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Recent updates on incubation of drug craving: a mini-review

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ABSTRACT

Cue-induced drug craving progressively increases after prolonged withdrawal from drug self-administration in laboratory animals, a behavioral phenomenon termed 'incubation of drug craving.' Studies over the years have revealed several important neural mechanisms contributing to incubation of drug craving. In this mini-review, we first discuss three excellent *Addiction Biology* publications on incubation of drug craving in both human and laboratory animals. We then review several key publications from the past year on behavioral and mechanistic findings related to incubation of drug craving.

Keywords Addiction, cocaine, dopamine, glutamate, incubation, relapse.

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Relapse to drug use is one of the major challenges in treating drug addiction and a common trigger for relapse is exposure to drug-associated cues and contexts. In 1986, Gawin & Kleber (1986) proposed that craving, triggered by drug-associated cues, progressively increases during the early days of abstinence and stays elevated after prolonged time periods. In 2001, Grimm *et al.* (2001) identified a similar behavioral phenomenon in rats: time-dependent increases in cue-induced cocaine seeking during the first few months of withdrawal from cocaine self-administration (also see Tran-Nguyen *et al.* 1998; Neisewander *et al.* 2000). This phenomenon, termed 'incubation of drug craving', is also observed in rats trained to self-administer heroin (Shalev *et al.* 2001), methamphetamine (Shepard *et al.* 2004), alcohol (Bienkowski *et al.* 2004) and nicotine (Abdolahi *et al.* 2010), as well as non-drug rewards such as sucrose (Grimm, Shaham & Hope 2002). Over the years, several groups have advanced our understanding of the underlying mechanisms of incubation of drug craving (Pickens *et al.* 2011; Marchant, Li & Shaham 2013). Recently, three excellent publications in *Addiction Biology* (Thiel *et al.* 2012; Xi *et al.* 2013; Li *et al.* 2014) further explore incubation in human and animal studies. Here, we review these three publications as well as extend our focus to other key publications related to incubation of drug craving since April 2013 (Lee *et al.* 2013; Theberge *et al.* 2013; Wang *et al.* 2013; Counotte *et al.* 2014; Guillem, Ahmed & Peoples 2014; Halbout *et al.* 2014; Krasnova *et al.* 2014; Loweth *et al.* 2014; Ma *et al.* 2014; Li *et al.* in press).

Clinical studies have confirmed that incubation of craving also occurs in human drug users. This phenomenon was first demonstrated in abstinent smokers (Bedi *et al.* 2011), and in 2013 Wang *et al.* (2013) reported that methamphetamine-dependent patients exhibited a time-dependent increase in cue-induced craving for up to 3 months of abstinence. A recent *Addiction Biology* study in alcoholics showed that cue-induced alcohol craving is higher after 60 days of abstinence than after 7 days (Li *et al.* 2014). Interestingly, in all three studies the time course of cue-induced drug craving (increasing over time) was opposite to that of baseline non-provoked drug craving (decreased over time). The demonstration of incubation in human studies provides support for the translational potential of therapeutic targets for relapse uncovered through rodent mechanistic studies.

In 2012, Thiel *et al.* (2012) demonstrated that environmental enrichment reduces incubation of cocaine craving. They first trained rats to self-administer cocaine for 15 days (3 hours/day) and then either housed rats individually or exposed them to an enriched environment

(group housing, toys and running wheels, etc.) during the withdrawal period. They found that environmental enrichment reduced cue-induced cocaine seeking on both withdrawal days 1 and 21, but did not completely block incubation of cocaine craving. These behavioral data extend previous findings about the protective effect of environmental enrichment against cocaine reward, cocaine-induced behavioral sensitization, and cocaine seeking and reinstatement (Solinas *et al*. 2008; Chauvet *et al*. 2009; Thiel *et al*. 2011). Interestingly, the protective effect of environmental enrichment against incubation of cocaine craving dissipated quickly after enrichment is discontinued (Chauvet *et al*. 2012), suggesting that the underlying neural mechanisms (see discussion later) are reversible.

At the mechanistic level, Thiel *et al*. (2012) found that environmental enrichment attenuates the increased phosphorylation of extracellular signal-regulated kinases (pERK) in amygdala. This finding is consistent with previous studies on the critical role of central amygdala (CeA) ERK activity in incubation of cocaine craving (Lu *et al*. 2005). Although Thiel *et al*. (2012) performed pERK analysis in whole amygdala, their findings highlight the critical role of ERK activation in amygdala in incubation of cocaine craving. Furthermore, their findings suggest that environmental enrichment may attenuate incubation of cocaine craving via suppression of enhanced cocaine cue reactivity in amygdala.

Indeed, several previous studies have demonstrated that the CeA plays a critical role in incubation of cocaine and sucrose craving using self-administration procedure (Lu *et al*. 2005, 2007; Uejima *et al*. 2007), and incubation of morphine craving using a conditioned place preference procedure (Li *et al*. 2008). Our recent study (Li *et al*. in press) extended these previous findings and showed that reversible inactivation of CeA, but not basolateral amygdala (BLA) by the GABAa + GABAb agonists muscimol + baclofen, decreased incubation of methamphetamine craving. Taken together, current evidence indicates that CeA neuronal activation serves as a common substrate for incubation of craving of both drug and non-drug rewards. Thus, future studies on CeA-centered neural circuits may uncover common neural pathways that contribute to incubation of craving for both drug and non-drug rewards.

In another *Addiction Biology* report, Xi *et al*. (2013) examined the role of dopamine D3 receptor (D3R) in incubation of cocaine craving. They found that systemic injection of the D3R antagonist SB-277011A decreased cue-induced cocaine seeking on withdrawal days 2, 10 and 30. Moreover, injections of the D3R antagonist into nucleus accumbens (NAc) core or shell or central amygdala (but not dorsal striatum or basolateral amygdala) decreased incubation of cocaine craving after 3–4 weeks withdrawal from cocaine self-administration. These findings demonstrate that D3R activity in these brain areas play a critical role in cue-induced cocaine seeking independent of the drug withdrawal period.

Adaptations of glutamate receptor systems are another focus of mechanistic studies in incubation of cocaine craving. Most notably, in NAc synapses, incubation of cocaine craving is accompanied by accumulation of Ca^{2+} permeable GluA2-lacking AMPA receptors (CP-AMPARs), which exhibits higher conductance than Ca^{2+} impermeable GluA2-containing AMPARs (CI-AMPARs); blockade of CP-AMPARs in NAc attenuates incubation of cocaine craving (Conrad *et al*. 2008). Importantly, a short-access cocaine self-administration regimen that did not elicit incubation is not associated with accumulation of CP-AMPARs in NAc during the withdrawal period (Purgianto *et al*. 2013).

Recently, Wolf and colleagues have further explored the neurobiological mechanism responsible for the accumulation of CP-AMPARs in NAc after withdrawal from cocaine. In this study, Loweth *et al.* (2014) demonstrated that the accumulation of CP-AMPARs during extended withdrawal is controlled in part by downregulation of activity at the metabotropic glutamate receptor 1 (mGluR1). Restoring mGluR1 function, via either systemic or NAc injection of mGluR1 positive allosteric modulators (PAM: Ro67-7476 or SYN119), prevents the accumulation of CP-AMPARs in NAc and decreases incubation of cocaine craving. These findings support the hypothesis that mGluR1 transmission in NAc serves as a negative regulator of cocaine craving. One critical mechanism of this negative regulation is through opposing the accumulation of CP-AMPARs in NAc synapses. These results may have translational significance because repeated systemic treatment with mGluR1 PAM (SYN119) during the withdrawal period decreased incubated cue-induced cocaine seeking.

At the neural circuit level, Dong and colleagues recently explored the role of glutamatergic BLA-to-NAc and mPFC-to-NAc projections. Their analysis focused on silent synapses, which are excitatory synapses that contain NMDA receptors with absent or labile AMPARs (Lee *et al*. 2013; Ma *et al*. 2014). In the first study, Lee *et al*. (2013) used *in vitro* optogenetic stimulation and observed increased silent synapses in BLA projections to NAc shell on withdrawal day 1 but not withdrawal day 45. Moreover, the unsilencing process that occurs after withdrawal from cocaine is associated with the insertion of CP-AMPARs. Re-silencing BLA-to-NAc shell synapses, by *in vivo* optogenetic stimulation, caused downregulation of CP-AMPARs and also decreased incubation of cocaine craving. These results are consistent with previous findings on the causal role of CP-AMPARs in NAc in incubation of cocaine craving (Conrad *et al*. 2008).

In the second study, Ma *et al.* (2014) observed increased silent synapses in both infralimibic (IL) and prelimibic (PrL) mPFC to NAc on withdrawal day 1 but not day 45. However, while the unsilencing process of IL-to-NAc during incubation of cocaine craving is associated with recruitment of CP-AMPARs, the unsilencing process of PrL-to-NAc is through recruitment of non CP-AMPARs. Moreover, re-silencing IL-to-NAc projections increased cue-induced cocaine seeking and re-silencing PrL-to-NAc projection decreased it. These findings suggest that the contribution of silent synapses in NAc to incubation of cocaine craving is projection specific. Thus, an important question for future research is whether maturation of silent synapses after withdrawal from cocaine also occurs in other glutatmatergic projections to NAc (e.g. ventral subiculum and thalamus).

In contrast to enhanced glutamate transmission in NAc during incubation of cocaine craving, Counotte *et al.* (2014) recently showed that incubation of sucrose craving, observed in both adult and adolescent rats under their experimental conditions, is associated with time-dependent decreases in AMPA/NMDA ratio in NAc, reflecting reduced excitatory transmission. These findings suggest that potentiating glutamate transmission such as through accumulation of CP-AMPARs in NAc synapse during prolonged withdrawal may be specific to incubation of cocaine craving, and more generally, that the mechanisms of incubation of drug and non-drug rewards are not the same.

Additionally, recent studies also advanced our understanding of mechanisms underlying incubation of craving for other drugs such as heroin. Theberge *et al.* (2013) examined the role of toll-like receptor 4 (TLR4), innate immune receptors expressed primarily on microglia, in incubation of heroin craving. They found that chronic administration of TLR4 antagonist (+)-naltrexone, via osmotic mini-pump, for 14 days after withdrawal from heroin self-administration decreased incubation of heroin craving. Interestingly, acute administration prior to incubation tests had no effect on incubation of heroin craving. However, chronic delivery of (+)-naltrexone during the withdrawal period had no effect on incubation of methamphetamine craving. These findings reveal a critical role of TLR4 in incubation of heroin craving and suggest that dissociable mechanisms control incubation of craving for different drugs of abuse. In support of this notion, we recently found that reversible inactivation of ventral medial prefrontal cortex (previously implicated in incubation of cocaine craving; Koya *et al.* 2009) and orbitofrontal cortex (previously implicated in incubation of heroin seeking, Fanous *et al.* 2012) had no effect on incubation of methamphetamine craving (Li *et al.* in press).

At the behavioral level, Krasnova *et al.* (2014) designed a procedure to study incubation of methamphetamine seeking after abstinence induced by negative consequences, which is a common factor contributing to abstinence in humans. In their procedure, rats underwent a punishment phase after extended access of methamphetamine or palatable food self-administration. During the punishment phase, every second reward delivery was paired with mild footshock, and shock intensity was gradually increased across the sessions. Then, they examined cue-induced methamphetamine seeking on withdrawal day 2 and day 21 following the punishment phase and observed that incubation of craving occurred after both methamphetamine and palatable food self-administration. This procedure provides a new platform for exploring neural mechanisms underlying relapse to drug and palatable food under conditions that more closely model human abstinence.

Most studies described above have used extended access cocaine self-administration procedure. Earlier evidence demonstrates that incubation of cocaine craving in rats is more pronounced after extended access of cocaine self-administration (6 hours/day for 10 days) than after limited access (2 hours/day for 10 days; Lu *et al.* 2004). However, more recently, Sorge & Stewart (2005), Hollander & Carelli (2007), Lee *et al.* (2013) and Ma *et al.* (2014) have each shown that incubation of cocaine craving is also reliably observed after limited access (2 hours daily sessions) training conditions. Most recently, Halbout *et al.* (2014) probed the minimum amount of cocaine self-administration experience needed to develop incubation of cocaine craving. The authors showed that a single extended access cocaine self-administration session (6 hours) led to incubation of cocaine craving in mice. This result questions the conventional idea that incubation of drug craving in rodent is critically dependent on the extended history of drug self-administration. Halbout *et al.* (2014) examined neuroadaptations associated with incubation after a single session. They found several similarities compared with previous findings about incubation after extended access (Conrad *et al.* 2008; Loweth *et al.* 2014). For example, they showed that AMPA receptor availability is significantly increased in NAc shell on withdrawal day 43. They also found a persistent suppression of mGluR1 mRNA expression in NAc during both early (day 9) and late withdrawal (day 43). More importantly, systemic administration of the mGluR1 antagonist (A-841720) during the first 7 days of withdrawal from a single day of cocaine self-administration enhanced cue-induced cocaine seeking on withdrawal day 43.

The observation of similar neuroadaptations after either single or extended access raise intriguing questions about the amount of cocaine experience that is necessary to trigger neuroadaptations associated with incubation of

cocaine craving. The Halbout *et al.* (2014) study provides additional support for the possibility that extended access, and subsequent escalation of drug intake, is not a determining factor for incubation of cocaine craving. Instead, these data imply that the neuroadaptations responsible for the persistence of drug seeking during abstinence, which potentially make drug addiction a chronically relapsing condition, are initiated after limited amounts of drug exposure. This has significant implications for our understanding of drug addiction because it suggests that the incubation phenomenon has the potential to contribute to the development of drug addiction during the early stages of drug use.

Supporting the above notion, Guillem *et al.* (2014) recently demonstrated that escalation of cocaine intake and incubation of cocaine craving in rats are dissociable at both the behavioral and neuronal levels. At the behavioral level, they observed escalation of cocaine intake during cocaine self-administration training. On withdrawal day 30, the rats exhibited 'incubated' drug seeking in the extinction test. However, on the same day, escalation of cocaine intake was no longer present when the rats were allowed to self-administer cocaine. At the neuronal level, incubation of cocaine craving was associated with increased neuronal activity in NAc core neurons that encode (time-locked phasic firing) cue-induced responses during incubation tests. In contrast, the absent escalation of cocaine intake after 30 withdrawal days was selectively associated with normalization of depressed NAc shell activity, which developed during prior escalation of cocaine intake.

In conclusion, we summarized results from several recent clinical and preclinical studies on incubation of craving of both drug and non-drug rewards. These recent findings confirm that incubation of drug craving occurs in human addicts. At the mechanistic level, results demonstrate a critical role of CeA in incubation methamphetamine craving, the identification of novel glutamatergic-related neuroadaptations in NAc and its efferent projections that contribute to incubation of cocaine craving. At the behavioral level, recent findings include the inhibitory effect of environmental enrichment on incubation of cocaine craving, incubation of methamphetamine and palatable food craving after punishment and the surprising demonstration of robust incubation of cocaine craving after a single day of cocaine self-administration. Lastly, a recent study demonstrates behavioral and neuronal dissociations between incubation of cocaine craving and escalation of cocaine intake.

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References

Abdolahi A, Acosta G, Breslin FJ, Hemby SE, Lynch WJ (2010) Incubation of nicotine seeking is associated with enhanced protein kinase A-regulated signaling of dopamine- and cAMP-regulated phosphoprotein of 32 kDa in the insular cortex. Eur J Neurosci 31:733–741.

Bedi G, Preston KL, Epstein DH, Heishman SJ, Marrone GF, Shaham Y, de Wit H (2011) Incubation of cue-induced cigarette craving during abstinence in human smokers. Biol Psychiatry 69:708–711.

Bienkowski P, Rogowski A, Korkosz A, Mierzejewski P, Radwanska K, Kaczmarek L, Bogucka-Bonikowska A, Kostowski W (2004) Time-dependent changes in alcohol-seeking behaviour during abstinence. Eur Neuropsychopharmacol 14:355–360.

Chauvet C, Lardeux V, Goldberg SR, Jaber M, Solinas M (2009) Environmental enrichment reduces cocaine seeking and reinstatement induced by cues and stress but not by cocaine. Neuropsychopharmacology 34:2767–2778.

Chauvet C, Goldberg SR, Jaber M, Solinas M (2012) Effects of environmental enrichment on the incubation of cocaine craving. Neuropharmacology 63:635–641.

Conrad KL, Tseng KY, Uejima JL, Reimers JM, Heng LJ, Shaham Y, Marinelli M, Wolf ME (2008) Formation of accumbens GluR2-lacking AMPA receptors mediates incubation of cocaine craving. Nature 454:118–121.

Counotte DS, Schiefer C, Shaham Y, O'Donnell P (2014) Time-dependent decreases in nucleus accumbens AMPA/NMDA ratio and incubation of sucrose craving in adolescent and adult rats. Psychopharmacology (Berl) 231:1675–1684.

Fanous S, Goldart EM, Theberge FR, Bossert JM, Shaham Y, Hope BT (2012) Role of orbitofrontal cortex neuronal ensembles in the expression of incubation of heroin craving. J Neurosci 32:11600–11609.

Gawin F, Kleber H (1986) Pharmacologic treatments of cocaine abuse. Psychiatr Clin North Am 9:573–583.

Grimm JW, Hope BT, Wise RA, Shaham Y (2001) Neuroadaptation. Incubation of cocaine craving after withdrawal. Nature 412:141–142.

Grimm JW, Shaham Y, Hope BT (2002) Effect of cocaine and sucrose withdrawal period on extinction behavior, cue-induced reinstatement, and protein levels of the dopamine transporter and tyrosine hydroxylase in limbic and cortical areas in rats. Behav Pharmacol 13:379–388.

Guillem K, Ahmed SH, Peoples LL (2014) Escalation of cocaine intake and incubation of cocaine seeking are correlated with dissociable neuronal processes in different accumbens subregions. Biol Psychiatry 76:31–39.

Halbout B, Bernardi RE, Hansson AC, Spanagel R (2014) Incubation of cocaine seeking following brief cocaine experience in mice is enhanced by mGluR1 blockade. J Neurosci 34:1781–1790.

Hollander JA, Carelli RM (2007) Cocaine-associated stimuli increase cocaine seeking and activate accumbens core neurons after abstinence. J Neurosci 27:3535–3539.

Koya E, Uejima JL, Wihbey KA, Bossert JM, Hope BT, Shaham Y (2009) Role of ventral medial prefrontal cortex in incubation of cocaine craving. Neuropharmacology 56 (Suppl. 1):177–185.

Krasnova IN, Marchant NJ, Ladenheim B, McCoy MT, Panlilio LV, Bossert JM, Shaham Y, Cadet JL (2014) Incubation of methamphetamine and palatable food craving after punishment-induced abstinence. Neuropsychopharmacology 39:2008–2016.

Lee BR, Ma YY, Huang YH, Wang X, Otaka M, Ishikawa M, Neumann PA, Graziane NM, Brown TE, Suska A, Guo C, Lobo MK, Sesack SR, Wolf ME, Nestler EJ, Shaham Y, Schluter OM, Dong Y (2013) Maturation of silent synapses in amygdala-accumbens projection contributes to incubation of cocaine craving. Nat Neurosci 16:1644–1651.

Li P, Wu P, Xin X, Fan YL, Wang GB, Wang F, Ma MY, Xue MM, Luo YX, Yang FD, Bao YP, Shi J, Sun HQ, Lu L (2014) Incubation of alcohol craving during abstinence in patients with alcohol dependence. Addict Biol doi: 10.1111/adb.12140.

Li X, Zeric T, Kambhampati S, Bossert JM, Shaham Y (in press) The central amygdala nucleus is critical for incubation of methamphetamine craving. Neuropsychopharmacology.

Li YQ, Li FQ, Wang XY, Wu P, Zhao M, Xu CM, Shaham Y, Lu L (2008) Central amygdala extracellular signal-regulated kinase signaling pathway is critical to incubation of opiate craving. J Neurosci 28:13248–13257.

Loweth JA, Scheyer AF, Milovanovic M, LaCrosse AL, Flores-Barrera E, Werner CT, Li X, Ford KA, Le T, Olive MF, Szumlinski KK, Tseng KY, Wolf ME (2014) Synaptic depression via mGluR1 positive allosteric modulation suppresses cue-induced cocaine craving. Nat Neurosci 17:73–80.

Lu L, Grimm JW, Dempsey J, Shaham Y (2004) Cocaine seeking over extended withdrawal periods in rats: different time courses of responding induced by cocaine cues versus cocaine priming over the first 6 months. Psychopharmacology (Berl) 176:101–108.

Lu L, Hope BT, Dempsey J, Liu SY, Bossert JM, Shaham Y (2005) Central amygdala ERK signaling pathway is critical to incubation of cocaine craving. Nat Neurosci 8:212–219.

Lu L, Uejima JL, Gray SM, Bossert JM, Shaham Y (2007) Systemic and central amygdala injections of the mGluR(2/3) agonist LY379268 attenuate the expression of incubation of cocaine craving. Biol Psychiatry 61:591–598.

Ma YY, Lee BR, Wang X, Guo C, Liu L, Cui R, Lan Y, Balcita-Pedicino JJ, Wolf ME, Sesack SR, Shaham Y, Schluter OM, Huang YH, Dong Y (2014) Bidirectional modulation of incubation of cocaine craving by silent synapse-based remodeling of prefrontal cortex to accumbens projections. Neuron 83:1453–1467.

Marchant NJ, Li X, Shaham Y (2013) Recent developments in animal models of drug relapse. Curr Opin Neurobiol 23:675–683.

Neisewander JL, Baker DA, Fuchs RA, Tran-Nguyen LT, Palmer A, Marshall JF (2000) Fos protein expression and cocaine-seeking behavior in rats after exposure to a cocaine self-administration environment. J Neurosci 20:798–805.

Pickens CL, Airavaara M, Theberge F, Fanous S, Hope BT, Shaham Y (2011) Neurobiology of the incubation of drug craving. Trends Neurosci 34:411–420.

Purgianto A, Scheyer AF, Loweth JA, Ford KA, Tseng KY, Wolf ME (2013) Different adaptations in AMPA receptor transmission in the nucleus accumbens after short versus long access cocaine self-administration regimens. Neuropsychopharmacology 38:1789–1797.

Shalev U, Morales M, Hope B, Yap J, Shaham Y (2001) Time-dependent changes in extinction behavior and stress-induced reinstatement of drug seeking following withdrawal from heroin in rats. Psychopharmacology (Berl) 156:98–107.

Shepard JD, Bossert JM, Liu SY, Shaham Y (2004) The anxiogenic drug yohimbine reinstates methamphetamine seeking in a rat model of drug relapse. Biol Psychiatry 55:1082–1089.

Solinas M, Chauvet C, Thiriet N, El Rawas R, Jaber M (2008) Reversal of cocaine addiction by environmental enrichment. Proc Natl Acad Sci U S A 105:17145–17150.

Sorge RE, Stewart J (2005) The contribution of drug history and time since termination of drug taking to footshock stress-induced cocaine seeking in rats. Psychopharmacology (Berl) 183:210–217.

Theberge FR, Li X, Kambhampati S, Pickens CL, St. Laurent R, Bossert JM, Baumann MH, Hutchinson MR, Rice KC, Watkins LR, Shaham Y (2013) Effect of chronic delivery of the toll-like receptor 4 antagonist (+)-naltrexone on incubation of heroin craving. Biol Psychiatry 73:729–737.

Thiel KJ, Engelhardt B, Hood LE, Peartree NA, Neisewander JL (2011) The interactive effects of environmental enrichment and extinction interventions in attenuating cue-elicited cocaine-seeking behavior in rats. Pharmacol Biochem Behav 97:595–602.

Thiel KJ, Painter MR, Pentkowski NS, Mitroi D, Crawford CA, Neisewander JL (2012) Environmental enrichment counters cocaine abstinence-induced stress and brain reactivity to cocaine cues but fails to prevent the incubation effect. Addict Biol 17:365–377.

Tran-Nguyen LT, Fuchs RA, Coffey GP, Baker DA, O'Dell LE, Neisewander JL (1998) Time-dependent changes in cocaine-seeking behavior and extracellular dopamine levels in the amygdala during cocaine withdrawal. Neuropsychopharmacology 19:48–59.

Uejima JL, Bossert JM, Poles GC, Lu L (2007) Systemic and central amygdala injections of the mGluR2/3 agonist LY379268 attenuate the expression of incubation of sucrose craving in rats. Behav Brain Res 181:292–296.

Wang G, Shi J, Chen N, Xu L, Li J, Li P, Sun Y, Lu L (2013) Effects of length of abstinence on decision-making and craving in methamphetamine abusers. PLoS ONE 8:e68791.

Xi Z-X, Li X, Li J, Peng X-Q, Song R, Gaál J, Gardner EL (2013) Blockade of dopamine D3 receptors in the nucleus accumbens and central amygdala inhibits incubation of cocaine craving in rats. Addict Biol 18:665–677.

