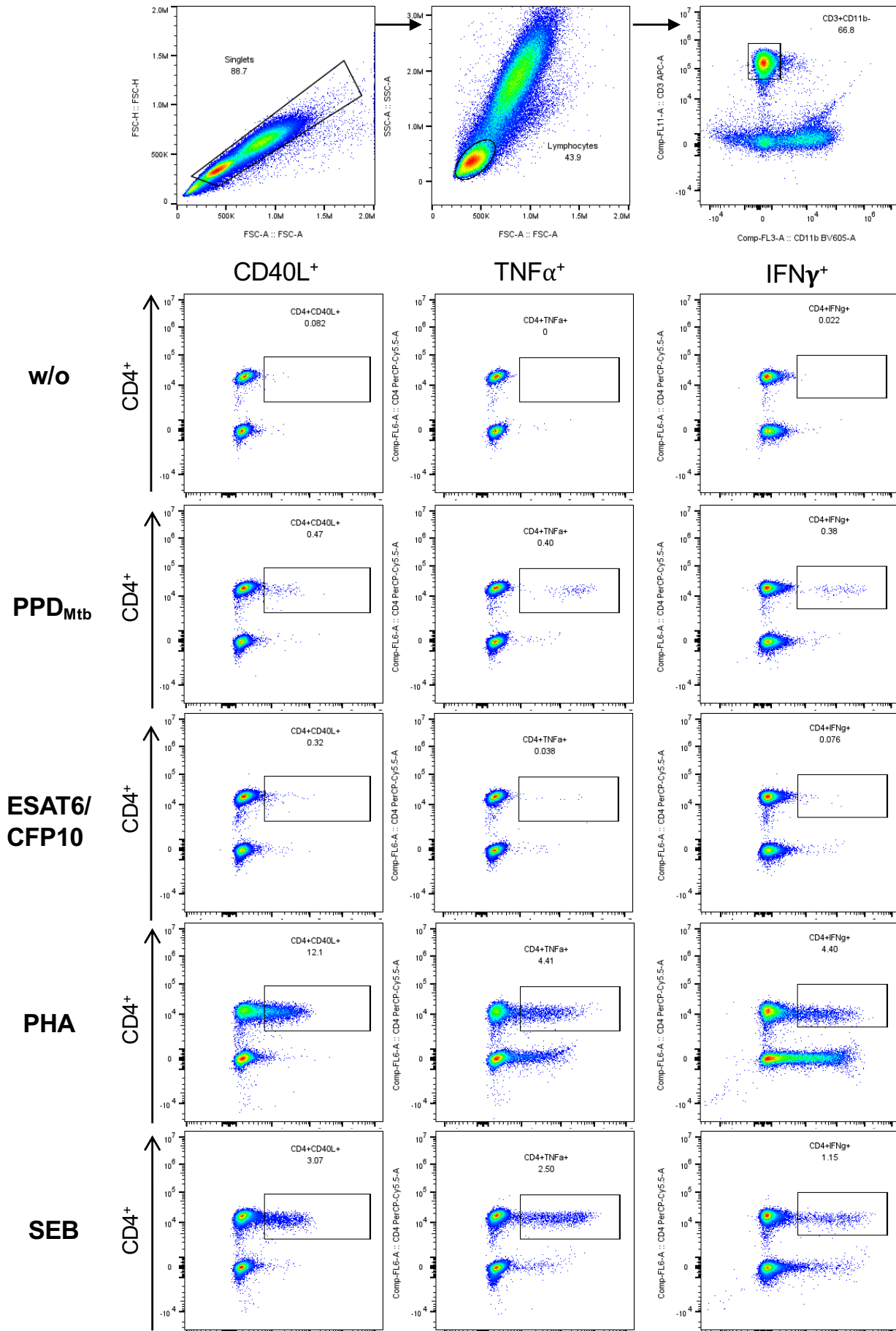
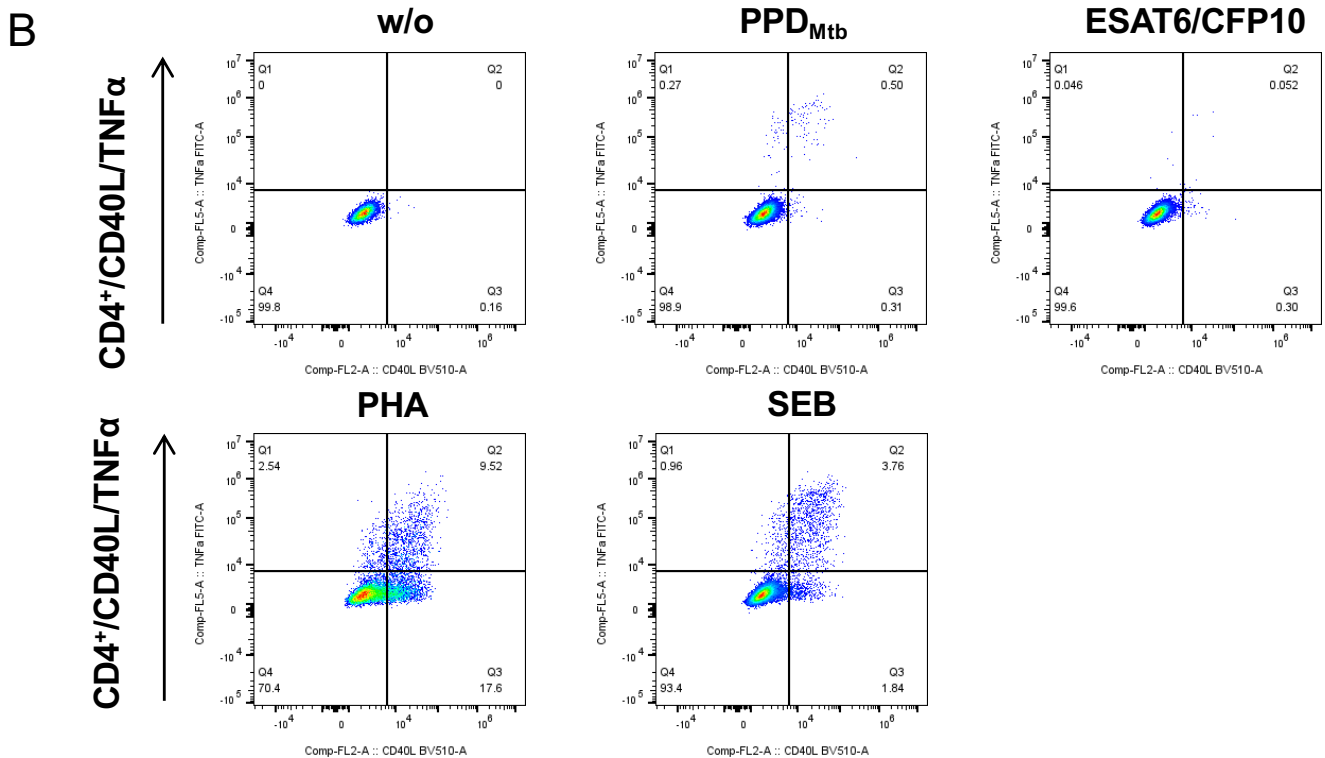
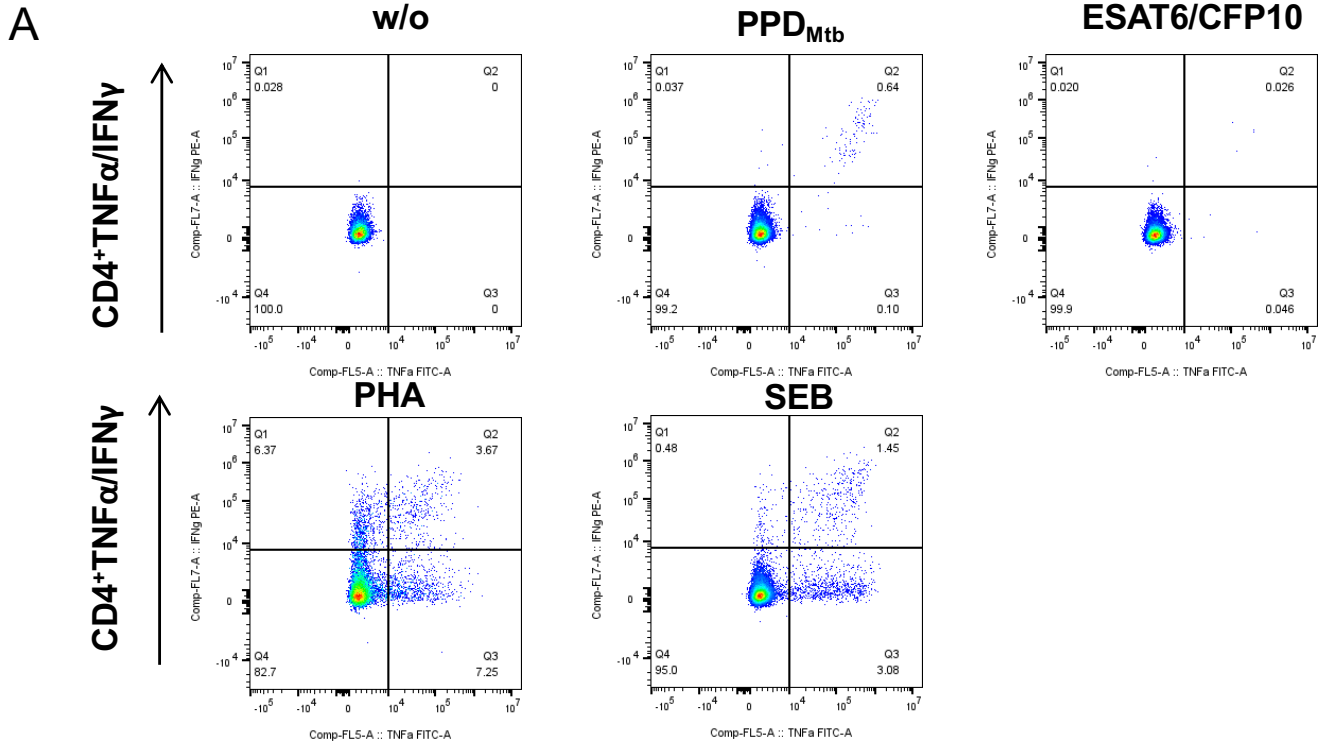
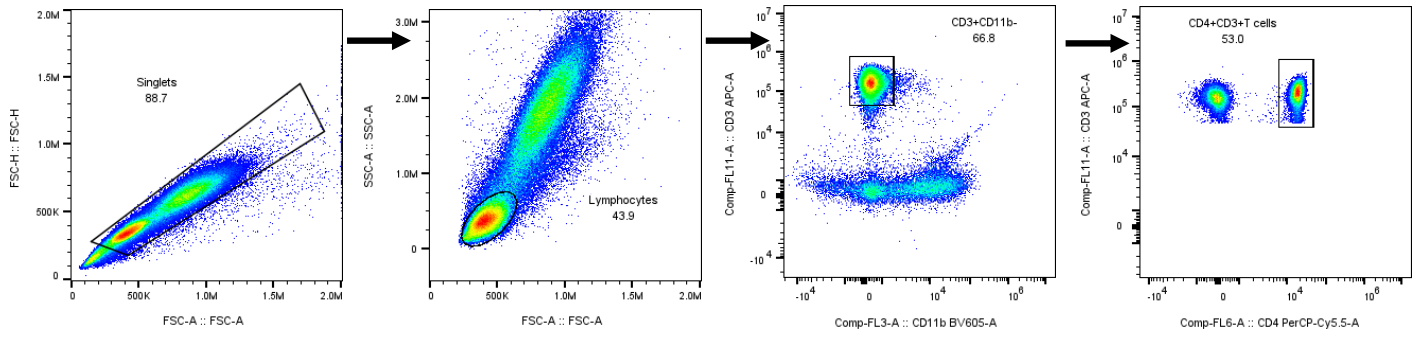


Supplementary Figure 1



T cell cytokine expression flow cytometry gating procedure, and data processing. Duplet cells were excluded using a forward scatter height (FSC-H) vs FSC area (FSC-A) dot plot. Next, lymphocyte-like cells were selected based on size (FSC-A) and granularity (Side scatter area, SSC-A). CD3⁺ lymphocytes were subsequently gated, and the proportions of CD4⁺ T-cells expressing CD40L, TNF α , and IFN- γ with or without stimulation were determined.

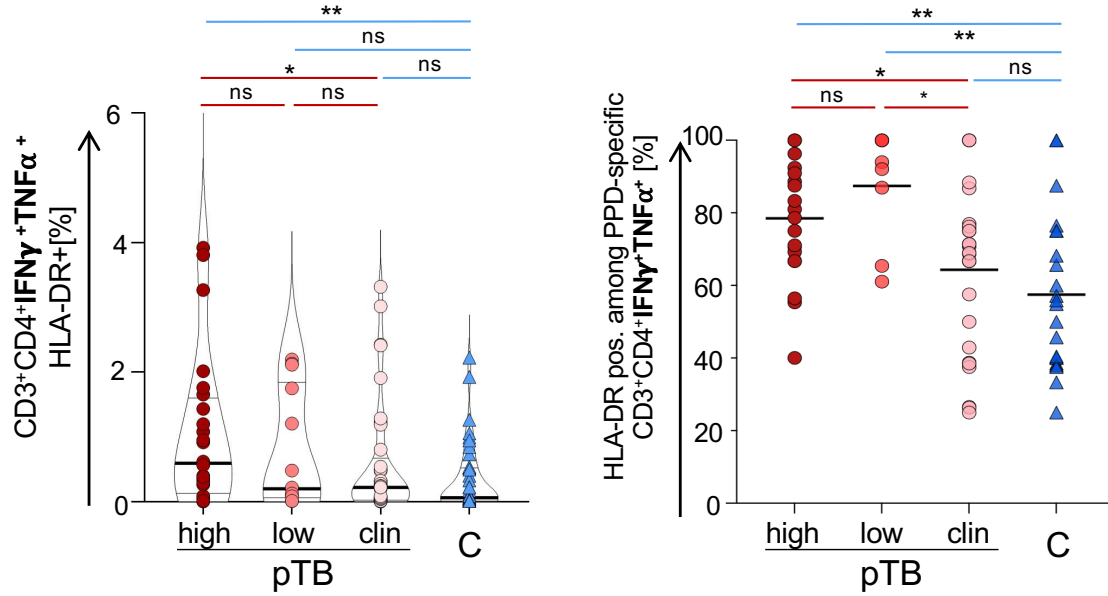
Supplementary Figure 2



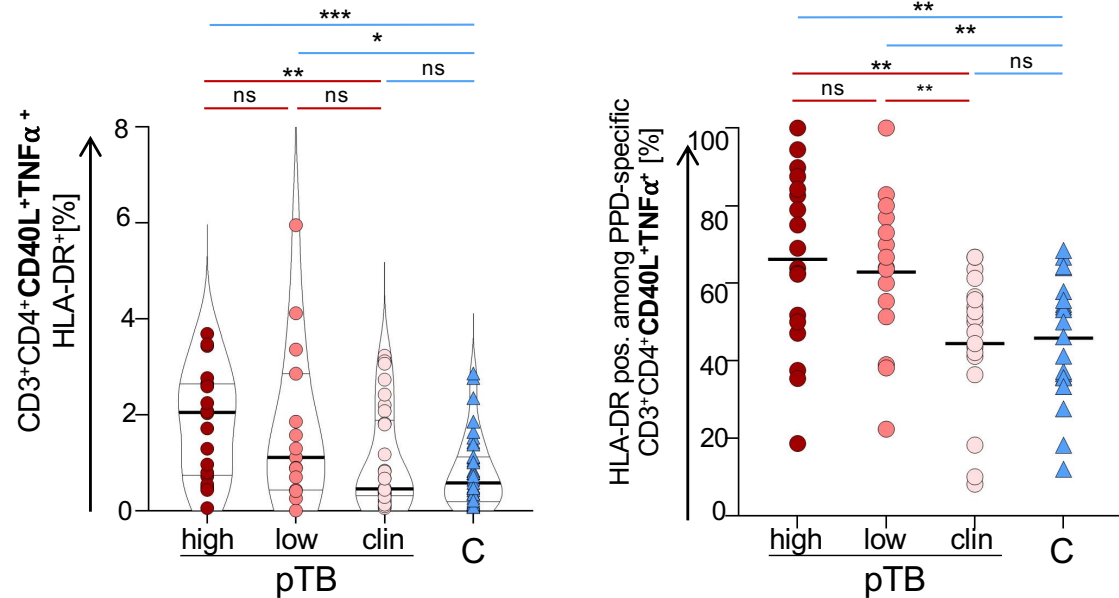
T cell cytokine expression flow cytometry gating procedure, and data processing. Initial gating from singlets to CD3⁺ lymphocytes was done as in Figure 1. Subsequently, CD4⁺ cells were gated on CD3⁺ lymphocytes, and the proportions of CD4⁺ T cells expressing TNFα/IFN-γ (Figure 2A) or CD40L/TNFα (Figure 2B) were determined, with or without stimulation.

Supplementary Figure 3

A



B

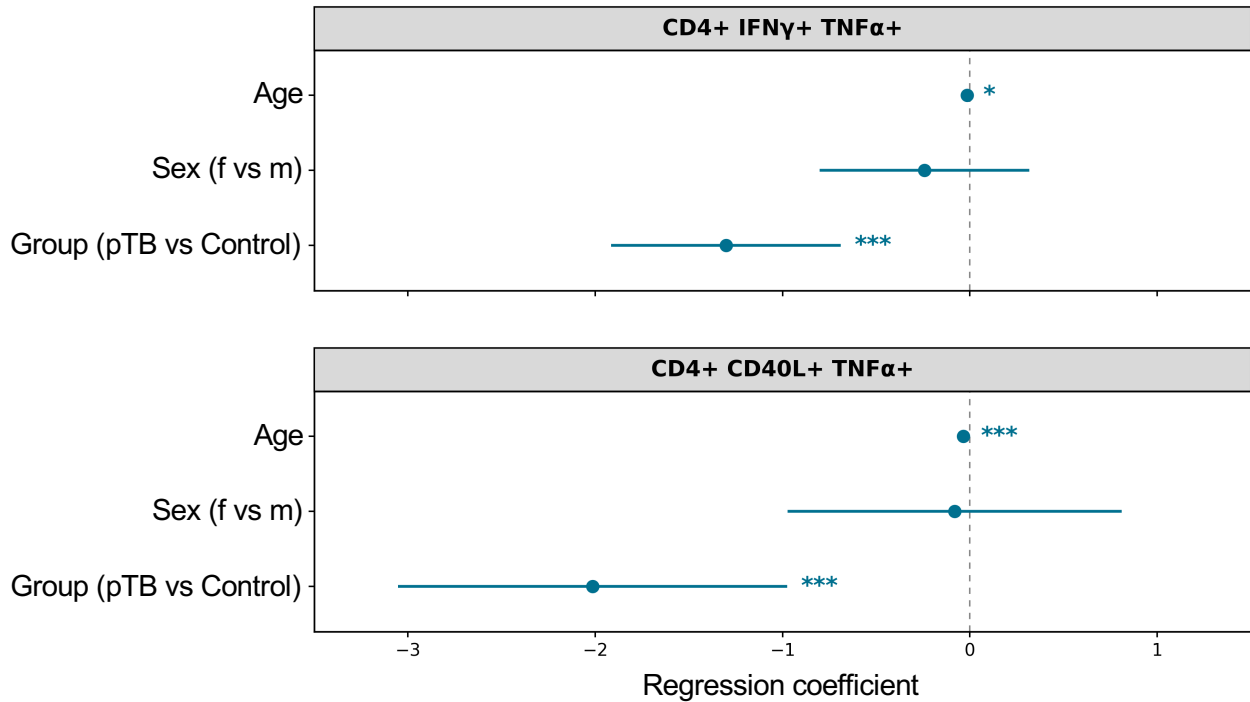


Comparison of the proportions and the relative proportions of HLA-DR positive cells on PPD_{Mtb}-specific cytokine-producing CD4⁺ T cells among subgroups of Tuberculosis patients and controls.

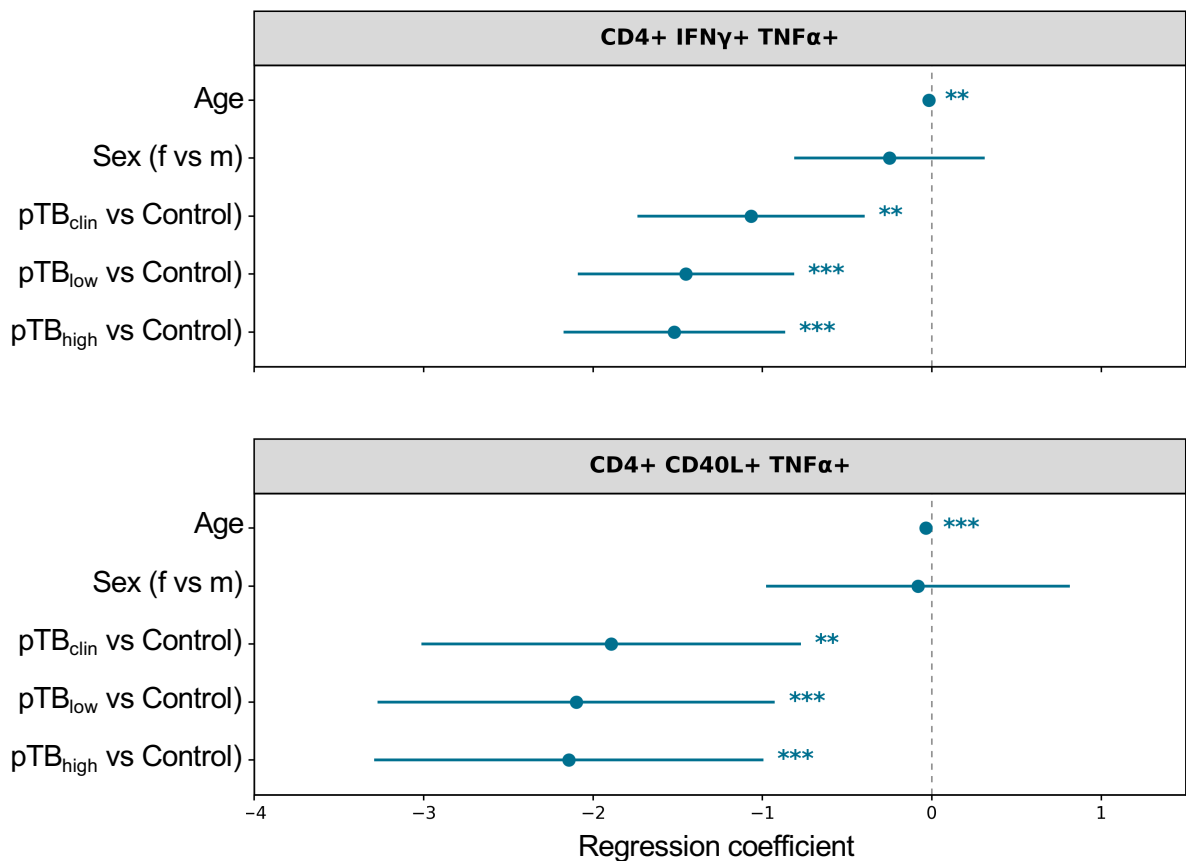
CD3⁺ CD4⁺ T cells expressing HLA-DR that produce the cytokines IFN γ /TNF α double positives (Supplementary Figure 3A, left panel) and CD40L/TNF α double positives (Supplementary Figure 3B, left panel) after PPD_{Mtb}-specific stimulation are shown for bacteriologically confirmed TB patients (TB_{high}, dark red circles; TB_{low}, bright red circles), non-bacteriologically confirmed clinically diagnosed TB patients (TB_{clin}, rose circles) and controls (HC, blue triangles). Violin symbol plots show the median value (bold line) as well as the 25th and 75th percentiles (thin lines). Again, the relative proportions of HLA-DR-positive PPD_{Mtb}-specific cytokine-producing CD4⁺ T cells are compared among the study groups using aligned dot plots, with the same codes as in the violin plots. Statistical differences between groups were assessed using the Mann-Whitney U test; asterisks denote significance levels (*, $p < 0.05$; **, $p < 0.01$; ***, $p < 0.001$; ****, $p < 0.0001$). It is noteworthy that 5 cases and 5 control samples were excluded from the analysis in Figure 3B due to a temporary shortage of CD40L antibody.

Supplementary Figure 4

A



B



Multivariable analysis of PHA-induced CD4⁺ T-cell cytokine responses. Forest plots showing regression coefficients (points) and 95% confidence intervals (horizontal bars) from multivariable linear regression models of PHA-induced CD4⁺ IFN γ /TNF α positive (upper panel) and CD4⁺ CD40L/TNF- α positive (lower panel) T-cell responses. **(A)** The pTB study group versus controls is shown **(B)** The pTB subgroups are shown versus controls. Each model includes age (years), sex (female vs male), and study group (pTB vs control) as predictors. The vertical dashed line indicates no effect ($\beta=0$). Significance is denoted by p-value asterisks (* $p<0.05$, ** $p<0.01$, *** $p<0.001$).