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Modulation of the locomotor activating effects of the noncompetitive NMDA receptor antagonist MK801 by dopamine D2/3 receptor agonists in mice

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Abstract

The noncompetitive NMDA receptor antagonist MK801 (dizocilpine) produces behavioral stimulation mediated, in part, through indirect activation of the dopamine (DA) system. Previous reports indicate that D2/3 agonists inhibit MK801-induced stereotypies; however, it is unclear if these agonists also attenuate MK801-induced locomotion. As such, the ability of the D2/3 agonists, quinelorane and quinpirole, and the partial D3 agonist, BP897, to attenuate the locomotor activating effects of MK801 was examined in mice. MK801 (0.1–1.0 mg/kg) produced a biphasic effect on total distance traveled with the intermediate dose of 0.3 mg/kg producing the greatest stimulation. The increase in MK801-induced total distance traveled was attenuated by the coadministration of quinelorane and quinpirole at doses that alone had no effect on activity. Similarly, the partial D3 agonist, BP897, blocked the effects of MK801. The D3-preferring antagonist, nafadotride, reversed the attenuation of quinelorane and partially reversed the attenuation of quinpirole. The D2-preferring antagonist, eticlopride, reversed the attenuating effects of quinelorane, but was not effective against quinpirole. Nafadotride and eticlopride were ineffective against the attenuating effects of BP897 on MK801-induced locomotion. Because BP897 is a partial agonist it was tested against quinelorane/MK801 and quinpirole/MK801 combinations. BP897 reversed the attenuating effects of quinelorane, but not those of quinpirole on MK801's effects. These results demonstrate that the DA system, through D2/3 receptor activation, modulates the locomotor activating effects produced by noncompetitive NMDA receptor blockade.

Keywords: MK801; D2/3 agonist; D2/3 antagonist; BP897; Nafadotride; Locomotor; Psychomotor; Mice

1. Introduction

There is considerable evidence that several neurotransmitter systems can indirectly modulate dopaminergic (DAergic) activity. Drugs such as mu opioid agonists, nicotine and Δ^9-THC, acting through their respective neurotransmitter systems, increase DA release in various brain regions (Di Chiara, 2000; Di Chiara et al., 1996; Di Chiara and Imperato, 1988; Navarro et al., 1993). The glutamate system also has been shown to be involved in regulating activity in the DA system. MK801 (dizocilpine), a noncompetitive antagonist at the NMDA receptor (Lodge and Johnson, 1990), indirectly stimulates DA release and turnover in the brain (Marcus et al., 2001; Rao et al., 1990), and these increases in DA are thought to mediate the conditioned place preference effects of MK801 (Suzuki et al., 2000) as well as the complex behavioral syndrome characterized by increased locomotion, stereotypy and decreased motor coordination following MK801 administration (e.g., Irifune et al., 1995; Verma and Kulkarni, 1992).

This complex behavioral syndrome that results from noncompetitive NMDA receptor antagonist administration has been proposed to represent a model of schizophrenic symptomatology (e.g., Javitt and Zukin, 1991), as there is pathophysiological evidence suggesting that deficits in glutamatergic neurotransmission may be a contributing factor (Carlsson, 1988; Marino and Conn, 2002). Thus, much work has focused on the interaction between the glutamatergic and DAergic systems. For example, MK801-induced locomotor activity is decreased in mice treated with reserpine, a drug that depletes monoamine stores (Narayanan et al., 1996; Starr and Starr, 1994), although this is not always observed (Carlsson and Carlsson, 1989). Moreover, Ouagazzal et al. (1993) demonstrated that MK801-induced hyperlocomotion was re-

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duced in rats by the D1 antagonist SCH23390 and the D2/3 antagonist raclopride, although high doses of raclopride that blocked locomotor activity also produced catalepsy. Similarly, Hoffman (1992) found that the D2/3 antagonists, haloperidol and eticlopride, attenuated MK801-induced locomotion in rats, although these antagonists were not tested alone and previous research indicates that both haloperidol and eticlopride alone at high doses can inhibit motor activity (e.g., Chausmer and Katz, 2001).

In addition to DA antagonists reducing DAergic activity there is evidence that, under some conditions, D2/3 agonists can suppress DAergic activity through inhibition of DA release (Dekeyne et al., 2001; Gainetdinov et al., 1996; Gilbert et al., 1995; Gobert et al., 1995). D2/3 agonists inhibit DAergic activity either through presynaptic mechanisms (Aretha et al., 1995; Gainetdinov et al., 1996; Gobert et al., 1995; Sotnikova et al., 2001) or a postsynaptic negative feedback pathway (Gobert et al., 1995; Koeltzow et al., 1998). As such, it is possible that D2/3 receptor stimulation may suppress the behavioral stimulant effects of MK801 if these effects are mediated through DAergic activity. Indeed, the D2/3 agonist, B-HT920, as well as PD128907, an agonist with selectivity for D3 over D2 receptors ranging from six- to several hundredfold (Levant, 1997), blocked MK801-induced sterotypies in mice (Verma and Kulkarni, 1992; Witkin et al., 1998).

Given the ability of D2/3 agonists to attenuate MK801-induced stereotypies, and the potential involvement of the DAergic system in mediating the effects of MK801, the present study was undertaken to examine the ability of the D2/3 agonists quinelorane and quinpirole to block the locomotor activating effects of MK801 in Swiss Webster mice. Because the D2/3 agonists attenuated MK801-induced locomotion, the ability of the D3-preferring antagonist, nafadotride (Audinot et al., 1998; Sautel et al., 1995b) and the D2-preferring antagonist, eticlopride (Levant, 1997), to reverse the attenuating effects of quinelorane and quinpirole on MK801-induced locomotion was assessed. Finally, BP897, a selective partial D3 receptor agonist (Pilla et al., 1999) was tested for its ability to not only attenuate the locomotor effects of MK801 but also for its ability to antagonize the attenuating effects of quinelorane and quinpirole.

2. Materials and methods

2.1. Subjects

Adult male Swiss Webster mice (Harlan Sprague Dawley, Indianapolis, IN) weighing 20–35 g were used. Mice were housed five per cage, had continuous access to food and water, and were allowed to acclimate to the vivarium environment one week prior to the start of any testing. The vivarium was temperature controlled (22–24 °C) and on a 12-h light–dark cycle. All testing occurred during the light component. Animals used in this study were cared for in accordance with the guidelines of the Institutional Animal Care and Use Committee of Virginia Commonwealth University, and the *Guide for the Care and Use of Laboratory Animals* (Institute of Laboratory Animal Resources, National Academy Press, 1996).

2.2. Locomotor procedure and analyses

Each mouse was tested with only one dose or one dose combination. Four commercially obtained, automated activity monitoring devices each enclosed in sound- and light-attenuating chambers were used (AccuScan Instruments, Columbus, OH). The interior of each device was divided into two separate $20 \times 20 \times 30$-cm arenas permitting the independent and simultaneous measurement of two mice. Sixteen photobeam sensors were spaced 2.5 cm apart along the walls of the chamber. On each test day, mice were randomly selected from the available stock in the vivarium and brought to the laboratory where they were allowed to acclimate for approximately 30 min. Nonhabituated mice were injected with vehicle or test compound(s) and, following various pretreatment intervals, placed into the test chambers where their activity was recorded a total of 60 min. Total distance traveled (cm) during the experimental session was recorded for each mouse and was summed across the 60-min session. Immediately following the test session each mouse was returned to its homecage and was briefly observed for signs of ataxia. A one-factor (dose) ANOVA was conducted followed by Dunnett's post hoc tests. The effects of MK801 alone and the DA agonists and antagonists alone were compared to their respective vehicle conditions using a two-tailed Dunnett's post hoc test to determine if the agonists or antagonists altered psychomotor activity relative to vehicle levels. Because MK801 produced biphasic effects on total distance traveled, the intermediate dose (0.3 mg/kg) was chosen for all drug combinations as it produced maximal psychomotor stimulant effects. The effects of 0.3 mg/kg MK801 in combination with either a DA agonist or a DA antagonist were compared to the effects of MK801 alone (0.3 mg/kg) using a two-tailed Dunnett's post hoc test. Two post hoc analyses were utilized to determine if a DA antagonist reversed the attenuation of MK801-induced effects produced by a DA agonist. Both post hoc comparisons were conducted so as to minimize false positive and false negative statistical outcomes. First, the effects of 0.3 mg/kg MK801 plus water and the effects of 0.3 mg/kg MK801 plus a DA agonist and a DA antagonist were compared to the effects of 0.3 mg/kg MK801 plus water using a two-tailed Dunnett's post hoc test. When the effects of MK801 plus a DA agonist and a DA antagonist were not significantly different than MK801 plus water it was deemed that the DA antagonist effectively reversed

the attenuation of the DA agonist on MK801-induced behaviors. Second, the effects of 0.3 mg/kg MK801 plus water and the effects of 0.3 mg/kg MK801 plus a DA agonist and a DA antagonist were compared to the effects of 0.3 mg/kg MK801 plus a DA agonist using a one-tailed Dunnett's post hoc test. When the effects of MK801 plus a DA agonist were significantly different compared to MK801 plus a DA agonist and a DA antagonist it was deemed that the DA antagonist effectively reversed the attenuation of the DA agonist on MK801-induced locomotion. Therefore, a DA antagonist was considered to have completely reversed the attenuating effects of a DA agonist on MK801-induced locomotion when (1) the effects of MK801 plus a DA agonist and DA antagonist were not significantly different than the effects of MK801 plus water based on a Dunnett's post hoc test and (2) when the effects of MK801 plus a DA agonist were significantly different than the effects of MK801 plus a DA agonist and a DA antagonist based on a Dunnett's post hoc test. A DA antagonist was considered to have produced a partial reversal when only one of the two post hoc conditions was satisfied. The alpha level for all comparisons was set at .05.

2.3. Drugs

(5*R*,10*S*)-(+)-5-Methyl-10,11-dihydro-5*H*-dibenzo[*a*,*d*]-]cyclohepten-5,10-imine hydrogen maleate [(+)-MK801 hydrogen maleate], quinelorane dihydrochloride, (−)-quinpirole hydrochloride, and (−)-eticlopride hydrochloride were purchased from Sigma-Aldrich (St. Louis, MO). Nafadotride and BP897 were generously supplied by Pierre Sokoloff (Unite de Neurobiologie et Pharmacologie Moleculaire, INSERM, Paris, France). (+)-MK801, quinelorane, (−)-quinpirole and eticlopride were dissolved in sterile water, and nafadotride was dissolved in sterile 0.09% saline. BP897 was first dissolved in 20% (wt/vol) hydroxypropyl-β-cyclodextrin (ENCAPSIN HPB, American Maize Products, Hammond, IN) in sterile water, at a maximal concentration of 5 mg/ml, to form a stock solution. Appropriate amounts of stock solution were added to sterile water to form test doses. All drugs were administered intraperitoneally, except for BP897, which was administered subcutaneously. Mice were injected with MK801 or vehicle and immediately placed into the test chambers. Mice tested with quinelorane, quinpirole and BP897 alone or in combination with MK801 were placed into the test chambers immediately following administration. During antagonism tests mice were injected with eticlopride, nafadotride or BP897 and returned to their homecage for 30 min, after which they were injected with a drug or drug combination and placed immediately into the test chambers. Drug routes of administration and pretreatment intervals were chosen based upon published (Cook and Beardsley, 2003) and unpublished observations from our laboratory.

3. Results

3.1. Effects of MK801 alone

Fig. 1 shows the effects of MK801 alone on total distance traveled when the data were summed across the entire 60-min session. A main effect was observed for dose on total distance traveled [$F(3,28) = 8.54$, $P < .05$] (Fig. 1) such that a dose of 0.3 mg/kg MK801 significantly increased activity. Following the testing of 1.0 mg/kg MK801 all mice exhibited signs of ataxia (characterized by a staggering, slow gait) upon return to their homecage.

3.2. Effects of quinelorane and quinpirole alone and in combination with MK801

Fig. 2 shows the effects of quinelorane and quinpirole alone when the data were summed across the entire 60-min session. Quinelorane [$F(4,35) = 2.39$, $P \geq .05$] and quinpirole [$F(3,28) = 1.94$, $P \geq .05$] failed to alter total distance traveled (Fig. 2A and B). Fig. 2A also shows the effects of quinelorane in combination with 0.3 mg/kg MK801. When quinelorane was combined with MK801 there was a main effect of dose for total distance traveled [$F(4,35) = 5.92$, $P < .05$] such that doses of 0.003 and 0.01 mg/kg quinelorane significantly reduced the locomotor effects of the 0.3 mg/kg dose of MK801. The combination of quinpirole and MK801 resulted in a main effect for total distance traveled [$F(3,28) = 4.63$, $P < .05$] (Figure 2B) such that a dose of 0.1 mg/kg quinpirole reduced the locomotor effects of MK801.

3.3. Effects of nafadotride and eticlopride pretreatment on the effects of vehicle or MK801 combinations

Fig. 3 shows the effects of nafadotride and eticlopride in combination with vehicle or MK801. When nafadotride was

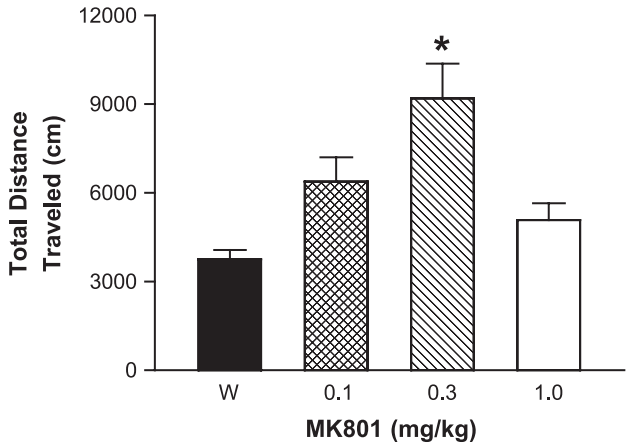

Fig. 1. Effects of MK801 on total distance traveled in mice ($n = 8$). Mice were injected intraperitoneally with water (W) or MK801 and placed immediately into the test chambers. Data depicted are the summed effects across the 60-min session. * Significant difference compared to water alone. Brackets indicate S.E.M.

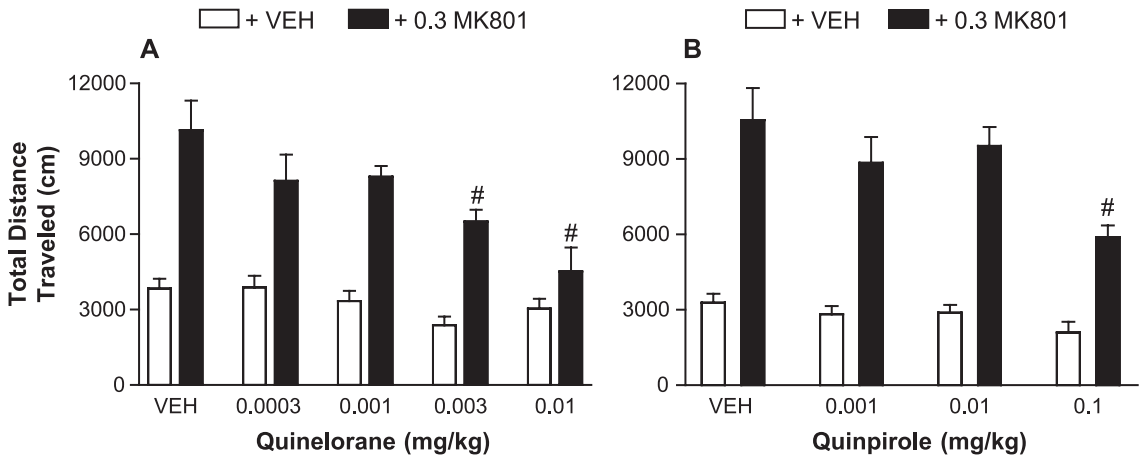

Fig. 2. Effects of quinelorane (panel A) and quinpirole (panel B) in combination with VEH (vehicle) or in combination with 0.3 mg/kg MK801 on total distance traveled in mice ($n=8$). Mice were injected intraperitoneally with vehicle in combination with quinelorane, quinpirole or vehicle as well as in combination with MK801 (0.3 mg/kg), and then were placed immediately into the test chambers. #Significant difference compared to 0.3 mg/kg MK801 alone. Other details as in Fig. 1.

administered as a pretreatment to vehicle there was no effect on total distance traveled [$F(3,28)=2.11$, $P\geq.05$] (Fig. 3A). Nafadotride pretreatment failed to alter MK801-induced (0.3 mg/kg) total distance traveled [$F(3,28)=0.52$, $P\geq.05$] (Fig. 3A). There was a main effect for eticlopride in combination with vehicle for total distance traveled [$F(4,35)=4.61$, $P<.05$] (Fig. 3B) such that a dose of 0.1 mg/kg eticlopride significantly reduced activity. Eticlopride pretreatment at a dose of 0.1 mg/kg significantly reduced MK801-induced total distance traveled [$F(4,35)=7.79$, $P<.05$] (Fig. 3B).

3.4. Effects of nafadotride pretreatment on the effects of MK801 in combination with quinelorane or quinpirole

Fig. 4 shows the effects of nafadotride on the locomotor effects of 0.3 mg/kg MK801 in combination with 0.003 mg/kg quinelorane or 0.1 mg/kg quinpirole. There was a main effect of drug treatment on total distance traveled [$F(5,42)=4.04$, $P<.05$] (Fig. 4A). Nafadotride reversed the attenuating effects of 0.003 mg/kg quinelorane on total distance traveled with maximal effects being obtained at a dose of 3.0 mg/kg. Nafadotride partially reversed the attenuating effects of 0.1 mg/kg quinpirole on MK801-induced total distance traveled [$F(5,42)=7.20$, $P<.05$] (Fig. 4B).

3.5. Effects of eticlopride pretreatment on the effects of MK801 in combination with quinelorane or quinpirole

Fig. 5A shows the effects of eticlopride on the stimulant effects of 0.3 mg/kg MK801 in combination with 0.003 mg/kg quinelorane and 0.1 mg/kg quinpirole. There was a main effect of drug treatment on total

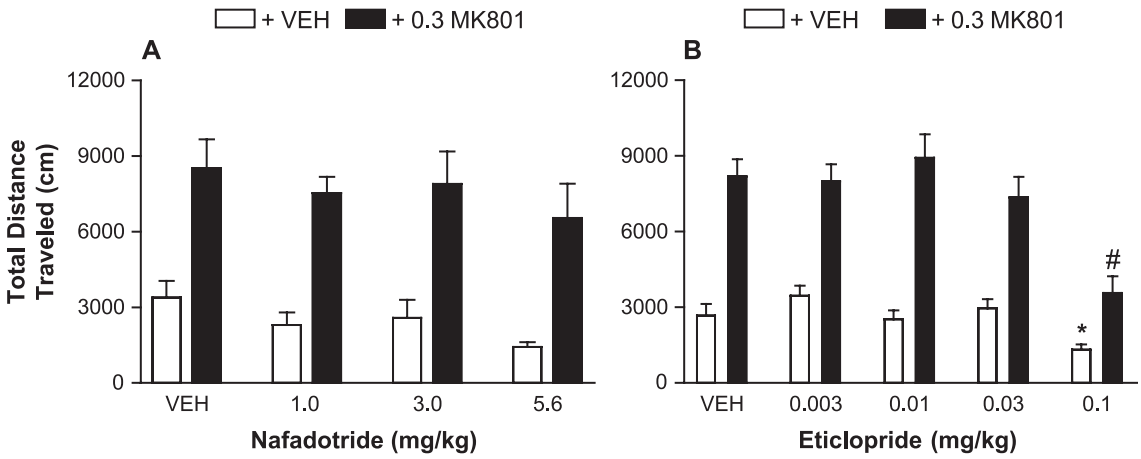

Fig. 3. Effects of nafadotride (panel A) and eticlopride (panel B) in combination with VEH (vehicle) or in combination with 0.3 mg/kg MK801 on total distance traveled in mice ($n=8$). Mice were injected intraperitoneally with vehicle in combination with nafadotride, eticlopride or vehicle as well as in combination with MK801 (0.3 mg/kg), and then were placed immediately into the test chambers. * Significant difference compared to vehicle alone. #Significant difference compared to 0.3 mg/kg MK801 alone. Other details as in Fig. 1.

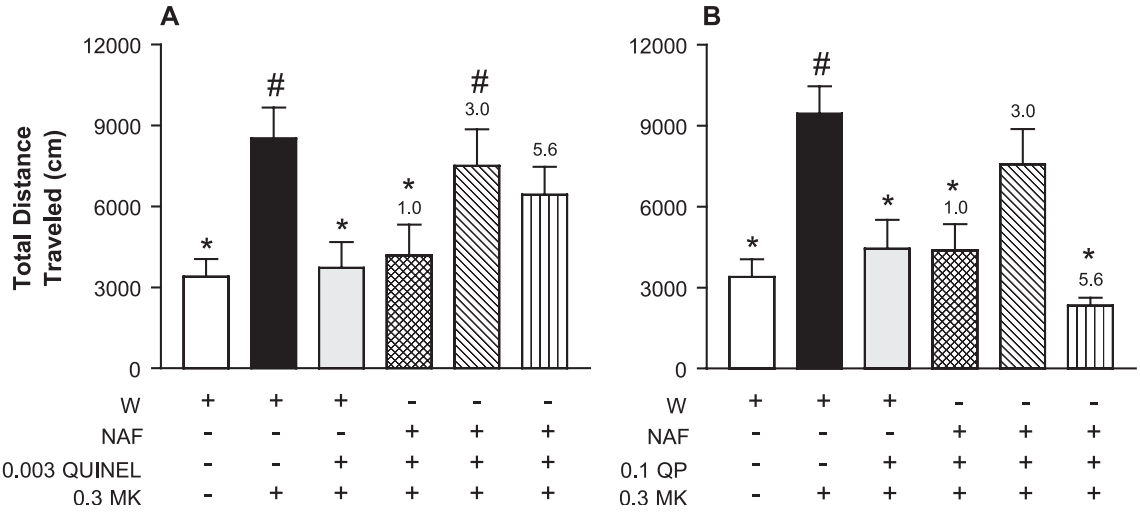

Fig. 4. Effects of nafadotride (NAF) and water (W) pretreatment (30 min) on 0.003 mg/kg quinelorane (QUINEL) plus 0.3 mg/kg MK801 combinations (panel A) or 0.1 mg/kg quinpirole (QP) plus 0.3 mg/kg MK801 combinations (panel B) on total distance traveled in mice ($n=8$). Mice were injected intraperitoneally with nafadotride or water and returned to their homecages for 30 min, and then were injected with the drug combinations and placed immediately into the test chambers. Numbers above the bars indicate doses (mg/kg) of nafadotride administered. The "+" indicates the drug was administered, whereas the "–" indicates the drug was not administered. * Significant difference compared to 0.3 mg/kg MK801 plus water. #Significant difference compared to 0.3 mg/kg MK801 plus quinelorane (or quinpirole). Other details as in Fig. 1.

distance traveled [$F(6,49)=11.54$, $P<.05$] (Fig. 5A). A dose of 0.03 mg/kg eticlopride reversed the attenuating effects of quinelorane on MK801-induced total distance traveled, whereas 0.1 mg/kg eticlopride enhanced the attenuating effects of 0.003 mg/kg quinelorane. There was a main effect of drug treatment for eticlopride pretreatment on 0.1 mg/kg quinpirole/MK801 combinations for total distance traveled [$F(6,49)=5.92$, $P<.05$] (Fig. 5B); however, post hoc tests failed to reveal that the attenuating effects of quinpirole on MK801-induced total distance traveled were reversed by eticlopride.

3.6. Effects of BP897 alone and in combination with MK801

Fig. 6 shows that when the data were summed across the 60-min session BP897 decreased total distance traveled [$F(4,35)=9.69$, $P<.05$] such that a dose of 10 mg/kg BP897 significantly decreased activity. When BP897 was

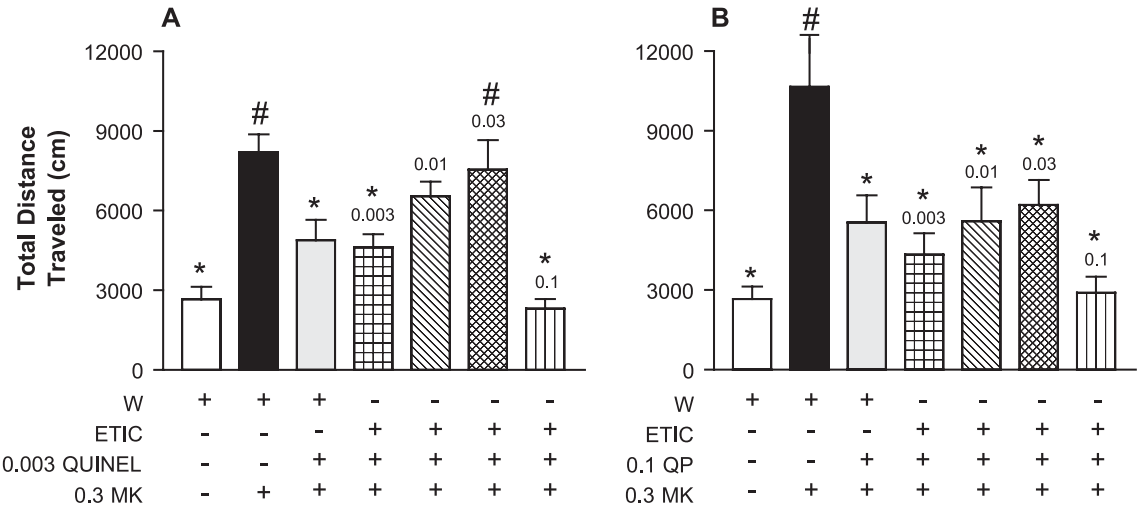

Fig. 5. Effects of eticlopride (ETIC) and water (W) pretreatment (30 min) on 0.003 mg/kg quinelorane (QUINEL) plus 0.3 mg/kg MK801 combinations (panel A) or 0.1 mg/kg quinpirole (QP) plus 0.3 mg/kg MK801 combinations (panel B) on total distance traveled in mice ($n=8$). Mice were injected intraperitoneally with eticlopride and returned to their homecages for 30 min, and then were injected with the drug combinations and placed immediately into the test chambers. Numbers above bars indicate doses (μg/kg) of eticlopride administered. * Significant difference compared to 0.3 mg/kg MK801 plus water. #Significant difference compared to 0.3 mg/kg MK801 plus quinelorane (or quinpirole). Other details as in Figs. 1 and 4.

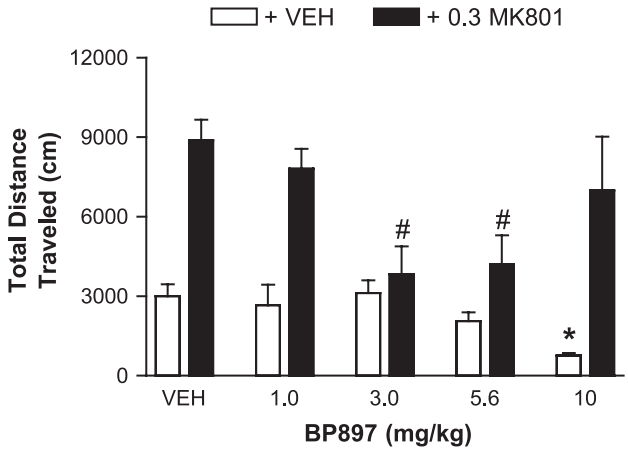

Fig. 6. Effects of BP897 in combination with VEH (vehicle) or in combination with 0.3 mg/kg MK801 on total distance traveled in mice ($n=8$). Mice were injected subcutaneously with vehicle in combination with BP897 or vehicle as well as in combination with MK801, and then were placed immediately into the test chambers. * Significant difference compared to vehicle alone. #Significant difference compared to 0.3 mg/kg MK801 plus vehicle. Other details as in Fig. 1.

combined with MK801 there was a main effect for total distance traveled [$F(4,35)=3.27$, $P<.05$] such that doses of 3.0 and 5.6 mg/kg BP897 significantly reduced MK801-induced locomotion.

3.7. Effects of nafadotride and eticlopride pretreatment on the effects of MK801 in combination with BP897

Fig. 7 shows the effects of nafadotride and eticlopride pretreatment on the combination of 0.3 mg/kg MK801 plus 5.6 mg/kg BP897. There was a main effect of drug treatment for nafadotride pretreatment and 5.6 mg/kg BP897/MK801 combinations on total distance traveled [$F(6,49)=7.90$, $P<.05$] (Fig. 7A); however, post hoc tests failed to reveal that the attenuating effects of 5.6 mg/kg BP897 on MK801-induced total distance traveled were reversed by nafadotride. There was main effect of drug treatment for eticlopride pretreatment on 5.6 mg/kg BP897/MK801 combinations for total distance traveled [$F(5,42)=13.88$, $P<.05$] (Fig. 7B); however, post hoc tests failed to reveal that the attenuating effects of 5.6 mg/kg BP897 on MK801-induced total distance traveled were reversed by eticlopride.

3.8. Effects of BP897 pretreatment on the effects of MK801 alone and combination with quinelorane or quinpirole

There was a main effect of BP897 pretreatment in combination with vehicle for distance traveled [$F(4,35)=5.37$, $P<.05$] with a dose of 10 mg/kg significantly reducing activity (data not shown). Pretreatment with BP897 signifi-

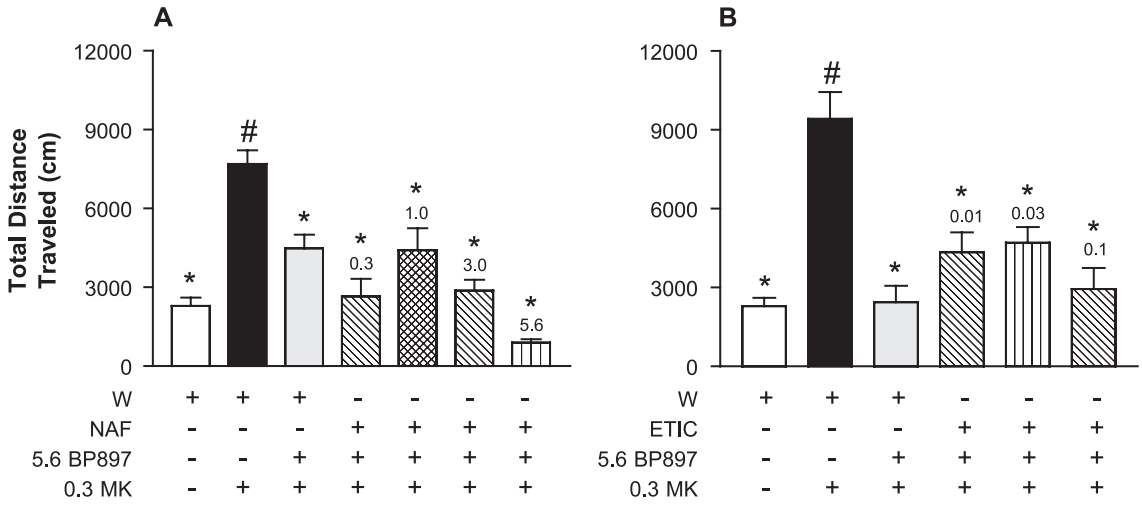

Fig. 7. Effects of nafadotride (NAF) (panel A) or eticlopride (ETIC) (panel B) pretreatment (30 min) on 5.6 mg/kg BP897 plus 0.3 mg/kgMK801 combinations on total distance traveled in mice ($n=8$). Mice were injected intraperitoneally with water (W), nafadotride or eticlopride and returned to their homecage for 30 min, and then were injected with the drug combinations and placed immediately into the test chambers. Numbers above the bars indicate doses (mg/kg) of nafadotride and eticlopride administered. * Significant difference compared to 0.3 mg/kg MK801 plus water. #Significant difference compared to 0.3 mg/kg MK801 plus BP897. Other details as in Figs. 1 and 4.

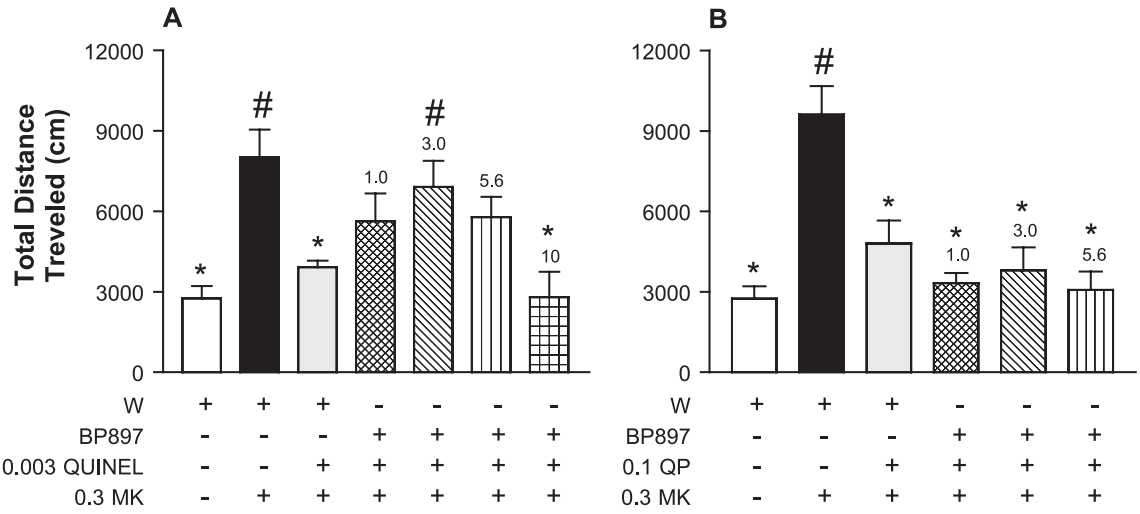

Fig. 8. Effects of BP897 and water pretreatment on 0.003 mg/kg quinelorane (QUINEL) plus 0.3 mg/kg MK801 combinations (panel A) or 0.1 mg/kg quinpirole (QP) plus 0.3 mg/kg MK801 combinations (panel B) on total distance traveled in mice ($n=8$). Mice were injected subcutaneously with water or BP897 and returned to their homecage for 30 min, and then were injected with the drug combinations and placed immediately into the test chambers. Numbers above bars indicate doses (μg/kg) of BP897 administered. * Significant difference compared to 0.3 mg/kg MK801 plus water. #Significant difference compared to 0.3 mg/kg MK801 plus quinelorane (or quinpirole). Other details as in Figs. 1 and 4.

cantly reduced MK801-induced total distance traveled [$F(4,35)=6.73$, $P<.05$] at a dose of 10 mg/kg (data not shown). Fig. 8 shows the effects of BP897 pretreatment on the locomotor effects of 0.3 mg/kg MK801 in combination with 0.003 mg/kg quinelorane or 0.1 mg/kg quinpirole. There was a main effect of drug treatment on total distance traveled [$F(6,49)=5.94$, $P<.05$] (Fig. 8A) such that a dose of 3.0 mg/kg BP897 completely reversed the attenuating effects of quinelorane on MK801. There was a main effect of drug treatment for BP897 pretreatment on 0.1 mg/kg quinpirole/MK801 combinations for total distance traveled [$F(5,42)=11.52$, $P<.05$] (Fig. 8B); however, post hoc tests failed to reveal that the attenuating effects of 0.1 mg/kg quinpirole on MK801-induced total distance traveled were reversed by BP897.

4. Discussion

The present study examined the modulatory actions of DA D2/3 agonists on the locomotor activating effects of the noncompetitive NMDA receptor antagonist MK801. MK801-induced locomotion was attenuated by the coadministration of the D2/3 agonists quinelorane and quinpirole as well as the partial D3 agonist, BP897. These results extend previous findings demonstrating that D2/3 agonists attenuate MK801-induced stereotypies (Verma and Kulkarni, 1992; Witkin et al., 1998) to now include MK801-induced locomotion.

MK801 (0.1–1.0 mg/kg) produced a biphasic effect on locomotor activity that was consistent with previous reports (Irifune et al., 1995; Starr and Starr, 1994; Verma and Kulkarni, 1992). A dose of 0.3 mg/kg MK801 produced maximal increases in total distance traveled. Although doses of 0.1 and 1.0 mg/kg resulted in quantitatively similar levels of activity, the 1.0 mg/kg dose produced signs of ataxia in several mice. This is consistent with previous findings at a dose of 1.0 mg/kg MK801 (Irifune et al., 1995; Starr and Starr, 1994).

The coadministration of the D2/3 agonists quinelorane or quinpirole with MK801 (0.3 mg/kg) significantly reduced the locomotor stimulant effects of MK801 at doses that alone had little effect on total distance traveled. Because MK801 produced a biphasic effect, the results indicating a reduction in the effects of 0.3 mg/kg MK801 by the D2/3 agonists could be conversely interpreted as an enhancement of MK801's effects (i.e., as functionally increasing the effects of 0.3 mg/kg to be similar to a larger dose given alone and on the downward limb of the dose–response curve). However, considering that administering 0.3 mg/kg MK801 in combination with the D2/3 agonists produced less locomotion than 0.3 mg/kg MK801 alone and did so in the absence of overt signs of ataxia suggests that the resulting levels of activity were functionally similar to a smaller and not to a larger dose of MK801. Similar results were obtained with BP897, a drug that has partial agonist activity at D3 receptors and weak antagonist properties at D2 receptors (Pilla et al., 1999; Wicke and Garcia-Ladona, 2001; Wood et al., 2000). It was not surprising that a dose of 10 mg/kg BP897 alone decreased spontaneous activity as the ED_{50} of BP897 to induce catalepsy in rodents is approximately 12 mg/kg (J. Costentin, personal communication to Pilla et al., 1999), and the ED_{50} to exhibit D2 receptor occupancy in the mouse striatum is approximately 15 mg/kg (Pilla et al., 1999). It appears likely that BP897, which attenuated the effects of MK801 at doses of 3.0 and 5.6 mg/kg, was producing effects primarily through its partial agonist activity at D3 receptors. However, in a recent

report a dose of 3.0 mg/kg BP897 reduced the locomotor activating effects of 0.3 mg/kg MK801 in D3 receptor knockout mice (Leriche et al., 2003). This suggests a potential role for D2 receptors in the effects of BP897 at doses of 3.0 mg/kg and larger.

Nafadotride, an antagonist exhibiting approximately 10-fold selectivity for D3 over D2 receptors (Audinot et al., 1998; Sautel et al., 1995b), did not alter the locomotor effects of MK801, but reversed or partially reversed the attenuating effects of quinelorane and quinpirole, respectively, on MK801-induced locomotion. In contrast, eticlopride, an antagonist with up to 15-fold selectivity for D2 over D3 receptors (Levant, 1997), decreased the effects of MK801 at the highest dose tested (0.1 mg/kg), which was also the dose that significantly inhibited behavior when administered alone. Therefore, this reduction of the locomotor activating effects of MK801 is more likely attributable to a nonspecific reduction in activity by eticlopride than its receptor interaction with MK801. Eticlopride, like nafadotride, reversed the effects of quinelorane on MK801-induced total distance traveled, but failed to reverse quinpirole's effect on MK801-induced total distance traveled. Previously, eticlopride had been shown to antagonize the locomotor stimulant effects of 0.1 mg/kg MK801 in rats (Hoffman, 1992); however, eticlopride was not tested alone in that study thus making interpretation of D2 receptor involvement in MK801's effects difficult. Neither nafadotride nor eticlopride reversed the attenuating effects of BP897 on MK801-induced total distance traveled, which was in contrast to their effects on quinelorane. It is presently unclear as to why nafadotride and eticlopride failed to reverse the attenuating effects of BP897.

Both quinelorane and quinpirole generally exhibit higher affinity for D3 than D2 receptors (see Levant, 1997), and in functional in vitro assays they are either as selective, or more selective for D3 versus D2 receptors (Chio et al., 1994; Sautel et al., 1995a). At doses of nafadotride up to 3.0 mg/kg ip (as in the present study) negligible binding at D2 receptors occurs (Levant and Vansell, 1997). As such, the reversal of quinelorane and quinpirole's effects on MK801 suggests that D3 receptor activation is involved in modulating MK801-induced locomotion. However, D2 receptor involvement cannot be eliminated as eticlopride (with a greater affinity for D2 versus D3 receptors) was effective as well against quinelorane and quinpirole.

Although it is unclear as to why BP897 was effective in reversing quinelorane's, but not quinpirole's attenuation of MK801's effects, there are data suggesting that quinpirole may bind to a novel site in addition to D2/3 receptors. For example, monoamine oxidase inhibitors inhibit [^{3}H]quinpirole binding, but fail to block quinpirole-stimulated GTPγS binding. BP897 can exhibit antagonistic actions at both D3 and D2 receptors (Pilla et al., 1999; Wood et al., 2000); thus it would be predicted that at least some partial antagonism of quinpirole might have been observed, but that was not the case. Although quinelorane and quinpirole produced similar effects in the present study, the interaction tests with BP897 suggest that their mechanisms may not be the same. Furthermore, there are other experimental conditions in which quinpirole and quinelorane differ in their effects. Quinelorane, unlike quinpirole, attenuates the antinociceptive effects of both mu and kappa opioid agonists (unpublished observations; Cook et al., 2000; Cook et al., 1999).

A possible explanation for the attenuation of MK801-induced locomotion by D2/3 agonists is that these agonists are able to presynaptically modulate the increases in DAergic activity induced by MK801, which ultimately results in reducing MK801's effects at the behavioral level. The potency with which quinelorane and quinpirole inhibit DA release in the striatum, nucleus accumbens and frontal cortex correlate with their affinity at D3, but not D2 receptors (Gobert et al., 1995). In addition to presynaptic modulation of DA release, it remains a possibility that postsynaptic D2/3 receptor activation, through a negative feedback loop, may alter DA release (Koeltzow et al., 1998). However, the apparent in vitro and in vivo selectivity relationship of D2/3 agonists such as quinelorane and quinpirole has been challenged by studies demonstrating behavioral and physiological effects in D3 but not D2 receptor knockout mice (Boulay et al., 1999a,b). Taken together, it remains unclear as to the extent to which D3 and/or D2 receptors contribute to the modulatory actions of the D2/3 agonists on MK801-induced locomotion. Nonetheless, these drugs are amongst the most widely tested and currently available drugs exhibiting degrees of selectivity for D2 and D3 receptors.

Given the evidence that the etiology of schizophrenia potentially involves hypoglutamatergic transmission that results in a hyperdopaminergic state, these data support the hypothesis that D2/3 agonists, which can inhibit DAergic activity, may serve as a mechanism for restoring the neurochemical imbalance within the schizophrenic brain. Although the relative contribution of D3 and/or D2 receptors in producing the observed attenuation in the present study remains unknown, there is clear evidence that D2 receptors (e.g., Seeman, 1987) and emerging evidence that D3 receptors (e.g., Schwartz et al., 2000) are involved in mediating schizophrenic symptomology. Presently, we are limited by the selectivity of existing ligands; however, as more D3 and D2 selective agents become available the relative contribution of each DA receptor subtype will be revealed.

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References

Aretha CW, Sinha A, Galloway MP. Dopamine D3-preferring ligands act at synthesis modulating autoreceptors. J Pharmacol Exp Ther 1995; 274:609–13.

Audinot V, Newman-Tancredi A, Gobert A, Rivet JM, Brocco M, Lejeune F, et al. A comparative in vitro and in vivo pharmacological characterization of the novel dopamine D3 receptor antagonists (+)-S 14297, nafadotride, GR 103,691 and U 99194. J Pharmacol Exp Ther 1998; 287:187–97.

Boulay D, Depoortere R, Perrault G, Borrelli E, Sanger DJ. Dopamine D2 receptor knock-out mice are insensitive to the hypolocomotor and hypothermic effects of dopamine D2/D3 receptor agonists. Neuropharmacology 1999a;38:1389–96.

Boulay D, Depoortere R, Rostene W, Perrault G, Sanger DJ. Dopamine D3 receptor agonists produce similar decreases in body temperature and locomotor activity in D3 knock-out and wild-type mice. Neuropharmacology 1999b;38:555–65.

Carlsson A. The current status of the dopamine hypothesis of schizophrenia. Neuropharmacology 1988;1.

Carlsson M, Carlsson A. The NMDA antagonist MK-801 causes marked locomotor stimulation in monoamine-depleted mice. J Neural Transm 1989;75:221–6.

Chausmer AL, Katz JL. The role of D2-like dopamine receptors in the locomotor stimulant effects of cocaine in mice. Psychopharmacology 2001;155:69–77.

Chio CL, Lajiness ME, Huff RM. Activation of heterologously expressed D3 dopamine receptors: comparison with D2 dopamine receptors. Mol Pharmacol 1994;45:51–60.

Cook CD, Beardsley PM. The modulatory actions of dopamine D2/3 agonists and antagonists on the locomotor-activating effects of morphine and caffeine in mice. Pharmacol Biochem Behav 2003;75:363–71.

Cook CD, Rodefer JS, Picker MJ. Selective attenuation of the antinociceptive effects of mu opioid by the putative dopamine D3 agonist 7-OH-DPAT. Psychopharmacology 1999;144:239–47.

Cook CD, Barrett AC, Syvanthong C, Picker MJ. Modulatory effects of dopamine D3/2 agonists on kappa opioid-induced antinociception and diuresis in the rat. Psychopharmacology 2000;152:14–23.

Dekeyne A, Rivet JM, Gobert A, Millan MJ. Generalization of serotonin (5-HT)1A agonists and the antipsychotics, clozapine, ziprasidone and S16924, but not haloperidol, to the discriminative stimuli elicited by PD128,907 and 7-OH-DPAT. Neuropharmacology 2001;40:899–910.

Di Chiara G. Role of dopamine in the behavioural actions of nicotine related to addiction. Eur J Pharmacol 2000;393:295–314.

Di Chiara G, Imperato A. Opposite effects of mu and kappa opiate agonists on dopamine release in the nucleus accumbens and in the dorsal caudate of freely moving rats. J Pharmacol Exp Ther 1988;244:1067–80.

Di Chiara G, Acquas E, Tanda G. Ethanol as a neurochemical surrogate of conventional reinforcers: the dopamine–opioid link. Alcohol 1996; 13:13–7.

Gainetdinov RR, Sotnikova TD, Grekhova TV, Rayevsky KS. In vivo evidence for preferential role of dopamine D3 receptor in the presynaptic regulation of dopamine release but not synthesis. Eur J Pharmacol 1996;308:261–9.

Gilbert DB, Millar J, Cooper SJ. The putative dopamine D3 agonist, 7-OH-DPAT, reduces dopamine release in the nucleus accumbens and electrical self-stimulation to the ventral tegmentum. Brain Res 1995;681:1–7.

Gobert A, Rivet JM, Audinot V, Cistarelli L, Spedding M, Vian J, et al. Functional correlates of dopamine D3 receptor activation in the rat in vivo and their modulation by the selective antagonist, (+)-S 14297: II. Both D2 and "silent" D3 autoreceptors control synthesis and release in mesolimbic, mesocortical and nigrostriatal pathways. J Pharmacol Exp Ther 1995;275:899–913.

Hoffman DC. Typical and atypical neuroleptics antagonize MK-801-induced locomotion and stereotypy in rats. J Neural Transm Gen Sect 1992;89:1–10.

Irifune M, Shimuzu T, Nomoto M, Fukuda T. Involvement of *N*-methyl-D-aspartate (NMDA) receptors in noncompetitive NMDA receptor antagonist-induced hyperlocomotion in mice. Pharmacol Biochem Behav 1995;51:291–6.

Javitt DC, Zukin SR. Recent advances in the phencyclidine model of schizophrenia. Am J Psychiatry 1991;148:1301–8.

Koeltzow TE, Xu M, Cooper DC, Hu X-T, Tonegawa S, Wolf ME, et al. Alterations in dopamine release but not dopamine autoreceptor function in dopamine D3 receptor mutant mice. J Neurosci 1998;18:2231–8.

Leriche L, Schwartz JC, Sokoloff P. The dopamine D(3) receptor mediates locomotor hyperactivity induced by NMDA receptor blockade. Neuropharmacology 2003;45:174–81.

Levant B. The D3 dopamine receptor: neurobiology and potential clinical relevance. Pharmacol Rev 1997;49:231–52.

Levant B, Vansell NR. In vivo occupancy of D2 dopamine receptors by nafadotride. Neuropsychopharmacology 1997;17:67–71.

Lodge D, Johnson KM. Noncompetitive excitatory amino acid receptor antagonists. Trends Pharmacol Sci 1990;11:81–6.

Marcus MM, Mathe JM, Nomikos GG, Svensson TH. Effects of competitive and non-competitive NMDA receptor antagonists on dopamine output in the shell and core subdivisions of the nucleus accumbens. Neuropharmacology 2001;40:482–90.

Marino MJ, Conn PJ. Direct and indirect modulation of the *N*-methyl-D-aspartate receptor. Curr Drug Target CNS Neurol Disord 2002;1: 1–16.

Narayanan S, Willins D, Dalia A, Wallace L, Uretsky N. Role of dopaminergic mechanisms in the stimulatory effects of MK-801 injected into the ventral tegmental area and the nucleus accumbens. Pharmacol Biochem Behav 1996;54:565–73.

Navarro M, Fernandez-Ruiz JJ, de Miguel R, Hernandez ML, Cebeira M, Ramos JA. An acute dose of delta 9-tetrahydrocannabinol affects behavioral and neurochemical indices of mesolimbic dopaminergic activity. Behav Brain Res 1993;57:37–46.

Ouagazzal A, Nieoullon A, Amalric M. Effects of dopamine D1 and D2 receptor blockade on MK-801-induced hyperlocomotion in rats. Psychopharmacology 1993;111:427–34.

Pilla M, Perachon S, Sautel F, Garrido F, Mann A, Wermuth CG, et al. Selective inhibition of cocaine-seeking behaviour by a partial dopamine D3 receptor agonist. Nature 1999;400:371–5.

Rao TS, Kim HS, Lehmann J, Martin LL, Wood PL. Interactions of phencyclidine receptor agonist MK-801 with dopaminergic system: regional studies in the rat. J Neurochem 1990;54:1157–62.

Sautel F, Griffon N, Levesque D, Pilon C, Schwartz JC, Sokoloff P. A functional test identifies dopamine agonists selective for D3 versus D2 receptors. Neuroreport 1995a;6:329–32.

Sautel F, Griffon N, Sokoloff P, Schwartz JC, Launay C, Simon P, et al. Nafadotride, a potent preferential dopamine D3 receptor antagonist, activates locomotion in rodents. J Pharmacol Exp Ther 1995b;275: 1239–46.

Schwartz JC, Diaz J, Pilon C, Sokoloff P. Possible implications of the dopamine D(3) receptor in schizophrenia and in antipsychotic drug actions. Brain Res Rev 2000;31:277–87.

Seeman P. Dopamine receptors and the dopamine hypothesis of schizophreniafs. Synapse 1987;1:133–52.

Sotnikova TD, Gainetdinov RR, Grekhova TV, Rayevsky KS. Effects of intrastriatal infusion of D2 and D3 dopamine receptor preferring antagonists on dopamine release in rat dorsal striatum (in vivo microdialysis study). Pharmacol Res 2001;43:283–90.

Starr MS, Starr BS. Comparison of the effects of NMDA and AMPA antagonists on the locomotor activity induced by selective D1 and D2 dopamine agonists in reserpine-treated mice. Psychopharmacology 1994;114:469–76.

Suzuki T, Kato H, Aoki T, Tsuda M, Narita M, Misawa M. Effects of the non-competitive NMDA receptor antagonist ketamine on morphine-induced place preference in mice. Life Sci 2000;67:383–9.

Verma A, Kulkarni SK. Modulation of MK-801 response by dopaminergic agents in mice. Psychopharmacology 1992;107:431–6.

Wicke K, Garcia-Ladona J. The dopamine D3 receptor partial agonist, BP 897, is an antagonist at human dopamine D3 receptors and at rat somatodendritic dopamine D3 receptors. Eur J Pharmacol 2001;424:85–90.

Witkin J, Gasior M, Acri J, Beekman M, Thurkauf A, Yuan J. Atypical antipsychotic-like effects of the dopamine D3 receptor agonist, (+)-PD 128,907. Eur J Pharmacol 1998;347:R1–3.

Wood MD, Boyfield I, Nash DJ, Jewitt FR, Avenell KY, Riley GJ. Evidence for antagonist activity of the dopamine D3 receptor partial agonist, BP 897, at human dopamine D3 receptor. Eur J Pharmacol 2000; 407:47–51.