

The purpose of translaminar imaging of olfactory cortex is to show that stimuli or stimulus features are differentially represented by different layers or by different groups of neurons within each layer. Depending on what we find, the goal is to suggest how these differences may be exploited downstream of the piriform or suggest a local role for interaction between layers in generating odor-evoked activity.

In functional studies of PIR cortex, LII/III is typically treated as a single entity despite evidence to the contrary from hodological and in vitro work. This work suggests that odors engage circuits that span LII/LIII primarily via LOT Ia. However, input from frontal cortical areas like OFC and Insula, as well as input from other limbic areas impinges primarily on LIII. This suggests that LII may be largely devoted to detection or encoding of odor identity while LIII may be required for attributing it with context. The fact that the cortical microcircuit spans layers, while inputs to this microcircuit are segregated by layer implies that translaminar circuits can be differentially engaged, reflecting the particular combination of active sources of input.

An odor representation requires translaminar processing of the incoming message. We know that the local contribution to the response is initiated by a small minority of mitral axon-recipient neurons that provide sufficient feedforward excitation, which, when combined with remaining sub threshold mitral inputs specifies the timing and identity of the evoked response. But it's not clear whether this local contribution is initiated entirely via the bulb or whether central inputs to layer III also shape the response.

*If such a residual contribution is present, it will tell us something about the way LII/LIII are coupled in a way that the mitral-piriform connection cannot. This can then aid with the larger question of whether this interaction contributes to more interesting operations like synthetic representation of mixtures or encoding of identity/intensity.*

*Experiment:*

Monitor translaminar activity in the presence and absence of sniffing. Is there a residual interaction between layers? Removing sniffing could result in uniform changes in both layers or preferential changes in one but not the other. The former could be explained by global attenuation in excitatory drive to the network and would imply that LII and LIII operate primarily in concert. The latter could reflect partitioning of labor based on source of input, and would imply that LII/ LIII function is more modular.

For example, spontaneous activity across LII/LIII can be abolished completely, can be attenuated, or can be biased in favor of internal inputs to Piriform. If we observe the first two possibilities, we would focus on the way in which active sampling sets the inter-laminar network state required for encoding sensory input. If we observe the third option, and this is more likely, we can disentangle

sampling effects from internal effects on spontaneous activity and determine the role these may have in shaping odor responses.

*Design:*

The following approach let's us distinguish between LII/LIII neurons, excitatory and inhibitory neurons, cortico-cortical neurons, and cortico-striatal neurons in the same FOV.

Green retrobeads are delivered to striatum, read beads, to either anterior piriform or insula; haven't decided yet. GC6 is delivered to posterior piriform which project to both of these targets. This is done in animals where excitatory neurons are labelled with tdtomato.

1-2 weeks later, volumes spanning the LII/LIII boundary in posterior piriform cortex are acquired as we:

1. toggle input from the olfactory periphery by turning off artificial sniffing
2. present 15 unique odors
3. present 4 unique odors across 4 orders of magnitude each
4. present a set of binary mixtures as well their constituent odors