2nd edition

Unmet challenges in high risk hematological malignancies: from benchside to clinical practice

Turin, September 13-14, 2021

Starhotels Majestic

Scientific board:
Marco Ladetto (Alessandria)
Umberto Vitolo (Candiolo-TO)

Disclosures of NAME SURNAME

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Novartis					**x**	**x**	
Abbvie						**x**	
Sanofi							**x**

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Biology of high risk myeloproliferative disorders

Paola Guglielmelli

CRIMM- Centro Ricerca e Innovazione delle Malattie Mieloproliferative

Azienda Ospedaliera Universitaria Careggi

Università di Firenze

What Do We Mean by “High Risk”?

Polycythemia Vera
Essential Thrombocythemia

Thrombosis

Bleeding

Primary Myelofibrosis
Secondary Myelofibrosis
(PPV-MF;PET-MF)

Overall Survival

Progression to MF
Overall Survival

Thrombosis
Bleeding

Progression to Acute Leukemia

"Conventional" Risk Stratification System for Patients with PV and ET

PV

Risk Category	Age >60y and/or Prior Thrombosis
Low	No
--	--
High	Yes

ET

Risk Category	Age >60y and/or Prior Thrombosis	Generic CV Risk Factors*
Low	No	--
Interm.	--	Yes
High	Yes	--

*, smoking, hypertension, metabolic syndrome, obesity

Low Risk (no factor) — Thrombosis incidence: ET: 1.5 ×100 p/y; PV: 2.5 ×100 p/y

High Risk (≥1 factor) — Thrombosis incidence: ET: 2.0 ×100 p/y; PV: 5.0-10.9 ×100 p/y

The HR in MPN patients aged ≥60 years was **2.4** (CI, 2.1 to 2.6, p<0.001) compared to MPN patients <60 years. MPN patients with a history of thrombosis had a **2.7-fold** increased risk of thrombosis (HR 2.7, CI 2.5 to 2.9, p<0.001) compared to those with no previous thrombosis. (Swedish registry)

Extreme thrombocytosis (>1.5M x10^9/L) is associated with increased risk of hemorrhage

Marchioli R et al. *J Clin Oncol*. 2005;23:2224. Barbui T et al. *J Clin Oncol*. 2011;29:761. Hultchrants M et al, Ann Int Med 2018..

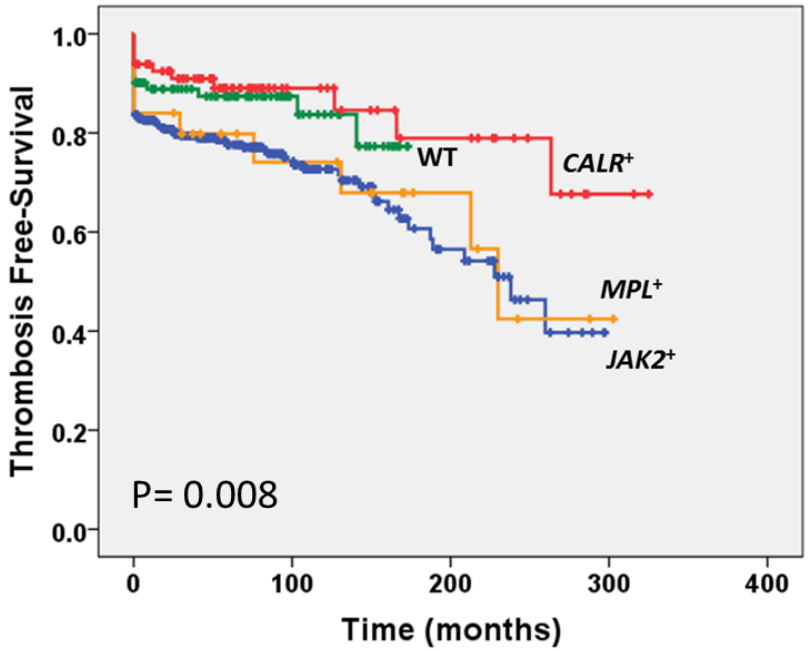
Limitations

- Above variables are «*not manageable*» (age, history)
- New factors have been proposed as candidate biomarkers for predicting vascular events (driver mutations, leucocytes count, inflammatory cytokines, CHIP(?))
- They do not account for the extreme heterogeneity of the disease
- Do not offer information in **conventionally defined low-risk patients**, that are thereby, by definition, the main «unmet» patient population
- Do not allow to infer about effectiveness of treatment

Does the Type of Driver Mutations Matter?

JAK2V617F	CALR
75% of all ET, PV, PMF	20% ET and PMF; extremely rare in PV
Older at presentation	Younger at presentation
Transformation to MF, AML ~ **10yr**	Transformation to MF, AML ~ **15-20 yr**
Thrombocytosis, polycythemia	Extreme thrombocytosis, ↓ hemoglobin
AVT, SVT, BCS tightly associated	AVT, SVT, BCS uncommonly associated
Leukocytosis tracks with JAK2V617F%	Leukocytosis less prominent, not related to VAF%

JAK2V617F Mutation Is Associated with Increased Risk of Thrombosis in ET



N=891

Risk factor	HR
Age > 60	1.50
CV risk factors	1.56
Previous thrombosis	**1.93**
***JAK2* V617F**	**2.04**

* Multivariate model adjusted for: sex, Hb, WBC and plt counts, HU and aspirin

- The cumulative incidence of thrombosis at 10 yr was 21.0% (95% CI, 16.6 to 25.7) **in *JAK2* mut** *Versus* **11.0%** (95% CI, 6.3 to 17.1) **in *CALR* mut** (P=0.003).

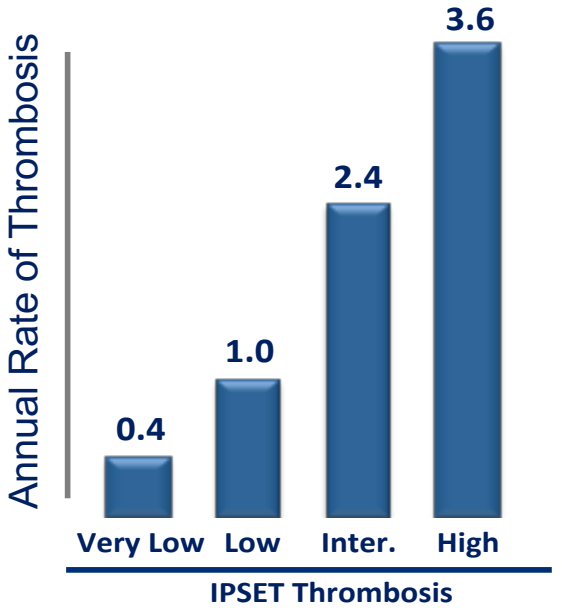
Vannucchi AM et al, Blood 2007; 110:840; Carobbio A *et al, Blood.* 2011;117:5857-9; Barbui T et al, Blood 2012.

Rotunno G, et al. *Blood.* 2014 Mar 6;123(10):1552-5; Rumi E, et al. *Blood. 2014* Apr 10;123(15):2416-9; Klampfl T, et al. *NEJM.* 2013;369:2379-90.

IPSET Score for ET

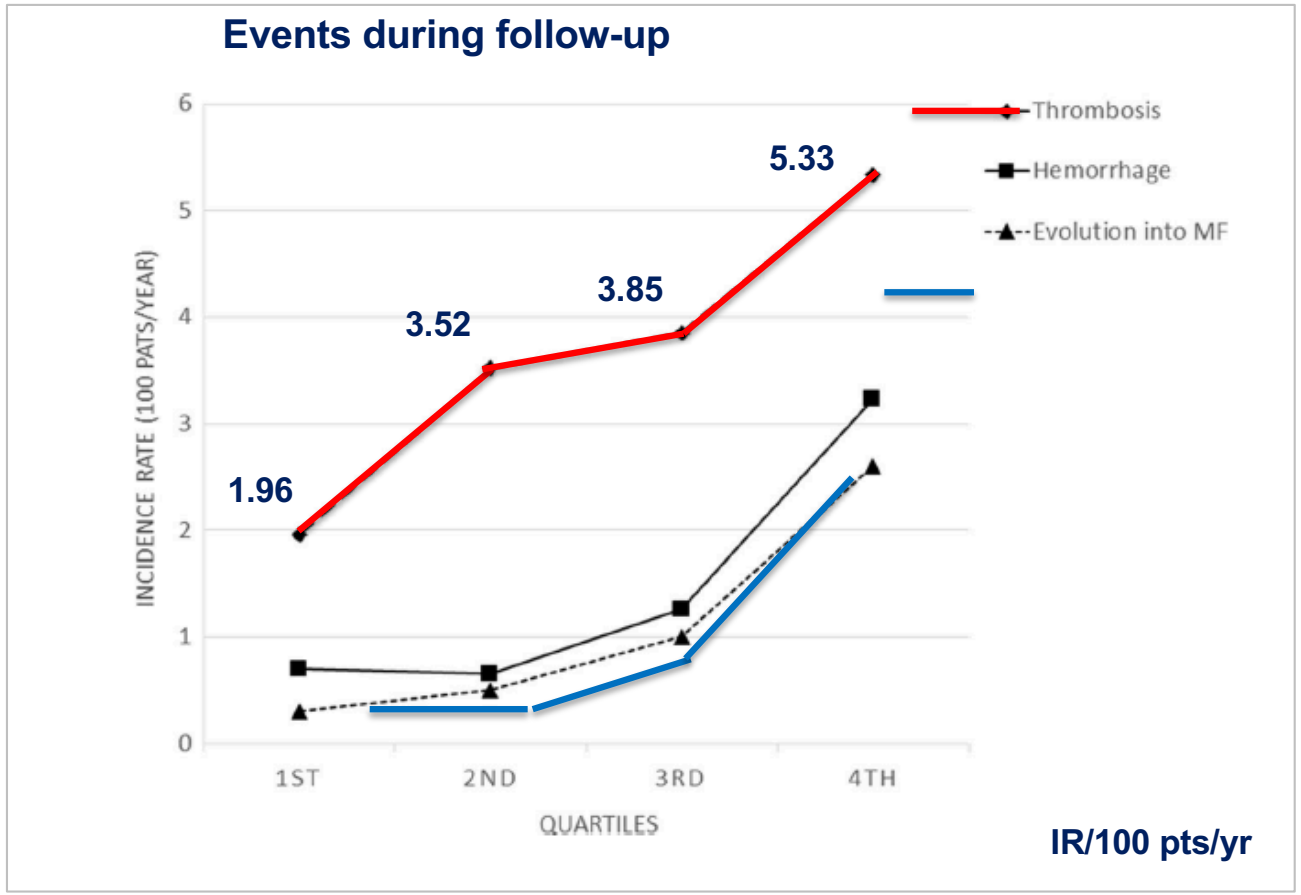
(IPSET= International Prognostic Score for Essential Thrombocytemia- Revised)

Risk Category	Variables included
Very-low	none
Low	*JAK2*V617F
Intermediate	Age >60 yr
High	Thrombosis history OR age + *JAK2*V617F



Tefferi A et al, AJH 2016; 91:390. Barbui et al. Blood 2012;120:5128.
Finazzi et al. Blood 2014 16;124:2611.;Barbui T et al BCJ 2015; 5:e369

Thrombosis and Hemorrhage are Common in MPN Patients with High *JAK2*V617F Allele Burden

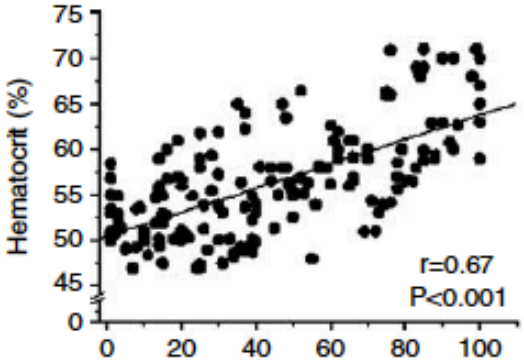


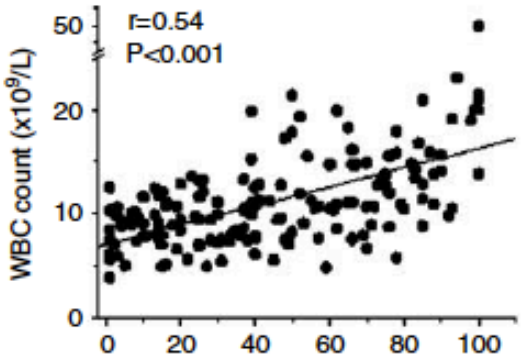
four quartiles (1st <25%, 2nd 26-50%, 3rd 51-75%, and 4th >75%)

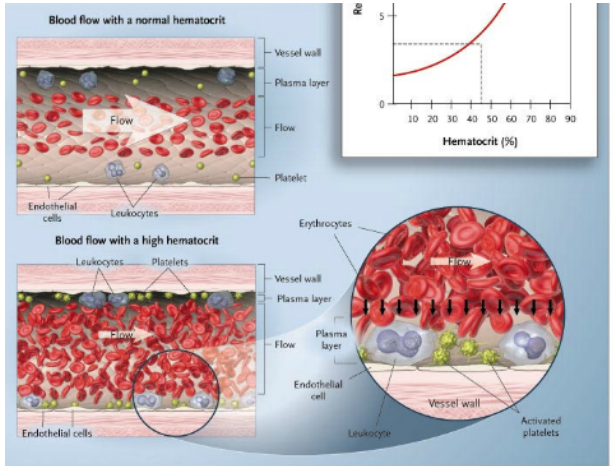
N=245, [ET=121, PV=124]

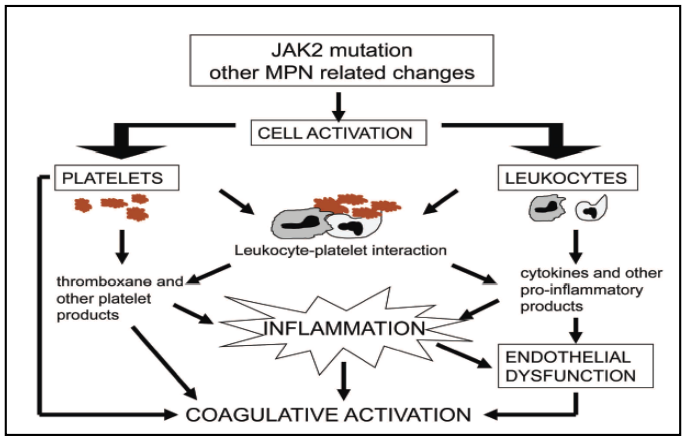
Bertozzi I et al, Ann Hematol 2017;42:E639;
Vannucchi AM et al, Blood 2007; 110:840; Vannucchi AM et al, Leukemia 2007; 21: 1952

Is the guilty *JAK2*V617F ± VAF *per se*, or the resulting downstream changes?





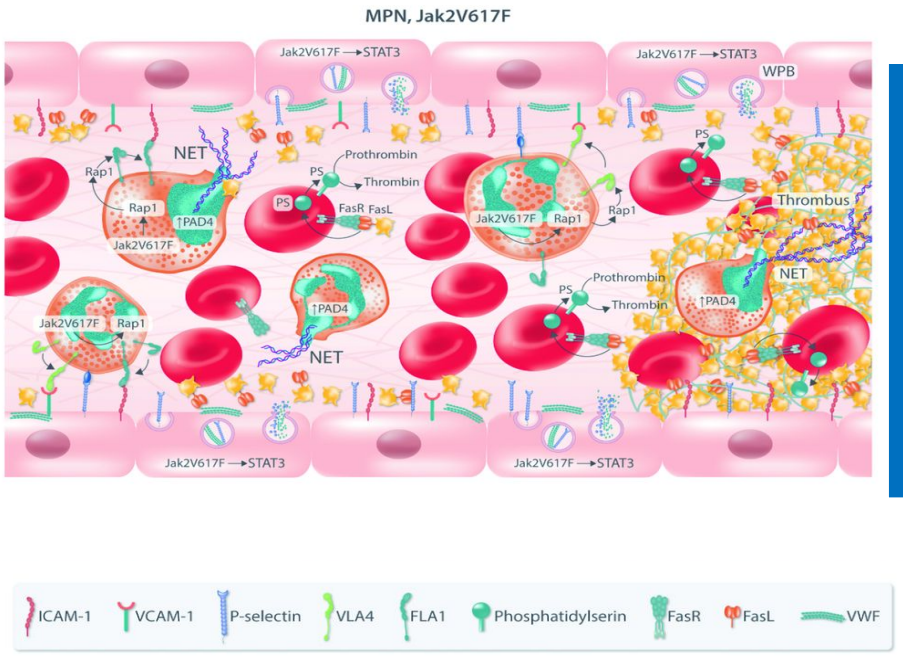




Vannucchi AM et al, Leukemia 2007; Spivak J, NEJM 2004.

Barbui et al, Haematologica 2011; Landolfi et al Haematologica 2011

The mechanism of thrombus formation in MPN



JAK2V617F

PLT

Increase in endothelial cell Weibel-Palade body degranulation of P-selectin and von Willebrand factor

WBC

Activation of the integrins LFA1 and VLA4; and increased neutrophil extracellular trap (NET) formation.

RBC

A red blood cell-platelet interaction through FasL/FasR

Bar-Natan M, Hoffman R. Haematologica 2019;104:3-6

Leucocytosis Is a Risk Factor for Thrombosis in PV and ET

- MPN is disease characterized by both **quantitative *and qualitative*** abnormalities in blood cells

- Studies addressing the relationship between WBC and thrombosis in MPNs have yielded somewhat consistent results.

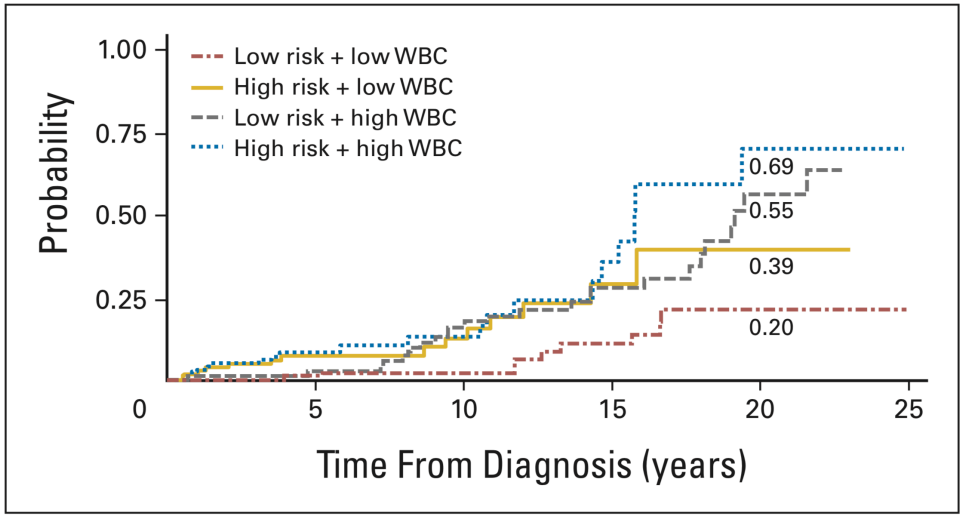
- Specific WBC threshold associated with increased risk remains unresolved

-> Mechanisms contributing to thrombosis likely to be complex

Table 3. Interaction of conventional risk categories and leukocyte count (multivariable model)

Risk factors	Hazard ratio (95% CI)
Low risk and low WBC count	1 (Reference)
Low risk and high WBC count*	3.1 (1.4-7.1)
High risk† and low WBC count	2.5 (1.0-6.0)
High-risk† and high WBC count*	5.0 (2.1-11.9)

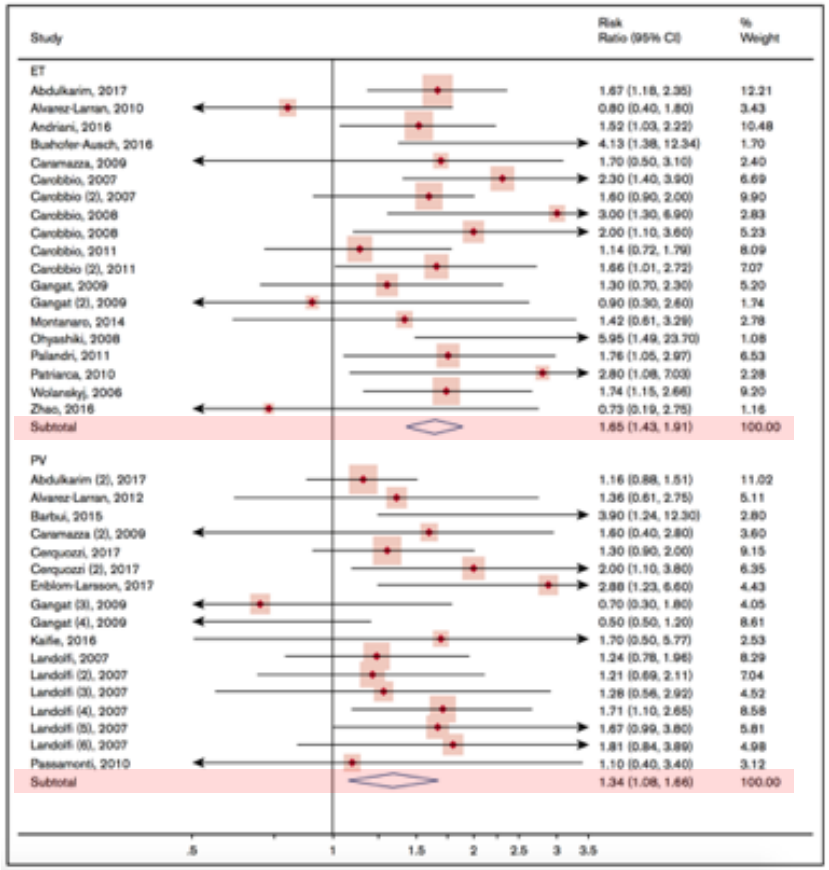
*White blood cell count greater than 8.7×10^9/L.
†Aged 60 years or older and/or previous thrombotic event.



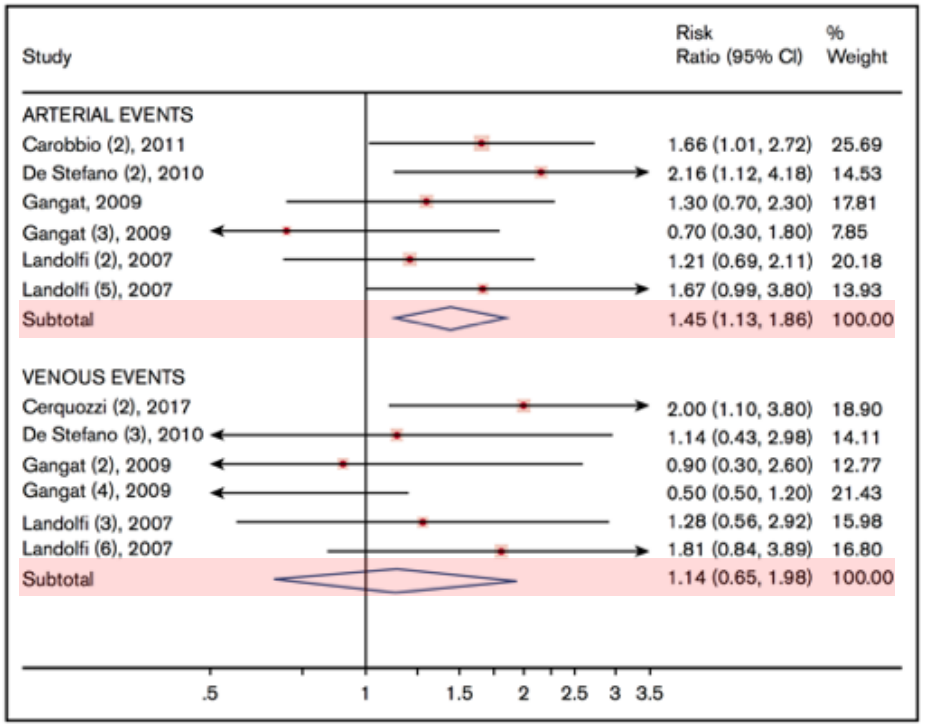
Carobbio et al. Blood 2007;109:2310-2313;
Carobbio et al. JCO 2008;26:2732-2736; Carobbio et al. Blood Adv 2019;3:1729-1737

The Effect of Leukocytosis Was Stronger in ET and in Arterial Events

Forest plot of the subgroup analysis on the primary outcome according to MPN diagnosis



Forest plot of the subgroup analysis on primary outcome according to type of thrombosis

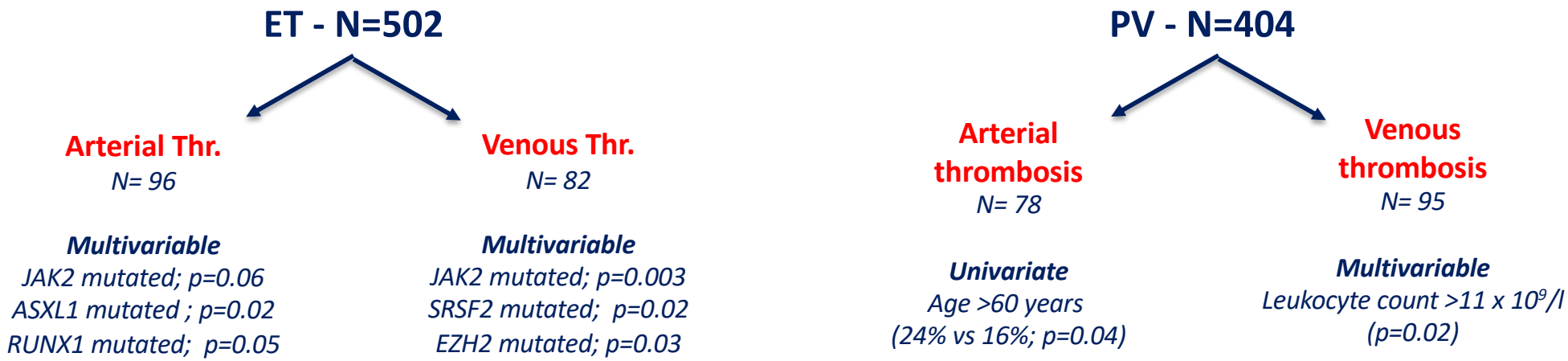


IPSET-thrombosis considered overall thrombosis without differentiating between arterial and venous events

a 60% expected increase of thrombosis risk in the presence of leukocytosis is a consistent estimate with nonnegligible clinical relevance that should be taken into account in classifying the thrombotic risk of these patients for arterial events and in ET cases.

Carobbio et al. Blood Adv 2019;3:1729-1737

Mutations and Thrombosis in ET and PV

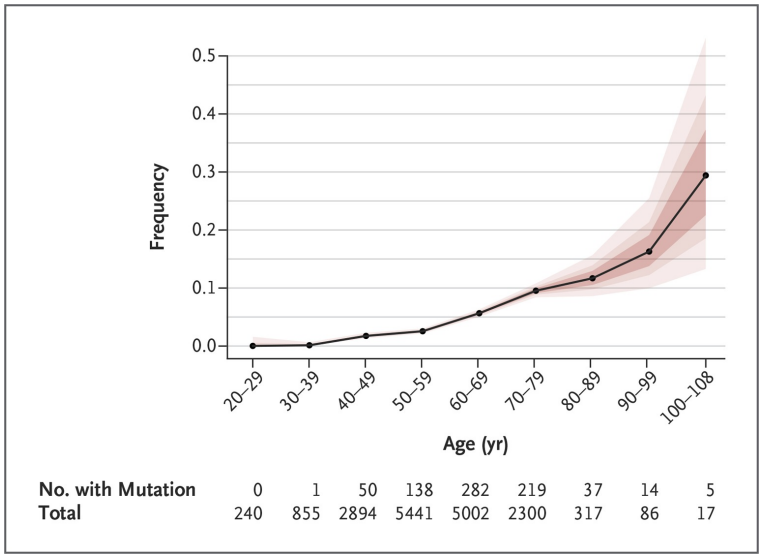


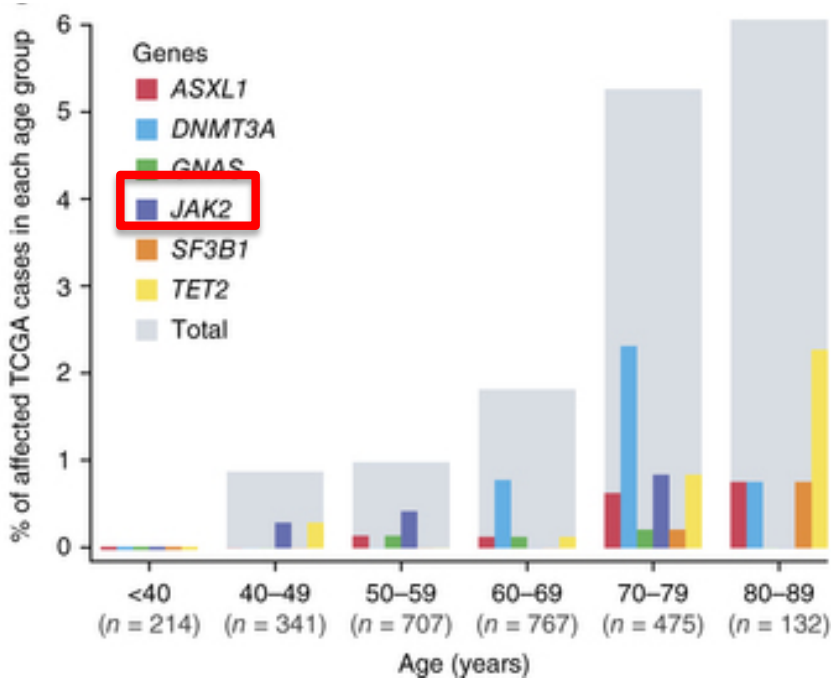
1. ***JAK2 mutated patients are at increased risk of both arterial and venous thrombosis***
2. ***High molecular risk mutations (ASXL1, RUNX1, SRSF2, EZH2) are associated with lower risk of thrombosis***
3. ***Neither age nor leukocyte count affected thrombosis risk***

1. ***Arterial thrombosis was affected by advanced age, only***
2. ***Venous thrombosis was affected by leukocytosis***

Ayalew Tefferi et al. Blood Adv 2016;1:21-30;P. Guglielmelli et al., ASH 2018

CHIP/ARCH
Clonal Hematopoiesis of Indetermined Potential/Age Related Clonal Hematopoiesis

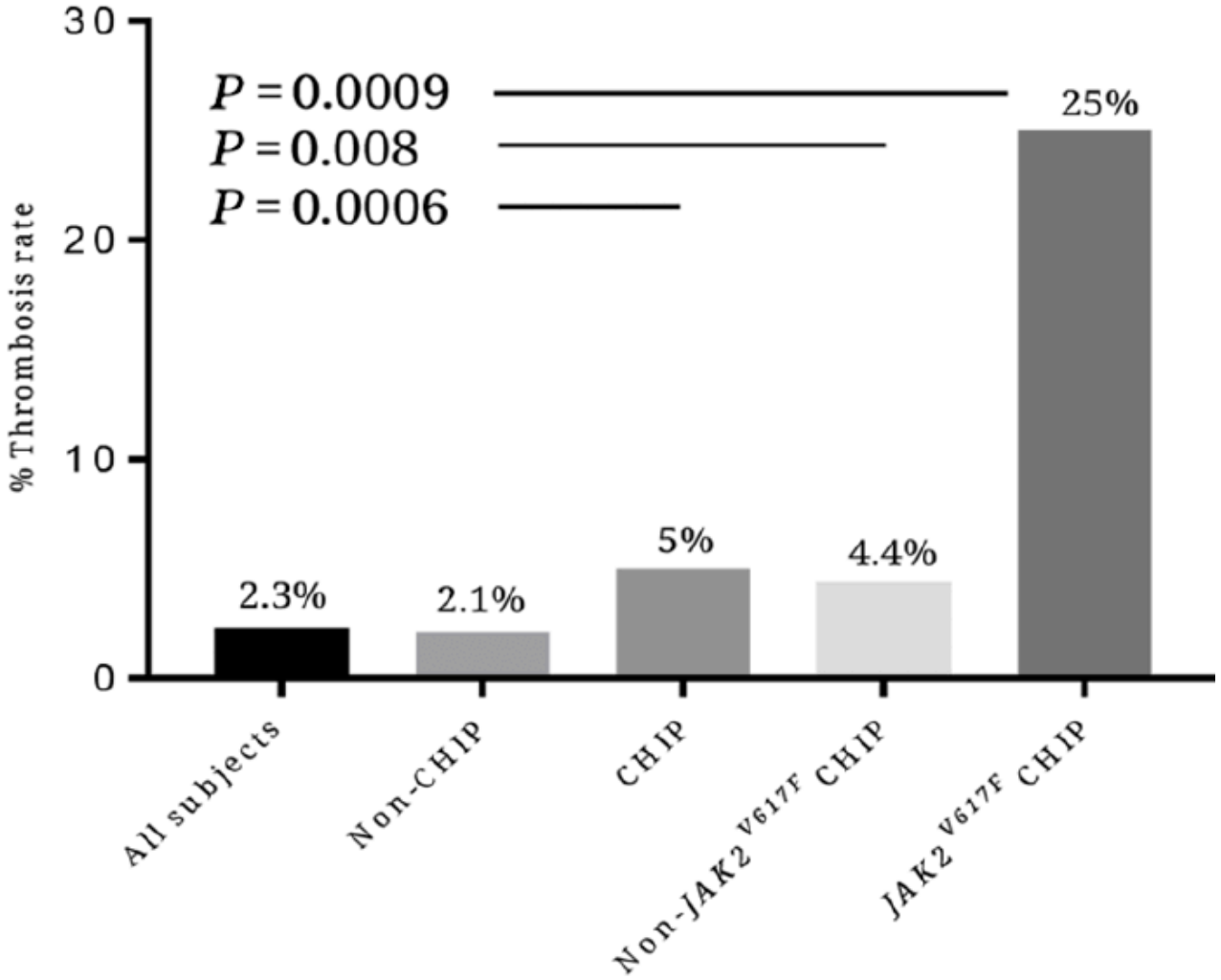




- Clonal hematopoiesis is increasingly common with aging (**10%** of persons older than 65 vs 1% of <50y).

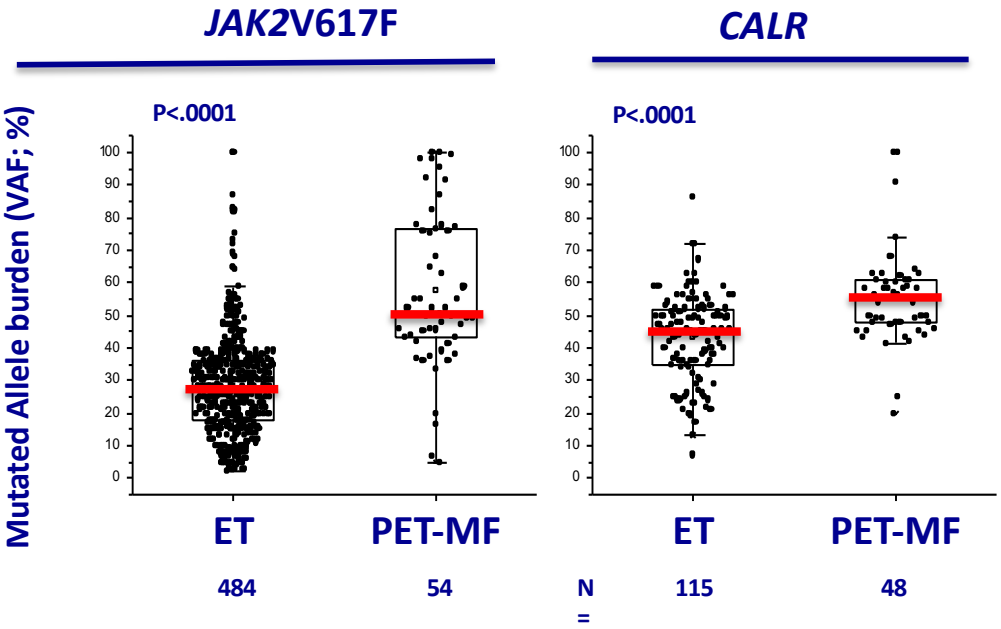
Thorsten Klampfl et al. *Blood* 2011;118:167-176; Xie M et al. *Nat Med*. 2014 ;20(12):1472-8; Jaiswal S et al. *N Engl J Med*. 2014 Dec 25;371(26):2488-98; Genovese G et al. *N Engl J Med*. 2014 25;371(26):2477-8; Jaiswal S et al, NEJM, 2017

JAK2V617F+ CHIP – venous thrombosis



Wolach et al. *Sci Transl Med*. 2018 Apr 11;10(436).

Accumulation of Mutated Alleles during Evolution to Myelofibrosis



- A *JAK2*V617F VAF >50% *vs* <50% is a risk factor for evolution: 14% *vs* 5% in ET.

Tefferi A, Cancer 2006; 106:331; Vannucchi AM, Blood 2007; 10:840;
Silver RT, Leuk Res 2010; Passamonti F, Leukemia 2010; 24:1574

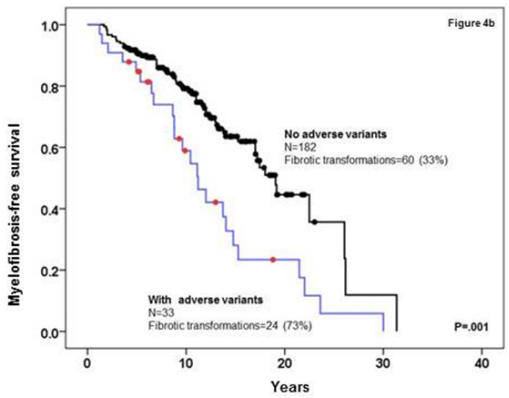
Non-Driver Mutations in Chronic Phase

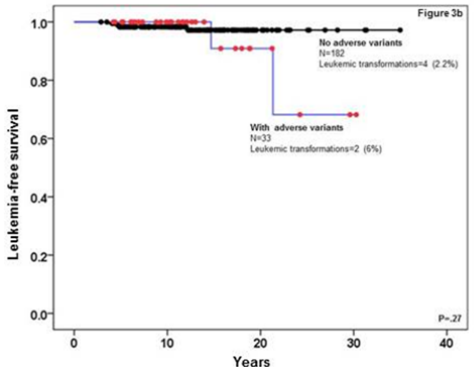
Gene (%)	PV -----→ PPV-MF		ET -----→ PET-MF	
N=	*133*	*158*	*183*	*134*
ASXL1	12%	17%	11%	29%
EZH2	0	4%	3%	10%
SRSF2	3%	1%	2%	4%
IDH1/2	2%	6%	1%	1%
TET2	22%	23%	16%	17%
LNK/SH3B3	2%	3%	0	0
ZRSR2	5%	8%	3%	0
SF3B1	3%	16%	5%	5%
SETBP1	2%	6%	2%	6%
DNMT3A	2%	0	6%	5%
CSF3R	3%	0	3%	8%
NRAS	0	0	1%	2%
CBL	1%	0	1%	16%
U2AF1	0	16%	1%	7%
RUNX1	2%	3%	2%	0
TP53	1%	0	2%	9%

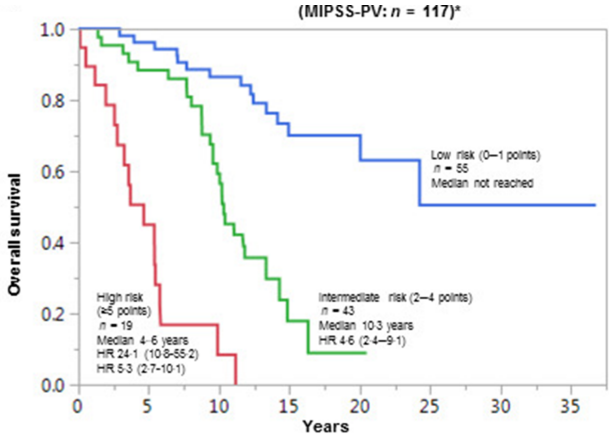
Vannucchi AM et al, Leukemia 2013; 27:1861-9. Tefferi A et al, Bood Adv 2016; 1:21-30;
Tefferi A et al, Blood Adv 2016; 1:105-111; Guglielmelli P et al, Blood; 2017:129:3227-3236

Impact of Mutations on Outcome in PV and ET

PV:
ASXL1, ***SRSF2,*** *IDH2*

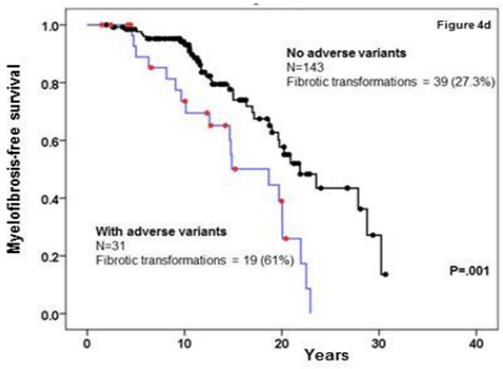


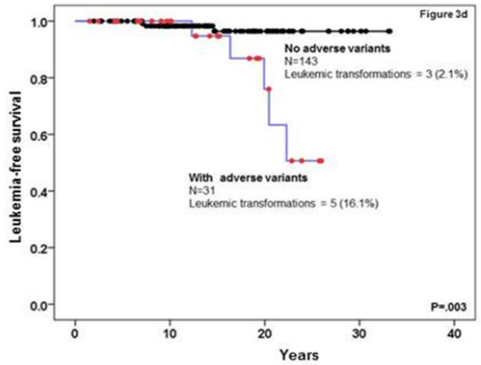


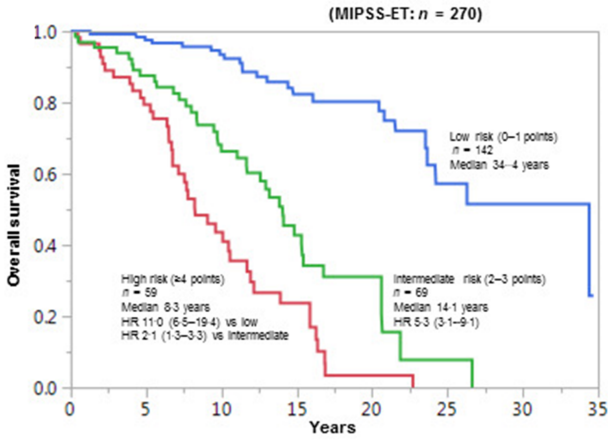


MIPSS-PV was based on four risk factors: presence of adverse mutations (*SRSF2*) (three points); age >67 years (two points); leukocyte count ≥15 × 10^9/l (one point) and thrombosis history (one point).

ET:
SH2B3, *IDH2,*
SF3B1,
U2AF1,
EZH2, ***TP53***





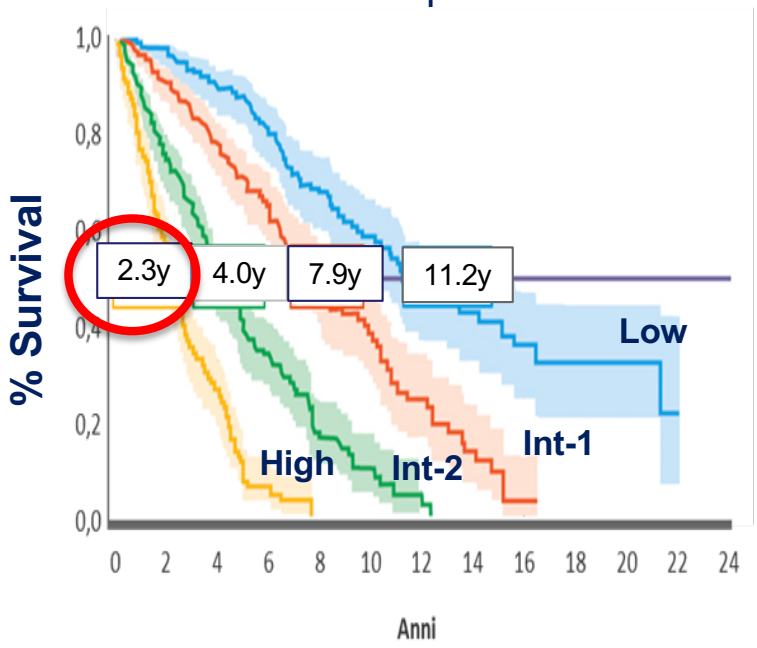


MIPSS-ET was based on three risk factors: presence of adverse mutations (SRSF2, SF3B1, U2AF1 and TP53) (two points); age >60 years (three points) and male sex (one point);

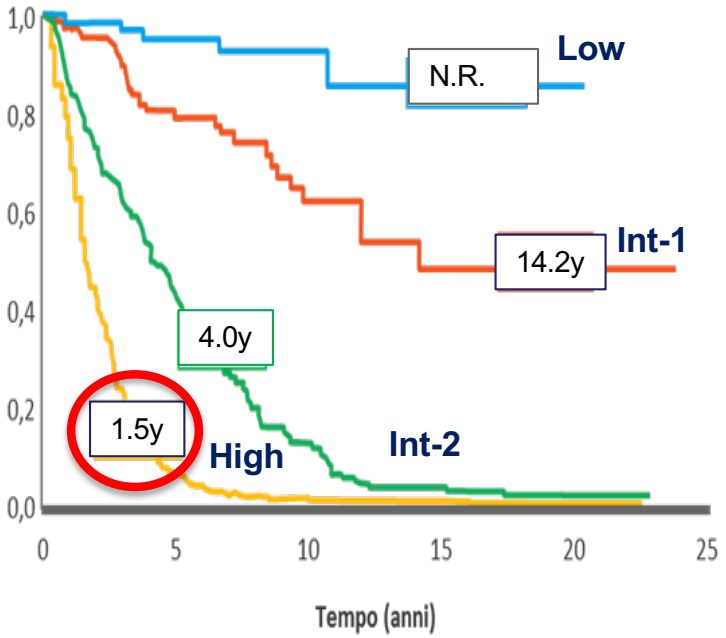
Ayalew Tefferi et al. Blood Adv 2016;1:21-30; Tefferi A et al. Br J Haematol. 2020

Survival in PMF according to IPSS and DIPSS

IPSS
(International Prognostic Scoring System)[1]



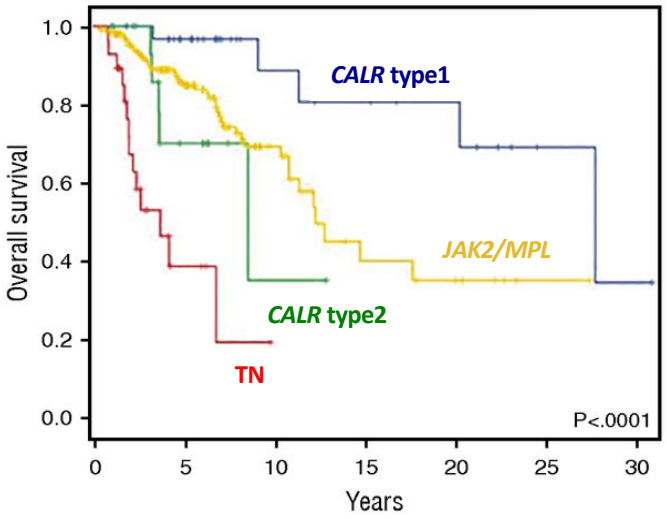
DIPSS
(Dynamic IPSS)²



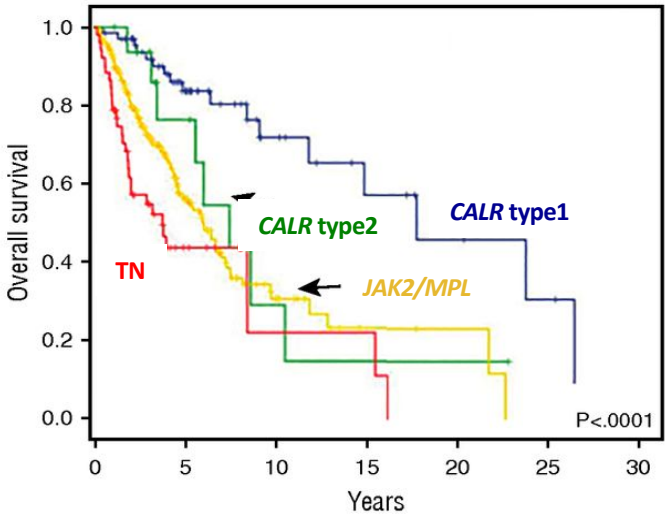
1.Cervantes F, et al. Blood 2009; 113(13): 2895-901; 2.Passamonti F et al, Blood 2010;115:1703-8

Impact of Driver Mutations on OS In Prefibrotic- and Overt-PMF

- Prefibrotic-PMF



- Overt PMF



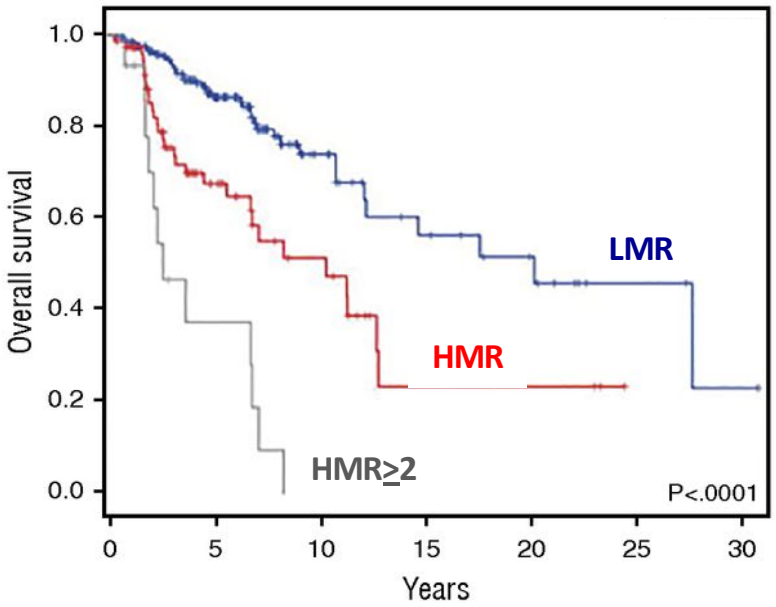
- In both pre-PMF and overt-PMF, *CALR* type 1 mutation was the most favorable, with median survival of 27.7 years and 17.8 years, respectively.
- In pre-PMF, TN had the worst outcome while an intermediate outcome was associated with *JAK2V617F* and *MPL*W515 mutations .
- In overt PMF, CALR type 1/like differed significantly from the other three mutation assets

Guglielmelli P et al. Blood 2017;129:3227-3236; Rumi E, et al. Blood. 2014 Aug 14;124(7):1062-9

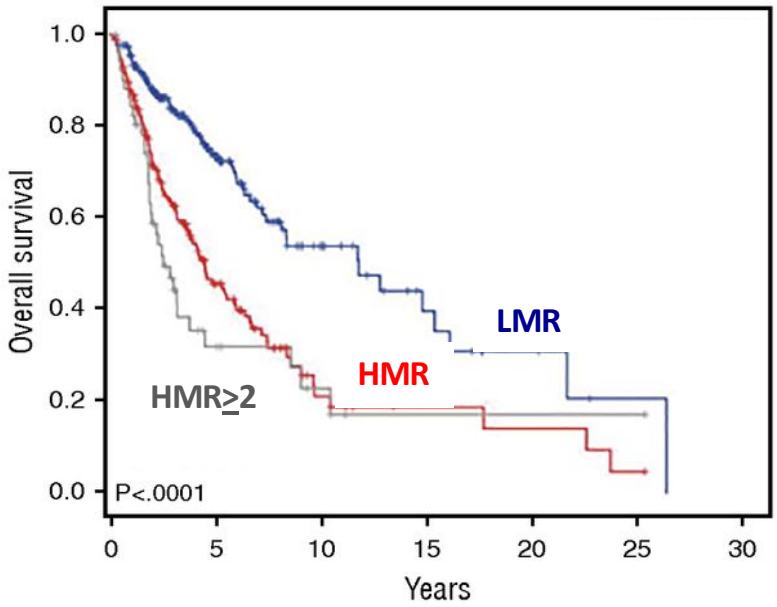
Impact of Non-Driver Mutations on OS In Prefibrotic- and Overt-PMF

harboring ≥1 mutation in any one of *ASXL1, EZH2, SRSF2, IDH1/2 (+ U2AF1)*

Prefibrotic-PMF



Overt PMF



- The HMR category was associated with significantly shorter survival in both pre-PMF and overt PMF independent of IPSS/DIPPS-plus scores

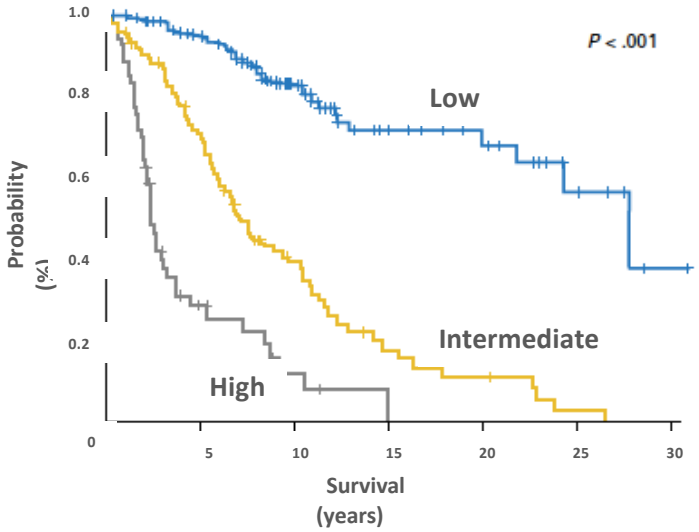
Vannucchi AM et al, Leukemia 2013;27:1861-9;
Guglielmelli P et al. Blood 2017;129:3227-3236; Guglielmelli P, et al. Leukemia 2014, 28:1804-10

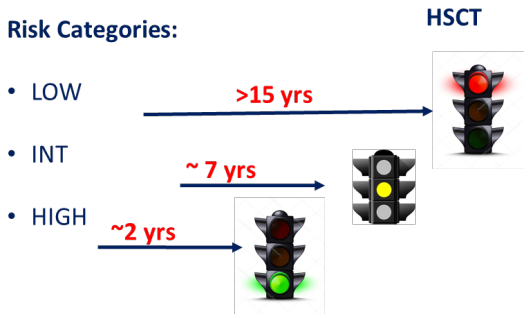
MIPSS70: Estimate of Survival for Transplant-Age Patients with PMF

Variables	Weighted value
Hb <100g/L	1
WBC >25x10^9/L	2
PLT <100x10^9/L	2
PB blasts ≥2%	1
Constitutional Symptoms	1
Grade ≥2 BM fibrosis	1
Absence *CALR* Type1	1
HMR category*	1
≥2 HMR mutations	2

Risk category	Score	OS (y)	HR
Low	0-1	27.7	1
Intermediate	2-4	7.1	5.5 (3.8-8.0)
High	≥5	2.3	16.0 (10.2-25.1)

* HMR category= any mutation in: *ASXL1*, *EZH2*, *SRSF2*, *IDH1/2*





Learning cohort: Italian cohort.

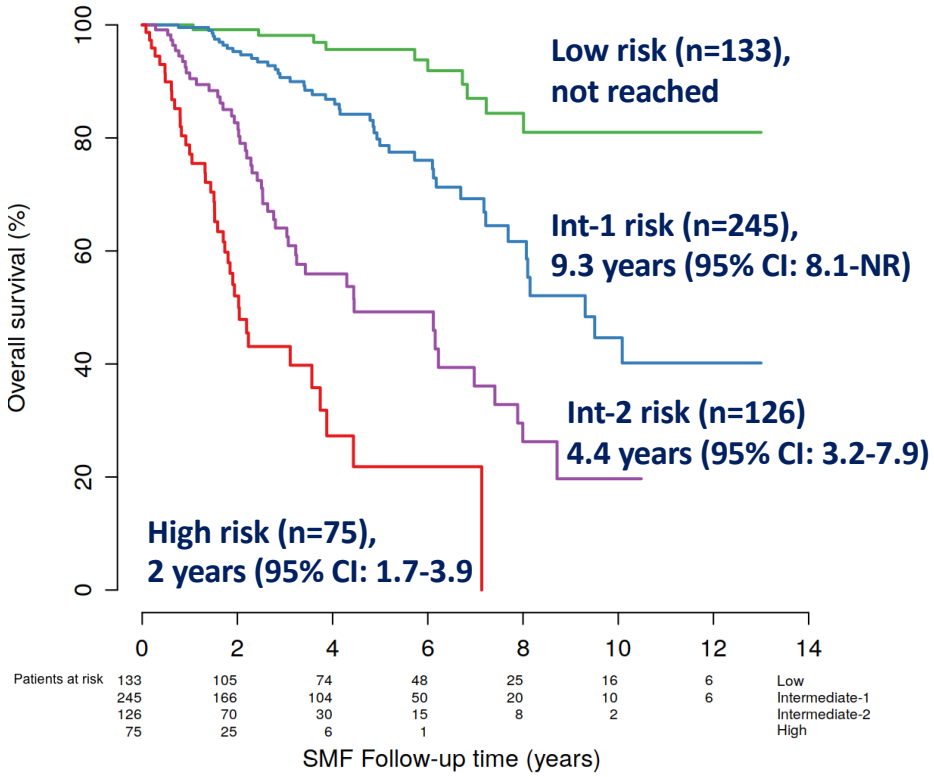
The higher categories included 30% of patients originally classified as low or intermediate IPSS risk; conversely, 6 patients (3.5%) included in intermediate-2 IPSS group were downgraded to the lowest risk categories of MIPSS70.

In **MIPSS70 plus Version 2.0** ***U2AF1*** mutations were included in HMR status and incorporates "very high risk (VHR)" Karyotype: single / multiple abnormalities of −7, i(17q), inv(3)/3q21, 12p−/12p11.2, 11q−/11q23, +21, or other autosomal trisomies, not including +8/+9;

http://www.mipss70score.it/

Guglielmelli P, et al. JCO 2018; ;36(4):310-318; Tefferi A JCO 2018 ; 36 (17): 1769

The MYSEC-PM estimate of survival in SMF



LR= <11 points
Int-1= 11-<14
Int-2= 14-<16
High= ≥16

Covariates	Points
Age, years	0.15
Hemoglobin <11 g/dL	2
Platelet < 150 x10^9/L	1
Circulating blast cells ≥ 3%	2
CALR-unmutated genotype	2
Constitutional symptoms	1

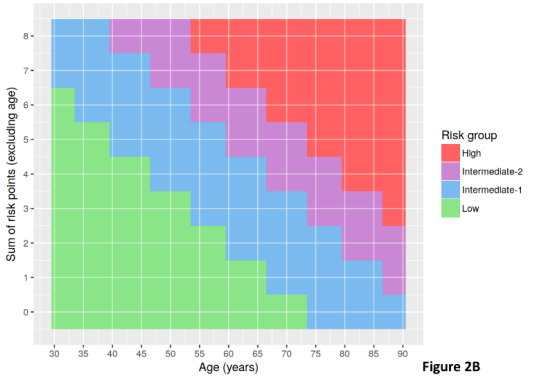
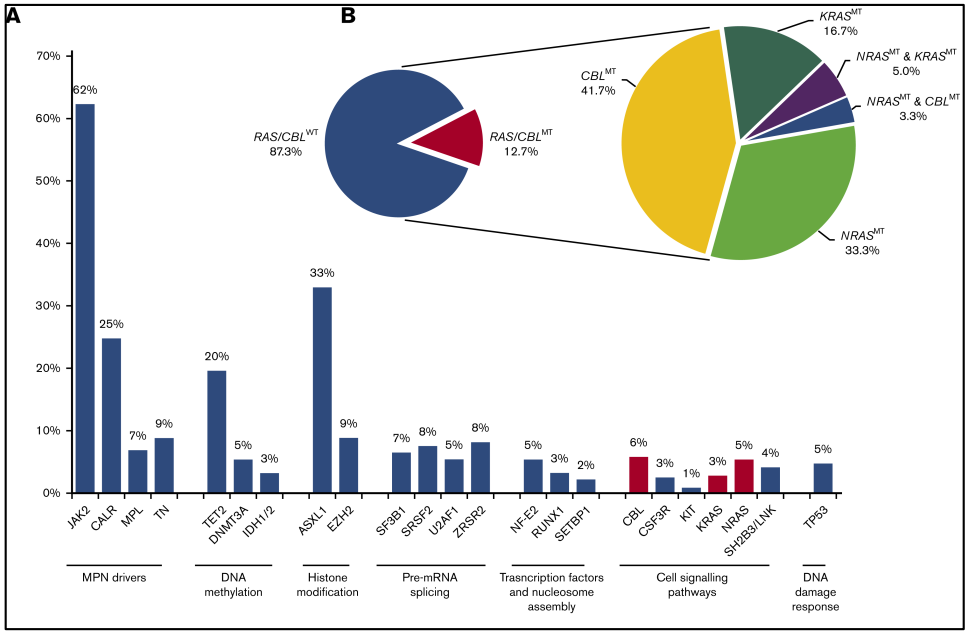


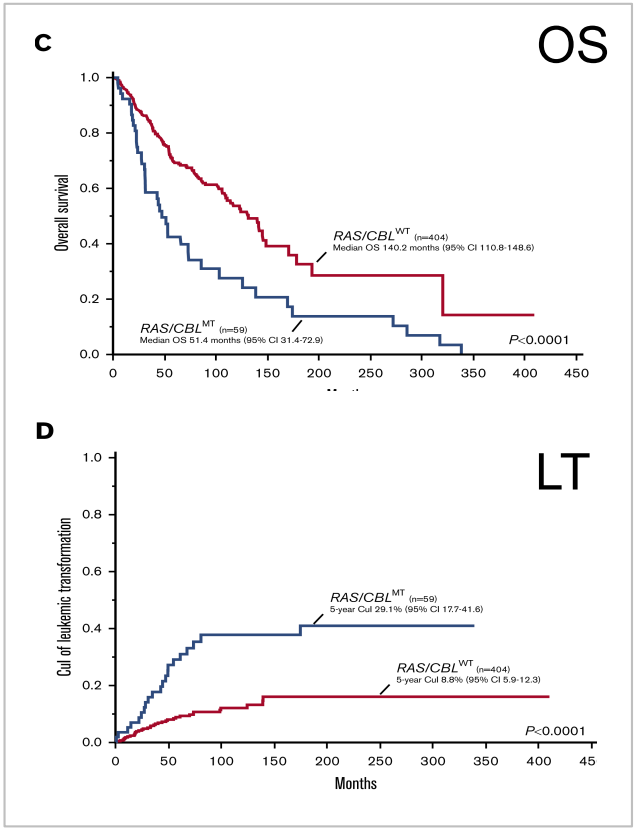
Figure 2B

http://www.mysec-pm.eu

Passamonti F et al. Leukemia. 2017 Dec;31(12):2726-2731.

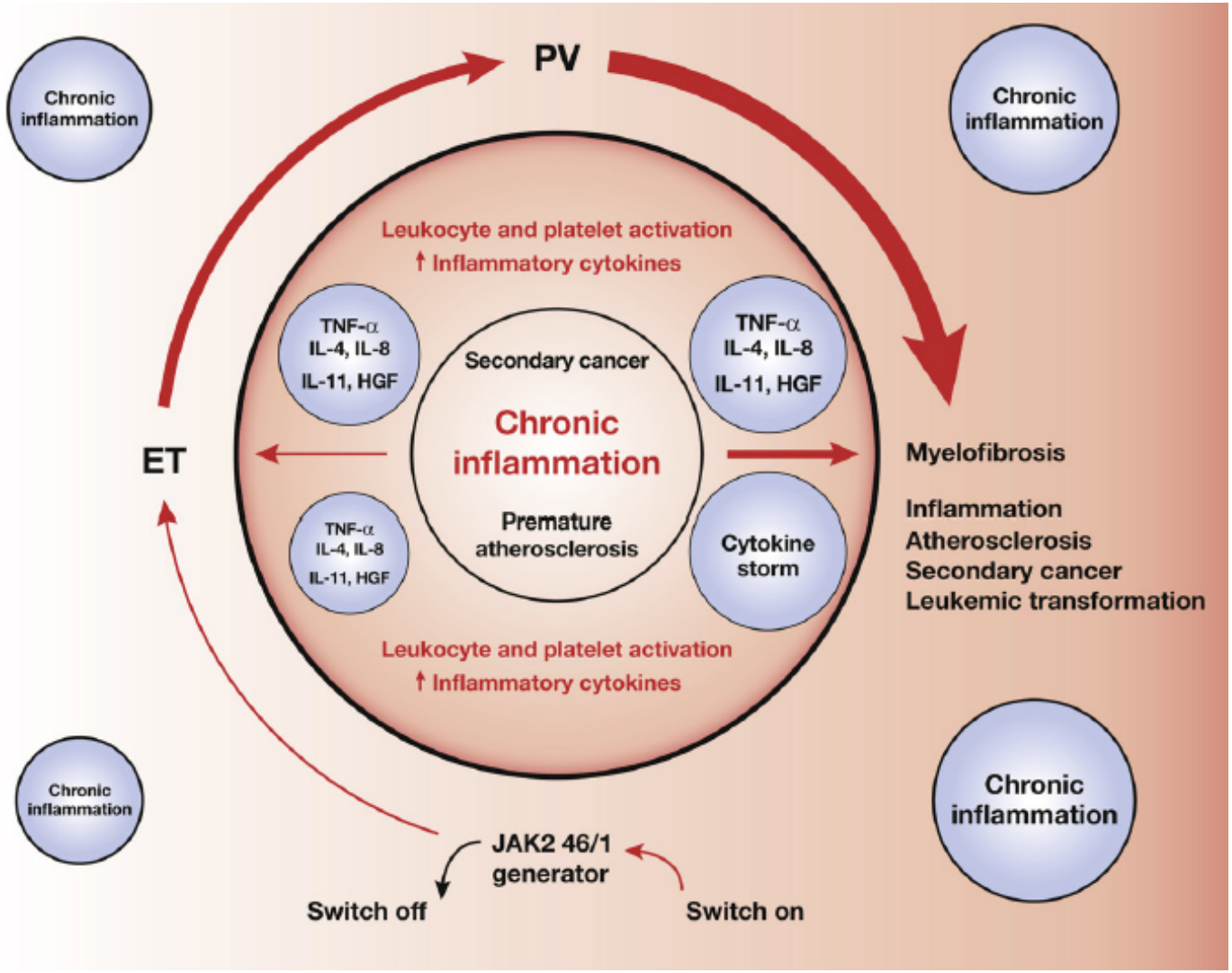
Prognostic Impact of *RAS/CBL* Mutations in MF





Coltro G et al. Blood Adv 2020; 4:3677.

MPNs Phenotype and Outcome Reflect The Underlying Biological Complexity



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AIRC Metastatic disease: the key unmet need in oncology Mynerva

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