Comparison of SARS-COV-2 humoral response between rheumatoid arthritis, psoriatic arthritis and spondyloarthritis patients and controls in two unvaccinated cohorts

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Abstract

Objective

To compare the humoral response after a SARS-CoV-2 infection in an inflammatory rheumatic disease population with a healthy control population in a case-control study.

Methods

Cases: between March and September 2021, all consecutive unvaccinated patients followed for rheumatoid arthritis (RA), spondyloarthritis (SpA) or psoriatic arthritis (PsA) in 16 hospitals in France were systematically screened with a SARS-CoV-2 serological test. Patients with a positive test were included in the COVID-RIC-2 cohort.
Controls: between June and July 2020, healthcare professionals working in the Toulouse University Hospital were screened with a SARS-CoV-2 serological test. Those with a positive test were included in the COVID-BIOTOUL cohort and matched to those from COVID-RIC-2 by age, sex and time-sampling on infection date.
Analyses: total SARS-CoV-2 antibody titres were centrally measured and compared.

Results

95 patients from COVID-RIC-2 (mean age 49 years, 76% females, median delay of COVID infection: 149 days) including 48 RA, 33 SpA and 14 PsA were compared to 95 matched controls. Globally, there was no significant difference of SARS-CoV-2 antibody titres between both populations: 155 Binding Antibody Units (BAU) (IQR:7-376) in COVID-RIC-2 vs. 120 BAU (IQR:35-320) in COVID-BIOTOUL. There was a trend towards higher antibody titres in patients from COVID-RIC-2 with severe COVID-19 symptoms. In COVID-RIC-2, there was no impact of age, sex, time-sampling or underlying disease on antibody titres and patients taking glucocorticoids, abatacept or rituximab trended toward having lower antibody titres after COVID-19 infection.

Conclusion

This study provides reassuring data on humoral response after COVID-19 infection in patients treated with disease-modifying anti-rheumatic drugs.

Key words

COVID-19, SARS-CoV-2, rheumatoid arthritis, spondyloarthritis, psoriatic arthritis, humoral response

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Introduction

Infection by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) was first detected at the end of 2019 (1) and became the 2019 Coronavirus disease (COVID-19) pandemic on March 2020 according to the World Health Organization (WHO) declaration (2). Risk factors for worse prognosis have been extensively documented for the general population including severe immunosuppression (3, 4). Patients with chronic inflammatory rheumatism under conventional synthetic (cs), targeted synthetic (ts) or biological (b) disease-modifying antirheumatic drugs (DMARDs) were initially been hypothesised to be at increased risk of occurrence of severe forms of COVID-19 (5, 6). However, many studies showed that the incidence and severity among patients with chronic inflammatory rheumatism were similar to the general population, except for patients taking particular treatments such as glucocorticoids, anti-CD20 and potentially JAK inhibitors (7-9).

Since early 2021, vaccination against SARS-CoV-2 considerably reduced the hospitalisation and mortality rates in the general population and became essential to limit the effects of the COVID-19 pandemic. Anti-spike (S) antibody concentrations have been shown to correlate with protection against COVID-19 (10). However, several studies showed that DMARDs could impair humoral response to vaccination in patients with inflammatory rheumatic disease, including glucocorticoids, methotrexate, leflunomide, anti-CD20, abatacept or JAK inhibitors (11-15). Furthermore, patients treated with anti-cytokine bDMARDs have a good humoral response following 2 doses of vaccine but higher decline of anti-S antibody levels in comparison to immunocompetent subjects, that can be compensated for by a third dose of vaccine (13). A recent publication studying the effect of four vaccine doses on humoral response in patients with immune-mediated inflammatory disease demonstrated a stronger humoral immune response in patients with hybrid immunisation with three vaccine doses followed by COVID-19 infection, than in infection-free patients in the vaccine group receiving four vaccine doses (12).

Although many studies investigated the impact of treatments on humoral response following SARS-CoV-2 vaccination, publications on natural immunisation following COVID-19 in patients with chronic inflammatory rheumatic disease and the impact of their treatments on humoral response are scarce.

The aim of the study was to compare the humoral response after SARS-CoV-2 infection in an unvaccinated population of rheumatoid arthritis (RA), spondyloarthritis (SpA) and psoriatic arthritis (PsA) patients to a healthy control unvaccinated population and to assess the impact of clinical factors and treatments on this humoral response.

Patients and methods

Study design

This is a case-control study comparing two observational longitudinal cohorts of unvaccinated subjects who had a positive SARS-CoV-2 serological test: the COVID-RIC-2 (COVID Chronic Inflammatory Rheumatism 2) cohort which included cases with inflammatory rheumatic disease and the COVID-BioToul cohort (COVID biological collection of Toulouse University Hospital) composed of healthy controls.

Participants

Cases from the COVID-RIC-2 cohort: between November 2020 and June 2021, all consecutive outpatients and inpatients aged 18 years or above, followed for RA, axial SpA or PsA in rheumatology departments of 16 hospitals in France, were invited to participate in a national multicentre study (COVID-RIC-1, NCT04655612), provided that they were treated with anti-inflammatory drugs (non-steroidal anti-inflammatory drugs or glucocorticoids) and/or DMARD (including targeted DMARD) and if they had never received any doses of a SARS-CoV-2 vaccine. After inclusion in COVID-RIC-1, patients underwent a local SARS-CoV-2 anti-S serological test, to assess SARS-CoV-2 prevalence in this specific population. Patients with a positive test, defined by the local SARS-CoV-2 kit, were then included between March and Septem-

ber 2021 in a 2-year longitudinal prospective study to assess long-term humoral response to SARS-CoV-2 (COVID-RIC-2, NCT04530682). The study protocol was implemented according to the Declaration of Helsinki and was approved by the Ethics Committee of Sud Méditerranée V Nice (no. CNRIPH 20.12.07:49339), all patients gave their consent.

Controls from COVID-BioToul cohort: between June 10th and July 10th 2020, 1 to 2 months after the end of the first lockdown in France, all healthcare professionals working in the Toulouse University Hospital (n=8758) underwent total serum anti-S antibody screening by enzyme linked immunosorbent assay (ELISA). At this time, none of them had been vaccinated against COVID-19 since vaccines were not available yet. Among this population, 276 had a seropositive test and were included in a two-year longitudinal study (COVID-BioToul, NCT04385108) (16). This study was approved by the French Research Ethics Committee Est-III (COVID BioToul, ID-RCB 2020-A01292-37), all patients gave their consent. Subjects with immunosuppressive treatments were excluded for this analysis.

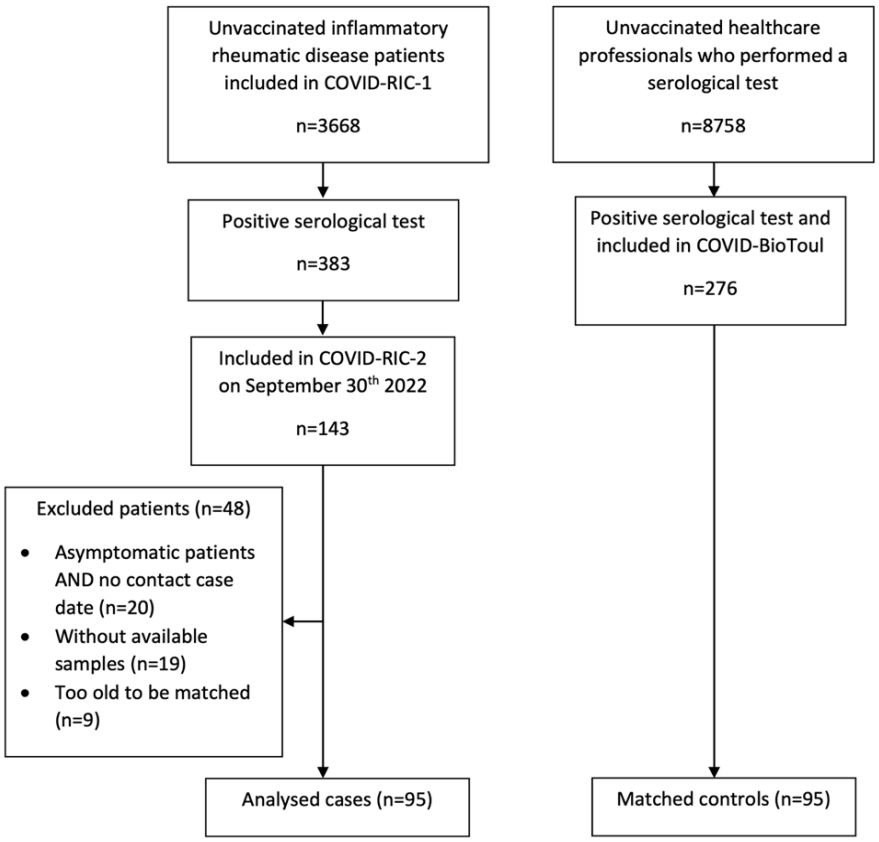

Fig. 1. Flow-chart of patient selection process

Participant selection and matching procedure

All patients with a positive serological test included in COVID-RIC-2 before September 30th 2021, with an available serum sample and a known date of COVID-19 infection (date of symptoms of COVID-19 or date of a positive SARS-CoV2 test or date of contact with a symptomatic case in asymptomatic patients) were eligible for this analysis as long as they were not vaccinated between COVID-RIC-1 and COVID-RIC-2 inclusion visits and individually matched to COVID-BioToul subjects by age (<60 *vs*. ≥60 years old), sex and the time between COVID-19 infection and the date of biological sample collection (<3, [3–6], [6–9], ≥9 months) with a ratio of 1:1. No medical file with comorbidities was available for these subjects. However, information about treatment was investigated and subjects with immunosuppressive treatments or congenital immunosuppression were excluded for this analysis.

Data collection

The following clinical characteristics of patients included in COVID-RIC-2 were collected during the inclusion visit: age, sex, date of COVID-19 infection when available, severity of COVID-19 symptoms classified into two categories: mild or severe (symptoms requiring a hospitalisation-whatever the type of hospitalisation - and oxygen therapy with or without intensive care unit hospitalisation), body mass index, smoking status, comorbidities assessed by the Charlson index, type of inflammatory rheumatic disease, disease activity score (disease activity score on 28 joints (DAS28) (17) for RA, Ankylosing Spondylitis Disease Activity Score (ASDAS) for axial SpA (18), Disease Activity in PSoriatic Arthritis (DAPSA) for PsA (19)), erythrocyte sedimentation rate (ESR), C-reactive protein (CRP) and treatments including corticosteroids, conventional synthetic DMARDs (csDMARDs), biologic DMARDs (bDMARDs) and targeted synthetic DMARDs (tsDMARDs). Anti-cytokine bDMARDs were grouped and included anti-TNF, anti-IL6R, anti-IL17 and anti-IL1. Other csDMARDs were grouped and included leflunomide, hydroxychloroquine and sulfasalazine.

Serum collection and SARS-CoV-2 antibody measurements

All patients included in COVID-RIC-2 had a biological sample collection during the inclusion visit. Sera were extracted after centrifugation and locally stored at -20°C for at least 6 months then sent to the Toulouse University Hospital for storage at -80°C. All subjects from COVID-BioToul had biological samples collected every three months after inclusion. Sera were extracted after centrifugation and were stored at -80°C at the Toulouse University Hospital. For each matched subject from COVID-BioToul, we chose the sample corresponding to the nearest time between the infection and the date of the sample collection of the COVID-RIC-2 matched patient. For example, if

Table I. demographic and disease characteristics of inflammatory rheumatic disease patients (COVID-RIC-2 cohort).

	COVID-RIC-2 patients n=95
Age, years, mean (SD)	49.4 (12.0)
Sex, number of females (%)	72 (76)
COVID-19 infection time, days, median (IQR)	149 (97-200)
Severity of COVID-19	
Mild symptoms, number (%)	79 (83)
Hospitalisation with oxygen therapy, n. (%)	11 (12)
Hospitalisation in intensive care unit, n. (%)	3 (3)
Missing data, n. (%)	2 (2)
Underlying disease	
RA[1], n. (%)	48 (50)
SpA[2], n. (%)	33 (35)
PsA[3], n. (%)	14 (15)
At least one comorbidity, n. (%)	68 (72)
Body mass index, kg/m^2, mean (SD)	27.2 (5.3)
Smoking status, number of current or past smokers, n (%)	38 (40)
Disease activity	
DAS28-CRP[4] (in 43 of the 48 RA), mean (SD)	3.00 (1.12)
ASDAS[5] (in 26 of the 33 SpA), mean (SD)	2.36 (0.70)
DAPSA[6] (in 12 of the 14 PsA)	11.88 (8.92)
Treatments	
Glucocorticoids, n. (%)	16 (17)
CsDMARDs without tsDMARD or bDMARD, n. (%)	18 (19)
Methotrexate as monotherapy, n. (%)	7 (7)
Other csDMARDs as monotherapy, n. (%)	5 (5)
Combination of csDMARDs, n. (%)	6 (6)
bDMARDs[8]	47 (51)
anti-cytokine as monotherapy, n. (%)	35 (37)
anti-cytokine in combination with a csDMARD, n. (%)	12 (13)
Rituximab[9], n. (%)	2 (2)
Abatacept[10], n. (%)	3 (3)
tsDMARDs[11]	
JAK inhibitors as monotherapy, number (%)	4 (4)
JAK inhibitors in combination with a csDMARD[10], n. (%)	6 (6)
Apremilast, n. (%)	1 (1)

1: RA: rheumatoid arthritis; 2: SpA: spondyloarthritis; 3: PsA: psoriatic arthritis; 4: DAS28-CRP: Disease Activity Score on 28 joints with CRP; 5: ASDAS: Ankylosing Spondylitis Disease Activity Score; 6: DAPSA: Disease Activity in PSoriatic Arthritis; 7: sDMARD: synthetic disease modifying anti-rheumatic drugs; 8: bDMARDs: biologic disease modifying anti-rheumatic drugs; 9: one patient received rituximab as monotherapy and one patient in combination with methotrexate; 10: one patient received abatacept as monotherapy, one patient in combination with methotrexate and one patient received methotrexate in combination with a sequence of abatacept then a JAK-inhibitor in the period between the infection date and the sample collection date; 11: tsDMARDs: targeted synthetic disease modifying anti-rheumatic drugs.

the delay between infection and blood collection was 9 months for a case from COVID-RIC-2, we used the sample of the matched-control from COVID-BioToul collected 9 months after the infection corresponding to the sample collected 6 months after the inclusion in COVID-BioToul.

Total anti-Spike SARS-CoV-2 antibodies were centrally measured in both cohorts with a quantitative enzyme-linked immunosorbent assay (ELISA) supplied by Wantai (Beijing Wantai Biological Pharmacy Enterprise Co., Ltd, China) (20) in the virology department of Toulouse University Hospital (CD) and expressed in binding antibody units (BAU) per ml using the WHO international standard (NIBSC code 20/136) (21). This ELISA kit has already been shown to have an excellent correlation with neutralising antibodies (22).

Statistical analyses

Characteristics of patients were described. Continuous variables were presented as means with standard deviation (SD) or medians with an interquartile range (IQR). Categorical variables were presented with frequencies and percentages (%).

Total SARS-CoV-2 antibodies were compared between the two groups using a Wilcoxon signed-rank test, after verifying the normality of the distribution assessed using the Shapiro-Wilk test. Sub-group analyses were performed according to patient age (below or above 60 years), sex, time between infection and the analysed sample, underlying disease (RA or SpA including PsA), COVID-19 severity (mild symptoms *versus* severe symptoms) and treatments. Biologic DMARDs were classified by mechanism of action and included anti-cytokine biologic DMARDs, abatacept and rituximab.

Results

Patients

On September 30th 2021, 3,668 unvaccinated patients were included in COVID-RIC-1, of whom 383 had a positive serological test and 143 patients accepted to be included in COVID-RIC-2 (Fig. 1). In these patients, 19 did not have available samples and it was not possible to precise the date of infection for 20 patients (no symptomatic infection and no contact with an infected subject). Furthermore, 9 patients were too old to be matched with a subject from COVID-BioToul.

The other 95 patients were included in this study and matched to 95 patients from COVID-BioToul. The 95 subjects from the COVID-BioToul cohort had a mean age of 49.3 years old (SD: 11.4), 72 were female (76%) and 30 subjects were smokers (32%). All had mild symptoms of COVID-19 or were asymptomatic. The characteristics of the subjects included in the COVID-RIC2 cohort are presented in Table I.

Comparison of total SARS-CoV-2 antibodies between inflammatory rheumatic disease patients and controls

The analyses did not show any significant differences of antibody titres between patients and matched controls (Fig. 2) in the main analysis. The sub-group analyses, depending on sex, age (above or below 60 years), time

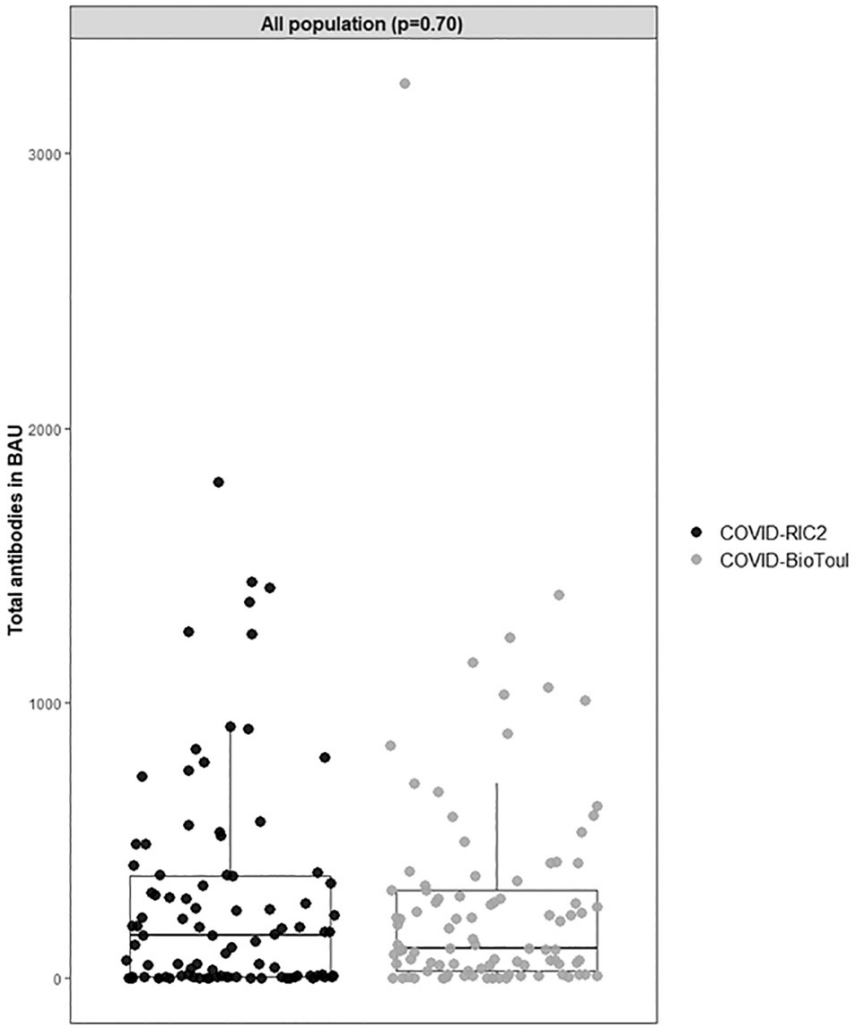

Fig. 2. Comparison of SARS-COV-2 total antibodies between patients from COVID-RIC-2 and COVID-BIOTOUL.
BAU: binding antibody units.

between infection and sample collection, underlying disease, we could not identify any differences between patients and matched controls (Fig. 3; Supplementary Table S1). A trend toward lower rates in RA patients compared with COVID-BIOTOUL controls was observed but was not statistically significant (p=0.14). In patients with severe COVID-19 symptoms (n=14) leading to hospitalisation for oxygen therapy or resuscitation, higher titres of SARS-CoV-2 antibodies were observed compared with matched controls, but the difference was not statistically significant (Fig. 3; Suppl. Table S1).

Effect of treatment on SARS-CoV-2 antibodies in inflammatory rheumatic disease patients

After stratification by treatment exposure during COVID-19 infection, we did not observe any difference between COVID-RIC-2 patients and their matched healthy controls from COVID-BioToul after stratification by treatment type or within the COVID-RIC-2 population according to treatment (Fig. 4; Suppl. Table S2), except for patients from COVID-RIC2 treated with tsDMARD (n=10) who had higher antibody titres in comparison with matched controls (median titre in COVID-RIC2: 454 BAU *vs*. 117 BAU in COVID-BIOTOUL, p=0.03). Furthermore, in patients treated with abatacept (n=3) or rituximab (n=2), the total antibody titres were very low (respectively, median: 6 BAU (IQR: 3–187) in abatacept-treated patients and 4 BAU (0–7) in rituximab-treated patients). Interestingly, antibody titres in JAK-inhibitor treated patients were significantly higher than matched-controls form COVID-BIOTOUL (p=0.03 for comparison, Fig. 4). There was no association between methotrexate use or other csDMARD use in the COVID-RIC2 population and antibody titres, in comparison with patients from COVID-BIOTOUL (Fig. 4; Suppl. Table S2). There was no correlation between the dose of methotrexate and antibody titres (Spearman correlation: r= -0.26, p=0.2). When analysing the COVID-RIC2 population, we did not observe any difference in antibody titres between methotrexate users (n=29) or non-users (n=66) (median titres in methotrexate users: 136 BAU *versus* 165 BAU in non-users, p=0.8). In patients using anti-cytokine bDMARDs, the use of methotrexate was not associated with a decrease in antibody titres (in n=10 patients, median titres: 138BAU) in comparison with patients using anti-cytokine bDMARDs as monotherapy (in n=37 patients, median titres: 49BAU, p=0.2 for the comparison). We also observed a trend toward lower total antibody titres in patients receiving glucocorticoids in comparison with their matched healthy controls (Fig. 4; Suppl. Table S2) but this difference was not significant (p-value for comparison = 0.07). Furthermore, in COVID-RIC2 patients, there was no correlation between cumulative glucocorticoid dose and antibody titres (Spearman correlation: r=-0.11, p=0.7).

Discussion

This study aimed to compare the SARS-CoV-2 total antibody titres in patients with RA, PsA and SpA to matched healthy controls after COVID-19 infection in an unvaccinated population. We could not identify any difference between the two populations, even after stratification by age, sex, time since infection or underlying disease. However, patients with RA tended to have lower titres compared with matched controls and this may be related to some therapies used preferentially in RA than in SpA such as RTX, ABA, and steroids. Indeed, we observed a trend toward higher SARS-CoV-2 total antibody titres in patients who had a severe COVID-19 disease, and lower titres in

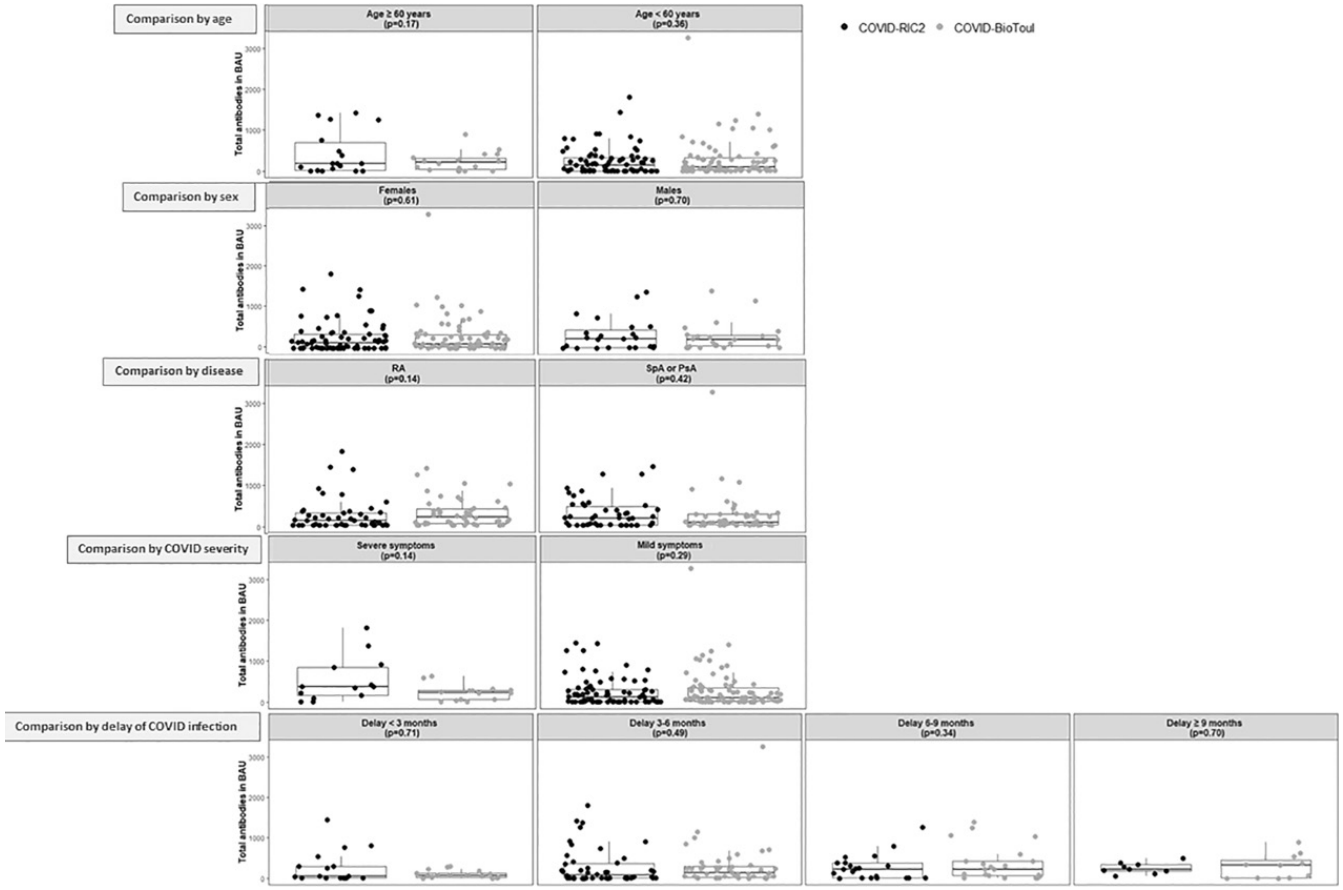

Fig. 3. Comparison of SARS-COV-2 total antibodies between patients from COVID-RIC-2 and COVID-BIOTOUL according to age, sex, underlying disease, COVID-19 severity, delay between infection and blood collection.
RA: rheumatoid arthritis; SpA: spondyloarthritis; PsA: psoriatic arthritis; BAU: binding antibody units

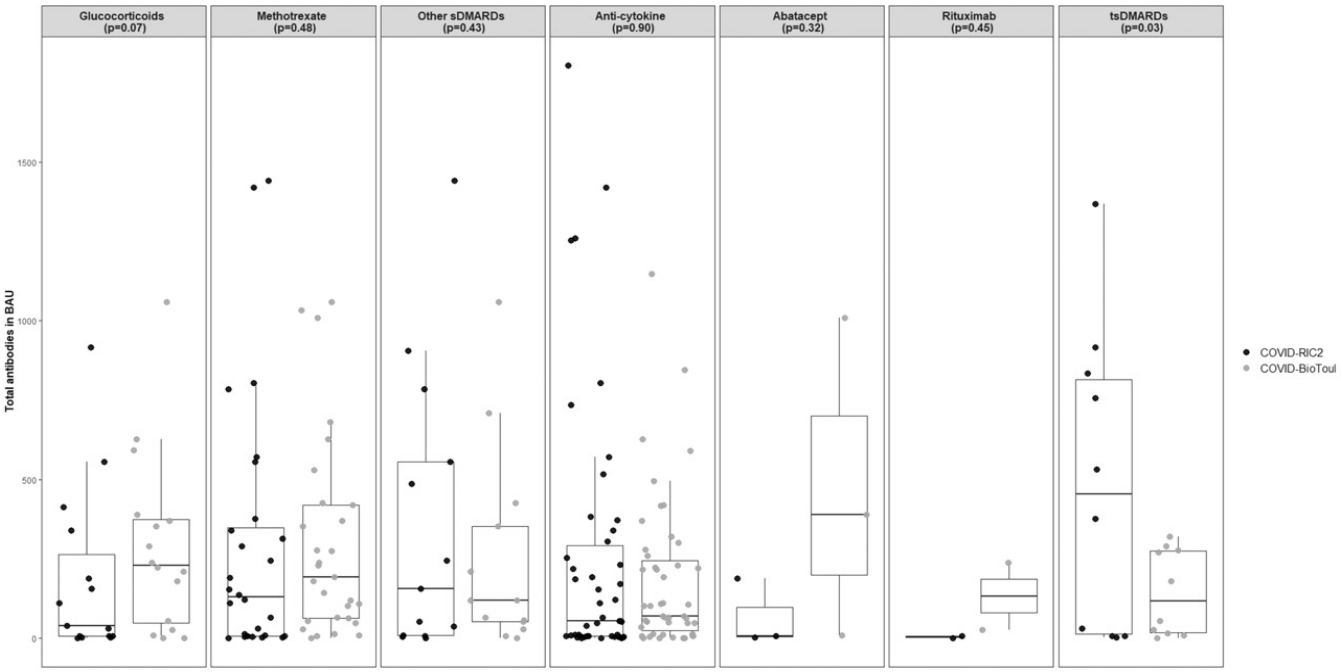

Fig. 4. Comparison of SARS-COV-2 total antibodies between patients from COVID-RIC-2 and COVID-BIOTOUL according to treatments during and after infection.
Other sDMARDs: other synthetic disease-modifying anti-rheumatic drugs included sulfasalazine, leflunomide, hydroxychloroquine.
tsDMARDs: targeted synthetic disease-modifying anti-rheumatic drugs included JAK inihibitors and apremilast.
BAU: binding antibody units.

patients exposed to glucocorticoids, as well as in the very limited population of patients treated with rituximab or abatacept.

At the beginning of the pandemic, concerns raised about the risk of severe COVID-19 infection in patients treated with inflammatory chronic diseases (5, 6). Several studies have sought to investigate the factors associated with SARS-CoV-2 antibody seroconversion in patients with immune-mediated inflammatory disease prior to the vaccination campaigns (23-29). Overall, the studies showed a lower prevalence of SARS-CoV-2 antibodies in patients with inflammatory disease in comparison with the general population. However, many factors might have an impact on seroconversion, the principal one being risk of SARS-CoV-2 exposure. Indeed, patients with immunosuppressive therapies were aware of the potential risk of a severe form of COVID-19 and were more likely to follow protection measures and socially isolate during the pandemic waves (23). In patients with documented SARS-CoV-2 infection, overall, the SARS-CoV-2 seroconversion rates were similar to those of the general population (24, 25, 28). However, one study showed that bDMARD exposure was associated with a low seroconversion rate, although the data were not confirmed by any other studies (28).

Only two studies measured SARS-CoV-2 antibodies in this specific population. Cruz-Machado *et al*. (24) assessed SARS-CoV-2 humoral response in 65 patients with rheumatic and musculoskeletal disease and a documented COVID-19 infection by a positive PCR test or a serological test. Among them, 56 had a positive serological test with SARS-CoV-2 antibody titres compared with a control population of 130 subjects, matched for age, sex and sample timing. In this study, all patients treated with glucocorticoids or rituximab had SARS-CoV-2 antibodies. However, they did not compare the antibody titres within the population according to treatment. Favalli *et al*. (28) recruited 66 patients with rheumatic or musculoskeletal disease and a positive SARS-CoV-2 serological test who were compared with 13 individuals with a documented SARS-CoV-2 infection as controls. The magnitude of the antibody response to SARS-CoV-2 proteins S and N was comparable between patients treated with b/tsDMARDs and csDMARDs and not impaired by glucocorticoid exposure. The anti-S antibody titres of patients were similar to the titres of the control group. In another study (27), the same group investigated humoral response after SARS-CoV-2 infection according to the different mechanisms of action of bDMARDs. Interestingly, they showed that patients treated with abatacept had a lower antibody response in comparison with control subjects. In this study, there were an insufficient number of patients to investigate the effect of rituximab on SARS-CoV-2 humoral response. The principal limitations of the study were the small number of control cases and no matching for age or time-of-sample.

As observed in studies assessing humoral response after vaccination in patients with immune-mediated inflammatory disease, we showed a trend toward a lower humoral response after SARS-CoV-2 infection in patients with RA and in patients taking glucocorticoids, rituximab or abatacept which are drugs preferentially used in RA. Furthermore, as has been shown by vaccination studies, anti-cytokine bDMARDs do not appear to have an impact on humoral response after SARS-CoV-2 infection (13). Vaccination studies have also shown an impact for DMARDs on decline of anti-S antibody levels in comparison with immunocompetent subjects (13). In this study, we did not observe any impact on the time between the SARS-CoV-2 infection onset and sampling on antibody titres, as was also shown by Cruz-Machado *et al*. (24). Vaccination studies have also shown that even when humoral response is impaired by an immunosuppressant such as rituximab (30) or methotrexate (31), while T-cell response appears to be maintained (14). While T-cell response seems to be maintained after a COVID-19 infection or vaccination, we still need standardised tests able to predict the risk of new COVID-19 infection in patients (32).

This study has several strengths. The first is the originality of the recruitment method: we systematically performed a serological test on all outpatients and inpatients coming to 16 hospitals in France for follow-up of their inflammatory rheumatic disease. This method of recruitment allowed us to include symptomatic – irrespective of symptom intensity – and asymptomatic cases while the majority of studies included only patients with symptomatic COVID-19. The second strength of this study was the creation of a control group of healthcare workers with the same method of recruitment, *i.e*. a serological test, with the same time-sampling and with an appropriate matching method. The major limitation of this study was a lower-than-expected sample size. The main reason was the accelerated vaccination campaign in France while the recruiting period of this study was in 2021. By the end of 2021, the majority of patients were vaccinated and could not be included in this study. Because of this small sample size, we could not identify any statistical differences according to treatment. However, as expected when reading vaccination studies, we observed very low SARS-CoV-2 antibody titres in patients treated with abatacept or rituximab and a statistical test may not be necessary to demonstrate the potential impact of these treatments on humoral response. We also had very few patients aged 60 and above, because we had no matching controls, which may impair the extrapolation of our findings to an older population. Finally, the majority of patients had mild symptoms and some of them were asymptomatic. In this case we used the date of a contact with an infected person as the potential date of the infection. This might have introduced a bias since we were not sure of the exact date of infection. Moreover, patients could have several infections over time that might increase the antibody titres.

This study provides reassuring data on humoral response after a COVID-19 infection in unvaccinated patients treated with DMARDs for a RA, PsA or SpA disease. However, as observed after SARS-CoV-2 vaccination, patients treated with abatacept or rituximab appear to have an altered humoral res-

ponse. Additional studies are needed to assess the impact of hybrid vaccination in comparison with conventional vaccination in this specific population.

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References

1. WU F, ZHAO S, YU B *et al.*: A new coronavirus associated with human respiratory disease in China. *Nature* 2020; 579(7798): 265-69. https://doi.org/10.1038/s41586-020-2008-3
2. GHEBREYESUS TA: WHO Director-General's opening remarks at the media briefing on COVID-19 - 11 March 2020: WHO; 2020 [Available from: https://www.who.int/director-general/speeches/detail/who-director-general-s-opening-remarks-at-the-media-briefing-on-covid-19---11-march-2020
3. BELSKY JA, TULLIUS BP, LAMB MG, SAYEGH R, STANEK JR, AULETTA JJ: COVID-19 in immunocompromised patients: A systematic review of cancer, hematopoietic cell and solid organ transplant patients. *J Infect* 2021; 82(3): 329-38. https://doi.org/10.1016/j.jinf.2021.01.022
4. ZHENG Z, PENG F, XU B *et al.*: Risk factors of critical & mortal COVID-19 cases: A systematic literature review and meta-analysis. *J Infect* 2020; 81(2): e16-e25. https://doi.org/10.1016/j.jinf.2020.04.021
5. PABLOS JL, GALINDO M, CARMONA L *et al.*: Clinical outcomes of hospitalised patients with COVID-19 and chronic inflammatory and autoimmune rheumatic diseases: a multicentric matched cohort study. *Ann Rheum Dis* 2020; 79(12): 1544-49. https://doi.org/10.1136/annrheumdis-2020-218296
6. ZANETTI A, CARRARA G, LANDOLFI G *et al.*: Increased COVID-19 mortality in patients with rheumatic diseases: results from the CONTROL-19 study by the Italian Society for Rheumatology. *Clin Exp Rheumatol* 2022; 40(11): 2038-43. https://doi.org/10.55563/clinexprheumatol/fmyozh
7. AKIYAMA S, HAMDEH S, MICIC D, SAKURABA A: Prevalence and clinical outcomes of COVID-19 in patients with autoimmune diseases: a systematic review and meta-analysis. *Ann Rheum Dis* 2021; 80(3): 384-91. https://doi.org/10.1136/annrheumdis-2020-218946
8. SHIN JI, KIM SE, LEE MH *et al.*: COVID-19 susceptibility and clinical outcomes in autoimmune inflammatory rheumatic diseases (AIRDs): a systematic review and meta-analysis. *Eur Rev Med Pharmacol Sci* 2022; 26(10): 3760-70. https://

doi.org/10.26355/eurrev_202205_28873
9. SPARKS JA, WALLACE ZS, SEET AM *et al*.: Associations of baseline use of biologic or targeted synthetic DMARDs with COVID-19 severity in rheumatoid arthritis: Results from the COVID-19 Global Rheumatology Alliance physician registry. *Ann Rheum Dis* 2021; 80(9): 1137-46. https://doi.org/10.1136/annrheumdis-2021-220418
10. FENG S, PHILLIPS DJ, WHITE T *et al*.: Correlates of protection against symptomatic and asymptomatic SARS-CoV-2 infection. *Nat Med* 2021; 27(11):2032-40. https://doi.org/10.1038/s41591-021-01540-1
11. AUROUX M, LAURENT B, COSTE B *et al*.: Serological response to SARS-CoV-2 vaccination in patients with inflammatory rheumatic disease treated with disease modifying anti-rheumatic drugs: A cohort study and a meta-analysis. *Joint Bone Spine* 2022; 89(5): 105380. https://doi.org/10.1016/j.jbspin.2022.105380
12. BJORLYKKE KH, ORBO HS, TVETER AT *et al*.: Four SARS-CoV-2 vaccine doses or hybrid immunity in patients on immunosuppressive therapies: a Norwegian cohort study. *Lancet Rheumatol* 2023; 5(1): e36-e46. https://doi.org/10.1016/S2665-9913(22)00330-7
13. FURER V, EVIATAR T, FREUND T *et al*.: Immunogenicity induced by two and three doses of the BNT162b2 mRNA vaccine in patients with autoimmune inflammatory rheumatic diseases and immunocompetent controls: a longitudinal multicentre study. *Ann Rheum Dis* 2022; 81(11): 1594-602. https://doi.org/10.1136/ard-2022-222550
14. JYSSUM I, KARED H, TRAN TT *et al*.: Humoral and cellular immune responses to two and three doses of SARS-CoV-2 vaccines in rituximab-treated patients with rheumatoid arthritis: a prospective, cohort study. *Lancet Rheumatol* 2022; 4(3): e177-e87. https://doi.org/10.1016/S2665-9913(21)00394-5
15. SYVERSEN SW, JYSSUM I, TVETER AT *et al*.: Immunogenicity and safety of a three-dose SARS-CoV-2 vaccination strategy in patients with immune-mediated inflammatory diseases on immunosuppressive therapy. *RMD Open* 2022; 8(2). https://doi.org/10.1136/rmdopen-2022-002417
16. DIMEGLIO C, HERIN F, MIEDOUGE M, MARTIN-BLONDEL G, SOULAT JM, IZOPET J: Protection of healthcare workers against Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) reinfection. *Clin Infect Dis* 2021; 73(7): 1323-24. https://doi.org/10.1093/cid/ciab069
17. ALETAHA D, LANDEWÉ R, KARONITSCH *et al*.: Reporting disease activity in clinical trials of patients with rheumatoid arthritis: EULAR/ACR collaborative recommendations. *Arthritis Rheum* 2008; 59(10): 1371-77. https://doi.org/10.1002/art.24123
18. LUKAS C, LANDEWE R, SIEPER J *et al*.: Development of an ASAS-endorsed disease activity score (ASDAS) in patients with ankylosing spondylitis. *Ann Rheum Dis* 2009; 68(1): 18-24. https://doi.org/10.1136/ard.2008.094870
19. SCHOELS M, ALETAHA D, FUNOVITS J, KAVANAUGH A, BAKER D, SMOLEN JS: Application of the DAREA/DAPSA score for assessment of disease activity in psoriatic arthritis. *Ann Rheum Dis* 2010; 69(8): 1441-47. https://doi.org/10.1136/ard.2009.122259
20. DIMEGLIO C, HERIN F, MIEDOUGE M *et al*.: Screening for SARS-CoV-2 antibodies among healthcare workers in a university hospital in southern France. *J Infect* 2021; 82(1): e29-32. https://doi.org/10.1016/j.jinf.2020.09.035
21. KUMAR A, BERNASCONI V, MANAK M, DE ALMEIDA ARANHA AP, KRISTIANSEN PA: The CEPI centralised laboratory network: supporting COVID-19 vaccine development. *Lancet* 2021; 397(10290): 2148-49. https://doi.org/10.1016/S0140-6736(21)00982-x
22. CHAPUY-REGAUD S, MIEDOUGE M, ABRAVANEL *et al*.: Evaluation of three quantitative anti-SARS-CoV-2 antibody immunoassays. *Microbiol Spectr* 2021; 9(3): e0137621. https://doi.org/10.1128/spectrum.01376-21
23. AMMITZBOLL C, ANDERSEN JB, VILS SR *et al*.: Isolation, behavioral changes, and low seroprevalence of SARS-CoV-2 antibodies in patients with systemic lupus erythematosus or rheumatoid arthritis. *Arthritis Care Res* (Hoboken) 2022; 74(11): 1780-85. https://doi.org/10.1002/acr.24716
24. CRUZ-MACHADO AR, BARREIRA SC, BANDEIRA M *et al*.: Risk factors for infection, predictors of severe disease, and antibody response to COVID-19 in patients with inflammatory rheumatic diseases in Portugal - a multicenter, nationwide study. *Front Med* (Lausanne) 2022; 9: 901817. https://doi.org/10.3389/fmed.2022.901817
25. D'SILVA KM, SERLING-BOYD N, HSU TY, SPARKS JA, WALLACE ZS: SARS-CoV-2 antibody response after COVID-19 in patients with rheumatic disease. *Ann Rheum Dis* 2021; 80(6): 817-19. https://doi.org/10.1136/annrheumdis-2020-219808
26. EVIATAR T, FURER V, POLACHEK A *et al*.: Seroprevalence of SARS-CoV-2 antibodies in patients with autoimmune inflammatory rheumatic diseases. *Clin Exp Rheumatol* 2022; 40(7): 1299-305. https://doi.org/10.55563/clinexprheumatol/ykin5p
27. FAVALLI A, FAVALLI EG, GOBBINI A *et al*.: Immunosuppressant treatment in rheumatic musculoskeletal diseases does not inhibit elicitation of humoral response to SARS-CoV-2 infection and preserves effector immune cell populations. *Front Immunol* 2022; 13: 873195. https://doi.org/10.3389/fimmu.2022.873195
28. FAVALLI EG, GOBBINI A, BOMBACI M *et al*.: The impact of anti-rheumatic drugs on the seroprevalence of anti-SARS-CoV-2 antibodies in a cohort of patients with inflammatory arthritis: the MAINSTREAM study. *Front Med* (Lausanne) 2022; 9: 850858. https://doi.org/10.3389/fmed.2022.850858
29. GROS C, MARIAGGI AA, MERITET JF *et al*.: Systematic assessment of the humoral response against SARS-CoV-2 in a French cohort of 283 patients with rheumatic diseases. *Joint Bone Spine* 2022; 89(3): 105312. https://doi.org/10.1016/j.jbspin.2021.105312
30. CONNOLLY CM, KOENIG D, RAVI SN *et al*.: Correspondence on "SARS-CoV-2 vaccination in rituximab-treated patients: evidence for impaired humoral but inducible cellular immune response" by Bonelli et al. *Ann Rheum Dis* 2021; 80(10): e164. https://doi.org/10.1136/annrheumdis-2021-220972
31. MOYON Q, STERLIN D, MIYARA M *et al*.: BNT162b2 vaccine-induced humoral and cellular responses against SARS-CoV-2 variants in systemic lupus erythematosus. *Ann Rheum Dis* 2022; 81(4): 575-83. https://doi.org/10.1136/annrheumdis-2021-221097
32. SETTE A, SIDNEY J, CROTTY S: T cell responses to SARS-CoV-2. *Annu Rev Immunol* 2023; 41: 343-73. https://doi.org/10.1146/annurev-immunol-101721-061120