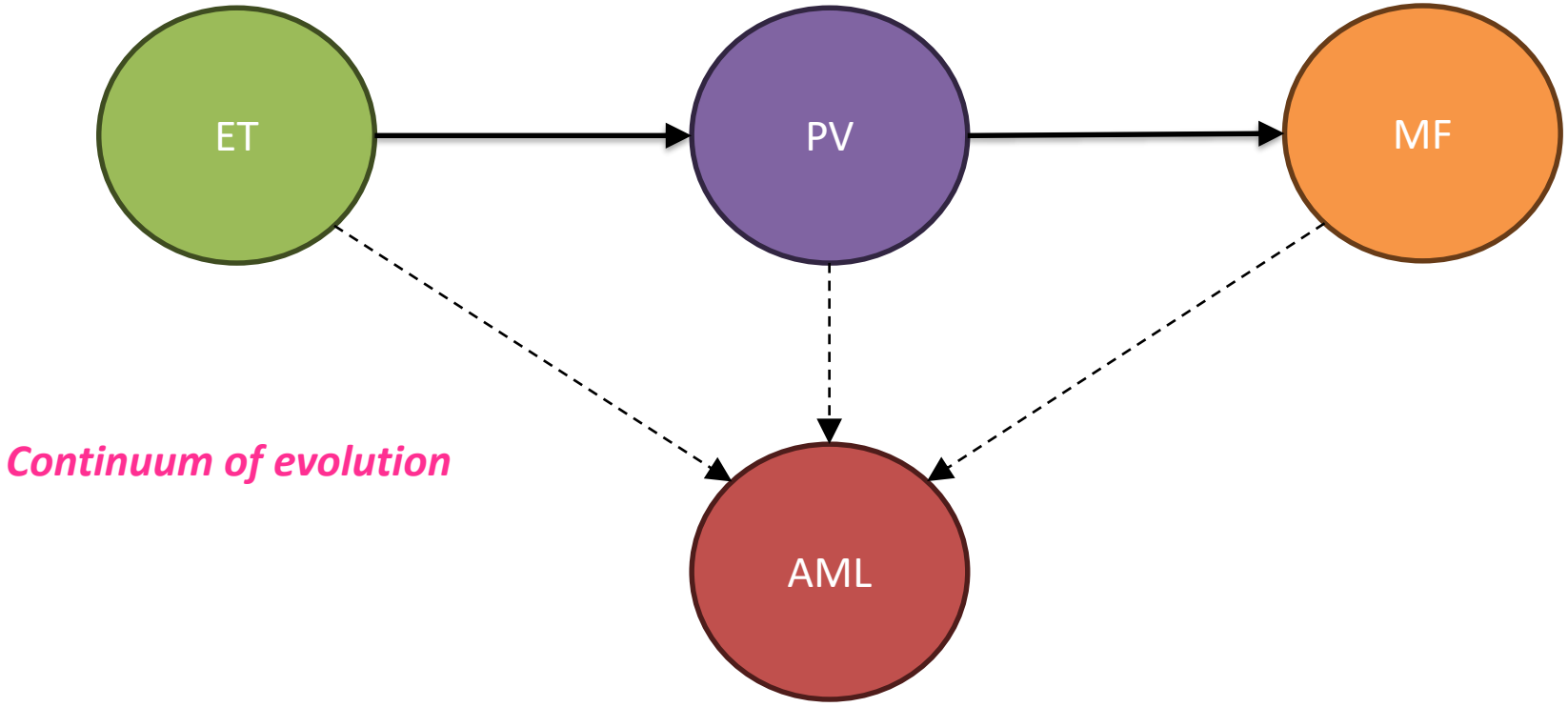
French & English SFGM-TC Day
«Role of transplantation in myelofibrosis »
07/02/2024

Genetic alterations in myelofibrosis

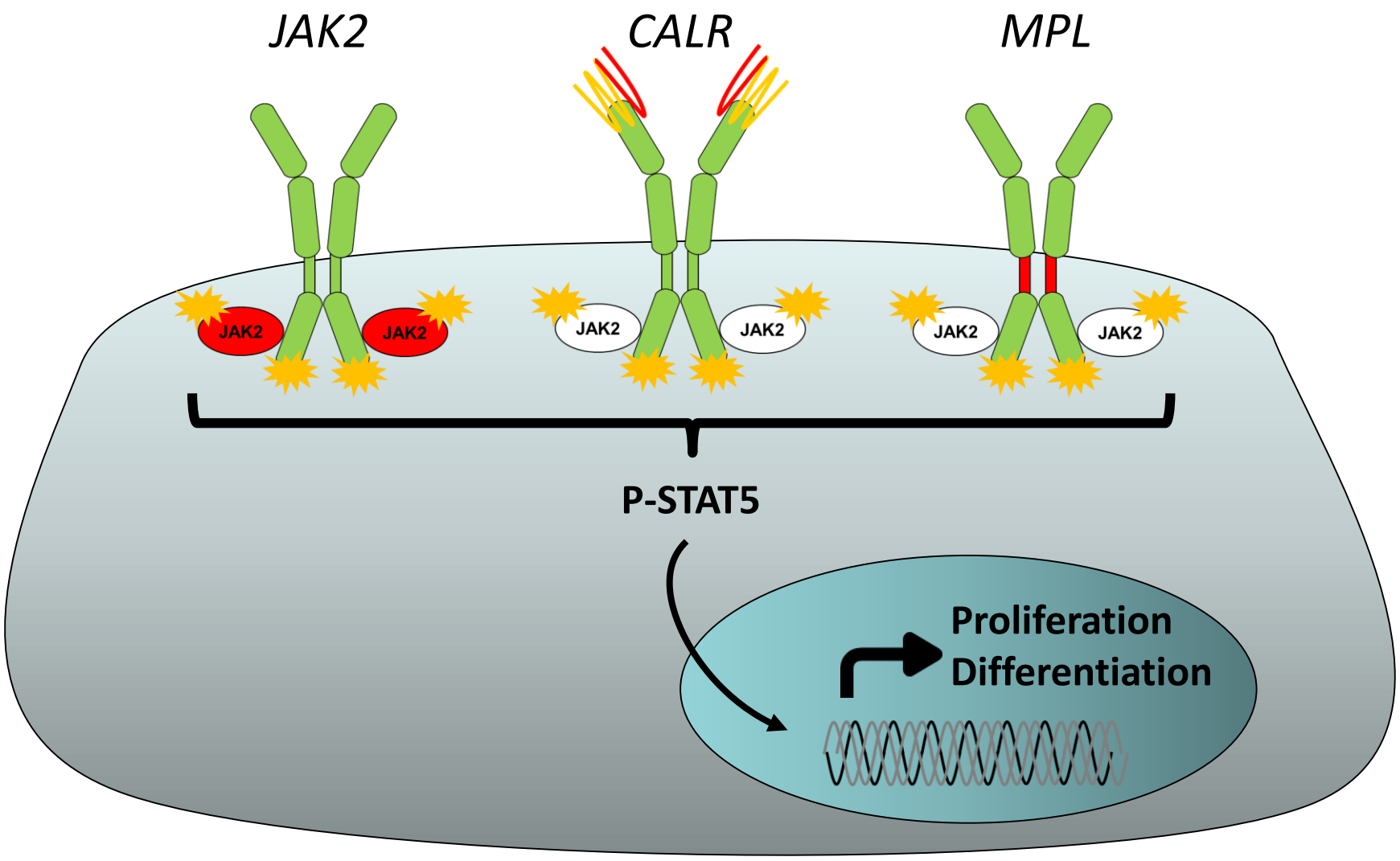
Damien Luque Paz
Laboratoire d'Hématologie – CHU d'Angers
INSERM CRCI2NA Eq 4 – Univ Angers

Myeloproliferative neoplasms (MPN)

- Chronic myeloid malignancies : essential thrombocythemia (ET), polycythemia Vera (PV) and myelofibrosis (MF)
- Clonal disease originating from one hematopoietic stem cell
- Excessive production of mature blood cells

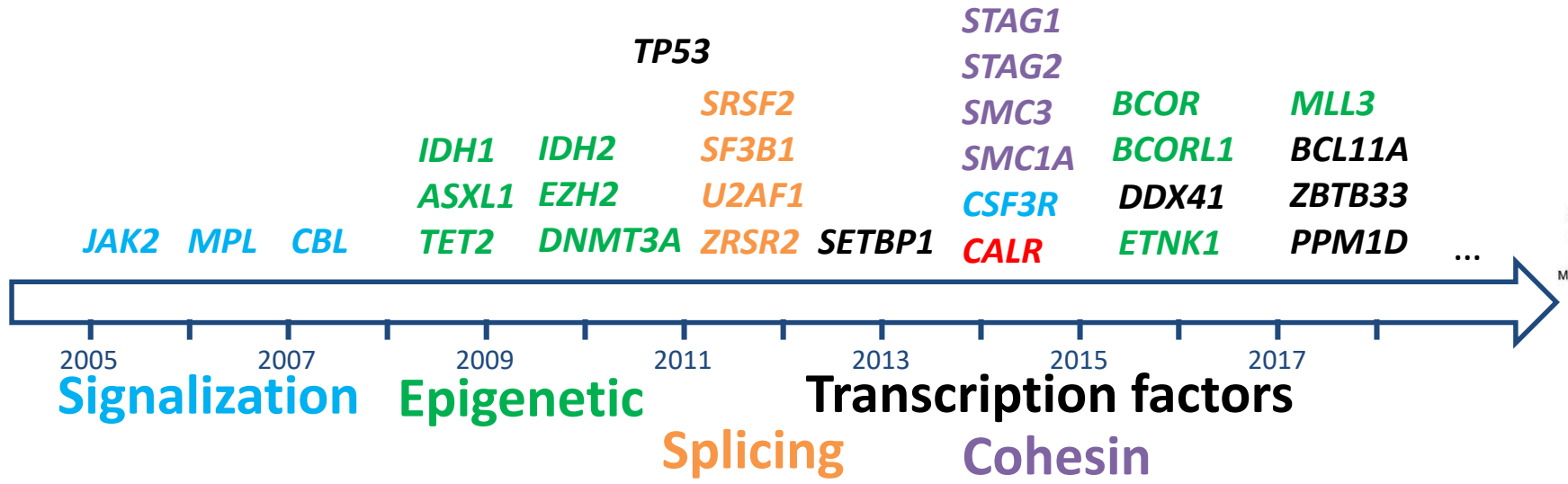


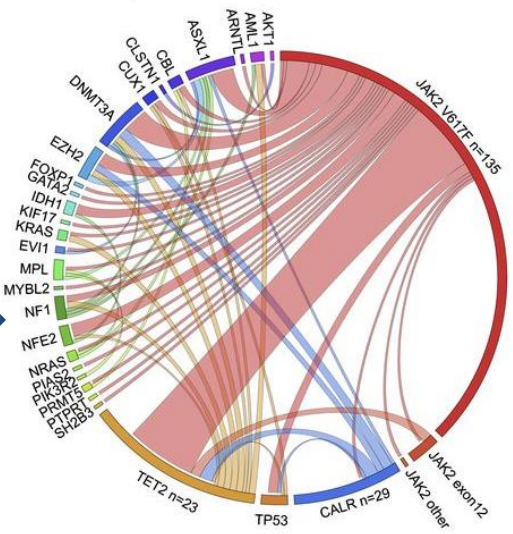
Driver mutations lead to constitutive activation of the JAK-STAT pathway

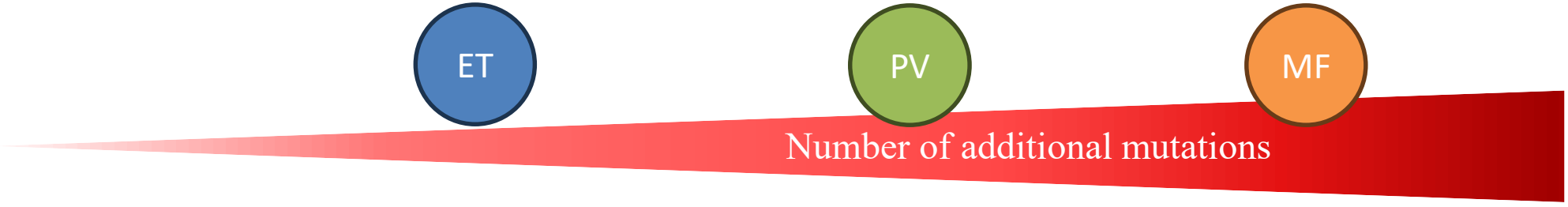


James et al. Nature 2005
Kralovics et al. NEJM 2005
Baxter et al. Lancet 2005
Levine et al. Cancer Cell 2005
Pikman et al. PLoS medecine 2006
Klampf et al. NEJM 2013
Nangalia et al. NEJM 2013

Additional mutations are detected in around 50% of MPN patients

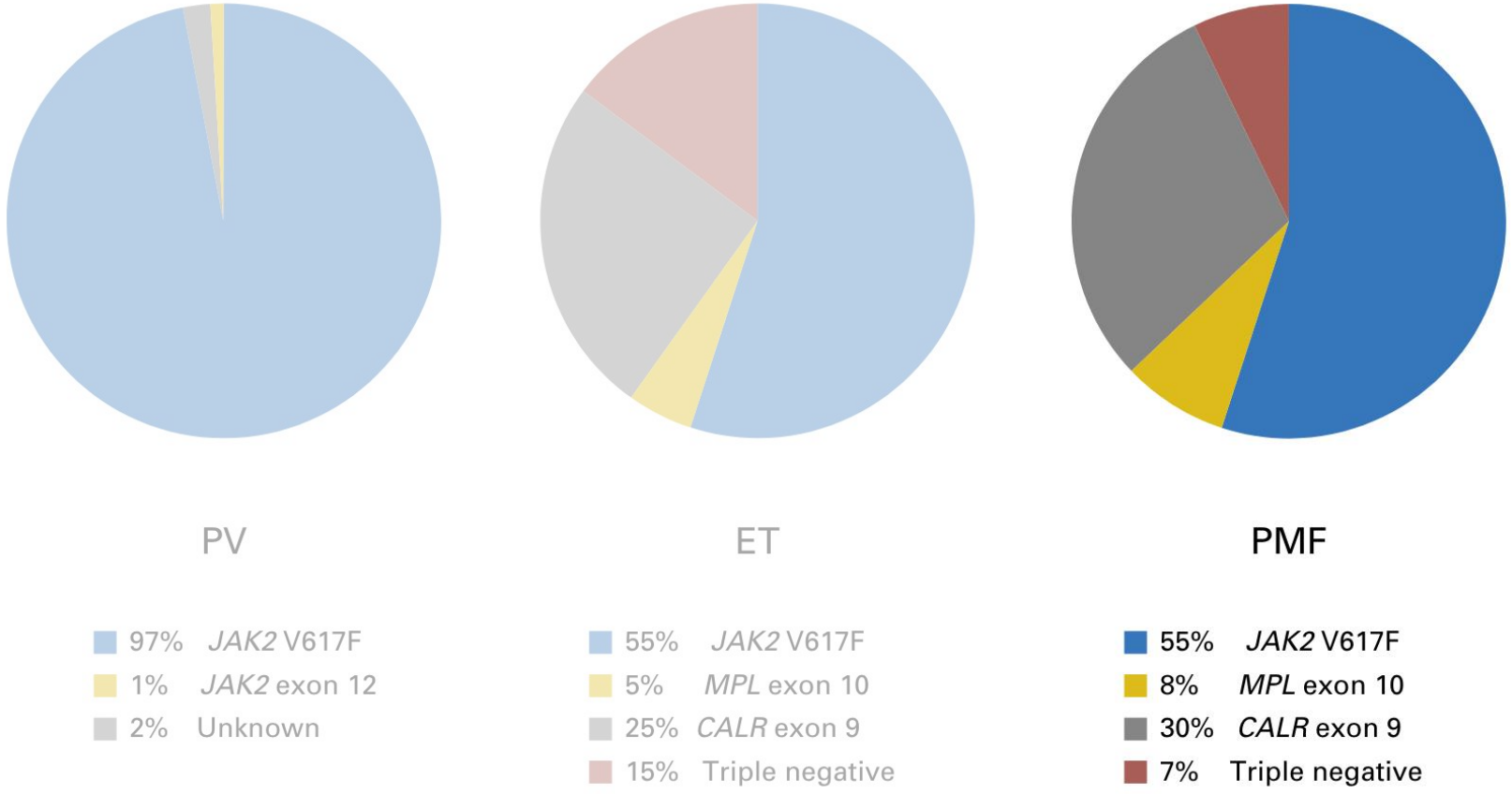






Lundberg, Blood 2014

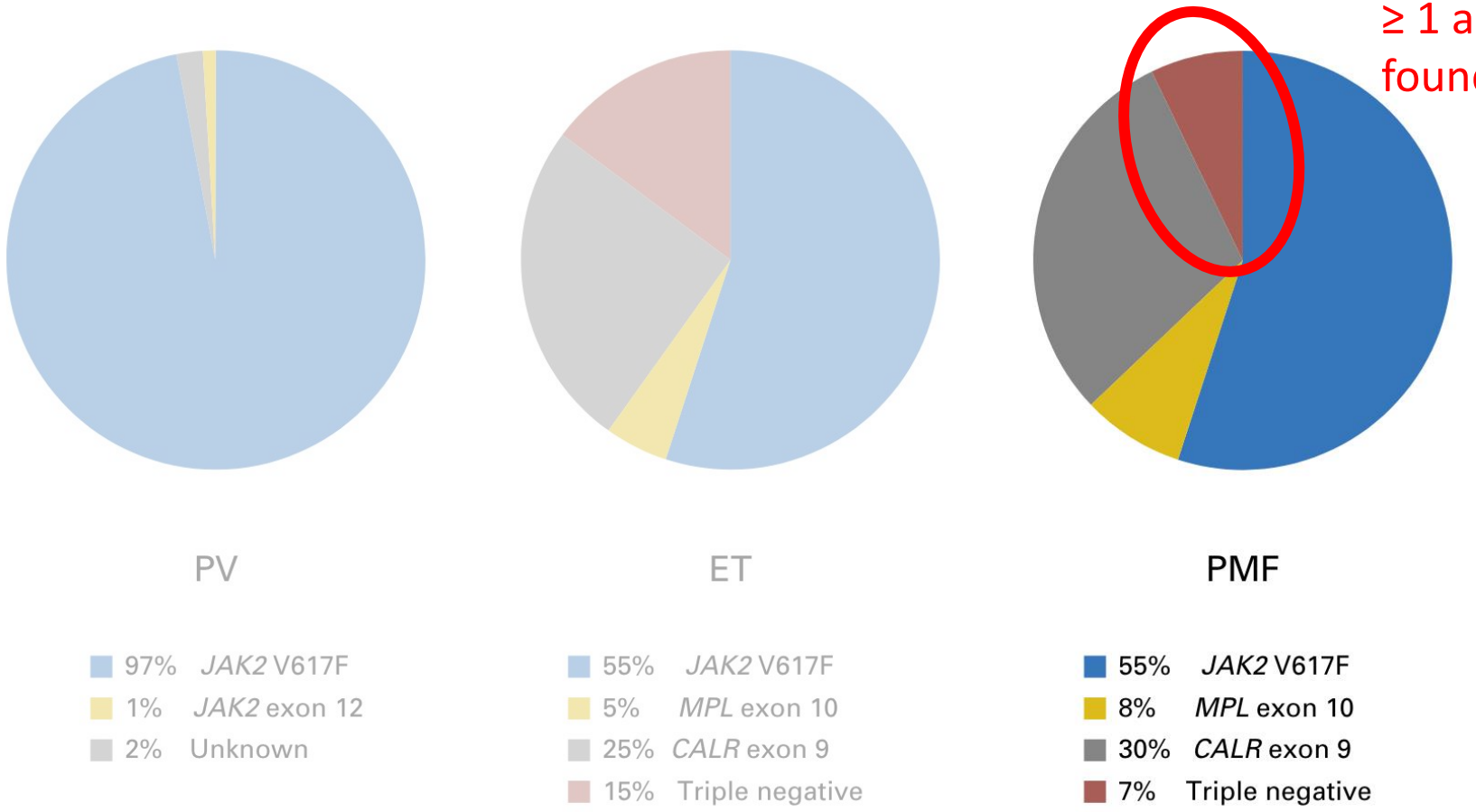
Myelofibrosis: distribution of driver mutations



Zoi & Cross, JCO 2017

Myelofibrosis: distribution of driver mutations

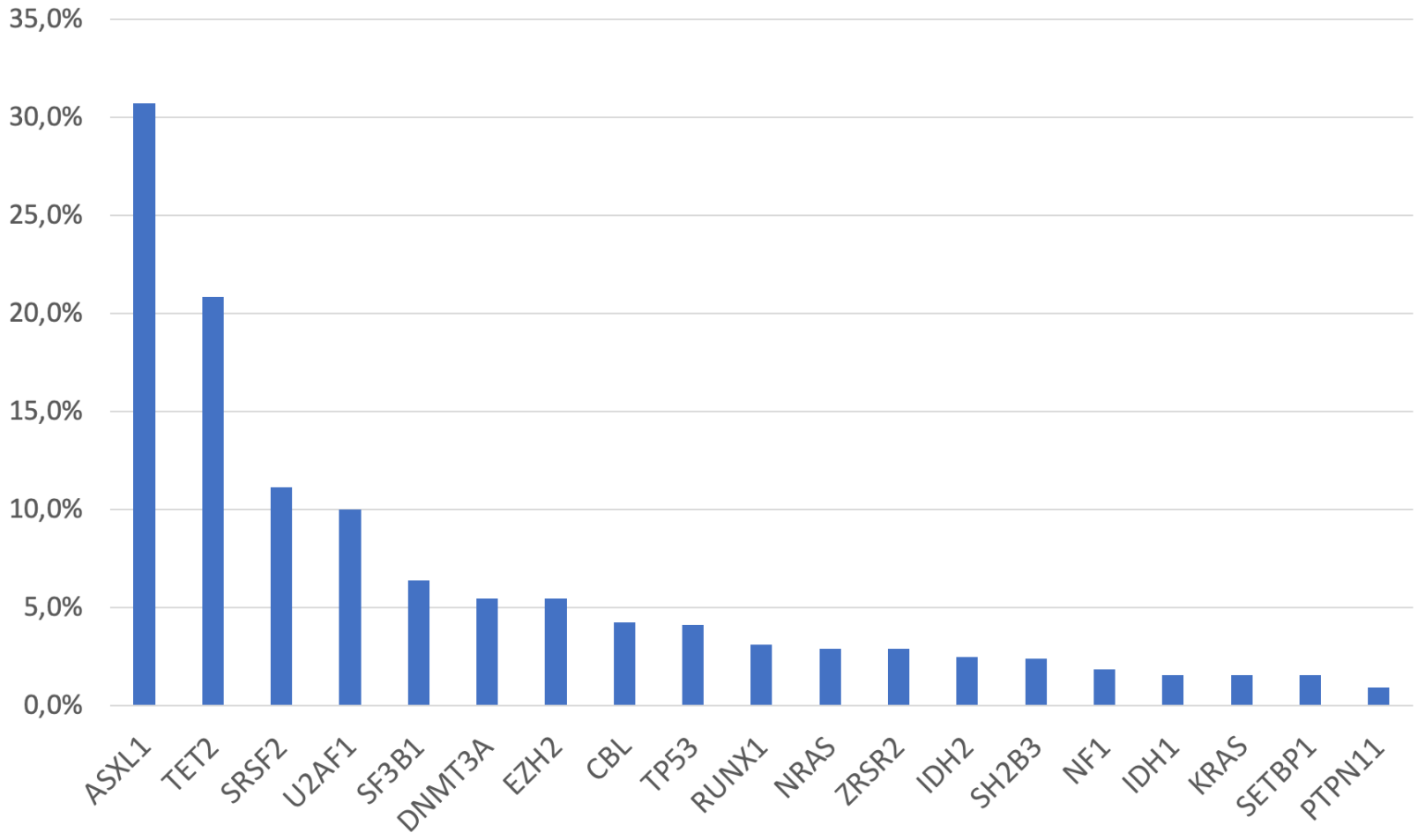
NGS in TN myelofibrosis:
≥ 1 additional mutation found in **60 à 90%** of cases



Zoi & Cross, JCO 2017

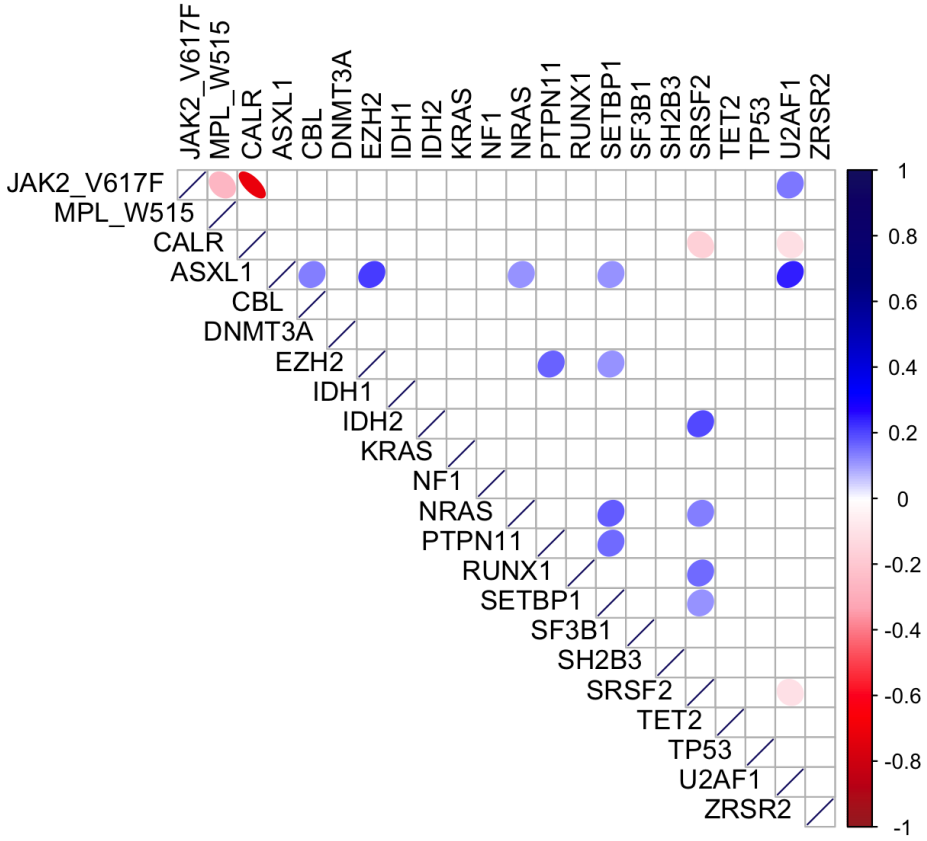
Myelofibrosis: distribution of additional mutations

Frequencies of mutations, aggregated data from 970 patients with myelofibrosis

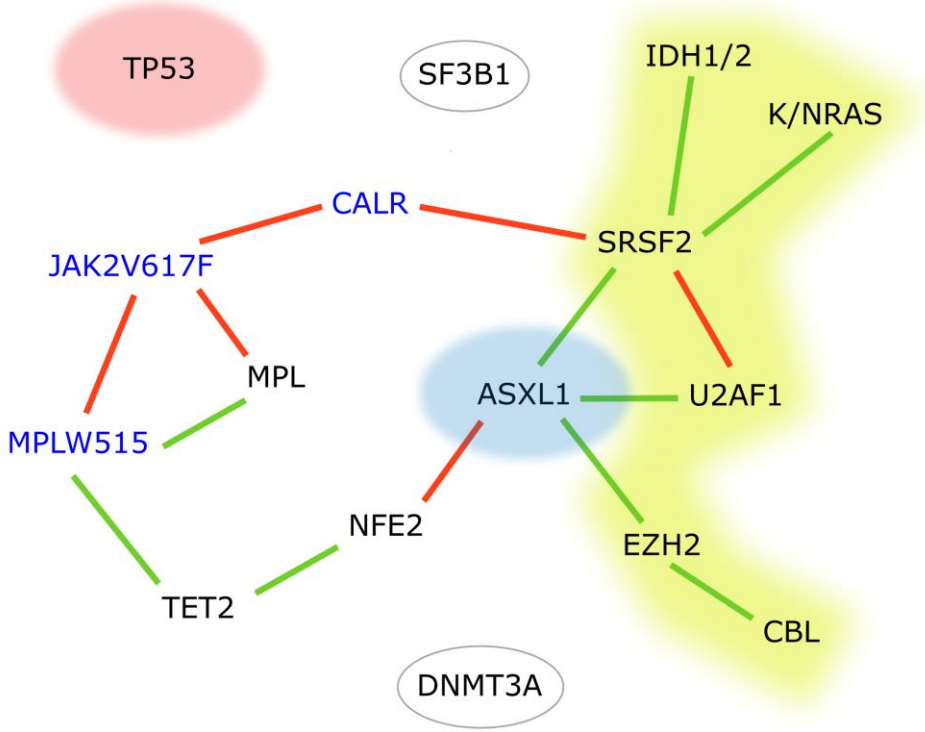


Lundberg, Blood 2014; Tefferi, Blood Advances 2016; Grinfeld, NEJM 2018; Santos, Leukemia 2020; Luque Paz Blood Advances 2021

ASXL1 mutations are the most common



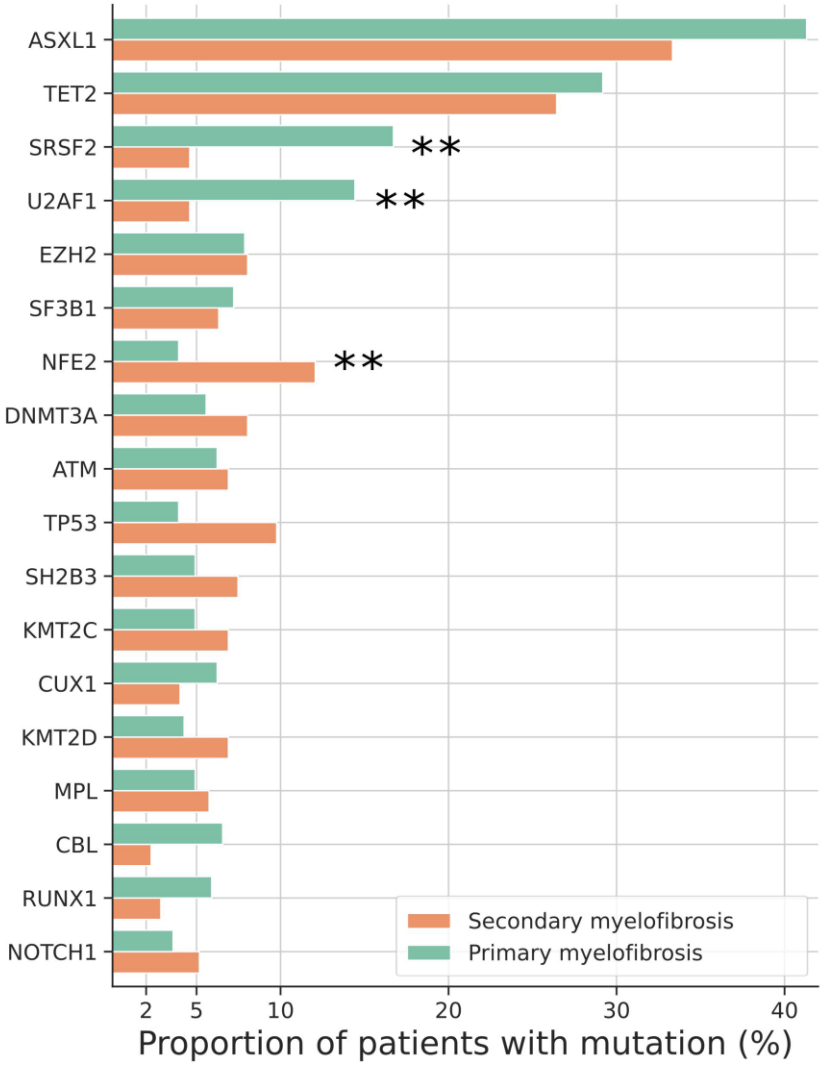
data from 970 MF patients

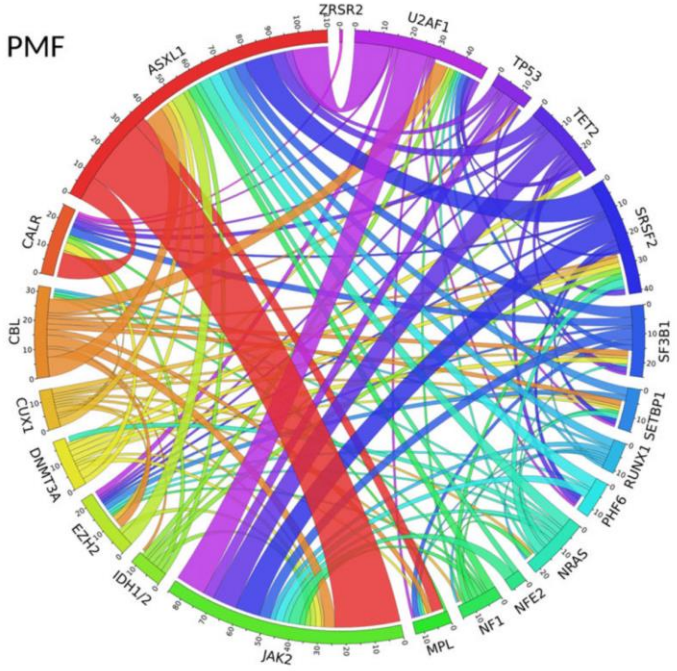


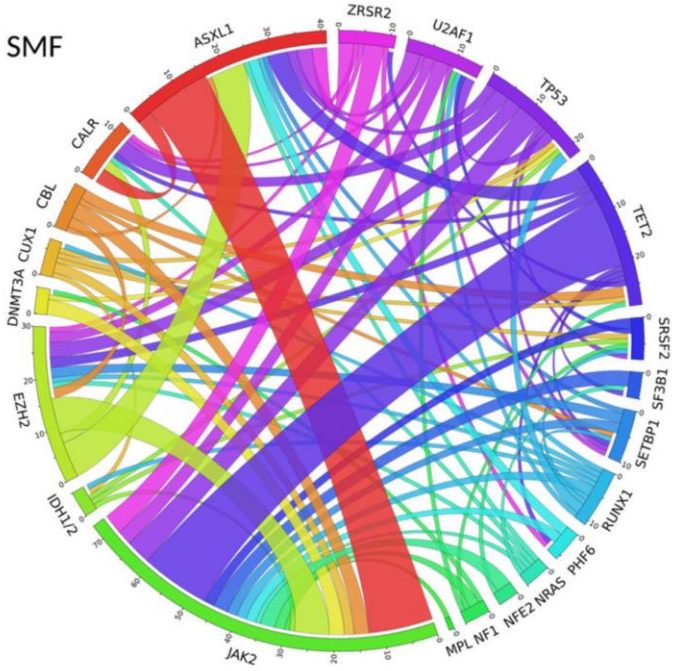
Luque Paz et al, n=479

Lundberg, Blood 2014; Tefferi, Blood Advances 2016; Grinfeld, NEJM 2018; Santos, Leukemia 2020; Luque Paz Blood Advances 2021

Differences between primary and secondary myelofibrosis







Courtier et al. GCC 2020; Luque Paz et al. Blood Advances 2021

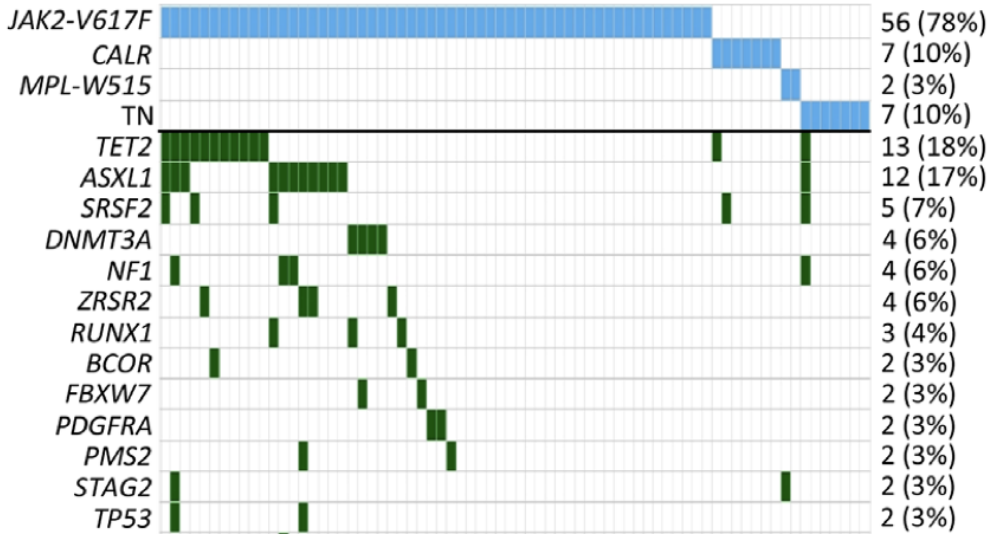
and in pre/early myelofibrosis?

Variables	Pre-PMF, N = 278	Overt PMF, N = 383	*P*
Mutated, n (%)			
ASXL1	50 (18.0)	129 (33.7)	<.0001
EZH2	10 (3.6)	46 (12.0)	<.0001
SRSF2	25 (9.0)	41 (10.7)	.28
IDH1/2	6 (2.2)	13 (3.4)	.24
HMR, n (%)			
=1	75 (27.0)	170 (44.4)	<.0001
≥2	15 (5.4)	52 (13.6)	<.0001

	MF0	MF1
ASXL1 mutated; *n* (%)	8 (14.5)	42 (18.8)
EZH2 mutated; *n* (%)	1 (1.8)	9 (4.0)
SRSF2 mutated; *n* (%)	2 (3.6)	23 (10.3)
IDH1/2 mutated; *n* (%)	0 (-)	6 (2.7)
HMR; *n* (%)	9 (16.4)	66 (29.6)
HMR ≥2; *n* (%)	2 (3.6)	13 (5.8)

Guglielmelli et al. Blood 2017

N=72 pre-PMF



Cheng et al. EJHaem 2021

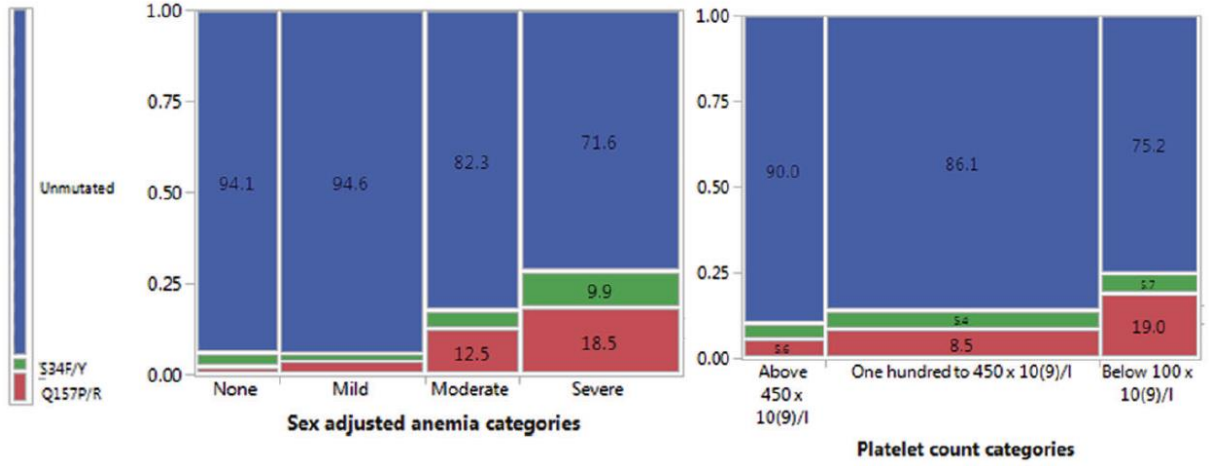
Association between mutations and clinical features

- Driver mutations
  - *CALR* mutations: younger patients, ↑ platelets
  - *JAK2* mutations: more constitutional symptoms, ↑ hemoglobin

Rumi et al. Blood 2014; Tefferi et al. Leukemia 2014

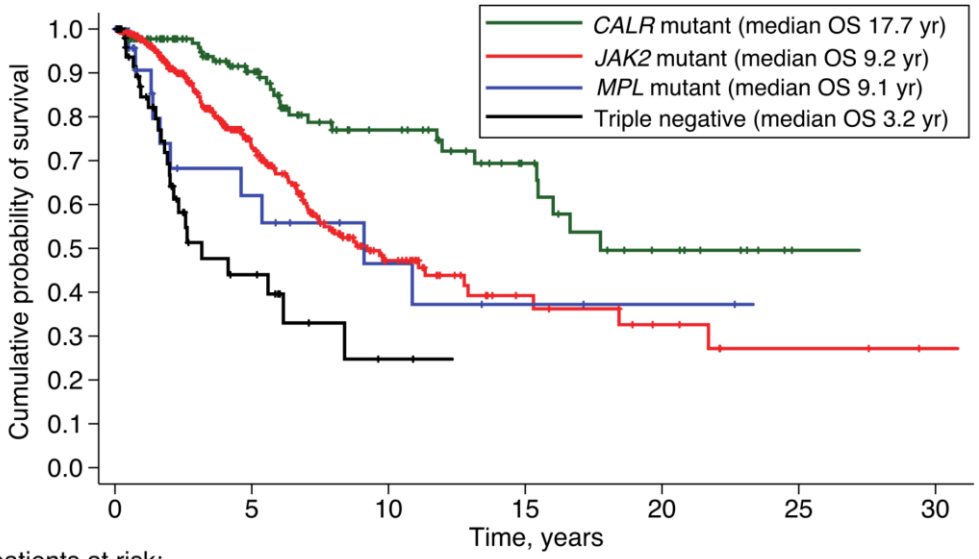
- *U2AF1* mutations <=> anemia, thrombocytopenia

Tefferi et al. Leukemia 2018



Prognostic impact of driver mutations

Overall survival



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

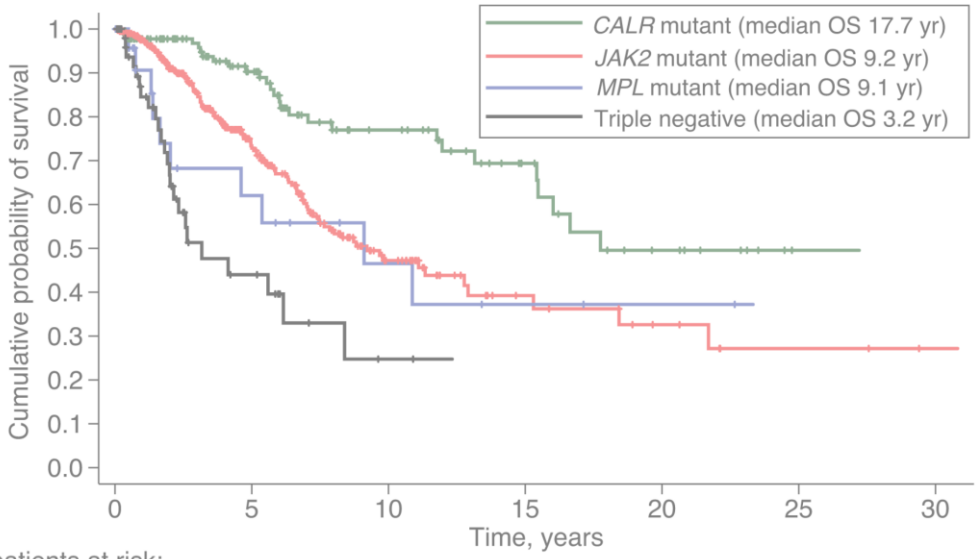
=> Independently of IPSS

For leukemic transformation => only triple-negative patients had a higher risk

Rumi et al. Blood 2014; Tefferi et al. Blood 2014

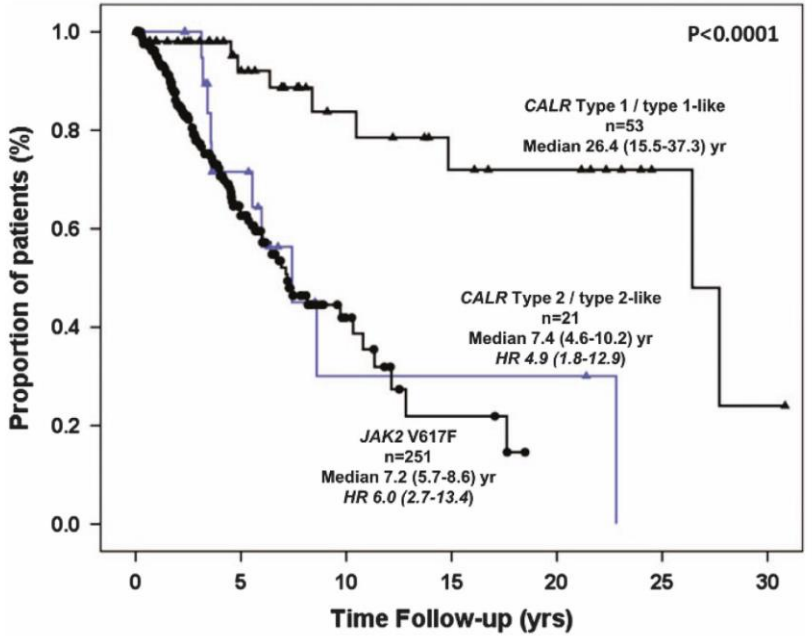
The better prognosis of *CALR* mutations is restricted to type-1 mutations

Overall survival



No. of patients at risk:

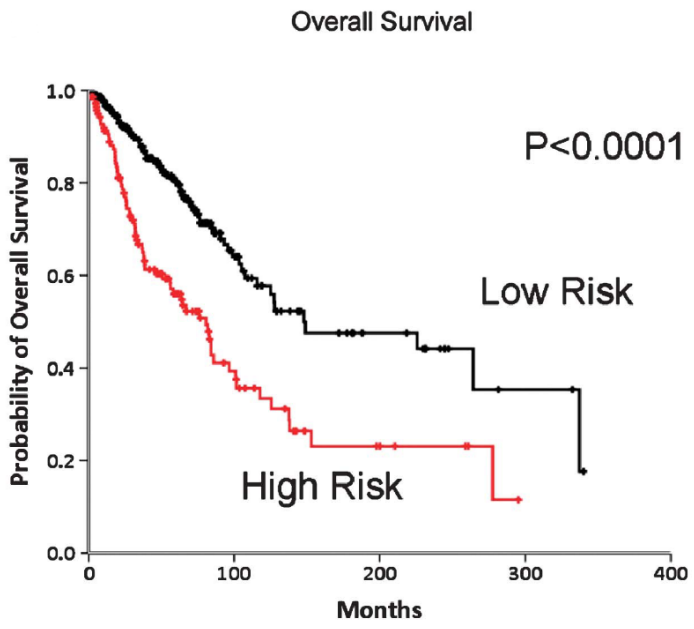
CALR mutant	140	72	37	19	9	1
JAK2 mutant	396	135	39	13	7	3
MPL mutant	25	10	5	3	2	0
Triple negative	53	11	2	0	0	0

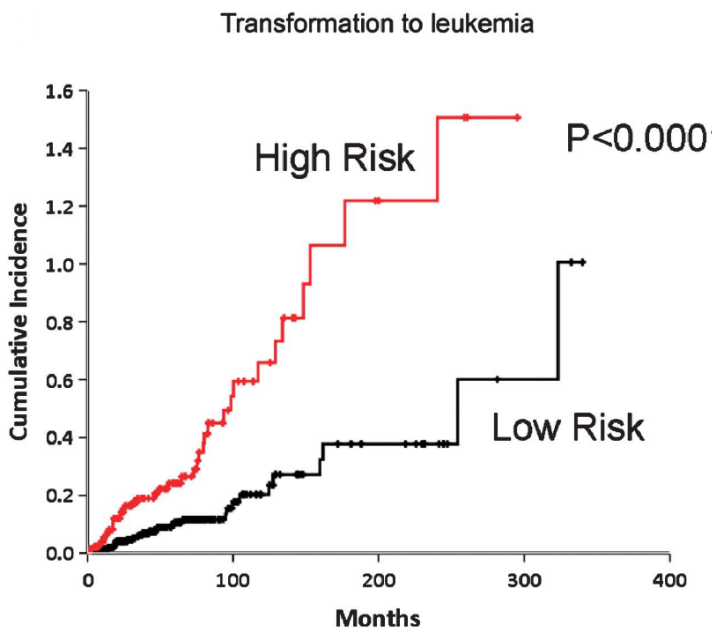


Rumi et al. Blood 2014; Tefferi et al. Blood 2014

Additional mutations: High Risk Mutations (HMR)

- HMR: mutations in *ASXL1, EZH2, IDH1/2* or *SRSF2* genes
- For PMF only
- Impact on overall survival and leukemic evolution

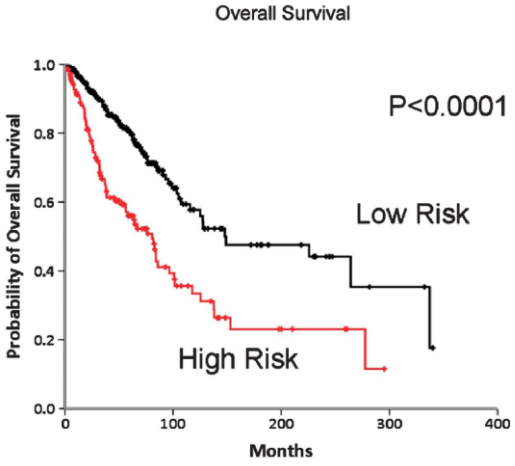




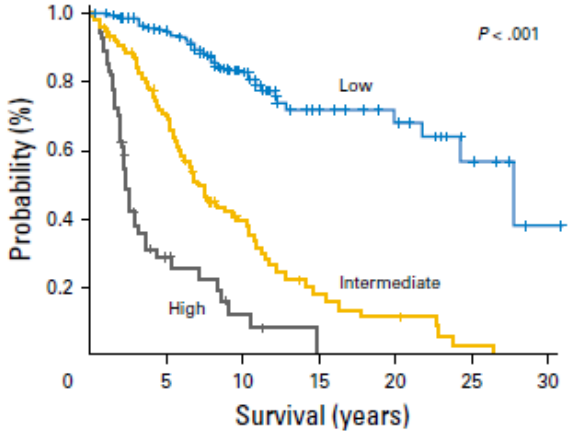
Vannucchi, Leukemia 2013

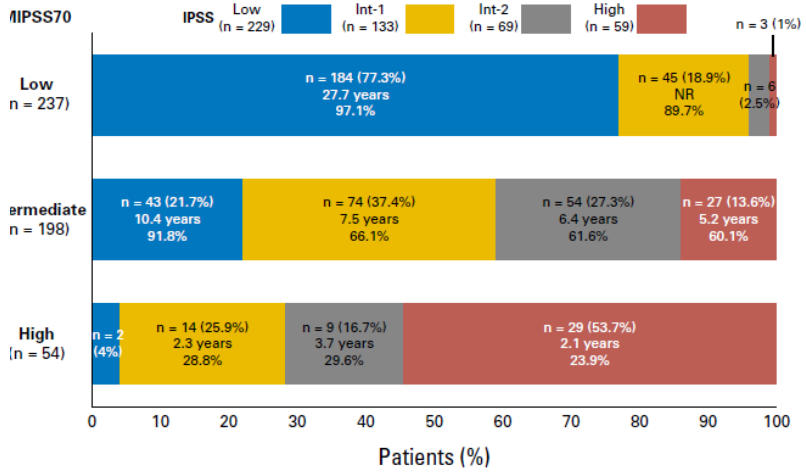
High Risk Mutations (HMR) => Integration in prognostic scores

- HMR: mutations in *ASXL1, EZH2, IDH1/2* or *SRSF2* genes
- Impact on overall survival and leukemic evolution



Vannucchi, Leukemia 2013





Guglielmelli, JCO 2018

<u>Several scores:</u>

MIPSS70

MIPSS70+

MIPSS70+v2

+U2AF1 Q157

GIPSS

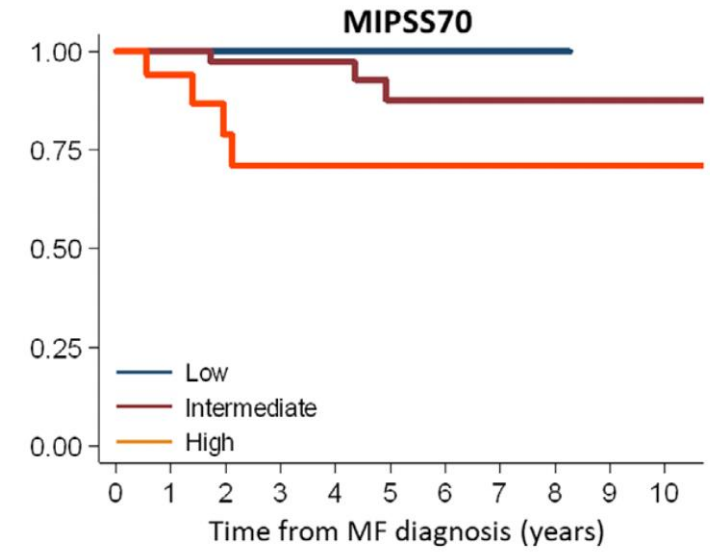
The prognostic scores with molecular data in PMF

⚠ *All scores have been developed in PMF patients at the time of diagnosis*

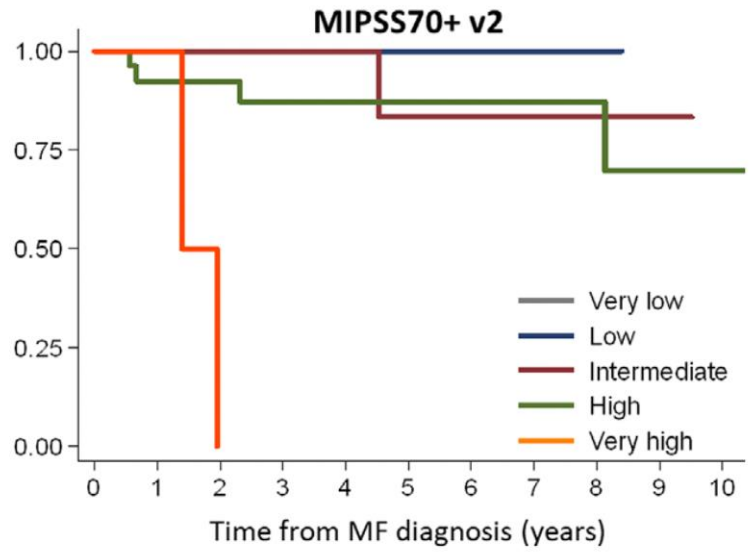
Score	Calculation		Groups, median OS	Reference
MIPSS70	Hb<10g/dL **1pt** Leuko>25G/L **2pts** Plat<100G/L **2pts** Blasts≥2%. **1pt** Symptoms **1pt**	BM fibrosis≥2 **1pt** HMR+ **1pt** ≥2 HMR **2pts** No CALR type1 **1pt**	Low (0-1pt): not reached Int (2-4pts): 6.3 years High (≥5pts): 3.1 years	*Guglielmelli, JCO 2018*
MIPSS70+v2	Severe anemia **2pts** Moderate anemia **1pt** Blasts≥2% **1pt** Symptoms **2pts** No CALR type1 **2pts**	One HMR **2pts** ≥2 HMR **3pts** Unfavorable Karyotype **4pts** VHR karyotype	Very low (0pt): NR Low (1-2pts): 16.4 years Int (3-4pts): 7.7 years High (5-8pts): 4.1 years Very high (≥9pts): 1.8 years	*Tefferi, JCO 2018*
GIPSS	No CALR type1 **1pt** Unfavorable **1pt** Karyotype **2pts** VHR karyotype	*ASXL1*-mut **1pt** *SRSF2*-mut **1pt** *U2AF1*-mut **1pt**	Low (0pt): 26.4 years Int-1 (1pt): 8 years Int-2 (2pts) : 4.2 years High (≥3pts): 2 years	*Tefferi, Leukemia 2018*

HMR: *ASXL1, EZH2, IDH1, IDH2, SRSF2* and *U2AF1* Q157 for MIPSS70+v2

MIPSS70 seemed to be applicable in secondary myelofibrosis



Number at risk						
Low	2	2	1	1	1	0
Interm	44	35	23	13	5	1
High	17	10	5	2	2	2

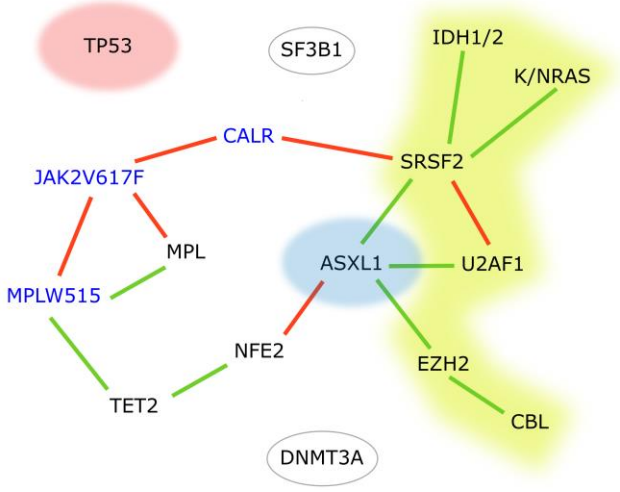


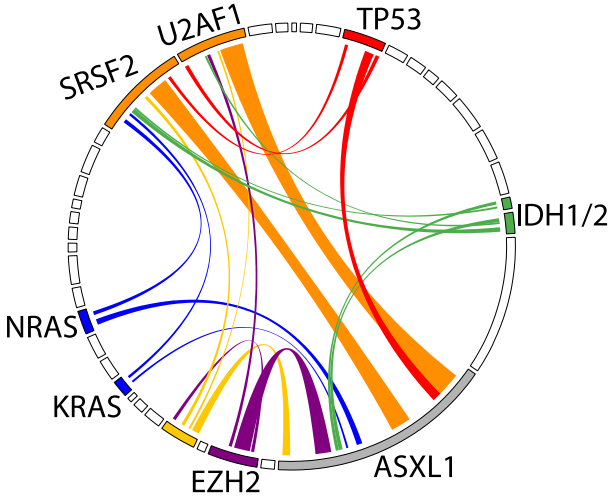
Number at risk						
Very low	1	1	1	1	1	0
Low	11	11	8	6	1	0
Interm	15	11	7	3	2	0
High	28	21	13	7	5	1
Very high	3	0	0	0	0	0

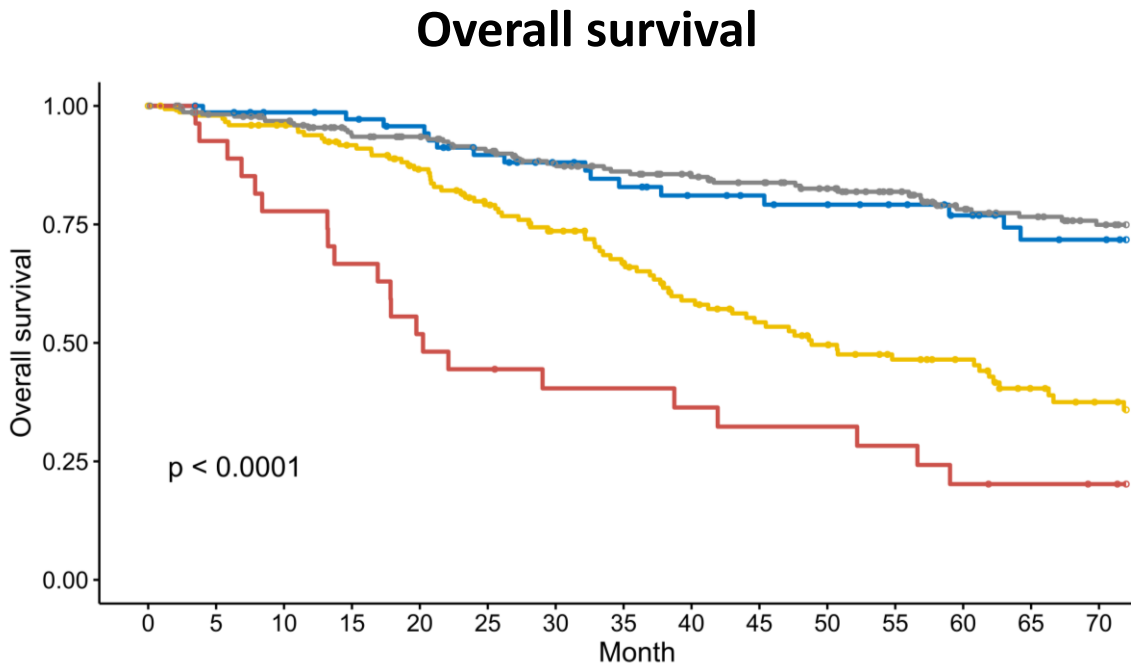
Guerra et al. AJH 2024

Redefinition of HMR? The impact of *ASXL1* mutations

- French (FIM) cohort of 479 myelofibrosis



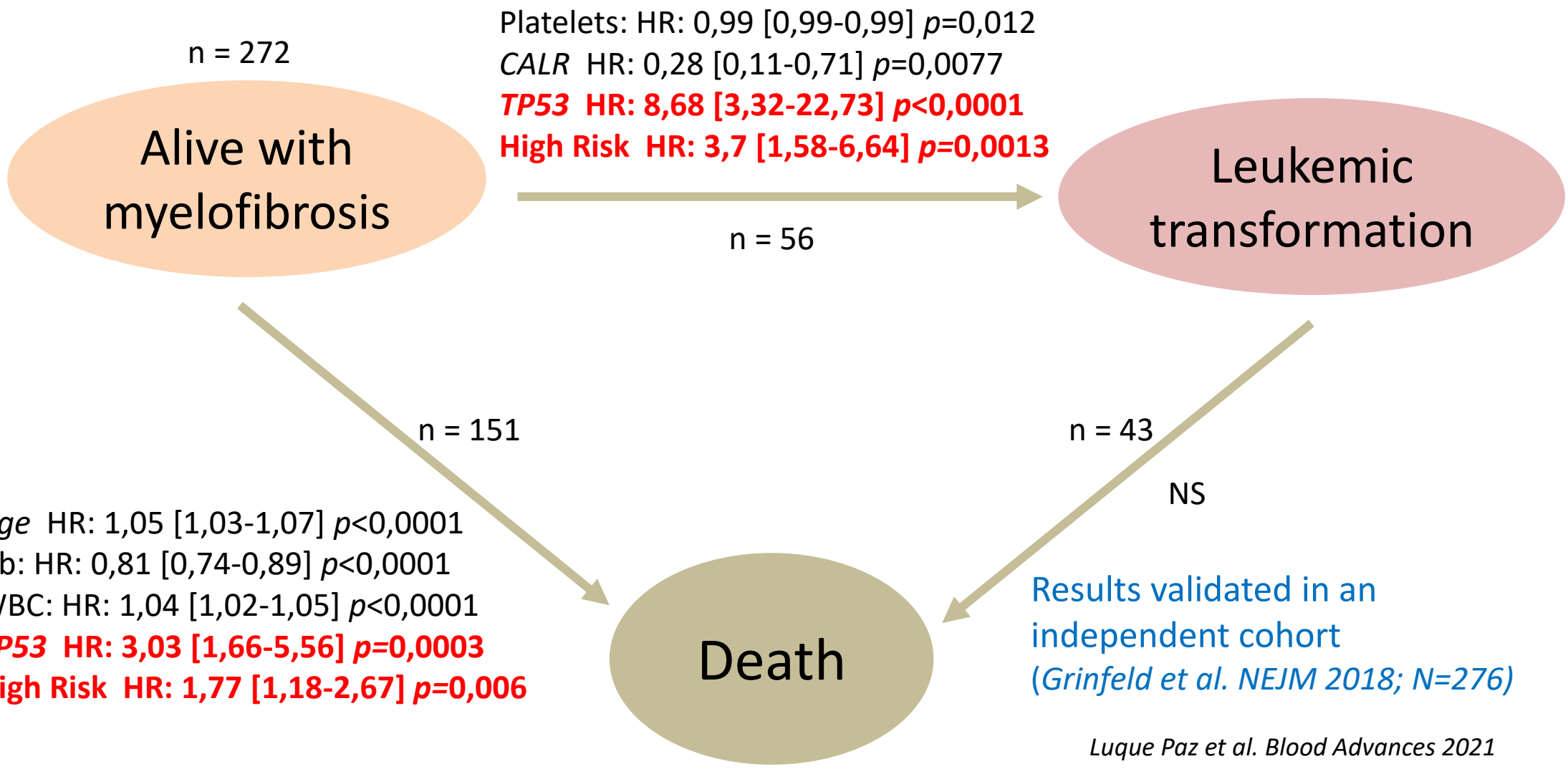




Luque Paz et al. Blood Advances 2021

Redefinition of HMR? The impact of *ASXL1* mutations

✓ Age, gender, hb, platelets, WBC, PMF/SMF, driver mutations, genomic groups

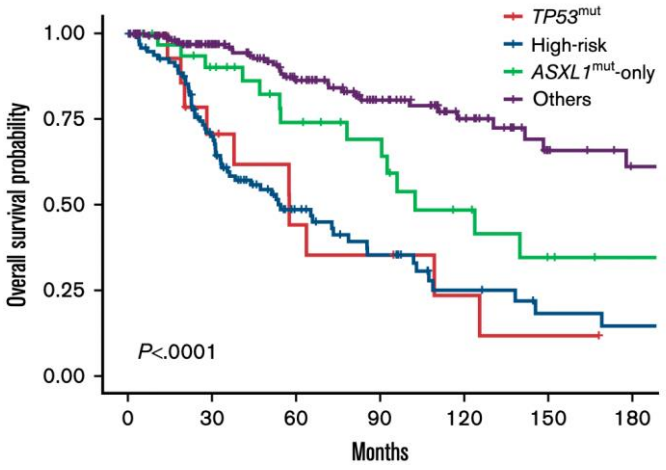


Results validated in an independent cohort
(Grinfeld et al. NEJM 2018; N=276)

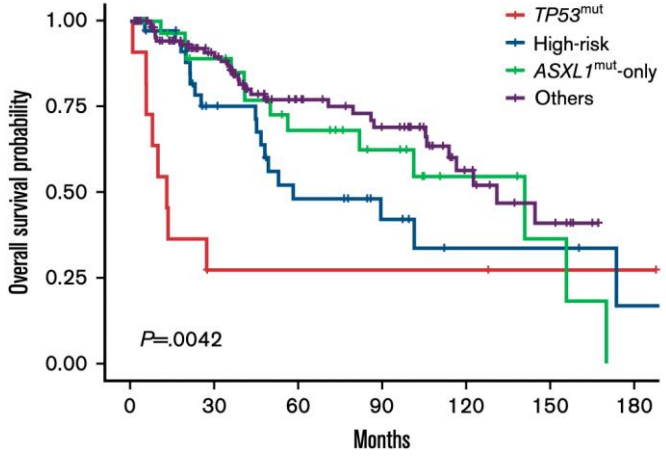
Luque Paz et al. Blood Advances 2021

Redefinition of HMR? The impact of *ASXL1* mutations

Primary myelofibrosis

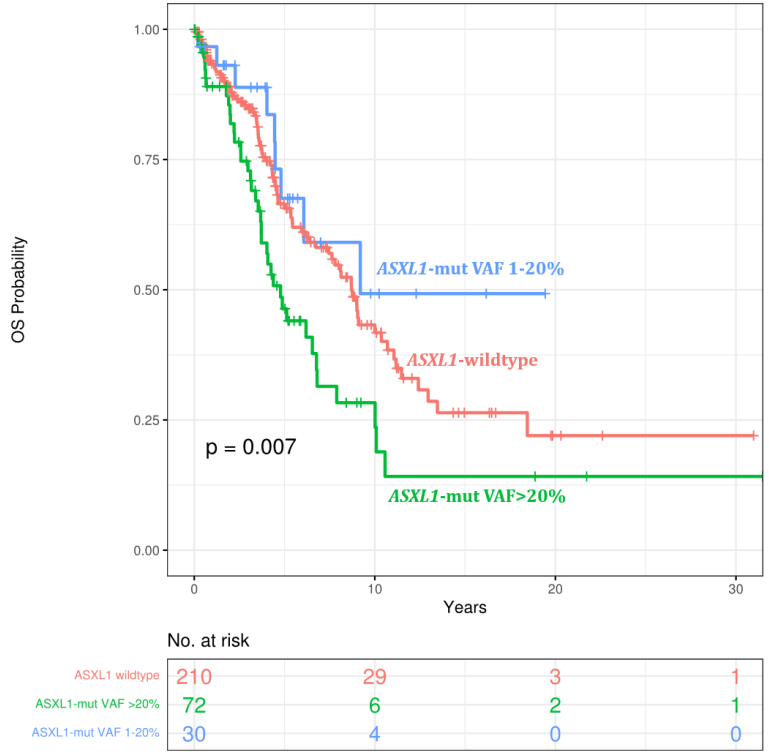


Secondary myelofibrosis



Guglielmelli, Blood Advances 2022

➢ A question of allele burden?

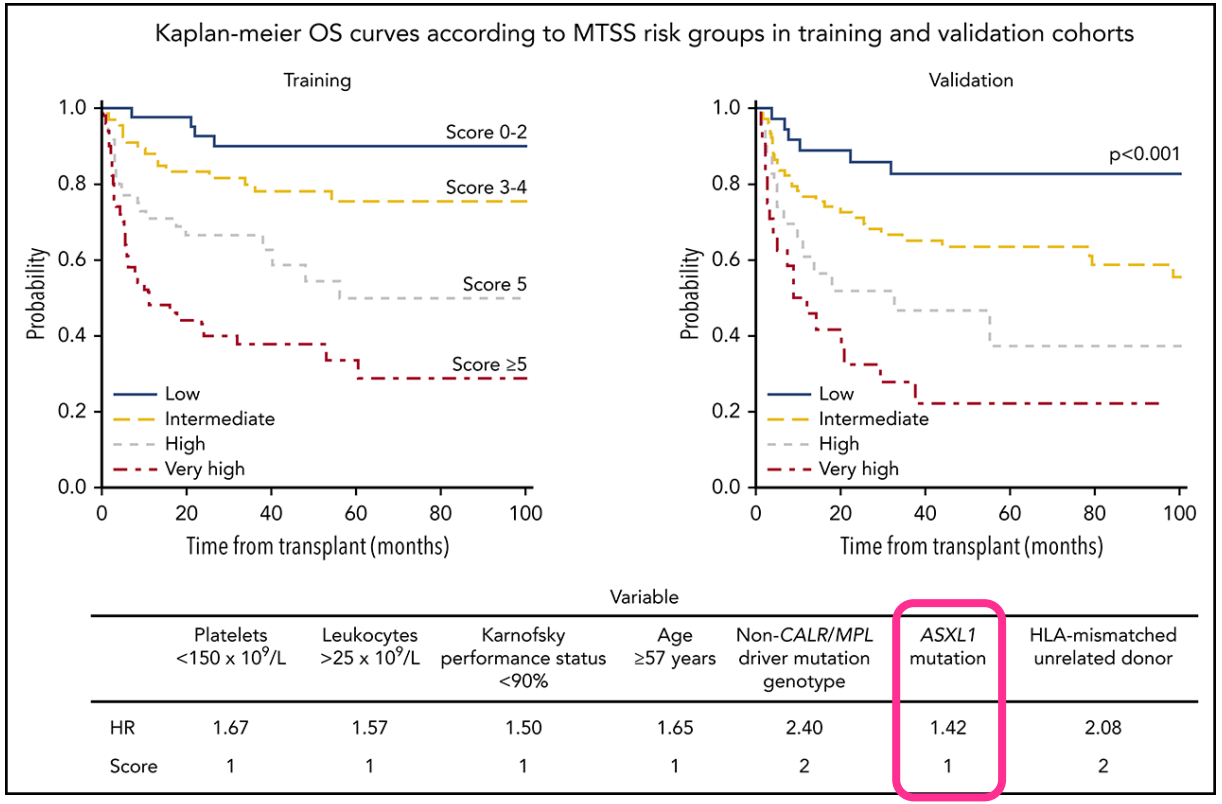


Hernández-Sánchez, AJH 2024

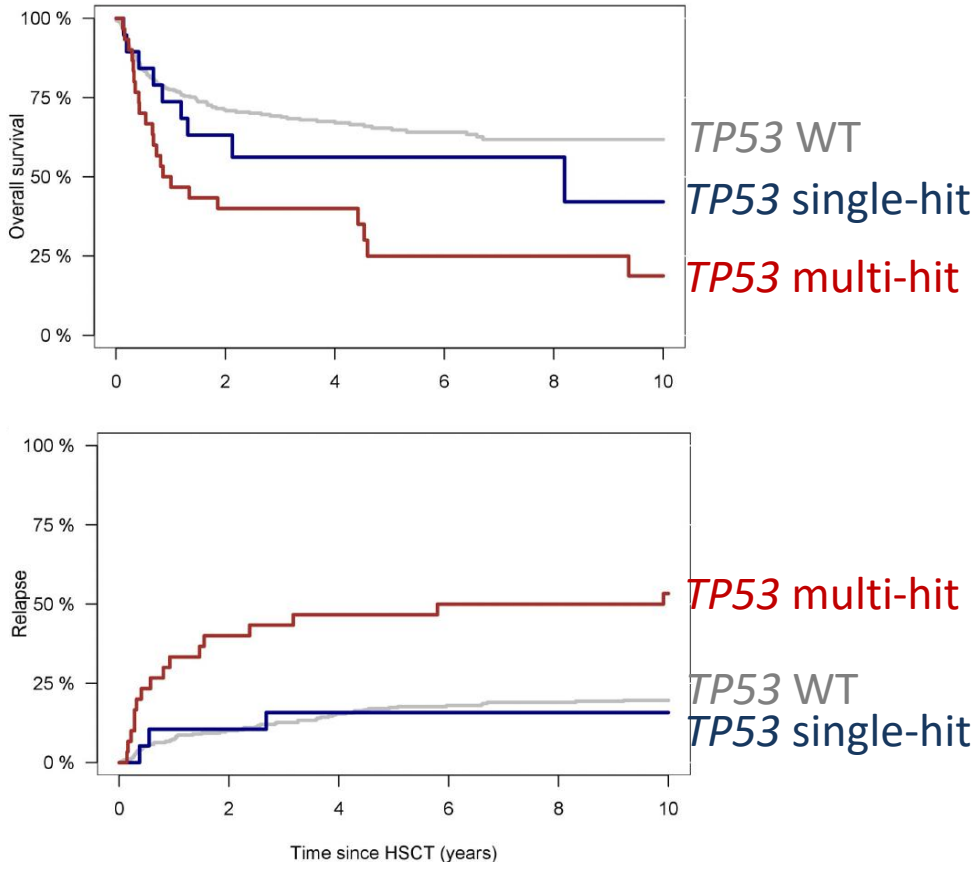
In the context of Stem Cell Transplantation

- MTSS score: *ASXL1* mutations
- *TP53* mutations: the number of allele matters

Gagelmann, Blood 2019 & Blood 2023



Variable	Platelets <150 x 10^9/L	Leukocytes >25 x 10^9/L	Karnofsky performance status <90%	Age ≥57 years	Non-*CALR/MPL* driver mutation genotype	*ASXL1* mutation	HLA-mismatched unrelated donor
HR	1.67	1.57	1.50	1.65	2.40	1.42	2.08
Score	1	1	1	1	2	1	2



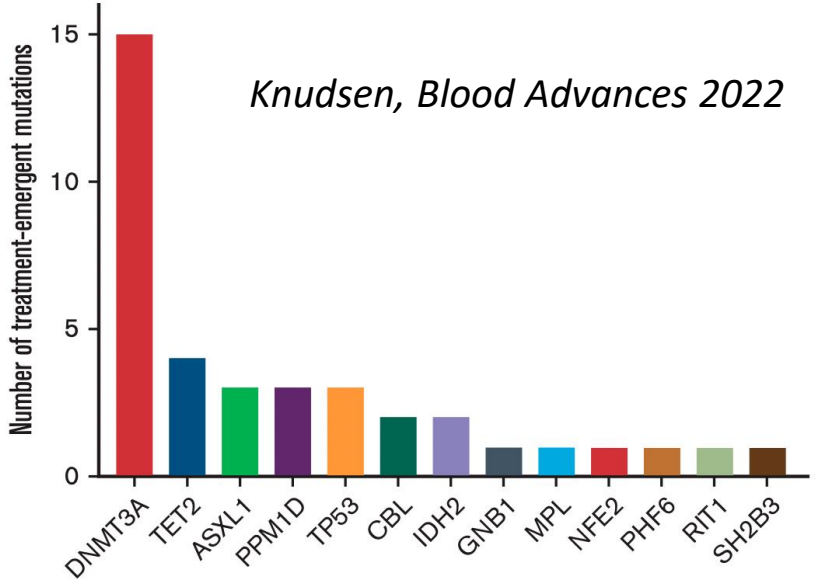
Additional mutations and response to therapy

- Presence of additional mutations is associated to resistance to HU, IFN and ruxolitinib

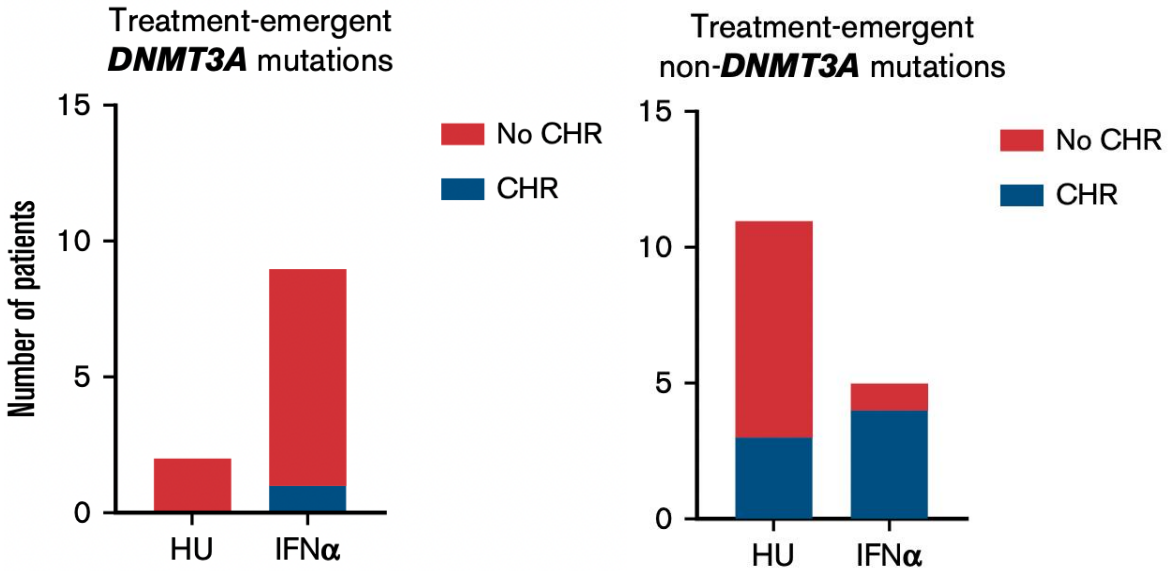
Quintas-Cardama, Blood 2013; Verger, Blood 2015; Newberry, Blood 2017; Alvarez-Larrán et al. Leukemia 2020

- During IFN therapy: emergence of *DNMT3A* mutations associated to resistance

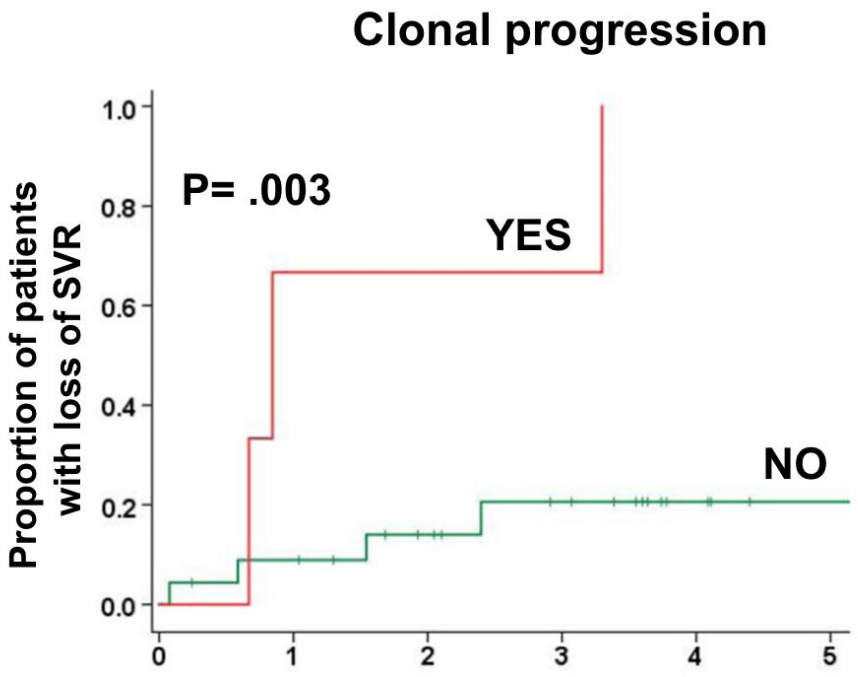
202 patients, NGS at diagnosis and at 24 months



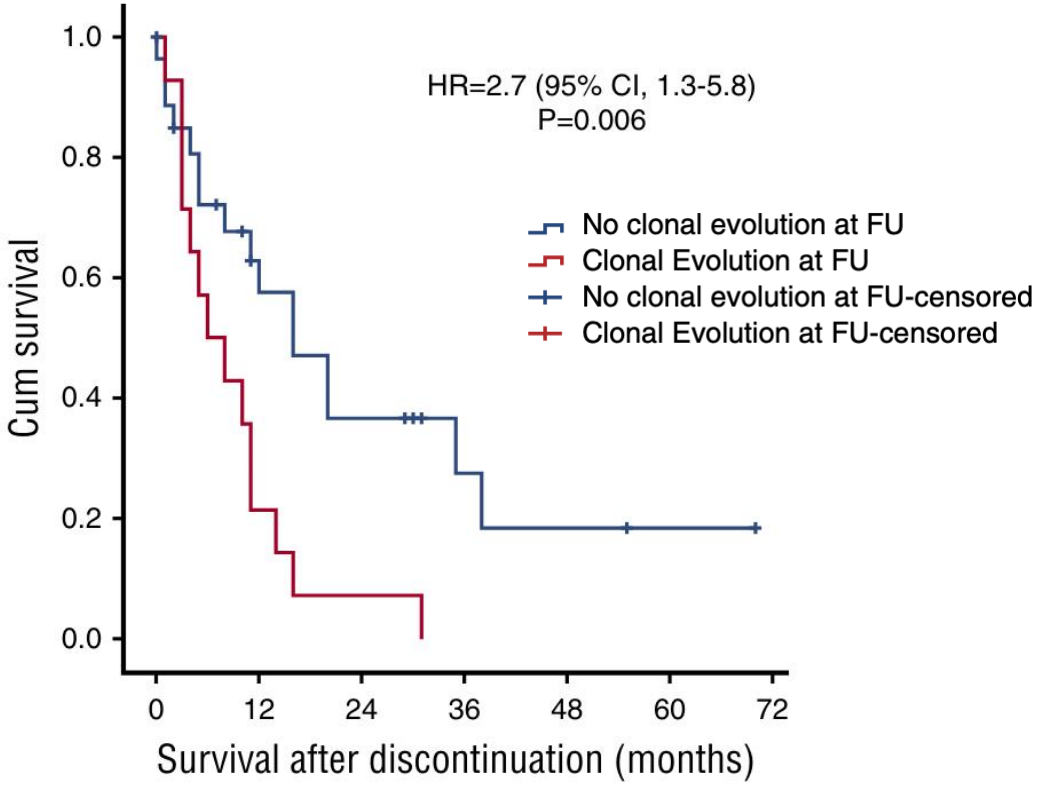
Knudsen, Blood Advances 2022



Acquisition of mutations during ruxolitinib treatment is associated with a worse prognosis



Pacilli, Blood Cancer Journal 2018



Newberry, Blood 2017

Conclusions

- Next-generation sequencing allows the detection of additional mutations
  - ➢ Diagnostic marker in triple-negative patients
  - ➢ Prognostic marker => molecular scores
- Extension of HMR? *TP53* multi-hit
- Indications for molecular evaluation during follow-up need to be defined

Thank you for your attention