Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Commercial immunoglobulin products contain cross-reactive but not neutralizing antibodies against SARS-CoV-2

To the Editor:

Coronavirus disease 2019 (COVID-19), caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), is one of the greatest modern public health crises. COVID-19–specific treatments, while being studied, are not yet readily available. We examined whether commercial prepandemic intravenous immunoglobulin (IVIG) contains cross-reactive antibodies that could bind and neutralize SARS-CoV-2. The receptor-binding domain (RBD), contained within the S1 subunit of the coronavirus spike protein, mediates viral entry by binding to the angiotensin-converting enzyme 2 receptor on host cells. The spike protein, including the RBD, is a known target for neutralizing antibodies in natural infection, and shares some epitopes with S proteins from common, circulating strains of human coronaviruses. We took 82 samples from 4 different brands manufactured in the United States and Europe (OctaPharma, Hoboken, NJ; Grifols, Barcelona, Spain and Durham, NC; CSL, Bern, Switzerland) and tested them for SARS-CoV-2 RBD binding using a standard ELISA. We found that all samples demonstrated the presence of cross-reactive antibodies above the negative controls; however, binding activity varied between individual lots and among brands (Fig 1, *A*).

To assess biological relevance, we took 7 samples (4 highest and 3 lowest RBD-binding) and examined neutralizing activity against a clinical isolate of SARS-CoV-2 in culture. Wells inoculated with virus alone or IVIG alone served as positive and negative controls, respectively. None of the 7 samples demonstrated SARS-CoV-2–neutralizing capacity at dilution 1:4 (Fig 1, *B*); meanwhile, convalescent patient serum neutralized virus at dilutions of 1:4, 1:16, and 1:64, but not at 1:256 (Fig 1, *C*). These results show that although IVIG contains cross-reactive antibodies against novel SARS-CoV-2, this does not confer viral neutralization.

Studies of immunoglobulin utility in other pandemics, such as the 2003 severe acute respiratory syndrome coronavirus and the 2012 Middle East respiratory syndrome coronavirus outbreaks, were largely inconclusive.[2] However, anti-infectious mechanisms by which immunoglobulin acts are complex and not limited to

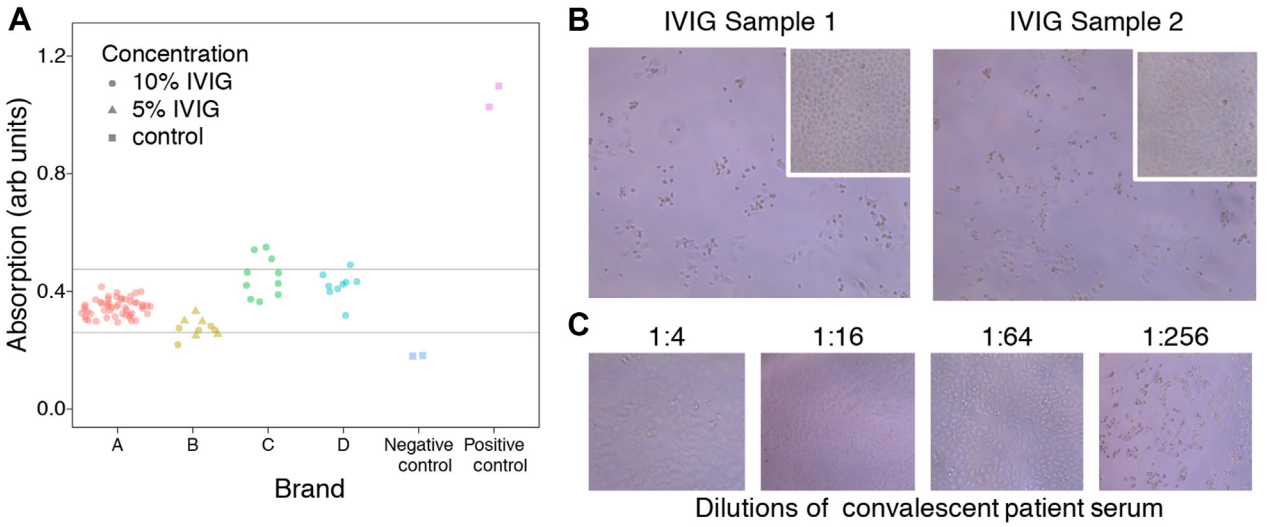

FIG 1. A, SARS-CoV-2 RBD antibody binding from commercially available IVIG at dilution 1:50 by ELISA. Each dot represents a different lot number of immunoglobulin product. The positive control is anti-spike antibody (CR3022, Creative Biolabs) at dilution 1:1000.[1] The negative control is human whole serum obtained from Sigma. Concentration indicates the protein percentage (wt/vol) in the commercial lot, before dilution for the ELISA. The 5% and 10% IVIG samples were first diluted to 0.66% (wt/vol) to achieve a common concentration of immunoglobulin, then diluted 1:50 for the ELISA. The dilution solution was PBS (Fisher Scientific, item no. BP3994) with 0.05% (vol/vol) Tween-20 (Sigma Aldrich, item no. P1379) and 1% (wt/vol) BSA (Rockland Antibodies & Assays, item no. BSA-50). The final concentration of immunoglobulin is approximately 0.0133% (wt/vol) or roughly 133 μg/mL of immunoglobulin. **B,** SARS-CoV-2 (Isolate USA-WA1/2020) was obtained from the Centers for Disease Control and Prevention and expanded in Vero E6 cells (ATCC). The virus titer used was 1×10^5 plaque-forming units (PFU)/mL. Vero cells were seeded at 7500/well in 96-well plates and cultured overnight. IVIG diluted 1:4 (20 μL) was mixed with 100 PFU of SARS-CoV-2 in 100 μL DMEM, incubated at 37 °C for 1 hour, and added to monolayers of Vero E6 cells in duplicate. Cytopathic effect was assessed after 3 days. Images of Vero E6 cells by microscopy (10×) after 72 hours in culture. Cytopathic effect (CPE) is visible in SARS-CoV-2–infected Vero E6 cells mixed with IVIG. Images show results of 2 of the 4 highest RBD-binding samples; the 3 lowest RBD-binding samples showed similar results (not shown). The insets show co-culture of Vero E6 cells with immunoglobulin but without virus, demonstrating that IVIG does not kill Vero E6 cells. **C,** Reduction of CPE in SARS-CoV-2–infected Vero E6 cells mixed with convalescent patient serum at dilutions of 1:4, 1:16, and 1:64; however, CPE was seen when serum was diluted to 1:256. *DMEM*, Dulbecco modified Eagle medium.

viral neutralization. For example, non-neutralizing influenza-specific antibodies can mediate complement fixation, phagocytosis, and antibody-dependent cellular cytotoxicity (ADCC). IVIG produced before the 2009 H1N1 pandemic had moderate titers of cross-reactive ADCC antibodies that eliminated H1N1-infected respiratory cells *in vitro*.[3] IVIG can also have anti-inflammatory effects that target immune-mediated pathology frequently seen during and after infection. Thus, evidence supports therapeutic antiviral and anti-inflammatory activity of IVIG beyond neutralization.

Data on the clinical utility of IVIG in COVID-19 are limited. IVIG from 1 manufacturer contained antibodies with reactivity to components of various coronaviruses but neutralization studies were not performed.[4] Over time, of course, all commercial immunoglobulin will contain SARS-CoV-2 antibodies. A case report described prompt recovery in a patient with severe COVID-19 after receiving plasma exchange and IVIG, suggesting that plasma exchange may clear pathogenic or inflammatory mediators while IVIG provides immunomodulatory and antiviral effects.[5] Although limited by study size and confounding variables, other case series reported that IVIG improved clinical outcomes in severe COVID-19, supporting its potential as adjuvant therapy.[6,7]

In summary, even prepandemic IVIG contains cross-reactive SARS-COV-2 RBD, but does not neutralize viral spread. Nonetheless, activities beyond neutralization such as ADCC, complement activation, and anti-inflammation may warrant its use in COVID-19.

Terrie S. Ahn, MD[a]
Brandon Han, BS[b,c]
Paul Krogstad, MD[b,d]
Chantana Bun, BS[a]
Lisa A. Kohn, MD, PhD[a]
Maria I. Garcia-Lloret, MD[a]
Robert Damoiseaux, PhD[b,c,e]
Manish J. Butte, MD, PhD[a,f]

From [a]the Division of Immunology, Allergy, and Rheumatology, Department of Pediatrics, [b]the Department of Molecular and Medical Pharmacology, [c]the California NanoSystems Institute, [d]the Division of Infectious Diseases, Department of Pediatrics, [e]the Department of Bioengineering, Samueli School of Engineering, and [f]the Department of Microbiology, Immunology, and Molecular Genetics, University of California Los Angeles, Los Angeles, Calif. E-mail: mbutte@mednet.ucla.edu.

This work was supported by the Jeffrey Modell Foundation, the UCLA AIDS Institute, and the Charity Treks fund.

Disclosure of potential conflict of interest: The authors declare that they have no relevant conflicts of interest. Immunoglobulin products were provided without financial support. The companies that provided immunoglobulin did not preview this manuscript and had no say in its content.

REFERENCES

1. Mustafa S, Balkhy H, Gabere MN. Current treatment options and the role of peptides as potential therapeutic components for Middle East respiratory syndrome (MERS): a review. J Infect Public Health 2018;11:9-17.
2. Jegaskanda S, Vandenberg K, Laurie KL, Loh L, Kramski M, Winnall WR, et al. Cross-reactive influenza-specific antibody-dependent cellular cytotoxicity in intravenous immunoglobulin as a potential therapeutic against emerging influenza viruses. J Infect Dis 2014;210:1811-22.
3. Díez JM, Romero C, Gajardo R. Currently available intravenous immunoglobulin contains antibodies reacting against severe acute respiratory syndrome coronavirus 2 antigens. Immunotherapy 2020;12:571-6.
4. Shi H, Zhou C, He P, Huang S, Duan Y, Wang X, et al. Successful treatment with plasma exchange followed by intravenous immunogloblin in a critically ill patient with 2019 novel coronavirus infection. Int J Antimicrob Agents 2020;56:105974.
5. Cao W, Liu X, Bai T, Fan H, Hong K, Song H, et al. High-dose intravenous immunoglobulin as a therapeutic option for deteriorating patients with coronavirus disease 2019. Open Forum Infect Dis 2020;7:ofaa102.
6. Xie Y, Cao S, Li Q, Chen E, Dong H, Zhang W, et al. Effect of regular intravenous immunoglobulin therapy on prognosis of severe pneumonia in patients with COVID-19. J Infect 2020;81:318-56.
7. Yuan M, Wu NC, Zhu X, Lee CC, So RT, Lv H, et al. A highly conserved cryptic epitope in the receptor binding domains of SARS-CoV-2 and SARS-CoV. Science 2020;368:630-3.

Available online Dec 17, 2020.
https://doi.org/10.1016/j.jaci.2020.12.003

mRNA COVID-19 vaccine is well tolerated in patients with cutaneous and systemic mastocytosis with mast cell activation symptoms and anaphylaxis

To the Editor:

Mastocytosis encompasses a heterogeneous group of diseases characterized by the presence of clonal mast cells (MCs) in tissues and symptoms of MC activation, including anaphylaxis.[1] Vaccination has been shown to cause exacerbation of MC mediator-related symptoms.[2] Vaccines against coronavirus disease 2019 (COVID-19) are the solution to the current pandemic, but reports of anaphylaxis following vaccination with the BNT162b2 Pfizer-BioNTech mRNA vaccine have emerged.[3] As such, it is important to provide evidence of the safety of mRNA vaccines in populations at risk for anaphylaxis, including patients with mastocytosis and MC activation symptoms.

We report here 2 cases of health care workers with direct contact with patients with COVID-19 and who had a diagnosis of cutaneous and systemic mastocytosis who had successful anti–COVID-19 vaccination. A 37-year-old female nurse with adult-onset monomorphic maculopapular cutaneous mastocytosis lesions, serum basal tryptase level of 12.4 ng/mL with indolent systemic mastocytosis, based on bone marrow MC aggregates with spindle-shape morphology, and negative KIT D816V mutation (World Health Organization major and minor criterion) presented with severe MC mediator-related symptoms including abdominal colicky pain, bloating and diarrhea, generalized pruritus and flare up of lesions, and osteopenia. She received the first dose of the Pfizer-BioNTech mRNA vaccine, BNT162b2, with premedication with H1 and H2 antihistamines, 1 hour before, and montelukast 10 mg, 1 and 24 hours, without side effects. A 47-year-old female nurse with adult-onset monomorphic maculopapular cutaneous mastocytosis, with serum basal tryptase level of 16.2 ng/mL and indolent systemic mastocytosis based on spindle-shaped bone marrow MCs, positive KIT D816V mutation, and MCs expressing CD25 and CD2 (World Health Organization 3 minor criteria) had a history of anaphylaxis with multiple drugs, and MC mediator-related symptoms including migraines, skin pruritus, gastroesophageal reflux, and osteopenia. She received the first dose of the same vaccine, with above premedication, and only had myalgias on the following day.

These 2 cases provide an initial evidence that mRNA COVID-19 vaccines are safe in patients with mastocytosis and MC activation symptoms, including anaphylaxis.

In patients with mastocytosis, the release of MC mediators following vaccination may be related to the activation of Toll-like receptors, noncanonical activation of FcεRI by superantigens bound to IgE, or complement activation.[2,3] Because the BNT162b2 vaccine contains polyethylene glycol (PEG),