

## **Supplementary Figure Legends**

### **Suppl. Fig 1. Expression patterns of EGFP in the brain following rAAV1-EGFP mediated transduction.**

**A-H.** Representative images of anti-EGFP immunoreactivity shows localized transduction of the choroid plexus and occasional neurons in the cortex, hippocampus and cerebellum (B) compared to PBS injected control mice (A) following rAAV1 injection into brains of neonatal mice on day P2. Representative high magnification images from ventricle (C), hippocampus and dentate (D), cortex (E), cerebellum (F), midbrain (G) and thalamus (H) depicting anti-EGFP staining is shown. *Scale Bar*, 600 $\mu$ m (A, B), 150 $\mu$ m (C-H).

**I-L.** Representative images of anti-EGFP immunoreactivity in the hippocampal pyramidal layer as well as neuronal projections in the cortex and thalamus following stereotaxic injection of rAAV1-EGFP into the hippocampus of 4 month old TgCRND8 mice (K, L) compared to PBS injected age matched controls (I, J). *Scale Bar*, 500 $\mu$ m (I, K), 125 $\mu$ m (J, L).

### **Suppl. Fig 2. rAAV1-mIFN $\gamma$ expression following stereotaxic injection in the hippocampus of adult TgCRND8 mice results in reactive astrogliosis and microgliosis.**

**A-L.** rAAV1-mIFN $\gamma$  or rAAV1-EGFP was injected into the hippocampus of 4 month old TgCRND8 mice and brain sections analyzed after 6 weeks (4→5.5 month). Whole brain panels showing a coronal section in the immediate vicinity of rAAV1 injection site was stained with anti-Iba-1 (A-F) or anti-GFAP (G-L) antibodies. Whole brain sections (top panels) and higher magnification (lower panels) highlight the activated glial morphology in the hippocampus of mIFN $\gamma$  expressing mice compared to EGFP expressing control mice. *Scale Bar*, 600 $\mu$ m (A, B, G, H), 150 $\mu$ m (C, D, I, J) and 25 $\mu$ m (E, F, K, L). *n*=5/group.

**M.** Quantitation of Iba-1 and GFAP immunoreactivity in paraffin embedded sections of 4→5.5 month old TgCRND8 mice expressing mIFN $\gamma$  or EGFP. ( $n=3/\text{group}$ ,  $*p<0.05$ ).

**N.** Levels of mIFN $\gamma$  in mIFN $\gamma$  expressing 4→5.5 month old TgCRND8 mice brains were increased compared to age-matched controls. Protein levels were analyzed using RIPA buffer solubilized brain lysates (dissected 1 mm anterior and 1 mm posterior to the injection site) by sandwich ELISA. ( $n=5/\text{group}$ ;  $*p<0.05$ ).

**O-P.** Representative immunoblot showing increased levels of CD11b and GFAP in 4→5.5 month old mIFN $\gamma$  expressing TgCRND8 mice compared to controls.  $\beta$ -actin has been used as a loading control (O). Intensity analysis of CD11b and GFAP immunoreactive bands normalized to  $\beta$  actin in 4→5.5 month old TgCRND8 mice is depicted (P). ( $n=3/\text{group}$ ,  $*p<0.05$ ).

**Suppl. Fig 3. Characterization of pathological features following chronic mIFN $\gamma$  expression in P2→5 month old TgCRND8 mice.**

**A-D.** Representative Nissl stained sections of rAAV1-mIFN $\gamma$  injected P2→5 month old TgCRND8 mice (A-B) and rAAV1-mIFN $\gamma$  injected 4→5.5 month old TgCRND8 mice (C-D). Lower panels (B, D) show higher magnification of thalamus (boxed area). P2→5 month old mIFN $\gamma$  expressing mice show basal ganglial calcification (arrowheads) whereas 4→5.5 month old rAAV1-mIFN $\gamma$  expressing TgCRND8 mice do not show any calcification. *Scale Bar*, 600 $\mu\text{m}$  (A, C), 100 $\mu\text{m}$  (B, D).  $n=5/\text{group}$ .

**E.** Representative von Kossa stained brain sections of a P2→5 month old mIFN $\gamma$  expressing TgCRND8 mouse showing no calcification in the hippocampus (inset 1), but extensive calcification in the thalamus (inset 2). *Scale Bar*, 600 $\mu\text{m}$  (E, left panel) and 150 $\mu\text{m}$  (insets 1 and 2).  $n=5/\text{group}$ .

**F-K.** Representative images of anti-CD3 immunostaining in brains of mIFN $\gamma$  expressing P2→5 month old TgCRND8 mice (G, J) or EGFP expressing control mice (F, I) shows no evidence of T cell infiltration. mTNF $\alpha$  expressing 5 month old TgCRND8 mice showing CD3 immunoreactive T cells (H, K) was used as positive control. *Scale Bar*, 600  $\mu$ m (F,G,H), 75 $\mu$ m (I,J,K).  $n=5$ /group.

**L-N.** Representative images of Nissl stained sections of hippocampal pyramidal cells in mIFN $\gamma$  expressing P2→5 month old TgCRND8 mice (M) compared to controls (L). Quantification of cell count and cell diameter of Nissl stained hippocampal pyramidal cells in P2→5 month old TgCRND8 mice expressing mIFN $\gamma$  compared to controls is depicted (N). ( $n=5$ /group,  $*p<0.05$ ). *Scale Bar*, 125 $\mu$ m.

**Suppl. Fig 4. Amyloid deposition is attenuated following acute focal expression of mIFN $\gamma$  in the hippocampus of TgCRND8 mice.**

**A-F.** 4 month old TgCRND8 mice were stereotaxically injected in the hippocampus with either rAAV1-mIFN $\gamma$  or rAAV1-EGFP and sacrificed after 6 weeks. Representative brain sections from EGFP and mIFN $\gamma$  expressing 5.5 month old mice as well as unmanipulated 4 month old mice were stained with anti A $\beta$ 42 antibody (A-C) and anti A $\beta$ 40 antibody (D-F). Insets depict individual plaques. *Scale Bar*, 600 $\mu$ m (A-F); inset, 25 $\mu$ m.  $n=5$ /group.

**G.** Forebrain A $\beta$  plaque burden analysis depicting alterations in A $\beta$ 42 and A $\beta$ 40 burden in 5.5 month old mIFN $\gamma$  and EGFP expressing mice compared to unmanipulated 4 month old TgCRND8 mice. ( $n=5$ /group,  $*p<0.05$  and  $**p<0.05$ ).

**Suppl. Fig 5. No change in endogenous APP or steady state A $\beta$  levels in mIFN $\gamma$  expressing APP non-transgenic mice or 3 month old transgenic Tg2576 mice.**

**A-B.** Representative anti CT20 immunoblot showing APP levels in P2→5 month old CRND8 nontransgenic mice expressing mIFN $\gamma$  and age-matched controls (A). Intensity analysis of APP bands normalized to  $\beta$ -actin show no significant changes in APP levels in P2→5mo nontransgenic mice injected with rAAV1-mIFN $\gamma$  compared to age-matched controls (B).  $n=4$ /group.

**C.** No change in soluble endogenous A $\beta$ 40 levels was seen in mIFN $\gamma$  expressing P2→5 month old CRND8 non-transgenic mice compared to age-matched controls.  $n=4$ /group.

**D-E.** Representative anti CT20 immunoblot showing APP and CTF levels in P0→3 month old Tg2576 mice expressing mIFN $\gamma$  and age-matched controls (D). Quantitative analysis of APP and CTF levels normalized to  $\beta$ -actin show no significant changes in P0→3 month old Tg2576 mice expressing mIFN $\gamma$  compared to age-matched controls (E).  $n=3$ /group.

**F.** Steady state levels of A $\beta$ 40 is not significantly altered in P0→3 mo mIFN $\gamma$  expressing Tg2576 mice as measured by ELISA using RIPA soluble and SDS soluble brain extracts.  $n=3$ /group.

**Suppl. Fig 6. No change in A $\beta$  degrading enzymes and BACE protein levels in mIFN $\gamma$  expressing P2→5 month old TgCRN8 mice.**

**A.** Q-PCR analysis of Neprilysin and IDE levels in brains of 5 month old TgCRND8 mice expressing EGFP or mIFN $\gamma$  show no significant changes in mRNA transcript levels. Relative quantitation of mRNA expression was performed using the comparative cycle threshold method. The expression levels of genes were normalized using  $\beta$ -actin levels from the corresponding samples. Data represents averaged fold change values obtained from IFN $\gamma$  expressing mice relative to averaged values obtained from EGFP expressing mice. ( $n=4$ /group).

**B-C.** Representative immunoblot of BACE1 immunoreactivity in P2→5 month TgCRND8 mice expressing mIFN $\gamma$  or EGFP (B). Intensity analysis of BACE1 bands normalized to  $\beta$ -actin show no significant changes in BACE1 levels in TgCRND8 mice expressing mIFN $\gamma$  mice compared to control mice (C).  $n=3$ /group.

**Suppl. Fig 7. mIFN $\gamma$  treatment of primary mouse microglia cells results in increased CD68 expression and efficacious A $\beta$  phagocytosis *in vitro***

**A-C.** Representative image of anti-CD68 immunostaining showing upregulation of CD68 (red fluorescence) in mIFN $\gamma$  treated primary mouse microglia cells (B) compared to untreated glia (A). Blue fluorescence depicts the DAPI stained glial nuclei. Quantitation of fluorescence intensity was done using Metamorph software (C). At least four different fields of view with ten or more cells per field were used for the analysis. Magnification 400X, \* $p<0.05$ .

**D-F.** mIFN $\gamma$  treatment of primary mouse microglia results in more efficient phagocytosis of fA $\beta$ 42-Hilyte568 (red fluorescence, E) compared to unstimulated glia (D). Blue fluorescence depicts the DAPI stained glial nuclei. Quantitation of fluorescence intensity was done using Metamorph software (F). At least ten different fields of view with two or more cells per field were used for the analysis. Magnification, 600X, \* $p<0.05$ .

**Suppl. Fig 8. No evidence of colocalization of peripheral macrophages with cored plaques in 5 month old mIFN $\gamma$  expressing mice.**

Representative sections from the brains of P2→5 month old EGFP (Control) or mIFN $\gamma$  expressing mice and spleen from 5 month old wild type BL6 mice depicting CD45 (A-C) and Ly-6C (D-F) immunoreactivity. The sections have been counterstained with hematoxylin and Congo Red. Insets show high magnification views of splenic macrophages (A, D) or Congo Red stained A $\beta$  plaques (B-C, E-F). *Scale Bar*, 120 $\mu$ m; inset, 15 $\mu$ m.  $n=5$ /group.