



Quality Assessment for Calcium Imaging Data (QACID)

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Introduction

Two-photon calcium imaging (2PCI) has become a vital tool in neuroscience, valued for its ability to capture detailed activity deep within brain tissue in real time. However, before analyzing this data, it is crucial to ensure its quality. Assessing the quality of raw 2PCI datasets helps avoid the use of flawed data, saving both time and computational resources. In this context, we introduce QACID, a pre-analysis pipeline designed to evaluate data quality by examining temporal and spatial features. QACID enhances the trustworthiness of research outcomes and increases efficiency by preventing the processing of inferior-quality datasets.

Objectives

To develop a robust assessment pipeline for evaluating the quality of raw data from two-photon calcium imaging. We aim to implement quantitative measures that accurately determine dataset quality, ensuring that subsequent data analyses are based on useful, reliable, and accurate information.

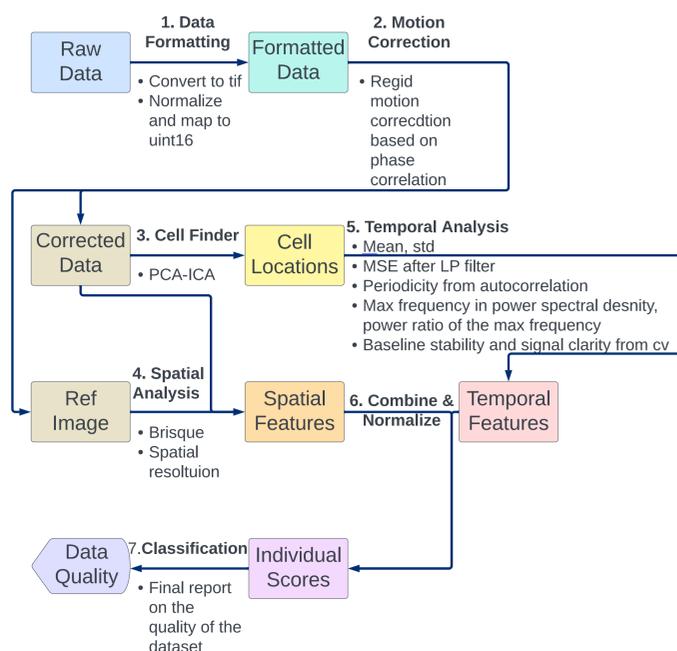
Datasets and Algorithms

152 real training sets from publications (45 CalmAn, 2 Liang, 50 PatchWarp, 55 Suite2p)

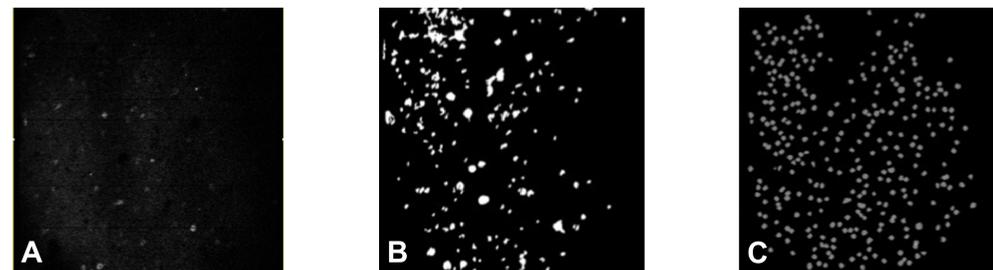
81 NAOMi-simulated training sets (varying volume, activity rate, alignment, excitation power, dark count, calcium indicator, electronics base noise variance, noise type)

70 testing sets (12 Dandi 000565, 13 Liang, 66 NeuroFinder, 1 from each of Dandi 000219, 000691, 000951, and 000728)

QACID pipeline (blocks show intermediate output, arrows show procedure)

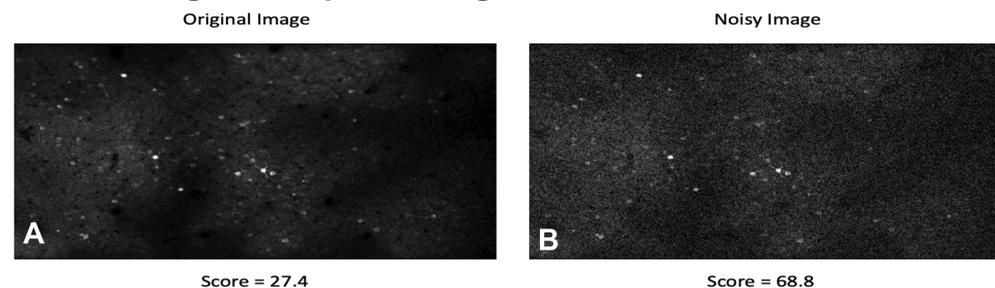


1. PCA-ICA is effective in neuron localization



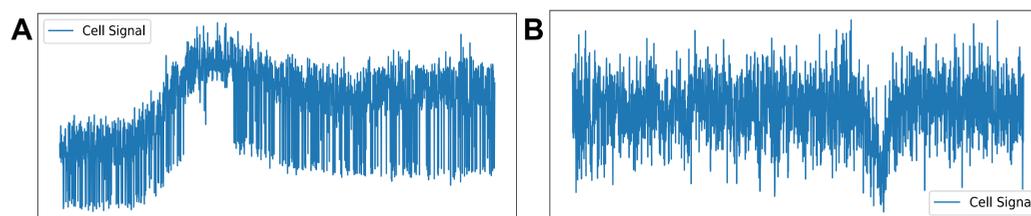
(A) Raw calcium imaging data. (B) Neuron locations identified by PCA-ICA. (C) Ground truth of neuron locations. Incremental PCA was used to acquire principal components. Then we applied ICA to find a matrix W that linearly transforms the mixed signals Y into statistically independent components.

2. BRISQUE algorithm quantifies global and local noise and distortion



(A) Single frame from a calcium imaging dataset. (B) Frame with Gaussian noise added. BRISQUE, a no-reference image quality assessment algorithm, was used on both the whole image and multiple sliding windows to compute a score for global and local noise and distortion. Adding Gaussian noise to the image resulted in a significantly higher BRISQUE score, indicating a lower-quality image.

3. Temporal metrics characterize the cell signals



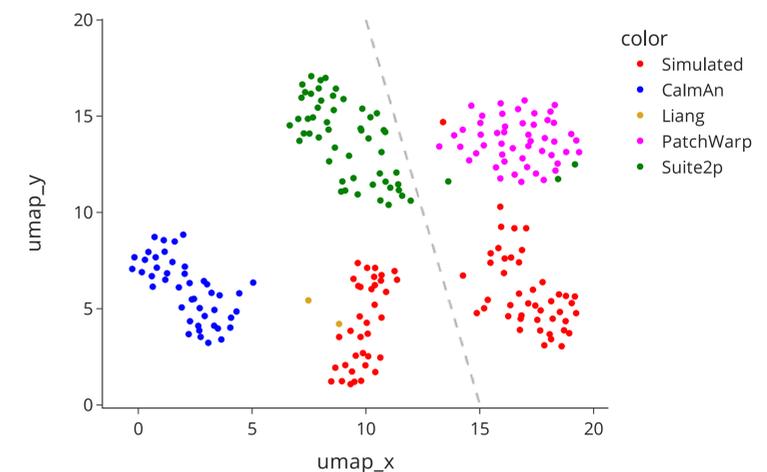
Signal	Mean	Std	Filter MSE	Period Score	Max PSF Freq	Freq Power Ratio	Stable CV	Spike CV	Total CV	Low Freq Pixel Prop
A	1760	407	4927	0.95	0.03	0.24	0.22	0.04	0.23	0.65
B	7664	4432	10 ⁷	0.11	0.03	0.007	0.56	0.12	0.58	0.15

(A) Signal from one cell in Naomi Simulation (B) Signal from one cell in CalmAn.

Filter MSE: The squared difference after applying the low-passing filter. **Period Score:** The periodicity of a signal. **Max PSF Freq:** the frequency with highest energy. **Freq Power Ratio:** the ratio of energy at the dominant frequency. **CV:** coefficient of variation. **Low Freq Pixel Prop:** proportion of pixel with low intensities, which reflects pixel distribution throughout the dataset.

4. UMAP clustering results for temporal and spatial features shows clear separation for datasets with good/poor qualities

- Datasets are clustered into 3 groups containing CalmAn on the bottom left, Good NAOMi+Suite2p+Liang in the middle, and Bad NAOMi+PatchWarp on the right, with some outliers
- Datasets to the left of the dashed line are considered as "Good" for labeling, others are considered as "Bad."
- Labels are used to training classification model.



5. Classification reveals quality of testing sets

Model	New Data Accuracy	Whole Test Accuracy
Logistic Regression	1	0.9306
SVM	1	0.9167
KNN	1	0.7083

Three common machine learning algorithms (Log Regression, SVM, and KNN) were implemented and compared. 30% of the original dataset and data from new independent sources were used for grid search and testing. New sets were labeled by inspection.

Conclusion

Through a comprehensive pipeline, we have successfully extracted quality characteristics from calcium imaging data and distinguished between high- and low-quality datasets through clustering. Using this workflow and trained data, we were able to develop a robust classifier that provides quality assessments to aid researchers.

For future work, we will refine our metrics and collect more data sets to enhance our efforts and improve the overall robustness of our work. We will also recruit experienced experts to provide more accurate labels to evaluate the accuracy of our clustering results.

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