*Pharmacology Biochemistry & Behavior*, Vol. 37, pp. 707–711. © Pergamon Press plc, 1990. Printed in the U.S.A. 0091-3057/90 $3.00 + .00

# Preexposure Sensitizes Rats to the Rewarding Effects of Cocaine

BRIAN A. HORGER, KEITH SHELTON AND SUSAN SCHENK

*Texas A&M University, Department of Psychology, College Station, TX 77843*

Received 26 April 1990

HORGER, B. A., K. SHELTON AND S. SCHENK. *Preexposure sensitizes rats to the rewarding effects of cocaine.* PHARMACOL BIOCHEM BEHAV **37**(4) 707–711, 1990.—During a preexposure period rats were injected once daily with either cocaine HCl (10 mg/kg, IP) or the saline vehicle for 12 consecutive days. Rats that were chronically exposed to cocaine during the pretreatment phase were more responsive to the motor activating effects of a subsequent injection of cocaine than were rats chronically treated with saline. In self-administration testing, saline-pretreated groups did not exhibit a significant preference for a lever producing a cocaine infusion relative to an inactive lever, suggesting that the doses tested (0.225 and 0.45 mg/kg/infusion) were subthreshold for cocaine reward. In contrast, subjects preexposed to cocaine had a higher rate of reinforced responses and exhibited a preference for a lever that resulted in a cocaine infusion. It was unlikely that the higher response rate was due to an elevation in nonspecific activity since inactive lever responding remained low and relatively invariant over the 9 days of testing. Thus the enhanced responding in the cocaine-preexposed rats suggests that the reinforcing effectiveness of the drug had increased. These data indicate that sensitivity to cocaine's behavioral effects can be enhanced and that predisposing factors to cocaine abuse can be manipulated.

Self-administration Cocaine Sensitization

THERE is substantial variability in the behavioral effects of cocaine. Some animals appear to be extremely sensitive in that they respond to very low doses of the drug, whereas others require higher doses to exhibit the same effect. That is, some rats appear sensitized or predisposed to the behavioral effects of cocaine.

Variability in response to cocaine's rewarding effect is apparent in that not all subjects learn to perform an operant to self-administer the drug. Thus the reinforcing effectiveness of cocaine is not the same for all rats. Animals that self-administer cocaine can be separated from their nonself-administering counterparts on the basis of housing conditions. Rats reared in a socially isolated condition were more prone to self-administer cocaine than rats reared in an aggregated condition (15). Thus a sensitization to cocaine reward has been experimentally produced.

Another means of inducing sensitization is by preexposing rats to cocaine. The bulk of work examining this phenomenon of reverse tolerance has measured the activating effects of this psychostimulant. At low and moderate doses single injections of cocaine increase activity (17). An augmentation of this effect is observed with subsequent administration, with chronic low dose exposures producing an intense form of behavioral sensitization (12). A prolonged period of preexposure is not required since sensitization has been observed after as little as a single, high dose exposure to cocaine (8). Sensitization to cocaine has also been demonstrated in the conditioned place preference paradigm (7).

These findings raised the possibility that cocaine preexposure may also serve to alter the reinforcing impact of the drug. The present experiment is an investigation of this hypothesis. We examined the effect of chronic cocaine exposure on its reinforcement effectiveness using the self-administration paradigm. In addition, some rats were tested to confirm behavioral sensitization to cocaine's motor-activating properties under identical preexposure parameters.

Preexposure for both behavioral indices of drug action was associated with specific cues in order to accommodate potential conditioning factors (5,13).

## METHOD

### *Subjects*

Male Sprague-Dawley rats (Harlan, TX) weighing 325–375 g were acclimated to the laboratory setting for one week prior to the onset of their respective procedures. The colony room was maintained on a 12-h light/dark schedule. The animals were housed individually with food and water available ad lib. The rats were randomly assigned to one of two testing procedures: 1) cocaine-induced locomotor activity and 2) cocaine self-administration.

### *Surgery*

Animals used in the self-administration phase of the procedure were implanted with chronic indwelling intravenous catheters according to a modification of Weeks (18). The rats were pretreated with atropine HCl (1.4 mg/kg, IP) 10 min before being anesthetized with separate injections of sodium pentobarbitol (20 mg/kg, IP) and ketamine (60 mg/kg, SC). A silastic catheter was inserted into the left jugular vein. The distal end was guided subcutaneously to an exposed portion of the skull and secured in place with dental acrylic. Each day following surgery the patency of the catheter was confirmed following a 0.05 ml infusion of a saline

solution containing heparin (1.25 units/ml) and penicillin G sodium (250,000 units/ml). All surgery was performed under sterile conditions following animal care guidelines.

*Apparatus*

*Activity monitors.* Ten 38.1 × 38.1 × 38.1 cm black wooden boxes with a metal grid floor served as behavioral testing chambers. These boxes were equipped with four sets of light sources and photocells located 5.1 cm above the floor and 12.7 cm from each corner. The photocells and receptors were arranged so that each pair of light beams crossed the other perpendicularly, dividing the activity chambers into nine equal spaces. As a beam was intersected it was recorded by a mechanical counter, providing a gross measure of locomotor activity. Testing was conducted under dimly lit conditions. Readings from the counters were taken every 10 min in an adjacent monitoring room.

*Self-administration chambers.* Self-administration testing was conducted in 16 operant boxes (Med Associates) enclosed in sound-attenuating chambers. Each box was equipped with two levers. Depression of one lever (the active lever) activated a mechanical infusion pump (Razel, model A equipped with 1 rpm motors and 20 ml syringes) which produced a 0.1 ml infusion of cocaine HCl through a swivel (2) suspended above each box. Coincident with the 12-s infusion, a stimulus light located above the lever was illuminated. Depression of the other lever (the inactive lever) was without effect. Drug delivery and data collection were controlled by the Operant Package for the Neurosciences software package [modified from (3,16)].

*Procedure*

*Activity measures.* The procedure followed that of Post and Rose (12). Before each session the activity boxes were wiped with a solution of 1% ammonium hydroxide in water to minimize the influence of remaining odors from preceding groups. Subjects were weighed and brought to the testing room in a carrying case. After being placed in the chambers, locomotor activity was monitored at 10-min intervals during a 20-min habituation period. Injections of either 10 mg/kg cocaine HCl (IP) (N = 10) or an equal volume (1 ml/kg) of the saline vehicle (N = 10) were then administered and activity recorded for the subsequent 40 minutes. This protocol was carried out for 12 days. On day 13 all animals received a drug challenge of 10 mg/kg cocaine HCl (IP) following the habituation period. Activity counts were again recorded for 40 min postinjection and compared for the saline and cocaine-preexposed rats.

*Self-administration.* Animals used in the self-administration procedure were divided into 2 groups. For 12 consecutive days the rats were injected with either cocaine HCl (10 mg/kg, IP) or saline and then were placed in the operant chambers for 30 min before being returned to the animal colony. Before each injection the operant chambers were wiped with a solution of 1% ammonium hydroxide in water to minimize potential odor cues. The levers were removed during this period. Following 12 days of preexposure to either cocaine or saline and an ensuing 2-day rest period, animals were tested for spontaneous acquisition of cocaine self-administration.

Doses available were 0.225 or 0.45 mg/kg/infusion. These doses were chosen since they fall below those that have been reported to reliably maintain self-administration (10,11). Thus if cocaine preexposure served to sensitize these rats to the reinforcing effects of cocaine, the use of these doses, at the low end of the dose/response curve should optimize the ability to observe an increase in reinforced responding.

During the self-administration phase, rats were given a single priming infusion of cocaine at the start of each 2-h daily session.

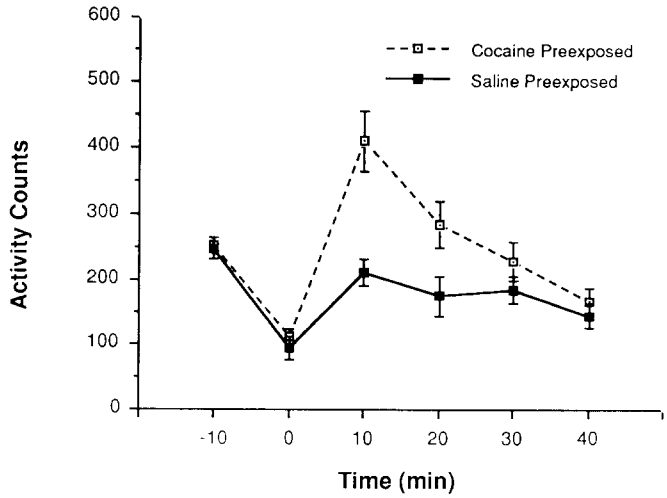


FIG. 1. Activity counts per 10 min during a 20-min preinjection and 40-min postinjection period. Data points represent the mean response (± SEM) to cocaine (10 mg/kg) in rats that had received 12 previous experiences with either cocaine or saline. The cocaine preexposed rats showed a greater response to the drug at 10 and 20 min postinjection ($p<0.05$).

Testing was continued for 9 days. Several rats from both conditions failed to complete the entire testing procedure due to catheter malfunction. The number of rats that completed the protocol were: cocaine-pretreated/0.225 mg/kg: 9; cocaine-pretreated/0.45 mg/kg: 5; saline-pretreated/0.225 mg/kg: 8; and saline-pretreated/0.45 mg/kg: 5.

## RESULTS

*Activity Measures*

Figure 1 shows the mean activity counts in response to the cocaine injection on day 13 for the cocaine and saline-pretreated rats. During the preinjection tests, motor activity was comparable for the two groups. Following the cocaine injection, activity increased in both groups, with the cocaine-pretreated animals exhibiting a higher cocaine-induced level of activity. A 2-way ANOVA (time × pretreatment condition) on the activity scores yielded a significant main effect for pretreatment condition, $F(1,18) = 6.7$, $p<0.05$, and a significant interaction between pretreatment condition and time, $F(5,90) = 7.5$, $p<0.01$. Scheffé post hoc comparisons revealed elevated activity counts for the cocaine-pretreated group at 10 and 20 min postinjection ($p<0.05$).

*Self-Administration*

Figure 2 depicts the mean number of reinforced and nonreinforced lever responses for each of the 4 groups (cocaine-pretreated/0.225 mg/kg, cocaine-pretreated/0.45 mg/kg, saline-pretreated/0.225 mg/kg, and saline-pretreated/0.45 mg/kg) over the 9 days of self-administration testing. Following an initial decrease in responding, reinforced lever responses increased over days for both of the groups that received cocaine pretreatment (left panels). Asymptotic responses rates on the active lever were reached by day 6 of testing. In contrast, inactive lever responses were low and relatively invariant over days. Individual 2-way ANOVAs (lever × days) on the response rates of each of the 2 groups showed that reinforced responses were significantly higher than nonrein-

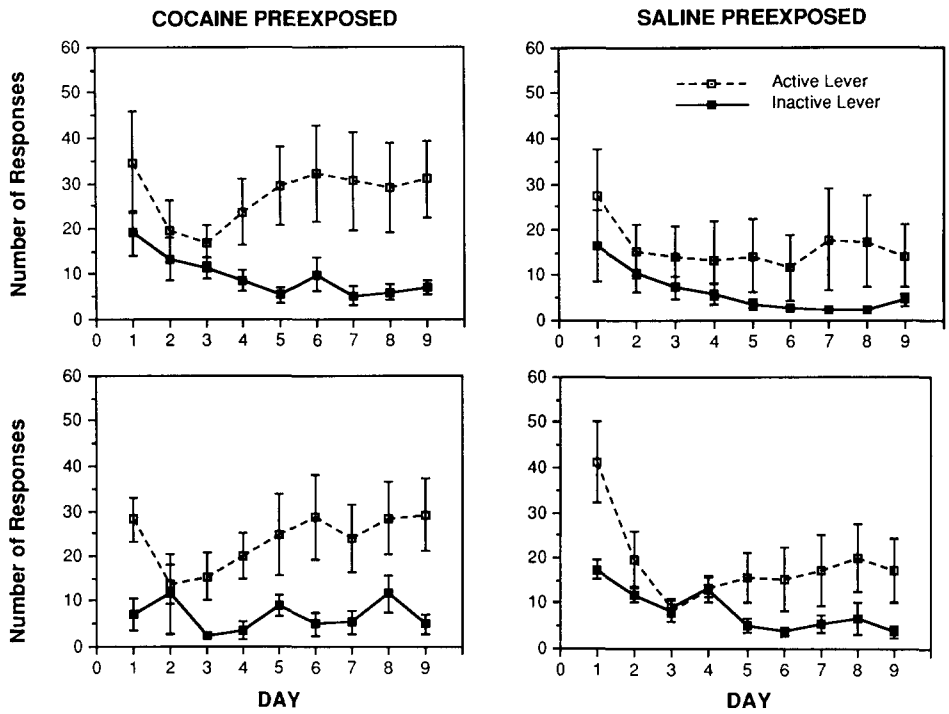


FIG. 2. Mean reinforced (active) and nonreinforced (inactive) lever responses (±SEM) for 1 of 2 intravenous doses of cocaine [0.225 (top) or 0.45 (bottom) mg/kg/infusion] as a function of days. Rats that had been pretreated with 12 daily injections of cocaine (10 mg/kg; IP) are depicted on the left side of the panel and the saline-pretreated groups are presented on the right. Rats in the cocaine-pretreated groups showed a preference for the active lever ($p<0.05$), whereas the saline-pretreated rats did not.

forced responses [0.225 mg/kg/infusion, $F(1,16)=5.5$, $p<0.05$; 0.45 mg/kg/infusion, $F(1,8)=5.6$, $p<0.05$]. In addition, a significant interaction between the 2 variables was found in cocaine-pretreated rats in the 0.225 mg/kg/infusion group, $F(8,128)=2.7$, $p<0.01$. Scheffé post hoc comparisons indicated elevated active lever responses relative to inactive lever responding on days 5 through 9 ($p<0.05$).

In contrast to the data from the cocaine-pretreated rats, neither of the 2 saline-pretreated groups (right panels) showed a significant preference for the active lever [0.225 mg/kg/infusion, $F(1,14)=1.5$, N.S.; 0.45 mg/kg/infusion dose, $F(1,8)=2.7$, N.S.].

A 3-way ANOVA (pretreatment condition × days × dose with repeated measures on the days factor) revealed a significant interaction between pretreatment condition and days, $F(8,184)=2.1$, $p<0.05$. Scheffé post hoc tests revealed that the number of reinforced responses in the cocaine-pretreated group was higher than in the saline-pretreated group on day 6 ($p<0.05$).

In order to determine the number of animals in each group which acquired the operant to self-administer cocaine, a criterion was defined based on the inactive lever responses. On the last day of self-administration testing, a 90% one-tailed confidence interval was placed about the mean number of inactive lever responses for each of the four groups. Subjects that responded on the reinforced lever at a rate above the upper limit of this confidence interval were operationally defined as having acquired cocaine self-administration.

A higher percentage of cocaine pretreated subjects acquired the operant to self-administer cocaine relative to the saline-pretreated animals at both doses tested. Seventy-eight and 60% of the cocaine-pretreated rats lever pressed above the criterion for a 0.225 and 0.45 mg/kg/infusion of cocaine, respectively. In contrast, only 25% (0.225 mg/kg/infusion) and 40% (0.45 mg/kg/infusion) of the saline-pretreated controls acquired cocaine self-administration at the same doses. Chi-square analysis indicated that these differences between cocaine and saline-pretreated rats were reliable, $\chi^2(3)=16.58$, $p<0.01$.

## DISCUSSION

Consistent with previous reports (5,12) the locomotor response to a cocaine challenge injection was enhanced by cocaine pretreatment. Thus the parameters and dose of the cocaine preexposure subsequently used in the self-administration tests were sufficient to produce behavioral sensitization.

In examining whether prior exposure could similarly sensitize rats to cocaine's reinforcing effects, low doses were employed during self-administration testing. These doses were subthreshold as indicated by their inability to sustain a preference for the reinforced lever in saline-pretreated rats.

In contrast, rats that had been preexposed to cocaine demonstrated higher rates of reinforced responding and a significant preference for the response that resulted in the delivery of a cocaine infusion at both doses tested. The acquisition of cocaine self-administration in these rats was gradual, with maximal reinforced response rates obtained by the sixth day of testing.

That the cocaine preexposed rats had higher reinforced response rates can be interpreted in at least two ways. First, it is possible that the enhanced responses simply reflect a more nonspecific effect of increased activity in response to cocaine, as the tests of cocaine-induced motor activation suggested [present results; (12)]. This possibility is unlikely since inactive lever responding remained low in both cocaine- and saline-pretreated groups.

A more likely possibility is that the pretreatment altered the reward impact of cocaine. An increase in responding for drug infusions has been interpreted to reflect both an increase and a decrease in reinforcement effectiveness of the drug, with the latter being the more common interpretation. Usually doses at the high end of the dose response curve are used in self-administration tests. As the dose is reduced or manipulations are performed which reduce the effectiveness of the drug dose, a shift to the right of the dose/response curve occurs, resulting in an increase in responding. However, responding is not an inverse function of dose across the entire dose/response curve. This relationship holds within the effective dose range. At the low end, subthreshold doses fail to support responding. Thus the curve is in the shape of an inverted U, with both high and low doses supporting low rates of responding. Depending on whether the dose being used is on the rising or falling portion of the dose/response curve, manipulations that increase responding can be interpreted to indicate either an increase or decrease, respectively, in the effectiveness of the self-administered drug.

Since the control rats in the present study failed to respond for cocaine at the doses tested (doses at the extreme left of the dose/response curve), within the time course of the present experiment (9 days), the results lend themselves to only one interpretation. An increase in responding indicates that the reward effectiveness of cocaine had increased as a result of prior cocaine exposure (the dose/response curve had shifted to the left).

Given the above argument, it was somewhat surprising that reinforced responding in the cocaine preexposed groups was not dose dependent. Response rates averaged about 30 in the 2-h test period. With a change in doses, compensatory responding has usually been reported. One possible explanation for the failure to observe similar compensatory responding in the present study is that the cocaine-preexposed subjects were responding for an alternative consistent reinforcer such as the illumination of the house light. However, this is unlikely since data from our lab has indicated a lack of responding for saline paired with house light illumination in rats previously trained to self-administer amphetamine (19). From present data it is not possible to determine why dose/response relationships were not present. We have, however, consistently observed this when acquisition of the behavior is tested. In contrast, when the dose is manipulated in experienced rats, dose-dependent responding is consistently found in our laboratory (15,19).

It is tempting to conclude that drug preexposure had sensitized rats to the reward impact of cocaine by altering relevant reward-related neural systems. Self-administration of cocaine has been linked specifically to the drug's effect on the mesolimbic dopamine system (20). Chronic preexposure under conditions which produce behavioral sensitization increased the response of this dopamine system to cocaine as measured by in vivo microdialysis (6). Consistent with this finding, a decrease in the ability of autoreceptor selective doses of apomorphine to attenuate firing rates of ventral tegmental area neurons has been observed following repeated cocaine administration (4). Auto receptor down-regulation and a reduction in inhibitory feedback influences from the nucleus accumbens are possible mechanisms mediating this effect.

A similar mechanism may account for the recent finding of sensitization to the rewarding properties of amphetamine induced by preexposure (9). Chronic amphetamine pretreatment also enhanced the response of the mesolimbic dopamine system to subsequent amphetamine injections (14). Thus sensitization to the rewarding properties of a number of drugs of abuse may be an important factor maintaining drug-taking behavior. Cross-sensitization between these drugs may also be expected.

This study provides a second demonstration of predisposing factors in cocaine self-administration. A previous report (15) showed that housing conditions in weanling rats could also explain variability in response to cocaine in the self-administration paradigm. However, the effect of this manipulation may be restricted to immature animals since rats that were differentially housed in maturity were equally sensitive to cocaine's reinforcing effectiveness (1). Thus it seems that the response to the reinforcing properties of cocaine can be experimentally manipulated under specific circumstances.

It will be of great interest to more fully investigate the conditions under which sensitization to the reinforcing effects of cocaine can be obtained. For example, conditioning factors have been demonstrated to play an important role in sensitization to cocaine's motor activating effects (5,13). An in depth examination of these factors was beyond the scope of the present study but should be investigated in subsequent experiments. Also of great interest will be to determine other, more benign forms of the manipulations which may be operating in the "normal" animal to predispose it to drug abuse.

ACKNOWLEDGEMENTS

The authors gratefully acknowledge the technical help of Steven Snow, Rachel Peltier, Laura Hickman and Craig Mortensen. This project was supported by a grant from the College of Liberal Arts, TAMU and NIDA 05548, to Susan Schenk. Portions of this work were presented at the Society for Neuroscience meeting, Phoenix, 1989.

## REFERENCES

1. Bozarth, M. A.; Murray, A.; Wise, R. A. Influence of housing conditions on the acquisition of intravenous heroin and cocaine self-administration in rats. Pharmacol. Biochem. Behav. 33:903–907; 1989.
2. Brown, Z. W.; Amit, Z.; Weeks, J. R. Simple flow through swivel for infusions into unrestrained animals. Pharmacol. Biochem. Behav. 5:363–365; 1976.
3. Emmett-Oglesby, M. W.; Spencer, D. G., Jr.; Arnoult, D. E. A TRS-80-based system for the control of behavioral experiments. Pharmacol. Biochem. Behav. 17:583–587; 1982.
4. Henry, D. J.; Greene, M. A.; White, F. J. Electrophysiological effects of cocaine in the mesoaccumbens dopamine system: Repeated administration. J. Pharmacol. Exp. Ther. 251:833–839; 1989.
5. Hinson, R. E.; Poulos, C. X. Sensitization to the behavioral effects of cocaine: Modification by pavlovian conditioning. Pharmacol. Biochem. Behav. 15:559–562; 1981.
6. Kalivas, P.; Duffy, P. Microdialysis studies on terminal field and somatodendritic dopamine release by acute and chronic cocaine. Current Separat. 9:82; 1989.
7. Lett, B. T. Repeated exposures intensify rather than diminish the rewarding effects of amphetamine, morphine, and cocaine. Psychopharmacology (Berlin) 98:357–362; 1989.
8. Lin-Chu, G.; Robinson, T. E.; Becker, J. B. Sensitization of rotational behavior produced by a single exposure to cocaine. Pharmacol. Biochem. Behav. 22:901–903; 1985.
9. Piazza, P. V.; Deminiere, J. Le Moal, M.; Simon, H.; Factors that predict individual vulnerability to amphetamine self-administration. Science 245:1511–1513; 1989.
10. Pickens, R.; Thompson, T. Cocaine-reinforced behavior in rats: Effects of reinforcement magnitude and fixed-ratio size. J. Pharmacol. Exp. Ther. 161:122–129; 1968.
11. Porrino, L. J.; Ritz, M. C.; Goodman, N. L.; Goodman, L. G.; Sharpe, M. J.; Goldberg, S. R. Differential effects of the pharmacological manipulation of serotonin systems on cocaine and amphetamine self-administration in rats. Life Sci. 45:1529–1535; 1989.
12. Post, R. M.; Rose, H. Increasing effects of repetitive cocaine administration in the rat. Nature 260:731–732; 1976.

13. Post, R. M.; Lockfeld, A.; Squillace, K. M.; Contel, N. R. Drug-environment interaction: Context dependency of cocaine-induced behavioral sensitization. Life Sci. 28:755–760; 1981.
14. Robinson, T. E.; Camp, D. M. The acute and chronic effects of amphetamine use on behavior and extracellular dopamine concentrations in the nucleus accumbens and dorsolateral striatum of freely moving rats. Current Separat. 9:84; 1989.
15. Schenk, S.; Lacelle, G.; Gorman, K.; Amit, Z. Cocaine self-administration in rats influenced by environmental conditions: Implications for the etiology of drug abuse. Neurosci. Lett. 81:227–231; 1987.
16. Spencer, D. G., Jr.; Emmett-Oglesby, M. W. Parallel processing strategies in the application of microcomputers to the behavioral laboratory. Behav. Res. Methods Instrum. 17:294–300; 1985.
17. Tatum, A. L.; Seevers, M. H. Experimental cocaine addiction. J. Pharmacol. Exp. Ther. 26:401–410; 1929.
18. Weeks, J. R. Experimental morphine addiction: Method for automatic intravenous injections in unrestrained rats. Science 138:143–144; 1962.
19. Wellman, P. J.; Shelton, K.; Schenk, S. Self-administration of phenylpropanolamine (PPA) by rats previously trained to self-administer amphetamine. Pharmacol. Biochem. Behav. 34:187–191; 1989.
20. Wise, R. A. Neural mechanisms of the reinforcing action of cocaine. NIDA Res. Monogr. 50:15–33; 1984.