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Running title: COVID-19 vaccination in IMIDs

Immunogenicity and safety of mixed COVID-19 vaccine regimens in immune mediated inflammatory diseases: an observational cohort

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CA Hitchon [1], C Mesa [2], CN Bernstein [1], RA Marrie [1,3], C Card [2,4], S O'Brien [5], J Kim [2]

1 Department of Internal Medicine, Max Rady College of Medicine, Rady Faculty of Health Sciences, University of Manitoba, Winnipeg, CAN

2 National Microbiology Laboratory Public Health Agency of Canada Winnipeg, CAN

3 Department of Community Health Sciences, Max Rady College of Medicine, Rady Faculty of Health Sciences, University of Manitoba, Winnipeg, CAN

4 Department of Medical Microbiology and Infectious Diseases, Rady Faculty of Health Sciences, University of Manitoba, Winnipeg, CAN

5 Canadian Blood Services, Ottawa, CAN

Authors:

Carol A Hitchon MD MSc Department of Internal Medicine, Max Rady College of Medicine, Rady Faculty of Health Sciences, University of Manitoba, Winnipeg, CAN

Christine Mesa, MSc National Microbiology Laboratory Public Health Agency of Canada Winnipeg, CAN

Charles N Bernstein, MD Department of Internal Medicine, Max Rady College of Medicine, Rady Faculty of Health Sciences, University of Manitoba, Winnipeg, CAN

Ruth Ann Marrie, MD PhD Department of Internal Medicine, and Department of Community Health Sciences, Max Rady College of Medicine, Rady Faculty of Health Sciences, University of Manitoba, Winnipeg, CAN

Sheila O'Brien, PhD Canadian Blood Services, Ottawa, CAN

Catherine Card, PhD, National Microbiology Laboratory, Public Health Agency of Canada Winnipeg, CAN and Department of Medical Microbiology and Infectious Diseases, Rady Faculty of Health Sciences, University of Manitoba, Winnipeg, CAN

John Kim PhD National Microbiology Laboratory Public Health Agency of Canada Winnipeg, CAN

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Corresponding author

CA Hitchon

RR149 800 Sherbrook Street

Winnipeg, Manitoba

email: carol.hitchon@umanitoba.ca

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Abstract

Objective: Among persons with immune-mediated inflammatory diseases (IMIDs) who received homologous or heterologous SARS-CoV-2 vaccines, we compared post-vaccine vaccine antibody responses and IMID disease activity/states.

Methods: In this single centre observational cohort study, we followed persons with diagnosed inflammatory arthritis (n= 78; 77% rheumatoid arthritis), systemic autoimmune rheumatic diseases (n=84; 63% lupus), inflammatory bowel disease (n= 93; 43% Crohn's), and multiple sclerosis (n= 72; 77% relapsing remitting) who received COVID-19 vaccinations between 03/2021-09/2022. Participants self-reported COVID-19 illness and exposure risks, IMID disease activity/state using disease-specific measures, and had anti-spike, -receptor binding domain (RBD) and -nucleocapsid (NC) IgG antibodies tested by multiplex immunoassays following each vaccination (V1, V2, V3, V4). Anti-SARS-CoV-2 responses were compared across vaccine regimens and to responses in 370 age-sex matched vaccinated blood donor controls.

Results: IMID participants were predominantly female (79.4%), with a mean (standard deviation) age of 56.0 (14.3) years. Most participants (66.1%) received homologous mRNA (BNT162b2 or mRNA1273) vaccines, 2.4% received homologous ChAdOx1, and 30.6% received heterologous vaccines (23.9% ChAdOx1/mRNA, 6.4% heterologous mRNA) for their first two vaccines. Seroconversion rates for 238 IMIDs increased 1 month post V2 (post-V1 anti-spike 52.0%, anti-RBD 58.9%; post-V2 anti-spike 91.5%, anti-RBD 90.22%, but remained lower than controls (post-V2 anti-Spike 98.1% $p<0.0001$). Antibody titers waned by 3 months post V2 but increased 1 month post V3 and 1 month post V4. If primed with a vector vaccine, a mRNA vaccine increased antibody titers to those comparable to homologous mRNA vaccines. Anti-RBD and Spike titers were higher in anti-NC seropositive (N=31; 25 participants) versus seronegative samples ($p<0.001$). IMID disease activity/state and rates of self- reported moderate or severe IMID flare were similar across vaccinations.

Interpretation: Heterologous COVID-19 vaccination improves seroconversion rates following a viral vector vaccine and does not lead to IMID disease flare. IMIDs benefit from at least three vaccines.

Funding: Research Manitoba, Public Health Agency of Canada

Key Words: COVID-19, vaccination, immune systems diseases

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Strengths and limitations

- Longitudinal cohort study with systematic collection of data on COVID-19 infection, IMID disease activity and with paired biosamples for anti-SARSCoV2 IgG assays following each vaccine for up to four vaccines.
- Cross disease comparisons of four IMIDs from different medical specialties which are treated with immunocompromising medications.
- Validated measures of self-report IMID disease activity/state used to assess vaccine safety and risk of post-vaccine IMID disease flare.
- Relatively small sample size for each IMID and predominantly female population (as expected for these IMIDs) limits analysis of sex and gender effects.

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Key Messages

What is already known about this subject?

Some treatments used for people with immune-mediated inflammatory diseases (IMIDs) such as autoimmune inflammatory arthritis, systemic autoimmune rheumatic diseases, inflammatory bowel disease, and multiple sclerosis may reduce COVID-19 vaccine-mediated immunogenicity.

What does this study add?

COVID-19 vaccines do not increase risk of disease flare for most IMIDs.

Across IMIDs, anti-SARS-CoV-2 antibody levels following a single vaccine were low but improved after receiving a second mRNA vaccine. Anti-SARS-CoV-2 antibody levels waned following the second vaccine and recovered after a third vaccine.

Vaccine induced anti-SARSCoV2 antibody titers are reduced in IMID participants taking immunosuppressants or biologics.

IMIDs who received an initial vector vaccine followed by a mRNA vaccine have vaccine induced anti-SARSCoV2 antibody titers comparable to those following a homologous mRNA vaccine course.

How might this impact on clinical practice?

These observations support the use of heterologous vaccine courses and third vaccines for people with IMIDs particularly in the setting of immunosuppression and biologic treatment.

These data can inform public health recommendations for COVID-19 vaccination that balance the need for optimal vaccine-induced protection with the need for feasible local vaccination programs and global vaccine equity.

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Introduction

COVID-19 vaccines have reduced rates of serious severe acute respiratory syndrome coronavirus (SARS-CoV-2) infection and mortality in the general population.[1-4] However, information on optimal vaccine strategies for immunocompromised individuals who are at increased risk of serious COVID-19 infection is limited.

Immune-mediated inflammatory diseases (IMIDs) such as autoimmune inflammatory arthritis (IA), systemic autoimmune rheumatic disease (SARD), inflammatory bowel disease (IBD), and multiple sclerosis (MS) affect 5% of the general population and share an autoimmune phenotype that affects multiple organ systems and treatment with immune therapies[5]. Due to immune-mediated disease and treatment, some people with IMIDs are at increased risk of vaccine preventable disease including serious COVID-19 infection[6-10]. While COVID-19 vaccines are effective in the general population, immune dysregulation from disease or treatment may impair vaccine responses in people with IMIDs. Due to evolving regional vaccination strategies in our region ([11] and Supplemental Figure 1), a high proportion of individuals with immunocompromised conditions received heterologous vaccine courses. Although available data on immunogenicity of these mixed vaccine courses in the general population are reassuring,[12-14] data are limited regarding the safety and immunogenicity of this strategy for immunocompromised patients as are data comparing these outcomes across diseases.

We established a cohort of patients diagnosed with any of IA, SARDs, IBD or MS to determine the safety (IMID flare) and humoral immunogenicity following COVID-19 vaccination and to assess the impact of mixing COVID-19 vaccine types. Herein, we report the clinical safety results and seroconversion results obtained after four vaccinations and compare seroconversion rates across initial vaccine combinations.

Methods:

Study Design: This single site observational cohort from Manitoba, Canada, was established in March 2021 and enrolled people diagnosed with any of IA, SARDs, IBD, or MS. Data was

collected until October 30th 2022. Vaccines were administered in accordance with provincial public health recommendations. Study participants were followed 1 and 3 months after each vaccine for up to four vaccines.

Clinical data collected: Demographic data, including birth date, sex, self-reported ethnic group according to Canadian Institute for Health Information guidelines[15], highest education level achieved, self-reported comorbidity and health behaviors such as smoking history were collected at baseline. Clinical data including IMID treatment, IMID-specific disease activity/state measures, participant reported interval COVID-19 infections, and biosamples were collected at each visit. IMID treatment was subcategorized as immunosuppressants, biologic or advanced therapies, other immunomodulators, and corticosteroids (Supplementary Table 1). Self-reported IMID disease activity/status was assessed with disease relevant validated measures (IA: Routine Assessment of Inflammatory Disease version 3 RAPID-3, Rheumatoid Arthritis Flare Core domain indices; SARDs: Systemic Lupus Activity Questionnaire SLAQ; IBD: Inflammatory Bowel Symptoms Severity Index-Short Form (IBDSI-SF), Manitoba IBD flare question; MS: self report disease activity, Expanded Disability Status Scale [EDSS])[16-20]. These disease-related questionnaires were fully completed for IBD (248/255 97% visits), IA (208/215; 97% visits), SARDs (239/260; 92% visits), and MS (291/327; 89% visits). Data were not imputed. Individuals could attend a study visit in person or submit their data by mailed paper forms or direct entry into a REDCap electronic database hosted at the University of Manitoba[21]. Participants had the option to participate in the safety study component only.

Biosamples: Participants not attending the clinic were provided kits to collect blood by finger poke on dried blood spot (DBS) cards for postal submission[22]. Participants attending the clinic had blood collected by venipuncture and DBS were prepared before processing blood for serum. Aliquoted serum and DBS cards were stored at -20C for batch testing.

Regional population control biosample data were obtained from 370 Canadian Blood Services blood donors who were age and sex-matched to the clinical cohort.

Serology: A bead-based multiplexed immunoassay was used to detect IgG anti-Spike (S1), Receptor Binding Domain (RBD) (reflecting response to vaccine), and Nucleocapsid (NC)

antibodies (reflecting response to infection) (Bio-Rad Bioplex 2200 SARS-CoV-2 IgG assay). This assay was chosen based on an evaluation of several multiplex platforms using a large panel of well-pedigreed plasma-DBS specimens[22]. The quantitative measurement of the antibody response used the WHO International standard for anti-SARS-CoV-2 antibody detection. Cut-offs for seropositivity for DBS samples were established using ROC curve analysis. Cut-offs for plasma from the company were used. Sera from IA (n=19) and lupus (n=31) patients collected before the pandemic were tested to evaluate the potential for false positive SARS-CoV-2 antibody tests in IMIDs with known autoantibodies. Concordance of antibody assays in paired DBS and serum samples was assessed using the Kendalls tau b test. The controls were tested using the Roche Elecysys® anti-SARS-CoV-2 spike protein semiquantitative immunoassay (Roche Diagnostics International Ltd, Rotkreuz, Switzerland) which measures total antibodies (including IgA, IgM and IgG) to the SARS-CoV2- spike protein (anti-S) and the Roche Elecsys® anti-SARS-CoV-2 qualitative immunoassay (Roche Diagnostics LTD Rotkreuz, Switzerland) which measures total antibodies (including IgA, IgM and IgG) to SARS-CoV-2 recombinant protein, nucleocapsid antigen (anti-NC).

Analysis: Demographic information of participants is reported using descriptive statistics including mean (standard deviation-sd), median (range or interquartile range-IQR), and counts (%). Non-parametric tests (Mann-Whitney U or Kruskal Wallis tests with Bonferroni adjustment [Badj] for multiple comparisons) were used to compare antibody levels across groups and visits. Wilcoxon signed rank tests were used to compare antibody levels across visits within individuals. Binary logistic regression was used to evaluate predictors of seroconversion one month following vaccine 2 (V2). Variables included sex, age greater than 65 years, IMID diagnosis, and treatment category (none versus biologics and advanced therapies versus immunosuppressants versus other agents), and vaccine mix (vector-mRNA versus mRNA-mRNA). Statistical analysis was conducted using IBM SPSS Statistics version 27 for Windows (IBM Corporation, Armonk, N.Y., USA).

All subjects provided informed consent. The study was approved by the University of Manitoba Health Research Ethics Board (HS24647-H2021-005), Manitoba Shared Health (SH2021:009),

and the Health Canada and Public Health Agency of Canada Research Ethics Board (REB 2021-018P).

Patient/public involvement: Patients and public informed the study questions but were not involved in study design, conduct or reporting.

Results:

Between March 12th 2021 and July 30 2022, we recruited 339 participants (Supplementary Figure 2). Vaccination and disease activity data for the time points reported herein were available for 327 participants (78 IA: 77% RA; 84 SARDS: 63 % lupus; 93 IBD: 43 % Crohn's and 72 MS: 77% Relapsing remitting) (Table 1). Most were female (79.4%) and white (84.7%) with a mean (sd) age of 56.0 (14.3) years. Nearly one-third (30.6%) of IMID participants were taking biologics or advanced therapies.

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Table 1 Demographics, vaccines administered, self reported IMID flare, and COVID-19 history of participants

	IA N=78	SARDs N=84	IBD N=93	MS N=72	All IMIDs N=327
Age, years (mean SD)	61.9(11.8)	55.7(13.4)	53,7(16.4)	53.1(13.5)	56.0 (14.3)
Female n (%)	66 (84.6)	75 (89.3)	59(63.4)	59 (81.9)	259 (79.4)
White n (%)	66(84.6)	64 (76.2)	79(84.9)	68 (94.4)	277 (84.7)
Education Years of school (median/IQR)	15.0(4.5)	16.0(4.3)	16.0(4.0)	16.0(5.0)	16.0(4.0)
Comorbidity (n %)[2]					
Cardiovascular	35 (44.9)	31 (36.9)	26 (28.0)	22 (30.5)	115(35.1)
Pulmonary	8 (10.2)	24 (28.6)	15 (16.1)	8 (11.1)	55(16.8)
Diabetes	13 (19.7)	3 (3.7)	3 (3.4)	5 (6.9)	24(7.3)
Other endocrine	25 (32.1)	24 (28.6)	20 (21.5)	20 (27.8)	90(27.4)
Renal disease	2 (3.0)	9 (11.0)	4 (4.3)	0 (0.0)	15(4.6)
Cancer	15 (19.2)	10 (11.9)	12 (12.9)	56.9)	43 (13.10
Mental Health	17 (21.8)	27 (32.9)	27 (29.0)	22 (30.6)	94(28.7)
Total (median range)	2.5(0-10)	3(0-10)	2(0-10)	2(0-6)	2(0-10)
BMI (mean SD)	27 (6.6)	28.0 (7.8)	27.2 (6.1)	28.0 (6.2)	27.7 (6.7)
IMID Treatment level [1] n (%)					
Immunomodulators	4(5.1)	25 (30.5)	20 (21.5)	28(38.9)	76 (23.5)
Immunosuppressants	27 (34.6)	42 (51.2)	10 (10.8)	0 (0.0)	79 (31.1)
Biologics/JAKi	37(47.4)	7 (8.5)	41 (44.1)	14 (19.4)	99 (30.6)
- anti-TNF (n)	21	0	28	0	49
- anti-B cell (n)	5	7	0	10	22
- other or JAKi (n)	11	0	13	4	28
None	10 (12.8)	8 (9.8)	22(23.7)	30 (41.7)	70 (21.6)
Vaccine type V1 n (%)[3]					
CHAdOx1	15 (19.5)	28 (33.3)	17 (18.3)	26 (36.1)	86 (26.4)
BNT	60 (77.9)	56 (66.7)	66 (71.0)	39 (54.2)	221 (67.8)
mRNA1273	2 (2.6)	6 (7.1)	10 (10.8)	7 (9.7)	25 (7.7)
Vaccine type V2 n (%)[3]					
CHAdOx1	2 (2.6)	3 (3.6)	3 (3.2)	1 (1.4)	9 (2.7)
BNT	65 (84.4)	58 (68.7)	60 (65.2)	54 (75.0)	236 (72.8)
mRNA1273	16 (13.0)	23 (27.7)	29 (31.5)	17 (23.6)	79(24.4)
Vaccine type V3 n/N (%)[3]					
CHAdOx1	0	0	1 (1.3)	1 (1.6)	2 (0.7)
BNT	54 (71)	58 (78.4)	49 (66,2)	39 (61.9)	200 (71.2)
mRNA1273	16 (21.1)	16 (21.6)	24 (32.4)	23 (36.5)	79 (28.1)
Other	0	0 (0)	1 (1.4)	1 (1.6)	2 (0.7)
Vaccine type V4 n/N (%)[3]					
CHAdOx1	0 / (0)	1 (2.7)	0 (0)	0 (0)	1 (0.8)
BNT	25 (69.4)	28 (75.7)	25 (64.1)	12 (70.6)	90 (69.8)

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mRNA1273	10 (27.8)	8 (21.6)	14 (35.9)	2 (11.8))	34 (26.4)
Other	-	-	-	3 (17.6)	3 (2.3)
Vaccines for V1 and V2 n (%)					
CHAdOx1-- BNT	9 (11.5)	10 (11.9)	4 (4.3)	17 (23.6)	40(12.2)
CHAdOx1 - mRNA1287	4 (5.1)	15(17.9)	11 (11.8)	8 (11.1)	38 (11.6)
BNT – mRNA1273 or mRNA1273-BNT	4 (5.1)	5 (6.0)	10 (10.8)	2 (2.8)	21 (6.4)
BNT-CHAdOx1	0(0)	0(0)	1 (1.1)	0(0)	1 (0.3)
CHAdOx1-CHAdOx1	2 (2.6)	3 (3.6)	2 (2.2)	1 (1.4)	8 (2.4)
BNT- BNT	56(71.8)	46 (54.8)	55 (59.1)	37 (51.4)	194 (59.3)
mRNA1273 -mRNA1273	2 (2.6)	4 (4.8)	9 (9.7)	7 (9.7)	22 (6.7)
Vaccine interval Days between V1 and V2 median (range)	66 (21-97)	62 (20-188)	57 (20-98)	59 (26-97)	60 (20-188)
Self-report IMID flare [4,5]					
1 month post V1 n/N (%)	12/40 (30.0)	10/39 (25.6)	8/41 (19.5)	1/17 (5.9)	31/137(22.6)
1 month post V2 n/N (%)	8/53 (15.5)	9/53 (17.0)	17/69(24.6)	2/65 (3.1)	36/240(15.0)
1 month post V3 n/N (%)	11/55 (20.0)	19/59 (32.2)	14/63 (22.2)	1/57 (1.8)	45/234 (19.2)
1 month post V4 n/N (%)	6/37 (16.2)	12/34 (35.3)	8/40 (20.0)	1/23 (4.3)	27/134 (20.1)
1 month post any V n/N(%)	37/185 (20.0)	50/185 (27.0)	47/213 (22.1)	5/162 (3.1)	139/745 (18.0)
COVID-19 illness -ever[5]					
None	43 (55.1)	38 (46.3)	46 (50.0)	31 (43.1)	158 (48.8)
Suspected but not tested	27 (34.6)	29 (35.4)	22 (23.9)	24 (33.3)	112(34.6)
COVID-19 PCR +ve	8 (10.3)	13 (15.9)	14 (15.2)	17 (23.6)	52 (16.0)
COVID-19 +ve hospitalized	0 (0.0)	2 (2.4)	0 (0.0)	0 (0.0)	2 (0.6)

IA=inflammatory arthritis; SARDs = systemic autoimmune rheumatic disease; IBD = inflammatory bowel disease; MS= multiple sclerosis; IMID = immune mediated inflammatory disease; FT=full time; PT= part time; BMI=body mass index; JAKi= janus kinase inhibitor; TNF= tumor necrosis factor inhibitor; BNT= BNT162b2 vaccine V1= first vaccine; V2 = second vaccine, V3 = third vaccine; V4 = fourth vaccine; NA=not available, PCR= polymerase chain reaction; IQR = interquartile range

[1] IMID treatment based on most aggressive combination if on multiple agents. 1 RA subject on only prednisone monotherapy. Medication data missing for 2 SARDs subjects

[2] cardiovascular disease includes ischemic heart disease, congestive heart failure, valvular heart disease peripheral vascular disease stroke or transient ischemic attack or hypertension; respiratory disease includes asthma or chronic obstructive pulmonary disease, other endocrine includes thyroid disease, hypercholesterolemia

[3] Vaccine data available for V1 N=326 (77 IA, 84 SARDS, 72 MS 93 IBD); V2 N=324 (77IA, 83 SARDS, 72 MS 92 IBD); V3 N=281 (76 IA, 74 SARDS, 63 MS, 74 IBD) V4 N=128 (35 IA, 37 SARDs 39 IBD 17 MS).

[4] Flare assessed by the following questions: IA: “Are you having a flare? SARDs: “In the past 3 months have you had a disease flare (A flare is when your disease gets worse)? Yes moderate or severe flare;

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IBD: In the past 6 months my disease has been: Constantly active, giving me symptoms every day, or often active, giving me symptoms most days or sometimes active, giving me symptoms on some days (for instance 1-2 days/week); MS Do you feel there has been a change in your MS since your last visit? "My multiple sclerosis is much worse and in a flare"

Flare rate across IMIDs 1 month after any vaccine Chi^2 36.5 $p<0.0001$; Flare rate post V1 across IMIDs non-significant. Flare rate post V2 across IMIDs Chi^2 12.4 $p=0.006$; Flare rate post V3 across IMIDs Chi^2 18.0 $p<0.001$; Flare rate post V4 across IMIDs Chi^2 8.8 $p=0.03$.

[5] Self-reported over course of study; COVID-19 illness data missing for 2 SARDS and 1 IBD participant.

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Samples of adequate quality were obtained following the first vaccine at 1 month (n=175) and 3 months (n=44), following the second vaccine at 1 month (n=234) and 3 months (n=246), following the third vaccine at 1 month (n=215) and following the fourth vaccine at 1 month (n=85). For 31 participants, DBS samples were of inadequate quality and a serum sample from the same day was substituted. In paired DBS and serum tests from 208 subjects, seroconversion and titers were highly concordant (Kendall's tau b correlation coefficient anti-RBD BAU/ml=0.93; anti-S1 BAU/ml= 0.92) (Supplementary Figure 3). Pre-pandemic sera were seronegative for anti-SarsCo-V2 antibodies.

Following the first vaccine, 60% of IMID participants seroconverted (Table 2). Following the second vaccine, seroconversion rates increased to 91% (p=0.1 across IMIDs for both anti-S1 and anti-RBD). The change in anti-S1 seropositivity between first and second vaccines was significant (anti-S1 all IMIDs Chi^2 82.2 p<0.0001; IA Chi^2 40.5 p<0.0001; SARDs Chi^2 18.5 p<0.0001; IBD $Chi^2$16.9 p<0.0001; MS Chi^2 4.1 p=0.04 and anti-RBD all IMIDs Chi^2 55.1 p<0.0001; IA Chi^2 31.6 p<0.0001; SARDS Chi^2 18.8 p<0.0001; IBD Chi^2 8.4 p<0.01; MS Chi^2 5.1 p=0.02). Of the 21 participants who were seronegative after the second vaccine and had data following the third vaccine, 10 (42%) seroconverted after the third vaccine. Seroconversion rates for both anti-S1 and anti-RBD after the third and fourth vaccines were greater than 95%.

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Table 2 Seroconversion rates one month following each COVID-19 vaccine

Seroconversion	IA	SARDs	IBD	MS	All IMIDs
Post V1					
Anti- S1	14/47 (29.8%)	21/47 (44.7%)	33/48(68.8%)	23/33 (69.7%)	91/175 (52.0%)
Anti-RBD	18/47 (38.2%)	22/47 (46.8%)	40/48 (83.3%)	23/33 (69.7%)	103/175 (58.9%)
Anti-NC	1/47 (2.1%)	0/48 (0%)	0/47 (0%)	1/33 (3%)	2/175 (1.1%)
Post V2 [1,]					
Anti-S1 [2]	50/55 (90.9%)	46/54 (85.1%)	62/64 (96.9%)	53/61 (86.9%)	214/234 (91.5%)
Anti-RBD [3]	50/55 (90.9%)	47/54 (87.0%)	63/64 (98.4%)	54/61 (88.5%)	211/234 (90.2%)
Anti-NC	1/55 (1.8%)	0/57 (0%)	1/64 (1.6%)	1/61 (1.6%)	3/234 (1.3%)
Post V3					
Anti-S1	51/52 (98.1%)	57/62 (91.9%)	56/56 (100.0%)	41/45 (91.1%)	205/215 (95.3%)
Anti-RBD	51/52 (98.1%)	56/62 (90.3%)	56/56 (100.0%)	41/45 (91.1%)	204/215 (94.9%)
Anti-NC	2/52 (3.8%)	3/62 (4.8%)	2/56 (3.6%)	2/45 (4.4%)	9/215 (4.2%)
Post V4					
Anti-S1	31/32 (96.9%)	21/23 (91.3%)	27/27 (100%)	3/3 (100%)	82/85 (96.5%)
Anti-RBD	31/32 (96.9%)	21/23 (91.3%)	27/27 (100%)	3/3 (100%)	82/85 (96.5%)
Anti-NC	3/32 (9.4%)	4/23 (17.4%)	3/27 (11.1%)	0/3 (0.0%)	10/85 (11.8%)

IA=inflammatory arthritis; SARDs = systemic autoimmune rheumatic disease; IBD = inflammatory bowel disease; MS= multiple sclerosis; IMID = immune mediated inflammatory disease; V1 = first vaccine; V2 = second vaccine; V3= third vaccine; V4=fourth vaccine

[1] seroconversion V1 to v2 across IMIDs anti-S1 Chi^2 82.2 p<0.0001; anti-RBD Chi^2 55.1 p<0.0001

[2] change in anti-S1 seropositivity V1 to V2 IA Chi^2 40.5 p<0.0001, SARDs Chi^2 18.5 p<0.0001 IBD $Chi^2$16.9 p<0.0001; MS Chi^2 4.1 p=0.04

[3] change in anti-RBD seropositivity V1 to V2 IA Chi^2 31.6 p<0.0001, SARDS Chi^2 18.8 p<0.0001, IBD Chi^2 8.4 p<0.01, MS Chi^2 5.1 p=0.02.

Post-V2 anti-S1 seroconversion rates for IMIDs were lower compared to those of age and sex-matched controls (anti-S1 seropositive controls 363/370 (98.1%) vs IMIDs X^2 14.5 p<0.0001) but similar for anti-NC (anti-NC seropositive controls 13/370 (3.5%) vs IMIDs Chisq-2.9 p=0.1). Matched population-based estimates were not available for subsequent vaccinations.

Anti-RBD and anti-S1 IgG titres of those who seroconverted increased between the 1 month post-V1 and 1 month post-V2 time points for combined IMIDs and for each IMID individually

(Figure 1) (p values all p<0.0001). Anti-RBD and anti-S1 titers declined by 3 months post-second vaccine (p values for combined IMIDs p<0.0001; SARDs p<0.001; IBD p<0.001, IA p<0.001 and MS p<0.001) but increased 1 month post-third vaccine (p value all p<0.0001). Titers of anti-RBD and anti-Spike were similar between 1 month post-third vaccine and 1 month post-vaccine fourth vaccine. Within individuals, paired analysis of titers across visits yielded similar findings.

Over the study, 99 (30.6%) IMID participants received heterologous/mixed vaccines for their first two vaccines (Table 1). For those with one month post-V2 serology data, individuals receiving homologous vector vaccines had the lowest seroconversion rates and titers for both anti-RBD and anti-S1 responses. Individuals receiving either mRNA vaccine following a vector vaccine had comparable anti-RBD and anti-Spike titers as those receiving two mRNA vaccines (Figure 2). Anti-RBD and anti-S1 seropositivity and titers one month following the third vaccine were similar among individuals receiving different combinations of vaccines for their first two vaccines (Supplemental Table 2, Supplemental Figure 4).

Participants over age 65 years, diagnosed with MS, or taking biologics, were less likely to seroconvert by the second vaccine in multivariable models (Table 3). Vaccine mix (vector-mRNA versus mRNA-mRNA) did not impact seroconversion when included in the regression models. Most of the 29 individuals who did not seroconvert after V2 were taking immunosuppressives (mycophenolate n=8; methotrexate n= 2, azathioprine n=2) or biologics (B cell targeting currently or previously n=12, anti-TNF n=4, other n= 1) (Supplementary Table 3).

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Table 3 Clinical variables associated with seroconversion one month following second vaccine .
RBD = Receptor binding domain; S1 = Spike protein; Ref = reference category; NA = not able to compute.

	Post second vaccine OR (95% CI)	**P value**
RBD seroconversion		
Sex		
Male	Ref	1.0
Female	1.0 (0.21-4.71)	
Age		
>65 years	Ref	0.002
≤ 65 years	7.4 (2.04-27.06)	
IMID		
IBD	Ref	<0.001
RA	0.2 (0.02-2.65)	
SARDs	0.05 (0.003-0.79)	
MS	0.009 (0.001-0.13)	
Immune therapy		
None	Ref	0.004
Other immunomodulator	NA	
Immunosuppressant	0.03 (0.002-0.33)	
Biologic	0.02 (0.002-0.16)	
S1 seroconversion		
Sex		
Male	Ref	0.85
Female	1.1 (0.29-4.52)	
Age		
>65 years	Ref	0.02
≤ 65 years	3.9 (1.29-11.62)	
IMID		
IBD	Ref	<0.001
RA	0.5 (0.07-3.02)	
SARDs	0.1 (0.01-0.91)	
MS	0.03 (0.003-0.20)	
Immune therapy		
None	Ref	0.01
Other immunomodulator	NA	
Immunosuppressant	0.06 (0.006-0.51)	
Biologic	0.04 (0.007-0.27)	

Twenty-five patients were seropositive for anti-NC antibodies on at least one visit (8 IA, 8 SARDs, 3 MS, 6 IBD) and for 4 of these individuals, seropositivity persisted with declining titers across consecutive visits spanning 3 to 6 months. All but one MS participant were also anti-RBD and anti-S1 seropositive. The anti-NC titers obtained closest to COVID-19 infection (i.e. the first positive sample) were lower than anti-S1 or anti-RBD titers at the same visit [median titer (25% and 75% quartile) BAU/ml first positive test anti-NC 28.5 (18.0, 62.2); anti-S1 12443.1 (7027.5, 27143.0); anti-RBD 13851.5 (6244.87, 35743.5) $p<0.001$ for both anti-S1 and anti RBD] and across all visits [median (25% and 75% quartile) BAU/ml anti-S1 1416.3 (470.9, 4090.6) anti-RBD 1230.1 (284.4, 3747.3) $p<0.001$ for both anti-S1 and anti-RBD]. Anti-RBD and anti-S1 titers were higher in anti-NC positive compared to anti-NC negative samples [median (range; IQR) anti-RBD 11755.3 (20373.1) vs 1248.0(27-78936.2; 53278.7); anti-Spike 11254.4 (77.3-68157.0; 15352.6) vs 1313.1 (37.4-87401.3; 3106.6)]. Although the rates of anti-NC positivity increased over the course of the study, anti-NC titers did not vary by vaccine status nor by date tested (Table 2, Supplementary Figure 5).

Most participants reported no COVID-19 infection symptoms during the study, including 6 individuals who tested seropositive for anti-NC antibodies, whereas 34.6% of participants reported mild symptoms consistent with COVID-19 infection but did not have community-based confirmatory testing by either polymerase chain testing before Dec 2021 or self-administered testing after Dec 2021. All COVID-19 infections with positive community-based testing were also anti-NC positive. Only two confirmed COVID-19 infections required hospitalization. Both had received 3 vaccines and had moderate levels of anti-NC (69.4 and 22.8 BAU/ml). (Table 1, Supplementary Figure 6)

Self-reported disease activity/status scores were similar across visits for each IMID (Figure 3). Rates of moderate or severe self-reported IMID flares were similar across vaccines and IMIDs. (Table 1).

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Discussion

This single center cohort study evaluated the safety and immunogenicity of SARS-COV-2 vaccines in IMIDs and confirmed relative safety with no increase in IMID disease activity despite self-reported disease flare rates of 14% following three vaccinations. Fewer than two-thirds seroconverted after the first vaccine. Seroconversion rates differed by vaccine type with higher titres of anti-RBD and anti-Spike responses generated by mRNA vaccines compared to the available vector vaccine. Individuals who received an initial vector vaccine followed by a mRNA vaccine had vaccine induced titers that were comparable to those following a homologous mRNA vaccine course and were higher than those who received homologous vector vaccines. Anti-SARS-CoV-2 antibody titers declined 3 months after the second vaccine but improved after the third and fourth vaccines. Most individuals who did not produce adequate humoral responses to the vaccine were taking immunosuppressants or biologics.

Our findings are consistent with emerging clinical trial and cohort data from the general population and other immunocompromised groups. Prior studies of rheumatic disease flare post-COVID-19 vaccination have produced mixed results however, most found no major concerns[7]. Data for IBD and MS are also reassuring[23 24]. Several studies have found lower seroconversion rates and anti-SARS-CoV-2 titers in IMIDs compared to the general population[25 26]. While older age plays a role, the primary reason for reduced responses appears due to medication use with the greatest impact from biologics, especially B-cell depleting therapies, fingolimod, anti-tumor necrosis factor agents, and Janus kinus inhibitors, and with DMARDs such as mycophenolate and methotrexate. Hydroxychloroquine does not impair vaccine responses. Proposed strategies to optimize responses for patients on these medications include holding medication around the time of vaccination and delaying vaccination following infusion of B-cell targeted therapies [27 28]. For B cell therapies, humoral immune responses remain suboptimal even after third doses especially for individuals with low pre-vaccination cell counts[29-31]. Reassuringly, T cell responses are induced although possibly impaired[26 31 32].

Heterologous vaccine administration increased as vaccine availability and data on safety and immunogenicity evolved, thereby allowing evaluation of the role of homologous versus

heterologous vaccine administration for people with IMIDs. In the general population, clinical trials and cohort studies of mixing vaccine types that compared homologous vector, homologous mRNA and heterologous vector/mRNA vaccine courses observed greater immune responses (humoral and cellular) with mRNA vaccines than vector vaccines and that in individuals receiving a vector vaccine first, a mRNA vaccine improved vaccine responses to levels comparable to those of the homologous mRNA vaccines[12-14 33]. Our observations in IMIDs confirm that second, and at least third vaccination courses are needed to generate acceptable humoral immunogenicity and that mRNA vaccines can overcome limited responses to vector vaccines.

The clinical findings of this report reflect data collected during intermittent public health mandated societal restrictions, before and during the early period when the Omicron variant was circulating in our region, and before bivalent vaccines were available[11]. Over one-third reported mild symptoms or were suspected to have had COVID-19 illness but less than 10% had confirmed COVID-19 infection. Despite complete vaccination, infection and concerning symptoms increased as public health restrictions were relaxed, the prevalence of SARS-CoV-2 virus increased, and new variants of concern emerged. This emphasizes the need for ongoing COVID-19 surveillance to inform personal health practices given the real concerns expressed by many people with IMIDs, even those who are fully vaccinated. Recent studies have described reduced sensitivity of the anti-NC assay following vaccination that is only partially explained by viral load.[34] In this study, all participants with COVID-19 infection confirmed with community based testing were also anti-NC positive and while the number of anti-NC +ve participants increased over the study, we did not see any difference in anti-NC IgG levels with number of vaccines or by calendar month.

We acknowledge limitations of this work. Our sample size was relatively small for each IMID; however, we have collected extensive patient-reported data combined with biologic samples from individuals representing four common IMID groups, allowing cross-disease and cross-specialty comparisons which are not widely reported. We assessed IMID disease activity using validated patient self-reported disease specific indices and flare questions although these questionnaires can be subject to recall bias. Ideally patient reported IMID activity would be

supplemented with clinician assessed measures however both patient preferences and COVID-19 pandemic travel restrictions impacted the feasibility of in person clinical assessments for all participants. Self reported of disease activity/state measures correlate with clinical assessment measures[19 35-37]. As expected for these IMIDs, our population was predominantly female thus we lack power to detect sex-based differences in our outcomes and there is uncertainty as to how they would reflect a male predominant cohort. We focused on humoral vaccine induced immunogenicity using antibody seropositivity and titres as surrogates for vaccine induced protection. Antibody binding titers have been shown to correlate with neutralizing and cellular responses which in turn correlate with vaccine efficacy[38]. Further work is needed to evaluate the neutralization capacity of vaccine induced antibodies to SARS-COV-2 and emerging variants of concern including Omicron. We did not evaluate cellular immune responses yet these are critical for long term anti-viral protection especially for individuals without robust antibody responses. Additional studies are needed to evaluate if there are important differences across mRNA vaccines and vaccine intervals for optimal protection against variants of concern to inform recommendations for additional vaccinations in IMIDs. Importantly, it is still unclear what level of humoral or cellular immunogenicity is optimal to protect IMIDs against serious COVID-19 infection although population-based vaccine efficacy data are emerging for some immunocompromised groups[39].

We conclude that most individuals with IMID can safely receive COVID-19 vaccines without risk of disease flare. At least two doses that include a mRNA vaccine, either homologous or mixed vaccine types are needed to generate humoral immunity comparable to the general population. The observed decline in humoral responses support the use of third and subsequent vaccine doses for IMIDs. These data can be used to direct vaccine policies in countries where vaccine rates have been lagging or where supply has been limited.

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Author contributions:

CA Hitchon: Conceptualization, Funding acquisition, Resources (recruited participants), Data curation Formal analysis (clinical and serologic data), writing- original draft, writing- review and editing

C Mesa: Investigation (Conducted SARS-CoV-2 assays) Formal analysis (analysis of anti-SARS-CoV-2 antibody data), Data curation writing- review and editing

C Bernstein: Conceptualization, Funding acquisition, Resources (recruited participants), writing review and editing

RA Marrie: Conceptualization, Funding acquisition, Resources (recruited participants), writing review and editing

C Card: writing review and editing

S OBrian: Resources (Provided data for controls) writing review and editing

J Kim: Conceptualization, Funding acquisition, Resources (anti-SARS-CoV-2 antibody assays) conducted and interpreted anti-SARSCoV2 antibody data. writing review and editing

All authors contributed to critical review of manuscript.

Data sharing statement: Limited deidentified data from consenting participants will be made available to qualified investigators for research purposes consistent with those of the original study documents and participant consent following submission of a formal proposal, and approval by the study team, the University of Manitoba, and Shared Health and completion of signed data access agreements.

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List of Tables and Figures

Table 1 Demographics, vaccines administered, self reported IMID flare, and COVID-19 history of participants

IA=inflammatory arthritis; SARDs = systemic autoimmune rheumatic disease; IBD = inflammatory bowel disease; MS= multiple sclerosis; IMID = immune mediated inflammatory disease; FT=full time; PT= part time; BMI=body mass index; JAKi= janus kinase inhibitor; TNF= tumor necrosis factor inhibitor; BNT= BNT162b2 vaccine V1= first vaccine; V2 = second vaccine, V3 = third vaccine; V4 = fourth vaccine; NA=not available, PCR= polymerase chain reaction; IQR = interquartile range

[1] IMID treatment based on most aggressive combination if on multiple agents. 1 RA subject on only prednisone monotherapy. Medication data missing for 2 SARDs subjects

[2] cardiovascular disease includes ischemic heart disease, congestive heart failure, valvular heart disease peripheral vascular disease stroke or transient ischemic attack or hypertension; respiratory disease includes asthma or chronic obstructive pulmonary disease, other endocrine includes thyroid disease, hypercholesterolemia

[3] Vaccine data available for V1 N=326 (77 IA, 84 SARDS, 72 MS 93 IBD); V2 N=324 (77IA, 83 SARDS, 72 MS 92 IBD); V3 N=281 (76 IA, 74 SARDS, 63 MS, 74 IBD) V4 N=128 (35 IA, 37 SARDs 39 IBD 17 MS).

[4] Flare assessed by the following questions: IA: “Are you having a flare? SARDs: “In the past 3 months have you had a disease flare (A flare is when your disease gets worse)? Yes moderate or severe flare; IBD: In the past 6 months my disease has been: Constantly active, giving me symptoms every day, or often active, giving me symptoms most days or sometimes active, giving me symptoms on some days (for instance 1-2 days/week); MS Do you feel there has been a change in your MS since your last visit? “My multiple sclerosis is much worse and in a flare”

Flare rate across IMIDs 1 month after any vaccine Chi^2 36.5 $p<0.0001$; Flare rate post V1 across IMIDs non-significant. Flare rate post V2 across IMIDs Chi^2 12.4 $p=0.006$; Flare rate post V3 across IMIDs Chi^2 18.0 $p<0.001$; Flare rate post V4 across IMIDs Chi^2 8.8 $p=0.03$.

[5] Self-reported over course of study; COVID-19 illness data missing for 2 SARDS and 1 IBD participant.

Running title: COVID-19 vaccination in IMIDs

Table 2 Seroconversion rates one month following each COVID-19 vaccine

IA=inflammatory arthritis; SARDs = systemic autoimmune rheumatic disease; IBD = inflammatory bowel disease; MS= multiple sclerosis; IMID = immune mediated inflammatory disease; V1 = first vaccine; V2 = second vaccine; V3= third vaccine; V4=fourth vaccine

[1] seroconversion V1 to v2 across IMIDs anti-S1 Chi^2 82.2 p<0.0001; anti-RBD Chi^2 55.1 p<0.0001

[2] change in anti-S1 seropositivity V1 to V2 IA Chi^2 40.5 p<0.0001, SARDs Chi^2 18.5 p<0.0001 IBD $Chi^2$16.9 p<0.0001; MS Chi^2 4.1 p=0.04

[3] change in anti-RBD seropositivity IA Chi^2 31.6 p<0.0001, SARDS Chi^2 18.8 p<0.0001, IBD Chi^2 8.4 p<0.01, MS Chi^2 5.1 p=0.02.

Table 3 Clinical variables associated with seroconversion.

RBD = Receptor binding domain; S1 = Spike protein; Ref = reference category; NA = not able to compute.

Figure 1 Titers of anti- spike and anti-receptor binding domain IgG levels following first, second third and fourth vaccination.

A. All IMIDs B. IA C. SARDs D. IBD E. MS

Data for seroconverters only. IgG levels natural log transformed. IMIDs=immune mediated inflammatory disease; IA = inflammatory arthritis; SARDs = systemic autoimmune rheumatic disease; IBD = inflammatory bowel disease; MS = multiple sclerosis; S1= spike; RBD= receptor binding domain; V1 = first vaccine; V2 = second vaccine; V3= third vaccine. Unadjusted p values * p<0.0001, ** p≤0.001, *** p≤0.00, **** p<0.05

Figure 2 Titers of anti- spike and anti-receptor binding domain IgG levels 1 month following second vaccination by vaccine mixture. A log anti-RBD B log anti-Spike

Data for seroconverters only. IgG levels natural log transformed. S1= spike; RBD= receptor binding domain BNT= BNT162b2, Unadjusted p values * p<0.01

Running title: COVID-19 vaccination in IMIDs

Figure 3 Disease activity before and after each vaccine

A Inflammatory arthritis B Systemic autoimmune rheumatic disease C Inflammatory bowel disease D Multiple Sclerosis

RAPID-3 Routine Assessment of Patient Index Data 3; SLAQ Systemic lupus activity questionnaire IBDSI-SF Inflammatory bowel disease symptom inventory – short form; EDSS Expanded disability status scale. V1 vaccine one; V2 vaccine two; V3 vaccine 3; V4 vaccine 4; mo = month(s).

Supplementary Tables and Figures

Supplementary Table 1 Categorization of IMID treatments

Supplementary Table 2 Seroconversion rates based on vaccine mixture between first and second vaccinations

RBD= receptor binding domain; BNT= BNT162b2

Homologous (any) versus heterologous (any) vaccine combination anti-Spike Chi^2 10.6 Fisher exact test $p<0.0001$; anti-RBD Chi^2 8.0 Fishers exact test $p=0.004$;

Across all vaccine combinations anti-Spike Chi^2 20.1 ($p=0.001$); anti-RBD Chi^2 =13.7 ($p=0.02$)

Supplementary Table 3 Clinical features of IMIDs not seroconverting after 2 vaccinations

IA= inflammatory arthritis; SARDs= Systemic autoimmune rheumatic disease; MS= multiple sclerosis; IBD= inflammatory bowel disease M=male; F=female

Supplementary Figure 1 Covid-19 vaccination timeline

HCW= health care workers; LTCR= long term care residents, GP= general population, IMID=immune mediated inflammatory disease; Bars indicate time when study participants (IMIDs) received vaccine. Light grey bars indicate time of first vaccine (V1); dark grey bars indicate time of second vaccine (V2).

Running title: COVID-19 vaccination in IMIDs

Supplementary Figure 2 Recruitment and sample acquisition chart

Supplementary Figure 3 Correlation between assays performed using serum and dried blood spot samples.
A. log Anti-S1 B log anti-RBD, C. anti-S1 BAU/ml D anti RBD BAU/ml (Kendalls tau b correlation co-efficient anti-S1 BAU/ml= 0.92; anti-RBD BAU/ml=0.93)

Supplementary Figure 4 Titers of anti- spike and anti-receptor binding domain IgG levels 1 month following third vaccination by first and second vaccine mixture.
A log anti-RBD B log anti-Spike all comparisons p=NS

Supplementary Figure 5 Anti-Nucleocapsid antibody levels and correlations with Anti-Spike and Anti-Receptor Binding Domain antibodies

A. Anti-NC titer by Calendar month/year B. Anti-NC titer by Study visit C. Correlation of anti-NC anti RBD and anti-S1 titers in samples seropositive for anti-NC D. Correlation of anti-RBD and anti-S1 titers in all seropositive samples.

NC = nucleocapsid S = Spike; RBD = receptor binding domain; ln = natural log. V1 = vaccine 1; V2 = vaccine 2; V3 = vaccine 3; V4= vaccine 4. Values are natural log transformed BAU/ml.

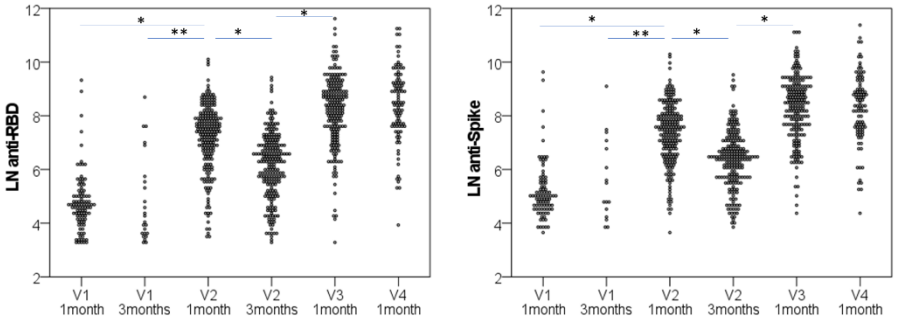
Spearman correlation coefficient anti-NC with anti-S1 = 0.06 (p=NS); anti-NC with anti-RBD = 0.03 (p=NS); anti-S with anti-RBD 0.96 p<0.001.

Supplementary Figure 6 COVID-19 infection

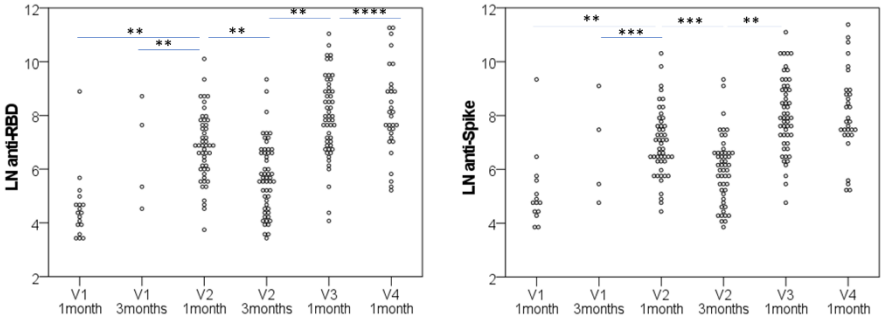
Grey line = no COVID-19 symptoms reported; Dashed grey line = COVID-19 suspected but not tested; dashed black line = COVID-19 infection confirmed by community based testing (rapid detection or polymerase chain reaction test); solid black line = proportion anti-nucleocapsid seropositive V1 vaccine one; V2 vaccine two; V3 vaccine 3; V4 vaccine 4; mo = month(s).

Running title: COVID-19 vaccination in IMIDs

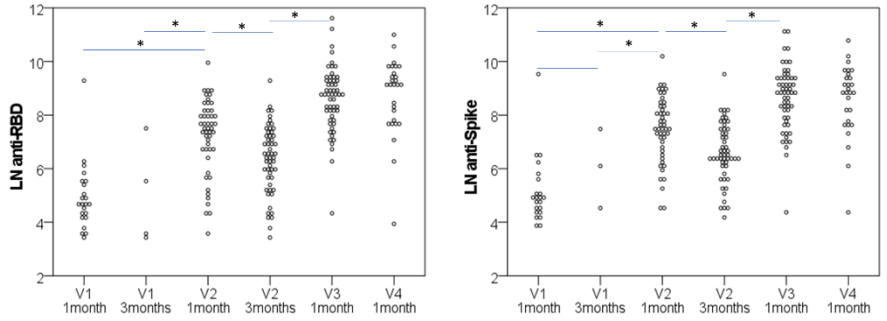
A


LN anti-RBD
LN anti-Spike
V1 1month
V1 3months
V2 1month
V2 3months
V3 1month
V4 1month

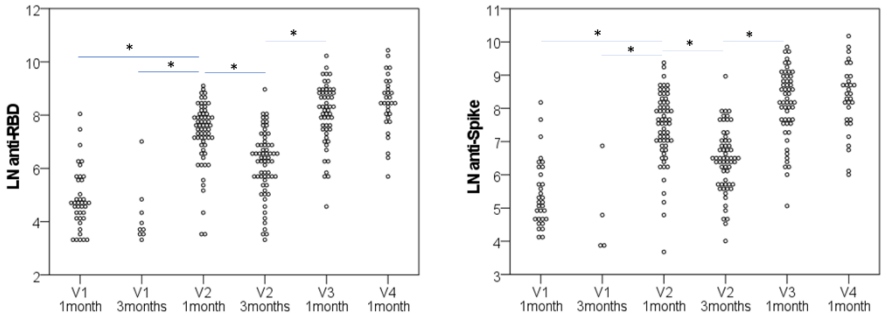
B


LN anti-RBD
LN anti-Spike
V1 1month
V1 3months
V2 1month
V2 3months
V3 1month
V4 1month

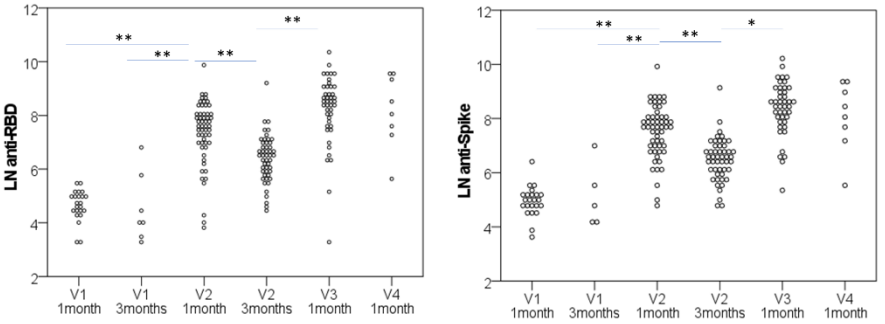
C

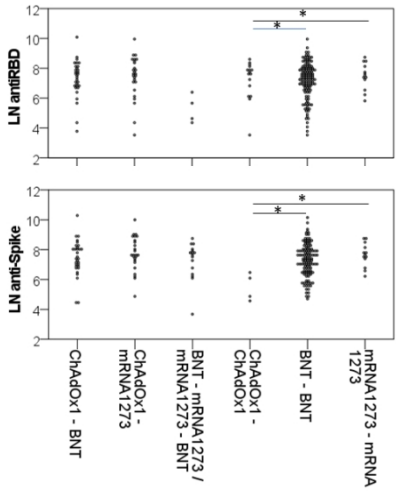

LN anti-RBD
LN anti-Spike
V1 1month
V1 3months
V2 1month
V2 3months
V3 1month
V4 1month

D


LN anti-RBD
LN anti-Spike
V1 1month
V1 3months
V2 1month
V2 3months
V3 1month
V4 1month

E


LN anti-RBD
LN anti-Spike
V1 1month
V1 3months
V2 1month
V2 3months
V3 1month
V4 1month



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Data for seroconverters only. IgG levels natural log transformed. S1= spike; RBD= receptor binding domain BNT= BNT162b2, Unadjusted p values * p<0.01

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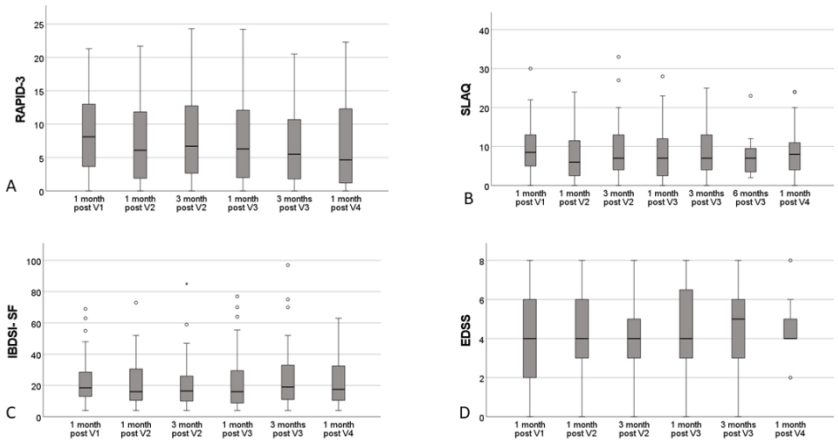

Figure 3 Disease activity before and after each vaccine
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855x481mm (38 x 38 DPI)

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Supplementary Tables and Figures

Supplementary Table 1 Categorization of IMID treatments

Treatment / Disease	Inflammatory Bowel Disease	Multiple Sclerosis	Rheumatoid Arthritis and SARDs
Corticosteroids[1]	Methylprednisolone Prednisolone) Prednisone Budesonide Hydrocortisone	Methylprednisolone Prednisolone Prednisone	Methylprednisolone) Prednisolone Prednisone Triamcinolone Cortisone
Anti-inflammatory or Immunomodulatory therapies	5-ASA Sulfasalazine	Glatiramer acetate interferon-beta 1a interferon-beta 1b dimethyl fumarate Teriflunomide Peg interferon-beta	Sulfasalazine sodium aurothiomalate auranofin aurothioglucose Penicillamine Hydroxychloroquine (
Traditional immunosuppressive therapies	Azathioprine Methotrexate 6-mercaptopurine Cyclosporine Tacrolimus	Azathioprine Methotrexate) Mitoxantrone Cyclophosphamide	Azathioprine methotrexate Cyclophosphamide Cyclosporine Leflunomide Mycophenolate Tacrolimus
Novel therapies/ Biologics	Infliximab adalimumab Golimumab Ustekinumab Vedolizumab Tofacitinib	Natalizumab Fingolimod[2] Alemtuzumab Cladribine Ocrelizumab	Infliximab) adalimumab Etanercept Anakinra Rituximab[3] Abatacept[3] Tocilizumab Tofacitinib Golimumab Certolizumab Upadacitinib Baricitinib Belimumab Sekukinumab

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Supplementary Tables and Figures

Supplementary Table 2 Seroconversion rates based on vaccine mixture between first and second vaccinations

RBD= receptor binding domain; BNT= BNT162b2

Homologous (any) versus heterologous (any) vaccine combination and seroconversion 1 month post V2 anti-Spike Chi^2 7.8 p<0.01; antiRBD Chi^2 6.8 p<0.01; 1 month post V3 anti-Spike Chi^2 0.5 p=NS; anti RBD Chi^2 0.2 p=NS

	1month post V2 Anti-Spike seroconverted n/N (%)	1month post V2 Anti-RBD seroconverted n/N (%)	1month post V3 Anti-Spike seroconverted n/N (%)	1month post V3 Anti-RBD seroconverted n/N (%)
Homologous	137/158 (86.7)	139/158 (88.0)	130/140 (92.9)	132/140 (94.3)
ChAdOX1-ChAdOX1	4/6 (66.7)	4/6 (66.7)	5/6 (83.3)	5/6 (83.3)
BNT-BNT	120/139 (84.3)	122/139 (87.8)	112/121 (92.6)	113/121 (93.4)
mRNA 1273-mRNA1273	13/13 (100)	13/13 (100)	13/13 (100.0)	13/13 (100.0)
Heterologous	69/70 (98.6)	69/70 (98.6)	64/65 (98.5)	64/65 (98.5)
ChAdOX1-BNT	30/31 (96.8)	30/31 (96.8)	27/28 (96.4)	27/28 (96.4)
ChAdOX1-mRNA1273	25/25 (100)	25/25 (100)	24/24 (100.0)	24/24 (100.0)
BNT-mRNA1273	14/14 (100)	14/14 (100)	13/13 (100.0)	13/13 (100.0)

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Supplementary Tables and Figures

Supplementary Table 3 Clinical features of IMIDs not seroconverting after 2 vaccinations

IA= inflammatory arthritis; SARDs= Systemic autoimmune rheumatic disease; MS= multiple sclerosis; IBD= inflammatory bowel disease M=male; F=female

IMID	Sex	Age baseline	Medication	vaccine combination
IA	M	82	Prednisone	BNT-BNT
IA	F	77	Methotrexate + rituximab	BNT-BNT
IA	F	76	tocilizumab	BNT-BNT
IA	F	70	methotrexate + anti-TNF	BNT-BNT
IA	F	61	mycophenolate	BNT-BNT
SARDs	F	66	rituximab + prednisone	BNT-BNT
SARDs	F	60	mycophenolate	Ch AdOX1-BNT
SARDs	F	60	mycophenolate	ChAdOX1-ChAdOX1
SARDs	F	31	rituximab + prednisone	BNT-BNT
SARDs	F	72	mycophenolate	BNT-BNT
SARDs	F	71	mycophenolate	BNT-BNT
SARDs	F	69	mycophenolate + IV Immune globulin	BNT-BNT
SARDs	F	28	mycophenolate + past rituximab	BNT-BNT
SARDs	F	69	azathioprine + past rituximab	BNT-BNT
SARDs	F	87	mycophenolate	BNT-BNT
MS	F	56	ocrelizumab	ChAdOX1-BNT
MS	M	57	ocrelizumab	BNT-BNT
MS	F	42	ocrelizumab	BNT-BNT
MS	F	74	none	BNT-BNT
MS	F	36	ocrelizumab	BNT-BNT
MS	F	66	none	BNT-BNT
MS	M	46	ocrelizumab	BNT-BNT
MS	F	48	fingolimod	BNT-BNT
MS	F	81	none	BNT-BNT
MS	F	34	ocrelizumab	BNT-BNT
IBD	F	47	anti-TNF	BNT-BNT
IBD	F	31	anti-TNF + prednisone	ChAdOX1-ChAdOX1
IBD	F	45	anti-TNF	BNT-BNT
IBD	M	79	azathioprine	BNT-BNT

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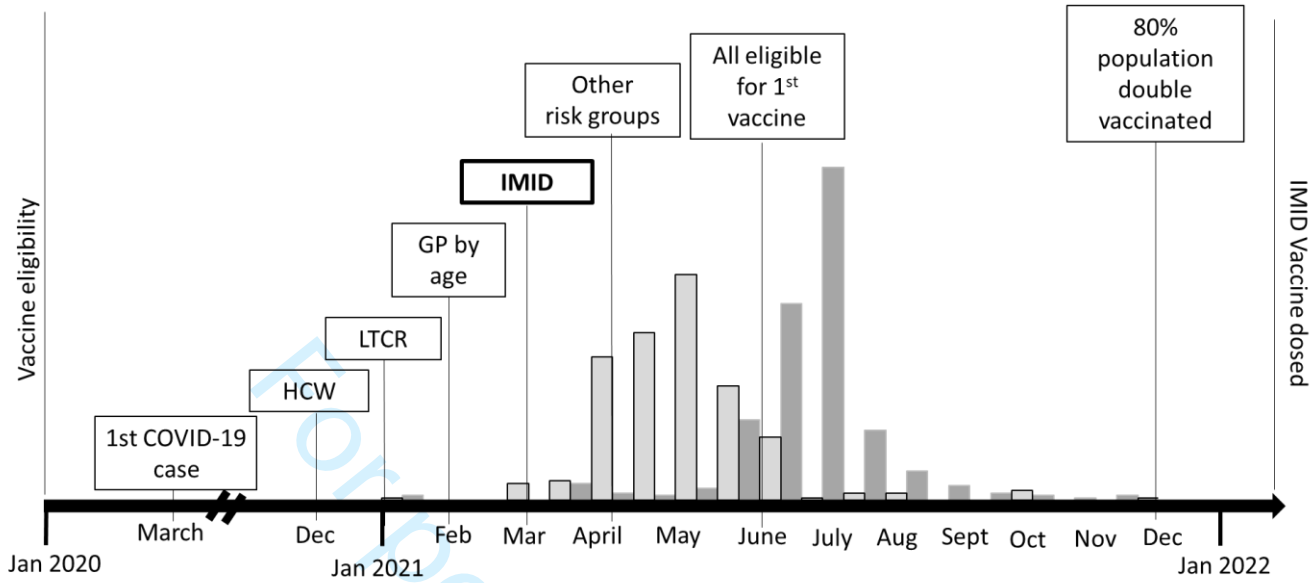
Supplementary Tables and Figures



Supplementary Figure 1 Covid-19 vaccination timeline

HCW= health care workers; LTCR= long term care residents, GP= general population, IMID=immune mediated inflammatory disease; Bars indicate time when study participants (IMIDs) received vaccine. Light grey bars indicate time of first vaccine (V1); dark grey bars indicate time of second vaccine (V2).

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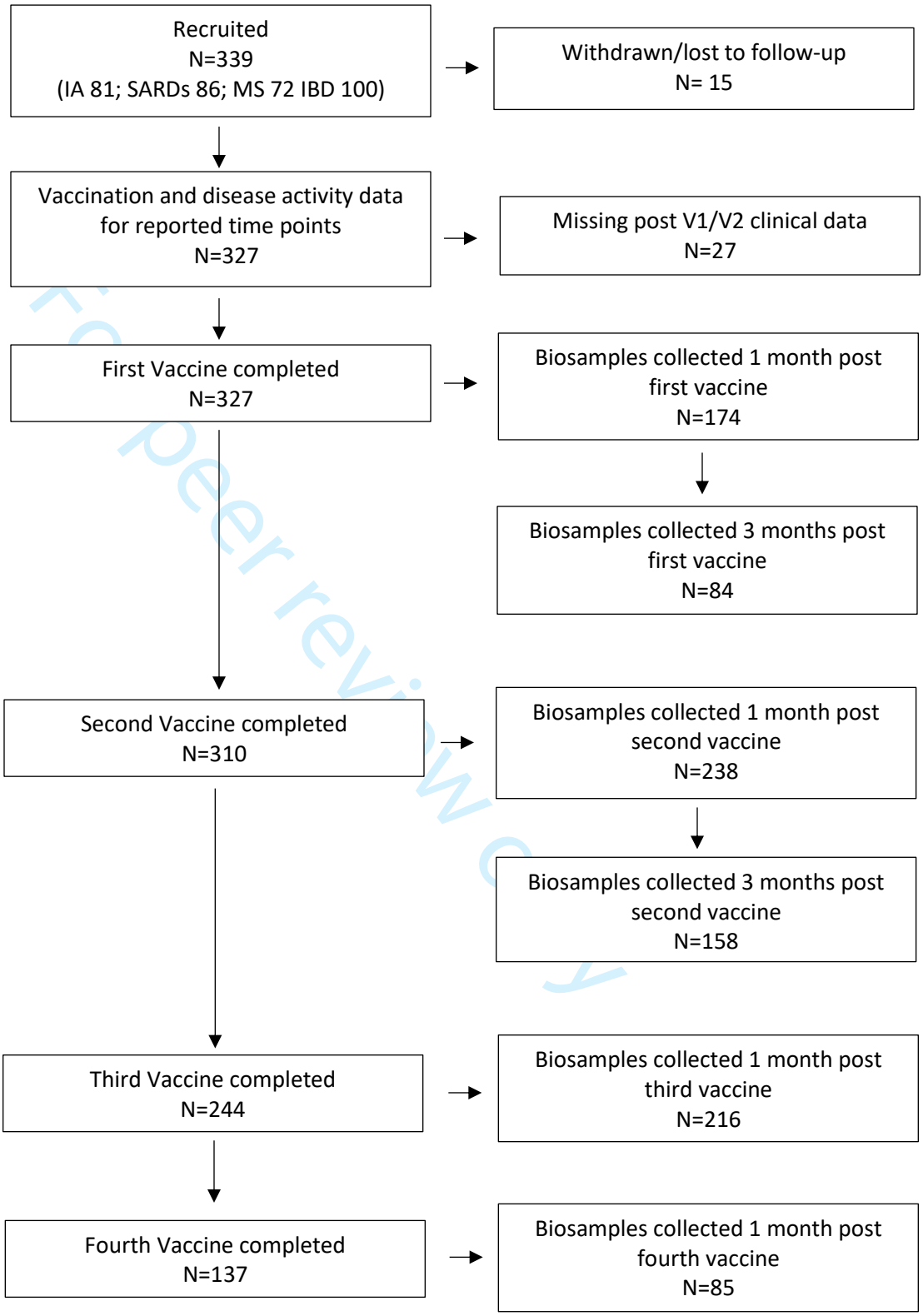
Supplementary Tables and Figures



Supplementary Figure 2 Recruitment and sample acquisition chart

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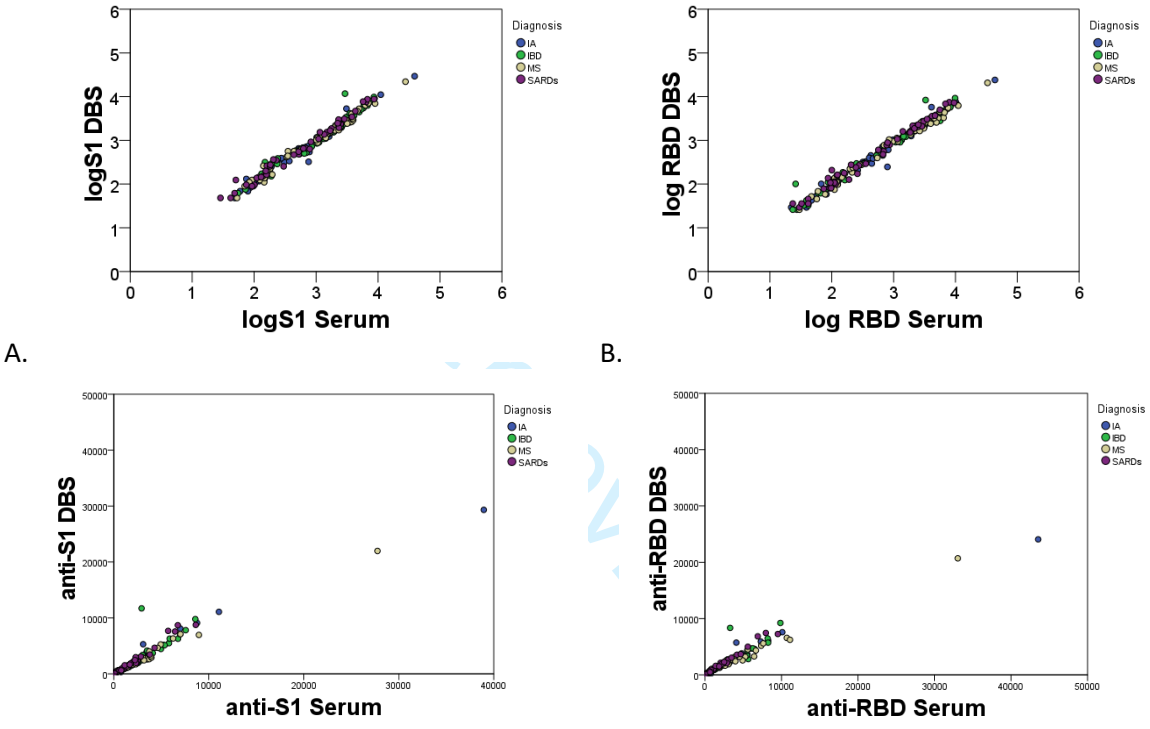
Supplementary Tables and Figures



Supplementary Figure 3 Correlation between assays performed using serum and dried blood spot samples. A. log Anti-S1 B log anti-RBD, C. anti-S1 BAU/ml D anti RBD BAU/ml (Kendalls tau b correlation co-efficient anti-S1 BAU/ml= 0.92; anti-RBD BAU/ml=0.93)

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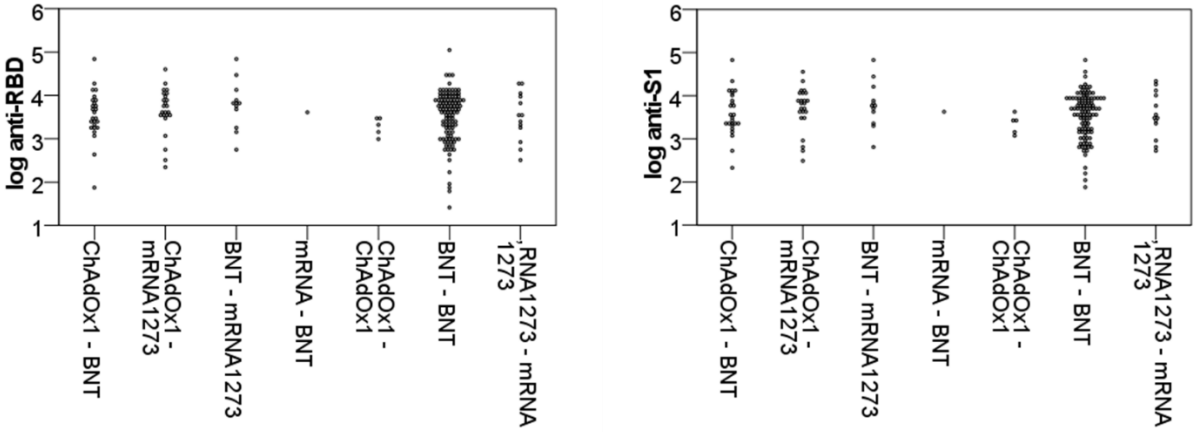
Supplementary Tables and Figures



Supplementary Figure 4 Anti-SARS-CoV2 titers 1 month post third vaccine based on first and second vaccine mixture.

RBD= Receptor binding domain; S1= Spike 1. All comparisons p=NS

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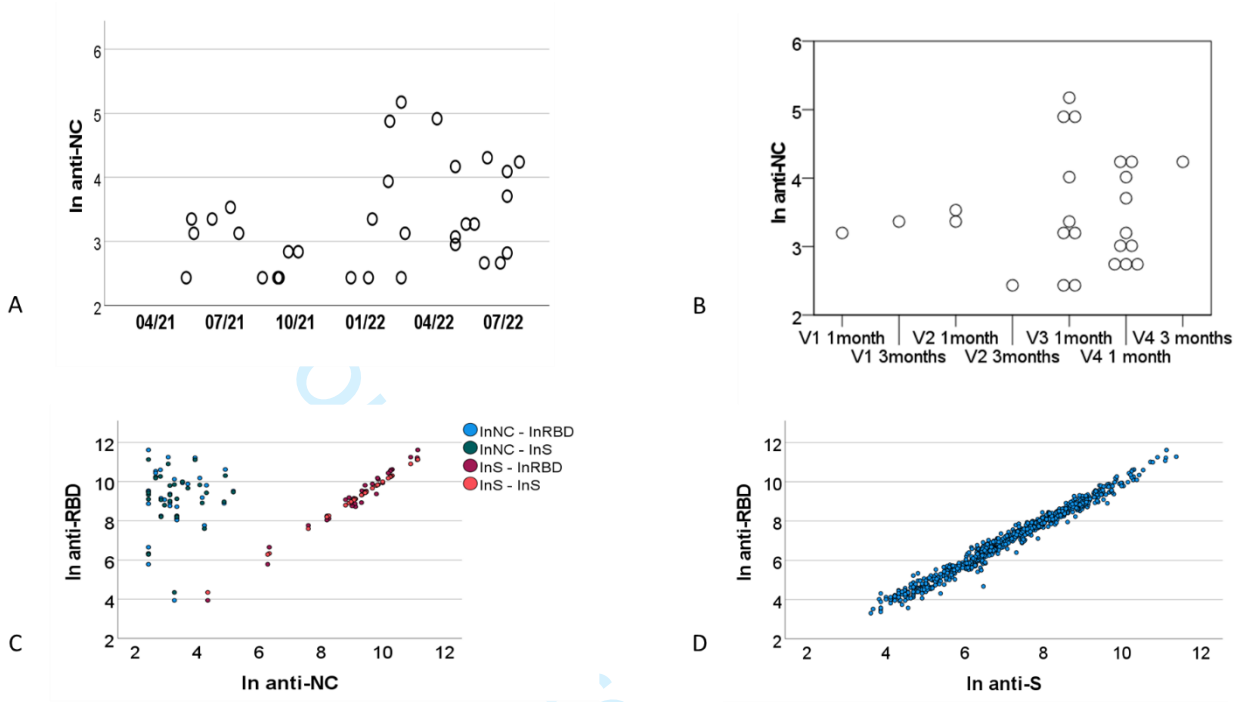
Supplementary Tables and Figures



Supplementary Figure 5 Anti-Nucleocapsid antibody levels and correlations with Anti-Spike and Anti-Receptor Binding Domain antibodies

A. Anti-NC titer by Calendar month/year B. Anti-NC titer by Study visit C. Correlation of anti-NC anti RBD and anti-S1 titers in samples seropositive for anti-NC D. Correlation of anti-RBD and anti-S1 titers in all seropositive samples.

NC = nucleocapsid S = Spike; RBD = receptor binding domain; ln = natural log. V1 = vaccine 1; V2 = vaccine 2; V3 = vaccine 3; V4= vaccine 4. Values are natural log transformed BAU/ml.

Spearman correlation coefficient anti-NC with anti-S1 = 0.06 (p=NS); anti-NC with anti-RBD = 0.03 (p=NS); anti-S with anti-RBD 0.96 p<0.001. Figure 3 Disease activity before and after each vaccine

A Inflammatory arthritis B Systemic autoimmune rheumatic disease C Inflammatory bowel disease D Multiple Sclerosis

RAPID-3 Routine Assessment of Patient Index Data 3; SLAQ Systemic lupus activity questionnaire IBDSI-SF Inflammatory bowel disease symptom inventory – short form; EDSS Expanded disability status scale. V1 vaccine one; V2 vaccine two; V3 vaccine 3; V4 vaccine 4; mo = month(s).

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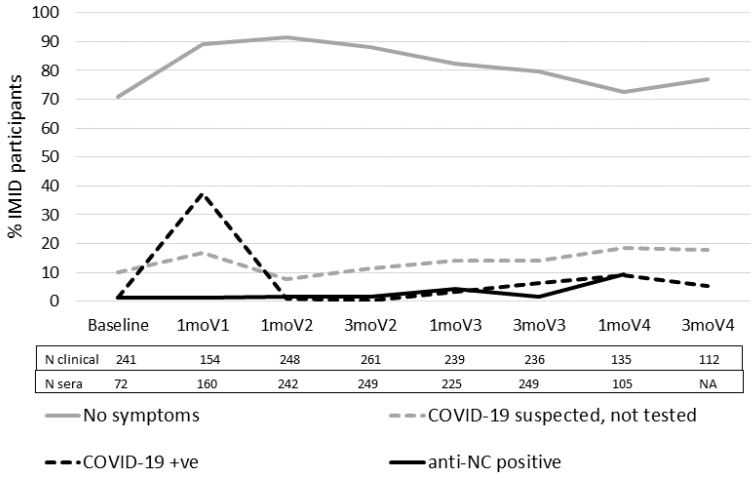

Supplementary Figure 6 COVID-19 infection

Grey line = no COVID-19 symptoms reported; Dashed grey line = COVID-19 suspected but not tested; dashed black line = COVID-19 infection confirmed by community based testing (rapid detection or polymerase chain reaction test); solid black line = proportion anti-nucleocapsid seropositive.

V1 vaccine one; V2 vaccine two; V3 vaccine 3; V4 vaccine 4.; mo = month(s)

STROBE Statement—checklist of items that should be included in reports of observational studies

	Item No	Recommendation	Page No
Title and abstract	1	(*a*) Indicate the study's design with a commonly used term in the title or the abstract	1
		(*b*) Provide in the abstract an informative and balanced summary of what was done and what was found	4
Introduction			
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported	7
Objectives	3	State specific objectives, including any prespecified hypotheses	7-8
Methods			
Study design	4	Present key elements of study design early in the paper	7-8
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection	7-8
Participants	6	(*a*) *Cohort study*—Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up *Case-control study*—Give the eligibility criteria, and the sources and methods of case ascertainment and control selection. Give the rationale for the choice of cases and controls *Cross-sectional study*—Give the eligibility criteria, and the sources and methods of selection of participants	7-8
		(*b*) *Cohort study*—For matched studies, give matching criteria and number of exposed and unexposed *Case-control study*—For matched studies, give matching criteria and the number of controls per case	
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable	8-9
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group	*7-9*
Bias	9	Describe any efforts to address potential sources of bias	7-9
Study size	10	Explain how the study size was arrived at	
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why	7-9
Statistical methods	12	(*a*) Describe all statistical methods, including those used to control for confounding	9
		(*b*) Describe any methods used to examine subgroups and interactions	9
		(*c*) Explain how missing data were addressed	8
		(*d*) *Cohort study*—If applicable, explain how loss to follow-up was addressed *Case-control study*—If applicable, explain how matching of cases and controls was addressed *Cross-sectional study*—If applicable, describe analytical methods taking account of sampling strategy	NA
		(*e*) Describe any sensitivity analyses	

Continued on next page

Results			
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed	10 and S Fig 2
		(b) Give reasons for non-participation at each stage	S Fig 2
		(c) Consider use of a flow diagram	S Fig2
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders	10 Table1
		(b) Indicate number of participants with missing data for each variable of interest	Table 1
		(c) *Cohort study*—Summarise follow-up time (eg, average and total amount)	
Outcome data	15*	*Cohort study*—Report numbers of outcome events or summary measures over time	*Table 1*
		Case-control study—Report numbers in each exposure category, or summary measures of exposure	
		Cross-sectional study—Report numbers of outcome events or summary measures	
Main results	16	(*a*) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included	10-12 Figure 1,2,4 Table 3
		(*b*) Report category boundaries when continuous variables were categorized	
		(*c*) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period	
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses	10-12
Discussion			
Key results	18	Summarise key results with reference to study objectives	12
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias	13-14
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence	13-14
Generalisability	21	Discuss the generalisability (external validity) of the study results	14
Other information			
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based	15

*Give information separately for cases and controls in case-control studies and, if applicable, for exposed and unexposed groups in cohort and cross-sectional studies.

Note: An Explanation and Elaboration article discusses each checklist item and gives methodological background and published examples of transparent reporting. The STROBE checklist is best used in conjunction with this article (freely available on the Web sites of PLoS Medicine at http://www.plosmedicine.org/, Annals of Internal Medicine at http://www.annals.org/, and Epidemiology at http://www.epidem.com/). Information on the STROBE Initiative is available at www.strobe-statement.org.

BMJ Open

Immunogenicity and safety of mixed COVID-19 vaccine regimens in patients with immune mediated inflammatory diseases: a single-centre prospective cohort study

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Date Submitted by the Author:	15-Apr-2023
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Primary Subject Heading:	Infectious diseases
Secondary Subject Heading:	Gastroenterology and hepatology, Neurology, Rheumatology, Immunology (including allergy)
Keywords:	COVID-19, RHEUMATOLOGY, GASTROENTEROLOGY, NEUROLOGY, IMMUNOLOGY

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Running title: COVID-19 vaccination in IMIDs

Immunogenicity and safety of mixed COVID-19 vaccine regimens in patients with immune mediated inflammatory diseases: a single-centre prospective cohort study

Running Title: COVID-19 vaccination in IMIDs

CA Hitchon [1], C Mesa [2], CN Bernstein [1], RA Marrie [1,3], C Card [2,4], S O'Brien [5], J Kim [2]

1 Department of Internal Medicine, Max Rady College of Medicine, Rady Faculty of Health Sciences, University of Manitoba, Winnipeg, CAN

2 National Microbiology Laboratory Public Health Agency of Canada Winnipeg, CAN

3 Department of Community Health Sciences, Max Rady College of Medicine, Rady Faculty of Health Sciences, University of Manitoba, Winnipeg, CAN

4 Department of Medical Microbiology and Infectious Diseases, Rady Faculty of Health Sciences, University of Manitoba, Winnipeg, CAN

5 Canadian Blood Services, Ottawa, CAN

Authors:

Carol A Hitchon MD MSc Department of Internal Medicine, Max Rady College of Medicine, Rady Faculty of Health Sciences, University of Manitoba, Winnipeg, CAN

Christine Mesa, MSc National Microbiology Laboratory Public Health Agency of Canada Winnipeg, CAN

Charles N Bernstein, MD Department of Internal Medicine, Max Rady College of Medicine, Rady Faculty of Health Sciences, University of Manitoba, Winnipeg, CAN

Ruth Ann Marrie, MD PhD Department of Internal Medicine, and Department of Community Health Sciences, Max Rady College of Medicine, Rady Faculty of Health Sciences, University of Manitoba, Winnipeg, CAN

Sheila O'Brien, PhD Canadian Blood Services, Ottawa, CAN

Catherine Card, PhD, National Microbiology Laboratory, Public Health Agency of Canada Winnipeg, CAN and Department of Medical Microbiology and Infectious Diseases, Rady Faculty of Health Sciences, University of Manitoba, Winnipeg, CAN

John Kim PhD National Microbiology Laboratory Public Health Agency of Canada Winnipeg, CAN

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Corresponding author

CA Hitchon

RR149 800 Sherbrook Street

Winnipeg, Manitoba

email: carol.hitchon@umanitoba.ca

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Abstract 300/300

Objective: Among persons with immune-mediated inflammatory diseases (IMIDs) who received SARS-CoV-2 vaccines, we compared post-vaccine antibody responses and IMID disease activity/states.

Design: Single centre prospective cohort study

Setting: Specialty ambulatory clinics in central Canada.

Participants: People with inflammatory arthritis (n=78; 77% rheumatoid arthritis), systemic autoimmune rheumatic diseases (n=84; 57% lupus), inflammatory bowel disease (n=93; 43% Crohn's), and multiple sclerosis (n=72; 71% relapsing-remitting) (female 79.4%, white 84.7%, mean (standard deviation) age 56.0(14.3 years) received COVID-19 vaccinations between 03/2021-09/2022.

Primary outcome: Post-vaccination anti-spike, -receptor binding domain (RBD) and -nucleocapsid (NC) IgG antibodies tested by multiplex immunoassays compared across vaccine regimens and to responses in 370 age-sex matched vaccinated controls.

Secondary Outcomes: COVID-19 infection and self-reported IMID disease activity/state.

Results: Most 216/327(66.1%) received homologous mRNA (BNT162b2 or mRNA1273) vaccines, 2.4% received homologous ChAdOx1, and 30.6% received heterologous vaccines (23.9% ChAdOx1/mRNA, 6.4% heterologous mRNA) for their first two vaccines (V1,V2). Seroconversion rates were: post-V1 anti-spike 91/175(52.0%), anti-RBD 103/175(58.9%); post-V2 anti-spike 214/234(91.5%), anti-RBD 211/234(90.2%), and were lower than controls (post-V2 anti-Spike 360/370(98.1%) p<0.0001. Antibody titers decreased 3 months post-V2 but increased 1 month post-V3 and 1 month post-V4 [BAU/ml median (interquartile range-IQR) 1 month post-V2, 3 months post-V2, 1 month post-V3, 1 month post V4 anti-S1 1835(2448), 629.1(883.4), 4757.5(7033.1), 4356.0(9393.4); anti-RBD 1686.8(2199.44), 555.8 (809.3), 4280.3(6380.6), 4792.2(11673.78)]. If primed with a vector vaccine, a mRNA vaccine increased antibody titers to those comparable to homologous mRNA vaccines. Anti-RBD and anti-Spike titers were higher in anti-NC seropositive (N=31; 25 participants) versus seronegative samples [BAU/ml median (IQR) anti-RBD 11755.3(20373.1) vs 1248.0(53278.7); anti-Spike 11254.4(15352.6) vs 1313.1(3106.6); both p<0.001). IMID disease activity/state and rates of self-reported moderate or severe IMID flare were similar across vaccinations.

Conclusion: Heterologous COVID-19 vaccination improves seroconversion rates following a vector vaccine and does not lead to IMID disease flare. IMIDs benefit from at least three vaccines.

Funding: Research Manitoba, Public Health Agency of Canada

Key Words: COVID-19, vaccination, immune systems diseases

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Strengths and limitations

- Longitudinal cohort study with systematic collection of data on COVID-19 infection, IMID disease activity and with paired biosamples for anti-SARSCoV2 IgG assays following each vaccine for up to four vaccines.
- Cross disease comparisons of four IMIDs from different medical specialties which are treated with immunocompromising medications.
- Validated measures of self-report IMID disease activity/state used to assess vaccine safety and risk of post-vaccine IMID disease flare.
- Relatively small sample size for each IMID and predominantly female population (as expected for these IMIDs) limits analysis of sex and gender effects.

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Introduction

COVID-19 vaccines have reduced rates of serious severe acute respiratory syndrome coronavirus (SARS-CoV-2) infection and mortality in the general population.[1-4] However, information on optimal vaccine strategies for immunocompromised individuals who are at increased risk of serious COVID-19 infection is limited.

Immune-mediated inflammatory diseases (IMIDs) such as autoimmune inflammatory arthritis (IA), systemic autoimmune rheumatic disease (SARD), inflammatory bowel disease (IBD), and multiple sclerosis (MS) affect 5% of the general population and share an autoimmune phenotype that affects multiple organ systems and treatment with immune therapies[5]. Due to immune-mediated disease and treatment, some people with IMIDs are at increased risk of vaccine preventable disease including serious COVID-19 infection[6-10]. While COVID-19 vaccines are effective in the general population, immune dysregulation from disease or treatment may impair vaccine responses in people with IMIDs. Due to evolving regional vaccination strategies in our region ([11] and Supplemental Figure 1), a high proportion of individuals with immunocompromised conditions received heterologous vaccine courses. Although available data on immunogenicity of these mixed vaccine courses in the general population are reassuring,[12-14] data are limited regarding the safety and immunogenicity of this strategy for immunocompromised patients as are data comparing these outcomes across diseases.

We established a cohort of patients diagnosed with any of IA, SARDs, IBD or MS to determine the safety (IMID flare) and humoral immunogenicity following COVID-19 vaccination and to assess the impact of mixing COVID-19 vaccine types. Herein, we report the clinical safety results and seroconversion results obtained after four vaccinations and compare seroconversion rates across initial vaccine combinations.

Methods:

Study Design: This single centre prospective cohort from Manitoba, Canada, was established in March 2021 and enrolled people diagnosed with any of IA, SARDs, IBD, or MS. Data was

collected until October 30th 2022. Vaccines were administered in accordance with provincial public health recommendations. Study participants were followed 1 and 3 months after each vaccine for up to four vaccines.

Study Objectives: The primary study objective was to compare post-vaccination anti-spike, -receptor binding domain (RBD) and -nucleocapsid (NC) IgG antibody seroconversion and titres across vaccine regimens. Secondary study objectives were to determine the kinetics of seropositivity and titers across vaccine doses, to compare immunogenicity across IMIDs, to determine the effect of vaccination on COVID-19 infection (efficacy), and to determine post vaccine IMID disease activity/state and self-reported IMID flare (safety).

Study Recruitment: We approached potential participants attending ambulatory clinics using multiple methods with an aim to enrol 400 individuals with a diagnosed IMID. Our sample size was limited by recruitment.

Clinical data collected: Demographic data, including birth date, sex, self-reported ethnic group according to Canadian Institute for Health Information guidelines[15], highest education level achieved, self-reported comorbidity and health behaviors such as smoking history were collected at baseline. Clinical data including IMID treatment, IMID-specific disease activity/state measures, participant reported interval COVID-19 infections with type of confirmatory test, and biosamples were collected at each visit. IMID treatment was subcategorized as anti-inflammatories and immunomodulators such as 5-ASA, sulfasalazine, hydroxychloroquine, glatiramer, and interferon therapies; traditional immunosuppressants such as methotrexate, leflunomide, azathioprine, and mycophenolate; biologic or advanced therapies such as anti-tumor necrosis factor agents, B cell depleting agents, vedolizumab, fingolimod, anti-cytokine therapies, other biologics and janus kinase inhibitors; and corticosteroids (Supplementary Table 1). Self-reported IMID disease activity/status was assessed with disease relevant validated measures (IA: Routine Assessment of Inflammatory Disease version 3 RAPID-3, Rheumatoid Arthritis Flare Core domain indices; SARDs: Systemic Lupus Activity Questionnaire SLAQ; IBD: Inflammatory Bowel Symptoms Severity Index-Short Form (IBDSI-SF), Manitoba IBD flare question; MS: self report disease activity, Expanded Disability Status Scale [EDSS])[16-20]. These

disease-related questionnaires were fully completed for IBD (248/255 97% visits), IA (208/215; 97% visits), SARDs (239/260; 92% visits), and MS (291/327; 89% visits). Data were not imputed. Individuals could attend a study visit in person or submit their data by mailed paper forms or direct entry into a REDCap electronic database hosted at the University of Manitoba[21]. Participants had the option to participate in the safety study component only.

Biosamples: Participants not attending the clinic were provided kits to collect blood by finger poke on dried blood spot (DBS) cards for postal submission[22]. Participants attending the clinic had blood collected by venipuncture and DBS were prepared before processing blood for serum. Aliquoted serum and DBS cards were stored at -20C for batch testing.

Regional population control biosample data were obtained from 370 vaccinated Canadian Blood Services blood donors who were age and sex-matched to the clinical cohort. Data on vaccine type administered was not available.

Serology: A bead-based multiplexed immunoassay was used to detect IgG anti-Spike (S1), Receptor Binding Domain (RBD) (reflecting response to vaccine), and Nucleocapsid (NC) antibodies (reflecting response to infection) (Bio-Rad Bioplex 2200 SARS-CoV-2 IgG assay). This assay was chosen based on an evaluation of several multiplex platforms using a large panel of well-pedigreed plasma-DBS specimens[22]. The quantitative measurement of the antibody response used the WHO International standard for anti-SARS-CoV-2 antibody detection. Cut-offs for seropositivity for DBS samples were established using ROC curve analysis. Cut-offs for plasma from the company were used. Sera from IA (n=19) and lupus (n=31) patients collected before the pandemic were tested to evaluate the potential for false positive SARS-CoV-2 antibody tests in IMIDs with known autoantibodies. Concordance of antibody assays in paired DBS and serum samples was assessed using the Kendalls tau b test. The controls were tested using the Roche Elecysys® anti-SARS-CoV-2 spike protein semiquantitative immunoassay (Roche Diagnostics International Ltd, Rotkreuz, Switzerland) which measures total antibodies (including IgA, IgM and IgG) to the SARS-CoV2- spike protein (anti-S) and the Roche Elecsys® anti-SARS-CoV-2 qualitative immunoassay (Roche Diagnostics LTD Rotkreuz, Switzerland) which measures

total antibodies (including IgA, IgM and IgG) to SARS-CoV-2 recombinant protein, nucleocapsid antigen (anti-NC).

Analysis: Demographic information of participants is reported using descriptive statistics including mean (standard deviation-sd), median (range or interquartile range-IQR), and counts (%). Non-parametric tests (Mann-Whitney U or Kruskal Wallis tests with Bonferroni adjustment [Badj] for multiple comparisons) were used to compare antibody levels across groups and visits. Wilcoxon signed rank tests were used to compare antibody levels across visits within individuals. Binary logistic regression was used to evaluate predictors of seroconversion one month following vaccine 2 (V2). Variables included sex, age greater than 65 years, IMID diagnosis, and treatment category (none versus biologics and advanced therapies versus immunosuppressants versus other agents), and vaccine mix (vector-mRNA versus mRNA-mRNA). Statistical analysis was conducted using IBM SPSS Statistics version 27 for Windows (IBM Corporation, Armonk, N.Y., USA).

All subjects provided informed consent. The study was approved by the University of Manitoba Health Research Ethics Board (HS24647-H2021-005), Manitoba Shared Health (SH2021:009), and the Health Canada and Public Health Agency of Canada Research Ethics Board (REB 2021-018P).

Patient/public involvement: None.

Results:

Between March 12th 2021 and July 30 2022, we recruited 339 participants (Supplementary Figure 2). Vaccination and disease activity data for the time points reported herein were available for 327 participants (78 IA: 77% RA; 84 SARDS: 57% % lupus; 93 IBD: 43 % Crohn's and 72 MS: 71% Relapsing remitting) (Table 1). Most were female (79.4%) and white (84.7%) with a mean (sd) age of 56.0 (14.3) years. Nearly one-third (30.6%) of IMID participants were taking biologics or advanced therapies.

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Table 1 Demographics, vaccines administered, self reported IMID flare, and COVID-19 history of participants

	IA N=78	SARDs N=84	IBD N=93	MS N=72	All IMIDs N=327
Age, years (mean SD)	61.9(11.8)	55.7(13.4)	53,7(16.4)	53.1(13.5)	56.0 (14.3)
Female n (%)	66 (84.6)	75 (89.3)	59(63.4)	59 (81.9)	259 (79.4)
White n (%)	66(84.6)	64 (76.2)	79(84.9)	68 (94.4)	277 (84.7)
Education Years of school (median/IQR)	15.0(4.5)	16.0(4.3)	16.0(4.0)	16.0(5.0)	16.0(4.0)
Comorbidity (n %)[2]					
Cardiovascular	35 (44.9)	31 (36.9)	26 (28.0)	22 (30.5)	115(35.1)
Pulmonary	8 (10.2)	24 (28.6)	15 (16.1)	8 (11.1)	55(16.8)
Diabetes	13 (19.7)	3 (3.7)	3 (3.4)	5 (6.9)	24(7.3)
Other endocrine	25 (32.1)	24 (28.6)	20 (21.5)	20 (27.8)	90(27.4)
Renal disease	2 (3.0)	9 (11.0)	4 (4.3)	0 (0.0)	15(4.6)
Cancer	15 (19.2)	10 (11.9)	12 (12.9)	56.9)	43 (13.10
Mental Health	17 (21.8)	27 (32.9)	27 (29.0)	22 (30.6)	94(28.7)
Total (median range)	2.5(0-10)	3(0-10)	2(0-10)	2(0-6)	2(0-10)
BMI (mean SD)	27 (6.6)	28.0 (7.8)	27.2 (6.1)	28.0 (6.2)	27.7 (6.7)
IMID Treatment level [1] n (%)					
Immunomodulators	4(5.1)	25 (30.5)	21 (22.6)	28(38.9)	78 (23.9)
Immunosuppressants	27 (34.6)	42 (51.2)	10 (10.8)	0 (0.0)	79 (24.2)
Biologics/JAKi	37(47.4)	7 (8.5)	41 (44.1)	14 (19.4)	99 (30.3)
- anti-TNF (n)	22	2	28	0	52
- anti-B cell (n)	5	6	0	10	21
- other or JAKi (n)	9	0	13	4	26
None	11 (14.1)	9 (10.7)	21(22.6)	30 (41.7)	71 (21.7)
Vaccine type V1 n (%)[3]					
CHAdOx1	15 (19.5)	28 (33.3)	17 (18.3)	26 (36.1)	86 (26.4)
BNT	60 (77.9)	56 (66.7)	66 (71.0)	39 (54.2)	221 (67.8)
mRNA1273	2 (2.6)	6 (7.1)	10 (10.8)	7 (9.7)	25 (7.7)
Vaccine type V2 n (%)[3]					
CHAdOx1	2 (2.6)	3 (3.6)	3 (3.2)	1 (1.4)	9 (2.7)
BNT	65 (84.4)	58 (68.7)	60 (65.2)	54 (75.0)	236 (72.8)
mRNA1273	16 (13.0)	23 (27.7)	29 (31.5)	17 (23.6)	79(24.4)
Vaccine type V3 n/N (%)[3]					
CHAdOx1	0	0	1 (1.3)	1 (1.6)	2 (0.7)
BNT	54 (71)	58 (78.4)	49 (66,2)	39 (61.9)	200 (71.2)
mRNA1273	16 (21.1)	16 (21.6)	24 (32.4)	23 (36.5)	79 (28.1)
Other	0	0 (0)	1 (1.4)	1 (1.6)	2 (0.7)
Vaccine type V4 n/N (%)[3]					
CHAdOx1	0 / (0)	1 (2.7)	0 (0)	0 (0)	1 (0.8)
BNT	25 (69.4)	28 (75.7)	25 (64.1)	12 (70.6)	90 (69.8)

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mRNA1273	10 (27.8)	8 (21.6)	14 (35.9)	2 (11.8))	34 (26.4)
Other	-	-	-	3 (17.6)	3 (2.3)
Vaccines for V1 and V2 n (%)					
CHAdOx1-- BNT	9 (11.5)	10 (11.9)	4 (4.3)	17 (23.6)	40(12.2)
CHAdOx1 - mRNA1287	4 (5.1)	15(17.9)	11 (11.8)	8 (11.1)	38 (11.6)
BNT – mRNA1273 or mRNA1273-BNT	4 (5.1)	5 (6.0)	10 (10.8)	2 (2.8)	21 (6.4)
BNT-CHAdOx1	0(0)	0(0)	1 (1.1)	0(0)	1 (0.3)
CHAdOx1-CHAdOx1	2 (2.6)	3 (3.6)	2 (2.2)	1 (1.4)	8 (2.4)
BNT- BNT	56(71.8)	46 (54.8)	55 (59.1)	37 (51.4)	194 (59.3)
mRNA1273 -mRNA1273	2 (2.6)	4 (4.8)	9 (9.7)	7 (9.7)	22 (6.7)
Vaccine interval Days between V1 and V2 median (range)	66 (21-97)	62 (20-188)	57 (20-98)	59 (26-97)	60 (20-188)
Self-report IMID flare [4,5]					
1 month post V1 n/N (%)	12/44 (27.3)	10/44 (22.7)	0/44 (0)	1/41 (2.4)	23/173 (13.3)
1 month post V2 n/N (%)	9/56 (16.1)	9/58 (8.6)	0/67 (0)	2/69 (2.9)	20/250 (8.0)
1 month post V3 n/N (%)	11/57 (19.3)	19/63 (30.2)	2/62 (3.3)	1/61 (1.6)	33/243 (13.6)
1 month post V4 n/N (%)	6/38 (15.8)	12/57 (21.1)	4/38 (8.3)	1/24 (4.2)	23/157 (14.6)
1 month post any V n/N(%)	38/195 (19.5)	50/202 (24.8)	6/211 (2.8)	5/129 (3.9)	99/803 (12.2)
COVID-19 illness -ever[5]					
None	43 (55.1)	38 (46.3)	46 (50.0)	31 (43.1)	158 (48.8)
Suspected but not tested	27 (34.6)	29 (35.4)	22 (23.9)	24 (33.3)	112(34.6)
COVID-19 PCR +ve	8 (10.3)	13 (15.9)	14 (15.2)	17 (23.6)	52 (16.0)
COVID-19 +ve hospitalized	0 (0.0)	2 (2.4)	0 (0.0)	0 (0.0)	2 (0.6)

IA=inflammatory arthritis (Rheumatoid Arthritis N=60, Psoriatic arthritis N=8, other Spondyloarthropathy N=3, other IA N=7); SARDs = systemic autoimmune rheumatic disease (systemic lupus erythematosus N=48, myopathy N=5, vasculitis N=7 other or unknown SARDs N=24) ; IBD = inflammatory bowel disease (crohn's disease N=43); MS= multiple sclerosis (relapsing-remitting N=51, secondary progressive N=11, primary progressive N=4, unknown N=2) ; IMID = immune mediated inflammatory disease; FT=full time; PT= part time; BMI=body mass index; JAKi= janus kinase inhibitor; TNF= tumor necrosis factor inhibitor; BNT= BNT162b2 vaccine V1= first vaccine; V2 = second vaccine, V3 = third vaccine; V4 = fourth vaccine; NA=not available, PCR= polymerase chain reaction; IQR = interquartile range

[1] IMID treatment based on most aggressive combination if on multiple agents. 1 RA subject on only prednisone monotherapy. Medication data missing for 2 SARDs subjects. Other therapy N=26 (ustekinumab n=6, vedolizumab n=7, abatacept n=1, tofacitinib n=4, tocilizumab n=3, fingolimod n=2, alemtuzumab n=1, natalizumab n=1, upadacitinib n=1)]

[2] cardiovascular disease includes ischemic heart disease, congestive heart failure, valvular heart disease peripheral vascular disease stroke or transient ischemic attack or hypertension; respiratory disease

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includes asthma or chronic obstructive pulmonary disease, other endocrine includes thyroid disease, hypercholesterolemia

[3] Vaccine data available for V1 N=326 (77 IA, 84 SARDS, 72 MS 93 IBD); V2 N=324 (77IA, 83 SARDS, 72 MS 92 IBD); V3 N=281 (76 IA, 74 SARDS, 63 MS, 74 IBD) V4 N=128 (35 IA, 37 SARDs 39 IBD 17 MS).

[4] Flare assessed by the following questions: IA: "Are you having a flare? SARDs: "In the past 3 months have you had a disease flare (A flare is when your disease gets worse)? Yes moderate or severe flare; IBD: In the past 6 months my disease has been: Constantly active, giving me symptoms every day, or often active, giving me symptoms most days or sometimes active, giving me symptoms on some days (for instance 1-2 days/week); MS Do you feel there has been a change in your MS since your last visit? "My multiple sclerosis is much worse and in a flare"

Flare rate across IMIDs 1 month after any vaccine Chi^2 72.9 $p<0.001$; Flare rate post V1 across IMIDs Chi^2 21.8 $p<0.001$. Flare rate post V2 across IMIDs Chi^2 17.7 $p<0.001$; Flare rate post V3 across IMIDs Chi^2 29.4 $p<0.001$; Flare rate post V4 across IMIDs Chi^2 10.3 $p=0.02$. IMID Flare rate across vaccination visits Chi^2 7.4 $p=0.06$.

[5] Self-reported over course of study; COVID-19 illness data missing for 2 SARDS and 1 IBD participant.

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Samples of adequate quality were obtained following the first vaccine at 1 month (n=175) and 3 months (n=44), following the second vaccine at 1 month (n=234) and 3 months (n=246), following the third vaccine at 1 month (n=215) and following the fourth vaccine at 1 month (n=85). For 31 participants, DBS samples were of inadequate quality and a serum sample from the same day was substituted. In paired DBS and serum tests from 208 subjects, seroconversion and titers were highly concordant (Kendall's tau b correlation coefficient anti-RBD BAU/ml=0.93; anti-S1 BAU/ml= 0.92) (Supplementary Figure 3). Pre-pandemic sera were seronegative for anti-SarsCo-V2 antibodies.

Following the first vaccine, 60% of IMID participants seroconverted (Table 2). Following the second vaccine, seroconversion rates increased to 91% (p=0.1 across IMIDs for both anti-S1 and anti-RBD). The change in anti-S1 seropositivity between first and second vaccines was significant (anti-S1 all IMIDs Chi^2 82.2 p<0.0001; IA Chi^2 40.5 p<0.0001; SARDs Chi^2 18.5 p<0.0001; IBD $Chi^2$16.9 p<0.0001; MS Chi^2 4.1 p=0.04 and anti-RBD all IMIDs Chi^2 55.1 p<0.0001; IA Chi^2 31.6 p<0.0001; SARDS Chi^2 18.8 p<0.0001; IBD Chi^2 8.4 p<0.01; MS Chi^2 5.1 p=0.02). Of the 20 participants who were seronegative after the second vaccine and had data following the third vaccine, 8/15 (53.3) seroconverted after the third vaccine. Seroconversion rates for both anti-S1 and anti-RBD after the third and fourth vaccines were greater than 95%.

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Table 2 Seroconversion rates one month following each COVID-19 vaccine

Seroconversion	IA	SARDs	IBD	MS	All IMIDs
Post V1					
Anti- S1	14/47 (29.8%)	21/47 (44.7%)	33/48(68.8%)	23/33 (69.7%)	91/175 (52.0%)
Anti-RBD	18/47 (38.2%)	22/47 (46.8%)	40/48 (83.3%)	23/33 (69.7%)	103/175 (58.9%)
Anti-NC	1/47 (2.1%)	0/48 (0%)	0/47 (0%)	1/33 (3%)	2/175 (1.1%)
Post V2 [1,]					
Anti-S1 [2]	50/55 (90.9%)	46/54 (85.1%)	62/64 (96.9%)	53/61 (86.9%)	214/234 (91.5%)
Anti-RBD [3]	50/55 (90.9%)	47/54 (87.0%)	63/64 (98.4%)	54/61 (88.5%)	211/234 (90.2%)
Anti-NC	1/55 (1.8%)	0/57 (0%)	1/64 (1.6%)	1/61 (1.6%)	3/234 (1.3%)
Post V3					
Anti-S1	51/52 (98.1%)	57/62 (91.9%)	56/56 (100.0%)	41/45 (91.1%)	205/215 (95.3%)
Anti-RBD	51/52 (98.1%)	56/62 (90.3%)	56/56 (100.0%)	41/45 (91.1%)	204/215 (94.9%)
Anti-NC	2/52 (3.8%)	3/62 (4.8%)	2/56 (3.6%)	2/45 (4.4%)	9/215 (4.2%)
Post V4					
Anti-S1	31/32 (96.9%)	21/23 (91.3%)	27/27 (100%)	3/3 (100%)	82/85 (96.5%)
Anti-RBD	31/32 (96.9%)	21/23 (91.3%)	27/27 (100%)	3/3 (100%)	82/85 (96.5%)
Anti-NC	3/32 (9.4%)	4/23 (17.4%)	3/27 (11.1%)	0/3 (0.0%)	10/85 (11.8%)

IA=inflammatory arthritis; SARDs = systemic autoimmune rheumatic disease; IBD = inflammatory bowel disease; MS= multiple sclerosis; IMID = immune mediated inflammatory disease; V1 = first vaccine; V2 = second vaccine; V3= third vaccine; V4=fourth vaccine

[1] seroconversion V1 to v2 across IMIDs anti-S1 Chi^2 82.2 p<0.0001; anti-RBD Chi^2 55.1 p<0.0001

[2] change in anti-S1 seropositivity V1 to V2 IA Chi^2 40.5 p<0.0001, SARDs Chi^2 18.5 p<0.0001 IBD $Chi^2$16.9 p<0.0001; MS Chi^2 4.1 p=0.04

[3] change in anti-RBD seropositivity V1 to V2 IA Chi^2 31.6 p<0.0001, SARDS Chi^2 18.8 p<0.0001, IBD Chi^2 8.4 p<0.01, MS Chi^2 5.1 p=0.02.

Post-V2 anti-S1 seroconversion rates for IMIDs were lower compared to those of age and sex-matched controls (anti-S1 seropositive controls 363/370 (98.1%) vs IMIDs X^2 14.5 p<0.0001) but similar for anti-NC (anti-NC seropositive controls 13/370 (3.5%) vs IMIDs Chisq-2.9 p=0.1). Matched population-based estimates were not available for subsequent vaccinations.

Anti-RBD and anti-S1 IgG titres of those who seroconverted increased between the 1 month post-V1 and 1 month post-V2 time points for combined IMIDs and for each IMID individually

(Figure 1) (p values all p<0.0001). Anti-RBD and anti-S1 titers declined by 3 months post-second vaccine (p values for combined IMIDs p<0.0001; SARDs p<0.001; IBD p<0.001, IA p<0.001 and MS p<0.001) but increased 1 month post-third vaccine (p value all p<0.0001). Titers of anti-RBD and anti-Spike were similar between 1 month post-third vaccine and 1 month post-vaccine fourth vaccine. Within individuals, paired analysis of titers across visits yielded similar findings.

Over the study, 99 (30.6%) IMID participants received heterologous/mixed vaccines for their first two vaccines (Table 1). For those with one month post-V2 serology data, individuals receiving homologous vector vaccines had the lowest seroconversion rates and titers for both anti-RBD and anti-S1 responses. Individuals receiving either mRNA vaccine following a vector vaccine had comparable anti-RBD and anti-Spike titers as those receiving two mRNA vaccines (Figure 2). Anti-RBD and anti-S1 seropositivity and titers one month following the third vaccine were similar among individuals receiving different combinations of vaccines for their first two vaccines (Supplemental Table 2, Supplemental Figure 4).

Participants over age 65 years, diagnosed with MS, or taking biologics, were less likely to seroconvert by the second vaccine in multivariable models. Results were similar if seroconversion was defined as seropositivity to anti-RBD and/or anti-Spike (Table 3). Vaccine mix (vector-mRNA versus mRNA-mRNA) did not impact seroconversion when included in the regression models. Most of the 20 individuals who did not seroconvert after V2 were taking immunosuppressives (mycophenolate n=5; methotrexate n= 2, azathioprine n=1) or biologics (B cell targeting currently or previously n=8, anti-TNF n=2, other n= 2) (Supplementary Table 3).

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Table 3 Clinical variables associated with seroconversion one month following second vaccine . RBD = Receptor binding domain; S1 = Spike protein; Ref = reference category; NA = not able to compute.

	Post second vaccine OR (95% CI)	**P value**
RBD seroconversion		
Sex		
Male	Ref	1.0
Female	1.0 (0.21-4.71)	
Age		
>65 years	Ref	0.002
≤ 65 years	7.4 (2.04-27.06)	
IMID		
IBD	Ref	<0.001
IA	0.2 (0.02-2.65)	
SARDs	0.05 (0.003-0.79)	
MS	0.009 (0.001-0.13)	
Immune therapy		
None	Ref	0.004
Other immunomodulator	NA	
Immunosuppressant	0.03 (0.002-0.33)	
Biologics/advanced therapies	0.02 (0.002-0.16)	
S1 seroconversion		
Sex		
Male	Ref	0.85
Female	1.1 (0.29-4.52)	
Age		
>65 years	Ref	0.02
≤ 65 years	3.9 (1.29-11.62)	
IMID		
IBD	Ref	<0.001
RA	0.5 (0.07-3.02)	
SARDs	0.1 (0.01-0.91)	
MS	0.03 (0.003-0.20)	
Immune therapy		
None	Ref	0.01
Other immunomodulator	NA	
Immunosuppressant	0.06 (0.006-0.51)	
Biologics/advanced therapies	0.04 (0.007-0.27)	

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We evaluated the impact of vaccine mix and treatment on waning of anti-SARSCo-V2 titers between 1 and 3 months following the second vaccination. In paired analysis the decline in titers between 1month post V2 and 3 months post V2 for anti-RBD and anti-S1 differed across vaccine mixtures (anti-RBD p=0.026; anti-S1 p=0.02) however this was mainly due to minimal titer changes for individuals receiving homologous vector vaccines who had lower titers overall. We observed greater titer change between those that received vector/mRNA versus mRNA/mRNA combinations for S1, but did not see differences in RBD titer change between those that received vector/mRNA versus mRNA/mRNA combinations. [median (IQR) anti-S1 mixed 1591.6 (3002.7) vs homologous mRNA 1086.3 (1608.8) p=0.021; anti-RBD mixed 1469.9 (2086.5) vs homologous mRNA 1124.5 (1402.4) p=0.051]. For individuals receiving homologous mRNA (BNT/BNT vs mRNA1273/mRNA1273) there was no difference in waning for anti-RBD or anti-S1. [RBD titer change anti-RBD: BNT/BNT 1080.6 (2405) vs mRNA1273/mRNA1273 1434.9 (2465.1) p=0.58; anti-S1 BNT/BNT 1051.9 (1674.1) vs mRNA1273/mRNA1273 1567.5 (2481.9) p=0.39]. There was no difference in waning for individuals receiving homologous mRNA vs mixed vector/mRNA of the same type for anti-RBD or anti-S1. There was no difference in titer change across different biologic categories (anti-TNF versus Bell depletion versus other biologic; anti-RBD p= 0.30; anti-S1 p=0.14) (Supplemental Figure 5).

Twenty-five participants were seropositive for anti-NC antibodies on at least one visit (8 IA, 8 SARDs, 3 MS, 6 IBD) and for 4 of these individuals, seropositivity persisted with declining titers across consecutive visits spanning 3 to 6 months. All but one MS participant were also anti-RBD and anti-S1 seropositive. Anti-RBD and anti-S1 titers were higher in anti-NC positive compared to anti-NC negative samples [median (range; IQR) anti-RBD 11755.3 (20373.1) vs 1248.0(27-78936.2; 53278.7); anti-Spike 11254.4 (77.3-68157.0; 15352.6) vs 1313.1 (37.4-87401.3; 3106.6)]. Of the 25 seropositive individuals, 9 were asymptomatic, 10 were taking biologics (5 anti-TNFs, 3 current or past B cell depletion, 4 other biologics/advanced therapies), 7 immunosuppressives (5 methotrexate, 2 azathioprine), 7 immunomodulating agents alone, and 1 MS participant was on no IMID medication (Supplemental Table 4). Although the rates of

anti-NC positivity increased over the course of the study, anti-NC titers did not vary by vaccine status (heterologous or homologous) nor by date tested (Table 2, Supplementary Figure 6).

Most participants reported no COVID-19 infection symptoms during the study, including 9 individuals who tested seropositive for anti-NC antibodies, whereas 113 (35.7%) of participants reported mild symptoms consistent with COVID-19 infection but did not have community-based confirmatory testing by either polymerase chain testing before Dec 2021 or self-administered testing after Dec 2021. All COVID-19 infections with positive community-based testing were also anti-NC positive. Only two confirmed COVID-19 infections required hospitalization. Both had received 3 vaccines and had moderate levels of anti-NC (69.4 and 22.8 BAU/ml). (Supplementary Figure 7)

Self-reported disease activity/status scores were similar across visits for each IMID (Figure 3). Rates of moderate or severe self-reported IMID flares were similar across vaccines. MS and IBD participants had lower rates of self-reported flare compared to IA and SARDs [n/N (%) with self-reported flare after any vaccine MS 5/129 (3.9), IA 38/195 (19.5), SARDs 50/202 (24.8); IBD 6/211 (2.8) (Chi^2 72.9 $p<0.001$]. Self reported IMID flare rates were similar following each vaccination (Table 1).

Discussion

This single center cohort study evaluated the safety and immunogenicity of SARS-COV-2 vaccines in IMIDs and confirmed relative safety with no increase in IMID disease activity despite self-reported disease flare rates of 12% following four vaccinations. Fewer than two-thirds seroconverted after the first vaccine. Seroconversion rates differed by vaccine type with higher titres of anti-RBD and anti-Spike responses generated by mRNA vaccines compared to the available vector vaccine. Individuals who received an initial vector vaccine followed by a mRNA vaccine had vaccine induced titers that were comparable to those following a homologous mRNA vaccine course and were higher than those who received homologous vector vaccines. Anti-SARS-CoV-2 antibody titers declined 3 months after the second vaccine but improved after

the third and fourth vaccines. Most individuals who did not produce adequate humoral responses to the vaccine were taking immunosuppressants or biologics.

Our findings are consistent with emerging clinical trial and cohort data from the general population and other immunocompromised groups. Prior studies of rheumatic disease flare post-COVID-19 vaccination have produced mixed results however, most found no major concerns[7]. Data for IBD and MS are also reassuring[23 24]. Several studies have found lower seroconversion rates and anti-SARS-CoV-2 titers in IMIDs compared to the general population[25 26]. While older age plays a role, the primary reason for reduced responses appears due to medication use with the greatest impact from biologics, especially B-cell depleting therapies, fingolimod, anti-tumor necrosis factor agents, and Janus kinus inhibitors, and with DMARDs such as mycophenolate and methotrexate. Hydroxychloroquine does not impair vaccine responses. Proposed strategies to optimize responses for patients on these medications include holding medication around the time of vaccination and delaying vaccination following infusion of B-cell targeted therapies [27 28]. For B cell therapies, humoral immune responses remain suboptimal even after third doses especially for individuals with low pre-vaccination cell counts[29-31]. Reassuringly, T cell responses are induced although possibly impaired[26 31 32].

Heterologous vaccine administration increased as vaccine availability and data on safety and immunogenicity evolved, thereby allowing evaluation of the role of homologous versus heterologous vaccine administration for people with IMIDs. In the general population, clinical trials and cohort studies of mixing vaccine types that compared homologous vector, homologous mRNA and heterologous vector/mRNA vaccine courses observed greater immune responses (humoral and cellular) with mRNA vaccines than vector vaccines and that in individuals receiving a vector vaccine first, a mRNA vaccine improved vaccine responses to levels comparable to those of the homologous mRNA vaccines[12-14 33]. Our observations in IMIDs confirm that second, and at least third vaccination courses are needed to generate acceptable humoral immunogenicity, that mRNA vaccines can overcome limited responses to vector vaccines, and that the type of mRNA administered has minimal impact on waning vaccine titers following the second vaccination.

The clinical findings of this report reflect data collected during intermittent public health mandated societal restrictions, before and during the early period when the Omicron variant was circulating in our region, and before bivalent vaccines were available[11]. Over one-third reported mild symptoms or were suspected to have had COVID-19 illness but less than 10% had confirmed COVID-19 infection. Despite complete vaccination, infection and concerning symptoms increased as public health restrictions were relaxed, the prevalence of SARS-CoV-2 virus increased, and new variants of concern emerged. This emphasizes the need for ongoing COVID-19 surveillance to inform personal health practices given the real concerns expressed by many people with IMIDs, even those who are fully vaccinated. Recent studies have described reduced sensitivity of the anti-NC assay following vaccination that is only partially explained by viral load.[34] In this study, all participants with COVID-19 infection confirmed with community based testing were also anti-NC positive and while the number of anti-NC +ve participants increased over the study, we did not see any difference in anti-NC IgG levels with number of vaccines or by calendar month.

We acknowledge limitations of this work. Our sample size was relatively small for each IMID; however, we have collected extensive patient-reported data combined with biologic samples from individuals representing four common IMID groups, allowing cross-disease and cross-specialty comparisons which are not widely reported. We assessed IMID disease activity using validated patient self-reported disease specific indices and flare questions although these questionnaires can be subject to recall bias. Ideally patient reported IMID activity would be supplemented with clinician assessed measures however both patient preferences and COVID-19 pandemic travel restrictions impacted the feasibility of in person clinical assessments for all participants. Self reported of disease activity/state measures correlate with clinical assessment measures[19 35-37]. As expected for these IMIDs, our population was predominantly female thus we lack power to detect sex-based differences in our outcomes and there is uncertainty as to how they would reflect a male predominant cohort. We focused on humoral vaccine induced immunogenicity using antibody seropositivity and titres as surrogates for vaccine induced protection. Antibody binding titers have been shown to correlate with neutralizing and cellular responses which in turn correlate with vaccine efficacy, although the titers needed to achieve

good vaccine efficacy may differ for anti-Spike and anti-RBD[38]. Further work is needed to evaluate the neutralization capacity of vaccine induced antibodies to SARS-COV-2 and emerging variants of concern including Omicron. We did not evaluate cellular immune responses yet these are critical for long term anti-viral protection especially for individuals without robust antibody responses. We were not able to confirm prior reports of the impact of different biologic categories on vaccine titers however our study was not powered for this question. Additional studies are needed to evaluate if there are important differences across mRNA vaccines and vaccine intervals for optimal protection against variants of concern to inform recommendations for additional vaccinations in IMIDs. Importantly, it is still unclear what level of humoral or cellular immunogenicity is optimal to protect IMIDs against serious COVID-19 infection although population-based vaccine efficacy data are emerging for some immunocompromised groups[39].

We conclude that most individuals with IMID can safely receive COVID-19 vaccines without risk of disease flare. At least two doses that include a mRNA vaccine, either homologous or mixed vaccine types are needed to generate humoral immunity comparable to the general population. The observed decline in humoral responses support the use of third and subsequent vaccine doses for IMIDs. These data can be used to direct vaccine policies in countries where vaccine rates have been lagging or where supply has been limited.

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Author contributions:

CA Hitchon contributed to the study conceptualization, funding acquisition, recruitment of participants, data analysis and interpretation, and writing of original and final manuscript drafts. C Mesa contributed to the data acquisition and interpretation by conducting and analyzing the SARS-CoV-2 assays and reviewed the draft manuscript. C Bernstein contributed to the study conceptualization, funding acquisition, recruitment of participants and both review and editing of the manuscript. RA Marrie contributed to the study conceptualization, funding acquisition, recruitment of participants and both review and editing of the manuscript. C Card contributed to the study conceptualization, review and editing of the manuscript. S OBrien contributed resources including data for the controls, and both review and editing of the manuscript. J Kim contributed to the study conceptualization, funding acquisition, provided resources for conducting and interpreting the SARS-CoV2 assays and contributed to the review and editing of the manuscript. All authors contributed to critical review of manuscript and approved the final version and are accountable for the work.

Data sharing statement: Limited deidentified data from consenting participants will be made available to qualified investigators for research purposes consistent with those of the original study documents and participant consent following submission of a formal proposal, and approval by the study team, the University of Manitoba, and Shared Health and completion of signed data access agreements.

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Running title: COVID-19 vaccination in IMIDs

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List of Tables and Figures

Table 1 Demographics, vaccines administered, self reported IMID flare, and COVID-19 history of participants

IA=inflammatory arthritis; SARDs = systemic autoimmune rheumatic disease; IBD = inflammatory bowel disease; MS= multiple sclerosis; IMID = immune mediated inflammatory disease; FT=full time; PT= part time; BMI=body mass index; JAKi= janus kinase inhibitor; TNF= tumor necrosis factor inhibitor; BNT= BNT162b2 vaccine V1= first vaccine; V2 = second vaccine, V3 = third vaccine; V4 = fourth vaccine; NA=not available, PCR= polymerase chain reaction; IQR = interquartile range

[1] IMID treatment based on most aggressive combination if on multiple agents. 1 RA subject on only prednisone monotherapy. Medication data missing for 2 SARDs subjects

[2] cardiovascular disease includes ischemic heart disease, congestive heart failure, valvular heart disease peripheral vascular disease stroke or transient ischemic attack or hypertension; respiratory disease includes asthma or chronic obstructive pulmonary disease, other endocrine includes thyroid disease, hypercholesterolemia

[3] Vaccine data available for V1 N=326 (77 IA, 84 SARDS, 72 MS 93 IBD); V2 N=324 (77IA, 83 SARDS, 72 MS 92 IBD); V3 N=281 (76 IA, 74 SARDS, 63 MS, 74 IBD) V4 N=128 (35 IA, 37 SARDs 39 IBD 17 MS).

[4] Flare assessed by the following questions: IA: “Are you having a flare? SARDs: “In the past 3 months have you had a disease flare (A flare is when your disease gets worse)? Yes moderate or severe flare; IBD: In the past 6 months my disease has been: Constantly active, giving me symptoms every day, or often active, giving me symptoms most days or sometimes active, giving me symptoms on some days (for instance 1-2 days/week); MS Do you feel there has been a change in your MS since your last visit? “My multiple sclerosis is much worse and in a flare”

Flare rate across IMIDs 1 month after any vaccine Chi^2 72.9 $p<0.001$; Flare rate post V1 across IMIDs Chi^2 21.8 $p<0.001$. Flare rate post V2 across IMIDs Chi^2 17.7 $p<0.001$; Flare rate post V3 across IMIDs Chi^2 29.4 $p<0.001$; Flare rate post V4 across IMIDs Chi^2 10.3 $p=0.02$. IMID Flare rate across vaccination visits Chi^2 7.4 $p=0.06$.

[5] Self-reported over course of study; COVID-19 illness data missing for 2 SARDS and 1 IBD participant.

Running title: COVID-19 vaccination in IMIDs

Table 2 Seroconversion rates one month following each COVID-19 vaccine

IA=inflammatory arthritis; SARDs = systemic autoimmune rheumatic disease; IBD = inflammatory bowel disease; MS= multiple sclerosis; IMID = immune mediated inflammatory disease; V1 = first vaccine; V2 = second vaccine; V3= third vaccine; V4=fourth vaccine

[1] seroconversion V1 to v2 across IMIDs anti-S1 Chi^2 82.2 p<0.0001; anti-RBD Chi^2 55.1 p<0.0001

[2] change in anti-S1 seropositivity V1 to V2 IA Chi^2 40.5 p<0.0001, SARDs Chi^2 18.5 p<0.0001 IBD $Chi^2$16.9 p<0.0001; MS Chi^2 4.1 p=0.04

[3] change in anti-RBD seropositivity IA Chi^2 31.6 p<0.0001, SARDS Chi^2 18.8 p<0.0001, IBD Chi^2 8.4 p<0.01, MS Chi^2 5.1 p=0.02.

Table 3 Clinical variables associated with seroconversion.

RBD = Receptor binding domain; S1 = Spike protein; Ref = reference category; NA = not able to compute.

Figure 1 Titers of anti- spike and anti-receptor binding domain IgG levels following first, second third and fourth vaccination.

A. All IMIDs B. IA C. SARDs D. IBD E. MS

Data for seroconverters only. IgG levels natural log transformed. IMIDs=immune mediated inflammatory disease; IA = inflammatory arthritis; SARDs = systemic autoimmune rheumatic disease; IBD = inflammatory bowel disease; MS = multiple sclerosis; S1= spike; RBD= receptor binding domain; V1 = first vaccine; V2 = second vaccine; V3= third vaccine. Unadjusted p values * p<0.0001, ** p≤0.001, *** p≤0.00, **** p<0.05

Figure 2 Titers of anti- spike and anti-receptor binding domain IgG levels 1 month following second vaccination by vaccine mixture. A log anti-RBD B log anti-Spike

Data for seroconverters only. IgG levels natural log transformed. S1= spike; RBD= receptor binding domain BNT= BNT162b2, Unadjusted p values * p<0.01

Running title: COVID-19 vaccination in IMIDs

Figure 3 Disease activity before and after each vaccine

A Inflammatory arthritis B Systemic autoimmune rheumatic disease C Inflammatory bowel disease D Multiple Sclerosis

RAPID-3 Routine Assessment of Patient Index Data 3; SLAQ Systemic lupus activity questionnaire IBDSI-SF Inflammatory bowel disease symptom inventory – short form; EDSS Expanded disability status scale. V1 vaccine one; V2 vaccine two; V3 vaccine 3; V4 vaccine 4; mo = month(s).

Supplementary Tables and Figures

Supplementary Table 1 Categorization of IMID treatments

Supplementary Table 2 Seroconversion rates based on vaccine mixture between first and second vaccinations

RBD= receptor binding domain; BNT= BNT162b2

Homologous (any) versus heterologous (any) vaccine combination anti-Spike Chi^2 10.6 Fisher exact test p<0.0001; anti-RBD Chi^2 8.0 Fishers exact test p=0.004;

Across all vaccine combinations anti-Spike Chi^2 20.1 (p=0.001); anti-RBD Chi^2 =13.7 (p=0.02)

Supplementary Table 3 Clinical features of IMIDs not seroconverting after 2 vaccines

IA= inflammatory arthritis; SARDs= Systemic autoimmune rheumatic disease; MS= multiple sclerosis; IBD= inflammatory bowel disease M=male; F=female

Supplementary Table 4: Clinical characteristics of anti-nucleocapsid seropositive participants

IMID = immune mediated inflammatory disease; IA= inflammatory arthritis; SARDs= Systemic autoimmune rheumatic disease; MS= multiple sclerosis; IBD= inflammatory bowel disease; NA not available (no prior visit with serology data). 1moV1 = 1 month post first vaccine, 1moV2 = 1 month post second vaccine; 3moV2 = 3 months post second vaccine; 1moV3 = 1 month post third vaccine; 1moV4 = 1 month post forth vaccine

[1] anti-Receptor Binding Domain BAU/ml; median (interquartile range) for all samples 1moV2 1686.8 (2199.44); 3moV2 555.8 (809.3); 1moV3 4280.3(6380.6)

[2] anti-Spike BAU/ml; median values for all samples 1moV2 1835 (2448); 3moV2 629.1 (883.4); 1moV3 4280.3(6380.6)

Running title: COVID-19 vaccination in IMIDs

Supplementary Figure 1 Covid-19 vaccination timeline

HCW= health care workers; LTCR= long term care residents, GP= general population, IMID=immune mediated inflammatory disease; Bars indicate time when study participants (IMIDs) received vaccine. Light grey bars indicate time of first vaccine (V1); dark grey bars indicate time of second vaccine (V2).

Supplementary Figure 2 Recruitment and sample acquisition chart

Supplementary Figure 3 Correlation between assays performed using serum and dried blood spot samples.
A. log Anti-S1 B log anti-RBD, C. anti-S1 BAU/ml D anti RBD BAU/ml (Kendalls tau b correlation co-efficient anti-S1 BAU/ml= 0.92; anti-RBD BAU/ml=0.93)

Supplementary Figure 4 Titers of anti- spike and anti-receptor binding domain IgG levels 1 month following third vaccination by first and second vaccine mixture.
A log anti-RBD B log anti-Spike all comparisons p=NS

Supplementary Figure 5 Anti-Nucleocapsid antibody levels and correlations with Anti-Spike and Anti-Receptor Binding Domain antibodies

A.Anti-NC titer by Calendar month/year B. Anti-NC titer by Study visit C. Correlation of anti-NC anti RBD and anti-S1 titers in samples seropositive for anti-NC D. Correlation of anti-RBD and anti-S1 titers in all seropositive samples.

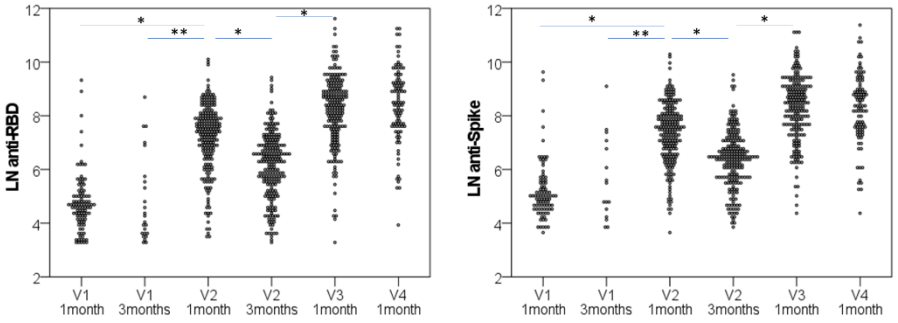
NC = nucleocapsid S = Spike; RBD = receptor binding domain; ln = natural log. V1 = vaccine 1; V2 = vaccine 2; V3 = vaccine 3; V4= vaccine 4. Values are natural log transformed BAU/ml.

Spearman correlation coefficient anti-NC with anti-S1 = 0.06 (p=NS); anti-NC with anti-RBD = 0.03 (p=NS); anti-S with anti-RBD 0.96 p<0.001.

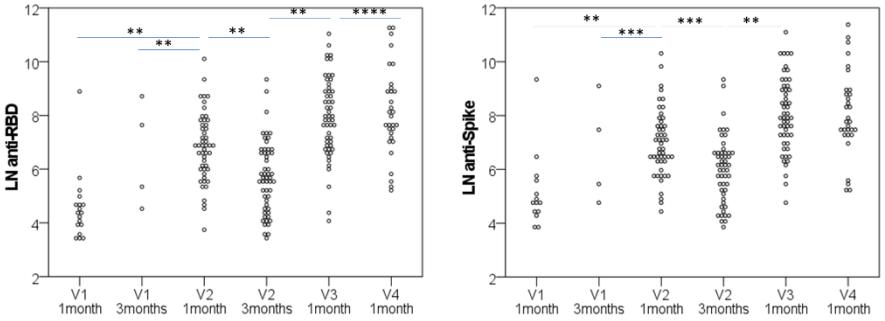
Supplementary Figure 6 COVID-19 infection

Grey line = no COVID-19 symptoms reported; Dashed grey line = COVID-19 suspected but not tested; dashed black line = COVID-19 infection confirmed by community based testing (rapid detection or polymerase chain reaction test); solid black line = proportion anti-nucleocapsid seropositive V1 vaccine one; V2 vaccine two; V3 vaccine 3; V4 vaccine 4; mo = month(s).

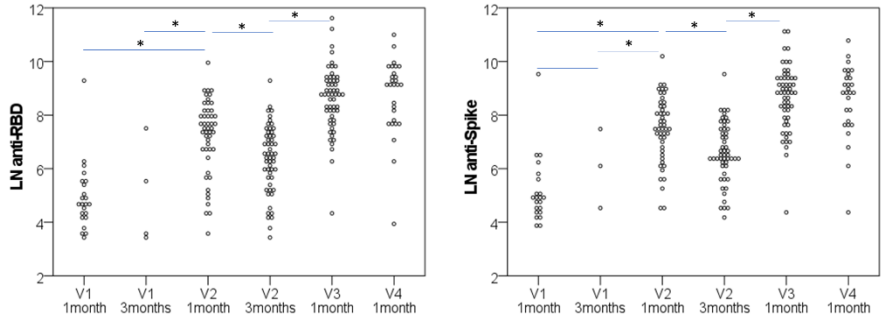
A


LN anti-RBD
LN anti-Spike
V1 1month
V1 3months
V2 1month
V2 3months
V3 1month
V4 1month

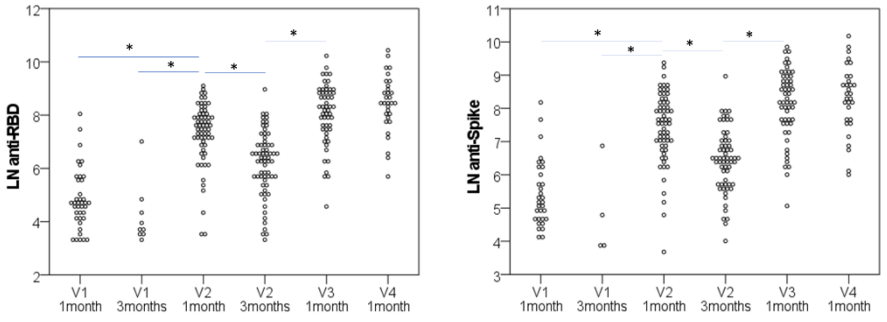
B


LN anti-RBD
LN anti-Spike
V1 1month
V1 3months
V2 1month
V2 3months
V3 1month
V4 1month

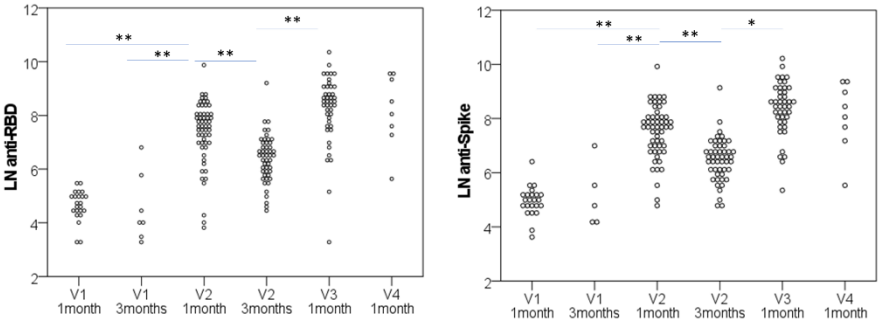
C


LN anti-RBD
LN anti-Spike
V1 1month
V1 3months
V2 1month
V2 3months
V3 1month
V4 1month

D


LN anti-RBD
LN anti-Spike
V1 1month
V1 3months
V2 1month
V2 3months
V3 1month
V4 1month

E


LN anti-RBD
LN anti-Spike
V1 1month
V1 3months
V2 1month
V2 3months
V3 1month
V4 1month

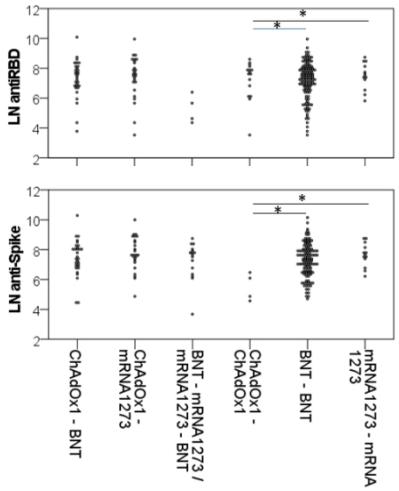


Figure 2 Titers of anti- spike and anti-receptor binding domain IgG levels 1 month following second vaccination by vaccine mixture. A log anti-RBD B log anti-Spike

Data for seroconverters only. IgG levels natural log transformed. S1= spike; RBD= receptor binding domain BNT= BNT162b2, Unadjusted p values * p<0.01

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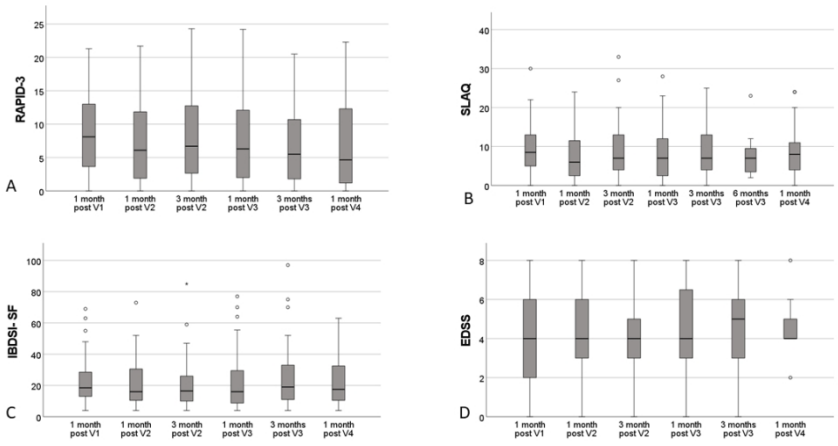

Figure 3 Disease activity before and after each vaccine
A Inflammatory arthritis B Systemic autoimmune rheumatic disease C Inflammatory bowel disease D Multiple Sclerosis
RAPID-3 Routine Assessment of Patient Index Data 3; SLAQ Systemic lupus activity questionnaire IBDSI-SF Inflammatory bowel disease symptom inventory – short form; EDSS Expanded disability status scale. V1 vaccine one; V2 vaccine two; V3 vaccine 3; V4 vaccine 4; mo = month(s).

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Supplementary Tables and Figures

Supplementary Table 1 Categorization of IMID treatments

Treatment / Disease	Inflammatory Bowel Disease	Multiple Sclerosis	Rheumatoid Arthritis and SARDs
Corticosteroids (N=37)	Methylprednisolone Prednisolone) Prednisone Budesonide Hydrocortisone	Methylprednisolone Prednisolone Prednisone	Methylprednisolone) Prednisolone Prednisone Triamcinolone Cortisone
Anti-inflammatory or Immunomodulatory therapies (N=77)	5-ASA Sulfasalazine	Glatiramer acetate interferon-beta 1a interferon-beta 1b dimethyl fumarate Teriflunomide Peg interferon-beta	Sulfasalazine sodium aurothiomalate auranofin aurothioglucose Penicillamine Hydroxychloroquine (
Traditional immunosuppressive therapies (N=78)	Azathioprine Methotrexate 6-mercaptopurine Cyclosporine Tacrolimus	Azathioprine Methotrexate) Mitoxantrone Cyclophosphamide	Azathioprine methotrexate Cyclophosphamide Cyclosporine Leflunomide Mycophenolate Tacrolimus
Novel therapies/ Biologics[1] (N=98)	Infliximab adalimumab Golimumab Ustekinumab Vedolizumab Tofacitinib	Natalizumab Fingolimod[2] Alemtuzumab Cladribine Ocrelizumab	Infliximab) Adalimumab Etanercept Anakinra Rituximab[3] Abatacept[3] Tocilizumab Tofacitinib Golimumab Certolizumab Upadacitinib Baricitinib Belimumab Sekukinumab Ustekinumab

[1.] Anti-TNF N= 51, B cell depletion N=21, Past B cell depletion N=9, Other therapy N=27 (ustekinumab n=6, vedolizumab n=6, tofacitinib n=4, tocilizumab n=2, fingolimod n=2, alemtuzumab n=1, natalizumab n=1, updacitinib n=1)

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Supplementary Tables and Figures

Supplementary Table 2 Seroconversion rates based on vaccine mixture between first and second vaccinations

RBD= receptor binding domain; BNT= BNT162b2

Homologous (any) versus heterologous (any) vaccine combination and seroconversion 1 month post V2 anti-Spike Chi^2 7.8 p<0.01; antiRBD Chi^2 6.8 p<0.01; 1 month post V3 anti-Spike Chi^2 0.5 p=NS; anti RBD Chi^2 0.2 p=NS

	1month post V2 Anti-Spike seroconverted n/N (%)	1month post V2 Anti-RBD seroconverted n/N (%)	1month post V3 Anti-Spike seroconverted n/N (%)	1month post V3 Anti-RBD seroconverted n/N (%)
Homologous	137/158 (86.7)	139/158 (88.0)	130/140 (92.9)	132/140 (94.3)
ChAdOX1-ChAdOX1	4/6 (66.7)	4/6 (66.7)	5/6 (83.3)	5/6 (83.3)
BNT-BNT	120/139 (84.3)	122/139 (87.8)	112/121 (92.6)	113/121 (93.4)
mRNA 1273-mRNA1273	13/13 (100)	13/13 (100)	13/13 (100.0)	13/13 (100.0)
Heterologous	69/70 (98.6)	69/70 (98.6)	64/65 (98.5)	64/65 (98.5)
ChAdOX1-BNT	30/31 (96.8)	30/31 (96.8)	27/28 (96.4)	27/28 (96.4)
ChAdOX1-mRNA1273	25/25 (100)	25/25 (100)	24/24 (100.0)	24/24 (100.0)
BNT-mRNA1273	14/14 (100)	14/14 (100)	13/13 (100.0)	13/13 (100.0)

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Supplementary Table 3 Clinical features of IMIDs not seroconverting after 2 vaccinations

IMID = immune mediated inflammatory disease; IA= inflammatory arthritis; SARDs= Systemic autoimmune rheumatic disease; MS= multiple sclerosis; IBD= inflammatory bowel disease M=male; F=female

IMID	Sex	Age baseline	Medication	vaccine combination
IA	M	82	Prednisone	BNT-BNT
IA	F	77	Methotrexate + rituximab	BNT-BNT
IA	F	76	tocilizumab	BNT-BNT
IA	F	70	methotrexate + anti-TNF	BNT-BNT
IA	F	61	mycophenolate	BNT-BNT
SARDs	F	66	rituximab + prednisone	BNT-BNT
SARDs	F	60	mycophenolate	ChAdOX1-ChAdOX1
SARDs	F	31	rituximab + prednisone	BNT-BNT
SARDs	F	72	mycophenolate	BNT-BNT
SARDs	F	71	mycophenolate	BNT-BNT
SARDs	F	69	mycophenolate + IV Immune globulin	BNT-BNT
SARDs	F	69	azathioprine + past rituximab	BNT-BNT
MS	F	56	ocrelizumab	ChAdOX1-BNT
MS	M	57	ocrelizumab	BNT-BNT
MS	F	74	none	BNT-BNT
MS	F	36	ocrelizumab	BNT-BNT
MS	F	48	fingolimod	BNT-BNT
MS	F	81	none	BNT-BNT
MS	F	34	ocrelizumab	BNT-BNT
IBD	F	31	anti-TNF + prednisone	ChAdOX1-ChAdOX1

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Supplementary table 4: Clinical characteristics of anti-nucleocapsid seropositive participants

IMID	Age (years)	First visit tested positive	Preceding visit with serology	Anti-RBD titer at preceding visit[1]	Anti-S1 titer at preceding visit[2]	First and second vaccine	Last vaccine prior to infection	IMID treatment
IA	63	1moV4	1moV3	76752	11941	v/mRNA	mRNA1273	JAKi
IA	63	1moV3	3moV1	NA	NA	mRNA/mRNA	mRNA1273	HDQ;MTX
IA	65	3moV4	3moV2	1547	1505	mRNA/mRNA	BNT	HDQ
IA	69	1moV4	1moV3	1365	1526	mRNA/mRNA	mRNA1273	HDQ aTNF
IA	53	1moV3	3moV2	923	178	mRNA/mRNA	BNT	HDQ, MTX
IA	68	1moV4	1moV3	4680	3403	mRNA/mRNA	BNT	aTNF. Pred
IA	49	1moV1	NA	NA	NA	mRNA/mRNA	BNT	HDQ MTX
IA	62	1moV1	NA	NA	NA	mRNA/mRNA	BNT	HDQ
SARDs	61	1moV4	1moV3	2756	2825	v/v	BNT	HDQ
SARDs	60	1moV3	3moV2	867	556	mRNA/mRNA	BNT	HDQ
SARDs	62	1moV4	1moV3	17969	12938	mRNA/mRNA	BNT	MTX
SARDs	44	1moV3	3moV2	172	254	mRNA/mRNA	BNT	MTX
SARDs	64	1moV4	3moV2	74	96	v/mRNA	BNT	Ritux
SARDs	38	1moV4	1moV3	3734	4104	v/mRNA	mRNA1273	AZA
SARDs	39	3moV2	NA	NA	NA	v/mRNA	mRNA1273	AZA
SARDs	30	1moV4	NA	NA	NA	mRNA/mRNA	BNT	aTNF
MS	53	1moV2	NA	NA	NA	v/mRNA	mRNA1273	GLA
MS	75	1moV1	NA	NA	NA	mRNA/mRNA	BNT	None
MS	56	1moV3	3moV3	2304	2811	v/mRNA	mRNA1273	INF
IBD	50	1moV2	NA	NA	NA	mRNA/mRNA	BNT	VED
IBD	67	1moV4	3moV2	591	584	mRNA/mRNA	mRNA1273	VED
IBD	30	1moV4	1moV3	1339	1464	mRNA/mRNA	BNT	UST
IBD	20	1moV4	1moV3	6221	6375	mRNA/mRNA	BNT	aTNF
IBD	64	1moV3	3moV2	497	536	v/mRNA	mRNA1273	5-ASA
IBD	45	1moV3	3moV2	Neg	neg	mRNA/mRNA	BNT	aTNF

IMID = immune mediated inflammatory disease; IA= inflammatory arthritis; SARDs= Systemic autoimmune rheumatic disease; MS= multiple sclerosis; IBD= inflammatory bowel disease; NA not available (no prior visit with serology data). 1moV1 = 1 month post first vaccine, 1moV2 = 1 month post second vaccine; 3moV2 = 3 months post second vaccine; 1moV3 = 1 month post third vaccine; 1moV4 = 1 month post forth vaccine

[1] anti-Receptor Binding Domain BAU/ml; median (interquartile range) for all samples 1moV2 1686.8 (2199.44); 3moV2 555.8 (809.3); 1moV3 4280.3(6380.6)

[2] anti-Spike BAU/ml; median values for all samples 1moV2 1835 (2448); 3moV2 629.1 (883.4); 1moV3 4280.3(6380.6)

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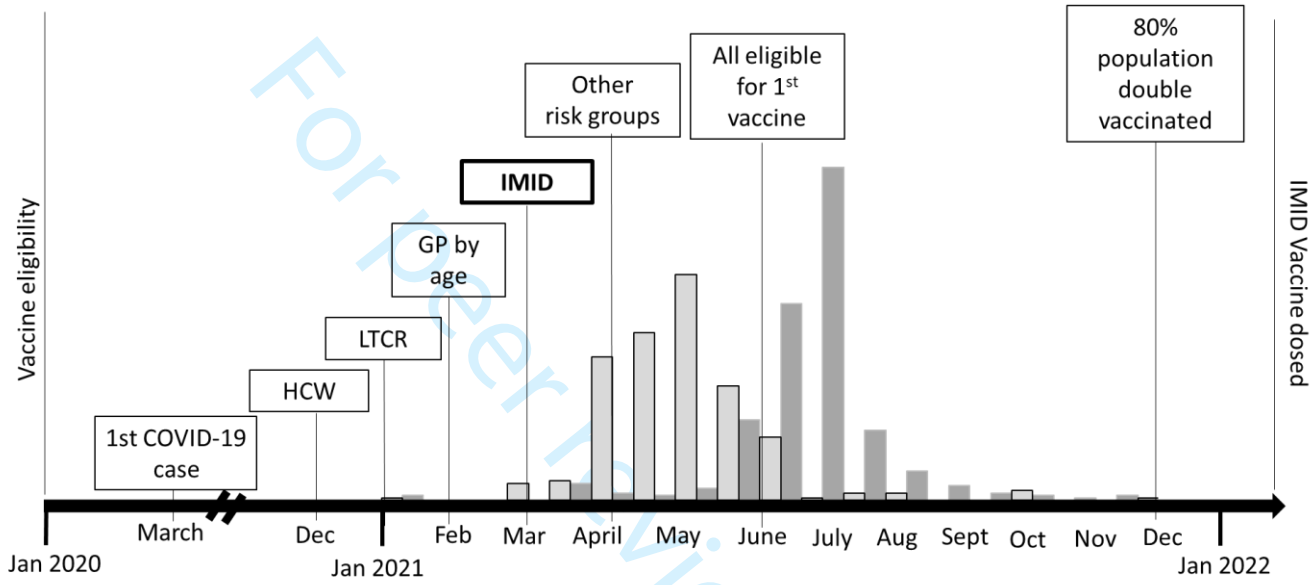

Supplementary Figure 1 Covid-19 vaccination timeline

HCW= health care workers; LTCR= long term care residents, GP= general population, IMID=immune mediated inflammatory disease; Bars indicate time when study participants (IMIDs) received vaccine. Light grey bars indicate time of first vaccine (V1); dark grey bars indicate time of second vaccine (V2).

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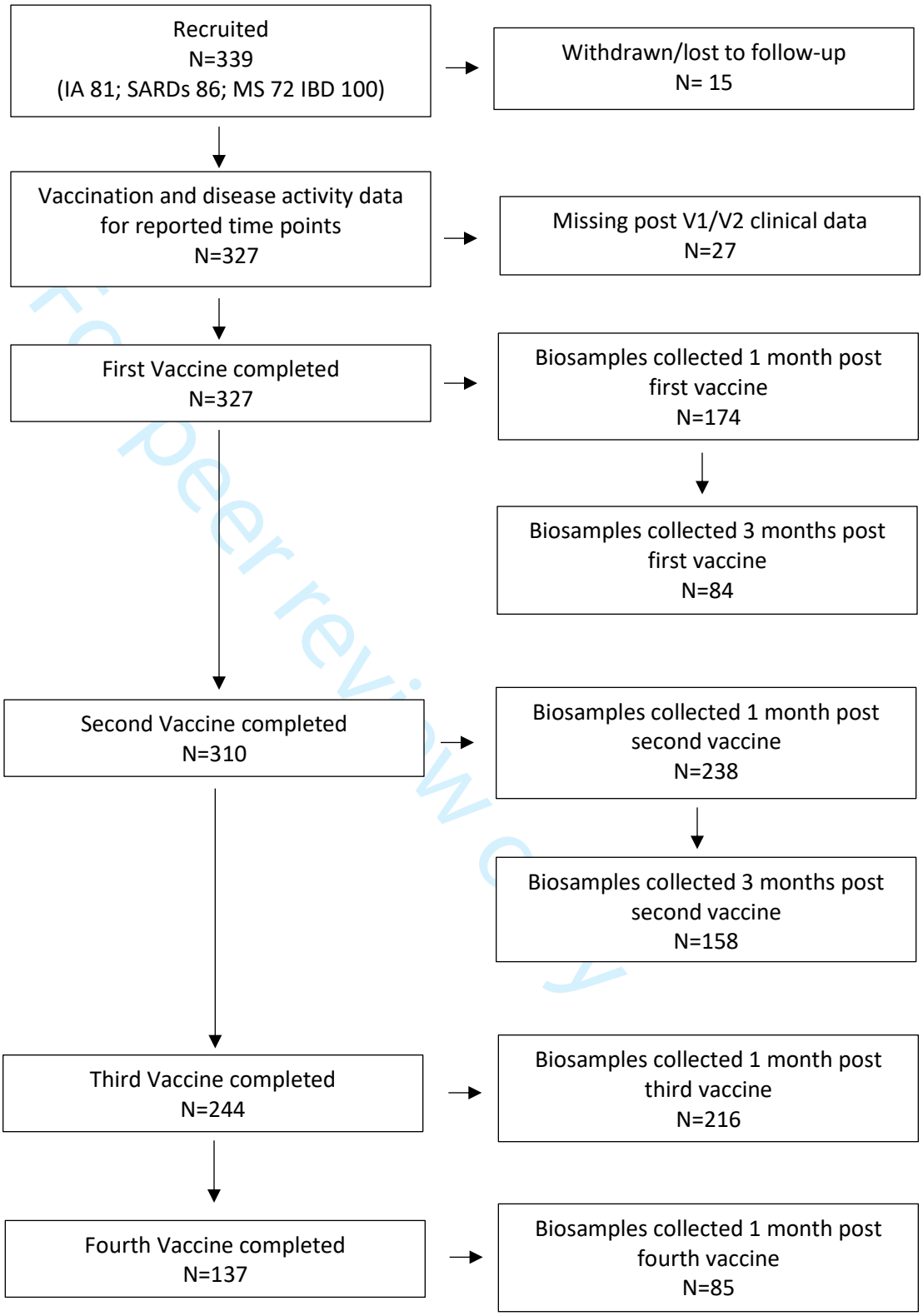
Supplementary Tables and Figures



Supplementary Figure 2 Recruitment and sample acquisition chart

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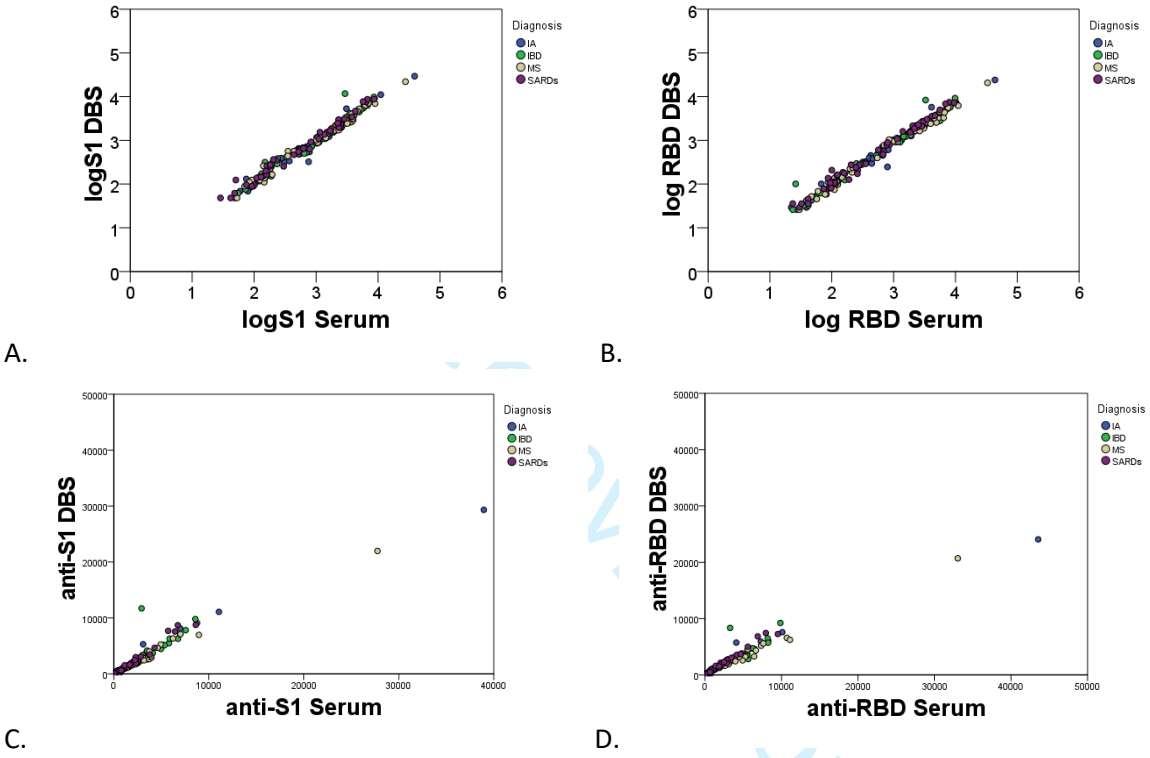

Supplementary Figure 3 Correlation between assays performed using serum and dried blood spot samples. A. log Anti-S1 B log anti-RBD, C. anti-S1 BAU/ml D anti RBD BAU/ml (Kendalls tau b correlation co-efficient anti-S1 BAU/ml= 0.92; anti-RBD BAU/ml=0.93)

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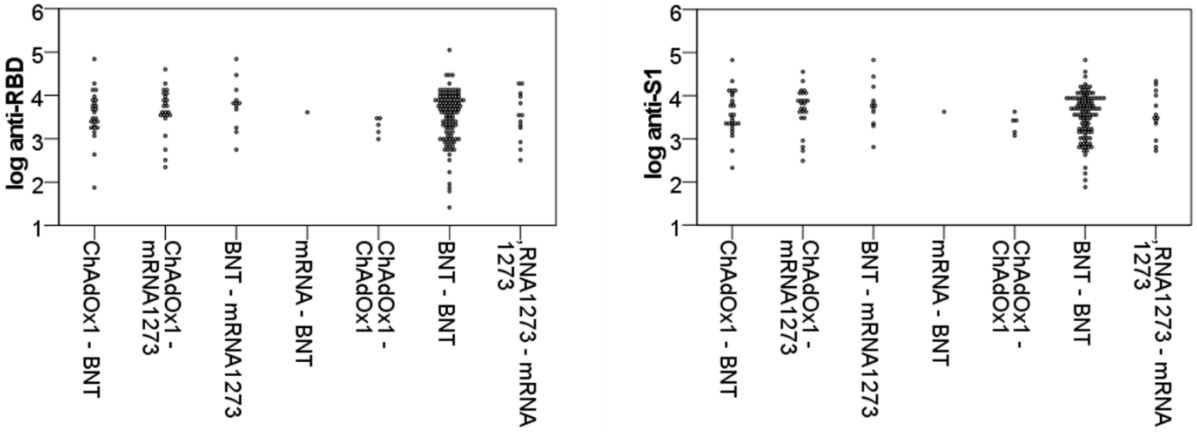

Supplementary Figure 4 Anti-SARS-CoV2 titers 1 month post third vaccine based on first and second vaccine mixture.

RBD= Receptor binding domain; S1= Spike 1. All comparisons p=NS

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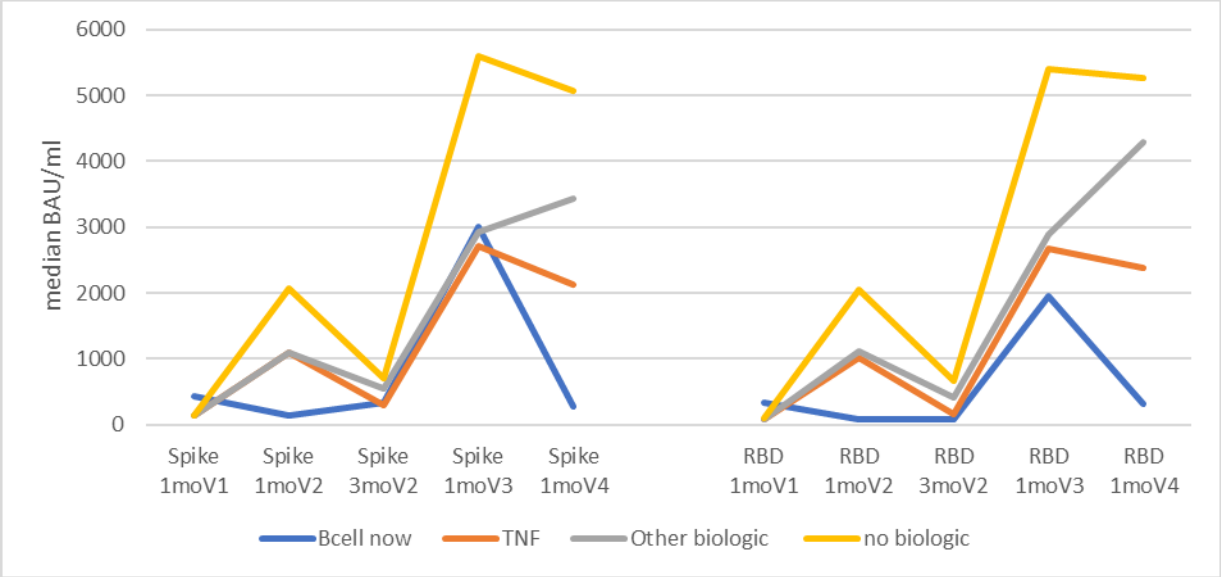

	B cell targeting now	anti-TNF	other biologic	no biologic	B cell targeting past
N 1moV1	2	14	8	77	2
N 1moV2	5	37	16	152	4
N 3moV2	5	34	20	154	5
N 1moV3	8	34	19	140	5
N 1moV4	5	18	11	46	5

Supplementary Figure 5 Median titers of anti-Spike and anti-RBD for individuals on different biologic categories and number participants in each treatment category for each visit.

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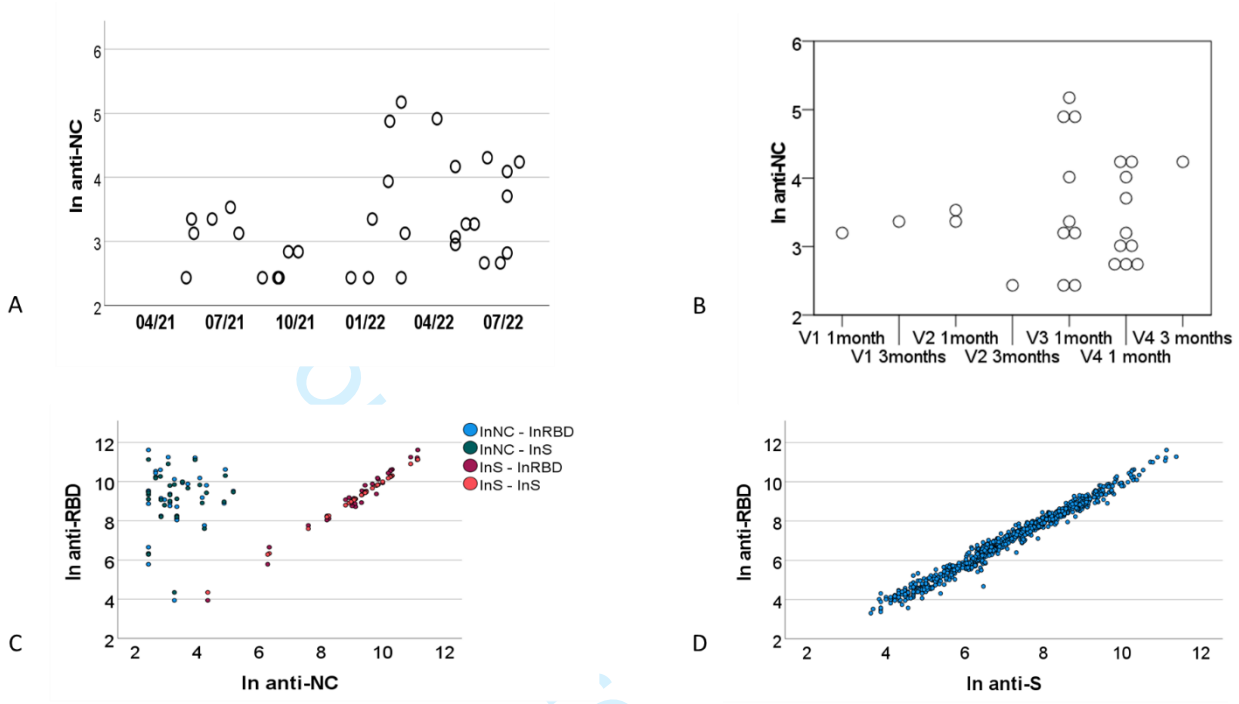
Supplementary Tables and Figures



Supplementary Figure 6 Anti-Nucleocapsid antibody levels and correlations with Anti-Spike and Anti-Receptor Binding Domain antibodies

A. Anti-NC titer by Calendar month/year B. Anti-NC titer by Study visit C. Correlation of anti-NC anti RBD and anti-S1 titers in samples seropositive for anti-NC D. Correlation of anti-RBD and anti-S1 titers in all seropositive samples.

NC = nucleocapsid S = Spike; RBD = receptor binding domain; ln = natural log. V1 = vaccine 1; V2 = vaccine 2; V3 = vaccine 3; V4= vaccine 4. Values are natural log transformed BAU/ml.

Spearman correlation coefficient anti-NC with anti-S1 = 0.06 (p=NS); anti-NC with anti-RBD = 0.03 (p=NS); anti-S with anti-RBD 0.96 p<0.001. Figure 3 Disease activity before and after each vaccine

A Inflammatory arthritis B Systemic autoimmune rheumatic disease C Inflammatory bowel disease D Multiple Sclerosis

RAPID-3 Routine Assessment of Patient Index Data 3; SLAQ Systemic lupus activity questionnaire IBDSI-SF Inflammatory bowel disease symptom inventory – short form; EDSS Expanded disability status scale. V1 vaccine one; V2 vaccine two; V3 vaccine 3; V4 vaccine 4; mo = month(s).

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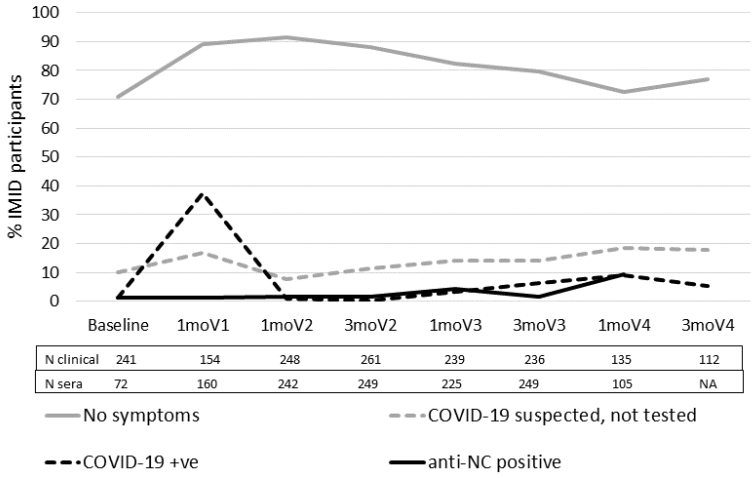
Supplementary Tables and Figures



Supplementary Figure 7 COVID-19 infection

Grey line = no COVID-19 symptoms reported; Dashed grey line = COVID-19 suspected but not tested; dashed black line = COVID-19 infection confirmed by community based testing (rapid detection or polymerase chain reaction test); solid black line = proportion anti-nucleocapsid seropositive.

V1 vaccine one; V2 vaccine two; V3 vaccine 3; V4 vaccine 4; mo = month(s); N = number

STROBE Statement—checklist of items that should be included in reports of observational studies

	Item No	Recommendation	Page No
Title and abstract	1	(*a*) Indicate the study's design with a commonly used term in the title or the abstract	1
		(*b*) Provide in the abstract an informative and balanced summary of what was done and what was found	4
Introduction			
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported	7
Objectives	3	State specific objectives, including any prespecified hypotheses	7-8
Methods			
Study design	4	Present key elements of study design early in the paper	7-8
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection	7-8
Participants	6	(*a*) *Cohort study*—Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up *Case-control study*—Give the eligibility criteria, and the sources and methods of case ascertainment and control selection. Give the rationale for the choice of cases and controls *Cross-sectional study*—Give the eligibility criteria, and the sources and methods of selection of participants	7-8
		(*b*) *Cohort study*—For matched studies, give matching criteria and number of exposed and unexposed *Case-control study*—For matched studies, give matching criteria and the number of controls per case	
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable	8-9
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group	*7-9*
Bias	9	Describe any efforts to address potential sources of bias	7-9
Study size	10	Explain how the study size was arrived at	
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why	7-9
Statistical methods	12	(*a*) Describe all statistical methods, including those used to control for confounding	9
		(*b*) Describe any methods used to examine subgroups and interactions	9
		(*c*) Explain how missing data were addressed	8
		(*d*) *Cohort study*—If applicable, explain how loss to follow-up was addressed *Case-control study*—If applicable, explain how matching of cases and controls was addressed *Cross-sectional study*—If applicable, describe analytical methods taking account of sampling strategy	NA
		(*e*) Describe any sensitivity analyses	

Continued on next page

Results			
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed	10 and S Fig 2
		(b) Give reasons for non-participation at each stage	S Fig 2
		(c) Consider use of a flow diagram	S Fig2
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders	10 Table1
		(b) Indicate number of participants with missing data for each variable of interest	Table 1
		(c) *Cohort study*—Summarise follow-up time (eg, average and total amount)	
Outcome data	15*	*Cohort study*—Report numbers of outcome events or summary measures over time	*Table 1*
		Case-control study—Report numbers in each exposure category, or summary measures of exposure	
		Cross-sectional study—Report numbers of outcome events or summary measures	
Main results	16	(*a*) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included	10-12 Figure 1,2,4 Table 3
		(*b*) Report category boundaries when continuous variables were categorized	
		(*c*) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period	
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses	10-12
Discussion			
Key results	18	Summarise key results with reference to study objectives	12
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias	13-14
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence	13-14
Generalisability	21	Discuss the generalisability (external validity) of the study results	14
Other information			
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based	15

*Give information separately for cases and controls in case-control studies and, if applicable, for exposed and unexposed groups in cohort and cross-sectional studies.

Note: An Explanation and Elaboration article discusses each checklist item and gives methodological background and published examples of transparent reporting. The STROBE checklist is best used in conjunction with this article (freely available on the Web sites of PLoS Medicine at http://www.plosmedicine.org/, Annals of Internal Medicine at http://www.annals.org/, and Epidemiology at http://www.epidem.com/). Information on the STROBE Initiative is available at www.strobe-statement.org.

BMJ Open

Immunogenicity and safety of mixed COVID-19 vaccine regimens in patients with immune mediated inflammatory diseases: a single-centre prospective cohort study

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Immunogenicity and safety of mixed COVID-19 vaccine regimens in patients with immune mediated inflammatory diseases: a single-centre prospective cohort study

CA Hitchon [1], C Mesa [2], CN Bernstein [1], RA Marrie [1,3], C Card [2,4], S O'Brien [5], J Kim [2]

1 Department of Internal Medicine, Max Rady College of Medicine, Rady Faculty of Health Sciences, University of Manitoba, Winnipeg, Canada

2 National Microbiology Laboratory Public Health Agency of Canada Winnipeg, Winnipeg, Canada

3 Department of Community Health Sciences, Max Rady College of Medicine, Rady Faculty of Health Sciences, University of Manitoba, Winnipeg, Canada

4 Department of Medical Microbiology and Infectious Diseases, Rady Faculty of Health Sciences, University of Manitoba, Winnipeg, Canada

5 Canadian Blood Services, Ottawa, Canada

Authors:

Carol A Hitchon, MD MSc, Department of Internal Medicine, Max Rady College of Medicine, Rady Faculty of Health Sciences, University of Manitoba, Winnipeg, Canada

Christine Mesa, MSc, National Microbiology Laboratory Public Health Agency of Canada Winnipeg, Winnipeg, Canada

Charles N Bernstein, MD, Department of Internal Medicine, Max Rady College of Medicine, Rady Faculty of Health Sciences, University of Manitoba, Winnipeg, Canada

Ruth Ann Marrie, MD PhD, Department of Internal Medicine, and Department of Community Health Sciences, Max Rady College of Medicine, Rady Faculty of Health Sciences, University of Manitoba, Winnipeg, Canada

Sheila O'Brien, PhD, Canadian Blood Services, Ottawa, Canada

Catherine Card, PhD, National Microbiology Laboratory, Public Health Agency of Canada Winnipeg, CAN and Department of Medical Microbiology and Infectious Diseases, Rady Faculty of Health Sciences, University of Manitoba, Winnipeg, Canada

John Kim, PhD, National Microbiology Laboratory Public Health Agency of Canada Winnipeg, Winnipeg, Canada

Correspondence to:

CA Hitchon

RR149 800 Sherbrook Street, Winnipeg, Manitoba, Canada

email: carol.hitchon@umanitoba.ca

Word count 3300 words

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Abstract

Objective: Among persons with immune-mediated inflammatory diseases (IMIDs) who received SARS-CoV-2 vaccines, we compared post-vaccine antibody responses and IMID disease activity/states.

Design: Single centre prospective cohort study.

Setting: Specialty ambulatory clinics in central Canada.

Participants: People with inflammatory arthritis (n=78; 77% rheumatoid arthritis), systemic autoimmune rheumatic diseases (n=84; 57% lupus), inflammatory bowel disease (n=93; 43% Crohn's), and multiple sclerosis (n=72; 71% relapsing-remitting) (female 79.4%, white 84.7%, mean (SD) age 56.0(14.3 years) received COVID-19 vaccinations between 03/2021-09/2022.

Primary outcome: Post-vaccination anti-spike, -receptor binding domain (RBD) and -nucleocapsid (NC) IgG antibodies tested by multiplex immunoassays compared across vaccine regimens and to responses in 370 age-sex matched vaccinated controls.

Secondary outcomes: COVID-19 infection and self-reported IMID disease activity/state.

Results: Most 216/327 (66.1%) received homologous mRNA (BNT162b2 or mRNA1273) vaccines, 2.4% received homologous ChAdOx1, and 30.6% received heterologous vaccines (23.9% ChAdOx1/mRNA, 6.4% heterologous mRNA) for their first two vaccines (V1, V2). Seroconversion rates were: post-V1 anti-spike 91/175 (52.0%), anti-RBD 103/175 (58.9%); post-V2 anti-spike 214/234 (91.5%), anti-RBD 211/234 (90.2%), and were lower than controls (post-V2 anti-Spike 360/370 [98.1%] p<0.0001). Antibody titers decreased 3 months post-V2 but increased 1 month post-V3 and 1 month post-V4 [BAU/ml median (IQR) 1 month post-V2, 3 months post-V2, 1 month post-V3, 1 month post V4 anti-S1 1835 (2448), 629.1 (883.4), 4757.5 (7033.1), 4356.0 (9393.4); anti-RBD 1686.8 (2199.44), 555.8 (809.3), 4280.3 (6380.6), 4792.2 (11673.78)]. If primed with a vector vaccine, a mRNA vaccine increased antibody titers to those comparable to homologous mRNA vaccines. Anti-RBD and anti-Spike titers were higher in anti-NC seropositive (N=31; 25 participants) versus seronegative samples [BAU/ml median (IQR) anti-RBD 11755.3 (20373.1) vs 1248.0 (53278.7); anti-Spike 11254.4 (15352.6) vs 1313.1 (3106.6); both p<0.001]. IMID disease activity/state and rates of self-reported moderate or severe IMID flare were similar across vaccinations.

Conclusion: Heterologous COVID-19 vaccination improves seroconversion rates following a vector vaccine and does not lead to IMID disease flare. IMIDs benefit from at least three vaccines.

Strengths and limitations of this study

- Longitudinal cohort study with systematic collection of data on COVID-19 infection, IMID disease activity and with paired biosamples for anti-SARSCoV2 IgG assays following each vaccine for up to four vaccines.
- Cross disease comparisons of four IMIDs from different medical specialties that are treated with immunocompromising medications.
- Validated measures of self-report IMID disease activity/state used to assess vaccine safety and risk of post-vaccine IMID disease flare.
- Relatively small sample size for each IMID and predominantly female population (as expected for these IMIDs) limits analysis of sex and gender effects.

Introduction

COVID-19 vaccines have reduced rates of serious severe acute respiratory syndrome coronavirus (SARS-CoV-2) infection and mortality in the general population.[1-4] However, information on optimal vaccine strategies for immunocompromised individuals who are at increased risk of serious COVID-19 infection is limited.

Immune-mediated inflammatory diseases (IMIDs) such as autoimmune inflammatory arthritis (IA), systemic autoimmune rheumatic disease (SARD), inflammatory bowel disease (IBD), and multiple sclerosis (MS) affect 5% of the general population and share an autoimmune phenotype that affects multiple organ systems and treatment with immune therapies[5]. Due to immune-mediated disease and treatment, some people with IMIDs are at increased risk of vaccine preventable disease including serious COVID-19 infection[6-10]. While COVID-19 vaccines are effective in the general population, immune dysregulation from disease or treatment may impair vaccine responses in people with IMIDs. Due to evolving regional vaccination strategies in our region ([11] and Supplemental Figure 1), a high proportion of individuals with immunocompromised conditions received heterologous vaccine courses. Although available data on immunogenicity of these mixed vaccine courses in the general population are reassuring,[12-14] data are limited regarding the safety and immunogenicity of this strategy for immunocompromised patients as are data comparing these outcomes across diseases.

We established a cohort of patients diagnosed with any of IA, SARDs, IBD or MS to determine the safety (IMID flare) and humoral immunogenicity following COVID-19 vaccination and to assess the impact of mixing COVID-19 vaccine types. Herein, we report the clinical safety results and seroconversion results obtained after four vaccinations and compare seroconversion rates across initial vaccine combinations.

Methods

Study design

This single centre prospective cohort from Manitoba, Canada, was established in March 2021 and enrolled people diagnosed with any of IA, SARDs, IBD, or MS. Data was collected until October 30th 2022. Vaccines were administered in accordance with provincial public health recommendations. Study participants were followed 1 and 3 months after each vaccine for up to four vaccines.

Study objectives

The primary study objective was to compare post-vaccination anti-spike, -receptor binding domain (RBD) and -nucleocapsid (NC) IgG antibody seroconversion and titres across vaccine regimens. Secondary study objectives were to determine the kinetics of seropositivity and titers across vaccine doses, to compare immunogenicity across IMIDs, to determine the effect of vaccination on COVID-19 infection (efficacy), and to determine post vaccine IMID disease activity/state and self-reported IMID flare (safety).

Recruitment

We approached potential participants attending ambulatory clinics using multiple methods with an aim to enrol 400 individuals with a diagnosed IMID. Our sample size was limited by recruitment.

Clinical data

Demographic data, including birth date, sex, self-reported ethnic group according to Canadian Institute for Health Information guidelines[15], highest education level achieved, self-reported comorbidity and health behaviors such as smoking history were collected at baseline. Clinical data including IMID treatment, IMID-specific disease activity/state measures, participant reported interval COVID-19 infections with type of confirmatory test, and biosamples were collected at each visit. IMID treatment was subcategorized as anti-inflammatories and immunomodulators such as 5-ASA, sulfasalazine, hydroxychloroquine, glatiramer, and interferon therapies; traditional immunosuppressants such as methotrexate, leflunomide, azathioprine, and mycophenolate; biologic or advanced therapies such as anti-tumor necrosis factor agents, B cell depleting agents, vedolizumab, fingolimod, anti-cytokine therapies, other

biologics and Janus kinase inhibitors; and corticosteroids (Supplementary Table 1). Self-reported IMID disease activity/status was assessed with disease relevant validated measures (IA: Routine Assessment of Inflammatory Disease version 3 RAPID-3, Rheumatoid Arthritis Flare Core domain indices; SARDs: Systemic Lupus Activity Questionnaire SLAQ; IBD: Inflammatory Bowel Symptoms Severity Index-Short Form (IBDSI-SF), Manitoba IBD flare question; MS: self report disease activity, Expanded Disability Status Scale [EDSS])[16-20]. These disease-related questionnaires were fully completed for IBD (248/255 97% visits), IA (208/215; 97% visits), SARDs (239/260; 92% visits), and MS (291/327; 89% visits). Data were not imputed. Individuals could attend a study visit in person or submit their data by mailed paper forms or direct entry into a REDCap electronic database hosted at the University of Manitoba[21]. Participants had the option to participate in the safety study component only.

Biosamples

Participants not attending the clinic were provided kits to collect blood by finger poke on dried blood spot (DBS) cards for postal submission[22]. Participants attending the clinic had blood collected by venipuncture and DBS were prepared before processing blood for serum. Aliquoted serum and DBS cards were stored at -20C for batch testing.

Regional population control biosample data were obtained from 370 vaccinated Canadian Blood Services blood donors who were age and sex-matched to the clinical cohort. Data on vaccine type administered was not available.

Serology

A bead-based multiplexed immunoassay was used to detect IgG anti-Spike (S1), Receptor Binding Domain (RBD) (reflecting response to vaccine), and Nucleocapsid (NC) antibodies (reflecting response to infection) (Bio-Rad Bioplex 2200 SARS-CoV-2 IgG assay). This assay was chosen based on an evaluation of several multiplex platforms using a large panel of well-pedigreed plasma-DBS specimens[22]. The quantitative measurement of the antibody response used the WHO International standard for anti-SARS-CoV-2 antibody detection. Cut-offs for seropositivity for DBS samples were established using ROC curve analysis. Cut-offs for plasma from the company were used. Sera from IA (n=19) and lupus (n=31) patients collected before

the pandemic were tested to evaluate the potential for false positive SARS-CoV-2 antibody tests in IMIDs with known autoantibodies. Concordance of antibody assays in paired DBS and serum samples was assessed using the Kendalls tau b test. The controls were tested using the Roche Elecsys® anti-SARS-CoV-2 spike protein semiquantitative immunoassay (Roche Diagnostics International Ltd, Rotkreuz, Switzerland) which measures total antibodies (including IgA, IgM and IgG) to the SARS-CoV2- spike protein (anti-S) and the Roche Elecsys® anti-SARS-CoV-2 qualitative immunoassay (Roche Diagnostics LTD Rotkreuz, Switzerland) which measures total antibodies (including IgA, IgM and IgG) to SARS-CoV-2 recombinant protein, nucleocapsid antigen (anti-NC).

Analysis

Demographic information of participants is reported using descriptive statistics including mean (standard deviation-sd), median (range or interquartile range-IQR), and counts (%). Non-parametric tests (Mann-Whitney U or Kruskal Wallis tests with Bonferroni adjustment [Badj] for multiple comparisons) were used to compare antibody levels across groups and visits. Wilcoxon signed rank tests were used to compare antibody levels across visits within individuals. Binary logistic regression was used to evaluate predictors of seroconversion one month following vaccine 2 (V2). Variables included sex, age greater than 65 years, IMID diagnosis, and treatment category (none versus biologics and advanced therapies versus immunosuppressants versus other agents), and vaccine mix (vector-mRNA versus mRNA-mRNA). Statistical analysis was conducted using IBM SPSS Statistics version 27 for Windows (IBM Corporation, Armonk, N.Y., USA).

Ethics and consent

All subjects provided informed consent. The study was approved by the University of Manitoba Health Research Ethics Board (HS24647-H2021-005), Manitoba Shared Health (SH2021:009), and the Health Canada and Public Health Agency of Canada Research Ethics Board (REB 2021-018P).

Patient and public involvement

Patients or the public were not involved in the study design or in the conduct or reporting of the research. Participants were informed of their serologic results.

Results

Between March 12th 2021 and July 30 2022, we recruited 339 participants (Supplementary Figure 2). Vaccination and disease activity data for the time points reported herein were available for 327 participants (78 IA: 77% RA; 84 SARDS: 57% % lupus; 93 IBD: 43 % Crohn's and 72 MS: 71% Relapsing remitting) (Table 1). Most were female (79.4%) and white (84.7%) with a mean (sd) age of 56.0 (14.3) years. Nearly one-third (30.6%) of IMID participants were taking biologics or advanced therapies.

Table 1. Demographics, vaccines administered, self reported IMID flare, and COVID-19 history of participants

	IA N=78	SARDs N=84	IBD N=93	MS N=72	All IMIDs N=327
Age, years (mean SD)	61.9(11.8)	55.7(13.4)	53,7(16.4)	53.1(13.5)	56.0 (14.3)
Female n (%)	66 (84.6)	75 (89.3)	59(63.4)	59 (81.9)	259 (79.4)
White n (%)	66(84.6)	64 (76.2)	79(84.9)	68 (94.4)	277 (84.7)
Education Years of school (median/IQR)	15.0(4.5)	16.0(4.3)	16.0(4.0)	16.0(5.0)	16.0(4.0)
Comorbidity (n %)[2]					
Cardiovascular	35 (44.9)	31 (36.9)	26 (28.0)	22 (30.5)	115(35.1)
Pulmonary	8 (10.2)	24 (28.6)	15 (16.1)	8 (11.1)	55(16.8)
Diabetes	13 (19.7)	3 (3.7)	3 (3.4)	5 (6.9)	24(7.3)
Other endocrine	25 (32.1)	24 (28.6)	20 (21.5)	20 (27.8)	90(27.4)
Renal disease	2 (3.0)	9 (11.0)	4 (4.3)	0 (0.0)	15(4.6)
Cancer	15 (19.2)	10 (11.9)	12 (12.9)	56.9)	43 (13.10
Mental Health	17 (21.8)	27 (32.9)	27 (29.0)	22 (30.6)	94(28.7)
Total (median range)	2.5(0-10)	3(0-10)	2(0-10)	2(0-6)	2(0-10)
BMI (mean SD)	27 (6.6)	28.0 (7.8)	27.2 (6.1)	28.0 (6.2)	27.7 (6.7)
IMID Treatment level [1] n (%)					
Immunomodulators	4(5.1)	25 (30.5)	21 (22.6)	28(38.9)	78 (23.9)
Immunosuppressants	27 (34.6)	42 (51.2)	10 (10.8)	0 (0.0)	79 (24.2)
Biologics/JAKi	37(47.4)	7 (8.5)	41 (44.1)	14 (19.4)	99 (30.3)
- anti-TNF (n)	22	2	28	0	52
- anti-B cell (n)	5	6	0	10	21
- other or JAKi (n)	9	0	13	4	26
None	11 (14.1)	9 (10.7)	21(22.6)	30 (41.7)	71 (21.7)
Vaccine type V1 n (%)[3]					
CHAdOx1	15 (19.5)	28 (33.3)	17 (18.3)	26 (36.1)	86 (26.4)
BNT	60 (77.9)	56 (66.7)	66 (71.0)	39 (54.2)	221 (67.8)
mRNA1273	2 (2.6)	6 (7.1)	10 (10.8)	7 (9.7)	25 (7.7)
Vaccine type V2 n (%)[3]					
CHAdOx1	2 (2.6)	3 (3.6)	3 (3.2)	1 (1.4)	9 (2.7)
BNT	65 (84.4)	58 (68.7)	60 (65.2)	54 (75.0)	236 (72.8)
mRNA1273	16 (13.0)	23 (27.7)	29 (31.5)	17 (23.6)	79(24.4)
Vaccine type V3 n/N (%)[3]					
CHAdOx1	0	0	1 (1.3)	1 (1.6)	2 (0.7)
BNT	54 (71)	58 (78.4)	49 (66,2)	39 (61.9)	200 (71.2)
mRNA1273	16 (21.1)	16 (21.6)	24 (32.4)	23 (36.5)	79 (28.1)
Other	0	0 (0)	1 (1.4)	1 (1.6)	2 (0.7)
Vaccine type V4 n/N (%)[3]					
CHAdOx1	0 / (0)	1 (2.7)	0 (0)	0 (0)	1 (0.8)
BNT	25 (69.4)	28 (75.7)	25 (64.1)	12 (70.6)	90 (69.8)

mRNA1273	10 (27.8)	8 (21.6)	14 (35.9)	2 (11.8))	34 (26.4)
Other	-	-	-	3 (17.6)	3 (2.3)
Vaccines for V1 and V2 n (%)					
CHAdOx1-- BNT	9 (11.5)	10 (11.9)	4 (4.3)	17 (23.6)	40(12.2)
CHAdOx1 - mRNA1287	4 (5.1)	15(17.9)	11 (11.8)	8 (11.1)	38 (11.6)
BNT – mRNA1273 or mRNA1273-BNT	4 (5.1)	5 (6.0)	10 (10.8)	2 (2.8)	21 (6.4)
BNT-CHAdOx1	0(0)	0(0)	1 (1.1)	0(0)	1 (0.3)
CHAdOx1-CHAdOx1	2 (2.6)	3 (3.6)	2 (2.2)	1 (1.4)	8 (2.4)
BNT- BNT	56(71.8)	46 (54.8)	55 (59.1)	37 (51.4)	194 (59.3)
mRNA1273 -mRNA1273	2 (2.6)	4 (4.8)	9 (9.7)	7 (9.7)	22 (6.7)
Vaccine interval Days between V1 and V2 median (range)	66 (21-97)	62 (20-188)	57 (20-98)	59 (26-97)	60 (20-188)
Self-report IMID flare [4,5]					
1 month post V1 n/N (%)	12/44 (27.3)	10/44 (22.7)	0/44 (0)	1/41 (2.4)	23/173 (13.3)
1 month post V2 n/N (%)	9/56 (16.1)	9/58 (8.6)	0/67 (0)	2/69 (2.9)	20/250 (8.0)
1 month post V3 n/N (%)	11/57 (19.3)	19/63 (30.2)	2/62 (3.3)	1/61 (1.6)	33/243 (13.6)
1 month post V4 n/N (%)	6/38 (15.8)	12/57 (21.1)	4/38 (8.3)	1/24 (4.2)	23/157 (14.6)
1 month post any V n/N(%)	38/195 (19.5)	50/202 (24.8)	6/211 (2.8)	5/129 (3.9)	99/803 (12.2)
COVID-19 illness -ever[5]					
None	43 (55.1)	38 (46.3)	46 (50.0)	31 (43.1)	158 (48.8)
Suspected but not tested	27 (34.6)	29 (35.4)	22 (23.9)	24 (33.3)	112(34.6)
COVID-19 PCR +ve	8 (10.3)	13 (15.9)	14 (15.2)	17 (23.6)	52 (16.0)
COVID-19 +ve hospitalized	0 (0.0)	2 (2.4)	0 (0.0)	0 (0.0)	2 (0.6)

IA=inflammatory arthritis (Rheumatoid Arthritis N=60, Psoriatic arthritis N=8, other Spondyloarthropathy N=3, other IA N=7); SARDs = systemic autoimmune rheumatic disease (systemic lupus erythematosus N=48, myopathy N=5, vasculitis N=7 other or unknown SARDs N=24) ; IBD = inflammatory bowel disease (crohn's disease N=43); MS= multiple sclerosis (relapsing-remitting N=51, secondary progressive N=11, primary progressive N=4, unknown N=2) ; IMID = immune mediated inflammatory disease; FT=full time; PT= part time; BMI=body mass index; JAKi= janus kinase inhibitor; TNF= tumor necrosis factor inhibitor; BNT= BNT162b2 vaccine V1= first vaccine; V2 = second vaccine, V3 = third vaccine; V4 = fourth vaccine; NA=not available, PCR= polymerase chain reaction; IQR = interquartile range.

[1] IMID treatment based on most aggressive combination if on multiple agents. 1 RA subject on only prednisone monotherapy. Medication data missing for 2 SARDs subjects. Other therapy N=26 (ustekinumab n=6, vedolizumab n=7, abatacept n=1, tofacitinib n=4, tocilizumab n=3, fingolimod n=2, alemtuzumab n=1, natalizumab n=1, upadacitinib n=1)].

[2] cardiovascular disease includes ischemic heart disease, congestive heart failure, valvular heart disease peripheral vascular disease stroke or transient ischemic attack or hypertension; respiratory disease

includes asthma or chronic obstructive pulmonary disease, other endocrine includes thyroid disease, hypercholesterolemia.

[3] Vaccine data available for V1 N=326 (77 IA, 84 SARDS, 72 MS 93 IBD); V2 N=324 (77IA, 83 SARDS, 72 MS 92 IBD); V3 N=281 (76 IA, 74 SARDS, 63 MS, 74 IBD) V4 N=128 (35 IA, 37 SARDs 39 IBD 17 MS).

[4] Flare assessed by the following questions: IA: "Are you having a flare? SARDs: "In the past 3 months have you had a disease flare (A flare is when your disease gets worse)? Yes moderate or severe flare; IBD: In the past 6 months my disease has been: Constantly active, giving me symptoms every day, or often active, giving me symptoms most days or sometimes active, giving me symptoms on some days (for instance 1-2 days/week); MS Do you feel there has been a change in your MS since your last visit? "My multiple sclerosis is much worse and in a flare".

Flare rate across IMIDs 1 month after any vaccine Chi^2 72.9 $p<0.001$; Flare rate post V1 across IMIDs Chi^2 21.8 $p<0.001$. Flare rate post V2 across IMIDs Chi^2 17.7 $p<0.001$; Flare rate post V3 across IMIDs Chi^2 29.4 $p<0.001$; Flare rate post V4 across IMIDs Chi^2 10.3 $p=0.02$. IMID Flare rate across vaccination visits Chi^2 7.4 $p=0.06$.

[5] Self-reported over course of study; COVID-19 illness data missing for 2 SARDS and 1 IBD participant.

Samples of adequate quality were obtained following the first vaccine at 1 month (n=175) and 3 months (n=44), following the second vaccine at 1 month (n=234) and 3 months (n=246), following the third vaccine at 1 month (n=215) and following the fourth vaccine at 1 month (n=85). For 31 participants, DBS samples were of inadequate quality and a serum sample from the same day was substituted. In paired DBS and serum tests from 208 subjects, seroconversion and titers were highly concordant (Kendall's tau b correlation coefficient anti-RBD BAU/ml=0.93; anti-S1 BAU/ml= 0.92) (Supplementary Figure 3). Pre-pandemic sera were seronegative for anti-SarsCo-V2 antibodies.

Following the first vaccine, 60% of IMID participants seroconverted (Table 2). Following the second vaccine, seroconversion rates increased to 91% (p=0.1 across IMIDs for both anti-S1 and anti-RBD). The change in anti-S1 seropositivity between first and second vaccines was significant (anti-S1 all IMIDs Chi^2 82.2 p<0.0001; IA Chi^2 40.5 p<0.0001; SARDs Chi^2 18.5 p<0.0001; IBD $Chi^2$16.9 p<0.0001; MS Chi^2 4.1 p=0.04 and anti-RBD all IMIDs Chi^2 55.1 p<0.0001; IA Chi^2 31.6 p<0.0001; SARDS Chi^2 18.8 p<0.0001; IBD Chi^2 8.4 p<0.01; MS Chi^2 5.1 p=0.02). Of the 20 participants who were seronegative after the second vaccine and had data following the third vaccine, 8/15 (53.3) seroconverted after the third vaccine. Seroconversion rates for both anti-S1 and anti-RBD after the third and fourth vaccines were greater than 95%.

Table 2. Seroconversion rates one month following each COVID-19 vaccine

Seroconversion	IA	SARDs	IBD	MS	All IMIDs
Post V1					
Anti- S1	14/47 (29.8%)	21/47 (44.7%)	33/48(68.8%)	23/33 (69.7%)	91/175 (52.0%)
Anti-RBD	18/47 (38.2%)	22/47 (46.8%)	40/48 (83.3%)	23/33 (69.7%)	103/175 (58.9%)
Anti-NC	1/47 (2.1%)	0/48 (0%)	0/47 (0%)	1/33 (3%)	2/175 (1.1%)
Post V2 [1,]					
Anti-S1 [2]	50/55 (90.9%)	46/54 (85.1%)	62/64 (96.9%)	53/61 (86.9%)	214/234 (91.5%)
Anti-RBD [3]	50/55 (90.9%)	47/54 (87.0%)	63/64 (98.4%)	54/61 (88.5%)	211/234 (90.2%)
Anti-NC	1/55 (1.8%)	0/57 (0%)	1/64 (1.6%)	1/61 (1.6%)	3/234 (1.3%)
Post V3					
Anti-S1	51/52 (98.1%)	57/62 (91.9%)	56/56 (100.0%)	41/45 (91.1%)	205/215 (95.3%)
Anti-RBD	51/52 (98.1%)	56/62 (90.3%)	56/56 (100.0%)	41/45 (91.1%)	204/215 (94.9%)
Anti-NC	2/52 (3.8%)	3/62 (4.8%)	2/56 (3.6%)	2/45 (4.4%)	9/215 (4.2%)
Post V4					
Anti-S1	31/32 (96.9%)	21/23 (91.3%)	27/27 (100%)	3/3 (100%)	82/85 (96.5%)
Anti-RBD	31/32 (96.9%)	21/23 (91.3%)	27/27 (100%)	3/3 (100%)	82/85 (96.5%)
Anti-NC	3/32 (9.4%)	4/23 (17.4%)	3/27 (11.1%)	0/3 (0.0%)	10/85 (11.8%)

IA=inflammatory arthritis; SARDs = systemic autoimmune rheumatic disease; IBD = inflammatory bowel disease; MS= multiple sclerosis; IMID = immune mediated inflammatory disease; V1 = first vaccine; V2 = second vaccine; V3= third vaccine; V4=fourth vaccine.

[1] seroconversion V1 to v2 across IMIDs anti-S1 Chi^2 82.2 p<0.0001; anti-RBD Chi^2 55.1 p<0.0001.

[2] change in anti-S1 seropositivity V1 to V2 IA Chi^2 40.5 p<0.0001, SARDs Chi^2 18.5 p<0.0001 IBD $Chi^2$16.9 p<0.0001; MS Chi^2 4.1 p=0.04.

[3] change in anti-RBD seropositivity V1 to V2 IA Chi^2 31.6 p<0.0001, SARDS Chi^2 18.8 p<0.0001, IBD Chi^2 8.4 p<0.01, MS Chi^2 5.1 p=0.02.

Post-V2 anti-S1 seroconversion rates for IMIDs were lower compared to those of age and sex-matched controls (anti-S1 seropositive controls 363/370 (98.1%) vs IMIDs X^2 14.5 p<0.0001) but similar for anti-NC (anti-NC seropositive controls 13/370 (3.5%) vs IMIDs Chisq-2.9 p=0.1). Matched population-based estimates were not available for subsequent vaccinations.

Anti-RBD and anti-S1 IgG titres of those who seroconverted increased between the 1 month post-V1 and 1 month post-V2 time points for combined IMIDs and for each IMID individually (Figure 1) (p values all p<0.0001). Anti-RBD and anti-S1 titers declined by 3 months post-second

vaccine (p values for combined IMIDs p<0.0001; SARDs p<0.001; IBD p<0.001, IA p<0.001 and MS p<0.001) but increased 1 month post-third vaccine (p value all p<0.0001). Titers of anti-RBD and anti-Spike were similar between 1 month post-third vaccine and 1 month post-vaccine fourth vaccine. Within individuals, paired analysis of titers across visits yielded similar findings.

Over the study, 99 (30.6%) IMID participants received heterologous/mixed vaccines for their first two vaccines (Table 1). For those with one month post-V2 serology data, individuals receiving homologous vector vaccines had the lowest seroconversion rates and titers for both anti-RBD and anti-S1 responses. Individuals receiving either mRNA vaccine following a vector vaccine had comparable anti-RBD and anti-Spike titers as those receiving two mRNA vaccines (Figure 2). Anti-RBD and anti-S1 seropositivity and titers one month following the third vaccine were similar among individuals receiving different combinations of vaccines for their first two vaccines (Supplemental Table 2, Supplemental Figure 4).

Participants over age 65 years, diagnosed with MS, or taking biologics, were less likely to seroconvert by the second vaccine in multivariable models (Table 3). Results were similar if seroconversion was defined as seropositivity to anti-RBD and/or anti-Spike. Vaccine mix (vector-mRNA versus mRNA-mRNA) did not impact seroconversion when included in the regression models. Most of the 20 individuals who did not seroconvert after V2 were taking immunosuppressives (mycophenolate n=5; methotrexate n= 2, azathioprine n=1) or biologics (B cell targeting currently or previously n=8, anti-TNF n=2, other n= 2) (Supplementary Table 3).

Table 3. Clinical variables associated with seroconversion one month following second vaccine

	Post second vaccine OR (95% CI)	P value
RBD seroconversion		
Sex		
Male	Ref	1.0
Female	1.0 (0.21-4.71)	
Age		
>65 years	Ref	0.002
≤ 65 years	7.4 (2.04-27.06)	
IMID		
IBD	Ref	<0.001
IA	0.2 (0.02-2.65)	
SARDs	0.05 (0.003-0.79)	
MS	0.009 (0.001-0.13)	
Immune therapy		
None	Ref	0.004
Other immunomodulator	NA	
Immunosuppressant	0.03 (0.002-0.33)	
Biologic	0.02 (0.002-0.16)	
S1 seroconversion		
Sex		
Male	Ref	0.85
Female	1.1 (0.29-4.52)	
Age		
>65 years	Ref	0.02
≤ 65 years	3.9 (1.29-11.62)	
IMID		
IBD	Ref	<0.001
RA	0.5 (0.07-3.02)	
SARDs	0.1 (0.01-0.91)	
MS	0.03 (0.003-0.20)	
Immune therapy		
None	Ref	0.01
Other immunomodulator	NA	
Immunosuppressant	0.06 (0.006-0.51)	
Biologic	0.04 (0.007-0.27)	

RBD = Receptor binding domain; S1 = Spike protein; Ref = reference category; NA = not able to compute. Biologics = Tumor necrosis factor antagonists, abatacept, tocilizumab, tofacitinib, upadacitinib, ustekinumab, apremilast, rituximab, anakinra, B cell targeted therapies (rituximab, belimumab, ocrelizumab). Immunosuppressants = methotrexate, leflunomide mycophenolate Immunomodulators = hydroxychloroquine, sulfasalazine.

We evaluated the impact of vaccine mix and treatment on waning of anti-SARSCo-V2 titers between 1 and 3 months following the second vaccination. In paired analysis the decline in titers between 1month post V2 and 3 months post V2 for anti-RBD and anti-S1 differed across vaccine mixtures (anti-RBD p=0.026; anti-S1 p=0.02) however this was mainly due to minimal titer changes for individuals receiving homologous vector vaccines who had lower titers overall. We observed greater titer change between those that received vector/mRNA versus mRNA/mRNA combinations for S1, but did not see differences in RBD titer change between those that received vector/mRNA versus mRNA/mRNA combinations. [median (IQR) anti-S1 mixed 1591.6 (3002.7) vs homologous mRNA 1086.3 (1608.8) p=0.021; anti-RBD mixed 1469.9 (2086.5) vs homologous mRNA 1124.5 (1402.4) p=0.051]. For individuals receiving homologous mRNA (BNT/BNT vs mRNA1273/mRNA1273) there was no difference in waning for anti-RBD or anti-S1. [RBD titer change anti-RBD: BNT/BNT 1080.6 (2405) vs mRNA1273/mRNA1273 1434.9 (2465.1) p=0.58; anti-S1 BNT/BNT 1051.9 (1674.1) vs mRNA1273/mRNA1273 1567.5 (2481.9) p=0.39]. There was no difference in waning for individuals receiving homologous mRNA vs mixed vector/mRNA of the same type for anti-RBD or anti-S1. There was no difference in titer change across different biologic categories (anti-TNF versus Bell depletion versus other biologic; anti-RBD p= 0.30; anti-S1 p=0.14) (Supplemental Figure 5).

Twenty-five participants were seropositive for anti-NC antibodies on at least one visit (8 IA, 8 SARDs, 3 MS, 6 IBD) and for 4 of these individuals, seropositivity persisted with declining titers across consecutive visits spanning 3 to 6 months. All but one MS participant were also anti-RBD and anti-S1 seropositive. Anti-RBD and anti-S1 titers were higher in anti-NC positive compared to anti-NC negative samples [median (range; IQR) anti-RBD 11755.3 (20373.1) vs 1248.0(27-78936.2; 53278.7); anti-Spike 11254.4 (77.3-68157.0; 15352.6) vs 1313.1 (37.4-87401.3; 3106.6)] (Supplemental Figure 6). Of the 25 seropositive individuals, 9 were asymptomatic, 10 were taking biologics (5 anti-TNFs, 3 current or past B cell depletion, 4 other biologics/advanced therapies), 7 immunosuppressives (5 methotrexate, 2 azathioprine), 7 immunomodulating agents alone, and 1 MS participant was on no IMID (Supplemental Table 4). Although the rates

of anti-NC positivity increased over the course of the study, anti-NC titers did not vary by vaccine status (heterologous or homologous) nor by date tested (Table 2, Supplemental Figure 7).

Most participants reported no COVID-19 infection symptoms during the study, including 9 individuals who tested seropositive for anti-NC antibodies, whereas 113 (35.7%) of participants reported mild symptoms consistent with COVID-19 infection but did not have community-based confirmatory testing by either polymerase chain testing before Dec 2021 or self-administered testing after Dec 2021. All COVID-19 infections with positive community-based testing were also anti-NC positive. Only two confirmed COVID-19 infections required hospitalization. Both had received 3 vaccines and had moderate levels of anti-NC (69.4 and 22.8 BAU/ml). (Table 1)

Self-reported disease activity/status scores were similar across visits for each IMID (Figure 3). Rates of moderate or severe self-reported IMID flares were similar across vaccines. MS and IBD participants had lower rates of self-reported flare compared to IA and SARDs [n/N (%) with self-reported flare after any vaccine MS 5/129 (3.9), IA 38/195 (19.5), SARDs 50/202 (24.8); IBD 6/211 (2.8) (Chi^2 72.9 $p<0.001$] IA and SARDs tended to have greater rates of self-reported flare (Table 1).

Discussion

This single center cohort study evaluated the safety and immunogenicity of SARS-COV-2 vaccines in IMIDs and confirmed relative safety with no increase in IMID disease activity despite self-reported disease flare rates of 12% following four vaccinations. Fewer than two-thirds seroconverted after the first vaccine. Seroconversion rates differed by vaccine type with higher titres of anti-RBD and anti-Spike responses generated by mRNA vaccines compared to the available vector vaccine. Individuals who received an initial vector vaccine followed by a mRNA vaccine had vaccine induced titers that were comparable to those following a homologous mRNA vaccine course and were higher than those who received homologous vector vaccines. Anti-SARS-CoV-2 antibody titers declined 3 months after the second vaccine but improved after

the third and fourth vaccines. Most individuals who did not produce adequate humoral responses to the vaccine were taking immunosuppressants or biologics.

Our findings are consistent with emerging clinical trial and cohort data from the general population and other immunocompromised groups. Prior studies of rheumatic disease flare post-COVID-19 vaccination have produced mixed results however, most found no major concerns[7]. Data for IBD and MS are also reassuring[23 24]. Several studies have found lower seroconversion rates and anti-SARS-CoV-2 titers in IMIDs compared to the general population[25 26]. While older age plays a role, the primary reason for reduced responses appears due to medication use with the greatest impact from biologics, especially B-cell depleting therapies, fingolimod, anti-tumor necrosis factor agents, and Janus kinus inhibitors, and with DMARDs such as mycophenolate and methotrexate. Hydroxychloroquine does not impair vaccine responses. Proposed strategies to optimize responses for patients on these medications include holding medication around the time of vaccination and delaying vaccination following infusion of B-cell targeted therapies[27 28]. For B cell therapies, humoral immune responses remain suboptimal even after third doses especially for individuals with low pre-vaccination cell counts[29-31]. Reassuringly, T cell responses are induced although possibly impaired[26 31 32].

Heterologous vaccine administration increased as vaccine availability and data on safety and immunogenicity evolved, thereby allowing evaluation of the role of homologous versus heterologous vaccine administration for people with IMIDs. In the general population, clinical trials and cohort studies of mixing vaccine types that compared homologous vector, homologous mRNA and heterologous vector/mRNA vaccine courses observed greater immune responses (humoral and cellular) with mRNA vaccines than vector vaccines and that in individuals receiving a vector vaccine first, a mRNA vaccine improved vaccine responses to levels comparable to those of the homologous mRNA vaccines[12-14 33]. Our observations in IMIDs confirm that second, and at least third vaccination courses are needed to generate acceptable humoral immunogenicity, that mRNA vaccines can overcome limited responses to vector vaccines, and that the type of mRNA administered has minimal impact on waning antibody titers following the second vaccination.

The clinical findings of this report reflect data collected during intermittent public health mandated societal restrictions, before and during the early period when the Omicron variant was circulating in our region, and before bivalent vaccines were available[11]. Over one-third reported mild symptoms or were suspected to have had COVID-19 illness but less than 10% had confirmed COVID-19 infection. Despite complete vaccination, infection and concerning symptoms increased as public health restrictions were relaxed, the prevalence of SARS-CoV-2 virus increased, and new variants of concern emerged. This emphasizes the need for ongoing COVID-19 surveillance to inform personal health practices given the real concerns expressed by many people with IMIDs, even those who are fully vaccinated. Recent studies have described reduced sensitivity of the anti-NC assay following vaccination that is only partially explained by viral load.[34] In this study, all participants with COVID-19 infection confirmed with community based testing were also anti-NC positive and while the number of anti-NC +ve participants increased over the study, we did not see any difference in anti-NC IgG levels with number of vaccines or by calendar month.

We acknowledge limitations of this work. Our sample size was relatively small for each IMID; however, we have collected extensive patient-reported data combined with biologic samples from individuals representing four common IMID groups, allowing cross-disease and cross-specialty comparisons which are not widely reported. We assessed IMID disease activity using validated patient self-reported disease specific indices and flare questions although these questionnaires can be subject to recall bias. Ideally patient reported IMID activity would be supplemented with clinician assessed measures however both patient preferences and COVID-19 pandemic travel restrictions impacted the feasibility of in person clinical assessments for all participants. Self reported of disease activity/state measures correlate with clinical assessment measures[19 35-37]. As expected for these IMIDs, our population was predominantly female thus we lack power to detect sex-based differences in our outcomes and there is uncertainty as to how they would reflect a male predominant cohort. We focused on humoral vaccine induced immunogenicity using antibody seropositivity and titres as surrogates for vaccine induced protection. Antibody binding titers have been shown to correlate with neutralizing and cellular responses which in turn correlate with vaccine efficacy, although the titers needed to achieve

good vaccine efficacy may differ for anti-Spike and anti-RBD[38]. Further work is needed to evaluate the neutralization capacity of vaccine induced antibodies to SARS-COV-2 and emerging variants of concern including Omicron. We did not evaluate cellular immune responses yet these are critical for long term anti-viral protection especially for individuals without robust antibody responses. We were not able to confirm prior reports of the impact of different biologic categories on antibody titers however our study was not powered for this question. Additional studies are needed to evaluate if there are important differences across mRNA vaccines and vaccine intervals for optimal protection against variants of concern to inform recommendations for additional vaccinations in IMIDs. Importantly, it is still unclear what level of humoral or cellular immunogenicity is optimal to protect IMIDs against serious COVID-19 infection although population-based vaccine efficacy data are emerging for some immunocompromised groups[39].

We conclude that most individuals with IMID can safely receive COVID-19 vaccines without risk of disease flare. At least two doses that include a mRNA vaccine, either homologous or mixed vaccine types are needed to generate humoral immunity comparable to the general population. The observed decline in humoral responses support the use of third and subsequent vaccine doses for IMIDs. These data can be used to direct vaccine policies in countries where vaccine rates have been lagging or where supply has been limited.

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Figure legends

Figure 1. Titers of anti- spike and anti-receptor binding domain IgG levels following first, second third and fourth vaccination

A. All IMIDs. B. IA. C. SARDs. D. IBD. E. MS.

Data for seroconverters only. IgG levels natural log transformed. IMIDs=immune mediated inflammatory disease; IA = inflammatory arthritis; SARDs = systemic autoimmune rheumatic disease; IBD = inflammatory bowel disease; MS = multiple sclerosis; S1= spike; RBD= receptor binding domain; V1 = first vaccine; V2 = second vaccine; V3= third vaccine. Unadjusted p values * $p<0.0001$, ** $p\leq0.001$, *** $p\leq0.00$, **** $p<0.05$.

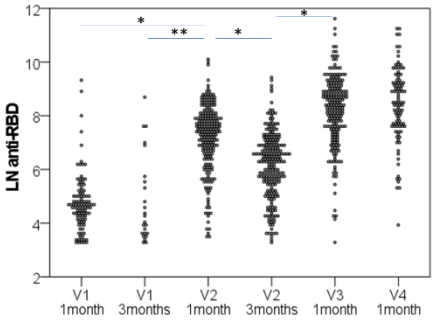
Figure 2. Titers of anti- spike and anti-receptor binding domain IgG levels 1 month following second vaccination by vaccine mixture
A. log anti-RBD. B. log anti-Spike.
Data for seroconverters only. IgG levels natural log transformed. S1= spike; RBD= receptor binding domain BNT= BNT162b2, Unadjusted p values * $p<0.01$.

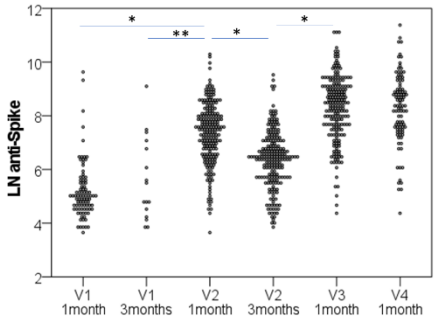
Figure 3. Disease activity before and after each vaccine

A. Inflammatory arthritis. B. Systemic autoimmune rheumatic disease. C. Inflammatory bowel disease. D. Multiple Sclerosis.

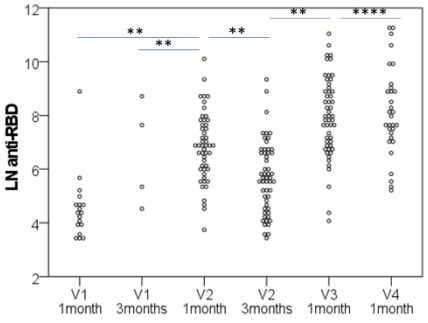
RAPID-3 Routine Assessment of Patient Index Data 3; SLAQ Systemic lupus activity questionnaire IBDSI-SF Inflammatory bowel disease symptom inventory – short form; EDSS Expanded disability status scale. V1 vaccine one; V2 vaccine two; V3 vaccine 3; V4 vaccine 4; mo = month(s).

A


LN anti-RBD
V1 1month
V1 3months
V2 1month
V2 3months
V3 1month
V4 1month


LN anti-Spike
V1 1month
V1 3months
V2 1month
V2 3months
V3 1month
V4 1month

B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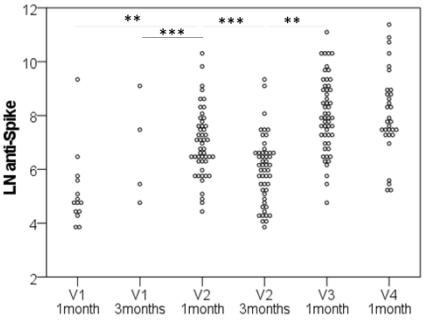
V1 1month
V1 3months
V2 1month
V2 3months
V3 1month
V4 1month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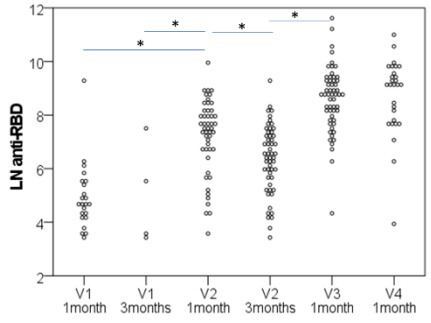
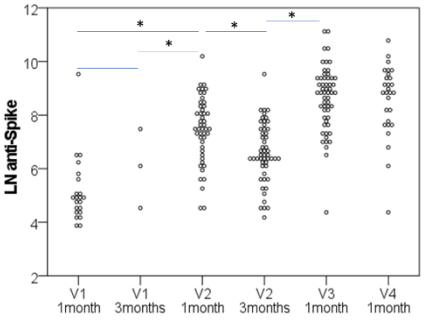
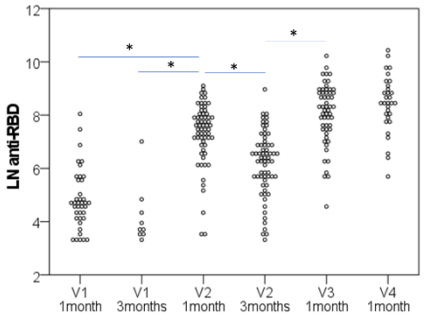
V1 1month
V1 3months
V2 1month
V2 3months
V3 1month
V4 1month

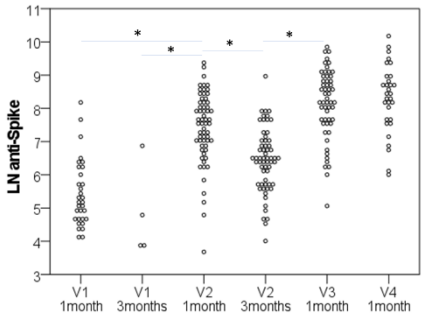
C


LN anti-RBD
V1 1month
V1 3months
V2 1month
V2 3months
V3 1month
V4 1month

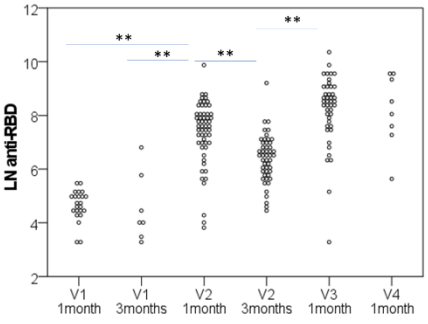

LN anti-Spike
V1 1month
V1 3months
V2 1month
V2 3months
V3 1month
V4 1month

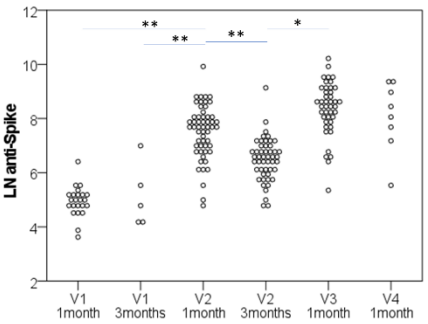
D


LN anti-RBD
V1 1month
V1 3months
V2 1month
V2 3months
V3 1month
V4 1month


LN anti-Spike
V1 1month
V1 3months
V2 1month
V2 3months
V3 1month
V4 1month

E


LN anti-RBD
V1 1month
V1 3months
V2 1month
V2 3months
V3 1month
V4 1month


LN anti-Spike
V1 1month
V1 3months
V2 1month
V2 3months
V3 1month
V4 1month

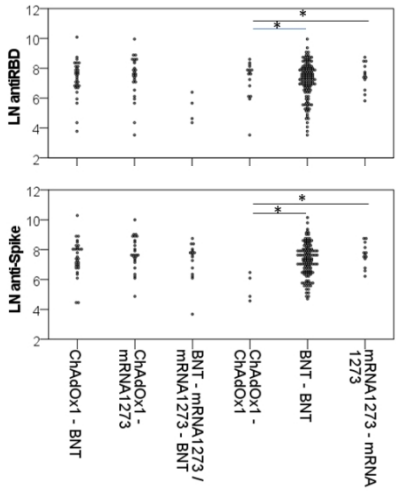


Figure 2 Titers of anti- spike and anti-receptor binding domain IgG levels 1 month following second vaccination by vaccine mixture. A log anti-RBD B log anti-Spike

Data for seroconverters only. IgG levels natural log transformed. S1= spike; RBD= receptor binding domain BNT= BNT162b2, Unadjusted p values * p<0.01

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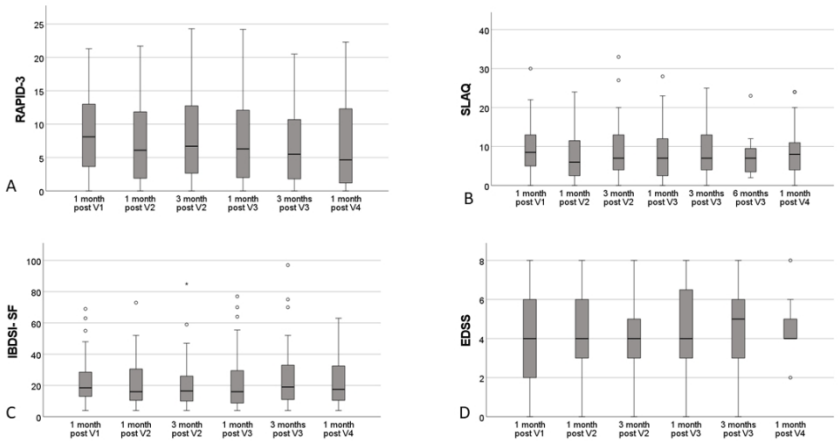

Figure 3 Disease activity before and after each vaccine
A Inflammatory arthritis B Systemic autoimmune rheumatic disease C Inflammatory bowel disease D Multiple Sclerosis
RAPID-3 Routine Assessment of Patient Index Data 3; SLAQ Systemic lupus activity questionnaire IBDSI-SF Inflammatory bowel disease symptom inventory – short form; EDSS Expanded disability status scale. V1 vaccine one; V2 vaccine two; V3 vaccine 3; V4 vaccine 4; mo = month(s).

855x481mm (38 x 38 DPI)

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Supplemental Tables and Figures

Supplemental Table 1 Categorization of IMID treatments

Treatment / Disease	Inflammatory Bowel Disease	Multiple Sclerosis	Rheumatoid Arthritis and SARDs
Corticosteroids (N=37)	Methylprednisolone Prednisolone) Prednisone Budesonide Hydrocortisone	Methylprednisolone Prednisolone Prednisone	Methylprednisolone) Prednisolone Prednisone Triamcinolone Cortisone
Anti-inflammatory or Immunomodulatory therapies (N=77)	5-ASA Sulfasalazine	Glatiramer acetate interferon-beta 1a interferon-beta 1b dimethyl fumarate Teriflunomide Peg interferon-beta	Sulfasalazine sodium aurothiomalate auranofin aurothioglucose Penicillamine Hydroxychloroquine (
Traditional immunosuppressive therapies (N=78)	Azathioprine Methotrexate 6-mercaptopurine Cyclosporine Tacrolimus	Azathioprine Methotrexate) Mitoxantrone Cyclophosphamide	Azathioprine methotrexate Cyclophosphamide Cyclosporine Leflunomide Mycophenolate Tacrolimus
Novel therapies/ Biologics[1] (N=98)	Infliximab adalimumab Golimumab Ustekinumab Vedolizumab Tofacitinib	Natalizumab Fingolimod[2] Alemtuzumab Cladribine Ocrelizumab	Infliximab) Adalimumab Etanercept Anakinra Rituximab[3] Abatacept[3] Tocilizumab Tofacitinib Golimumab Certolizumab Upadacitinib Baricitinib Belimumab Sekukinumab Ustekinumab

[1.] Anti-TNF N= 51, B cell depletion N=21, Past B cell depletion N=9, Other therapy N=27 (ustekinumab n=6, vedolizumab n=6, tofacitinib n=4, tocilizumab n=2, fingolimod n=2, alemtuzumab n=1, natalizumab n=1, updacitinib n=1)

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Supplemental Tables and Figures

Supplemental Table 2 Seroconversion rates based on vaccine mixture between first and second vaccinations

RBD= receptor binding domain; BNT= BNT162b2

Homologous (any) versus heterologous (any) vaccine combination and seroconversion 1 month post V2 anti-Spike Chi^2 7.8 p<0.01; antiRBD Chi^2 6.8 p<0.01; 1 month post V3 anti-Spike Chi^2 0.5 p=NS; anti RBD Chi^2 0.2 p=NS

	1month post V2 Anti-Spike seroconverted n/N (%)	1month post V2 Anti-RBD seroconverted n/N (%)	1month post V3 Anti-Spike seroconverted n/N (%)	1month post V3 Anti-RBD seroconverted n/N (%)
Homologous	137/158 (86.7)	139/158 (88.0)	130/140 (92.9)	132/140 (94.3)
ChAdOX1-ChAdOX1	4/6 (66.7)	4/6 (66.7)	5/6 (83.3)	5/6 (83.3)
BNT-BNT	120/139 (84.3)	122/139 (87.8)	112/121 (92.6)	113/121 (93.4)
mRNA 1273-mRNA1273	13/13 (100)	13/13 (100)	13/13 (100.0)	13/13 (100.0)
Heterologous	69/70 (98.6)	69/70 (98.6)	64/65 (98.5)	64/65 (98.5)
ChAdOX1-BNT	30/31 (96.8)	30/31 (96.8)	27/28 (96.4)	27/28 (96.4)
ChAdOX1-mRNA1273	25/25 (100)	25/25 (100)	24/24 (100.0)	24/24 (100.0)
BNT-mRNA1273	14/14 (100)	14/14 (100)	13/13 (100.0)	13/13 (100.0)

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Supplemental Tables and Figures

Supplemental Table 3 Clinical features of IMIDs not seroconverting after 2 vaccinations

IMID = immune mediated inflammatory disease; IA= inflammatory arthritis; SARDs= Systemic autoimmune rheumatic disease; MS= multiple sclerosis; IBD= inflammatory bowel disease M=male; F=female

IMID	Sex	Age baseline	Medication	vaccine combination
IA	M	82	Prednisone	BNT-BNT
IA	F	77	Methotrexate + rituximab	BNT-BNT
IA	F	76	tocilizumab	BNT-BNT
IA	F	70	methotrexate + anti-TNF	BNT-BNT
IA	F	61	mycophenolate	BNT-BNT
SARDs	F	66	rituximab + prednisone	BNT-BNT
SARDs	F	60	mycophenolate	ChAdOX1-ChAdOX1
SARDs	F	31	rituximab + prednisone	BNT-BNT
SARDs	F	72	mycophenolate	BNT-BNT
SARDs	F	71	mycophenolate	BNT-BNT
SARDs	F	69	mycophenolate + IV Immune globulin	BNT-BNT
SARDs	F	69	azathioprine + past rituximab	BNT-BNT
MS	F	56	ocrelizumab	ChAdOX1-BNT
MS	M	57	ocrelizumab	BNT-BNT
MS	F	74	none	BNT-BNT
MS	F	36	ocrelizumab	BNT-BNT
MS	F	48	fingolimod	BNT-BNT
MS	F	81	none	BNT-BNT
MS	F	34	ocrelizumab	BNT-BNT
IBD	F	31	anti-TNF + prednisone	ChAdOX1-ChAdOX1

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Supplemental Tables and Figures

Supplemental table 4: Clinical characteristics of anti-nucleocapsid seropositive participants

IMID	Age (years)	First visit tested positive	Preceding visit with serology	Anti-RBD titer at preceding visit[1]	Anti-S1 titer at preceding visit[2]	First and second vaccine	Last vaccine prior to infection	IMID treatment
IA	63	1moV4	1moV3	76752	11941	v/mRNA	mRNA1273	JAKi
IA	63	1moV3	3moV1	NA	NA	mRNA/mRNA	mRNA1273	HDQ;MTX
IA	65	3moV4	3moV2	1547	1505	mRNA/mRNA	BNT	HDQ
IA	69	1moV4	1moV3	1365	1526	mRNA/mRNA	mRNA1273	HDQ aTNF
IA	53	1moV3	3moV2	923	178	mRNA/mRNA	BNT	HDQ, MTX
IA	68	1moV4	1moV3	4680	3403	mRNA/mRNA	BNT	aTNF. Pred
IA	49	1moV1	NA	NA	NA	mRNA/mRNA	BNT	HDQ MTX
IA	62	1moV1	NA	NA	NA	mRNA/mRNA	BNT	HDQ
SARDs	61	1moV4	1moV3	2756	2825	v/v	BNT	HDQ
SARDs	60	1moV3	3moV2	867	556	mRNA/mRNA	BNT	HDQ
SARDs	62	1moV4	1moV3	17969	12938	mRNA/mRNA	BNT	MTX
SARDs	44	1moV3	3moV2	172	254	mRNA/mRNA	BNT	MTX
SARDs	64	1moV4	3moV2	74	96	v/mRNA	BNT	Ritux
SARDs	38	1moV4	1moV3	3734	4104	v/mRNA	mRNA1273	AZA
SARDs	39	3moV2	NA	NA	NA	v/mRNA	mRNA1273	AZA
SARDs	30	1moV4	NA	NA	NA	mRNA/mRNA	BNT	aTNF
MS	53	1moV2	NA	NA	NA	v/mRNA	mRNA1273	GLA
MS	75	1moV1	NA	NA	NA	mRNA/mRNA	BNT	None
MS	56	1moV3	3moV3	2304	2811	v/mRNA	mRNA1273	INF
IBD	50	1moV2	NA	NA	NA	mRNA/mRNA	BNT	VED
IBD	67	1moV4	3moV2	591	584	mRNA/mRNA	mRNA1273	VED
IBD	30	1moV4	1moV3	1339	1464	mRNA/mRNA	BNT	UST
IBD	20	1moV4	1moV3	6221	6375	mRNA/mRNA	BNT	aTNF
IBD	64	1moV3	3moV2	497	536	v/mRNA	mRNA1273	5-ASA
IBD	45	1moV3	3moV2	Neg	neg	mRNA/mRNA	BNT	aTNF

IMID = immune mediated inflammatory disease; IA= inflammatory arthritis; SARDs= Systemic autoimmune rheumatic disease; MS= multiple sclerosis; IBD= inflammatory bowel disease; NA not available (no prior visit with serology data). 1moV1 = 1 month post first vaccine, 1moV2 = 1 month post second vaccine; 3moV2 = 3 months post second vaccine; 1moV3 = 1 month post third vaccine; 1moV4 = 1 month post forth vaccine

[1] anti-Receptor Binding Domain BAU/ml; median (interquartile range) for all samples 1moV2 1686.8 (2199.44); 3moV2 555.8 (809.3); 1moV3 4280.3(6380.6)

[2] anti-Spike BAU/ml; median values for all samples 1moV2 1835 (2448); 3moV2 629.1 (883.4); 1moV3 4280.3(6380.6)

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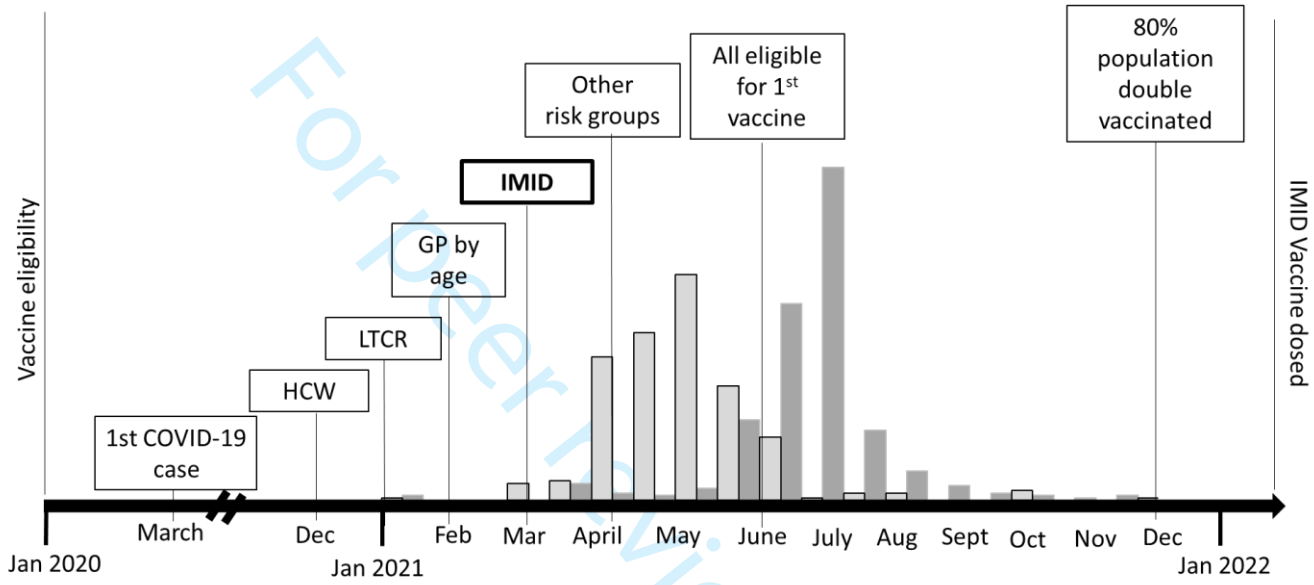

Supplemental Figure 1 Covid-19 vaccination timeline

HCW= health care workers; LTCR= long term care residents, GP= general population, IMID=immune mediated inflammatory disease; Bars indicate time when study participants (IMIDs) received vaccine. Light grey bars indicate time of first vaccine (V1); dark grey bars indicate time of second vaccine (V2).

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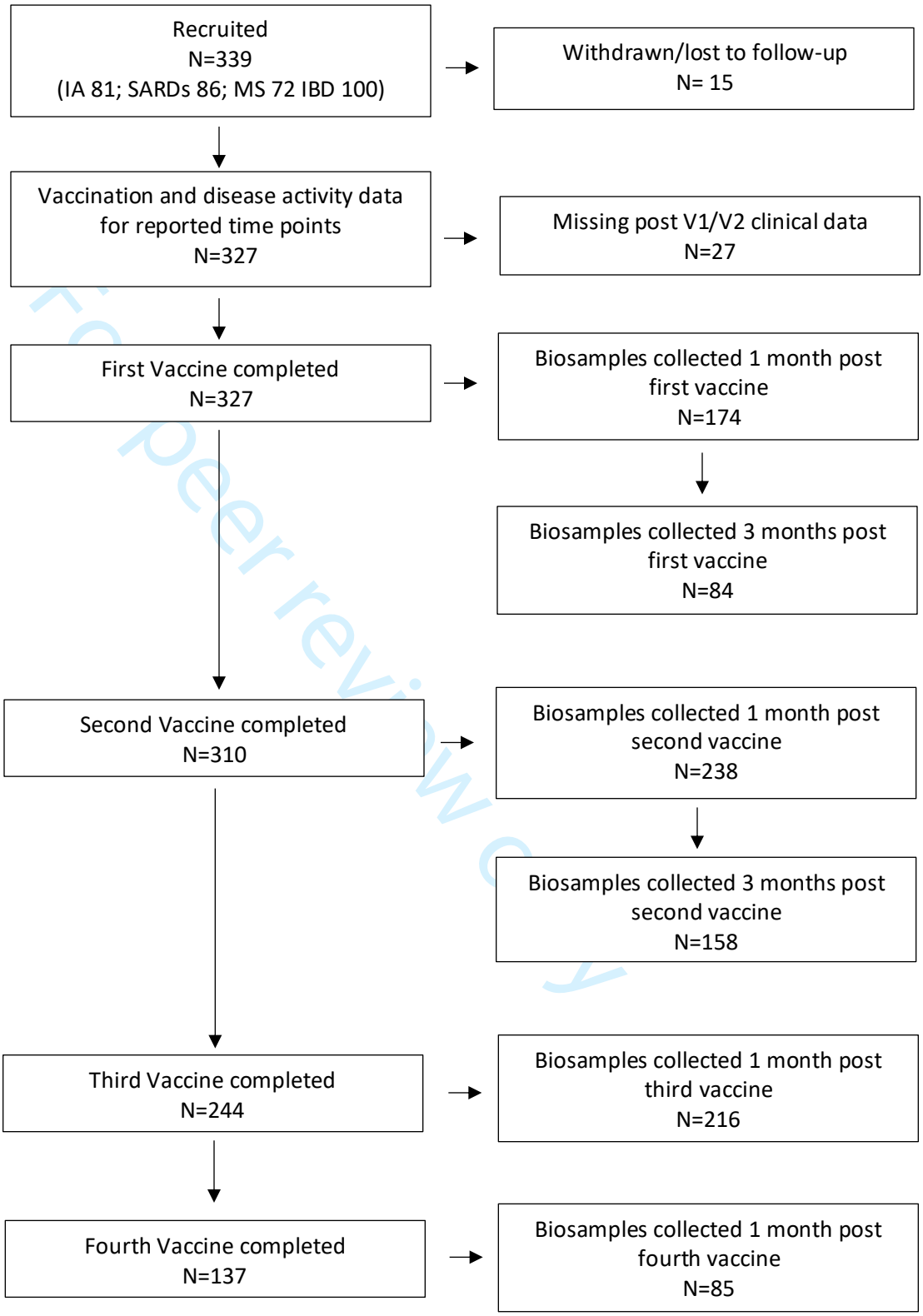

Supplemental Figure 2 Recruitment and sample acquisition chart

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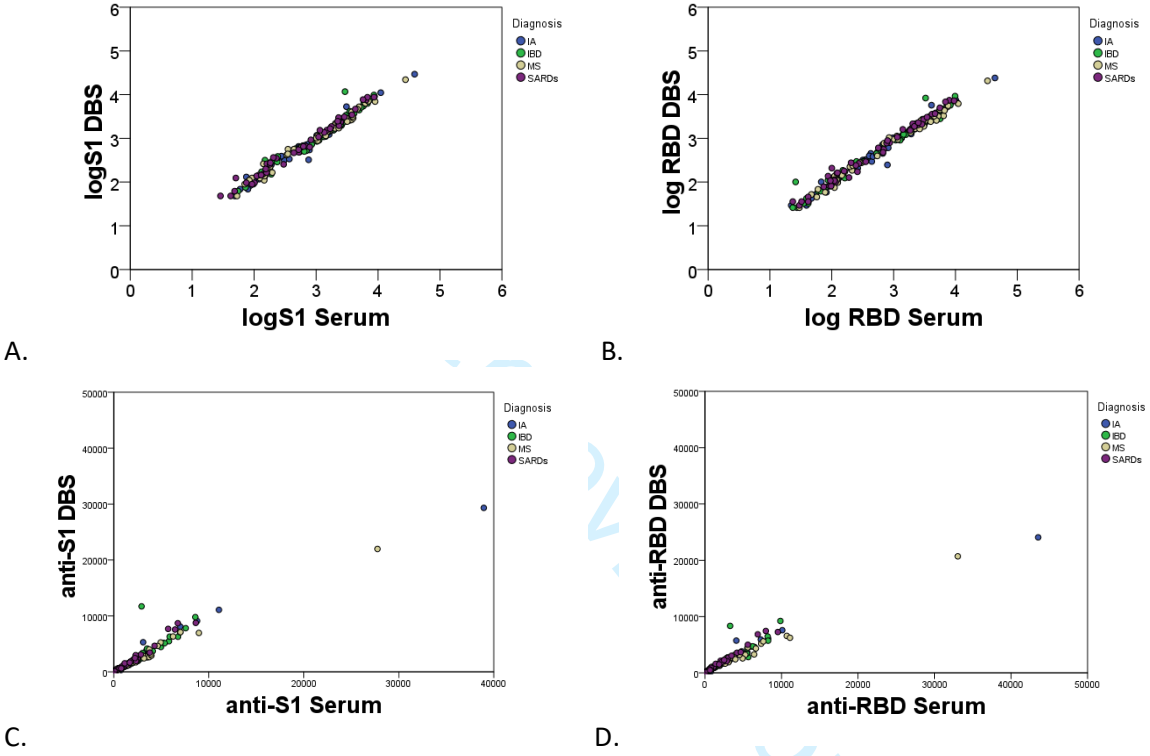
Supplemental Tables and Figures



Supplemental Figure 3 Correlation between assays performed using serum and dried blood spot samples. A. log Anti-S1 B log anti-RBD, C. anti-S1 BAU/ml D anti RBD BAU/ml (Kendalls tau b correlation co-efficient anti-S1 BAU/ml= 0.92; anti-RBD BAU/ml=0.93)

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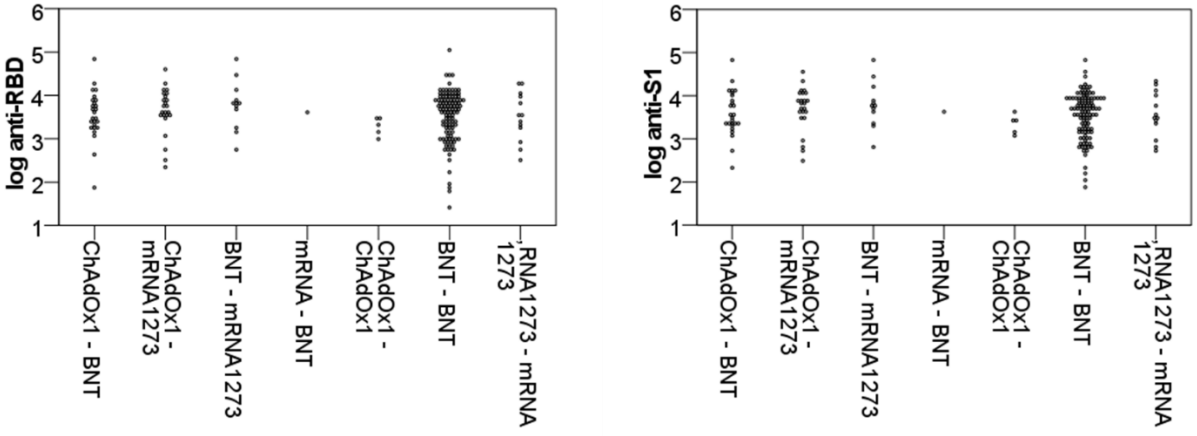
Supplemental Tables and Figures



Supplemental Figure 4 Anti-SARS-CoV2 titers 1 month post third vaccine based on first and second vaccine mixture.

RBD= Receptor binding domain; S1= Spike 1. All comparisons p=NS

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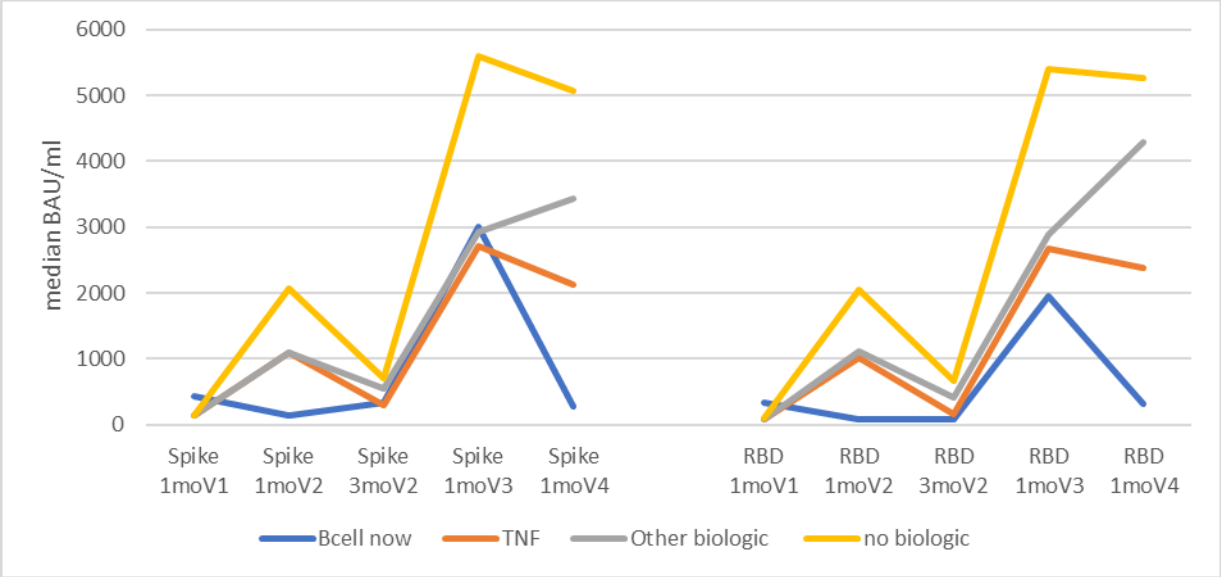

	B cell targeting now	anti-TNF	other biologic	no biologic	B cell targeting past
N 1moV1	2	14	8	77	2
N 1moV2	5	37	16	152	4
N 3moV2	5	34	20	154	5
N 1moV3	8	34	19	140	5
N 1moV4	5	18	11	46	5

Supplemental Figure 5 Median titers of anti-Spike and anti-RBD for individuals on different biologic categories and number participants in each treatment category for each visit.

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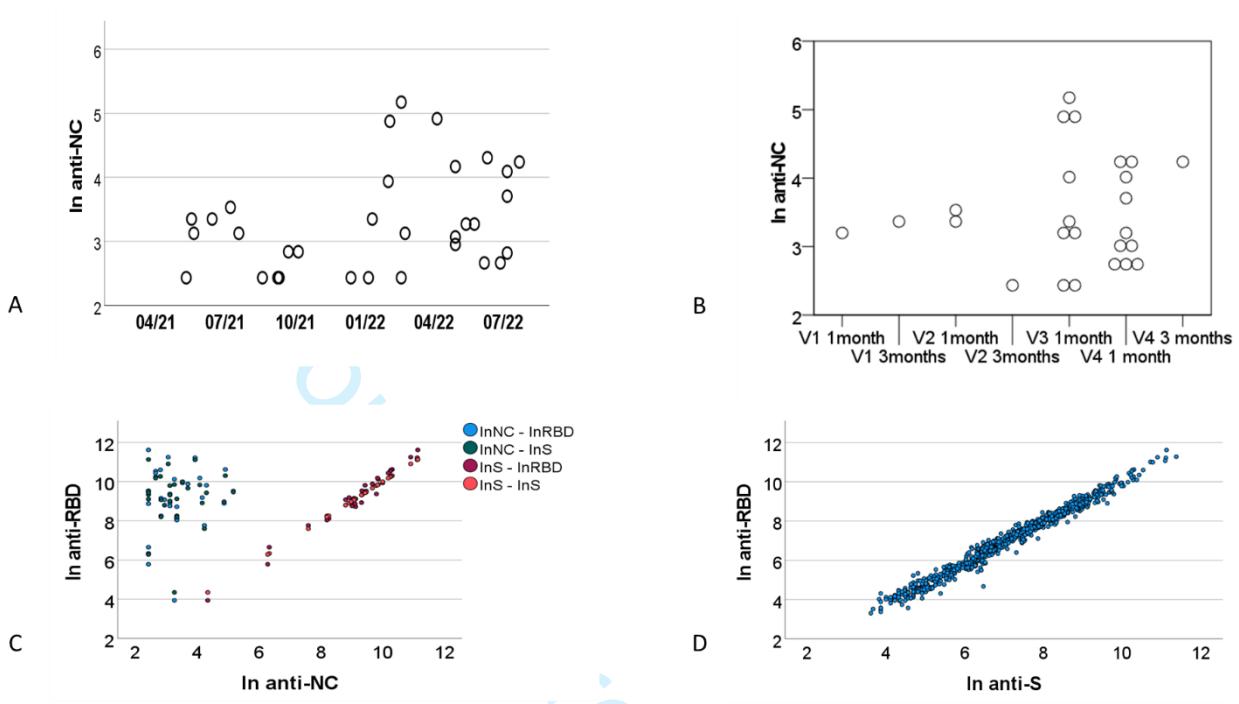

Supplemental Figure 6 Anti-Nucleocapsid antibody levels and correlations with Anti-Spike and Anti-Receptor Binding Domain antibodies

A. Anti-NC titer by Calendar month/year B. Anti-NC titer by Study visit C. Correlation of anti-NC anti RBD and anti-S1 titers in samples seropositive for anti-NC D. Correlation of anti-RBD and anti-S1 titers in all seropositive samples.

NC = nucleocapsid S = Spike; RBD = receptor binding domain; ln = natural log. V1 = vaccine 1; V2 = vaccine 2; V3 = vaccine 3; V4= vaccine 4. Values are natural log transformed BAU/ml.

Spearman correlation coefficient anti-NC with anti-S1 = 0.06 (p=NS); anti-NC with anti-RBD = 0.03 (p=NS); anti-S with anti-RBD 0.96 p<0.001. Figure 3 Disease activity before and after each vaccine

A Inflammatory arthritis B Systemic autoimmune rheumatic disease C Inflammatory bowel disease D Multiple Sclerosis

RAPID-3 Routine Assessment of Patient Index Data 3; SLAQ Systemic lupus activity questionnaire IBDSI-SF Inflammatory bowel disease symptom inventory – short form; EDSS Expanded disability status scale. V1 vaccine one; V2 vaccine two; V3 vaccine 3; V4 vaccine 4; mo = month(s).

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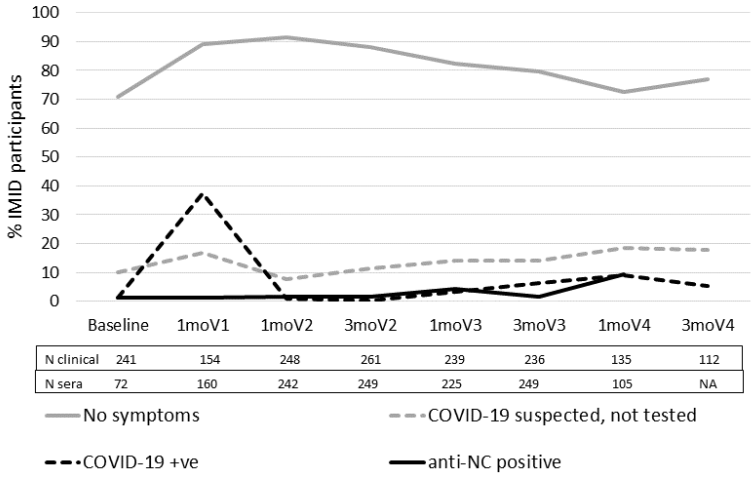

Supplemental Figure 7 COVID-19 infection

Grey line = no COVID-19 symptoms reported; Dashed grey line = COVID-19 suspected but not tested; dashed black line = COVID-19 infection confirmed by community based testing (rapid detection or polymerase chain reaction test); solid black line = proportion anti-nucleocapsid seropositive.

V1 vaccine one; V2 vaccine two; V3 vaccine 3; V4 vaccine 4; mo = month(s); N = number

STROBE Statement—checklist of items that should be included in reports of observational studies

	Item No	Recommendation	Page No
Title and abstract	1	(*a*) Indicate the study's design with a commonly used term in the title or the abstract	1
		(*b*) Provide in the abstract an informative and balanced summary of what was done and what was found	4
Introduction			
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported	7
Objectives	3	State specific objectives, including any prespecified hypotheses	7-8
Methods			
Study design	4	Present key elements of study design early in the paper	7-8
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection	7-8
Participants	6	(*a*) *Cohort study*—Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up *Case-control study*—Give the eligibility criteria, and the sources and methods of case ascertainment and control selection. Give the rationale for the choice of cases and controls *Cross-sectional study*—Give the eligibility criteria, and the sources and methods of selection of participants	7-8
		(*b*) *Cohort study*—For matched studies, give matching criteria and number of exposed and unexposed *Case-control study*—For matched studies, give matching criteria and the number of controls per case	
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable	8-9
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group	*7-9*
Bias	9	Describe any efforts to address potential sources of bias	7-9
Study size	10	Explain how the study size was arrived at	
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why	7-9
Statistical methods	12	(*a*) Describe all statistical methods, including those used to control for confounding	9
		(*b*) Describe any methods used to examine subgroups and interactions	9
		(*c*) Explain how missing data were addressed	8
		(*d*) *Cohort study*—If applicable, explain how loss to follow-up was addressed *Case-control study*—If applicable, explain how matching of cases and controls was addressed *Cross-sectional study*—If applicable, describe analytical methods taking account of sampling strategy	NA
		(*e*) Describe any sensitivity analyses	

Continued on next page

Results			
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed	10 and S Fig 2
		(b) Give reasons for non-participation at each stage	S Fig 2
		(c) Consider use of a flow diagram	S Fig2
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders	10 Table1
		(b) Indicate number of participants with missing data for each variable of interest	Table 1
		(c) *Cohort study*—Summarise follow-up time (eg, average and total amount)	
Outcome data	15*	*Cohort study*—Report numbers of outcome events or summary measures over time	*Table 1*
		Case-control study—Report numbers in each exposure category, or summary measures of exposure	
		Cross-sectional study—Report numbers of outcome events or summary measures	
Main results	16	(*a*) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included	10-12 Figure 1,2,4 Table 3
		(*b*) Report category boundaries when continuous variables were categorized	
		(*c*) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period	
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses	10-12
Discussion			
Key results	18	Summarise key results with reference to study objectives	12
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias	13-14
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence	13-14
Generalisability	21	Discuss the generalisability (external validity) of the study results	14
Other information			
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based	15

*Give information separately for cases and controls in case-control studies and, if applicable, for exposed and unexposed groups in cohort and cross-sectional studies.

Note: An Explanation and Elaboration article discusses each checklist item and gives methodological background and published examples of transparent reporting. The STROBE checklist is best used in conjunction with this article (freely available on the Web sites of PLoS Medicine at http://www.plosmedicine.org/, Annals of Internal Medicine at http://www.annals.org/, and Epidemiology at http://www.epidem.com/). Information on the STROBE Initiative is available at www.strobe-statement.org.