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Efficacy of Eculizumab in Coexisting Complement C3 Glomerulopathy and Atypical Hemolytic Uremic Syndrome

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INTRODUCTION

Complement-mediated kidney diseases are a currently evolving area in nephrology. Two kidney diseases associated with dysregulation of the complement alternative pathway are atypical hemolytic uremic syndrome (aHUS) and C3 glomerulopathy (C3G, including C3 glomerulonephritis and dense deposit disease [DDD]).[1]

Atypical hemolytic uremic syndrome is very rare and is characterized by acute kidney injury, thrombocytopenia, and microangiopathic hemolytic anemia. Most patients carry an underlying inherited, acquired complement abnormality, or both, leading to dysregulated activity of the alternative pathway. C3 glomerulopathy is rare as well, and represents a glomerular disease caused by uncontrolled activation of the complement cascade that leads to C3 deposition within the glomerulus. The position and density of the glomerular C3 deposits allows distinction into 2 subtypes, C3 glomerulonephritis and DDD. Most frequently, dysregulation occurs at the level of the C3-convertase of the alternative pathway and is driven by genetic or acquired defects, or both. Improvement in understanding the common pathophysiology leads to the hypothesis that patients can transition between diseases within this spectrum.[2]

We describe 2 pediatric patients who presented with both renal clinical pictures related to complement alternative pathway dysregulation—in 1 with simultaneous onset, and in the other at subsequent times. Table 1 summarizes the main clinical features.

CASE PRESENTATION

Case 1

A 9-year-old boy with a negative family history of nephropathy had the first occasional observation of significant circulating C3 reduction (15 mg/dl, range 90–180) with a normal C4 level. Three years later (12 years old) the patient was admitted after the appearance of nonnephrotic proteinuria (0.6 g/24 h) with normal renal function and persistent reduction of C3 levels (9 mg/dl). Histologic examination showed a membranoproliferative glomerulonephritis pattern by light microscopy with C3 deposition by immunofluorescence (Figure 1a), consistent with a diagnosis of C3 glomerulopathy; electron microscopy showed ribbon-like dense intramembranous deposits as observed in DDD (Figure 1b). The patient was treated with oral prednisone, achieving complete remission of proteinuria within 2 months. After gradual tapering, steroids were discontinued after 6 months, leaving only angiotensin-converting enzyme inhibitor treatment.

Study of the complement alternative pathway showed positive C3 nephritic factor (C3 Ag, 188 mg/l; 20% of residual convertase activity [400 μg immunoglobulin G], according to the method described by Fremeaux-Bacchi) and normal C5b9 deposition on resting and activated endothelial cells. Regarding genes involved in complement system activity regulation, no pathogenetic mutations of *MCP, CF1, C3, CFB,* or *THBD* were found, although a polymorphism associated with DDD was identified on the *CFH* gene

Table 1. Clinical characteristics

Characteristic	Case 1	Case 2
Age at onset, yr	9	6
Sex	Male	Male
Simultaneous onset of aHUS/C3G	No	Yes
C3 levels at onset	Low	Low
C3 levels after eculizumab	Low	Normal
C3 nephritic factor	+	−
Complete remission[a]	Yes	Yes
Time of complete remission, mo	4	4

aHUS, atypical hemolytic uremic syndrome; C3, complement C3; C3G, C3 glomerulopathy.
[a]Defined according to Kidney Disease: Improving Global Guidelines 2012 Guidelines.[S1]

(heterozygous polymorphism p.V62I). Screening results for ocular drusen were negative.

The patient was readmitted 1 year later for severe nephrotic syndrome and acute kidney injury (serum creatinine 1.25 mg/dl) requiring albumin and diuretic administration. Reduction of the C3 level was confirmed (8 mg/dl) with a normal C4 level. A second kidney biopsy was performed. Histologic examination confirmed the previous picture. Electron microscopy revealed features of coexisting DDD and type 1 membranoproliferative glomerulonephritis with subendothelial deposits (not shown). Three boli of methylprednisolone were administered and treatment was then tapered to oral prednisone, associated with mycophenolate mofetil. No remission of proteinuria was observed.

The patient was admitted again 2 weeks later in an anasarcatic state with severe arterial hypertension requiring 3 drugs to reach normal targets. The patient also experienced seizures owing to posterior reversible encephalopathy syndrome, which required intensive care and antiepileptic treatment with levetiracetam. During hospitalization, worsening of anemia, low platelet count, and elevated hemolysis indexes were found, with elevated soluble C5b9 levels (1269 ng/ml). A third kidney biopsy was performed, confirming previous findings associated with concentrically hypertrophied arteriolar walls and fibrinoid eosinophilic deposits in capillary lumen, focal positivity of fibrinogen by immunofluorescence, and subendothelial expansion of capillary walls by electron microscopy,

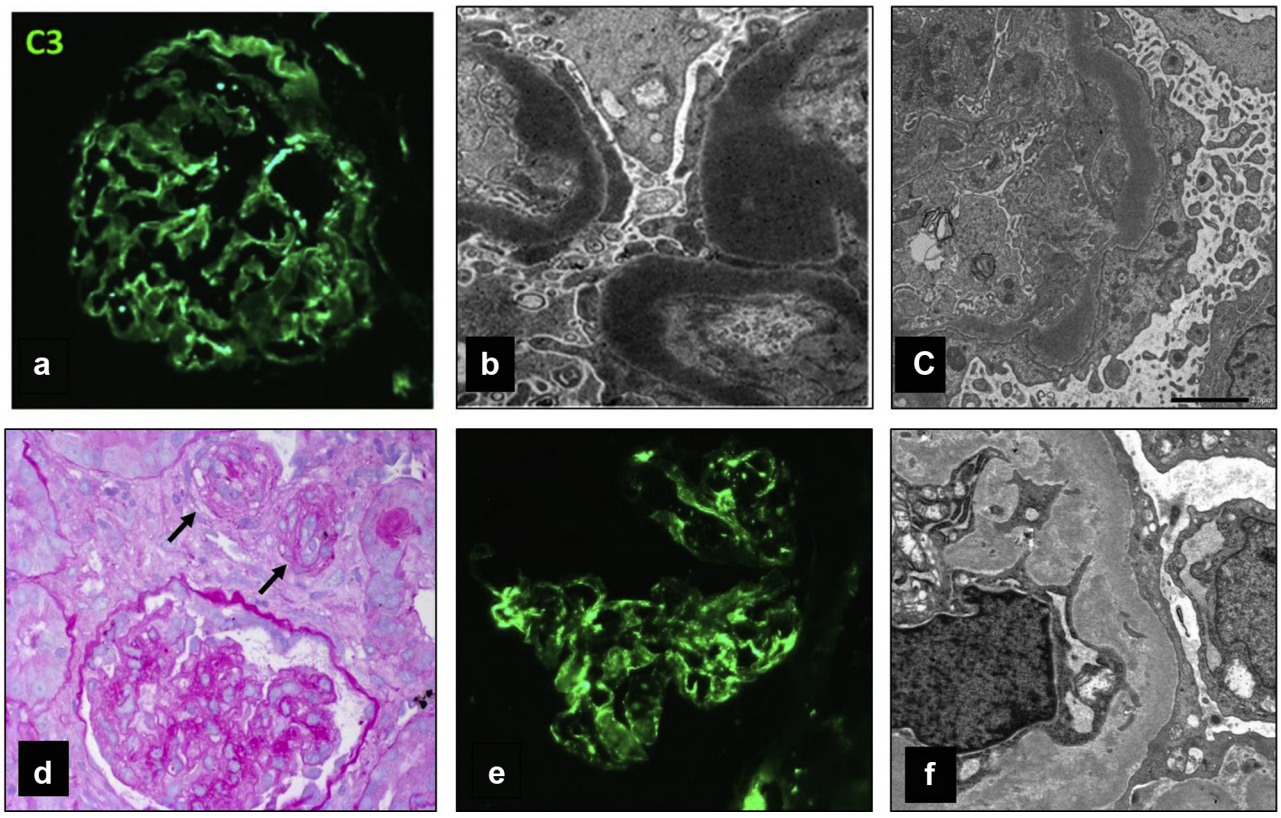

Figure 1. Case 1. Findings of the first renal biopsy (a). C3 immunofluorescence shows strong and diffuse positivity of both capillaries and mesangium. At first renal biopsy, electron microscopy highlights ribbon-like intramembranous dense deposits consistent with dense deposit disease (b). At third renal biopsy, electron microscopy shows dense deposit disease and thickening of capillary wall for expansion of subendothelial space, endothelial swelling, and loss of fenestration, all features of thrombotic microangiopathy (c). Case 2. Histologic examination shows proliferative glomerulonephritis and concentrically hypertrophic arteriolar narrowing (arrows) (d) (periodic acid–Schiff stain, original magnification ×20). C3 immunofluorescence shows strong and diffuse positivity of both capillaries and mesangium (e). Thickening of the capillary wall for endothelial hyperplasia, subendothelial deposits, and cellular interposition indicate membranoproliferative C3 glomerulonephritis.

consistent with the clinical hypothesis of aHUS (Figure 1c). In addition to administration of prednisone and mycophenolate mofetil, treatment with anti-C5 monoclonal antibody (eculizumab) was prescribed at the standard dosage. The patient showed immediate benefit with reduction of proteinuria and increase in serum albumin levels as well as improvement in blood pressure and in hemolysis. Complete remission of nephrotic syndrome (urine albumin-to-creatinine ratio <0.2 mg/mg) was achieved 4 months after the first eculizumab infusion. C3 serum levels were persistently low. The prednisone dose was tapered and then discontinued after 9 months; infusions of eculizumab were discontinued after 12 months.

To date (19 months after the patient's last eculizumab infusion), nephrotic syndrome is still in complete remission with normal renal function and normal blood pressure levels. Maintenance therapy consists of mycophenolate mofetil associated with double renin-angiotensin-aldosterone system blockade. C3 serum levels remain persistently low.

Case 2

A 6-year-old boy with no family history nephropathy was admitted for acute kidney injury with anemia, low platelet count, and an elevated hemolysis index (haptoglobin, <3 mg/dl; lactate dehydrogenase, 3500 U/l; and high indirect bilirubin, 3.2 mg/dl). Renal function rapidly worsened, requiring renal replacement therapy with hemodialysis. In the absence of diarrhea and owing to clinical suspicion of aHUS, therapy with eculizumab was initiated with prompt improvement of renal function. Mild arterial hypertension was treated with amlodipine. The circulating C3 level was low with a normal C4 level. Results of fecal cultures and serologic testing were negative for Shiga or Shiga-like toxins or antibodies. The disintegrin and metalloproteinase with thrombospondin type 1 motif, 13 (ADAMTS13) level was normal. No mutations associated with aHUS were identified in genes regulating the pathway of complement (*CFH, MCP, CFB, C3, CFI,* and *THBD*). Given the presence of persistent nephrotic-range proteinuria, a kidney biopsy was performed. Light microscopy showed proliferative glomerulonephritis associated with remodeling or narrowing of the arteriolar wall as post-acute signs of thrombotic microangiopathy (Figure 1c). A positive C3 level (++) with mild positivity (+/–) for immunoglobulin and fibrinogen was observed by immunofluorescence (Figure 1e). Electron microscopy showed coarse subendothelial deposits, cellular interposition, glomerular basement membrane layering, and podocyte foot process effacement, consistent with C3 glomerulonephritis (Figure 1f). Three methylprednisolone boli were administered. The patient had a complete response in terms of renal function, proteinuria, and blood pressure; the serum creatinine level returned to normal values (0.5 mg/dl) within 2 months from the first eculizumab infusion. Complete remission of nephrotic syndrome was achieved within 4 months. Circulating C3 levels returned to normal after 10 months. Prednisone was gradually tapered until discontinuation in 6 months; eculizumab was discontinued after 12 months.

To date, the patient is 11 years old. The last dose of eculizumab was infused 4 years ago, and maintenance therapy consists of a low-dose of an angiotensin-converting enzyme inhibitor. Serum creatinine, urinalysis, circulating complement levels, and blood pressure are normal.

DISCUSSION

In the past decade, advances in the understanding of the complement system and its pathways of activation have led to major progress in unraveling the pathogenesis of many kidney diseases that previously were considered idiopathic. Moreover, blockade of the complement system has led to major progress in treatment outcomes. Alternative pathway dysregulation has been identified in most cases of aHUS and C3G, and mutations involving complement regulatory genes are identified in at least 50% of patients, highlighting a common genetic background.[3] Hyperactivation of the complement alternative pathway may be spontaneous or triggered by infections, pregnancy, childbirth, or the development of monoclonal gammopathy.[4] Atypical hemolytic uremic syndrome is a systemic disorder originating from endothelial damage, leading to injury in target organs (renal, gastrointestinal tract, liver, pancreas, and brain); consumptive thrombocytopenia; and microangiopathic hemolytic anemia. C3 glomerulopathy is a recently acknowledged class of glomerulonephritis that often present with hypertension, proteinuria (both nephrotic and nonnephrotic), hematuria, and renal dysfunction. There is evidence that C3G differs from aHUS in where the alternate pathway dysregulation occurs (peripherally in C3G and on the endothelial surface in aHUS) and in the type of underlying mutation.[5] However, some clinical observations, including the 2 patients we describe, suggest that at least in some cases the diseases are 2 faces of the same medal.[6]

Despite the common pathogenetic pathway, few cases with coexisting or shifting C3G or aHUS are described.[2] To the best of our knowledge, these are the first 2 case descriptions of pediatric patients in whom both conditions developed and were treated with eculizumab; main features are resumed in Table 2. No alterations in genes involved in complement regulation were found in either

Table 2. Teaching points

- Complement-mediated kidney diseases are a currently evolving area in nephrology. Kidney diseases associated with dysregulation of the complement alternative pathway are atypical hemolytic uremic syndrome and C3 glomerulopathy (including C3 glomerulonephritis and dense deposit disease).
- Atypical hemolytic uremic syndrome is a rare disease, characterized by acute kidney injury, thrombocytopenia, and microangiopathic hemolytic anemia. Most patients carry an underlying inherited or acquired complement abnormality, or both, which leads to dysregulated activity of the alternative pathway.
- C3 glomerulopathy is a glomerular disease caused by uncontrolled activation of the complement cascade that leads to C3 deposition within the glomerulus. The position and density of the glomerular C3 deposits allows distinction into 2 subtypes: C3 glomerulonephritis and dense deposit disease.
- Only a few cases of patients with coexisting or shifting C3 glomerulopathy and atypical hemolytic uremic syndrome have been described. These 2 cases are the first descriptions of pediatric patients with both conditions who were successfully treated with eculizumab.

patient, but in case 1 only C3 nephritic factor was positive. Both patients had low C3 levels at disease onset. Despite the complete response to therapy, in case 1 the C3 levels were persistently low, whereas C3 levels in case 2 normalized after treatment. Many cases of aHUS with normal complement levels are described, and no association between levels of C3 and severity of the disease has been found. On the contrary, in C3glomerulopathy a persistently low C3 level may be a sign of unresolved peripheral alternative pathway hyperactivation.

The humanized monoclonal antibody eculizumab targeting C5 has shown dramatic efficacy in complement-driven forms of aHUS.[7,8] Conversely, in patients with C3G the efficacy of eculizumab is not well established, and a recent randomized controlled trial (Evaluating the Morphofunctional Effects of Eculizumab Therapy in Primary Membranoproliferative Glomerulonephritis [EAGLE] Study) yielded disappointing results.[9] In contrast with our cases, the EAGLE study involved patients with C3G without associated aHUS features. Both of our patients showed a rapid and sustained response to eculizumab, not only in terms of intravascular hemolysis, as expected, but also in terms of nephrotic-range proteinuria. The efficacy of eculizumab may be explained by the presence of aHUS features, a hallmark of endothelial activation of the complement alternative pathway, which may have made our patients susceptible to eculizumab treatment, as is well established in patients with isolated aHUS. Indeed, our group has previously described a child with a very unusual form of aHUS caused by a thrombomodulin mutation who presented with nephrotic syndrome. In this patient, treatment with eculizumab was effective, even though the patient's proteinuria had resolved with conservative therapy before use of eculizumab.[S2] Whether or not eculizumab may also be beneficial for glomerular inflammation and therefore in reducing proteinuria is less clear.

At last follow-up, both patients were in complete remission, having discontinued eculizumab therapy. Both have normal renal function: 1 with maintenance therapy with mycophenolate mofetil and angiotensin-converting enzyme inhibitor and the other with angiotensin-converting enzyme inhibitor alone. These findings suggest that eculizumab, in addition to being effective in treatment of aHUS, may also be effective in the rare cases of coexisting C3G or aHUS.

DISCLOSURE

MV has received consulting or advisory fees from Alexion, Achillion Pharmaceuticals, Novartis, and F. Hoffmann-La Roche AG that covered only travel expenses or financed a patient nonprofit organization and in no way influenced the content of this study. All the other authors declared no competing interests.

SUPPLEMENTARY MATERIAL

Supplementary File (Word)

Supplementary References.

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