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Effects of terguride, ropinirole, and acetyl-L-carnitine on methamphetamine withdrawal in the rat

Michael E. Hoefer [1], Steven J. Voskanian [1], George F. Koob, Luigi Pulvirenti *

Molecular and Integrative Neurosciences Department, SP30-2400, The Scripps Research Institute, 10550 North Torrey Pines Road, La Jolla, CA 92037, USA

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Abstract

Withdrawal from psychostimulants, including methamphetamine, induces a depressive state associated with lethargy, dysphoria, *hyperphagia* and psychomotor retardation. Previous work with repeated administration of amphetamine in rats has shown that amphetamine withdrawal produces decreased motivation to work for a non-drug reward, and this withdrawal is reversed by administration of a dopamine partial agonist. The purpose of the present study was to examine decreased motivation to work for a non-drug reward during methamphetamine withdrawal and explore the effects of a dopamine agonist, dopamine partial agonist, and indirect monoamine agonist on methamphetamine withdrawal. During withdrawal from repeated methamphetamine administration, rats showed reduced responding for a sweet solution in a progressive-ratio schedule of reinforcement, and this effect was significantly more pronounced than previously observed with amphetamine. Repeated systemic treatment with the dopamine partial agonist terguride (0.2 and 0.4 mg/kg, i.p., twice daily), the full dopamine agonist ropinirole (1 mg/kg, i.p., twice daily), and acetyl-L-carnitine (60 and 100 mg/kg, i.p.), a compound with a potential antidepressant effect, during methamphetamine withdrawal restored responding for the sweet solution, suggesting that these drugs may represent potential therapeutic strategies for the treatment of methamphetamine addiction during the withdrawal phase.

Keywords: Dopamine; Partial agonist; Methamphetamine; Acetyl-L-carnitine; Withdrawal; Drug addiction

1. Introduction

Several studies have shown that withdrawal from exposure to high doses of amphetamines causes psychomotor retardation in both humans and laboratory animals (Pulvirenti and Koob, 1993; Newton et al., 2004), and in rodents an increase of intracranial self-stimulation (ICSS) thresholds has been reported, interpreted as a functional deficit of the brain reward system (Wise and Munn, 1995; Markou and Koob, 1991). Withdrawal from repeated exposure to amphetamine also has been shown to decrease operant responding for a natural reward (sucrose solution) on a progressive-ratio schedule of reinforcement for a period of up to five days (Barr and Phillips, 1999). The progressive-ratio schedule is a behavioral paradigm specifically tailored to reflect a measure of the relative reinforcing properties of a self-administered drug (Bedford et al., 1978; Richardson and Roberts, 1996) as well as palatable reinforcers (Hodos, 1961) and ICSS (Hodos, 1965). The reduction in operant responding observed during amphetamine withdrawal demonstrates a decreased motivation to respond for a natural reward and may be interpreted as evidence of decreased functioning of the brain reward system or the presence of a reward deficit state (Orsini et al., 2001; Barr and Phillips, 1999). One hypothesis under test in the present study is that repeated exposure to methamphetamine will have similar effects as amphetamine in producing a withdrawal response as measured by decreased operant responding for a natural reward (i.e., sucrose solution).

Although several studies have shown that the mesolimbic dopamine system appears to be critically involved in the development of psychostimulant dependence (Wise and Bozarth, 1987; Wise, 1996; Koob and Le Moal, 1997), only limited clinical efficacy has been reported with the use of dopamine agonists, dopamine antagonists, and tricyclic antidepressants in the treatment of psychostimulant dependence (Wesson and Smith, 1978; Mello and Negus, 1996; Cretzmeyer et al., 2003).

* Corresponding author. Tel.: +1 858 784 7144; fax: +1 858 784 7405.
E-mail address: lpulvirenti@scripps.edu (L. Pulvirenti).
[1] M.E.H. and S.J.V. equally contributed to this work.

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The limited efficacy of direct dopamine agonist drugs to treat the core symptoms of psychostimulant dependence and the recent successes of the partial opioid agonist buprenorphine in the treatment of opiate dependence (Davids and Gastpar, 2004) has generated a great deal of interest in the use of partial dopamine agonists for the treatment of psychostimulant dependence. Partial dopamine agonists bind to the dopamine receptor with high affinity and low intrinsic activity (Hoyer and Boddeke, 1993), and thus exhibit a modulatory role on dopaminergic function (Clark et al., 1991; Svensson et al., 1991; Pulvirenti and Koob, 1994; Wacan et al., 2006). The partial dopamine agonists terguride and SDZ 208–911 have been shown to work similarly to classic dopamine antagonists in rats trained to self-administer cocaine and amphetamine under different schedules of reinforcement (Pulvirenti and Koob, 1994; Pulvirenti et al., 1998; Izzo et al., 2001), and terguride was able to counteract the reward deficit observed during amphetamine withdrawal as measured by restored operant responding for a sweet solution on a progressive-ratio schedule (Orsini et al., 2001). A second hypothesis under test in the present study is that terguride, a dopamine partial agonist, will reverse the effects of methamphetamine withdrawal.

Acetyl-L-carnitine is the acetyl ester of carnitine, an endogenous component present in high concentrations in the brain, and which has been involved in a number of neural functions (Virmani et al., 2003). Evidence suggesting the potential clinical use of acetyl-L-carnitine in depression derives from different lines of investigation. Acetyl-L-carnitine exerts an indirect effect on dopamine neurotransmission (Harsing et al., 1992) and appears to possess antidepressant effects in some but not all experimental conditions (Gambarana et al., 2001; Tolu et al., 2002). In preclinical studies, acetyl-L-carnitine reduced the immobility time in Porsolt's forced swimming test (Pulvirenti et al., 1990) both after acute and chronic treatment. In addition, altered lipid metabolism as well as membrane dynamics, two parameters significantly modified by acetyl-L-carnitine, are thought to be impaired in various forms of affective illness (for review, see Pettegrew et al., 2000), and acetyl-L-carnitine shares with lithium similar effects on the breakdown of phosphoinositides and choline (Pettegrew et al., 2000). These preclinical observations also are supported by a number of clinical studies which have shown that acetyl-L-carnitine has significant antidepressant effects in different patient populations (Tempesta et al., 1987; Villardita et al., 1983; Nasca et al., 1989; Fulgente et al., 1990; Garzya et al., 1990; Bianchetti et al., 2003). Although its precise mechanism of action has yet to be elucidated, repeated treatment with acetyl-L-carnitine has been shown to increase extracellular dopamine and serotonin levels in the shell of the nucleus accumbens, while preventing the development of avoidance deficit induced by acute unavoidable stress (Tolu et al., 2002). This effect is shared by antidepressants such as imipramine and fluoxetine (Gambarana et al., 2001). Acetyl-L-carnitine also increases dopamine efflux in the corpus striatum (Harsing et al., 1992). Acetyl-L-carnitine also benefits from a low side-effect profile and wide therapeutic index (De Grandis and Minardi, 2002; Bonavita, 1986). A third hypothesis under test in the present study is that acetyl-L-carnitine may have efficacy in reversing methamphetamine withdrawal.

Thus, the overall aim of the present study was to investigate the potential therapeutic role of the partial dopamine agonist terguride and the full dopamine agonist ropinirole in the reduced motivation to seek a natural reinforcer following abrupt interruption of a binge-like schedule of methamphetamine administration. The potential role of acetyl-L-carnitine also was explored following a binge-like schedule using lower doses of methamphetamine.

2. Material and methods

2.1. Animals

Eighty-seven male Wistar Rats (Charles River, Kingston, NY) were housed on a reversed light/dark cycle (lights on 10 PM to 10 AM), and body weights were 300–350 g at the start of each experiment, except for one experiment where starting weights were 250–300 g. Rats were group housed 2–3 per cage, and testing was performed nightly between 6 PM and 10 PM. All procedures met the guidelines of the *National Institutes of Health Guide for the Care and Use of Laboratory Animals* (NIH Publication number 85–23, revised 1996) and the *Principles of Laboratory Animal Care* as well as the European Community guidelines for the use of experimental animals. All procedures were approved by the Institutional Animal Care and Use Committee of The Scripps Research Institute.

2.2. Operant procedure

Subjects were given an initial 48 h exposure to a sweet solution (3% sucrose; Fisher Scientific) and 0.125% saccharin (Sigma-RBI, St. Louis, MO). All rats then were water deprived for 20 h daily before being placed in standard operant chambers (Coulbourn Instruments, Allentown, PA). Operant chambers were housed in sound-attenuated storage cubicles, and syringe pumps (Razel Scientific Instruments, Stamford, CT) were calibrated to dispense 0.1 ml of sweet solution per reinforcement. A single retractable lever mounted next to a stainless steel drinking cup was utilized for delivery of sweet solution, and a computer controlled data collection and delivery of the sweet solution. There was a timeout period of 5 s after each reinforcement, where subsequent responding produced no effect.

Subjects first were trained on a fixed-ratio schedule of reinforcement for 1 h daily for 10 days under the following design: Day 1–3 (FR1), Day 4–6 (FR3), Day 7–10 (FR10). Two animals that did not acquire self-administration of the sweet solution were not included in the study. After fixed-ratio training, subjects were placed on a progressive-ratio schedule for the sweet solution for 2 h daily, and the response requirements necessary to receive a single reinforcement increased according to the following progression: $5e^{0.2(\text{total rewards}+1)}-5$. This resulted in the following response requirement progression: 1, 2, 4, 6, 9, 12, 15, 20, 25, 32, 40, 50, 62, 77, 95, 118, 145, 178, 219, 268, etc. The break point was defined as the last ratio attained by the rat prior to a 60 min period during which a ratio

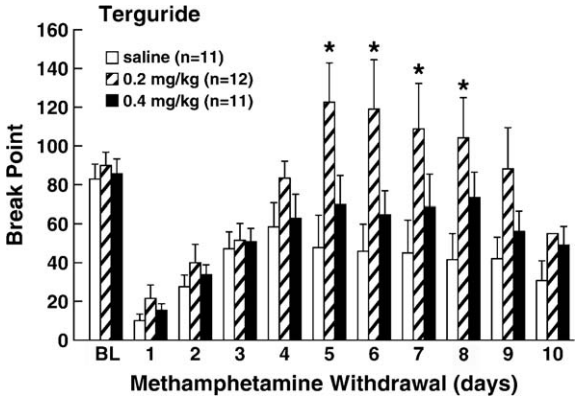

Fig. 1. Effect of repeated treatment with terguride (0.2 and 0.4 mg/kg, i.p., twice daily for five days) on progressive-ratio responding (2 h nightly) for a sweet solution (3% sucrose + 0.125% saccharin) during methamphetamine withdrawal. Break points were defined as the highest number of lever presses subjects performed for a single reinforcement ± SEM (n = 10–12/group). Terguride reversed the decrease in progressive-ratio responding for a sweet solution during methamphetamine withdrawal, and a significant difference between terguride at the dose of 0.2 mg/kg and saline was reached at Day 5 (* p < 0.01), Day 6 (* p < 0.05), Day 7 (* p < 0.05), and Day 8 (* p < 0.05) of methamphetamine withdrawal.

was not completed. Progressive-ratio sessions were conducted at 6 PM. After stable response was maintained for three consecutive days (±10% with a break point >40) this was taken as baseline and rats were injected in the intraperitoneal cavity (i.p.) with methamphetamine three times per day (9 AM, 5 PM, 12 AM), starting with a dose of 1 mg/kg/inj and increasing by 1 mg/kg/inj for 10 injections, three injections per day, reaching a total cumulative dose of 55 mg/kg over four days (Experiments 1 and 2) and 40 mg/kg total cumulative dose (Experiment 3) (Barr and Phillips, 1999; Segal and Kuczenski, 1977). During methamphetamine treatment, animals were housed with free exposure to food and water.

In the first study, at the end of methamphetamine treatment, rats were treated i.p. twice daily (9:30 AM and 9:30 PM) for five days with either saline (n = 11) or terguride at the dose of 0.2 mg/kg (n = 12) or 0.4 mg/kg (n = 11), and progressive-ratio responding was monitored for 10 days. In a second study, rats were treated i.p twice daily for five days with either saline (n = 14) or ropinirole at a dose of 1 mg/kg (n = 10), and progressive-ratio responding was monitored for 10 days.

In experiments which started during the conduction of the studies described in the present manuscript and currently in progress, rats were trained to self-administer methamphetamine to explore whether, upon exposure to longer time schedules of methamphetamine self-administration, a withdrawal syndrome measurable as reduction of motivation to obtain a natural reinforcer emerged. It was observed that in those conditions the amount of methamphetamine self-administered was lower compared to the amounts used in Experiments 1 and 2 of the present study. In the third study, the dose of methamphetamine was therefore adjusted accordingly. Rats were treated i.p. once a day (9:30 PM) with either saline (n = 9) or acetyl-L-carnitine at a dose of 60 mg/kg (n = 10) and 100 mg/kg (n = 10) for 10 days, and progressive-ratio responding was monitored for 10 days. Here, rats were exposed to a binge-like schedule of methamphetamine withdrawal using lower doses of the drug. Methamphetamine was administered 10 times over four days, as described above, except that dose was escalated from 1 to 5 mg/kg for the first five injections as in previous experiments, then maintained at 5 mg/kg for the last five injections, reaching a total cumulative dose of 40 mg/kg over 10 days. In experiments which started during the conduction of the studies described in the present manuscript and currently in progress, rats were trained to self-administer methamphetamine to explore whether, upon exposure to longer time schedules of methamphetamine self-administration, a withdrawal syndrome measurable as reduction of motivation to obtain a natural reinforcer emerged. It was observed that in those conditions the amount of methamphetamine self-administered was lower compared to the amounts used in Experiments 1 and 2 of the present study. In the third experiment the dose of methamphetamine was therefore adjusted accordingly.

2.3. Drugs

Ropinirole, and methamphetamine hydrochloride (Sigma-RBI, St. Louis, MO) and acetyl-L-carnitine (a generous gift of Sigma-Tau Pharmaceuticals, Pomezia, Italy), were dissolved in saline. Terguride (Research Biochemicals International, Natick, MA) was dissolved in distilled water and 1 N HCl, and the pH was adjusted to 5.5 with 1 M NaOH. All drugs were injected in a volume of 1.0 ml/kg.

2.4. Data analysis

Using SPSS software designed for Windows, a two-way (*Dose* vs. *Time*) mixed ANOVA (with time as the repeated measure) was performed on the data for each experiment. If significance was reached, simple effects analyses were performed to determine differences between dose groups at

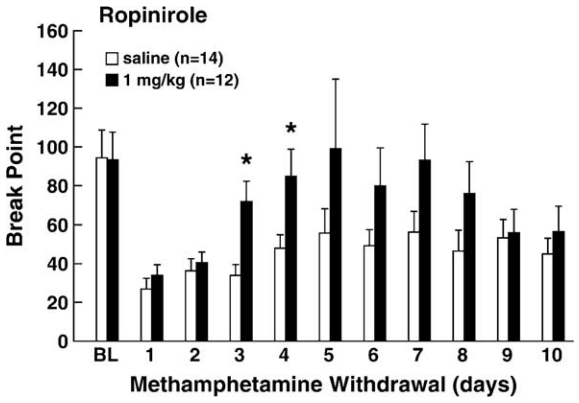

Fig. 2. Effect of repeated treatment with ropinirole (1.0 mg/kg, i.p., twice daily for five days) on progressive-ratio responding (2 h nightly) for a sweet solution (3% sucrose + 0.125% saccharin) during methamphetamine withdrawal. Break points were defined as the highest number of lever presses subjects performed for a single reinforcement ± SEM (n = 10–14/group). Ropinirole reversed the decrease in progressive-ratio responding for a sweet solution in rats during methamphetamine withdrawal, and a significant difference between ropinirole and saline was reached at Day 3 (* p < 0.01) and Day 4 (* p < 0.05) of methamphetamine withdrawal.

each day of methamphetamine withdrawal. When appropriate, post hoc analyses for between-subjects comparisons were performed using Bonferroni-corrected paired *t*-tests.

3. Results

3.1. Effects of terguride on progressive-ratio responding for a sweet solution during methamphetamine withdrawal

Fig. 1 shows the effects of treatment with terguride on progressive-ratio responding for a sweet solution during methamphetamine withdrawal. ANOVA revealed a main effect of Time ($F_{10,310}=14.035$, $p<0.001$), a main effect of Drug ($F_{2,31}=3.95$, $p<0.05$) and a significant Drug × Time interaction ($F_{20,310}=2.16$, $p<0.01$). Post hoc comparison revealed a significant difference between post-methamphetamine and baseline responding at Days 1,2,7,8,9 and 10 in the saline group, thus showing that the schedule of administration using increasing doses 1–10 mg/kg significantly attenuates responding throughout the 10-day observation period, with the exception of days 3–6. Post hoc comparison also revealed a significant difference between terguride at a dose of 0.2 mg/kg and saline on Day 5 ($p<0.01$), Day 6 ($p<0.05$), Day 7 ($p<0.05$), and Day 8 ($p<0.05$).

3.2. Effects of ropinirole on responding for a sweet solution during methamphetamine withdrawal

Fig. 2 shows the effects of treatment with ropinirole at a dose of 1.0 mg/kg on progressive-ratio responding for a sweet solution during methamphetamine withdrawal. ANOVA revealed a main effect of Drug ($F_{1,22}=3.453$, $p<0.05$), a main effect of Time ($F_{10,\ 220}=9.52$, $p<0.01$) and no significant Drug × Time interaction ($F_{20,220}<1.6$). Post hoc analysis showed a significant difference at Day 3 ($p<0.01$) and Day 4 ($p<0.05$), indicating that ropinirole restored responding for a natural reward during methamphetamine withdrawal faster than saline.

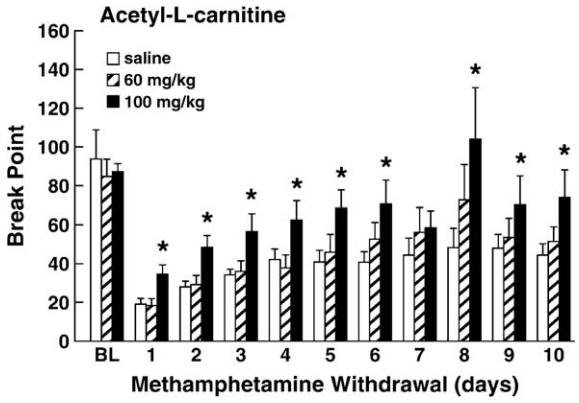

Fig. 3. Effects of acetyl-L-carnitine on mild methamphetamine withdrawal. Methamphetamine was administered 10 times with the dose escalated from 1–5 mg/kg for the first five injectons as in previous experiments, then maintained at 5 mg/kg for the last five injections. *$p<0.05$ vs. saline. Acetyl-L-carnitine reversed the decrease in progressive-ratio responding for a sweet solution during methamphetamine withdrawal, and a significant difference between the dose of 100 mg/kg and saline was reached at all days of methamphetamine withdrawal (with the exception of Day 7).

3.3. Effects of acetyl-L-carnitine on responding for a sweet solution during methamphetamine withdrawal at lower dosage

Fig. 3 shows the effects of treatment with acetyl-L-carnitine (0, 60, and 100 mg/kg) on progressive-ratio responding for a sweet solution during methamphetamine withdrawal. ANOVA revealed a main effect of Time ($F_{10,\ 260}=13.82$, $p<0.01$), a main effect of Drug ($F_{2,26}=3.23$, $p<0.05$) and no significant Drug × Time interaction ($F_{20,260}<1$). Post hoc comparison revealed a significant difference between post-methamphetamine and baseline responding at all Days 1–10 in the saline group, thus showing that the schedule of administration using increasing 1–5 mg/kg doses significantly attenuates responding throughout the 10-day observation period. Post hoc analysis revealed a significant difference between acetyl-L-carnitine at the dose of 100 mg/kg and saline ($p<0.05$) at all days of methamphetamine withdrawal, with the exception of day 7.

4. Discussion

The results of the studies presented here demonstrate that withdrawal from repeated high doses of methamphetamine causes a substantial decrease in responding for a sweet solution reward on a progressive-ratio schedule of reinforcement. This decrease in responding for a sweet solution may reflect a lack of motivation to seek a naturally rewarding stimulus and was similar to that previously observed with other psychostimulant drugs and may be a useful model for the dysregulation of the reward-seeking state associated with psychostimulant withdrawal.

Compared to amphetamine, withdrawal from repeated doses of methamphetamine induced a more profound and persistent decrease in responding for a sweet solution (Barr and Phillips, 1999; Orsini et al., 2001). In previous studies, the motivation to seek the reinforcer, measured as maximum number of responses performed (break point) was reported as a decrease of approximately 35% of pre-amphetamine values and lasted for approximately the first 48–72 h of amphetamine withdrawal (Barr and Phillips, 1999; Orsini et al., 2001). Following a sustained schedule of methamphetamine administration, both at the higher and lower doses used in the present study, a decrease of approximately 70% compared to pre-binge levels was observed, and values did not recover to pre-methamphetamine levels throughout the 10-day observation period. It is possible that the more profound and persistent withdrawal effects elicited by methamphetamine play a significant role in its higher abuse potential and higher relapse rates among users (Murray, 1998). Both a total dose of 55 mg/kg/day (experiments 1 and 2) and a total dose of 40 mg/kg/day (experiment 3) produced similar results, suggesting that even lower doses of methamphetamine may be effective in producing a hedonic-like withdrawal effect.

Terguride produced the peak of its effect at Days 5–8 (i.e., after termination of terguride administration, Days 1–5). During this time frame the dose of 0.4 mg/kg also appeared to increase

responding for the sweet solution compared to saline, but this did not reach statistical significance. Stimulation of dopamine D_2 receptors during the initial withdrawal phase appears to represent, therefore, a potential strategy for restoring motivation to seek natural reinforcers. The restoration in responding expressed itself fully during the following days, even in the absence of drug treatment, in the design used in the present study.

Terguride also restored responding for a sweet solution during amphetamine withdrawal in a previous study (Orsini et al., 2001). A significant effect of terguride was observed on the second day of withdrawal, while in the present study a significant effect of terguride was not detected until the fifth day. In addition, terguride was effective in the present study at the dose of 0.2 mg/kg, while the dose of 0.4 mg/kg was ineffective. A U-shaped curve in behavioral or biochemical studies has not been reported to our knowledge with terguride in normal rats or in studies using rats with dopamine depletion. Although speculative at present, it is possible that after methamphetamine treatment the higher dose of terguride may start showing antagonistic effects at the dopamine receptor and thus partially reducing its "corrective" action as a dopamine agonist. It is also interesting that terguride was effective at the dose of 0.4 mg/kg, but not 0.2 mg/kg during amphetamine withdrawal (Orsini et al., 2001). The reason for these differences remains unclear, but one possibility is the low level of amphetamine withdrawal observed in the Orsini et al. (2001) paper which only lasted for 72 h, thus making a direct comparison impossible. Terguride also appeared to show a tendency to increase the maximum FR above baseline at days 5–8, although this did not reach statistical significance. This is not a novel observation since a similar effect had been noted in amphetamine withdrawal using terguride (Orsini et al., 2001). In that study an apparent increase of maximum FR compared to baseline was observed during the last two days of treatment with terguride but, again, it did not reach statistical significance. Half-life of terguride in the rat is 50 min (Krause and Humpel, 1988) and pharmakinetics alone is unlikely to explain effects of terguride 72 hours after administration. Half-lives of ropinirole and acetyl-L-carnitine are also relatively short, namely 30 min and 4.2 h respectively (Ramji et al., 1999; Kwon and Chung, 2004), and are unlikely to explain differences in the results observed.

The fact that ropinirole, a dopamine full agonist, was also able to restore responding for the natural reinforcer during methamphetamine withdrawal provides further evidence supporting the hypothesis that withdrawal from methamphetamine is associated with a decrease in dopamine neurotransmission. Ropinirole appeared to restore responding for the natural reward more rapidly than terguride since statistical significance was reached after 3 days of administration, but the effects also appeared to dissipate more rapidly following the termination of the treatment. The reason for this difference cannot be simply accounted for by difference in pharmacokinetics, and further studies will be needed to confirm whether partial agonists possess longer-lasting effects on psychostimulant withdrawal. It is also noteworthy that ropinirole has been shown to be effective as an adjunct medication in treatment-resistant depression (Cassano et al., 2005), further supporting the hypothesis that drugs acting as dopamine agonists may serve as candidates for the depressive syndrome associated with methamphetamine withdrawal. Indeed, terguride is thought to be effective during psychostimulant withdrawal because of its intrinsic agonistic activity, which is evident in conditions of dopamine depletion, such as during amphetamine and methamphetamine withdrawal.

Acetyl-L-carnitine significantly reduced the severity and duration of methamphetamine withdrawal at the dose of 100 mg/kg using a schedule of lower doses of methamphetamine exposure. The precise biochemical mechanism underlying the effects of acetyl-L-carnitine remains at present elusive. However, acetyl-L-carnitine, similar to other antidepressants, has been shown to increase dopamine release in the brain (Harsing et al., 1992), to prevent the behavioral deficit observed following inescapable stress (Gambarana et al., 2001; Tolu et al., 2002), and to possess effects similar to antidepressant drugs in various animal models of depression (Pulvirenti et al., 1990; Tolu et al., 2002; Masi et al., 2003). Acetyl-L-carnitine also has been shown to prevent methamphetamine-induced neurotoxicity (Virmani et al., 2003). On the basis of the available evidence, it is tempting to speculate that the effects of acetyl-L-carnitine reported here may be at least partially due to the previously reported activation of dopamine neurotransmission in mesolimbic areas, an effect which may be dependent upon activation of 5-HT_{1A} receptors (Tolu et al., 2002). It is possible, however, that other components also contributed to the effects reported here, including protection from methamphetamine toxicity (Virmani et al., 2003). It is indeed possible that a short-term methamphetamine withdrawal effect lasting 24–48 h is superimposed on changes in responding due to methamphetamine-induced neurotoxicity. The fact that responding did not fully recover to baseline levels even after 10 days might be consistent with this. Indeed acetyl-L-carnitine restored responding at later time points (day 10 post-methamphetamine) and it also possesses anti-neurodegenerative effects. However fascinating, the hypothesis that anti-neurodegenerative effect might play a role remains at present speculative. Terguride and ropinirole were ineffective at later time points but they were also administered for a shorter time (5 days). In the present design with purely behavioral measures it is not possible to specifically isolate a neurodegenerative component in the design used. A possible neurotoxic effect, which may not be a confounding effect but an intrinsic component of methamphetamine action (also in humans) will need to be investigated in future studies. In addition, the observation period was limited to 10 days in the present study. The results encourage longer observation periods in future investigation.

Withdrawal from psychostimulants may cause psychomotor retardation per se (Paulson et al., 1991; Pulvirenti and Koob, 1993), which itself could account for the reductions in responding observed during methamphetamine withdrawal. However, it is unlikely that psychomotor retardation alone can explain the reduced motivation to seek a reinforcer. For example, rats have been shown to maintain high levels of responding for ICSS during amphetamine withdrawal (Markou and Koob, 1992). Additionally, the pattern of responding

observed by Barr and Phillips (1999) during amphetamine withdrawal did not correspond to the pattern of behavior observed following a locomotor-retarding dose of a neuroleptic (Fowler and Mortell, 1992). In the present study, subjects were given a full hour to complete responding for a final reinforcement and were not required to respond at levels that typically require an exhaustive amount of energy.

Additionally, it is important to note that suppression of motivated responding for a natural reinforcer such as food, water or sweetened solutions may be due to a devaluation of the rewarding properties of the reinforcer, but may also be due to a variety of other nonspecific factors, including general malaise associated with drug withdrawal. An alternative explanation, therefore, cannot be excluded a priori since the measure used is not a direct assessment of reward deficit only. Also, hyperphagia is certainly one component of clinical withdrawal from psychostimulants but it is not commonly observed during the first days of withdrawal where exhaustion is the predominant clinical feature.

Exposing animals to dopamine agonists leads to long-lasting alterations in dopaminergic function. These have most often been assessed as increased locomotor activity and enhanced dopamine overflow in brain limbic areas and are defined as behavioral sensitization, a progressive, enduring enhancement of behaviors that develops following repeated stimulant administration (Vezina, 2004; Richtand et al., 2001). It appears to be mediated in part by dopaminergic pathways and may reflect a number of psychiatric conditions including the development of psychosis. Biochemical and behavioral evidence also indicates that a relationship may exist between the sensitization of midbrain dopamine neurons and various aspects of psychostimulant exposure including drug self-administration, craving and reinstatement (Vezina, 2004). Dopamine D1, D2 and D3 receptors are thought to participate in behavioral sensitization (Richtand et al., 2001). While no specific information is available for the full agonist ropinirole, the partial agonist terguride did not appear to produce sensitization per se in the rat (Sibole et al., 2003). Although the efficacy of terguride in the present study suggests that sensitization does not play a major role in the restored responding for a natural reinforcer observed here, further studies using dopamine agonists and full agonists will be needed to clarify whether the development of sensitization plays a significant role using multiple administration of dopamine agonists in psychostimulant withdrawal.

In summary, the present results show that repeated methamphetamine administration produces a robust motivational withdrawal syndrome reflected in decreased responding for a highly palatable sweet solution. A dopamine agonist, partial agonist, and indirect agonist reversed this withdrawal, suggesting an important role for loss of dopamine function in methamphetamine withdrawal and a potential therapeutic target.

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