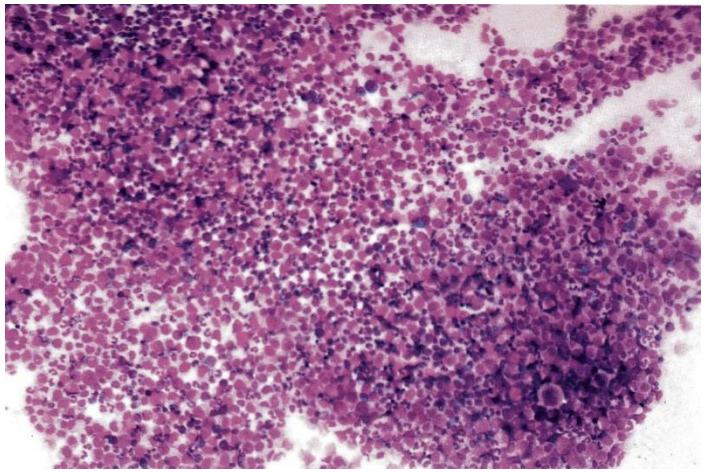


Neoplasie mieloproliferative croniche

prof. Gian Matteo Rigolin

The 2016 revision to the World Health Organization Classification of Myeloid Neoplasms and acute leukemias

1. Myeloproliferative neoplasms
2. Myeloid/lymphoid neoplasms with eosinophilia and rearrangement of PDGFRA, PDGFRB, or FGFR1, or with PCM1-JAK2
3. Myelodysplastic/myeloproliferative neoplasms (MDS/MPN)
4. Myelodysplastic syndromes (MDS)
5. Blastic plasmacytoid dendritic cell neoplasm
6. Acute leukemias of ambiguous lineage
7. Acute myeloid leukemia (AML) and related neoplasms
8. B-lymphoblastic leukemia/lymphoma
9. T-lymphoblastic leukemia/lymphoma

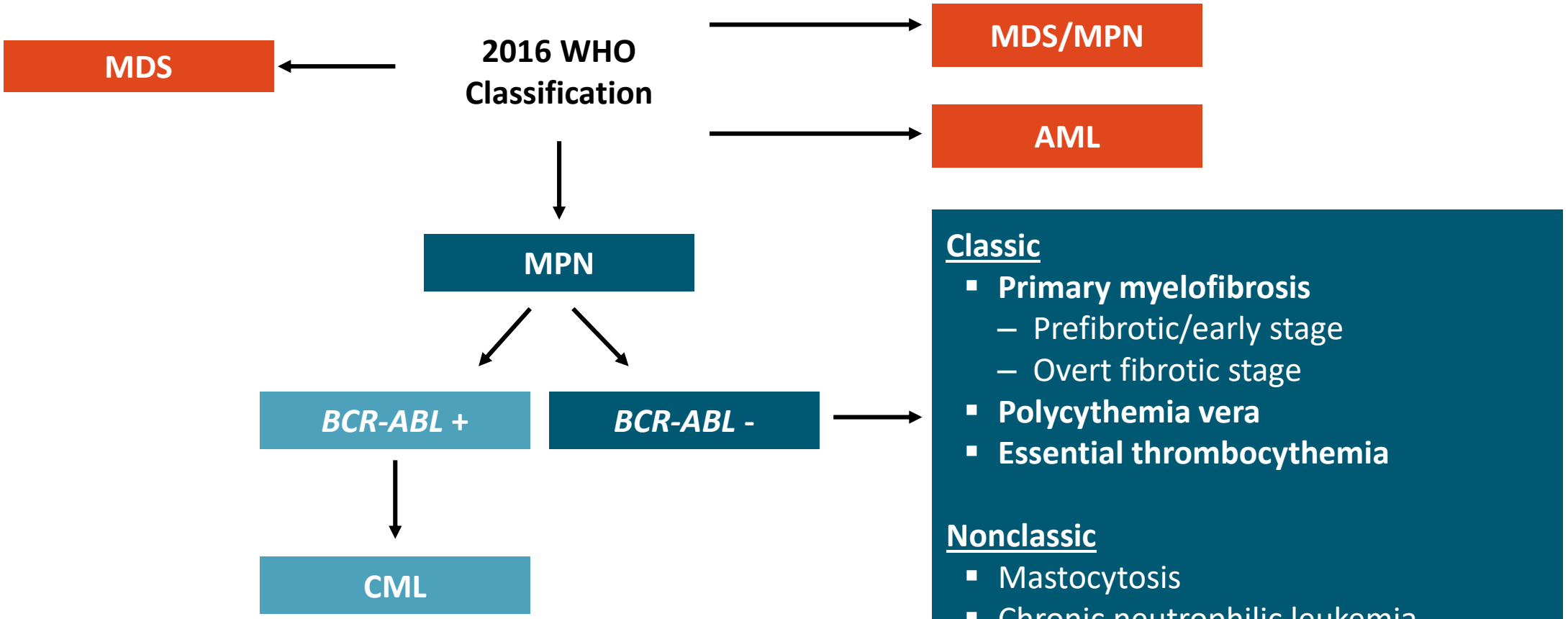
Arber DA et al. Blood. 2016; 127(20):2391-2405

The 2016 revision to the WHO classification of myeloid neoplasms and acute leukemia

Myeloproliferative neoplasms (MPN)
Chronic myeloid leukemia (CML), *BCR-ABL1*$^{+}$
Chronic neutrophilic leukemia (CNL)
Polycythemia vera (PV)
Primary myelofibrosis (PMF)
PMF, prefibrotic/early stage
PMF, overt fibrotic stage
Essential thrombocythemia (ET)
Chronic eosinophilic leukemia, not otherwise specified (NOS)
MPN, unclassifiable
Mastocytosis

Arber DA et al. Blood. 2016; 127(20):2391-2405

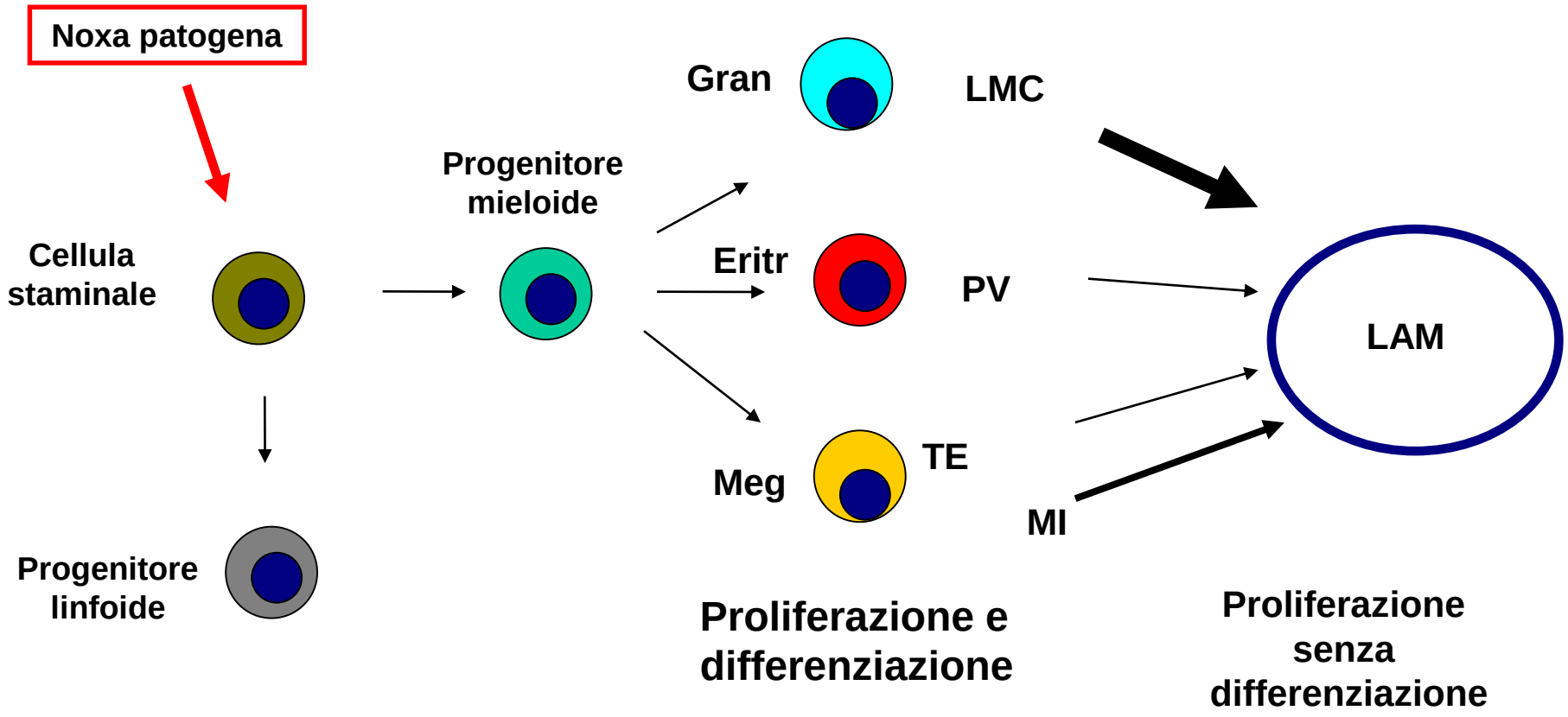
Myeloid Malignancies



Arber. Blood. 2016;127:2391.

Slide credit: clinicaloptions.com

SMP: definizione



Varietà di disordini clonali acquisiti della cellula staminale pluripotente, contrassegnati dalla proliferazione clonale di uno o più progenitori emopoietici nel midollo ed in sedi extramidollari

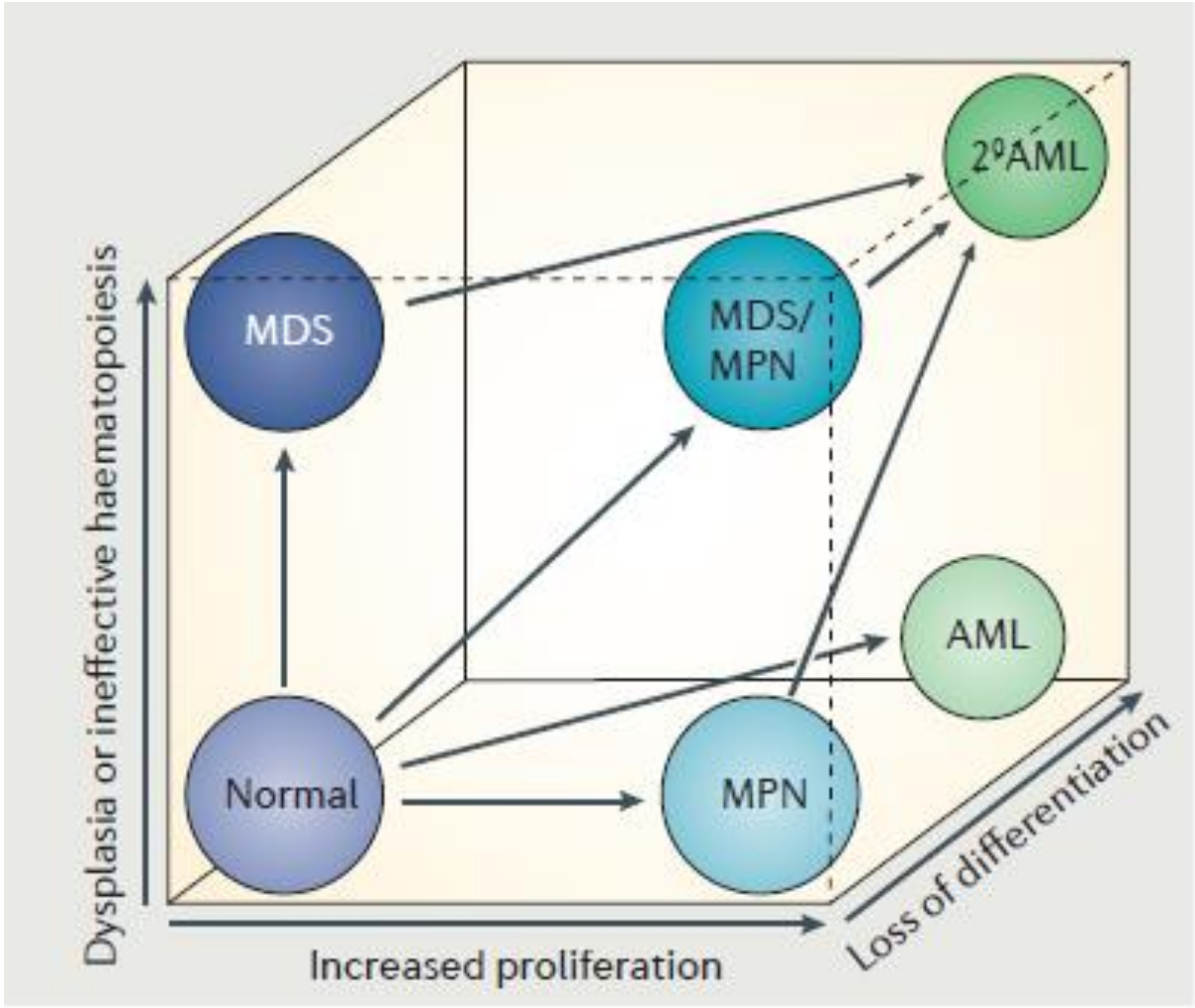
Differentiation and proliferation

	MDS	AML	MPD
Differentiation	Impaired	Impaired	Normal ← ←
Proliferation/survival	Impaired → →	Preserved	Increased

Arrows indicate where a second hit could result in progression to AML.

Nimer SD. Blood 2008;111:4841.

Normal myelopoiesis and myeloid malignancies



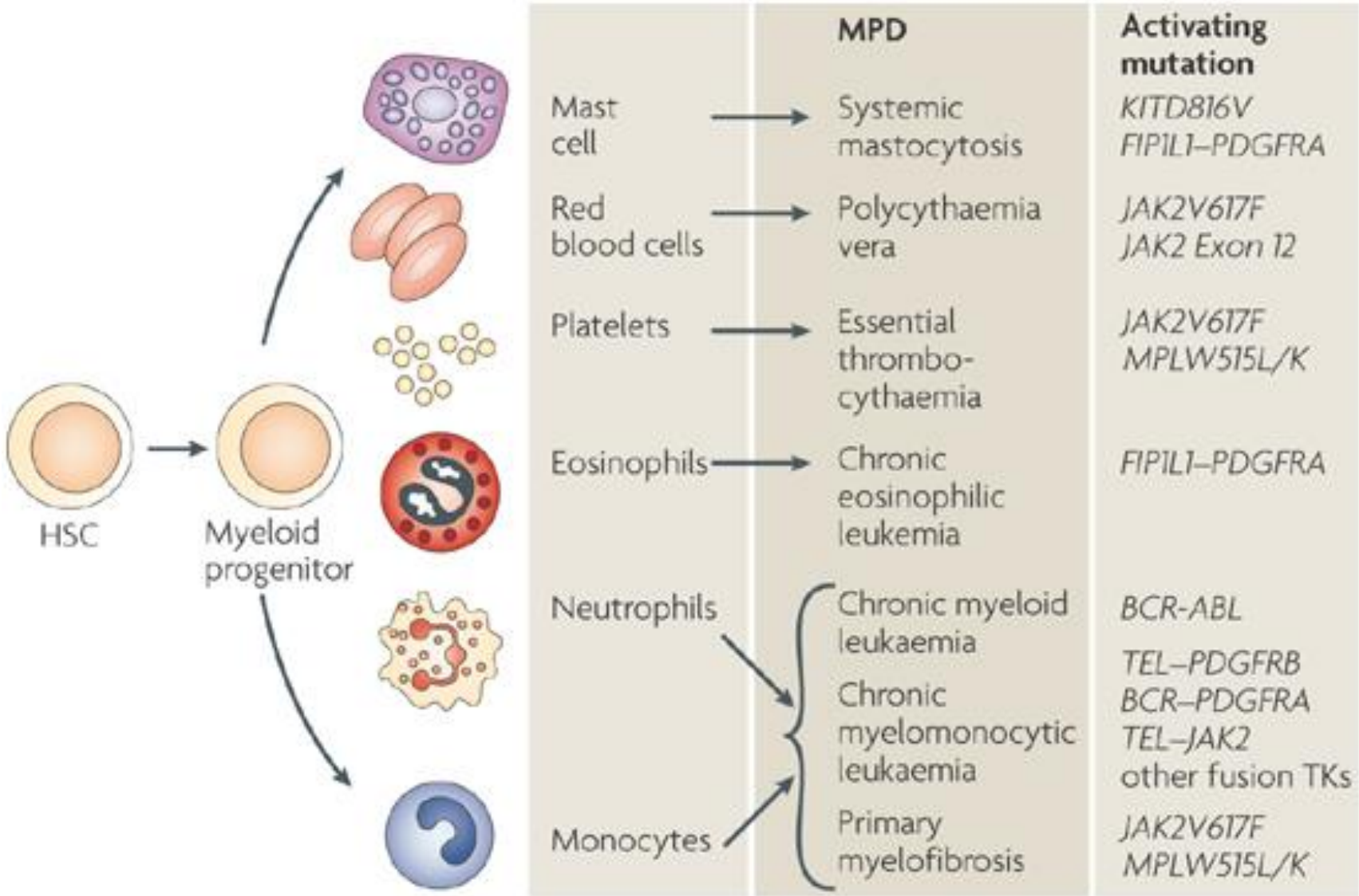
Deininger MWN et al. Nature Review Cancer. 2017;17:425

- Myeloproliferative neoplasms (MPN) are characterized by excess proliferation in one or more of the myeloid lineages and frequently by extramedullary haematopoiesis. Blood cell morphology is normal and differentiation is maintained.
- Myelodysplastic syndromes (MDS) exhibit decreased numbers of cells in the blood, whereas their bone marrow is frequently hypercellular (ineffective haematopoiesis).
- Acute myeloid leukaemia (AML) is characterized by differentiation arrest and accumulation of primitive undifferentiated myeloid cells (myeloblasts)
- MDS/MPN display a combination of the features of MDS and MPN with dysplasia and excess production of blood cells in at least one of the myeloid lineages.

Tyrosine kinase genes in MPN

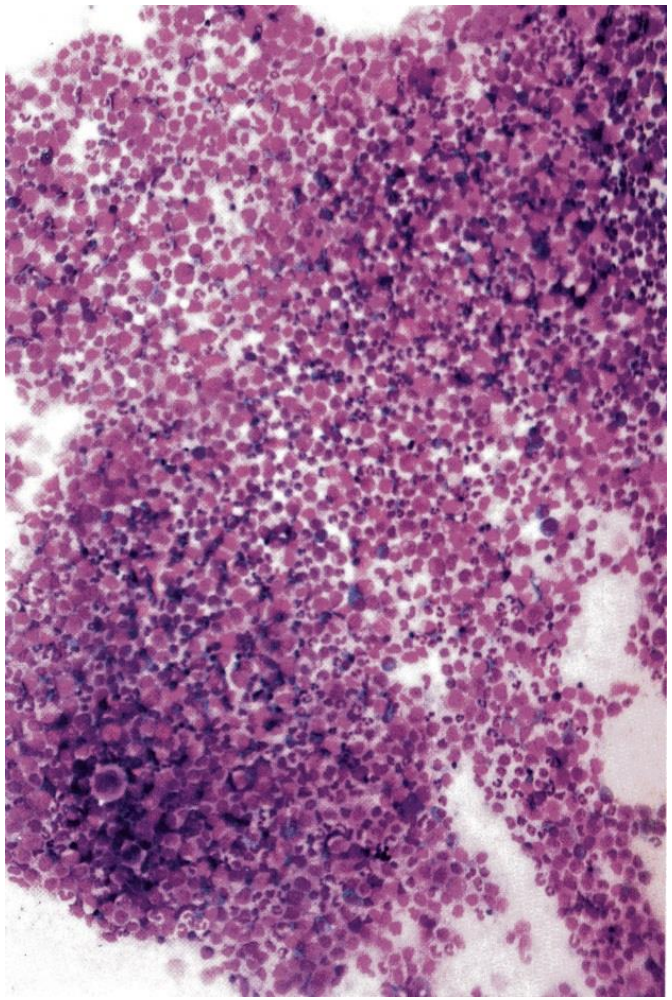
- **9q34: ABL1**
  - t(9;22)(q34;q11): BCR-ABL1 CML
- **9p24: JAK2**
  - JAK2(V617F): PV, ET, IM
- **5q33: PDGFRB**
  - t(5:12)(q33;p13): ETV6-PDGFRB CMML with eosinophilia
- **8p11: FGFR1**
  - t(8;13)(p11;q12): 8p11 CMPD
- **4q12: PDGFRA**
  - del4q12: FIP1L1-PDGFRA: HES
- **4q12: KIT**
  - KIT (D816V): systemic mastocytosis

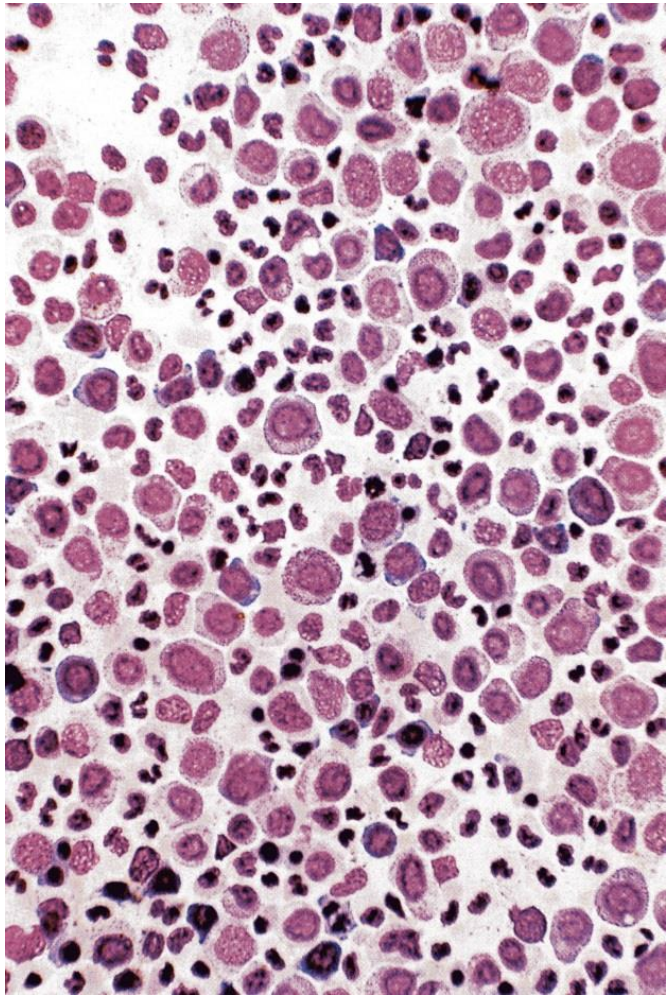
Classification and molecular pathogenesis of the MPD



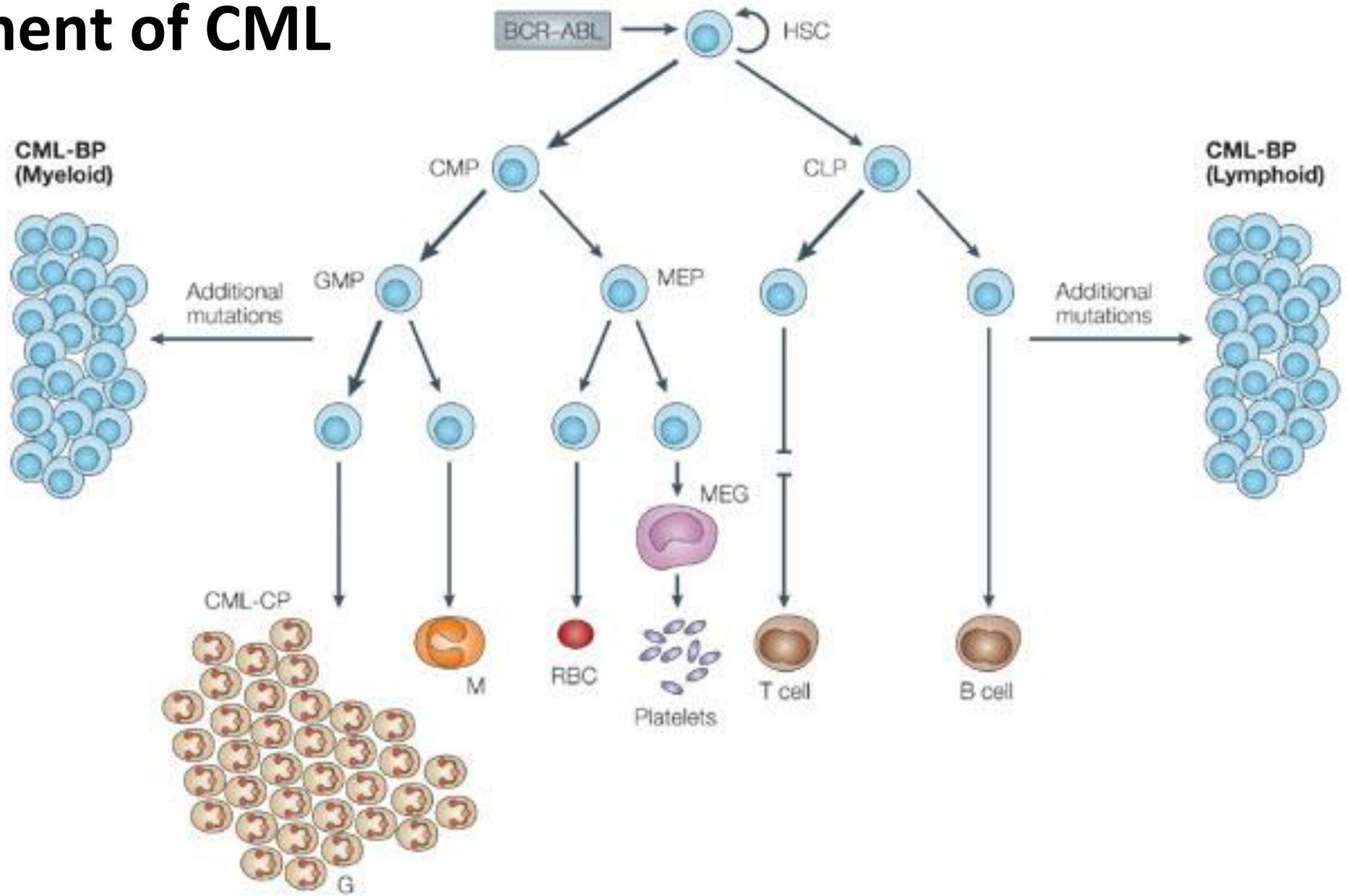
Levine RL et al. Nat Rev Cancer. 2007;7:673-83

LEUCEMIA MIELOIDE CRONICA



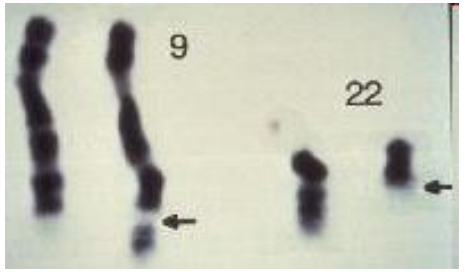


The development of CML



Nature Reviews | Cancer

Milestones in the history of CML



1960: Abnormal chrom. 22 (Philadelphia chrom.) identified and associated with CML

1973: Translocation 9;22 defined

1983: Molecular studies of fusion abnormality of breakpoint cluster gene (bcr) with cellular abl gene (c-abl)

1984: Fusion cytoplasmic protein BCR-ABL found to alter cell proliferation, adhesion and survival

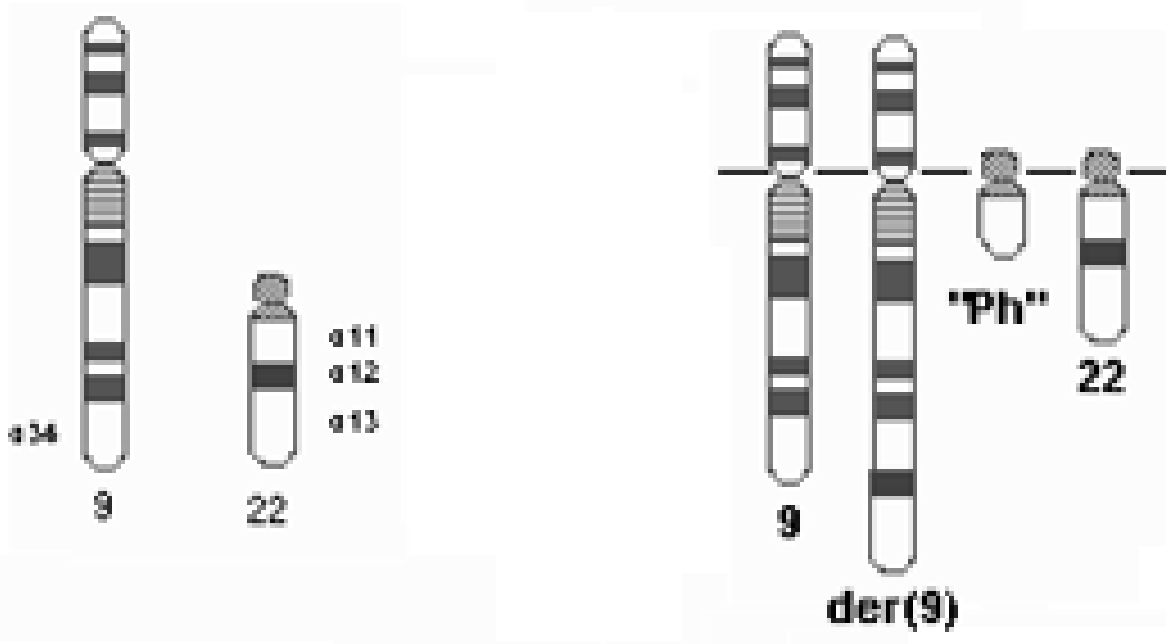
1984: Constitutive abnormal BCR-ABL tyrosine kinase activity defined

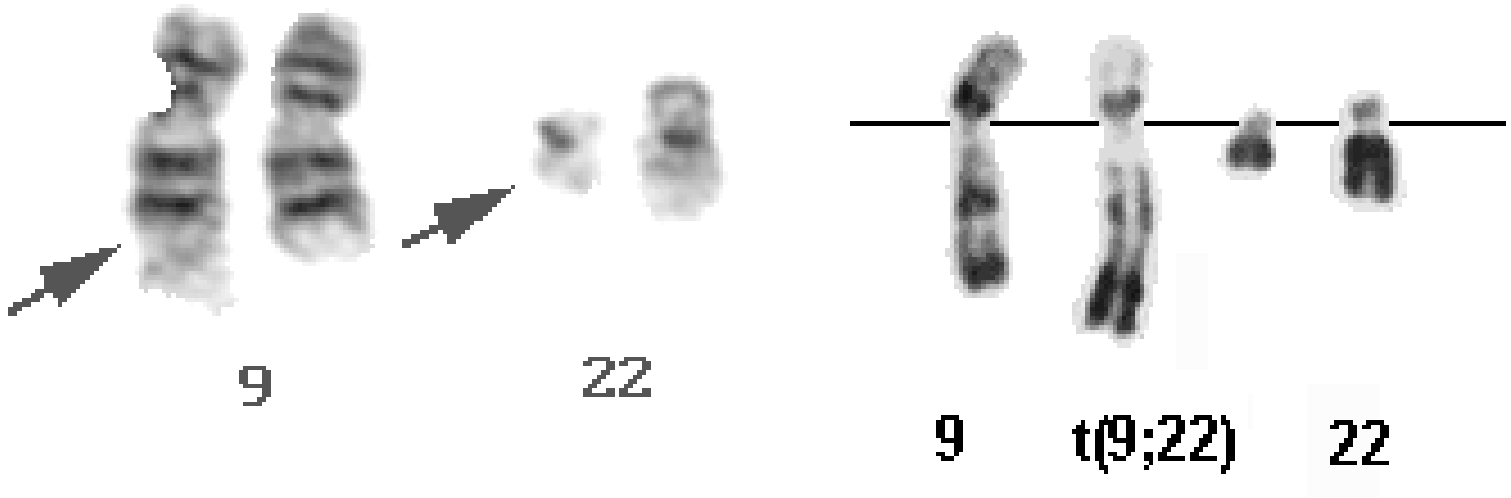
1988: Development of synthetic pharmacologic inhibitors that target tyrosine kinases

1998: Phase I clinical trials using STI-571 initiated

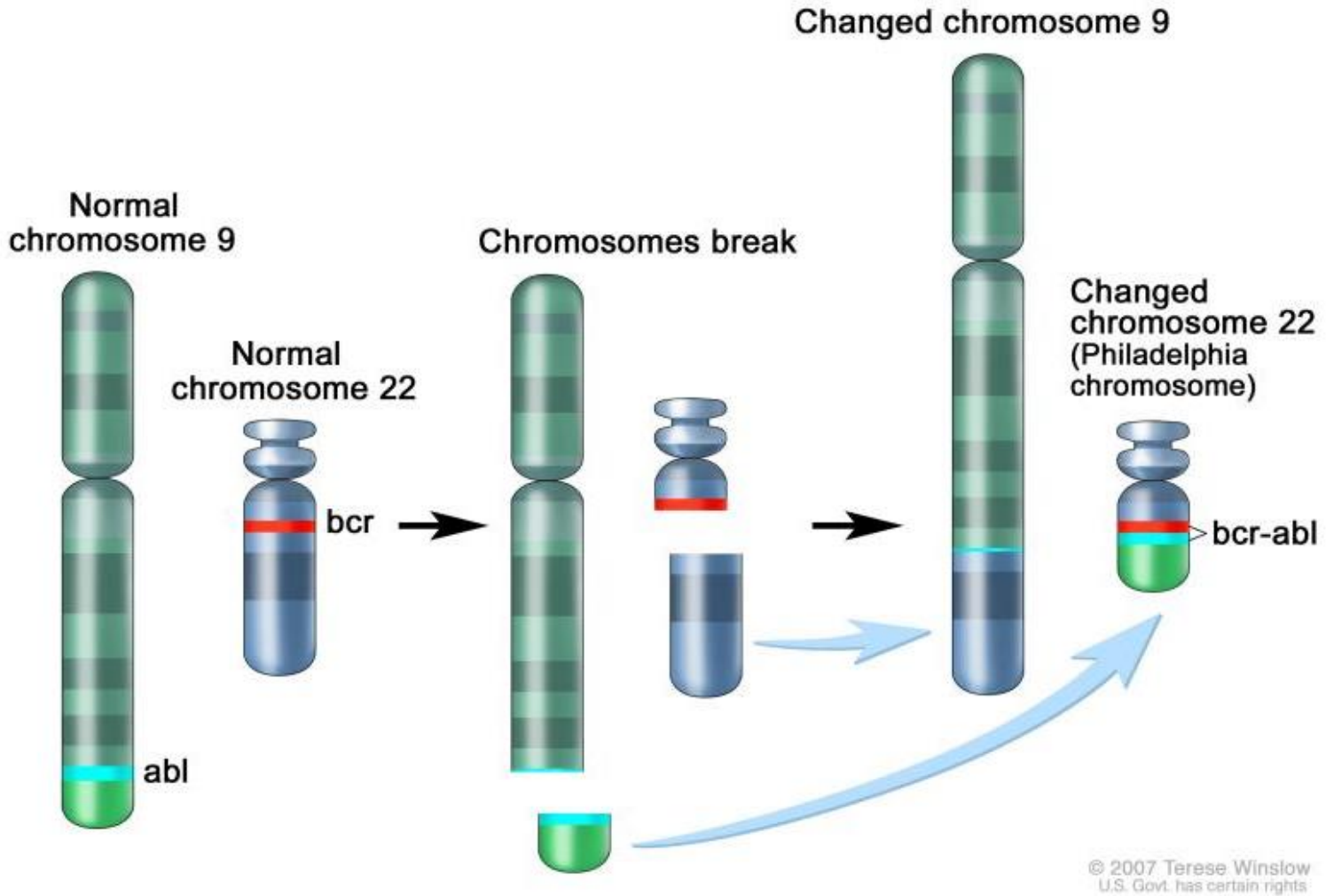
2001: STI571 is approved for treatment of CML that is refractory to IFN-therapy

t(9;22)



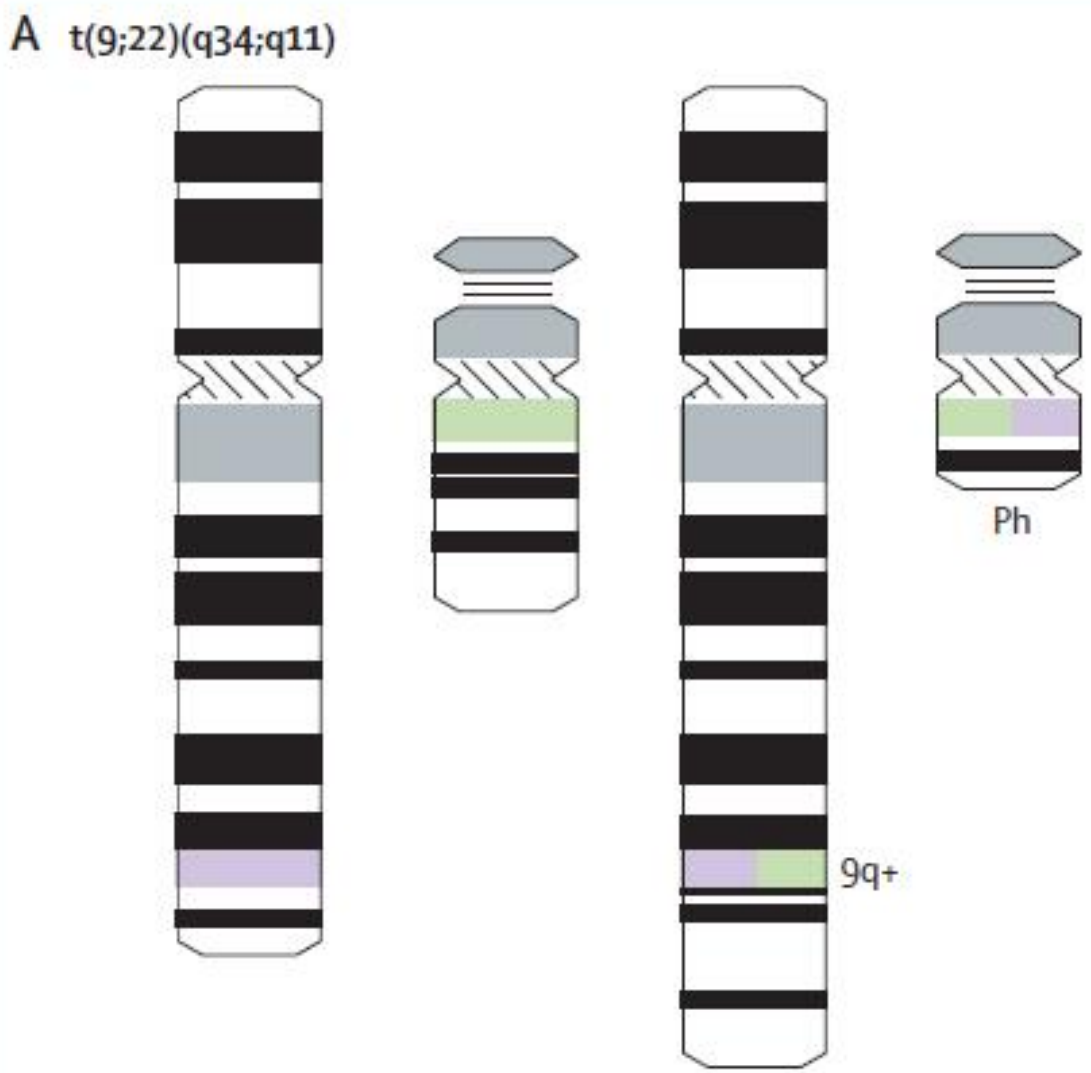


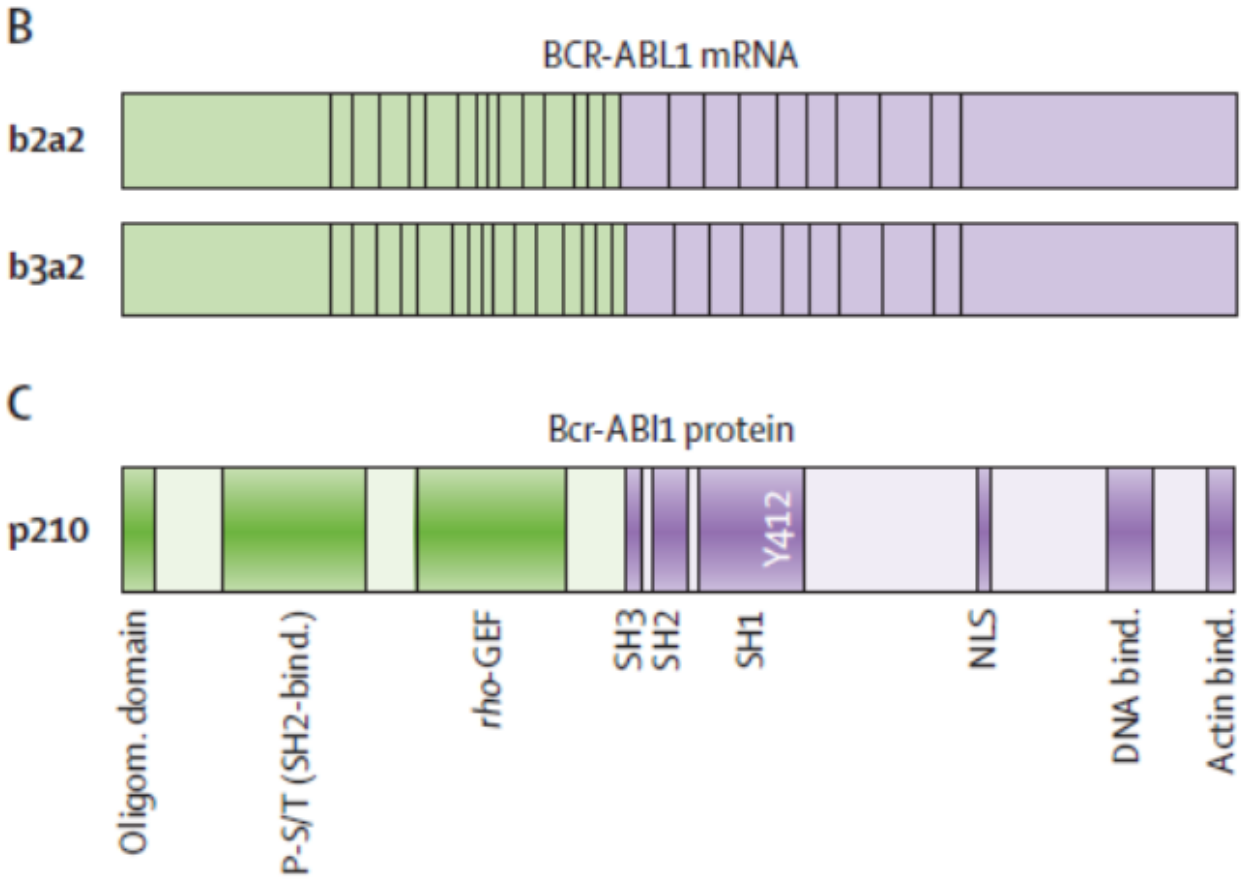
Schematic diagram of the translocation that creates the Philadelphia chromosome.



Pathogenesis of CML

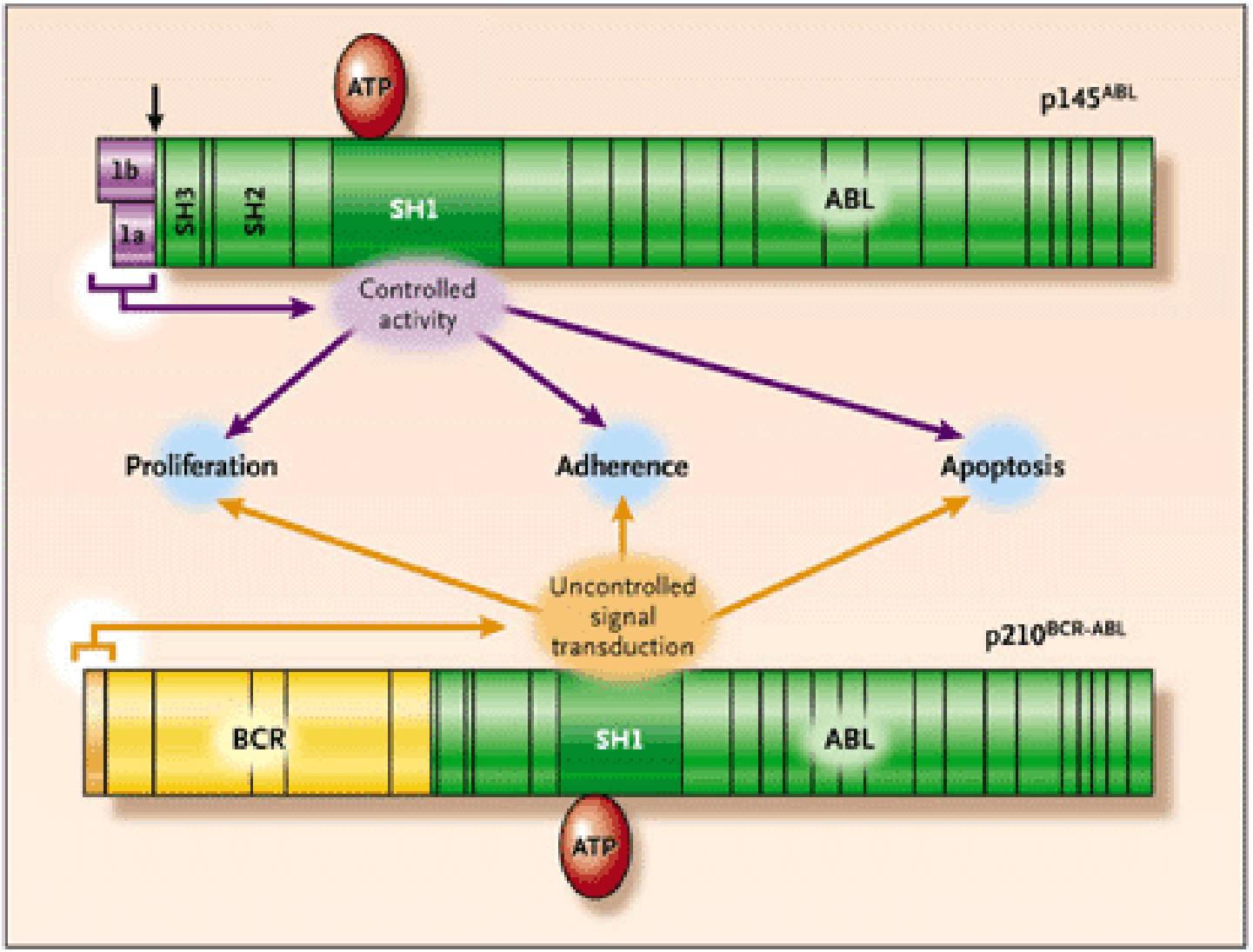
The t(9;22) reciprocal translocation (A) results in the creation of the *BCR-ABL1* fusion gene, which is in turn transcribed to a BCR-ABL1 mRNA (B), and translated to the Bcr-Abl protein (C)





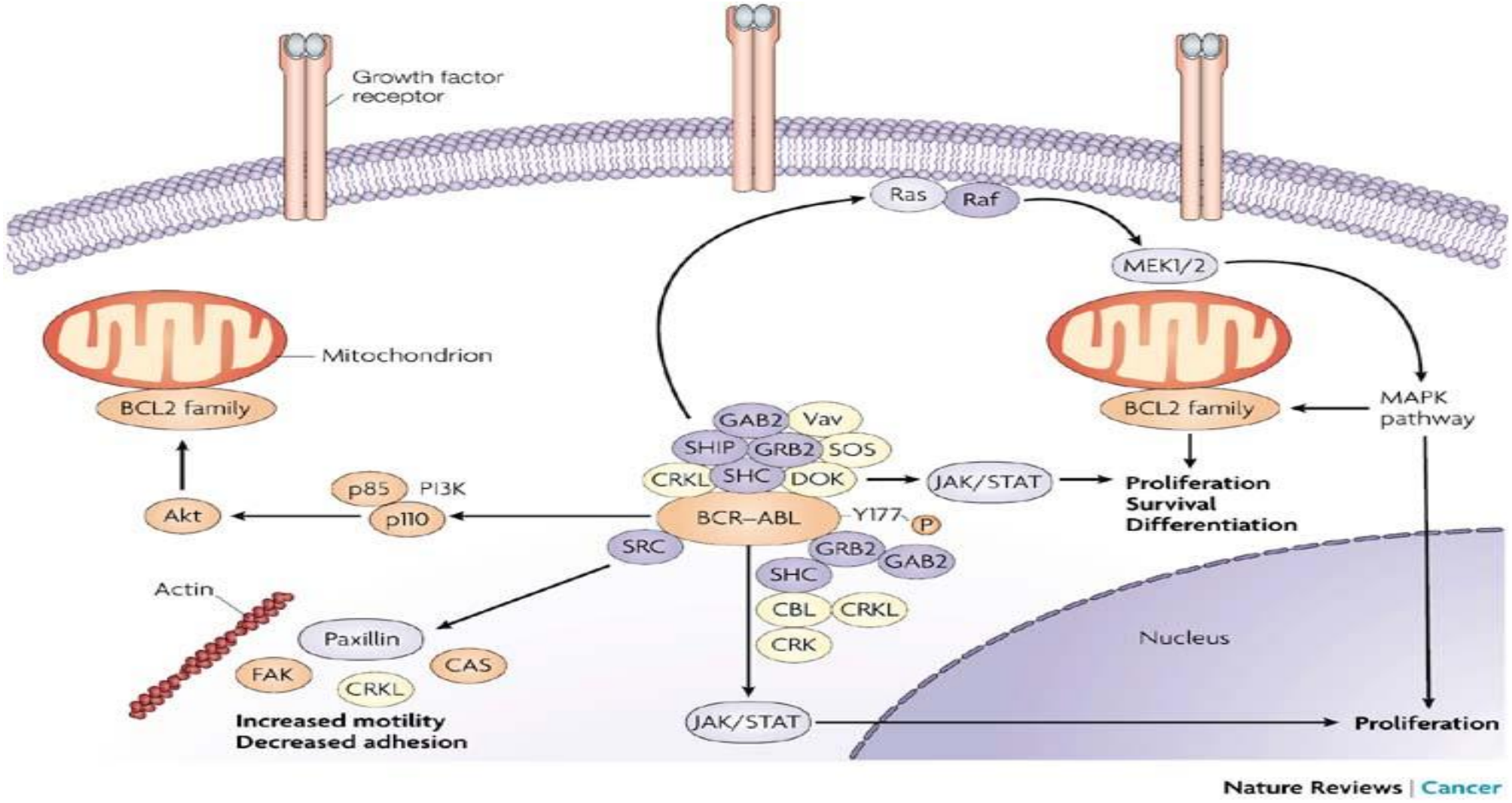
Apperley JF. Lancet 2015; 385: 1447–59

Deregulation by BCR-ABL of proliferation, adherence, and apoptosis



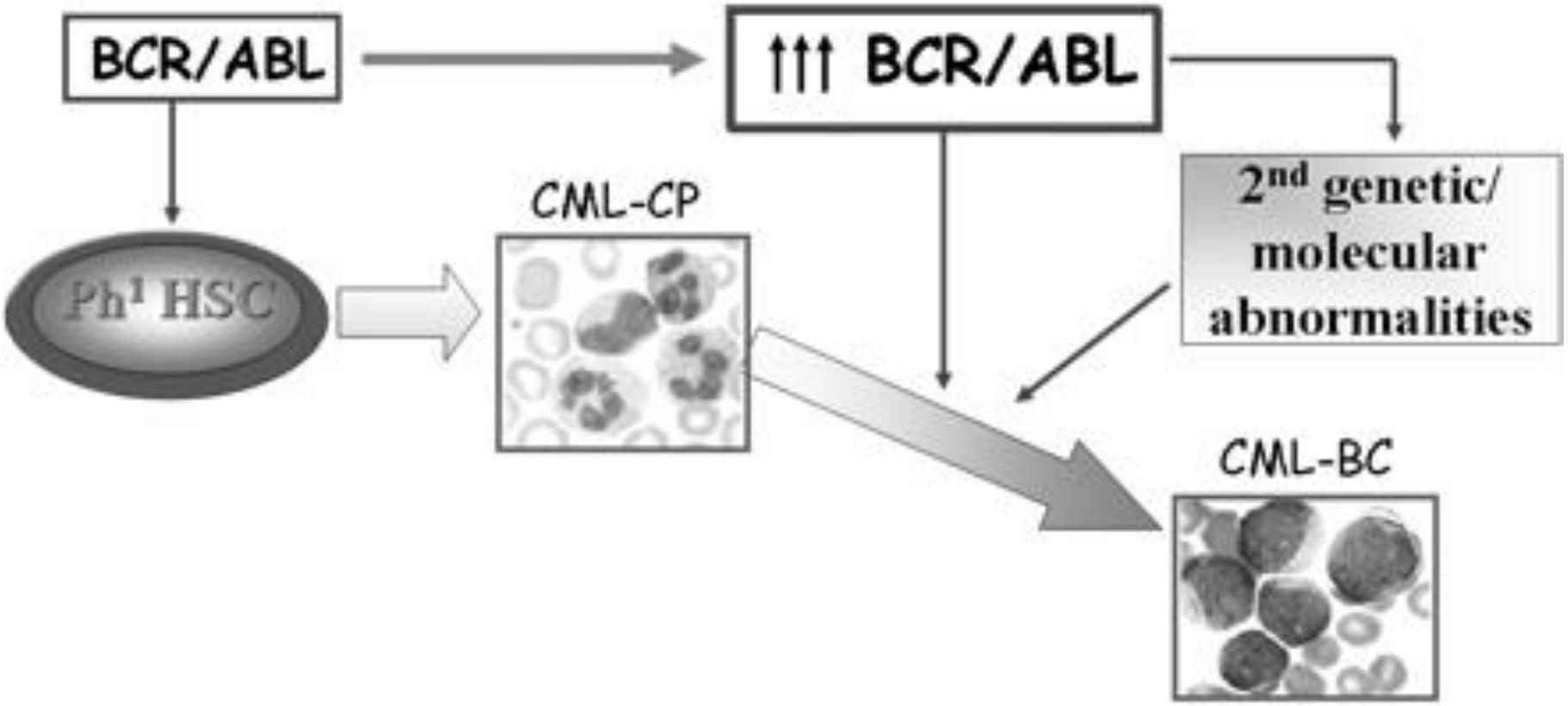
Goldman JM, Melo JV. NEJM. 2003;349:1451-64

Main BCR/ABL-activated pathways regulating proliferation and survival of hematopoietic cells



Weisberg E et al. Nature Reviews Cancer 7:345-356;2007.

Possible mechanisms of CML disease progression



LMC: epidemiologia

- Rappresenta il 15-20% di tutte le leucemie
- Incidenza 1-1,5 casi/100.000 individui anno
- M>F
- Età mediana: 50 anni

Panel 1: Presenting symptoms and signs of chronic myeloid leukaemia

Frequent

- Fatigue
- Night sweats
- Malaise and weight loss
- Left upper quadrant pain, discomfort, satiety
- Splenomegaly

Less frequent

- Priapism
- Retinal haemorrhages
- Thrombosis, bleeding, or both
- Bone pain*
- Hepatomegaly
- Lymphadenopathy*
- Skin infiltration*
- Extramedullary mass (chloroma)*

*Should raise suspicions of presentation with advanced phase disease.

LMC: clinica

Asintomatica in un terzo dei casi

- Leucocitosi di diversa entità
- Splenomegalia

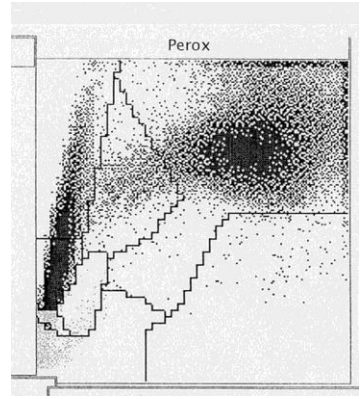
- **Fase accelerata/blastica:**
  - come leucemia

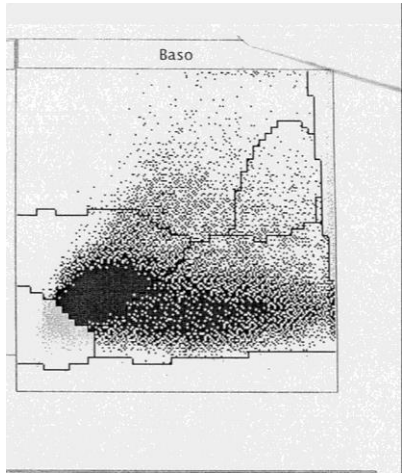
Apperley JF. Lancet 2015; 385: 1447–59

TEST	RISULT	PAT	NORMALI	UNITA'
WBC		107.0	(5.2 - 12.4)	x10.e3 /uL
RBC		3.71	(4.2 - 6.1)	x10.e6 /uL
HGB		11.7	(12 - 18)	g/dL
HCT		34.1	(37 - 50)	%
MCV	91.9		(80 - 99)	fL
MCH		31.5	(27 - 31)	pg
MCHC	34.3		(33 - 37)	g/dL
CHCM	34.0		(33 - 37)	g/dL
RDW		15.4	(11.5 - 14.5)	%
HDW	3.10		(2.2 - 3.2)	g/dL
PLT	177		(130 - 400)	x10.e3 /uL
MPV	8.1		(7.2 - 11.1)	fL
			(40 - 74)	%
			(19 - 48)	%
			(3.4 - 9)	%
			(0 - 7)	%
			(0 - 1.5)	%
			(0 - 4)	%
			(1.9 - 8)	x10.e3 /uL
			(0.9 - 5.2)	x10.e3 /uL
			(0.16 - 1)	x10.e3 /uL
			(0 - 0.8)	x10.e3 /uL
			(0 - 0.2)	x10.e3 /uL
			(0 - 0.4)	x10.e3 /uL
LI	2.42		(1.90 - 3)	
MPXI	9.5		(-10 - 10)	
WBCPEROX	106.2			
WBC BASO	107.0			

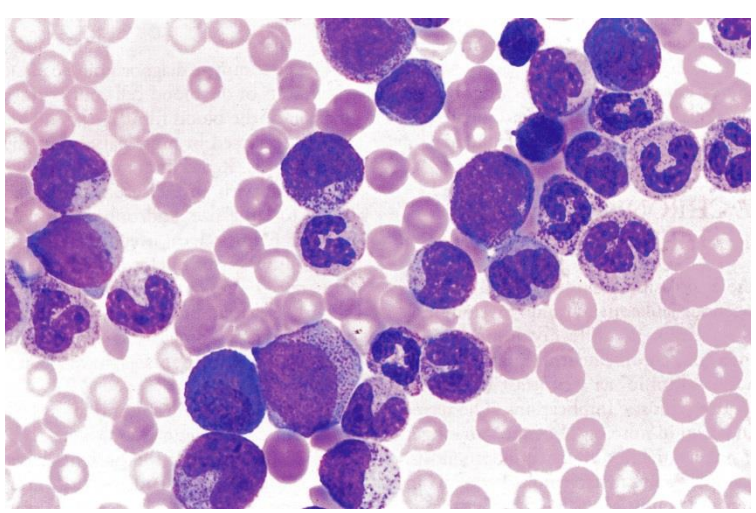
Formula al microscopio ottico
Neutrofili 66%
Promielociti 5%
Mielociti 6%
Metamielociti 6%
Blasti 1%
Linfociti 4%
Monociti 8%
Eosinofili 1%
Basofili 3%

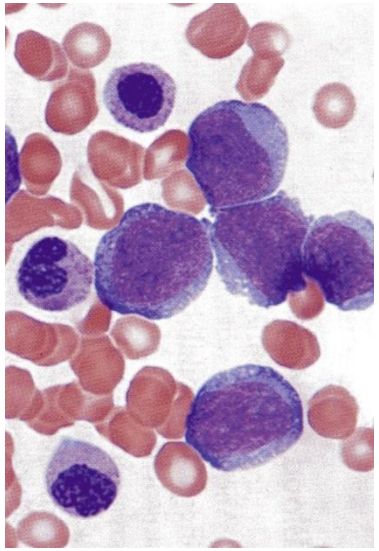
IG	+++
LS	+
ATYP	++
BLASTS	++

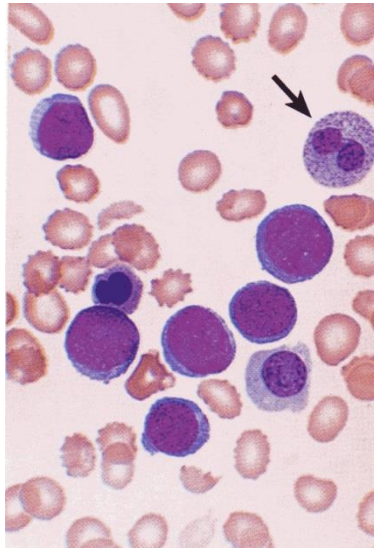


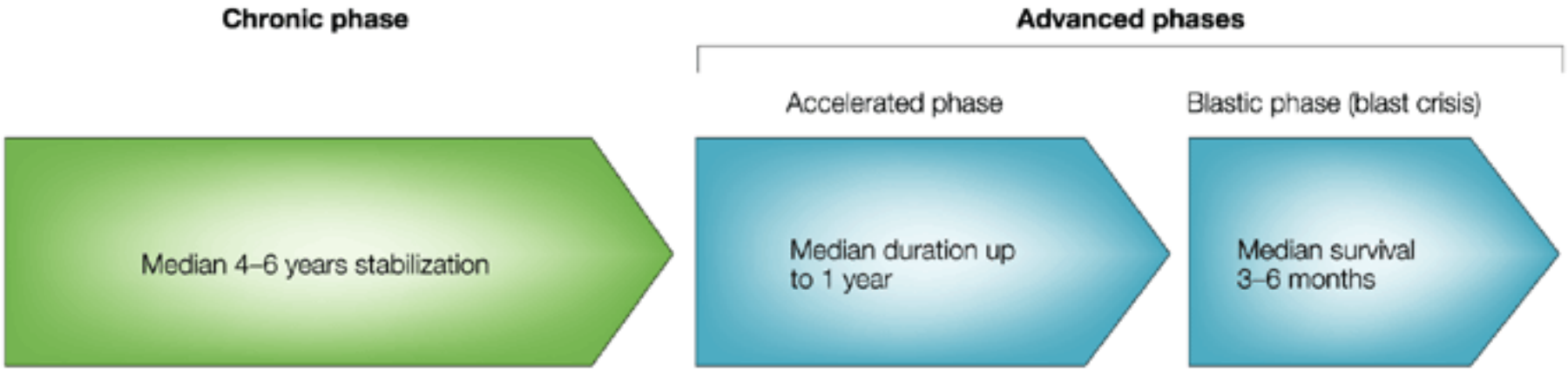


Clinical course of CML



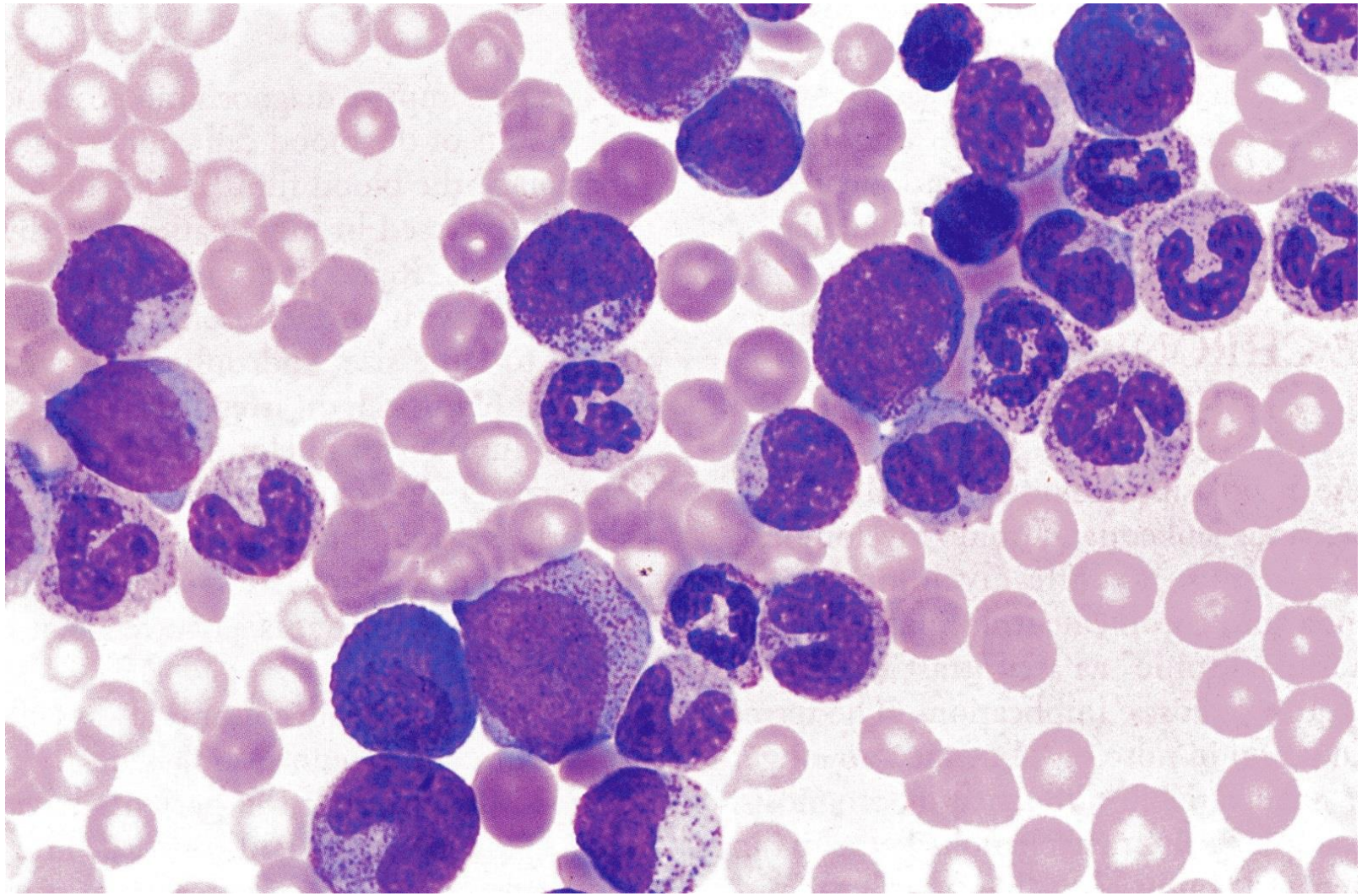






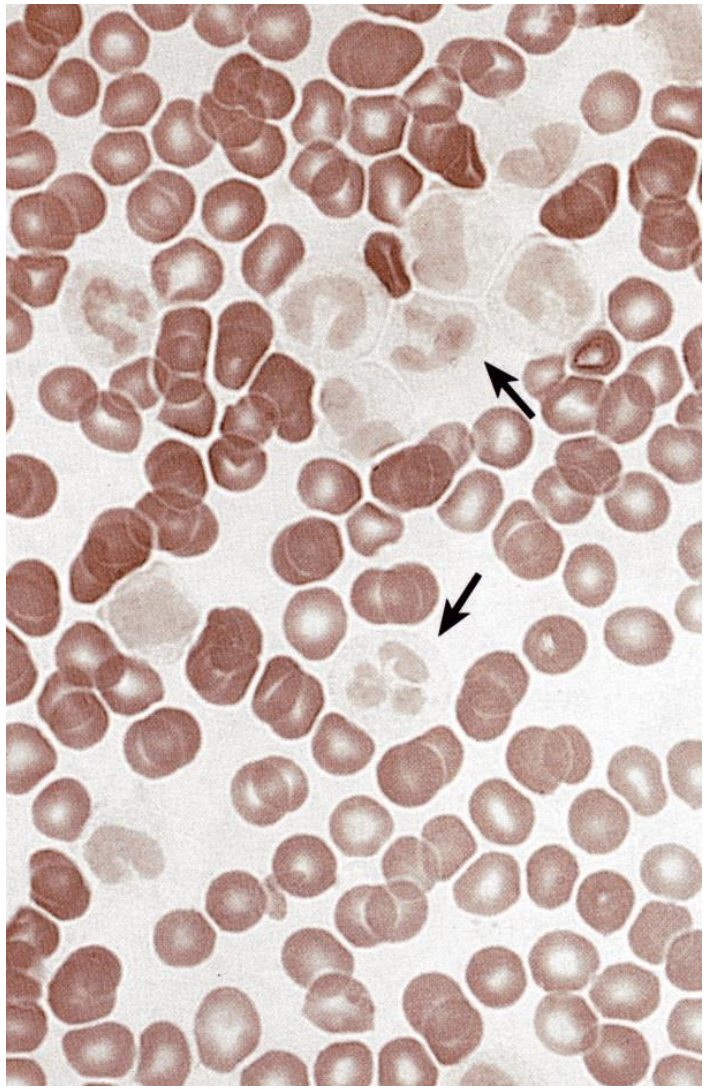
Nature Reviews | Drug Discovery

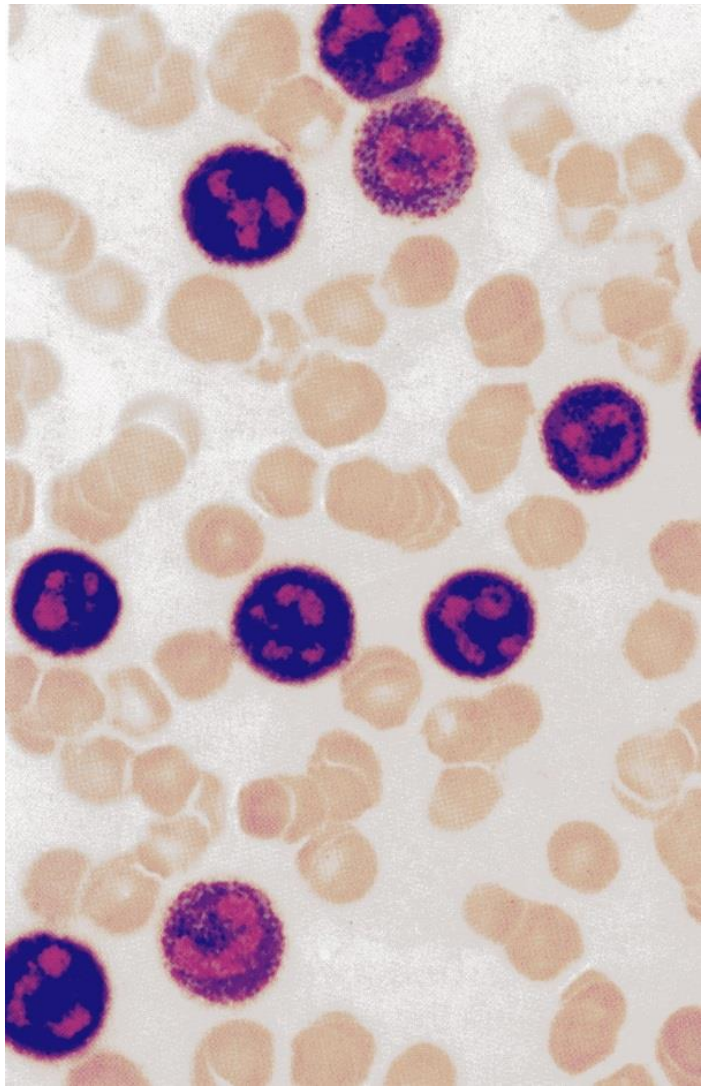
LEUCEMIA MIELOIDE CRONICA: fase cronica



CML: fosfatasi alcalina leucocitaria

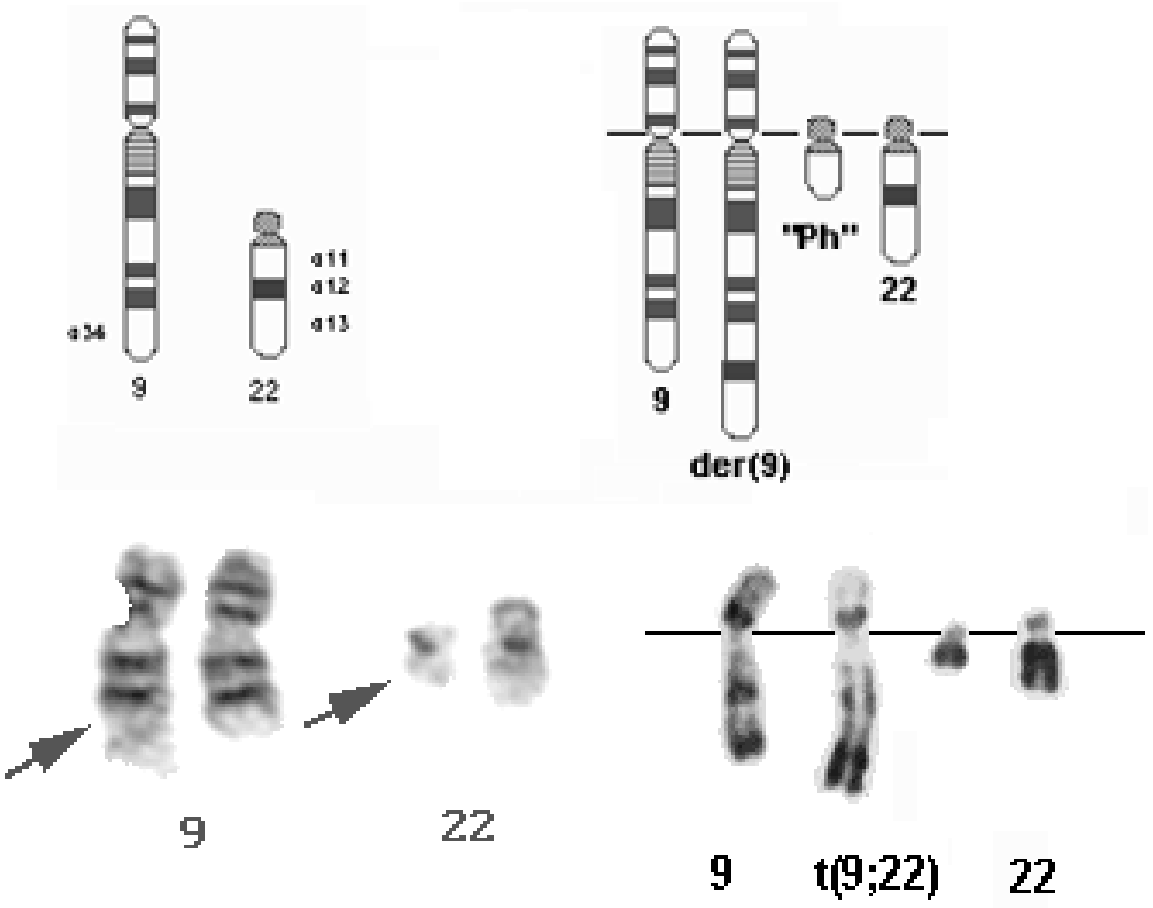
Leucemia mieloide cronica



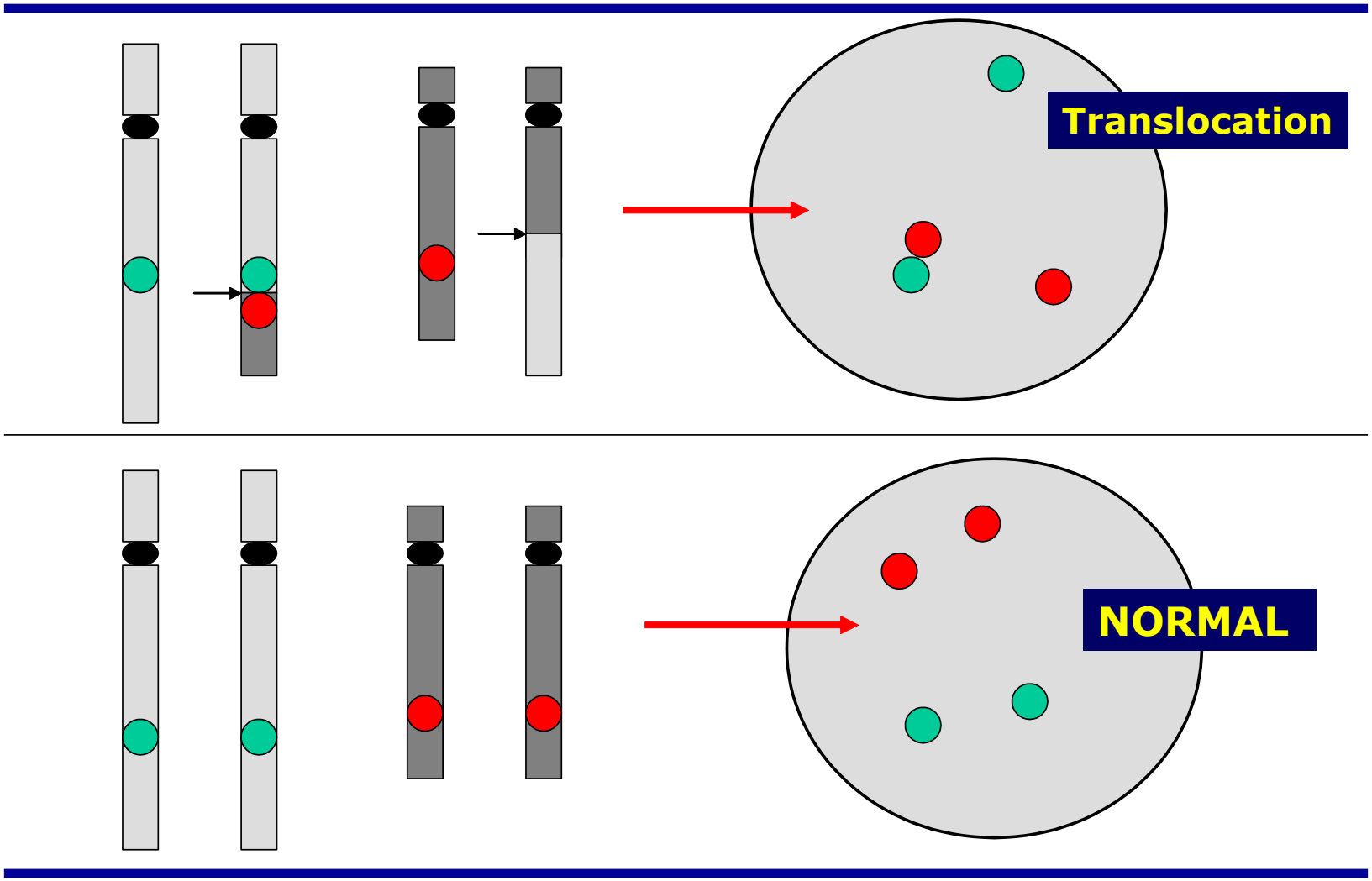


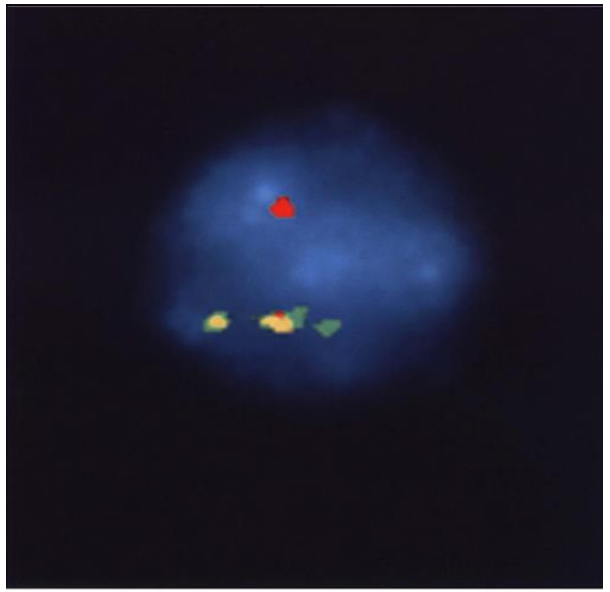
Policitemia vera

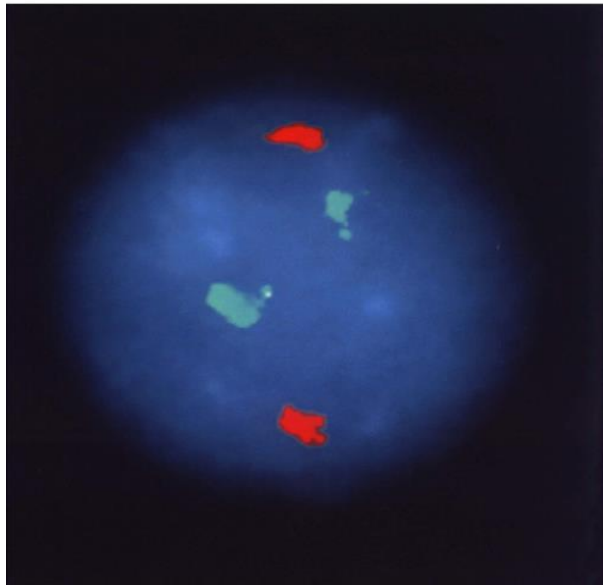
t(9;22): citogenetica



DUAL COLOUR FISH TO DETECT CHROMOSOME TRANSLOCATIONS
Fusion gene detection







Mandatory diagnostic tests for CML

1. **Blood count with blood film differential.**
   - **This will typically show a so-called left shift of the myeloid series with the presence of rare blasts, promyelocytes, myelocytes and metamyelocytes, basophils, and eosinophils.**
   - **these must be accurately quantified as the results contribute to accurate identification of disease stage and prognostic scoring systems.**

2. **Bone marrow aspirate with differential**
   - **to include percentages of blasts, promyelocytes, myelocytes, eosinophils, and basophils.**

3. **Cytogenetics and karyotyping by G banding:**
   - **FISH is not sufficient at diagnosis as it is unable to identify chromosomal abnormalities in addition to the t(9;22) translocation**

4. **Reverse transcriptase PCR for BCR-ABL1 mRNA transcripts.**

Apperley JF. Lancet 2015; 385: 1447–59

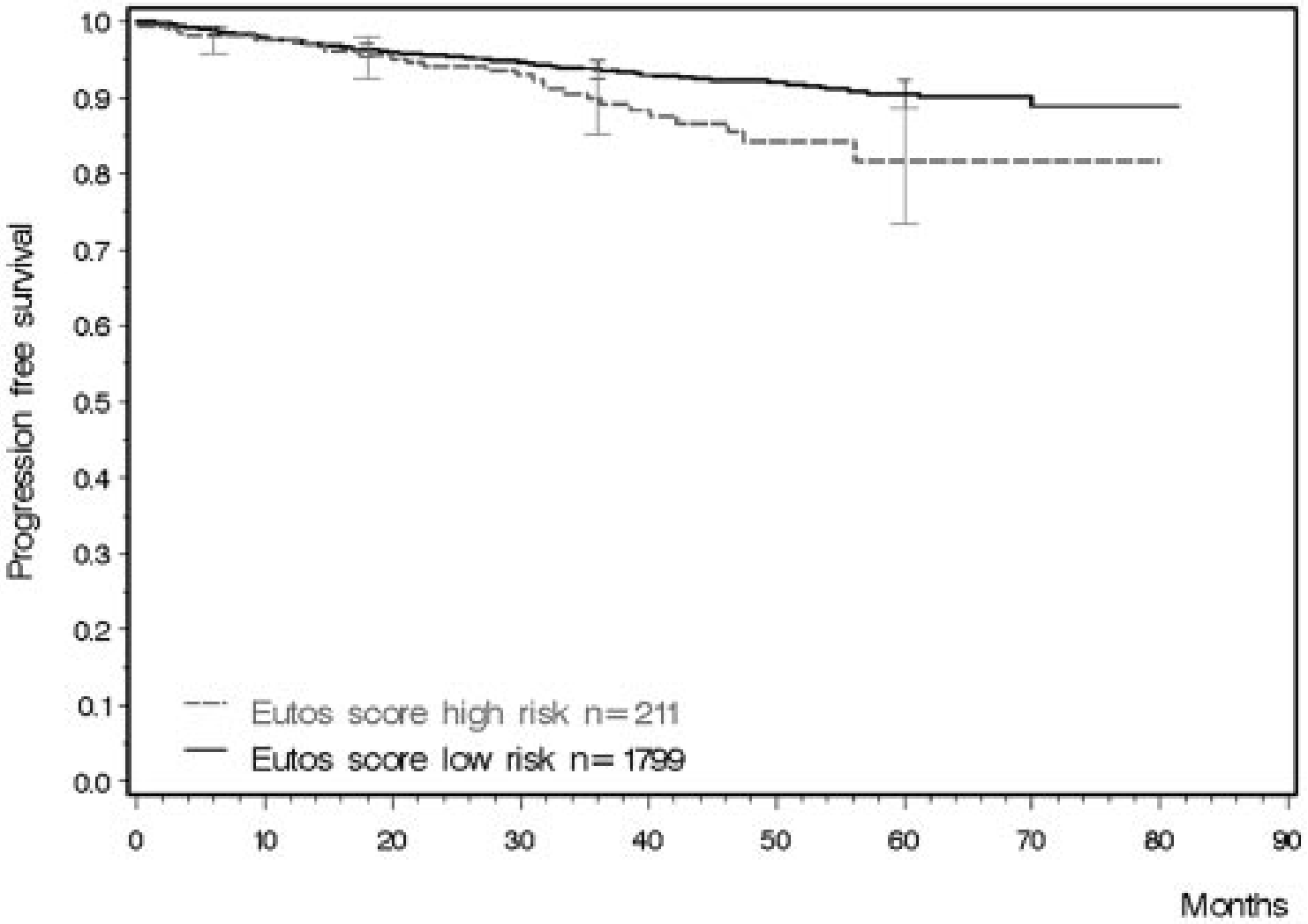
	Sokal[8]*	Hasford[9]*	EUTOS[10] †
Age (years)	0·116 × (age 43·4 years)	0·666 when >50 years	..
Spleen (cm below costal margin)	0·0345 × (spleen size − 7·51)	0·042 × spleen size	..
Platelets x 10^9/L	0·188 × [(plts − 700)^2 − 0·563]	1·0956 when >1500	..
Peripheral blood basophils %	Not included	0·20399 when >3%	7 × %
Peripheral blood eosinophils %	Not included	0·0413×%	4 × spleen
Low risk	<0·8	≤780	≤87
Intermediate risk	0·8–1·2	781–1480	..
High risk	>1·2	>1480	>87

*Calculations for Sokal and Hasford done with the European LeukemiaNet risk calculator. †Calculations for EUTOS done at European LeukemiaNet.

Table 2: Scoring systems validated for parameters at diagnosis for treatment with busulfan (Sokal), interferon α (Hasford), and imatinib (EUTOS)

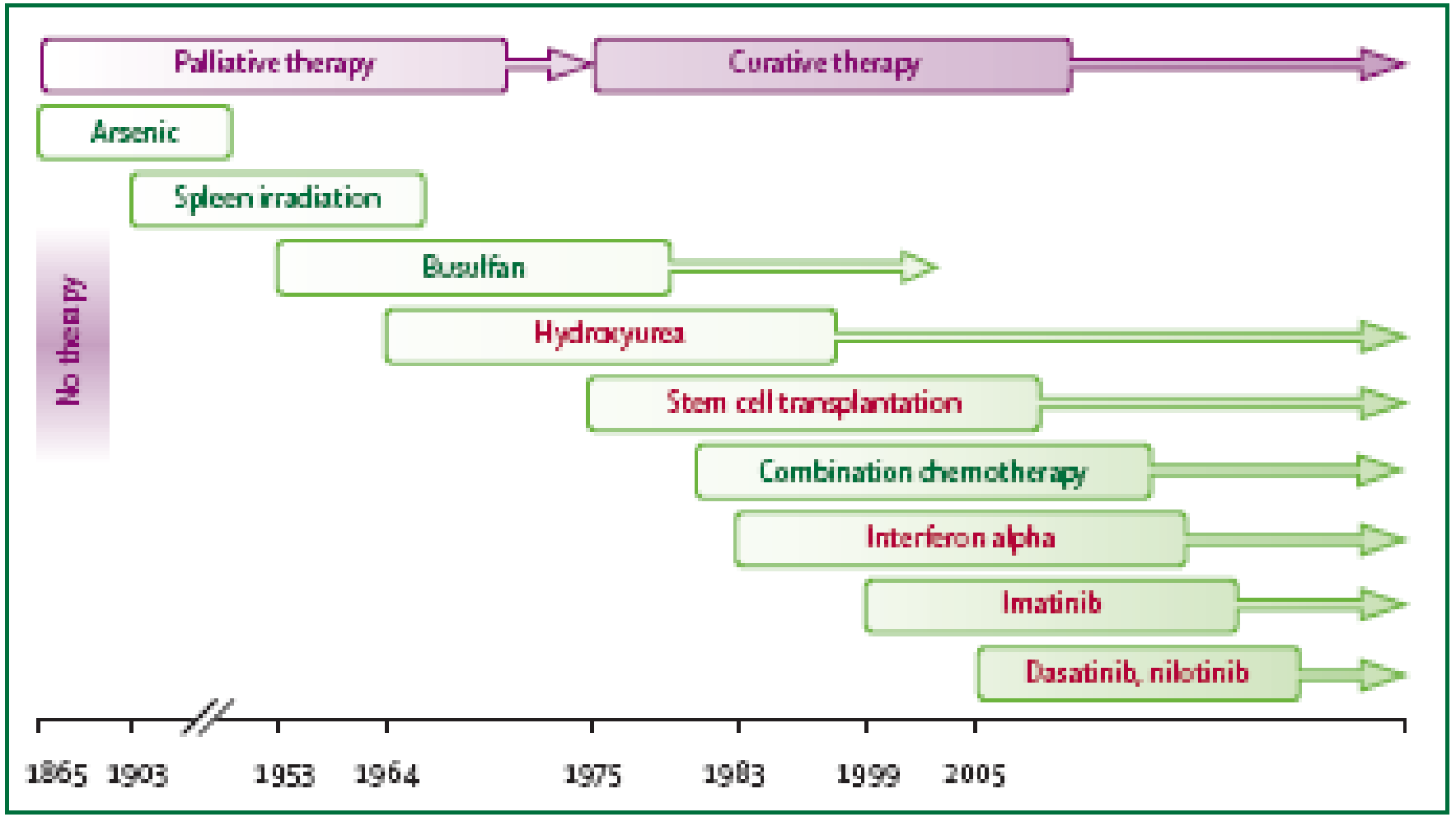
Apperley JF. Lancet 2015; 385: 1447–59

PFS calculated for 2010 pts with follow-up (*P* .0069).



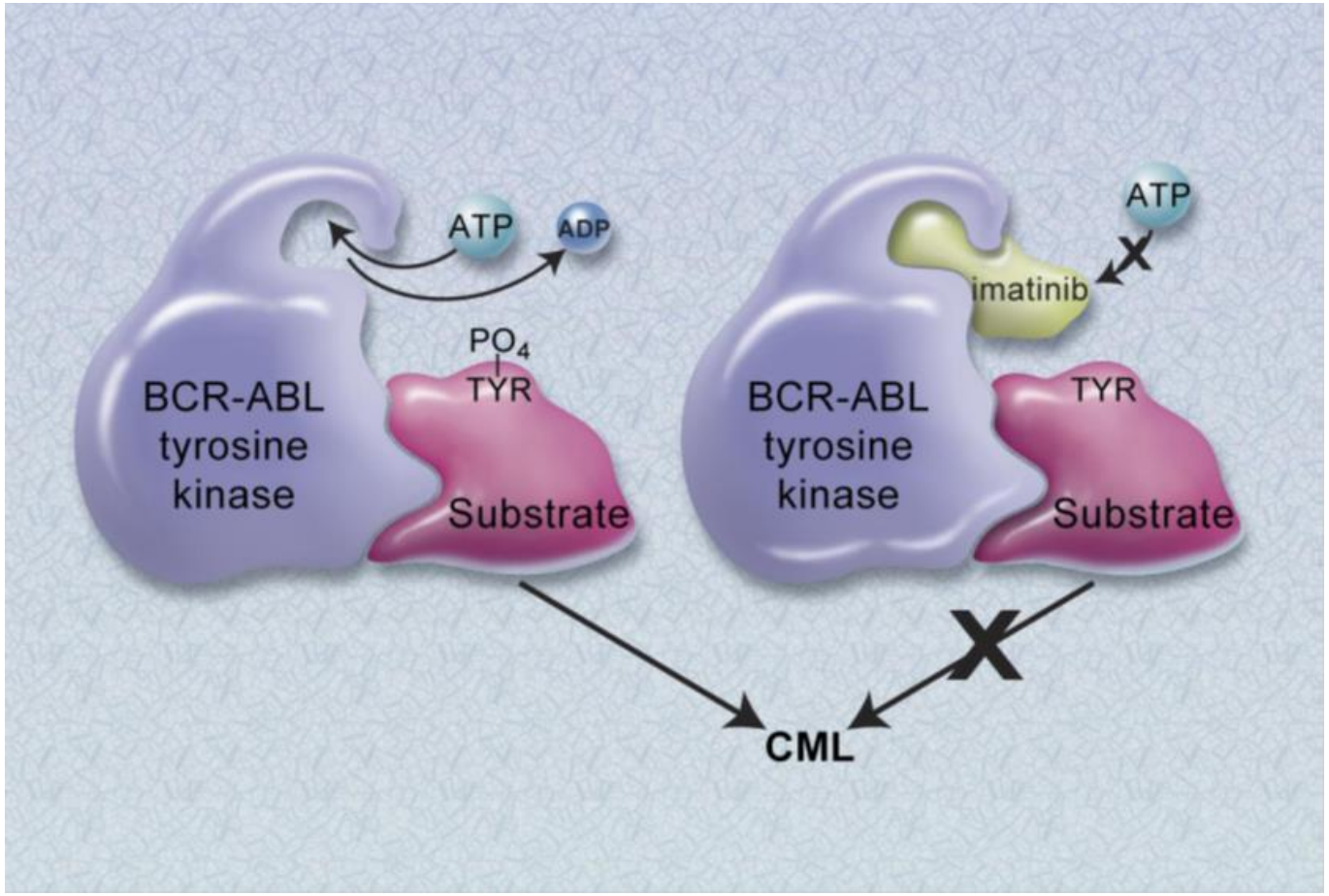
Hasford et al *Blood*. 2011;118(3):686-692)

Development of treatments for CML

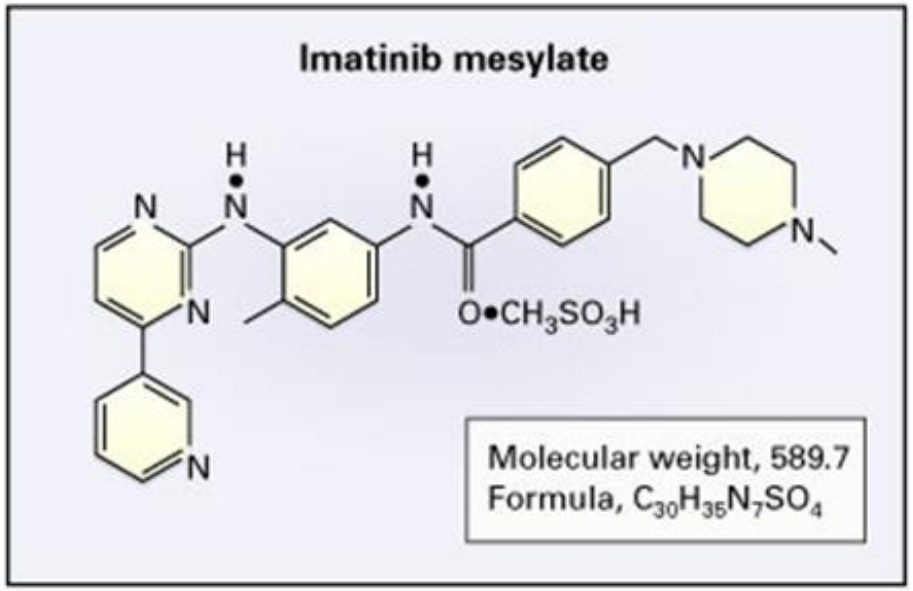


Lancet 2007; 370: 342–50

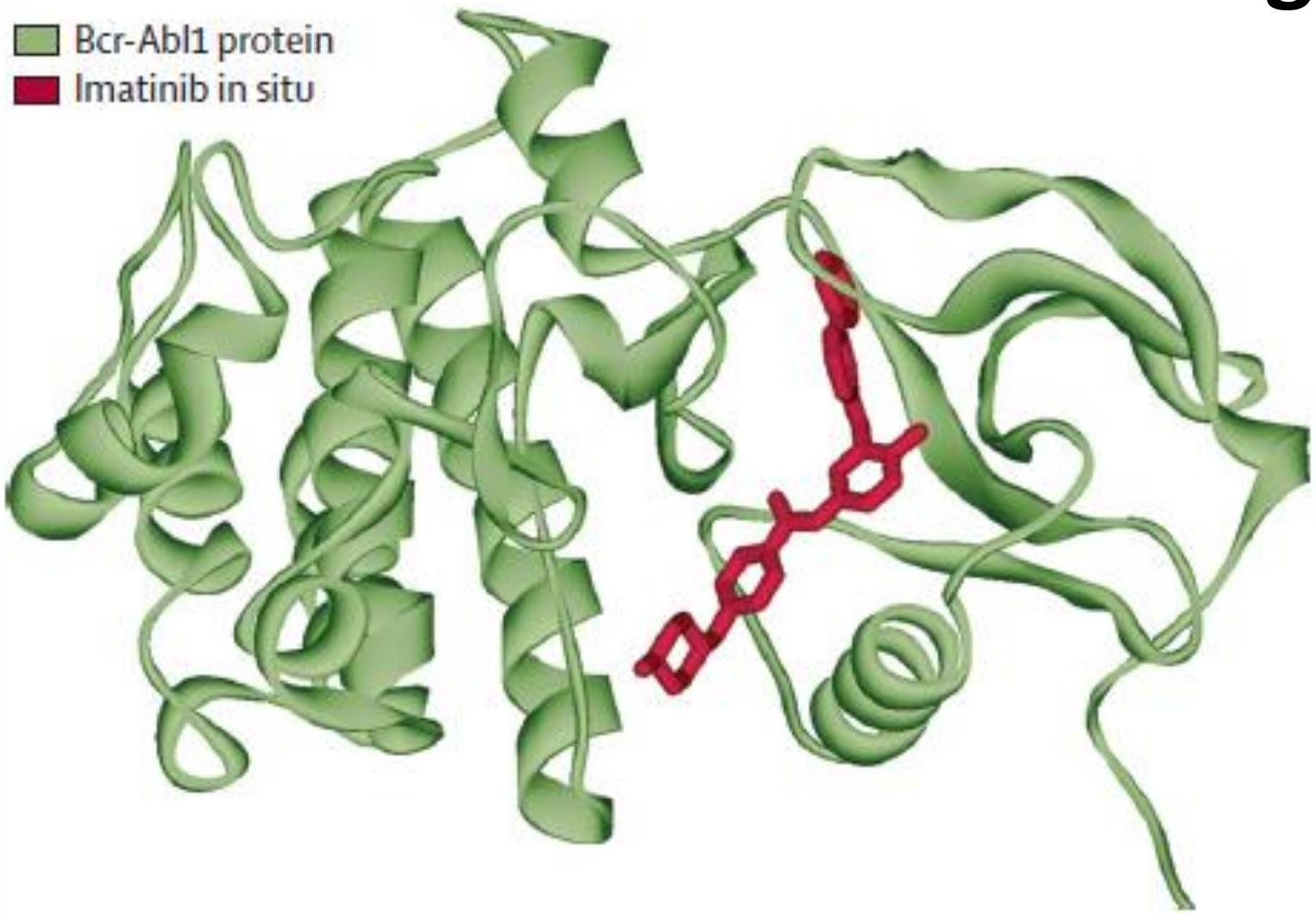
Imatinib Mesylate



Druker BJ. Blood. 2008;112:4808-4817)



Pathogenesis of CML



Schematic of protein with imatinib in ATP binding loop

Apperley JF. Lancet 2015; 385: 1447–59

Approved BCR-ABL TKIs for CML Therapy: Indications

TKI	First Line	Second or Later Lines	AP	BP	Dosing (Adult)
Imatinib	Yes	After failure with interferon	Yes	Yes	CP-CML: 400 PO QD AP-CML or BP-CML: 600 PO QD *(Consider dose escalation in select patients based on response)*
Dasatinib	Yes	Resistance to prior TKIs including imatinib	Yes	Yes	CP-CML: 100 mg PO QD *(No hematologic/cytogenetic response consider escalating to 140 mg PO QD)* AP-CML or BP-CML: 140 mg PO QD
Nilotinib	Yes	Resistance or intolerance to prior TKIs including imatinib	Yes	No	Newly diagnosed: 300 mg PO BID Resistant/intolerant: 400 mg PO BID
Bosutinib	Yes	Previously treated with one or more TKIs where imatinib, dasatinib, and nilotinib are clinically inappropriate	Yes	Yes	Newly diagnosed: 400 mg PO QD Resistant/intolerant: 500 mg PO QD *(Consider dose escalation in select patients based on response)*
Ponatinib	No	Patients with *T315I* mutation or resistance or intolerance to dasatinib or nilotinib and for whom subsequent treatment with imatinib is clinically inappropriate	Yes	Yes	45 mg PO QD Hepatic impairment: 30 mg PO QD

Garcia-Gutierrez. Front Oncol. 2019;9:603.

Slide credit: clinicaloptions.com

FDA-Approved BCR-ABL TKIs for CML Therapy: Efficacy

TKI	Inhibition	Efficacy	Notes
Imatinib	c-ABL, BCR-ABL, PDGF-R, c-KIT	Improved OS, PFS vs IFN-a/LDAC in IRIS	Progression on imatinib due to compliance issues or development of resistance mutations (eg, in ABL kinase domain)
Dasatinib	BCR-ABL, ABL kinase domain point mutants, SRK kinases, other RTKs	Higher rates of CCyR, MMR vs imatinib in DASISION, but PFS and OS not improved	325-fold more potent at inhibiting BCR-ABL vs imatinib
Nilotinib	KIT and PDGFR (weaker than imatinib), BCR-ABL point mutations	Higher rates of MMR vs imatinib in ENESTnd, but PFS and OS not improved	Rationally designed from chemical structure of imatinib: nilotinib has ~ 30-fold greater potency than imatinib, with improved selectivity
Bosutinib	BCR-ABL	Higher rates of and shorter times to MMR and CCyR vs imatinib in BFORE (OS, PFS NE)	Inhibits BCR-ABL with greater potency than imatinib; lacks activity vs PDGFRα/β and c-KIT
Ponatinib	BCR-ABL including T315I-mutant ABL, VEGFR, PDGFR, FGFR, SRC, others.	CCyR 46% and MMR 39% in patients with resistant or intolerant CP-CML in single-arm, open-label multicenter trial	Not indicated for patients with ND CP-CML; only for patients with CML not indicated for any other TKI, or with T315I+ CML.

Kennedy. Curr Hematol Malig Rep. 2018;13:202. Ponatinib PI.

Slide credit: clinicaloptions.com

TKIs in CML: Overview

- The introduction of TKIs for CML has profoundly changed the patient experience and clinical outcomes
  - Life expectancy for most patients with CP-CML is nearing that of the general population
  - Differences between TKIs can be combined with disease-specific and patient-specific factors to individualize treatment

Bosutinib PI. 2017. Cortes. J Clin Oncol. 2018;36:231.

Slide credit: clinicaloptions.com

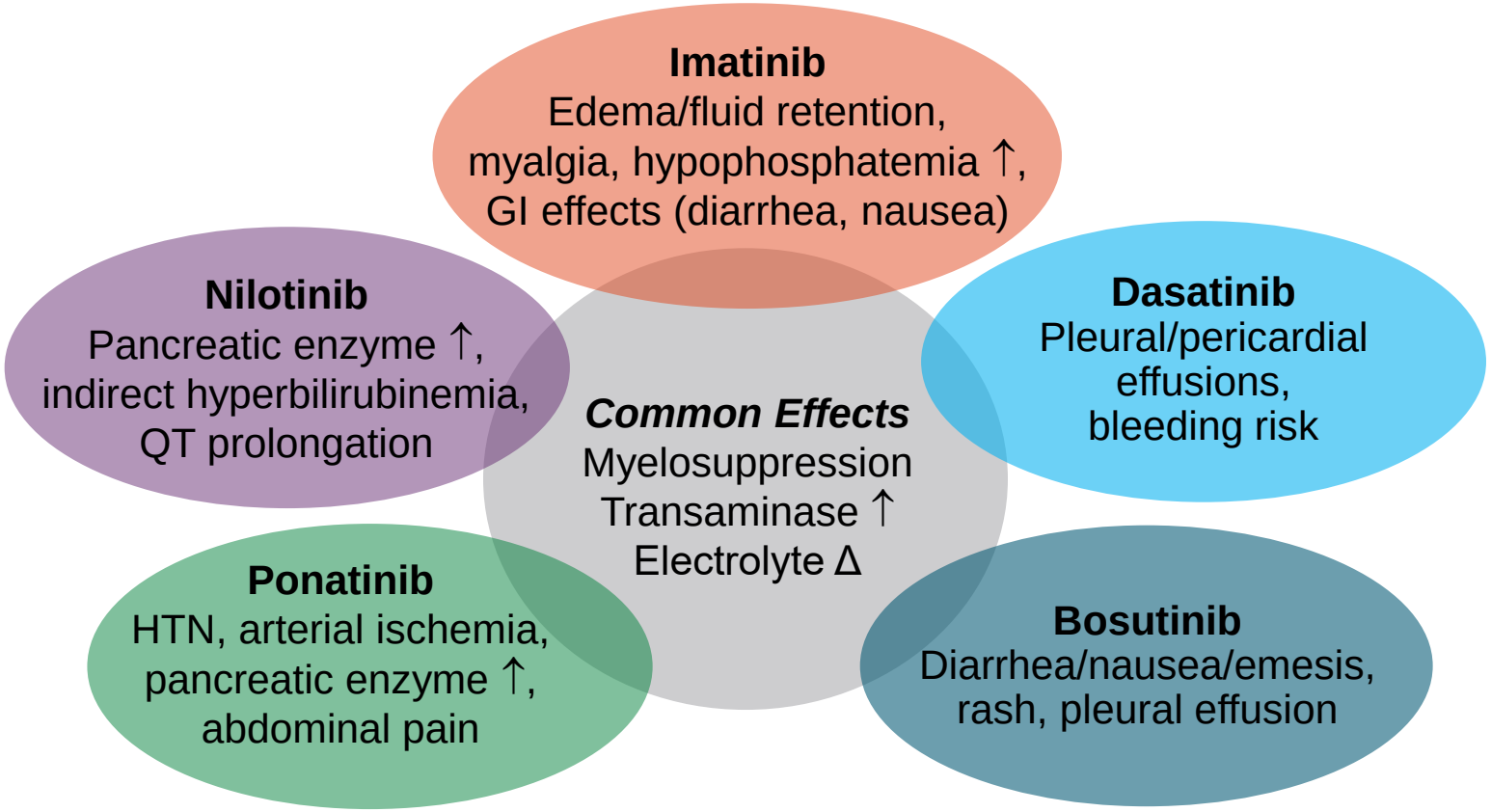
TKIs in CML: Overview

- Considerations regarding initial TKI treatment
  - Monitoring of response, management of toxicities, and adherence are essential to maximize benefit
  - A multidisciplinary team approach with an engaged patient is critical, given that CML is a chronic disease with multiple decision points and the need for changes to occur quickly
  - Optimal treatment selection, adherence, and monitoring will lead to the need to consider:
    - How to safely discontinue treatment
    - Switching to a new treatment due to disease progression or intolerance

Bosutinib PI. 2017. Cortes. J Clin Oncol. 2018;36:231.

Slide credit: clinicaloptions.com

Toxicity Spectrum of TKIs in CML



Imatinib PI. Dasatinib PI. Nilotinib PI. Bosutinib PI. Ponatinib PI.

Slide credit: clinicaloptions.com

Individualizing Therapy in CML: TKIs for Patients With Specific History or Comorbidities

- When selecting first-line TKI, consider age, medical history, comorbidities, and AE profiles

Comorbidity	Considerations for TKI Selection
Risk for pleural effusions (eg, lung disorders, uncontrolled hypertension)	Avoid dasatinib
Pulmonary arterial hypertension	Avoid dasatinib
Risk of hemorrhagic complications (eg, patients taking anti-coagulants)	Increased with dasatinib, imatinib
Uncontrolled diabetes mellitus	Use nilotinib with caution (and only on empty stomach)
Coronary, cerebrovascular, or peripheral arterial disease	Use nilotinib with caution
Hepatic impairment	Dose reduce bosutinib (200 mg QD)
Renal impairment	Dose reduce imatinib (50% decrease in recommended starting dose); use imatinib with caution in severe cases

Hochhaus. Ann Oncol. 2017;28(suppl 4):iv41. Bosutinib PI. Imatinib PI.

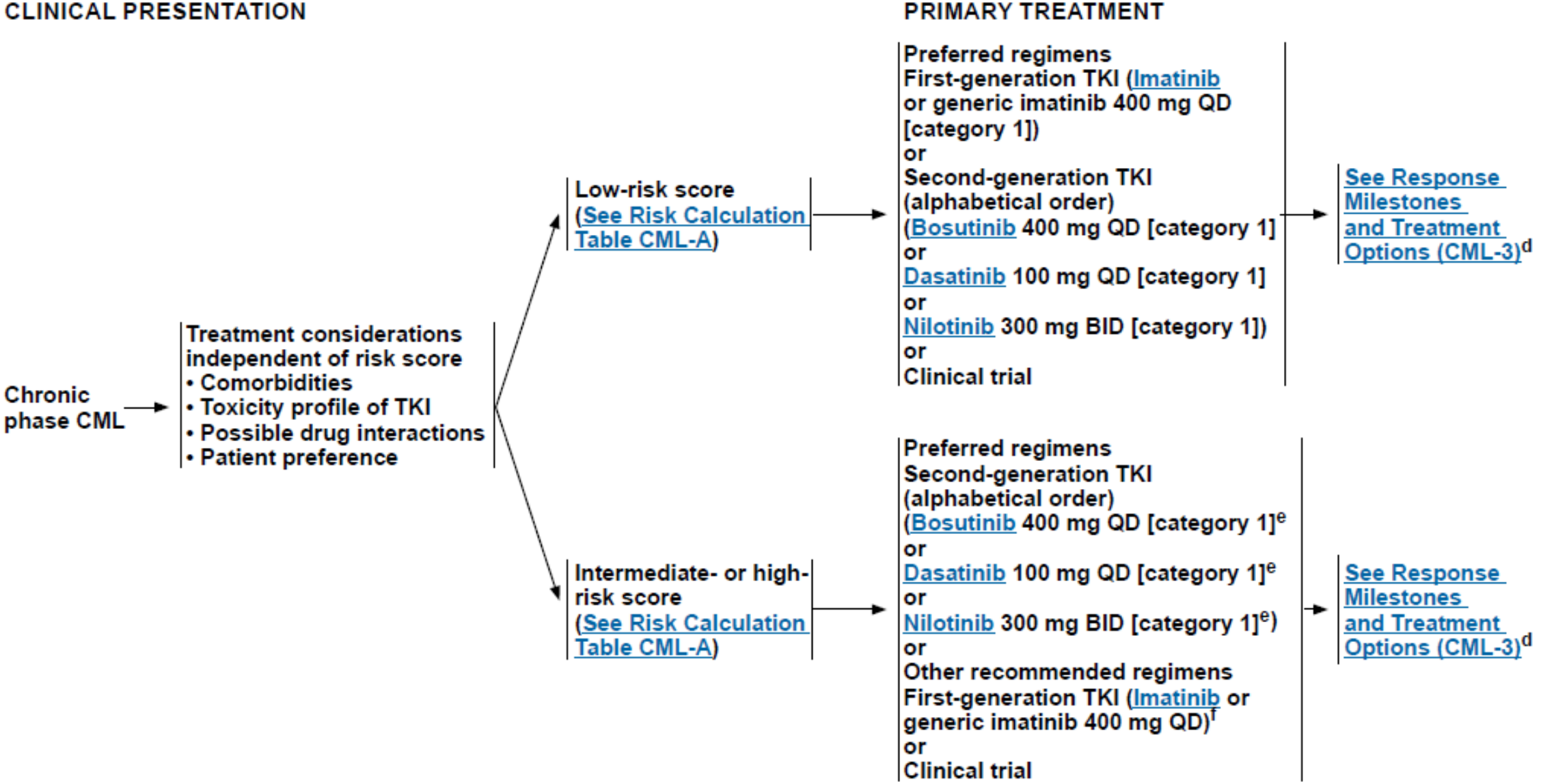
Slide credit: clinicaloptions.com

	Imatinib		Dasatinib		Nilotinib		Bosutinib		Ponatinib	
	All grades	Grade 3/4	All grades	Grade 3/4	All grades	Grade 3/4	All grades	Grade 3/4	All grades	Grade 3/4
Fatigue	++++	+	+++	+	++++	-	NR	NR	++++	++
Rash	++++	++	+++	+	++++	-	++++	++	++++	++
Headache	+++	-	++++	-	++++	-	++++	++	++++	++
Myalgia and arthralgia	+++++	-	++++	-	NR	NR	++	-	++++	++
Bone pain	+++	++	NR	NR	NR	NR	++	-	NR	NR
Diarrhoea	++++	++	++++	+	+++	+	+++++	++++	NR	NR
Nausea	++++	-	++++	-	+++	+	++++	++	++++	+
Vomiting	+++	-	+++	-	++	-	++++	++	NR	NR
Abdominal pain	++	-	NR	NR	NR	NR	++++	++	++++	+++
Pancreatitis	+	+	NR	NR	++	++	NR	NR	+++	+++
Bleeding events (GI, CNS)	+	+	++	++	++	+	NR	NR	NR	NR
Oedema	++++	++	++++	++	+++	-	+++	++	NR	NR
Pleural effusion	++	+	++++	++	++	+	NR	NR	NR	NR
PAH	NR	NR	+	+	NR	NR	NR	NR	NR	NR
QT prolongation	+	NK	++	NK	++	NK	NR	NR	NR	NR
Hypertension	NR	NR	NR	NR	NR	NR	NR	NR	+++	++
PAOD	-	-	NR	NR	++	++	NR	NR	++++	++++
Elevated lipase	++++	+++	NG	-	++++	+++	++++	+++	++++	++++
Elevated ALT	++++	++	NG	+	+++++	+++	+++++	++++	++++	++
Low phosphate	+++++	++++	NG	+++	++++	+++	++++	++	NR	NR
Raised glucose	-	-	-	-	++++	+++	-	-	NR	NR
Anaemia	+++++	+++	+++++	++++	++++	++	+++++	+++	+++	+++
Neutropenia	+++++	++++	+++++	++++	++++	+++	++++	++++	++++	++++
Thrombocytopenia	+++++	++++	+++++	++++	++++	+++	++++++	++++	++++	++++
Abn platelet function	+++++	NK	+++++	NK	-	-	++++	NK	NR	NR
LGL expansion	NR	NR	++++	NK	NR	NR	NR	NR	NR	NR

Data derived from studies of first line use with the exception of ponatinib (so far used only as second or subsequent line) and rare events such as PAH, PAOD, and abnormal platelet function.[72,78,79,82,85,86,92,93] +=<1% of patients. ++=1–5%. +++=5–10%. ++++=10–50%. +++++=50–100% =specifically reported as absent. NR=not reported. GI=gastrointestinal. PAH=pulmonary arterial hypertension. NK=effect of side-effect not known. PAOD=peripheral arterial occlusive disease. NG=data not given. ALT=alanine transaminase. Abn=abnormal. LGL=large granular lymphocytes.

Table 4: **Most frequently reported side-effects of tyrosine-kinase inhibitors**

Apperley JF. Lancet 2015; 385: 1447–59


CLINICAL PRESENTATION
PRIMARY TREATMENT
Chronic phase CML
Treatment considerations independent of risk score
• Comorbidities
• Toxicity profile of TKI
• Possible drug interactions
• Patient preference
Low-risk score (See Risk Calculation Table CML-A)
Preferred regimens
First-generation TKI (Imatinib or generic imatinib 400 mg QD [category 1])
or
Second-generation TKI (alphabetical order)
(Bosutinib 400 mg QD [category 1]
or
Dasatinib 100 mg QD [category 1]
or
Nilotinib 300 mg BID [category 1])
or
Clinical trial
See Response Milestones and Treatment Options (CML-3)[d]
Intermediate- or high-risk score (See Risk Calculation Table CML-A)
Preferred regimens
Second-generation TKI (alphabetical order)
(Bosutinib 400 mg QD [category 1][e]
or
Dasatinib 100 mg QD [category 1][e]
or
Nilotinib 300 mg BID [category 1][e])
or
Other recommended regimens
First-generation TKI (Imatinib or generic imatinib 400 mg QD)[f]
or
Clinical trial
See Response Milestones and Treatment Options (CML-3)[d]

CRITERIA FOR HEMATOLOGIC, CYTOGENETIC, AND MOLECULAR RESPONSE AND RELAPSE

Complete hematologic response[1]

- Complete normalization of peripheral blood counts with leukocyte count <10 x 10^9/L
- Platelet count <450 x 10^9/L
- No immature cells, such as myelocytes, promyelocytes, or blasts in peripheral blood
- No signs and symptoms of disease with resolution of palpable splenomegaly

Cytogenetic response[2,3]

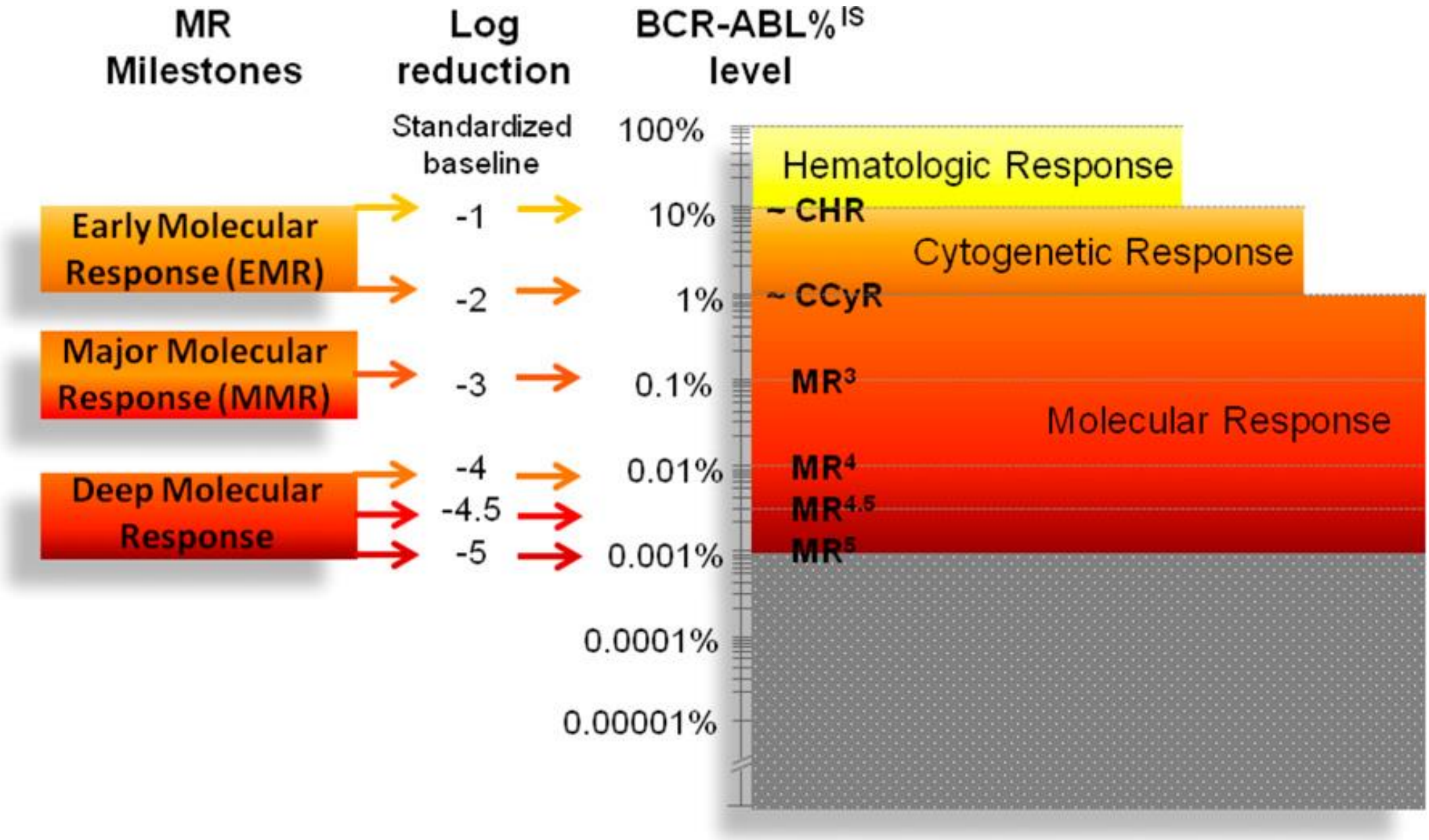
- Complete cytogenetic response (CCyR) - No Ph-positive metaphases[4]
- Major cytogenetic response (MCyR) - 0%–35% Ph-positive metaphases
- Partial cytogenetic response (PCyR) - 1%–35% Ph-positive metaphases
- Minor cytogenetic response - >35%–65% Ph-positive metaphases

Molecular response[5,6]

- Early molecular response (EMR) - *BCR-ABL1* (IS) ≤10% at 3 and 6 months
- Major molecular response (MMR) - *BCR-ABL1* (IS) ≤0.1% or ≥3-log reduction in *BCR-ABL1* mRNA from the standardized baseline, if qPCR (IS) is not available
- Complete molecular response (CMR) is variably described, and is best defined by the assay's level of sensitivity (eg, MR4.5)

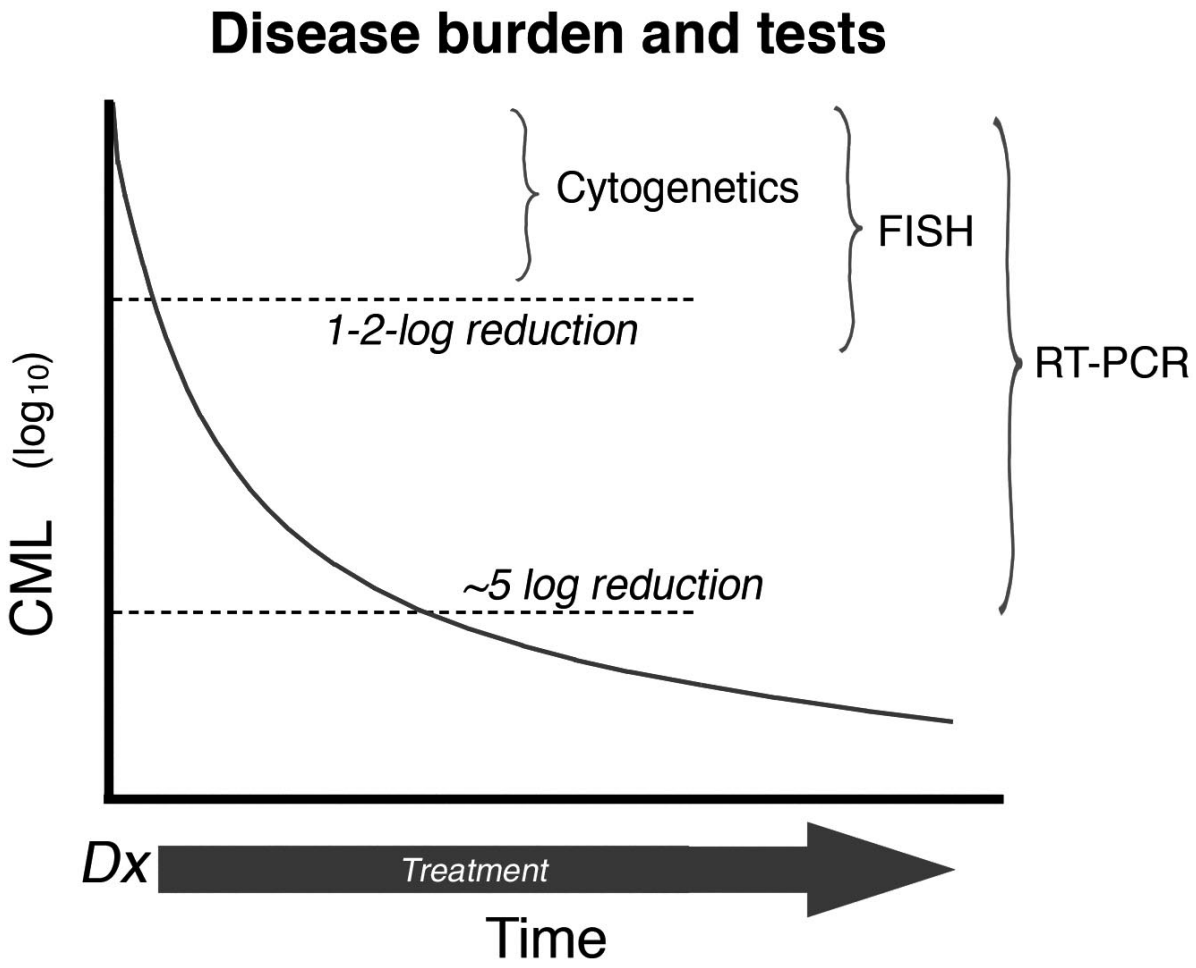
Relapse

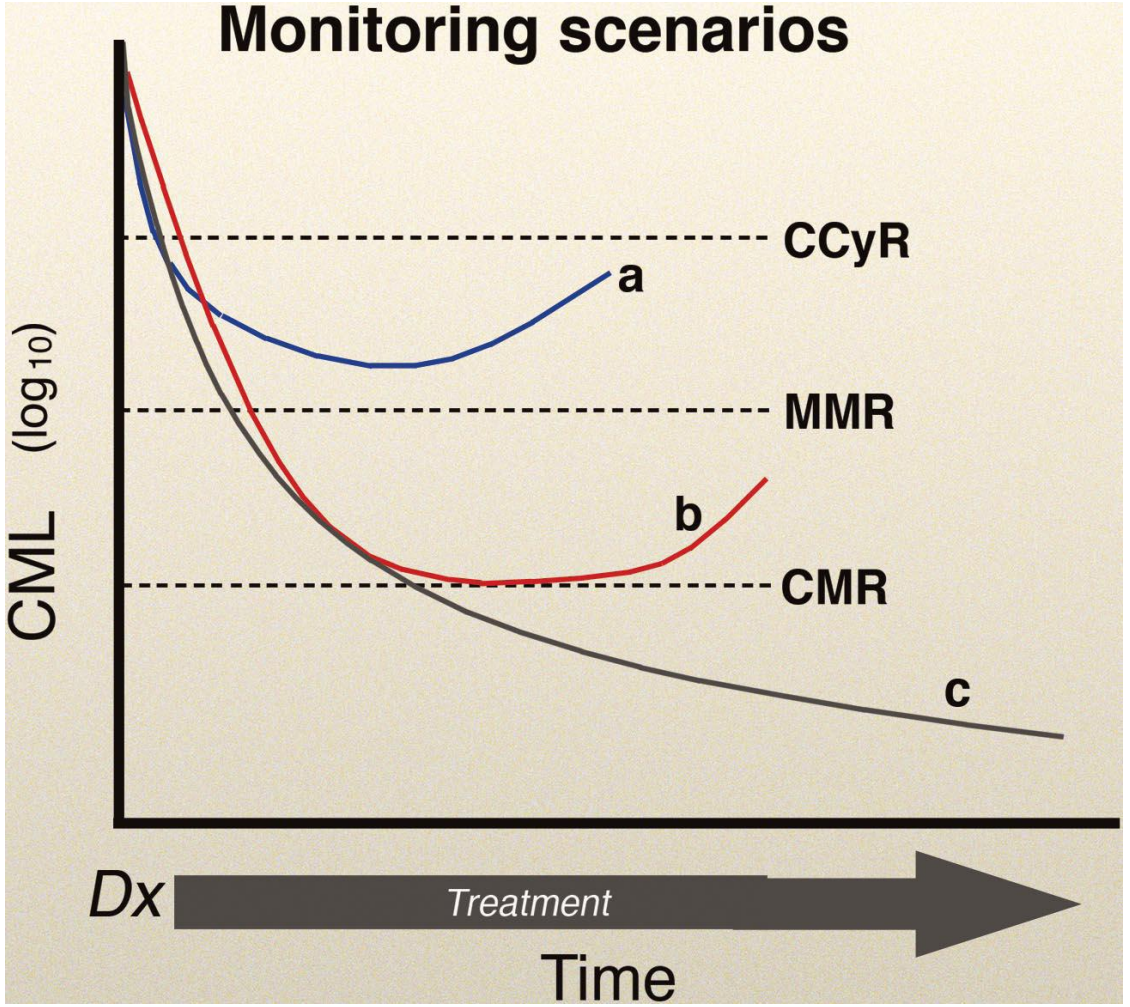
- Any sign of loss of response (defined as hematologic or cytogenetic relapse)
- 1-log increase in *BCR-ABL1* transcript levels with loss of MMR should prompt bone marrow evaluation for loss of CCyR but is not itself defined as relapse (eg, hematologic or cytogenetic relapse)



Soverini et al. The Oncologist 2016

Disease burden and tests.





Blood. 2009;114:3376-3381

When to Consider Changing Therapy

Response Category	Clinical Actions	When to Take Clinical Actions				
		Mo 3	Mo 6	Mo 12	After Mo 18	Anytime
Warning	▪ Monitor carefully ▪ Prepare to consider changing therapy ▪ Assess adherence ▪ Assess for drug–drug interactions	▪ Ph+: 36% to 95% ▪ *BCR-ABL1:* > 10%	▪ Ph+: 1% to 65% ▪ *BCR-ABL1*: 1% to 10%	▪ *BCR-ABL1*: 0.1% to 10%	▪ *BCR-ABL1*: 0.1% to 1%	
Failure	▪ Switch to alternate TKI ▪ Assess for alloSCT ▪ BM exam to assess CML phase, clonal evolution ▪ Evaluate *BCR-ABL1* mutational profile	▪ No complete hematologic response ▪ Ph+ > 95%	▪ Ph+: > 35% ▪ *BCR-ABL1"* > 10%	▪ Ph+: ≥ 1% ▪ *BCR-ABL1:* > 1%		▪ Relapse ▪ Loss of MMR

Hochhaus. Ann Oncol. 2017;28(suppl 4):iv41.

Slide credit: clinicaloptions.com

MONITORING RESPONSE TO TKI THERAPY AND MUTATIONAL ANALYSIS

Test	Recommendation
Bone marrow cytogenetics[1]	• At diagnosis • Failure to reach response milestones • Any sign of loss of response (defined as hematologic or cytogenetic relapse)
qPCR using IS	• At diagnosis • Every 3 months after initiating treatment. After *BCR-ABL1* (IS) ≤1% (>0.1%–1%) has been achieved, every 3 months for 2 years and every 3–6 months thereafter • If there is 1-log increase in *BCR-ABL1* transcript levels with MMR, qPCR should be repeated in 1–3 months
BCR-ABL1 kinase domain mutation analysis	• Chronic phase ‣ Failure to reach response milestones ‣ Any sign of loss of response (defined as hematologic or cytogenetic relapse) ‣ 1-log increase in *BCR-ABL1* transcript levels and loss of MMR • Disease progression to accelerated or blast phase[2]

EARLY TREATMENT RESPONSE MILESTONES[d,g]

BCR-ABL1 (IS)	3 months	6 months	12 months[h]	≥15 months
>10%[i]	YELLOW	RED		
>1%–10%	GREEN		YELLOW	RED
≤1%	GREEN			

COLOR	CONCERN	CLINICAL CONSIDERATIONS	SECOND-LINE TREATMENT
RED	TKI-resistant disease	• Evaluate patient compliance and drug interactions • Consider mutational analysis	Switch to alternate TKI (CML-5) and evaluate for allogeneic HCT
YELLOW	Possible TKI resistance	• Evaluate patient compliance and drug interactions • Consider mutational analysis • Consider bone marrow cytogenetic analysis to assess for MCyR at 3 mo or CCyR at 12 mo	Switch to alternate TKI (CML-5) or Continue same TKI (other than imatinib) (CML-F)[j] or Increase imatinib dose to a max of 800 mg and Consider evaluation for allogeneic HCT
GREEN	TKI-sensitive disease	• Monitor response (CML-C) and side effects	Continue same TKI (CML-F)[k]

In vitro sensitivity of unmutated BCR-ABL1 and of some more frequent BCR-ABL1 kinase domain mutants to imatinib, nilotinib, dasatinib, bosutinib, and ponatinib

BCR-ABL1	Imatinib IC_{50}, range (nM)	Nilotinib IC_{50}, range (nM)	Dasatinib IC_{50}, range (nM)	Bosutinib IC_{50} (nM)	Ponatinib IC_{50} (nM)
Unmutated	260-678	<10-25	0.8-1.8	41.6	0.5
M244V*	1600-3100	38-39	1.3	147.4	2.2
L248V	1866-10 000	49.5-919	9.4	NA	NA
G250E*	1350 to >20 000	48-219	1.8-8.1	179.2	4.1
Q252H	734-3120	16-70	3.4-5.6	33.7	2.2
Y253F	>6400-8953	182-725	6.3-11	40	2.8
Y253H*	>6400-17 700	450-1300	1.3-10	NA	6.2
E255K*	3174-12 100	118-566	5.6-13	394	14
E255V	6111-8953	430-725	6.3-11	230.1	36
D276G	1147	35.3	2.6	25	NA
E279K	1872	36.5-75	3	39.7	NA
V299L	540-814	23.7	15.8-18	1086	NA
F311I	480-1300	23	1.3	NA	NA
T315I*	>6400 to >20 000	697 to >10 000	137 to >1000	1890	11
T315A	125	N.A.	760	NA	1.6
F317L*	810-7500	39.2-91	7.4-18	100.7	1.1
F317V	500	350	NA	NA	10
M351T*	880-4900	7.8-38	1.1-1.6	29.1	1.5
F359V*	1400-1825	91-175	2.2-2.7	38.6	10
V379I	1000-1,630	51	0.8	NA	NA
L384M*	674-2800	39-41.2	4	19.5	NA
L387M	1000-1100	49	2	NA	NA
H396R*	1750-5400	41-55	1.3-3	33.7	NA
H396P	850-4300	41-43	0.6-2	18.1	1.1
F486S	2728-9100	32.8-87	5.6	96.1	NA
Plasma drug concentration					
Cmin	2062 ± 1334	1923 ± 1233	5.5 ± 1.4	268 (30-1533)	64.3 ± 29.2
Cmax	4402 ± 1272	2329 ± 772	133 ± 73.9	392 (80-1858)	145.4 ± 72.6

The half maximal inhibitory concentration (IC_{50}) shown here is universally regarded as a measure of the degree of sensitivity of a *BCR-ABL1* mutant to a given TKI and is experimentally determined by quantifying the TKI concentration required to reduce by 50% viability of a Ba/F3 mouse lymphoblastoid cell line engineered to express that mutant form of *BCR-ABL1*. The table lists all of the *BCR-ABL1* mutants for which the IC_{50} values of at least 2 TKIs are available. For imatinib, dasatinib, and nilotinib, ranges of IC_{50} values were provided when differences in IC_{50} values reported by different studies were observed (reviewed in Baccarani et al[5]). For bosutinib and ponatinib, IC_{50} values come from a single study each.[68,71] Plasma drug concentration is also given in nM. Values of plasma drug concentration are mean ± standard deviation for imatinib (400 mg once daily), nilotinib (300 mg twice daily), dasatinib (100 mg once daily), and ponatinib (45 mg once daily), and median (range) for bosutinib (500 mg once daily).[34,50,72-75]

NA, not available.

*Representative of the 10 most frequent mutations.[56,59]

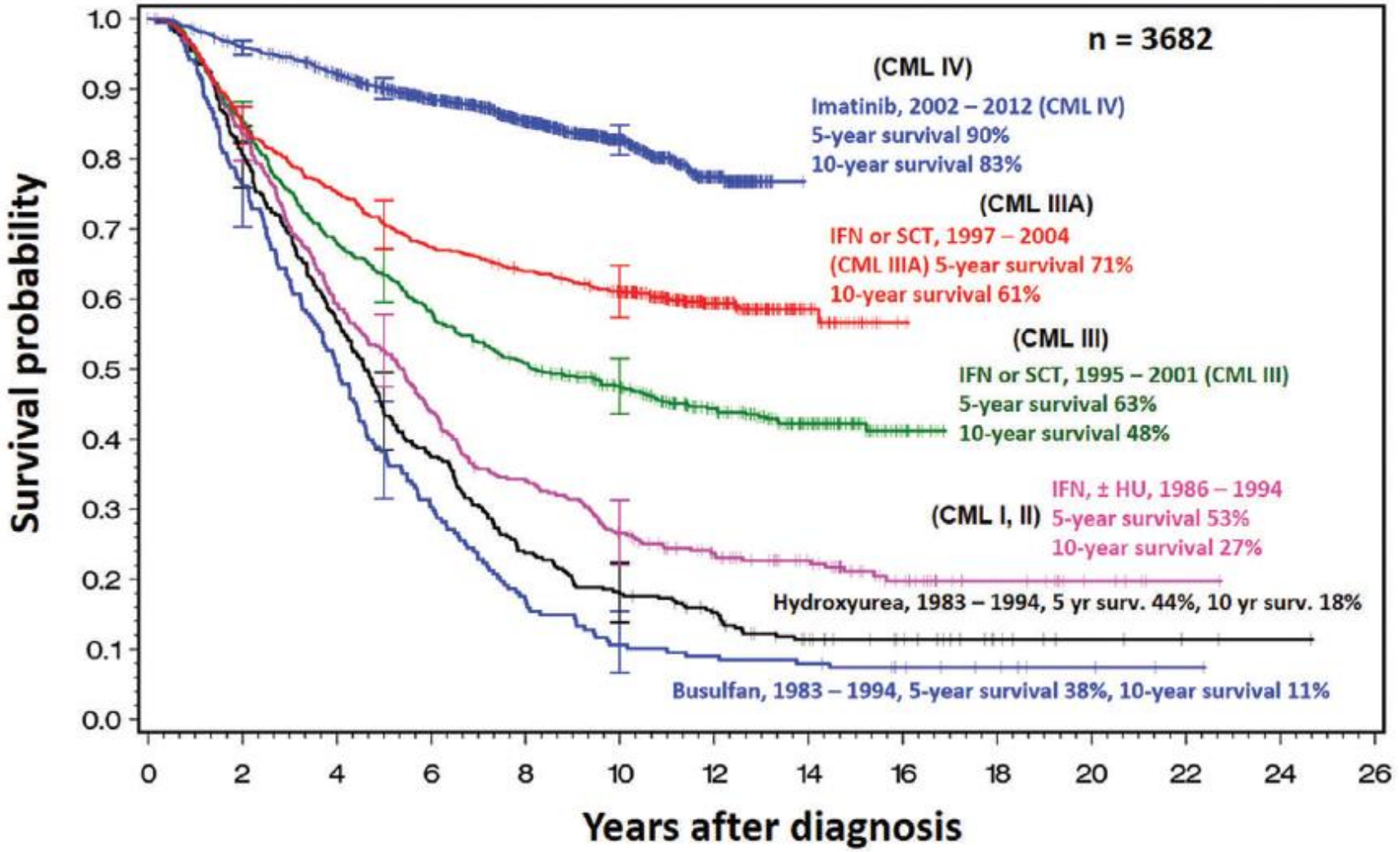
Baccarani et al. Blood. 2013;122(6):872-884)

TREATMENT RECOMMENDATIONS BASED ON *BCR-ABL1* MUTATION PROFILE

- **Patients with disease resistant to primary treatment with imatinib should be treated with bosutinib, dasatinib, or nilotinib in the second-line setting, taking into account *BCR-ABL1* mutation status.**
- **Patients with disease resistant to primary treatment with bosutinib, dasatinib, or nilotinib can be treated with an alternate TKI (other than imatinib) in the second line setting, taking into account *BCR-ABL1* mutation status. The durability of these responses is frequently limited.**
- **The table below lists the *BCR-ABL1* mutations that should NOT be treated with bosutinib, dasatinib or nilotinib in the second-line setting.**

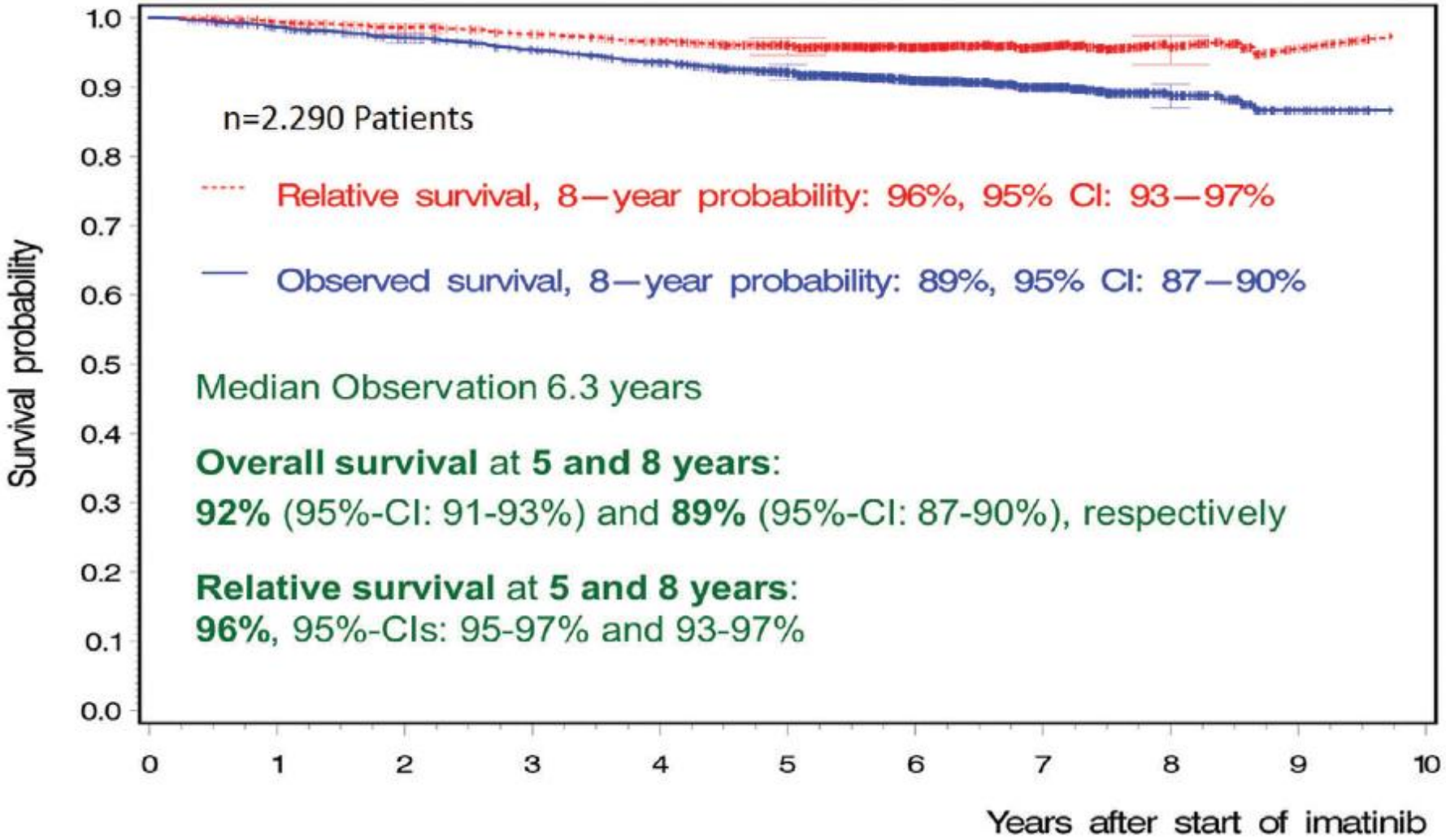
THERAPY	CONTRAINDICATED mutations[o]
Bosutinib	*T315I, V299L, G250E or F317L*[p]
Dasatinib	*T315I/A, F317L/V/I/C* or *V299L*
Nilotinib	*T315I, Y253H, E255K/V*, or *F359V/C/I or G250E*
Ponatinib,[q] Omacetaxine,[r] allogeneic HCT (CML-6), or clinical trial	None

Survival with CML in five consecutive randomized studies of the German CML Study Group since 1983; update 2016.



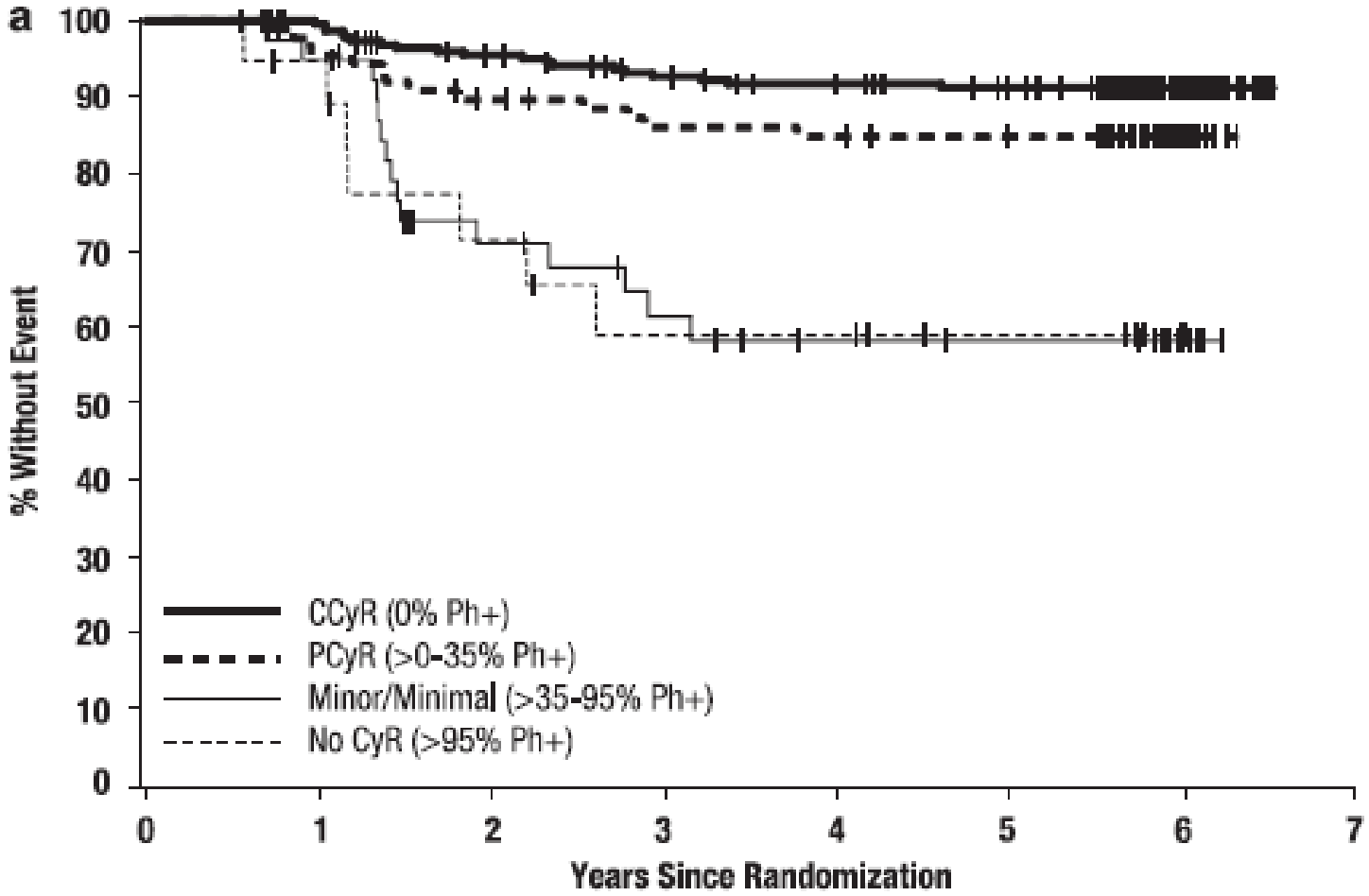
Rüdiger Hehlmann. Haematologica. 2016;101:657-9

Relative and overall survival of 2290 CML patients from the EUTOS Study for CML treated with imatinib



Rüdiger Hehlmann. Haematologica. 2016;101:657-9

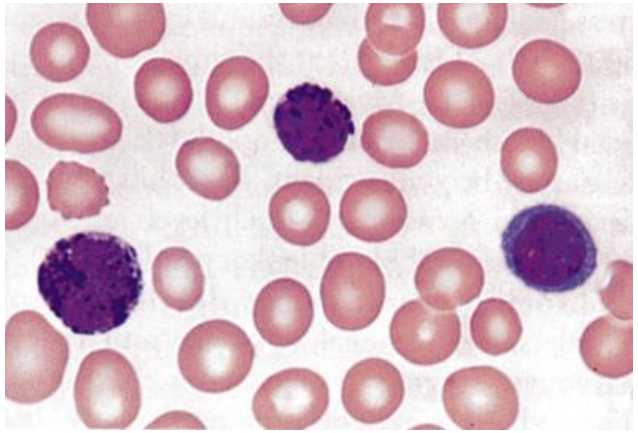
Event-free survival by level of cytogenetic response at 6 months after the initiation of imatinib treatment.

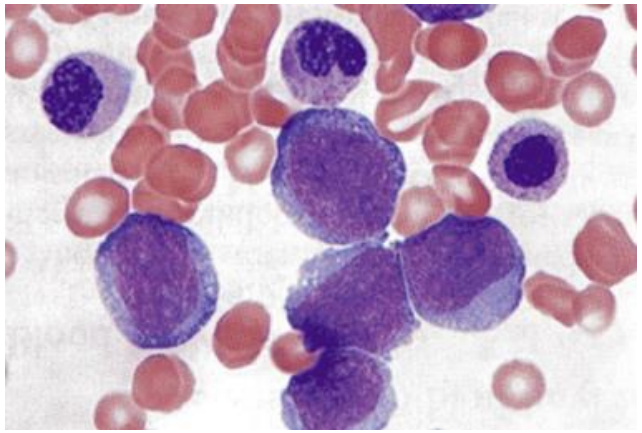


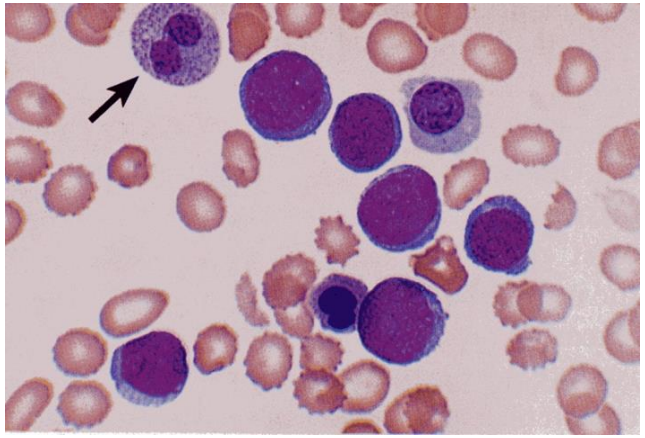
Hochhau et al. Leukemia (2009) 23, 1054–1061;

	WHO criteria[5]	European Leukaemia Net criteria[6]
Accelerated phase		
Blasts in peripheral blood or bone marrow	10–19%	15–29% or blasts plus promyelocytes in peripheral blood or bone marrow >30% with blasts <30%
Basophils in peripheral blood	≥20%	≥20%
Platelets	<100 × 10^9/L not attributable to treatment, or platelets >1000 × 10^9/L uncontrolled on treatment	<100 × 10^9/L not attributable to treatment
Additional chromosomal abnormalities	Occurring on treatment	Occurring on treatment
White cell count and spleen size	Increasing and uncontrolled on treatment	..
Blast crisis		
Blasts in peripheral blood or bone marrow	≥20%	≥30%
Blast proliferation	Extramedullary, except spleen	Extramedullary, except spleen
Large foci of blasts	Bone marrow or spleen	..

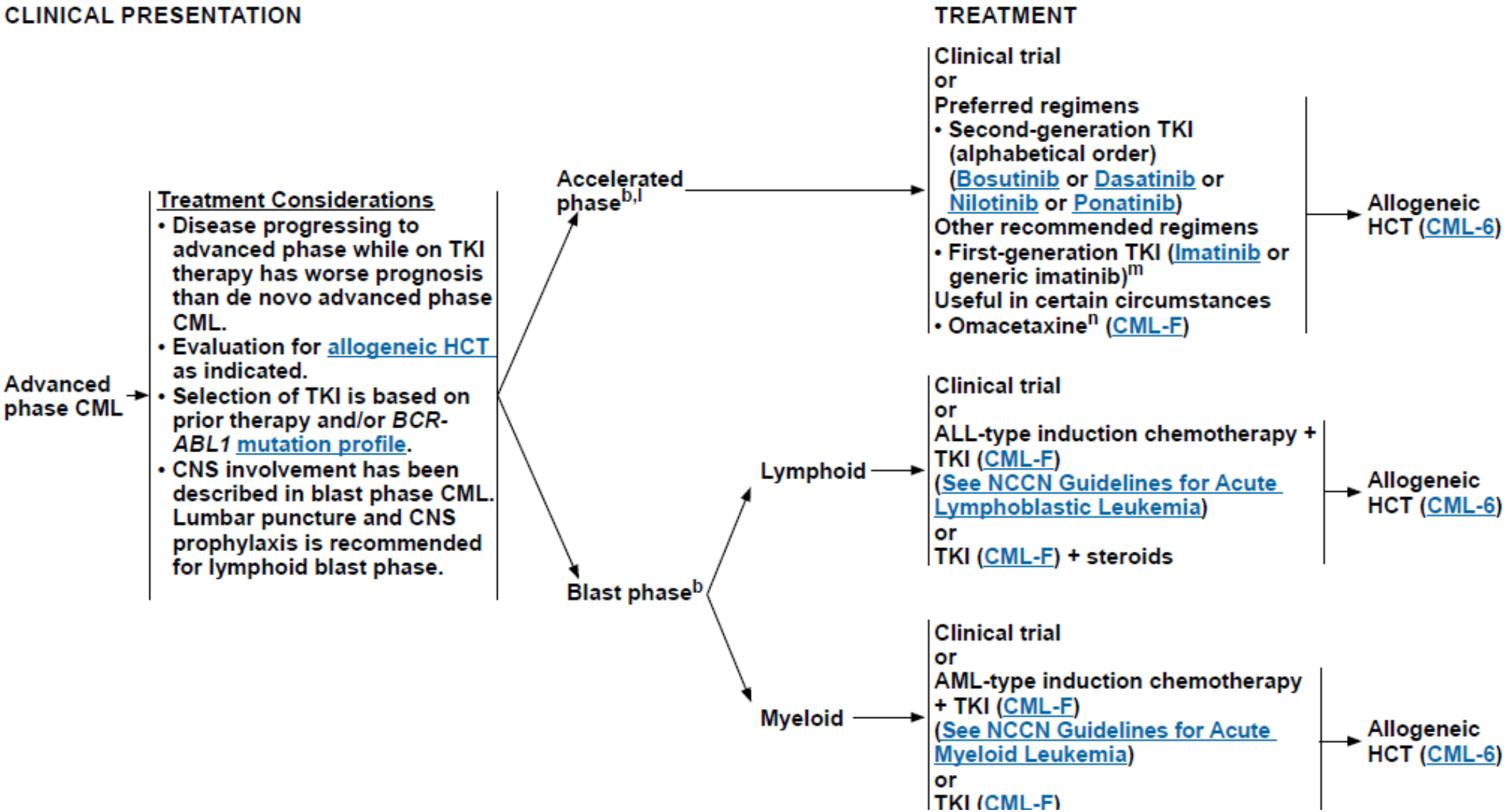
Table 1: **Definitions of accelerated phase and blast crisis according to present classification systems**







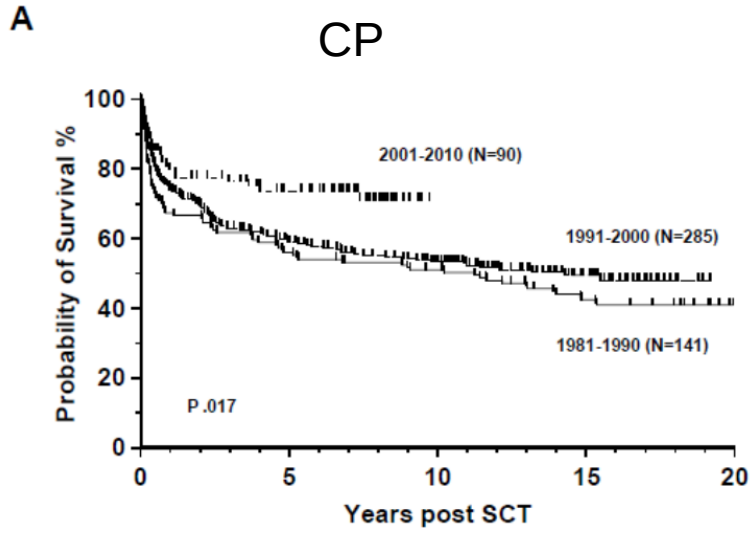
Apperley JF. Lancet 2015; 385: 1447–59

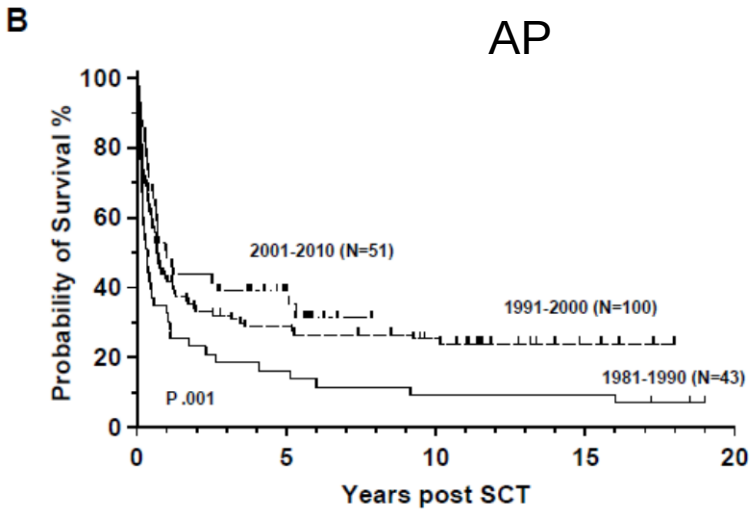

CLINICAL PRESENTATION
TREATMENT
Advanced phase CML
Treatment Considerations
• Disease progressing to advanced phase while on TKI therapy has worse prognosis than de novo advanced phase CML.
• Evaluation for allogeneic HCT as indicated.
• Selection of TKI is based on prior therapy and/or BCR-ABL1 mutation profile.
• CNS involvement has been described in blast phase CML. Lumbar puncture and CNS prophylaxis is recommended for lymphoid blast phase.
Accelerated phase[b,l]
Blast phase[b]
Lymphoid
Myeloid
Clinical trial
or
Preferred regimens
• Second-generation TKI (alphabetical order) (Bosutinib or Dasatinib or Nilotinib or Ponatinib)
Other recommended regimens
• First-generation TKI (Imatinib or generic imatinib)[m]
Useful in certain circumstances
• Omacetaxine[n] (CML-F)
Allogeneic HCT (CML-6)
Clinical trial
or
ALL-type induction chemotherapy + TKI (CML-F) (See NCCN Guidelines for Acute Lymphoblastic Leukemia)
or
TKI (CML-F) + steroids
Allogeneic HCT (CML-6)
Clinical trial
or
AML-type induction chemotherapy + TKI (CML-F) (See NCCN Guidelines for Acute Myeloid Leukemia)
or
TKI (CML-F)
Allogeneic HCT (CML-6)

Indication for allo-SCT in CML

CML phase	Clinical situation	TKI and chemotherapy management	HLA typing and donor search	Immediate allo-SCT referral
CP	First failure of imatinib, high risk	Second-line TKI	Yes	No
	First failure of nilotinib or dasatinib	Second-line TKI	Yes	Yes
	Failure to 2 TKIs	Third-line TKI	Yes	Yes
	T315I mutation	Ponatinib or omacetaxine	Yes	Yes
AP	TKI naïve	TKI ± chemotherapy	Yes	Yes
	TKI naïve, without optimal response	Second-line TKI ± chemotherapy	Yes	Yes
	TKI pretreated	Second-line TKI ± chemotherapy	Yes	Yes
BP	TKI naïve or pretreated	Induction chemotherapy, TKI	Yes	Yes

Barrett Blood. 2015;125(21):3230-3235)



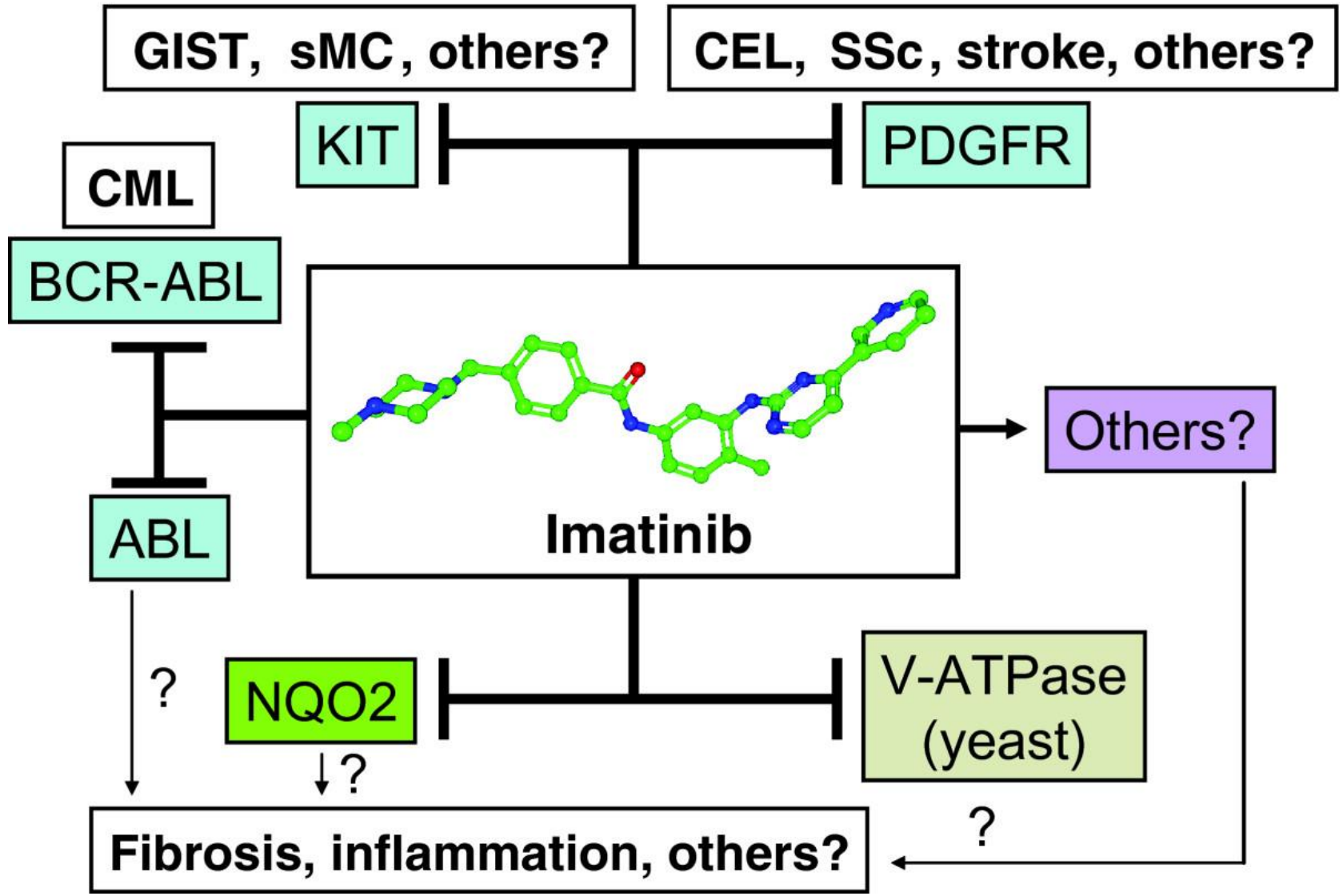


	Score*
Age (years)	
<20	0
20–40	1
>40	2
Disease phase	
Chronic phase	0
Acceleration, second of subsequent chronic phase	1
Blast crisis	2
Stem cell source	
HLA-matched sibling	0
Volunteer unrelated donor or mismatched family member	1
Donor-recipient sex combinations	
Male to male	0
Male to female	0
Female to female	0
Female to male	1
Time from diagnosis to transplant	
<12 months	0
>12 months	1

Taken from Gratwohl/European Group for Blood and Marrow Transplantation score.[30] *Total score will be in the range 0–7.

Table 3: **Factors affecting transplant outcome in chronic myeloid leukaemia**

Apperley JF. Lancet 2015; 385: 1447–59


GIST, sMC, others?
CEL, SSc, stroke, others?
KIT
PDGFR
CML
BCR-ABL
ABL
Imatinib
Others?
?
NQO2
?
V-ATPase
(yeast)
?
Fibrosis, inflammation, others?

Molecular genetic abnormalities in myeloid/lymphoid neoplasms associated with eosinophilia

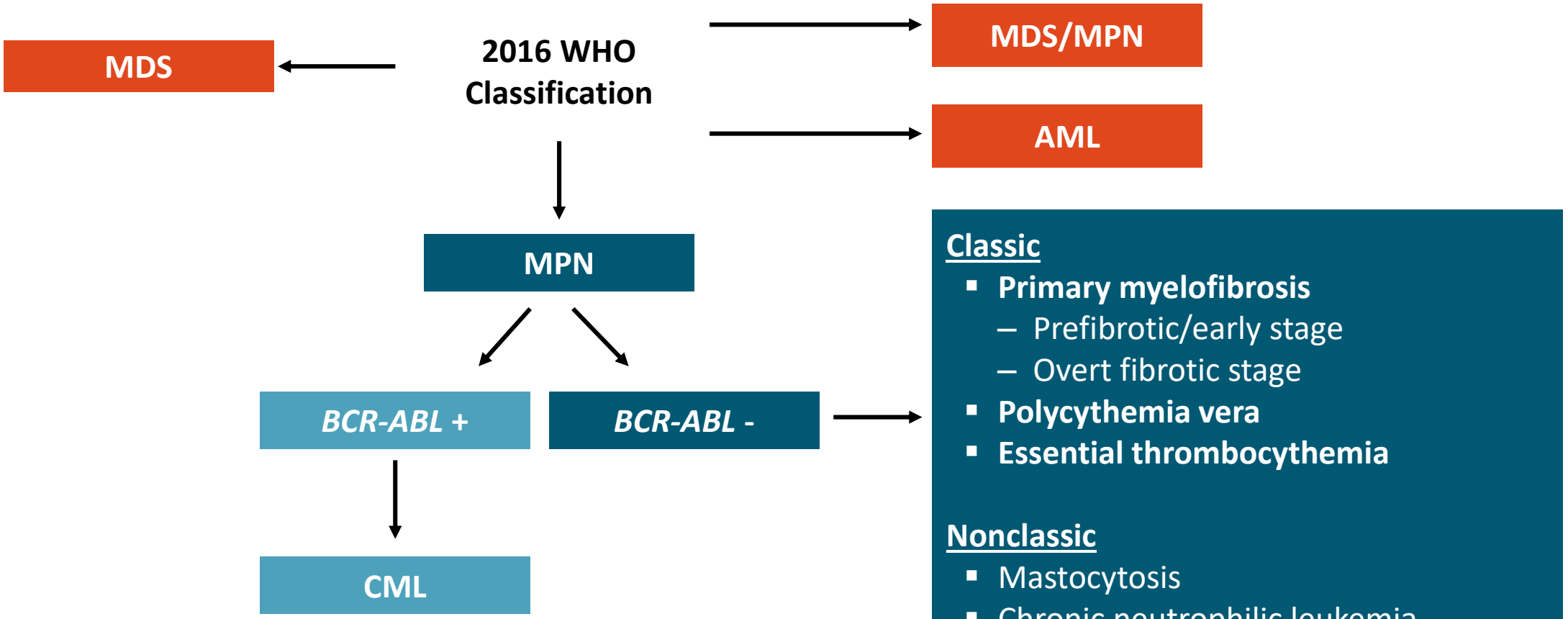
Table 10. Molecular genetic abnormalities in myeloid/lymphoid neoplasms associated with eosinophilia

Disease	Presentation	Genetics	Treatment
PDGFRA	Eosinophilia ↑Serum tryptase ↑Marrow mast cells	Cryptic deletion at 4q12 *FIP1L1-PDGFRA*, at least 66 other partners	Respond to TKI
PDGFRB	Eosinophilia Monocytosis mimicking CMML	t(5;12)(q32;p13.2) *ETV6-PDGFRB*, at least 25 other partners	Respond to TKI
FGFR1	Eosinophilia Often presents with T-ALL or AML	Translocations of 8p11.2 *FGFR1*-various partners	Poor prognosis; do not respond to TKI
PCM1-JAK2	Eosinophilia Rarely presents with T-LBL or B-ALL Bone marrow shows left-shifted erythroid predominance and lymphoid aggregates	t(8;9)(p22;p24.1) *PCM1-JAK2*	May respond to JAK2 inhibitors

↑, Increased.

Arber DA et al. Blood. 2016; 127(20):2391-2405

Myeloid Malignancies

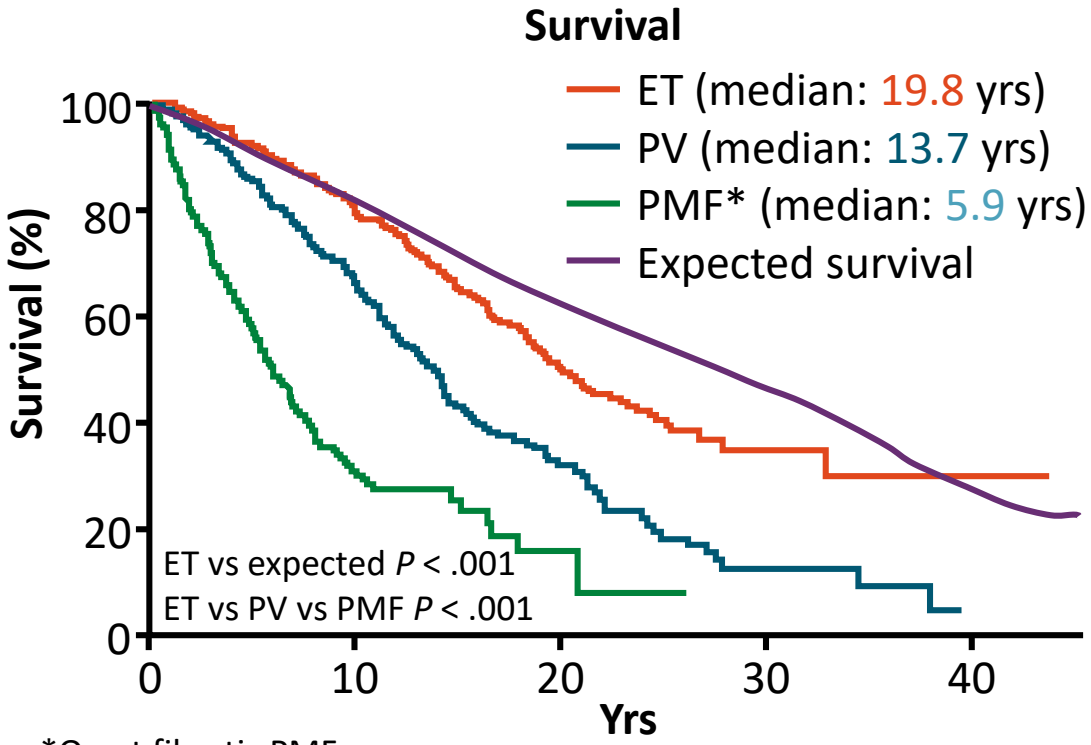


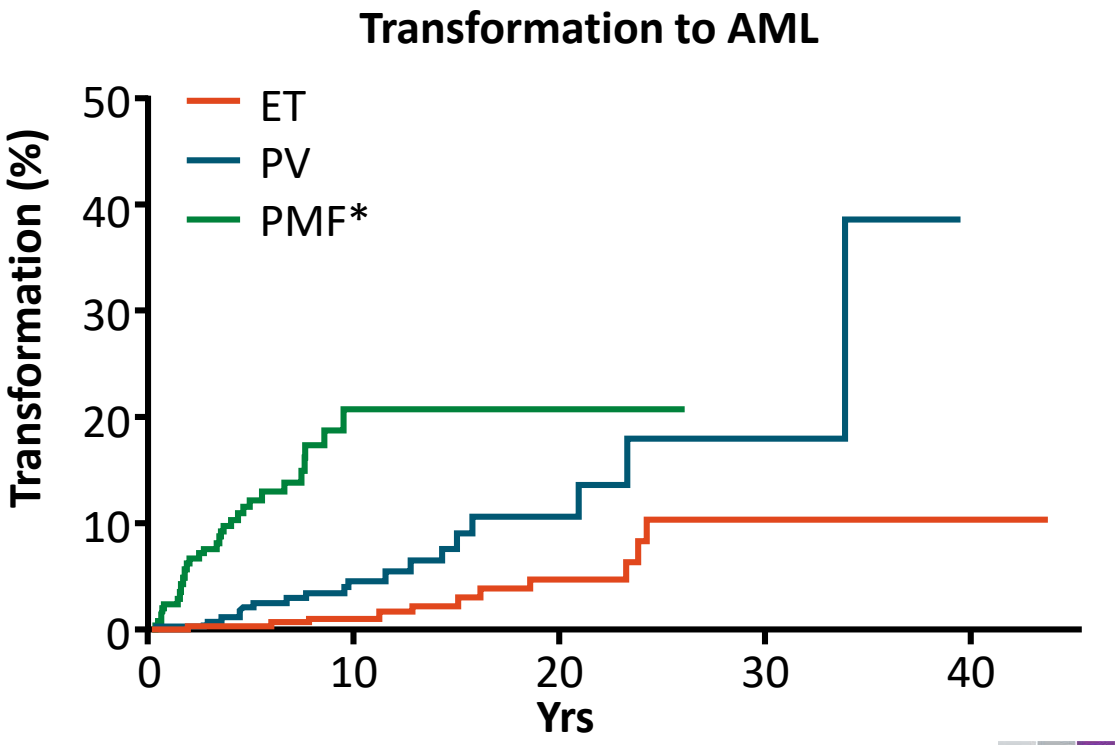
Arber. Blood. 2016;127:2391.

Slide credit: clinicaloptions.com

Survival and Disease Progression With PV, MF, and ET

- Although similarities exist in the molecular signature and presentation of PV, MF, and ET, important to distinguish between these conditions as prognosis and management can differ
- Assessment of survival and progression in patients with PV, MF, or ET at Mayo Clinic (N = 826)



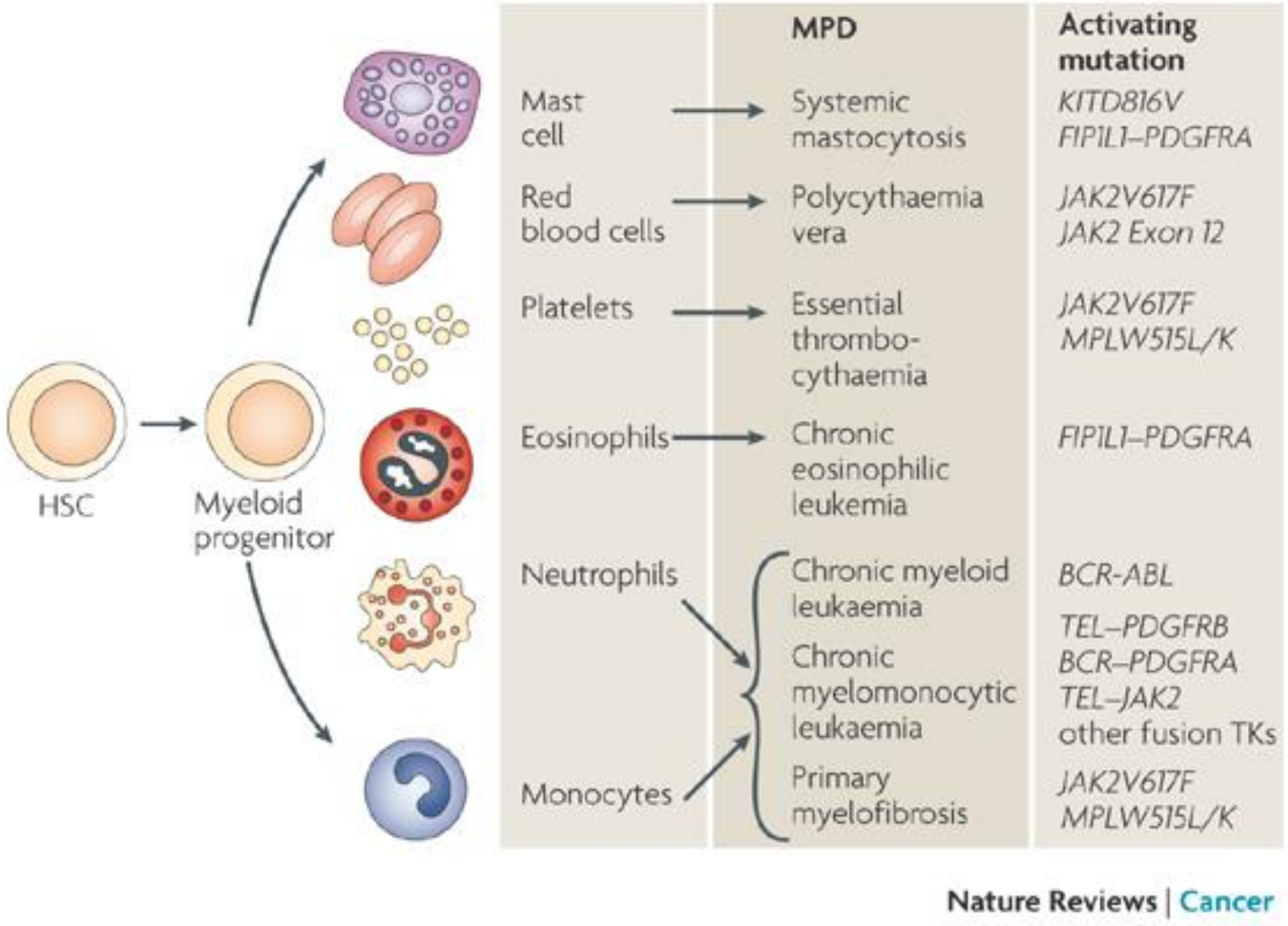


*Overt fibrotic PMF.

Tefferi. Blood. 2014;124:2507.

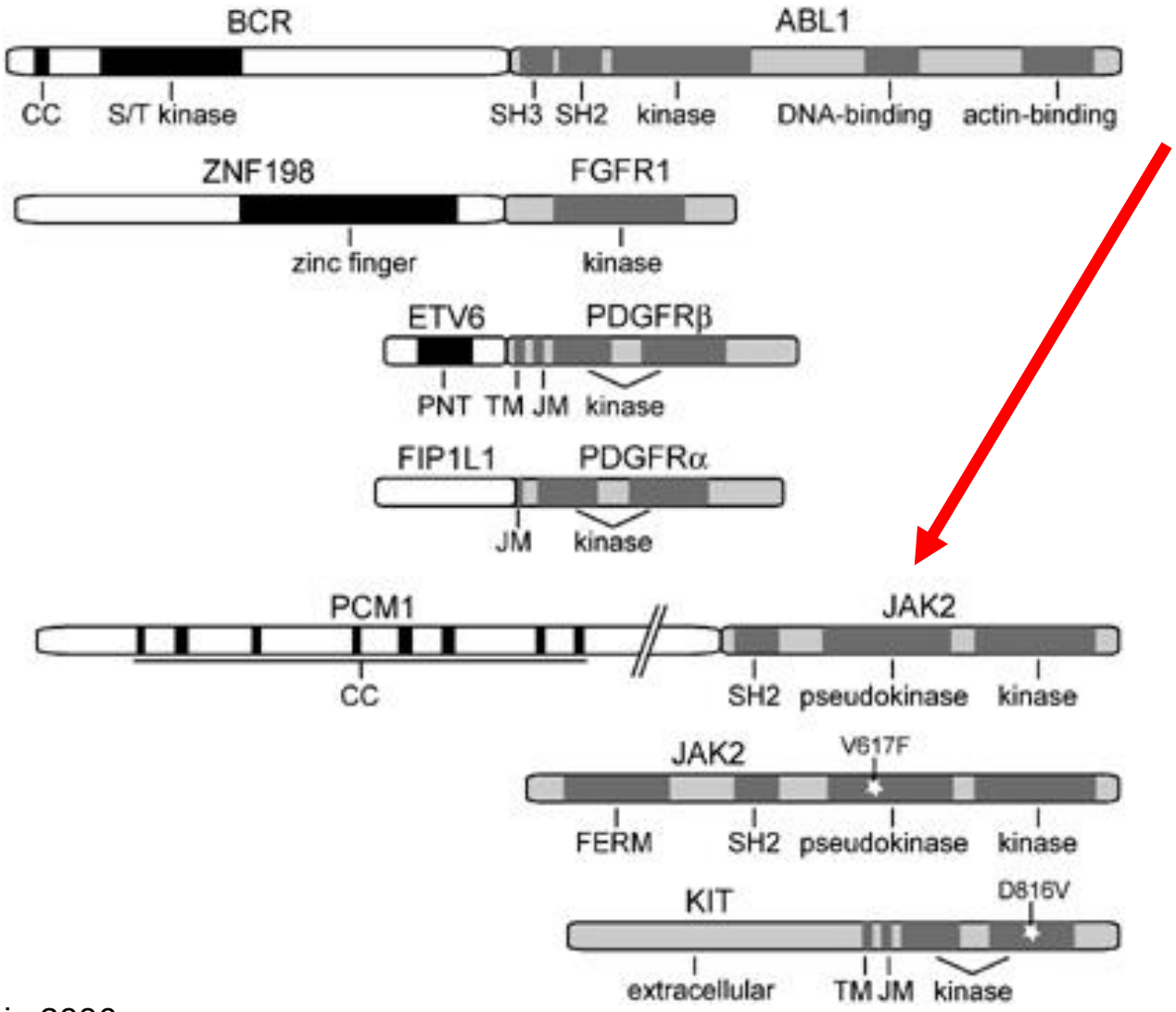
Slide credit: clinicaloptions.com

Classification and molecular pathogenesis of the MPD



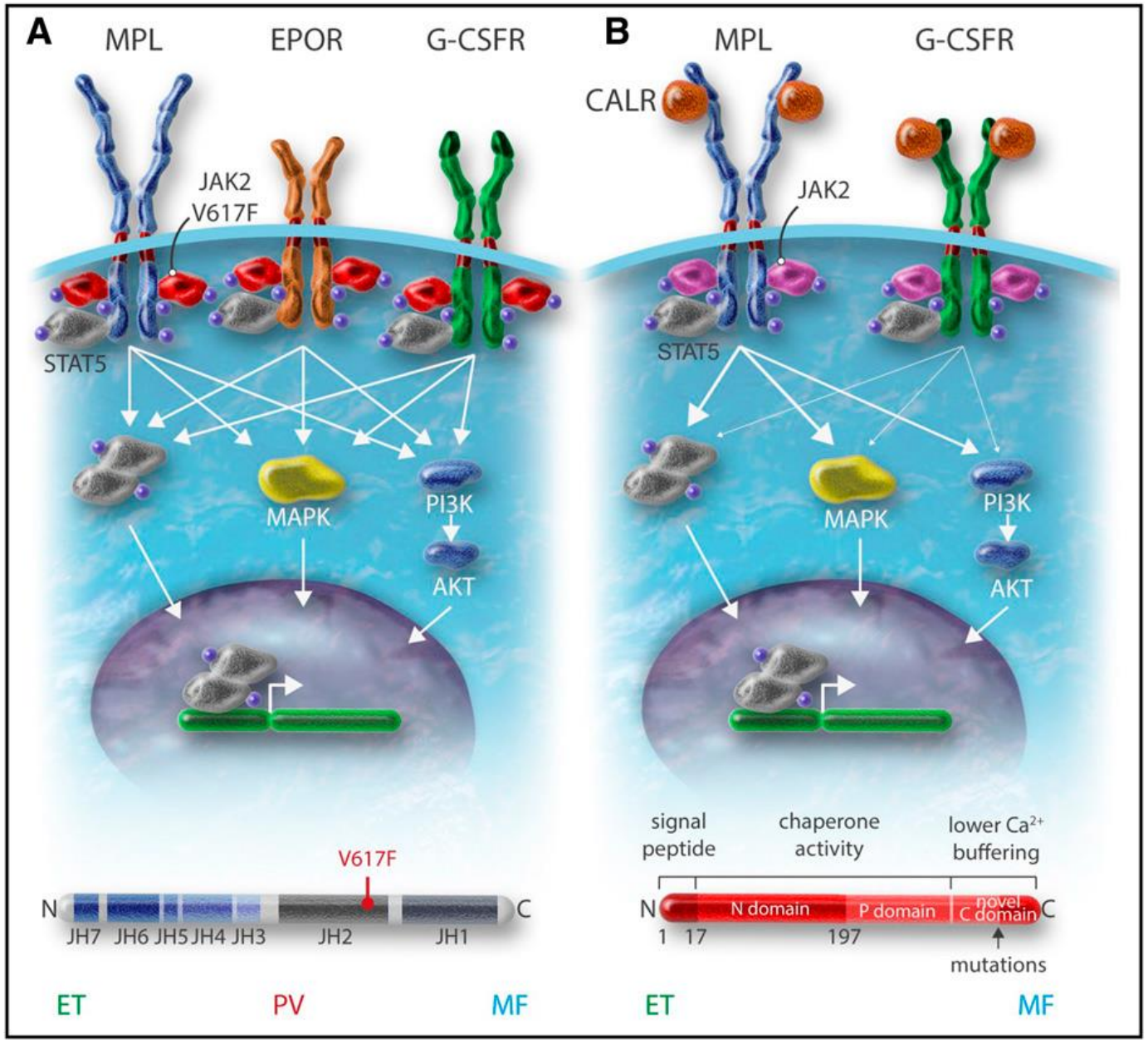
Levine RL et al. Nat Rev Cancer. 2007;7:673-83

Tyrosine kinase involved in the pathogenesis of CMPD



Keersmaecker Leukemia 2006

Role of cytokine receptors in the oncogenic properties of JAK2V617F and CALR mutants.



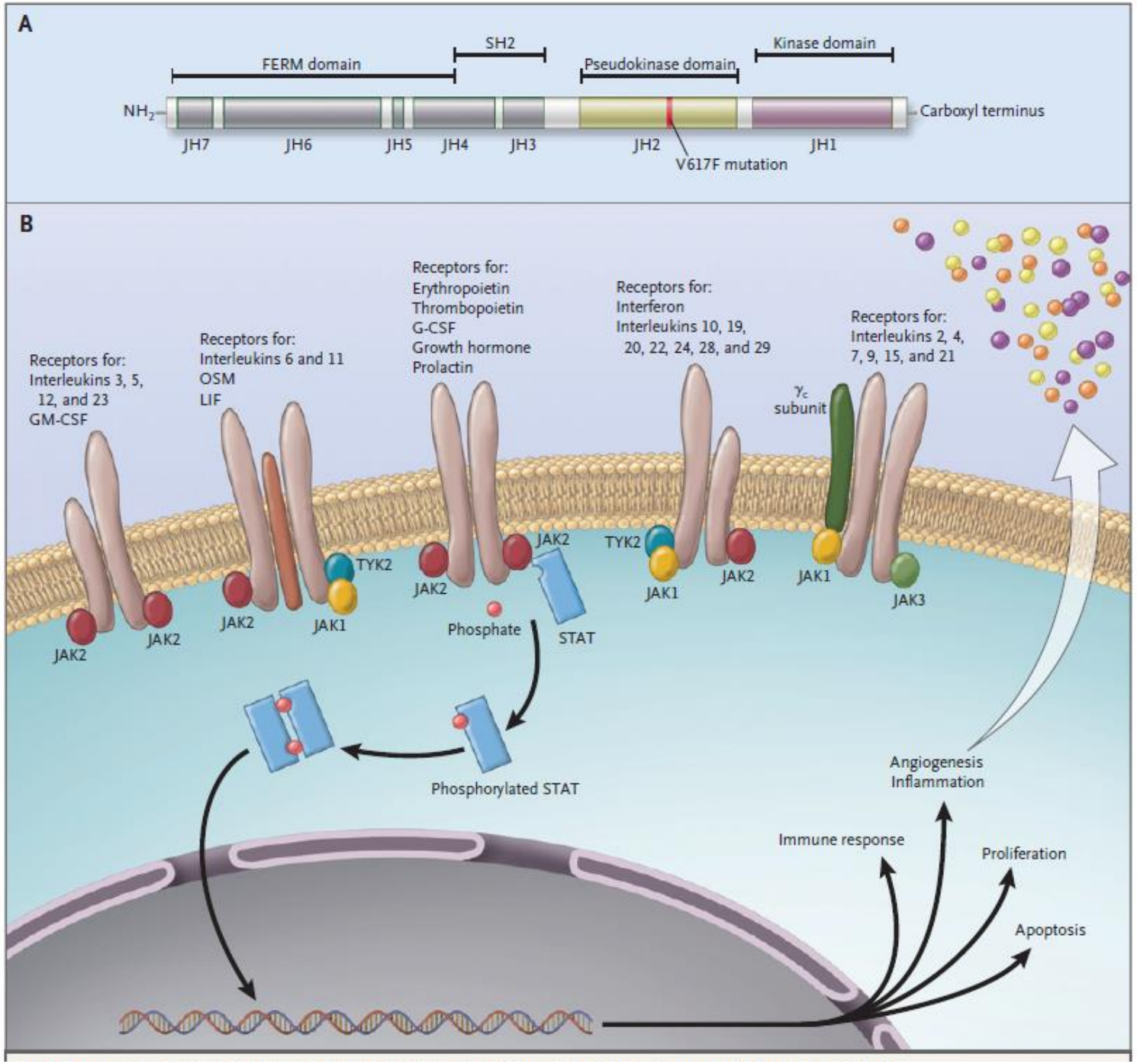
(A) JAK2V617F activates signaling through the 3 main homodimeric receptors EPOR, MPL, and G-CSFR, which are involved in erythrocytosis, thrombocytosis, and neutrophilia, respectively.

(B) The CALR mutants mainly activate MPL and at a low level the G-CSFR but not the EPOR, explaining the thrombocytosis associated with these mutants

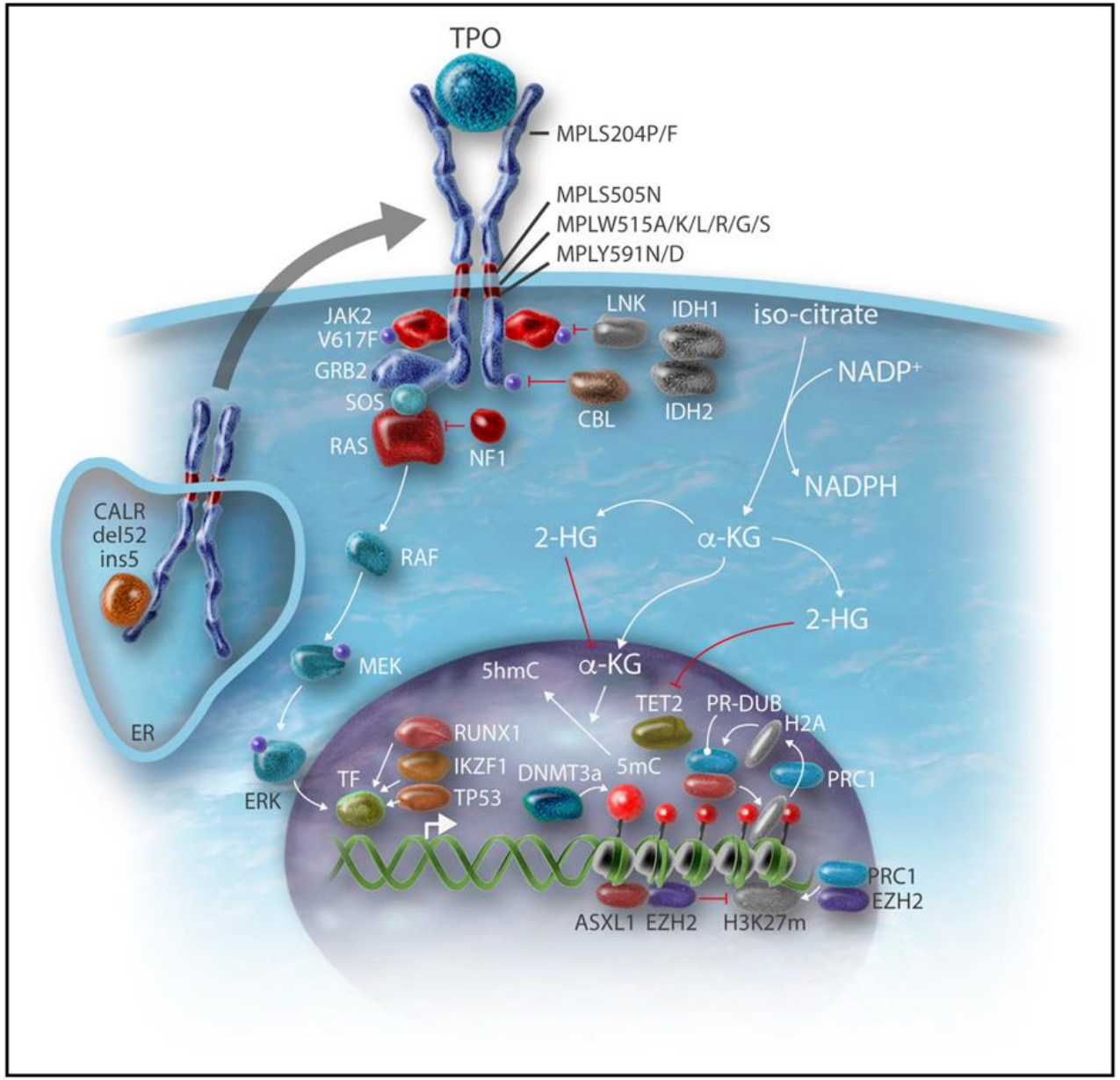
Vainchenker W. Blood. 2017;129(6):667-679

JAK2 V617F

Vannucchi AM. NEJM, 2010;3623:1180



Genes involved in epigenetic regulation and leukemic transformation.



The mechanisms by which the genes involved in the epigenetic regulation lead to modifications in gene regulation are detailed.
Some genes involved in leukemic transformation (N-Ras pathway and transcription factors such as p53, RUNX1) are also described.
MEK, MAPK/ERK-kinase; RAF, rapidly accelerated fibrosarcoma; SOS, Son of Sevenless; TF, transcription factor.

Vainchenker W. Blood. 2017;129(6):667-679

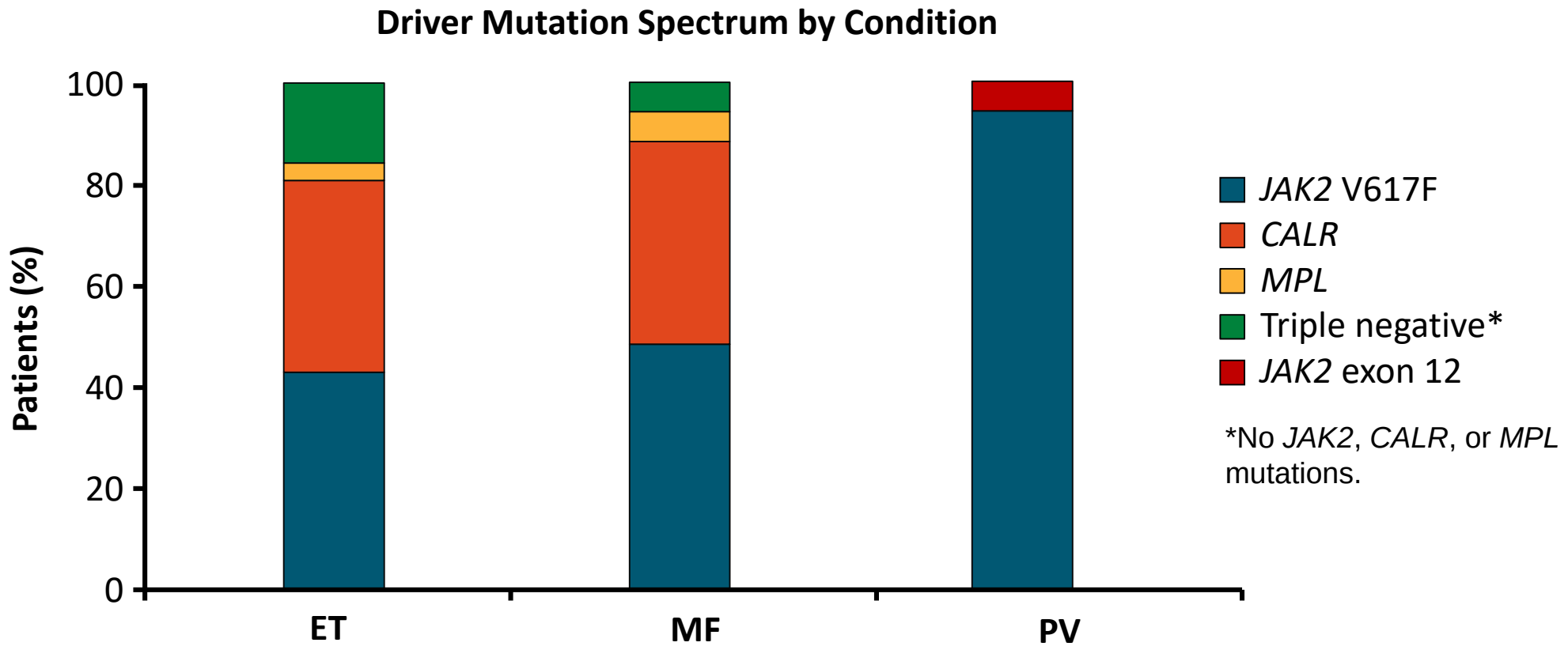
Commonly mutated genes in the myeloproliferative neoplasms

Gene	Chromosome location	Mutation location	Frequency (%)		
			PV	ET	PMF
JAK2	9p24	exon 14	97	50–60	55–60
JAK2	9p24	exon 12	1–2	rare	rare
MPL	1p34	exon 10	rare	3–5	5–10
CALR	19p13	exon 9	rare	20–30	25–35
TET2	4q24	all coding regions	10–20	5	10–20
IDH1/IDH2	2q33/15q26	exons 4	rare	rare	5
DNMT3A	2p23	exons 7–23	5–10	1–5	5–10
ASXL1	20q11	exon 13	2–5	2–5	15–30
EZH2	7q35-q36	all coding regions	1–3	rare	5–10
CBL	11q23	exons 8–9	rare	rare	5–10
SH2B3	12q24	exon 2	rare	rare	rare
SF3B1	2q33	exons 12–16	rare	rare	5–10
SRSF2	17q25	exon 1	rare	rare	10–15
U2AF1	21q22	exons 2–7	rare	rare	5–15

PV, polycythemia vera; ET, essential thrombocythemia; PMF, primary myelofibrosis

Langabeer et al EJH 2015:95;270–279

Phenotype Driver Mutations Activating the JAK-STAT Pathway in MPNs



- A very small percentage of PV patients may have *LNK* or *CALR* driver mutations
- Nondriver mutations mostly frequently occurring in MPNs: *TET2, ASXL1, DNMT3A*

Grinfeld. Haematologica. 2017;102:7.

Slide credit: clinicaloptions.com

2016 Revised WHO Diagnostic Criteria for Myeloproliferative Neoplasms

Arber et al. Blood 2016;127:2391

		Polycythemia Vera (PV)		Essential Thrombocythemia (ET)		Primary Myelofibrosis (PMF) (overt)	Primary Myelofibrosis (prefibrotic) (prePMF)
Major criteria	1	Hemoglobin (Hgb) >16.5 g/dL (men) >16 g/dL (women) or Hematocrit >49% (men) >48% (women) or ↑ red cell mass >25% above mean	1	Platelet count ≥450 x 10^9/L	1	Megakaryocyte proliferation and atypia*** and ≥ grade 2 reticulin/collagen fibrosis ***megakaryocytes with aberrant nuclear/cytoplasmic ratio and hyperchromatic and irregularly folded nuclei and dense clustering	Megakaryocyte proliferation and atypia*** and ≤ grade 1 reticulin/collagen fibrosis, Increased cellularity, granulocytic proliferation and decreased erythropoiesis
	2	Bone marrow (BM) tri-lineage myeloproliferation with pleomorphic mature megakaryocytes*	2	BM megakaryocyte proliferation with large and mature morphology and hyper-lobulated nuclei. Reticulin fibrosis grade should be ≤1	2	Not meeting WHO criteria for other myeloid neoplasm	Not meeting WHO criteria for other myeloid neoplasm
	3	Presence of *JAK2* mutation	3	Not meeting WHO criteria for other myeloid neoplasms	3	Presence of *JAK2*, *CALR* or *MPL* mutation or presence of another clonal marker or absence of evidence for reactive bone marrow fibrosis	Presence of *JAK2*, *CALR* or *MPL* mutation or presence of another clonal marker or absence of evidence for reactive bone marrow fibrosis
			4	Presence of *JAK2*, *CALR* or *MPL* mutation			
Minor criteria		1. Subnormal serum erythropoietin level		1. Presence of a clonal marker or absence of evidence for reactive thrombocytosis	1	1. Anemia not otherwise attributed 2. Leukocytosis ≥11 x 10^9/L 3. Palpable splenomegaly 4. Increased lactate dehydrogenase (LDH), above upper normal limit 5. Leukoerythroblastosis	1. Anemia not otherwise attributed 2. Leukocytosis ≥11 x 10^9/L 3. Palpable splenomegaly 4. Increased lactate dehydrogenase (LDH), above upper normal limit

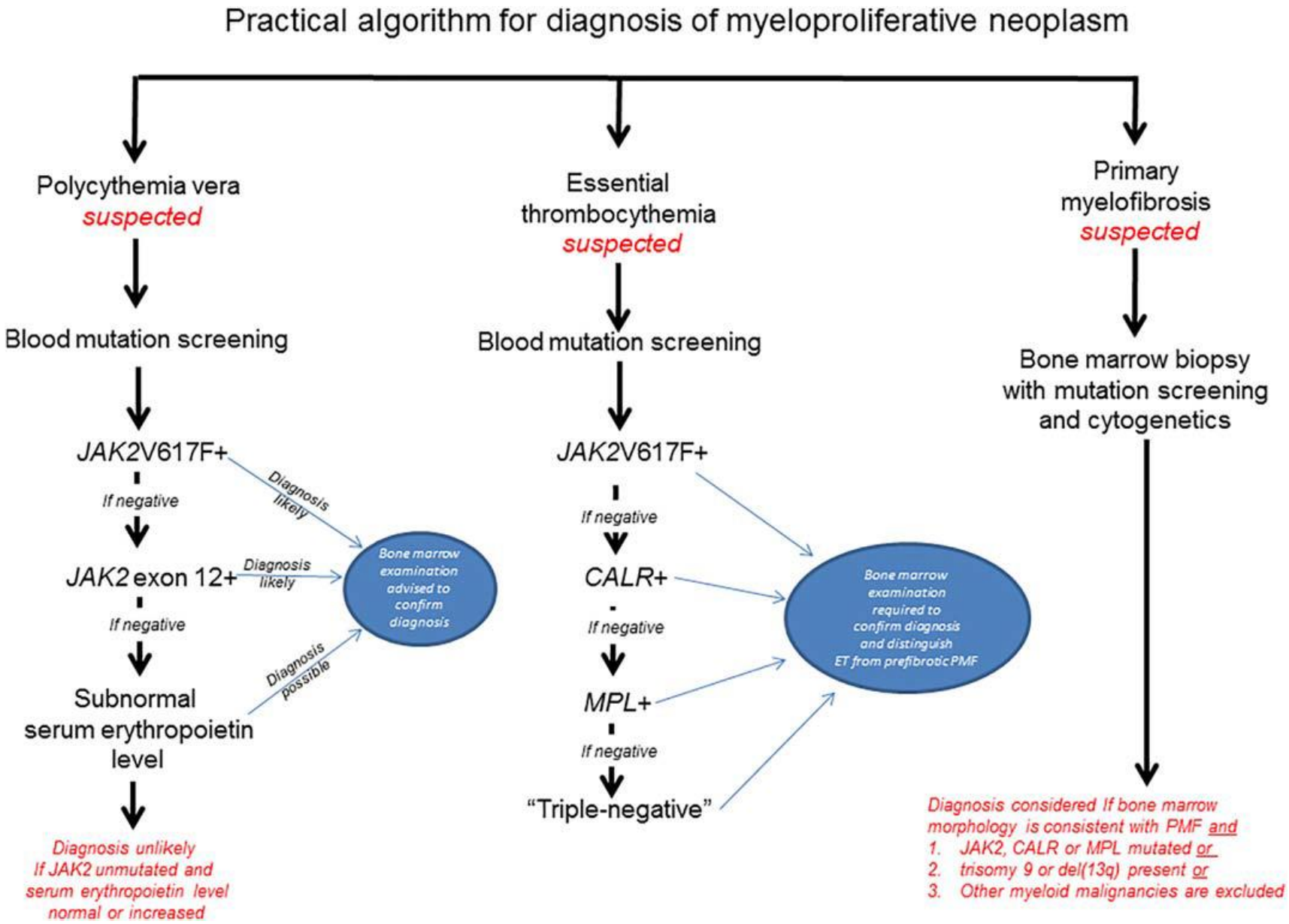
PV diagnosis requires meeting all three major criteria or the first two major criteria and one minor criterion.
*BM biopsy may not be not required if Hb >16.5 g/dL in men or 16.5 in women (Hct >55.5 in men and 49.5 in women)

ET diagnosis requires meeting all 4 major criteria or first three major criteria and one minor criterion

PMF diagnosis requires meeting all 3 major criteria and at least one minor criterion

prePMF diagnosis requires meeting all 3 major criteria and at least one minor criterion

Tefferi & Barbui AJH 2016



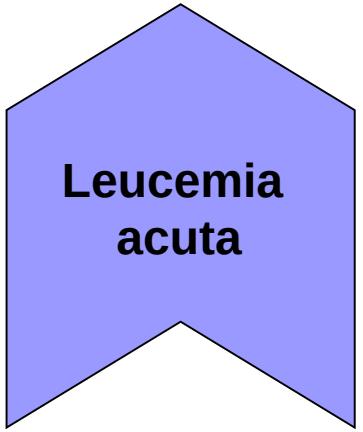
Tefferi & Barbui Am. J. Hematol. 92:95–108, 2016.

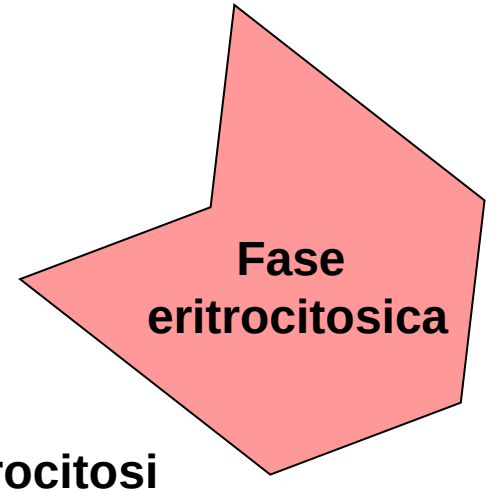
Policitemia Vera (PV)

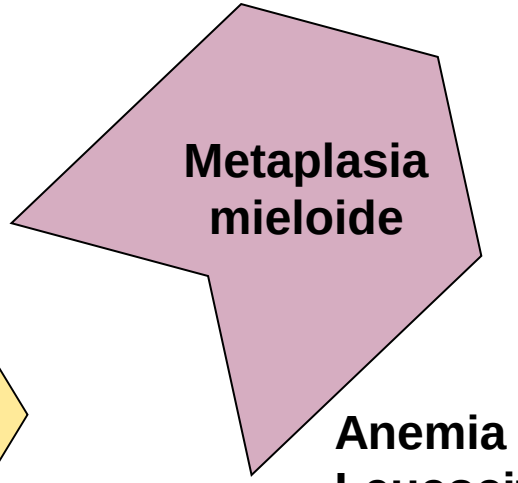
- **Definizione**
  - Malattia neoplastica derivata dall'espansione clonale della cellula staminale trasformata e caratterizzata soprattutto da incremento della massa eritrocitaria.

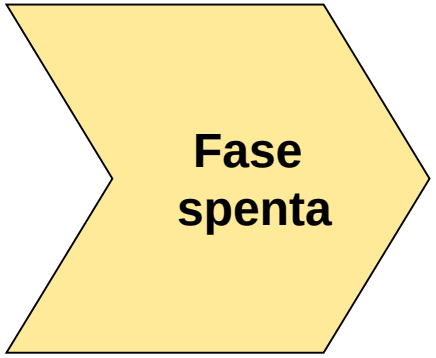
- **Epidemiologia**
  - Incidenza in Europa: 8-10 casi/1,000,000 per anno (2 in Giappone, 13 in Australia)

PV: decorso









Eritrocitosi
Leucocitosi
Piastrinosi
Splenomegalia
Trombosi
Emorragie
Prurito

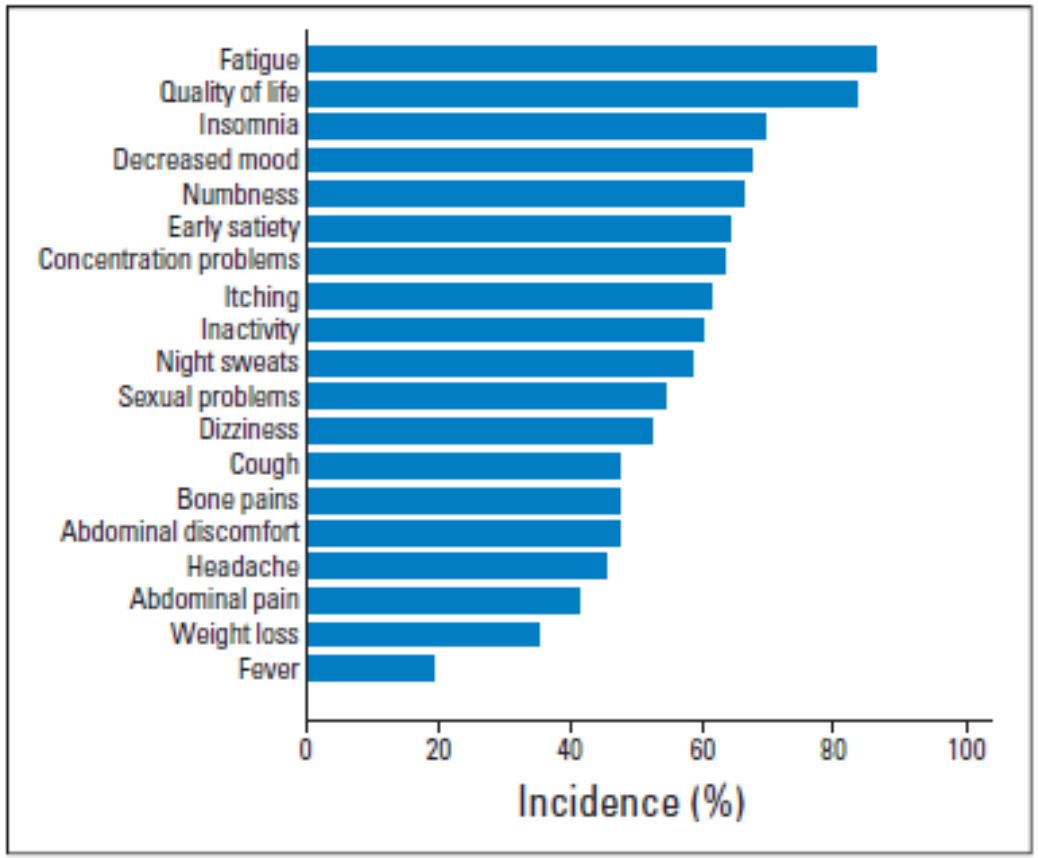
Parametri ematologici stabili
Nessuna terapia

Anemia
Leucocitosi
Piastrinosi o piastrinopenia
Splenomegalia
Febbre
Calo ponderale

PV: clinica

- **Età media 60 anni,**
- **M/F 2:1**

- **Esordio**
  - **Asintomatico**
  - **Sintomatico**
    - **Cefalea, acufeni, vertigini, disturbi visivi, (scotomi, diplopia) da iperviscosità ematica**
    - **Episodi vascolari (trombotici e/o emorragici) di diversa gravità (40% dei casi causa di morte)**
    - **Prurito**
    - **Ipertensione**
    - **rubeosi**



Stein et al, JCO 2015

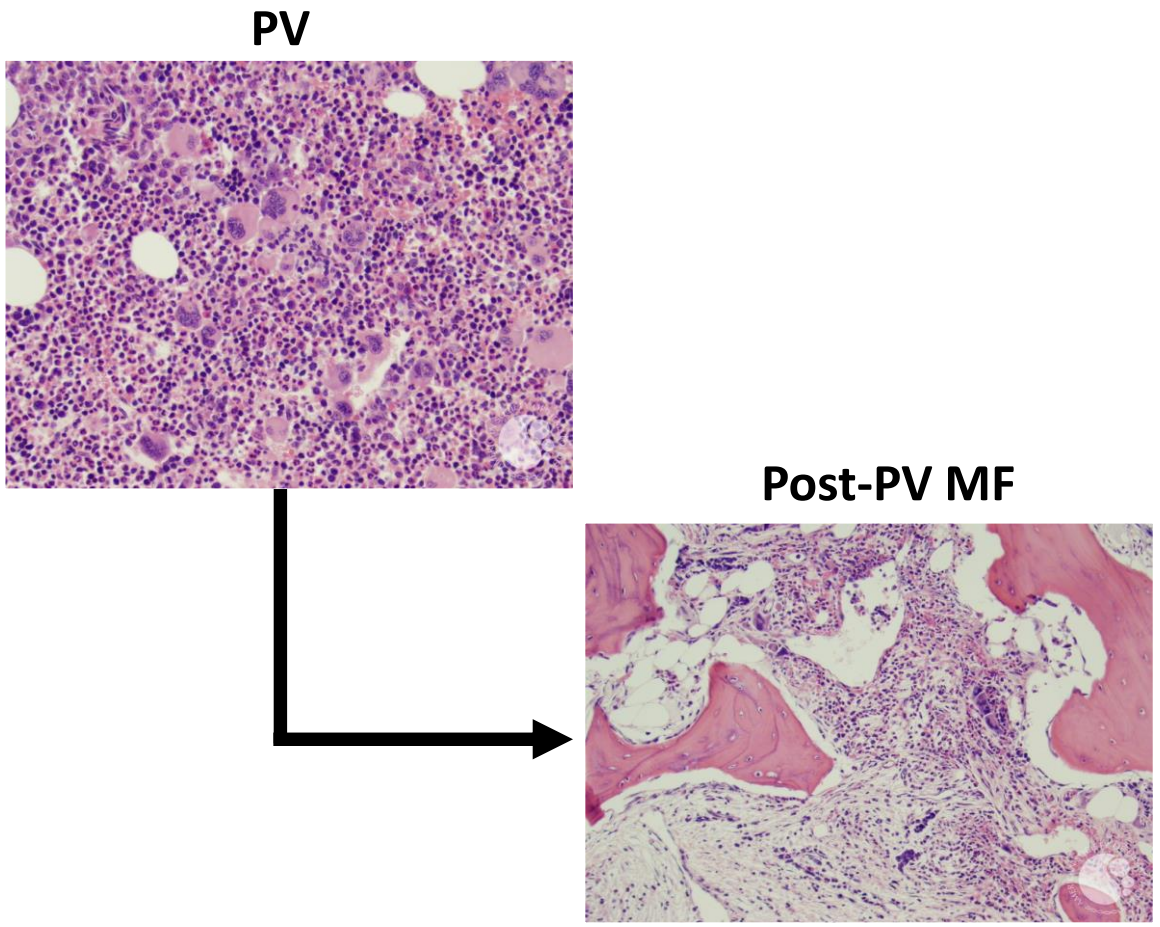
Evolution of WHO PV Diagnostic Criteria

2008 WHO[1]	2016 WHO[2]
Requirement for diagnosis	
▪ 2 major and 1 minor criteria OR 1 major and 2 minor criteria	▪ All 3 major criteria OR first 2 major criteria and the minor criterion
Major criteria	
1. Hb > 18.5 g/dL (Men); > 16.5 g/dL (Female) *2.* *JAK2* V617F mutation or similar (*JAK2* exon 12)	1. Hb > 16.5 g/dL or Hct > 49% (men); or Hb > 16.0 g/dL or Hct > 48% (women); or increased red cell mass 2. BM biopsy showing hypercellularity, trilineage growth (panmyelosis) with erythroid, granulocytic, and pleomorphic, mature megakaryocytic proliferation *3.* *JAK2* V617F or *JAK2* exon 12 mutation
Minor criteria	
1. Subnormal serum EPO level 2. BM trilineage proliferation 3. Endogenous erythroid colony growth	1. Subnormal serum EPO level

1. Thiele J, et al. Curr Hematol Malig Rep. 2009;4:33-40. 2. Arber DA, et al. Blood. 2016;127:2391-2405. Slide credit: clinicaloptions.com

Bone Marrow Testing in PV Diagnosis

- **Bone marrow biopsy may not be required for diagnosis** if sustained Hb levels > 18.5 g/dL (men) or > 16.5 g/dL (women) where *JAK2* mutated and EPO suppressed[1]
- **Biopsy may identify fibrosis at diagnosis**
  - Prevalence: 14% to 48% with grade 1 fibrosis at diagnosis; consequences include a higher rate of overt, fibrotic progression[2,3]
- **Biopsy required to diagnose post-PV MF**[4]
  - Progression prevalence: 5% to 19% at 15 yrs
  - Note that high-grade bone marrow fibrosis alone not enough to diagnose post-PV MF; see WHO MF diagnostic criteria on next slide



1. Arber. Blood. 2016;127:2391. 2. Barbui. Blood. 2012;119:2239. 3. Barraco. Blood Cancer J. 2017;7:e538. 4. Cerquozzi. Blood Cancer J. 2015;5:e366.

Slide credit: clinicaloptions.com

Familial polycythemia (rare)

- **High Epo levels**
  - Low P50: increased affinity of hemoglobin for oxygen
    - High-O2-affinity hemoglobin variants
    - 2,3-bisphosphoglycerate (2,3-BPG) deficiency
    - Methemoglobinemia
  - Normal P50: defects in oxygen sensing
    - Homozygous Chuvash *VHL* mutation
    - Other *VHL* mutations
- **Low or normal Epo levels**
  - *Epo-R* mutations: primary familial and congenital polycythemias

Table II. Germline mutations causing MPN-like disorders.

Gene	Disease	Inheritance	Representative references
Hereditary erythrocytosis			
EPOR	ECYT1: Primary familial and congenital polycythaemia (PFCP)	AD	de la Chapelle *et al* (1993)
VHL	ECYT2: von Hippel-Lindau disease	AR	Ang *et al* (2002)
			Pastore *et al* (2003)
			Percy *et al* (2003)
			Perrotta *et al* (2006)
EGLN1 (*PHD2*)	ECYT3	AD	Percy *et al* (2006)
			Percy *et al* (2007)
EPAS1 (*HIF2α*)	ECYT4	AD	Percy *et al* (2008)
HBB	High oxygen affinity variants	AD	Rumi *et al* (2009)
BPGM	2,3 DPG deficiency	AR-AD	Max-Audit *et al* (1980)
Hereditary thrombocytosis			
THPO	THCYT1	AD	Wiestner *et al* (1998)
			Kondo *et al* (1998)
			Ghilardi and Skoda (1999)
			Ghilardi *et al* (1999)
			Liu *et al* (2008)
MPL	THCYT2 (*MPL* S505N)	AD	Ding *et al* (2004)
			Teofili *et al* (2007)
	MPL Baltimore (*MPL* K39N)	Functional SNP*	Moliterno *et al* (2004)
	MPL P106L	Functional SNP*	El-Harith *et al* (2009)

AD, autosomal dominant; AR, autosomal recessive; ECYT, familial erythrocytosis; MPN, myeloproliferative neoplasm; SNP, single nucleotide polymorphism; THCYT, thrombocythaemia.

*Mild thrombocytosis in heterozygous individuals, severe thrombocytosis in homozygous individuals.

Rumi & Cazzola BJH 2017

Secondary polycythemia

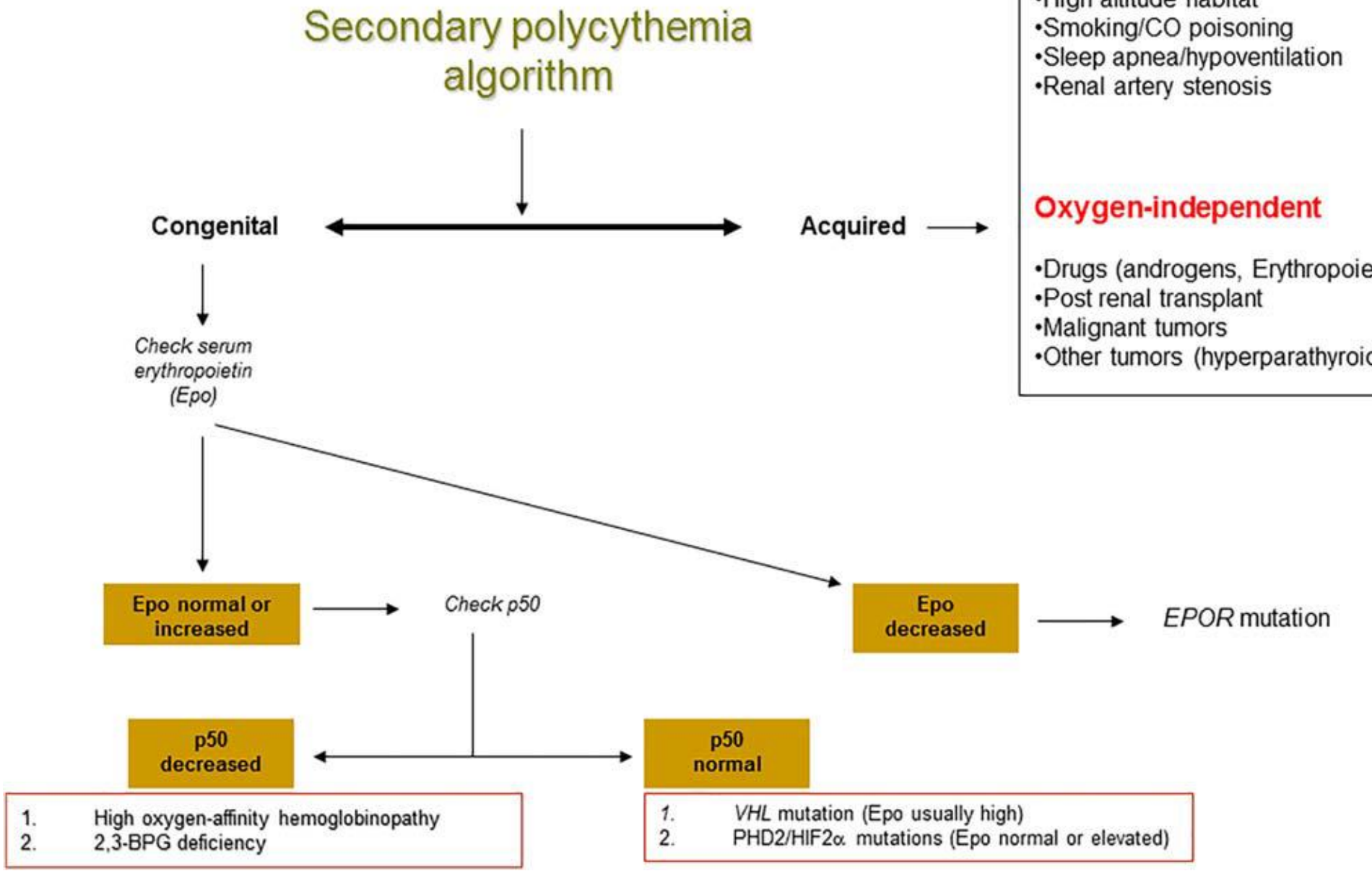
- **Physiologically inappropriate EPO increase**
  - **Tumors:**
    - renal cell carcinoma,
    - Wilms tumor,
    - hepatoma,
    - uterine fibroma,
    - cerebellar hemangioma,
    - atrial myxoma
  - **Benign renal disease:**
    - polycystic kidney disease,
    - hydronephrosis,
    - renal artery stenosis (rare)
  - **Postrenal transplantation erythrocytosis**
  - **Endocrine disorders:**
    - pheochromocytoma,
    - primary aldosteronism,
    - Bartter syndrome,
    - Cushing syndrome
  - **Erythropoiesis-stimulating hormones**
    - Epo, androgens

Secondary polycythemia

- **Physiologically appropriate EPO increase: response to hypoxia**
  - **Reduced PaO2**:
    - chronic lung disease,
    - pickwickian (obesity-hypoventilation) syndrome,
    - sleep apnea,
    - high altitude,
    - cyanotic heart disease
  - **Normal PaO2**:
    - smokers' and CO-induced polycythemia

diagnostic algorithm for secondary erythrocytosis

Tefferi & Barbui Am. J. Hematol. 92:95–108, 2016.

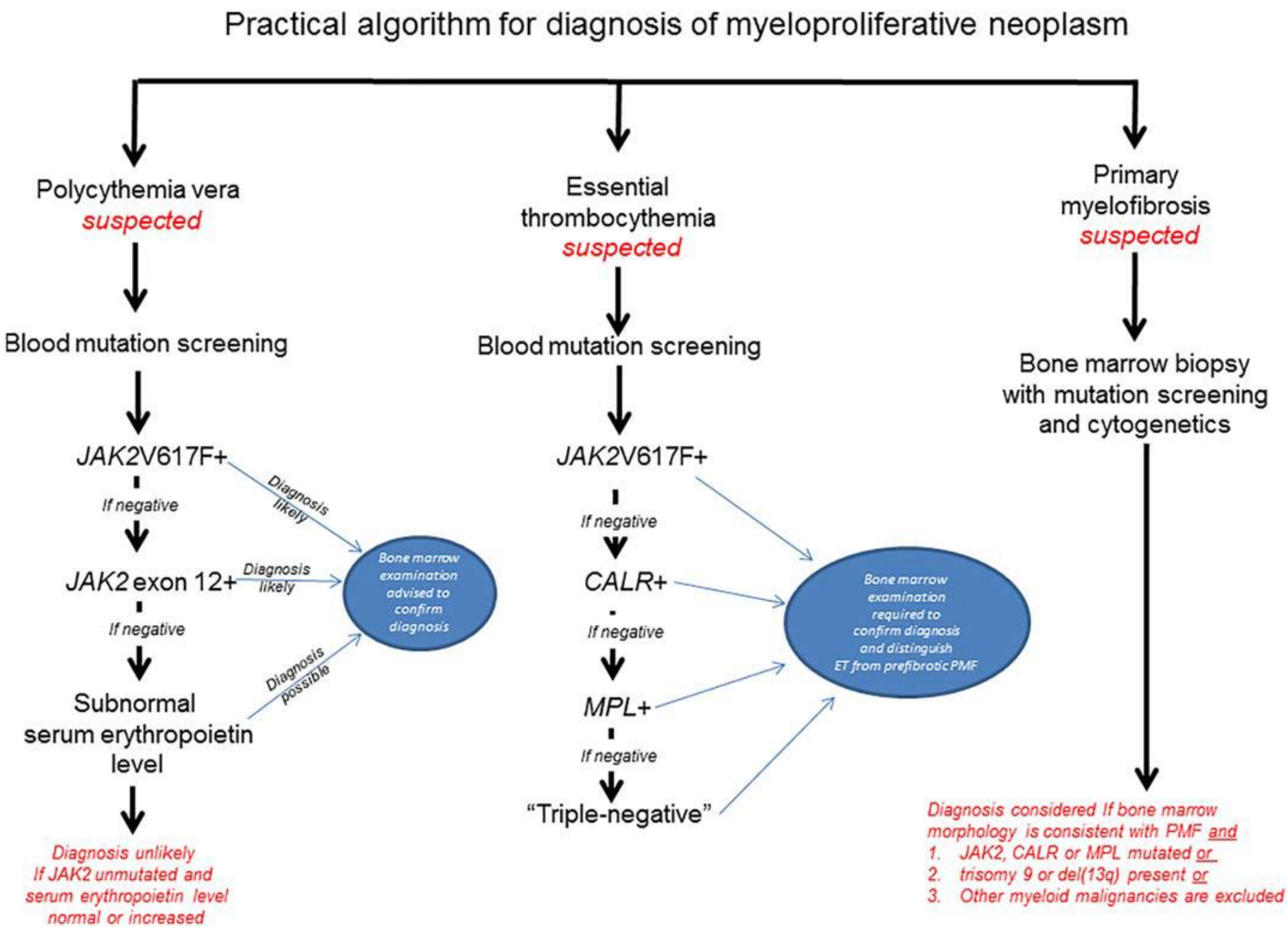


Hypoxia-driven

- Cardiac or pulmonary disease
- High altitude habitat
- Smoking/CO poisoning
- Sleep apnea/hypoventilation
- Renal artery stenosis

Oxygen-independent

- Drugs (androgens, Erythropoietin)
- Post renal transplant
- Malignant tumors
- Other tumors (hyperparathyroidism)



Tefferi & Barbui Am. J. Hematol. 92:95–108, 2016.

Table 1. Risk Factors Associated With Increased Morbidity and Mortality in Patients With Polycythemia Vera

Risk Factors
For thrombosis
Age > 60 years
Previous history of thrombosis
Leukocytosis[39-42]*
Increased *JAK2 V617F* allele burden[6,43-45]*
High-risk gene expression profile[46]*
For transformation to myelofibrosis or secondary acute myeloid leukemia
Older age[47]
Longer disease duration[48]
Leukocytosis[41]
Exposure to phosphorus-32, pipobroman, or chlorambucil[7,48]
Risk factor associated with decreased survival
Older age[7]
Leukocytosis[7]
History of venous thrombosis[7]
Abnormal karyotype[7]

*Emerging or controversial risk factor.

Stein et al, JCO 2015

Thrombosis Risk-Adapted Management of PV

Category	Characteristics	Treatment
Low risk	Age ≤ 60 yrs AND no history of thrombosis	▪ Therapeutic phlebotomy (goal Hct < 45%) ▪ ASA 81 mg daily ▪ Address CV modifiable risk factors
High risk	Age > 60 yrs OR history of thrombosis	▪ All the above, AND ▪ Cytoreductive therapy ▪ First line ▪ Hydroxyurea ▪ IFN-α ▪ Busulfan* ▪ Second line ▪ Ruxolitinib ▪ IFN-α

*For pts > 70 yrs of age.

- Indications for cytoreduction in low-risk pts with:
  - Frequent phlebotomy requirement
  - Progressive leukocytosis
  - Platelets > 1500 x 10^9/L (risk of bleeding)
  - Severe disease-related symptoms

Barbui T, et al. J Clin Oncol. 2011;29:761-770. Tefferi A, et al. Am J Hematol. 2015;90:162-173.

Recommendations for Second-line PV Therapy

Pt Characteristics	Options
Inadequate response or intolerance to HU	Ruxolitinib, IFN-α[1,2]
Inadequate response or intolerance to IFN-α	HU[1]
Short life expectancy	Busulfan, pipobroman, or ^{32}P[1]

1. Barbui T, et al. J Clin Oncol. 2011;29:761-770.
2. Ruxolitinib [package insert].

Slide credit: clinicaloptions.com

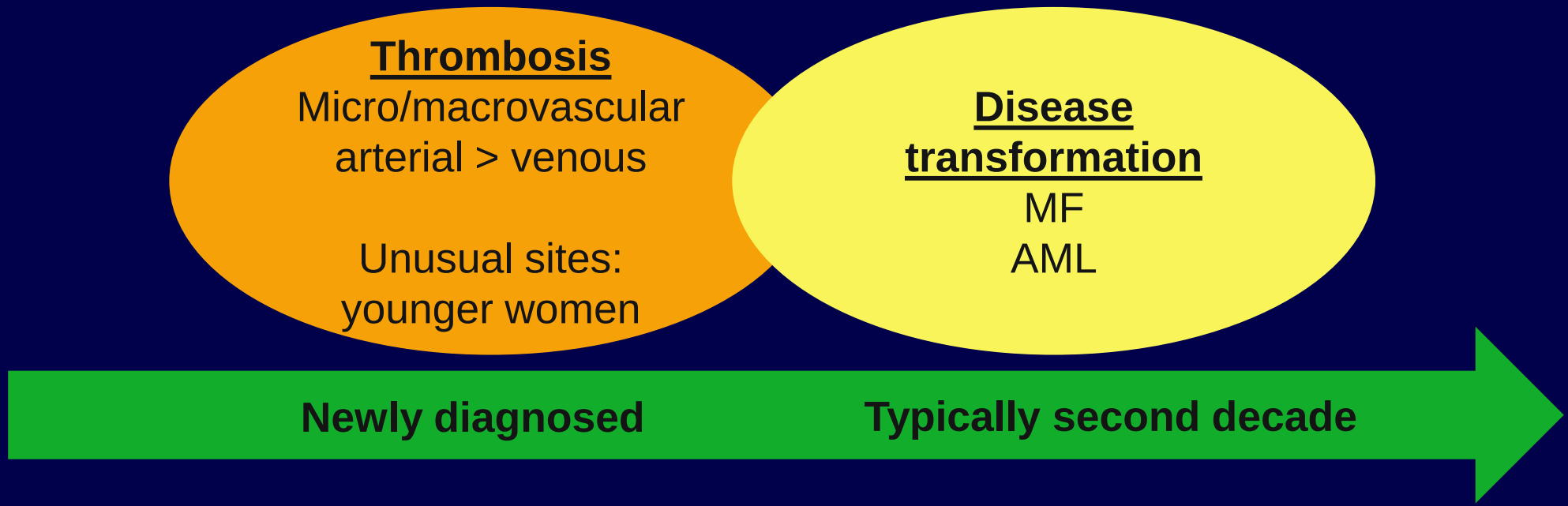
Clinical Complications of PV

Symptoms (*Independent of Risk*)

Cytokine: fatigue, pruritus, constitutional symptoms, bone pain

Vascular: headache, dizziness, numbness, decreased concentration, low mood, sexual issues

Disease evolution: splenomegaly, constitutional symptoms

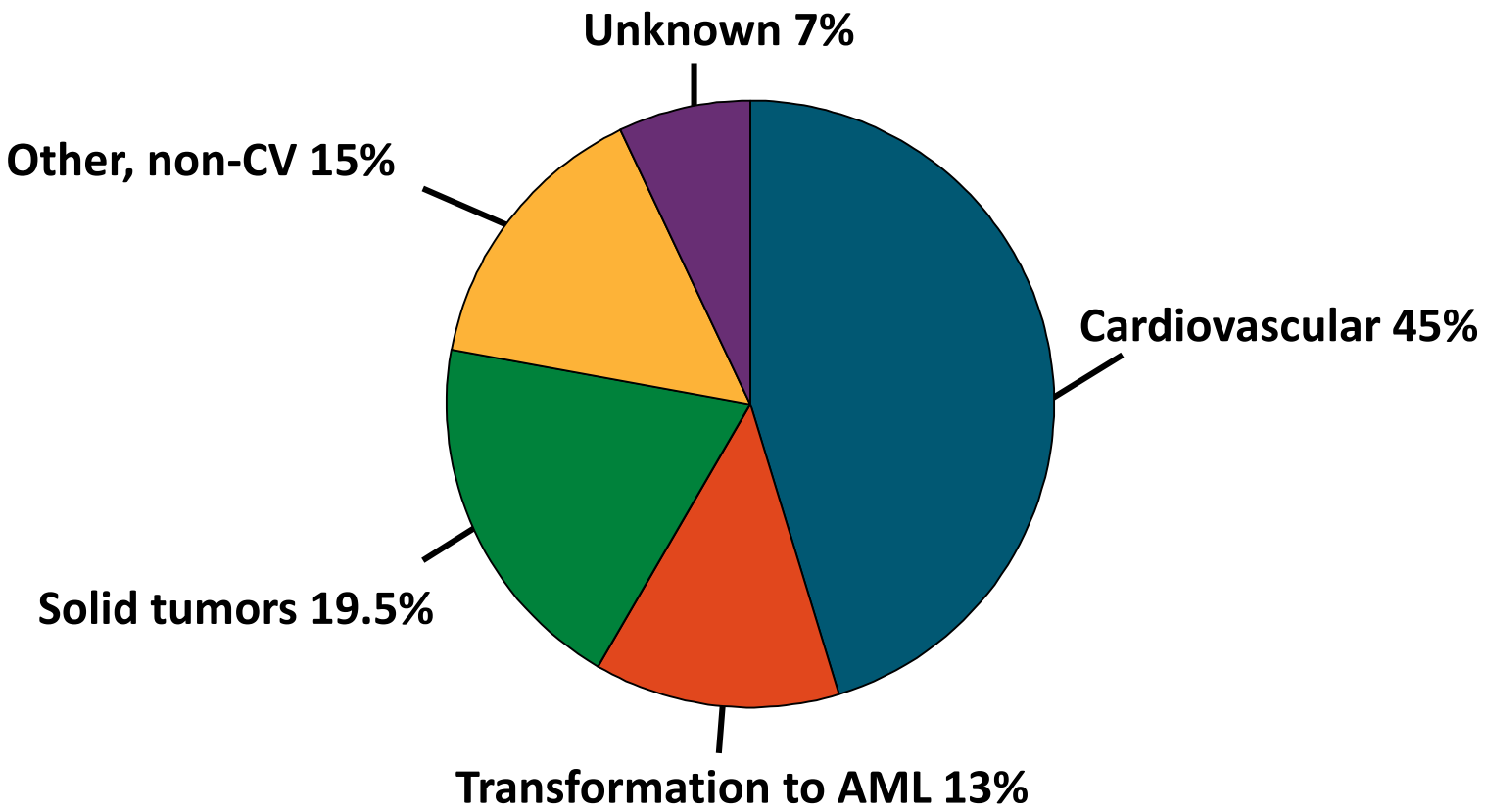


Mesa RA, et al. Cancer. 2007;109:68-76. Scherber R, et al. Blood. 2011;118:401-408. Geyer HL, et al. Blood. 2014;124:3529-3537. Stein BL, et al. Ann Hematol. 2014;93: 1965-1976. Stein BL, et al. Leuk Lymphoma. 2013;54:1989-1995.

Slide credit: clinicaloptions.com

Thrombosis: A Major Cause of Mortality in PV

- Data from large prospective multicenter project in PV (ECLAP trial); 164 of 1638 patients deceased at time of analysis



Marchioli. JCO. 2005;23:2224.

Slide credit: clinicaloptions.com

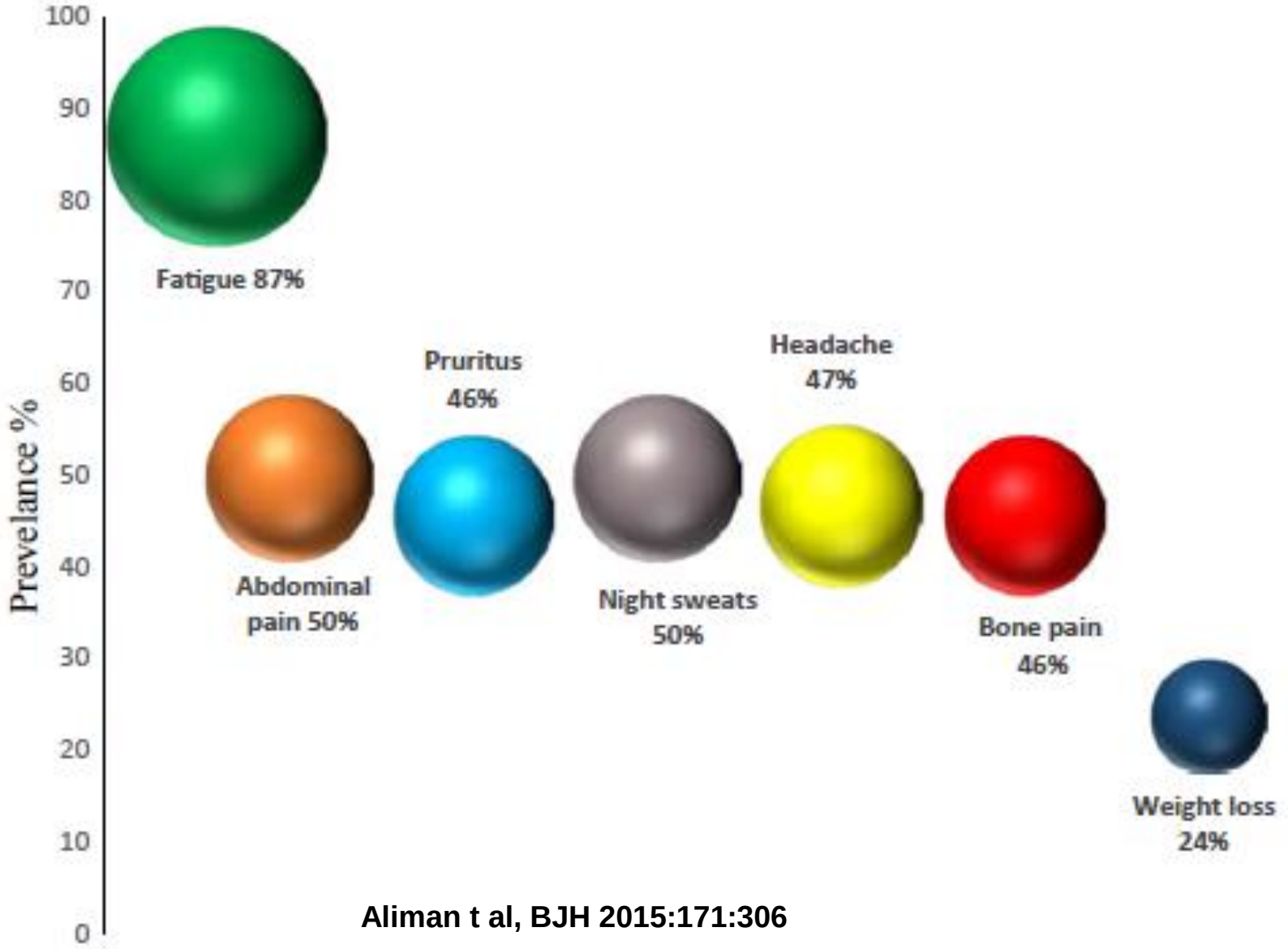
Trombocitemia essenziale TE

- **Definizione**
  - Disordine clonale mieloproliferativo cronico caratterizzato da trombocitosi (pst > 450.000 μL) con iperplasia megacariocitaria nel midollo.
- **Incidenza**
  - 1-2.5 casi 100.000 individui anno

TE: clinica

- Età: media 50 anni (range 40-70)
- M=F
- Esordio
  - Asintomatico
  - Manifestazioni trombotiche arteriose e venose (1/3 dei casi)
    - Distretti mesenterico, renale, portale, plenico
  - Manifestazioni emorragiche cutanee e mucose
    - Ematemesi e melena
    - S. di von Willebrand acquisita (Plt > 1.000.000 μL)
  - Manifestazioni neurologiche
    - Cefalea, parestesie, instabilità microcircolo piedi e emani (eritromelalgia), eritema e dolore urente alle estremità
    - Aborti in gravidanza
  - Splenomegalia

Prevalence of constitutional symptoms reported by ET patients



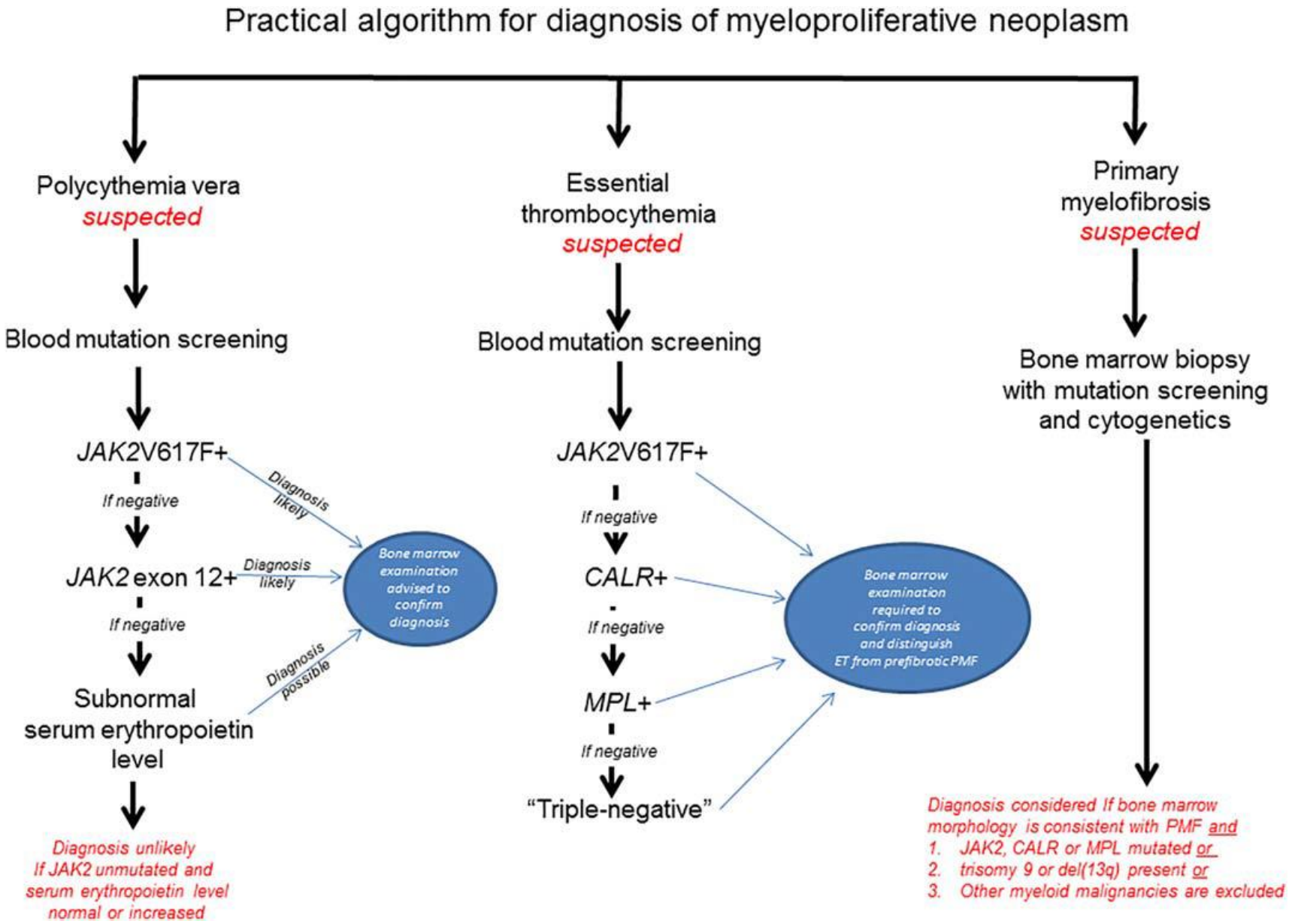
Aliman t al, BJH 2015:171:306

WHO Diagnostic Criteria: ET

ET Diagnosis
Requirement for diagnosis
▪ All 4 major criteria OR first 3 major criteria and the minor criterion
Major criteria
1. Platelet count ≥ 450 x 10^9/L 2. Proliferation mainly of megakaryocyte lineage with increased numbers of enlarged, mature megakaryocytes with hyperlobulated nuclei; no significant increase or left shift in neutrophil granulopoiesis or erythropoiesis and very rarely minor (grade 1) increase in reticulin fibers 3. Not meeting WHO criteria for other myeloid neoplasms 4. Presence of *JAK2, CALR,* or *MPL* mutation
Minor criteria
1. Presence of a clonal marker or absence of evidence for reactive thrombocytosis

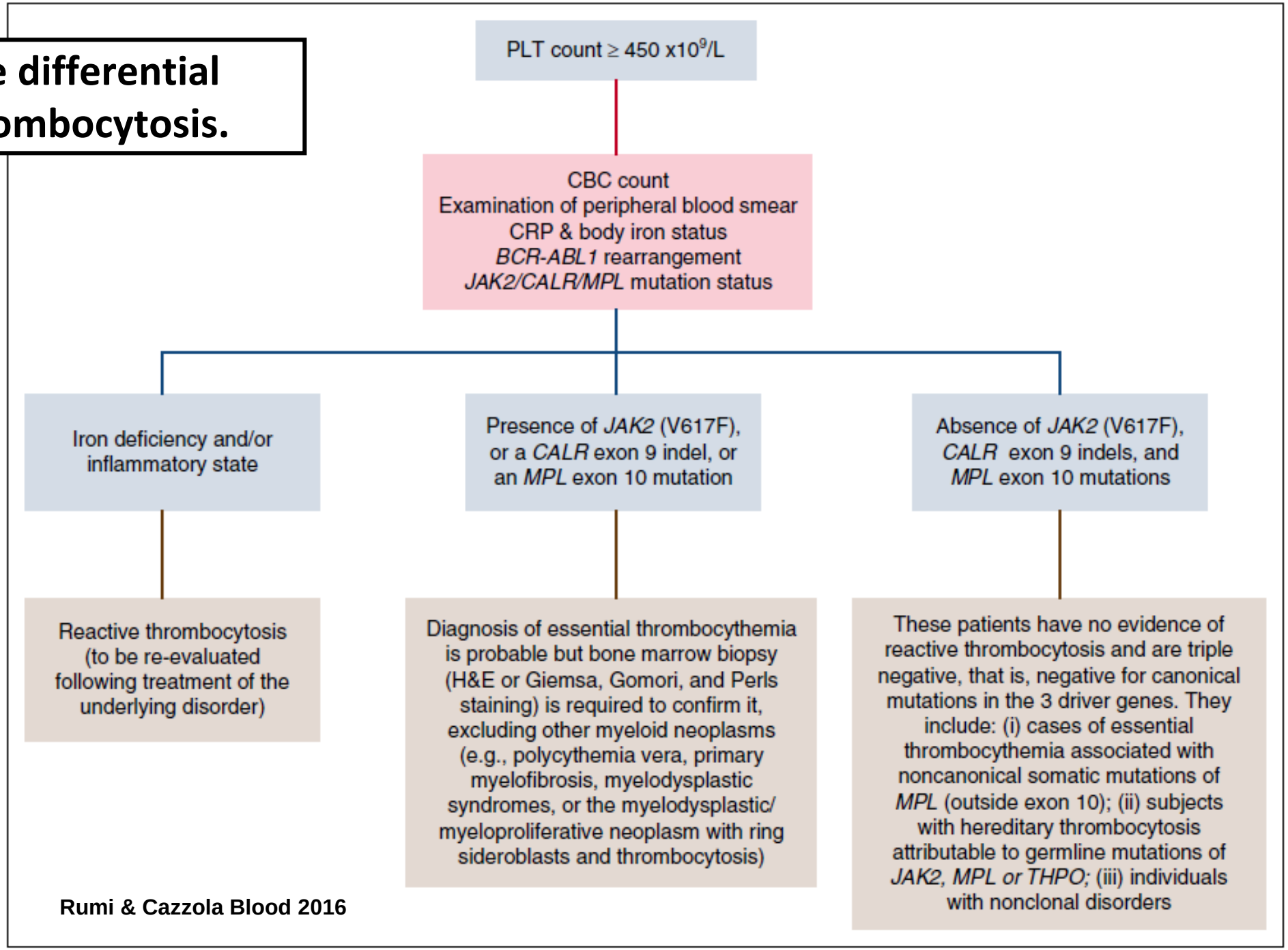
Footnotes in slidenotes.

Arber. Blood. 2016;127:2391.



Tefferi & Barbui Am. J. Hematol. 92:95–108, 2016.

approach to the differential diagnosis of thrombocytosis.



Rumi & Cazzola Blood 2016

Familial thrombocytosis

- **Familial thrombocytosis (rare)**
  - High *Tpo* levels:
    - *Tpo* gene mutations
  - Activating mutation of *c-Mpl (Tpo-R)*
  - Others

Secondary thrombocytosis

- **Secondary thrombocytosis**

  - **Transient processes**
    - Acute blood loss
    - Recovery ("rebound") from thrombocytopenia
    - Acute infection or inflammation
    - Response to exercise
    - Drug reactions

  - **Sustained processes**
    - Iron deficiency
    - Hemolytic anemia
    - Asplenic state (eg, after splenectomy)
    - Chronic inflammatory or infectious diseases
    - Cancer

Differential Diagnosis of Thrombocytosis

Reactive Causes

- Iron deficiency anemia
- Post-surgery
- Splenectomy
- Infection
- Inflammation
- Connective tissue disease
- Metastatic cancer
- Lymphoproliferative disorders

Other Myeloid Disorders

- PV
- Primary MF
- Chronic myeloid leukemia
- MDS with deletion of 5q
- Refractory anemia with ring sideroblasts and thrombocytosis

Bleeker JS, et al. Thrombosis. 2011;2011:536062.

Slide credit: clinicaloptions.com

Criteria for evaluating the thrombotic risk of ET (IPSET)

Risk factor	HR	Score
Age > 60 years	1.50	1
Cardiovascular risk factors	1.56	1
Previous thrombosis	1.93	2
JAK2V617F	2.04	2

Low risk implies a score = 0–1; intermediate risk, score = 2; and high risk, score ≥ 3

CV risk factors: Hypertension, hypercolesterolemia, diabetes, smoking, congestive heart failure

Barbui et al. Leukemia. 2018;32:1057-1069.

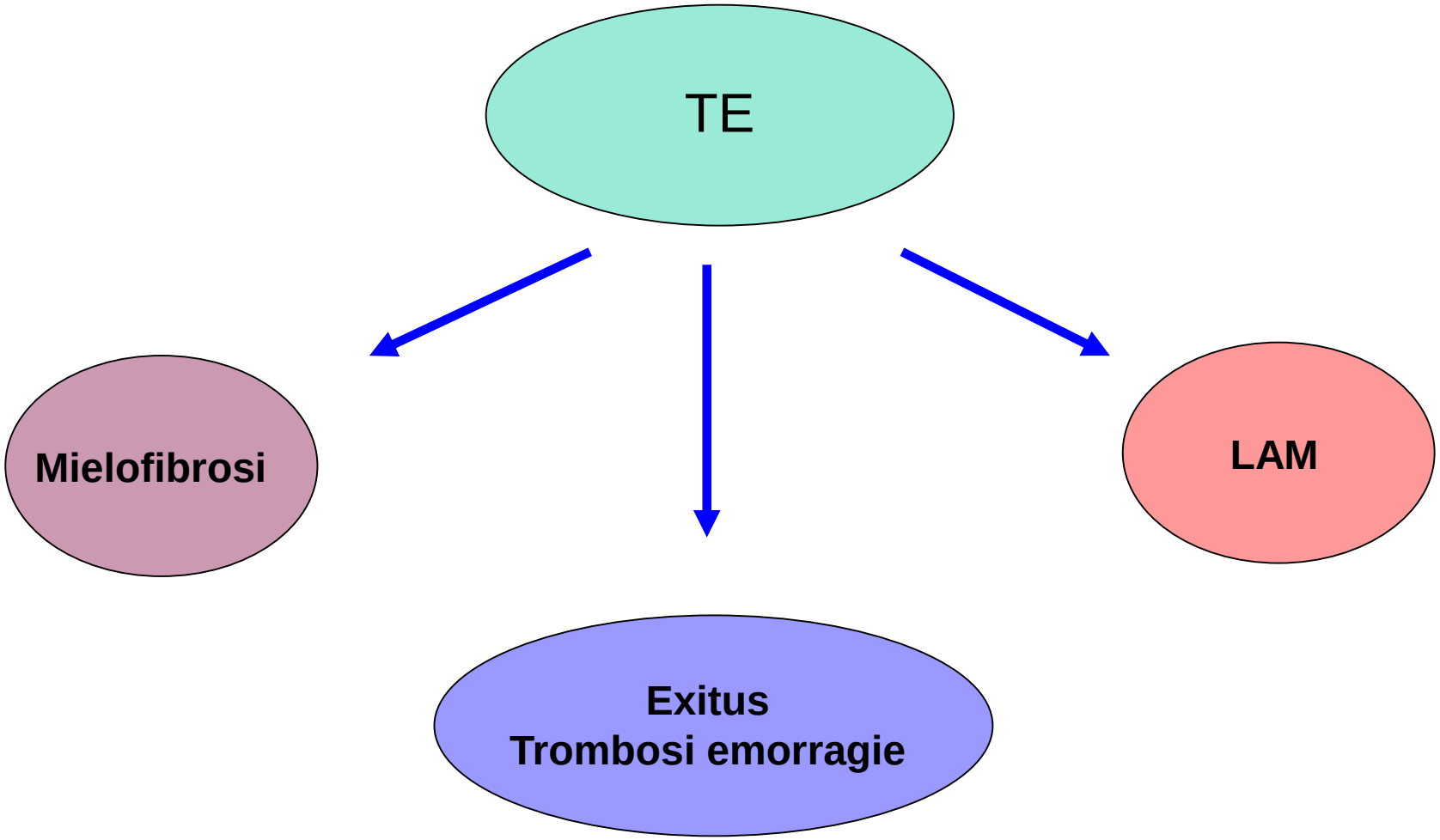
Thrombosis Risk-Adapted Management of ET and PV

<table>
<tr><th>Category</th><th>Characteristics</th><th colspan="2">Treatment</th></tr>
<tr><td>Low risk</td><td>Age ≤ 60 yrs AND no history of thrombosis</td><td colspan="2">▪ Therapeutic phlebotomy (goal Hct < 45%) in PV
▪ Aspirin 81 mg/day for ET/PV
▪ Address CV modifiable risk factors for ET/PV</td></tr>
<tr><td rowspan="3">High risk</td><td rowspan="3">Age > 60 yrs OR history of thrombosis</td><td colspan="2">▪ All the above AND cytoreductive therapy</td></tr>
<tr><td colspan="2">Cytoreductive therapy</td></tr>
<tr><td>First line</td><td>Second line</td></tr>
<tr><td>▪ Hydroxyurea for ET/PV
▪ Anagrelide for ET
▪ PegIFN for ET/PV</td><td>• Ruxolitinib for PV
• PegIFN for ET/PV
• Busulfan (age > 70 yrs) for ET/PV</td></tr>
</table>

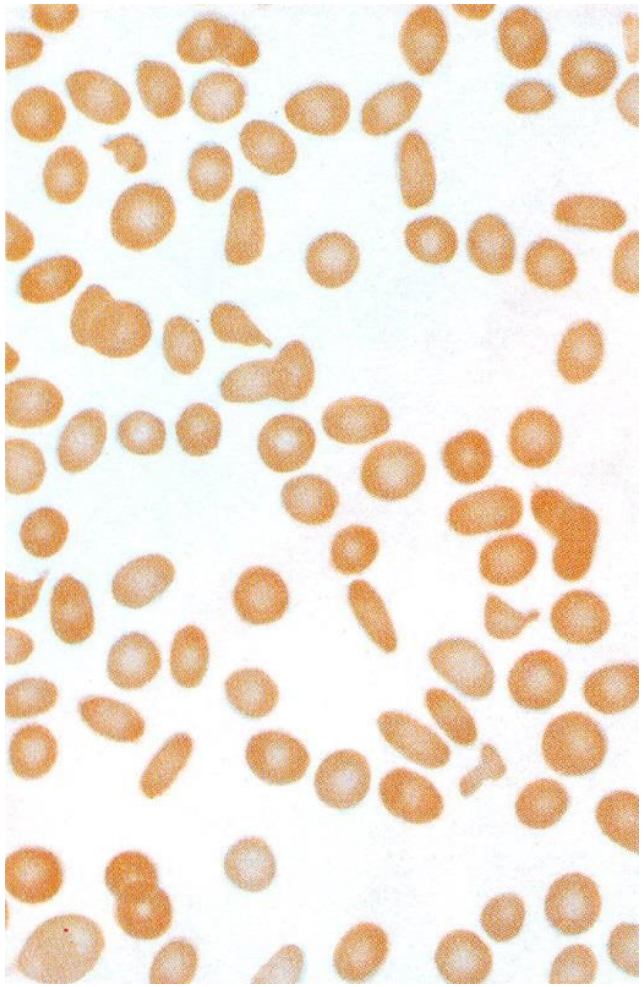
Barbui. JCO. 2011;29:761. Tefferi. Am J Hematol. 2015;90:162.

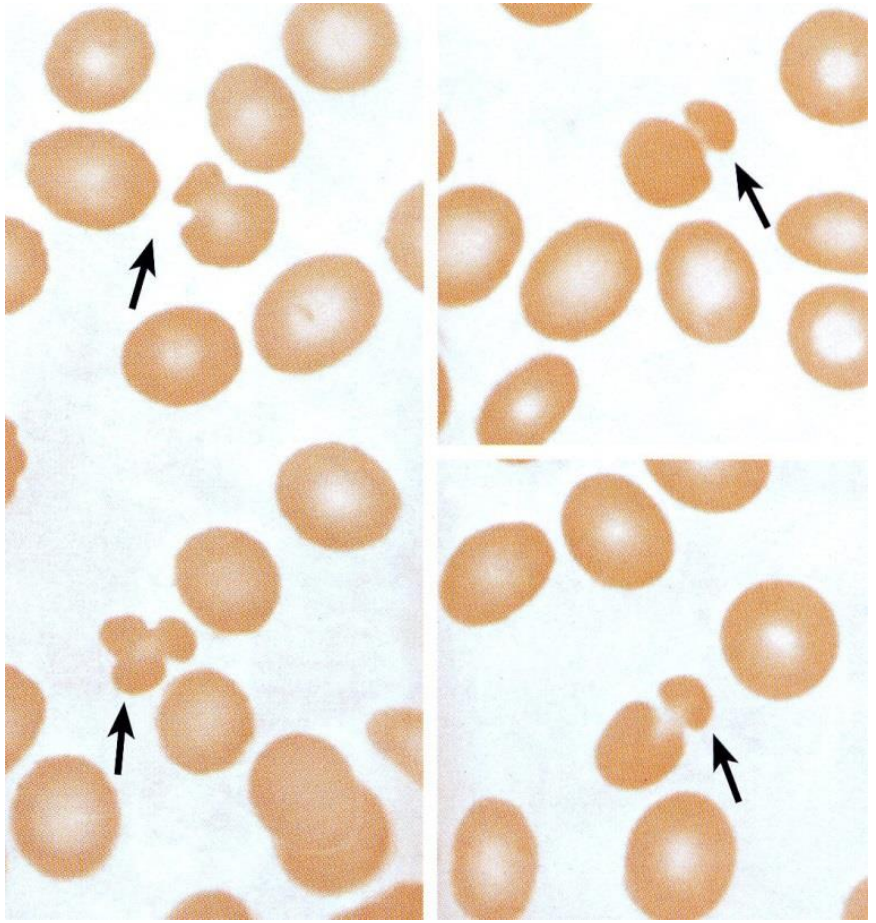
Slide credit: clinicaloptions.com

TE: evoluzione



Idiopathic myelofibrosis





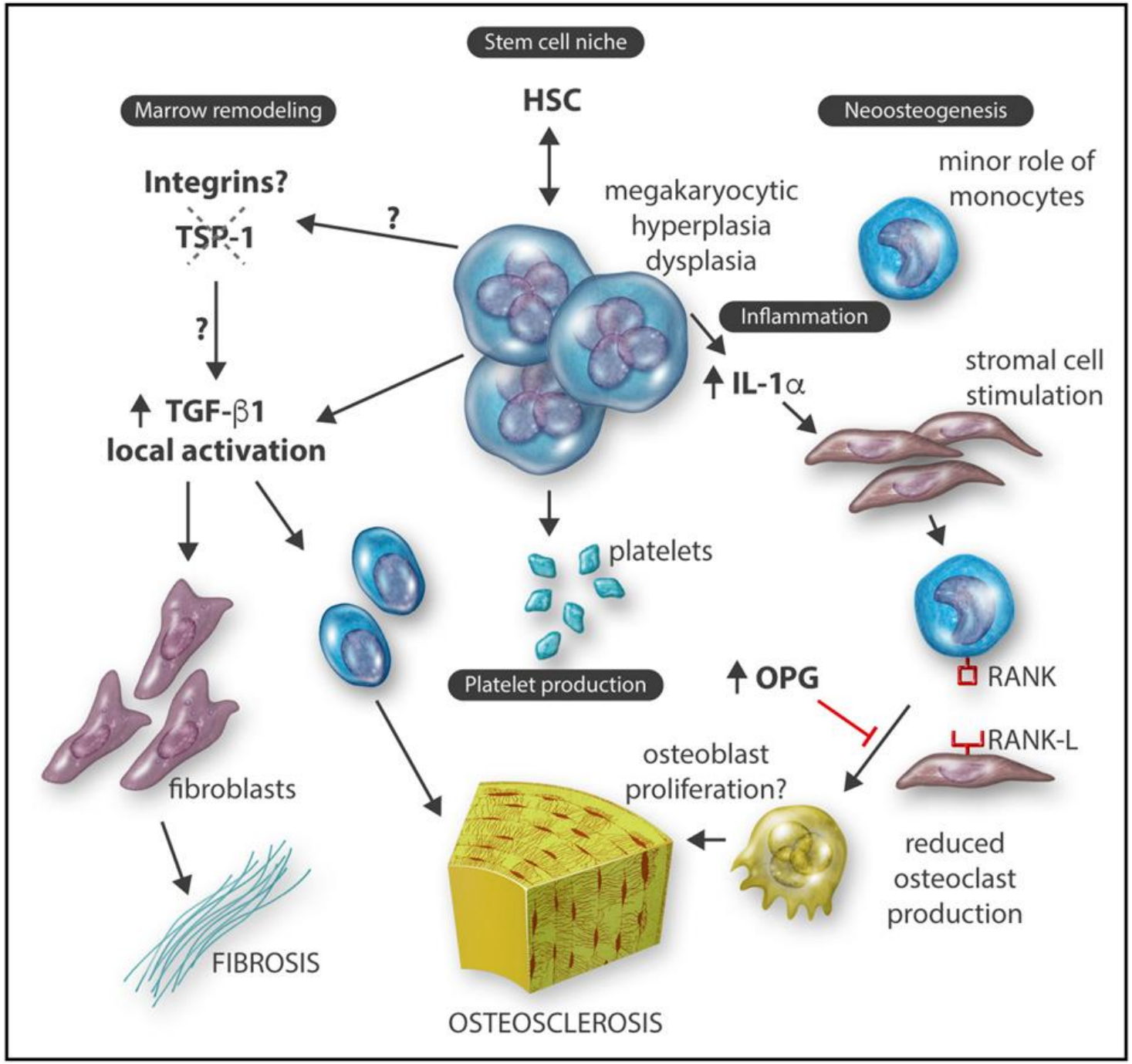
definition

- **Chronic myeloproliferative disorder characterised by:**
  - **Anemia**
  - **Splenomegaly**
  - **Immature granulocytes, erythroblasts, teardrop-shaped red cells and an increase in CD34+ cells in the blood**
  - **Marrow fibrosis**
  - **Osteosclerosis**
  - **Fibrohematopoietic tumors that can occur in virtually any tissue**

epidemiology

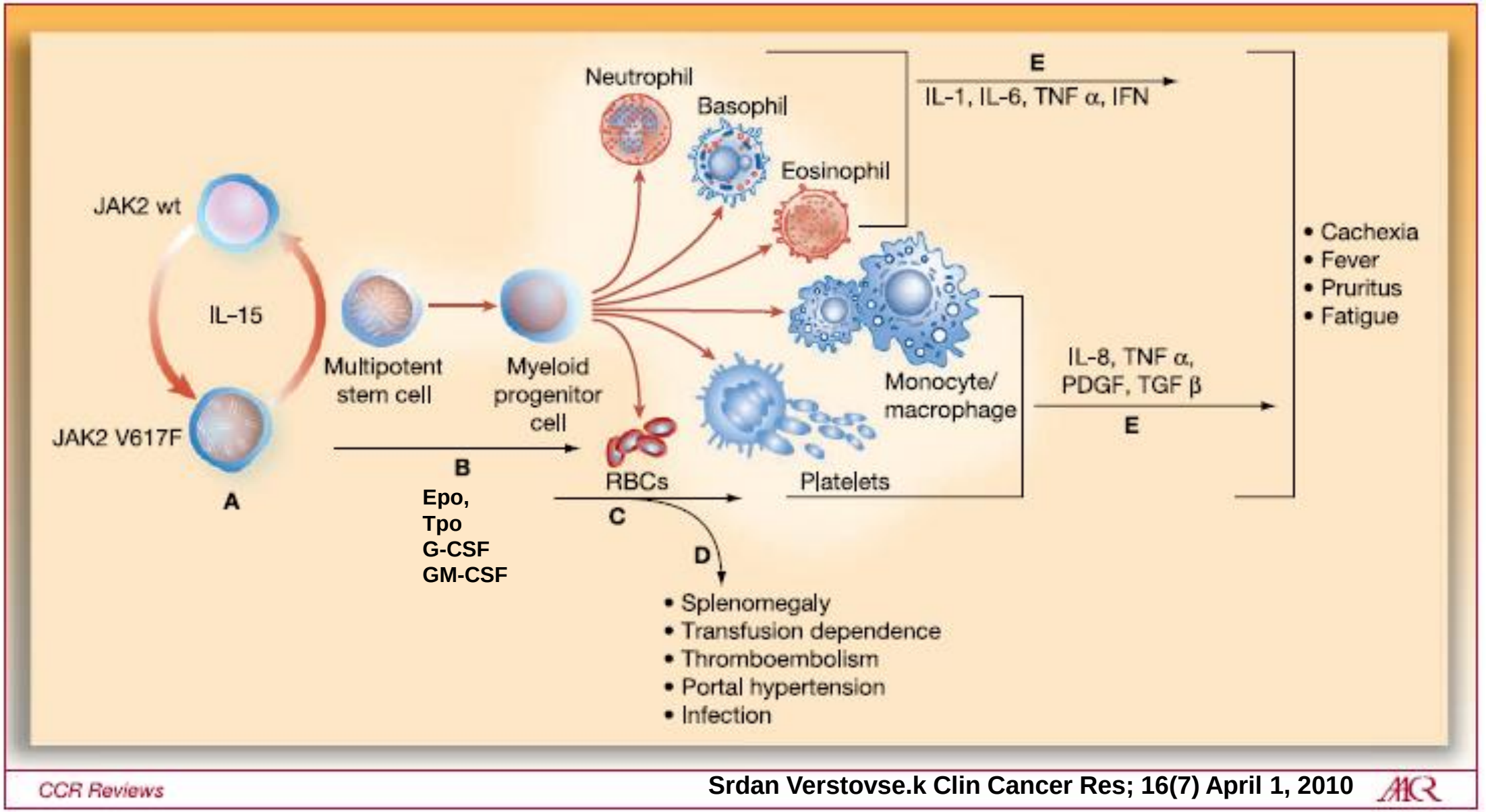
- **Incidence in western countries**
  - 0.4-0.7 new cases per 100.000 person/year
- **Median age at presentation 65 years**
  - 22% of patients are aged 55 years or less
- **Secondary complication** of polycythemia vera and essential thrombocythemia (rate 10-20% after 15-20 years of follow-up)
- 10-20% of patients have **leukemic transformation** in the first 10 years

MKs play a central role in MI pathogenesis.



Vainchenker W. Blood. 2017;129(6):667-679

Role of JAK2 signaling in the pathogenesis of splenomegaly, clinical manifestations, and constitutional symptoms in myelofibrosis.



CCR Reviews

Srdan Verstovse.k Clin Cancer Res; 16(7) April 1, 2010 AACR

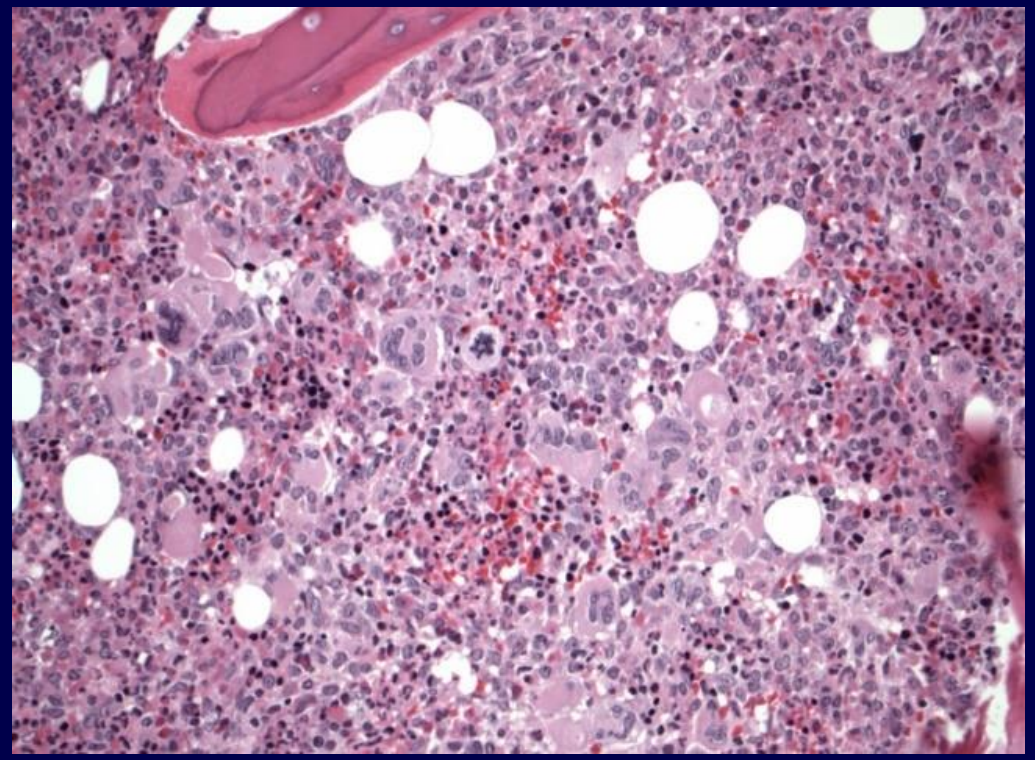
WHO Diagnostic Criteria: MF

Primary MF Diagnosis	
Requirement for diagnosis	
▪ All 3 major criteria AND ≥ 1 minor criteria	
Major criteria	
1. Megakaryocytic proliferation and atypia, **without reticulin fibrosis > grade 1 (prefibrotic PMF)** or **with reticulin and/or collagen fibrosis grade 2/3 (overt fibrotic PMF)**	
2. *JAK2*, *CALR*, or *MPL* mutation, presence of other clonal markers* OR absence of reactive MF	
3. Not meeting WHO criteria for other myeloid malignancies	
Minor criteria	
1. Anemia not attributed to a comorbid condition	3. Palpable splenomegaly
2. Leukocytosis ≥ 11 x 10^9/L	4. LDH increased above ULN
	5. **Leukoerythroblastosis (overt fibrotic PMF)**

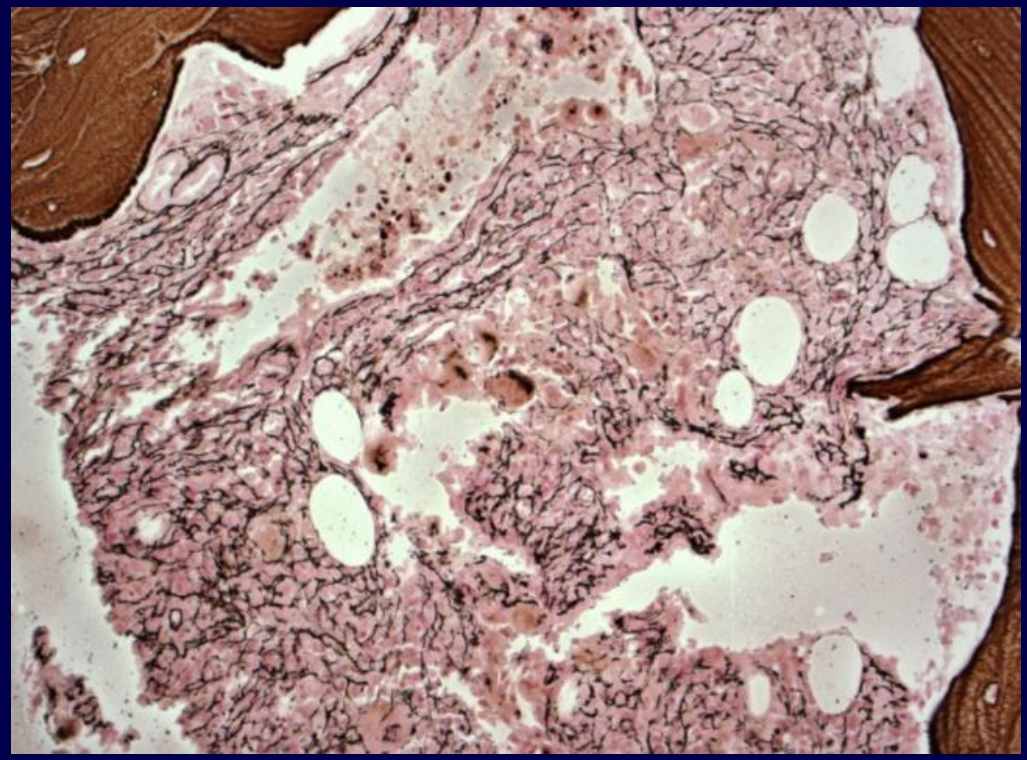
*eg, *ASXL1, EZH2, TET2, IDH1/IDH2, SRSF2, SF3B1*.

Arber. Blood. 2016;127:2391.

Primary MF



Clustering, atypical megakaryocytes



Bone marrow reticulin fibrosis

Faculty communication.

Slide credit: clinicaloptions.com

Myelofibrosis grading	
MF-0	Scattered linear reticulin with no intersections (crossovers) corresponding to normal BM
MF-1	Loose network of reticulin with many intersections, especially in perivascular areas
MF-2	Diffuse and dense increase in reticulin with extensive intersections, occasionally with focal bundles of thick fibers mostly consistent with collagen, and/or focal osteosclerosis*
MF-3	Diffuse and dense increase in reticulin with extensive intersections and coarse bundles of thick fibers consistent with collagen, usually associated with osteosclerosis*

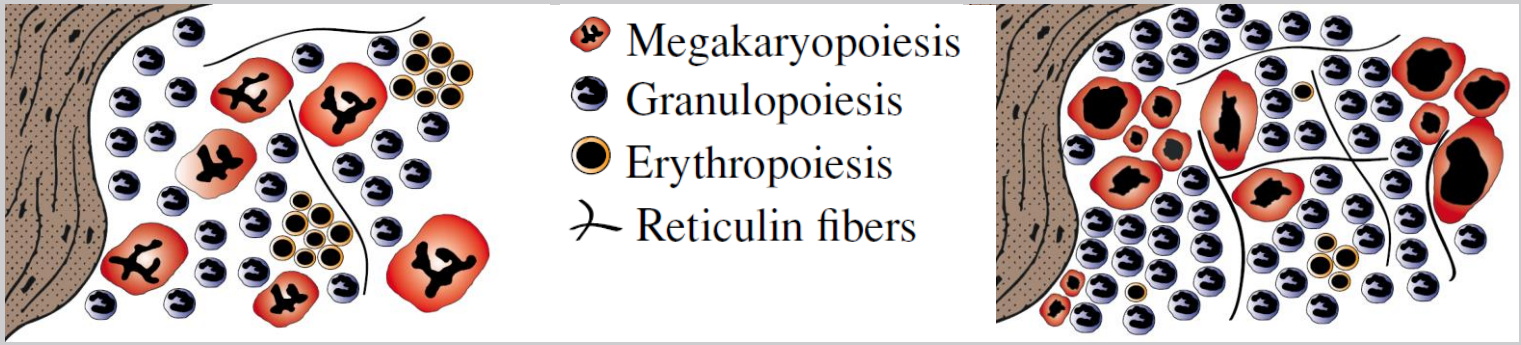
Semiquantitative grading of BM fibrosis (MF) with minor modifications concerning collagen and osteosclerosis. Fiber density should be assessed only in hematopoietic areas.

*In grades MF-2 or MF-3 an additional trichrome stain is recommended.

Arber DA et al. Blood. 2016; 127(20):2391-2405

ET vs Prefibrotic PMF: Morphologic Characteristics

Characteristic	ET	Prefibrotic PMF
Age-matched cellularity	▪ No or slight increase	▪ Marked increase
Granulopoiesis/ erythropoiesis	▪ No significant increase	▪ Pronounced proliferation of granulopoiesis, reduction of erythroid precursors
Histology	▪ Large/giant mature megakaryocytes ○ Hyperlobulated or deeply folded nuclei ○ Dispersed or loosely clustered in the marrow space	▪ Medium to giant megakaryocytes ○ Hypolobulated, hyperchromatic, bulbous, or irregularly folded nuclei with aberrant nuclear/cytoplasmic ratio ○ Dense or loose clustering and frequent endosteal translocation
Increase in reticulin fibers	▪ None or minor	▪ None or not significant



Thiele. Blood. 2011;117:5710. Republished with permission of American Society of Hematology, from Thiele. Blood. 2011;117:5710; permission conveyed through Copyright Clearance Center, Inc.

Slide credit: clinicaloptions.com

Post-ET vs Post-PV Myelofibrosis

Parameter	Post-ET MF	Post-PV MF
Clinical features	2 of the following: ▪ ≥ 1 constitutional symptom ▪ Increasing splenomegaly (> 5 cm, or newly palpable) ▪ Anemia and Hb decline ≥ 2 g/dL ▪ Increased LDH ▪ Leucoerythroblastic blood smear	2 of the following: ▪ ≥ 1 constitutional symptom ▪ Increasing splenomegaly (≥ 5 cm, or newly palpable) ▪ Anemia or loss of phlebotomy/ cytoreductive requirement ▪ Leucoerythroblastic blood smear
Bone marrow fibrosis	Grade 2/3 on a 0-3 scale	
Prognosis	Is the prognosis different than that of primary MF?	

Note: documentation of prior diagnosis of ET or PV (WHO criteria) required.

Barosi G, et al. Leukemia. 2008;22:437-438.

If Diagnostic Criteria for MF Not Met, What Are Other Causes of Fibrosis?

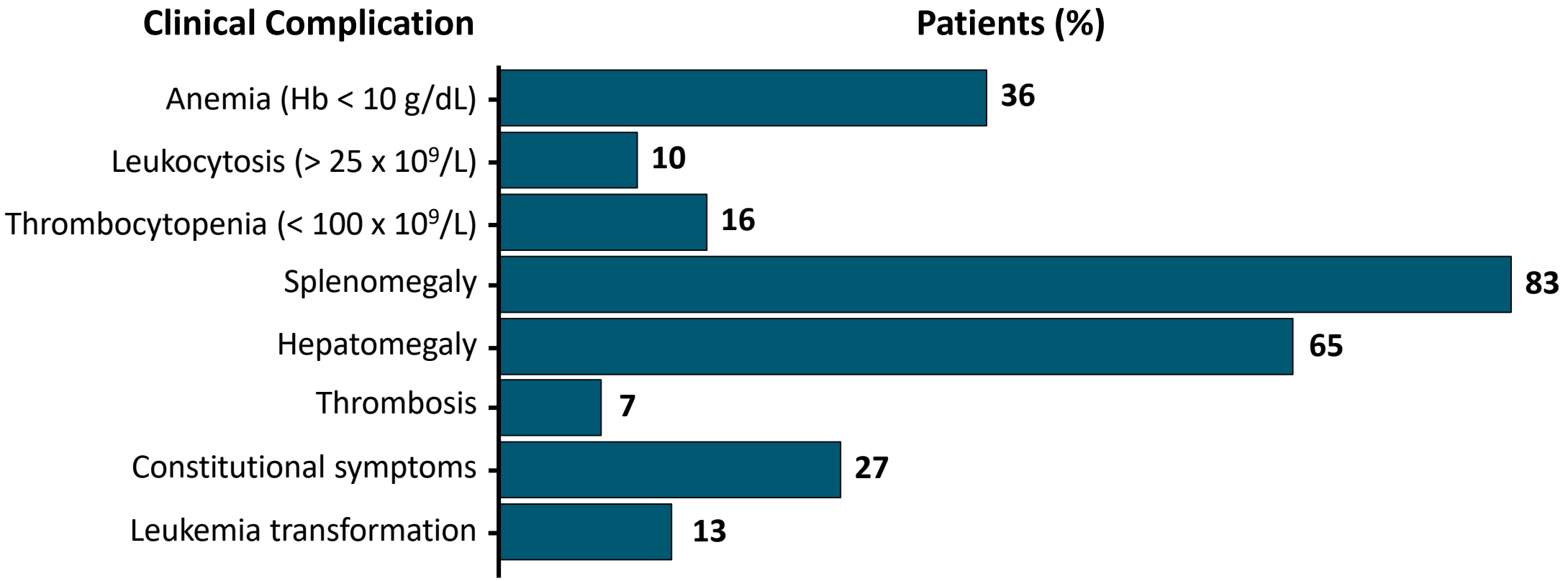
- Autoimmune MF
- Acute panmyelosis
- MDS with fibrosis
- Hodgkin and non-Hodgkin lymphoma
- Hairy cell leukemia
- Bone marrow metastases
- Secondary hyperparathyroidism
- HIV, tuberculosis
- Medication induced (TPO agonists)

Kuter DJ, et al. Br J Haematol. 2007;139:351-362.

presentation

- Heterogeneous presentation
  - Asymptomatic patients
  - Symptomatic patients
    - Splenomegaly
    - Anemia
    - Constitutional symptoms

Main Clinical Complications in MF



- Common symptoms derived from complications: bone pain, pruritus (myeloproliferation), night sweats, weight loss, fever (constitutional), early satiety, abdominal discomfort (splenomegaly), fatigue, insomnia

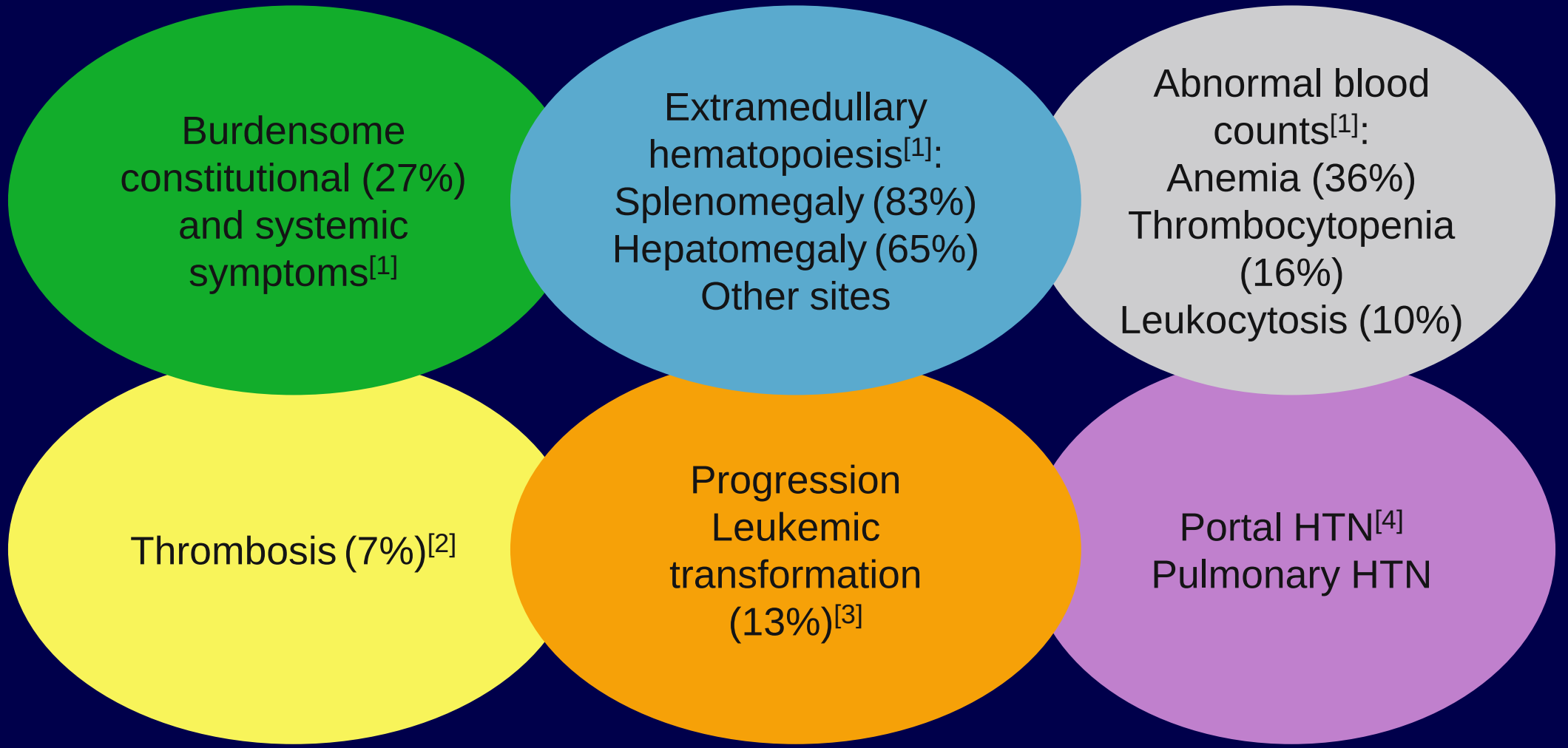
Passamonti. Blood. 2010;115:1703. Barbui. Blood. 2010;115:778. Passamonti. Blood. 2010;116:2857. Scherber. Blood. 2011;118:401.

Slide credit: clinicaloptions.com

Hematologic features

- Leukopenia
- Leukocytosis
  - (leukoerythroblastosis)
- Thrombocytopenia
- Thrombocytosis
- Anemia
  - (dacriocytes)
- Increased LDH

Clinical Complications of MF



1. Passamonti F, et al. Blood. 2010;115:1703-1708. 2. Barbui T, et al. Blood. 2010;115:778-782.
3. Passamonti F, et al. Blood. 2010;116:2857-2858. 4. Mesa RA. Blood. 2009;113:5394-5400.

Prognosis

- Median survival: 3.5-5 years
  - Wide variability
- Adverse prognostic factors
  - Constitutional symptoms
  - Hb < 10 g/dL
  - WBC count < 4 or > 30 x 10^9/L
  - Blood blasts > 1%
  - cytogenetics
  - Type of mutation

International Prognostic Scoring System: Risk Classification of MF at Presentation

- Risk factors
  - Older than 65 yrs of age
  - Constitutional symptoms
  - Hb < 10 g/dL
  - WBC count > 25 x 10^9/L
  - Peripheral blood blasts ≥ 1%

No. Risk Factors	Risk Group	Median Survival, Yrs
0	Low	11.3
1	Intermediate-1	7.9
2	Intermediate-2	4.0
≥ 3	High	2.3

Cervantes F, et al. Blood. 2009;113:2895-2901.

IPSS and Dinamic IPSS (plus) prognostic scoring systems

Risk factors	Point value		IPSS			DIPSS		
	IPSS	DIPSS	Risk group	Risk score	Median survival	Risk group	Risk score	Median survival
Age >65	1	1	Low	0	11.3 years	Low	0	Not reached
Constitutional symptoms[a]	1	1	Intermediate-1	1	7.9 years	Intermediate-1	1 to 2	14.2 years
Hb <10 g/dL	1	2	Intermediate-2	2	4 years	Intermediate-2	3 to 4	4 years
WBC count >25 $\times$ 10^9/L	1	1	High	$\geq$ 3	2.3 years	High	$\geq$ 5	1.5 years
Blood blasts $\geq$ 1%	1	1						

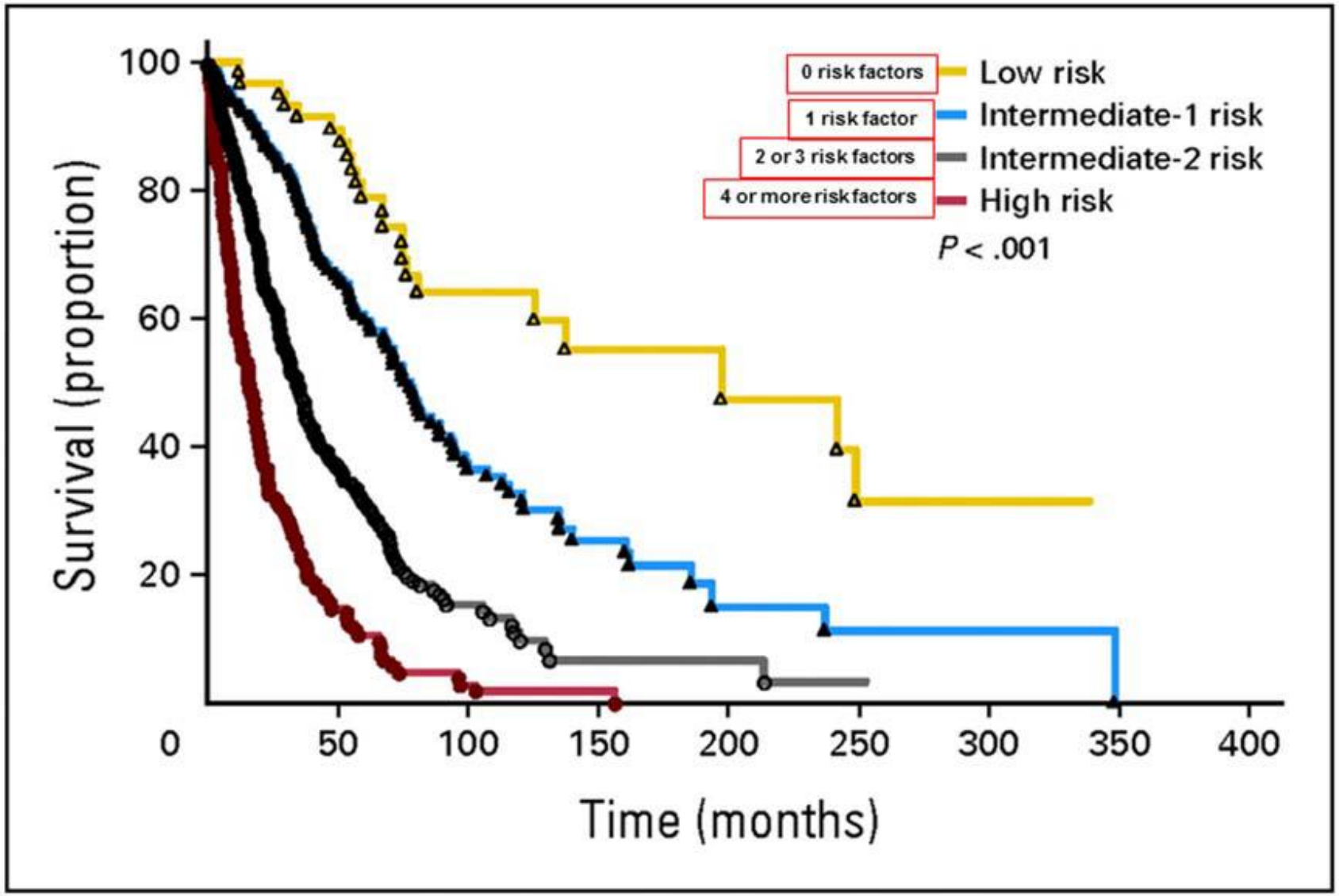
[a] Constitutional symptoms defined as weight loss >10% of the baseline value in the year preceding PMF diagnosis and/or unexplained fever or excessive sweats persisting for more than 1 month.

Risk factors	Points	DIPSS plus		
		Risk group	Risk score	Median survival
DIPSS intermediate-1	1	Low risk	0	15.4 years
DIPSS intermediate-2	2	Intermediate-1	1	6.5 years
High risk	3	Intermediate-2	2 to 3	2.9 years
Unfavorable karyotype[a]	1	High	4 to 6	1.3 years
Platelets <100 $\times$ 10^9/L	1			
RBC transfusion dependent	1			

[a] Unfavorable karyotype = complex karyotype or single or two abnormalities that include +8, −7/7q-, i(17q), −5/5q, 12p-, inv(3) or 11q23 rearrangement.

Survival data of 793 patients with primary myelofibrosis evaluated at time of their first Mayo Clinic referral and stratified by their Dynamic International Prognostic Scoring System (DIPSS-plus) that employs eight variables:

Age >65 yrs; Hgb <10 g/dL; RBC transfusion-dependent; platelets <100 x 10(9)/L; WBC > 25 x 10(9)/L; ≥1% circulating blasts; constitutional symptoms; karyotype.



Median
15.4 years
6.5 years
2.9 years
1.3 years

Gangat N et al. JCO 2011;29:392-397

Prognostic Impact of Driver and High Molecular Risk Nondriver Mutations in Primary MF

- Analysis of association between driver mutations and survival in pts with primary MF (N = 617)[1]

Driver Mutation	Pts, %	Median OS, Yrs
CALR mutated	22.7	17.7
JAK2 mutated	64.7	9.2
MPL mutated	4.0	9.1
Triple negative	8.6	3.2

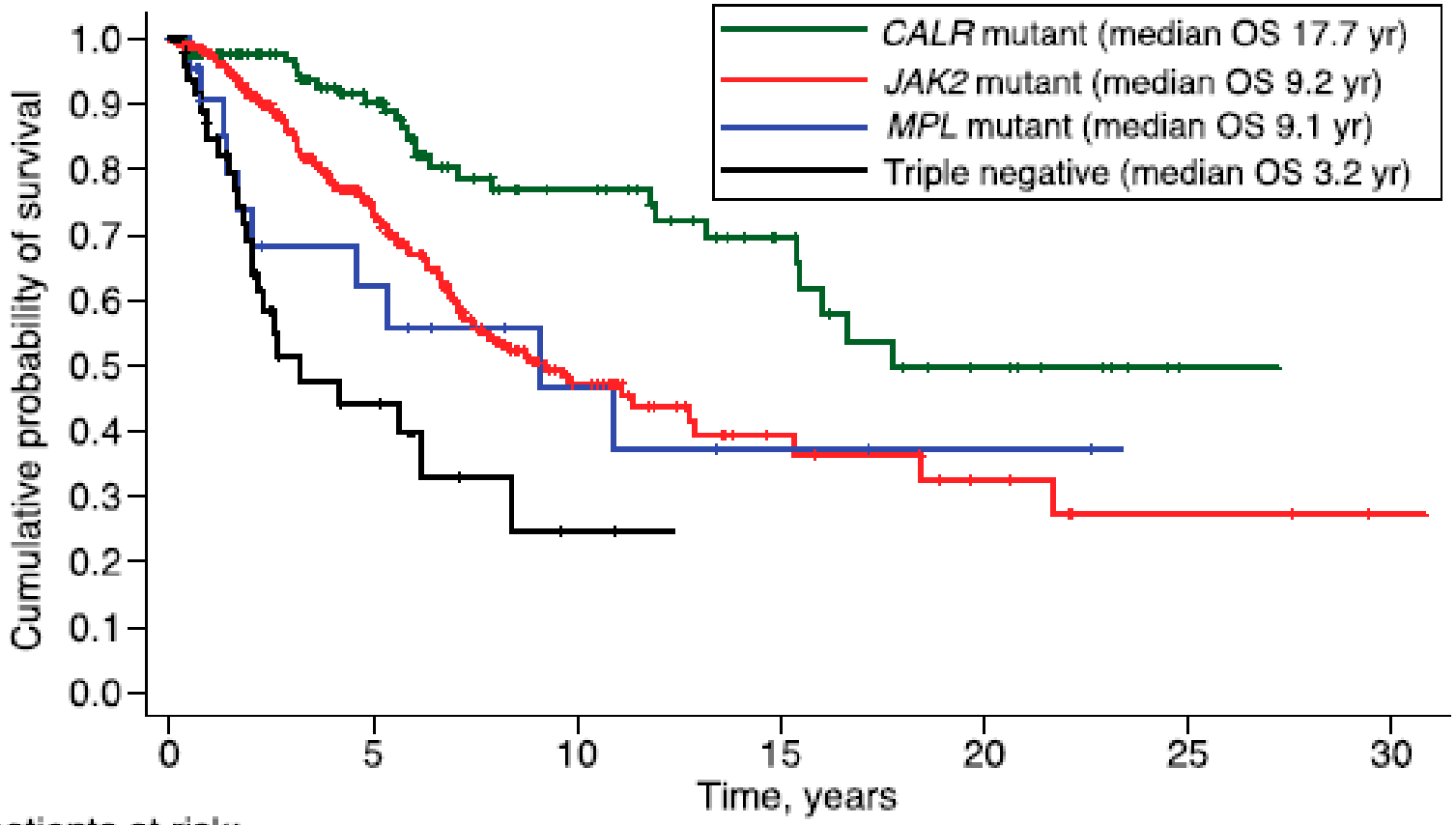
- Analysis of association between set of nondriver mutations (*IDH*, *EZH2*, *ASXL1*, *SRSF2)* and survival in pts with PMF (N = 797)[2]
  - Presence of mutations predicted decreased survival; ≥ 2 mutations predicted worst survival

1. Rumi E, et al. Blood. 2014;124:1062-1069. 2. Guglielmelli P, et al. Leukemia. 2014;28:1804-1810.

Slide credit: clinicaloptions.com

Survival in MI

Rumi et al, Blood. 2014;124(7):1062-1069)



No. of patients at risk:

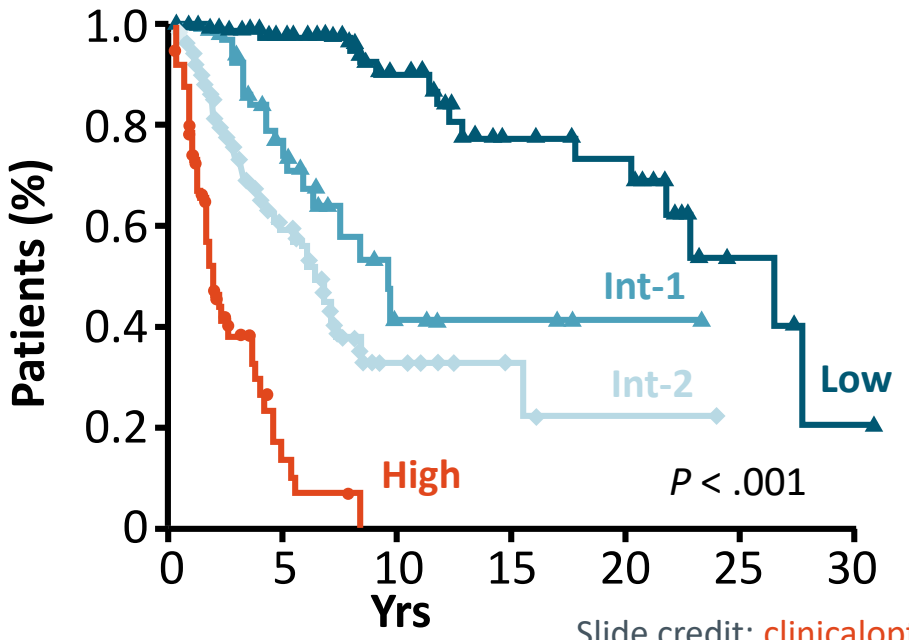
CALR mutant	140	72	37	19	9	1
JAK2 mutant	396	135	39	13	7	3
MPL mutant	25	10	5	3	2	0
Triple negative	53	11	2	0	0	0

MIPSS: Incorporating Mutational Data Into Risk Stratification

Variables	Multivariate Analysis HR (95% CI)	*P* Value	Weighted Value
Age > 60 yrs	3.8 (2.60-5.51)	< .0001	1.5
Hb < 100 g/L	1.4 (1.01-1.99)	.04	0.5
Constitutional symptoms	1.5 (1.13-2.16)	.007	0.5
Plt < 200 x 10^9/L	2.5 (1.77-3.42)	< .0001	1.0
Triple negativity	3.9 (2.20-6.80)	< .0001	1.5
JAK2/MPL mutation	1.8 (1.11-2.90)	.016	0.5
ASXL1 mutation	1.4 (1.06-1.99)	.02	0.5
SRSF2 mutation	1.7 (1.08-2.58)	.02	0.5

Akaike information criterion indicated that MIPSS performed better than IPSS in predicting survival (1611.6 *vs* 1649.0).

Risk Category	Score	% Pts	OS, Yrs	HR
Low	0-0.5	27	26.4	1
Int-1	1-1.5	13	9.7	4.7
Int-2	2-3.5	46	6.4	9.9
High	≥ 4	13	1.9	36.5



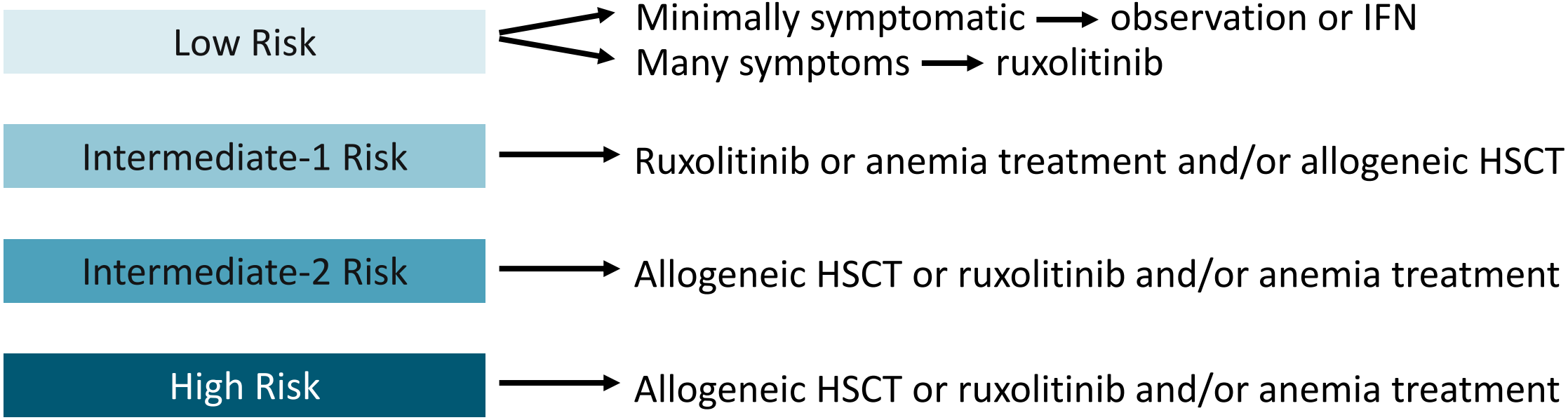
Vannucchi. ASH 2014. Abstr 405.

Slide credit: clinicaloptions.com

treatments

- Treatment options:
  - Supportive treatments (EPO)
  - Hydrossycarbamide
  - Steroids
  - JAK2 inhibitors (ruxolitinib)
  - Immunomodulating drugs (Imids)
  - Splenectomy
  - Radiotherapy
  - Allo-BMT
  - Telomerase inhibitors

MF Treatment: Based on Risk and MF-Related Symptoms/Signs



Mesa. Leuk Lymphoma. 2013;54:242. Geyer. Hematology Am Soc Hematol Educ Program. 2014;2014:277.

Slide credit: clinicaloptions.com

Needs-Oriented Therapy for MF

Clinical Issue	Treatments	
Anemia	▪ ESAs ▪ Corticosteroids ▪ Danazol	▪ Thalidomide, lenalidomide (IMiDs)
Symptomatic splenomegaly	▪ Ruxolitinib ▪ Hydroxyurea	▪ Cladribine, IMiDs ▪ Splenectomy
Constitutional symptoms/QoL	▪ Ruxolitinib ▪ Corticosteroids	
Extramedullary hematopoiesis	▪ Radiation therapy	
Hyperproliferative (early) disease	▪ Interferon	
Risk of thrombosis	▪ Low-dose ASA	
Accelerated/blastic phase	▪ Hypomethylating agents	
Improved survival	▪ Allogeneic HSCT ▪ Ruxolitinib	

Slide credit: clinicaloptions.com

Allogeneic HSCT for Patients With MF

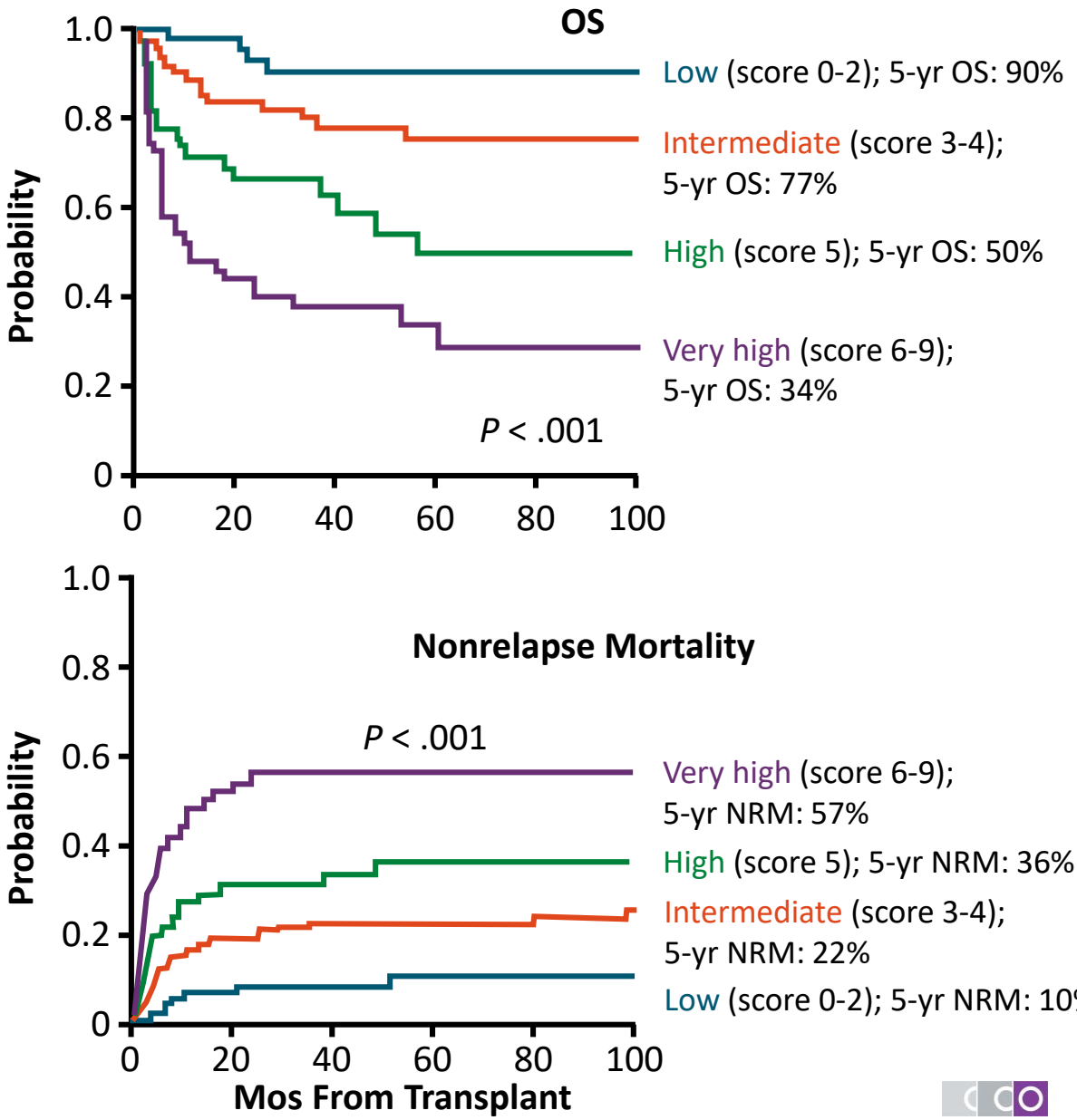
- **Who**: consider HSCT in **younger patients whose survival is expected to be < 5 yrs** (generally intermediate-2–risk/high-risk patients)[1]
- **But**: **very few MF patients undergo HSCT**
  - Traditionally **limited to younger patients** < 60 yrs of age and those with HLA-identical sibling match (although now possible up to 75 yrs of age)
  - **High transplant-related mortality and morbidity** associated with transplantation due to acute and chronic GvHD[2]
    - 1-yr NRM rate: 12% (completely matched donors) to 38% (mismatched)
    - 5-yr survival rate: 56% (matched sibling donors) to 34% (partially matched/ mismatched)

1. Cervantes. Blood. 2009;113:2895. 2. Kröger. Leukemia. 2015;29:2126.

Slide credit: clinicaloptions.com

Myelofibrosis Transplant Scoring System (MTSS)

Variable	Multivariable		Score
	HR (95% CI)	*P*	
Leukocyte count > 25 x 10^9/L	1.57 (1.16-2.41)	.007	1
Platelet count < 150 x 10^9/L	1.67 (1.16-2.40)	.006	1
Performance status < 90%	1.50 (1.06-2.13)	.021	1
CALR or *MPL*			
▪ Present			
▪ Absent	2.40 (1.30-4.71)	.012	2
Age ≥ 57 yrs	1.65 (1.15-2.36)	.006	1
HLA-mismatched unrelated	2.08 (1.45-2.97)	<.001	2
ASXL1	1.42 (1.01-2.01)	.041	1
Internal validation	AIC: 688.629 C-index original: 0.723 Bootstrap C-index: 0.712		



Gagelmann. ASH 2018. Abstr 689.

Slide credit: clinicaloptions.com