LETTER

Consequences of NMO-IgG binding to aquaporin-4 in neuromyelitis optica

Hinson et al. (1) suggested a unique mechanism for the pathogenesis of neuromyelitis optica (NMO), a neuroinflammatory disease caused by binding of an autoantibody (NMO-IgG) to aquaporin-4 (AQP4) on astrocyte plasma membranes. AQP4 is expressed as long (called M1) and short (called M23) isoforms, the latter forming orthogonal arrays of particles (OAPs). Hinson et al. (1) reported that M1-AQP4 is rapidly internalized in cells whereas M23-AQP4 resists internalization, and that NMO-IgG inhibits AQP4 water permeability. They propose that NMO pathogenesis involves NMO-IgG–induced internalization of M1-AQP4 and direct inhibition of AQP4 water permeability, resulting in increased OAP size, enhanced complement-dependent cytotoxicity, and tissue swelling. Although their mechanism is unique and interesting, data from our laboratory and others challenge each of their major findings and hence the validity of their model.

Differential internalization of M1-AQP4 and M23-AQP4 following NMO-IgG exposure is quite unexpected, as M1-AQP4 and M23-AQP4 form tight heterotetramers in cell plasma membranes. Differential internalization conflicts with published data from the same group showing full internalization of AQP4 (2), and from our laboratory showing rapid and comparable internalization of M1-AQP4 and M23-AQP4 in transfected cells (3, 4). We found that relative resistance of complement-dependent cytotoxicity in cells expressing M1-AQP4 vs. M23-AQP4 is not caused by differential internalization, but instead by enhanced C1q binding to clustered NMO-IgG on OAPs (4). A major technical problem with the study of Hinson et al. (1) is the use of N-terminus GFP-labeled AQP4, as the GFP label prevents OAP formation. We recently reexamined the differential internalization issue in multiple cell types expressing M1-AQP4 and M23-AQP4, and did not find preferential M1-AQP4 internalization following NMO-IgG exposure. Also, OAP size did not change following NMO-IgG application by superresolution imaging of mouse astrocytes using stochastic reconstruction microscopy (five NMO sera studied).

With regard to NMO-IgG inhibition of AQP4 water permeability, it is unlikely that significant inhibition is sterically possible in view of the large size of NMO-IgG compared with AQP4 tetramers, which contain four separate water pores. The time-to-lysis assay of oocyte osmosis Hinson et al. (1) reported is an inaccurate surrogate of water permeability. We measured water permeability by stopped-flow light scattering on plasma membrane vesicles isolated from AQP4-expressing CHO cells, an approach that can detect changes in water permeability as small as 5% and is not confounded by internalization effects. Exposure of AQP4 to high concentrations of NMO-IgG from six different seropositive NMO patients did not reduce M1-AQP4 or M23-AQP4 water permeability. Another published report also concluded that NMO-IgG does not inhibit AQP4 water permeability (5).

Last, Hinson et al. (1) argued that punctate AQP4, cell edema, and intramyelinic edema occur in human NMO when NMO-IgG binds AQP4 without complement. This contradicts data in mice, in which such changes occur only when complement is present. Punctate AQP4 is a nonspecific finding seen in damaged astrocytes, as are cellular and intramyelinic edema, which are seen in other CNS diseases, such as Alexander disease, in which NMO-IgG is absent. The mechanisms linking NMO-IgG binding to AQP4 to NMO pathology thus require further investigation.

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