ (n=6) or saline (n=7) before RES; *b*) intrastriatal injection of $MgSO_4$ (n=6) or saline (n=7) during RES. *p<0.05, **p<0.01 in comparison with baseline.

The brain was stimulated epidurally by repeated 10-sec trains of low-frequency stimuli (8 pulses/sec) delivered with a 10-min interval. The thresholds were tested 24 h after stimulation, then 5 additional series of RES were applied. In the experiments with intrastriatal injections, $MgSO_4$ (0.5 mg/μl in 0.2 μl) was infused during each RES series (total volume 4 μl) using the following stereotaxic coordinates [17]: A=1.7-1.9, L=2.0, H=5.5. Control animals received saline according to the same schedule. Behavioral observation began on the next day after the second RES series was completed and continued for a month.

Behavioral changes after the intraperitoneal administration of 1 mg/kg haloperidol (Gedeon Richter) and 100 mg/kg L-DOPA (Madopar) were observed for 24 h except night. Locomotor activity was assessed by the number of crossed squares in an Orto-Varimex apparatus (Columbus Instruments). The index of haloperidol-induced catalepsy was determined as the number of animals retaining a given posture (rearing with the forepaws placed on an upper plane of a 10-cm cube) for more than 300 sec. Student's t test was applied to analyze the intergroup difference.

RESULTS

In the first series we studied the effect of $MgSO_4$ administered by different routes on cortical electrogenesis and characteristics of delayed stereotyped activities. Intraperitoneal administration of $MgSO_4$ did not change the thresholds of DR and TCR and the latency and characteristics of afterdischarges (the data not shown). However, intrastriatal injections of Mg^{2+} noticeably modulated poststimulus changes in the cortical excitability (Fig. 1, *b*). Instead of a standard decrease in DR thresholds after 20 RES series [1,2], which was also observed in rats receiving intrastriatal saline, 5 of 6 animals receiving intrastriatal $MgSO_4$ exhibited increased DR thresholds (Fig. 1, *b*). However, both groups showed similar behavioral disturbances with locomotor stereotypies and psychoaffective disorders (Table 1). Behavioral changes in animals receiving RES after intraperitoneal $MgSO_4$ were also similar to those observed in controls.

The response to test drugs in rats with stereotyped behavior and control animals were different. In rats with stereotypy (n=11), intraperitoneal $MgSO_4$ caused short-term tonic body extensions, followed by stereotyped chewing, gnawing, tremor-like sniffing, and enhanced sex grooming, while in control rats (n=10) the same dose of $MgSO_4$ caused only a sedative effect. On the next day experimental rats demonstrated significantly increased locomotor activity in the open field test (Fig. 2, *a*).

L-DOPA inhibited locomotor activity in control rats, but sharply enhanced it in rats with developed

TABLE 1. Development of Stereotyped Locomotor Acts after Rhythmic Electrical Stimulation of the Sensorimotor Cortex after Administration of Saline (Control, n=4) and $MgSO_4$ (n=5) to the Head of the Right Caudate Nucleus

Experimental series			Latency, days	Quantitative characteristics of stereotypy		
				first manifestations	after 1 day	after 5 days
Head jerkings per min	Control		4.6±2.2	13.4±9.8	15.2±10.0	20.8±16.8
	$MgSO_4$		4.0±3.2	12.0±9.6	14.5±11.3	N.d.
Rotations for 5 min	Control	to the right	5.0±2.4	2.7±2.1	1.6±0.6	8.0±10.4
		to the left		12.3±6.4	8.3±2.9	19.3±18.9
	$MgSO_4$	to the right	2.7±0.8	3.0±4.8	4.5±7.0	N.d.
		to the left		13.7±11.0	13.5±9.5	N.d.

Note. N.d. — not determined. Latency of stereotypy was measured from the end of RES.

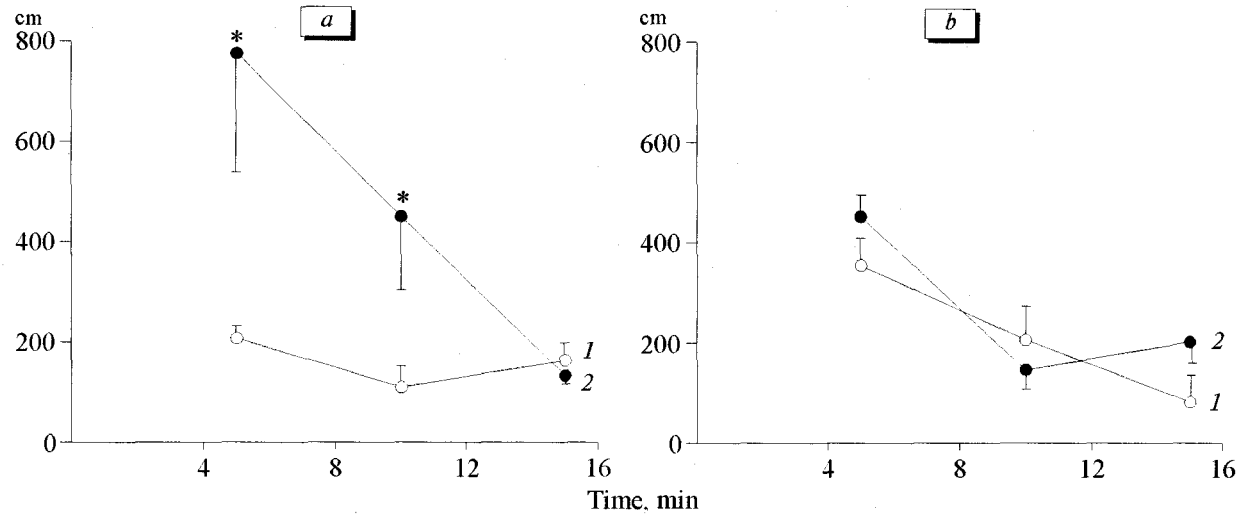

Fig. 2. Effect of $MgSO_4$ (150 mg/kg, intraperitoneally) on locomotor activity in open field. *a*) rats with stimulation-induced stereotypy; *b*) control rats; *1, 2* observation days 1 and 2. Ordinate: total length of ambulations for 5 min. *$p<0.02$ in comparison with day 1.

syndrome (Fig. 3, *a, 1, 2, 5*). If the right-hemispheric RES caused only wry-neck or no symptoms at all, the animals responded to L-DOPA like the controls (Fig. 3, *a, 3, 4*). In the control and RES-subjected rats with developed stereotypy, intraperitoneal injection of haloperidol suppressed both voluntary and involuntary motions (Fig. 3, *c*) and caused catalepsy. However, in the experimental group, the number of animals with the maximum duration of catalepsy was lower than in the control group (Fig. 3, *b, 2*) and the residual suppression of locomotor activity tested after 24 h was less pronounced (Fig. 3, *c*).

The development of delayed stereotyped behavior can be caused by RES-induced excitotoxic damage to striatal and SMC neurons. The long latency and long-term persistence of these locomotor disorders [3] are similar to the characteristics of delayed behavioral reactions observed after intrastriatal administration of glutamate receptor agonists [7]. The hyperactivity of corticofugal glutamatergic pathways can damage the neurons receiving these projections [15,18], while the intrastriatal injection of glutamate receptor agonists (in particular, NMDA) can cause death of GABAergic projection neurons in the striatum and the development of various hyperkineses [7,10].

The excitotoxic damage to cerebral neurons mediated by NMDA receptors can be prevented by Mg^{2+} [13], which prompted us to study its electrophysiological and behavioral effects. When administered intrastriatally, this noncompetitive NMDA receptor antagonist [14] exert no significant effects on cortical epileptogenesis, but prevented the RES-induced increase in

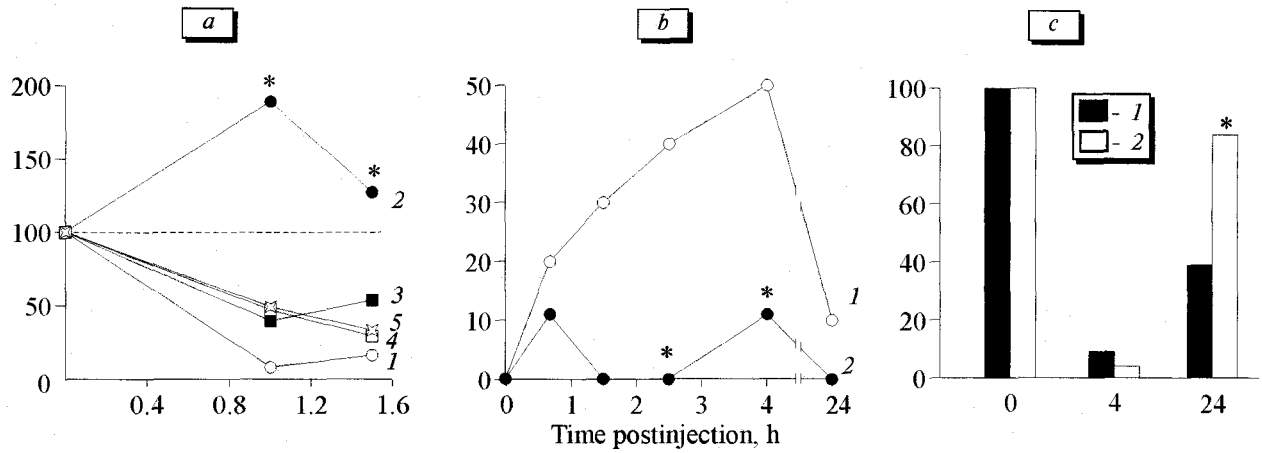

Fig. 3. Effect of L-DOPA (*a*) and haloperidol (*b, c*) on behavioral indices in rats with stereotypy. Points represent total activity for 5 min. Ordinates: *a, c*) locomotor activity, %; *b*) number of animals retaining given posture (rearing with forepaws on the upper plane of a 10-cm cube) for more than 300 sec, %; *a: 1*) control group (n=6); *2*) rats with stereotypy (n=6); *3*), rats without behavioral disturbances after rhythmical stimulation (n=7); *4*) rats with wry-neck syndrome (n=7); *5*), control rats (n=10) receiving saline. *$p<0.05$ in comparison with *1. b, c: 1*) control group (n=10), 2) rats with stereotypy (n=9), *$p<0.05$ in comparison with the control.

excitability observed in our previous studies [1-3] and in the control group in these experiments. Since striatum is the central element of the strio-pallido-thalamic positive feedback loop [16], this effect of Mg^{2+} implies the important role of the striatum in RES-induced elevation of SMC excitability.

The failure of prevention of RES-induced stereotyped behavior with Mg^{2+} (Table 1) suggests that NMDA glutamate receptors do not participate in this process. These data are surprising, since cortico-striatal excitatory influences are predominantly mediated by quisqualate glutamate receptors [8]. On the other hand, Mg^{2+}-induced increase in locomotor activity (Fig. 1, *a*) and orofacial diskinesia in RES-subjected rats imply that NMDA receptors can participate in the realization of stereotyped behavior. It should be noted, that behavioral responses to Mg^{2+} revealed in animals with the RES-induced syndrome (Fig. 2) are similar to the effects induced by organic NMDA antagonists [9,11].

Functional dopaminergic hyperactivity in stimulated rats (Fig. 2) resembles enhanced responses to dopamine agonists in patients with Huntington's chorea and in monkey with excitotoxic striatal damage [7,10]. The weakened effects of haloperidol on cataleptic reaction and locomotor activity observed in stimulated rats (Fig. 3, *b, c*) is of special interest, since haloperidol-induced akinesia and muscular tone disturbances model the corresponding extrapyramidal symptoms in parkinsonism. It should be noted that haloperidol induced no catalepsy after excitotoxic striatal damage [5] or after pharmacological blockade of glutamatergic transmission in the anterodorsal striatum [12] receiving SMC projections [16]. The ability of RES to prevent haloperidol-induced catalepsy agrees with clinical data on the correction of parkinsonian symptoms in the course of electroconvulsive therapy [6,9]. It is suggested, that the antiparkinsonian effect of electroconvulsive treatment is due to selective excitotoxic damage to the certain striatal and SMC neurons [6].

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