

Fig. S1 (related to Figs. 1, 2, 3). Wnt/β-catenin signaling is activated in retinal ECs and mural cells, but not in astrocytes. **A)** Analyses of a P14 WT retinal single cell RNAseq database (see Results and Materials and Methods) to explore expression profiles of downstream Wnt/β-catenin targets (*Lef1*, *Apcdd1*, *Axin2*) in endothelial cells (ECs), pericytes (PCs) and astrocytes. **B-E)** P10 WT retinal flat-mounts were stained for *Lef1* and *Pdgfrβ* (A= artery, V= vein, C= capillary). Solid arrowheads point to *Lef1*⁺ mural cells. Dotted bar graph shows number of *Lef1*⁺ mural cells in unit length for each vessel type (n=4 animals). **F-K)** Fluorescence *in situ* hybridization of P10 WT retinal sections with antisense probe against *Apcdd1* mRNA followed by immunostaining for Caveolin-1 or *Pdgfrβ* proteins. Solid arrowheads and empty arrowheads point to localization of *Apcdd1* mRNA inside and outside the corresponding cells marker, respectively. Student's t-test: **p<0.02, NS: not significant. Graphs show mean +/- SEM. Scale bars: B-D = 41 μm.

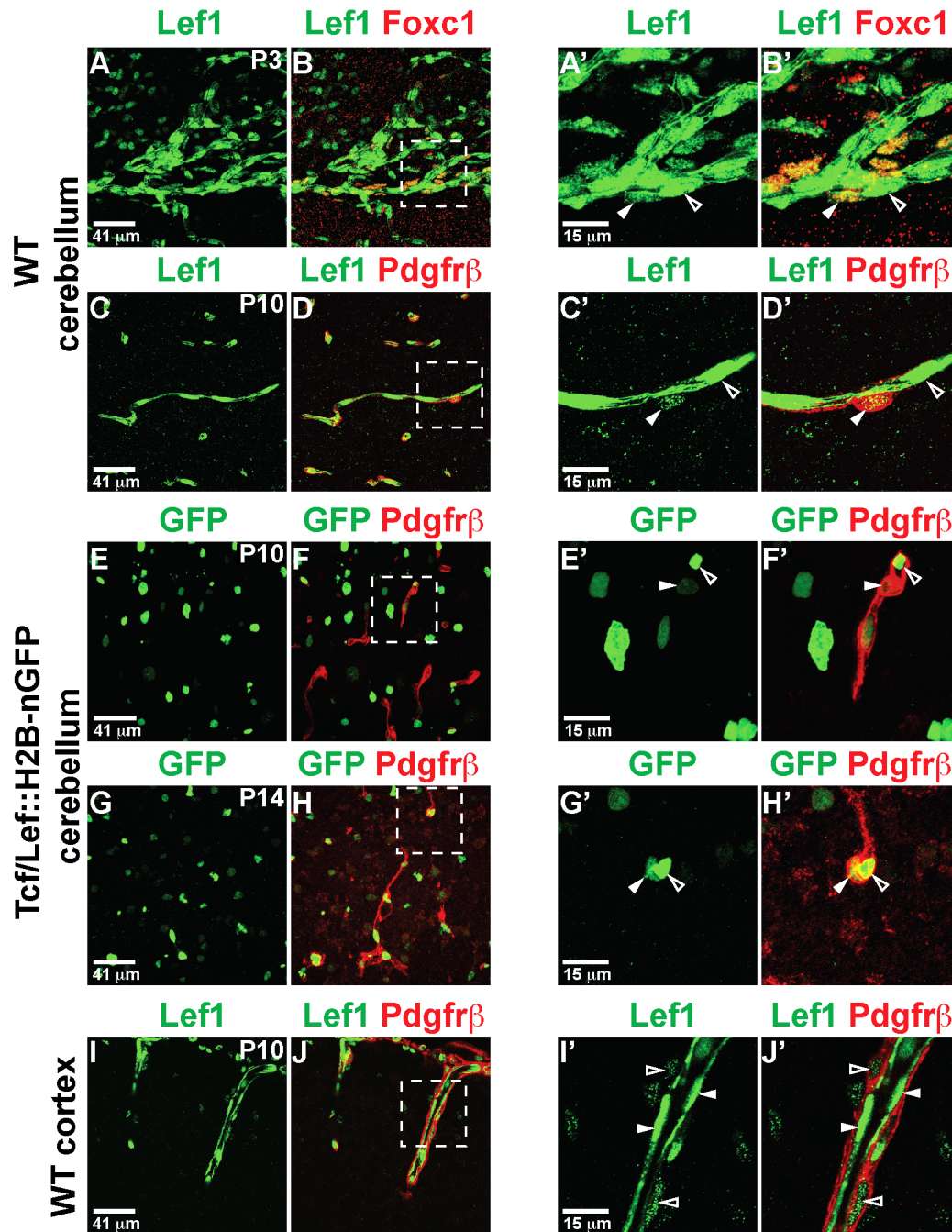


Fig. S2 (related to Figs. 1, 2). Wnt/ β -catenin signaling is activated in cerebellar and cortical mural cells. **A-B')** P3 WT cerebellar sections were stained for Lef1 and Foxc1. **C-D')** P10 WT cerebellar sections were stained for Lef1 and Pdgfr β . **E-H')** P10 and P14 Tcf/Lef::H2B-nGFP cerebellar sections were stained for GFP and Pdgfr β . In all images empty arrowheads point to Lef1⁺ or GFP⁺ ECs, whereas solid arrowheads point to Lef1⁺ or GFP⁺ mural cells. Scale bars: A-J = 41 μ m; A'-J' = 15 μ m.

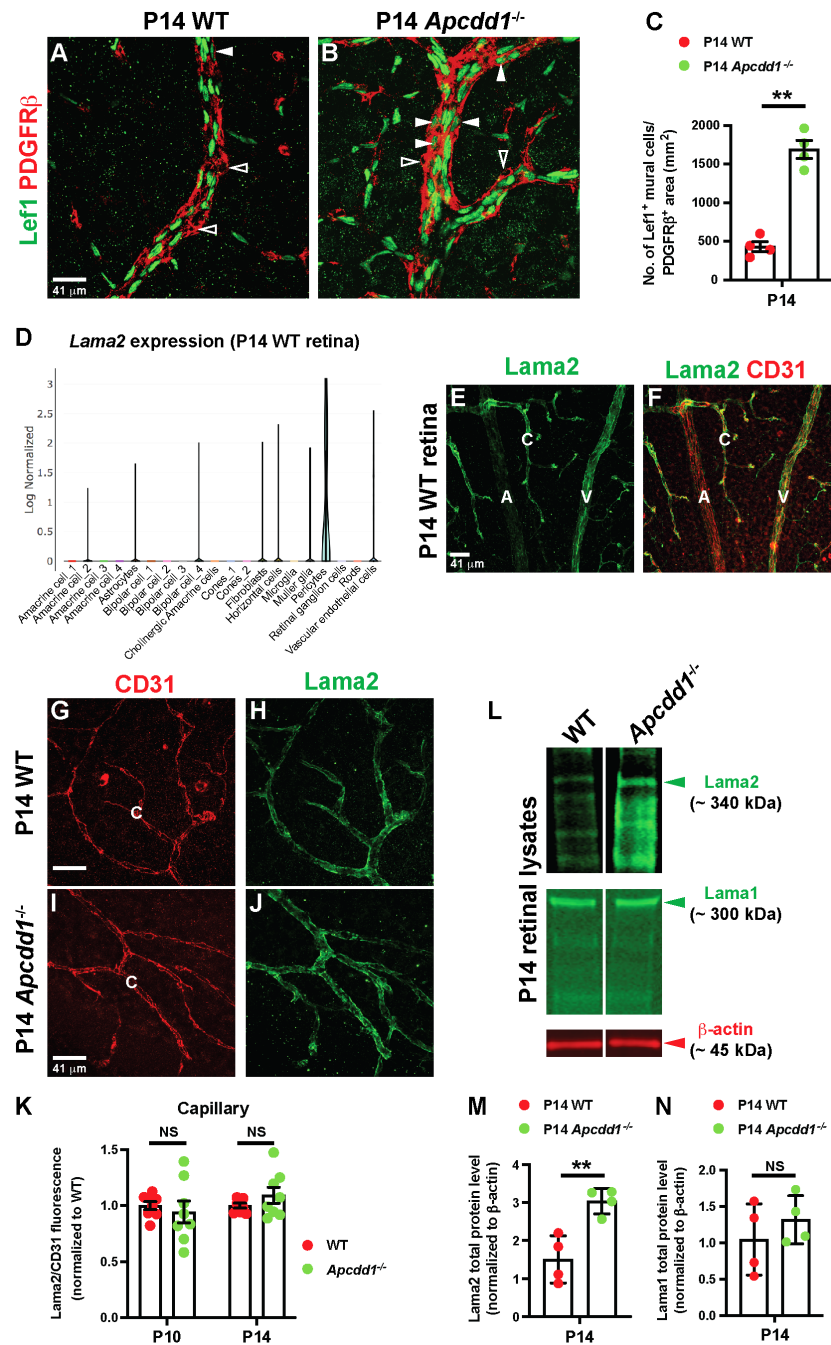


Fig. S3 (related to Fig. 3). Lama2 (*Lama2*) is predominantly expressed in retinal PCs, but not in ECs, and is upregulated in the *Apcdd1*^{-/-} retina. A-C) P14 WT and *Apcdd1*^{-/-} retinal flat-mounts were stained

for *Lef1* and *Pdgfr β* . Solid arrowheads point to *Lef1*⁺ mural cells, empty arrowheads point to *Lef1*⁻ mural cells. Dotted bar graph shows the number of *Lef1*⁺ mural cells (n=4 animals/genotype). **D**) Expression profile of *Lama2* mRNA in P14 WT retinal cells from the published retina single cell RNAseq database. **E**, **F**) P14 WT retinal flat mounts were stained for CD31 and Lama2 proteins. A= artery, V= vein, C= capillary. **G-K**) P14 WT and *Apcdd1*^{-/-} retinal flat-mounts were labelled for Lama2 and CD31, showing retinal capillaries (C). Dotted bar graphs show the ratio of Lama2/CD31 M.F.I in capillaries normalized to the WT average values at P10 and P14 (8 fields of capillary network from n=4 mice/genotype). **L**) P14 WT and *Apcdd1*^{-/-} whole retinal lysates were probed for Lama1 or Lama2 (green bands) and β -actin (red bands) by western blot. **M, N**) Quantification of Lama1 and Lama2 protein levels (normalized to β -actin) by western blotting (n=4 samples/group). Student's t-test: **p<0.02, NS= not significant. Graphs show mean \pm SEM (C, K) or mean \pm SD (M, N). Scale bars: 41 μ m.

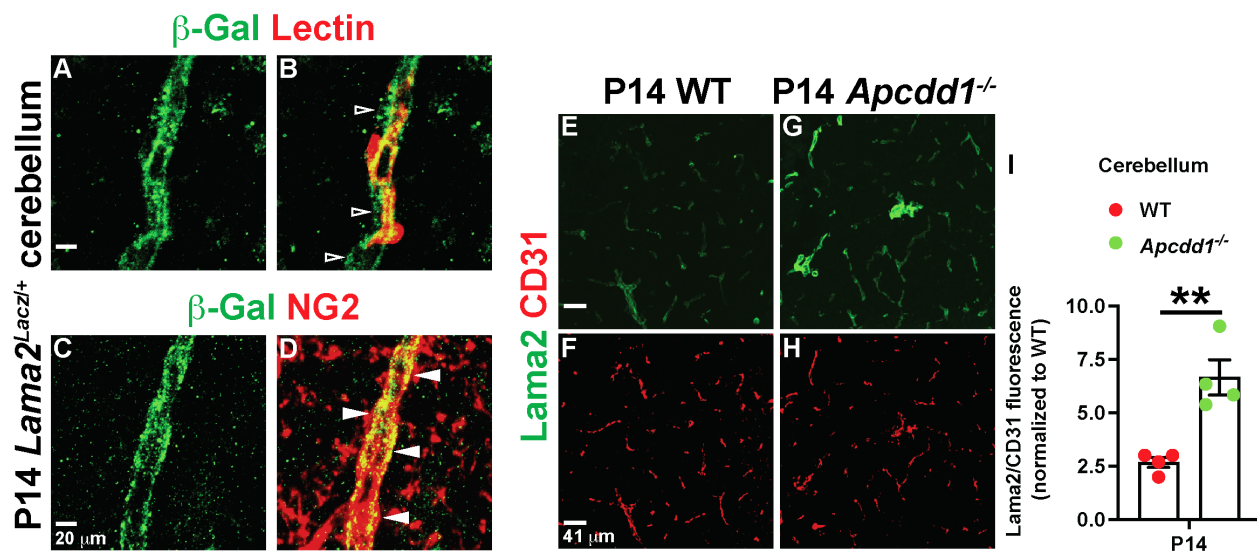


Fig. S4 (related to Fig. 3). Mural cell-derived Lama2 expression and deposition are upregulated in the *Apcdd1^{-/-}* cerebellar vascular basement membrane. A-D) P14 *Lama2^{LacZ/+}* cerebellar sections were stained for β-galactosidase (β-Gal) and either Lectin (A-B) or NG2 (C-D). Empty arrowheads point to β-Gal⁻ ECs. Solid arrowheads point to β-Gal⁺ mural cells. E-I) P14 WT and *Apcdd1^{-/-}* cerebellar sections were immunolabelled for Lama2 and CD31. Dotted bar graph shows the ratio of Lama2/CD31 M.F.I. in, normalized to the WT values (n=4 mice/genotype); Students' t-test: **p<0.02. Graph shows mean±SEM. Scale bars: A-D= 20 μm; E-H= 41 μm.

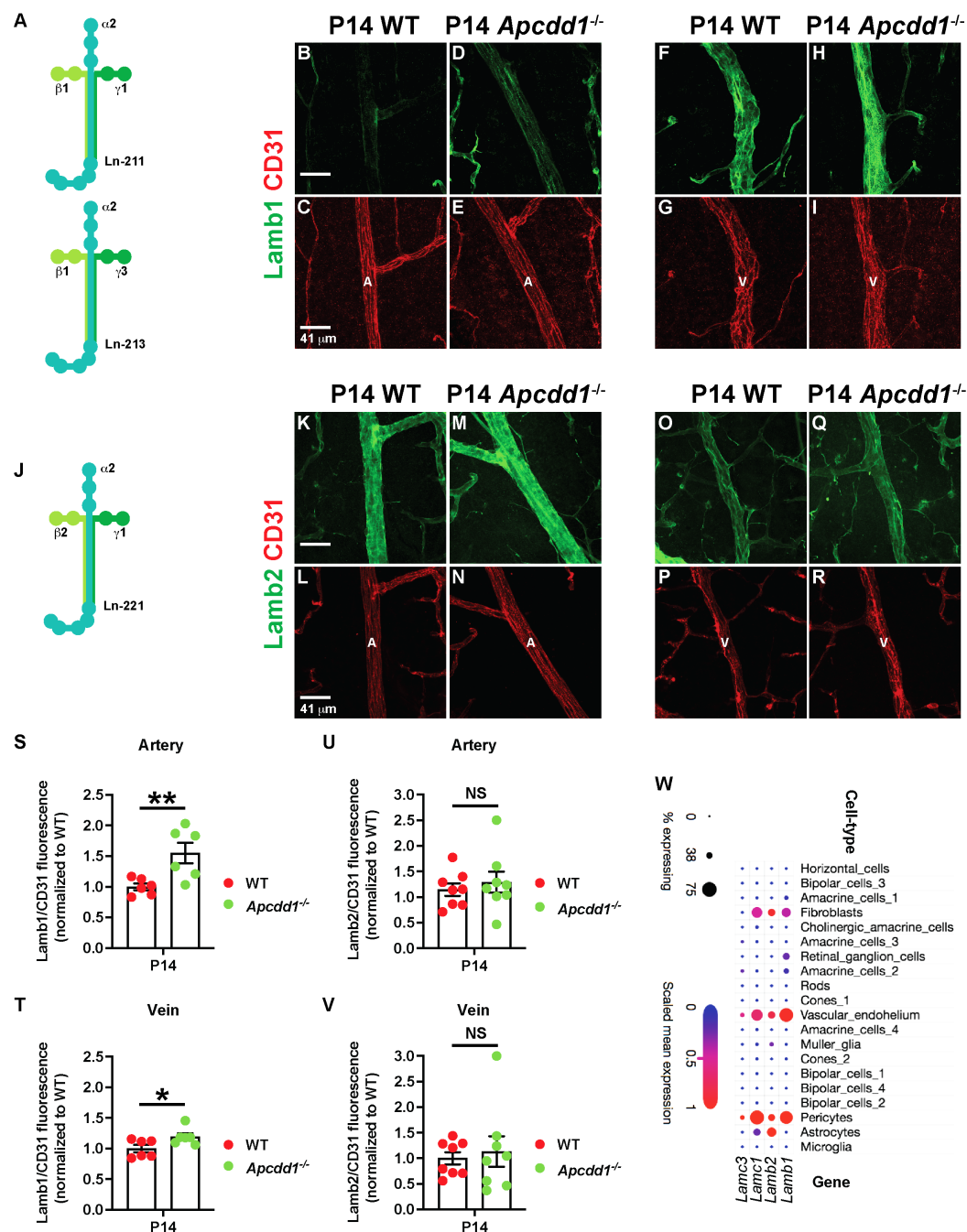


Fig. S5 (related to Fig. 3). Laminin-211 is likely the most affected isoform in the *Apcdd1*^{-/-} retina. **A)** Schematic diagram of two Laminin heterotrimers (211 and 213) containing $\alpha 2$ and $\beta 1$. **B-I)** P14 WT and *Apcdd1*^{-/-} retinal flat-mounts were labelled for Lamb1 and CD31. **J)** Schematic diagram of one $\alpha 2$ - and $\beta 2$ -

containing Laminin heterotrimer (221). **K-R**) P14 WT and *Apcdd1*^{-/-} retinal flat-mounts were labelled for Lamb2 and CD31. **S, T**) Dotted bar graphs show the ratio of Lamb1/CD31 M.F.I in arteries and veins at P14 normalized to the WT average values (6 arteries or veins analyzed from n=3 mice/group). **U, V**) Dotted bar graphs show the ratio of Lamb2/CD31 M.F.I in arteries and veins at P14 normalized to the WT average values (8 arteries and veins analyzed from n=4 mice/group). **W**) Analyses of the published P14 WT retinal single cell RNAseq database (see Results and Materials and Methods sections) to explore the expression profiles of *Lamb1*, *Lamb2*, *Lamc1* and *Lamc3*. A= artery, V= vein. Student's t-test: *p<0.05, **p<0.02, NS= not significant. Graphs show mean +/- SEM. Scale bars: 41 μ m.

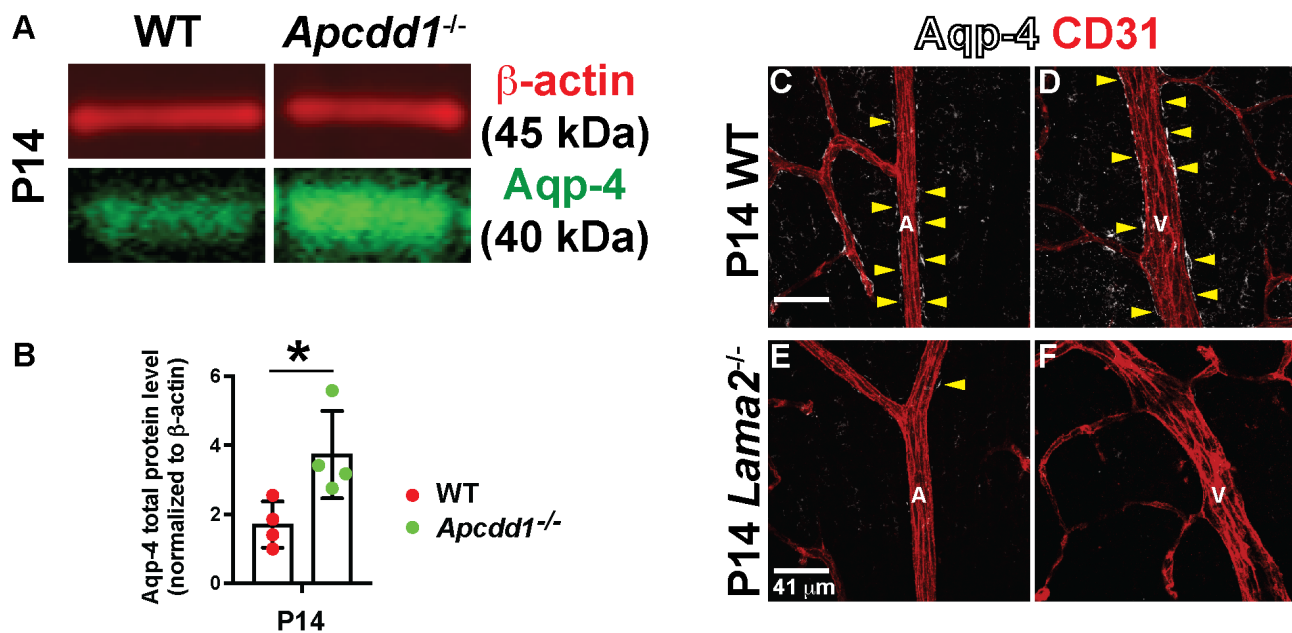


Fig. S6 (related to Fig. 6). Increased Aqp-4 in the *Apcdd1*^{-/-} retina and decreased Aqp-4⁺ astrocyte endfeet polarization in the *Lama2*^{-/-} retina. **A)** P14 WT and *Apcdd1*^{-/-} whole retinal lysates were probed for Aqp-4 (green bands) and β -actin (red bands) by western blot. **B)** Quantification of Lama1 and Lama2 protein levels (normalized to β -actin) by western blotting (n=4 samples/group). Student's t-test: *p<0.05. Graphs show mean \pm SD. **C-F)** P14 WT and *Lama2*^{-/-} retinal flat-mounts were labelled for Aqp-4 and CD31. Yellow arrowheads point at Aqp-4⁺ astrocyte endfeet around retinal blood vessels. A= arteries and V= veins. Scale bars: 41 μ m.

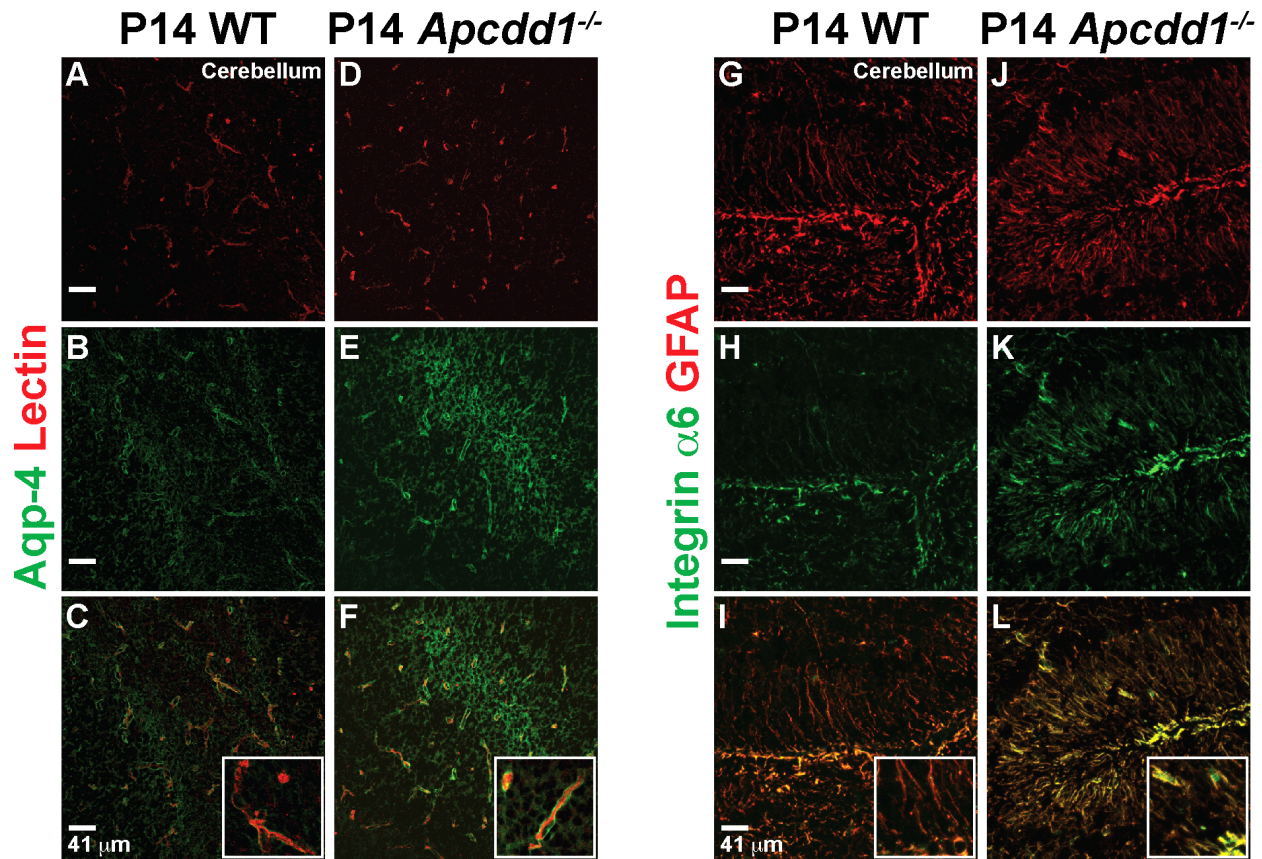


Fig. S7 (related to Figs. 6 and 8). Increased expression of Aqp4 in astrocyte endfeet and Integrin $\alpha 6$ in *Apcdd1*^{-/-} astrocytes in the cerebellum. A-F) P14 WT and *Apcdd1*^{-/-} cerebellar sections were labelled for Aqp-4 and Lectin. Insets show higher magnification images of single vessels. G-L) P14 WT and *Apcdd1*^{-/-} cerebellar sections were labelled for Integrin $\alpha 6$ and GFAP. Insets show higher magnification images. Scale bar: 41 μ m.

Table S1 (related to Figs. 2, 3, 6 and 8). List of significantly differentially expressed genes between wild-type and *Apcdd1*^{-/-} retinas at P10 and P14. Negative values mean that the gene is lower in *Apcdd1*^{-/-} compared to wild-type retinas, and positive values mean that the gene is higher in *Apcdd1*^{-/-} compared to wild-type retinas.

[Click here to download Table S1](#)

Table S2 (related to Fig. 2). List of significant differentially expressed putative endothelial genes between wild-type and *Apcdd1*^{-/-} retinas at P10 and P14. The genes are ranked by log₂Fold Change from the lowest to the highest value. Negative values mean that the gene is lower in *Apcdd1*^{-/-} compared to wild-type retinas, and positive values mean that the gene is higher in *Apcdd1*^{-/-} compared to wild-type retinas.

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Table S3 (related to Figs. 2 and 3). List of significant differentially expressed putative pericyte genes between wild-type and *Apcdd1*^{-/-} retinas at P10 and P14. The genes are listed by log₂Fold Change from the lowest to the highest value. Negative values mean that the gene is lower in *Apcdd1*^{-/-} compared to wild-type retinas, and positive values mean that the gene is higher in *Apcdd1*^{-/-} compared to wild-type retinas.

[Click here to download Table S3](#)

Table S4 (related to Figs. 2, 3 and 8). List of significant differentially expressed extracellular matrix genes (ECM) between wild-type and *Apcdd1*^{-/-} retinas at P10 and P14. The genes are listed by log₂Fold Change from the lowest to the highest value. Negative values mean that the gene is lower in *Apcdd1*^{-/-} compared to wild-type retinas, and positive values mean that the gene is higher in *Apcdd1*^{-/-} compared to wild-type retinas.

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Table S5 (related to Fig. 6). List of significant differentially expressed astrocyte maturity genes between wild-type and *Apcdd1*^{-/-} retinas at P10 and P14. The genes are listed by log₂Fold Change from the lowest to the highest value. Negative values mean that the gene is lower in *Apcdd1*^{-/-} compared to wild-type retinas, and positive values mean that the gene is higher in *Apcdd1*^{-/-} compared to wild-type retinas.

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