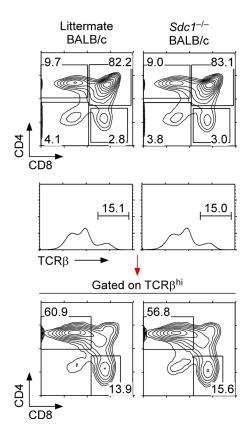


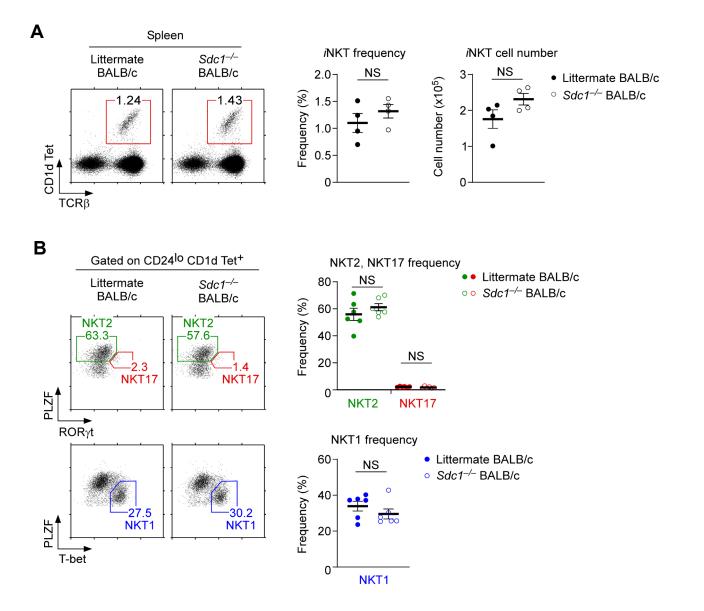
Supplementary Figure 1. CD138 expression in BALB/c thymocytes

- (A) The graph shows the frequency of CD138⁺ $\gamma\delta$ T cells among total thymic $\gamma\delta$ T cells of BALB/c mice. Data are summary of 3 independent experiments with a total of 8 BALB/c mice.
- (**B**)The graph shows the frequencies of CD138⁺ iNKT cells among total thymic iNKT cells of BALB/c mice. Data are summary of 4 independent experiments with a total of 9 BALB/c mice.
- **(C)** The graphs show frequencies and cell numbers of *i*NKT cells in *Rorc*^{-/-} and WT littermate BALB/c thymocytes. Results are the summary of 3 independent experiments with a total of 5 *Rorc*^{-/-} and 4 WT littermate mice.
- **(D)** Thymic $\gamma\delta$ T cell numbers in $Rorc^{-/-}$ and WT littermate BALB/c mice. Results are summary of 3 independent experiments with a total of 5 $Rorc^{-/-}$ and 4 WT littermate mice.
- (E) The graph shows the frequency of CD138⁺ $\gamma\delta$ T cells among total thymic $\gamma\delta$ T cells of $Rorc^{-/-}$ and WT littermate BALB/c mice. Results are summary of 3 independent experiments with a total of 5 $Rorc^{-/-}$ and 4 WT littermate mice.



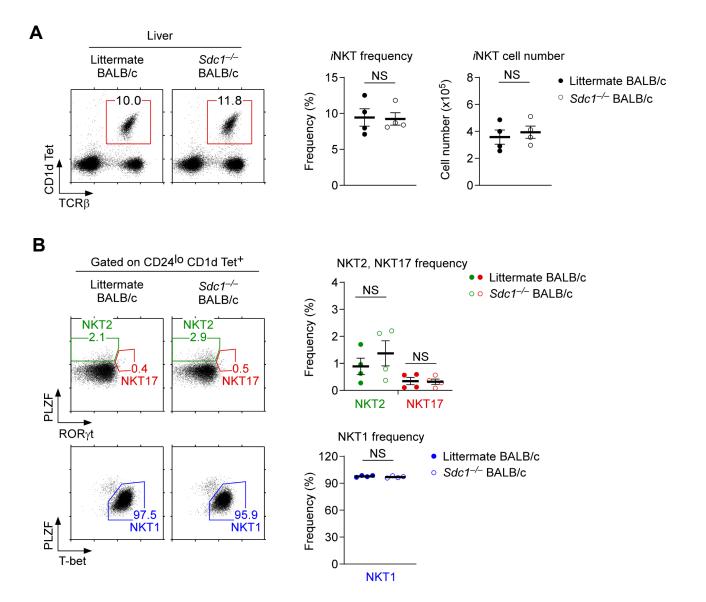
Supplementary Figure 2. T cell development in Sdc1-/- BALB/c mice

Contour plots show CD8 versus CD4 profiles of total (top) and TCR β^{hi} -gated (bottom) $Sdc1^{-/-}$ and WT littermate BALB/c thymocytes. The results are representative of 6 independent experiments with a total of $10\ Sdc1^{-/-}$ and $10\ WT$ littermate BALB/c mice.



Supplementary Figure 3. Splenic iNKT cells in Sdc1-/- BALB/c mice

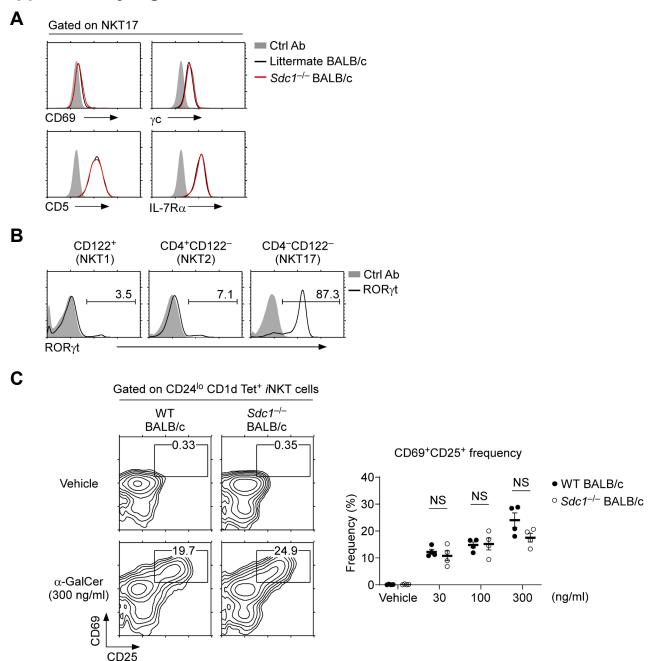
- (A) Identification of *i*NKT cells among B cell-depleted splenocytes of $Sdc1^{-/-}$ and WT littermate BALB/c mice. The dot plots are representative (left), and the *i*NKT frequency and number graphs (right) show the summary of 2 independent experiments with a total of $4 Sdc1^{-/-}$ and 4 WT littermate BALB/c mice. Numbers in the box show frequencies of *i*NKT cells among B cell-depleted splenocytes.
- **(B)** Subset distribution in $Sdc1^{-/-}$ and WT littermate BALB/c splenic *i*NKT cells. The frequencies of NKT1, NKT2, and NKT17 cells were determined by T-bet versus PLZF and ROR γ t versus PLZF expression. The dot plots are representative (left), and the graphs show the summary (right) of 3 independent experiments with a total of 6 $Sdc1^{-/-}$ and 6 WT littermate BALB/c mice.



Supplementary Figure 4. Liver iNKT cells in Sdc1-/- BALB/c mice

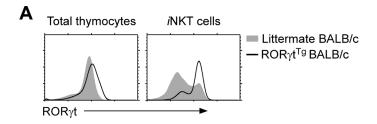
- (A) Identification of *i*NKT cells in the liver of $Sdc1^{-/-}$ and WT littermate BALB/c mice. The dot plots are representative (left), and the *i*NKT frequency and number graphs (right) show the summary of 2 independent experiments with a total of $4 Sdc1^{-/-}$ and 4 WT littermate BALB/c mice. Numbers in the box show frequencies of *i*NKT cells among total CD45⁺ liver lymphocytes.
- **(B)** *i*NKT subset distribution in $Sdc1^{-/-}$ and WT littermate BALB/c liver cells. The frequencies of NKT1, NKT2, and NKT17 cells were determined by T-bet versus PLZF and ROR γ t versus PLZF expression. The dot plots are representative (left), and the graphs show the summary (right) of 2 independent experiments with a total of 4 $Sdc1^{-/-}$ and 4 WT littermate BALB/c mice.

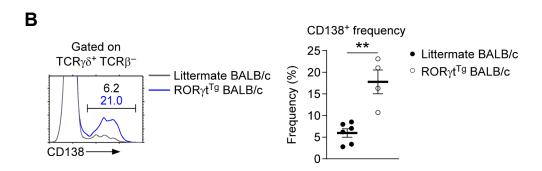
BALB/c mice.

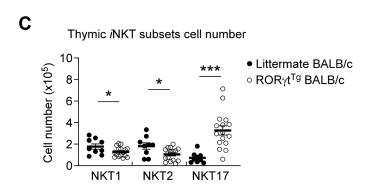


Supplementary Figure 5. Characterization of thymic *i*NKT subsets in $Sdc1^{-/-}$ BALB/c mice (A) CD69, CD5, γ c and IL-7R α expression were assessed on NKT17 cells of $Sdc1^{-/-}$ and WT littermate BALB/c thymocytes. The histograms are representative of 2 independent experiments with a total of 2 $Sdc1^{-/-}$ and 2 WT littermate BALB/c mice.

(B) ROR γ t expression in thymic *i*NKT subsets as identified by CD122 and CD4 staining. The histograms are representative of 2 independent experiments with a total of 3 BALB/c mice. (C) Surface CD69 and CD25 expression was assessed on mature *i*NKT cells of $Sdc1^{-/-}$ and WT littermate BALB/c thymocytes upon overnight *in vitro* stimulation with the indicated amounts of α -GalCer. Contour plots are representative, and the graph show the summary of 3 independent experiments. Results are summary of 3 independent experiments with a total of 4 $Sdc1^{-/-}$ and 4 WT

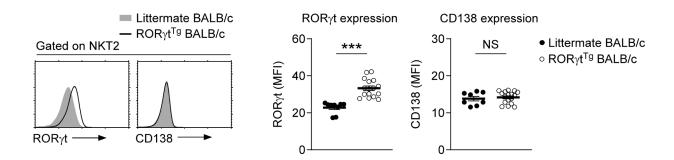






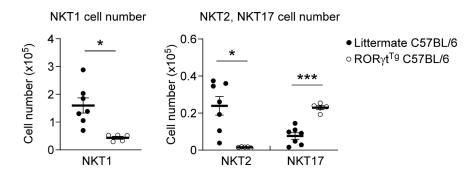
Supplementary Figure 6. iNKT and γδ T cell analysis in RORγt^{Tg} BALB/c thymocytes

- (A) The intracellular ROR γ t contents in total thymocytes and total *i*NKT cells were assessed in ROR γ t^{Tg} and WT littermate BALB/c mice. The histograms are representative of 2 independent experiments with a total of 5 ROR γ t^{Tg} and 3 WT littermate BALB/c mice.
- **(B)** CD138 expression on thymic $\gamma\delta$ T cells of ROR γ t^{Tg} BALB/c thymocytes. The histogram (left) identifies CD138⁺ $\gamma\delta$ T cells, and the graph (right) summarizes the frequency of CD138⁺ $\gamma\delta$ T cells. The results are representative of 2 independent experiments with a total of 4 ROR γ t^{Tg} and 6 littermate WT BALB/c mice.
- (C) Thymic NKT1, NKT2 and NKT17 cell numbers were determined in RORγt^{Tg} and WT littermate BALB/c mice. The results show summary of 4 independent experiments with a total of 16 RORγt^{Tg} and 9 WT littermate BALB/c mice.



Supplementary Figure 7. ROR γ t and CD138 expression in NKT2 cells of ROR γ t BALB/c thymocytes

The histograms show ROR γ t and CD138 expression in thymic NKT2 cells (left). The graphs show the MFI of ROR γ t and CD138 in NKT2 cells of the indicated mice (right). The results show summary of 4 independent experiments with a total of 16 ROR γ t^{Tg} and 9 WT littermate BALB/c mice.



Supplementary Figure 8. Characterization of thymic *i*NKT subsets in RORγt^{Tg} C57BL/6 mice Thymic NKT1, NKT2 and NKT17 cell numbers were determined in RORγt^{Tg} and WT littermate C57BL/6 mice. The results show summary of 3 independent experiments with a total of 5 RORγt^{Tg} and 7 WT littermate C57BL/6 mice.