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Chronic methamphetamine self-administration alters cognitive flexibility in male rats

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Abstract

Rationale—Methamphetamine (meth) addiction is a chronically relapsing disorder that often produces persistent cognitive deficits. These include decreased cognitive flexibility, which may prevent meth addicts from altering their habitual drug abuse and leave them more susceptible to relapse. Multiple factors including low rates of compliance with research study participation and varied drug use patterns make the relationship between cognitive flexibility and relapse difficult to establish in clinical populations.

Objectives—Here, we combined an extended-access meth self-administration paradigm with an automated set-shifting task in rats to directly compare cognitive flexibility performance with meth-seeking behavior.

Methods—Rats were pre-trained on an automated visual discrimination task, followed by 14 days of extended access (6 h/day) of meth or sucrose self-administration. They were then tested in the set-shifting task on strategy shift and reversal and subsequently assessed for cue-induced reinstatement of meth seeking.

Results—Rats with a history of meth, but not sucrose, self-administration had selective deficits in reversal learning. Specifically, meth rats had an increase in the total number of errors and perseverative errors (corresponding to the old stimulus-reward association) following the reversal shift, which correlated with prior stable meth self-administration. However, no relationship was seen between errors during the reversal and cue-induced reinstatement.

Conclusion—The lack of association between meth-induced reversal deficits and cue-induced reinstatement to meth seeking indicates that these two domains may constitute independent pathologies of meth addiction.

Compliance with ethical standards Procedures were conducted in accordance with the “Guide for the Care and Use of Laboratory Rats” (Institute of Laboratory Animal Resources on Life Sciences, National Resources Council, 1996) and approved by the IACUC of the Medical University of South Carolina.

Keywords

Cognitive flexibility; Methamphetamine; Reinstatement; Reversal; Self-administration

Introduction

Methamphetamine (meth) addiction has been associated with a variety of cognitive deficits, including impaired performance on tests of cognitive flexibility, which measure the ability to adjust behavior when new information is presented. These deficits may impair meth addicts from altering their habitual drug abuse behavior and ultimately lead to an inability to initiate abstinence or resist relapse (Kalechstein et al. 2003; Paulus et al. 2005).

Different forms of cognitive flexibility comprise key aspects of executive function that are mediated in large part by regions of the prefrontal cortex (PFC) in humans, non-human primates, and rodents (Robbins and Arnsten 2009). Brain imaging studies demonstrated that meth addicts have decreased basal PFC activity (Paulus et al. 2003) and exhibit cognitive deficits in PFC-dependent tasks that require attention, working memory, or cognitive flexibility (McCann et al. 2008; Nordahl et al. 2003; Salo et al. 2009). One form of cognitive flexibility entails the ability to shift strategies in response to rule changes, and in humans, this function can be assessed using extradimensional-intradimensional (ED/ID) tasks or the Wisconsin Card Sorting Test (WCST). Reversal learning tasks assess a relatively simpler form of flexibility entailing shifts between different stimulus-reward associations (Cools et al. 2002; Morris and Dolan 2004). In general, meth addicts have difficulty shifting responses according to rule changes. Specifically, they make more perseverative errors (i.e., following the old rule) in the WCST (Chung et al. 2007; Henry et al. 2010; Kim et al. 2005, 2011) and exhibit deficits on tasks with reversal learning components (Bechara et al. 2001; London et al. 2000). Although cognitive inflexibility appears to be common among meth addicts (Chung et al. 2007; Henry et al. 2010; Kim et al. 2005, 2011), clinical studies have not directly examined the relationship between cognitive flexibility and relapse.

Several set-shifting tasks have been developed to assess cognitive flexibility in rodents. Studies using these behavioral measures have established that distinct subregions of PFC mediate different components of behavioral flexibility. In rats, lesions or inactivation of medial PFC disrupt shifts between different rules, strategies, or attentional sets (Birrell and Brown 2000; Dias et al. 1996; Floresco et al. 2008), whereas orbitofrontal cortex (OFC) lesions disrupt reversal learning (Boulougouris et al. 2007; Ghods-Sharifi et al. 2008; Schoenbaum et al. 2002). A rodent “digging” task version of the ED/ID shift test was patterned after similar tasks used in humans (Roberts et al. 1988; Slamecka 1968) and relies on intrinsically salient stimuli, such as digging media and scents (Birrell and Brown 2000). However, detailed analyses of specific types of ED shift errors cannot be measured in the ED/ID shift task (as in the WCST) because the stimuli change throughout the task and not enough trials occur to adequately quantify these errors. Thus, a limitation of the ED/ID task is the difficulty in ascertaining whether impairments in shifting induced by a particular manipulation result from increased perseveration, impairments in set maintenance, or acquiring a novel strategy. An alternative approach entails having subjects switch between

different discrimination strategies either on a maze or in an operant chamber, originally referred to as a non-reversal shift (Mackintosh and Holgate 1968; Mackintosh et al. 1968) and more recently termed a “strategy shift” (Floresco et al. 2008, 2009). An automated version of this procedure patterned after similar procedures used with a T-maze has been developed for an operant conditioning chamber and uses both visual and spatial dimensions (Brady and Floresco 2015). As in both maze-based procedures and the ED phase of the “digging” task, performance during the strategy shift is disrupted by manipulations of the medial PFC (Birrell and Brown 2000; Floresco et al. 2008). Notably, this automated approach allows partitioning of different types of errors made during shifts.

Although it has been proposed that impaired cognitive flexibility seen in meth addicts may contribute to relapse (Chung et al. 2007; Henry et al. 2010; Kim et al. 2005, 2011), surprisingly few studies have examined cognitive flexibility in animal models of meth addiction. Izquierdo et al. (2010) demonstrated that an acute ‘binge’ regimen (4 meth injections $\times$ 2 days) resulted in specific reversal deficits in both the attentional set-shift task and in a touchscreen task (Izquierdo et al. 2010). Likewise, previous work by our group revealed impairments during the ED phase of the ED/ID attentional set-shift task following long access (6 h/day) meth self-administration (Parsegian et al. 2011). Since analysis of specific error types was not possible, it was unclear whether these impairments were associated with increased perseverative behavior or other types of impairments. Furthermore, the design of this prior study precluded a detailed analysis of how deficits in cognitive flexibility may relate to reinstatement of drug-seeking behavior. Thus, in the current study, we used an operant-based set-shifting task in combination with the meth self-administration and reinstatement model to examine potential relationships between cognitive flexibility, meth intake, and relapse-like behavior.

Methods and materials

Subjects

Male Long-Evans rats (Charles River, Raleigh, NC; $n = 43$) weighing 250–300 g at the time of arrival were used. All rats were individually housed on a reverse 12:12-h light-dark cycle in a temperature- and humidity-controlled vivarium. Water was available ad libitum throughout the study. During all set-shifting phases of the study and acquisition of self-administration, rats were maintained with rat chow (Harlan, Indianapolis, IN, USA) at 85 % of their free-feeding body weight. For all other phases of the study, rats were maintained ad libitum. Procedures were conducted in accordance with the “Guide for the Care and Use of Laboratory Rats” (Institute of Laboratory Animal Resources on Life Sciences, National Resources Council, 1996) and approved by the IACUC of the Medical University of South Carolina.

Behavioral test apparatus

All set-shifting and self-administration experiments were conducted during the rats’ active (dark) cycle in standard Plexiglas self-administration chambers (30 $\times$ 20 $\times$ 20 cm) that were enclosed in sound attenuating cubicles with a ventilation fan (Med Associates, St. Albans, Vermont) and linked to a computerized data collection program (MED PC, Med Associates).

Importantly, the self-administration chambers were located in a different room from the set-shifting chambers, and stimuli in the chambers were altered to prevent contextual association with the set-shifting chamber.

Initial lever press training and visual cue discrimination

All set-shifting procedures have been previously described (Floresco et al. 2008, 2009). However, 100 μ l of 15 % sucrose solution was delivered to the reward port upon a correct response rather than a sucrose pellet. Rats first underwent initial lever pressing training in the self-administration chambers, each equipped with a house light, two retractable levers with a white stimulus light above each lever, and a liquid reward port located between the levers. For initial training (1 h/day, ~2 weeks), rats were trained under a fixed ratio 1 schedule of reinforcement on each lever (left/right, counterbalanced by day) to a criterion of 50 rewards or 30 min for each lever. In the second phase of training, rats learned to press the levers within 10 s after insertion to receive a sucrose reward. These sessions consisted of 90 trials, and rats were required to press either the left or right lever within 10 s or the trial was counted as an omission. Completion of this stage of training occurred when a rat achieved <5 omissions/session, with a minimum of 5 days of training. Rats were then tested for side bias, whereby both levers were presented and responses on either lever resulted in a reward. The lever chosen most often during this session was considered their preferred lever.

The following day, rats were trained on visual cue discrimination. Every 15 s, one of the two stimulus lights above a lever was illuminated and 3 s later, the houselight was turned on and both levers were inserted into the chamber. Rats had a maximum of 10 s to respond on one of the levers. A response on the lever under the stimulus light caused both levers to retract and deliver sucrose reward, with the houselight extinguished 3 s later. An erroneous response on the other lever caused retraction of levers, houselight termination, and no reward delivery. Rats were trained for a minimum of 30 trials and to a criterion of 10 consecutive correct responses (excluding trial omissions) for 1–4 days. Rats were assigned to either the sucrose or meth group in a counterbalanced fashion based on performance on the visual discrimination.

Surgery

Following visual discrimination training, rats were anesthetized and implanted with indwelling jugular catheters using previously described methods (Reichel et al. 2012) and were allowed to recover for at least 5 days. All surgical procedures were identical for sham surgeries except that no catheter was inserted.

Self-administration

The self-administration chambers were located in a different room from the set-shifting chambers and were equipped with two nose ports located on opposite walls with white stimulus lights above each nose port (used as the houselights) and a tone generator. For meth self-administration, infusion tubing enclosed in steel spring leashes (Plastics One Inc., Roanoke, VA) was connected to the infusion harness and a weighted swivel apparatus (Instech, Plymouth Meeting, PA) was suspended above the box to allow for free movement within the chamber. During daily 6-h meth self-administration sessions, a response on the

active (meth-paired) nose port resulted in a meth infusion (0.06 mg/kg) and presentation of a 5-s tone (78 dB, 4.5 kHz), followed by a 20-s time-out period with house lights off. During the time-out period, responses on the active and inactive nose ports were recorded, but had no scheduled consequences. A separate group of rats were trained to respond for sucrose pellets, using procedures similar to those for meth self-administration. Sucrose-trained rats underwent the same procedures, except that they underwent a sham surgery and self-administered banana-flavored pellets. For these subjects, a pellet dispenser was located to the left of the active nose port. These rats received flavored pellets during this phase (different from the liquid sucrose reward used in the set-shifting task) to maximize the differences between tasks. The number of pellets that rats self-administered was restricted to the average number of infusions for meth rats (i.e., 60 pellets) to help control for the effect of the operant learning experience on set-shifting. Self-administration was complete upon reaching a criterion of >25 infusions/pellets for >14 days.

Set shift and reversal

After self-administration, rats underwent 3 days of abstinence before resumption of testing in the automated set-shift task (ASST) chambers where rats had learned visual discrimination previously. First, meth and sucrose rats underwent a reminder visual discrimination task. After reestablishing criterion performance (10 consecutive correct responses) on the visual discrimination, the rats were tested on the strategy set shift the following day. During the initial phase of this session, subjects received 20 “reminder” trials where they were required to use the visual rule to assess their ability to remember the rule learned the previous day (Enomoto et al. 2011). On the 21st trial, reinforcement contingencies shifted, so that the rat was required to discontinue use of the previous rule (follow the light) and shift to a new rule using a spatially oriented response strategy (location of lever) to receive a reward. For this shift, responses on the lever opposite to the rat’s side bias elicited a reward, regardless of the location of the light. All other aspects of the procedure were identical to those used during visual discrimination training. Successful performance required 10 consecutive correct responses, not including omissions. There was no maximum trial limit on these tests; thus, all rats completed each shift on the same day.

The day after the strategy shift, rats performed the reversal test, where the correct response was now the lever opposite the previously correct lever (during strategy set shift), regardless of the location of light. Again, rats received 20 reminder trials at the start of the session such that the correct lever was the same as during the set-shift test the previous day. On the 21st trial, the reinforcement contingencies switched and all subsequent trials following the new rule.

Extinction of responding and reinstatement

After the reversal test, rats underwent 2-h daily extinction sessions in the self-administration chambers, whereby responses on both active and inactive nose ports were recorded but had no scheduled consequences. Extinction criterion consisted of a minimum of 7 days with <25 nose pokes for two consecutive days. After extinction, rats with a history of meth or sucrose self-administration underwent testing for reinstatement elicited by cues (2-h sessions; tone/

lights on upon active nose poke). All rats received one non-contingent cue at the beginning of the session, with all subsequent cues only in response to active nose pokes.

Error analysis

Error analysis was based on criteria established by Floresco et al. (2008). Errors made during a shift were categorized into three subtypes: perseverative, regressive, and never reinforced. Perseverative errors reflected a rat's inability to shift away from the previously learned strategy or rule, and regressive errors measured the ability of the rat to maintain the new rule. To measure perseveration, trials in which a perseverative error is possible were counted and separated into blocks of 8; perseverative errors were scored when a rat pressed the incorrect lever on 6 or more trials per block of 8 trials. Once a rat made <6 perseverative errors in a block, all-subsequent errors were scored as regressive errors. Never reinforced errors were scored during the set shift when a rat selected an incorrect lever when the cue light was illuminated over the correct lever and serve as an index of how rapidly rats can ascertain the novel strategy following a shift. For the reversal test, we only analyzed perseverative and regressive errors, using similar criteria as above.

Data analysis

Daily meth intake (mg/kg), rates of responding on the reinforced and non-reinforced nose ports, and the number of reinforcers during SA sessions were analyzed using repeated measures analysis of variance (ANOVA) with Bonferroni-corrected post hoc comparisons between individual data points when appropriate. Data for the various stages of the set-shifting task were analyzed by *t* test or ANOVA. Pearson correlations and linear regression were used to assess the relationship between behavioral tests. Data points were eliminated if the rat lost catheter patency or failed to extinguish prior to reinstatement testing. All post hoc analyses were conducted using Bonferroni tests with the alpha set at 0.05 and all data are expressed as the mean $\pm$ SEM.

Results

The full experimental timeline is summarized in Fig. 1.

Visual cue discrimination training

Meth and sucrose rats were trained on visual cue discrimination until they achieved the criterion of 10 consecutive correct responses (excluding omissions) with a maximum of 200 trials per day. Meth and sucrose rats were trained for 1–4 days and did not differ in numbers of trials (meth = 250.5 ± 53.8 , sucrose = 205.2 ± 37.1 ; $t_{39} = 0.72$, $p > 0.05$) or errors (meth = 94.9 ± 22.5 , sucrose = 86.0 ± 15.8 ; $t_{39} = 0.34$, $p > 0.05$) to reach criterion during the initial visual discrimination sessions prior to self-administration.

Self-administration, extinction, and cue-induced reinstatement

Meth and sucrose rats quickly acquired meth or sucrose pellet self-administration over the 14 sessions. Across the 6-h daily sessions, meth rats escalated their intake from 3.01 ± 0.33 mg/kg/day on day 1 to 5.46 ± 0.30 mg/kg/day by day 14 (Fig. 2a). The escalation in total meth intake was statistically significant ($F_{(13,169)} = 11.33$, $p < 0.0001$), with differences

between day 1 and days 6–14 ($p < 0.01$ – 0.0001). After testing on set-shifting and reversal learning procedures, rats experienced extinction of self-administration training. Here, active nose pokes predictably decreased in both meth and sucrose rats (Fig. 2b, c, shaded area). Rats showed robust cue-induced reinstatement when comparing active nose pokes on the last day of extinction vs cue-induced reinstatement for meth seeking ($t_{16} = 8.55$, $p < 0.0001$) and sucrose seeking ($t_{24} = 11.90$, $p < 0.0001$; Fig. 2b, c).

Visual discrimination recall, strategy set shift, and reversal

Visual cue discrimination recall—Meth and sucrose rats were trained for 1–5 days and did not differ in the number of trials (meth = 188.5 ± 36.3 , sucrose = 152.4 ± 26.3 ; $t_{37} = 0.83$, $p > 0.05$) or errors (meth = 69.9 ± 14.2 , sucrose = 53.4 ± 9.0 ; $t_{37} = 1.02$, $p > 0.05$) to reach criterion during the visual discrimination recall sessions after self-administration and reinstatement. Thus, meth self-administration did not affect the ability to recall a previously acquired visual cue discrimination rule.

Strategy set shift (visual cue shift to lever position)—There were no differences in the proportion of correct responses during the pre-shift (first 20 trials of the session before the “rule” shift) between the meth and sucrose groups (meth = 79.9 ± 2.0 %, sucrose = 79.4 ± 2.7 %; $t_{37} = 0.15$, $p > 0.05$; data not shown). Following the rule shift, no differences were observed between groups for total number of errors to reach criterion ($t_{37} = 0.34$, $p > 0.05$; Fig. 3a), nor were differences observed when the errors were broken down into subtypes: perseverative errors ($t_{37} = 0.23$, $p > 0.05$); regressive errors ($t_{37} = 0.92$, $p > 0.05$) (Fig. 3b). Curiously, there was a difference between groups for the never reinforced errors ($t_{37} = 2.480$, $p < 0.05$). Thus, a history of meth self-administration did not appear to affect the retrieval of the visual cue “rule” from the previous day, nor did it affect shifting from a visual cue to a spatial response discrimination strategy. The difference in never reinforced errors may reflect a difference in exploratory behavior between the two groups.

Reversal—Similar to the strategy set-shift test, there was no difference between groups for the proportion of correct responses during 20 reminder trials that were given prior to the reversal shift (meth = 73.3 ± 2.5 %, sucrose = 78.0 ± 2.1 %; $t_{35} = 1.46$, $p > 0.05$). However, analysis of data following the rule shift revealed a significant increase in the total number of errors to reach criterion ($t_{35} = 2.47$, $p = 0.02$) in meth rats as compared to sucrose controls (Fig. 3c). Further analysis of the error subtypes showed that this increase in the number of errors was due to an increase in perseverative errors ($t_{35} = 2.95$, $p = 0.006$), but not regressive errors ($t_{35} = 0.10$, $p = 0.92$) (Fig. 3d). Therefore, meth self-administration did not affect retrieval of an egocentric spatial discrimination acquired during the set shift on the previous day, but did impair reversal of this association by retarding the ability to cease responding on the previously correct lever.

Relationship between meth seeking and cognitive flexibility

For rats with a meth history, a significant positive correlation was found between the mean number of active nose pokes on the last 5 days of self-administration (when meth taking had stabilized) and the number of total errors ($r^2 = 0.328$, $p = 0.02$) and perseverative errors ($r^2 = 0.327$, $p = 0.02$) made during the reversal (Fig. 4a, b). Additionally, meth intake on the last 5

days correlated with the total number of errors during the reversal ($r^2 = 0.252$, $p = 0.04$). In sucrose-experienced rats, no correlations were found between mean nose pokes on the last 5 days of self-administration and number of total errors ($r^2 = 0.058$, $p > 0.05$) or perseverative errors ($r^2 = 0.062$, $p > 0.05$) made during the reversal (Fig. 4c, d). Intake correlations were not calculated for sucrose controls, since all rats were limited to 60 sucrose pellets per day. There were no other significant correlations for either group between seeking during self-administration (or intake for meth rats) and any measure during the set shift. Thus, a selective positive relationship was only seen between meth seeking and intake with increased number of errors made during the reversal. Additionally, there were no significant correlations between the first day of extinction or number of days to reach extinction criterion and any of the behavioral flexibility measures.

Next, we assessed possible relationships between behavior during the reversal learning and reinstatement behavior. Specifically, we examined potential relationships between the number of total errors and perseverative errors during the reversal (in which the meth group showed a deficit) and cue-induced reinstatement. There were no significant correlations between reinstatement and the number of total errors (meth, $r^2 = 0.020$, $p > 0.05$; sucrose, $r^2 = 0.043$, $p > 0.05$) or perseverative errors (meth, $r^2 = 0.0007$, $p > 0.05$; sucrose, $r^2 = 0.004$, $p > 0.05$) (Fig. 5). Additionally, no significant correlations were seen in either group between measures of set shift and cue-induced reinstatement (data not shown).

Discussion

Using an operant-based set-shifting task in combination with a self-administration and reinstatement model, we showed that rats with a history of long access meth self-administration had selective deficits in reversal learning. Specifically, meth rats had an increase in the total number of errors and perseverative errors following the rule shift, which correlated with meth seeking as measured by average number of active nose pokes during stable meth self-administration. However, we found no relationship between errors made during the reversal performance and subsequent cue-induced reinstatement to meth seeking. These findings suggest that while prolonged meth self-administration causes deficits in reversal learning, these

A variety of rodent set-shifting and reversal learning tasks have been developed, using different stimuli and procedures. Yet, one commonality among them is that lesions or inactivation of the medial PFC disrupts extradimensional shift performance (Birrell and Brown 2000; Dias et al. 1996; Floresco et al. 2008), whereas OFC lesions disrupt reversal learning (Boulougouris et al. 2007; Ghods-Sharifi et al. 2008; McAlonan and Brown 2003; Schoenbaum et al. 2002). In the present study, we observed that rats with a history of meth self-administration demonstrated selective deficits in reversal learning known to be dependent on the functional integrity of the OFC. As such, these results suggest that extended access to meth induces OFC dysfunction, a finding consistent with what has been observed in human meth addicts. Imaging studies have demonstrated that meth addicts have altered metabolism in the OFC (London et al. 2004; Volkow et al. 2001) and exhibit deficits in cognitive flexibility on tasks thought to be OFC mediated, such as the Iowa gambling task (Bechara et al. 2001; London et al. 2000).

Meth-induced reversal deficits have been previously shown after a binge regimen of meth in both a touchscreen reversal learning task and an attentional set-shifting task (Izquierdo et al. 2010). Reversal deficits have also been observed after sensitizing regimens of amphetamine (Fletcher et al. 2005) or cocaine (Schoenbaum et al. 2004) and after cocaine self-administration (Calu et al. 2007). The procedures used in our study allowed us to analyze specific error types to obtain additional insight into the underlying nature of these deficits. During the reversal, meth rats had an increase in total errors compared to sucrose controls. More specifically, the increase manifested as more perseverative, but not regressive errors. This distinction indicates that the meth rats were slower to disengage from the previously learned rule as quickly as the sucrose controls, but could maintain the rule once initial learning had occurred. In this regard, lesions to the OFC also induce an increase in perseverative errors on a similar reversal learning task (Boulougouris et al. 2007), providing additional evidence that prolonged meth self-administration perturbs OFC functioning.

In contrast to the effects of meth self-administration on reversal learning, we observed no effect of these treatments on set-shifting functions dependent on the medial PFC assessed with an operant procedure where rats were required to switch between different discrimination rules.

We observed a minor difference in never reinforced errors, which could reflect a subtle difference in exploratory behavior. However, with the lack of differences in all other measures, we are hesitant to infer this difference as a deficit. The overall lack of a deficit in the strategy set shift was somewhat surprising, considering that other studies have found that meth addicts also show deficits in shifting tasks such as the WCST primarily mediated by the lateral prefrontal cortex (Henry et al. 2010), which at least with respect to set-shifting has been proposed to be functionally homologous to the medial PFC in rats (Brown and Bowman 2002).

We previously reported a deficit during the ED phase of the attentional set-shifting task, developed by Birrell and Brown (2000), after long access meth self-administration (Parsegian et al. 2011). Several reasons could account for the differences in results of meth exposure on strategy set-shifting in these studies. First, the two paradigms are very different in regard to the nature of the learning experience, the specific type of responses required to obtain reward (digging vs lever pressing), and the type of stimuli used (odor and tactile cues vs primarily visuospatial cues). In addition, the ED deficit that we previously reported was observed 1 day after completion of daily meth self-administration and showed some degree of recovery over time. In contrast, in the current study, rats were tested on the set-shifting task 5–7 days after meth self-administration. Thus, the ED deficit may have been a result of early abstinence, while the deficit that we observed in reversal learning may be due to chronic meth exposure. Of perhaps greater relevance, in our previous study, rats were trained to approach a particular stimulus dimension over multiple phases of the ED/ID task (simple and compound discriminations, reversals, ID) prior to receiving an extradimensional shift, whereas in the present study, rats acquired the initial rule in a single day of training. Thus, set-shifting functions mediated by the medial PFC may be more susceptible to the deleterious effects of meth self-administration after extended experience with an initial rule or attentional set. Alternatively, it is equally plausible that functions related to formation and

shifting of attentional sets are more sensitive to repeated meth exposure than those related to shifting between different rules or strategies.

Deficits in cognitive flexibility observed in meth addicts are thought to relate to their inability to alter their behavior despite negative consequences and contribute to the difficulty initiating or maintaining abstinence. In cocaine abusers, drug craving during early abstinence has been associated with increased OFC metabolism (Volkow et al. 1991). Given the meth-induced impairments in OFC-dependent reversal learning observed in the present study, we expected that a relationship might emerge between the cognitive deficits after meth self-administration and reinstatement to meth seeking. In this regard, we observed a relationship between increased meth seeking during self-administration and a more pronounced reversal deficit. Specifically, we observed a positive correlation between meth intake (as measured by number of active nose pokes) and the number of total and perseverative errors during the reversal, an effect not observed in the sucrose control group. This is consistent with many human studies where greater lifetime meth use resulted in more pronounced cognitive deficits (Ornstein et al. 2000; Stewart et al. 2014). The current data provide additional support for the notion that impairments in cognitive functioning observed in psychostimulant abusers are due in part to repeated drug exposure, as opposed to pre-existing cognitive abnormalities (Rogers and Robbins 2001).

The present findings that cognitive dysfunction was more related to prior meth intake rather than relapse is interesting in light of recent findings obtained with human meth addicts (Stewart et al. 2014). In that study, recently abstinent meth addicts were measured on a task comprised of win/stay and lose/shift components and relapse behavior was monitored after 1 year. Meth addicts with the greatest lifetime meth use made more perseverative errors in the task, similar to what we observed in the present study. Importantly, no relationship was seen between the number of perseverative errors made and the risk of relapse. These results indicate that chronic meth-induced deficits that produce increased perseveration by meth intake may be more related to the inability to initiate abstinence, rather than increased predilection to relapse. However, it is important to note that cognitive flexibility in our study and in Stewart et al. (2014) was measured earlier in abstinence than at the time when relapse testing (or actual relapse) occurred. We cannot therefore discount the possibility that an association with relapse behavior may exist if measured in a similar timeframe.

The mechanism by which chronic meth is producing these effects is currently unclear. However, altered OFC metabolism in meth addicts has been shown to be associated with decreased D2 receptor binding in the striatum (Volkow et al. 2001). Similarly, low D2 receptor availability predicts poorer performance in reversal learning tasks in animals (Groman et al. 2011; Laughlin et al. 2011) and humans (Jocham et al. 2009). Although it is tempting to speculate that dysfunctions of the dopamine system underlie this deficit, chronic meth affects many neurotransmitter systems that are known to contribute to reversal learning, including serotonin and norepinephrine (Izquierdo and Jentsch 2012).

To summarize, the present study revealed that long-access meth self-administration in rats perturbs certain forms of cognitive flexibility mediated by the OFC, whereas shifts between different discrimination strategies (mediated by the medial PFC) are relatively spared.

Moreover, the magnitude of impairment observed in meth-experienced rats was related to the amount of meth intake, suggesting that prior drug exposure can lead to deficits in frontal lobe function. On the other hand, we did not observe a relationship between impairments in reversal learning and cue-induced reinstatement of drug-seeking behavior, leaving open the possibility that other aspects of relapse not related to discrete drug-related cues may be linked to OFC dysfunction. Collectively, these findings provide additional insight into how meth addiction may perturb cognitive function mediated by different regions of the frontal lobes and point to new avenues of research into how this dysfunction may be related to drug taking and relapse.

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Fig. 1.

Experimental timeline. The *top* portion depicts behavioral training and testing that occurred in separate behavioral chambers for the ASST. The *bottom* portion depicts events relating to the self-administration (SA) paradigm. Rats were first pre-trained for the ASST, underwent side-bias testing, and trained on the visual discrimination (*Visual Discrim.*). Following training, rats underwent catheter (or sham) surgery and 14 days of long-access SA (meth or sucrose-pellet control). After 3 days of abstinence, rats were re-trained on the visual discrimination rule (*Re-train*; rule: light) and then tested on strategy set-shift test (*Shift*; rule: light to lever) and reversal test (*Rev.*; rule: lever to opposite lever). Rats then experienced extinction trials in the SA chambers and cue-induced reinstatement testing

Fig. 2. Self-administration (SA) in meth ($n = 19$) and sucrose ($n = 24$) rats. **a** Meth intake (mg/kg) during 14 days of self-administration. Significant differences from the first day of long access are indicated ($*p < 0.05$). Average nose pokes during self-administration, extinction, and cue-induced reinstatement are shown for meth (**b**) and sucrose (**c**) rats

Fig. 3.

Performance during the strategy set shift and reversal following self-administration ($n = 19$ – 23 per group). During the set shift, the rat shifts from the previous rule (light) to response strategy (side of lever). **a** Errors to criterion following the rule change during the set shift. **b** Comparison of the types of errors committed during the ED. During the reversal test, the rat shifts from the previous rule (side of lever) to the opposite lever. **c** Errors to criterion following the rule change during the reversal. **d** Comparison of the types of errors committed during the reversal. * $p < 0.05$; significantly different from sucrose

Fig. 4.

Scatter plots and correlation coefficients for the relationships between stable self-administration (last 5 days) and errors committed during the reversal. Meth rats ($n = 18$) showed significant positive correlations between the number of active nose pokes and total number of errors (a) and perseverative errors (b) committed during the reversal. Sucrose control rats ($n = 21$) showed no significant correlations between self-administration and total number of errors (c) or perseverative errors (d) committed during the reversal.

Fig. 5.

Scatter plots and correlation coefficients for the relationships between cue-induced reinstatement and errors committed during the reversal in meth ($n = 15$) and sucrose ($n = 21$) rats. No significant correlations were seen between the number of active nose pokes during cue-induced reinstatement and total number of errors or perseverative errors committed during the reversal for meth rats (**a, b**) or sucrose rats (**c, d**) changes in cognitive flexibility may reflect symptoms that are independent of motivated meth seeking maintained by drug-paired cues.