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Figure S5 Phenotypic analysis of ICK Nes CKO mice.  (A–D) ICK mRNA expression levels in the P4 control and ICK Nes CKO cerebellum (A), in the P4 control and ICK Nes CKO hippocampus (B), in the P4 control and ICK Nes CKO cerebral cortex (C), and in the P4 control and ICK Nes CKO whole brain (D) were analyzed by Q-PCR. Error bars show the SD.  (E) ICK protein expression in the P4 control and ICK Nes CKO brain was examined by Western blot analysis. α-tubulin was used as a loading control.  (F) Ccrk mRNA expression levels in the P4 control and ICK Nes CKO brain were analyzed by Q-PCR. There was no significant difference between the P4 control and ICK Nes CKO brain.  (G) Growth retardation observed in ICK Nes CKO mice (right) compared to control mice (left) at the age of one month.  (H, I) Nissl staining of sagittal sections from the P21 control (H) and ICK Nes CKO (I) brain. There was no obvious difference in the size of lateral ventricles between the control and ICK Nes CKO brain.  (J, K) Nissl staining of sagittal cerebellar sections from P4 control (J) and ICK Nes CKO (K) mice. Defects in cerebellar foliation are observed in ICK Nes CKO mice.  (L, M) The numbers of Ki67-positive cells (L) and PH3-positive cells (M) in control and ICK Nes CKO EGL were counted. Ki67-positive cells and PH3-positive cells significantly decreased in ICK Nes CKO mice.  (N) Gli1 expression was reduced in the ICK Nes CKO cerebellum.  (O–T) Immunohistochemical analysis of the P4 ICK Nes CKO hippocampal DG. Sagittal hippocampal sections from P4 control and ICK Nes CKO mice were immunostained with antibodies against Ki67 (green in O, P) and PH3 (green in Q, R). Cell proliferation was decreased in the ICK Nes CKO hippocampal DG. The numbers of Ki67-positive cells (S) and PH3-positive cells (T) in the P4 control and ICK Nes CKO hippocampal DG were counted.  (U) Gli1 expression in the hippocampus was measured by Q-PCR.  (V, W) Cilia in ependymal cells are normally formed in the ICK Nes CKO brain. Sagittal sections from the P4 control and ICK Nes CKO brain were immunostained with an anti-acetylated α-tubulin antibody.  (X–Y’) Ciliary localization of GPCR in the ICK Nes CKO cerebral cortex.
Sections from cerebral cortex were stained with ciliary GPCR, SSTR3 (red). The cilia were stained with an anti-AC3 antibody (green). SSTR3 (arrowheads) were localized in the cilia properly both in control and ICK Nes CKO mice. Nuclei were stained with DAPI (blue). Scale bars, 20 mm (G), 1mm (H, I), 500 µm (J, K), 200 µm (O–R) and 10 µm (V–Y'). Error bars show the SD. **p < 0.03, *p < 0.05, n.s., not significant. LV, lateral ventricle.