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ORIGINAL INVESTIGATION

M.T. Bardo · J.M. Valone · R.A. Bevins

Locomotion and conditioned place preference produced by acute intravenous amphetamine: role of dopamine receptors and individual differences in amphetamine self-administration

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Abstract Although previous studies have shown that dopamine (DA) antagonists block amphetamine reward, these studies have utilized animal models that involve repeated exposures to amphetamine. The present investigation examined the effect of DA antagonists on single-trial conditioned place preference (CPP) produced by acute intravenous (IV) amphetamine in rats. In the first experiment, rats were prepared with a jugular catheter and then received an acute IV injection of amphetamine (0.1–3 mg/kg) paired with one compartment of a CPP apparatus. Relative to sham controls (no IV catheter), amphetamine produced a dose-dependent increase in locomotor activity and CPP. Two further experiments demonstrated that both effects of amphetamine were completely blocked by pretreating rats with the D_1 DA antagonist SCH-23390 (0.025 and 0.25 mg/kg) or the D_2 DA antagonist eticlopride (0.2 and 2 mg/kg) on the conditioning trial. In a final experiment, single-trial amphetamine CPP did not predict subsequent self-administration of IV amphetamine (10–50 µg/infusion) using either a fixed ratio (FR) 1 or progressive ratio (PR) schedule of reinforcement. Thus, while sharing a similar DA receptor mechanism, the present results indicate that single-trial CPP and self-administration are dissociable effects of IV amphetamine.

Key words Amphetamine · Dopamine · Locomotor activity · Conditioned place preference · Self administration · Dopamine receptor

M.T. Bardo (✉) · J.M. Valone · R.A. Bevins[1]
Department of Psychology, University of Kentucky, Lexington, KY 40506, USA
e-mail: mbardo@pop.uky.edu, Fax: +1-606-323-1979

Present address:
[1] Department of Psychology, University of Nebraska, Lincoln, NE 68588-0308, USA

Introduction

Clinical evidence suggests that vulnerability to drug abuse may be predicted by the degree of positive reward derived from the initial drug experience (Haertzen et al. 1983). Individual differences in the acute rewarding effect of various drugs of abuse probably reflect, at least in part, neuropharmacological differences related to both genetic and environmental factors. Unfortunately, investigations into the neuropharmacological mechanisms that underlie acute drug reward are lacking. In humans, most studies of drug reward involve subjects that have an extensive history of drug-taking. Similarly, animal models of drug reward, such as conditioned place preference (CPP) or self-administration (Yokel 1987; Carr et al. 1989), typically require repeated drug exposures. Since repeated drug exposure may produce either tolerance or sensitization to the behavioral effect of various drugs (Stewart and Badiani 1993; Ramsay and Woods 1997; Schenk and Partridge 1997), these approaches do not allow for assessment of the neuropharmacological mechanisms of acute drug reward.

One potential strategy to assess acute drug reward in laboratory animals is to utilize the single-trial CPP procedure. Although CPP typically requires multiple drug conditioning trials, evidence indicates that acute IV injection of a relatively high dose of morphine (4–8 mg/kg) induces CPP in rats (Mucha et al. 1982; Bardo and Neisewander 1986). This preference is established by rapidly infusing the drug at the beginning of a 30-min placement into a distinct stimulus compartment. On the next day, rats are given equivalent exposure (without drug) to a different stimulus compartment. When rats are subsequently allowed to choose between the two compartments, they show a preference for the drug compartment relative to the no-drug compartment. Establishment of this CPP is blocked by naloxone (Bardo and Neisewander 1986), indicating that opiate receptors mediate acute morphine reward. At present, it is unclear if single-trial CPP is unique to opiates or whether it may also be obtained with other drug classes.

The major purpose of the present study was to determine if IV amphetamine produces single-trial CPP and to assess if this effect is blocked by dopamine (DA) antagonists selective for either the D_1 receptor family (D_1 and D_5) or the D_2 receptor family (D_2, D_3 or D_4). Previous work has shown that amphetamine reward is attenuated by either D_1 or D_2 DA antagonists (Yokel and Wise 1975; Spyraki et al. 1982; Hoffman and Beninger 1989). In all of these studies, however, amphetamine reward was assessed with repeated drug treatment in either a CPP or self-administration model. It is well known that repeated amphetamine treatment induces behavioral sensitization and alters activity of the mesolimbic DA system (Robinson and Becker 1986; Kalivas et al. 1993; Cador et al. 1995; Segal and Kuczenski 1997). Thus, it not clear if the antagonist-induced attenuation in amphetamine reward would also be obtained in a non-sensitized animal model. A secondary purpose of the present study was to determine if the acute rewarding effect of IV amphetamine, assessed by single-trial CPP, predicts subsequent amphetamine abuse liability, assessed by repeated self-administration.

Materials and methods

Animals

Adult male Sprague-Dawley rats (200–225 g body weight) were obtained from Harlan Industries (Indianapolis, Ind., USA) and were caged individually with free access to food and water in the home cage. The colony room was controlled for temperature (24°C) and relative humidity (45%), with lights on from 0700 to 1900 hours. Prior to the start of each experiment, animals were acclimated to the colony room for at least 1 week and were handled for 2 days. Behavioral testing was conducted during the light phase of the cycle. All procedures were approved by the University of Kentucky Institutional Animal Care and Use Committee.

Surgery

Animals were anesthetized (100 mg/kg ketamine, 5 mg/kg diazepam, IP) and implanted with a catheter into the jugular vein. In the CPP experiments that required only a single injection of amphetamine, a polyethylene tube (PE-50) was inserted into the vein and exited out the mid-scapular region of the back. A sterile piece of stainless steel tubing was used to close the ending. Sham controls received the same surgical treatment, but they did not receive the catheter insertion. In the self-administration experiments that required repeated injections of amphetamine, a Silastic tube was inserted into the vein and exited out the top of a head mount that was affixed to the top of the skull with dental acrylic and metal screws. Daily infusions of heparinized saline and streptokinase (Pharmacia, Columbus, Ohio, USA; 250 000 IU, 2 mg/ml heparinized saline, 0.1 ml/rat per day) were used to maintain patency of the Silastic catheter. At the end of each experiment, each animal was injected with IV morphine (15 mg/kg) and presence of a rapid cataleptic response was used to confirm catheter patency.

Apparatus

For assessment of locomotor activity and CPP, two similar conditioning apparatus were used. Each apparatus had three different wooden compartments separated by removable partitions. The two end compartments measured 24×30×45 cm high, while the middle compartment was smaller and measured 24×10×45 cm high. One end compartment had white walls, a wire mesh floor, and pine bedding beneath the floor. The other end compartment had black walls, a metal rod floor, and cedar bedding beneath the floor. The middle compartment had gray walls and a solid wood floor. The solid partitions could be replaced with similar partitions containing a 10×10 cm opening, which allowed the animals access to all compartments. The apparatus was located in a laboratory room that was separate from the colony room and was equipped with a white noise generator and audio speaker (ambient background of 70 dB). Suspended from the ceiling above the apparatus was a video camera which was used to record the experimental sessions.

For assessment of amphetamine self-administration, 12 operant chambers (ENV-001; Med Associates, St Albans, Vt., USA) enclosed in a sound attenuating environment were used. Located in the bottom center of the front panel in each chamber was a 5×4.2 cm opening to a recessed food tray. Two metal response levers were located on the front panel, one on each side of the food tray. The center of each lever was mounted 7.3 cm from the grid floor. A 28-V cue light, 3 cm in diameter, was centered 6 cm above each lever. Drug infusions were delivered using a syringe pump (Med Associates; PHM-100) and a water-tight swivel that allowed a catheter to be attached from the syringe (10 ml) to the head mount of the animal in the operant chamber. A personal computer, using Med Associates interface, controlled the experimental sessions and collected data.

Behavioral procedures

For CPP, the conditioning procedure was conducted over 2 consecutive days. Because preliminary data from our laboratory indicated that drug-naive animals tend to show a slight preference for the black compartment, amphetamine conditioning was established in the white compartment. On day 1, animals were placed individually into either the white or black compartments (counterbalanced within treatment groups) for 30 min with the solid partitions inserted between the compartments. On day 2, animals received equal exposure to the opposite compartment. Conditioned animals were injected IV with amphetamine immediately following placement into the white compartment and were injected with saline immediately following placement into the black compartment. Control animals received either no injection in either compartment (sham control) or received saline in both compartments (saline control). To assess the effect of antagonist drugs (SCH-23390 or eticlopride), the 2-day conditioning procedure was similar, except that the antagonist drug was administered IV 5 min before placement into the white compartment. Locomotor activity in the white compartment was videotaped and later scored by an observer who was unaware of each animal's individual treatment. Locomotor activity was quantified by counting the number of times that each animal crossed over a line drawn on the video monitor screen that bisected the white compartment in the plane parallel to the end wall. A line cross was operationally defined as both front shoulders crossing the line.

On the day following the 2-day conditioning procedure, each animal was tested for CPP. The rat was placed into the center gray compartment and allowed to enter all compartments of the apparatus for 15 min. Test sessions were videotaped and the duration spent in each compartment was determined by an observer who was unaware of each animal's individual treatment. Entry into a compartment was operationally defined as having both front shoulders in the compartment.

To assess the potential correlation between single-trial amphetamine CPP and amphetamine self-administration, a group of rats were run in both behavioral procedures. Rats were first implanted with a Silastic jugular catheter exiting through a head mount. After 2 days of recovery, each animal was assessed for CPP with a single dose of amphetamine (1 mg/kg, IV) as described previously, except that rats were first given a 15-min preference test prior to conditioning to establish the baseline preference for each compartment. A baseline preference test was used in this experiment

in order to obtain a measure of amphetamine CPP from all rats in the sample, i.e., no rats were assigned to a saline control group. Following baseline testing, amphetamine was subsequently paired with the non-preferred compartment and saline was paired with the preferred compartment. Following this conditioning procedure, rats were again tested for place preference. The magnitude of amphetamine CPP for each individual rat was expressed as a percent change in duration spent in the non-preferred compartment from the preconditioning test to the post-conditioning test.

On the day following the single-trial CPP test, daily amphetamine self-administration sessions (3 h/session) were initiated. Rats were first trained to self-administer amphetamine (30 µg/infusion, 0.1 ml/infusion, 10 s infusion) on a fixed ratio (FR) 1 schedule using a two-lever choice procedure. Depression of one lever delivered amphetamine and depression of the other lever led to no reinforcement; the levers were counterbalanced for drug reinforcement across rats. Training continued until stable responding on the FR1 was established. Stable responding was defined as 15% or less variability in the number of responses on the drug lever across three consecutive sessions. After this criterion was reached, each rat was tested for self-administration of the training dose of amphetamine (30 µg/infusion) on a progressive ratio (PR) schedule of drug reinforcement across three consecutive sessions (5 h/session). Within each PR session, the number of responses required to obtain an amphetamine infusion increased incrementally (1, 2, 4, 6, 9, 12, 15, 20, etc.; see Roberts and Richardson 1992). The last ratio value completed for amphetamine infusion within each session was defined as the breakpoint. After the breakpoint value was determined using 30 µg amphetamine, rats were tested with two other amphetamine doses (10 and 50 µg/infusion) across consecutive daily sessions. For each dose, rats were first stabilized on an FR1 schedule and then were tested for PR responding as described previously.

Drugs

Amphetamine sulfate and morphine sulfate were obtained from the National Institute on Drug Abuse (Rockville, Md., USA); *R*(+)-SCH-23390 hydrochloride and *S*(–)-eticlopride hydrochloride were purchased from Research Biochemicals International (Natick, Mass., USA); ketamine hydrochloride (100 mg/ml injectable) was purchased from Fort Dodge Laboratories (Fort Dodge, Iowa, USA); and diazepam (5 mg/ml injectable) was purchased from Steris Laboratories (Phoenix, Ariz., USA). For CPP, drugs were prepared in sterile heparinized saline (0.9% NaCl) and injected IV in a volume of 1 ml/kg body weight. For self-administration, amphetamine was prepared in sterile saline and injected IV in a volume of 0.1 ml/infusion. All dosages were calculated using the salt form of the drug.

Statistics

In all of the single-trial CPP experiments, locomotor activity and preference data were analyzed by separate factorial analyses of variance (ANOVAs). Pairwise comparisons among treatment groups were performed using Tukey's HSD test (Kirk 1968). In these analyses, CPP data were expressed as either an absolute measure of preference (total duration in white compartment) or a relative measure of preference (total duration in white compartment divided by total duration in white+black compartments). However, since these two measures yielded essentially equivalent results across experiments, only the absolute preference data are presented in graphic form.

To determine if individual differences in amphetamine CPP predicted subsequent amphetamine self-administration, a CPP score was derived for each animal by subtracting the duration spent in the non-preferred compartment prior to conditioning from the duration spent in the non-preferred compartment after conditioning. Pearson product-moment correlation coefficients were then derived by correlating the shift in preference with the number of infusions on the FR1 and PR schedules. Separate correlation coefficients were determined for each self-administration test dose of amphetamine (10, 30 and 50 µg/infusion).

Results

Amphetamine dose-effect curves

Separate groups of rats (n=9–16/group) were conditioned with a single dose of amphetamine (0.1, 0.3, 1 or 3 mg/kg) or received no injections (sham control). Within this dose range, there was a dose-dependent increase in locomotor activity [$F(4,46)=15.80$, $P<0.0001$], with an apparent maximal increase at 1 mg/kg amphetamine (see Fig. 1A). Pairwise comparisons between groups revealed that, relative to the sham control, there was a significant increase in activity following 0.3, 1 and 3 mg/kg amphetamine, but not following 0.1 mg/kg amphetamine.

As shown in Fig. 1B, there was also a significant dose-dependent CPP with amphetamine [$F(4,48)=11.46$, $P<0.0001$]. Pairwise comparisons between groups revealed that, relative to the sham control, there was a sig-

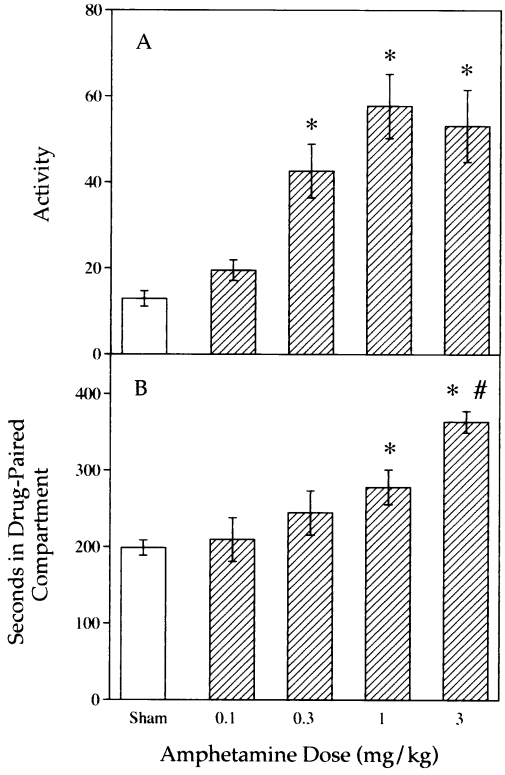

Fig. 1A, B Dose-effect curves for locomotor activity and CPP with acute IV amphetamine. **A** Mean level of activity (±SEM) measured in the white compartment immediately following amphetamine or sham injection. **B** Duration of time spent in the drug-paired white compartment on the test day following single-trial amphetamine conditioning. In both panels, an *asterisk* (*) represents a significant difference from the sham control group and a *hash* (#) represents a significant difference from 1 mg/kg amphetamine group [Tukey's test, $P<0.05$]

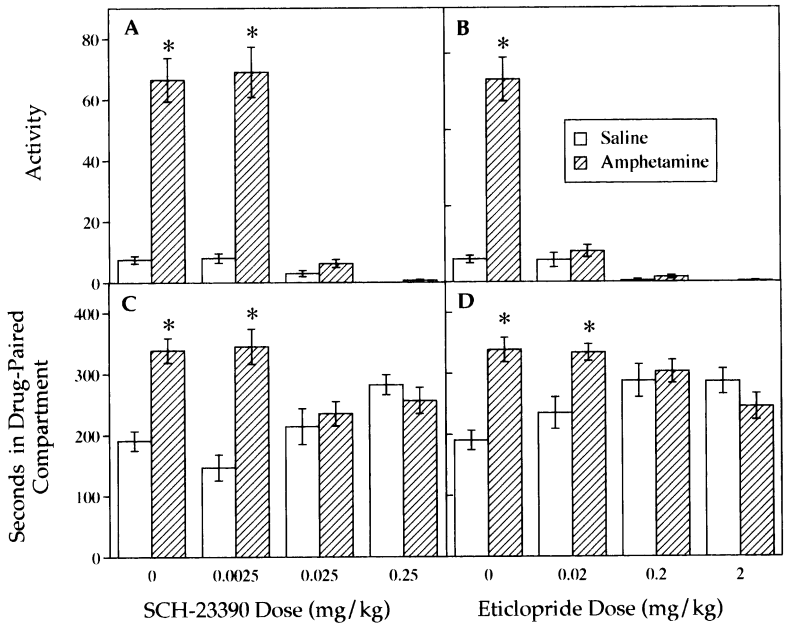

Fig. 2A–D Effect of varying doses of SCH-23390 or eticlopride on locomotor activity and CPP produced by acute IV amphetamine (1 mg/kg). **A** Mean level of activity (±SEM) on the conditioning day in rats pretreated with SCH-23390 or saline and then placed in the white compartment following either amphetamine (*hatched columns*) or saline (*clear columns*). **B** Mean level of activity (±SEM) on the conditioning day in rats pretreated with eticlopride or saline and then placed in the white compartment following either amphetamine or saline. **C** Duration of time spent in the drug-paired compartment on the test day in rats previously pretreated with SCH-23390 or saline on the conditioning day with amphetamine or saline. **D** Duration of time spent in the drug-paired compartment on the test day in rats previously pretreated with eticlopride or saline on the conditioning day with amphetamine or saline. In all panels, an *asterisk* (*) represents a significant difference from the saline conditioned group pretreated with the same dose of antagonist (Tukey's test, $P<0.05$)

nificant increase in preference following 1 and 3 mg/kg amphetamine, but not following either 0.1 or 0.3 mg/kg amphetamine. There was also a significant difference between groups conditioned with either 1 or 3 mg/kg amphetamine.

Effects of DA antagonists

To assess the role of DA receptor subtypes in the acute effects of IV amphetamine, separate groups of rats (n=8–10/group) were pretreated with either the D_1 antagonist SCH-23390 (0, 0.0025, 0.025 or 0.25 mg/kg) or the D_2 antagonist eticlopride (0, 0.02, 0.2 or 2 mg/kg) prior to conditioning with amphetamine (1 mg/kg) or saline. As shown in Fig. 2A, SCH-23390 decreased the locomotor stimulant effect of amphetamine. The overall ANOVA for locomotor activity revealed a significant interaction between conditioning drug (amphetamine or saline) and pretreatment dose of SCH-23390 [$F(7,76)$= 10.71, $P<0.0001$]. In saline-pretreated animals, amphetamine produced a significant increase in activity. This amphetamine-induced increase was not significantly altered by the lowest dose of SCH-23390 (0.0025 mg/kg). Pretreatment with 0.025 mg/kg SCH-23390 blocked the locomotor stimulant effect of amphetamine; this dose of SCH-23390 did not significantly decrease activity in saline controls. At the highest pretreatment dose of SCH-23390 (0.25 mg/kg), locomotor activity was almost completely depressed in both amphetamine and saline conditioned groups.

As shown in Fig. 2C, SCH-23390 also blocked amphetamine CPP. The overall ANOVA for CPP revealed a significant interaction between conditioning drug (amphetamine or saline) and pretreatment dose of SCH-23390 [$F(7,76)=44.45$, $P<0.0001$]. With saline pretreatment, amphetamine conditioned animals showed a preference for the drug-paired compartment relative to saline controls. This amphetamine CPP was not significantly altered by the lowest dose of SCH-23390 (0.0025 mg/kg). Pretreatment with either 0.025 or 0.25 mg/kg SCH-23390 blocked the amphetamine CPP. Pretreatment with SCH-23390 alone (open bars in Fig. 2C) also tended to increase preference; however, none of the SCH-23390 alone groups differed significantly from saline control.

Similar to SCH-23390, pretreatment with eticlopride decreased the locomotor stimulant effect of amphetamine (see Fig. 2B). The overall ANOVA revealed a significant interaction between conditioning drug (amphetamine or saline) and pretreatment dose of eticlopride [$F(7,77)=42.67$, $P<0.0001$]. In saline pretreated animals, amphetamine produced a marked increase in activity. This amphetamine-induced increase was blocked by the lowest dose of eticlopride (0.02 mg/kg). Higher pretreatment doses of eticlopride (0.2 and 2 mg/kg) also blocked the locomotor stimulant effect of amphetamine. Although the high eticlopride doses also decreased activity in saline controls, these differences did not reach statistical significance.

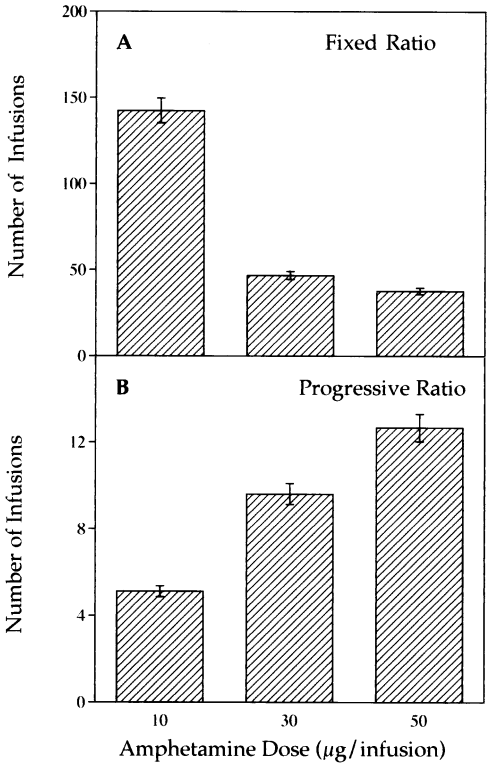

Fig. 3A, B Dose-effect curves for IV amphetamine self-administration using either an FR1 **A** or PR **B** schedule of reinforcement. The mean PR breakpoint values corresponding to the amphetamine doses of 10, 30 and 50 g/infusion were 9, 28.5 and 60, respectively. Significant dose-dependent differences in the number of infusions were obtained on both the FR1 and PR schedules (*F*-test, $P<0.05$)

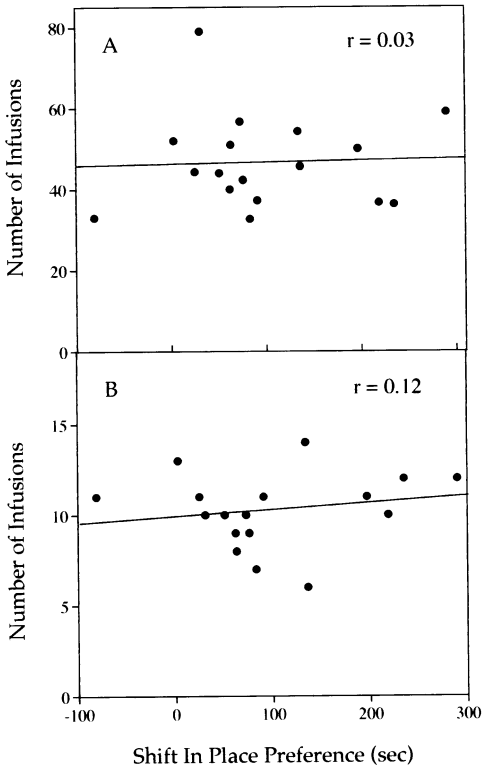

Fig. 4A, B Relationship between single-trial CPP using IV amphetamine (1 mg/kg) and self-administration of IV amphetamine (30 g/infusion). **A** Scatterplot of individual data points and a best-fit line derived from rats tested for amphetamine CPP and self-administration on a FR1 schedule. **B** Scatterplot of individual data points and a best-fit line derived from rats tested for amphetamine CPP and self-administration on a PR schedule. In both panels, the degree of relationship was not significant (Pearson *r*, $P>0.05$)

As shown in Fig. 2D, eticlopride blocked amphetamine CPP. The overall ANOVA revealed a significant interaction between conditioning drug (amphetamine or saline) and pretreatment dose of eticlopride [$F(7,77)=7.33$, $P<0.0001$]. With saline pretreatment, amphetamine-conditioned animals showed a preference for the drug-paired compartment relative to saline controls. Amphetamine CPP was also evident in animals pretreated with 0.02 mg/kg eticlopride, but not in animals pretreated with either 0.2 or 2 mg/kg eticlopride. Pretreatment with either 0.2 or 2 mg/kg eticlopride alone (open bars in Fig. 2D) produced a significant increase in preference compared to saline control.

Individual differences in amphetamine CPP and self-administration

Individual differences in single-trial amphetamine CPP and amphetamine self-administration were correlated in a group of rats ($n=17$) that were assessed in both behavioral procedures as described previously. Examination of the CPP group data revealed that the shift in preference for the non-preferred compartment after conditioning ranged from –81 to 290 s, with an average shift of 99.3 s (data not shown); this within-subject preference shift was statistically significant [$F(1,16)=19.22$, $P<0.001$]. Examination of the self-administration group data, displayed in Figs. 3A and B, revealed that the number of infusions varied as a function of amphetamine dose on the FR1 schedule [$F(2,10)=118.62$, $P<0.0001$] and the PR schedule [$F(2,10)=7.87$, $P<0.01$]. There was no significant relationship between amphetamine CPP and number of amphetamine infusions at any dose on either the FR1 or PR schedule. Scatterplots of the data from the training dose of amphetamine (30 μg/infusion) are presented in Figs. 4A and B.

Discussion

Previous work has demonstrated that single-trial CPP is obtained following acute IV morphine (Mucha et al. 1982; Bardo and Neisewander 1986). The present results indicate that single-trial CPP is not specific to opiate drugs, but that it is also evident following acute IV amphetamine. The amphetamine dose-effect curve for single-trial CPP was graded within the dose range tested (0.1–3 mg/kg). Within a similar dose range, other studies have shown that the dose-effect curve for multiple-trial

CPP following IP or SC injections of amphetamine is also graded (Bardo et al. 1995). These results counter the argument that CPP as a measure of drug reward is relatively insensitive to drug dosage (Wise 1989). Despite the graded effect, however, it should be noted that a plateau in the dose-effect curve defining the maximal conditioning effect was not apparent in the present study. Doses higher than 3 mg/kg amphetamine were not tested because of the potential for seizures, as well as evidence indicating that higher doses given repeatedly may produce a conditioned place aversion (Bardo et al. 1995).

In contrast to the present results, at least one report found that acute IV cocaine does not induce single-trial CPP (Nomikos and Spyraki 1988). While this finding suggests that single-trial CPP may not generalize to all stimulant drugs, procedural differences between the cocaine study conducted by Nomikos and Spyraki (1988) and the present amphetamine study may account for the differential outcomes. First, Nomikos and Spyraki (1988) tested only a single dose of cocaine (0.5 mg/kg), whereas the present report tested a full range of amphetamine doses (0.1–3 mg/kg). Second, Nomikos and Spyraki (1988) injected the rats outside of the conditioning apparatus, whereas the present report injected the rats inside of the apparatus. Perhaps having the onset of the IV drug effect in the test apparatus, rather than outside the apparatus, led to more robust conditioning in the present study. This possibility is supported by other work showing backward pairing of a conditioned stimulus (apparatus compartment) with an unconditioned stimulus (drug effect), as used in the Nomikos and Spyraki (1988) study, typically produces negligible conditioning (Mackintosh 1974). Given these procedural differences, it seems premature to conclude that IV cocaine CPP cannot be obtained with a single trial. Further parametric work will be needed to resolve this issue.

Although there has been some debate in the literature about the potential influence of conditioned locomotor responses on the expression of CPP (Swerdlow and Koob 1984; Carr et al. 1989), the present results with IV amphetamine show a clear dissociation between locomotor activity and CPP. Most important, the dose-effect curves for amphetamine-induced activity and CPP differed across the dose range tested. The lowest dose of amphetamine that increased activity was a half-log unit lower than that needed to produce CPP. In addition, while a clear plateau in locomotor stimulation was evident at 1 mg/kg amphetamine, no plateau in CPP was apparent up to 3 mg/kg amphetamine. These results suggest that, although drugs of abuse may increase locomotor activity and produce reward by activating a similar DA substrate in the brain (Wise and Bozarth 1987), expression of these different behaviors also involves some separate neuropharmacological mechanisms.

The present results also provide evidence about the role of D_1 and D_2 DA receptor families on the locomotor stimulant and rewarding effects of acute IV amphetamine. With locomotor activity, pretreatment with either SCH-23390 or eticlopride completely blocked the locomotor activity induced by IV amphetamine. These results are in accord with previous work showing that selective D_1 and D_2 DA antagonists are potent blockers of the hyperactivity observed with administration of acute amphetamine via other routes (Beninger and Hahn 1983; Mithani et al. 1986; Stewart and Vezina 1987; Vezina and Stewart 1989; Mazurski and Beninger 1991). Despite their similar blockade of hyperactivity following acute amphetamine, it is important to note that selective D_1 and D_2 DA antagonists also have a differential effect on the locomotor sensitization obtained with repeated amphetamine injections. That is, amphetamine-induced locomotor sensitization is blocked by D_1 antagonists, but not by D_2 antagonists (Stewart and Vezina 1987; Ujike et al. 1989; Vezina and Stewart 1989; Drew and Glick 1990). These findings indicate that locomotor activity following acute amphetamine and locomotor sensitization following repeated amphetamine injections involves, at least in part, separate neuropharmacological mechanisms.

Similar to their effects on amphetamine-induced locomotion, SCH-23390 or eticlopride pretreatments blocked completely the CPP induced by acute IV amphetamine. These findings are consistent with previous work showing that both D_1 and D_2 receptors play a role in amphetamine reward. In particular, pretreatment with selective D_1 or D_2 DA antagonists has been shown to attenuate amphetamine reward assessed by multiple-trial CPP (Spyraki et al. 1982; Mithani et al. 1986; Leone and Di Chiara 1987; Hoffman and Beninger 1989; Hiroi and White 1991; Acquas and Di Chiara 1994) and self-administration (Yokel and Wise 1975; Phillips et al. 1994). In these previous studies, however, it is important to note that the rewarding effect of amphetamine was assessed across repeated injections. Since the rewarding effect of amphetamine becomes sensitized across repeated administrations (Woolverton et al. 1984; Lett 1989; Strakowski et al. 1996), it is unclear if the antagonist effects observed in these previous studies reflect either a blockade of the acute rewarding effect of amphetamine or a blockade of the sensitization produced by repeated amphetamine injections. The present study directly addressed this issue by using a single-trial CPP procedure that rules out the contribution of any sensitization that occurs with repeated amphetamine injections. Since pretreatment with either SCH-23390 or eticlopride blocked single-trial amphetamine CPP, these results provide evidence that both D_1 and D_2 DA receptors mediate the primary rewarding effect induced by the first amphetamine experience.

While it seems likely that SCH-23390 and eticlopride disrupted single-trial amphetamine CPP by attenuating the acute rewarding effect of amphetamine, we cannot rule out the possibility that these antagonists may have impaired learning of the CPP behavior independent of any direct effect on amphetamine reward. In particular, previous work has shown that SCH-23390 may disrupt learning in various behavioral tasks across different species (Sanger 1987; Ichihara et al. 1989; Sawaguchi and Goldman-Rakic 1991). Perhaps most relevant to the

present study, Lin et al. (1994) examined the effect of SCH-23390 and raclopride on single-trial amphetamine conditioned taste aversion in rats. Like CPP, amphetamine conditioned taste aversion is thought to involve the acquisition of a Pavlovian association between a conditioned stimulus with amphetamine. In the study by Lin et al. (1994), pretreatment with SCH-23390 or raclopride did not alter the conditioned taste aversion produced by acute amphetamine. These results indicate that blockade of either D_1 or D_2 DA receptors does not produce a generalized impairment in the ability of animals to form a Pavlovian association between a conditioned stimulus and amphetamine following a single trial. Thus, it seems more likely that the DA antagonists used in the present report blocked the acute rewarding effect of amphetamine rather than impairing learning.

One unexpected finding from the present study was that eticlopride alone (no amphetamine) produced a significant preference for the drug-paired compartment. A similar trend was observed with SCH-23390. However, it is important to note that the DA antagonist doses that produced the apparent CPP also abolished activity almost completely. This antagonist-induced immobility may have prevented habituation to the stimulus compartment on the conditioning day, thus making the compartment relatively more novel on the test day. Since rats prefer a novel compartment relative to a familiar compartment (Bardo et al. 1989; Parker 1992), the apparent antagonist-induced CPP observed here may reflect a preference for novelty.

In addition to assessing the role of D_1 and D_2 DA receptors, the present study also examined the potential correlation between single-trial amphetamine CPP and amphetamine self-administration. With human subjects, the degree of self-reported positive reward derived from the initial drug experience is related to drug abuse vulnerability (Haertzen et al. 1983). To assess this predictive relationship in a controlled setting, individual differences in single-trial amphetamine CPP were correlated with subsequent rates of amphetamine self-administration on an FR1 and PR schedule. Although the number of amphetamine self-infusions varied as a function of dose, we found no evidence that individual differences in single-trial amphetamine CPP correlated with subsequent amphetamine self-administration rates under either the FR1 or PR schedule. Thus, these results in rats challenge the idea that individual differences in drug abuse vulnerability are related to the degree of reward derived from the first drug experience.

Finally, any conclusion based upon the present correlational results should be tempered because the CPP and self-administration paradigms are not equivalent measures of drug reward. While there seems to be reasonable correspondence between these paradigms in their ability to identify drugs that have abuse liability (cf. Yokel 1987; Carr et al. 1989), both methodological and theoretical differences have tended to prevent direct comparison of results obtained with each paradigm. Recent evidence from monkeys indicates that the relationship between CPP and self-administration is not completely concordant (Evans and Foltin 1997; Foltin and Evans 1997). Evidence also indicates that the neural mechanisms that underlie CPP and self-administration do not overlap completely (Bardo 1998). Thus, the failure to find any relationship between single-trial amphetamine CPP and subsequent amphetamine self-administration may be related to inherent differences between the paradigms, rather than to differences in the acute and chronic rewarding effects of amphetamine.

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