Expanded View Figures

Figure EV1. Mononuclear phagocyte-resident NRP1 expression in LysM-Cre/*Nrp1*^{fl/fl}.

A   Gating scheme explaining the identification of the Ly6G$^-$, F4/80$^+$, CD11b$^+$, and NRP1$^+$ mononuclear phagocytes in retinas and sclera-choroid-RPE cell complexes. 1. Gating of live cells, 2. Removal of doublets, 3. Selection of viable cells, 4. Exclusion of neutrophils, 5. Gating of mononuclear phagocytes, 6. Gating of NRP1$^+$ mononuclear macrophages.

B   FACS histogram of APC-conjugated rat IgG2A isotype control (red) versus anti-mNRP1 APC-conjugated rat IgG2A (R&D systems) (blue).

C   FACS histogram of FMO (fluorescence minus one) versus anti-mNRP1 APC-conjugated rat IgG2A.

D, E   Quantification of NRP1-positive microglia (Ly6G$^-$, F4/80$^+$, CD11b$^+$, CX3CR1^{hi}, CD45^{lo}, NRP1$^+$) in retinas and sclera-choroid-RPE cell complexes in Naïve (non-burned) mice (D); $n = 4$ and at D3 (E); $n = 4$.

F, G   Quantification of mononuclear phagocytes (Ly6G$^-$, F4/80$^+$, CD11b$^+$) in retinas and sclera-choroid-RPE cell complexes in Naïve (non-burned) mice (F); $n = 5$ (LysM-Cre/*Nrp1*$^{+/+}$), 4 (LysM-Cre/*Nrp1*$^{fl/fl}$) and at D3 (G); $n = 4$.

H   Compilation of representative compressed Z-stack confocal images of FITC–dextran-labeled CNV and IB4-stained laser impact area from LysM-Cre/*Nrp1*$^{+/+}$ and LysM-Cre/*Nrp1*$^{fl/fl}$ mice at D7. Scale bar: 20 μm.

I–K   Quantification of area of FITC–dextran-labeled CNV (I), isolectin B4 (IB4)-stained laser impact area (J) and the ratio of FITC/IB4 per laser burn (K) relative to LysM-Cre/*Nrp1*$^{+/+}$ at D7; $n = 14$ burns (LysM-Cre/*Nrp1*$^{+/+}$), $n = 20$ burns (LysM-Cre/*Nrp1*$^{fl/fl}$).

Data information: All comparisons between groups were analyzed using a Student's unpaired *t*-test; **$P < 0.01$, ***$P < 0.001$; error bars represent mean $\pm$ SEM; exact P values listed in Table 2.
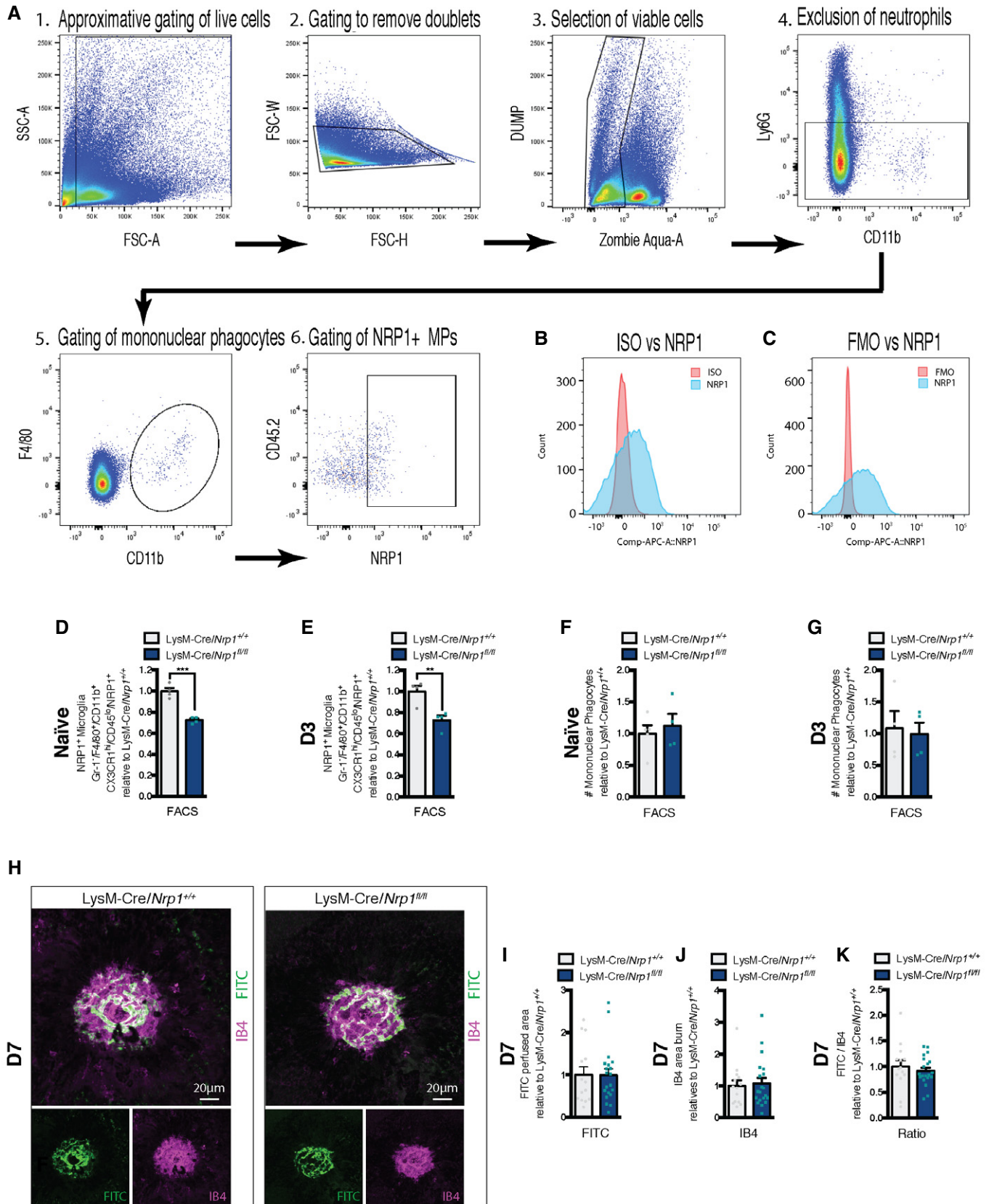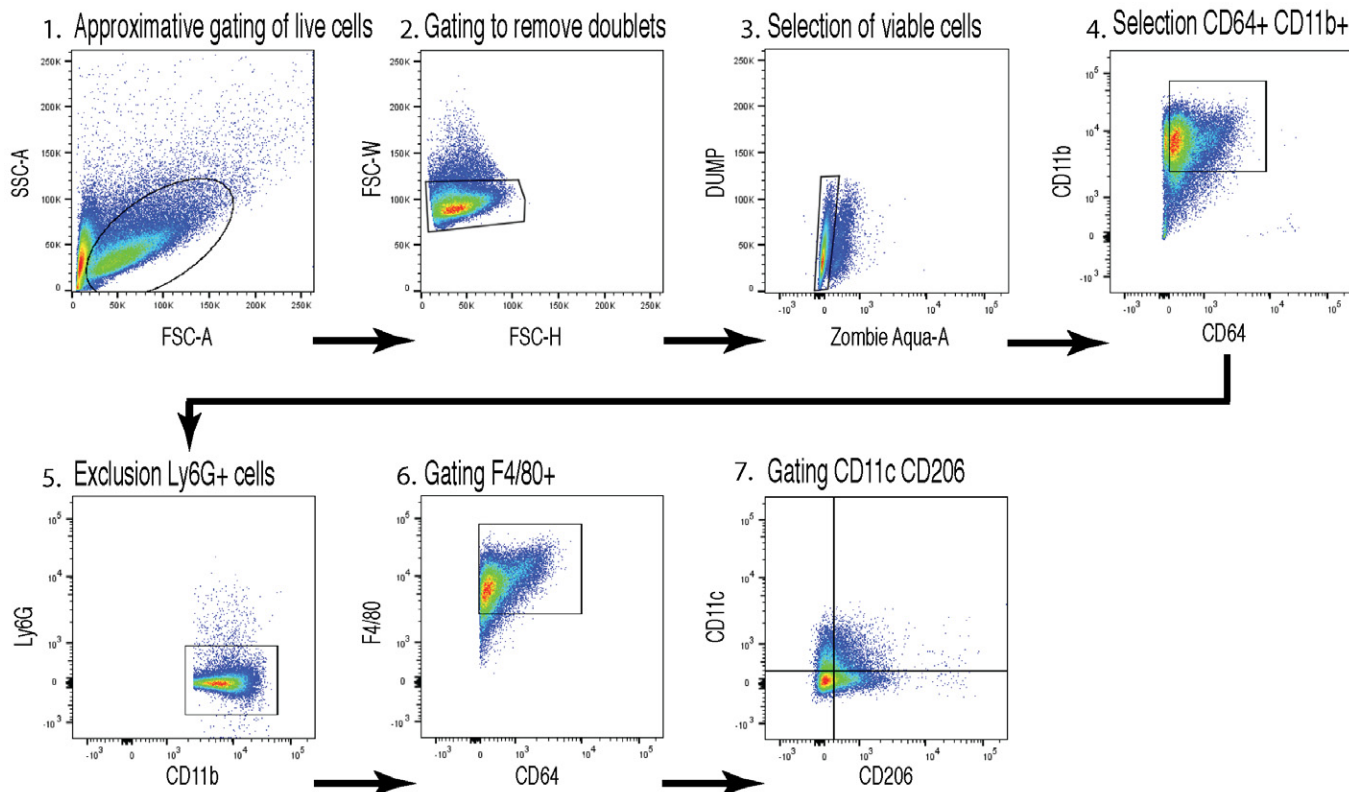
Figure EV1.

Figure EV2. Gating scheme explaining the identification of the Ly6G⁻, F4/80⁺, CD11b⁺, CD11c⁺, CD206⁻ and Ly6G⁻, F4/80⁺, CD11b⁺, CD11c⁻, CD206⁺ BMDMs.

1. gating of live cells, 2. removal of doublets, 3. selection of viable cells, 4. Selection of myeloid cells (CD64+) 5. exclusion of neutrophils, 6. gating of mononuclear macrophages, 7. Gating on CD11c and CD206.
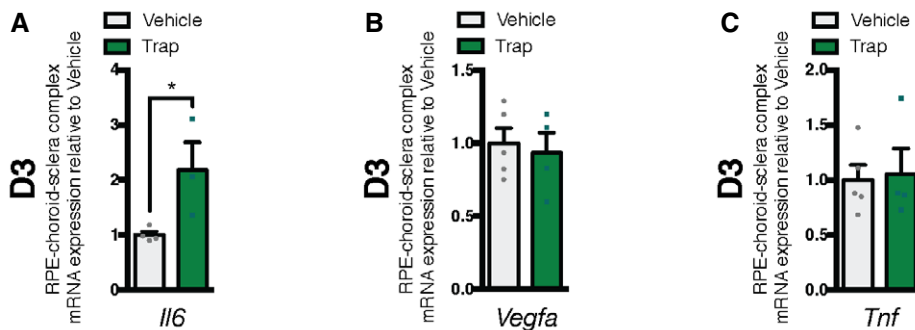

Figure EV3. Inflammatory state of RPE-choroid-sclera complexes following treatment with trap.

A–C mRNA expression of inflammation markers relative to vehicle in mouse RPE-choroid-sclera complexes at D3 for *Il6* (A); *n* = 4 (vehicle), *n* = 4 (trap), *Vegfa* (B); *n* = 5 (vehicle), *n* = 4 (trap), *Tnf* (C); *n* = 5 (vehicle), *n* = 4 (trap).

Data information: All comparisons between groups were analyzed using a Student's unpaired *t*-test; *P < 0.05; error bars represent mean ± SEM; exact *P* values listed in Table 2.