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Intermittent and Continuous Cocaine Administration: Residual Behavioral States During Withdrawal

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KING, G. R., C. J. JOYNER, T. H. LEE, C. KUHN AND E. H. ELLINWOOD, JR. *Intermittent and continuous cocaine administration: Residual behavioral states during withdrawal.* PHARMACOL BIOCHEM BEHAV **43**(1) 243-248, 1992. – Rats were pretreated with 40 mg/kg/day cocaine for 14 days by either subcutaneous injections or osmotic minipumps. Rats were then withdrawn from the pretreatment regime for 1 or 7 days and given a 20-mg/kg IP cocaine challenge (day 1) or a 0-, 10-, 20-, or 40-mg/kg IP cocaine challenge (day 7). The results indicate that rats receiving intermittent, daily injections exhibited sensitization to the behavioral effects of a cocaine challenge on days 1 and 7 of withdrawal. In contrast, rats receiving continuous cocaine exhibited tolerance to the behavioral effects of a cocaine challenge on days 1 and 7 of withdrawal. The present results support and extend previous research that indicates that the route and temporal pattern of administration influences the effects of chronic cocaine. Furthermore, the present results indicate that the continuous infusion paradigm may represent an alternative animal model of some aspects of high-dose cocaine abuse, as compared to the typical procedure of single, or multiple, daily cocaine injections.

Cocaine Behavior Withdrawal Tolerance

RESEARCH on chronic cocaine administration indicates that both the dose and route of administration influence the effects of chronic cocaine [see (7) for a review]. For example, daily intermittent IP injections of cocaine produce sensitization (i.e., reverse tolerance) to the locomotor and stereotypy-inducing properties of cocaine (8,13,16). Schedule-induced cocaine intake has also been found to produce sensitization; however, the oral administration of a single daily dose of cocaine does not produce sensitization (3,8). Lastly, subcutaneous injections also produce sensitization to the locomotor effects of cocaine, while continuous infusion of cocaine via minipump produces tolerance (14). These results indicate that the behavioral effects (sensitization or tolerance) of chronic cocaine are partially dependent upon the method of administration and may be related to distinct residual behavioral states.

Chronic stimulant administration also results in an altered neurobiological/behavioral state that not only produces an acute withdrawal syndrome (4) but also results in a protracted residual state that may be the substratum for the high rate of recidivism of cocaine abusers. For example, Reith et al. (15) examined the hyperactivity (i.e., general activity) of mice receiving cocaine via SC injections or a subcutaneous minipump for 18 days. During the first 11 days of cocaine infusion, the continuous infusion mice exhibited marked daytime hyperactivity; however, daytime activity returned to basal levels during the remaining 8 days. These same animals also exhibited residual tolerance to a cocaine challenge administered approximately 7 days after cessation of cocaine infusions. In contrast to subjects receiving continuous cocaine, subjects receiving daily SC injections of cocaine exhibited sensitization to a cocaine challenge approximately 7 days after cessation of treatment.

Although the Reith et al. (15) data suggest that the continuous infusion subjects became tolerant to the stimulant effects of a cocaine challenge, this conclusion remains tentative. First, the experiments only examined general activity. Indices of changes in general activity only measure some of the effects of cocaine administration. Measures of changes in the structure of behavior are also necessary, and this is especially true given the stereotypy-inducing properties of cocaine (16). Second, that study only measured the behavioral effects of cocaine for a 10-min period. Therefore, it is not clear if subjects were only tolerant during the onset phase of cocaine distribution but would have exhibited the same maximal activity levels as control subjects if a longer sampling period had been used. Lastly, only a single cocaine challenge dose was used; thus, any conclusions regarding tolerance must remain tentative in the absence of demonstrated changes in behavior over a dose-response analysis. Therefore, it is of interest to determine if

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tolerance to the behavioral effects of cocaine can be demonstrated over a dose-response analysis and over a longer behavioral sampling period.

The present experiments examine the residual behavioral states produced by withdrawal from different routes of cocaine administration (i.e., continuous or intermittent cocaine). These residual states were assessed by behavior ratings over several challenge doses of cocaine. The dose-response curves were determined on the first or seventh day of withdrawal from intermittent or continuous cocaine administration to examine the duration (robustness) of these residual states.

METHOD

Animals

Male Sprague-Dawley rats, initially weighing 100–125 g (Charles River Laboratories), were acclimated to the vivarium on a 12 L : 12 D cycle (light between 7 a.m.–7 p.m.) for 1 week prior to treatment. They were housed in pairs in plastic cages with continuous access to food and water.

Drug

Cocaine HCl (received from NIDA) was dissolved in 0.9% sterile saline. All doses are calculated as the salt, and injection volume was based upon body weight.

Minipump Preparation

Alzet Osmotic pumps (model 2ML2) from Alza Corporation (Palo Alto, CA) were filled with either 2 ml 100 mg/ml cocaine HCL or saline (0.9%); the infusion rate was 2μl/h, resulting in an overall, average dose of 40 mg/kg/day for the cocaine pumps. The pump was primed by warming in a beaker of saline in a waterbath at 37°C for 4 h prior to surgical implantation.

Surgery

Animals were shaved and injected locally with (0.2 cc) lidocaine (Abbott Corp., North Chicago, IL) at the dorsal midline incision site. Animals were then anesthetized by inhalation with methoxyflurane (Metofane). A 2-cm vertical incision was made with scissors and a large subcutaneous pocket was formed with the scissors. The minipump was inserted into this pocket with the delivery portal toward the head. The opening was closed with metal surgical autoclips.

On day 14, pumps were surgically removed using the same procedure and the residual amount of cocaine measured. The amount was consistently less than 15% of the original volume, indicating that rats approximately received the programmed daily dose.

Pretreatment

Treatment was for a 14-day period. On day 1 of treatment, animals were either: a) implanted with 2ML2 Alzet minipumps continuously infusing cocaine at a rate of 40 mg/kg/day (pump groups), b) injected SC once daily with 40 mg/kg cocaine HCl (injection group), or c) either injected SC with 0.9% saline once daily or implanted with a saline minipump as the controls. Treatment with a saline minipump controlled for any possible confounding effects of the implantation (surgical) procedure on rats' behavior.

Behavioral Testing

On either day 1 or 7 following pretreatment, animals were acclimated to the test room in their home cage for 30 min under normal light conditions and then placed in the test cage for a further 20-min adaptation period. The test cages were standard, clear plastic laboratory animal housing cages, 28 × 18 × 12 cm, with a second cage taped, upside down, in place on top. The top cage had five air holes drilled uniformly on either side. Four of these test cages were placed in a row 12 in. apart. Following adaptation to the test cages, the rating procedure began. The Ellinwood and Balster Rating Scale (2) was used (Table 1). A rating was given to each of the animals at 5 min preinjection and at 5-min intervals thereafter for a total of 90 min. The observation period was for 20 s with 10 s between cages. The observer was blind to the treatment and test dose each animal received.

For the test session on day 1 following pretreatment, all rats received an IP injection of 20 mg/kg cocaine. For the test session on day 7 following pretreatment, each rat received one

TABLE 1

ELLINWOOD AND BALSTER (2) RATING SCALE

Score		Definition
1	Asleep	Lying down; eyes closed
2	Inactive	Lying down; eyes open
3	Inplace activities (grooming)	Normal grooming or chewing cage litter
4	Normal; alert; active	Moving about cage, sniffing; rearing
5	Hyperactive	Running movement characterized by rapid changes in position (jerky)
6	Slow patterned	Repetitive exploration of the cage at normal level of activity
7	Fast patterned	Repetitive exploration of the cage with hyperactivity
8	Dyskinetic reactive	Backing up; jumping; seizures; abnormally maintained postures; dyskinetic movements

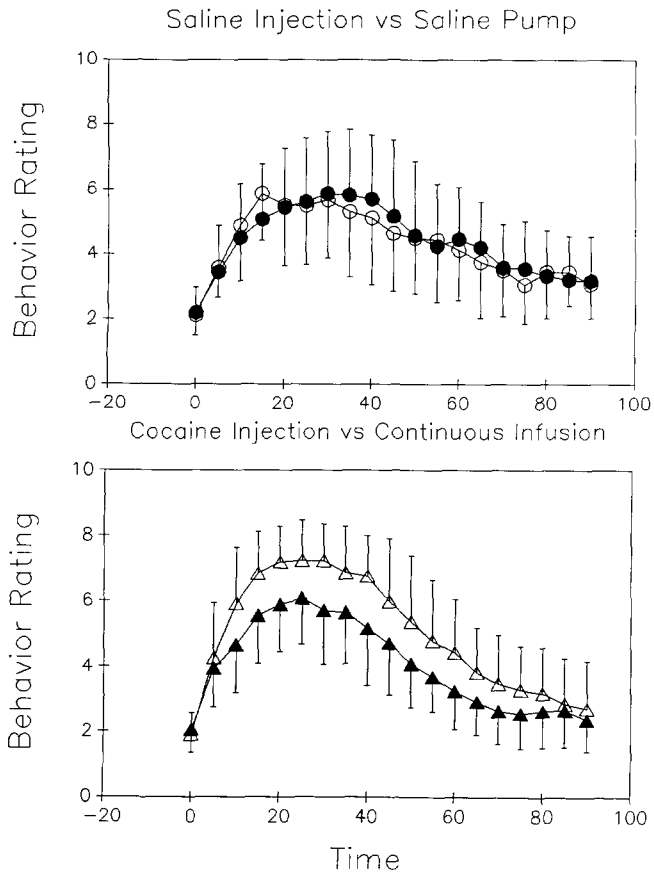

FIG. 1. Mean behavior rating for each group following a 20-mg/kg cocaine challenge 24 h after cessation of treatment. The bars represent one SD for each point. (○), saline pump pretreatment rats; (●), saline injection pretreatment rats; (△), cocaine injection pretreatment rats; (▲), continuous infusion pretreatment rats.

of the following doses of cocaine IP: 0, 10, 20, or 40 mg/kg. For each test session, the subject types (i.e., injection, pump, saline) were randomized according to a Latin square design; doses for each test session were also randomized by a Latin square design. The significance level was set at $p < 0.05$ for all comparisons.

RESULTS

Day 1

Figure 1 presents the mean behavior rating for each treatment group following a 20-mg/kg cocaine challenge 24 h after cessation of treatment. To examine whether the implantation procedure affected subjects' behavior, Mann-Whitney *U*-tests were performed comparing the behavior ratings of saline injection ($n = 16$) and saline pump ($n \approx 24$) subjects separately at each time point. The results of these comparisons indicated no significant differences between the two groups at any time point. Therefore, these subjects were collapsed into a single control group for all subsequent comparisons.

The results of Mann-Whitney comparisons indicate that the cocaine injection group ($n = 40$) exhibits significantly higher behavior ratings than the saline control group at 5-45 min and significantly higher behavior ratings than the cocaine pump group ($n = 38$) at 10-80 min. Furthermore, the cocaine pump group exhibits significantly lower behavior ratings than the saline control group at 60-90 min.

Day 7

Figure 2 presents the mean behavior ratings for each dose separately for each group on the seventh day of withdrawal from the chronic dosing regime. This figure indicates that each group demonstrated a dose response to cocaine challenges. In other words, increasing doses of cocaine resulted in increasing behavior ratings.

Figure 3 presents the mean behavior rating for each treatment group, separately for each dose, 7 days after cessation of treatment. Panel A presents the behavior ratings of subjects receiving a vehicle injection. Mann-Whitney comparisons indicate that the cocaine injection group ($n = 12$) exhibits a significantly higher behavior rating compared to the saline control group ($n = 12$) at 10 min and significantly higher behavior ratings than the cocaine pump group ($n = 10$) at 10 and 15 min.

Panel B presents the behavior ratings for subjects receiving a 10-mg/kg cocaine challenge dose. The results of Mann-Whitney comparisons indicate that the cocaine injection group

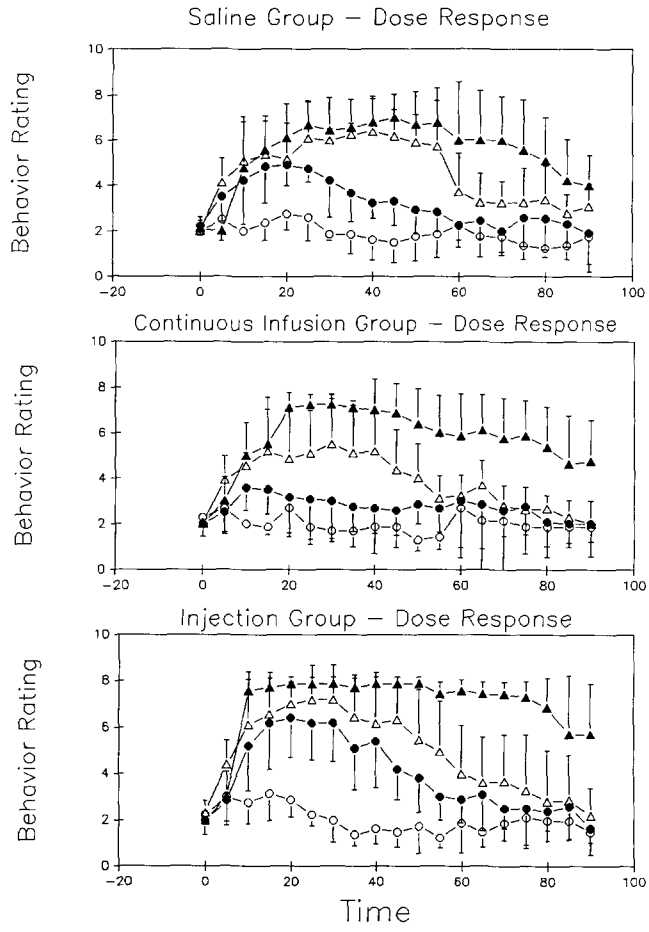

FIG. 2. Mean behavior ratings for each dose separately for each group on the seventh day of withdrawal from the chronic dosing regime. (○), vehicle challenge dose; (●), 10-mg/kg challenge dose; (△), 20-mg/kg challenge dose; (▲), 40-mg/kg challenge dose.

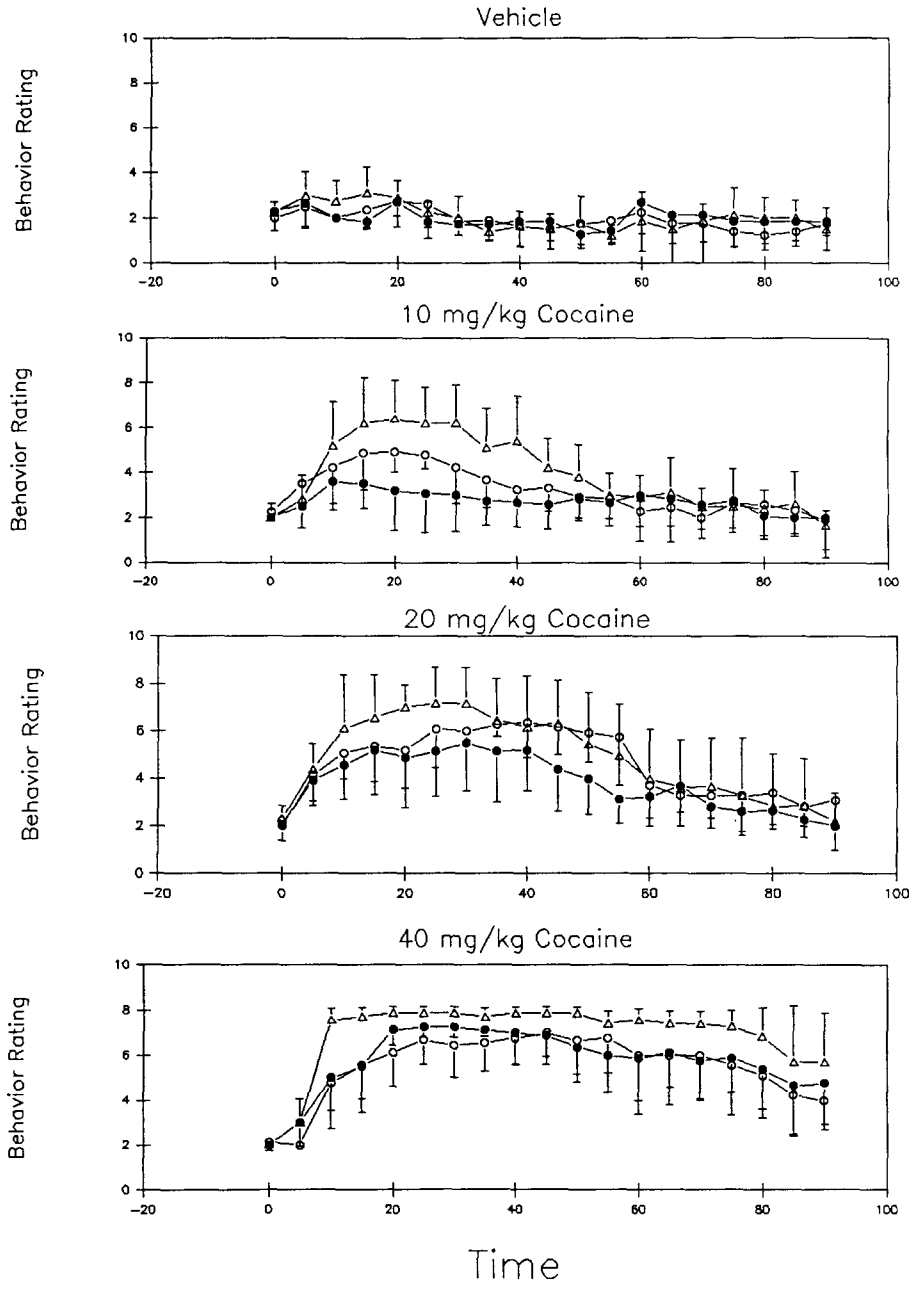

FIG. 3. Mean behavior rating for each treatment group, separately for each dose, 7 days after cessation of treatment. (●), continuous infusion pretreatment rats; (○), saline control rats; (△), cocaine injection pretreatment rats.

(n = 11) exhibits significantly higher behavior ratings than the saline control group (n = 11) at 20–30 and 40 min and significantly higher behavior ratings than the cocaine pump (n = 12) group at 15–45 min. Furthermore, the cocaine pump group exhibits significantly lower behavior ratings than the saline control group at 20 and 25 min.

Panel C presents the behavior ratings for subjects receiving a 20-mg/kg cocaine challenge dose. The results of Mann-Whitney comparisons indicate that the cocaine injection group (n = 13) exhibits significantly higher behavior ratings than the saline control group (n = 11) at 15–30 min and significantly higher behavior ratings than the cocaine pump group (n = 12) at 10–55 min compared to the cocaine pump group. Furthermore, the cocaine pump group exhibits significantly lower behavior ratings than the saline control group at 40–55 min.

Panel D presents the behavior ratings for subjects receiving a 40-mg/kg cocaine challenge dose. The results of Mann-Whitney comparisons indicate that the cocaine injection group (n = 11) exhibits significantly higher behavior ratings than

the saline control group (n = 13) at 10–30 min and significantly higher behavior ratings than the cocaine pump group (n = 12) at 10–35, 45–50, 60, and 70 min compared to the cocaine pump group. The cocaine pump and saline control groups are not significantly different at any time point.

DISCUSSION

The present results support, and extend, previous findings that indicate that the effects of chronic cocaine depend upon route of administration. Chronic, daily SC injections of cocaine produce sensitization to subsequent cocaine challenges 24 h and 7 days following pretreatment with chronic cocaine; this is consistent with previous research using daily, intermittent injections of cocaine (8,13,16). In contrast to these results, continuous infusion of an overall, equivalent daily dose of cocaine produces tolerance to subsequent cocaine challenges 24 h and 7 days following pretreatment with chronic cocaine; these results are consistent with the study by Reith et al. (15), but extend those results to a dose-response analysis and a longer sampling period.

The sensitization exhibited by the injection group tends to be characterized by a rapid onset and, with the exception of the 40-mg/kg challenge dose, is generally restricted to the initial 30–35 min of the observation session. In other words, the sensitization manifests itself within the first 10–15 min of the session and lasts approximately 30 min. In contrast, the tolerance exhibited by the continuous infusion subjects generally manifests itself in the latter portions of the session (i.e., the offset phase); the tolerance is characterized, generally, as a more rapid offset of cocaine's effects, relative to control subjects.

These results are probably not solely due to changes in the pharmacokinetics of cocaine. First, no consistent differences have been found in the early dispositional phase characteristics of cocaine following chronic treatment. For example, no differences in cocaine levels in brain, cerebrospinal fluid, and tissue in acutely and chronically treated subjects have been reported (9,10,12). Furthermore, consistent changes in cocaine half-life have not been reported: The half-life of cocaine following chronic treatment has been reported to be both increased (6) and decreased (10). Second, Reith et al. (15) did not report any differences in cerebral cocaine or benzoylecgonine between subjects receiving SC injections or continuous infusion for 18 days. Finally, although Pettit et al. (11), utilizing dialysis, found an increase in cocaine levels in chronically treated animals, the dopamine : cocaine ratio was even larger. Thus, changes in cocaine disposition alone are probably not sufficient to explain the present results.

Injection subjects receiving vehicle injections exhibited a significant increase in behavior ratings during the initial 10 min of the observation session. This short-term, initial increase probably reflects a conditioned increase in behavior (1,5,14). These subjects had received 14 injections of cocaine. Some aspects of the locomotor-stimulating properties of cocaine could have become conditioned to the injection process. Thus, the increase in the behavior in injection subjects during the initial 10 min of the session would reflect this short-term conditioned increase. The subsequent, rapid return to baseline activity levels reflects the fact that subjects did not receive the drug and therefore did not experience the locomotor-stimulating properties of the drug.

The failure to find tolerance in the continuous infusion subjects at the highest dose tested is not surprising. The 40-mg/kg challenge was an extremely large dose that could simply overwhelm the residual neuroadaptations. This result indicates that subtle residual effects of chronic cocaine administration may be masked by subsequent large drug challenges and that dose–response analyses should be used when examining the residual effects of chronic cocaine administration.

The present results also indicate that the continuous infusion paradigm may represent an alternative animal model of some aspects of high-dose cocaine abuse than the typical procedure of single, or multiple, daily injections of cocaine. First, cocaine abuse is sometimes characterized by a "binge" pattern of drug consumption. A binge is characterized by the readministration of the drug approximately every half hour or less. In recreational users, a binge typically lasts 12 h, whereas in high-dose users it not infrequently lasts days (4). Thus, during a binge, although the plasma cocaine levels will fluctuate as a function of an oscillating pattern of self-administration, the abuser nonetheless is maintaining a reasonably sustained plasma cocaine level, not infrequently over prolonged periods. This aspect of cocaine abuse is not represented by intermittent cocaine injections but is represented by the continuous infusion paradigm. Second, binges are thought to occur because of acute tolerance; the effects of the drug may rapidly diminish with each successive administration. This acute tolerance, coupled with the memory of the immediately preceding "high," produces the desire to reinstate the drug effect; this is accomplished by the repeated consumption of the drug (4). Daily intermittent injections produce sensitization, and not tolerance, to the effects of cocaine. However, the continuous infusion paradigm does result in tolerance to the stimulant effects of cocaine. Thus, the continuous infusion paradigm may represent a good alternative animal model of high-dose cocaine addiction (4).

In summary, the present results indicate that daily, intermittent SC injections produce sensitization to subsequent challenge doses of cocaine administered either 1 or 7 days after cessation of chronic treatment. In contrast to these results, continuous infusion of equivalent daily doses of cocaine results in tolerance to subsequent challenge doses of cocaine. Thus, the continuous infusion paradigm may represent a more valid animal model of some aspects of human cocaine abuse.

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