Application of data science to multi-brain area simultaneous recordings in the mouse during movement planning

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1 Introdution

In recent years, neuroscience has seen remarkable progress, particularly with the advent of advanced technologies that allow for extensive recording of neuronal activities, either sequentially or simultaneously. Despite these advancements, directly observing functional interactions at the cellular level throughout the brain remains a formidable challenge. This is largely due to the complexity of recording neural activities across numerous regions simultaneously, which limits our ability to fully capture the dynamics of brain networks involved in complex behaviors like decision-making and motor planning (Siegle et al., 2021).

Technological innovations in multi-electrode and optical recording have made it possible to monitor neuronal activities in both cortical and, in some

cases, deeper structures. As these recording technologies continue to advance, they promise to exponentially increase the number of neurons that can be recorded simultaneously (Cunningham and Yu, 2014). This development is essential for understanding brain functions at a network level, where different regions work together to support cognition and behavior. Studies such as those by Steinmetz et al. (2019) and Stringer et al. (2019) have demonstrated the power of these technologies in capturing brainwide activity patterns, offering insights into how spontaneous and task-driven activities are encoded across multiple regions.

Previously, analytical techniques like cross-correlation and dimensionality reduction (e.g., Principal Component Analysis) have been widely employed to study neural dynamics. These techniques have proven effective for single-region data but fall short when applied to multi-region recordings due to limitations in capturing the full scope of interactions between distinct brain areas. Traditionally, single-probe recordings could only capture local neural dynamics, limiting researchers' ability to analyze how multiple regions interact in coordinated and synchronous patterns (Svoboda and Li, 2018). Multiprobe recordings, however, enable simultaneous monitoring across multiple brain areas, enhancing spatial and temporal resolution and allowing researchers to study coordinated neuronal activities with greater detail and fidelity. This capability provides a comprehensive perspective on brain dynamics, which is essential for understanding complex neural functions and addressing research questions about inter-regional neural interactions (Chen et al., 2023).

The primary objective of this thesis is to unravel the mechanisms underlying movement planning processes involved in memory-guided tasks, using advanced statistical and machine learning methods. Specifically, the research focuses on understanding the contributions of individual neurons and population-level activity, shedding light on interactions within the Anteromedial Prefrontal Cortex (ALM) and its involvement in decision-making. Furthermore, this study seeks to examine how other brain regions, such as the Thalamus, interact with the ALM during decision-making processes. By considering additional factors like anatomical location and intrinsic timescales, this research aims to advance our comprehension of how these areas contribute to decision-related neural computations.

2 Recent Work

Advances in recording technologies, such as multiprobe and high-density electrode arrays, have enabled brain-wide neural recordings with improved spatial and temporal resolution, allowing researchers to capture distributed coding of choice and action across multiple brain regions (Steinmetz et al., 2019; Stringer et al., 2019). These large-scale, multi-region recordings have highlighted the importance of studying neural activity across interconnected areas to understand complex behaviors like movement planning. Despite these advancements, analyzing high-dimensional data remains challenging, prompting the use of dimensionality reduction techniques, such as Principal Component Analysis (PCA), to distill essential patterns from large-scale recordings (Cunningham and Yu, 2014). Although widely used, PCA alone may not fully capture complex, multi-region interactions, leading researchers to explore methods like Canonical Correlation Analysis (CCA), which examines shared neural variance between regions, thereby enhancing insights into inter-regional connectivity (Pehlevan et al., 2023).

The concept of intrinsic timescales has also emerged as a key factor in understanding how different brain regions process information over time, with research showing a hierarchical organization across cortical areas. Higher-order regions exhibit longer timescales, which support sustained cognitive processes such as decision-making and memory (Murray et al., 2014). This temporal hierarchy provides a framework for studying regions like the ALM and Thalamus, which may require different timescales for coordinated activity during tasks that involve memory-guided movement planning. Further studies on intrinsic timescales highlight the role of regions with longer timescales in sustaining task engagement, which is particularly relevant to interpreting temporal integration differences in ALM-Thalamus interactions during decision-related tasks (Hasson et al., 2015; Chaudhuri et al., 2015).

To understand inter-regional interactions, researchers have also investigated neural signal transmission across brain regions. Cross-Correlogram analysis, for example, evaluates temporal correlations and functional connectivity between neural populations, potentially revealing whether activity in one area, such as the ALM, is temporally correlated with activity in another, such as the Thalamus (Amarasingham et al., 2009). This method sheds light on the directional flow of signals, which is crucial for understanding brain network dynamics. Granger causality analysis further complements this approach by identifying whether neural activity in one region causally drives

activity in another, making it a valuable tool for examining directed connectivity between the ALM and Thalamus (Gokcen et al., 2023). Together, these methods advance our understanding of the functional relationships that underlie complex behaviors.

While previous studies have demonstrated the importance of multi-region recordings and dimensionality reduction, a gap remains in understanding the specific interactions between regions like the ALM and Thalamus during memory-guided movement tasks. Many existing studies lack the simultaneous recordings necessary to capture nuanced inter-regional dynamics involved in decision-making. Moreover, techniques such as PCA and CCA have been individually applied but seldom combined with connectivity-focused methods to study ALM-Thalamus interactions in depth.

This research addresses these gaps by leveraging simultaneous recordings and an ensemble approach—integrating Cross-Correlogram analysis, jitter analysis, p-value computation, and fluorescence signal analysis—to robustly quantify ALM-Thalamus connectivity. This combination enhances the reliability of our findings, offering a more comprehensive understanding of the neural basis of movement planning in mice.

3 Data

3.1 Datasets

We are using a public dataset that contains pre-recorded neural and behavioral data from 28 mice, collected by the Janelia Research Campus and described by Guo et al. (2014). The subjects included 25 VGAT-ChR2 EYFP mice (Jackson Laboratory, JAX #014548), one C57BL/6J (JAX #000664), one Sst IRES-Cre (JAX #013044) crossed with reporter mouse Ai32 (Rosa26-LSL-ChR2-EYFP, JAX #012569), and one Emx1-Cre (JAX #005628) crossed with R26-LNL-GtACR1 Fred-Kv2.1 reporter mouse (JAX #033089).

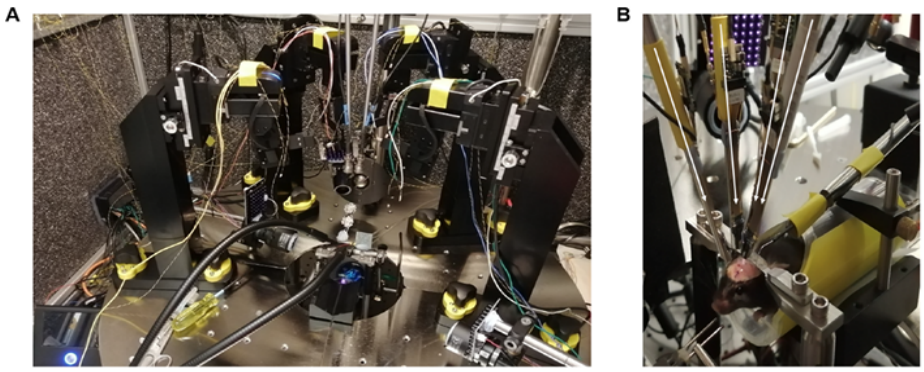

Figure 3.1: A.) The recording apparatus with four towers holding neuropixels probes. B.) Zoomed-in view showing head-restrained mouse and a recording with four neuropixels probes. Source: [2].

3.2 Experiment Setting

The data collection setup involved head-restrained, awake mice performing a memory-guided, auditory delayed response task (Inagaki et al., 2018). During the sample epoch (0.65 seconds), instruction tones of either 3 kHz or 12 kHz were played three times (150 ms each, with 100 ms intervals). This was followed by a 1.2-second delay period, concluding with a 'Go' cue (6 kHz carrier frequency, 360 Hz modulation, 100 ms duration), which prompted the mice to indicate the instruction by licking one of two ports. The response epoch lasted 1.5 seconds, during which correct responses were rewarded with a small water reward (0.1–0.2 µL), while incorrect responses triggered a time-out (1–3 seconds).

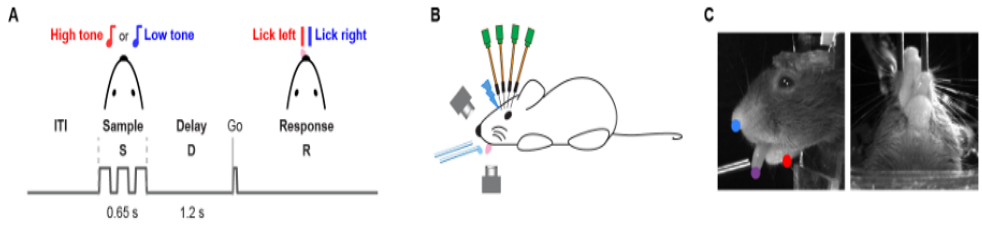

Figure 3.2: A.) Memory-guided directional licking task. Mice were trained to lick to one of two possible lickports instructed by one of two auditory stimuli (high or low tone), presented during the sample epoch (S). Mice respond after a 'Go' cue in the response epoch (R), after a delay epoch (D). Between trials is the inter-trial interval (ITI). B.) Typical recording with four Neuropixels probes during behavior. Electrophysiological measurements were combined with movement tracking using high-speed videography and laser-based photoinhibition of ALM (lightning bolt). C.) Video images, showing three tracked keypoints (nose, tongue, jaw) in the side view. Source: [2].

3.3 Data Acquistion

The datasets were stored in DANDI repository where to retrieve data we need to use either the Python Command Line Interface (CLI) Client or the DANDI Web application. Specific subject data can be identified and downloaded by querying the DANDI web platform or using CLI commands to streamline retrieval. After determining the relevant subject ID, individual file or session links can be used to access specific datasets through the DANDI CLI.

Each session's data is stored in the Neurodata Without Borders (NWB) format, an open standard designed for organizing large-scale neurophysiological data. Each .nwb file typically contains comprehensive time-series and video data, amounting to hundreds of gigabytes per session, which includes electrophysiological recordings, behavioral events, and high-speed video captures. This format facilitates the integration of data from different modalities and time scales, allowing for standardized storage and efficient data access.

3.4 Data Curation, Cleaning and Preprocessing

Following data retrieval, the next step involves curating and organizing the data for each mouse. The curation process consolidates individual session data into a main Data Table, preserving the details of each session while

providing a structured overview at the subject level. This structured organization aids in subsequent analyses, enabling consistent access to session-specific details while maintaining a comprehensive dataset framework.

[AND MORE STUFF ABOUT CLEANING AND PROCESSING FROM VERSION3 DOCUMENT]

4 Methodology

4.1 Firing Rate

In this section, we focus on the analysis of spike statistics, specifically employing firing rate calculations and Gaussian kernel smoothing to represent neural activity in a meaningful way. Neuronal spikes, or action potentials, serve as the fundamental unit of information in the brain, which encodes information through the timing and frequency of these events (Abbott, 2001). Spikes can be grouped into spike trains or collective patterns of activity that reveal the underlying structure and function of neural circuits. Understanding these patterns through spike statistics is critical for decoding the neural representations and processing mechanisms within brain regions such as the Anteromedial Prefrontal Cortex (ALM) and Thalamus.

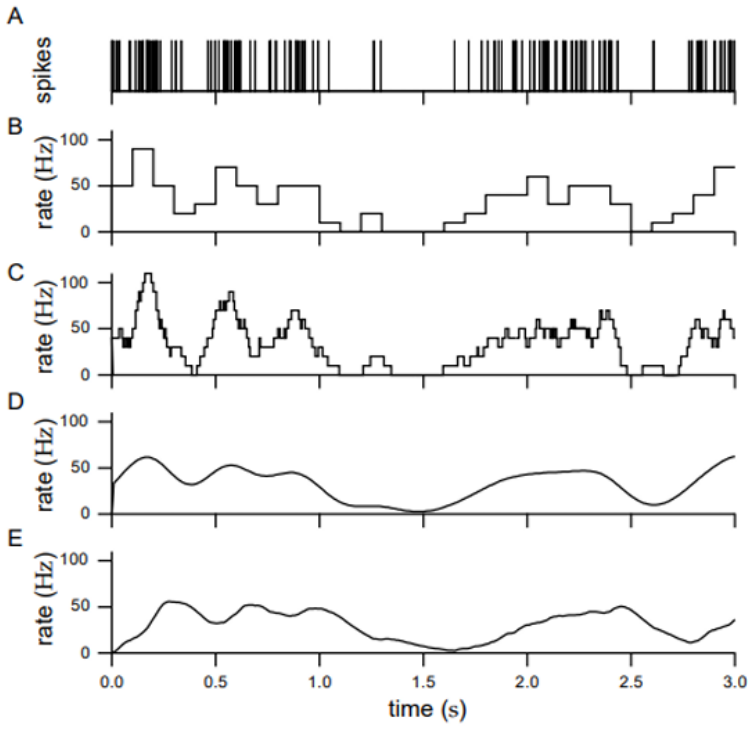

Figure 4.1: A.) spike train from a neuron in the inferotemporal cortex of a monkey recorded while that animal watched a video on a monitor under free viewing conditions. B.) Discrete time firing rate obtained by binning time and counting spikes with 1t = 100 ms. C.) Approximate firing rate determined by sliding a rectangular window function along the spike train with 1t = 100 ms. D.) Approximate firing rate computed using a Gaussian window function with σt = 100 ms. E.) Approximate firing rate using the causal window function with $1/\alpha = 100$ ms. Source: [1].

One of the key metrics used in spike train analysis is the firing rate. The firing rate measures the frequency of action potentials fired by a neuron over a defined period, providing a quantitative basis for interpreting neural activity patterns. Formally, the firing rate (FR) is calculated as:

$$FR = \frac{S}{T}$$

where:

- FR is the firing rate (spikes per second),
- S is the total number of spikes observed,
- T is the time interval over which the spikes are counted.

By determining the firing rate, we can observe trends in neuronal activity across different behavioral epochs, such as the sample, delay, and response phases in memory-guided tasks. In this study, histograms and window functions are applied to visualize the firing rates, which allows for an intuitive interpretation of how neuronal populations respond to task-relevant stimuli and periods.

4.1.1 Gaussian kernel Smoothing

To obtain a continuous and smooth representation of the firing rate, we apply Gaussian kernel smoothing. This technique involves convolving the discrete spike train with a Gaussian kernel, providing a smoothed approximation of the underlying firing pattern. Gaussian kernel smoothing is especially valuable in studies with high-frequency spike data, as it reduces noise and enhances the interpretability of neural activity over time.

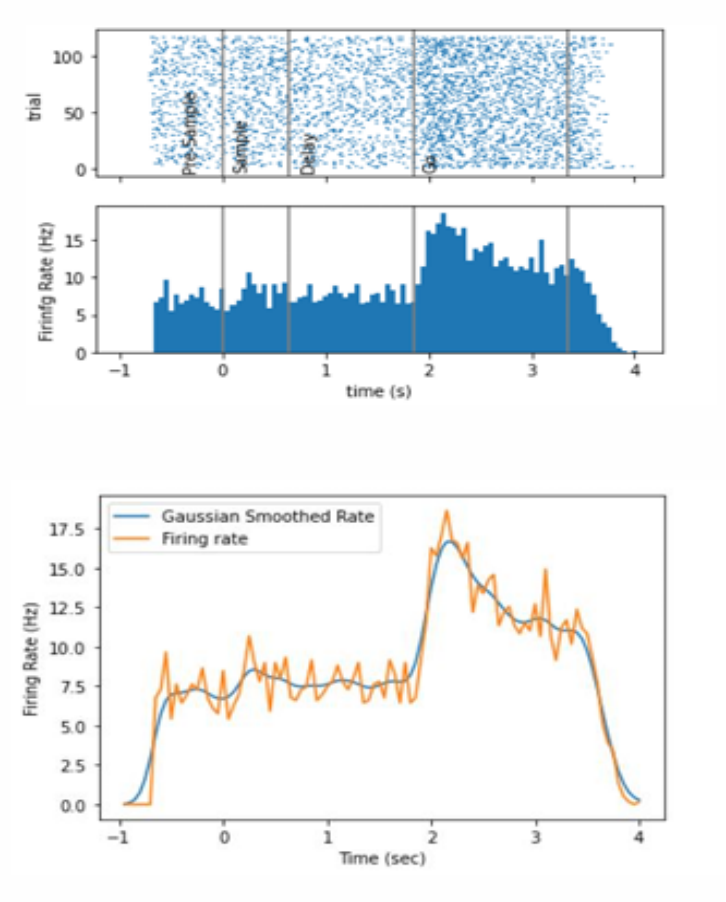

Figure 4.2: (Top) Raster plot showing spike times for a single neuron across 100+ trials over the full task duration, with key task phases marked & histogram of firing rates aggregated over time. (bottom) The smoothed firing rate (blue) using a Gaussian kernel ($\sigma = 0.1$) and the raw firing rate (orange).

In our analysis, we applied a Gaussian kernel with a sigma (σ) of 0.1[CITE FIGURE], a parameter chosen to balance temporal resolution with smoothness. The kernel width effectively determines how much the firing rate at each point is influenced by surrounding time points, with a smaller sigma providing higher resolution and a larger sigma yielding a broader, smoother curve. By adjusting sigma, we optimize the representation of activity trends without losing critical temporal details.

4.2 Population Analysis

In this analysis, population analysis is used to investigate the collective behavior of neurons during decision-making and motor planning tasks. By examining activity patterns across populations of neurons, we aim to capture higher-level neural dynamics that contribute to task-specific outcomes. This analysis employs two primary techniques: Coding Direction and Principal Component Analysis (PCA), which are effective for extracting meaningful patterns from complex, high-dimensional neural data.

4.2.1 Coding Direction

The Coding Direction (CD) method is used to characterize and distinguish neural responses associated with specific behavioral outcomes, such as leftward or rightward licks, in the context of a memory-guided task. In this analysis, a set of orthogonal directions in the multi-dimensional activity space is defined, each represented by an n×1 vector, where n is the number of neurons. These vectors are designed to maximize the separation between response vectors for lick-left and lick-right trials at task-relevant time points within this n-dimensional activity space (Chen, 2023).

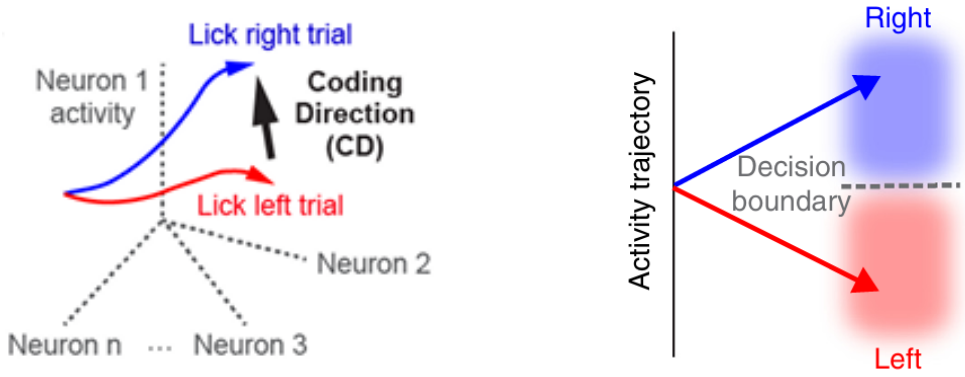

Figure 4.3: Left, the definition of coding direction (CD), a vector in activity space that best separates activity trajectories for different task conditions. Source: [2]. Right, the activity trajectory has a clear separation boundary to select its movement direction. Source: [4]

To compute the Coding Direction, we performed the following steps:

- **Mean Spike Count Calculation**: For each trial type (lick-left and lick-right), we calculated the mean spike count vector separately for each neuron. This provides two distinct mean response vectors: $\vec{x}_{\text{lick-left}}$ and $\vec{x}_{\text{lick-right}}$.
- **Direction Calculation**: At each time point, we computed the difference between these mean vectors, yielding the vector $\vec{w}_t = \vec{x}_{\text{lick-left}} - \vec{x}_{\text{lick-right}}$. This vector $\vec{w}_t$ represents the Coding Direction that maximally separates the two trial types.
- **Averaging Across Epochs**: To obtain a robust representation of the Coding Direction over the task period, we averaged $\vec{w}_t$ across specified epochs (e.g., delay and go periods).
- **Projection onto the Coding Direction**: Finally, we projected the activity of each trial onto the Coding Direction vector. This projection facilitated a quantitative assessment of how well the neural population activity aligned with the task demands, specifically the discrimination between lick-left and lick-right trials (Chen, 2023).

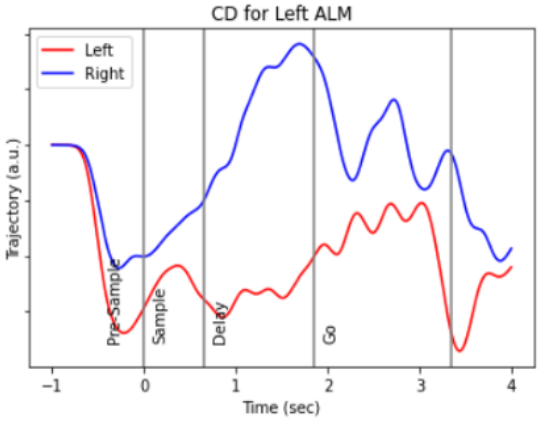

Figure 4.4: Left and right lick's firing rate projection onto Coding Direction CD_{Delay} (Averaged over delay epoch)

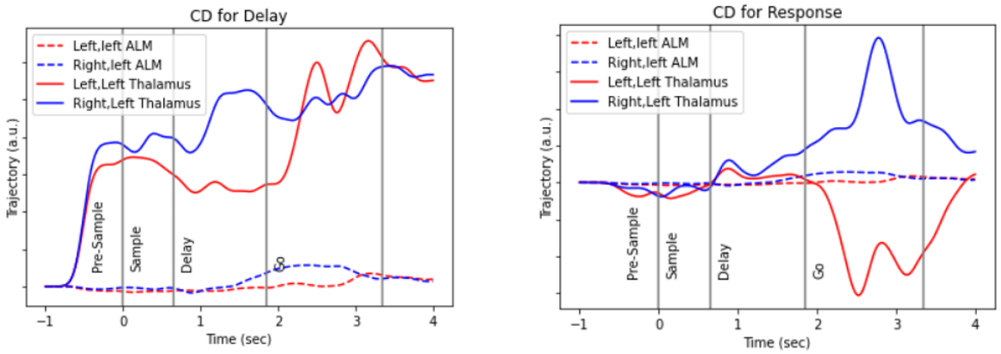

Figure 4.5: Red and blue indicate trial instruction(correct trials) left and right lick respectively. (Left) Derived trajectory by projecting onto Coding direction for late delay(last 0.6sec of Delay epoch). (Right) Derived trajectory by projecting onto Coding direction for early go(first 0.6sec of Go epoch).

Through this process, we observed that the population activity in regions like the anterior lateral motor cortex (ALM) and the Thalamus could be visualized in an activity space where trajectories reflect decision-relevant dynamics. For instance, in the ALM, the Coding Direction showed distinct clustering of trajectories corresponding to lick-left and lick-right trials, especially during the late delay epoch (last 0.6 seconds), allowing us to identify clear boundaries between the two trial types. By applying this approach to both the ALM and Thalamus, we found that Coding Direction analysis revealed distinct neural representations for each response type across critical task phases, particularly in the late delay and early go periods, making it ideal for examining selectivity.

4.3 Principal Component Analysis

Principal Component Analysis (PCA) is a widely used dimensionality reduction technique that identifies orthogonal directions (principal components) in the data that capture the greatest variance. In this analysis, we applied PCA to simplify the high-dimensional neural activity data, representing complex firing patterns through a reduced set of key components. By capturing the primary variance in the neural firing rates, PCA allows us to represent underlying trends in population activity, aiding in extracting patterns relevant to memory-guided tasks.

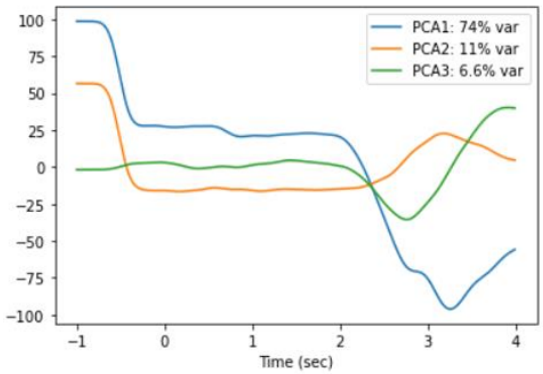

Figure 4.6: PCA components of firing rate vector of 100's of neurons(Correct trials).

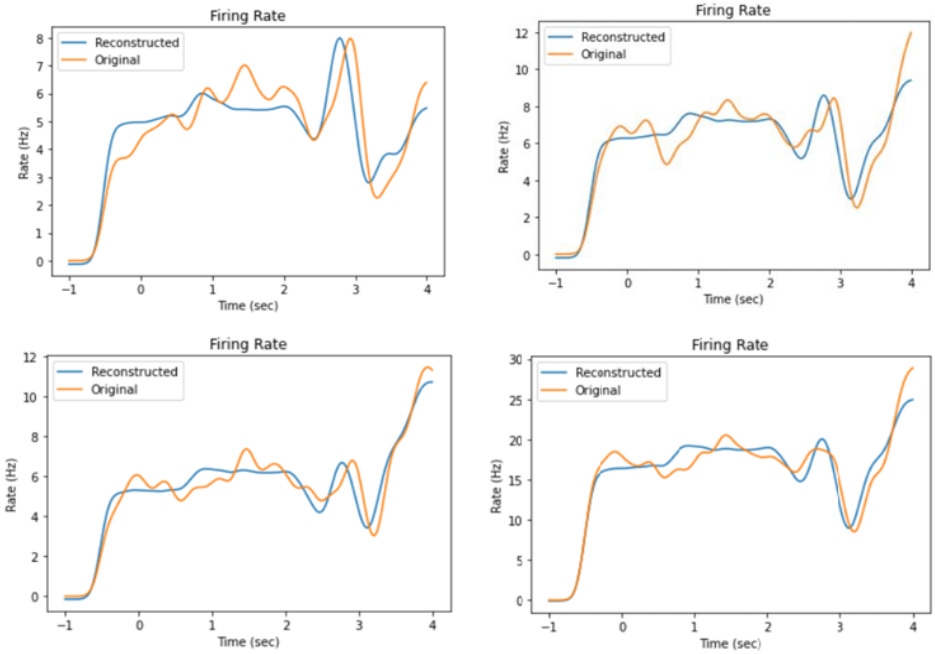

Figure 4.7: Reconstructed Firing rate from derived top 3 PCA components.

The following steps were conducted to apply PCA to the neural activity data:

- **Variance Maximization**: PCA identifies the directions that maximize the variance in the data. Each principal component (PC) represents an axis in the lower-dimensional space that captures significant variance, allowing us to simplify complex population activity into a few key components (Cunningham & Yu, 2014).

- **Low-Dimensional Projection**: Neural activity data for each trial was projected onto the first few principal components, reducing dimensionality while retaining the most informative variance. This projection highlights patterns in firing rates across trials, revealing population-level structures within the data.

- **Interpretation of Principal Components**: Each principal component was analyzed to understand its contribution to the overall variance. In this study, the initial appearance of neuron firing rates was seemingly random and high-dimensional. However, PCA revealed that the underlying structure could be distilled into a few principal components, with these components effectively capturing relevant trends across different behavioral conditions.

[ADD ENCODING, DECODING INFORMATION PART]

While PCA is highly effective in reducing dimensionality, it is important to note that the variance captured by PCA includes all types of variability, including both task-relevant and spontaneous fluctuations in firing rates (Cunningham & Yu, 2014). This means that the low-dimensional space identified by PCA may include variability unrelated to task events, which could introduce noise into the interpretation of neural dynamics. Despite this limitation, PCA remains a valuable tool for summarizing neural population activity and identifying general patterns across trials.

4.4 Multimodal Analysis

4.4.1 Cross-correlograms

Cross-correlogram analysis reveals significant time-lagged correlations between the Anterior Lateral Motor Cortex (ALM) and Thalamus, indicating potential directional signal transmission during the memory-guided task.

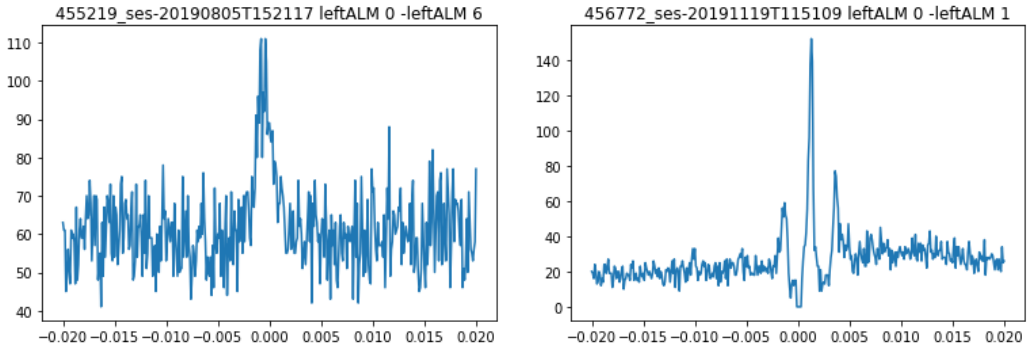

Figure 4.8: ALM-ALM Cross-correlograms with time(sec) centered on go epoch start time

4.4.2 Jitter Corrected CCG

Jitter-corrected Cross-Correlogram analysis demonstrates significant functional connectivity and directional signal flow between ALM and Thalamus during the memory-guided task.

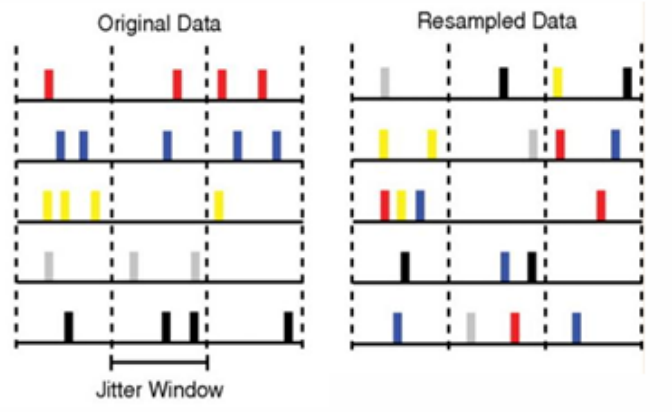

Figure 4.9: Diagram of the method used for CCG correction. Data from trials is shown on the left, with a different color labeling for the spikes from each trial. The trials were divided into bins based on the jitter window size (dashed vertical lines). From the original data, a raw CCG was computed. The original data were then resampled. For each spike in each jitter bin, a new spike was chosen randomly, without replacement, from the set of all spikes in the same jitter bin on all trials. Source: [3].

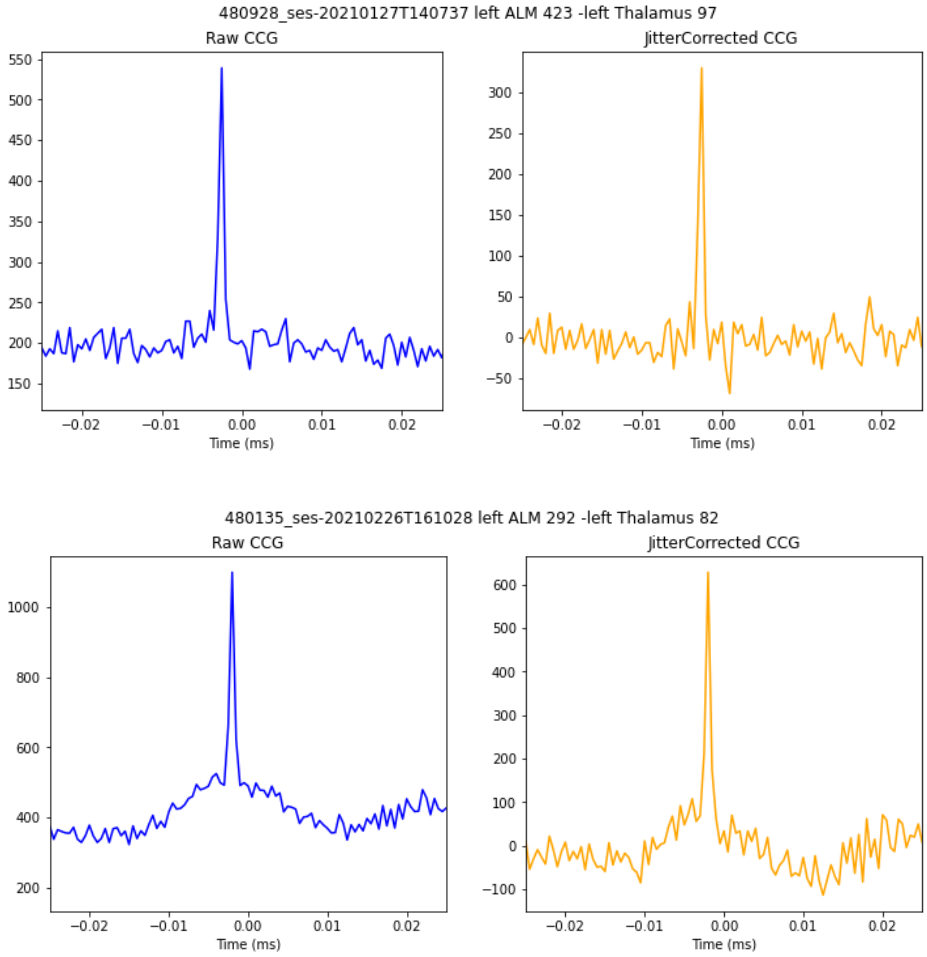

Figure 4.10: Title of graphs contains subject and session of recorded neurons. Blue, represents Raw CCG calculated from the original data. Orange, represents Jitter corrected CCG(jitter window size = 20ms).

4.4.3 Autocorrelogram and Intrinsic Timescale(τ)

Autocorrelogram analysis, through intrinsic timescale (τ) measurements, reveals the memory retention capacity of ALM and Thalamus, indicating how long these regions retain neural patterns during the memory-guided task.

4.5 Fluorescence Analysis

Fluorescence analysis identifies and maps projection zones in the brain regions receiving inputs from the ALM, highlighting the areas involved in sustaining neural activity essential for memory retention and movement planning.

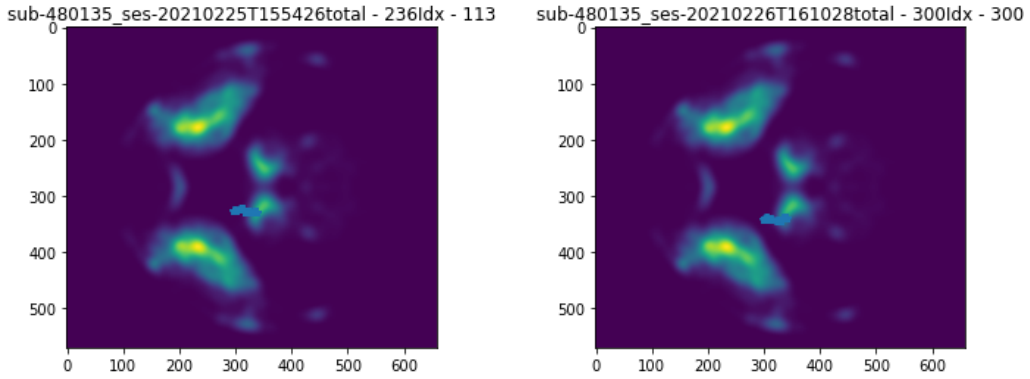

Figure 4.11: total and idx represent total no. of neurons and no. of overlapped neurons (on condition: radius = 100μm, strength of fluorescence ¿ 0.25) with ALM project zone. The fluorescence map represents the ALM projection zones while dots are recorded neurons plotted based on their CCF locations.

4.6 P-values

By evaluating the significance of coincident spikes and CCG peaks, we can identify meaningful neural interactions, distinguishing genuine connectivity from random chance.

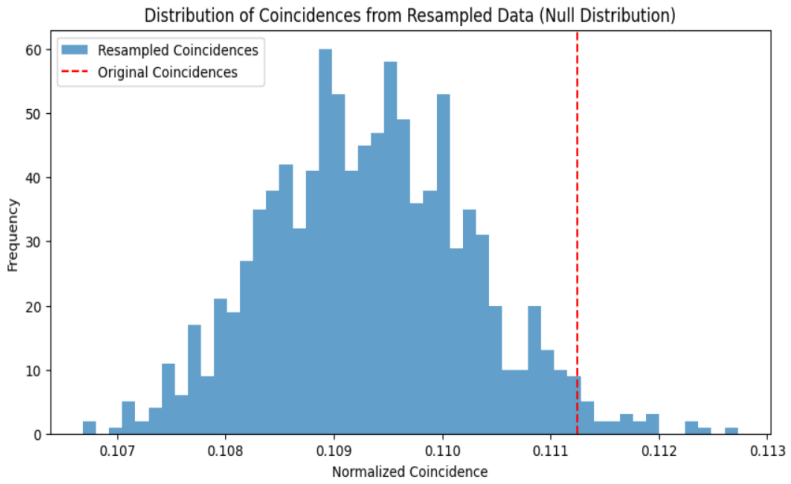

Figure 4.12: The distribution of coincidental spike occurrences (normalized coincidence) generated from resampled data(number of sample = 1000), representing the null distribution of coincidences. The blue bars indicate the frequency of coincidental spike values expected under the null hypothesis, while the red dashed line represents the observed coincidental spike value from the original data.

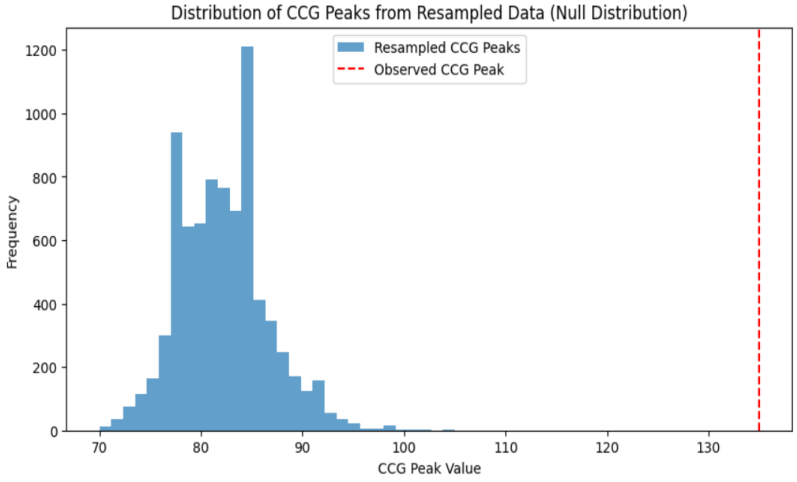

Figure 4.13: The distribution of CCG peaks generated from 10,000 resampled (jittered) data with a jitter window of 20 ms. The blue bars depict the frequency of CCG peak values under the null hypothesis. The red dashed line marks the observed CCG peak from the original data.

5 Discussion

5.1 Results

5.2 Future Work

6 Conclusion

References

[1] Larry F Abbott. "Theoretical neuroscience rising". In: *Nature Neuroscience* 4.11s (2001), pp. 1186–1190. ISSN: 1097-6256. DOI: 10.1038/nn1101-1186. URL: https://doi.org/10.1038/nn1101-1186.

[2] Susu Chen et al. "Brain-wide neural activity underlying memory-guided movement". In: (Mar. 2023). DOI: 10.1101/2023.03.01.530520. URL: http://dx.doi.org/10.1101/2023.03.01.530520.

[3] Matthew Smith and Adam Kohn. "Spatial and Temporal Scales of Neuronal Correlation in Primary Visual Cortex". In: *The Journal of Neuroscience 28* (2008). URL: https://doi.org/10.1523/JNEUROSCI.2929-08.2008.

[4] Karel Svoboda and Nuo Li. "Neural mechanisms of movement planning: motor cortex and beyond". In: *Current Opinion in Neurobiology* 49 (Apr. 2018), pp. 33–41. ISSN: 0959-4388. DOI: 10.1016/j.conb.2017.10.023. URL: http://dx.doi.org/10.1016/j.conb.2017.10.023.