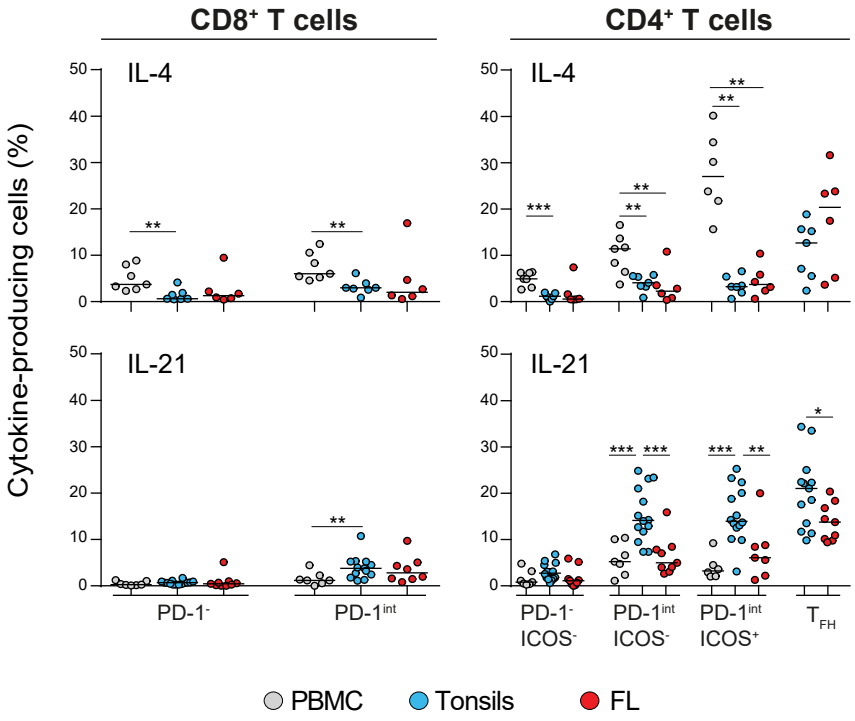
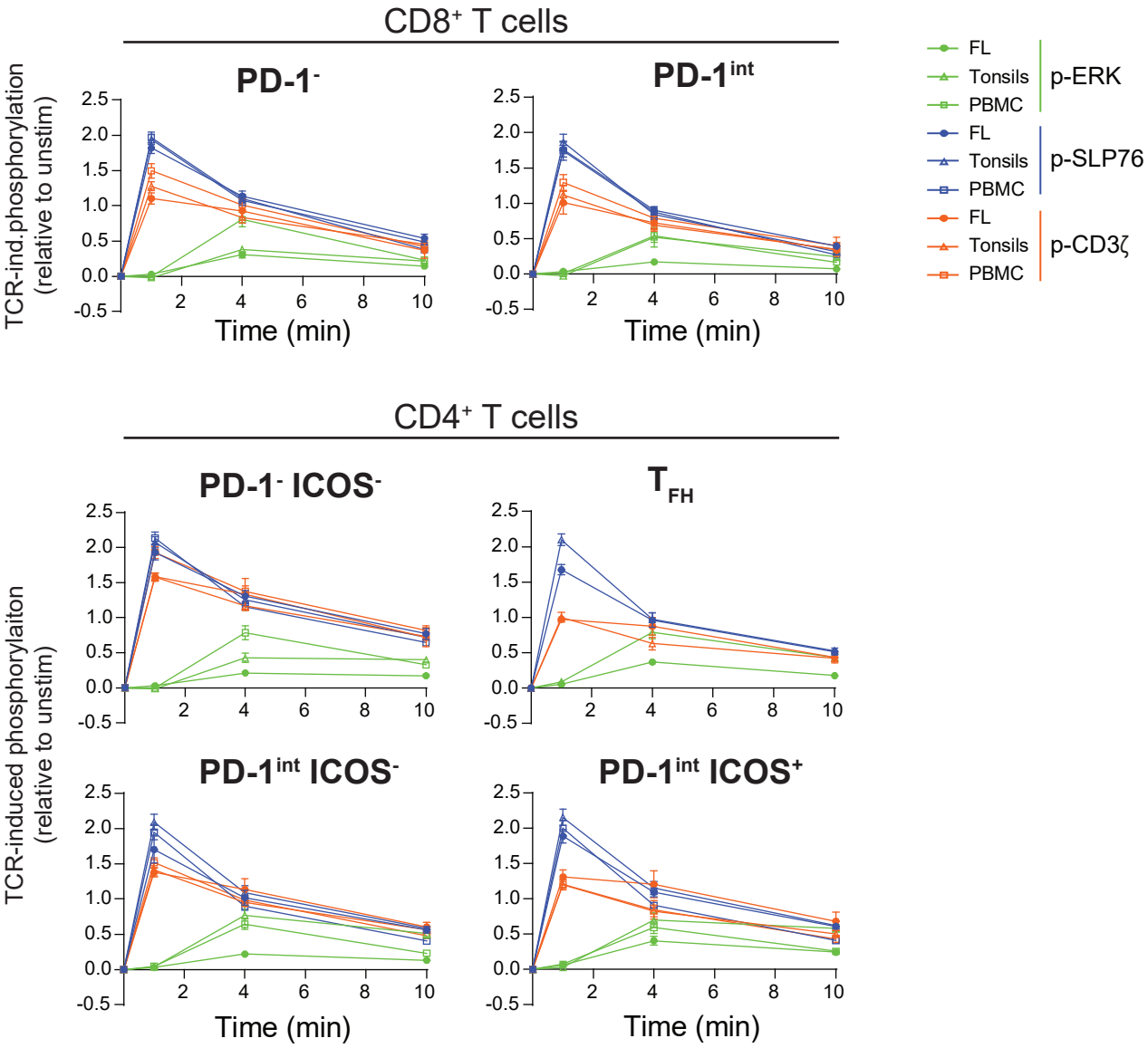
Supplementary Figure 1



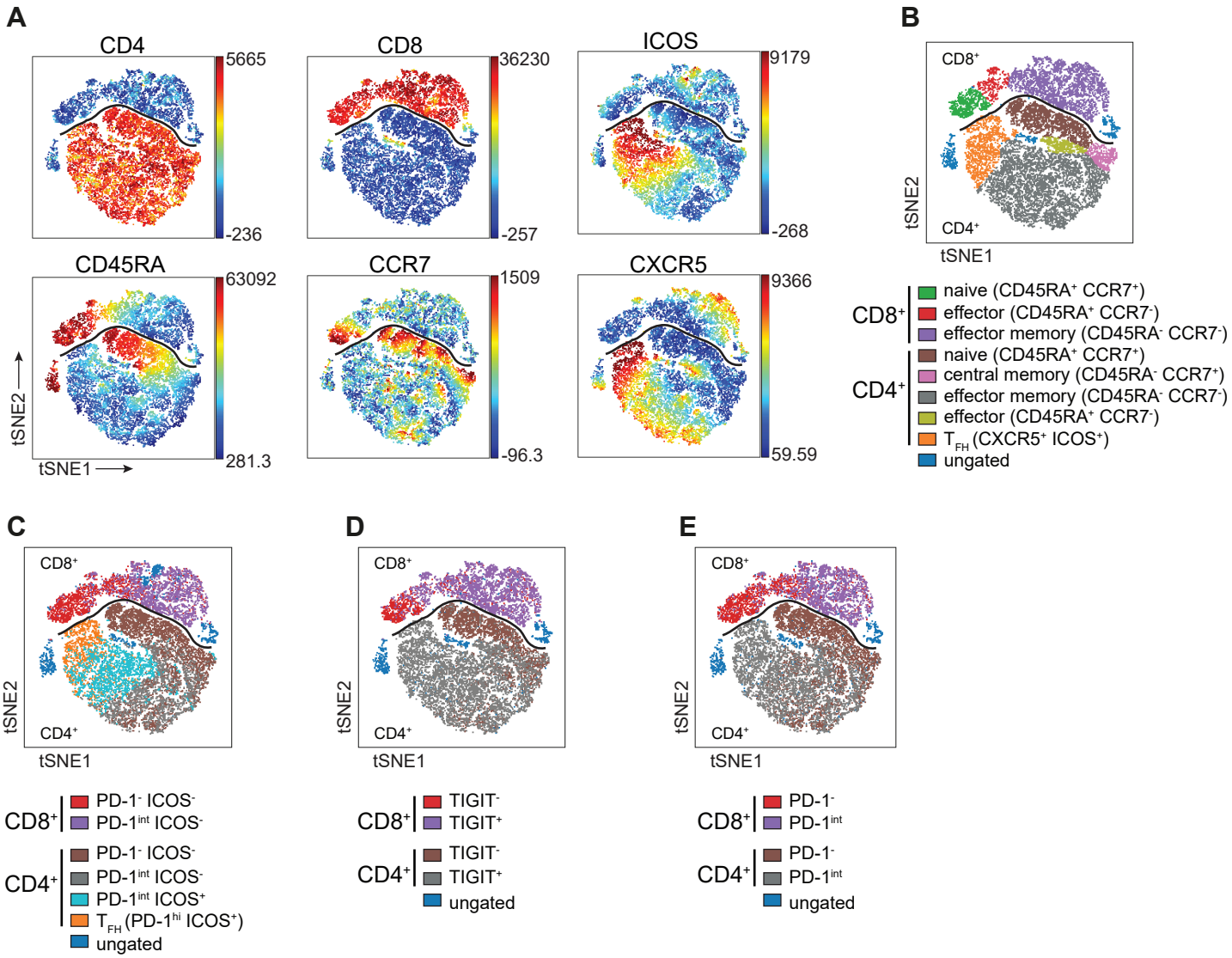
Supplementary Figure 1. IL-21 production is reduced in CD4 FL T cells. Single cell suspensions from FL LN and healthy donors (tonsils and PBMC) were cultured with or without PMA and ionomycin for a total of 6 hours, with GolgiPlug present for the last 4 hours. Levels of intracellular IL-4 and IL-21 were measured by fluorescence flow cytometry. Each data point represents a single donor. Statistical differences were calculated using Mann-Whitney non-parametric test; *$p < 0.05$, **$p < 0.01$, ***$p < 0.001$.

Supplementary Figure 2



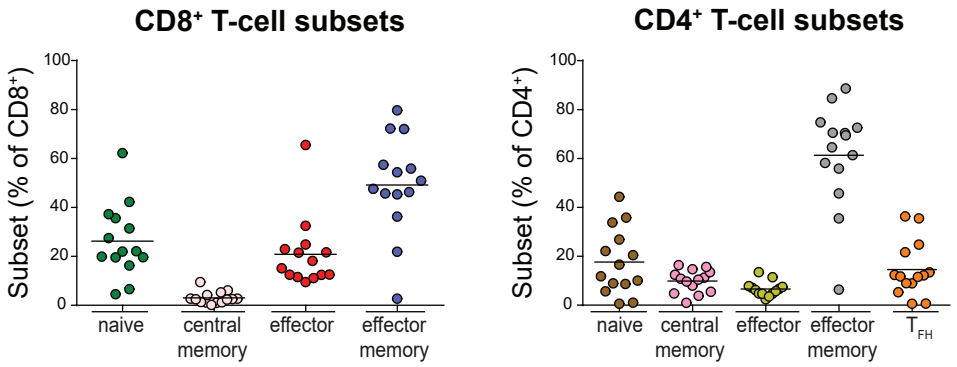
Supplementary Figure 2. TCR-induced signaling effectors have different kinetic. Single cell suspensions from FL LN (*n* = 9), tonsils (*n* = 11) and PBMC (*n* = 9) were cultured with or without α-CD3+α-CD28 antibodies for 2 minutes, followed by avidin crosslinking for 1, 4 or 10 minutes and then assayed for TCR-induced phosphorylation of CD3ζ, SLP76 and ERK using phospho-flow cytometry. Diagrams show kinetic of the three signaling effectors in CD8 and CD4 T-cell subsets as median fold change (FC) induction relative to unstimulated cells. Mean FC ± SEM.

Supplementary Figure 3



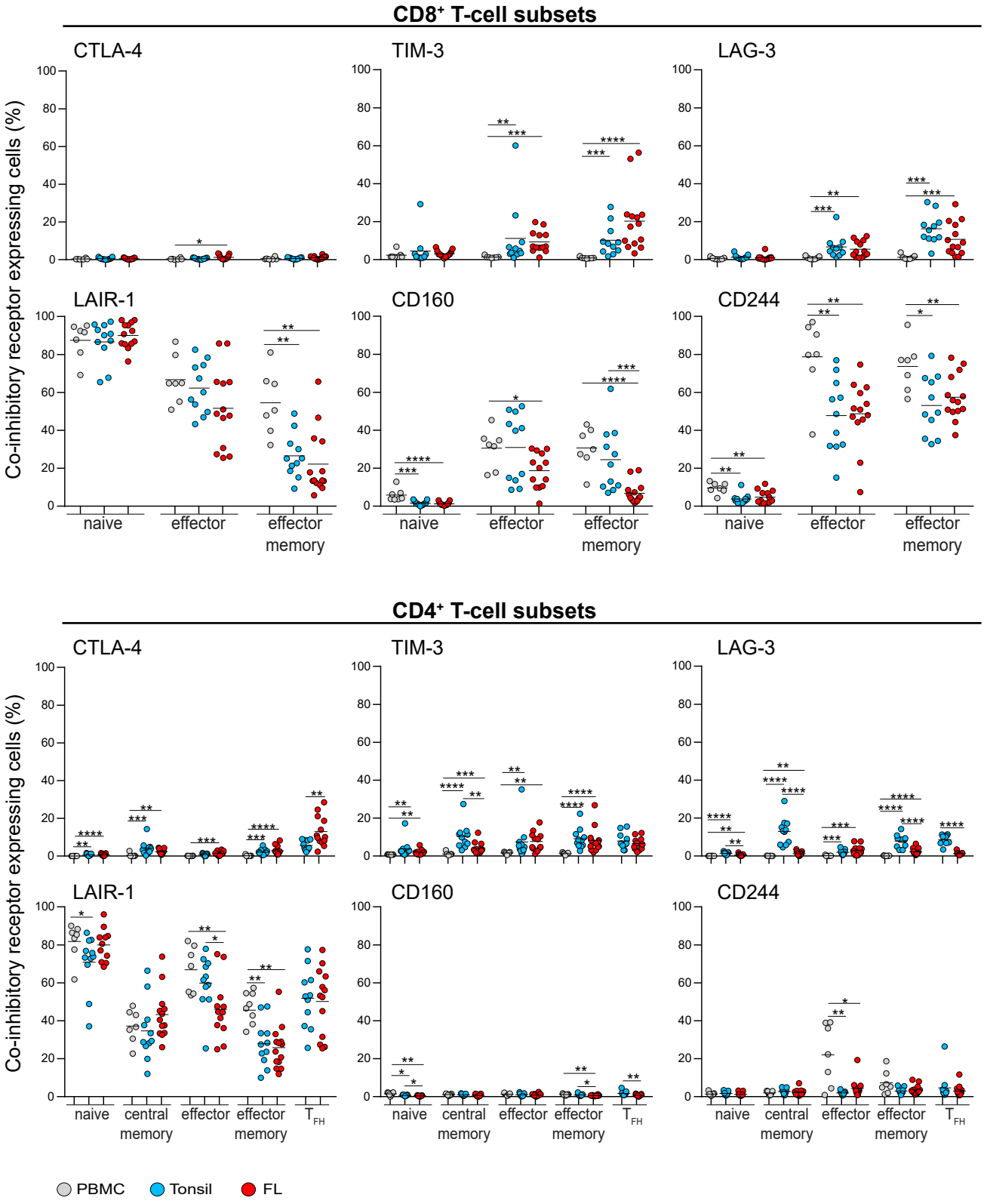
Supplementary Figure 3. Identification of T-cell subsets. Single cell suspensions from FL were analyzed by fluorescence flow cytometry. A viSNE analysis was performed on the CD3$^+$ cells, based on expression of CD4, CD8, CXCR5, ICOS, CD45RA and CCR7. Figure shows viSNE map of 4 FL samples combined. The manually added line in the viSNE plots marks the distinction between CD8 and CD4 T cells. Color represents fluorescence intensity with separate scale for each marker, with max set to highest measured signal. **(A)** Intensity of markers included in viSNE analysis and used for gating of conventional T-cell subsets (corresponding to subsets used in Figure 4). **(B)** Each gated T-cell subset, identified in (A), is shown with a separate color. **(C)** T-cell subsets were gated on biaxial plots based on expression of PD-1 and ICOS as in Figure 1, and shown on the viSNE map. **(D)** T cells were gated on biaxial plots based on expression of CD4/CD8 and TIGIT and shown on viSNE map. **(E)** T cells were gated on biaxial plots based on expression of CD4/CD8 and PD-1 and shown on viSNE map.

Supplementary Figure 4



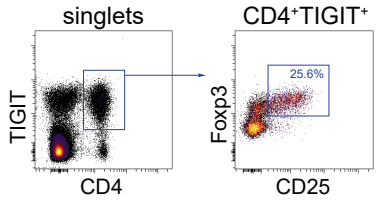
Supplementary Figure 4. Distribution of T-cell subsets in FL LN. Single cell suspensions from FL LN (*n* = 14) were analyzed by fluorescence flow cytometry. Conventional subsets of naive T cells, T central memory cells, T effector cells, T effector memory cells and T follicular helper cells (T_{FH}) were gated on biaxial plots, based on differential expression of CD4, CD8, CXCR5, ICOS, CD45RA and CCR7. Each data point represents a single donor.

Supplementary Figure 5



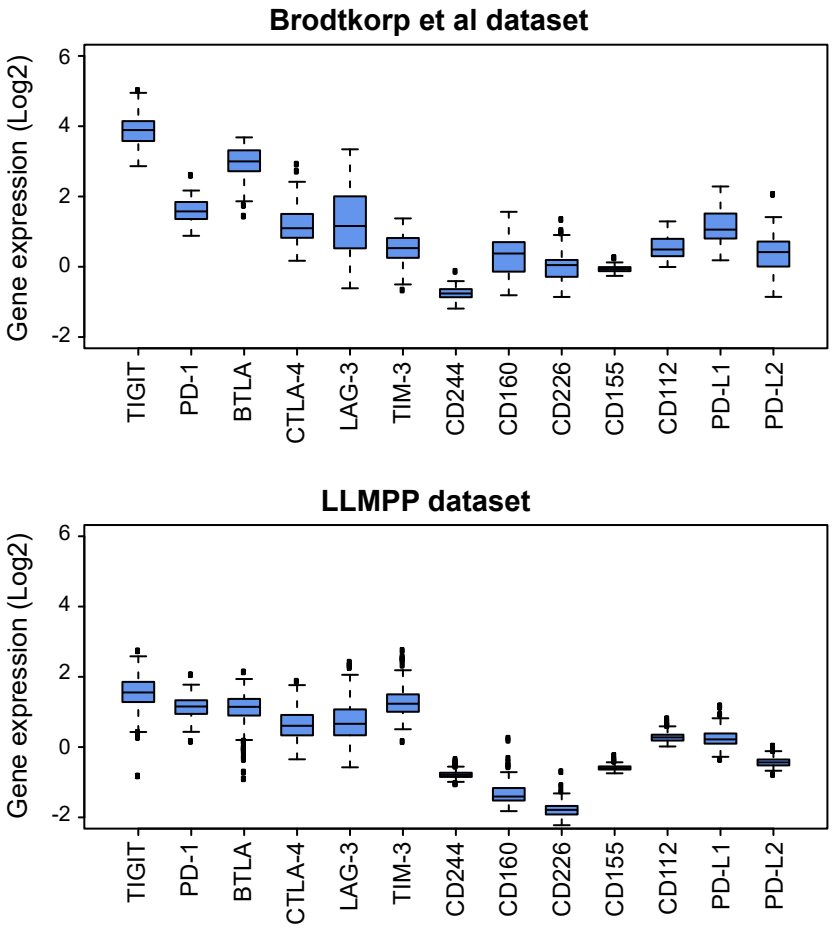
Supplementary Figure 5. Expression patterns of co-inhibitory receptors in CD8 and CD4 T-cell subsets. Co-Inhibitory receptor expression was analyzed by 11-parameter flow cytometry in single cell suspensions from FL LN and non-tumoral controls (tonsils and PBMC). Each data point represents a single donor. Statistical differences were calculated using Mann-Whitney non-parametric test *$p < 0.05$, **$p < 0.01$, ***$p < 0.001$, ****$p < 0.0001$.

Supplementary Figure 6



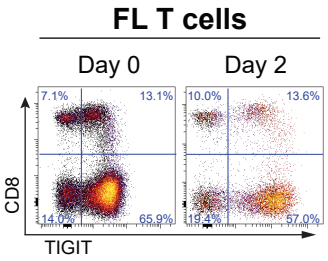
Supplementary Figure 6. Contribution of Tregs among CD4⁺TIGIT⁺ T cells
Single cell suspensions were analyzed by flow cytometry. Plots show one representative FL sample.

Supplementary Figure 7



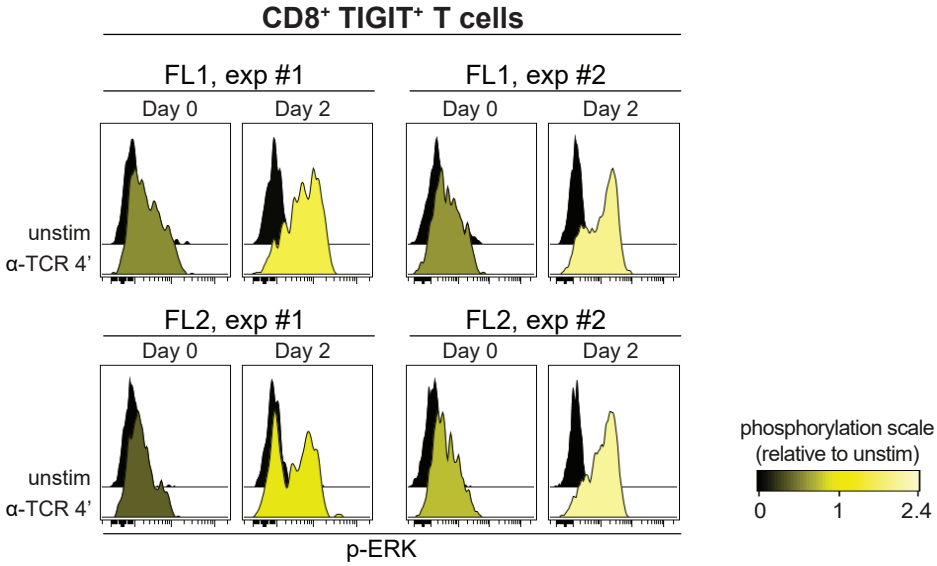
Supplementary Figure 7. TIGIT is a highly expressed co-inhibitory receptor in FL. Boxplots represent gene expression in 44 pre-treatment FL biopsies (Brodtkorb et al.) (upper panel), and 194 pre-treatment FL biopsies (Dave et al.) (lower panel). Values are median-centered across patients.

Supplementary Figure 8



Supplementary Figure 8. TIGIT expression is stable over time.
Single cell suspensions from FL LN were analyzed by flow cytometry. TIGIT expression was measured at day 0 and after 48 h *in vitro* culture.

Supplementary Figure 9



Supplementary Figure 9. Recovery of TCR-induced signaling in TIGIT$^+$ CD8$^+$ FL T cells is robust. Single cell suspensions from FL LN were assayed for TCR-induced signaling and analyzed by phospho-flow cytometry at day 0 and after 48 hour *in vitro* culture. Signaling was induced using α-CD3+α-CD28 antibodies for 2 minutes, followed by avidin crosslinking for 4 minutes. The same FL specimens (n = 2) were analyzed in two separate experiments. The histograms show median fold change (FC) induction relative to unstimulated cells, using arcsinh transformed data.