Supplement

Preserved T-cell Response in anti-CD20 treated Multiple Sclerosis Patients following SARS-CoV-2 Vaccination

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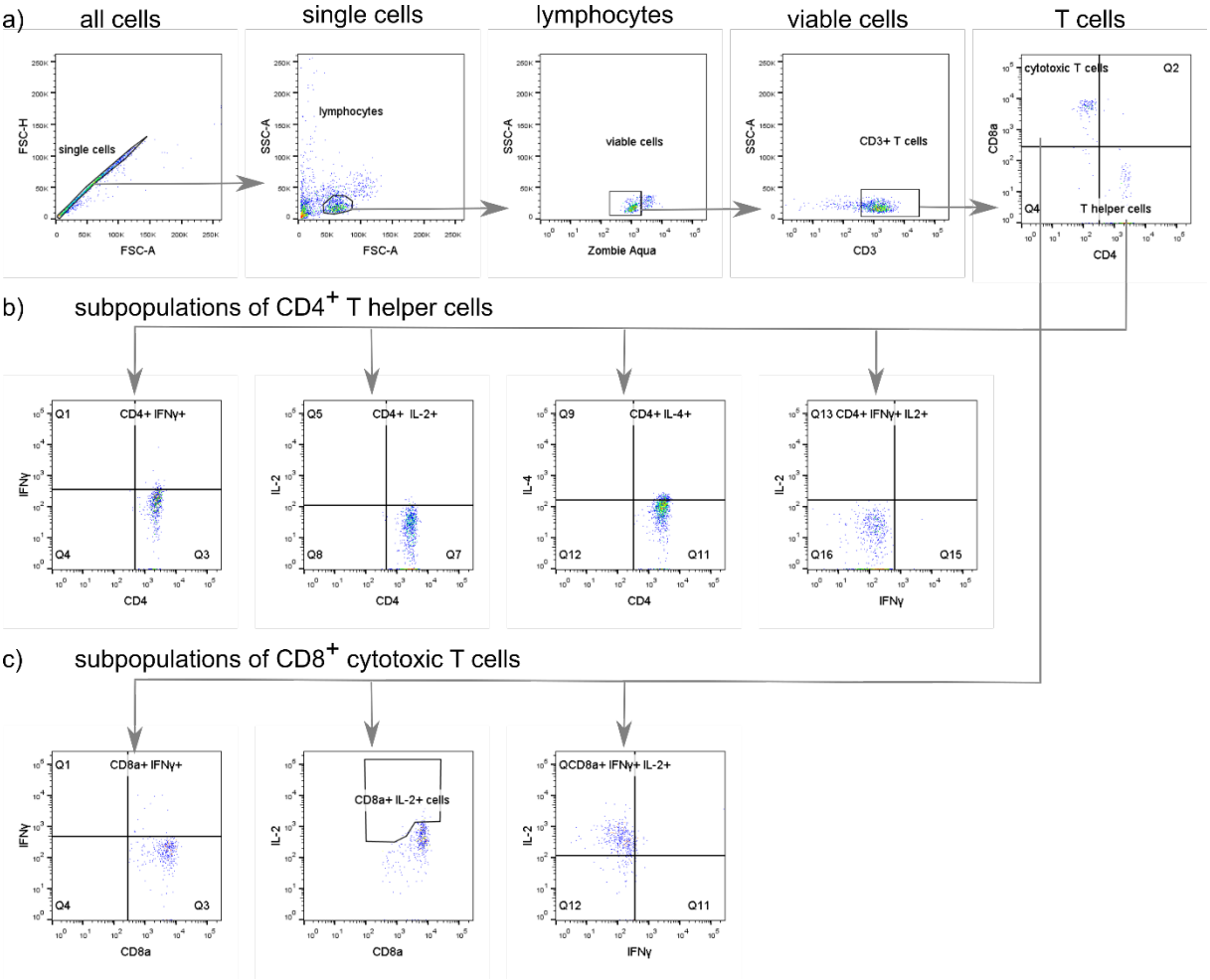
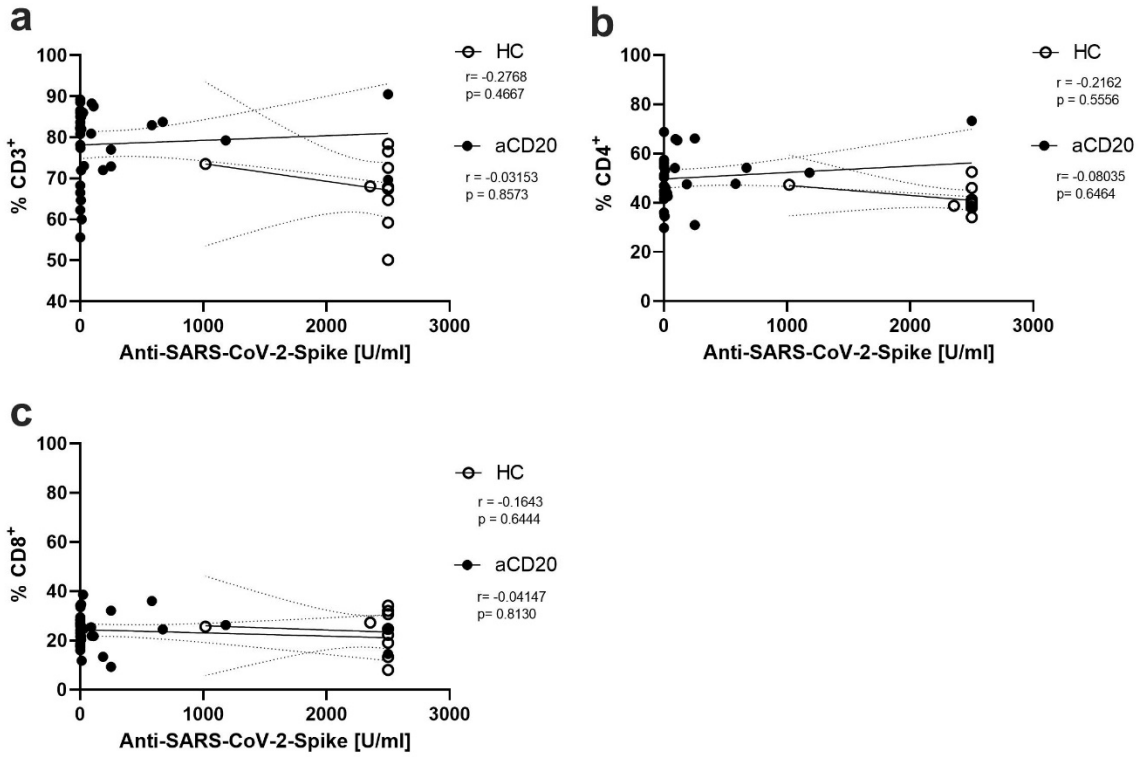
Supplementary Table 1: Antibodies used for flow cytometry.

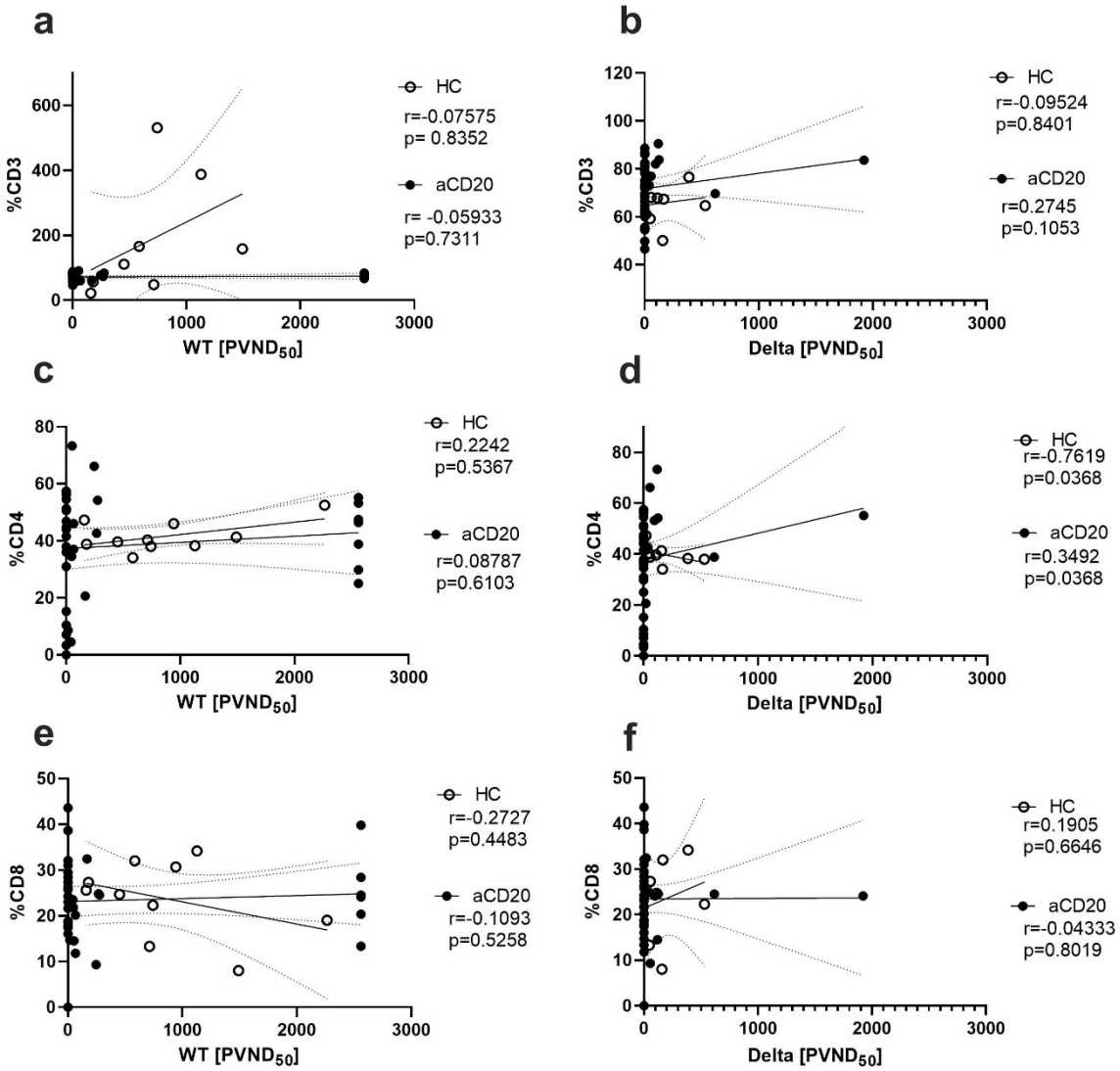
	antibody	conjugate/dye	dilution
Viability		Zombie Aqua™	1/1,500
Extracellular staining	anti-CD3	BV785	1/200
	anti-CD4	FITC	1/200
	anti-CD8a	APC/Fire750	1/200
Intracellular staining	anti-IL-2	APC	1/50
	anti-IL-4	PE	1/50
	anti-IFN-γ	PerCP-Cy5.5	1/50



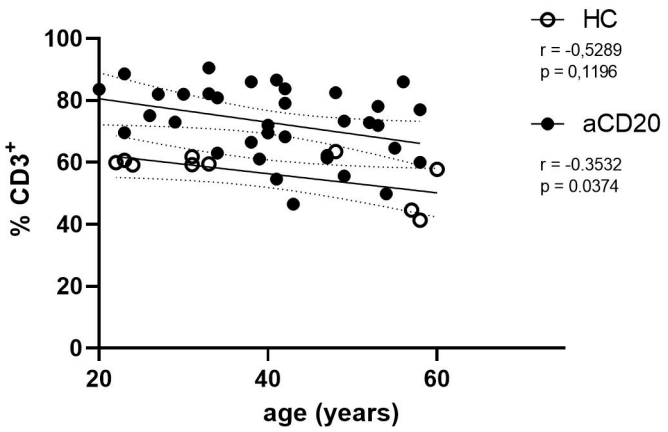
Supplementary Figure 1: Gating strategy of T cell subpopulations in SARS-CoV-2 peptide stimulated PBMCs after flow cytometry measurements. The gating strategy was applied to data shown in Figures 3-5. a) $CD8^+$ and $CD4^+$ T cells were gated within viable single T cells. b) Subpopulations of $CD4^+$ T helper cells. c) Identification of subpopulations of $CD8^+$ cytotoxic T cells.



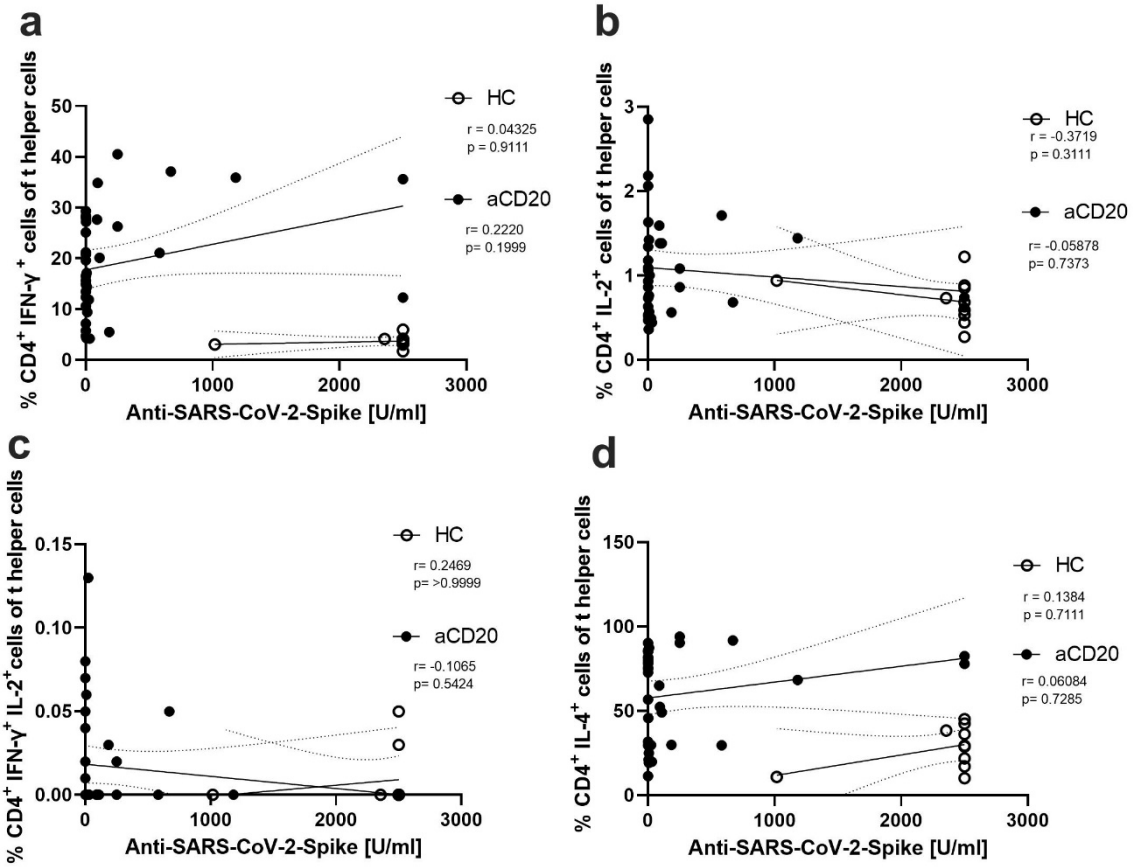
Supplementary Figure 2: Correlation of Anti-SARS-CoV-2-Spike titer and T cell populations in HC and aCD20 treated MS patients. No correlation of anti-SARS-CoV-2-Spike titers and a) $CD3^+$ T cell frequencies, b) $CD4^+$ Th helper cell frequencies and c) $CD8^+$ cytotoxic T cell frequencies. Reference range of Anti-SARS-CoV2 antibodies: <0,8-2500 U/mL. Data were analyzed with non-parametric, two-tailed Spearman correlation.



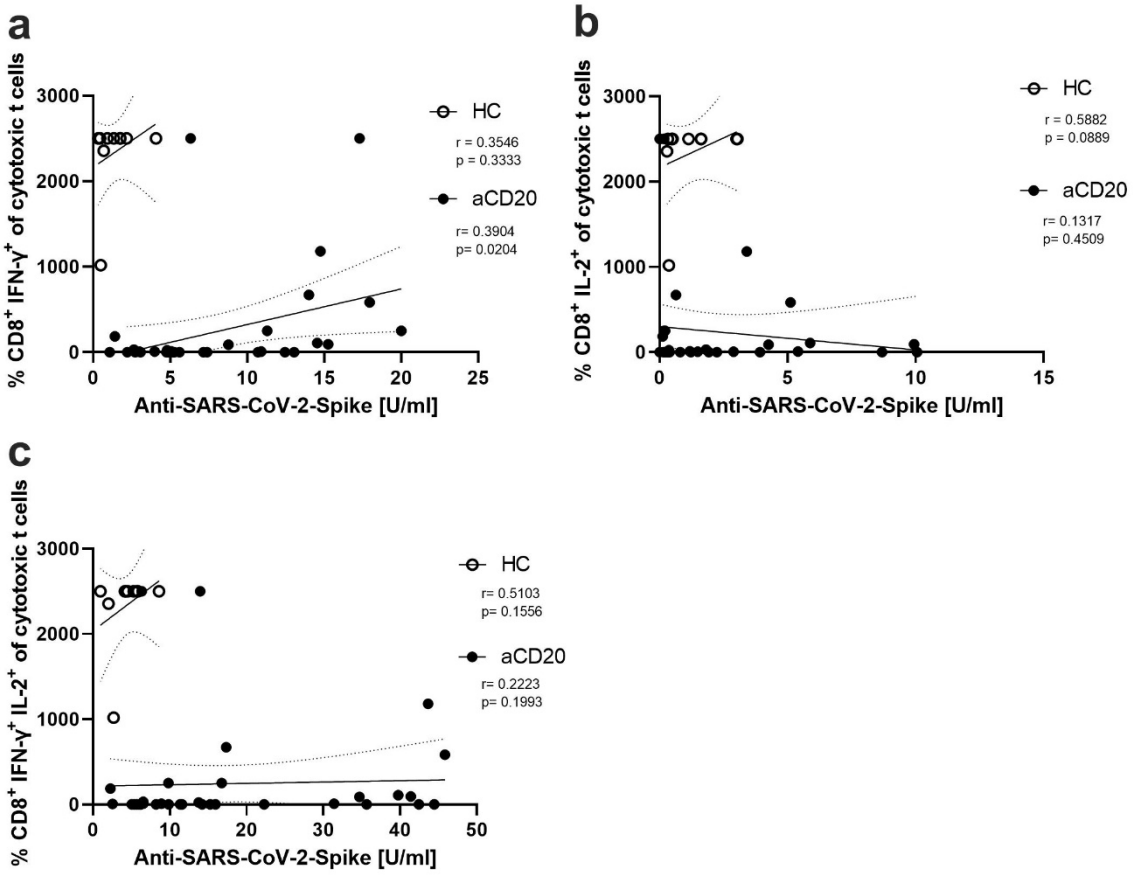
Supplementary Figure 3: Correlation of neutralization capacity and frequencies of CD3$^+$, CD4$^+$ and CD8$^+$ T cell populations. Correlation of WT neutralization with frequencies of a) CD3$^+$, c) CD4$^+$ and e) CD8$^+$ T cells and correlation of delta neutralization with frequencies of b) CD3$^+$, d) CD4$^+$ and f) CD8$^+$ T cells. Data were analyzed with non-parametric, two-tailed Spearman correlation.



Supplementary Figure 4: Correlation of age with CD3$^+$ T cell population. Negative correlation of age with CD3$^+$ T cell populations in aCD20 treated MS patients. Data were analyzed with non-parametric, two-tailed Spearman correlation.



Supplementary Figure 5: Correlation of anti-SARS-CoV-2-Spike titer and CD4$^+$ Thelper cell subpopulations. Anti-SARS-CoV-2-Spike titers did not correlate with frequencies of a) CD4$^+$IFN-γ$^+$ cells, b) CD4$^+$IL-2$^+$ cells, c) CD4$^+$IFN-γ$^+$IL-2$^+$ cells or d) CD4$^+$IL-4$^+$ cells. Reference range of anti-SARS-CoV-2 antibodies: <0,8-2500 U/mL. Data were analyzed with non-parametric, two-tailed Spearman correlation.



Supplementary Figure 6: Correlation of anti-SARS-CoV-2-Spike titer and $CD8^+$ Tc cell subpopulations. Anti-SARS-CoV-2-Spike titers correlated with frequencies of a) $CD8^+IFN\text{-}\gamma^+$ cells in aCD20 treated MS patients but not in HC. No correlation of anti-SARS-CoV-2-Spike titers with b) $CD8^+IL\text{-}2^+$ cells or c) $CD8^+IFN\text{-}\gamma^+IL\text{-}2^+$ cells. Reference range of anti-SARS-CoV-2 antibodies: <0,8-2500 U/mL. Data were analyzed with non-parametric, two-tailed Spearman correlation.