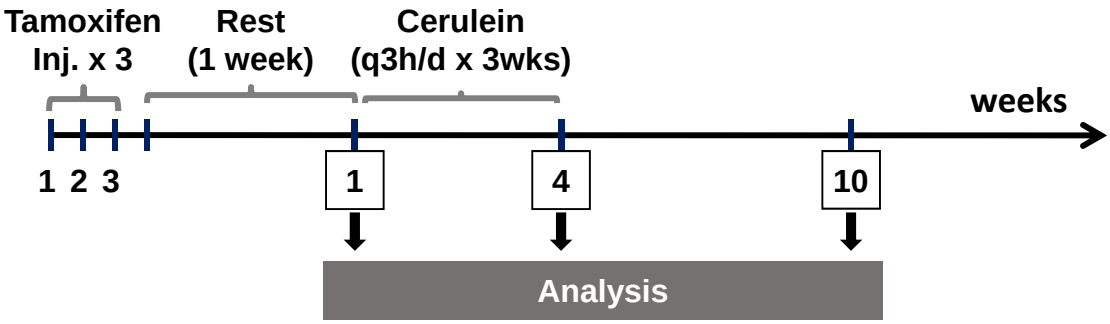
Supplementary Figure 1

a



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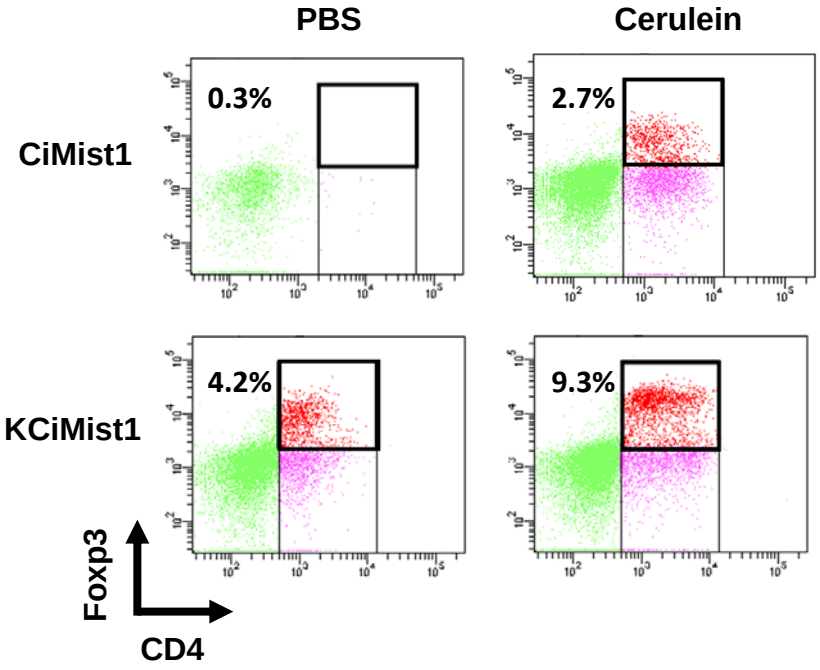

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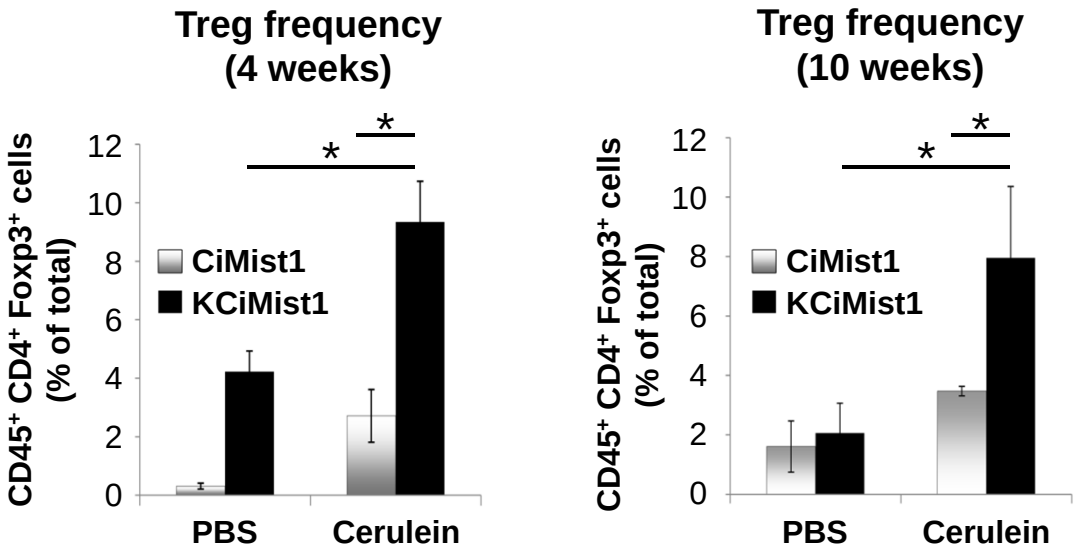
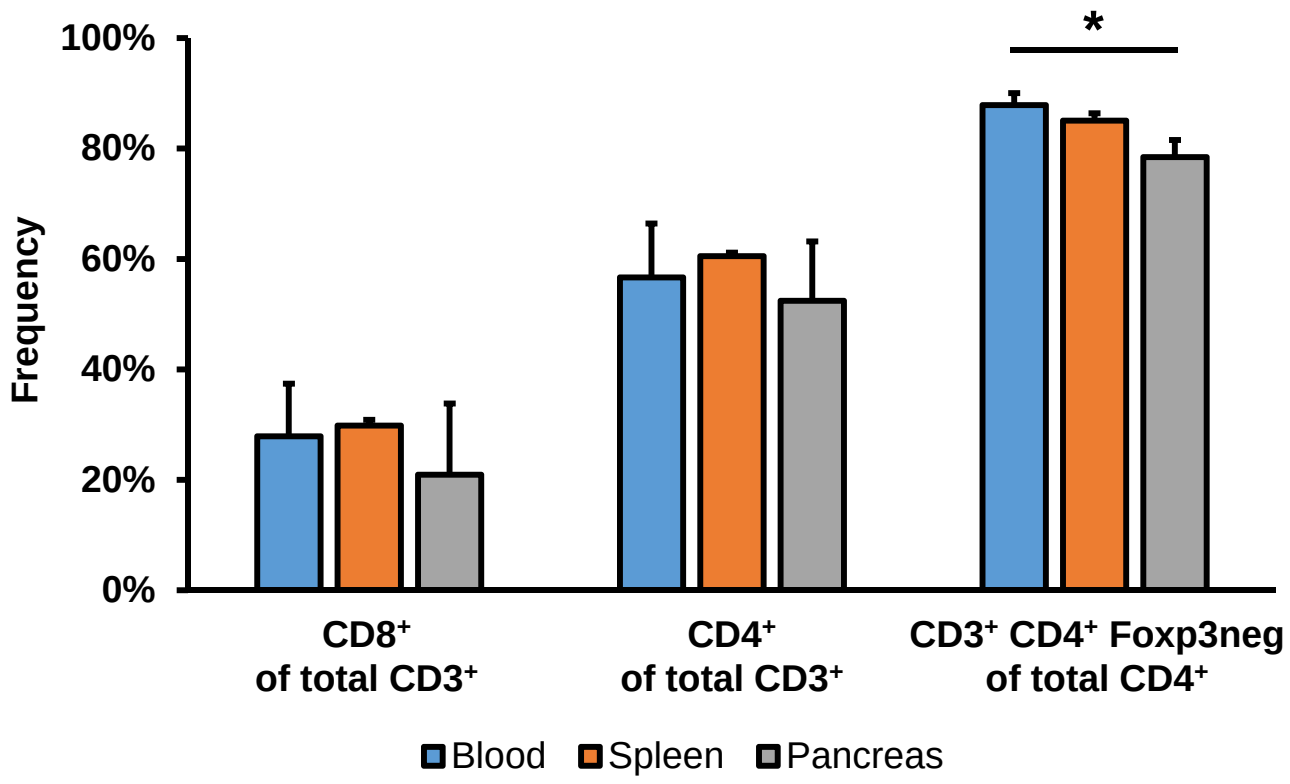


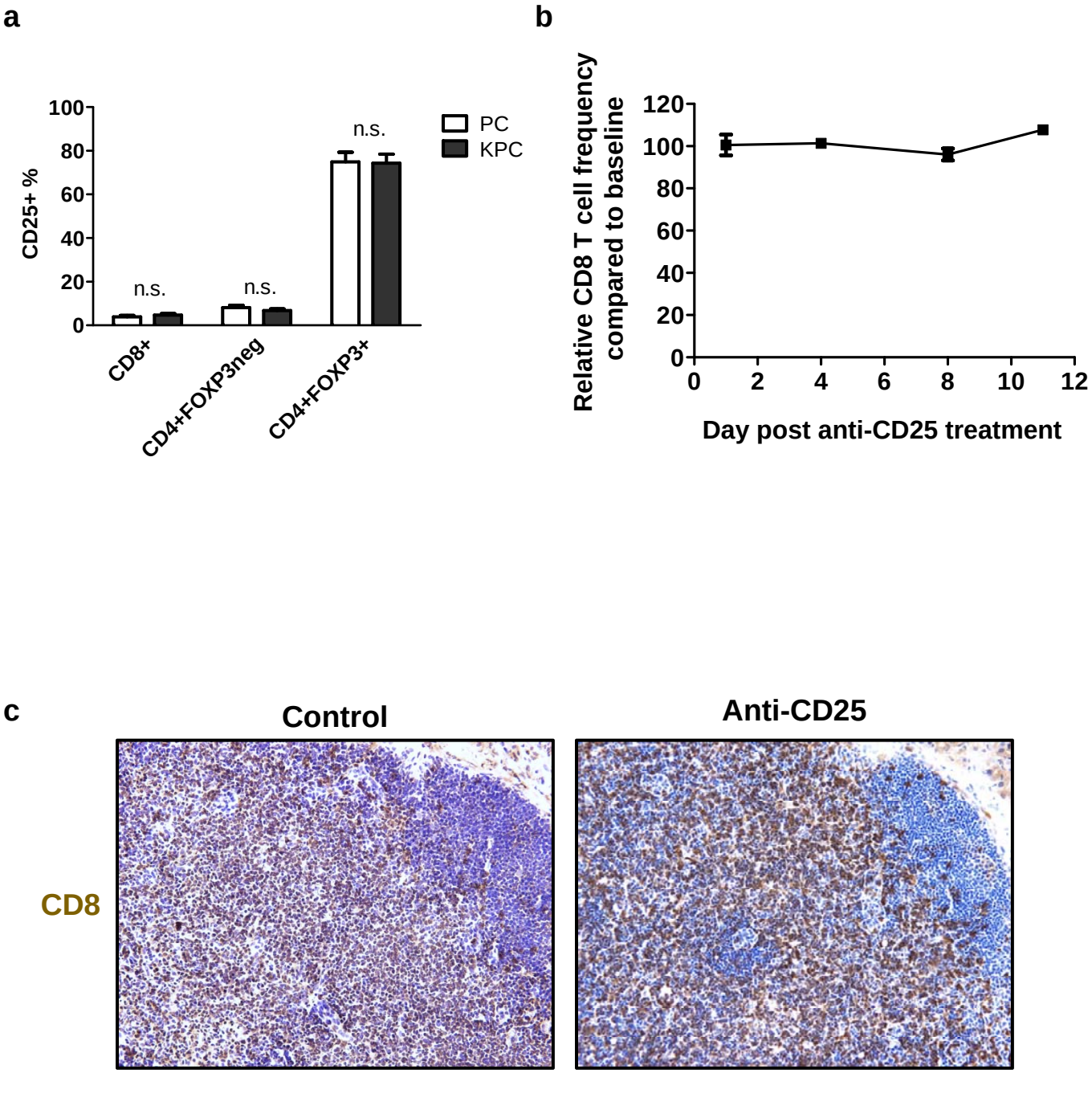
Supplementary Figure 1. FOXP3^{+} CD4^{+} regulatory T cells accumulate in the pancreas of *Mist1Cre ERT2; LSL-Kras$^{G12D/+}$* (KCi Mist1) mice during chronic pancreatic inflammation induced with cerulean. a, Schematic showing the study design for treatment with tamoxifen and cerulein (treatment for 3 weeks, 3 times/week, 6 injections/day) to induce expression of Kras^{G21D} and chronic pancreatitis, respectively. **b,** Representative flow cytometry dot plots showing FOXP3^{+}CD4^{+} Tregs in the pancreas of CiMist (controls) and KCiMist1 mice upon activation of Kras with or without chronic exposure to cerulein. **c,** Quantification of Treg frequency in CiMist and KCiMist1 mice at 4 weeks (left panel) and 10 weeks (right panel) after Kras activation with or without chronic exposure to cerulein. n = 4-6 mice/group. Significance was determined by Students *t* test. *, $p<0.05$.

Supplementary Figure 2



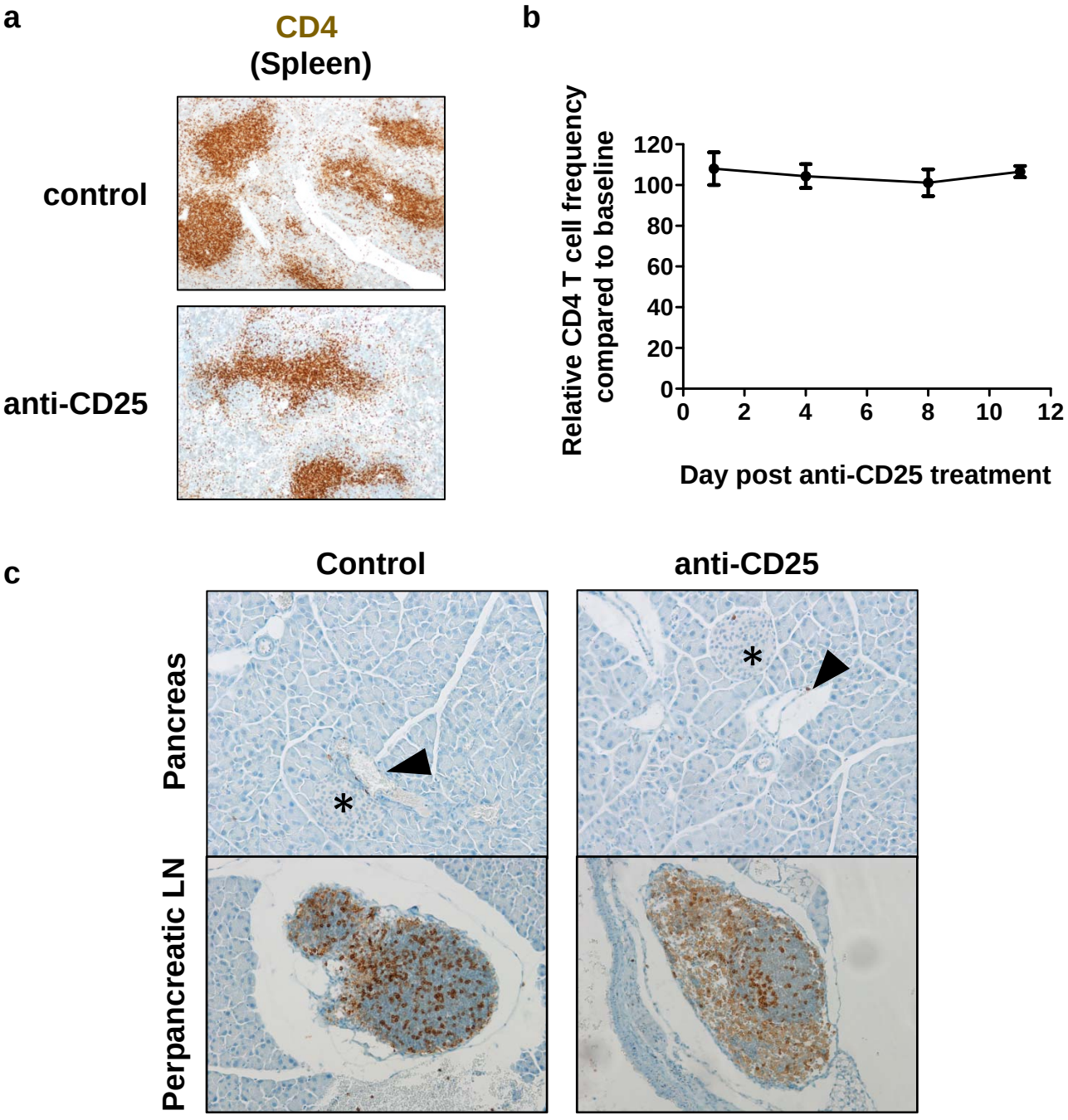
Supplemental Figure 2. T cell subset frequencies in blood, spleen and pancreas of tumor-bearing KPC mice. Cell frequencies were determined by flow cytometry of single cell suspensions obtained from the indicated tissues. Statistical significance was determined by non-parametric *t* test. *, $p < 0.05$. *n* = 3/group.

Supplementary Figure 3



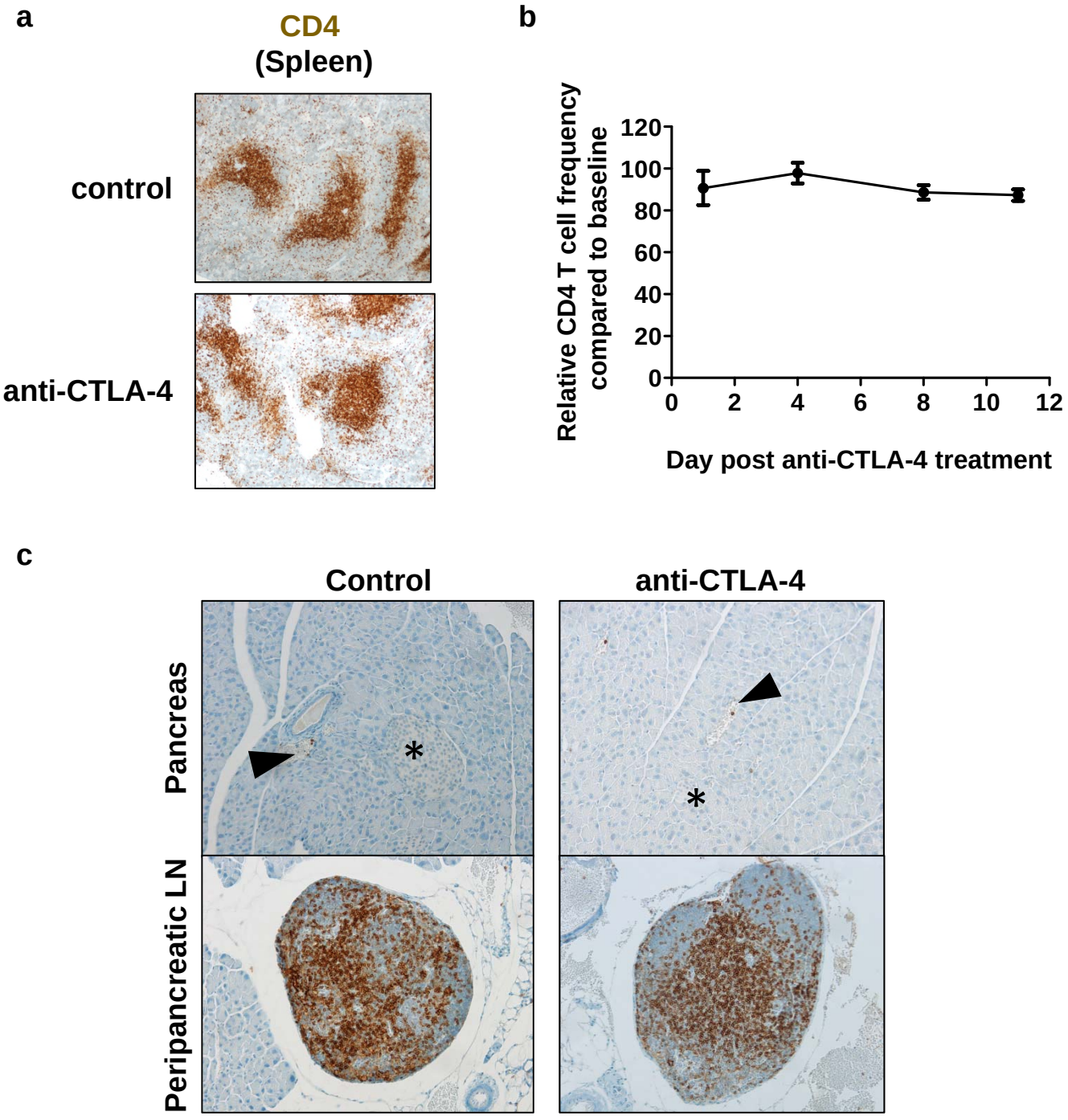
Supplementary Figure 3. CD25 antibodies do not alter CD8 T cell frequencies *in vivo*. **a,** CD25 expression by T cell subsets in peripheral blood of healthy PC and tumor-bearing KPC mice (*n* = 5 mice per group). **b,** CD8 T cell frequency in peripheral blood after treatment with anti-CD25 antibody PC61 (*n* = 5 mice per group). **c,** Representative images showing immunohistochemical staining for CD8 T cells in peri-tumoral lymph nodes detected adjacent to KPC tumors in mice at 14±2 days after treatment with isotype control or anti-CD25 antibodies. Statistical significance was determined by *t*-test. n.s., not significant.

Supplementary Figure 4



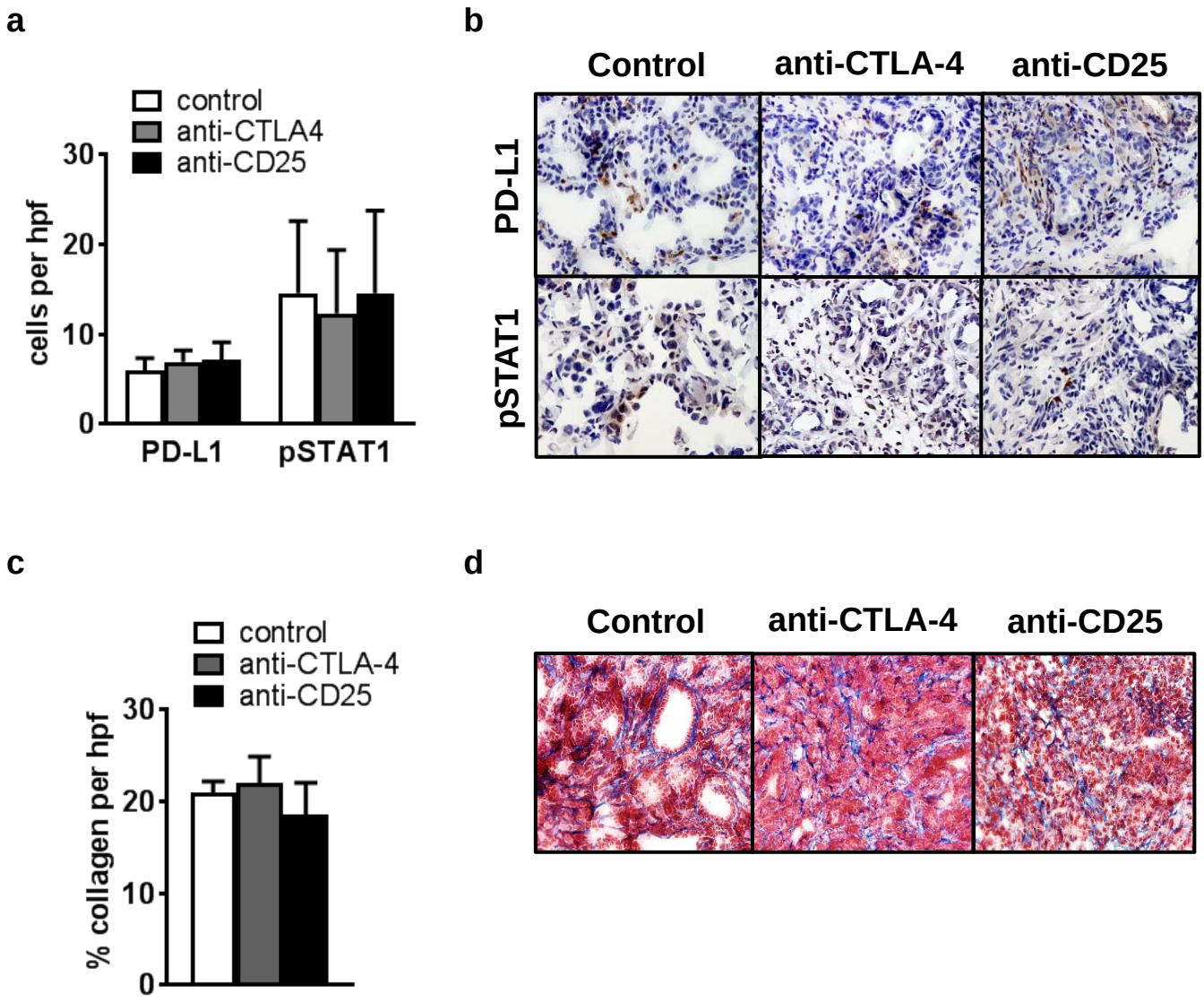
Supplementary Figure 4: Treatment with anti-CD25 antibodies does not alter CD4 T cell frequencies in healthy mice. PC mice were treated with isotype control or anti-CD25 (PC61) antibodies (*n*=4 mice per group). Peripheral blood was collected on days 1, 4, 8 and 11. Spleens and pancreata were harvested from mice at 14 days after treatment. **a,** Representative immunohistochemical images showing CD4+ cells in spleen tissue. **b,** CD4 T cell frequency in peripheral blood after treatment with anti-CD25 antibody. **c,** Representative immunohistochemical images showing CD4+ cells in pancreas (top) and peripancreatic lymph nodes (bottom). In the pancreas, CD4 T cells were rare and mainly confined to blood vessels (arrowhead) and peripancreatic lymph nodes without evidence of involvement of pancreatic islets (asterisks).

Supplementary Figure 5



Supplementary Figure 5: Treatment with anti-CTLA-4 antibodies does not alter CD4 T cell frequencies in healthy mice. PC mice were treated with isotype control or anti-CTLA-4 (9H10) antibodies (*n*=4 mice per group). Peripheral blood was collected on days 1, 4, 8 and 11. Spleens and pancreata were harvested from mice at 14 days after treatment. **a,** Representative immunohistochemical images to detect CD4+ cells in spleen tissue. **b,** CD4 T cell frequency in peripheral blood after treatment with anti-CTLA-4 antibody. **c,** Representative immunohistochemical images showing CD4+ cells in pancreas (top) and peripancreatic lymph nodes (bottom). In the pancreas, CD4 T cells were rare and mainly confined to blood vessels (arrowheads) and peripancreatic lymph nodes without evidence of involvement of pancreatic islets (asterisks).

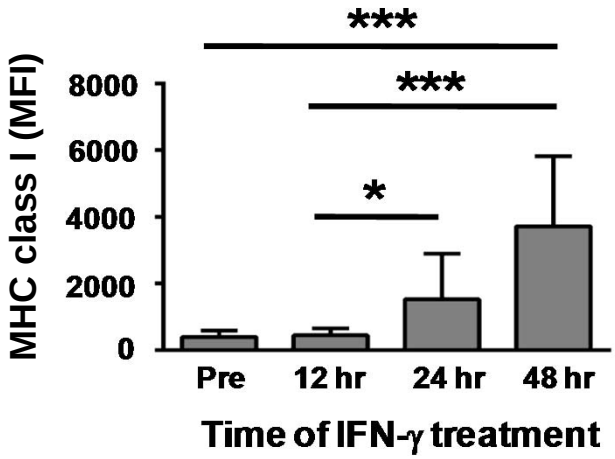
Supplementary Figure 6



Supplementary Figure 6: Treatment with anti-CTLA-4 or anti-CD25 antibodies does not alter PD-L1 expression, STAT1 activation or alter collagen-based fibrosis in PDAC tumors from KPC mice. KPC mice were treated with control, anti-CD25 (PC61), or anti-CTLA-4 (9H10) antibodies. Pancreatic tumors were harvested from mice at 14 days after treatment (*n*=4-6 mice per group). **a,** Quantification and **b,** representative immunohistochemical images to detect PD-L1 and phosphorylated STAT1 (pSTAT1) expression in tumor tissue. **c,** Quantification and **d,** representative images of Masson's Trichrome staining to detect collagen deposition. No significant changes in PD-L1, pSTAT1, or extracellular matrix were seen with treatment compared to control. Statistical significance was measured by one-way ANOVA with Tukey post-hoc test.

Supplementary Figure 7

a



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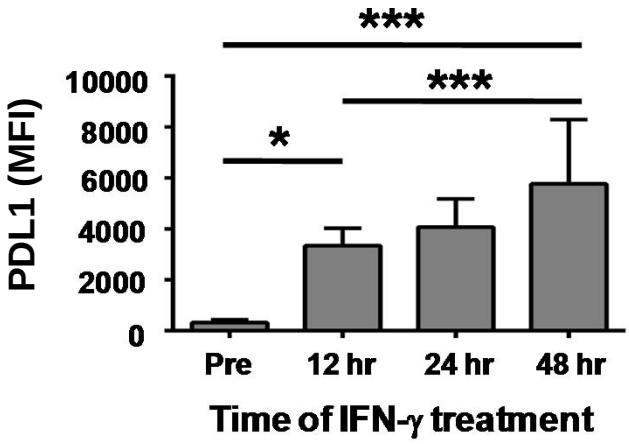

Supplementary Figure 7. KPC PDAC cell lines were treated *in vitro* with or without IFN-γ and analyzed by flow cytometry. Shown is the mean fluorescence intensity (MFI) for **a,** MHC class I and **b,** PDL1 molecules detected prior to treatment (Pre) and 12, 24, and 48 hours after IFN-γ treatment. Statistical significance was determined by one-way ANOVA with Tukey post-hoc test. *, $p<0.05$; ***, $p<0.001$.