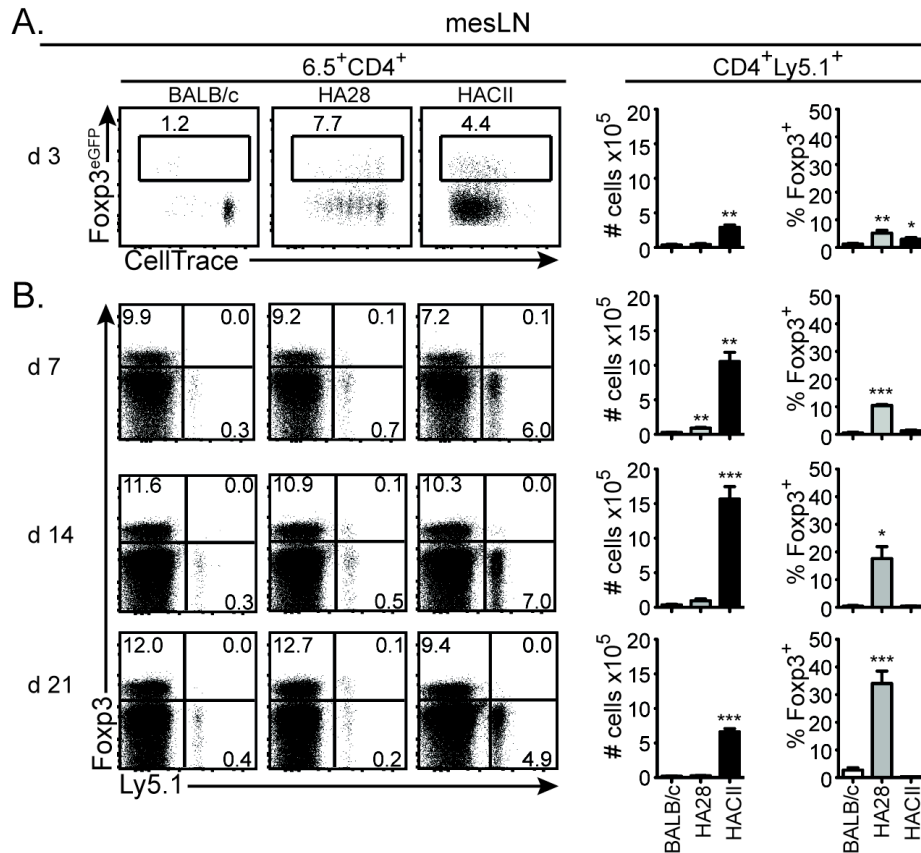


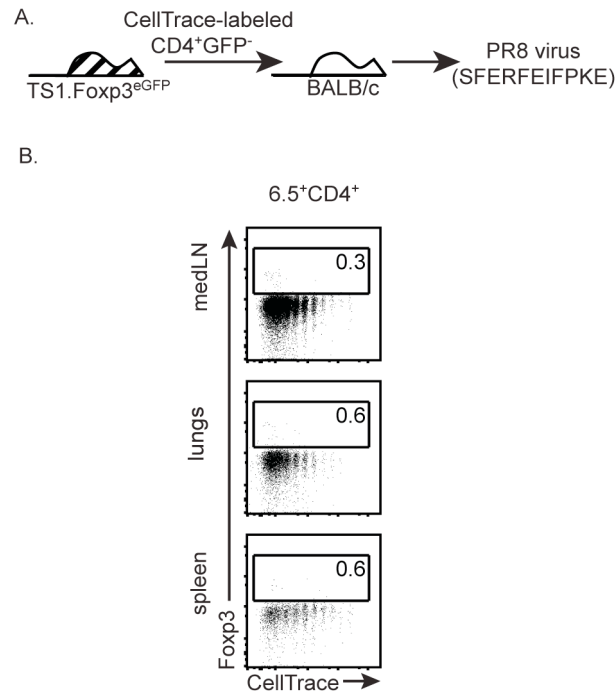
Supplemental Figure 1.



pTreg formation occurs with similar efficiency in the mesLNs as the pLNs of HA28 and HACII mice. Upper panels show dot plots of eGFP versus Cell Trace Violet levels on 6.5⁺CD4⁺ mesLN cells isolated from BALB/c, HA28 and HACII mice 3 days post-transfer of CellTrace Violet-labeled 6.5⁺CD4⁺CD25⁻eGFP⁻ cells from TS1.Foxp3^{eGFP}.Ly5.1 mice. Lower panels show dot plots of Fxp3 versus Ly5.1 expression by CD4⁺ pLN cells 7 days post-transfer of CellTrace Violet-labeled 6.5⁺CD4⁺CD25⁻eGFP⁻ cells from TS1.Foxp3^{eGFP}.Ly5.1 mice into BALB/c, HA28 and HACII mice, and 14 or 21 days post-transfer into BALB/c.Foxp3^{eGFP}, HA28.Foxp3^{eGFP}, and HACII.Foxp3^{eGFP} mice. Graphs show mean numbers or percentages ±SEM of indicated cell types (n=4-8 for each recipient from at least

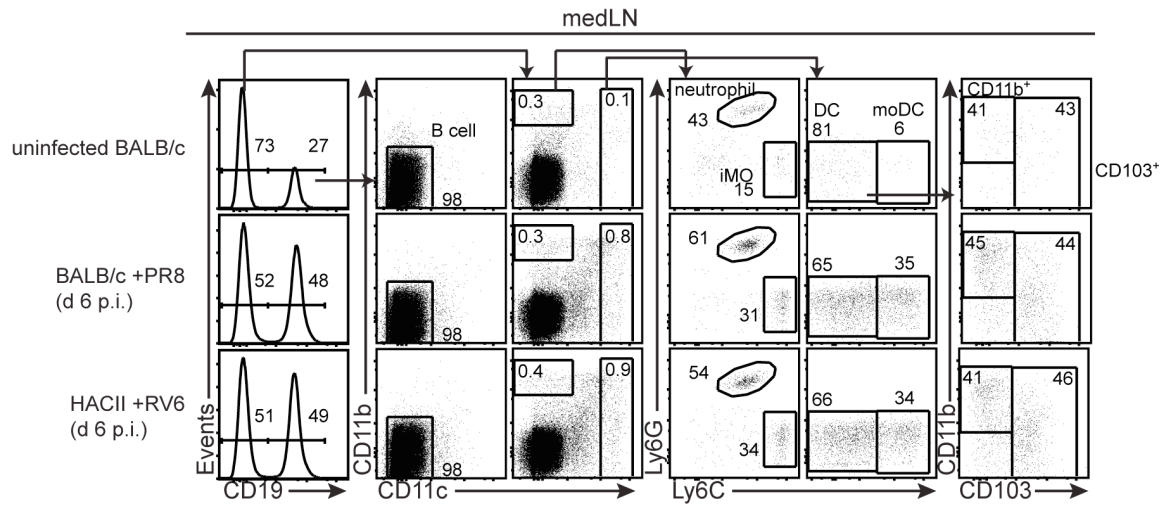
2 independent experiments at each time point). Percentages of cells in indicated gates are shown.

Supplemental Figure 2.



Recognition of PR8 viral-HA does not induce 6.5⁺CD4⁺Foxp3⁻ cells to upregulate Foxp3. **(A)** Schematic denotes transfer of CD4⁺CD25⁻eGFP⁻ cells isolated from TS1.Foxp3^{eGFP} mice into BALB/c mice followed 24 hours later by PR8 infection and analysis at d 5 p.i.. **(B)** Dot plots show Foxp3 versus CellTrace Violet staining by 6.5⁺CD4⁺ cells obtained from medLNs, lungs or spleens 5 d p.i with PR8 virus, and percentages of cells in indicated gates are shown. Panels are representative of 3 independent experiments with 1-2 mice of each strain/experiment.

Supplemental Figure 3.



Gating strategy used to identify different APC populations. Histograms and dot plots show staining for indicated molecules by cells isolated from the medLNs of uninfected BALB/c mice, of BALB/c mice d 6 p.i. with PR8 virus, and of HACII mice d 6 p.i. with RV6 virus. Arrows show gates used to identify and then further fractionate cell populations as indicated. B cells are identified as CD19⁺CD11b⁻CD11c⁻ cells; DCs are identified as CD19⁻CD11c⁺Ly6C⁻Ly6G⁻ cells, and further differentiated as being CD103⁺ or CD103⁻CD11b⁺; moDCs are identified as CD19⁻CD11c⁺Ly6C⁺Ly6G⁻ cells; neutrophils are identified as CD19⁻CD11b⁺CD11c⁻Ly6C^{low}Ly6G⁺ cells; iMOs are identified as CD19⁻CD11b⁺CD11c⁻Ly6C^{high}Ly6G⁻ cells.