Supplemental Information

A novel role for IL-6 receptor classic signaling: induction of RORyt+Foxp3+ Tregs with enhanced suppressive capacity

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Supplementary Figure S1. IL-6Ra expression on naive lymphocytes

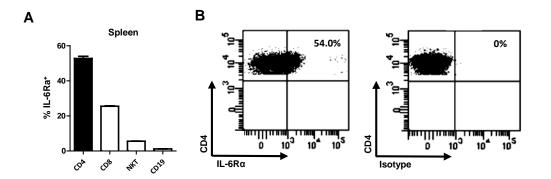
Supplementary Figure S2. CD4^{Cre} mediated excision of IL-6Ra on different splenic leukocyte subsets

Supplementary Figure S3. CD4^{Cre} mediated excision of IL-6Ra on different renal leukocyte subsets

Supplementary Figure S4. Renal T cell responses in recipients of IL-6Ra^{-/-} whole CD4⁺ T cells after NTN induction

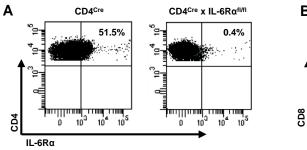
Supplementary Figure S5. Systemic Treg percentages remain unaffected by IL-6Ra deficiency and in IL-6^{-/-} mice

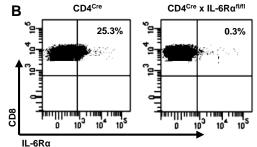
Supplementary Figure S6. Th17 responses are unchanged in nephritic mice after transfer of IL-6Ra^{-/-} Tregs



Supplementary Figure S1. (A) FACS analysis of IL-6Ra expression on indicated lymphocyte subsets from spleens of naive mice. (B) Representative FACS plot of IL-6Ra expression (left) and isotype control (right) on naive splenic CD4⁺ T cells.

T cell subpopulations

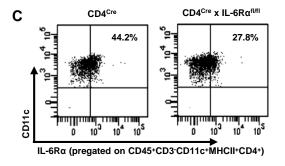


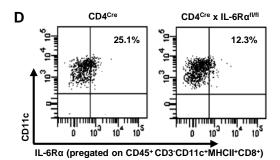


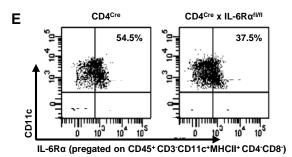
Supplementary Figure S2. (A-H) Representative FACS plots IL-6Ra of expression on indicated lymphocyte subsets from spleens of naive CD4^{Cre} wild type and CD4^{Cre} x IL-6R $\alpha^{fl/fl}$ mice.



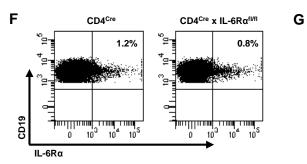
CD4

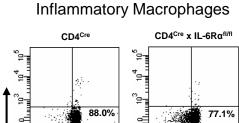






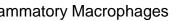
B cells





חתרותוותיים, 10⁴ 10 0 0 0

IL-6Rα (pregated on CD45+CD11b+Ly6Chigh)

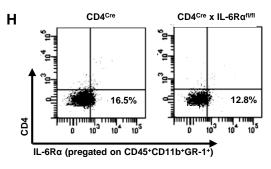


لستتت<u>ا</u> 10⁰

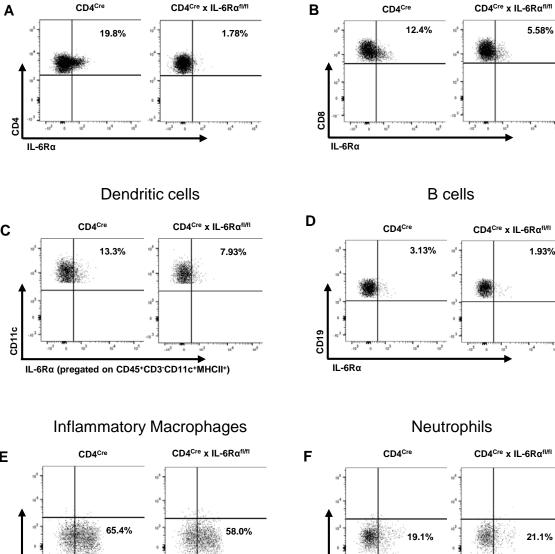
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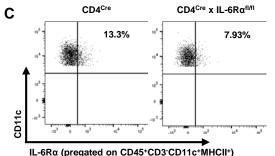
10⁴ 10⁵

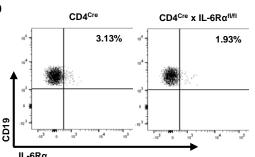
Neutrophils



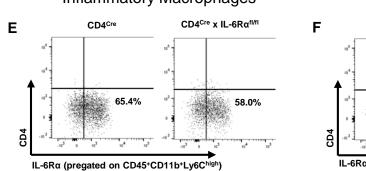
T cell subpopulations

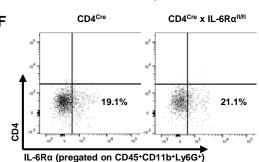




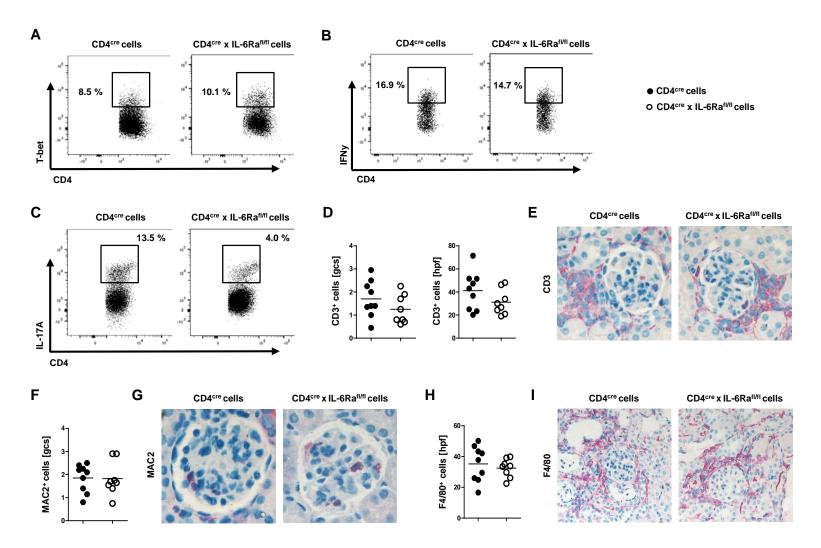


5.58%

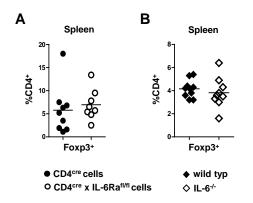




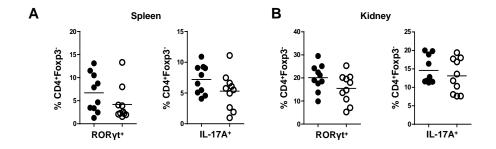
Supplementary Figure S3. (A-F) Representative FACS plots of IL-6Ra expression on indicated lymphocyte subsets from kidneys of naive CD4^{Cre} wild type and CD4^{Cre} x IL-6R $\alpha^{fl/fl}$ mice.



Supplementary Figure S4. (A-C) Representative FACS plots of renal (A) T-bet (B) IFNγ and (C) IL-17A expressing CD4⁺ Teff of the indicated recipient groups. (D) Quantification of renal glomerular and interstitial infiltration of CD3⁺ T cells. (E) Representative immunohistology of renal CD3⁺ T cells (red) of the indicated groups. (F) Quantification of infiltrating glomerular MAC2⁺ macrophages. (G) Representative immunohistology of glomerular MAC2⁺ macrophages (red) of the indicated groups. (H) Quantification of infiltrating interstitial F4/80⁺ Macrophages. (I) Representative immunohistology of interstitial F4/80⁺ macrophages (red) of the indicated groups.



Supplementary Figure S5. FACS analyses of splenic Tregs from (A) nephritic Rag1^{-/-} recipients of the indicated whole CD4⁺ T cell populations and (B) nephritic wild type and IL-6^{-/-} mice.



Supplementary Figure S6. FACS analyses of Th17 cells by quantification of RORyt activation and secretion of IL-17A as indicated in (A) spleens and (B) kidneys.