ATYPICAL HEMOLYTIC UREMIC SYNDROME AND C3 GLOMERULOPATHY

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OVERVIEW

1. INTRODUCTION
2. RENAL PATHOLOGY
3. CLINICAL PHENOTYPE & ASSESSMENT
4. GENETIC & ACQUIRED DRIVES OF DISEASE
5. TREATMENT
6. RESEARCH RECOMMENDATIONS

Kidney Disease: Improving Global Outcomes
PART 1:
INTRODUCTION
Complement Activation

Alternative Pathway

Classical Pathway

Lectin Pathway

C3b

C3bB

C3bBb

C5a

C3a

Inflammation anaphylatoxins

Opsonization

Cell Lysis

MAC
ATYPICAL HEMOLYTIC UREMIC SYNDROME (aHUS)

• Ultra-rare disease (UK- 0.42/million population/yr)
  – acute kidney injury
  – thrombocytopenia
  – microangiopathic hemolytic anemia.
• At least 50% of aHUS patients have an underlying inherited and/or acquired complement abnormality.
• Historically prognosis poor- most rapid ESRF
• Eculizumab, a humanized mAb against C5 changed natural history of disease
C3 GLOMERULOPATHY (C3G)

- C3G ultra rare (1/million population/yr)
- C3G comprises a group of kidney diseases driven by uncontrolled activation of the complement cascade that leads to C3 deposition within the glomerulus.
- The dysregulation of C3 convertase is driven by genetic and/or acquired defects.
- A biopsy is required to make the diagnosis.
- Two major subtypes dense deposit disease (DDD) and C3 glomerulonephritis (C3GN).
PART 2:
RENAL PATHOLOGY
aHUS PATHOLOGY

- aHUS is a thrombotic microangiopathy (TMA).
- Pathology - tissue response to endothelial injury.
- Overt thrombosis not always seen
  - Suggested microangiopathy +/- thrombosis
- In general, it is not possible to determine etiology from morphology.
- The presence of C5b-9 staining is not a reliable indicator of aHUS.
# Morphological Features in Microangiopathy

## Active Lesions

### Glomeruli
- Thrombi
- Endothelial swelling or denudation
- Fragmented red blood cells
- Subendothelial flocculent material by EM
- Mesangiolysis
- Microaneurysms

### Arterioles
- Thrombi
- Endothelial swelling or denudation
- Intramural fibrin
- Fragmented red blood cells
- Intimal swelling
- Myocyte necrosis

### Arteries
- Thrombi
- Myxoid intimal swelling
- Intramural fibrin
- Fragmented red blood cells

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**Thrombi**

**Fibrin**

**Microaneurysms**

**Bloodless / fragments**

**Flocculent material**

**Mesangiolysis**
**MORPHOLOGICAL FEATURES IN MICROANGIOPATHY**

<table>
<thead>
<tr>
<th>Chronic Lesions</th>
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<tbody>
<tr>
<td><strong>Glomeruli</strong></td>
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<tr>
<td>• Double contours of peripheral capillary walls by LM, with variable mesangial interposition</td>
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<tr>
<td>• New subendothelial basement membrane by EM</td>
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<tr>
<td>• Widening of the subendothelial zone by EM</td>
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<tr>
<td><strong>Arterioles</strong></td>
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<tr>
<td>• Hyaline deposits</td>
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<tr>
<td><strong>Arteries</strong></td>
</tr>
<tr>
<td>• Fibrous intimal thickening with concentric lamination (onion skin)</td>
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</table>

- Kidney Disease: Improving Global Outcomes
**C3G Pathologies**

- The C3G disease spectrum is caused by abnormal control of complement activation, deposition or degradation.

- Light microscopy - diverse.

- Predominant glomerular C3 fragment deposition on IF:
  - (x2 greater than other immunoreactants IgG, IgM, IgA C1q)
  - 90% DDD, less C3GN.

- Electron microscopy (EM) is used to sub-classify C3G as DDD or C3GN.

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**C3 & Immunoglobulin deposition**

**Dominant C3 Deposition**
MORPHOLOGICAL FEATURES OF C3G

**Light Microscopy**

*Active lesions*
- Mesangial expansion with or without hypercellularity
- Endocapillary hypercellularity including monocytes and/or neutrophils
- Capillary wall thickening with double contours (the combination of capillary wall thickening and mesangial increase is referred to as a membranoproliferative pattern)
- Necrosis
- Cellular/fibrocellular crescents

*Chronic lesions*
- Segmental or global glomerulosclerosis
- Fibrous crescents

**Immunofluorescence Microscopy**
- Typically dominant C3 staining

**Electron Microscopy**
- DDD: Dense osmiophilic mesangial and intramembranous electron dense deposits
- C3GN: Amorphous mesangial with or without capillary wall deposits including subendothelial, intramembranous and subepithelial electron dense deposits
- Sub-epithelial ‘humps’ may be seen in both DDD and C3GN (not pathognomonic of post infectious GN)
C3G Pathology: Controversies

- Correlations between renal biopsy appearances, etiology and clinical outcome are ill-defined.

- IF staining is subjective and semiquantitative.
  - Well-defined for dense deposit disease (DDD).
  - Not clear if characteristic for C3 glomerulonephritis (C3GN).

- Role of C4d staining in distinguishing C3G from IC MPGN requires further investigation

- Pronase digestion should be considered in all cases
  - Masked monotype Ig deposits
PART 3: CLINICAL PHENOTYPE AND ASSESSMENT
• Disease penetrance for an acute episode of aHUS in carriers of known pathogenic mutations increases with age.
Current classifications of aHUS reflect a better understanding of disease mechanisms, including the impact of genetic background and etiologic triggers.

Triggers e.g. pregnancy, infections.
Complement Activation

Thrombotic Microangiopathies

Complement Genetic or Acquired Predisposition

~30%
~70%

Multiple associations
Unmasking latent complement defect
Or
Direct effect

KDIGO
aHUS

• Eculizumab has changed the natural history of disease
  – Previously most rapidly progressed to ESRF
  – Prompt presentation usually leads to prompt resolution
  – Unclear what will happen on Ecu removal

• The time course and persistence of an aHUS episode are not well understood.
  – i.e. acute vs chronic disease
  – Some, but not all, patients may be at life-long risk for recurrent acute presentation.
C3G

• C3G generally follows a chronic, indolent course with persistent AP activation

• 10-year renal survival of approximately 50%.

• There are, however, cases of C3G that present as a rapidly progressive GN.
EXTRARENAL MANIFESTATIONS

aHUS

- Extrarenal manifestations are reported in up to 20% of patients.
- It is unclear whether these manifestations are a direct consequence of complement activation, TMA, or other factors such as severe hypertension and uremia.

C3G

- Acquired partial lipodystrophy (APL) and retinal drusen are reported and appear to be direct consequences of complement activation.
LABORATORY ANALYSIS

- Determine underlying etiology / excluding other diagnoses.
- Measure ADAMTS13
- STEC-HUS investigations in all (5% of STEC- no diarrhoea)
PART 4:

GENETIC AND ACQUIRED DRIVERS OF DISEASE
aHUS

- Studies of hundreds of aHUS patients have provided an excellent understanding of genetic drivers of disease, leading to the development of individualized care.

- Genetic screening and molecular diagnostics, with expert interpretation of the results, should inform therapeutic decisions.
C3G

- Understanding of the genetics of C3G is not yet comparable to that of aHUS.

- There is no clear benefit to performing genetic analysis in all cases of C3G.
GENETIC TESTING

- In aHUS and C3G
  - Screen CFH, CD46, CFI, C3, CFB, THBD, CFHR1, CFHR5, & DGKE.
- In aHUS,
  - Screen risk haplotypes CFH-CFHR3 and MCP_{ggaac}
    - modify disease penetrance and severity.
- In both aHUS and C3G,
  - copy number variation analysis
  - hybrid genes and other complex genomic rearrangements in the CFH/CFHRs genomic region must be included in the genetic testing.
GENETIC TESTING

• In aHUS identification of pathogenic variant
  – Reinforces diagnosis & establishes cause of disease
  – Facilitates management & genetic counseling

• In aHUS genetic analysis is essential in living-related kidney donor transplantation.
  – Only consider if causative genetic factor identified & absent in donor

• In aHUS, genetic testing is recommended for patients in whom discontinuation of Eculizumab is being considered.

• In C3G less clear
UNDERSTANDING GENETIC VARIANTS

• Genetic variants should be classified as “benign,” “likely benign,” “variant of uncertain significance (VUS),” “likely pathogenic,” or “pathogenic,” following international guidelines.

• Genetic analysis should be interpreted by a laboratory with expertise in aHUS and C3G.
**ACQUIRED DRIVERS OF DISEASE**

- In both aHUS and C3G, acquired drivers of disease are autoantibodies to complement proteins or protein complexes that impair normal function.

**Complement Activation**

- Cell Lysis
- Membrane Attack Complex (C5b-9)
- Inflammation
- Anaphylatoxins (C3a, C5a)
- Opsonization
  - (C3b)

**Complement Regulation**

- αCFH Ab
- CFI Ab
- CFH
- CFI
- MCP

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Kidney Disease: Improving Global Outcomes
ACQUIRED DRIVERS OF DISEASE

In aHUS, the best-studied acquired drivers are FH autoantibodies, which are usually seen in association with deletion of the $CFHR3$ and $CFHR1$ genes.

- The deletion of $CFHR3$ and $CFHR1$ is a common copy number variation that can be identified on genetic testing.
- The finding of FH autoantibodies should be confirmed in a second sample at least 4 weeks after the initial sample.
ACQUIRED DRIVERS OF DISEASE

• In C3G, the most common autoantibodies are to C3 convertase, a serine protease formed from C3b and Bb
  – These autoantibodies are called C3Nefs
  – They stabilize C3 convertase and prolong its half-life
• Other antibodies in C3G include FH autoantibodies and C4Nefs
• In older adults, serum free light chains (FLC) should be assayed.
  – Serum FLC assays have contributed to major improvements in care for patients with monoclonal gammopathy.
  – Serum FLC assays can be used as a first-line test in screening pathways for a light chain clone in older adults with kidney disease
PART 5:
TREATMENT STRATEGIES
TREATMENT: aHUS

• All patients with a clinical diagnosis of primary aHUS are eligible for treatment with eculizumab.
  – The dosing schedule as per Eculizumab registration trials

• Treatment duration is controversial as there is no evidence to support life-long therapy in all aHUS patients.
  – Two options for long-term dosing have been considered:
    • The minimal dose required to achieve complement blockade
    • A discontinuation dosing schedule.
  – Dose reduction or discontinuation require ongoing monitoring
TREATMENT: aHUS

• If access to eculizumab is unavailable, plasma therapy can be used.

• The use of plasma exchange when eculizumab is available may be associated with some improvement but delaying use of eculizumab may lead to a suboptimal therapeutic outcome.

• Eculizumab increases the risk of meningococcal infection.
  – Patients should receive vaccination against meningococcus (including Type B); however, vaccination should not delay the start of eculizumab therapy.
  – Antibiotic prophylaxis is mandated during the first 2 weeks.
Kidney transplantation should be delayed for at least 6 months after the start of dialysis as limited renal recovery is possible several months after starting eculizumab.

Living-related kidney donation carries a risk for recurrence in the recipient and a risk of de novo disease in the donor should the donor carry an at-risk genetic variant.

Liver transplant remains an option in patients with liver-derived complement protein abnormalities, in particular for renal transplant recipients with uncontrolled disease activity despite eculizumab therapy.
Clinical diagnosis of aHUS

High titer FH autoantibody

Plasma therapy

Simultaneous start of anticellular therapy

Continue plasma therapy indefinitely

Periodic monitoring of FH autoantibody level

Discontinue therapy when antibody titer falls below a pathogenic titer for at least 6 months

Eculizumab

Simultaneous start of anticellular therapy

Continue eculizumab therapy indefinitely

Periodic monitoring of FH autoantibody level

Discontinue therapy when antibody titer falls below a pathogenic titer for at least 6 months

Kidney Disease: Improving Global Outcomes
# Treatment: C3G: All Patients

<table>
<thead>
<tr>
<th>All Patients</th>
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<tbody>
<tr>
<td>• Optimal blood pressure control (suggested blood pressure below the 90% in children and ≤120/80 in adults)</td>
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<tr>
<td>• Priority agents include angiotensin converting enzyme inhibitors and angiotensin receptor blockers</td>
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<tr>
<td>• Optimal nutrition for both normal growth in children, healthy weight in adults</td>
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<td>• Lipid control</td>
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<tr>
<th>Moderate Disease</th>
<th>Description</th>
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<tr>
<td>• Urine protein over 500 mg/24 hours despite supportive therapy</td>
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<td></td>
<td><strong>OR</strong></td>
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<td>• Moderate inflammation on renal biopsy</td>
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<td><strong>OR</strong></td>
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<td>• Recent increase in serum creatinine suggesting risk for progressive disease</td>
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<tr>
<th>Recommendation</th>
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<tr>
<td>• Prednisone</td>
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<tr>
<td>• Mycophenolate mofetil</td>
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<tr>
<th>Severe Disease</th>
<th>Description</th>
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<tr>
<td>• Urine protein over 2000 mg/24 hours despite immunosuppression and supportive therapy</td>
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<td></td>
<td><strong>OR</strong></td>
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<tr>
<td>• Severe inflammation represented by marked endo- or extracapillary proliferation with or without crescent formation despite immunosuppression and supportive therapy</td>
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<tr>
<td></td>
<td><strong>OR</strong></td>
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<tr>
<td>• Increased serum creatinine suggesting risk for progressive disease at onset despite immunosuppression and supportive therapy</td>
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<table>
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<tr>
<td>• Methylprednisolone pulse dosing as well as other anti-cellular immune suppressants have had limited success in rapidly progressive disease</td>
</tr>
<tr>
<td>• Data are insufficient to recommend eculizumab as a first-line agent for the treatment of rapidly progressive disease</td>
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</table>
**TREATMENT: C3G**

- A retrospective study supports the effectiveness of mycophenolate mofetil in C3GN patients.
- No specific recommendation can be made for plasma therapy or rituximab (an anti-CD20 antibody).
- Since the pathogenesis of C3G is due to dysregulation and hyperactivity of the alternative pathway of complement, eculizumab has been tried in a limited number of patients with varied results.
TREATMENT: C3G: TRANSPLANT

- No specific data are available to inform decisions surrounding transplantation in C3G.
- Recommendations reflect expert opinion and limited case reports.
- C3G recurs in allografts at a high rate, leading to graft loss in ~50% of patients.
RESEARCH RECOMMENDATIONS: SUMMARY

- **aHUS**
  - A comparative study of biopsies from patients with well-documented malignant hypertension and patients with well-documented alternative complement pathway disease
  - A longitudinal study of patients with features of chronic microangiopathy on biopsy but without a history of acute presentation

- **C3G**
  - A multicenter study analyzing biopsies to define the relationship of morphology to etiology, clinical course and response to therapy
  - Comprehensive genetic testing to fill the knowledge gap in establishing robust phenotype-genotype correlations
Research Recommendations: Summary

• Clinical studies – aHUS
  – Define how complement biomarkers correlate with current or impending aHUS relapse and/or renal involvement
  – Identify risk factors for relapse upon cessation of anti-complement therapy
  – Identify alternative anti-complement therapeutics

• Clinical studies – C3G
  – Assess the value of proximal (at the level of the AP) anti-complement therapy
  • Development and trial novel complement inhibitors
  – Determine value of complement biomarkers to inform clinical outcome in C3G patients and stratify them into targeted treatment groups
CONCLUSIONS

• While there are knowledge gaps in both aHUS and C3G, the evidence base for the management of patients with C3G lags behind that of aHUS; addressing this disparity should be a priority.

• Although these two diseases are presented as distinct entities, there is substantial overlap in their pathogenesis and clinical presentation.
Atypical hemolytic uremic syndrome and C3 glomerulopathy: conclusions from a “Kidney Disease: Improving Global Outcomes” (KDIGO) Controversies Conference

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