Insula-Bnst Circuit Regulation of Stress-Induced Susceptibility to Negative Affect in Ethanol Abstinence

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The concept of "preaddiction" is under consideration to assess and quantify the risk of developing a substance use disorder. Given the prevalence of problematic alcohol use and the transition to alcohol use disorder (AUD) in some individuals, early intervention is crucial. Addiction involves complex overlapping dysfunctional patterns, making it difficult to pinpoint cause and effect, or quantifiable biomarkers. To overcome this, we must understand how behavioral traits relate to underlying neuroadaptations. Neurocircuits are the basic functional units that encode behavior. Could discrete neurocircuit signatures engaged by AUD-related behaviors represent signatures that predict AUD susceptibility?

The insula, a hub of the salience network involved in interoception, decision-making, stress response, and reward processing, is a promising target for pre-AUD. Threat-, stress-, and alcohol cue-induced insula activation are linked to future alcohol-drinking behavior. We and others have defined a role for the insula projection to the extended amygdala, specifically the BNST, in regulating stress-coping behavior, binge drinking, and negative affective behaviors during abstinence. The clear intertwined connectivity between the insula and BNST, and an emerging role for this pathway in negative affect warrants studying this circuitry as a potential pre-AUD imaging locus.

To begin, we tested the effects of chronic restraint stress on ethanol drinking and abstinence-induced negative affect in a chronic drinking-forced abstinence (CDFA) model in C57BL/6J mice. Stress did not impact ethanol drinking, however, it did induce higher negative affect-like behavior in abstinence. Interestingly, this effect was driven by ~50% of mice, suggesting two population clusters- one susceptible and one resilient to stress before ethanol drinking. Behavioral performance in abstinence in susceptible mice was positively correlated with ethanol consumption and stress-coping behavior, revealing a direct parallel between basal stress response, drinking patterns, and abstinence-induced negative affect. Notably, these effects were specific to female mice, outlining unique sex differences in stress susceptibility.

We hypothesized that stress-induced recruitment of insula-BNST pathway activity underlies susceptibility to abstinence-induced negative affect. Chemogenetically inhibiting insula-BNST neurons with hM4Di 1h before each stress exposure prevented the emergence of a stress susceptible population in abstinence. Insula-BNST neurons may differentially integrate the valence of a stressor, an effect that can persist and reemerge as increased negative affect-like behavior in abstinence. Next, we used in vivo fiber photometry with the GCaMP calcium indicator to measure the relationship between insula-BNST activity during stress, and subsequent drinking patterns and negative affect in abstinence. Peak GCaMP amplitude time-locked to an active coping bout during restraint stress was negatively correlated with ethanol consumption during CDFA, and positively correlated with latency to feed on NSFT in protracted abstinence.

These findings suggest the insula-BNST may encode susceptibility to alcohol abstinence-induced hyperkatifeia. Stress-induced recruitment of the insula-BNST pathway relates to future drinking behavior and negative affect-like behavior in abstinence, and disengaging the circuit during stress exposure prevents negative affect in abstinence. Current studies are examining the unique transcriptomic and neurophysiological signatures of insula-BNST neurons. Shifting strategies away from reactive treatments, and towards preventative measures to mitigate risks of AUD is essential.