Webinars
Constitutional thrombocytopenia
EuroBloodNet

Thrombocytopenia due to transcription factor defects and leukemia predisposition

Paul Saultier, MD PhD
APHM – Pediatric Hematology, Immunology, and Oncology
Reference Center for Inherited Platelet Disorders (CRPP)
Aix Marseille University – Inserm 1263 – C2VN
ERN-EuroBloodNet
Marseille – France
February 21, 2024

Co-funded by
the Health Programme
of the European Union

CRPP
MHEMO

European
Reference
Network
for rare or low prevalence
complex diseases
Network
Hematological
Diseases (ERN EuroBloodNet)

Conflicts of interest

- Investigator in studies sponsored by Biomarin, Celgene, Gilead, Sanofi, and Takeda

- Support for attending scientific meetings and educational events: Baxalta-Shire, Global Blood Therapeutics, Novo Nordisk, Pfizer, Octapharma, Roche, Servier, and Sobi

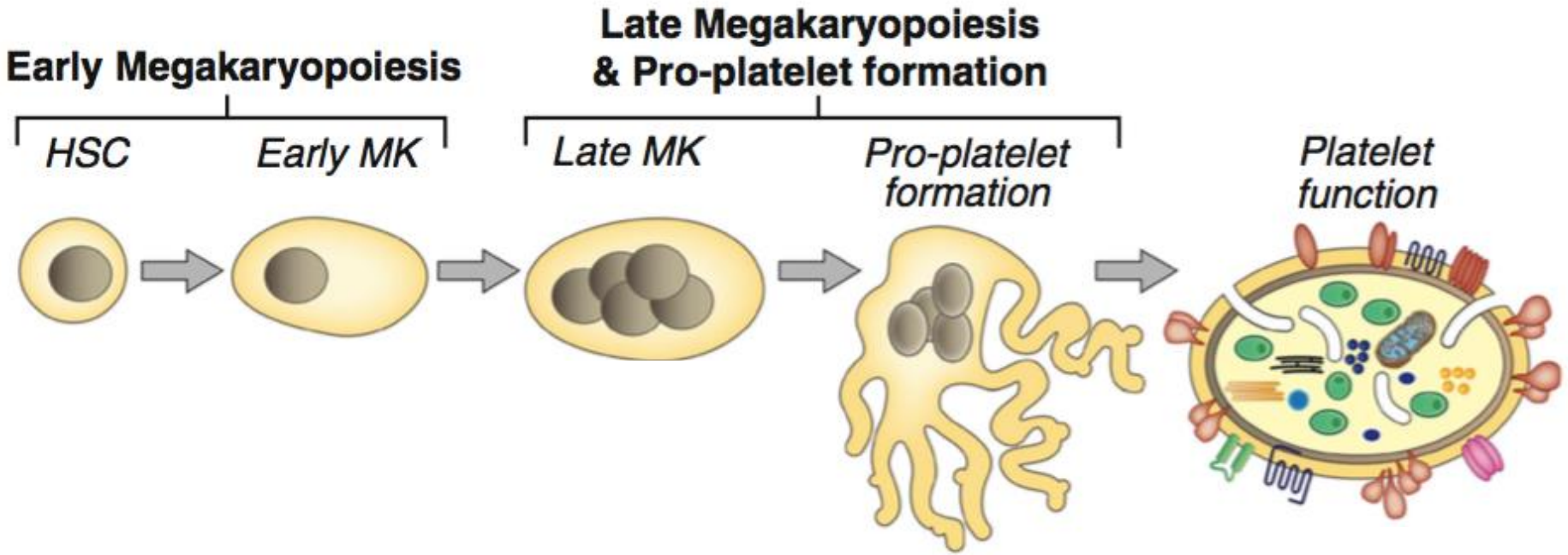
- Honoraria (consultant / advisory boards) from Octapharma, Novartis, Servier, Clinigen, Sobi, and Jazz.

Constitutional thrombocytopenias associated with leukemia predisposition

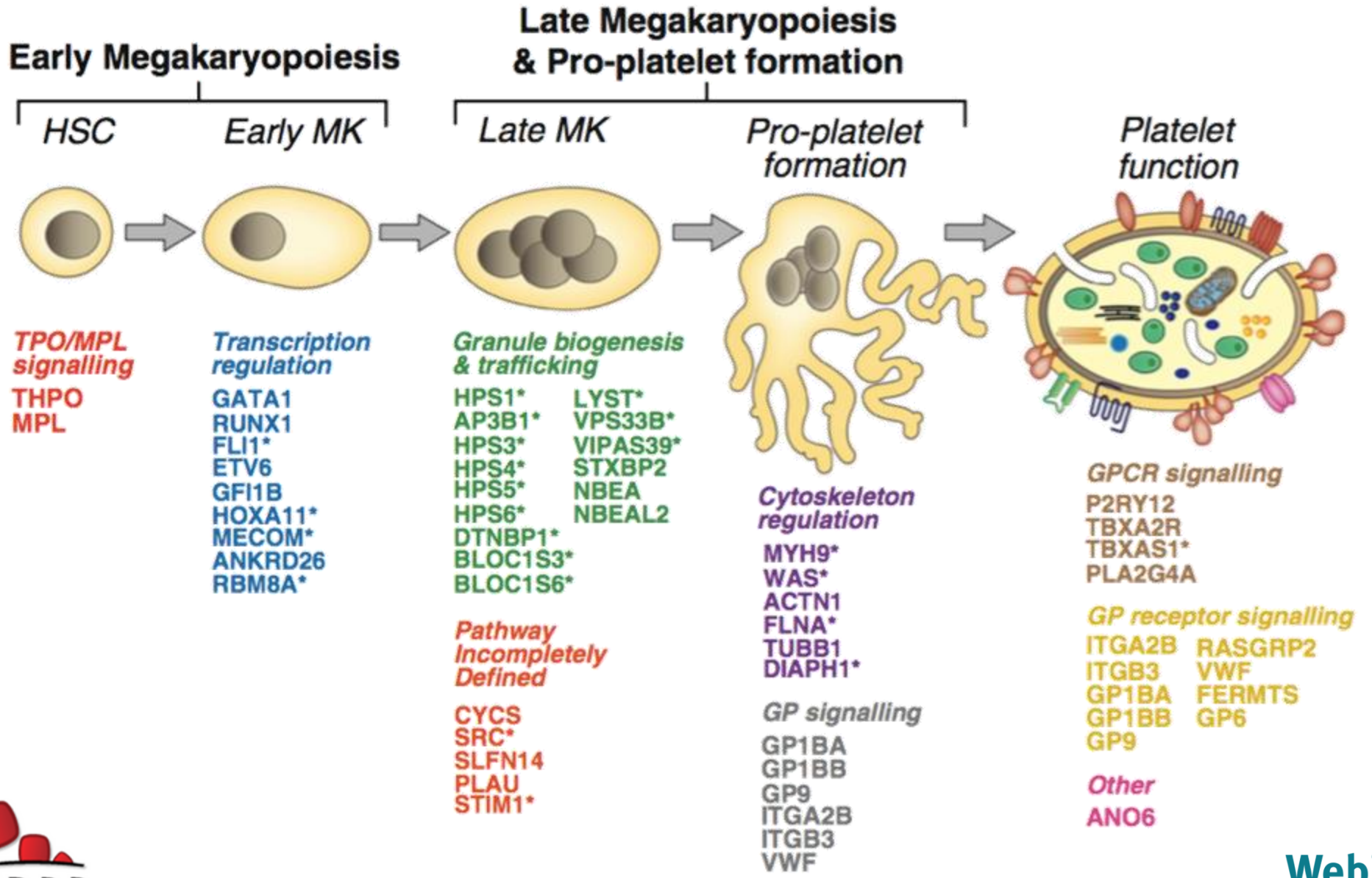
1. **Epidemiology and principal characteristics**
2. **How to diagnose these conditions?**

   The pediatrician / hospitalist / hemostasis specialist perspective

   The Hem/Onc perspective
3. **How to manage the bleeding risk?**
4. **Which hematological follow-up?**



Lentaigne et al. Blood. 2016;127(23):2814–23.



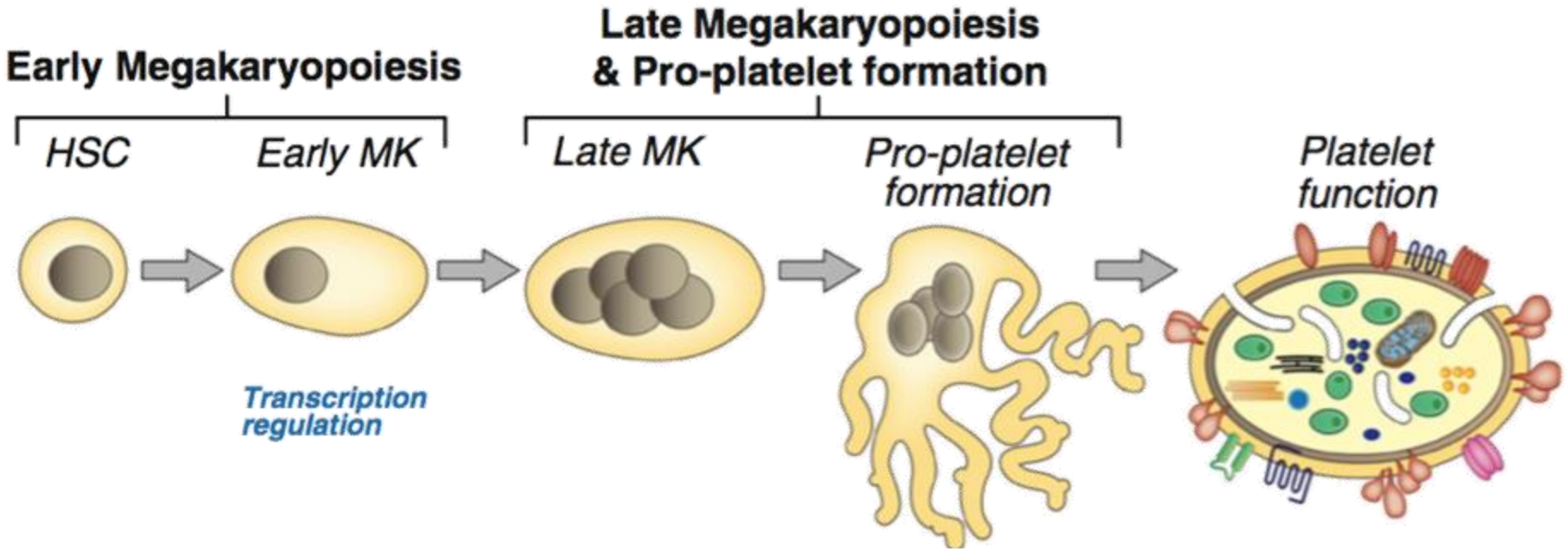
Lentaigne et al. Blood. 2016;127(23):2814–23.

European Reference Network for rare or low prevalence complex diseases

Network Hematological Diseases (ERN EuroBloodNet)

CRPP MHEMO

Webinars EuroBloodNet



RUNX1

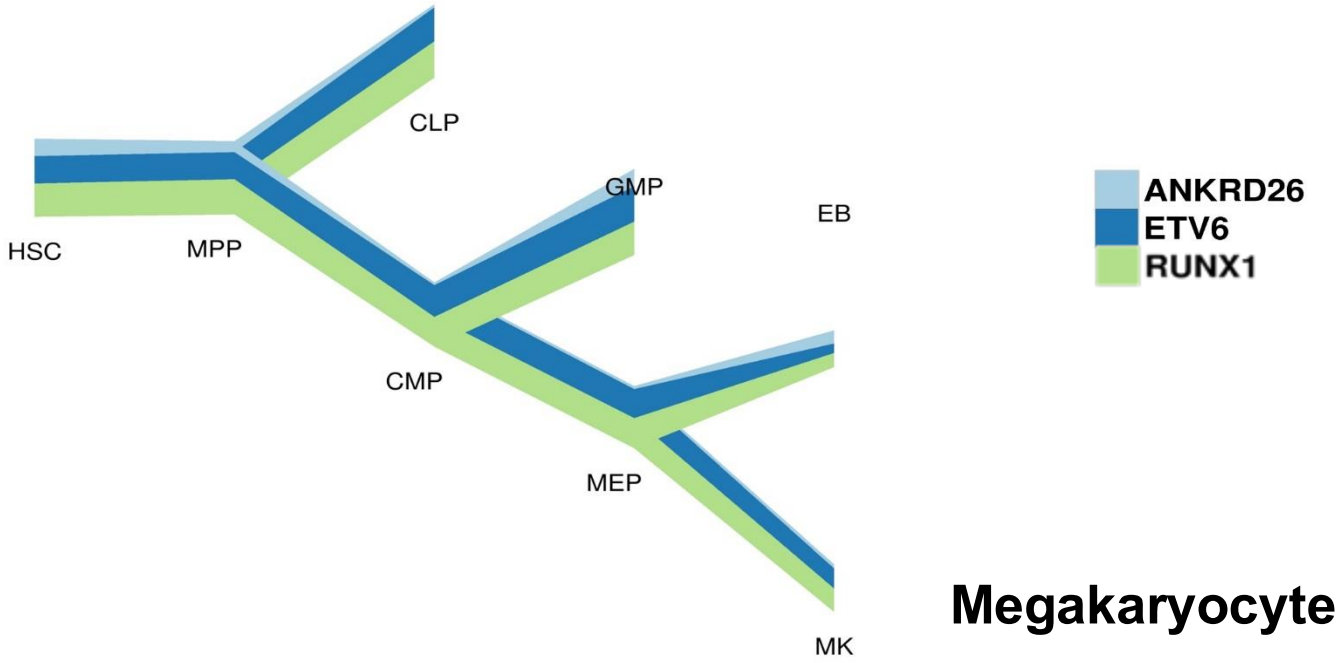
ETV6

ANKRD26

Lentaigne et al. Blood. 2016;127(23):2814–23.

Expression in the hematopoietic lineage

Hematopoietic stem cell



Megakaryocyte

RUNX1, ANKRD26, and ETV6 are crucial regulators of different stages of hematopoiesis

Riverplot generated using Blueprint Progenitors data and the online tool available at http://blueprint.haem.cam.ac.uk. (Chen L et al.. Science. 2014 26;345(6204):1251033)

Constitutional thrombocytopenias associated with leukemia predisposition

Epidemiology and principal characteristics

European Reference Network for rare or low prevalence complex diseases

Network Hematological Diseases (ERN EuroBloodNet)

CRPP MHEMO

Webinars EuroBloodNet

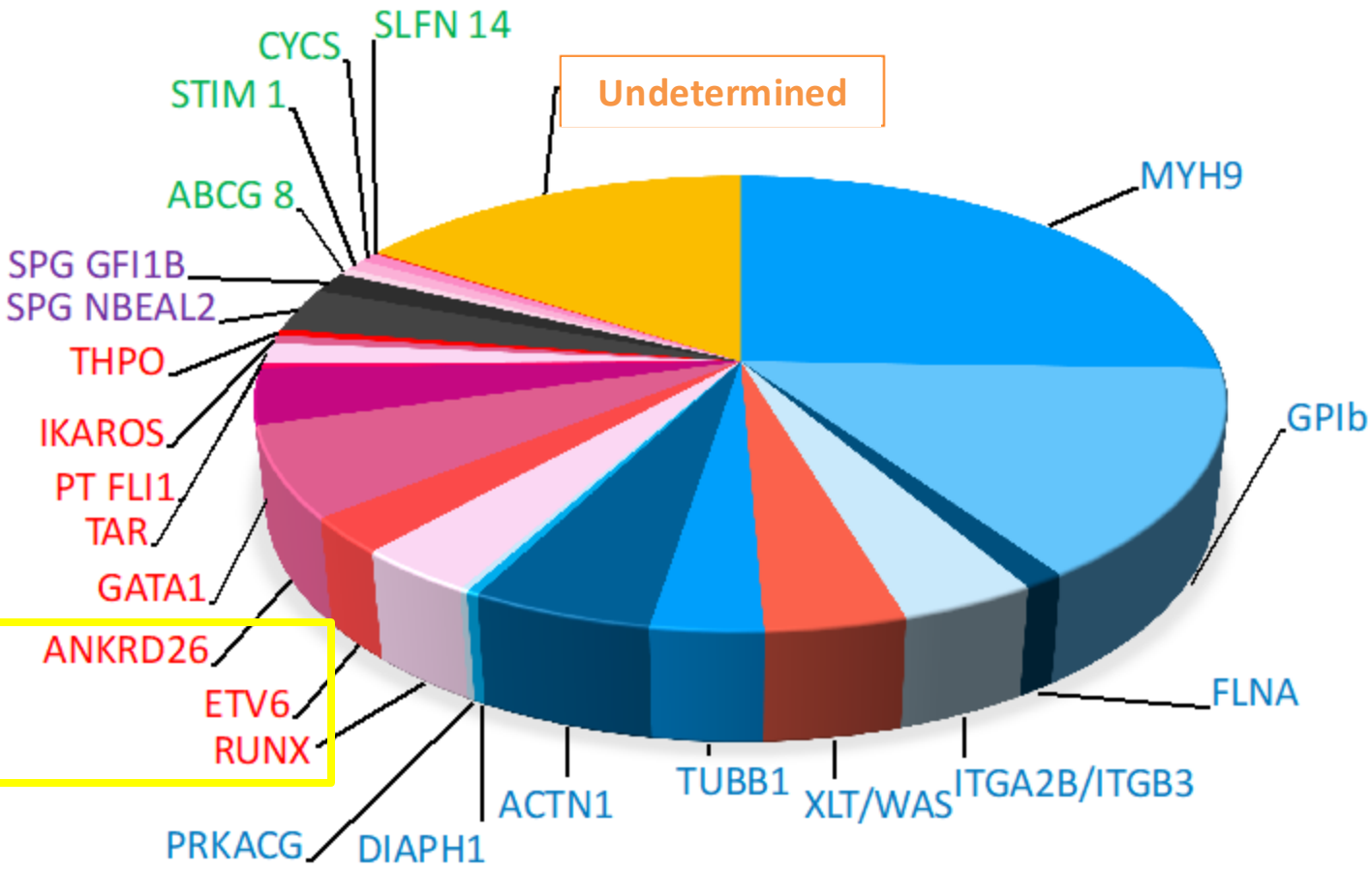
Epidemiology

n=859

Grey platelet
~5%

Transcription
≈ 15%

Leukemia predisposition ≈ 12%



Cytoskeleton ≈ 60%

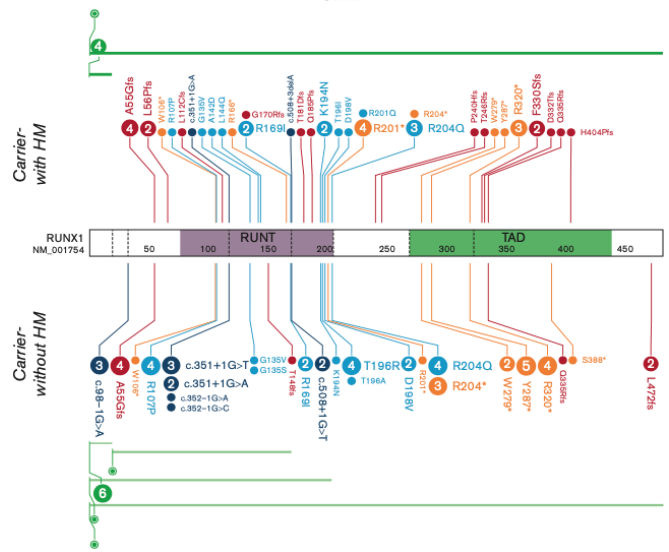
C. Falaise – APHM, Marseille, France– Unpublished data – 2023

European Reference Network for rare or low prevalence complex diseases
Network Hematological Diseases (ERN EuroBloodNet)
CRPP MHEMO
Webinars EuroBloodNet

RUNX1 thrombocytopenia - FPD/MM

- **Autosomal dominant**

No clear genotype-phenotype relationship



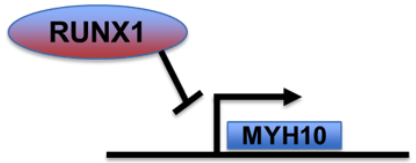
- **Thrombocytopenia**

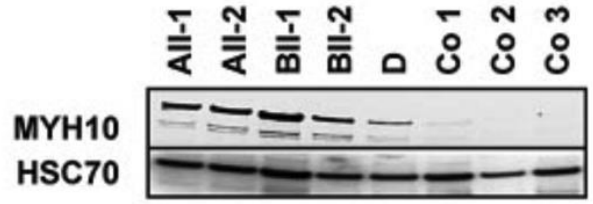
Mild to moderate, normal platelet volume

Platelet dysfunction (aspirin-like defect, α/δ-granule deficiency)

Platelet biomarker: MYH10

Extra-hematologic features: asthma and eczema





- **Hematological malignancies**

Myeloid +++ MDS, AML, T-ALL, MPAL , T-NHL, CLL, HCL

Penetrance ~40% (variable according to families (10-100%)

European Reference Network for rare or low prevalence complex diseases

Network Hematological Diseases (ERN EuroBloodNet)

CRPP MHEMO

Godley. Semin Hematol. 2014;51(4):306–321.
Song et al. Nat Genet. 1999;23(2):166–175.
Antony-Debré et al. Leukemia. 2016;30(4):999–1002.
Antony-Debré et al. Blood. 2012;120(13):2719–2722.
Cunningham L et al. Blood. 2023;142(25):2146-2158.

Webinars EuroBloodNet

ANKRD26 thrombocytopenia (THC2)

- **Relatively frequent among constitutional thrombocytopenias (≈10%)**

- **Autosomal dominant**

Variants affect the 5'UTR region (promoter)

Alter TPO signaling

- **Thrombocytopenia**

Mild to severe (10-100 G/L)

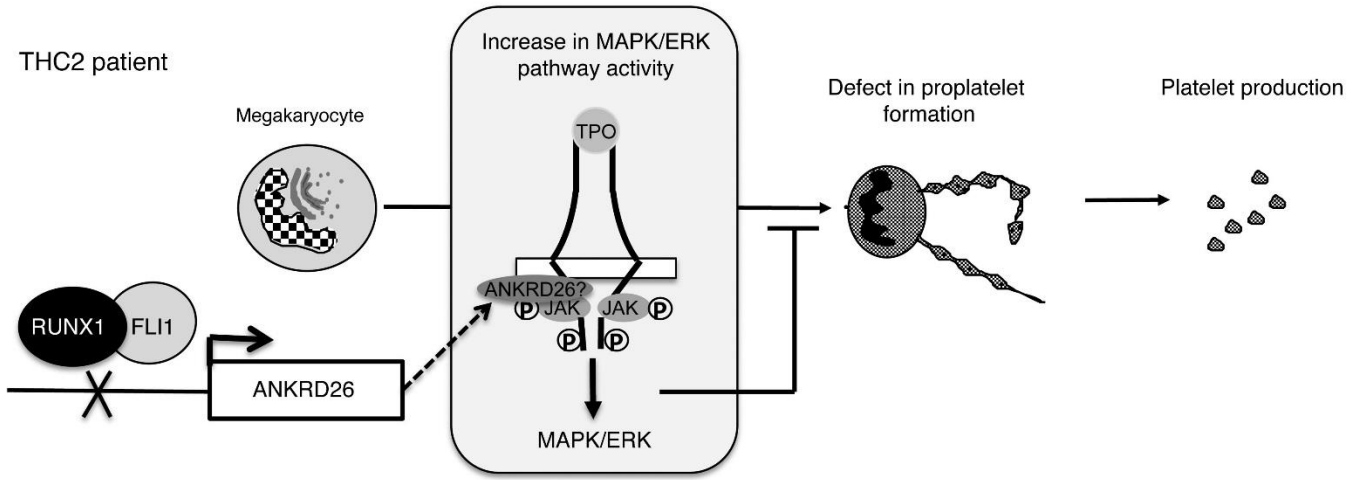
Normal platelet volume

Absent/moderate bleeding

- **Hematological malignancies**

Myeloid +++: MDS, AML, CML

Penetrance ≈10%



Noris et al. Blood. 2011;117(24):6673–6680.
Noris Blood. 2013;122(11):1987–1989.
Bluteau D et al. J Clin Invest. 2014;124(2):580-91.
Boutroux et al. Eur J Pediatr. 2015;174(10):1399–1403.
Basso-Valentina F et al. Haematologica. 2023 (ahead of print)

European Reference Network for rare or low prevalence complex diseases

Network Hematological Diseases (ERN EuroBloodNet)

CRPP MHEMO

Webinars EuroBloodNet

ETV6 thrombocytopenia (THC5)

- **Autosomal dominant**

Most variants affect the ETS DNA-binding domain

- **Thrombocytopenia**

Mild to moderate, normal platelet volume

Absent to moderate hemorrhagic syndrome

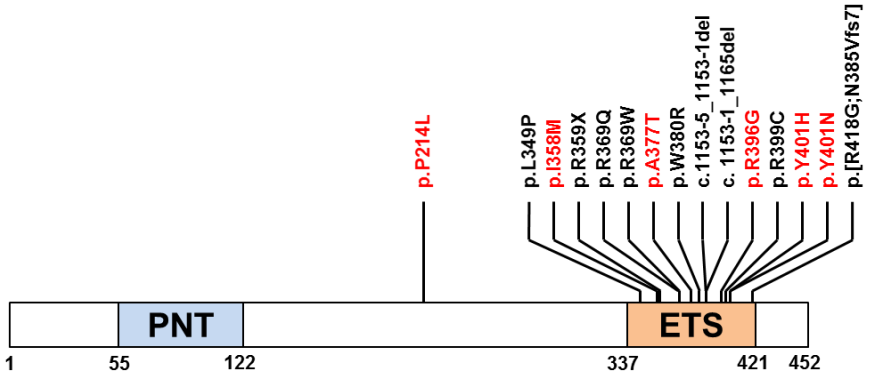
Variable platelet function defect

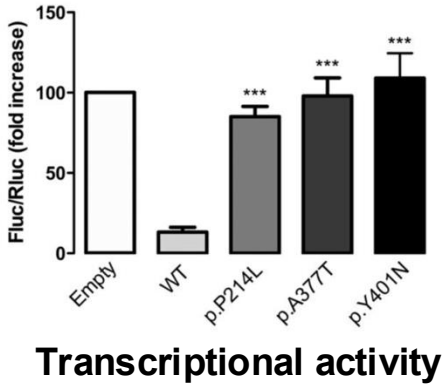
Increased ETV6 expression in platelets

- **Hematological malignancies :**

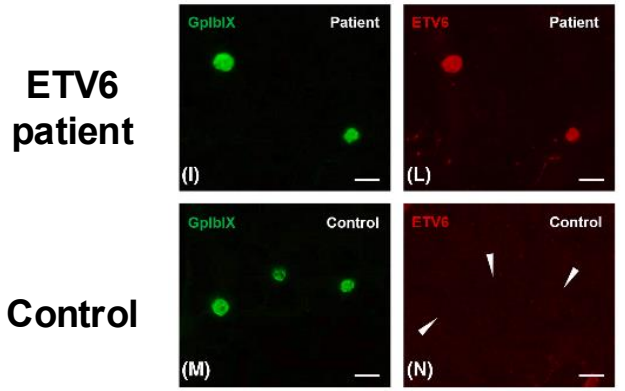
Childhood ALL +++, MDS, AML, MM, CMML

15/18 families, penetrance ≈15-25%





Transcriptional activity



Zhang et al. Nat Genet. 2015;47(2):180–185.
Noetzli et al. Nat Genet. 2015;47(5):535–538.
Poggi et al. Haematologica. 2017;102(2):282–294.
Zaninetti C et al. Br J Haematol. 2024;204(2):710-714.

European Reference Network for rare or low prevalence complex diseases

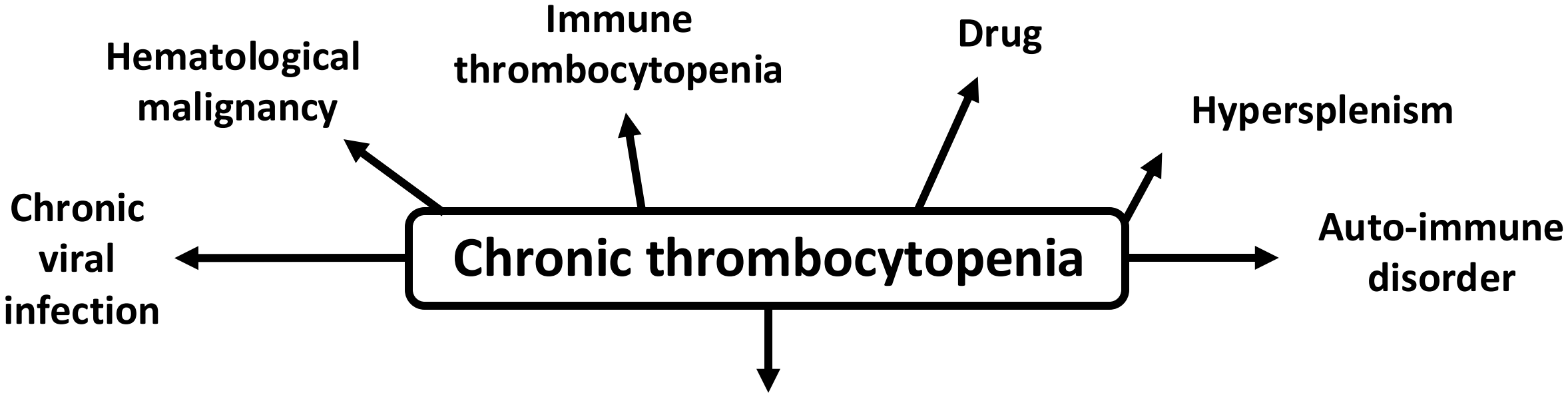
Network Hematological Diseases (ERN EuroBloodNet)

CRPP MHEMO

Webinars EuroBloodNet

How to diagnose constitutional thrombocytopenias associated with leukemia predisposition?

The pediatrician / hospitalist / hemostasis specialist perspective



Constitutional thrombocytopenia…

Family history of thrombocytopenia / abnormal bleeding
Early onset of thrombocytopenia
Too symptomatic (abnormal platelet function)
Mild to moderate thrombocytopenia
Poor response to IVIG/corticoids

…with leukemia predisposition?

Personal or family **history of hematological malignancy**
Associated abnormality: other cell line abnormality
Eczema, asthma

Variant classification and resolution of VUS

Data sharing initiatives and consortia

Myeloid Malignancy Variant Curation Expert Panel: specific classification rules for assessing RUNX1 variants pathogenicity

Functional assays:
- Established luciferase-based transactivation assays with known RUNX1 target gene (included in ACMG guidelines)
- Luciferase assays also established for assessment of ETV6 and ANKRD26 variants

Homan CC et al. Blood. 2023;141(13):1533-1543.
Luo X et al. Blood Adv. 2019 22;3(20):2962-2979.
Poggi et al. Haematologica. 2017;102(2):282–294.
Bluteau D et al. J Clin Invest. 2014;124(2):580-91.

How to manage the bleeding risk?

Managing bleeding risk in FPD-MM, THC2, and THC5

- **Patients often present with mild-to-moderate platelet counts and mild-to-moderate bleeding risk**
- **Main risk: surgeries or childbirth**
- **Antifibrinolytics and platelet transfusions as clinically indicated**
- **Short course of TPO receptor agonists** (ie, eltrombopag and romiplostim) **before surgery may have clinical utility** (off-label)

  Phase II clinical data included patients with ANRKD26 thrombocytopenia (n=9), lower response than MYH9/mBSS
  Further long-term clinical data needed, especially with long-term administration
  Usually, platelet response is observed within 3 weeks

Zaninetti C et al. Haematologica. 2020;105(3):820-828. `
Fiore M et al. Platelets. 2016;27(8):828-829.
Bastida JM et al. Int J Mol Sci. 2021;22(9):4330.

How to diagnose constitutional thrombocytopenias associated with leukemia predisposition?

The Hem/Onc perspective

European Reference Network for rare or low prevalence complex diseases
Network Hematological Diseases (ERN EuroBloodNet)
CRPP MHEMO
Webinars EuroBloodNet

WHO 2022 classification of myeloid neoplasms

→ **Toward a genetic basis for defining diseases**

Myeloid neoplasms with germline predisposition without a pre-existing platelet disorder or organ dysfunction

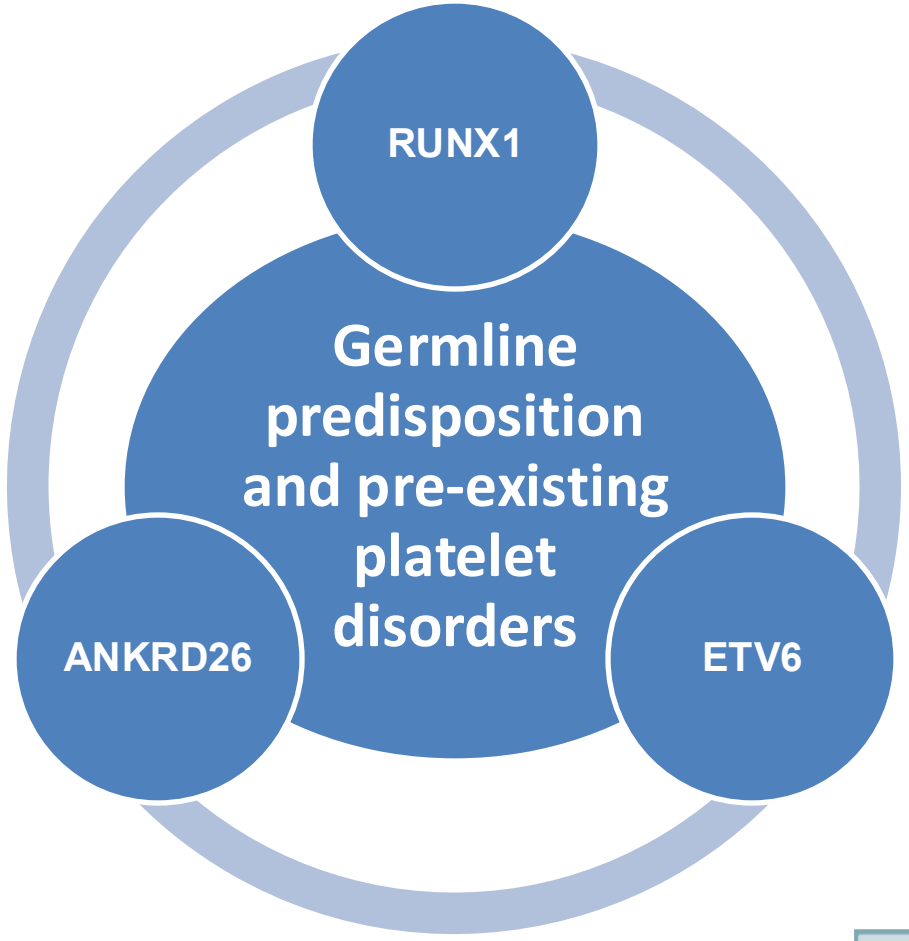
- Germline *CEBPA* P/LP variant (CEBPA-associated familial AML)
- Germline *DDX41* P/LP variant[a]
- Germline *TP53* P/LP variant[a] (Li-Fraumeni syndrome)

Myeloid neoplasms with germline predisposition and pre-existing platelet disorder

- Germline *RUNX1* P/LP variant[a] (familial platelet disorder with associated myeloid malignancy, FPD-MM)
- Germline *ANKRD26* P/LP variant[a] (Thrombocytopenia 2)
- Germline *ETV6* P/LP variant[a] (Thrombocytopenia 5)

Myeloid neoplasms with germline predisposition and potential organ dysfunction

- Germline *GATA2* P/LP variant (GATA2-deficiency)
- Bone marrow failure syndromes
  - Severe congenital neutropenia (SCN)
  - Shwachman-Diamond syndrome (SDS)
  - Fanconi anaemia (FA)
- Telomere biology disorders
- RASopathies (Neurofibromatosis type 1, CBL syndrome, Noonan syndrome or Noonan syndrome-like disorders[a,b])
- Down syndrome[a,b]
- Germline *SAMD9* P/LP variant (MIRAGE Syndrome)
- Germline *SAMD9L* P/LP variant (SAMD9L-related Ataxia Pancytopenia Syndrome)[c]
- Biallelic germline *BLM* P/LP variant (Bloom syndrome)



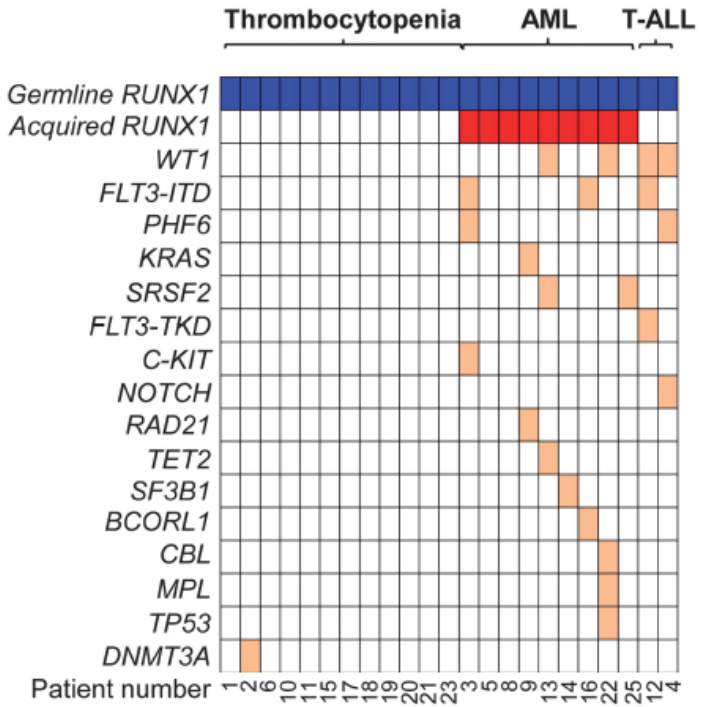
Khoury JD et al. Leukemia. 2022;36(7):1703-1719.

Webinars
EuroBloodNet

Screening at leukemia diagnosis

- **Family or personal history of chronic thrombocytopenia**
- **Routine use somatic cancer gene panels at leukemia diagnosis**
  - **Scrutiny for ETV6, RUNX1, ANKRD26 variants with high VAF (>30%)**
  - **Variant suspicious of being germline:**
    → germline confirmation
    → genetics referral for patient and family

RUNX1 variants induce genomic instability



Genomic instability

Additional somatic variants in myeloid MDS/AML :

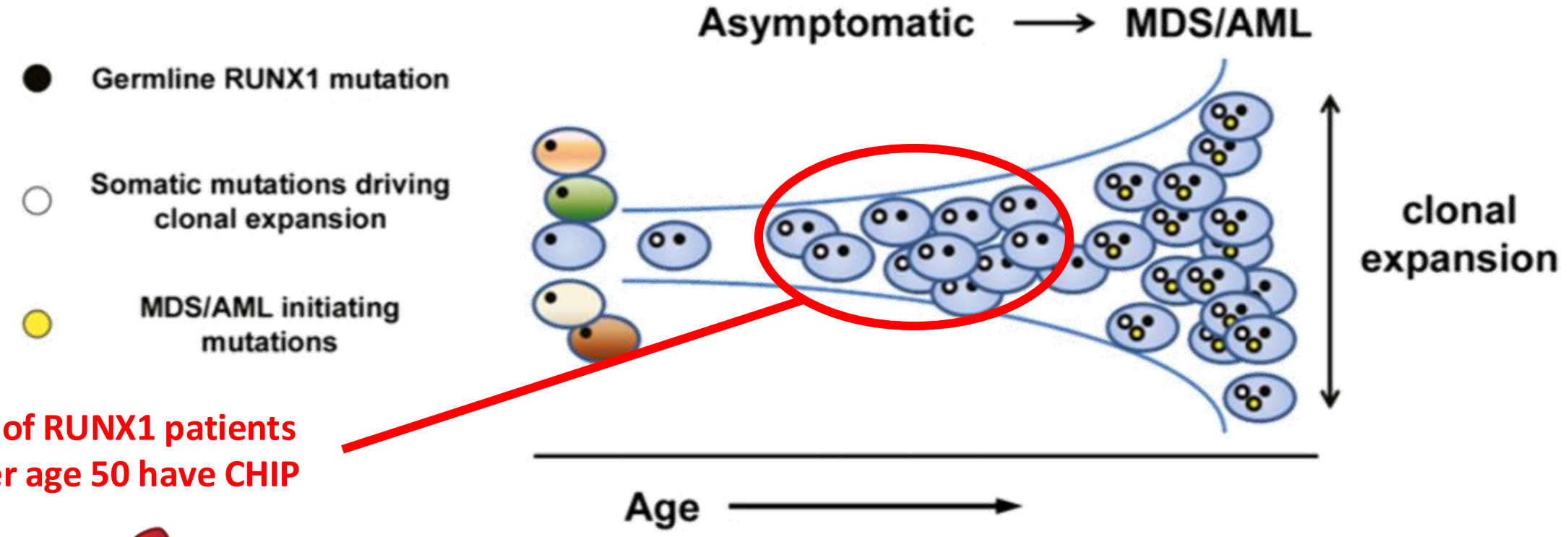
- RUNX1 +++ (biallelic alteration)

- Other variants : TET2, ASXL1, BCOR, PHF6, SRSF2

Antony-Debré et al. Leukemia. 2016;30(4):999–1002
Förster A et al. Cancers (Basel). 2022;14(14):3431.

European Reference Network for rare or low prevalence complex diseases
Network Hematological Diseases (ERN EuroBloodNet)
CRPP MHEMO
Webinars EuroBloodNet

RUNX1 variants induce genomic instability and early onset clonal hematopoiesis (CHIP)



67% of RUNX1 patients under age 50 have CHIP

Churpek JE. Blood. 2015 Nov 26;126(22):2484-90

European Reference Network for rare or low prevalence complex diseases

Network Hematological Diseases (ERN EuroBloodNet)

CRPP MHEMO

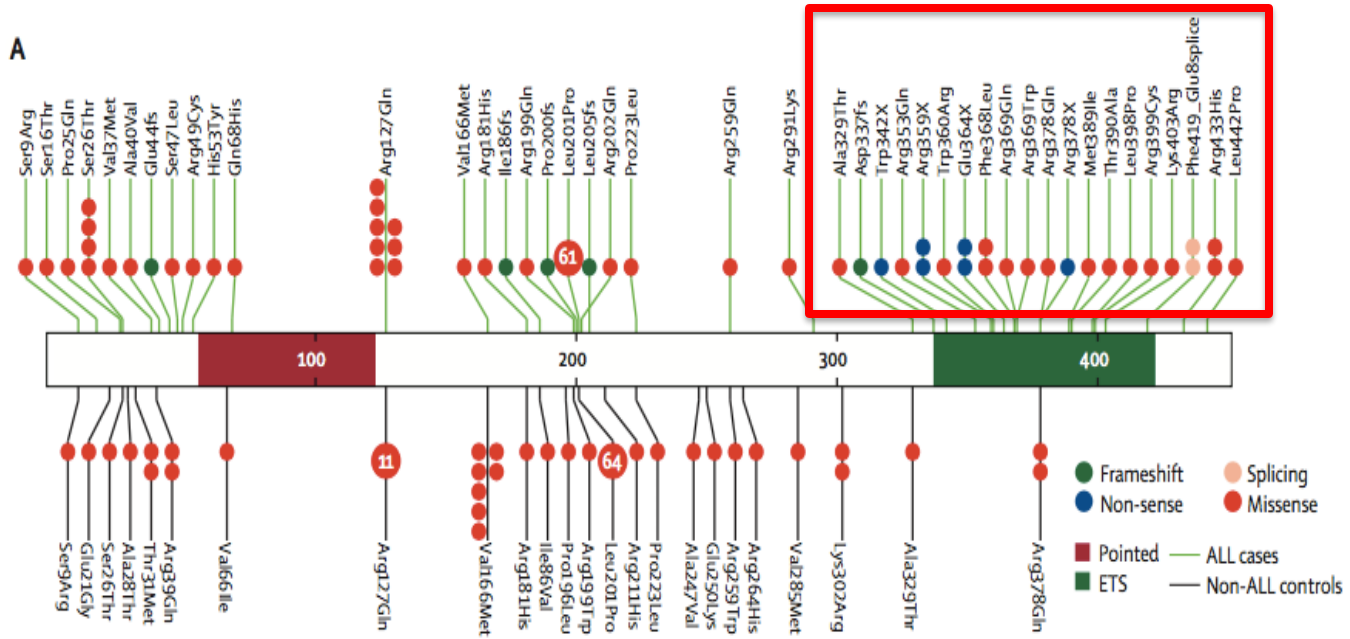
Webinars EuroBloodNet

ETV6 predisposition and childhood ALL

Childhood ALL (n=4405) : *ETV6* sequencing (constitutional)

CASES
(childhood ALL)

CONTROLS
(general population)



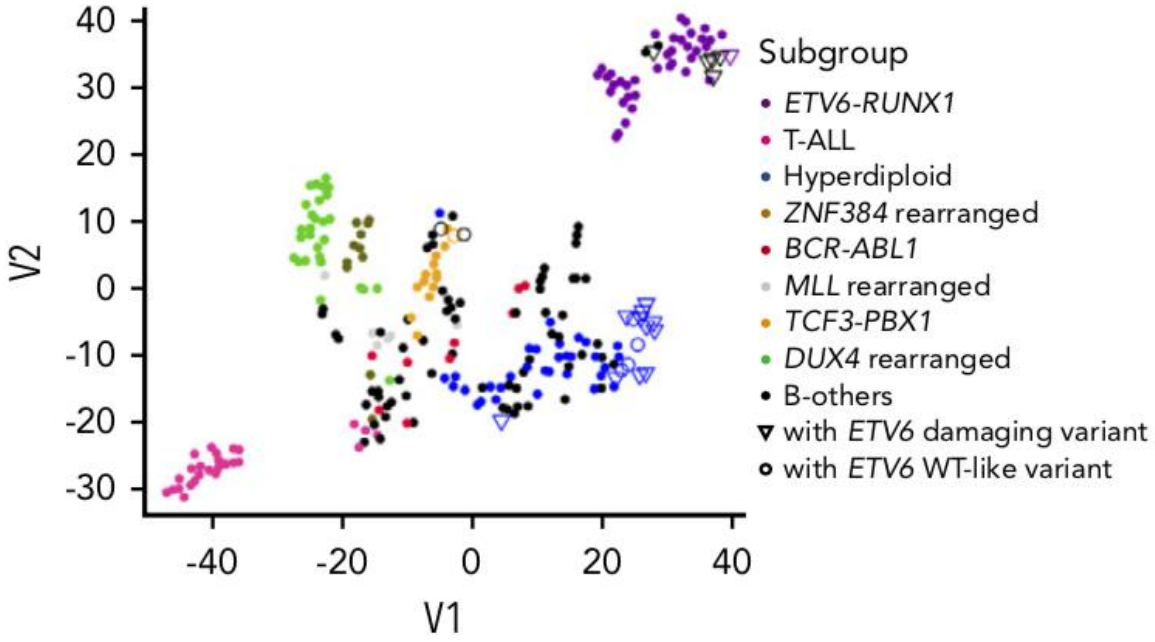
Childhood ALL: ~1% *ETV6* constitutional variant

Hyperdiploid B-ALL of older children
No impact on early response / relapse

Moriyama et al. Lancet Oncol. 2015;16(16):1659–1666.

European Reference Network for rare or low prevalence complex diseases
Network Hematological Diseases (ERN EuroBloodNet)
CRPP MHEMO
Webinars EuroBloodNet

ETV6 predisposition and childhood ALL



ETV6 predisposition somatic signature

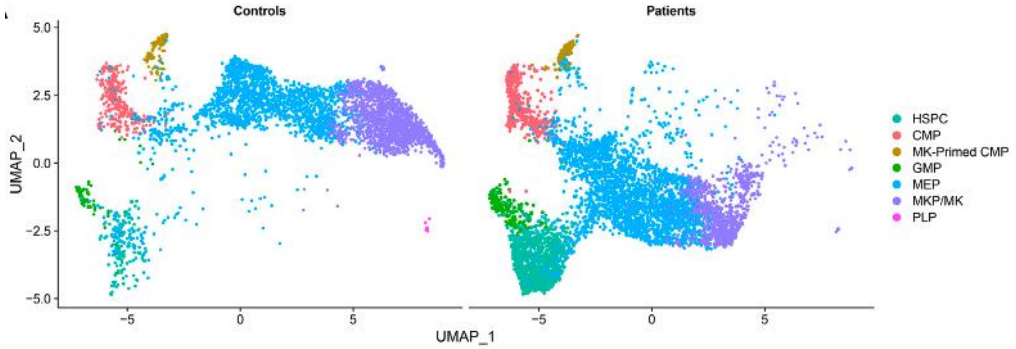
1) Hyperdiploid
variants RAS pathway
2) Diploid
Focal deletion ETV6, PAX5

Nishii R et al. Blood. 2021;137(3):364-373.

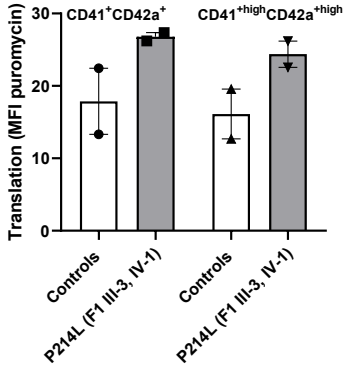
ETV6 variants alter ribosome biogenesis and translation machinery

Strong transcriptomic differences in the MEPs and MKs from ETV6 patients

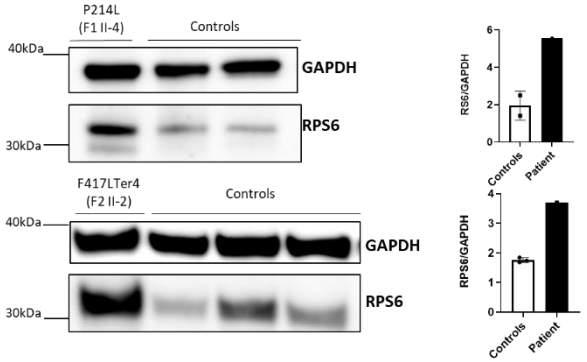
Dysregulated “DNA repair” and “translation” pathways



Increased translation levels



Increased ribosomal protein RPS6 expression in MKs / platelets



MC Alessi | Marjorie Poggi | Timothée Bigot | Elisa Gabinaud

C2VN, Inserm 1263

Bigot T et al. J Thromb Haemost. 2023;21(9):2528-2544.

Constitutional thrombocytopenia with leukemia predisposition

	RUNX1	ETV6	ANKRD26
Transmission	----------	Autosomal dominant	----------
Bleeding	0 to ++	0 to ++	0 to +
Thrombocytopenia	0 to +	+	+ to +++
Platelet volume	----------	Normal	----------
Hematological malignancy	~40% Myeloid +++ All age groups (m=35y)	15 - 25% ? Pediatric B-ALL +++ AML/MDS in adults	~10% Myeloid +++ older adult (m=40y)
Somatic signature	Loss of RUNX1 WT allele	Hyperdiploidy Loss of ETV6 WT allele	

European Reference Network
for rare or low prevalence complex diseases
Network Hematological Diseases (ERN EuroBloodNet)

CRPP
MHEMO

Webinars
EuroBloodNet

Which hematological follow-up for constitutional thrombocytopenias associated with leukemia predisposition?

Hematological initial workup and follow-up

No evidence-based guidelines

Hematological initial workup and follow-up

At diagnosis: clinical examination + CBC + BM assessment + cytogenetics + somatic NGS panel

Follow-up /6-12 months : clinical examination + CBC + somatic NGS panel in peripheral blood

In case of clinical/CBC abnormality or clonal evolution (NGS): repeat BM assessment

Clinical utility of assessment of clonal evolution using NGS need further prospective data

In case of hematological malignancy

Identification of a related donor who does not carry the germline variant

Indication for HSCT in CR1 to be discussed (RUNX1+++, ETV6?, ANKRD26?)

European Reference Network for rare or low prevalence complex diseases

Network Hematological Diseases (ERN EuroBloodNet)

CRPP MHEMO

Homan CC et al. Blood. 2023;141(13):1533-1543.
Porter et al. Clinical Cancer Research. 2017;23(11):e14–e22.
Furutani et al. J Clin Oncol. 2017;35(9):1018–1028.
Churpek et al. Blood. 2016;128(14):1800–1813.

Webinars EuroBloodNet

Conclusion

Thrombocytopenia should be explored even in the absence of bleeding (after patient information)

Clinical and biological diagnosis
- Family history, autosomal dominant transmission, normal platelet volume and smears, gene sequencing+++
- At hematological malignancy stage: somatic signatures

Pathophysiology: transcription/signaling defect, genomic instability, secondary events, loss of WT allele

Bleeding risk management :
- Mainly for surgeries or childbirth
- Antifibrinolytics and platelet transfusion as clinically indicated
- Short course TPO receptor agonists (ie, eltrombopag and romiplostim) before surgery may have clinical utility

Hematological initial workup and follow-up (incl. sequential NGS), anticipate HSCT

Perspectives
- prediction / monitoring of progression to MDS/leukemia
- gene therapy / pharmaceutical approach to restore RUNX1/ETV6/ANKRD26 gene function
- targeted therapies for clonal secondary events

Acknowledgments

C2VN lab, Inserm 1263 , Aix Marseille Université –
CRPP (French Reference Center for Inherited Platelet Disorders)
Laboratory of Hematology, La Timone Hospital, APHM, Marseille France

Marie Christine Alessi
Marjorie Poggi
Pierre Emmanuel Morange
Timothée Bigot
Elisa Gabinaud
Céline Falaise
Pierre Morange
Noémie Saut
Marie Loosveld

French Reference Center
for Inherited Platelet Disorders

Co-funded by
the Health Programme
of the European Union

Department of Pediatric Hematology, Immunology, Oncology, La Timone Children Hospital : Hervé Chambost, Gérard Michel

EuroBloodNet: Mariangela Pellegrini, Maryam Lalem, Pierre Fenaux, Béatrice Gulbis