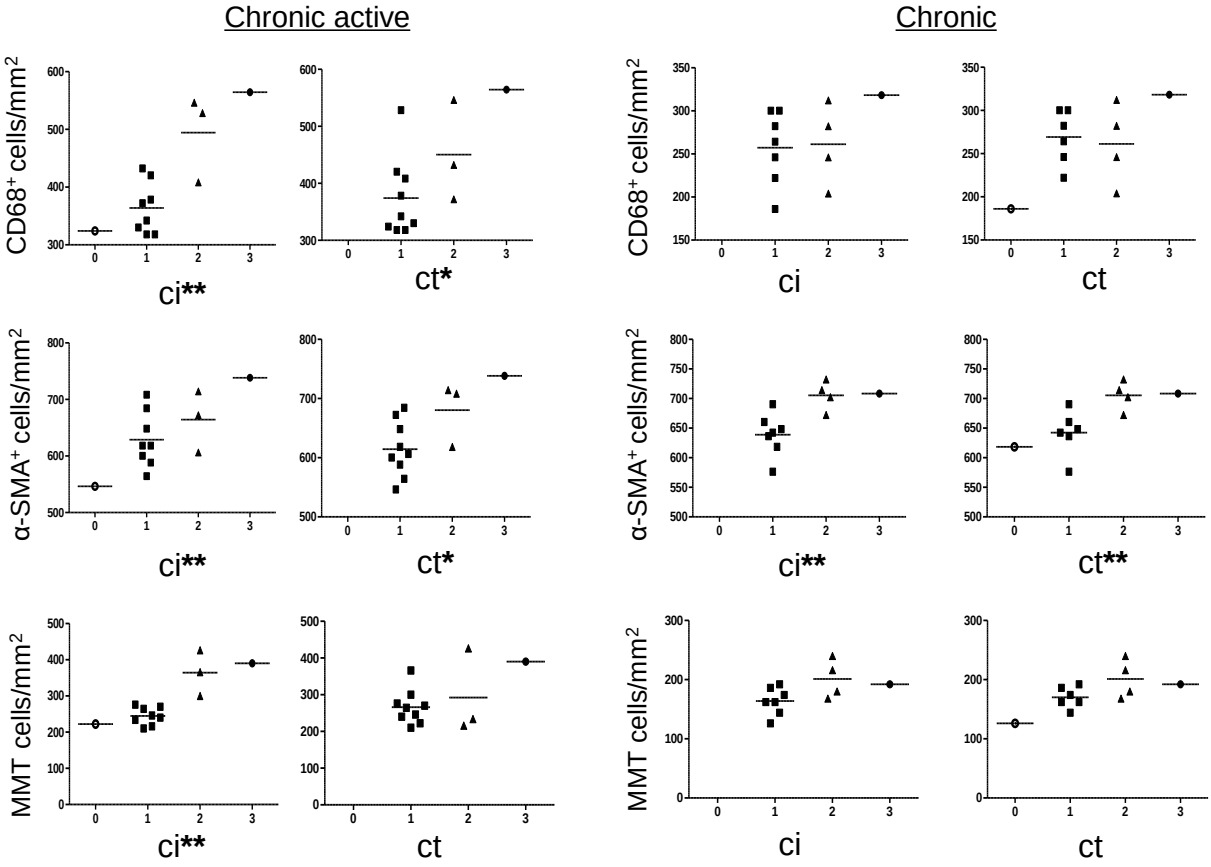

Figure S1. Correlation between Banff scores and number of MMT cells. In biopsies with chronic active transplant rejection, the number of CD68$^+$ macrophages and the number of α-SMA$^+$ myofibroblasts correlate with interstitial fibrosis (ci) and tubular atrophy (ct) scores, while the number of CD68$^+$α-SMA$^+$ MMT cells correlates with only the ci score. In biopsies with chronic allograft injury without evidence of active rejection (chronic), the scores of ci and ct correlate only with the number of α-SMA$^+$ myofibroblasts. Data represent mean for biopsies of same score. *p<0.05, **p<0.01 by Spearman's correlation coefficient.

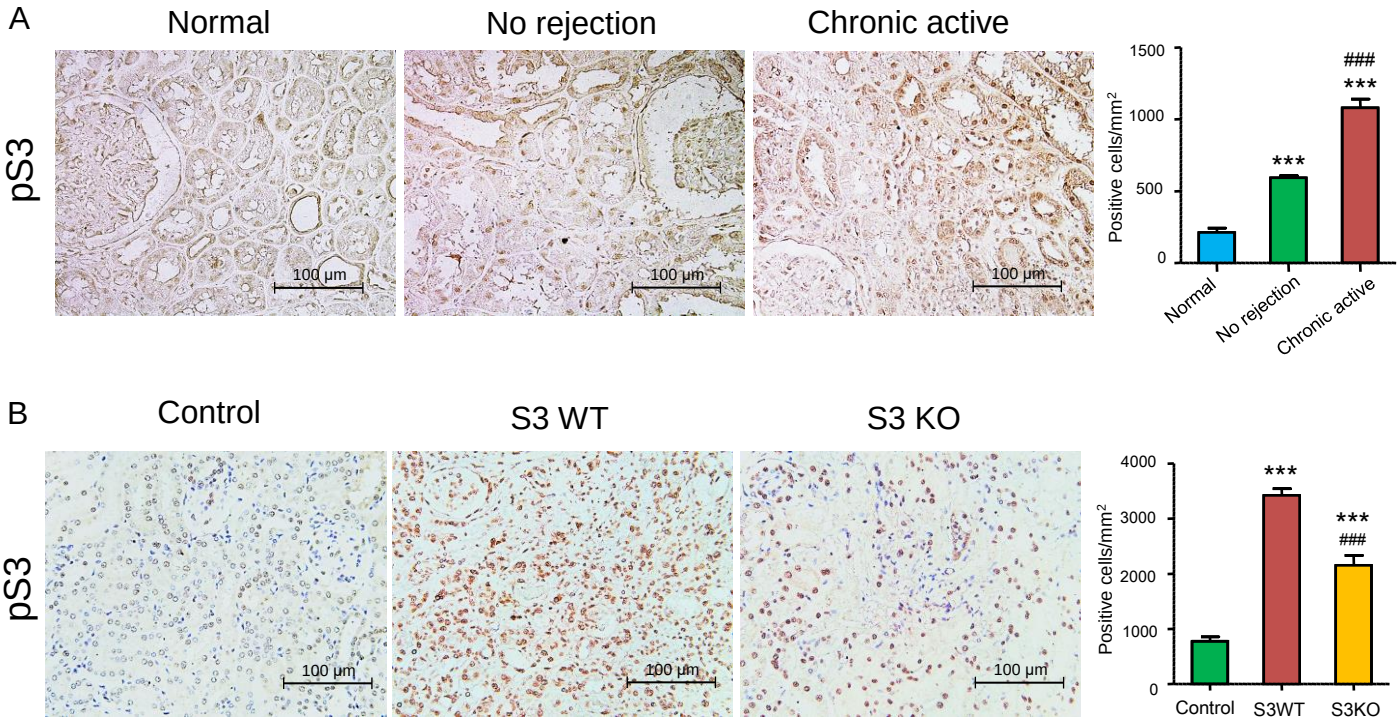

Figure S2. Smad3 is activated in renal allografts in both human and mouse samples. (A) Immunohistochemical staining of phospho-Smad3 (pS3) in human samples from normal kidneys, renal allografts without rejection and renal allografts with chronic active rejection. (B) Immunohistochemical staining of phospho-Smad3 in an isograft control and allografts in Smad3 wild type (WT) or Smad3 knockout (KO) recipient mice 28 days after transplantation. Graphs show quantification of the number of phospho-Smad3 positive cells. Data are mean ± SEM for groups of patients (A) as indicated in the Methods and groups of 6-8 mice (B). ***p<0.001 versus normal or isograft control; ###p<0.001 versus grafts from patients without rejection or Smad3 WT mice.

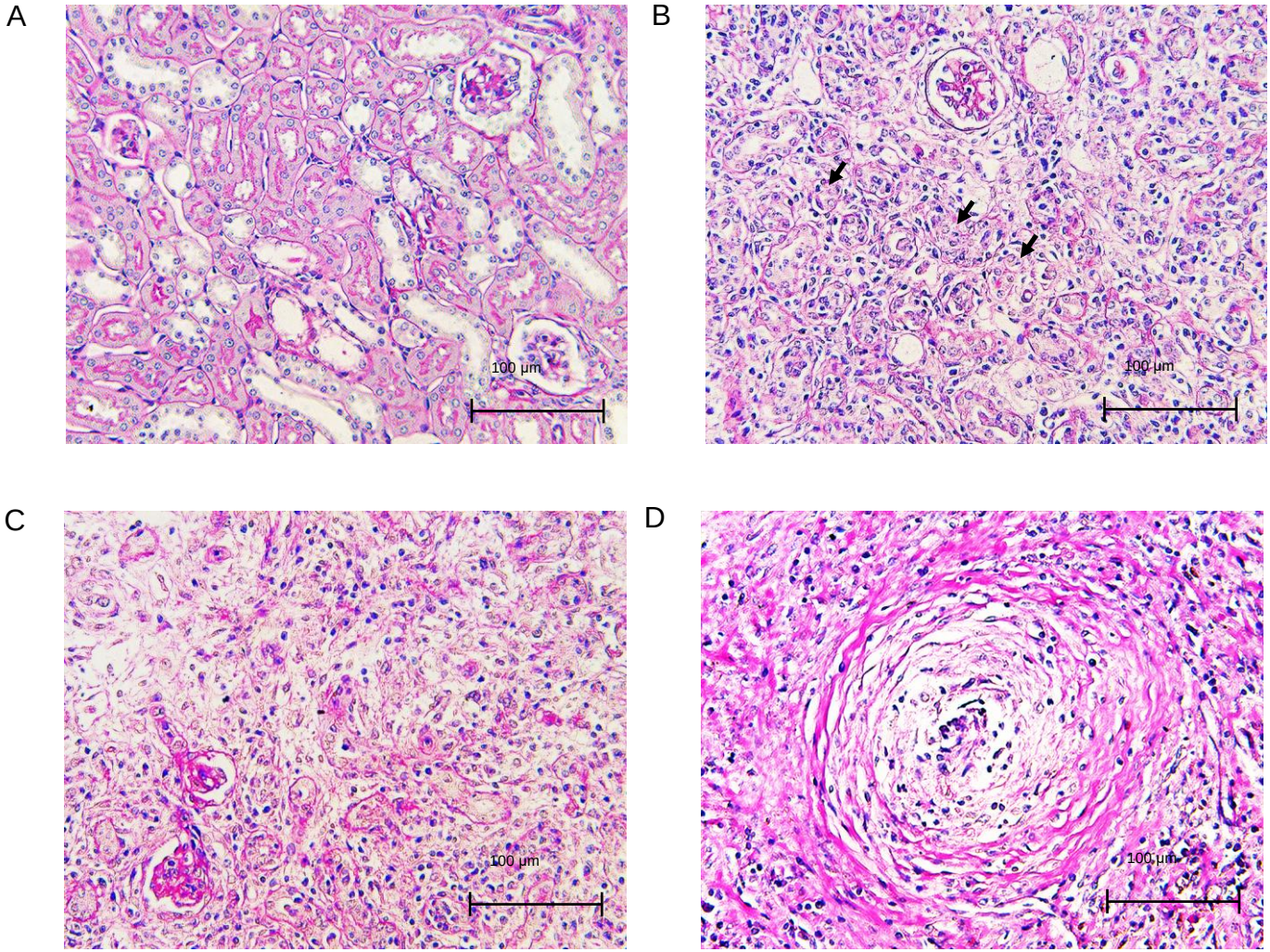

Figure S3. Typical allograft injuries in mouse renal transplantation. PAS staining for isograft (A) and allografts in Smad3 wild type recipients 28 days after transplantation(B, C and D). (A) Comparatively normal renal tissue. (B) Tubular atrophy (arrowheads); (C) severe interstitial fibrosis and (D) severe ateriopathy reveal chronic allograft injuries. Data represent a group of 6 mice.

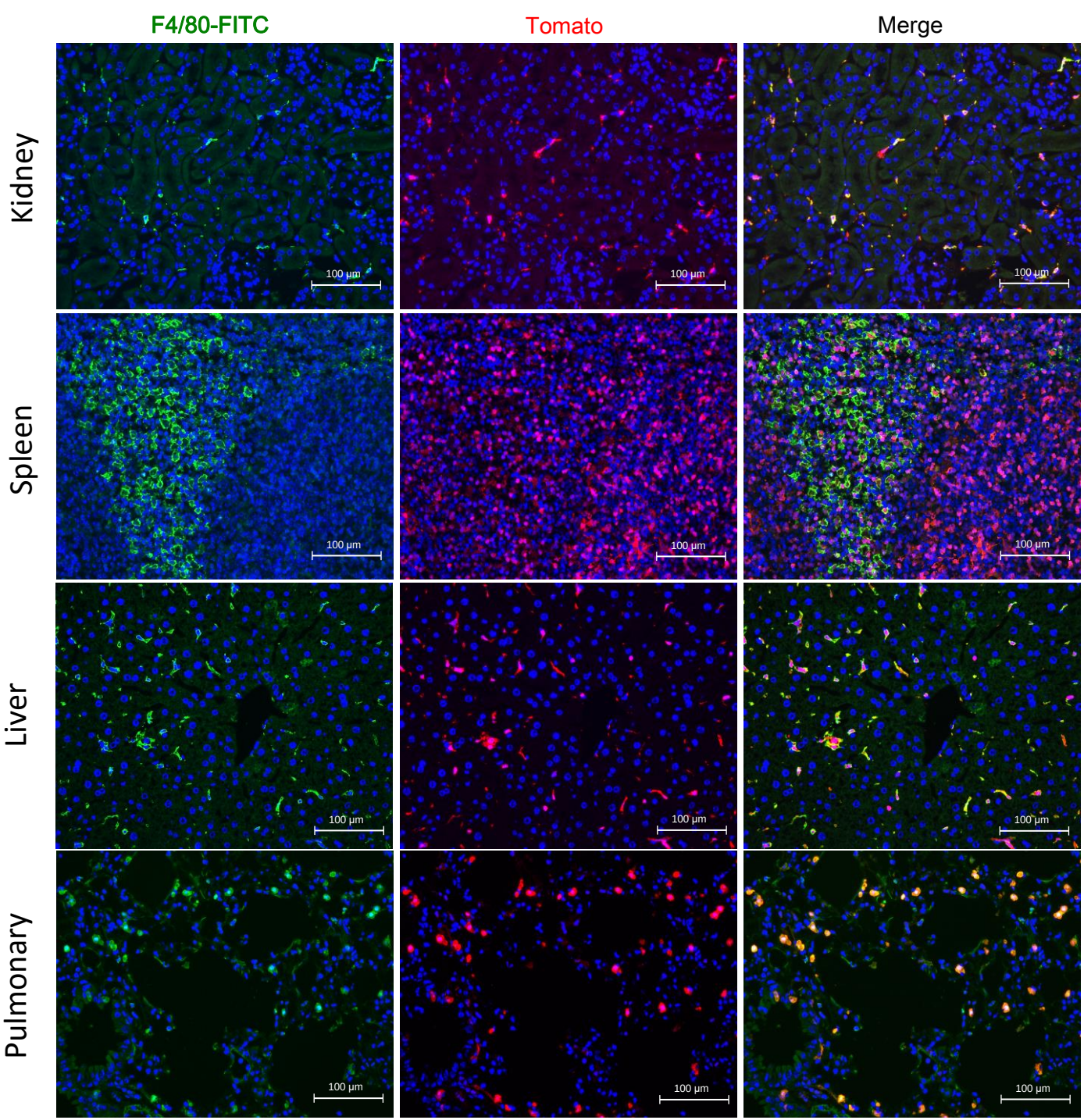

Figure S4. Lineage distribution of Tomato-expressing macrophages in normal tissues. Two-color immunofluorescence shows that almost all F4/80+ macrophages in the kidney, spleen, liver, and Lung tissues are co-expressing Tomato in normal Lyz2-Cre/Rosa26-Tomato. Note that many Tomato+ cells in spleen germinal centers are lack of F4/80, presumably dendritic cells. Results represent at least 6 mice. Scale bar, 100μm.

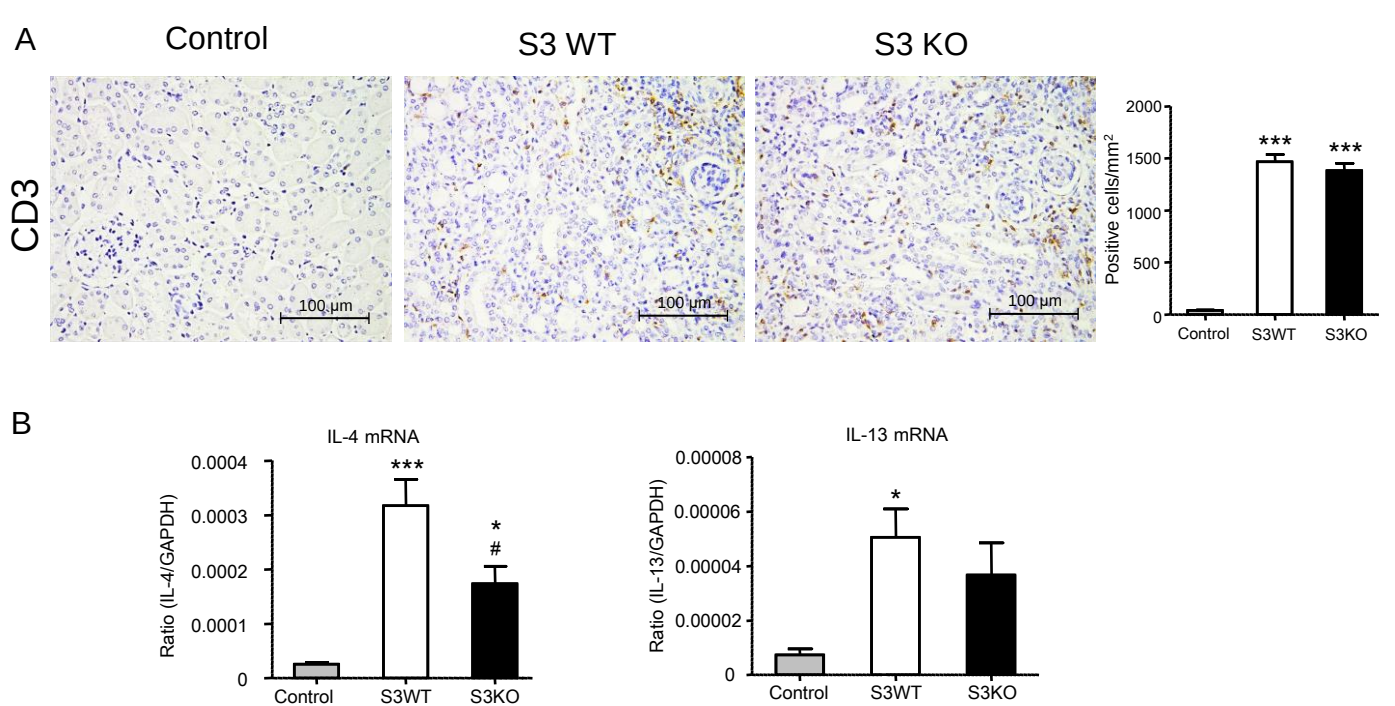

Figure S5. Deletion of Smad3 in the recipient reduces IL-4 expression without altering IL-13 expression and T cells infiltration in mouse chronic renal allograft rejection. Kidney allografts were transplanted into Smad3 wild type (WT) or Smad3 knockout (KO) recipient mice and examined 28 days later. An isograft group was used as a control. (A) Immunohistochemistry stain for $CD3^+$ cells. Graphs show quantification of the number of $CD3^+$ cells. (B) Real-time PCR analysis of RNA extracted from whole renal allograft tissue for IL-4 and IL-13. Data are mean $\pm$ SEM for groups of 6-8 mice. *$p<0.05$, ***$p<0.001$ versus isograft control; $^{\#}p<0.05$ versus Smad3 WT mice.

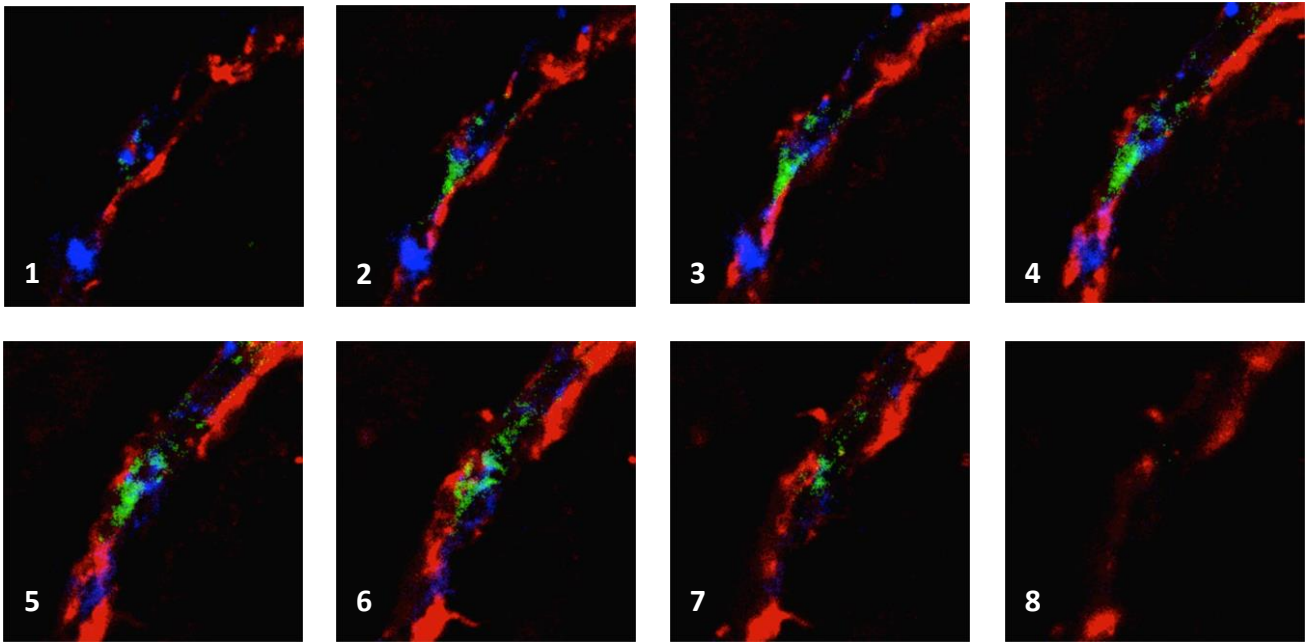

Figure S6. A Z-stack image shows a M2 macrophages (CD68+CD206+) co-expressing α-SMA in human chronic renal allograft rejection. CD68 (green), CD206 (blue), a-SMA (red). A Z-stack image is also shown in Supplementary video 4.

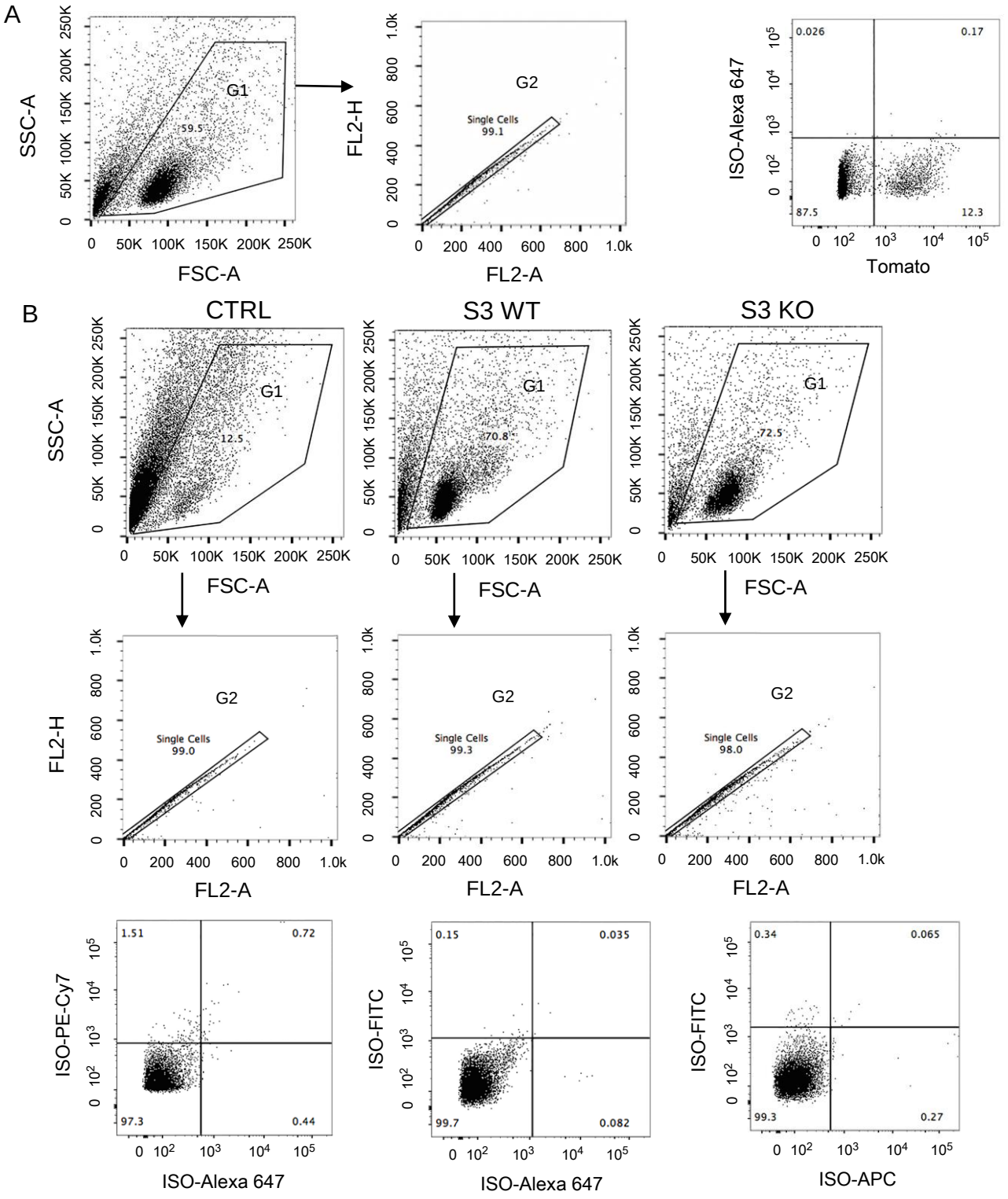

Figure S7. Flow cytometry gating strategy and isoform controls. (A) Cells isolated from renal grafts in Lyz2-Cre/Rosa26-Tomato recipient mice are gated (G1) on a forward scatter/side scatter (FSC-A/SSC-A). The G1 events are gated (G2) on FL2-A/FL2-H as singlet. The G2 events are visualized in isoform control for CD68-Alexa 647 and Figure 3 (D); (B) Cells isolated from renal grafts in control, S3 WT and S3KO recipient mice are gated (G1) on FSC-A/SSC-A. The G1 events are gated (G2) on FL2-A/FL2-H as singlet. The G2 events are visualized in isoform control for CD68-Alexa 647, α-SMA-Cy7, Collagen I-FITC, F4/80-APC and CD206-FITC and Figure 7(A), Figure 10(A).