



Commentary

A (class-)switch in the antibody response may distinguish primary from secondary dengue virus infection

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The four circulating serotypes of dengue virus (DENV1-4) are mosquito-borne flaviviruses that cause 390 million human infections annually [1]. Approximately 25% of these infections result in symptoms ranging from a mild fever to a potentially fatal disease characterized by hemorrhagic fever or shock syndrome [1]. Although antibodies are a critical component for flavivirus immunity, complex antibody responses to DENV1-4 hinder the design of effective vaccines. Epidemiological studies have established that intermediate levels of antibodies from a prior DENV infection with one serotype are associated with the development of severe disease following subsequent infection with a different DENV serotype [2]. The prevailing theory suggests that during secondary exposure, pre-existing cross-reactive antibodies of the IgG isotype can bind to DENV particles from a different serotype without neutralizing infectivity. Instead, these antibodies can promote viral uptake into target cells expressing Fc gamma receptors in a process called antibody-dependent enhancement (ADE). Understanding the components and functions of the antibody response to DENV is important for informing the design of safe and effective antibody-based vaccines and therapies.

Unlike previous work, which was mostly restricted to analysis of B cells expressing the IgG antibody isotype, in the April 2020 issue of *EBioMedicine*, Waickman, Gromowski, et al. used single-cell RNA sequencing to obtain a more unbiased profile of the overall B cell repertoire of six individuals who had experienced primary or secondary DENV infection [3]. The authors examined paired heavy- and light-chain antibody sequences from over 9000 B cells, including short-lived plasmablasts generated early after infection, as well as memory B cells that persist long after the infection has resolved. Among memory B cells, there were no appreciable differences in isotype distribution in primary versus secondary infection. Consistent with a previous study, there was a low prevalence of IgA- relative to IgG-expressing plasmablasts upon secondary infection [4]. In contrast,

following primary infection, this new study found an unexpectedly high proportion of plasmablasts expressing IgA antibodies, many of which were extensively hypermutated, suggesting a recall response despite no known prior DENV exposure. Further studies are warranted to investigate the origin of these hypermutated IgA plasmablasts detected following primary infection.

While IgA antibodies have recently been shown to contribute to the overall serum neutralizing activity against HIV [5], the functional significance of IgA antibodies in the context of DENV infection remains to be determined. Waickmann, Gromowski, et al. posited that plasmablast-derived DENV-specific IgA antibodies may be protective: as they appeared to recognize epitopes commonly targeted by IgG antibodies, by virtue of their inability to bind to Fc gamma receptors on relevant target cells, IgA antibodies may compete for binding to DENV with their IgG counterparts to abrogate the Fc gamma receptor-mediated ADE pathway. However, it is difficult to reconcile this proposed protective mechanism given the fact that except in the case of infants born to DENV-immune mothers [6], ADE is mostly implicated following secondary infection [2], during which IgA antibodies are apparently less prevalent. Indeed, given the reported characteristics of DENV-specific IgA antibodies in the current study, including 1) abundance during primary infection, 2) overlap in epitope specificity with IgG, and 3) limited neutralizing capacity (at least when tested with IgG Fc), it is equally possible that IgA antibodies could inhibit IgG-mediated neutralization of DENV. Additional studies to deconvolute the contribution of different antibody isotypes [5] in the humoral response to DENV infection or vaccination will help define their functional significance.

A limitation of the study by Waickman, Gromowski, et al. is its restricted sample size and population: six pediatric patients of whom five were infected with DENV1. Studies with a larger sample size that includes multiple age groups infected with other DENV serotypes will be needed to confirm the findings reported here and to ultimately draw correlations with disease outcomes. Nevertheless, this study highlights the utility of single-cell RNA sequencing to efficiently profile a large number of B cells in an unbiased manner, capturing diverse antibody isotypes and cellular states. Other recent studies further demonstrated this technology's capacity to link B cell receptor sequences [7] or transcriptional profiles [10] to antigen specificity. In the future, it would be interesting to investigate whether particular B cell transcriptomic signatures can predict antibody functions such as direct neutralization and Fc-dependent effector mechanisms. In addition to single-cell genomics, a 'systems serology' approach [8] to probe

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the biochemical and biophysical modifications to antibodies will also be important, given the emerging role of Fc glycoforms in regulating dengue disease [9]. Rapid advances in profiling humoral immunity at high throughput and resolution hold promise for comprehensively identifying the correlates of antibody-mediated protection and pathogenesis in DENV and other infections.

Declaration of Competing Interests

Authors have no conflicts of interest to disclose.

References

- [1] Bhatt S, Gething PW, Brady OJ, Messina JP, Farlow AW, Moyes CL, et al. The global distribution and burden of dengue. *Nature* 2013;496:504–7 <https://doi.org/10.1038/nature12060>.
- [2] Katzelnick LC, Gresh L, Halloran ME, Mercado JC, Kuan G, Gordon A, et al. Antibody-dependent enhancement of severe dengue disease in humans. *Science* 2017;358:929–32 <https://doi.org/10.1126/science.aan6836>.
- [3] Waickman AT, Gromowski GD, Rutvisuttinunt W, Li T, Siegfried H, Victor K, et al. Transcriptional and clonal characterization of B cell plasmablast diversity following primary and secondary natural DENV infection. *EBioMedicine* 2020;54:102733 <https://doi.org/10.1016/j.ebiom.2020.102733>.
- [4] Wrarmert J, Onlamoon N, Akondy RS, Perng GC, Polsrila K, Chandele A, et al. Rapid and massive virus-specific plasmablast responses during acute dengue virus infection in humans. *J Virol* 2012;86:2911–8 <https://doi.org/10.1128/JVI.06075-11>.
- [5] Jia M, Liberatore RA, Guo Y, Chan K-W, Pan R, Lu H, et al. VSV-displayed HIV-1 envelope identifies broadly neutralizing antibodies class-switched to IgG and IgA. *Cell Host Microbe* 2020;0 <https://doi.org/10.1016/j.chom.2020.03.024>.
- [6] Kliks SC, Nimmanitya S, Nisalak A, Burke DS. Evidence that maternal dengue antibodies are important in the development of dengue hemorrhagic fever in infants. *Am J Trop Med Hyg* 1988;38:411–9 <https://doi.org/10.4269/ajtmh.1988.38.411>.
- [7] Setliff I, Shiakolas AR, Pilewski KA, Murji AA, Mapengo RE, Janowska K, et al. High-throughput mapping of B cell receptor sequences to antigen specificity. *Cell* 2019;179:1636–46 e15 <https://doi.org/10.1016/j.cell.2019.11.003>.
- [8] Arnold KB, Chung AW. Prospects from systems serology research. *Immunology* 2018;153:279–89 <https://doi.org/10.1111/imm.12861>.
- [9] Wang TT, Sewatanon J, Memoli MJ, Wrarmert J, Bournazos S, Bhaumik SK, et al. IgG antibodies to dengue enhanced for FcγRIIIA binding determine disease severity. *Science* 2017;355:395–8 <https://doi.org/10.1126/science.aai8128>.
- [10] Neu KE, Guthmiller JJ, Huang M, La J, Vieira MC, Kim K, et al. Spec-seq unveils transcriptional subpopulations of antibody-secreting cells following influenza vaccination. *Journal of Clinical Investigation* 2019;129(1):93–105. doi: [10.1172/JCI121341](https://doi.org/10.1172/JCI121341).