The role of complement activation in IgM M-protein associated neuropathies

Authors

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Background
Polyneuropathy associated with an immunoglobin M (IgM) monoclonal gammopathy is characterized by slowly progressive, predominantly distal sensorimotor deficits, sensory ataxia, and electrophysiological features of demyelination. IgM antibodies against myelin-associated glycoprotein (MAG) are present in serum from the majority of patients. Nerve damage most likely results from the concerted action of binding of anti-MAG antibodies to nerves followed by complement activation. The interaction of anti-MAG antibodies with complement activation and their relation to clinical neurological characteristics have not been studied in detail.

Methods
We used serum samples from 101 patients who were previously included in the IMAGiNe study, a prospective observational cohort study of patients with polyneuropathy and an IgM M-protein (both IgM MGUS and Waldenström's Macroglobulinemia). We analyzed patient sera to assess IgM anti-MAG titers by Bühlmann enzyme-linked immunosorbent assay (ELISA) and antibody-mediated complement activation using both an in-house ELISA-based system and an in-house cell-based system of primate peripheral nerve slides.

In both systems, we used patient sera to opsonize and added IgG/IgM-depleted serum as a complement source. For each opsonization sample, a negative EDTA control was included to correct for background complement activation. We measured the level of complement activation by labelling C3 and assessing optical density (OD) under the microscope. Complement activation was depicted as ratio between the OD_{C3} and the OD_{EDTA} as $ratio_{C3/EDTA}$. We studied correlations of complement activation with anti-MAG ELISA titers and clinical neurological characteristics.

Results
Patients (75.2% men) had a median age of 69 years at the moment of serum sampling, and harbored an IgM-kappa M-protein in the majority of cases (74.4%), followed by IgM-lambda (16.7%) or both IgM-kappa and -lambda (9.9%). Total IgM ranged from 0.65 g/L to 23 g/L, with a median of 3.9 g/L. Anti-MAG antibodies were present in 62.4% of patients (titer ≥10,000 Bühlmann titer units), anti-ganglioside antibodies (GM1, GM2, GQ1b or GD1a) were present in 8.2% of patients.

IgM anti-MAG titers varied from negative to strongly positive (up to 2,303,333 Bühlmann titer units). Complement activation in the ELISA-based system correlated significantly with anti-MAG antibody titer (Spearman's rho 0.80; $p < 0.0001$) despite large variability between serum samples with comparable anti-MAG titers (figure 1). This variability was even larger in the cell-based assay, which also showed complement activation in IgM anti-MAG negative patients (figure 2), and indicates the presence of complement activating autoantibodies directed against epitopes other than MAG in a subset of patients with IgM associated polyneuropathy. Similarly, some anti-MAG positive patients did not show an elevated complement activation level, suggesting differing complement activating properties between patients.

Neurological characteristics did not correlate with anti-MAG titers or complement activation. This may be caused by a lack of sufficiently sensitive clinimetrics, but could also reflect a lack of direct influence of anti-MAG titer or complement activation on neurological characteristics.

Conclusions
Anti-MAG antibody titers correlate with the level of complement activation. Neurological characteristics of IgM associated polyneuropathy do not correlate with titers or the level of complement deposition.

Figures

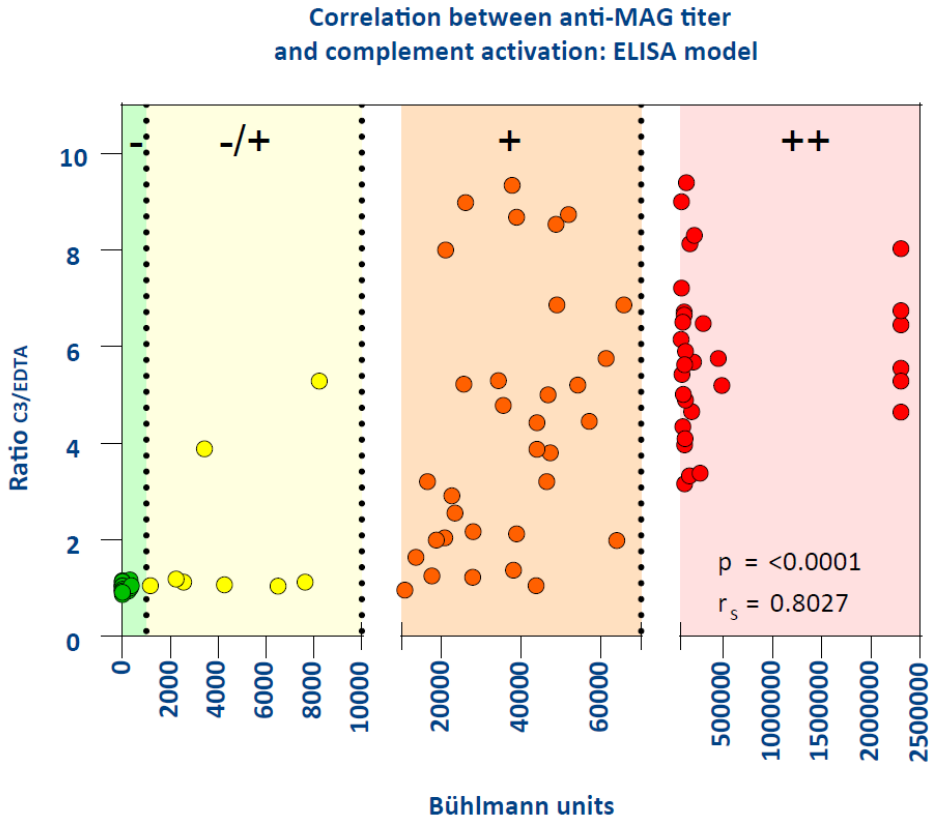

Figure 1: correlation between anti-MAG titer and complement activation (ELISA model)

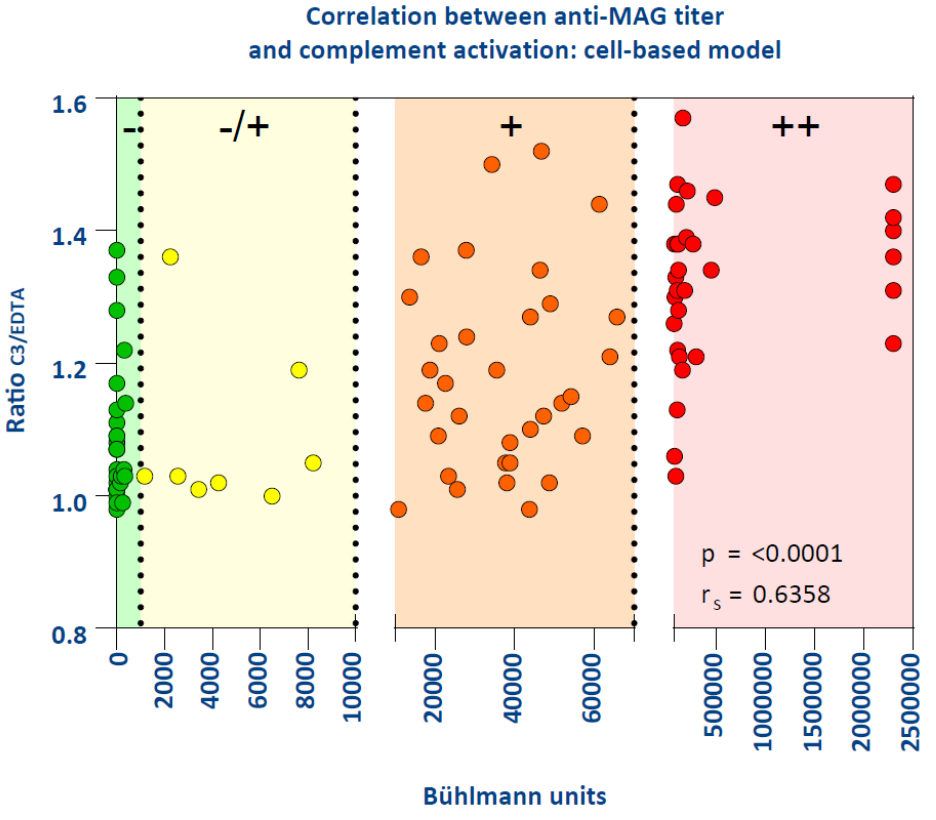

Figure 2: correlation between anti-MAG titer and complement activation (cell-based model)