Title

Discordant Virus-Specific Antibody Levels, Antibody Neutralization Capacity, and T-cell Responses Following 3 Doses of SARS-CoV-2 Vaccination in a Patient With Connective Tissue Disease

Permalink

https://escholarship.org/uc/item/88j08892

Journal

Open Forum Infectious Diseases, 8(8)

ISSN

2328-8957

Authors

Peluso, Michael J
Munter, Sadie E
Lynch, Kara L
et al.

Publication Date

2021-08-01

DOI

10.1093/ofid/ofab393

Peer reviewed

Discordant Virus-Specific Antibody Levels, Antibody Neutralization Capacity, and T-cell Responses Following 3 Doses of SARS-CoV-2 Vaccination in a Patient With Connective Tissue Disease

Michael J. Peluso,[1,a] Sadie E. Munter,[2,a] Kara L. Lynch,[3] Cassandra Yun,[3] Leonel Torres,[2] Nikita S. Iyer,[2] Joanna Donatelli,[2] Lindsay Ryan,[4] Amelia N. Deitchman,[5] Steven G. Deeks,[1] Rachel L. Rutishauser,[2] and Timothy J. Henrich[2]

[1]Division of HIV, Infectious Disease and Global Medicine, University of California San Francisco, San Francisco, California, USA, [2]Division of Experimental Medicine, University of California San Francisco, San Francisco, California, USA, [3]Department of Laboratory Medicine, University of California San Francisco, San Francisco, California, USA, [4]Department of Medicine, University of California San Francisco, San Francisco, California, USA, and [5]Department of Clinical Pharmacology, University of California San Francisco, San Francisco, California, USA

We report a patient with connective tissue disease who developed modest severe acute respiratory syndrome coronavirus 2 receptor binding domain–specific antibody levels and a lack of neutralization capacity, despite having received 3 mRNA coronavirus disease 2019 vaccines and holding anti-B-cell therapy for >7 months before vaccination. The patient developed virus-specific T-cell responses.

Keywords. COVID-19 vaccine; neutralizing antibodies; rheumatological disease; SARS-CoV-2; T-cell immunity.

Coronavirus disease 2019 (COVID-19) vaccines have been shown to be extremely successful in eliciting strong humoral immune responses that protect against individual and community-based infection [1, 2]. However, rapidly emerging data suggest that people on immunomodulatory medications for a variety of underlying medical conditions, including solid organ transplantation (SOT) and rheumatologic disease, may not develop severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2)–specific humoral or cellular responses to full COVID-19 vaccination [3–7]. For example, nearly one-half of SOT recipients failed to develop detectable anti-spike antibody responses after a single dose of mRNA vaccine in a recent

study [8], although many of these individuals were taking multiple immunosuppressive medications (eg, glucocorticoids, mycophenolate, calcineurin inhibitors). Reductions in antibody responses have also been observed in patients with rheumatologic and inflammatory disorders on a variety of biological and nonbiological disease-modifying antirheumatic drugs including those on a single agent [5, 7]. Overall, antibody neutralization appears to correlate with SARS-CoV-2 anti-receptor binding domain (RBD) antibody levels and virus-specific CD4 T-cell responses following vaccination and natural infection [2, 4, 9, 10].

There are several important questions that arise from these emerging data that are now commonly being asked by patients and clinicians alike. First, it is not known whether alternative vaccine strategies, such as giving a third dose of the same or a different vaccine, will lead to increased humoral and cellular immune responses in those with suboptimal responses after completing full vaccination. Second, it is not clear if stopping various immunosuppressive medications for a set period around vaccination will allow a more robust vaccine response. Third, it is not known whether detectable antibody or T-cell responses will be fully protective in immunocompromised individuals or mirror neutralization capacity of such a response [10]. In order to provide insight into these questions and spur discussion about how to manage vaccination in immunocompromised individuals, we performed longitudinal, in-depth humoral and cellular immune characterization before and after mRNA COVID-19 vaccination, including an additional third dose, in a patient on anti-B-cell therapy and low-dose steroids for connective tissue disease.

CASE REPORT

The patient is a 37-year-old female with a medical history of antinuclear antibody (ANA)–positive undifferentiated connective tissue disease who presented in October 2019 with acute onset of ventricular arrhythmias, marked exertional intolerance and postural tachycardia, shivering, hyperhidrosis, and neuropathic pain and paresthesia, with later development of myokymia. Subsequent workup revealed substantially elevated and rising titers of anti-GAD-65 and anti-SSA/anti-SSB antibodies and central autonomic dysfunction. Initial infusion of intravenous immunoglobulin (IVIG) enabled the patient to ambulate well enough for hospital discharge and resolved the paresthesia, but did not produce durable improvement, with temporary improvement after repeat IVIG infusions but then relapse and an overall downward trajectory. After trials of pulse steroids and plasma exchange therapy, the patient improved on a combination of the anti-B-cell monoclonal

antibody, rituximab (starting April 2020), oral steroid taper, hydroxychloroquine, and regular IVIG infusions, on which she remained at the time of SARS-CoV-2 vaccination. The patient's last rituximab dose was 231 days before her first dose of mRNA COVID-19 vaccine (Moderna), but she had a CD-19 B-cell percentage of ≤1% before and after vaccination (Figure 1). On advice from the patient's clinical team and clinical laboratory testing, she received a third mRNA vaccine dose (Moderna) as part of her clinical care. The decision to receive a third dose was independent from her participation in this study.

In order to determine humoral responses to COVID-19 vaccination, informed consent was obtained from the patient to collect plasma and peripheral blood mononuclear cells at several time points before and after vaccination and to collect information on clinical laboratory testing. The UCSF Institutional Review Board approved this study, and informed consent was obtained from the patient and vaccine control participants. Total SARS-CoV-2 anti-RBD antibody

levels were tested using a Pylon enhanced fluorescence assay, and we determined neutralization capacity using a surrogate virus neutralization test (sVNT) based on antibody-mediated blockage of the ACE2–spike protein–protein interaction, as previously described [11]. As shown in Figure 1A, the patient developed a low but detectable total antibody response 20 days after the first mRNA vaccine dose (31 relative light units [RLU]). Total antibody levels peaked 13 days after the second Moderna mRNA vaccine dose (2802 RLU) but started to decline by 27 days after this second administration (392 RLU). A third dose of Moderna vaccine was administered 47 days after the second vaccine, but total antibody levels continued to decline as shown in Figure 1A, decreasing to 135 RLU 43 days after the third vaccination and 13 days after rituximab infusion. In contrast, antibody levels in 2 healthy control participants not taking immunosuppressive medications that were measured 36 and 47 days after the second dose of mRNA vaccine (Moderna) were >2.5 times higher
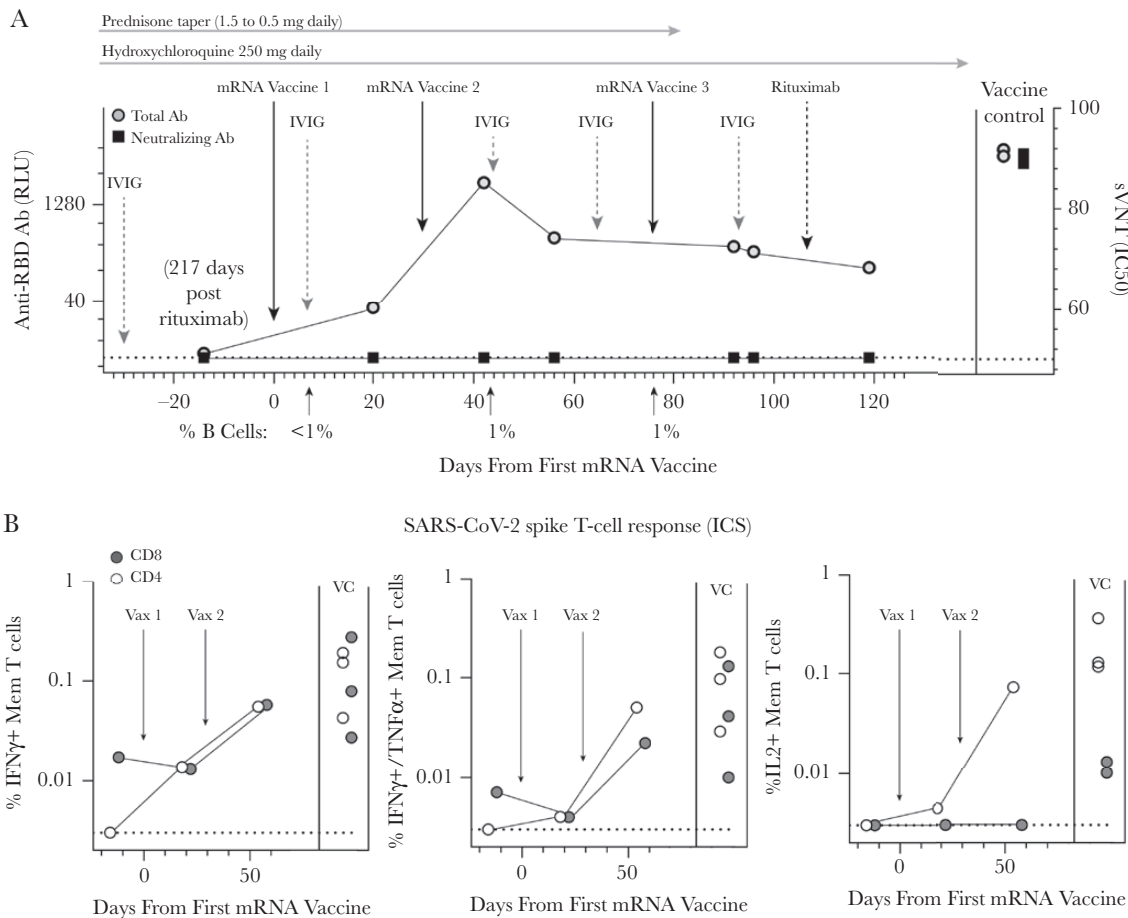

Figure 1. Longitudinal humoral and cellular immunity following SARS-CoV-2 vaccination in a participant with connective tissue disease on immunosuppressive medications. A timeline of SARS-CoV-2 RBD total antibody levels and neutralization IC50 with timing of vaccine administration and immunosuppressive medication use is shown in (A). SARS-CoV-2 spike peptide pool CD4 and CD8 T-cell responses as measures by an ICS assay are shown in (B). Immune responses are compared with vaccine control samples from participants following full mRNA vaccination. Abbreviations: COVID-19, coronavirus disease 2019; ICS, intracellular cytokine staining; IFN, interferon; RBD, receptor binding domain; RLU, relative light units; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2; sVNT, surrogate viral neutralization test; TNF, tumor necrosis factor; Vax, COVID-19 vaccination; VC, vaccine controls.

(9190 and 7285 RLU). Surprisingly, the patient had no detectable antibody neutralization by sVNT at any time point after vaccination, whereas the controls demonstrated robust neutralization (IC50 of 89 and 91).

We measured SARS-CoV-2 spike-specific CD4 and CD8 T-cell responses before and after the first and second doses of vaccine by intracellular cytokine staining (Figure 1B), as previously described [12]. Despite waning antibody levels and a lack of neutralization as discussed above, an increase in the frequency of interferon γ (IFNγ)–producing and IFNγ/tumor necrosis factor alpha (TNFα)–producing memory CD4 and CD8 T-cells after the second vaccine administration was observed following 8 hours of S peptide pool stimulation (PepTivator SARS-CoV-2 Prot_S; Miltenyi Biotec), similar to levels in 3 control participants' samples a median of 38 days after completion of mRNA vaccination. An increase in the frequency of memory CD4 T-cells expressing IL-2 in response to vaccination was also observed (Figure 1B). The patient felt well without clinical symptoms of COVID-19 before and during sample collection.

DISCUSSION

Although case reports are now emerging of suboptimal humoral and cellular immune responses to COVID-19 vaccination in immunocompromised individuals [4–8, 13], there is a paucity of data regarding the impact of a third dose of vaccine and correlations between commercial antibody assays and neutralization capacity in this population. This case provides insight into several of the outstanding but critically important questions regarding vaccine responses in the setting of immunomodulatory medications. First, despite developing a detectable antibody response after completing 2 doses of mRNA COVID-19 vaccination, these levels were lower than observed in several immunocompetent participants not taking immunosuppression following full mRNA vaccination and declined after the second vaccine dose. Total antibody levels continued to decline despite a third vaccine dose. Second, despite detectable, albeit waning, antibody levels, this patient mounted no detectable neutralization activity. Neutralization capacity is one of the best predictors of in vivo vaccine efficacy [2], and without neutralizing antibodies, it is not known if this patient will be protected from acquisition of COVID-19 or will demonstrate reduced severity if infected. Of note, antibody levels and neutralization are generally concordant in healthy and immunocompromised individuals and also correlate highly with viral-specific CD4 T-cell responses [9, 10, 12]. Third, despite suboptimal antibody responses, the patient developed SARS-CoV-2 spike-specific polyfunctional CD4 and CD8 T-cell responses that were generally similar to the postvaccine control samples. It is not known if these adaptive cellular responses are protective against COVID-19 acquisition or severe disease, as cutoffs between T-cell responses and vaccine efficacy have not been defined in any population [10]. A discordance between the development of anti-spike antibodies and T-cell immunity has been previously reported in SOT recipients, with up to 50% of patients who did not develop antibody responses still having SARS-CoV-2-specific T-cells [4]. Nonetheless, case reports are emerging of severe disease in transplant recipients despite full vaccination [13], suggesting that these patients may be vulnerable regardless of development of humoral or cellular responses.

The patient was on a low-dose prednisone taper and daily oral hydroxychloroquine therapy before and during vaccination but had no rituximab infusion for 231 days before vaccine initiation. Anti-B-cell antibody therapies have been associated with reduced antibody responses in those with rheumatologic conditions, but this case suggests that very low percentages of circulating B-cells, which can persist for many months following rituximab therapy, may cause impaired humoral vaccine responses. It is also possible that ongoing hydroxychloroquine therapy led to bone marrow suppression contributing to low circulating B-cell levels. The timing of the IVIG infusions and blood draws largely precludes any possibility that the more robust, though short-lived, antibody response after the second dose was due to passive transfer of antibodies from the IVIG; SARS-CoV-2-specific antibody levels were not available from the infused units. A limitation of the study was the use of a surrogate neutralization assay in lieu of measuring direct neutralization of infectious virus, and there may only be modest correlations between the surrogate and more direct assays [9].

Regardless of the pharmacologic drivers of suboptimal immune responses observed in this case, further clinical studies investigating the protective capacity of COVID-19 vaccination in immunosuppressed individuals and the potential utility of alternative vaccine strategies (ie, using a third homologous or heterologous vaccine dose) are urgently needed. Furthermore, the data highlight the utmost importance of widespread vaccination to obtain herd immunity to protect the proportion of the population who may have inadequate responses.

References

1. Ogbe A, Kronsteiner B, Skelly DT, et al; Oxford Immunology Network Covid-19 Response T Cell Consortium; Oxford Protective T Cell Immunology for COVID-19 (OPTIC) Clinical Team. T cell assays differentiate clinical and subclinical SARS-CoV-2 infections from cross-reactive antiviral responses. Nat Commun **2021**; 12:2055.

2. Khoury DS, Cromer D, Reynaldi A, et al. Neutralizing antibody levels are highly predictive of immune protection from symptomatic SARS-CoV-2 infection. Nat Med **2021**; 27:1205–11.

3. Boyarsky BJ, Werbel WA, Avery RK, et al. Antibody response to 2-dose SARS-CoV-2 mRNA vaccine series in solid organ transplant recipients. JAMA **2021**; 325:2204–6.

4. Cucchiari D, Egri N, Bodro M, et al. Cellular and humoral response after mRNA-1273 SARS-CoV-2 vaccine in kidney transplant recipients. Am J Transplant **2021**; 21:2727–39.

5. Haberman RH, Herati RS, Simon D, et al. Methotrexate hampers immunogenicity to BNT162b2 mRNA COVID-19 vaccine in immune-mediated inflammatory disease. Ann Rheum Dis **2021**; annrheumdis-2021-220597

6. Miele M, Busa R, Russelli G, et al. Impaired anti-SARS-CoV-2 humoral and cellular immune response induced by Pfizer-BioNTech BNT162b2 mRNA vaccine in solid organ transplanted patients. Am J Transplant **2021**; 10.1111/ajt.16702.

7. Simon D, Tascilar K, Fagni F, et al. SARS-CoV-2 vaccination responses in untreated, conventionally treated and anticytokine-treated patients with immune-mediated inflammatory diseases. Ann Rheum Dis **2021**; annrheumdis-2021-220461.

8. Boyarsky BJ, Werbel WA, Avery RK, et al. Immunogenicity of a single dose of SARS-CoV-2 messenger RNA vaccine in solid organ transplant recipients. JAMA **2021**; 325:1784–6.

9. Patel EU, Bloch EM, Clarke W, et al. Comparative performance of five commercially available serologic assays to detect antibodies to SARS-CoV-2 and identify individuals with high neutralizing titers. J Clin Microbiol **2021**; 59:e02257–20.

10. Sette A, Crotty S. Adaptive immunity to SARS-CoV-2 and COVID-19. Cell **2021**; 184:861–80.

11. Luo YR, Yun C, Chakraborty I, et al. A SARS-CoV-2 label-free surrogate virus neutralization test and a longitudinal study of antibody characteristics in COVID-19 Patients. J Clin Microbiol **2021**; 59:e0019321.

12. Peluso MJ, Deitchman AN, Torres L, et al. Long-term SARS-CoV-2-specific immune and inflammatory responses in individuals recovering from COVID-19 with and without post-acute symptoms. Cell Reports **2021**. doi:10.1016/j.celrep.2021.109518

13. Ali NM, Alnazari N, Mehta SA, et al. Development of COVID-19 Infection in Transplant Recipients After SARS-CoV-2 Vaccination. Transplantation. **2021**. doi:10.1097/TP.0000000000003836