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ORIGINAL INVESTIGATION

Dopamine D2/D3 receptor agonist quinpirole impairs spatial reversal learning in rats: investigation of D3 receptor involvement in persistent behavior

Vasileios Boulougouris · Anna Castañé · Trevor W. Robbins

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Abstract
Rationale Dopamine is strongly implicated in the ability to shift behavior in response to changing stimulus-reward contingencies.
Objectives We investigated the effects of systemic administration of the D2/D3 receptor agonist quinpirole (0.1, 0.3 mg/kg), the D2/D3 receptor antagonist raclopride (0.1, 0.3 mg/kg), the selective D3 antagonist nafadotride (0.3, 1.0 mg/kg), and combined administration of raclopride (0.1 mg/kg) or nafadotride (1.0 mg/kg) with quinpirole (0.3 mg/kg) on spatial discrimination and reversal learning.
Materials and methods Rats were trained on an instrumental two-lever spatial discrimination and reversal learning task. Both levers were presented, only one of which was reinforced. The rat was required to respond on the reinforced lever under a fixed ratio 3 schedule of reinforcement. Following attainment of criterion, a reversal was introduced.
Results None of the drugs altered performance during retention of the previously reinforced contingencies. Quinpirole (0.3 mg/kg) significantly impaired reversal learning by increasing both trials and incorrect responses to criterion in reversal phase, a pattern of behavior manifested as increased perseverative responding on the previously reinforced lever. In contrast, neither raclopride nor nafadotride when administered alone altered reversal performance. However, raclopride blocked the quinpirole-induced reversal deficit, whereas combined administration of nafadotride and quinpirole affected not only performance during the reversal but also the retention phase. The reversal impairment resulting from co-administration of nafadotride and quinpirole was associated with both perseverative and learning errors.
Conclusions Our data indicate distinct roles for D2 and D3 receptors in the capacity to modify behavior flexibly in the face of environmental change.

Keywords Reversal learning · Perseveration · Learning · Discrimination · Dopamine · Quinpirole · Raclopride · Nafadotride · Obsessive-compulsive disorder · Animal model

V. Boulougouris (✉) · A. Castañé · T. W. Robbins
Department of Experimental Psychology,
Behavioral and Clinical Neuroscience Institute (BCNI),
University of Cambridge,
Downing Street,
CB2 3EB Cambridge, UK
e-mail: vb257@cam.ac.uk

A. Castañé
Department of Neurochemistry and Neuropharmacology,
Institut d'Investigacions Biomediques de Barcelona (CSIC),
IDIBAPS,
Rosselló, 161,
08036 Barcelona, Spain

Introduction

Reversal learning paradigms, where subjects have to inhibit a previously learned response and emit responses originally not reinforced, provide a valid measure of behavioral flexibility in humans (Rolls et al. 1994; Rogers et al. 2000; Murphy et al. 2002; Fellows and Farah 2003), nonhuman primates (Jones and Mishkin 1972; Butter 1969; Dias et al. 1996; Clarke et al. 2004, 2005, 2007; Lee et al. 2007), and rats (Birrell and Brown 2000; Chudasama and Robbins 2003; McAlonan and Brown 2003; Idris et al. 2005; van der Meulen et al. 2006; Boulougouris et al. 2007, 2008). Converging evidence suggests that the striatopallidal pathway mediates reversal learning, as inactivation or lesions of the dorsomedial

striatum impairs animals' ability to flexibly adapt their behavior to changes in reinforcement contingencies (Kirkby 1969; Kolb 1997; Ragozzino and Choi 2004; Ragozzino et al. 2002a, b; Divac et al. 1967; Dunnett and Iversen 1980; Ferry et al. 2002).

Accumulating evidence suggests a strong involvement of D2-like receptors in reversal learning: Ridley et al. (1981) demonstrated impaired visual reversal performance following the D2 receptor antagonist haloperidol, while Lee et al. (2007) showed that the selective D2/D3 receptor antagonist, raclopride, also leads to reversal deficits. Furthermore, Floresco et al. (2006) showed that the D2/D3 receptor antagonist eticlopride potently impaired the ability to change behavior in response to a conditional change of rule in a set-shifting task. Moreover, D-amphetamine, which increases dopamine release in the striatum, has also been shown to impair reversal learning (Idris et al. 2005), but the findings are equivocal as some authors show enhancement instead (Weiner and Feldon 1986). Finally, deletion of the D2 receptor gene in knockout mice impaired both initial and reversal learning with a larger deficit in reversal learning (Izquierdo et al. 2006; Kruzich and Grandy 2004). In contrast, although D1-like receptors are implicated in set-shifting (Ragozzino 2002; Floresco et al. 2006), they seem not to mediate reversal learning, as the D1/D5 receptor antagonist SCH 23390 affected neither discrimination nor reversal of visual stimuli (Lee et al. 2007).

Based on these lines of evidence, our study had the objective of investigating the contribution of D2-like receptors on the performance of rats in an instrumental two-lever spatial discrimination and reversal learning task via systemic administration of the D2/D3 receptor agonist quinpirole. We hypothesized that quinpirole, which has been consistently implicated in compulsive behavior (Szechtman et al. 1998, 2001; Joel et al. 2001; Kontis et al. 2008), would specifically impair response inhibition and that drug effects would be apparent during reversal but not retention of the original discrimination. Further, we aimed to investigate the distinct involvement of D2 and D3 receptor subtypes in spatial reversal learning by (a) testing the effects of systemic administration of the D2/D3 receptor antagonist raclopride (selective D2 receptor agonists and antagonists not being available) and determining whether it abolishes the quinpirole-induced reversal impairment following combined administration and (b) determining the effects of systemic administration of the selective D3 receptor antagonist nafadotride in combination with quinpirole. If nafadotride ameliorates the quinpirole-induced impairment, this would support the hypothesis of D3 receptor involvement in reversal learning. However, if the reversal impairment was blocked by raclopride but not nafadotride, this would support its mediation by D2 receptors.

Materials and methods

Subjects

Eighty-eight experimentally naïve adult male Lister Hooded rats (Charles River, UK), weighting 280–320 g at the start of the experiment, were pair-housed under a reversed light cycle (lights on from 19:00 to 07:00). Prior to the beginning of training, rats were handled for ≈5 min daily for 3 days and were put on to a food-restriction schedule (18 g of Purina lab chow per day). Water was available ad libitum, and testing took place between 13:00 and 16:00 7 days per week. One animal was excluded due to computer failure during testing, and one group of nine animals was also excluded (see "Results" section: *Raclopride*). The total number of animals used for each group is explicitly shown in Table 1. The work was carried out under a UK Home Office Project license (PPL 80/1767) in accordance with the UK Animals (Scientific Procedures) Act 1986.

Behavioral apparatus

The behavioral apparatus consisted of eight operant conditioning chambers (30×24×30 cm; Med Associates, Georgia, VT, USA), each enclosed within a sound-attenuating wooden box fitted with a fan for ventilation and masking of extraneous noise. Each chamber was fitted with two retractable levers located on either side of a centrally positioned food magazine, into which an external pellet dispenser could deliver 45 mg sucrose pellets (Noyes dustless pellets; Sandown Scientific, Middlesex, UK), a light-emitting diode (LED), which was positioned centrally above each lever, a magazine light, and a houselight. Magazine entry was detected by an infrared photocell beam located horizontally across the entrance. The apparatus was controlled by Whisker control software (http://www.whiskercontrol.com), and the task was programmed in Visual C++ (v.6).

Behavioral procedure

Rats were trained on the instrumental two-lever spatial discrimination and serial reversal learning task as described and illustrated previously (Boulougouris et al. 2008). Briefly, rats were initially trained to nose-poke in the central magazine in order to trigger presentation of the retractable levers and to respond on them under a fixed ratio 3 (FR-3) schedule for food delivery (pretraining). The FR-3 schedule was used to preclude the possibility of reinforcing single, accidental presses on the correct lever and to render the reversal task more difficult, as the change in reversal contingencies cannot be detected from a single lever press.

Acquisition of spatial discrimination Training continued with the acquisition of a two-lever discrimination task. Now both levers were presented at trial onset, and the rat had to learn that three lever presses on only one of these levers would result in reward.

Each session lasted 20 min and consisted of a maximum of five ten-trial blocks. Each trial began with the presentation of both levers and a visual stimulus (a lit LED). The lit LED was used as a distractor, and its location (left/right) varied from trial to trial according to a pseudo-random schedule so that the light was presented an equal number of times on each side for the session. Thus, the only stimulus with informational value for the discrimination at this phase was the spatial position of the retractable levers. Throughout the session, three lever presses on one lever (lever A) would produce a single pellet reward (correct responses) and the retraction of both levers, whereas three responses on lever B would result in lever retraction without reward delivery (incorrect responses). The position of the reinforced lever (left or right) was kept constant for each rat, but was counterbalanced between subjects.

Each rat had one training session per day and was trained to a criterion of nine correct trials in one block of ten trials (binomial distribution $p<0.01$, likelihood of attaining criterion in a ten-trial block). Once this criterion was reached, this *initial* discrimination phase was considered complete, and the animal was returned to the home cage. If the criterion was not achieved, this phase was repeated the next day until criterion attainment (Fig. 1). Animals needed 1–3 days for criterion attainment during this phase.

Within session serial reversal learning task In the next training session, reversal learning was introduced. By definition, reversal learning presupposes retention of a previously acquired discrimination. Accordingly, in the reversal session, animals were again exposed to the initial discrimination task described above (with the same lever rewarded as before: discrimination retention). This initial retention phase preceding reversal also comprised a maximum of five ten-trial blocks. Once the criterion of nine correct trials in a ten-trial block was achieved, the position of the reinforced lever was reversed (*reversal phase*). The reversal phase also consisted of a maximum of five ten-trial blocks. The learning criterion was the same as in the initial phase (nine correct trials in a ten-trial block; Fig. 1). Animals always required more than one session to reach criterion on the reversal phase. Thus, they received multiple, separate training sessions, the data of which were summed together to produce the final results. During these sessions, the initial contingency was determined by retention performance. For example:

Day 1: A+, B− (retention without reversal-criterion achieved)
Day 2: A+, B− (retention preceding reversal-criterion achieved)
A−, B+ (reversal phase-criterion NOT achieved)
Day 3: A+, B− (retention preceding reversal-criterion achieved)
A−, B+ (reversal phase-criterion achieved)

Trials and incorrect responses to criterion would be added for days 2 and 3 in the example.

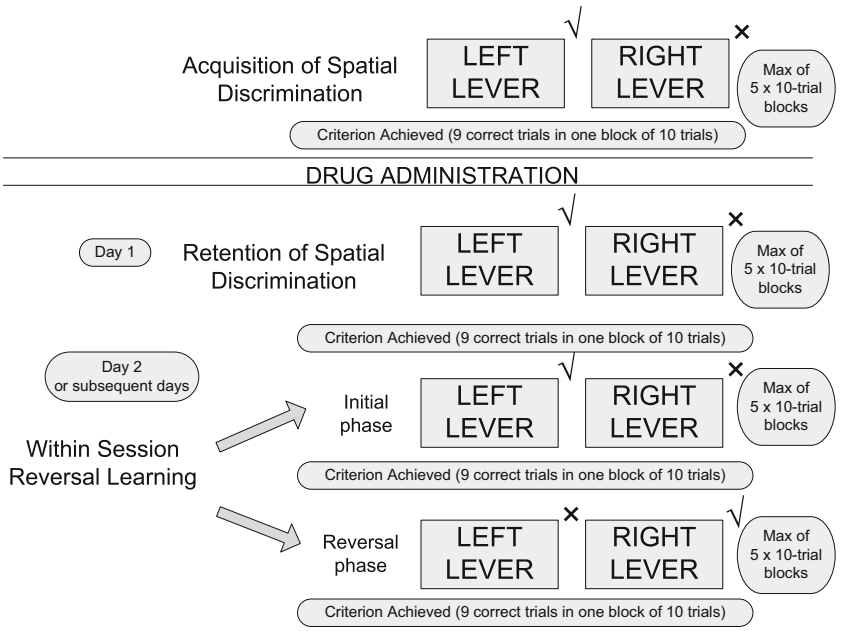

Fig. 1 Flow diagram of the behavioral procedure. Rats responded to levers under a FR3 schedule to obtain a pellet reward. The √ and × symbols indicate which lever was correct and incorrect at each stage. The correct lever was counterbalanced across rats

Drugs

The D2/D3 receptor agonist quinpirole (quinpirole hydrochloride, Q-102, Sigma Chemical, St. Louis, MO, USA), the D2/D3 receptor antagonist raclopride-L-tartrate (Sigma Chemical), and the selective D3 receptor antagonist (*S*)-nafadotride-tartrate (Sigma Chemical) were tested in one experiment. Prior to drug administration, animals were divided into eight groups, matched for their performance during the acquisition of the spatial discrimination. Each group received i.p. injections of either vehicle or quinpirole (0.1, 0.3 mg/kg), raclopride (0.1, 0.3 mg/kg), nafadotride (0.3, 1.0 mg/kg), a combination of both quinpirole 0.3 mg/kg and raclopride 0.1 mg/kg, or a combination of quinpirole 0.3 mg/kg and nafadotride 1.0 mg/kg. All drugs were administered daily 20 min prior to the start of the behavioral task in the designated area. In case of combined administration, animals were injected with raclopride or nafadotride 10 min before quinpirole administration. During this period prior to behavioral testing, animals were singly housed in clean holding cages. Following initiation of drug testing, animals required 1–3 days to achieve criterion in the retention phase and 6–9 days to achieve criterion in the reversal phase (total days of drug administration, 8 to 12).

Quinpirole and raclopride were dissolved in physiological saline while nafadotride in minimal 1.0 M HCl. All drugs were given by systemic injections in a volume of 1 ml/kg. Determination of doses was based on previous studies using the same drugs (quinpirole: Joel et al. 2001; Kurylo and Tanquay 2003; raclopride: Chang and Liao 2003; Fowler and Liou 1998; nafadotride: Levant and Vansell 1997; Bari et al. unpublished observations).

Statistical analysis

The main measures of the animals' ability to learn the discrimination and reversals were (a) the number of trials to criterion, (b) the total number of errors (i.e., incorrect trials) to criterion, and (c) the total number of incorrect responses to criterion on completed (correct and incorrect) trials (i.e., 1 incorrect trial=3 incorrect responses). Type of errors was further analyzed as described previously (Boulougouris et al. 2007) according to the method of Dias et al. (1996) and Bussey et al. (1997), modified from Jones and Mishkin (1972). In this analysis, errors during reversal learning were broken down into two learning stages: errors committed before the attainment of chance level performance (<50% correct trials) and errors committed above chance (≥50% correct trials). Jones and Mishkin regarded errors made during the first stage of learning as indicative of perseverative responses to the previously reinforced stimulus. Thus, stage 1 errors are termed "perseverative errors", whereas stage 2 errors are termed "learning errors". Additional secondary measures recorded for each trial were (d) the latency to respond, (e) the latency to collect the reward, and (f) the number of omissions.

Data for each variable were subjected to a repeated-measures ANOVA. Where significant interactions were detected, they were further explored through Newman–Keuls post hoc comparisons to establish simple effects. For all comparisons, significant difference was assumed at $p<0.05$. The between-subject factor was Group [eight levels: two different doses of each drug (one for raclopride) plus vehicle and quinpirole combined with raclopride 0.1 mg/kg or nafadotride 1.0 mg/kg], and the within-subject factors were either Retention Phase without reversal, Retention Phase preceding reversal, or Reversal Phase. Perseverative and learning errors were subjected to one-way ANOVAs followed by Newman–Keuls post hoc comparisons. The between-subject factor was Group (eight levels: as described above).

Results

Prior to drug administration, the groups did not differ in the number of incorrect responses to reach performance criterion in the acquisition of spatial discrimination ($F_{7,70}=0.60$, $p=0.75$, data not shown).

Retention without reversal

Drugs had no significant effects, at any dose, on retention (without reversal) of the drug-free spatial discrimination as indicated by a lack of effect on the number of trials (Fig. 2) or the number of incorrect responses (Fig. 3) to reach criterion.

Retention preceding reversal

Quinpirole Animals treated with quinpirole did not exhibit any significant differences on any measure during retention, preceding the reversal phase. While quinpirole (0.3 mg/kg) did appear to increase trials, but not the more robust measure of incorrect responses, to criterion, this was non-significant.

Raclopride The high dose of raclopride (0.3 mg/kg) prevented animals from initiating trials and responding, and for this reason, this group was excluded from all the analyses. Raclopride (0.1 mg/kg) increased number of trials, but not incorrect responses, to criterion on the retention preceding reversal phase (vehicle vs. raclopride 0.1 mg/kg: $p<0.001$; Fig. 2). Co-administration of raclopride (0.1 mg/kg) and quinpirole (0.3 mg/kg) yielded no significant differences on any measure during the retention preceding reversal phase.

Nafadotride Administration of nafadotride alone, at any dose, did not affect any measure of retention preceding

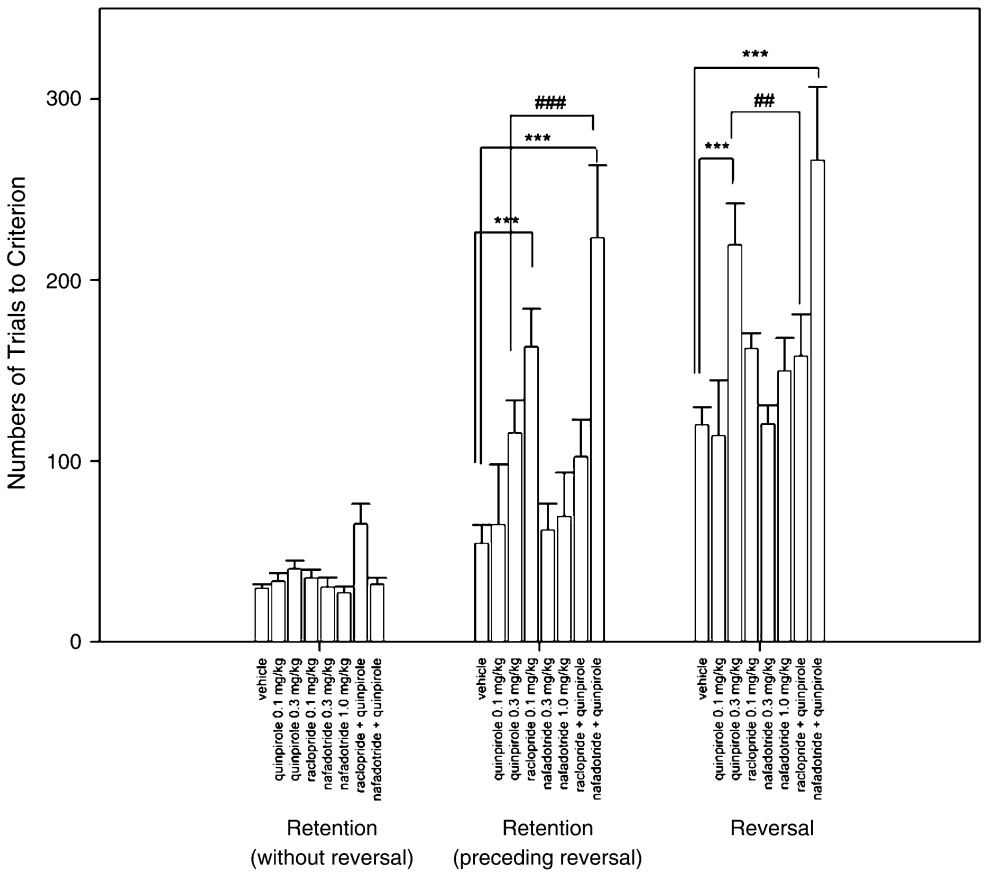

Fig. 2 Number of trials to criterion through (a) the retention phase (without reversal), (b) retention (initial) phase (preceding reversal), and (c) reversal phase. Data are presented as mean values±SEM. There were significant main effects of group and phase ($F_{7,70}$=6.78, p<0.001 and $F_{1,70}$=159.89, p<0.001, respectively) and a significant group×phase interaction ($F_{7,70}$=5.61, p<0.001). *Symbols* denote significant differences following Newman–Keuls post hoc comparisons (***,###p<0.001; ##p<0.01)

reversal phase. However, co-administration of nafadotride (1.0 mg/kg) and quinpirole (0.3 mg/kg) impaired performance on the retention preceding the reversal phase by increasing both the number of trials (vehicle vs. nafadotride+quinpirole: p<0.001; quinpirole 0.3 mg/kg vs. nafadotride+quinpirole: p<0.001; Fig. 2) and incorrect responses to criterion (vehicle vs. nafadotride+quinpirole: p<0.001; quinpirole 0.3 mg/kg vs. nafadotride+quinpirole: p<0.001; Fig. 3).

Reversal phase

Quinpirole Animals treated with quinpirole (0.3 but not 0.1 mg/kg) exhibited a highly significant impairment of performance in the reversal learning phase. Specifically, quinpirole (0.3 mg/kg) significantly increased both trials (vehicle vs. quinpirole 0.3 mg/kg: p<0.001; Fig. 2) and incorrect responses to criterion (vehicle vs. quinpirole 0.3 mg/kg: p<0.001; Fig. 3) compared to vehicle controls.

Table 1 Mean values±SEM of omissions and average latencies to respond (in seconds) during Retention [collapsed retention without reversal and retention (initial phase) preceding reversal] and Reversal Phase

Group	*N*	Omissions retention	Omissions reversal	Latencies retention	Latencies reversal
Vehicle	17	9.06±4.65	19.47±4.81	2.16±0.22	2.43±0.20
Quinpirole 0.1 mg/kg	9	10.67±3.16	24.67±5.24	3.11±0.41	3.93±0.49
Quinpirole 0.3 mg/kg	10	64.5±12.78	47.0±13.48	3.85±0.41	3.66±0.30
Raclopride 0.1 mg/kg	9	88.11±17.1	91.88±21.31	2.65±0.33	2.61±0.20
Nafadotride 0.3 mg/kg	8	8.0±4.57	8.38±2.51	1.59±0.13	2.20±0.18
Nafadotride 1.0 mg/kg	8	3.88±2.89	24.0±4.46	2.44±0.40	2.82±0.36
Raclopride+quinpirole	10	70±11.86	67.20±23.71	3.00±0.32	2.70±0.28
Nafadotride+quinpirole	7	16.14±3.63	18.86±9.49	2.77±0.37	2.78±0.30

Omissions There was a significant main effect of group ($F_{7,70}$=11.86, p<0.001) but no significant main effect of phase or group×phase interaction ($F_{1,70}$=0.63, p=0.43 and $F_{7,70}$=0.72, p=0.66, respectively), *Latencies* there was a significant main effect of group ($F_{7,70}$=5.02, p<0.001) but no significant main effect of phase or group×phase interaction ($F_{1,70}$=3.07, p=0.084 and $F_{7,70}$=1.58, p=0.16, respectively)

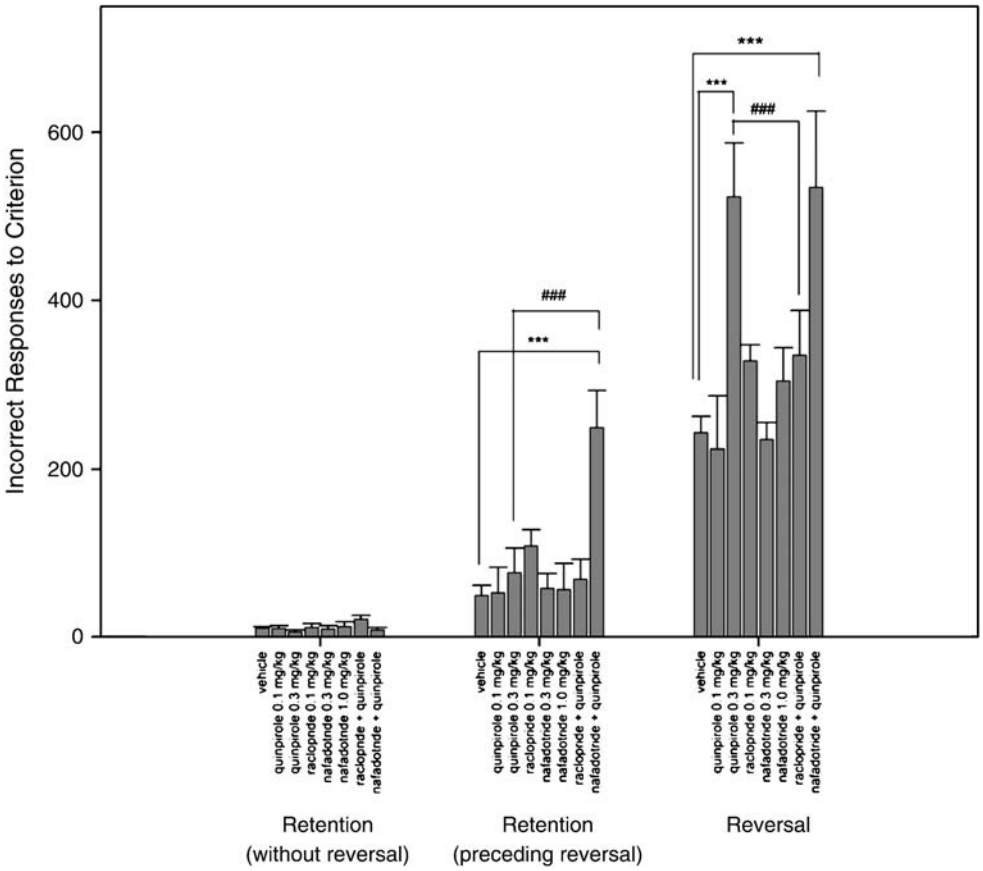

Fig. 3 Incorrect responses of completed trials through (a) the retention phase (without reversal), (b) retention (initial) phase (preceding reversal), and (c) reversal phase. Data are presented as mean values±SEM. There were significant main effects of group and phase ($F_{7,70}$=6.78, p<0.001 and $F_{1,70}$=298.84, p<0.001, respectively) and a significant group×phase interaction ($F_{7,70}$=5.93, p<0.001). *Symbols* denote significant differences following Newman–Keuls post hoc comparisons (***,### p<0.001)

Animals treated with quinpirole (0.3 mg/kg) made significantly more perseverative errors (i.e., <50% correct) than controls in reversal phase (vehicle vs. quinpirole 0.3 mg/kg: p=0.0019; Fig. 4). No differences were noted in learning errors (Fig. 4).

Raclopride Raclopride (0.1 mg/kg), when administered alone, did not alter significantly reversal learning performance. However, raclopride (0.1 mg/kg) when co-administered with quinpirole (0.3 mg/kg) blocked the quinpirole-induced reversal deficit as indicated by the number of trials (quinpirole 0.3 mg/kg vs. raclopride+quinpirole: p=0.012; Fig. 2), incorrect responses to criterion (quinpirole 0.3 mg/kg vs. raclopride+quinpirole: p<0.001; Fig. 3), and perseverative errors (quinpirole 0.3 mg/kg vs. raclopride+quinpirole: p=0.015; Fig. 4).

Nafadotride Administration of nafadotride alone, at any dose, did not affect any measure of reversal performance. Co-administration, however, of nafadotride (1.0 mg/kg) and quinpirole (0.3 mg/kg) had an additive effect on the quinpirole-induced reversal deficit by increasing not only the number of trials (vehicle vs. nafadotride+quinpirole: p< 0.001; Fig. 2), incorrect responses (vehicle vs. nafadotride+quinpirole: p<0.001; Fig. 3), and perseverative errors to criterion (vehicle vs. nafadotride+quinpirole: p=0.036; Fig. 4) compared to vehicle controls but also learning errors (vehicle vs. nafadotride+quinpirole: p<0.001; quinpirole 0.3 mg/kg vs. nafadotride+quinpirole: p<0.001; Fig. 4).

There were no significant differences between groups regarding the number of omitted trials and the latencies to make a response at any stage of the experiment (Table 1).

Discussion

In this study, we have demonstrated that stimulation of the D2/D3 receptors via administration of the D2/D3 receptor agonist quinpirole (0.3 mg/kg) impaired reversal learning performance by increasing the number of trials and incorrect responses to criterion under reversal conditions. This impairment, perseverative in nature, occurred in the absence of significant effects on retention of previous stimulus-reward contingencies, suggesting that it was not a generalized impairment of the ability to retain associative relationships between stimuli and rewards. On the contrary, although the D2/D3 receptor antagonist raclopride, when administered alone, increased the number of trials to criterion in retention preceding the reversal phase, it had no significant effects, neither on the more robust measure of incorrect responses to criterion nor on any other measure of reversal performance. However, raclopride protected

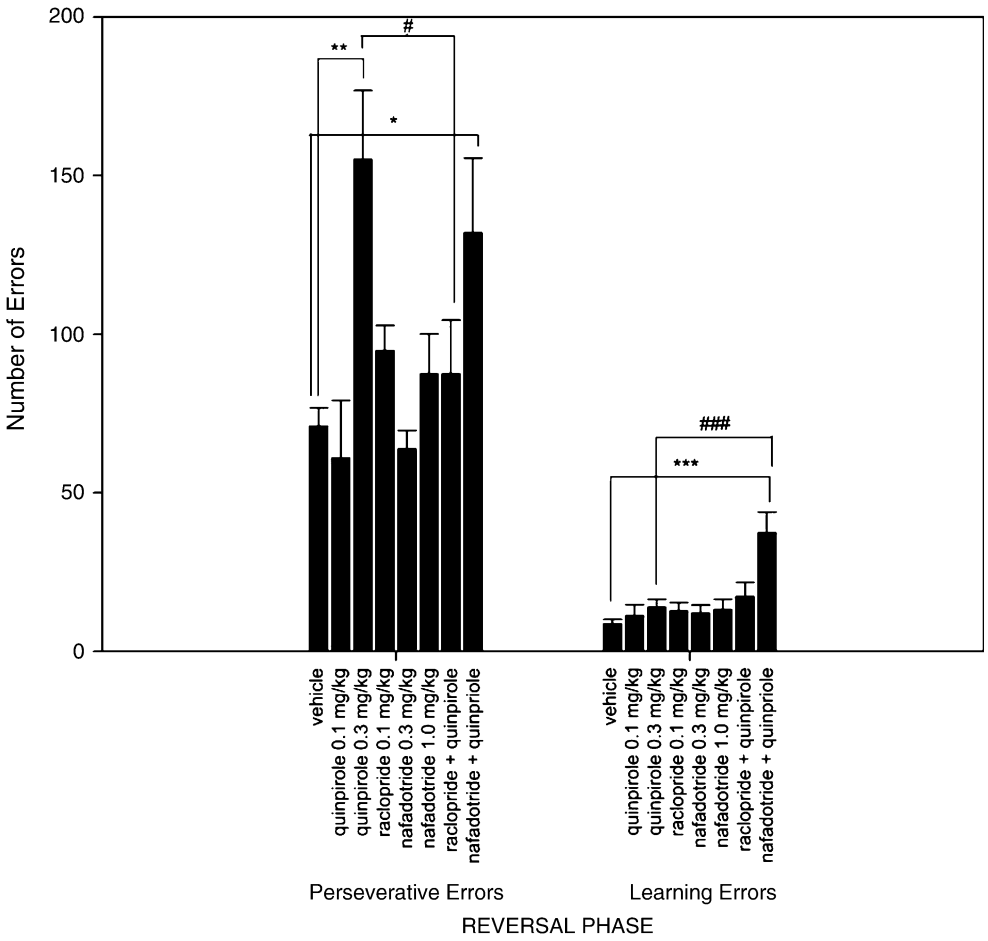

Fig. 4 Mean error scores±SEM of all groups during each learning stage of reversal performance: (a) perseveration (<50% correct) and (b) learning (≥50% correct). Perseveration stage: There was a significant main effect of group ($F_{7,70}$= 5.33, p<0.001). Learning stage: There was a significant main effect of group ($F_{7,70}$=6.60, p< 0.001). *Symbols* denote significant differences following Newman–Keuls post hoc comparisons (***,$^{\#\#\#}p$<0.001; **p<0.01; *,$^{\#}p$<0.05)

against the quinpirole-mediated reversal impairment following their combined administration. Finally, although the selective D3 receptor antagonist nafadotride when administered alone had no significant effects on retention or reversal conditions, it impaired both retention and reversal performance following co-administration with quinpirole by increasing both number of trials and incorrect responses during the retention preceding reversal and reversal phases. Moreover, the reversal impairment resulting from co-administration of nafadotride and quinpirole was general in nature as reflected by increases in both perseverative and learning errors. This finding delineates a dissociation of the persistent deficit from one of new learning in the context of instrumental discrimination: Quinpirole-treated animals were not impaired in learning the new stimulus-reward contingencies once the perseverative tendency had been overridden. In contrast, when quinpirole was co-administered with the selective D3 receptor antagonist nafadotride, animals were impaired in learning the new stimulus-reward contingencies even when they had overridden the perseverative stage. Taken together, our findings might suggest that the D2 receptors play a crucial role in the ability of animals to inhibit a prepotent learned response, whereas D3 receptors are more likely involved in the modulation of the learning process during changing reward contingencies. This interpretation, however, is made with caution given the fact that raclopride is a D2/D3 receptor antagonist and the unavailability of any completely selective D2 receptor antagonists. It could also be argued that the paradoxical finding that selective blockade of D3 receptor via co-administration of nafadotride and quinpirole led to a more severe deficit than the quinpirole-induced impairment is probably due to possible actions of nafadotride on other receptors besides D3.

To our knowledge, there have been no previous studies on the effects of quinpirole on reversal learning and acquisition. Although the present experiments did not investigate the effects of quinpirole on acquisition as drug-free performance in acquisition was the matching criterion for group formation, pilot studies have shown no significant differences on acquisition of the spatial discrimination under quinpirole treatment. However, Jentsch and colleagues have shown that administration of quinpirole to monkeys impaired both acquisition and reversal learning of a three-choice visual discrimination (personal communication). A possible explanation for this discrepancy might be that the non-selective quinpirole deficit observed by Jentsch and colleagues is probably due to the more "taxing" nature of their retention (three-choice visual vs. two-choice spatial discrimination).

D2-like receptors and persistent behavior: implications for obsessive-compulsive disorder

The present findings may be relevant to various neuropsychiatric disorders where inflexible behavior is a feature, such as obsessive-compulsive disorder (OCD). Treatment of patients with OCD with dopaminergic antagonists, especially for those with concurrent psychotic spectrum disorders or with comorbid chronic tic disorders such as Tourette's syndrome, has been shown to have therapeutic effects (McDougle et al. 1994a, b). Moreover, abnormalities in the binding potential of the dopamine D2 receptor in the striatum of OCD patients have also been reported (Denys et al. 2004b).

In the present study, quinpirole affected neither the number of incorrect responses to criterion nor the late phases (i.e., "learning" phase) of reversal learning. However, it significantly increased number of trials and incorrect responses in the reversal learning phase. Perseverative errors in the early reversal stage were also affected. The present results are consistent with results obtained with other animal models of persistent behavior that have linked changes in the dopaminergic system to compulsive-like behaviors. Prolonged, but not acute administration, of quinpirole induces directional persistence in the T-maze (Kontis et al. 2008), "surplus" lever pressing in the signal attenuation model (Joel et al. 2001), and compulsive behavior in rats without evidence of stereotypy (Szechtman et al. 1998, 2001). It has been proposed that sensitization to quinpirole produces these effects by affecting post-synaptic D2 receptors, resulting in a suppression of basal ganglia function (Eilam et al. 1989; Sullivan et al. 1998). This suppression, together with excessive cortical stimulation, has been suggested to underlie compulsive behavior (Modell et al. 1989). In these studies, the effects of quinpirole emerged gradually (following 10 days of administration) rather than acutely (perhaps due to a sensitization effect). In the present study, the quinpirole-induced differences were evident from the second day of administration of the drug. This discrepancy could probably be attributed to the fact that the effects of quinpirole on these models were tested after animals had achieved a baseline performance on the test behavior. In our task, animals are exposed to the reversal learning task when drugs are administered without having any previous experience of the behavioral procedure.

Given that quinpirole is a D2/D3 receptor agonist, the dopaminergic contribution to persistent behavior in reversal learning appears to involve either D2 or D3 receptors or both. The contribution of a D3 receptor involvement could not be excluded on the basis of the findings of Szechtman et al. (1998, 2001). The D3 receptor has also been implicated in the pathogenesis of OCD based on genetic comorbidity between OCD and Tourette's syndrome (Pauls et al. 1986): Tourette's syndrome has been associated with a polymorphism in the dopamine D3 receptor gene. However, after evaluation of the frequency of this polymorphism in OCD patients and controls, Catalano et al. (1994) concluded that there is no association with OCD and the D3 receptor gene. This finding was also supported by the suggestion of an association of OCD with the D4 receptor gene (Billett et al. 1998). Although the possibility of a D3 receptor contribution to persistent behavior and to OCD pathogenesis could not be definitely excluded at this point, it does not seem to be very likely. Moreover, in the present study, we showed that selective blockade of the D3 receptor by nafadotride did not protect against the quinpirole-induced deficit in reversal learning. In fact, it led to a more generalized impairment, not only in the retention condition but also in the late phase (learning phase) of reversal learning. This effect suggests that D2 receptors are more likely to play a crucial role in the ability of animals to inhibit prepotent learned responses (persistence), whereas D3 receptors are possibly implicated in modulating the learning process during changing reward contingencies.

In summary, our data indicate distinct role for D2 and D3 receptors in the capacity to modify behavior flexibly in the face of environmental change. In addition, the present results may be relevant to the pathophysiology of OCD, suggesting a potential role for D2 receptors when considering possible treatment strategies for this disorder.

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