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Prolonged Attenuation of the Reinforcing Strength of Cocaine by Chronic *d*-Amphetamine in Rhesus Monkeys

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Chronic treatment with the indirect dopamine agonist *d*-amphetamine can reduce cocaine use in clinical trials and, in preclinical studies in laboratory animals, attenuates daily cocaine self-administration. The present study extended previous results to conditions designed to reflect a more clinically relevant experience of cocaine exposure and *d*-amphetamine treatment. Each morning, monkeys pressed a lever to receive food pellets under a 50-response fixed-ratio schedule of reinforcement. After determining a dose-response curve for cocaine (0.003–0.56 mg/kg per injection, i.v.) under a progressive-ratio (PR) schedule of reinforcement in the evening, cocaine self-administration sessions were suspended and *d*-amphetamine (0.01–0.056 mg/kg/h, i.v.) was administered continuously for at least 24 days, except during cocaine self-administration sessions, which were conducted using the PR schedule once every 8 days. When a persistent decrease in self-administration was observed, the cocaine dose-effect curve was redetermined. Cocaine- and food-maintained responding were also examined after discontinuation of *d*-amphetamine. Although individual differences in sensitivity were observed, *d*-amphetamine produced selective, qualitatively similar decreases in the reinforcing strength of cocaine in all monkeys that persisted at least 4 weeks. Moreover, cocaine dose-effect curves were shifted downward and/or to the right. For 2 weeks following discontinuation of *d*-amphetamine treatment, the reinforcing strength of cocaine varied within and across individuals, however, on the whole no increased sensitivity was apparent. These data provide further support for the use of agonist medications for cocaine abuse, and extend the conditions under which such treatment is successful to those that incorporate clinically relevant patterns of cocaine use and drug treatment.
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INTRODUCTION

Cocaine abuse persists as a major public health problem, for which no pharmacotherapy has proven to be sufficiently effective (Vocci and Ling, 2005). The success of methadone and nicotine replacement therapies in the treatment of opiate and nicotine addiction, respectively, has encouraged efforts to develop an indirect dopamine agonist medication to treat stimulant abuse (Grabowski *et al*., 2004a; Rothman and Glowa, 1995; Herin *et al*., 2010). Although efforts are ongoing to develop novel dopamine indirect agonists for this purpose (eg, Carroll *et al*., 1999; Platt *et al*., 2002; Lile and Nader, 2003; Rothman *et al*., 2005; Negus *et al*., 2007), several clinical studies have supported the safety and efficacy of *d*-amphetamine as a treatment for cocaine dependence (eg, Fleming and Roberts, 1994; White, 2000), including three double-blind, placebo-controlled studies that used a sustained-release preparation (Grabowski *et al*., 2001, 2004b; Shearer *et al*., 2003).

Chronic treatment with *d*-amphetamine has also been found to decrease cocaine self-administration in laboratory animals under several conditions, including progressive-ratio (PR) and second-order schedules of reinforcement, as well as food-cocaine choice procedures (Negus, 2003; Negus and Mello, 2003a, b; Chiodo *et al*., 2008; Chiodo and Roberts, 2009; Czoty *et al*., 2010). Importantly, *d*-amphetamine was administered chronically in these studies. Using chronic drug administration in preclinical models is critical not only because chronic administration better reflects ultimate clinical use, but also because acute administration of *d*-amphetamine can enhance the reinforcing effects of cocaine in self-administration and reinstatement paradigms (Gerber and Stretch, 1975; de Wit and Stewart, 1983; Barrett *et al*., 2004; DC Roberts, personal communication). Under other conditions in which acute *d*-amphetamine was reported to decrease cocaine self-administration, comparable reductions in food-maintained responding were

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observed (Foltin and Evans, 1999; Mansbach and Balster, 1993). The latter findings highlight the importance of concurrently studying behavior reinforced by a non-drug stimulus, disruptions of which may indicate potential for side effects that decrease compliance.

The present study was designed to further characterize the effects of chronic *d*-amphetamine treatment on cocaine self-administration under a PR schedule of reinforcement in rhesus monkeys, using a regimen of access to cocaine and an approach to *d*-amphetamine treatment designed to better reflect the clinical experience in several ways. First, unlike other studies, access to cocaine was suspended during *d*-amphetamine administration to model a treatment scenario in which an addict is able to refrain from using cocaine during a brief, initial period due to incarceration, hospitalization, or temporarily enhanced motivation to quit. Rather than assessing cocaine self-administration daily, the reinforcing strength of cocaine was assessed only once every 8 days, permitting 7 consecutive days of *d*-amphetamine treatment in the absence of cocaine. Second, food-reinforced responding was monitored daily to exclude the possibility that any observed decreases in cocaine-reinforced responding were due to behavioral disruption that could indicate a likelihood of side effects in a clinical population. Third, rather then employing a group design in which all monkeys received the same *d*-amphetamine doses for predetermined lengths of time, the *d*-amphetamine dose was adjusted for each subject based on the observed effect (or lack thereof) on cocaine-reinforced responding. If no decrease in cocaine self-administration was observed within 4 weeks, or if tolerance had developed to an initial decrease, the *d*-amphetamine dose was increased until decreases in self-administration were observed. This approach was implemented to reflect the clinical reality that individual differences in patients' sensitivity to medications typically result in adjustments in dose. When a *d*-amphetamine dose was reached that resulted in a prolonged attenuation of the reinforcing strength of cocaine, other cocaine doses were tested to generate a dose-effect curve, which was compared with the curve generated in that monkey before *d*-amphetamine treatment. Additional features of the present studies that more closely recapitulate the clinical situation than most self-administration procedures include the use of cocaine-experienced (*vs* drug-naïve) subjects, the use of the PR schedule to measure the reinforcing strength of cocaine (*vs* fixed-ratio, fixed-interval or second-order schedules of reinforcement that assess presence/absence of reinforcing effects) and the continued assessment of cocaine self-administration after termination of *d*-amphetamine treatment to assess whether discontinuation resulted in specific effects on behavior such as rebound increases in the reinforcing effects of cocaine or disruption of food-reinforced responding.

MATERIALS AND METHODS

Subjects and Apparatus

Subjects were four adult male rhesus monkeys (*Macaca mulatta*), each prepared with a chronic indwelling venous catheter and subcutaneous vascular access port (Access Technologies, Skokie, IL) under sterile surgical conditions as described previously (Czoty *et al*., 2006). Two subjects (R-1429 and R-1425) had $\sim$6 months of experience self-administering cocaine at the outset of the present study and two subjects (R-1427 and R-1268) had self-administered cocaine for over 4.5 years. Monkeys were housed individually in sound-attenuating chambers ($0.91 \times 0.91 \times 0.91$ m; Plas Labs, Lansing, MI). The front wall of each cubicle was constructed of Plexiglas to allow the monkey visual access to the laboratory. Each cubicle was equipped with two response levers (BRS/LVE, Beltsville, MD). Four stimulus lights, alternating white and red, were located in a horizontal row above each lever. A receptacle located between the levers was connected via tygon tubing to a pellet dispenser located outside the chamber for response-contingent delivery of food pellets. Each animal was fitted with a stainless-steel restraint harness and spring arm (Restorations Unlimited, Chicago, IL) that attached to the rear of the cubicle. A peristaltic infusion pump (Cole-Parmer Instrument Co., Vernon Hills, IL) was located on the top of the chamber for delivering injections at a rate of $\sim$1.5 ml/10 s. Monkeys received fresh fruit, peanuts, and vegetables several days per week and water was available *ad libitum*. Animal housing and handling and all experimental procedures were performed in accordance with the 2003 National Research Council *Guidelines for the Care and Use of Mammals in Neuroscience and Behavioral Research* and were approved by the Animal Care and Use Committee of Wake Forest University. Environmental enrichment was provided as outlined in the Animal Care and Use Committee of Wake Forest University Non-Human Primate Environmental Enrichment Plan.

Food-Reinforced Responding

Monkeys were trained under a reinforcement schedule in which 50 responses on the left lever resulted in delivery of one food pellet, ie, a 50-response fixed-ratio (FR 50) schedule. Under this schedule, white stimulus lights above the left lever were illuminated and 50 responses resulted in food pellet delivery, extinguishing of white lights and illumination of red stimulus lights for 10 s, followed by a 10-sec timeout (TO) period during which no lights were illuminated and responding had no scheduled consequences. Sessions began at 0830 hours each day and lasted up to 23 h or until the maximum number of allowed food reinforcers was earned. Thus, only food was available from 0830 hours to 1500 hours when the self-administration session began (see below). At that point, food and cocaine were concurrently available if the monkey had not yet received the maximum number of pellets. The maximum number of pellets that could be earned was determined for each monkey as that required to provide enough food to maintain body weight, measured at least monthly, at $\sim$95% of free-feeding levels. When monkeys earned fewer than the maximum number of food pellets, supplementary food (Purina Monkey Chow) was given at $\sim$0800 hours in an amount calculated to raise the total grams of food to the desired level. Target food amounts for the monkeys in the present study were 130 or 140 g for three monkeys and 180–190 g for the fourth.

Cocaine Self-Administration

Monkeys self-administered (−)cocaine HCl (National Institute on Drug Abuse, Bethesda, MD, dissolved in sterile 0.9% saline) under a PR schedule of reinforcement in sessions that began at 1500 hours each day. Under this schedule, white stimulus lights were illuminated above the right lever and 50 responses on that lever resulted in the first injection of the maintenance dose of cocaine (0.03 or 0.1 mg/kg per injection in ~1.5 ml over 10 s), extinguishing of white lights and illumination of red stimulus lights for 10 s, followed by a 10-min TO. The response requirement for subsequent injections was determined by the equation used by Richardson and Roberts (1996): ratio = $[5 \times e^{(R \times 0.2)}] - 5$, where e is the mathematical constant and R is equal to the reinforcer number. For the present studies, the first response requirement (50 responses) corresponds to the 12th value given by this equation and was followed by 62, 77, 95, 117, 144, 177, 218, 267, 328, 402, 492, 602, 737, 901, 1102, etc. Sessions ended when 2 h elapsed without an injection.

Chronic *d*-Amphetamine Treatment

Initially, 0.03 or 0.1 mg/kg cocaine was made available in evening PR sessions until responding stabilized (3 consecutive days on which the number of injections was within 2 of the 3-day mean, with no upward or downward trend). Subsequently, other doses of cocaine were substituted for the maintenance dose for at least 4 days and until the number of injections stabilized. The maintenance dose of cocaine was situated on the ascending limb of the dose-response curve, permitting the detection of either increases or decreases in the reinforcing strength of cocaine. At ~0830 hours on the next day, the external part of the catheter was connected to a syringe in an infusion pump (Cole-Parmer Instrument, Vernon Hills, IL) outside the chamber and *d*-amphetamine was infused at a rate of 0.4 ml/h such that monkeys received a dose of 0.01 mg/kg/h. Food-reinforced responding was studied throughout treatment. On day 8, the solution being infused was changed from *d*-amphetamine to saline at approximately noon. This time was selected because it takes ~3 h to infuse the volume of *d*-amphetamine in the catheter (~1.2 ml). Thus, *d*-amphetamine continued to be infused until ~1455 hours, at which time the catheter was filled with the maintenance dose of cocaine. This procedure minimized the amount of *d*-amphetamine infused as a bolus when the catheter was filled with cocaine immediately before the start of the self-administration session. At 1500 hours, the maintenance dose of cocaine was again made available for self-administration under the PR schedule of reinforcement, signaled by the illumination of the white stimulus lights above the right lever. Thus, *d*-amphetamine treatment continued until the start of the cocaine self-administration session. At ~0830 hours on the next day, the catheter was flushed with heparin/saline solution, after which administration of the same dose of *d*-amphetamine was continued. For the first monkey tested (R-1427) a 6-day interval was used during treatment with 0.01 mg/kg/h *d*-amphetamine. During *d*-amphetamine treatment, monkeys were observed daily to assess unconditioned effects such as locomotor activation, agitation, stereotopies, or other unconditioned behavioral effects.

This procedure (availability of the maintenance dose of cocaine for one session) was repeated every 8 days until day 24. If, at that time, the number of cocaine injections was decreased from baseline by ~30% or more, treatment was continued another 8 days. Whether or not treatment had been extended to 32 days, if no decrease in cocaine self-administration had been observed, or if tolerance to initial decreases had developed, the dose of *d*-amphetamine was then increased to 0.03 mg/kg/h and the effects of this dose of *d*-amphetamine were similarly assessed for 32 days. If treatment with this dose had no effect or if tolerance developed, the dose was increased to 0.056 mg/kg/h. When a dose was reached at which the number of cocaine injections remained decreased at day 32, higher cocaine doses (up to 0.56 mg/kg per injection) were made available for a single day at 3-day intervals while *d*-amphetamine treatment was continued. At 2 days after the curve had been completed, *d*-amphetamine treatment was discontinued and self-administration of the maintenance dose of cocaine was again examined on post-treatment days 3, 7, and 15.

Data Analysis

The dependent variable of primary interest was the number of cocaine injections earned under the PR schedule of reinforcement. In addition, the number of food reinforcers received was recorded in hourly bins. Because individual differences in sensitivity to *d*-amphetamine resulted in different regimens of *d*-amphetamine treatment in the four monkeys, individual data are shown.

RESULTS

Baseline Food- and Cocaine-Reinforced Responding

Under baseline conditions, monkeys earned all available food pellets, typically within the first 3 h of availability (data not shown); in all cases, all food reinforcers had been received before cocaine availability. In cocaine self-administration sessions, the number of injections received increased significantly as a function of the available cocaine dose, up to 0.56 mg/kg cocaine, in all monkeys (Figure 1, closed symbols). In all cases, the cocaine self-administration session had concluded by 0730 hours on the morning following its 1500 hours start. Typically, when a new cocaine dose was substituted for the training dose, responding stabilized within 10 sessions.

Effects of *d*-Amphetamine on Food- and Cocaine-Reinforced Responding

At no time during the present studies were any overt unconditioned effects of *d*-amphetamine noted. These data are consistent with an earlier study (Czoty *et al*., 2010), in which only mild agitation was observed in three of four subjects during treatment with 0.1 mg/kg *d*-amphetamine. The only exception was subject R-1268, which showed transient motor effects (intermittent periods of laying down with eyes closed or leaning against cage wall) during days 3 and 4 of 0.056 mg/kg *d*-amphetamine treatment, which

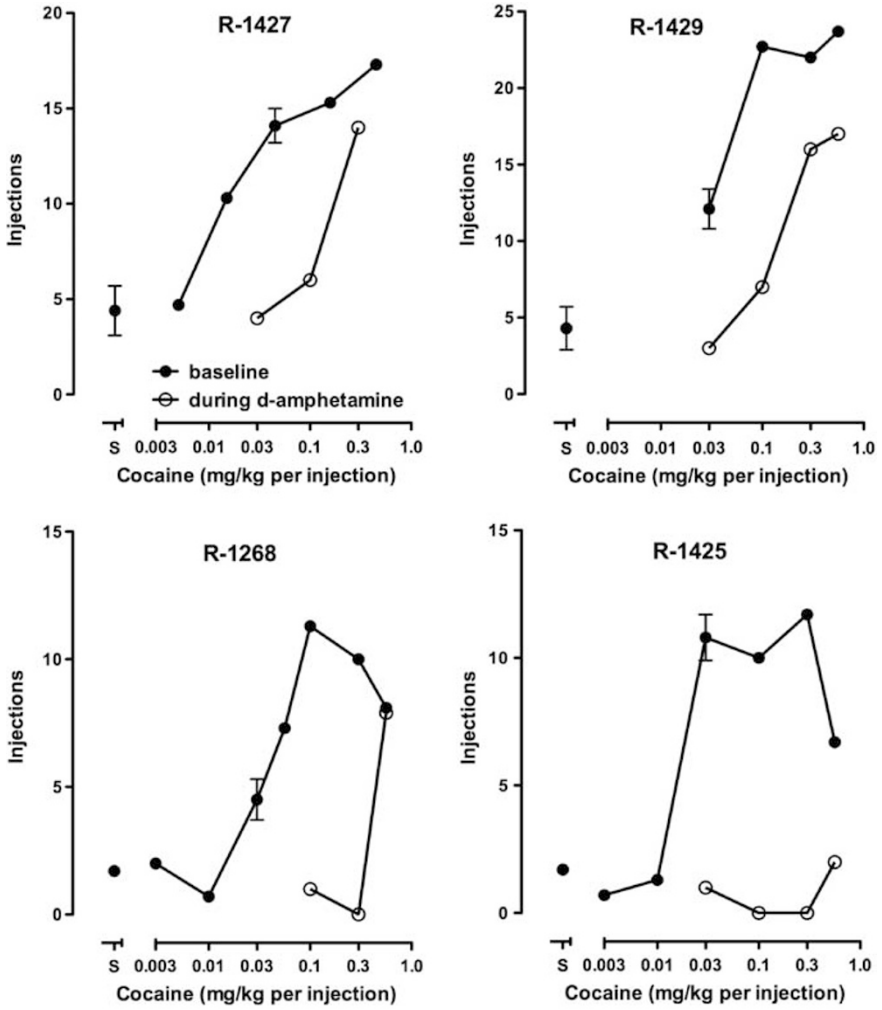

Figure 1 Dose-effect curves for cocaine self-administration before (closed symbols) and during (open symbols) *d*-amphetamine treatment in four monkeys. Each point is the mean of the last three days of availability of a cocaine dose or saline. When a cocaine dose or saline was made available on more than one occasion, error bars indicate the SEM. Ordinates: number of cocaine injections earned; abscissae, available cocaine dose.

coincided with disruptions in food-maintained responding (see below and Figure 2, third panel) and dissipated by day 5.

In subject R-1427, food-reinforced responding was not affected during 24 days of treatment with 0.01 mg/kg/h *d*-amphetamine (Figure 2, top panel). The number of 0.03 mg/kg cocaine injections earned was decreased from baseline on day 6, but by day 24 had returned to a level slightly above baseline. When the *d*-amphetamine dose was increased to 0.03 mg/kg/h, the number of food reinforcers earned under the FR 50 schedule decreased to near zero by day 5; tolerance gradually developed to this effect such that the number of food reinforcers earned returned to baseline by day 12. When food-maintained responding was disrupted, monkeys ate all supplemental chows on the following day as described previously (Czoty *et al*., 2010). When first examined on day 8 of treatment with 0.03 mg/kg/h *d*-amphetamine, the reinforcing strength of 0.03 mg/kg cocaine was decreased substantially. In contrast to the tolerance that developed to *d*-amphetamine-induced decreases in food-reinforced responding, the decrease in cocaine self-administration persisted for over 4 weeks of treatment.

Effects of *d*-amphetamine on the reinforcing strength of cocaine were qualitatively similar in R-1429 (Figure 2, second panel). Treatment with the lowest dose of *d*-amphetamine for 24 days decreased the number of 0.03 mg/kg cocaine injections earned by ~40%. Extending treatment for an additional 8 days resulted in neither tolerance to nor augmentation of this effect. Next, the *d*-amphetamine dose was increased, and following 2 weeks of treatment, the number of reinforcers earned on the PR schedule was reduced to two (data not shown). Shortly thereafter, however, the catheter lost patency and was replaced. Because we could not be certain that the decrease was due to effects of *d*-amphetamine rather than catheter failure, 0.03 mg/kg cocaine was again made available until responding again stabilized (BL2). At this point, treatment with 0.03 mg/kg/h *d*-amphetamine was begun again. The reinforcing strength of cocaine gradually decreased up to day 32. In this monkey, unlike the other subjects, food-reinforced responding was unaffected by *d*-amphetamine treatment.

As in R-1427 and R-1429, treatment with 0.01 mg/kg/h *d*-amphetamine for 24 days had no effects on food-maintained behavior and only slightly decreased the

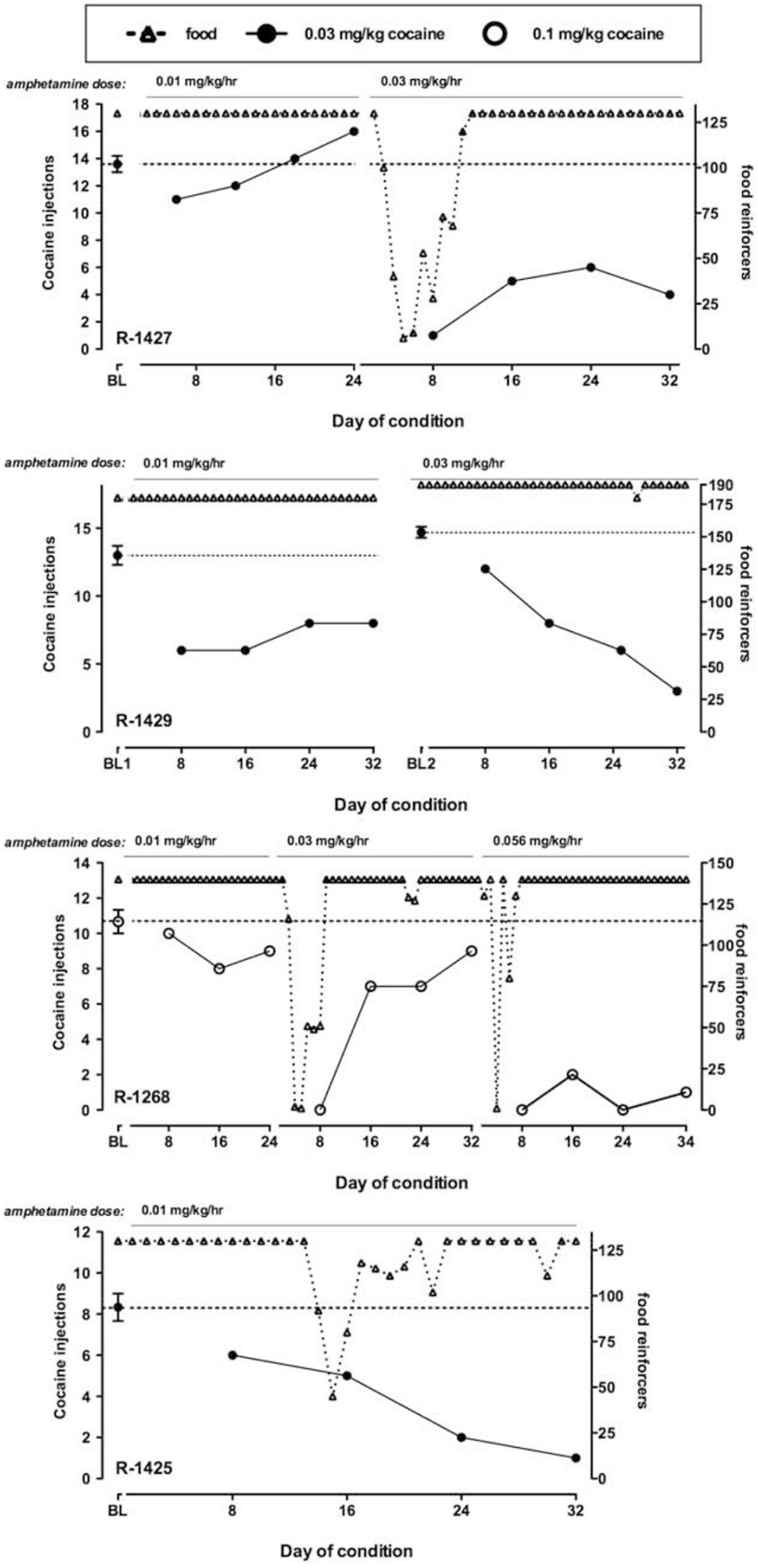

Figure 2 Cocaine self-administration and food-maintained responding during *d*-amphetamine treatment in four monkeys. Left ordinates: number of cocaine injections received; right ordinates: number of food reinforcers earned; abscissae: day of exposure to each condition, which is indicated across the top of each panel. BL represents average (±SEM) number of injections earned at baseline before *d*-amphetamine treatment. BL2 for R-1429 indicates a redetermined baseline after catheter replacement (see Results). The dashed line in each graph indicates the mean number of injections received under baseline conditions (ie, before *d*-amphetamine treatment).

reinforcing strength of cocaine (0.1 mg/kg) in R-1268 (Figure 2, third panel). As observed in R-1427, increasing the *d*-amphetamine dose to 0.03 mg/kg/h resulted in immediate decreases in food- and cocaine-reinforced responding, and tolerance developed to the effect on food-maintained responding. Unlike in R-1427, however, tolerance also developed to the decrease in cocaine self-administration such that the number of 0.1 mg/kg cocaine injections earned approached baseline levels by day 32. When the *d*-amphetamine dose was further increased to 0.056 mg/kg/h, a transient decrease in food-maintained responding was again observed, but the initial decrease in cocaine self-administration was maintained for at least 32 days.

In subject R-1425, treatment with *d*-amphetamine also resulted in prolonged decreases in self-administration of 0.03 mg/kg cocaine and a decrease in food-reinforced responding to which tolerance developed (Figure 2, bottom panel). In this monkey, however, effects were observed during treatment with the lowest dose, 0.01 mg/kg/h *d*-amphetamine.

Redetermination of Dose-Effect Curves

Once a dose of *d*-amphetamine was found that decreased cocaine self-administration for at least 32 days, additional cocaine doses were made available for a single self-administration session until a complete dose-effect curve was determined. In all monkeys, the reinforcing strength of several cocaine doses was found to be decreased compared with baseline values, such that the dose-effect curves were shifted downward and/or to the right (Figure 1, open symbols).

Recovery of Cocaine Self-Administration After *d*-Amphetamine Treatment

Food- and cocaine-maintained responding were examined in the 2 weeks after termination of *d*-amphetamine treatment to characterize the time course of recovery of cocaine self-administration and to assess whether discontinuation of *d*-amphetamine treatment resulted in any rebound increases in the reinforcing strength of cocaine or disruption of food-maintained responding. Although individual differences were observed across monkeys (Figure 3), the reinforcing strength of the maintenance dose of cocaine generally remained decreased below baseline for the first 3–7 days following discontinuation of *d*-amphetamine treatment and was near baseline by day 15 after treatment termination in three of four monkeys; no disruption of food-reinforced responding was observed in these subjects. In the fourth monkey (R-1425), the reinforcing strength of cocaine remained substantially decreased on day 15 after discontinuing *d*-amphetamine treatment, and an initial disruption of food-maintained responding was observed, which dissipated gradually. It is noteworthy that the subject which was most sensitive to *d*-amphetamine during treatment (R-1425) also showed prolonged disruptions in food- and cocaine-maintained responding after termination of treatment.

DISCUSSION

Accumulating evidence from preclinical studies and clinical trials indicates that *d*-amphetamine possesses therapeutic potential in the treatment of cocaine dependence (see Introduction). The results of the present study extend these findings to a novel animal model designed to incorporate features likely to be encountered clinically. In all monkeys, a dose of *d*-amphetamine was reached that decreased the reinforcing strength of cocaine for over 1 month. This effect was selective for cocaine-maintained responding in that decreases in food-maintained responding either dissipated over several days or were absent. When a range of cocaine doses was examined during *d*-amphetamine treatment, dose-effect curves for cocaine self-administration were shifted downward and/or to the right in all monkeys. Finally, discontinuation of *d*-amphetamine treatment was not associated with disruptions in food-maintained responding in three of four monkeys, and although cocaine self-administration gradually returned to baseline levels, no rebound increase in the reinforcing strength of cocaine was observed.

A primary goal of the present studies was to develop an animal model of cocaine addiction that incorporated features of human cocaine use and treatment approaches that have not been used in current models. The impetus for this effort was the observation that, although intravenous self-administration techniques have demonstrated impressive predictive validity with respect to the abuse liability of drugs (eg, Griffiths *et al*., 1980; Ator and Griffiths, 2003),

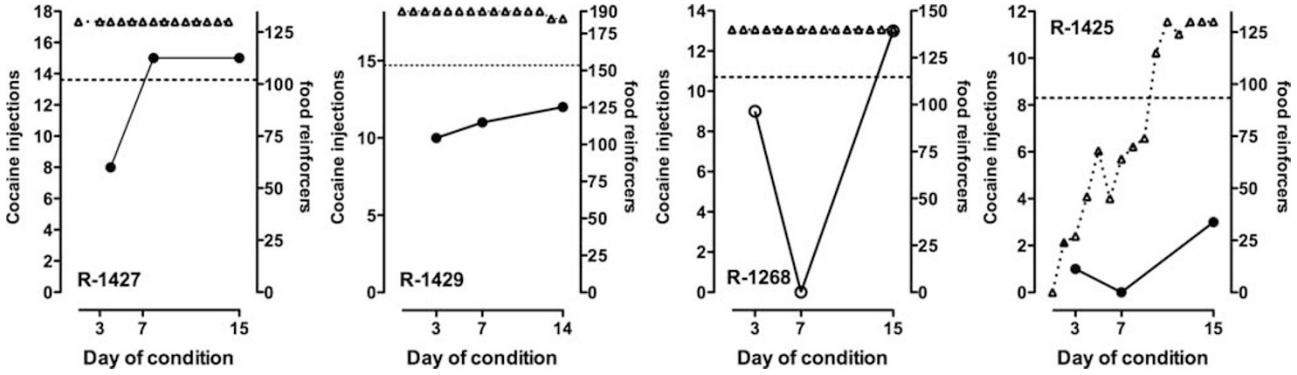

Figure 3 Cocaine- and food-maintained responding after discontinuation of *d*-amphetamine treatment in individual monkeys. Ordinates, symbols and lines as in Figure 2; abscissae: days after discontinuation of *d*-amphetamine treatment.

techniques designed for this purpose have been less successful in identifying drugs that will serve as effective medications for cocaine dependence. Moreover, features known or recommended to increase predictive validity, such as chronic treatment with putative pharmacotherapies, assessment of selectivity of medication effects through concurrent assessment of non-drug maintained behaviors, single-subject designs and examination of treatment drug effects on a range of self-administered cocaine doses (c.f. Mello and Negus, 1996; Ator and Griffiths, 2003; Haney and Spealman, 2008), are typically not included in preclinical assessments. For example, the present studies used cocaine-experienced monkeys, which would be expected to better model a long-term cocaine user than drug-naïve animals. In addition, unlike the majority of preclinical studies in which animals self-administer cocaine daily under limited-access conditions (eg, a predetermined maximum cocaine intake or amount of time), the present study examined cocaine self-administration using a PR schedule, in which the end of the session was determined by the monkey. Moreover, in the present study cocaine self-administration was suspended during the first week of treatment. This feature was implemented to model the frequent (though not universal) clinical situation, in which a cocaine addict is able to abstain from using cocaine for a brief period of time during the initial phase of treatment due to hospitalization, incarceration, or motivation to remain abstinent when starting treatment. It is unlikely that the decreases in cocaine self-administration were simply due to discontinuation of cocaine alone or sporadic behavioral testing. In previous studies using identical procedures, discontinuation of self-administration sessions for up to 1 week did not alter the cocaine dose-response curve (Czoty *et al*., 2006). An additional advantage, from a pharmacological point of view, is that suspending cocaine treatment provides a more pure assessment of the effects of the pretreatment drug, as neuroadaptations that are ostensibly induced by chronic *d*-amphetamine treatment are not affected by cocaine during this initial period.

In addition to providing a more clinically relevant pattern of access to cocaine, the present studies employed an approach to drug treatment designed to better reflect clinical use. The response of each subject to *d*-amphetamine treatment was monitored individually and adjustments to the treatment regimen (ie, increasing the *d*-amphetamine dose) were made based on the individual's response (ie, a single-subject design, c.f. Ator and Griffiths, 2003). This approach contrasts with the vast majority of animal self-administration studies, which use a group design in which all subjects receive a predetermined range of doses and data are averaged across animals for analysis. In the present study, although individual differences in sensitivity to *d*-amphetamine necessitated that different doses were effective, *d*-amphetamine produced similar effects in all monkeys. Had data for each *d*-amphetamine dose been averaged across subjects, this conclusion would have been obscured. More importantly, an individualized approach to treatment, with adjustments in medication dose guided by efficacy and side effects, is more similar to a physician's approach to treating a patient.

Perhaps most critically, the present studies examined chronic administration of *d*-amphetamine, unlike many preclinical studies that assess the effects of acute drug pretreatments on cocaine self-administration. In addition to the face validity provided by using chronic treatment, it has become apparent that *d*-amphetamine has opposite effects on cocaine administration when administered acutely *vs* chronically. Whereas chronic regiments of *d*-amphetamine have been consistently shown to decrease cocaine self-administration across a range of conditions, acute administration of *d*-amphetamine can enhance the reinforcing effects of cocaine in self-administration and reinstatement paradigms (eg, de Wit and Stewart 1983; Barrett *et al*., 2004). Contrasting effects of acute *vs* chronic drug treatment on cocaine self-administration have also been observed with other dopaminergic drugs, including the dopamine D1 receptor antagonist ecopipam (eg, Romach *et al*., 1999; Haney *et al*., 2001) and the low-efficacy D2 receptor agonist aripiprazole (Sorensen *et al*., 2008; Bergman, 2008; Thomsen *et al*., 2008).

Chronic treatment also proved critical to assessing the selectivity of *d*-amphetamine's effects on drug- *vs* food-maintained behavior, which was assessed by comparing self-administration to monkeys' daily responding maintained by presentation of food pellets on a separate response lever. In three of four monkeys, treatment with the dose of *d*-amphetamine that proved effective in decreasing the reinforcing strength of cocaine produced decreases in food-maintained responding. However, in all cases tolerance developed to this effect while cocaine self-administration remained decreased, similar to the findings of Negus and Mello (2003b). As observed in a previous study that examined 5 days of *d*-amphetamine treatment (Czoty *et al*., 2010) effects on food-maintained responding were likely due to disruptive effects of *d*-amphetamine on responding rather than to a reduction in appetite or the appetitive value of food pellets, as monkeys routinely took and ate chows and preferred foods when offered by a technician. Taken together, these data suggest that *d*-amphetamine treatment may be associated with some side effects during initial treatment, but that tolerance is likely to develop to these effects, whereas the ability of *d*-amphetamine to reduce the reinforcing strength of cocaine would be maintained. One caveat to this conclusion is that food and cocaine were available under different schedules of reinforcement, which may have contributed to the apparent selectivity of *d*-amphetamine. However, Negus (2003) studied cocaine-food choice in which reinforcers were available under different FR schedules and found that *d*-amphetamine selectively reduced cocaine choice. Finally, in the present study, *d*-amphetamine was administered continuously to provide a near-constant level of drug in the brain. Unlike the study by Negus (2003), *d*-amphetamine treatment was terminated before cocaine availability. Considering the elimination half-life is $\sim$4.5 h for intravenous *d*-amphetamine (Beckett and Rowland, 1965), appreciable levels of drug were most likely present during most of the session. It would be possible and worthwhile in future studies to examine whether comparable effects would be observed with different treatment regimens, including those that more accurately reflect the pharmacokinetics of drugs taken once daily by humans.

Once a *d*-amphetamine dose was identified that decreased the reinforcing strength of the maintenance dose of cocaine,

self-administration of higher cocaine doses was assessed while *d*-amphetamine treatment was continued. This phase of the experiment served two purposes. First, studying a single cocaine dose generates ambiguous data with respect to the interaction between *d*-amphetamine and cocaine; a decrease in self-administration could be interpreted as resulting from sedation, toxicity, or behavioral disruption as well as a decrease in the reinforcing effects of cocaine. Thus, a range of cocaine doses was studied in combination with *d*-amphetamine to gain a more complete understanding of the drug interaction. Second, it was deemed important to determine whether or not the effects of *d*-amphetamine were insurmountable. An important clinical consideration regarding use of a medication that decreases cocaine use is whether that effect could be overcome by taking higher cocaine doses. In addition to circumventing the therapeutic effects of the treatment drug, increasing the ingested cocaine dose in combination with an agonist therapy could result in lethal additive effects including cardiovascular crises or seizures. In the present studies, however, the ability of *d*-amphetamine to attenuate the reinforcing strength of cocaine could not be fully surmounted by increasing the cocaine dose. That is, in the presence of *d*-amphetamine, no dose of cocaine resulted in a number of injections as high as the dose that had peak reinforcing strength under baseline conditions. In all monkeys, chronic *d*-amphetamine treatment resulted in a downward and/or rightward displacement of the cocaine dose-effect curve compared with a dose-effect curve generated before *d*-amphetamine treatment. A caveat to this description of effects is that higher cocaine doses, in combination with *d*-amphetamine, may have resulted in a number of cocaine injections comparable to that seen at baseline. However, in previous studies in rhesus monkeys using this PR schedule (eg, Lile *et al*., 2003; Martelle *et al*., 2008; Czoty *et al*., 2010), dose-effect curves have been observed to plateau or have a descending limb when doses $\geqslant$0.56 mg/kg were tested (as seen in Figure 1 for R-1268 and R-1425). Another reason doses $>$0.56 mg/kg were not examined was the concerns regarding toxicity when made available in combination with *d*-amphetamine. Finally, when *d*-amphetamine treatment was discontinued, cocaine self-administration gradually returned to baseline levels as observed previously (Negus and Mello, 2003b; Czoty *et al*., 2010). Importantly, no rebound increases in cocaine self-administration were observed after *d*-amphetamine treatment, which could be hypothesized under a PR schedule of reinforcement if termination of *d*-amphetamine treatment resulted in a withdrawal state in which monkeys were hypersensitive to cocaine. Moreover, no disruptions in food-maintained responding were observed in three of four monkeys, providing preliminary evidence that termination of treatment with amphetamine-like drugs may not lead to a discontinuation syndrome, sometimes observed with antidepressant drugs that act on brain monoamine systems (eg, Haddad, 1998).

Taken together, the present results extend the conditions under which *d*-amphetamine selectively decreases cocaine self-administration. Importantly, several modifications to typical self-administration studies were employed in this model that render the experimental conditions more like the clinical situation, although there are many other aspects that have not been, and possibly cannot be, incorporated into animal models (eg, uniquely human negative consequences of drug use on employment and progressive psychiatric consequences). With regard to animal models, increasing face validity does not guarantee enhanced predictive validity. However, refining the model so that key features are more similar to those experienced by human cocaine users may strengthen predictive validity by increasing the overlap with neurobiological substrates and mechanisms involved in human drug use and dependence. Overall, we hypothesize that, by more closely resembling key clinical factors of cocaine use and treatment, the model described here will be better able to identify drugs that will serve as effective medications for cocaine dependence compared with typical animal models. Testing this hypothesis will require assessment of a variety of drugs that have and have not demonstrated success in the clinic and/or in typically used laboratory animal self-administration procedures. The current results with *d*-amphetamine are an encouraging sign that data obtained from this model will be concordant with clinical studies.

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DISCLOSURE

The authors declare no conflict of interest.

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