

EPN E INSUFFICIENZE MIDOLLARI

descrizione e casi clinici

Francesco Lanza, U.O. Ematologia

Ospedale S.M. Delle Croci –Ravenna

Rete Trapianti Metropolitana della Romagna



CLASSIFICATION OF HAEMOLYTIC ANAEMIAS

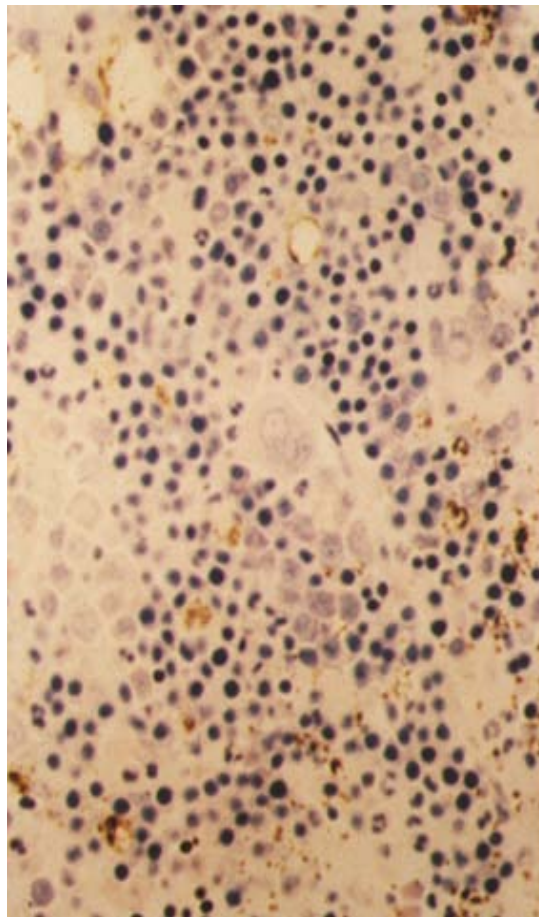
	<i>Intracorporeal causes</i>	<i>Extracorporeal causes</i>
Hereditary	<ul style="list-style-type: none">• Hemoglobinopathies• Enzymopathies• Membranopathies• Other (Rare PNH ?)	Familial HUS (due to defective C reg protein)
Acquired	Paroxysmal Nocturnal Hemoglobinuria (PNH)	<ul style="list-style-type: none">• Malaria• Auto-immune• Drug-induced• Micro-angiopathic• Other

PNH Classification

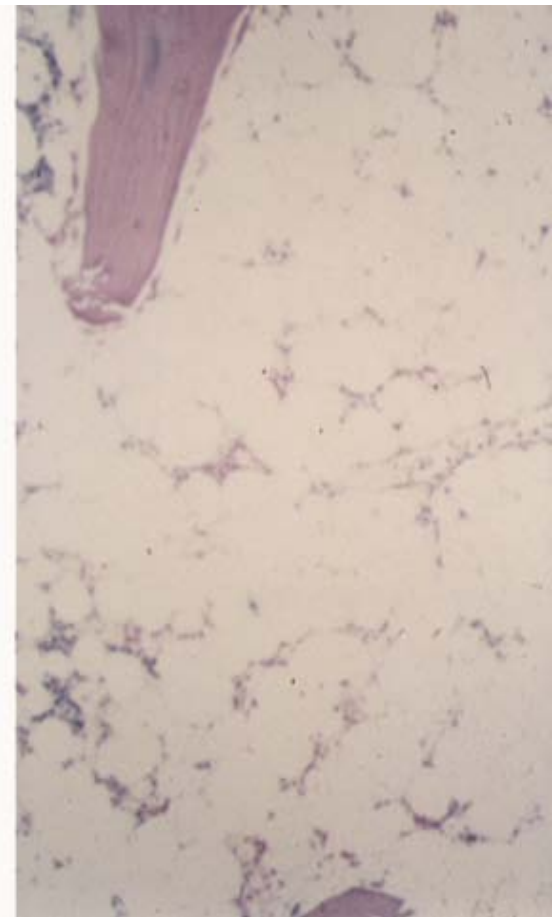
Parker et al, Blood 2005

Category	Hemolysis	BMF
1) Classical (Florid PH)	+	-
2) PNH associated to BM disorders (MDS, AA)	+	+
3) Subclinical	-	+

There are two major forms of PNH

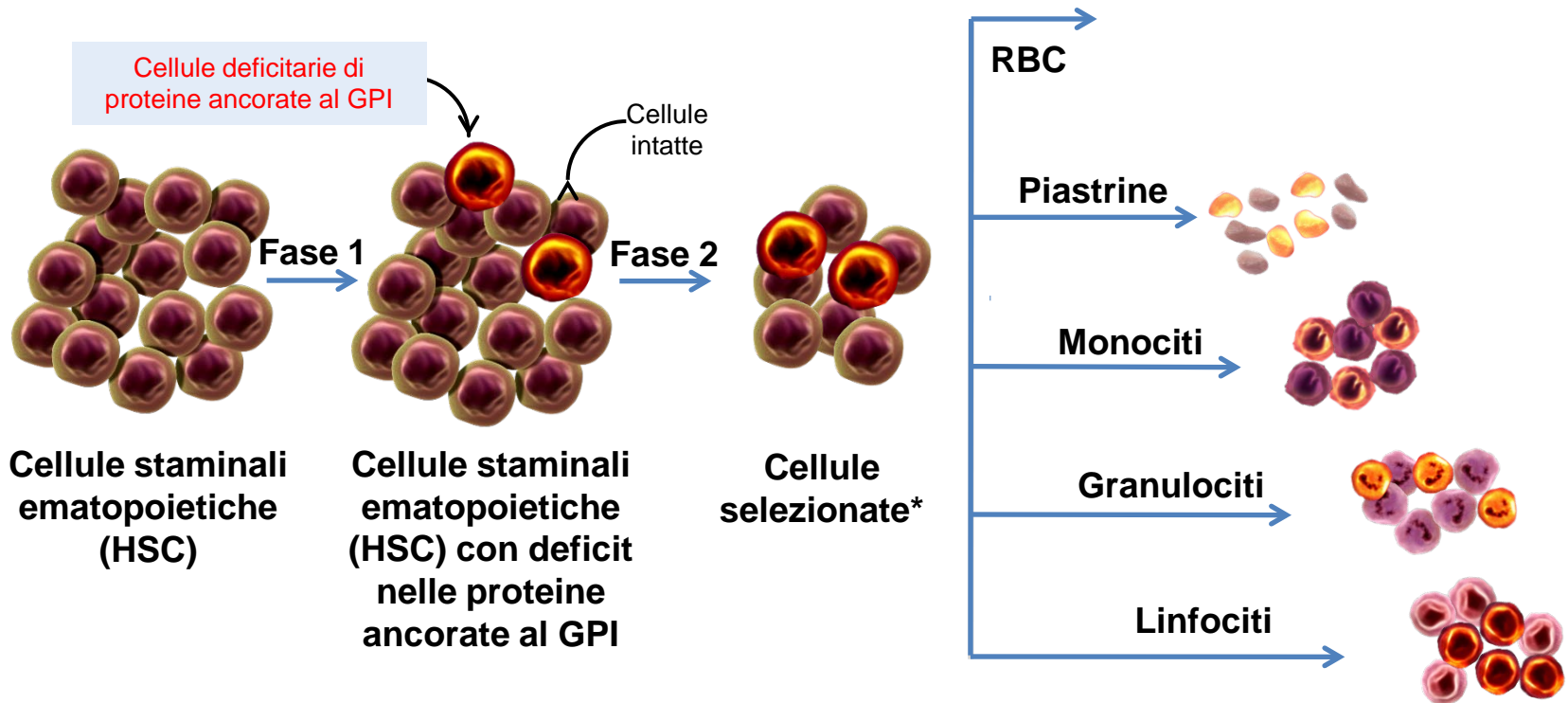


1. Florid haemolytic



2. PNH/aplastic anaemia syndrome

Le cellule emopoietiche presentano un deficit nelle proteine ancorate al GPI



Mutazione somatica
nel *PIG-A*

**Attacco
autoimmune**

Espansione clonale
tramite
immunoselezione

Espansione benigna simil-tumorale

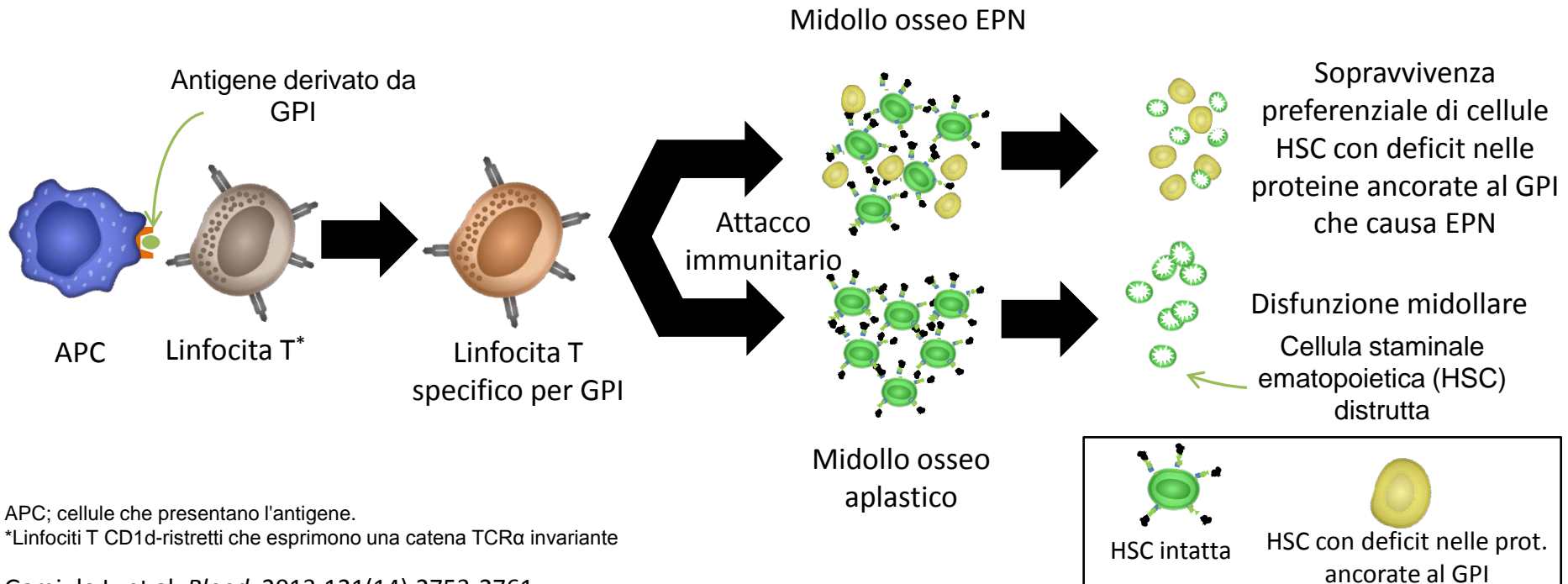
*Cloni che si presume abbiano un vantaggio di crescita conferito, in aggiunta alla mutazione del gene *PIG-A*, con conseguente deficit nelle proteine ancorate al GPI.

Inoue N, et al. *Int J Hematol.* 2003;77(2):107-112.

L'ancora GPI può essere il target dell'attacco immunitario in pazienti con EPN

- Cellule con deficit nelle proteine ancorate al GPI possono avere una sopravvivenza preferenziale a causa di un processo autoimmune mediato dai linfociti T, che ha come target le cellule staminali ematopoietiche GPI positive, ma non quelle GPI negative¹
- In pazienti con EPN è stato osservato un numero elevato di linfociti T reattivi verso il GPI*¹

Potenziale meccanismo dell'attacco immunitario nell'EPN

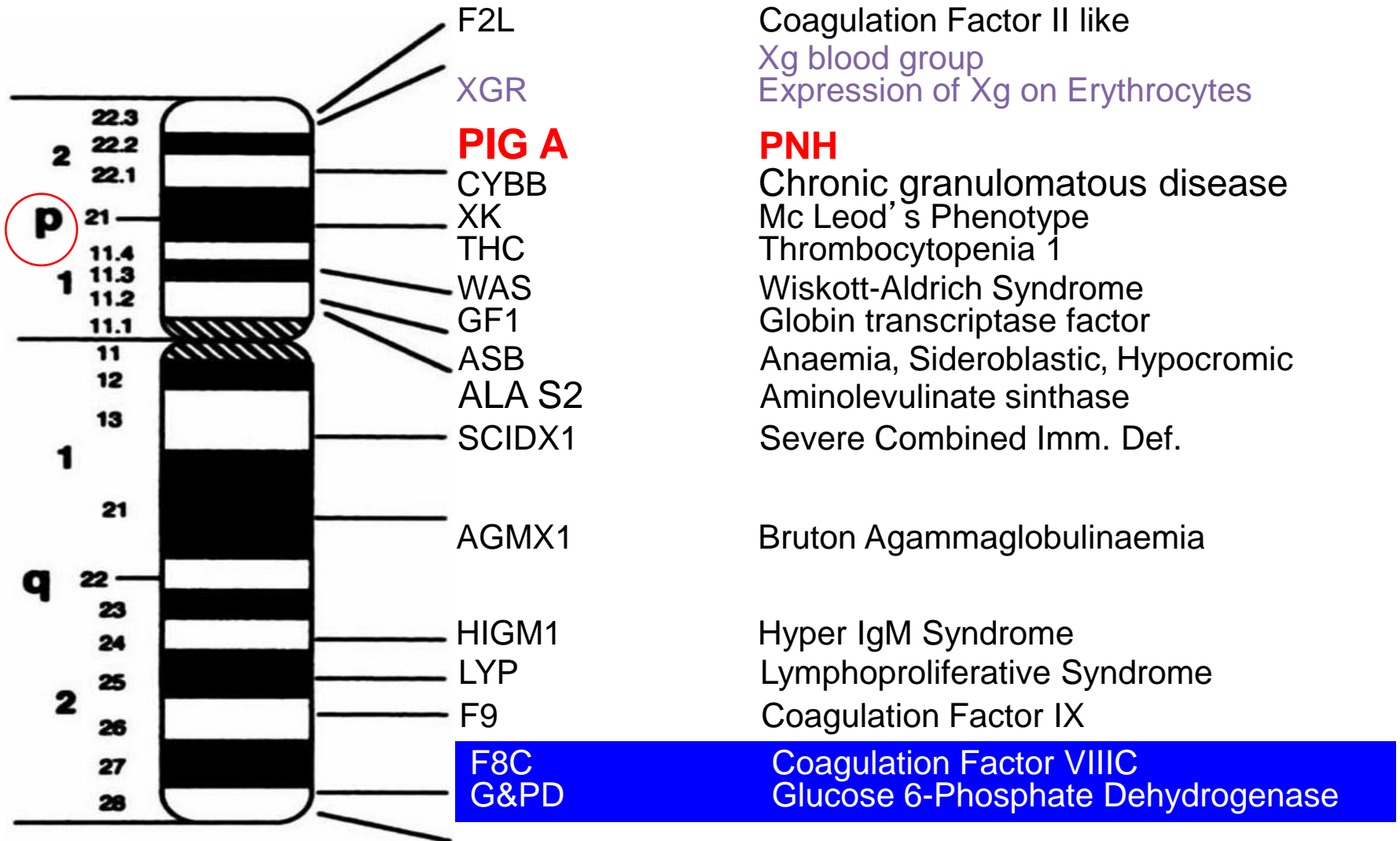


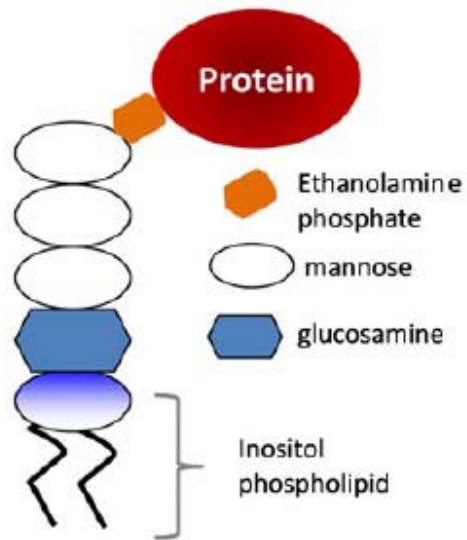
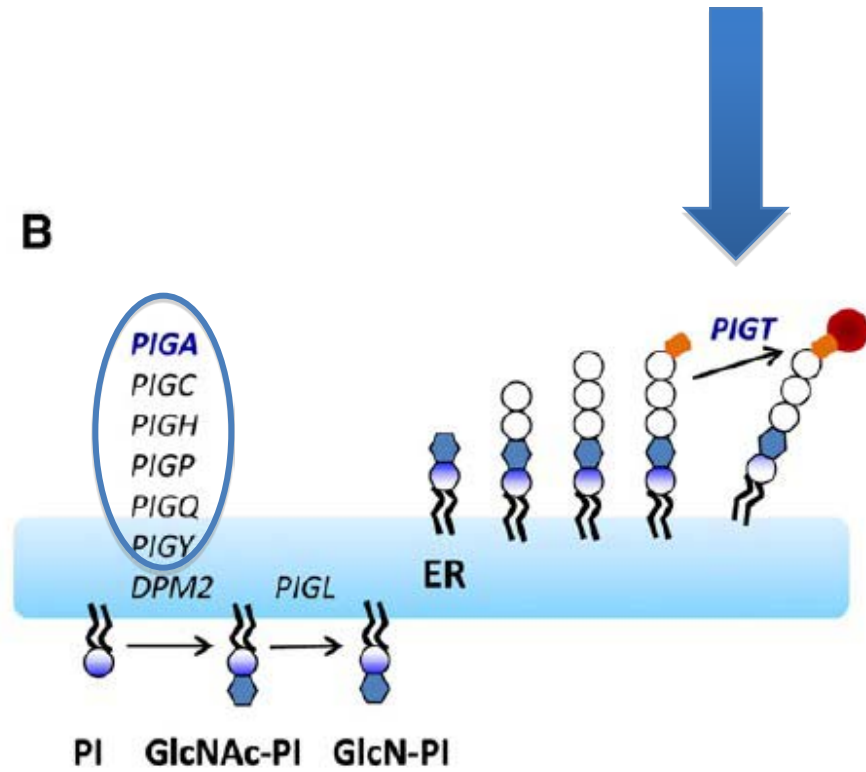
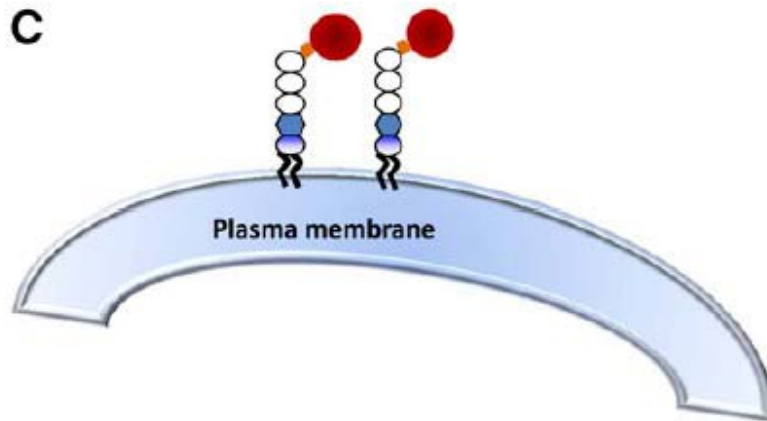
APC; cellule che presentano l'antigene.

*Linfociti T CD1d-ristretti che esprimono una catena TCR α invariante

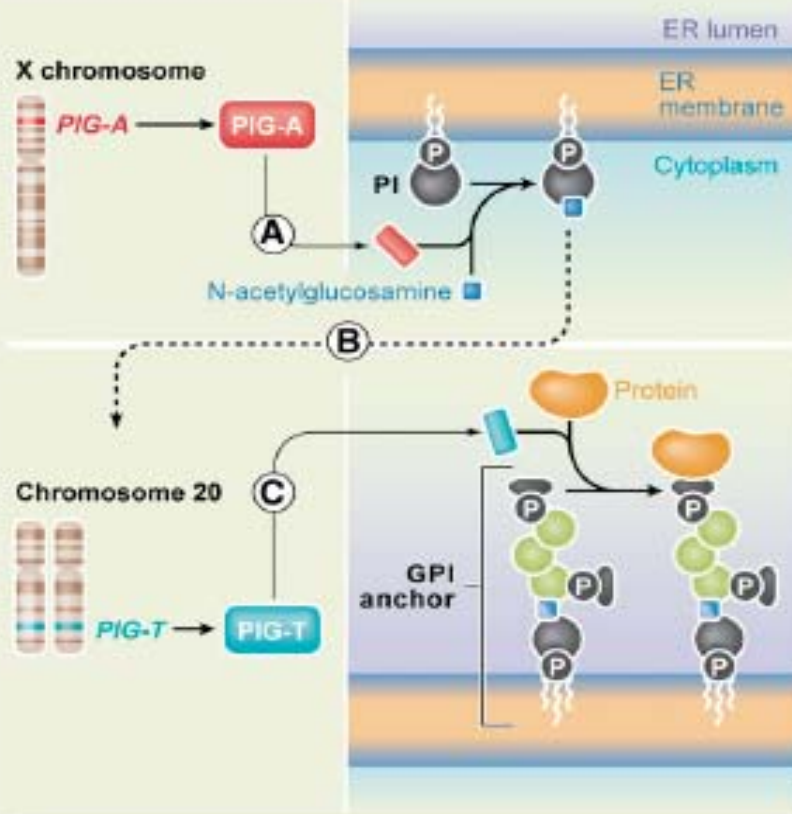
Gargiulo L, et al. *Blood*. 2013;121(14):2753-2761.

Somatic mutation(s) in the X-linked PIG-A gene

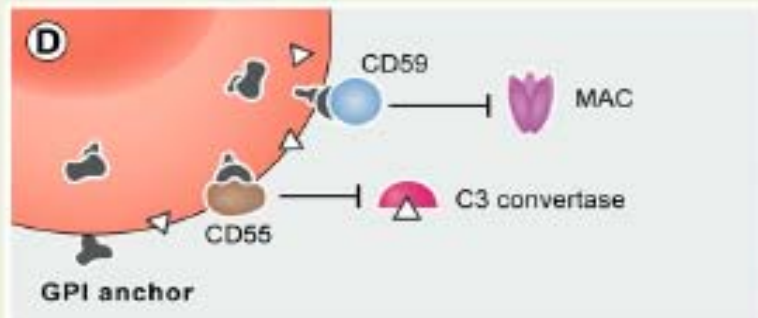


A**B****C**

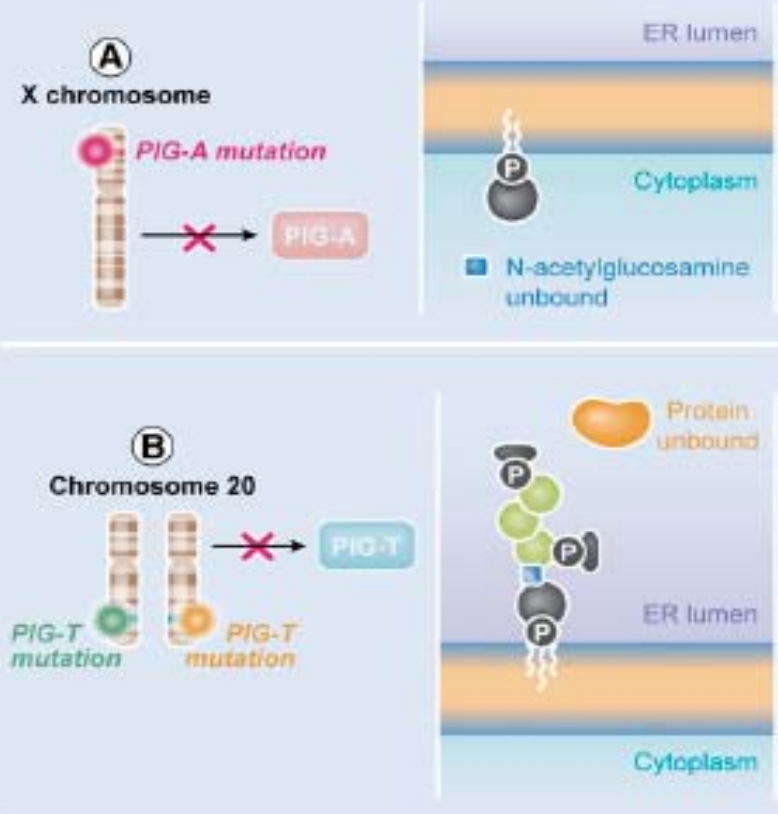
NORMAL ERYTHROCYTE



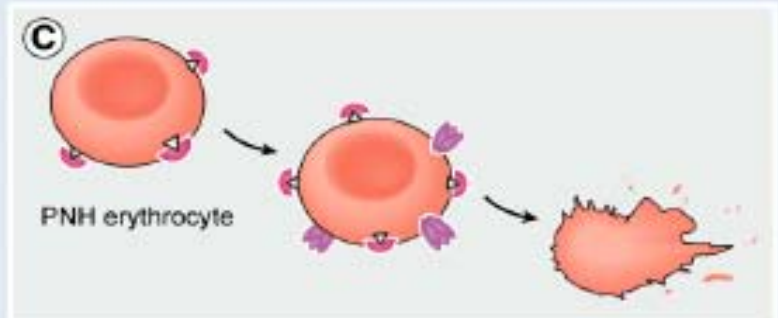
GPI anchor tethers C-regulatory proteins



PNH ERYTHROCYTE



No GPI anchor



RED CELLS, IRON, AND ERYTHROPOIESIS

A case of paroxysmal nocturnal hemoglobinuria caused by a germline mutation and a somatic mutation in *PIGT*

Peter M. Krawitz,¹ Britta Höchsmann,² Yoshiko Murakami,³ Britta Teubner,¹ Ulrike Krüger,¹ Eva Klopocki,⁴ Heidemarie Neitzel,¹ Alexander Hoellein,⁵ Christina Schneider,² Dmitri Parkhomchuk,¹ Jochen Hecht,⁶ Peter N. Robinson,¹ Stefan Mundlos,¹ Taroh Kinoshita,³ and Hubert Schrezenmeier²

Key Points

- A carrier of a deleterious splice site mutation in *PIGT* acquired a second hit in *PIGT* and developed PNH.

To ascertain the genetic basis of a paroxysmal nocturnal hemoglobinuria (PNH) case without somatic mutations in *PIGA*, we performed deep next-generation sequencing on all exons of known genes of the glycosylphosphatidylinositol (GPI) anchor synthesis pathway. We identified a heterozygous germline splice site mutation in *PIGT* and a somatic 8-MB deletion in granulocytes affecting the other copy of *PIGT*. *PIGA* is essential for GPI anchor synthesis, whereas *PIGT* is essential for attachment of the preassembled GPI anchor to proteins. Although a single mutation event in the

● ● ● RED CELLS, IRON, & ERYTHROPOIESIS

Comment on Krawitz et al, page 1312

PNH from mutations of another *PIG* gene

Lucio Luzzatto¹ ¹ISTITUTO TOSCANO TUMORI

In this issue of *Blood*, Krawitz et al report on a patient with paroxysmal nocturnal hemoglobinuria (PNH) who does not have a mutation of *PIG-A*, but in whom instead both alleles of *PIG-T* (another gene involved in glucosylphosphatidylinositol [GPI] biosynthesis) have inactivating mutations, one in the germ line and one somatic.¹

Deep sequencing reveals stepwise mutation acquisition in paroxysmal nocturnal hemoglobinuria

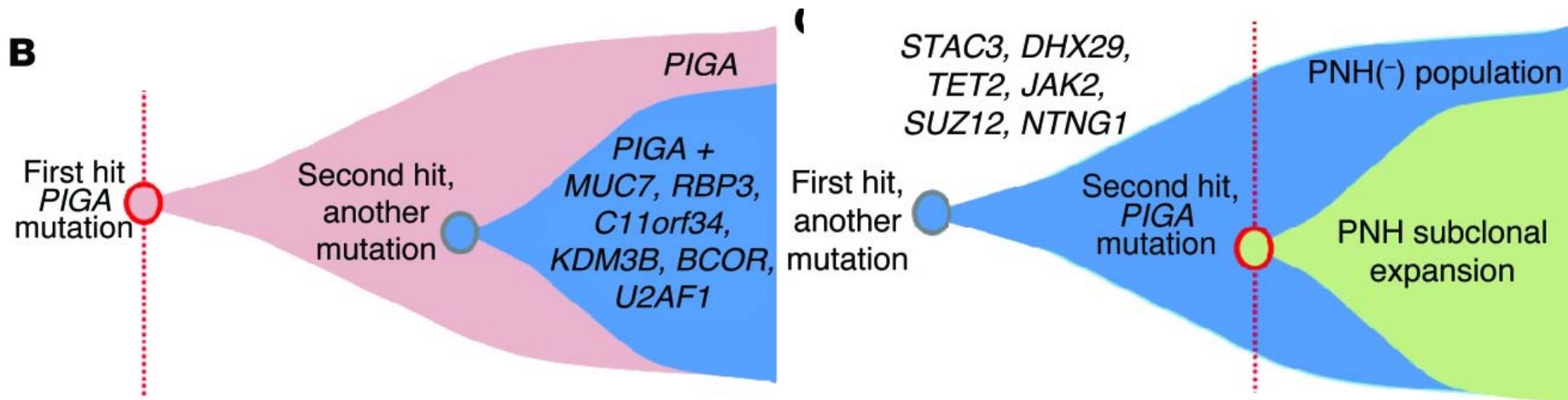
Wenyi Shen,^{1,2} Michael J. Clemente,¹ Naoko Hosono,¹ Kenichi Yoshida,³ Bartłomiej Przychodzen,¹ Tetsuichi Yoshizato,³ Yuichi Shiraishi,⁴ Satoru Miyano,^{4,5} Seishi Ogawa,³ Jaroslaw P. Maciejewski,¹ and Hideki Makishima¹

¹Department of Translational Hematology and Oncology Research, Taussig Cancer Institute, Cleveland Clinic, Cleveland, Ohio, USA. ²Department of Hematology, The First Affiliated Hospital of Nanjing Medical University, Nanjing, Jiangsu, China. ³Department of Pathology and Tumor Biology, Graduate School of Medicine, Kyoto University, Kyoto, Japan. ⁴Laboratory of DNA Information Analysis, Human Genome Center and ⁵Laboratory of Sequence Analysis, Human Genome Center, Institute of Medical Science, The University of Tokyo, Tokyo, Japan.

Paroxysmal nocturnal hemoglobinuria (PNH) is a nonmalignant clonal disease of hematopoietic stem cells that is associated with hemolysis, marrow failure, and thrombophilia. PNH has been considered a monogenic disease that results from somatic mutations in the gene encoding *PIGA*, which is required for biosynthesis of glycosylphosphatidylinositol-anchored (GPI-anchored) proteins. The loss of certain GPI-anchored proteins is hypothesized to provide the mutant clone with an extrinsic growth advantage, but some features of PNH argue that there are intrinsic drivers of clonal expansion. Here, we performed whole-exome sequencing of paired PNH⁺ and PNH⁻ fractions on samples taken from 12 patients as well as targeted deep sequencing of an additional 36 PNH patients. We identified additional somatic mutations that resulted in a complex hierarchical clonal architecture, similar to that observed in myeloid neoplasms. In addition to mutations in *PIGA*, mutations were found in genes known to be involved in myeloid neoplasm pathogenesis, including *TET2*, *SUZ12*, *U2AF1*, and *JAK2*. Clonal analysis indicated that these additional mutations arose either as a subclone within the *PIGA*-mutant population, or prior to *PIGA* mutation. Together, our data indicate that in addition to *PIGA* mutations, accessory genetic events are frequent in PNH, suggesting a stepwise clonal evolution derived from a singular stem cell clone.

SELECTIVE GROWTH ADVANTAGE

- **PIG-A mutations confer a growth advantage**
- **A second anti-apoptotic mutation occurs**



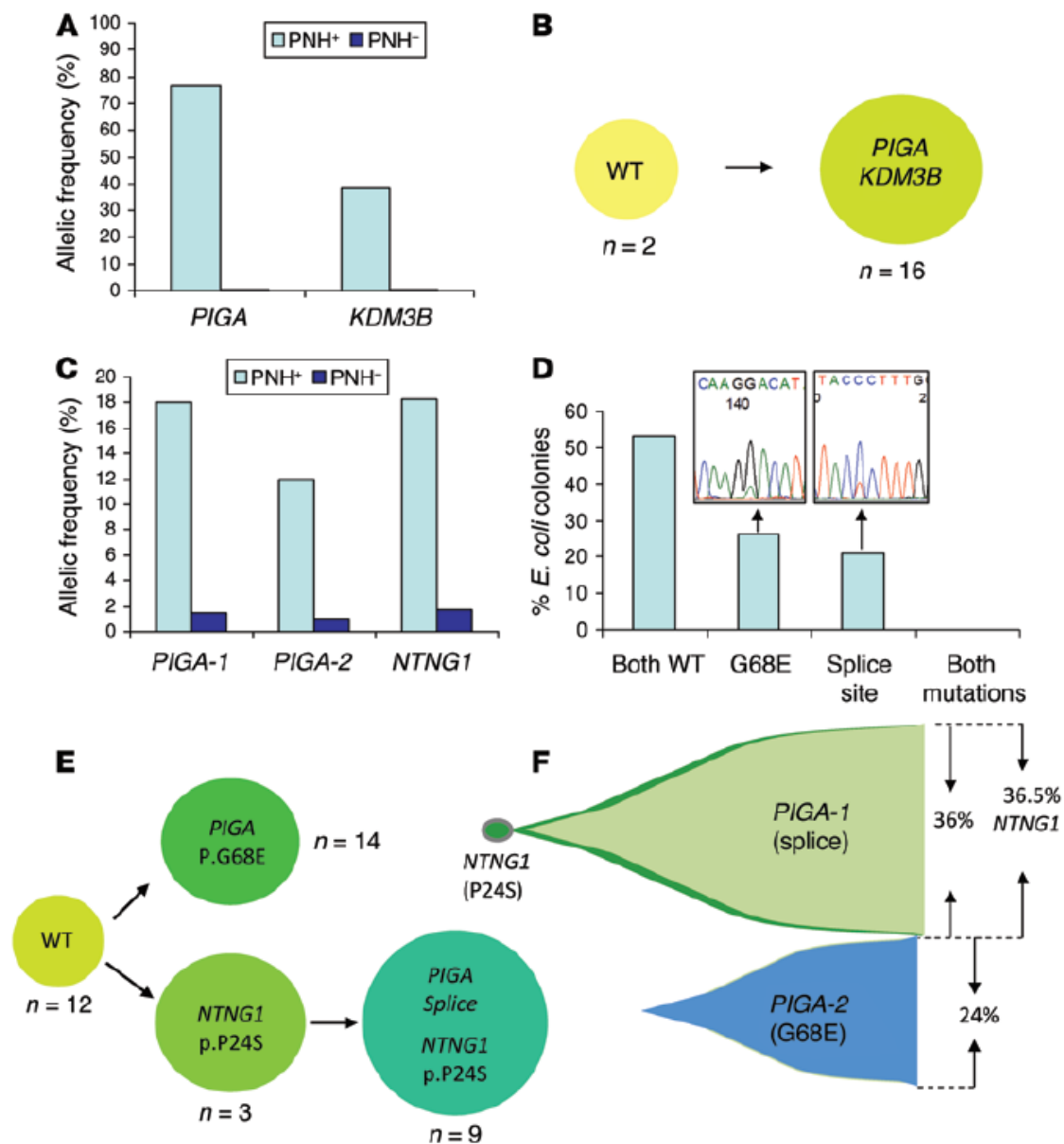
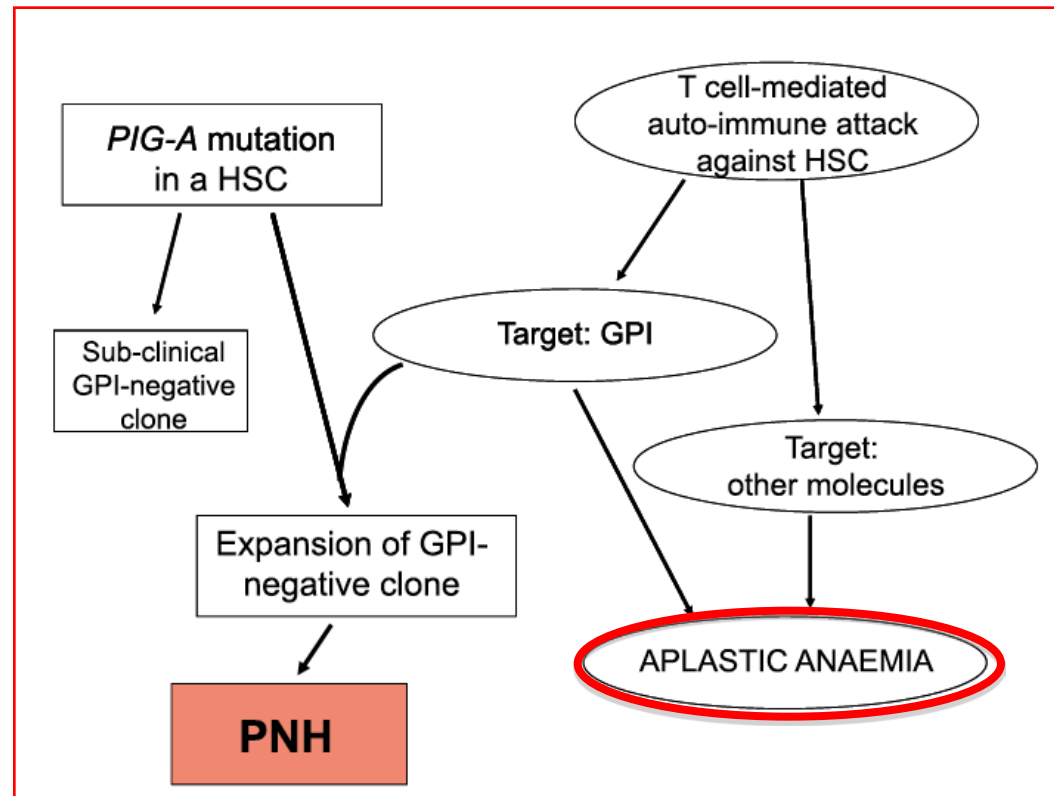


Figure 1. *PIGA* mutations can be either primary or secondary events. The primary event (PNH5) is represented in **A** and **B**; the secondary event (PNH1) is represented in **C–F**. **(A)** Analysis of VAFs of the mutations identified in case PNH5 indicated that the *KDM3B* mutation was present at a lower frequency than the *PIGA* mutation, and both mutations were almost exclusively confined to the sorted PNH⁺ (CD59⁺) fraction. **(B)** Single-colony sequencing results confirmed that the *PIGA* and *KDM3B* mutations were present in the same cell population. **(C)** Deep sequencing VAFs for *PIGA-1* (G68E), *PIGA-2* (splice site), and *NTNG1* (P24S) mutations, all of which were primarily present in the PNH fraction in the PNH1 case. **(D)** Bacterial subcloning and Sanger sequencing results demonstrated that the *PIGA* mutations in this case were independent, suggesting the presence of 2 separate PNH clones. **(E)** Single-colony sequencing further confirmed that 2 independent PNH clones were present and also suggested that the *PIGA* splice site mutation appeared to be a secondary event preceded by a *NTNG1* mutation. **(F)** The combination of deep sequencing data with single-colony sequencing allowed for a representation of the clonal architecture in PNH1.

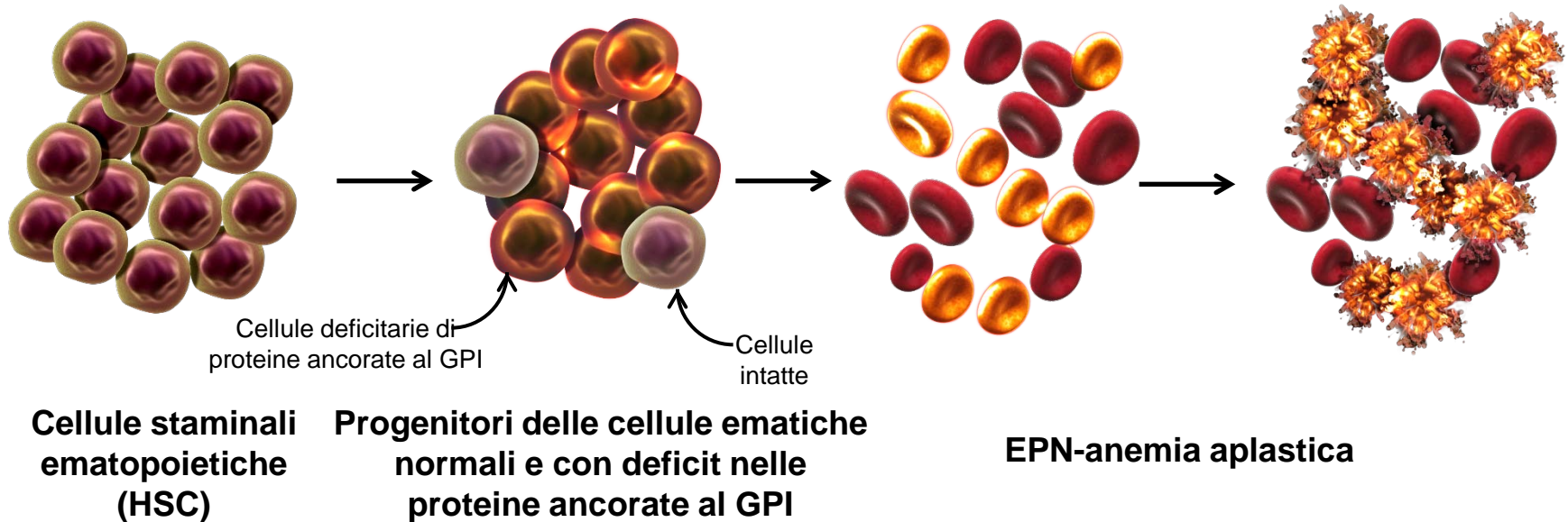
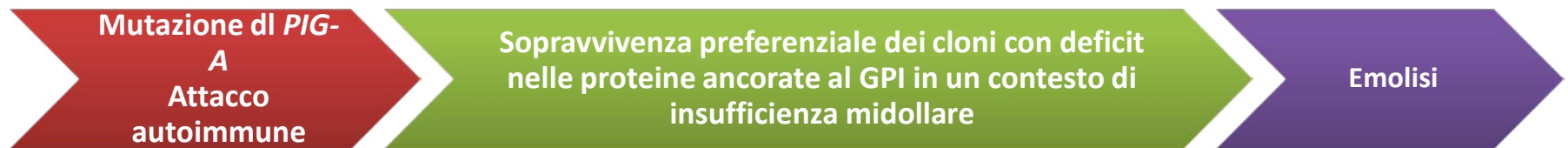
CONDITIONAL GROWTH ADVANTAGE

- PIG-A mutations are a benign event
- PNH cells undergo clonal expansion in the setting of immune selection
 - GPI anchored antigens are the targets of immune attack



Un contesto di disfunzione midollare può promuovere l'espansione di un clone con deficit nelle proteine ancorate al GPI in pazienti con AA¹⁻³

Le cellule staminali con deficit nelle proteine ancorate al GPI hanno un vantaggio competitivo in un contesto di disfunzione midollare²



1. Brodsky RA, Jones RJ. *The Lancet*. 2005;365(9471):1647-1656.
2. Kinoshita T, Inoue N. *Int. J. Hematol*. 2002;75(2):117-122.
3. Tiu R et al. *Leucemia*. 2007;21:1648-1657.

Development of paroxysmal nocturnal hemoglobinuria in CALR-positive myeloproliferative neoplasm

Fraiman et al.

Abstract: Paroxysmal nocturnal hemoglobinuria (PNH), a disease characterized by intravascular hemolysis, thrombosis, and bone marrow failure, is associated with mutations in the *PIG-A* gene, resulting in a deficiency of glycosylphosphatidylinositol-anchored proteins. Many hypotheses have been posed as to whether PNH and *PIG-A* mutations result in an intrinsic survival benefit of CD55⁻/CD59⁻ cells or an extrinsic permissive environment that allows for their clonal expansion within the bone marrow compartment. Recent data have identified the concurrence of *PIG-A* mutations with additional genetic mutations associated with myeloproliferative disorders, suggesting that some presentations of PNH are the result of a stepwise progression of genetic mutations similar to other myelodysplastic or myeloproliferative syndromes. We report for the first time in the literature the development of clinically significant PNH in a patient with *JAK2*^{V617F}-negative, *CALR*-positive essential thrombocythemia, providing further support to

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www.nature.com/bcj

LETTER TO THE EDITOR

Paroxysmal nocturnal hemoglobinuria and concurrent *JAK2*^{V617F} mutation

Sugimuri et al. 2012

Blood Cancer Journal (2012) 2, e63; doi:10.1038/bcj.2012.7;
published online 23 March 2012

oncogenesis and may be supported by the observation that patients with PNH can harbor clonal cytogenetic abnormalities.⁸ However, no specific gene mutations other than *PIG-A* have been

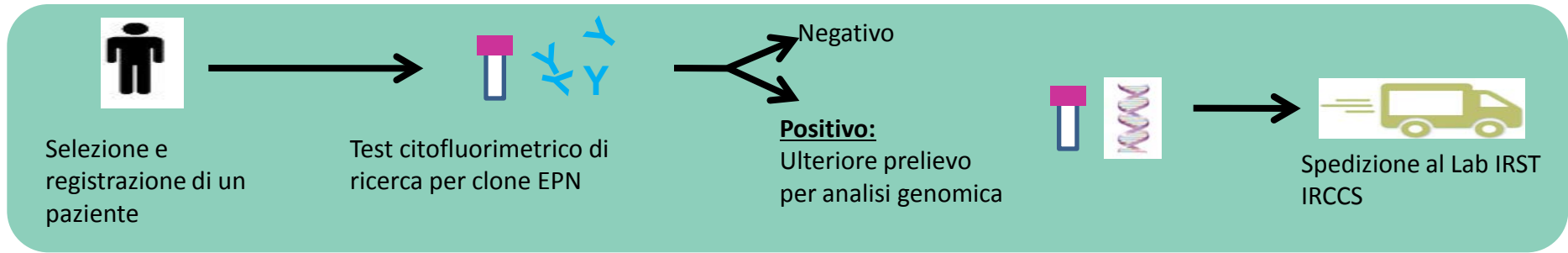
STUDIO DI PREVALENZA DEI CLONI EPN IN PAZIENTI CON NEOPLASIE MIELOPROLIFERATIVE PHILADELPHIA-NEGATIVE

1. **UOC Ematologia Ospedale S. Maria delle Croci Ravenna (centro coordinatore)**
 2. UOC Ematologia Ospedale Infermi **Rimini**
 3. UO di Ematologia IRST IRCCS – **Meldola Forli**
 4. UO Ematologia Azienda Ospedaliero Universitaria Integrata- **Verona -**
 5. UO Ematologia Azienda Ospedaliero Universitaria- **Siena**
 6. Azienda Ospedaliero Universitaria- **Udine**
 7. Ospedale “S. Bortolo”, - **Vicenza**
 8. Azienda Ospedaliero Universitaria - **Padova**
- Laboratorio Unico Pievesestina (Lab Citometria) :
 - Laboratorio Bioscienze IRST IRCCS
 - Laboratorio di Siena, Verona, Vicenza, Padova e Udine

- **Lo studio**
- **Procedure**
- **Avanzamento**
- **GDPR**
- **Contatti**



WBC	FLAER	CD157	CD64	CD15	CD45
Strumentazione Beckman Coulter	Alexa-488 FITC	PE	PC7	APC	5 canale
Strumentazione Beckton Dickinson	Alexa-488 FITC	PE	PC7	APC	5 canale

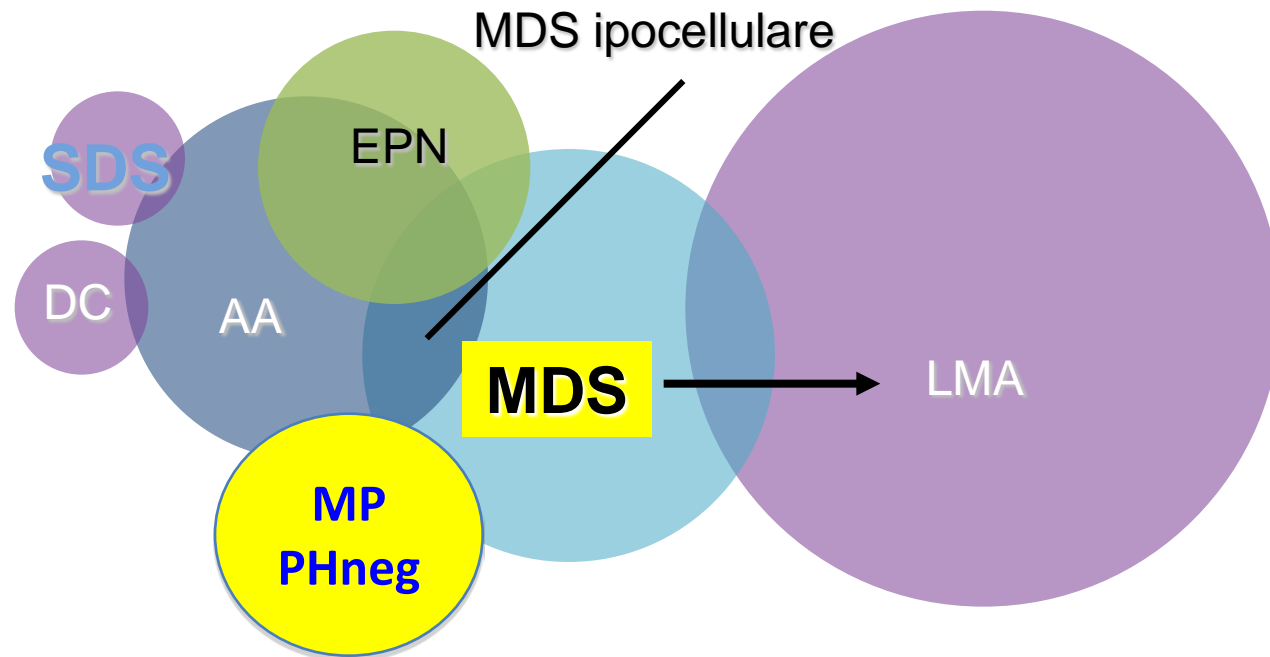


1
2.
3

RBC	CD235a	CD59	CD45
Strumentazione Beckman Coulter	FITC	PE	APC
Strumentazione Beckton Dickinson	FITC	PE	APC

PNH vs other BM failure disorders (BMF)

PNH-related sign & symptoms may be similar to those of other BMF disorders, and the clinical course may change significantly over time ^{1,2}

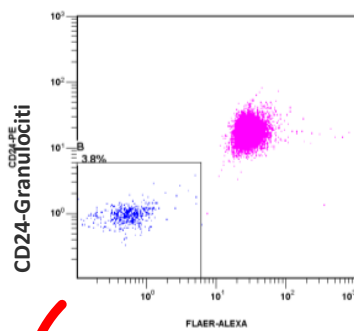


LMA, leucemia mieloide acuta; AA, anemia aplastica; DC, discheratosi congenita; MDS, sindrome mielodisplastica; SDS, sindrome di Shwachman-Diamond.

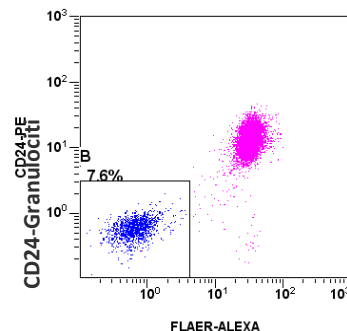
I cloni con deficit nelle proteine ancorate al GPI possono espandersi rapidamente o più raramente estinguersi

- I cloni con deficit nelle proteine ancorate al GPI potrebbero espandersi rapidamente nel corso del tempo in pazienti con AA¹⁻³

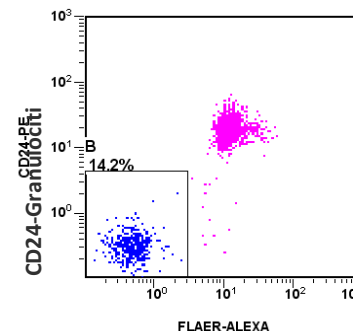
Espansione di cloni con deficit nelle proteine ancorate al GPI⁴



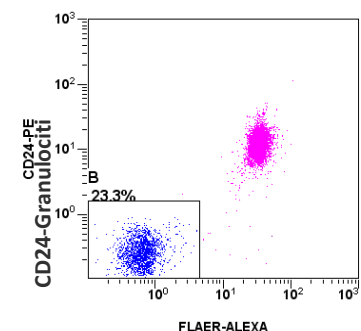
FLAER: Marcatore che si lega all'ancora di GPI



FLAER: Marcatore che si lega all'ancora di GPI



FLAER: Marcatore che si lega all'ancora di GPI



FLAER: Marcatore che si lega all'ancora di GPI

Dimensioni clone granulocitario

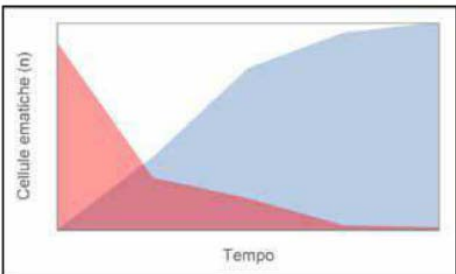
3,8%

7,6%

14,2%

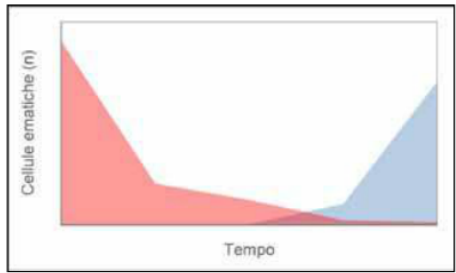
23,3%

9 mesi

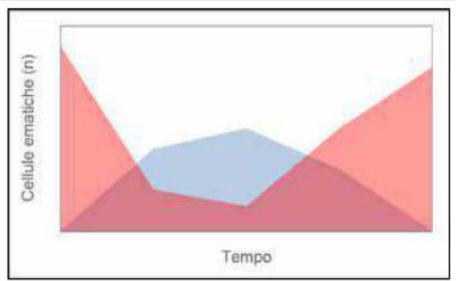


**Fast and complete
escape**

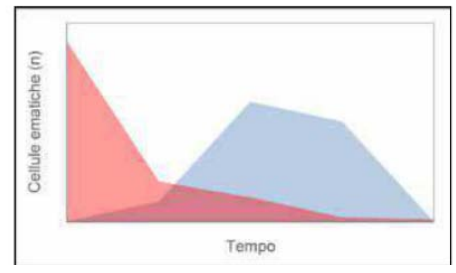
Flord PNH



**Late and complete
escape**

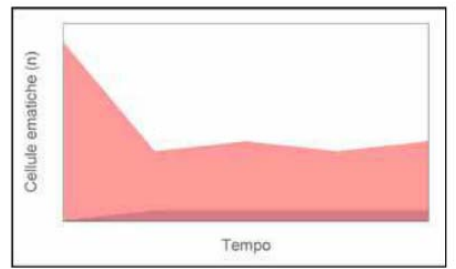


**Complete but temporary
escape, with normal
hematopoietic reconstitution**



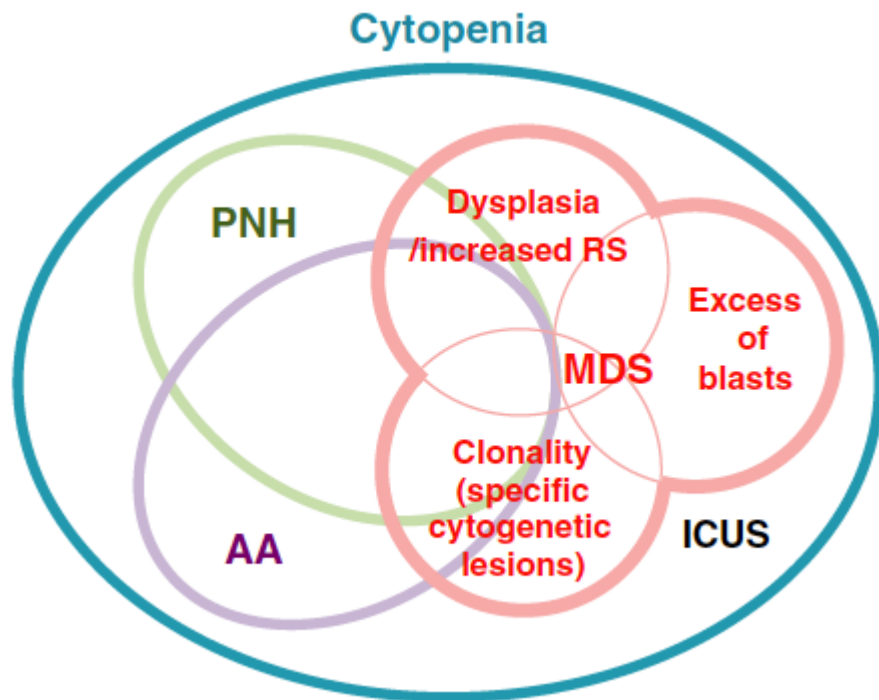
**Complete but
temporary escape**

**SAA/AA-
PNH
sc-PNH**



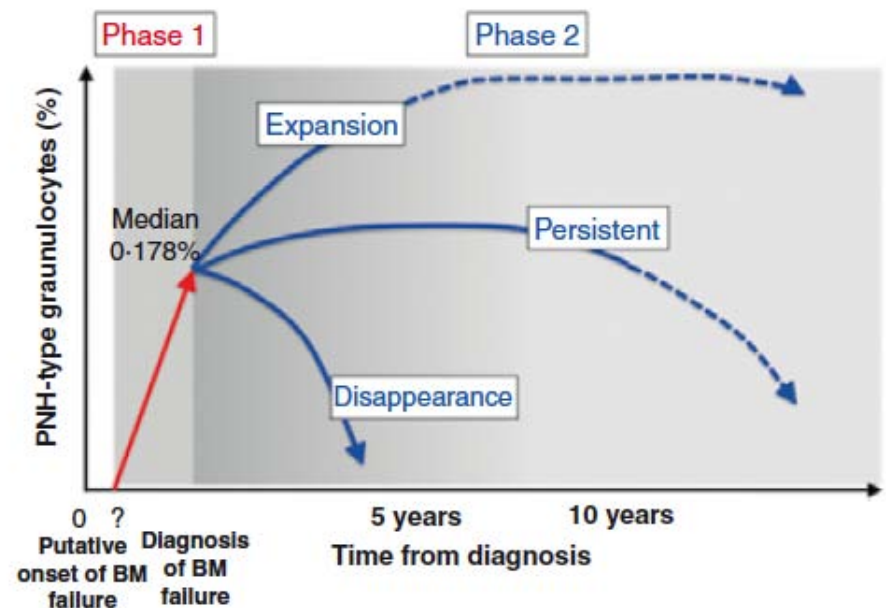
NO escape

1. Overlap tra AA, PNH, MDS



Ogawa S, Blood 2016

2. Evoluzione clonale

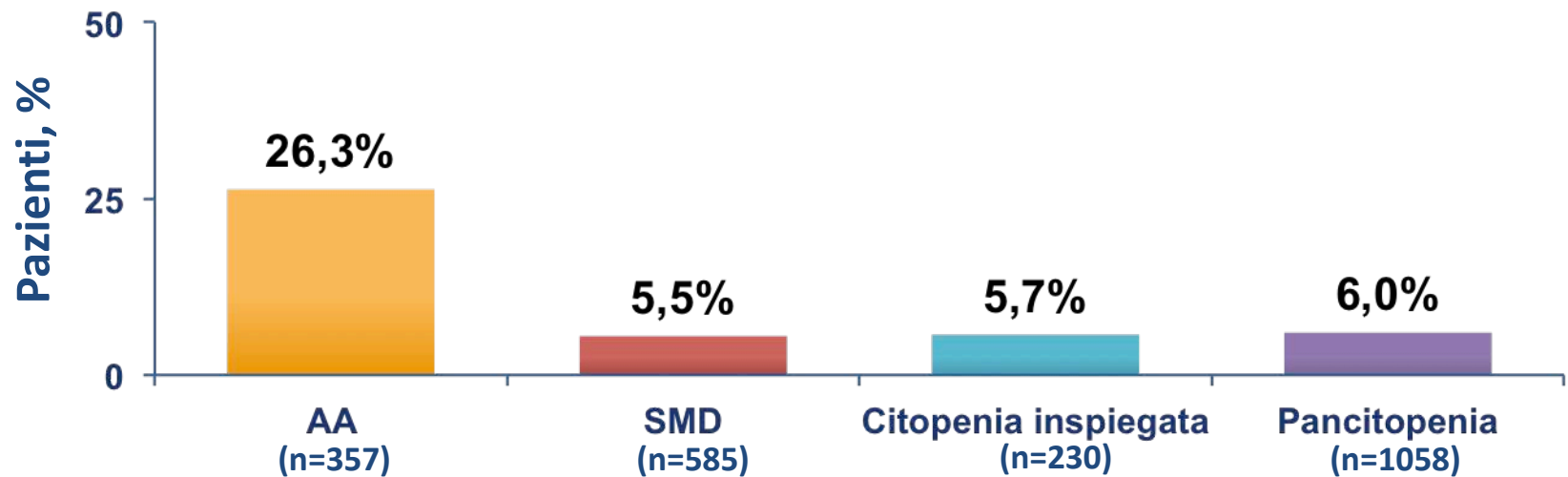


Sugimori C et al, BJH 2009

In pazienti affetti da insufficienza midollare (BMF) sono individuate cellule con deficit nelle proteine ancorate al GPI

- I pazienti con AA, sindrome mielodisplastica (MDS), o altri disturbi da BMF hanno spesso cloni con deficit nelle proteine ancorate al GPI individuabili^{1,2}

Percentuale di pazienti con cellule con deficit nelle proteine ancorate al GPI > 0,01% alla diagnosi³



Codice diagnostico ICD-9*

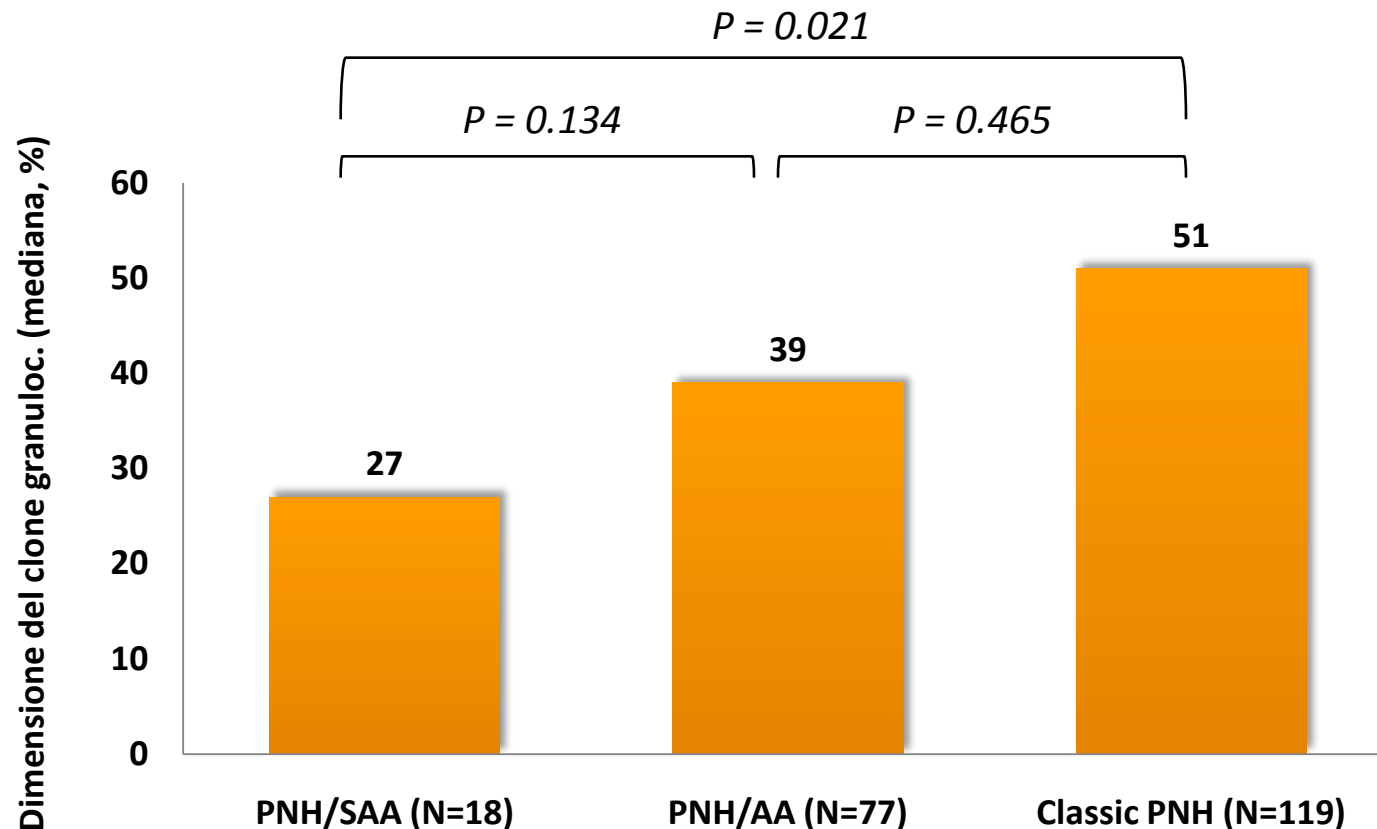
BMF, BoneMarrow Failure

ICD (International Classification of Diseases), Classificazione internazionale delle malattie.

* I pazienti potrebbero avere avuto più di 1 codice ICD-9 associato.

- Sugimori C, et al. *Br. J. Haematol.* 2009;147(1):102-112.
- Raza A, et al. *Cytometry B Clin. Cytom.* 2014;86(3):175-182.
- Movalia MK, et al. *Blood.* 2011;118(21):1033.

Pazienti con PNH ed AA: dimensione del clone

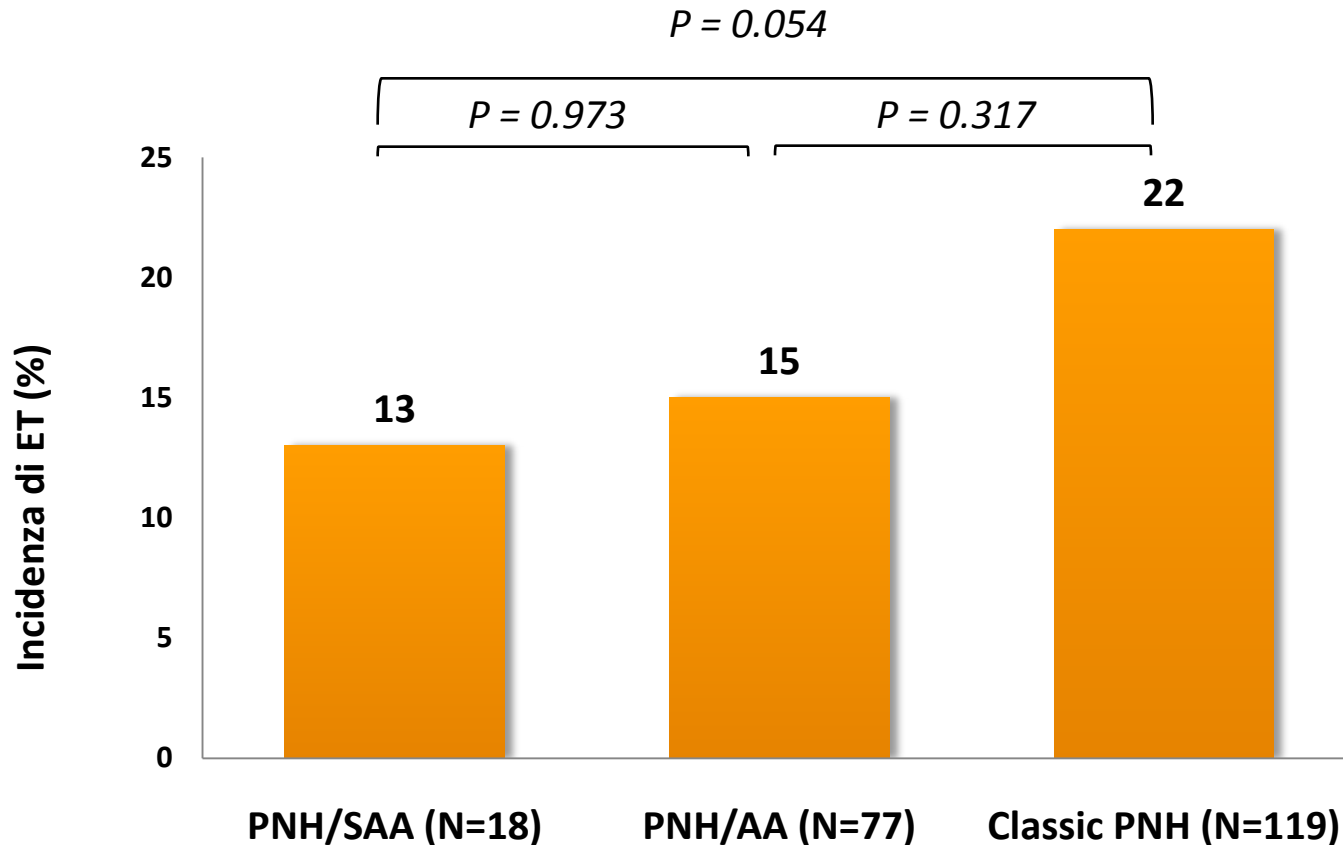


- I pazienti con PNH classica presentano dimensione del clone simile ai pazienti con PNH/AA, mentre è stata osservata una differenza significativa rispetto al sottogruppo con PNH/SAA

PNH/SAA: evidenza alla diagnosi di almeno 2 dei seguenti parametri: Hb ≤ 8 g/dL, ANC $< 0.5 \times 10^9/L$, PLTs $< 20 \times 10^9/L$.

PNH/AA: evidenza alla diagnosi di almeno 2 dei seguenti parametri: Hb ≤ 10 g/dL, ANC $0.5-1.5 \times 10^9/L$, PLTs $20-100 \times 10^9/L$.

L'incidenza di ET è simile nei sottogruppi di pazienti con PNH con o senza AA

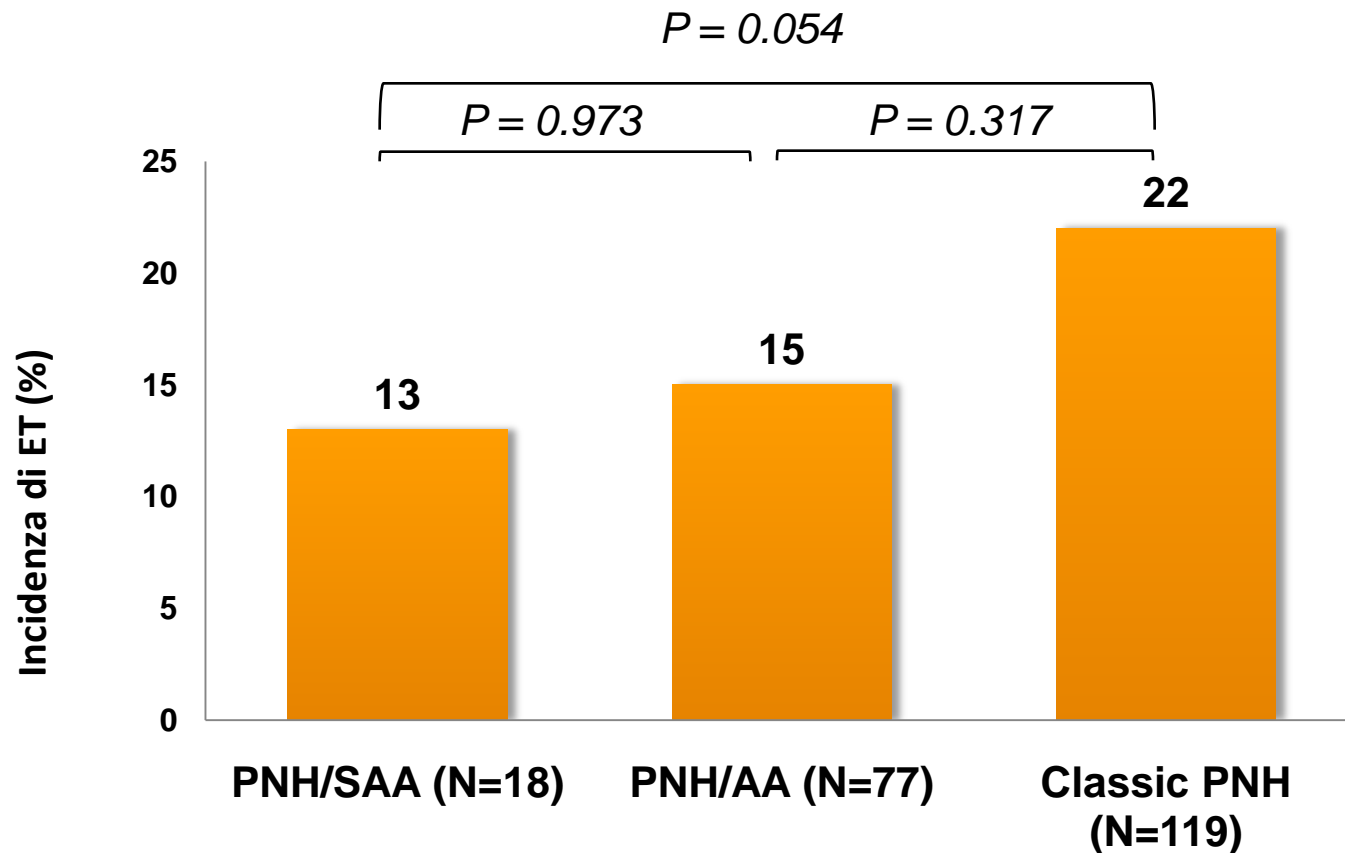


- Non è stata osservata alcuna differenza significativa nell'incidenza di TE tra i pazienti con PNH classica ed i sottogruppi PNH/SAA e PNH/AA

PNH/SAA: evidenza alla diagnosi di almeno 2 dei seguenti parametri: Hb ≤ 8 g/dL, ANC $< 0.5 \times 10^9/L$, PLTs $< 20 \times 10^9/L$.

PNH/AA: evidenza alla diagnosi di almeno 2 dei seguenti parametri: Hb ≤ 10 g/dL, ANC $0.5-1.5 \times 10^9/L$, PLTs $20-100 \times 10^9/L$.

L'incidenza di EVENTI TROMBOTICI è simile nei sottogruppi di pazienti con PNH con o senza AA



Non è stata osservata alcuna differenza significativa nell'incidenza di TE tra i pazienti con PNH classica ed i sottogruppi PNH/SAA e PNH/AA

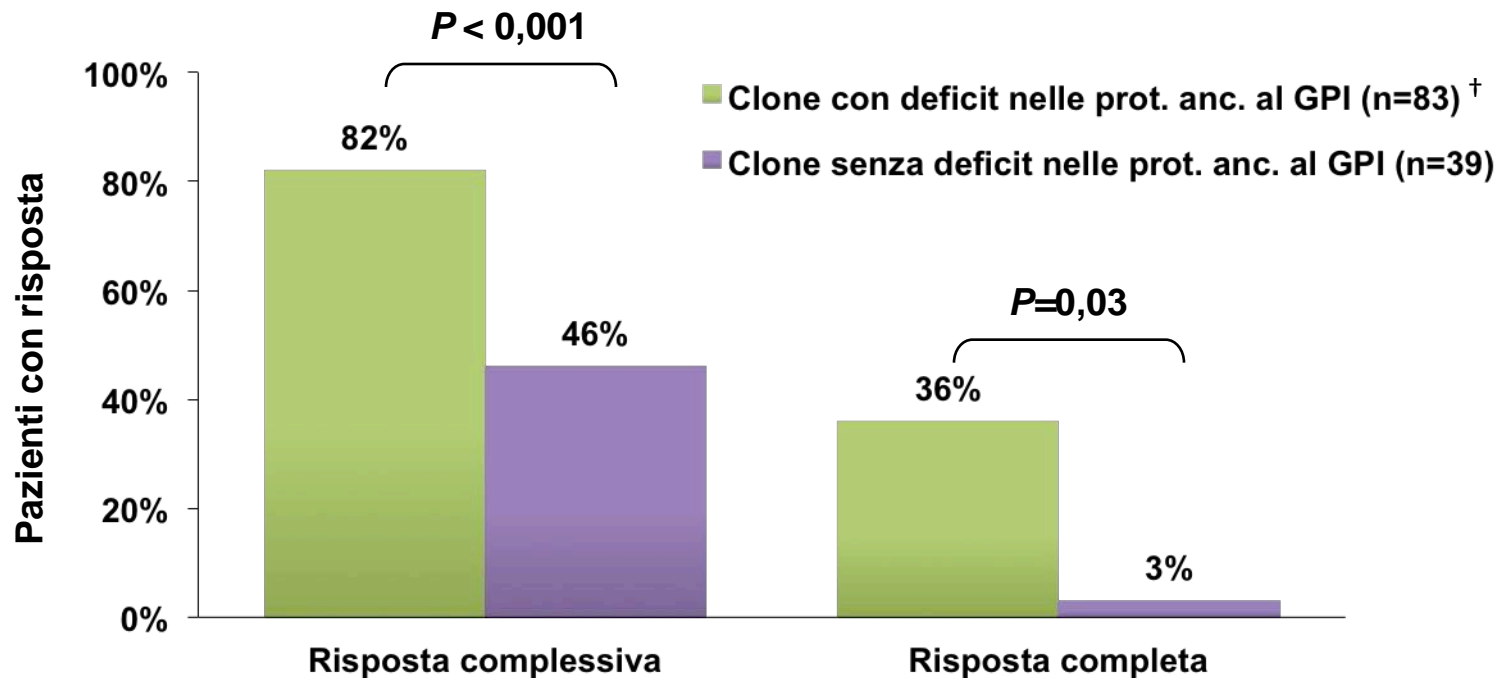
PNH/SAA: evidenza alla diagnosi di almeno 2 dei seguenti parametri: Hb ≤ 8 g/dL, ANC $< 0.5 \times 10^9/L$, PLTs $< 20 \times 10^9/L$.

PNH/AA: evidenza alla diagnosi di almeno 2 dei seguenti parametri: Hb ≤ 10 g/dL, ANC $0.5-1.5 \times 10^9/L$, PLTs $20-100 \times 10^9/L$.

I cloni con deficit nelle proteine ancorate al GPI potrebbero rappresentare un fattore predittivo di risposta alla terapia immunosoppressiva (IST)

- La presenza di cloni con deficit nelle proteine ancorate al GPI è stato l'unico significativo fattore predittivo della risposta alla IST in 122 pazienti con AA ($P < 0,01$) in un'analisi multivariata

Risposta* alla IST in pazienti con cloni con deficit nelle proteine ancorate al GPI



IST, terapia immunosoppressiva.

*Risposta completa (CR): emoglobina normale per l'età, conta dei neutrofili maggiore di $1,5 \times 10^9/L$, e conta piastrinica maggiore di $150 \times 10^9/L$; Risposta parziale (PR), indipendenza da trasfusioni e criteri per la classificazione come malattia grave in pazienti con AA grave non più soddisfatti. Risposta complessiva = CR + PR.

†Presenza di cloni con deficit nelle proteine ancorate al GPI definita tale quando $> 0,1\%$

Sugimori C, et al. *Blood*. 2006;107(4):1308-1314.

**STUDIO CITOFLUORIMETRICO DEI
CLONI GPI-CARENTI (CON FENOTIPO
EPN) IN CORSO DI ANEMIA APLASTICA**



Clonoteca EPN

Riassunto attività per regione



Lab Pievesestina

48% delle analisi per ricerca cloni EPN in Emilia Romagna

0/0 N. analisi / N. centri con inserimenti negli ultimi 12 mesi

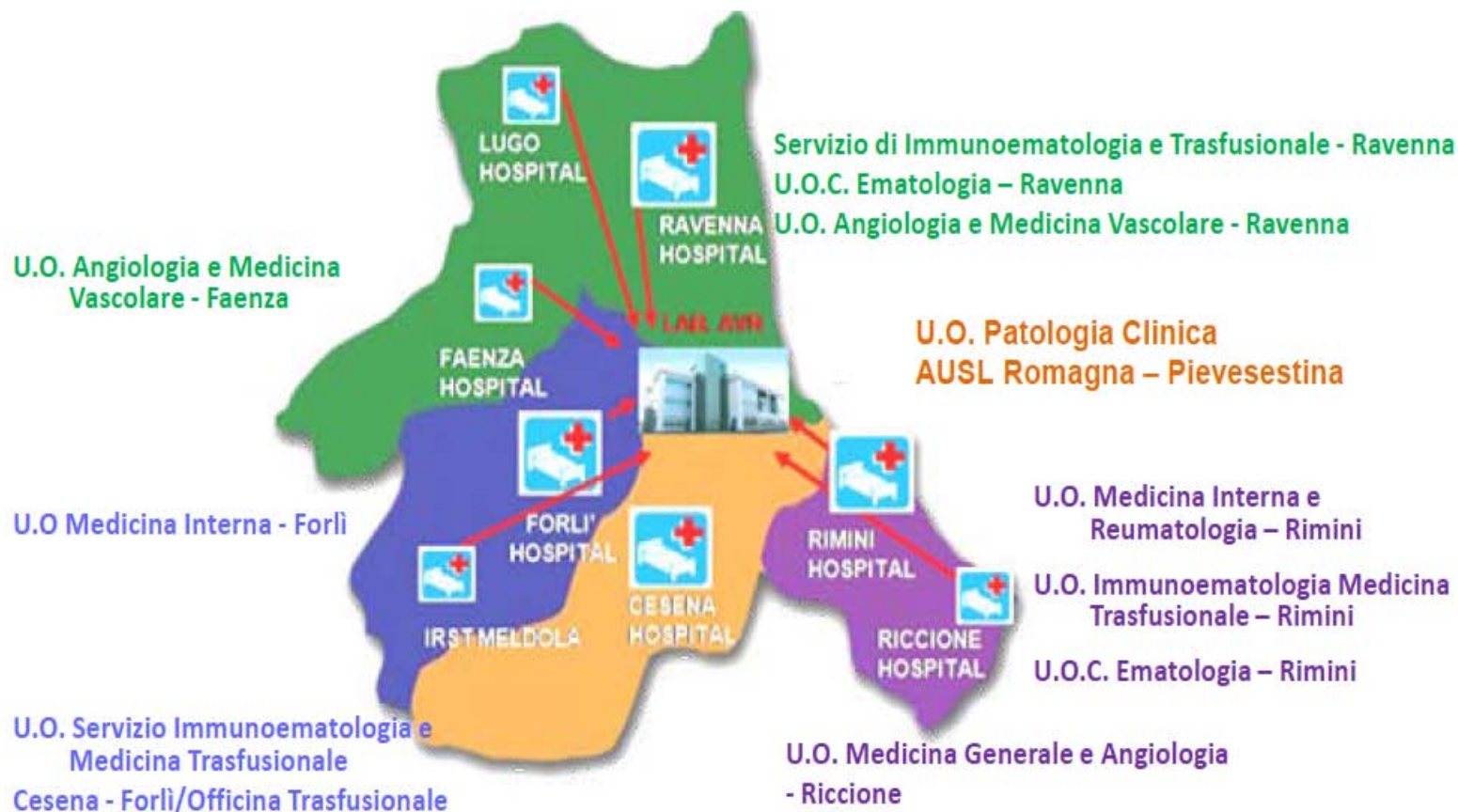
Emoglobinuria Parossistica Notturna: STUDIO AVR FLAER

Newsletter Settembre 2017

PROGETTO AUSL ROMAGNA PER LO SCREENING DI CLONI EPN IN PAZIENTI CON CITOPENIA DI DIVERSA NATURA (IDIOPATICA, IN CORSO DI ANEMIA IPO-PLASTICA O DI MIELODISPLASIA A BASSO GRADO IPSS), O ASSOCIATA A TROMBOSI VENOSA IDIOPATICA ATIPICA O CITOPENIA CON SEGNI DI EMOLISI INTRAVASCOLARE:

STUDIO DI VALIDAZIONE DI UN TEST RAPIDO IN CITOMETRIA A FLUSSO 3 COLORI

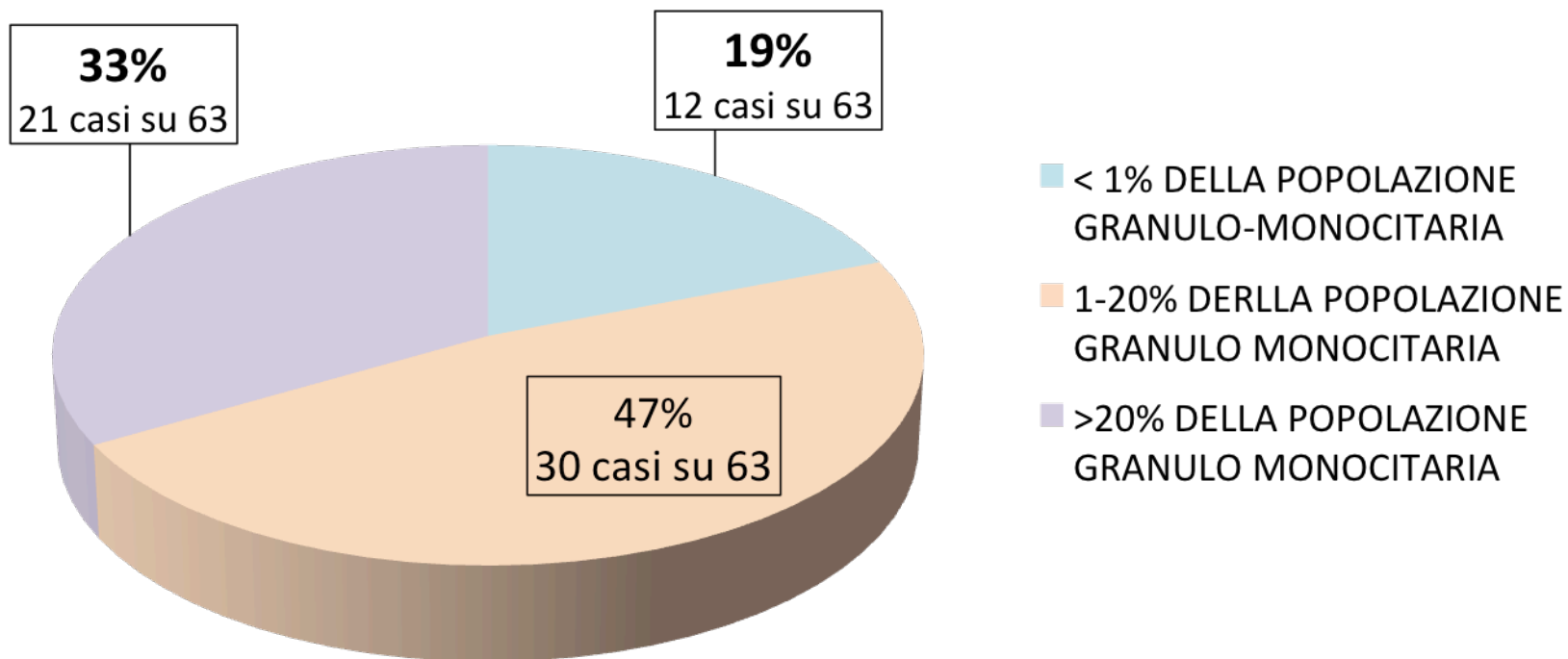
12 centri in AUSL Romagna



Obiettivi dello studio

- *Obiettivo primario*
 - **Determinare la prevalenza di cloni con fenotipo EPN** in pazienti con diagnosi nota o nuova di anemia aplastica afferenti ai Centri della Rete Ematologica Lombarda partecipanti allo studio
- *Obiettivi secondari*
 - **Determinare le linee cellulari coinvolte** nell'espressione del clone EPN.
 - Valutare **la dimensione dei cloni in termini di conta cellulare assoluta.**

IPOPLASIA- APLASIA midollare: studio cloni EPN: 5 COLORI



CASO CLINICO n. 1- EPN

P.A. F, anni 45

Anamnesi familiare: 1 sorella con TVP “idiopatica”

Anamnesi fisiologica, farmacologica, patologica remota:
nulla di rilevante da segnalare

Anamnesi patologica prossima:

- **Giugno 2007:** inviata in P.S. per dolore addominale più accentuato in regione periombelicale

E.O.: dolore alla palpazione superficiale e profonda, più evidente in regione mesogastrica; fegato palpabile all' arcata costale, non splenomegalia

Esami ematochimici e strumentali in urgenza:

Hb: 10.2 g/dl con MCV=91 μ^3 , **WBC: 3.830/mmc**

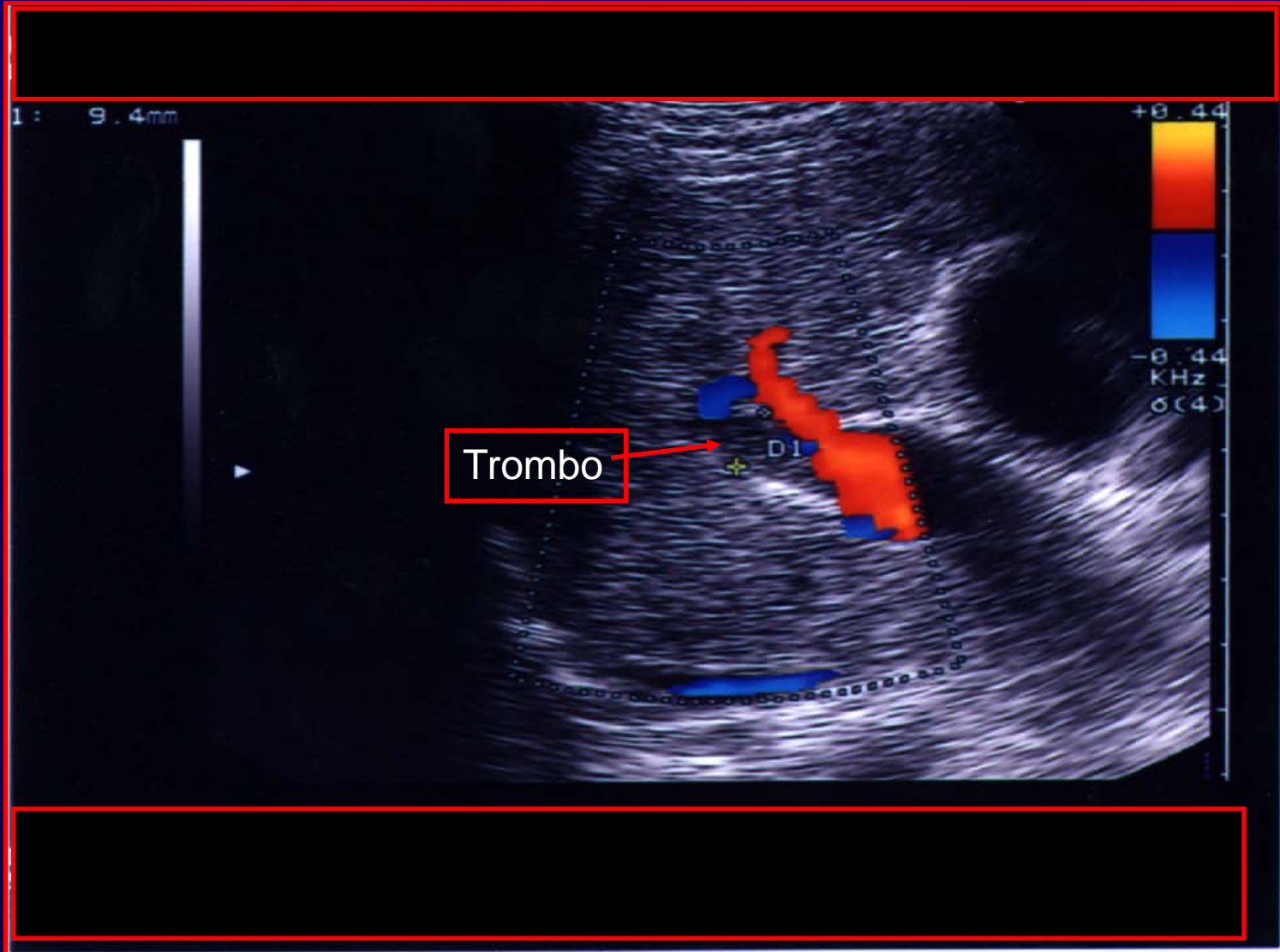
PLT: 83.000/mmc, Formula l.: N 30%, L 65%, M 5%, E 0%,

Indici di funzionalità epatica e renale: nella norma

PT, aPTT, fibrinogeno: nei limiti di norma; **D-dimero: elevato**

Rx torace: negativo: **RICOVERO IN REP MEDICINA**

Ecografia addome con color-doppler: trombosi ramo portale destro



Esami ematochimici e strumentali in degenza:

VES: 41 mm/h

Reticolociti: 27‰

Ferritina: 11 ng/ml

Eritropoietina sierica: 49 UI

LDH: 679 U/L (v.n. ≤ 245 U/L)

Test di Coombs diretto ed indiretto: negativi

Aptoglobina: < 30 mg/ml

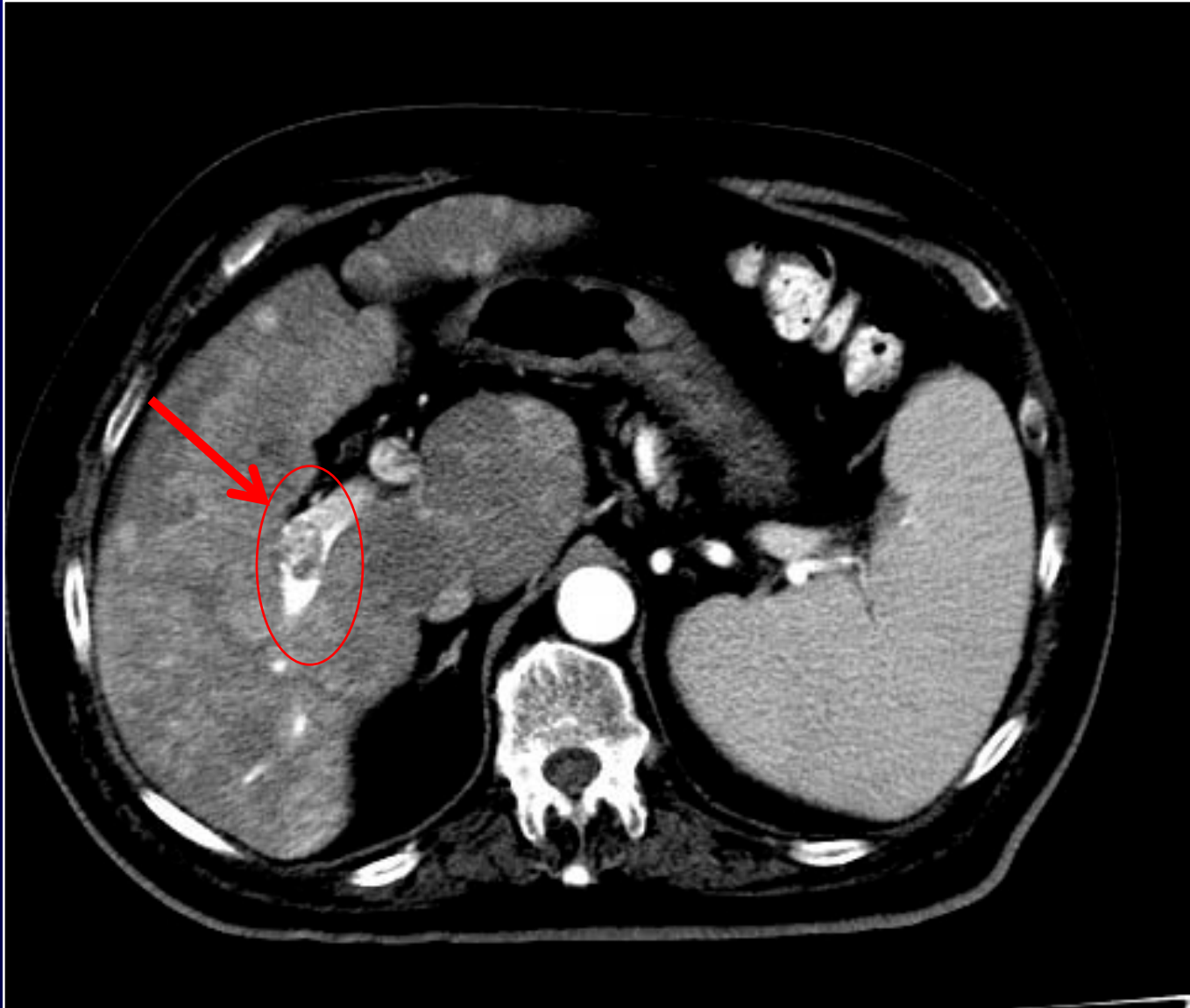
Bilirubina totale ed indiretta: nei limiti della norma

C3: consumato

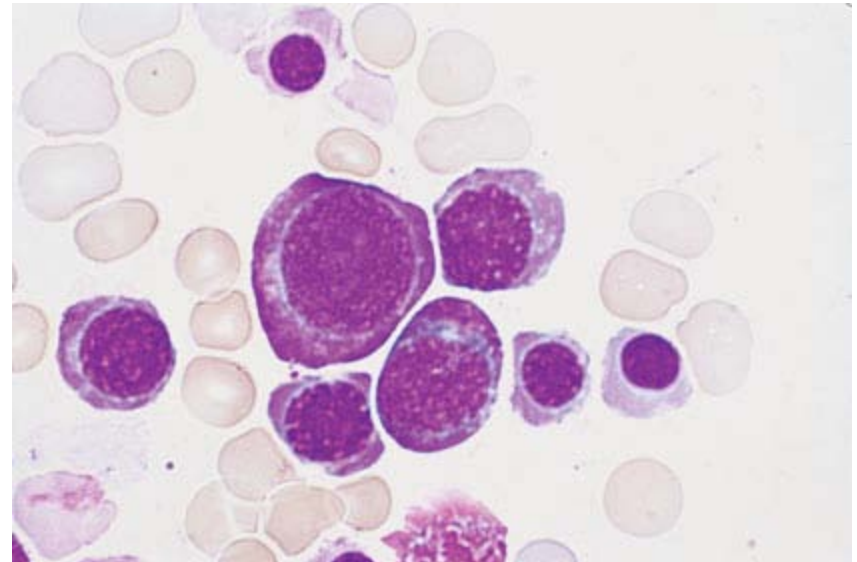
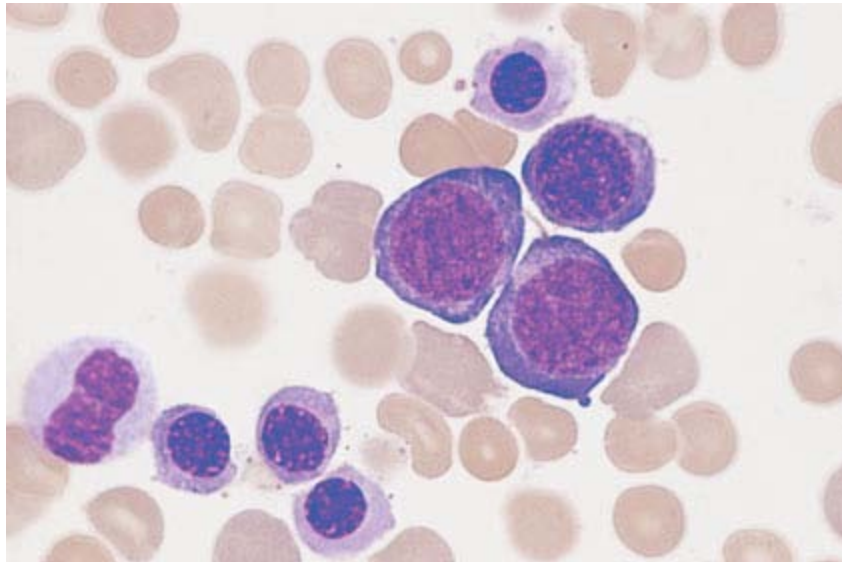
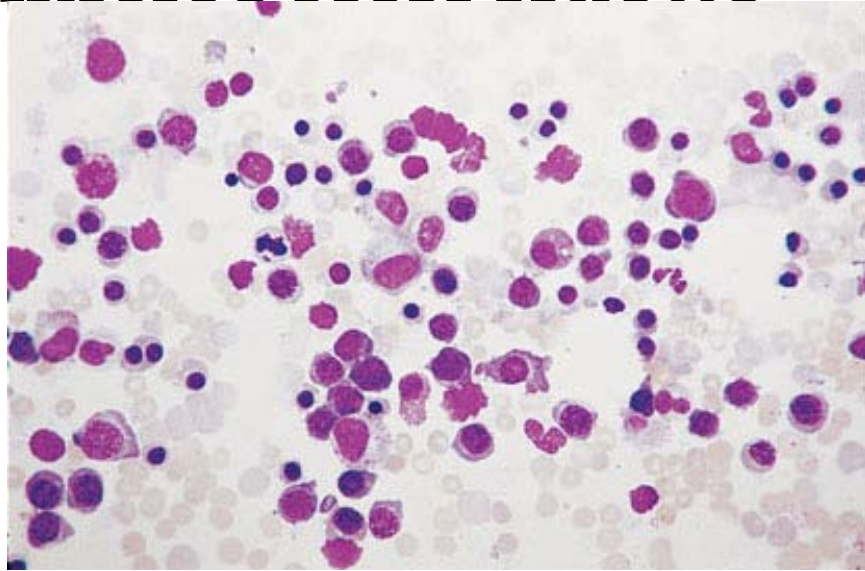
C4: nella norma

ANA, ENA, dsDNA, ASMA, APCA, AMA: assenti

**TC addome in fase arteriosa:
trombosi ramo portale destro**



**BM :iperplasia
eritroide, diseritropoiesi, megacario-
mielopoiesi ridotte**



Trombosi in sede atipica: iter diagnostico



Trombosi eredo-familiari

ATIII

Proteina C

Proteina S

PCA-R

FV Leiden

Protombina mutata

Iperomocisteinemia

Più rare:

↑ lipoproteina (a)

Ridotta fibrinolisi

(↑ di PAI-1 o ↓ di tPA)



Stati trombofilici acquisiti

Diabete mellito

Sindrome da APA (atc anti-fosfolipidi)

Neoplasie (pancreas, stomaco, polmone)

Neoplasie mieloproliferative Phneg

Sindrome nefrosica

MPD Jak2+

Protesi valvolari

Morbo di Crohn

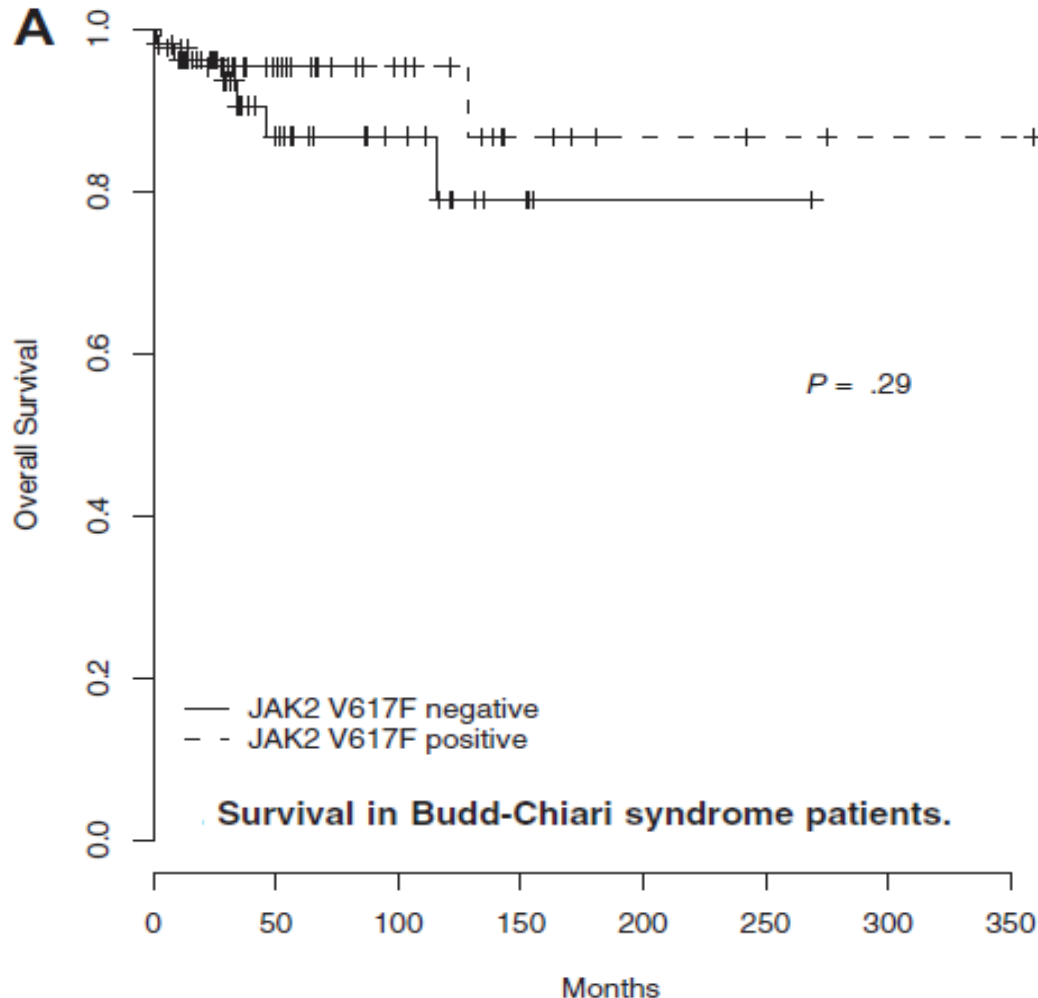
Sindrome di Behcet

Trombocitopenia da eparina

EPN

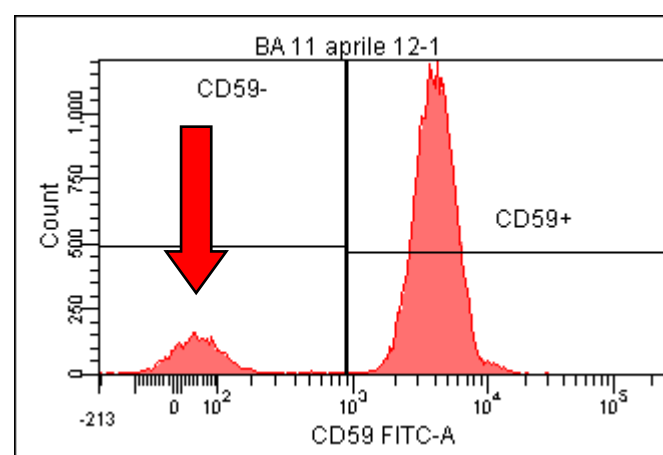
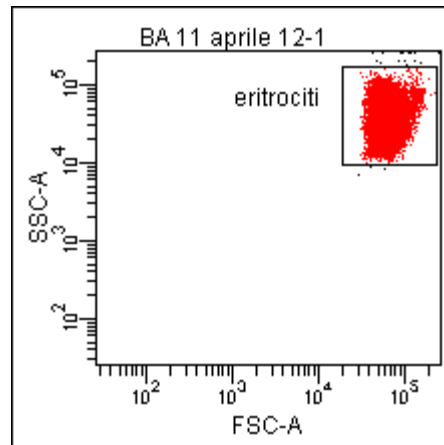
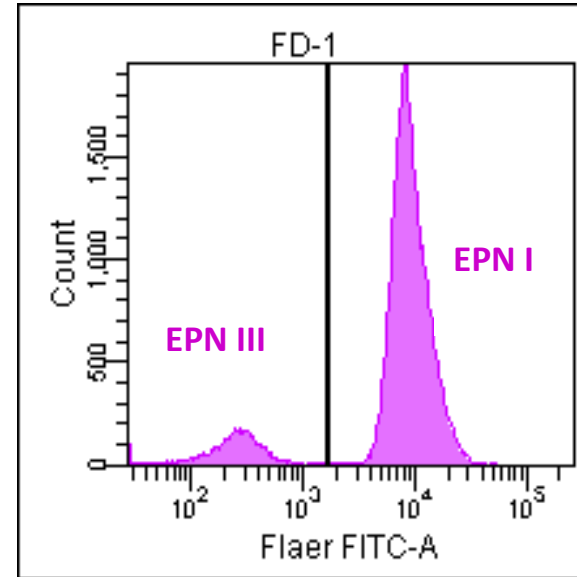
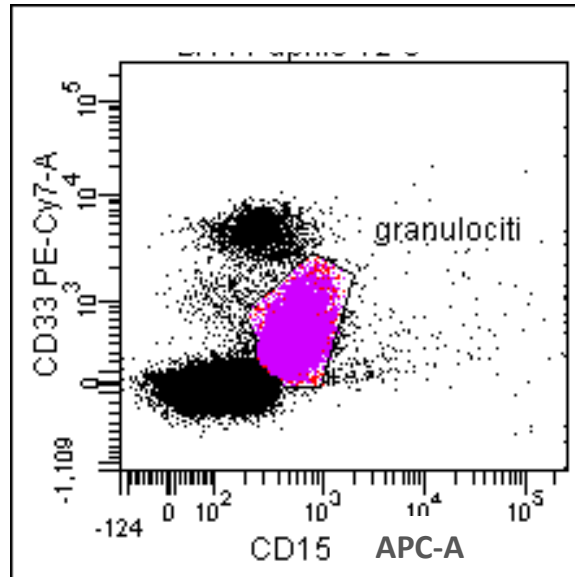
The impact of JAK2 and MPL mutations on diagnosis and prognosis of splanchnic vein thrombosis: a report on 241 cases

Jean-Jacques Kiladjian, Francisco Cervantes, Franck W. G. Leebeek, Christophe Marzac, Bruno Cassinat, Sylvie Chevret, Dominique Cazals-Hatem, Aurélie Plessier, Juan-Carlos Garcia-Pagan, Sarwa Darwish Murad, Sebastian Raffa, Harry L. A. Janssen, Claude Gardin, Sophie Cereja, Carole Tonetti, Stéphane Giraudier, Bertrand Condat, Nicole Casadevall, Pierre Fenaux and Dominique C. Valla



Jak 2 +:
34% PVT
45% BCS

Analisi multiparametrica citometrica (sp): FLAER sui granulociti e CD59 su globuli rossi



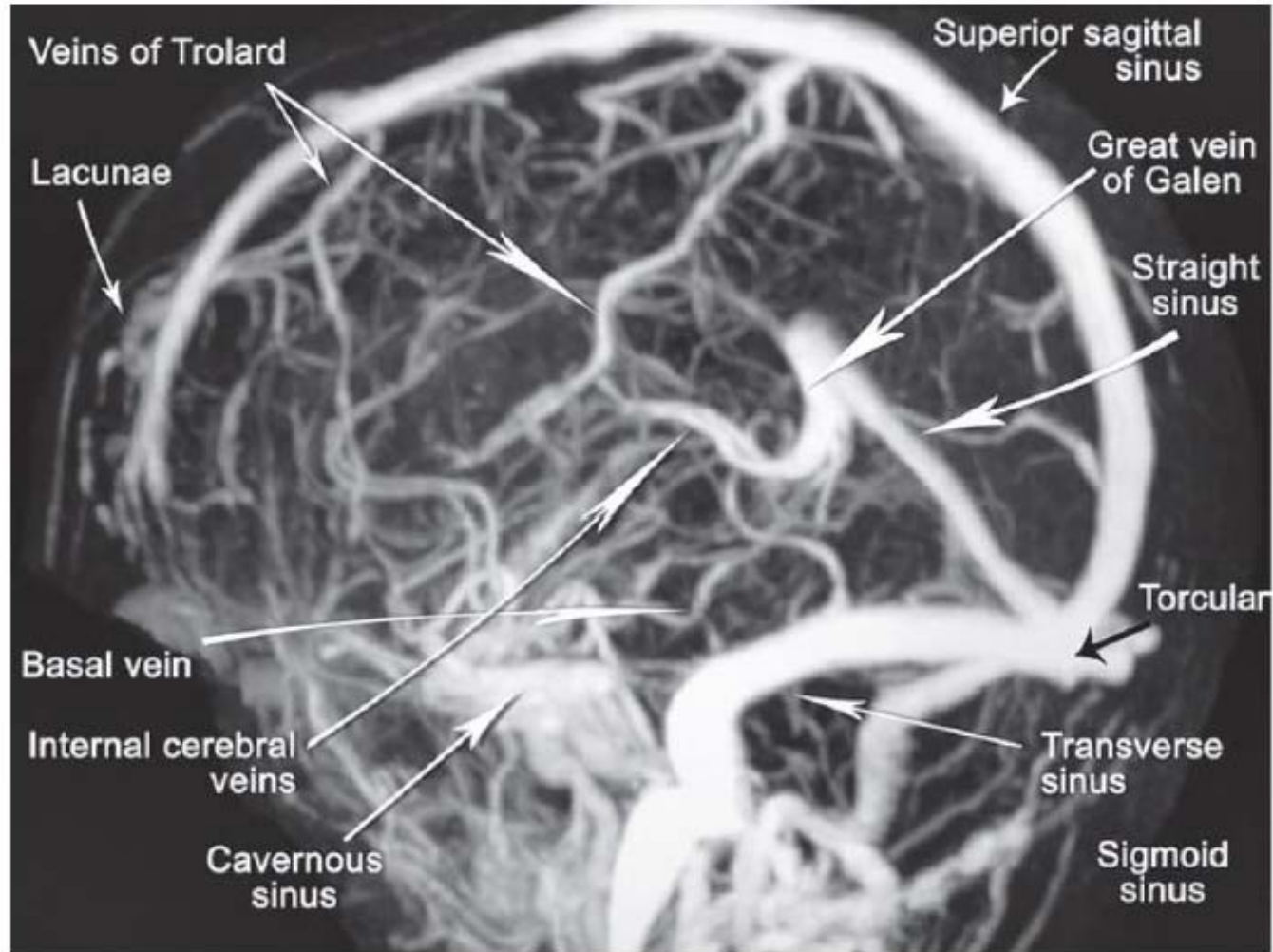
Caso 2

- **Anni 69. Esordio Marzo 2018 con dolore addominale:** esami: leucopenia (WBC 2.2), piastrinopenia (88), anemia (Hb 9.5) ,
insuff epatica: ipertransaminasemia. ittero, epatomegalia, splenomegalia, ascite, ipertensione portale, Insuff renale grado lieve.
- **Ricovero: S di Budd Chiari (trombosi 3 vene sovraepatiche).**
- **Citometria: clone EPN 80% in Gr neutrofili e monociti, 30% in GR.**

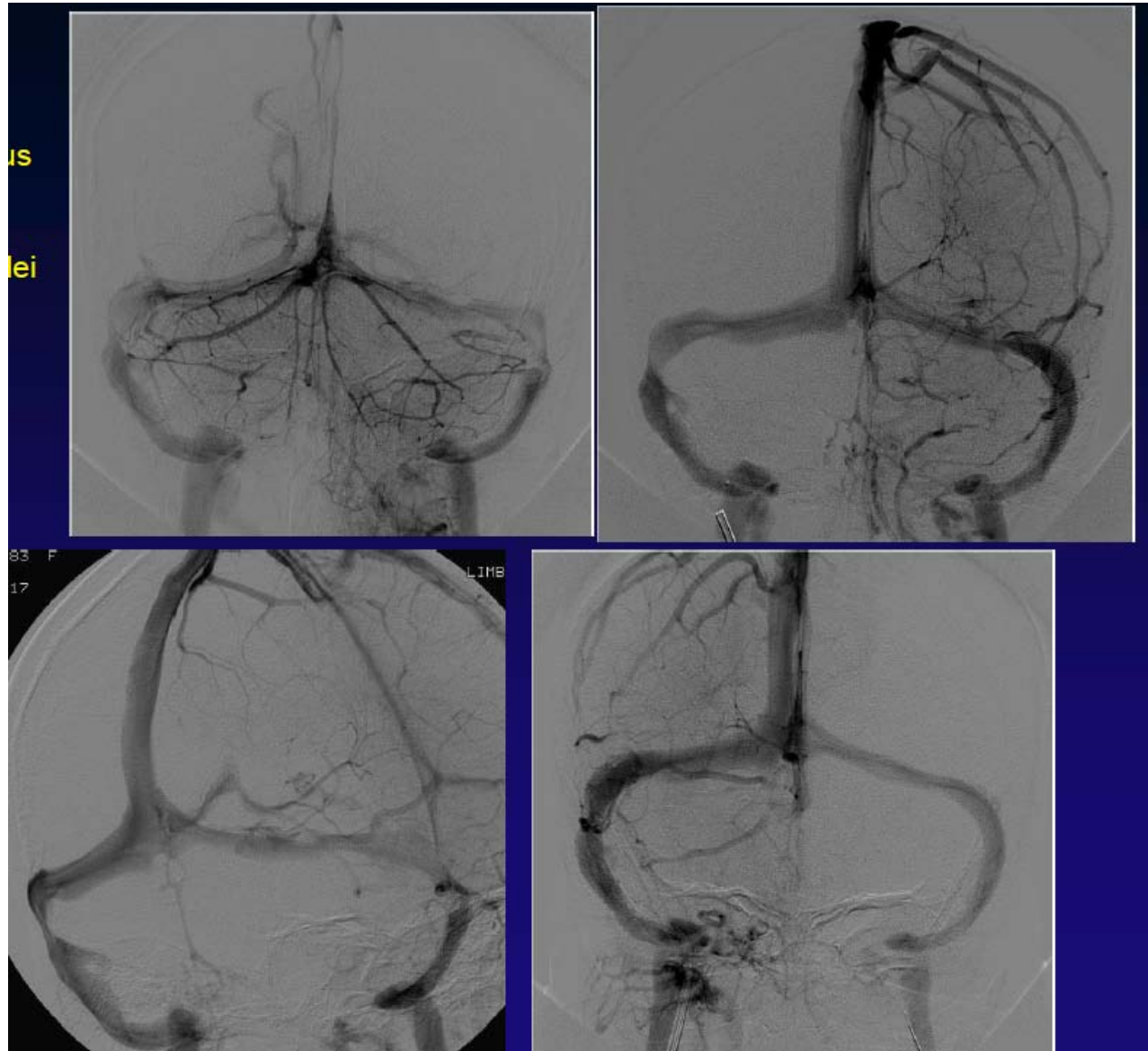
Caso 3

- Storia negli ultimi **24 mesi di oltre 10 episodi trombotici** (vene dei seni cerebrali, splancnici multipli, TVP, TIA, s.coronarica)
- Screening trombofilico negativo
- **Consulenza ematologica Marzo 2016: clone EPN 85%,**
- **Aprile 2016: inizio terapia con eculizumab**
- Nessun evento trombotico dal 2016 ad oggi

VENE CEREBRALI



Stenting bilaterale seni trasversi e sigmoidei



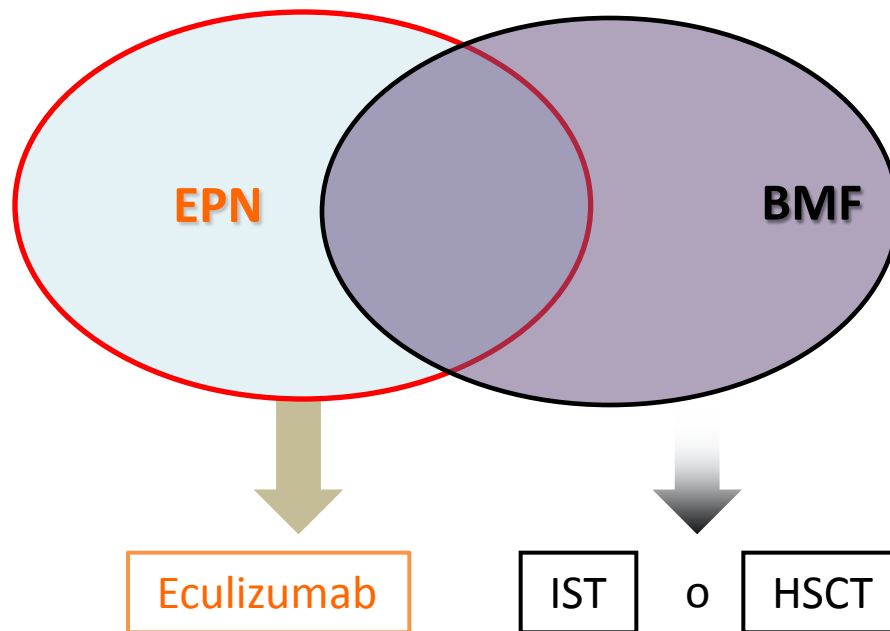
Caso 4

- Esordio aplasia severa e clone EPN 5-45 % in 9 mesi : ATG CyA : no risposta ; ATG cavallo-CyA
- No risposta_ Cy A e boli endxan e VCR no risposta ; eculizumab : no risposta sulla serie rossa
- 2 anni dopo: clone 90%, AA non severa: N 700/ul, Pst 40.000/mmc, Hb 8 g/dl: ottima risposta ematologica.

Gestione di pazienti con EPN e insufficienza midollare senza emolisi

- In pazienti con emolisi minima, gli interventi devono focalizzarsi sulla disfunzione midollare sottostante¹

Algoritmo di trattamento²⁻⁴



FACTORS THAT MAY INFLUENCE THE START OF TREATMENT

- **Size of PNH red cell population**
- **Prevalence of PNH III *versus* PNH II**
- **LDH level above 2500-3000**
- **Thrombotic episodes**
- **Syntoms related to acute hemolysis**
- **Transfusion dependency**
- **Renal failure, pulmonary hypertension, severe dysphagia**

Conclusions

- **Small and large PNH clones may be detected in aplastic- hypoplastic form, and less frequently in low-risk MDS.**
- **Several “PNH-like” signs and symptoms are independent from the clone size**
- **Monocyte (neutrophil) only may be detected**
- **Severe AA should be treated with IS therapy (Ecu may be used in later phases)**
- **Consensus on “small clone” definition is lacking**
 - **Clinical (e.g. absence of hemolysis)**
 - **Technical (<10%, <1%, <0.1% according to the detection limit)**
- **Clinical significance of small clones to be determined**