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Source Location as a Feature for the Classification of Multi-sensor Extracellular Action Potentials

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Abstract— **Extracellular action potentials (EAPs) must be classified before they can yield any useful information on neuronal function and organization. Neuronal source classification therefore represents a critical step in the analysis of electrophysiological data. This study demonstrates the efficacy of a multi-sensor EAP classification scheme using source location as a classification feature. Localization was performed using the multiple signal classification (MUSIC) algorithm. Six distinct source neurons were classified from 20 seconds of extracellular, four-sensor (tetrode) recordings. On average, 89.5% of the waveforms making up each class matched the shape of the average class waveform. These results indicate that this classification scheme can successfully identify individual neurons from multi-sensor EAP recordings.**

I. Introduction

Extracellular recording is one of the most commonly used techniques for studying neural activity *in vivo* [1]. Its main advantage is allowing action potentials (APs) to be recorded and resolved for multiple neurons simultaneously [2]. The advent of multi-sensor extracellular recording has made this process easier and more effective [3]. However, in order to determine functional relationships and neural interactions, the recorded APs must be classified. Many techniques have been proposed for this purpose, ranging from principal component analysis (PCA) [4], to expectation-maximization based clustering [5].

Although these methods differ in their actual classification algorithms, the defining feature that sets most classification schemes apart is their feature selection algorithm. Choosing the correct feature largely determines the efficacy of extracellular action potential (EAP) classification. While commonly used abstract mathematical features, such as the principal components in PCA [6], or various template scores [7], can be used for classification purposes, their calculated features may not be unique to single neurons, and may change significantly with sensor location and across trials. Features that can reliably represent a single neuron, as well as remain invariant across trials and sensor positions are preferable for classification purposes.

One such classification feature, previously explored by Chelaru and Jog in the context of tetrode recordings [8], is neuron location. Source location is a superior classification feature to mathematical abstractions for several reasons. First of all, source location remains constant even if the recording sensors are moved with respect to the neurons of interest. This can account for unexpected movements, which are a common occurrence during extracellular recording experiments. Secondly, source location can be used to identify and follow single neurons across trials. In chronic recordings, where neuron populations may migrate with time, this can provide both information on neural migration trends and allow for the migrating neurons to be classified accurately, and grouped consistently across many consecutive trials. Note also that localization does not require spike alignment, as location features are extracted independently for each recorded EAP.

The localization approach taken by Chelaru and Jog applies a simplified monopole-like model to the recordings and proceeds to estimate the source location by solving a nonlinear system of equations. In contrast to this numerical solution, our previous work showed that the monopole-model could be inverted exactly, resulting in a closed-form solution [9]. Unfortunately, this method is very sensitive to noise. Likewise, solutions come in pairs, one of which is spurious. Identifying which solution is the accurate one can be tedious and sometimes impossible.

Due to these limitations we explored a different localization method rooted in statistical signal processing [10]. The multiple signal classification (MUSIC) algorithm used in this study has proven effective in both electroencephalogram (EEG) and magnetoencephalogram (MEG) source localization [11], as well as in our preliminary source localization experiments with tetrodes [10]. The MUSIC algorithm is more robust against noise than the closed-form solution, and generates a single localization result, eliminating the need to identify the accurate and spurious solutions from a pair.

The work presented here builds on our previous localization work, and uses MUSIC-derived source locations as EAP classification features. Classification efficacy was quantitatively assessed by within- and inter-class analyses.

II. Methods

A. Data Collection

Data used in this experiment is publicly available online [12]. A planar silicon probe, developed by the Center for Neural Communication and Technology of the University of Michigan (now manufactured by NeuroNexus), was used for recording. The probe was placed below the surface (∼50-100 μm) of an adult locust's antennal lobe. Recordings were sampled at 15 KHz and bandpass filtered from 300 - 5,000 Hz. A total of 20 seconds of data was provided

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from four of the probe tip sensors. For a more detailed data collection procedure please refer to [13]. Data analysis, including detection, feature extraction, and classification, was performed in MATALB.

B. Detection

Spike detection was performed using a supervised matched filter for multi-sensor data. Half of the collected data was used for training, and the remaining half was used for further analysis. 25 EAPs, 2.7 ms each, were selected from the training data, and used to generate a matched template, $\mathbf{s}$. Similarly, 20 noise samples, roughly 50 ms each, were used to generate a noise covariance matrix, $\mathbf{C}$. To simplify the noise covariance calculation, noise processes were assumed to be stationary, and uncorrelated in space. The noise processes' temporal statistics were also assumed to be identical across the four sensors. Detection was performed by thresholding the matched filter output at three standard deviations above the noise mean

$$\mathbf{s}^T\mathbf{C}^{-1}\mathbf{x} > \gamma \tag{1}$$

where $\mathbf{x}$ is the signal, and γ is the threshold. A more detailed derivation of the test statistic can be found in [14].

C. Feature Extraction

The classification scheme presented here uses neuron location as a classification feature. The source of each detected EAP was localized using the MUSIC algorithm. Briefly, MUSIC assumes that an EAP, $\psi(t)$, recorded with a c-sensor array, can be described by the static linear system

$$\psi(t) = m(r)s(t) + w(t) \tag{2}$$

where t is the time instant, $m(r) \in \mathbb{R}^{c\times 1}$ is the lead field vector (LFV) [11], [15] representing the system's response to a unitary signal input, $s(t) \in \mathbb{R}$ is the neuron's firing strength, and $w(t) \in \mathbb{R}^{c\times 1}$ is zero-mean noise. The LFV can represent a response to an arbitrary number of various source types, from a single monopole-like source to complicated multipole, multi-source systems. Similarly, no constraint is placed on the number of sensors being used. Due to its simplicity, both mathematically and computationally, our preliminary localization work concentrated on a monopole LFV model. Our results indicated that this model is sufficient for the localization of signals recorded from a four-sensor microelectrode, or tetrode [10]. Therefore a monopole LFV model was also used in this study. A more complex model, such as the commonly used dipole model, could also be appropriate here but may require data from more than four sensors [16].

The MUSIC algorithm localizes the EAP source by finding the source location, $r^\star$, with an LFV most orthogonal to the noise subspace

$$r^\star = \arg\min_r \frac{m(r)^T E_N {E_N}^T m(r)}{m(r)^T m(r)} \tag{3}$$

where $E_N \in \mathbb{R}^{c\times(c-1)}$ is the noise subspace, and is calculated via singular value decomposition of the recorded EAP, $\psi(t)$.

Note that EAPs are not filtered or aligned prior to localization. A more detailed derivation of this method can be found in our previous work [10].

D. Classification

MUSIC-derived locations from each detected EAP were classified using an expectation-maximization (EM) algorithm. The EM algorithm assumes Gaussian distributed clusters for EAPs coming from a specific neuron, and a uniformly distributed cluster of outliers. The EM algorithm was used to group points into several different cluster models, and the optimal cluster model, or number of clusters, was determined by maximizing the Bayes Information Criterion across all models [17]. For a detailed derivation of this classifier please refer to our prior work [5].

E. Analysis

After classification each cluster was analyzed for average location and spread. The spread was quantified by a standard radius, which is the norm of each cluster's x, y, and z standard deviations. The EAPs from each cluster were aligned to their peak values and averaged to demonstrate the representative waveforms for each cluster.

Waveform signatures, representing the relative signal power across the four sensors, were then calculated for each EAP. This signature was used to determine the consistency of multi-sensor EAP shapes within a given cluster, and was selected based on its robustness against both large noise variations and possible variations in a single neuron's firing strength.

III. Results

All 1040 EAPs detected in the 20 second data stream were successfully localized using MUSIC. The EAP locations, projected onto the x-y plane, are shown in Fig. 1 (Top). This data was classified yielding 6 distinct location clusters, centered at (0.4 36.4 0.5), (21.3 19.3 -0.1), (9.5 -25.7 -0.1), (32.1 -8.0 -0.7), (-24.8 -14.4 0.2), (-25.4 13.5 -0.2) μm with standard radii of 11.5, 10.1, 6.7, 12.3, 9.0, and 11.2 μm respectively [Fig. 1 (Bottom), Table I]. Only 34 source locations were classified as outliers, representing 3% of the data set.

The underlying EAP waveforms representing each source location in a given cluster were analyzed to assess classification efficacy. The average waveforms are unique to each location and distinguishable from each other (Fig. 2). This is strong evidence that each cluster represents a unique neuron. Furthermore, localization results for each cluster make physical sense. Cluster N3 is localized closest to S1, and displays the strongest signal at S1 (Fig. 2). Likewise, N3 displays a stronger signal at S1 than any other cluster. Similar observations are true for the remaining five clusters. These results imply that neurons are localized accurately not only with respect to the sensors, but also with respect to each other.

Although the average waveforms are unique, all six have relatively high standard deviations at the EAP peaks (up to

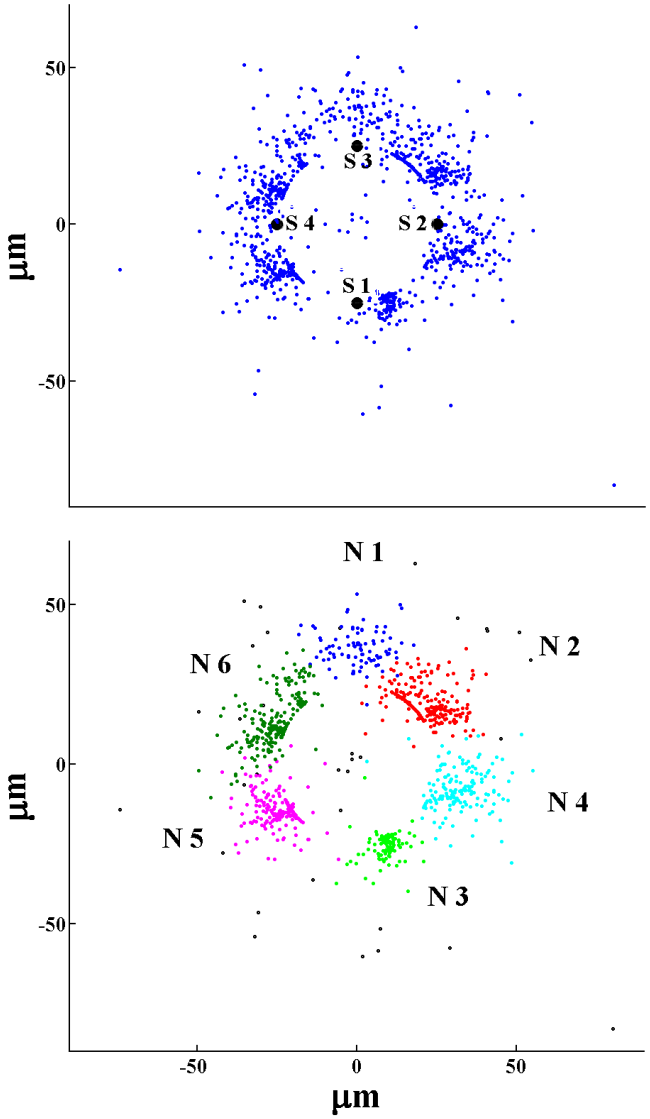

Fig. 1: **Top:** MUSIC source localization results for all 1040 detected EAPs (blue). Sensor locations are depicted in black and marked S1-4. **Bottom:** Classification results for EAP source locations shown above. Black points represent outliers, and all other colors represent distinct source location clusters, labeled N1-6.

TABLE I: Location, standard radius, and classification accuracy of each class N1-6

Source		N1	N2	N3	N4	N5	N6
Location	x (μm)	0.4	21.3	9.5	32.1	-24.8	-25.4
	y (μm)	36.4	19.3	-25.7	-8.0	-14.4	13.5
	z (μm)	0.5	-0.1	-0.1	-0.7	0.2	-0.2
Std. Radius (μm)		11.5	10.1	6.7	12.3	9.0	11.2
Accuracy (%)		98.1	77.5	95.4	98.1	90.1	77.2

4.9 standard deviations of the noise). This is not surprising as the waveforms were crudely aligned by peak EAP values, greatly increasing the peak variance. Additionally, biological noise is known to increase during spikes due to the correlated activity of nearby neurons [18]. To determine more conclusively if this high waveform variance implies miss-classification, waveform signatures, defined here as the signal power across the four sensors for each EAP, were compared within clusters.

For clusters N1, N3, N4, and N5, the waveform signatures are consistent with the average waveform signature among 98.9%, 95.4%, 98.1%, and 90.1% of the EAPs in each cluster, respectively (Table I). This implies that the variance in EAP waveforms across these clusters is most likely due to miss-alignment or biological noise. It is reasonable to conclude that these clusters represent unique and singular neurons.

The remaining two clusters were slightly less consistent, with 77.5% and 77.2% of the EAPs in N2 and N6, respectively, matching their average waveform signatures. For N6 the remaining 22.8% of waveforms match the waveform signature of N1, and therefore seem to be miss-classified. N2 on the other hand had a broad spectrum of EAPs not matching the average waveform. This may imply that some EAPs classified as N2 account for a different nearby neuron, or a superposition from two or more neurons. Although these clusters were less internally consistent, the results still indicate that they represent distinct neurons.

Overall, classification based on our feature extraction method successfully differentiated 6 unique and distinct neurons from tetrode recorded EAPs. Furthermore, MUSIC-derived neuron locations are physically relevant, implying localization accuracy.

IV. Discussion

The miss-classification observed in clusters N2 and N6 may be due in part to the geometry of the planar sensor array used for recording. In general, the MUSIC algorithm needs data from a three dimensional recording sensor array to produce accurate localization results. However, the algorithm will produce solutions, albeit less reliable ones, given data from a planar array. Our preliminary work has shown that solutions derived from a planar array have poor resolution on the z-axis, assuming the sensor array is in the x-y plane, and form short arcs of symmetry running though the true source location [19].

This phenomenon can be seen in the localization results presented here. The solutions form a rough circle around the recording sensors, with each cluster assuming the general outline of an arc. Likewise, the clusters are all located just above or just below the recording array (Table I), giving very little resolution on the z-axis. If a three-dimensional array is used for recording, the clusters are expected to form tighter, more Gaussian-like spheres, and be more reliably localized on the z-axis, thus presenting a more constructive classification environment. This would likely decrease the miss-classification rates, and the localization variance observed in

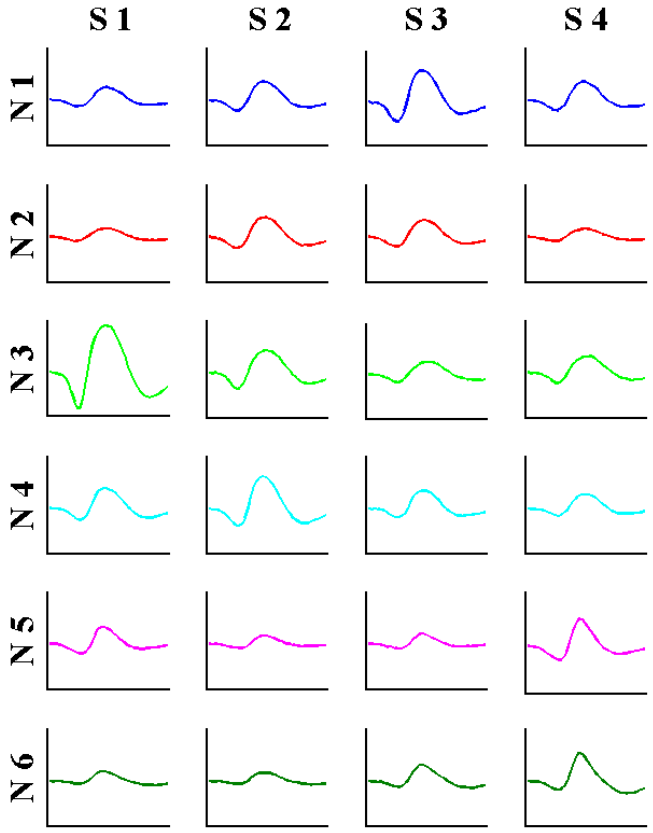

Fig. 2: Average waveforms for each cluster shown in Fig. 1 (Bottom). Waveforms are color matched to the clusters. Note that each cluster has a unique and distinguishable waveform signature.

the present study.

The use of arrays with more than 4 sensors may also decrease localization error and miss-classification rates. As 4 sensor localization is sensitive to outlying data, a larger number of sensors may mitigate the effects of noise and decrease localization variance.

Another notable characteristic of this feature extraction scheme is its limited sensitivity to noisy or outlying signals. Unlike the approach presented by Chelaru and Jog [8], where 39% of recorded spikes were filtered out and discarded as outliers prior to analysis, the feature extraction scheme presented here did not filter out any spikes prior to analysis. Furthermore, only 3% of the classified spikes were identified as outliers. Conserving most of the detected EAPs will increase the reliability of further analysis, and improve the amount of information that can be gathered from a given recording session.

V. Conclusion

Six distinct EAP sources were successfully classified, with an average accuracy of 89.5%, using source location as a classification feature. This was achieved in spite of an unfavorable recording sensor arrangement. Furthermore, the classification scheme used here does not rely on any prior knowledge of EAP class characteristics, and does not require the signals to be aligned prior to classification. This decreases the amount of signal processing necessary for classification, allows a broader range of units to be identified, and permits the algorithm to be completely unsupervised. Given our results, this technique presents itself as a strong candidate for broad use in extracellular signal analysis. Future work will quantify the algorithm's performance on more favorable data sets, collected using a three-dimensional sensor array, and on data sets from arrays having more than 4 sensors. The algorithm's performance under these conditions is expected to significantly improve.

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