Neuronal circuit interrogation by whole-brain calcium imaging and real-time data-driven optical manipulation at the single-cell level

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We developed a microscopy framework that integrates multi-view light-sheet imaging of the nervous system [1-3] with cellular-resolution optical manipulation using wide-field patterned illumination and three-dimensional multi-photon excitation. Furthermore, by developing a computational control layer that analyzes and classifies calcium imaging data in real time, we designed experiments that facilitate real-time image-based behavior detection and closed-loop optogenetic manipulation of neuronal activity during behavior.

We demonstrated the utility of this framework by dissecting motor programs in larval *Drosophila*. We performed high-resolution calcium imaging of entire central nervous system (CNS) explants and detected the execution of different motor programs in real time. By using this real-time behavior classification to instruct the timing of optogenetic intervention, we systematically targeted different subsets of neurons involved in motor control and determined their role in executing motor behaviors and modulating the state of the nervous system. Through this concurrent use of whole-CNS functional imaging, real-time behavior classification and precise, data-driven optogenetic intervention, we discovered a state-dependency in the behavioral choices available to the nervous system and systematically mapped neuronal activity patterns across the CNS underlying switches between these states.

We furthermore employed our method to perturb functionally-defined populations of neurons throughout the brain of larval zebrafish using two-photon optogenetic stimulation while measuring the brainwide effects of these perturbations with whole-brain functional imaging. These experiments show that concurrent whole-brain activity and causality mapping in the same animal enables delineating the contributions of neurons to brainwide circuit dynamics and behavior.

References:

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