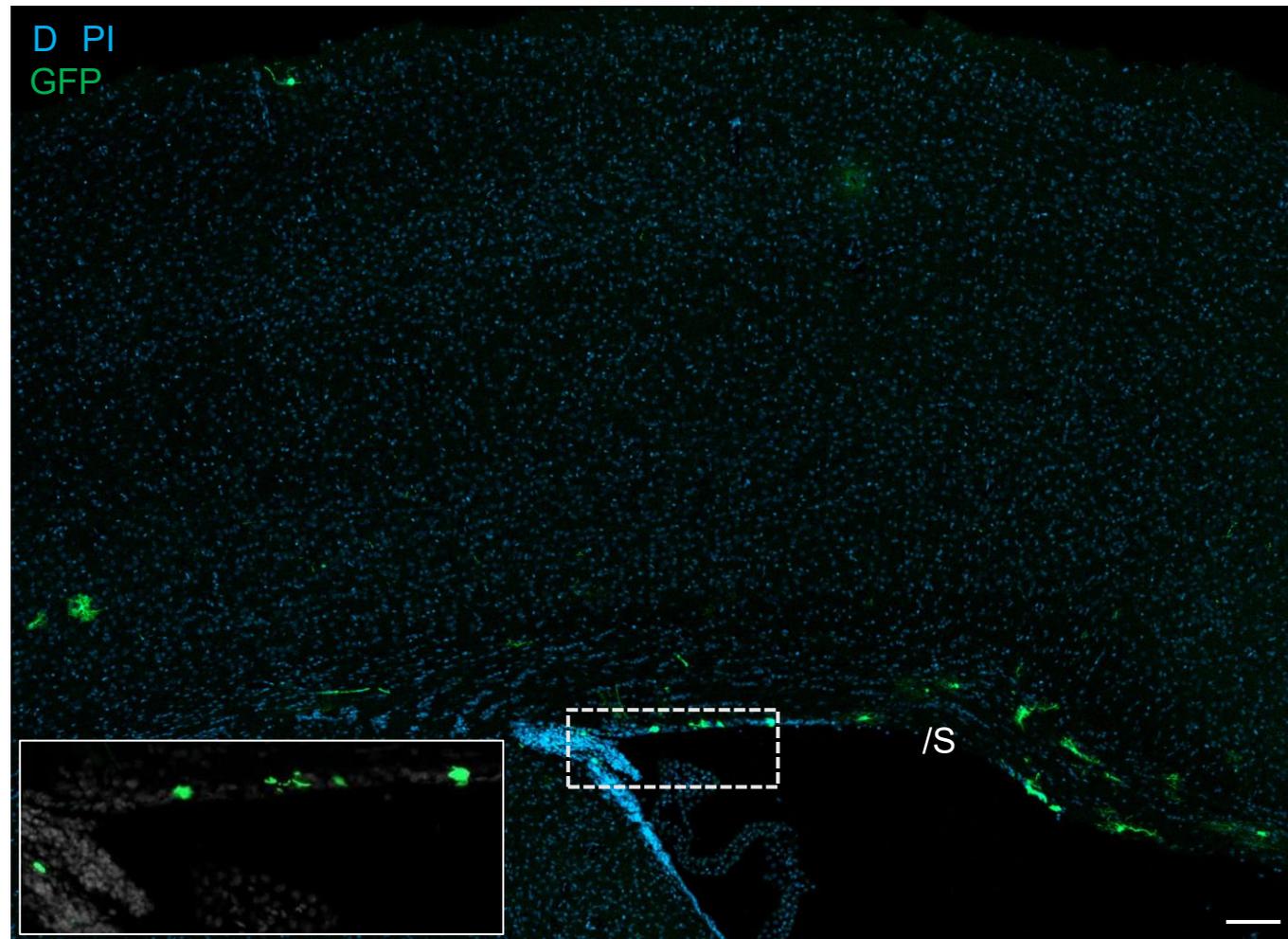


P - G EGFP P0 P21



P - G EGFP E16.5 P21

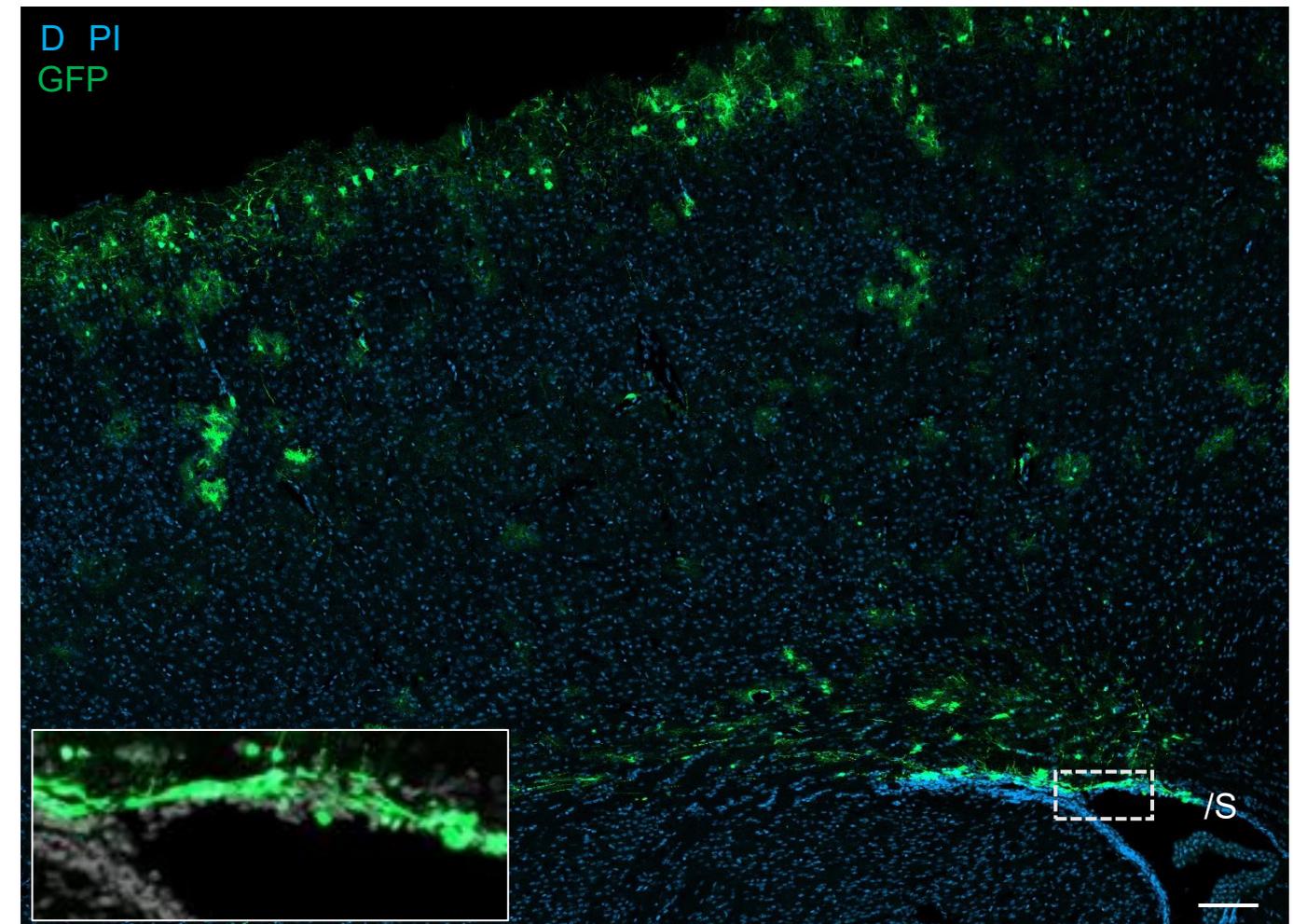


FIGURE S1. Piggy back fate mapping of β -S progenitor cells observed at P21.

Representative images show distribution of EGFP+ cells in E16.5 (left) and P0 (right) electroporated cortices at P21. Magnification images show labeled EGFP+ progenitors in the β -S corpus callosum (cc). Scale bar = 100 μ m.

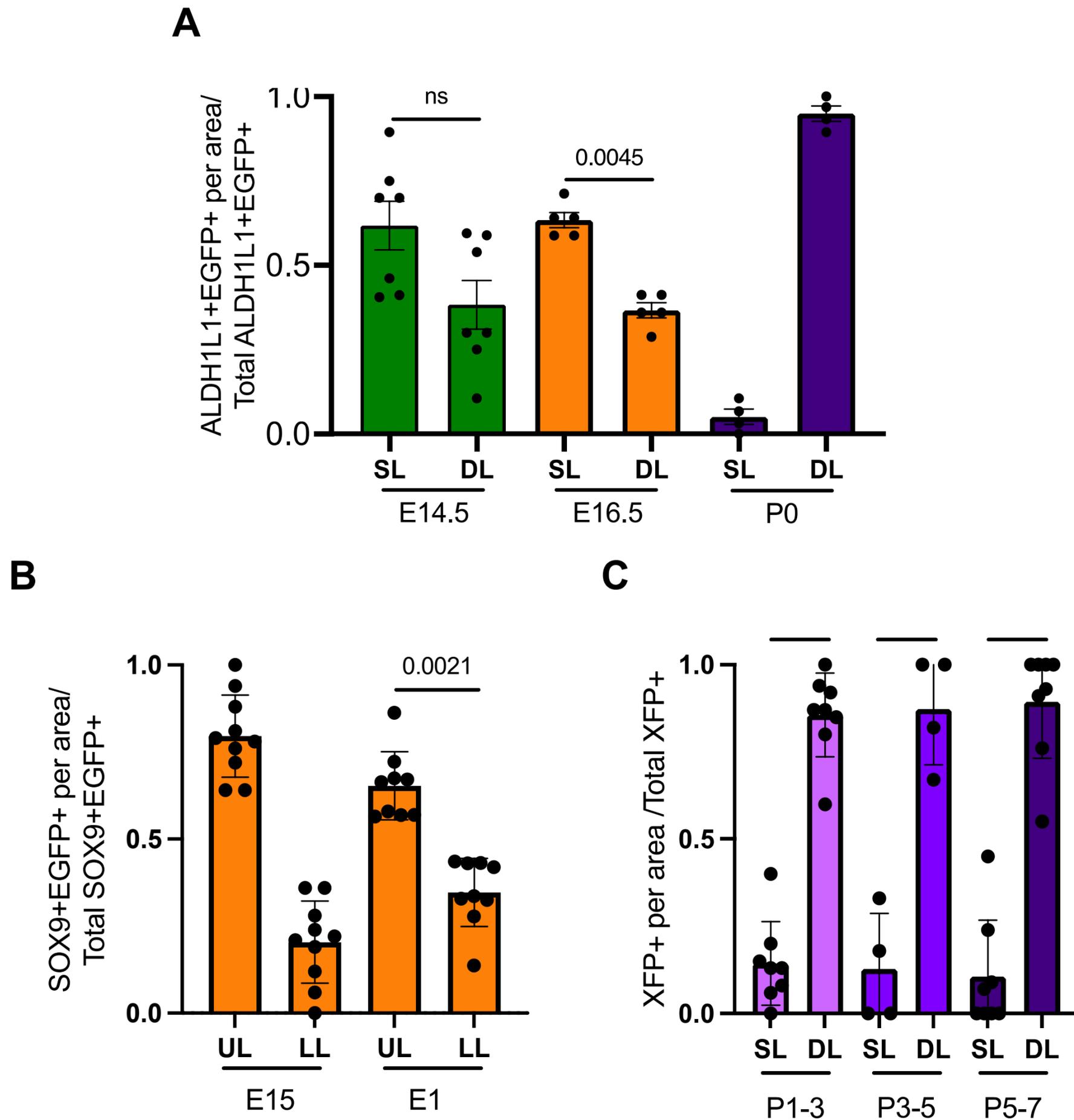


FIGURE S2. *PiggyBac* fate mapping of -S progenitor cells from E14.5 to postnatal period.

(a) Quantification of E14.5, E16.5, and P0-labeled EGFP+ LD 1L1+ astrocytes at P21.

(b) Quantification of E15.5- and E1 .5-labeled EGFP+SOX9+ astrocytes at P21.

(c) Quantification of XFP+ labeled astrocytes following T administration during three postnatal windows P1 P3, P3 P5, and P5 P .

Source data is provided in Table S1.

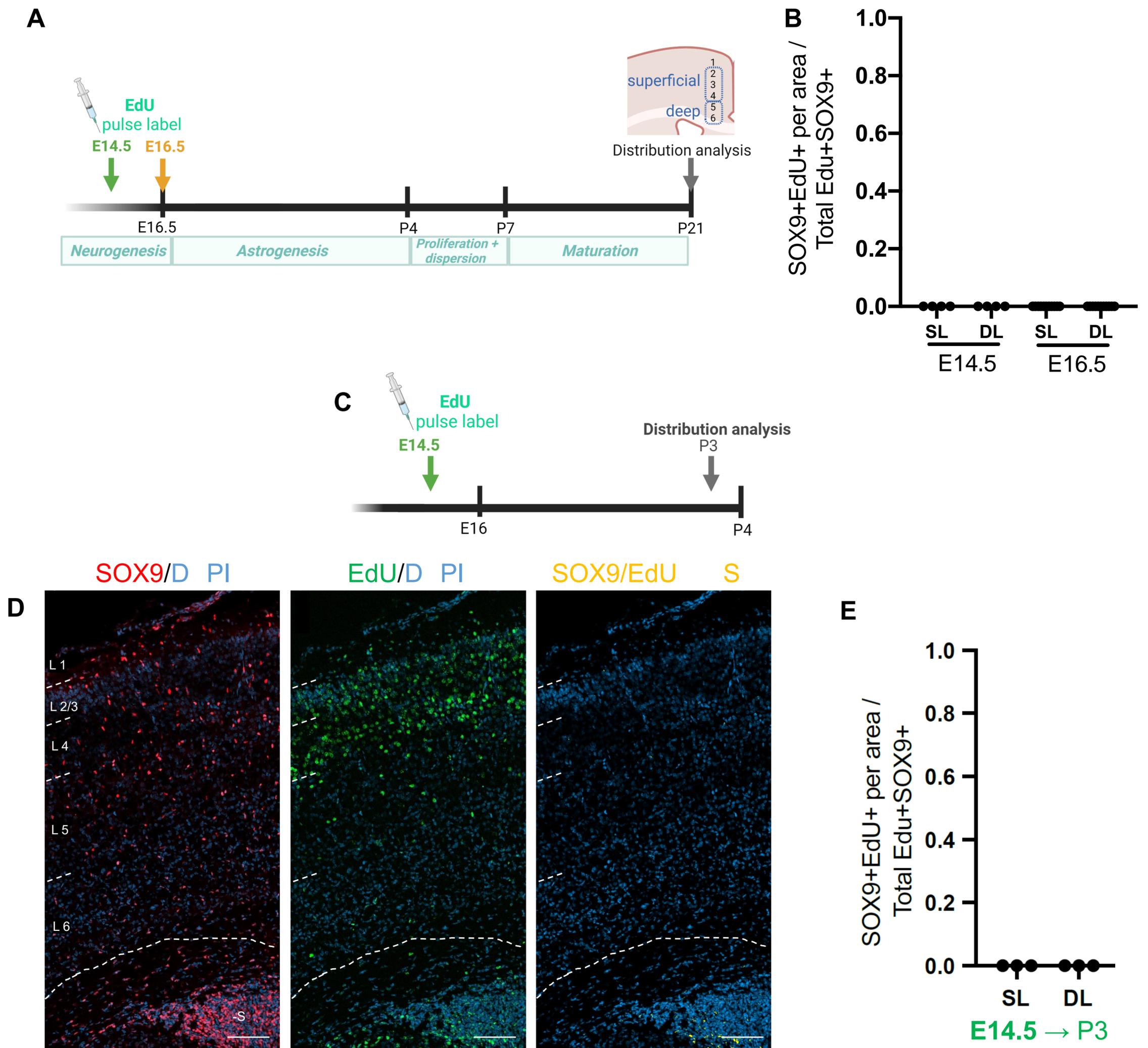


FIGURE S3. EdU labeling at E14.5 or E16.5 is not detected in SOX9+ astrocytes at P21.

(a) Schematic of experimental design. EdU pulse-labeling was performed at E14.5 or E16.5 and cortices were analyzed at P21.

(b) Quantification of E14.5- and E16.5-labeled SOX9+EdU+ astrocytes at P21.

(c) Schematic of experimental design. EdU pulse-labeling was performed at E14.5. Cortices were analyzed at P3.

(d) Representative images show distribution of SOX9+ astrocytes in an E14.5 EdU pulse-labeled cortex at P3. Third panel shows absence of EdU+SOX9+ cells represented by absence of colocalization mask in yellow. Dotted lines show demarcation of cortical layers (L1, L2/3, L4, L5, and L6) and corpus callosum (cc). Labeled in left most panel. Scale bar = 100 μ m.

(e) Quantification of E14.5-labeled SOX9+EdU+ astrocytes at P3.

Source data is provided in Table S1.

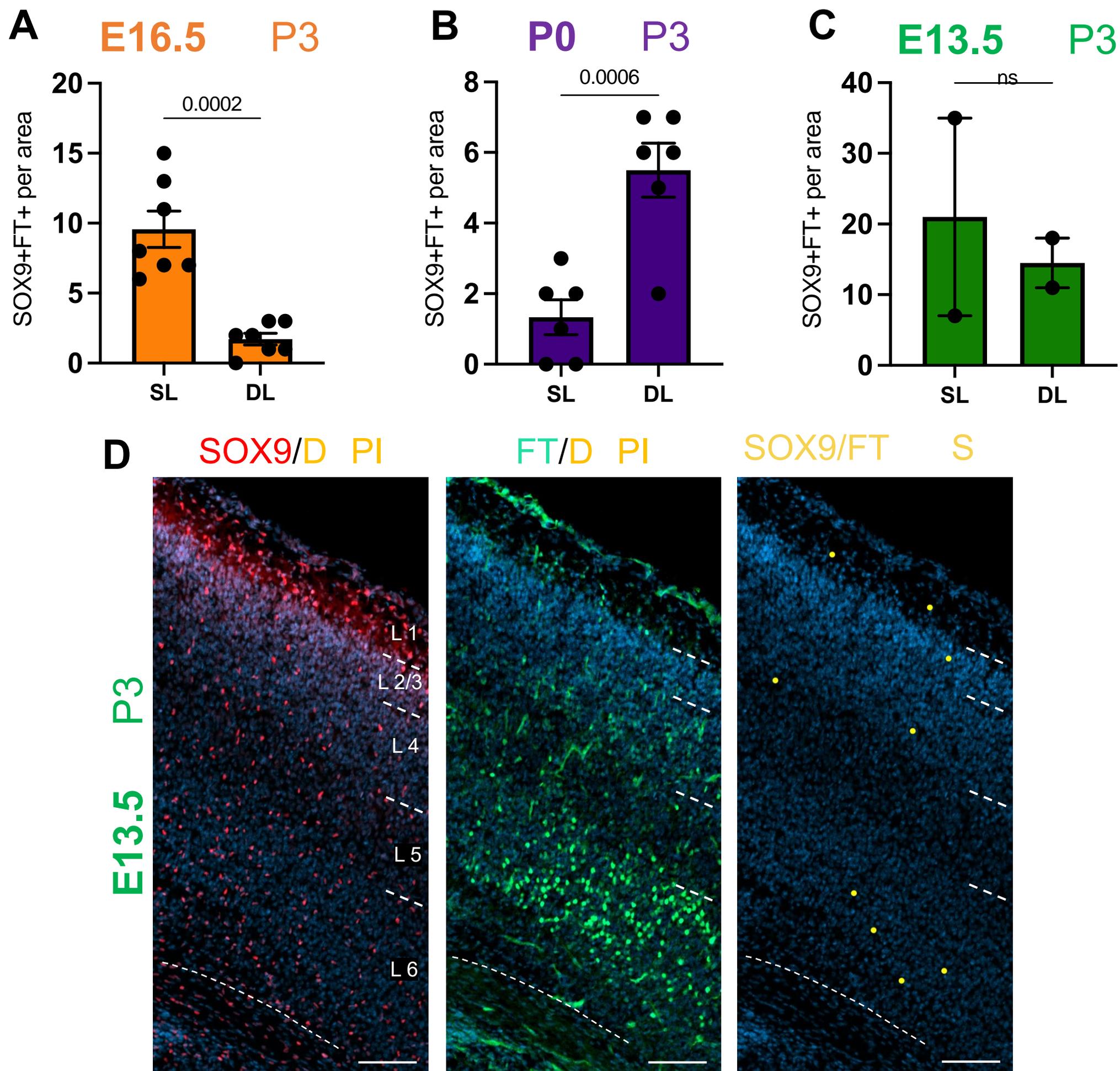


FIGURE S4. FlashTag labeling of E13.5 apical radial glia shows no preferential localization of astrocytes in the superficial and deep cortical layers at P3.

(a) Quantification of E16.5-labeled FT+SOX9+ astrocytes at P3.

(b) Quantification of P0-labeled FT+SOX9+ astrocytes at P3.

(c) Quantification of E13.5-labeled FT+SOX9+ astrocytes at P3.

(d) Representative images show distribution of SOX9+ astrocytes in a E13.5 FT pulse-labeled cortex at P3. Third panel shows distribution of FT+SOX9+ cells represented by yellow mask. Dotted lines show demarcation of cortical layers (L1, L2/3, L4, L5, and L6) and corpus callosum (cc). Labeled in left most panel. Scale bar = 100 μ m.

Source data is provided in Table S1.

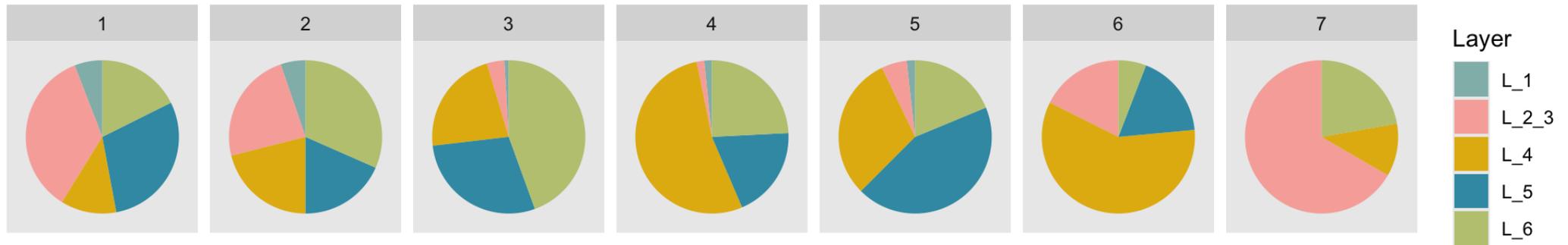
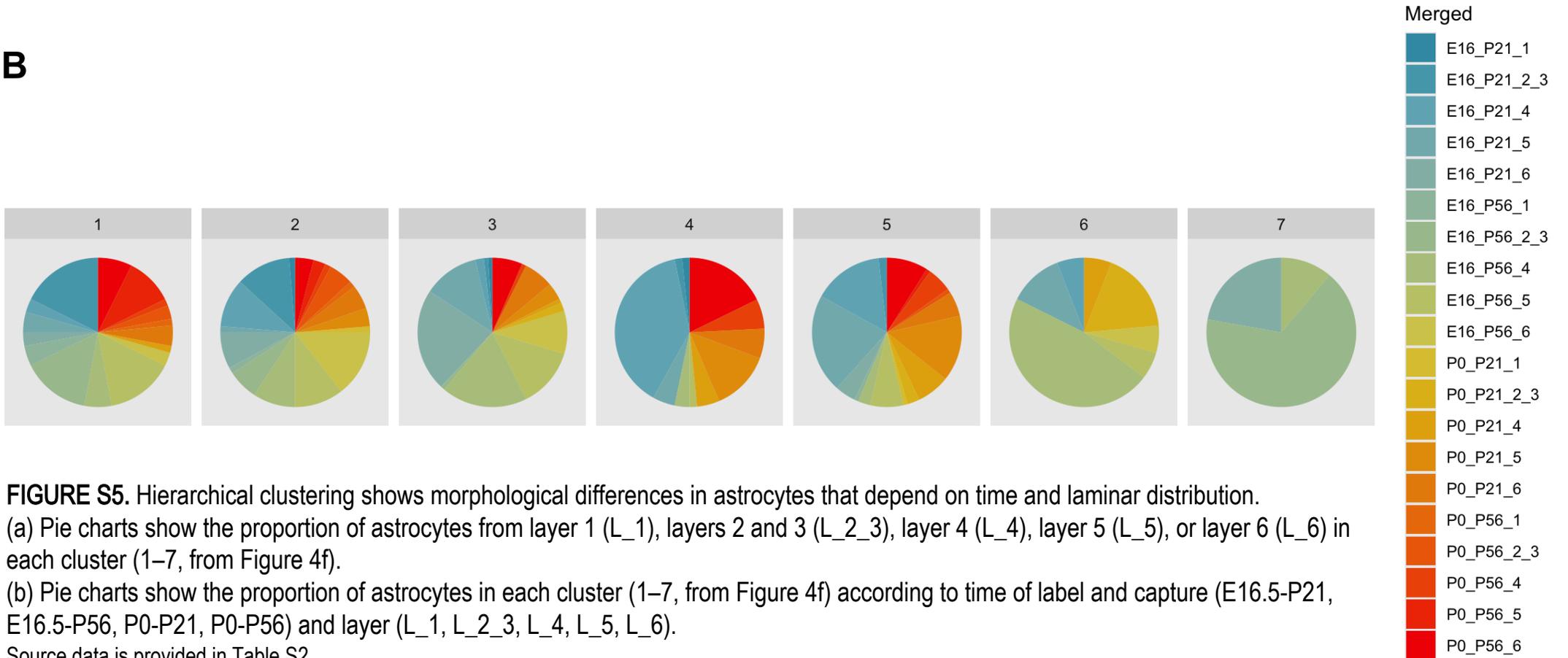
A**B**

FIGURE S5. Hierarchical clustering shows morphological differences in astrocytes that depend on time and laminar distribution.

(a) Pie charts show the proportion of astrocytes from layer 1 (L_1), layers 2 and 3 (L_2_3), layer 4 (L_4), layer 5 (L_5), or layer 6 (L_6) in each cluster (1–7, from Figure 4f).

(b) Pie charts show the proportion of astrocytes in each cluster (1–7, from Figure 4f) according to time of label and capture (E16.5-P21, E16.5-P56, P0-P21, P0-P56) and layer (L_1, L_2_3, L_4, L_5, L_6).

Source data is provided in Table S2.