Jul 11, 2024

Tile Scan Imaging and Cell Counting

DOI
dx.doi.org/10.17504/protocols.io.bp2l6231kgqe/v1

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OPEN ACCESS

DOI: dx.doi.org/10.17504/protocols.io.bp2l6231kgqe/v1

Protocol Citation: Shiyi Wang 2024. Tile Scan Imaging and Cell Counting. **protocols.io** https://dx.doi.org/10.17504/protocols.io.bp2l6231kgqe/v1

Protocol status: Working
We use this protocol and it's working

Created: July 11, 2024

Last Modified: July 11, 2024

Protocol Integer ID: 103200

Keywords: ASAPCRN

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

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Abstract

Tile Scan Imaging and Cell Counting

1 **Sample Preparation**

1.1 - Prepare 30 µm thick coronal sections containing the anterior cingulate cortex (ACC) and primary motor cortex (MOp) from P21 WT and LRRK2 G2019Ski/ki; Aldh1l1-eGFP mice.

2 **Confocal Microscopy**

2.1 - Acquire tile scan images using a confocal Leica SP8 STED microscope equipped with a galvo scanner and a 20x objective.

2.2 - Ensure consistent settings for image acquisition across all samples.

3 **Double Positive Cell Identification**

3.1 - Identify cells labeled with ALDH1L1-EGFP (astrocyte marker) and SOX9 (astrocyte marker) in the ACC and MOp regions.

3.2 - Use FIJI software with the cell counter tool for manual cell counting.

4 **Data Collection**

4.1 - Count ALDH1L1-EGFP and SOX9 double positive cells manually in 2-4 sections per brain.

4.2 - Analyze sections from 3 sex- and age-matched mice per genotype.