Supplemental material

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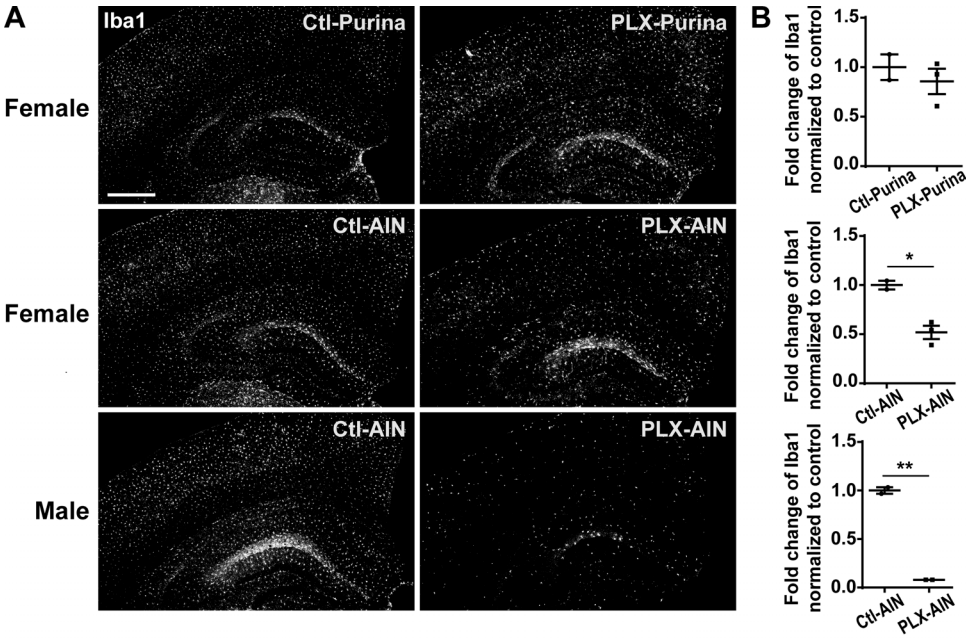

C

Female mice	Purina	AIN	Purina/AIN (%)
Drug incorporation rate in diet (%)	70	90	78
Average drug uptake per day μg (drug) / g (mouse weight)	23.7	29.6	80
Average plasma drug concentration (μg/ml)	134.3	299.7	45
Microglial depletion rate (%)	14	48	30

D

AIN chow	Female	Male	Female/Male (%)
Average drug uptake per day μg (drug) / g (mouse weight)	29.6	32	93
Average plasma drug concentration (μg/ml)	299.7	580	52
Microglial depletion rate (%)	48	92	52

Figure S1. **Mouse sex and chow formula impact the efficiency of microglial depletion by PLX3397. (A)** Iba1 staining images for 3–4-mo-old female or male TE4 mice treated with 300 mg/kg PLX3397 supplemented in either Purina-5053 or AIN-76A diet for 7 d (n = 2–3 mice per group). Scale bar = 500 μm for all images. **(B)** Quantification of Iba1-covered area in the hippocampus. **(C and D)** Effects of chow formula and sex on serum PLX3397 levels in relation to microglial depletion rate. Data are expressed as means ± SEM. One-way ANOVA with Tukey's post hoc test. *, P < 0.05; **, P < 0.01. The experiments in A and B were repeated three times.

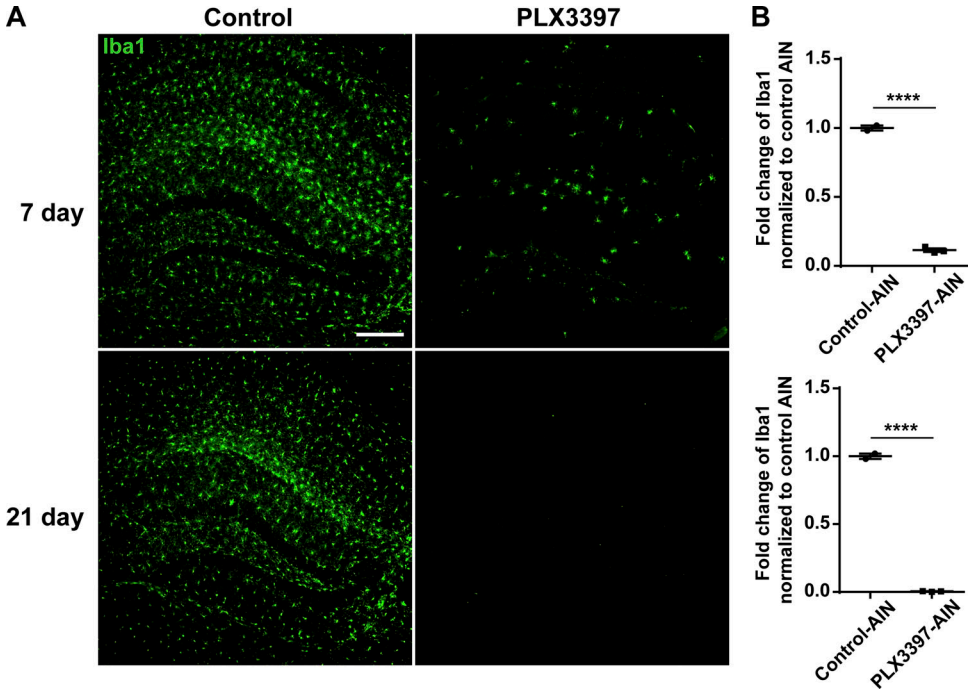

Figure S2. **PLX3397 supplemented in the AIN-76A diet at 400 mg/kg chow depletes microglia with high efficiency. (A)** Iba1 staining images for 5–6-mo-old male E4 mice treated with 400 mg/kg PLX3397 supplemented in the AIN-76A diet for 7 or 21 d (n = 2–3 mice per group). Scale bar = 200 µm for all images. **(B)** Quantification of Iba1-covered area in the hippocampus. Data are expressed as means ± SEM. One-way ANOVA with Tukey's post hoc test. ****, P < 0.0001. This characterization experiment was performed once.

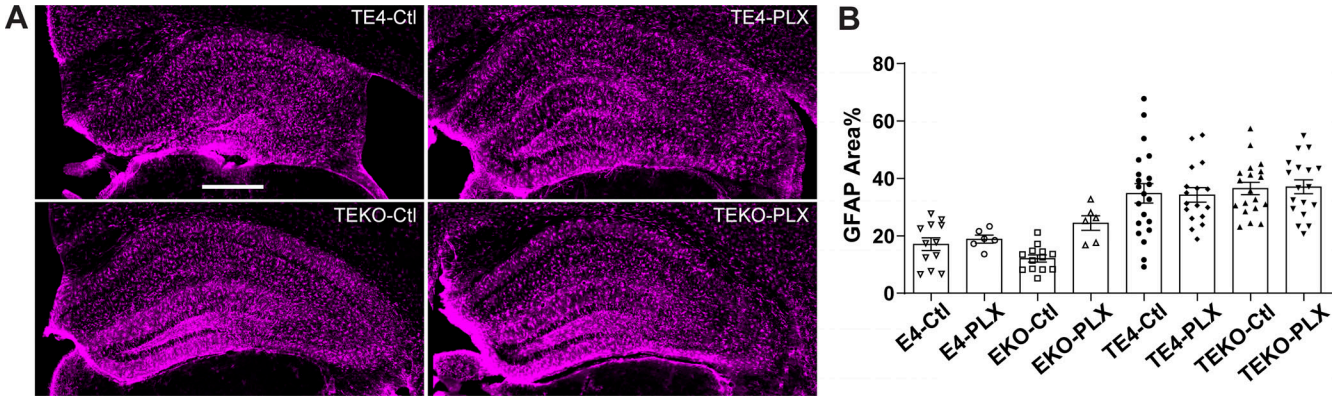

Figure S3. **PLX3397 treatment does not affect GFAP⁺ astrocytes. (A)** Representative images of GFAP staining for 9.5-mo-old TE4 and TEKO mice treated with control (Ctx) or PLX3397-supplemented (PLX) chow. Scale bar = 500 µm for all images. **(B)** Quantification of GFAP-covered area in the hippocampus of all mice (TE4-Ctl: n = 21; TE4-PLX: n = 17; TEKO-Ctl: n = 19; TEKO-PLX: n = 18; E4-Ctl: n = 12; E4-PLX: n = 6; EKO-Ctl: n = 13; EKO-PLX: n = 6). Data are expressed as means ± SEM. One-way ANOVA with Tukey's post hoc test. The GFAP staining was performed once.

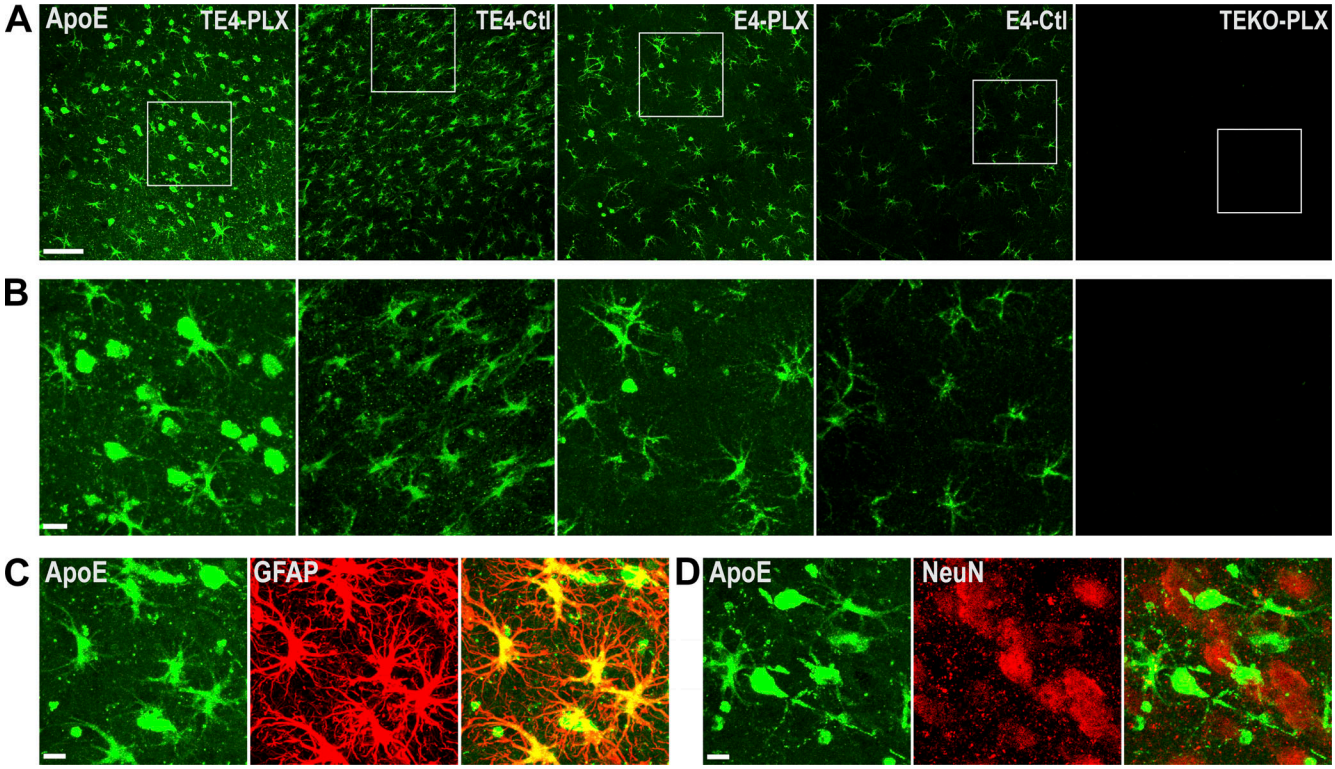

Figure S4. **ApoE is elevated in astrocytes and neurons in the piriform/entorhinal cortex of PLX3397-treated TE4 and E4 mice. (A)** Representative images of apoE staining in the piriform/entorhinal cortex of 9.5-mo-old TE4-PLX, TE4-Ctl, E4-PLX, E4-Ctl, and TEKO-PLX mice, respectively. Scale bar = 50 µm for all images. **(B)** Zoom-in of the selected area in A. Scale bar = 10 µm for all images. **(C)** Co-localization of apoE with GFAP$^+$ astrocytes in the piriform/entorhinal cortex of TE4-PLX mouse. Scale bar = 10 µm. **(D)** Non-co-localization between apoE and NeuN in the piriform/entorhinal cortex of TE4-PLX mouse. Scale bar = 10 µm. ApoE staining was repeated seven times using different antibodies.

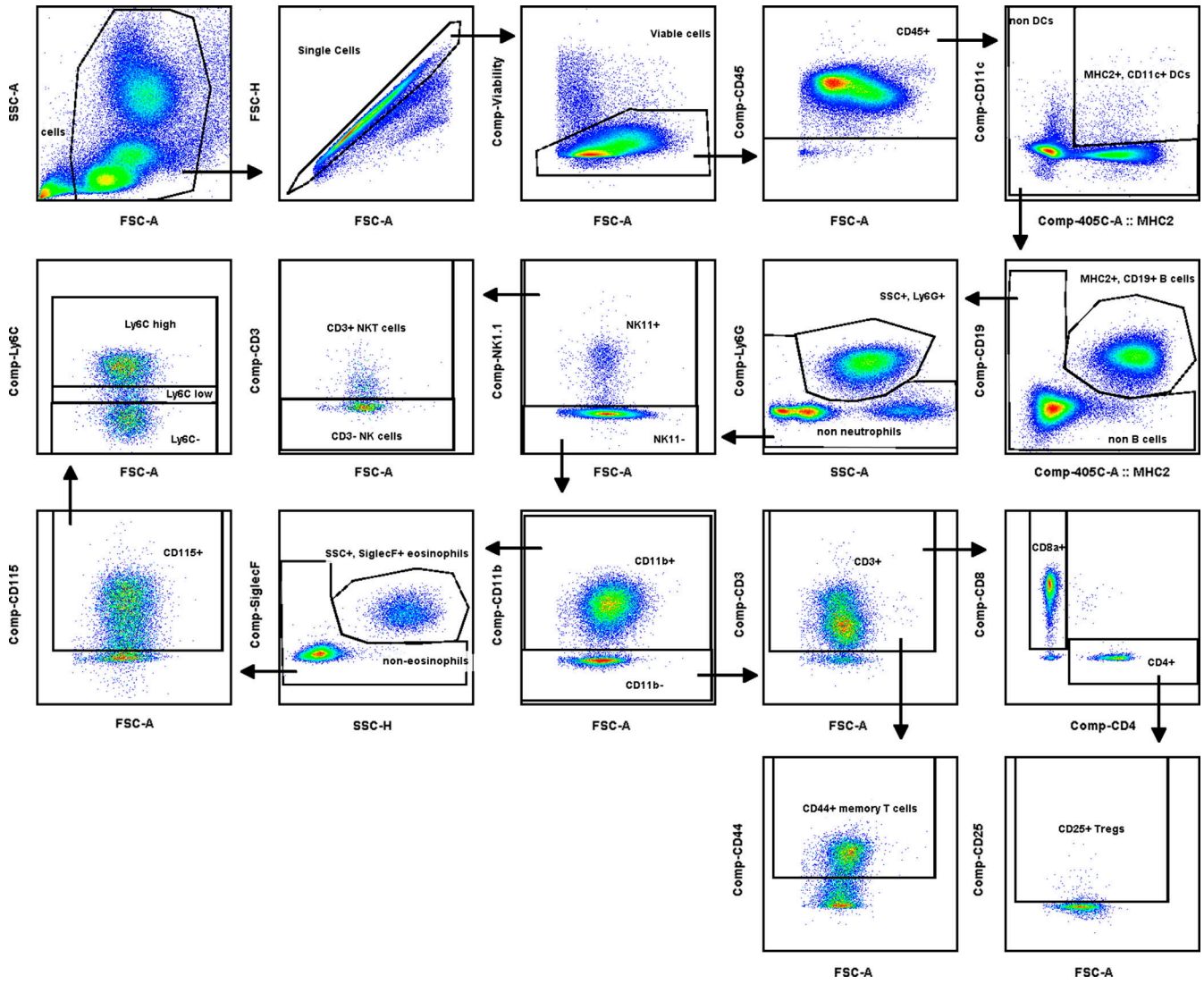

Figure S5. **Gating strategies for isolating blood cell types using flow cytometry.** Blood cells were sequentially gated through total cells, single cells, viable cells, and CD45$^+$ cells. From the CD45$^+$ gate, DCs were isolated as the MHC2$^+$CD11c$^+$ population. From the non-DC gate, B cells were isolated as the MHC2$^+$CD19$^+$ population. From the non-B cell gate, neutrophils were isolated as the SSC^{high}Ly6G$^+$ population. From the nonneutrophil gate, NK1.1$^+$ cells were isolated and further divided into CD3$^+$ NK T cells and CD3$^-$ NK cells. The NK1.1$^-$ gate was further separated into CD11b$^+$ and CD11b$^-$ gates. From the CD11b$^+$ gate, eosinophils were isolated as the SSC^{high}SiglecF$^+$ population. From the noneosinophil gate, monocytes were isolated as the CD115$^+$ population, which was further divided into Ly6C^{hi}, Ly6C^{lo}, and Ly6C$^-$ populations. From the CD11b$^-$ gate, T cells were isolated as the CD3$^+$ population, which was further separated into CD4$^+$ and CD8$^+$ T cells, or CD44$^+$ memory T cells. From the CD4$^+$ T cell gate, T reg cells were isolated as the CD25$^+$ population. SSC-A, side scatter area; FSC-A, forward scatter area; SSC-H, side scatter height.