Name: Christopher Cowan Email: cowanc@musc.edu

# HDAC5 intrinsic enzymatic activity limits drug-seeking behavior

Daniel Wood[1], Evgeny Tsvetkov[1], Susana Comte-Walters[2], Lauren Ball[2], Ethan Anderson[1], Makoto Taniguchi[1], Christopher Cowan[1]

[1]Department of Neuroscience Medical University of South Carolina; [2]Department of Cell and Molecular Pharmacology and Experimental Therapeutics Medical University of South Carolina

Repeated use of illicit drugs produces long-lasting associations between the drug experience and environmental features through stable neuroadaptations in the brain's reward circuitry. Drug-cue associations can serve as potent motivators of drug seeking in abstinent individuals long after drug cessation. In rodents, the epigenetic enzyme histone deacetylase 5 (HDAC5) functions in the nucleus accumbens (NAc) during active drug use to limit future cue-induced drug seeking. HDAC5 shuttles steadily between the cytoplasm and the nucleus, but cocaine and heroin produce a nuclear accumulation of HDAC5 that limits drug-cue associations. In the nucleus, HDAC5 represses numerous target genes, but HDAC5's intrinsic deacetylase activity is much lower than class I HDACs leading some to propose that class IIa HDACs, like HDAC5, function largely as protein scaffolds for recruitment of class I HDACs, like HDAC3, to genomic sites. Using tandem mass spectrometry, we observed that two conserved cysteines within HDAC5's enzymatic domain form an intramolecular disulfide bond *in vitro* and *in vivo*. Mutation of these cysteines abolishes HDAC5 deacetylase activity without disrupting HDAC3 binding. Unlike enzyme-active nuclear HDAC5, viral-mediated expression of the deacetylase-dead HDAC5 in the adult rat NAc fails to reduce NAc medium spiny neuron intrinsic excitability, a recently identified candidate mechanism by which HDAC5 limits drug-cue memory formation or stability. These data support a novel role for the intrinsic enzymatic activity of HDAC5 in decreasing relapse-like behavior, possibly through the modulation of chromatin structure and expression of genes linked to intrinsic excitability.