IgG4-associated complement activation in membranous nephropathy—A Fab phenomenon?

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Abstract

Introduction
Immunoglobulin G4-related diseases encompass a growing list of organ involvement including membranous nephropathy, a disease entity generally associated with circulating antibodies to glomerular antigens and localised activation of complements. Although both immunoglobulin G4 and complements were present in the renal biopsy specimens, the mechanistic association link between them remains uncertain, particularly because immunoglobulin G4 is known to inhibit complement activation. The solution to this conundrum may lie in the recently discovered Fab arm exchange phenomenon.

Hypothesis
In this article, we hypothesise that immunoglobulin G4 molecules undergo structural perturbations via the dynamic Fab arm exchange phenomenon, which might then render it capable of complement activation and hence have a direct role in the pathogenesis of immunoglobulin G4-related membranous nephropathy.

Evaluation of Hypothesis
Investigation of the hypothesis is based on recent discovery of newer biological properties of IgG4.

Conclusion
Membranous nephropathy may be just another endproduct of the newly discovered proinflammatory properties of IgG4.

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Introduction
One hundred and fifty years after the discovery of immunoglobulins (Ig), their role in disease conditions continues to evolve. Particular focus of interest is the IgGs whose subclasses IgG1, IgG2, IgG3 and IgG4 were discovered in 1950s and comprise 71%, 18%, 8% and 3% of serum IgG concentration, respectively. Recent discovery of the association between IgG4 and autoimmune pancreatitis and the subsequent report of IgG4-associated idiopathic tubulointerstitial nephritis (IgG4-TIN) have renewed interest on the role that IgG4 plays in renal diseases. Distinctive pathological features of IgG4-TIN have been described, including elevated serum IgG4 concentrations, plasma cell-rich tubulointerstitial nephritis with >10 IgG4+ plasma cells/ per high power field in the most concentrated field, tubular basement membrane immune complex deposits by immunofluorescence (IF), immunohistochemistry, and/or electron microscopy. The latest twist is the association of IgG4 with membranous nephropathy (IgG-MN), a disease entity that is generally associated with circulating antibodies to glomerular antigens and local complement activation by subepithelial immune deposit. Although circumstantial evidences suggest that IgG4 could have a pathogenetic role in MN, the inability of IgG4 to activate complement raises questions about the relevance of IgG4 in the pathogenesis of MN.

Hypothesis
We hypothesise that IgG4 molecules undergo structural perturbations via the dynamic Fab arm exchange phenomenon, which then renders it capable of complement activation and hence a direct role in the pathogenesis of IgG4-related MN.

Evaluation of Hypothesis
Evaluation of the hypothesis is based on a review of renal pathology findings and their interpretation in light of recent knowledge of the physicochemical and biological properties of IgG4. MN is characterized by glomerular subepithelial immune deposits that locally activate complement to cause podocyte injury. M-type phospholipase A2 receptor protein (PLA2R1) expressed on the surface of native glomerular podocytes appear to be the dominant autoantigen, and antibodies to PLA2R1 are found in about 80% of patients with primary MN. Majority of the antibodies are of the IgG4 subclass, and titres of IgG4, but not other subtypes, correlate with baseline proteinuria and remission, suggesting a pathogenetic role of these antibodies. Most of the patients with IgG4-related renal disease have low serum complement levels. Moreover, in renal biopsies of patients with IgG4-TIN, IgG4 deposits (as well as IgG1, IgG2 and IgG3) in the interstitium, Bowman capsule and tubular basement membranes were associated with deposits of C3. In patients with IgG4-MN, IF staining for IgG4 and C3 in the glomerular basement membrane were common findings. In multiple animal models of MN, depletion of complement factors mitigated proteinuria and normalised tubular function. Although these circumstantial evidences suggest a role for complement activation in the formation of immune deposits in IgG4-MN, others have shown that IgG4 does not cause complement activation. Resolving this apparent
Nephropathy—a Fab phenomenon?

Discussion

Although the above evidences support the participation of IgG4 in the activation of the complement pathway, its relative contribution in the co-presence of IgG1 and IgG3 remains to be elucidated. Human IgG subclasses have differing abilities to activate complements, decreasing in the order: IgG3>IgG1>IgG2>IgG4.21,22 These differences in binding capability to C1q and FcR are due to differences in their amino acid residues and inter-heavy chain disulphide bridges in the hinge region of IgG subclasses that restrict antigen-binding fragments Fab-Fab and Fab-Fc flexibility that triggers effector functions, such as complement activation and Fc-receptor binding. IgG4 has a short hinge region and displays only limited flexibility as compared with the other IgG subclasses. IgG4 does have a unique property known as the Fab arm exchange phenomenon, whereby a heavy chain and attached light chain (half-molecule) are exchanged with a heavy–light chain pair from another molecule resulting in bispecific antibodies.23 These functionally monovalent antibodies are unable to cross-link antigen and have less predilection to form immune complexes.22,24 How does then IgG4 activate the complement system? A recent review by Karsten and Kohl provides valuable insights into structural features of Iggs that facilitate C1q binding.23

Accordingly, IgG4 hinge regions may potentially attain greater flexibility through dynamic Fab arm exchange via the alternate and lectin pathways. The intensity of C1q deposits was higher in segmental MN vs. global MN, while the intensity of factor B and MBL was higher in global vs. segmental MN. Thus, it was suggested that IgG1 and IgG3 are associated with complement activation via the classical pathway; whereas IgG2 and IgG4 are associated with complement activation via alternate and lectin pathways.

Conclusion

There is emerging clinical and pathological evidence of a role of IgG4 in complement activation in MN. Evidence suggests that the structural basis for differences among IgG subclasses with respect to effector functions appears to be located within the C2 domain of the Ig molecule. Moreover, amino acid composition of the C2 domain seems to be responsible for isotype variation. Replacing the C2 portion of human IgG4 with that from IgG1 resulted in an IgG mutant that was capable of activating complements as potently as the IgG1 molecule.25 There is also evidence that amino acids (Ser313Pro replacement) outside the conserved C1q binding sites may also be involved in complement activation. Furthermore, an important factor in complement activation appears to be N-glycosylation of IgG molecules in the C2 domain at Asn297.26 The N-glycans within the C2 domain required for alternate complement activation. The N-glycans are heterogeneous and contain variable terminal sugar residues, which can define whether MBLs (alternate pathway) can recognize Iggs through their polysaccharide-recognition domains. When the terminal sugar is N-acetyl-glucosamine, MBL can bind to IgG and activate complements via the lectin (alternate) pathway; when it is galactose, C1q binding (classical) can occur; mannose causes abrogation of C1q binding while agalactosylation results in reduced or no complement activation. Therefore, the dynamic Fab arm exchange can cause structural alterations to the IgG4 molecule enabling it to fine-tune its response at C2 glycan level by activating either the classical or alternate pathways.
exchange allows IgG4 to gain a half-molecule with a heavy and a light-chain, comprising a C3,2 region with a heterogeneous N-glycan structure with differing terminal sugar moieties that can lead to aggregation or activation of complements via the classical or alternate pathways. MN may be just another endproduct of the newly discovered proinflammatory properties of IgG4. Clearly, much work lies ahead.

**Abbreviations list**

IF, immunofluorescence; Ig, immunoglobulin; IgG-MN, IgG4 with membranous nephropathy; IgG4-TIN, IgG4-associated idiopathic tubulointerstitial nephritis; MBL, mannose-binding lectin ligand; MN, membranous nephropathy; PLAZR1, phospholipase A2 receptor protein.

**References**