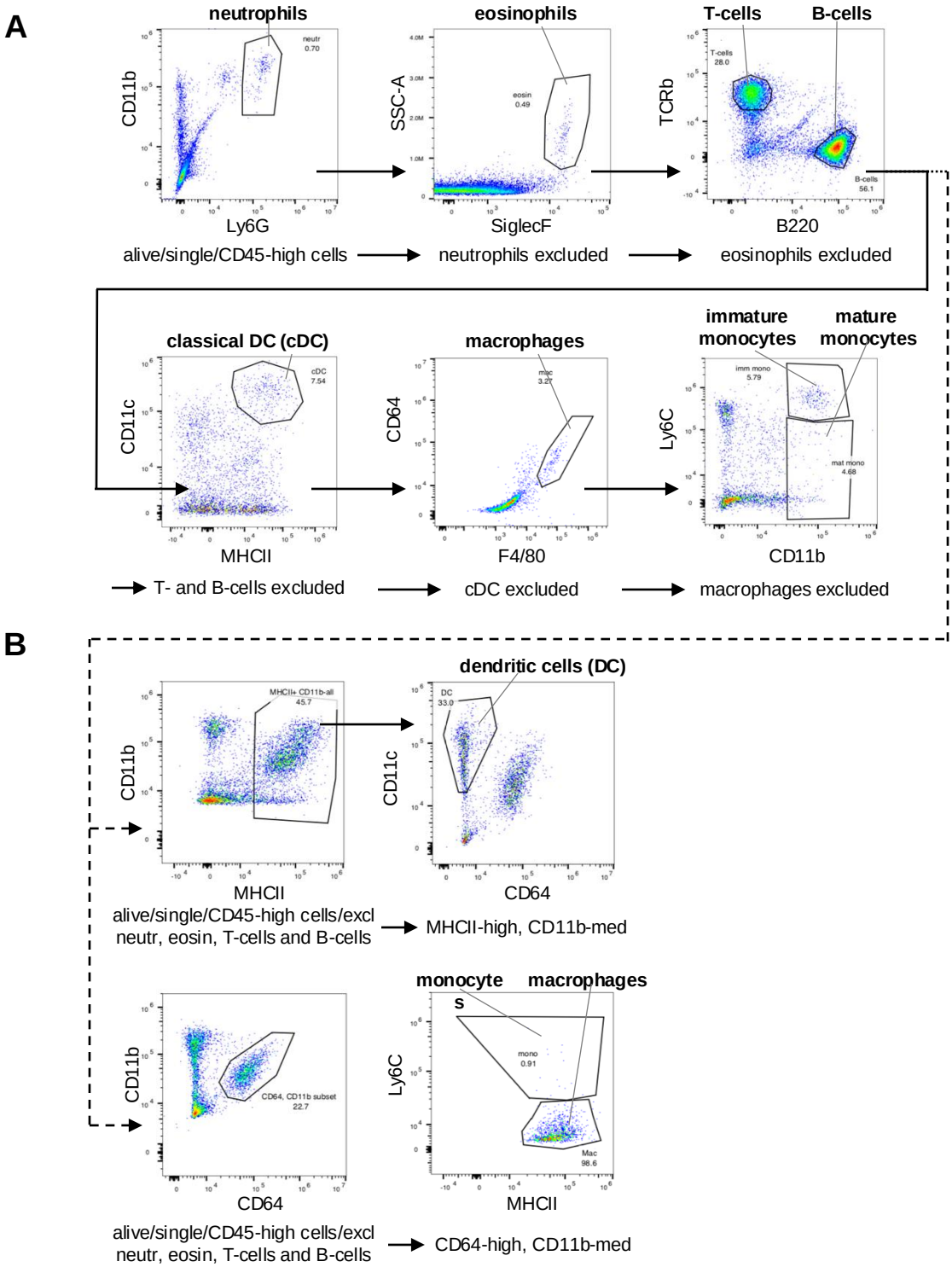
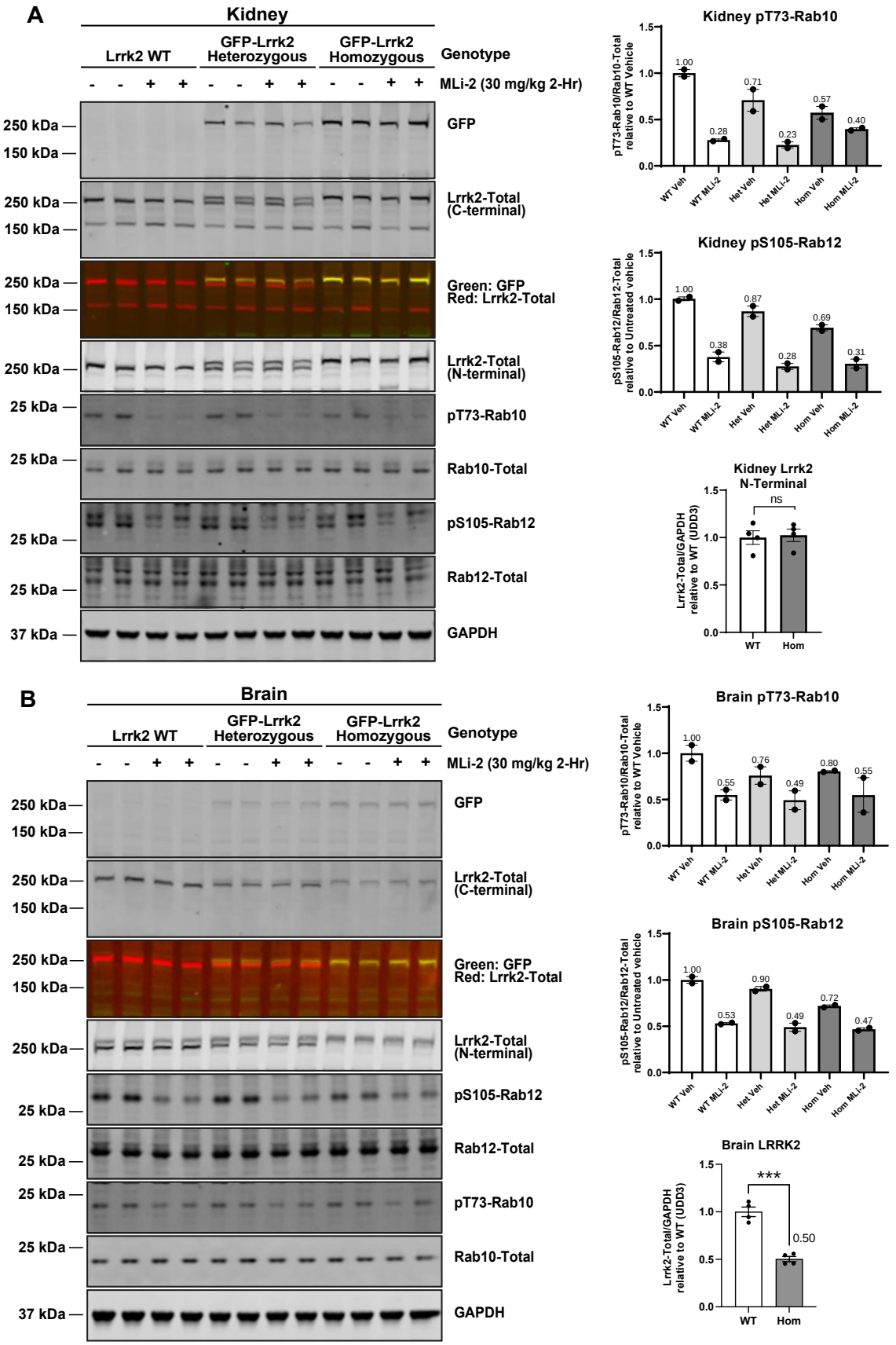
Dikovskaya et al. Inflammation and IL-4 regulate Parkinson's and Crohn's disease associated kinase LRRK2.

Appendix

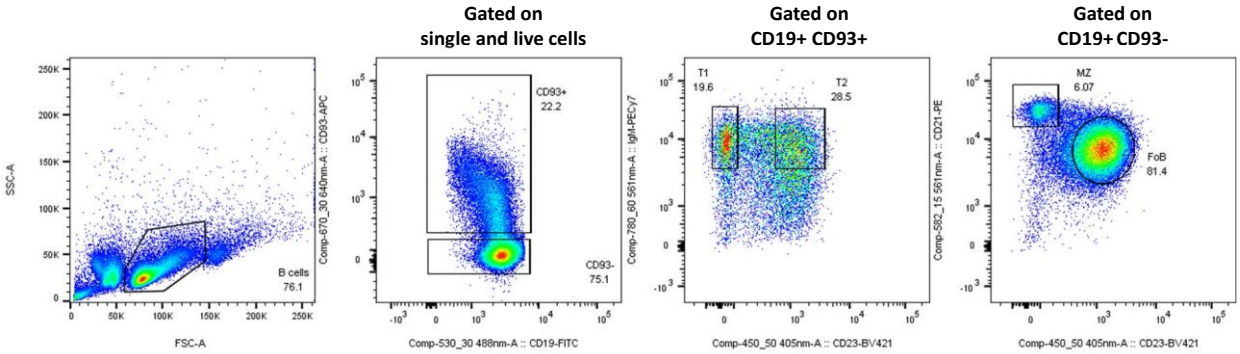
Table of context:



Appendix Figure S1. Gating strategy for splenocytes and lamina propria cells. **A.** Splenocytes gating. Immune cells were gated from live, single CD45+ cells followed by sequential identification and exclusion of: neutrophils ($CD11b^{+}/Ly6G^{hi}$); eosinophils ($SiglecF^{+}$/ SSC-high); T- cells (TCRb+) and B-cells (B220-high); classical dendritic cells (cDC, $CD11b^{hi}/MHCII^{hi}$); macrophages (CD64+/F4/80+) and immature and mature monocytes ($CD11b^{hi}/Ly6C^{hi}$ and $CD11b^{hi}/Ly6C^{lo}$ respectively). **B.** Lamina propria cells gating. The staining and initial gating was identical to A up to the exclusion of T - and B-cells, after which DCs were defined as $CD11b^{+/-}/MHCII^{hi}$ then gating on $CD11c^{hi}/CD64^{-}$ cells. Macrophages and monocytes were identified as CD64+/ $CD11^{int}$ cells, then $Ly6C^{+}$ for monocytes and for $Ly6C^{-}/MHCII^{+}$ for macrophages.



Appendix Figure S2. Analysis of Lrrk2 pathway in kidneys (A) and brain (B) of EGFP-LRRK2-KI mouse. 3-month-old WT, Heterozygous and Homozygous EGFP-LRRK2-KI mice were treated with or without 30 mg/kg MLi-2 for 2h prior to tissue collection and processed as in Fig. EV4. Quantification of Lrrk2-substrate phosphorylation of pT73-Rab10 and pS105-Rab12 relative to total levels (n = 2), and total Lrrk2 levels relative to GAPDH for both N-terminal antibodies (n = 4) are shown as mean ± SEM for each tissue. Statistical significance calculated by unpaired 2-tailed T-test, with the values *** - p = 0.0003 or "ns" - nonsignificant. Each lane indicated sample derived from a different mouse tissue.



Appendix Figure S3. Gating strategy for mouse splenic B-cell subtypes. Splenic B-cells were isolated using pan B-cell isolation kit by negative selection and stained with DAPI and antibodies for surface markers. Live single CD19^{+} B-cells were digitally separated into CD93^{hi}/CD23^{-} T1 transitional B-cells and CD93^{hi}/CD23^{hi} T2 transitional B-cells, and CD93^{-} mature B-cells were further subdivided CD23^{hi}/CD21^{+} follicular B-cells (Fo) and CD23^{-}/CD21^{hi} marginal zone B-cells (MZ).