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MK-801 interferes with the acquisition of amphetamine- and lithium-induced place conditioning

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The place conditioning paradigm was used to assess the ability of the noncompetitive NMDA receptor antagonist, MK-801, to interfere with drug–place associations. MK-801 (0.1 mg/kg, i.p.) attenuated a place preference produced by high, but not low to moderate, doses of amphetamine. Interference with amphetamine place preference learning was not the result of state-dependent retrieval produced by MK-801. Furthermore, MK-801 did not interfere with the expression of a previously established amphetamine place preference. Pretreatment with MK-801 also interfered with lithium-induced conditioned place aversion learning. These results suggest that the attenuation of place preference conditioning produced by MK-801 is the result of its interference with learning rather than with the rewarding properties of drugs.

The *N*-methyl-D-aspartate (NMDA) glutamate receptor plays a critical role in learning and memory processes (see, e.g., Lynch & Baudry, 1984; Morris, Anderson, Lynch, & Baudry, 1986; Robinson, Crooks, Shinkman, & Gallagher, 1989). Both competitive and noncompetitive antagonists of the NMDA receptor produce deficits in acquisition of behavior indicative of olfactory conditioning (Staubli, Thibault, DiLorenzo, & Lynch, 1989), spatial learning (Butelman, 1989; Davis, Butcher, & Morris, 1992; Heale & Harley, 1990; Morris et al., 1986; Shapiro & Caramanos, 1990), conditioned fear (J. J. Kim, DeCola, Landeira-Fernandez, & Faneslow, 1991; Miserendino, Sananes, Melia, & Davis, 1990), extinction of conditioned fear (Falls, Miserendino, & Davis, 1992), and conditioned taste avoidance (Welzl, Alessandri, & Bättig, 1990). Blockade of this receptor by antagonists also impedes the induction of long-term potentiation (LTP), a physiological model that has been used to characterize learning at the synaptic level (Collingridge & Bliss, 1987; Collingridge, Kehl, & McLennan, 1983; Morris et al., 1986). A number of investigators have suggested that learning and memory deficits produced by NMDA antagonists may be related to the blockade of LTP (e.g., Morris et al., 1986), although this relation is a matter of controversy (e.g., Keith & Rudy, 1990). In the present study, we investigated the potential of NMDA antagonists to modify the association produced between a place and a rewarding (amphetamine) or aversive (lithium chloride and naloxone) drug in the classical conditioning paradigm of place conditioning.

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MK-801 and Drug Association

Blockade of the NMDA receptor also modifies long-lasting behavioral plasticity associated with chronic drug administration, including the acquisition of tolerance to the analgesic effects of morphine (Trujillo & Akil, 1991), as well as the sensitization to the locomotor activity effects of morphine (Jeziorski, White, & Wolf, 1994), cocaine (Karler, Calder, Chaudhry, & Turkanis, 1989; Pudiak & Bozarth, 1993), and amphetamine (Karler et al., 1989; Stewart & Druhan, 1993). The NMDA antagonist MK-801 also interferes with sensitization to the rewarding properties of cocaine in the self-administration paradigm (Schenk et al., 1993), suggesting that the disruption of NMDA function might attenuate properties of drugs that contribute to abuse liability. NMDA receptor antagonism also interferes with the establishment, but not with the expression of amphetamine-induced conditioned locomotion (Stewart & Druhan, 1993). These findings suggest that NMDA antagonists interfere with associative learning (Popik, Layer, & Skolnick, 1995; Trujillo & Akil, 1991).

MK-801 and Place Conditioning

There is considerable recent evidence that MK-801 interferes with the establishment of both morphine (Clavier, Nores, Olsen, & Vaccarino, 1996; H. S. Kim, Jang, & Park, 1996; Tzschentke & Schmidt, 1995) and cocaine (Cervo & Samanin, 1995) induced conditioned place preference learning. This effect appears to be specific to the antagonism of the NMDA receptor because it is stereo-

selective (Del Pozo, Barrios, & Baeyens, 1996). The place conditioning paradigm is a measure of the rewarding or aversive properties of drugs (for reviews, see Carr, Fibiger, & Phillips, 1989, and van der Kooy, 1987). With this procedure, during a training period, rats are confined to one distinctive compartment following injection of a drug, and to an alternative compartment following injection of an inert substance. Their preference for either chamber is subsequently evaluated during a test when they are undrugged and are allowed to explore both compartments. It is inferred that the drug is rewarding if the rats display a preference for the drug-paired compartment (and aversive if the rats display a preference for the alternative compartment).

There is abundant evidence that amphetamine is effective in inducing a conditioned place preference (see Carr et al., 1989; Reicher & Holman, 1977). The strength of the preference appears to increase with amphetamine dose (Erb & Parker, 1994; Gilbert & Cooper, 1983; Hoffman & Beninger, 1988; Laviola, Dell'Omo, Chiarotti, & Bignami, 1994; Richardson et al., 1993; Spyraki, Fibiger, & Phillips, 1982; although see Costello, Carlson, Glick, & Bryda, 1989, and Wall, Hinson, Schmidt, Johnston, & Streather, 1990); in fact Erb and Parker (1994) reported that the strength of an amphetamine-induced place preference increased with doses ranging from 1 to 10 mg/kg, even though at the higher doses amphetamine-induced stereotypy is apparent during conditioning.

Although MK-801 interferes with morphine- and cocaine-induced place preference learning (Cervo & Samanin, 1995; Clavier et al., 1996; H. S. Kim et al., 1996; Tzschentke & Schmidt, 1995), it has been reported that MK-801 is *ineffective* in interfering with amphetamine-induced place preference learning (Hoffman, 1994). Since MK-801 does interfere with amphetamine-induced sensitization (Karler et al., 1989; Stewart & Druhan, 1993) and conditioned locomotion (Stewart & Druhan, 1993), it is surprising that it does not interfere with amphetamine-induced place preference learning.

The following experiments were conducted to reexamine the potential of MK-801 to interfere with amphetamine-induced place preference learning. In Experiment 1, we evaluated the ability of MK-801 to interfere with a two-trial amphetamine- (3 mg/kg) induced place preference. Like Hoffman (1994), we found that MK-801 did not interfere with the establishment of a place preference produced by a moderate dose of amphetamine. However, in subsequent experiments, we found that when the dose of amphetamine was increased to 10 mg/kg, which produced a stronger place preference, MK-801 did effectively interfere with place preference learning.

EXPERIMENT 1

In Experiment 1, we examined the ability of a low dose of MK-801 (0.1 mg/kg, i.p.) to interfere with the establishment of one- or two-trial amphetamine- (3 mg/kg, i.p.) induced place preference learning. Because of controversial side effects of MK-801 on sensorimotor processes (Hargreaves & Cain, 1992; Keith & Rudy, 1990), we selected the lowest dose that has been shown to effectively modify place preferences produced by other drugs (Cervo & Samanin, 1995; H. S. Kim et al., 1996; Tzschentke & Schmidt, 1995). At this dose of MK-801, groups are found to be hyperactive in comparison with saline controls, but this level of hyperactivity is minimal when compared with that of groups receiving the higher doses used in the literature (Hargreaves & Cain, 1992).

Method

Subjects. The subjects in Experiment 1 were 56 male Sprague–Dawley rats (Charles River Labs, Quebec) weighing 238–285 g on the 1st day of conditioning. The rats were housed in pairs in transparent plastic cages with woodchip bedding, and they were provided with food (Lab Diet, PMI Feeds Inc.) and water ad lib. throughout the study. A 12:12-h light:dark schedule (0800–2000) was in effect in the temperature-controlled colony room (21° ± 1° C). After 5 days of adaptation to the laboratory, the experimental procedures began. All rats were conditioned and tested at the same time of day during the light cycle.

Apparatus. The place conditioning apparatus (see Parker, 1992) consisted of four wooden shuttleboxes, each bisected into two distinctive chambers (35 × 25 × 29 cm) by four removable dividers. All chamber surfaces were painted flat black, and were identical except for floor texture (5-cm black sandpaper strips or 0.5-cm wire mesh). When evaluated by group means, the preferences displayed for these chamber cues did not significantly differ when the rats were tested for 15 min following two pairings of each chamber with saline (mean time in sandpaper chamber, 440 sec; mean time in mesh chamber, 460 sec). The number of seconds spent in each chamber during the testing phase was recorded by an overhead videocamera with the signal analyzed by a video-tracking system (Videomex-V, Columbus Instruments).

Drugs. MK-801 (Research Biochemicals, Natick, MA) and *d*-amphetamine sulphate (NIDA, Research Triangle Park, NC) were dissolved in physiological saline (0.9%) and delivered at volumes of 1 ml/kg and 2 ml/kg, respectively. The dose of MK-801 was 0.1 mg/kg. The amphetamine dose was 3 mg/kg. All drug injections were administered i.p.

Procedure. The rats received either one ($n = 32$) or two ($n = 24$) cycles of conditioning trials prior to being tested for a place preference. A conditioning cycle consisted of a saline conditioning trial, followed 24 h later by an amphetamine conditioning trial. For the rats that received two conditioning cycles, the cycles were separated by 48 h. On a saline conditioning trial, all rats received two injections of saline, spaced 30 min apart. Five minutes after the second injection, the rats were placed into one of the chambers (sandpaper or mesh) of the conditioning apparatus for 30 min. On an amphetamine conditioning trial, all rats were injected with 3 mg/kg of amphetamine, 5 min prior to a 30-min confinement in the chamber opposite to that which they experienced on the saline conditioning trial. Thirty minutes prior to the amphetamine injection, half of the rats in each group were injected with MK-801, and half of the rats in each group were injected with physiological saline.

The place preference test occurred 3 days after the final conditioning trial. During the test, the dividers were removed between the boxes, and the rats were placed at the intersection of the two chambers. The rats were allowed to explore both chambers, drug free, for a 15-min period.

Data analysis. The data were converted to difference scores that represented the mean seconds in the amphetamine-paired chamber minus the mean seconds in the saline-paired chamber. A place preference was determined by comparing the mean difference score for

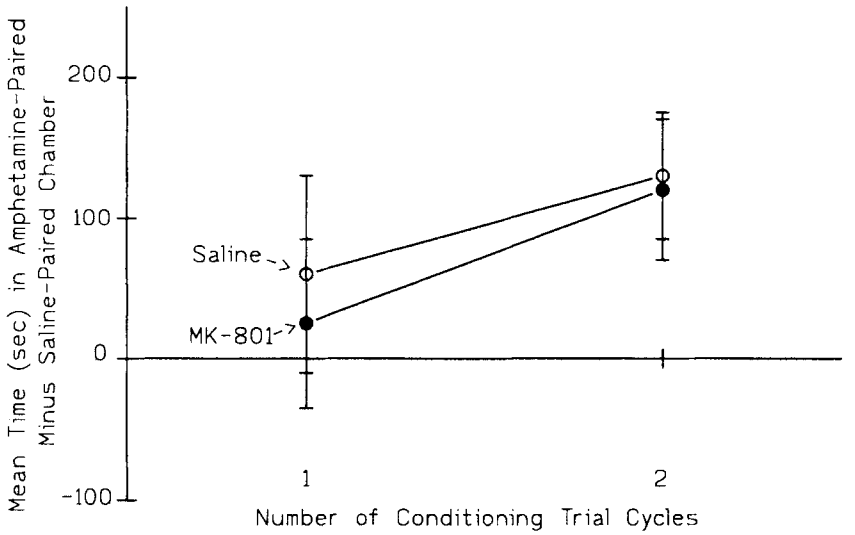

Figure 1. Mean difference scores (+*SEM*) during the place preference test in Experiment 1. The groups previously received one or two conditioning cycles with amphetamine (3 mg/kg) that were preceded by an injection of MK-801 (.1 mg/kg) or saline.

each group from a value of zero by means of *t* tests. Since the side paired with amphetamine was counterbalanced, any unconditional side preferences should be equally distributed among the groups. Therefore, a zero baseline was the value expected if the rats displayed neither a place preference nor a place aversion.

Results

MK-801 did not modify the strength of an amphetamine (3 mg/kg) induced place preference following one or two conditioning trials in Experiment 1. Figure 1 presents the mean number of seconds that the rats pretreated with MK-801 or saline during one or two conditioning trial cycles spent in the amphetamine-paired chamber minus the number of seconds that they spent in the saline-paired chamber (difference score $\pm SEM$) during the place preference test of Experiment 1. Following two, but not one, conditioning trial cycles, rats pretreated with both MK-801 ($p < .025$) and saline ($p < .01$) displayed difference scores that were significantly greater than zero. However, a 2 × 2 between groups analysis of variance (ANOVA) with the factors of pretreatment condition (MK-801 or saline) and number of conditioning trial cycles (one or two) revealed no significant effects.

Discussion

MK- 801 did not interfere with the place preference produced by 3 mg/kg of amphetamine. These findings agree with those of Hoffman (1994), who also failed to find that MK-801 interfered with a two-trial place preference established with a relatively low dose of amphetamine (2 mg/kg).

EXPERIMENT 2

MK-801 did not interfere with the establishment of a place preference produced by a moderate dose of amphetamine. However, an evaluation of the results of experiments in which MK-801 successfully interfered with place preference learning suggested, paradoxically, that a sufficiently strong baseline association must be established before such interference was apparent. In the studies that have demonstrated interference with place preference learning by MK-801 pretreatment, rats received three to four conditioning trials (Cervo & Samanin, 1995; H. S. Kim et al., 1996; Tzschentke & Schmidt, 1995), whereas, in Hoffman's (1994) study and in Experiment 1 above, rats received only two conditioning trials with a relatively low dose of amphetamine (2–3 mg/kg). In fact, Clavier et al. (1996) have reported that MK-801 did not interfere with the acquisition of a one-trial morphine place preference, but did interfere with acquisition of a stronger two-trial morphine place preference. In Experiment 2, the potential of MK-801 to interfere with a two-trial place preference produced by 5 and 10 mg/kg of amphetamine was investigated. Higher doses of amphetamine produce stronger place preferences than lower doses (see, e.g., Erb & Parker, 1994).

Method

Subjects. Ninety-three male Sprague–Dawley rats, weighing 250–270 g on the 1st day of conditioning, were treated identically as in Experiment 1 except as indicated. As in Experiment 1, MK-801 and *d*-amphetamine sulphate were dissolved in physiological saline (0.9%) and delivered at volumes of 1 ml/kg and 2 ml/kg, respectively. The dose of MK-801 was 0.1 mg/kg. The amphetamine dose was 5 mg/kg or 10 mg/kg. All drug injections were administered i.p.

Procedure. As in Experiment 1, a conditioning cycle consisted of a saline conditioning trial followed 24 h later by an amphetamine conditioning trial. The two conditioning cycles were separated by 48 h. On a saline conditioning trial, all rats received two injections of saline spaced 30 min apart. Five minutes after the second injection, the rats were placed into one of the chambers (sandpaper or mesh) of the conditioning apparatus for 30 min. On an amphetamine

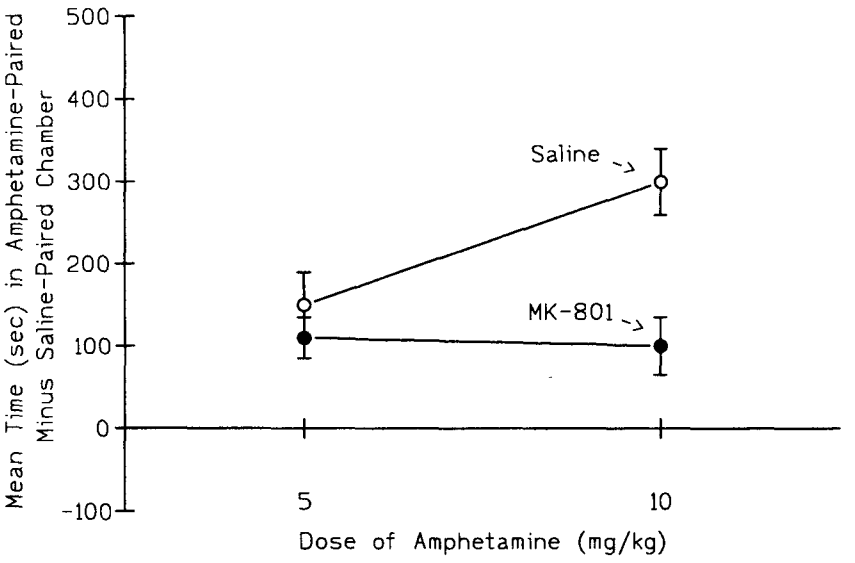

Figure 2. Mean difference scores (+*SEM*) during the place preference test of Experiment 2. The groups received two conditioning cycles with 5- or 10-mg/kg amphetamine that were preceded by an injection of MK-801 or saline.

conditioning trial, rats were injected with 5 mg/kg (n = 47) or 10 mg/kg (n = 46) of amphetamine, 5 min prior to 30-min confinement in the chamber opposite to that which they experienced on the saline conditioning trial. Thirty minutes prior to the amphetamine injection, half of the rats in each group were injected with MK-801 (n = 24), and half were injected with physiological saline (n = 23). The rats injected with 10 mg/kg of *d*-amphetamine displayed stereotyped behaviors when returned to their home cages for a number of hours on the day of conditioning. However, a period of 48 h intervened between the amphetamine injection and the subsequent saline trial to prevent carryover effects of the drug to the nondrug trial.

The place preference test occurred 3 days after the final conditioning trial. During the test, the dividers were removed between the boxes, and the rats were placed at the intersection of the two chambers. The rats were allowed to explore both chambers, drug free, for a 15-min period.

Results

MK-801 interfered with the establishment of a place preference produced by 10 mg/kg of *d*-amphetamine, but not 5 mg/kg of *d*-amphetamine. Figure 2 presents the mean difference score (+*SEM*) for the MK-801– and the saline-pretreated groups conditioned with 5 or 10 mg/kg of amphetamine on each of two conditioning trial cycles during the preference test of Experiment 2. A 2 × 2 between-groups ANOVA with the factors of pretreatment condition (MK-801 or saline) and dose of amphetamine (5 or 10 mg/kg) revealed a pretreatment condition effect [$F(1,89) = 14.0, p < .01$] and a pretreatment condition × dose interaction [$F(1,89) = 5.1, p < .025$]. Subsequent Newman–Keuls pairwise comparison tests revealed that for rats conditioned at a dose of 10 mg/kg of amphetamine, but not 5 mg/kg of amphetamine, the saline-pretreated rats displayed a greater amphetamine-induced place preference than did the MK-801–pretreated rats ($p < .01$). Furthermore, among the rats pretreated with saline, but not the rats pretreated with MK-801, 10 mg/kg of amphetamine produced a significantly greater place preference than did 5 mg/kg of amphetamine ($p < .05$). However, MK-801 did not block the establishment of an amphetamine-induced place preference because the MK-801–pretreated groups conditioned with both 5 and 10 mg/kg of *d*-amphetamine displayed difference scores greater than zero ($ps < .05$).

Discussion

Although MK-801 did not interfere with the establishment of an amphetamine place preference produced by 3 (Experiment 1) or 5 (Experiment 2) mg/kg of *d*-amphetamine, it did attenuate the establishment of a preference produced by 10 mg/kg of *d*-amphetamine. As previously reported by Erb and Parker (1994), the saline-pretreated group in Experiment 2 displayed a stronger amphetamine place preference when conditioned with 10 mg/kg of amphetamine than when conditioned with 5 mg/kg. Because the strength of amphetamine place conditioning increases as the dose of amphetamine increases, it is possible that MK-801 more effectively interferes with stronger than weaker place–drug associations. In support of this suggestion, it has recently been reported that MK-801 (0.1 mg/kg) interferes with a morphine (5 mg/kg) place preference produced by two training trials, but not a preference produced by a single training trial (Clavier et al., 1996). Our findings suggest that Hoffman's (1994) failure to detect MK-801–induced disruption of place preference learning may have been due to the weakness of the place preference produced by 2 mg/kg of amphetamine across only two conditioning cycles. It should be noted that in other reports of MK-801–induced interference with morphine- (Del Pozo et al., 1996; H. S.

Kim et al., 1996; Tzschentke & Schmidt, 1995) or cocaine- (Cervo & Samanin, 1995) induced place preference learning, the rats were tested following three to four conditioning cycles.

EXPERIMENT 3

MK-801 appeared to interfere with the acquisition of an amphetamine place preference, suggesting that it interfered with learning. Conversely, the hedonic properties of MK-801 alone might contribute to its ability to interfere with place preference learning. If MK-801 is aversive (under the conditions of our findings), it would be expected to attenuate the amphetamine place preference without necessarily modifying learning. The summation of aversive properties of MK-801 and the rewarding properties of amphetamine might have prevented the establishment of an amphetamine place preference in the MK-801 pretreated groups in Experiment 2. In Experiment 3, we evaluated the effects of MK-801 alone administered under the same conditions as in Experiment 2.

Method

Twelve male Sprague–Dawley rats, weighing 255–265 g on the 1st day of conditioning, were treated identically as in Experiment 2 except as specified. As in Experiment 2, all rats received two cycles of conditioning trials separated by 48 h. The first trial of a cycle was a saline conditioning trial, conducted identically to that of Experiment 2. However, on the second trial of each cycle, all rats received saline injections instead of amphetamine injections following pretreatment with 0.1 mg/kg of MK-801. A 15-min, drug-free place preference test occurred 3 days after the final conditioning trial.

Results and Discussion

MK-801 alone produced neither a place preference nor a place aversion. The mean seconds spent in the MK-801–paired chamber minus the saline-paired chamber (-20.3 ± 54.3 sec) did not significantly differ from a value of zero.

The attenuation of amphetamine place preference learning produced by MK-801 cannot be attributed to the hedonic properties of MK-801 summating with those of amphetamine because MK-801 alone produced neither a place preference nor a place aversion. In fact, other investigators have reported that when assessed under different conditions and following a greater number of training trials, MK-801 can produce a place *preference* at a similar dose (Layer, Kaddis, & Wallace, 1993; Steinpreis, Kramer, Mix, & Piwowarczyk, 1995). It is likely that, with further training, MK-801 alone would also have produced a place preference in Experiment 3. However, clearly the attenuation of amphetamine-induced place preference learning by MK-801 is not the result of the aversive hedonic properties of MK-801.

EXPERIMENT 4

The results of Experiment 2 suggest that pretreatment with MK-801 attenuated the establishment of a place preference. However, an alternative explanation based on state-dependent retrieval is also possible. During conditioning, rats experienced the amphetamine-paired chambers while in an MK-801 state; however, they were tested drug free. Both the MK-801– and the saline-pretreated groups were trained with amphetamine and tested drug free. Since the saline-pretreated groups displayed a clear preference for the amphetamine-paired side, our results show that amphetamine did not produce a state-dependent deficit. However, since MK-801 has been shown to produce state-dependent retrieval (Jackson, Koek, & Colpaert, 1992), it is conceivable that the absence of the MK-801 state during testing resulted in a state-dependent deficit in the expression of a learned preference. In fact, Carlezon, Mendrek, and Wise (1995) have recently reported that MK-801 produces state-dependent sensitization of locomotor activation by bromocriptine, although their dose of MK-801 was 2.5 times higher than that of the present investigation.

In Experiment 4, we examined the potential of state-dependent retrieval to account for the interference with place preference learning produced by MK-801. If state-dependent retrieval is responsible for the MK-801–induced interference with amphetamine place preference learning, rats both trained and tested in the MK-801 state should display a greater amphetamine place preference than should rats trained and tested under different states (MK-801 and saline).

Method

Forty-eight male Sprague–Dawley rats weighing 235–278 g on the 1st day of conditioning served as subjects. They were treated in a manner similar to that of Experiment 2 except as indicated. The rats received two conditioning trial cycles in which half of the rats were injected with MK-801 and half were injected with saline 30 min prior to the amphetamine (10 mg/kg) conditioning trial.

The place preference test occurred 3 days after the final conditioning trial. Thirty minutes prior to the test, half ($n = 12$) of the rats in each pretreatment group were injected with MK-801 (0.1 mg/kg, i.p.) and half ($n = 12$) were injected with saline.

Results

MK-801 interfered with the establishment, but not with the expression, of an amphetamine-induced place preference, and this effect was not state dependent. Figure 3 presents the mean difference scores during the place preference test of the rats pretreated with MK-801 or saline during each of the two conditioning trial cycles and tested with MK-801 or saline in Experiment 2. A 2×2 between-groups ANOVA revealed only a pretreatment condition effect [$F(1,44) = 7.4$, $p < .01$]. Regardless of the test drug, rats pretreated with saline during conditioning displayed a stronger amphetamine-induced place preference than did rats pretreated with MK-801 during conditioning. MK-801 during testing did not affect the expression of the amphetamine-induced place preference.

However, MK-801 pretreatment during conditioning did not prevent the establishment of an amphetamine-induced place preference, as is evidenced by the mean

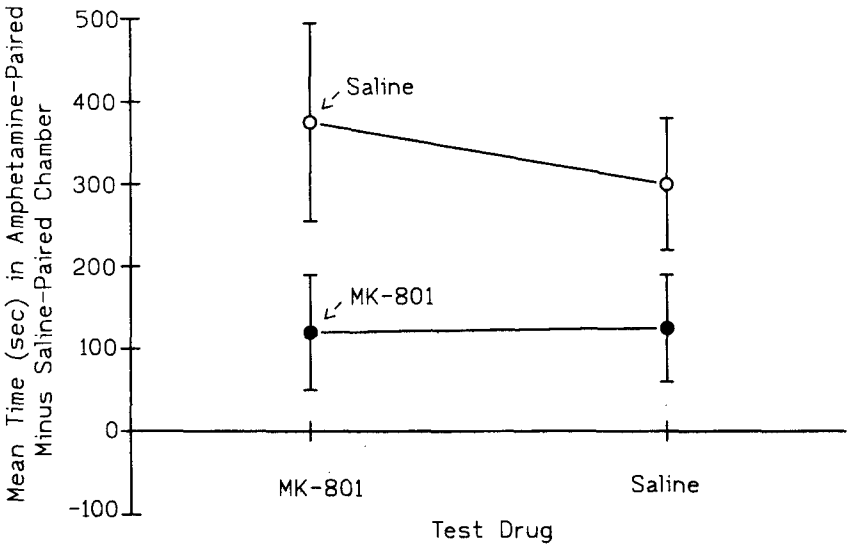

Figure 3. Mean difference scores (+*SEM*) during the place preference test for the rats pretreated with MK-801 or saline during each of the two conditioning trial cycles and tested with MK-801 or saline in Experiment 4.

difference scores of rats pretreated with MK-801 during conditioning which were greater than a value of zero, regardless of whether they were tested following injections of MK-801 or saline ($ps < .05$).

Discussion

Consistent with the results of Experiment 2, Experiment 4 demonstrated that MK-801 interfered with the establishment of amphetamine-induced place preference learning. This effect was probably not the result of state-dependent retrieval, because it was not modulated by the drug state in which the rats were tested. If interference by MK-801 was due to state-dependent retrieval, rats conditioned and tested in the MK-801 state should have displayed a greater place preference than did the rats conditioned in the MK-801 state and tested in the saline state. This pattern of results did not occur. However, although amphetamine alone did not produce a state-dependent deficit in place preference learning in the saline-pretreated group, it remained possible that the state produced by the combined MK-801 and amphetamine might have produced a state-dependent learning deficit.

In this experiment, we also assessed the ability of MK-801 to interfere with the expression of a previously established amphetamine-induced place preference. The rats that were trained in a saline state, but tested in an MK-801 state, assessed the potential of MK-801 to interfere with the display of a previously learned place preference. Clearly, MK-801 did not interfere with the expression of a place preference produced by 10 mg/kg of amphetamine, although it did attenuate the establishment of that preference. This finding is consistent with others in the literature suggesting that MK-801 interferes with the establishment, but not with the expression of learning (e.g., J. J. Kim et al., 1991; Shapiro & Caramanos, 1990; Stewart & Druhan, 1993).

EXPERIMENT 5

The attenuation of place preference learning produced by MK-801 might be the result of MK-801–induced interference with the rewarding properties of the conditioning drug or might be the result of MK-801–induced interference with learning. If MK-801 should interfere with the acquisition of drug–place associations rather than with reward, it would interfere not only with place preference learning, but also with place aversion learning. Indeed, Higgins, Nguyen, and Sellers (1992) reported that MK-801 (0.1 mg/kg) attenuated the establishment of a conditioned place aversion produced by naloxone-precipitated morphine withdrawal. In Experiment 5, we assessed the ability of MK-801 to interfere with the establishment of a conditioned place aversion produced by lithium chloride (LiCl).

Method

The subjects were 24 male Sprague–Dawley rats weighing 240–290 g on the 1st day of conditioning. The procedures of Experiment 5 were similar to those of Experiment 2, except that the conditioning drug was LiCl (75 or 127 mg/kg of a 0.15 M solution), instead of amphetamine. On each of two conditioning trial cycles, rats received saline trials on Monday and Thursday; they were injected i.p. with 1 ml/kg of saline 30 min prior to another i.p. injection of either 12 ($n = 12$) or 20 ($n = 12$) ml/kg of saline, 5 min prior to placement in the appropriate chamber for 30 min. They received the drug conditioning trial on Tuesday and Friday. On each trial, they were injected i.p. with either saline or MK-801 (0.1 mg/kg) 30 min prior to an i.p. injection of either 12 or 20 ml/kg of 0.15 M LiCl, 5 min prior to placement in the alternate chamber for 30 min. Three days after the final conditioning trial, they were given a 15-min place preference test.

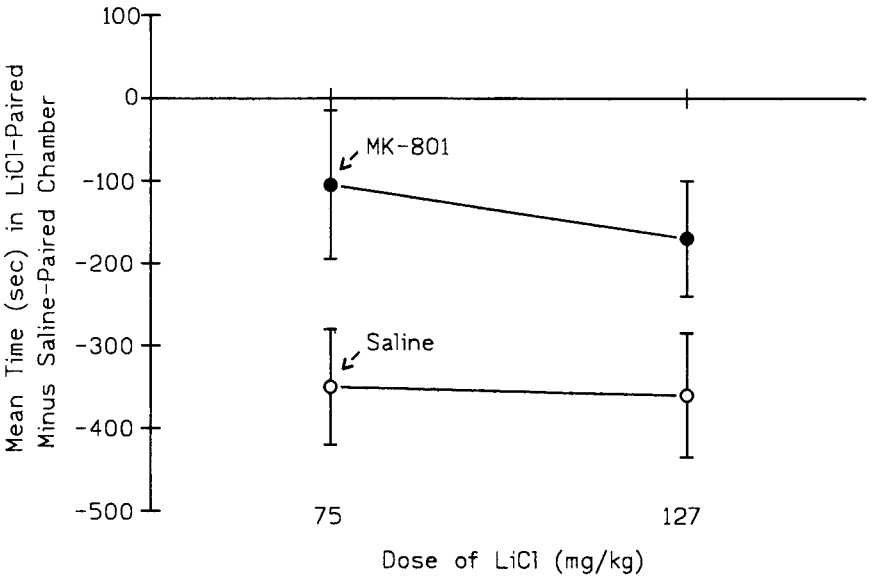

Figure 4. Mean difference scores (+*SEM*) during the preference test of Experiment 5. The groups were pretreated with MK-801 or saline during the two conditioning cycles with LiCl (75 or 127 mg/kg).

Results

MK-801 interfered with the establishment of a place aversion produced by LiCl. Figure 4 presents the mean seconds in the LiCl-paired minus the saline-paired chamber for the rats pretreated with MK-801 or saline during conditioning with either 75 or 127 mg/kg of LiCl. A 2 × 2 between-groups ANOVA revealed only a pretreatment condition effect [$F(1,20) = 4.5, p < .05$]. MK-801 interfered with, but did not eliminate, a LiCl-induced aversion; that is, the mean difference scores of the MK-801 pretreated group conditioned with 127 mg/kg of LiCl, but not 75 mg/kg of LiCl, were significantly less than zero ($ps < .05$).

Discussion

MK-801 attenuated the establishment of a place aversion produced by 75 or 127 mg/kg of LiCl. The NMDA antagonist appears to interfere with learning processes in general because it interferes with the establishment of place preference learning produced by amphetamine, morphine (Clavier et al., 1996; H. S. Kim et al., 1996; Tzschentke & Schmidt, 1995), and cocaine (Cervo & Samanin, 1995), as well as with place aversion learning produced by lithium and naloxone-precipitated morphine withdrawal (Higgins et al., 1992).

GENERAL DISCUSSION

When administered at a low dose, MK-801 interferes with the establishment of a lithium-induced conditioned place aversion and an amphetamine-induced conditioned place preference. This effect appears to be the result of interference with learning, because MK-801 did not modify the expression of a previously established amphetamine-induced conditioned place preference. Furthermore, the decrement in place preference learning was not the result of MK-801–induced state dependent learning.

Paradoxically, it appears that the efficacy of MK-801 to attenuate amphetamine place preference learning is reliant on a sufficiently strong baseline association. In the present study, MK-801 interfered with a two-trial place preference established with a dose of 10 mg/kg of amphetamine, but did not interfere with a weaker place preference established with doses of 3–5 mg/kg of amphetamine. These findings agree with those of Hoffman (1994), who also failed to find that MK-801 attenuated a two-trial place preference established with a relatively low dose of amphetamine (2 mg/kg). It should be noted that in other instances of MK-801–induced interference with morphine- or cocaine-induced place preference learning, the rats received three to four pairings of the drug with the chamber (Cervo & Samanin, 1995; Del Pozo et al., 1996; H. S. Kim et al., 1996; Tzschentke & Schmidt, 1995). Furthermore, Clavier et al. (1996) reported that MK-801 did not interfere with the acquisition of a morphine place preference after one conditioning trial, but that it did attenuate the place preference produced after the second conditioning trial (which strengthened the association).

The attenuation of amphetamine-induced place preference learning does not appear to be simply the result of MK-801–induced state-dependent retrieval. In Experiment 4, we examined the strength of a place preference in rats tested under the influence of MK-801 or saline. If the attenuation of an amphetamine-induced place preference in rats treated with MK-801 during conditioning

was the result of state-dependent retrieval (see, e.g., Carlezon et al., 1995), the preference should have been greater when the rats were tested in the same MK-801 state that they experienced during conditioning. This did not occur. The only effect that was apparent in Experiment 4 was that the animals pretreated with MK-801 during conditioning displayed a weaker amphetamine place preference than did the animals pretreated with saline during conditioning, regardless of the drug condition under which the rats were tested. Although amphetamine alone did not produce a state-dependent deficit in the saline-pretreated rats, it does remain possible that the combined MK-801 and amphetamine state distinctively produced a state-dependent deficit.

The results of Experiment 4 also indicate that MK-801 selectively interfered with the acquisition of amphetamine-induced place preference learning, but not with the expression of a previously learned place preference. This finding is consistent with others in the literature (e.g., J. J. Kim et al., 1991; Shapiro & Caramanos, 1990; Stewart & Druhan, 1993), suggesting that antagonism of NMDA receptors may selectively interfere with learning.

Although the dose of MK-801 used in the present experiments was low, there is some evidence in the literature that this dose modifies activity levels in rats (Hargreaves & Cain, 1992), although it is near the threshold for such effects. One might argue that nonspecific sensorimotor deficits produced during conditioning interfered with the ability of rats to form the association between amphetamine or lithium and the chamber. However, if such is the case, one must also explain why these sensorimotor deficits selectively affected the establishment of an association between the context and amphetamine in the groups conditioned with 10 mg/kg of amphetamine, but not in the groups conditioned with 3–5 mg/kg of amphetamine, as well as the context and lithium in Experiment 4. Furthermore, if sensorimotor deficits produced by MK-801 interfered with the ability of rats to form an association during conditioning, one might expect such deficits to also interfere with the ability of rats to display the established association when administered prior to a place preference test. However, MK-801 did not modify the expression of an amphetamine-induced place preference in Experiment 2.

MK-801 could disrupt the establishment of place preference learning produced by amphetamine, morphine (Del Pozo et al., 1996; H. S. Kim et al., 1996; Tzschentke & Schmidt, 1995), and cocaine (Cervo & Samanin, 1995) by interfering with learning or by interfering with the drug-induced reward. Because MK-801 interferes with the establishment of a place aversion produced by lithium (Experiment 3) and naloxone-precipitated morphine withdrawal (Higgins et al., 1992), it is most likely that MK-801 directly modifies the place cues–drug association. In fact, there is evidence that MK-801 actually potentiates responding maintained by cocaine (Ranaldi, French, & Roberts, 1996) and by brain stimulation reward (Corbett, 1989; Herberg & Rose, 1989), as well as the ability of both morphine (Carlezon & Wise, 1993) and cocaine (Ranaldi, Bauco, & Wise, 1996) to enhance brain stimulation reward in operant tasks. Unlike place conditioning, these paradigms assess the ability of MK-801 to maintain steady state responding of a behavior that is *already learned*. MK-801 does not appear to interfere with drug-induced reward.

In contrast, place conditioning, on the other hand, measures the ability of drugs to establish a preference or aversion to a place. Therefore, this paradigm assesses the ability of MK-801 to interfere with the acquisition of a learned preference or aversion. Because MK-801 interfered not only with amphetamine-induced place preference learning, but also with lithium-induced place aversion learning, it appears to interfere with learning processes rather than with reward.

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