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Dopaminergic basis for the facilitation of brain stimulation reward by the NMDA receptor antagonist, MK-801

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Abstract

MK-801 (dizocilpine maleate), an antagonist of the NMDA receptor, was given alone or in combination with dopamine D_1 and D_2 receptor antagonists to rats self-stimulating in lateral hypothalamus to determine whether the dopamine neurons play a role in mediating the effects of MK-801. MK-801 given at a dose of 0.1 mg/kg i.p. to self-stimulators induced a prolonged facilitation of lever-pressing, but given to non-self-stimulators, the drug had no effects. Pretreatment of self-stimulators with the dopamine D_1 receptor antagonist Schering 23390 (SCH 23390), 0.2 mg/kg given i.p. 15 min before MK-801, prevented the facilitation seen with MK-801, but did not suppress self-stimulation. SCH 23390 given alone suppressed self-stimulation. Pretreatment of self-stimulators with the dopamine D_1/D_2 receptor antagonist, haloperidol, 0.2 mg/kg given i.p. 15 min before MK-801, also prevented the facilitation of self-stimulation induced by MK-801 yet did not suppress self-stimulation. Haloperidol given alone suppressed self-stimulation. Pretreatment of self-stimulators with both SCH 23390 and haloperidol 15 min before MK-801 prevented the facilitation seen with MK-801 and suppressed self-stimulation. The combined treatment with SCH 23390 and haloperidol (without MK-801) suppressed self-stimulation, and the suppression lasted longer than the suppression seen when the two dopamine receptor antagonists were given as pretreatment, before MK-801. Pretreating self-stimulators with the combination of SCH 23390 and haloperidol 15 min before amphetamine (2 mg/kg) prevented the facilitatory response and suppressed responding for the brain reward. The suppression was of shorter duration than the suppression seen after the injection of SCH 23390 plus haloperidol. The treatment of self-stimulators with both MK-801 and amphetamine resulted in a greater and longer-lasting facilitation than the increase in responding produced by either drug alone. The similarity between the effects of MK-801 and those of amphetamine and between the effects of pretreatment with the dopamine receptor antagonists before MK-801 and before amphetamine suggests that dopaminergic activity played a significant role in the action underlying the effects of MK-801 on brain stimulation reward.

Keywords: Dopamine; Haloperidol; MK-801; SCH 23390; Self-stimulation

1. Introduction

Much interest has recently been shown in the notion of functional cooperation between the glutamate (Glu) and the dopamine projections to the striatum and related structures (Carlsson and Carlsson, 1990). The attention given to striatum is based on its involvement with specific aspects of motor function and especially with effects of psychostimulants on this in rodents (Iversen, 1977). The notion of cooperativity derives from anatomical, biochemical, and behavioral findings. The anatomical organization of the striatum shows that it receives a massive projection from the neocortex which contains Glu and is excitatory in nature (Cottman et al., 1987; Fagg, 1983; Fonnum, 1984). This organization also shows that the striatum receives a massive projection, from the substantia nigra, pars compacta, which contains dopamine and is inhibitory in nature (Beckstead et al., 1979; Lindvall and Björklund, 1974). Anatomical evidence shows that terminals from both systems converge onto the same neurons, suggesting that this convergence has functional significance (Freund et al., 1984; Smith and Bolam, 1990). Additional support for the notion of cooperativity comes from biochemical findings showing that activity in the Glu striatal projection leads to the release of dopamine in that structure, and activity in the dopamine projection to the striatum leads to a decreased local release of Glu (Araneda and Bustos, 1989; Kashihara et al., 1990; Hernandez et al., 1988; Hiramatsu et al., 1989; Löscher et al., 1991; Rao et al., 1990a,b). Electrophysiological evidence supports the notion that the

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two transmitter systems act cooperatively in striatum, in that Glu receptor antagonists that act at the NMDA receptor site can increase the firing rate of the midbrain dopamine neurons in rodents (Freeman and Bunney, 1984). The notion of cooperativity in the striatum is also supported by evidence showing that the same NMDA receptor antagonists can induce motor hyperactivity in rats (Hoffman, 1992; Liljequist et al., 1991; Tricklebank et al., 1989; Wishaw and Auer, 1989), in many respects similar to the hyperactivity induced by amphetamine and apomorphine (Iversen, 1977). Thus, while evidence is available for the notion of cooperativity between the Glu and the dopamine systems in the regulation of striatal motor function, the idea is not yet widely accepted.

The nucleus accumbens is considered a component of the striatal complex on the basis of the similarity between its anatomical and biochemical organization and that of the striatum. Although there are some differences between the two structures, these differences do not contradict the notion that, in accumbens and striatum, the Glu and the dopamine systems act cooperatively. One of the similarities between the two structures is that both receive a cortical excitatory input containing Glu as the transmitter, and both receive an inhibitory input from the mesencephalon containing dopamine as the transmitter. In nucleus accumbens, however, the cortical projection derives from the allocortex and the mesencephalic projection from the ventral tegmental area (Beckstead et al., 1979; Cottman et al., 1987; Fagg, 1983; Lindvall and Björklund, 1974; Oades and Halliday, 1987). In both structures, the terminals of the Glu and the dopamine neurons converge onto the same intrinsic neurons, and in both structures, Glu antagonists acting at the NMDA receptor site can increase the firing rate of the dopamine neurons in the midbrain that are responsible for striatal and accumbens innervation (Freeman and Bunney, 1984; Zhang et al., 1992).

Considerable evidence is available showing that n. accumbens, like striatum, is involved in motor function (Iversen, 1977). In the case of the striatum, the aspects of motor activity involved are those seen in drug-induced oral stereotypy, whereas in the case of the n. accumbens, the aspects are those seen in drug-induced locomotion (Iversen, 1977). It is not surprising, therefore, that the behavioral evidence for cooperativity between the Glu and the dopamine systems relates mainly to motor function in striatum. On the other hand, it is not unreasonable to believe that evidence relating to another function, this time in n. accumbens, should serve to gain wider acceptance for the notion of cooperativity at this brain level.

Besides being involved in the locomotor aspects of psychostimulants, the n. accumbens has also been shown to be involved in brain stimulation reward (self-stimulation). Early on, after self-stimulation was discovered, it was thought that the noradrenergic neurons of the locus coeruleus mediated the rewarding effects of brain stimulation (Olds and Olds, 1963; German and Bowden, 1974; Stein et al., 1977; Wise, 1978; Stellar and Stellar, 1985), but cumulating evidence in the past two decades has served to direct attention to the dopamine neurons of the ventral tegmental area and their projection to n. accumbens as critical for brain stimulation reward (Mogenson et al., 1979; Phillips and Fibiger, 1978; Wise and Bozarth, 1982). In view of the fact that the anatomical organization of the n. accumbens is similar to that of the striatum, it is surprising that the role of the Glu system as it relates to self-stimulation, has not been explicitly addressed.

The capacity of Glu receptor antagonists to influence self-stimulation has been investigated, but only in the context of their capacity to become drugs abused by humans. In this context, it was reported that MK-801 (dizocilpine maleate), a compound of this class, facilitates self-stimulation behavior (Corbett, 1989; Herberg and Rose, 1989). The drug is a Glu antagonist acting at the NMDA receptor to block its ion channel and, therefore, to produce a reduction of glutamatergic signal flow depending on the activation of that particular receptor (Wong et al., 1986, 1988). The capacity of MK-801 to facilitate self-stimulation might have been due to depressed Glu activity in n. accumbens or to some other action. The question of what action of MK-801 was responsible was, however, not addressed in the self-stimulation studies dealing with its potential as drug of abuse. The purpose of the present study was investigate the basis for the effects of MK-801 on self-stimulation, the assumption being that the evidence would reveal whether the Glu and the dopamine systems interact to modulate a function that is not motor in nature, yet depends on the integrity of the n. accumbens.

2. Materials and methods

2.1. Subjects

Male Sprague-Dawley rats (350–400 g at the time of electrode implantation) were individually housed in a temperature-controlled room (23°C) with lights set on a 12 : 12-h cycle (07:00–19:00 h light-dark). Food and water were available on demand.

2.2. Electrodes

Each subject was implanted with one bipolar electrode made from two stainless steel wires (250 μm) twisted together (Olds and Olds, 1963; Olds, 1979; Olds and Yuwiler, 1992). Implantation was carried out under pentobarbital anesthesia (50 mg/kg i.p.) using stereotaxic coordinates (König and Klippel, 1970) to place the probe in the medial forebrain bundle. The stereotaxic coordinates used to guide the implantation were 3.5 mm posterior to the bregma, 1.5 mm lateral to the midline, and 8.3 mm from the top of the skull. After 8–10 days of recovery from the surgery, training to self-stimulate began.

2.3. Apparatus and training

The chambers in which training to self-stimulate and drug tests were carried out were rectangular boxes made from Plexiglas, and fitted with a metal lever on the front wall (Olds, 1979). Each lever press delivered a 0.25-s train of 60-Hz sine waves to the medial forebrain bundle. Lever presses made during the delivery of a brain stimulus were ineffective. Throughout training and during drug tests, water, but not food, was available. All self-stimulation sessions started late each afternoon and finished the next morning, 13 h later. Training sessions were given each night, and self-stimulation session were also given nightly regardless of whether the session was a drug test. Autoshaping and autotraining were used to train the animals and resulted in the animals showing highly stable patterns of lever-pressing behavior over these extended sessions. The reason for sessions of such long duration was the need to gauge the full extent of the effects of treatment with a single drug or a combination of drugs.

In the first training session, the intensity of stimulation available was 50 μA. A computer-generated score showing the total number of responses made in the 13-h session was ready for the experimenter in the morning to help with the decision whether to increase the intensity of the stimulus in the second training session or to leave it unchanged. If the first training session failed to produce a response score of 5000 or more for the 13-h session, the intensity of the stimulus was raised by 20 μA in the second session. The same criterion and procedures were applied in each succeeding session to a maximum of 100 μA, following which current intensity was not further increased. All subjects received 10 training sessions with the last four at the intensity meeting the criterion for no further increase or at the maximum 100 μA intensity.

At the end of training, the scores of the last four sessions were used to classify animals as self-stimulators or non-self-stimulators. The criteria required 5000 or more lever presses to obtain brain stimulation in each of sessions 6–10 and a distributed response pattern extending from the first h to the last h of the session. If there were pauses, these had to be evenly distributed and of approximately equal duration. The animals that met the requirements were kept for the drug tests. Some of the animals which showed scores of less than 500/session in the last four training sessions were also continued with, to determine drug effects on non-self-stimulators. Sessions 9 and 10 were preceded by the i.p. injection of 0.4 ml of 0.9% saline to control for the effects of the injection procedure. The results from these sessions showed that the injections of saline did not alter the total self-stimulation score or the pattern of self-stimulation throughout these lengthy sessions. Therefore, the scores achieved in these sessions were pooled with those from sessions 7 and 8 to compute the baseline hourly self-stimulation rates and, based on these, the group data for animals that received the same drug treatment. The values in the figures show group mean and S.E.M. values. Statistical analysis was carried out using group mean and S.D. values. Some of the animals were tested more than once. In this case, new baseline mean values were computed, using the scores obtained in the daily self-stimulation sessions given between drug tests. The new baseline scores represented the hourly self-stimulation rates in the last two sessions preceding the second or third drug test. There was no evidence of bias confounding the data. Training and drug tests were carried out concurrently in 7–8 subjects, and several groups of animals, each undergoing identical training, were used in this study.

A PDP-11 34 A computer was used to collect, store, and sum lever-pressing counts for each self-stimulation session, with hard copy showing 10-min and hourly scores for each animal at the end of the session.

2.4. Histology

The effects of the various streatments were determined only for the animals in which electrodes had been confirmed to be correctly located in the medial forebrain bundle. Therefore, at the end of a series of drug tests, the animals in a group were deeply anesthetized with an overdose of pentobarbital and then transcardially perfused with saline followed by formalin. The brains were then dissected out, stored in formalin, and sliced. Electrode tracks were localized on 60-μm-thick tissue sections stained with Cresyl violet. In self-stimulators, the deepest penetration made by the electrode proved to be in the medial forebrain bundle at the level of the lateral hypothalamus. In the non-self-stimulators used to test for the motor effects of MK-801, the deepest point was outside the lateral hypothalamus, either too dorsal or too lateral.

2.5. Treatments

Vehicle or drug injection was given immediately before the self-stimulation session began. MK-801 was injected i.p. 0.1 mg/kg. This dose was chosen on the basis of evidence in the literature that it is facilitatory for self-stimulation, whereas a lower dose is without effect and a higher dose induces motor disabilities (Corbett, 1989; Herberg and Rose, 1989). Furthermore, pilot studies in our laboratory had shown that the dose of 0.25 mg/kg facilitated self-stimulation in some but not all animals, had no effect in a few animals, and depressed responding in others, thus, producing unpredictable effects for investigating effects of combined drug treatments. On the other hand, the 0.1 mg/kg dose could be counted on to produce facilitation that was stable, long-lasting and robust in most subjects. MK-801 was freshly dissolved in 0.9% saline before administration.

Haloperidol was given at a dose of 0.2 mg/kg i.p. If given as pretreatment, it was injected 15 min before the

second drug was injected, following which the self-stimulation session was started. Haloperidol was dissolved in a few drops of 0.3% tartaric acid and then brought up to volume with 0.9% saline. SCH 23390 was given at a dose of 0.2 mg/kg i.p. As a pretreatment, it was given 15 min before the second drug. It was dissolved in 0.9% saline. Amphetamine was given at a dose of 2.0 mg/kg i.p. This dose was selected on the basis of findings in a previous study from this laboratory showing that this dose is highly effective to produce facilitation of self-stimulation (Olds and Yuwiler, 1992), and reports in the literature also describing the effectiveness of this dose on self-stimulation (Wise, 1978). Amphetamine was dissolved in 0.9% saline before its administration.

2.6. Data analysis

The responses in each session were summed for each hour of the 13-h session. Group baseline scores were computed from individual scores of drug-naive animals. Group baseline scores for drug-experienced animals were computed from individual scores in the last two sessions preceding each drug treatment. Group test scores were computed from the individual scores of animals given the same drug treatment. The statistical test used to evaluate the significance of the changes in the rate of self-stimulation by different treatments was the analysis of variance (ANOVA) for repeated measures (Winer, 1962).

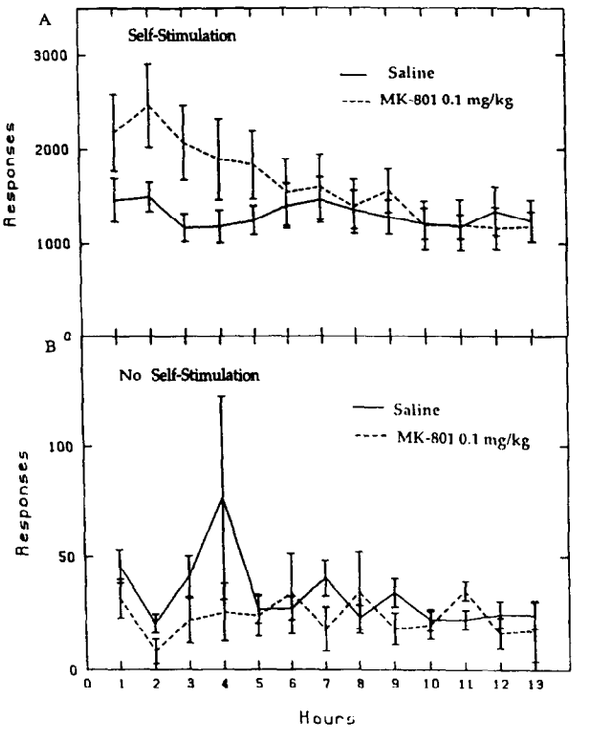

Fig. 1. Effects of MK-801, 0.1 mg/kg i.p., on the lever-pressing behavior of self-stimulators and on the operant lever-pressing behavior of non-self-stimulators. (A) Effects in self-stimulators. (B) Effects in non-self-stimulators.

3. Results

3.1. Effects of MK-801

MK-801 (0.1 mg/kg i.p.) given to self-stimulators produced a large and long-lasting facilitation of self-stimulation and did so in all subjects (Fig. 1A, $n = 5$). The response occurred early in the session, lasted several hours, and was not followed by an attenuation of lever-pressing behavior but by a return to the baseline rate. The differences between the baseline and the test self-stimulation rates show that the rates after MK-801 were signficantly higher than the rates achieved in the control sessions ($F(12,96) = 2.08$, $P < 0.02$). There was also a significant interaction between time and treatment (Fig. 1A, $F(12,96) = 2.51$, $P < 0.006$).

MK-801 given to non-self-stimulators tested concurrently with self-stimulators did not produce an increase in lever-pressing behavior in spite of the very low level at which these animals pressed the lever (operant rate) (Fig. 1B, $n = 3$). The low rate of lever-pressing in these animals was due to brain stimulation not being rewarding and to their being fully habituated to the test environment, having received the same number of sessions as the self-stimulators. In these animals, pressing was accidental, not purposeful. Far from increasing the pressing rate, MK-801 seems to have further decreased of the already low operant rate.

3.2. Effects of SCH 23390 given before MK-801

SCH 23390, injected i.p. at the dose of 0.2 mg/kg 15 min before MK-801, depressed self-stimulation, but only briefly, and then led to recovery to the baseline lever-pressing rate reached 5 h after the injection of MK-801. Recovery was brief, and was followed by a long-lasting attenuation of responding for the brain reward (Fig. 2A, $n = 3$, $F(3,12) = 3.76$, $P < 0.04$, analysis for the first 4 h of the 13-h test). The pretreatment with SCH 23390 thus prevented the facilitation seen with MK-801 given alone and led to a protracted reduction of the self-stimulation rate but, interestingly, did not abolish self-stimulation behavior.

SCH 23390 not used as pretreatment but tested alone at the dose of 0.2 mg/kg i.p., abolished self-stimulation for the first 4 h of the session. This was followed by a gradual return to the baseline rate of self-stimulation 10 h after injection of the dopamine antagonist. Statistical evaluation of the self-stimulation scores achieved with SCH 23390 and the baseline scores revealed that the difference between the two sets of scores was highly significant (Fig. 2B, $n = 3$, $F(12,48) = 2.52$, $P < 0.01$, analysis using the scores for the full 13 h of the session). There was also a

significant interaction between time and treatment (Fig. 2B, $n = 3$, $F(12,48 = 2.45$, $P < 0.01$).

These data show that MK-801 can prevent the blocking of self-stimulation by SCH 23390 and that SCH 23390 can prevent the facilitation of self-stimulation by MK-801.

3.3. Effects of haloperidol given before MK-801

The administration of haloperidol, 0.2 mg/kg i.p.. 15 min before MK-801, 0.1 mg/kg i.p., prevented the facilitation of self-stimulation seen when MK-801 was given alone, but did not suppress self-stimulation (Fig. 3A, $n = 5$). At the beginning of the session, started after the administration of MK-801, self-stimulation was briefly facilitated, then it returned to a rate of responding not significantly different from the baseline rate. The brief facilitation seen with this treatment was absent when the treatment was SCH 23390 followed by MK-801. Thus, haloperidol proved effective in blocking the robust facilitation of self-stimulation by MK-801 except for some residual action seen briefly at the beginning of the session. The scores in the drug tests did not differ significantly from the baseline scores.

Haloperidol given alone at the 0.2 mg/kg dose i.p. abolished self-stimulation for the first 3 h of the session.

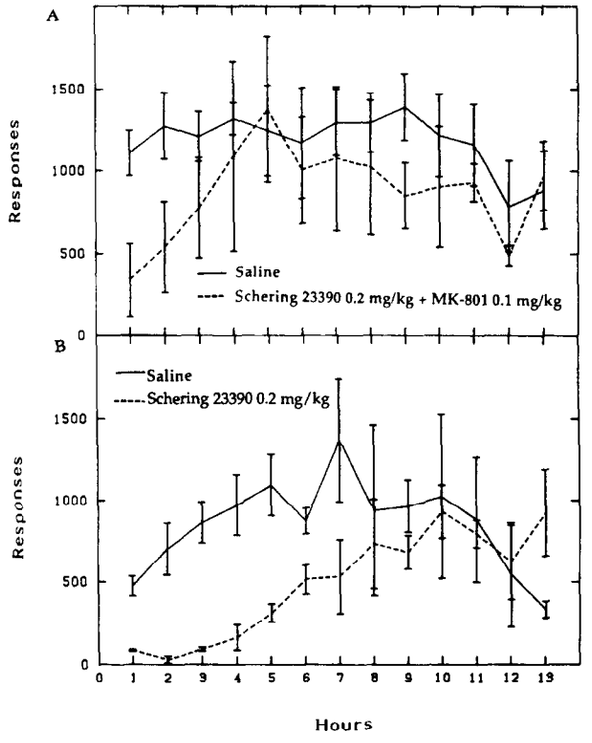

Fig. 2. Effects of SCH 23390, 0.2 mg/kg i.p., given 15 min before MK-801, 0.1 mg/kg i.p., on self-stimulation. (A) Effects of SCH 23390 as pretreatment to MK-801. The facilitation of self-stimulation seen with MK-801 given alone was blocked (cf. Fig. 1) but baseline self-stimulation was preserved. (B) Effects of SCH 23390, given alone. Self-stimulation was not preserved.

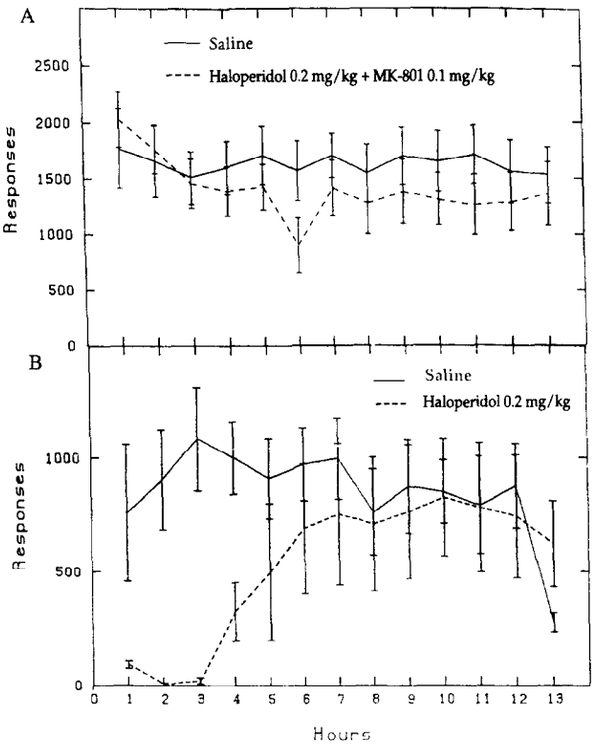

Fig. 3. Effects of haloperidol, 0.2 mg/kg i.p., given 15 min before MK-801, 0.1 mg/kg i.p., on self-stimulation. (A) Effects of haloperidol as pretreatment to MK-801. The facilitation of self-stimulation by MK-801 given alone was blocked (cf. Fig. 1) but self-stimulation was preserved. (B) Effects of haloperidol, given alone. Self-stimulation was not preserved.

Responding then gradually recovered, being again at the level of the baseline rate 10 h after injection of the dopamine antagonist (Fig. 3B, $n = 4$, $F(12,72) = 3.47$, $P < 0.0004$). The analysis also showed a significant interaction between response rate and treatment ($F(12, 72) = 5.26$, $P < 0.0001$). Haloperidol, thus, proved highly effective against self-stimulation when given alone, but was considerably less effective when given before MK-801.

3.4. Effects of MK-801 and amphetamine combined

The combined administration of MK-801, 0.1 mg/kg i.p., and amphetamine, 2 mg/kg i.p., produced facilitation of self-stimulation similar, or in some cases greater than the facilitation produced by either drug given alone (Fig. 4A, $n = 4$, $F(12,72) = 3.35$, $P < 0.0006$, test scores compared to baseline scores). The combined treatment produced a peak effect of longer duration than the peak effect produced by either drug given alone. Analysis of the data also showed a significant interaction between the self-stimulation rate over the course of the session and the treatment ($F(12,72) = 4,1$, $P < 0.001$). Thus, this test showed that the normal facilitation seen when amphetamine was given to self-stimulators was potentiated by

the coadministration of the NMDA receptor antagonist, MK-801.

Amphetamine, given alone at the 2 mg/kg i.p. dose, produced marked facilitation of self-stimulation with a peak effect seen 2 h after injection (Fig. 4B, $n = 4$, $F(12,120) = 15.75$, $P < 0.0001$). The response to amphetamine was of shorter duration, however, than the facilitatory response to the treatment that combined amphetamine and MK-801 (cf. Fig. 4A and Fig. 4B). The difference between the facilitation produced under the two conditions supports the notion that the two compounds, when given in combination, synergized to prolong their separate actions in the brain reward pathways.

3.5. Effects of SCH 23390 plus haloperidol before MK-801 and amphetamine

The pretreatment combining SCH 23390, 0.2 mg/kg i.p., and haloperidol, 0.2 mg/kg i.p., both injected 15 min before MK-801, 0.1 mg/kg i.p., resulted in the suppression of self-stimulation for 1–2 h and this was followed by a return to responding, but at rates below the baseline rates (Fig. 5A, $n = 3$, $F(1,4) = 9.18$, $P < 0.03$, analysis for the first 5 h of the session). The two dopamine receptor antagonists given together as pretreatment to MK-801, thus, not only prevented the facilitation normally seen with MK-801, but their actions synergized to block the protective action of MK-801 against each antagonist separately, resulting in lever-pressing rates below the baseline rates.

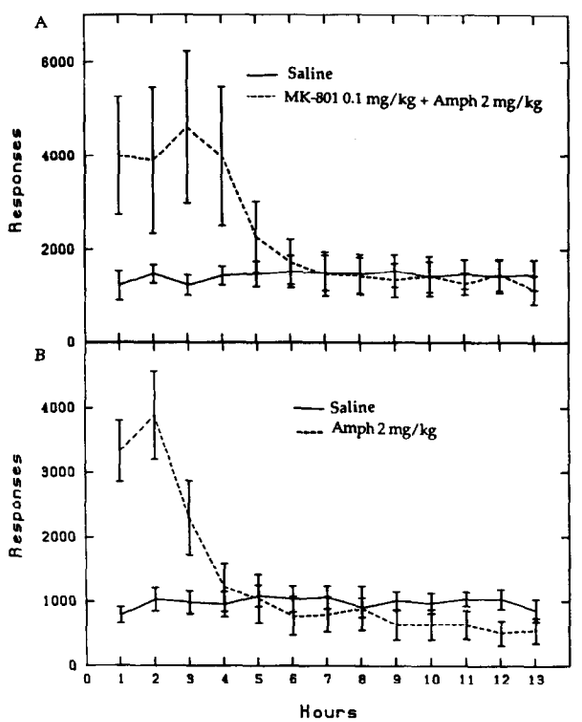

Fig. 4. Effects of treatment with both MK-801, 0.1 mg/kg i.p. and amphetamine, 2 mg/kg i.p., on self-stimulation. (A) Effects of the two compounds combined. The facilitation lasted longer than with either drug given alone (cf. Fig. 1). (B) Effects of amphetamine given alone.

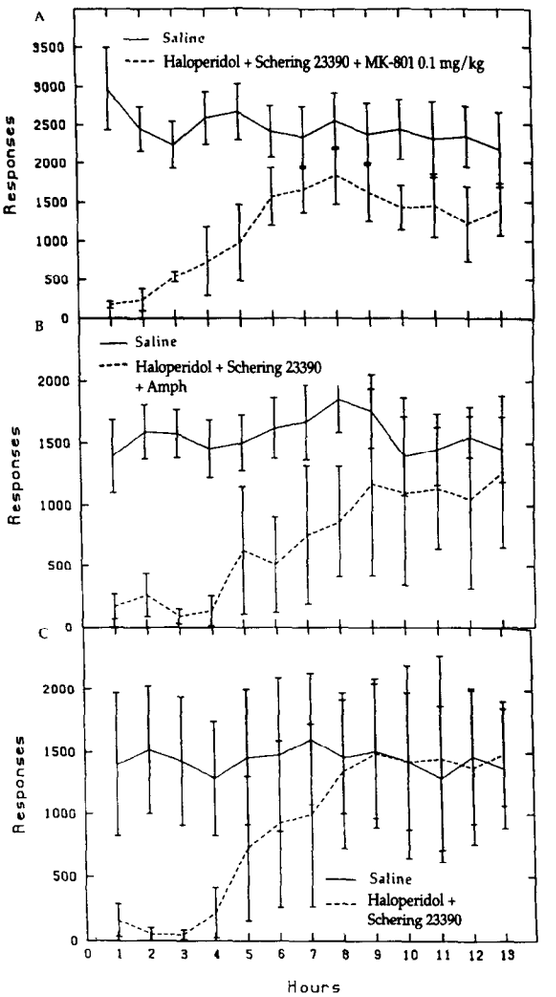

Fig. 5. Effects of pretreatment with SCH 23390, 0.2 mg/kg i.p., and haloperidol, 0.2 mg/kg i.p., given 15 min before MK-801, 0.1 mg/kg i.p. or before amphetamine, 2 mg/kg i.p., on self-stimulation. (A) Effects of the pretreatment with SCH 23390 and haloperidol given before MK-801. The facilitation seen with MK-801 given alone was blocked and self-stimulation was suppressed. (B) Effects of the pretreatment with SCH 23390 and haloperidol given before amphetamine. The facilitation normally seen with amphetamine was blocked and self-stimulation was suppressed. (C) Effects of SCH 23390 and haloperidol combined on self-stimulation. Self-stimulation was blocked for a longer time than when the two dopamine antagonists were given as the combined pretreatment to MK-801 or amphetamine.

The same pretreatment given before the administration of amphetamine, 2 mg/kg i.p. produced the same results. Again, the facilitation of self-stimulation by amphetamine was blocked and self-stimulation itself was suppressed for a time (Fig. 5B, $n = 3$, $F(1,4) = 7.58$, $P < 0.05$, analysis for the first 5 h of the session).

Tests carried out to determine the effects of giving both SCH 23390 and haloperidol, without MK-801 or amphetamine, showed that they suppressed self-stimulation

for the first 3 h after their administration, and that the recovery to the baseline self-stimulation rate was achieved only 9 h after the injection (Fig. 5C, $n = 3$, $F(12,48) = 3.17$, $P < 0.002$). Analysis of the results also revealed that there was a significant interaction between the self-stimulation rate and time of treatment (Fig. 5C, $n = 3$, $F(12,48) = 3.54$, $P < 0.0008$). These results show that neither MK-801 nor amphetamine had effects on self-stimulation powerful enough to overcome the combined actions of the two dopamine antagonists used in this study.

4. Discussion

The results showed that MK-801 can facilitate self-stimulation when given at a low dose and that this effect is robust, long-lasting and not different in important respects from the facilitation of self-stimulation produced by amphetamine. The results further showed that MK-801 does not facilitate the lever-pressing behavior of non-self-stimulators. In addition, the data showed that the dopamine receptor antagonists, SCH 23390 and haloperidol, can block the facilitation of self-stimulation by MK-801 in a manner similar to their ability to block the facilitation of the same behavior by amphetamine. In both situations, facilitation was blocked but not self-stimulation itself, showing that MK-801 and amphetamine provide equal protection against the negative effects of the two dopamine receptor antagonists. Finally, the findings obtained with the combined treatment with MK-801 and amphetamine showed that the facilitation of self-stimulation produced under these conditions was greater than the facilitation produced by either drug alone, suggesting their ability to synergize in this context. These findings, thus, emphasize the similarity between the actions of MK-801 and amphetamine and between their capacity to exert reciprocal regulation of their actions on self-stimulation. Based on these results, it is concluded that the functional cooperativity between the glutamate and the dopamine systems presumed to exist in striatum for the motor function may well extend to n. accumbens for the reward function.

The facilitatory effects of MK-801 on self-stimulation reported here confirm similar findings reported earlier (Corbett, 1989; Herberg and Rose, 1989). The present data, however, extend the earlier observations by showing the long duration of this effect, the pattern of self-stimulation that followed the facilitation, the effects of pretreatment with the dopamine receptor antagonists SCH 23390 and haloperidol, and the interaction between MK-801 and amphetamine to effect reciprocal modulation of self-stimulation.

The results showing that MK-801 did not facilitate the lever-pressing behavior in non-self-stimulators indicate that the facilitation was specific to brain stimulation reward and was not the consequence of gross behavioral activation, a finding confirming the view expressed in the earlier reports of the effects of MK-801 on self-stimulation (Corbett, 1989; Herberg and Rose, 1989). The problem of distinguishing motor effects from the rewarding effects of a compound on self-stimulation is particularly important when the treatment depresses responding because the effect might be due to some pathological condition induced by the compound. However, when the effect of the treatment on self-stimulation is facilitatory, leading to consistent and sustained lever-pressing behavior extending over several hours, the effect is unlikely to be due to an increase in gross motor activity, leading to such sustained, purposeful behavior. Furthermore, it is well known that a drug-induced increase in gross motor behavior disrupts self-stimulation, as noted in tests with high doses of amphetamine or apomorphine (Wise, 1978; Wise and Bozarth, 1982). These considerations support the conclusion that the failure of MK-801 to facilitate responding in non-self-stimulators can be taken to mean that the drug acted in the brain stimulation reward pathway when it induced facilitation, although this does not preclude its ability to also act in motor pathways in other situations.

The results of the tests in which the dopamine receptor antagonists were given as pretreatment revealed that SCH 23390 could block the facilitation of self-stimulation by MK-801. After this treatment, self-stimulation throughout the session was attenuated compared to the baseline rate. The failure of SCH 23390 to abolish self-stimulation itself under these conditions was in sharp contrast with the suppression of this behavior when SCH 23390 was given alone. This difference in the effects seen with SCH 23390 + MK-801 and with SCH 23390 given alone, shows that MK-801 had the capacity to protect self-stimulation but no longer the capacity to facilitate it. These findings support two conclusions. The first is that the D_1 dopamine receptor plays a role in self-stimulation since its blockade by SCH 23390 led to the suppression of self-stimulation. The second is that an action of MK-801 on the dopamine neurons involved in brain stimulation reward, no matter how indirect, counteracted the blockade of the D_1 dopamine receptor, resulting in self-stimulation being maintained instead of being suppressed.

The results obtained when haloperidol was the pretreatment before MK-801 underscore the important role played by the D_2/D_1 dopamine receptors in the brain reward function. Haloperidol, given alone, suppressed self-stimulation for several hours, but when given before MK-801, it failed to suppress self-stimulation although the facilitatory aspect of the response was now absent. These results show that MK-801 can protect the brain reward function against haloperidol. Here, as when SCH 23390 was the pretreatment, MK-801 blunted the negative action of haloperidol.

However, the data obtained when SCH 23390 and haloperidol were both given before MK-801 imply that the two types of dopamine receptors are necessary for self-stimulation. The evidence supporting this conclusion is

that, when both dopamine receptor antagonists were used as pretreatment, MK-801 was ineffective in inducing facilitation of self-stimulation and also was ineffective in providing the action required for the continued occurrence of self-stimulation challenged by the combination of SCH 23390 and haloperidol. These findings suggest that one of these two receptors has to be available for activation of self-stimulation and, that under normal conditions, the two types act cooperatively, a conclusion also reached with respect to striatal motor function (Arnt and Hytell, 1984, 1986; Molloy and Waddington, 1985; Waddington et al., 1986).

Finally, the synergism between MK-801 and amphetamine suggests that the two compounds act to enhance brain stimulation reward by different actions. For amphetamine, the action is relatively clear. It action prompted the release of dopamine in an environment in which the transmitter was also released with each application of the stimulus in the medial forebrain bundle (Iversen, 1977; Wise, 1978; Wise and Bozarth, 1982). For MK-801, several actions may have been responsible and which in fact was responsible is unclear.

One action may have been the capacity to increase the release of dopamine from terminals in n. accumbens as MK-801 was shown to be capable of doing in striatum (Hernandez et al., 1988; Kashihara et al., 1990; Liljequist et al., 1991). In the situation where MK-801 was given alone, the amount of dopamine released by stimulation of the medial forebrain bundle (Iversen, 1977) and released by MK-801 would have led to greatly enhanced dopaminergic activity. However, MK-801 has not been shown to have a greater capacity than amphetamine for inducing the release of dopamine yet the facilitation by MK-801 was as robust as that seen with amphetamine and the peak effect lasted much longer. Furthermore, when MK-801 and amphetamine were given together, enough dopamine must have been available to be released by MK-801 over and above the release due to amphetamine to stimulate the medial forebrain bundle. Such a situation seems improbable because of the many hours during which the facilitation of self-stimulation lasted, suggesting that the synergism between MK-801 and amphetamine may have been due to some action of MK-801 other than promotion of the release of dopamine. These considerations argue against the notion of release of dopamine stimulated by MK-801 as the principal reason why the facilitation of self-stimulation by this compound lasted longer than the facilitation by amphetamine.

MK-801 has, however, been reported to have several actions. Thus, it may also have activated the phencyclidine site on the NMDA receptor (Deutsch et al., 1987; Loo et al., 1987; Rao et al., 1990a,b), with as consequence the facilitation of self-stimulation. The capacity of MK-801 to activate the phencyclidine site has been shown to lead to the release of dopamine, and may be the reason for the similarity between the effects of amphetamine and non-competitive NMDA receptor antagonists on dopamine-dependent behaviors. But the arguments raised earlier against the notion of release of dopamine from terminals as the basis for the effects of MK-801 on self-stimulation also apply here. Not enough dopamine is likely to have been available for release by brain stimulation and MK-801 to sustain a facilitation that lasted longer than the facilitation of self-stimulation produced by amphetamine, or to account for the synergism between amphetamine and MK-801 on self-stimulation.

Still another action of MK-801 may have been responsible, at least in part, for its striking effect on the reward function. This action is its capacity to block the ion channel of the NMDA receptor (Wong et al., 1986, 1988), with as consequence a reduced excitatory input to the forebrain structures receiving dopaminergic innervation from the midbrain dopamine neurons, including the n. acccumbens, the structure considered a critical node in the circuitry responsible for brain stimulation reward. A reduced excitatory input to the n. accumbens would have the consequence of reducing the inhibitory regulation exerted by n. accumbens on the dopamine neurons of the ventral tegmental area, thus disinhibiting the dopamine neurons (Cottman et al., 1987; Fagg, 1983; Johnson and North, 1992; Oades and Halliday, 1987). Thus, the capacity of MK-801 to block the ion channel of the NMDA receptor, leading to disinhibition of the ventral midbrain dopamine neurons, has been viewed as the basis for the increase in the firing rate of the dopamine neurons in the ventral midbrain seen after MK-801 (Tanaka and North, 1993; Freeman and Bunney, 1984; Zhang et al., 1992). It seems likely that the same action of MK-801 was responsible for its effects on self-stimulation and for its capacity to counteract the negative effect of SCH 23390 and haloperidol. The reason for this conclusion is that disinhibition of the dopamine neurons may last longer than the release of dopamine due to drug action, and this disinhibition may account for enhanced synthetic activity, and prolongation of the capacity to sustain the dopamine-dependent behavior. Under these conditions, the facilitation by MK-801 of self-stimulation would last longer than the facilitation produced by amphetamine that was observed. Self-stimulation would, thus, continue, in spite of the challenge by SCH 23390 and haloperidol, as was also observed. Also, under such conditions of disinhibition of the relevant dopamine neurons, the conditions would exist for MK-801 and amphetamine to synergize.

In conclusion, the results support the notion of cooperativity of the dopamine and Glu neurons in the regulation of excitability at the reward sites and, thus, in the determination of self-stimulation rates. The other actions of MK-801 related to dopaminergic activity may have contributed to the final result, but for the reasons given earlier, disinhibition of the midbrain dopamine neurons seems to have been the principal factor determining the effects of MK-801 in the present context.

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