Supplementary methods.

Serological testing

Testing was performed according to the manufacturer's instruction locally adapted for performance at half-volume.

T cell analysis

PBMC were thawed and viable cells resuspended in AIM-V serum free media onto the ELISpot plate ($2.5x10^5$ viable cells/well) and exposed to negative control, antigen mixtures containing peptides derived from either the Spike protein or NP, or PHA (positive control). Plates were incubated overnight at 37ºC with 5% CO_2. After cell removal, alkaline phosphate conjugated anti IFN gamma antibody incubation, followed by BCIP/NBTplus substrate incubation, the plates were dried and spots (SFU) counted. The reference range was determined by the manufacturer. A significant test was determined by subtracting the SFU in the negative control from the number of SFU in the stimulated wells. A signal of greater than 10 SFU in the negative control invalidated that sample. Results were interpreted according to the manufacturer as: negative 0-4 SFU, borderline 5-7 SFU, weak positive 8-15 SFU, positive 16-30 SFU and strong positive >30 SFU.

Interferon (IFN) core analysis

IFN score was calculated as the average of the natural logarithm of IFN-induced chemokines (6). High IFN scores (>2STDEV of healthy control population) predict worsening of clinical outcome of IFN-related dcSSc disease (7). Samples were diluted 1:2 and assayed according to the manufacturer's instructions and analysed using a Luminex 200 instrument with xPonent 4.2.

Results

Responses after second SARS-CoV-2 vaccination in patients with absent seroconversion.

Antibody response after second vaccine: were as follows: abatacept 3/6 (50%), RTX 7/14 (50%), anti-TNF 5/9 (56%), JAKi 2/3 (67%) and anti-IL6 2/3 (67%), and p=0.971. Eight of 15 (53%) patients on concomitant MTX seroconverted. For patients treated with RTX; treatment time ≤ 6months compared to >6 months was associated with a reduced seroconversion rate 3/9 (33%) vs. 6/9 (67%) p=0.094.

Efficacy post vaccine

The participants were questioned whether they had contracted PCR confirmed COVID-19 infection, 9 months after the start of the study. In addition, the patient electronic case notes were reviewed for evidence of COVID-19 infection. There were 8 SARS -CoV-2 confirmed symptomatic cases during the study follow-up. Four patients on RTX, 3 with anti-TNF, 1 with anti-IL6 and 1 healthy control. Of the 2 patients with absent seroconversion (both received treatment with RTX) after the first does of the vaccine, one patient failed to convert after the second dose but had strong T cell responses and had mild symptoms. The other patient had void T cell responses and didn't attend for the post second vaccine bloods as was already

C:\Users\SaleemB\AppData\Local\Temp\MicrosoftEdgeDownloads\e97da3e9-dba1-4eee-a646-6648b338dac4\RMDREsubmissionSupplementary FINAL.docx

unwell with COVID symptoms. The latter patient was the only patient to be admitted to hospital. Of note none of 8 cases had pre vaccine SARS-CoV-2 exposure.

C:\Users\SaleemB\AppData\Local\Temp\MicrosoftEdgeDownloads\e97da3e9-dba1-4eee-a646-6648b338dac4\RMDREsubmissionSupplementary FINAL.docx