Supplemental material

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Supplemental material

Methods: T cell responses

Peripheral blood mononuclear cells (PBMC) were thawed, resuspended in R10 medium (RPMI, 10% FCS) and rested for 2 hours at 37ºC, 5% CO_2 before plating at $2x10^5$ cells in duplicate per restimulation condition at a final concentration of: 1.5 µg/ml SARS-CoV-2 Spike Glycoprotein peptide mix (JPT, Germany), 0.5 µg/ml SEB peptides (Sigma Aldrich, Japan) or R10 with 0.4% DMSO. After 18 – 20 hours incubation, secreted IFN-γ was detected using 1 µg/ml anti-IFN-γ biotinylated detection mAb (7-B6-1-biotin, Mabtech, Sweden) followed by 1 µg/ml streptavidin-alkaline phosphatase for 1 hour (Mabtech, Sweden). Plates were developed using NBT/BCIP substrate (ThermoFischer, USA) and scanned on an AID ELISpot Reader. Results are reported as IFN-γ spot forming cells (s.f.c) per 10^6 PBMC as determined by the mean number of spots over background.

Results: SARS-CoV-2 specific IFN-γ release

PBMC were available in 66 patients and 21 HC. IFN-γ-secreting cells were quantified after stimulation with a SARS-CoV-2 spike peptide mix. Few baseline spike-specific IFN-γ-secreting T cells were detected pre-vaccination, indicative of cross-reactivity with common cold coronavirus spike epitopes (Suppl Fig. 4A).[1,2] The number of IFN-γ-spot forming cells (s.f.c) was comparable between HC and RA patients, with a peak two weeks after the second vaccine dose (T2) and decrease thereafter. In RA patients, a more rapid decline was noted at week 24 compared to HC (Suppl Fig 4B). No significant differences regarding the T cell response between RA patients on csDMARDs-mono, anti-cytokine bDMARDs or tsDMARDs were detected (Suppl Fig. 4C). After the first vaccination (T1), but not at subsequent visits, we observed that RA patients treated with csDMARDs-mono exhibited a decrease in the number of peripheral blood IFN-γ-secreting T cells, as compared to HC or RA patients treated with tsDMARDs or anti-cytokine bDMARDs-combo

(Suppl Fig. 4D). No differences were observed in the spike-specific cellular responses between RA patients treated with anti-cytokine bDMARDs-mono or combo versus RA patients on csDMARDs-mono (Suppl Fig. 4E). RA patients treated with tsDMARDs exhibited a decreased T cell response three weeks following the first vaccination compared to HC and RA patients treated with csDMARDs, although these differences were no longer evident after the second vaccine dose (Suppl Fig. 4F).

References

1. Grifoni A, Weiskopf D, Ramirez SI, et al. Targets of T Cell Responses to SARS-CoV-2 Coronavirus in Humans with COVID-19 Disease and Unexposed Individuals. *Cell* 2020; 181(7): 1489-1501 e15.

2. Bacher P, Rosati E, Esser D, et al. Low-Avidity CD4(+) T Cell Responses to SARS-CoV-2 in Unexposed Individuals and Humans with Severe COVID-19. *Immunity* 2020; 53(6): 1258-1271 e5.

Legends to supplemental figures

Legend to Supplement Fig 1:

RA disease activity as assessed by CDAI (median) at baseline (T0), 3 weeks (T1), 6 (T2) and 12

weeks (T3) after the first vaccine dose

Red horizontal lines indicate median, black horizontal lines indicate 1st and 3rd IQR

***p < 0.001, **** p < 0.0001

Legend to Supplement Fig 2:

Longitudinal antibody response in RA patients and healthy controls.

Dynamics of antibody binding titers against SARS-CoV-2 antigen S1 as assessed by Elecsys Anti-

SARS-CoV-2 (S) assay (U/ml) in healthy controls (A) and all RA patients (B) as well as stratified

by treatment (C) csDMARDs (D) bDMARDs (E) Abatacept (F) tsDMARDs.

Legend to Supplement Fig 3:

(A) At all timepoints following vaccination (T1-T4), RA patients had significantly lower anti-S1

levels compared to age and vaccination matched healthy controls, whereas no differences in T

cell responses (SARS-CoV-2 specific IFN-γ release) were detected (B)

*$p<0.05$, **$p<0.01$, ***$p<0.001$.

Legend to Supplement Fig 4:

SARS-CoV-2-specific IFN gamma production over 24 weeks.

(A) Quantification of the number of IFN-γ spot forming cells (s.f.c) per 10^6 PBMC at baseline (T0)

and the indicated time-points following vaccination in HC or patients with RA.
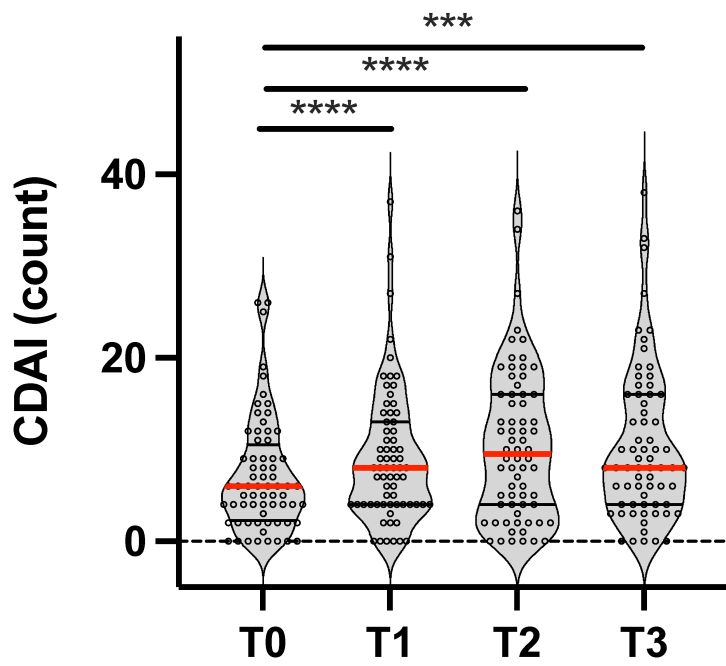
(B) Representative ELISPOT wells following PBMC restimulation with SARS-CoV-2 Spike

peptide mix at the indicated time points for HC and RA patients.

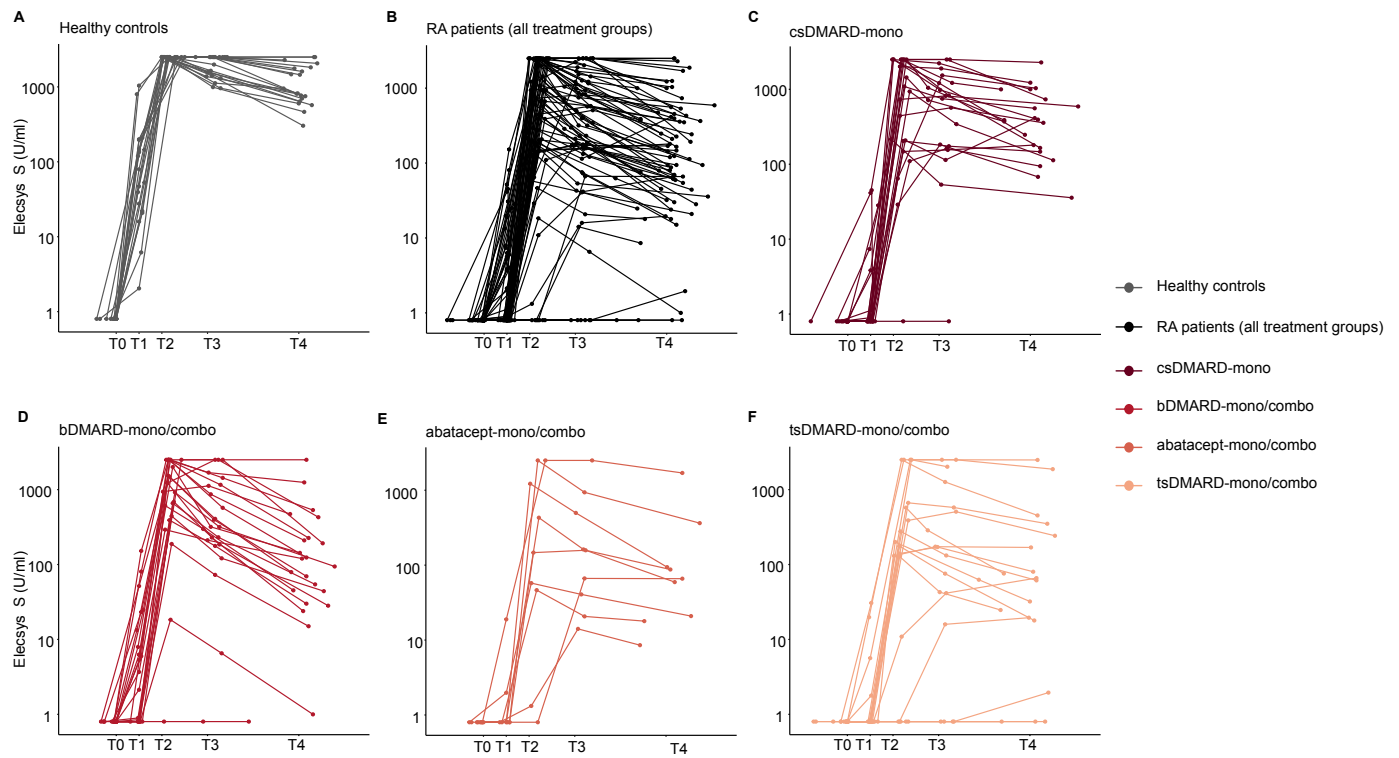(C) Quantification of IFN-γ s.f.c in individuals with RA stratified according to different DMARD categories.

(D-F) Quantification of IFN-γ s.f.c in RA patients treated with csDMARDs (D), anticytokine bDMARDs (E), or tsDMARDs (F).

*p<0.033, **p<0.002

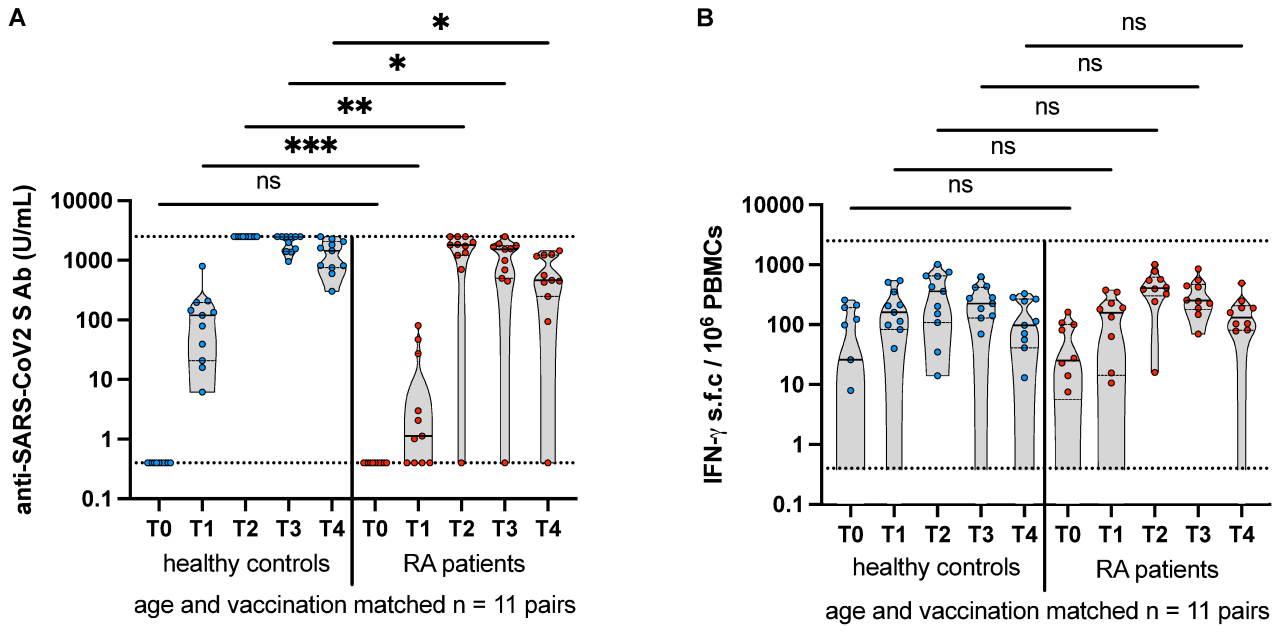Supplemental material

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Supplement Fig 1

Supplement Fig 2

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Supplement Fig 4