The effect of a full agonist/antagonist of the D₁ receptor on locomotor activity, sensorimotor gating and cognitive function in dizocilpine-treated rats

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Abstract

Cognitive impairment has been found across all subtypes of schizophrenia. The location and function of dopamine-1 receptors (D₁Rs) make them attractive targets for the treatment of cognitive impairment in schizophrenia. Here we investigate the systemic effect of a D₁R agonist (A77636) and antagonist (SCH 23390) on hyperlocomotor activity and cognitive deficit induced by an NMDA receptor antagonist (MK-801). Wistar rats (250–300 g) received A77636 (0.1, 0.5 or 1 mg/kg) or SCH 23390 (0.02 or 0.05 mg/kg) with MK-801 (0.1 mg/kg) or saline for 4 d. On day 4 we assessed the prepulse inhibition of the acoustic startle response, locomotor activity in a novel arena and active allothetic place avoidance (spatial memory task) 15 min after the last injection. Systematic administration of the D₁R agonist at 0.1 mg/kg ameliorates cognitive dysfunction in our model of schizophrenia, but increases stereotypy and locomotor activity (model of psychotic symptoms) at higher doses (0.5 or 1 mg/kg). Administration of the D₁R antagonist had no effect on cognitive function, but decreased hyperlocomotion induced by MK-801. Thus, based on our results, over-activation of D₁Rs may exacerbate psychotic symptoms in patients with schizophrenia.

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Key words: Animal model, cognitive function, D₁ receptor, MK-801, schizophrenia.

Introduction

Schizophrenia is a complex neuropsychiatric illness that affects 1% of the population. Except for positive symptoms, cognitive impairment has been found across all subtypes of the disease (Breier, 1999; Harvey et al. 2001). Patients with schizophrenia are deficient in abstraction, executive function, verbal memory, language function, vigilance and attention (Breier, 1999). Moreover, short-term place memory is impaired in schizophrenia (McGurk et al. 2005) and a deficit in spatial working memory has even been proposed as an endophenotypic marker for schizophrenia (Glahn et al. 2003). Spatial memory is of particular interest in schizophrenia because this cognitive domain is utilized in everyday activities, such as using public transport and other daily activities that are impaired in this illness (McGurk & Mueser, 2004).

Dopaminergic signalling in the central nervous system plays a clear role in many aspects of behaviour and cognition. Dopamine modulates the activity of structures important for attention, cognition, and sensorimotor gating such as the hippocampus, thalamus and prefrontal cortex (Goldman-Rakic et al. 2004; Kodama et al. 2002; Swerdlow et al. 2001). Dopamine acts upon neurons through two families of dopamine receptors, i.e. D₁-like receptors (D₁Rs) and D₂-like receptors (D₂Rs). The D₁-like receptor family consists of D₁ and D₅ receptors, which are abundantly expressed in the hippocampus, thalamus and prefrontal cortex of humans and rodents (Mengod et al. 1992;
The prefrontal cortex, hippocampus and thalamus are important in cognitive functions. Besides the dopaminergic fibres, these structures also subcortical areas. At the cellular level, the interaction between D₁ receptors and N-methyl-d-aspartate receptors (NMDARs) is well established (Missale et al. 2006). Activation of D₁Rs phosphorylate subunits of NMDARs through secondary messengers contributes to potentiation of the NMDA-evoked response (Missale et al. 2006). Moreover, the activation of D₁Rs results in the rapid translocation of NMDARs to post-synaptic membranes and activation of NMDARs recruits the D₁Rs to the plasma membrane (Dunah et al. 2004). NMDAR stimulation enhances dopamine D₁Rs-mediated cAMP accumulation in both co-transfected cells and primary hippocampal cultures (Pei et al. 2004). A protein–protein interaction between D₃ and NR₃ and the NR₃A subunit of NMDARs in the striatum and hippocampus has also been described. Two regions in the D₁Rs carboxyl tail can directly and selectively couple to NMDAR subunits NR₃ and NR₃A. While one interaction through a NR₃A subunit is involved in the inhibition of NMDAR-gated currents, the other through a NR₃A subunit is implicated in the attenuation of NMDAR-mediated excitotoxicity through a phosphatidylinositol-3 kinase-dependent pathway (Lee et al. 2002). Both D₃ and NMDA receptors form a heteromeric complex and activation of NMDARs increase the expression of D₃Rs (Pei et al. 2004). Taken together, the activation of D₃Rs reinforces NMDA neurotransmission in the cortex, providing a target for new therapies for cognitive dysfunction in schizophrenia. Changes in NMDAR and expression of its subunits have been documented in patients with schizophrenia (Kristiansen et al. 2007; Pilowsky et al. 2006). Moreover, experiments with antagonists of NMDARs in animals have shown an enduring cognitive deficit after the administration of phencyclidine, dizocilpine (MK-801) or ketamine (Jentsch et al. 2007; Stefani & Moghaddam, 2005; Vales et al. 2006).

In the present study we show the effect of an agonist/antagonist of D₃Rs in an animal model of cognitive deficit in schizophrenia. In this model, experimental psychosis is induced by repeated systemic administration of dizocilpine (MK-801), a non-competitive NMDA antagonist. The model is based on the glutamatergic hypothesis of the origin of schizophrenia (Carlsson et al. 2001; Javitt & Zukin, 1991), which presumes that chronic inhibition of NMDA reduces inhibitor GABA neurotransmission. This induced disinhibition of the neurotransmitter system could cause psychosis (Carlsson et al. 2001). Administration of MK-801 increases locomotor activity, impairs sensorimotor gating and causes other alterations of neural functions that can be reversed by antipsychotics (Bubeníková et al. 2005; Swerdlow et al. 1998; Vales et al. 2006). Therefore, animals treated with MK-801 or similar NMDA antagonists might be an excellent pharmacological model of cognitive dysfunction in schizophrenia (Bubeníková-Valesová et al. 2008a).

The aim of the present study was to investigate the effect of a full D₃Rs agonist and antagonist on hyperlocomotor activity, on the deficit in cognitive function and prepulse inhibition of the startle response (PPI) and hyperlocomotion induced by the NMDAR antagonist (MK-801).

In this study we used the active allocthotic place avoidance (AAPA) tasks (Stuchlik et al. 2004; Vales et al. 2006) to assess cognitive function in rats. The AAPA task is a spatial memory test that can be used for determining the ability of animals to efficiently organize their behaviour in both normal and pathological conditions. A unique feature of this task is that the rats have to solve a conflict between two discordant subsets of spatial stimuli. Thus for successful performance in the AAPA task the rat has to differentiate between relevant and irrelevant stimuli (Stuchlik et al. 2004). This differentiation between stimuli is disturbed in schizophrenia patients (Light & Braff, 1999; Schwartz et al. 2001).

PPI measures sensorimotor gating, which is thought to regulate environmental inputs and selectively allocate attentional resources to salient stimuli (Braff et al. 2001). Deficits in sensorimotor gating have been observed in patients with several neuropsychiatric disorders including schizophrenia (Swerdlow et al. 1998). Measuring spatial learning and memory is dependent on the locomotor activity of animals. Therefore, it is necessary to assess locomotor activity during the AAPA test. Animals showing an inhibition or hyperlocomotion due to pharmacological intervention could not perform this spatial cognitive task properly. Moreover, locomotor activity and thigmotaxis (a parameter describing anxiety) in a novel environment may provide new information about the effect of mild stress on MK-801-induced psychoses.
Materials and methods

Drugs

MK-801 (Dizocilpine maleate; [5R,10S]-[+]-5-methyl-10,11-dihydro-5H-dibenzo[a,d] cyclohepten-5,10-imine; Sigma-Aldrich, Czech Republic) at 0.1 mg/kg was dissolved in saline and injected i.p. at 1 ml/kg 30 min prior to the AAPA test or 15 min prior to the PPI or locomotor activity test. The D1R agonist A77636 was dissolved in saline and administered at 0.1, 0.5 and 1 mg/kg s.c. in 1 ml/kg as above. The D1R antagonist SCH 23390 was dissolved in saline and administered at 0.02 and 0.05 mg/kg s.c. in 1 ml/kg as above. All animals received the same volume of liquid per kg of body weight. Control animals received saline.

Animals

Male Wistar rats (200–250 g, specific pathogen-free animals; Hannover breed Konárovice, Czech Republic) were used in this study. Cages containing two male rats each were housed in a temperature-controlled room (21–22 °C) with a 12 h light/dark regime (lights on 07:00 hours) and free access to food (ST-1 diet) and water. Each rat was experimentally naive and was tested only once. All manipulations respected the Guidelines of the European Union Council (86/609/EU) and followed the instructions of the National Committee for the Care and Use of Laboratory Animals.

Apparatus and behavioural procedures

Locomotor activity in novel environment

Locomotor activity, expressed as the total distance covered by the rat during 30 min in a box (68 cm × 68 cm × 30 cm) located in a soundproof room, was measured using a video tracking system for automation of the behavioural experiments (Noldus, EthoVision Colour Pro-version 3.1, USA).

PPI

All testing occurred within a startle chamber (SR-LAB, San Diego Instruments, USA). All rats were initially tested in a short session (5-min acclimatization period plus five single stimuli; 120 dB) 2 d before the experiment. The experimental design was as described previously (Bubeníková et al. 2005). Briefly, the acclimatization period (62 dB) was presented for 5 min. After this the test began with five initial startle stimuli (120 dB) followed by four different trial types presented in a pseudo-random order: (1) single pulse: 120 dB broadband burst, 20 ms duration; (2) prepulse: 13 dB above the background noise, 20 ms duration, presented 100 ms before the onset of the pulse alone; (3) prepulse alone: 13 dB above the background noise, 20 ms duration; (4) no stimulus. Five presentations of each trial type were given with an interstimulus interval between 25 s and 30 s. The PPI was measured as the difference between the average values of the single pulse and prepulse-pulse trials and expressed as a percentage of the PPI:

\[
100 \times \frac{\text{mean response for prepulse-pulse trials} - \text{startle response for single pulse trials}}{\text{startle response for single pulse trials}}
\]

Four single-pulse trials at the beginning of the test session were not included in the calculation of the PPI and acoustic startle response (ASR) values. Animals that had an average value >10 mV were removed from the calculation of the PPI and were marked as non-responders (~3% of the total). The number of animals removed did not differ between treatment groups.

AAPA task

The AAPA apparatus is a dry arena task, in which animals are trained to avoid a room-frame fixed stable sector on a continuously rotating arena. This consisted of a smooth metallic circular arena (82 cm in diameter), enclosed with a 30 cm high transparent Plexiglas wall and elevated 1 m above the floor of a 4 × 5 m room containing numerous of extra-maze cues. Each animal was initially placed opposite the shock sector in the arena which started to rotate. Animals had to avoid an unmarked, 60° sector identified solely by its relationship to distal room cues.

The shock sector was at the north of the four arbitrarily defined cardinal compass directions and remained in a stable spatial position throughout training. Rats wore a latex harness, which carried an infrared light-emitting diode (LED) between their shoulders. A computer-based tracking system (iTrack; Bio-Signal Group, USA) located in an adjacent room recorded the rat’s position every 40 ms. Position timeseries were stored for offline analysis (TrackAnalysis; Bio-Signal Group). Whenever the rat entered the shock sector for >500 ms, the tracking system delivered a mild, constant-current foot-shock (50 Hz, 0.5 s, 0.4–0.7 mA) and recorded an entrance. If the rat did not leave the sector, additional shocks were given every 1200 ms, but no more entrances were recorded until the rat left the sector for >300 ms. Shocks were delivered through the implanted needle and the arena floor (the highest voltage drop was between the rat’s paws and grounded floor). We used a compact floor
instead of a grid, in order to allow accumulation of intra-maze landmarks (a condition necessary for generation of a conflict between arena and room frames. The current was individualized for each rat to elicit a rapid escape response but to prevent freezing. In most cases, animals responded appropriately to 0.6 mA.

**Design of experiments and data analyses**

**Locomotor activity**

Rats received the drugs or saline for 4 d and were tested on day 4 in the arena (68 cm × 68 cm × 30 cm), 15 min after the last injection. We analysed total distance travelled in the arena during 30 min. In addition, we calculated thigmotaxis \( t = f_{\text{peripheral zones}} / f_{\text{all zones}} \). Thigmotaxis varies from 0 to 1 and indicates a probability of an appearance in any of the peripheral zones within the arena. For statistical analysis, groups were divided into two independent experimental groups (A77636 and SCH 23390). For each of the groups, the same control and MK-801-treated rats were used as reference. Each group contained ten animals. The data were statistically evaluated by two-way analyses of variance (ANOVA) with the drug A77636 or SCH 23390 as one factor and MK-801 as the second factor. When appropriate, comparisons between treatment groups were conducted using the Student-Newman-Keuls (SNK) method post-hoc test; \( p < 0.05 \) was considered significant (SigmaStat 3.0).

**Startle response and PPI**

Rats were treated and tested as above. The data for statistical evaluation was measured as the difference between the average values of the single-pulse and prepulse-pulse trials and was expressed as a percentage of the PPI (see above equation).

Groups were divided into two independent experimental groups (A77636 and SCH 23390). For each of the groups, the same control and MK-801-treated rats were used as reference. The data were evaluated as described above.

**AAPA task**

Rats received drugs or saline for 4 d and on each day were trained in the AAPA task, with the shock sector stretching from the centre of the arena to its northern edge. Experimental sessions lasted 20 min and each rat had one session every day, carried out during daylight hours. Groups were divided into two independent experimental groups (A77636 and SCH 23390). For each of the groups, the same control and MK-801-treated rats were used as reference. Each group contained eight animals. Control animals received 1 ml/kg saline 30 min prior to behavioural testing each day.

The total distance travelled in a session (total distance) was measured in the arena frame (which only takes into account the active locomotion of rats). The number of entries into the shock sector (number of errors) reflected the efficiency of spatial performance in the AAPA task. The maximum time a rat spent in the safe part of the arena between two errors in a particular session was also recorded (maximum time of avoidance). This reflected the ability to remember the shock sector location and to avoid it. We suggest that the latter gives the best indication of the ability of rats to appropriately solve the task.

**Results**

**Locomotor activity and thigmotaxis**

Locomotor activity was measured as total distance travelled during the 30 min. The two-way ANOVA for the low dose of D₁ agonist A77636 (0.1 mg/kg) showed a significant effect of MK-801 \( (F_{1,36}=7, p < 0.05) \), but no effect of A77636 \( (F_{1,36}=0.007) \) or the interaction of A77636 × MK-801 \( (F_{1,36}=0.0013) \) on locomotor activity. The SNK \( t \) test showed no effect of drugs on locomotor activity (Fig. 1a). Two-way ANOVA for the high dose of D₁ agonist A77636 (1 mg/kg) showed a significant effect of MK-801 \( (F_{1,36}=15.7, p < 0.001) \), A77636 \( (F_{1,36}=6.6, p < 0.001) \) and the interaction of A77636 × MK-801 \( (F_{1,36}=4.6, p < 0.001) \) on locomotor activity. The SNK \( t \) test showed no effect of MK-801 and D₁ agonist alone on locomotor activity (Fig. 1a). However, there was a significant increase in locomotor activity after co-administration of A77636 (1 mg/kg) and MK-801 compared to control rats \( (p < 0.001) \) or to MK-801 treated rats \( (p < 0.01) \) (Fig. 1a).

Thigmotaxis parameters showed a preference for peripheral zones in the open-field task. The two-way ANOVA for the low dose of A77636 did not show any significant effect of A77636, MK-801 or an interaction between treatments. However, a two-way ANOVA for the high dose of A77636 (1 mg/kg) showed an effect of A77636 \( (F_{1,36}=9.3, p < 0.01) \), but no effect of MK-801 or an interaction between treatments. The SNK \( t \) test showed an increase of thigmotaxis in group A77636+MK-801 compared to the control and MK-801 groups \( (p < 0.05) \) (Table 1).

A two-way ANOVA for the low dose of D₁ antagonist SCH 23390 (0.02 mg/kg) showed a significant effect of SCH 23390 \( (F_{1,36}=30.1, p < 0.001) \), but no...
The effect of D₁ receptors on MK-801-induced behaviour

Table 1. The effect of an agonist/antagonist on thigmotaxis (ℓ=f/ peripheral zones/f all zones; an interval of 0–1)

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Saline</th>
<th>MK-801</th>
</tr>
</thead>
<tbody>
<tr>
<td>Saline</td>
<td>0.85±0.01</td>
<td>0.86±0.03</td>
</tr>
<tr>
<td>A77636 (0.1 mg/kg)</td>
<td>0.83±0.02</td>
<td>0.85±0.02</td>
</tr>
<tr>
<td>A77636 (1 mg/kg)</td>
<td>0.89±0.01</td>
<td>0.92±0.01</td>
</tr>
<tr>
<td>SCH 23390 (0.02 mg/kg)</td>
<td>0.84±0.02</td>
<td>0.87±0.03</td>
</tr>
<tr>
<td>SCH 23390 (0.05 mg/kg)</td>
<td>0.78±0.05</td>
<td>0.68±0.1</td>
</tr>
</tbody>
</table>

Each group contained ten animals. Data were analysed by two-way ANOVA for agonist/antagonist at different doses with the same control group and MK-801 group. The Student–Newman–Keuls post-hoc test was used to determine differences between groups (*p < 0.05 compared to MK-801 group).

ASR and PPI

There was no effect of any drug treatments on the ASR (data not shown). A two-way ANOVA for the low dose of D₁ agonist A77636 (0.1 mg/kg) showed a significant effect of MK-801 (F₁,₃₆ =9.7, *p < 0.01), no effect of A77636 (F₁,₃₆ =0.41), and a significant interaction of A77636 × MK-801 (F₁,₃₆ =8.13, *p < 0.01) on the PPI. The SNK t test showed that MK-801 decreased the level of PPI compared to controls (*p < 0.001) and administration of the low dose of A77636 disturbed the effect of MK-801 (*p < 0.05 compared to the MK-801 group) (Fig. 2a). A two-way ANOVA for the high dose of D₁ agonist A77636 (1 mg/kg) showed a significant effect of MK-801 (F₁,₃₆ =17.2, *p < 0.001), but no effect of A77636 (F₁,₃₆ =1.1) or the interaction of A77636 × MK-801 (F₁,₃₆ =0.9) on the PPI. The SNK t test only showed a decrease in PPI after MK-801 administration (*p < 0.001).

A two-way ANOVA for the low dose of D₁ antagonist SCH 23390 (0.02 mg/kg) showed a significant effect of MK-801 (F₁,₃₆ =8.9, *p < 0.01) and an interaction of SCH 23390 × MK-801 (F₁,₃₆ =12.6, *p < 0.001), but no effect of SCH 23390 (F₁,₃₆ =1.7) on the PPI. The SNK t test showed a decrease in PPI after administration of SCH 23390 alone (*p < 0.01) and in combination with

The effect of MK-801 on locomotor activity over 30 min in a novel arena. (a) Shows that the chronic administration of a D₁ agonist (A77636) had no effect on spontaneous activity. Although the administration of MK-801 (0.1 mg/kg) had no effect on locomotor activity, the combination of MK-801 with the high dose of A77636 (1 mg/kg) increased locomotor activity compared to the MK-801 group (**p < 0.01) or the control group (***p < 0.001). (b) Shows that the chronic administration of a D₁ antagonist (SCH 23390) decreased spontaneous locomotor activity at both doses (0.02 and 0.05 mg/kg) compared to the control groups (**p < 0.01, ***p < 0.001). Administration of MK-801 (0.1 mg/kg) slightly increased locomotor activity (*p < 0.05). Administration of SCH 23390 decreased locomotor activity in MK-801-treated rats (###p < 0.001 compared to the MK-801 group).

The effect after administration of D₁(D₁) agonist/antagonist on locomotor activity over 30 min in a novel arena. (a) Shows that the chronic administration of a D₁ agonist (A77636) had no effect on spontaneous activity. Although the administration of MK-801 (0.1 mg/kg) had no effect on locomotor activity, the combination of MK-801 with the high dose of A77636 (1 mg/kg) increased locomotor activity compared to the MK-801 group (**p < 0.01) or the control group (***p < 0.001). (b) Shows that the chronic administration of a D₁ antagonist (SCH 23390) decreased spontaneous locomotor activity at both doses (0.02 and 0.05 mg/kg) compared to the control groups (**p < 0.01, ***p < 0.001). Administration of MK-801 (0.1 mg/kg) slightly increased locomotor activity (*p < 0.05). Administration of SCH 23390 decreased locomotor activity in MK-801-treated rats (###p < 0.001 compared to the MK-801 group).
MK-801 ($p<0.001$) (Fig. 1b). A two-way ANOVA for the high dose of D$_1$ antagonist SCH 23390 (0.05 mg/kg) showed a significant effect of MK-801 ($F_{1,36}=25.5$, $p<0.001$), but no effect of SCH 23390 ($F_{1,36}=0.006$) or the interaction of SCH 23390 × MK-801 ($F_{1,36}=0.34$) on the PPI. The SNK $t$ test showed a decrease in PPI level after MK-801 treatment ($p<0.001$) (Fig. 1b).

**AAPA task**

A two-way ANOVA for the low dose of D$_1$ agonist A77636 (0.1 mg/kg) showed no effect of drugs or their combination on locomotor activity during the AAPA task (Fig. 3a). There was a significant effect of the interaction A77636 × MK-801 ($F_{1,28}=27.8$, $p<0.001$), but no effect of MK-801 ($F_{1,28}=2.3$) or A77636 ($F_{1,28}=0.05$) on the number of entrances into the shock sector (number of errors). The SNK $t$ test showed that MK-801 increased the number of errors ($p<0.001$) (Fig. 3b). The administration of the D$_1$ agonist blocked the effect of MK-801 ($p<0.001$), but the D$_1$ agonist...
alone increased the number of errors ($p<0.01$) (Fig. 3b). There was a significant effect of the interaction $A77636 \times MK-801$ ($F_{1,28} = 13.8, p < 0.001$), but no effect of $MK-801$ ($F_{1,28} = 0.94$) or $A77636$ ($F_{1,28} = 0.05$) on the maximum time of avoidance. The SNK $t$ test showed that administration of $MK-801$ ($p < 0.01$) and $D_1$ agonist alone ($p < 0.05$) decreased the parameter (Fig. 3c). Administration of $A77636$ blocked the effect of $MK-801$ ($p < 0.05$).

A two-way ANOVA for the high dose of $D_1$ agonist $A77636$ (0.5 mg/kg) showed no effect of the drugs or their combination on locomotor activity during the AAPA task (Fig. 3a). There was a significant effect of $MK-801$ ($F_{1,28} = 11.5, p < 0.001$), but no effect of $A77636$ ($F_{1,28} = 0.14$) or the interaction $A77636 \times MK-801$ ($F_{1,28} = 3.1$) on the number of entrances into the shock sector (number of errors). The SNK $t$ test showed that $MK-801$ increased the number of errors ($p < 0.01$) (Fig. 3b). Administration of the $D_1$ agonist had no effect on the parameter in $MK-801$-treated rats (Fig. 3b). There was a significant effect of the interaction $A77636 \times MK-801$ ($F_{1,28} = 7.1, p < 0.01$) and $MK-801$ ($F_{1,28} = 6.5, p < 0.01$), but no effect of $A77636$ ($F_{1,28} = 0.03$) on the maximum time of avoidance. The SNK $t$ test showed that administration of $MK-801$ ($p < 0.01$) decreased the parameter. However, there was no effect of $A77636$ at the high dose (Fig. 3c).

A two-way ANOVA for the low dose of $D_1$ antagonist $SCH 23390$ (0.02 mg/kg) showed a significant effect of $MK-801$ ($F_{1,28} = 32.2, p < 0.001$), but no effect of the interaction of $SCH 23390 \times MK-801$ ($F_{1,28} = 0.72$) or $MK-801$ ($F_{1,28} = 0.2$) on locomotor activity. The SNK $t$ test showed that $SCH 23390$ decreased locomotor activity with or without $MK-801$ co-treatment (Fig. 4a).

A two-way ANOVA for the low dose of $D_1$ antagonist $SCH 23390$ (0.02 mg/kg) showed a significant effect of $MK-801$ ($F_{1,28} = 19.8, p < 0.001$) and the interaction of $SCH 23390 \times MK-801$ ($F_{1,28} = 19.4, p < 0.001$), but no effect of $SCH 23390$ ($F_{1,28} = 3.3$) on the number of errors. The SNK $t$ test showed an increase in the number of errors after administration of $SCH 23390$ without $MK-801$ ($p < 0.001$) and $MK-801$ alone ($p < 0.001$) (Fig. 4b). There was no interaction between $MK-801$ and $D_1$ antagonist at the low dose. There was a significant effect of the interaction $SCH 23390 \times MK-801$ ($F_{1,28} = 21.8, p < 0.001$), $MK-801$ ($F_{1,28} = 24.2, p < 0.001$) and $SCH 23390$ ($F_{1,28} = 33.9, p < 0.001$) on the maximum time of avoidance. The SNK $t$ test showed that $SCH 23390$ decreased the maximum time avoided when administered alone ($p < 0.001$) or in combination with $MK-801$ ($p < 0.001$) (Fig. 4c).

A two-way ANOVA for the high dose of $D_1$ antagonist $SCH 23390$ (0.05 mg/kg) showed a significant effect of $SCH 23390$ ($F_{1,28} = 22.2, p < 0.001$), but no effect of the interaction of $SCH 23390 \times MK-801$ ($F_{1,28} = 0.35$) or $MK-801$ ($F_{1,28} = 3.2$) on locomotor activity. The SNK $t$ test showed that $SCH 23390$ decreased locomotor activity with or without $MK-801$ co-treatment (Fig. 4a).
Two-way ANOVA for the high dose of D₁ antagonist SCH 23390 (0.05 mg/kg) showed a significant effect of MK-801 \( (F_{1,28} = 14.9, p < 0.001) \) and the interaction of SCH 23390 × MK-801 \( (F_{1,28} = 19.5, p < 0.001) \), but no effect of SCH 23390 \( (F_{1,28} = 5.5) \) on the number of errors. The SNK \( t \) test showed an increase in the number of errors after administration of SCH 23390 both with and without MK-801 \( (p < 0.001 \text{ in both cases}) \) (Fig. 4b). There was no interaction between MK-801 and the D₁ antagonist at the low dose. There was a significant effect of the interaction SCH 23390 × MK-801 \( (F_{1,28} = 21.6, p < 0.001) \), MK-801 \( (F_{1,28} = 24.6, p < 0.001) \) and SCH 23390 \( (F_{1,28} = 34.9, p < 0.001) \) on the maximum time of avoidance. The SNK \( t \) test showed that SCH 23390 decreased the maximum time avoided when administered alone \( (p < 0.001) \) or in combination with MK-801 \( (p < 0.001) \) (Fig. 4c).

**Discussion**

The location and function of D₁Rs make them promising targets for the treatment of negative symptoms and cognitive impairments in schizophrenia. Here we investigated the effect of a full agonist of D₁R \( (A77636) \) on hyperlocomotor activity and cognitive deficit induced by an NMDAR antagonist (MK-801). D₁Rs are the most abundant dopamine receptors in the brain. Except for the prefrontal cortex and hippocampus, D₁ mRNA has been found in the striatum and nucleus accumbens (Missale et al. 1998), structures that control motor function (Pijnenburg et al. 1976). We found that subchronic administration of a high dose of D₁R agonist enhanced the effect of MK-801 on locomotor activity, but had no effect itself. The low dose of D₁ agonist had no effect on locomotor activity with or without MK-801 treatment. Similar findings were observed after administration of a partial D₁R agonist SKF 38393 (Goodwin et al. 1992; Starr & Starr, 1993). The local infusion of a partial D₁ agonist (SKF 38393) into the nucleus accumbens or striatum leads to an increase in spontaneous locomotor activity (Kreipke & Walker, 2004; Rouillon et al. 2008). Unlike systematic injection, the infusion of a NMDAR antagonist (D-AP5) into the core of the nucleus accumbens reduces the effect of the D₁R agonist on locomotor activity (David et al. 2004). A similar effect on locomotor activity was observed after infusion of a D₁R agonist and MK-801 into the striatum (Campbell et al. 2006; Kreipke & Walker, 2004). These data show that local infusion of a D₁R agonist into the structures that control motor function has the opposite effect to systematic administration on MK-801-induced hyperlocomotion. To date, this difference between the local and systemic action of a D₁R agonist has not been explained.

Systematic subchronic administration of the D₁R antagonist (SCH 23390) decreased both spontaneous and MK-801-induced locomotor activity. Inhibition of locomotor activity in rats was observed after systemic or local infusion of SCH 23390 into the striatum and nucleus accumbens (Meyer et al. 1993; Shapovalova & Kamkina, 2008). In addition, acute administration of SCH 23390 decreased locomotor sensitization produced by the NMDA antagonist phencyclidine (Phillips et al. 2001).

Locomotor activity in a novel environment is stressful for rats. The parameter that describes anxiety or other changes in exploratory activity is thigmotaxis. A high thigmotaxis score (close to 1) indicates that the frequency of occurrence of the animal in the central part of the arena is small. Co-administration of the high dose of D₁R agonist \( (A77636) \) with MK-801 increased thigmotaxis, which could indicate higher anxiety in these rats. However, an increased thigmotaxis score is connected with hyperlocomotion and therefore this behaviour may be due to stereotypy. Administration of the D₁R antagonist had no effect on thigmotaxis even when SCH 23390 inhibited locomotor activity.

Hyperlocomotion and stereotypy induced by an NMDAR antagonist represent a model of the positive symptoms with high predictive validity in antipsychotic effect (Bubenikova-Valesova et al. 2008a; Lipska & Weinberger, 2000). Therefore, our data documenting that the high dose of D₁R agonist increases these parameters indicate that D₁ agonists, especially in higher doses, would not be effective in the treatment of positive symptoms in schizophrenia or that they would even potentiate this modality.

PPI measures processing of sensory information and is reduced in several neuropsychiatric disorders (Braff et al. 2001). Systemic administration of NMDA antagonists as a model of schizophrenia-like behaviour also reduced PPI (Bubenikova-Valesova et al. 2008a). However, there is evidence that the D₁R rather than D₂R regulates PPI in rats. A study on D₁R knock-out mice showed that MK-801 reduced PPI in mice lacking D₁ and D₂ receptors, which means that the MK-801-produced deficit in PPI is independent of D₁ and D₂ receptors (Ralph-Williams et al. 2002). However, in the present study we found that subchronic administration of a low dose D₁R agonist blocked the effect of MK-801, but the high dose of A77636 had no effect. In addition, the D₁R agonist alone had no effect on PPI. In contrast, an intense study using a partial agonist of D₁R \( (SKF 38393) \) showed that the agonist
potentiated the effect of MK-801 on PPI (Bortolato et al. 2005). In the Bortolato et al. study, one dose of a partial agonist SKF 38393 (10 mg/kg) was used, therefore we do not know if the lower dose has the same effect. Moreover, SKF 38393 potentiated the disruptive effect of MK-801 only at the lower dose where MK-801 alone had no effect on PPI. At a higher dose, which is comparable with the present study (0.15 mg/kg), there was no interaction between SKF 38393 and MK-801. In our study, as in the literature, the administration of a D\textsubscript{1}R antagonist (SCH 23390) had no effect on PPI deficit induced by MK-801. Moreover, local administration of a D\textsubscript{1}R antagonist (SCH 23390) into the prefrontal cortex had no effect on PPI deficit induced by systemic administration of MK-801 (de Jong & van den Buuse, 2006).

Several studies have demonstrated an improvement in cognitive processes after mild stimulation of D\textsubscript{1}Rs in both monkeys and rats (Goldman-Rakic et al. 2004; Williams & Castner, 2006). However, excessive stimulation of D\textsubscript{1}R function is known to cause a cognitive deficit (Zahrt et al. 1997). Another study revealed facilitation of spatial cognition by a full D\textsubscript{1}R agonist dihydrexidine. This corresponded to the increased release of acetylcholine (Steele et al. 1997). Systematic as well as local administration of D\textsubscript{1}R antagonist into the prefrontal cortex disrupts cognitive processes in several tasks (Rinaldi et al. 2007; Sawaguchi et al. 1990; Stuchlik et al. 2007). In accordance with the literature, subchronic administration of D\textsubscript{1}R antagonist in the present study disrupted the performance in the cognitive task in intact animals, but this effect could be due to large hypolocomotor activity in these animals.

In the present study we found that subchronic systematic administration of a low dose of D\textsubscript{1}R agonist (A77636) impaired cognitive function in the AAPA task (an increase in number of errors and a decrease in maximum time avoided) in intact animals, but ameliorated the cognitive deficit induced by MK-801. Administration of the high dose of A77636 had no effect on the MK-801-induced deficit in cognitive function. Our results confirmed that supposedly pro-cognitive drugs would not be effective in intact animals. Similar findings were published with antipsychotics, where the pro-cognitive effect was observed only in MK-801-treated animals or in animals with fimbria fornix lesions (Addy et al. 2005; Bubenikova-Valesova et al. 2008b). In addition, chronic administration of NMDA antagonist lowers dopamine levels and increases D\textsubscript{1}R in prefrontal cortex. Therefore, administration of D\textsubscript{1}R agonist could block the effect of MK-801 on cognitive function (Tsukada et al. 2005).

Our results indicate that moderate activation of D\textsubscript{1}Rs could ameliorate cognitive dysfunction in patients with schizophrenia. The biphasic effect of low and high doses of D\textsubscript{1}R agonist could be explained by the concept of a nonlinear, inverted U-shaped relationship between the intensity of D\textsubscript{1} dopamine transmission in the prefrontal cortex and efficiency of cognitive processes, suggesting that there is a specific range of D\textsubscript{1} activity implementing optimal performance in the cognitive tasks (Vijayraghavan et al. 2007). In relation to this concept, D\textsubscript{1} agonists may enhance cognition in conditions where D\textsubscript{1} function in the prefrontal cortex is insufficient, and a D\textsubscript{1} blockade can improve cognitive processes in situations where D\textsubscript{1} transmission is too intense. A decreased level of D\textsubscript{1}R-like binding in the prefrontal cortex in drug-naive patients with schizophrenia was measured with positron emission tomography imaging, and was found to be correlated with the severity of negative symptoms and cognitive dysfunction but not with the severity of positive symptoms (Okubo et al. 1997). Therefore, activation of D\textsubscript{1}Rs should be effective as a treatment for cognitive dysfunction in schizophrenia.

Taken together, our data show that subchronic systematic administration of a D\textsubscript{1}R agonist ameliorates cognitive dysfunction in our model of schizophrenia, but increases stereotypy and locomotor activity (model of psychotic-like symptoms). The administration of a D\textsubscript{1}R antagonist had no effect on cognitive function, but decreased the hyperlocomotion induced by MK-801. It should be noted that the combination of D\textsubscript{1} activation with antipsychotics such as haloperidol or risperidone could abolish this pro-psychotic effect of D\textsubscript{1}R activation.

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Statement of Interest

None.

References


