## Automated Computer-Vision Analysis Pipeline of Calcium Imaging Neuronal Network Activity Data

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Abstract : Introduction: Calcium imaging is an established technique in neuroscience research for detecting activity in neural networks. Bursts of action potentials in neurons lead to transient increases in intracellular calcium visualized with fluorescent indicators. Manual identification of cell bodies and their contours by experts typically takes 10-20 minutes per calcium imaging recording. Our aim, therefore, was to design an automated pipeline to facilitate and optimize calcium imaging data analysis. Our pipeline aims to accelerate cell body and contour identification and production of graphical representations reflecting changes in neuronal calcium-based fluorescence. Methods: We created a Python-based pipeline that uses OpenCV (a computer vision Python package) to accurately (1) detect neuron contours, (2) extract the mean fluorescence within the contour, and (3) identify transient changes in the fluorescence due to neuronal activity. The pipeline consisted of 3 Python scripts that could both be easily accessed through a Python Jupyter notebook. In total, we tested this pipeline on ten separate calcium imaging datasets from murine dissociate cortical cultures. We next compared our automated pipeline outputs with the outputs of manually labeled data for neuronal cell location and corresponding fluorescent times series generated by an expert neuroscientist. Results: Our results show that our automated pipeline efficiently pinpoints neuronal cell body location and neuronal contours and provides a graphical representation of neural network metrics accurately reflecting changes in neuronal calcium-based fluorescence. The pipeline detected the shape, area, and location of most neuronal cell body contours by using binary thresholding and grayscale image conversion to allow computer vision to better distinguish between cells and non-cells. Its results were also comparable to manually analyzed results but with significantly reduced result acquisition times of 2-5 minutes per recording versus 10-20 minutes per recording. Based on these findings, our next step is to precisely measure the specificity and sensitivity of the automated pipeline's cell body and contour detection to extract more robust neural network metrics and dynamics. Conclusion: Our Python-based pipeline performed automated computer vision-based analysis of calcium image recordings from neuronal cell bodies in neuronal cell cultures. Our new goal is to improve cell body and contour detection to produce more robust, accurate neural network metrics and dynamic graphs.

Keywords : calcium imaging, computer vision, neural activity, neural networks

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