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Sex differences in the escalation of intravenous cocaine intake following long- or short-access to cocaine self-administration

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Abstract

Preclinical data have indicated that extended access to cocaine self-administration (e.g., 6–12 h/day) facilitates an escalation in daily cocaine intake that is not seen when rats are given shorter (e.g., 1–2 h/day) access to cocaine for self-administration. Data from studies with rats have shown that females self-administer more cocaine than males during all phases of drug abuse (e.g., acquisition, maintenance, and reinstatement). The purpose of this study was to examine potential differences between males and females in the escalation of intravenous cocaine intake following a differential access (e.g., 1 vs. 6 h) period of cocaine self-administration. Four groups of rats were compared: (1) long-access (LgA; 6 h) females; (2) LgA males; (3) short-access (ShA; 1 h) females; and (4) ShA males. Animals were given LgA or ShA to intravenous cocaine (0.5 mg/kg/infusion) self-administration under an FR 1 schedule for 21 days. Subsequently, access conditions were made equal (3 h) across groups, and dose–response curves for cocaine were compared. Results revealed that the LgA groups' dose–response curves were significantly elevated above those of ShA groups. Additionally, the dose–response curve of LgA female rats was significantly elevated above that of LgA male rats. These results suggest that female rats are more sensitive than male rats to factors that contribute to the escalation of cocaine intake (e.g., extended access conditions).

Keywords: Sex differences; Cocaine; Escalation; Intravenous; Gender; Differential access; Self-administration

1. Introduction

Cocaine is a highly addictive illicit drug that produces rapid, euphoric effects shortly after it is administered via insufflation (Oliveto et al., 2001), intravenous injection (Resnick and Kestenbaum, 1977; Walsh et al., 1996), or smoking (Foltin and Fischman, 1991). In order to experience the same euphoric effect over time, the user consumes more of the drug, often soon after the last administration. However, the euphoria produced by the first cocaine dose is not duplicated when subsequent doses are spaced too closely (Brower et al., 1986; Foltin et al., 2003; Trinkoff et al., 1990), and this frequently causes the user to take the drug in 'binges' or cycles of frequent, repeated dosing (Gawin and Kleber, 1985). These patterns of use result in chronic exposure to the drug that can lead to uncontrolled accelerated patterns of drug intake (e.g., escalation), which are defining features of addiction (Edwards, 1986; Gawin, 1991; Marlatt et al., 1988). In 2002, an estimated two million people or 0.9% of the population in the United States were current cocaine users (National Survey on Drug Use and Health (NSDUH), 2002). Of these users, 25.2% were classified as cocaine abusers or cocaine dependent (NSDUH, 2002). Therefore, identifying factors that may contribute to the progression from cocaine use to cocaine addiction, such as the escalation in drug intake, may be important for developing prevention and treatment strategies for cocaine addiction.

Sex is one factor that may be important to consider when designing strategies to prevent and treat cocaine addiction. For example, women reported greater negative physiological effects (e.g., nervousness) following cocaine use than men (Kosten et al., 1996). Women also took longer to detect the subjective effects of cocaine, and they reported less euphoria and dysphoria following cocaine administration compared to men (Lukas et al., 1996). Similar sex differences were reported in the subjective responses to smoked

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cocaine (Sofuoglu et al., 1999) and repeated 'binge'-smoked cocaine (Evans et al., 1999). It is possible that women may be more likely than men to escalate their cocaine use and to use the drug in an uncontrolled, erratic manner to achieve the euphoria they first experienced since their subjective euphoric responses to the drug are initially diminished relative to males.

Preclinical data have revealed sex differences during all phases of drug addiction in animal models of drug self-administration, and these differences typically occurred during transition periods of drug use (e.g., acquisition, controlled to uncontrolled intake, relapse). For example, female rats took less time than male rats to acquire intravenous cocaine, heroin (Lynch and Carroll, 1999), nicotine (Donny et al., 2000), and methamphetamine (Roth and Carroll, 2004) self-administration. During the maintenance of stimulant self-administration, female rats worked harder than males for single injections of cocaine (Roberts et al., 1989) or methamphetamine (Roth and Carroll, 2004), as indicated by higher breakpoints (BP) (maximum behavioral output) under progressive ratio (PR) schedules. Female rats regulated their intake of intravenous cocaine to a lesser extent than male rats (Lynch et al., 2000), and they exhibited greater levels of reinstatement for cocaine self-administration than males following a cocaine-free period (Lynch and Carroll, 2000).

One method that has been used to examine the progression from controlled drug intake to uncontrolled, escalated drug intake in animal models of drug self-administration was to vary the total time the animals had access to drug self-administration each day (Ahmed and Koob, 1998, 1999). Results from studies using this method revealed that animals given extended access (6 h/day) to cocaine self-administration displayed elevated dose–response curves under equal access conditions compared to animals previously given limited access (1 h/day) (Ahmed and Koob, 1998, 1999). These studies were extended to examine the effects of access conditions on the escalation and reinstatement of heroin self-administration (Ahmed et al., 2000). Rats given long (11 h) versus short (1 h) access to heroin self-administration also displayed an increased escalation in heroin intake, a slower extinction response following the cessation of heroin availability, and an increased reinstatement of heroin self-administration following exposure to stress (Ahmed et al., 2000). Mantsch et al. (2001) also used an extended access procedure to examine individual differences in cocaine self-administration in rats. The authors compared high responders (HR) and low responders (LR) to novelty during several phases of cocaine self-administration (e.g., acquisition, maintenance, escalation) during daily 10-h sessions. Higher levels of responding in HR versus LR rats were found under the extended access (10 h/day) conditions only at low cocaine doses, and they were surmountable by increasing the dose (Mantsch et al., 2001). In contract, recent work by Paterson and Markou (2004) using similar procedures with nicotine did not find escalation in male rats.

A differential access paradigm similar to that used by Ahmed and Koob (1998, 1999) was used in the present study to examine sex differences in the escalation of cocaine intake. Male and female rats were given either short access (ShA; 1 h) or long access (LgA; 6 h) to daily cocaine self-administration. Subsequently, daily access conditions were made equal (3 h) for all groups of rats, and dose–response curves for cocaine were obtained and compared across access groups. Males and females were compared to determine if differential access to cocaine self-administration affected the escalation of cocaine and if sex differences existed in the sensitivity to factors that contribute to the escalation of cocaine intake (e.g., extended access conditions).

2. Method

2.1. Animals

Thirty-three sexually mature (>90 days old), female ($n = 17$) and male ($n = 16$) Wistar (Harlan Sprague–Dawley) rats were used as subjects. Female rats weighed approximately 240–400 g, and male rats weighted approximately 340–540 g at the beginning of the experiment. Rats were same-sex pair housed for a minimum of 5 days upon arrival to the laboratory in plastic home cages with ad libitum access to food and water. After the acclimation period, each rat was implanted with a chronic indwelling catheter into the right jugular vein. Following catheterization, the rats were placed in individual test chambers where they remained for the duration of the experiment. The experimental rooms were temperature (24 °C)- and humidity-controlled with a 12:12-h light/dark cycle with lights on at 7:00 a.m. Rats received a daily ration (females = 16 g and males = 20 g) of ground Purina laboratory chow (Purina Mills, Minneapolis, MN) at 3:00 p.m. that maintained them at approximately 85% of their free-feeding body weights. The rats' reduced weights remained relatively stable throughout the entire experiment and were monitored by weighing each animal at least one time per week. Water was freely available throughout the experiment. The experimental protocol was approved by the University of Minnesota Institutional Care and Use Committee. Laboratory facilities were accredited by the Association for Assessment and Accreditation of Laboratory Animal Care (AAALAC), and experiments were conducted in accordance with the accepted principles of laboratory animal care (National Research Council, 2003).

2.2. Apparatus

Experimental test chambers were octagonally shaped with alternating Plexiglas and stainless steel walls that contained openings for the insertion of a drinking spout, a jar containing ground food, and two response levers that

remained extended into the test chambers at all times (Coulbourn Instruments, Lehigh Valley, PA). Above each lever was a light panel consisting of three colored stimulus lights (Coulbourn Instruments) that were illuminated during drug infusions. A house light (4.76 W) was located at the top of the chamber and was constantly illuminated during the experimental sessions. Each experimental chamber was enclosed in a sound-attenuating wooden box that contained a fan for ventilation and white noise. An infusion pump (RHSYOCKC, Fluid Metering, Oyster Bay, NY) was attached to a 500-ml reservoir containing the drug solution and was mounted outside the chamber. The reservoir was equipped with Tygon tubing (1.52 mm od; 0.51 mm id, Fisher Scientific, Springfield, NJ) that connected to a swivel (050–0022, Alice King Chatham, Hawthorne, CA) mounted at the top of the chamber. A spring-covered tether (C313CS, Plastic Products, Roanoke, VA) extended from the swivel and was connected to the rat by a plastic connector and section of needle tubing (C3236, Plastic One, Roanoke, VA) embedded in the center of a plastic infusion harness that protected the connection (Instech Laboratories, Plymouth Meeting, PA). The indwelling catheter in the rat was attached to the needle tubing embedded in the harness. An IBM-compatible computer with Med-PC interface (Med Associates, St. Albans, VT) was used for programming, data collection, and storage.

2.3. Drugs

Cocaine was provided by the National Institute on Drug Abuse (Research Triangle Institute, Research Triangle Park, NC) and was dissolved in sterile saline solution. Cocaine infusions (0.25, 0.5, 1.0, and 2.0 mg/kg/infusion) were delivered at a rate of 0.025 ml/s and the infusion duration was 1 s/100 g of body weight. Infusion durations ranged from 2.4 to 5.4 s, and volumes ranged from 0.06 to 0.135 ml. Cocaine solutions were made weekly and refrigerated, but they were added to individual drug reservoirs at room temperature.

2.4. Procedure

Prior to the catheter implantation, rats were anesthetized with ketamine (90 mg/kg) and pentobarbital (10 mg/kg). Atropine (0.02 mg/kg) was also administered to facilitate respiration. Subsequently, a chronic, indwelling silicon catheter was inserted into the right jugular vein as previously described (Carroll and Boe, 1982; Lynch et al., 2000; Weeks, 1972). One end of the catheter terminated at the opening of the right atrium. The free tip of the catheter was led subcutaneously to a medial incision approximately 1 cm caudal to the scapulae, and from there it was attached to the needle tubing embedded in the plastic harness. The day after surgery, the tether system was attached to the needle tubing of the plastic harness. Rats were allowed to recover from surgery for 3 days and they were administered gentamycin (2 mg/kg iv) and heparinized saline (10 IU/kg iv) daily during this recovery period to prevent infection and catheter blockage.

Following recovery from surgery, rats were assigned to one of 4 groups: (1) LgA females ($n=9$); (2) ShA females ($n=8$); (3) LgA males ($n=7$); and (4) ShA males ($n=9$). Experimental sessions were conducted 7 days per week, beginning at 10:00 a.m. Body weights were monitored weekly, and catheter patency was confirmed approximately every 7 days with an injection of sodium methohexital (5 mg/kg iv). Patency of the catheter was assumed if there was an immediate loss of the righting reflex. If the catheter was not patent, rats were reanesthetized, a catheter was implanted in their left jugular vein, and they were returned to the experiment following 3 days of recovery.

2.4.1. Predifferential access

Initially, all rats were given 2 h (10:00 a.m. to 12:00 p.m.) daily access to intravenous cocaine (0.5 mg/kg/infusion) self-administration under a fixed ratio (FR) 1 schedule of reinforcement. Following the administration of each cocaine infusion, a 20-s timeout occurred during which stimulus lights remained on, and responses were recorded but had no programmed consequences. Rats were required to administer an average of 40 infusions/day over 4 consecutive days before they moved on to the differential access phase of the experiment.

2.4.2. Differential access

After rats met the infusion criterion for the predifferential access phase of the experiment, they were placed on their respective self-administration access procedures. The LgA groups had access to 6 h (10:00 a.m. to 4:00 p.m.) of cocaine (0.5 mg/kg/infusion) self-administration, and the ShA groups had access to 1 h (10:00 to 11:00 a.m.) of cocaine self-administration. The behavioral schedule remained at an FR 1 with a 20-s timeout period following each infusion during which stimulus lights remained illuminated and responses were recorded but not reinforced. These conditions were held constant for 21 days to allow enough exposure to the differential access conditions in order to assess escalation of drug intake.

2.4.3. Postdifferential access/dose–response

Following the differential access phase of the experiment, all groups of rats were switched to equal access (3 h, 10:00 a.m. to 1:00 p.m.) self-administration sessions. One of 4 cocaine doses (0.25, 0.5, 1.0, or 2.0 mg/kg/infusion) was initially available and the presentation of the initial dose was counterbalanced across rats. An FR 1 schedule with a 20-s timeout consisting of the same conditions described above was used for this portion of the experiment. Following the collection of 4 days of stable responding (no increasing or decreasing trends in responding), the cocaine dose was nonsystematically changed from the initial dose to each of the other three doses (0.25, 0.5, 1.0, or 2.0 mg/kg/infusion). In order to account for possible order effects, the lowest

dose of cocaine (0.25 mg/kg/infusion) never immediately followed the highest dose (2.0 mg/kg/infusion). Responding was allowed to stabilize for 4 days at each dose of cocaine in order to construct dose–response curves for each group of rats.

2.5. Data analysis

All statistical analyses were computed using GB Stat (Dynamic Microsystems, Silver Spring, MD). Independent measures consisted of sex (male vs. female), access condition (LgA vs. ShA), day (1–21), hour (1–6), and cocaine dose (0.25, 0.5, 1.0, and 2.0 mg/kg/infusion). Dependent measures were number of cocaine infusions and cocaine intake (mg/kg). The effects of sex and access condition on the number of cocaine infusions self-administered during Days 1–21 of the differential access period and on the first (Day 1) and last (Day 21) day of the differential access period were determined using three-way repeated measures analyses of variance (ANOVA). A three-way repeated measures ANOVA was also used to assess the effects of access condition and sex on the number of cocaine infusions self-administered at each dose of cocaine during equal access self-administration sessions. A two-way repeated measures ANOVA was used to examine the effects of sex and hour (1–6) on number of cocaine infusions self-administered in the LgA groups. Separate two-tailed Student's *t* tests were used for additional a priori group comparisons. Fisher's least significant difference (LSD) protected *t* tests were used for all post hoc analyses. A value of $P<.05$ determined statistical significance.

3. Results

3.1. Differential access period (Days 1–21)

Fig. 1 illustrates the mean (± S.E.M.) number of cocaine (0.5 mg/kg/infusion) infusions self-administered over the 21-day differential access period for each group of rats. Statistical analysis revealed that there was a significant main effect of sex [$F(1,692)=3.56$, $P<.05$] and access condition [$F(1,692)=149.01$, $P<.05$], but not day (i.e., 1–21) on number of infusions self-administered across the 21-day differential access period. There were no significant interactions among sex, day, and/or access condition. Both the LgA male and LgA female rats self-administered significantly more cocaine infusions than ShA male and ShA female rats across the 21 days. Post hoc analyses revealed that within the LgA groups, females self-administered significantly more cocaine infusions than males at Days 7, 8, 20, and 21 ($t=2.52$, $df=15$, $P<.05$; $t=6.13$, $df=15$, $P<.05$; $t=2.81$, $df=15$, $P<.05$; and $t=3.38$, $df=15$, $P<.05$, respectively).

The effects of access condition and sex on the increase in cocaine self-administration from Days 1 to 21 were also assessed. There was a significant main effect of access condition [$F(1,71)=87.38$, $P<.05$], but not sex or day on number of cocaine infusions self-administered on Days 1 versus 21. There were no significant interactions among sex, day, and/or access condition. Post hoc tests indicated that LgA females self-administered significantly more cocaine infusions on Day 1 than ShA females and ShA males ($t=3.92$, $df=16$, $P<.05$ and $t=3.69$, $df=17$, $P<.05$, respectively). The LgA females also self-administered significantly more infusions on Day 21 relative to LgA males, ShA females, and ShA males ($t=2.27$, $df=15$, $P<.05$; $t=6.30$, $df=16$, $P<.05$; and $t=6.88$, $df=17$, $P<.05$, respectively). The LgA males self-administered significantly more cocaine infusions than ShA females and ShA males on Day 1 ($t=3.70$, $df=14$, $P<.05$ and $t=3.41$, $df=15$, $P<.05$, respectively) and on Day 21 ($t=3.70$, $df=14$, $P<.05$ and $t=4.16$, $df=15$, $P<.05$, respectively). Over the entire 21-day period, LgA female rats progressively increased their cocaine intake from an average of 85.89 (± 15.34) infusions on Day 1 to an average of 171.33 (± 40.91) infusions on Day 21. The LgA male rats self-administered an average of 85.14 (± 14.63) and 91.0 (± 7.61) infusions on Days 1 and 21, respectively.

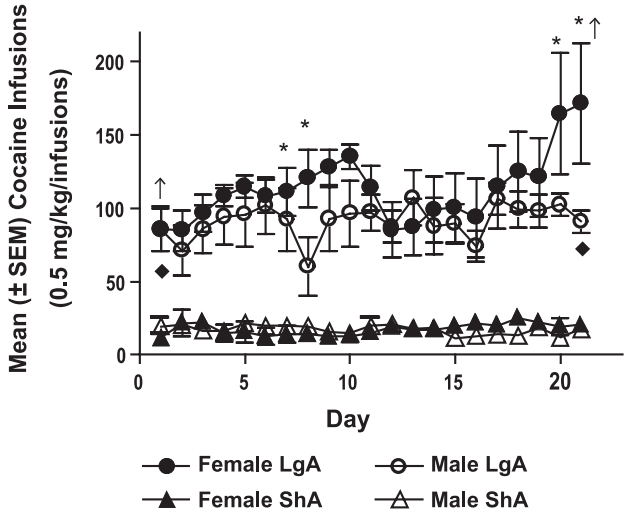

Fig. 1. Mean (± S.E.M.) daily cocaine (0.5 mg/kg/infusion) infusions over the 21-day differential access period for female (filled symbols) and male (open symbols) ShA (triangles) and LgA (circles) groups. Asterisks (*) indicate that LgA female rats self-administered significantly more cocaine than LgA male rats at Days 7, 8, 20, and 21. The diamonds (♦) indicate that LgA males self-administered significantly more cocaine than ShA males and ShA females on Days 1 and 21. The arrows (↑) indicate that LgA females self-administered significantly more cocaine than ShA males and ShA females on Days 1 and 21.

The top frame of Fig. 2 illustrates mean (± S.E.M.) number of daily cocaine infusions and mean (± S.E.M.) total cocaine intake (mg/kg) during daily sessions across the 21-day differential access period for each group of rats. The LgA female rats self-administered significantly more infusions ($t=2.52$, $df=15$, $P<.05$) and consumed significantly more cocaine in milligram per kilogram ($t=3.08$, $df=15$,

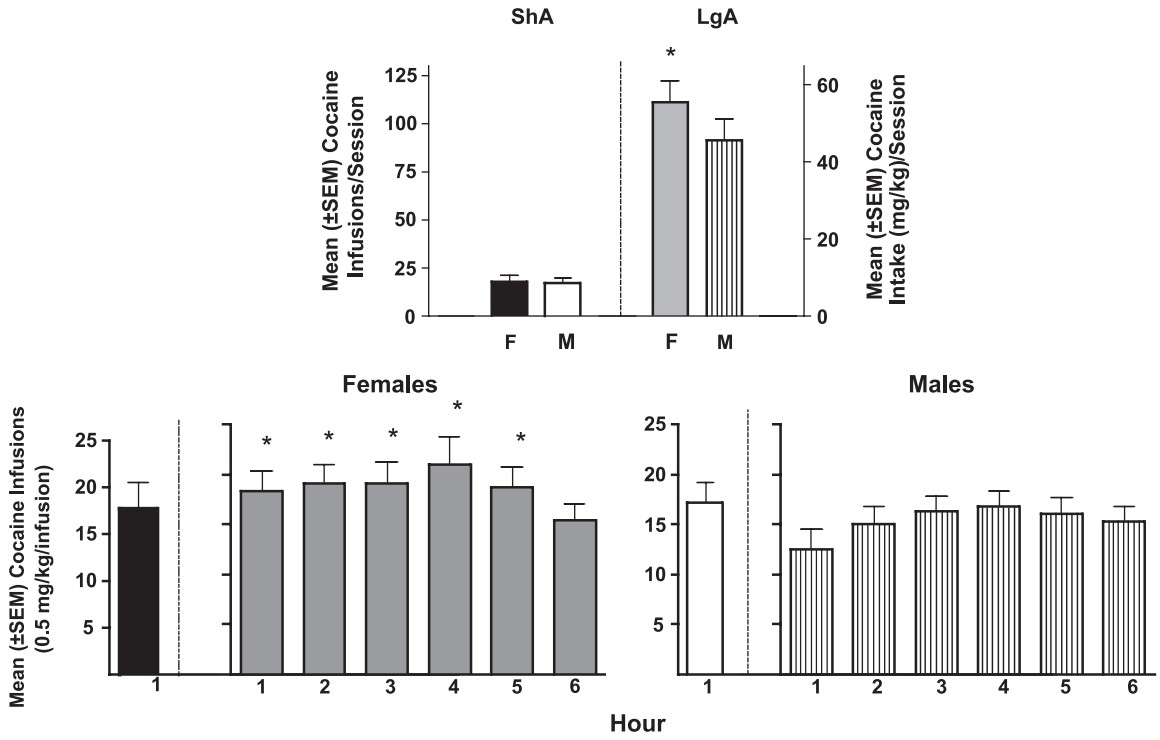

Fig. 2. The upper frame represents mean total (± S.E.M.) daily cocaine infusions (left *y*-axis) and intake (mg/kg) (right *y*-axis) for each group of rats during the 21-day differential access period. The asterisk (*) indicates that LgA female rats (gray bar) self-administered significantly more cocaine infusions and consumed significantly more cocaine (mg/kg) than LgA male rats (striped bar). Lower frames present mean (± S.E.M.) cocaine (0.5 mg/kg/infusion) infusions as a function of individual hours in both ShA and LgA groups. The asterisks (*) indicate that female LgA rats (gray bars) self-administered significantly more cocaine than male LgA rats (striped bars) during hours 1–5 of their daily sessions.

$P<.05$) compared to the LgA males during the differential access period. As expected, the LgA female and male rats self-administered significantly more cocaine than ShA female and male rats during this period ($t=23.62$, $df=16$, $P<.05$ and $t=20.82$, $df=15$, $P<.05$, respectively). LgA female and male rats also consumed significantly more cocaine in milligrams per kilogram during the differential access period than the ShA female and male rats ($t=19.26$, $df=16$, $P<.05$ and $t=20.50$, $df=15$, $P<.05$, respectively).

The bottom frame of Fig. 2 depicts the hourly distribution of the mean (± S.E.M.) number of cocaine (0.5 mg/kg/infusion) infusions self-administered for ShA female (black bars), LgA female (gray bars), ShA male (white bars), and LgA male (striped bars) rats during each hour of their daily self-administration sessions during the 21-day differential access period. There was a significant main effect of sex on the number of cocaine infusions self-administered in LgA rats [$F(1,1907)=15.13$, $P<.05$]. There was also a significant main effect of hour [$F(5,1907)=7.22$, $P<.05$] and a significant Sex × Hour interaction [$F(5,1907)=4.92$, $P<.05$]. Overall, LgA females self-administered significantly more cocaine infusions than LgA males. Post hoc tests revealed that LgA females self-administered significantly more cocaine infusions than LgA males during Hours 1, 2, 3, 4, and 5 ($t=7.22$, $df=15$, $P<.05$; $t=4.31$, $df=15$, $P<.05$; $t=3.44$, $df=15$, $P<.05$; $t=3.48$, $df=15$, $P<.05$; and $t=3.20$, $df=15$, $P<.05$, respectively). Within the LgA female group, rats self-administered significantly more cocaine infusions during Hours 1, 2, 3, 4, and 5 compared to Hour 6 ($t=4.58$, $df=8$, $P<.05$; $t=4.19$, $df=8$, $P<.05$; $t=4.83$, $df=8$, $P<.05$; $t=5.64$, $df=8$, $P<.05$; and $t=3.76$, $df=8$, $P<.05$, respectively). Additionally, within the LgA male group, rats self-administered significantly more cocaine infusions during Hours 2, 3, 4, 5, and 6 ($t=2.40$, $df=6$, $P<.05$; $t=3.77$, $df=6$, $P<.05$; $t=4.44$, $df=6$, $P<.05$; $t=3.06$, $df=6$, $P<.05$; and $t=2.48$, $df=6$, $P<.05$, respectively) compared to Hour 1.

3.2. Escalation of cocaine intake

In Fig. 3, mean cocaine infusions are presented for each of the four groups of rats across 4 cocaine infusion doses. The left panels of Fig. 3 represent the mean (± S.E.M.) number of cocaine infusions self-administered by female (top left) LgA (filled circles) and ShA (filled triangles) rats and male (bottom left) LgA (open circles) and ShA (open triangles) rats during the 3-h self-administration sessions. In the right panels of Fig. 3, the mean (± S.E.M.) number of cocaine infusions is compared for ShA (top right) female (filled triangles) and male (open triangles) rats and LgA (bottom right) female (filled circles) and male (open circles) rats during the daily 3-h self-administration sessions. Statistical analysis revealed that there were main effects of sex and cocaine dose (0.25, 0.5, 1.0, and 2.0 mg/kg/infusion),

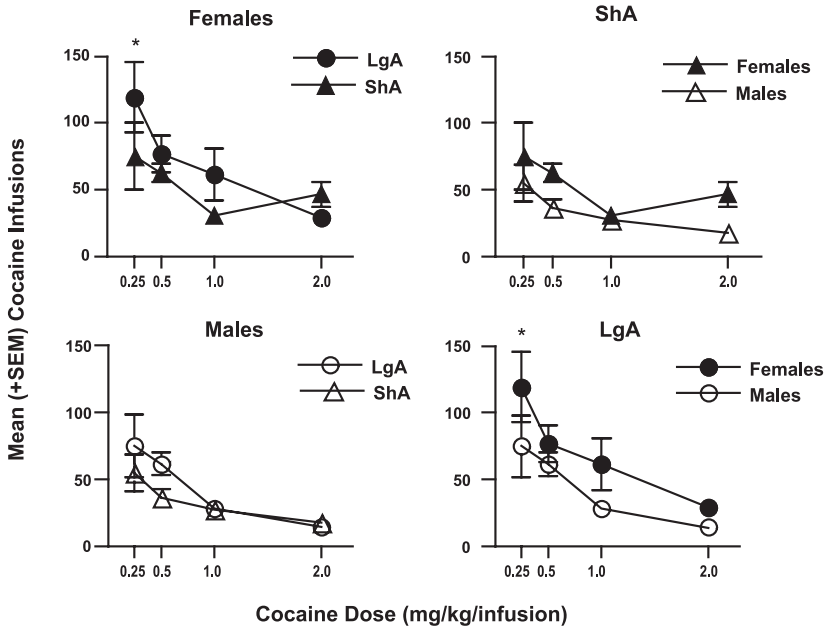

Fig. 3. Mean (± S.E.M.) cocaine infusions under an FR 1 schedule during the 3-h equal access conditions are presented as a function of cocaine dose for each group of rats. The left panels compare female (top left) LgA (filled circles) and ShA (filled triangles) rats and male (bottom left) LgA (open circles) and ShA (open triangles) rats. The asterisk (*) indicates that LgA females self-administered significantly more cocaine than ShA females at the 0.25 mg/kg/infusion dose. The right panels compare ShA (top right) female (filled triangles) and male (open triangles) rats and LgA (bottom right) female (filled circles) and male (open circles) rats. The asterisk (*) indicates that LgA females self-administered significantly more 0.25 mg/kg/infusion cocaine compared to LgA males.

but not access condition on number of cocaine infusions self-administered during the 3-h self-administration sessions [$F(1,131) = 9.00$, $P < .05$ and $F(3,131) = 18.33$, $P < .05$, respectively]. There were no significant interactions among sex, cocaine dose, and/or access condition. Post hoc tests revealed that LgA female rats self-administered significantly more cocaine infusions than LgA males, ShA males, and ShA females at the 0.25 mg/kg/infusion dose ($t = 2.47$, $df = 15$, $P < .05$; $t = 3.91$, $df = 17$, $P < .05$; and $t = 2.57$, $df = 16$, $P < .05$, respectively).

4. Discussion

Previous escalation studies have been focused mainly upon male rats. In this study, we extended the escalation paradigm to female rats. In general, under equal access conditions, rats that had previously been allowed LgA to cocaine self-administration displayed dose–response curves that were shifted vertically upward relative to the ShA rats. However, female rats were more sensitive than male rats to the extended access conditions, as indicated by a significant increase in the number of cocaine infusions self-administered at the 0.25 mg/kg/infusion dose in the LgA females compared to the ShA females. In the male groups (ShA and LgA), prior length of access to cocaine self-administration did not significantly affect the subsequent escalation of cocaine intake at any of the doses tested. Additionally, the dose–response curve of the LgA female rats was shifted vertically upward relative to the LgA male rats, with LgA females self-administering significantly more cocaine infusions at the lowest dose tested (0.25 mg/kg/infusion). During the differential access period (21 days) in the present study, LgA female rats increased their cocaine intake from Days 1 to 21 to a significantly greater extent than LgA males. Also, LgA female rats self-administered significantly more cocaine infusions and consumed more cocaine in milligram per kilogram body weight than LgA male rats under an FR 1 schedule of reinforcement. Therefore, these results concur with those from previous studies showing that female rats self-administer more cocaine than male rats (Carroll et al., 2002; Lynch and Carroll, 1999; Morse et al., 1993; Roberts et al., 1989).

In the present study, daily duration of access to cocaine self-administration (1 vs. 6 h) did not significantly influence subsequent cocaine intake in male rats. These results differ from those obtained by Ahmed and Koob (1998, 1999). One reason for this may have been attributable to differences in procedures between the present study and those of Ahmed and Koob's (1998, 1999). In the previous studies, dose–response testing sessions alternated with days of 1 (ShA)- or 6-h (LgA) sessions of self-administration. This may have allowed the LgA rats to maintain dependence upon cocaine. In the present study, dose–response testing was conducted during the 3-h sessions without any alternation between access conditions (1 vs. 6 h). This may have resulted in diminished differences between access conditions and may have obscured escalation effects in male rats. Another reason for differences in results between the present study and Ahmed and Koob's (1998, 1999) studies may be the

dose of cocaine that was used. In the present study, all of the rats received 0.5 mg/kg/infusion cocaine during their respective access conditions. In Ahmed and Koob's (1998, 1999) studies, male rats received a fixed amount of cocaine (0.25 mg) and they weighed between 280 and 460 g; therefore, the unit dose of cocaine was higher (approximately 0.54–0.89 mg/kg/infusion) and may have produced increased self-administration. It has been reported that lower, threshold doses of cocaine are optimal for examining sex differences in rats (Donny et al., 2000; Lynch and Carroll, 1999; Roth and Carroll, 2004) and in fact, the significant sex difference in the LgA rats (Fig. 3) was found at the lowest cocaine dose (0.25 mg/kg). It should be noted that the lowest dose of cocaine (0.25 mg/kg/infusion) used for the dose–response curve in the present study was not low enough to produce an ascending limb on the curve. This limits the interpretation of the data due to the fact that it is not possible to conclude whether there was a horizontal shift of the curve to the right or the left across groups of rats. The dose–response function in Ahmed and Koob's (1998, 1999) studies included a lower dose of cocaine (approximately 0.07–0.11 mg/kg/infusion, depending upon body weight). In one of these studies, the dose–response curve included an ascending limb (1998); however, in the other study, there was no ascending limb in the curves (1999). There was no indication of a horizontal shift in the functions between LgA and ShA groups in either of the studies referred to above (Ahmed and Koob, 1998, 1999). It is important that future studies investigating sex differences in the escalation of cocaine intake include lower doses to determine whether there are horizontal shifts in the dose–response curves between males and females and different access conditions (e.g., LgA vs. ShA).

In other studies using a differential access procedure similar to that used in the present study, drug consumption in LgA rats was characterized by an increased early (e.g., Hour 1 of the 6-h sessions) drug loading period (Ahmed and Koob, 1998, 1999). However, in the present study, the only difference observed during Hour 1 of cocaine self-administration under the 6-h access condition was that female rats self-administered significantly more cocaine infusions compared to males. In fact, cocaine infusions were relatively stable across each hour of the 6-h sessions in both LgA male and female rats. In other studies using this procedure, cocaine consumption during the first hour was evaluated by 10-min intervals (Ahmed and Koob, 1998, 1999). In the present study, total cocaine infusions for each hour were available and the data were not evaluated by shorter time intervals; therefore, drug loading during the first part of the first hour may have been obscured. Differences in the temporal patterns of responding during long-access self-administration in previous studies may also have been due to cocaine dose, strain of rat, and/or other differences in experimental conditions. It should also be noted that in the present study, rats were maintained at approximately 85% of their free feeding body weights. Since food restriction has been demonstrated to increase drug self-administration in animals (Carroll, 1985; Carroll et al., 1979; Comer et al., 1995), reduced food availability may have facilitated stress-induced cocaine self-administration in all of the groups. Additionally, female rats have been shown to be more sensitive to food restriction conditions (Carroll et al., 2001). Thus, in the present study, food restriction may have increased the probability for females to self-administer more cocaine. Finally, there is evidence suggesting reduced behavioral responding to stimulants and opioids under different testing environments. Specifically, acute psychomotor responses and the development of sensitization to amphetamine (Crombag et al., 2000) and morphine (Badiani et al., 2000) are attenuated if the drugs are administered in a rats' home cage versus a separate testing environment. In the present study, rats lived in their experimental test chambers. Although it has been shown that there was no effect of environment on acute psychomotor responses to intravenous cocaine, cocaine-induced sensitization was diminished in rats presented with the drug in their home environment compared to their testing environment (Browman et al., 1998). This may have diminished the responding for cocaine in the present study since animals lived in their testing environment and cues associated with drug access may not have been as salient as when different environments are used.

The sex differences observed in the present study correspond to data obtained from other studies using animal models of stimulant self-administration. Specifically, female rats consumed more caffeine (mg/kg) compared to males (Heppner et al., 1986), and female rats that had previously shown an increased rate of acquisition of cocaine self-administration maintained higher levels of cocaine intake (mg/kg) than males under an FR 1 schedule of reinforcement (Lynch and Carroll, 1999). Sex differences have also been reported in responding for stimulants under PR schedules of reinforcement where BPs were used to evaluate the reinforcing strength of the drug. Specifically, female rats reached higher BPs than male rats under a PR schedule for cocaine (Carroll et al., 2002; Hecht et al., 1999; Roberts et al., 1989), nicotine (Donny et al., 2000), and methamphetamine (Roth and Carroll, 2004) self-administration.

There has only been one other study in which sex differences in the intake patterns of cocaine self-administration in rats were examined. In this study, a two-lever self-administration procedure that allowed animals to control both the dose size and interdose interval of the drug infusion was used to examine sex and hormonal effects on the regulation and dysregulation of cocaine intake (Lynch et al., 2000). The dysregulation of cocaine self-administration was defined as a lower correlation between dose size and interdose interval. Typically, the rats consumed more cocaine because the postreinforcement pause was shortened. The results showed that after stable responding for cocaine was achieved, the regulation of cocaine intake was disrupted

more in female rats than in male rats, and the greatest disruption occurred during the estrus phase (levels of estrogen rapidly decline from peak levels that occurred in proestrus) of the estrous cycle in females (Lynch et al., 2000).

It has also been suggested that there are sex differences in the neural systems mediating cocaine reinforcement. For example, stimulant-induced dopamine release in brain regions implicated to be important in drug abuse (e.g., striatum) was greater in female than male rats (Becker, 1999; Becker and Ramirez, 1981). In vivo and in vitro data revealed that dopamine release and uptake rates were enhanced in the caudate nucleus of female versus male rats (Walker et al., 2000). Sex differences also occurred independently of gonadal hormones. Specifically, ovariectomized female rats acquired cocaine self-administration faster (Hu et al., 2003), exhibited greater behavioral sensitization to cocaine (Hu and Becker, 2003), and displayed enhanced dopamine functioning in the striatum (Bazzett and Becker, 1994; Castner et al., 1993; McDermott et al., 1994) than castrated male rats.

In the present study, the effects of ovarian hormones on the escalation of cocaine self-administration in female rats were not examined; however, estrogen has been implicated in the observed sex differences in animal models of stimulant self-administration (see Donny et al., 2000; Hecht et al., 1999; Lynch et al., 2000; 2001; Roberts et al., 1989). Estrogen also enhanced stimulant-induced dopamine release in the striatum (Becker, 1999) and dopamine reuptake in the nucleus accumbens (Thompson, 1999) in female rats. In future studies, it will be important to investigate the role that estrogen and the estrous cycle may play in the sex differences observed in the escalation of drug intake in rats.

It has been suggested that the regulation of drug intake implies that each individual has an endogenous set point for a preferred level of drug effects (Ahmed and Koob, 1998). The progression from drug use to drug addiction may involve the alteration in this set point along with the subsequent permanent deviation from regular patterns of drug use. The sex differences presented in this paper suggest that female rats may be more vulnerable than male rats to conditions that facilitate escalated patterns of cocaine intake (e.g., extended access conditions). This potential vulnerability in females may be related to differences in cocaine's effects. For instance, preclinical data revealed that female rats displayed a decreased ability to regulate cocaine intake relative to male rats after stable levels of responding were achieved (Lynch et al., 2000). Also, clinical data indicated decreased subjective ratings of euphoria in women relative to men following repeated exposure to cocaine (Evans et al., 1999; Lukas et al., 1996; Sofuoglu et al., 1999). It is also possible that female rats' increased vulnerability to the escalation in cocaine intake that occurs following prolonged exposure to the drug is a direct result of a greater propensity for prolonged drug exposure to change endogenous set points in the ability of cocaine to produce its reinforcing effects in female rats relative to male rats.

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