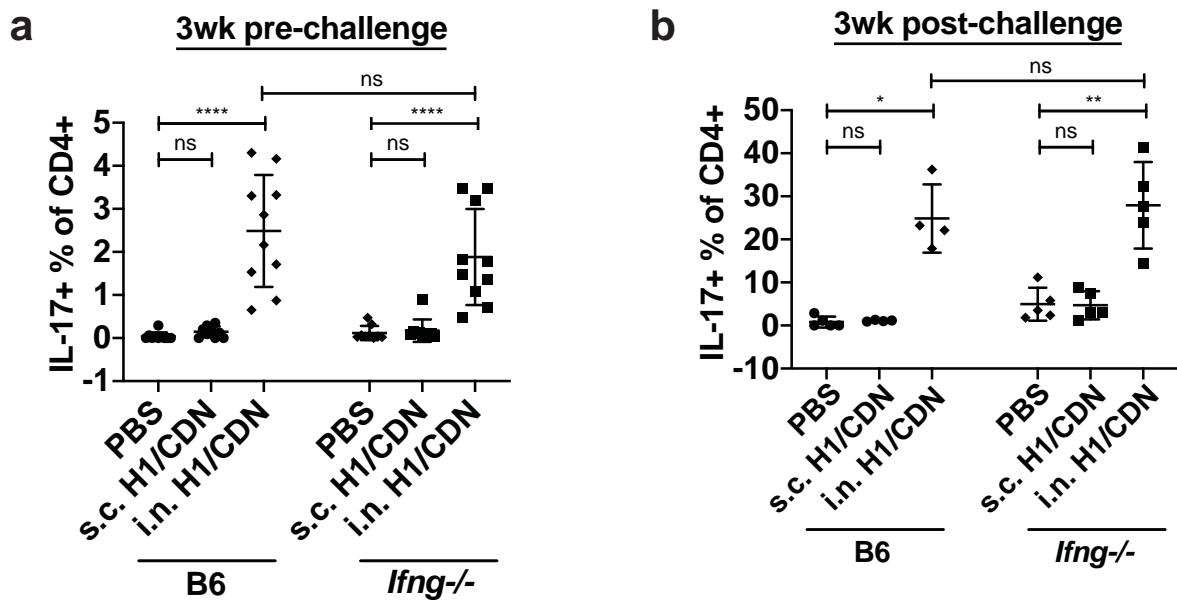
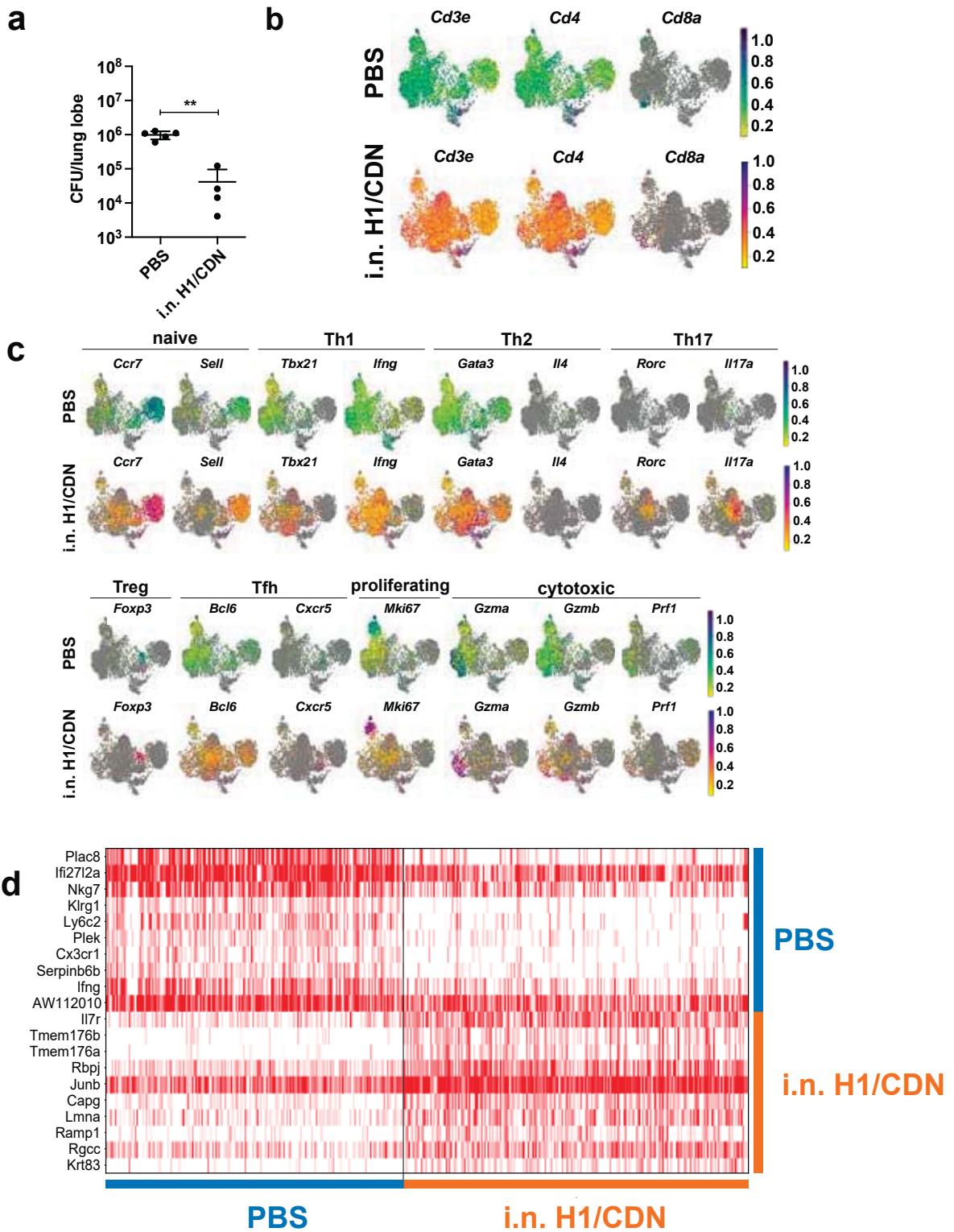


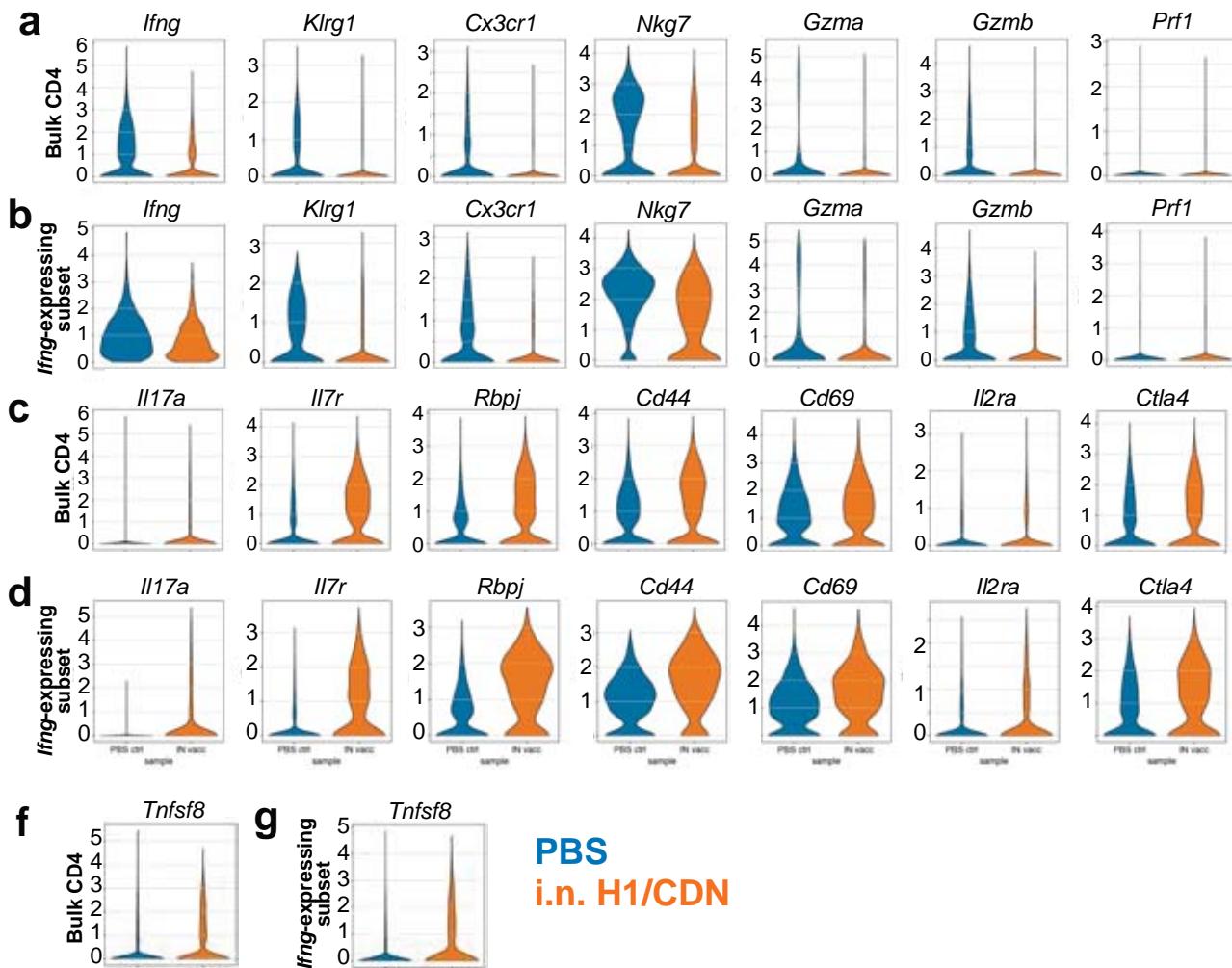
**Figure S1. Semi-quantitative histopathological analysis of vaccinated mouse lungs.** Formalin-fixed paraffin-embedded sections of mouse lungs 4 weeks post *M. tuberculosis* challenge were stained with H&E and assessed using a semi-quantitative scoring method for (a) inflammation area, (b, g) lymphoid follicle, lymphoid nodule, or granuloma presence, and (d-f) localized inflammation. Data are expressed as mean ( $\pm$  SD) of eight to ten mice per group from two independent experiments. Kruskal Wallis test followed by Dunn multiple comparison posthoc p-values; \* $p \leq 0.05$ , \*\* $p \leq 0.001$ , \*\*\* $p \leq 0.0001$ .



**Figure S2. Evaluation of T cells in the lungs of WT and *Ifng*<sup>-/-</sup> mice.** CD4 T cells in the lungs of mice at the indicated timepoints were evaluated for IL-17 production using ICS and flow cytometry. The mice depicted are the same as those depicted in Figure 3a and 3b. Data are expressed as mean ( $\pm$  SD) of eight to ten mice per group from two independent experiments with value from each individual animal displayed (a) or as a representative experiment with each animal displayed (b). Mann-Whitney t test p values; \* $p \leq 0.05$ , \*\* $p \leq 0.01$ , \*\*\* $p \leq 0.001$ .



**Figure S3.** (a) CFU from the scRNA-seq experiment at 4 weeks post challenge. Subsets of CD4 T cells were manually annotated based on signature expression of lineage-specific transcription factor, cytokine, and cell surface markers. (a) UMAP plots of CD4 T cell subsets isolated from mock (PBS) or (b) i.n. H1/ML-RR-cGAMP (i.n. H1/CDN) vaccinated mice 3 weeks post infection. (c) Signature gene expression of CD4 T cell subtypes in mock and i.n. H1/CDN vaccinated mice. The color scale indicates the percentile expression level for each cell that has non-zero expression. (d) heat map depicting genes that are most differentially expressed in mock and i.n. H1/CDN vaccinated mice.



**Figure S4.** Markers of Th1 lineage, terminal differentiation, and cytotoxicity in (a) bulk and (b) *Ifng*-expressing CD4 T cell populations in mock (blue, PBS) and i.n. H1/ML-RR-cGAMP vaccinated (orange, i.n. H1/CDN) infected mouse lungs. Markers of Th17 lineage, memory, and activation in (c) bulk and (d) *Ifng*-expressing CD4 populations. *Tnfsf8* expression in (f) bulk and (g) *Ifng*-expressing CD4 populations.