

Sex differences in limbic circuit modulation of alcohol drinking and anxiety

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Binge alcohol drinking is the most common form of excessive alcohol consumption and a leading risk factor for the development of alcohol use disorder, anxiety, and other stress-related mood disorders. The comorbid expression of these neuropsychiatric diseases is higher in women than men, however the reason for this increased vulnerability is unknown. Female mice binge drink more and have greater basal anxiety than males, and these behaviors are driven by neurons that synthesize the neuropeptide corticotropin-releasing factor (CRF) in the bed nucleus of the stria terminalis (BNST), a sexually-dimorphic limbic brain region. In order to determine whether the excitability of BNST CRF neurons provide a mechanism for the observed sex difference in behavior, we performed *ex vivo* slice electrophysiology in CRF-reporter mice following three cycles of binge alcohol (or water control) drinking. We show that BNST CRF neurons were more likely to be tonically active at baseline in females than males and that repeated binge drinking increased the proportion of active neurons in both sexes—male neurons from alcohol-exposed mice adopted a “female-like” baseline phenotype, at a time point when escalation in alcohol intake to baseline female levels emerges, and female neurons became even more excitable. In addition, BNST CRF neurons from female alcohol-exposed mice had a lower threshold for firing than other groups, which may account for their further excitability. Interestingly, recordings from BNST neurons of chronic alcohol drinking rhesus monkeys showed similar alterations in excitability, suggesting that this is a translationally relevant neuronal mechanism that may be related to the increased risk for comorbid alcohol use disorder and anxiety in women compared to men.

Using neuronal tracing and *ex vivo* optogenetics + slice electrophysiology in mice, we also mapped a dense, direct projection of glutamatergic neurons from the paraventricular nucleus of the thalamus (PVT) to the BNST that produced a monosynaptic excitatory but larger polysynaptic inhibitory input to BNST CRF neurons. The synaptic input from the PVT to BNST CRF neurons was larger in males than females, suggesting that the PVT provides less of a “break” on BNST CRF neurons in females leading to increased neuronal activity. We hypothesize that decreased excitability of BNST-projecting PVT neurons disinhibits BNST CRF neurons to drive binge drinking and stress reactivity, conferring increased risk of comorbid addiction and anxiety disorders in females and increased susceptibility to these diseases in males following chronic alcohol exposure. Current studies are examining the effects of chronic alcohol drinking on these PVT neurons and their synaptic inputs to the BNST in mice and monkeys. We are also examining the role of estrogen signaling within this circuit on the neuronal function and plasticity related to these behaviors and neuropsychiatric disease states.

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