Cellular Mechanisms Underlying the Opioid Modulation of the Thalamo-cortico-striatal Circuit

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The ongoing opioid crisis calls for an in-depth understanding of opioid actions at the cellular and circuit levels. The striatum integrates excitatory inputs from the interconnected cortex and thalamus to form a triangular circuit loop that mediates motor control, affective pain, decision-making, and reward. Opioids impose strong modulation of this circuit. However, where and how opioids act are not fully understood. We aim to elucidate cellular and subcellular mechanisms underlying opioid modulation of the individual elements within the thalamo-cortico-striatal circuit and how these modulations integrate to alter the function of the entire circuit. Towards this goal, in addition to electrophysiological pharmacology, we have developed novel imaging capability to directly visualize the subcellular cAMP/PKA signaling downstream of opioids in living tissue.

Methods: Imaging experiments were performed using custom-built two-photon fluorescent lifetime imaging microscopy (2pFLIM). Genetically encoded cAMP/PKA indicators were developed with protein engineering and protein targeting strategies. Viral reagents were generated. For acute slice experiments, brain slices (300 μm) were obtained from virally injected mice. Whole-cell patch clamp recordings were used to characterize the physiological properties of the thalamo-cortico-striatal circuits. For in vivo 2pFLIM imaging, craniotomy was performed for cortical imaging experiments, and GRIN lens were used for subcortical imaging experiments.

Results: We found that MOR agonists potently inhibited thalamic inputs to the individual striatal medium spiny neurons. MOR activation also inhibited thalamic inputs to the pyramidal neurons in the anterior cingulate cortex (ACC). In contrast, DOR agonists disinhibited ACC pyramidal neuron responses to thalamic inputs by suppressing local feed-forward GABA signaling from parvalbumin-positive interneurons. To further dissect the underlying cellular and subcellular mechanisms, we generated novel PKA indicators, called tAKARα that can report intracellular opioids responses. 2pFLIM imaging of tAKARα can track bidirectional PKA activities in individual neurons in awake mice. We observed compartmentalized PKA responses to opioids that are both cell-type specific and opioid receptor specific in the thalamo-cortico-striatal circuit.

Discussions: The specific functional consequences of individual opioid receptors will be discussed. The novel imaging modality to investigate opioid-induced signal dynamics may fill a critical knowledge gap regarding the differential signaling mechanisms underlying the action of different opioids, and provide a novel readout for 'biased signaling' of specific opioid agonists.

References:

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