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Letter

Highly variable SARS-CoV-2 spike antibody responses to two doses of COVID-19 RNA vaccination in patients with multiple myeloma

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COVID-19 mRNA vaccines are highly efficacious in preventing COVID-19 morbidity and mortality in phase 3 clinical studies as well as in real-world settings. Emerging evidence suggests that some individuals with underlying comorbidities may mount suboptimal antibody responses to SARS-CoV-2 immunization (Addeo et al., 2021; Monin et al., 2021; Thakkar et al., 2021). Indeed, patients with multiple myeloma (MM) are immuno-compromised due to defects in humoral and cellular immunity as well as due to immunosuppressive therapy. Preliminary reports indicate that the antibody response in MM after the initial dose of SARS-CoV-2 mRNA vaccine is attenuated and delayed compared to healthy controls (Bird et al., 2021; Terpos et al., 2021). Moreover, MM patients who receive anti-CD38 monoclonal antibodies may have poorer vaccine-induced antibody responses even after completion of the full two-dose mRNA vaccine regimen (Pimpinelli et al., 2021). The kinetics of the vaccine responses in MM patients with prior COVID-19 infection and the impact of treatments, including BCMA-targeting agents, to vaccine response remain unknown.

We analyzed SARS-CoV-2 spike-binding IgG antibody levels in 320 MM patients who received COVID-19 vaccinations in early 2021 (69.1% BNT162b2 by Pfizer-BioNTech, 27.2% mRNA-1273 by Moderna, and 3.8% unknown) through the use of the COVID-SeroKlir Kantaro SARS-CoV-2 IgG test (Amanat et al., 2020). Of these patients, 18.8% (60/320) had COVID-19 prior to immunization. A detailed description of the patients and their clinical MM disease characteristics is presented in Table S1A. Of 320 MM patients, 260 (81.3%) had SARS-CoV-2 spike-binding IgG antibody levels measured at least 10 days after receiving the second vaccine dose (median 51 days, range 11–118 days). Of the fully immunized MM patients, 84.2% (219/260) mounted measurable SARS-CoV-2 spike-binding IgG antibody levels which varied by three orders of magnitude (median 149 AU/mL, range: 5–7,882 AU/mL), and 41 individuals (15.8%) had values below the level of detection (Figure S1A). Vaccine-induced antibody responses in the control group of 67 health care workers selected from an ongoing observational study to best match the MM population were, in comparison, more homogeneous (median 300 AU/mL, range: 21–3,335 AU/mL), and no individuals had antibody levels below the level of detection. Notably, antibody levels in the 38 fully vaccinated MM patients with prior reported COVID-19 infections were 10 times higher than those of MM patients that were naive at the time of vaccination (median for COVID-19 survivors: 801 AU/mL [range: 0–7,882 AU/mL] versus median for COVID-19 naive MM patients: 68.5 AU/mL [range: 0–3,174 AU/mL], $p < 0.001$, Mann-Whitney *U* test). This difference has been described previously for healthy vaccinated individuals (Ebinger et al., 2021; Krammer et al., 2021). Repeat antibody measurements from before the initial vaccination to 60 days after the second vaccination confirm delayed and suboptimal antibody responses, particularly in patients with MM and without prior SARS-CoV-2 infection compared to vaccinated healthy individuals without co-morbidities (Figure S1C).

Patients receiving myeloma treatment had significantly lower SARS-CoV-2 spike-binding IgG antibody levels after two vaccine doses ($p = 0.004$, Mann-Whitney *U* test) compared to patients not receiving anti-myeloma therapy (median antibody level on active therapy: 70 AU/mL, compared to MM patients without active therapy: 183 AU/mL). Looking at treatment categories, we found significantly lower antibody levels for patients receiving anti-CD38-containing regimens ($p < 0.001$) and BCMA-targeted therapy ($p = 0.003$) but not for the other treatments ($p = 0.55$) compared to patients not actively receiving anti-myeloma therapy (Figure S1B).

Of note, 15.8% of the MM patients (41/260) failed to develop any SARS-CoV-2 spike-binding IgG antibodies despite having received both doses of mRNA vaccines. 24/41 (58.5%) of these "non-responders" were on anti-CD38 antibody-containing therapy at the time of vaccination, 13/41 (31.7%) were on anti-BCMA bispecific antibody therapy, and 4/41 (9.8%) had undergone anti-BCMA CAR-T therapy more than three months prior. Univariate analysis showed a significant association of the following disease-related factors with the absence of SARS-CoV-2 spike-binding IgG despite completing the full immunization schedule: more previous lines of treatment (>3 lines, $p = 0.035$ or >5 lines, $p = 0.009$), receiving active MM treatment ($p = 0.005$), grade 3 lymphopenia at time of vaccination ($p = 0.018$), receiving anti-CD38 monoclonal antibody therapy ($p = 0.042$), and receiving BCMA-targeted therapy ($p < 0.001$). Multivariate logistic regression found that, after correcting for age, vaccine type, lines of treatment, time since MM diagnosis, response status, and lymphopenia, anti-CD38-containing treatment ($p = 0.005$, odds ratio [OR] = 4.258) and BCMA-targeted treatment ($p < 0.001$, OR = 10.269) remained significantly associated with the probability of not developing antibodies after vaccination (Table S1A).

The clinical relevance of these observations is further emphasized by the fact that we observed 10 cases of COVID-19 in MM patients after one (n = 7) or both (n = 3, no anti-spike IgG antibodies at the time of infection) doses of the mRNA vaccination (Table S1B). Six patients received outpatient treatment (including infusion of anti-spike monoclonal antibodies in 4/6 patients). However, four of these patients required subsequent hospitalization due to severe COVID-19, and one patient, who had no detectable SARS-CoV-2 spike-binding IgG antibodies >10 days after full vaccination, died after prolonged intubation for hypoxic respiratory failure. None of the other MM patient developed symptoms suggestive of COVID-19 after vaccination with a median follow-up of 122 days (range 13–185 days) after the first dose.

This evidence, taken together, shows that MM patients mount a highly variable antibody response after completing the recommended two-dose COVID-19 vaccination regimen, and 15.8% develop no detectable SARS-CoV-2 spike IgG antibodies. The current analysis represents a real-world, convenience sample in which not all participants were able to give samples for all time points. Of note, the COVID-SeroKlir Kantaro SARS-CoV-2 IgG Ab has a large dynamic range, but some antibody values included were capped at 125 AU/mL for technical reasons. These capped antibody values could potentially mask a bigger difference between MM patients and healthy controls. Only two out of the 260 MM patients shown in Figure S1A had capped test values (>125 AU/mL). It is important to note that the current report focuses on the quantification of spike-binding IgG antibody levels, but determination of virus neutralization, IgG subtype, and T cell immunity is needed in order to fully understand COVID-19-vaccine-induced immune responses in MM patients. These studies are ongoing and will complement the data presented. Follow-up studies will demonstrate the durability of vaccine-induced antibody responses beyond three months after the second vaccine dose. It is possible that individuals that mount low-to-modest antibody responses will "sero-revert" more rapidly than those with very high antibody titers.

Our findings underscore the need for routine serological monitoring of MM patients following COVID-19 vaccination to allow for personalized risk reduction measures in the context of relaxing mask and social distancing mandates for vaccinated individuals. The combination of specific risk factors in the MM population and the potential for cancer-directed therapies to hamper vaccine responses more broadly (Addeo et al., 2021; Thakkar et al., 2021) support the need for clinical trials that assess the use of prophylactic strategies (e.g., monoclonal antibodies) to mitigate SARS-CoV-2 infection risk in patients who are likely to have suboptimal vaccine response as well as studies that explore additional immunization strategies with different vaccine types or booster vaccinations (Werbel et al., 2021).

SUPPLEMENTAL INFORMATION

Supplemental Information can be found online at https://doi.org/10.1016/j.ccell.2021.06.014.

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DECLARATION OF INTERESTS

The Icahn School of Medicine at Mount Sinai has filed patent applications relating to SARS-CoV-2 serological assays and NDV-based SARS-CoV-2 vaccines, and these list Florian Krammer as co-inventor. Viviana Simon and Carlos Cordon-Cardo are listed on the serological assay patent application as co-inventors. Mount Sinai has spun out a company, Kantaro, to market serological tests for SARS-CoV-2. Florian Krammer has consulted for Merck and Pfizer (before 2020) and is currently consulting for Seqirus and Avimex. The Krammer laboratory is collaborating with Pfizer on animal models of SARS-CoV-2. Bo Wang reports consulting fees for Sanofi Genzyme. Ajai Chari reports grants and personal fees from Janssen, Bristol Myers Squibb (Celgene), Amgen, Seattle Genetics, and Millennium Pharmaceuticals/Takeda and personal fees from Karyopharm, Sanofi, Oncopeptides, Antengene, Glaxo Smith Kline, Secura Bio, Shattuck Labs, Genentech, and Abbvie. Florian Krammer reports grants and personal fees from Pfizer and personal fees from Seqirus and Avimex. Sundar Jagannath reports consulting fees for Bristol Myers Squibb (Celgene), Janssen, Karyopharm Therapeutics, Legend Biotech, Sanofi, and Takeda. Samir Parekh reports consulting fees from Foundation Medicine and research funding from Bristol Myers Squibb (Celgene), Karyopharm, and Amgen. Other authors report no relevant conflicts of interest.

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Supplemental information

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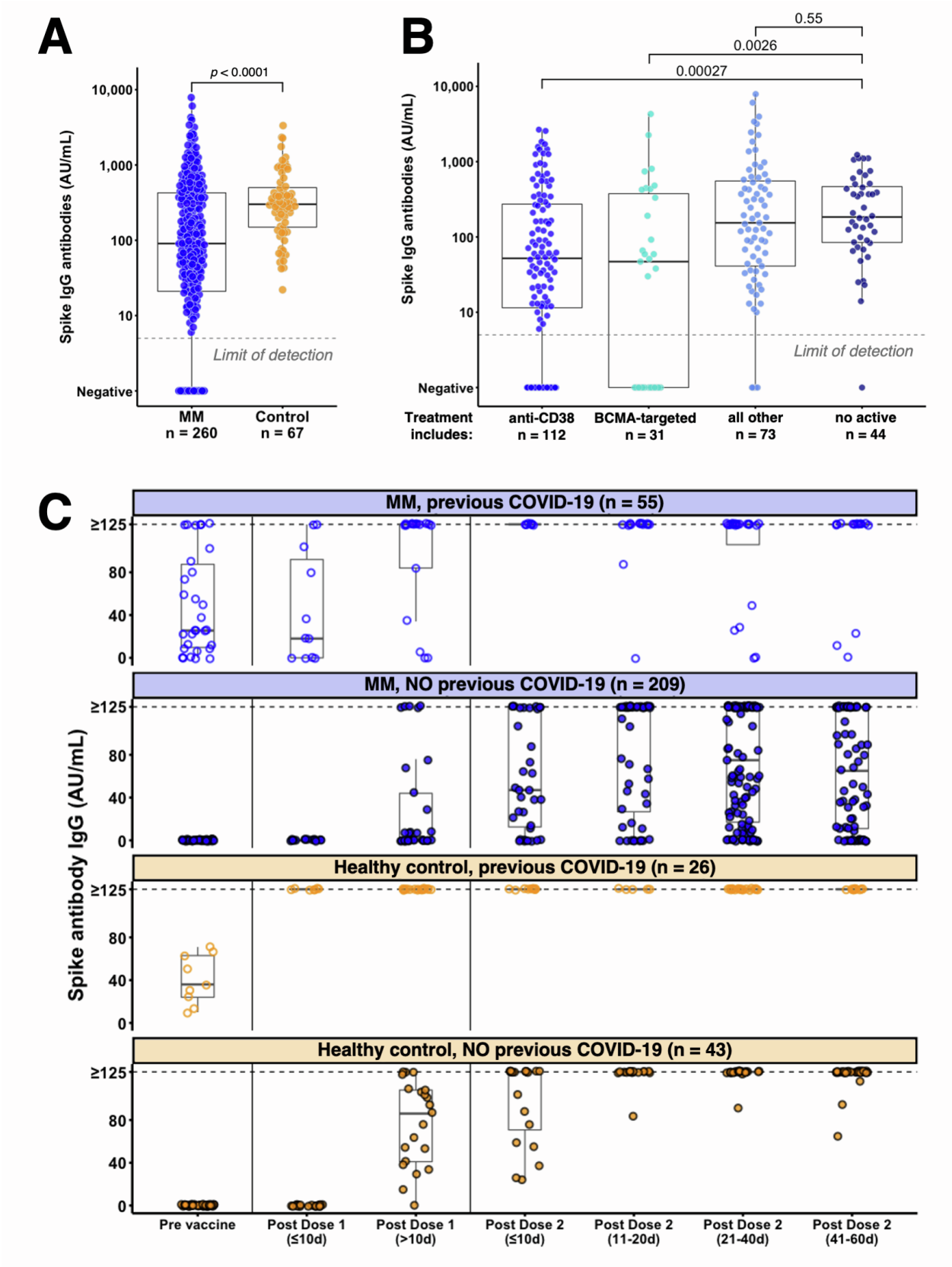
A
p < 0.0001
Spike IgG antibodies (AU/mL)
10,000
1,000
100
10
Negative
Limit of detection
MM
n = 260
Control
n = 67
B
0.55
0.0026
0.00027
Spike IgG antibodies (AU/mL)
Limit of detection
Treatment includes:
anti-CD38
n = 112
BCMA-targeted
n = 31
all other
n = 73
no active
n = 44
C
MM, previous COVID-19 (n = 55)
MM, NO previous COVID-19 (n = 209)
Healthy control, previous COVID-19 (n = 26)
Healthy control, NO previous COVID-19 (n = 43)
Spike antibody IgG (AU/mL)
≥125
80
40
0
Pre vaccine
Post Dose 1 (≤10d)
Post Dose 1 (>10d)
Post Dose 2 (≤10d)
Post Dose 2 (11-20d)
Post Dose 2 (21-40d)
Post Dose 2 (41-60d)

Figure S1. SARS-CoV-2 anti-spike IgG antibody responses in patients with multiple myeloma (MM) before and after COVID-19 RNA vaccination.

(**A**) Anti-SARS-CoV-2 spike IgG antibody level at least 10 days after receiving two doses of COVID-19 mRNA vaccine in MM patients versus healthy controls. Antibody concentrations measured in artificial units per mL (AU/mL) are depicted on a log-10 scale to better capture the full range of responses.

(**B**) Anti-SARS-CoV-2 spike IgG antibody levels at least 10 days after receiving two doses of SARS-CoV-2 mRNA vaccine in MM patients are split according to major treatment groups.

(**C**) Time course of anti-SARS-CoV-2 spike IgG antibody levels in MM patients with COVID-19 (112 antibody measurements in 55 patients), in MM patients without prior COVID-19 (349 antibody measurements in 209 patients), in healthy controls that were seropositive (78 antibody measurements in 26 individuals) and seronegative (172 antibody measurements in 43 individuals) prior to vaccination. Sample collection time points are binned into seven distinct time intervals. Antibody concentrations shown are capped at 125 AU/mL (any value >40 AU/mL is categorized as strong positive). For each panel, the lower and upper hinges of the boxplot correspond to the first and third quartiles (the 25th and 75th percentiles) with a bold horizontal line indicating the median. Vertical whiskers are extended up to 1.5 times the interquartile range (IQR). Values above the brackets correspond to the *p* value of a Mann-Whitney *U* test for comparison of 2 independent samples.

Table S1A. Demographics, vaccine type, disease and treatment characteristics of the study participants included in this report with univariate and multivariate statistical analysis (i.e. multivariate logistic regression model with absence of detectable IgG antibody levels > 10 days after full vaccination as dichotomized outcome).

Table S1B. Characteristics of patients that developed COVID-19 at some time after receiving at least one mRNA vaccine dose.

Supplemental Materials and Methods

Study information and patient selection

Patients with multiple myeloma (MM): The study cohort consisted of 320 patients with and without previously documented COVID-19 pooled from two different non-interventional Institutional Review Board (IRB) approved study protocols at The Icahn School of Medicine at Mount Sinai.

A total of 112 MM patients were enrolled after obtaining written informed consent for an ongoing longitudinal study at our institution (IRB-16-00791). Patients had blood taken for analysis at multiple time points before or after administration of the SARS-CoV-2 mRNA vaccine. All specimens were coded prior to processing and antibody testing for all serum specimen was performed in a blinded manner. All participants with, at least, one post vaccine antibody data point available at the time of writing this report were included in the analysis.

The remaining 208 MM patients were identified under a retrospective study (IRB: GCO#: 11-1433) by conducting a chart review for patients at our MM clinic who had SARS-CoV-2 spike IgG results at various time points around SARS-CoV-2 mRNA vaccine administration. Chart review was conducted to retrieve patient clinical characteristics. The cohort may contain an overrepresentation of patients on BCMA-targeted treatments as we are a tertiary care center for MM where patients are routinely referred for treatment on clinical trials.

Control group: The PARIS (***P****rotection* ***A****ssociated with* ***R****apid* ***I****mmunity to* ***S****ARS-CoV-2*) study seeks to investigate the durability and effectiveness of SARS-CoV-2 immune responses in health care workers over time. The study was reviewed and approved by the Mount Sinai Hospital Institutional Review Board (IRB-20-03374). All participants provided informed consent prior to collection of data and specimen. All specimens were coded prior to processing and antibody testing for all serum specimen was performed in a blinded manner. The current report includes serological data from a subgroup selected to best match the demographics and age of the MM patient population. 69 participants over the age of 50 years without known underlying immune modulatory co-morbidities and, at least, one post vaccine dose 2 antibody test were included.

SARS-CoV-2 antibody ELISA: Anti-SARS-CoV-2 spike protein IgG antibody testing was performed using an anti-IgG assay developed at Mount Sinai Health System Department of Pathology in collaboration with the Icahn School of Medicine at Mount Sinai Department of Microbiology under a Food and Drug Administration (FDA) Emergency Use Authorization. The antibodies to SARS-CoV-2 spike protein were detected using an established two-step ELISA (COVID-SeroKlir Kantaro SARS-CoV-2 IgG Ab Kit 100% Prev, Cat. Nr. COV219). The assay shows a performance of 100% specificity and 95% sensitivity in in-house evaluation. The qualitative results are reported out as negative (less than 5 AU/mL), weak positive (5-15 AU/mL), moderate positive (16-39 AU/mL) and strong positive (40 AU/mL and above) as described in the FDA emergency authorization. In addition, quantitative results are reported in artificial Units/ml (AU/mL). Starting Mid-February 2021, the dynamic range of the spike binding antibody test was extended in order to better capture the high antibody titers mounted by many individuals upon receiving SARS-CoV-2 vaccination (standard range up to 125 AU/ml; extended range: no upper limit). Thus, while the antibody test is the same, the upper limit of detection changed depending on when the antibody test was performed (before or after February 2021). Figure 1A and 1B show antibody data generated with the extended dynamic range with the exception of two MM patients who had values of >125 AU/mL. The data shown in Fig. 1C summarizing the longitudinal antibody responses upon COVID-19 vaccination were capped at 125 AU/mL because for several of the MM patients we had antibody values of >125 AU/mL.

Statistical analysis: Continuous variables are presented throughout the manuscript as median with the range of values being shown by disclosing minimum and maximum values. In addition, individual antibody data points are shown. Categorical variables are shown as a percentage and the absolute number of subjects. Wherever two outcome groups are compared, Fisher's exact test was used to determine significance and where applicable odds ratios (ORs) were reported.

The Mann-Whitney U test was used to determine significance for continuous variables. A two-sided alpha < 0.05 was considered statistically significant. All statistical analyses were done using R (v4.0.2). Multivariate statistical analysis was done using the presence or absence of measurable anti-spike IgG response > 10 days after dose 2 as a binary outcome variable and conducted using the glm(…, family = “binomial”) function in R (v4.0.2).