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Intermittent social stress produces different short- and long-term effects on effort-based reward-seeking behavior.

Christopher Lemon¹, Alberto Del Arco^{1,2,3}

¹HSERM, School of Applied Sciences, University of Mississippi, Oxford, MS

²Neurobiology and Anatomical Sciences, Medical School, University of Mississippi Medical Center, Jackson, MS

³Department of Biomolecular Sciences, School of Pharmacy, University of Mississippi, Oxford, MS

Abstract

Previous studies show that intermittent social defeat (ISD) stress increases self-administration of psychostimulants, which suggests that ISD promotes reward-seeking behavior and, ultimately, increases vulnerability to develop drug abuse. The present study investigates whether ISD alters cost/benefit evaluations to promote reward-seeking behavior and whether these alterations are time-dependent. Male rats performed two different tasks that assessed their motivation to seek and consume food rewards. An effort-discounting task in which rats chose between less and more effortful options (i.e., 1 lever-press versus 2, 5, 10 or 20 lever-presses) associated with low- and high-reward (i.e., 1 sugar pellet versus 3 sugar pellets), respectively; and a progressive ratio task in which rats had to increase their effort (more lever presses) to obtain a sugar pellet. ISD consisted of exposing animals to social defeat once every three days for ten days (4 stress episodes). Rats were tested 24-48 h after stress episodes, and 1 week and 6 weeks after the last stress episode. In the effort-discounting task, stressed animals showed a decrease in their preference for high rewards associated with more effort (i.e., 10 and 20 lever-presses). These effects were transient and not maintained one week after stress. In the progressive ratio task, stressed animals showed an increase in the number of lever presses to obtain rewards that emerged six weeks after the last stress episode. These results suggest different short- and long-term effects on the motivation for rewards after ISD and indicate temporal dynamic adaptations in the function of the brain reward system.

Keywords

social stress; motivation; decision-making; effort; reward-seeking

Correspondence author: Alberto Del Arco, Ph.D., 215 Turner Center, P.O. Box 1848, University, MS 38677, USA, adelarco@olemiss.edu.

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INTRODUCTION

The repeated exposure to social adversity increases vulnerability to develop substance use disorders [1]. Studies in humans and animals show that stressful experiences increase drug-seeking behavior and relapse [1-4]. In particular, intermittent (episodic) social defeat (ISD) stress in rats has been consistently shown to increase drug self-administration (i.e., psychostimulants) [5-7] as well as the activity of the mesolimbic dopamine system [5,8], which suggests that ISD enhances the motivation to seek rewards. However, how the intermittent exposure to social stress changes reward processing is unclear [9] and therefore, how ISD increases reward-seeking behavior is poorly understood. Pharmacological studies show that the activation of the mesolimbic dopamine system alters cost/benefit evaluations and increases the motivation to pursue food as well as drug rewards [10-15]. Based on these studies, the present study investigates whether ISD alters cost/benefit evaluations in order to promote reward-seeking behavior. Clarifying this will help understand the transition from social adversity to drug abuse.

Previous studies assessing the effects of stress on cost/benefit decision-making focused on acute and chronic stress [16]. Thus, by using an effort-discounting task, it has been shown that acute restraint stress decreases the preference for high magnitude rewards when animals are required to put more effort (i.e. more lever presses) to earn them [17,18]. Similarly, by using a progressive ratio schedule, studies have shown that acute restraint and chronic social stress [6,19,20] as well as chronic corticosterone administration [21,22], decrease the motivation to work for rewards, as they reduce the number of lever presses that animals are willing to make to obtain rewards. The effects of chronic social stress have been associated with depression-like behaviors and anhedonia [23-26]. Overall, these studies suggest that stress alters effort-based reward seeking and decreases the motivation to work for rewards. It is important to emphasize, however, that the effects of repeated social stress on motivation depend critically on the stress protocol utilized –continuous (i.e., every day) versus intermittent exposure to social defeat [6]–, and therefore, these studies do not provide evidence of whether ISD changes effort-based reward-seeking behavior.

Importantly, the effects of repeated social stress on motivation and reward seeking seem to depend on the time when these effects are evaluated [25-29]. In fact, it has been shown that the alterations in reward processing (i.e., anhedonia) produced by chronic social defeat are transient in some animals (i.e., resilient to social defeat) and not maintained after the last stress episode [25,26]. Moreover, one study shows that chronic social defeat increased the motivation for rewards ten weeks after stress [29], while the opposite was found when the effects of social defeat were evaluated immediately or up to three weeks after stress [6,26]. These studies point to long-term changes in the function of the brain reward system during the post-stress period and open the possibility that ISD produces time-dependent effects on effort-based reward-seeking behavior [30].

The present study investigates whether ISD alters cost/benefit decision-making and the motivation to pursue rewards (i.e. sugar pellets) and whether these alterations are time dependent. In the current study we focus on the intermittent exposure to social defeat (i.e., ISD) versus other social defeat protocols (i.e., chronic social defeat) because ISD

consistently increases reward seeking (i.e., drug-self administration) [5-7], while the opposite is found (i.e., depressive-like anhedonia) by most studies utilizing chronic social defeat [6,26,31,32]. ISD is also an optimal protocol for investigating time-dependent and cumulative effects of stress since animals can be tested on the days in between stress episodes as well as weeks after stress. We trained animals in an effort-discounting task [18] and then exposed them to ISD as in our previous work [30]. In this task, animals choose between high- and low-reward choices based on the effort (i.e., number of lever presses) to obtain the high-reward option. The terms low-reward and high-reward refer to the magnitude of rewards (i.e., one sugar pellet versus three sugar pellets). Food deprived rats value higher magnitude rewards more than lower magnitude. Additionally, we tested animals in a progressive ratio task [18] to evaluate whether they were more motivated to work (i.e. press lever more times) for rewards. To assess the temporal profile of the effects of ISD on behavior, we tested animals' performance in both tasks in between social stress episodes and one and six weeks after the last stress episode. This timeline is based on previous studies showing that ISD increases drug self-administration five weeks after the last stress episode [5,33].

2. METHODS

2.1 Animals

Sixteen adult male Long Evans rats (Envigo, Indianapolis, IN) (375–425 g) were pair-housed in standard polycarbonate cages (45 x 24 x 20 cm) on a 12 h light/dark cycle (lights on at 9:00 P.M.). All experiments were performed during the dark phase when the animals are most active. The rats were placed on a mild food-restricted diet (15 g of chow per rat and day) two days before starting behavioral experiments [30]. This feeding regime –extended throughout all phases of the experiment–, resulted in a 7-10% weight loss for both groups of rats (basal: Control= 528 ±11, Stress= 540 ±14; pre-ISD: Control= 483 ±11, Stress= 503 ±14; post-ISD: Control= 477 ±11, Stress= 500 ±10). All procedures were approved by the University of Mississippi Institutional Animal Review Board and were conducted in accordance with the National Institute of Health Guide for the Care and Use of Laboratory Animals.

2.2 Experimental design

Two-three weeks after their arrival to the animal facility, all rats were handled for at least three days and then habituated to the operant chambers and trained in the effort-discounting task. Once animals reached stable performance (see below), they were divided in two groups: control (n= 8) and stress (n= 8). The stress group was exposed to ISD following the protocol used in our previous study [30] and depicted in Figure 1A. The control group was taken to a different room and handled for 5 min. Rats were tested in the effort-discounting task 24-48 h after stress episodes and one week after the end of the stress protocol. Rats were tested in the progressive ratio task 48 h after the second and fourth stress episodes, and one and six weeks after the end of the stress protocol.

2.3 Effort-discounting task

We utilized an effort-discounting task modified from Bryce and Floresco [18] (Figure 1B). The apparatus consisted of an operant chamber with two retractable levers (right and left) and a food trough in between them in the same wall. The chamber contained a house light that was on during the entire session. First, animals were trained to establish the lever–response contingency through a reward-shaping procedure using both levers (3-4 days) (dustless sugar pellets, 45mg; Bio-Serv). Second, they were trained to discriminate between a high-reward lever (HR) and a low-reward lever (LR) (FR1, 30 min sessions). Pressing the HR delivered 3 sugar pellets while pressing the LR delivered 1 sugar pellet. This training phase lasted until they pressed the LR <25% of the trials (5-6 days). Then, the effort-discounting training started. The task consisted of 4 blocks of 12 trials each (48 trials total, 32 min sessions). In each block, the first 2 trials were *force choice* trials in which only one of the two levers was randomly extended. The right lever was associated with a low reward (1 sugar pellet) and the left lever was associated with a high reward (3 sugar pellets). The value of the levers varied for each animal. The remaining 10 trials were *free choice* trials in which both levers were extended, and animals were required to make a choice. For all sessions and blocks, one lever was designated as the LR, and the other lever was designated as the HR. This designation was counterbalanced among animals. Every trial had a duration of 40 s and started with a variable intertrial interval (25-35 s) after which the levers were extended for a maximum of 10 s. If the animals press the LR, both levers were retracted, and a single food pellet was delivered to the food trough after 1 s. The light of the food trough was turned on then. If the animals pressed the HR only the LR retracted, and the HR remained extended until the animals made the required number of presses to obtain 3 food pellets. If animals did not press any lever within the 10 s both levers were retracted, and the trial was considered an omission. Similarly, if the animals did not perform the required number of presses, the lever retracted, and no pellet was delivered. The number of presses required to earn rewards (ratio) increased over blocks, initially set at 2 presses, and increasing to 5, 10, and 20 presses for subsequent blocks. Animals were trained until stable performance (3 consecutive sessions choosing the HR lever at least 80% of trials during the first block), which typically took 5-7 days. Choice latency (the time between lever extension and choice) and rates of pressing on the HR lever were also evaluated.

2.4 Progressive ratio schedule

Control and stressed rats were tested in a progressive ratio schedule to assess changes in the motivation to work for rewards (Figure 1C). In the progressive ratio the number of presses required to obtain a pellet increased exponentially. The ratio was modified from previous work [18] and increased in the following manner: 2, 5, 10, 20, 31, 44, 56, 78 and 100 presses. Rats had a maximum of 10 min to complete each ratio and obtain rewards. The sessions ended when rats failed to complete a ratio. The total number of lever presses and pellets obtained over the course of a session and the last ratio obtained before a session terminated (breakpoint) were measured.

2.5 Intermittent social defeat stress (ISD)

After stable performance in the effort-discounting task, rats were exposed to repeated social defeat stress [30,34] (Resident-Intruder paradigm, modified from Miczek et al., [6]). Rats were housed individually one week before starting the stress protocol. Resident males (Long Evans retired breeders; 500-600 g) were housed in transparent PVC cages (H x L x W: 45 x 61 x 61 cm) with females for at least 10 days before starting the procedure. Female rats were previously sterilized by ligation of the oviducts. Animals of the stress group were exposed to social defeat once every three days for ten days. Control animals were moved to a new room and handled for 5 min. Every social stress session started by removing the female rat from the resident cage at least 30 min before. Then, first, the intruder rat was placed in the cage with the resident male separated by a divider wall that contained wire mesh (allowing sensory exposure) –both rats could see and smell each other–, for 10 minutes. Second, the divider wall was removed, allowing the rats to interact. The interaction was stopped when either 6 attacks were witnessed, the intruder was in supine position for 5 seconds, or 5 minutes had elapsed. To avoid injuries, the interaction was also stopped if an aggressive bite occurred. The latency to the first attack was also recorded. Third, the divider wall was reinserted, and the intruder remained in the cage for 10 more minutes. After this time, intruders returned to their home cages. Intruders were not exposed to the same resident more than twice.

2.6 Data analysis

A three-way ANOVA with repeated measures was utilized to analyze performance in the effort-discounting and progressive ratio tasks. This analysis considered group (Control and Stress) as between subjects and dependent variables measured in the effort-discounting task (i.e., number of choices for high reward lever, choice latency, rate of lever presses) and the progressive ratio task (i.e., lever presses, pellets consumed, break point) as within subjects; and focused on the cumulative (i.e., after the second stress episode) and time-dependent (i.e., across sessions) effects of ISD. One- and Two- way ANOVAs with repeated measures as well as Student *t* tests were also utilized for further analysis. The statistical computations were performed with SPSS software and the statistical significance was set at $p = 0.05$.

RESULTS

Effects of ISD on the effort-discounting task

The rats were tested in the effort-discounting task during the days in between stress episodes and one week after the last stress episode (see Figure 1A). A three-way ANOVA with repeated measures with group (Control, Stress) as between subjects and ratio (2, 5, 10 and 20) and session (S2-Post 1Week) as within subjects was performed to compare the choice of the more effortful option (choice HR lever) as well as choice latency and rate of lever presses. As shown in Figure 2A, animals decreased their preference for the HR lever when they were required to put more effort (i.e., press the lever more times) to receive a food pellet (ratio effect: $F_{(3,42)} = 21.45$, $p < 0.001$, $\eta_p^2 = 0.61$). The three-way ANOVA did not detect overall significant effects across sessions ($F_{(3,42)} = 2.34$, $p = 0.082$, $\eta_p^2 = 0.14$). However, it did reveal a significant session x group interaction ($F_{(3,42)} = 3.44$, $p = 0.025$,

$\eta_p^2 = 0.20$) indicating that the effects of stress on the choice HR lever were different across sessions.

We performed two-way ANOVAs to further assess how stress changed the preference for the HR lever across sessions for every ratio (Figure 3). We found that stress decreased the preference for HR compared to controls at the 10 presses ratio (session x group interaction: $F_{(3,42)} = 3.40$, $p = 0.026$, $\eta_p^2 = 0.20$). This effect was significant after the third stress episode (S3) ($t_{14} = 2.75$, $p = 0.016$, $\eta_p^2 = 0.35$; independent t test) (Figures 2A and 3). We also found that the preference for HR changed across sessions at the 20 presses ratio (session effect: $F_{(3,42)} = 4.79$, $p = 0.006$, $\eta_p^2 = 0.25$). This effect was significant for stressed animals ($F_{(3,21)} = 6.47$, $p = 0.003$, $\eta_p^2 = 0.48$; one-way ANOVA with repeated measures), but not controls ($F_{(3,21)} = 0.94$, $p = 0.44$, $\eta_p^2 = 0.12$; one-way ANOVA with repeated measures), which indicates a decrease in the preference for HR across sessions in the Stress group. As shown in Figure 3, this decrease was lowest after the last stress episode (S4) compared to the previous sessions S3 ($t_7 = 2.34$, $p = 0.050$) and S2 ($t_7 = 3.26$, $p = 0.014$; paired t test). This effect was not maintained 1 week after the last stress episode since HR choices increased significantly at 1 week compared to S4 ($t_7 = 2.55$, $p = 0.038$; paired t test). The two-way ANOVA did not find group differences for HR preference across sessions at the 20 presses ratio (session x group interaction: $F_{(3,42)} = 2.13$, $p = 0.110$, $\eta_p^2 = 0.13$).

Figure 2B shows the choice latency to the HR lever for every ratio across sessions. The three-way ANOVA analysis did not detect significant differences for group ($F_{(1,11)} = 0.28$, $p = 0.60$, $\eta_p^2 = 0.02$) or session ($F_{(3,33)} = 0.85$, $p = 0.47$, $\eta_p^2 = 0.07$). However, there was a ratio effect ($F_{(3,33)} = 11.92$, $p < 0.001$, $\eta_p^2 = 0.52$) indicating that the choice latency increased with higher ratios.

We also show the rate of lever presses in both groups for every ratio across sessions (Table 1). The three-way ANOVA analysis did not detect significant differences for group ($F_{(1,11)} = 0.65$, $p = 0.43$, $\eta_p^2 = 0.05$) or session ($F_{(3,33)} = 0.53$, $p = 0.66$, $\eta_p^2 = 0.04$). However, there was a ratio effect ($F_{(3,33)} = 11.61$, $p < 0.001$, $\eta_p^2 = 0.51$) indicating that animals in both groups reduce the number of presses per second at higher ratios. In addition, we also evaluated the number of omissions during the task after the third and fourth stress episodes. There were few omitted trials after the third (control = 0.25 ± 0.25 ; stress = 0.75 ± 0.75) and fourth (control = 0.62 ± 0.37 , stress = 1.62 ± 1.16) stress episodes in both groups. The number of omitted trials was not different between groups (group effect: $F_{(1,14)} = 0.58$, $p = 0.45$, $\eta_p^2 = 0.04$; session x group interaction: $F_{(1,14)} = 0.67$, $p = 0.42$, $\eta_p^2 = 0.04$) or across sessions ($F_{(1,14)} = 4.21$, $p = 0.060$, $\eta_p^2 = 0.23$).

Effects of ISD on the progressive ratio task

Rats were tested in a progressive ratio schedule after the second (S2) and fourth (S4) stress episodes, and 1 and 6 weeks after the last stress episode. A three-way ANOVA with repeated measures with group (Control, Stress) as between subjects and ratio (2, 5, 10, 20, 31) and session (S2-Post 6Weeks) as within subjects, was performed to compare the number of lever presses during the progressive ratio task. As shown in Figure 4A, in every session, both control and stressed animals pressed the lever more times whenever the ratio to receive a reward (i.e., press/g) was lower (ratio effect: $F_{(4,168)} = 24.59$, $p < 0.001$, $\eta_p^2 = 0.63$). Also,

there was an increase in the number of lever presses across sessions (session effect: $F_{(3,168)}=3.88$, $p=0.015$, $\eta_p^2=0.22$). The three-way ANOVA did not detect overall group effects ($F_{(1,14)}=0.64$, $p=0.43$, $\eta_p^2=0.04$), or significant group x ratio ($F_{(4,56)}=0.46$, $p=0.76$, $\eta_p^2=0.03$) and group x session ($F_{(3,42)}=1.84$, $p=0.15$, $\eta_p^2=0.11$) interactions. However, this analysis did reveal a significant session x ratio interaction ($F_{(3,12)}=3.41$, $p=0.001$, $\eta_p^2=0.19$), and therefore we performed two-way ANOVAs with repeated measures to further assess how the number of lever presses changed across sessions for every ratio (2, 5, 10, 20 and 31) in both groups. The two-way ANOVAs demonstrated increases in lever presses across sessions for ratio 2 ($F_{(3,42)}=6.76$, $p=0.001$, $\eta_p^2=0.32$), ratio 5 ($F_{(3,42)}=4.89$, $p=0.005$, $\eta_p^2=0.60$) and ratio 10 ($F_{(3,42)}=6.76$, $p=0.001$, $\eta_p^2=0.32$). They also revealed a session x group interaction for ratio 5 (price of 100 press/g) ($F_{(3,42)}=3.39$, $p=0.027$, $\eta_p^2=0.19$) indicating that stressed animals pressed the lever more times than controls at this ratio. Specifically, stressed animals pressed the lever more times compared to controls 6 weeks after the last stress episode ($t_{14}=2.86$, $p=0.012$, $\eta_p^2=0.37$; independent t test), when the price to receive rewards was low (Figure 4A).

We also analyzed whether social stress changed the total number of lever presses and the total number of pellets consumed across sessions by performing the corresponding two-way ANOVAs with group and sessions as between and within subjects, respectively. Figure 4B shows the total number of lever presses across sessions. There was an increase in the total number of lever presses across sessions (session effect: $F_{(3,42)}=4.56$, $p=0.007$, $\eta_p^2=0.24$). However, this analysis did not detect overall group effects ($F_{(1,14)}=0.66$, $p=0.42$, $\eta_p^2=0.045$) or significant session x group interaction ($F_{(3,42)}=1.80$, $p=0.161$, $\eta_p^2=0.11$).

Figure 4C shows the total number of pellets consumed across sessions. Similar to total lever presses, there were not overall group effects ($F_{(3,42)}=1.33$, $p=0.26$, $\eta_p^2=0.09$) or significant session x group interaction ($F_{(3,42)}=2.27$, $p=0.094$, $\eta_p^2=0.14$), but there was a significant change in the number of pellets consumed across sessions (session effect: $F_{(3,42)}=6.95$, $p=0.001$, $\eta_p^2=0.33$). This effect was significant for stressed ($F_{(3,21)}=4.54$, $p=0.013$, $\eta_p^2=0.39$) and control ($F_{(3,21)}=4.72$, $p=0.011$, $\eta_p^2=0.40$; one-way ANOVA with repeated measures) animals. However, a further analysis revealed that stressed animals, but not controls, consumed significantly more pellets at 6 weeks (stressed: $t_7=2.64$, $p=0.033$; control: $t_7=0.17$, $p=0.860$; paired t test) and 1 week (stressed: $t_7=3.50$, $p=0.010$; control: $t_7=1.59$, $p=0.160$; paired t test) compared to the previous sessions S2 and S4, respectively. Furthermore, there was a strong tendency for stressed animals to consume significantly more pellets than controls 6 weeks after the last stress episode ($t_{13}=2.07$, $p=0.056$, independent t test).

Figure 4D shows the break point for stressed and control animals, which is the ratio (price) at which animals stop pressing the lever. We performed a two-way ANOVA with group and sessions as between and within subjects, respectively, and found that there were not significant differences for group ($F_{(1,13)}=0.10$, $p=0.75$, $\eta_p^2=0.008$) or session x group interaction ($F_{(3,39)}=0.39$, $p=0.76$, $\eta_p^2=0.02$), but there was a significant effect across sessions ($F_{(3,39)}=3.64$, $p=0.021$, $\eta_p^2=0.22$). According to these results animals stop pressing the lever later at later sessions but both groups of animals stop pressing the lever at similar ratios (i.e., price).

DISCUSSION

We assessed whether the exposure to intermittent episodes of social stress changes the effort that animals are willing to make to obtain rewards (i.e., sugar pellets). In the effort-discounting task, we found that ISD decreases the preference for larger rewards associated with a larger effort (i.e., more lever presses), which suggests that ISD biases animal's choices to move away from costlier rewards. Yet these effects were observed 24-48 h after stress but not one week after the last stress episode. In addition, we found that ISD increases the number of lever presses and the total number of food pellets consumed during the progressive ratio task six weeks after stress, but not before, which suggests that ISD increases reward seeking in the long term. These results suggest that ISD produces different short- and long-term effects on the motivation for rewards and point to temporal dynamic adaptations in the function of the brain reward system that are relevant to better understand the transition from social adversity to drug abuse.

During the effort-discounting task animals are required to choose between low effort/low reward and high effort/high reward [10,18]; the high reward choice requires animals to press the lever more times in order to earn a larger reward (i.e. three sugar pellets instead of one). Food deprived rats value higher magnitude rewards more than lower magnitude. As expected, both control and stressed animals decreased their preference for the high reward option as the effort requirements increased from two to twenty lever presses. This effect paralleled an increase in the choice latency to the high reward lever. These results agree with previous studies using the same task [17,18], and confirm that animals are making their choices according to cost/benefit subjective evaluations.

Exposing animals to ISD decreased the preference for high rewards at higher ratios (i.e. higher prices) 24-48 h after the third and fourth stress episodes, which indicates that the effects of ISD on task performance are cumulative and depend on the effort involved in the choice. Similar effects have been found after acute restraint and chronic social stress [17,18,20]. Furthermore, these effects of ISD were time-dependent since they were not observed one week after stress. In addition, we also found that ISD did not change performance in the progressive ratio task 48 h after single stress episodes. The progressive ratio task evaluates whether animals are willing to work harder to earn rewards [12,14]. As shown, stressed and control animals made similar number of lever presses and put the same effort to obtain sugar pellets (i.e. lever presses). Taken together, our results indicate that ISD alters effort-based decision-making in the short term and biases animal's behavior to avoid higher effort options when a lower effort (lower reward) option is concurrently available but does not change the motivation for rewards. These results resemble previous studies which suggest that stress reduces effort-related motivated behaviors [18,20,22].

In contrast to the short-term effects, the animals exposed to ISD were more motivated to seek rewards in the long term. Thus, stressed animals pressed the lever more times (at low ratios) to obtain rewards and consumed more sugar pellets than control animals, during the progressive ratio task. Importantly, these effects were observed six weeks after the termination of the stress protocol. In line with these results, it has been shown that chronic social defeat can increase the motivation for rewards ten weeks after stress [29]. The

increased consumption of sugar pellets long after the termination of stress could be related to metabolic changes after ISD. Thus, it has been shown that chronic social stress alters metabolism and increases food consumption, and that this effect can be facilitated by using highly palatable foods like sugar [35,36]. However, it is currently unknown whether ISD and chronic social stress produce similar effects on metabolism.

The increased reward-seeking behavior after ISD shown here can be interpreted in the context of previous studies showing that ISD increases drug-self administration. Thus, by using the same ISD protocol as the one utilized here, studies have shown that ISD increases cocaine self-administration during a 24-h continuous access session (i.e. binge) several weeks after the last stress episode. Interestingly, the same studies show that ISD does not change the breakpoint for cocaine infusions in a progressive ratio schedule [5-7,37,38], which is in agreement with our results using food. In fact, as shown here, stressed animals did not “pay” higher prices for sugar pellets and stopped pressing the lever at similar ratios (i.e. breakpoint) compared to controls. These results suggest that stressed animals have more motivation to seek out food and drug rewards but not to work harder to obtain them. These results also point to long-term adaptations in the function of the brain reward system leading to an enhanced reward-seeking behavior. In this context, the enhanced seeking and consumption of rewards (i.e., food, drugs) after ISD might function as a stress-reliever, which ultimately could increase the risk for developing drug abuse [2,9,39].

At the mechanistic level, and based on previous work, ISD could alter effort-based reward seeking behavior by changing the activity of the hypothalamus-pituitary-adrenal (HPA) axis, and the release of the corticotropic-releasing-factor (CRF) and glucocorticoids [5,16,18,40]. Thus, previous studies suggest that increases of CRF during stress alter cost/benefit decision-making and decreases the preference for high magnitude rewards [18,41]. Similarly, it has been shown that the long-term administration of corticosterone biases animal’s behavior to low-cost rewards [22]. In addition, studies have also demonstrated that the activation of CRF receptors in the ventral tegmental area can increase drug-seeking behavior [5,8] and have suggested a link between ISD effects and long-term changes in the function of the mesolimbic dopamine system. Overall, these studies support the idea that both short- and long-term effects of ISD on effort-based reward-seeking behavior are produced by dynamic adaptations in the function of the HPA axis and the mesolimbic dopamine system. Critically, both the HPA axis (i.e., CRF, glucocorticoids) and the mesocorticolimbic dopamine system are involved in drug abuse vulnerability and substance use disorders [1,42-44].

Several methodological limitations should be considered for the present study. First, our experiments were performed in male rats and therefore the reported effects on effort-based reward-seeking behavior cannot be extended to both sexes. In fact, sex differences have been reported on reward seeking after social stress [20,45]. Second, some studies suggest that the effort-discounting task can be influenced by the order of experience on effort choice [46]. In the current study, both stressed and control animals experienced the task in the same order every session and therefore, it is difficult to know the actual impact of this factor on our results. Third, previous studies have shown that food-deprived animals can develop sucrose craving [36,47,48]. It is possible that food deprivation modulates performance in the

progressive ration task by having a stronger impact on stressed compared to control animals. Yet whether food deprivation interacts with the effects of other treatments, such as stress, is uncertain and requires further investigation. Finally, our results show an important individual variability, especially for stressed animals, during task performance. Individual differences most likely reflect the complexity of the neurobiological and physiological mechanisms involved in the effects of stress [49], and ultimately suggest that our results should be interpreted cautiously since all animals are not equally affected by ISD [50,51].

In summary, our study shows that ISD produces time-dependent changes in effort-based reward-seeking behavior by decreasing the motivation for high magnitude rewards—associated with high effort, in the short term, and increasing the motivation to obtain and consume rewards in the long term. These results suggest dynamic changes in the function of the reward system (i.e., CRF, dopamine) during and after the repeated exposure to social stress. It can be hypothesized that the short-term effects on effort-based decision-making after ISD can predict the long-term effects on the motivation for rewards. For instance, a decrease in the preference for high rewards shortly after stress might be associated with lower risk to increasing reward-seeking behavior and, ultimately lower vulnerability to drug abuse, in the long term. Further studies will be needed to substantiate this hypothesis. Our findings are relevant to better understand the link between social adversity and vulnerability to drug abuse, which is essential to help predict and prevent the development of substance use disorders.

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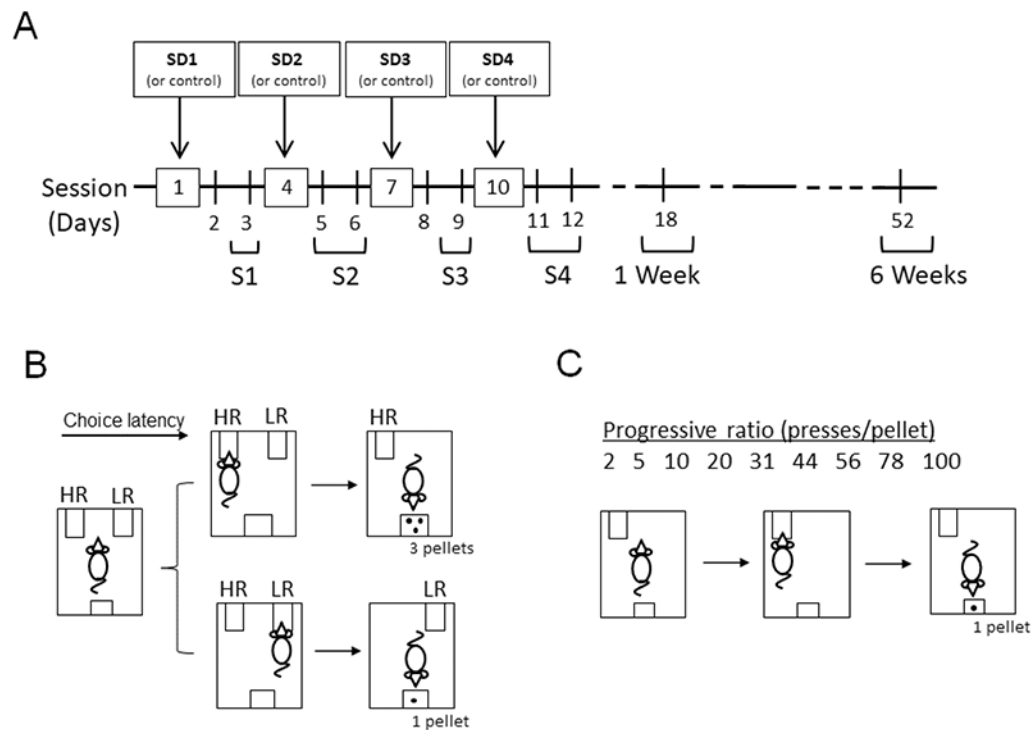
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**Figure 1:**

(A) Timeline for the intermittent social stress and the effort-based reward-seeking behavior testing sessions. Rats were exposed to social defeat stress once every three days (SD1-4) for ten days. Control animals were handled. (B) Effort-discounting task protocol. Rats chose between a High-Reward/High-Effort lever (HR) and a Low-Reward/Low-Effort lever (LR). Choosing the HR delivered three sugar pellets and choosing the LR delivered one sugar pellet. The effort-discounting task was performed 24-48 h after stress episodes and 1 week after the last stress episode (days 3, 5, 9, 11 and 18). (C) Progressive ratio task protocol. Rats were required to increase their effort (presses per pellet) to obtain rewards (one sugar pellet). The progressive ratio task was performed 48 h after the second and the fourth stress episodes, and 1 and 6 weeks after the last stress episode (days 6, 12, 18 and 52).

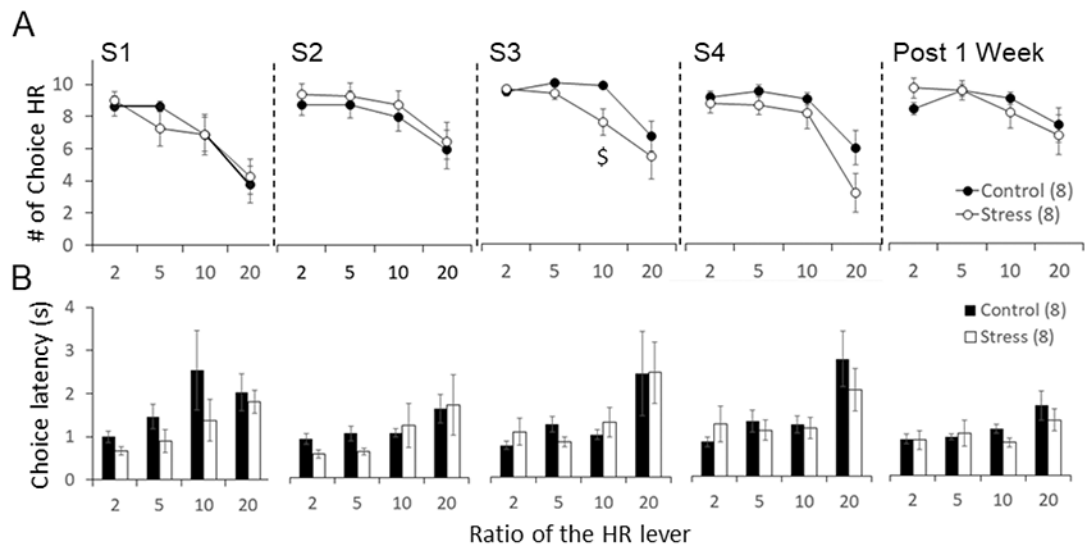


Figure 2:

Effort-discounting task performance for control and stressed animals. **(A)** Preference for the more effortful option (HR lever) at different ratios (2-20) and across sessions (S1-Post 1 Week). Data are the mean \pm SEM of the number of HR choices. **(B)** Choice latency at different ratios (2-20) and across sessions (S1-Post 1 Week). Data are the mean \pm SEM of the choice latency in seconds. Number of animals per group in parenthesis. \$ $p < 0.05$ compared to control.

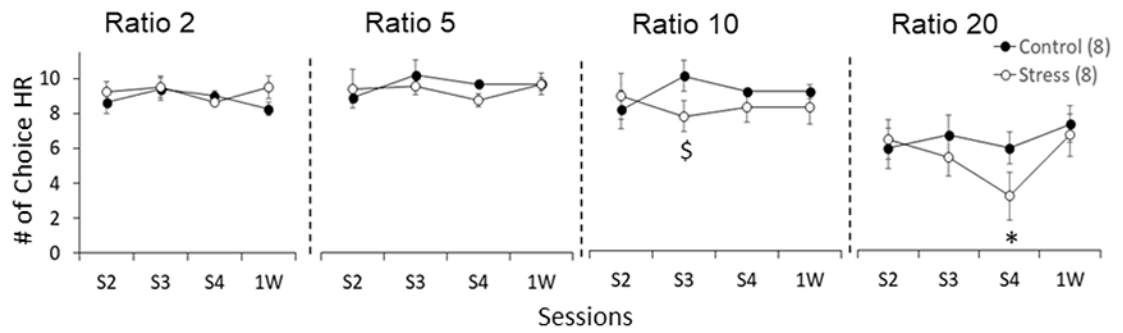
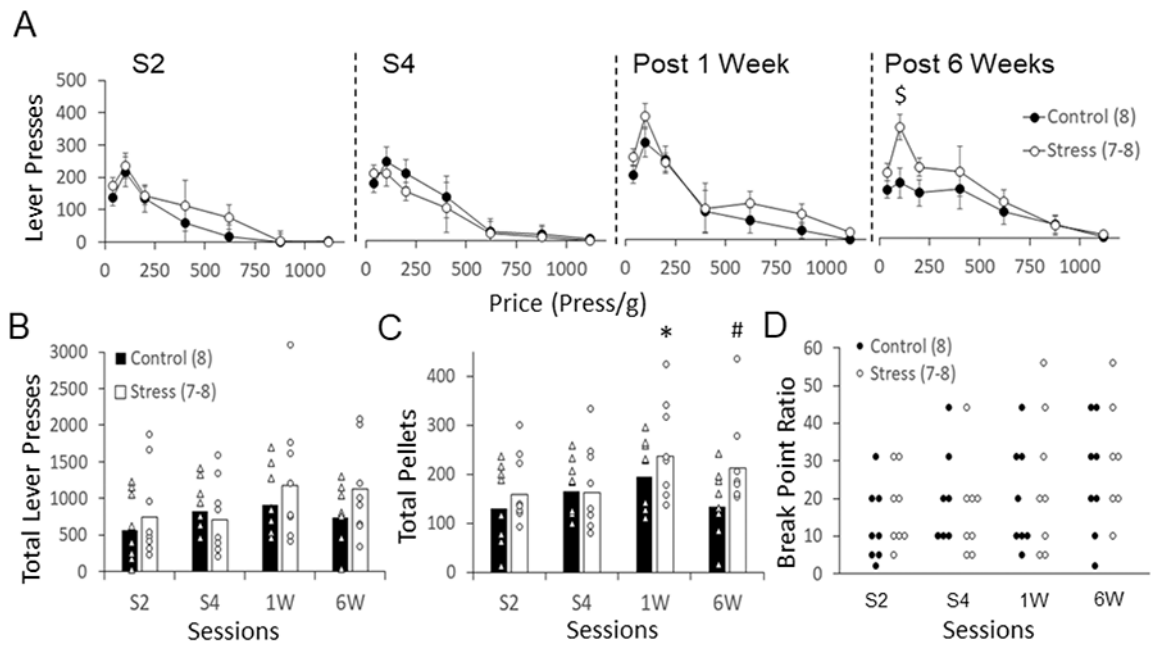


Figure 3:

Effort-discounting task performance across sessions (S2-Post 1 week) for each ratio (2-20) for control and stressed animals. Data are the mean \pm SEM of the number of HR choices. \$ $p < 0.05$ compared to control. Number of animals per group in parenthesis. \$ $p < 0.05$ compared to control; * $p < 0.05$ compared to S3 and 1W sessions.

**Figure 4:**

Progressive ratio task performance for control and stressed animals. **(A)** Lever presses at different prices (presses per grams of sugar) and across sessions (S2-Post 6 Weeks). Data are the mean \pm SEM of the number of lever presses. **(B)** Total number of lever presses across sessions. Data are the mean \pm SEM of the total number of lever presses. **(C)** Total number of pellets consumed across sessions. Data are the mean \pm SEM of the total number of pellets consumed. **(D)** Break point ratio across sessions. Each dot represents one rat. Number of animals per group in parenthesis. \$ $p < 0.05$ compared to control; * $p < 0.05$ compared to S4; # $p = 0.056$ compared to control.

Table 1.

Rate of lever presses (presses/s) during the effort-discounting task for every ratio (2-20) scheduled across sessions (S1-Post 1 Week). Animals decreased their rate of lever presses at higher ratios ($F_{(3,33)}= 11.61$, $p < 0.001$, $\eta_p^2 = 0.51$). Data are the mean \pm SEM (n= 8 per group).

		S1	S2	S3	S4	Post 1 Week
Ratio 2	Control	3.40 \pm 0.58	4.22 \pm 0.76	3.44 \pm 0.55	3.67 \pm 0.46	3.60 \pm 0.57
	Stress	3.58 \pm 0.56	3.97 \pm 0.53	3.82 \pm 0.63	3.60 \pm 0.73	3.07 \pm 0.40
Ratio 5	Control	2.37 \pm 0.28	2.25 \pm 0.29	2.51 \pm 0.21	2.48 \pm 0.27	2.64 \pm 0.20
	Stress	3.16 \pm 0.40	3.35 \pm 0.41	3.06 \pm 0.29	2.95 \pm 0.38	2.77 \pm 0.37
Ratio 10	Control	2.22 \pm 0.21	2.39 \pm 0.26	2.54 \pm 0.22	2.70 \pm 0.29	2.55 \pm 0.21
	Stress	3.21 \pm 0.48	3.08 \pm 0.45	2.87 \pm 0.29	3.16 \pm 0.58	3.07 \pm 0.47
Ratio 20	Control	1.95 \pm 0.24	1.96 \pm 0.16	2.16 \pm 0.21	1.99 \pm 0.18	2.10 \pm 0.13
	Stress	2.67 \pm 0.46	2.67 \pm 0.39	2.93 \pm 0.73	2.62 \pm 0.44	2.85 \pm 0.39