MPN – origins and evolution

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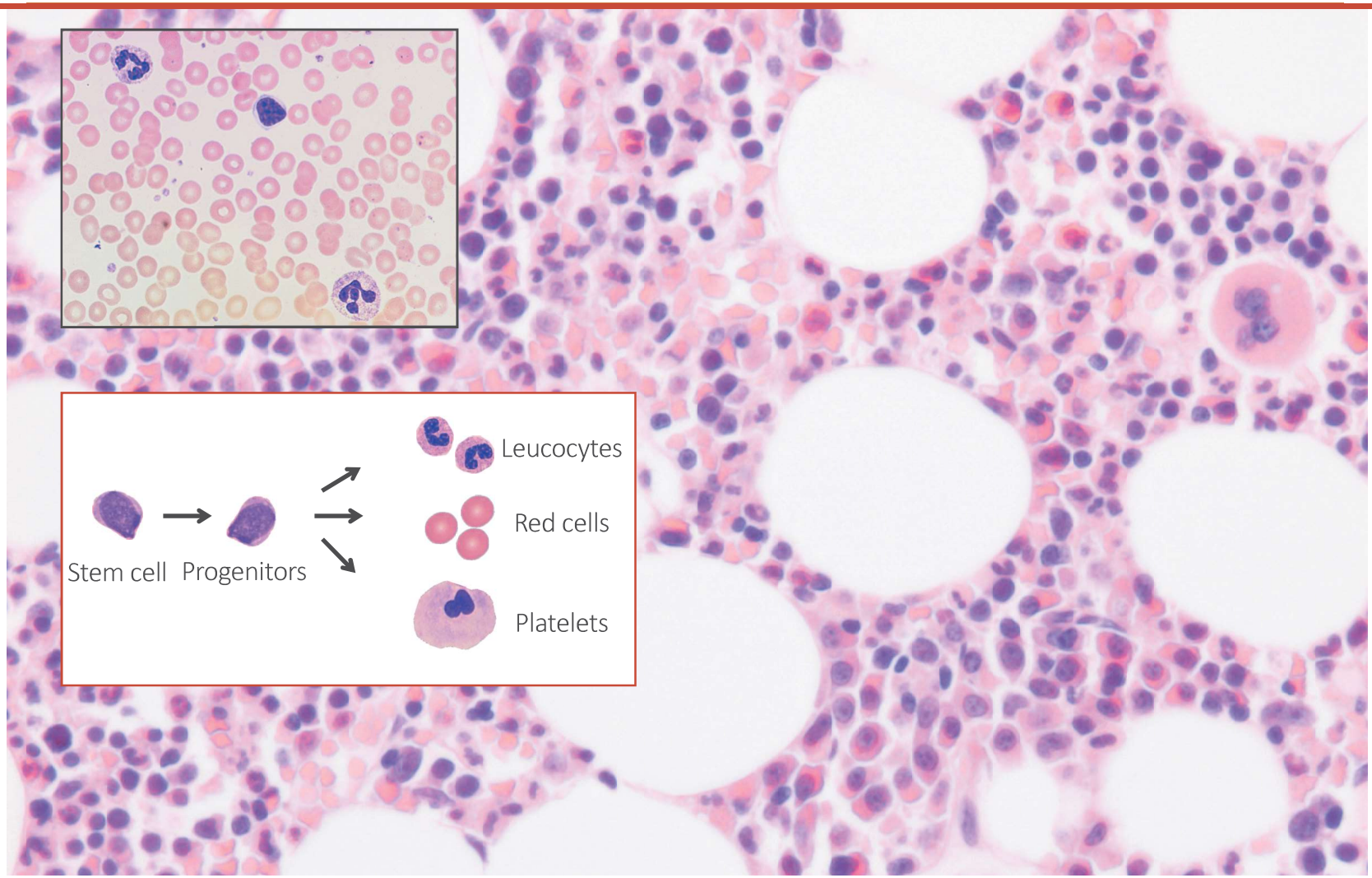
wellcome sanger institute

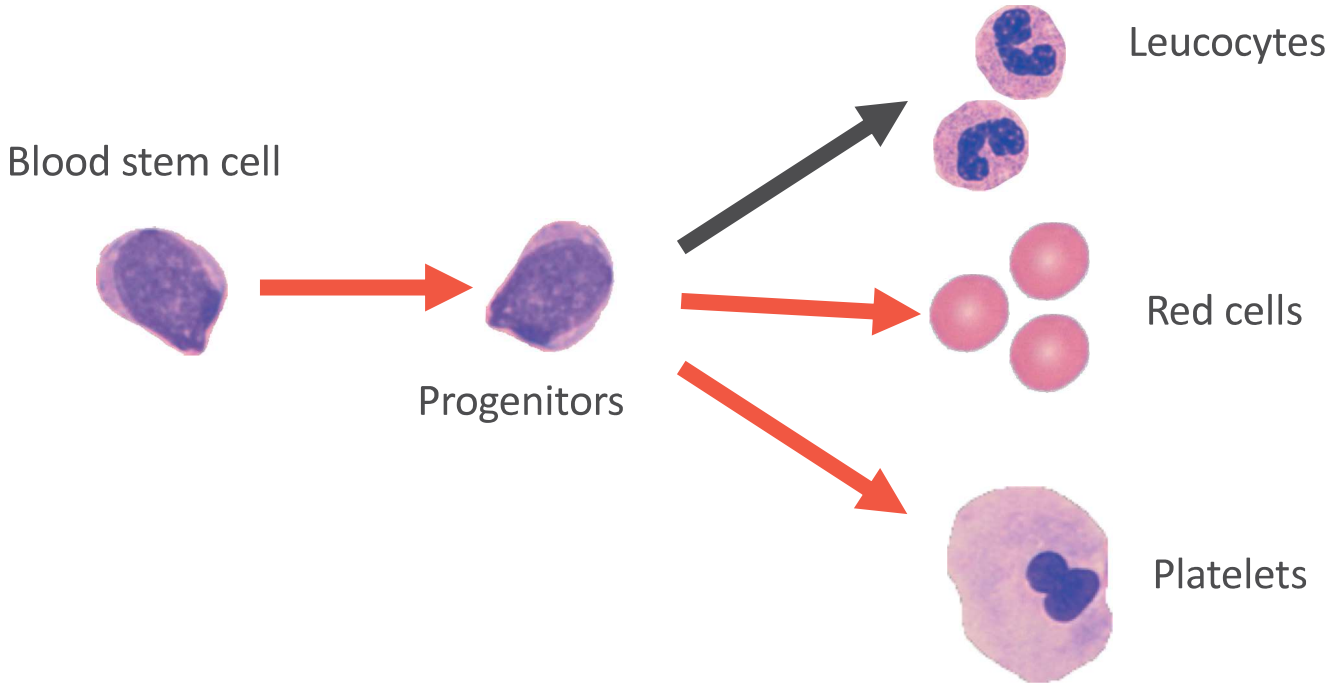
Wellcome - MRC Cambridge Stem Cell Institute

UNIVERSITY OF CAMBRIDGE

Blood and bone marrow

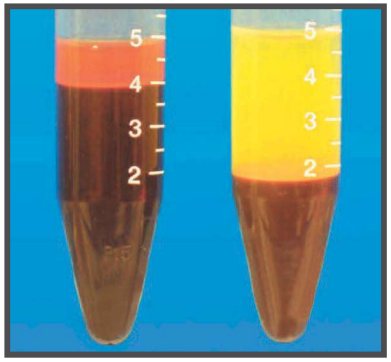


Myeloproliferative neoplasms

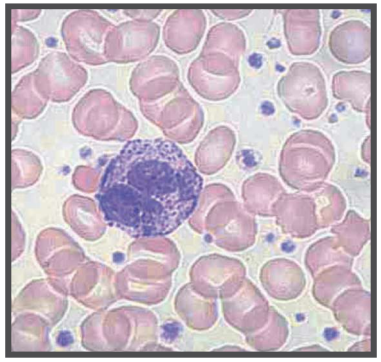


Myeloproliferative neoplasms

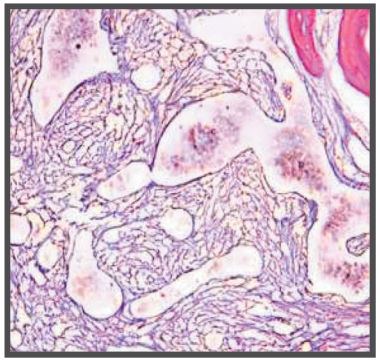
PV



ET



MF



- *What causes it?*
- *How long have I had it for?*
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Myeloproliferative neoplasms

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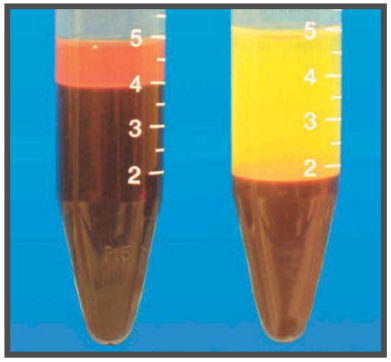

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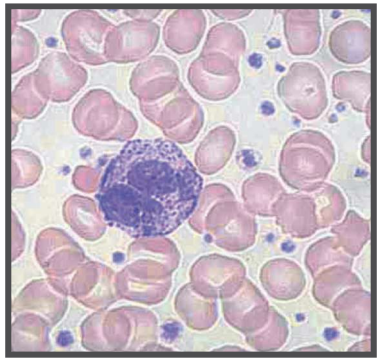

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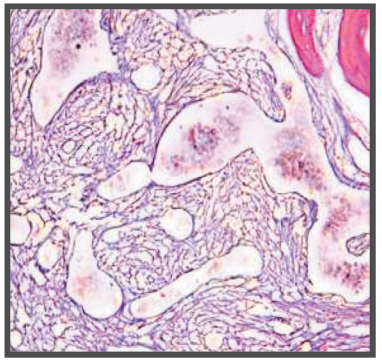
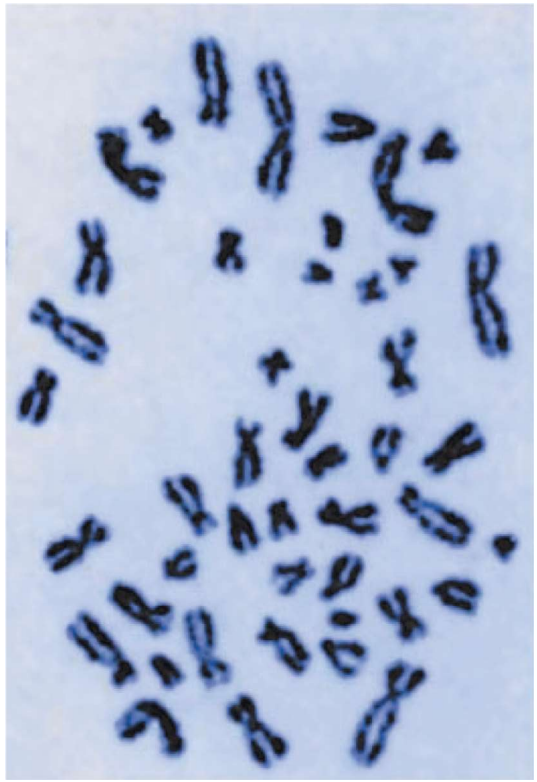
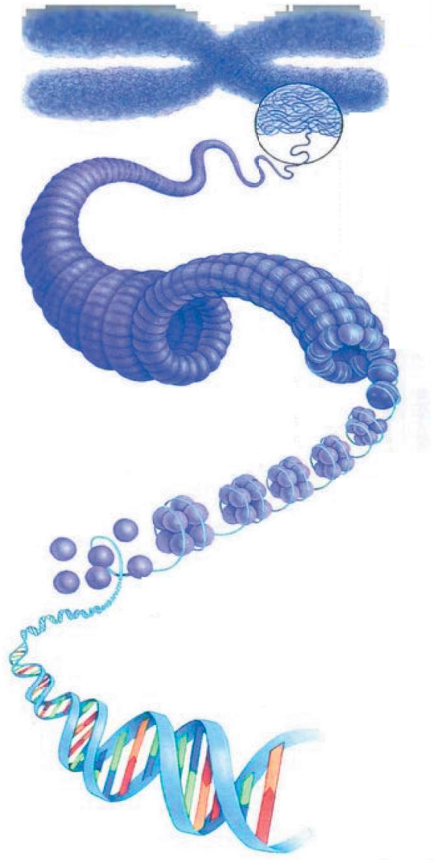


- ***What causes it?***
- *How long have I had it for?*
- *How fast did it grow?*

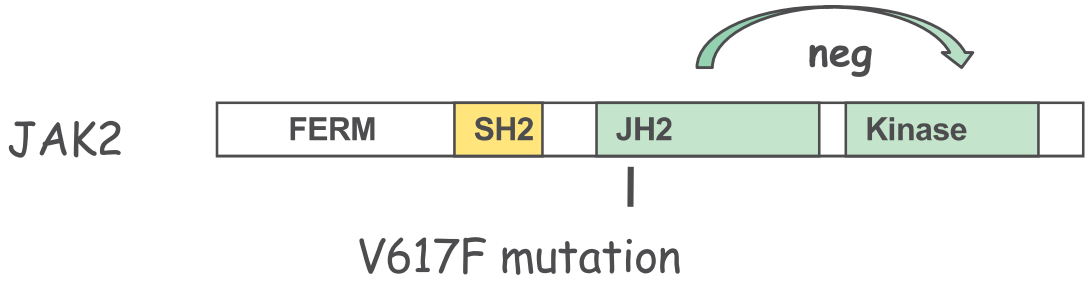
The code of life – counting chromosomes



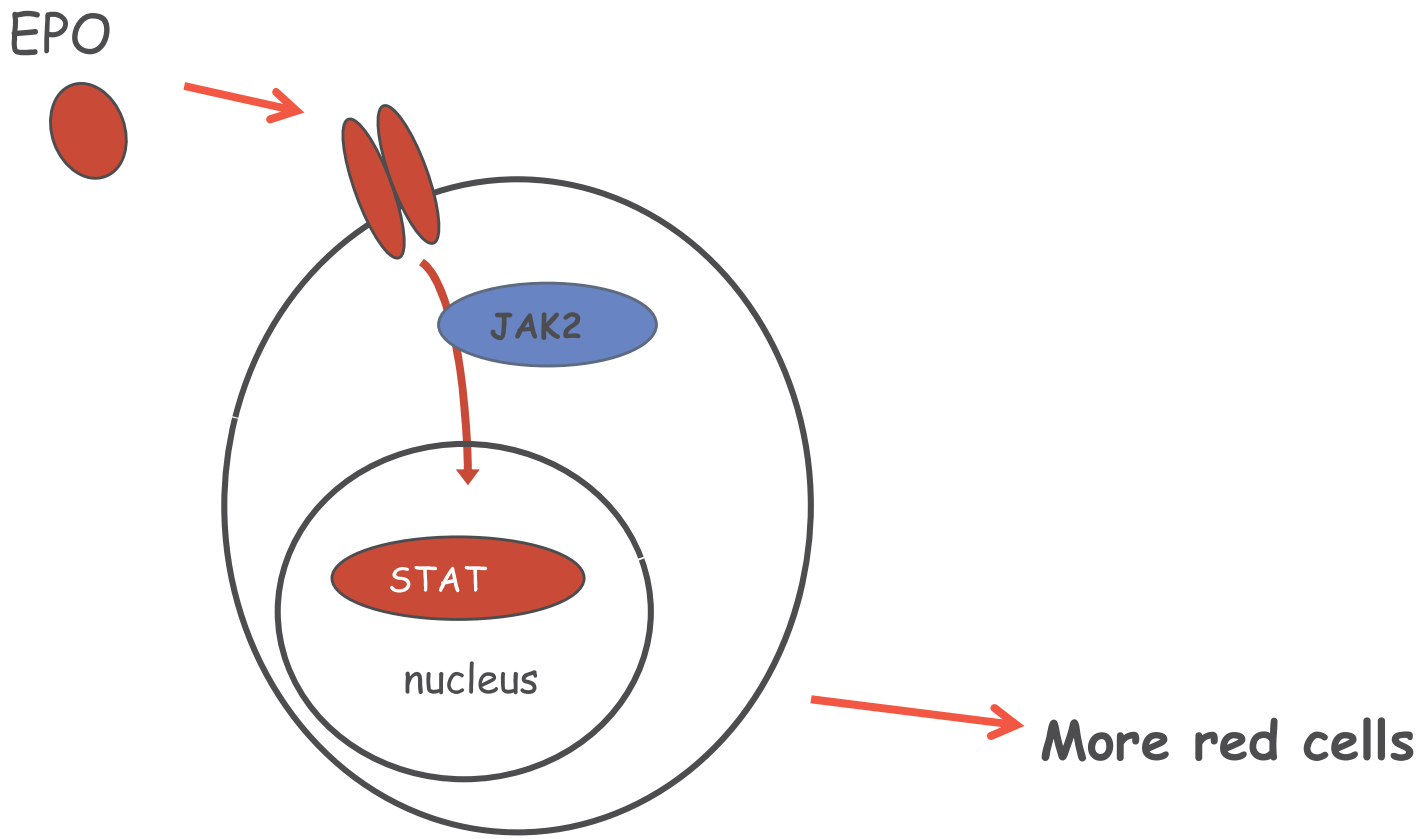
Tjio and Levan 1955



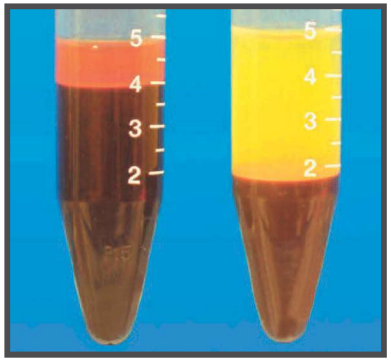
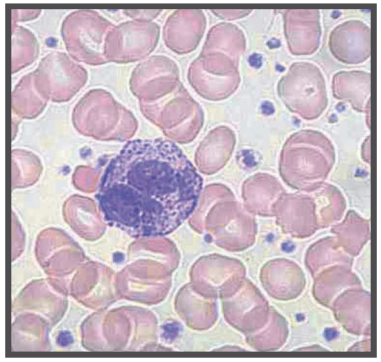
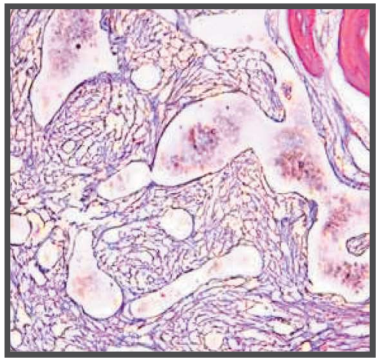
A change in the gene *JAK2*



“It all starts to make sense”



JAK2 mutations in MPN

PV	ET	MF
		
JAK2: 98%	50-60%	50-60%

2005 → 2010

Identification of JAK2 mutation

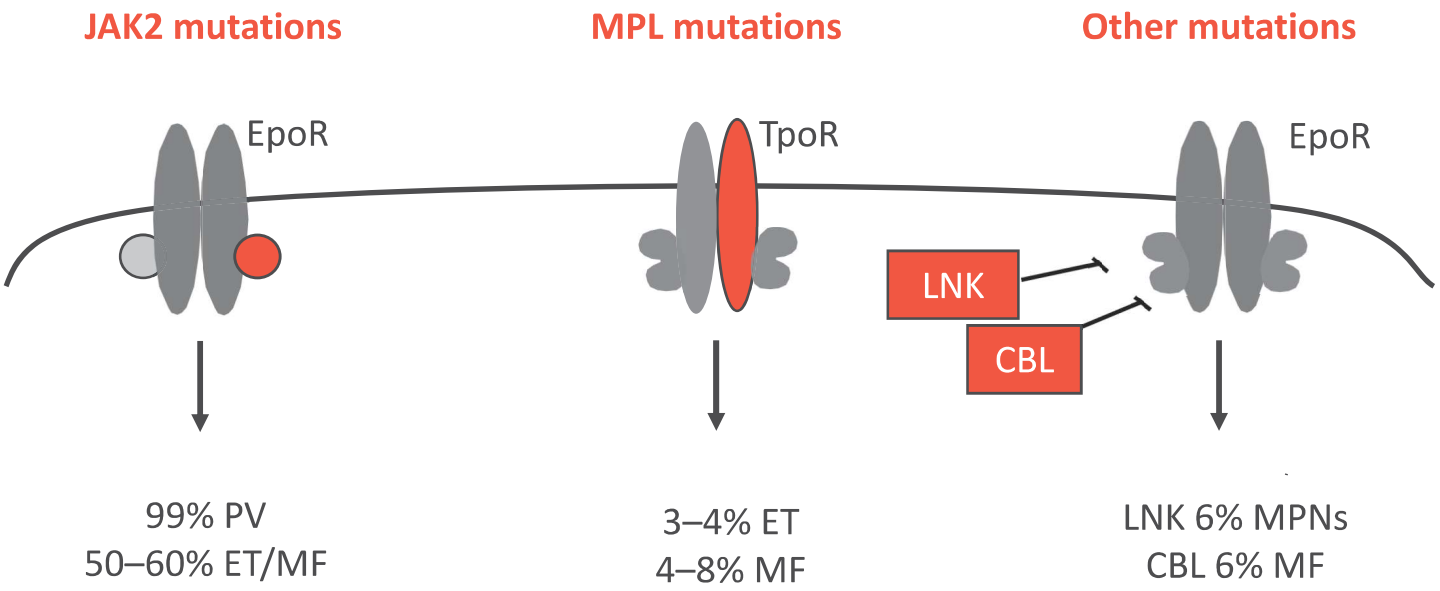
Recognition of new disease subtypes

Molecular testing in hospitals

Therapeutic JAK2 inhibitors

James et al, 2005; Baxter et al, 2005; Kralovics et al, 2005; Levine et al, 2005

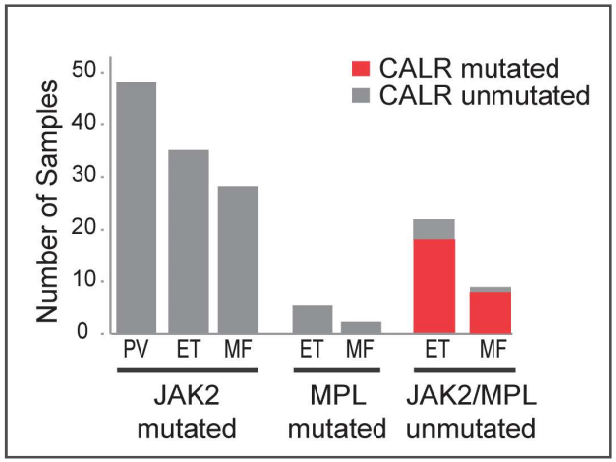
The theme repeated… increased growth signal to cells

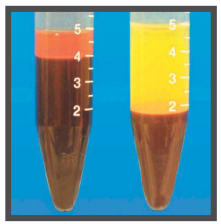


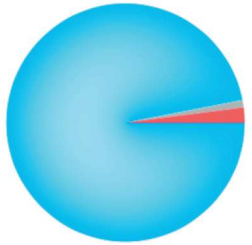
? Pathogenic mechanism of 50% of ET and MF

James et al 2005; Levine et al 2005; Baxter et al 2005; Kralovics R, et al. *Blood* 2005;106:3374–3376; Scott et al 2007; Pikman Y, et al. *PLoS Med* 2006;3:e270; Beer et al 2008; Boyd et al 2010; Oh et al 2010; Lasho TL, et al. *N Engl J Med* 2010;363:1189–1190; Pardanani A, et al. *Leukemia* 2010;25:218–225; Sanada et al 2009; Grand et al 2009

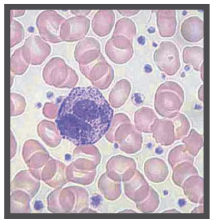
CALR mutations in majority of *JAK2*-unmutated MPNs

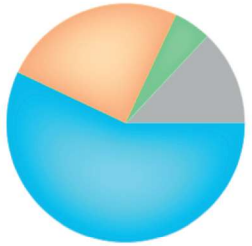




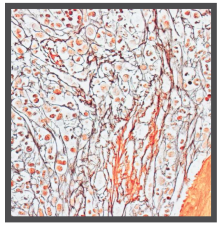


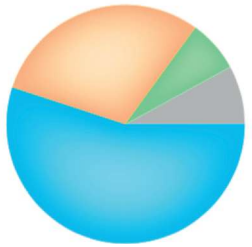
PV





ET





MF

CALR

JAK2 exon 12

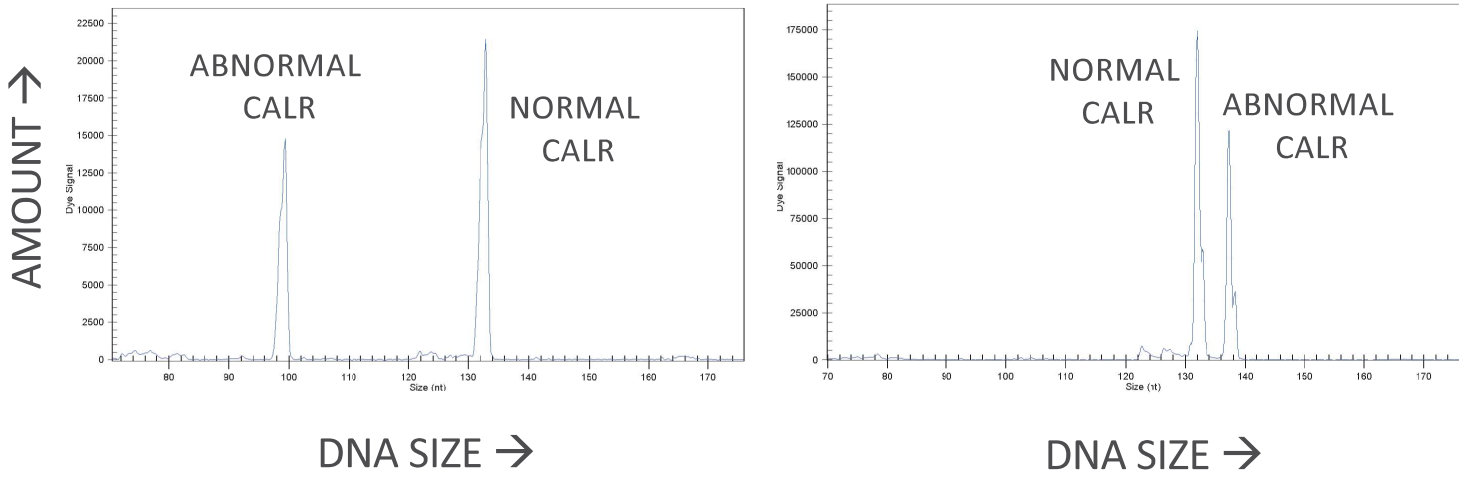
$JAK2^{V617F}$

JAK2/MPL/CALR unmutated

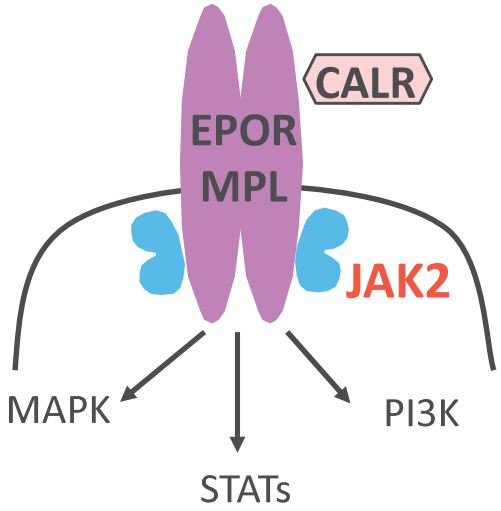
MPL

Nangalia, Massie et al NEJM 2013; Klampfl et al NEJM 2013

A new test for the clinic and patients….



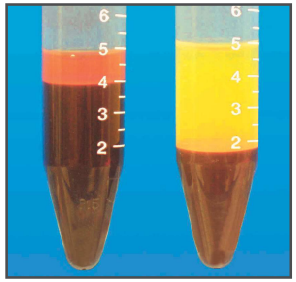
Changes in the JAK2, CALR and MPL genes *drive* MPNs….

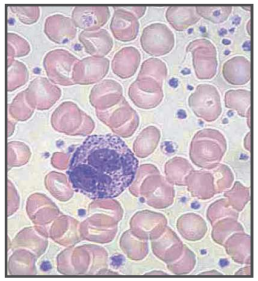


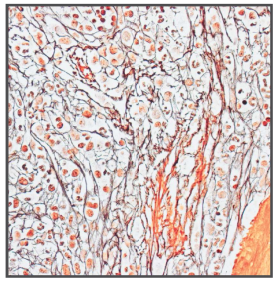
Polycythaemia Vera (PV)

Essential thrombocythaemia (ET)

Myelofibrosis (MF)

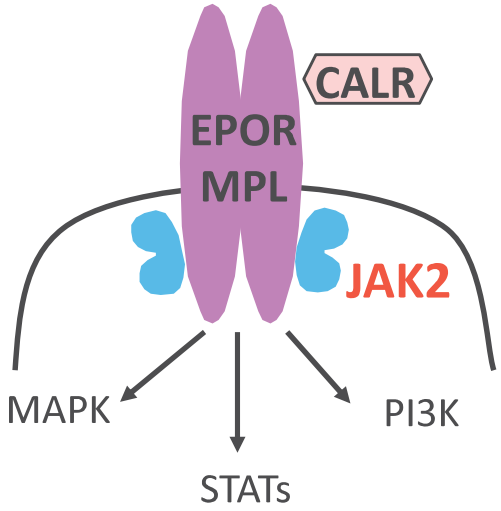






- >90% patients have mutations in *JAK/CALR/MPL*

Additional mutations in other genes also found….



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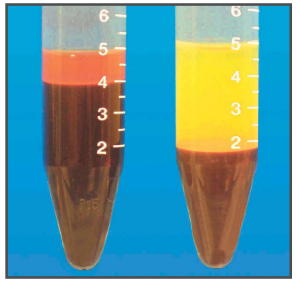

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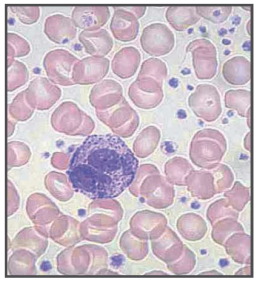
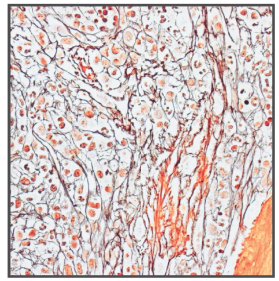
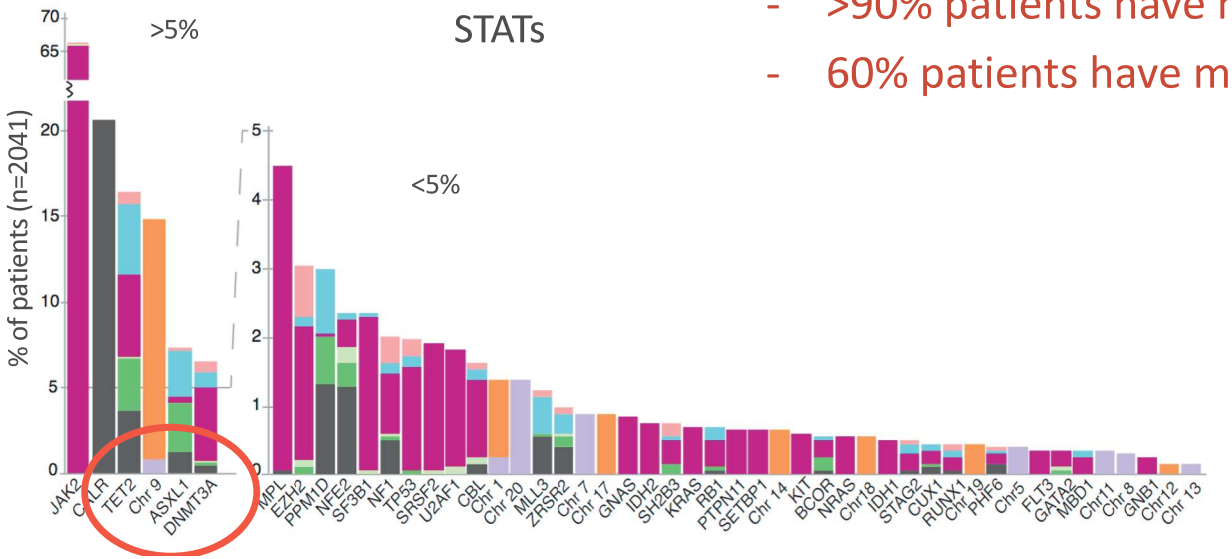


Myelofibrosis (MF)



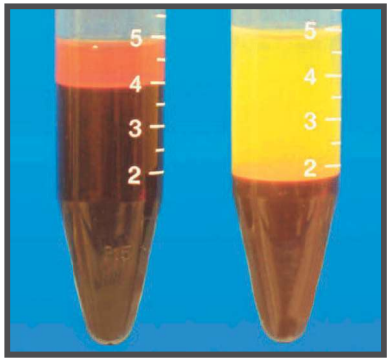
- >90% patients have mutations in *JAK/CALR/MPL*
- 60% patients have mutations in additional genes



Nangalia et al, NEJM 2013

Myeloproliferative neoplasms

PV



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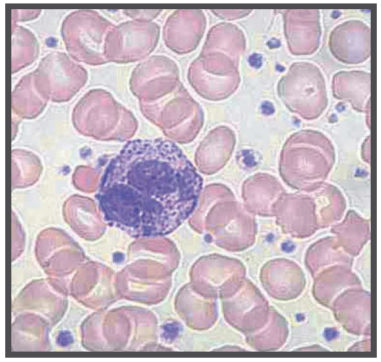

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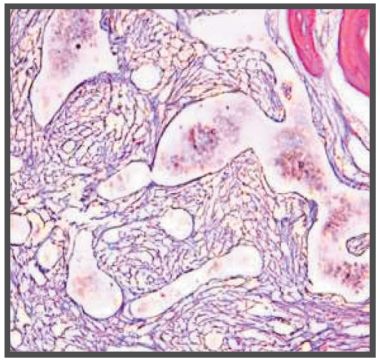

- *What causes it?*
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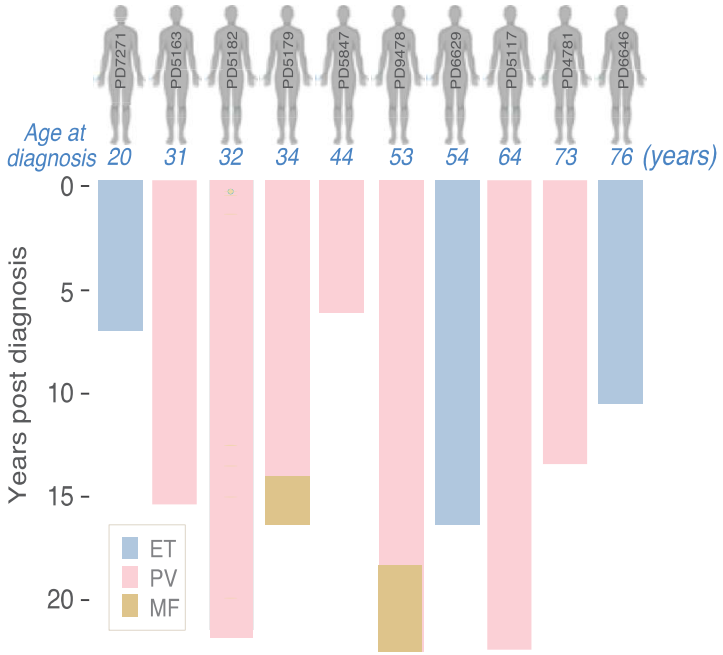
Patient cohort and experimental design



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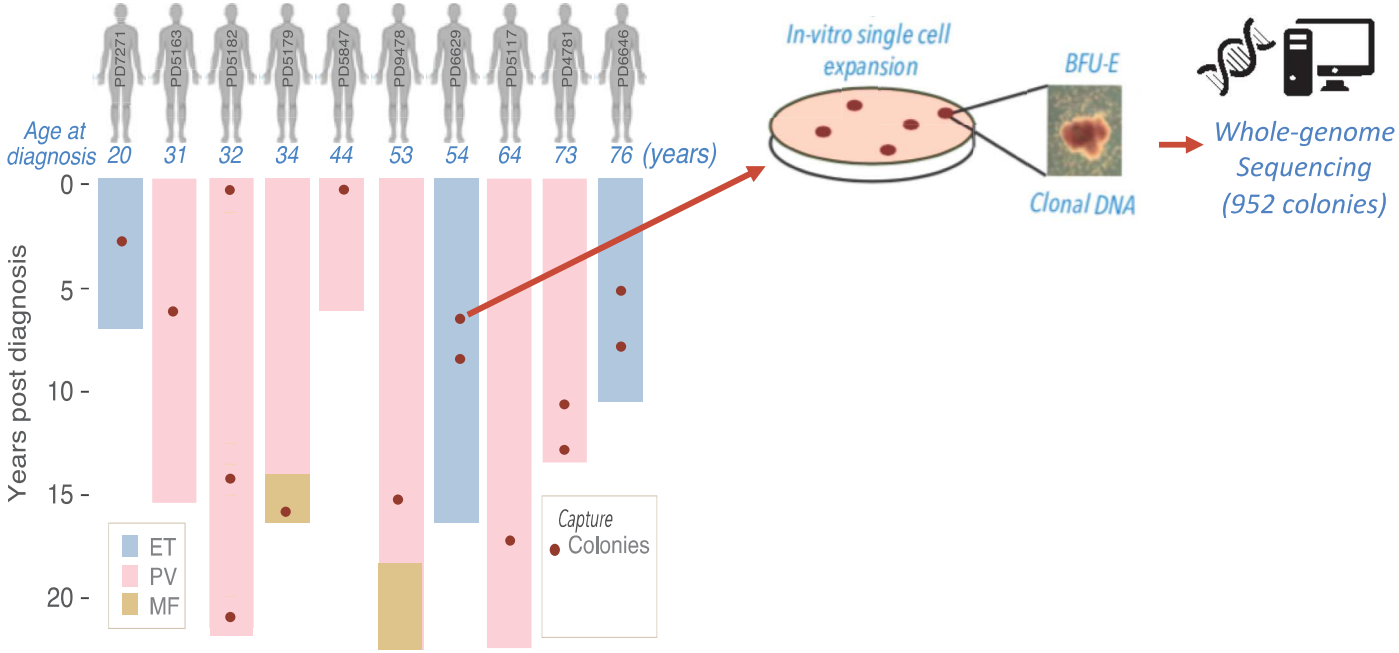

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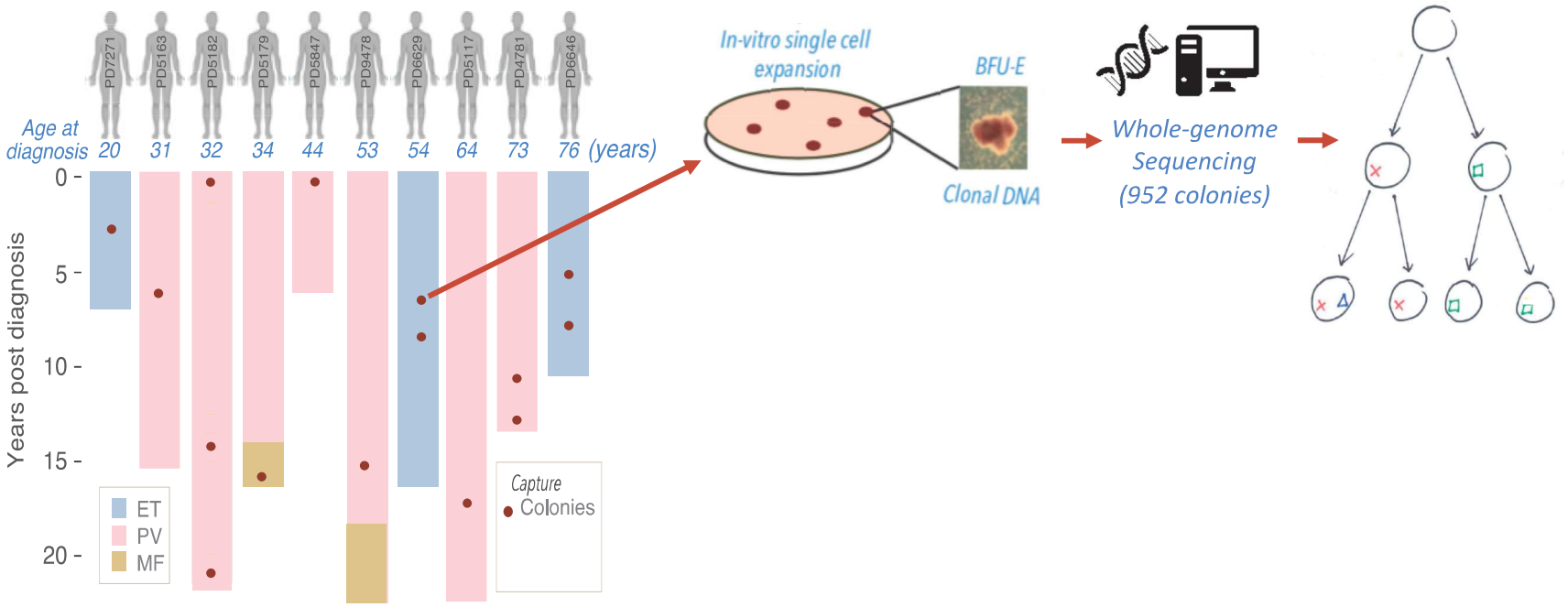

- Starting from the zygote, all cells are acquiring mutations
- Mutations in individual cells act as a natural barcodes

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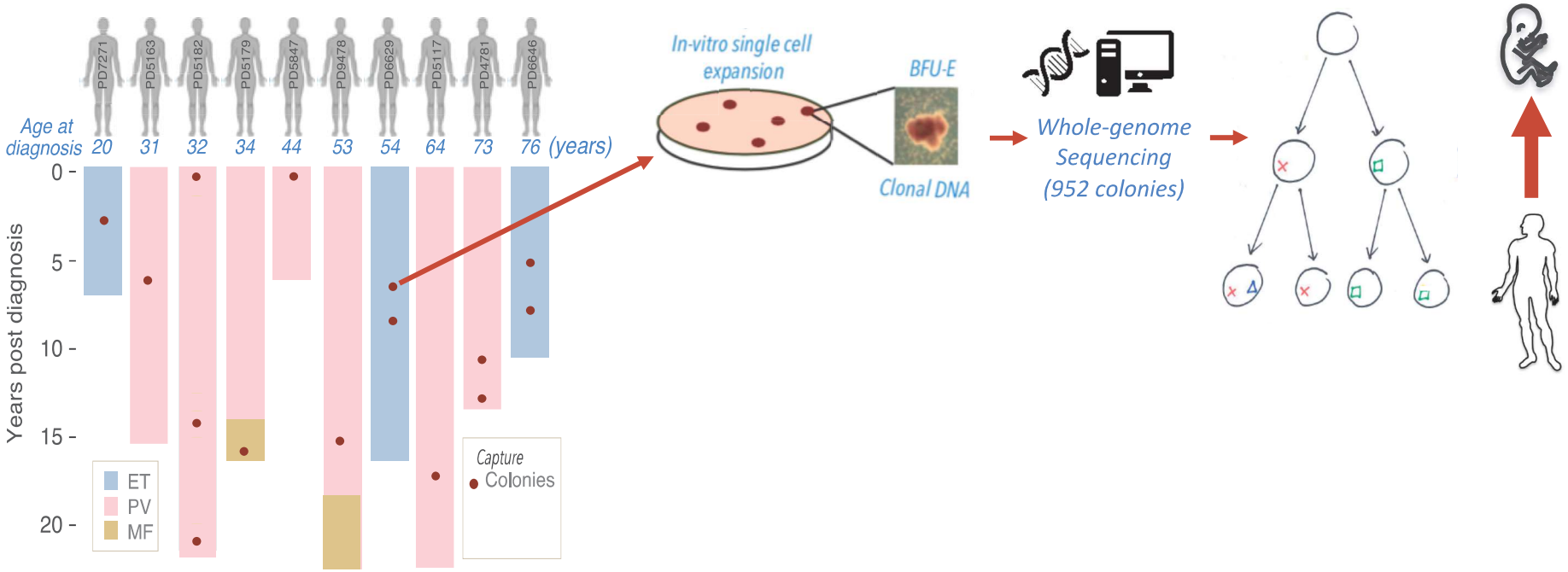
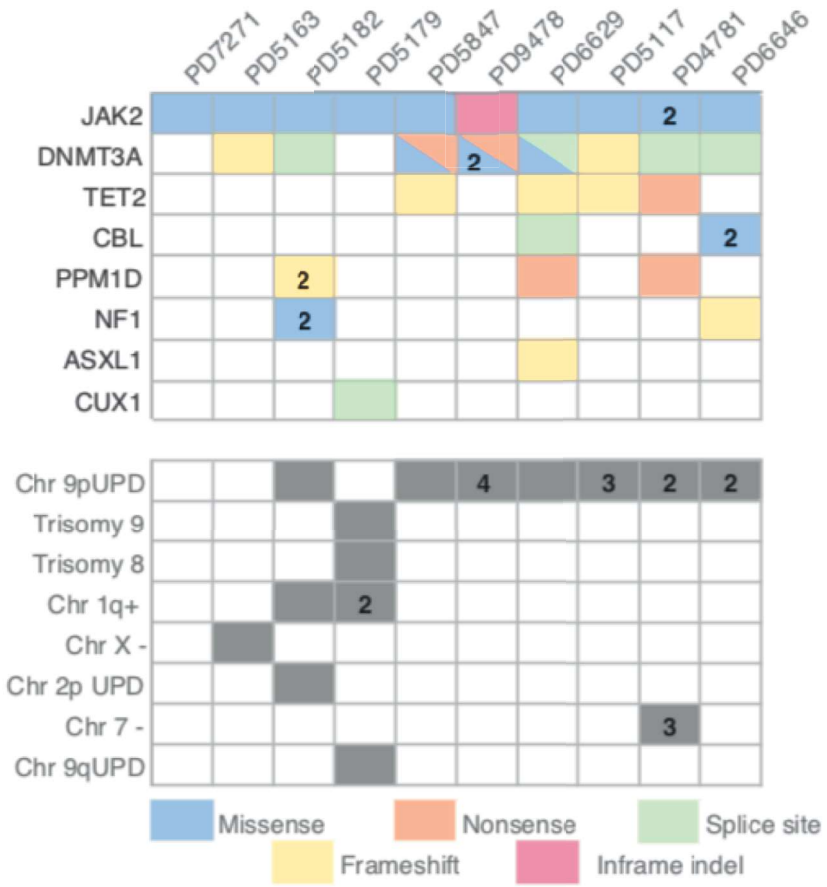
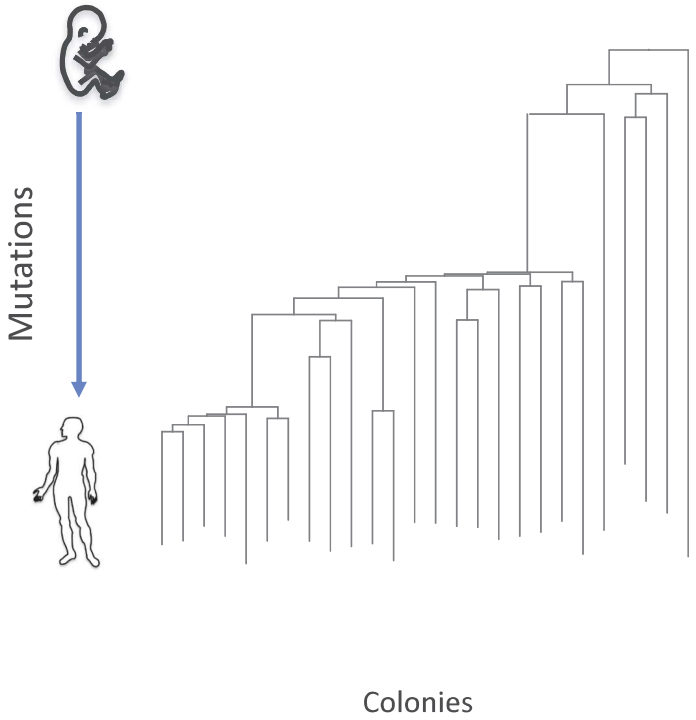


- Starting from the zygote, all cells are acquiring mutations
- Mutations in individual cells act as a natural barcodes
- Mutations can trace family relationships back to start of life

Rich mutational landscape at the clonal level



Using somatic mutations to build a phylogenetic “tree”



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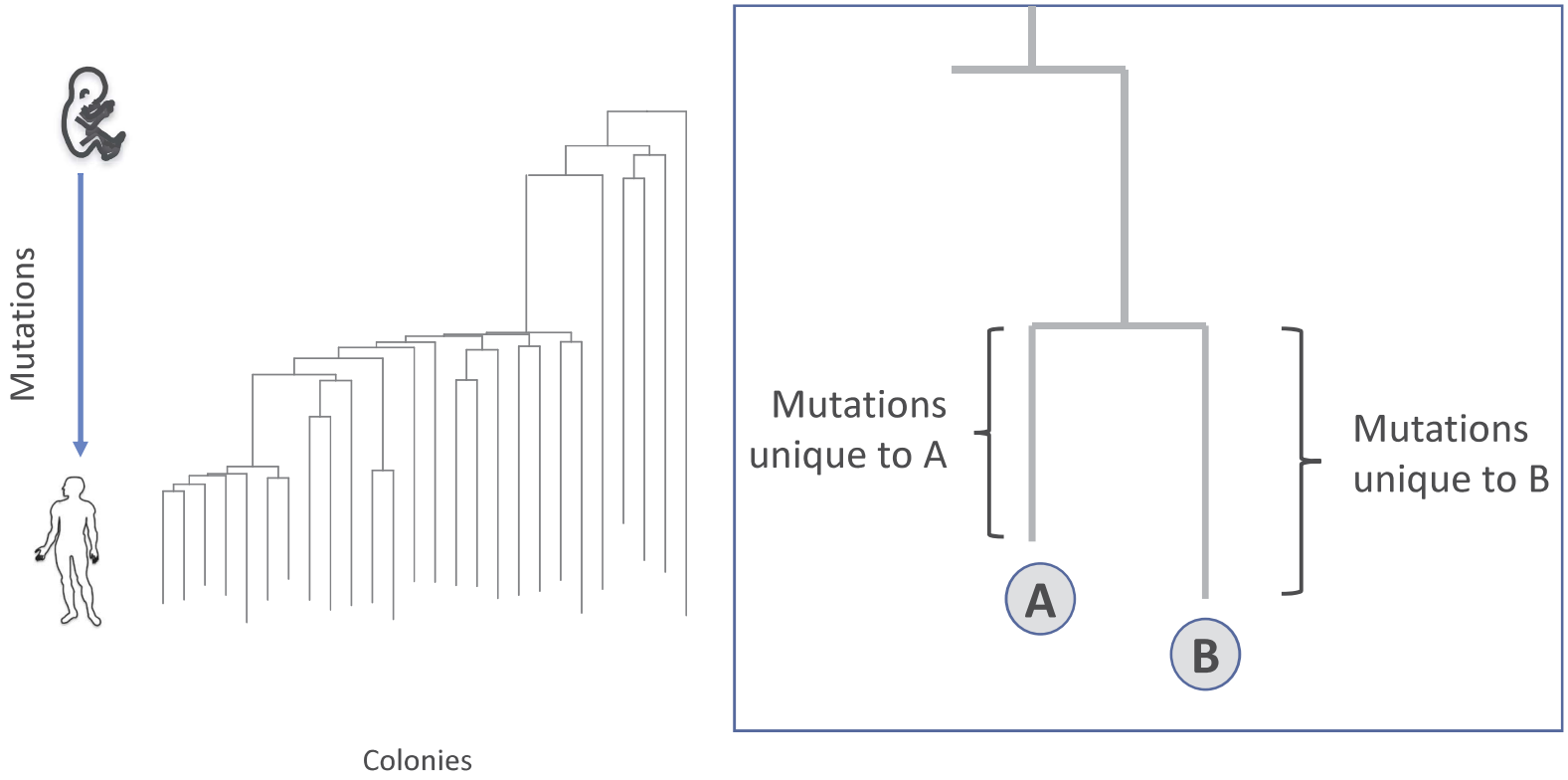

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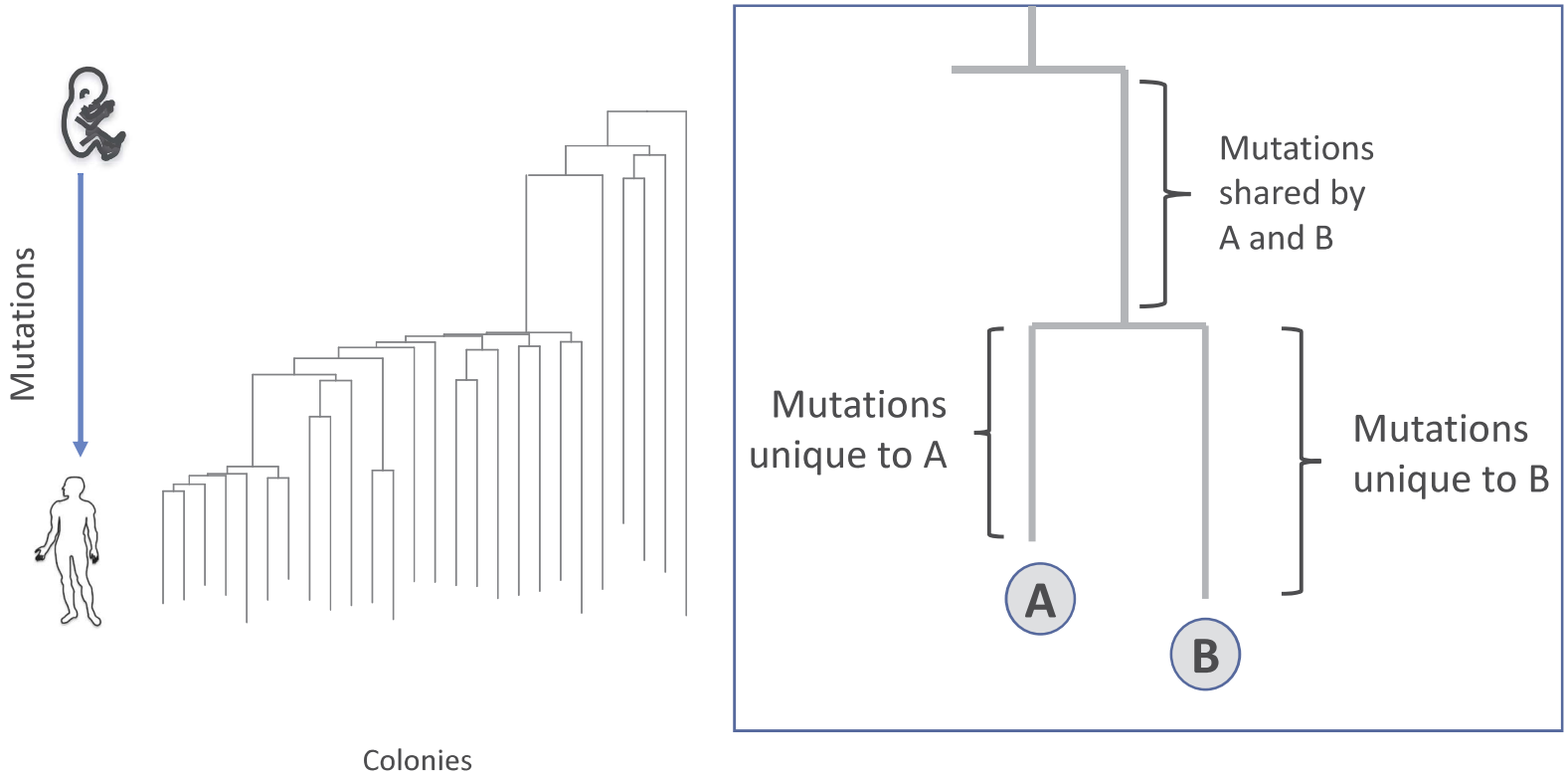
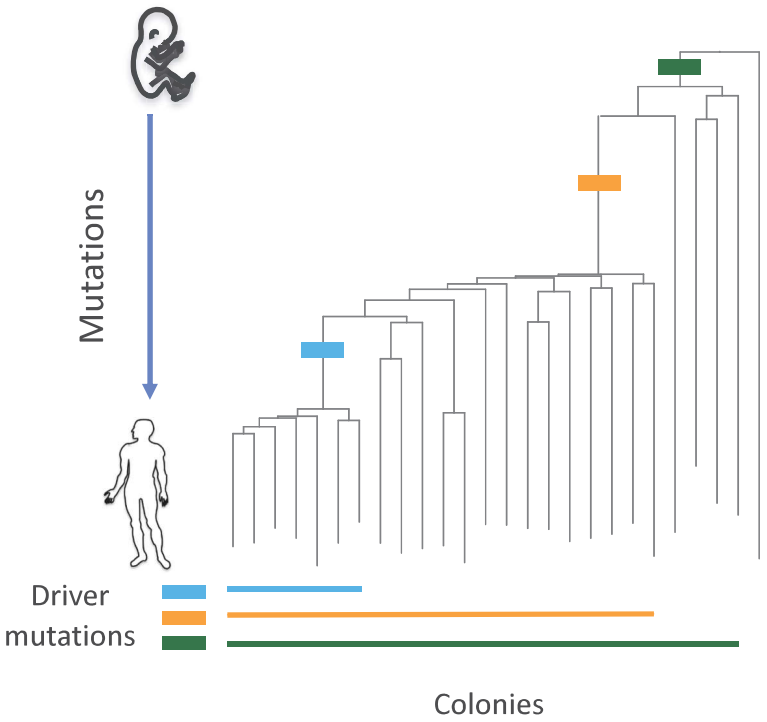
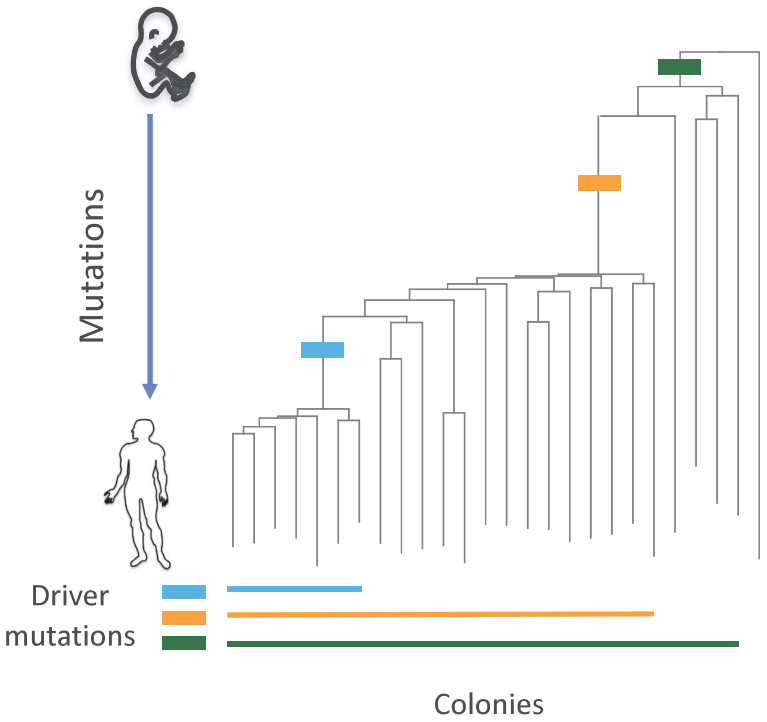
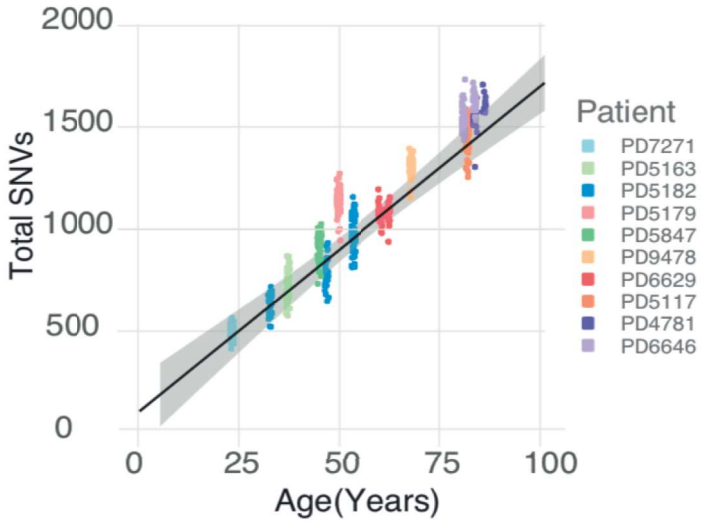


Putting driver mutations on “branches” of the “tree”



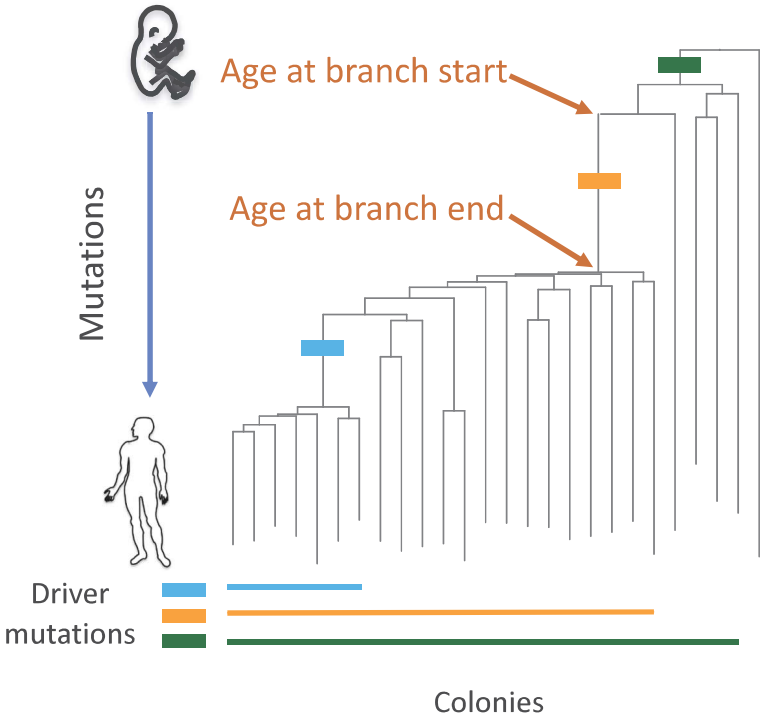
A molecular clock to *time* the 'branches' of the tree

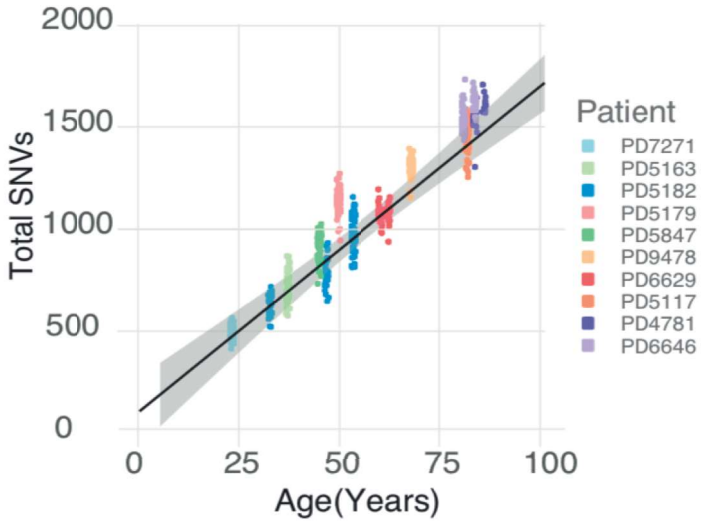




~18 mutations per year

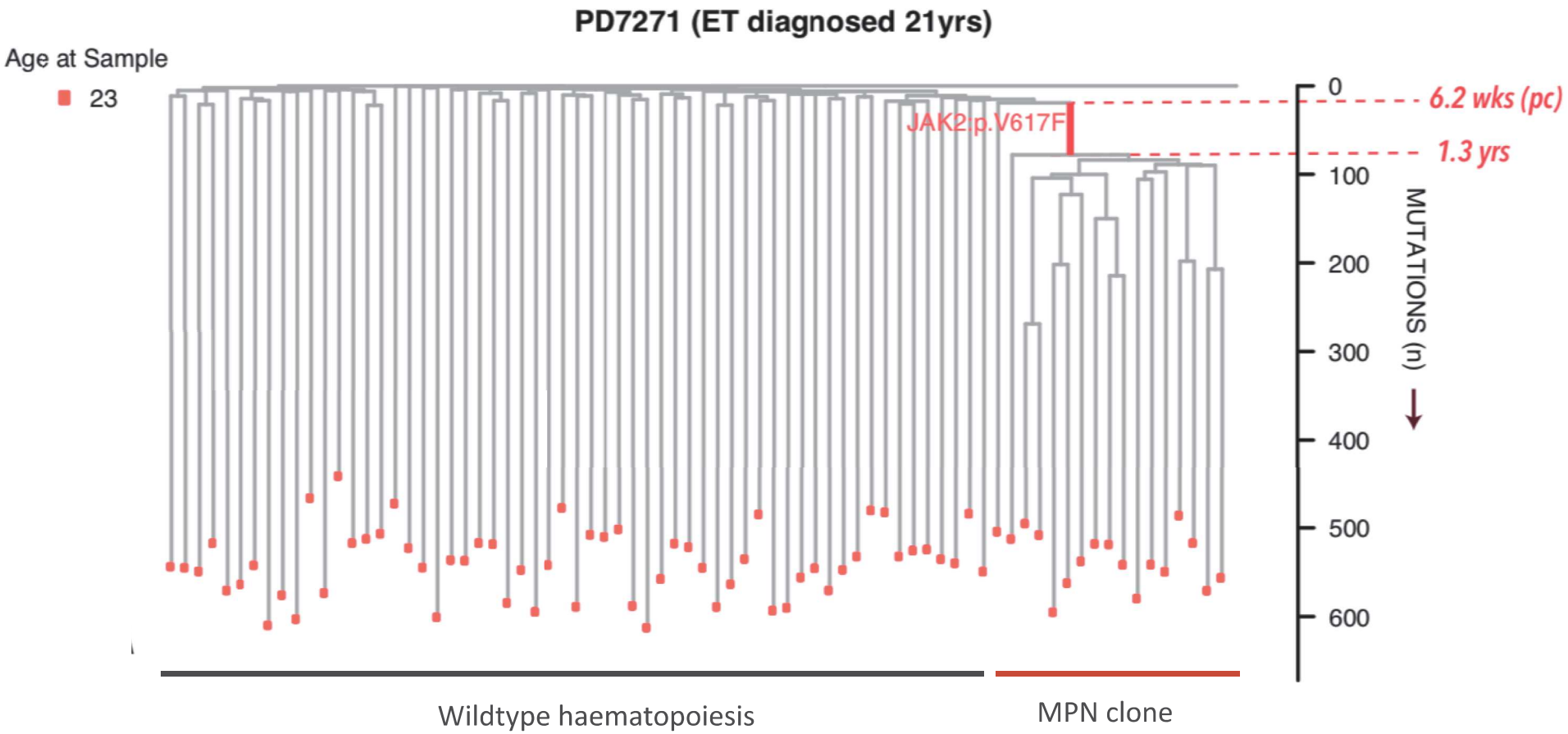
A molecular clock to *time* the 'branches' of the tree





~18 mutations per year

JAK2^{V617F} is acquired in early life in MPN



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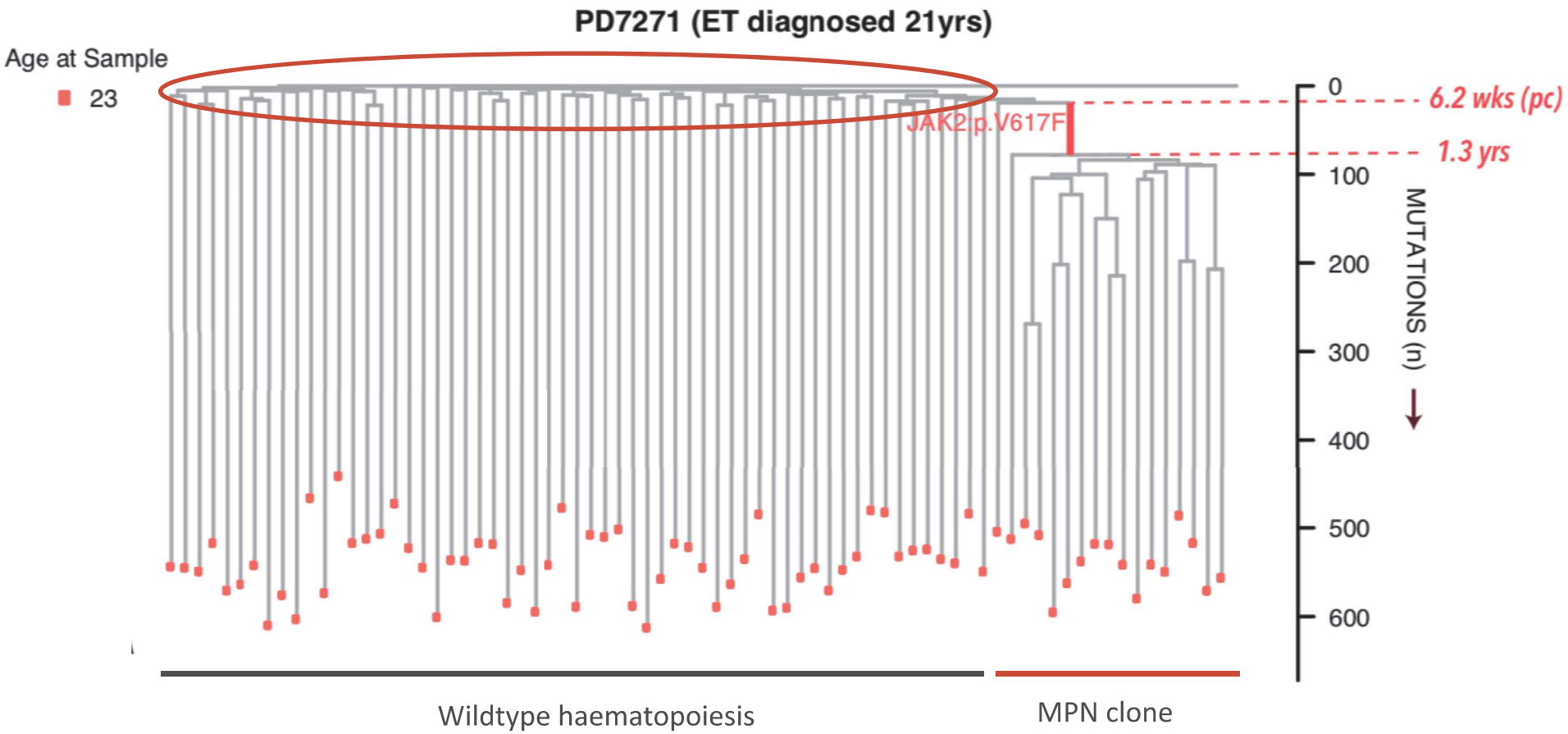

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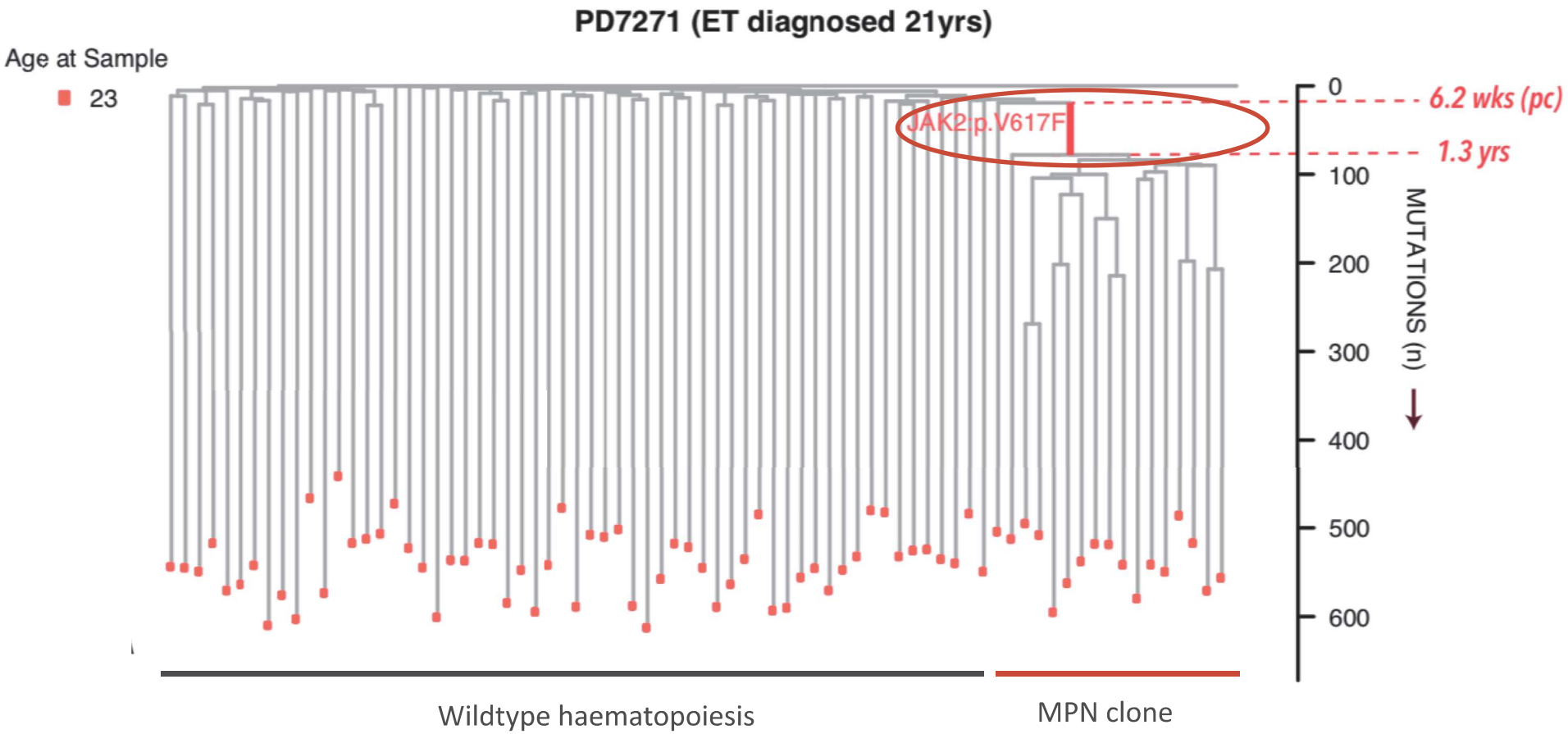

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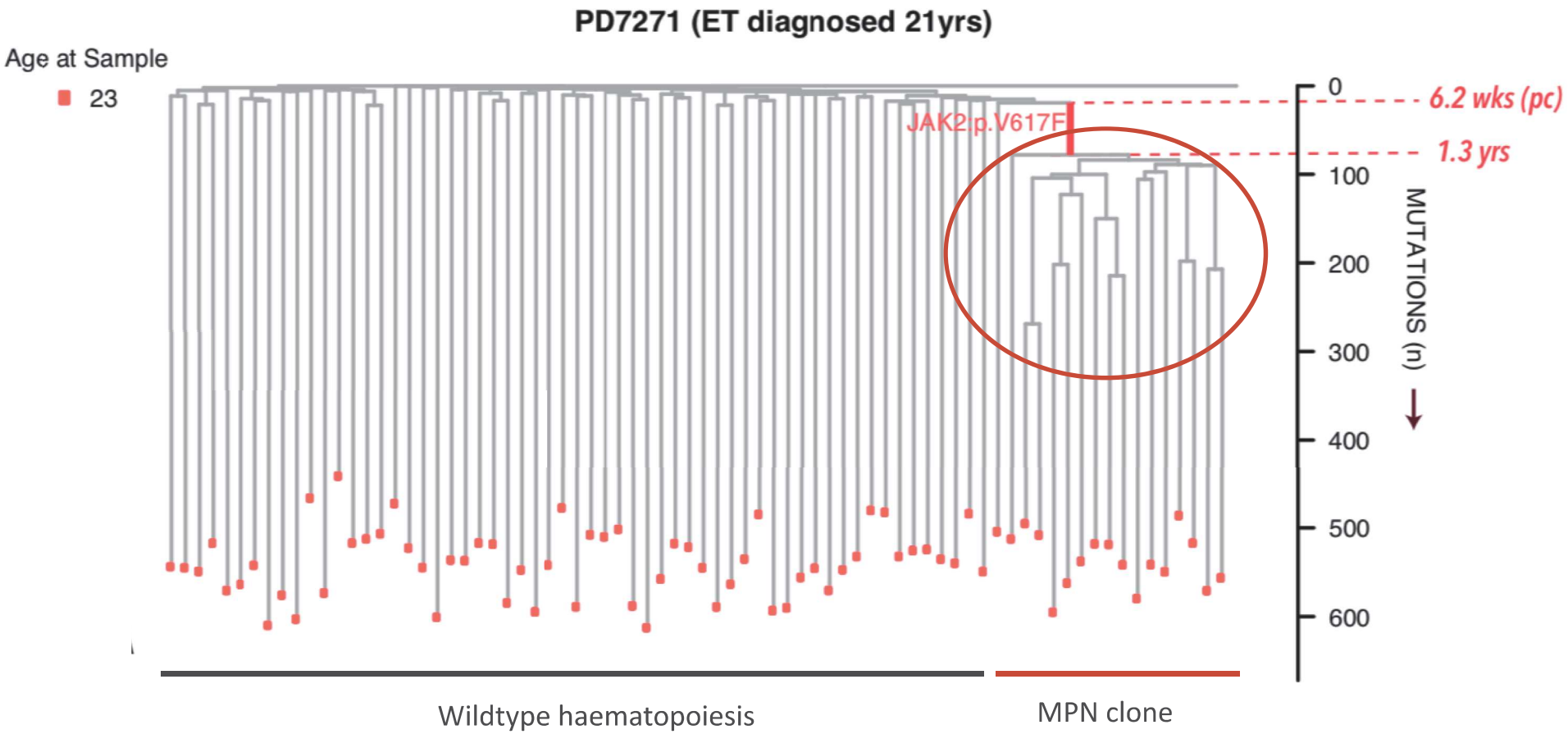
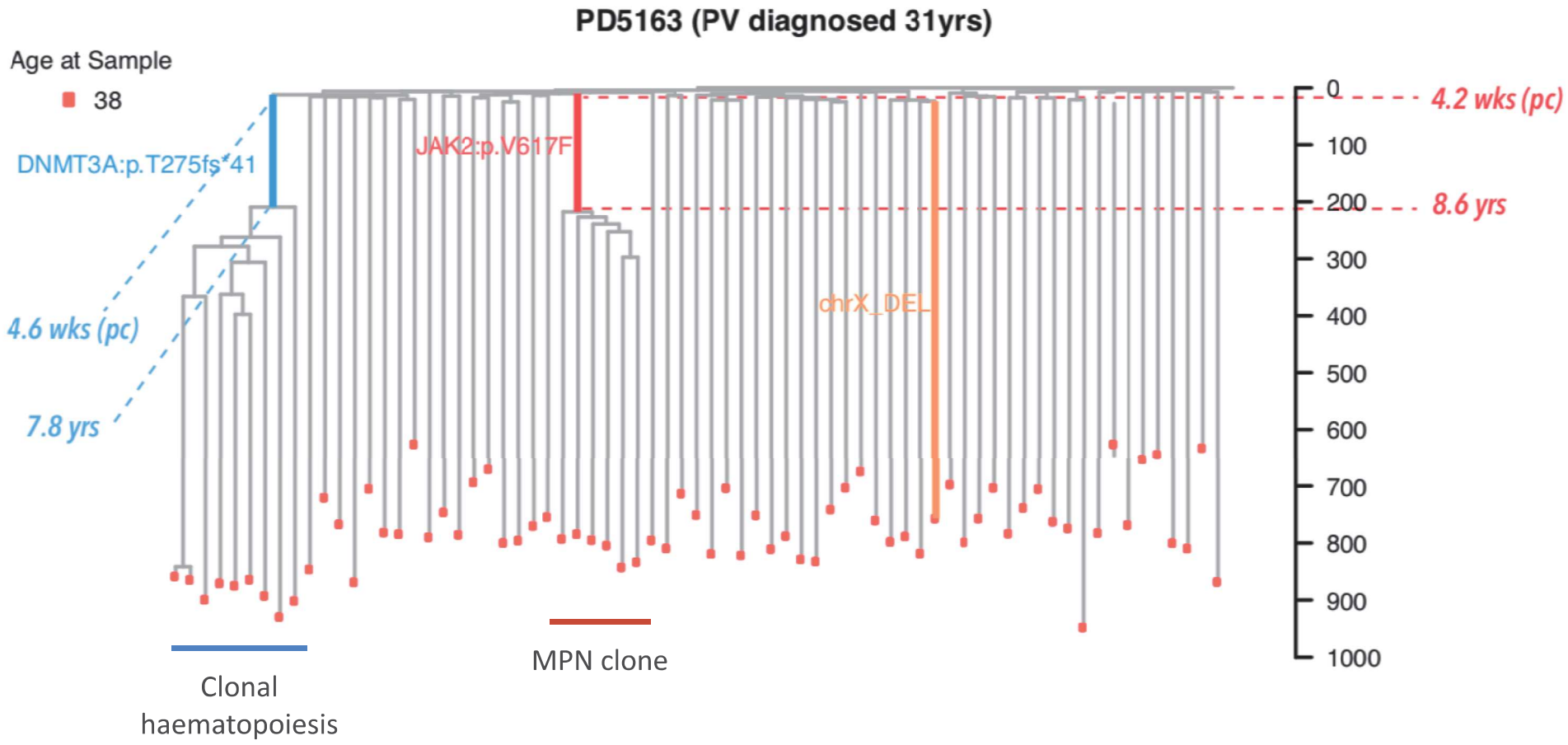
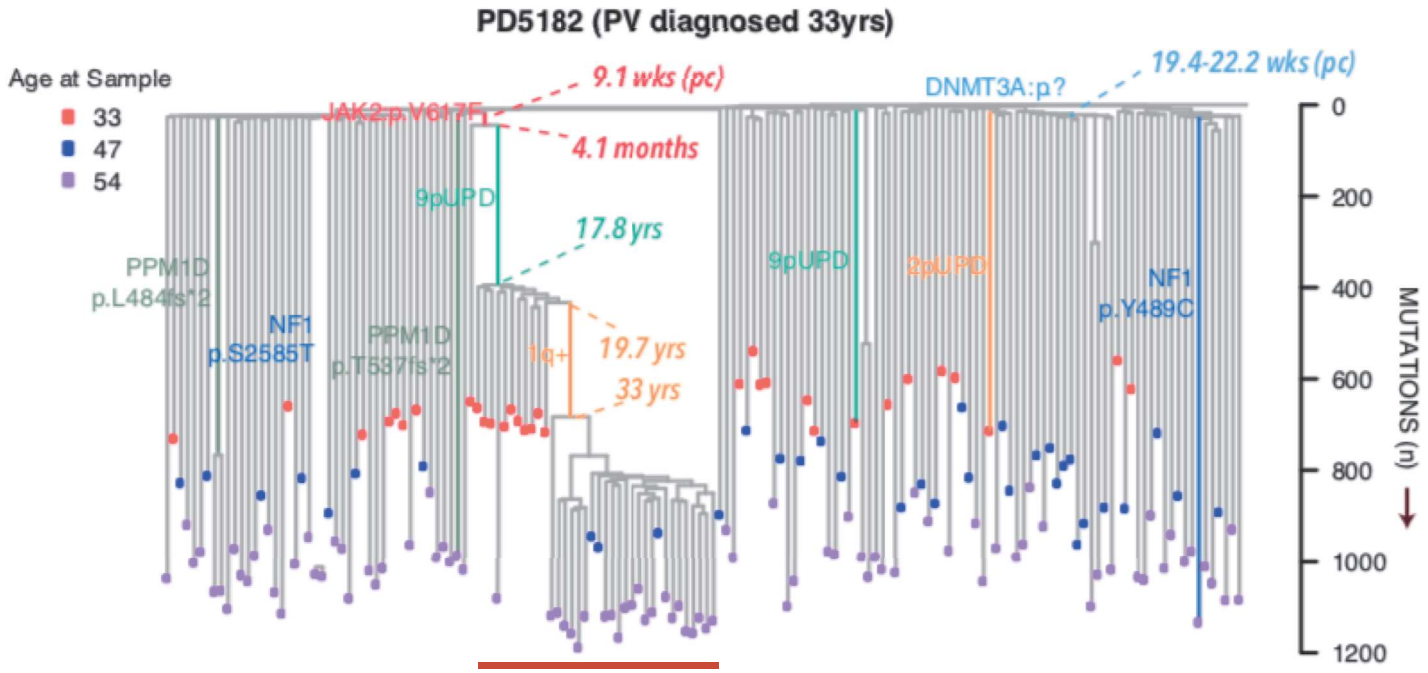


DNMT3A – clonal haematopoiesis acquired early in life



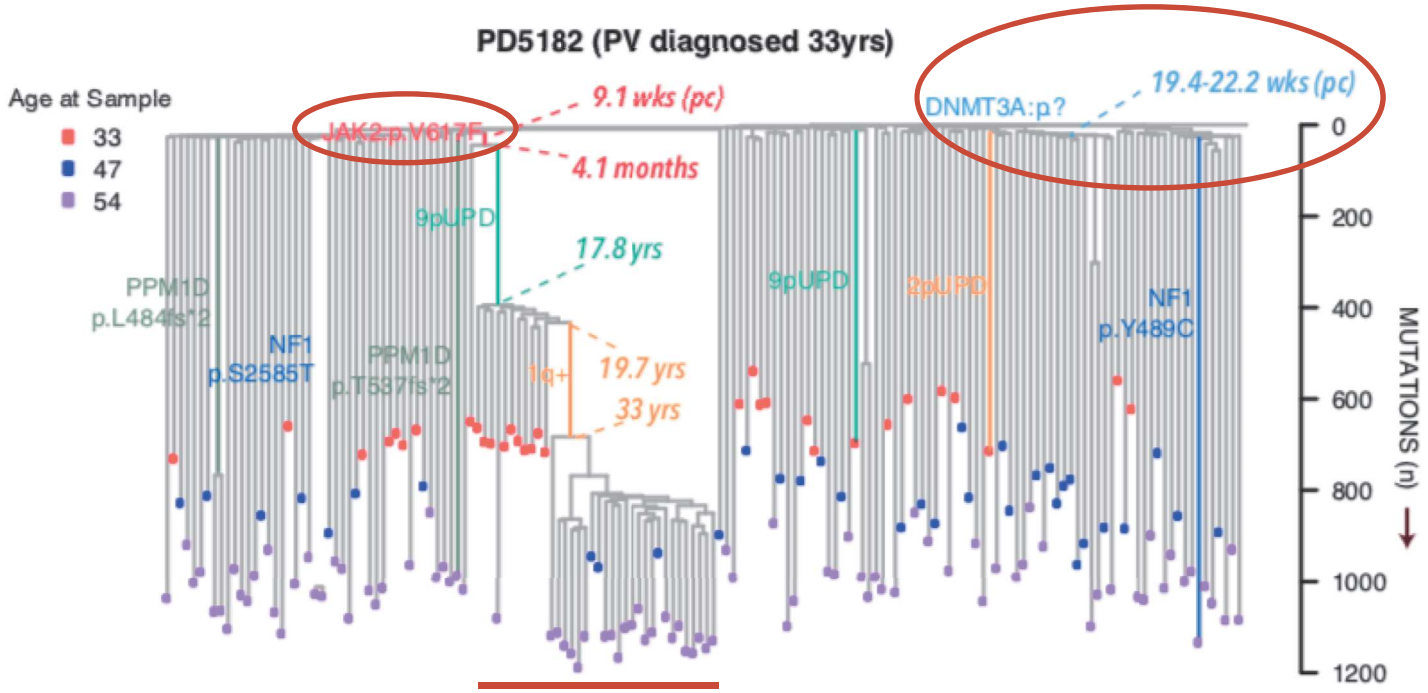
JAK2 and *DNMT3A* mutations can be acquired *in utero*



MPN clone

- Rich driver mutation landscape

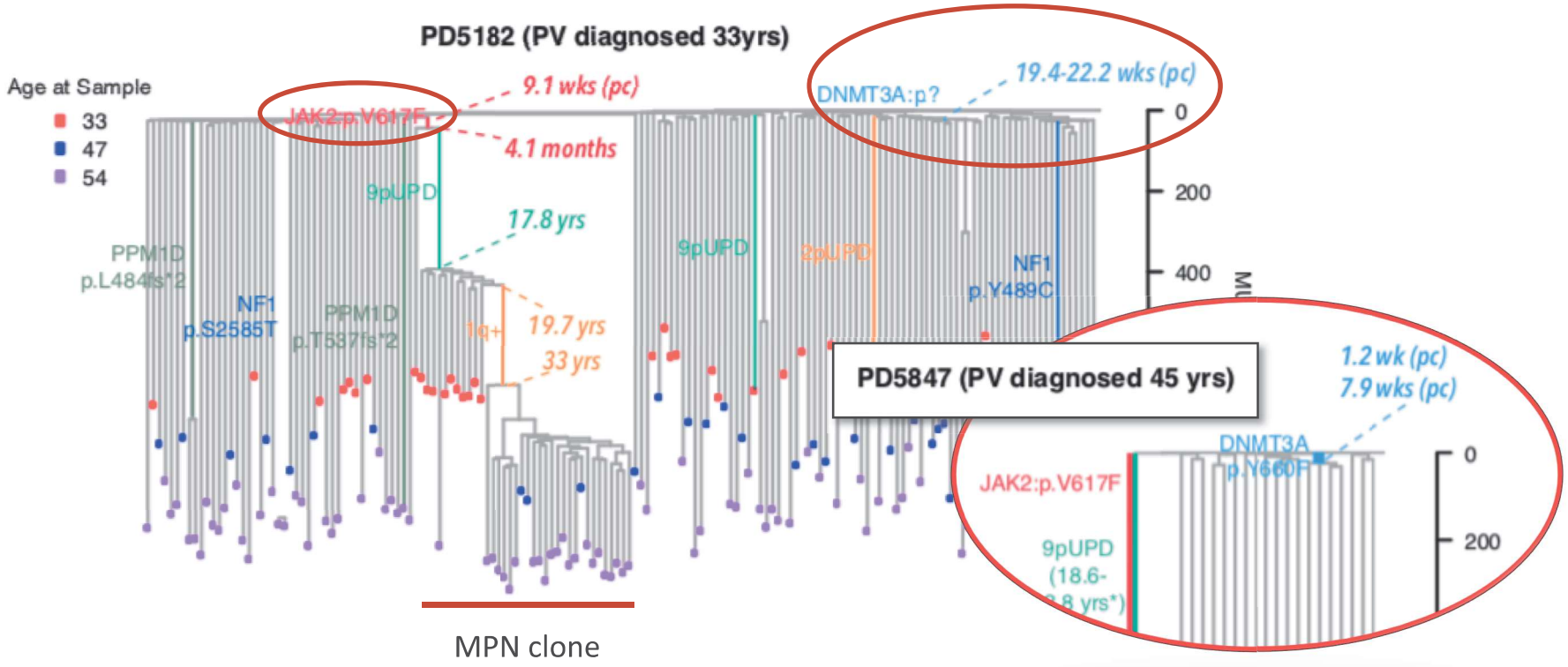
JAK2 and *DNMT3A* mutations can be acquired *in utero*



MPN clone

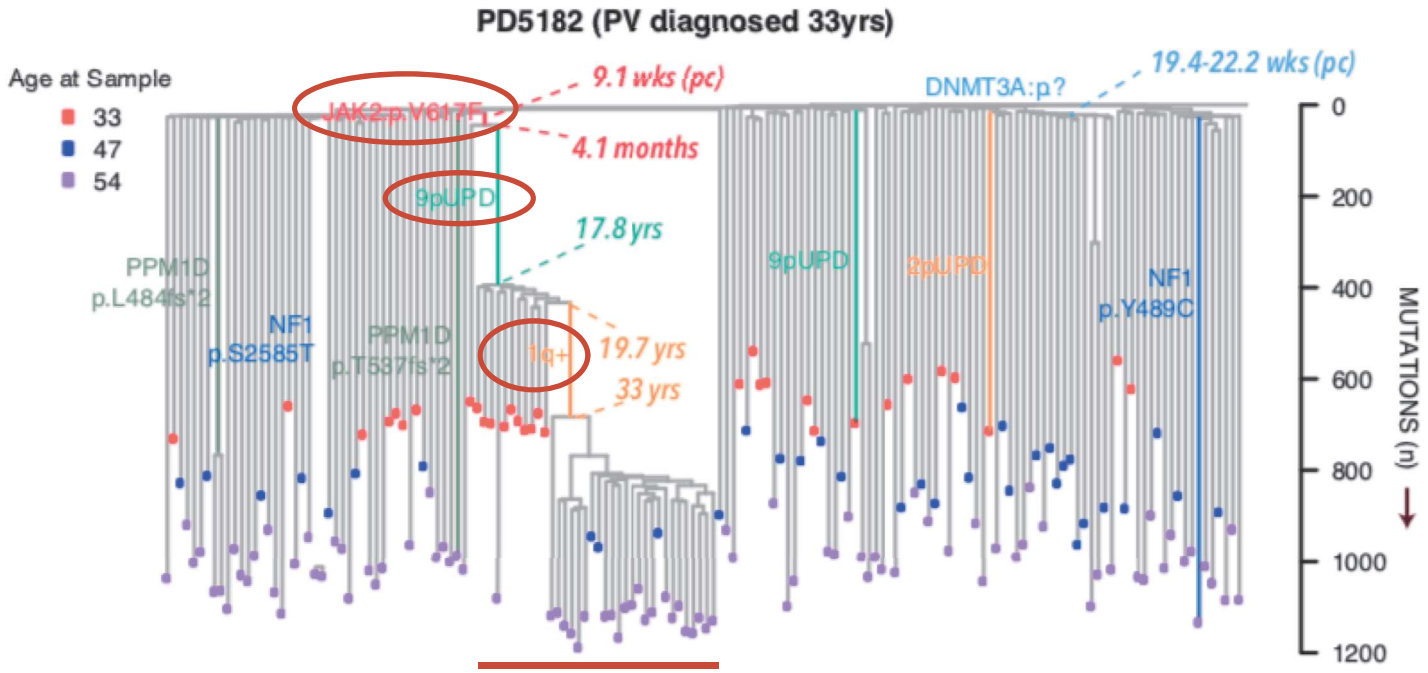
- Rich driver mutation landscape
- *JAK2* and *DNMT3A* mutations acquired very early in life – including *in utero*

JAK2 and *DNMT3A* mutations can be acquired *in utero*



- Rich driver mutation landscape
- *JAK2* and *DNMT3A* mutations acquired very early in life – including *in utero*

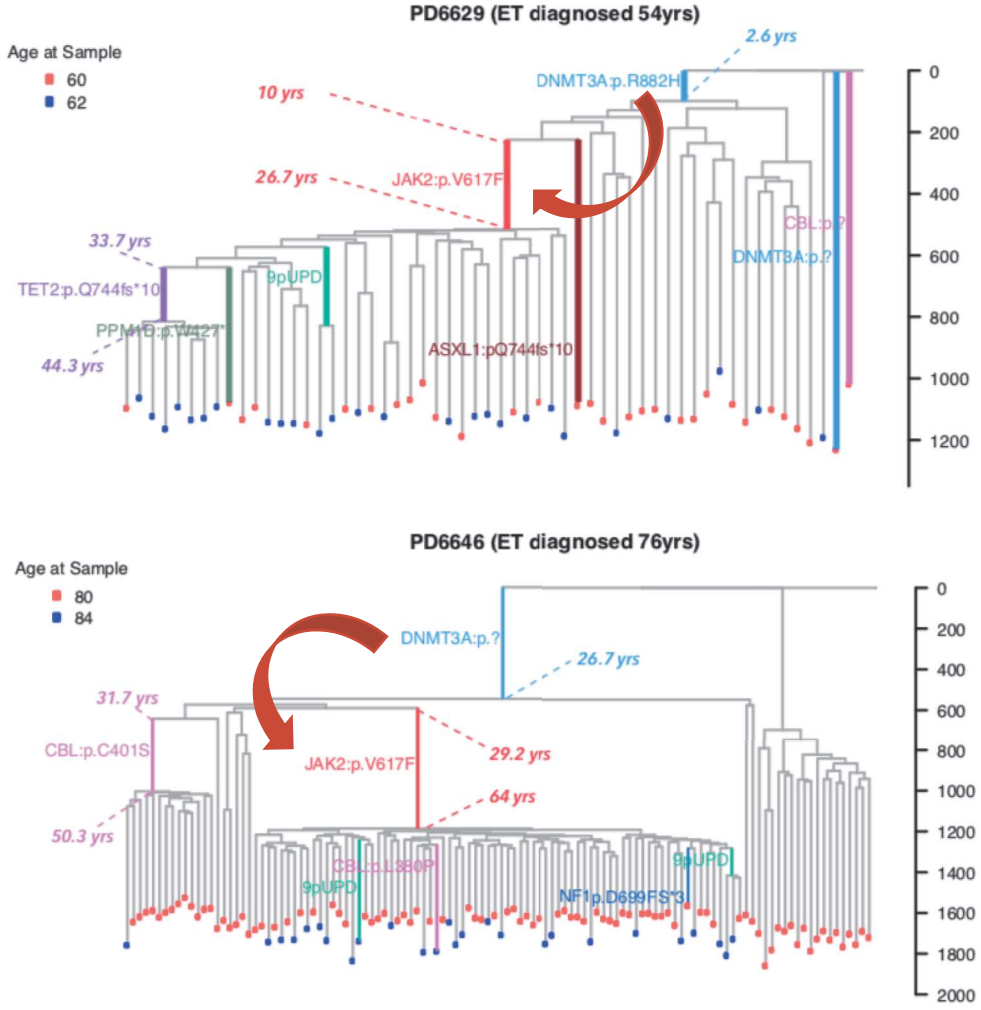
JAK2 and *DNMT3A* mutations can be acquired *in utero*



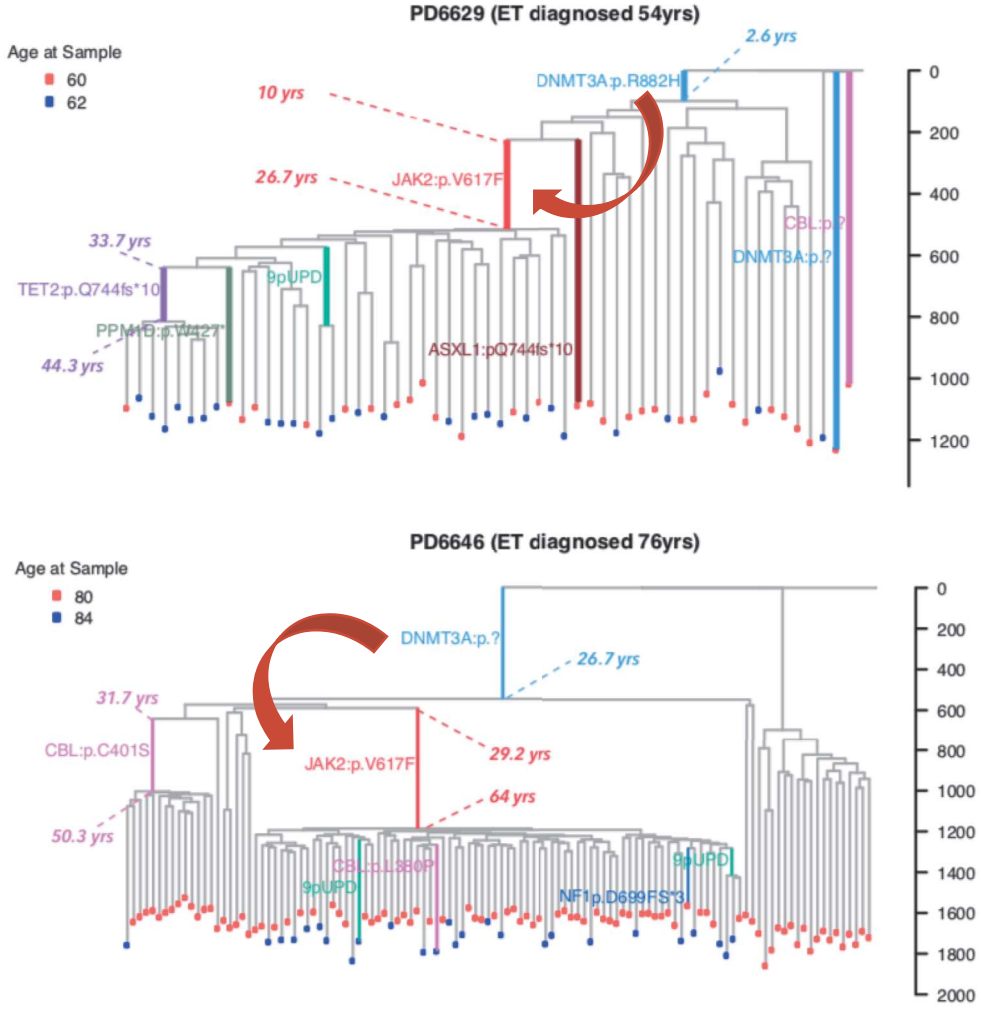
MPN clone

- Rich driver mutation landscape
- *JAK2* and *DNMT3A* mutations acquired very early in life – including *in utero*
- Additional driver mutation acquisitions separated by decades

JAK2^{V617F} acquired "second"

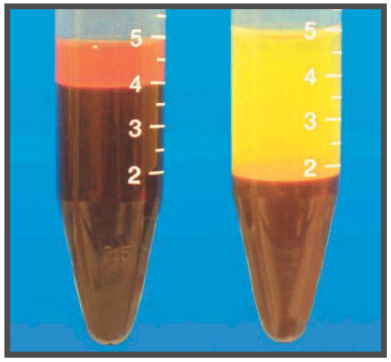


JAK2^{V617F} acquired "second"



Myeloproliferative neoplasms

PV



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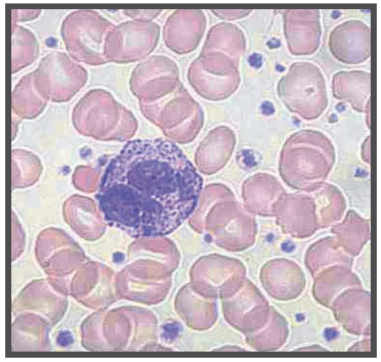
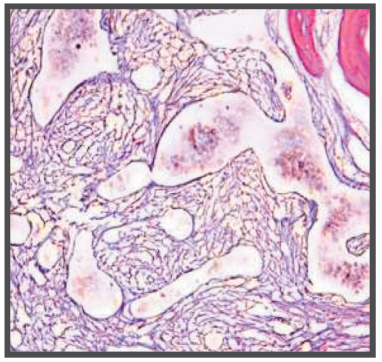
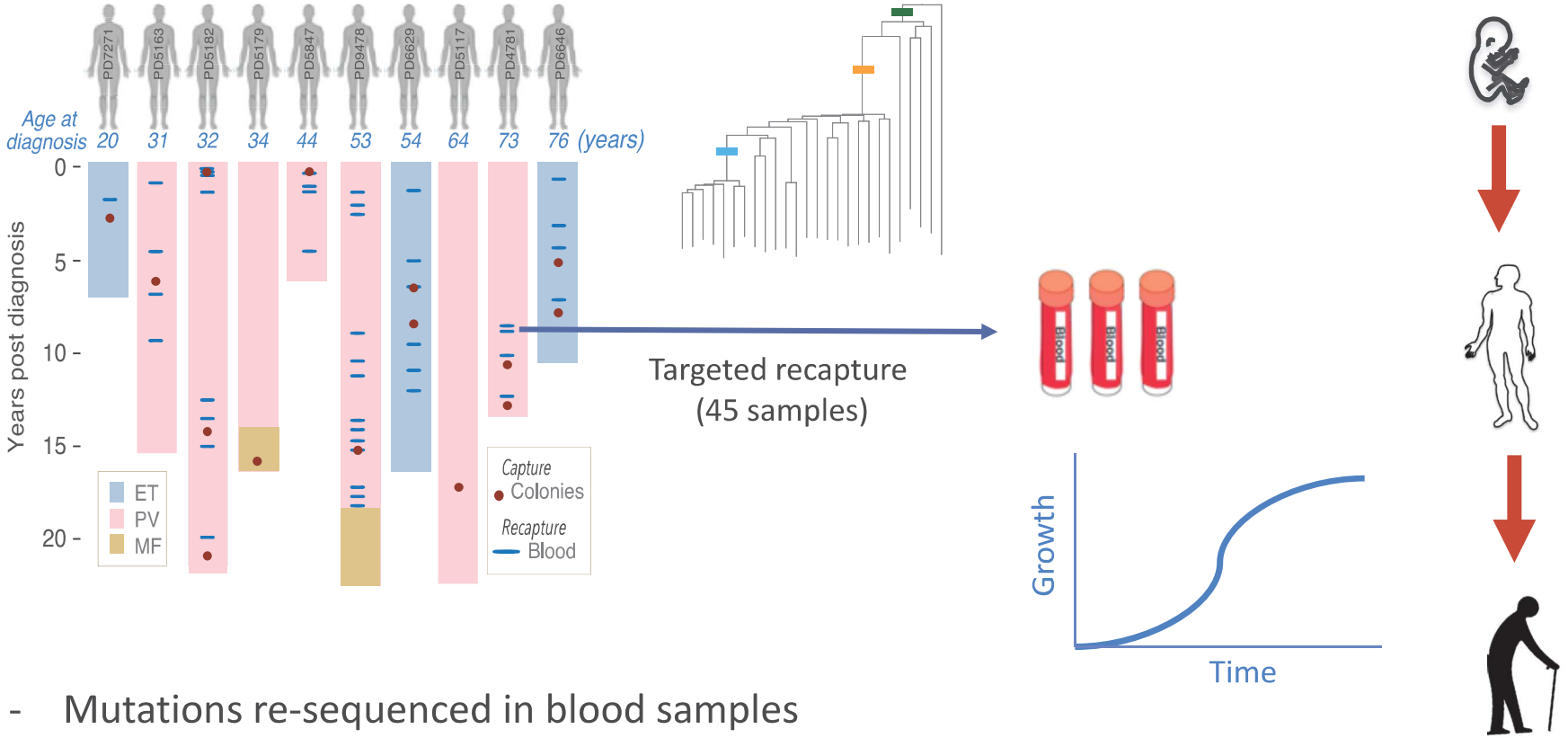


MF



- *What causes it?*
- *How long have I had it for?*
- ***How fast did it grow?***

Estimating growth rates of the clones



- Mutations re-sequenced in blood samples
- Pattern of branching in the tree + mutation fractions in blood inform growth rates of mutant clones

Measures of growth rates (*fitness*) of clones

Patient	Clade	% additional growth per yr	95% CI
PD5847	*JAK2*^{V617F}, *9pUPD*, *TET2*^{N281fs*1}	**233**	143-360
PD5182	*JAK2*^{V617F}, 9pUPD	**142**	75-279
PD6646	*DNMT3A*$^{p?}$, *JAK2*^{V617F}	**128**	92-266
PD4781	*JAK2*^{V617F}, 9pUPD, *TET2*^{Q1632*}, 7q-, 7p-	**119**	76-289
PD5179	*JAK2*^{V617F}, 1q+	**115**	86-170
PD6629	*DNMT3A*^{R882H}, *JAK2*^{V617F}, *TET2*^{Q744fs*10}	**83**	36-166
PD9478	*JAK2*$^{Exon\ 12}$, *DNMT3A*^{Y908*}	**71**	54-96
PD6646	*DNMT3A*$^{p?}$, *CBL*^{C401S}	**70**	34-369
PD7271	*JAK2*^{V617F}	**68**	41-95
PD5847	*JAK2*^{V617F}, 9pUPD	**67**	6-246
PD5163	*JAK2*^{V617F}	**43**	19-65
PD6629	*DNMT3A*^{R882H}, *JAK2*^{V617F}	**40**	27-56
PD5163	*DNMT3A*^{T275fs*41}	**38**	22-61
PD6629	*DNMT3A*^{R882H}	**26**	19-35
PD5117	*JAK2*^{V617F}	**18**	13-23
PD5847	*DNMT3A*^{Y660F}	**9**	5-25

Additional driver mutations promote rapid growth

Patient	Clade	% additional growth per yr	95% CI
PD5847	*JAK2*^{V617F}, *9pUPD*, *TET2*^{N281fs*1}	**233**	143-360
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Patient specific factors influence consequences of *JAK2*

Patient	Clade	% additional growth per yr	95% CI
PD5847	*JAK2*^{V617F}, *9pUPD*, *TET2*^{N281fs*1}	**233**	143-360
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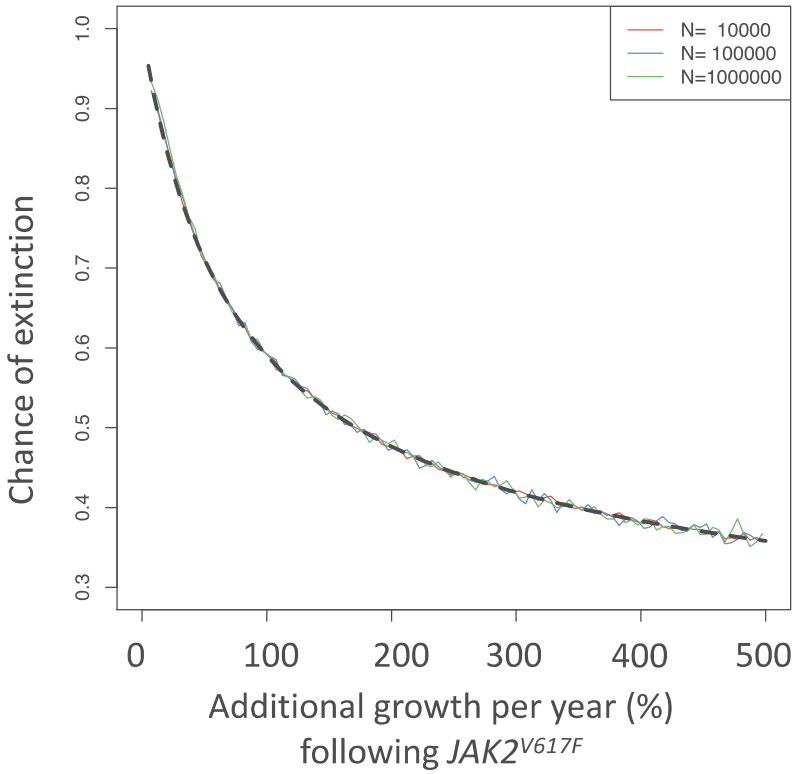
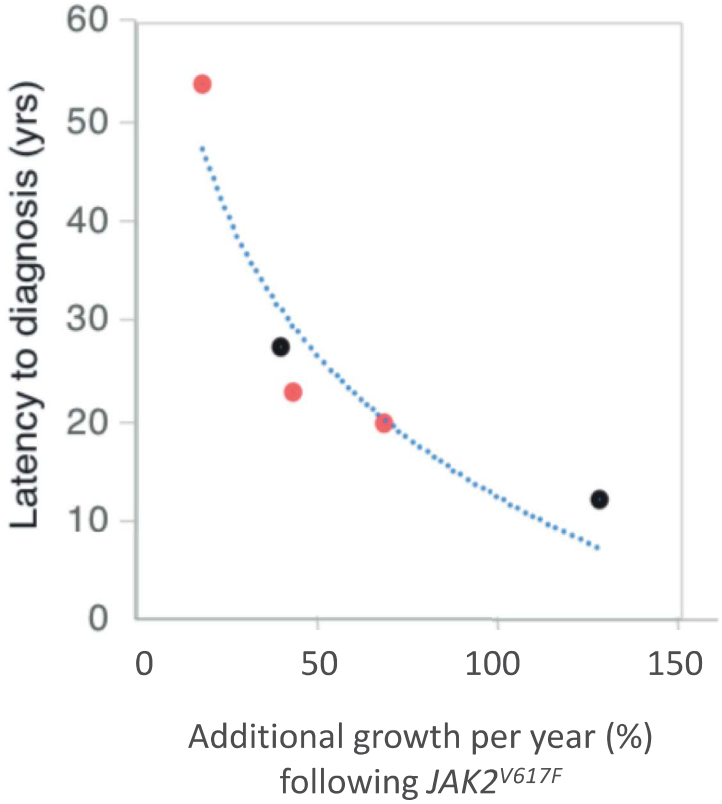
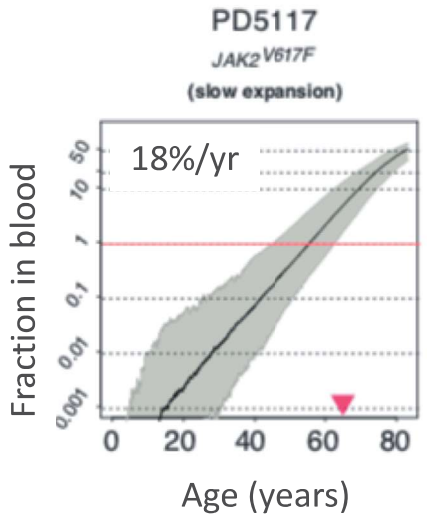
Fitness influences both CHANCE and TIME to disease



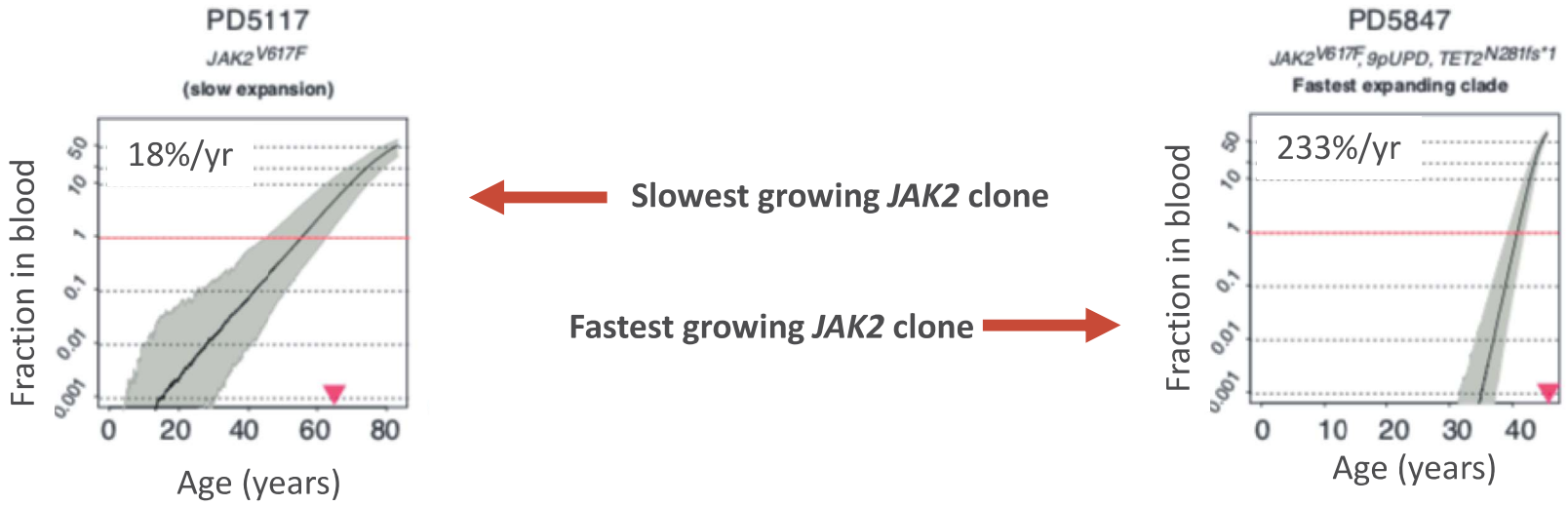


Detection decades in advance of disease presentation

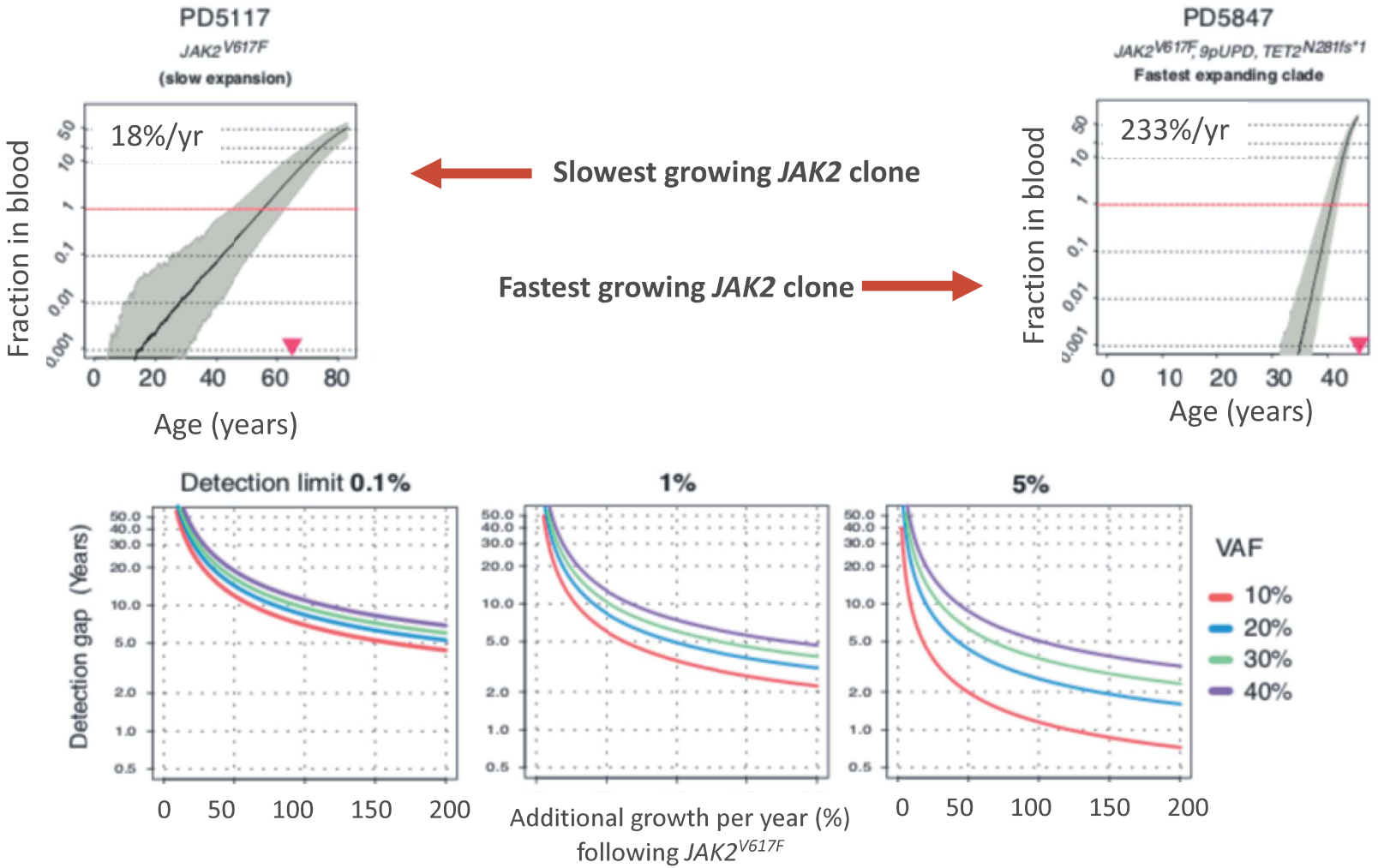


Slowest growing *JAK2* clone

Detection decades in advance of disease presentation



Early detection *AND* measurement of growth rates



Summary

- *In utero* and childhood driver mutation acquisition for *JAK2* and *DNMT3A.*
- Variable rates of clonal expansion – likely combination of *DRIVER* + *PATIENT* effects.
- Sequential driver mutation acquisition separated by decades with rapid growth.
- Expansion rates determine latency to diagnosis.
- Mutation detection *AND* rates of expansion could enable preventative strategies.

Nick Williams

Nangalia lab
Tony Green and Peter Campbell
Sanger core facility and IT
Our patients and funders

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https://www.biorxiv.org/content/10.1101/2020.11.09.374710v1

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