PHYSIOLOGY

N-Methyl-D-Aspartate Receptors and Amnesia in Mice with Depression-Like State

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We studied the effect of activation (N-methyl-D-aspartic acid and D-cycloserine) and blockade (dizocilpine and 7-chlorokynurenic acid) of N-methyl-D-aspartate receptors on the development of amnesia in intact and depressive mice under conditions of conditioned passive avoidance response. Agonists and antagonists of N-methyl-D-aspartate receptors produce a strong antiamnesic effect in mice with behavioral despair. In intact animals, only N-methyl-D-aspartic acid and D-cycloserine improved passive avoidance performance.

Key words: *memory; amnesia; depression; N-methyl-D-aspartate receptors*

There is a large body of evidence that NMDA receptors play a role in the pathogenesis of depression. This hypothesis is taken into account in the development of new potent antidepressants [1,9]. Depressive-like state is induced in experimental animals for the analysis of behavior and memory [4,5,7]. High anxiety, passive behavioral pattern, anhedonia, and delayed extinction of fear memory are typical of depression-like state [2,7,10]. However, the role of NMDA receptors in learning and memory of animals with a depressive-like state is poorly understood. At the same time, published data show that the effectiveness of neuropharmacological treatment depends on initial behavioral characteristics [3,6]. It remains unclear whether the drugs modulating activity of NMDA receptors affect the resistance to amnesic factors in depression. There are conflicting data on the antiamnesic effect of NMDA receptor agonists and antagonists [11-13]. We believe that this discrepancy results not only from specific features of amnesic factors and memory tests, but also from the difference in the initial functional state of animals, which was not taken into account in previous researches.

Here we compared the effects of activation and blockade of NMDA receptors on the development of amnesia in intact mice and animals with a depression-like state.

MATERIALS AND METHODS

Experiments were performed on 120 male C57Bl/6J mice weighing 20-25 g. The animals had free access to water and standard food. Our study was performed in accordance to the principles of humanity and Rules for Studies with Experimental Animals (Supplement to the Order of the Ministry of Health of the USSR, No. 755, 12.08.1977) and approved by the Committee of Biomedical Ethics (Institute of Physiology).

Behavioral despair was induced by forced swimming in a reservoir with cold water (3 days, 5 min a day, water temperature 22°C). Memory changes were studied during 1 session of passive avoidance (PA) conditioning in a chamber with dark and light compartments. The mouse was placed in a light

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compartment. Electrical stimulation (0.5 mA, 2 sec) was delivered through an electrode floor when the mouse moved to the dark compartment. Amnesia was induced by restraining of the mouse in a dark compartment for 5 min immediately after electrocutaneous stimulation on the day of training. PA performance was tested over 180 sec after 24 and 48 h. The latency of transition into the dark compartment was recorded.

We studied the effect of activation and blockade of NMDA receptors on the development of amnesia in intact mice and animals with behavioral despair. The mice were divided into 10 groups of 10-15 specimens each. Groups 1-5 included intact mice receiving physiological saline (control), NMDA receptor antagonists dizocilpine (MK-801, ion channel blocker, 0.15 mg/kg) and 7-chlorokynurenic acid (7KYN, modulator of the receptor glycine site, 10 mg/kg), and NMDA receptor agonists N-methyl-D-aspartic acid (NMDA, glutamate-binding site, 25 mg/kg) and D-cycloserine (glycine site partial agonist, 20 mg/kg), respectively. Group 6-10 mice with behavioral despair received these compounds in the same doses. The test compounds were dissolved in physiological saline 30 min before PA training and amnesic treatment.

The results were analyzed by two-way analysis of variance ANOVA (factor 1, group; factor 2, time of testing) and subsequent comparative tests.

RESULTS

Restraining of control and depressive mice in a dangerous compartment of the chamber on the day of training was accompanied by blockade of PA performance after 24 and 48 h (Fig. 1, groups 1 and 6). No differences were found in the latency of transition on days of training and testing ($p>0.05$), which reflected the development of total amnesia.

NMDA receptor blockade with dizocilpine (group 2) and 7KYN (group 3) had no effect on amnesia in intact mice. The latency of transition in these animals practically did not differ from the control. Dizocilpine (group 7) and 7KYN (group 8) produced an antiamnesic effect in mice with behavioral despair. After 24 and 48 h these mice exhibited longer latency of transition compared to control depressive animals (group 6) and intact mice receiving injections of dizocilpine and 7KYN (groups 2 and 3).

NMDA receptor activation in intact and depressive mice (groups 4 and 9) prevented the development of amnesia and increased the latency of transition compared to the control (groups 1 and 6). Global statistical analysis of parameters for groups 1 and 4 and groups 6 and 9 revealed a significant influence of the group factor ($F_{1,23}=6.90$, $p<0.02$; $F_{1,23}=9.18$, $p<0.006$) and time factor ($F_{2,46}=7.35$, $p<0.002$; $F_{2,46}=11.52$, $p<0.0001$) and interaction between these factors ($F_{2,46}=5.29$, $p<0.009$; $F_{2,46}=5.17$, $p<0.01$). It should be emphasized that the positive effect on PA performance was most significant in mice with behavioral despair. The antiamnesic effect was delayed in intact mice. These animals significantly differed from the control only after 48 h. However, depressive mice differed from controls after 24 h.

NMDA receptor agonist D-cycloserine abolished the effect of amnesic treatment and improved PA performance in intact and depressive mice (groups 5 and 10) compared to the control (groups 1 and 6).

Our results indicate that activation of NMDA receptors increases the resistance to amnesic treat-

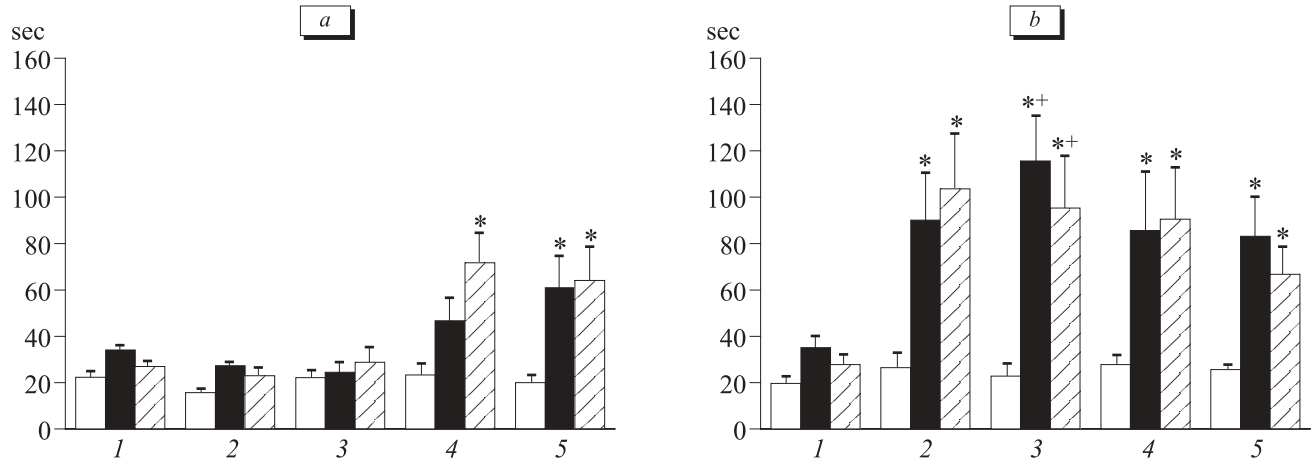

Fig 1. Effect of activation and blockade of NMDA receptors on PA performance (latency of transition) during amnesia. Intact mice (*a*) and mice with behavioral despair (*b*). Control (*1*); dizocilpine, 0.15 mg/kg (*2*); 7KYN, 10 mg/kg (*3*); NMDA, 25 mg/kg (*4*); and D-cycloserine, 20 mg/kg (*5*). Light bars, day of training; dark bars, after 24 h; shaded bars, after 48 h. $p<0.05$: *compared to the control; +compared to dizocilpine-treated animals.

ment (restraining of the animal in a dangerous compartment) independently on the initial behavioral pattern. Previous experiments showed that NMDA receptor agonists produce an antiamnesic effect during amnesia of various types. Partial agonists were most potent in this respect [11-13]. We found that the antiamnesic effect of NMDA in intact mice developed more slowly compared to that of D-cycloserine, which was consistent with published data. The mechanism of this phenomenon remains unknown. The latency of transition remained unchanged after administration of the test compounds on the day of training. It can be hypothesized that improvement of cognitive processes is not associated with locomotor activity or sensitivity to pain punishment.

It is interesting that antagonists with various sites of action on the NMDA receptor complex, dizocilpine and 7KYN, produce a selective antiamnesic effect in mice with behavioral despair. The selective neuroprotective effect of NMDA receptor blockade seems paradoxical taking into account published data on the negative effect of these compounds on learning and memory in intact animals [3,6,13]. However, dizocilpine and 7KYN produce a positive effect in animals with impaired retrieval of memory trace (*e.g.*, during hypoxia, pentylenetetrazole treatment, and brain trauma) [8,12]. Various effects of NMDA receptor antagonists in intact and depressive mice are probably related to individual emotional reactions under negative conditions. The animals with a depression-like state are characterized by dysfunction of the reinforcement systems in the brain and increased sensitivity to stress factors [7,10]. Dizocilpine and 7KYN that produce a strong antidepressant effect and increase activity of the reinforcement system [1,9] probably contribute to the recovery of associative relations in PA training of depressive mice. This effect manifested in the resistance to amnesia.

We conclude that the developed depression-like state determines a role of NMDA receptors in prevention of amnesia.

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