Cite as: M. C. van Zelm, *Sci. Immunol.* 10.1126/sciimmunol.abq5901 (2022).

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Immune memory to SARS-CoV-2 Omicron BA.1 breakthrough infections: To change the vaccine or not?

Menno C. van Zelm[1,2]*

[1] Department of Immunology and Pathology, Central Clinical School, Monash University, Melbourne, VIC, Australia. [2] Department of Allergy, Immunology and Respiratory Medicine, Central Clinical School, Monash University, and Alfred Hospital, Melbourne, VIC, Australia.

***Corresponding author. Email: menno.vanzelm@monash.edu**

Analysis of memory B cell responses to Spike antigen after Omicron BA.1 breakthrough infections provides clues on whether "original antigenic sin" is in play.

Two studies recently published in *Science Immunology* address how well vaccine-induced immune memory can recognize and respond to breakthrough infections with the SARS-CoV-2 Omicron BA.1 variant (*1*, *2*). The COVID-19 pandemic has presented the global population with multiple challenges over the past two years. Following the development of novel and highly effective vaccines eliciting immune responses to the SARS-CoV-2 Spike protein, and addressing the challenges in their global rollout and uptake, we are now facing escape mutations in the Spike proteins of viral variants of concern (VoC), most notably in the Omicron variants.

The studies by Quandt *et al.* (*1*) and Kaku *et al.* (*2*) both start by evaluating the vaccine-induced antibody response in double- or triple-dose mRNA-vaccinated individuals (BNT162b2 or mRNA-1273). Both demonstrate that antibody amounts decline between one and six months after the second dose but are boosted by the third dose. Importantly, the relative recognition of VoC is strongly boosted, demonstrating an improved capacity for protection including against Omicron.

Quandt *et al.* examined separate groups of individuals that experienced omicron BA.1 breakthrough infection after their second or third dose of BNT162b2 (*1*). These individuals displayed high variant recognition with similar degrees of neutralization between the Wuhan, Beta, Delta and Omicron variants after dose 3 or after Omicron infection. The authors then went on to evaluate the memory B cell (Bmem) compartment with a series of full Spike or receptor binding domain (RBD) constructs that were tetramerized with a fluorochrome-conjugated streptavidin. This enabled flow cytometric evaluation of Bmem cells that recognized each of the VoC. Similar frequencies of Bmem cells specific for each of the variants were found. While Omicron infection led to great increases in Spike- and RBD-specific Bmem, these frequencies were expanded for each of the variants (Wuhan, Alpha, Delta, Omicron BA.1). Through combinatorial gating, it was shown that the majority of Bmem cells recognize each of the variants. Thus, Quandt *et al.* demonstrate that the circulating Bmem compartment induced by vaccination has the capacity to respond to each of the current VoC and is the likely source of the increased Omicron neutralization capacity after a third vaccination or Omicron infection.

Kaku *et al.* performed detailed cellular and molecular analysis on the Bmem cells after breakthrough infection, showing a relative expansion of RBD-reactive Bmem at the expense of N-terminal domain (NTD) or S2 recognition (*2*). Still, the relative frequencies of RBD Wuhan-specific Bmem that recognized the Omicron BA.1 variant were similarly high as after triple-dose vaccination (65-75%). Through single-cell sorting of 410 RBD specific Bmem cells from five donors, Kaku *et al.* evaluated their immunoglobulin gene sequences and the binding capacities of the corresponding recombinantly-produced antibodies. Nearly all antibody genes contained somatic mutations, and nearly all antibodies bound with higher affinity to Wuhan RBD than to the Omicron BA.1 variant. Thus, the breakthrough infection response seems to be dominated by a recall of vaccine-induced Bmem with the capacity to recognize Omicron.

The emergence of variants provides a challenge to the current vaccines and vaccination strategies because there is a risk of decline in vaccine efficacy as the number of mutations within Spike epitopes rises. Potentially, vaccine strain updates will be required in a manner similar to the annual influenza program. However, decision-making should be based on both real world and experimental evidence. Recent data demonstrate that the current COVID-19 vaccines reduce the risk of infection, but this protection is incomplete and wanes rapidly within weeks after the second dose (*3*). In contrast, the initial double-dose mRNA vaccination is highly protective against severe disease for >5 months from Wuhan and Delta variants. This efficacy is lower and lasts shorter against Omicron infection, but is highly boosted by third dose vaccination (*3*). Thus, it is not realistic to expect durable protection by COVID-19 vaccines from SARS-CoV-2 infection but instead, the aim for COVID-19 vaccines should be to provide maximum efficacy and durability against severe disease by new

variants, including Omicron.

Currently, there are no markers for protection against severe disease nor its durability. Considering the rapid contraction of the antibody response, serum IgG amounts will not be the best immune correlate. Circulating memory B and T cells persist for months after infection or vaccination and might provide insights into the nature of the response and clues for biomarkers (*4*–*6*). The publications by Quandt *et al.* and Kaku *et al.* give the first insights into the capacity of COVID-19 vaccine-immune memory to respond to SARS-CoV-2 variants (*1*, *2*). Both double- and triple-dosed vaccinated individuals mounted strong antibody and Bmem responses to Omicron infection. Furthermore, Kaku *et al.* demonstrate that the antibodies expressed by Bmem after Omicron infection have a higher affinity to the Wuhan Spike (*2*), indicating that these are derived from vaccine-induced Bmem and have a sufficiently high affinity to overcome the mutations in the Omicron Spike. These observations also hint at the potential mechanism by which the third vaccine dose boosts the relative capacity to bind to Omicron (*7*): the renewed response induces further affinity maturation to the Wuhan Spike, resulting in sufficiently high affinity to the mutated Omicron Spike (**Fig. 1**). Still, not all vaccine-induced Bmem can recognize Omicron and it remains unclear if this quantitative difference will affect vaccine effectiveness. Furthermore, it is unclear how durable the capacity is of Bmem to respond to Omicron after the third dose. These unknowns are important for decision-making on booster vaccine formulations and frequency, and will undoubtedly be addressed over the next few months.

Theoretically, a variant-specific vaccine has the capacity to generate more optimal immune memory to both conserved and new epitopes. However, if this vaccine is provided as a booster on top of previous Wuhan vaccinations, the new specificities would need to be derived from naïve B cells, which would compete with existing Bmem against conserved epitopes. By nature, Bmem have a pre-activated phenotype (*8*), allowing them to respond more rapidly and outcompete naïve B cells, thereby restricting the response to the conserved epitope. This phenomenon of 'original antigenic sin' was originally posed by Thomas Francis on observations from influenza infections, and subsequently described for influenza vaccinations, where it is dubbed "negative antigenic interaction" (*9*). The data from Kaku *et al.* provide the first indication that original antigenic sin occurs after Omicron infection in that they did not find unique Omicron-specific antibodies (*2*). If this were the same for vaccination with an Omicron variant vaccine, it would mean that this will boost a restricted number of Bmem cells (Fig. 1). Thus, it would restrict the number of unique specificities, even more than boosting with a Wuhan vaccine. Potentially, this restriction would limit the capacity to respond to new variants rather than extend it, which would have been the goal. Clearly more data will be needed, especially longer after infection or vaccination, as these time points would better reveal new specificities derived from the delayed kinetics of the naïve B cell response.

In conclusion, the data from Quandt *et al.* and Kaku *et al.* provide a first glimpse into the Bmem after vaccine boosters and their capacity to respond to Omicron infection (*1*, *2*). The upside is that this provides us with critical information into the immunological basis of vaccine efficacy against the current VoC while also illustrating that the capacity to respond to variants is based on antibody affinity as well as diversity. Improving affinity can be achieved by vaccine boosters; enhancing diversity will not be so straightforward. Responses to new specificities will require engagement of naive B cells and we might need methods to boost their responses relative to those of existing Bmem. Alternatively, there is the potential to alter vaccine formulations to go beyond Spike variants. The B cell response is highly dependent on T-cell help, and it has become clear that T cells recognizing other targets than Spike are important in the response to SARS-CoV-2 infection (*10*). Inclusion of other structural (Nucleocapsid, Membrane protein) and nonstructural targets (e.g., nsp3, nsp4, ORF3a, and ORF8) would strengthen T-cell responses, but also diversify the Bmem and antibody responses. While these targets do not induce neutralizing responses, these are less prone to mutation and will generate Bmem that are relevant against future variants, supporting other aspects of the immune response.

Although the challenges created by SARS-CoV-2 variants are urgent, these problems also provide unique opportunities to address long-standing paradigms, unknowns and challenges in the field. Let's seize them!

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Acknowledgments: I would like to thank Profs. Robyn O'Hehir and David Tarlinton and Dr. Emily Edwards for their continuous support and critical feedback.
Funding: MCvZ is supported by a National Health and Medical Research Council (NHMRC) Ideas grant (1185294) and an unrestricted grant from BD Biosciences.
Competing interests: MCvZ is an inventor on a patent application related to generation of fluorescently labeled antigen tetramers used for flow cytometric analysis of antigen-specific B cells.

Published First Release 2 June 2022
10.1126/sciimmunol.abq5901

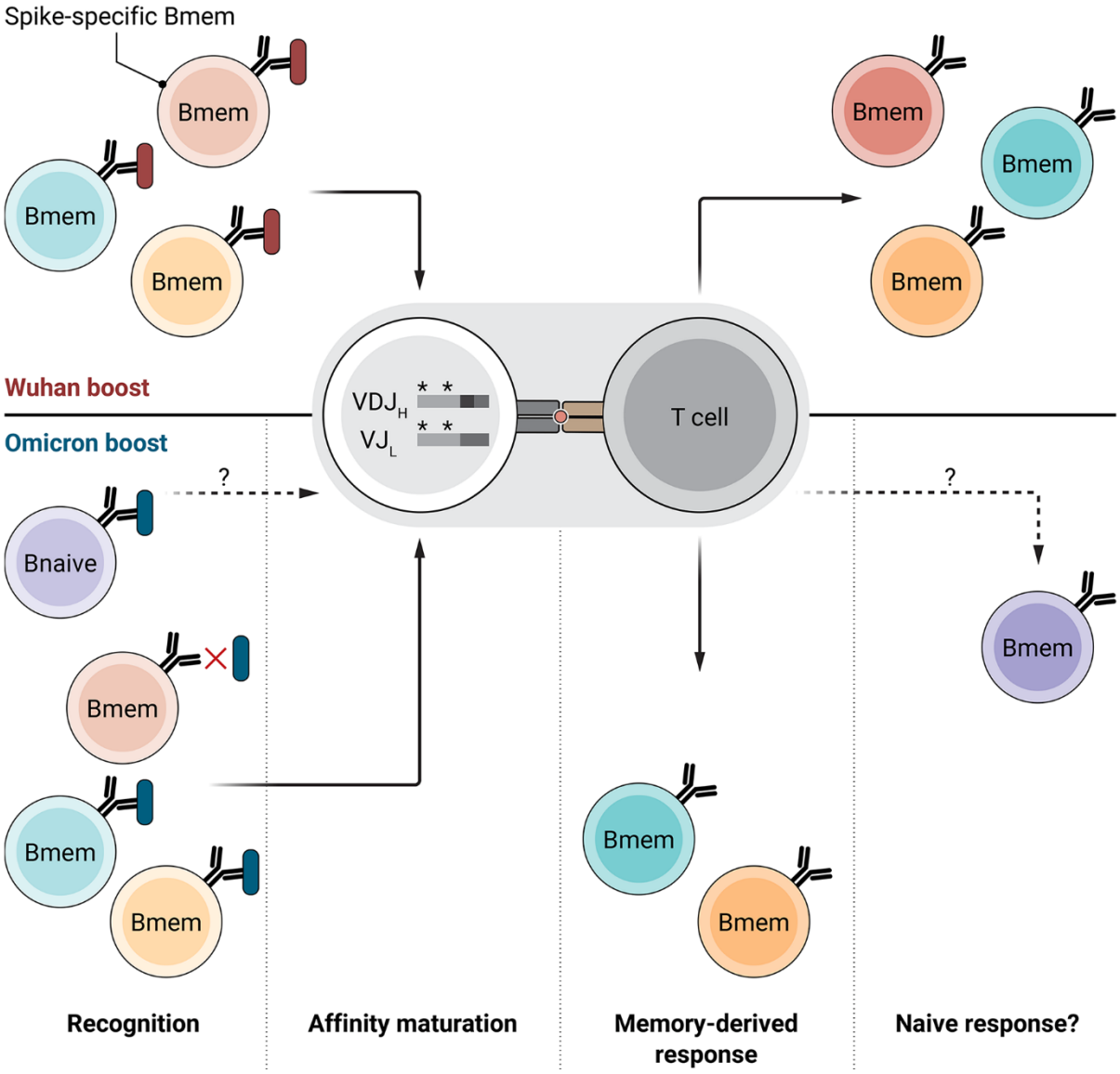

Fig. 1. Model for Bmem responses to Wuhan vs variant SARS-CoV-2 (Omicron). Top. Booster vaccination with Wuhan Spike has the potential to include all previously-formed Bmem, which undergo further affinity maturation through somatic hypermutations in their Ig genes in the germinal center. Bottom. Omicron infection in previously vaccinated individuals reactivates those Bmem that can still recognize the variant despite many mutations. Activation of naive B cells with new specificities is needed to prevent reduction of Bmem diversity. CREDIT: A. MASTIN/*SCIENCE IMMUNOLOGY*

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Sci. Immunol., **Ahead of Print** • DOI: 10.1126/sciimmunol.abq5901