

Complement-mediated HUS

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Fondazione IRCCS Ca' Granda
Ospedale Maggiore Policlinico

Sistema Socio Sanitario



Regione
Lombardia



The TMAs

1. TTP (congenital and acquired)

2. HUS

Shigatoxin-related-HUS (tHUS, D+HUS, VTEC-HUS, eHUS)

Atypical HUS

Complement mediated

Secondary (Malignancies, Autoimmune diseases, Drugs
Infections, Hypertension)

Idiopathic

Coagulation-mediated (DGKe, THBD)

Pneumonia-related

Cobalamine dismetabolism (MMA & deficiency)

3. TA-TMA

4. HELLP

5. APS

6. Scleroderma

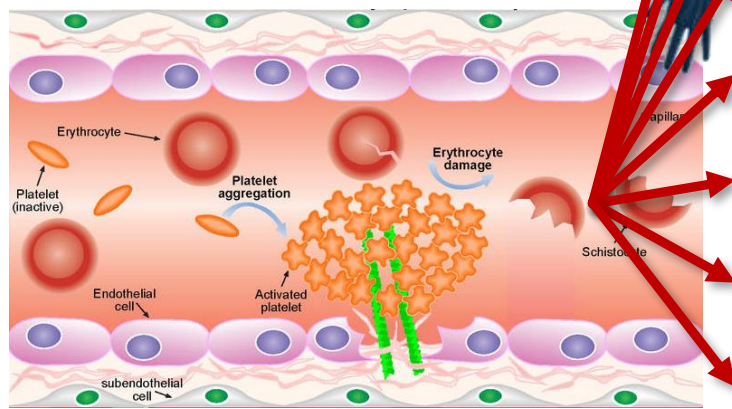
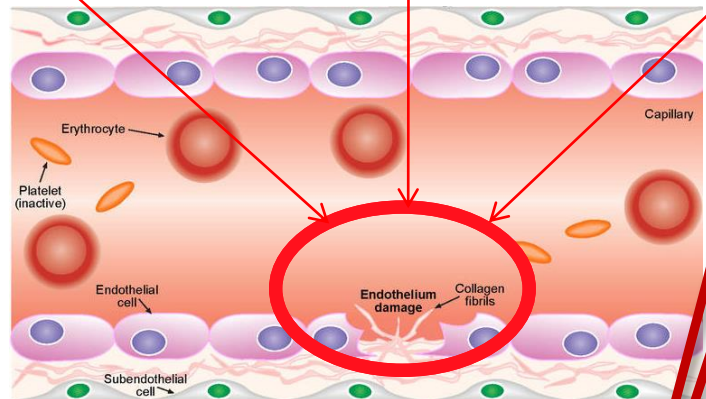
7. DIC

Thrombotic Microangiopathies (MAT): Endothelial Diseases

Shigatoxin

Complement Abnormalities

TA-R, Pneumonia-R,
Coagul-R, Cobalamin-R
Drugs-R, Idiopathic,
Secondary, etc.



Endothelial Damage



Thrombosis in the
Microcirculation



Platelet
consumption



Hypoxic/
ischemic
organ
damage



Mechanical
hemolysis

Blood

- Haemolysis
- Decreased platelets

Renal

- Proteinuria
- Malignant hypertension
- Renal failure

Pulmonary

- Pulmonary haemorrhage
- Pulmonary oedema

CNS

- Confusion
- Seizures
- Stroke
- Encephalopathy

Cardiovascular

- Myocardial infarction
- Thromboembolism
- Cardiomyopathy

Gastrointestinal

- Liver necrosis
- Pancreatitis
- IDDM
- Colitis

Skin

- Ulcerations

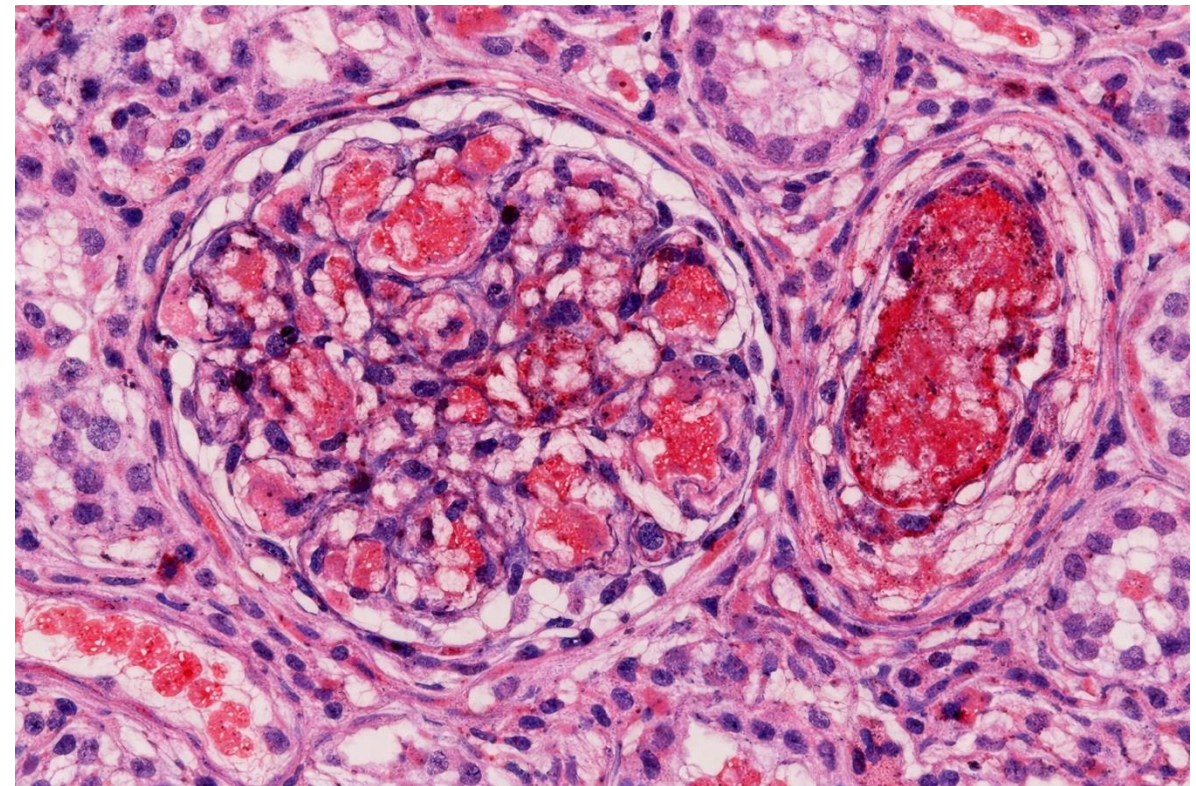
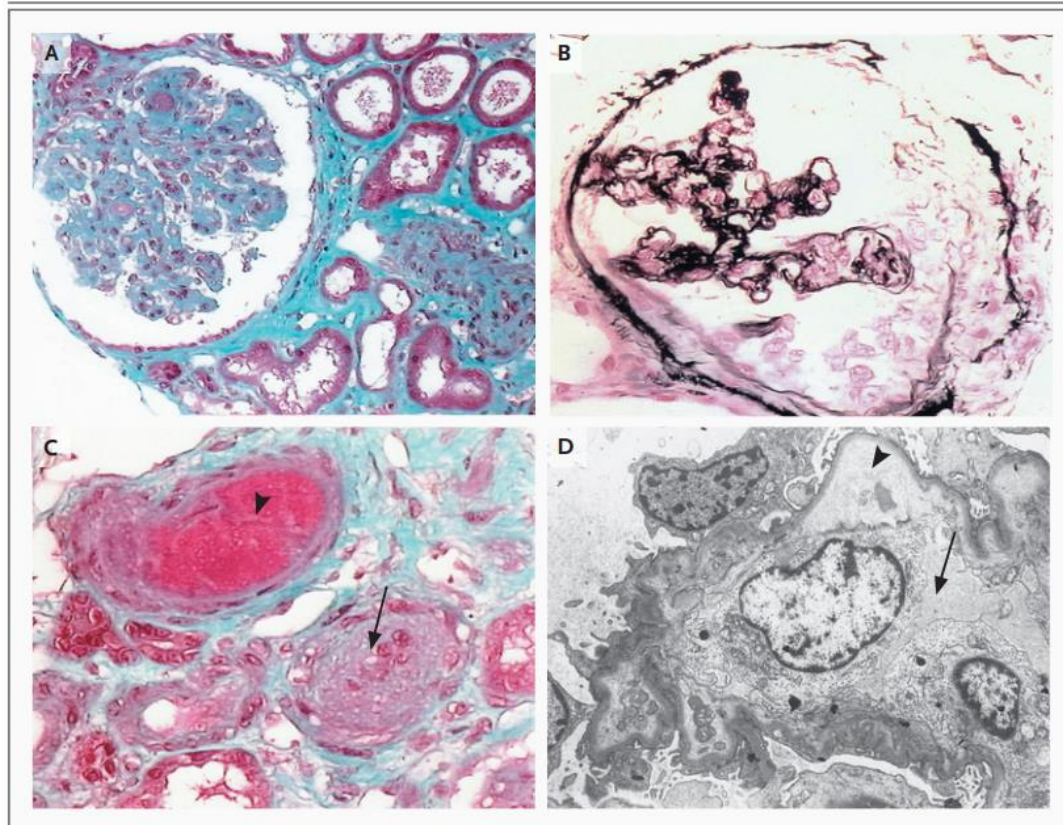
Atypical Hemolytic–Uremic Syndrome

Marina Noris, Ph.D., and Giuseppe Remuzzi, M.D.

N Engl J Med 2009;361:1676-87.

THE NEW ENGLAND JOURNAL of MEDICINE

The core histologic pattern: glomerular and arteriolar thrombotic microangiopathy (endothelial swelling, capillary lumen occlusion secondary to platelet fibrin thrombi) acute tubular injury, **no immune deposition**.



The diagnostic triad

- **Thrombocytopenia**
- **Hemolytic Anemia**
- **AKI**



Time to change the definition of hemolytic uremic syndrome

Gianluigi Ardissino ¹, Ilaria Possenti ², Francesca Tel ³, Sara Testa ⁴, Fabio Paglialonga ⁵

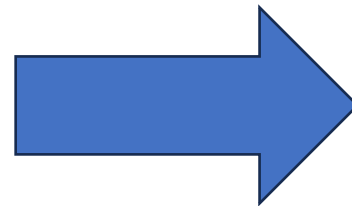
	<u>tHUS</u>	<u>aHUS</u>	<u>Total</u>
<u>Total</u>	116	16	132
<u>Hb > LASR</u>	24%	31%	25%
<u>No low PLT</u>	11%	6%	11%
<u>Normal renal function</u>	14%	19%	14%
<u>All 3 criteria</u>	60%	50%	59%

The correct definition of HUS may not be a critical for TMA specialists but the frontline health care providers are not the specialists and the current definition does not help to promptly identify patients

The diagnostic triad

Before

- **Thrombocytopenia**
- **Hemolytic Anemia**
- **AKI**



Now

Platelet consumption

Thrombocytopenia
Acute reduction in platelet count
IPF >3% o MPV >11 fl

Hemolysis

Anemia
Schistocytes
↑ LDH
Haptoglobin consumption

Kidney Damage

Renal failure
Hematuria/Proteinuria

Platelets consumption

Hemolysis

DCT negative

Kidney damage

(+/- multiorgan)

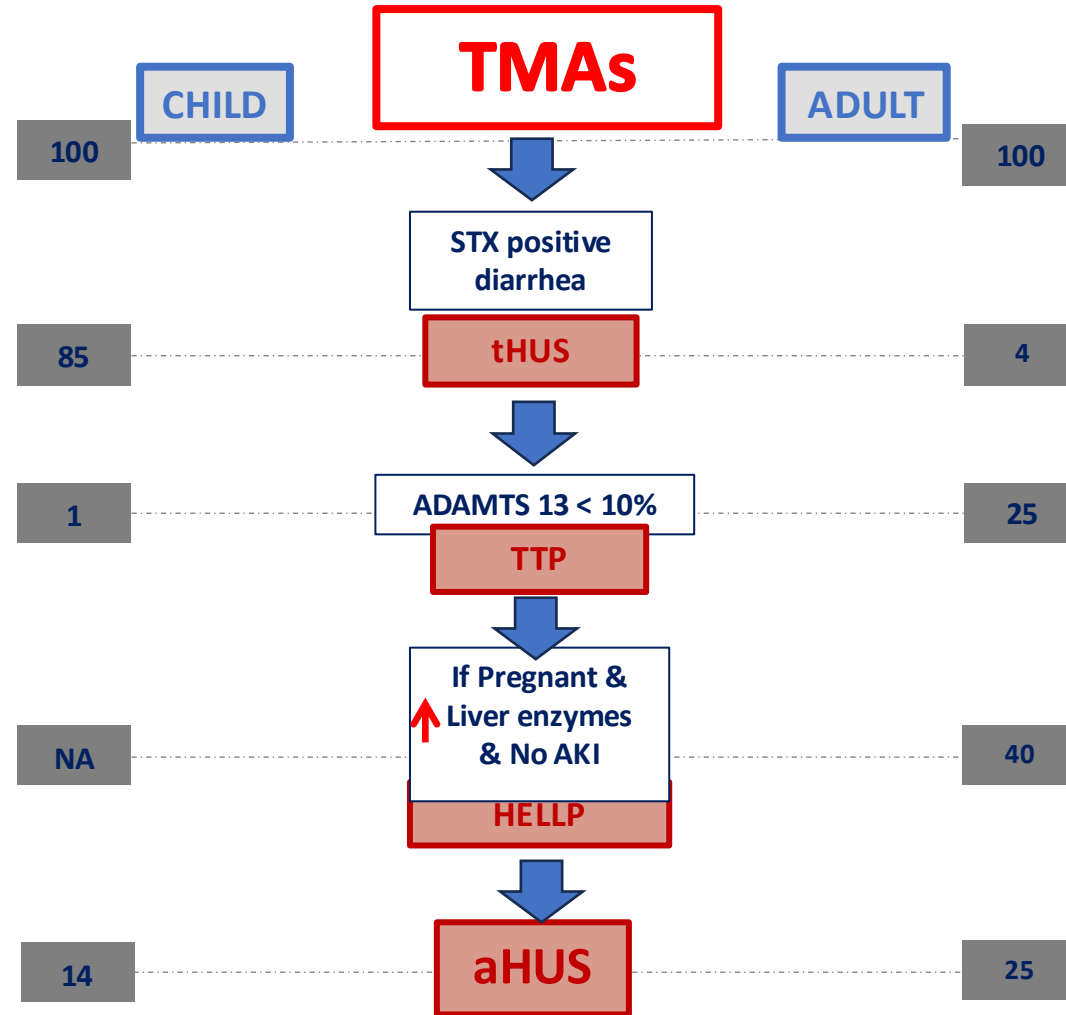
Without diarrhea or with Stx negative diarrhea



Atypical HUS

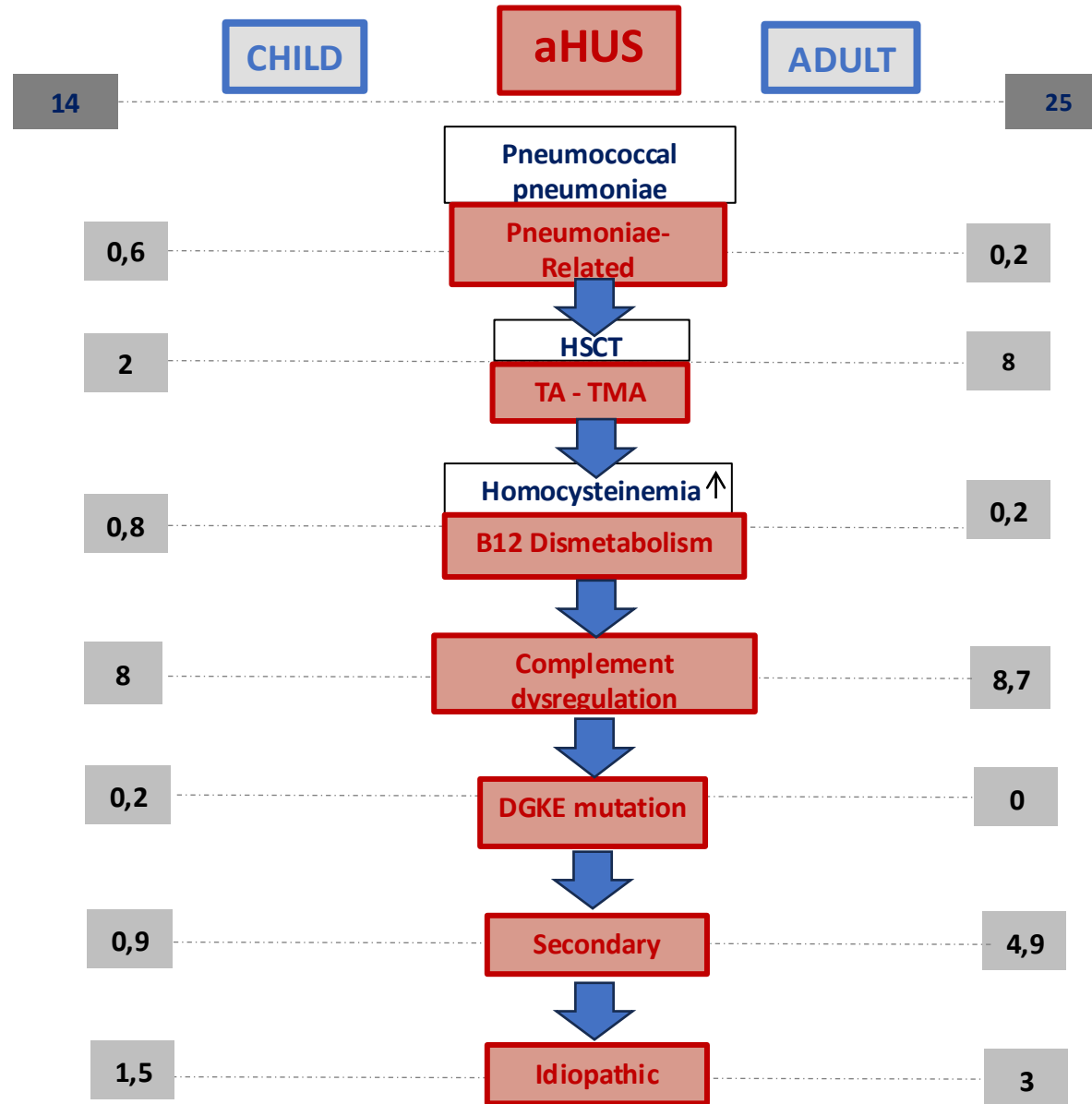
Rule out TTP (ADAMTS13 >10%)

Epidemiology-based diagnostic algorithm of TMAs



HELLP: Hemolysis, elevated liver enzymes, low platelets

Epidemiology-based diagnostic algorithm of TMAs



Atypical Hemolytic–Uremic Syndrome

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N Engl J Med 2009;361:1676-87.

Uncontrolled over-activation of the alternative complement pathway:

1. Genetic abnormalities (50%)

Loss of function

(CFH & CFHR, CFI, MCP)

Gain of function

(C3, CFB)

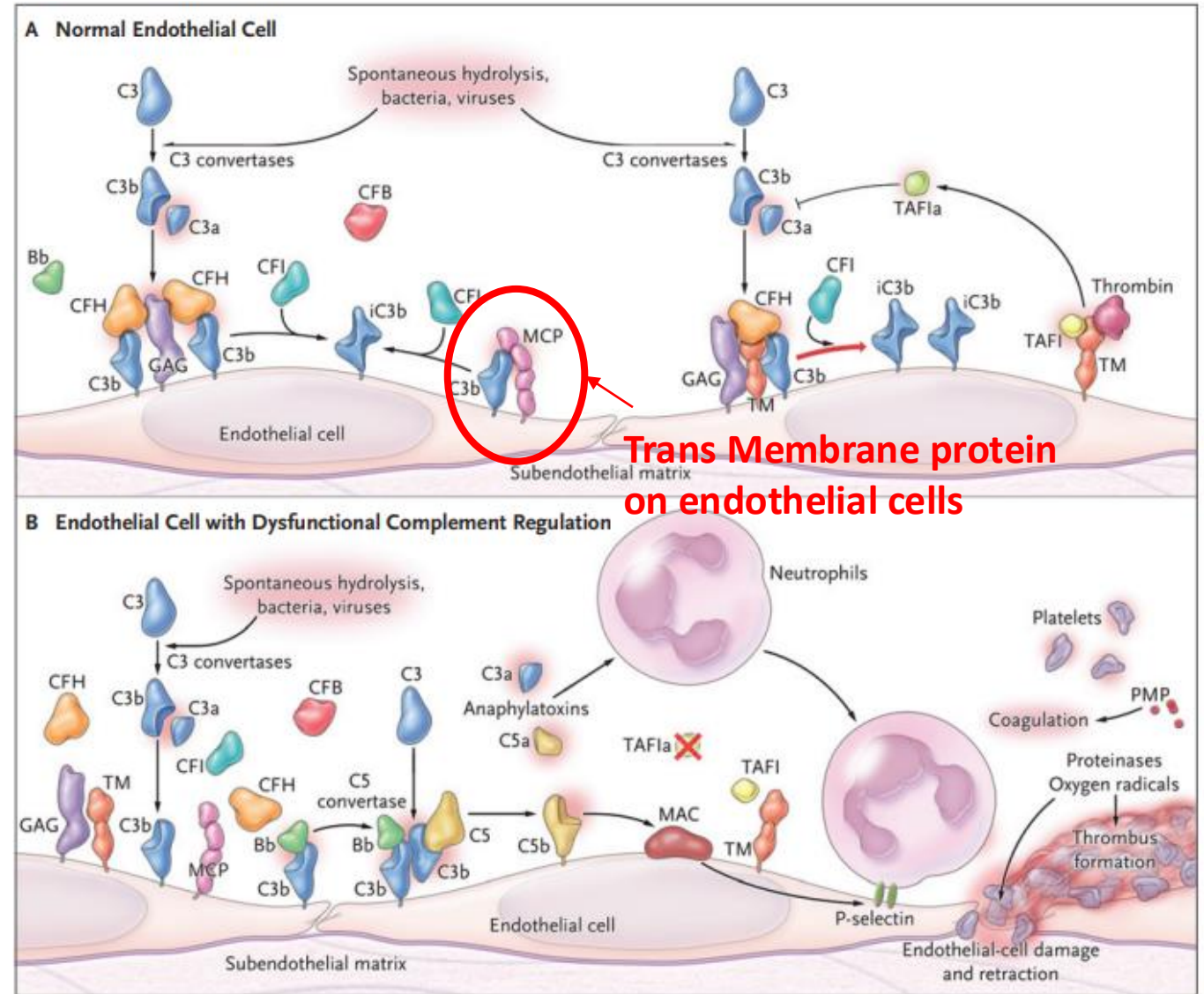
2. AutoAntibodies (<10%)

(Anti-CFH) associated with CFHR1-3 deletions

3. Idiopathic (40%)

CFH: Primary brake on C activation

CFHR: Fine tuners proteins



Outcome of atypical hemolytic uremic syndrome: role of triggers and complement abnormalities in the response to C5 inhibition

Gianluigi Ardissino¹ · Donata Cresseri² · Maria Cristina Mancuso¹ · Valentina Capone¹ · Luigi Porcaro³ · Valeria Amico³ · Marianna Tangredi² · Elena Grovetti⁴ · Samantha Griffini⁴ · Giuseppe Castellano² · Giovanni Montini⁵ · Dario Consonni⁶ · Massimo Cugno⁴ · on behalf of the HUS-ItalKid Network.

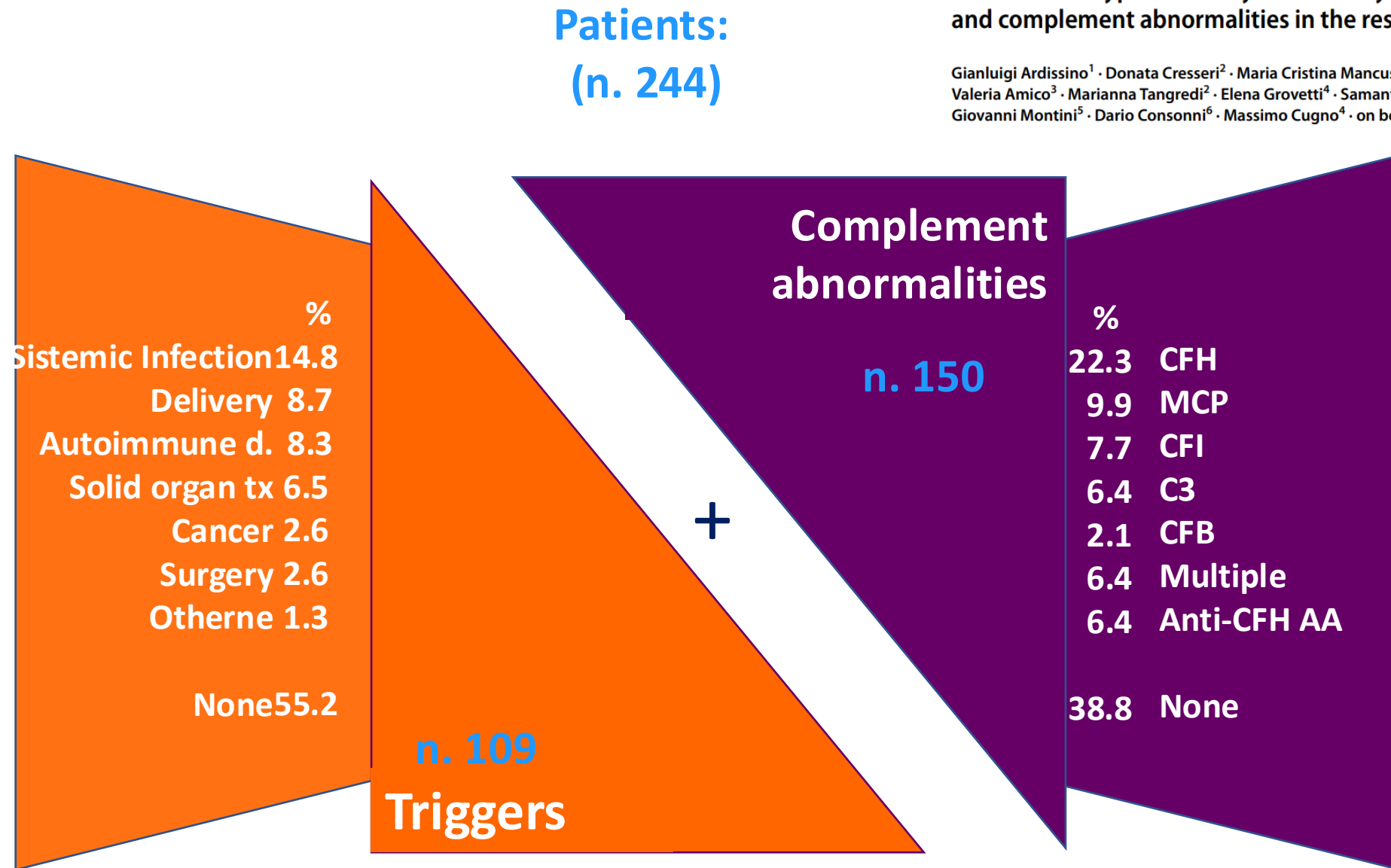
Journal of Nephrology (2024) 37:1017–1026
<https://doi.org/10.1007/s40620-023-01873-9>

ORIGINAL ARTICLE

		No. of patients (%)
Patients		244 (100)
Sex (Female)		133 (54.5)
Pediatric Onset		73 (29.9)
Recurrent		50 (20.5)
Complement Abnormality		150 (61.5)
	CFH	54 (22.1)
	MCP	26 (10.7)
	CFI	18 (7.4)
	C3	14 (5.7)
	CFB	5 (2.0)
	Multiple	16 (6.5)
	Anti-CFH Ab	17 (7.0)
	None	94 (38.5)
Trigger		109 (44.7)
	Infection	38 (15.6)
	Pregnancy/abortion/delivery	22 (9.0)
	Autoimmune disease	18 (7.4)
	Solid organ transplant	15 (6.1)
	Cancer	7 (2.9)
	Surgery	6 (2.5)
	Other	3 (1.2)
	None	135 (55.3)
Conventional treatment		102 (41.8)
C5 inhibitor		142 (58.2)

Outcome of atypical hemolytic uremic syndrome: role of triggers and complement abnormalities in the response to C5 inhibition

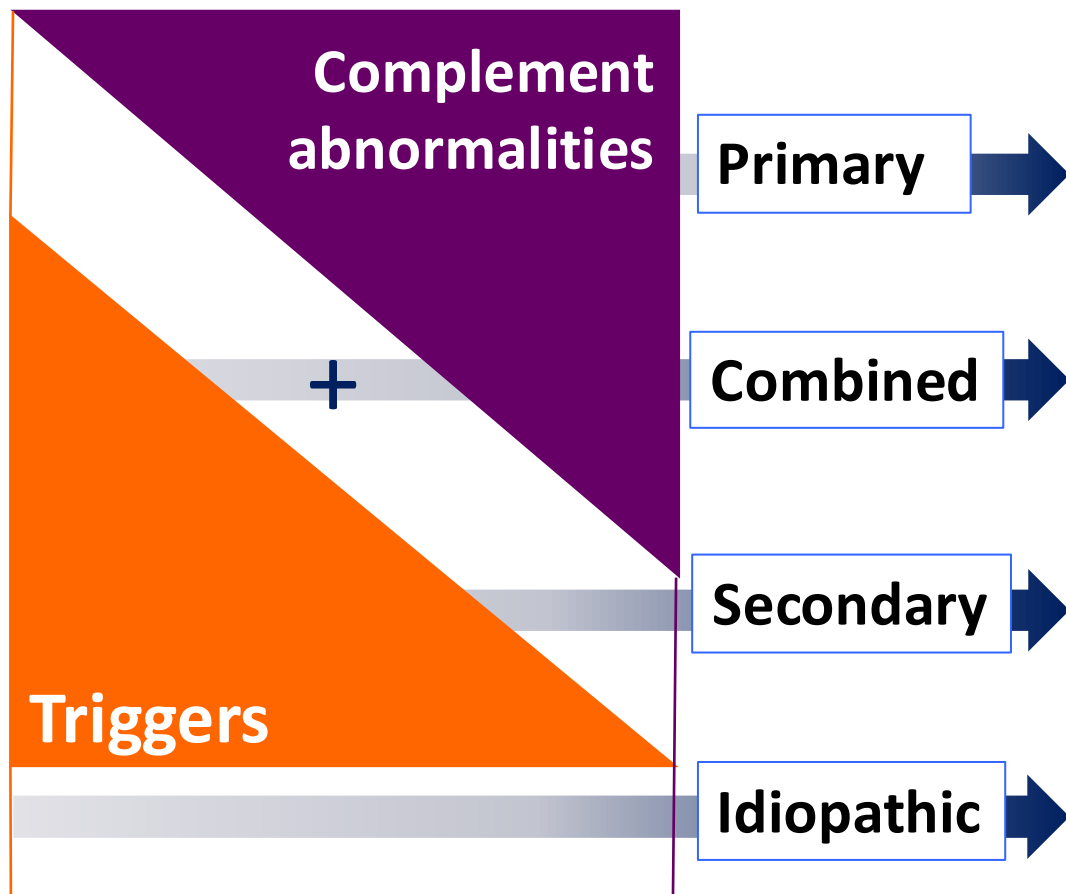
Gianluigi Ardissino¹ · Donata Cresseri² · Maria Cristina Mancuso¹ · Valentina Capone¹ · Luigi Porcaro³ · Valeria Amico³ · Marianna Tangredi² · Elena Grovetti⁴ · Samantha Griffini⁴ · Giuseppe Castellano² · Giovanni Montini⁵ · Dario Consonni⁶ · Massimo Cugno⁴ · on behalf of the HUS-ItaKid Network.



* Trigger: any acute or chronic condition present at the event (HUS) but not regularly encountered in the general population

Outcome of atypical hemolytic uremic syndrome: role of triggers and complement abnormalities in the response to C5 inhibition

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Total n: 244 (100%)	Pediatric (n: 73)	Adult (n: 171)
99 (40.6)	46 (63.0)	53 (31.0)
49 (20.1)	14 (19.2)	35 (20.5)
58 (23.8)	5 (6.8)	53 (31.0)
38 (15.6)	8 (11.0)	30 (17.5)

.....which analysis?

Next Generation Sequencing (NGS):

CFH (NM_000186.4)

MCP/CD46 (NM_002389.4)

CFI (NM_000204.5)

+ THBD (NM_000361.3)

C3 (NM_000064.4)

+ DGKE (NM_003647.3) autosomal recessive!!

CFB (NM_001710.65)

CFHR1 (da Ex2 a Ex6) (NM_002113.3), CFHR3 (NM_021023.6), CFHR5 (NM_030787.4)

Autosomal dominant

Analytical sensitivity and specificity >98%

Multiplex Ligation-Dependent Probe Amplification Analysis (MLPA):

CFH, CFHR3, -R1, -R2, -R4 e -R5 for the search for macrodeletions/macro duplications and hybrid genes

Sensitivity 90%

+ haplotypes at risk CFH-H3 e MCPgg

Ab anti CFH

Associated with CFHR3-CFHR1 omo deletion

Complement Gene Variants and Shiga Toxin–Producing *Escherichia coli*–Associated Hemolytic Uremic Syndrome

Retrospective Genetic and Clinical Study

www.cjasn.org Vol 14 March, 2019

Véronique Frémeaux-Bacchi,^{1,2} Anne-Laure Sellier-Leclerc,³ Paula Vieira-Martins,¹ Sophie Limou,⁴ Theresa Kwon,⁵ Annie Lahoche,⁶ Robert Novo,⁶ Brigitte Llanas,⁷ François Nobili,⁸ Gwenaëlle Roussey,⁹ Mathilde Cailliez,¹⁰ Tim Ulinski,¹¹ Georges Deschênes,⁵ Corinne Alberti,¹² François-Xavier Weill,¹³ Patricia Mariani,¹⁴ and Chantal Loirat⁵

Complement Abnormalities	Postdiarrheal HUS, Total Cohort, <i>n</i> =108 ^a	French Controls, <i>n</i> =80	1000 Genomes Controls, <i>n</i> =503
	<i>n</i> (%)	<i>n</i> (%)	<i>n</i> (%)
One rare variant per patient or control			
Complement factor H	3 (3)	2 (3)	8 (2)
Membrane cofactor protein	1 (0.9)	0	2 (0.4)
Complement factor I	1 (0.9)	2 (3)	13 (3)
C3	8 ^d (7)	4 (5)	16 (3)
Complement factor B	1 (0.9)	0	7 (1)
Thrombomodulin	3 (3)	1 (1)	9 (2)
Two rare variants per patient or control	1 ^e (0.9)	2 ^f (3)	5 (1)
Total	18 (17)	11 (14)	60 (12)
Anti-factor H antibodies	3 ^d (3)	0	

Complement Gene Variants and Shiga Toxin–Producing *Escherichia coli*–Associated Hemolytic Uremic Syndrome Retrospective Genetic and Clinical Study

www.cjasn.org Vol 14 March, 2019

Véronique Frémeaux-Bacchi,^{1,2} Anne-Laure Sellier-Leclerc,³ Paula Vieira-Martins,¹ Sophie Limou,⁴ Theresa Kwon,⁵ Annie Lahoche,⁶ Robert Novo,⁶ Brigitte Llanas,⁷ François Nobili,⁸ Gwenaëlle Roussey,⁹ Mathilde Cailliez,¹⁰ Tim Ulinski,¹¹ Georges Deschênes,⁵ Corinne Alberti,¹² François-Xavier Weill,¹³ Patricia Mariani,¹⁴ and Chantal Loirat⁵

- **very rare pathogenic variants** with minor allele frequency ,0.1% are more frequent in Shiga toxin–positive patients
- role of **complement activation** in Shiga toxin–associated HUS
- in rare cases **genetic abnormalities may contribute to C activation** in Shiga toxin–associated HUS
- the only patient who progressed to **ESKD** carried a factor H pathogenic variant leading to functional factor H deficiency
- **genetic screening** should be considered in post-diarrheal HUS patients who progress rapidly to ESKD.

Differential Diagnosis

	STEC-HUS	Complement-relatedHUS
Renal Damage	+	+
Haptoglobin	↓	↓
LDH	↑	↑
C3	↓/=	↓/=
PLT	Spontaneous normalization with the overcome of the infection	Normalization only after C5i

SUPPORTIVE TREATMENT

“Ad interim” recommendation for RBC transfusion

- When Hb < 7 gr/dL
- Target Hb 9-10 gr/dL

“Ad interim” recommendation for Platelets Transfusion

- Active bleeding
- Significant surgical procedures (Not CVC!)

Atypical Hemolytic–Uremic Syndrome

Marina Noris, Ph.D., and Giuseppe Remuzzi, M.D.

Table 2. Genetic Abnormalities and Clinical Outcome in Patients with Atypical Hemolytic–Uremic Syndrome.*

Gene	Protein Affected	Main Effect	Frequency %	Response to Short-Term Plasma Therapy†	Long-Term Outcome‡	Outcome of Kidney Transplantation
<i>CFH</i>	Factor H	No binding to endothelium	20–30	Rate of remission: 60% (dose and timing dependent)	Rate of death or ESRD: 70–80%	Rate of recurrence: 80–90%§
<i>CFHR1/3</i>	Factor HR1, R3	Anti-factor H antibodies	6	Rate of remission: 70–80% (plasma exchange combined with immunosuppression)	Rate of ESRD: 30–40%	Rate of recurrence: 20%¶
<i>MCP</i>	Membrane cofactor protein	No surface expression	10–15	No definitive indication for therapy	Rate of death or ESRD: <20%	Rate of recurrence: 15–20%¶
<i>CFI</i>	Factor I	Low level or low cofactor activity	4–10	Rate of remission: 30–40%	Rate of death or ESRD: 60–70%	Rate of recurrence: 70–80%§
<i>CFB</i>	Factor B	C3 convertase stabilization	1–2	Rate of remission: 30%	Rate of death or ESRD: 70%	Recurrence in one case
<i>C3</i>	Complement C3	Resistance to C3b inactivation	5–10	Rate of remission: 40–50%	Rate of death or ESRD: 60%	Rate of recurrence: 40–50%
<i>THBD</i>	Thrombomodulin	Reduced C3b inactivation	5	Rate of remission: 60%	Rate of death or ESRD: 60%	Recurrence in one case

Eculizumab (AntiC5Ab) 2009: the changeover



The NEW ENGLAND
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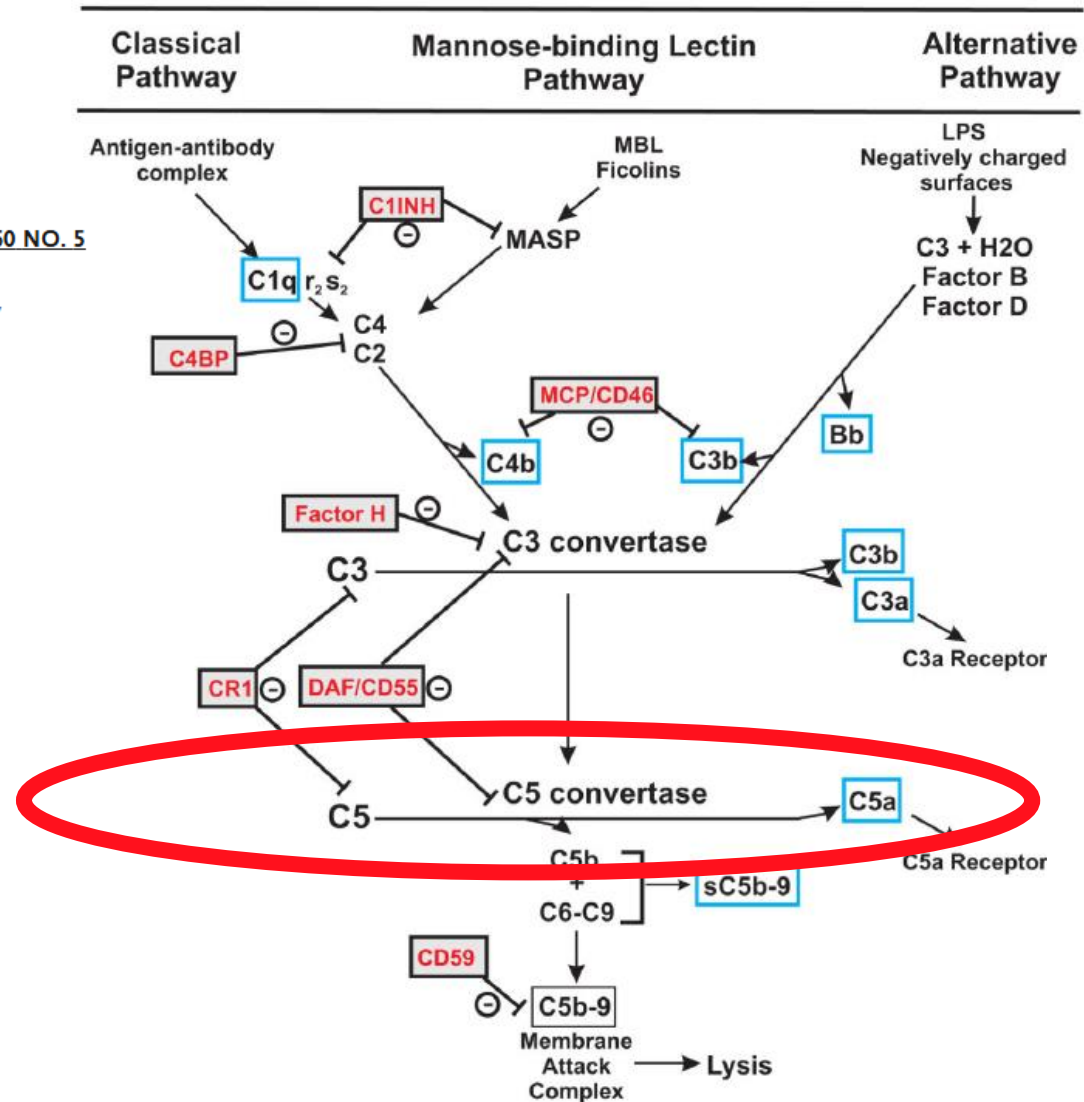
Eculizumab for Atypical Hemolytic–Uremic Syndrome

Syndrome

Published January 29, 2009 | N Engl J Med 2009;360:542-544 | DOI: 10.1056/NEJMc0808527 | VOL. 360 NO. 5

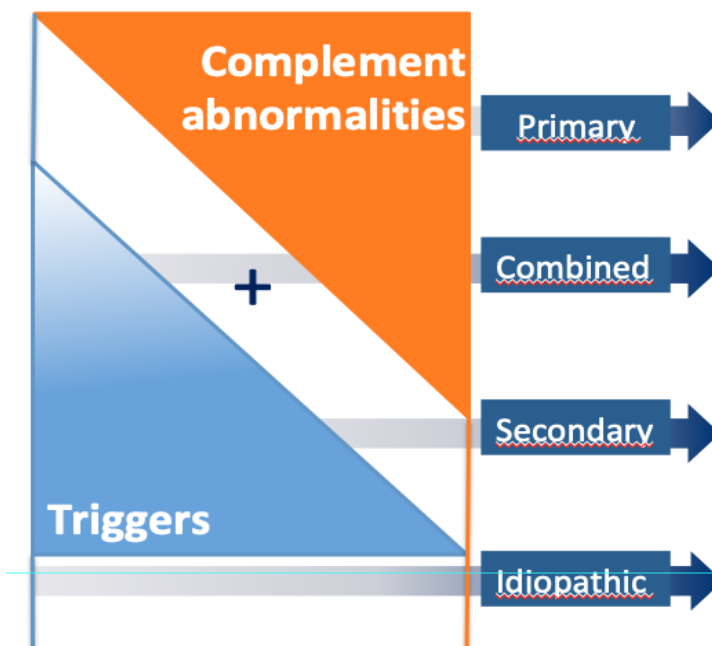
Jens Nürnbergger, Thomas Philipp, Oliver Witzke, Anabelle Opazo Saez, Udo Vester, Hideo Andreas Baba, Andreas Kribben, Lothar Bernd Zimmerhackl, Andreas R Janecke, Mato Nagel, Michael Kirschfink

- humanized monoclonal IgG antibody, which targets C5, preventing its cleavage
- pediatric eculizumab dosing (aHUS) ≥ 40 kg
 - **Induction:** 900 mg IV weekly \times 4 weeks
 - **Week 5:** 1,200 mg IV
 - **Maintenance:** 1,200 mg every 2 weeks



Outcome of atypical hemolytic uremic syndrome: role of triggers and complement abnormalities in the response to C5 inhibition

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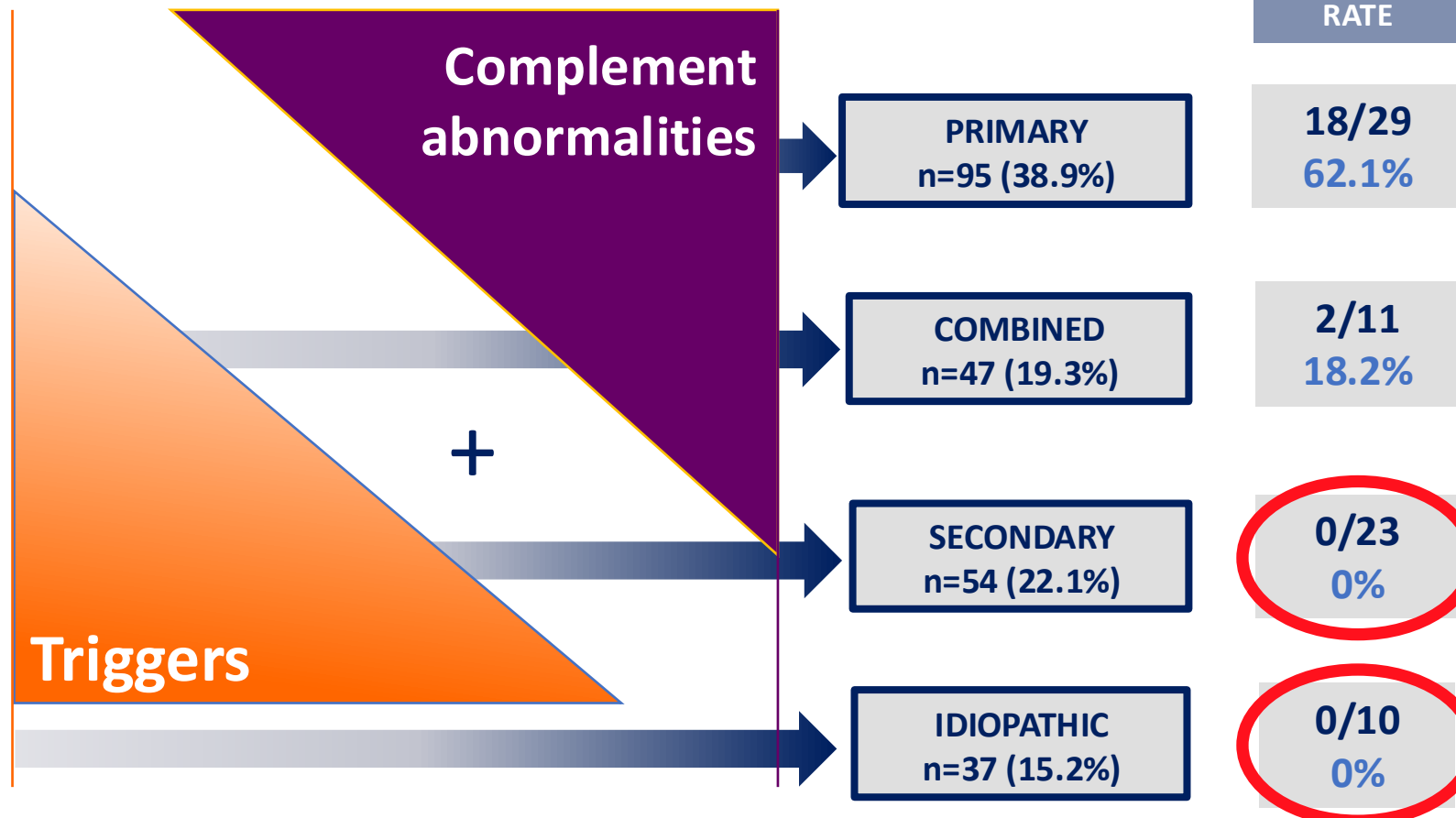
Long-term outcome								
Conventional treatment n: 102				C5 inhibitor n: 142				P*
sCr <1.5 mg/dL	sCr ≥1.5 mg/dL	ESKD	Death	sCr <1.5 mg/dL	sCr ≥1.5 mg/dL	ESKD	Death	
15 (30.0)	12 (24.0)	19 (38.0)	4 (8.0)	32 (65.3)	12 (24.5)	4 (8.2)	1 (2.0)	<0.001
9 (37.5)	4 (16.7)	10 (41.6)	1 (4.2)	17 (60.7)	8 (28.6)	2 (7.1)	1 (3.6)	0.031
5 (35.7)	5 (35.7)	2 (14.3)	2 (14.3)	24 (58.6)	6 (14.6)	10 (24.4)	1 (2.4)	0.089
4 (28.6)	4 (28.6)	6 (42.8)	0 (0.0)	8 (33.3)	8 (33.3)	7 (29.2)	1 (4.2)	0.754

What is the best treatment schedule? Life long???

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Discontinuation



*Triggers defined as an acute or chronic condition that is thought to have contributed to the cause of disease.

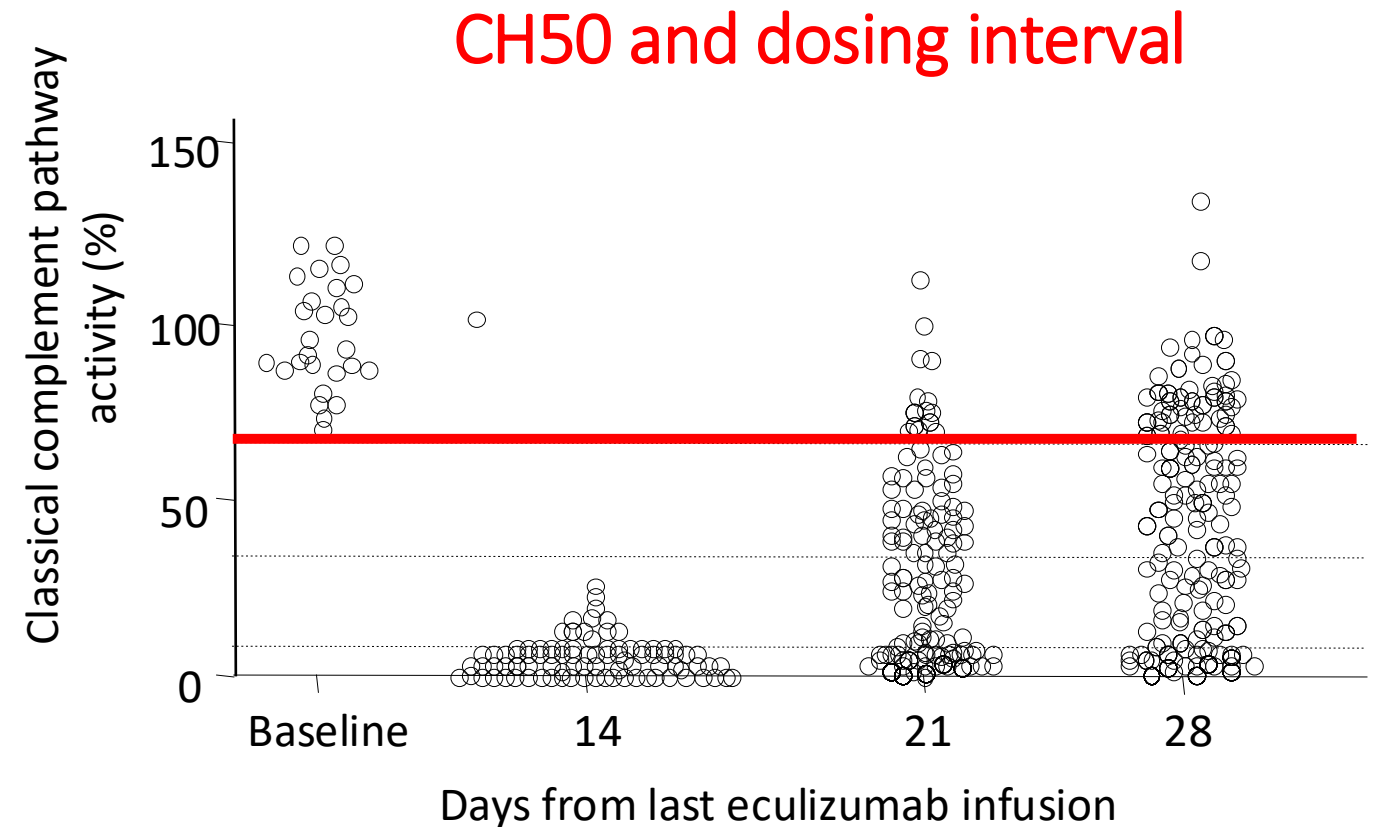
Eculizumab treatment in atypical hemolytic uremic syndrome: correlation between functional complement tests and drug levels

Massimo Cugno¹  · Valentina Capone² · Samantha Griffini¹ · Elena Grovetti¹ · Giulia Pintarelli³ · Luigi Porcaro³
Emilio Clementi⁴ · Gianluigi Ardissino²

Journal of Nephrology (2022) 35:1205–1211
<https://doi.org/10.1007/s40620-021-01187-8>

ORIGINAL ARTICLE

- the interval between doses was adjusted on classical complement pathway (CCP) activity, targeted to < 30% for the prevention of relapses
- 38 patients with aHUS (median age:25.0 years)
- 22 patients receive eculizumab every 28 days and 16 receive it every 21
- an inverse correlation between CCP activity and eculizumab circulating levels was present
- complement activity measurement can be used as a proxy for circulating levels of the drug



**Eculizumab treatment in atypical hemolytic uremic syndrome:
correlation between functional complement tests and drug levels**

**ALGORITHM
FOR TAILORING ECULIZUMAB TREATMENT**

If CH50 is $\leq 2\%$, the interval between administrations is increased of 1 wk

If CH50 is 3-30%, the interval is confirmed

If CH50 is $>30\%$, the interval is shortened of 1 wk

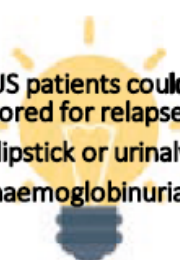
With the combination of discontinuation and tailored C5i the cost is reduced to 50%

Careful monitoring for disease relapse

Haemoglobinuria for the early identification of aHUS relapse.
Data from the ItaKid-HUS Network.

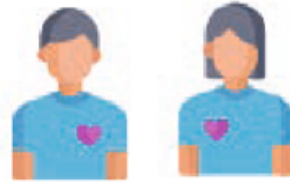
Journal of NEPHROLOGY


aHUS is a thrombotic microangiopathy involving the glomerulus and associated with renal damage. Thus it cannot take place without haematuria.

 aHUS patients could be monitored for relapses with urine dipstick or urinalysis for haemoglobinuria.

Retrospective study to analyse our experience on aHUS patients

- who have never be treated with C5 inhibition;
- who are on treatment;
- who have discontinued treatment.



84 patients



1517 determinations



8904 patient-month




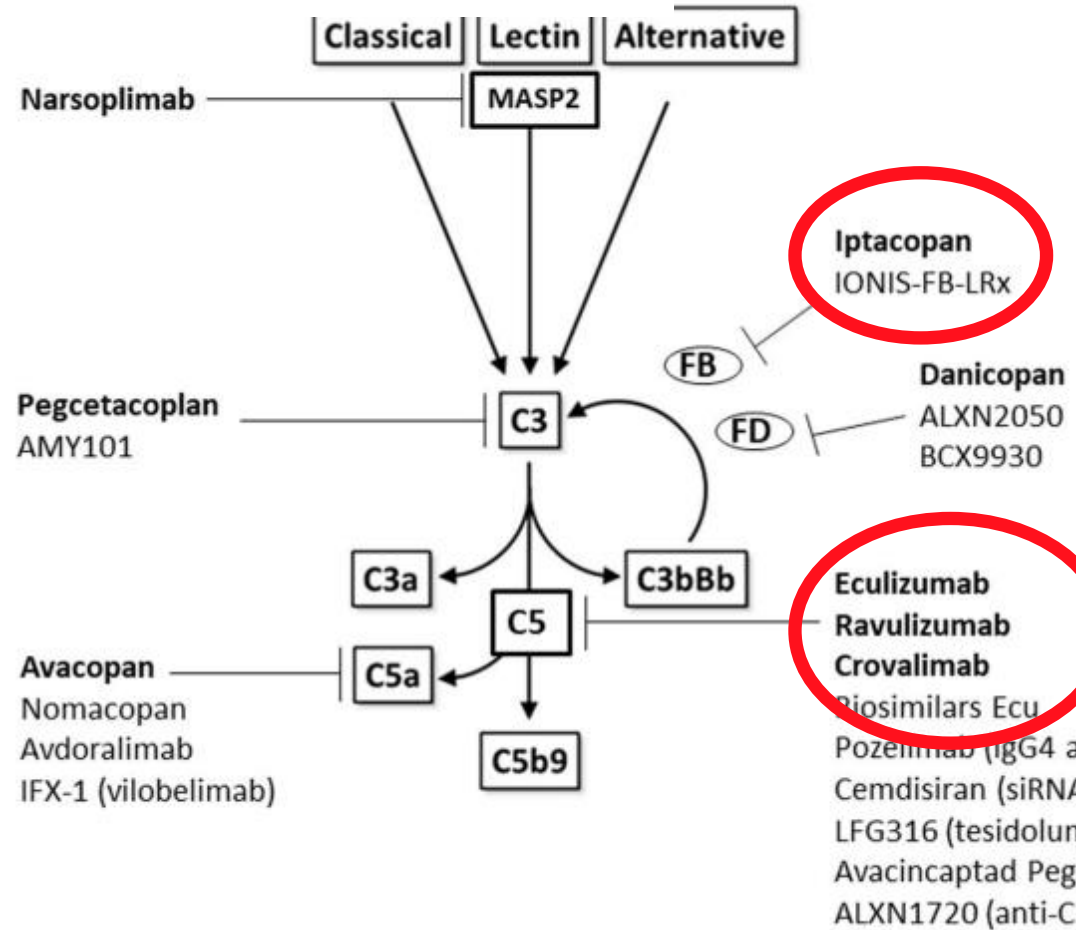
	Relapse n. (%)	No relapse n. (%)	Total
uHb+	21 (10.0)	188 (90.0)	209
uHb-	0 (0)	1308 (100)	1308
Total	21 (1.4)	1496 (98.6)	1517

Sensitivity 100% and Specificity 87.4%
PPV 10.5% and NPV 100 %.
Accuracy 87.6%.

Conclusion: Haemoglobinuria is a very sensitive and acceptably specific marker of aHUS relapse. This finding and its validation may have a positive impact on patients' quality of life and on the outcome of this life threatening disease via an early diagnosis of relapses.

Complement inhibitors in pediatric kidney diseases: new therapeutic opportunities

Luca Antonucci^{1,2} · Joshua M. Thurman³ · Marina Vivarelli^{1,4} 



Phase III for relapse-prevention
CFB inhibitor
OS daily
Not yet standard in acute aHUS

iv
iv, same target, longer half life, maintenance every 8 w
New kid, sc, every 4 weeks

Complement is especially critical for:

- Encapsulated bacteria
- Neisseria species
- Opsonization and immune complex clearance

Universal peculiarity (important !)

All terminal complement inhibitors (C5 blockers):

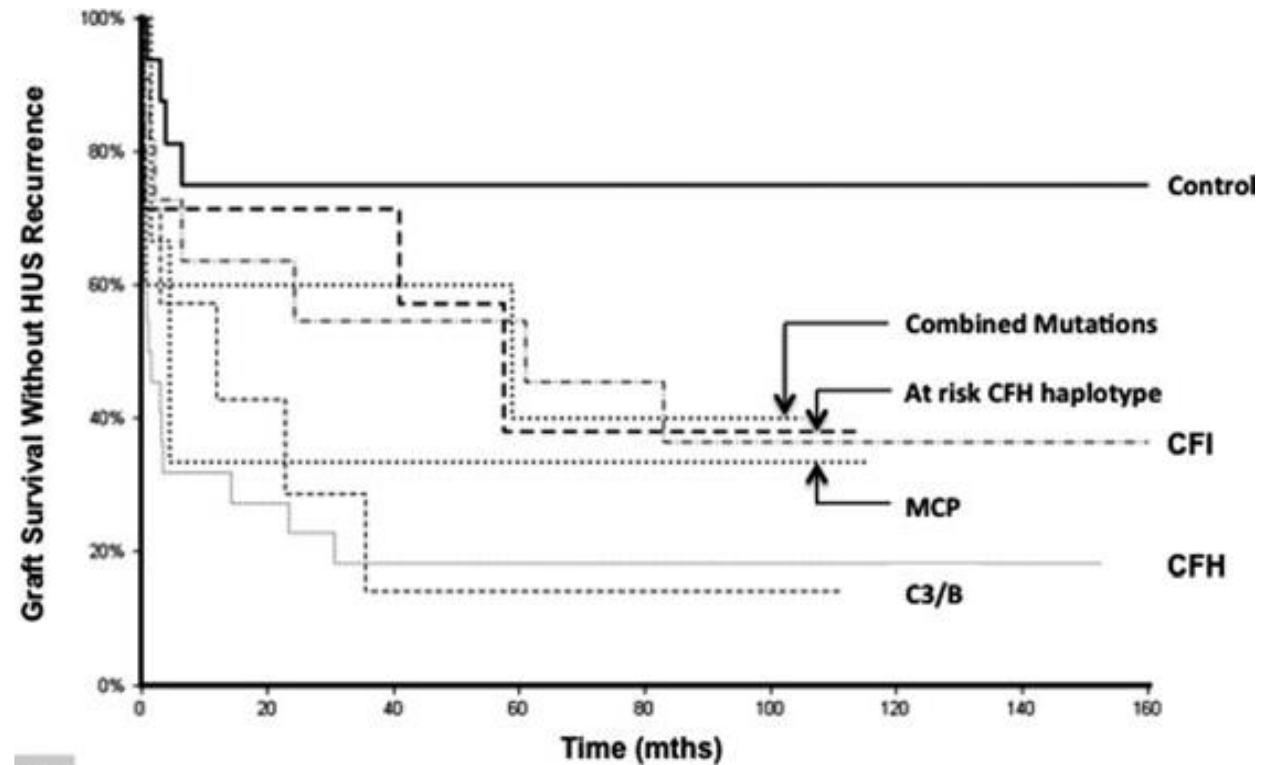
- ↑ risk of Neisseria infections
- Require MenACWY + MenB vaccines
- Consider antibiotic prophylaxis

.....For decades aHUS has been considered
a specific contraindication to kidney
transplantat for the very high risk of disease
recurrence and graft failure

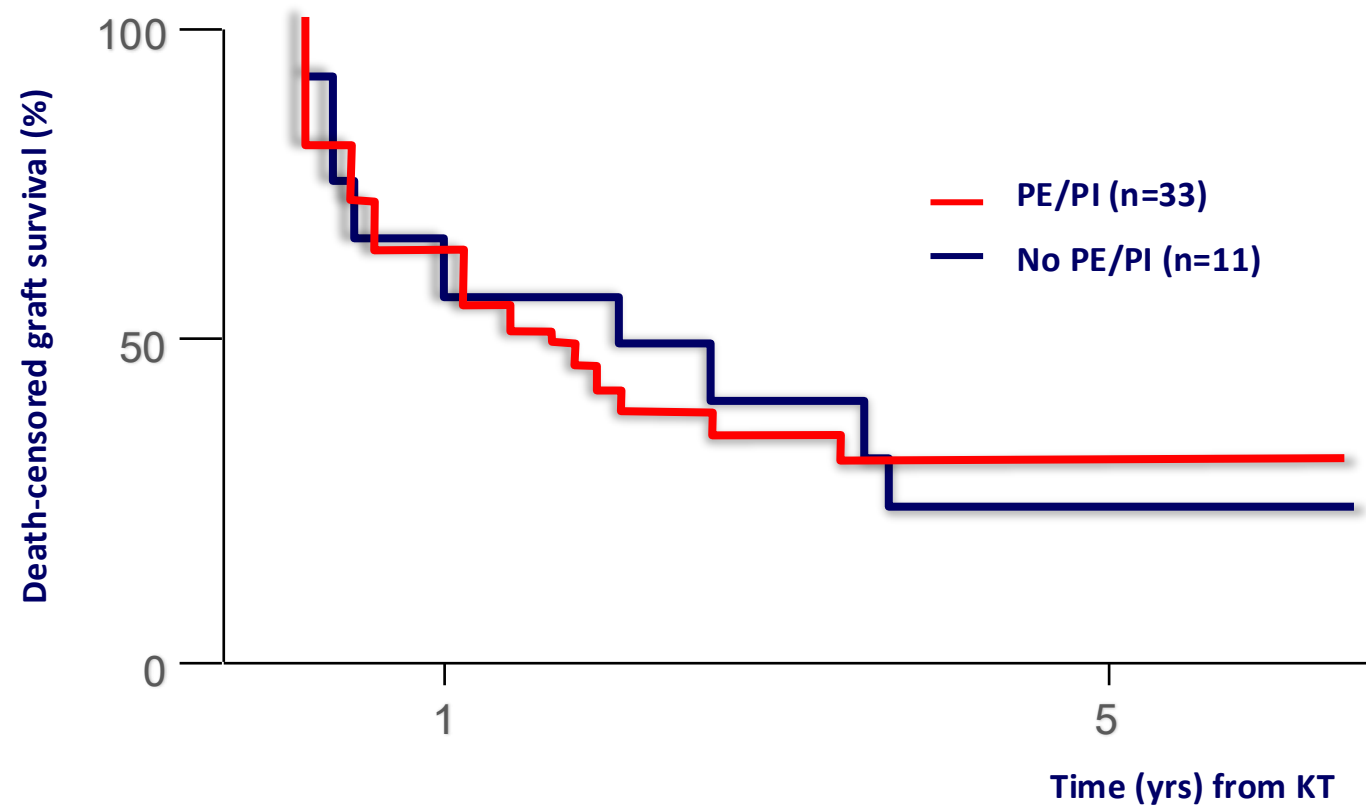
aHUS prognosis in the pre-Eculizumab era

Protein	ESRD (%)	Recurrence on KTx (%)
Factor H	75	90
Factor I	70	75
C3	60	50
MCP	<20	15

For many years, aHUS has been considered an absolute contraindication to KTx due to the high rate of recurrence and graft loss.



Graft survival following aHUS recurrence in patients treated or not with curative plasma therapy

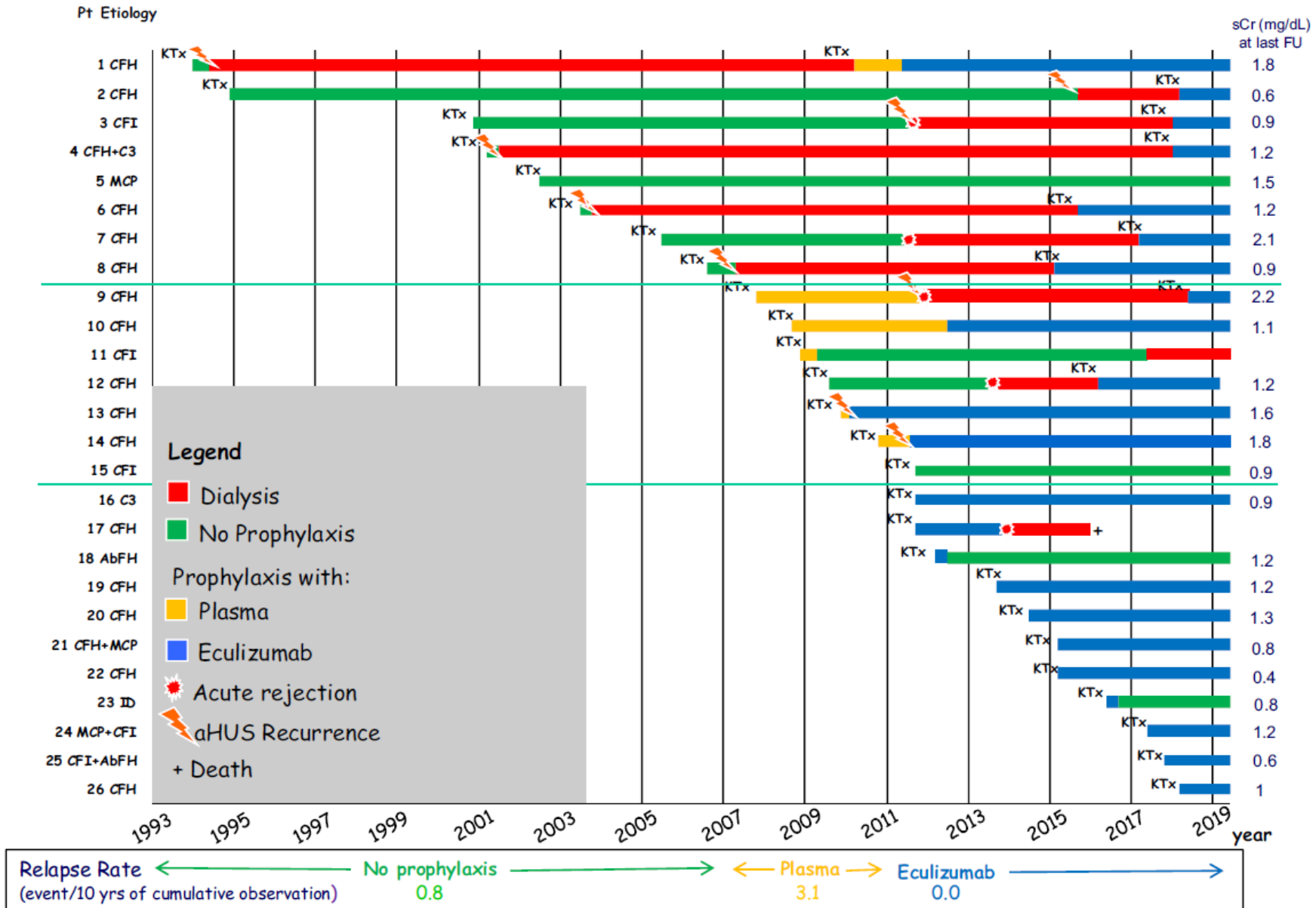


Historical representation of the experience with kidney transplantation in aHUS at Milano HUS Center

Kidney transplant in patients with atypical hemolytic uremic syndrome in the anti-C5 era: single-center experience with tailored Eculizumab

Gianluigi Ardissino¹ · Donata Cresseri² · Francesca Tel¹ · Antenore Giussani³ · Stefania Salardi⁴ · Martina Sgarbanti⁴ · Bice Strumbo⁴ · Sara Testa¹ · Valentina Capone¹ · Samantha Griffini⁵ · Elena Grovetti⁵ · Massimo Cugno⁵ · Mirco Belingheri² · Chiara Tamburello¹ · Evangeline Millicent Rodrigues¹ · Michela Perrone⁶ · Massimo Cardillo⁷ · Grazia Corti⁸ · Dario Consonni⁹ · Lucrezia Furian¹⁰ · Silvana Tedeschi⁴ · Piergiorgio Messa² · Claudio Beretta³

N. of pts.	26 (16 F)
N. of KTx:	35
Median time on RRT (yrs)	5.5
Median age at KTx (yrs)	33.5



AHUS PATIENTS' MANAGEMENT AIMED AT KTX: OUR EXPERIENCE

Ardissino G et al *Pediatr Nephrol* 2018;33:457–461

Pre-KTx: - identification and characterization of patients:

NGS for CFH, CFI, CFB, C3, MCP, THBD, DGKe
MLPA for macrodeletions and hybrid genes
Ab anti-CFH research (IgG, IgA and IgM)

- pre-KTx anti-C5 treatment

Peri-KTx: - Confirmation of remission (PLT, haptoglobin, LDH)

- Preoperative eculizumab at a dose appropriate to the patient's weight (adult 900 mg)
- Standard immunosuppressive regimen

P.O.:- Daily monitoring of disease recurrence indicators (PLT, haptoglobin, LDH)

- Subsequent doses of eculizumab based on the genetic abnormality, clinical condition, and evidence of complement suppression (CH50)

In MCP mutation, Ab AntiCFH (levels < 1000 IU/mL) or idiopathic forms: only pre-KTx administration

Post-KTx: - Maintaining anti-C5 therapy (Ravulizumab or Eculizumab)

- Careful monitoring for recurrence

Take home messages

- aHUS is a clinical entity that includes several different diseases that should be actively ruled-in or out
- Complement-mediated aHUS has an excellent outcome with C5i but treatment should be started early for a better outcome
- Idiopathic aHUS do not respond to C5i
- Both Secondary and Idiopathic aHUS do not relapse after discontinuation of C5i

Take home messages

- Urine dip-stick for uHb is very sensitive to identify relapses when patients are off treatment
- The interval between doses of C5i can be increased if CH50 activity is regularly monitored
- New drugs will be available for the prevention of relapses of complement-mediated aHUS among which FBi seems the most promising

Complement-mediated HUS

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