

# ***Hemolytic Uremic Syndrome (HUS) and Thrombotic Thrombocytopenic Purpura (TTP)***

***Jean Francis MD***

***Boston University Medical Center***

***Professor of Medicine, Renal Section***

***Co-director of ARC on Thrombosis and TMA Collaborative***

***Associate physician BWH***

# Jean Francis, MD



- *Boston University Chobanian and Avedisian School of Medicine*
- *Medicine Residency @Yale-HSR Campus*
- *Nephrology Fellowship @ Yale-HSR Campus*
- *Transplant Fellowship @ BIDMC*
- *Professor of Medicine @ BUSM*
  - Clinical focus: kidney and pancreas Transplant
  - Research focus: Thrombosis/TMA

# DISCLOSURE

## Relevant Financial Relationships

Advisory board and consulting: Alexion  
Vertex, Novartis, Apellis, UpToDate

## Off Label Usage

Yes

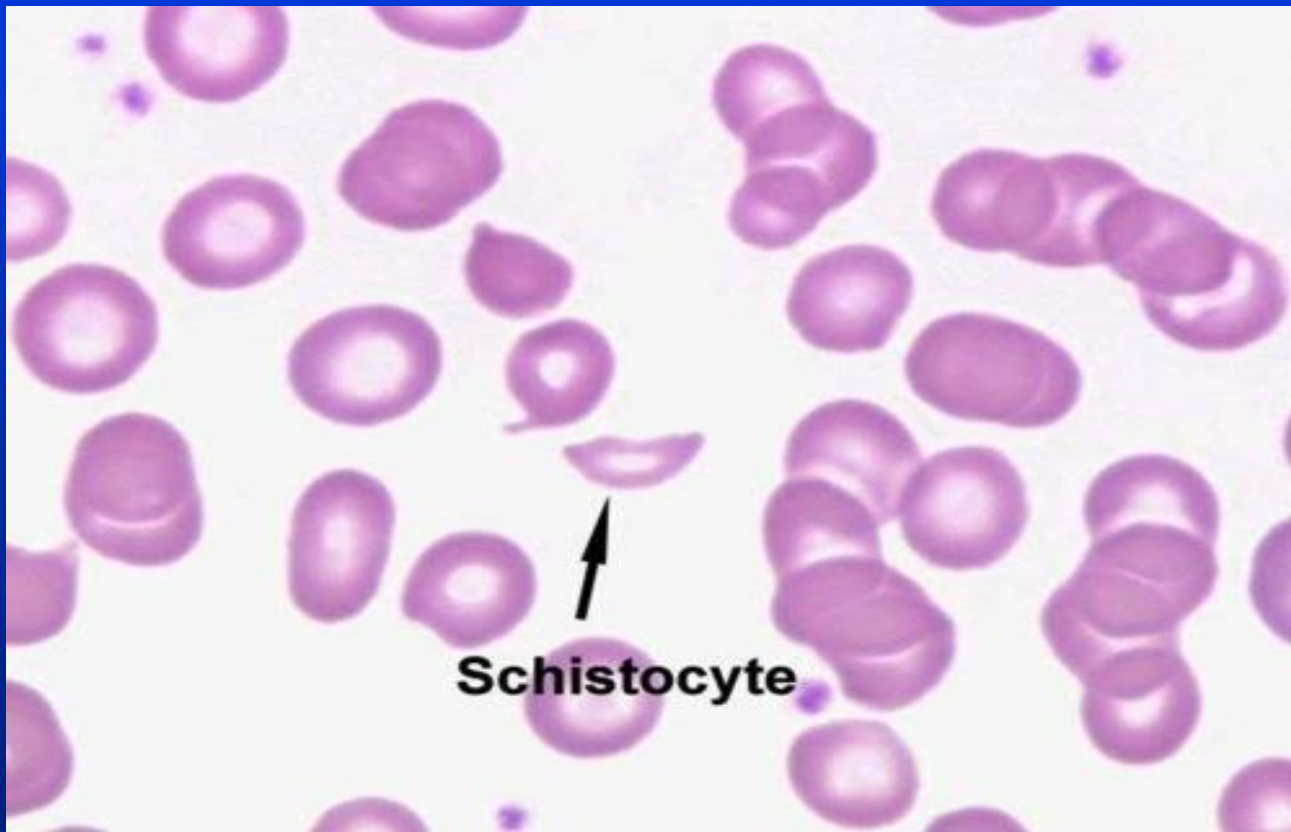
# Case Presentation: 1

- 44 years old previously healthy woman admitted to the hospital with weakness, fatigue, and malaise for few days. She had no fever, chills, and no recent travel or sick contacts. She reports that a week ago she developed some upper respiratory viral infection which resolved in 3 days.
- She has no known past medical history and currently she is not taking any medications.
- Physical exam: BP 157/87, HR 98, Temp 99F, RR 16, Pale, no icterus, AAOx3
- Lungs CTA bilaterally
- Heart: normal exam, except for mild tachycardia
- GI: Normal exam
- Skin no rash
- Neurological exam: normal

# Case Presentation: 1

- Blood workup demonstrated:
- AKI, anemia and thrombocytopenia
  - Creatinine 3.9 mg/dL from baseline 1.0 mg/dL
  - Platelets 64 k/uL, Hb 6.8 g/L, PT and PTT normal, fibrinogen normal,
- Workup of anemia: LDH 648, haptoglobin <8
- Coombs negative
- Schistocytes seen on peripheral smear
- Serum creatinine continued to increase
  - Creatinine 8.5 mg/dL on Hospital Day #5
  - C3 and C4 normal
  - ANA, dsDNA negative, SS-A, SS-B neg

# Peripheral Smear



2 or > schistocytes/hpf on blood smear

# Case Presentation: 1-

## Question 1

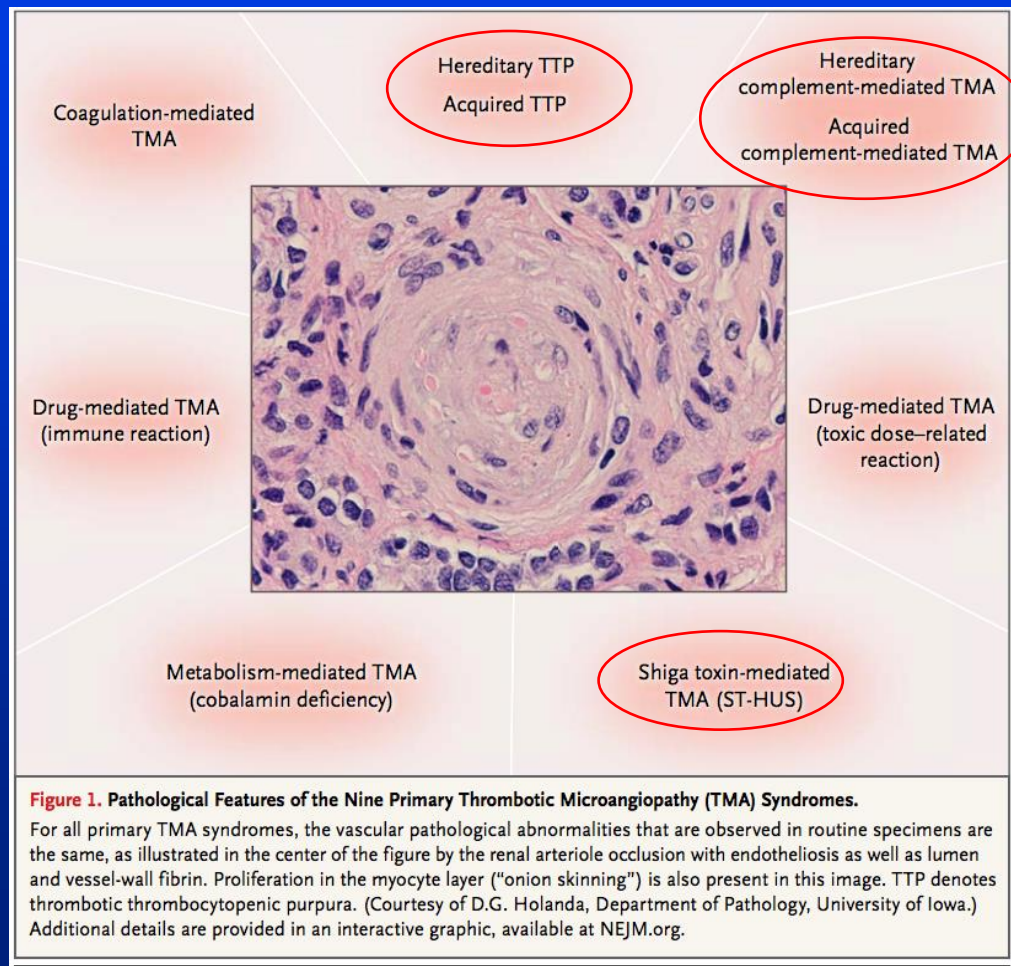
- What is the underlying pathophysiologic entity associated with this presentation:
  - a- Autoimmune hemolytic anemia
  - b- Thrombotic microangiopathy (TMA)
  - c- Hemolytic uremic syndrome (HUS)
  - d- Thrombotic thrombocytopenic prurpura (*TTP*)

# TMA syndromes

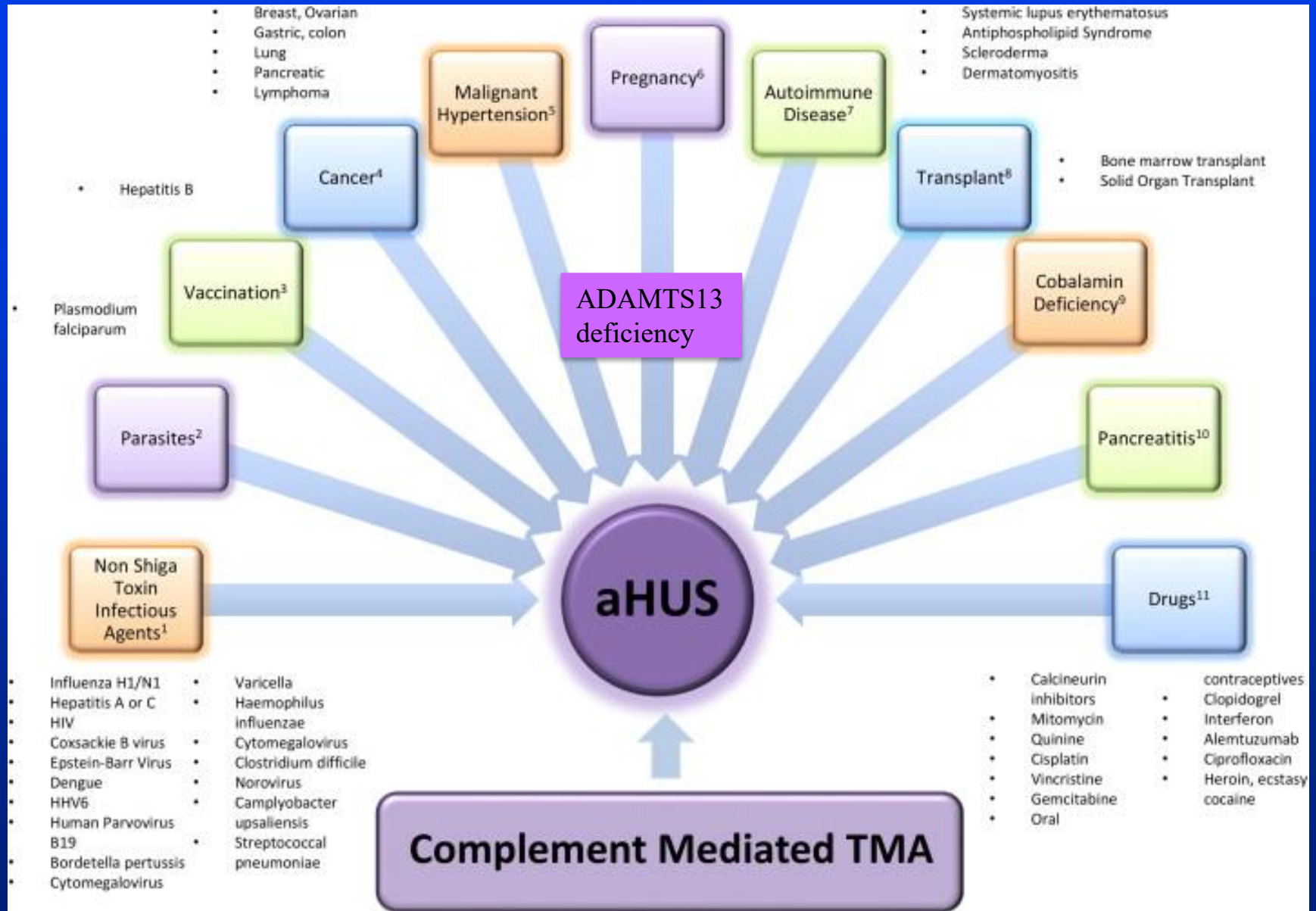
- Extremely diverse syndromes
- Hereditary or acquired
- Affect children and adults
- Clinically: Microangiopathic hemolytic anemia (MAHA), thrombocytopenia, organ injury
- Pathology: arteriolar and capillary microthrombi, endothelial injury



# TMA Pathology



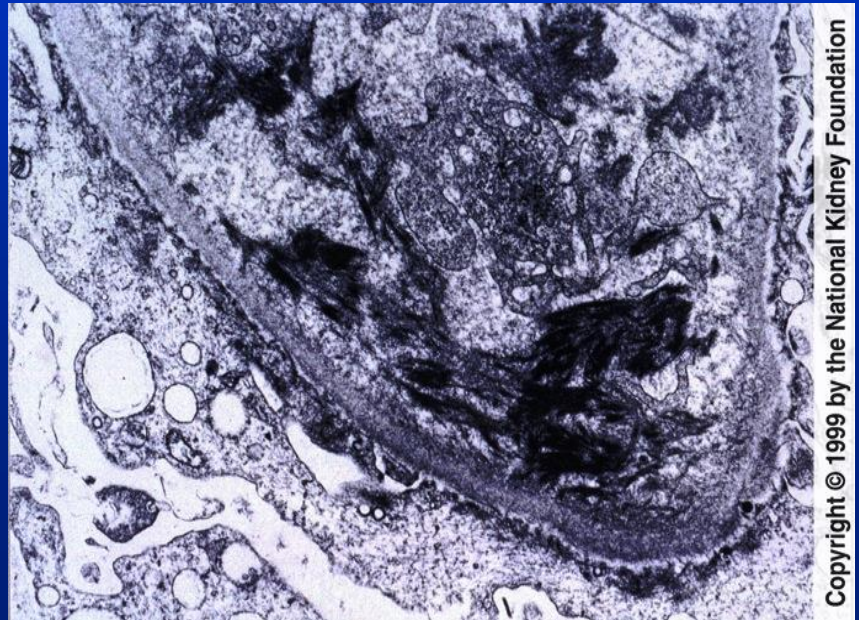
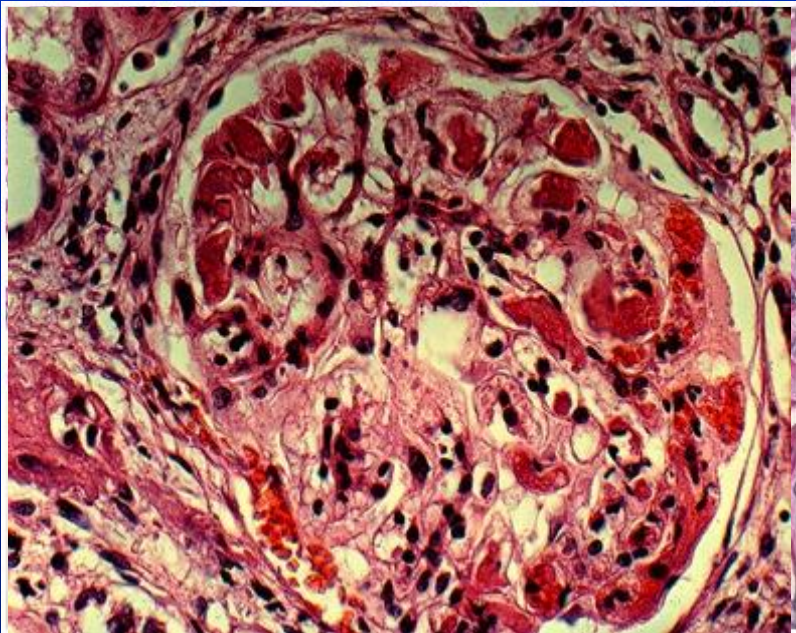
# Differential Diagnosis of TMA





# Case 1 continued...

- Kidney biopsy performed on hospital day #5
- Demonstrated active and chronic TMA



# Case Presentation: 1-

## Question 2

- Plasma exchange (PEX) started on Hospital Day #5
- Hemodialysis initiated on HD #5
  - MAHA and thrombocytopenia persisted despite PEX therapy daily for 5 days, and patients remained dialysis dependent
  - ADAMTS13 78% and APLA neg, STEC neg

What is the best next step?

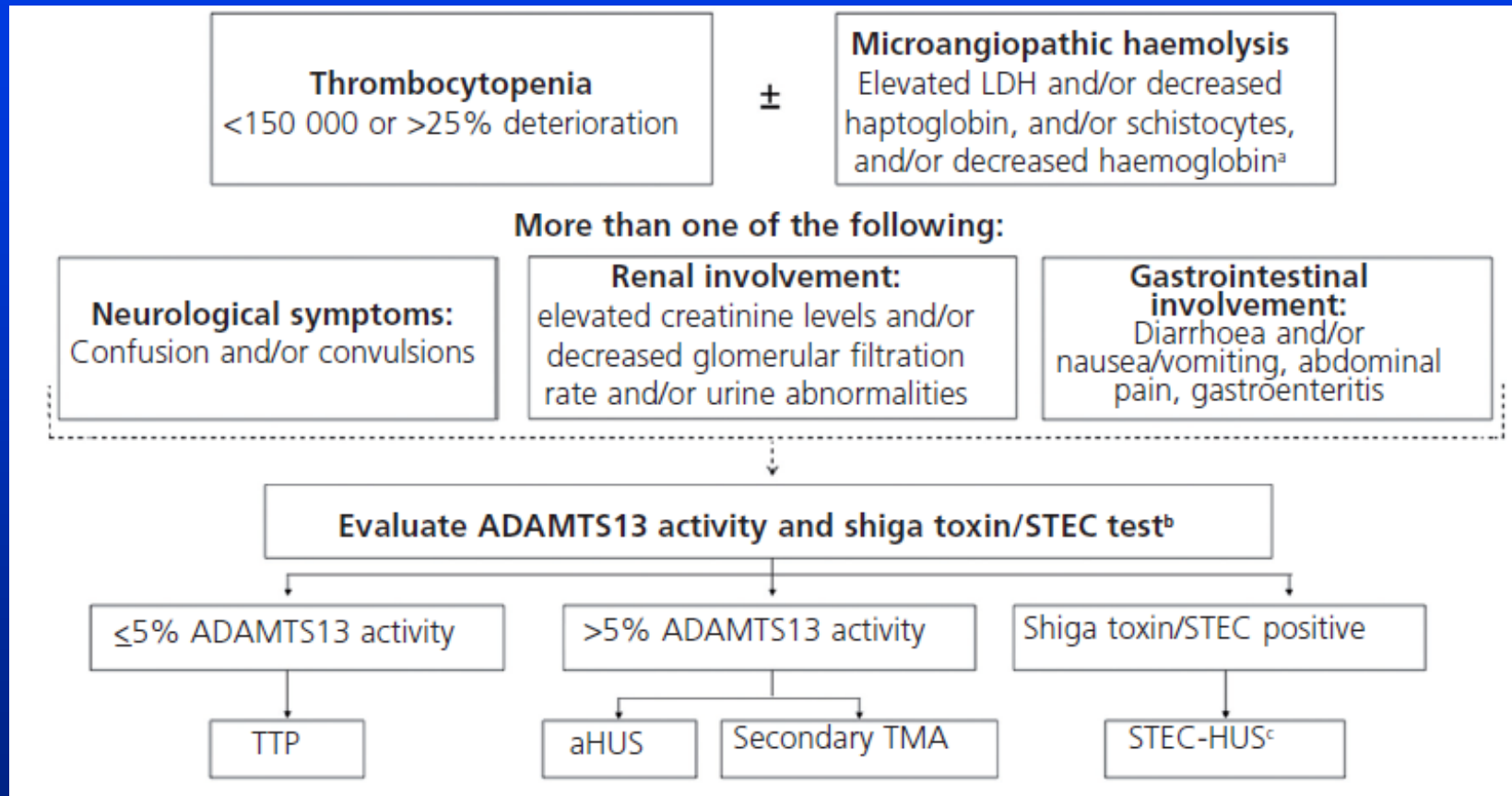
- a- continue PEX for another week
- b- Start plasma infusion
- c- Consider initiation of rituximab
- d- Eculizumab

# Case Presentation: 1-

## Question 3

- Before the administration of eculizumab, you should:
  - a- vaccinate the patient for *Neisseria meningitidis*
  - b- vaccinate the patient for *Neisseria meningitidis* and start at least 2 weeks of antibiotic prophylaxis for *N. Meningitidis* (e.g ciprofloxacin)
  - c- Monitor for any signs of meningitis and treat as needed
  - d- no further intervention is needed, proceed with eculizumab

# Summary: Evaluation of TMA



While waiting for  
ADAMTS13 result Plts>  
30,000 and creatinine >2.25  
mg/dl almost rules out TTP

Campistol Nefrologia 2013, Nester C Blood Purif 2013

Cataland et al, blood, 2014 x Volume 123, Number 16

# Plasmic Score

| Variables                                                                                 | Points* |
|-------------------------------------------------------------------------------------------|---------|
| Platelet count <30 x 10 <sup>9</sup> per L                                                | 1       |
| Hemolysis variable†                                                                       | 1       |
| No active cancer                                                                          | 1       |
| No history of solid-organ or stem-cell transplant                                         | 1       |
| MCV <90 fL                                                                                | 1       |
| INR <1.5                                                                                  | 1       |
| Creatinine <2.0 mg/dL                                                                     | 1       |
| INR: International normalized ratio. MCV: Mean corpuscular volume.                        |         |
| †Reticulocyte count >2.5%, or haptoglobin undetectable, or indirect bilirubin >2.0 mg/dL. |         |

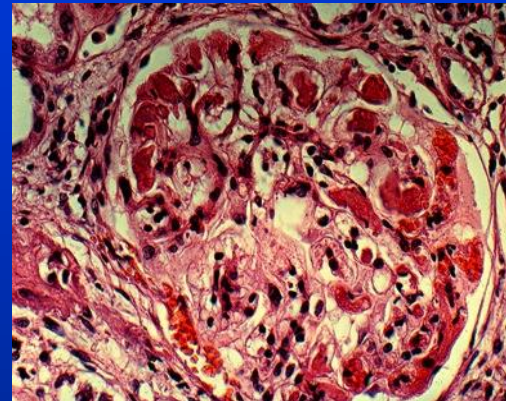
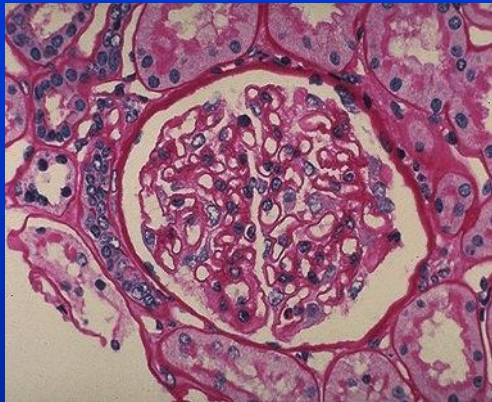
| Score | Risk Category | Risk of severe ADAMTS13 deficiency (≤ 10%) |
|-------|---------------|--------------------------------------------|
| 0-4   | Low           | 4.3%                                       |
| 5-6   | Intermediate  | 56.8%                                      |
| 7     | High          | 96.2%                                      |



# Hemolytic Uremic Syndrome (HUS)

A thrombotic microangiopathy manifesting with:

- Micro-angiopathic hemolytic anemia
- Thrombocytopenia
- Acute renal failure



Classic HUS-D<sup>+</sup> - diarrheal prodrome associated with shiga toxin producing 0157:H7 E.coli; most cases recover.

Atypical HUS - May be associated with mutations in complement regulatory proteins. Poor prognosis, 50% ESRD, may recur after transplantation.



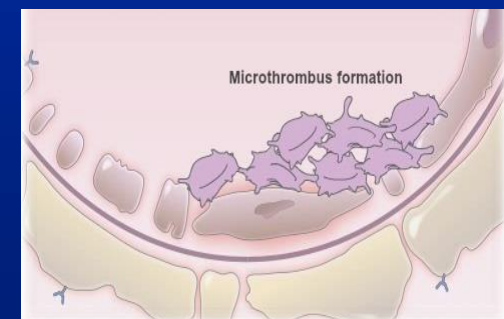
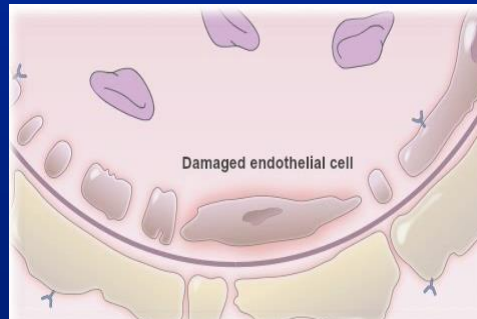
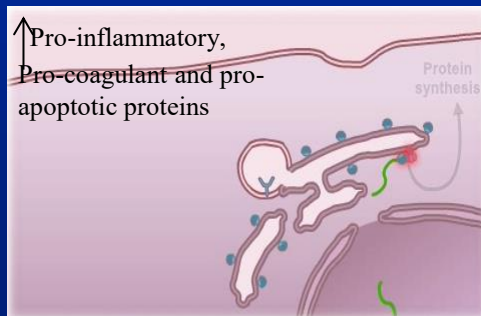
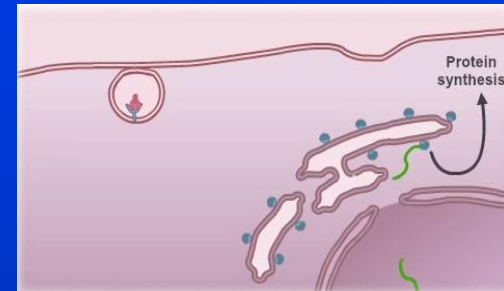
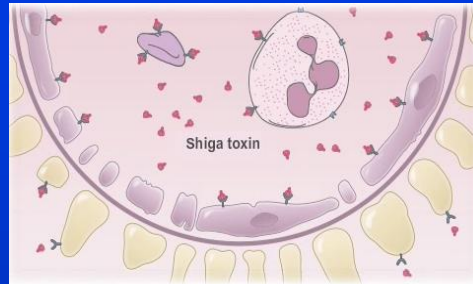
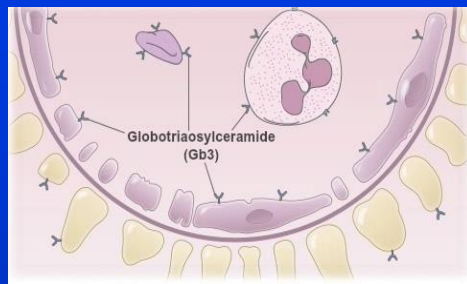
# Shiga Toxin HUS

- Induced by enteric infection with shiga toxin secreting strain of Enterohemorrhagic E Coli (O157:H7, O104:H4) or Shigella (contaminated water, vegetables, beef products etc..)
- Common in children presenting with AKI
- 6-9% of STEC infected children develop HUS

# STEC-HUS

- Bloody diarrhea prodrome 5-10 days
- 60% require dialysis, mean time on dialysis: 10 d.
- 25% of affected patients have neurological symptoms
- 4% mortality
- 5-25% with long term morbidity (HTN, proteinuria, decreased GFR)
- Treatment is supportive
- Role of PEX or complement inhibition uncertain

# Shiga Toxin HUS: Pathophysiology



# Case Presentation: 1-

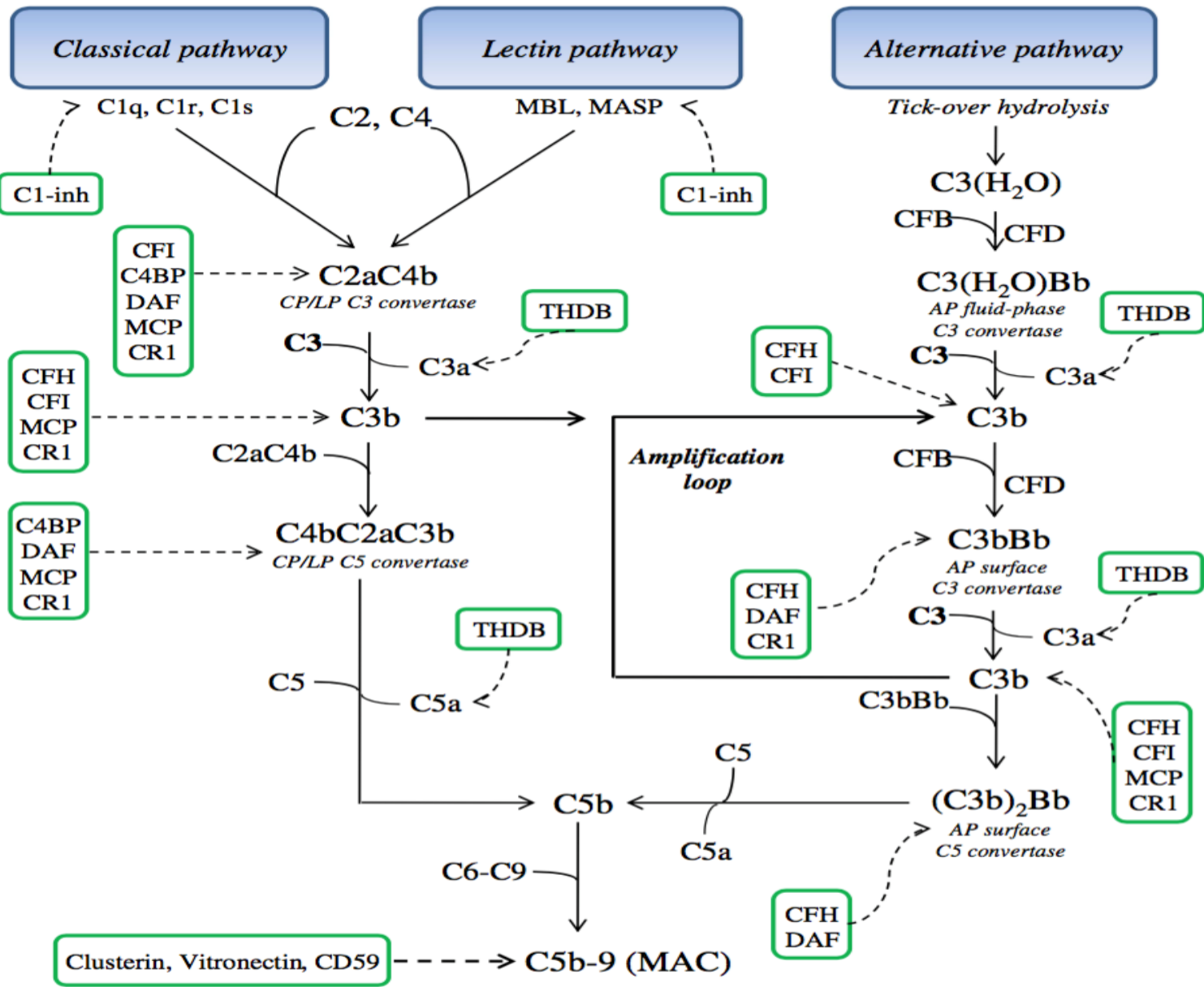
## Question 4

- Which one of the following statements is correct:
  - a- Normal complement levels rule out aHUS
  - b- The absence of complement system related genetic mutations rules out aHUS
  - c- the absence of family history of aHUS, rules out aHUS
  - d- none of the above

# Atypical HUS

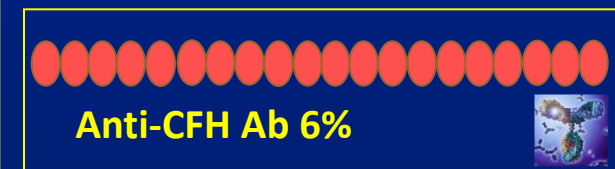
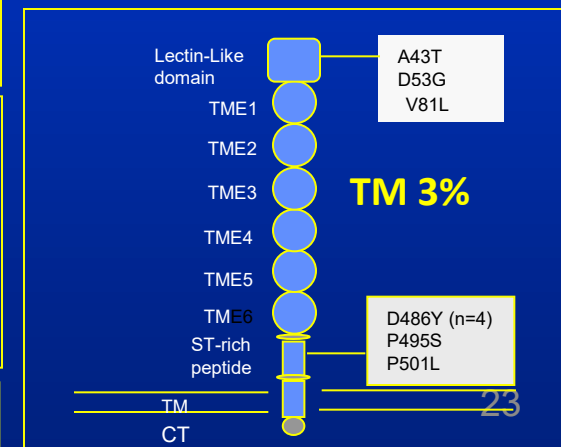
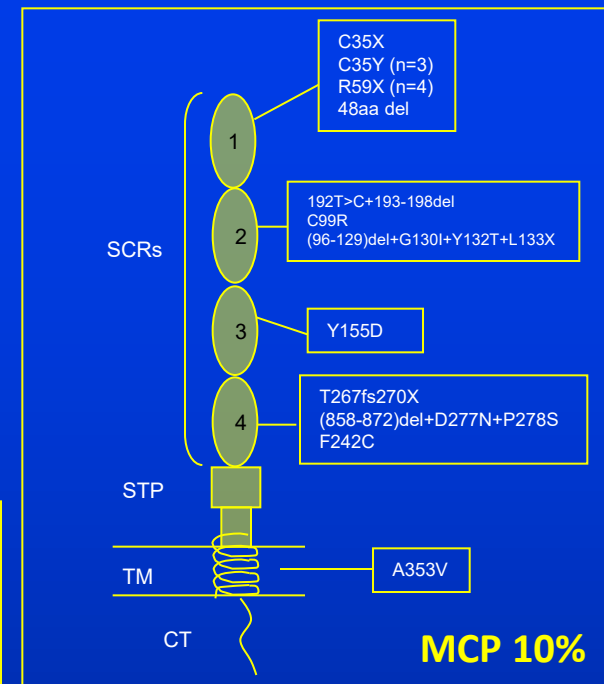
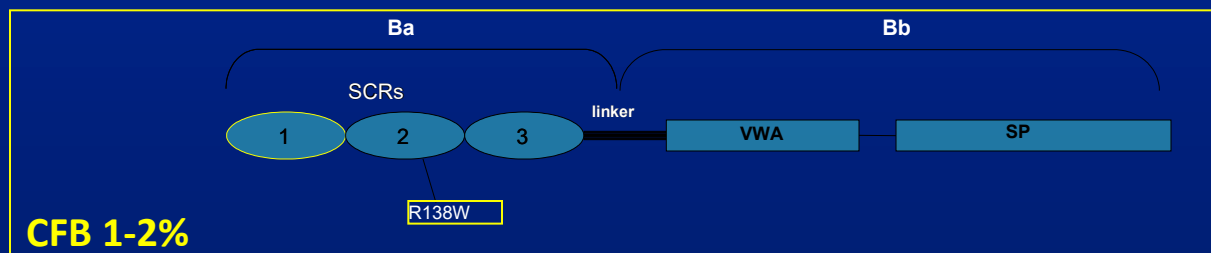
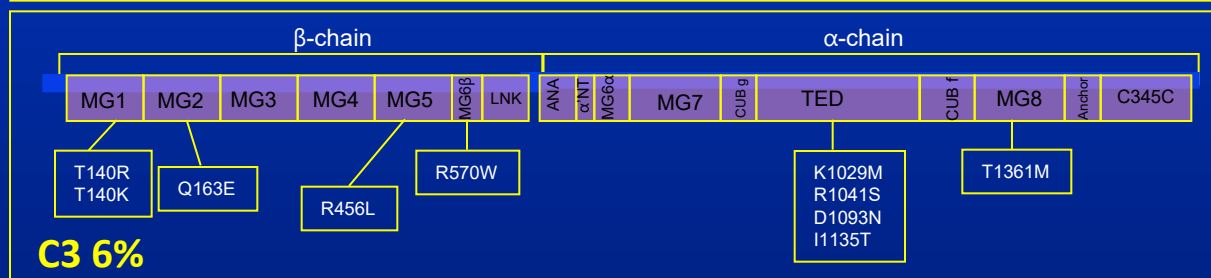
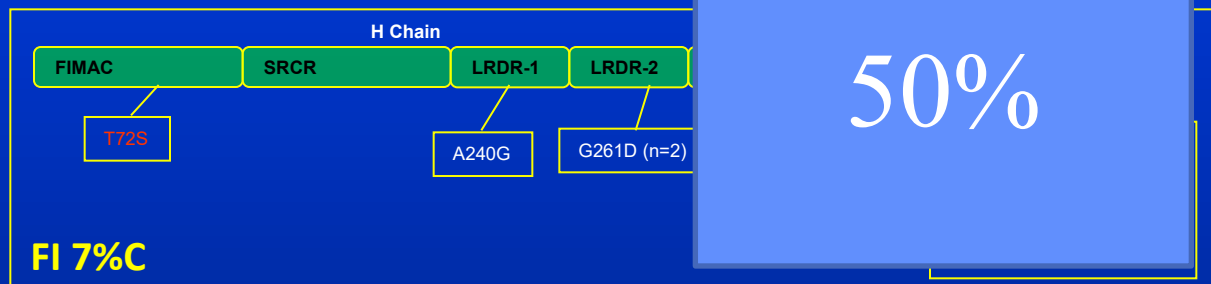
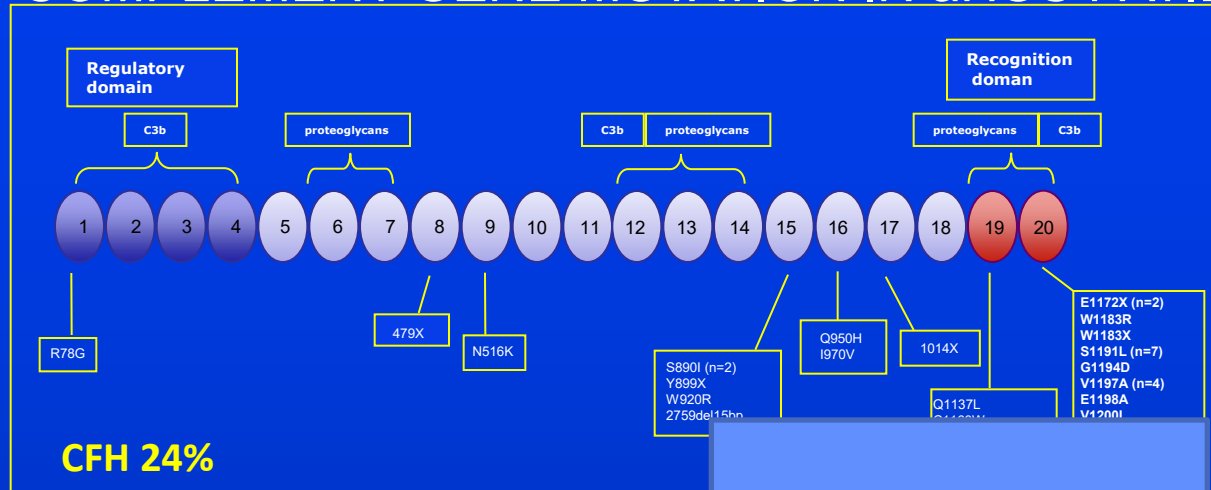
## Atypical HUS:

- is not induced by Shiga toxin+
- is often recurrent, 50% of patient have normal complement level and normal sC5b-9 (sMAC)
- may cause permanent kidney damage
- may cause neurological and other organ damage
- frequently recurs after transplantation
- may be sporadic or familial
- linked to abnormalities of complement regulation

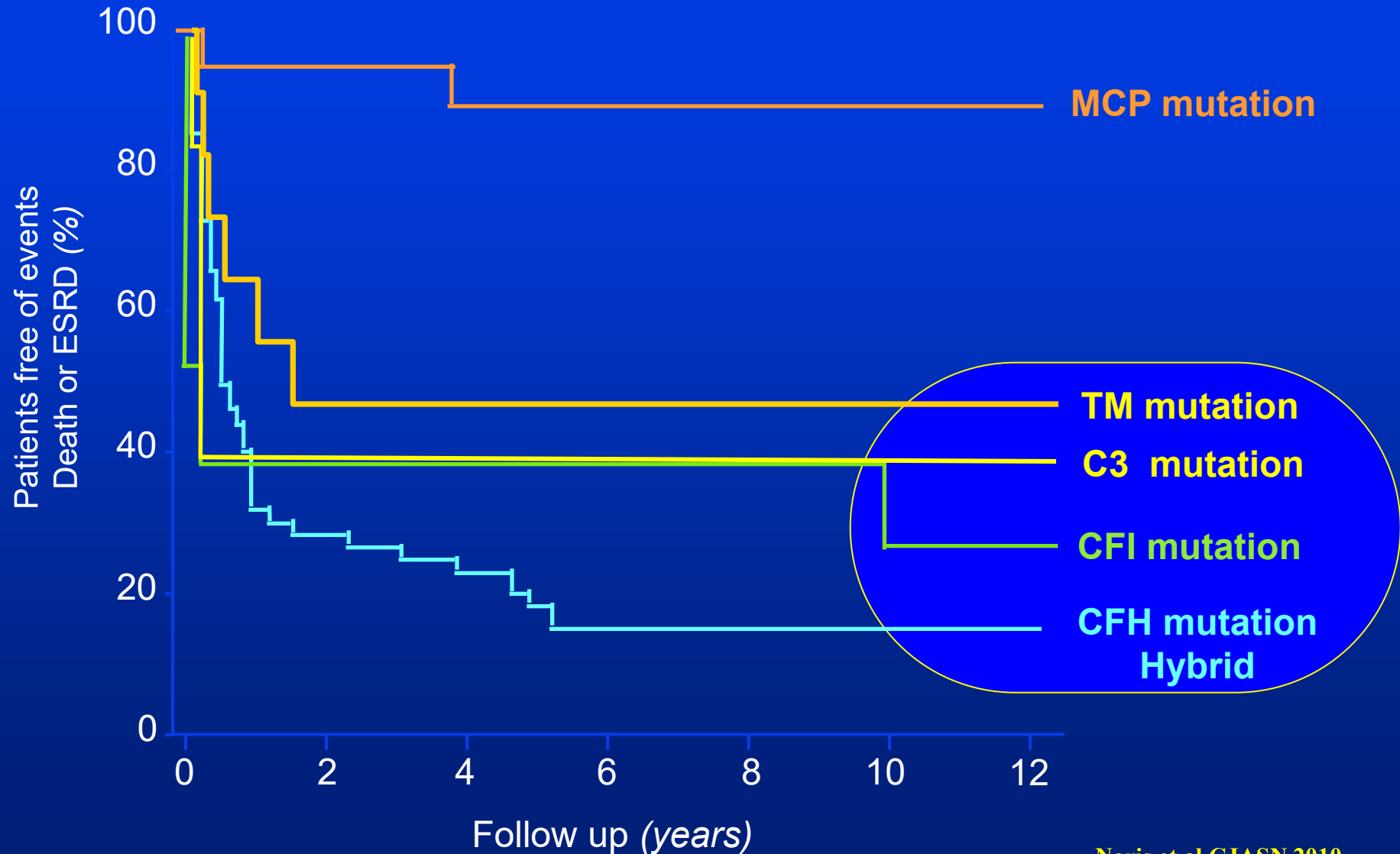




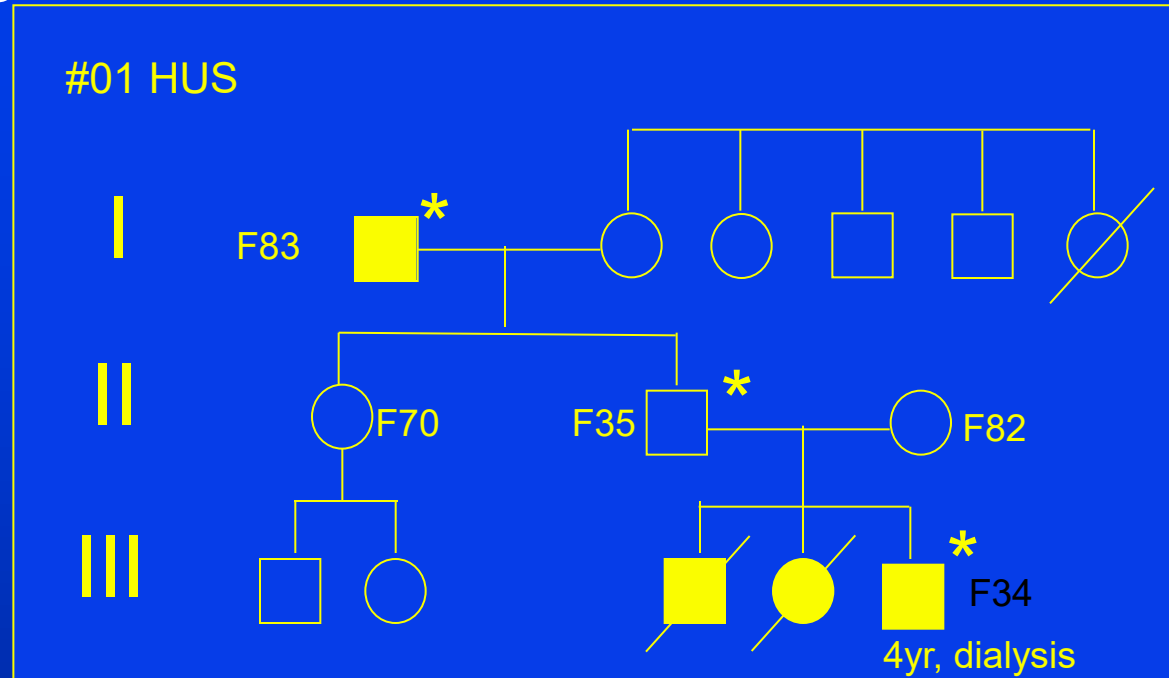
# COMPLEMENT GENE MUTATION IN aHUS PATIENTS



# LONG TERM OUTCOME OF aHUS PATIENTS



# INCOMPLETE PENETRANCE OF aHUS IN MUTATION CARRIERS



\* R1215Q change in CFH

- 3 subjects in the III generation developed aHUS in infancy: 2 died, 1 reached ESRD
- F35 never developed aHUS
- Subject F83, carrier of the R1215Q mutation developed aHUS and died at 82 years of age

# Atypical HUS may arise when there is:

---

- Deficiency, dysfunction or autoantibody-mediated inhibition of one or more of the regulatory proteins
- Resistance of C3b or factor B to decay by factors H, I and MCP
- Mutation of thrombomodulin, an endothelial glycoprotein with cytoprotective and anticoagulant properties
- The disease may be quiescent for months or years, even in patients with homozygous mutations and complete deficiency, only to be triggered by an otherwise innocuous infection, drug exposure or pregnancy

## *Eculizumab/Ravulizumab*

The diagram illustrates the activation of the complement system and its downstream effects on endothelial cells. It is divided into two main sections: the left section shows the activation of the complement cascade, and the right section shows the effects of the resulting complement components on endothelial cells.

**Complement Activation (Left Section):**

- Trigger:** A yellow arrow labeled "Trigger" points to the C3 protein (blue circle).
- C3 Activation:** C3 is cleaved into C3a (small blue fragment) and C3b (larger blue fragment).
- C3 Convertase:** C3b binds to CFB (red oval) and CFI (grey dashed circle) to form the C3 convertase.
- C3 Convertase Action:** The C3 convertase cleaves C3 into C3a and C3b.
- C5 Activation:** C3b binds to Bb (red oval) and C5 (green circle) to form the C5 convertase.
- C5 Convertase Action:** The C5 convertase cleaves C5 into C5a (small green fragment) and C5b (larger green fragment).

**Effects on Endothelial Cells (Right Section):**

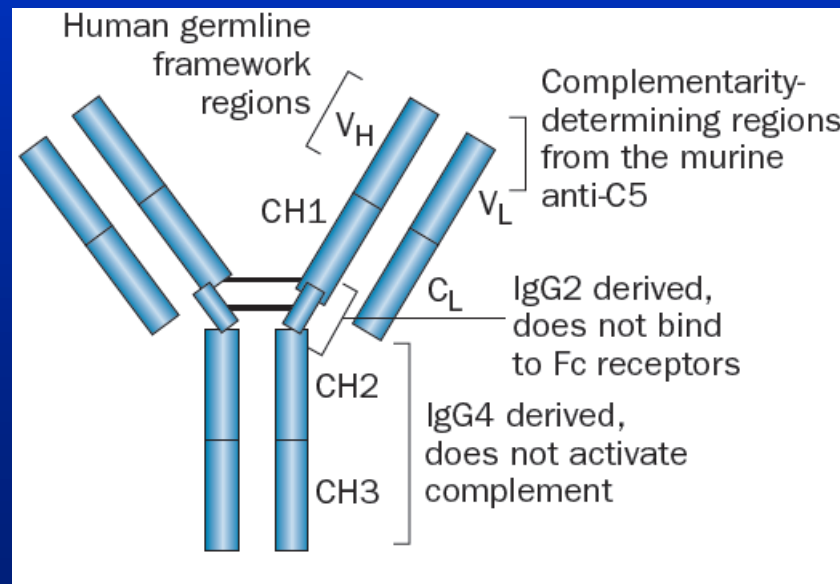
- Coagulation:** C5a (small green fragment) binds to the endothelial cell surface, leading to the activation of the coagulation cascade (represented by a yellow arrow).
- TM shedding:** C5b-9 (yellow oval) binds to the endothelial cell surface, leading to the shedding of TM (thrombomodulin, red structure).
- VWF shedding:** C5b-9 binds to the endothelial cell surface, leading to the shedding of VWF (von Willebrand factor, purple structure).
- P-selectin:** C5b-9 binds to the endothelial cell surface, leading to the shedding of P-selectin (purple structure).

**Endothelial cell:** The diagram shows two endothelial cells (purple structures) with various receptors and proteins on their surface.

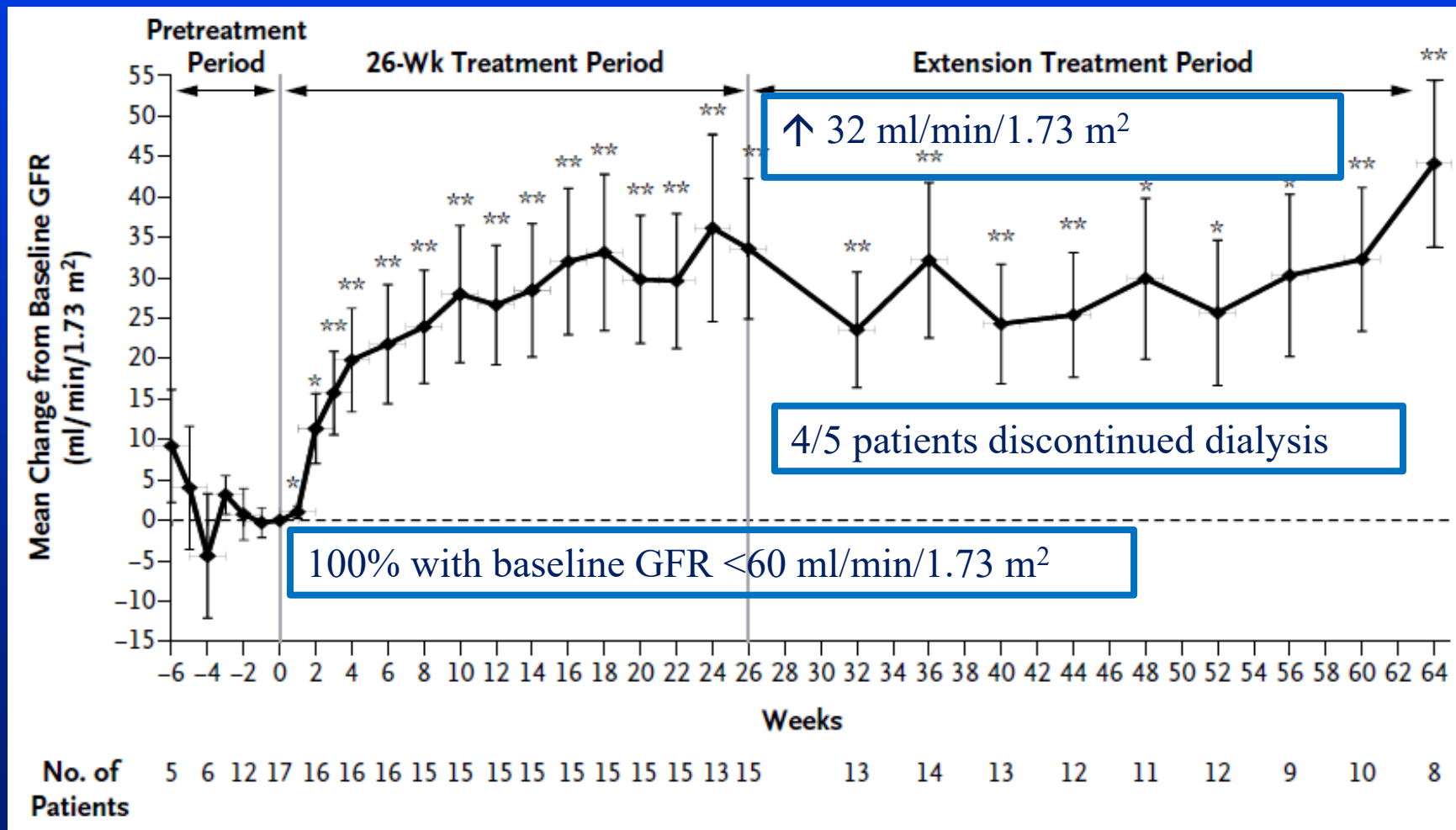
Legendre CM et al., *N Engl J Med*, 2013  
Licht C et al., *Kidney Int*, 2015

# Eculizumab

- Recombinant fully humanized hybrid IgG2/IgG4 monoclonal antibody against C5
- Prevents formation of C5a and C5b, component of the membrane attack complex

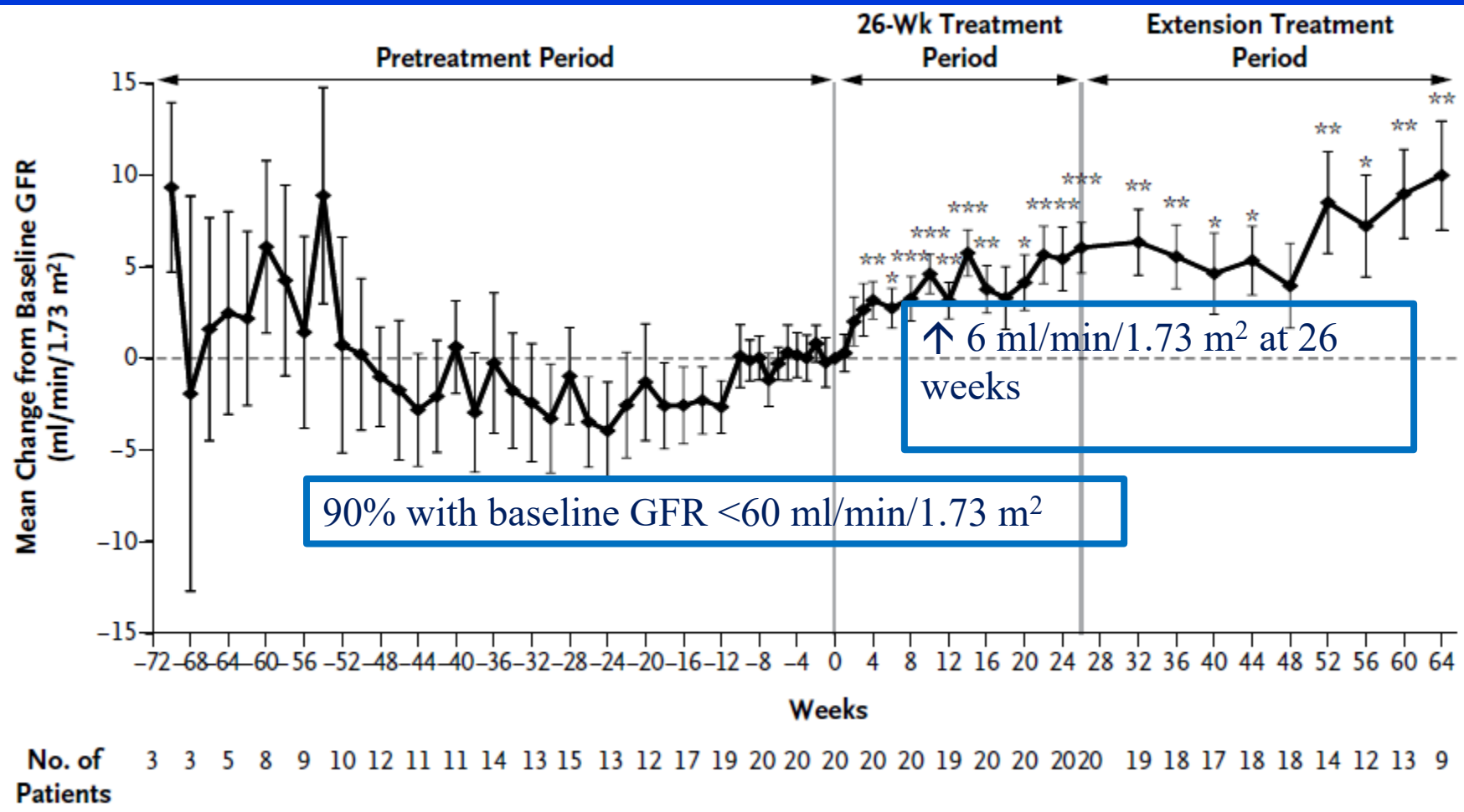


# Plasma-Resistant Trial: eGFR

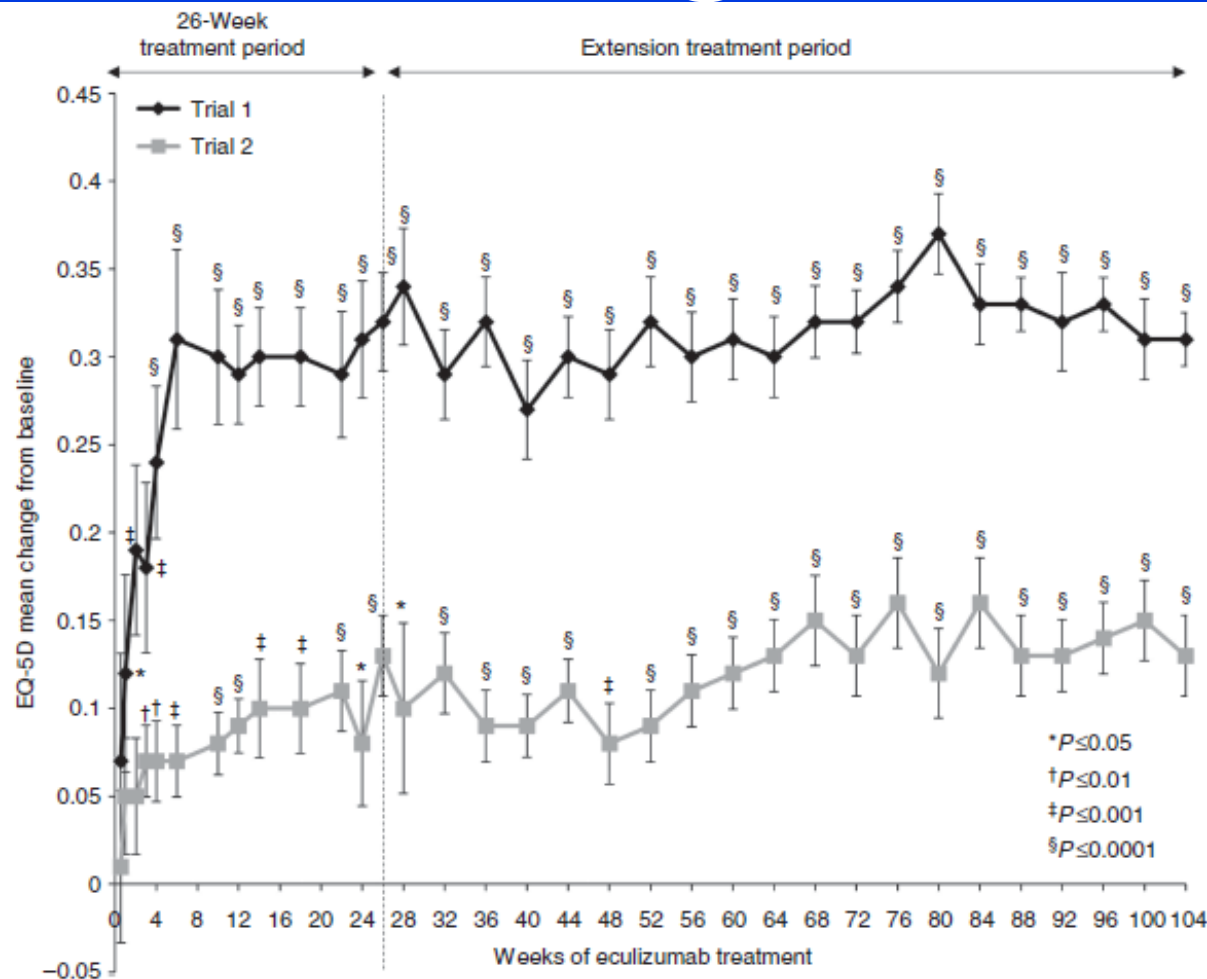




# Plasma-Sensitive Trial: eGFR



# Change in HRQoL



Clinically meaningful increase in 87%

Clinically meaningful increase in 73%

\* $P \leq 0.05$   
 $\dagger P \leq 0.01$   
 $\ddagger P \leq 0.001$   
 $\S P \leq 0.0001$

Trial 1 8 15 14 14 14 12 13 13 13 12 7 12 4 9 9 9 8 8 10 11 8 9 11 10 11 11 10 9 9 7 7 8  
 Trial 2 18 20 20 19 19 19 19 17 16 18 5 17 3 19 17 19 18 19 18 17 17 16 18 18 18 16 17 18 17 16 15

# Black Box Warning

## PATIENT SAFETY CARD

### PATIENT SAFETY INFORMATION CARD



Important Safety Information for  
Patients Taking Soliris®

Soliris can lower the ability of your immune system to fight infections, **especially meningococcal infection, which requires immediate medical attention.** If you experience any of the following symptoms, you should immediately call your doctor or seek emergency medical care, preferably in a major emergency medical care center.

- headache with nausea or vomiting
- headache and a fever
- headache with a stiff neck or stiff back
- fever of 103°F (39.4°C) or higher
- fever and a rash
- confusion
- muscle aches with flu-like symptoms
- eyes sensitive to light



**Get emergency medical care right away if you have any of these signs or symptoms and show this card.**

**Even if you stop using Soliris,** keep this card with you for 3 months after your last Soliris dose. Your risk of meningococcal infection may continue for several weeks after your last dose of Soliris.

# TRANSPLANTATION OUTCOMES

*aHUS recurrence  
(% failure)*

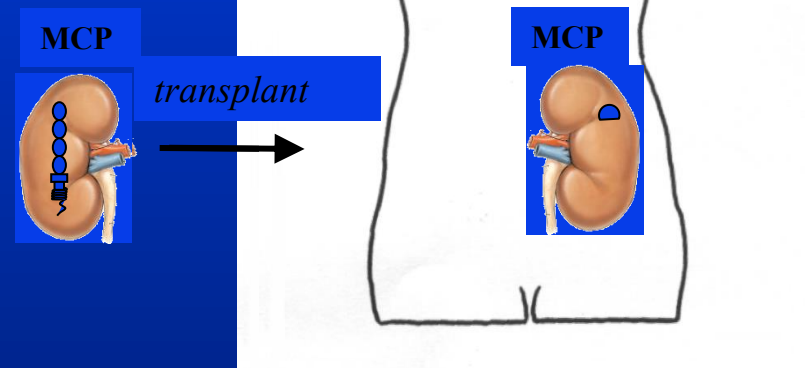
Patients with :

|       |            |                 |
|-------|------------|-----------------|
| - CFH | mutations  | 49 out 76 (82%) |
| - CFI | mutations  | 19 out 26 (95%) |
| - CFB | mutations  | 4 out 4 (100%)  |
| - C3  | mutations  | 16 out 30 (75%) |
| - CFH | antibodies | 5 out 17 (80%)  |
| - MCP | mutations  | 3 out 17 (66%)  |

22.6%  
■ single  
combined



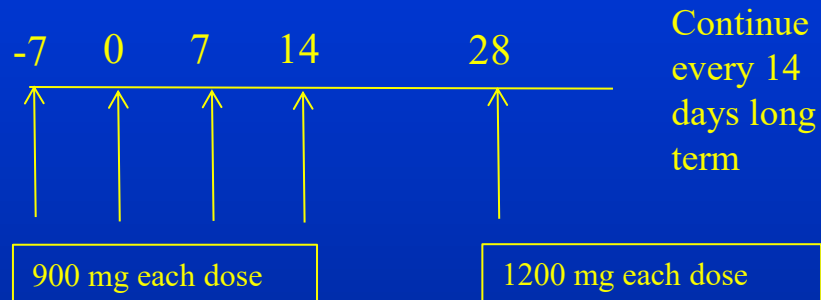
**MCP?**



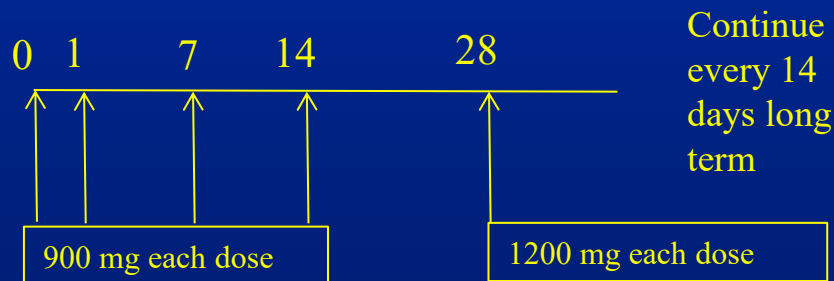
FH normal  
FH mutated

# Treatment Plan

- *Living donor*

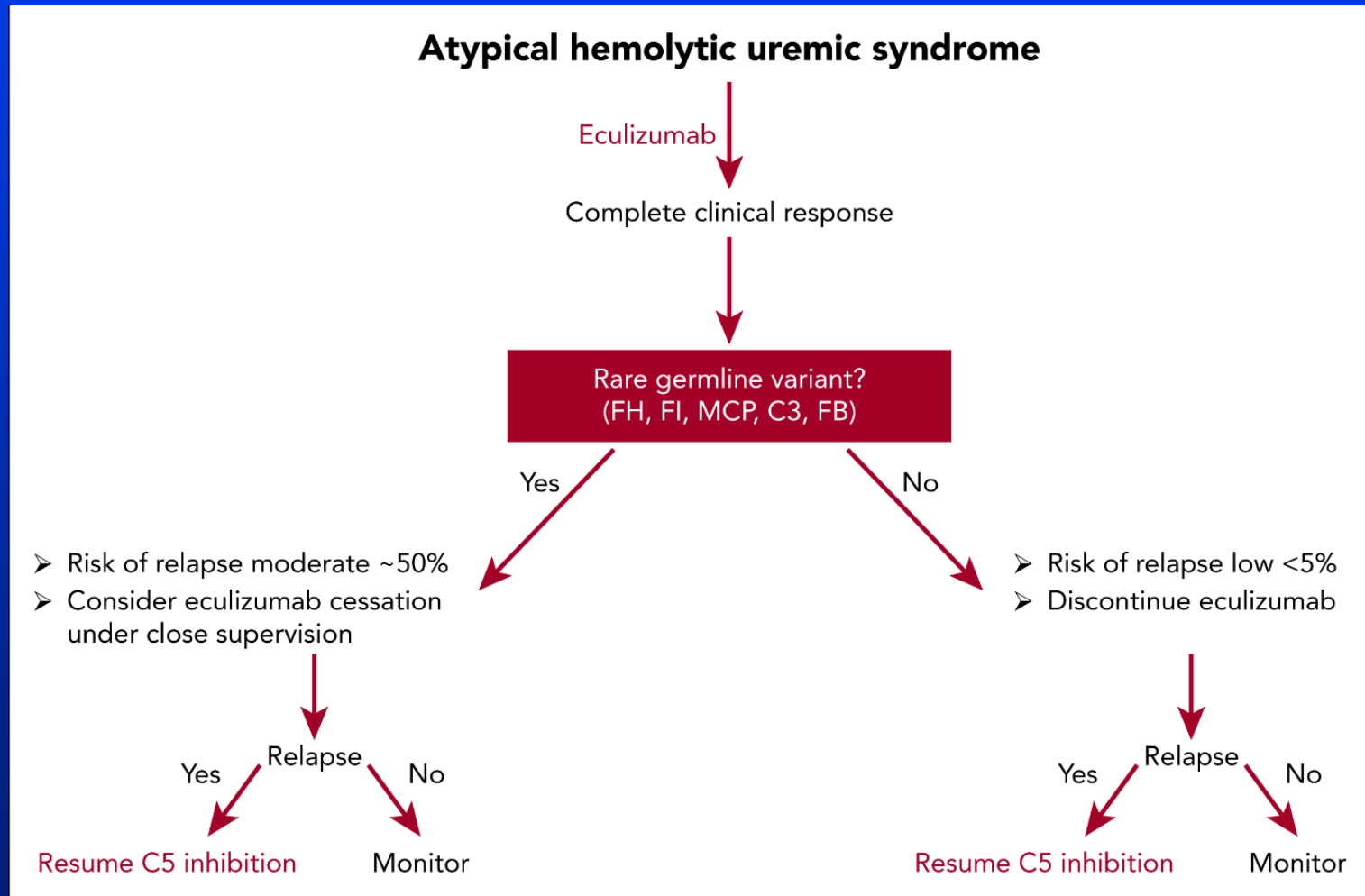


- *Deceased donor:*



Laboratory test samples must be drawn before plasma therapy and before eculizumab pre-transplant, 24 hours before transplant and post transplant to monitor eculizumab therapy

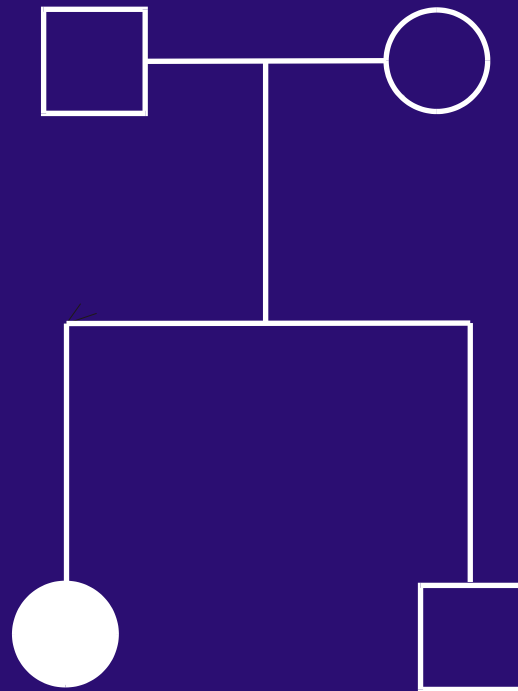
# Can we stop Eculizumab?



# Case 3

- 12 years old male patient who developed ESRD secondary to aHUS. He is now undergoing an evaluation for kidney transplantation at your center. Which of the following statement is correct:

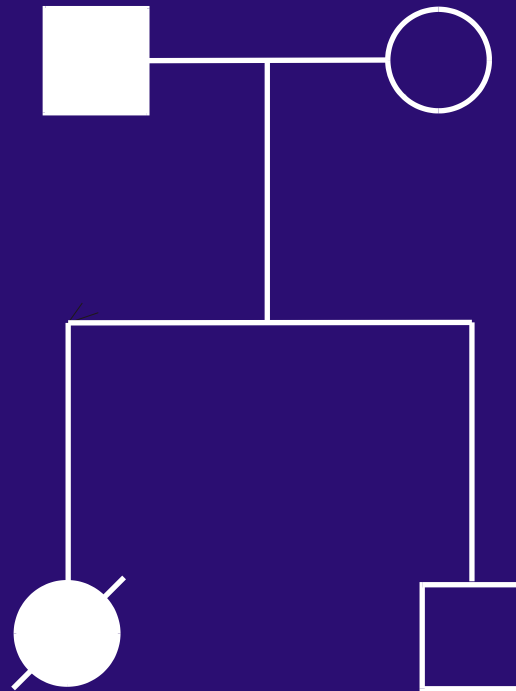
- A- Living unrelated kidney donor should be avoided
- b- Living related kidney transplant should be avoided
- c- He can be transplanted regardless of the source of donor as long as he receives eculizumab after kidney transplantation
- d- The risk of recurrent aHUS after kidney transplantation is very low



Sporadic HUS  
a. 2. LRD from  
father



Father 1y later  
develops HUS



Graft lost to  
recurrent HUS

# Mutations in Complement System Proteins in Atypical HUS

| Protein               | Synthesis Site | Function  | Effect of Mutation | Mutation (%) |
|-----------------------|----------------|-----------|--------------------|--------------|
| <b>CFH</b>            | Liver (plasma) | Cofactor  | Inactivating       | 30           |
| <b>MCP</b>            | Cells          | Cofactor  | Inactivating       | 12           |
| <b>CFI</b>            | Liver (plasma) | Protease  | Inactivating       | 5-10         |
| <b>C3</b>             | Liver (plasma) | Component | Activating         | 5            |
| <b>CFB</b>            | Liver (plasma) | Component | Activating         | rare         |
| <b>THBD</b>           | Cells          | Cofactor  | Inactivating       | 3-5          |
| <b>CFHR1<br/>/CFH</b> | Liver (plasma) | -         | Competitor         | 3-5          |

# Kidney transplant alone with Eculizumab or Combined liver Kidney Transplant

| Isolated kidney with chronic eculizumab                           | Combined liver – kidney transplant                                 |
|-------------------------------------------------------------------|--------------------------------------------------------------------|
| Lower short-term risk                                             | Higher short-term mortality                                        |
| Long-term outcomes unknown                                        | Long-term outcomes stable over 10-20 years                         |
| Long-term dependence on eculizumab to prevent recurrences of aHUS | aHUS recurrence unlikely                                           |
| Complications of immunosuppressive agents                         | ? Fewer complications of immunosuppressive agents                  |
| Chronic rejection                                                 | ? Reduced risk of chronic rejection                                |
| IV infusion every 2 weeks                                         | Better lifestyle with no infusions                                 |
| Limited worldwide availability                                    | More widely available<br>But scarce liver plus kidney availability |
| Extremely expensive                                               | Less expensive                                                     |

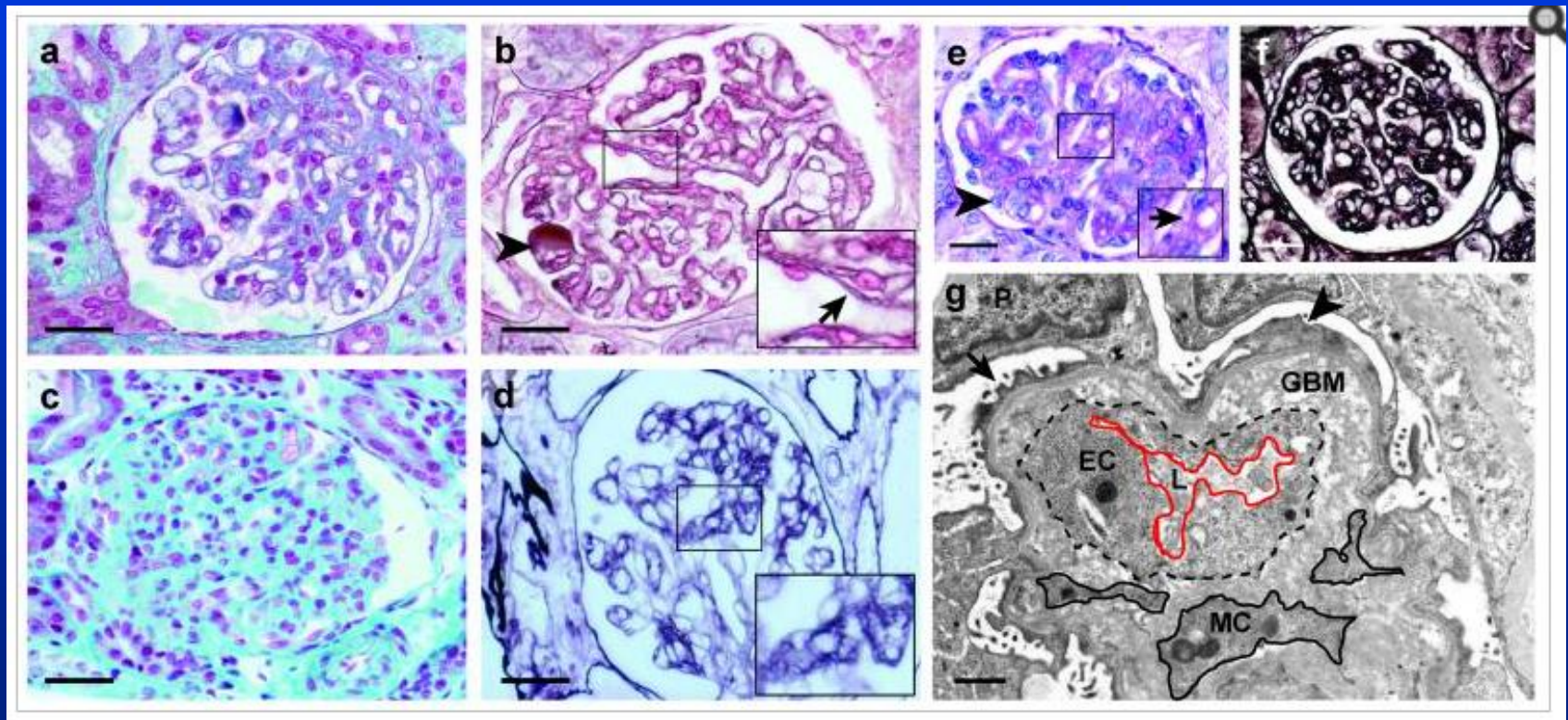
# Complement based therapeutics in the pipeline for renal disorders

| Drug Class                            | Name                                 | Current use                                        | Potential use                                                              |
|---------------------------------------|--------------------------------------|----------------------------------------------------|----------------------------------------------------------------------------|
| <b>C1 Esterase Inhibitor</b>          | Berinert, Conestat Alfa, Cinryze     | - Acute and chronic graft injury [Transplantation] |                                                                            |
| <b>MASP2 Inhibitor</b>                | OMS721                               | - TMA                                              | - aHUS, HSCT-TMA<br>- IgA Nephropathy                                      |
| <b>Anti-C5 antibody</b>               | Eculizumab                           | - PNH<br>- AHUS                                    | - Delayed AMR<br>- Transplant associated TMA<br>- Idiopathic Membranous GN |
| <b>C5aR antibody</b>                  | CCX168                               |                                                    | - ANCA vasculitis<br>- AHUS [with congenital complement abnormality]       |
| <b>C3 inhibiting peptide</b>          | AMY 101<br>Compstatin, Pegcetacoplan |                                                    | - PNH<br>- C3GP, aHUS                                                      |
| <b>C3 soluble complement receptor</b> | TP 10                                |                                                    | - DDD                                                                      |
| <b>C5 inhibitor</b>                   | Coversin                             |                                                    | - PNH                                                                      |
| <b>Factor D inhibitor</b>             | ACH-4471 (Danicopan)                 |                                                    | - PNH                                                                      |

# Case 4

- 10 months old male enfant presents with MAHA, thrombocytopenia, and AKI. He has no known family history of aHUS. The patient had persistent hypertension, hematuria and also nephrotic range proteinuria. All genetic complement testing for aHUS were negative and he had no circulating anti-CFH antibodies. C3 and C4 are normal.
- a Kidney biopsy was performed.

# Case 4



# Case 4: Question 1

- What is the most likely diagnosis:
  - a- complement mediated aHUS
  - b- TTP
  - c- HUS secondary to diacylglycerol kinase  $\epsilon$  (DGKE) mutation
  - d- Lupus nephritis

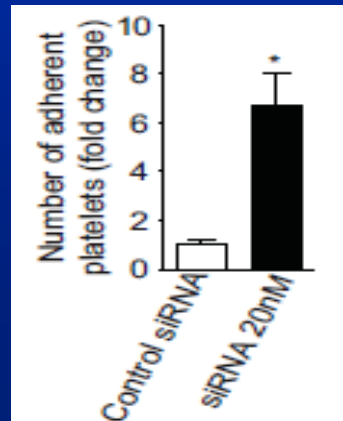
# RECESSIVE MUTATIONS IN *DGKE* CAUSE aHUS SYNDROME

- Homozygous or compound heterozygous mutations in *DGKE* (encoding diacylglycerol kinase  $\epsilon$ ) were found in 27% of aHUS cases with onset in the first year of life
- Peculiar clinical phenotype: recurrent disease in childhood, development of proteinuria sometimes with the nephrotic syndrome
- *DGKE* is not an integral protein of complement and patients did not show complement consumption and one relapsed while on eculizumab

Lemaire M et al, Nature Genet 2013

## ***Consequences of *DGKE* deficiency***

- *DGKE* knock down in endothelial cells up-regulated ICAM-1 and tissue factor expression and resulted in platelet adhesion without inducing complement deposition.



Bruneau et al, Blood, 2015



# TTP

- Described in 1924 by Moschowitz
- Associated with CNS involvement
- In the 1980's TTP was a disease in search of an etiology
- Large von Willebrand multimers were initially identified.

# TTP Pentad

- Severe Thrombocytopenia (10-30k)
- MAHA
- Neurological involvement (headache, confusion, TIA, seizure, etc..)
- Renal failure
- Fever

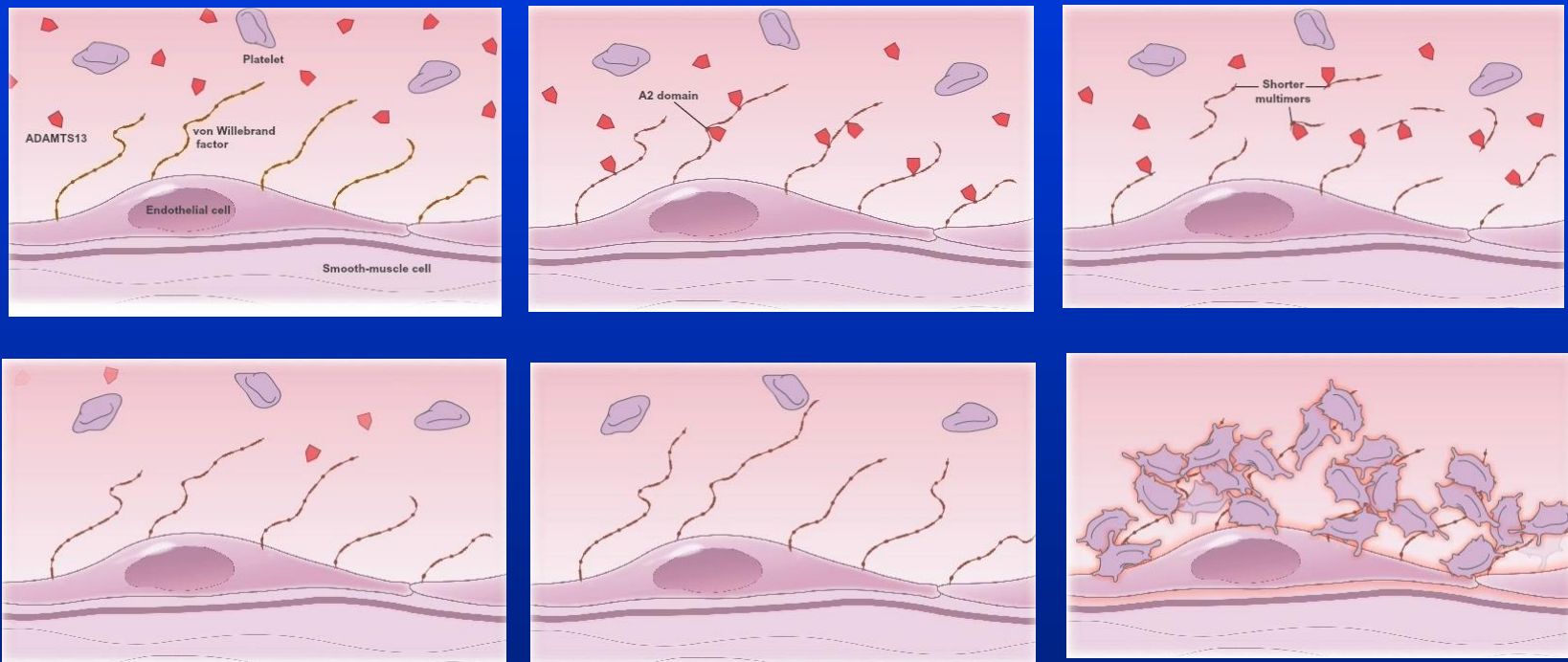
# Von Willebrand Factor (vWF)

- Large glycoprotein
- Dimers form in the ER
- Multimers form in the Golgi
- vWF is released in the plasma as large multimers with high MW, the biologically active form
- A disintegrin and metalloproteinase with a thrombospondin type 1 motif, member 13 (ADAMTS13) usually cleaves vWF multimers

# Hereditary Thrombotic Thrombocytopenic Purpura (TTP)

- Homozygous or compound heterozygous mutations of ADAMTS13 (Upshaw-Schulman syndrome) causing ADAMTS13 deficiency in the absence of auto-antibody
- Usually presents in children can present in adults (e.g. precipitated by pregnancy)
- Patients with heterozygous mutations are normal

# Hereditary TTP: Pathophysiology

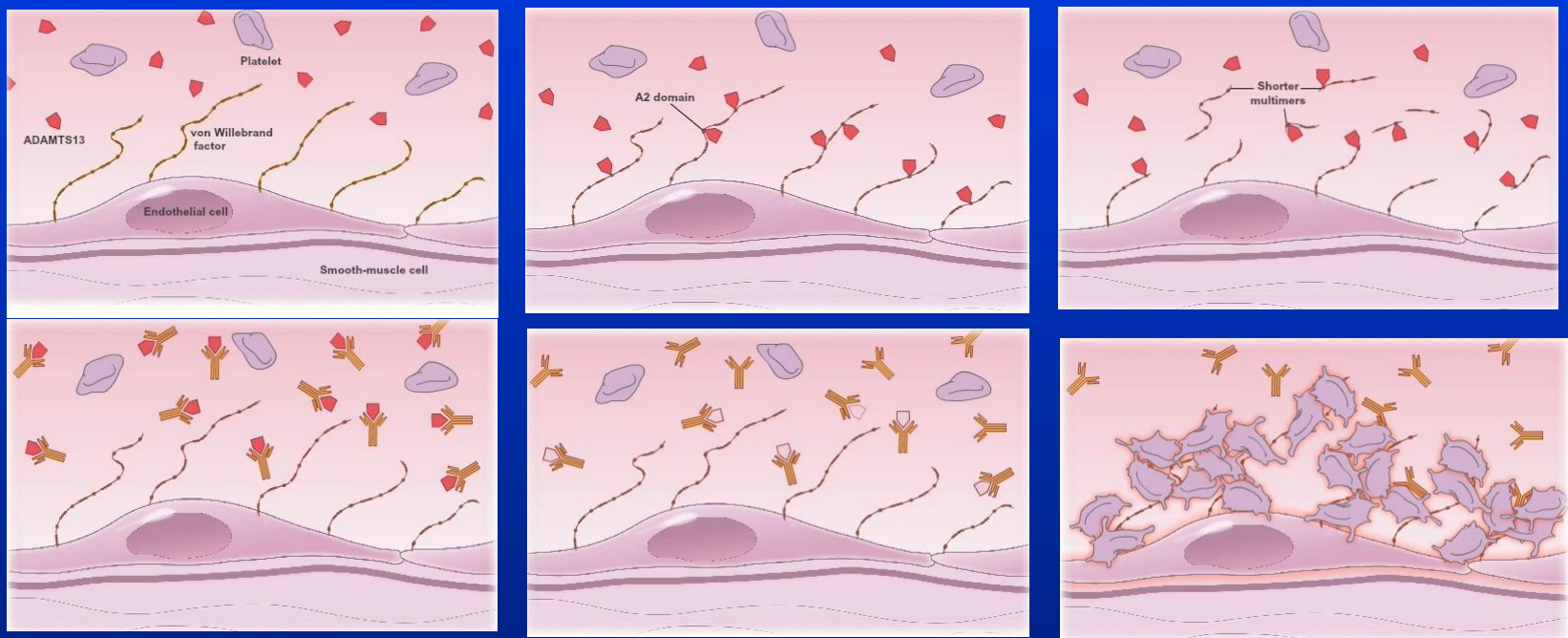


Treatment: Plasma infusion or Plasma derived factor VIII concentrate (if plasma allergy) which contains ADAMTS13

# Acquired Thrombotic Thrombocytopenic Purpura (TTP)

- Variable presentation (weakness, GI symptoms, purpura, focal neurological deficit)
- Normal or transient mild renal failure!
- 1/3 have no neurological findings
- ADAMTS13 activity  $<10\%$  caused by an autoantibody blocking ADAMTS13 or accelerating its clearance

# Acquired TTP: Pathophysiology



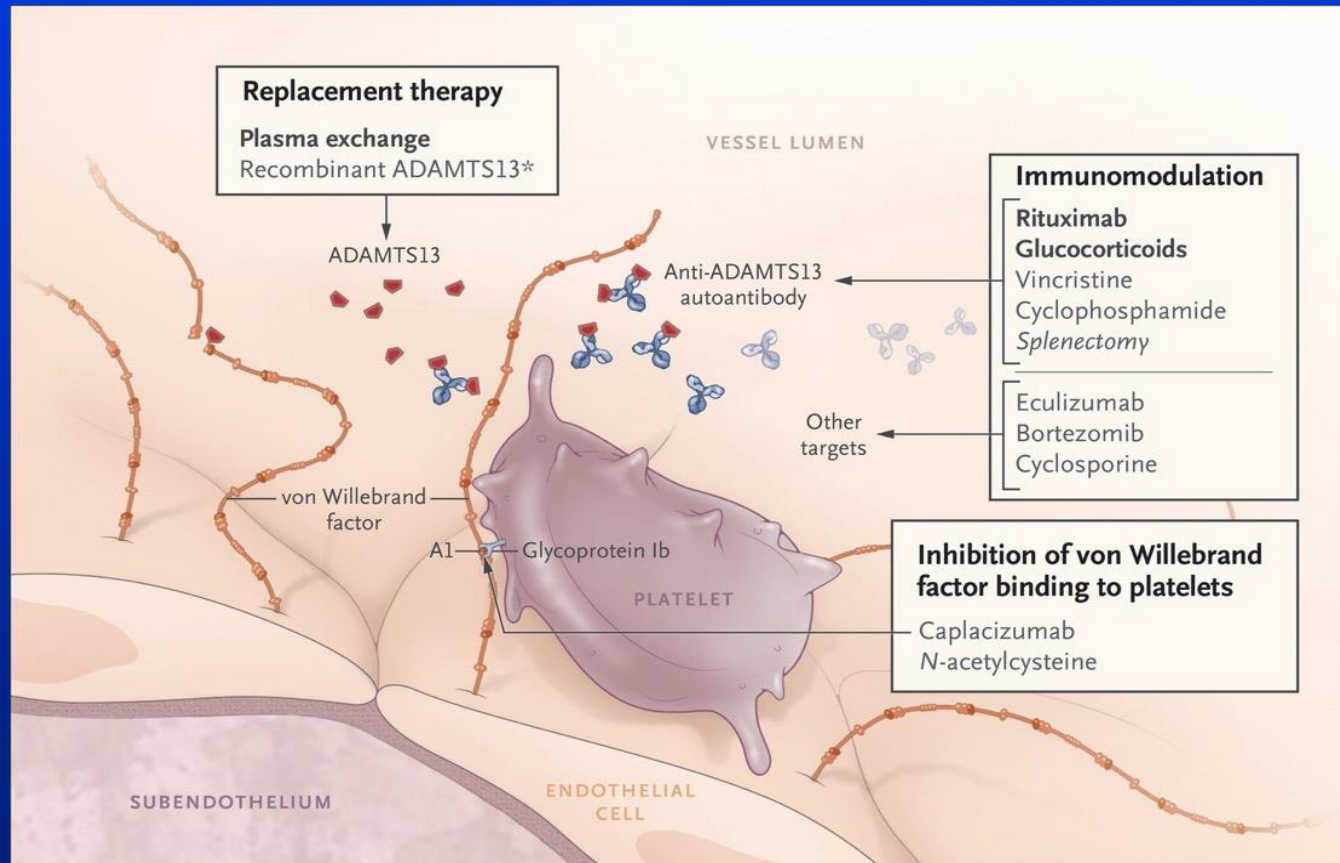
Treatment: without PEX mortality close to 90%, PEX (standard of trt) 60-90% survival. Steroids, rituximab, bortezomib, other IS can be used. Dialysis rarely required

# Therapy of Acquired TTP

- Adults with TTP- plasma exchange (removes large multimers of vWF and autoantibodies to ADAMTS13 and allows infusion of metalloprotease), 90 % survival with PEX for TTP (mortality 90% in the past), PEX performed daily and continued until platelets are normal ( $>150000$ )
- Steroids are frequently used
- Rituximab (Anti-CD 20) often used for severe cases (severe neurological symptoms)
- Aspirin or anticoagulants may increase bleeding risk , platelets transfusion can be used in severe thrombocytopenia



# Treatment for TTP



# Case 5

35 y/o female with no PMH presents with 5d headache, N/V and non-bloody diarrhea

- No recent travel
- Husband and kids are healthy, and ate same foods
- Has never had this constellation of symptoms before

# Case 5, continued

Medications: none

PMH: 3 full-term vaginal deliveries, no miscarriages

SH: occasional EtOH, no tobacco, no illicit drugs

FH: father with HTN, CAD; siblings and children healthy

# Physical Exam

VS: T 100.0 HR 90 BP 145/90 RR 16 O<sub>2</sub> sat 98%  
(RA)

60 kg, no apparent distress, mild pallor

Lungs: clear

Heart: RRR, no MGR, normal pulses

Abd: diffusely tender with deep palpation, no rebound

Extr: no edema, no rashes

Neuro: no focal deficit

# Laboratory tests

Creatinine 2.0, BUN 36

WBC 11.0 / Hgb 9.0 / Plts 30

LDH 1800, haptoglobin <8

Normal amylase, lipase, LFTs

UA: SG 1.015, pH 5.5, 1+ protein, 2+ blood, trace  
LE

> 20 RBC/hpf, > 10 WBC/hpf, several granular casts

# Case 5, continued

- Blood and urine cultures negative
- Stool cultures negative for bacterial dysentery, STEC negative
- INR 1.1, PTT 28, D-dimer normal
- Samples sent for ADAMTS13 activity and anti-phospholipid antibodies

# Case 5: Question 1

- What is your first therapeutic approach?
  - a- Plasma exchange with steroids
  - b- plasma infusion
  - c- steroids and rituximab
  - d- Eculizumab

# Case 5, continued

- Plasma exchange initiated with FFP
- Rapid improvement in headache
- Platelet count 180 by day 4, LDH down to 280
- On day 5, prior to plasma exchange, Plts dropped to 140
- LDH increased to 480
- Return of headache and < 10 min episode of left-sided weakness
- ADAMTS13 activity returned < 5% of normal
- ADAMTS13 inhibitor present, consistent with acquired TTP



## Case 5, Question 2

- What is the best therapeutic approach for this refractory TTP
  - a- Increase the dose of PEX to twice daily
  - b- Continue current dose of PE, high dose steroids and rituximab given at end of PEX
  - c- Eculizumab
  - d- reconsider your diagnosis.

# Case 6

- The best treatment of the first relapse of acquired TTP (usually occurring >30 day after initial episode recovery), after an initial good response to PEX and steroids during the first presentation is:
  - a- PEX and steroids
  - b- PEX/steroids and Rituximab
  - c- splenectomy
  - d- cyclophosphamide

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# Thank you!

- *Questions*