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SHORT COMMUNICATION

Kynurenic Acid Prevented Social Recognition Deficits Induced by MK-801 in Rats

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Summary

MK-801 impaired social recognition potency of adult male rats when given immediately after the initial interaction with a juvenile rat. Administration of kynurenic acid prior to the initial interaction protected the adults against recognition deficits induced by MK-801. When re-exposed at a delay of 30 min to the familiar juvenile, social investigation in the adults was significantly reduced. Thus, the adults are able to remember olfactory stimuli emitted by juvenile conspecifics.

Key words

Amnesia • Kynurenic acid • MK-801 • Social recognition • Working memory

It is generally accepted that the NMDA receptor complex is regulated by several modulatory sites including ligand binding and allosteric sites. One of these modulatory sites is located within the receptor-associated ion channel where a non-competitive receptor antagonist MK-801 binds to block the channel in its open state. MK-801 has been shown to disrupt learning and memory of animals in a number of behavioral tests (Murray and Ridley 1997, Carey *et al.* 1998). Furthermore, kynurenic acid (KYN) and its derivatives interact as antagonists with the strychnine-insensitive glycine binding site on the NMDA receptor complex (Birch *et al.* 1988, Kemp *et al.* 1988). Unfortunately, there is no uniformity in the effect of kynurenines on cognitive functions in rodents. Both beneficial effects on learning and memory tasks (Beninger *et al.* 1986, Wirsching *et al.* 1989, Wood *et al.* 1993, Smith *et al.* 1993, Viu *et al.* 2000) and their disruption (Watanabe *et al.* 1992, Ohno *et al.* 1994, Bannerman *et al.* 1997) have been reported.

In rats, the olfactory sense is considered as the most important sensory modality while solving learning and memory tasks of communicative character (Thor and Holloway 1982). We have previously shown that while MK-801 impaired social recognition potency of adult male rats (Hliňák and Krejčí 1994), kynurenic acid improved the ability of animals to recognize a familiar juvenile (Hliňák and Krejčí 1995).

In the present study, two experiments were performed using the social recognition paradigm. First, we determined deficits in recognition potency of animals given different doses of MK-801 immediately after the acquisition session (AS). Second, we evaluated whether

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kynurenic acid given before the acquisition session could attenuate or even prevent recognition deficits produced by a chosen dose of MK-801.

Experimentally naive male Hannover-Wistar rats (Konárovice Breeding, Czech Republic), 120 days old (250-300 g) were used. They were housed in a temperature controlled (20-22 ^{o}C) room in standard plastic cages, three animals per cage and maintained on a natural photoperiod for at least two weeks before the start of the experiment. Commercial pellet food and water were available *ad libitum*. Juvenile male rats, 21-24 days old, were used as social partners. The experiments were conducted in agreement with the Ethical Direction of State Law 246/l992 (CR).

MK-801 (Experiment 1: 0.05, 0.1 or 0.2 mg/kg, Experiment 2: 0.1 mg/kg) dissolved in 0.9 % saline was injected intraperitoneally, always immediately after the AS. Kynurenic acid (Experiment 2: 10 or 30 mg/kg) dissolved in 0.5 N sodium hydroxide and adjusted to pH 7.5 with tartaric acid was administered subcutaneously 60 min before the AS. Both compounds and saline or vehicle (when appropriate) were injected in a volume of 1 ml/kg.

Table 1. The effect of MK-801 and kynurenic acid (KYN) on the social investigation in rats.

Group	Dose mg/kg	N	Juvenile	Social investigation time (s)		Ratio index
				Acquisition	Retention	
Experiment 1						
Control		8	Same	131.3 ± 11.5	73.0 ± 6.7^{ab}	0.56 ± 0.04^{b}
		8	Novel	138.4 ± 12.6	176.0 ± 9.6	1.31 ± 0.09
MK-801	0.05	8	Same	192.2 ± 42.7	103.7 ± 5.7^{abc}	0.65 ± 0.08^{bc}
		8	Novel	144.0 ± 9.7	162.6 ±11.9	1.14 ± 0.07
MK-801	0.1	8	Same	164.2 ± 8.7	157.9 ± 4.8^{cd}	0.98 ± 0.06^{cd}
		8	Novel	139.3 ± 12.4	138.9 ± 12.9	1.01 ± 0.07
MK-801	0.2	8	Same	138.6 ± 14.3	124.3 ± 17.3^{cd}	0.91 ± 0.10^{cd}
		8	Novel	131.7 ± 14.1	148.9 ±16.8	1.14 ± 0.10
Experiment 2						
Control		10	Same	120.9 ± 7.8	54.7 ± 6.8^{a}	0.45 ± 0.05
MK-801	0.1	10	Same	105.6 ± 8.2	116.0 ± 5.2^{c}	1.14 ± 0.06^{c}
KYN	10	10	Same	111.0 ± 8.0	67.0 ± 6.9^{ae}	0.62 ± 0.06^{e}
KYN	30	10	Same	121.0 ± 7.5	54.8 ± 5.4^{ae}	0.46 ± 0.05^{e}
KYN+MK	10+0.1	10	Same	120.1 ± 10.1	65.5 ± 9.4^{ae}	0.56 ± 0.07^{e}
KYN+MK	30+0.1	10	Same	115.0 ± 7.2	45.9 ± 3.9^{ae}	0.41 ± 0.04^{e}

MK-801 or saline were administered (i.p.) immediately after the AS (Experiments 1 and 2). Kynurenic acid or vehicle were injected (s.c.) 60 min before the AS (Experiment 2). The RS was performed 30 min after the AS. Same juvenile: during the RS adult males were interacted with the same juvenile as in the AS. Novel juvenile: during the RS adult males were interacted with a novel juvenile. Ratio index means the ratio of the investigation time during the RS to that during the AS. Data are expressed as mean ± SEM values. Statistical significance, P<0.05: Wilcoxon test, [a] the RS vs. the AS (Experiments 1 and 2); Kruskal-Wallis ANOVA test (df 7 and 5, respectively), [b] vs. corresponding novel group (Experiment 1), [c] vs. corresponding control group (Experiments 1 and 2), [d] vs. corresponding 0.05 mg/kg MK-801 group (Experiment 1), [e] vs. MK-801 group (Experiment 2).

The procedure was identical to that described previously (Hliňák and Krejčí 2002). Briefly, 24 h before the start of testing the adult males were housed individually. During the AS each adult male was exposed to a juvenile. The retention session (RS) followed 30 min later: the adult males were re-exposed to the same or a novel juvenile (Experiment 1). In Experiment 2, the adults were interacted with the same juvenile only. Each

session lasted 5 min. The testing was conducted between 08:00 and 12:00 h in the room illuminated with a 25-W fluorescent tube. An experienced observer recorded the duration of behaviors oriented toward the juvenile (head and body sniffing, genital exploration, close pursuing, touching the flanks with the snout, manipulation with the forepaws), defined as the total time of social investigation.

To compare the difference within groups (AS vs. RS) the Wilcoxon matched-pairs signed ranks test was used. To compare the absolute time within sessions (as well as the ratio index) the Kruskal-Wallis analysis of variance followed by Dunn's test was used. Statistical significance was accepted when $P<0.05$.

The data on the social investigation of adult male rats are summarized in Table 1. *Experiment 1:* The overall analysis revealed a significant difference in the effect of MK-801 ($H=36.7$, $P<0.0001$ for the RS, and $H=36.9$, $P<0.0001$ for ratio index; df 7). When re-exposed to same juveniles the investigation time of the adults given 0.1 and 0.2 mg/kg of MK-801 corresponded to that measured during the AS. Further, the investigation time of the controls and of males given 0.05 mg/kg dose was significantly reduced. When re-exposed to novel juveniles the investigation time in all MK-801 treated groups as well as in the control group was similar to the time observed in the AS. Data on the ratio index confirm the above-mentioned differences. Therefore, the dose of 0.1 mg/kg was chosen as effective in inducing the deficit in social recognition potency. Also, the present results confirm our previous findings (Hliňák and Krejčí 1994). *Experiment 2:* The overall analysis revealed no significant difference in the AS ($H=2.4$, $P=0.79$, df 5) which implies that KYN administered 60 min before had no effect on social investigation of the adults. However, significant differences in the absolute investigation time during the re-exposure to same juveniles ($H=26.5$, df 5, $P<0.0001$) as well as in the ratio index ($H=29.6$, df 5, $P<0.0001$) were revealed. A significant reduction in social investigation was measured in the controls, KYN alone treated, and KYN plus MK-801 treated animals as compared to those treated only with MK-801.

The present data show that KYN given 60 min before the initial interaction prevented amnesia for recognition of familiar juveniles produced by MK-801. Namely, KYN plus MK-801 treated adult males were able to recognize a juvenile that they had encountered 30 min before. This result extends our previous finding that systemic administration of KYN improves working memory for olfactory stimuli elicited by juvenile conspecifics (Hliňák and Krejčí 1995). Since MK-801 was administered immediately after the AS, the effect of KYN evidently relates to both the consolidation of acquired olfactory information and the retention of memory traces. Consequently, the retrieval of memory traces seems to be facilitated. However, we cannot exclude that the administration of KYN before the AS reinforced perception of olfactory stimuli that is not accompanied by an increase of social investigation. In every case, the present results are in agreement with findings reporting anti-amnesia and beneficial effects of KYN in cognitive processes (Beninger *et al.* 1986, Smith *et al.* 1993, Wood *et al.* 1993, Viu *et al.* 2000). Since KYN was applied prior to the MK-801, the question arises whether MK-801 can act at its binding site at all. It is possible that KYN could reduce the accessibility of this open-channel blocker.

Agonists, such as milacemide (Handelmann *et al.* 1989), D-cycloserine (Monahan *et al.* 1989, Flood *et al.* 1992, Kawabe *et al.* 1998) and 1-aminocyclopropanecarboxylic acid (Viu *et al.* 2000), acting at the strychnine-insensitive glycine site, also facilitated the performance of animals in various learning and memory tasks. It seems that compounds acting in an antagonistic or agonistic way on the binding sites of the NMDA receptor-channel complex can contribute with different potencies to the expression of behavior related to memory/recognition performances.

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Reprint requests

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