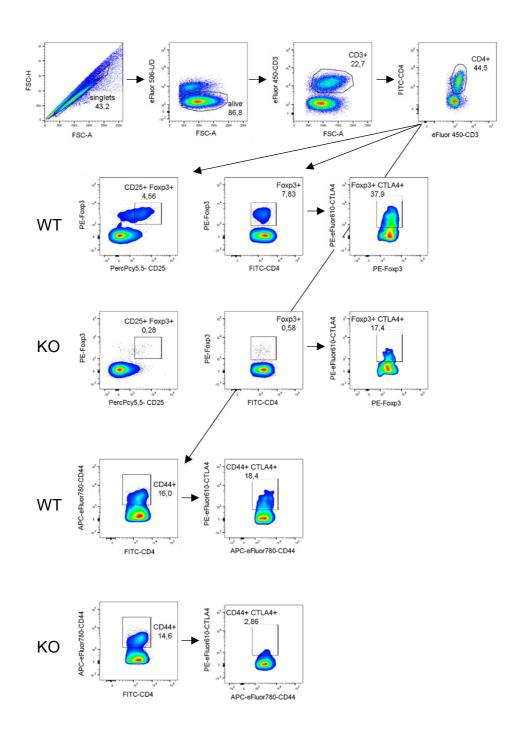


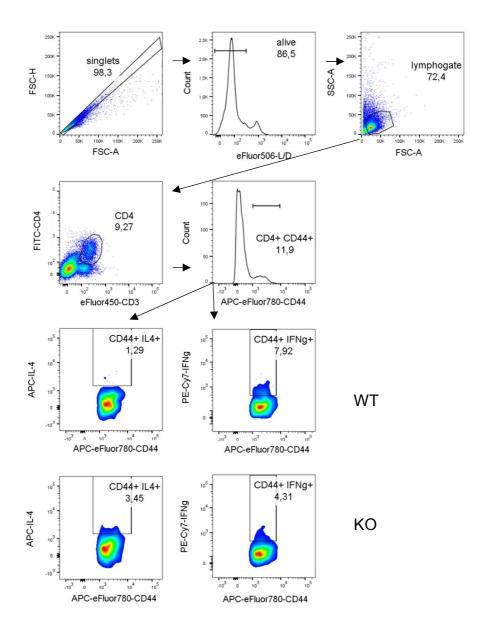
Supplementary Figure 1: Gating strategy for thymic Tregs (CD4⁺ CD8⁻ CD25⁺ FoxP3⁺) of *Malt1*-KO and WT mice

Singlets: FSC-A and FSC-H; cells: FSC-A and SSC-A; living cells: FSC-A and L/D eFluor506 negative; CD4⁺ CD8⁻ T cells: anti-CD4-FITC positive and anti-CD8-PE-Cy7 negative; Tregs (Foxp3⁺CD25⁺CD4⁺ T cells): anti-Foxp3-PE and anti-CD25-PercPcy5.5 positive.



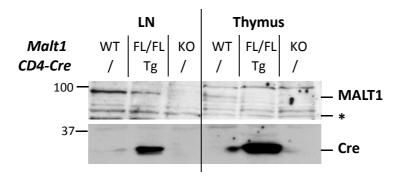
Supplementary Figure 2: Gating strategy for CTLA4⁺ Foxp3⁺ CD4⁺ T cells and CTLA-4⁺ CD44⁺ CD4⁺ T cells of *Malt1*-KO and WT mice

Singlets: FSC-A and FSC-H; cells: FSC-A and SSC-A; living cells: FSC-A and L/D eFluor506 negative; CD4⁺ T cells: anti-CD3-eFluor and anti-CD4-FITC positive; Tregs (Foxp3⁺ CD25⁺ CD4⁺ T cells): anti-Foxp3-PE and anti-CD25-PercPcy5.5 positive; surface CTLA4⁺ Tregs: anti-Foxp3-PE and anti-CTLA4-PEeFluor610; surface CTLA4⁺ CD44⁺ CD4⁺ T cells: anti-CD44-APC-eFluor780 and anti-CTLA4-PEeFluor610 positive.



Supplementary Figure 3: Gating strategy for Th1 and Th2 cells of *Malt1*-KO and WT mice

Singlets: FSC-A and FSC-H; living cells: histogram L/D eFluor506 negative; cells (lymphogate): FSC-A and SSC-A; CD4 $^+$ T cells: anti-CD3-eFluor and anti-CD4-FITC positive; CD44 $^+$ CD4 $^+$ T cells: histogram anti-CD44-APCeFluor780 positive; Th1 cells: anti-CD44-APCeFluor and anti-IFN- γ -PEcy7 positive; Th2 cells: anti-CD44-APCeFluor and anti-IL-4-APC positive.



Supplementary Figure 4: Western blot showing *CD4-Cre* specific MALT1 loss in lymph nodes and thymus

Thymus (T cells) or lymph nodes (LN; mainly T cells and B cells) from *Malt1*-WT, *Malt1*^{FL/FL} *CD4*-*Cre*^{Tg/+} and *Malt1*-KO mice were lysed in E1A buffer and equal amounts of protein (100μg) were run on SDS-PAGE and blotted. The western blot was developed with an anti-MALT1 antibody recognizing murine MALT1 (top panel) and an anti-Cre recombinase antibody (lower panel). Aspecific band (*) was used as loading control.