Pathogenesis and clinical phenotype of familial myeloproliferative neoplasms

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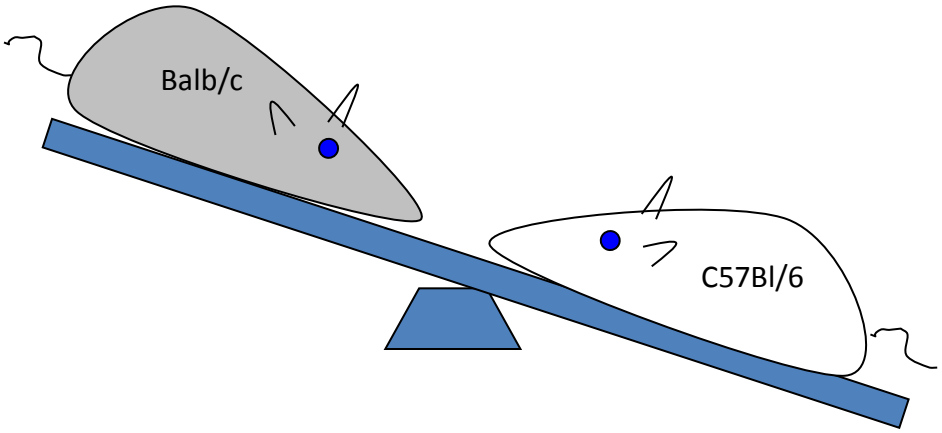
Why hereditary predisposition to MPN?

Several lines of evidence:

- Phenotypic diversity of MPN (ET, PV, PMF) despite a common somatic mutation (*JAK2* V617F)
- Existence of biclonal MPN with 2 independent clones in the same patient
- Common polymorphims predisposing to MPN (*JAK2* GGCC haplotype)
- Familial clustering of MPN

Hereditary predisposition to MPN
1st line of evidence: host modifying influences

JAK2 V617F Mouse model



Strain specific modifiers:
Balb/c mice show markedly elevated WBC count, splenomegaly, fibrosis compared to C57Bl/6 mice

Werning et al, Blood 2006

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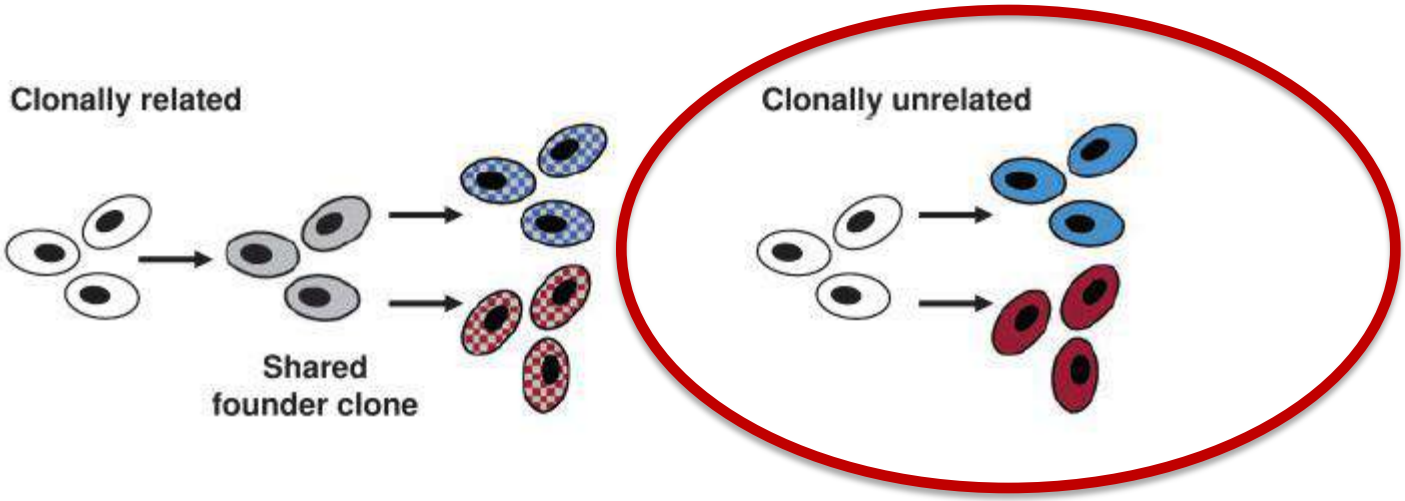
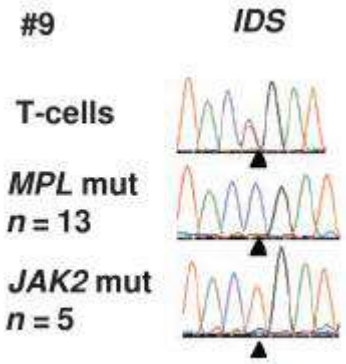
Hereditary predisposition to MPN
2nd line of evidence: biclonal MPN



The mutation-bearing clones express different alleles of the X-linked SNP (rs11549009 in *IDS*)→ the 2 mutations had arisen in unrelated hematopoietic stem cells:

mutation-bearing clones are clonally unrelated



Beer et al, BJH 2009

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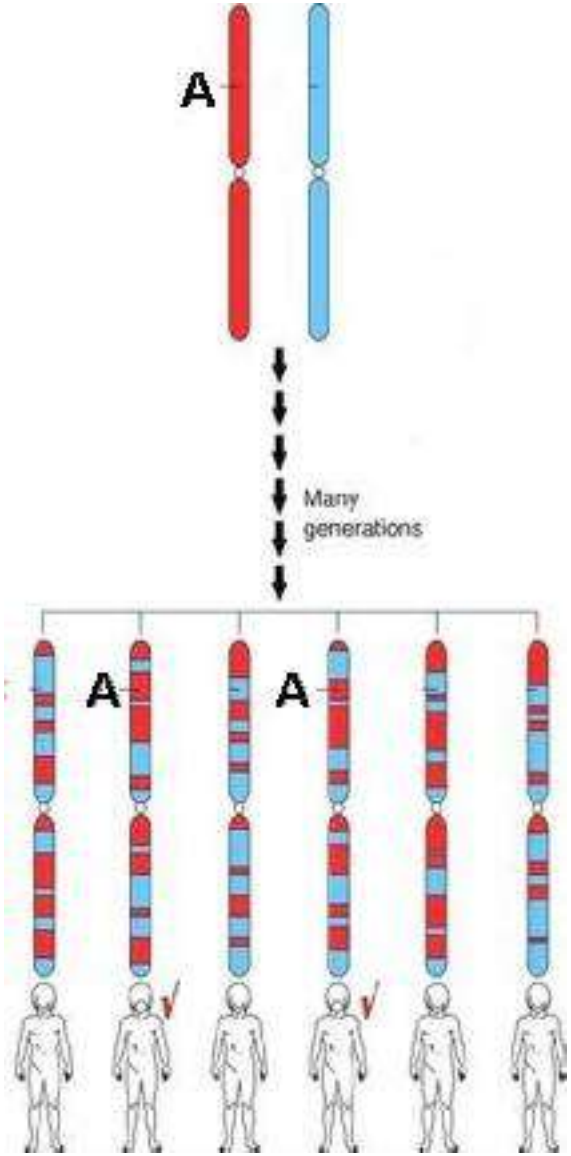
- Existence of biclonal MPN with 2 independent clones in the same patient
- Common polymorphims predisposing to MPN (e.g. *JAK2* GGCC haplotype, *TERT, MECOM, HBS1L-MYB*)
- Familial clustering of MPN

What haplotype is?

- Set of DNA sequences (one or more loci or SNP) very close along the chromosome
- They tend to be inherited together due to absence of recombination during crossover (genetic linkage)



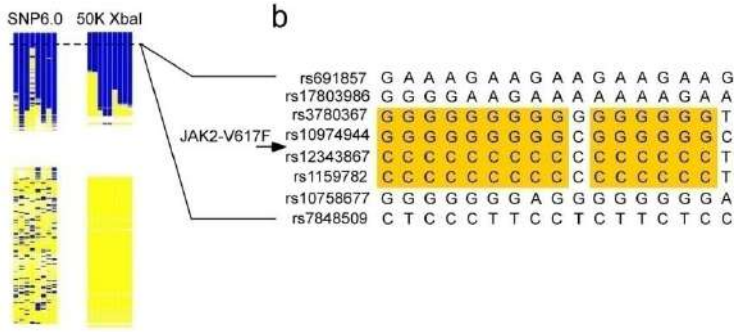
http://hapmap.ncbi.nlm.nih.gov/originhaplotype.html.en

Hereditary predisposition to MPN

3rd line of evidence: *JAK2* haplotype predisposing to MPN

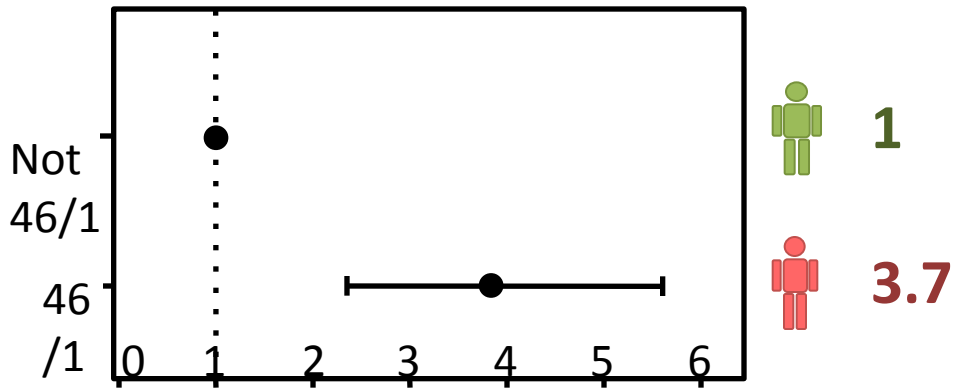
88% of all *JAK2*-V617F mutations occur on the "GGCC" *JAK2* haplotype

Olcaydu et al, Nat Genet 2009

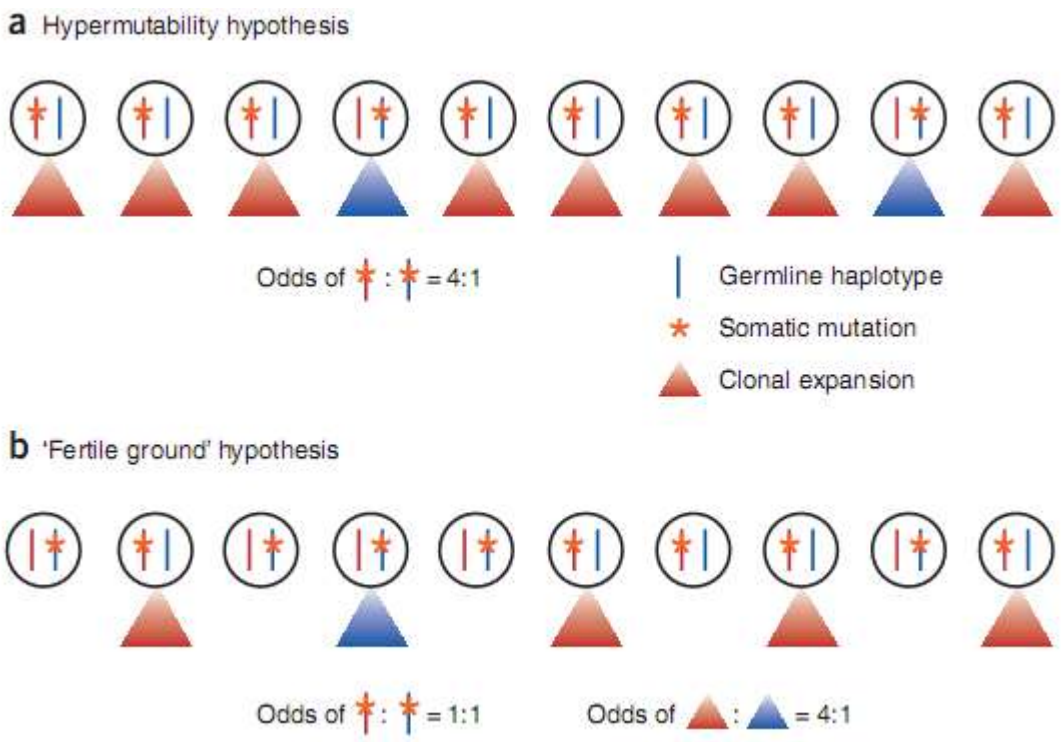


46/1 is a significant constitutional risk factor for development of V617F positive MPNs [OR =3.7; 95% CI, 3.1 - 4.3]

Jones et al, Nat Genet 2009



Role of the *JAK2* haplotype: 2 hypotheses



Hypermutability: there is a specific mutational mechanism by which V617F preferentially arises on a 46/1 haplotype

Fertile ground: mutation rate on 46/1 and non-46/1 haplotypes is the same but the probability of clinically manifest disease is higher if V617F arises on 46/1

Campbell, Nat Genet 2009

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Hereditary predisposition to MPN
4th line of evidence: familial clustering of MPN

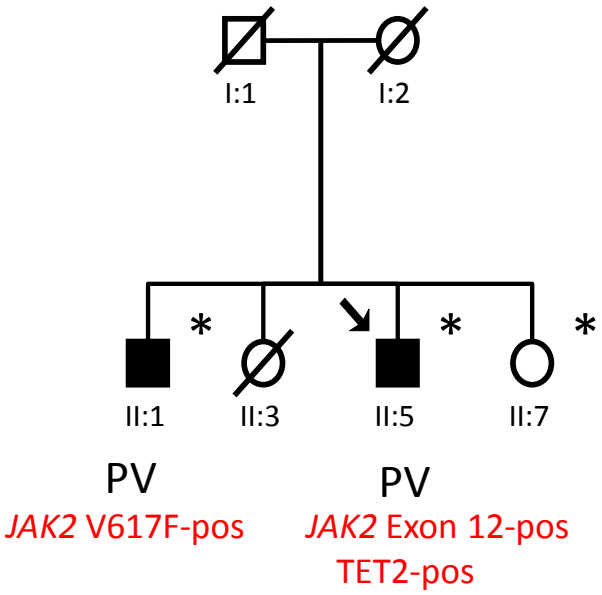
- Familial clustering in 7.6% of apparently sporadic MPN
- Relatives of MPN patients have a 5- to 7-fold increased risk of developing MPN

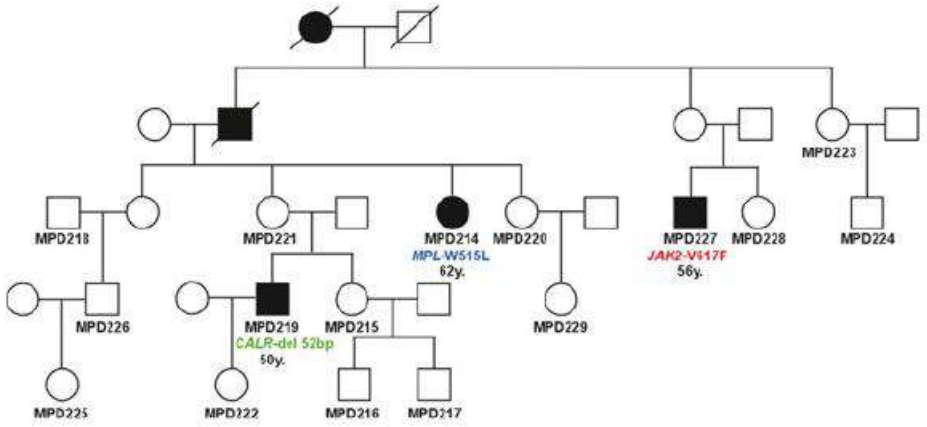
Rumi et al, JCO 2007

Landgren et al, Blood 2008

Familial MPN: pathogenesis

- The known recurrent MPN-associated mutations (*JAK2*, *CALR*, *MPL*) are acquired as somatic mutations also in familial MPN
- A genetic predisposition to acquisition of *JAK2/CALR/MPL* mutations may be inherited





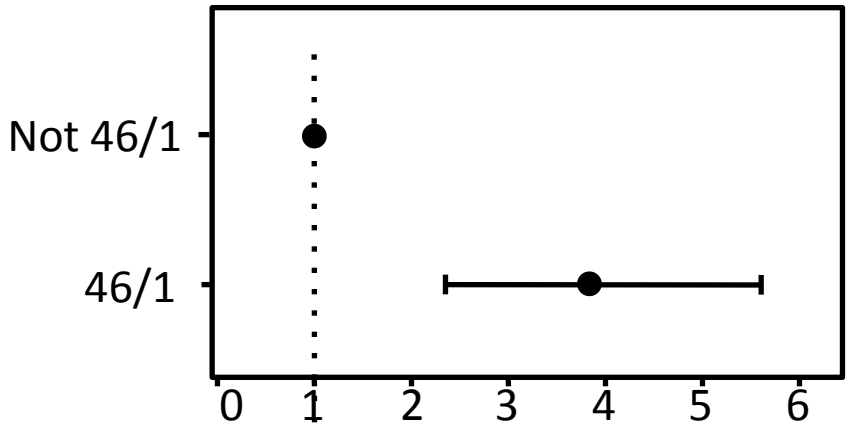
Bellanne-Chantelot et al, Blood 2006

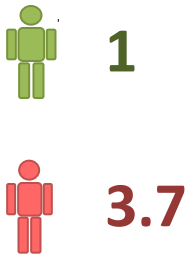
Rumi et al, JCO 2007

Harutyunyan et al, Blood 2015

Familial MPN: pathogenesis→predisposing SNP

Sporadic





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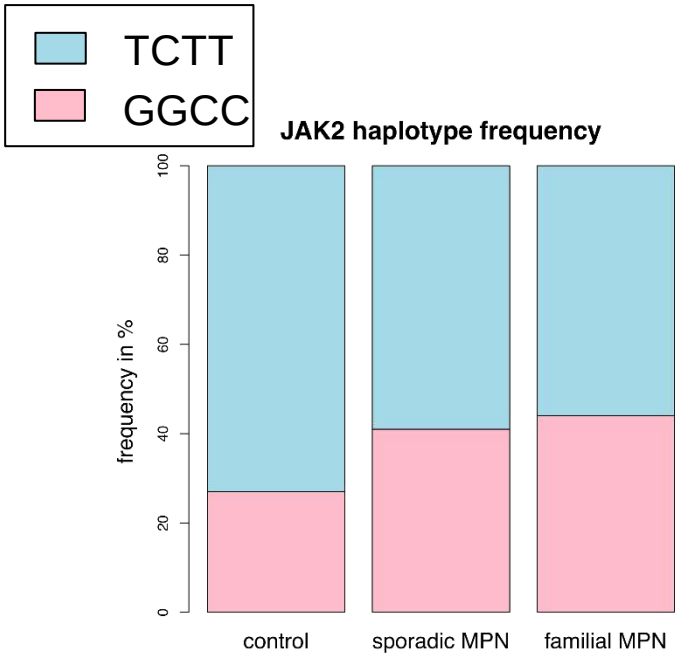
Jones et al, Nat Genet 2009

Familial

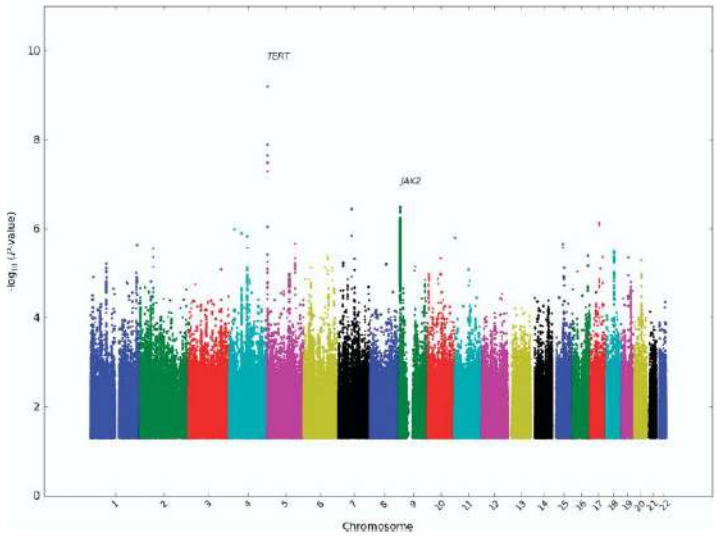


- The GGCC haplotype frequency was higher in familial (0.44) and sporadic (0.42) MPN than in control (0.27)
- Familial MPN did not differ significantly from sporadic MPN in disease risk conferred by the GGCC haplotype (*P ns*)

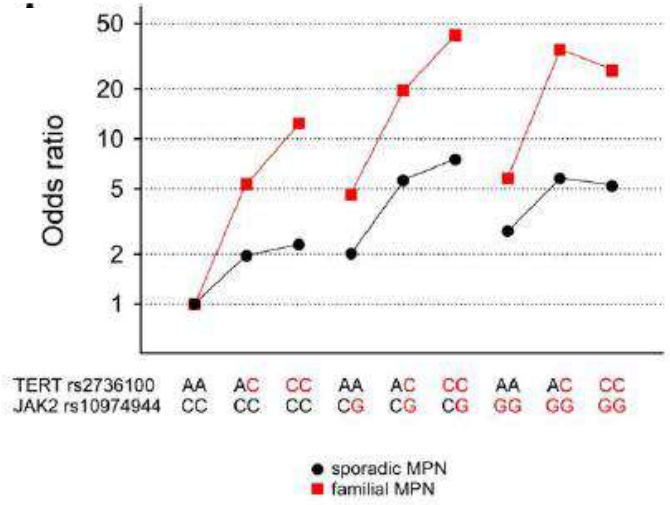
Olcaydu, Rumi et al, Haematologica 2011

Familial MPN: pathogenesis→predisposing SNP

Sporadic



Familial



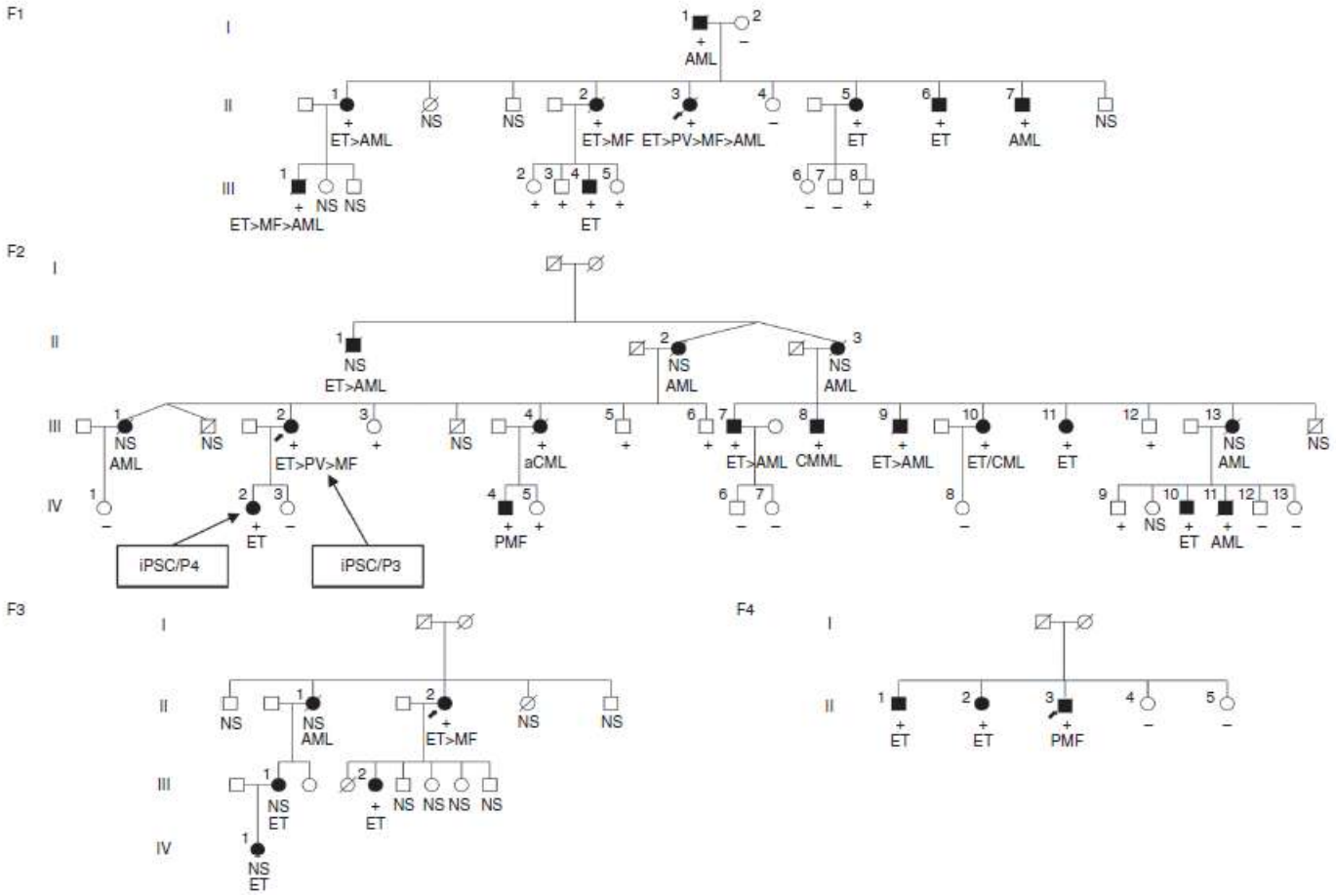
- *TERT* rs2736100_C and *JAK2* GGCC are independently predisposing to MPN.

- *TERT* rs2736100_C significantly enriched (P=0.0090) in familial MPN compared to sporadic MPN→ low-penetrance variants may be responsible for a substantial part of familial clustering in MPN.

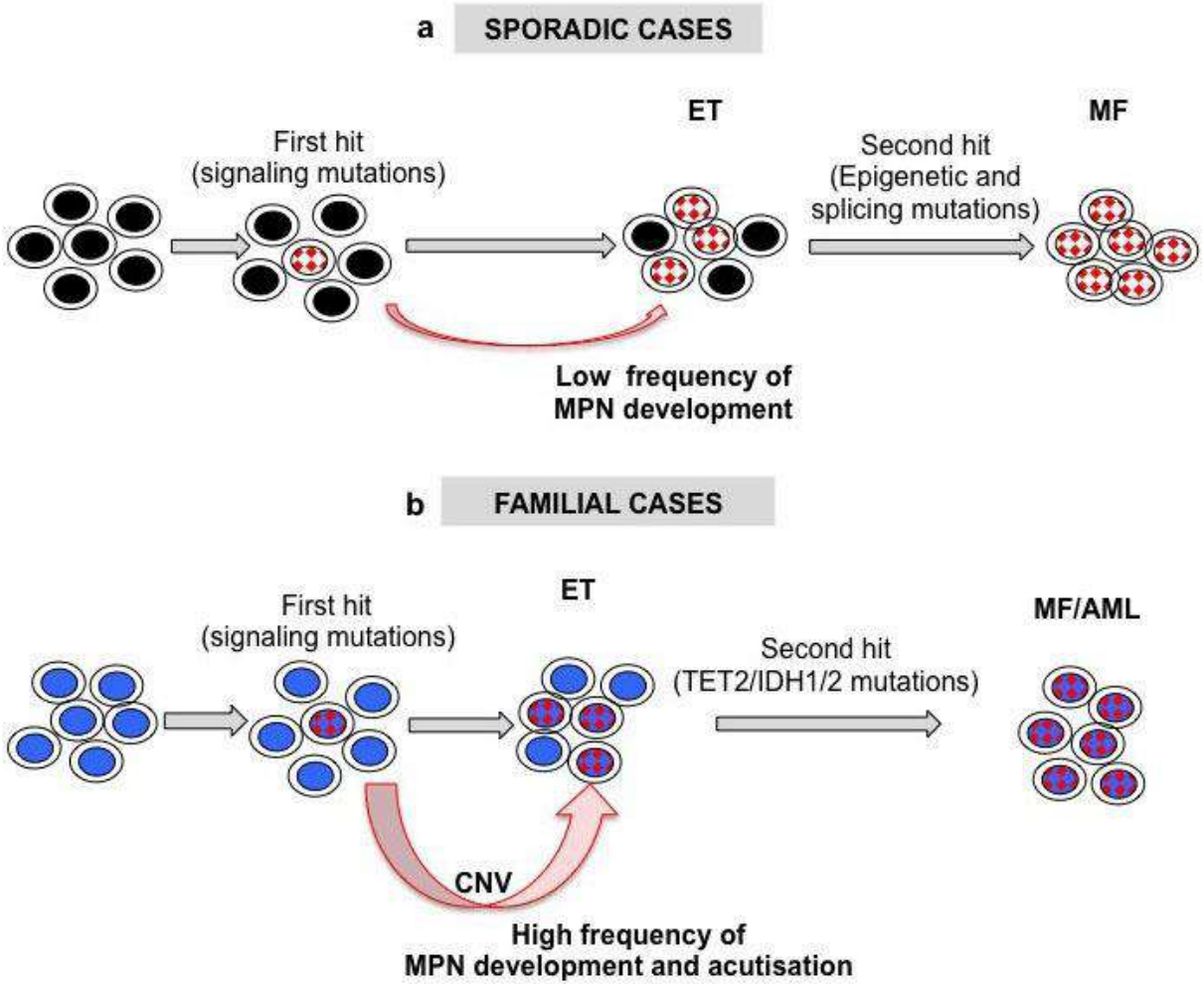
Oddsson al, Luekemia 2014

Jager al, AJH 2014

The first identified germline genomic alteration in familial MPN: duplication of ATG2B and GSKIP

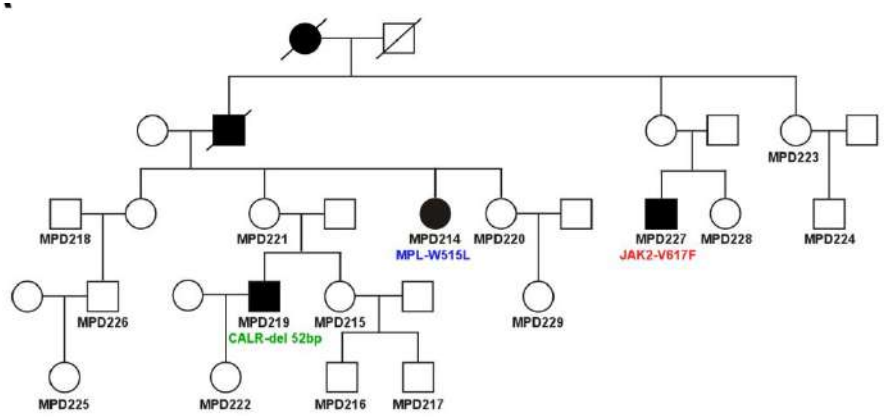


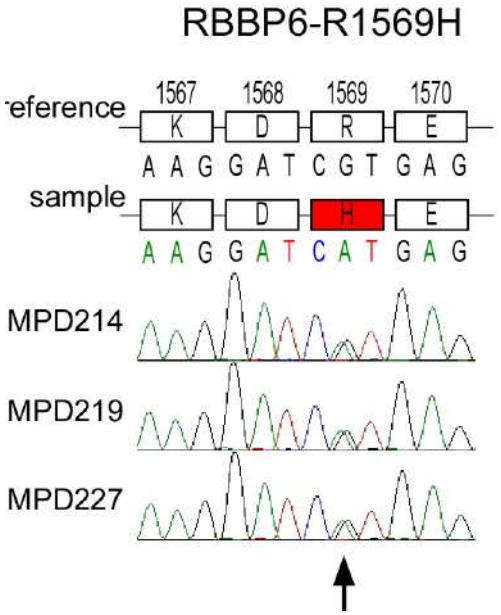
Saliba et al, Nature Gen 2015

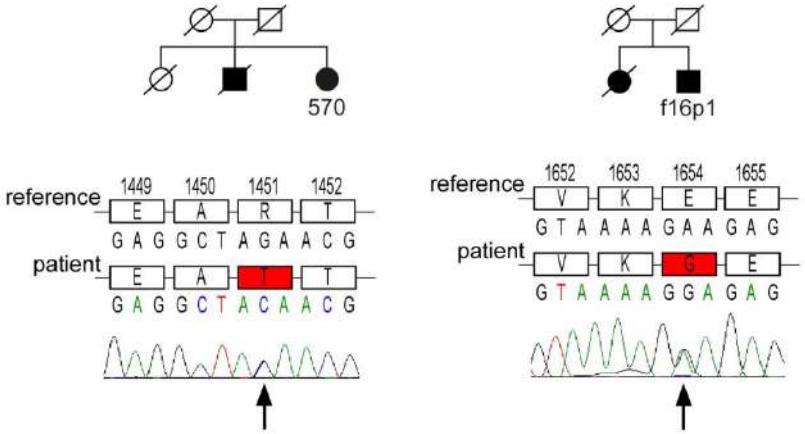


Saliba et al, Nature Gen 2015

The second identified germline genomic alteration in familial MPN: RBBP6 mutations







Germline *RBBP6* mutations in about 5% (3/67) of familial MPN cases.

Harutyunyan et al, Blood 2015

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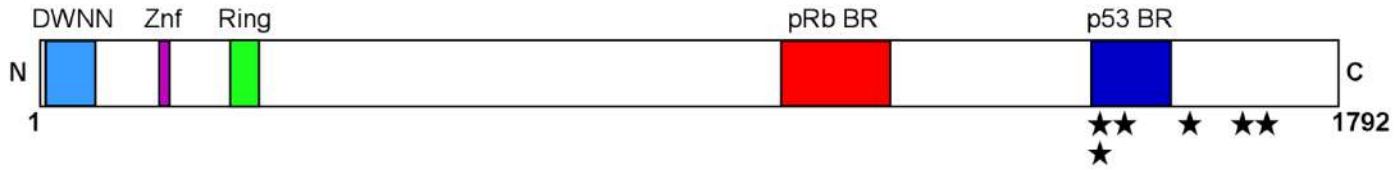


RBBP6 ubiquitinates and degrades p53.

The *RBBP6* mutations are all located in the vicinity of its p53-binding domain so they may affect p53 functions.

Mutant RBBP6 might cause an elevation in somatic mutagenesis rates through inhibition of p53 function and deregulation of cell cycle.

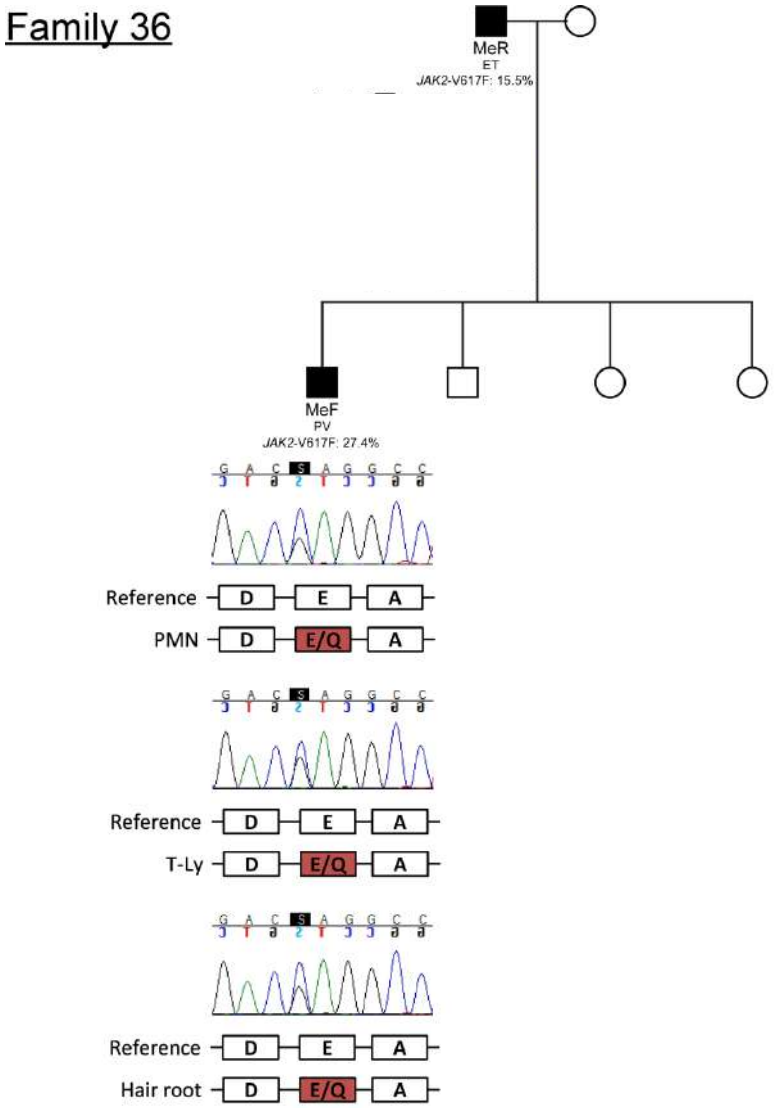
Harutyunyan et al, Blood 2015

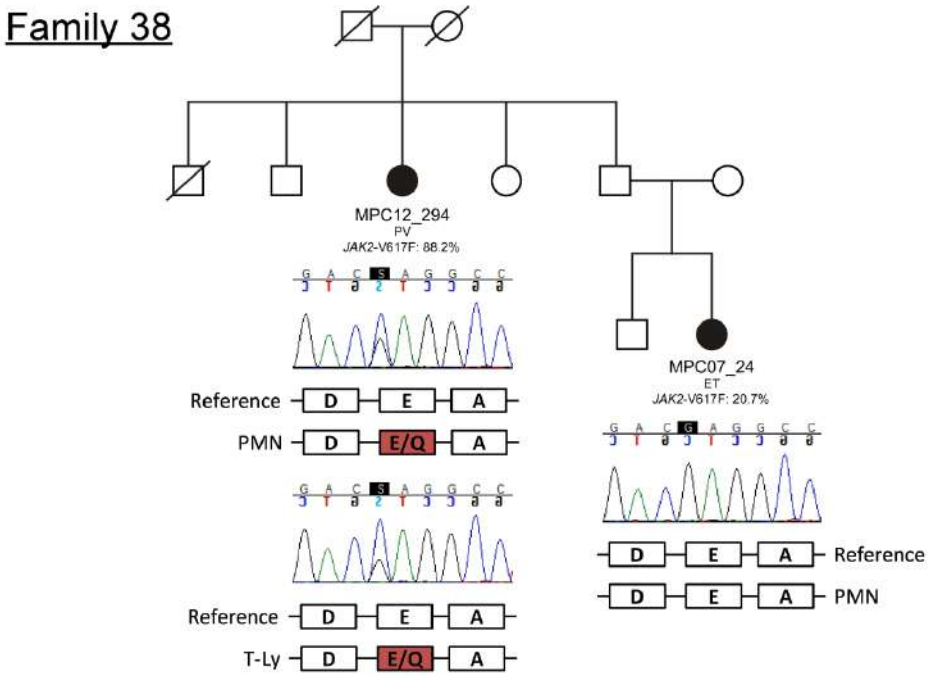
The third identified germline genomic alteration in familial MPN: LNK mutation

WES in 16 families.

Some mutations in known genes (*JAK2*, *LNK*, *TET2*, *DNMT3A*, *IDH1* …)

First gene validated with Sanger sequencing: *LNK* E208Q in one patient belonging to family 36.



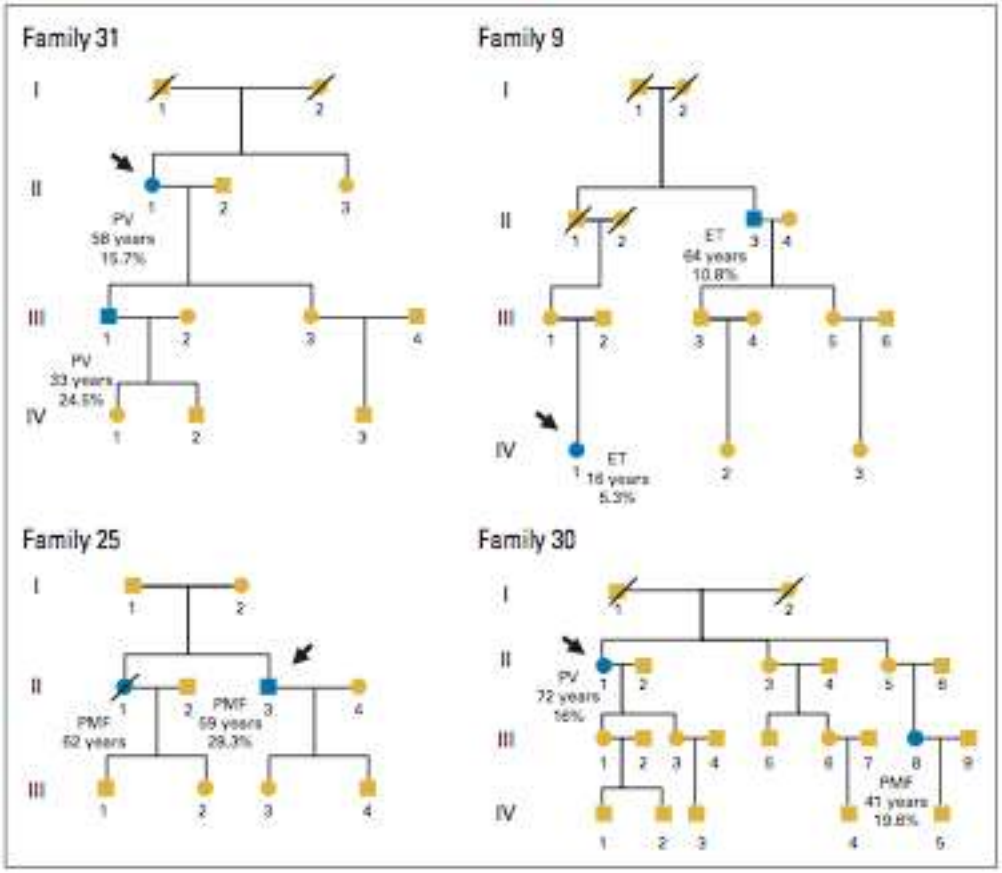


94 families (149 pts) screened for *LNK* mutations

Germline *LNK* mutations in 2/94 (2%) MPN families.

Rumi et al., Blood 2016

Familial clustering of MPN: clinical phenotype



Clinical phenotype may be homogeneous (all relatives affected have the same MPN) or mixed (different MPNs in the same pedigree)

Rumi et al, JCO 2007

Familial clustering of MPN: clinical phenotype at diagnosis

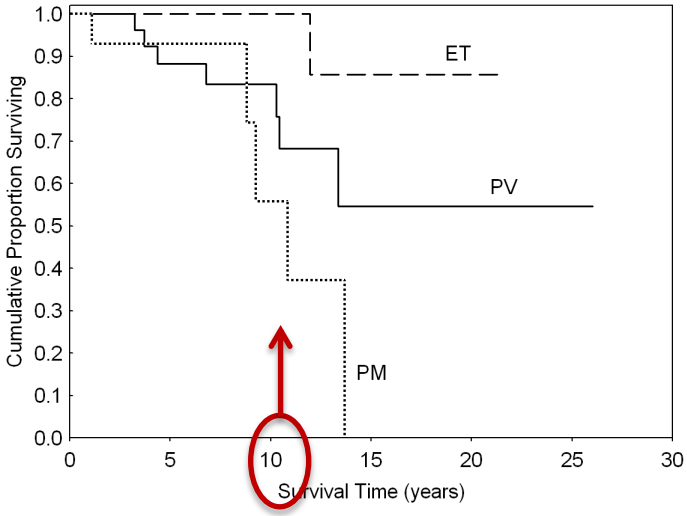
	Familial PV (n=36)	Sporadic PV (n=188)	*P*	Familial ET (n=24)	Sporadic ET (n=157)	*P*	Familial PMF (n=15)	Sporadic PMF (n=78)	*P*
Age, years Median Range	 59 24-77	 57 18-85	 0.1	 58 16-74	 51 17-83	 0.1	 57 23-78	 57 15-83	 0.1
Sex Male Female	 26 10	 100 88	 0.06	 8 16	 54 103	 0.9	 8 7	 37 41	 0.7
WBC, x10^9/l Median Range	 8.4 4-17.8	 10.1 4.1-32.2	 0.1	 6.9 5.1-12.5	 8.5 4.6-18.3	 0.1	 11.7 4.5-37.7	 9.2 2.2-19.7	 0.1
Hb, g/dl Median Range	 18.6 16.6-23	 18.4 16.5-23.6	 0.1	 14.3 12.2-17.2	 14.1 12.7-17.5	 0.1	 13 10.1-15.9	 13.4 6.6-16.3	 0.1
PLT x10^9/l Median Range	 306 133-1000	 458 137-2000	 0.1	 816 499-1498	 756 451-2541	 0.1	 500 33-1832	 714 46-3279	 0.1
Splenomegaly, No. (%)	7 (19)	47 (25)	0.5	2 (8)	3 (2)	0.1	9 (60)	48 (62)	0.1
Thrombosis, No. (%)	10 (28)	44 (23)	0.6	2 (8)	23 (14)	0.5	3 (20)	12 (15)	0.7
Hemorrhage, No. (%)	1 (3)	15 (8)	0.4	1 (4)	5 (3)	0.5	0	5 (6)	0.5

Familial MPN similar to sporadic MPN

Rumi et al, JCO 2007

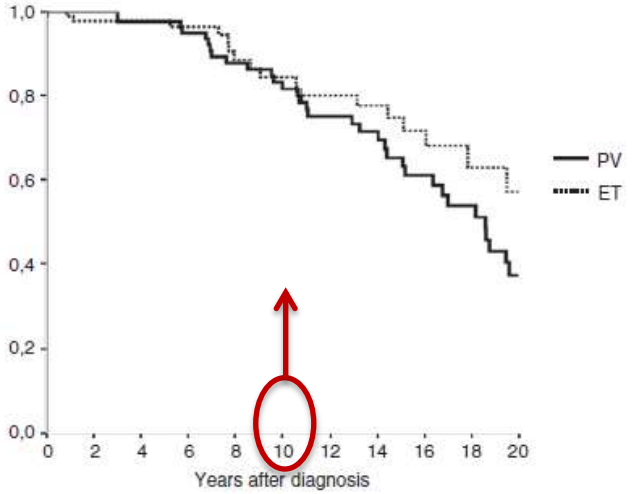
Familial clustering of MPN: overall survival

ET 100%; PV 83%; PMF 56%



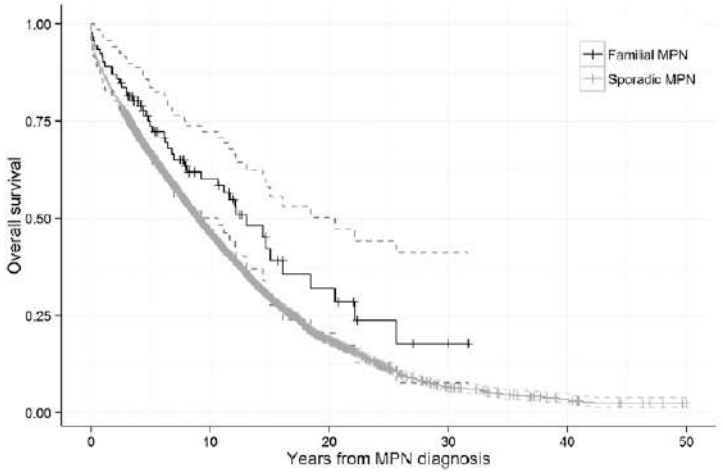
Rumi et al, JCO 2007

ET 83%; PV 83%



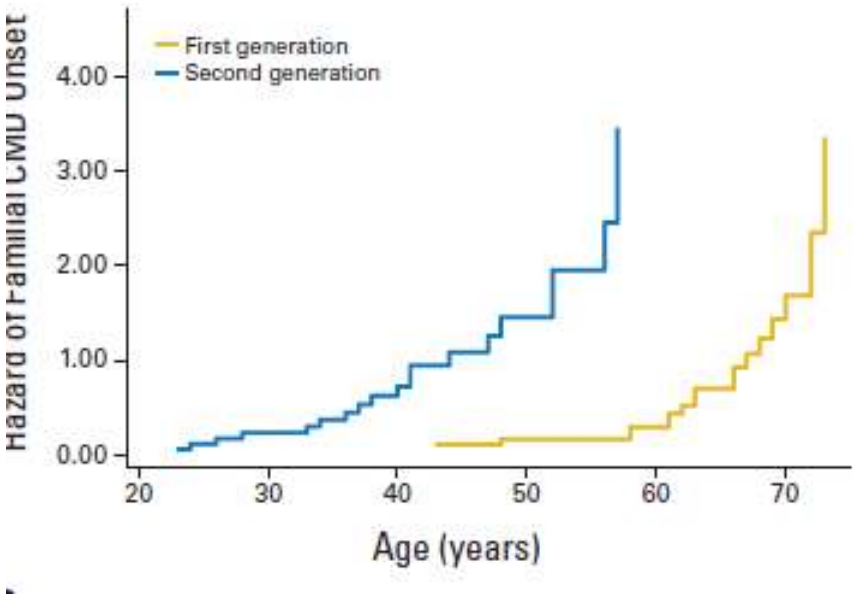
Malak et al, Blood Cell Mol Dis 2012

HR 1.1, 95% CI 0.9-1.5, *P*=0.39



Hultcrantz et al, Blood 2015

Familial clustering of MPN: disease anticipation

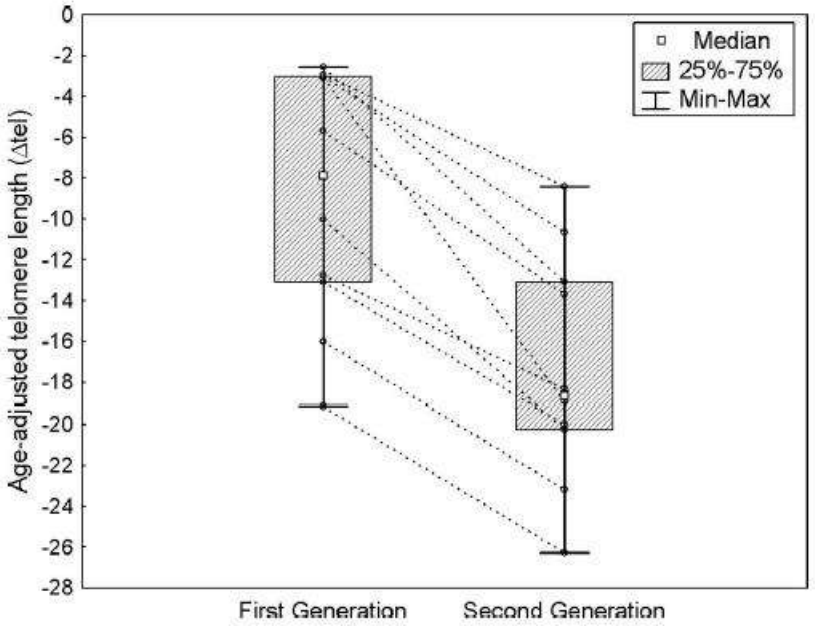


The second-generation patients have significantly younger age at diagnosis than first generation patients (64 years vs 40 years, *P*<.001).

Nelson-Aalen method shows that the age-dependent hazard of MPN onset is significantly higher in second-generation than in first-generation patients (*P*<.001).

Rumi et al, JCO 2007

Familial clustering of MPN: disease anticipation



Age-adjusted telomere length in 10 families with 2-generation pairs.

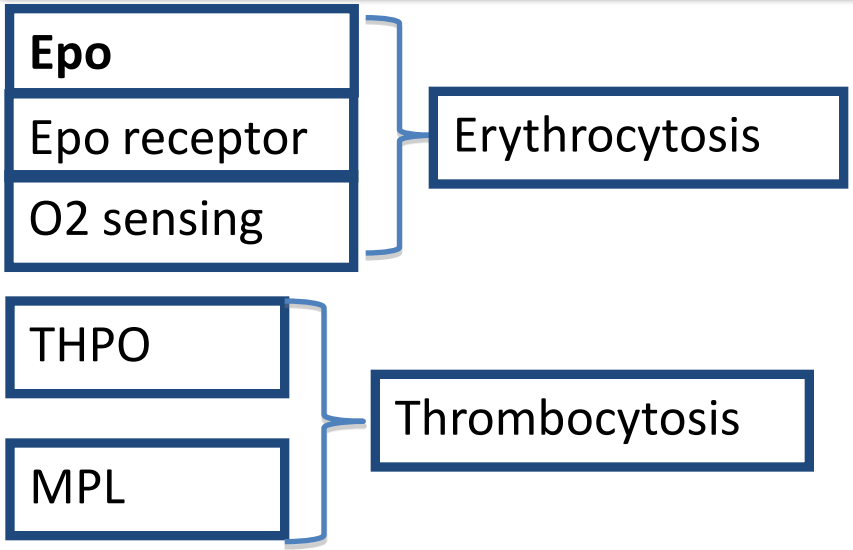
Telomere length is adjusted for age to avoid the age effect on telomere shortening (it is expressed as Δtel=difference between the observed length and the age-adjusted normal telomere length predicted from the linear regression line).

Second-generation patients have significantly shorter telomeres than first-generation patients (*P*=.005).

Rumi et al, Blood 2008

Familial clustering of MPN: differential diagnosis

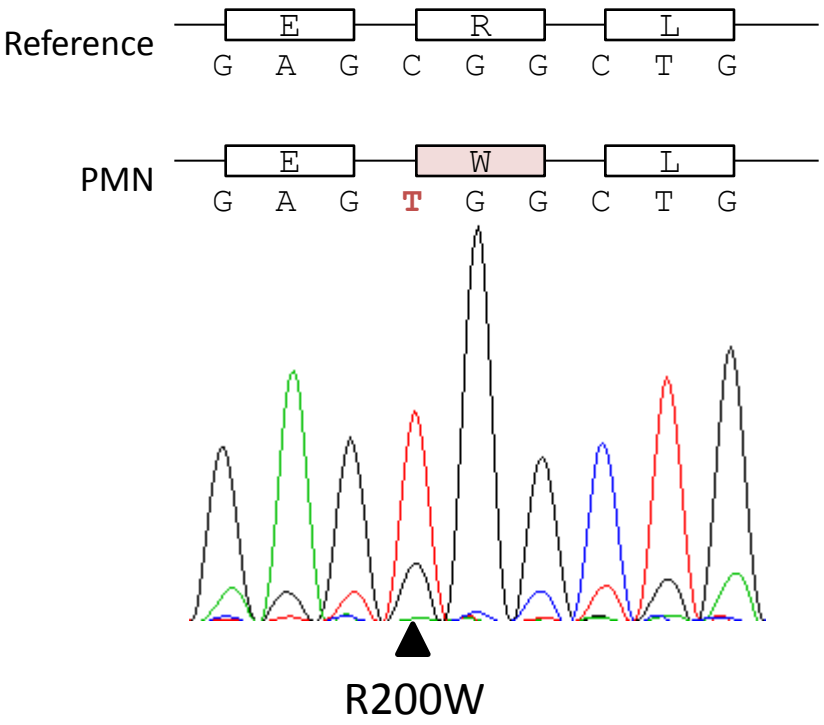
Hereditary thrombocytosis/ erythrocytosis	Familial MPN
1. Polyclonal hematopoiesis	1. Clonal hematopoiesis
2. Mendelian inheritance	2. Complex inheritance
3. *MPL, THPO, EPOR,* ***EPO*** mutations are disease causing genes	3. *JAK2*,*CALR*,*MPL* mutations are secondary genetic events

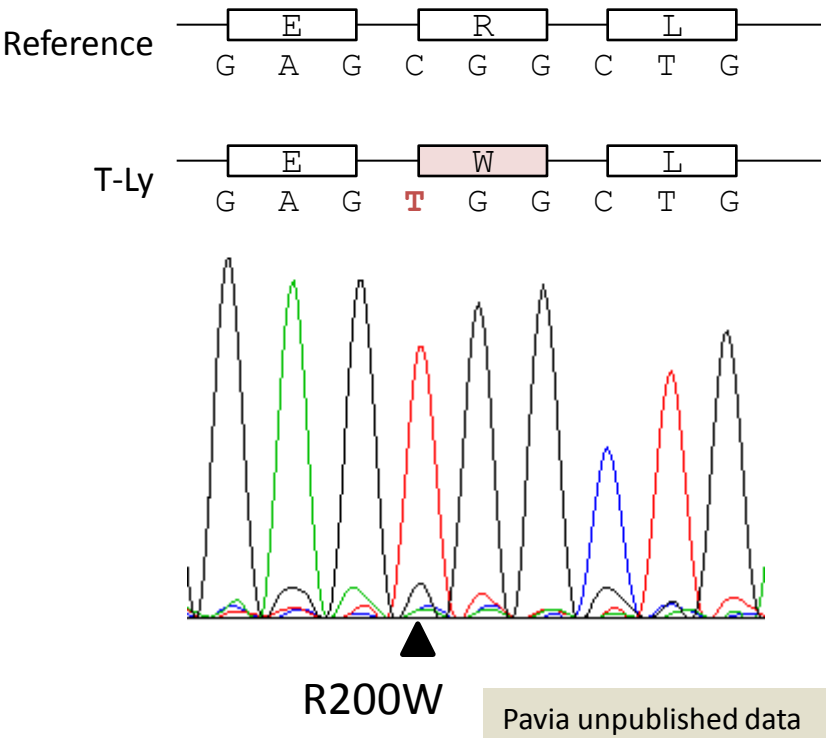


- Essential thrombocythemia
- Polycythemia vera
- Primary myelofibrosis

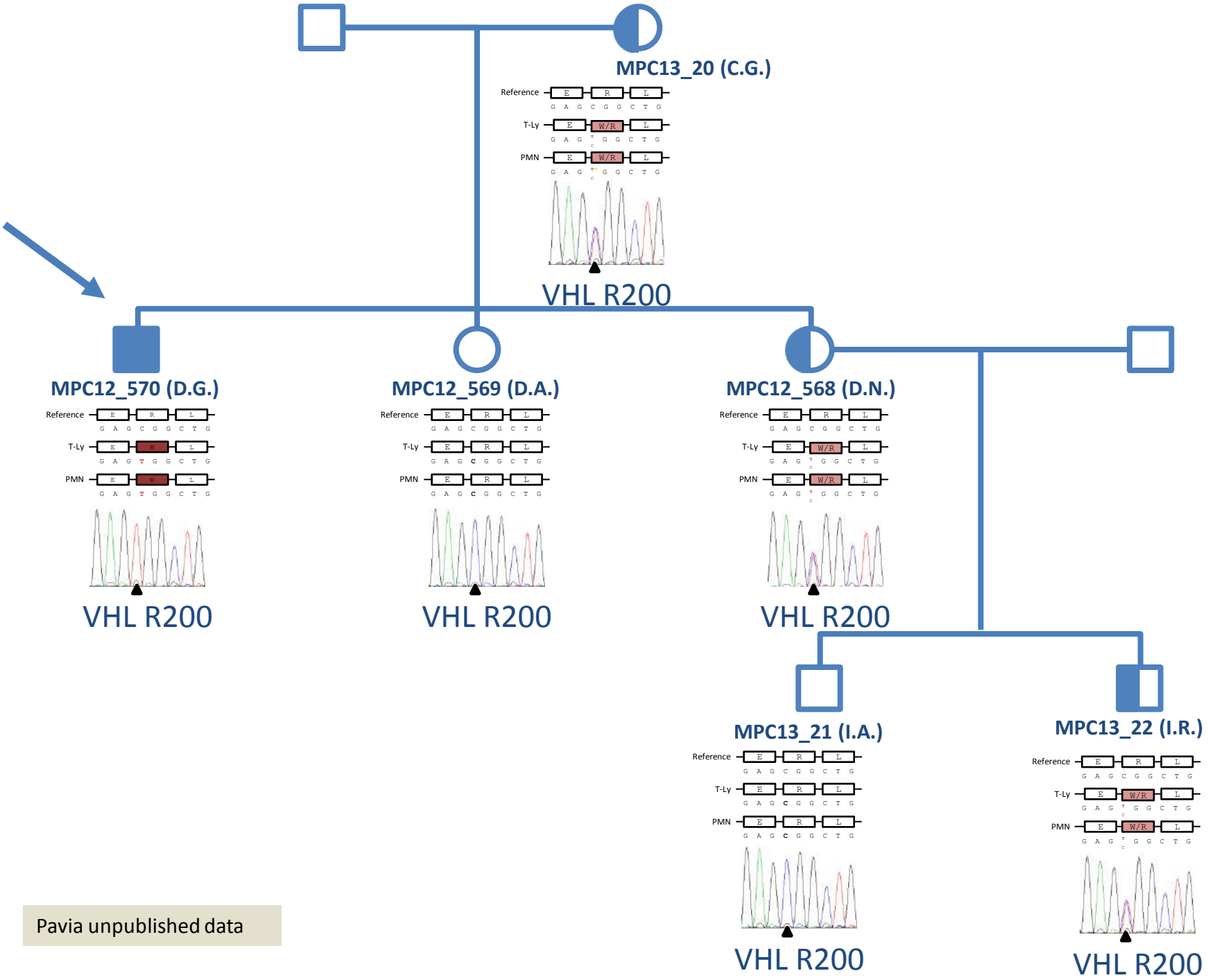
Chuvash Polycythemia: case report

- ♂ 36 yr, initial diagnosis: PV, treated with IFN because eccessive need of phlebotomy
- *JAK2* V617F and exon 12 neg
- Normal P50, Epo 115 mU/Ml
- Bone marrow biopsy not consistent with PV
- Sequencing VHL gene: VHL mutation





Pavia unpublished data



Pavia unpublished data

Conclusions

- A genetic predisposition to acquisition of *JAK2/MPL/CALR* is inherited
- TERT s2736100_C contributes in part to familial clustering
- Germline duplication of *ATG2B* e *GSKIP* and germline mutations of *RBBP6* and *LNK* in a small % of familial MPN
- We still do not know whether a unique “predisposing gene” exists, or if there is an extreme genetic heterogeneity (many different germline mutations in a small % of familial MPN)
- genetic alterations in non coding regions might be responsible for familial predisposition→ WGS in the most informative families→project GR-2016-02361272

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GR-2016-02361272

And all our patients…