

Age group, years	PI	NPI
18-45	33	19
65-74	30	17
> 74	13	16

Table S1: Distribution of pre-infected (PI) and non-pre-infected (NPI) patients in the Pfizer Cohort.

Out of 145 participants, 17 were excluded for various reasons: i/ positive IgG-N at any visit until D57 (n = 3), ii/ not analyzed by ELISpot (n = 6), iii/ not analyzed by Luminex (n = 8). Thus, 128 patients were included in this study.

The average time between the date of the positive test (Infection) and the first vaccination for the PI patients was 9.3 months [7,2; 12,2].

The V3/V1 ratio for each cytokine

Cytokine	Median	[Q1 ; Q3]
	N = 128	
IL-15	1.00	[1.00 ; 1.34]
IL-5	1.00	[1.00 ; 1.62]
PDGF-BB	1.00	[0.77 ; 2.36]
IL-9	1.12	[0.88 ; 2.22]
IL-6	1.27	[0.75 ; 2.33]
VEGF	1.00	[0.85 ; 1.80]
G-CSF	1.39	[0.92 ; 1.98]
MP-1 β	1.37	[1.04 ; 1.86]
MCP-1	1.14	[0.93 ; 2.02]
MIP-1 α	1.48	[0.99 ; 2.30]
RANTES	1.03	[1.00 ; 3.56]
IL-2	1.00	[0.83 ; 3.95]
IL-8	2.69	[1.42 ; 6.65]
TNF α	2.80	[1.50 ; 6.31]
IP-10	3.99	[1.42 ; 17.99]
ELI IFN γ	4.53	[2.28 ; 10.46]

Table S2: Type of cytokines inducing a vaccine CD4⁺T cell response following BNT162b2 vaccination.

A total of 128 volunteers were vaccinated, and T cell responses were measured as shown in Figure 1.

The median induction factor for vaccine response is depicted for each cytokine between V3 and V1. A response factor of at least 2 between V3 and V1 was required to define a T cell vaccine response. The red box highlights for which cytokines a vaccine T cell response (median fold change ≥ 2) was observed in over half of the volunteer population. All cytokines were quantified using Luminex, except for IFN- γ , which was assed by ELISpot.

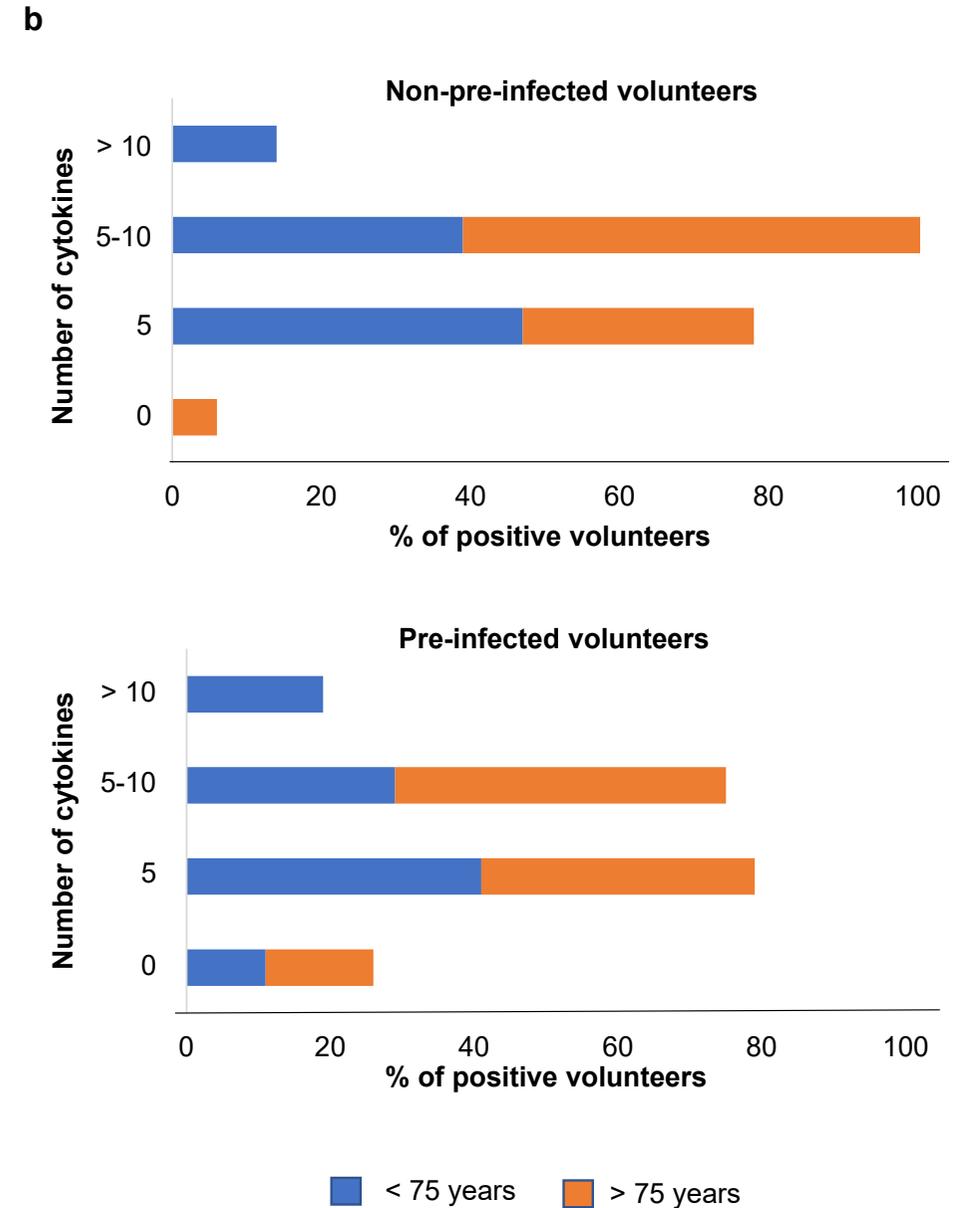
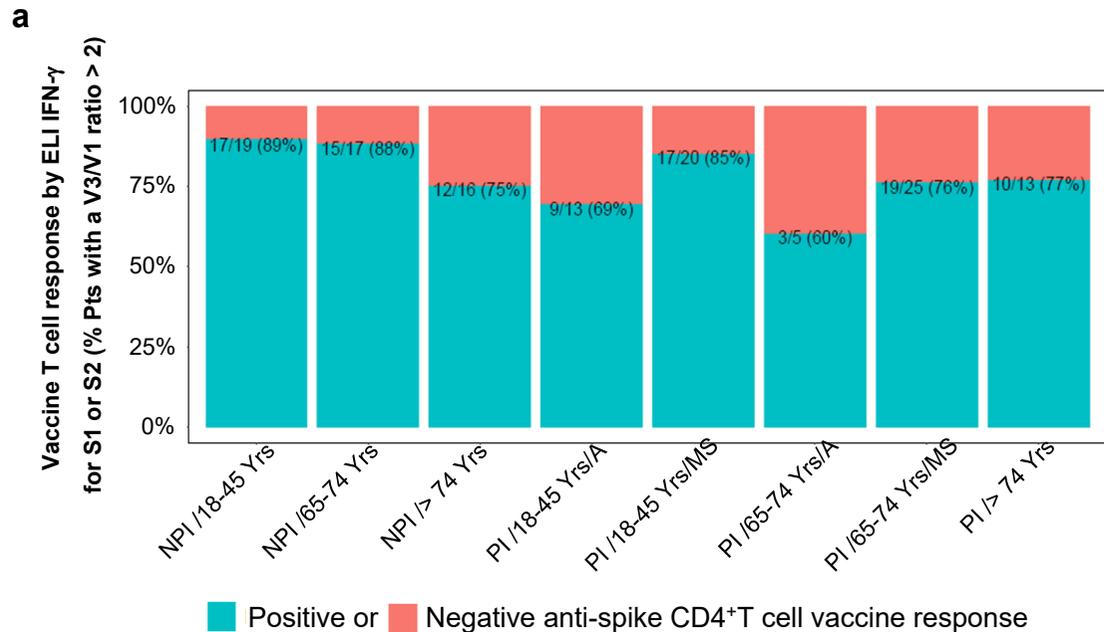
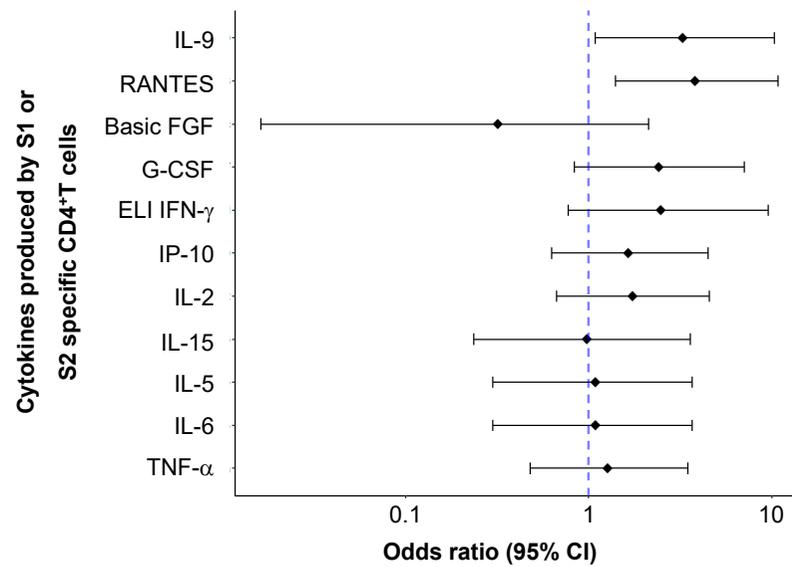


Figure S1: Age and disease severity do not affect CD4⁺T cell response in pre-infected individuals at D57 post mRNA vaccination.

(a) Volunteers were divided into several groups based on whether they were pre-infected (PI) or not (NPI), and into three age groups: 18-45 years, 65-74 years, and > 74 years. Disease severity was categorized into two groups: asymptomatic ([A]) or symptomatic (moderate or severe; MS). The percentage of vaccine CD4⁺T cell responses, measured by ELISpot IFN- γ in these different groups, is shown. Purified CD4⁺T cells were purified and sensitized with the megapool (MP8) of peptides derived from the S1 or S2 proteins. Positive reactions to either S1 or S2 MP were considered as positive. **(b)** The number of cytokines produced by CD4⁺T cells after two BNT162b2 vaccine doses was classified into four groups: 0, 5, 5-10, and > 10 cytokines. Patients were divided into two subgroups: < 75 years (blue box) or > 75 years (orange box). The percentage of patients in each cytokine group is shown within the subpopulation of vaccinated and pre-infected (B Bottom) or not (B Top) patients.



Association between each specific cytokines and vaccine-induced CD8⁺T cell in the PI group

Figure S2: Cytokine profile detection derived from CD4⁺T cells and vaccine-induced CD8⁺T cells.

A correlation was sought between the positivity (defined by the V3/VA ratio) of each cytokine produced by CD4⁺T cells sensitized by S1 and S2 following vaccination (D57) in pre-infected volunteers and the concurrent induction of CD8⁺T cell specific to S1 or S2. The Cox statistical test was used for the analysis.

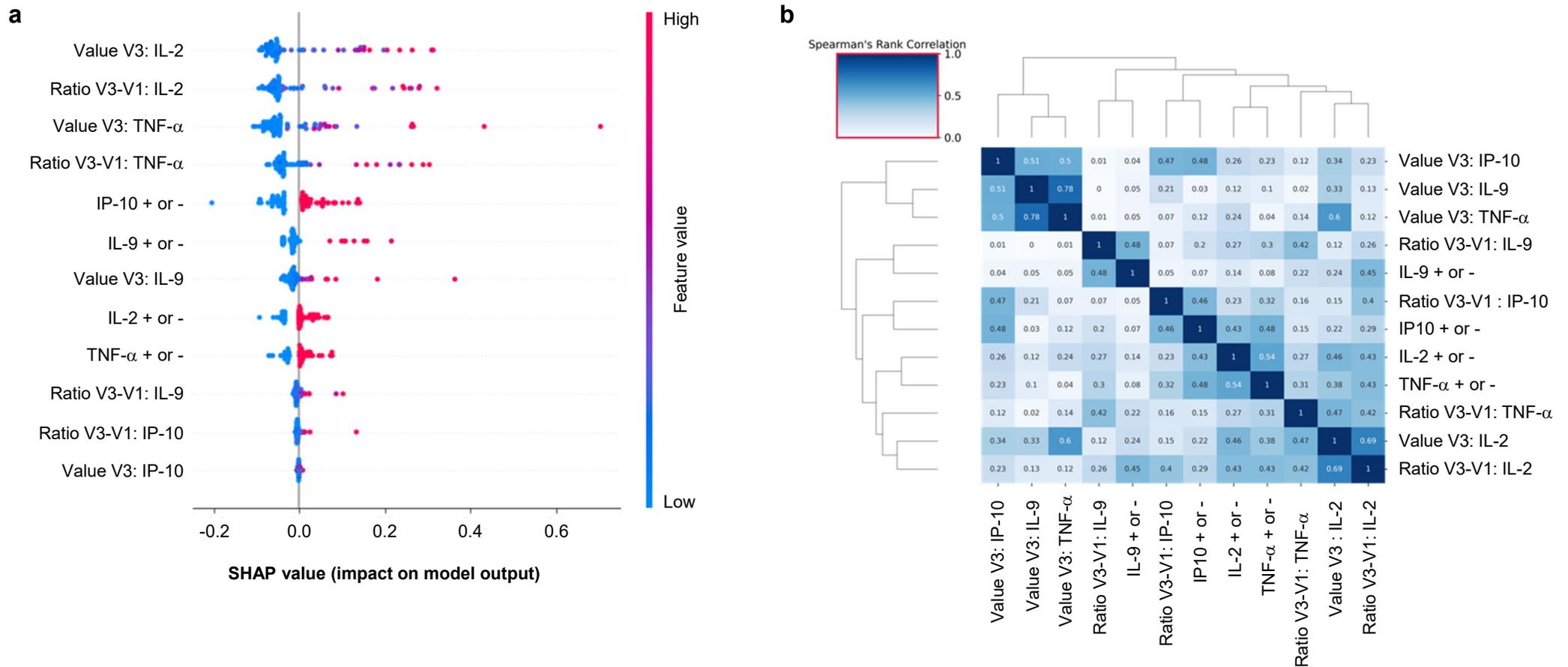


Figure S3: A Shapley additive (SHAP) analysis for the machine learning model setup in this study.

a) The impact of each parameter (V3/V1 ratio, absolute value at V3, positivity or negativity of the test for each cytokine) on the classification is shown through a swarm plot of their SHAP values and distribution. The color of the dots represents the relative value of the feature in the Moderna dataset, with high values shown in red and low values in blue. The horizontal location of the dots indicates whether the effect of that feature value contributed positively or negatively to that prediction instance (x-axis). **b)** A heatmap displays the Spearman correlation coefficients obtained between the features, where dark blue corresponds to a Spearman correlation of 1 and white corresponds to a Spearman correlation of 0. Attached to the heatmap is a dendrogram that illustrates the hierarchies of feature clusters determined through agglomerative hierarchical clustering analysis.

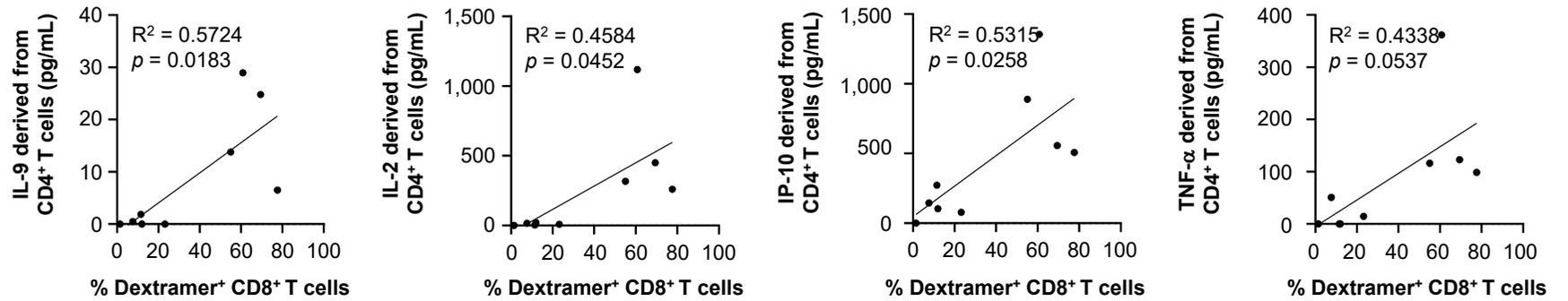
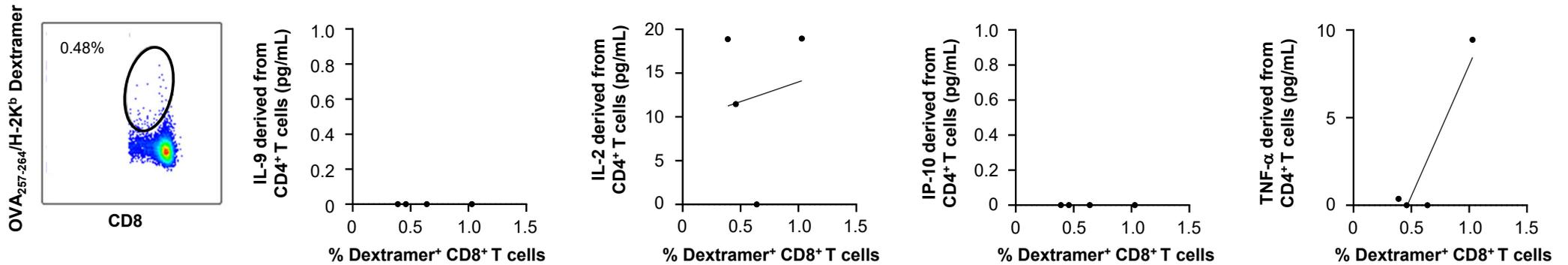
a**b**

Figure S4: Correlation between the cytokine signature of CD4⁺T cells and the percentages of ovalbumin (OVA)-specific CD8⁺T cells in the context of high or low frequency of dextramer⁺ positive CD8⁺T cells.

Mice were intranasally immunized as described in Figure 7. OVA (100 μ g) was administered alone or combined with the adjuvant C-di-GMP (10 μ g) (**a**) or with STxB-OVA without adjuvant (20 μ g) (**b**). After 21 days, concentrations of IL-9, IL-2, TNF- α , and IP-10 were measured in the supernatants of CD4⁺T cells sensitized for 36 hours with the OVA₂₆₅₋₂₈₀ peptide, which is restricted by CMHII. The Pearson statistical test was used for the analysis.