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BRIEF COMMUNICATION

NMDA Receptor Inhibition Prevents Tolerance to Cocaine

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DE MONTIS, M. G., P. DEVOTO, D. MELONI, C. GAMBARANA, G. GIORGI AND A. TAGLIAMONTE. *NMDA receptor inhibition prevents tolerance to cocaine.* PHARMACOL BIOCHEM BEHAV 42(1) 179–182, 1992.—Male rats were treated with cocaine by utilizing two different experimental paradigms. One group of animals received a low dose (10 mg/kg, IP) of cocaine for 7 days. A second group received 40 mg/kg IP of cocaine for 3 days. In both experimental groups, half the animals were concomitantly treated with 0.25 mg/kg IP (+)-5methyl-10,11-dihydro-5*H*-dibenzo-[*a*,*d*]-cyclohepten-5,10-imine maleate (MK-801), a noncompetitive NMDA receptor antagonist. Rats treated with the low dose of cocaine after 7 days developed tolerance to the stimulation of locomotor activity induced by cocaine and by the dopamine D_2 agonist quinpirole. Rats treated with 40 mg/kg of cocaine showed a marked behavioral sensitization. Both these effects, tolerance and sensitization, were prevented by coadministration of MK-801, thus suggesting these two phenomena are different aspects of a common neuronal response in which NMDA transmission plays a crucial role.

Central stimulants Tolerance NMDA receptor antagonists Behavioral sensitization Quinpirole

IT is well established that repeated, intermittent administrations of *d*-amphetamine, cocaine, and related central stimulants sensitize animals to both the increased locomotion and stereotyped activity induced by these compounds (9,11). The behavioral sensitization produced by amphetamine and cocaine, also known as reverse tolerance, has been linked to an increased dopaminergic neurotransmission (7), which would result in a D_2 receptor supersensitivity (14). However, biochemical studies carried out in animals made supersensitive to central stimulants indicate an involvement of both dopaminergic receptor subtypes (1,5). The complexity of these results prompted the search for other mechanisms underlying the behavioral phenomenon of reverse tolerance. Recently, sensitization was equated to the phenomenon of long-term potentiation since, like this model of neuronal memory (4), its occurrence can be prevented by *N*-methyl-D-aspartate (NMDA) receptor blockade (6). In fact, the development of sensitization to amphetamine or cocaine could be prevented in different animal models (3,6) by coadministering (+)-5 methyl-10,11-dihydro-5*H*-dibenzo-[*a*,*d*]-cyclohepten-5, 10-imine maleate (MK-801), a potent noncompetitive NMDA receptor blocker, with the central stimulant (15). In addition, MD-801 also prevented the occurrence of both tolerance to and dependence on morphine (13).

Repeated administration of central stimulants may initially produce desensitization to the acute effects, that is, tolerance, which is usually followed by the appearance of sensitization (10). The treatment paradigm is crucial to observe the two different phenomena. In fact, the closer together in time drug injections are given the more likely tolerance will develop and the later sensitization will occur.

The present report confirms that MK-801, coadministered daily with cocaine, prevented the development of sensitization and, in addition, shows that pretreatment with MK-801 prevented the occurrence of tolerance to cocaine effects.

METHOD

Animals

Male Sprague-Dawley rats (Charles River, Como) weighing 200–250 g were used. Animals were kept on a 12 L : 12 D cycle (lights on from 0800 to 2000 h) with free access to food and water. Testing was conducted between 1000–1700 h.

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TABLE 1

EFFECT OF MK-801 ON THE DEVELOPMENT OF TOLERANCE TO COCAINE-INDUCED HYPERMOTILITY

	First Day of Treatment		Fourth Day of Treatment		Eighth Day of Treatment		Quinpirole (0.3 mg/kg) on Day 8	
Pretreatment	Motility Counts (40 min)	Stereotypy Scores	Motility Counts (40 min)	Stereotypy Scores	Motility Counts (40 min)	Stereotypy Scores	Motility Counts (40 min)	Stereotypy Scores
Saline	1264 ± 119	1.81 ± 0.30	1350 ± 150	2.00 ± 0.25	2476 ± 182	5.15 ± 0.32	2205 ± 205	4.50 ± 0.12
Cocaine 10 mg/kg	2545 ± 260*	3.35 ± 0.38*	1605 ± 300†	3.80 ± 0.32	854 ± 113‡	4.00 ± 0.16	1008 ± 151*	3.50 ± 0.26
MK-801 0.25 mg/kg	1483 ± 257	2.83 ± 0.35	1620 ± 194	2.60 ± 0.18	2635 ± 289†	5.33 ± 0.15	1980 ± 227†	4.60 ± 0.15
MK-801 + cocaine	5906 ± 723‡	5.50 ± 0.26‡	4289 ± 630‡	5.00 ± 0.25‡	2372 ± 424†	4.83 ± 0.17	2351 ± 207‡	4.00 ± 0.28

Animals were treated as described in the Methods Section. Saline and MK-801 groups received 10 mg/kg cocaine (or quinpirole) only on day 8. Each value represents the mean ± SEM of 24 animals, except for day 8 experiments, where rats were divided into two subgroups of 12 animals each and challenged with cocaine (10 mg/kg) and quinpirole (0.3 mg/kg), respectively.

*Value is significantly different from control ($p < 0.05$).

†Value is significantly different from cocaine ($p < 0.05$).

‡Value is significantly different from control ($p < 0.01$).

Behavioral Equipment and Testing

Motor activity was measured by placing animals individually in motility cages (M/P40 Fc Electronic Motility Meter, Motron Products, Stockolm). Each cage had 40 photoconductive sensors placed in the floor area (21 × 32 cm) at a fixed distance of 4 cm. The sensors were lit uniformly by an incandescent lamp mounted 60 cm above them. Motor activity was defined as the number of interruptions of a beam. On the test day, animals were observed for 30 min before and 40 min after treatment. Motility was evaluated also on a qualitative basis and stereotyped movements (i.e., sniffing, rearing, licking, and chewing syndromes) were scored distinctly, from 0–6, according to the Creese and Iversen (2) rating scale.

Drugs

Cocaine hydrochloride (Sigma Chemical, St. Louis, MO), quinpirole (Ly-17-1555, Eli-Lilly & Co., Indianapolis, IN), and MK-801 (RBI Res. Biochem. Inc., Natick, MA) were dissolved in 0.9% saline and injected IP in a volume of 0.1 ml/100 g rat body weight.

Procedures

Two different experimental paradigms were used:

1. Ninety-six rats were divided into four groups of 24 animals each. The first group was treated with 10 mg/kg IP cocaine, once a day, administered 30 min after 0.25 ml saline IP. The second group received a daily injection of MK-801 (0.25 mg/kg, IP), followed by 0.25 ml saline. The third group received MK-801 and cocaine, injected in this order, at 30-min intervals. The effect of each treatment on motility was scored the first day to have a baseline value of each animal and every fourth day to evaluate possible changes in response to treatments. Controls and MK-801 groups were tested for cocaine response only on day 8.
2. Seventy-two rats were divided into three groups of 24 animals each. The first group was treated with 40 mg/kg IP cocaine once a day for 3 days administered 30 min after 0.25 ml saline given IP. The second group received the same dose of cocaine 30 min after MK-801 (0.25 mg/kg, IP). The third group (controls) received two daily injections of 0.25 ml saline IP at 30-min intervals. All treatments were carried out in the test cage during a 70-min period to strengthen environmental contingencies and favor the occurrence of reverse tolerance (8).

Twenty-four hours after the last treatment in both experimental paradigms, each group of animals was divided into two subgroups and tested for motor activity after acute challenge with cocaine (10 mg/kg, IP) or quinpirole (0.25 mg/kg, IP).

Statistics

All data are expressed as mean ± SEM. As multiple drug treatments were compared with values from a single control group, statistical comparisons were made by analysis of variance (ANOVA) followed by post hoc analysis using the Bonferroni test ($p < 0.05$).

RESULTS

As previously observed (6), cocaine given acutely at the dose of 10 mg/kg produced an increase of coordinated motor activity, while at the dose of 40 mg/kg cocaine-induced intense motor activity was disrupted by frequent bursts of stereotyped movements. MK-801 (0.25 mg/kg) injected alone never showed significant effects on spontaneous motor activity. However, MK-801 administered 30 min before cocaine (10 mg/kg, IP) clearly potentiated the hypermotility produced by this dose of cocaine, both acutely and after 8 days of treatment. Rats treated for 8 days in their home cage with 10 mg/kg of cocaine IP once a day were tested for locomotor activity. Table 1 shows that animals had developed a complete tolerance to the increased locomotor activity produced both by cocaine (given at the same dose) and by 0.3 mg/kg quinpirole, a centrally acting dopamine D_2 receptor agonist (12). The decreased hypermotility was real and not related to an increase of stereotyped movements; in fact, 10–15 min after treatment most animals were lying in a corner of the cage. On the other hand, MK-801 (0.25 mg/kg) coadministered with cocaine (10 mg/kg) for 8 days prevented the development of tolerance to the acute effects of cocaine.

Table 2 shows that injections of 40 mg/kg cocaine IP for 3 days produced a marked sensitization to the acute effect on locomotor activity of both cocaine (10 mg/kg) and quinpirole (0.3 mg/kg). The two compounds, injected IP 24 h after the last treatment, induced stereotypies in more than 50% of the

TABLE 2

EFFECT OF MK-801 ON COCAINE-INDUCED SENSITIZATION

	Acute Cocaine 10 mg/kg		Acute Quinpirole	
Pretreatment	Motility Counts (40 min)	Stereotypy Scores	Motility Counts (40 min)	Stereotypy Scores
Saline	2007 ± 122	4.57 ± 0.25	1668 ± 318	3.80 ± 0.26
Cocaine 40 mg/kg	3098 ± 337*	8.67 ± 0.43†	2624 ± 251*	6.42 ± 0.22†
MK-801 + Cocaine	1806 ± 257‡	4.25 ± 0.10§	1735 ± 331‡	4.20 ± 0.35§

Animals were treated as described in methods. Cocaine and quinipirole values reported in the table refer to the fourth day of treatment. Each value represents the mean ± SEM of 12 animals.

*Value is significantly different from control ($p < 0.05$).

†Value is significantly different from control ($p < 0.01$).

‡Value is significantly different from cocaine ($p < 0.05$).

§Value is significantly different from cocaine ($p < 0.01$).

animals. Coadministration of MK-801 prevented the occurrence of reverse tolerance to cocaine, in agreement with previous observations (6).

DISCUSSION

Daily administration of cocaine at a relatively low dose (10 mg/kg) for 8 days resulted in a markedly reduced response to both cocaine and quinpirole, a selective dopamine D_2 receptor agonist. On the other hand, as expected, cocaine (40 mg/kg) injected daily for 3 consecutive days produced marked supersensitivity to the stimulant effect of both cocaine (10 mg/kg) and quinpirole (0.3 mg/kg).

The occurrence of both these effects was significantly prevented by pretreating rats 30 min before each dose of cocaine with MK-801, a selective, noncompetitive NMDA receptor antagonist. The fact that MK-801 significantly potentiated the stimulatory effect of 10 mg/kg cocaine on motility might suggest that the lack of tolerance observed after 8 days of treatment is an effect on the expression of tolerance, not on its occurrence. On the other hand, 0.25 mg/kg MK-801, which per se had no significant effect on motility by potentiating cocaine, would be expected to favor the development of supersensitivity. Such a mechanism could explain the failure of tolerance to occur. The effect present after 8 days of treatment would represent the algebraic sum of tolerance plus reverse tolerance. However, such an explanation appears paradoxical in view of the fact that MK-801 also prevented the development of supersensitivity to cocaine. Moreover, at this point it is relevant to emphasize that in rats MK-801 is able to prevent the occurrence of tolerance to the analgesic effect of morphine as measured by the tail-flick test and of opiate dependence as measured by the naloxone-precipitated abstinence syndrome (13). All reported findings substantiate the role of excitatory amino acid systems in behavioral changes and neuronal plasticity produced by long-term exposure to different psychotropic drugs.

The fact that NMDA receptor blockade prevented the development of both sensitization and tolerance produced by repeated cocaine administration suggests the two phenomena represent distinct aspects of a common neuronal response in which NMDA transmission plays a crucial role.

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REFERENCES

1. Barnett, J. V.; Kuczenski, R. Desensitization of rat striatal dopamine-stimulated adenylate cyclase after acute amphetamine administration. J. Pharmacol. Exp. Ther. 237:820–825; 1986.
2. Creese, I.; Iversen, S. D. The pharmacological and anatomical substrates of the amphetamine response in the rat. Brain Res. 83: 419–436; 1975.
3. Criswell, H. E.; Mueller, R. A.; Breese, G. R. Long-term D_1-dopamine receptor sensitization in neonatal 6-OHDA-lesioned rats is blocked by an NMDA antagonist. Brain Res. 512:284–290; 1990.
4. Collingridge, G. L.; Davies, S. N. NMDA receptors and long-term potentiation in the hippocampus. In: Watkins, J. C.; Collingridge, G. L., eds. The NMDA receptor. Oxford: Oxford University Press; 1989:123–135.
5. Howlett, D. R.; Nahorski, S. R. Acute and chronic amphetamine treatments modulate striatal dopamine receptor binding sites. Brain Res. 161:173–178; 1989.
6. Karler, R.; Calder, L. D.; Chandry, I. A.; Turkanis, S. A. Blockade of "reverse tolerance" to cocaine and amphetamine by MK-801. Life Sci. 45:599–606; 1989.
7. Kazahaya, Y.; Akimoto, K.; Otsuki, S. Subchronic methamphetamine treatment enhances methamphetamine or cocaine-induced dopamine influx in vivo. Biol. Psychiatry 25:903–912; 1989.
8. Post, R. M.; Lochkfeld, A.; Squillace, K. M.; Contel, N. R. Drug-environment interaction: Context dependency of cocaine-induced behavioral sensitization. Life Sci. 28:755–760; 1981.
9. Post, R. M.; Rose, H. Increasing effects of repetitive cocaine administration in the rat. Nature 260:731–732; 1976.
10. Robinson, T. E.; Becker, J. B. Enduring changes in brain and behavior produced by chronic amphetamine administration: A review and evaluation of animal models of amphetamine psychosis. Brain Res. Rev. 11:157–198; 1986.
11. Segal, D. S.; Mandell, J. A. Long-term administration of d-amphetamine: Progressive augmentation of motor activity and stereotypy. Pharmacol. Biochem. Behav. 2:249–255; 1974.
12. Spiraky, C.; Fibiger, H. C. Behavioral evidence of supersensitivity of postsynaptic dopamine receptors in the mesolimbic system after chronic administration of desipramine. Eur. J. Pharmacol. 74:195–206; 1981.
13. Trujillo, K. A.; Akil, H. Inhibition of morphine tolerance and dependence by the NMDA receptor antagonist MK-801. Science 251:85–87; 1991.
14. Ujike, H.; Akiyama, K.; Otsuki, S. D_2 but not D_1 dopamine agonists produce augmented behavioral response in rats after subchronic treatment with methamphetamine or cocaine. Psychopharmacology (Berl.) 102:459–464; 1990.
15. Wong, E. H. F.; Kemp, J. A.; Pristley, T.; Knight, A. R.; Woodruff, G. N.; Iversen, L. L. The anticonvulsant MK-801 is a potent *N*-methyl-D-aspartate antagonist. Proc. Natl. Acad. Sci. USA 83: 7104–7108; 1986.