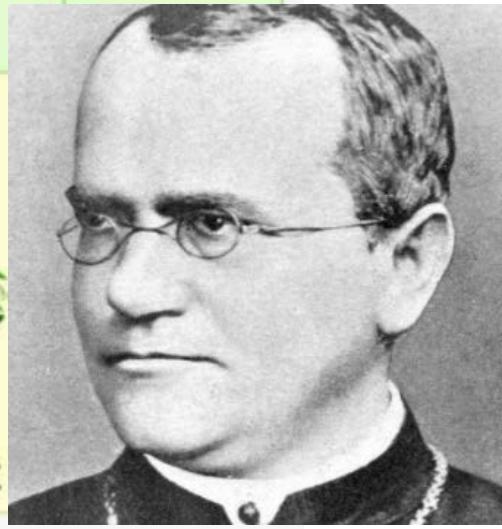
THE ROLE OF MUTATIONS IN BLOOD DISEASES IN AFRICA AND ELSEWHERE

Lucio Luzzatto
Professor of Hematology
Muhimbili University of Health and Allied Siences
Dar-es-Salaam, TANZANIA

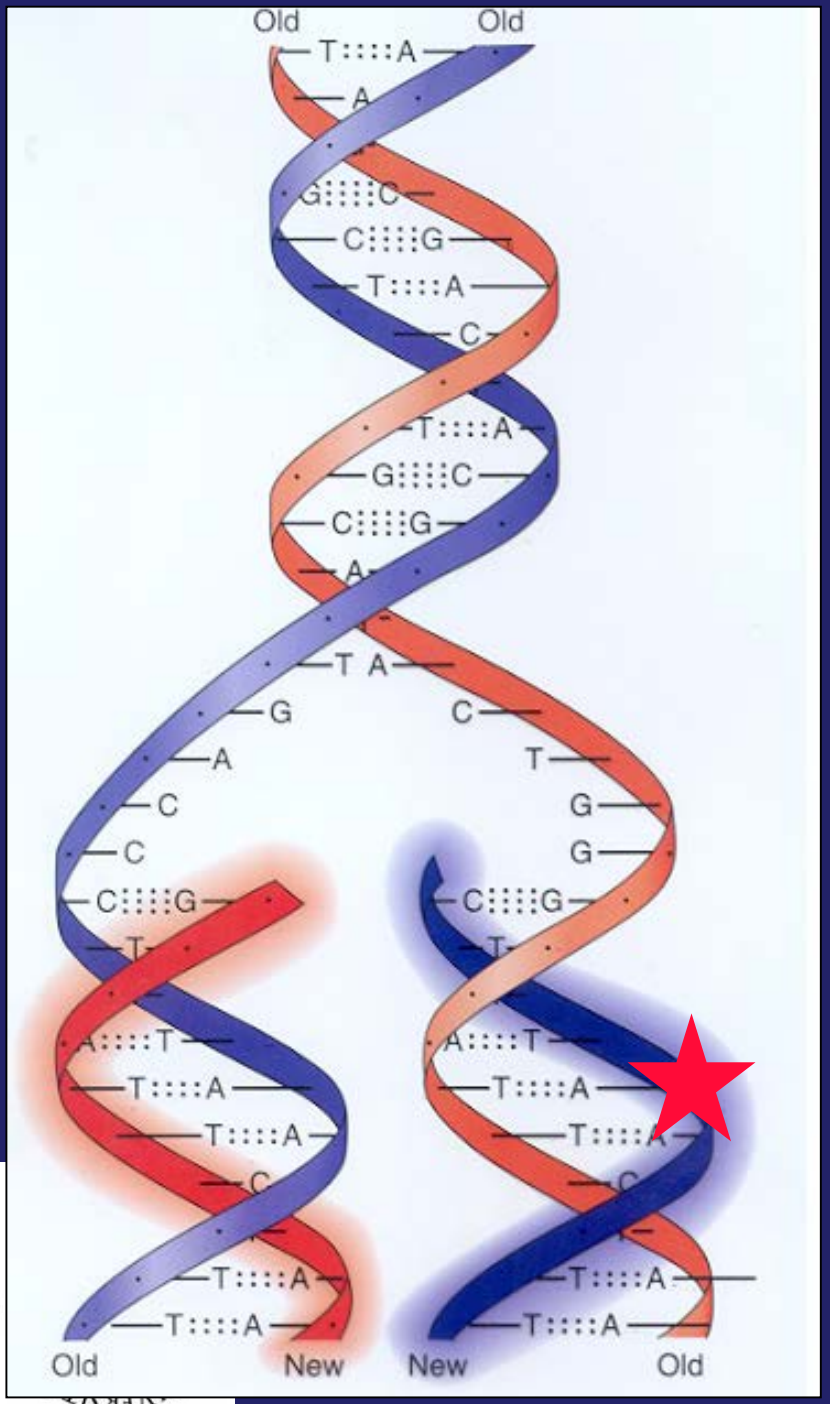
IRCCS Policlinico San Matteo
Pavia, 10 febbraio 2023

THE DAWN OF CONTEMPORARY GENETICS

	Flower color	Pea shape	Pea color	Pod color	Pod shape	Plant height	Flower position
DOMINANT	Purple	Round	Yellow	Green	Inflated	Tall	
RECESSIVE	White	Wrinkled	Green	Yellow	Constricted	Short	



Gregor Johann **Mendel** (Hynčice, 20 July 1822 – Brno, 6 January 1884)



A physical apparatus cannot be 'perfect': in spite of proofreading and mismatch repair, very rarely an error can take place;

SPONTANEOUS MUTATION

RARE *versus* COMMON MUTATIONS IN HUMAN POPULATIONS

- It is estimated that there are more than 6000 **rare diseases**: most of them are caused by germ-line mutations
- Inherited diseases can be also caused by genes that, in certain populations, are common, or *polymorphic*
- The rigorous definition of a polymorphic gene is *one whose frequency is greater than can be accounted for by recurrent mutation*; for convenience, an arbitrary threshold of 1% is widely adopted

Mutations in Hereditary Amyloidosis

Mutations in Transthyretin Gene *(TTR)* MIM *176300

Search

NCBI Reference Sequences:

Location:	**18q12.1**	
Protein:	**ACCESSION**	**NP_000362.1**
mRNA:	**ACCESSION**	**NM_000371.3**
Genomic DNA:	**ACCESSION**	**NG_009490**

TTR exon 1

Mutations associated with amyloidosis shown in red
Non-amyloidogenic mutations shown in green

AMYLOIDOGENIC AND NON-AMYLOIDOGENIC MUTATIONS IN EXON 4 OF THE *TTR* GENE

TTR exon 4

	A	C			TG				A	G	A
337	GTG	GTA	TTC	ACA	GCC	AAC	GAC	TCC	GGC	CCC	CGC
	V93M	V94A	F	T	A97G A97S	N	D	S	G101S	P102R	R103S
											C
	TA			T/G G	T	A/TT		A	T	A	CG
370	CGC	TAC	ACC	ATT	GCC	GCC	CTG	CTG	AGC	CCC	TAC
	R104H R104C	Y	T	I107V I107F I107M	A108A	A109S A109T A109V	L	L111M	S112I	P113T	Y114H Y114C Y114S
		C			T	T		AC			T
403	TCC	TAT	TCC	ACC	ACG	GCT	GTC	GTC	ACC	AAT	CCC
	S	Y116S	S	T	T119M	A120S	V	V122I V122A V122del	T	N	P125S
436	AAG	GAA	TGA								
	K	E	STOP								

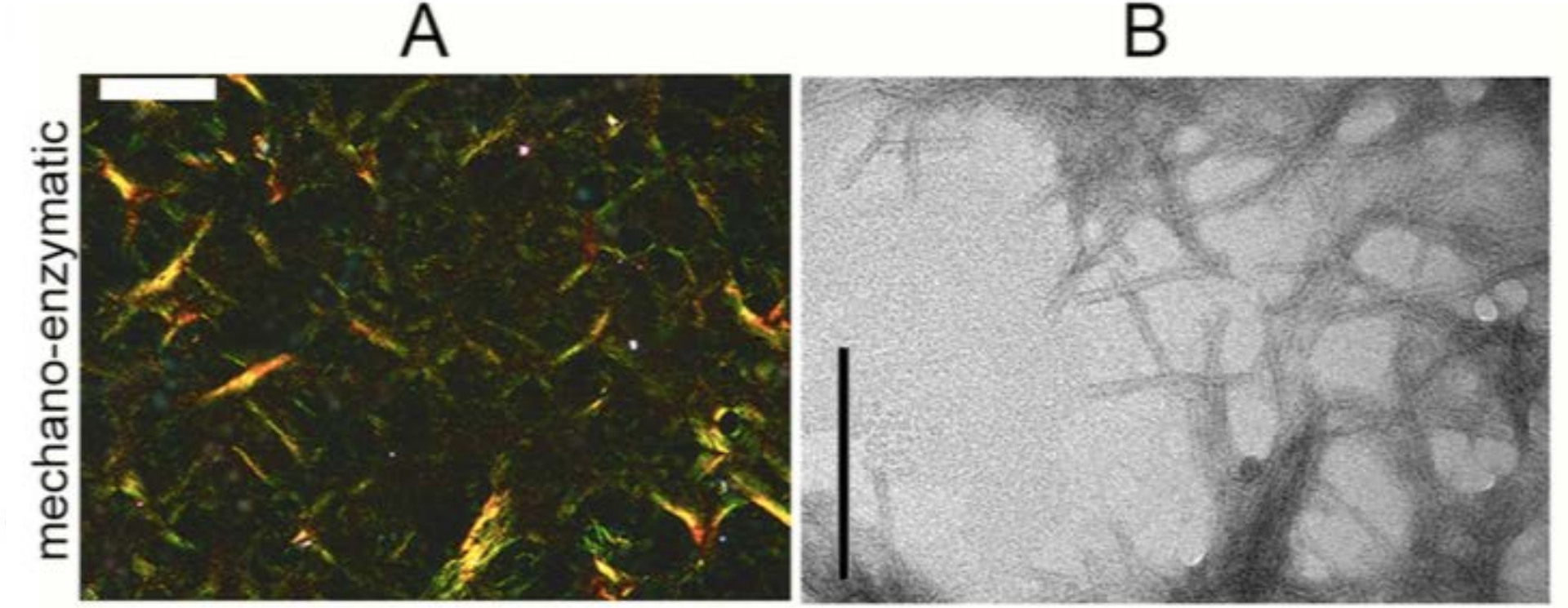
JBC EDITORS' PICK

Comparative study of the stabilities of synthetic *in vitro* and natural *ex vivo* transthyretin amyloid fibrils

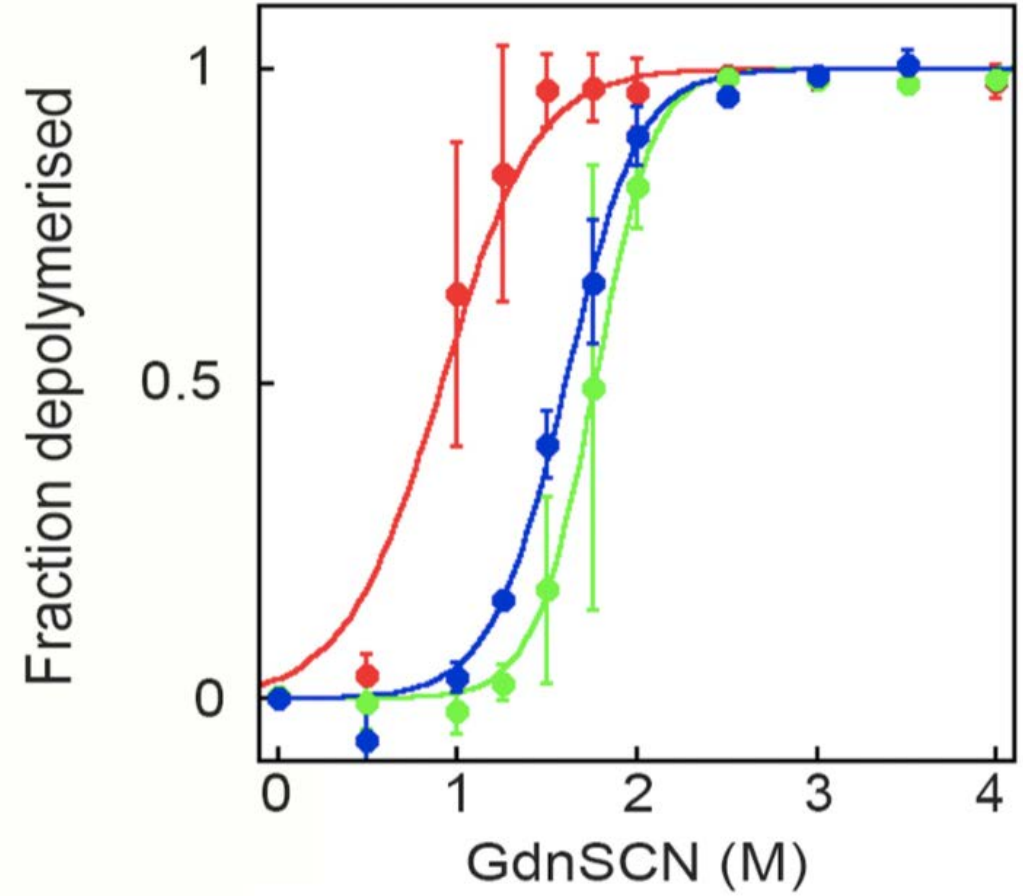
Received for publication, April 23, 2020, and in revised form, June 17, 2020 Published, Papers in Press, June 22, 2020, DOI 10.1074/jbc.RA120.014026

Sara Raimondi[1,‡], P. Patrizia Mangione[1,2,‡], Guglielmo Verona[2,‡], Diana Canetti[2], Paola Nocerino[2], Loredana Marchese[1], Rebecca Piccarducci[2,3], Valentina Mondani[1,2], Giulia Faravelli[1], Graham W. Taylor[2], Julian D. Gillmore[4], Alessandra Corazza[2,5,6], Mark B. Pepys[2,4], Sofia Giorgetti[1,6,*], and Vittorio Bellotti[1,2,*]

J. Biol. Chem. (2020) 295(33) 11379–11387



By a mechano-enzymatic approach one can form in vitro *from a* TTR^{V122I} *mutant protein amyloid fibrils similar to those found* in vivo

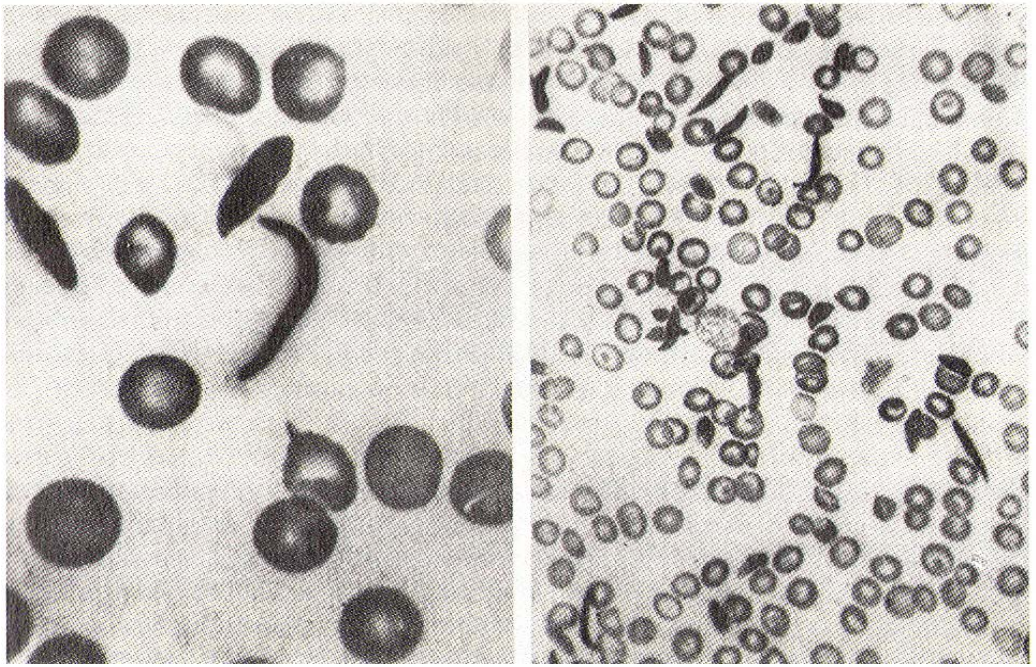


(From Raimondi et al. *JBC* **295**:11379,2020)

First description of Sickle Cell Disease from microscopy of red cells

James B Herrick. Peculiar elongated and sickle-shaped red blood corpuscles in a case of severe anemia. (Arch Intern Med. **6**: 517, 1910).

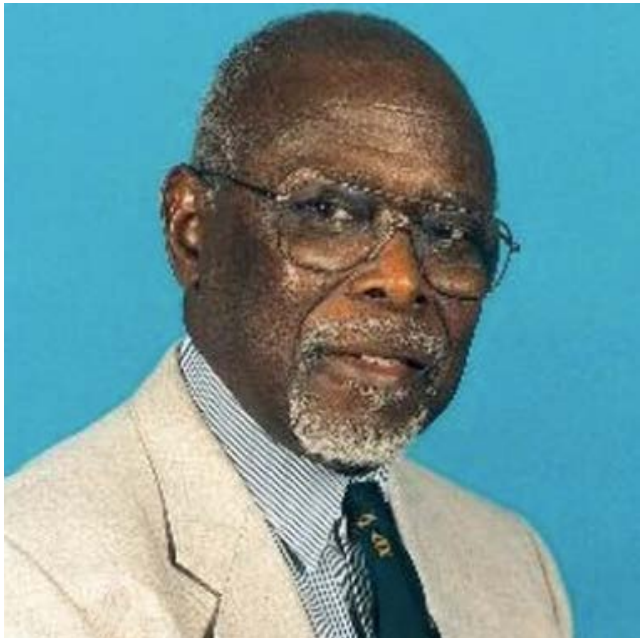
Walter Clement Noel,
Student of Dentistry, U of Chicago;
originally from Grenada, West Indies



Sickle cells first seen and sketched by ***Ernest E Irons***

Personal View

So the names of the disease, known in African tribes centuries before Herrick observed the "peculiar sickle shaped red cells" in the blood of a West Indian in 1910, were, and still are, *Chwechweechwe* pronounced chway-chway-chway (Ga language), *Nuidudui* (Ewe), *Nwiiwii* pronounced nweewee (Fante), *Ahotutuo* (Twi) etc. Onomatopoeia plays a handsome part in



F.I.D. KONOTEY-AHULU

Accra, Ghana

Sickle Cell Anemia, a Molecular Disease[1]

Linus Pauling, Harvey A. Itano,[2] S. J. Singer,[2] and Ibert C. Wells[3]

Gates and Crellin Laboratories of Chemistry,
California Institute of Technology, Pasadena, California[4]

THE ERYTHROCYTES of certain individuals possess the capacity to undergo reversible changes in shape in response to changes in the partial pressure of oxygen. When the oxygen pressure is lowered, these cells change their forms from the normal biconcave disk to crescent, holly wreath, and other forms. This process is known as sickling. About 8 percent of American Negroes possess this characteristic; usually they exhibit no pathological consequences ascribable to it. These people are said to have sicklemia, or sickle cell trait. However, about 1 in 40 (*4*) of these individuals whose cells are capable of sickling suffer from a severe chronic anemia resulting from excessive destruction of their erythrocytes; the term sickle cell anemia is applied to their condition.

The main observable difference between the erythrocytes of sickle cell trait and sickle cell anemia has been that a considerably greater reduction in the partial pressure of oxygen is required for a major fraction of the trait cells to sickle than for the anemia cells (*11*). Tests *in vivo* have demonstrated that between 30 and 60 percent of the erythrocytes in the venous circulation of sickle cell anemic individuals, but less than 1 percent of those in the venous circulation of sicklemic individuals, are normally sickled. Experiments *in vitro* indicate that under sufficiently low oxygen pressure, however, all the cells of both types assume the sickled form.

The evidence available at the time that our investigation was begun indicated that the process of sickling might be intimately associated with the state and the nature of the hemoglobin within the erythrocyte. Sickle cell erythrocytes in which the hemoglobin is combined with oxygen or carbon monoxide have the biconcave disk contour and are indistinguishable in that form from normal erythrocytes. In this condition they are termed promeniscocytes. The hemoglobin appears to be uniformly distributed and randomly oriented within normal cells and promeniscocytes, and no birefringence is observed. Both types of cells are very flexible. If the oxygen or carbon monoxide is removed, however, transforming the hemoglobin to the uncombined state, the promeniscocytes undergo sickling. The hemoglobin within the sickled cells appears to aggregate into one or more foci, and the cell membranes collapse. The cells become birefringent (*11*) and quite rigid. The addition of oxygen or carbon monoxide to these cells reverses these phenomena. Thus the physical effects just described depend on the state of combination of the hemoglobin, and only secondarily, if at all, on the cell membrane. This conclusion is supported by the observation that sickled cells when lysed with water produce discoidal, rather than sickle-shaped, ghosts (*10*).

It was decided, therefore, to examine the physical and chemical properties of the hemoglobins of individuals with sicklemia and sickle cell anemia, and to compare them with the hemoglobin of normal individuals to determine whether any significant differences might be observed.

Experimental Methods

The experimental work reported in this paper deals largely with an electrophoretic study of these hemoglobins. In the first phase of the investigation, which concerned the comparison of normal and sickle cell anemia hemoglobins, three types of experiments were performed: 1) with carbonmonoxyhemoglobins; 2) with uncombined ferrohemoglobins in the presence of dithionite ion, to prevent oxidation to methemoglobins; and 3) with carbonmonoxyhemoglobins in the presence of dithionite ion. The experiments of type 3 were performed and compared with those of type 1 in order to ascertain whether the dithionite ion itself causes any specific electrophoretic effect.

Samples of blood were obtained from sickle cell anemic individuals who had not been transfused within three months prior to the time of sampling. Stroma-free concentrated solutions of human adult hemoglobin were prepared by the method used by Drabkin (*3*). These solutions were diluted just before use with the

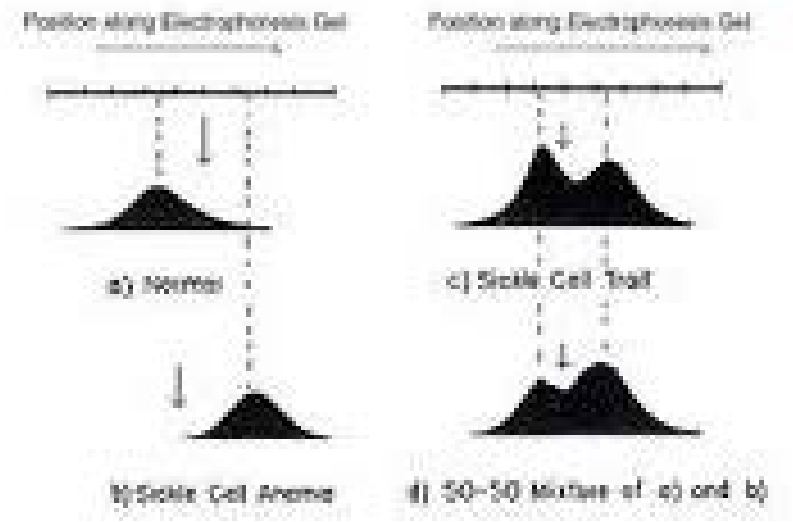
[1] This research was carried out with the aid of a grant from the United States Public Health Service. The authors are grateful to Professor Ray D. Owen, of the Biology Division of this Institute, for his helpful suggestions. We are indebted to Dr. Edward R. Evans, of Pasadena, Dr. Travis Winsor, of Los Angeles, and Dr. G. E. Burch, of the Tulane University School of Medicine, New Orleans, for their aid in obtaining the blood used in these experiments.

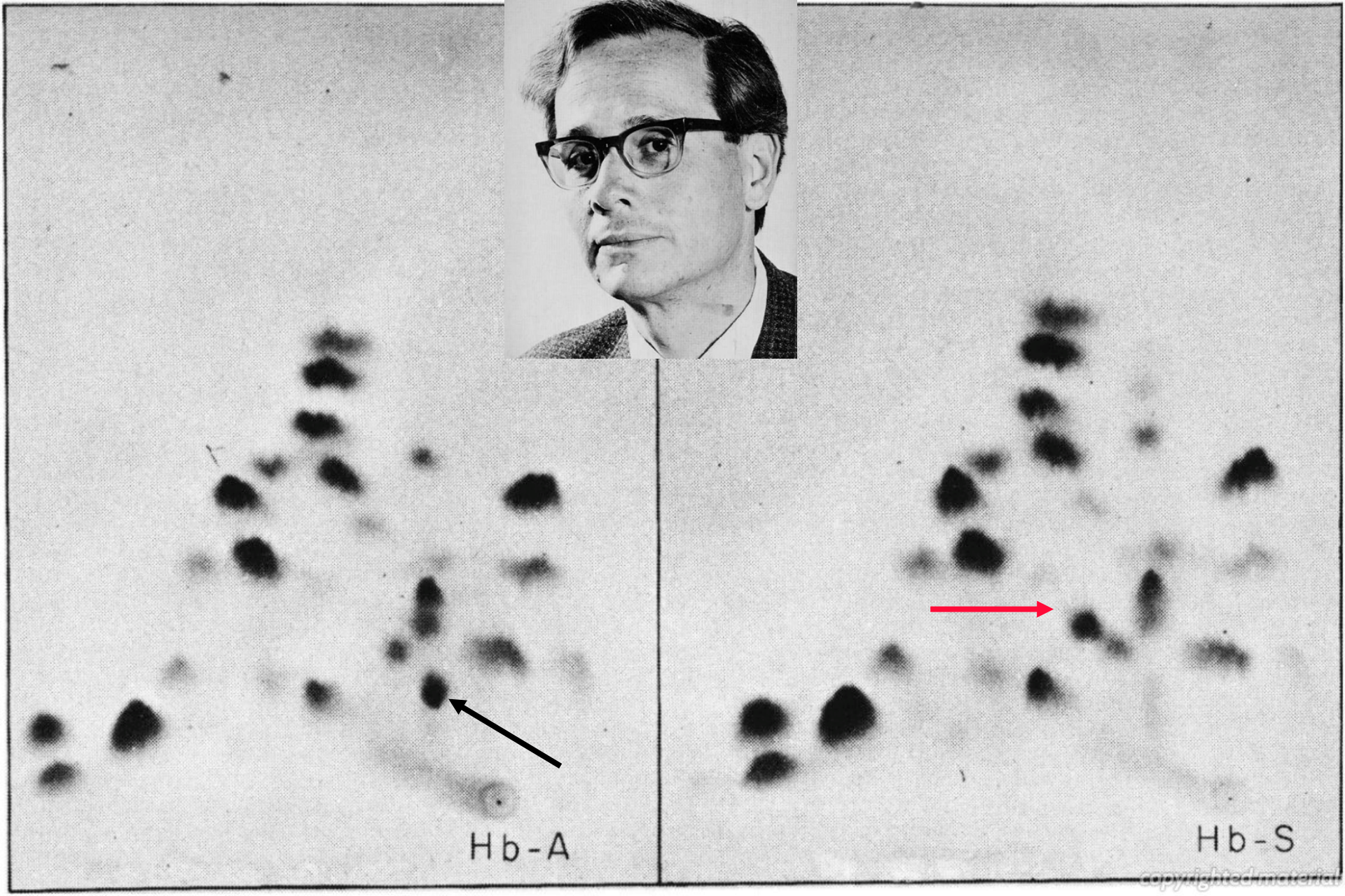
[2] U. S. Public Health Service postdoctoral fellow of the National Institutes of Health.

[3] Postdoctoral fellow of the Division of Medical Sciences of the National Research Council.

[4] Contribution No. 1333.

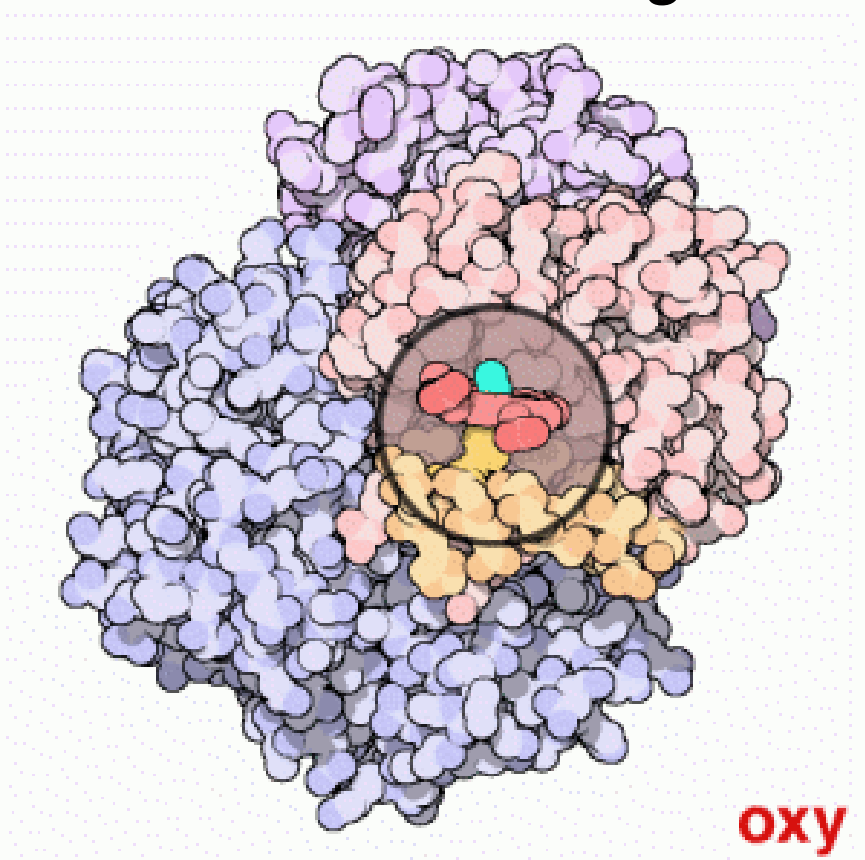
1949





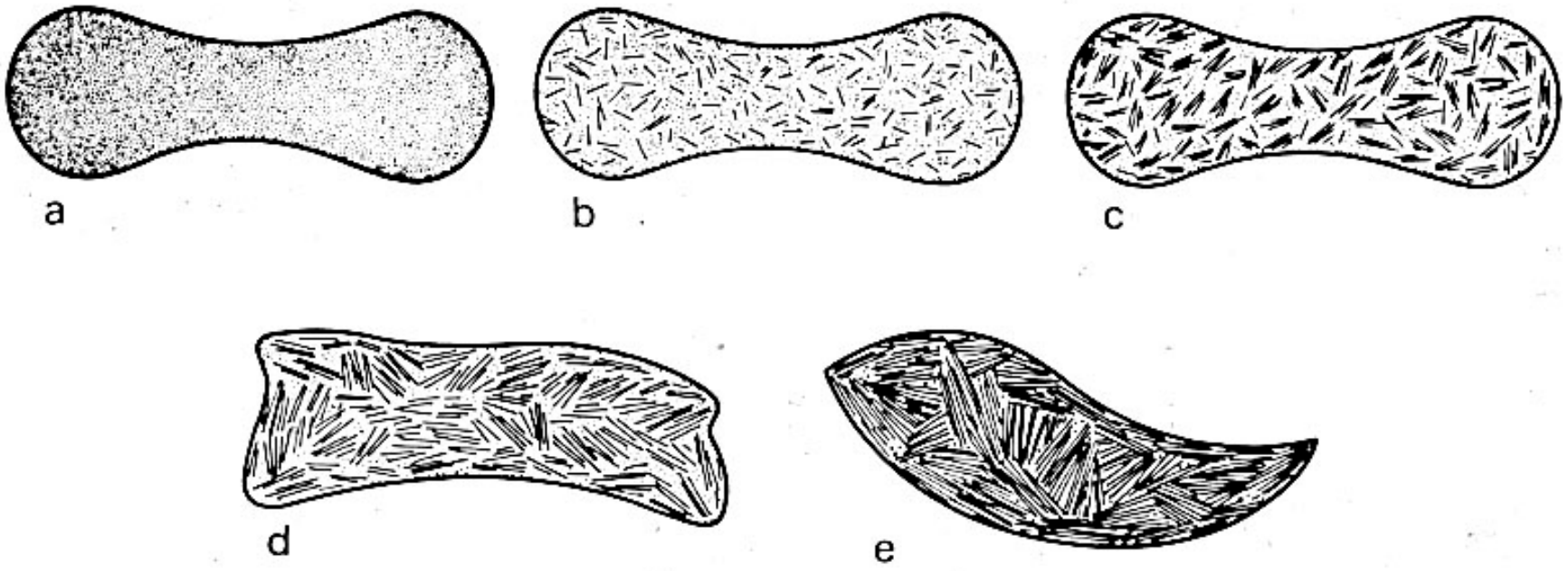
Fingerprints of Hb A and Hb S: Vernon INGRAM, 1956

Binding of oxygen to haeme produces a conformational change of the entire haemoglobin molecule

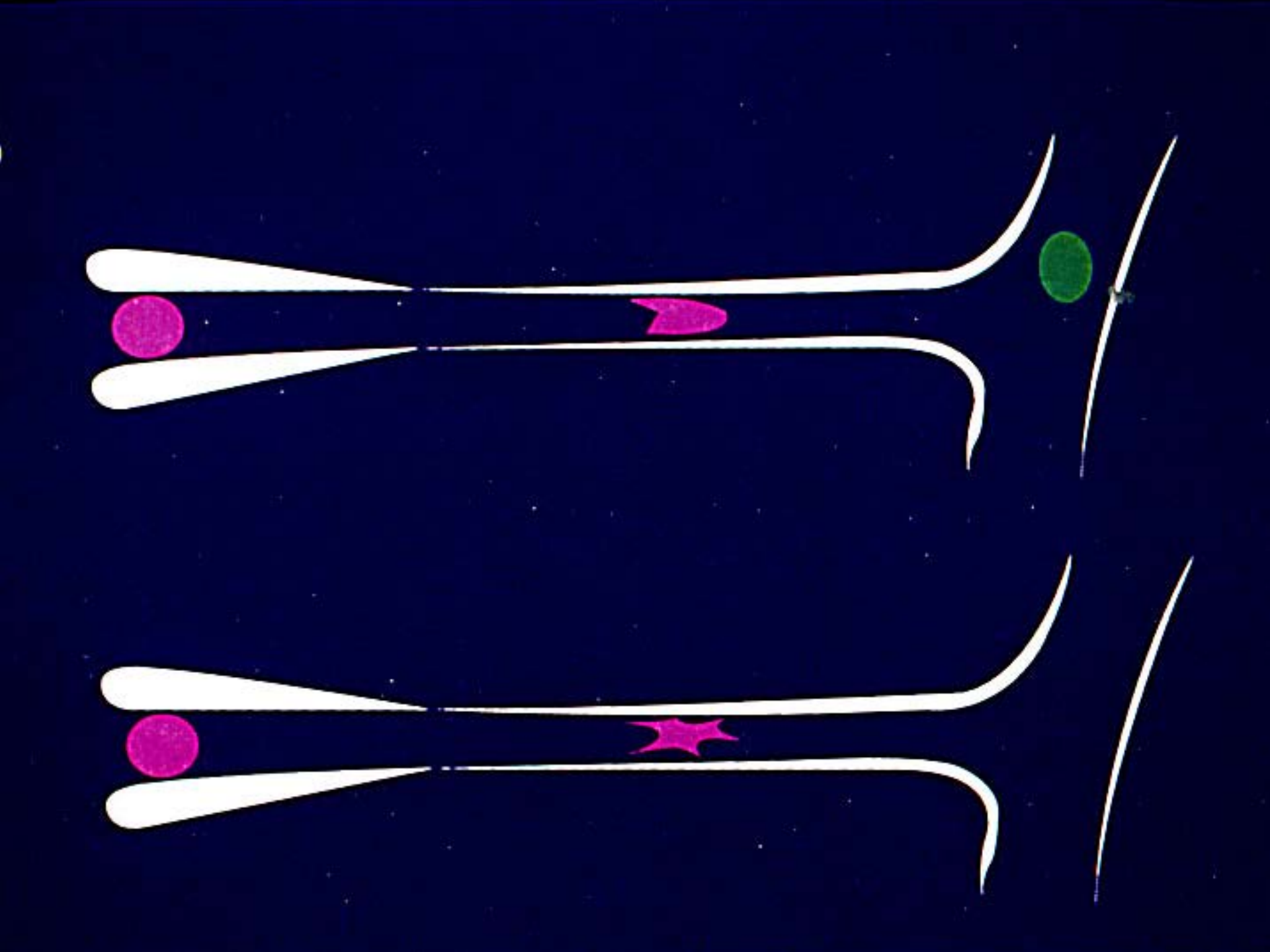


The tetrameric structure enables the hemoglobin molecule to 'breathe'

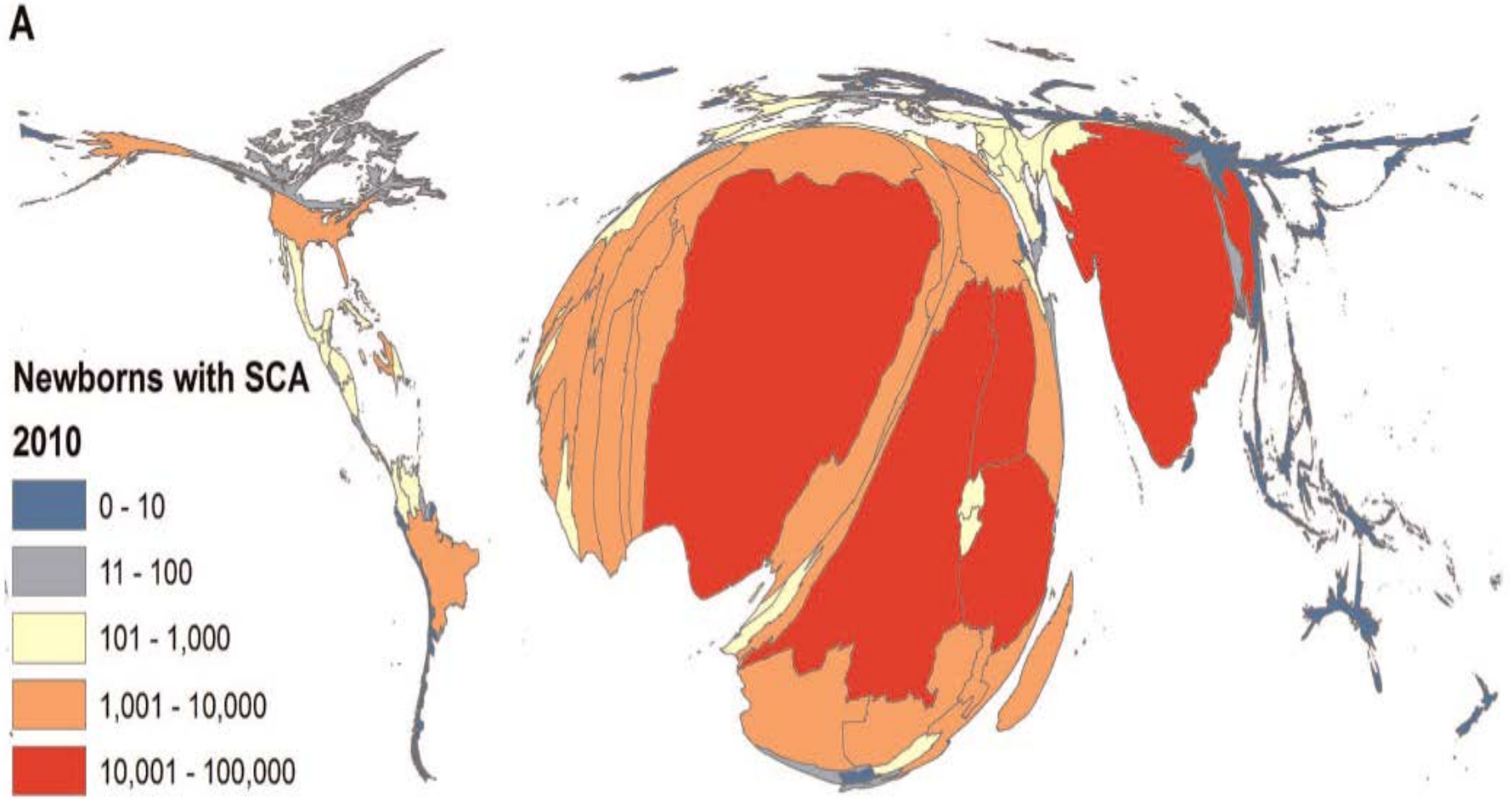
Polymerization of deoxyhaemoglobin S is a fast reaction; sickling of a red cell is a slower gradual process



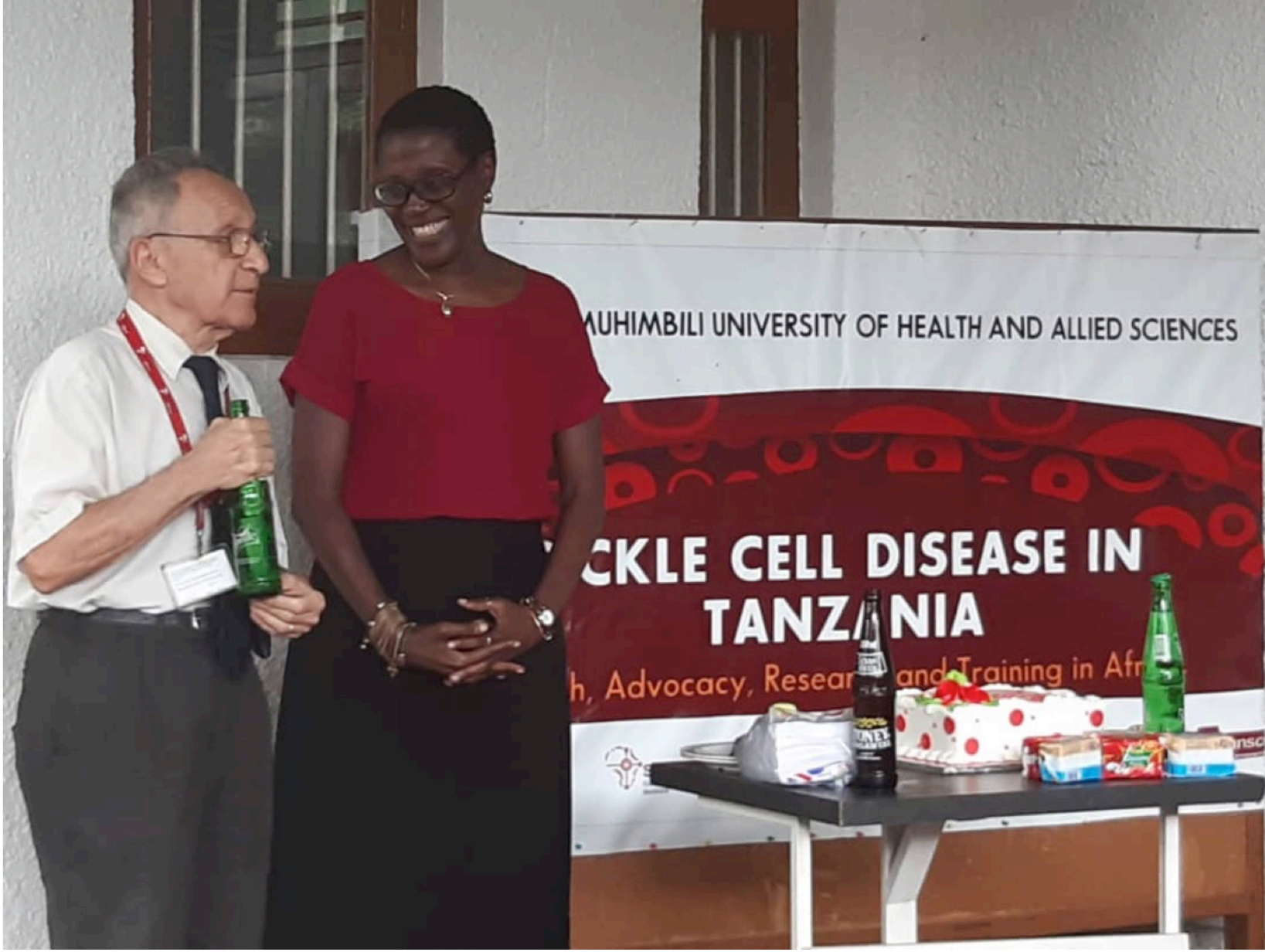
From Noguchi and Schechter (1981)



THE SICKLE CELL ANAEMIA BURDEN IN THE WORLD



(From Piel et al., *PLoS Medicine* :e1001484,2013)


AUHIMBILI UNIVERSITY OF HEALTH AND ALLIED SCIENCES
CKLE CELL DISEASE IN
TANZA NIA

Makani et al. BMC Hematology (2018) 18:33
https://doi.org/10.1186/s12878-018-0125-0

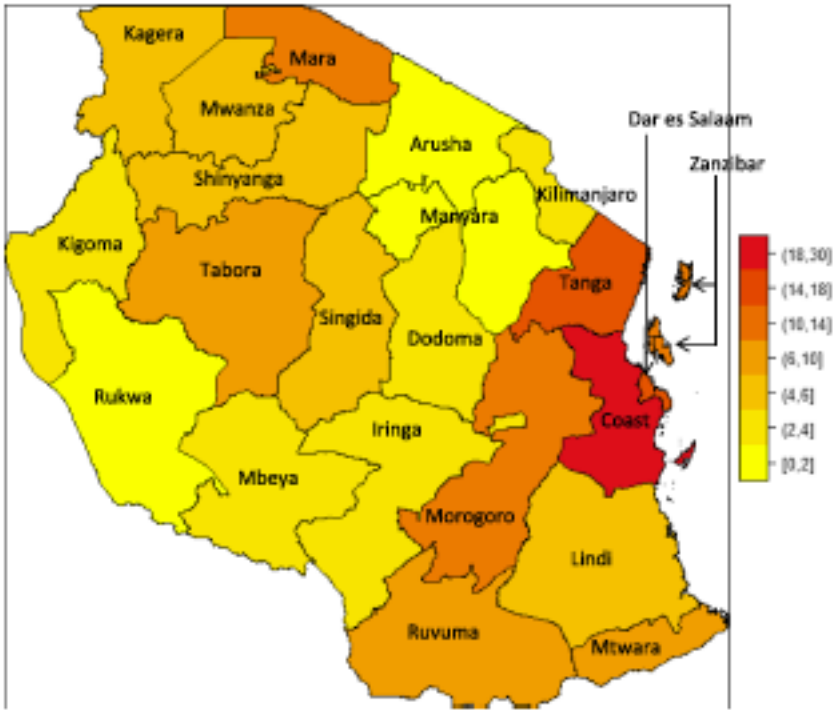
BMC Hematology

RESEARCH ARTICLE Open Access

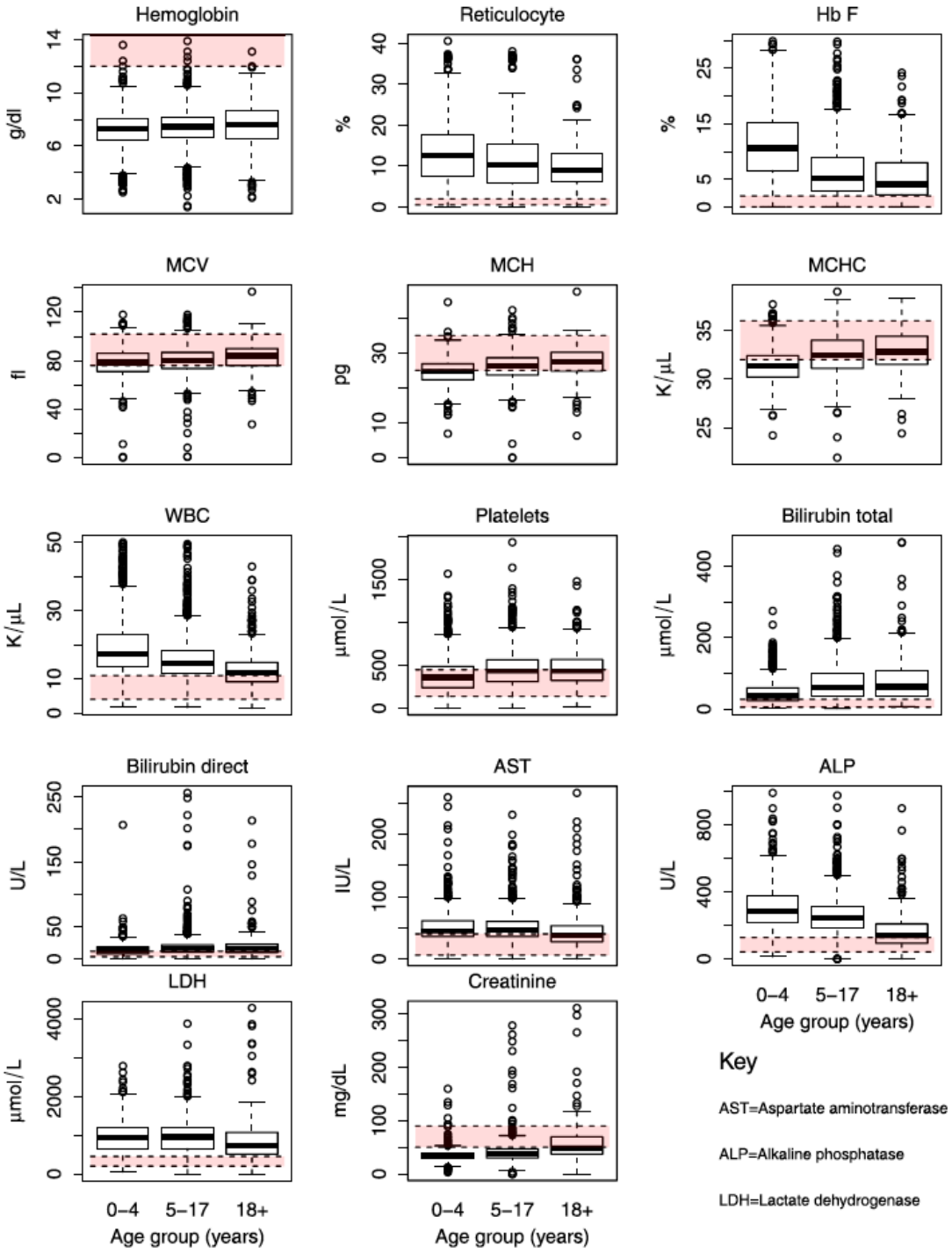
A ten year review of the sickle cell program in Muhimbili National Hospital, Tanzania

Julie Makani[1,2,3*], Furahini Tluway[1], Abel Makubi[1], Deogratius Soka[1], Siana Nkya[1,4], Raphael Sangeda[1], Josephine Mgaya[1], Stella Rwezaula[1,3], Fenella J. Kirkham[5], Christina Kindole[1,3], Elisha Osati[1,3], Elineema Meda[3], Robert W. Snow[2,6], Charles R. Newton[2,6], David Roberts[2], Muhsin Aboud[1], Swee Lay Thein[7], Sharon E. Cox[8], Lucio Luzzatto[1†] and Bruno P. Mmbando[1,9†]

- A study of 3751 patients with SCD
- At least 90% homozygous SS
- Co-existing α-thalassaemia in nearly 40% of cases

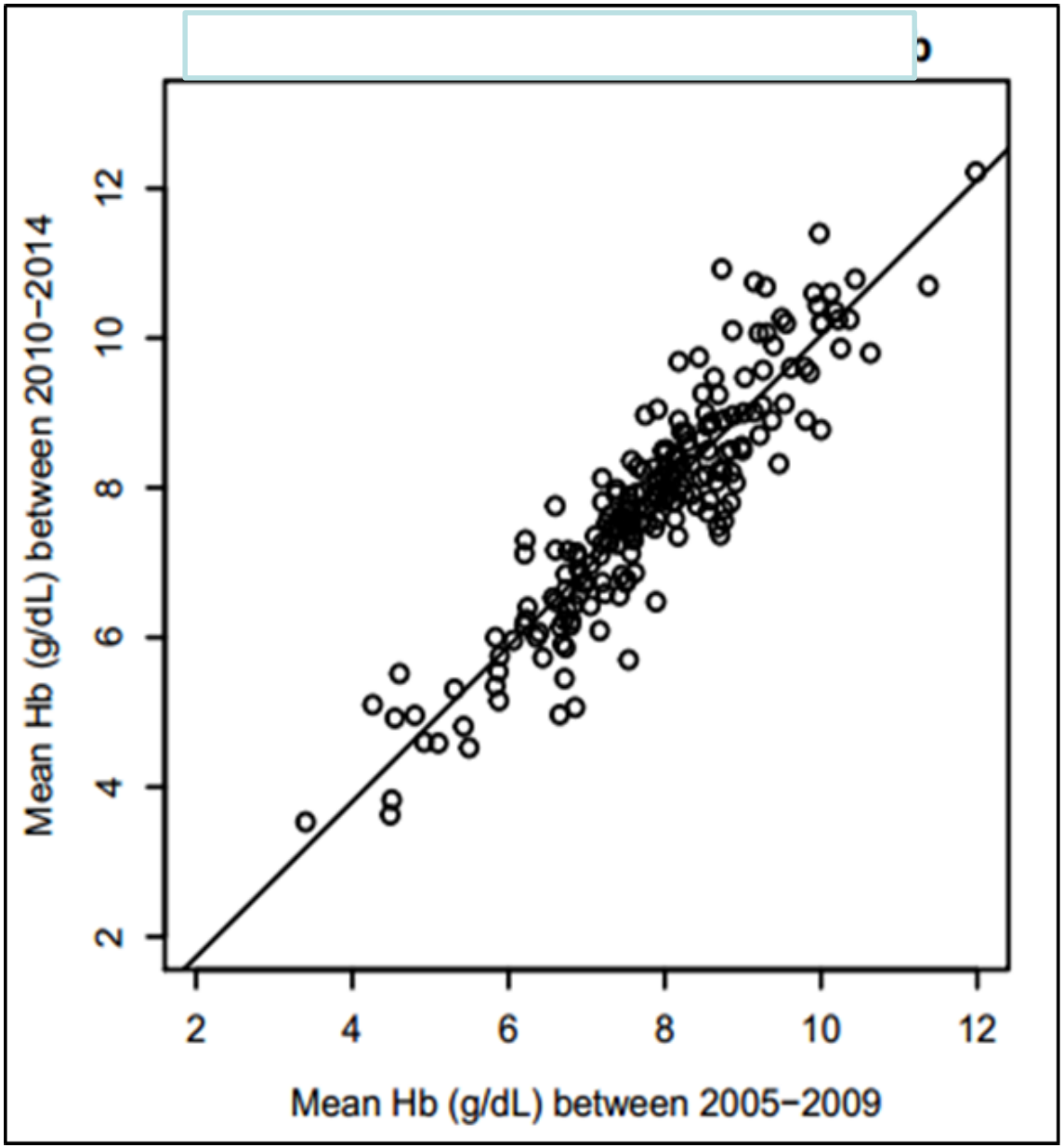


Several laboratory parameters in sickle cell disease are influenced by age (n = 3751)

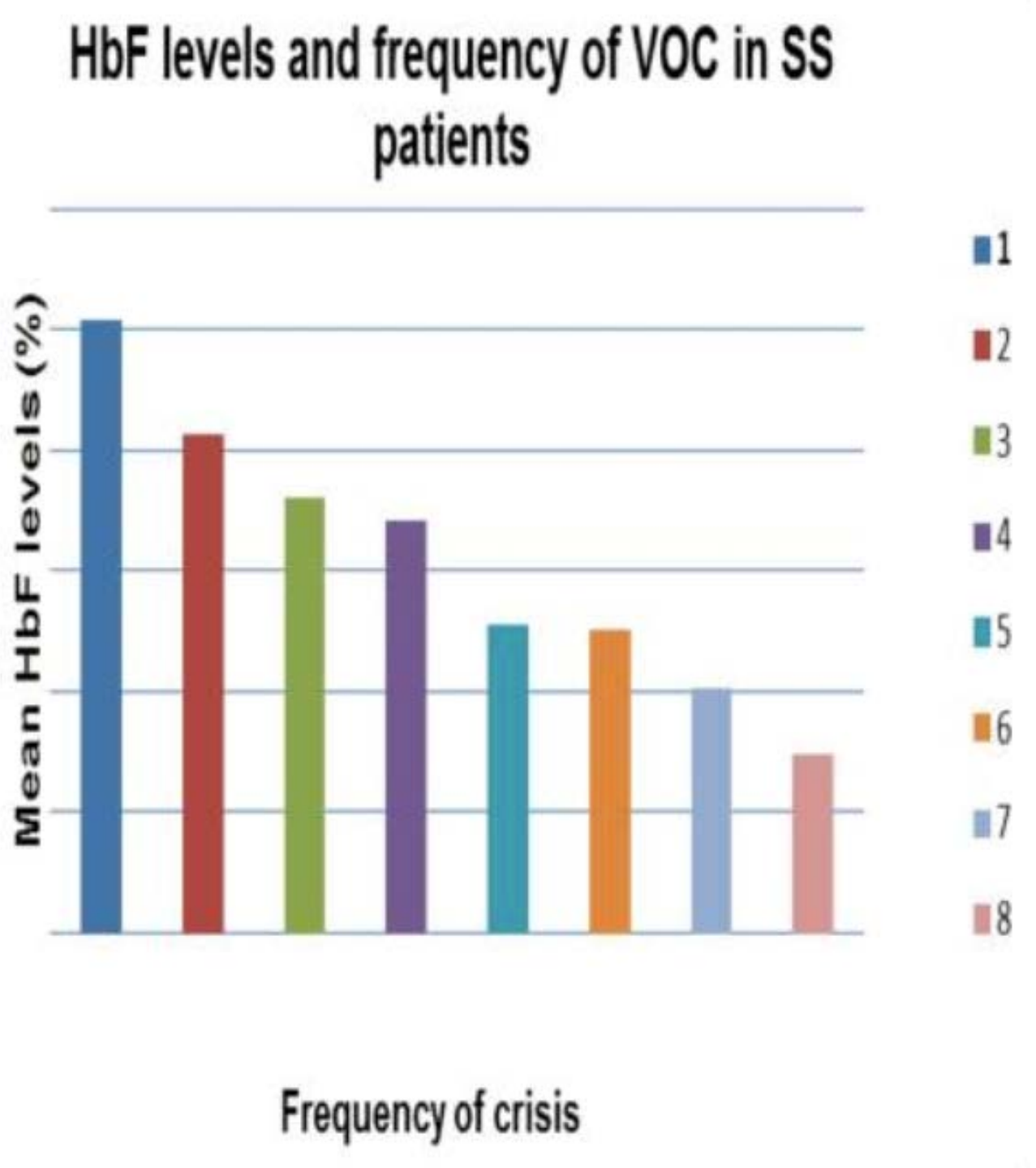


(From Makani *et al.*, *BMC Hematology* **18**:33,2018)

The ***steady-state Hb*** *is a robust characteristic of each individual patient with SCD (n = 245, age>15)*

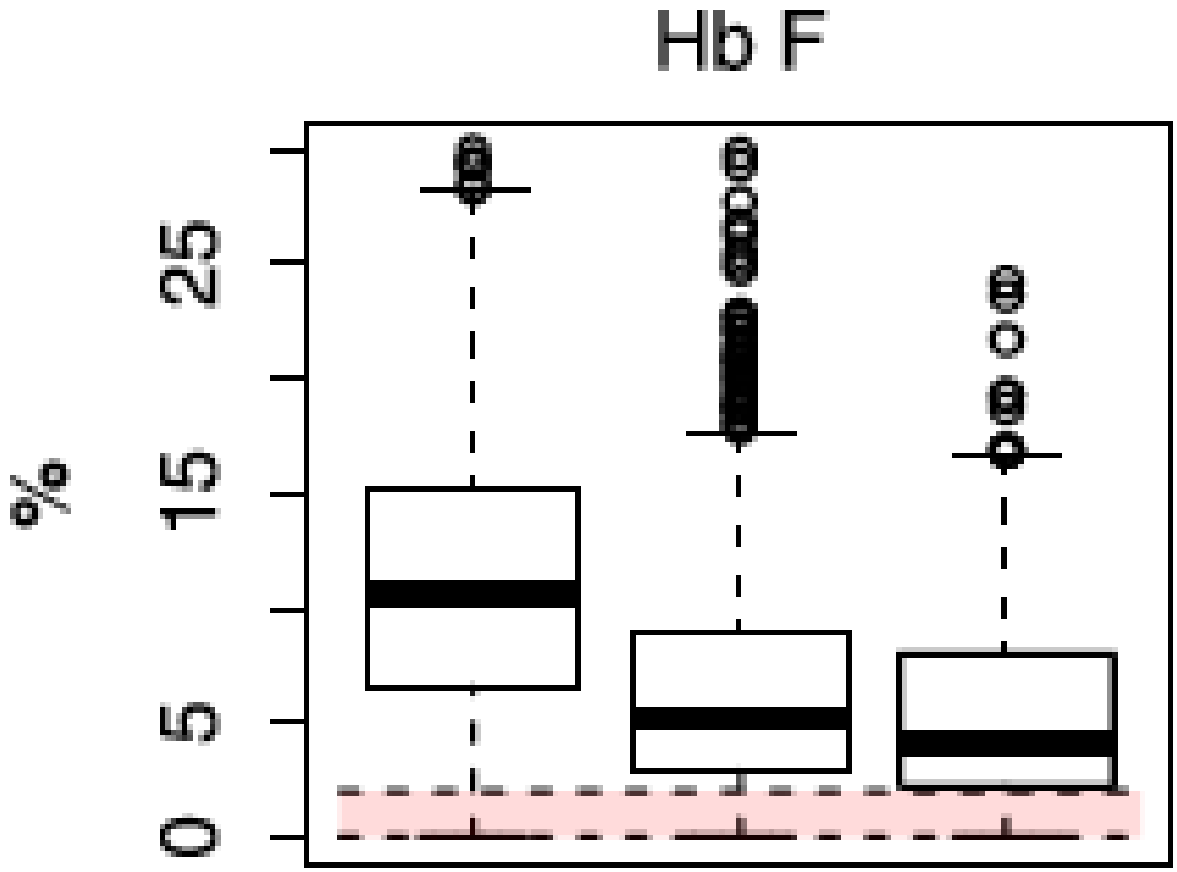


(J Makani, B Mmbando and L Luzzatto, *unpublished*)



(From Antwi-Bosiako et al., *Ghana Med J* **49**:102,2015)

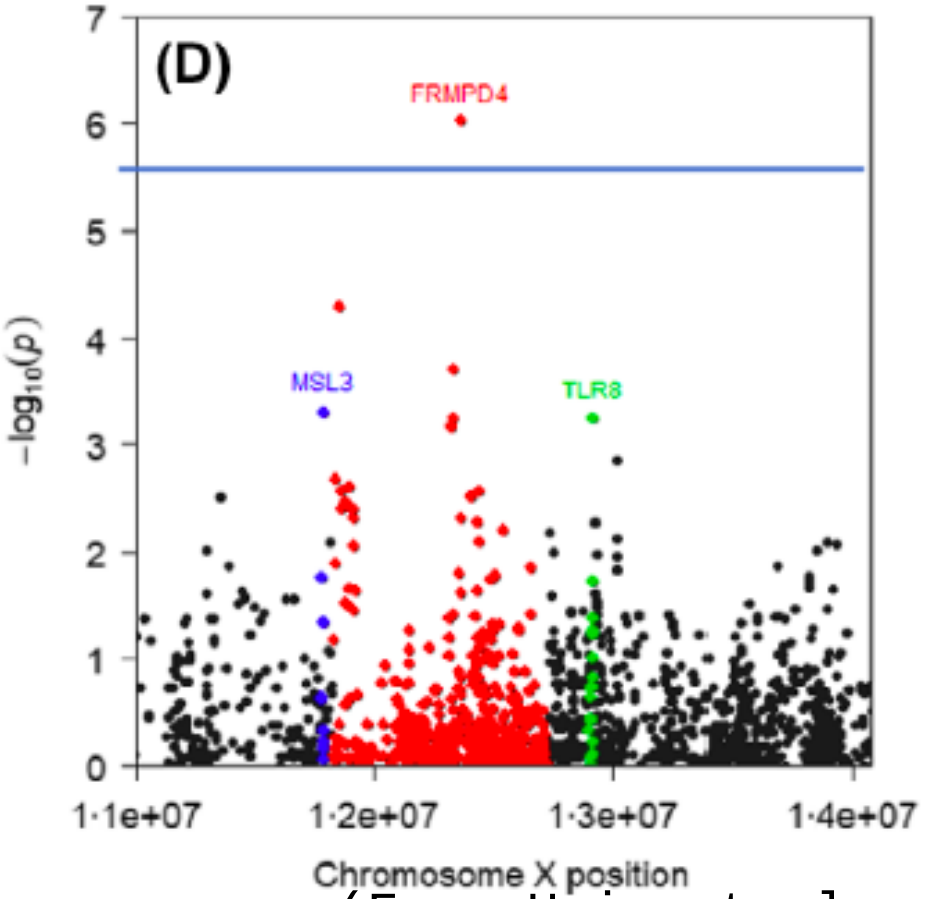
DELAYED DECREASE, IN CHILDREN WITH SCD, OF FETAL HAEMOGLOBIN, A MAJOR MODULATOR OF DISEASE SEVERITY



(From Makani *et al., BMC Hematology* **18**:33,2018)

Three SCD modifier genes have been previously identified, that significantly affect Hb F synthesis:

- *The* γ globin gene itself on 11p15.5
- *HBS1L-MYB* intergenic region on 6q23
- *BCL11A on* 2p16.1



(From Urio et al, *Am J Hematology* **191**:888,2020)

Thus, we may now have a fourth gene:

- *FRMPD4* on Xp22.2

Hydroxyurea Affordable for SCD in Tanzania is Essential (HASTE)

MUHAS, University of Verona. Support from Aurelio Maggio

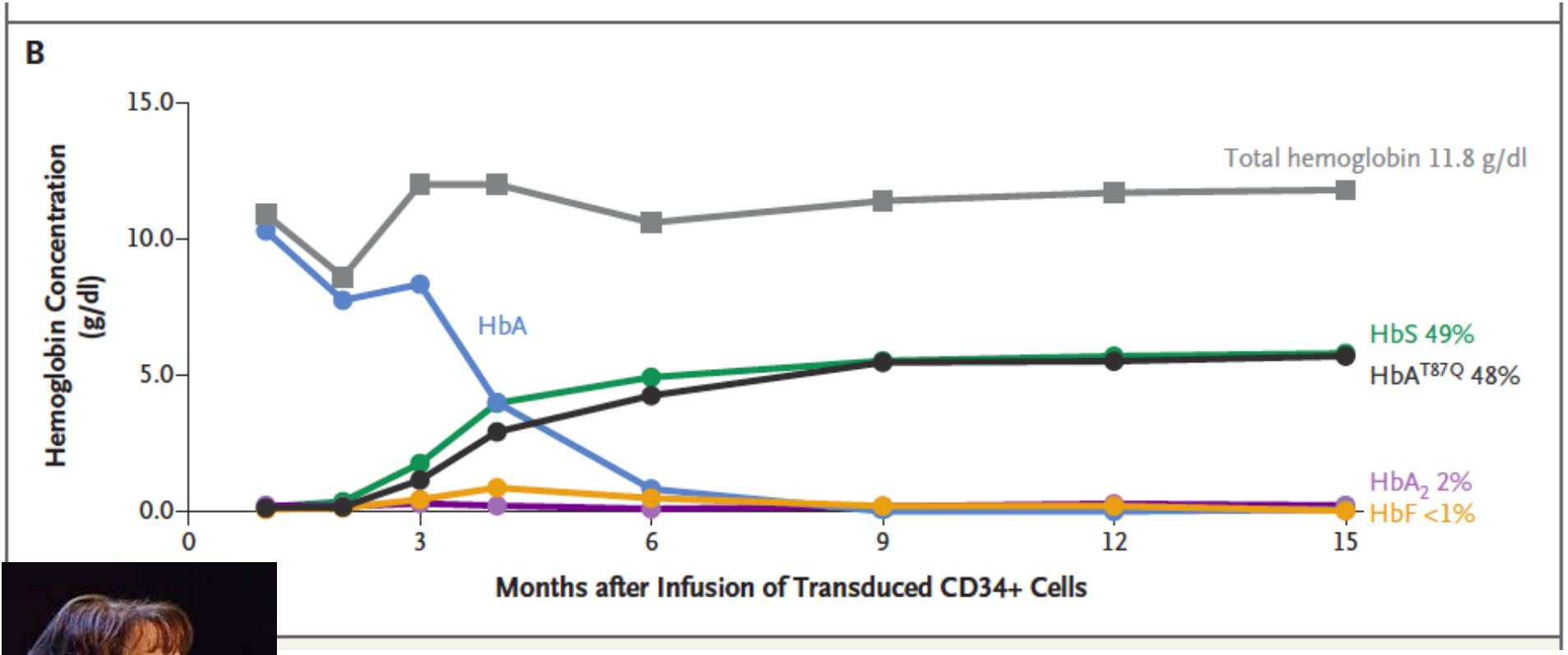
See Costa et al., *Am J Hematol* 2020 Sep 24

Specific proposals aiming to reduce the gap between potential and reality

(Modified from Luzzatto & Makani, *Front Pharmacol* **12**:770640,2022)

- Adding **SCD**
  to the triad of conditions (HIV, tuberculosis, malaria)
  for which cost of treatment is born by the Global Fund.

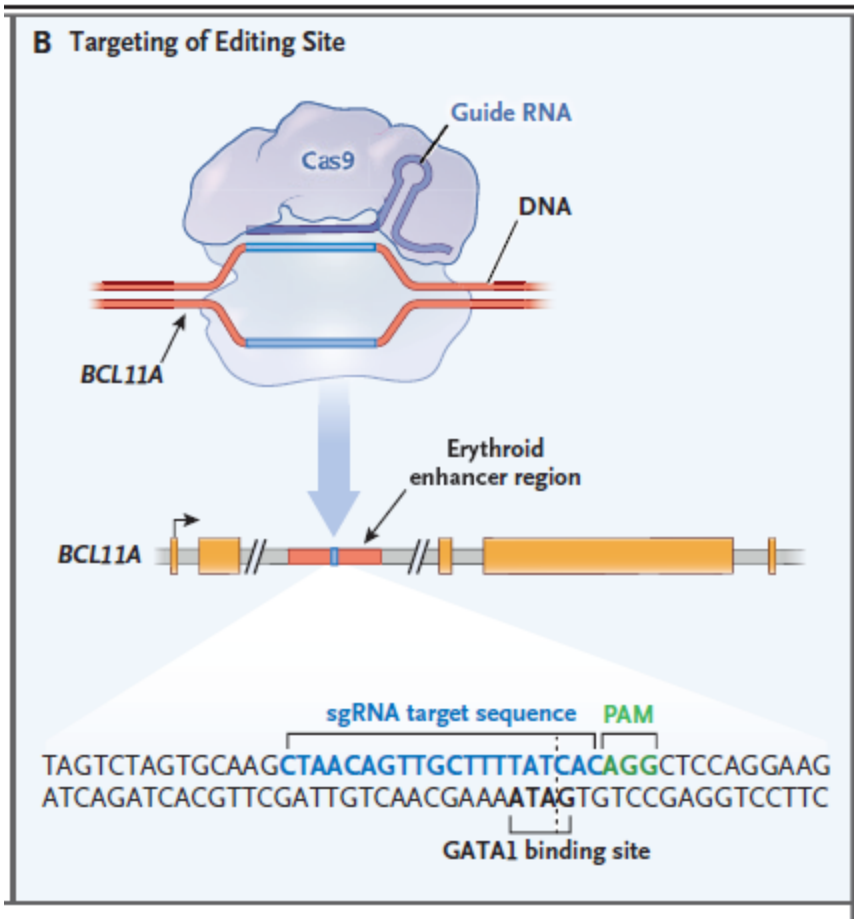
SUCCESFUL GENE THERAPY
IN A PATIENT WITH SICKLE CELL ANAEMIA



Courtesy of ***Marina Cavazzana-Calvo***

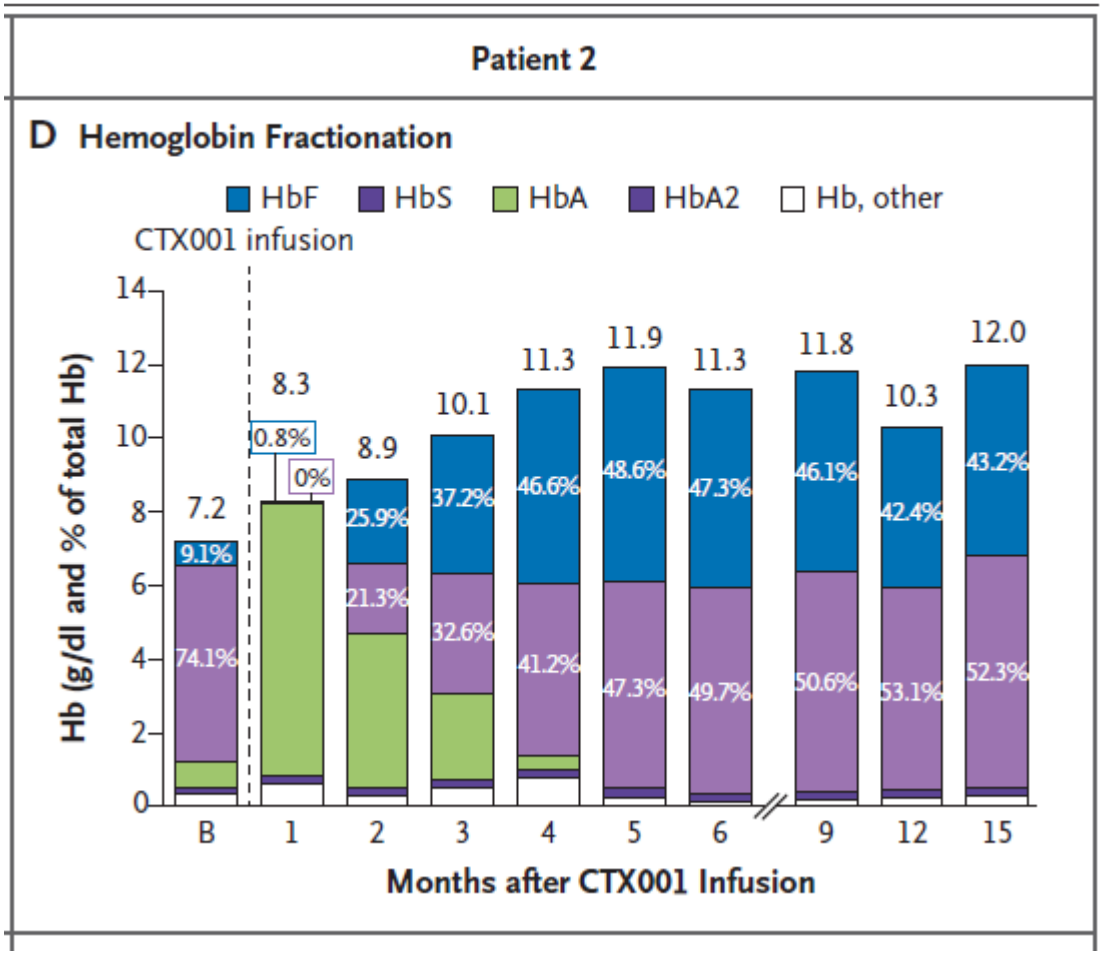
(Ribeil et al, NEJM **376***:* 848, 2017)

RNA-guided inactivation of BCL11A *(close to the GATA1 binding site) in hematopoietic stem cells*



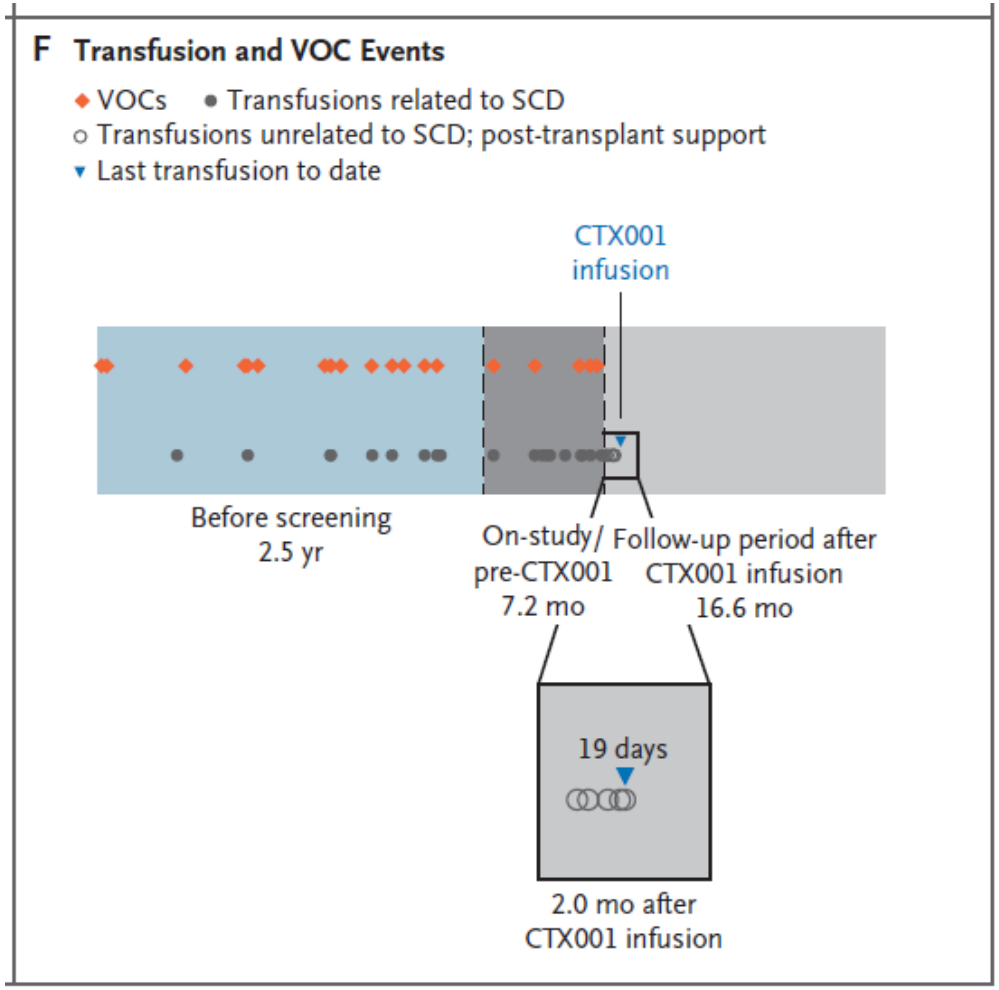
(From Frangoul et al., *NEJM*, 05.12.2020)

CRISPR-mediated inactivation of BCL11A *causes impressive increase in Hb F in a patient with severe SCD*



(From Frangoul et al., *NEJM*, 05.12.2020)

No more bone pain crises and no need for blood transfusion after CRISPR-mediated inactivation of BCL11A



(From Frangoul et al., *NEJM*, 05.12.2020)

Specific proposals aiming to reduce the gap between potential and reality

(Modified from Luzzatto & Makani, *Front Pharmacol* **12**:770640,2022)

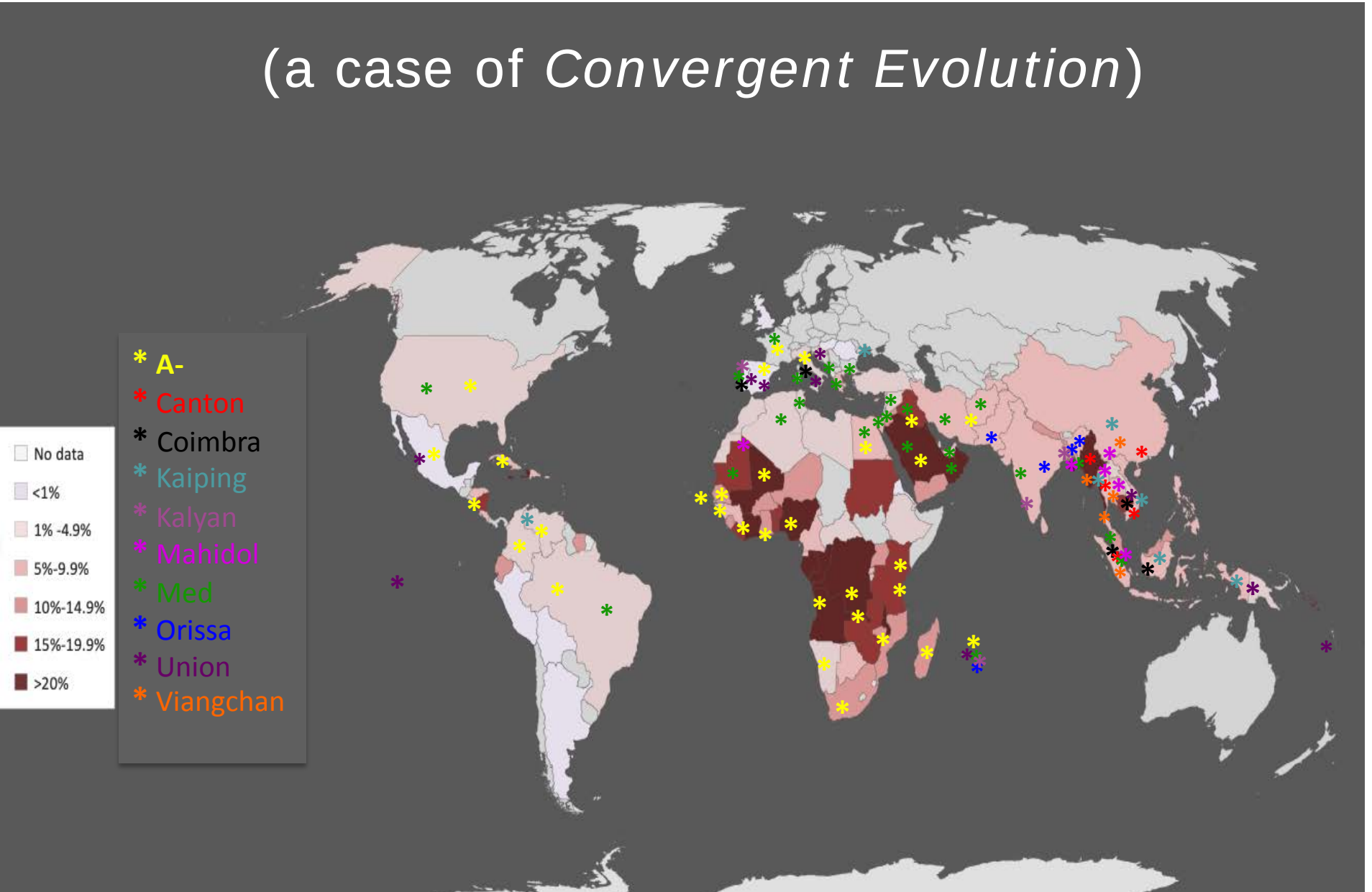
- Adding **SCD**
  to the triad of conditions (HIV, tuberculosis, malaria)
  for which cost of treatment is born by the Global Fund.

- BMT solidarity programme:
  for every BMT (HSCT) procedure in Europe/US,
  0.1% of the expense could be deposited into a fund
  to support BMT in accredited centers in Africa.

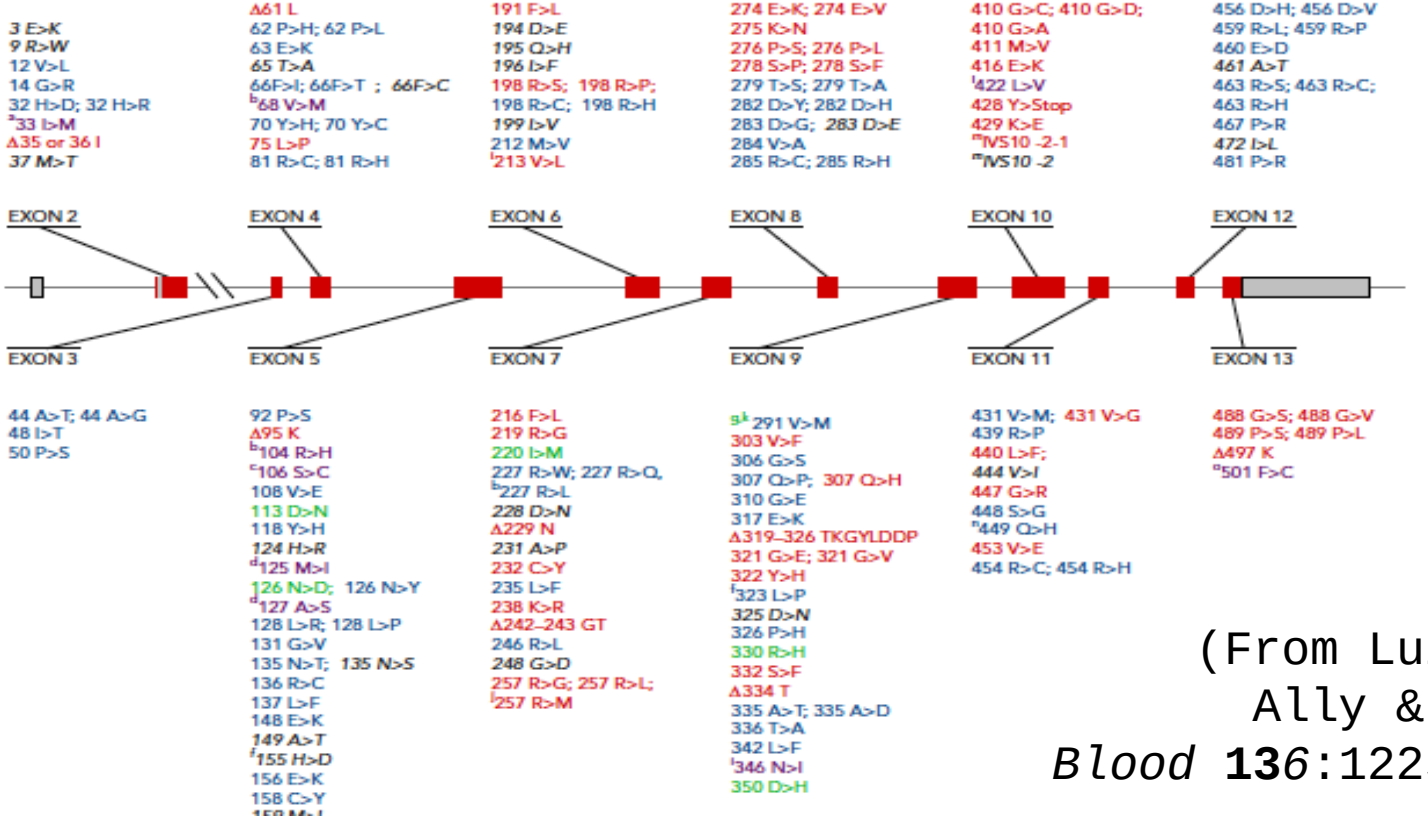
1874. **Charles DARWIN**
was elected foreign member
of Accademia dei Lincei

The frequency of independently arisen G6PD mutations has increased wherever malaria was endemic

(a case of *Convergent Evolution*)

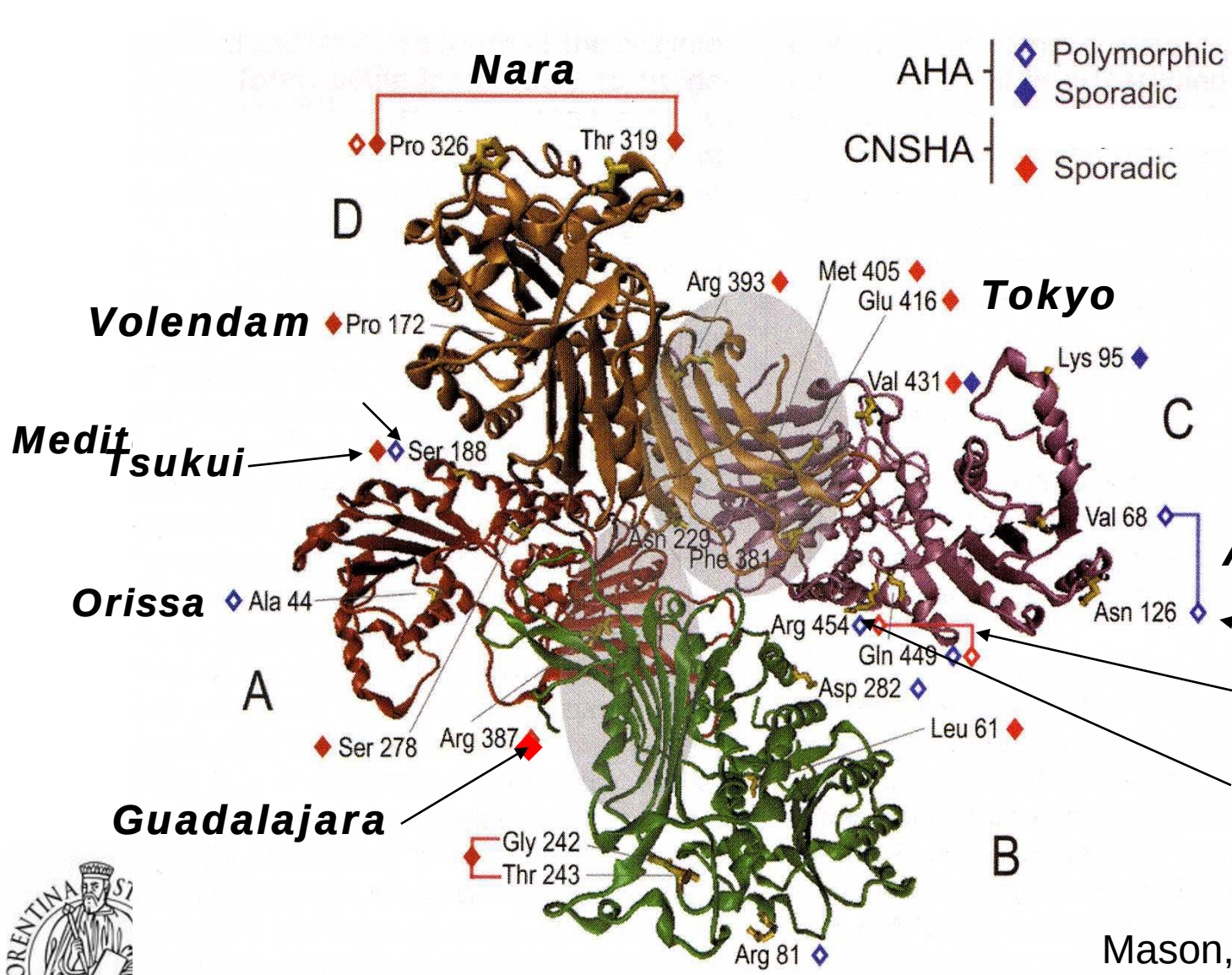


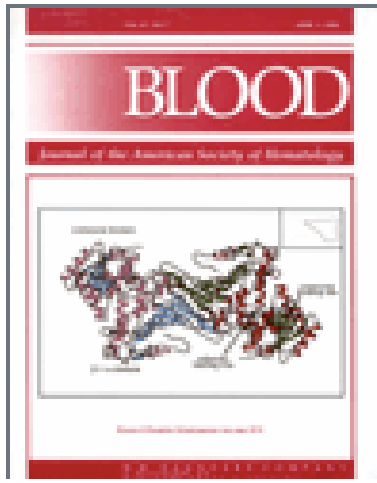
POINT MUTATIONS IN *G6PD*



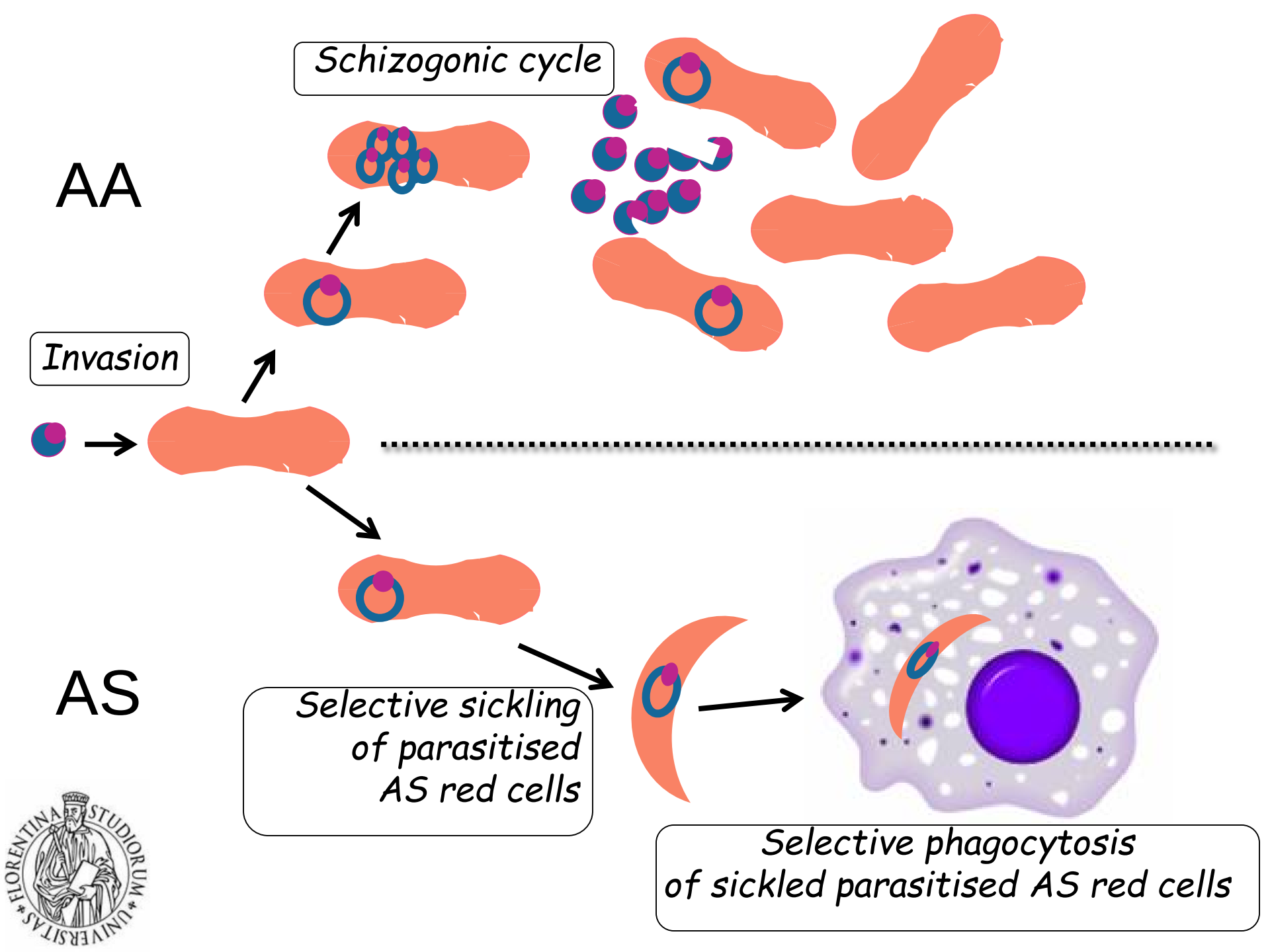
(From Luzzatto, Ally & Notaro *Blood* **136**:1225,2020)

G6PD DEFICIENCY: GENOTYPE-PHENOTYPE CORRELATIONS AT THE MOLECULAR LEVEL

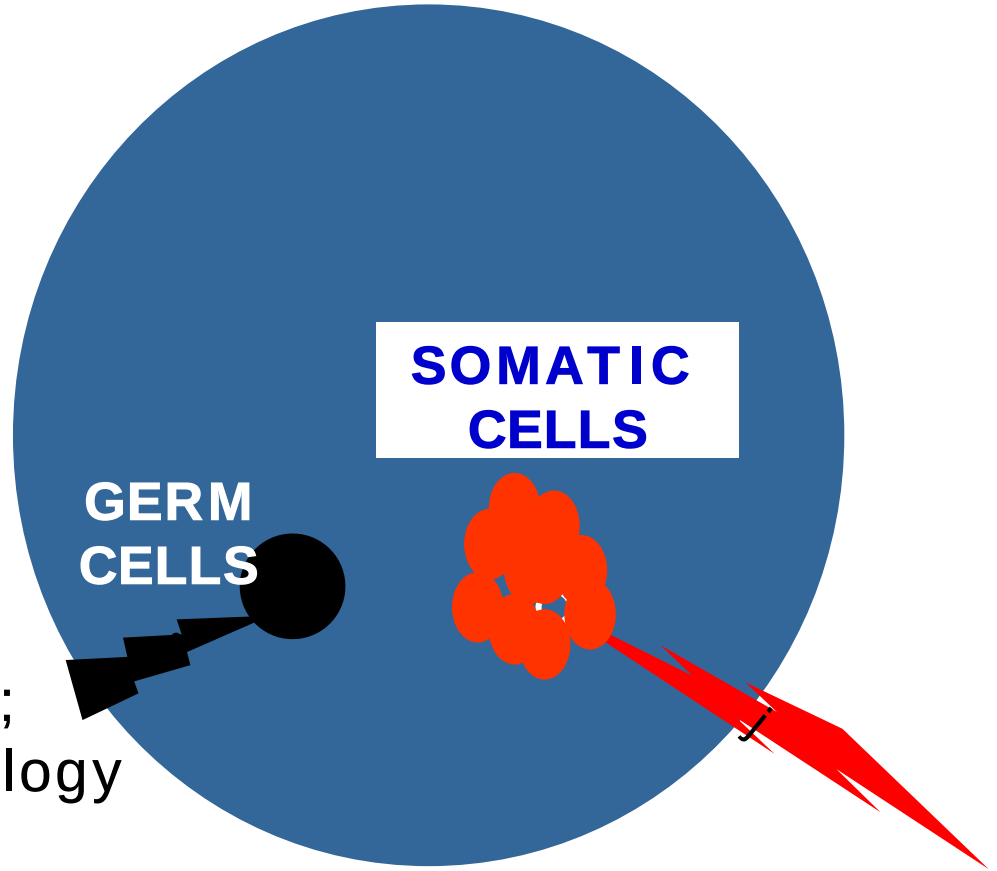




(Modified from Mason, Bautista & Gilsanz, *Blood Revs* **21**: 267, 2007)


Schizogonic cycle
AA
Invasion
AS
Selective sickling
of parasitised
AS red cells
Selective phagocytosis
of sickled parasitised AS red cells

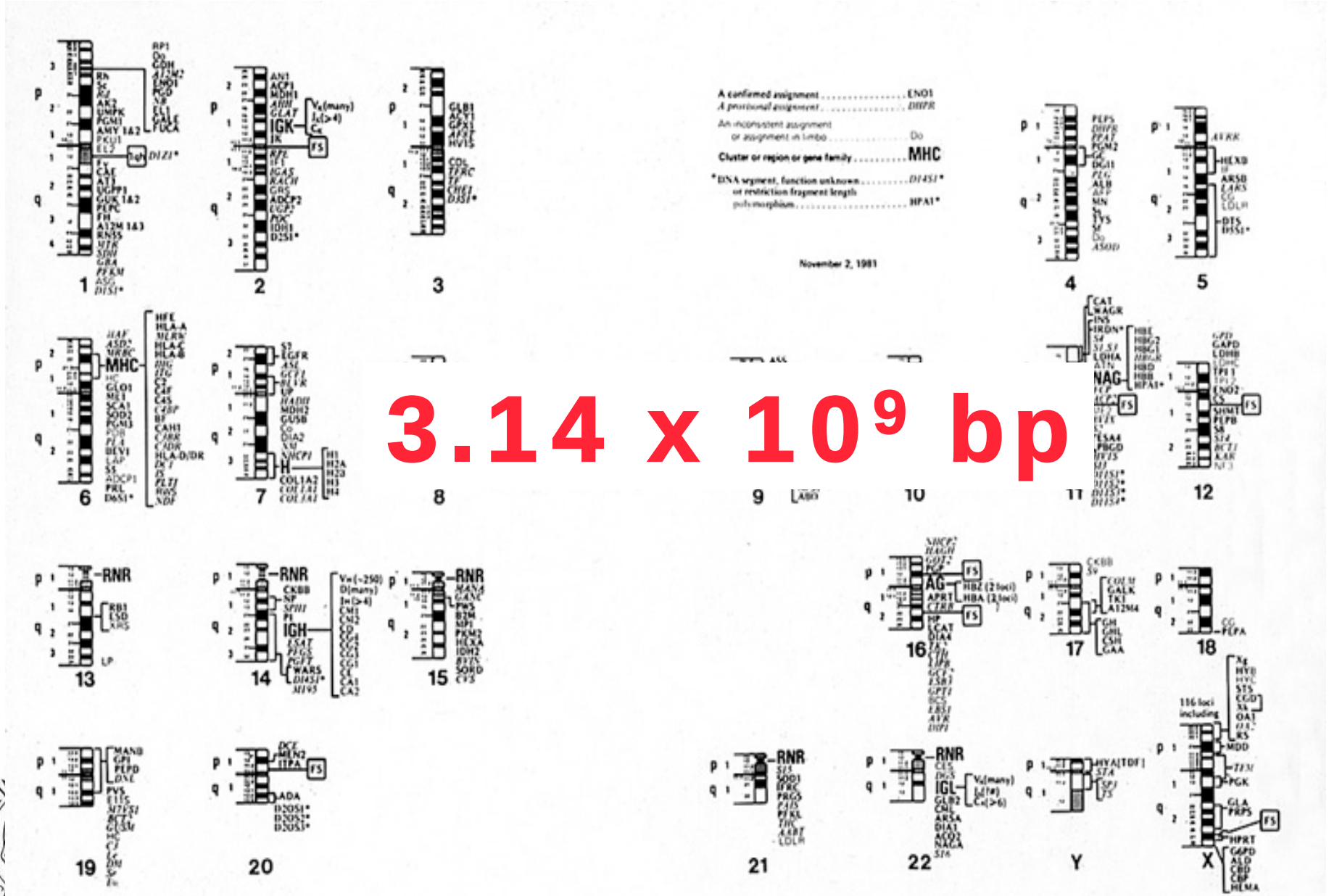
GENETICS AND INHERITANCE



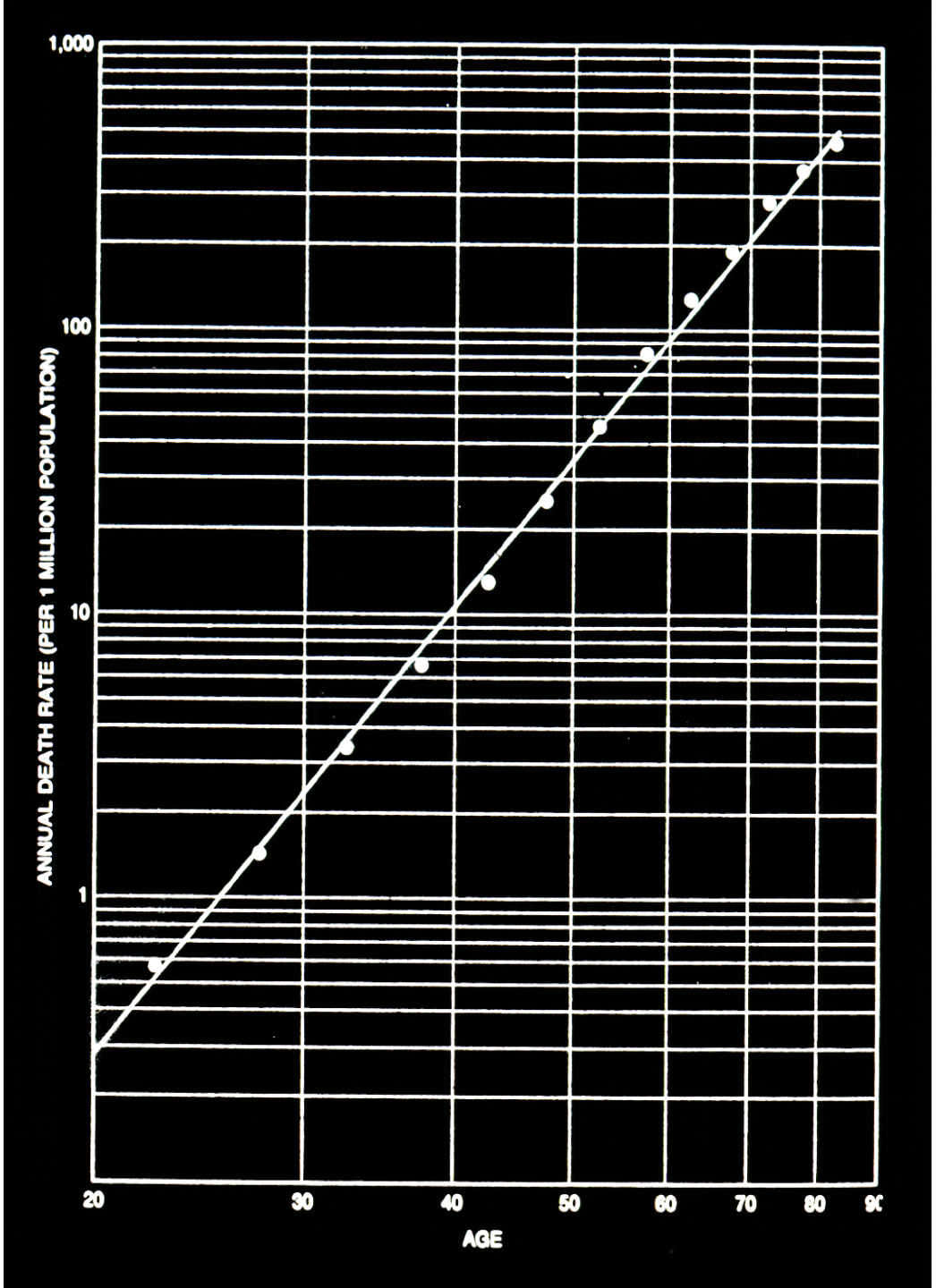
No pathology; potential pathology in offspring

No transmission to offspring; **potential pathology in host**

THE HUMAN GENOME



The incidence of cancer is a function of age



From Richard Peto, *Scientific American*, 1968

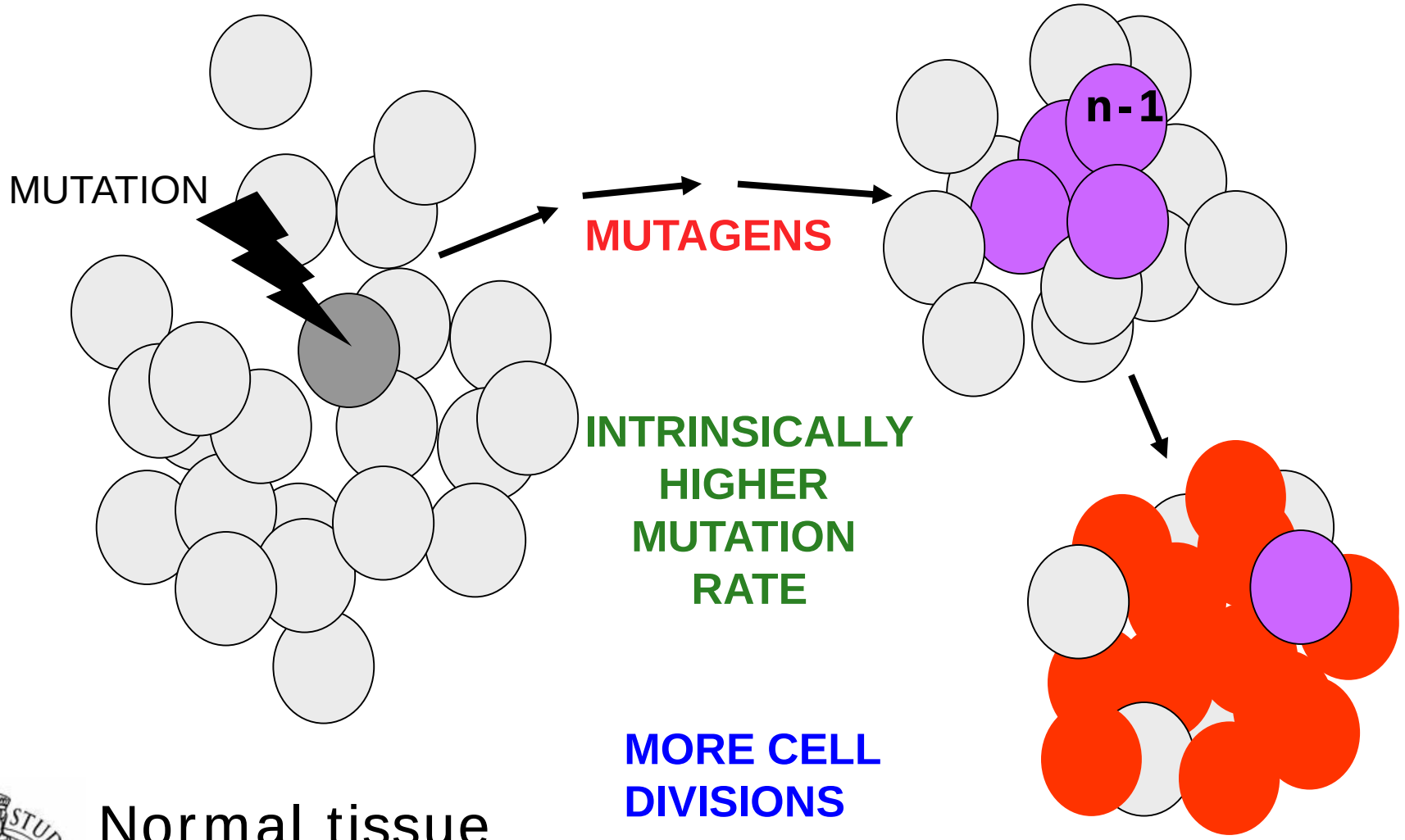
review article

Mutation selection and the natural history of cancer

John Cairns*

Survival of the rapidly renewing tissues of long-lived animals like man requires that they be protected against the natural selection of fitter variant cells (that is, the spontaneous appearance of cancer). This article discusses three possible protective mechanisms and shows how they could explain various features of the natural history of certain common cancers of man.

Inherited, acquired and environmental factors can favor/accelerate oncogenesis



SOUNDING BOARD

Causality and Chance in the Development of Cancer

Lucio Luzzatto, M.D., and Pier Paolo Pandolfi, M.D., Ph.D.

N ENGL J MED 373;1 NEJM.ORG JULY 2, 2015

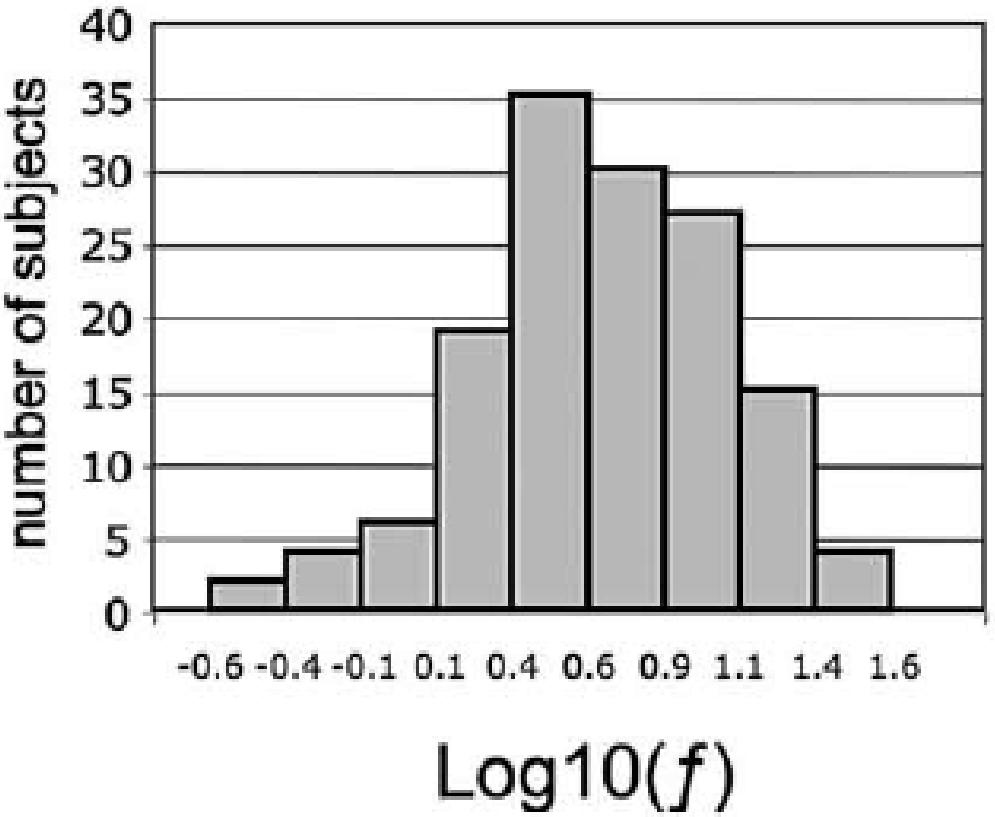
$$M = \mu D \quad (1)$$

$$M' = (\mu + \mu_e)D \quad (2)$$

The log normal distribution of values of f,

a proxy for μ,

is a (probably polygenic) quantitative trait



(From Rondelli et al., *PLoS ONE* **8**: e54046, 2013)

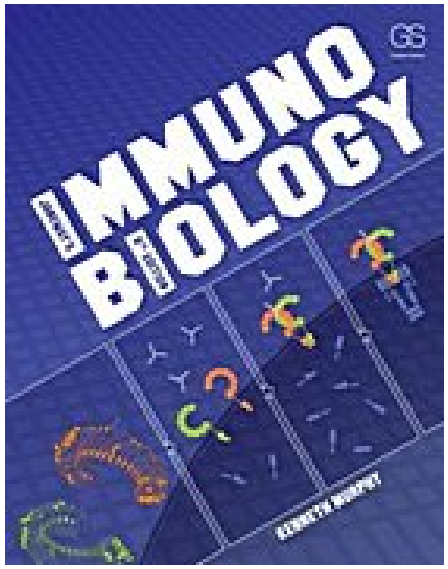
WHY μ IS IMPORTANT WITH RESPECT TO CANCER

- A higher value of μ may correlate with a higher risk of cancer
- If there are *n* mutational steps in the formation of a tumor the estimated risk will be proportional to μ^n
- It may be possible to identify the genetic determinants of μ

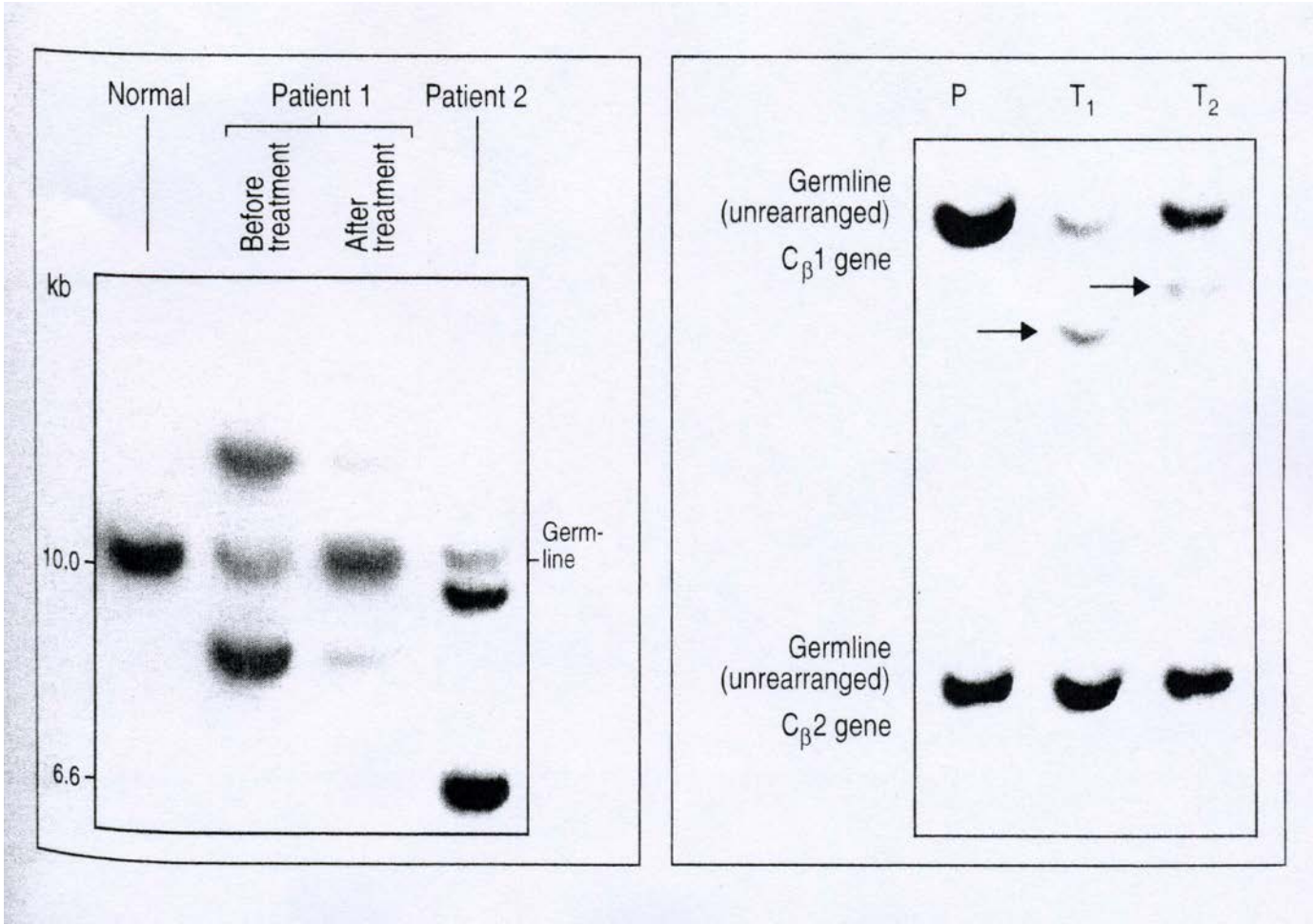
μ may be significantly affected by environmental factors

- One might find agents that decrease μ

A POPULATION OF LYMPHOID CELLS CAN BE SHOWN UNAMBIGUOUSLY TO BE CLONAL

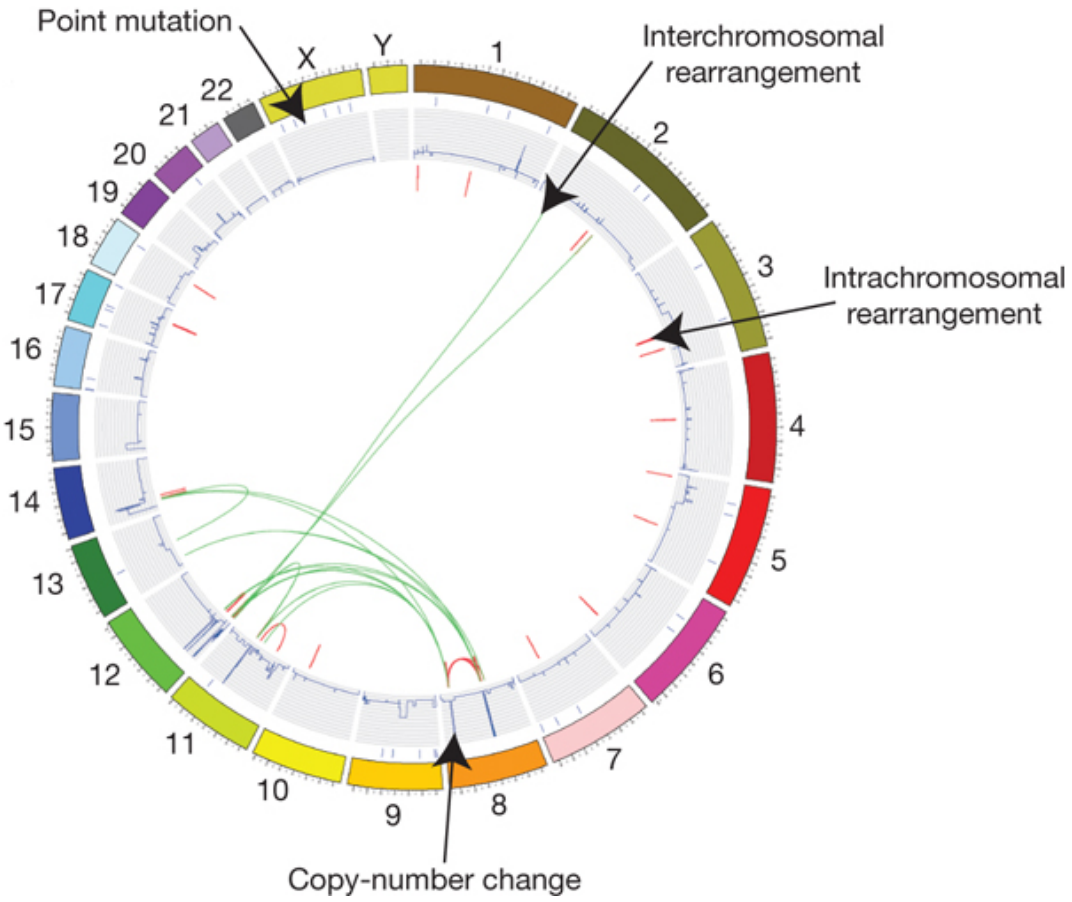


(From L Foroni, T J Vulliamy, L Luzzatto, 1987 *Janeway's IMMUNOBIOLOGY, 2007)*



THE LANDSCAPE OF SOMATIC MUTATIONS PRESENT IN A SINGLE CANCER GENOME.

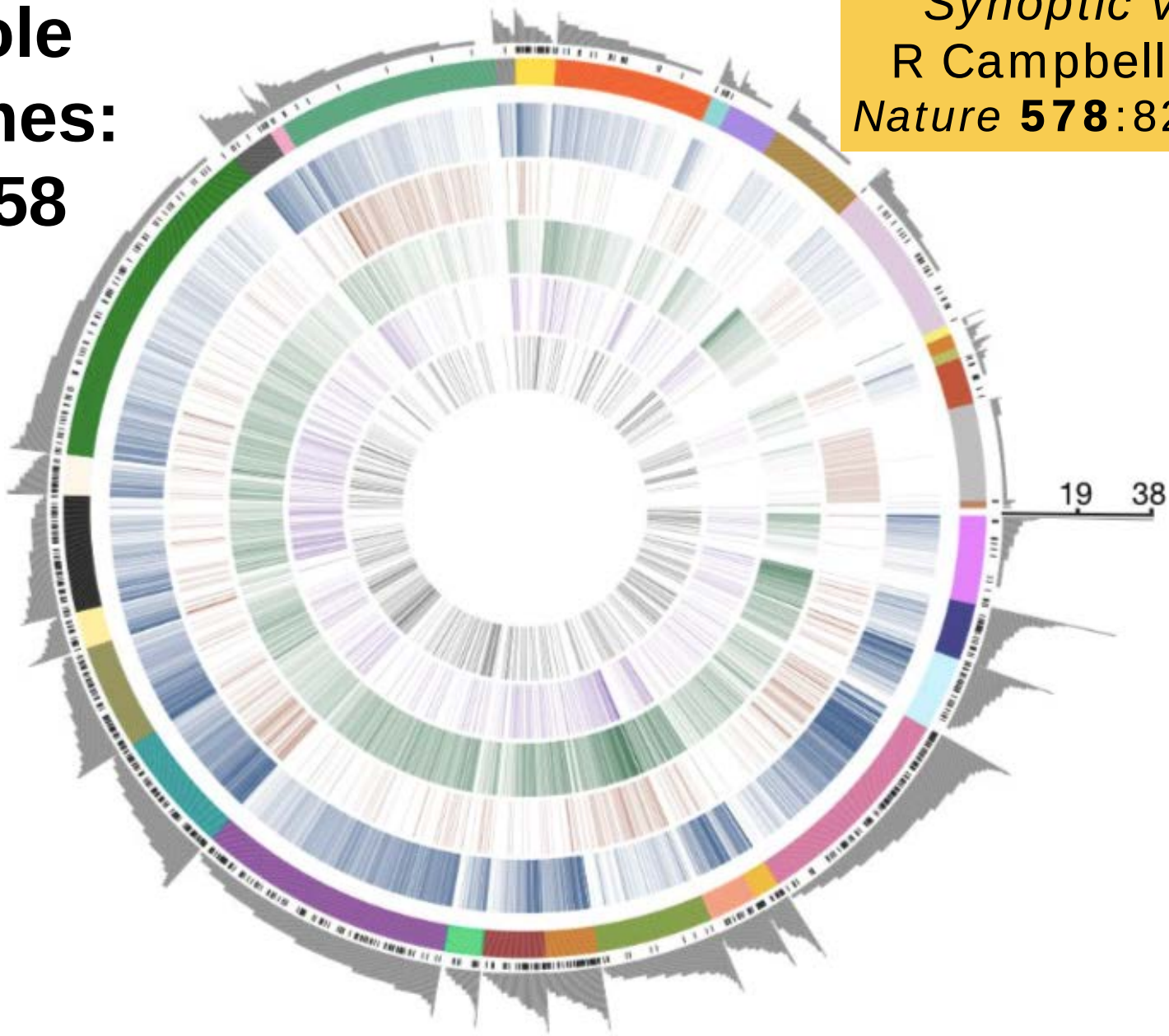
Stratton *et al.*
Nature **458**,719-724
(2009)

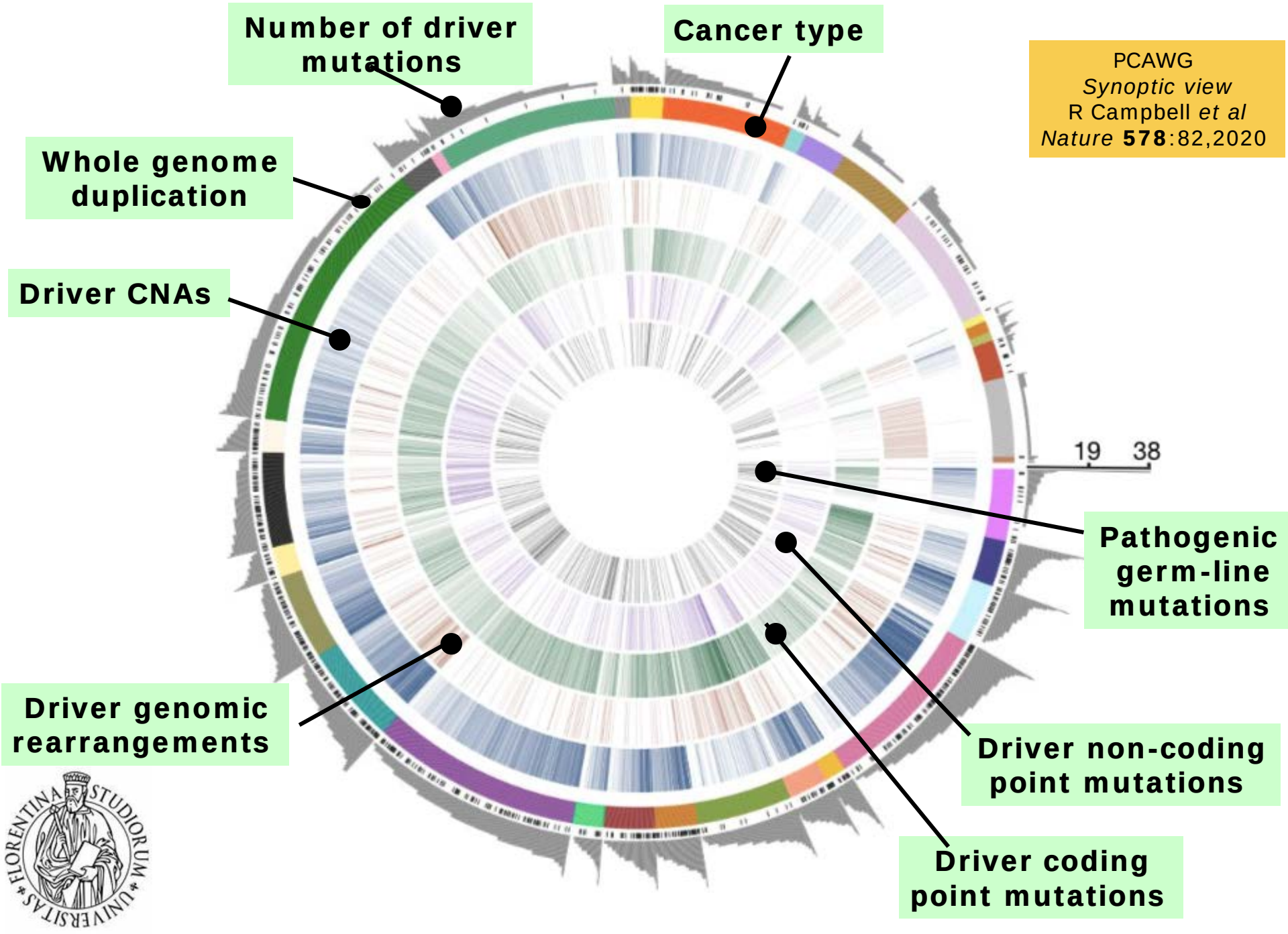


The ultimate nosography of a tumor is the set of somatic mutations in that tumor

Pan-cancer analysis of whole genomes: n = 2658

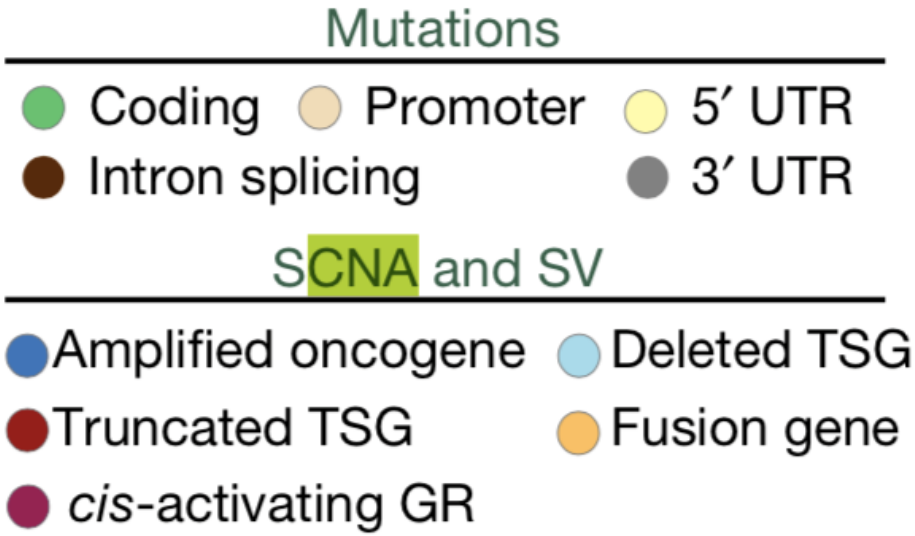
PCAWG
Synoptic view
R Campbell *et al*
Nature **578**:82,2020

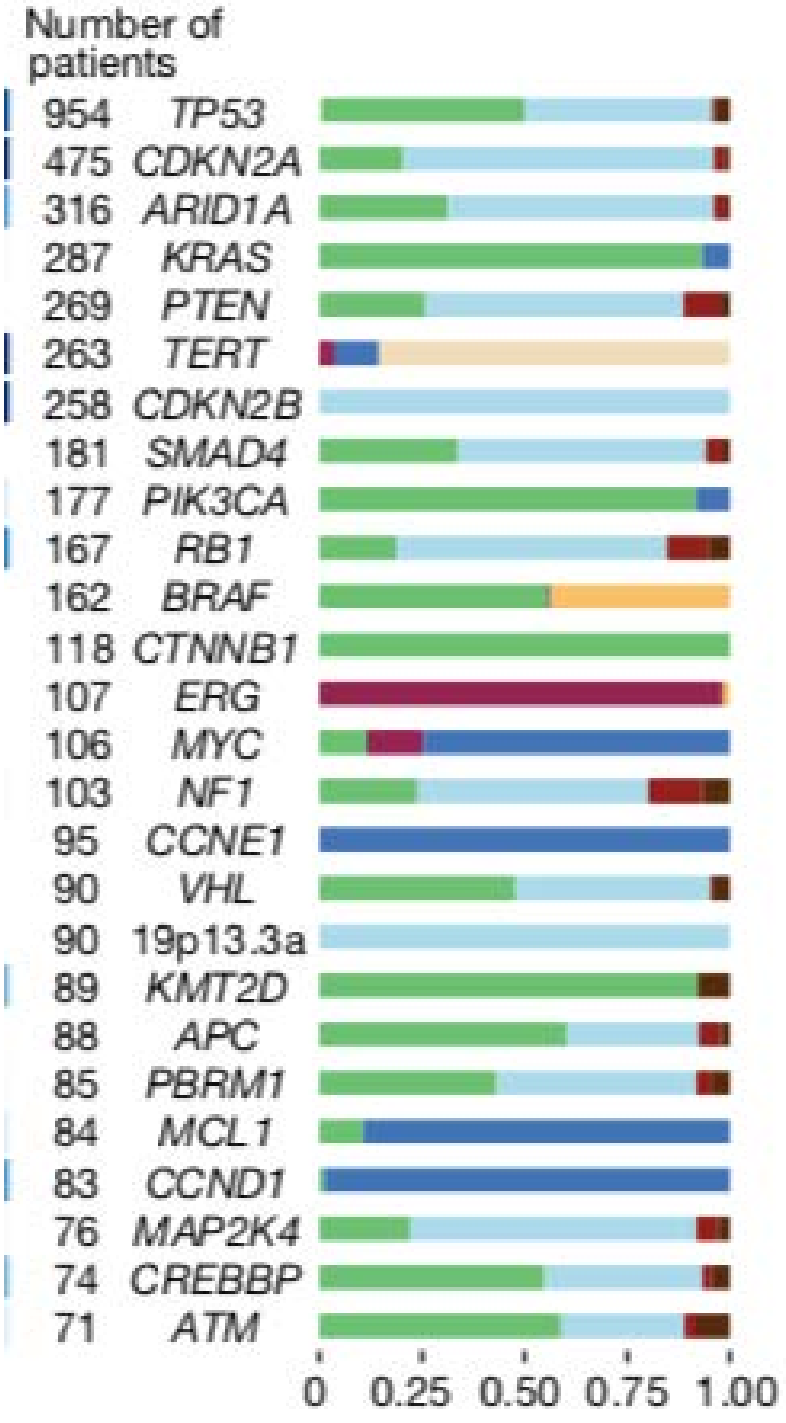




PCAWG
Synoptic view
R Campbell *et al*
Nature **578**:82,2020

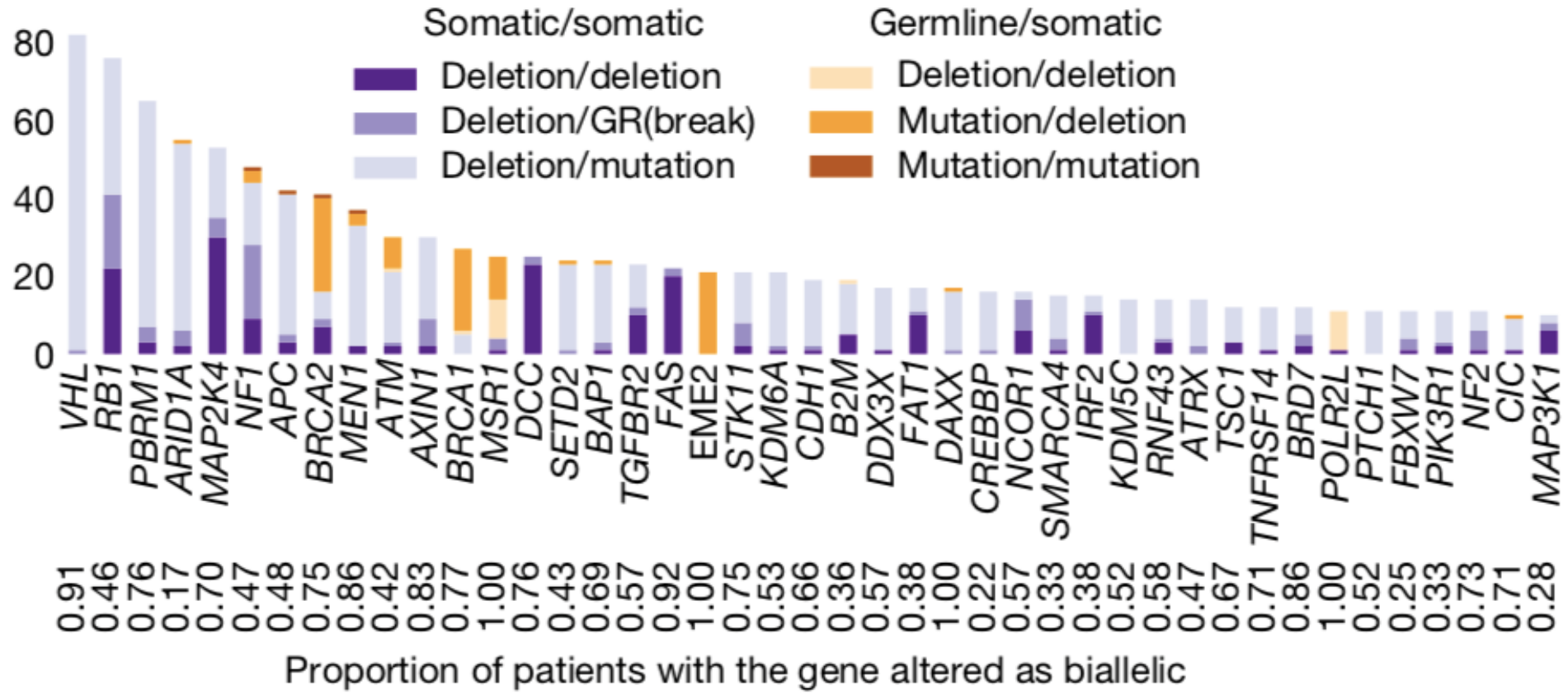
Different proportions of various types of mutation are found in individual 'cancer genes'





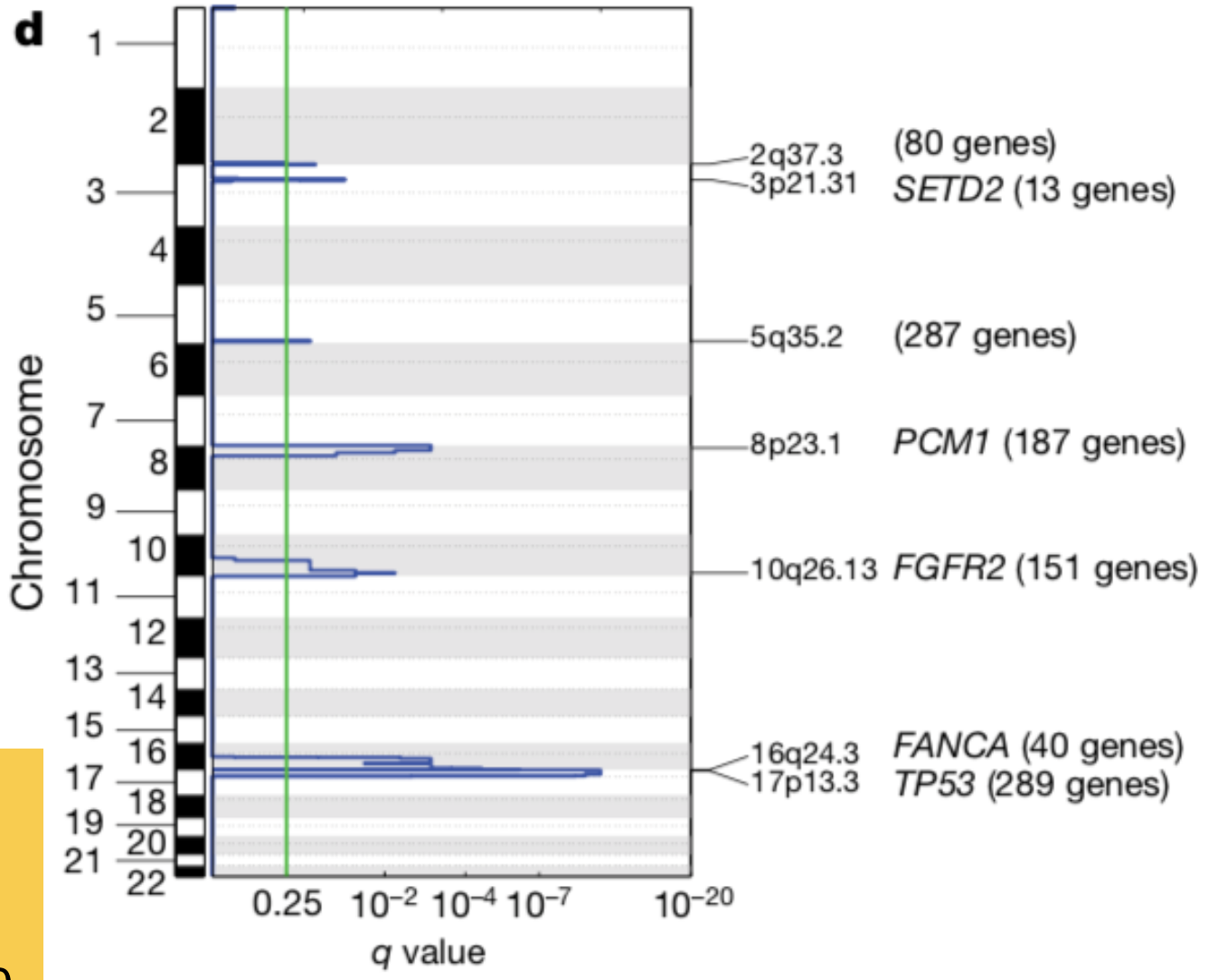
PCAWG
Synoptic view
R Campbell *et al*
Nature **578**:82,2020

Bi-allelic mutations in tumor-suppressor genes are not rare



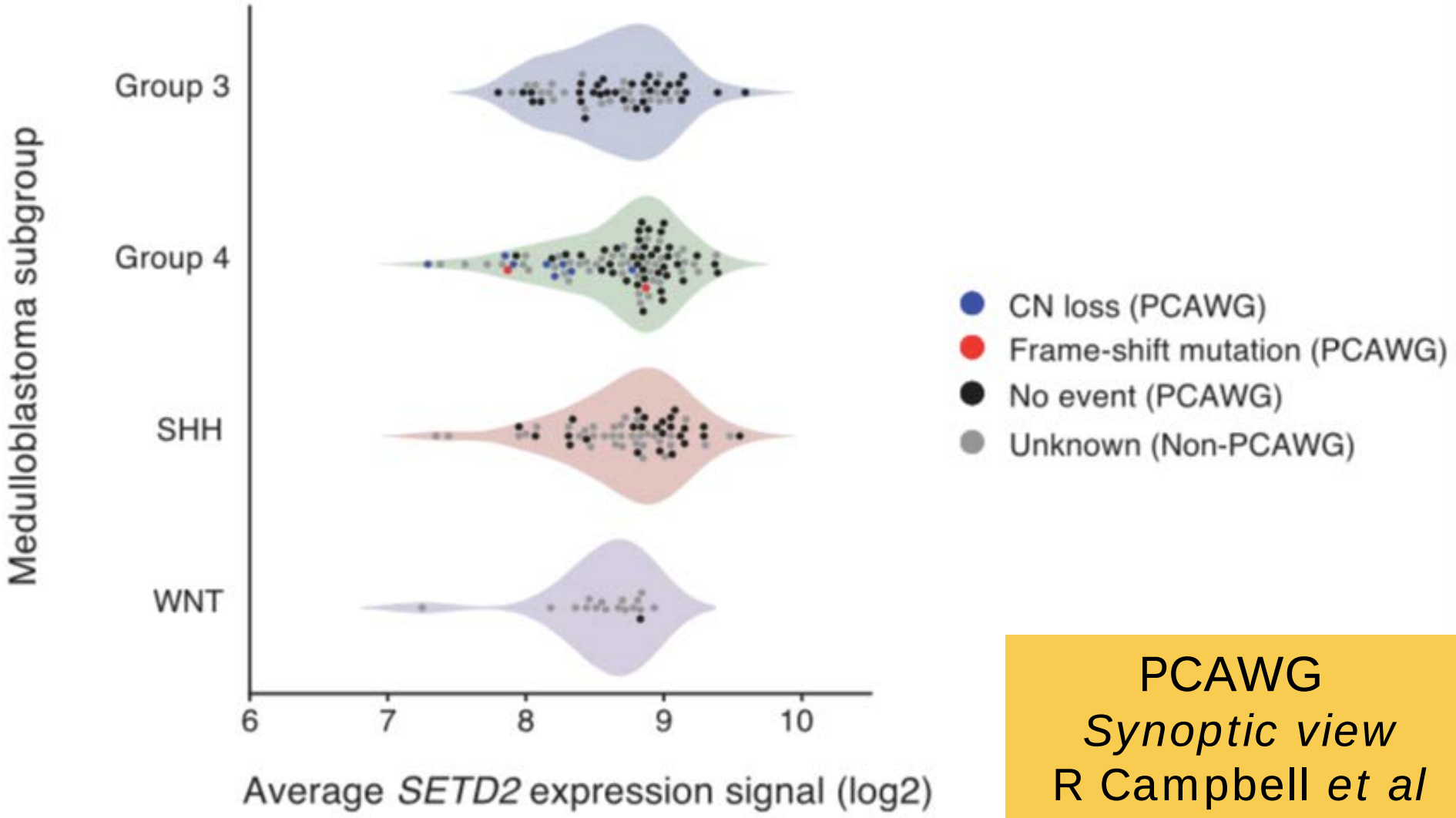
PCAWG
Synoptic view
R Campbell *et al*
Nature **578**:82,2020

COPY NUMBER LOSSES IN TUMORS WITH NO DETECTED DRIVER MUTATIONS



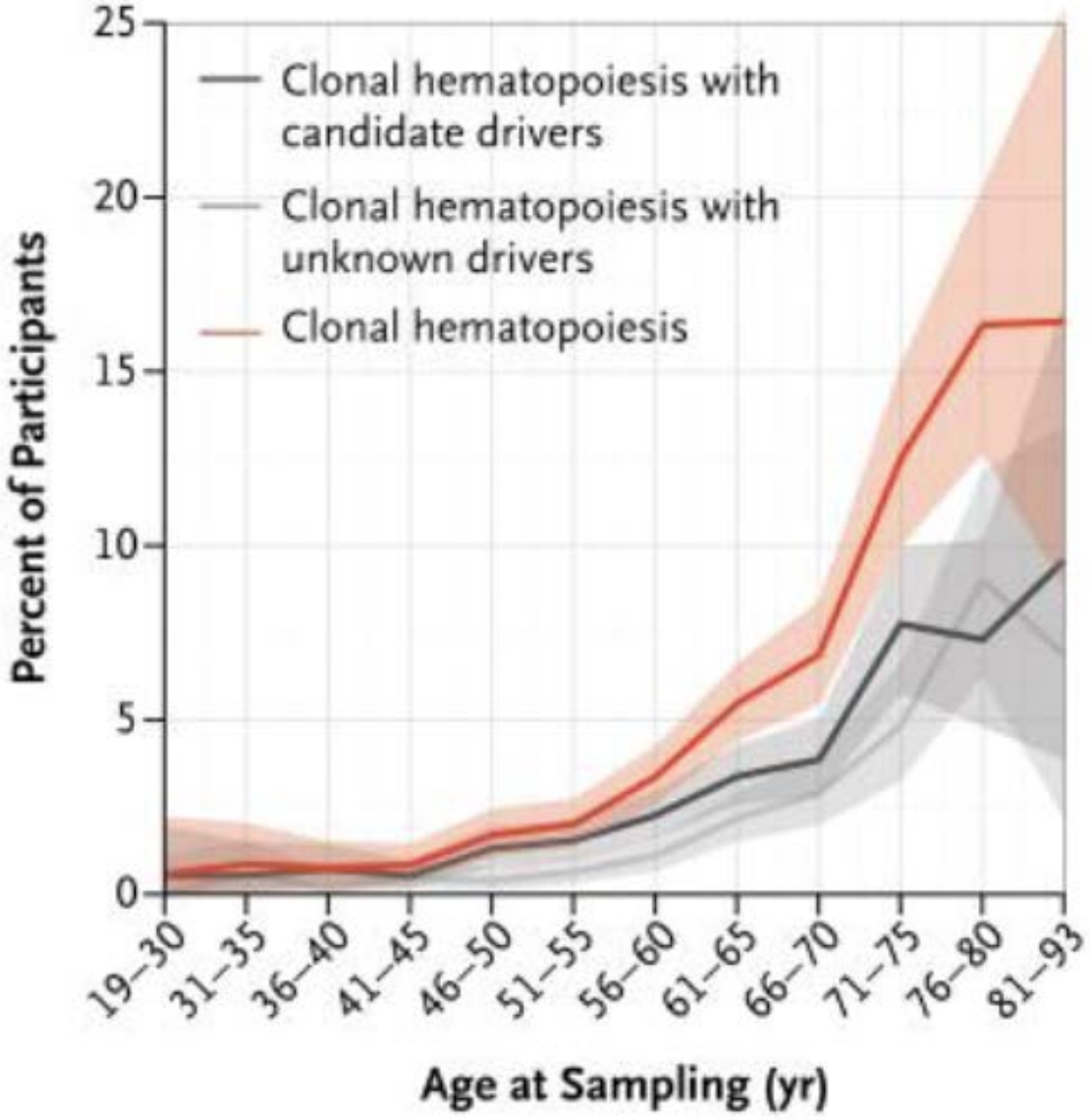
PCAWG
Synoptic view
R Campbell *et al*
Nature **578**:82,2020

Inactivating mutations of SETD2, *a gene that encodes a histone lysine methyltransferase, are found in a subset of medulloblastoma tumors*



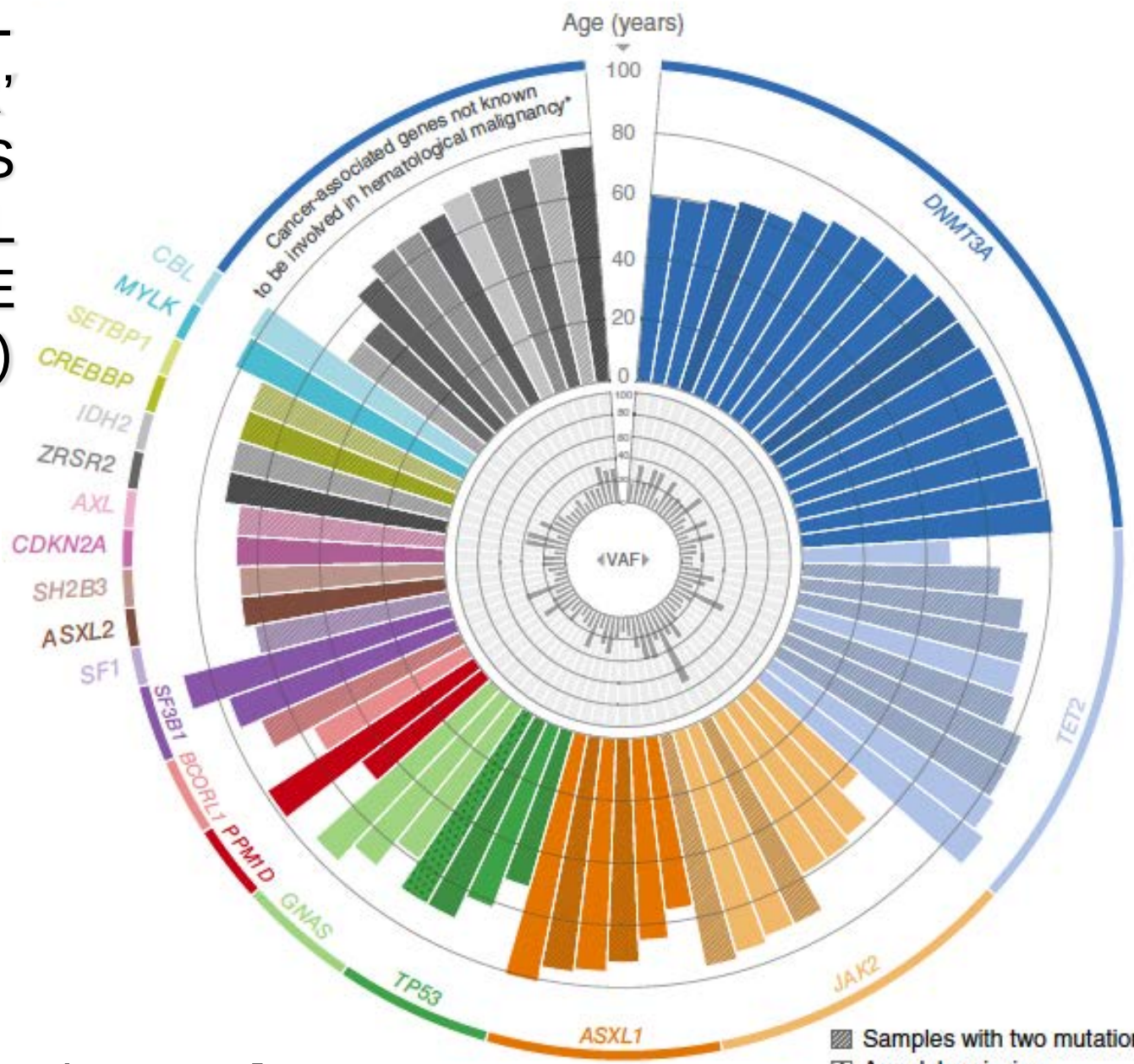
PCAWG
Synoptic view
R Campbell *et al*
Nature **578**:82,2020

Clones
that can be identified
within hematopoiesis
increase with age



(From Genovese etal.,
NEJM **371**:2477,2014)

‘BLOOD-CANCER’ MUTATIONS IN NORMAL PEOPLE (58/2728)



(From Xie *et al.*, *Nature Medicine* **20**:1472,2014)

Acta
Haematologica

Original Paper

Acta Haematol 2023;146:14–24
DOI: 10.1159/000527284

Received: March 16, 2022
Accepted: September 8, 2022
Published online: November 7, 2022

Clonal Megakaryocyte Dysplasia with Isolated Thrombocytosis Is a Distinct Myeloproliferative Neoplasm Phenotype

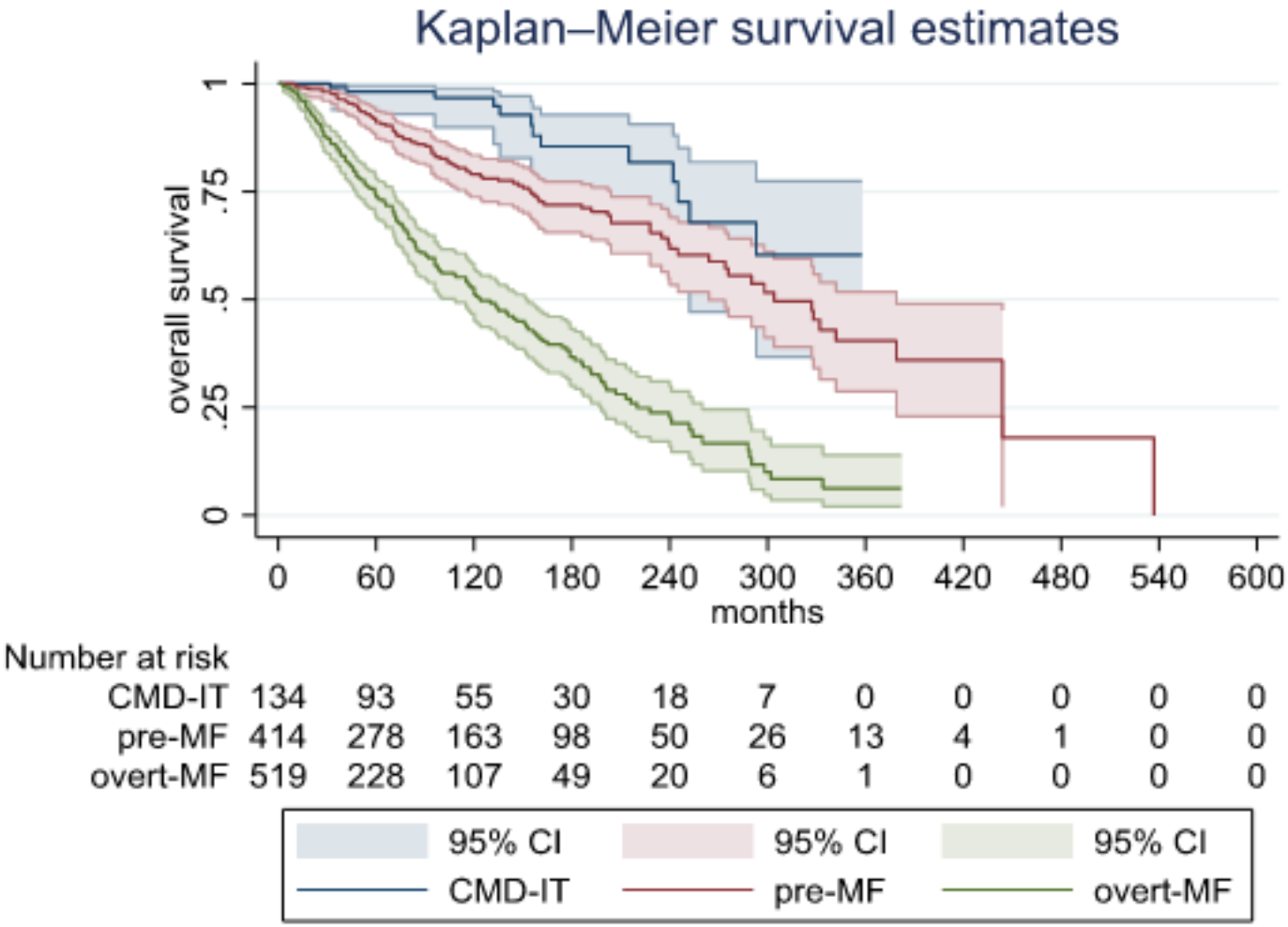
Giovanni Barosi[a] Rita Campanelli[a] Margherita Massa[b] Paolo Catarsi[a]
Adriana Carolei[a] Carlotta Abbà[a] Laura Villani[a] Umberto Magrini[a]
Giuliana Gregato[c] Francesco Bertolini[c] Annalisa de Silvestri[d]
Robert Peter Gale[e] Vittorio Rosti[a]

Table 2. Somatic mutations and cytogenetics of CMD-IT and PMF stratified as pre-MF and overt-MF

	CMD-IT	Pre-MF	Overt-MF	Comparison of CMD-IT with pre-MF	Overall (*p* value for trend)
JAK2^{V617F} mutants, *N* (%)	*N* = 126 71 (55.5)	*N* = 373 286 (76.7)	*N* = 417 264 (63)	OR = 0.30 (0.23–0.55) *p* < 0. 001	0.82
JAK2^{V617F} VAF %, median (IQR)	*N* = 67 27 (15–44)	*N* = 215 37 (22–66)	*N* = 193 48 (31–71)	*p* = 0.002	<0.001
JAK2^{V617F} mutants with VAF >50%, *N* (% of JAK2V^{617F} mutants)	*N* = 71 12 (16.9)	*N* = 281 106 (37.7)	*N* = 262 120 (45.4)	OR = 0.25 (0.13–0.47) *p* < 0.001	<0.001
CALR mutation, *N* (%)	*N* = 124 47 (38)	*N* = 372 49 (13)	*N* = 416 84 (20)	OR = 3.97 (2.43–6.38) *p* < 0.001	0.015
CALR-type 1, *N* (% of *CALR* mutated)	*N* = 44 25 (57)	*N* = 45 36 (80)	*N* = 80 62 (77.5)	*p* = 0.023	0.024
MPL mutation, *N* (%)	*N* = 129 6 (5)	*N* = 363 13 (4)	*N* = 417 29 (7)	OR = 1.31 (0.49–3.54) *p* = 0.58	0.103
Triple negative, *N* (%)	*N* = 129 4 (3)	*N* = 363 22 (6)	*N* = 417 40 (10)	OR = 0.49 (0.17–1.47) *p* = 0.21	0.006
NGS-detected HMR mutations, *N* (%)	*N* = 46 6 (13)	*N* = 105 17 (16)	*N* = 87 27 (31)	OR = 0.77 (0.28–2.11) *p* = 0.62	0.011
Abnormal cytogenetics, *N* (%)	*N* = 46 6 (13)	*N* = 144 35 (24)	*N* = 140 62 (44)	*OR = 0.46* (0.18-1.19) *p* = 0.11	0.001

(From Barosi et al., *Acta Haematol* **146**:14,2023)

CMD-IT *(Clonal Megakaryocytic Dysplasia with Isolated Thrombocytosis) has a clinical course significantly different from that of other myeloproliferative dirorders*



(From Barosi et al., *Acta Haematol* **146**:14,2023)

blood
Special Report

Genomic profiling for clinical decision making in myeloid neoplasms and acute leukemia

Eric J. Duncavage,[1] Adam Bagg,[2] Robert P. Hasserjian,[3] Courtney D. DiNardo,[4] Lucy A. Godley,[5] Ilaria Iacobucci,[6] Siddhartha Jaiswal,[7] Luca Malcovati,[8] Alessandro M. Vannucchi,[9] Keyur P. Patel,[10] Daniel A. Arber,[11] Maria E. Arcila,[12] Rafael Bejar,[13] Nancy Berliner,[14] Michael J. Borowitz,[15,16] Susan Branford,[17] Anna L. Brown,[18] Catherine A. Cargo,[19] Hartmut Döhner,[20] Brunangelo Falini,[21] Guillermo Garcia-Manero,[22] Torsten Haferlach,[23] Eva Hellström-Lindberg,[24] Annette S. Kim,[25] Jeffery M. Klco,[6] Rami Komrokji,[26] Mignon Lee-Cheun Loh,[27] Sanam Loghavi,[10] Charles G. Mullighan,[6] Seishi Ogawa,[28] Attilio Orazi,[29] Elli Papaemmanuil,[30] Andreas Reiter,[31] David M. Ross,[32] Michael Savona,[33] Akiko Shimamura,[34] Radek C. Skoda,[35] Francesc Solé,[36] Richard M. Stone,[37] Ayalew Tefferi,[38] Matthew J. Walter,[39] David Wu,[40] Benjamin L. Ebert,[41] and Mario Cazzola[42]

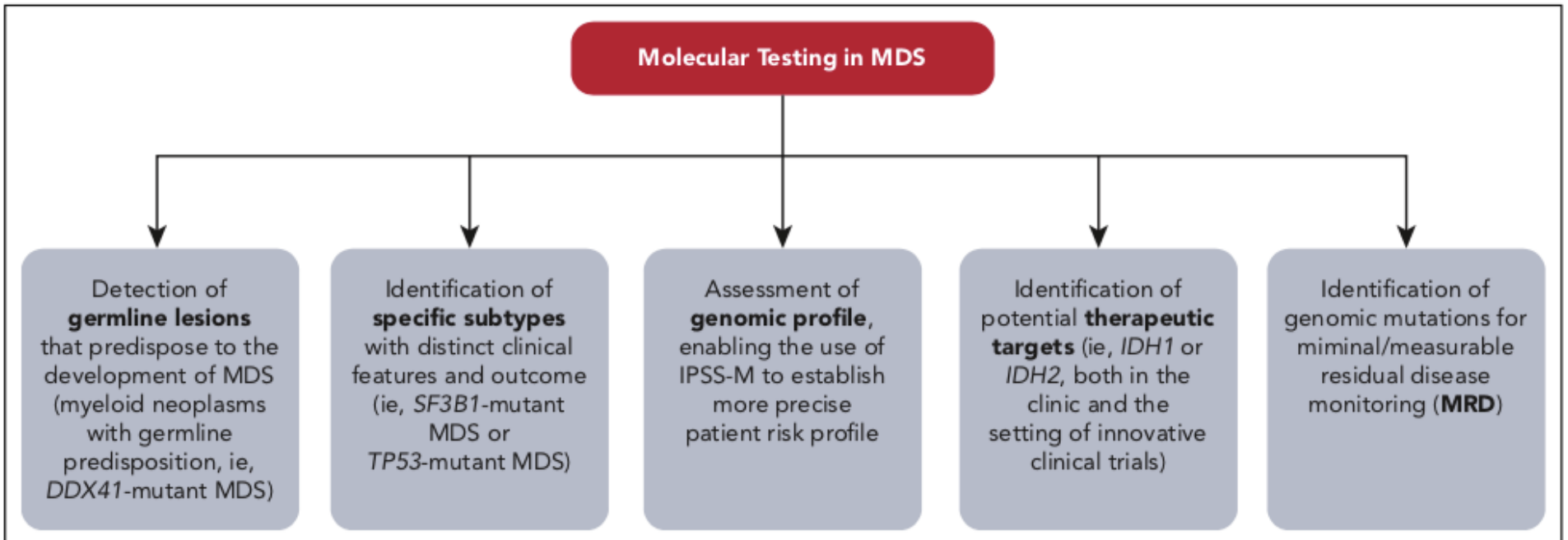
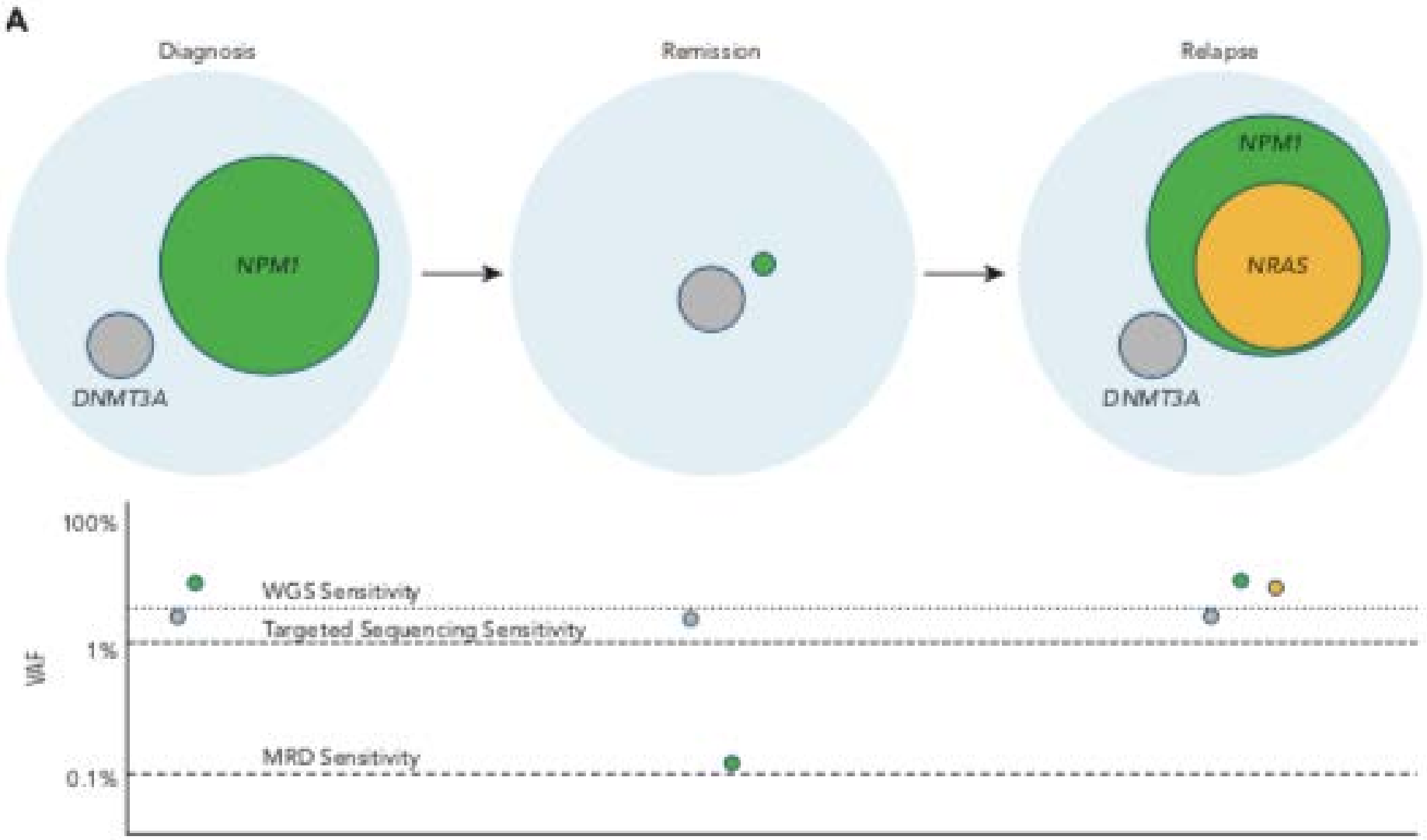


Table 2. Gene mutations in myeloid neoplasms and leukemia indicated for clinical testing

Indication	Single gene mutations	Structural variants*
MDS, MDS/MPN, cytopenia	*ASXL1, BCOR, BCORL1, CBL, CEBPA, CSF3R, DDX41, DMNT3A, ETV6, ETNK1, EZH2, FLT3-ITD, FLT3-TKD, GATA2, GNB1, IDH1, IDH2, JAK2, KIT, KRAS, KMT2A-PTD, NF1, NPM1, NRAS, PHF6, PPM1D, PRPF8, PTPN11, RAD21, RUNX1, SAMD9†, SAMD9L†, SETBP1, SF3B1, SRSF2, STAG2, TET2, TP53, U2AF1, UBA1, WT1, ZRSR2*	
MPN and mastocytosis‡	*ASXL1, CALR, CBL, CSF3R, DNMT3A, EZH2, IDH1, IDH2 JAK2§, KIT, KRAS, MPL, NRAS, PTPN11, RUNX1, SETBP1, SF3B1, SH2B3, SRSF2, TET2, U2AF1, ZRSR2*	*BCR::ABL1§*
Eosinophilia	*ASXL1, CBL, DNMT3A, EZH2, KRAS, NRAS, RUNX1, SF3B1, SRSF2, STAT5B, TET2, U2AF1*	*BCR::ABL1§, FGFR1::R, FLT3::R, JAK2::R, PDGFRA::R, PDGFRB::R*
AML	**Genes required for diagnosis and risk stratification:** *ASXL1, BCOR, CEBPA, DDX41, EZH2, FLT3-ITD§, FLT3-TKD§, IDH1§, IDH2§, NPM1, RUNX1, SF3B1, SRSF2, STAG2, TP53, U2AF1, ZRSR2* **Additional genes recommended to test for at diagnosis and for use in disease monitoring:** *ANKRD26, BCORL1, BRAF, CBL, CSF3R, DNMT3A, ETV6, GATA2, JAK2, KIT, KRAS, NRAS, NF1, PHF6, PPM1D, PTPN11, RAD21, SETBP1, TET2, WT1*	*BCR::ABL1§, CBFB::MYH11, DEK::NUP214 MECOM::R, KMT2A::R, NUP98::R, RUNX1::RUNX1T1, PML::RARA§*
B-ALL	*CREBBP, CRLF2, FLT3, IDH1, IDH2, IKZF1, IL7R, JAK1, JAK2, JAK3, KMT2D, KRAS, NF1, NRAS, PAX5, PTPN11, SETD2, SH2B3, TP53*	*ABL1::R§, ABL2::R, CRLF2::R, CSF1R::R, DUX4::R, EPOR::R, ETV6::R, JAK2::R, KMT2A::R, MEF2D::R, NUTM1::R, PAX5::R, PDGFRA::R, PDGFRB::R, TCF3::R, ZNF384::R*
T-ALL	*DNMT3A, ETV6, EZH2, FBXW7, FLT3, IDH1, IDH2, IL7R, JAK1, JAK3, KRAS, MSH2, NOTCH1, NRAS, PHF6, PTEN, U2AF1, WT1*	*BCL11B::R, LMO2::R, MYB::R, NUP::ABL1, NUP214::R, STIL::R, TAL::R, TLX1::R, TLX3::R*

(From Duncavage et al., *Blood* **140**:2228,2022)

MONITORING TUMOR BURDEN IN MYELOID NEOPLASMS



(From Duncavage et al., *Blood* **140**:2228,2022)

FINANCIAL BURDEN OF MONITORING TUMOR BURDEN IN MYELOID NEOPLASMS

	Cost	Breadth	Depth	Sensitivity	Emerging Mutations	Structural Variants
WGS	$$$$			10%	+	+
Gene panel	$$			2%	+	+/–
MRD panel	$$$			~0.1%	+	–
Patient-specific MRD	$$$$			<0.1%	–	–

(From Duncavage et al., *Blood* **140**:2228,2022)

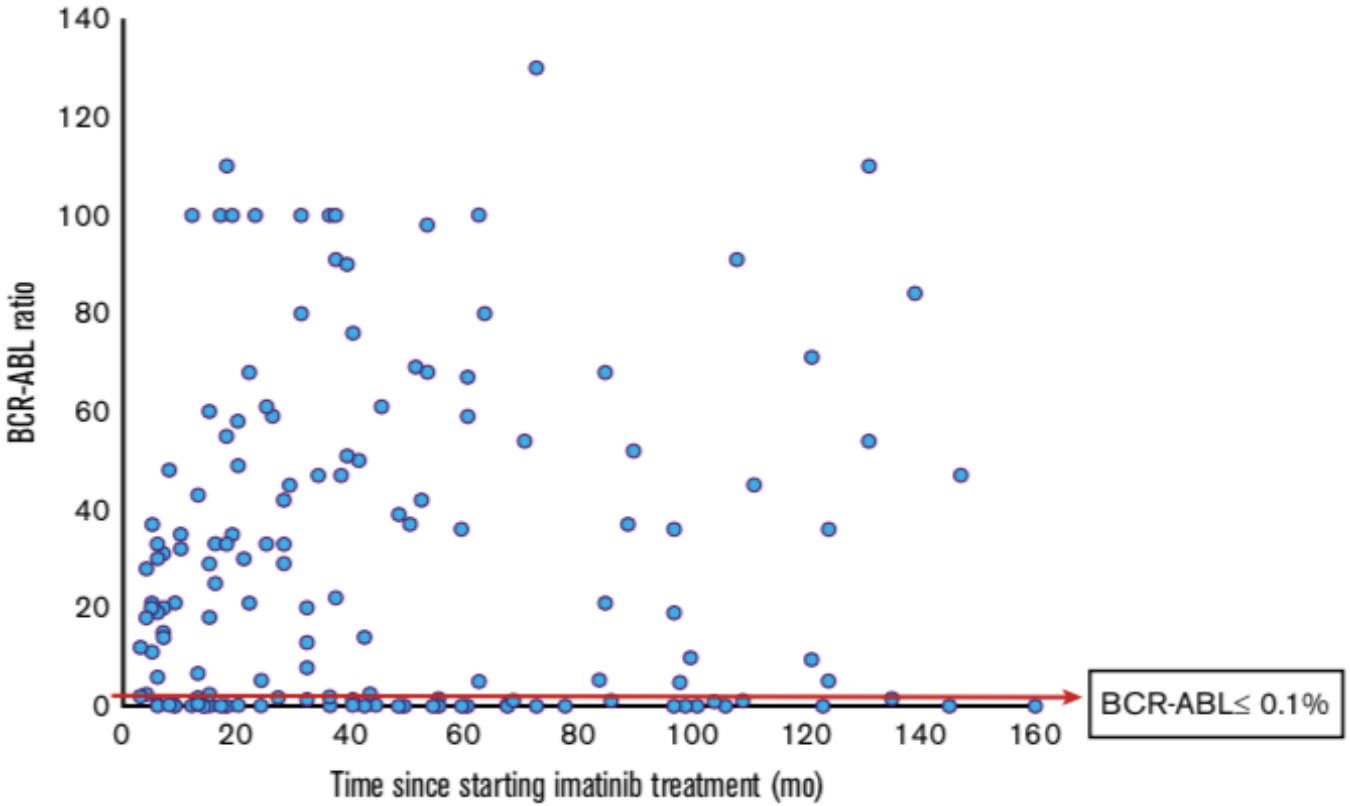
REGULAR ARTICLE

blood advances

Check for updates

Molecular response to imatinib in patients with chronic myeloid leukemia in Tanzania

The rate of ***major molecular response*** *to imatinib is rather low in patients with CML in Tanzania*

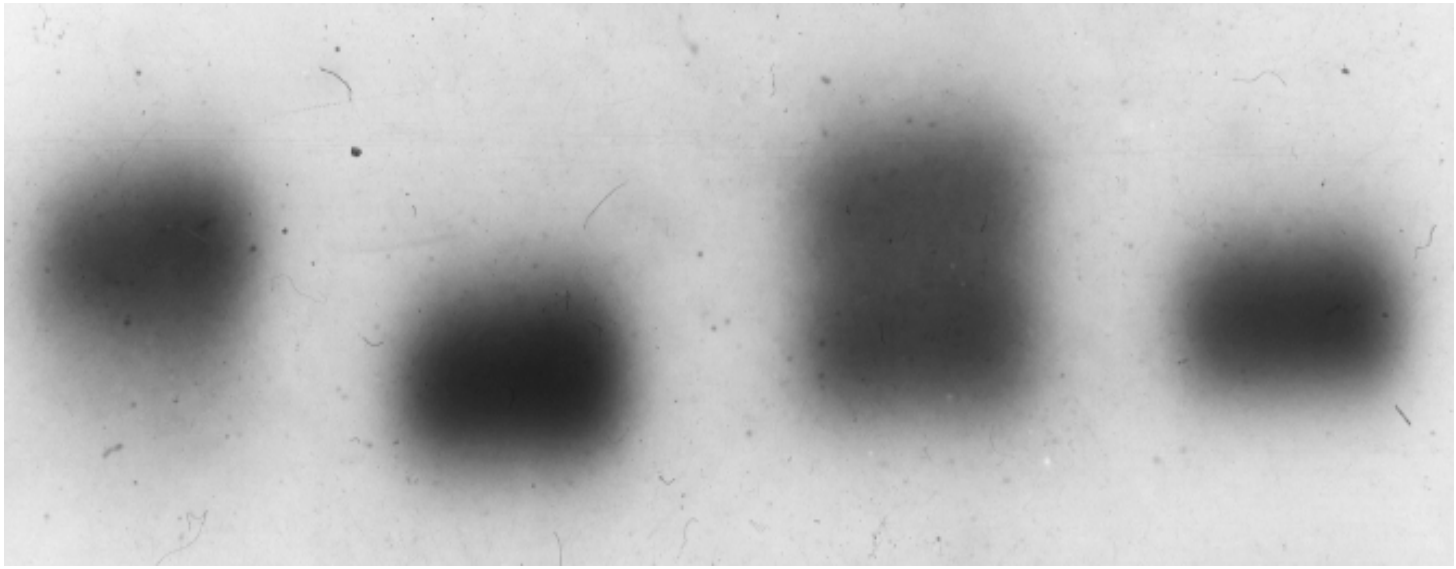


(From Nasser *et al.*, *Blood Advances* **5**:1403,2021)

Paroxysmal Nocturnal Hemoglobinuria: Evidence for Monoclonal Origin of Abnormal Red Cells

By S. B. Oni, B. O. Osunkoya and L. Luzzatto

Blood, Vol. 36, No. 2 (August), 1970

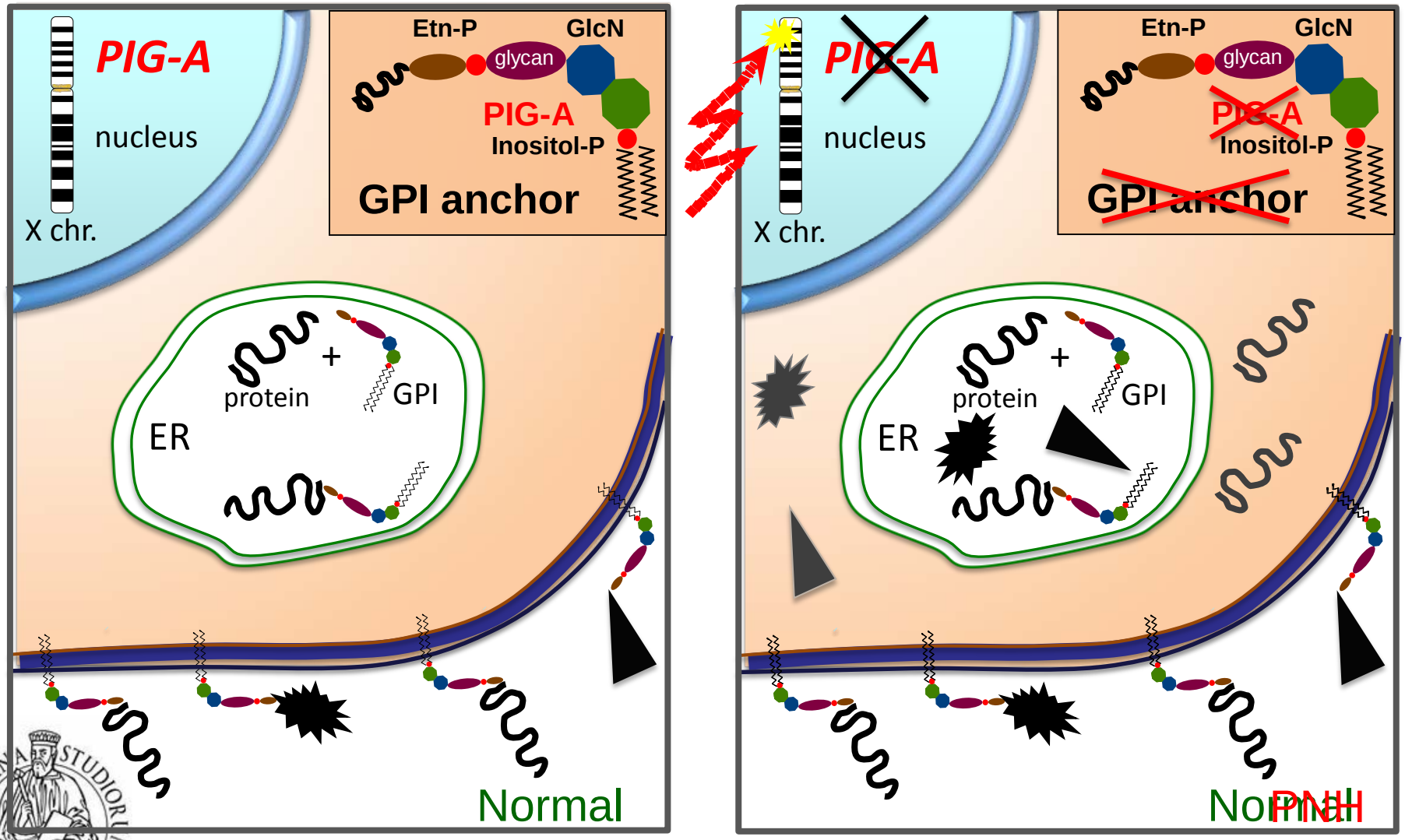


G6PD-A G6PD-B Whole RBC PNH RBC

Controls **Patient**

From the Subdepartment of Hematology, Department of Pathology, University College Hospital, Ibadan, Nigeria.

PATHOGENESIS OF A PNH CELL



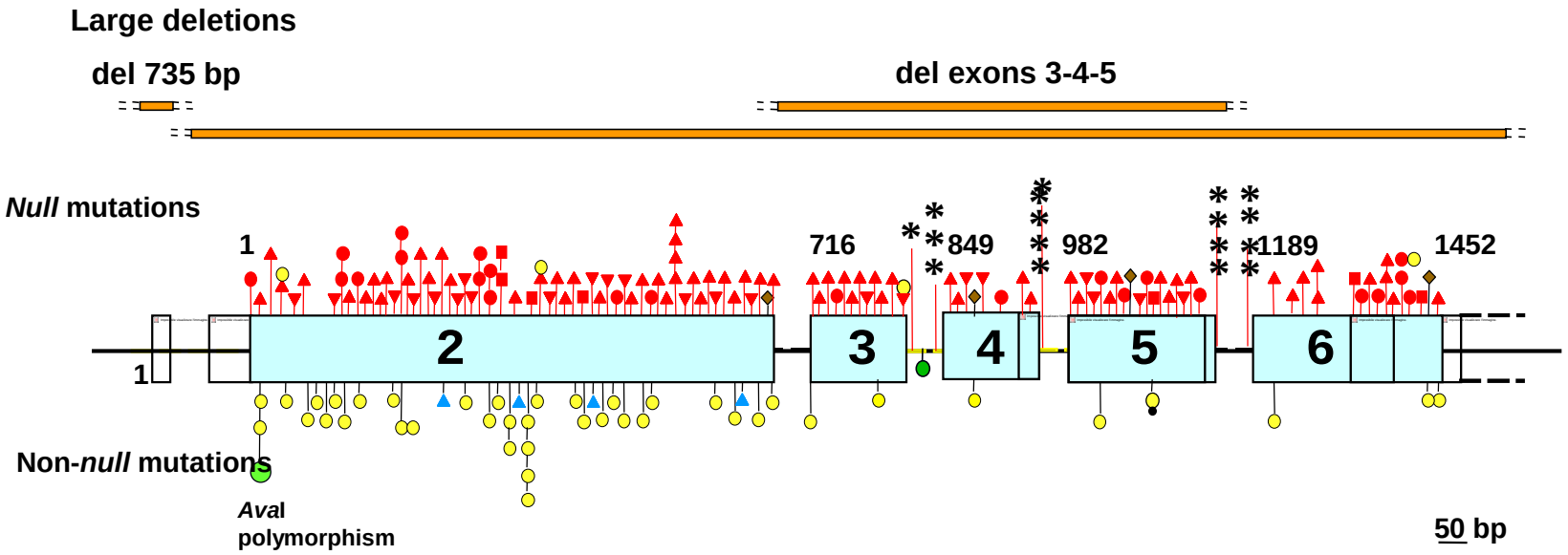
Haematopoietic stem cell

Cell, Vol. 73, 703–711, May 21, 1993, Copyright © 1993 by Cell Press

Deficiency of the GPI Anchor Caused by a Somatic Mutation of the *PIG-A* Gene in Paroxysmal Nocturnal Hemoglobinuria

Junji Takeda,* Toshio Miyata,*
Kazuyoshi Kawagoe,* Yoshiyasu Iida,*†
Yuichi Endo,‡ Teizo Fujita,‡ Minoru Takahashi,*
Teruo Kitani,§ and Taroh Kinoshita*
*Department of Immunoregulation
§Department of Internal Medicine
Research Institute for Microbial Diseases
Osaka University

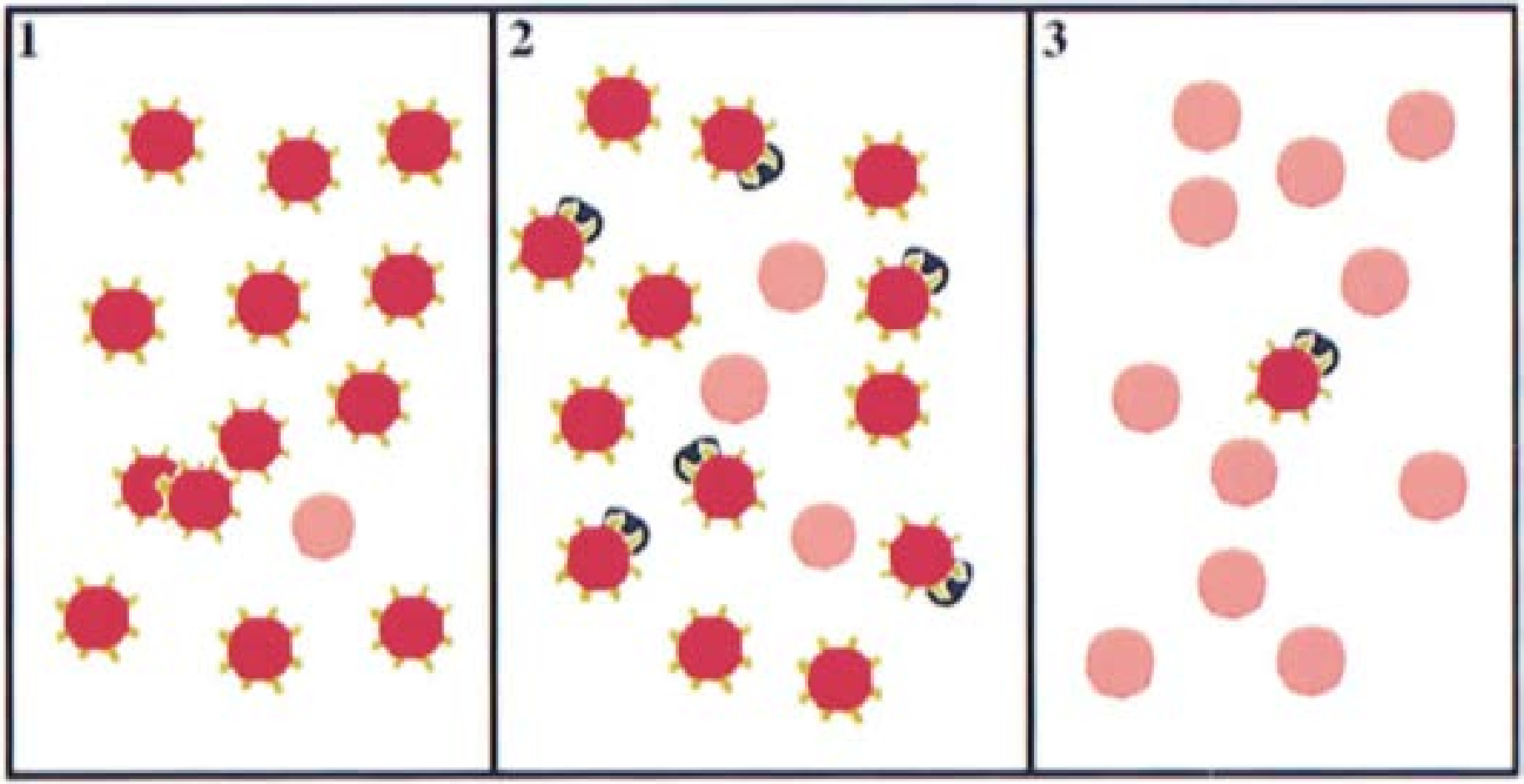
MUTATIONS IN THE *PIG-A* GENE



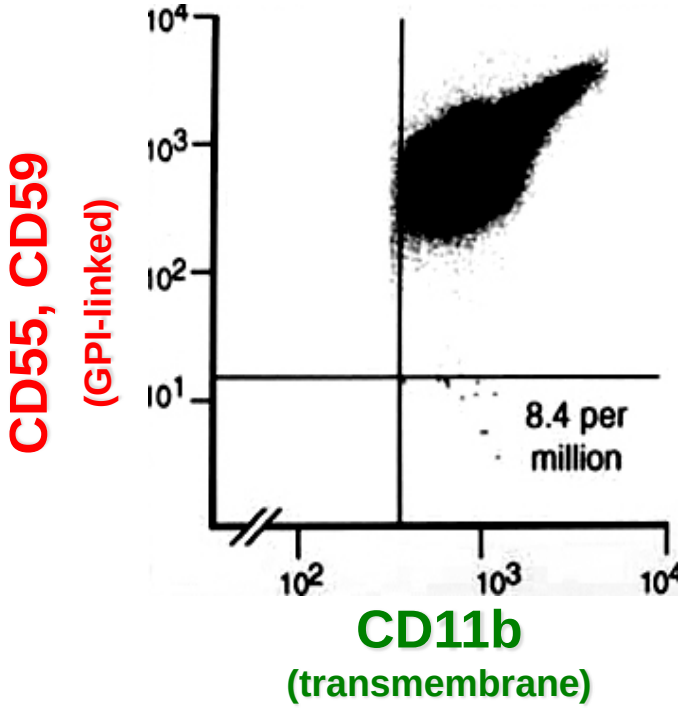
(From Luzzatto & Nafa (2000), Genetics of PNH, *in PNH and the GPI-linked Proteins,* N S Young and J Moss, Eds., Acad Press.)

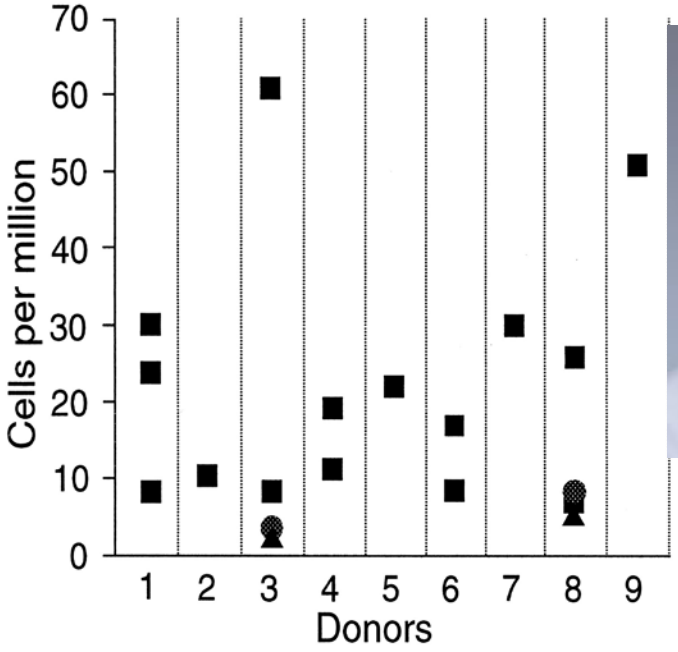
Cell, Vol. 88, 1–4, January 10, 1997, Copyright ©1997 by Cell Press

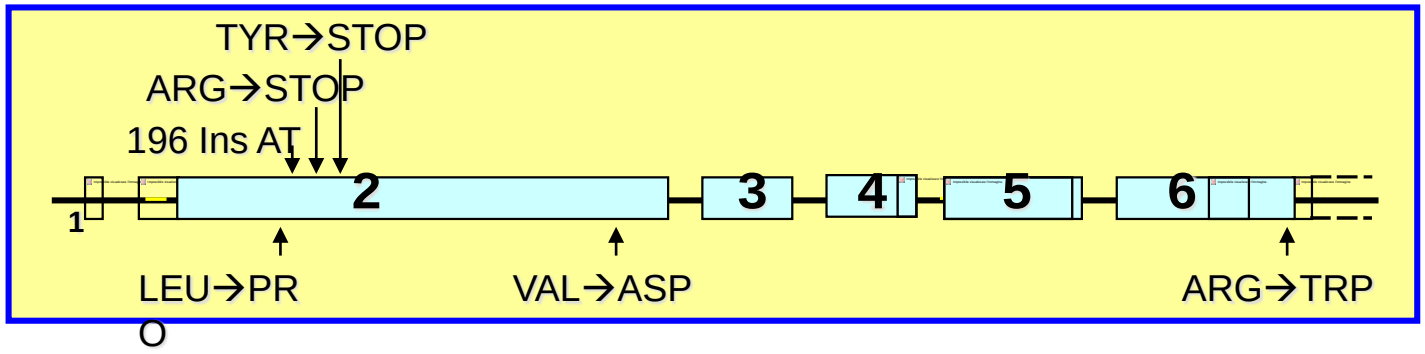
Somatic Mutations in Paroxysmal Nocturnal Hemoglobinuria: A Blessing in Disguise?



GPI(–) granulocytes are found in normal people

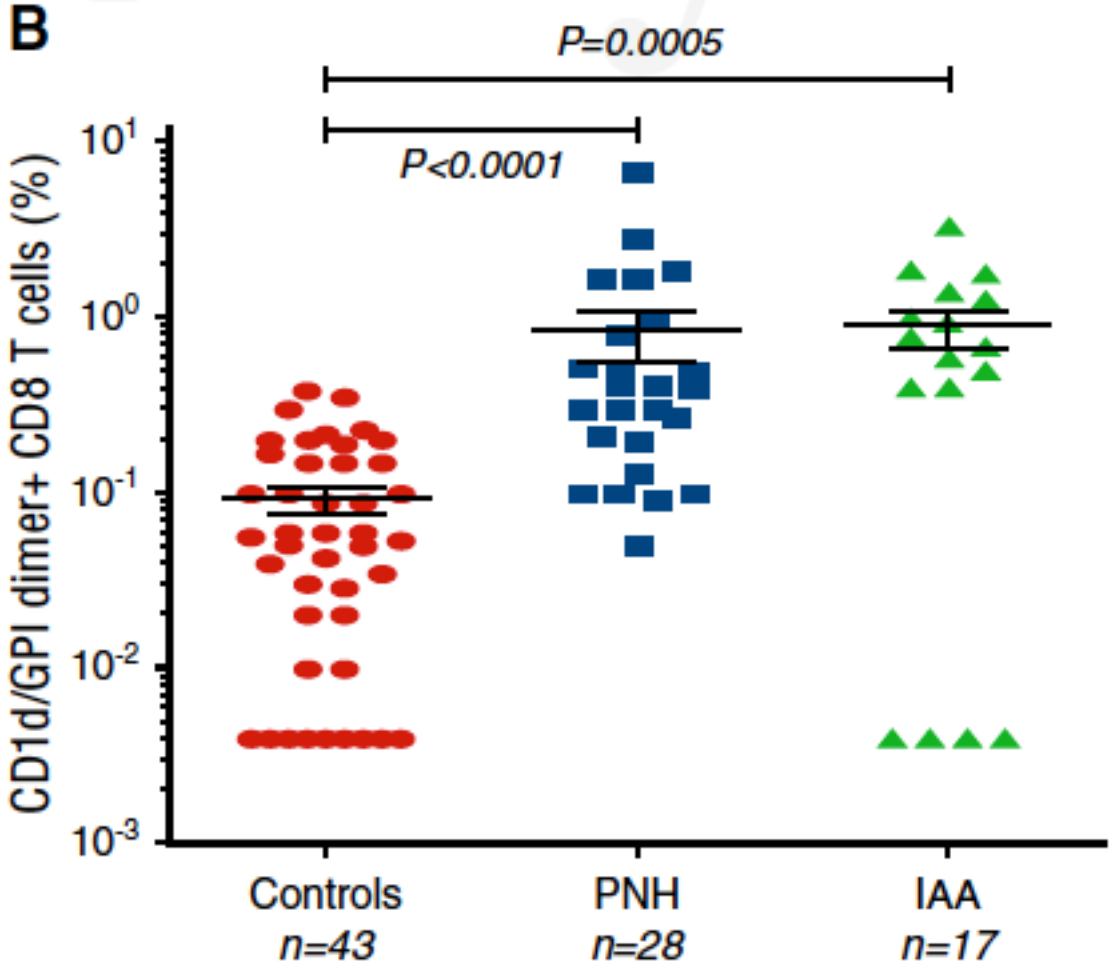






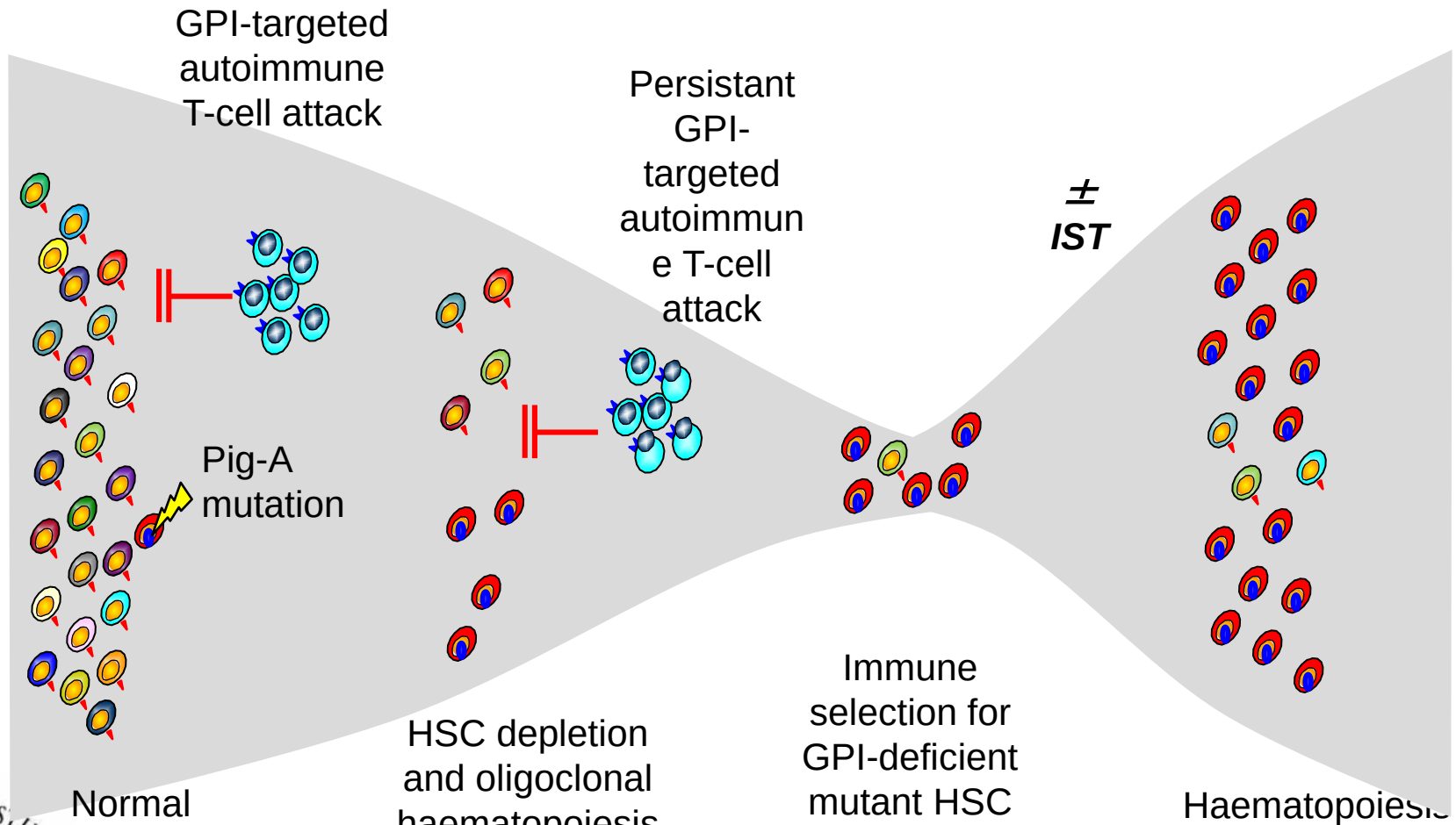
(From Araten et al.,*PNAS* **96**:5209,1999)

The abundance of GPI-specific T cells is similar in patients with PNH and in a subset of patients with idiopathic aplastic anaemia



(From Gargiulo et al., *Blood* **129**:388,2017)

APLASTIC ANEMIA WITH GPI-DEFICIENT MUTANT CLONES AND THE DEVELOPMENT OF PNH



(From Luzzatto & Risitano, *Brit J Haematology* **182**:758,2018)

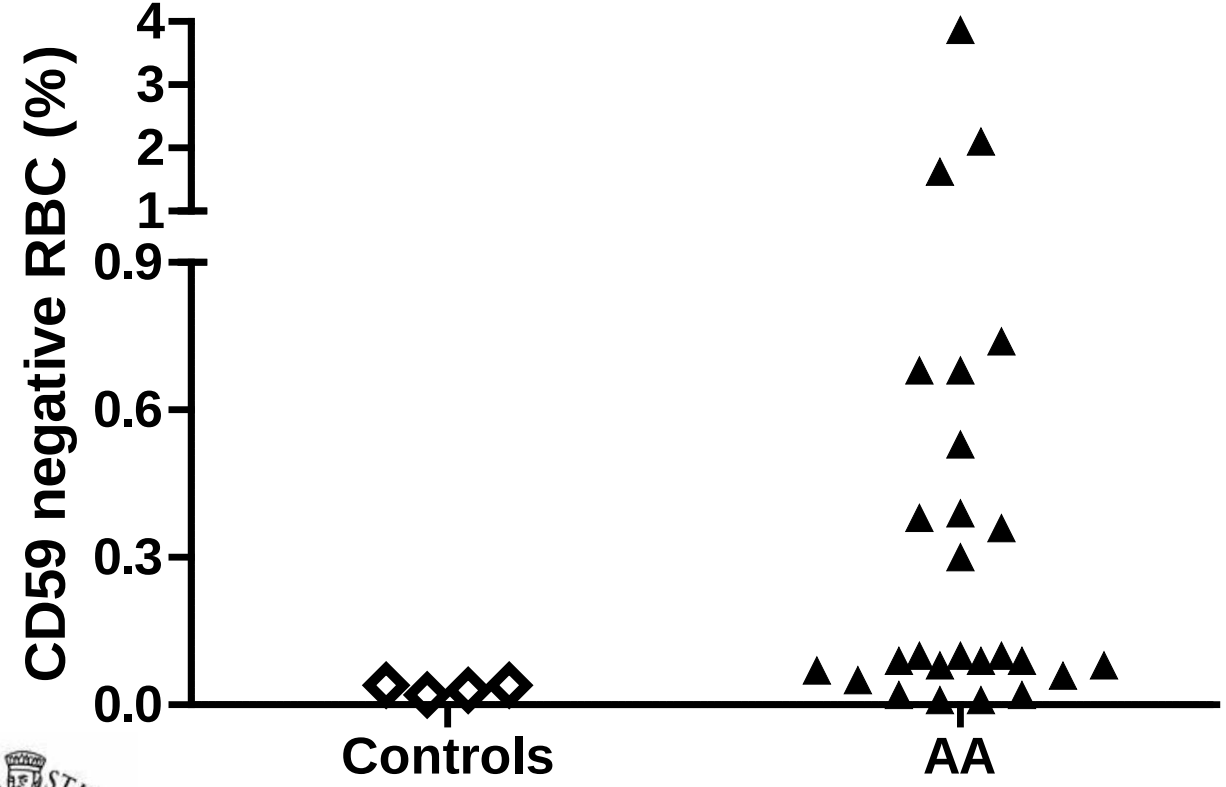
POPULATION GENETICS OF ORGANISMS/SOMATIC CELLS

Feature	Properties	In a population of organisms		In a population of somatic cells	
		Consequences	*Examples*	*Cosequences*	*Examples*
Genetic drift	Mutant neutral	Depends on population size	'Founder effects'	Clone may expand when normal cells few	Mutant clones in aplastic anaemia
Darwinian selection	Mutant has higher fitness	Mutant progeny may take over	Gene fixation	Clone will expand	Oncogenic mutations
	Conditional advantage	Depends on environment	*HBB*^{S} gene if malaria endemic	Depends on micro-environment	
	Convergent evolution	Increased frequency of independently arisen mutant genes	Genes of melano-genesis	Clones with mutations in different genes may expand	

POPULATION GENETICS OF ORGANISMS/SOMATIC CELLS

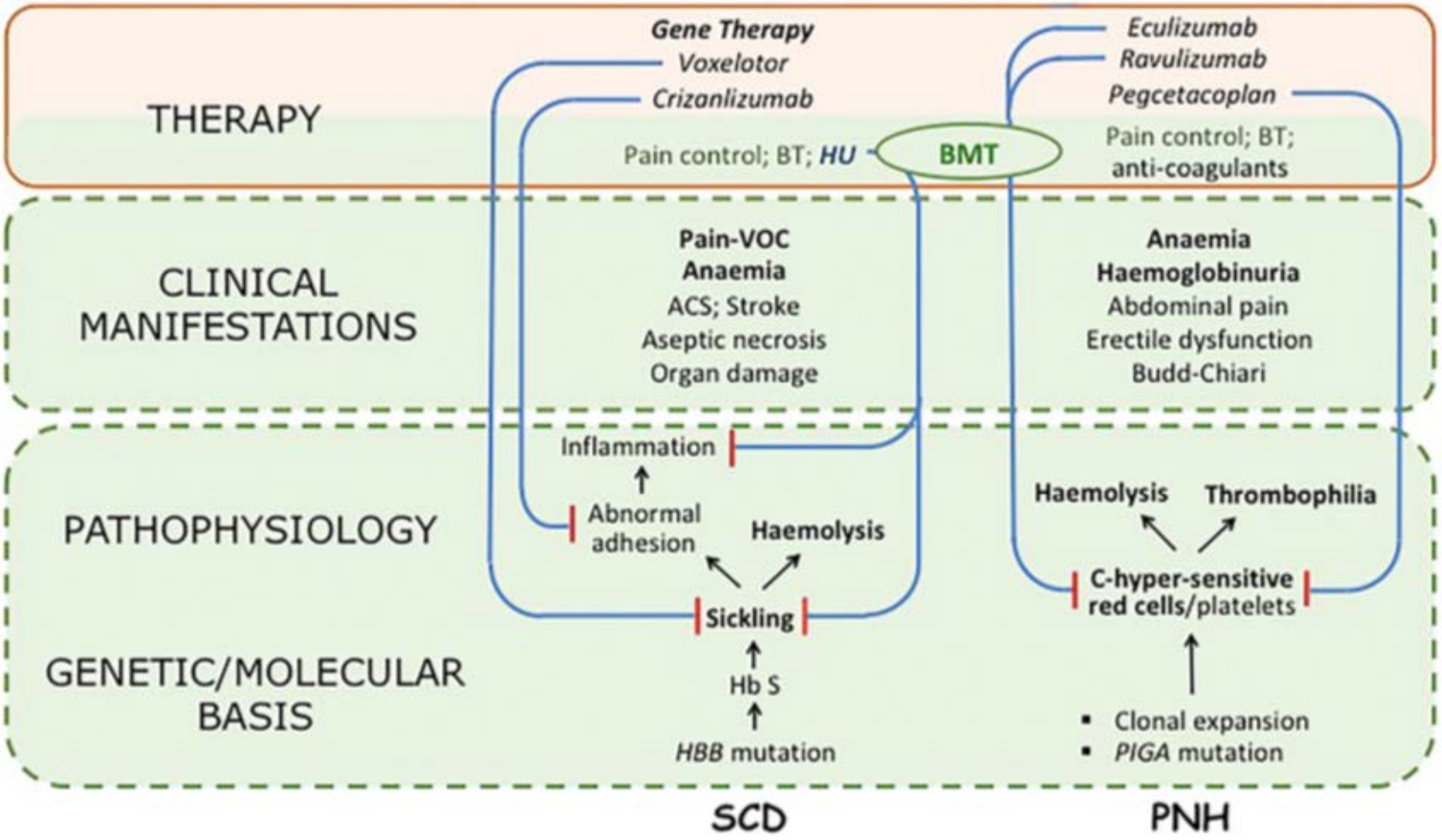
Feature	Properties	In a population of organisms		In a population of somatic cells	
		Consequences	*Examples*	*Cosequences*	*Examples*
Genetic drift	Mutant neutral	Depends on population size	'Founder effects'	Clone may expand when normal cells few	Mutant clones in aplastic anaemia
Darwinian selection	Mutant has higher fitness	Mutant progeny may take over	Gene fixation	Clone will expand	Oncogenic mutations
	Conditional advantage	Depends on environment	*HBB*^{S} gene if malaria endemic	Depends on micro-environment	*PIGA* mutant clone in PNH
	Convergent evolution	Increased frequency of independently arisen mutant genes	Genes of melano-genesis	Clones with mutations in different genes may expand	Many genes can cause leukemia

PNH RED CELLS IN AA PATIENTS IN TANZANIA



(From Ally, Magesa & Luzzatto, *Am J Hematol* **94**:E86,2019)

SYNOPTIC VIEW OF TWO 'RARE' DISEASES



(Luzzatto & Makani, *Front Pharmacol* **12**:770640,2022)

Specific proposals aiming to reduce the gap between potential and reality

(Modified from Luzzatto & Makani, *Front Pharmacol* **12**:770640,2022)

- Adding **SCD**
  to the triad of conditions (HIV, tuberculosis, malaria)
  for which cost of treatment is born by the Global Fund.

- BMT solidarity programme:
  for every BMT (HSCT) procedure in Europe/US,
  0.1% of the expense could be deposited into a fund
  to support BMT in accredited centers in Africa.

- Rare Disease treatment matching programme:
  for every patient treated with a super-expensive drug
  (e.g. eculizumab) reimbursed by NHS/insurance, the
  manufacturer offers the drug to one patient with the
  same disease in Africa.

Two major factors in the evolution of organisms or of somatic cells

Fortuna

τύχη

CHANCE

DARWINIAN SELECTION

In the Yoruba culture of SW Nigeria the *Ifa* priest analyzes your life based on 2^8 combinations of cowrie shells that fall randomly on *recto* or *verso*.

THANK YOU!

Ibadan

Adeyinka AFOLAYAN
Olaniyi BABALOLA
John BEETLESTONE
Adeynka FALUSI
Adetokunbo LUCAS

Dar-es-Salaam

Mwashungi ALLY
Julie MAKANI
Ahlam NASSRE
Florence URIO

Napoli

FiorellaA ALFINITO
Michele D'URSO
Maria Grazia PERSICO
RobertoROBLEDO
Bruno ROTOLI
Ugo TESTA

Firenze

Piero DOLARA
Luciana GARGIULO
Giacomo GIANFALDONI
Caterina NANNELLI
Rosario NOTARO
Michela SICA

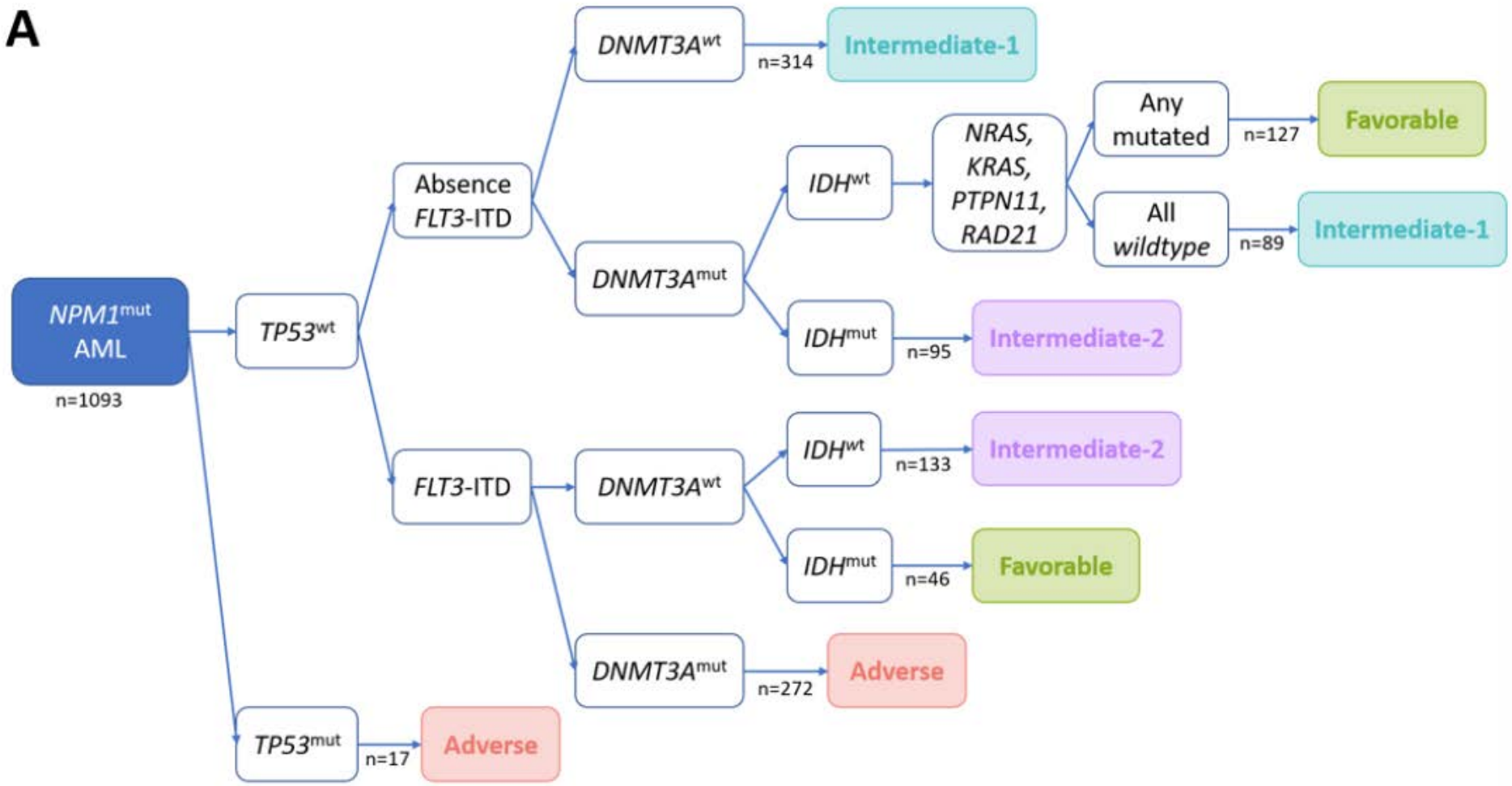
London

Tim COX
Inderjeet DOKAL
Letizia FORONI
Frank GROSVELD
Peter HILLMEN
Letizia LONGO
Philip MASON
Tom VULLIAMY
Winifred WATKINS

New York

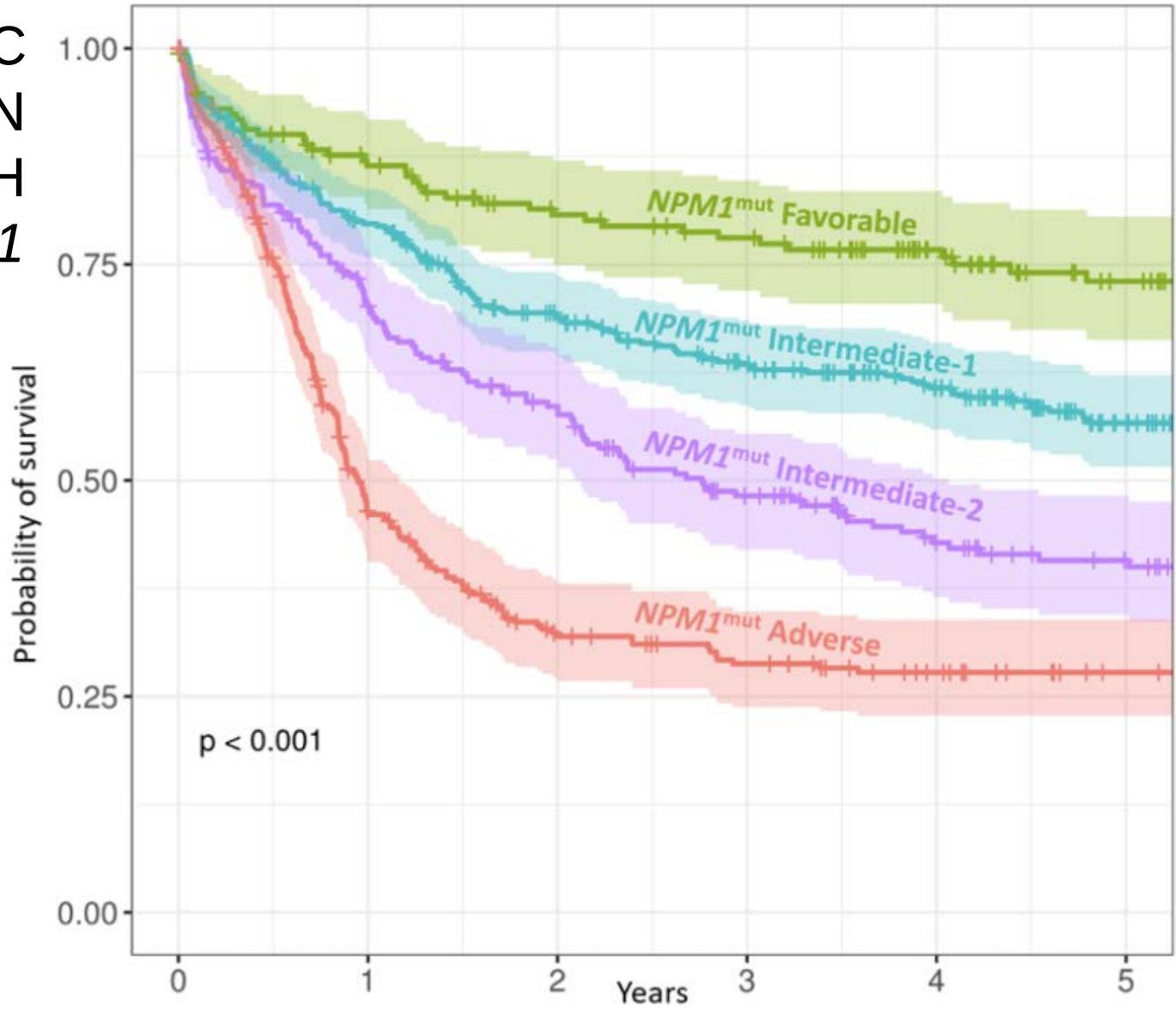
David ARATEN
Monica BESSLER
Anasyasios KARADIMITRIS
Khedouja NAFA
Rosario NOTARO
PierPaolo PANDOLFI
Vittorio ROSTI
Michel SADELAIN
Gabi TREMML

SOMATIC MUTATIONS IN AML WITH MUTANT *NPM1*



(From Alberto Hernández Sánchez et al., Abstract 304, ASH 2022)

PROGNOSTIC CLASSIFICATION OF AML WITH MUTANT *NPM1*



(From Alberto Hernández Sánchez et al., Abstract 304, ASH 2022)


FLORENTINA STUDIORUM UNIVERSITAS


FLORENTINA STUDIORUM UNIVERSITAS


FLORENTINA STUDIORUM UNIVERSITAS