Anti-craving effects of environmental enrichment

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Abstract

We hypothesized that environmental enrichment in rats may reduce cocaine-seeking behaviour elicited by cocaine-priming injections and by cocaine-associated cues. Rats trained to self-administer cocaine while housed in isolated conditions were then assigned to live in isolation or an enriched environment for 21 d of forced abstinence. Subsequently, extinction and reinstatement of cocaine-seeking behaviour (operant responses without cocaine available) were assessed. Expt 1 showed that enrichment resulted in less cocaine-seeking behaviour during extinction and cue-elicited reinstatement compared to continued isolation housing, but had no effect on cocaine-primed reinstatement. A subsequent experiment, which included a pair-housed group to control for potential isolation stress, again demonstrated that enrichment attenuated cocaine seeking during extinction, but not cocaine-primed reinstatement, relative to both isolation and pair-housed conditions. The findings suggest that enrichment reduces the impact of cocaine-associated environmental stimuli, and hence it may be a useful intervention for attenuating cue-elicited craving in humans.

Keywords
Cocaine; drug-seeking behaviour; environmental enrichment; reinstatement; self-administration

Introduction

Cocaine addiction is difficult to treat because relapse often occurs even after long periods of abstinence (Dackis & O'Brien, 2001). A major factor contributing to relapse is craving, which can be elicited either by sampling cocaine or exposure to environmental cues associated with cocaine (Ehrman et al. 1992). Therapies to extinguish cue-elicited craving have been unsuccessful overall in preventing relapse (Conklin & Tiffany, 2002). An alternative treatment may be environmental enrichment to promote physical and emotional well-being such that individuals can better resist cocaine and cope with craving elicited by drug-associated cues.

Preclinically, environmental enrichment refers to living conditions in which animals are housed in large cages with access to social stimulation, novel objects, and exercise (van Praag et al. 2000). Environmental enrichment enhances brain function (Laviola et al. 2008; van Praag et al. 2000) and attenuates addiction-relevant processes including drug reward and reinforcement, and drug-seeking behaviour (Green et al. 2002; Howes et al. 2000; Stairs et al. 2006; Xu et al. 2007). Two recent studies suggest that environmental enrichment may reduce responses to reward-associated stimuli. Solinas et al. (2008) used environmental enrichment to block
expression of a previously acquired cocaine-conditioned place preference; Grimm et al. (2008) used environmental enrichment to block cue-elicited sucrose-seeking behaviour.

This study examined whether environmental enrichment introduced during forced abstinence after cocaine self-administration can reduce motivation to seek cocaine. The extinction/reinstatement model was used to measure cocaine-seeking behaviour elicited by response-contingent cues or cocaine priming.

Method

Animals and surgery

Adult male Sprague–Dawley rats weighing 225–250 g upon arrival were housed under standard isolated conditions of 1 rat/cage (21.6 × 45.7 × 17.8 cm) with only food and water available in a colony with a 12-h reversed light/dark cycle (lights on 19:00 hours). Care and housing were in adherence to the Guide for the Care and Use of Laboratory Animals (1996). Rats were habituated to handling for 5 d prior to surgically implanting intravenous (i.v.) catheters under 2–3% isoflurane anaesthesia using procedures described previously (Zavala et al. 2007).

Expt 1

Self-administration—All rats remained housed in isolated conditions throughout self-administration training. After recovery from surgery, rats underwent 15 d cocaine self-administration training for 3 h/d during their dark cycle. Initially, sessions began with an FR1 schedule of reinforcement and progressed to a VR5 schedule based on individual performance, with the latter in effect exclusively during the last 5–8 sessions. Schedule completions on a designated lever (i.e. active lever) resulted in simultaneous presentation of a tone (500 Hz, 10 dB above background), cue light above the lever, and house light, followed 1 s later by a cocaine infusion (0.75 mg/kg per 0.1 ml i.v.). Upon completion of the 6-s infusion, the cue light and tone ceased, but the house light remained on for an additional 20-s time-out. Responses on another lever (i.e. inactive lever) produced no consequences. Rats were restricted to 16 g food/d beginning 2 d before training to facilitate exploration. A rat remained food-restricted until a criterion of ≥21 infusions/3 h was achieved on two consecutive days, after which food was available ad libitum in the home cage throughout the remainder of the experiment. All rats had reached this criterion by the 10th session. One rat was omitted from the analyses due to catheter failure but was later used as a social partner in environmental enrichment.

Abstinence—The day after completing self-administration, rats were assigned to one of two housing conditions, counterbalanced for previous cocaine intake: Isolated (n = 9) or Enriched (n = 9). Enriched animals lived in large plastic tubs (74 × 91 × 36 cm) that housed five rats and contained bedding, nesting material, three PVC pipes, two running wheels, two water bottles, two food dishes, and two small plastic toys. Toys were changed 3 times/wk to maintain novelty. All rats were habituated to intraperitoneal (i.p.) injections by receiving saline (1 ml/kg i.p.) on each of the 2 d prior to testing.

Test phases—After 21 d of abstinence, testing occurred across three consecutive phases: (1) extinction, (2) cue reinstatement, and (3) cocaine reinstatement. The extinction phase began immediately after the rats were returned to the self-administration chambers. Responses on the active lever were recorded but produced no scheduled consequences. After 2 h, the cue reinstatement phase began with a programmed presentation of the stimulus complex previously paired with cocaine. Thereafter, cues were presented after each active lever press for 2 h. Following the cue reinstatement phase, rats were given a cocaine injection (10 mg/kg i.p.), and then returned to the chamber. Subsequent non-reinforced lever presses were recorded for 1 h.
Expt 2

The procedures used for this experiment were identical to those used in expt 1, with the exception that animals were assigned to one of three housing conditions during abstinence, counterbalanced for previous cocaine intake: Isolated ($n = 9$), Pair-housed ($n = 9$), or Enriched ($n = 9$). The Pair-housed condition was identical to the Isolated condition except that there were two rats/cage. Two rats were omitted from the analyses due to catheter failure but were used for the living conditions during abstinence. Testing occurred across three consecutive phases: (1) extinction, (2) saline-primed reinstatement, and (3) cocaine-primed reinstatement. The extinction phase was identical to that used in expt 1. After 2 h, rats were removed and given a saline injection (1 ml/kg i.p.) in order to examine reinstatement from injection stress. Subsequent non-reinforced lever presses were recorded for 1 h. Rats were again removed and given a cocaine-priming injection (10 mg/kg i.p.), and subsequent non-reinforced lever presses were recorded for 2 h.

Data analysis

Cocaine-seeking behaviour was operationally defined as active lever responses in the absence of cocaine reinforcement. Separate mixed-factorial ANOVAs were used to analyse active and inactive lever responses during each test phase and at the transition from one phase to the next with abstinence living condition as a between-subjects factor and 20-min time-interval as a repeated measure. Total active lever presses for each test phase were analysed using $t$ tests (expt 1) or one-way ANOVAs (expt 2). Significant interactions were further analysed using Newman–Keuls tests.

Results

Expt 1

Cocaine intake—Group assignment was counterbalanced such that there were no differences in cocaine intake across Isolated and Enriched groups. For the last five self-administration sessions, the Isolated and Enriched groups, respectively, had a mean (± S.E.M.) cocaine infusions/session of 44.3 ± 3.4 and 42.5 ± 3.7 and a mean (± S.E.M.) active lever presses of 240.6 ± 38.5 and 237.8 ± 20.9.

Cocaine-seeking behaviour—Cocaine-seeking behaviour across time, as well as in total for each of the test phases, is illustrated in Fig. 1(a, b), respectively. Cocaine-seeking behaviour was reduced in the Enriched group relative to the Isolated group during extinction, which was most evident in the first hour of testing. There was a living condition × time interaction [$F(5, 80) = 3.82, p<0.01$] for active lever presses and post-hoc analyses indicated more cocaine-seeking behaviour in the Isolated vs. Enriched group during intervals 1–3 ($p<0.05$, Newman-Keuls; Fig. 1a). Inactive lever presses also decreased across time more rapidly for the Enriched group (i.e. interval 2) than for the Isolated group [living condition × time interaction: $F(5, 80) = 2.87, p<0.01$, followed by Newman–Keuls tests, $p<0.05$; Fig. 1a]. Total cocaine-seeking behaviour during the extinction phase was reduced in the Enriched vs. Isolated group [$t(16) = 3.98, p<0.001$; Fig. 1b].

Although both the Isolated and Enriched groups demonstrated cue-elicited reinstatement of cocaine-seeking behaviour, the magnitude of reinstatement was reduced in the Enriched group relative to the Isolated group. Analyses of cocaine-seeking behaviour during the transition from the last 20 min of extinction to the first 20 min of cue reinstatement, as well as across time during the cue reinstatement phase, revealed living condition × time interactions [$F(1, 16) = 12.93, p<0.001$ and $F(5, 80) = 7.28, p<0.001$, respectively]. Post-hoc analyses indicated more cocaine-seeking behaviour in the Isolated vs. Enriched group during intervals 1–2 (Newman–Keuls, $p<0.05$; Fig. 1a). There was no effect of living condition for inactive lever presses during
this phase. Total cocaine-seeking behaviour during cue reinstatement was also greater for the Isolated vs. Enriched group \( t(16) = 3.63, p<0.001; \) Fig. 1b.

Rats in both the Isolated and Enriched groups demonstrated similar magnitudes of cocaine-primed reinstatement during the transition to this test phase [main effect of time only: \( F(1, 16) = 18.90, p<0.001; \) Fig. 1a], as well as throughout the test phase [main effect of time only: \( F(2, 32) = 5.47, p<0.01; \) Fig. 1a]. There were no group differences in inactive lever presses or total cocaine-seeking behaviour across this phase (Fig. 1b).

**Expt 2**

**Cocaine intake**—For the last 5 d of self-administration, mean (± s.e.m.) cocaine infusions/session for the Isolated, Pair-housed, and Enriched groups, respectively, were 38.4 ± 3.2, 38.1 ± 1.8, and 37.9 ± 2.9 and active lever presses were 228.2 ± 49.9, 229.7 ± 17.3, and 229.8 ± 32.9.

**Cocaine-seeking behaviour**—Cocaine-seeking behaviour across time, as well as in total for each of the test phases, is illustrated in Fig. 2(a, b), respectively. All groups exhibited cocaine-seeking behaviour after being placed in the self-administration context, which gradually extinguished by the end of the extinction phase. There was a living condition × time interaction \( [F(10,120) = 8.04, p<0.001] \) for active lever presses during this phase. The Isolated group exhibited more cocaine-seeking behaviour than the Enriched group during intervals 1–2 (\( p<0.05, \) Newman–Keuls; Fig. 2a). The Pair-housed group also exhibited more cocaine-seeking behaviour than the Enriched group during interval 1 (\( p<0.05, \) Newman–Keuls; Fig. 2a). Inactive lever pressing decreased across this phase at a similar rate for all groups [main effect of time: \( F(5, 120) = 3.54, p<0.01; \) Fig. 2a]. Total cocaine-seeking behaviour during the extinction phase differed depending on living condition \( [F(2, 24) = 34.49, p<0.001] \), with Isolated and Pair-housed groups higher than the Enriched group (\( p<0.05, \) Newman–Keuls; Fig. 2b).

Across the saline reinstatement phase, there were no group differences in active or inactive lever pressing (Fig. 2a, b). All groups demonstrated cocaine-primed reinstatement of cocaine-seeking behaviour during the transition to this test phase [main effect of time: \( F(1, 24) = 46.56, p<0.001; \) Fig. 2a] that decreased at similar rates for all groups. There was no living condition × time interaction. There were also no group differences in inactive lever presses or in total cocaine-seeking behaviour (Fig. 2b).

**Discussion**

The findings are consistent with the hypothesis that enrichment during abstinence attenuates cocaine-seeking behaviour after exposure to the self-administration environment during extinction, as well as during discrete cue-induced reinstatement of cocaine-seeking behaviour. Our results are in agreement with Solinas et al. (2008) and Grimm et al. (2008), who demonstrated that environmental enrichment introduced as an intervention strategy attenuates expression of cocaine-conditioned place preference and sucrose reinforcement. Our results are also in agreement with Stairs et al. (2006) who demonstrated that environmental enrichment attenuates context/cue-elicited drug-seeking behaviour relative to isolation housing.

Expt 1 results suggest that environmental enrichment had no effect on cocaine-primed reinstatement. Expt 2 confirmed these results in rats that did not undergo cue reinstatement prior to cocaine-primed reinstatement. We had expected environmental enrichment would decrease cocaine-primed reinstatement given that Stairs et al. (2006) found this manipulation blunts amphetamine-primed reinstatement. There were many differences between the present study and that of Stairs et al. that may have contributed to the different effects observed with
enrichment. We suggest the most critical difference is that Stairs et al.’s Enriched group were reared and maintained under enrichment throughout the experiment. In contrast, the Enriched group in the present study were isolated prior to and throughout training, and were introduced to environmental enrichment only during forced abstinence. It is likely that training history and timing of environmental enrichment influence its impact on drug-seeking behaviour elicited by priming or drug-associated stimuli. The benefit of environmental enrichment observed in the present study is particularly relevant to the clinical use of enrichment as an anti-relapse treatment, which would begin during abstinence.

The Pair-housed group was included in expt 2 to determine whether the group differences in context-elicited cocaine-seeking behaviour observed during extinction in expt 1 were due to an isolation stress-induced increase or an enrichment-induced decrease in the motivational impact of the contextual stimuli associated with cocaine. Expt 2 results demonstrated that the Enriched group exhibited less context-elicited cocaine-seeking behaviour during extinction relative to both the Isolated and Pair-housed groups. These findings suggest that environmental enrichment may protect against craving elicited by the drug-associated context.

There are several possible mechanisms by which environmental enrichment may attenuate context/cue-elicited cocaine-seeking behaviour, including reducing either impulsivity (Perry et al. 2008; Wood et al. 2006), negative affect or stress (Belz et al. 2003), and/or behavioural sensitization resulting from repeated drug exposure (Bardo et al. 1995). Indeed, impulsivity is considered an important factor in vulnerability to drug use and relapse (de Wit & Richards, 2004). Furthermore, stress and negative affect can elicit cocaine relapse in humans (Sinha, 2001) as well as reinstatement of drug-seeking behaviour in rodents (Koob & Le Moal, 2008). Therefore, manipulations that blunt responses to stress and anxiety during abstinence may aid in preventing relapse. To this end, environmental enrichment promotes overall emotional stability (Mohammed et al. 1993), is rewarding to animals (van der Harst et al. 2003), and attenuates stress-related responses (Belz et al. 2003; Benaroya-Milshtein et al. 2004).

In regard to sensitization, this phenomenon is related to incentive motivational effects of drug and drug-associated cues (Robinson & Berridge, 2008). Stairs et al. (2006) suggest that the attenuation of extinction responding that they observed with environmental enrichment may reflect attenuated sensitivity to the conditioned reinforcing effects of drug-paired cues. They reasoned that environmental enrichment housing during abstinence may attenuate the incentive value of amphetamine, in turn reducing the incentive value of drug-conditioned stimuli. Indeed, Stairs et al. (2006) demonstrated that environmental enrichment attenuated reinstatement at a threshold amphetamine priming dose, which they suggest reflects enrichment-induced reversal of sensitization to the incentive motivational effects of amphetamine. The priming dose of cocaine used in the present study may not have been sensitive enough to detect enrichment-induced attenuation of cocaine-primed reinstatement. Studies examining lower doses of cocaine priming will be necessary to conclusively determine whether enrichment decreases the potency of cocaine priming. Regardless, the findings that moderate doses of amphetamine or cocaine restate drug-seeking behaviour in Enriched animals suggests that clinically, enrichment may not be an effective treatment for countering craving elicited by sampling drug.

The present findings offer strong preclinical support for the idea that environmental enrichment may be a useful intervention for reducing craving elicited by cocaine-associated environmental stimuli. Reducing cue-elicited craving with enrichment in humans should be a useful adjunct to treatments aimed at preventing relapse. An important characteristic of drug-seeking behaviour during abstinence is that it increases, or ‘incubates’, over time (Grimm et al. 2001; Tran-Nguyen et al. 1998). We plan to examine whether environmental enrichment across various durations of abstinence can be used to block this incubation effect.
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References


Fig. 1.
Extinction, cue reinstatement, and cocaine reinstatement for expt 1. (a) Cocaine-seeking behaviour illustrated as active lever presses (±S.E.M.) across 20-min time-intervals; inactive lever presses are illustrated below for comparison. (b) Total cocaine-seeking behaviour (i.e. active lever presses ± S.E.M.) collapsed across the time-intervals for each test phase (i.e. 2 h extinction, 2 h cue reinstatement, and 1 h cocaine reinstatement). The extinction phase began when rats were placed into the self-administration chambers. The cue reinstatement phase began 2 h later by presenting the light/tone cues previously associated with cocaine infusions, and thereafter response-contingent cues were presented on a FR1 schedule. The cocaine reinstatement phase followed 2 h later and began with administration of a priming injection of cocaine (10 mg/kg...
i.p.). Active lever responses during extinction and cocaine-primed reinstatement produced no consequences. * Difference from Isolated group ($p<0.05$); † difference from previous 20-min interval (time main effect, $p<0.05$).
Fig. 2.
Extinction, saline reinstatement, and cocaine reinstatement for expt 2. (a) Cocaine-seeking behaviour illustrated as active lever presses (± SEM) across 20-min intervals; inactive lever presses are illustrated below for comparison. (b) Total cocaine-seeking behaviour (i.e. active lever presses ± SEM) collapsed across time for each test phase (i.e. 2 h extinction, 1 h saline reinstatement, and 2 h cocaine reinstatement). The extinction phase began when rats were placed into the self-administration chambers. The saline reinstatement phase began 2 h later with administration of a saline priming injection (1 ml/kg). The cocaine reinstatement phase followed 1 h later and began with the administration of a priming injection of cocaine (10 mg/kg i.p.). Active lever responses during all phases produced no consequences. * Difference from
Isolated group ($p<0.05$); § difference from Pair-housed group ($p<0.05$); † difference from previous 20-min interval (time main effect, $p<0.05$).