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Adalimumab and Infliximab Impair SARS-CoV-2 Antibody Responses: Results from a Therapeutic Drug Monitoring Study in 11 422 Biologic-Treated Patients

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Abstract

Background and Aims: Infliximab attenuates serological responses to SARS-CoV-2 infection. Whether this is a class effect, or if anti-tumour necrosis factor [anti-TNF] level influences serological responses, remains unknown.

Methods: Seroprevalence and the magnitude of SARS-CoV-2 nucleocapsid antibody responses were measured in surplus serum from 11 422 (53.3% [6084] male; median age 36.8 years)

patients with immune-mediated inflammatory diseases, stored at six therapeutic drug monitoring laboratories between January 29 and September 30, 2020. Data were linked to nationally held SARS-CoV-2 PCR results to July 11, 2021.

Results: Rates of PCR-confirmed SARS-CoV-2 infection were similar across treatment groups. Seroprevalence rates were lower in infliximab- and adalimumab- than vedolizumab-treated patients (infliximab: 3.0% [178/5893], adalimumab: 3.0% [152/5074], vedolizumab: 6.7% [25/375], $p = 0.003$). The magnitude of SARS-CoV-2 reactivity was similar in infliximab- vs adalimumab-treated patients (median 4.30 cut-off index [COI] [1.94–9.96] vs 5.02 [2.18–18.70], $p = 0.164$), but higher in vedolizumab-treated patients (median 21.60 COI [4.39–68.10, $p < 0.004$). Compared to patients with detectable infliximab and adalimumab drug levels, patients with undetectable drug levels [<0.8 mg/L] were more likely to be seropositive for SARS-CoV-2 antibodies. One-third of patients who had PCR testing prior to antibody testing failed to seroconvert, all were treated with anti-TNF. Subsequent positive PCR-confirmed SARS-CoV-2 was seen in 7.9% [12/152] of patients after a median time of 183.5 days [129.8–235.3], without differences between drugs.

Conclusion: Anti-TNF treatment is associated with lower SARS-CoV-2 nucleocapsid seroprevalence and antibody reactivity when compared to vedolizumab-treated patients. Higher seropositivity rates in patients with undetectable anti-TNF levels support a causal relationship, although confounding factors, such as combination therapy with a immunomodulator, may have influenced the results.

Key Words: CLARITY; biologic; infliximab; adalimumab; vedolizumab; COVID-19; immunosuppression; vaccination; inflammatory bowel disease

1. Introduction

The increased transmissibility of the dominant delta variant of SARS-CoV-2 means that > 80% of the UK population will need to be fully vaccinated to achieve herd immunity.[1] Anti-tumour necrosis factor [anti-TNF] drugs impair protective immunity following pneumococcal,[2] influenza[3,4] and viral hepatitis[5] vaccinations and increase the risk of serious respiratory infections.[6] By suppressing immune responses, biologic and immunosuppression therapies increase the reservoir for viral transmission and have been implicated in the evolution and emergence of novel variants of SARS-CoV-2.[7]

We have recently reported that seroprevalence, seroconversion rates and the magnitude of SARS-CoV-2 nucleocapsid [N] antibodies following SARS-CoV-2 infection are reduced in patients with inflammatory bowel disease [IBD] treated with infliximab compared to vedolizumab.[8] Vedolizumab is a gut-selective anti-integrin α4β7 monoclonal antibody and, unlike anti-TNF therapy, is not associated with increased susceptibility to systemic infection or attenuated serological responses to SARS-CoV-2 vaccination.[9] Because we observed similar rates of SARS-CoV-2 infection and hospitalizations between infliximab- and vedolizumab-treated patients, our findings suggest that infliximab directly influences the serological response to SARS-CoV-2 infection. In the same cohort of IBD patients, SARS-CoV-2 spike [S] antibody levels and rates of seroconversion were also lower after a single-dose of either the BNT162b2 [Pfizer] or ChAdOx1 nCoV-19 [AstraZeneca/Oxford] vaccines in patients treated with infliximab than vedolizumab.[10]

Whether antibody responses following SARS-CoV-2 infection are also impaired in patients treated with other biopharmaceuticals, including other anti-TNF therapies such as adalimumab, and whether biologic drug levels influence the magnitude of SARS-CoV-2 [N] antibody responses, remain unknown.

2. Objectives

In patients with immune-mediated inflammatory diseases [IMIDs], we aimed to define whether biologic class impacted the:

[i] seroprevalence of SARS-CoV-2 antibodies
[ii] magnitude of SARS-CoV-2 antibodies, stratified by biologic drug levels
[iii] seroconversion and subsequent positive polymerase chain reaction (PCR)-confirmed SARS-CoV-2

3. Methods

3.1. Study design and population

CLARITY IBD is a UK-wide, multicentre, observational cohort study investigating the impact of biologics and/or concomitant immunomodulators on SARS-CoV-2 acquisition, illness and immunity in patients with IBD [www.clarityibd.org].

Here, we report data from a retrospective cohort of patients with IMIDs who had serum stored following routine therapeutic drug monitoring [TDM] tests as part of clinical care during the early phase of the COVID-19 pandemic. Surplus serum samples were obtained from six UK laboratories [Barts Health NHS Trust, NHS Greater Glasgow and Clyde, Guy's and St Thomas' NHS Foundation Trust, North West London Pathology, Royal Devon and Exeter NHS Foundation Trust, and Royal Wolverhampton NHS Trust] that offer TDM for infliximab, adalimumab, ustekinumab or vedolizumab. Samples archived between January 29, 2020, shortly after the first case of COVID-19 was reported in the UK,[11] to September 30, 2020 were included. Surplus samples were transferred to the Academic Department of Blood Sciences at the Royal Devon and Exeter NHS Foundation Trust and serum was tested for SARS-CoV-2 [N] antibodies. Samples with adequate linked clinical data, of more than 150 µL, the minimum volume required to undertake the assay, and not contaminated by haemolysis, were processed.

3.2. Outcomes

The primary outcome was the proportion of patients with a positive SARS-CoV-2 [N] antibody test. Secondary outcomes were the impact of biologic drug levels on seropositivity and the magnitude of

SARS-CoV-2 antibodies, and seroconversion and rates of subsequent positive PCR-confirmed SARS-CoV-2.

3.3. Variables and case definition

We recorded the patient's national patient identifier (National Health Service [NHS] number or Community Health Index [CHI]), sex, date of birth, postcode, date of serum sample and referring hospital. Where missing, these data were obtained from the NHS Digital Data Access Request Service. The following variables, where available from the TDM requisition form, were also recorded: diagnosis (IBD [Crohn's disease, ulcerative colitis or IBD-unclassified], non-IBD [ankylosing spondylitis, Bechet's disease, hidradenitis suppurativa, juvenile idiopathic arthritis, malignancy, psoriatic arthritis, psoriasis, rheumatoid arthritis, sarcoidosis or systemic lupus erythematosus], treatment and results from TDM [biologic drug and anti-drug antibody testing] performed at the referring site.

We linked our data by NHS number or CHI to data held by Public Health England, Scotland and Wales, which archive dates and results of SARS-CoV-2 PCR tests undertaken. Confirmed cases were patients with a positive PCR test to SARS-CoV-2. Due to differences in nationally held public health databases, we received: all negative and positive PCR test results from Public Health Wales [March 23, 2020 to May 4, 2021] and Public Health Scotland [March 14, 2020 to July 11, 2021], and all negative PCR test results up to and including the first positive PCR test result from Public Health England [February 26, 2020 to April 18, 2021].

3.4. Laboratory methods

We used the Roche Elecsys Anti-SARS-CoV-2 [N] immunoassay to detect antibodies to SARS-CoV-2. This sandwich electrochemiluminescence immunoassay uses a recombinant protein of the nucleocapsid antigen for determination of antibodies against SARS-CoV-2.[12] The electrochemiluminescence signal from a negative and positive calibrator are assigned a value of 0.8 and 1.2, respectively, and a cut-off index [COI] is set at a signal equivalent to 1. The manufacturer reports clinical sensitivity of 99.5% [97.0–100] ≥ 14 days post-PCR confirmation and specificity of 99.8% [95% CI 99.7–99.9].[12]

In-house assay validation experiments demonstrated the intra- and inter-assay coefficient of variation were 2.2 and 7.0%, respectively. No effect was observed on recovery of SARS-CoV-2 antibodies following four freeze/thaw cycles. SARS-CoV-2 antibodies were stable in uncentrifuged blood and serum at ambient temperature for up to 7 days, permitting postal transport from research sites to the central laboratory. No analytical interference was observed for the detection of SARS-CoV-2 antibodies with infliximab, adalimumab or vedolizumab up to 10 000, 8000 and 60 000 mg/L, respectively, or with anti-drug antibodies to infliximab, adalimumab or vedolizumab up to 400, 200 and 38 AU/mL respectively. For anti-TNF-treated patients, absence of drug was defined using a cut-off of < 0.8 mg/L.[13] For vedolizumab-treated patients, absence of drug was defined using a cut-off of < 3.1 mg/L. Anti-drug antibody levels, recorded as positive or negative, were supplied by the referring laboratory.

3.5. Statistical analysis

A priori sample size calculations were not undertaken for this study; rather we collected all available samples saved through the early phase of the pandemic. Statistical analyses were undertaken in R 4.0.5 [R Foundation for Statistical Computing]. All tests were two tailed and p-values < 0.05 were considered significant. We included patients with missing clinical data in analyses for which they had data and have specified the denominator for each variable. Continuous data were reported as median and interquartile range [IQR], and discrete data as numbers and percentages, unless otherwise stated. We used patients' postcodes to assign them to one of ten UK administrative regions and present seroprevalence rates mapped to these regions. We also used postcodes to derive patients' income and employment deprivation scores using combined English and Welsh data from 2019[14] and Scottish data from 2020.[15]

Seroprevalence of SARS-CoV-2 antibodies was estimated as the proportion of samples with a positive SARS-CoV-2 antibody result. Univariable analyses, using Fisher's exact and Mann–Whitney U tests, were used to identify demographic and treatment-related factors, including TDM, associated with SARS-CoV-2 seropositivity. We explored the magnitude of antibody reactivity using density plots, stratified by drug exposure among patients with a positive SARS-CoV-2 antibody result. We performed a sensitivity analysis restricting the cohort to patients treated with an anti-TNF who were known to have IBD, and all vedolizumab-treated patients, which is only licensed in the UK for treatment of IBD.

3.6. Ethics and role of the funding source

CLARITY IBD is an investigator-led, UK National Institute for Health Research COVID-19 urgent public health study, funded by the Royal Devon and Exeter NHS Foundation Trust, and Hull University Teaching Hospital NHS Trust, and by unrestricted educational grants from F. Hoffmann-La Roche AG [Switzerland], Biogen Inc. [USA], Celltrion Healthcare [South Korea], Takeda [UK] and Galapagos NV [Belgium]. None of our funding bodies had any role in study design, data collection, or analysis, writing or decision to submit for publication.

Data were provided to the Royal Devon and Exeter NHS Foundation trust under Regulation 3 [4] of the Health Service Control of Patient Information [COPI] Regulations 2002 to facilitate a COVID-19 research purpose. The Surrey Borders Research Ethics committee approved the study [REC reference: REC 20/HRA/3114] in September 2020. The sponsor was the Royal Devon and Exeter NHS Foundation Trust. The protocol is available online at https//www.clarityibd.org. The study was registered with the ISRCTN registry, ISRCTN45176516.

4. Results

In total, 14 106 surplus samples were received; 4.2% of samples [597/14 106] were excluded because of insufficient demographic or clinical information, insufficient volume or haemolysis, leaving 13 509 samples from 11 600 patients to be analysed. Of these, 1.5% [178/11 600] patients did not have adequate treatment details [n = 176] or were treated with etanercept [n = 2], and therefore excluded. In total, 13 316 samples from 11 422 unique patients were included in the final analysis [Figure 1; Supplementary Figures 1 and 2].

4.1 Patient characteristics

Overall, 53.3% [6084/11 422] of patients were male with a median age of 36.8 years [IQR 25.5–51.5]. The median income deprivation score was 0.10 [IQR 0.06–0.17]. Diagnosis was not recorded in 79.3% [9061/11 422] of patients; 19.5% [2231/11 422] of patients had IBD and 1.1% [130/11 422] had a non-IBD diagnosis. In total, 51.6% [5893/11 422] of patients were treated with infliximab, 44.4% [5074/11 422] adalimumab, 3.3% [375/11 422] vedolizumab and 0.7% [80/11 422] ustekinumab. Baseline characteristics stratified by biologic drug are shown in Table 1.

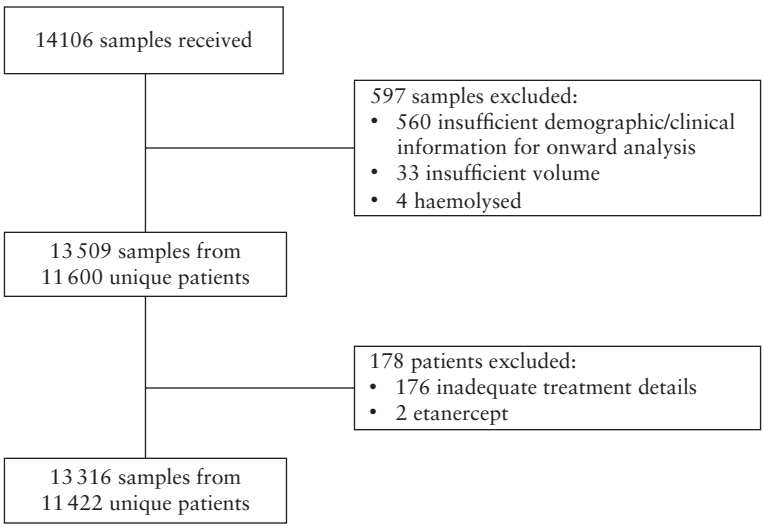

Figure 1. Study profile.

In total, 60.2% [6875/11 422] patients had undergone PCR testing across England, Scotland and Wales, of whom 11.2% [770/6875] had a positive PCR test. No differences were observed in the proportion of patients who tested positive for SARS-CoV-2 (infliximab: 11.2% [402/3600], adalimumab: 11.4% [342/2990], ustekinumab: 4.2% [2/48], vedolizumab: 10.1% [24/237], $p = 0.467$).

4.2 SARS-CoV-2 seroprevalence

Seropositivity to SARS-CoV-2 was first observed on February 3, 2020 and seroprevalence increased to 3.1% by September 30, 2020 [Supplementary Figure 3]. Univariable analyses demonstrated that the proportion of patients with a positive SARS-CoV-2 antibody test was lower in anti-TNF- and ustekinumab-treated patients than vedolizumab-treated patients (infliximab: 3.0% [178/5893], adalimumab: 3.0% [152/5074], ustekinumab: 1.3% [1/80], vedolizumab: 6.7% [25/375], $p = 0.003$) [Table 2]. The magnitude of SARS-CoV-2 reactivity was similar in infliximab- vs adalimumab-treated patients (median 4.30 COI [1.94–9.96] vs 5.02 [2.18–18.70], $p = 0.164$), and for both drugs, was lower than the vedolizumab-treated group (median 21.60 COI [4.39–68.10], $p = 0.004$) [Figures 2A and 3]. Seropositivity was also associated with UK region and calendar month [Table 2; Supplementary Figure 4].

4.3. Sensitivity analysis

The diagnosis of IBD was recorded in 19.6% [1153/5893] and 18.2% [923/5074] of infliximab- and adalimumab-treated patients, respectively. Univariable analyses demonstrated that the proportion of patients with a positive SARS-CoV-2 antibody test was lower in anti-TNF- than vedolizumab-treated patients (infliximab: 2.3% [27/1153]; adalimumab: 2.3% [21/923]; vedolizumab: 6.7% [25/375], $p < 0.001$). The magnitude of SARS-CoV-2 reactivity was similar in infliximab- vs adalimumab-treated patients (median 2.66 COI [1.65–7.29] vs 4.38 [2.31–16.20], $p = 0.127$), and for both drugs, was lower than the vedolizumab-treated group (median 21.60 COI [4.39–68.10], $p = 0.004$).

4.4. Impact of biologic drug level on seropositivity and magnitude of SARS-CoV-2 antibodies

Of 11 422 patients in the study, 95.6% [5636/5893] infliximab-, 97.0% [4923/5074] adalimumab- and 89.1% [334/375] vedolizumab-treated patients had biologic drug level data available for analysis. Overall, 12.1% [681/5636] of infliximab- and 7.0% [347/4923] of adalimumab-treated patients had undetectable drug levels, of whom 54.8% [373/681] and 39.2% [136/347] had detectable anti-infliximab and anti-adalimumab antibodies, respectively. In total, 10.8% [36/334] of vedolizumab-treated patients had undetectable drug levels. Compared to patients with detectable infliximab drug levels, patients with undetectable drug levels [<0.8 mg/L] were more likely to be seropositive for SARS-CoV-2 antibodies (odds ratio [OR] 1.73, 95% confidence interval [CI] 1.13–2.56, $p = 0.009$) [Figure 2B] and had a higher magnitude of SARS-CoV-2 antibodies (median COI 7.72 [3.05–41.6] vs 3.54 [1.77–8.70], $p = 0.002$). Sensitivity analyses showed that the effect size was greater when only patients with undetectable drug and an anti-infliximab antibody were included (OR 2.02, 95% CI 1.20–3.26, $p = 0.007$; median COI 9.26 [5.47–44.80], $p = 0.001$).

Similarly, compared to patients with detectable adalimumab drug levels [≥ 0.8 mg/L], patients with undetectable drug levels were more likely to be seropositive for SARS-CoV-2 antibodies [OR 1.72, 95% CI 0.96–2.90, $p = 0.04$; Figure 2B], but there was no difference in the magnitude of SARS-CoV-2 antibodies (median COI 8.49 [3.21–25.5] vs 4.80 [1.93–18.5], $p = 0.11$).

There was no association between vedolizumab drug levels and seropositivity or the magnitude of SARS-CoV-2 antibodies [Figure 2B]. Compared to vedolizumab-treated patients, infliximab- and adalimumab-treated patients with undetectable drug levels had similar seropositivity rates (infliximab: 4.8% [33/681], adalimumab: 4.9% [17/347], vedolizumab: 6.7% [25/375], $p = 0.43$) and the magnitude of SARS-CoV-2 titres (infliximab: median COI 7.72 [3.05–41.60], adalimumab: median COI 8.49 [3.21–25.50], vedolizumab: median COI 21.6 [4.39–68.10], $p = 0.38$).

4.3. Seroconversion and subsequent positive PCR-confirmed SARS-CoV-2

Overall, 1.6% [23/1428] of patients had a positive PCR test prior to collection of the sample used for SARS-CoV-2 antibody testing. Of those with a positive PCR test, all of whom were treated with an anti-TNF drug, 65.2% [15/23] patients seroconverted. There was no difference in seroconversion, stratified by time from PCR testing to SARS-CoV-2 [N] antibody testing (positive antibody: 100 days [75.7–145.5] vs negative antibody: median 64 days [27.5–129.0], $p = 0.42$). Moreover, there was no correlation between time to

Table 1. Baseline characteristics of patients, stratified by biologic therapy

Variable		Adalimumab	Infliximab	Ustekinumab	Vedolizumab	Total
Sex	Female	49.66% [2520/5074]	44.24% [2607/5893]	48.75% [39/80]	42.13% [158/375]	46.61% [5324/11 422]
	Male	50.24% [2549/5074]	55.69% [3282/5893]	51.25% [41/80]	56.53% [212/375]	53.27% [6084/11 422]
	Unknown	0.10% [5/5074]	0.07% [4/5893]	0.00% [0/80]	1.33% [5/375]	0.12% [14/11 422]
Diagnosis	IBD	18.19% [923/5074]	19.57% [1153/5893]	56.25% [45/80]	29.33% [110/375]	19.53% [2231/11 422]
	Non-IBD	1.99% [101/5074]	0.49% [29/5893]	0.00% [0/80]	0.00% [0/375]	1.14% [130/11 422]
	Unknown	79.82% [4050/5073]	79.94% [4711/5893]	43.75% [35/80]	70.67% [265/375]	79.33% [9061/11 422]
Age [years]		38.80 [27.44–52.42]	34.73 [23.07–50.14]	36.82 [24.65–57.68]	40.33 [29.66–57.78]	36.77 [25.53–51.48]
Income deprivation score		0.10 [0.06–0.17]	0.10 [0.06–0.17]	0.09 [0.06–0.14]	0.11 [0.07–0.19]	0.10 [0.06–0.17]
UK region	East Midlands	8.58% [422/4921]	6.51% [373/5732]	0.00% [0/79]	0.82% [3/366]	7.19% [798/11 098]
	East of England	12.17% [599/4921]	10.62% [609/5732]	10.13% [8/79]	6.83% [25/366]	11.18% [1241/11 098]
	London	12.48% [614/4921]	18.55% [1063/5732]	5.06% [4/79]	36.89% [135/366]	16.36% [1816/11 098]
	North East	2.89% [142/4921]	1.90% [109/5732]	1.27% [1/79]	0.00% [0/366]	2.27% [252/11 098]
	North West	9.33% [459/4921]	8.30% [476/5732]	0.00% [0/79]	2.73% [10/366]	8.52% [945/11 098]
	Scotland	26.19% [1289/4921]	19.23% [1102/5732]	0.00% [0/79]	0.00% [0/366]	21.54% [2391/11 098]
	South East	9.29% [457/4921]	12.07% [692/5732]	10.13% [8/79]	6.83% [25/366]	10.65% [1182/11 098]
	South West	7.82% [385/4921]	9.19% [527/5732]	53.16% [42/79]	16.94% [62/366]	9.15% [1016/11 098]
	Wales	2.01% [99/4921]	2.16% [124/5732]	0.00% [0/79]	0.82% [3/366]	2.04% [226/11 098]
	West Midlands	6.34% [312/4921]	6.07% [348/5732]	0.00% [0/79]	16.67% [61/366]	6.50% [721/11 098]
	Yorkshire and the Humber	2.91% [143/4921]	5.39% [309/5732]	20.25% [16/79]	11.48% [42/366]	4.60% [510/11 098]

IBD = inflammatory bowel disease.

SARS-CoV-2 [N] antibody test and the magnitude of SARS-CoV-2 antibodies [Spearman's rho $R = 0.034$, $p = 0.88$].

Subsequent positive PCR-confirmed that SARS-CoV-2 was seen in 7.9% [12/152] of patients. The median magnitude of SARS-CoV-2 antibody reactivity prior to a positive PCR test was 1.74 COI [1.14–15.48], with a median time from positive SARS-CoV-2 antibody to positive PCR test of 183.5 days [129.8–235.3]. There was no association between biologic class (anti-TNF 7.4% [10/135] vs vedolizumab 11.8% [2/17], $p = 0.35$), or the magnitude of SARS-CoV-2 antibody reactivity [$p = 0.13$], and a subsequent positive PCR test.

5. Discussion

We have shown that patients with IMIDs treated with infliximab and adalimumab have attenuated serological responses to SARS-CoV-2 infection with lower seroprevalence and antibody reactivity when compared to vedolizumab-treated patients. Amongst patients

Table 2. Seroprevalence of SARS-CoV-2 antibodies, stratified by baseline characteristics

Variable	Seroprevalence	*p* value
Biologic therapy		
Adalimumab	3.00% [152/5074]	0.003
Infliximab	3.02% [178/5893]	
Ustekinumab	1.25% [1/80]	
Vedolizumab	6.67% [25/375]	
Sex		
Female	2.97% [158/5324]	0.27
Male	3.24% [197/6084]	
Unknown	7.14% [1/14]	
Diagnosis		
IBD	2.38% [53/2231]	0.07
Non-IBD	3.08% [4/130]	
Unknown	3.30% [299/9061]	
Region		
East Midlands	1.88% [15/798]	<0.001
East of England	2.74% [34/1241]	
London	7.93% [144/1816]	
North East	2.38% [6/252]	
North West	3.28% [31/945]	
Scotland	1.42% [34/2391]	
South East	1.86% [22/1182]	
South West	1.48% [15/1016]	
Wales	2.21% [5/226]	
West Midlands	4.72% [34/721]	
Yorkshire and the Humber	2.16% [11/510]	
Income score	0.11 [0.06–0.19]	0.05
Age > 70 years	1.8% [10/555]	0.08
Calendar month sample tested		
January	0.00% [0/51]	<0.001
February	0.61% [2/330]	
March	0.20% [1/491]	
April	3.53% [20/566]	
May	3.74% [38/1015]	
June	4.60% [87/1893]	
July	3.19% [82/2570]	
August	2.20% [49/2225]	
September	3.37% [77/2282]	

IBD = inflammatory bowel disease.

treated with adalimumab and infliximab, seropositivity rates were highest in patients with undetectable drug levels and were similar to those observed in patients treated with vedolizumab. One-third of our cohort who had PCR-confirmed SARS-CoV-2 infection, all of whom were treated with anti-TNF therapy, subsequently did not develop SARS-CoV-2 antibodies. Subsequent positive PCR-confirmed SARS-CoV-2 was observed in 8% patients.

Like infliximab,[8] adalimumab impairs antibody responses following SARS-CoV-2 infection, and we observed that higher SARS-CoV-2 antibody levels were associated with undetectable infliximab and adalimumab drug levels. This is biologically plausible since anti-TNF drugs directly impede the immune mechanisms responsible for generating antibody responses, including maturation of antigen presenting cells and co-stimulation of antigen-specific T-cells.[16–18] TNF neutralization, or genetic ablation, results in reduced B-cells in primary follicles in germinal centres and the periphery, and B-cell immunoglobulin synthesis.[16] In keeping with this hypothesis, in infliximab-treated patients, the highest SARS-CoV-2 antibody concentrations were seen in patients with undetectable drug levels in the presence of anti-infliximab antibodies where drug is absent.[19,20] It is possible that this cohort of patients were less likely to be treated with an immunomodulator, which we have previously shown is independently associated with SARS-CoV-2 seroconversion in infliximab-treated patients with IBD.[8] An alternative explanation for our results is that anti-TNF agents in IMIDs prevent severe COVID-19 infection and consequently immune responses.[21] Against this postulate, we previously observed no difference in rates of hospitalization for confirmed COVID-19 amongst infliximab- compared to vedolizumab-treated patients with IBD, and that vaccine responses were similarly impaired in anti-TNF-treated patients.[8,10]

Even after PCR-confirmed infection, one-third of patients who were subsequently tested for SARS-CoV-2 antibodies, and all of whom were treated with either adalimumab or infliximab, failed to mount an antibody response. Whilst this might be explained by antibody decay in the period between the positive PCR test and SARS-CoV-2 antibody test, we reported similar findings in our prospective cohort of patients with IBD, where 52% [42/81] of infliximab-treated patients did not mount an antibody response following PCR-confirmed infection.[8] Whether a failure to seroconvert after infection predisposes people to recurrent SARS-CoV-2 infection cannot be determined in this cohort because of a paucity of PCR testing in the early phase of the pandemic. However, following a positive SARS-CoV-2 antibody test, over 7% of patients subsequently had PCR-confirmed SARS-CoV-2. We acknowledge that none of these 12 patients had a positive PCR test prior to their initial SARS-CoV-2 antibody test and it is therefore possible that these patients may have had false positive antibody tests. An alternative explanation is that these patients may have failed to clear a primary SARS-CoV-2 infection or had a second infection.

The main strength of this study was analysis of SARS-CoV-2 antibodies on more than 13 000 samples from 11 422 unique patients with IMIDs treated with biologic therapy during the early phase of the pandemic. Other strengths include correlation with comprehensive biologic drug-level data, and linkage with SARS-CoV-2 public health testing data. We acknowledge, however, the following limitations. First, because this was an analysis of surplus serum, clinical details were infrequently entered on requisition forms. We therefore did not have access to comprehensive clinical data for study subjects including comorbidities, ethnicity, diagnosis, symptoms of suspected COVID-19, and indications for, and duration of, biologic and concomitant therapies. Second, as serum samples for this study were collected early in the pandemic, a limited number of subjects had PCR-confirmed infection. Third, our ability to interpret SARS-CoV-2 antibody durability and risk of re-infection was limited by the duration of follow-up, frequency of sampling and the availability of the first positive PCR test results conducted in England. Finally, as this study involved surplus serum samples used for TDM, limited data were available for patients treated with therapies for which TDM is not widely used, including ustekinumab.

From a public health perspective, attention has turned from natural infection to vaccine effectiveness in the face of novel SARS-CoV-2 variants. Several groups have shown that most patients with IBD can mount an effective immune response in the short term following both licensed doses of SARS-CoV-2 vaccine.[22–27] Urgent research is needed to understand the factors linked to vaccine non-response. For patients who need to start anti-TNF therapy, they and their families should receive SARS-CoV-2 vaccines without a delay between vaccine doses, wherever possible before anti-TNF therapies are started. Whether timing booster doses towards the end of an anti-TNF treatment cycle when drug levels are lowest,[28] and/or the temporary discontinuation of immunomodulators,[29] potentiate

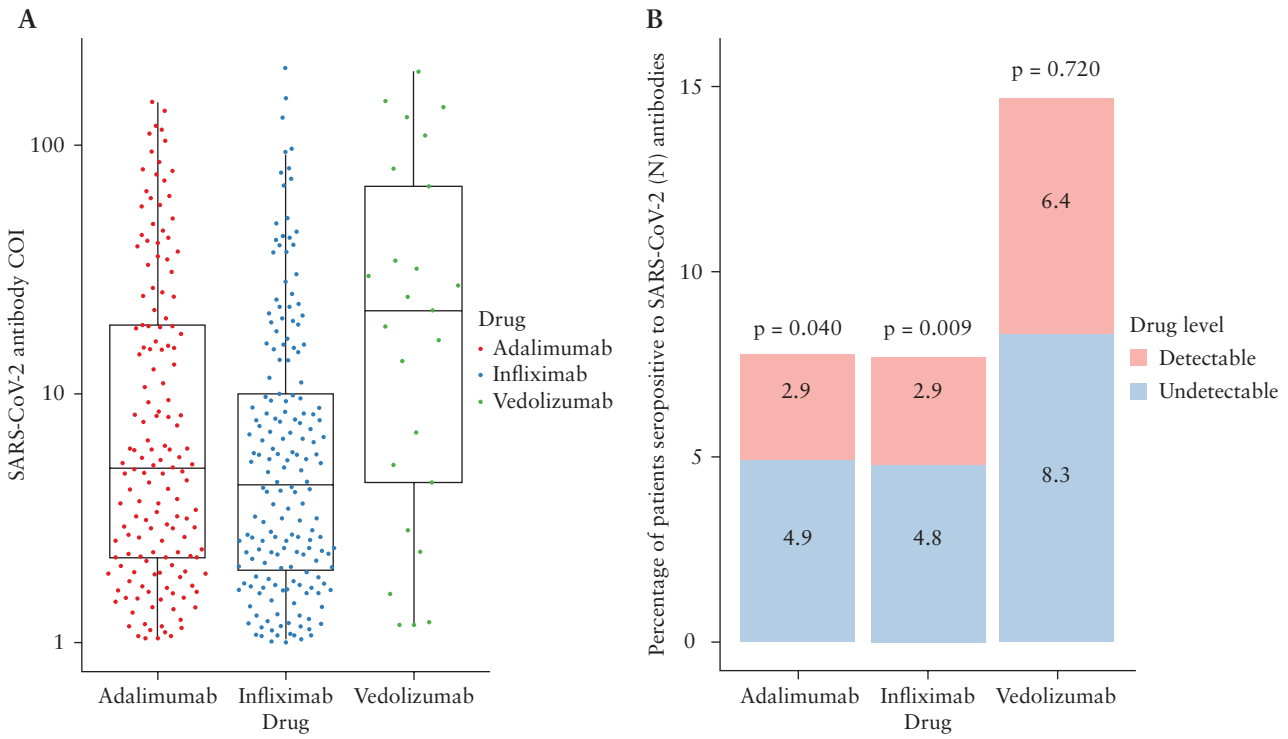

Figure 2. [A] Boxplot of the magnitude of SARS-CoV-2 antibody reactivity, stratified by biologic therapy. [B] Percentage of patients with seropositivity, defined by a SARS-CoV-2 nucleocapsid antibody concentration ≥ 10 U/mL, stratified by biologic drug and drug level. The *P* value above each bar represents a within-drug comparison between patients with detectable or undetectable drug levels and SARS-CoV-2 antibodies.

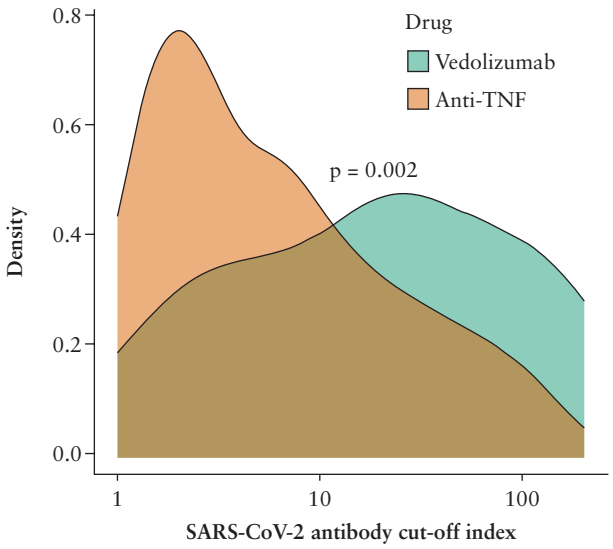

Figure 3. Density plot of the magnitude of SARS-CoV-2 antibody reactivity, stratified by biologic therapy.

long-term immunogenicity warrants further study. So too does the use of higher-dose vaccines,[3] adjuvants including the influenza vaccines [ComFluCOV][30] and/or switching between vaccines with different mechanisms of action.[31]

6. Conclusion

Patients with IMIDs treated with infliximab and adalimumab have attenuated serological responses to SARS-CoV-2 when compared to vedolizumab-treated patients. Seropositivity rates were highest in patients with undetectable drug levels and were similar to those observed in patients treated with vedolizumab, supporting a causal relationship between anti-TNF use and attenuated antibody responses to infection, although confounding factors, such as combination therapy with an immunomodulator, may have influenced the results.

Conflict of Interest

Dr Lin reports non-financial support from Pfizer, non-financial support from Ferring, outside the submitted work. Dr Chee reports non-financial support from Ferring, personal fees and non-financial support from Pfizer, outside the submitted work. Dr Derikx has served on the advisory board for Sandoz, outside the submitted work. Dr Kelleher reports financial support from Pfizer, Wellcome Trust, UKRI and non-financial support from Oxford Immunotech outside the submitted work. Dr Kok reports personal fees from Janssen, personal fees from Takeda, personal fees from PredictImmune, and personal fees from Amgen, outside the submitted work. Dr Lees reports personal fees from Abbvie, personal fees from Janssen, personal fees from Pfizer, personal fees from Takeda, grants from Gilead, personal fees from Gilead, personal fees from Galapagos, personal fees from Iterative Scopes, personal fees from Trellus Health, personal fees from Celltion, personal fees from Ferring, and personal fees from BMS, during the conduct of the study. Dr Macdonald reports grants and personal fees from Takeda Pharmaceuticals, grants and personal fees from Biogen, personal fees and non-financial support from AbbVie, personal fees from Grifols, personal fees from Sandoz, personal fees from Celltrion, personal fees and non-financial support from Janssen, personal fees from Vifor Pharmaceuticals, personal fees from Predictimmune, personal fees from Bristol Myers Squibb, and non-financial support from Ferring Pharmaceuticals, outside the submitted work. Dr Shaji reports grants from Takeda, Abbvie, AMGEN, Tillots Pharma, and personal fees from Jaansen, Takeda, Galapagos, Celltrion, Falk Pharma, Tillots pharma, Cellgene, Pfizer and Phamrmacocosmos, outside the submitted work. Dr Philip J. Smith reports speaker fees and advisory board sponsorship from Janssen, Celltrion and

Takeda outside the submitted work. Dr Irving reports grants and personal fees from Takeda, grants from MSD, grants and personal fees from Pfizer, personal fees from Galapagos, personal fees from Gilead, personal fees from Abbvie, personal fees from Janssen, personal fees from Boehringer Ingelheim, personal fees from Topivert, personal fees from VH2, personal fees from Celgene, personal fees from Arena, personal fees from Samsung Bioepis, personal fees from Sandoz, personal fees from Procise and personal fees from Prometheus, outside the submitted work. Dr Powell reports personal fees from Takeda, personal fees from Janssen, personal fees from Pfizer, personal fees from Bristol-Myers Squibb, personal fees from Abbvie, personal fees from Roche, personal fees from Lilly, personal fees from Allergan and personal fees from Celgene, outside the submitted work; and Dr Powell has served as a speaker/advisory board member for Abbvie, Allergan, Bristol Myers Squibb, Celgene, Falk, Ferring, Janssen, Pfizer, Tillotts, Takeda and Vifor Pharma. Dr Kennedy reports grants from F. Hoffmann-La Roche AG, grants from Biogen Inc, grants from Celltrion Healthcare, grants from Galapagos NV and non-financial support from Immundiagnostik, during the conduct of the study; grants and non-financial support from AbbVie, grants and personal fees from Celltrion, personal fees and non-financial support from Janssen, personal fees from Takeda, and personal fees and non-financial support from Dr Falk, outside the submitted work. Dr Goodhand reports grants from F. Hoffmann-La Roche AG, grants from Biogen Inc, grants from Celltrion Healthcare, grants from Galapagos NV and non-financial support from Immundiagnostik, during the conduct of the study. Dr Ahmad reports grants and non-financial support from F. Hoffmann-La Roche AG, grants from Biogen Inc, grants from Celltrion Healthcare, grants from Galapagos NV and non-financial support from Immundiagnostik, during the conduct of the study; personal fees from Biogen inc, grants and personal fees from Celltrion Healthcare, personal fees and non-financial support from Immundiagnostik, personal fees from Takeda, personal fees from ARENA, personal fees from Gilead, personal fees from Adcock Ingram Healthcare, personal fees from Pfizer, personal fees from Genentech and non-financial support from Tillotts, outside the submitted work. Neil Chanchlani, Ben Hamilton, Rachel Nice, Zehra Arkir, Claire Bewshea, Bessie Cipriano, Allan Dunlop, Louise Greathead, Rachel Griffiths, Hajir Ibraheim and Timothy J McDonald have no conflicts of interest.

Author Contributions

N.C., S.L., C.B., S.S., N.P., N.A.K., J.R.G. and T.A. participated in the conception and design of this study. C.B. was the project manager. R.N. and T.J.M. coordinated all biochemical analyses and central laboratory aspects of the project. Sample collection and coordination was done by R.N., Z.A., C.B., B.C., A.D., L.G., R.G., P.K., K.B.K., J.M., T.J.M. and P.M.I. N.C., S.L., D.C., R.N., H.I., P.J.S., P.M.I., N.P., N.A.K., J.R.G. and T.A. were involved in the acquisition, analysis or interpretation of data. Data analysis was done by N.C., S.L. and N.A.K. Drafting of the manuscript was done by N.C., S.L., L.A.A.P.D., N.A.K., J.R.G. and T.A. T.A. obtained the funding for the study. All the authors contributed to the critical review and final approval of the manuscript. N.C., S.L., N.A.K. and T.A. have verified the underlying data.

Patient Involvement

The Exeter IBD Patient Panel reviewed the study protocol. A member of the Exeter IBD Patient Panel sits on the study management committee, ensuring patient involvement in all aspects of study delivery, data analysis and dissemination of findings.

Data Sharing

The study protocol including the statistical analysis plan is available at www.clarityibd.org. Individual participant de-identified data that underlie the results reported in this article will be available immediately after publication for a period of 5 years. The data will be made available to investigators whose proposed use of the data has been approved by an independent review committee. Analyses will be restricted to the aims in the approved proposal. Proposals should be directed to tariq.ahmad1@nhs.net; to gain access data requestors will need to sign a data access agreement.

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Supplementary Data

Supplementary data are available at *ECCO-JCC* online.

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