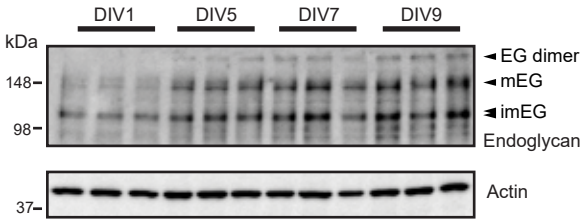
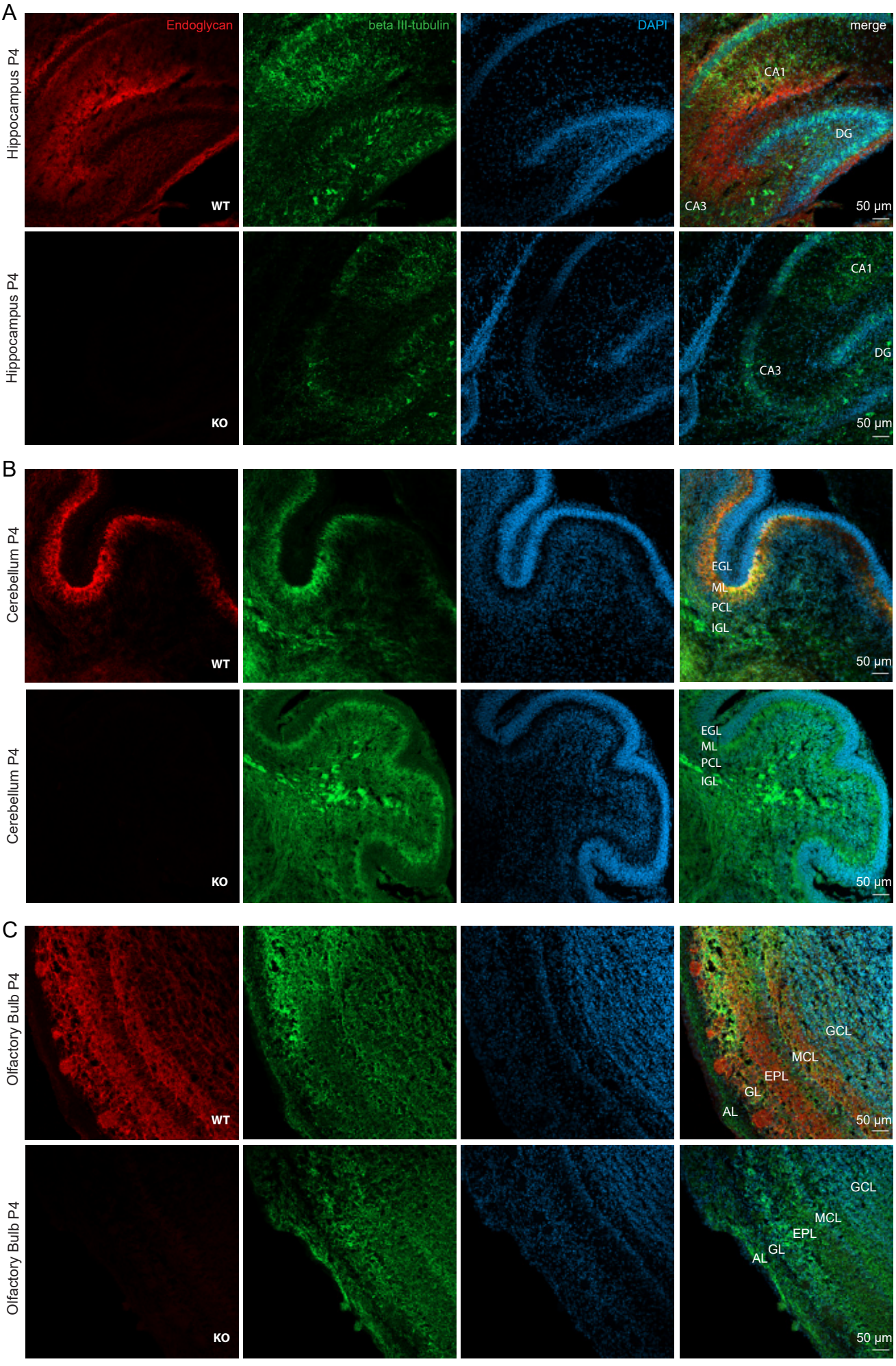
Supplementary Figures

Suppl. Figure S1



Supplementary Figure S1: Time-dependent expression of endoglycan in cultured neurons. Neurons were prepared from E16 mice, and collected at days in vitro (DIV)1, 5, 7, and 9. Actin was used as loading control for lysates. mEG: mature, fully glycosylated endoglycan; imEG: immature, partially glycosylated endoglycan.

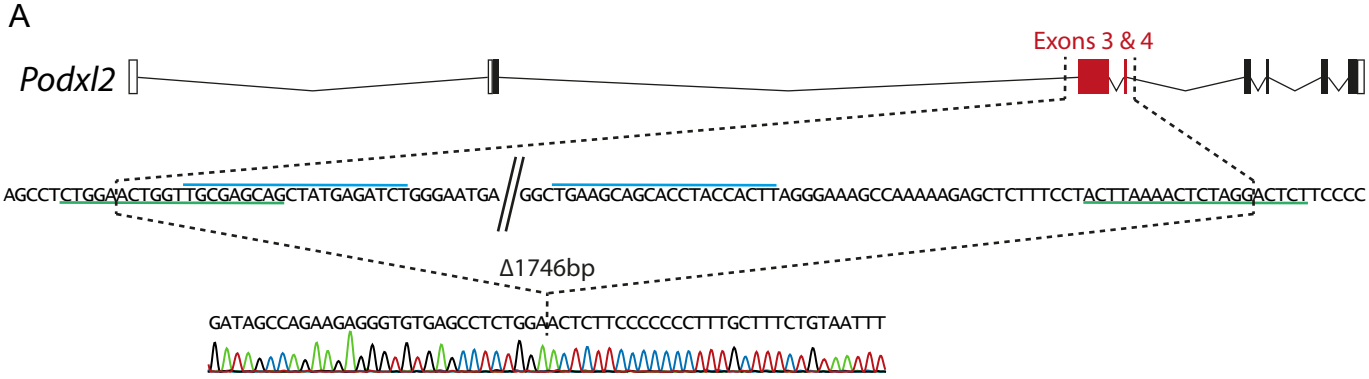
Suppl. Figure S2



Supplementary Figure S2: Immunohistochemistry of P4 mouse brains. (A-C) Immunostaining of endoglycan (red), beta III-tubulin (green), and DAPI (blue) of brain slices from 4 days old endoglycan WT (upper panels) and littermate KO mice (lower panels) within the hippocampal area (A), the cerebellar area (B), and the olfactory bulb (C). Confocal images are shown with a 10X magnification. Signals from endoglycan immunostaining were largely absent in the KO brain, indicating the specificity of the antibody in immunohistochemistry. Scale bars, 50 µm. *CA1*: cornu ammonis 1, *CA3*: cornu ammonis 3, *DG*: dentate gyrus, *EGL*: external granule layer, *ML*: molecular layer, *PCL*: Purkinje cell layer, *IGL*: internal granule layer, *AL*: axonal layer, *GL*: glomerular layer, *EPL*: external plexiform layer, *MCL*: mitral cell layer, *GCL*: granule cell layer

Suppl. Figure S3

A

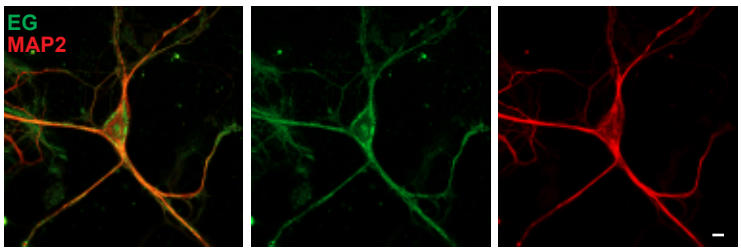


B

	WT	HET	KO	Total
Male	12	19	8	39
Female	2	11	1	14
Total	14	30	9	53

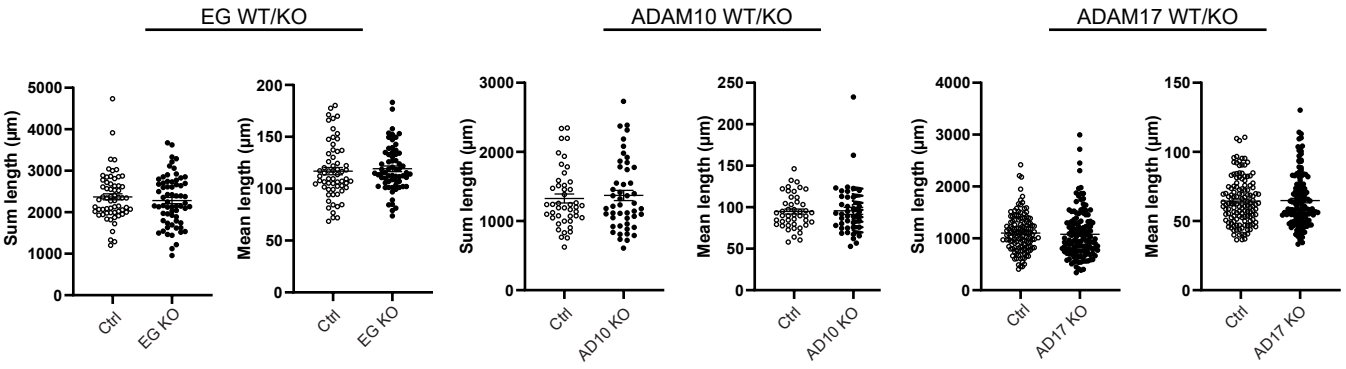
Supplementary Figure S3: Endoglycan knockout mouse. (A) Generation of the endoglycan (Podxl2) knock-out mouse line. Protospacer sequences targeting upstream of exon 3 and downstream of exon 4 (shown as red boxes) of the Podxl2 gene were indicated with blue (sense) and green (antisense) lines. KO allele was confirmed by sequencing and had a deletion of 1746 base pairs. (B) Resulting offspring genotypes from eight litters of endoglycan heterozygotes to heterozygotes (HET×HET) mating.

Suppl. Figure S4



Supplementary Figure S4: Co-localization of MAP2 and endoglycan in primary neurons. Immunostaining of endoglycan and MAP2 on mouse primary neurons at DIV7. Scale bars, 5 µm

Suppl. Figure S5



Supplementary Figure S5 : Quantification of neurite length in neurons lacking endoglycan (EG), ADAM10 or ADAM17. Neuronal images captured for Sholl analysis in Fig. 4 were used for manual neurite tracing. Panels show the summarized length or the mean length of neurites for each knock-out (KO) versus control. N=45-168. Shown are individual measurements as well as mean and error bars representing standard error of the mean. Results were analyzed by unpaired t-test.