Correspondence
Maurizio Zangari, Myeloma Center, University of Arkansas for Medical Sciences, 4301 W Markham St., Little Rock, AR 72223, USA.
Email: mzangari@uams.edu

ORCID

Meera Mohan https://orcid.org/0000-0002-6913-6526
Sharmilan Thanendrarajan https://orcid.org/0000-0003-2593-411X
Carolina Schinke https://orcid.org/0000-0002-2699-1741
Frits van Rhee https://orcid.org/0000-0001-9959-1282
Maurizio Zangari https://orcid.org/0000-0002-1720-3854

REFERENCES

1. Avet-Loiseau H, San-Miguel J, Casneuf T, et al. Evaluation of sustained minimal residual disease negativity with daratumumab-combination regimens in relapsed and/or refractory multiple myeloma: analysis of POLLUX and CASTOR. *J Clin Oncol*. 2021;39(10):1139-1149.
2. Kumar S, Paiva B, Anderson KC, et al. International Myeloma Working Group consensus criteria for response and minimal residual disease assessment in multiple myeloma. *Lancet Oncol*. 2016;17(8):e328-e346.
3. Rasche L, Alapat D, Kumar M, et al. Combination of flow cytometry and functional imaging for monitoring of residual disease in myeloma. *Leukemia*. 2019;33(7):1713-1722.
4. Nishimura KK, Barlogie B, van Rhee F, et al. Long-term outcomes after autologous stem cell transplantation for multiple myeloma. *Blood Adv*. 2020;4(2):422-431.
5. Paiva B, Gutiérrez NC, Rosiñol L, et al. High-risk cytogenetics and persistent minimal residual disease by multiparameter flow cytometry predict unsustained complete response after autologous stem cell transplantation in multiple myeloma. *Blood*. 2012;119(3):687-691.
6. Schinke C, Hoering A, Wang H, et al. The prognostic value of the depth of response in multiple myeloma depends on the time of assessment, risk status and molecular subtype. *Haematologica*. 2017;102(8):e313-e316.
7. Zhan F, Huang Y, Colla S, et al. The molecular classification of multiple myeloma. *Blood*. 2006;108(6):2020-2028.
8. Munshi NC, Avet-Loiseau H, Anderson KC, et al. A large meta-analysis establishes the role of MRD negativity in long-term survival outcomes in patients with multiple myeloma. *Blood Adv*. 2020;4(23):5988-5999.
9. Munshi NC, Avet-Loiseau H, Rawstron AC, et al. Association of minimal residual disease with superior survival outcomes in patients with multiple myeloma: a meta-analysis. *JAMA Oncol*. 2017;3(1):28-35.
10. Lahuerta JJ, Paiva B, Vidriales MB, et al. Depth of response in multiple myeloma: a pooled analysis of three PETHEMA/GEM clinical trials. *J Clin Oncol*. 2017;35(25):2900-2910.
11. Paiva B, Corchete LA, Vidriales MB, et al. Phenotypic and genomic analysis of multiple myeloma minimal residual disease tumor cells: a new model to understand chemoresistance. *Blood*. 2016;127(15):1896-1906.
12. Goicoechea I, Puig N, Cedena MT, et al. Deep MRD profiling defines outcome and unveils different modes of treatment resistance in standard and high risk myeloma. *Blood*. 2021;137(1):49-60.
13. Weinhold N, Ashby C, Rasche L, et al. Clonal selection and double-hit events involving tumor suppressor genes underlie relapse in myeloma. *Blood*. 2016;128(13):1735-1744.
14. Weinhold N, Heuck CJ, Rosenthal A, et al. Clinical value of molecular subtyping multiple myeloma using gene expression profiling. *Leukemia*. 2016;30(2):423-430.
15. Mohan M, Yarlagadda NK, Atwal D, et al. Clinical implications of loss of minimal residual disease (MRD) negativity in multiple myeloma. *J Clin Oncol*. 2020;38(15 suppl):8514.
16. Mohan M, Kendrick S, Pandey Y, et al. Late relapsing multiple myeloma ≥ 10 years after treatment on total therapy protocols are associated with good outcome. *Blood*. 2020;136(suppl 1):11-12.
17. Jurczyszyn A, Waszczuk-Gajda A, Castillo JJ, et al. Primary refractory multiple myeloma: a real-world experience with 85 cases. *Leuk Lymphoma*. 2020;61(12):2868-2875.
18. Majithia N, Vincent Rajkumar S, Lacy MQ, et al. Outcomes of primary refractory multiple myeloma and the impact of novel therapies. *Am J Hematol*. 2015;90(11):981-985.
19. Wudhikarn K, Mailankody S, Smith EL. Future of CAR T cells in multiple myeloma. *Hematology Am Soc Hematol Educ Program*. 2020;2020(1):272-279.

SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of this article.

Received: 19 May 2021 | Revised: 25 May 2021 | Accepted: 27 May 2021
DOI: 10.1002/ajh.26262

SARS-CoV-2 vaccination induces breakthrough hemolysis in paroxysmal nocturnal hemoglobinuria on complement inhibitor

To the Editor:

The SARS-CoV-2 infection and vaccination have raised concerns in complement mediated hemolytic anemias (i.e., paroxysmal nocturnal hemoglobinuria, PNH, and cold agglutinin disease, CAD), particularly if on treatment with complement inhibitors.[1–3] Among complement amplifying triggers (infections, traumas, and surgery[4,5]) an emerging causative agent may be represented by SARS-CoV-2 vaccines. In PNH patients on complement inhibitors, pharmacodynamic breakthrough hemolysis (BTH) has been defined as hemolysis reactivation with hemoglobin drop due to increased complement activity.[6] In CAD, where complement inhibitors are in clinical trials, a true BTH has not been defined yet, although hemolytic flares have been described under treatment.[7] Gerber et al.[5] recently reported six PNH patients, either untreated or on complement inhibitors, who experienced abrupt hemolytic crisis following SARS-CoV-2 vaccination (four Pfizer-BioNTech and two Moderna vaccine). Five patients were on intravenous ravulizumab and developed BTH after either the first

(N = 1), the second (N = 2), or after both doses (N = 2) of vaccine. Two patients were also receiving oral factor D inhibitor danicopan, and one experienced BTH only after the second dose, after interruption of danicopan (two missed doses) due to concerns about interaction with vaccine efficacy. Pérez-Lamas et al. described a hemolytic crisis occurring 2 days after the first dose of an mRNA Covid-19 vaccination in a CAD patient not under complement inhibition.[8] Here we describe a classic hemolytic PNH patient on C5 inhibitor, who developed severe BTH one day after the second dose of Moderna SARS-CoV-2 vaccine. This patient belongs to a cohort of 16 patients (13 PNH and three CAD) who received mRNA SARS-CoV-2 vaccine from March 2021. All of them were actively treated with complement inhibitors (five eculizumab, three intravenous ravulizumab, four subcutaneous ravulizumab, one factor B inhibitor iptacopan, and three C1s inhibitor sutimlimab) at our Hematology unit in Milan, Italy. The study was conducted in accordance with Helsinki Declaration and patients gave informed consent.

A 45-year-old lady suffering from PNH presented to the emergency room on April 21 the day after receiving the second dose of Moderna mRNA SARS-CoV-2 vaccine. The patient had a diagnosis of classic hemolytic PNH since 1996, and was on C5 complement inhibition from 2016 with suboptimal response and complete control of intravascular hemolysis (average Hb 9.5 g/dL with normal LDH, inadequate reticulocytosis). Since 2019 she has been enrolled in a clinical trial with subcutaneous ravulizumab, administered weekly. The patient had had no side effects following her first dose of the Moderna vaccine (March, 24). At presentation, she complained of hyperpyrexia (38.8°C) and several episodes of vomiting, abdominal pain and dark urine. Vital signs were normal and blood counts (Figure S1) displayed severe anemia with significant intravascular hemolysis (LDH 2.3 × ULN), consistent with BTH. Moderate neutropenia (0.83 × 10^9/L), and mild thrombocytopenia (100 × 10^9/L) were also noted, along with slightly increased C reactive protein (3.6 mg/dL), prolonged prothrombin time ratio (1.46), and increased D dimer (895 g/L). Molecular testing on nasopharyngeal swab for SARS-CoV-2 was negative, as well as chest X-ray, blood and urine cultures. The patient was started on intravenous antibiotics, hydration, and paracetamol with progressive amelioration of symptoms, and admitted to the hematology ward. BTH persisted, and low molecular weight heparin was started. On day+4 from presentation, Hb dropped to 6.7 g/dL with suboptimal reticulocyte counts (106 × 10^9/L). Recombinant human erythropoietin (rhEPO, epoetin alpha 40 000 IU subcutaneously) was administered with hematological improvement and repeated on day+14. Of note, subcutaneous ravulizumab was regularly continued along admission.

The occurrence of post-vaccine BTH in PNH patients should not discourage vaccination of this population. In fact, whilst Covid-19 may be fatal and difficult to handle,[1,2] BTH is more known and seems manageable with supportive care, as in the case reported. In our patient, neutropenia deserved hospital admission, and recombinant erythropoietin was helpful to improve anemia avoiding transfusions. Moreover, the prompt recognition of BTH is pivotal, since, besides anemia, active intravascular hemolysis puts patients at higher risk of thrombosis, through various mechanisms including free Hb release and nitric oxide depletion with consequent endothelial dysfunction.[6] Of note, increased thrombotic risk is also reported in CAD, although its management has not been clearly established. Thrombotic risk may be even higher during Covid-19, and after certain SARS-CoV-2 vaccines that may mimic autoimmune heparin-induced thrombocytopenia.[9] In our patient, prophylaxis with LMWH was therefore instituted and no thrombosis occurred.

Regarding the mechanisms of post-vaccine BTH, it has been described that SARS-CoV-2 spike protein may bind on nucleated cells and amplify complement alternative pathway by interfering with factor H.[10] However, the addition of spike protein subunit 1 did not increase lysis of PNH erythrocytes ex vivo,[5] and did not appear to bind red cells, weakening the hypothesis of a direct vaccine effect on erythrocytes. On the contrary, complement amplification triggered by the inflammatory/immune response following vaccine seems prominent. In our patient, no BTH was reported after the first vaccine administration, suggesting a pivotal role of higher immune response following the "booster" dose. In keeping with this hypothesis, pharmacodynamic BTH has been observed even after ravulizumab that has a half-life four times longer than eculizumab, and is associated with reduced pharmacokinetic BTH events.[11] On the other hand, a protective effect of proximal complement inhibitors (i.e. C1 inhibitors, C3 inhibitors, factor D and factor B inhibitors) may be hypothesized, although definite data are not available. Consistently, no hemolytic events occurred after either vaccine doses in the three CAD patients on the C1s inhibitor sutimlimab at our center.

Concerning timing of administration, in the experience by Gerber et al.,[5] all patients had received ravulizumab ≥4 weeks before vaccine, suggesting that administration of SARS-CoV-2 vaccination may be safer within 4 weeks from the last ravulizumab. However, our patient had received weekly subcutaneous ravulizumab the day immediately before both vaccine doses. It is therefore difficult to advise a proper timing of vaccine administrations, and close monitoring and patient education remain the only mandatory procedures.

In conclusion, this report shows that hemolytic flares may occur after SARS-CoV-2 vaccines and should be promptly recognized and managed through medical suspicion, patient education, and clinical monitoring.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

AUTHOR CONTRIBUTIONS

All authors followed patients, wrote the article and revised it for important intellectual content.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

Bruno Fattizzo[1,2], Juri Alessandro Giannotta[1], Nicola Cecchi[2], Wilma Barcellini[1]

[1]*Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy*

[2]Department of Oncology and Oncohematology, University of Milan, Milan, Italy

Correspondence
Bruno Fattizzo, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, via Francesco Sforza 35, 20100, Milan, Italy.
Email: bruno.fattizzo@unimi.it

ORCID
Bruno Fattizzo https://orcid.org/0000-0003-0857-8379
Juri Alessandro Giannotta https://orcid.org/0000-0001-9349-1627
Wilma Barcellini https://orcid.org/0000-0003-1428-9944

REFERENCES

1. Pike A, Muus P, Munir T, et al. COVID-19 infection in patients on anti-complement therapy: the Leeds National Paroxysmal Nocturnal Hemoglobinuria service experience. *Br J Hematol*. 2020;191(1):e1-e4.
2. Genthon A, Chiarabini T, Baylac P, et al. Severe COVID-19 infection in a patient with paroxysmal nocturnal hemoglobinuria on eculizumab therapy. *Leuk Lymphoma*. 2021;8:1-4.
3. Barcellini W, Giannotta JA, Fattizzo B. Are patients with autoimmune cytopenias at higher risk of COVID-19 pneumonia? The experience of a reference center in Northern Italy and review of the literature. *Front Immunol*. 2021;11:609198.
4. Berentsen S. Complement activation and inhibition in autoimmune hemolytic anemia: focus on cold agglutinin disease. *Semin Hematol*. 2018;55(3):141-149.
5. Gerber GF, Yuan X, Yu J, et al. COVID-19 vaccines induce severe hemolysis in paroxysmal nocturnal hemoglobinuria. *Blood*. 2021. https://doi.org/10.1182/blood.2021011548. [Epub ahead of print].
6. Risitano AM, Marotta S, Ricci P, et al. Anti-complement treatment for paroxysmal nocturnal hemoglobinuria: time for proximal complement inhibition? A position paper from the SAAWP of the EBMT. *Front Immunol*. 2019;10:1157.
7. Röth A, Barcellini W, D'Sa S, et al. Sutimlimab in cold agglutinin disease. *N Engl J Med*. 2021;384(14):1323-1334.
8. Pérez-Lamas L, Moreno-Jiménez G, Tenorio-Núñez MC, et al. Hemolytic crisis due to Covid-19 vaccination in a woman with cold agglutinin disease. *Am J Hematol*. 2021. https://doi.org/10.1002/ajh.26214. [Epub ahead of print].
9. Scully M, Singh D, Lown R, et al. Pathologic antibodies to platelet factor 4 after ChAdOx1 nCoV-19 vaccination. *N Engl J Med*. 2021;384:2202-2211. https://doi.org/10.1056/NEJMoa2105385
10. Yu J, Yuan X, Chen H, Chaturvedi S, Braunstein EM, Brodsky RA. Direct activation of the alternative complement pathway by SARS-CoV-2 spike proteins is blocked by factor D inhibition. *Blood*. 2020;136(18):2080-2089.
11. Brodsky RA, Peffault De Latour R, Rottinghaus ST, et al. Characterization of breakthrough hemolysis events observed in the phase 3 randomized studies of ravulizumab versus eculizumab in adults with paroxysmal nocturnal hemoglobinuria. *Hematologica*. 2020;106(1):230-237.

SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of this article.

Received: 12 May 2021 | Revised: 2 June 2021 | Accepted: 3 June 2021
DOI: 10.1002/ajh.26265

Vaccine-induced thrombotic thrombocytopenia following Ad26.COV2.S vaccine in a man presenting as acute venous thromboembolism

To the Editor:

The emergent need to combat the coronavirus disease 2019 (COVID-19) pandemic led to the development of several highly effective vaccines at unprecedented speeds, using highly advanced technology and scientific research. The initial trials demonstrated the safety of these vaccines, and the risk of serious adverse effects remains significantly low even after the vaccination of more than 1.5 billion people worldwide to date. The Ad26.COV2.S (Johnson & Johnson/Janssen) vaccine, a recombinant replication-incompetent adenovirus type 26 vector COVID-19 vaccine, was issued Emergency Use Authorization by the US Food and Drug Administration (FDA) on February 27, 2021, and has been administered to more than 10 million individuals as of June 1st, 2021.

In March 2021, a rare, life-threatening syndrome was first described by the European Medicines Agency as thrombotic thrombocytopenic syndrome (TTS) following ChAdOx1 nCoV-19 vaccination (AstraZeneca), a recombinant replication-deficient chimpanzee adenovirus vector.[1] The syndrome, now recognized as vaccine-induced thrombotic thrombocytopenia (VITT), results in pathologic anti-platelet factor 4 (PF4) antibodies leading to thrombocytopenia and thrombosis in the absence of heparin exposure, a mechanism similar to "autoimmune" heparin-induced thrombocytopenia (HIT).[2,3]

On April 13th, 2021, six cases of VITT were reported in the United States following vaccination by the Ad26.COV2.S vaccine, all of whom were women who developed cerebral venous sinus thrombosis (CVST).[4] This led to an 11-day pause in the administration of the vaccine, during which a thorough investigation led to its resumption by the Centers for Disease Control (CDC) and FDA, but revised to include a warning about this rare side effect. By April 27th, 2021, the number of VITT cases following Ad26.COV2.S vaccination reported in the US had risen to 15, all of whom were women of ages 20 to 50 years, and included 12 incidences of CVST.[5,6] To date, the only report of suspected VITT in a male Ad26.COV2.S vaccine recipient in the medical literature comes from the initial Johnson & Johnson phase 3 trials. One of the male trial participants who developed CVST, thrombocytopenia, and