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ORIGINAL INVESTIGATION

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The influence of environment on the induction of sensitization to the psychomotor activating effects of intravenous cocaine in rats is dose-dependent

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Abstract The acute psychomotor response and development of sensitization to amphetamine is attenuated if IP injections are given in the cage where a rat lives relative to when injections are given in a novel but physically identical test environment. Furthermore, when the environmental cues predicting IP injections are completely eliminated by using remotely activated IV injections in the home cage, 1.0 mg/kg amphetamine produces a very small acute response and no sensitization. The same treatments do produce sensitization if IV injections are signaled by placement of the rat in a novel test cage. The present experiment was designed to determine if there is a similar effect of environmental condition on the response to IV cocaine, and to what extent the effect may be dose-dependent. This was accomplished by comparing the psychomotor activating effects (rotational behavior) of repeated IV administrations of one of eight doses of cocaine (0.0, 0.3, 0.6, 1.2, 2.4, 3.6, 4.8, or 7.2 mg/kg) given in the home cage, with infusions of the same doses given in a novel test cage. There was no effect of environment on the acute psychomotor response to cocaine. There was, however, a significant effect of environment on the induction of sensitization. A higher dose of cocaine was required to induce sensitization when IV administrations were given in the home cage than when they were given in a physically identical but novel test environment. At high doses, however, cocaine induced sensitization regardless of environmental condition. The results suggest that the effect of this environmental manipulation is to shift the dose-effect curve for the induction of sensitization, and support the notion that the ability of psychostimulant drugs to induce sensitization can be modulated by the circumstances surrounding drug administration.

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Introduction

The repeated administration of psychomotor stimulant drugs, such as amphetamine or cocaine, results in a progressive and persistent increase in their psychomotor activating effects, a phenomenon known as behavioral sensitization (Robinson and Becker 1986; Stewart and Badiani 1993). Although the exact mechanism responsible for this phenomenon is not known, behavioral sensitization is accompanied by long-lasting neuroadaptations, including changes in dopamine (DA) neurotransmission (Kalivas and Stewart 1991), and structural modifications in nucleus accumbens and prefrontal cortex neurons (Robinson and Kolb 1997).

There has been considerable research on both the behavioral pharmacology and the neurobiology of sensitization, but the conditions necessary for its induction and expression are still not well understood. Because behavioral sensitization is accompanied by drug-induced neuroadaptations, it is tempting to think of sensitization as an inevitable consequence of exposure to psychostimulant drugs. There is, however, increasing evidence that the ability of drugs to produce their behavioral effects can be powerfully modulated by the circumstances surrounding drug administration (Barrett 1987; Falk and Feingold 1987). For example, both the acute psychomotor response to amphetamine and the rate of sensitization to the psychomotor activating effects of IP amphetamine and cocaine are attenuated if rats are given drug treatments in their home cage, relative to rats treated in a physically identical but novel test environment (Badiani et al. 1995a, b, c). In addition, we reported recently that if the cues associated with IP treatments are eliminated by using unsignaled IV infusions, low doses of amphetamine (0.5–1.0 mg/kg) fail to induce sensitization. The same doses given in a physically identical but novel environment do induce sensitization (Crombag et al. 1996; Robinson et al. 1998).

The study by Crombag and colleagues (1996) clearly establishes that the circumstances surrounding drug administration can determine whether a given dose of amphetamine is capable of inducing sensitization. Indeed, this study suggests it might be impossible to induce sensitization if unsignaled IV infusions of a psychostimulant drug are given in the home environment. Alternatively, the effect of this environmental manipulation may not be to gate sensitization in an all-or-none fashion, but to shift the dose-effect curve for inducing sensitization. The purpose of this experiment, therefore, was two-fold: 1) to determine if the effects of IV infusions of cocaine given in a home or novel environment are similar to those previously reported for amphetamine, and 2) to determine if these effects are dose-dependent.

Materials and methods

Subjects

Male Sprague-Dawley rats (Harlan Sprague-Dawley Inc., Indianapolis, Ind., USA), weighing 200–225 g upon arrival were housed in a room with a 14-h light/10-h dark cycle (lights on from 0600 to 2000 hours), with ad libitum access to food and water.

Surgical and screening procedures

After 1 week of habituation to the main animal colony, all rats were pretreated with atropine methyl nitrate (dissolved in 0.5 mg/ml in saline and administered IP; Sigma, St Louis, Mo., USA), and then anesthetized with sodium pentobarbital (Nembutal, 50 mg/kg, IP) supplemented with methoxyfluorothane. A 21 gauge stainless steel guide cannula was positioned above the nigrostriatal bundle using the following coordinates, measured from bregma: anterior/posterior −3.0 mm; medial/lateral ±1.8; ventral −1.0 mm (Paxinos 1986). In half of the animals, the cannula was positioned in the left hemisphere and in the other half it was positioned in the right hemisphere. Along with the guide cannula, a 15 gauge piece of hypodermic tubing bent at a 45 degree angle and an L-shaped piece of plastic tubing were affixed to the skull using dental cement and jeweler's screws attached to the skull. The guide cannula was capped with a stainless steel stylet to maintain patency between procedures.

At least 2 days following surgery, all rats received a unilateral 6-hydroxydopamine (6-OHDA) lesion of the mesostriatal dopamine system. Briefly, awake animals were pretreated with desipramine hydrochloride IP (15 mg/kg in distilled water: Sigma) 30–60 min before receiving 6-OHDA (Breese and Traylor 1971). A 29 gauge stainless steel cannula was inserted into the guide cannula so that its tip was located 8.3 mm ventral from the surface of the skull. 6-OHDA HBr, 8 µg in 4 µl of a saline-ascorbate solution, was infused over an 8-min period. The infusion cannula was left in place for 2 min following the infusion and then was removed from the guide cannula, and the stylet reinserted.

The purpose of the 6-OHDA lesion was so cocaine-induced rotational behavior could be used as an index of the psychomotor activating effects of cocaine. As discussed in detail elsewhere (Badiani et al. 1995a), the quantification of rotational behavior in rats with a unilateral 6-OHDA lesion offers a number of advantages over more traditional measures of psychomotor activation. For example, in rats with a unilateral 6-OHDA lesion, a progressive increase in drug effect is seen as a progressive increase in rotational behavior (Robinson 1984). In intact rats, however, a progressive increase in drug effect is not necessarily characterized by a progressive increase in locomotor activity, which can make it difficult to quantify and interpret the development of sensitization. Also, the unconditioned rotational response produced by an injection of saline in a novel test environment is negligible in rats with a unilateral 6-OHDA lesion, whereas the locomotor response in intact rats is usually very large (Badiani et al. 1995a).

The animals were allowed to recover from the lesion for at least 1 week, and were then tested with 0.05 mg/kg apomorphine to assess the development of dopamine receptor supersensitivity (denervation supersensitivity), as expressed by the appearance of contraversive rotational behavior. Denervation supersensitivity is a good indicator of the size of the lesion, because with this dose it is seen only after 90–95% of dopamine terminals are destroyed (Marshall and Ungerstedt, 1977). Ten minutes after a subcutaneous injection of apomorphine, the number of full rotations were counted for 2 min. Animals that did not make at least five rotations were re-lesioned and re-screened. If they did not make at least five rotations when given apomorphine a second time, they were excluded from the study.

Within 1–4 days, all rats received an indwelling IV catheter in their right jugular vein using procedures described previously (Weeks 1972). Briefly, under ether anesthesia (supplemented with methoxyfluorothane), the silicone end of the catheter was inserted into the right external jugular vein. The PE 20 end of the tubing was passed subcutaneously, exiting through the skin at the nape of the neck, and was secured through the L-shaped tubing imbedded in the skull cap. Before being returned to their cage following surgery, the catheter was filled with 50 µl of a solution containing 50 mg/ml gentamicin. Catheters were then flushed daily with 0.1 ml of a heparin solution (30 USP/ml heparin in 0.9% saline, pH 7.4).

To check for catheter patency at the end of the experiment, animals received an IV infusion of 0.2 ml thiopental sodium (40 mg/kg dissolved in sterile saline). Animals that did not become ataxic within 10 s were excluded from the experiment.

Behavioral test procedures

Behavioral testing took a total of 17 days and was divided into the following phases.

Phase 1: habituation

At the beginning of the 5-day habituation period, the animals were assigned to one of two groups. The animals in one group (IV-Home) were transported to test chambers, located in a sound attenuated testing room, where they were housed for the duration of the experiment. Each test chamber consisted of a circular plastic bucket with a diameter of 25 cm at the base with granulated corn cob bedding. Food and water were available ad libitum and extraneous noises were masked using white noise. On day 2, each rat was tethered to a liquid swivel (modified from Brown et al. 1976) fixed to a moveable counter-balanced arm suspended above the animal by a lightweight flexible cable secured to the post imbedded in the skull cap. These animals remained tethered for the remainder of the experiment.

Every morning, beginning with habituation day 3, between 0800 and 0900 hours, each animal had its catheter flushed manually with 0.1 ml heparin solution, and it was attached to an infusion line (a length of PE 20 tubing) filled with saline, and fixed to the tether that connected the catheter to the liquid swivel. PE 20 tubing also connected the liquid swivel to a syringe mounted on a remote controlled syringe pump. The experimenter then left the room and did not return until the end of the day. The syringe pump was activated from outside of the room at 1100, 1300, or 1500 hours (in a counter-balanced order). The pump was programmed to deliver a total of 60 µl over 6 min. At the end of the day (between 1700 and 1800 hours) the experimenter entered the testing room and disconnected the infusion line from the catheter and the catheter was re-sealed with a stylet.

During the habituation period, the second group of animals (group IV-Novel) were housed in stainless steel hanging cages in the main animal colony and their catheters were flushed manually

with the heparin solution using the same schedule as for animals in the IV-Home group.

Phase 2: treatment

At the beginning of the treatment phase the animals in the IV-Home group and the IV-Novel group were assigned to one of eight subgroups (a total of 16 independent groups), representing eight dose conditions. Thus, depending on their group assignment each animal received: 0.0 (saline), 0.3, 0.6, 1.2, 2.4, 3.6, 4.8 or 7.2 mg/kg cocaine, once each day for a total of 5 consecutive days, using the following procedures. For animals in the IV-Home group, the procedure was essentially the same as during the habituation phase, except the lines were filled with one of the eight doses listed above. Every morning between 0800 and 0900 hours, each catheter was flushed with heparin, the portion of the line nearest the catheter was filled with cocaine (or saline, 0.0 mg/kg), and the remainder of the line was filled with the heparin solution and connected to the catheter and liquid swivel. The experimenter then left the room and did not return until the end of the day. The syringe pumps and behavioral monitoring equipment were turned on using controls located outside of the testing room. Animals were infused at 1100, 1300 or 1500 hours (counter-balanced across days) for 6 min at a rate of 10 µl/min. Thus, each daily infusion consisted of the 20 µl heparin solution that filled the catheter, followed by 9 µl of the drug/saline solution, and finally by an additional 31 µl heparin solution (a total of 60 µl). At the end of the day (1700–1800 hours), the animals were disconnected from the infusion line, and their catheters were again re-sealed. This procedure was repeated on each treatment day. Thus, neither the apparatus, the injection ritual nor the time of day reliably predicted drug administration in this group.

At the same time each day as animals in the IV-Home group were tested, animals in the IV-Novel group were taken from the animal room and transported to a testing room containing chambers identical to those in which the IV-Home group was housed. Their catheters were flushed with 0.1 ml heparin solution and the animals were then tethered and infused with one of the eight doses of cocaine/saline, exactly as for animals in the IV-Home group. Each test session lasted for a total of 60 min, after which time the animals in the IV-Novel group were disconnected from the infusion line, the catheters were re-sealed with stylets, and the animals were returned to the main animal room. Thus, for this group drug administration was "signaled" by transport to the testing room and by placement into the test chamber.

Phase 3: saline challenge

On the day following the last treatment, all animals received an infusion of saline to test for a conditioned rotational response to an infusion of saline. The procedures were identical to those described in the treatment phase above, except that all animals received saline.

Phase 4: drug challenge

Following the saline challenge, all animals were treated exactly as described for the Habituation phase (see above) for 4 days. The drug challenge was administered on day 6 after the last drug treatment. On the challenge test day, all animals in both the IV-Home and IV-Novel groups (both saline and cocaine pretreated animals) received an infusion of 0.6 mg/kg cocaine to test for the expression of sensitization as a function of treatment dose and environment. On the challenge test day, exactly the same procedures were used as described above for treatment days, the only difference being that all animals received 0.6 mg/kg cocaine.

Animals in the IV-Home groups were videotaped during each treatment and challenge test days. The video tapes were used to score the occurrence of convulsions in the IV-Home group. The experimenter used direct observation to score the occurrence of convulsions in the IV-Novel group. For both groups, rotational behavior was quantified using an automated device described previously (McFarlane et al. 1992) and one rotation was defined as four consecutive 90º turns in the same direction.

Results

Data are included only for those animals who passed the apomorphine screen and the catheter patency test. Also, complete data were not always available for every animal because of occasional equipment malfunction. Therefore, the final *n* for each group vary somewhat for different analyses, and the final *n* is indicated in the figure captions.

Doses of 3.6 mg/kg cocaine and higher produced convulsions in at least some animals. Approximately 50–60% of animals infused with doses of 3.6 mg/kg and 4.8 mg/kg showed convulsions, and all animals that received 7.2 mg/kg showed convulsions. Because doses of 3.6 mg/kg and higher were clearly toxic, the psychomotor activating effects of these doses were not analyzed. There was, however, an interesting effect of environment on cocaine lethality. As shown in Table 1, all animals that received 7.2 mg/kg showed convulsions, regardless of environmental condition. However, this dose of cocaine resulted in a significantly greater incidence of lethality when given in the IV-Home condition than in the IV-Novel condition. The two rats in the IV-Novel group died after the fifth injection of cocaine. Of the seven rats in the IV-Home group that died, one died after the first injection, two died after the second injection, two died after the fourth injection and two died after the fifth injection.

Effect of environment on the acute psychomotor response

Figure 1 shows the effects of the first IV infusion of cocaine or saline. The mean number of rotations were averaged over the first 15 min following drug administration, as this captured the entire time course of the drug response (see Figs. 2A and 3A). A two-way ANOVA resulted in no effect of Environment (F=1.180, P=0.2801), a significant effect of Drug dose (F=6.549, P=0.0001) and no interaction (F=0.167, P=0.9564). Thus, there was no significant effect of environment on the acute psychomotor response to cocaine.

Table 1 Incidence of cocaine-induced convulsions and lethality

	Convulse		Lethal*	
	Yes	No	Yes	No
IV–Home	9	0	7	2
IV–Novel	9	0	2	7

* P=0.03, Fisher exact probability test

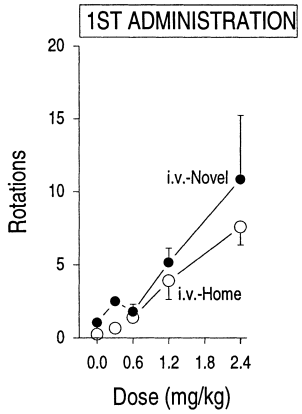

Fig. 1 The mean (±SEM) number of rotations per 3-min interval (averaged over the first 15 min of the test session) produced by an acute IV infusion of one of five different doses of cocaine (or saline). The *open circles* represent animals in the IV-Home group and the *closed circles* animals in the IV-Novel group. There was no effect of environment on the acute dose-effect curve. The *n* values for each group are: 0.0 mg/kg (Home *n*=11; Novel *n*=7), 0.3 mg/kg (Home *n*=7; Novel *n*=6), 0.6 mg/kg (Home *n*=8; Novel *n*=8), 1.2 mg/kg (Home *n*=10; Novel *n*=8), 2.4 mg/kg (Home *n*=12; Novel *n*=11)

Effect of environment on sensitization: within-subjects analysis

One index of the sensitization produced by repeated cocaine administration is provided by a within-subjects comparison of the psychomotor response on the first day of drug treatment with that on the last day of drug treatment (i.e., day 1 versus day 5 of treatment in the present study). This analysis is shown for the IV-Home group in Fig. 2 and for the IV-Novel group in Fig. 3.

Figure 2 shows the time course of the psychomotor response on the first (A) and the fifth (B) day of treatment for the IV-Home group. It is apparent that the psychomotor response to all doses of cocaine peaked very rapidly (within 3–6 min), and for most doses activity returned to control levels within 6–9 min. For the highest dose (2.4 mg/kg), the psychomotor response persisted a little longer (approximately 15 min). Sensitization is characterized by both an increase in the magnitude of a psychomotor response and a more rapid onset of the response (Leith and Kuczenski 1982), and therefore, a comparison of the rotational response during the first 3-min interval (as a function of dose and day of treatment) is shown in Fig. 2C. In this comparison, sensitization is indicated by a significant increase in psychomotor response between day 1 and day 5 of treatment. A two-way ANOVA yielded a main effect of Day (F=7.34, P=0.019), an effect of Dose (F=9.365, P<0.0001) and a significant Day by Dose interaction (F=4.766, P=0.0026). A comparison of the responses on day 1 versus day 5 (for a given dose) using *t*-tests with Bonferroni corrections indicated that only rats treated with 2.4 mg/kg showed a significant difference between day 1 and day 5 (t=–4.670, P=0.035). That is, only this group showed evidence of sensitization. It is obvious from a comparison of Fig. 2A and B that in rats

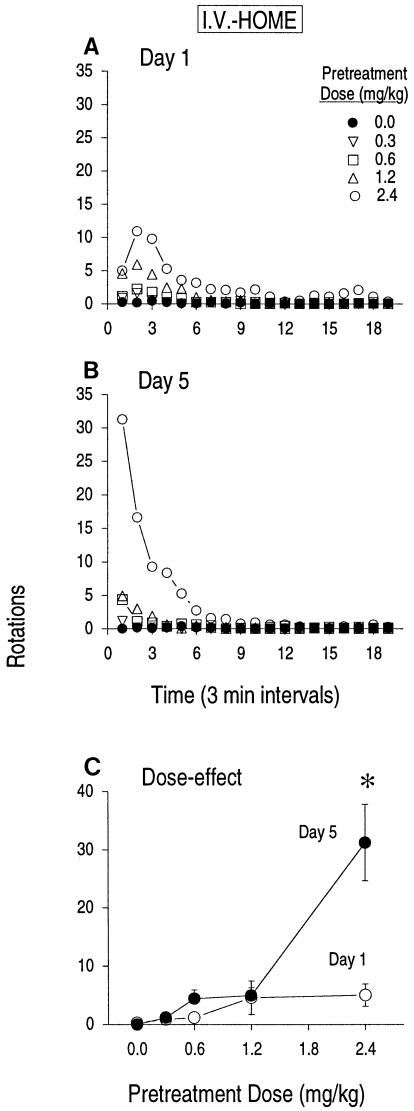

Fig. 2A–C The effects of different doses of cocaine on rotational behavior following the first versus fifth injection in the IV-Home group. **A** The mean number of rotations per 3-min interval over the 1-h test session on day 1 in animals receiving saline (dose 0.0 mg/kg; *n*=11) or 0.3 (*n*=7), 0.6 (*n*=8), 1.2 (*n*=10), or 2.4 (*n*=12) mg/kg cocaine IV. The group that received saline is indicated by the *closed circles*. **B** The mean number of rotations per 3-min interval on day 5 for the same animals shown in **A**. **C** Mean (±SEM) number of rotations for the first 3-min interval on day 1 (*open circles*) and day 5 (*closed circles*) as a function of dose. The *asterisk* (∗) indicates that only rats treated with 2.4 mg/kg showed a significant difference in their response on day 1 versus day 5

treated with 2.4 mg/kg sensitization was characterized by both an increase in the magnitude of the response and by a more rapid onset of rotational behavior.

Figure 3 illustrates the time course of the psychomotor response on day 1 (A) and day 5 (B) of treatment for the IV-Novel group. It is apparent that the psychomotor response to all doses of cocaine peaked very rapidly (within 3–6 min), and for most doses activity returned to control levels within 6–12 min. For the highest dose (2.4 mg/kg),

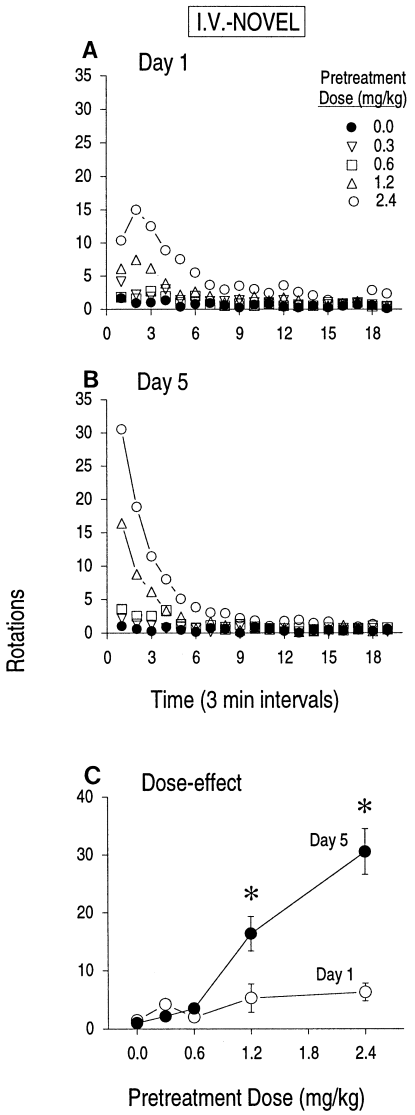

Fig. 3A–C The effects of different doses of cocaine on rotational behavior following the first versus fifth infusion in the IV-Novel group. **A** The mean number of rotations per 3-min interval over the 1-h test session on day 1 in animals receiving saline (dose 0.0 mg/kg; *n*=7) or 0.3 (*n*=6), 0.6 (*n*=8), 1.2 (*n*=8), or 2.4 (*n*=11) mg/kg cocaine IV. The group that received saline is indicated by the *closed circles*. **B** The mean number of rotations per 3-min interval on day 5 for the same animals shown in **A**. **C** Depicts the mean (±SEM) number of rotations for the first 3-min interval on day 1 (*open circles*) and day 5 (*closed circles*) as a function of dose. The *asterisks* (∗) indicate that rats treated with doses of 1.2 and 2.4 m/kg showed a significant difference in their response on day 1 versus day 5

the psychomotor response persisted a little longer (approximately 18–21 min). For the comparison in Fig. 3C, sensitization is indicated by a significant increase in the psychomotor response between day 1 and day 5 of treatment. An overall ANOVA yielded a significant effect of Day (F=12.773, P=0.0051), a significant effect of Dose (F=16.990, P<0.001) and a significant Day by Dose interaction (F=9.575, P<0.0001). A comparison of day 1 versus day 5 responses (for a given dose) using t-tests with Bonferroni corrections indicated that rats treated with both 1.2 mg/kg (t=–4.127, P=0.0176) and 2.4 mg/kg (t=–7.654, P<0.001) showed a significant difference between day 1 and day 5. It is obvious from a comparison of Fig. 3A and B that in rats treated with 1.2 mg/kg and 2.4 mg/kg sensitization was characterized by both an increase in the magnitude of the response and by a more rapid onset of rotational behavior.

In summary, the within-subjects analysis indicates that for the IV-Home group a dose of 2.4 mg/kg was necessary to induce sensitization, whereas for the IV-Novel group a dose of 1.2 mg/kg was sufficient.

Effect of environment on sensitization: between-subjects analysis

A second, and perhaps more conservative index of sensitization, is to compare the response of saline and drug-pretreated animals to a challenge infusion of a fixed dose of the drug. In this case, sensitization is indicated if drug-pretreated animals show a significantly greater psychomotor response to the drug challenge than saline-pretreated animals. In addition, in the present experiment animals were pretreated with different doses of cocaine, and therefore, one can also construct a dose-effect curve for the *induction* of sensitization; i.e., determine what pretreatment dose is necessary to induce sensitization.

Figure 4 shows the effects of a challenge infusion of 0.6 mg/kg of cocaine as a function of pretreatment dose and environmental condition. Figure 4A and B show the time course of the behavioral response in IV-Home and IV-Novel groups, respectively. It is obvious from inspection of panels A and B that in all groups the peak psychomotor response occurred in the first 3-min interval after drug administration, and then declined rapidly to negligible levels (within 9 min). Figure 4C shows the dose-effect curve for the induction of sensitization, based on analysis of the first 3-min interval (i.e., the peak drug effect). In this analysis, sensitization is indicated if the response of drug-pretreated animals is significantly greater than the response of animals pretreated with a dose of 0.0 mg/kg (saline). A two-way ANOVA resulted in a non-significant overall effect of Environment (F=1.945, P=0.1672), a significant main effect of Dose (F=10.32, P<0.0001) and a significant Environment by drug Dose interaction (F=2.873, P=0.0284). Follow-up tests were made using Bonferroni-corrected t-tests. These indicated that in the IV-Home group, only animals pretreated with 2.4 mg/kg differed significantly from the saline-pretreated controls (t=–3.794, P=0.0064). In the IV-Novel group, however, animals pretreated with 1.2 mg/kg (t=–4.268, P=0.002) and 2.4 mg/kg (t=–2.529, P=0.0648) differed from saline-pretreated controls (for all other comparisons, P>0.2). It is obvious from inspection of Fig. 4A and B (compare the response to a 0.6 mg/kg cocaine challenge in saline-pretreated animals to that in cocaine-pretreated animals) that sensitization was characterized primarily by an increase in the peak drug effect during the first 3-min interval.

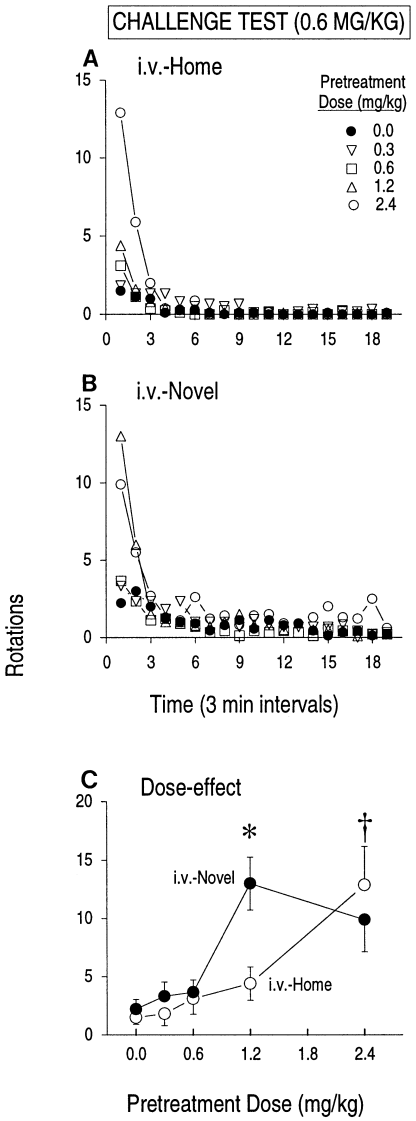

Fig. 4A–C The effects of a challenge infusion of 0.6 mg/kg as a function of treatment with different doses of cocaine on rotational behavior in both the IV-Home and IV-Novel groups. **A** The mean number of rotations per 3-min interval over the 1-h test session in response to 0.6 mg/kg in animals treated with saline (dose 0.0 mg/kg; *n*=10), 0.3 (*n*=6), 0.6 (*n*=8), 1.2 (*n*=10), or 2.4 (*n*=8) mg/kg cocaine IV in the IV-Home group. The group that received saline is indicated by the *closed circles*. **B** The mean number of rotations per 3-min interval over the 1-h test session in response to 0.6 mg/kg in animals treated with saline (dose 0.0 mg/kg; *n*=9), 0.3 (*n*=6), 0.6 (*n*=9), 1.2 (*n*=10), or 2.4 (*n*=10) mg/kg cocaine IV in the IV-Novel group. The group that received saline is indicated by the *closed circles*. **C** Depicts the mean (±SEM) number of rotations for the first 3-min interval in response to a fixed dose of cocaine as a function of pretreatment dose. The *open circles* represent animals in the IV-Home group, while the *closed circles* reflect the same data for animals challenged in the IV-Novel group. † Indicates that in the IV-Home group only rats treated with 2.4 mg/kg differed from the saline-pretreated control group. ∗ Indicates that in the IV-Novel group rats treated with 1.2 mg/kg differed from saline-treated animals

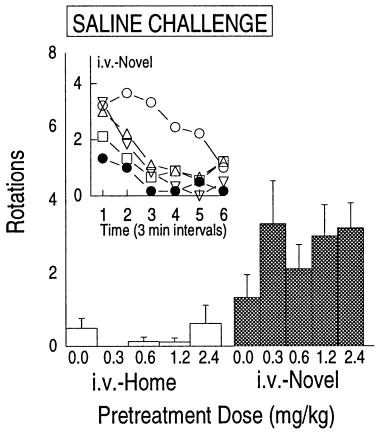

Fig. 5 The mean (±SEM) number of rotations produced by an infusion of saline as a function of cocaine pretreatment and environmental condition. The *open bars* represent the peak response (first 3-min interval) of animals in the IV-Home group pretreated with one of five doses of cocaine (or saline). The *closed bars* represent the response of animals in the IV-Novel group. The *n* values are as follows: saline (Home *n*=9; Novel *n*=6), 0.3 mg/kg (Home *n*=6; Novel *n*=6), 0.6 mg/kg (Home *n*=8; Novel *n*=9), 1.2 mg/kg (Home *n*=9; Novel *n*=10), and 2.4 mg/kg (Home *n*=8; Novel *n*=9). The *insert* illustrates the time course (in 3-min intervals) of the behavioral response to an infusion of saline as a function of pretreatment condition only for animals in the IV-Novel group. The *closed circles* represent the animals pretreated with saline and the *open symbols* animals pretreated with cocaine, ▽ 0.3 mg/kg, □ 0.6 mg/kg, △ 1.2 mg/kg, and ○ 2.4 mg/kg

In summary, this between-subjects analysis was consistent with the within-subjects analysis presented above. That is, for the IV-Home group a dose of 2.4 mg/kg was necessary to induce sensitization whereas for the IV-Novel group a dose of 1.2 mg/kg was sufficient.

Effect of environment on development of conditioned psychomotor activation

To determine whether the drug administration procedure resulted in conditioned psychomotor activation, animals in both groups received a "saline challenge" following the drug treatment phase. The response to the saline challenge is shown in Fig. 5. In the IV-Home group there was essentially no behavioral effect of an infusion of saline and no effect of cocaine pretreatment (F=0.931, P=0.4571). In the IV-Novel group there was no significant effect of pretreatment when only the peak effect was considered (i.e., the first 3-min interval; see Fig. 5; F=0.998, P=0.4219). When the entire time course was analyzed, however (Fig. 5 insert), there was evidence for a small but statistically significant conditioned response only in animals pretreated with 2.4 mg/kg cocaine (F=4.621, P=0.0510).

Discussion

In earlier studies, we found that the unsignaled IV administration of low doses of amphetamine (0.5–1.0

mg/kg) failed to induce sensitization when given to rats in their home cage (Crombag et al. 1996; Robinson et al. 1998). These doses were sufficient, however, to induce sensitization if drug administration was signaled by placement of the animal into a novel test environment. The purpose of the present experiment was two-fold: first, to determine whether the ability of IV *cocaine* to induce sensitization is modified by the circumstances surrounding drug administration in a similar way as are the effects of amphetamine, and second, to determine whether it is impossible to induce sensitization if cocaine administration is unsignaled, or, whether the effect of environment is to shift the dose-effect curve for the induction of sensitization. The results were clear. First, the ability of cocaine to induce sensitization was modified by the circumstances surrounding drug administration in a similar way as reported previously with amphetamine. Second, the effect of environment was to shift the dose-effect curve for the induction of sensitization. That is, a higher dose was required to induce sensitization if cocaine was given at home than if it was given in a novel test environment. At the highest dose tested, however, cocaine induced sensitization regardless of environmental condition. We have recently obtained a similar effect with amphetamine (Browman et al. 1997).

These results are consistent with our previous report that environmental condition can modify cocaine sensitization. Badiani and colleagues (1995b) reported that the IP administration of 20 mg/kg cocaine to animals living in the test environment (i.e., at home), induced sensitization, but significantly less robust sensitization than if the same dose was given in a novel test environment. In that study, however, an IP injection was accompanied by a number of cues predictive of drug administration even in animals living in the test chambers, for example, the appearance of an experimenter, handling, and a needle prick. In the present experiment, these cues were eliminated, and the absence of these cues may magnify the effect of environment on sensitization. Nevertheless, pretreatment with 2.4 mg/kg was sufficient to induce sensitization in both the IV-Home and IV-Novel groups, and the magnitude of the behavioral response to a 0.6 mg/kg challenge was comparable in both groups, suggesting a comparable degree of sensitization. However, a challenge test with a single dose is really not sufficient to make strong claims about the effect of environment on the *magnitude* of sensitization. To address this issue properly would require generating a dose-effect curve for the *expression* of sensitization as a function of pretreatment condition.

It is interesting to note that the dose required to induce sensitization in the IV-Home group (2.4 mg/kg) was very high. The next highest dose tested (3.6 mg/kg) produced convulsions in some animals. This suggests that, depending on the conditions under which drugs are administered, very high doses may be required to induce sensitization. It also suggests that treatment with a behaviorally effective dose (that is, a dose capable of producing psychomotor activation) is not necessarily sufficient to produce sensitization. Of course, in the present experiment the animals were given only five treatments, and it is possible that with more treatments lower doses may begin to induce the neuroadaptive processes that are responsible for behavioral sensitization. We know, for example, that many factors can influence the induction of sensitization besides dose and environmental condition (for review, see Robinson 1988). Nevertheless, the present results emphasize that the ability of a given dose of cocaine to induce sensitization can be powerfully modulated by the circumstances surrounding drug administration, and therefore, in studying sensitization the use of relatively high doses may sometimes be advantageous (Robinson et al. 1998).

Although the environmental conditions manipulated here influenced the susceptibility to sensitization, it is important to emphasize that there was no effect of environment on the *acute* psychomotor response to cocaine. This is consistent with the earlier study by Badiani and colleagues (1995b), who reported that the initial psychomotor response to 20 mg/kg cocaine IP was the same in animals given the drug at home or in a novel environment. This is in contrast to the effect of environment on the acute psychomotor response to amphetamine (Badiani et al. 1995a, b; Crombag et al. 1996). Nevertheless, even in the case of amphetamine, the susceptibility to sensitization appears to be independent of the acute drug response (Badiani et al. 1995a; Robinson 1984, 1988). Perhaps the most compelling evidence that the susceptibility of sensitization is independent of the acute psychomotor response to stimulant drugs comes from studies with inbred strains of mice. For example, based on studies with recombinant inbred lines, Schuster et al. (1977, p. 185) suggested that, “the initial response to cocaine and the development of sensitization are controlled by different genetic determinants (also see, Short and Schuster 1976).” This suggestion has been supported by a number of more recent studies (Logan et al. 1988; Phillips et al. 1995).

It is not clear by what mechanism(s) environment (IV-Home versus IV-Novel; home versus novel) modulates the induction of sensitization. It is possible that the differences between the IV-Home and IV-Novel groups in the induction of sensitization may be related to differences in the availability of associative cues in the two environments (Badiani et al. 1995b). To the extent that classical conditioning contributes to sensitization, one might expect this to have a greater effect when drug administrations are signaled then when they are unsignaled. Consistent with this idea, only animals in which drug administrations were signaled by placement in a novel environment showed a conditioned response when given a challenge infusion of saline. On the other hand, the only group that showed a significant conditioned response was that pretreated with 2.4 mg/kg cocaine. Animals pretreated with 1.2 mg/kg in the novel environment also developed sensitization, but did not show a conditioned response. Furthermore, the unsignaled administration of 2.4 mg/kg at home was sufficient to induce sensitization,

but these animals did not exhibit a conditioned response. This suggests that the development of a conditioned response is not necessary for a sensitized response.

Another variable that may contribute to the effect of environment on sensitization is the action of a novel environment as a stressor. It is known that exposure to a novel environment is a potent stimulus for activating the hypothalamic-pituitary-adrenal (HPA)-axis, producing neuroendocrine and neural changes indicative of stress. Animals reliably exhibit elevated levels of corticosterone following their first exposure to a novel environment (Friedman and Ader 1967), and a greater increase in the plasma corticosterone response with repeated intermittent exposures (Hennessy 1991). It has further been suggested that the development of behavioral sensitization is enhanced by stress-induced corticosterone secretions under some conditions (for review, see Piazza and LeMoal 1996), and cross-sensitization has been reported between stress and psychostimulant drugs (Robinson 1988). Whatever the contribution of a novel environment as a stressor, it appears that a stress-induced increase in plasma corticosterone is not a significant factor. Adrenalectomy has no effect on the rate of sensitization in novel or home environments (Badiani et al. 1995c). It is possible, however, that another stress-related factor does contribute to the effect of environment on sensitization such as stress-induced corticotropin-releasing factor secretion in non-hypothalamic systems (Cador et al. 1993).

One additional interesting finding in the present study was the effect of environment on cocaine lethality. There was no effect of environment on the incidence of cocaine-induced convulsions. The administration of 3.6, 4.8 or 7.2 mg/kg cocaine IV produced a similar incidence of convulsions regardless of environmental condition. The extent to which convulsions were lethal, however, was greatly influenced by environmental condition. Although all animals that received 7.2 mg/kg had convulsions, 77% of the animals died in the IV-Home group, whereas only 22% of the animals in the IV-Novel group died. Dworkin and colleagues (1995) reported a related effect of environment on cocaine-induced lethality. They found that animals self-administering cocaine were significantly less likely to die than animals which received experimenter-delivered (yoked) cocaine. Dworkin and colleagues (1995) suggested that the critical variable may be whether drug administration is response-dependent or response-independent. The current findings suggest, however, that the effect of environment on lethality may not be due to response-dependent factors, but to the predictability of infusions. In the present study, cocaine administration was not contingent upon the action of the animal in either group, but there were still marked group differences in cocaine lethality. It is not clear what effects of cocaine are responsible for its lethality, and further studies will be necessary to determine the nature of the critical variables.

In conclusion, the current study further supports the notion that environmental factors can powerfully modulate the induction of sensitization to the psychomotor activating effects of psychostimulant drugs. Although it is not clear what mechanisms mediate environmental control of sensitization, the results reported here emphasize that sensitization is not a simple inevitable consequence of exposure to psychostimulant drugs, but is the result of complex interactions between the neuropharmacological effects of drugs and the circumstances surrounding drug administration. To the extent that sensitization-related neuroadaptations in brain reward systems contribute to the development of addiction (Robinson and Berridge 1993), further study of how environmental factors contribute to sensitization may yield important new insights into how they may also contribute to addiction.

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