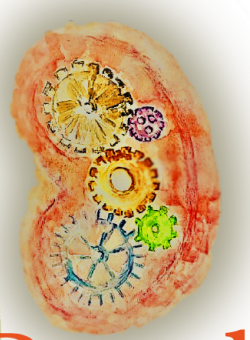
Anno VII

Cadernos de Patologia Renal



COMPLEMENTO E DOENÇAS RENAIS.

08 a 10 de maio, 2023
Hospital do Rim,
São Paulo, SP

Protocolos de Intervenção MAT:
passado, presente e futuro

Dra. Lilian Monteiro P. Palma

Nefrologista Pediátrica – UNICAMP e LUME Nefrologia e Dialise

Realização: hrim FUNDAÇÃO OSWALDO RAMOS NÚCLEO DE ENSINO E PESQUISA

Apoio: Sociedade Brasileira de Nefrologia

SONESP Sociedade de Nefrologia do Estado de São Paulo

Conflitos de interesse

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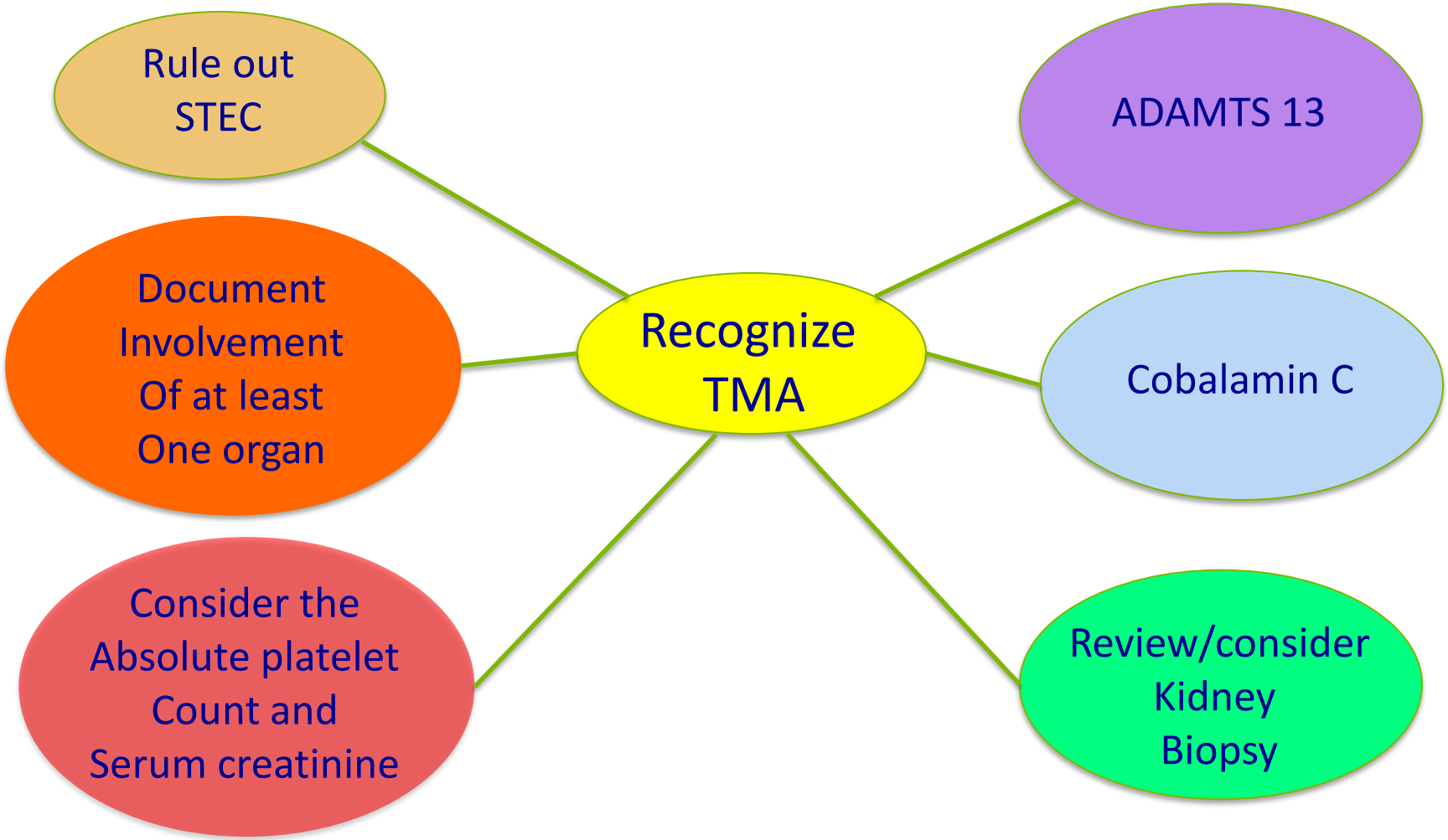
COMPLEMENTO E DOENÇAS RENAIS.

- Palestrante Síndrome Hemolítico Urêmica atípica para Alexion
- Consultora científica Glomerulopatia C3 para Apellis
- Esta não é uma aula patrocinada

Atypical Hemolytic Uremic Syndrome (aHUS): Essential Aspects of an Accurate Diagnosis

Jeffrey Laurence, MD, Hermann Haller, MD, Pier Mannuccio Mannucci, MD, Masaomi Nangaku, MD, PhD, Manuel Praga, MD, and Santiago Rodriguez de Cordoba, PhD



Laurence J. *Clinical Advances in Hematology and Oncology,* 2016 sup 11

Revolutionary

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ORIGINAL ARTICLE

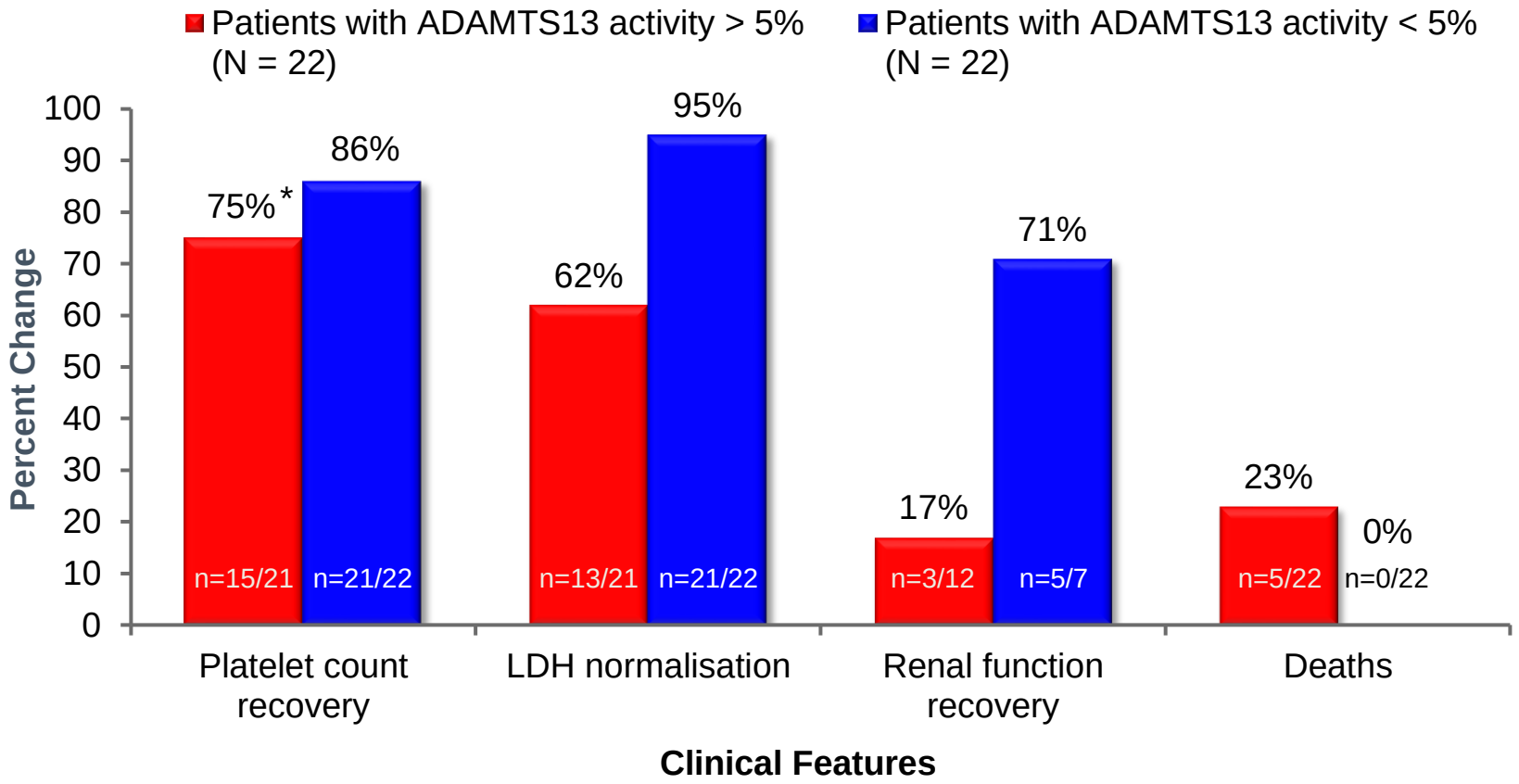
Improved Survival in Thrombotic Thrombocytopenic Purpura–Hemolytic Uremic Syndrome — Clinical Experience in 108 Patients

William R. Bell, M.D., Hayden G. Braine, M.D., Paul M. Ness, M.D., and Thomas S. Kickler, M.D.

August 8, 1991
N Engl J Med 1991; 325:398-403
DOI: 10.1056/NEJM199108083250605

Among Patients With TMA Treated With PE/PI, Those With ADAMTS13 Activity > 5% Have Increased Rates of ESKD and Premature Mortality

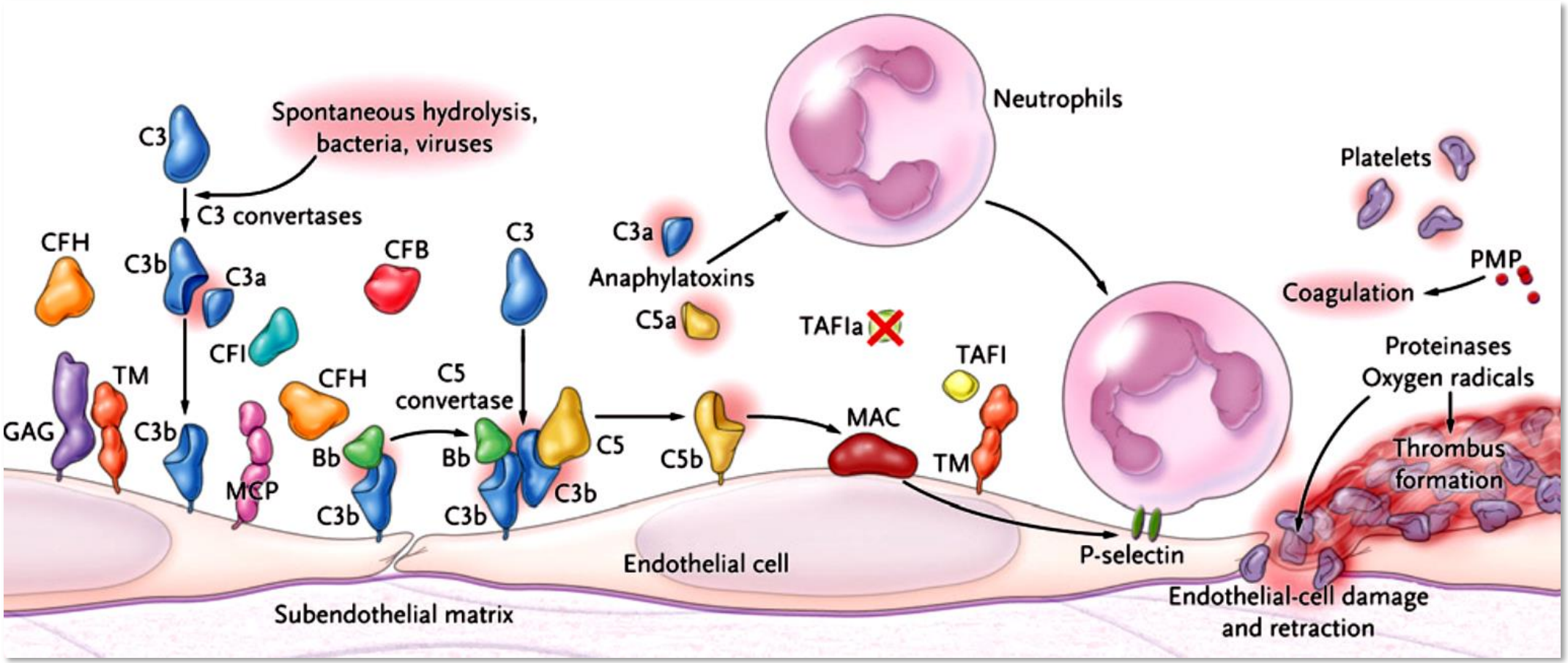
A retrospective analysis of 44 patients undergoing PE/PI for the treatment of TMA examined recovery time as determined by laboratory results



*Of 21 patients with available data.

Reference: 1. Pishko AM, et al. *Blood*. 2014;124(21):4192.

Alternative Complement Pathway dysregulation



Noris M & Remuzzi G. *N Engl J Med,* 2009

Review

Pediatric Atypical Hemolytic Uremic Syndrome Advances

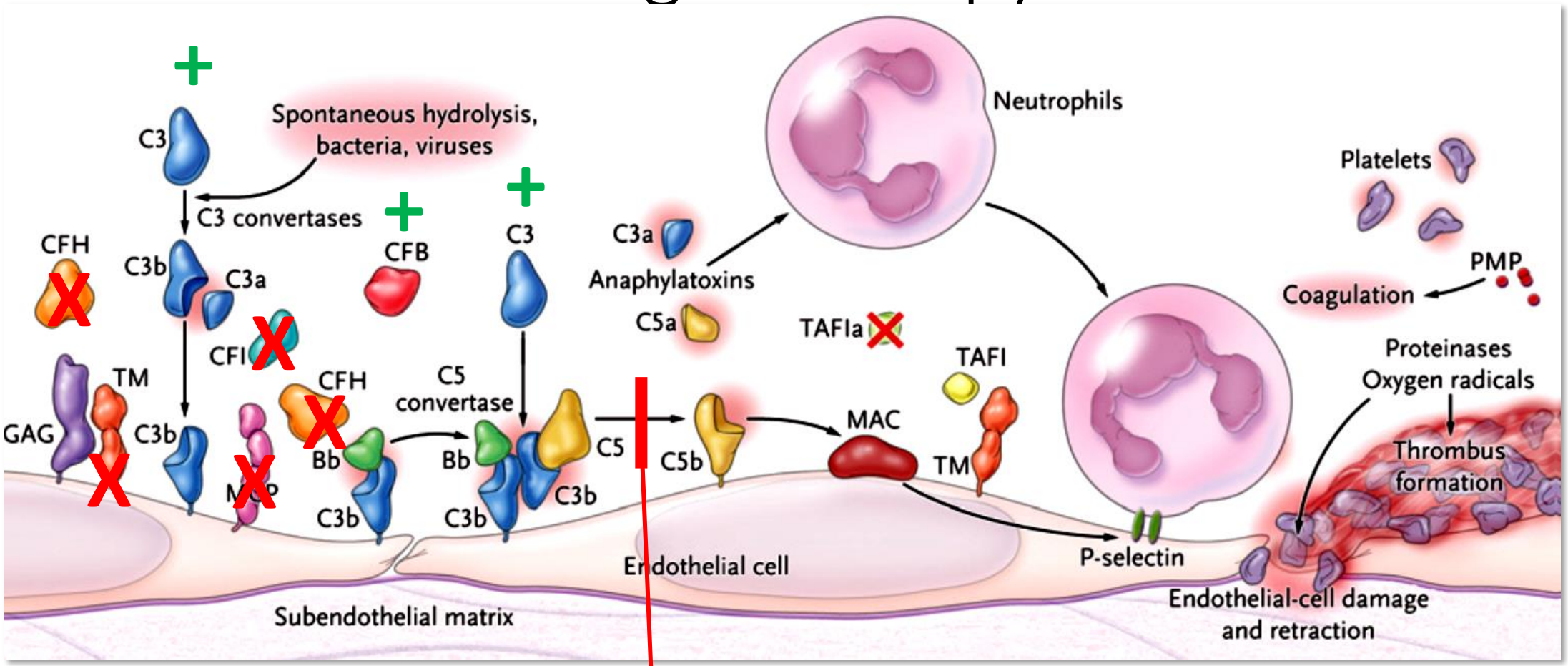
Rupesh Raina [1,2,*], Nina Vijayvargiya [1], Amrit Khooblall [1], Manasa Melachuri [3], Shweta Deshpande [1], Divya Sharma [3], Kashin Mathur [1], Manav Arora [1], Sidharth Kumar Sethi [4] and Sonia Sandhu [5]

	Current Therapeutics	Drug Class	Pathophysiology/ Mechanism of Action	Complement Pathway Proteins Affected
Current Therapeutics.	Eculizumab	Monoclonal Antibody, terminal complement inhibitor	Binds to C5 and prevents cleavage to C5a and C5b	C5a and C5b levels
	Ravulizumab		Prevents the cleavage of C5 into C5a and C5b	
	Nomacopan	C5aR1 antagonist	Inhibits C3a, C4a, and C5a protein function	C3a, C4a, and C5a levels
	Avocapan	Recombinant protein derived from a tick C5 inhibitor	Inhibits C5 and leukotriene B4	C5 and leukotriene B4 levels
	Cemdisiran	Short sequences of interfering RNA	Match mRNA for the C5 protein, with N-acetylgalactosamine	C5 levels
Biosimilars	ABP 959	Biosimilar to FDA-licensed Eculizumab	Binds to C5 and prevents cleavage to C5a and C5b	C5a and C5b levels
	Elizaria	Russian biosimilar to Eculizumab	Binds to C5 and prevents cleavage to C5a and C5b	

Future Therapeutics			
ALXN1720	Anti-C5 mini body	Binds to C5 protein and blocks its activation	C5 levels
Pozelimab	C5 antibody	Decrease hemolysis and C5 level	
Tesidolumab	C5 monoclonal IgG1 antibody	Binds to C5 preventing its cleavage	C5a and C5b levels
Crovalimab	Binds to a C5 epitope	Binds to C5b and prevents the formation of the MAC complex	C5a, C5b, and MAC complex proteins
IFX-1	Targets C5a protein directly	Binds to C5a	C5a levels
Zilucoplan	Binds to the C5b protein and the C5b part of C5	Inhibits C5b binding on C5 by binding to its C5 domain	C5b levels
Avacincaptad Pegol	Binds to and inhibits the C5 protein	Prevents cleavage of C5	C5 levels
Avdoralimab	Anti-C5aR1 antibody	Blocks T-cell and natural killer cell activity through C5aR1 suppression	C5aR1 levels
MAC Inhbitor HMR59	Promotes CD59 production	Enhances synthesis of CD59, which blocks C5b-9 formation	C5b-9 formation

Raina R et al. *Cells*, 2021 (10)

Atypical Uremic Hemolytic Syndrome (aHUS) Target Therapy



Noris M & Remuzzi G. *N Engl J Med,* 2009

Eculizumab aHUS Dosing Schedule

Administration

Eculizumab should be administered at the recommended dosage regime time points, or within 2 days of these time points

aHUS Dosing Schedule for Adults (≥18 years of age)

Pretreatment	Induction Phase					Maintenance Phase				
≥2 weeks before induction	Week	1	2	3	4	5	6	7	8	9+
Neisseria meningitidis vaccination	Soliris® Dose	900 mg	900 mg	900 mg	900 mg	1200 mg	–	1200 mg	–	1200 mg

Q2W

aHUS Weight-based Dosing Schedule for Patients < 18 Years

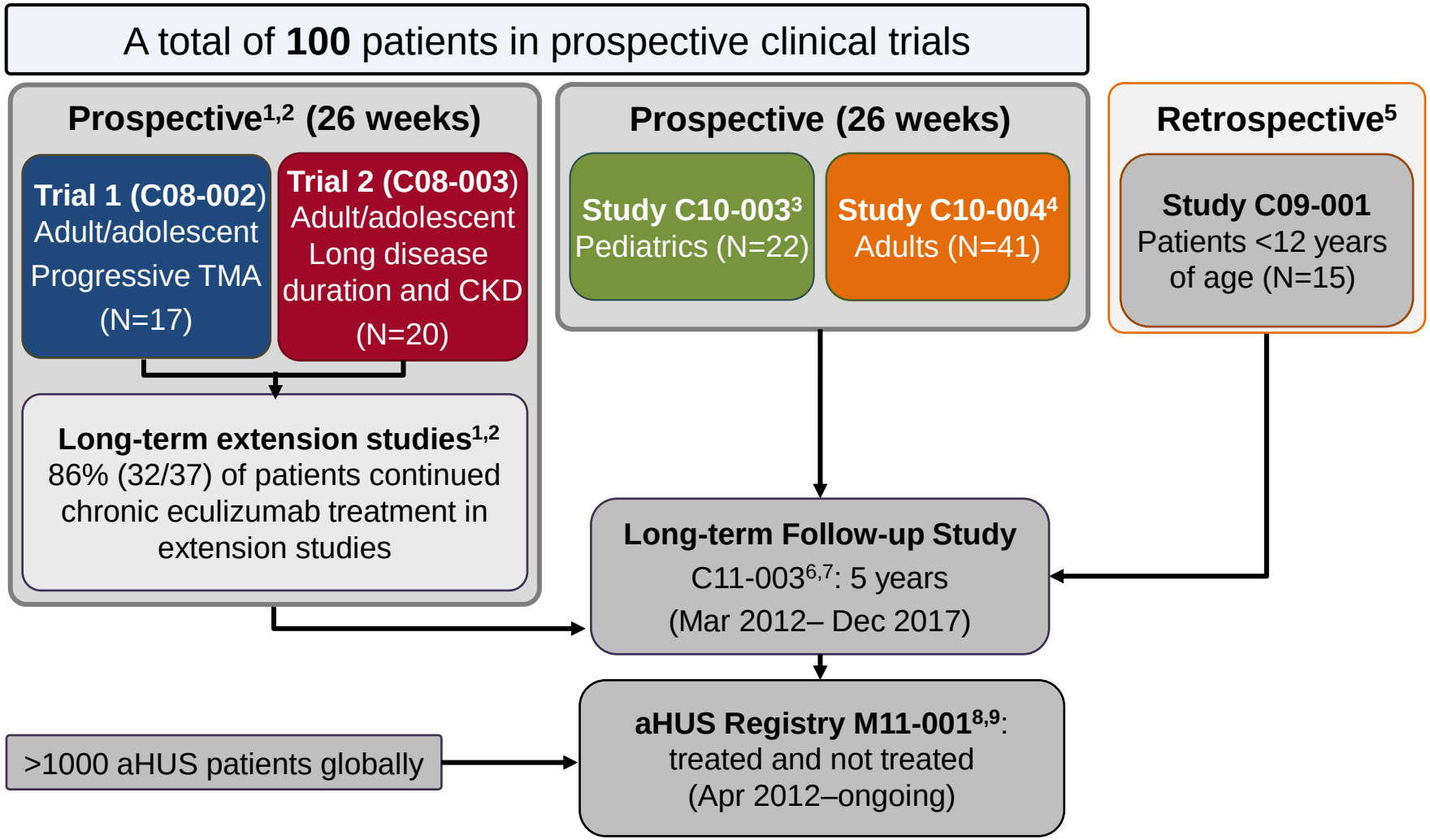
Body Weight	Induction Phase	Maintenance Phase
40 kg and over	900 mg weekly × 4 doses	1200 mg at week 5; then 1200 mg every 2 weeks (Q2W)
30 kg to <40 kg	600 mg weekly × 2 doses	900 mg at week 3; then 900 mg Q2W
20 kg to <30 kg	600 mg weekly × 2 doses	600 mg at week 3; then 600 mg Q2W
10 kg to <20 kg	600 mg weekly × 1 dose	300 mg at week 2; then 300 mg Q2W
5 kg to <10 kg	300 mg weekly × 1 dose	300 mg at week 2; then 300 mg Q3W

aHUS, atypical hemolytic uremic syndrome; Q2W, once every 2 weeks; Q3W, once every 3 weeks.

US Food and Drug Administration. Soliris (eculizumab) [prescribing information]. New Haven, CT: Alexion Pharmaceuticals, Inc., 2016.

Please See Summary of Product Characteristics for Soliris®, including Special Warnings and Precautions for use. Alexion 2015.

Eculizumab Clinical Development Program (2011–Ongoing)



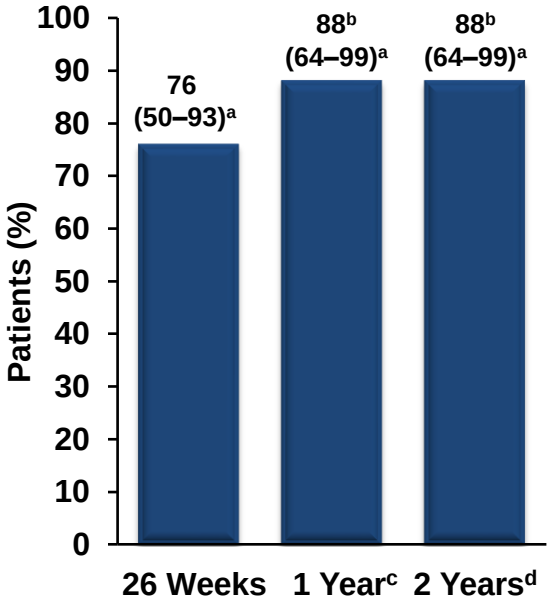
aHUS, atypical hemolytic uremic syndrome; CKD, chronic kidney disease; TMA, thrombotic microangiopathy.

1. Legendre et al. *N Engl J Med.* 2013;368:2169–81. 2. Licht et al. *Kidney Int.* 2015;87:1061-73. 3. Greenbaum et al. *Kidney Int*. 2016;89:701-11. 4. Fakhouri et al. *Am J Kidney Dis*. 2016;68:84-93. 5. Eculizumab Summary of Product Characteristics. Alexion Europe SAS, 2014. 6. Menne et al. *J Am Soc Nephrol.* 2015;26:458A. 7. http://clinicaltrials.gov/show/NCT01522170. 8. Licht et al. *BMC Nephrol.* 2015;16:207. 9. https://clinicaltrials.gov/ct2/show/NCT01522183.

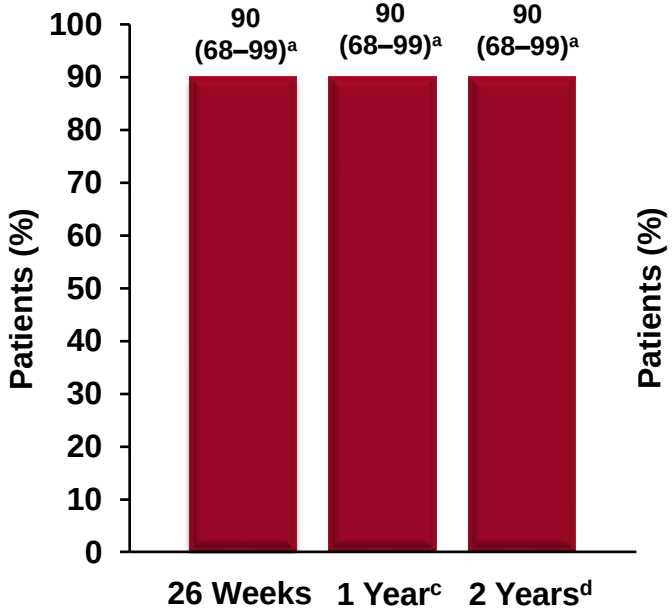
Hematologic Normalization Achieved With Ongoing Eculizumab

Hematologic normalization: Normal platelet and LDH levels (≥2 consecutive measurements, ≥4 weeks apart)

C08-002[1,2] (N=17): Progressing TMA



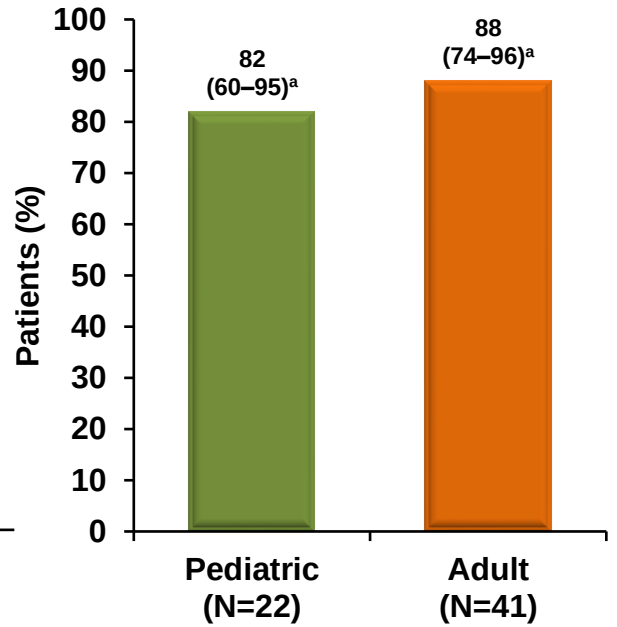
C08-003[1,2] (N=20): Long Duration of aHUS and CKD



C10-003[3] (N=22): Pediatric

C10-004[4] (N=41): Adult

(at 26 weeks)



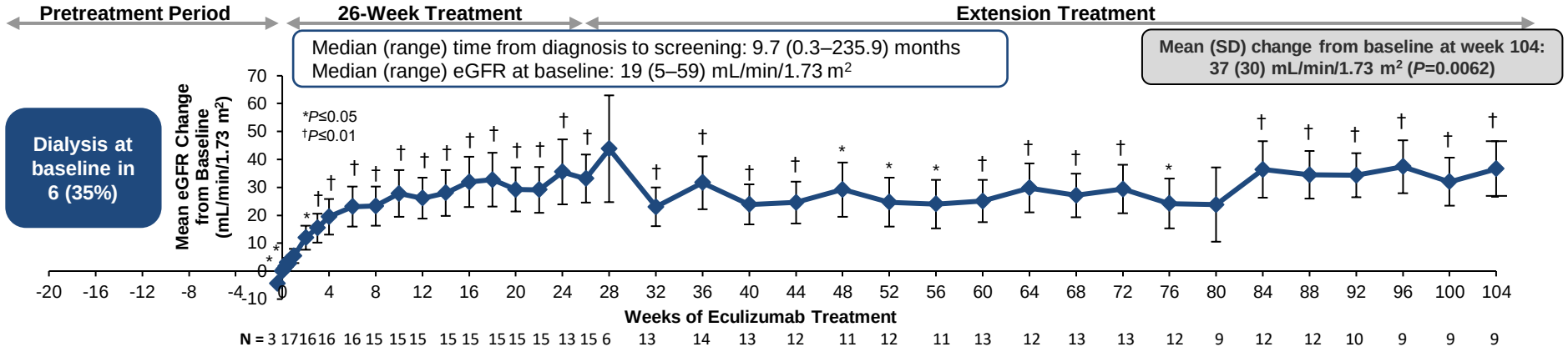
In <u>all studies</u>, hematologic normalization was achieved regardless of the identification of a complement abnormality

[a]95% CI. [b]The 2 patients from C08-002 who did not achieve hematologic normalization at years 1 and 2 were those who withdrew from the study within the initial 26-week treatment period. [c]Median duration 64 weeks. [d]Median duration 100 weeks.
aHUS, atypical hemolytic uremic syndrome; CI, confidence interval; CKD, chronic kidney disease; LDH, lactate dehydrogenase; TMA, thrombotic microangiopathy.

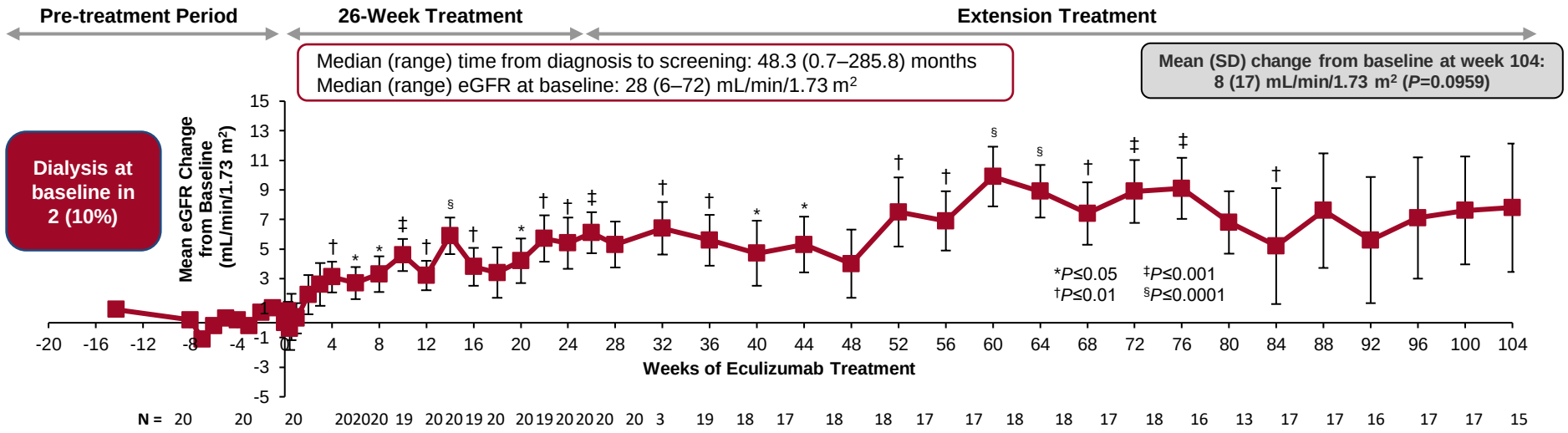
1. Legendre et al. *N Engl J Med*. 2013;369(14):1379-80. 2. Licht et al. *Kidney Int.* 2015;87(5):1061-1073. 3. Greenbaum et al. *Kidney Int*. 2016;89(3):701-11. 4. Fakhouri et al. *Am J Kidney Dis*. 2016;68(1):84-93.

Continued Improvement in eGFR Over 2 Years With Eculizumab Therapy

C08-002 (N=17): Progressing TMA



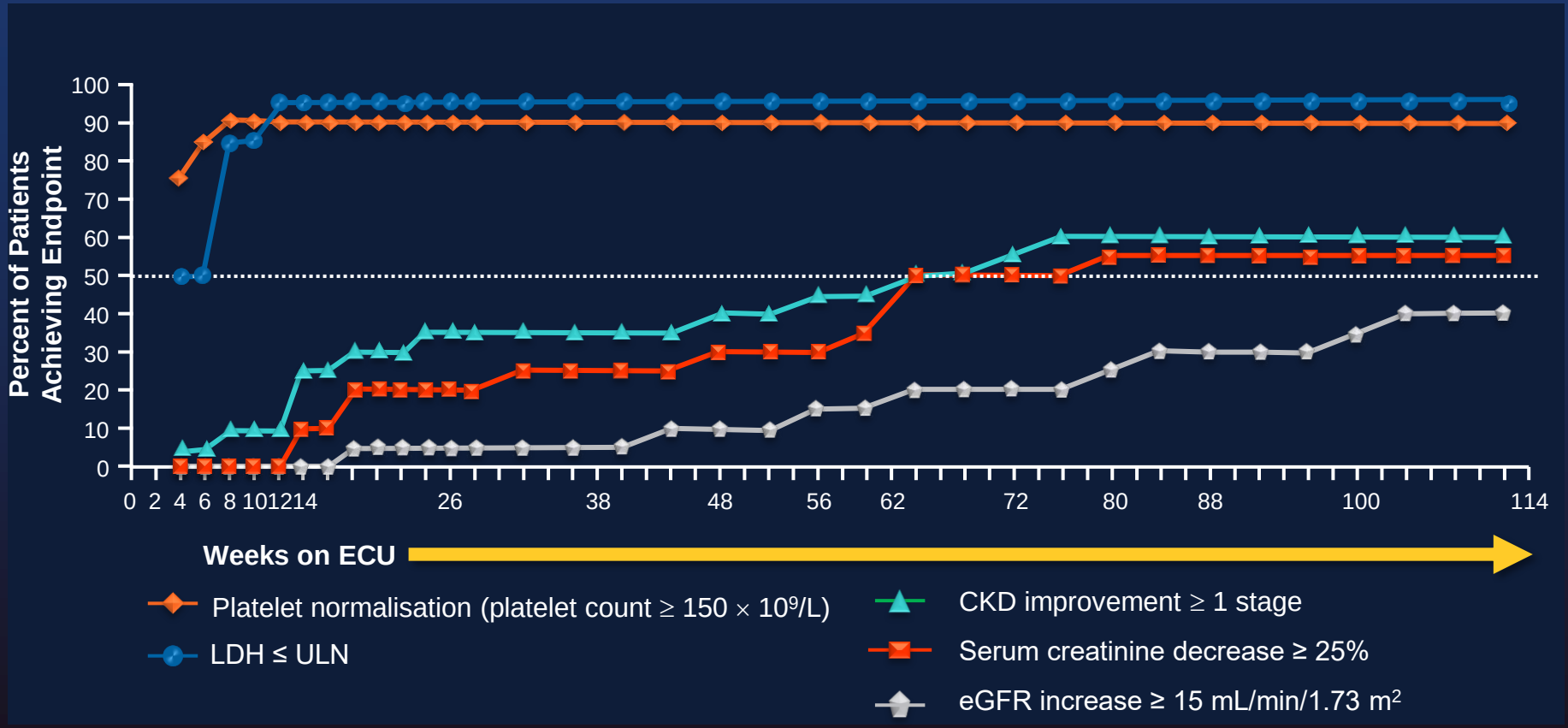
C08-003 (N=20): Long Duration of aHUS and CKD



aHUS, atypical hemolytic uremic syndrome; CKD, chronic kidney disease; eGFR, estimated glomerular filtration rate; TMA, thrombotic microangiopathy.

Legendre et al. *N Engl J Med*. 2013;369(14):1379-80. Licht et al. *Kidney Int*. 2015;87(5):1061-73

Sustained Improvements in Hematologic Markers of Complement-mediated TMA Followed by Continued Improvement in Renal Function with Ongoing Eculizumab Treatment

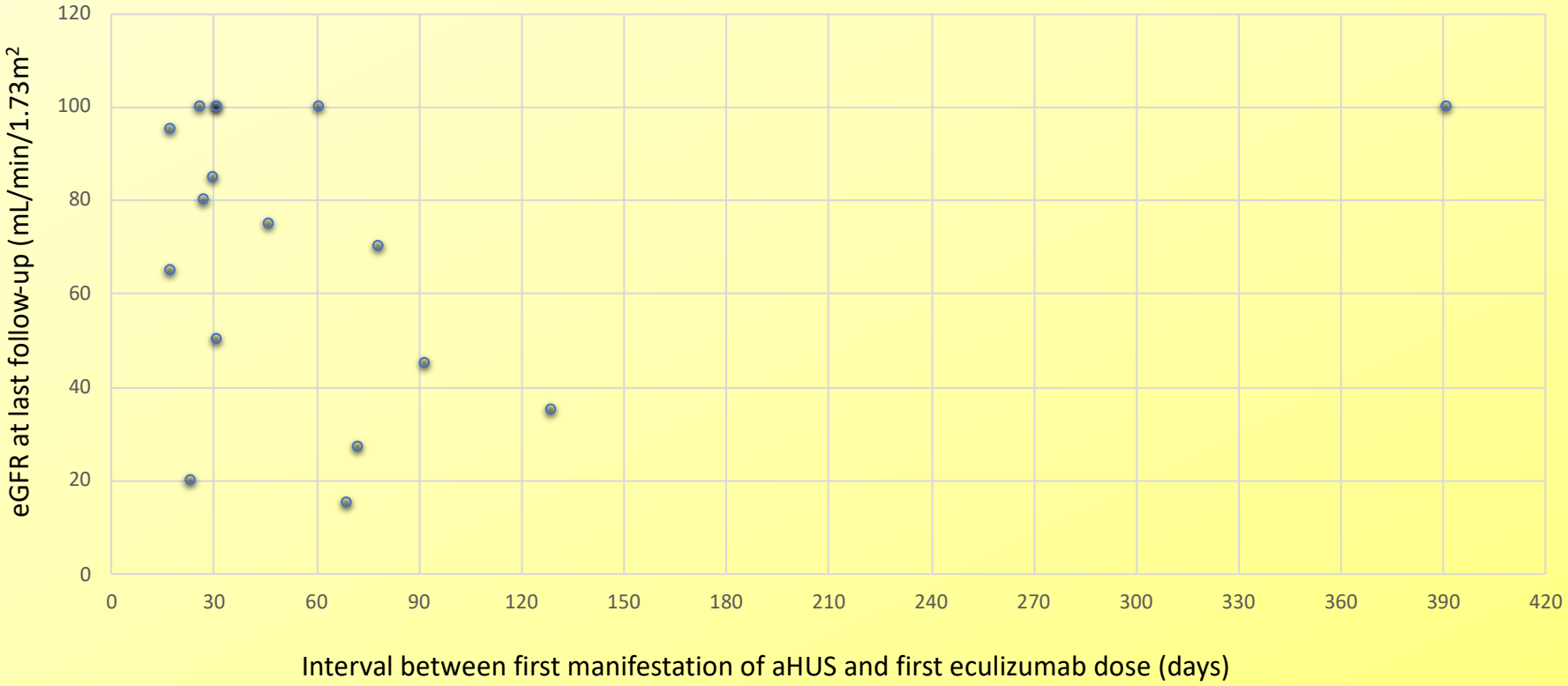


CKD, chronic kidney disease; eGFR, estimated glomerular filtration rate; LDH, lactate dehydrogenase.

Reference: 1. Licht C, et al. Poster presented at: 55th ASH Annual Meeting and Exposition; December 7-10, 2013; New Orleans, LA. Poster 2186.

Kidney function in patients who stopped dialysis

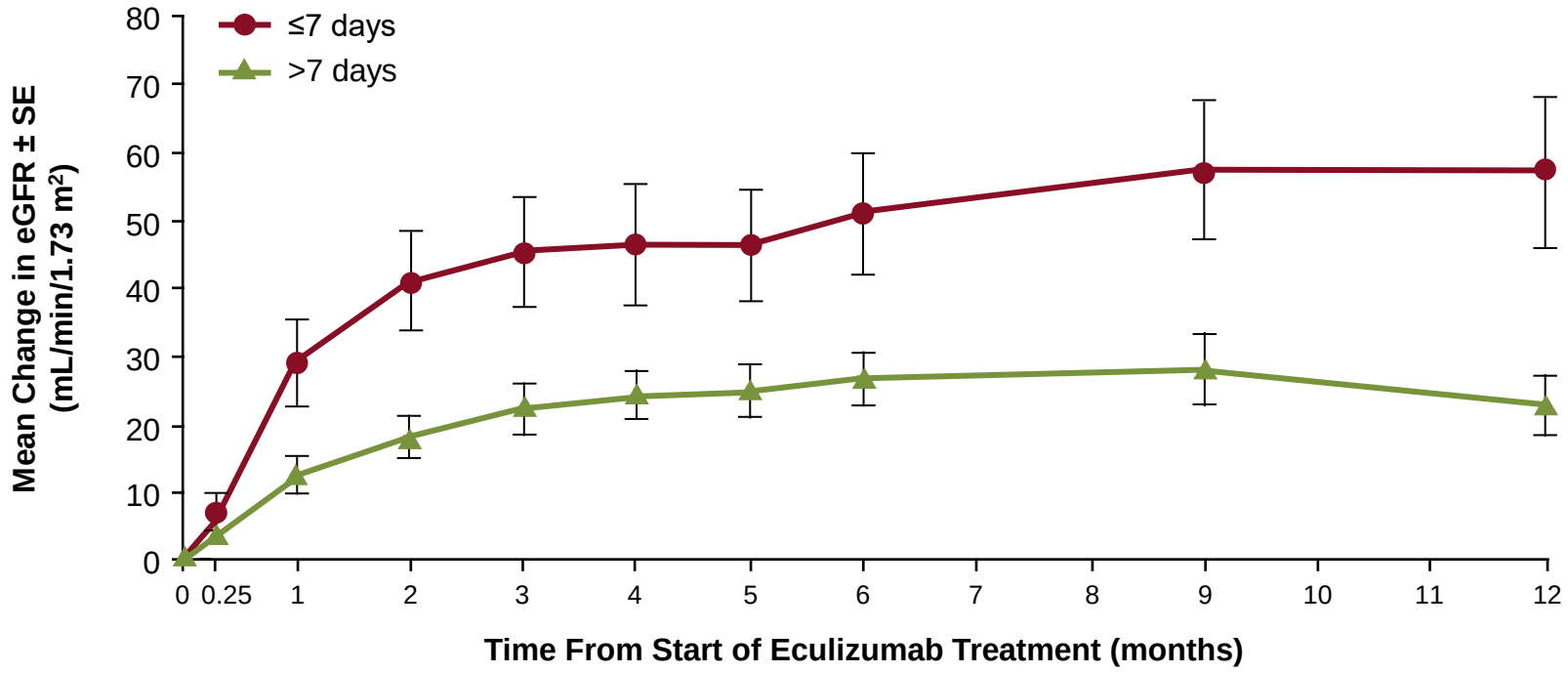
Figure 1 – eGFR at last follow-up of all patients treated with eculizumab who stopped dialysis (Adult Group n= 7; Pediatric Group n=10))



Clinical Kidney Journal, in press

Earlier Eculizumab Initiation Leads to Improved Renal Recovery

- Retrospective analysis with pooled data from 4 prospective clinical studies
- Evaluated changes in eGFR in patients initiating eculizumab ≤7 days or >7 days after onset of last TMA manifestation



Patients (N)												
Treatment Initiated in	≤7 days	21	20	18	20	20	19	19	20	17		14
	>7 days	76	74	69	74	74	75	72	74	60		54

eGFR, estimated glomerular filtration rate; SE, standard error; TMA, thrombotic microangiopathy.

Vande Walle et al. *J Nephrol*. 2017;30(1):127-34

Eculizumab decreases progression to End Stage Renal Disease when compared to historical treatment in Adults and Children

	Children			Adults				
	Pre-eculizumab era		Eculizumab	Pre-eculizumab era		Eculizumab		
	French cohort[2] (n=89)	Italian cohort[3] (n=149)	Trial 3[139,140] (n=22)	French Cohort[2] (n=89)	Italian cohort[3] (n=149)	Trial 1[141,142] (n=17)	Trial 2[141,142] (n=20)	Trial 4[143,144] (n=41)
First episode	16%	..	..	46%	..	..	..	..
6-month follow-up	..	..	9%	..	..	6%	10%	15%
1-year follow-up	29%	..	9%	56%	..	6%	10%	15%
2-year follow-up	..	..	9%	..	..	12%	10%	6-15%
3-year follow-up	..	48%	..	..	67%	..	..	..
5-year follow-up	36%	..	..	64%	..	..	..	..

30-50%

56-67%

For a detailed table legend see the appendix (pp 27,28). HUS=haemolytic uraemic syndrome.

Table 2: **Percentage of patients with atypical HUS who progressed to end-stage renal disease or who died in four prospective trials of eculizumab compared with the Italian and French registries of the pre-eculizumab era**

Fakhouri et al. Lancet, Feb 2017

Safety Profile of Eculizumab in the 100 Trial Patients With aHUS[1-4]

- No unexpected safety signals
- Most TEAEs were mild/moderate
- 1 death deemed unrelated to eculizumab (adult)
- 2 meningococcal infections (adults; both recovered)
- 1 patient had low positive values for human anti-human antibodies to eculizumab (pediatric)
  - Overall, there has been no observed correlation of antibody development to clinical response or adverse events
- Adverse events were reported with less frequency over time from week 26 to the 2-year update in studies C08-002 and C08-003

aHUS, atypical hemolytic uremic syndrome; TEAEs, treatment-emergent adverse events.

1. Legendre et al. *N Engl J Med*. 2013;369(14):1379-80. 2. Licht et al. *Kidney Int*. 2015;87(5):1061-73. 3. Greenbaum et al. *Kidney Int*. 2016;89(3):701-11.
4. Fakhouri et al. *Am J Kidney Dis*. 2016;68(1):84-93.

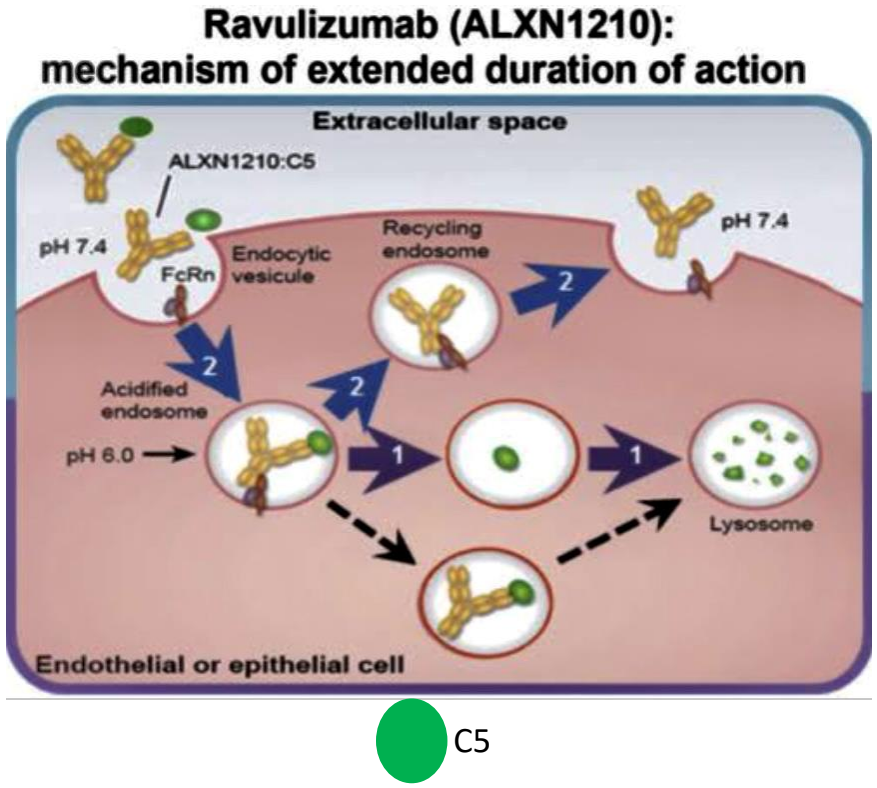
Subgroups of Patients and Specific Topics

Group/Topic	Available Evidence	Results
Pediatric[1]	Clinical Trial[2] and Case Reports	• n=38 Case Reports • **Hematologic response to eculizumab ≈100%** • **Kidney response to eculizumab ≈100%**
Adults[1]	Clinical Trial[3] and Case Reports	• n=39 Case Reports • **Hematologic response to eculizumab ≈90%** • **Kidney response to eculizumab ≈56%**
Lack of response	Case Reports	• Cobalamin C metabolism disease[4] • Mutation c.2654G→A in C5 in patients with PNH[5]
Monitoring	Ex vivo Assays	• Biomarkers of AP activity Noris et al[6] • Not agreed upon

AP, alternative pathway; PNH, paroxysmal nocturnal hemoglobinuria.

1. Palma, Langman. *J Blood Med*. 2016;7:39-72. 2. Greenbaum et al. *Kidney Int*. 2016;89(3):701-11. 3. Fakhouri et al. *Am J Kidney Dis*. 2016;68(1):84-93. 4. Cornec-Le Gall et al. *Am J Kidney Dis*. 2014;63:119-23. 5. Nishimura et al. *N Engl J Med*. 2014;370:632-39. 6. Noris et al. *Blood*. 2014;124:1715-26.

Ravulizumab (Ultomiris)



- Ravulizumab differs from eculizumab by the substitution of 4 amino acids, which alters the pharmacokinetics and pharmacodynamics of the molecule
- Mechanistically, these amino acid substitutions promote endosomal dissociation of the ravulizumab-C5 complex and result in lysosomal degradation of C5, while allowing recycling of ravulizumab to the vascular space due to enhanced affinity of ravulizumab for the neonatal Fc receptor (FcRn)
- These modifications resulted in a novel antibody against C5 with **a terminal half-life that is 4 times longer than that of eculizumab**.
- The molecule is thus designed to provide highly specific C5 inhibition over extended dosing intervals.

Roth A *et al*. *Blood Adv.* 2018

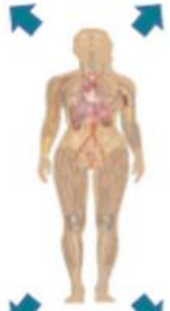
The long-acting C5 inhibitor, Ravulizumab, is effective and safe in adult patients with atypical hemolytic uremic syndrome naive to complement inhibitor treatment

Who was tested?

56 patients with aHUS

Reduced hemoglobin

Platelets <150 x 10^9/L



Increased Scr

Increased LDH

All patients with acute TMA

88% with eGFR <29

52% on dialysis

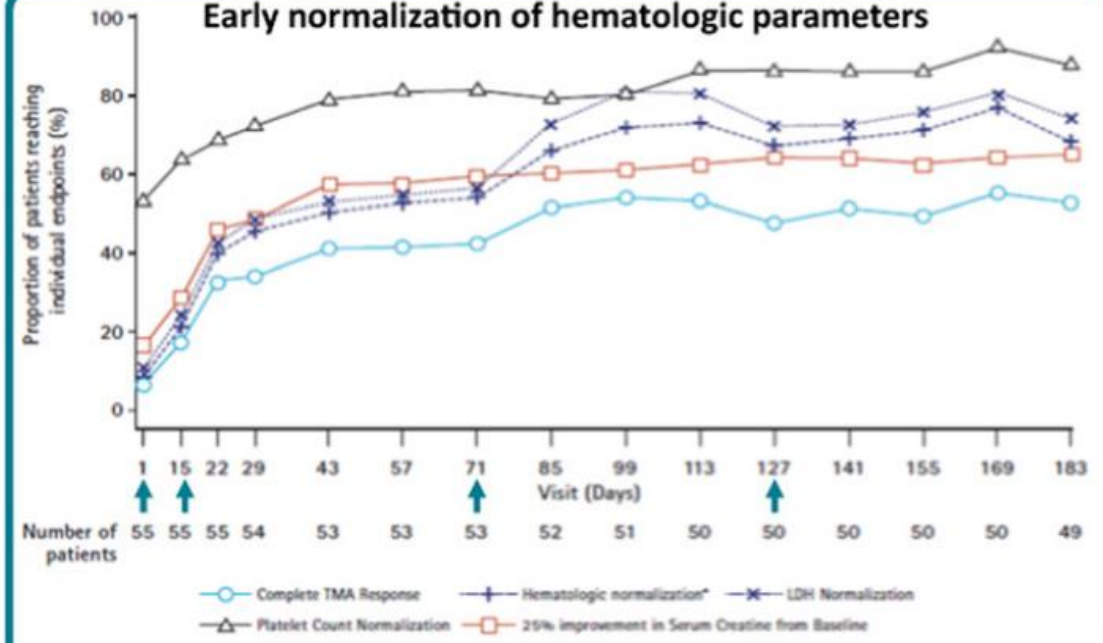
What was done?

Ravulizumab induction dose — Day 1

Ravulizumab maintenance doses — Day 15, Day 71, Day 127

Complete TMA response was achieved in 54% of patients and 59% of patients on dialysis at baseline came off dialysis by Day 183

Early normalization of hematologic parameters



*Hematologic normalization is defined as normalization of both platelet count (≥150 x 109/L) and LDH (≤246 U/L).

Safety

No unexpected adverse events identified

No meningococcal infections observed

Four deaths unrelated to ravulizumab treatment

CONCLUSIONS:

- Ravulizumab provided immediate and complete inhibition of C5 (defined as free C5 <0.5µg) sustained over the 8-week dosing interval
- Substantial improvement was achieved in platelet count, LDH, serum creatinine and renal function and no unexpected adverse events were identified
- The results of this study support the use of ravulizumab at 8-weekly dosing intervals in adult patients with aHUS

ISN kidney INTERNATIONAL
OFFICIAL JOURNAL OF THE INTERNATIONAL SOCIETY OF NEPHROLOGY

Rondeau, 2019

The long-acting C5 inhibitor, ravulizumab, is efficacious and safe in pediatric patients with atypical hemolytic uremic syndrome previously treated with eculizumab

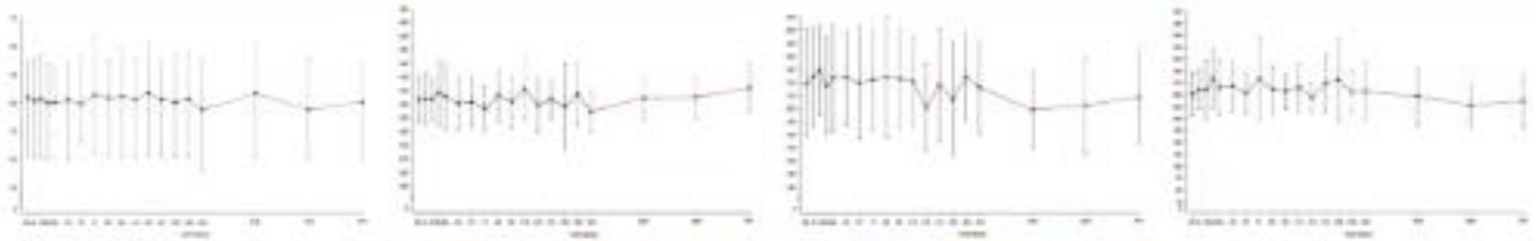
Graphical Abstract

HYPOTHESIS: Ravulizumab is safe and efficacious in patients switching from eculizumab treatment

DESIGN & OUTCOMES:

Ravulizumab Efficacy following Switch from Eculizumab*

eGFR mL/min/1.73m²	Hemoglobin g/L	Lactate Dehydrogenase U/L	Platelet Count X10³/L



*Eculizumab discontinued 2-weeks pre-baseline

Ravulizumab Safety following Switch from Eculizumab*

	Overall (N = 10)	
	n (%)	Events
Any AE	10 (100)	66
Any serious AE	1 (10.0)	5
TESAEs resulting in drug discontinuation	0	0
TEAEs resulting in trial discontinuation	0	0
TEAEs during trial drug infusion	2 (20.0)	4
TESAEs during trial drug infusion	0	0
Treatment-related AEs	2 (20.0)	4
Meningococcal infections	0	0

CONCLUSION: Treatment with ravulizumab in pediatric patients with aHUS previously treated with eculizumab resulted in stable renal and hematologic parameters, with no unexpected safety concerns when administered every 4–8 weeks

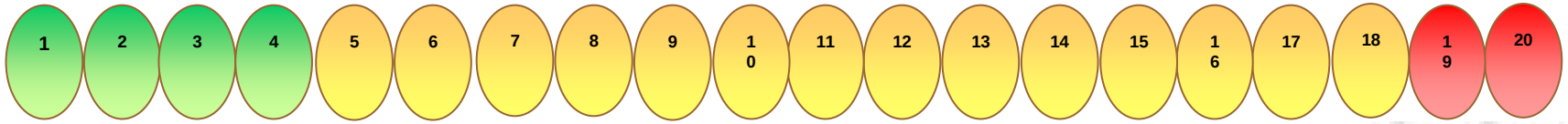
Tanaka et al. 2019

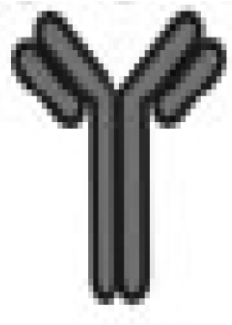
Pediatric Nephrology
Journal of the
IPNA International Pediatric Nephrology Association

Autoantibodies to Factor H in aHUS Patients

Complement regulation

Ligand recognition





Anti-factor H IgG antibodies

- HUS-associated factor H autoantibodies (described mainly in children) mimic the effect of C-terminal factor H mutations, as they inhibit the regulatory function of factor H at cell surfaces by blocking its C-terminal recognition region
- Associated with CFHR1,3 mutations.

Dragon-Durey et al., *JASN,* 2005*;* Jozsi et al., *Blood,* 2007.
Hofer, *Clin J Am Soc Nephrol* 8: 407–415, 2013

aHUS in India

Atypical Hemolytic Uremic Syndrome: A Meta-Analysis of Case Reports Confirms the Prevalence of Genetic Mutations and the Shift of Treatment Regimens

259 patients with aHUS

- 54% at least 1 functional mutation in a complement gene
- 21% Factor H
- 19% anti-Factor H antibody

Krishnappa V. et al Therapeutic Apheresis and Dialysis, 2017
Raina R et al. Cells, 2021

Clinical and Immunological Profile of Anti-factor H Antibody Associated Atypical Hemolytic Uremic Syndrome: A Nationwide Database

781 patients < 18 y aHUS

- 55.8 % anti-Factor H antibody
- Titers correlate with outcome
- Impact on treatment

Puraswani M et al. Frontiers in Immunology, 2019

Anti-factor H antibody and its role in atypical hemolytic uremic syndrome

Geographical Differences

- 5-25% in Europe
- ~50% in South Asia
- May have therapeutic implications

Raina R et al. Frontiers in Immunology, 2022

Anti-complement Therapy – Initiation and Amplification

Clinical Trials in Brazil

Narsoplimab (anti MASP2)

- C3G
- Lupus Neph
- MN
- IgAN

Pegcetacoplan (C3 Inhibitor)

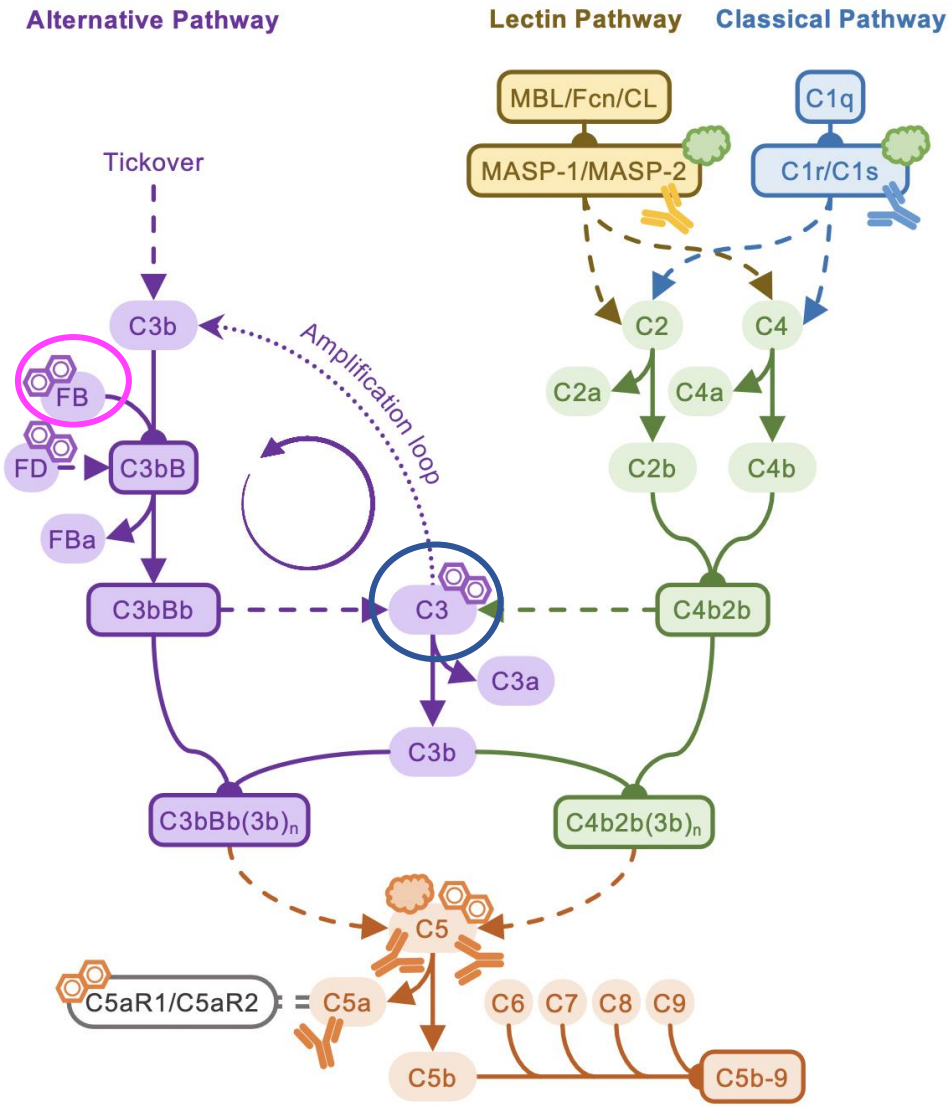
- C3G
- IC-MPGN
- Recurrence after Kidney Tx

Iptacopan (Factor B inhibitor)

- C3G
- aHUS
- IgAN

Danicopan (Factor D inhibitor)

- Paroxysmal Nocturnal Hemoglobinuria (PNH)



Initiation Inhibitors

- **Narsoplimab** (anti-MASP2)
- **Sutimlimab** (anti-C1s)
- **C1 esterase inhibitors**

Amplification Inhibitors

- **Pegcetacoplan** (C3 inhibitor)
- **Iptacopan** (FB inhibitor)
- **Danicopan** (FD inhibitor)

Effector Inhibitors

- **Eculizumab** (anti-C5)
- **Ravulizumab** (anti-C5)
- **Crovalimab** (anti-C5)
- **Nomacopan** (C5 inhibitor)
- **Zilucoplan** (C5 inhibitor)
- **Zimura** (C5 inhibitor)
- **Vilobelimab** (anti-C5a)
- **Avacopan** (C5aR1 antagonist)

ClinicalTrials.gov Search Results 09/21/2022

Richard B.P., Daniel R. Tipping the balance: intricate roles of the complement system in disease and therapy. Seminars in Immunopathology (2021)

Courtesy of Dr. Silvana Miranda

Anti-complement Therapy – Effector Inhibitors

Clinical Trials in Brazil

Ravulizumab (anti C5)

- aHUS
- PNH

Crovalimab (anti C5)

- aHUS
- PNH
- Sickle cell disease

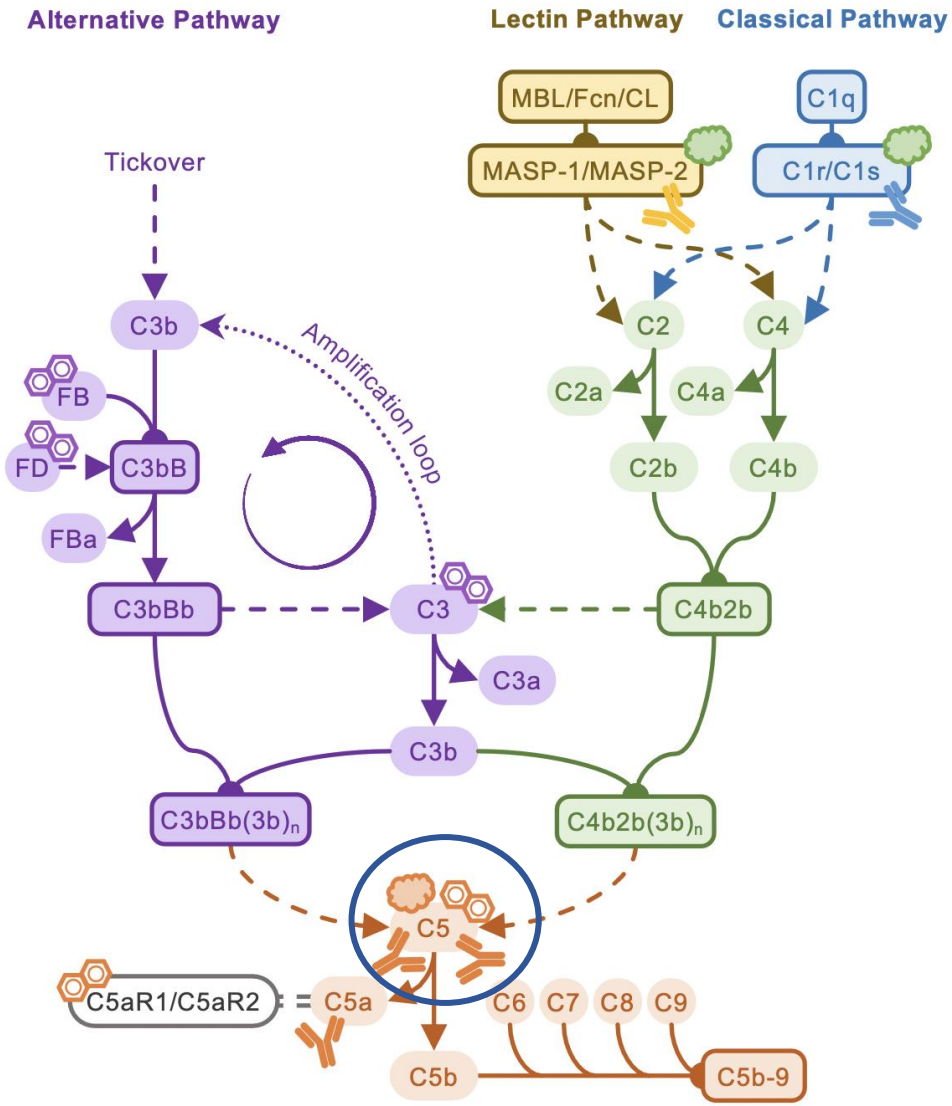
Nomacopan (inibidor C5)

- TMA

Vilobelimab (anti C5a)

- Microscopic poliangiitis*
- Granulomatous poliangiitis*

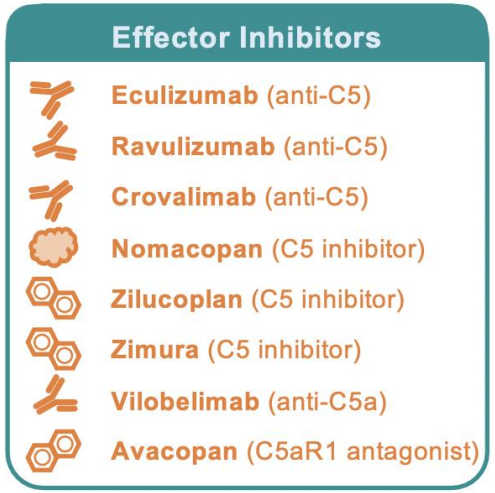
* complete



Avacopan (antagonista C5aR1)

- C3G*
- Vasculitis*
- IgAN*
- aHUS*

* complete



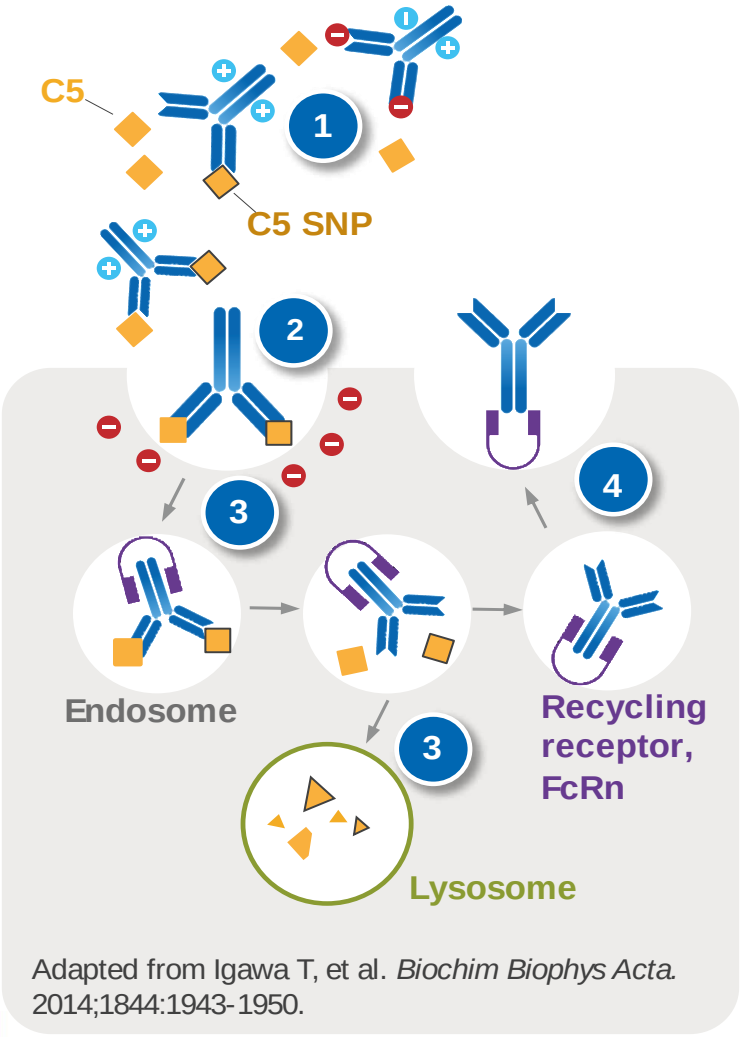
ClinicalTrials.gov Search Results 09/21/2022

Richard B.P., Daniel R. Tipping the balance: intricate roles of the complement system in disease and therapy. Seminars in Immunopathology (2021)

Courtesy of Dr. Silvana Miranda

Crovalimab – Mechanism of Action

Anno VII
Cadernos de Patologia Renal
COMPLEMENTO E DOENÇAS RENAIS.



Adapted from Igawa T, et al. *Biochim Biophys Acta.* 2014;1844:1943-1950.

1 High-affinity binding

- Crovalimab is engineered to **optimize binding of C5** in the plasma through affinity maturation.

2 Preferential antibody uptake (proprietary innovative engineering)

- Crovalimab charge is engineered to favor **increased endocytosis/ recycling of antibody bound to two C5 molecules.**

3 Acid-sensitive binding and antigen degradation

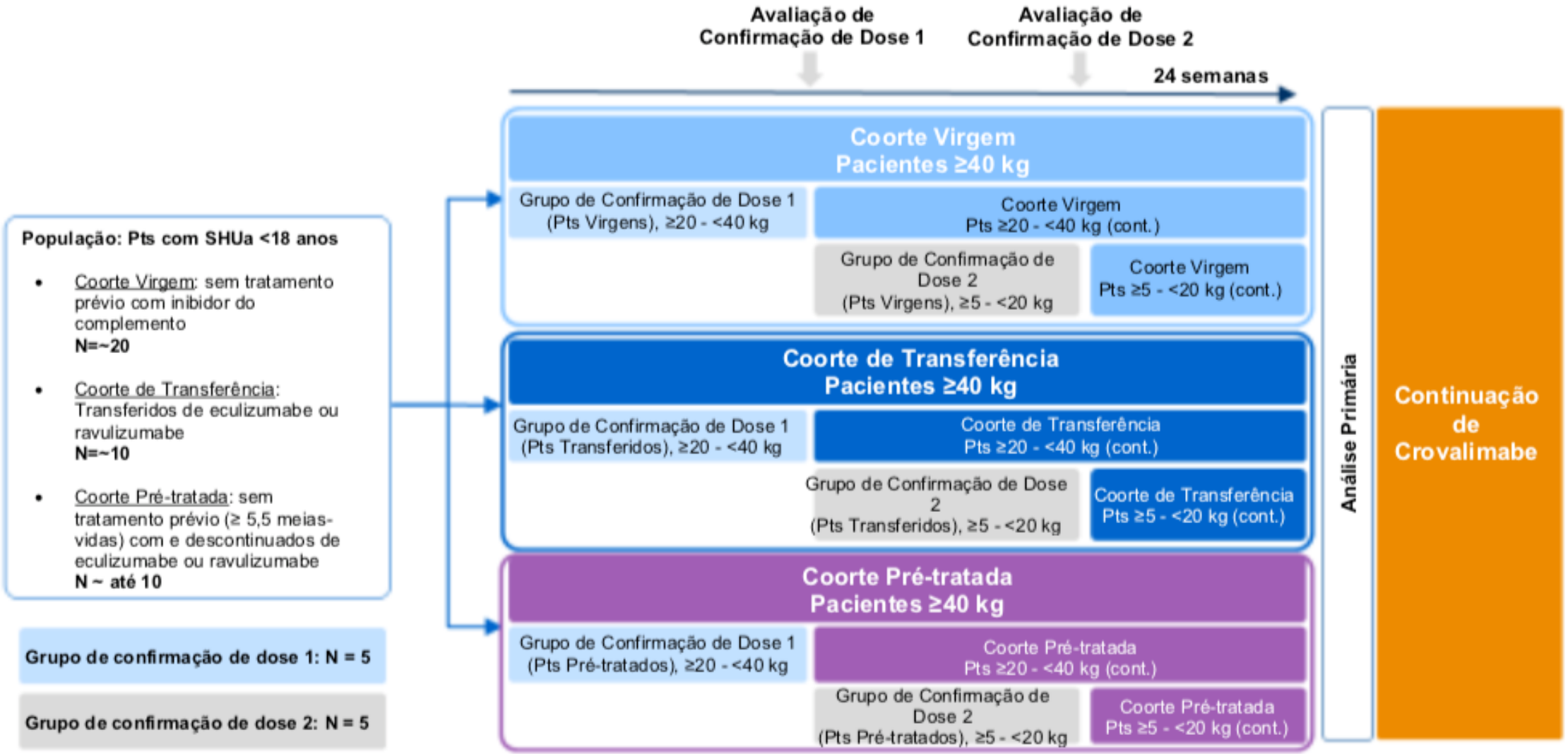
- Crovalimab is engineered to **dissociate from C5 in the acidic pH** of the endosome.
- C5 is degraded in the lysosome.

4 Antibody recycling by FcRn engineering

- Crovalimab is **recycled** and returned to the plasma through FcRn to **extend its half-life**.
- **Binding to FcRn** is optimized.

SNP, single-nucleotide polymorphism.

Courtesy of Dr. MH Vaisbich

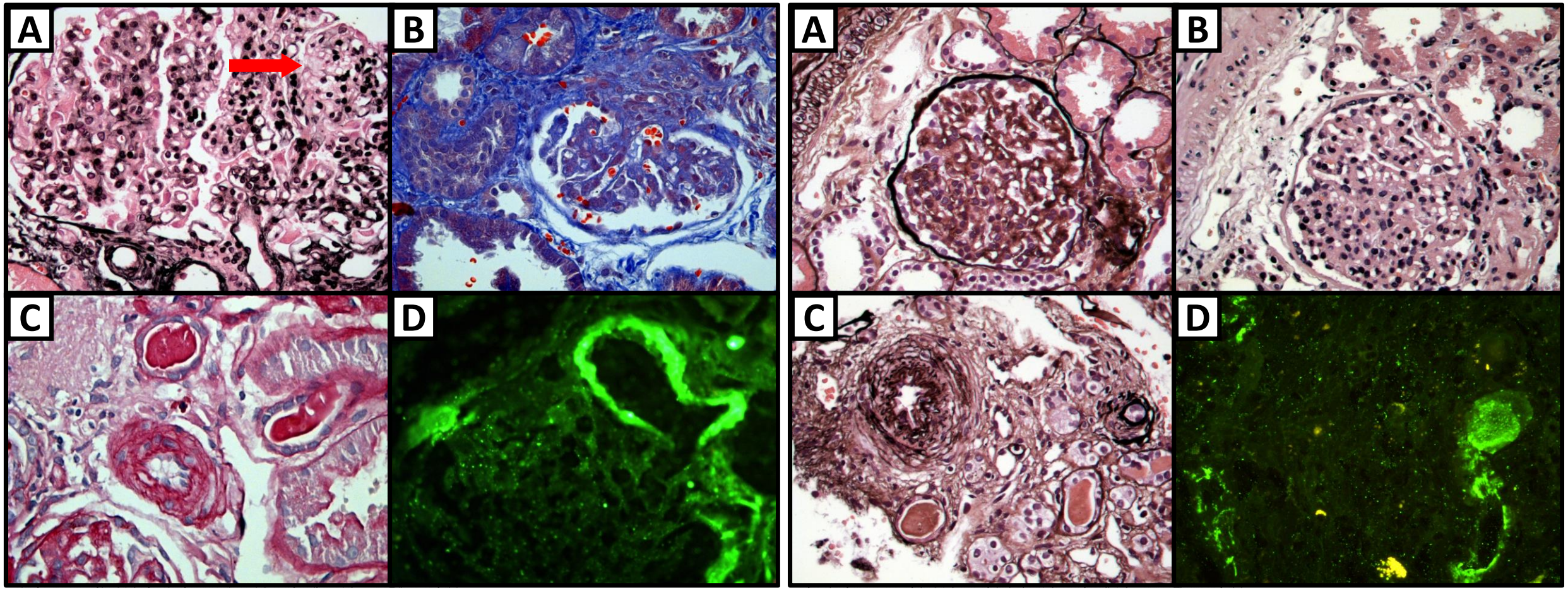


SHUa = síndrome hemolítica urêmica atípica; cont. = continuação; N = número de pacientes; Pts = pacientes.

OBSERVAÇÃO: O número total de pacientes em cada braço inclui os pacientes avaliáveis dos Grupos de confirmação de dose.

Courtesy of Dr. MH Vaisbich

17 yo, male, 3 episodes of TMA (1st at 6 months)



2016 → 2018

Eculizumab

De Holanda et al, *Clinical Kidney Journal*, 2019

17 yo, male, 3 episodes of TMA (1st at 6 months)

DGKe – p.356AspLysfs*6 (c.1069_1071del)

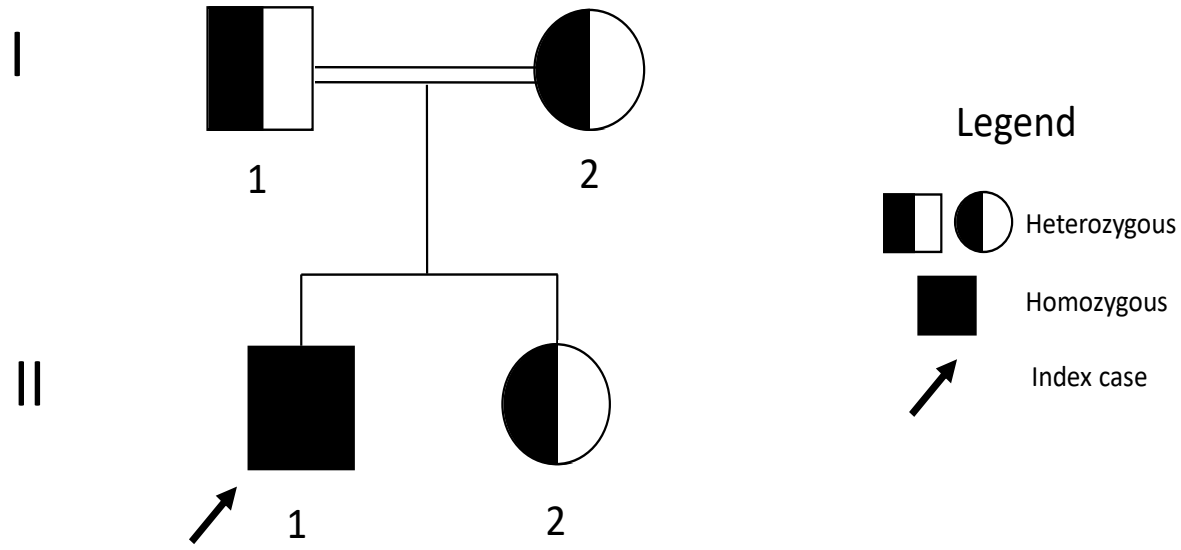


Figure 3. **Family pedigree.** Index case (II-1) developed first symptoms of aHUS at six months. His genetic diagnosis prompted the analysis of the family members, uncovering the presence of the variant p.356AspLysfs*6 (c.1069_1071del) in all of them.

De Holanda et al, *Clinical Kidney Journal*, 2019

Archives of Pediatrics
Palma LMP, et al. Arch Pediatr 8: 246
www.doi.org/10.29011/2575-825X.100246
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Case Report

Diacylglycerol kinase Epsilon (DGKe) Nephropathy: Rare Cause of Thrombotic Microangiopathy

Lilian Monteiro Pereira Palma[1*], Nathalia Montibeler Ferreira[2], Vera Maria Santoro Belangero[3], Sumara Zuanazi Pinto Rigatto[3], João Bosco Pesquero[4], Caio Perez Gomes[4]

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[2]Pediatria, Departamento de Pediatria, Universidade Estadual de Campinas, Brazil

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[4]Center for Research and Diagnosis of Genetic Diseases - Escola Paulista de Medicina - Universidade Federal de São Paulo, Brazil

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Citation: Pereira Palma LM, Ferreira NM, Santoro Belangero VM, Pinto Rigatto SZ, Pesquero JB, et al. (2023) Diacylglycerol kinase epsilon (DGKe) Nephropathy: Rare cause of Thrombotic Microangiopathy. Arch Pediatr 8: 246. DOI: 10.29011/2575-825X.100246

Received Date: 14 February 2023; **Accepted Date:** 20 February 2023; **Published Date:** 24 February 2023.

○ Complete blood count ○ Peripheral smear (schistocytes) ○ Reticulocyte count ○ Bilirrubin ○ Lactic Dehydrogenase ○ Haptoglobin ○ Coagulation tests ○ Direct antiglobulin test (Coombs test)
○ Anamnesis and family history, consaguinity ○ Complete physical examination with Blood Pressure and Neurological Assessment ○ Kidney function ○ Urinalysis ○ Proteinúria (24 h and/or spot urine protein/creatinine ratio) ○ Hepatic enzymes ○ Pancreatic enzymes ○ Blood gases ○ Troponin, EKG, echocardiography ○ Ophtalmologic evaluation, fundoscopy
○ Stool culture and shigatoxin test ○ Activity and inhibitor to ADAMTS13 ○ Complement, blood levels ○ Cultures: blood, urine, cephalorachidic liquid, abscesses ○ Latex for *Streptococcus pneumoniae* ○ Tests for H1N1, COVID19, viral panels ○ According to epidemiology: dengue, leptospirosis, brucelosis ○ Homocystein and vitamin B12 blood levels ○ Aminoacid chromatography (blood, urine) ○ Genetic Tests: Hemolytic Uremic Syndrome panel or Whole Exome Sequencing ○ Anti-Factor H antibody ○ Kidney biospy in selected cases

Table 1: Checklist of exams and procedures to detect Thrombotic Microangiopathy, evaluate organ damage and define underlying etiology; Legend: EKG: electrocardiogram; H1N1: influenza vírus; COVID19: coronavírus 19.

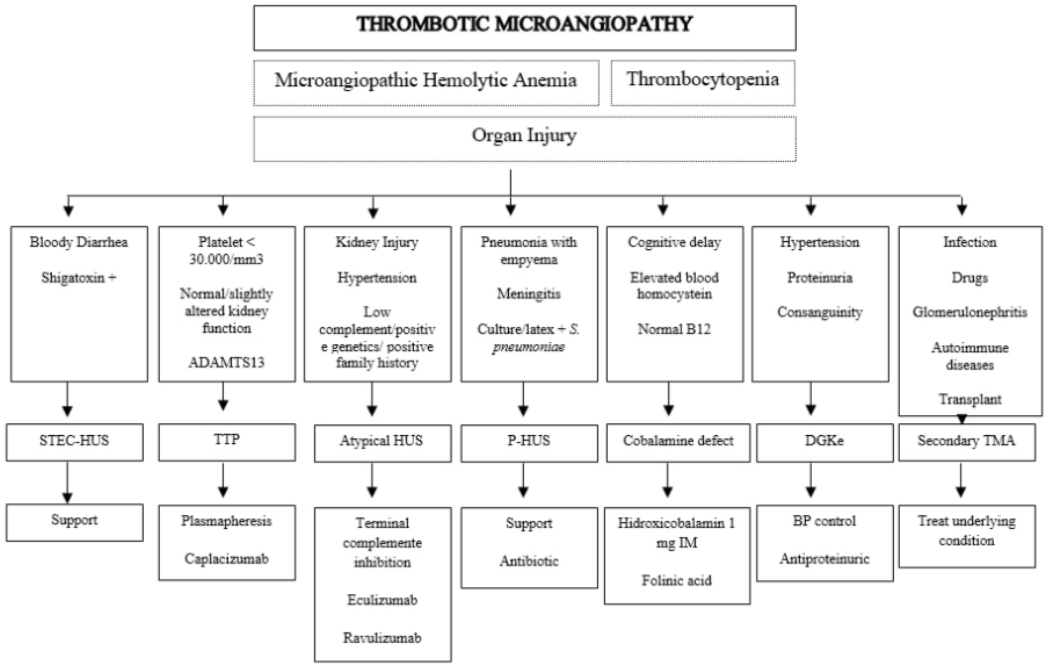
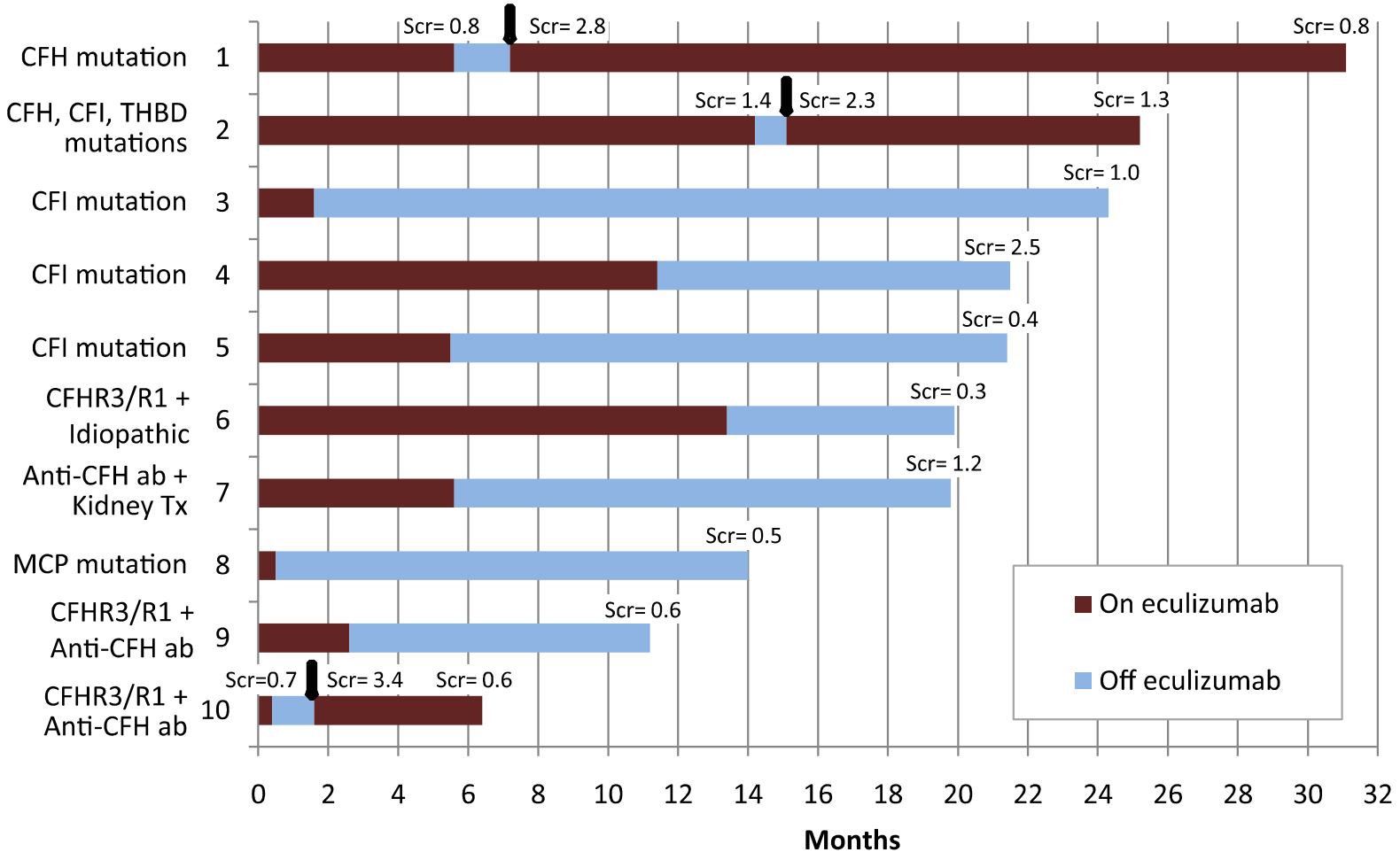


Figure 1: Definition of Thrombotic Microangiopathy and main causes in the pediatric population (clinical presentation, diagnosis and treatment).

Palma L et al, *Archives of Ped*, 2023

22 pts treated – 10 stopped

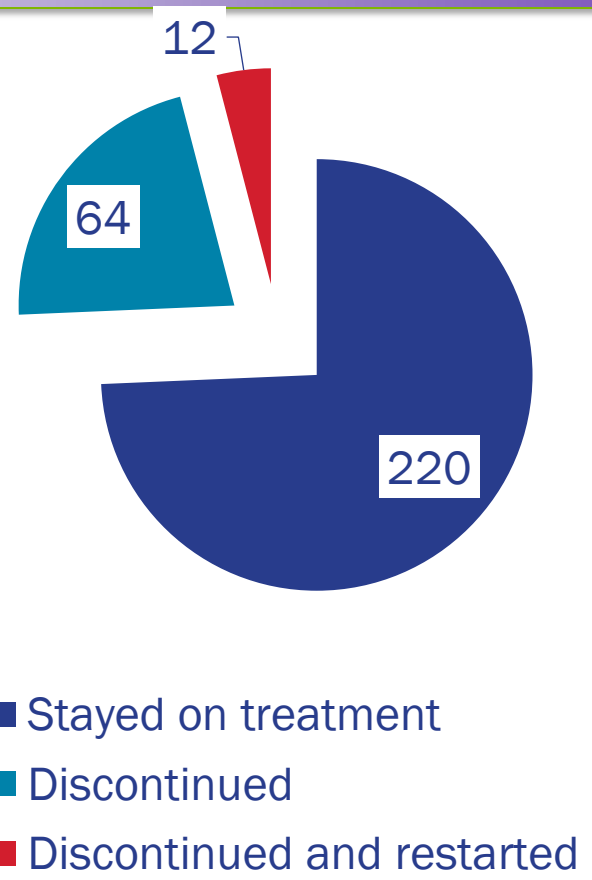


Ardissino G et al. AJKD, 2014

Discontinuations and restarts in the global ahus registry

3. The global aHUS Registry

- 28 (24%) patients aged <18 years discontinued, of whom 7(25%) restarted eculizumab treatment
- 48 (27%) adult patients discontinued and 5 (10%) subsequently restarted eculizumab treatment



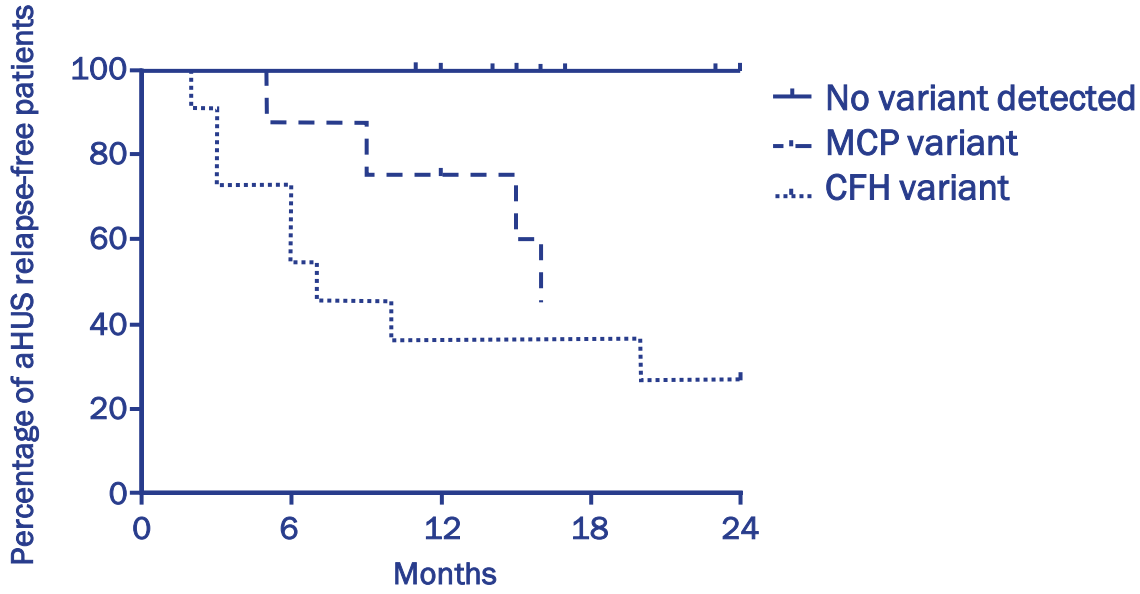
Macia M et al. *CKJ*. 2016

Pathogenic Variants in Complement Genes and Risk of Atypical Hemolytic Uremic Syndrome Relapse After Eculizumab Discontinuation

Fadi Fakhouri, Marc Fila, François Provôt, Yahsou Delmas, Christelle Barbet, Valérie Châtelet, Cédric Rafat, Mathilde Cailliez, Julien Hogan, Aude Servais, Alexandre Karras, Raifah Makdassi, Feriell Louillet, Jean-Philippe Coindre, Eric Rondeau, Chantal Loirat, and Véronique Frémeaux-Bacchi

CJASN. 2016. [Epub ahead of print]
doi: 10.2215/CJN.06440616

Risk of “Relapse” After Discontinuation by Identified Variant



Number at risk						
CFH	11	7	5	2	0	OR=80 (95% CI, 3.7–1737; *P*<0.001)
MCP	8	7	5	3	1	OR=25 (95% CI, 1.1–594; *P*=0.001)
No variant	16	15	14	8	6	Reference

Kaplan–Meier estimate of atypical hemolytic uremic syndrome (aHUS) relapse-free survival after eculizumab discontinuation in patients with pathogenic variants in complement factor H (CFH) or membrane cofactor protein (MCP) or no detected pathogenic variants in complement genes.

Eculizumab discontinuation in children and adults with atypical haemolytic uremic syndrome: a prospective multicentric study

Prospective
55 patients

19 pediatric

36 adults

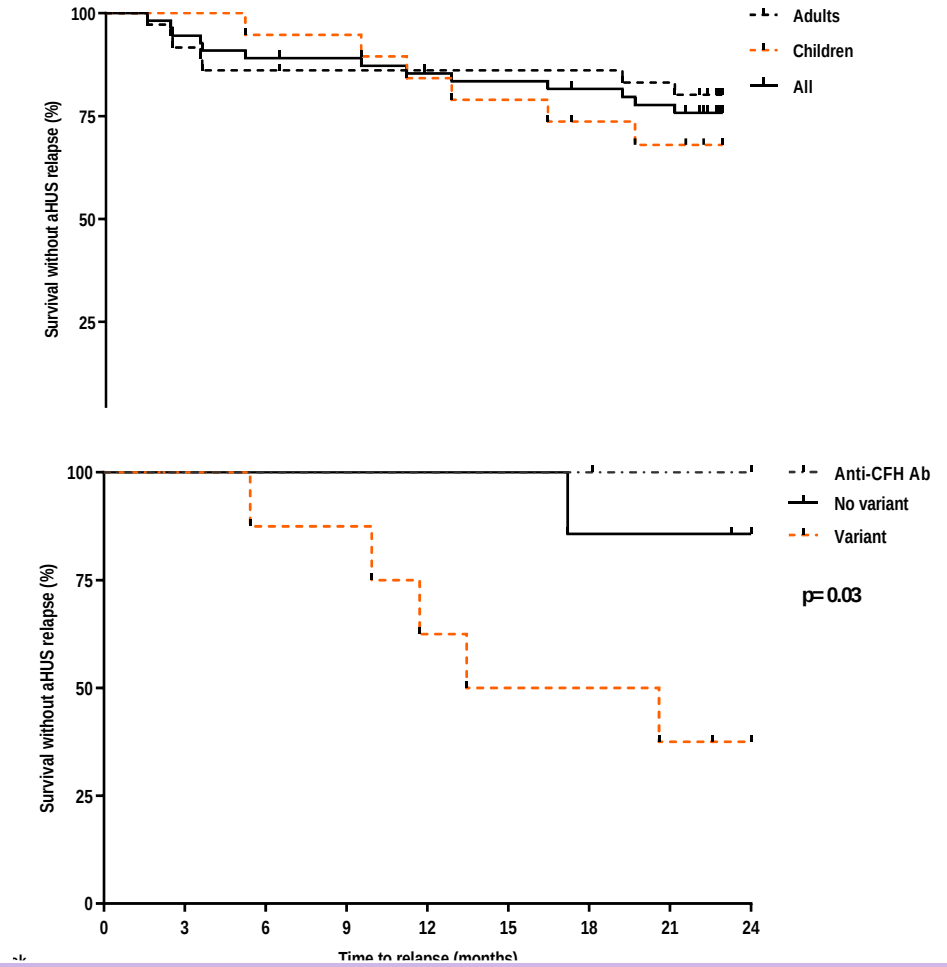
6 recurrences
(31%)

7 recurrences
(19%)

All patients with AKI on recurrence
2 progressive CKD

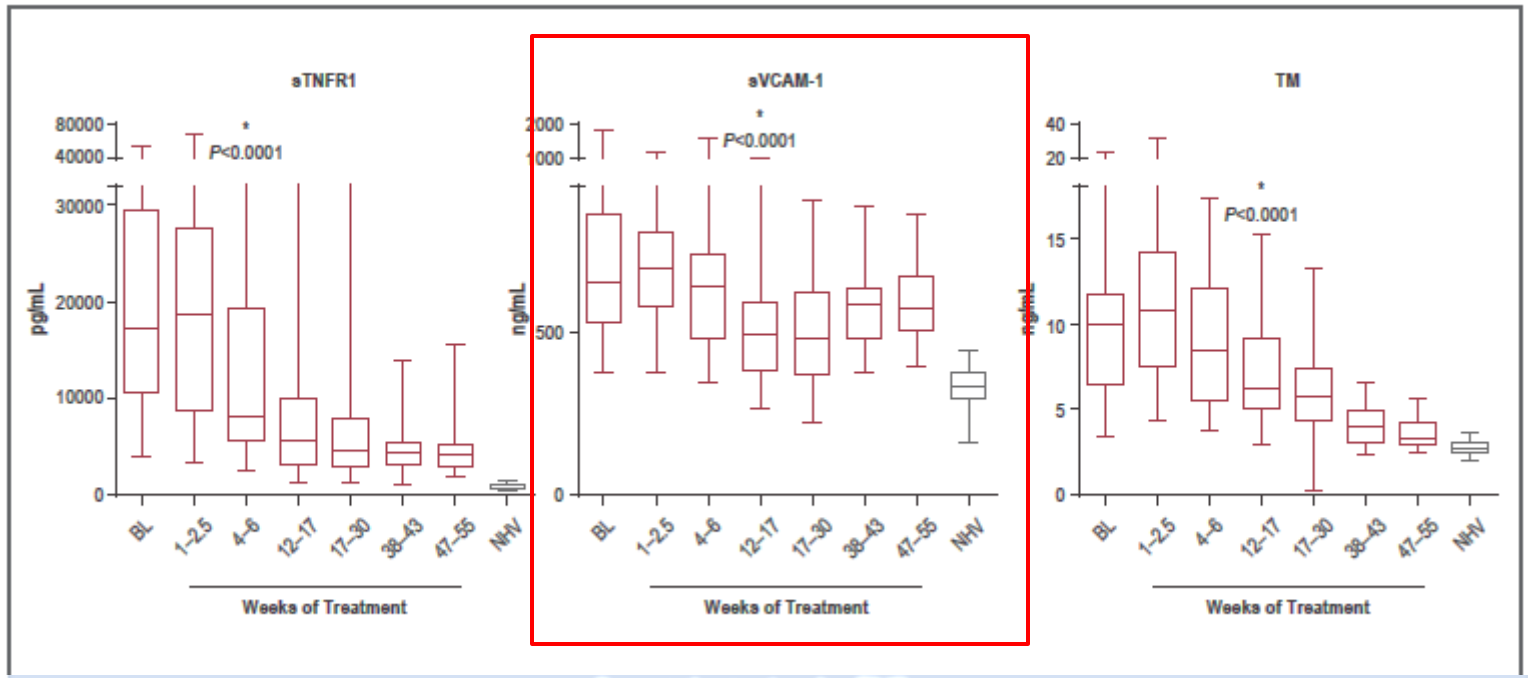
RISK FACTORS FOR RECURRENCE:

- Pathogenic variant
- Female
- **Elevated sC5b-9 at discontinuation**



Fakhouri F et al. Blood. 2021 May 6;137(18):2438-2449

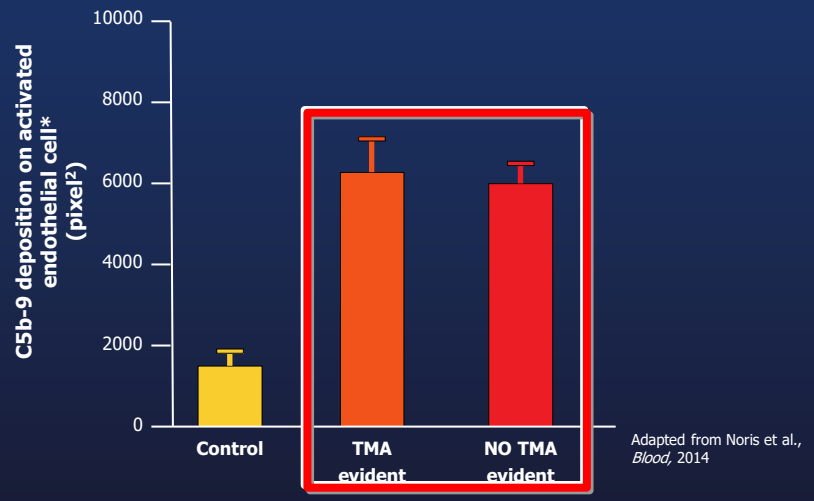
Eculizumab and biomarkers – pivotal studies



Inflammation	Soluble Tumor Necrosis Factor-1 (sTNFR1)	Surrogate marker for TNF-α TNF-α is pro-inflammatory; associated with vascular[13] and chronic renal inflammation and progression of renal failure[14-16] TNF-α upregulated by complement[17]
Endothelial Activation	Soluble Vascular Cell Adhesion Molecule (sVCAM-1)	Adhesion molecule released by activated endothelial cells Upregulated by TNF-α and terminal complement[18,19]
Endothelial Cell Damage	Thrombomodulin (TM)	Protective against thrombotic risk when membrane bound[20]; released by damaged endothelial cells[21] Upregulated by TNF-α[22]

Cofiell R *et al*. Blood, 2015: 125 (21)

In patients with aHUS, there is complement deposition on activated endothelial cells with or without overt TMA[1]



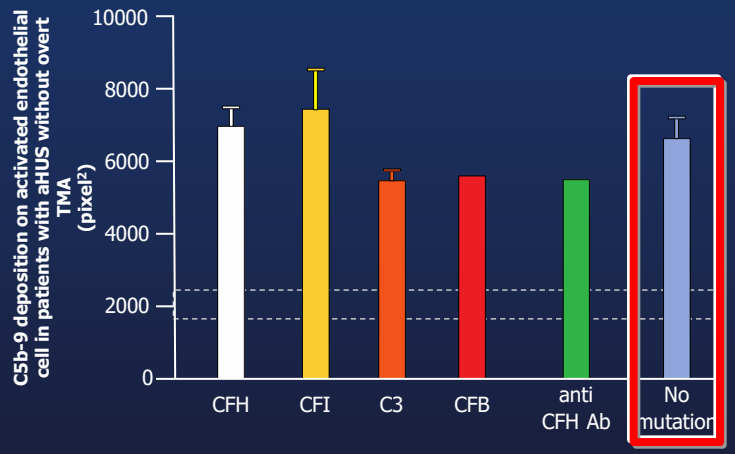
Adapted from Noris et al., *Blood,* 2014

*C5b-9 deposition on activated endothelial cell is a test of complement activation in patients with aHUS

[redacted]

Noris M *et al*, *Blood*, 2014c

In patients with aHUS, there is complement deposition on activated endothelial cells with or without mutations[1]

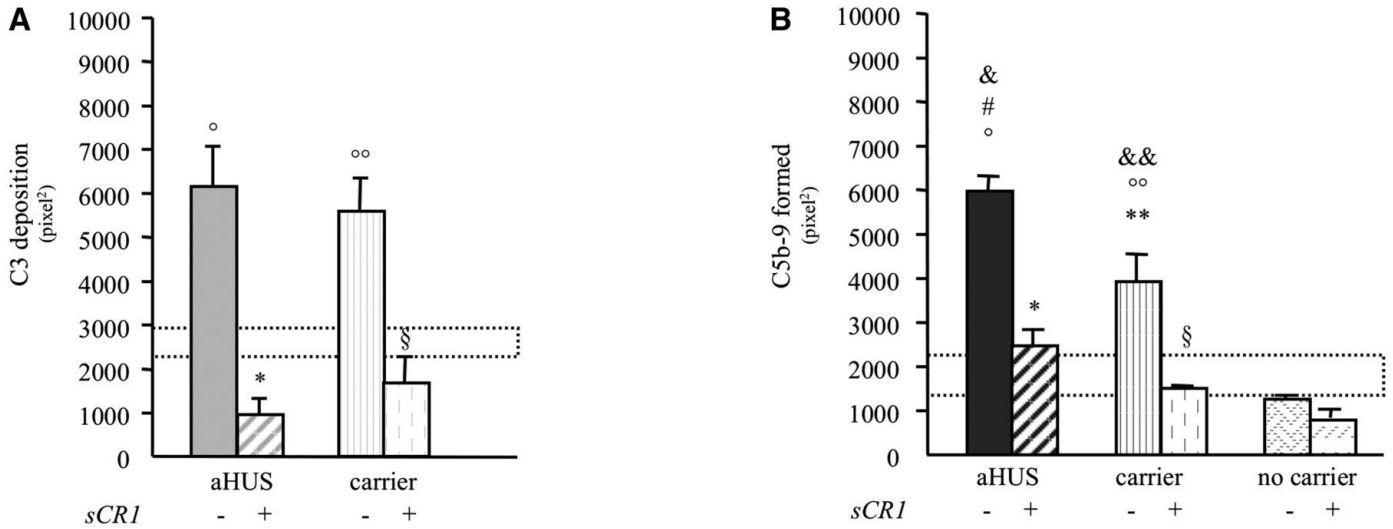


Adapted from Noris et al., *Blood,* 2014

*Dotted horizontal areas: Range of C5b-9 deposition induced by healthy control serum (mean+/-SE)

1. Noris M *et al, Blood*, 2014

Serum from healthy carriers of complement gene mutations induces C3 and C5b-9 deposition on ADP-activated microvascular endothelial cells (HMEC-1).

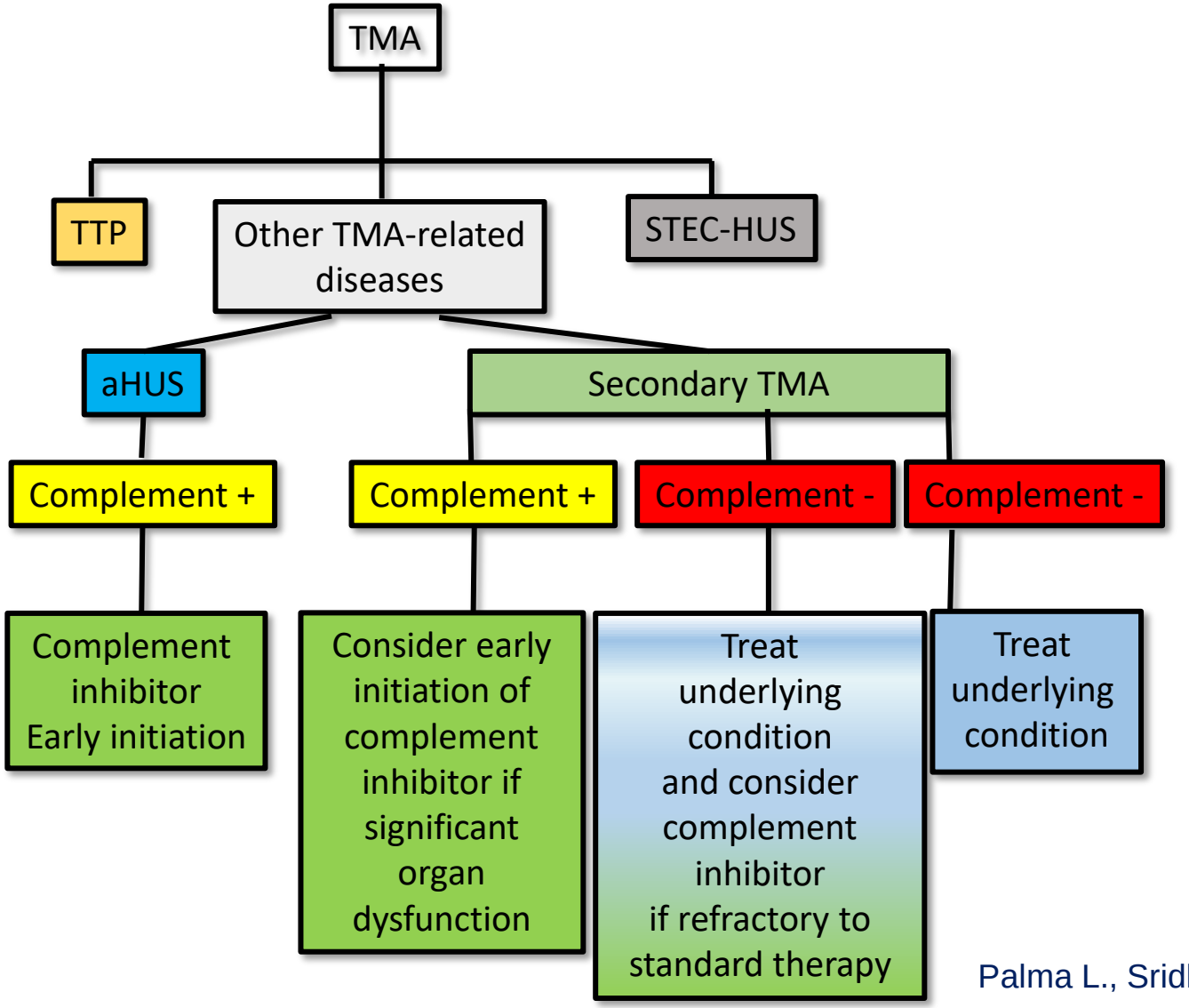


C

Patient No.	Mutations or anti CFH Ab	Degree of kinship	Complement parameters: plasma SC5b-9 (127-400 ng/ml)°	Endothelial C3 deposits £ (% of the control)	Endothelial C5b-9 deposits £ (% of the control)
Healthy mutation carrier 1	*CFH* - R1210C	brother patient 1	200	236% *	n.d.
Healthy mutation carrier 2	*CFH* -S1191L	sister patients 15/16	1158	189% *	478% *
Healthy mutation carrier 3	*CFH* -S1191L	mother patients 15/16	980	200% *	284% *
Healthy mutation carrier 4	*C3* - T140R	mother patient 29	n.a.	303% *	276% *
Healthy mutation carrier 5	*CFI* - G261D	father patient 24	609	313% *	305% *
Healthy mutation carrier 6	*CFH* - R78G	father patient 3	n.a.	n.d.	240% *
Healthy mutation carrier 7	*CFHR1/CFH* hybrid	son patient 23	242	n.d.	216% *
Healthy no carrier 1	no mut/no anti CFH Ab	mother patient 3	n.a.	n.d.	83%
Healthy no carrier 2	no mut/no anti CFH Ab	sister patient 3	n.a.	n.d.	76%
Healthy no carrier 3	no mut/no anti CFH Ab	mother patient 1	n.a.	n.d.	92%
Healthy no carrier 4	no mut/no anti CFH Ab	wife patient 23	194	n.d.	64%
Healthy no carrier 5	no mut/no anti CFH Ab	niece patient 1	n.a.	n.d.	83%
Healthy no carrier 6	no mut/no anti CFH Ab	mother patient 24	n.a.	n.d.	98%
Healthy no carrier 7	no mut/no anti CFH Ab	sister patients 15/16	n.a.	n.d.	102%

Marina Noris et al. Blood 2014;124:1715-1726

Approach to TMA management according to evidence of complement involvement



Complement +:

- **Genetics**: pathogenic/likely pathogenic variant or risk haplotype in alternative complement pathway genes
- **Antibody**: autoantibodies to complement factors (mainly anti-Factor H and anti-Factor B)
- **Functional Assays:** fluid vs. solid
- **Biopsy:** C4d, C5b-9?
- **Proteomics:** may be the answer to precision medicine in order to determine the complement pathway and burden

Palma L., Sridharan M., Sethi S. et al. *Kidney International Reports*, 2021

História clínica

- Sexo feminino, 18 anos, sem doenças prévias
- 1 mês antes da internação na Nefrologia apresentou febre, dor abdominal difusa, náusea, vômitos e disúria.
- UPA - Iniciou cefalexina por 14 dias – após o tratamento apresentou *rash* difuso, petéquias e oligúria.
- Emergência do HFB em 07/05/14 – trombocitopenia, anemia e IRA
- Iniciou hemodiálise

De Holanda, Palma et al. *Clinical Rheumatology*, 2017 Sep

Investigação e diagnóstico

- Hematúria e proteinúria
- Creatinina: 4,8 mg/dL; Ureia: 191 mg/dL
- Plaquetas: 50.000/mm^3
- Hemoglobina: 6,2 g/dL
- LDH 946 U/L
- Coombs negativo

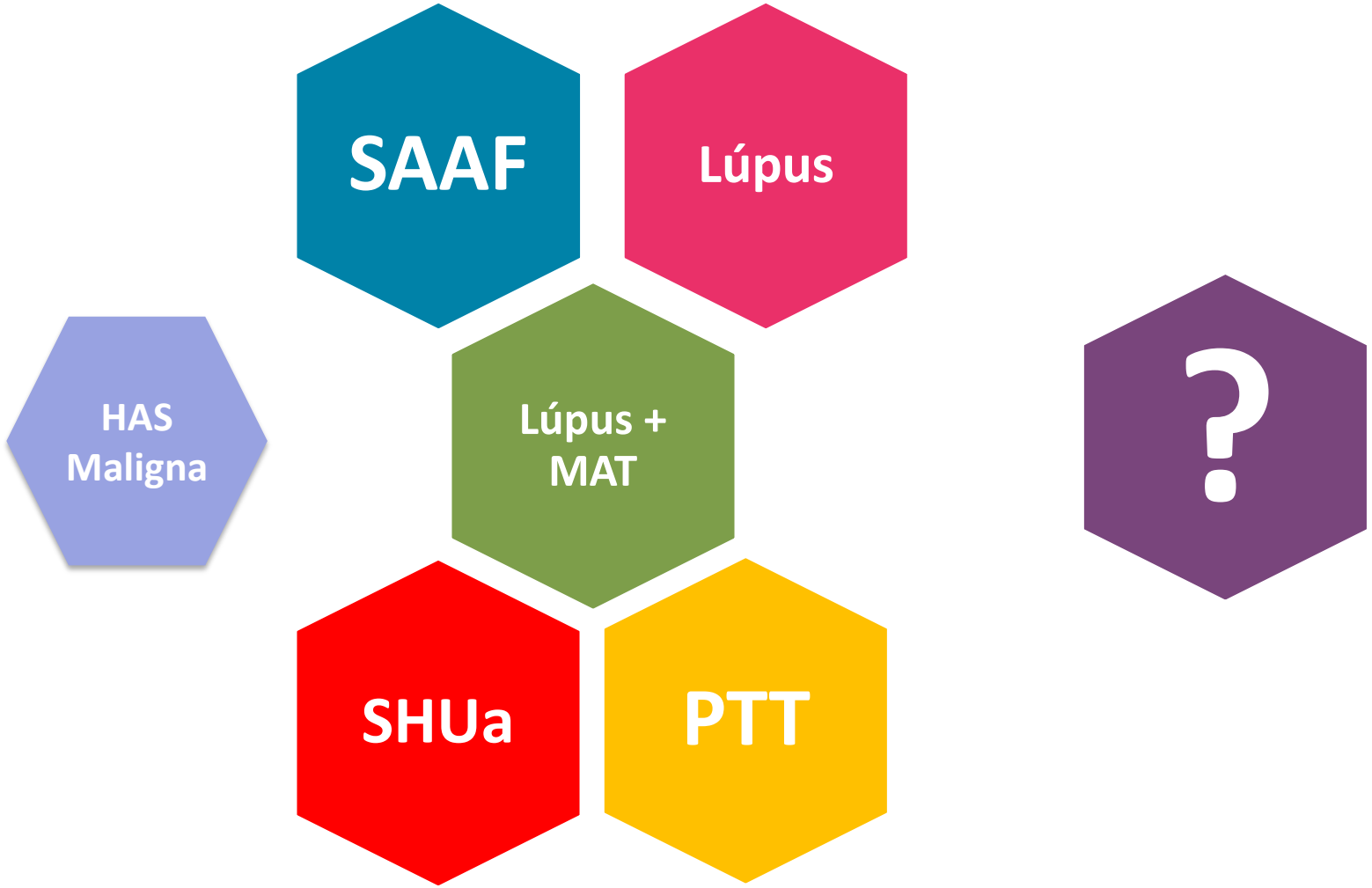
De Holanda, Palma et al. *Clinical Rheumatology*, 2017 Sep

Investigação e diagnóstico

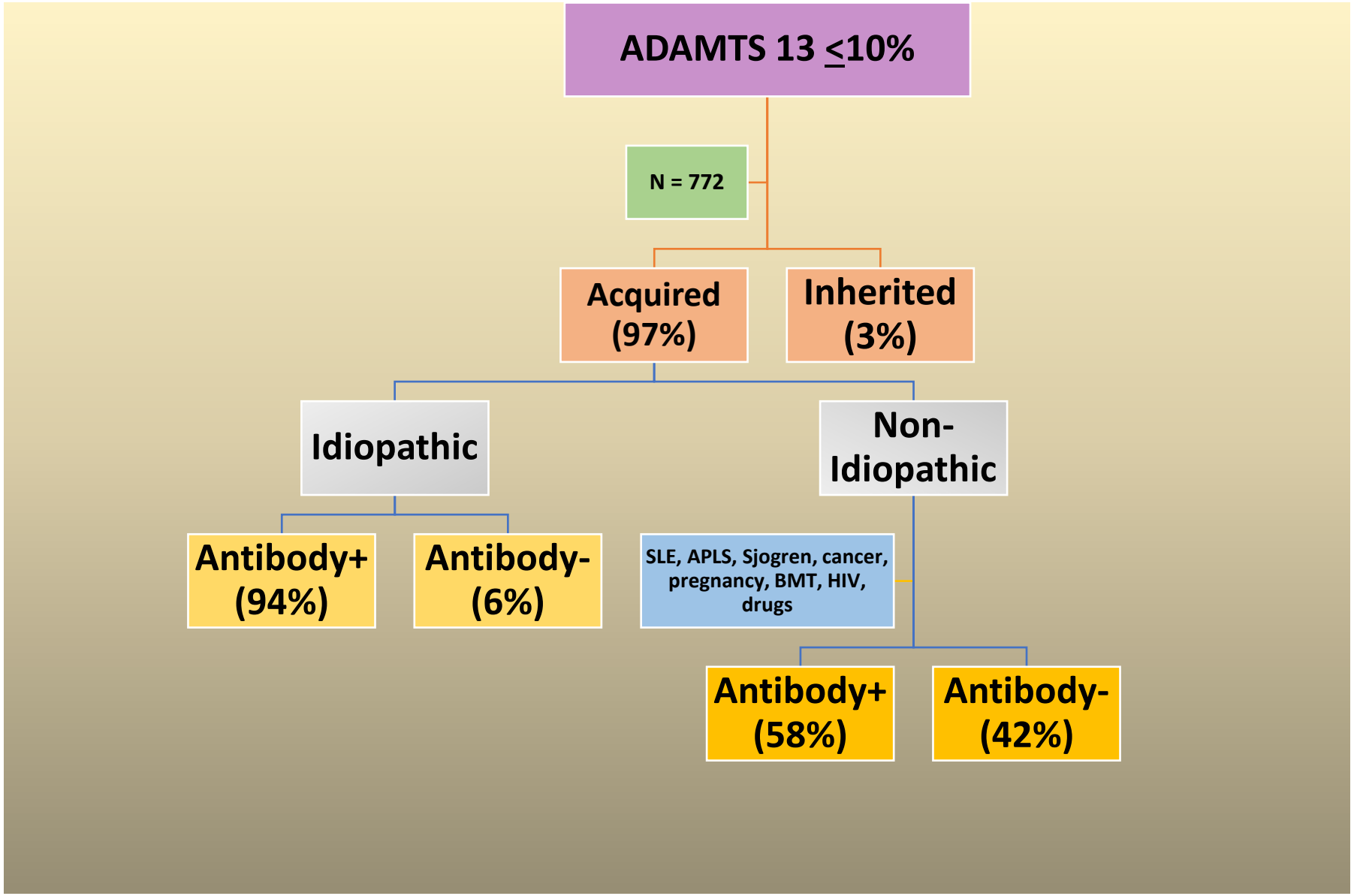
- Sorologia para hepatite – negativo
- Anti-HIV negativo
- **C3- 21 (90-180)**
- **C4- 5 (10-40)**
- **FAN- positivo 1:1280**
- **Anti-DNAds – positivo 1:40**
- VDRL negativo
- Sangue periférico – esquizócito 1+
- USG rins e vias urinárias: normal

De Holanda, Palma et al. *Clinical Rheumatology*, 2017 Sep

LES e Microangiopatia Trombótica (MAT): Diagnósticos Diferenciais



HM: hipertensão maligna; SAAF: síndrome do anticorpo antifosfolipídio; PTT: púrpura trombocitopênica trombótica; SHUa: síndrome hemolítico urêmica atípica



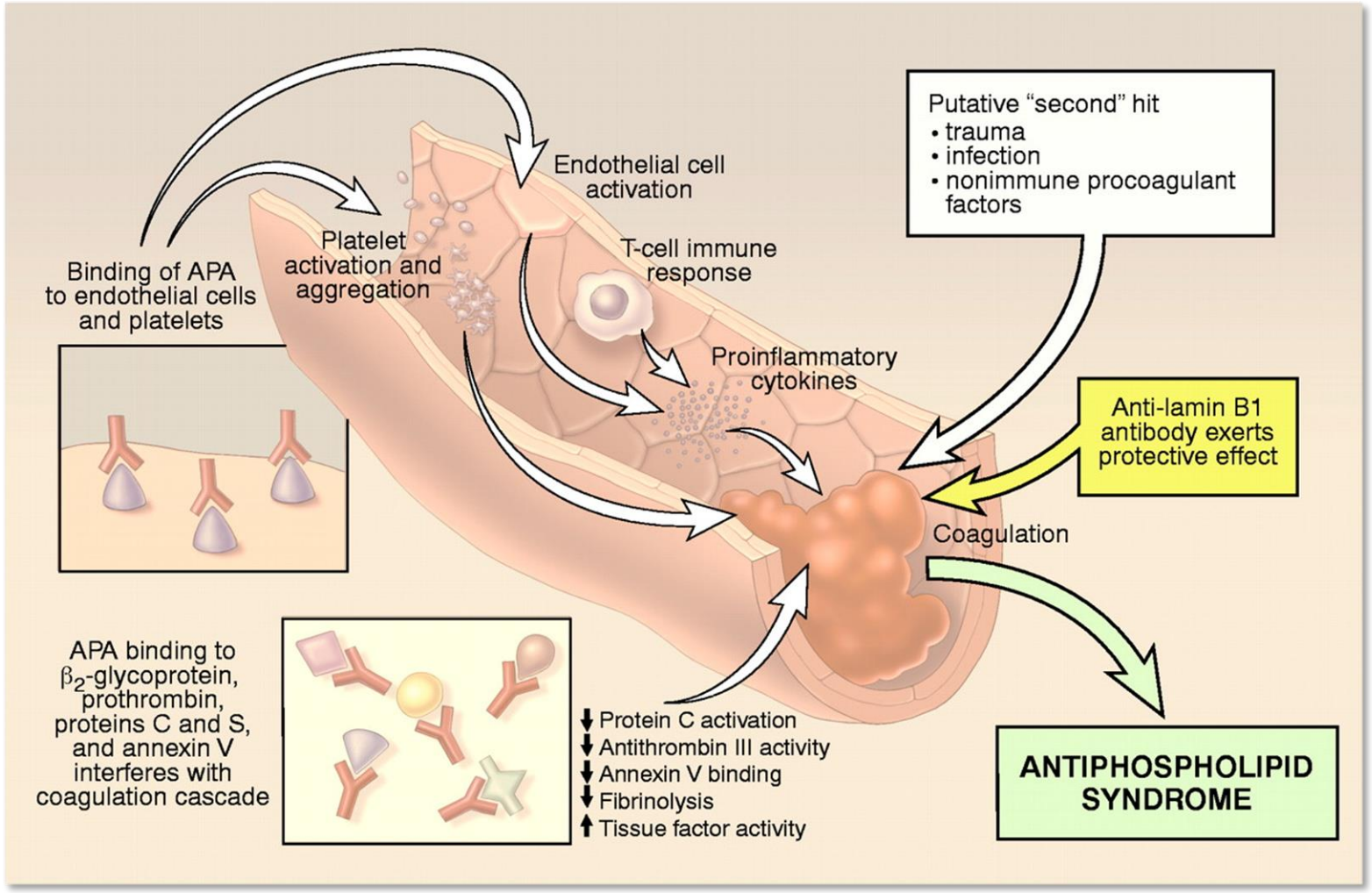
Mariotte E, et al. Lancet Haematol 2016;3:e237-e245.

Investigação e diagnóstico

- Sorologia para hepatite – negativo
- Anti-HIV negativo
- C3- 21 (90-180)
- C4- 5 (10-40)
- FAN- positivo 1:1280
- Anti-DNAds – positivo 1:40
- VDRL negativo
- Sangue periférico – esquizócito 1+
- USG rins e vias urinárias: normal
- **ADAMST13 – 58 %**

De Holanda, Palma et al. *Clinical Rheumatology*, 2017 Sep

A combinação de fatores leva à trombose na SAAF



Anticorpos contra um grupo heterogêneo de proteínas ligantes a fosfolipídios

- Cardiolipina
- $Beta_2$ glicoproteína 2
- Protrombina
- Anticoagulante lúpico

Ali Nayer et al, 2014;2-Saigal et al, 2010

Investigação e diagnóstico

- Sorologia para hepatite – negativo
- Anti-HIV negativo
- C3- 21 (90-180)
- C4- 5 (10-40)
- FAN- positivo 1:1280
- Anti-DNAds – positivo 1:40
- VDRL negativo
- Sangue periférico – esquizócito 1+
- USG rins e vias urinárias: normal
- ADAMST13 – 58 %
- **Anticorpos antifosfolipídios – negativos**
- **Anticorpos anticardiolipina – negativo**

De Holanda, Palma et al. *Clinical Rheumatology*, 2017 Sep

Diagnóstico e manejo inicial

- Lupus eritematoso sistêmico + microangiopatia trombótica
  - Metilprednisolona IV 1g x 3 dias (2 ciclos)
  - (19/20/21 maio – 26/27/28 maio)
  - Ciclofosfamida 1 g IV (22/05)
  - Plasmaferese 5 sessões
  - Micofenolato/azatioprina
- Sem biópsia (trombocitopenia)

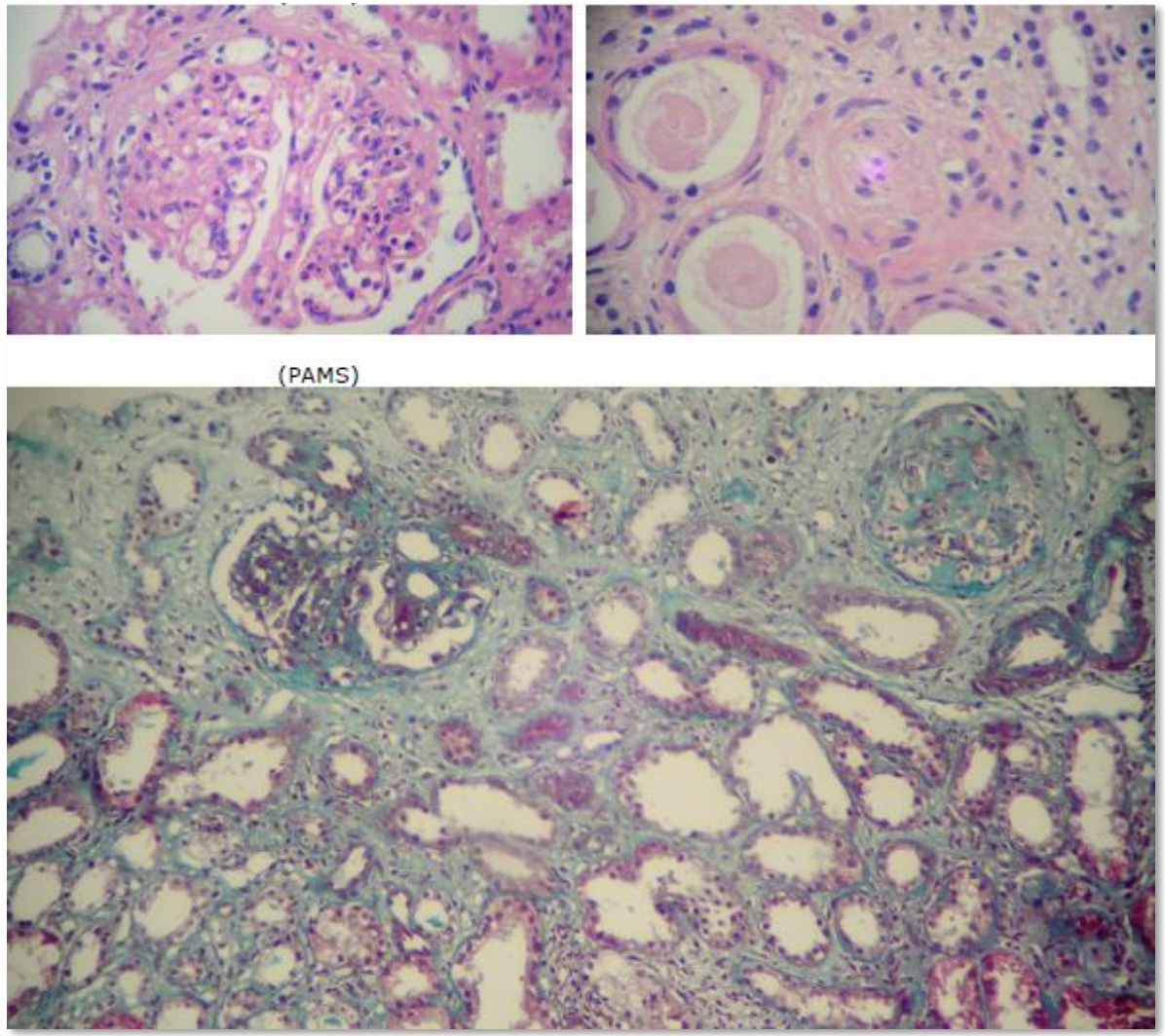
De Holanda, Palma et al. *Clinical Rheumatology*, 2017 Sep

Evolução da paciente

- **55 dias depois de iniciar imunossupressão:**
  - IRA – oligoanúria
  - Anemia (Hemoglobina: 6,6 g/dl)
  - Plaquetas 81.000
  - LDH > 1000
  - Haptoglobina < 6
- Biópsia renal (15/07)

De Holanda, Palma et al. *Clinical Rheumatology*, 2017 Sep

Nefrite Lúpica classe IV e Microangiopatia Trombótica

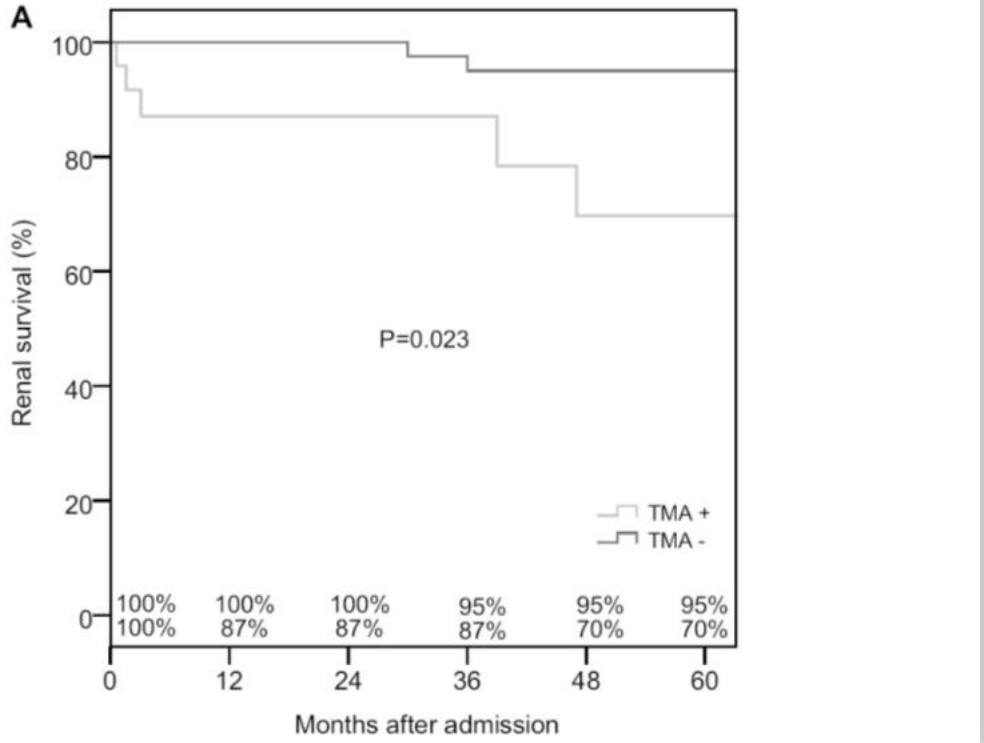


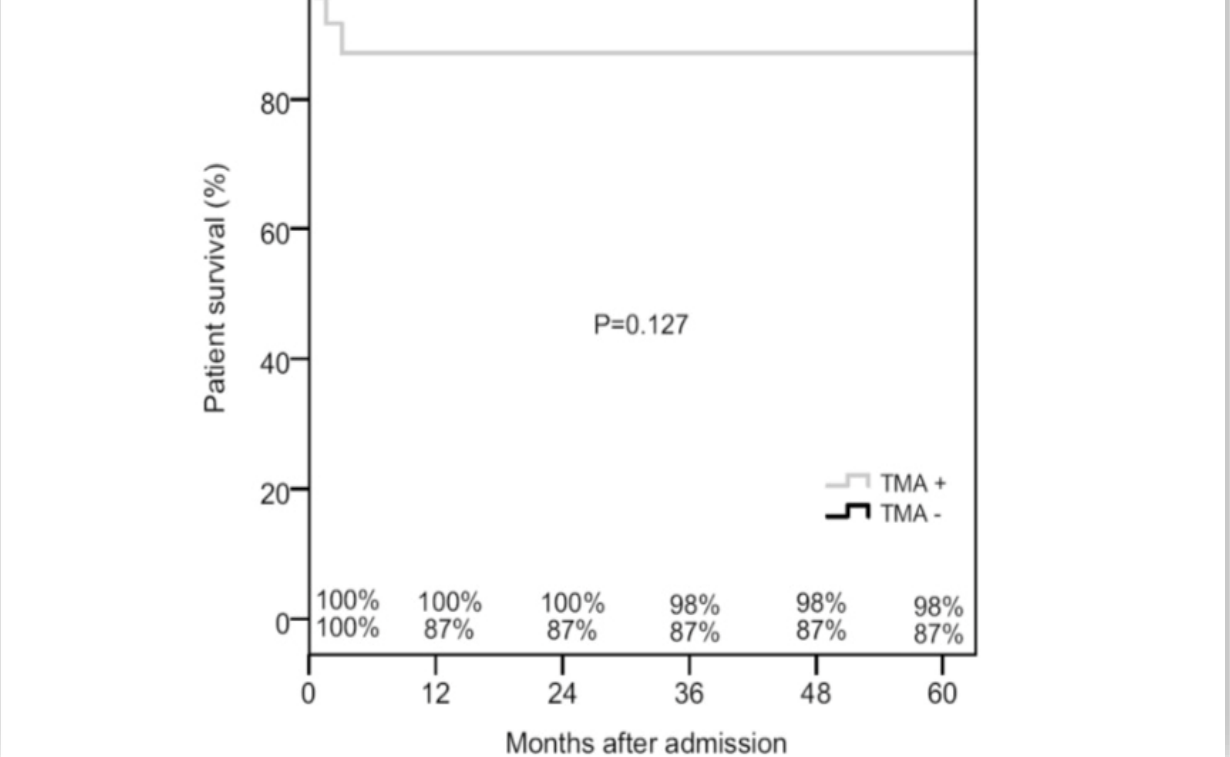
De Holanda, Palma et al. *Clinical Rheumatology*, 2017 Sep

The Journal of Rheumatology

Clinical Outcomes and Clinico-pathological Correlations in Lupus Nephritis with Kidney Biopsy showing Thrombotic Microangiopathy

Chao Li, Desmond Y.H. Yap, Gavin Chan, Yu-bing Wen, Hang Li, Colin Tang, Xue-mei Li, Xue-wang Li and Tak Mao Chan





Genética das doenças glomerulares imunes: foco no complemento

Variações no gene do complemento associadas a IgAN e Nefrite Lúpica					
IgAN	**Gene**	**Variante/Anormalidade**	**Principal efeito**	**Impacto**	**Frequência**
-	**CFH**	c. 1696+2019ª (c. 1696 + 2019G > A, rs 6677604)	/	Protetor	SNP comum
-	**CFHR1/CFHR3**	Deleção	Menor competição com FH	-	-
-	**CFHR5**	Raras variantes funcionais	Aumento da competição com FH	Risco	9%
NL	-	Anormalidade	-	-	-
-	**CFH**	Mutações homozigóticas	Comprometimento da fase fluida da via alternativa (AP) de regulação do complemento	Patogênica	Muito rara
-	**CFHR1/CFHR3**	Deleção	Anticorpos anti-FH? Hiperatividade da célula B?	Risco	Deleção comum
-	**CFI**	Mutações homozigóticas	Comprometimento da fase fluida de regulação do complemento	Patogênica	Muito rara
-	**C1Q, C1R, C1S**	Mutações homozigóticas	Deficiência de C1; sem ativação da via clássica (CP)	Alto risco de LES e NL	Rara
-	**C4A, C4B**	Mutações homozigóticas	Deficiência de C4; sem ativação da CP nem da via da lectina (LP)	Alto risco de LES e NL	Rara
-	**C2**	Mutações homozigóticas	Deficiência de C2; sem ativação da CP nem da LP	Risco de LES e NL	Rara

Marina Noris PhD, Giuseppe Remuzzi MD
Seminars in Nephrology, Vol 37, Nº 5, September 2017, pp 447-463

Genetic Analysis of the patient with SLE and TMA

Request for Atypical hemolytic uremic syndrome

Clinical information: thrombotic microangiopathy, lupus, generalized seizures, vasculitis, anemia, pancytopenia.

Result details:

Atypical hemolytic uremic syndrome panel (sequencing)

ADAMTS13, C3, CD46, CFHR1, CFHR2, CFHR3, CFHR5, CFI, DGKE, PIGA, THBD, CFH	no pathogenic mutation

Atypical hemolytic uremic syndrome panel (MLPA)

CFHR1 and CFHR3	heterozygous large deletion encompassing entire CFHR1 and CFHR3 genes
CFH, CFHR2, CFHR5	no deletion/duplication

We detected a heterozygous deletion encompassing the *CFHR1* and *CFHR3* genes. We urgently recommend analyzing in the next step for deletion/duplication in the genes ADAMTS13, C3, CD46, CFHR1, CFHR2, CFHR3, CFHR5, CFI, DGKE, PIGA, THBD, CFH.

Given the results we recommend
- genetic counselling
- deletion/duplication analysis of the CFI gene.

Interpretation:

We detected a heterozygous deletion encompassing the *CFHR1* and *CFHR3* genes. Zipfel et al. (2007) found that an 84-kb deletion of the CFHR1 and CFHR3 genes was associated with an increased risk of atypical hemolytic-uremic syndrome in 2 independent European cohorts. In the first group, 19 (16%) of 121 aHUS patients had the deletion compared to 2 of 100 control individuals. Three of the patients had a homozygous deletion. All patients had normal serum factor H levels. In the second group comprising 66 patients, 28% had the deletion compared to 6% of controls. Ten percent and 2% of patients and controls, respectively, were homozygous for the deletion.
In vitro functional expression studies showed that CFHR1/CFHR3-deficient plasma had decreased protective activity against erythrocyte lysis, suggesting a defective regulation of complement activation. Of 147 patients

Evolução da paciente

Eculizumabe 12/07

Eculizumabe 18/07 25/07 01/08

Eculizumabe 11/08

	12/07	16/07	17/07	02/08	04/08	11/08	10/03
Hemoglobina	6,6	4,5	7,9	8,0	8,5	10,6	10,6
Plaquetas	81.000	98.000	79.000	172.000	183.000	146.000	215.000
Diurese (mL)	100	350	710	880	1.375	1.310 Cr: 2,3	2.000 Cr: 2,3

- 11/08 - paciente recebeu alta após 1 semana fora de hemodiálise
- 10/03 - Boa qualidade de vida – tratamento de LES e eculizumabe 2x por mês (Cr: 2,3)
- Agosto/2016 – nova biópsia renal – sem atividade de doença, proteinúria residual (Cr: 2,3)
- Janeiro 2017 – Cr: 2,5 mg/dL; proteinúria em torno de 1,2 g/24h

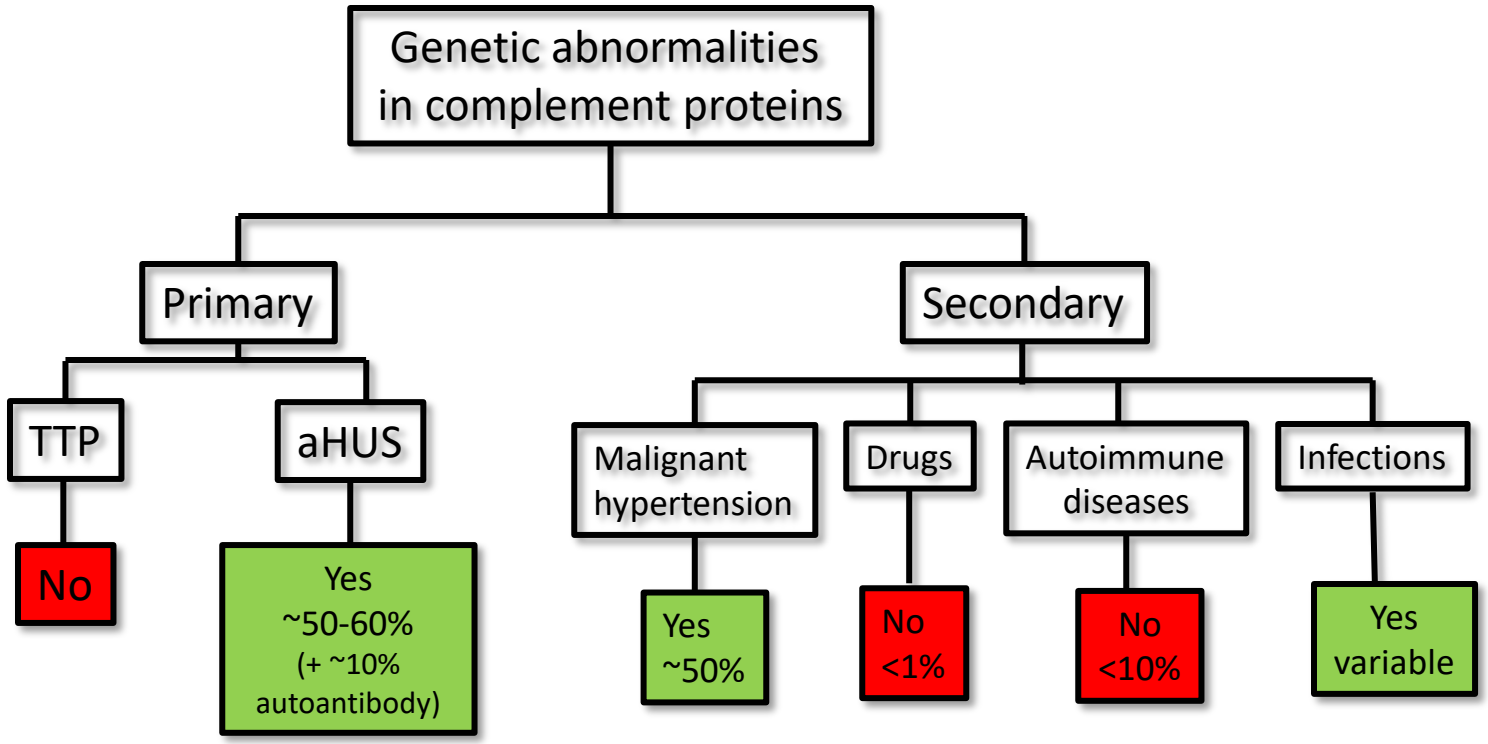
De Holanda, Palma et al. Clinical Rheumatology, 2017 Sep

SLE, aHUS & eculizumab

Coppo *et al.* *Pediatr Nephrol, 2015*	4 yo fem SLE & nephritis TMA, cardiovascular, neurologic and pulmonary GT negative No response to rituxumab	Rapid disapperance of TMA Hemato and kidney normalization TMA recurred after eculizumab stopped – resumed with improvement
El Husseini *et al. Am J Kidney Dis, 2015*	24 yo fem SLE and TMA (clinical and biopsy) No response to cyclophosphamide, PE and steroids	Complete normalization in six months of eculizumab treatment Discontinued
Hadaya *et al.* *Am J Transplant, 2011*	27 yo fem ESRD and SLE TMA on biopsy GT negative TMA persisted after Ktx, no response to PE	Complete improvement with eculizumab

Asif J et al. *J Nephrol*, 2016

Complement Genetics in TMA



Palma L., Sridharan M., Sethi S. et al. *Kidney Int Reports*, 2021

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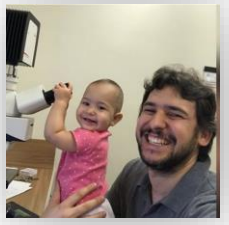

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COMDORA

Brazilian aHUS Registry
comdora-sbn.org