REPLY TO PANDEY:

Possible functional impact of IgG3 allotype constant region

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In a letter to the editor (1), Janardan Pandey comments on our recent study demonstrating that IgG3 switch variants of a severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) specific antibody (Ab) are superior to other IgG subclasses in binding to the SARS-COV-2 spike protein (SP) in virus neutralization activity (2).

Understanding the effect of individual IgG subclasses to modulate antigen (Ag) binding and activity of IgG Abs is an essential basis for interpreting polyclonal immune responses that occur in vivo. For example, strong IgG3 and IgG4 responses were identified in the sera of COVID-19 patients, which need to be interpreted with respect to their protective potential. In the long term, knowledge about IgG subclass activity may help in designing more informed vaccination strategies boosting select IgG subclasses against prevalent and emerging viral diseases.

Pandey (1) correctly points out that, even within each IgG subclass, a great heterogeneity exists within the human population. With 13 allotypes, IgG3 is indeed the most polymorphic IgG subclass, and our current study focuses on only one of these variants, GM5. He further suggests that the amino acid alteration in the Fc portion of IgG3-GM5 may impact Ag binding and hence extend its effects to the variable domain. Indeed, our study shows that the IgG3 switch variant has an approximately fivefold higher affinity for the SARS-CoV-2 SP, compared to the other IgG subtypes, which may explain, at least in part, the increased neutralizing activity. Further along these lines, we fully agree with Pandey that there may be other IgG3 allotypes that show an even stronger effect on Ag binding compared to GM5. We would even go beyond his statements and stress that in vivo factors such as binding to Fcγ receptors or FcRn may play a critical role for IgG3 allotype activity (3). Moreover, Ab modifications of a more subtle nature, like posttranslational modifications, may also impact functional activities of IgG3 allotypes. In this context, several studies point to a strong Ag-binding effect of, for example, IgG1 tyrosine sulfation (4), and single sugar residues may even reverse activities, from proinflammatory to antiinflammatory (5). Notably, IgG3 is the only IgG subtype that carries, next to conserved Fc *N*-glycans, a heavily O-glycosylated hinge region, in close proximity to the Ag-binding domain.

Thus, it would indeed be very interesting to perform in-depth studies of all 13 known IgG3 allotypes carrying identical variable domains to elucidate the impact of Fc modifications not only on Fc-mediated activities but also on Ag binding. The plant expression system as described by Kallolimath et al. (2, 6) provides high flexibility and speed in recombinant Ab expression and is certainly well suited to pursue such an approach.

Acknowledgments

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