Journal Club

Editor's Note: These short, critical reviews of recent papers in the *Journal*, written exclusively by graduate students or postdoctoral fellows, are intended to summarize the important findings of the paper and provide additional insight and commentary. For more information on the format and purpose of the Journal Club, please see http://www.jneurosci.org/misc/ifa_features.shtml.

Social Stress-Induced Alterations in CRF Signaling in the VTA Facilitate the Emergence of Addiction-like Behavior

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Review of Holly et al.

Stress increases the vulnerability to addiction in humans (for review, see Sinha, 2008). In preclinical studies, stressors can enhance drug self-administration and induce relapse (Miczek et al., 2008; Mantsch et al., 2016). The brain region-specific molecular mechanisms that mediate the interaction between stress and cocaine self-administration are multifaceted, and their elucidation is a work in progress (Lu et al., 2003; Miczek et al., 2008).

One potential mediator is corticotropin-releasing factor (CRF), a neuropeptide associated both with behavioral and physiological responses to stress (Vale et al., 1981; Sutton et al., 1982) and with escalation of drug self-administration (Koob et al., 2014). CRF binds to CRF₁ and CRF₂ receptors and acts on the hypothalamic-pituitary-adrenal axis, as well as extrahypothalamic areas, including the amygdala, bed nucleus of stria terminalis, and ventral tegmental area (VTA) (Bale and Vale, 2004). CRF signaling in the VTA is a particularly compelling candidate mechanism because drugs of abuse and stressful/aversive stimuli generate simi-

lar activation in the VTA. Furthermore, recent work has shown that CRF₁ and CRF₂ receptors are involved in the VTA response to social stress (Holly et al., 2015) and that this response facilitates later cocaine “binge” self-administration (i.e., increased cocaine taking when rats are given 24 h of unlimited drug access) (Boyson et al., 2014).

A recent study in *The Journal of Neuroscience* by Holly et al. (2016) tested the hypothesis that stress-induced CRF signaling in the VTA drives cocaine addiction-like behaviors in a VTA subregion and CRF receptor subtype-specific manner. The VTA is a morphologically and functionally heterogeneous structure (Ikemoto, 2007), with the posterior (pVTA) and anterior (aVTA) regions differentially required both in the case of reinforcement (e.g., drugs of abuse) and aversion (e.g., footshock) (Holly and Miczek, 2016). Additionally, there are conflicting reports in the literature on the involvement of specific CRF receptor subtypes within the VTA in the interaction between stress and cocaine addiction-like behaviors. For example, some have reported that CRF₁, but not CRF₂, receptors are required for stress-induced reinstatement of cocaine seeking (Blacktop et al., 2011), whereas others have found the opposite (Wang et al., 2007). These discrepant findings may be due to functional heterogeneity of CRF signaling in the VTA along the anterior/posterior axis.

In Experiment 1, Holly et al. (2016) used *in vivo* microdialysis to investigate how VTA CRF levels change over the course of repeated exposure to social defeat stress. Dur-

ing the first session, CRF levels in the pVTA, but not aVTA, increased dramatically during stress and remained elevated afterward. By the fourth session, however, CRF in the aVTA was significantly elevated during stress, whereas pVTA CRF decreased following stress. Furthermore, both the aVTA and pVTA had increased basal CRF release by the fourth session of social stress. These results show that the CRF response in the VTA to social stress is dynamically regulated (ON/OFF changes in signaling within 1 h), enduring (higher basal CRF after repeated stress), and adjusted in a subregion-specific manner over repeated trials (the aVTA appears to be recruited with repeated episodes, whereas the pVTA response is blunted).

In Experiment 2, the authors demonstrated that socially stressed rats administered significantly more cocaine during a 24 h binge session compared with non-stressed controls. The effect of stress was notably long-lasting, as the self-administration test was conducted at least 30 d after stress. The authors observed a double dissociation between the requirements for specific CRF receptor subtypes along the anterior/posterior VTA axis. CRF₁ receptors in the pVTA, but not aVTA, and CRF₂ receptors in the aVTA, but not pVTA, were necessary for increased cocaine intake in stressed rats.

In Experiment 3, the authors showed that persistent changes in VTA CRF signaling in previously stressed rats are necessary for long-term stress-induced facilitation of cocaine seeking. Similar to Experiment 2, CRF₁ receptors in the pVTA, but not aVTA, and CRF₂ receptors in the aVTA, but not

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pVTA, were required for increased cocaine seeking following 15 d of cocaine abstinence. Previously stressed rats had enhanced tonic pVTA and aVTA CRF signaling compared with nonstressed groups, immediately before and throughout the cocaine-seeking test. Interestingly, neither stressed nor nonstressed rats exhibited phasic changes in VTA CRF during the test. The enhanced cocaine-seeking behavior observed in previously stressed rats may reflect a stable, persistent state mediated by chronically elevated VTA CRF signaling induced by stress/cocaine exposure rather than one that was activated in response to the drug-seeking test. Consistent with the notion that propensity to seek cocaine is a stable state, in a choice-reinstatement procedure, a strong correlation was found between preference for previously cocaine- and food-reinforced levers over the course of extinction responding and during reinstatement tests that were conducted in subsequent weeks (Tunstall and Kearns, 2014).

The findings of Holly et al. (2016) raise the question: how does increased CRF release during and after stress lead to increased cocaine taking and seeking? CRF is critically involved in compensatory opponent or “rebound” mechanisms that are engaged in an effort to counteract the effects of cocaine following intense/prolonged drug self-administration (Koob and Le Moal, 2008). Enhanced CRF signaling contributes to the aversive state experienced in cocaine withdrawal (opposite the positive hedonic state experienced during drug intoxication), which may motivate a subject to seek and take more cocaine to alleviate negative affect (i.e., negative reinforcement). We hypothesize that, when subjects with already elevated CRF levels (i.e., previously stressed rats) self-administer sufficient amounts of cocaine, cessation of cocaine taking produces an augmented aversive state, which facilitates addiction-like behavior.

Enhanced CRF could contribute to an aversive state driving addiction-like behaviors via modulation of mesolimbic dopamine function. CRF can modulate VTA dopamine neurons (Ungless et al., 2003; Wang et al., 2005) and can enhance dopamine signaling in the nucleus accumbens, similarly to drugs of abuse (Deutch and Roth, 1990). It is possible that CRF could sensitize mesolimbic dopamine function, which would be expected to enhance the positive rewarding properties of cocaine. However, VTA CRF can also decrease dopamine release in the nucleus accumbens, depending on the upstream input to the VTA (Wanat et al., 2013; Grieder et al., 2014). Furthermore, CRF injected intracerebroventricularly can decrease brain stimula-

tion reward (Macey et al., 2000). It remains unclear whether altered CRF signaling in the VTA observed by Holly and Miczek (2016) affected VTA dopamine release or mesolimbic reward function. It is also possible that enhanced CRF contributes to an augmented negative affective state through dopamine-independent, prostress effects. Critical experiments to extend the present results would be to assess alterations in VTA dopamine signaling (e.g., using *in vivo* microdialysis) and mesolimbic reward function (e.g., using intracranial self-stimulation) during the critical stages of the present experimental design, particularly following binge-access to cocaine and during cocaine abstinence. We predict that previously stressed rats would be more hypohedonic and display more blunted mesolimbic dopamine function than nonstressed controls following this cocaine exposure.

The role of CRF signaling during repeated social-defeat stress and later cocaine self-administration behavior is compelling. Future experiments could directly test the hypothesis that repeated social stress strengthens opponent processes activated by cocaine. This could be achieved by comparing previously nonstressed versus stressed groups in the strength of their response to the delayed, aversive effects of cocaine injections (Su et al., 2013; Zhou et al., 2014). If stress sensitizes the compensatory mechanisms activated by cocaine (i.e., CRF signaling), we would expect previously stressed rats to demonstrate an enhanced aversion response. The contracted timescale of a place aversion design may also be useful in future experiments designed to explore neurobiological mechanisms that may buffer against or facilitate the CRF response to stress. For example, oxytocin is a candidate buffer mechanism, having antistress properties that may be mediated in part by interference with stress-induced CRF transcription (Jurek et al., 2015). In contrast, vasopressin is a candidate facilitation mechanism, as vasopressin is known to be potentially synergistic with CRF in causing release of adrenocorticotrophic hormone (Gillies et al., 1982). Vasopressin has also been shown to be upregulated in response to repeated stress, resulting in increased colocalization of vasopressin in CRF neurons in the median eminence (de Goeij et al., 1991) and paraventricular nucleus (de Goeij et al., 1992).

Finally, if we consider cocaine self-administration, particularly the negative emotional experience associated with the engagement of opponent processes, as a stressful experience, then we may consider the results of Holly et al. (2016) as a charac-

terization of the long-lasting “priming” of a stress signaling system. In the case of social defeat, we may observe the effect of enhanced stress signaling through enhanced motivation to avoid or escape a stressful situation. In the case of cocaine self-administration, we would expect the effects of social stress to combine with opponent processes stress-signaling, further enhancing the motivation to seek and self-administer cocaine.

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