Cocaine Is Self-Administered into the Shell but Not the Core of the Nucleus Accumbens of Wistar Rats

ZACHARY A. RODD-HENRICKS, DAVID L. MCKINZIE, TING-KAI LI, JAMES M. MURPHY, and WILLIAM J. MCBRIDE

Institute of Psychiatric Research and Department of Psychiatry (Z.A.R.-H., D.L.M., J.M.M., W.J.M.), and Departments of Medicine (T.-K.L.) and Biochemistry (W.J.M.), Indiana University School of Medicine, Indianapolis, Indiana; and Department of Psychology (J.M.M.), Purdue School of Science, Indiana University-Purdue University at Indianapolis, Indianapolis, Indiana

Received May 15, 2002; accepted July 16, 2002

ABSTRACT

The rewarding properties of cocaine have been postulated to be regulated, in part, by the mesolimbic dopamine system. However, the possibility that the rewarding properties of cocaine are mediated by direct activation of this system has yielded contradictory findings. The intracranial self-administration technique is used to identify specific brain regions involved in the initiation of response-contingent behaviors for the delivery of a reinforcer. The present study assessed whether adult Wistar rats would self-administer cocaine directly into the nucleus accumbens shell (AcbSh) and core (AcbC). For each subregion, subjects were placed in standard two-lever operant chambers and randomly assigned to one of five groups for each site that were given either artificial cerebrospinal fluid (aCSF), or 400, 800, 1200, or 1600 pmol of cocaine/100 nl to self-administer. The data indicate that rats with placements within the AcbSh readily self-administered 800 to 1600 pmol of cocaine/100 nl and responded significantly more on the active than inactive lever. These subjects also decreased responding on the active lever when aCSF was substituted for cocaine and reinstated responding on the active lever when cocaine was reintroduced. Coinfusion of the D2-like receptor antagonist sulpiride inhibited cocaine self-infusion in the AcbSh. In contrast to the AcbSh data, rats failed to self-administer any tested dose of cocaine into the AcbC or areas ventral to the AcbSh. These findings suggest that the AcbSh is a neuroanatomical substrate for the reinforcing effects of cocaine and that activation of D2-like receptors is involved.
wyler, 1994) and DA levels in the Acb (Wise et al., 1995; Hemby et al., 1997). Additionally, the initiation of cocaine self-infusions is predicated upon the extracellular DA level in the Acb (Wise et al., 1995; Peoples and West, 1996). Moreover, neural firing of Acb cells may reflect changes in cocaine levels and contribute to the temporal spacing of cocaine self-administration (Nicola and Deadwyler, 2000). Furthermore, research has indicated that cells within the Acb display distinct, independent firing patterns in response to i.v. cocaine administration compared with water and food reinforcements (Carelli et al., 2000).

In the initial cocaine ICSA study (Goeders and Smith, 1983), placements seemed to be mainly within the AcbC. However, since the publication of the Goeders and Smith (1983) study, several reports have demonstrated functional differences between the two subregions of the Acb with respect to DA neurotransmission, psychostimulant-induced DA transmission, and behavioral effects of local application of psychostimulants. For example, cocaine-induced locomotor activity is reduced by microinjections of NMDA antagonists directly into the AcbC, but not into the AcbSh (Pulvirenti et al., 1994), whereas i.v. administration of cocaine, morphine, and amphetamine preferentially increase extracellular DA in the AcbSh compared with the AcbC (Pontieri et al., 1995). Similarly, i.p.-administered cocaine significantly increased DA levels in the AcbSh, whereas only a slight increase was observed in the AcbC (Hedou et al., 1999). Additionally, cocaine-induced increases of extracellular DA levels within the AcbSh were associated with an increase in locomotor activity (Hedou et al., 1999), and the AcbSh is also more responsive to systemically administered D1 dopamine receptor agonists and antagonists than the AcbC when observing acetylcholine output (Consolo et al., 1999). Last, evidence for an important difference between the accumbens subregions in the ability to support self-administration behavior emerges from a study reporting that the ICSA of nomifensine, a DA uptake inhibitor, occurred in the AcbSh, but not in the AcbC (Carlezon et al., 1995).

The present study was undertaken to examine the ICSA of cocaine into the AcbSh and AcbC and to determine the effects of a D2/D3 receptor antagonist on this ICSA behavior. The hypothesis to be tested is that the AcbSh is a site supporting the reinforcing effects of cocaine and these effects are mediated in part by D2/D3 receptors.

Materials and Methods

Animals

Experimentally naive female Wistar rats (Harlan, Indianapolis, IN), weighing 250 to 320 g at time of surgery, were used. Rats were double-housed upon arrival and maintained on a 12-h reverse light/dark cycle (lights off at 9:00 AM). Female rats were used because they maintain their body size better than males, which allows for more accurate cannula placements. Although not systematically studied, the estrus cycle did not seem to have a significant effect on ICSA behavior in the present study or in previous studies (Gatto et al., 1994; Ikemoto et al., 1997; Rodd-Henricks et al., 2000, 2002), as indicated by no obvious fluctuations in ICSA behavior in rats given similar doses of the same agent for two or more sessions conducted every other day. Food and water were freely available except in the test chamber. Protocols were approved by the institutional animal care and use committee and are in accordance with the guidelines of the Institutional Care and Use Committee of the National Institute on Drug Abuse, National Institutes of Health, and the Guide for the Care and Use of Laboratory Animals (National Research Council, 1996).

Data for rats that did not complete all experimental test sessions were eliminated from the analyses. The number of animals indicated for each experiment represents approximately 95% of the total number that underwent surgery; about 5% of the animals were not included for analyses mainly due to the loss of the guide cannula before completion of all experimental sessions. The data for these animals were not used because their injection sites could not be verified due to the loss of the guide cannula.

Drug and Vehicle

The artificial cerebrospinal fluid (aCSF) consisted of 120.0 mM NaCl, 4.8 mM KCl, 1.2 mM KH2PO4, 1.2 mM MgSO4, 25.0 mM NaHCO3, 2.5 mM CaCl2, and 10.0 mM d-glucose. Cocaine hydrochloride (National Institute on Drug Abuse) and the D2/D3 DA receptor antagonist sulpiride (Sigma-Aldrich, St. Louis, MO) were dissolved in the aCSF solution. When necessary, 0.1 M HCl or 0.1 M NaOH was added to the solutions to adjust pH levels to 7.4 ± 0.1.

Apparatus

The test chambers (30 x 30 x 26 cm; width x height x depth) were situated in a sound-attenuating cubicule (64 x 60 x 50 cm; Coulbourn Instruments, Allentown, PA) and illuminated by a dim house light during testing. Two identical levers (3.5 x 1.8 cm) were mounted on the same wall of the test chamber, 15 cm above a grid floor, and separated by 12 cm. Levers were raised to this level to avoid inadvertent bar pressing and to reduce responses as a result of general locomotor activation. Directly above each lever was a row of three different-colored cue lights. The light (red) to the far right over the active bar was illuminated during resting conditions and was extinguished when the active lever was pressed. A desktop computer equipped with an operant control system (L2T2; Coulbourn Instruments) recorded the data and controlled the delivery of infusate in relation to lever response.

An electrolytic microinfusion transducer (EMIT) system (Bozarth and Wise, 1980) was used to control microinfusions of drug or vehicle. Briefly, two platinum electrodes were placed in an infusate-filled cylinder container (28 mm in length x 6 mm in diameter) equipped with a 28-gauge injection cannula (Plastics One, Roanoke, VA). The electrodes were connected by a spring-coated cable (Plastics One) and a swivel (model 2005; Mercotac, Carlsbad, CA) to a constant current generator (MNC, Shreveport, LA), which delivered 6 μA of quiescent current and 200 μA of infusion current between the electrodes. Depression of the active lever delivered the infusion current for 5 s, which led to the rapid generation of H2 gas (increasing the pressure inside the airtight cylinder), and, in turn, forcing 100 nl of the infusate through the injection cannula. EMIT units were calibrated weekly by sampling after a 60- and 5-s infusion test. During, the 5-s infusion and additional 5-s time-out period, the house light and right cue light (red) were extinguished and the left cue light (green) over the active lever flashed on and off at 0.5-s intervals. A 55-s time-out period was used in one experiment to provide evidence that responding on the active lever was not solely a result of reflexive responding (Goeders and Smith, 1987). The conditions for the 55-s time-out were the same as the 5-s time-out period, except for the duration.

Animal Preparation

While under halothane anesthesia, a unilateral 22-gauge guide cannula (Plastics One) was stereotaxically implanted in the right hemisphere of each subject, aimed 1.0 mm above the target region. Coordinates (Paxinos and Watson, 1986) for placements into the AcbC were 1.7 mm anterior to bregma, 2.4 mm lateral to the midline, and 7.5 mm ventral from the surface of the skull at a 10° angle to the vertical. Coordinates for placements into the AcbSh were 1.7 mm anterior to bregma, 2.4 mm lateral to the midline, and 7.5 mm ventral from the surface of the skull at a 10° angle to the vertical.
1.7 mm anterior to bregma, 2.7 mm lateral to the midline, and 6.5 mm ventral from the surface of the skull at a 10° angle to the vertical. Between experimental sessions, a 28-gauge stylet was placed in the guide cannula and extended 0.5 mm beyond the tip of the guide. After surgery, all rats were individually housed and allowed to recover for 7 to 10 days. Animals were handled for at least 5 min daily after the fourth recovery day. Subjects were not acclimated to the test chamber before the commencement of data collection nor were they trained on any other operant paradigm.

**General Test Condition**

For testing, subjects were brought to the testing room, the stylet was removed, and the injection cannula was screwed into place. Rats were then placed in the operant chamber. To avoid trapping air at the tip of the injection cannula, the infusion current was delivered for 5 s during insertion of the injector, which extended 1.0 mm beyond the tip of the guide. Depression of the “active lever” on a fixed ratio 1 schedule of reinforcement caused the delivery of a 100-nl bolus of infusate over a 5-s period followed by a 5-s time-out period. During both the 5-s infusion period and 5-s time-out period, responses on the active lever did not produce further infusions, but were recorded. Responses on the “inactive lever” were recorded, but did not result in infusions. The assignment of active and inactive lever with respect to the left or right position was counterbalanced among subjects. However, the active and inactive levers remained the same for each rat throughout the experiment. No shaping technique was used to facilitate the acquisition of lever responses. The number of infusions and responses on the active and inactive bar were recorded throughout each session. The duration of each operant session was 4 h and sessions occurred every 48 h.

**Treatment Procedures**

**Dose Response.** Animals were randomly assigned to one of five groups per injection site (shell, n = 10–12/group; core, n = 6–7/group). A vehicle group received infusions of 100 nl of aCSF for all seven sessions. The other groups received infusions of 400, 800, 1200, or 1600 pmol of cocaine/100 nl (4, 8, 12, or 16 mM cocaine) for the first four sessions, followed by infusions of 100 nl of aCSF during the fifth and sixth sessions; in the seventh session, rats were allowed to respond for their originally assigned infusate. A total of 110 rats completed the training procedure. Thirteen rats from various groups (aCSF, n = 2; for cocaine: 400 pmol, n = 3; 800 pmol, n = 3; 1200 pmol, n = 2; and 1600 pmol, n = 2) had placements ventral to the AcbSh. For the rats with placements ventral to the AcbSh, data from the 400 to 1600 pmol of cocaine/100 nl of aCSF groups were collapsed across infuscate groups and served as neuroanatomical controls.

**Sulpiride Coadministration.** A separate group of rats with guide cannulae aimed at the AcbSh, self-administered 1200 pmol of cocaine/100 nl for the initial four sessions (n = 9). During the fifth and sixth sessions, 400 pmol of sulpiride/100 nl was added to the cocaine solution, and the rats were allowed to self-administer the combined solution. On the seventh and eighth sessions, rats were given only the 1200 pmol of cocaine/100 nl. Another group of rats (n = 8) was given 400 pmol/100 nl of sulpiride alone for the entire eight sessions.

**Extended Time-Out.** An additional control experiment was conducted to determine whether extending the time-out period between infusions would alter self-administration of cocaine into the AcbSh. Reflexive responding, particularly with psychostimulants, may underlie the self-infusion of cocaine into the AcbSh (Goeders and Smith, 1987). Therefore, a recommended control experiment is to extend the interinfusion interval by increasing the time-out period (Goeders and Smith, 1987). Thus, during this experiment, depression of the active lever (fixed ratio 1 schedule of reinforcement) caused the delivery of a 100-nl bolus of infusate over a 5-s period followed by a 55-s time-out period. During both the 5-s infusion period and 55-s time-out period, responses on the active lever were recorded but did not produce further infusions. Responses on the inactive lever were recorded but did not result in infusions. Under these conditions, a group of rats (n = 8) received infusions of 1200 pmol of cocaine/100 nl of aCSF for 11 consecutive sessions.

**Histology.** At the termination of the experiment, 1% bromophenol blue (0.5 µl) was injected into the infusion site. Subsequently, the animals were given a fatal dose of Nembutal and then decapitated. Brains were removed and immediately frozen at −70° C. Frozen brains were equilibrated at −15°C in a cryostat microtome and then sliced into 40-µm sections. Sections were then stained with cresyl violet and examined under a light microscope for verification of the injector site using the rat brain atlas of Paxinos and Watson (1986).

**Results**

**Placements.** Cannula placements in and around the AcbC and AcbSh are depicted in Fig. 1. Cannula placements outside the Acb were ventral to the AcbSh. Primary locations of the cannula tips were between +1.0 and +1.6 mm relative to bregma. Additionally, approximately 95% of all subjects successfully completed the experimental paradigms and had cannula tip locations within the AcbC, AcbSh, or area immediately ventral to the AcbSh. Photomicrographic images represent the cannula tract and angle of descent for placements in the AcbC, AcbSh, and ventral to the AcbSh (Fig. 1, left). The right side of Fig. 1 shows the distribution of placements.

**Dose Response.** A select range of cocaine concentrations supported response-contingent behaviors in the AcbSh. For rats receiving infusions into the AcbSh, an ANOVA on the average number of infusions (Fig. 2) received during the initial four test days (acquisition) revealed a significant effect of dose (F(4,48) = 3.53; p = 0.013). Post hoc comparisons (Tukey’s b; p < 0.05) indicated that the 400-, 800-, 1200-, and 1600-pmol groups received significantly more infusions than the aCSF controls. Additionally, the post hoc comparisons revealed that the 1200-pmol group received significantly more infusions than the 400-pmol group. In contrast, an ANOVA performed on the average number of infusions received during acquisition by rats self-administering into the AcbC (Fig. 2) revealed no significant effect of dose (F(4,49) = 1.65; p = 0.19). Although not included in any of the analyses, rats with cannulae implanted ventral to the AcbSh (Fig. 2) received a comparable amount of infusions as those administering aCSF into either the AcbSh or AcbC.

**Extinction and Reinstatement.** For subjects receiving infusions into the AcbSh, a repeated measures ANOVA performed on the number of infusions across the seven sessions revealed that there was a significant effect of dose (F(4,48) = 4.5; p = 0.004), an effect of session (F(6,288) = 6.3; p < 0.001), and a session × dose interaction (F(24,288) = 3.9; p < 0.001). Decomposing the interaction term by holding sessions constant resulted in individual ANOVAs for each acquisition session being performed. The analysis revealed a significant effect of dose for each session in which cocaine was self-administered (sessions 1–4 and 7; F(4,48) values > 2.65; p values < 0.045). Post hoc comparisons revealed that during session 4, all four cocaine groups received significantly more infusions than the aCSF group, and the 800- and 1200-pmol cocaine groups received significantly more infusions than the 400-pmol group (Fig. 3). In contrast, when aCSF was substituted for cocaine in sessions 5 and 6, there were no significant differences between the dose groups on the amount of infusions administered (F(4,48) values < 2.19; p values > 0.09).
When cocaine was restored in session 7, the 400-, 800-, and 1200-pmol cocaine groups showed an increase (Fig. 3) in the number of infusions during session 7 compared with session 6 ($F_{1,9}$ values = 11.51; $p$ values = 0.008), whereas the 1600-pmol cocaine group did not. Additionally, an ANOVA performed on the number of infusions received during session 7 revealed a significant effect of dose ($F_{4,48}$ = 2.92; $p$ = 0.03). Post hoc comparisons indicated that all cocaine infusate groups were significantly higher than the aCSF group and that the 1200-pmol group received significantly more infusions than the 1600-pmol group (Tukey’s $b$; $p$ < 0.05). Of particular interest, the 400-pmol group displayed a more than 3-fold increase ($F_{3,27}$ = 7.93; $p$ < 0.001) in the number of infusions received during session 7 (43 ± 6) than during session 4 (14 ± 3).

**Patterns of Infusions.** The temporal patterns of infusions (in 30-min blocks) by rats given aCSF, 400 or 1200 pmol of cocaine into the AcbSh or 1200 pmol of cocaine into the AcbC are depicted in Fig. 4. Throughout all sessions, rats self-administering aCSF into the AcbSh closely resembled the infusion pattern for rats that were self-administering 1200 pmol of cocaine into the AcbC. For the group self-infusing 1200 pmol of cocaine into the AcbSh, approximately 2 h into the first session, this group diverged from the other groups and displayed signs of acquiring self-administration (Fig. 4, top). During session 4, rats readily administered 1200 pmol of cocaine/100 nl within the first 30-min block maintained a higher level of responding throughout the session and displayed an increase in responding during the last 30-min block. During the second aCSF substitution session (session 6), the number of infusions was low and most were administered during the initial two 30-min periods. When 1200 pmol of cocaine was restored in session 7, rats rapidly reinstated self-administration behavior and exhibited an infusion pattern similar to acquisition session 4, except for the high number of infusions in the last 30-min period. With regard to the group that was self-infusing 400 pmol of cocaine/100 nl into the AcbSh, the pattern of self-administration during session 1 and 4 only slightly deviated from that observed with aCSF self-administration with higher infu-
During the fourth session (acquisition), the 400-, 800-, 1200-, and 1600-pmol of cocaine/100 nl directly into the AcbSh readily acquired response-contingent behavior (Fig. 7). During the second through fourth sessions, rats discriminated between the active and inactive lever was reduced in sessions 5 and 6 compared with session 4 ($F_{2,16} = 9.1; p = 0.002$) and lever discrimination was no longer apparent ($F_{1,8} < 1.7; p > 0.23$). As a measure of the overall effects of sulpiride on general locomotor activity, an analysis preformed on the number of inactive lever response during sessions 4 to 6 indicated no alterations in number of responses ($F_{2,16} = 2.6; p = 0.11$). In addition, a general comparison of the number of active lever responses in rats self-administering 1200 pmol of cocaine within the initial 30 min of the first sulpiride coadministration or aCSF substitution session (data not shown) indicated that the effect of sulpiride coadministration was not different from aCSF substitution ($22.2 \pm 8.6$ and $31.6 \pm 10.4$, respectively). Giving 1200 pmol/100 nl of cocaine alone in sessions 7 and 8 resulted in the recovery of responding on the active lever ($F_{2,16} = 9.1; p = 0.002$) and the re-establishment of lever discrimination ($F_{1,8} > 9.3; p < 0.016$). Rats

Effects of Sulpiride on Cocaine Self-Infusions. Replicating the initial finding, rats allowed to self-administer 1200 pmol of cocaine/100 nl directly into the AcbSh readily acquired response-contingent behavior (Fig. 7). During the second through fourth sessions, rats discriminated between the active and inactive lever ($F_{1,8} > 13.0; p < 0.007$). When 400 pmol/100 nl of sulpiride was coadministered with cocaine, responding on the active lever was reduced in sessions 5 and 6 compared with session 4 ($F_{2,16} = 9.1; p = 0.002$) and lever discrimination was no longer apparent ($F_{1,8} < 1.7; p > 0.23$). As a measure of the overall effects of sulpiride on general locomotor activity, an analysis performed on the number of inactive lever response during sessions 4 to 6 indicated no alterations in number of responses ($F_{2,16} = 2.6; p = 0.11$). In addition, a general comparison of the number of active lever responses in rats self-administering 1200 pmol of cocaine within the initial 30 min of the first sulpiride coadministration or aCSF substitution session (data not shown) indicated that the effect of sulpiride coadministration was not different from aCSF substitution ($22.2 \pm 8.6$ and $31.6 \pm 10.4$, respectively). Giving 1200 pmol/100 nl of cocaine alone in sessions 7 and 8 resulted in the recovery of responding on the active lever ($F_{2,16} = 9.1; p = 0.002$) and the re-establishment of lever discrimination ($F_{1,8} > 9.3; p < 0.016$). Rats
self-administering sulpride alone throughout all eight sessions (data not shown) had low levels of responding on both levers (<20 responses/session) and did not discriminate between active and inactive levers during any session ($F_{1,7}$ values < 0.07; $p$ values > 0.79).

**Extended Time-Out.** Extending the time-out period from 5 to 55 s retarded the acquisition of 1200 pmol of cocaine/100-nl self-administration into the AcbSh (Fig. 8), as indicated by the finding that responses on the active lever were not greater than responses on the inactive lever until the fourth session ($F_{1,8}$ values = 15.3; $p$ values < 0.004). There was a significant effect of session on the number of infusions administered ($F_{10,80}$ = 23.8; $p$ < 0.0001) and the number of active lever responses ($F_{10,80}$ = 19.2; $p$ < 0.0001). Post hoc comparisons (Tukey's) revealed that both the number of infusions administered and the number of active lever responses were greater during the last three sessions (sessions 9–11) than after the initial signs of acquisition of self-administration (sessions 4–7) and had reached asymptotic levels.

**Discussion**

The results of the current study indicate that Wistar rats will initiate and maintain self-administration of cocaine directly into the AcbSh, but not the AcbC or areas ventral to the AcbSh (Fig. 2), suggesting that the AcbSh is a site supporting the reinforcing actions of cocaine. The self-infusion of cocaine into the AcbSh does not seem to be a result of a general increase in behavioral activity because rats learned to discriminate the active from the inactive lever during acquisition for the self-infusion of 800 to 1200 pmol of cocaine (Figs. 5 and 6), and self-administration was still evident with an increase in the interval between reinforcements (Fig. 8). Additionally, the 800- and 1200-pmol cocaine groups decreased responding on the active lever when aCSF was substituted for cocaine and reinstated responding on the active lever when cocaine was restored (Figs. 5 and 6). Furthermore, the coadministration of the D$_2$-like receptor antagonist sulpiride reversibly reduced cocaine self-administration, which was reinstated when sulpiride was removed from the infusate (Fig. 7), suggesting that the reinforcing properties of cocaine in the AcbSh involves activation of D$_2$-like receptors.

The behavioral effects of cocaine have been linked to an indirect increase of DA levels that activates both D$_1$ and D$_2$ receptors postsynaptically (Spealman et al., 1992). Subsequently, cocaine self-administration (i.v.) is altered by either systemic (Haile and Kosten, 2001) or intra-accumbens (McGregor and Roberts, 1993; Caine and Koob, 1994; Phillips et al., 1994b) administration of D$_1$ and D$_2$ receptor agents. Self-administration of the DA-reuptake inhibitor nomifensine into the AcbSh is reduced by coadministration of the D$_2$ receptor antagonist sulpiride (Carlezon et al., 1995). Similarly, amphetamine ICSA into the Acb (shell-core boundary) is reduced by coadministration of either agonist alone did not support ICSA, suggesting that activation of both the D$_1$ and D$_2$ receptors in the Acb is required to produce reinforcing effects within the AcbSh. In the current study, only a D$_2$/3 receptor antagonist was used to reduce cocaine ICSA into the AcbSh, a D$_1$ receptor antagonist may have also reduced cocaine ICSA into the

![Figure 4](image-url)
It is possible that the coadministration of sulpiride reduced cocaine self-administration through a nonspecific reduction in activity or a general negative effect of the drug. However, this explanation seems unlikely because the initial level of responding was comparable after coadministration of sulpiride and aCSF substitution (Figs. 6 and 7), and sulpiride was self-administered at a slightly higher rate than aCSF.

In general, local administration of a drug can produce very different effects compared with systemic administration of the same drug. For example, there is a marked contrast between the effects of systemic (Haile and Kosten, 2001) or intra-acum-bens (McGregor and Roberts, 1993; Caine and Koob, 1994; Phillips et al., 1994b) administration of D₁ and D₂ receptor antagonists on i.v. self-administration of cocaine and cocaine ICSA. Under i.v. self-administration paradigms, administration of D₁ and D₂ receptor antagonists typically increases the amount of responding, and number of infusions received for cocaine (McGregor and Roberts, 1993; Caine and Koob, 1994; Phillips et al., 1994b; Haile and Kosten, 2002). The increase in responding and number of infusions has been asserted as a compensatory reaction so that the organism maintains the previously established level of reinforcement (Caine and Koob, 1994; Phillips et al., 1994b). Under ICSA conditions, coadministration of D₂/3 receptor antagonists reduced cocaine self-administration in the mPFC (Goeders and Smith, 1986; Goeders et al., 1986) and in the AcbSh. Additionally, increasing the concentration of cocaine/infusion did not alleviate the reduction of self-administration after coadministration of sulpiride with cocaine in the mPFC (Goeders et al., 1986). Conceptually, the likelihood of a compensatory increase in self-administration under ICSA conditions is less than under i.v. self-administration because the ICSA paradigm results in a local administration of both drug and neurotransmitter agent. Therefore, the animal cannot “overcome” the effects of the agent on the reinforcing properties of the reinforcer by increasing self-administration of the reinforcer because an equal amount of agent will also be directly administered into the neuroanatomically specific location after each operant response.

Behaviorally, the rapid acquisition of self-administration of reinforcers directly into specific neuroanatomical loci has previously been shown for the self-administration of ethanol (Gatto et al., 1994; Rodd-Henricks et al., 2000), acetaldehyde (Rodd-Henricks et al., 2002), morphine (Bozarth and Wise, 1981), and other opioid agonists (Devine and Wise, 1994) into the VTA. Goeders and Smith (1983) reported that cocaine self-administration into the mPFC occurred within the first 8-h session. Additionally, self-administration of the DA up-

---

**Fig. 5.** Depicted is the number of active and inactive lever responses (mean ± S.E.M.) for rats self-administering aCSF, or 400 or 800 pmol of cocaine/100 nl of aCSF into the AcbSh or AcbC. Asterisks (p < 0.05; Tukey’s test) represent significant difference in responses on the active lever versus responses on the inactive lever and responding observed for rats self-administering aCSF (p values < 0.05; determined by one-way ANOVAs performed on individual sessions).
take inhibitor nomifensine into the AcbSh was apparent within the second 3-h session (Carlezon et al., 1995).

The delay in the acquisition of self-administration when the time-out period between infusions was extended (Fig. 8) is predicted by most learning theories, which hold that prolonged separations between reinforcements should delay the acquisition of learning, i.e., partial reinforcement and development of superstitious behaviors (Macintosh, 1977; c.f. Domjan and Burkhard, 1982: discussion on probability learning theory, Pavlovian temporal contiguity, and instrumental learning variables). Examination of the pattern of self-administration for 1200 pmol of cocaine (Fig. 4) indicates an initial high period of drug delivery maintained a lower level of self-infusion, followed by a return to a higher level of self-administration at the conclusion of the session. Previously, we reported a similar pattern of self-administration for

Fig. 6. Depicted is the number of active and inactive lever responses (mean ± S.E.M.) for rats self-administering 1200 or 1600 pmol cocaine/100 nl of aCSF into the AcbSh or AcbC. Asterisks represent significant ($p < 0.05$; Tukey’s test) difference in responses on the active lever versus responses on the inactive lever and responding observed for rats self-administering aCSF ($p$ values < 0.05; determined by one-way ANOVAs performed on individual sessions).

Fig. 7. Depicted is the number of active and inactive lever responses (mean ± S.E.M.) for rats self-administering 1200 pmol/100 nl alone (sessions 1–4 and 7–8) or coadministering 400 pmol/100 nl of sulpiride and 1200 pmol of cocaine/100 nl (sessions 5–6) into the AcbSh. Asterisks represent significant ($p < 0.05$; Tukey’s test) difference in responses on the active versus inactive lever and from responding observed for rats self-administering sulpiride alone ($p$ values < 0.05).
ethanol (Rodd-Henricks et al., 2000) and acetaldehyde (Rodd-Henricks et al., 2002) into the posterior VTA. Additionally, Carlezon et al. (1995) reported that nomifensine self-administration into the AcbSh was pronounced during the initial 30-min period, but they did not observe the higher self-administration near the end of the session. Previous research attempting to determine whether cocaine would be self-administered into the Acb varied slightly from the current study. In the initial study (Goeders and Smith, 1983), it was reported that concentrations between 0 and 5000 pmol/100 nl of cocaine failed to support self-administration in the Acb. However, from the histological presentation of cannulae placements, it would seem that most of the placements from that study were located in the AcbC, an area that did not support self-administration of cocaine in the current study. Yet, there are a number of methodological distinctions, besides injector location, between the current study and the reports of Goeders and Smith. Goeders and Smith's experiments used male Fisher-344 rats (Goeders and Smith, 1983, 1986; Goeders et al., 1986), whereas the current experiment used female Wistar rats. Fisher-344 rats are not as responsive to cocaine as many other rat lines (i.e., Lewis rats) and are less likely to acquire i.v. self-administration of various drugs of abuse, including cocaine (Kosten et al., 1994, 1997; Ambrosio et al., 1995). Recently, Fisher-344 rats, which were selected and highly trained to self-administer cocaine, were found to differ from Lewis rats in response to D₁ and D₂ receptor agents on cocaine discrimination and i.v. self-administration (Haile and Kosten, 2001). Additionally, the initial ICSA studies were conducted with older EMIT units, used different environmental cues, and different temporal patterns than the current study (i.e., Goeders and Smith, 1983; session every 3rd day, session length 8 h). Thus, injector location, the use of a different rat line, and other experimental parameters may have contributed to different findings regarding the self-administration of cocaine into the Acb.

In addition to examining nomifensine self-administration in the AcbSh, Carlezon et al. (1995) reported some preliminary results examining cocaine self-administration into the AcbSh. Using a within-subject design to gradually increase the amount of cocaine/100 nl, during the 22nd to 24th session, Long-Evans rats reliably responded for 300 pmol/100 nl of cocaine (Carlezon et al., 1995). However, the current study, although using slightly higher concentrations of cocaine, established patterns of responding closely resembling cocaine self-administration into the mPFC (Goeders and Smith, 1983) that were obtained within seven sessions, which would lend greater assurance of injector site viability (Bozarth and Wise, 1981).

The concentration of cocaine that supported self-administration in the current study is approximately 8 to 12 mM. Cocaine self-administration in the mPFC was reported to occur at concentrations ranging from 0.5 to 0.9 mM (Goeders and Smith, 1983). Additionally, microinjection of cocaine directly into the nucleus accumbens, not delineated between shell and core, can induce locomotor activation at concentrations ranging from 16 to 35 mM (Delfs et al., 1990; Hemby et al., 1992) and condition a place preference between 40 and 65 mM (Liao et al., 2000). Thus, the concentrations used in the current study seem to be higher than effective doses of cocaine in other brain areas but are lower than microinjected concentrations of cocaine needed to induce locomotor activation and condition a place preference.

In addition to the reinforcing properties, cocaine can act as...
a local anesthetic agent (Wesson and Smith, 1977). It is possible that the lack of lever discrimination observed in rats self-administering cocaine into the AcbSh at the 1600 pmol/100-nl dose may be a result of nonspecific effects (e.g., local anesthetic) at this high concentration (Fig. 5). However, the fact that the D2 receptor antagonist sulpiride could effectively block the ICSA of cocaine would argue against a nonspecific anesthetic effect.

Repeated, intermittent exposure to cocaine has been shown to result in numerous molecular, neurochemical, and behavioral alterations that have been hypothesized to be the basis for cocaine sensitization (for reviews, see Koob and Nestler, 1997; Pierce and Kalivas, 1997). Repeated administration of cocaine results in a supersensitivity in the efficacy of reuptake blockade by cocaine (Henry and White, 1995). Similarly, repeat injections of amphetamine produced sensitization to the locomotor stimulation effect of a challenge dose of amphetamine administered directly into the Acb (Paulson and Robinson, 1991). In the current study, rats self-administering 400 pmol of cocaine displayed a unique response pattern after aCSF substitution (Figs. 3–5). The 400-pmol group received an average of 14 infusions (Figs. 3 and 4) and did not clearly demonstrate lever discrimination (Fig. 5) during the initial four sessions. However, when 400 pmol of cocaine was given after two extinction sessions, these rats increased responding on the active lever, showed robust lever discrimination (Fig. 5) and self-administered over 43 infusions (Fig. 3). In general, the data indicate that after a period of drug abstinence (a total of 6 days from session 4 to 7) a concentration of cocaine that produced low levels of self-infusions during acquisition was now able to support robust response-contingent behaviors. Therefore, the present data suggest that neurobiological alterations occurred directly in the AcbSh or in other areas associated with the AcbSh (e.g., feedback loops) over the course of the experiment that enhanced the reinforcing properties of cocaine at this dose. It is not clear why only the 400-pmol cocaine dose showed, after extinction, the increased infusions in session 7 compared with session 4 (Fig. 3). Perhaps this subthreshold dose, during the acquisition period, produced a “kindling-like” effect within the AcbSh. At the higher doses (800 and 1200 pmol), cocaine may be producing similar effects, but these effects are masked compared with the already high number of infusions received in sessions 2 to 4, or may occur during the acquisition period.

Past research has indicated that the Acb in general is important in the construction of instrumental associative learning between behaviors and reinforcing sensation (Colwill and Rescorla, 1989). The AcbSh has been hypothesized to coordinate movements and the encoding of incentive value of performed operants (Parkinson et al., 1998; Corbit et al., 2001). In contrast, the AcbSh has been hypothesized to mediate the excitatory effects of stimuli, the anticipation of reward, and goal-directed behaviors (Johnson et al., 1995; Sokoloski and Salamone, 1998; Sokoloski et al., 1998; Corbit et al., 2001). Therefore, the current finding that the AcbSh directly supports cocaine reinforcement is perhaps linked to the mediation of excitatory, anticipatory, and/or goal-directed behaviors of this structure. Although, the AcbSh did not support cocaine self-administration, it would be inappropriate to state that the AcbSh is not involved in the reinforcing properties of cocaine. Summarily, the present data suggest that the AcbSh is a neuronatomical substrate for the reinforcing properties of cocaine, involving the activation of D2-like receptors.

Acknowledgments
We thank Robert S. Crile and Bradley Glazier for assistance. Separate, corollary, preliminary findings concerning cocaine ICSA into the AcbSh were reported at the New York Academy of Science meeting in 1999.

References


