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Optically Induced CRISPR Tool for Epigenome Editing in Cocaine Action

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Drug abuse is a debilitating chronic disease which is a leading cause of disability around the world. Studies have shown that, at the molecular level, repeated exposure of drugs, such as cocaine, leads to changes in epigenetic processes, including nucleosome modifications, and alter cocaine-induced behavior. However, the lack of spatiotemporal precise tools to induce histone modifications is a major barrier in studying the complex molecular and behavioral dynamics occurring during the addictive process. In this study, we developed an optically induced CRISPR mediated histone modification tool that can be targeted to precise loci in selective neuron subtypes using the CRY2-CIBN blue light heterodimerizing complex system. These novel constructs are packaged into AAVs for *in vivo* delivery into the brain. In cell culture, our novel constructs, which code for catalytically dead (d)Cas9-CIBN and CRY2-KDM1A fusion proteins on a Cre dependent backbone, were co-transfected into Neuro2A cells along with sgRNA targeting for early growth response 3 (Egr3) and NGFIA-binding protein 2 (Nab2). After 1 hour of blue light exposure, Egr3 mRNA level was significantly down regulated in cells transfected with the sgRNA targeting Egr3. Consistent with this, Nab2 mRNA level was significantly down regulated in cells with Nab2 targeting sgRNA. Interchanging with CRY2-p300, which contains the truncated functional core of histone acetylation enzyme p300 fused with CRY2, Egr3 and Nab2 mRNA levels were significantly increased with sgRNA targeted for Egr3 and Nab2, respectively, after 1 hour or 4 hours of blue light exposure.