Long-term calcium imaging of distinct neuronal populations in the mouse olfactory bulb

Yoav Adam and Adi Mizrahi

Department of Neurobiology, Inst. for Life Sciences and The Edmond and Lily Safra Center for Brain Sciences, The Hebrew University of Jerusalem, Edmond J. Safra Campus, Givat Ram, Jerusalem, 91904, Israel.

Fine plastic events in the brain are hard to measure because they may be masked by the awesome heterogeneity in neuronal networks as well as by inter-animal variability. Studying neuronal plasticity has benefited tremendously from recent developments in time lapse approaches. For example, time lapse *in vivo* two-photon imaging of dendrites and spines revealed the dynamic nature of synapses in adult brains. But how do neurons maintain their functional response profiles in the face of continuous structural dynamics? Or do they? This question is of particular interest in the mammalian olfactory bulb, the first station of olfactory processing in the brain, because it receives constant supply of new neurons in the process of adult neurogenesis. These neurons integrate into the existing network, causing continuous structural plasticity with unknown functional significance. Here we follow the response profiles of distinct neuronal populations in the OB repeatedly over many weeks. We selectively targeted projection neurons (mitral and tufted cells) or local neurons (periglomerular cells) using viruses to induce expression of the genetically encoded calcium indicator GCaMP3.0. As assessed by immunohistochemistry, lentivirus-GCaMP3.0 infected selectively mitral and tufted cells. In contrast, AAV-GCaMP3.0 was expressed exclusively in interneurons. We then combined viral injections with a chronic window preparation in order to follow the response profiles of single neurons using two-photon imaging. Basal odor responses were imaged repeatedly from distinct populations of neurons, revealing unique response signatures for different neuronal subtypes. Repeated imaging of the same neurons over several weeks revealed remarkable stability in the response profiles of neurons to a small set of odors and concentrations. This preparation is currently used to test whether and how neuronal profiles change in the OB following simple associative learning tasks. During the tenure of Japan-Israeli grant we will use this method to assess the functional changes in the network following genetic depletion of the neuronal stem cells niche (in collaboration with Prof. Kageyama).