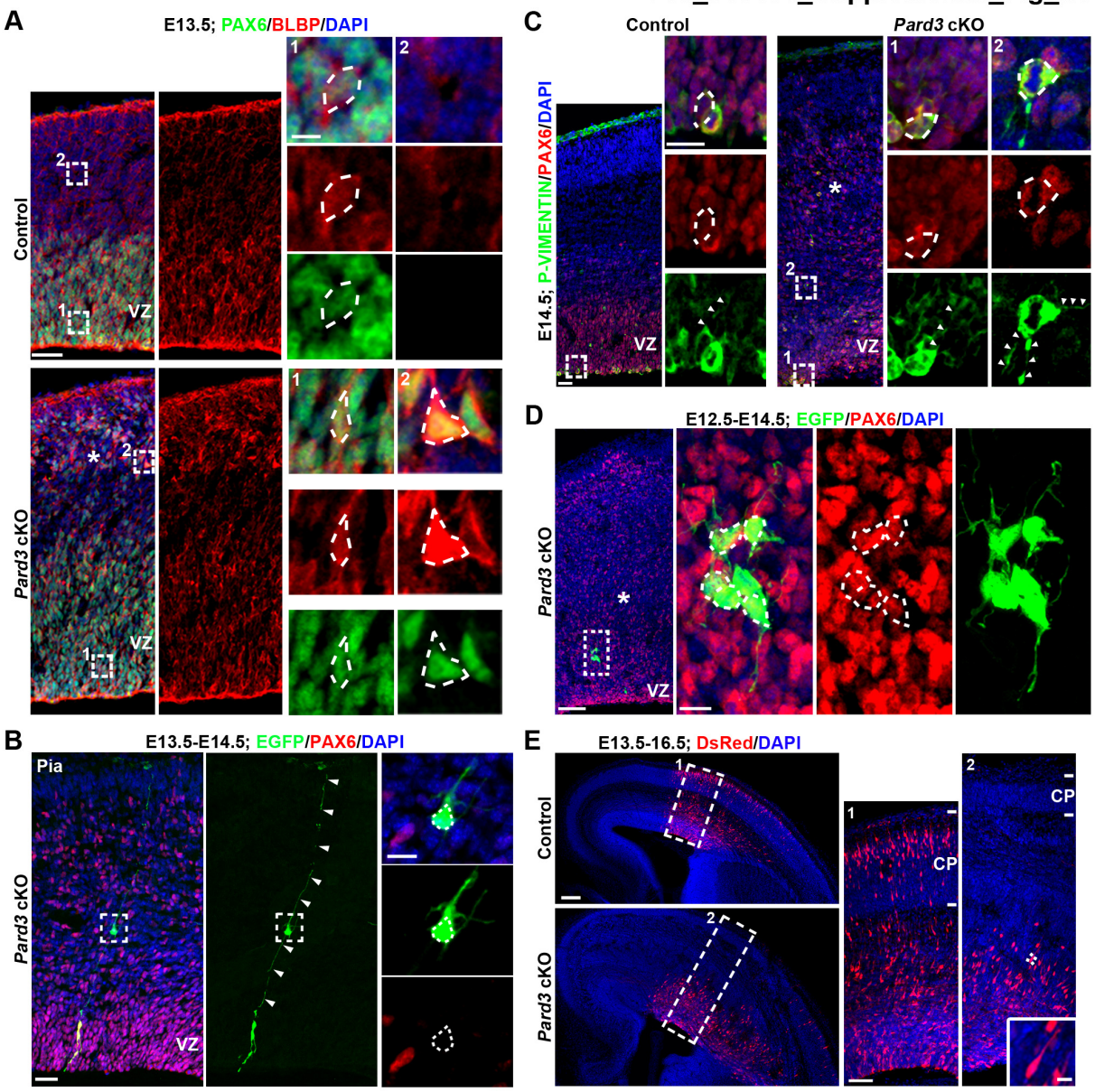
Liu_313171_Supplemental_Fig_S3



Supplemental Figure S3: Morphological analysis of RGPs in *Pard3* cKO cortex (A) Representative confocal images of E13.5 control and *Pard3* cKO cortices stained for PAX6 (green) and a bona-fide RGP marker BLBP (red), and counter-stained for DAPI (blue). High-magnification images of the VZ (areas 1) and extra-VZ (areas 2) are shown to the right. Broken line polygons indicate the cell bodies of representative progenitors in each group. Scale bars: 30 μm (left) and 5 μm (right). **(B)** Representative confocal images of the *Pard3* cKO cortex received *in utero* intraventricular injection of low-titer EGFP-expressing retrovirus at E13.5, stained for PAX6 (red) and EGFP (green), and counter-stained with DAPI (blue) at E14.5. Filled arrowheads show the long radial glial fiber of a $PAX6^+$ RGP in the VZ reaching the pia surface. High-magnification images of an EGFP-expressing, $PAX6^-$ differentiating progeny closely associated with the radial glial fiber (broken line square) are shown to the right. Scale bars: 30 μm (left) and 10 μm (right). **(C)** Representative confocal images of E14.5 control and *Pard3* cKO cortices stained for PAX6 (red) and p-VIMENTIN (green), and counter-stained for DAPI (blue). High-magnification images of dividing RGPs in the VZ (area 1) and extra-VZ (area 2) are shown to the right. Broken line polygons indicate the cell bodies of representative dividing RGPs in each group. RGP fibers are highlighted by arrowheads. Asterisk indicates ectopic $PAX6^+$ progenitors in *Pard3* cKO cortex. Scale bars: 30 μm (left) and 10 μm (right). **(D)** Representative confocal images of the *Pard3* cKO cortex received *in utero* intraventricular injection of EGFP-expressing retrovirus at E12.5, stained for PAX6 (red) and EGFP (green), and counter-stained for DAPI (blue) at E14.5. High-magnification images of EGFP-expressing $PAX6^+$ RGPs in extra-VZ (broken line rectangle) are shown to the right. Broken line polygons indicate the cell bodies of the representative RGPs. Note that the ectopic $PAX6^+$ cells grow multiple short processes. Scale bars: 30 μm (left) and 10 μm (right). **(E)** Representative confocal images of the control and

Pard3 cKO cortices received *in utero* intraventricular electroporation of DsRed-expressing plasmid at E13.5, stained for PAX6 (red) and EGFP (green), and counter-stained with DAPI (blue) at E16.5. High-magnification images (broken line areas 1 and 2) are shown to the right. Image of representative cells with a bipolar morphology in the SVZ/IZ is shown as the inset. Note that, while a significant fraction of DsRed-expressing cells migrate to the cortical plate in the control cortex, very few DsRed-expressing cells are found in the cortical plate of the *Pard3* cKO cortex (control, 31.0±2.4%; *Pard3* cKO, 1.9±1.1%; n=3 per genotype; $p<0.0001$; unpaired two-tailed t-test with Welch's correction; mean ± SEM). Scale bars: 150 μm (left) and 30 μm (middle) and 10 μm (right).