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SARS-CoV-2 seroprevalence, seroconversion and neutralizing antibodies in a systemic lupus erythematosus cohort and comparison to controls

Sir,

At the outset of the SARS-CoV-2 pandemic, it was speculated that systemic lupus erythematosus (SLE) patients may be at significant risk of COVID-19 due to underlying immune dysregulation and immunosuppressive therapies (reviewed in[1]). It was unclear how these factors would alter B/T cell responses, risk of infection, and/or development of neutralizing antibodies.[2,3] In this study, we examined the prevalence of SARS-CoV-2 antibodies using multiple assays, RT-PCR positivity, and neutralizing antibodies in 173 SLE patients (94.8% female, mean age 48.5 years, mean disease duration 11.7 years, 42.8% non-White race/ethnicity, 83.2% prescribed hydroxychloroquine, 28.9% corticosteroids, and 43.9% other immunomodulators) prior to vaccination compared to controls.

Pre-pandemic serum samples biobanked prior to 01/01/2020 and intra-pandemic samples collected from 03/15/2020–01/31/2021 were tested for SARS-CoV-2 antibodies using an ELISA measuring IgA and IgG anti-spike 1 (S1) protein (Euroimmun AG, Lübeck, Germany) and an assay detecting IgG antibodies to nucleocapsid (N), S1 receptor binding domain (RBD), and S1 (XMAP®: Luminex Corporation, Austin, TX) and conventional SLE autoantibodies (anti-Ro52, -SSA/Ro60, -SSB/La, -Sm, -U1RNP, -ribosomal P, and -dsDNA). One hundred pre-pandemic and 148 intra-pandemic sera (i.e., 248 unique individuals) from unselected ambulatory individuals undergoing autoantibody testing served as controls. RT-PCR tests were performed on the SLE cohort if clinically indicated and results retrospectively collected until 01/31/2021. Pre-pandemic and intra-pandemic SLE and control samples with antibodies to at least one SARS-CoV-2 antigen were tested for neutralizing antibodies using the Surrogate Virus Neutralization Test (GenScript Biotech Corporation, Piscataway, NJ, USA).[4]

None of the SLE patients had pre-pandemic SARS-CoV-2 antibodies versus 6% of controls (difference −6.0%, 95% CI: −10.7%, −1.4%; Table 1). Comparable proportions of SLE patients and controls had at least one intra-pandemic SARS-CoV-2 antibody (3.5% versus 4.7%, difference −1.2%, 95% CI: −5.6%, 3.2%). A sensitivity analysis age and sex-matching controls to SLE patients (2:1) yielded similar results. Intra-pandemic seroprevalence of IgG antibodies to the N protein in SLE patients was lower than in the general population (Calgary, AB Canada) over a similar observation interval[5] (0.6% vs 2.9%, difference −2.3%, 95% CI: −3.6%, −1.0%). 7.5% (6/80) of SLE patients had a positive RT-PCR. None of the 173 SLE patients in the cohort, including the nine SLE patients with either intra-pandemic SARS-CoV-2 antibodies and/or a positive RT-PCR (Supplementary Table 1) were hospitalized for SARS-CoV-2 infection.

Two of six SLE patients with at least one SARS-CoV-2 intra-pandemic antibody developed neutralizing antibodies (medication profiles in Supplementary Table 1); both had IgG antibodies to the RBD of SARS-CoV-2 (Supplementary Table 2). None of six controls with at least one pre-pandemic antibody to SARS-CoV-2 had neutralizing antibodies, whereas 4/7 controls with at least one intra-pandemic SARS-CoV-2 antibody had neutralizing antibodies, three of which had IgG antibodies to RBD. As shown in Supplementary Table 3, there was no statistical difference (chi-squared test) in frequency of any pre- or intra-pandemic SLE-related autoantibodies between SLE patients with and without SARS-CoV2 positivity. This and the absence of SARS-CoV2 antibodies in pre-pandemic sera suggests that molecular mimicry is an unlikely explanation for SARS-CoV-2 seropositivity.[6]

Like other reports,[3,7,8] our SLE cohort had a lower rate of seropositivity pre-pandemic and a slightly lower to similar rate of seropositivity intra-pandemic compared

Table 1. SLE cohort patients and controls with pre- and/or intra-pandemic SARS-CoV-2 antibodies.[a]

			SLE, n (%) (n = 173)	Controls, n (%) (pre-pandemic, n = 100; intra-pandemic, n = 148)	Difference between cohort and controls, % (95% CI)
Pre-pandemic	ELISA	*Anti-S1 IgA*[b]	0 (0.0)	3 (3.0)	−3.0 (−6.3, 0.3)
		Anti-S1 IgG[c]	0 (0.0)	4 (4.0)	−4.0 (−7.8, 0.1)
	xMAP®	*Anti-N IgG*[d]	0 (0.0)	2 (2.0)	−2 (−4.7, 0.7)
		Anti-RBD IgG[d]	0 (0.0)	0 (0.0)	0. (0.0, 0)
		Anti-S1 IgG[d]	0 (0.0)	0 (0.0)	0. (0.0, 0)
	Patients with at least one antibody[e]		0 (0.0)	6 (6.0)	**−6.0 (-10.7, -1.4)**
	Neutralizing antibodies[f]		0 (0.0[g])	0 (0.0[g])	NA
Intra-pandemic	ELISA	*Anti-S1 IgA*[b]	1 (0.6)	3 (2.0)	−1.4 (−3.9, 1.1)
		Anti-S1 IgG[c]	5 (2.9)	5 (3.4)	−0.5 (−4.3, 3.3)
	xMAP®	*Anti-N IgG*[d]	1 (0.6)	5 (3.4)	−2.8 (−5.9, 0.3)
		Anti-RBD IgG[d]	2 (1.2)	3 (2.0)	−0.8 (−3.6, 2.0)
		Anti-S1 IgG[d]	0 (0.0)	1 (0.7)	**−0.7 (-11.1, -2.9)**
	Patients with at least one antibody[e]		6 (3.5)	7(4.7)	−1.2 (−5.6, 3.2)
	Neutralizing antibodies[f]		2 (33.3)	4 (57.1)	Not done[h]
	RT-PCR (+/−)		6 (7.5[i])	NA	

Bolded differences indicate statistical significance.
ELISA: enzyme-linked immunosorbent assay; IgA: immunoglobulin A; IgG: immunoglobulin G; N: nucleocapsid; NA: not applicable; RBD: receptor binding domain; RT-PCR: reverse transcription-polymerase chain reaction; S1: spike 1 protein; SLE: systemic lupus erythematosus; XMAP: Luminex addressable laser bead imunoassay.
[a]None of the SLE patients received a COVID vaccination prior to intra-pandemic testing. None of the controls received a COVID vaccination prior to pre-pandemic sampling. We were unable to verify the vaccination status of the intra-pandemic control samples, but there was very limited vaccine available in Canada prior to 31 January 2021.
[b]Cut-off for positivity: 1.9 OD ratio.
[c]Cut-off for positivity: 0.8 OD ratio.
[d]Cut-off for positivity: 700 MFI.
[e]identified by either ELISA or xMAP® assays.
[f]Cut-off for positivity: 20%.
[g]Only patients positive for SARS-CoV-2 antibodies pre- and/or intra-pandemic were assessed for neutralizing antibodies (pre-pandemic, n = 6 controls, n = 0 SLE; intra-pandemic, n = 7 controls, n = 6 SLE).
[h]Statistical testing not done due to low n.
[i]80 patients had a RT-PCR performed.

to contemporaneous controls. In contrast, over a similar observation period, others reported that 4% (4/100) SLE patients had PCR-confirmed infection, but 36% showed SARS-CoV-2 antibodies of at least one isotype, particularly IgA and IgM.[2] However, these antibodies were also detected in pre-pandemic samples and had low neutralizing activity. We also measured IgM antibodies but found them to be an unreliable indicator of SARS-CoV-2 exposure and IgA cut-offs needed to be increased according to local controls. Although there is emerging evidence of higher rates of SARS-CoV2 infections and increased odds of mortality in rheumatic disease patients,[9] it is unclear which factors influence SARS-CoV-2 infection in SLE,[2,3,7,8] However, as no pre-pandemic SARS-CoV2- IgG antibodies were observed in our SLE cohort, this seems an unlikely explanation for protection against COVID-19. Current efforts are focusing on vaccine responses in SLE.[10,11]

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Declaration of conflicting interests

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Ethics

Anonymized data was used and presented. The study was performed in accord with the World Medical Association Declaration of Helsinki.

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Supplemental material

Underlying research materials related to this manuscript (data, samples or models) are in Supplemental Tables published online or can be accessed by contacting the corresponding author.

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