Neural Signals, Systems and Technology

EEE 5283/4800

Project Proposal

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Title: Odor-Evoked Neuronal Responses in the Mouse Piriform Cortex: Analysis of Odor Selectivity and Classification Using Calcium Imaging

1. Problem Statement

Understanding how the brain processes olfactory stimuli is fundamental to uncovering the neural mechanisms underlying sensory perception, learning, and memory. Unlike other sensory modalities, the olfactory system projects directly to cortical areas without a thalamic relay, making it a valuable model for studying sensory processing in the cortex. The piriform cortex (PCx), a primary region involved in olfactory perception, plays a key role in encoding and interpreting odor identity [1, 2]. Despite its importance, the mechanisms underlying odor selectivity, the organization of odor representations across neuronal populations, and the temporal dynamics of odor-evoked activity in the PCx remain incompletely understood. In particular, how individual neurons contribute to the encoding of specific odors, and how these responses can be integrated to support odor classification, are open questions [3, 4].

This study aims to analyze in vivo two-photon calcium imaging data from the mouse piriform cortex during odor stimulation. Specifically, we seek to: (1) quantify the selectivity of individual neurons to different odor stimuli, (2) characterize how odors are represented across neuronal populations.

The dataset used for this project consist of two-photon calcium imaging recordings from the pyramidal neurons in the piriform cortex of mice. The data was collected under head-fixed conditions during passive odor presentations, where 10 different odors were delivered across multiple trials (8 trials per odor). Each trial lasted 30 seconds, with a 10-second stimulus window. Neuronal activity was recorded at single-cell resolution, allowing for the analysis of odor-evoked responses across the population. The data was preprocessed using Suite2p, and is well-suited for studying sensory encoding, population dynamics, and stimulus-specific neural representations.

2. Background and Literature Review

Olfaction is one of the oldest and most evolutionarily conserved senses in mammals, directly influencing behavior, survival, and memory [5]. The PCx plays a central role in the representation and discrimination of odor stimuli. Unlike the topographic organization of other sensory cortices, the PCx exhibits non-topographic and highly distributed representations, allowing for the encoding of a vast array of odors through population coding [4, 6-8].

Recent advances in in vivo two-photon calcium imaging have enabled the monitoring of large populations of neurons simultaneously at single-cell resolution in awake, behaving animals, facilitating a detailed understanding of how odors are represented at both the single-cell and network level [9]. Despite these advances, several critical questions remain. Odor Selectivity: Prior studies have shown that neurons in the PCx respond to multiple odors, but with varying degrees of selectivity. Quantifying this selectivity systematically remains a challenge due to trial variability and overlapping tuning profiles [1, 3, 10]. Odor Representation: A key open question is whether distinct odors elicit reliably separable population-level activity patterns in the PCx, and whether these neural patterns can support accurate classification of odor identity. Neuronal response dynamics: It remains to be determined whether the temporal dynamics of neural activity during odor presentation encode additional information about odor identity, contributing to the overall representation.

Previous studies have shown that neurons in the piriform cortex respond to multiple odors with varying levels of selectivity, suggesting a distributed coding scheme [10, 11]. Dimensionality reduction techniques such as Principal Component Analysis (PCA) and Uniform Manifold Approximation and Projection (UMAP) have been employed to visualize and analyze population-level responses to odors, revealing structure in the neural activity space [12-15].

3. Research and Method Design

3.1 Odor-Evoked ΔF/F Analysis

To assess how individual neurons in the piriform cortex respond to different odor stimuli, odor-evoked activity will be quantified using the ΔF/F method, a standard approach for measuring changes in calcium fluorescence relative to baseline. We will use the neuronal responses recorded during the presentation of 10 distinct odors, and ΔF/F traces computed for each neuron across multiple trials. For each odor, the ΔF/F responses will be averaged across trials to obtain a robust estimate of stimulus-evoked activity per neuron. This analysis will enable the evaluation of the relative strength of neuronal activation across different odors and help characterize patterns of odor selectivity at the single-cell level.

3.2 Heatmap of Odor Responses

To visualize the diversity and specificity of neuronal responses to odor stimuli, a heatmap will be constructed in which each row corresponds to an individual neuron and each column to one of the ten odor stimuli. The color intensity will represent the average ΔF/F response strength, providing a clear visual representation of how different neurons respond to each odor. This analysis will help reveal patterns of selectivity, redundancy, and overlap across the neuronal population, and will serve as an initial step in identifying odor-tuned neurons and characterizing the structure of odor representations in the piriform cortex.

3.3 Time-Aligned Raster Plot and PSTH

To examine the temporal dynamics of odor-evoked neuronal activity, neural responses will be aligned to the onset of each odor stimulus and visualized using raster plots and peri-stimulus time histograms (PSTHs). Raster plots will display spike timing or inferred activity across trials for individual neurons, while PSTHs will summarize the average response over time relative to stimulus onset. This analysis will allow for the assessment of how response patterns evolve following odor presentation, providing insight into the timing and duration of odor-evoked activity and its potential role in encoding odor identity.

3.4 Odor Selectivity Index (OSI)

To quantify how selectively individual neurons respond to different odors, an Odor Selectivity Index (OSI) will be calculated for each neuron. The OSI will be defined as:

$$\mathrm{OSI} = \frac{R_{\max} - R_{\mathrm{mean\_others}}}{R_{\max} + R_{\mathrm{mean\_others}}}$$

R_{max} is the response to the neuron's preferred (most effective) odor, and R_{mean_others} is the average response to all other odors. This metric will provide a normalized measure of selectivity ranging from 0 (non-selective) to 1 (highly selective).

Neurons will be ranked based on their OSI values to assess the distribution of odor selectivity across the population and to identify putatively narrowly tuned versus broadly tuned neurons.

3.5 Dimensionality Reduction (PCA/UMAP)

To investigate how different odors are represented at the population level in the piriform cortex, dimensionality reduction techniques will be applied to the neural activity data. Principal Component Analysis (PCA) and Uniform Manifold Approximation and Projection (UMAP) will be used to reduce the high-dimensional population activity matrix into a low-dimensional space that preserves key patterns of variability across trials. This analysis will allow for visualization of the neural response structure, enabling assessment of whether trials corresponding to different odors form distinct and separable clusters. Clear separation in this reduced space would suggest that population activity contains sufficient information to distinguish between odor identities.

3.5.1 Trial-to-Trial Similarity of Odor-Evoked Neural Responses

To evaluate the consistency of neuronal responses to the same odor, we will compute similarity using correlation-based measures (e.g., Pearson or Spearman correlation) on population activity vectors. This will help quantify and identify variability in response patterns (stability or drift).

3.5.2 Representational Stability and Odor Category Clustering

We will apply clustering techniques, such as hierarchical clustering and k-means, to group odors based on the similarity of their evoked neural activity. This may reveal functional odor "categories," where odors that elicit similar neural patterns are grouped together, potentially reflecting perceptual or molecular similarity.

4. Milestones, Metrics of Success, and Timeline

Time Period	Task	Metrics of Success
Week 1	Data Preprocessing & ΔF/F Calculation	Preprocess data, calculate average ΔF/F responses for each odor.
Week 2	Heatmap & PSTH Generation	Generate heatmap and raster plots/PSTH for each odor and trial.
Week 3	OSI Calculation & Odor Selectivity	Calculate and rank neurons based on their odor selectivity index (OSI).
Week 4-5	Dimensionality Reduction (PCA/UMAP)	Visualize reduced-dimensional space and check if odors are separable, test the similarity and clustering.
Week 6	Final Analysis & Report Writing	Finalize analysis and write the report.

5. References

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