Complement in Membranous Nephropathy
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Membranous Nephropathy
- Autoimmune disease
- Common cause of nephrotic syndrome in adults
- Idiopathic
- Secondary
  - Lupus nephritis – Class V
  - Hepatitis B-associated
  - Drugs
  - Cancer-associated

Pathological Features
- Light microscopy
  - Normal → diffuse GBM thickening (spikes)
- Immunofluorescence
  - Granular capillary IgG and complement
- Electron microscopy
  - Subepithelial electron-dense deposits
  - Podocyte foot process effacement, cytoskeletal condensation and apical microvillous transformation

Pathogenesis
- In-situ immune complex formation
- Circulating antibodies
- Target antigen on podocytes
  - Rats – megalin
  - Neonatal MN – alloimmune response to neutral endopeptidase (NEP)
  - Idiopathic and secondary MN – unknown

Experimental Membranous Nephropathy
- Susceptible rat strains immunized with tubular brush border (Fx1A) develop proteinuria after 6-8 weeks and an immune complex GN indistinguishable from human MN
- Rats injected with anti-Fx1A develop passive HN and proteinuria within 5 days
- Subepithelial immune deposits form in situ when circulating antibody binds to an intrinsic glomerular antigen
Role of Complement in Experimental MN

- Passive Heymann nephritis (PHN) in rats
  - Complement depletion prevents proteinuria
  - Proteinuria and sublethal podocyte injury require formation of C5b-9
  - Depletion of C6 prevents proteinuria
  - Urinary excretion of C5b-9 corresponds to complement-mediated proteinuria

- Active Heymann nephritis
  - Only rats with complement deposits become proteinuric
  - Proteinuria depends on neutralization of Crry (C-regulatory protein)

- “Planted” subepithelial antigen
  - C6-deficient rabbits protected

Evidence for a Role for Complement in Human Membranous Nephropathy

- C3 and C5b-9 are detected in most cases of recent onset MN
- C3d (stable product of C3) is present in all cases of MN
- C5b-9 is detectable in the urine of cases of recent onset MN
- Development of proteinuria in neonatal anti-NEP-induced MN is dependent on the deposition of C-fixing IgG1 antibodies

Caveats

- The PVG strain of C6-deficient rats develop proteinuria after induction of PHN
- The predominant antibodies in human MN are non-C-fixing IgG4

Effects of C5b-9 in Cultured Glomerular Epithelial Cells

- Increased intracellular calcium (influx and release from stores)
- Activation of
  - Protein kinases – RTks, PKC, ERK, JNK, p38
  - Phospholipases – PLC, cPLA2
  - Transcription factors – NFkB
  - Enzymes – Cox-2, NADPH oxidase, MMP-9, heparanase
  - Stress pathways – ER stress, Hsp27
  - Growth factors – PDGF-B, HB-EGF, TGF-β
  - CDK inhibitors – p21, p27
- Matrix protein production – type IV collagen, laminin, heparan sulfate proteoglycans
- DNA damage
- Disruption of the actin cytoskeleton and cell-matrix adhesion complexes
Effect of Complement on the Podocyte Slit Diaphragm in Experimental Membranous Nephropathy

- Nephrin, a major podocyte slit-diaphragm protein, is linked to the actin cytoskeleton by CD2AP and anchored in the plasma membrane by podocin
- Slit-diaphragms are dislocated at the onset of proteinuria in PHN
- Complement-dependent podocyte injury in PHN causes nephrin to dissociate from actin, which may explain the dislocation of slit-diaphragms and account for the onset of proteinuria
- The amount of nephrin is reduced, in part due to loss in the urine

Treatment of Idiopathic Membranous Nephropathy

Standard Treatment

- ACEi or ARB, diuretic, statin
- High-risk patients:
  - Pulse steroid alternating with cytotoxic (Ponticelli)
  - Oral cyclophosphamide
  - Cyclosporin
  - Mycophenolate mofetil?

Targeted Treatment

- Anti-CD20 (Rituximab)
- Humanized anti-C5 monoclonal antibody (eculizumab)

Treatment of Idiopathic Membranous Nephropathy with Complement Inhibitor anti-C5 monoclonal antibody (eculizumab)

A multi-center double-blind controlled study of eculizumab was performed in 130 patients with idiopathic MN. While there was no effect on the primary endpoint of proteinuria after 16 weeks of therapy, some patients given up to one-year of this agent in an open-label extension had impressive responses (unpublished).

Treatment of Idiopathic Membranous Nephropathy with Rituximab (anti-CD20)

- Two small case series induced a complete or partial remission of proteinuria in 50-60% of patients.
- There was no way of predicting which patients would respond

Future Directions

- Identify the membranous nephropathy antigen
- Develop a sensitive and specific immunoassay for anti-MN antibodies
- Screen MN patients for circulating MN antibodies
- Select only those with high titers of anti-MN antibodies (active disease) for therapy with complement inhibitors or anti-CD20
Selected references


