Figure S1

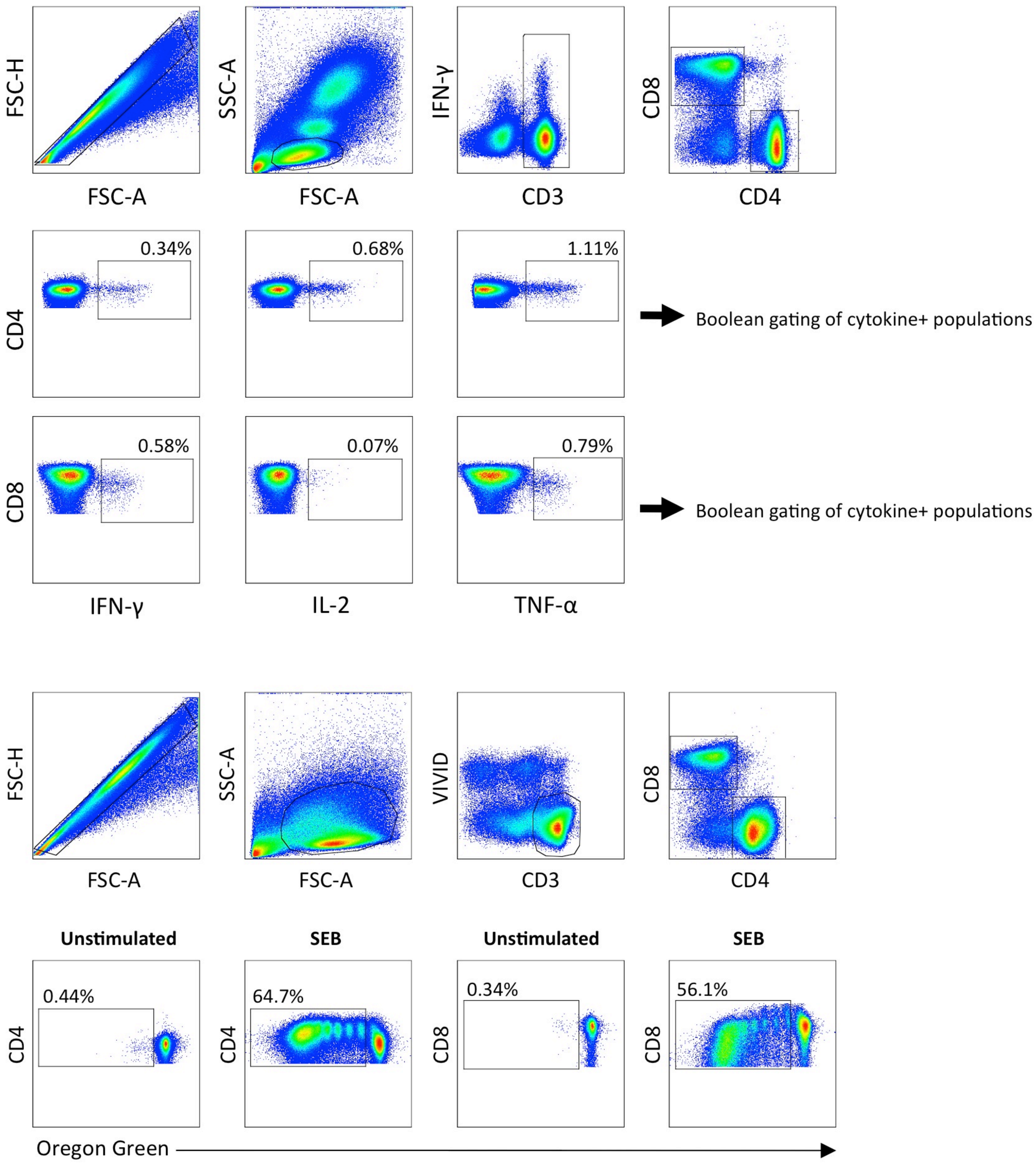

Figure S1. Flow cytometry gating strategies. (**A**) Gating strategy for the whole blood intracellular cytokine staining assay. Forward scatter area (FSC-A) versus forward scatter height (FSC-H) was used to select singlets, followed by FSC-A versus side scatter area (SSC-A) to select lymphocytes. CD3 T cells were gated within the lymphocyte population, followed by gating on CD4 or CD8 T cells. Total populations of IFN-γ, IL-2, and TNF-α-producing CD4 and CD8 T cells were selected, followed by Boolean gating to generate all possible combinations of cytokine-producing T cell populations. Representative data from whole blood of an LTBI donor stimulated for 8 hrs with SEB are shown. Percentages of cytokine-positive cells shown are after subtraction of background cytokine production in the unstimulated negative control. (**B**) Gating strategy for the PBMC proliferation assay. FSC-A versus forward FSC-H was used to select singlets, followed by FSC-A versus SSC-A to select lymphocytes. Live CD3 T cells were defined as $CD3^{+}VIVID^{low}$ lymphocytes; CD4 or CD8 T cells were gated from the live CD3 T cell population. Proliferating cells were defined as Oregon $Green^{low}$ (OG^{low}) CD4 or CD8 T cells. Representative data from freshly isolated PBMC of an LTBI donor are shown for unstimulated (no antigen) and SEB-stimulated cells.

Figure S2

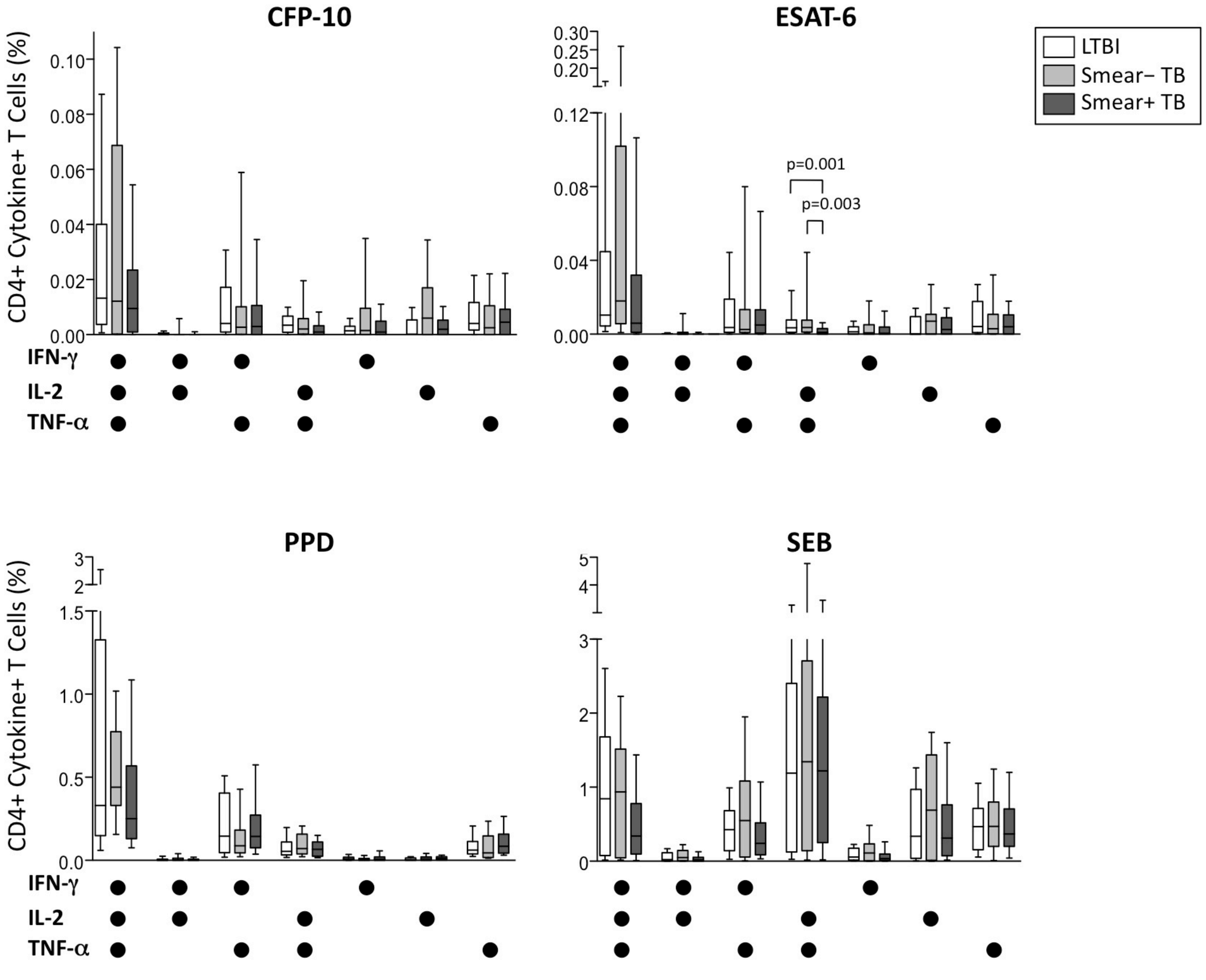

Figure S2. Frequency of cytokine-producing *Mtb*-specific CD4 T cell populations in whole blood *ex vivo*. Whole blood was stimulated for 8 hrs with overlapping CFP-10 and ESAT-6 peptide pools, PPD, or SEB. The frequencies of each CD4 T cell subset producing all possible combinations of IFN-γ, IL-2, and TNF-α are shown. Background cytokine production from the negative control sample has been subtracted. Statistical analysis between LTBI, smear− TB and smear+ TB donors was first performed by the Kruskal-Wallis test, and if significance was found ($p<0.05$), the Mann-Whitney test was used for comparisons between two groups.