Drp1-mediated mitochondrial fission controls cortical astrocyte organization and development

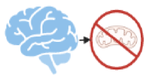
Sprihaa Kolanukuduru[1,2], Pia Rodriguez[2,3], Cagla Eroglu[3,4,5]

1. Department of Psychology and Neuroscience, UNC-Chapel Hill; 2. Cell and Molecular Biology Program, Duke University; 3. Department of Cell Biology, Duke University; 4. Department of Neurobiology, Duke University; 5. Howard Hughes Medical Institute

Background

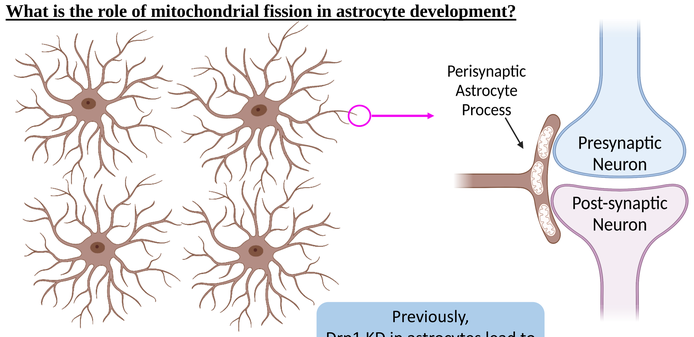
Mitochondrial fission is required for proper brain development

Fission (Drp1)



Mitochondrial defects cause neurodevelopmental disorders:
- Autism Spectrum Disorder
- Childhood Refractory Epilepsy

What is the role of mitochondrial fission in astrocyte development?



Previously, Drp1 KD in astrocytes lead to astrocyte clusters up to P21

How does Drp1-mediated mitochondrial fission regulate astrocyte organization and development in the mouse cortex?

Drp1 KD caused astrocyte clustering in the adult mouse cortex

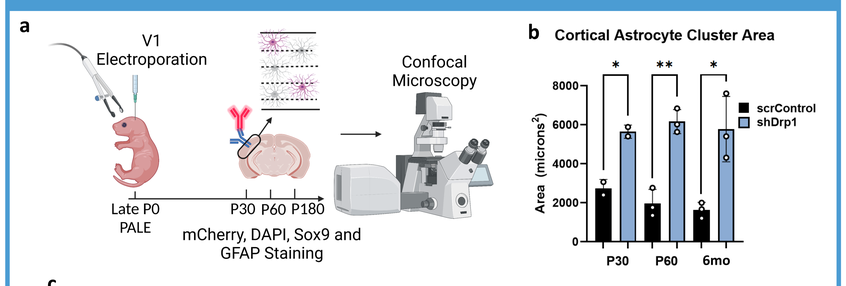


Figure 1. Knockdown of Drp1-mediated mitochondrial fission disrupts astrocyte tiling. a. PALE was performed on P0 mice pups injected with either scrControl or shDrp1 constructs and collected at P21, P30, P60, and P180. b. Quantification of cluster area in control and shDrp1 astrocytes, n=3 animals per condition, *p<0.05, unpaired t-test. Bars are Mean ± SEM c. Representative images of control (top) and shDrp1 (bottom) P30, P60, and P180 visual cortex PALE brains.

Drp1 KD disrupts cortical astrocyte organization in development and adulthood

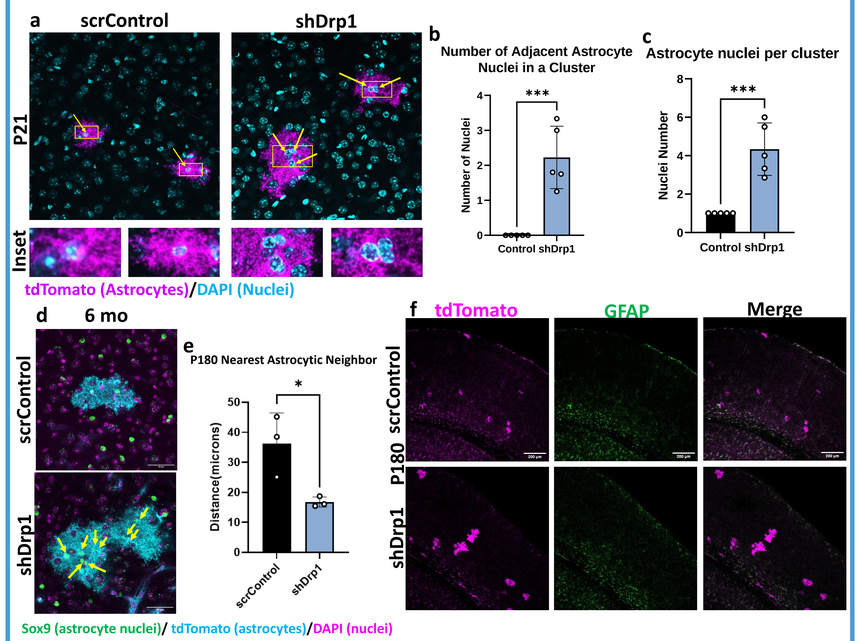


Figure 2. Sparse knockdown of Drp1-mediated mitochondrial fission disrupts astrocyte tiling. a. Representative images of control (left) and shDrp1 (right) astrocyte clusters with zoomed in images of nuclei (bottom) in P21 visual cortex PALE brains. b. Quantification of number of adjacent astrocyte nuclei per cluster in control and shDrp1 astrocytes, n=5 animals per condition, ***p<0.001, unpaired t-test. Bars are Mean ± SEM. c. Quantification of number of astrocyte nuclei per cluster in control and shDrp1 astrocytes, n=5 animals per condition, ***p<0.001, unpaired t-test. Bars are Mean ± SEM. d. Representative images for Sox9 (green) and Dapi (magenta) in P180 control (top)and shDrp1 (bottom) astrocyte clusters (cyan). e. Quantification of nearest neighbor distance between Sox9+ astrocytic nuclei in P180 control vs. shDrp1 astrocyte clusters, n=3 animals per condition, *p<0.05, unpaired t-test. Bars are Mean ± SEM f. Representative images of P180 control (top) and shDrp1 (bottom) visual cortex PALE brain astrocytes (magenta) and GFAP (green) expression.

Astrocytic-specific Drp1 conditional knockout increases GFAP expression and decreases cortical astrocyte coverage

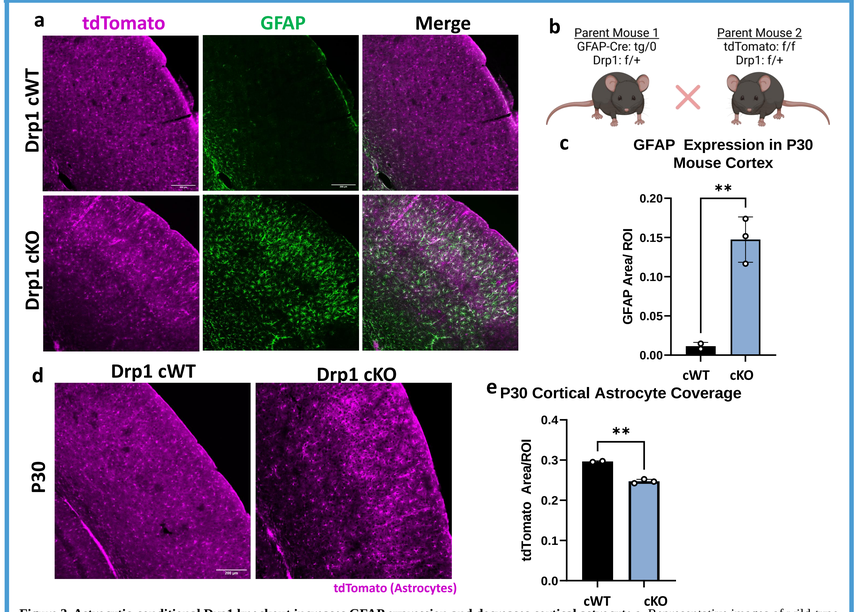
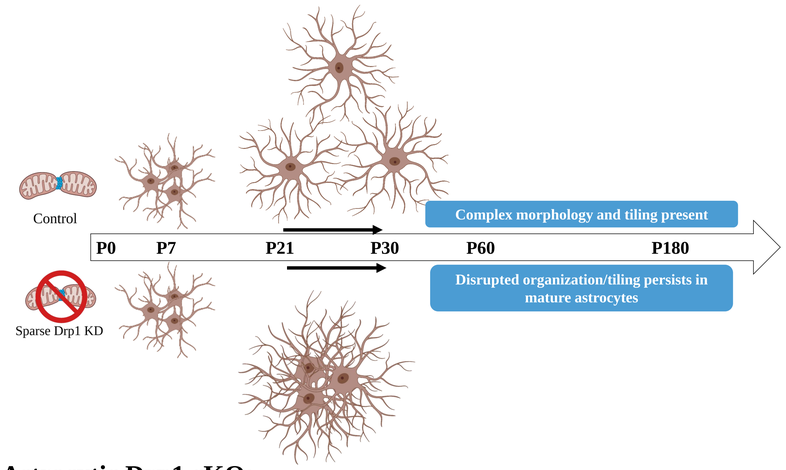


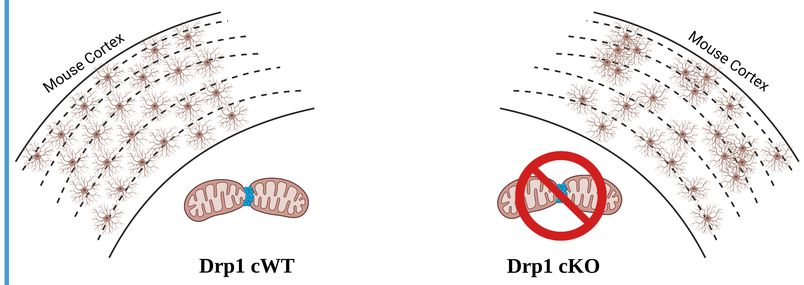
Figure 3. Astrocytic-conditional Drp1 knockout increases GFAP expression and decreases cortical astrocyte a. Representative images of wild-type (top) and Drp1 cKO (bottom) P30 visual cortex astrocytes (magenta) and GFAP (green) expression. **coverage** b. Breeding scheme of mice for collection and analysis. c. Quantification of GFAP expression in wild-type and Drp1 cKO astrocytes, n=2/3 animals per condition, unpaired t-test, **p<0.01. Bars are Mean ± SEM. d. Representative images of P30 wild-type (left) and Drp1 cKO (right) visual cortex astrocytes (magenta) e. Quantification of total coverage of labeled astrocytes in wild-type and Drp1 cKO P30 visual cortex, unpaired t-test, **p<0.01. Bars are Mean ± SEM.

Key Findings

Sparse Astrocyte Drp1 KD:



Astrocytic Drp1 cKO:



- Even coverage of astrocytes across the cortex
- Little to no cortical GFAP expression

- Decreased astrocyte coverage
- Increased GFAP expression
  - Dysfunction?

Future Directions

- Does astrocytic cKO of Drp1 cause disorganization of astrocytes in the cortex?
- Do Drp1-deficient astrocytes have decreased synaptogenic ability compared to the wild-type?
- Does the disruption of astrocytic tiling induced by Drp1 KD in astrocytes disrupt gap junction coupling?

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