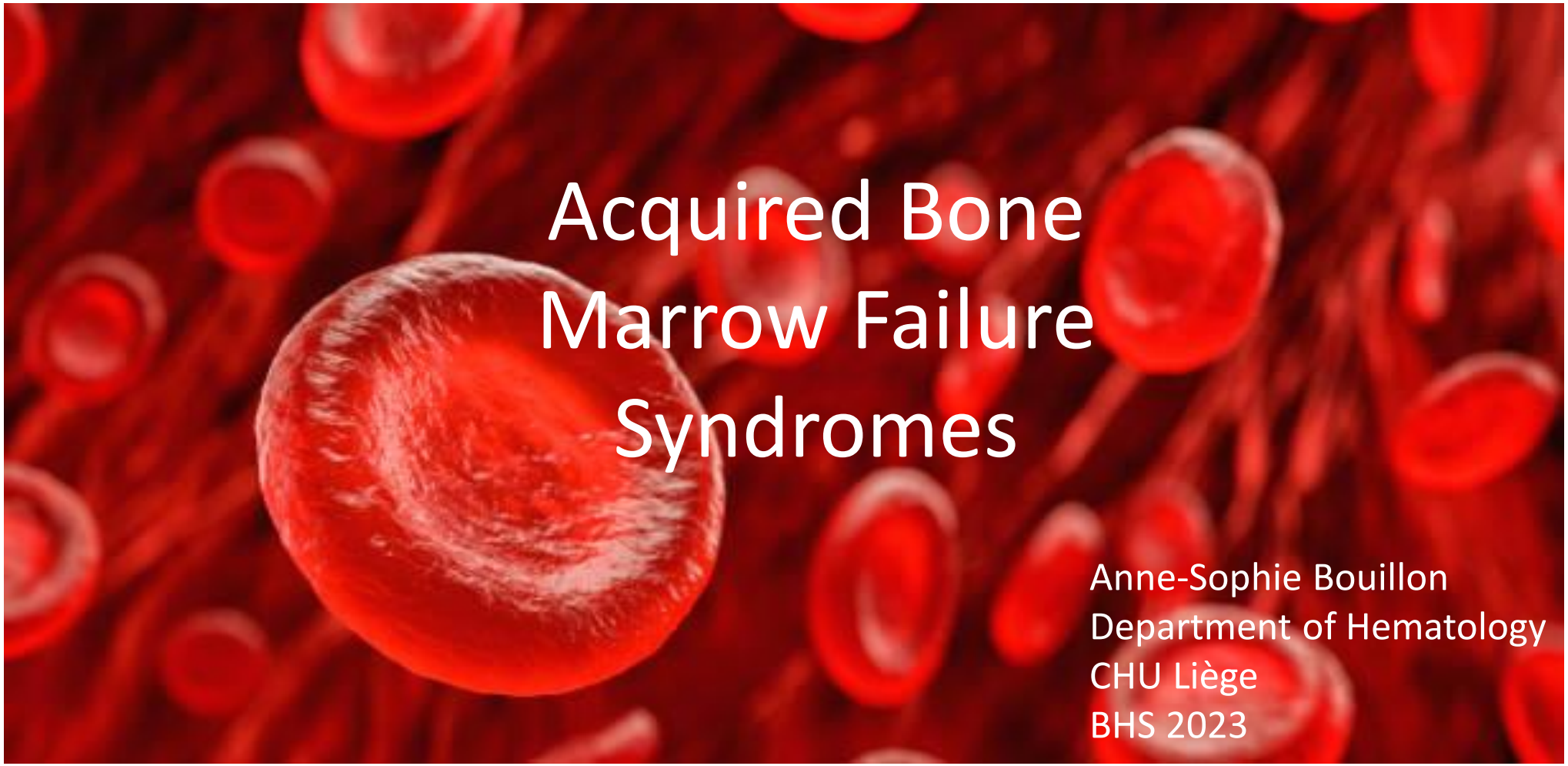


Acquired Bone Marrow Failure Syndromes

Anne-Sophie Bouillon
Department of Hematology
CHU Liège
BHS 2023

Idiopathic aplastic anemia

Paroxysmal nocturnal hemoglobinuria

Case report (I)

10/2020: 40 years old patient came to emergency department with epistaxis

- Blood count: Hb 9,9g/dl, platelets 10000/mm³, WBC 3760/mm³ with 1620 neutrophils.
- Rtc 73000/mm³
- Serum chemistries without abnormality

- Macrocytosis
- No blasts
- No vitamine deficiency
- No schistocytes
- HIV, hepatitis, CMV, EBV, parvovirus negative

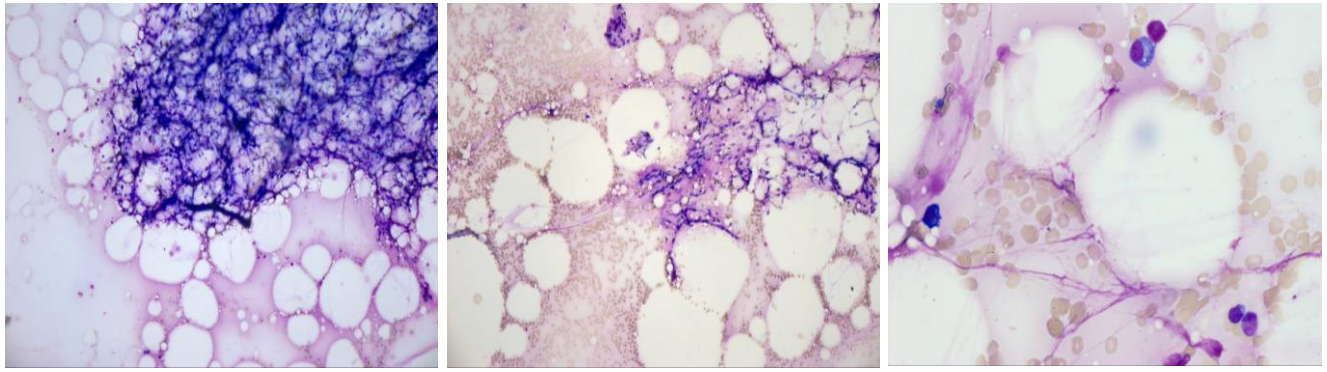
→ Suspicion of ITP: Administration of Immunoglobulines and steroids

NO RESPONSE

Case report (II)

Bone marrow examination:

- Cytology: Aplasia of megacaryocytes and erythropoiesis, hypoplasia in granulocytes
- Biopsy: aplasia (hypocellularity <10%)



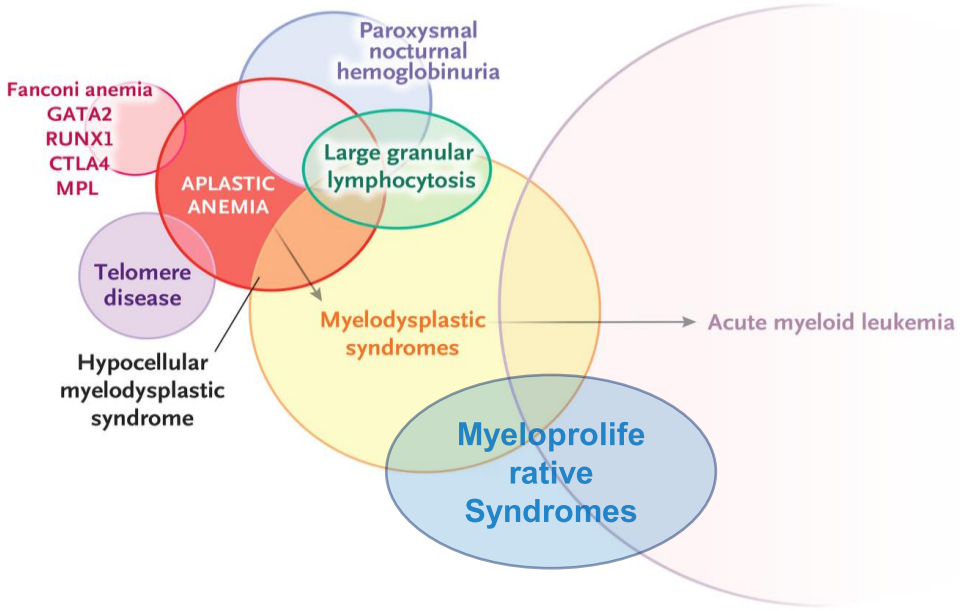
- → **Aplastic anemia**

Aplastic Anemia

Rare disorder

Loss of hematopoietic stem cell

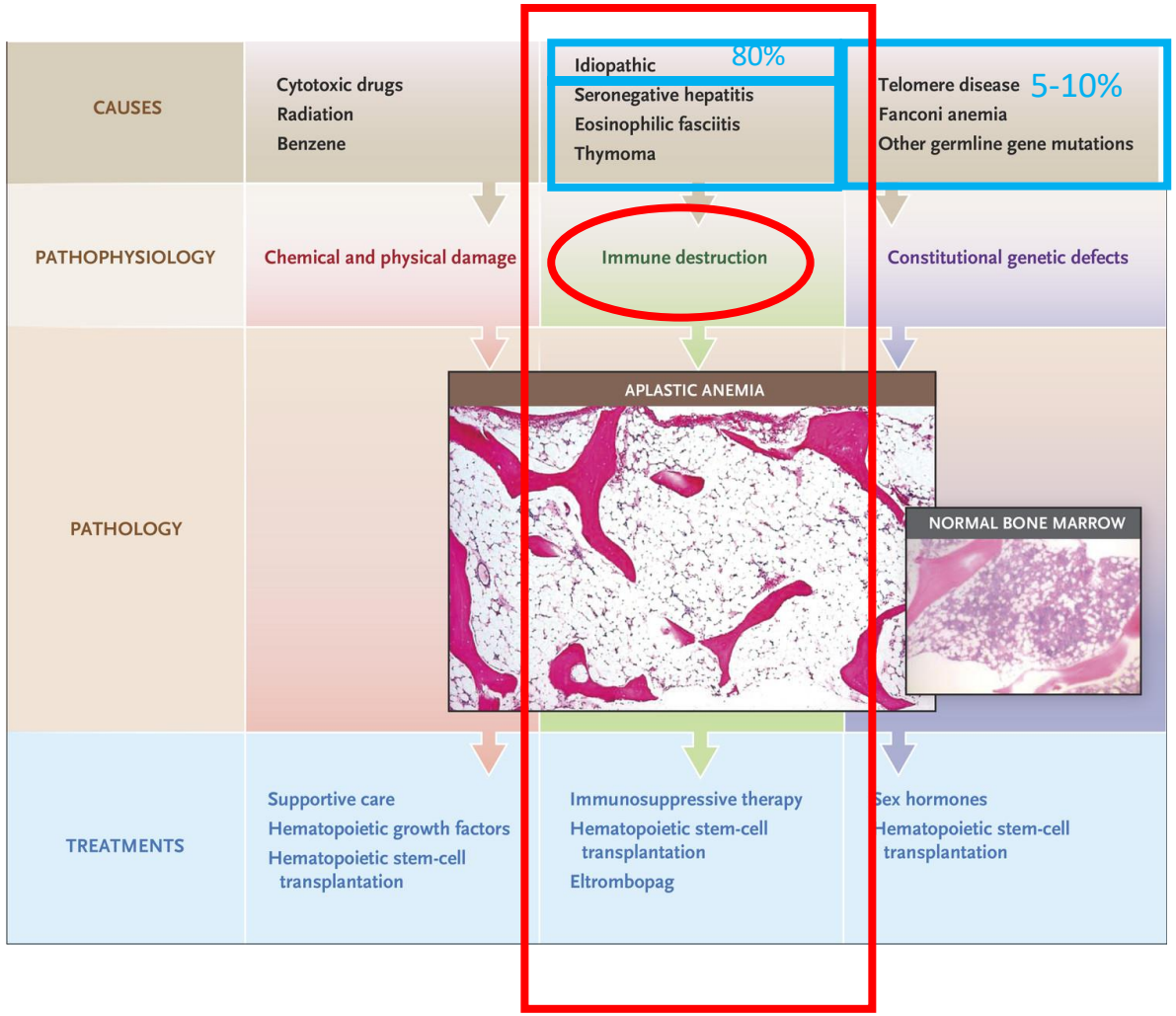
→ Cytopenia in association with bone marrow aplasia/hypoplasia



NS Young. N Engl J Med 2018;379:1643-1656.

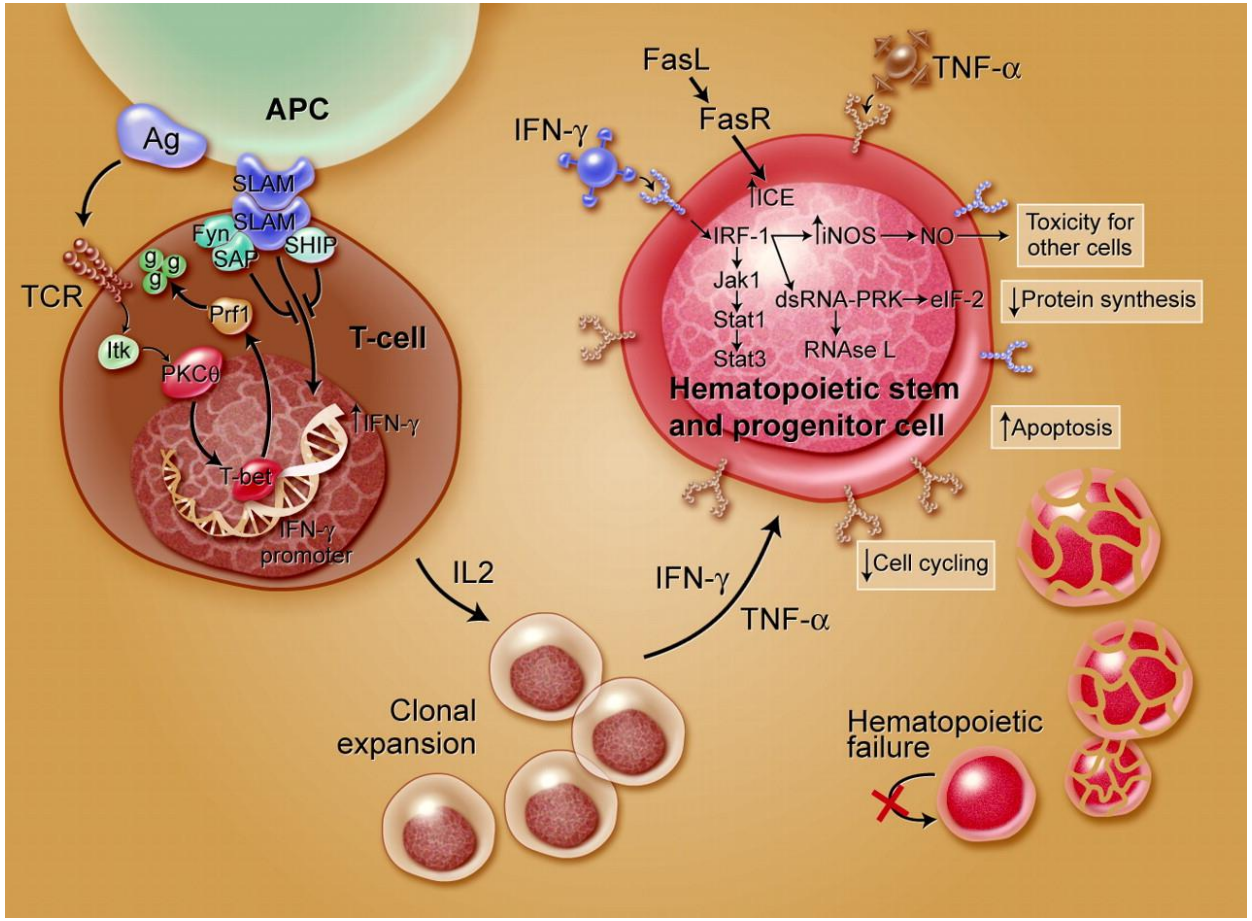
Etiology

- Idiopathic (> 80%)
- Drugs (<20%), idiosyncratic reactions
- Post infection (<5%)
- Late onset inherited BMF - syndromes



NS Young. N Engl J Med 2018;379:1643-1656.

Idiopathic Aplastic Anemia Pathophysiology



- Immune mediated disease
- oligoclonal T cell expansion
- Inhibitory cytokines TNF-α and interferon-γ
- Increased apoptosis and suppression of normal HSC growth
- Destruction of HSC
- PNH Clone by immune escape mechanism

Young NS et al. Blood 2006

Evaluation

- History: Begin?, symptoms ?, infections? drugs?, autoimmun diseases?, familiar history?
- Clinical examination, signs of inherited bone marrow failure?
- Laboratory studies:
  - Blood count:
    - Cytopenia
    - Macrocytosis
    - Reticulocytopenia
  - Hemolysis?
  - Quick, PTT, Fibrinogen
  - CRP
  - Serum chemistries, including electrolytes, liver function tests (including lactate dehydrogenase [LDH]) and renal function tests
  - Proteines and electrophoresis, immunoglobulines
  - Ferritin
  - Vitamin B12 and folate
  - ANA, ANCA, RF, …
  - Blood group and Coombs Test
- Infectious diseases: EBV, CMV, Hepatitis A, Hepatitis B, Hepatitis C, Hepatitis E, HIV, Parvovirus B19
- PNH ?

Evaluation- Bone marrow examination

Bone marrow examination

- Cytology
- Biopsy required!
- Cytogenetics/ FISH
- NGS

Hypocellular bone marrow, mostly all lines

Fat cells

Residual hematopoeitic cells are mostly morphologically normal → dyserythropoiesis possible

No infiltration with malignant cells or fibrosis

Infiltration by lymphocytes

Diagnostic criteria idiopathic aplastic anemia

- Cytopenia
- Hypocellular bone marrow
- Exclusion of inherited bone ma
- Exclusion of other etiologies →

- Clonal markers?
- PNH?

! Are not considered as Aplastic Anemia…

- Cytotoxic medications and/or ionizing radiation with predictable and reversible damage to HSCs and acute hematopoietic failure
- Hypoplastic MDS
- BMF due to Anorexia nervosa
- Pure red cell aplasia
- Isolated PNH
- Autoimmune diseases
- BM infiltration by other cancers

Classification

Classification	Criteria
Severe	Bone marrow (BM) cellularity <25% (or <50% if <30% of BM are haematopoietic cells) And two or more of the following: Peripheral blood neutrophil count $<0.5 \times 10^9$/L Peripheral blood platelet count $<20 \times 10^9$/L Peripheral blood reticulocyte count $<20 \times 10^9$/L
Very severe	As above, but peripheral blood neutrophil count must be $<0.2 \times 10^9$/L
Non-severe	Hypocellular BM with peripheral blood values not meeting criteria for severe aplastic anaemia

Furlong et al. J Paed Child Health 2020

Case report (III)

10/2020:

- Hb 9,9g/dl, platelets 10000/mm³, WBC 3760/mm³ with 1620 neutrophils.
- Rtc 73000/mm³

Exclusion of infections, cytotoxics, drugs,…

No vitamine deficiency

No sign of autoimmune or rheumatic diseases

No Fanconi anemia, no telomeropathy

PNH Clone 0,3%, without relevant hemolysis

Moderate aplastic anemia

Severe aplastic anemia

Evolution 12/2020:

- Hb 6,9 g/dl, Rtc 15000/mm³, plt 17000/mm³, neutrophiles 400/mm³

Treatment

Indication of treatment:

✓Severe and very severe aplastic anemia

✓Non severe aplastic anemia with risk due to cytopenia like

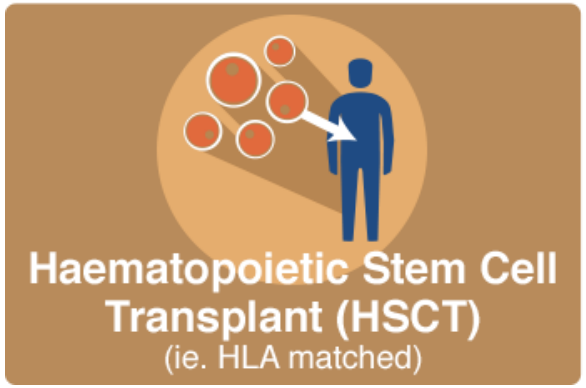
- Bleeding
- Infections
- Transfusions

✓ Transition nsAA to sAA

sAA without treatment → poor prognosis!

Treatment

- Transfusions
- Prevention of infections
- Prevention of bleeding
- Iron overloading



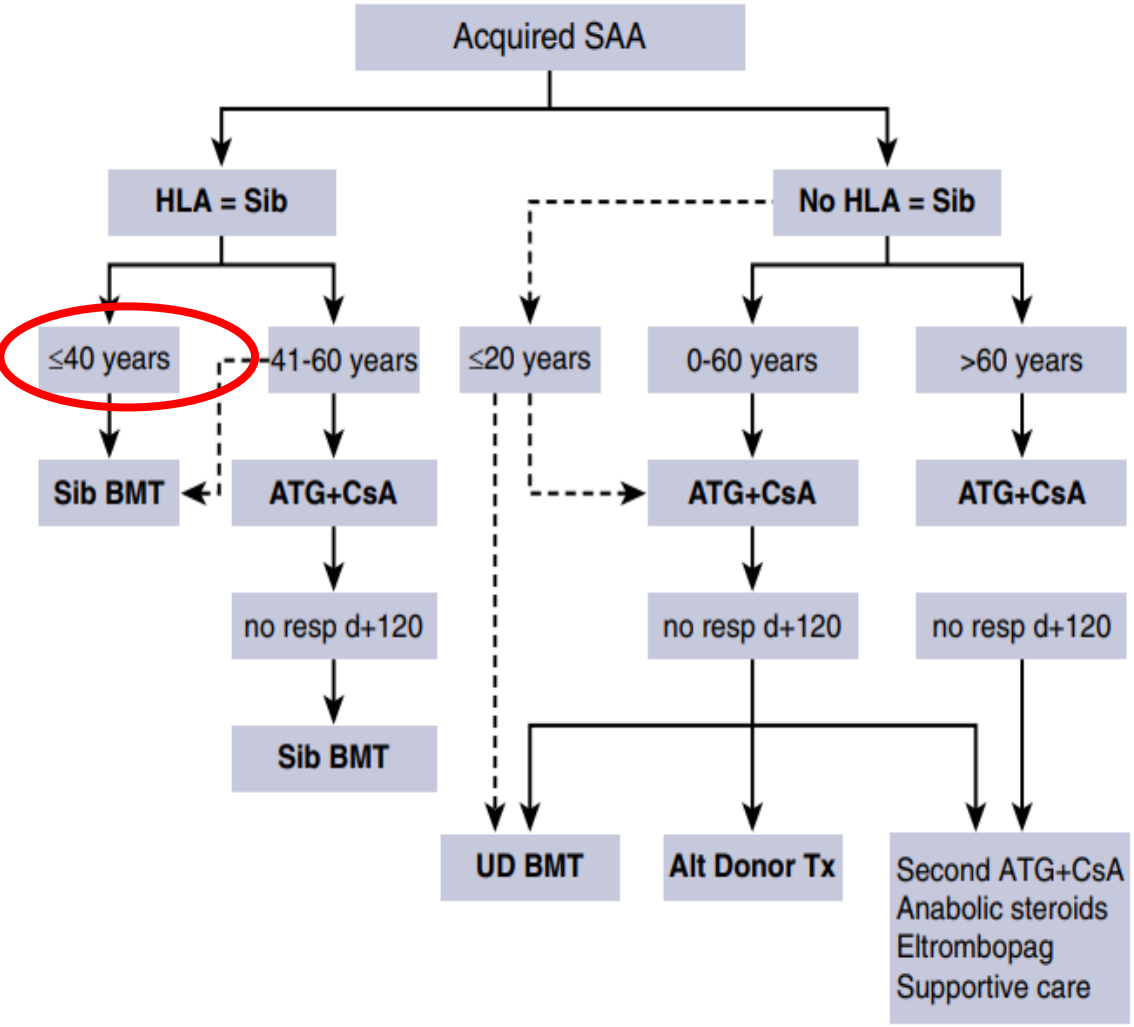
Hematopoietic stem cell transplantation

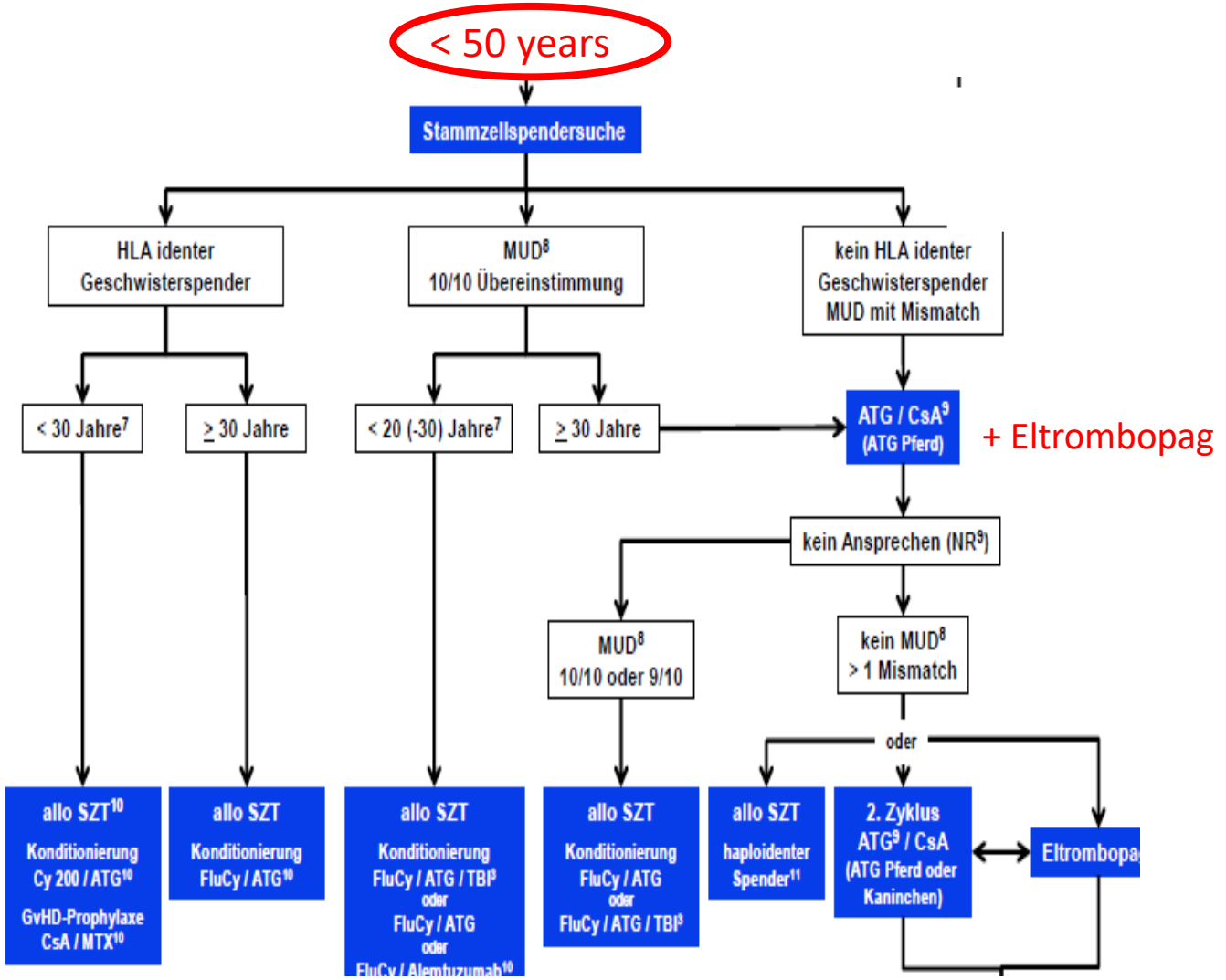
First line treatment for patients < 40-50 years
sAA and vsAA

Influencing factors

- Sibling donor
- Bone marrow transplant vs. PBSCT

Treatment of newly diagnosed or relapsed AA should be discussed with specialized center

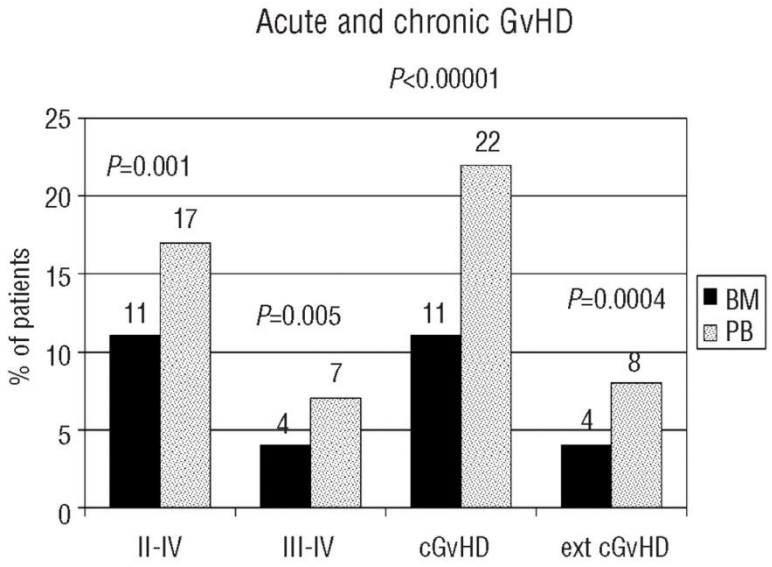


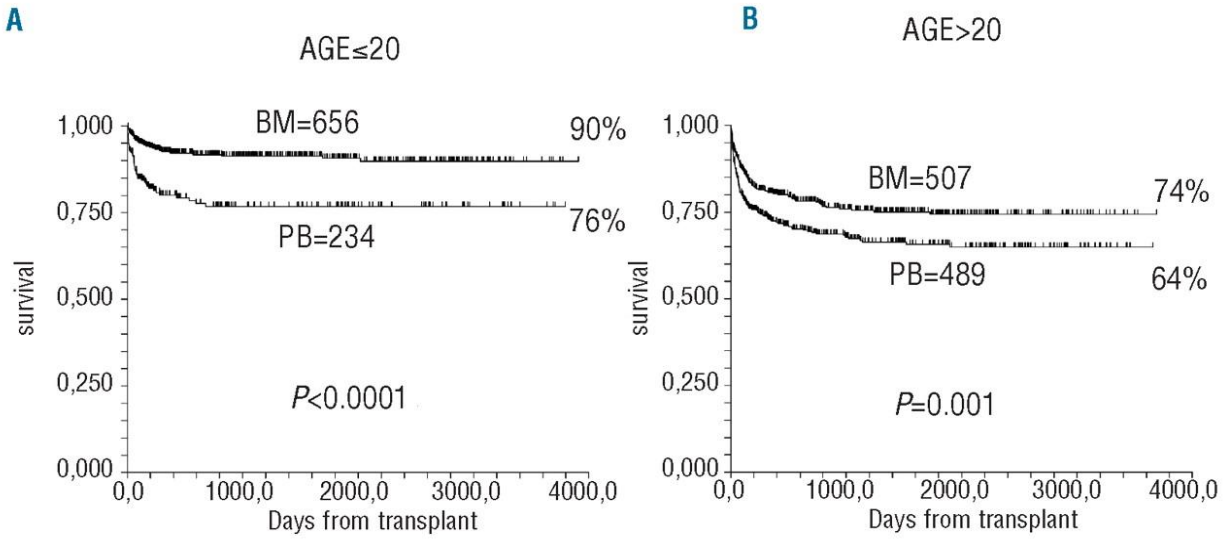


Andrea Bacigalupo, Blood First Edition paper, 17 January 2017; DOI 10.1182/blood-2016-08- 693481

https://www.onkopedia.com/de/onkopedia/guidelines/aplastische-anaemie

Bone marrow vs peripheral stem cells





Bone marrow transplantation ist the first choice !

Bacigalupo A et al. *Haematologica* 2012;97:1142-8.

Immunosuppressive Therapy
Antithymocyte Globulin (ATG) and Cyclosporine A

Primary therapy

- vSAA oder SAA > 40- 50 years or without HLA sibling donor
- nSAA with severe cytopenia

No age limitation

Standard is **horse ATG/CSA (not rabbit ATG in first line)**

Eltrombopag

Good results in second line therapy

First line for sAA/vsAA

The NEW ENGLAND JOURNAL of MEDICINE

ORIGINAL ARTICLE

Horse versus Rabbit Antithymocyte Globulin in Acquired Aplastic Anemia

Phillip Scheinberg, M.D., Olga Nunez,
Priscila Scheinberg, M.S., Angélique B
and Neal S. Y

Table 2. Hematologic Response at 3 and 6 Months to Horse ATG and Rabbit ATG.

Response	Horse ATG (N=60)	95% CI	Rabbit ATG (N=60)	95% CI	P Value
	no. (%)		*no. (%)*		
At 3 mo	37 (62)	49–74	20 (33)	21–46	0.002
At 6 mo	41 (68)	56–80	22 (37)	24–49	<0.001

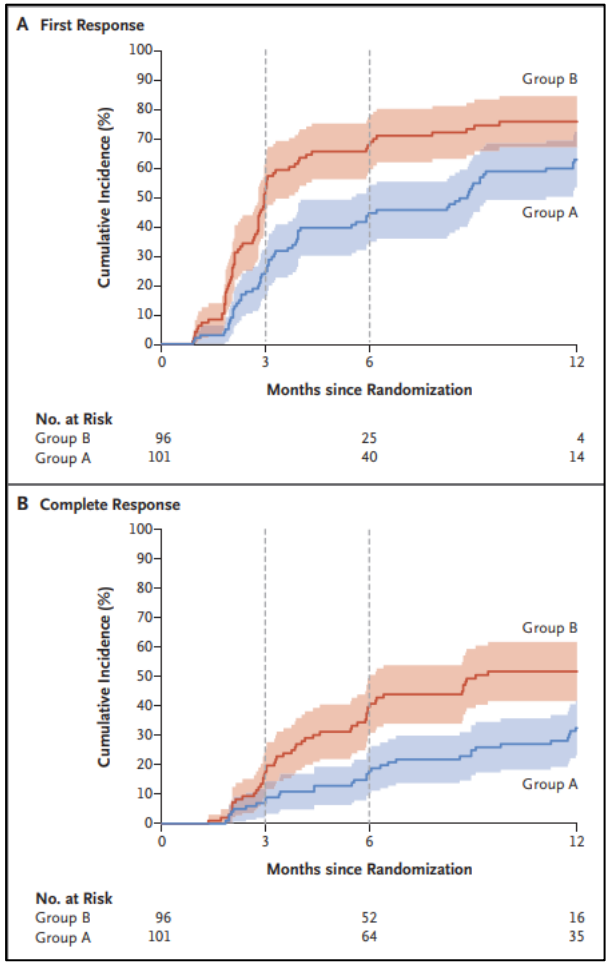
Future…

The NEW ENGLAND JOURNAL of MEDICINE

ESTABLISHED IN 1812 JANUARY 6, 2022 VOL. 386 NO. 1

Eltrombopag Added to Immunosuppression in Severe Aplastic Anemia

R. Peffault de Latour, A. Kulasekararaj, S. Iacobelli, S.R. Terwel, R. Cook, M. Griffin, C.J.M. Halkes, C. Recher, F. Barraco, E. Forcade, J.-C. Vallejo, B. Drexler, J.-B. Mear, A.E. Smith, E. Angelucci, R.A.P. Raymakers, M.R. de Groot, E. Daguindau, E. Nur, W. Barcellini, N.H. Russell, L. Terriou, A.-P. Iori, U. La Rocca, A. Sureda, I. Sánchez-Ortega, B. Xicoy, I. Jarque, J. Cavenagh, F. Sicre de Fontbrune, S. Marotta, T. Munir, J.M.L. Tjon, S. Tavitian, A. Praire, L. Clement, F. Rabian, L. Marano, A. Hill, E. Palmisani, P. Muus, F. Cacace, C. Frieri, M.-T. van Lint, J.R. Passweg, J.C.W. Marsh, G. Socié, G.J. Mufti, C. Dufour, and A.M. Risitano, for the Severe Aplastic Anemia Working Party of the European Society for Blood and Marrow Transplantation*



Eltrombopag 150 mg daily !

Non severe aplastic anemia

Treatment only in case of relevant cytopenia

First choice: Immunosuppressive treatment by hATG and Cyclosporin A

EMAA trial: CSA mono vs CSA+Eltrombopag in first line

Follow up

- Response evaluation 4-6 month after IST
- In most cases not a complete remission
- Bone marrow examination 1x/year to exclude clonal evolution
- PNH

- 30 % of relapse after IST

Take home message

- Idiopathic aplastic anemia is a rare disorder characterized by cytopenia and hypoplasia/aplasia of bone marrow
- Exclusion of other bone marrow failure etiologies and especially inherited BMF
- Exclude PNH/AA overlap
- Treatment only if sAA/vsAA or relevant cytopenia
- Considering HSCT at first line treatment < 40-50 years, sibling donor
- IST for patients > 50 years, nsAA, non eligible for HSCT
- ATG/CSA/ + Eltrombopag in first line
- Treatment in specialized centers , trials !


Paroxysmal
Nocturnal
Hemoglobinuria

Case report (I)

27 years old, female patient with fatigue

- Hb 10,4 g/dl, platelets 80000/mm³, neutrophils 1630/mm³
- LDH 450 UI/L, haptoglobin < 0,01 g/L ; reticulocytes 280000/mm³

Free hemoglobin present

Coombs negative

No schistocytes

No infectious diseases

Paroxysmal nocturnal hemoglobinuria?

Definition and pathophysiology

Acquired disorder in hematopoietic stem cells

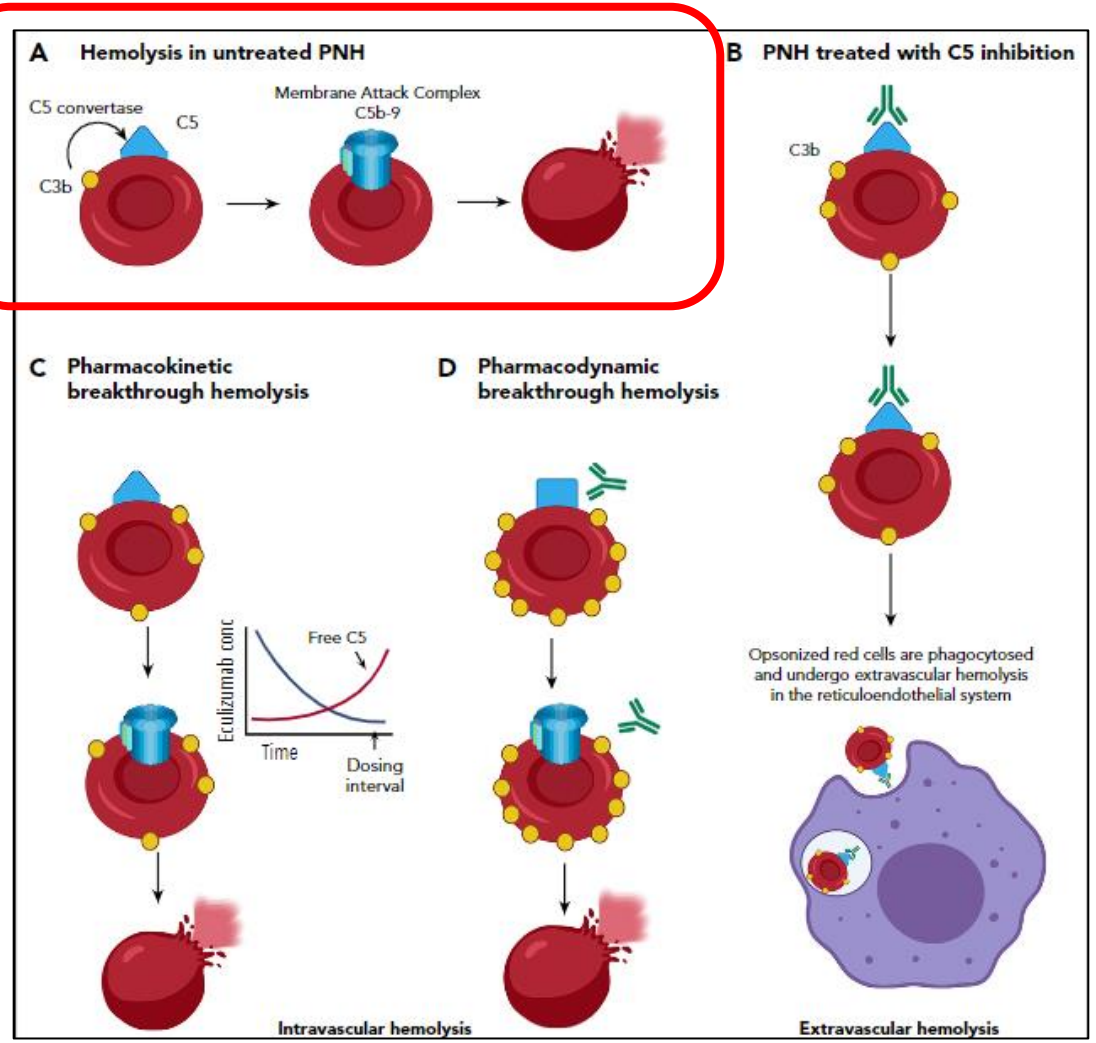
Mutation in PIG-A gen → reduced or absent GPI-anchored proteins on cell membrane

↓

Loss of the GPI-linked complement inhibitors, CD55 and CD59, on red blood cells

↓

Intravascular complement driven hemolysis and thromboembolic risk



Brodsky, blood® 11 MARCH 2021 | VOLUME 137, NUMBER 10

Symptoms

Intravascular hemolysis

- Nocturnal hemoglobinuria +- 25%
- Anemia related symptoms
- Vasospasm due to NO depletion

Thrombosis, typical and atypical localisations

- Venous> arterial
- Increased C5 levels→proinflammatory and prothrombotic cytokines
- NO depletion → dysfonction of endothel cells and platelets
- Inhibition ADAMTS13
- Pro coagulating agents
- → thrombotic risk correlates with clone size

Cytopenia/Bone marrow dysfunction

- BM dysfunction frequent
- Overlap syndrom with AA/less common MDS

Abdominal pain/dysphagia

Smooth muscle dystonia

Erectile dysfunction

Pulmonary hypertension

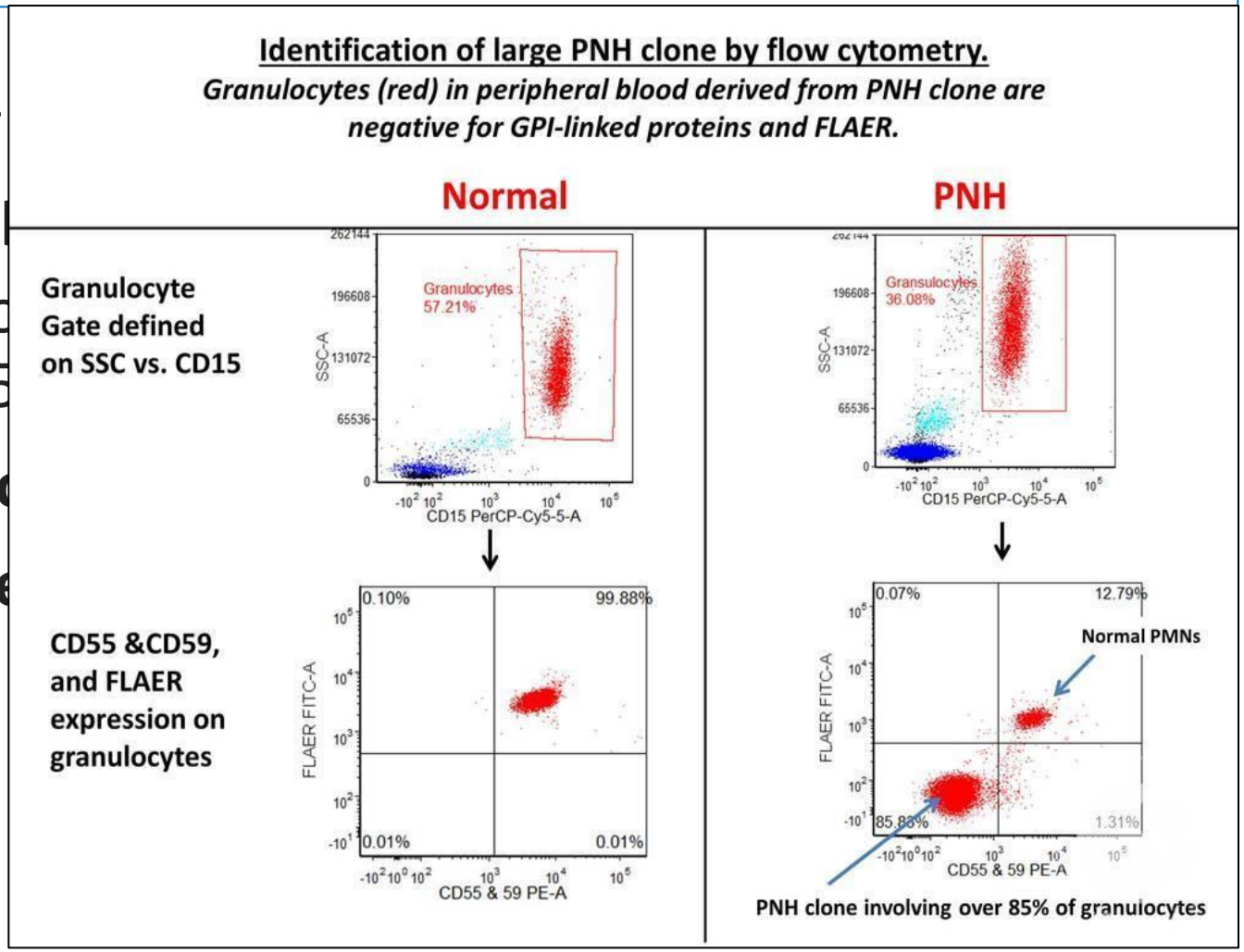
Renal insufficiency

Evaluation

- History:
  - Symptomes due to anemia, erectile dysfunction, abdominal pain,…
- Clinical examination
- Laboratory
  - Complete blood count, blood smear, reticulocytes, schistocytes
  - Coombs
  - Haptoglobin, LDH, Bilirubin, free hemoglobin
  - Iron
  - Urine for hemoglobin and hemosiderin

Flow cytometry/FLAER

- Detect reduction or loss of
  define the size of the PNH
- fluorescently labeled monc
  anchored proteins (eg, CD5
- **two independent flow cyt**
- **two cell lines need to be te**
  - **Red blood cells**
  - **Granulocytes**
  - **Monocytes**



ASH, image bank

In case of cytopenia → **bone marrow examination** to exclude AA /MDS related

- Clonal expansion of PNH clone favorised by autoimmune setting
- Autoimmune mediated bone marrow failure contributes to cytopenia
- About 40 % of PNH patients develop AA
- 70% of AA patients with PNH clone

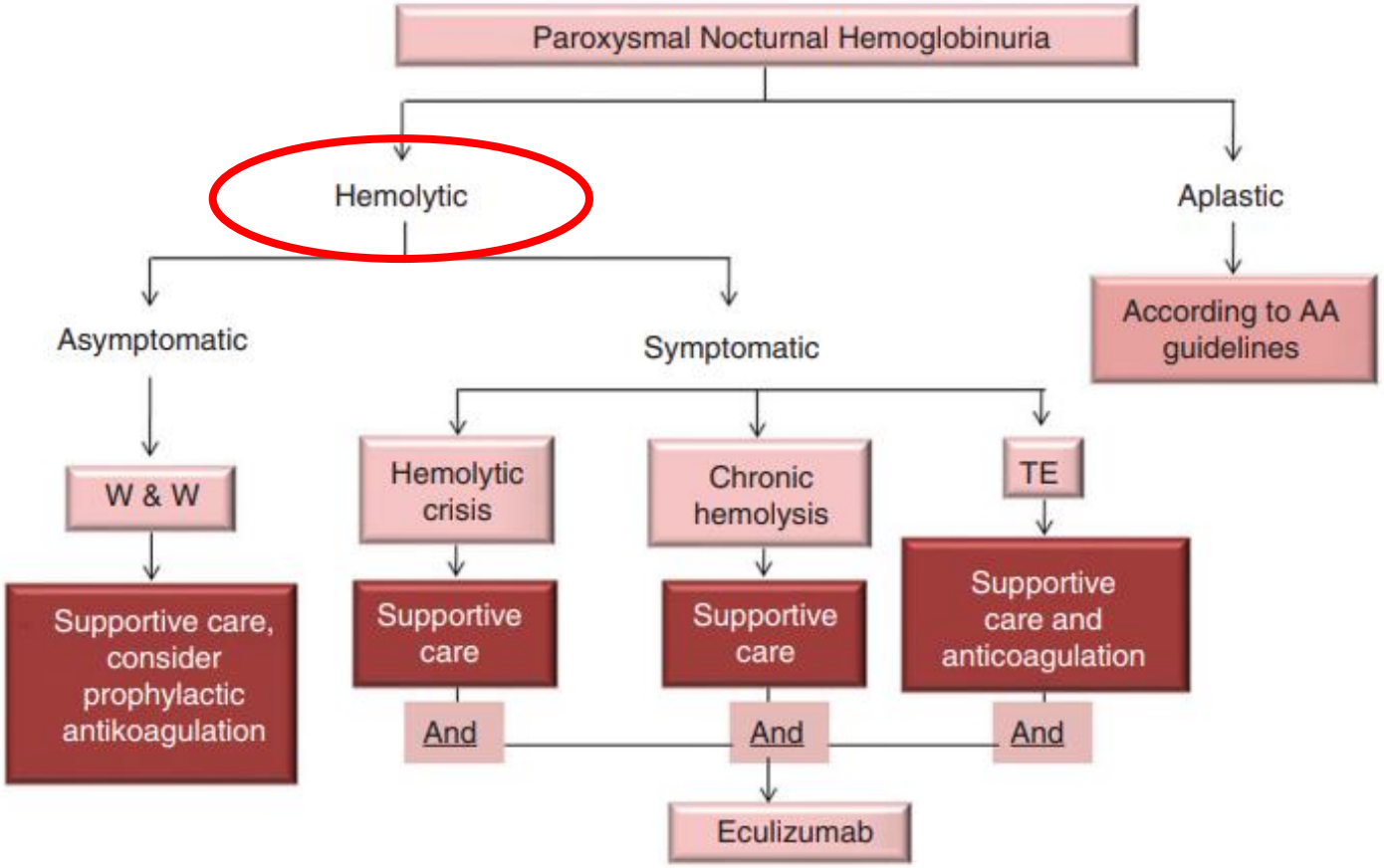
Case report (II)

PNH Clone 28,7% neutrophils; 25,4 % monocytes

Diagnosis of PNH confirmed!

Treatment?

Treatment



Schrezenmeier et al, Treatment algorithm adopted from the German PNH guidelines

Treatment

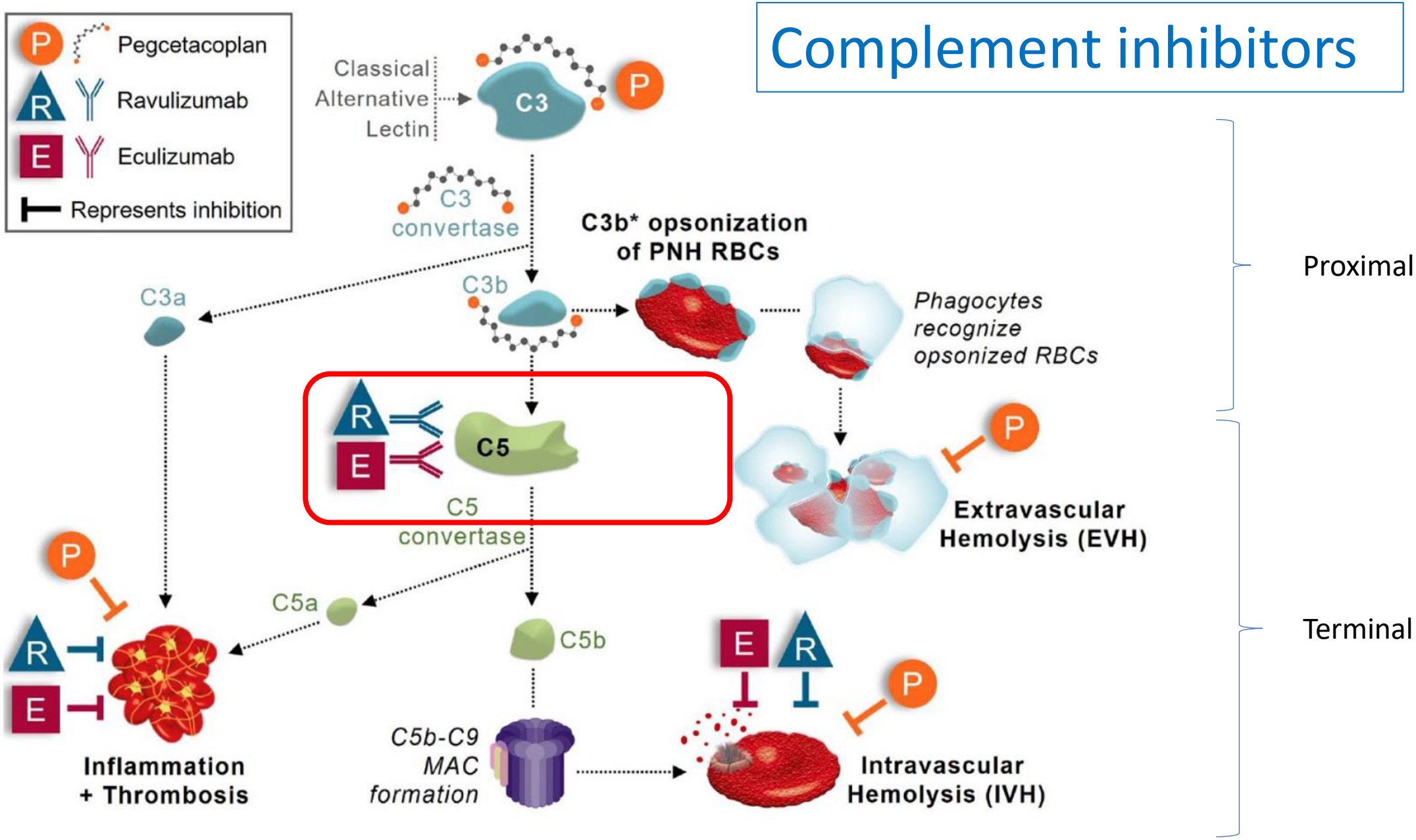
HSCT is the only curative option → use limited by treatment-related mobidity and mortality

- Key treatment if AA/PNH overlap
- Option when no access to complement inhibitors

Complement inhibitors

- To reduce hemolysis
- To reduce thromboembolic risk

Complement inhibitors



Raymond S.M. Wong. Ther Adv Hematol 2022, Vol. 13: 1–17 DOI: 10.1177/ 20406207221114673

Eculizumab

- Terminal complement inhibitor targeting C5
- Prevention of MAC assemblys and so intravascular hemolysis
- Stabilization of Hb and less transfusions
- Resolution of related symptoms
- Reduce thromboembolic risk

- ! Vaccination against Neisseria meningitides
- 600 mg weekly for 4 weeks, then 900 mg/2 weeks

Ravulizumab

- Second generation therapeutic complement inhibitor targeting C5
- Half life benefit → administration /8 weeks
- Stable responders on Eculizumab or newly diagnosed and severe PNH

Response to anti-complement agents in PNH

TABLE 1 | Tentative classification of hematological response to anti-complement agents in PNH.

Response category	Red blood cell transfusions	Hemoglobin level	LDH level*‡	ARC*
Complete response	None	≥12 g/dL	≤1.5x ULN	***and*** ≤150,000/μL§
Major response	None	≥12 g/dL	>1.5x ULN	***or*** >150,000/μL§
Good response	None	≥10 and <12 g/dL	A. ≤1.5x ULN B. >1.5x ULN	Rule out bone marrow failure°
Partial response	None or occasional (≤2 every 6 months)	≥8 and <10 g/dL	A. ≤1.5x ULN B. >1.5x ULN	Rule out bone marrow failure°
Minor response#	None or occasional (≤2 every 6 months) Regular (3–6 every 6 months) Reduction by ≥50%^	<8 g/dL <10 g/dL <10 g/dL	A. ≤1.5x ULN B. >1.5x ULN	Rule out bone marrow failure°
No response#	Regular (>6 every 6 months)	<10 g/dL	A. ≤1.5x ULN B. >1.5x ULN	Rule out bone marrow failure°

Risitano et al. Anti-complement Treatment for Paroxysmal Nocturnal Hemoglobinuria: Time for Proximal Complement Inhibition? A Position Paper From the SAAWP of the EBMT. Front. Immunol., 14 June 2019
Sec. Molecular Innate Immunity
https://doi.org/10.3389/fimmu.2019.01157

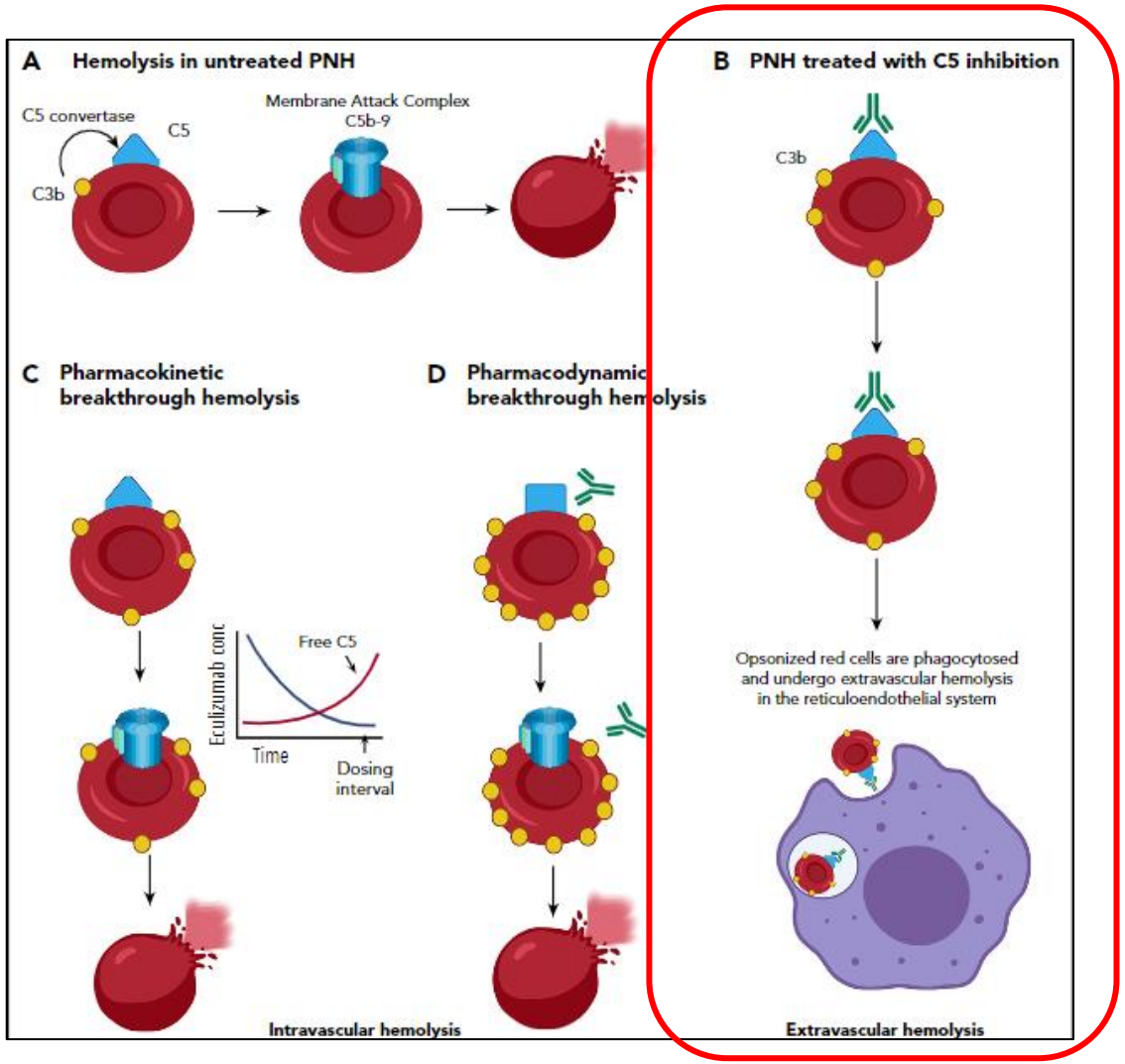
No response to treatment ?

Intravascular Hemolysis during Eculizumab treatment

- Minimal residual intravascular hemolysis is normal
- Breakthrough hemolysis

Pharmakokinetic breakthrough	Suboptimal plasma level until next administration
Pharmacodynamic breakthrough	Massive complement activation

C3- Mediated Extravascular Hemolysis



- Surface complement activation on affected cells continues
- Phagocytosis of opsonized cells
- Extravascular hemolysis

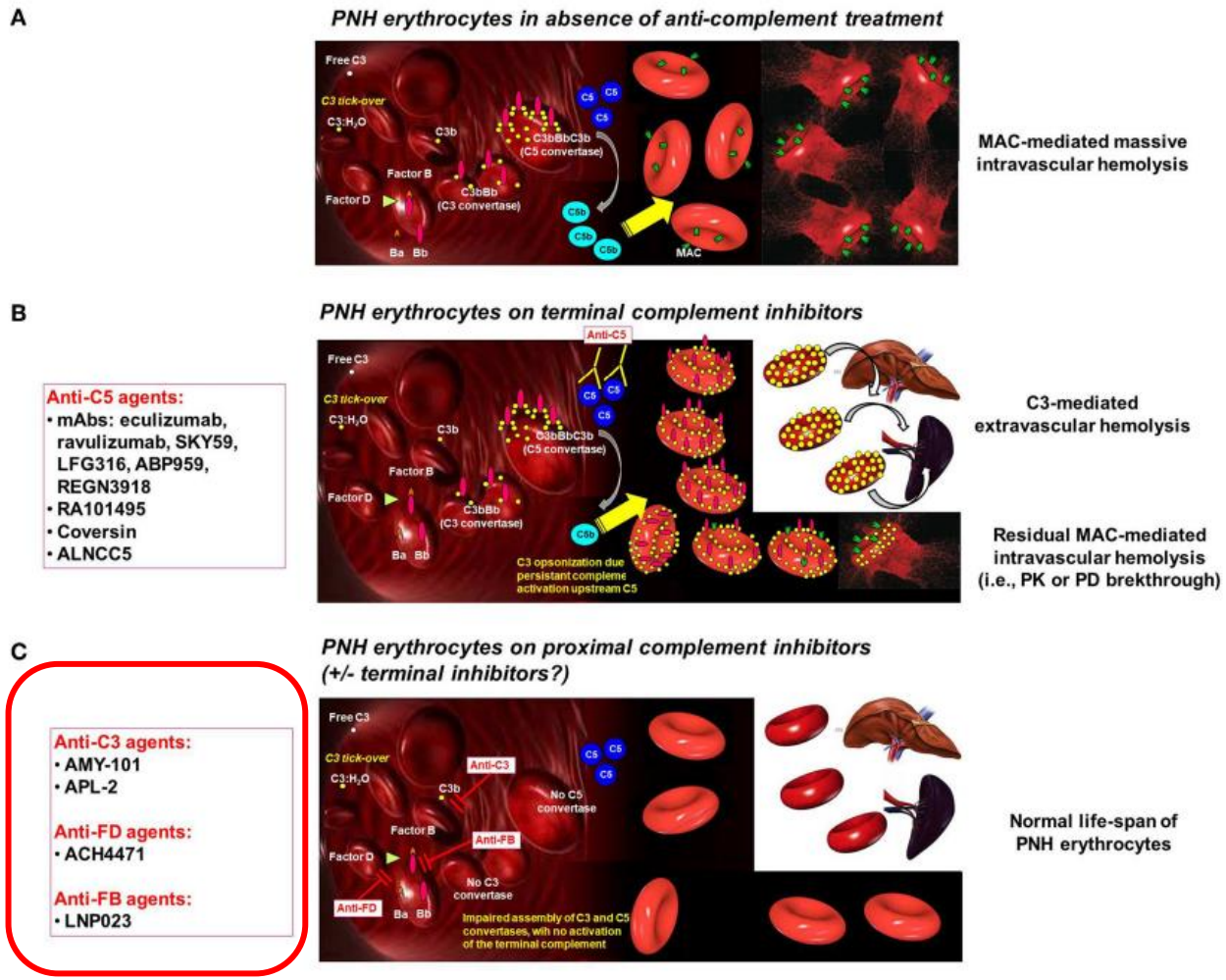
Underlying bone marrow failure

Robert A Brodsky. blood® 11 MARCH 2021 | VOLUME 137, NUMBER 10

Future of PNH treatment …

Pegcetacoplan C3
(Pegasus)

Iptacopan Factor B
(Apply PNH-Study)



Risitano et al. Anti-complement Treatment for Paroxysmal Nocturnal Hemoglobinuria: Time for Proximal Complement Inhibition? A Position Paper From the SAAWP of the EBMT. Front. Immunol., 14 June 2019 Sec. Molecular Innate Immunity
https://doi.org/10.3389/fimmu.2019.01157

What‘s about case report?

Three month later…

- Significant hemolysis with transfusions
- Platelets 36000/mm³

Bone marrow:

- Cytology with mild signs of dysgranulopoesis
- Biopsy: hypocellular <60 % with increased erythropoesis and infiltration by T lymphocytes

PNH with development of non severe aplastic anemia?

PNH/ AA overlap syndrome

- Treatment of the most relevant problem
- In this case hemolysis and so complement inhibitor
- If sAA or vsAA → treatment for aplastic anemia
- ! Response criteria could be influenced by underlying bone marrow failure syndrome

Take Home message

- Acquired disorder of HSC leading to intravascular hemolysis and thromboembolic complications
- Often accompanied by cytopenia with risk of developement AA
- Diagnosis by flow cytometry
- Treatment by complement inhibitors for symptomatic PNH (hemolysis or thrombosis)
- Future → proximal complement inhibitors
- Treatment like AA in aplastic forms

Thank you for your attention