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Enhanced cocaine self-administration in adult rats with adolescent isolation experience

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Abstract

It is widely accepted that early environmental influences may affect the behavior of adult animals and their responses to psychotropic drugs. Rearing animals in isolation is a relevant paradigm for studying early life stress and for understanding the development of certain neurological and psychiatric diseases. The present study evaluated the effect of adolescent isolation on intravenous cocaine self-administration in adult rats. Male Sprague–Dawley rats were raised from postnatal day 22 to 55 either alone (isolated) or in groups of four per cage (grouped). Then, rats were trained for cocaine self-administration. Our results showed that both isolated and grouped rats acquired stable cocaine self-administration during 5 days of self-administration training. Numbers of both lever presses and cocaine infusions in isolated rats were significantly more than those in grouped rats. Especially, numbers of incorrect lever presses in isolated rats were significantly more than those in grouped rats. In addition, the intervals of inter-reinforcement for cocaine in isolated rats were significantly shorter as compared with grouped rats. These results indicate that rats with adolescent isolation experience have enhanced cocaine self-administration behavior.

Keywords: Cocaine; Intravenous self-administration; Isolation

1. Introduction

Events experienced in early life may contribute to the expression or exacerbation of a variety of physical and psychological disorders. A strong association between psychosocial stressors in early life and increased risks for depression, anxiety and substance abuse in adulthood exists (Kendler et al., 2000). Rearing animals in isolation is a relevant paradigm for studying early life stress and for understanding the genesis of certain neurological and psychiatric diseases (Myhrer, 1998; Whitaker-Azmitia et al., 2000).

Previous studies reported that neonatal isolation (rats with isolation stress on postnatal days 2 to 9) increases the rate of self-administrated addictive drugs such as ethanol (Schenk et al., 1990), morphine (Alexander et al., 1981), heroin (Bozarth et al., 1989), and cocaine (Schenk et al., 1987) in adult rats. In particularly, many studies suggest that neonatal isolation increases the reinforcing efficacy of cocaine and results in the enhancement of the propensity to self-administer cocaine (Matthews et al., 1999; Kosten et al., 2000; Zhang et al., 2005). In contrast, adulthood isolation has been demonstrated not to affect the self-administration of cocaine (Bozarth et al., 1989). Whereas, the effect of adolescent isolation (rats with isolation stress on postnatal days 22 to 55), which could induce significant modification in neurogenesis (Lu et al., 2003; Lapiz et al., 2003), on self-administration of addictive drugs is still unclear. Thus, in the present study, we investigate the effect of adolescent isolation on intravenous self-administration of cocaine in adult rats.

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2. Materials and methods

2.1. Animals and drug

Male Sprague–Dawley rats (Shanghai Center of Experimental Animals, Chinese Academy of Sciences) were maintained on a constant light/dark cycle with constant temperature and humidity. All animal treatments were strictly in accordance with the National Institutes of Health Guide for the Care and Use of Laboratory Animals. Cocaine hydrochloride was purchased from Qinghai Pharmaceutical Factory, China, and dissolved in 0.9% saline.

2.2. Apparatus for self-administration

Eight operant chambers (32 cm wide × 25 cm high × 25 cm deep, Med Associates) contained within a sound attenuating box with a ventilation fan were used in the experiment. Each chamber was equipped with a standard lever, a stimulus light above the lever, a house light and a beep. A software-operated infusion pump placed outside the sound-attenuating box delivered intravenous cocaine infusion, through a counter-balanced single-channel liquid swivel. Animals were tethered to the counter-balanced arm by a metal spring and skull-mounted plastic-post. A lab computer using the software provided by Med Associates controlled the apparatus.

2.3. Isolation procedure

Rats were divided into two groups and reared respectively under two different conditions from postnatal day 22 to 55: isolation rearing (in opaque plastic containers, one per container) and group rearing (in wire-net fencing cages, four per cage). From postnatal day 56 (the day of intravenous catheter implantation surgery) to the end of the experiment (postnatal day 65) all animals were housed singly in wire-net cages placed side-by-side. From postnatal day 22 to 50, the rats were given free access to water and food until their body weights reached approximately 250 g. Thereafter, they were given food at the end of each day at 15–20 g/day during food training and cocaine self-administration testing (postnatal days 51–55 and 61–65, respectively). They were given food and water available ad libitum during the period of surgery and recovery (postnatal days 56–60).

2.4. Training procedures

During the last 5 days under isolation and group housing conditions (from postnatal day 51 to 55), animals were trained to press lever for food pellets. Food training was carried out on a continuous reinforcement schedule (Fixed-Ratio 1) with time-out duration of 5 s until the subjects reached the criterion of over 100 lever presses per hour in all 3 1-h periods in a daily 3-h training session. Food training typically took 5 days. Once reaching the criterion of food training, animals were subjected to intravenous catheter implantation surgery (Caine and Koob, 1993) and allowed to recover for 5 days before cocaine self-administration training.

From postnatal day 61 to 65, rats were trained to acquire cocaine self-administration under a continuous reinforcement schedule (Fixed-Ratio 1) with time-out duration of 5 s during a daily 3-h session. The beginning of the session was marked by illumination of the house light and two intravenous priming injections of cocaine to displace the heparinized saline and fill the catheter with drug. Subsequent depression of the lever press resulted in the intravenous cocaine infusion (0.5 mg/kg/infusion, 50 μl over 4 s) accompanied by the extinction of the house light, illumination of the stimulus light, and simultaneously the presence of an audible tone for 4 s. In the 5 s time-out period, the house light was illuminated, and the stimulus light and the audible tone were extinguished. During cocaine infusions and time-out phases, the lever presses were recorded but had no responses. The criterion for acquisition of cocaine self-administration was 21 or more infusions during a self-administration session. This acquisition criterion was based on our work conducted in this laboratory on acquisition of cocaine self-administration and consulted other references (Carroll and Lac, 1997, 1998).

2.5. Data analysis

Data are expressed as mean ± S.E.M. Analysis of data was performed using two-way analysis of variance to compared numbers of lever responses and cocaine infusions between isolated and grouped rats. In all statistical tests, a value of $P < 0.05$ was considered to be significant.

3. Results

3.1. Increased lever presses and cocaine infusions in isolated rats

Isolation- or group-reared rats were trained for intravenous cocaine self-administration (3 h/day) at 0.5 mg/kg/infusion under the continuous reinforcement schedule (Fixed-Ratio 1) for 5 days. Numbers of lever presses in isolated and grouped rats for each training day were recorded and the percentages of attaining self-administration criterion for each training day in isolated and grouped rats were calculated. The criterion for acquisition of cocaine self-administration was 21 or more infusions during a self-administration session. The data showed that the percentages of attaining self-administration criterion between isolated and grouped rats for each training day were not significantly different ($F_{9,109} = 1.642$, $P > 0.05$) (Fig. 1), which suggests both isolated and grouped rats could acquire stable cocaine self-administration.

As shown in Fig. 2A, numbers of lever presses in isolated rats were more than those in grouped rats throughout 5 days of testing ($F_{8,57} = 11.646$, $P < 0.001$). Numbers of lever presses in isolated rats were ~100% and ~300% higher than those in grouped rats on the 1st and 2nd day, respectively. Isolated rats maintained stable cocaine self-administration during 3rd–5th

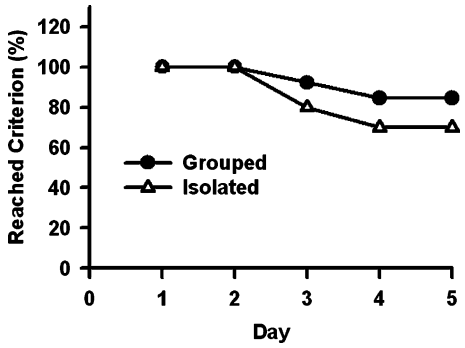

Fig. 1. Both isolated and grouped rats could acquire stable cocaine self-administration. Rats isolation- or group-reared from postnatal day 22 to 55 were trained for intravenous cocaine self-administration (3 h/day) at 0.5 mg/kg/infusion under the continuous reinforcement schedule for 5 days. The criterion for acquisition of cocaine self-administration was 21 or more infusions during a self-administration session. The percentages of attaining self-administration criterion in isolated and grouped rats for each training day were calculated. Data represent the percentages of attaining self-administration criterion in isolated ($n=11$) and grouped rats ($n=13$).

days, but they pressed lever at higher rates as compared with grouped rats. Numbers of cocaine infusions throughout cocaine self-administration session under the continuous reinforcement were shown in Fig. 2B. Numbers of cocaine infusions in isolated rats were ~100% higher than those in grouped rats throughout the 5 training days ($F_{9,55}=9.217$, $P<0.001$). In conclusion, the cocaine self-administration behavior in isolated rats was enhanced as compared with grouped rats throughout the 5 testing days and maintained at relatively lower levels during 3rd–5th days. In addition, numbers of incorrect lever presses during cocaine infusions and time-out phases were shown in Fig. 2C. Numbers of incorrect lever presses in isolated rats were significantly more than those in grouped rats ($F_{9,53}=7.281$, $P<0.001$).

3.2. Reduced intervals of inter-reinforcement for cocaine in isolated rats

Our results showed that intervals of inter-reinforcement for cocaine in isolated rats were shorter than those in grouped rats throughout the 5 testing days. Fig. 3A shows the representative intervals of inter-reinforcement for cocaine in isolated rats at the 1st hour on the 5th training day ($P<0.01$). In addition, isolated rats exhibited rapid burst-like pattern of responding on the lever presses and grouped rats showed temporally dispersed pattern of responding. The increased numbers of cocaine infusions corresponding to the reduced intervals of inter-reinforcement for cocaine in isolated rats suggest the reinforcing properties of cocaine were decreased in isolated rats. The representative response pattern of isolated rats throughout cocaine self-administration sessions of the 1st and 5th day in comparison with grouped rat is presented in Fig. 3B.

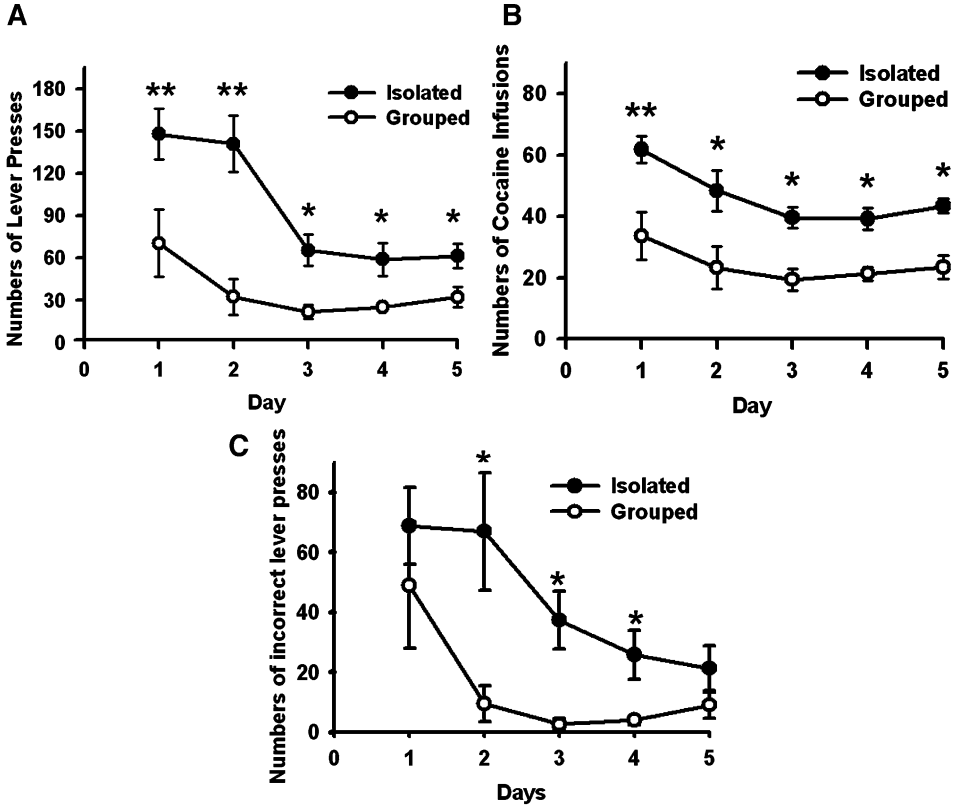

Fig. 2. Increased lever presses and cocaine infusions in isolated rats. Rats isolation- or group-reared from postnatal day 22 to 55 were trained for intravenous cocaine self-administration (3 h/day) at 0.5 mg/kg/infusion under the continuous reinforcement schedule for 5 days. Data represent the mean (±S.E.M.) numbers of lever presses (A), cocaine infusions (B), and incorrect lever presses (C) in isolated ($n=7$) and grouped rats' ($n=11$) attained criterion throughout 5 training days. *$P<0.05$, **$P<0.01$.

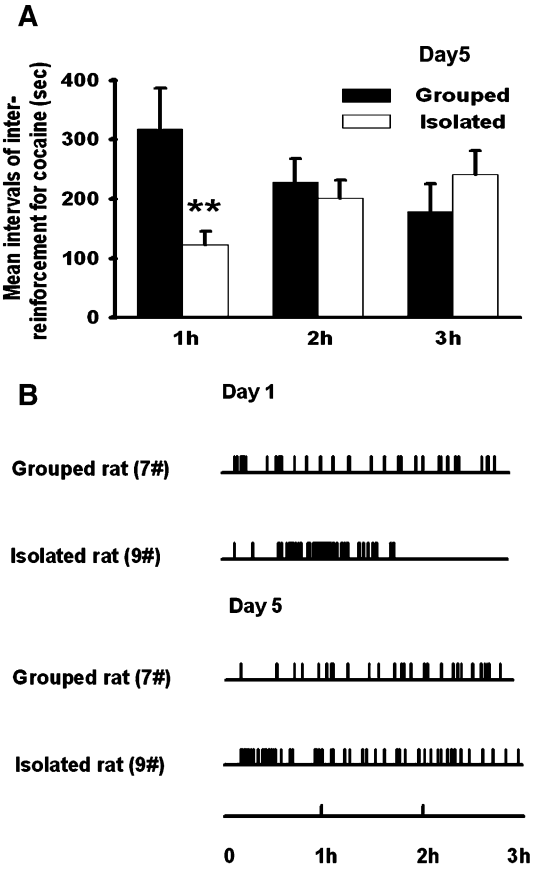

Fig. 3. Reduced intervals of inter-reinforcement for cocaine in isolated rats. Rats isolation- or group-reared from postnatal day 22 to 55 were trained for intravenous cocaine self-administration (3 h/day) at 0.5 mg/kg/infusion under the continuous reinforcement schedule for 5 days. Data represent the mean (±S.E.M.) intervals of inter-reinforcement for cocaine in isolated rats and grouped rats during the 1st, 2nd and 3rd hour on the 5th day of testing (A). $^{**}P<0.01$. (B) shows the response patterns of one representative isolated rat (9#) and one representative grouped rat (7#) throughout cocaine self-administration session (3 h) of the 1st and 5th day. Each mark represents an infusion of cocaine.

3.3. Decreased ratios of cocaine infusions versus lever presses in isolated rats

The difference of ratios of cocaine infusions versus lever presses between isolated and grouped rats is shown in Fig. 4. The ratios of cocaine infusions versus lever presses in isolated rats were significantly lower than those in grouped rats during the first 2 days ($F_{9,52}=3.668$, $P<0.001$), but ultimately, accorded with grouped rats on the 5th day.

4. Discussion

The present study assessed the effect of adolescent isolation on intravenous cocaine self-administration under the continuous reinforcement schedule. Our results showed lever presses and cocaine infusions as well as incorrect lever presses in isolated rats were significantly more than those in grouped rats. In addition, the intervals of inter-reinforcement for cocaine in isolated rats were significantly shorter than those in grouped rats.

The results indicate adolescent isolated rats have enhanced cocaine self-administration behavior, which suggests adolescent isolation may cause greater impulsivity or motivation for cocaine in adult rats (Schenk et al., 1987). Whereas, the cocaine self-administration behavior in grouped and isolated rats were higher during 1st–2nd testing days and maintained at relatively lower levels during 3rd–5th days. The motivation for food at the extinction of food reward in the initial two days of drug training may contribute to the higher numbers of lever presses. In addition, the intervals of inter-reinforcement for cocaine were significantly shorter in isolated rats, corresponding to the increased cocaine infusions, suggesting that adolescent isolation may decrease the reinforcing properties of cocaine. This is in agreement with the previous reports on the effect of adolescent isolation on morphine (Wongwitdecha and Marsden, 1996), cocaine (Berry and Marsden, 1994; Schenk et al., 1986), amphetamine (Wongwitdecha and Marsden, 1995), and heroin (Schenk et al., 1986) using a conditioned place preference test and other studies using cocaine self-administration (Phillips et al., 1994). Previous studies demonstrated that neonatal isolated rats could acquire cocaine self-administration at low and modest doses (0.125–0.25 mg/kg/infusion) and grouped rats could only acquire at a higher dose (0.5 mg/kg/infusion), which suggests neonatal isolation increases the reinforcing properties of cocaine and results in the enhancement of the propensity to self-administer cocaine in neonatal isolated rats as compared with grouped rats (Matthews et al., 1999; Kosten et al., 2000; Zhang et al., 2005). Our study suggests that the effect of adolescent isolation on the reinforcing properties of cocaine may differ from that of neonatal isolation, although the underlining mechanisms are to be understood.

In addition, our results showed incorrect lever presses in isolated rats were significantly more than those in grouped rats. This could be a result of greater impulsivity and motivation in isolated rat, but other factors such as impaired learning and memory or hyperactivity in these rats may also contribute to it.

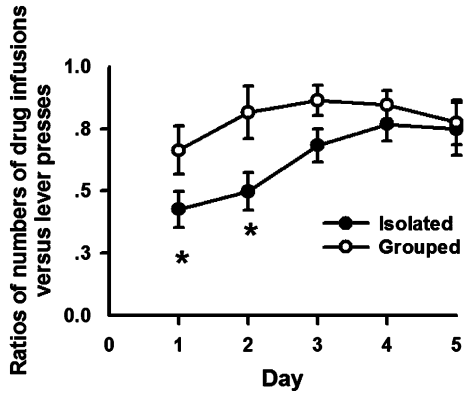

Fig. 4. Decreased ratios of cocaine infusions versus lever presses in isolated rats. Rats isolation- or group-reared from postnatal day 22 to 55 were trained for intravenous cocaine self-administration (3 h/day) at 0.5 mg/kg/infusion under the continuous reinforcement schedule for 5 days. Data represent the mean (±S.E.M.) ratios of numbers of cocaine infusions versus lever responses in isolated ($n=7$) and grouped rats ($n=11$) throughout 5 training days. $^{*}P<0.05$.

Both our and other studies demonstrated that adolescent isolation could induce neurogenesis and neurotransmission modification in brain regions such as hippocampus, frontal cortex, and amygdala (Lapiz et al., 2003; Nilsson et al., 1999; Varty et al., 1999; Lu et al., 2003). Adolescent isolation could result reduced long-term potentiation, a neural model of learning and memory (Lu et al., 2003). Previous studies demonstrated that rats reared under the condition of adolescent isolation have impairment of learning and memory (Nilsson et al., 1999; Varty et al., 1999; Lu et al., 2003). In our study, incorrect lever presses in isolated rats progressively decreased during 1st–4th days and approached to the levels of grouped rats on the 5th day, which is consistent with the previous observation that isolated rats possess lower learning and memory capability and suggests that impaired learning and memory capability may be also a possible cause of the increased incorrect lever presses in isolated rats.

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