Jul 10, 2024

Mouse astrocyte territory volume analysis

DOI
dx.doi.org/10.17504/protocols.io.q26g7yoo3gwz/v1

Shiyi Wang[1]

[1]Duke University

ASAP Collaborative Rese…

Shiyi Wang
Duke University

OPEN ACCESS

DOI: dx.doi.org/10.17504/protocols.io.q26g7yoo3gwz/v1

Document Citation: Shiyi Wang 2024. Mouse astrocyte territory volume analysis. **protocols.io** https://dx.doi.org/10.17504/protocols.io.q26g7yoo3gwz/v1

Created: May 23, 2023

Last Modified: July 10, 2024

Document Integer ID: 82331

Keywords: ASAPCRN

Funders Acknowledgement:
Aligning Science Across Parkinson's (ASAP) initiative
Grant ID: ASAP-020607

Disclaimer

DISCLAIMER – FOR INFORMATIONAL PURPOSES ONLY; USE AT YOUR OWN RISK

The protocol content here is for informational purposes only and does not constitute legal, medical, clinical, or safety advice, or otherwise; content added to **protocols.io** is not peer reviewed and may not have undergone a formal approval of any kind. Information presented in this protocol should not substitute for independent professional judgment, advice, diagnosis, or treatment. Any action you take or refrain from taking using or relying upon the information presented here is strictly at your own risk. You agree that neither the Company nor any of the authors, contributors, administrators, or anyone else associated with **protocols.io**, can be held responsible for your use of the information contained in or linked to this protocol or any of our Sites/Apps and Services.

Abstract

Mouse astrocyte territory volume analysis

1. To assess the territory volume of individual astrocytes in the mouse cortex, 100 µm-thick floating sections containing anterior cingulate cortex (ACC) and primary motor cortex (MOp) astrocytes labeled sparsely via PALE with mCherry-CAAX were collected.

2. High-magnification images containing an entire astrocyte (50-60 µm z-stack) were acquired with an Olympus FV 3000 microscope with the 60× objective.

3. Criteria for data inclusion required that the entirety of the astrocyte could be captured within a single brain section and that the astrocyte was in layer 2/3 of the ACC or MOp.

4. Astrocytes in which the entire astrocyte could not be captured within the section or was in other layers or outside of the ACC or MOp were not imaged.

5. Imaged astrocytes were analyzed using Imaris Bitplane software.

6. Surface reconstructions were generated, and the Imaris Xtensions “Visualize Surface Spots” and “Convex Hull” were used to create an additional surface render representing the territory of the astrocyte.

7. The volume of each territory was recorded, and astrocyte territory sizes from biological replicates were analyzed across experimental conditions using a nested two-way ANOVA followed by Bonferroni posthoc test.

8. For 3D Sholl analysis of individual PALE astrocytes, we first loaded images onto Imaris and then created a surface.

9. After generating the surface of astrocytes, we created filaments using ‘Add new filament (leaf icon)’.

10. For the quantification of complexity, we clicked on the gear tool on Imaris to display Sholl intersections.

11. The number of animals and cells/animals analyzed are specified in the figure legend for each experiment.