Published in final edited form as:

Nat Rev Nephrol. 2015 November ; 11(11): 633–634. doi:10.1038/nrneph.2015.149.

The glomerulus reveals some secrets

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Abstract

In the past decade, major advances have been made in defining the antigens and pathogenesis of immune complex diseases such as membranous nephropathy and IgA nephropathy. Probing of rare genetic diseases has revealed new pathways of injury in proteinuric conditions, including channel abnormalities in the podocyte and complement dysregulation underlying proliferative glomerular lesions.

Novel approaches to the assessment of human renal biopsy samples have led to new insights into several common glomerular diseases. In primary membranous nephropathy (MN), the most common target antigen, phospholipase A2 receptor (PLA2R), has been identified.^{1,2} Moreover, mutations in transient receptor cation potential channel 6 (TRPC6) were discovered to cause familial focal segmental glomerulosclerosis (FSGS),³ expanding the paradigm beyond previous data showing that genetic forms of this disease are caused by mutations in structural proteins of the podocyte. The markedly increased incidence of some chronic kidney diseases (CKDs) in African Americans was linked to risk allele variants of *APOL1*, which encodes apolipoprotein L1.⁴ The pathogenesis of IgA nephropathy (IgAN) was elucidated, implicating antibody binding to an abnormally glycosylated region of IgA-1—the main IgA subtype present in the circulation.⁵ An unbiased classification approach to the varying biopsy appearances of IgAN has become a model for exploring the significance of diverse patterns of injury.⁶ Aberrations in complement C3 regulation are now recognized to underlie many proliferative glomerular injuries.^{7,8} Rapidly expanding translational research is aimed at directing these new insights into the molecular pathogenesis of disease towards individualized therapies.

A major goal in nephrology has been to identify the target antigen in primary MN. Animal models were developed but the antigens (for example megalin in Heymann nephritis) were not present in human MN. Using an elegant and laborious approach, Beck *et al.* collected normal glomeruli from non-transplanted deceased donor kidneys and screened protein extracts with serum samples from patients with primary or secondary MN.¹ They identified the antigen PLA2R, which is normally expressed on podocytes, using additional techniques, including mass spectrometry and elution of IgG from the deposits. About 70% of patients with clinically defined primary MN had deposits containing PLA2R and circulating antibodies to this receptor. Importantly, patients with MN secondary to lupus erythematosus

Competing interests

The author declares no competing interests.

did not show PLA2R positivity, and only a very minor proportion of patients with cancer-associated MN had PLA2R-positive deposits or serum PLA2R antibodies. Using a similar approach, another antigen, thrombospondin type-1 domain-containing 7A, was subsequently identified in approximately 10% of PLA2R-negative patients with primary MN.² These studies indicate that extensive screening and mass spectrometry of large numbers of samples will likely identify additional pathogenic antigens in patients without a clinically evident secondary cause of MN. Studies are currently underway to define the utility of assessing serum levels versus biopsy demonstration of PLA2R in delineating specific causality and tracking responses to intervention. Understanding inherited (for example HLA-DQ1-associated risks⁹) or acquired (for example drug or infection-associated) mechanisms underlying autoantibody formation will yield additional insights and may provide targets for early detection or even prevention in susceptible populations.

Mass spectrometry was also crucial in clarifying other types of proteins deposited in glomeruli. For example, patients with non-AL, non-AA amyloid can now be correctly classified according to amyloid type, identifying rare familial and other forms of amyloid that require differing therapeutic approaches. This technique has transformed the approach to characterizing unusual deposits within the kidney, including those in dense deposit disease (DDD).¹⁰

Primary FSGS is a common glomerulopathy caused by an as yet not definitively identified circulating factor or factors. However, patients with familial FSGS may offer insights into possible pathogenic pathways that may be perturbed in the more common sporadic FSGS. Most cases of familial FSGS are linked to mutations in structural proteins of the podocyte and slit diaphragms or in components that anchor podocytes to the glomerular basement membrane, such as nephrin, podocin and α -actinin-4. Recently, a missense mutation in the cation channel TRPC6 was discovered in a kindred with familial adult-onset FSGS.³ This mutation alters calcium signalling, increasing the response to agonists such as angiotensin II. It is possible that TRPC6 abnormalities also occur in sporadic FSGS or other proteinuric diseases. Thus, increased TRPC6 activity and resulting high calcium influx may contribute to podocyte injury and play a role in proteinuria and progressive sclerosis. Pharmacologic targeting of a channel is an attractive and attainable goal; manipulating TRPC6 expression and activity thus offers promise as a therapy for numerous proteinuric diseases characterized by podocyte dysfunction and loss.

Genetic approaches have also shed light on non-familial kidney diseases. Compared with Caucasians, African Americans have a 4–5-fold increased risk of some CKDs, including hypertension-associated CKD, HIV-associated nephropathy and FSGS. Genome-wide association studies found linkage of this excess risk to the *APOL1* allele variants G1 and G2.⁴ These variants, unlike the GO variant common in Caucasians, are trypanolytic for *Trypanosoma brucei rhodesiense*, suggesting possible evolutionary pressures underlying their high prevalence in populations of African ancestry. Although the mechanisms for increased disease risk have not been elucidated, ongoing studies postulate an effect on podocytes and/or innate immunity.

"Mass spectrometry ... has transformed the approach to characterizing unusual deposits within the kidney..."

IgAN has a highly variable clinical course and a range of morphological expressions. A working group formed by the International IgA Nephropathy Network and the Renal Pathology Society undertook a novel approach to classification of this disease.⁶ In a non-biased method, all pathological lesions present within a cohort of archival IgAN biopsy samples were defined to determine the variables that could be reproducibly scored. The six pathologic variables thus defined were then assessed for correlation with clinical outcomes. Four variables correlated independently with increasing loss of glomerular filtration rate: mesangial hypercellularity, endocapillary hypercellularity, segmental sclerosis and significant tubular atrophy/interstitial fibrosis (>25%). These variables were used to define the so-called MEST score. Subsequent studies validated and refined this scoring approach, which could form the basis for patient stratification, analysis of therapeutic responses and assessment of progression risk, as well as possibly provide insights into the pathogenesis of various lesions. Similar to the approach commonly used in oncology, patients with similar stage and severity of disease may be grouped together in clinical trials.

The pathogenesis of IgAN has also been elucidated. The hinge region of normal IgA-1 has numerous serine and threonine residues with O-glycosylation. However, in patients with IgAN, abnormally glycosylated IgA-1 with deficient galactose is bound by anti-glycan antibodies, including IgG, which may have a role in complement activation and a subsequent cascade of injury.⁵ This abnormal IgA-1 is not efficiently cleared by receptors in the liver, which may enhance its deposition within the mesangium. These insights provide a basis for investigation of events leading to the generation of glycan-specific antibodies, not only as disease markers, but also as a pathogenic mechanism and therapeutic target.

Membranoproliferative glomerulonephritis (MPGN) is characterized by mesangial and subendothelial deposits, double contours of glomerular basement membranes due to reactions to these deposits, endocapillary hypercellularity and increased matrix. MPGN was previously thought to represent immune complex diseases of various types, designated as MPGN I, MPGN II (now known as DDD) and MPGN III. However, some patients with an MPGN pattern of injury have isolated C3 deposits.⁷ Mass spectrometry studies demonstrated that deposits in DDD contain soluble components of the terminal complement complex. Aberrations in complement regulatory proteins were identified in a group of patients with dominant C3 deposits or C3 deposits only, including heterozygous mutations in factor H and CD46 (key regulators of C3 activation) and expression of an autoantibody that inhibits C3 convertase, C3 nephritic factor.⁷ Subsequent studies in animal models and in patients led to a new paradigm for approaching these C3 dominant glomerulonephritides, now called C3 glomerulopathy.⁸ Patients show a spectrum of lesions from DDD to deposits of complement components that overlap with the ultrastructural appearance of immune complexes. Various genetic or acquired alterations in complement regulation have been recognized, including familial forms with mutations in complement factor H-related protein 5. The therapeutic approach in these patients may be controlling complement dysregulation, rather than searching for underlying lupus, collagen vascular diseases, chronic infections or other triggers of MPGN.

The field of nephrology—and specifically glomerular diseases—is catching up with the state-of-the-art in oncology, in which specific mutation profiles and aberrant signalling pathways form the basis for personalized therapy. We are at the dawn of a new beginning in molecular understanding of the pathogenesis of some of the common diseases afflicting the glomerulus. These key observations form the basis for rapid progress in the development of new insights into the pathogenesis of disease and new targets for intervention.

References

1. Beck LH, Jr et al. M-type phospholipase A2 receptor as target antigen in idiopathic membranous nephropathy. *N. Engl. J. Med* 361, 11–21 (2009). [PubMed: 19571279]
2. Tomas NM et al. Thrombospondin type-1 domain-containing 7A in idiopathic membranous nephropathy. *N. Engl. J. Med* 371, 2277–2287 (2014). [PubMed: 25394321]
3. Winn MP et al. A mutation in the TRPC6 cation channel causes familial focal segmental glomerulosclerosis. *Science* 308, 1801–1804 (2005). [PubMed: 15879175]
4. Genovese G et al. Association of trypanolytic ApoL1 variants with kidney disease in African Americans. *Science* 329, 841–845 (2010). [PubMed: 20647424]
5. Suzuki H et al. Aberrantly glycosylated IgA1 in IgA nephropathy patients is recognized by IgG antibodies with restricted heterogeneity. *J. Clin. Invest* 119, 1668–1677 (2009). [PubMed: 19478457]
6. Working Group of the International IgA Nephropathy Network and the Renal Pathology Society. The Oxford classification of IgA nephropathy: pathology definitions, correlations, and reproducibility. *Kidney Int* 76, 546–556 (2009). [PubMed: 19571790]
7. Servais A et al. Primary glomerulonephritis with isolated C3 deposits: a new entity which shares common genetic risk factors with haemolytic uraemic syndrome. *J. Med. Genet* 44, 193–199 (2007). [PubMed: 17018561]
8. Pickering MC et al. C3 glomerulopathy: consensus report. *Kidney Int* 84, 1079–1089 (2013). [PubMed: 24172683]
9. Stanescu HC et al. Risk HLA-DQA1 and PLA₂R1 alleles in idiopathic membranous nephropathy. *N. Engl. J. Med* 364, 616–626 (2011). [PubMed: 21323541]
10. Sethi S et al. Laser microdissection and mass spectrometry-based proteomics aids the diagnosis and typing of renal amyloidosis. *Kidney Int* 82, 226–234 (2012). [PubMed: 22495291]

Immunofluorescence image showing granular capillary wall staining for PLA2R in primary membranous nephropathy.