Supplemental Figure Legend

Supplemental Figure 1: Splenic and renal BTLA expression
(A) Percentage of $BTLA^+$ cells and (B) MFI of BTLA expression in splenic Tregs ($CD3^+CD4^+Foxp3^+$), Th17 ($CD3^+CD4^+RORyt^+$), Th1 cells ($CD3^+CD4^+Tbet^+$), cDC1 ($MHC2^+CD11c^+Xcr1^+$), cDC2 ($MHC2^+CD11c^+CD11b^+$) and B cells ($CD19^+$) during homeostasis (d0) and at different time points throughout NTN. (C) Proportion of $BTLA^+$ and (D) MFI of BTLA in indicated renal dendritic cell subpopulations (cDC1 and cDC2) and B cells. Depicted is one representative experiment of a total of two experiments. Student's t test *p<0,05 (determined by Student's t-Test)

Supplemental Figure 2: *Btla*^{KO} mice show increased splenic T cell frequencies but do not develop spontaneous nephritis
(A) Albumin-to-creatinin ratio and (B) BUN of unchallenged *Btla*^{KO} and littermate controls. (C) Quantification of renal histological damage. (D) Representative kidney PAS stainings. (n=3 vs 4) (E) Total splenocyte numbers, counted using a Neubauer counting chamber. (F) Flow cytometry analysis of splenic total T helper cells ($CD3^+CD4^+$), $RORyt^+$ Th17 cells, $Tbet^+$ Th1 cells and $Foxp3^+$ Tregs. (G) Concentration of indicated cytokines in the supernatant of splenocytes isolated from healthy wt and *Btla*^{KO} mice and stimulated *in vitro* with anti-CD3 for 72h (n= 4 vs 5). All experiments were carried out using 8- month- old, male *Btla*^{KO} mice and littermate controls.
**p<0,01 (determined by Student's t-Test)

Supplemental Figure 3: Glomerular deposition of nephrotoxic sheep IgG
Immunofluorescence staining of sheep IgG (magenta) and podocin (green) in kidneys of (A) WT and (B) *Btla*^{KO} animals after 10 days of NTN. No difference in sheep IgG deposition was observed in the two groups.

Supplemental Figure 4: Renal immunity and NTS specific humoral response after 10 days of NTN
(A) IHC stainings reveal significantly more MAC-2+ glomerular macrophages in the kidney of Btla^{KO} mice. Flow cytometry analysis reveals no changes in renal (B) NKT cells, cytotoxic T cells, (C) yd T cells and upregulation of (D) CXCR3+ Tregs in Btla^{KO} mice. (E) Frequencies of renal $Xcr1^+$ cDC1, $CD11b^+$ cDC2 and $Ly6C^{hi}CD11b^+$ monocytes. (F) Serum titers of NTS-specific mouse IgM and total IgG as determined by ELISA. All analysis were carried out after 10 days of NTN. *p<0,05; ** p<0,01 (determined by Student's t-Test)

Supplemental Figure 5: Representative images of PAS stainings of BTLA^{KO} and antibody treatment studies
Representative low power images of renal damage after 10 days (A) and 21 days (B) of NTN in littermate controls and *Btla*^{KO} mice. (C) Representative images of healthy littermate control (left) and day 3 of NTN (right). Acute tubular damage and intratubular protein casts are visible. In addition, glomeruli show signs of mesangial expansion, representing early glomerular damage. (D) Histological damage after 14 days of NTN in control (left) and anti-BTLA-antibody treated mice (right).

Supplemental Figure 6: ACR and immunophenotyping of *Btla*^{KO} and WT after 21 days of NTN
Additional experiment showing course of ACR as well as histological damage and flow cytometry analysis at day 21 of NTN. (A) Development of ACR up to 20 days after NTS injection in *Btla*^{KO} and WT littermates reveals significant increase at days 7 and 10 in *Btla*^{KO}. (B) Representative images and quantitative scoring of glomerular damage at day 21 of NTN. (C) Renal immune cell composition as determined by flow cytometry 21 days after

NTS injection. *Btla*^{KO} mice show a significant increase in F4/80^{+} macrophages at this late stage of NTN, whereas no significant changes are observed in the other immune cell populations. (C) Splenic immune cell composition show a significant decrease in systemic Foxp3^{+} Treg frequencies. * p<0,05, ** p<0,01 (determined by Student's t-Test)

Supplemental Figure 7: scRNAseq analysis of renal T cell subclusters

(A) Schematic workflow of scRNAseq experiment. Nephritic Btla^{KO} and wild type mice (n= 2 vs 2) were sacrificed at day 10 of NTN. Renal immune cells were isolated using MACS and FACS before library preparation and sequencing. (B) Heatmap of the T cell cluster showing the top 5 markers of each subcluster. 9 subclusters were identified using an unsupervised clustering approach. (C) 4 T cell clusters are relatively expandet in *Btla*^{KO} mice, identified as Teff1 (Th1 cells), Tregs, cycling T cells and Teff3 (Th2-like cells). Volcano plots of pseudobulk analysis showing significantly up (red)- and downregulated (blue) genes in (D) T effector 1, (E) T effector 3 and (F) Treg clusters. (G) Treemap displaying the positively enriched gene ontology (GO) terms in Tregs between *Btla*^{KO} and WT mice samples. The size of rectangles in the treemap are proportional to the significance of the enrichment.

Supplemental Figure 8: Top 20 GO:Terms upregulated in total T cells and Tregs List of the 20 most significantly enriched GO:Terms of total T cells and Tregs, showing the genes associated with each term and p-values.

Supplemental Figure 9: BTLA deficient nephritogenic T cells show similar rates of apoptosis

Splenocytes isolated from nephritic Btla^{KO} and wild type mice were re-stimulated in vitro with sheep IgG and stained for apoptotic marker annexin V after 24h hours and measured using flow cytometry. Tregs were gated as CD4^{+}CD25^{+}CD127^{-} cells. 7- AAD^{+} events represent dead cells, annexin V^{+}, 7-AAD^{-} events represent apoptotic cells. Three biological replicates per group.

Supplemental Figure 10: Glomerular macrophage infiltration and C3 deposition in anti-BTLA antibody treated mice

IHC staining for MAC-2+ glomerular macrophages reveal reduced numbers in the intervention group. (A) Representative images of glomerular macrophages and quantification of MAC-2+ cells per glomerulus (B) Representative images of C3 deposition as detected by IHC. No differences were observed between the groups.

Supplemental Figure 11: Treg and Th1 survival and humoral responses in anti-BTLA-antibody treated mice

Anti-BTLA antibody administration has no impact on frequencies of living Tregs and Th1 cells in the kidney (A) and in the spleen (B). Percentages of living cells were determined by flow cytometry.

In addition, splenic B cell frequencies and NTS specific humoral responses are not affected by antibody stimulation. (C) Splenic B cell frequencies of indicated groups as determined by flow cytometry. NTS specific total mouse IgG (D) and IgG subtypes (E) as determined by standard ELISA techniques.

Gating strategy: renal Tregs CD45^{+}CD4^{+}CD3^{+}Foxp3^{+}RORyt^{-}; splenic Tregs CD45^{+}CD4^{+}CD3^{+}Foxp3^{+}; renal and splenic Th1 cells: CD45^{+}CD4^{+}CD3^{+}Tbet^{+}. Fixable viability dye was used to gate for living cells.

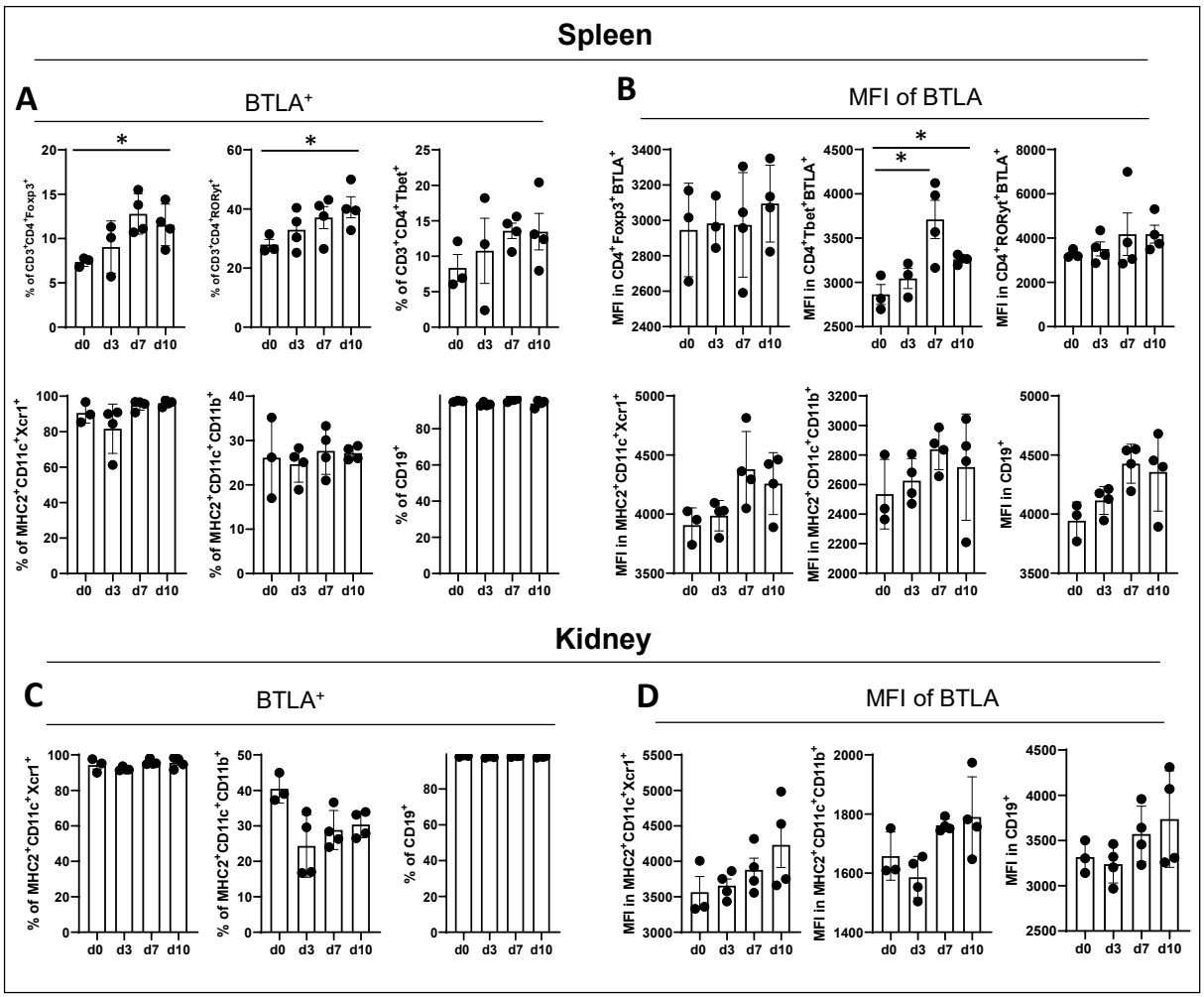
Supplemental Figure 12: Prophylactic anti-BTLA-antibody administration attenuate NTN

Administration of an agonistic anti-BTLA antibody attenuates NTN as shown by significantly reduced (A) albumin-to-creatinin ratios and (B) BUN after 10 days of NTN.

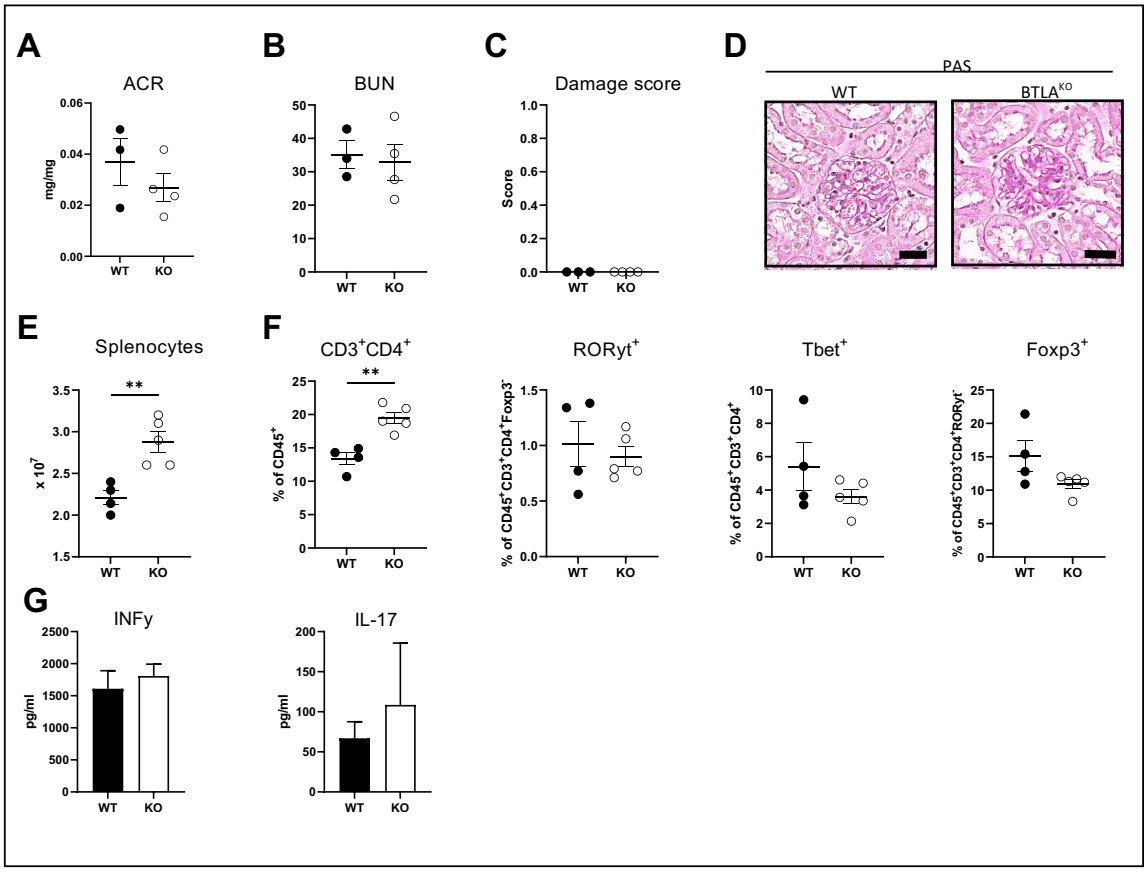
(C) Representative renal PAS stainings of indicated groups after 10 days of NTN. (D) Mice treated with the agonistic antibody show significantly reduced glomerular damage after 10 days of NTN. (E) Reduced numbers of glomerular and interstitial CD3+ cells are observed in the treatment group compared to controls, detected with IHC. (F) Representative IHC stainings for CD3 of indicated groups. Scale bars represent 20µm.
* p<0,05, ** p<0,01 (determined by Student's t-Test)

Supplemental Figure 13: Flow cytometry analysis of prophylactic BTLA-antibody study

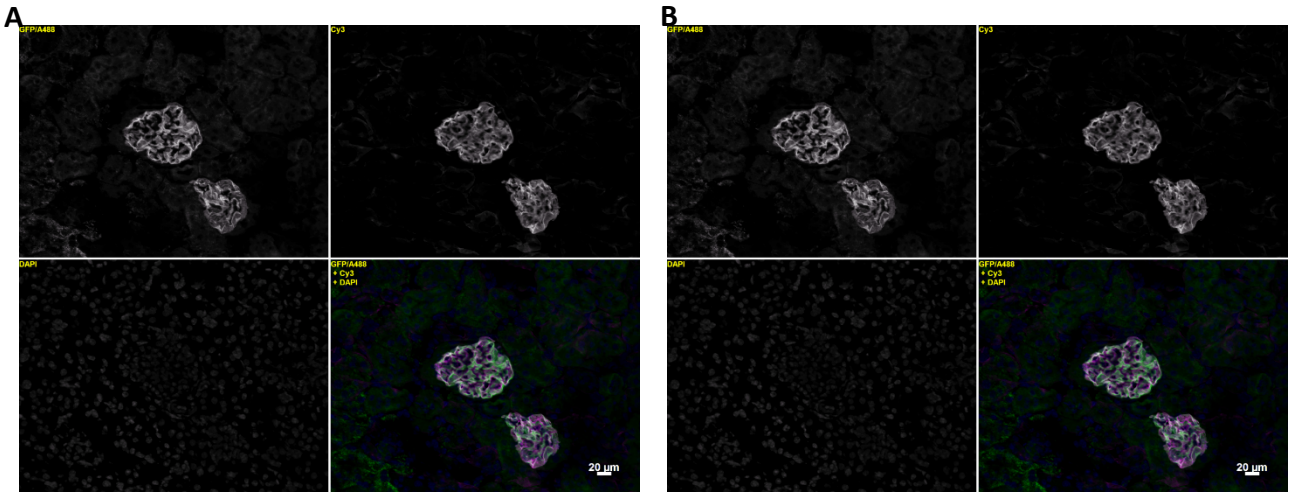
Flow cytometry analysis of renal (A) macrophages, (B) dendritic cells (total, $Xcr1^+$ cDC1 and $CD11b^+$ cDC2) and (C) B cells show a significant reduction of macrophages in the treatment group. (D) Frequencies of renal T helper subsets $ROR\gamma t^+$ Th17 and $Tbet^+$ Th1 cells are significantly reduced in the treatment group with no changes seen in $Foxp3^+$ Tregs. (E) Representative dot plots of renal $Tbet^+$ T helper cells. (F) Analysis of splenic RORyt+ Th17 and Tbet+ Th1 reveal no differences between the groups, while $Foxp3^+$ Tregs are significantly increased in the treatment group. (G) Representative dot plots of splenic $Foxp3^+$ Tregs. * p<0,05 (determined by Student's t-Test)



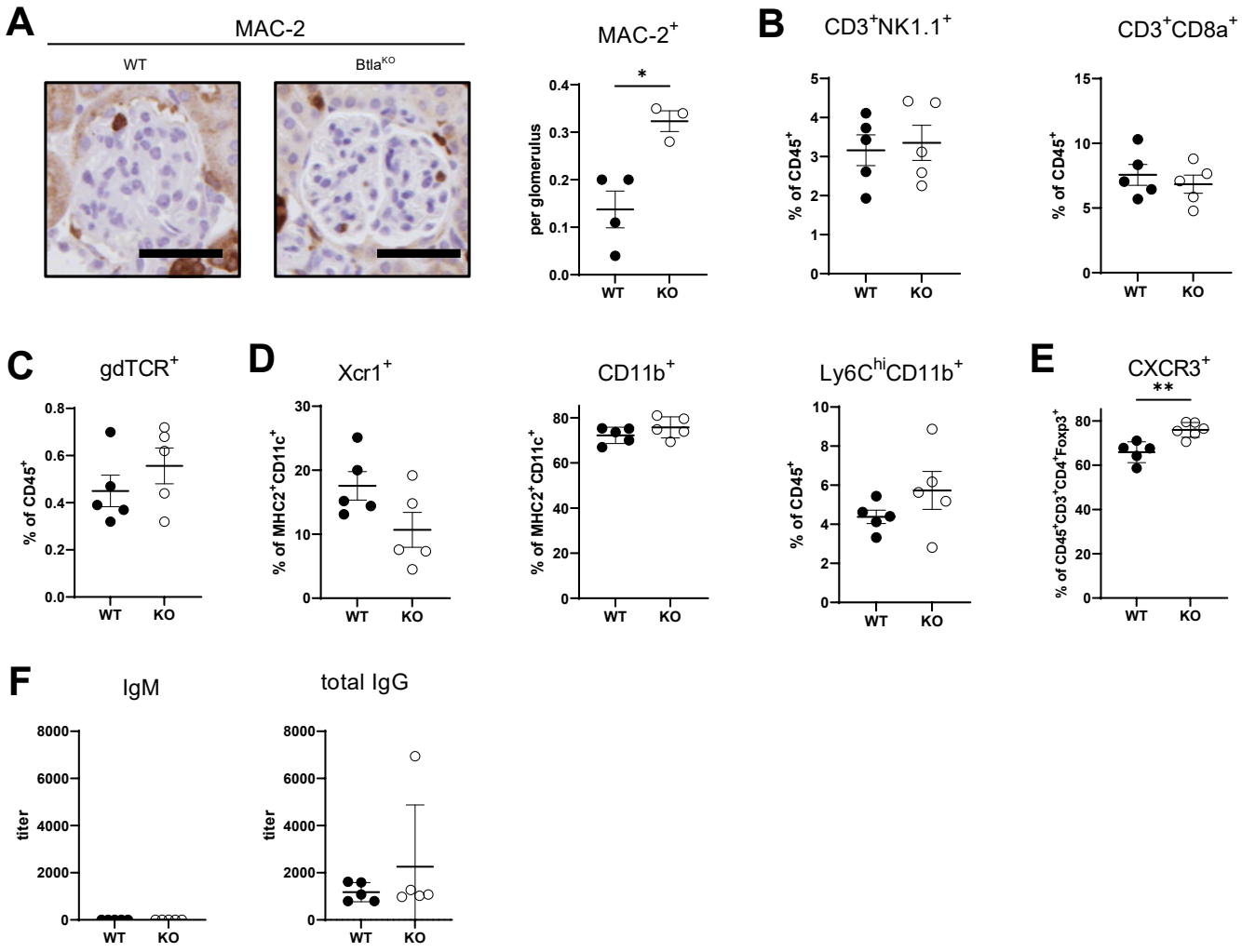
Supplemental Figure 1: Splenic and renal BTLA expression



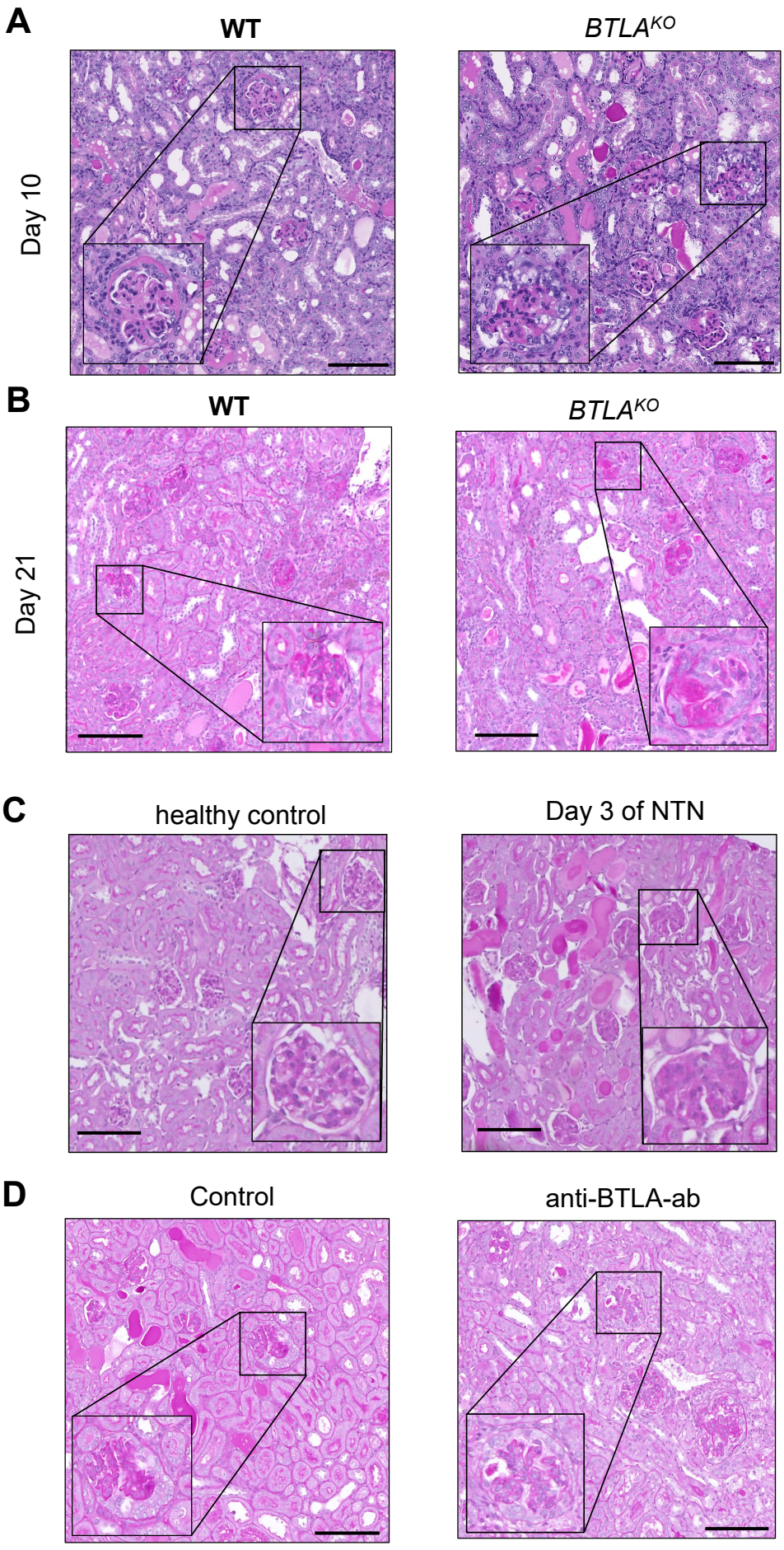
Supplemental Figure 2: Btla^{KO} mice show increased splenic T cell frequencies but do not develop spontaneous nephritis



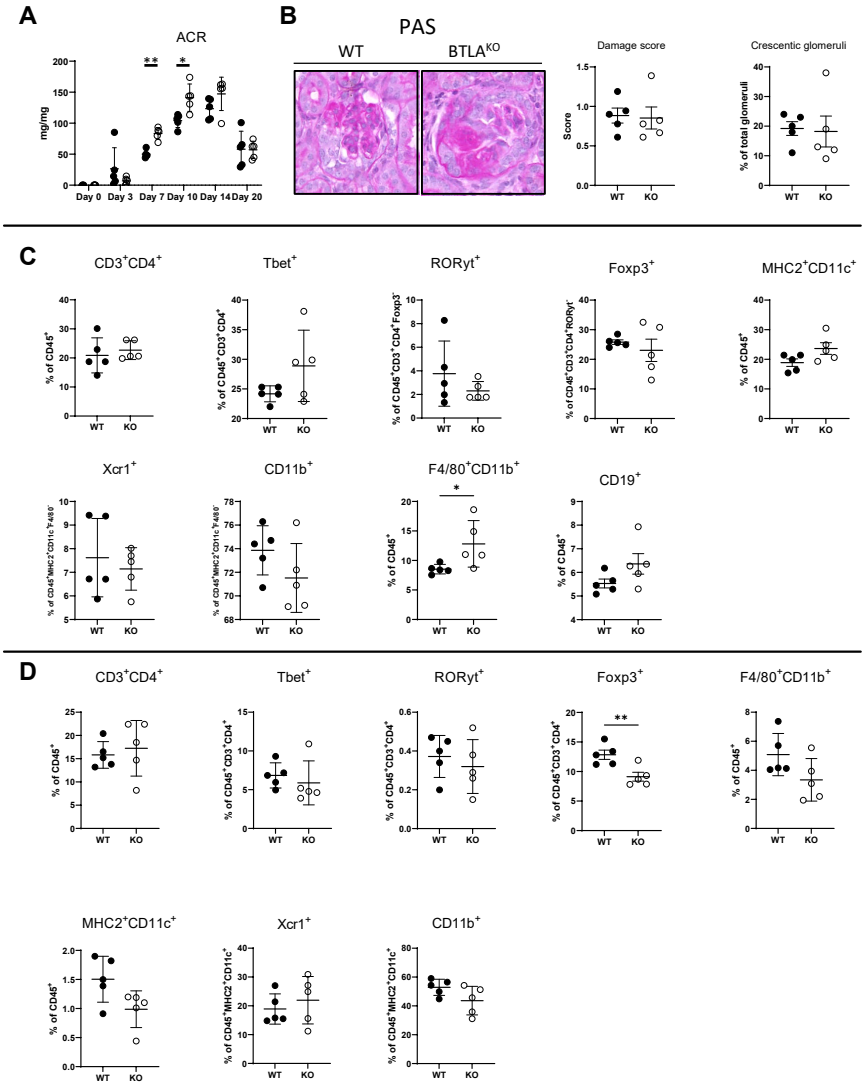
Supplemental Figure 3: Glomerular deposition of nephrotoxic sheep IgG



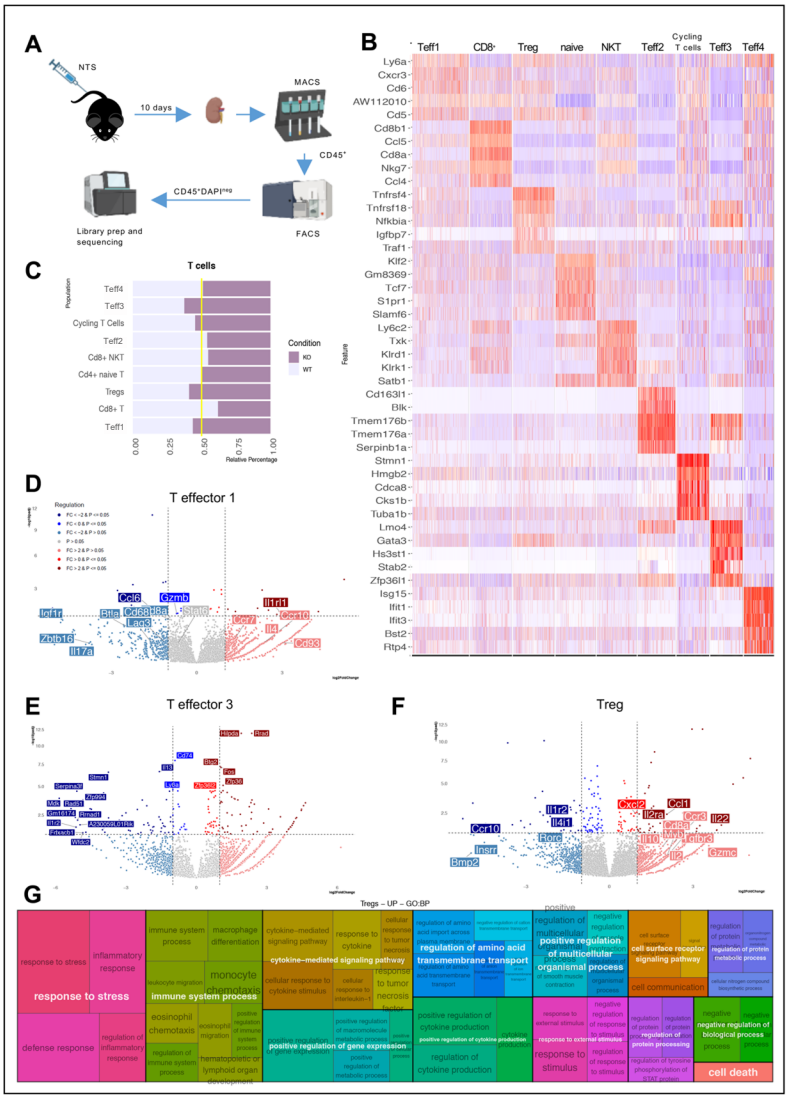
Supplemental Figure 4: Renal immunity and NTS specific humoral response after 10 days of NTN



Supplemental Figure 5: Representative images of PAS stainings of *Btla*^{KO} and antibody treatment studies



Supplemental Figure 6: ACR and immunophenotyping of *Btla*^{KO} and WT mice after 21 days of NTN



Supplemental Figure 7: scRNAseq analysis of renal T cell subclusters

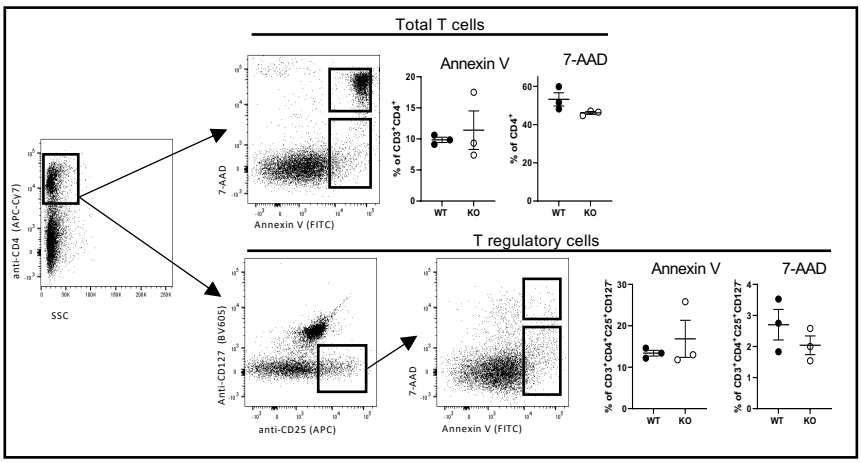
T cell cluster

GeneSetID	GeneSetName	Database	Regulation	P_value	Intersection
GO:0002684	positive regulation of immune system process	GO:BP	UP	2,18268E-10	Il1rl1,Gata3,Hes1,Ctla4,Il10,Il2ra,Il2,Tmem64,Tnfrsf18,Cxcl1,Ccl24,Calca,Arg1,Il5,Ccl1,Zfp36l1,Tnfsf11, Rgcc,Tfrc,Nfkbiz,Cd5,Cd6,Foxp3
GO:0006950	response to stress	GO:BP	UP	3,89141E-10	Il1rl1,Gata3,Areg,Tff1,mtCo1,Rbm3,Hilpda,Dst,Ctla4,Il10,Il2ra,Skil,Il2,Nfkb1,C1qtnf12,Tnfrsf4,Cxcl1,Ccl24,Rad18,Lig1,Ptgir,Calca,Casp3,Mt2,Arg1,Cirbp,Gadd45b,Il5,Mybbp1a,Ccl1,Nfkbia,Zfp36l1,Tnfsf11,Rgcc,Tfrc,Nfkbiz,Cd6,Foxp3,Pgk1,mt-Nd1,mt-Co2,mt-Atp6,mt-Nd4,mt-Nd5,mt-Cytb
GO:0006954	inflammatory response	GO:BP	UP	4,96771E-10	Il1rl1,Gata3,Il10,Il2ra,Il2,Nfkb1,C1qtnf12,Tnfrsf4,Cxcl1,Ccl24,Ptgir,Calca,Il5,Ccl1,Nfkbia,Tnfsf11,Tfrc,Nfkbiz,Cd6,Foxp3
GO:0002682	regulation of immune system process	GO:BP	UP	5,50817E-10	Il1rl1,Gata3,Hes1,Ctla4,Il10,Il2ra,Il2,Tmem64,Tnfrsf9,Tnfrsf18,Cxcl1,Ccl24,Calca,Casp3,Arg1,Il5,Ccl1,Nfkbia,Zfp36l1,Prxl2a,Tnfsf11,Rgcc,Tfrc,Nfkbiz,Cd5,Cd6,Foxp3
GO:0070661	leukocyte proliferation	GO:BP	UP	1,72356E-09	Hes1,Ctla4,Il10,Il2ra,Il2,Tnfrsf9,Tnfrsf4,Casp3,Arg1,Il5,Csf2,Tnfsf11,Tfrc,Cd6,Foxp3
GO:0070663	regulation of leukocyte proliferation	GO:BP	UP	6,22355E-09	Hes1,Ctla4,Il10,Il2ra,Il2,Tnfrsf9,Casp3,Arg1,Il5,Csf2,Tfrc,Cd6,Foxp3
GO:0051249	regulation of lymphocyte activation	GO:BP	UP	1,60846E-08	Gata3,Hes1,Ctla4,Il10,Il2ra,Il2,Tnfrsf9,Casp3,Arg1,Il5,Zfp36l1,Tnfsf11,Tfrc,Nfkbiz,Cd5,Cd6,Foxp3
GO:0051240	positive regulation of multicellular organismal process	GO:BP	UP	1,61194E-08	Il1rl1,Gata3,Hes1,Fabp5,Hilpda,Il10,Rgs2,Il2ra,Skil,Il2,Tmem64,Ncmap,Ccl24,Furin,Calca,Tent5a,Il5,Csf2,Ccl1,Zfp36l1,Tnfsf11,Rgcc,Tfrc,Nfkbiz,Cd6,Foxp3
GO:0050863	regulation of T cell activation	GO:BP	UP	1,75836E-08	Gata3,Hes1,Ctla4,Il2ra,Il2,Tnfrsf9,Casp3,Arg1,Tnfsf11,Tfrc,Nfkbiz,Cd5,Cd6,Foxp3
GO:0098609	cell-cell adhesion	GO:BP	UP	1,75993E-08	Gata3,Lgals7,Hes1,Ctla4,Il10,Il2ra,Il2,Calca,Pkp3,Casp3,Arg1,Zfp36l1,Tnfsf11,Rgcc,Tfrc,Stfa3,Nfkbiz, Cd5,Cd6,Foxp3
GO:0002694	regulation of leukocyte activation	GO:BP	UP	1,93044E-08	Il1rl1,Gata3,Hes1,Ctla4,Il10,Il2ra,Il2,Tnfrsf9,Casp3,Arg1,Il5,Zfp36l1,Tnfsf11,Tfrc,Nfkbiz,Cd5,Cd6,Foxp3
GO:0048522	positive regulation of cellular process	GO:BP	UP	2,09567E-08	Il1rl1,Gata3,Areg,Hes1,Ikzf2,Rbm3,Fabp5,Hilpda,Ctla4,Il10,Rgs2,Il2ra,Sema6d,Dstn,Skil,Il2,Crtc2,Nfkb1, Tmem64,Ncmap,C1qtnf12,Tnfrsf18,Cxcl1,Ccl24,Rad18,Fosb,Furin,Calca,Casp3,Tent5a,Arg1,Cirbp, Gadd45b,Socs2,Phlda1,Il5,Csf2,Mybbp1a,Ccl1,Nfkbia,Zfp36l1,Tnfsf11,Rgcc,Tfrc,Nfkbiz,Tiam1,Cd5, Cd6,Foxp3
GO:0008283	cell population proliferation	GO:BP	UP	2,28113E-08	Il1rl1,Gata3,Areg,Hes1,Tff1,Hilpda,Ctla4,Il10,Il2ra,Il2,Tnfrsf9,Tnfrsf4,Cxcl1,Ccl24,Ptgir,Calca,Mki67, Casp3,Arg1,Il5,Csf2,Nfkbia,Zfp36l1,Tnfsf11,Rgcc,Tfrc,Tiam1,Cd6,Foxp3
GO:0051239	regulation of multicellular organismal process	GO:BP	UP	2,53067E-08	Il1rl1,Gata3,Hes1,Fabp5,Hilpda,Ctla4,Il10,Rgs2,Il2ra,Sema6d,Skil,Il2,Nfkb1,Tmem64,Ncmap,Ccl24,Furin, Calca,Tent5a,Arg1,Socs2,Il5,Csf2,Ccl1,Nfkbia,Zfp36l1,Prxl2a,Tnfsf11,Rgcc,Tfrc,Nfkbiz,Cd6,Foxp3, Pgk1
GO:0048534	hematopoietic or lymphoid organ development	GO:BP	UP	2,71585E-08	Gata3,Hes1,Ctla4,Il10,Il2ra,Il2,Nfkb1,Tmem64,Tnfrsf9,Cxcl1,Calca,Casp3,Il5,Csf2,Nfkbia,Zfp36l1,Prxl2a, Tnfsf11,Tfrc,Nfkbiz,Foxp3
GO:0050670	regulation of lymphocyte proliferation	GO:BP	UP	2,93274E-08	Hes1,Ctla4,Il10,Il2ra,Il2,Tnfrsf9,Casp3,Arg1,Il5,Tfrc,Cd6,Foxp3
GO:0007155	cell adhesion	GO:BP	UP	3,74204E-08	Gata3,Lgals7,Hes1,Dst,Ctla4,Il10,Il2ra,Il2,Tnfrsf18,Calca,Pkp3,Casp3,Izumo1r,Arg1,Zfp36l1,Tnfsf11, Rgcc,Tfrc,Stfa3,Nfkbiz,Tiam1,Cd5,Cd6,Foxp3
GO:0032944	regulation of mononuclear cell proliferation	GO:BP	UP	3,74509E-08	Hes1,Ctla4,Il10,Il2ra,Il2,Tnfrsf9,Casp3,Arg1,Il5,Tfrc,Cd6,Foxp3
GO:0042110	T cell activation	GO:BP	UP	5,04715E-08	Gata3,Hes1,Ctla4,Il2ra,Il2,Tnfrsf9,Tnfrsf4,Casp3,Arg1,Zfp36l1,Tnfsf11,Tfrc,Nfkbiz,Cd5,Cd6,Foxp3
GO:0071356	cellular response to tumor necrosis factor	GO:BP	UP	5,94239E-08	Gata3,Hes1,Nfkb1,Tnfrsf4,Tnfrsf18,Ccl24,Calca,Ccl1,Nfkbia,Zfp36l1,Tnfsf11

Treg cluster

GeneSetID	GeneSetName	Database	Regulation	P_value	Intersection
GO:0006950	response to stress	GO:BP	UP	4,44816E-07	Calca,Il1rl1,Il2ra,Nfkb1,Plk3,AI506816,Cxcl2,Ruvbl1,Klrg1,Dctpp1,Eif4ebp1,Mt2,Cotl1, Itgb1,Il22,Il5,Ccl1,Ccl3,Ddx1,Ctla2a,Lgals3,Ltb4r1,Gzmb,Myh9,Tff1,Gnl1,Malt1,Jak2,Rbm3,mt-Nd1, mt-Co2,mt-Atp6,mt-Cytb
GO:0006954	inflammatory response	GO:BP	UP	1,1257E-05	Calca,Il1rl1,Il2ra,Nfkb1,Cxcl2,Itgb1,Il22,Il5,Ccl1,Ccl3,Ctla2a,Ltb4r1
GO:0006952	defense response	GO:BP	UP	1,60705E-05	Calca,Il1rl1,Il2ra,Nfkb1,Cxcl2,Klrg1,Cotl1,Itgb1,Il22,Il5,Ccl1,Ccl3,Ddx1,Ctla2a,Lgals3,Ltb4r1,Gzmb
GO:0010628	positive regulation of gene expression	GO:BP	UP	4,22456E-05	Calca,Csf2,Il1rl1,Nfkb1,Rpl5,Ruvbl1,Furin,Il5,Ccl1,Ccl3,Ddx1,Myh9,Cd200,Malt1,Jak2,Rbm3
GO:0001819	positive regulation of cytokine production	GO:BP	UP	0,000379323	Calca,Csf2,Il1rl1,Furin,Ccl1,Ccl3,Ddx1,Cd200,Malt1,Jak2
GO:0019221	cytokine-mediated signaling pathway	GO:BP	UP	0,000849728	Csf2,Il1rl1,Il2ra,Tnfrsf18,Cxcl2,Il5,Ccl1,Ccl3
GO:0007166	cell surface receptor signaling pathway	GO:BP	UP	0,000977241	Csf2,Il1rl1,Il2ra,Tbl1xr1,Nfkb1,Tln1,Tnfrsf18,Cxcl2,Ruvbl1,Klrg1,Furin,Eif4ebp1,Itgb1,Il22,Il5,Ccl1,Ccl3
GO:0071345	cellular response to cytokine stimulus	GO:BP	UP	0,001236818	Calca,Csf2,Il1rl1,Il2ra,Nfkb1,Tnfrsf18,Cxcl2,Il5,Ccl1,Ccl3,Kat6b,Rnmt,Jak2
GO:0050727	regulation of inflammatory response	GO:BP	UP	0,001889437	Il1rl1,Il2ra,Nfkb1,Il22,Ccl1,Ccl3,Ctla2a,Jak2
GO:0002376	immune system process	GO:BP	UP	0,001996746	Calca,Csf2,Il1rl1,Il2ra,Nfkb1,Tnfrsf18,Cxcl2,Klrg1,Itgb1,Il5,Ccl1,Ccl3,Ddx1,Ctla2a,Kat6b,Lgals3,Gzmb, Myh9,Cd200,Malt1,Jak2
GO:0001817	regulation of cytokine production	GO:BP	UP	0,002254711	Calca,Csf2,Il1rl1,Nfkb1,Furin,Ccl1,Ccl3,Ddx1,Cd200,Malt1,Jak2
GO:0001816	cytokine production	GO:BP	UP	0,002433826	Calca,Csf2,Il1rl1,Nfkb1,Furin,Ccl1,Ccl3,Ddx1,Cd200,Malt1,Jak2
GO:0048519	negative regulation of biological process	GO:BP	UP	0,002546899	Calca,Csf2,Il1rl1,Rgs2,Il2ra,Tbl1xr1,Nfkb1,Plk3,Tnfrsf18,Rpl5,Zfp422,Furin,Eif4ebp1,Mt2,Rrad,Itgb1, Crtap,Il22,Slc43a2,Ccl3,Birc5,Ctla2a,Kat6b,Lgals3,Myh9,H1f0,Cd200,Tff1,Ier3,Jak2,Eif3a,Rbm3
GO:0034097	response to cytokine	GO:BP	UP	0,003754015	Calca,Csf2,Il1rl1,Il2ra,Nfkb1,Tnfrsf18,Cxcl2,Il5,Ccl1,Ccl3,Kat6b,Rnmt,Jak2
GO:0030225	macrophage differentiation	GO:BP	UP	0,004216399	Calca,Csf2
GO:1903789	regulation of amino acid transmembrane transport	GO:BP	UP	0,005313144	Rgs2,Itgb1,Slc43a2
GO:0010958	regulation of amino acid import across plasma membrane	GO:BP	UP	0,005313144	Rgs2,Itgb1,Slc43a2
GO:0050900	leukocyte migration	GO:BP	UP	0,007388864	Calca,Tnfrsf18,Cxcl2,Itgb1,Ccl1,Ccl3,Lgals3
GO:0051240	positive regulation of multicellular organismal process	GO:BP	UP	0,007674145	Calca,Csf2,Il1rl1,Rgs2,Il2ra,Ncmap,Furin,Itgb1,Il5,Ccl1,Ccl3,Ddx1,Cd200,Malt1,Jak2
GO:0002548	monocyte chemotaxis	GO:BP	UP	0,007784988	Calca,Ccl1,Ccl3,Lgals3

Supplemental Figure 8: Top 20 GO:Terms upregulated in total T cells and Tregs



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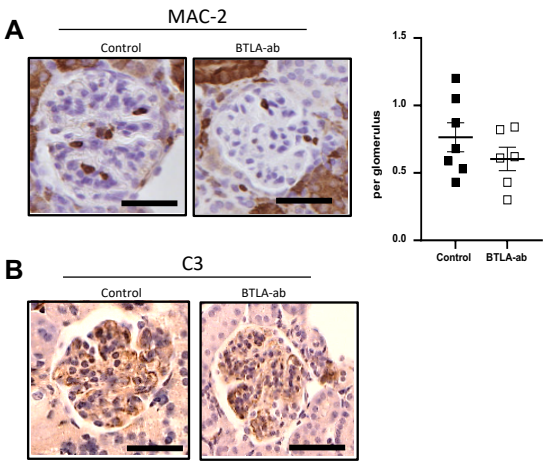

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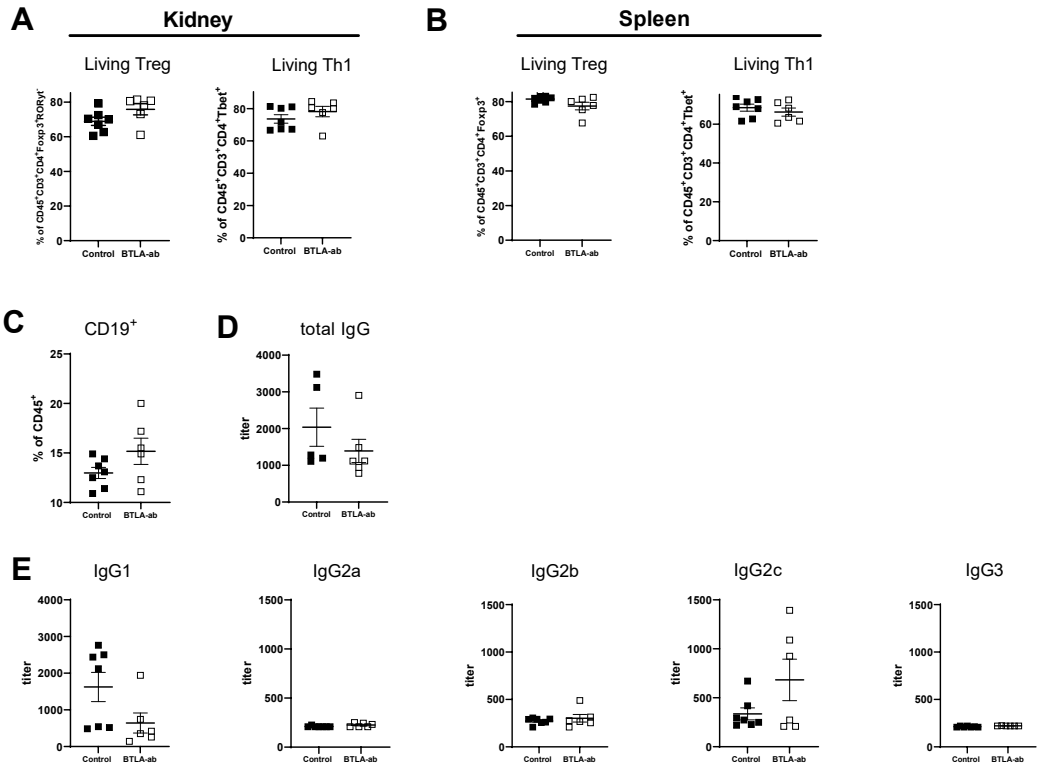

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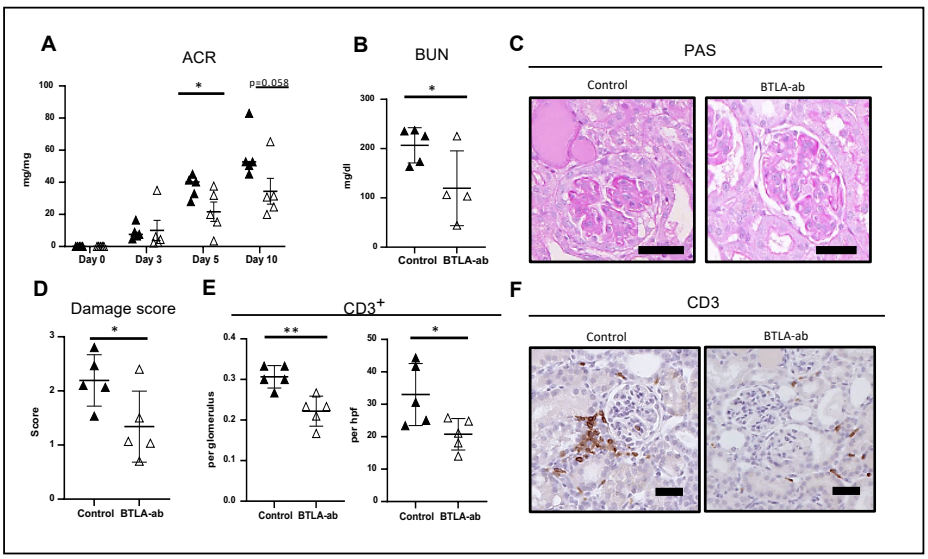
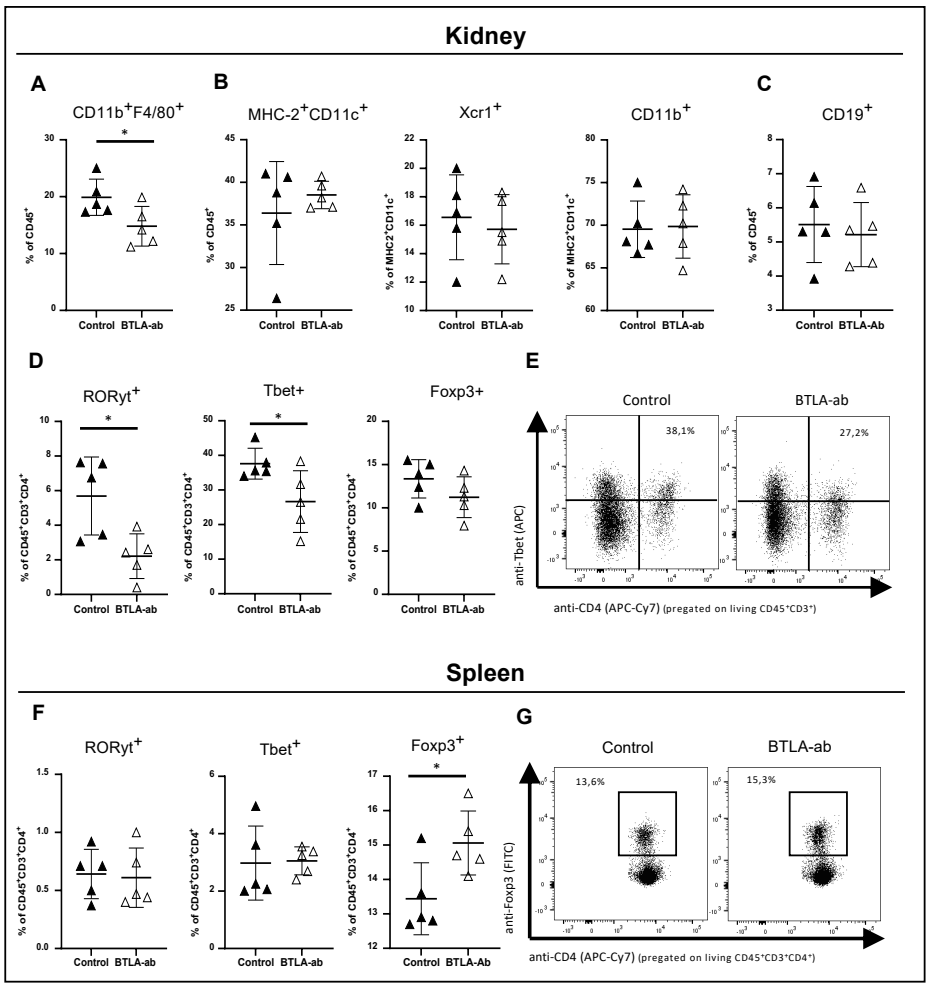


Supplemental Figure 12: Prophylactic BTLA-antibody administration attenuates NTN



Supplemental Figure 13: Flow cytometry analysis of prophylactic BTLA-antibody study