National University Cancer Institute, Singapore

NUHS National University Health System

NUH kids National University Hospital

Acute Lymphoblastic Leukaemia in Singapore: Why we need local trials

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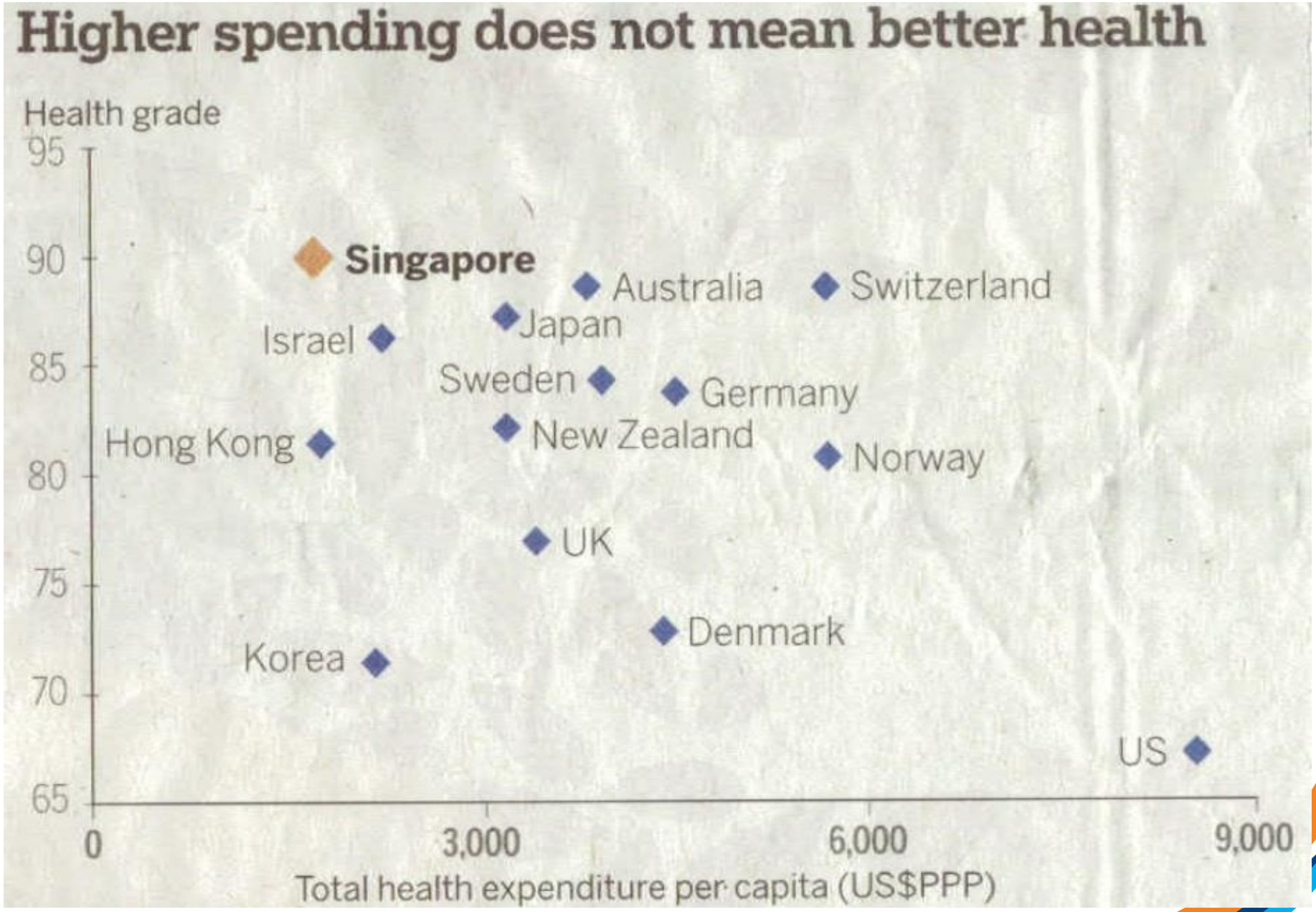
Khoo Teck Puat-National University Children's Medical Institute, NUH
Yong Loo Lin School of Medicine, NUS
National University Health System

Singapore Healthcare: Better value, better health

- 4 M
  - Medisave
  - Medishield, Medishield+
  - Medifund
  - MediLife
- Asset rich, cash poor – locked up in housing
- Tiered care – subsidized and private
- Medical Insurance


Higher spending does not mean better health
Health grade
95
90
85
80
75
70
65
Singapore
Australia
Switzerland
Japan
Israel
Sweden
Germany
New Zealand
Hong Kong
Norway
UK
Denmark
Korea
US
0
3,000
6,000
9,000
Total health expenditure per capita (US$PPP)

Health and Biomedical Science Research

5 Ps

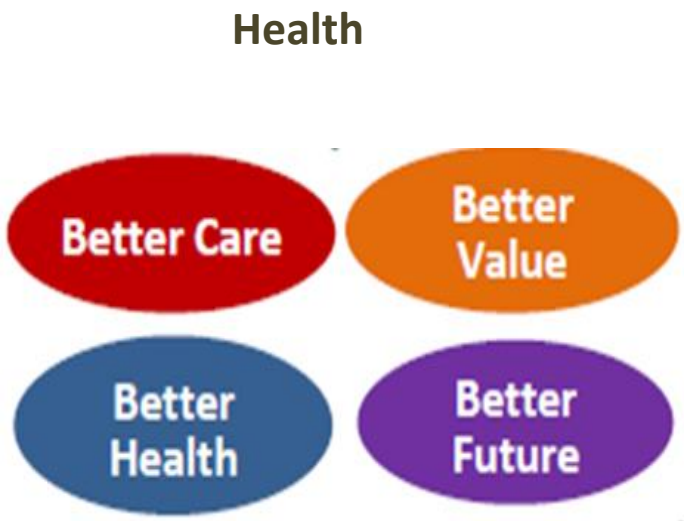
Publication.

Practice

Product

Policy

Population



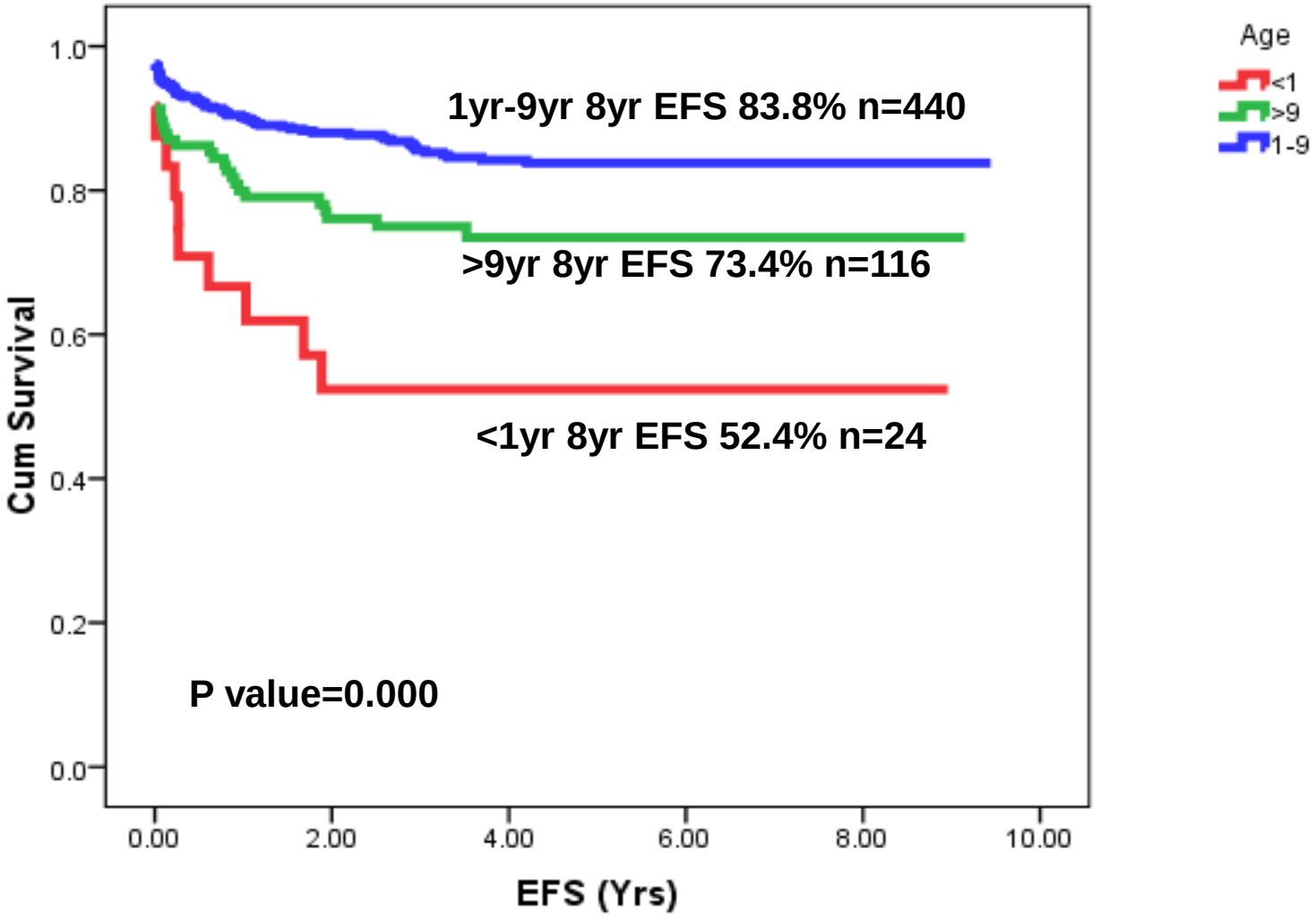
Accurate risk assignment in ALL: Stratified Medicine

NCI criteria – Age 1-10 years old & WBC < 50k

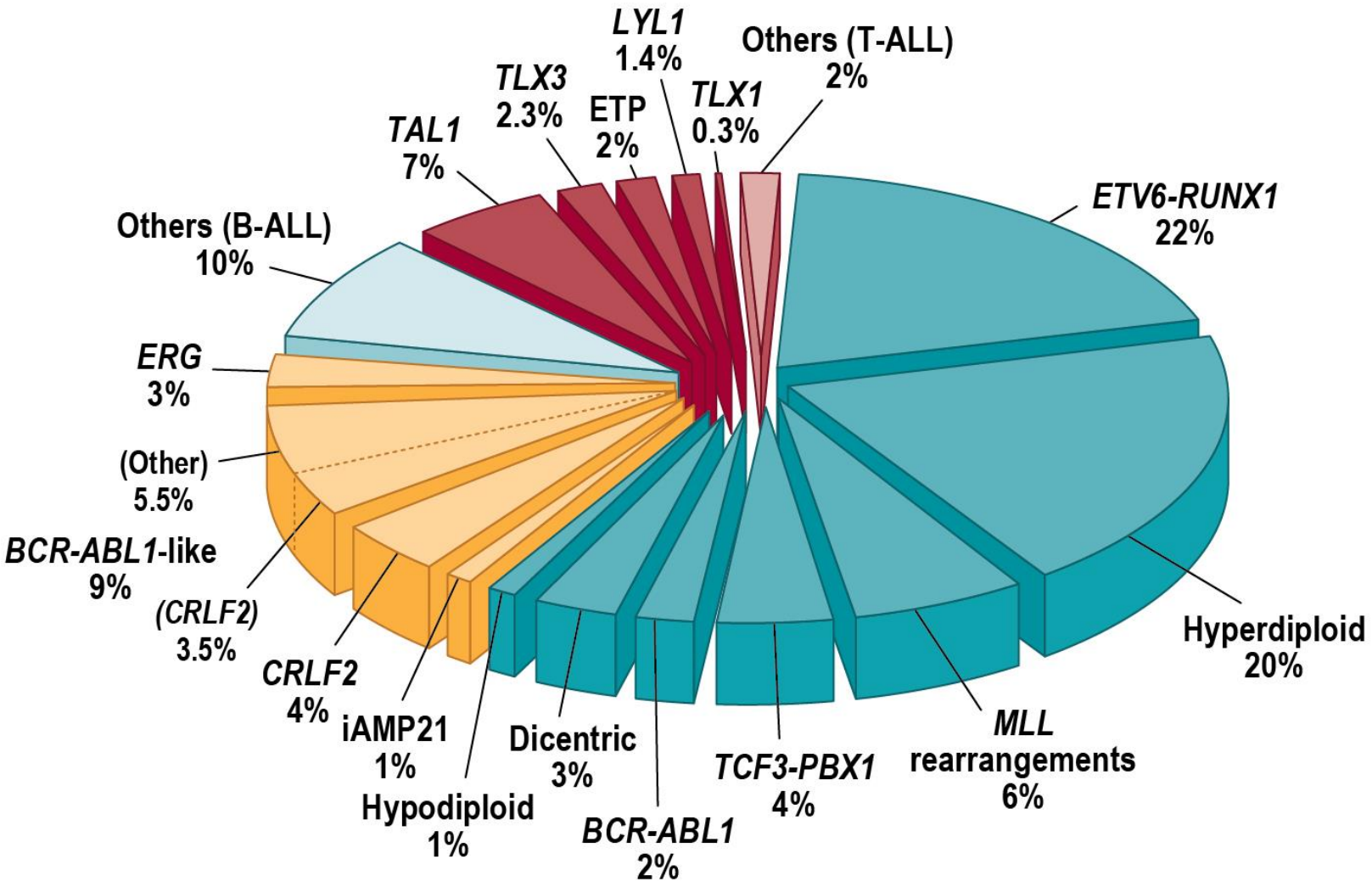
Cytogenetics
Favourable – Hyperdiploid>50; TEL-AML1, E2A-PBX1
Unfavorable – Hypodiploid < 44, BCR-ABL1, MLL-R

Minimal residual disease (MRD) quantitation

EFS by Age Groups

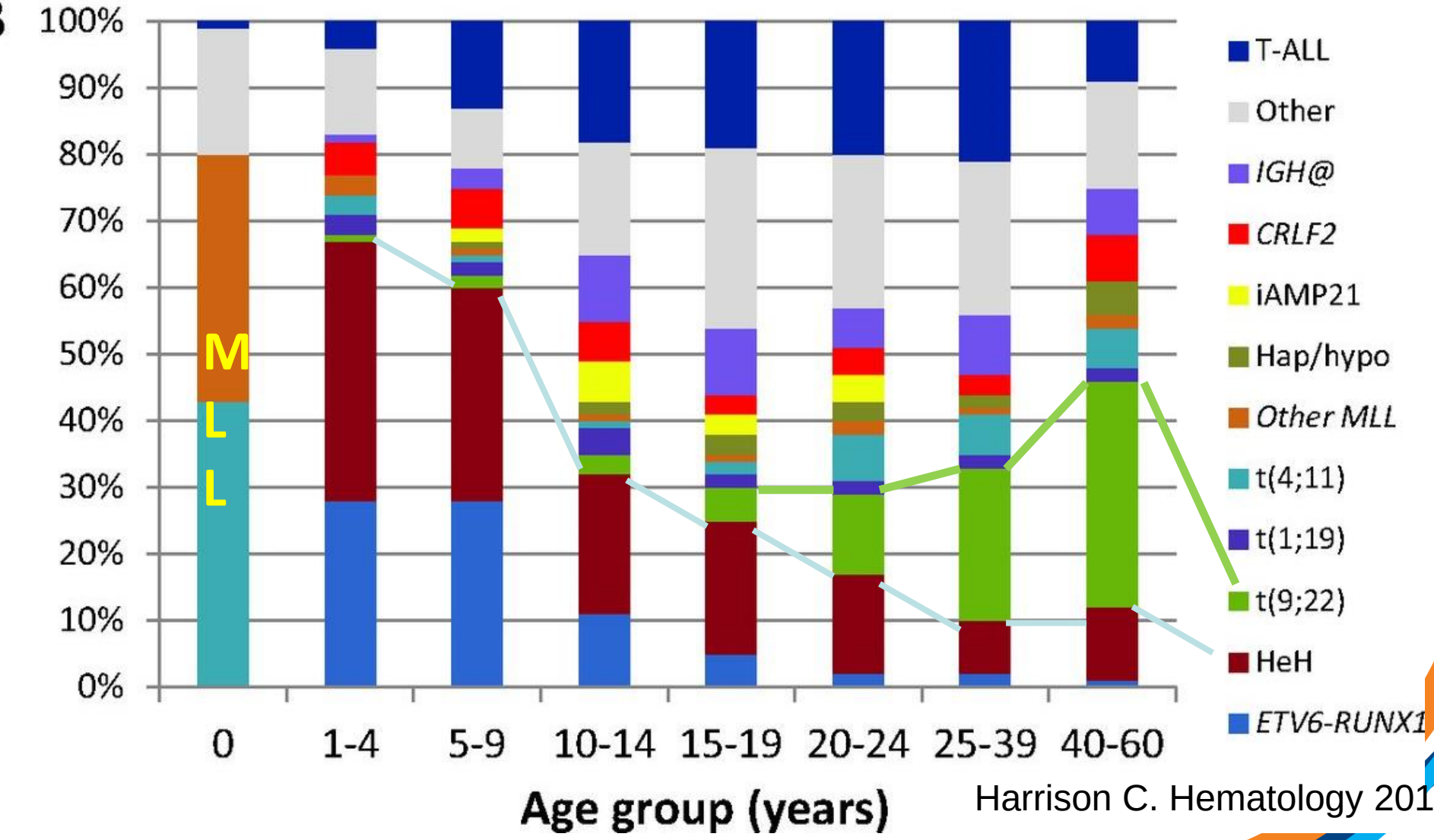


Genetic Classification of ALL



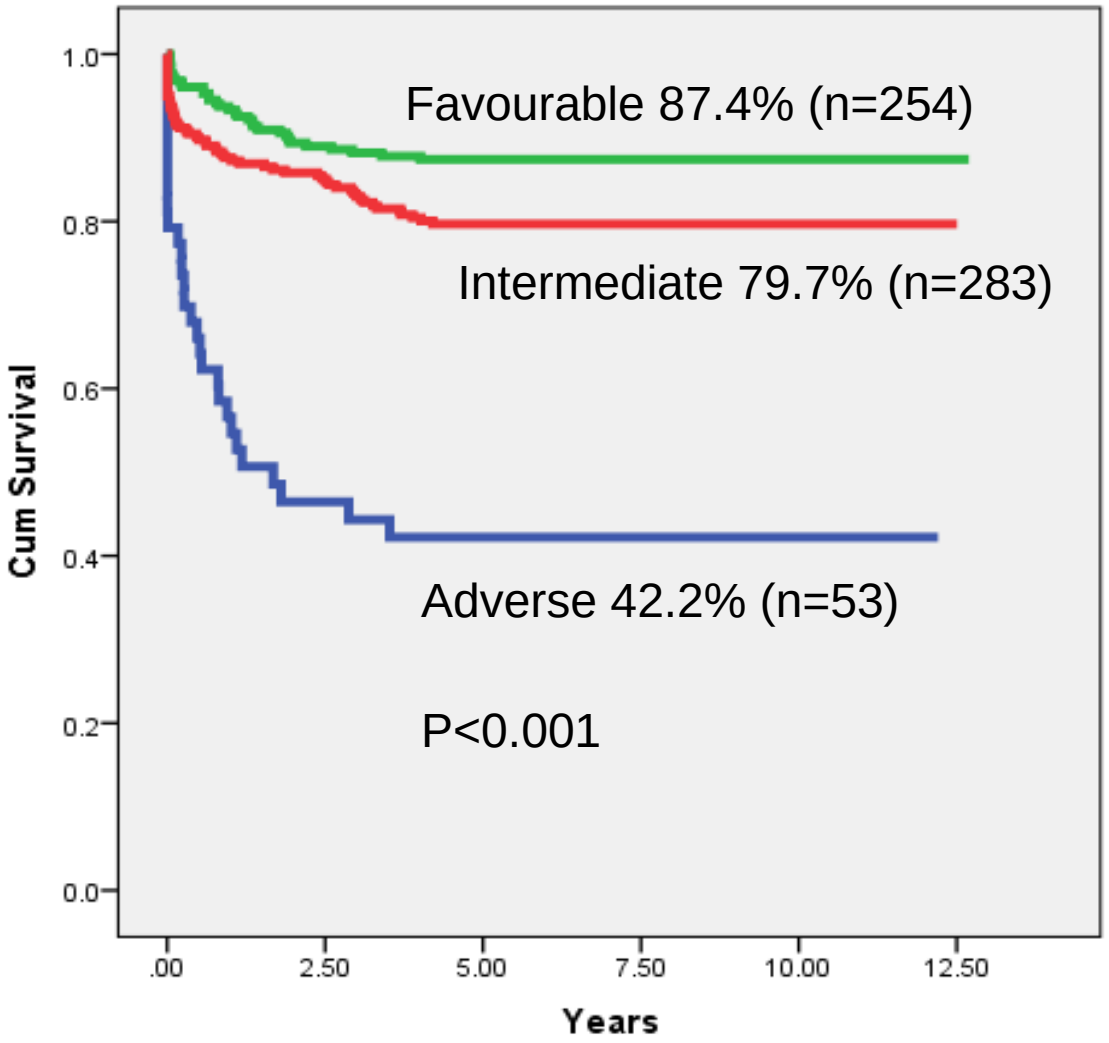
Pui et al. J Clin Oncol

Cytogenetics and age in ALL



Harrison C. Hematology 2014

Ma-Spore ALL 2003 5y- EFS
by cytogenetics/molecular

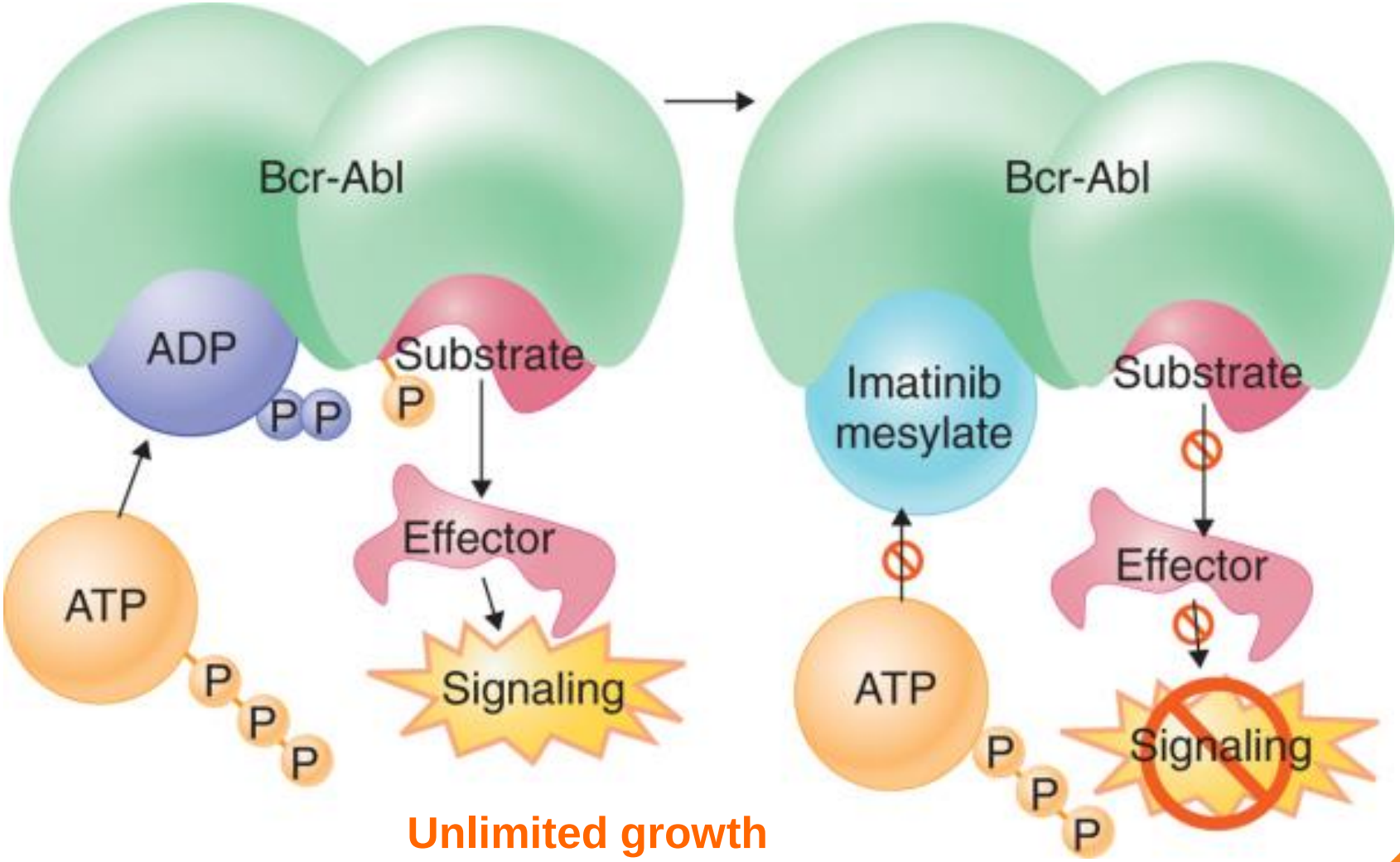


Favourable
- high hyperdiploid,
- ETV6-RUNX1,
- TCF3-PBX1)

Intermediate
- normal karyotype
- Others
-

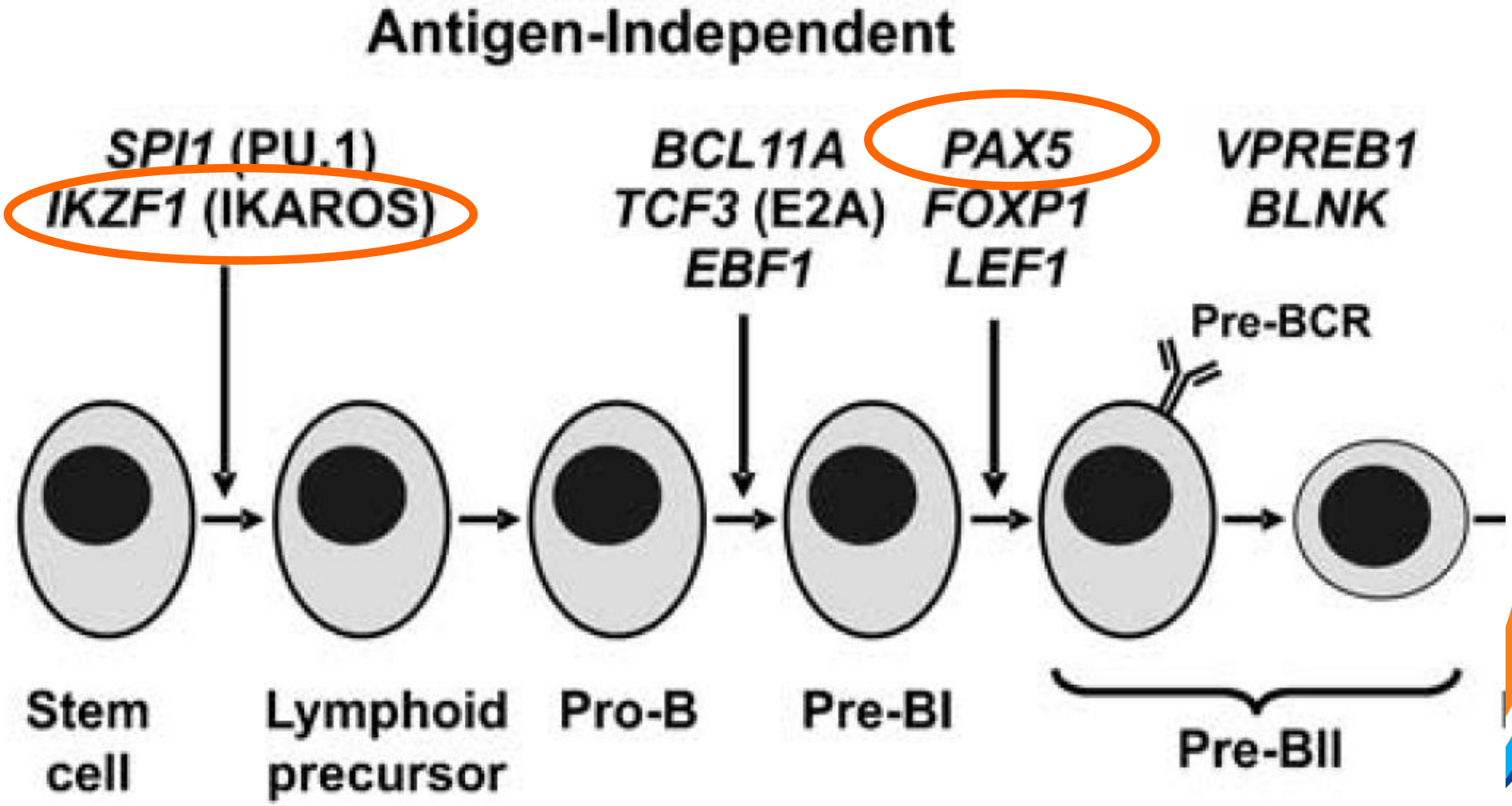
Unfavorable
- Hypodiploid <45
- MLL-rearrangement,
- BCR-ABL1 and
- SIL-TAL1
- subgroups

Imatinib – targeted therapy for BCR-ABL1



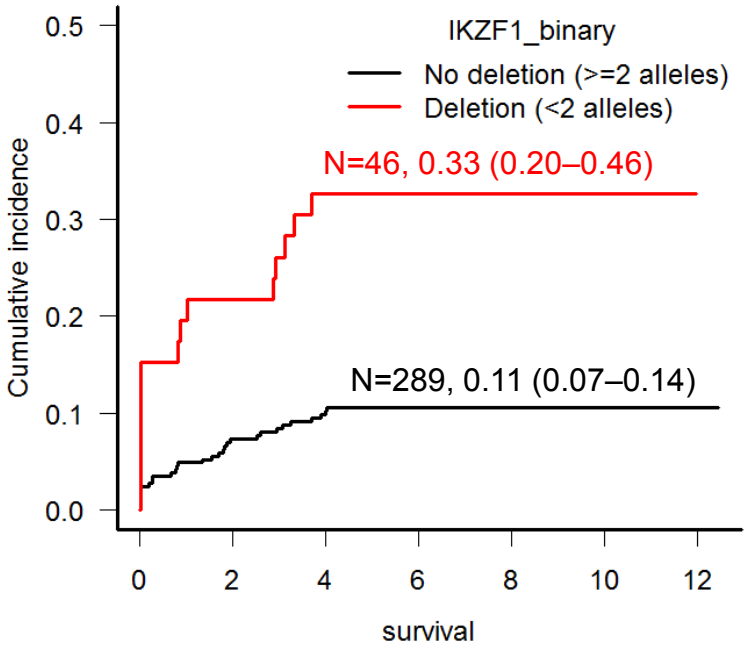
Unlimited growth

Alteration in B-cell development genes -> ALL

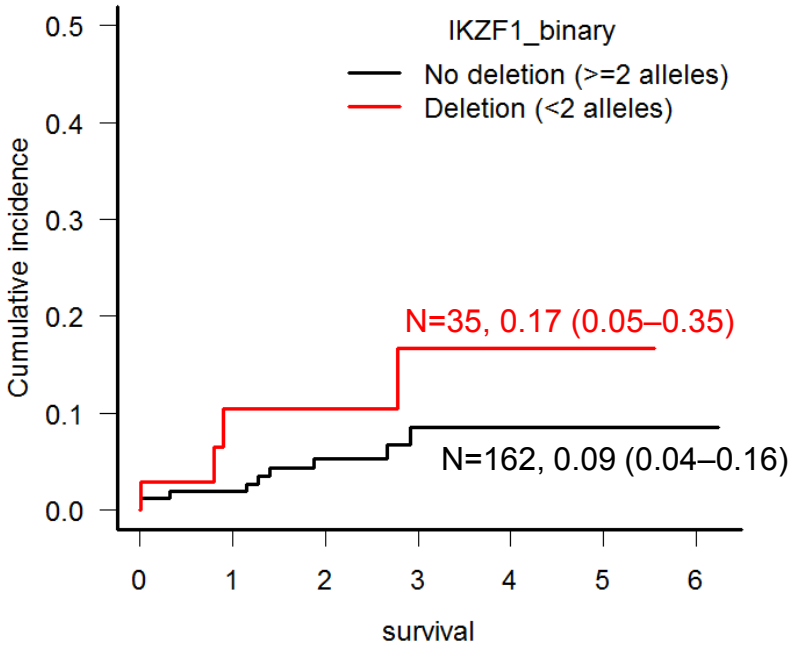


Intervening by intensification Ikaros deletion (Ma-Spore ALL 2003 vs 2010)

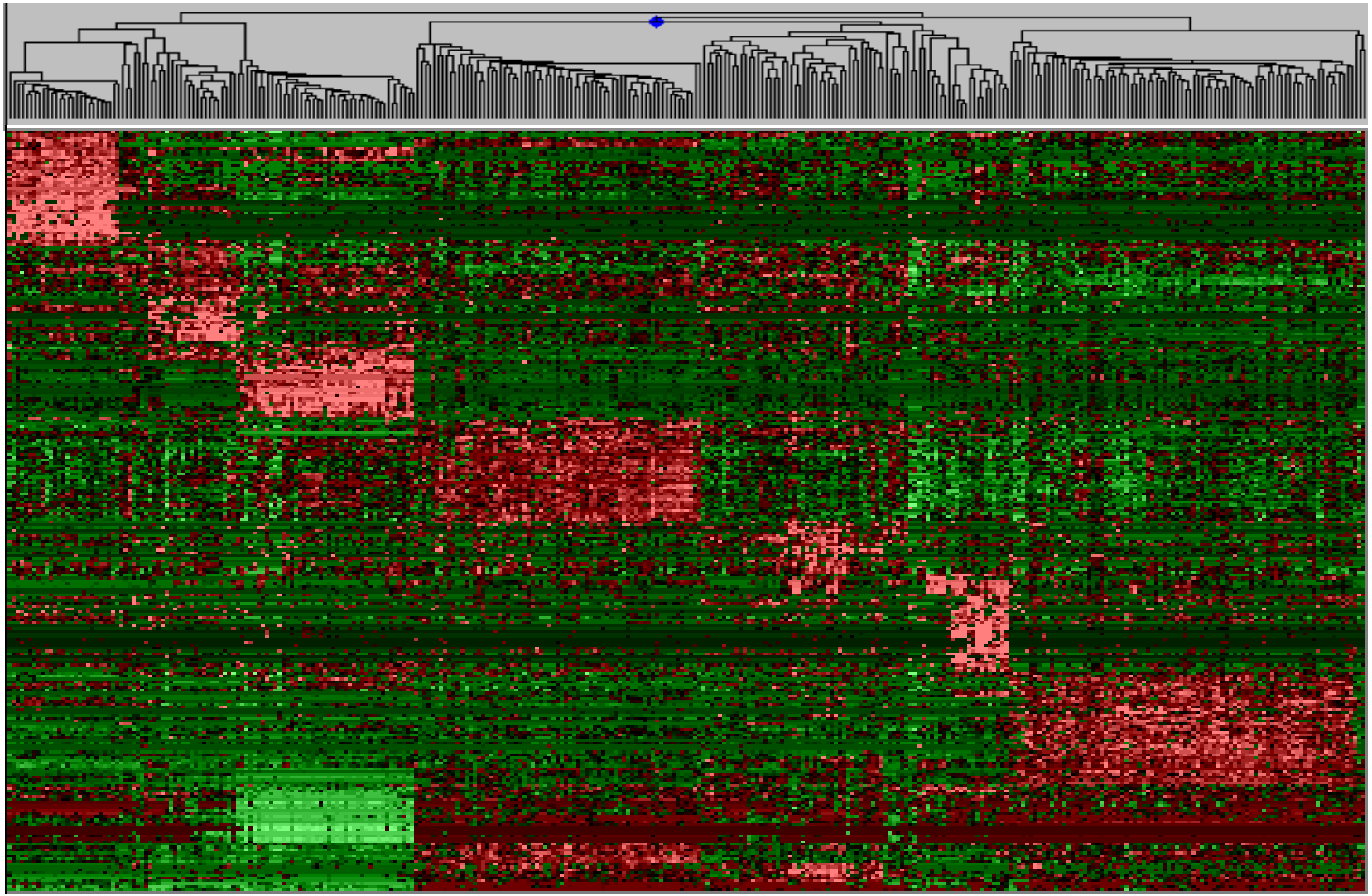
IKZF1 (P<0.001)



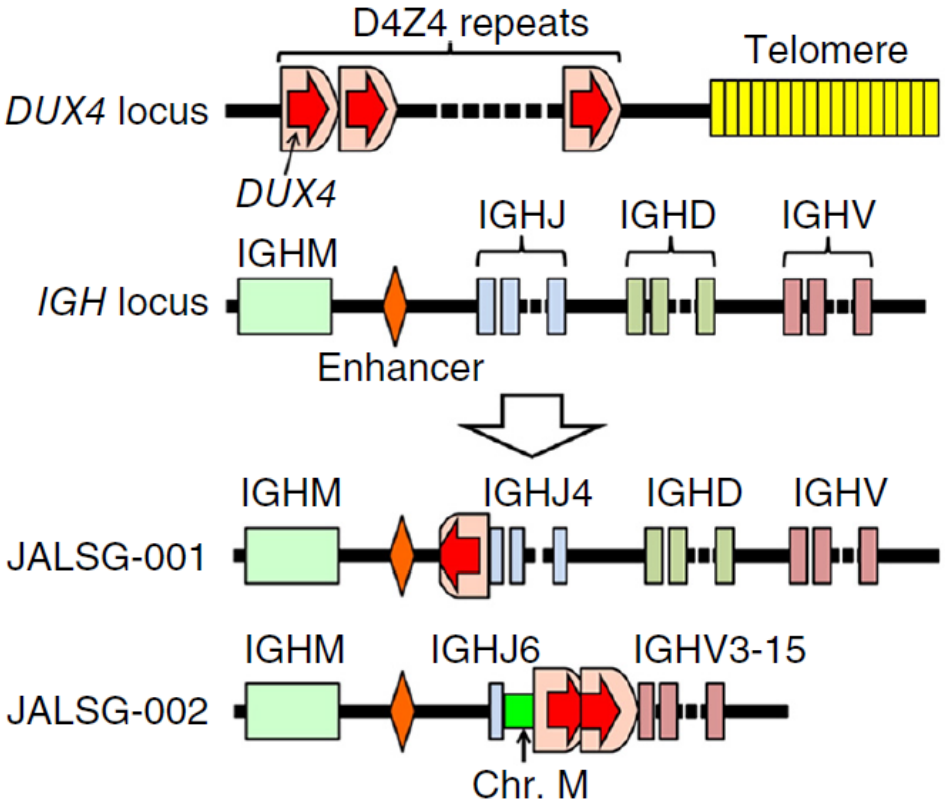
IKZF1 (P=0.178)

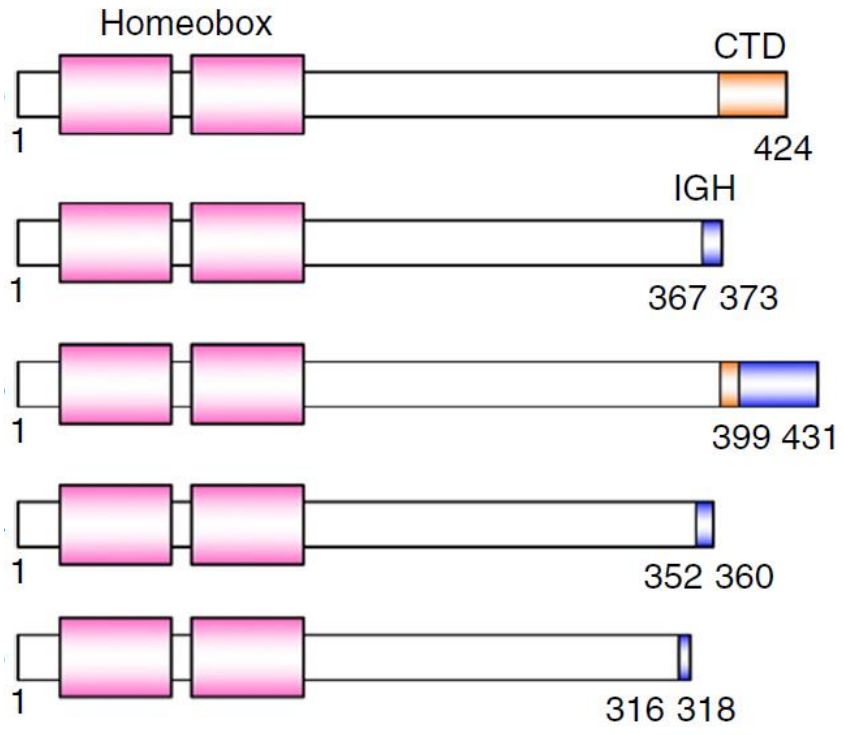


Diagnostic ALL BM Samples (n = 327)

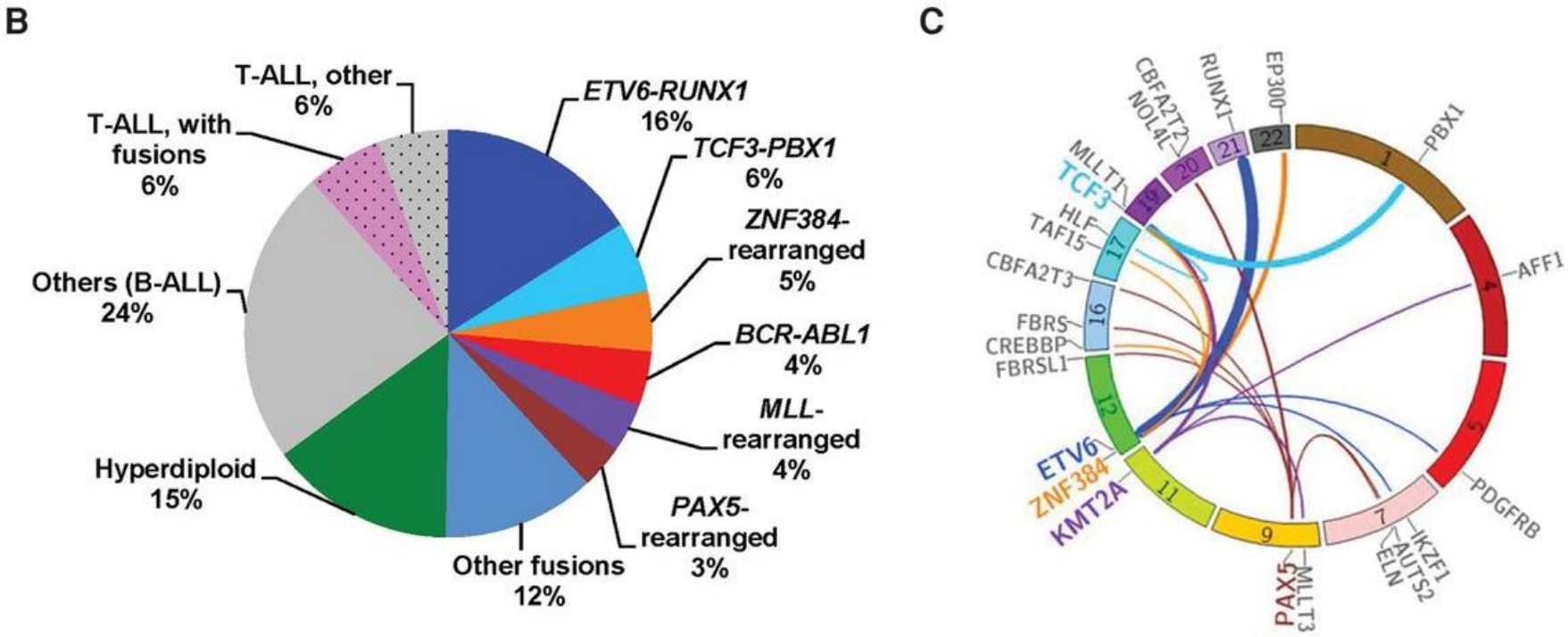


D4Z4 repeats inserted after enhancer of IgH
In B-ALL, IgH enhancer active,
increase DUX4 expression



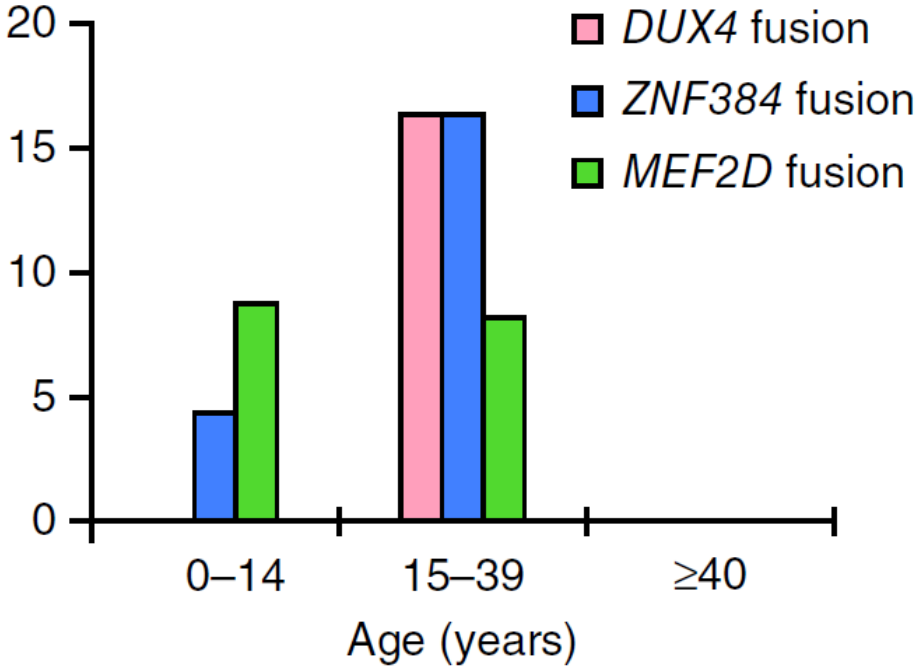


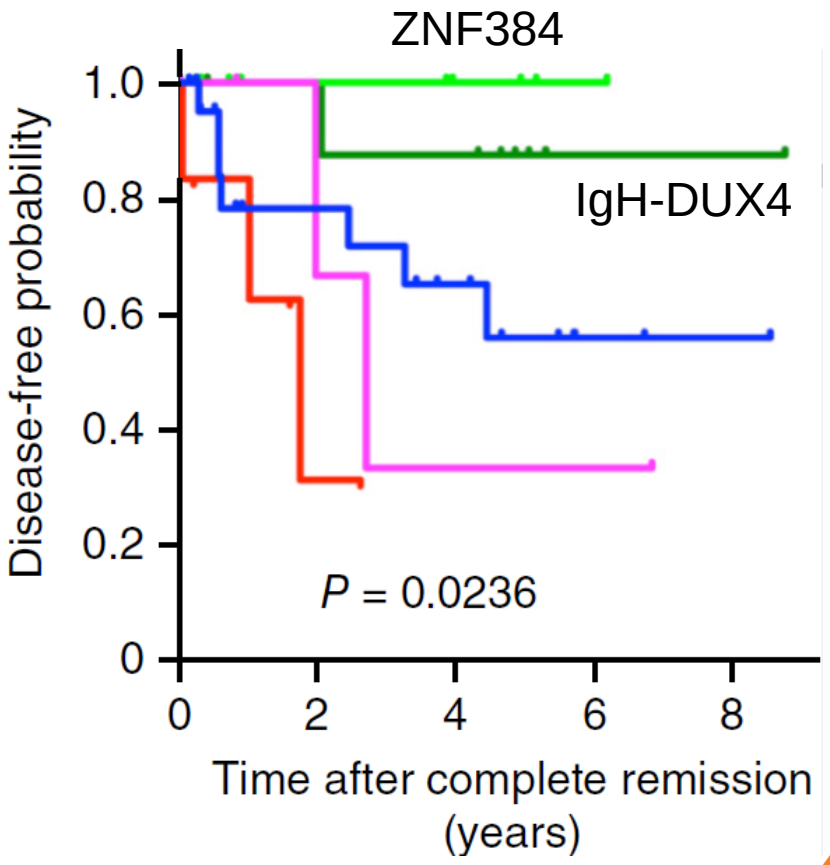
ZNF384-rearranged leukaemia – good prognosis



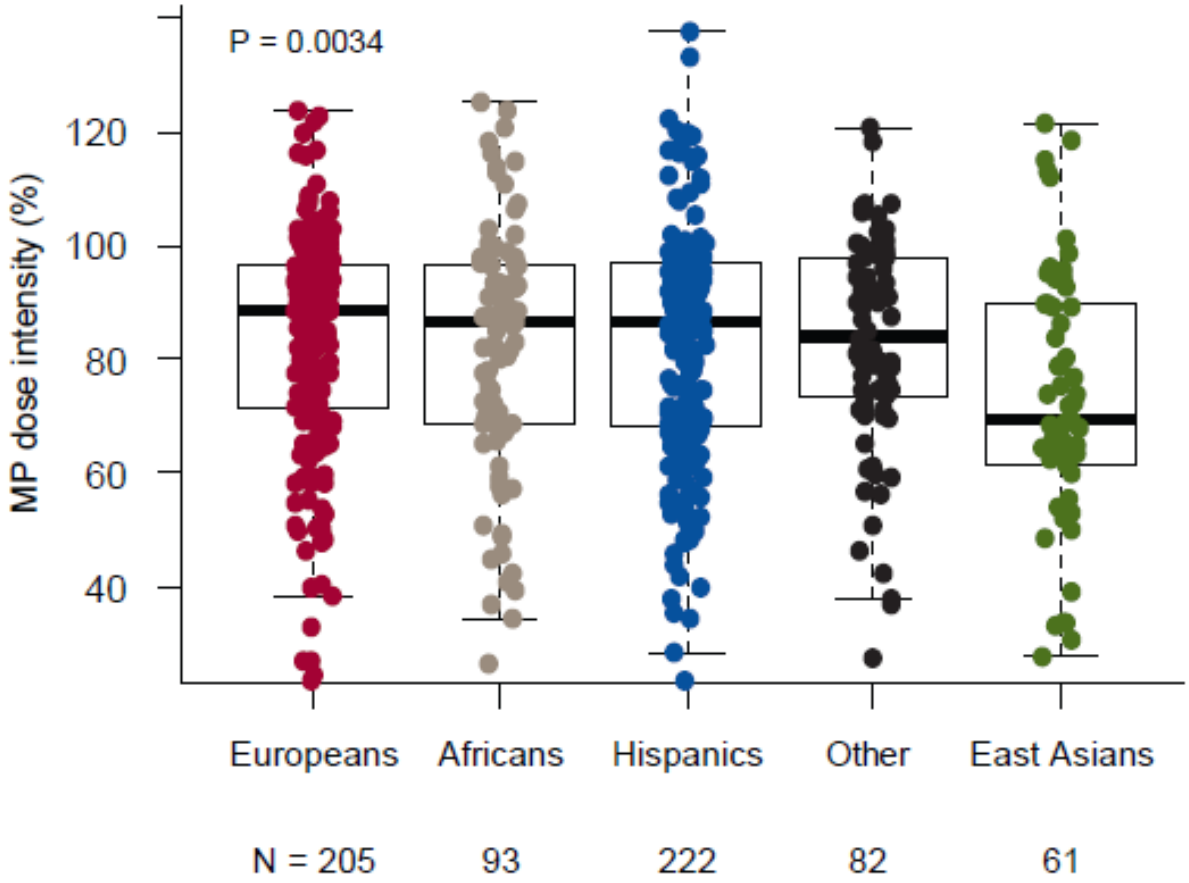
Qian M, … Quah TC, Yeoh AEJ, Yang J. Gen Res 2017;27(2):185

IgH-DUX4 – a/w ERG deletion
IgH-DUX4, ZNF384 excellent outcome



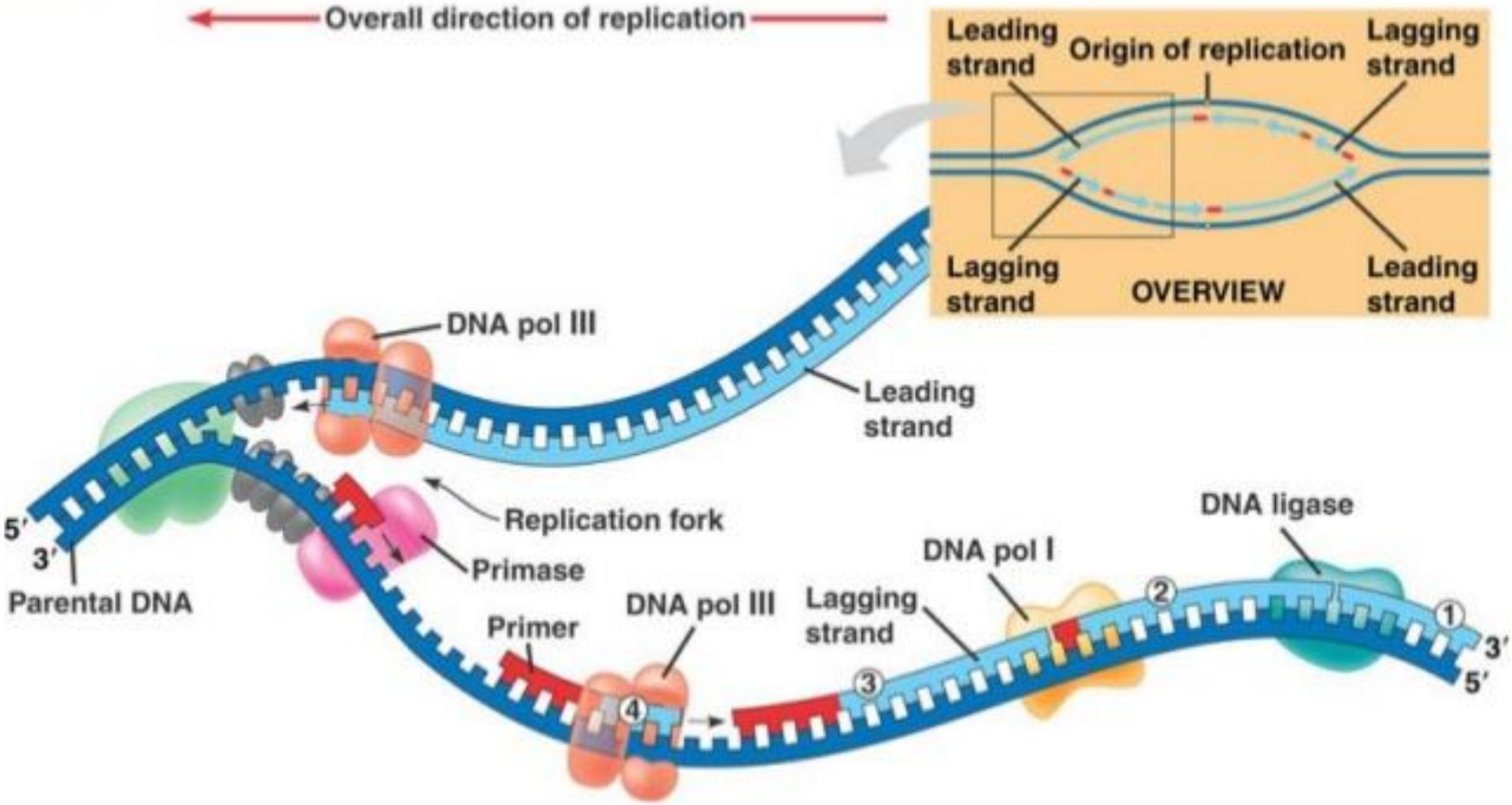


East Asian Ancestry has Lower 6MP Tolerance



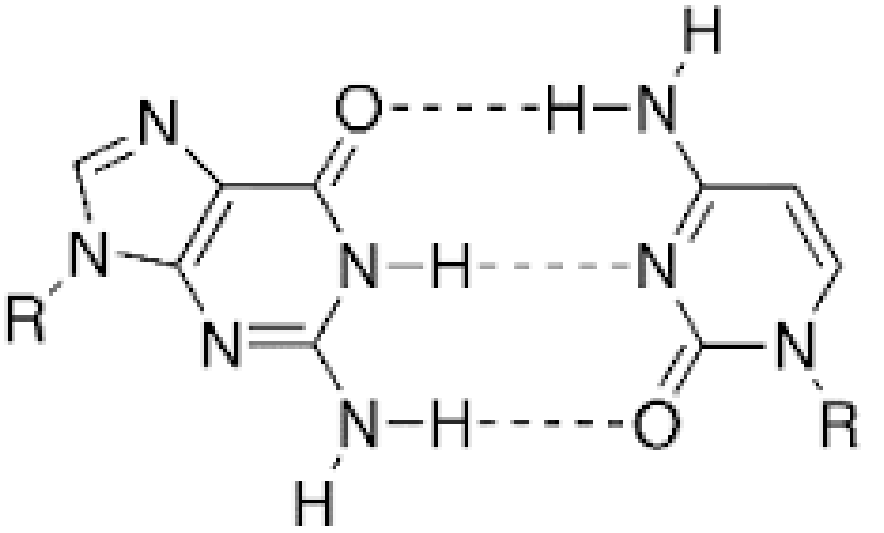
Yang et al., J Clin Oncol 2015

Replication Fork



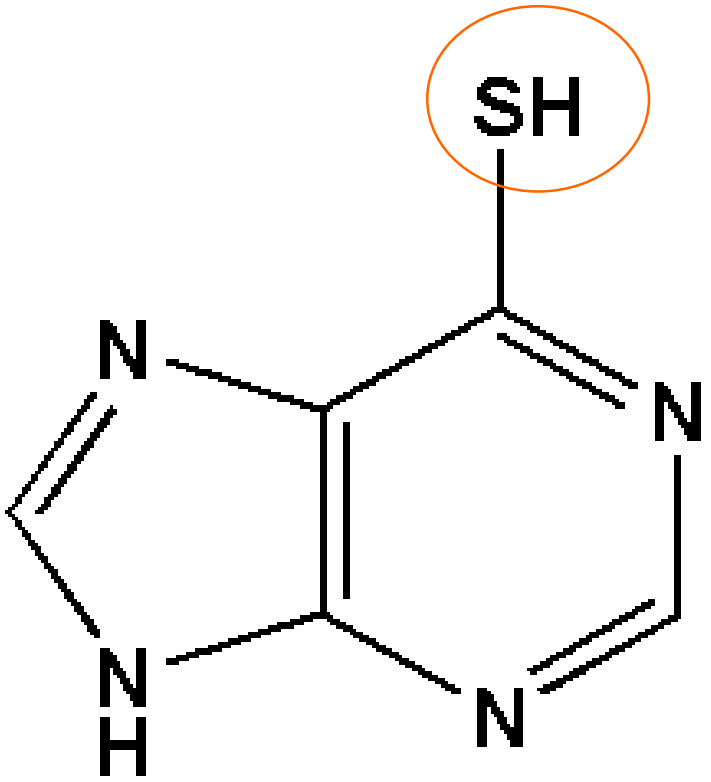
http://kvhs.nbed.nb.ca/gallant/biology/replication_overview.jpg Fig. 16.16

Guanine pairs with cytosine – 3 hydrogen bonds



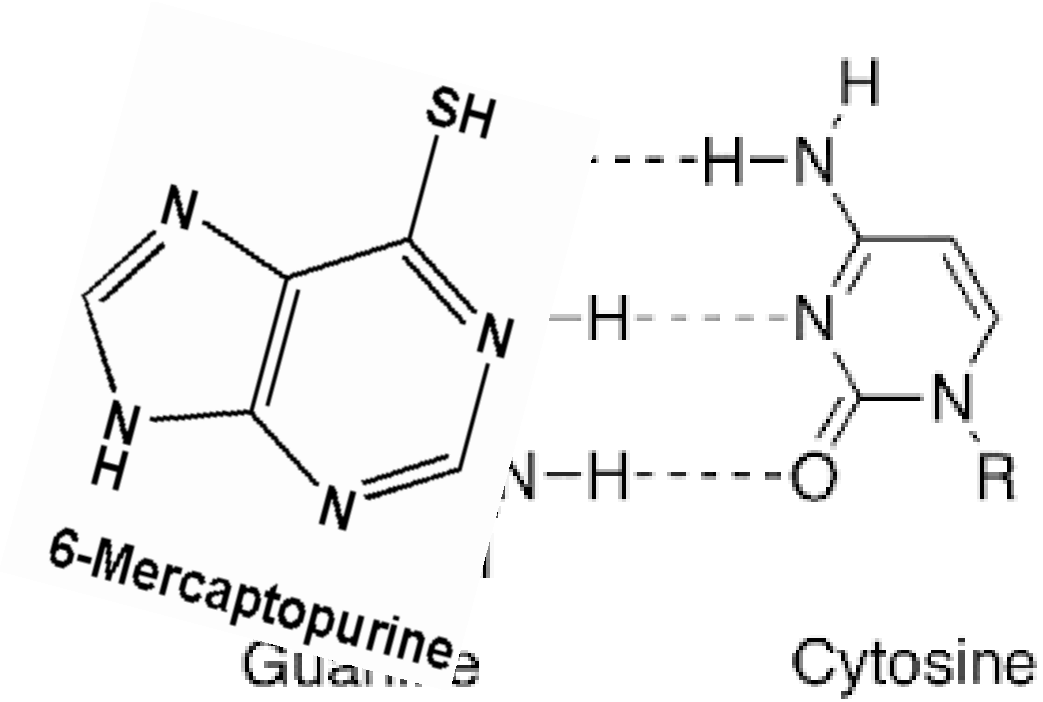
Guanine Cytosine

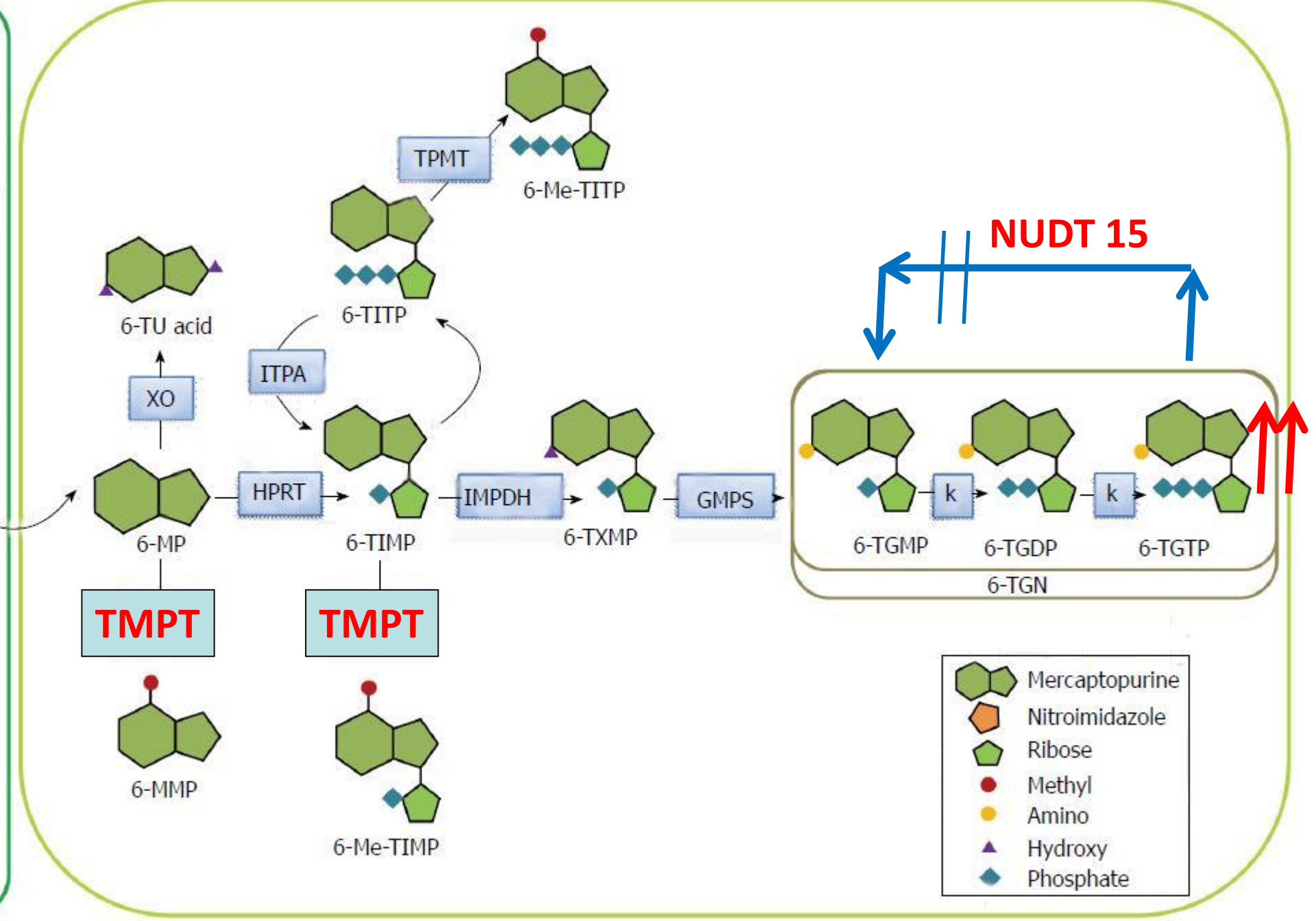
Gertrude Elion – Nobel Prize 1988



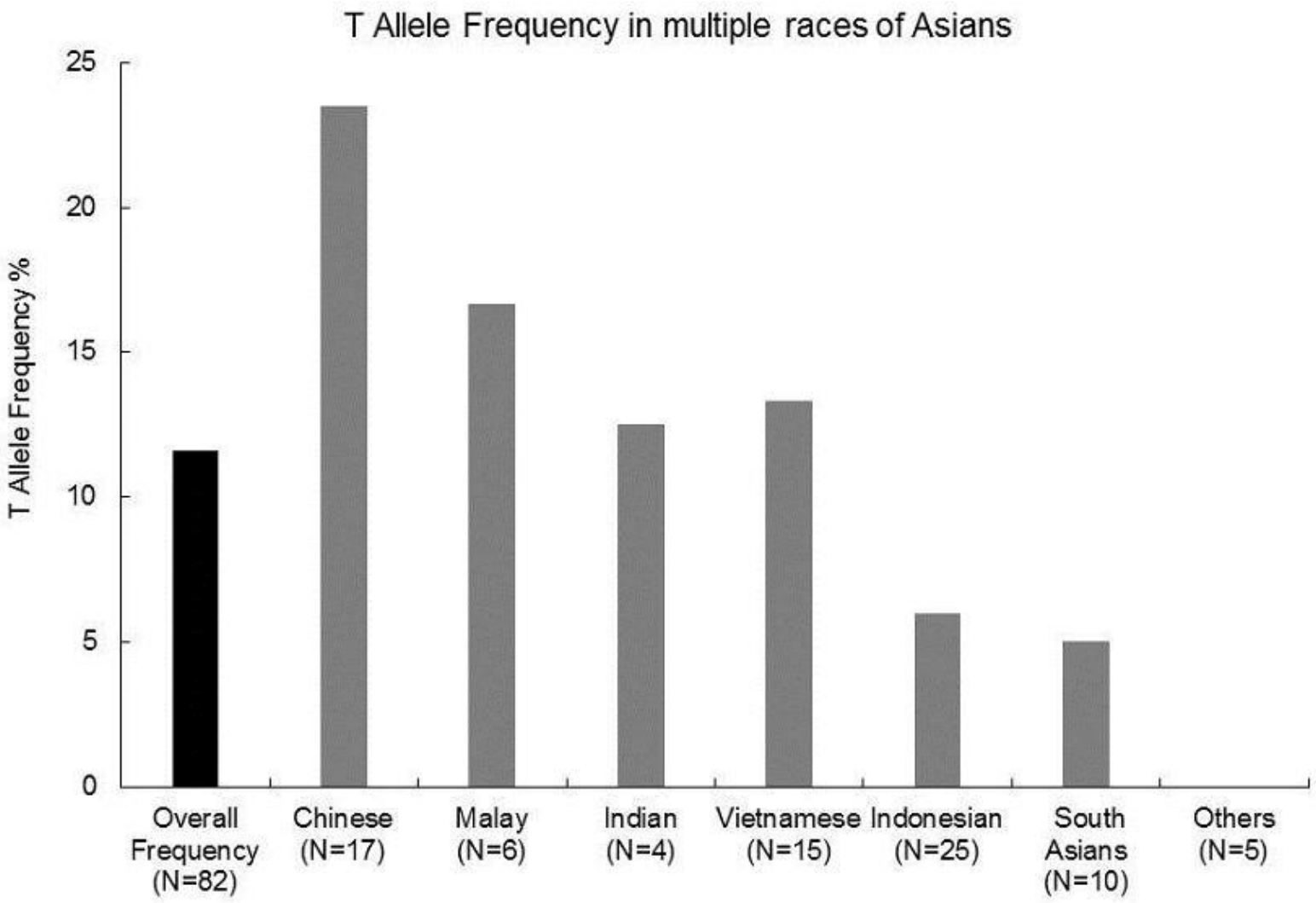
6-Mercaptopurine

Mercaptopurine – designed analog to block pairing
Stuck Zipper – fail DNA replication

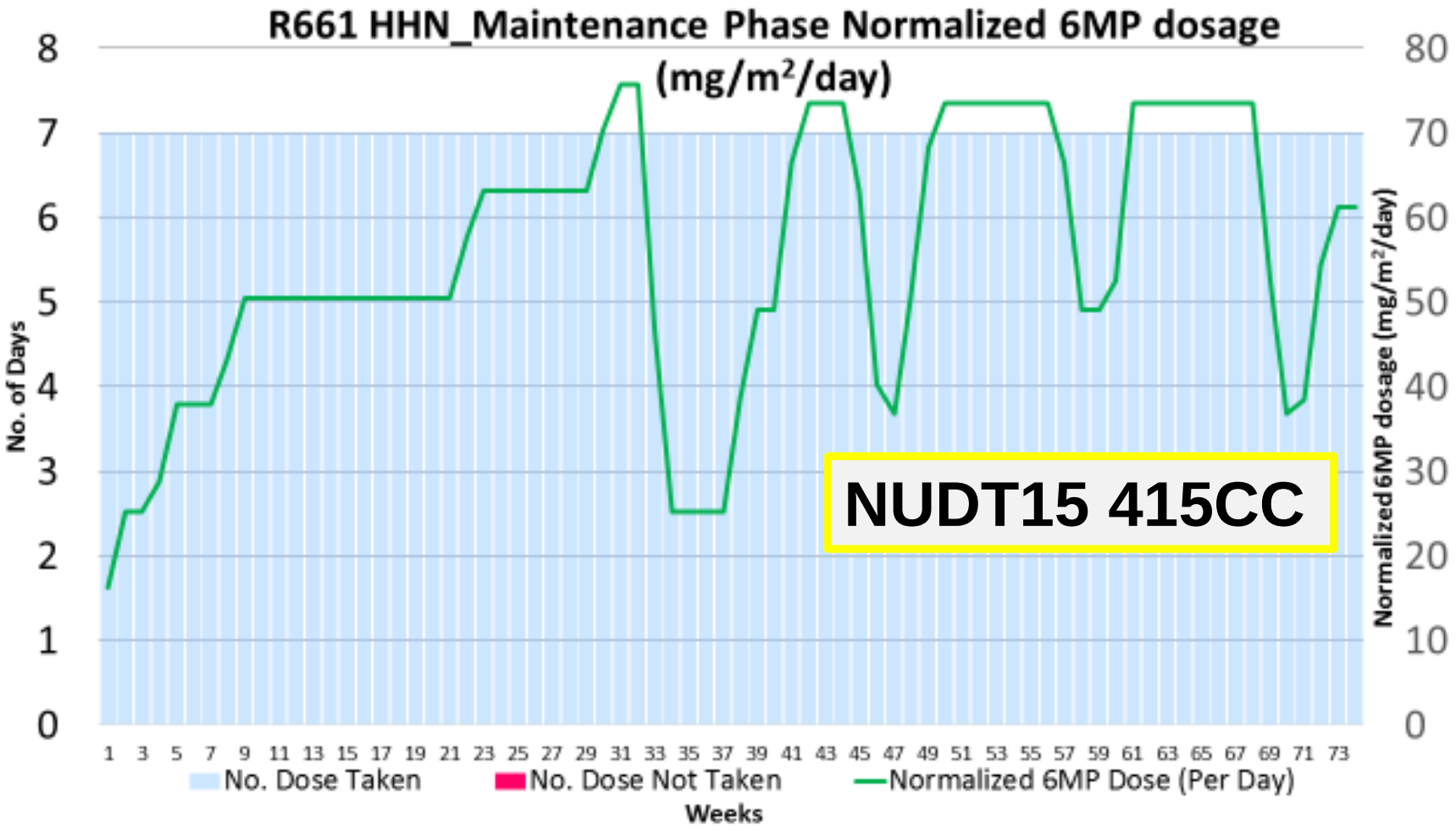


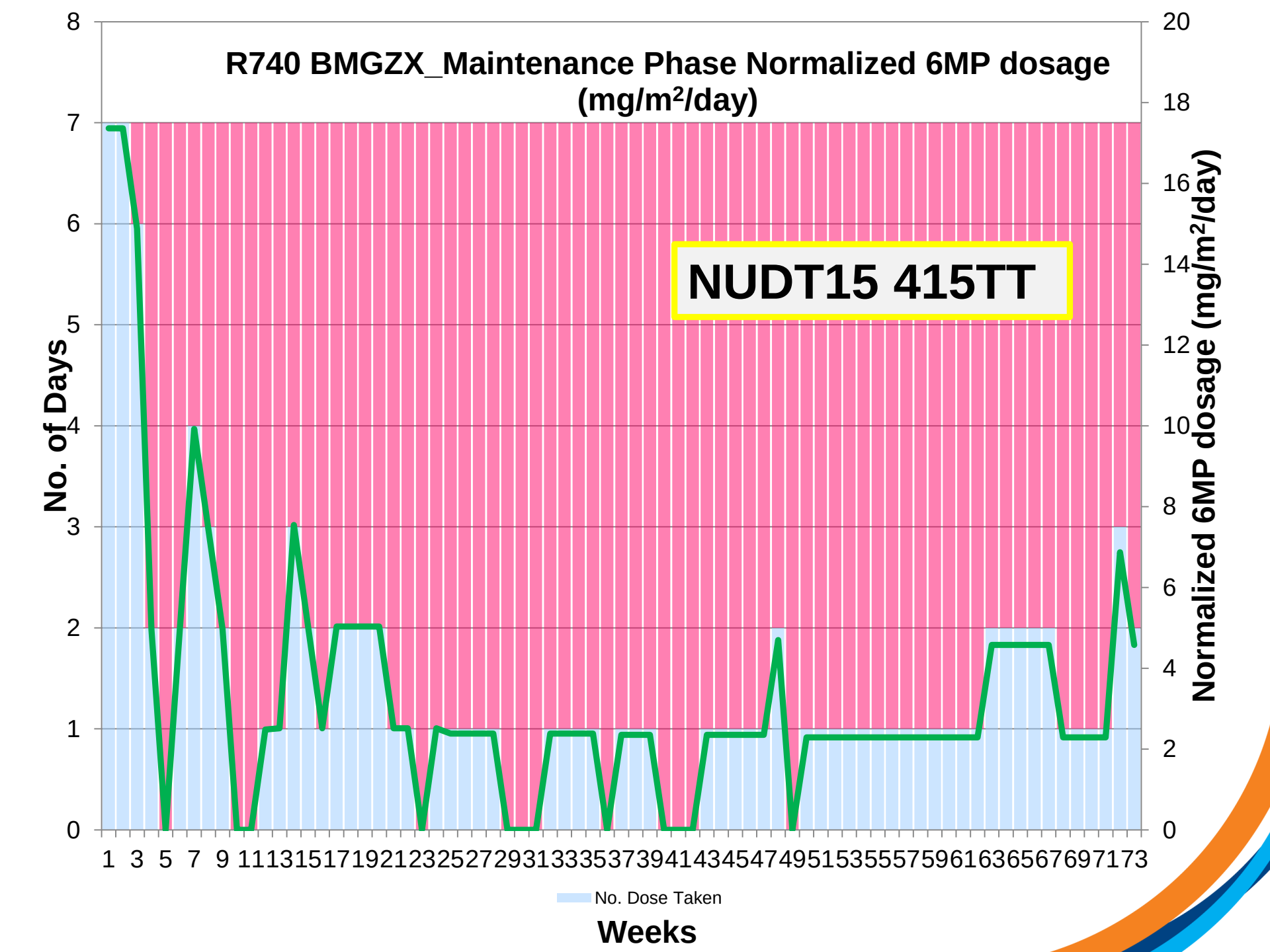

TPMT
6-Me-TITP
6-TITP
6-TU acid
XO
ITPA
HPRT
6-MP
6-TIMP
IMPDH
6-TXMP
GMPS
NUDT 15
k
k
6-TGMP
6-TGDP
6-TGTP
6-TGN
TMPT
TMPT
6-MMP
6-Me-TIMP
Mercaptopurine
Nitroimidazole
Ribose
Methyl
Amino
Hydroxy
Phosphate

Frequency of NUDT15 415T in NUH

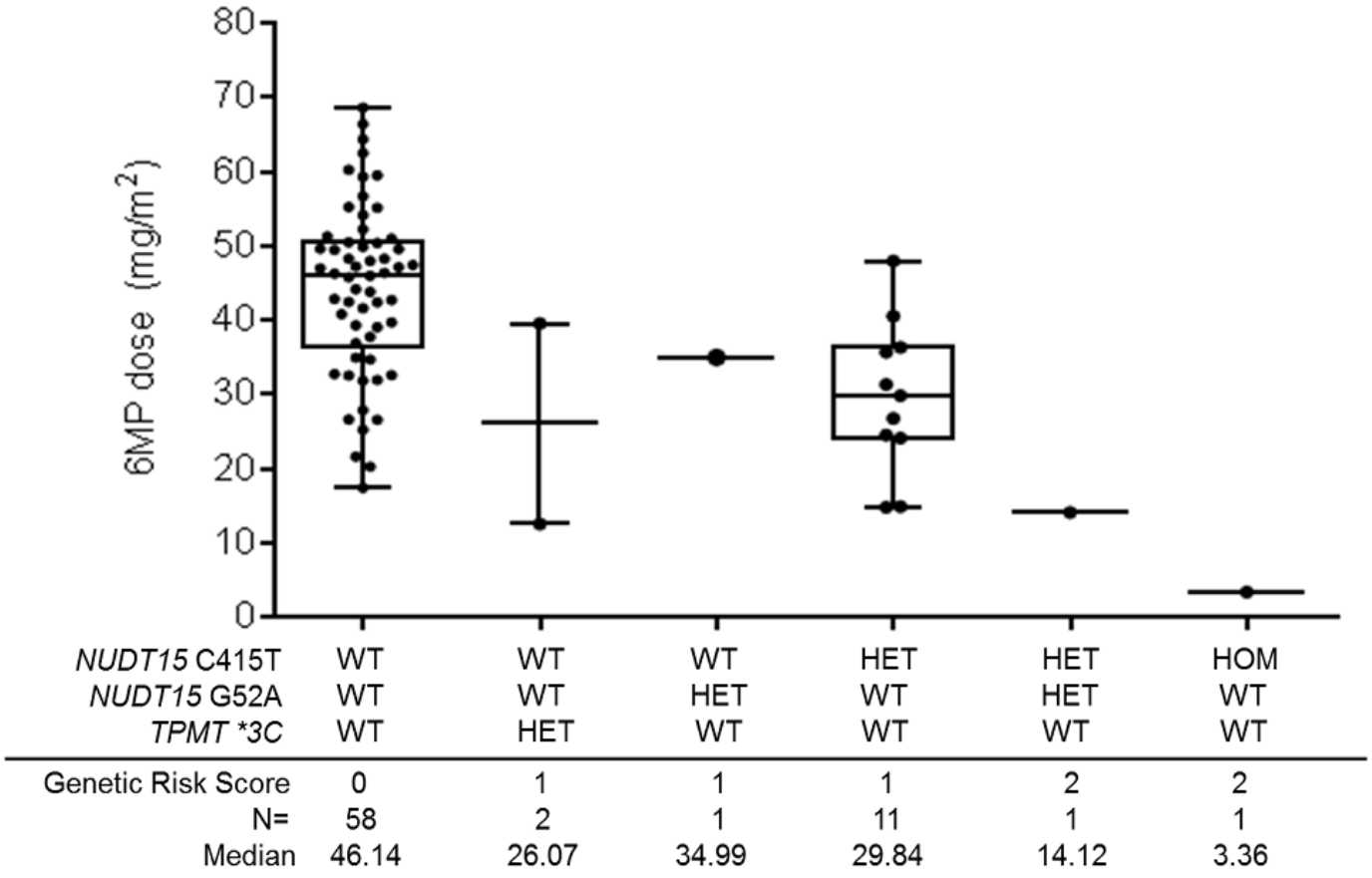


NUDT15 and TPMT wild type: Genetic score 0




R740 BMGZX_Maintenance Phase Normalized 6MP dosage (mg/m²/day)
NUDT15 415TT
No. of Days
Normalized 6MP dosage (mg/m²/day)
No. Dose Taken
Weeks
0
1
2
3
4
5
6
7
8
0
2
4
6
8
10
12
14
16
18
20
1 3 5 7 9 11 13 15 17 19 21 23 25 27 29 31 33 35 37 39 41 43 45 47 49 51 53 55 57 59 61 63 65 67 69 71 73

Combined *NUDT15* and *TPMT* on MP Tolerance; Ma-Spore ALL 2003 experience



NUDT15 C415T	WT	WT	WT	HET	HET	HOM
NUDT15 G52A	WT	WT	HET	WT	HET	WT
TPMT *3C	WT	HET	WT	WT	WT	WT
Genetic Risk Score	0	1	1	1	2	2
N=	58	2	1	11	1	1
Median	46.14	26.07	34.99	29.84	14.12	3.36

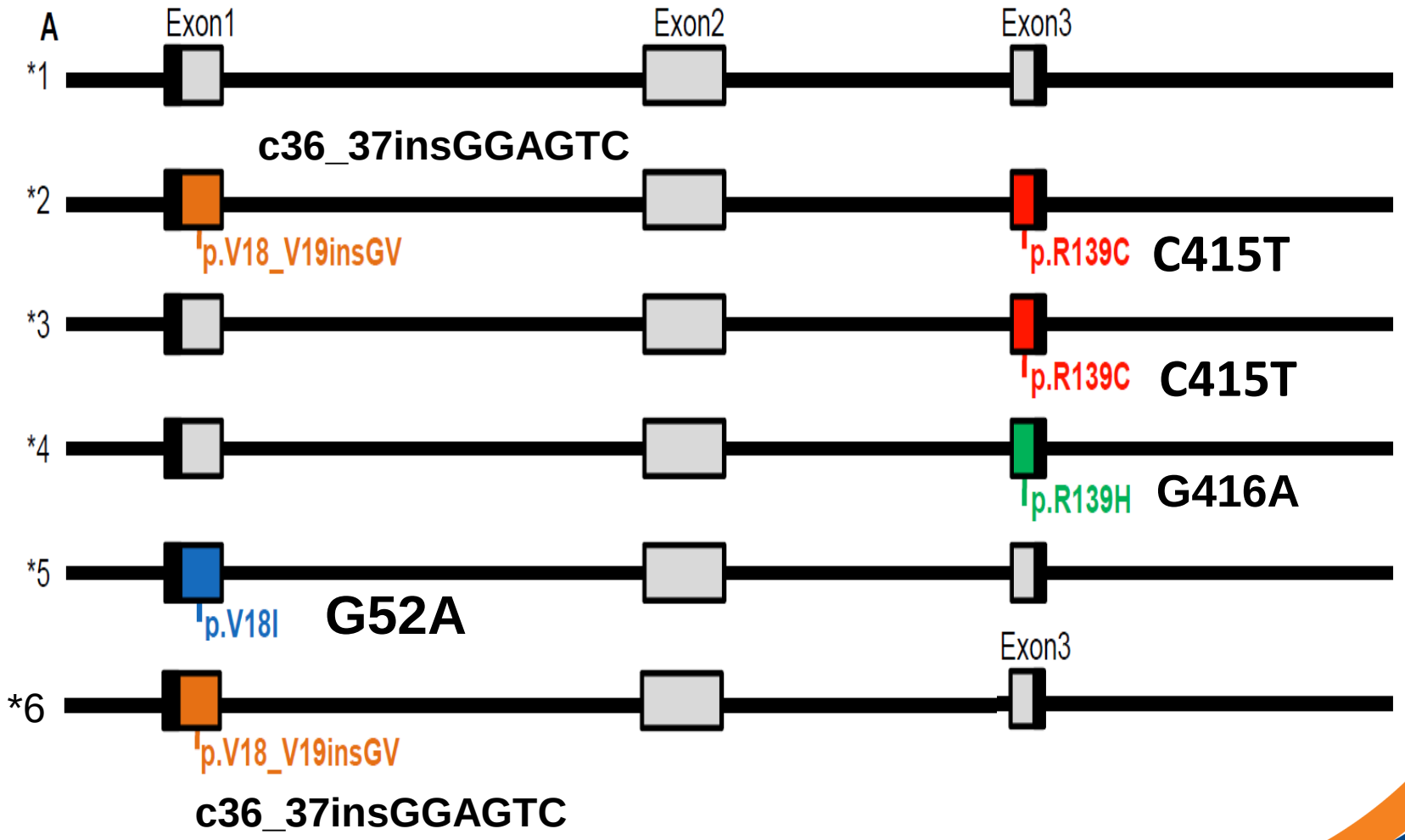
Moriyama et al. Nat Gen 2016

NUDT15 polymorphisms alter thiopurine metabolism and hematopoietic toxicity

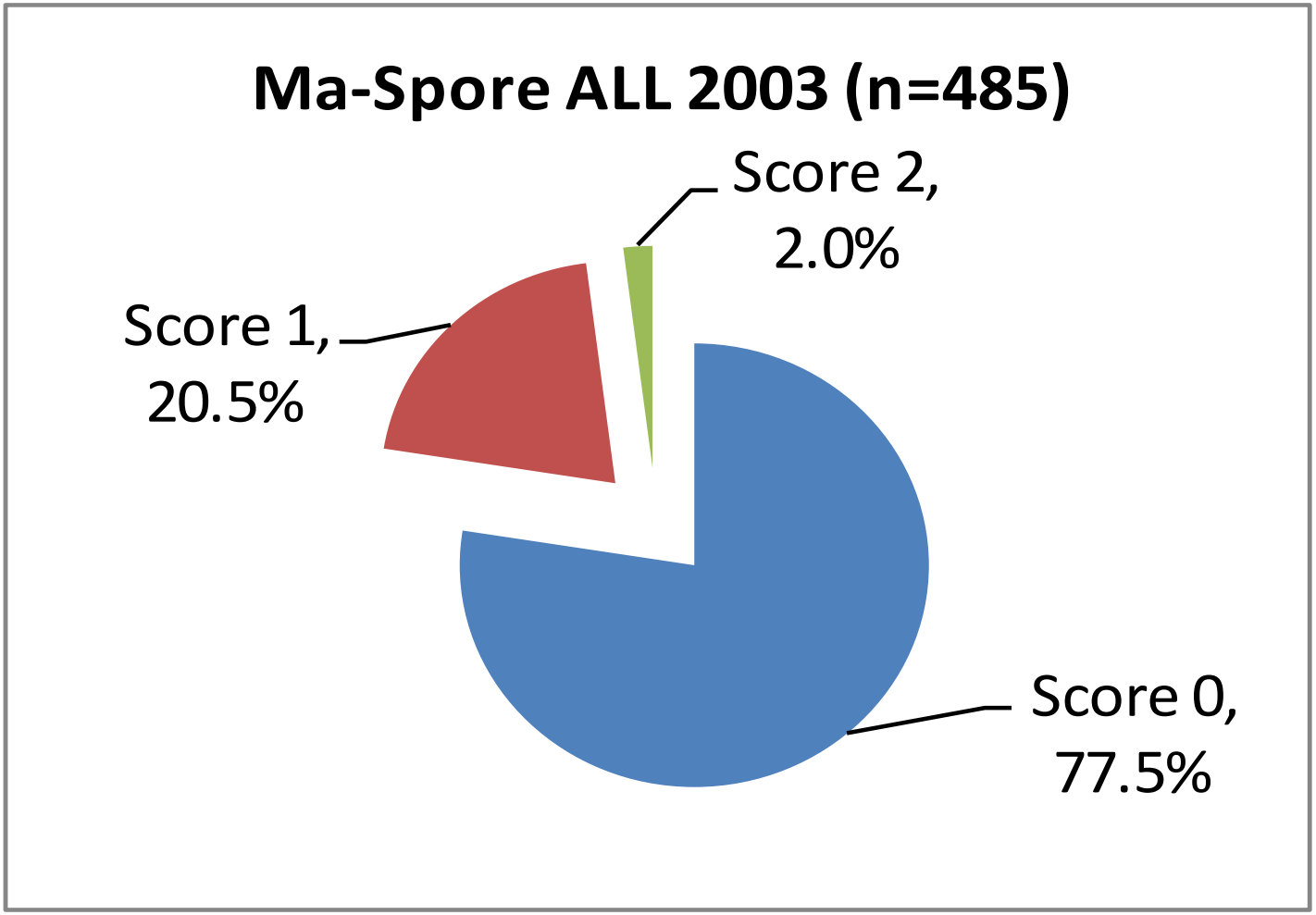
Takaya Moriyama[1,2], Rina Nishii[1,3,22], Virginia Perez-Andreu[1,22], Wenjian Yang[1], Federico Antillon Klussmann[4,5], Xujie Zhao[1], Ting-Nien Lin[1], Keito Hoshitsuki[1,6], Jacob Nersting[7], Kentaro Kihira[2], Ute Hofmann[8,9], Yoshihiro Komada[2], Motohiro Kato[10], Robert McCorkle[1], Lie Li[1], Katsuyoshi Koh[11], Cesar Rolando Najera[4], Shirley Kow-Yin Kham[12], Tomoya Isobe[13], Zhiwei Chen[12], Edwynn Kean-Hui Chiew[12], Deepa Bhojwani[14], Cynthia Jeffries[15], Yan Lu[15], Matthias Schwab[8,9,16,17], Hiroto Inaba[18], Ching-Hon Pui[18], Mary V Relling[1], Atsushi Manabe[19], Hiroki Hori[2], Kjeld Schmiegelow[7,20], Allen E J Yeoh[12,21], William E Evans[1] & Jun J Yang[1]

Widely used as anticancer and immunosuppressive agents, thiopurines have narrow therapeutic indices owing to frequent toxicities, partly explained by *TPMT* genetic polymorphisms. Recent studies identified germline *NUDT15* variation as another critical determinant of thiopurine intolerance, but the underlying molecular mechanisms and the clinical implications of this pharmacogenetic association remain unknown. In 270 children enrolled in clinical trials for acute lymphoblastic leukemia in Guatemala, Singapore and Japan, we identified four *NUDT15* coding variants (p.Arg139Cys, p.Arg139His, p.Val18Ile and p.Val18_Val19insGlyVal) that resulted in 74.4–100% loss of nucleotide diphosphatase activity. Loss-of-function *NUDT15* diplotypes were consistently associated with thiopurine intolerance across the three cohorts ($P = 0.021$, 2.1×10^{-5} and 0.0054, respectively; meta-analysis $P = 4.45 \times 10^{-8}$, allelic effect size = −11.5). Mechanistically, NUDT15 inactivated thiopurine metabolites and decreased thiopurine cytotoxicity *in vitro*, and patients with defective *NUDT15* alleles showed excessive levels of thiopurine active metabolites and toxicity. Taken together, these results indicate that a comprehensive pharmacogenetic model integrating *NUDT15* variants may inform personalized thiopurine therapy.

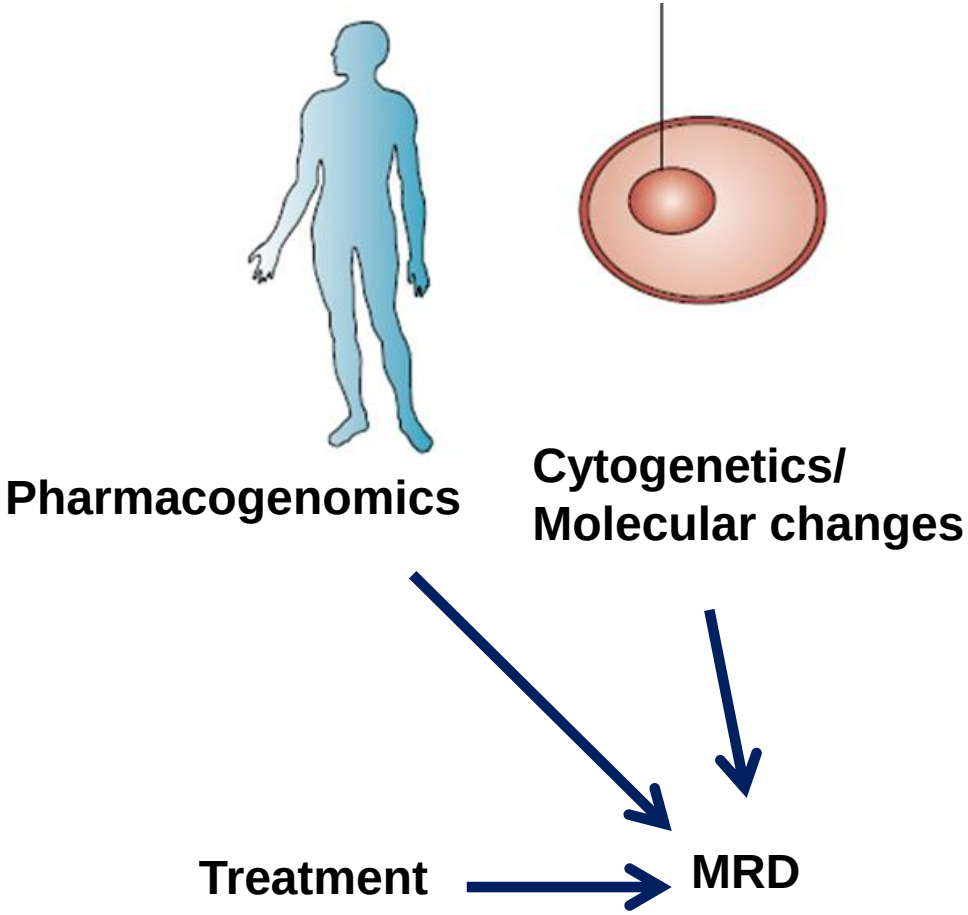
NUDT15 variants



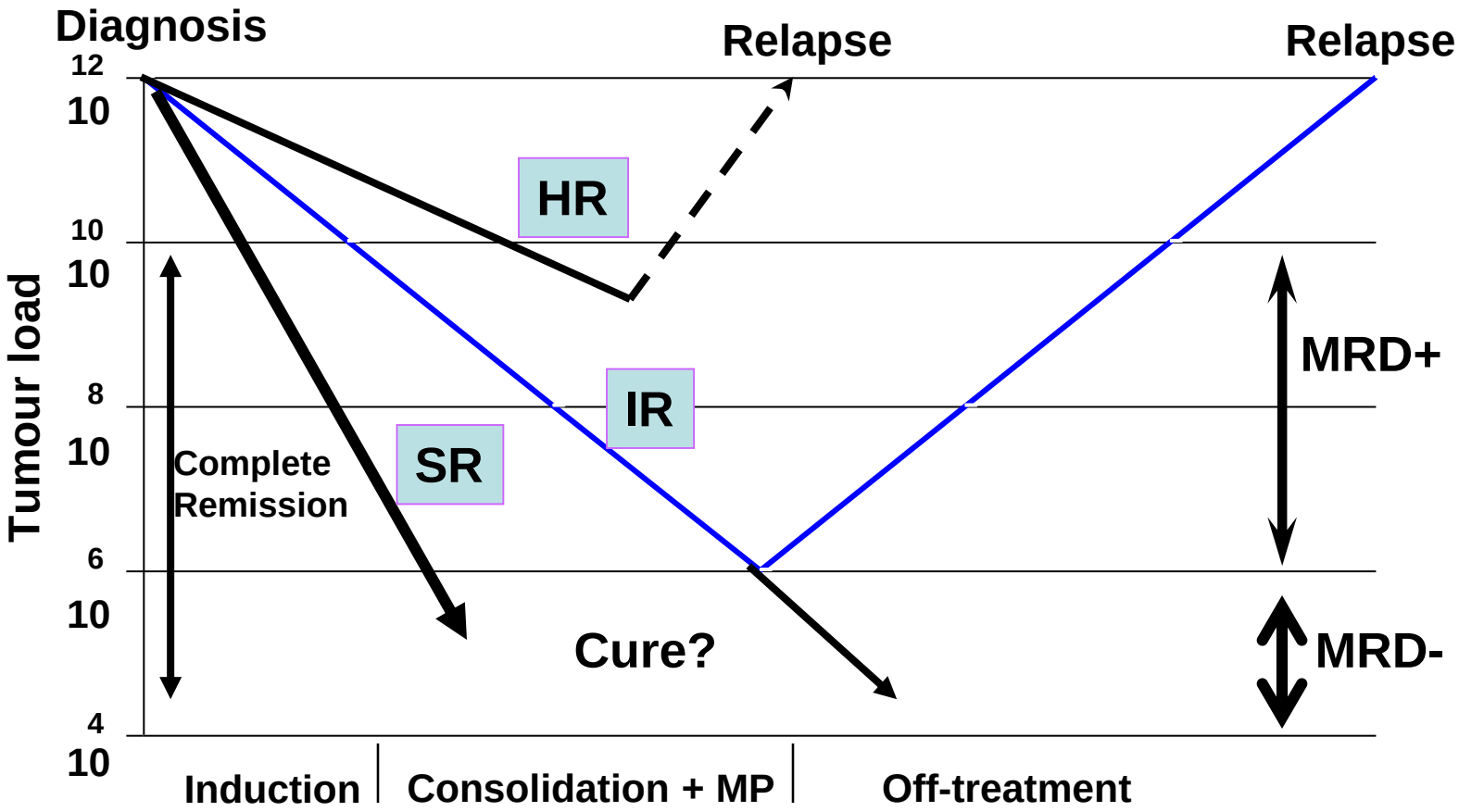
Combining NUDT15 and TPMT variants
Genetic score – 1 in 50 very sensitive to MP



Minimal Residual Disease – quantitating submicroscopic disease



MRD Kinetics – Flow or PCR MRD



JOURNAL OF CLINICAL ONCOLOGY

ORIGINAL REPORT

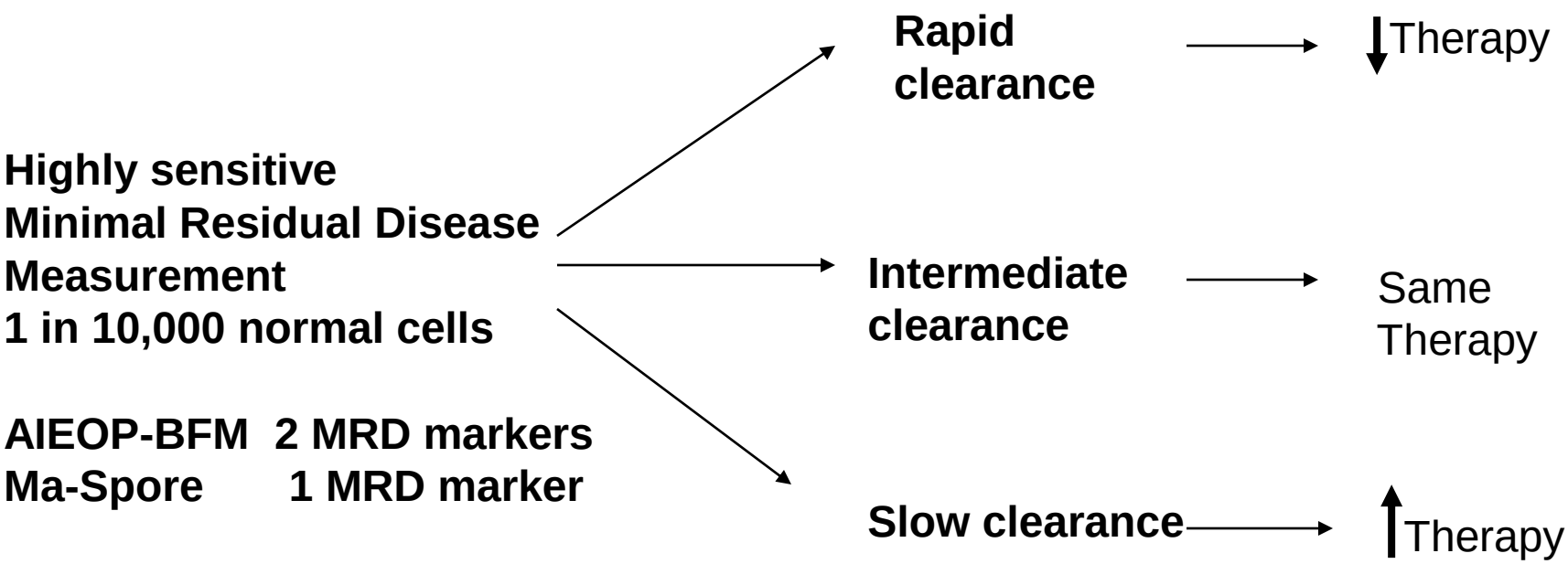
Minimal Residual Disease–Guided Treatment Deintensification for Children With Acute Lymphoblastic Leukemia: Results From the Malaysia-Singapore Acute Lymphoblastic Leukemia 2003 Study

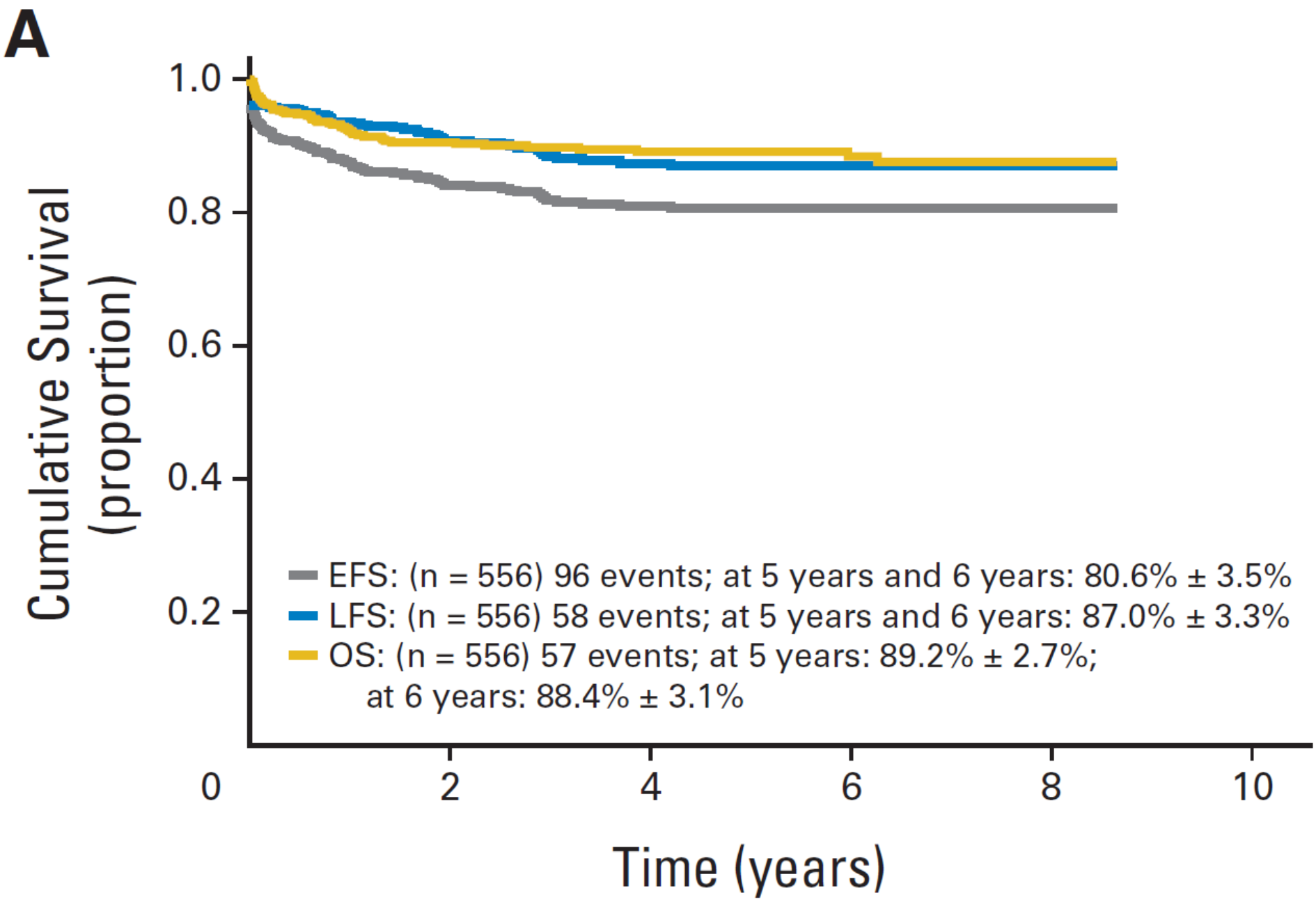
Allen Eng Juh Yeoh, Hany Ariffin, Elaine Li Leng Chai, Cecilia Sze Nga Kwok, Yiong Huak Chan, Kuperan Ponnudurai, Dario Campana, Poh Lin Tan, Mei Yoke Chan, Shirley Kow Yin Kham, Lee Ai Chong, Ah Moy Tan, Hai Peng Lin, and Thuan Chong Quah

A truly Asian effort – NUS, NUH, KKH – Singaporeans, Asian children

University Malaya – Hany Ariffin, Wan Ariffin and team ,
Sime Darby Medical Centre, KL – Lin Hai Pheng, Chan Lee Lee

Tailoring leukaemia treatment



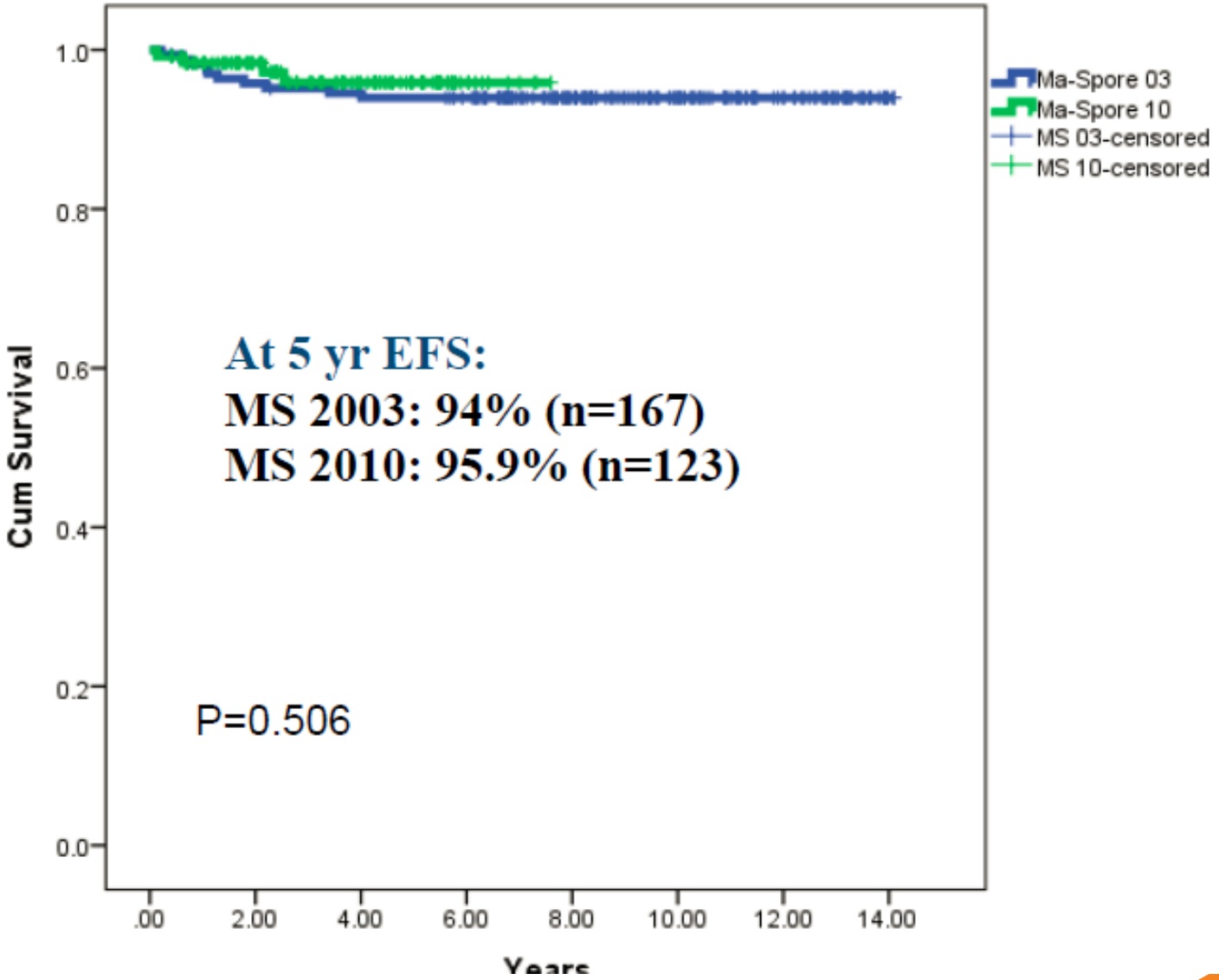

A
Cumulative Survival (proportion)
1.0
0.8
0.6
0.4
0.2
0
2
4
6
8
10
Time (years)
EFS: (n = 556) 96 events; at 5 years and 6 years: 80.6% ± 3.5%
LFS: (n = 556) 58 events; at 5 years and 6 years: 87.0% ± 3.3%
OS: (n = 556) 57 events; at 5 years: 89.2% ± 2.7%;
at 6 years: 88.4% ± 3.1%

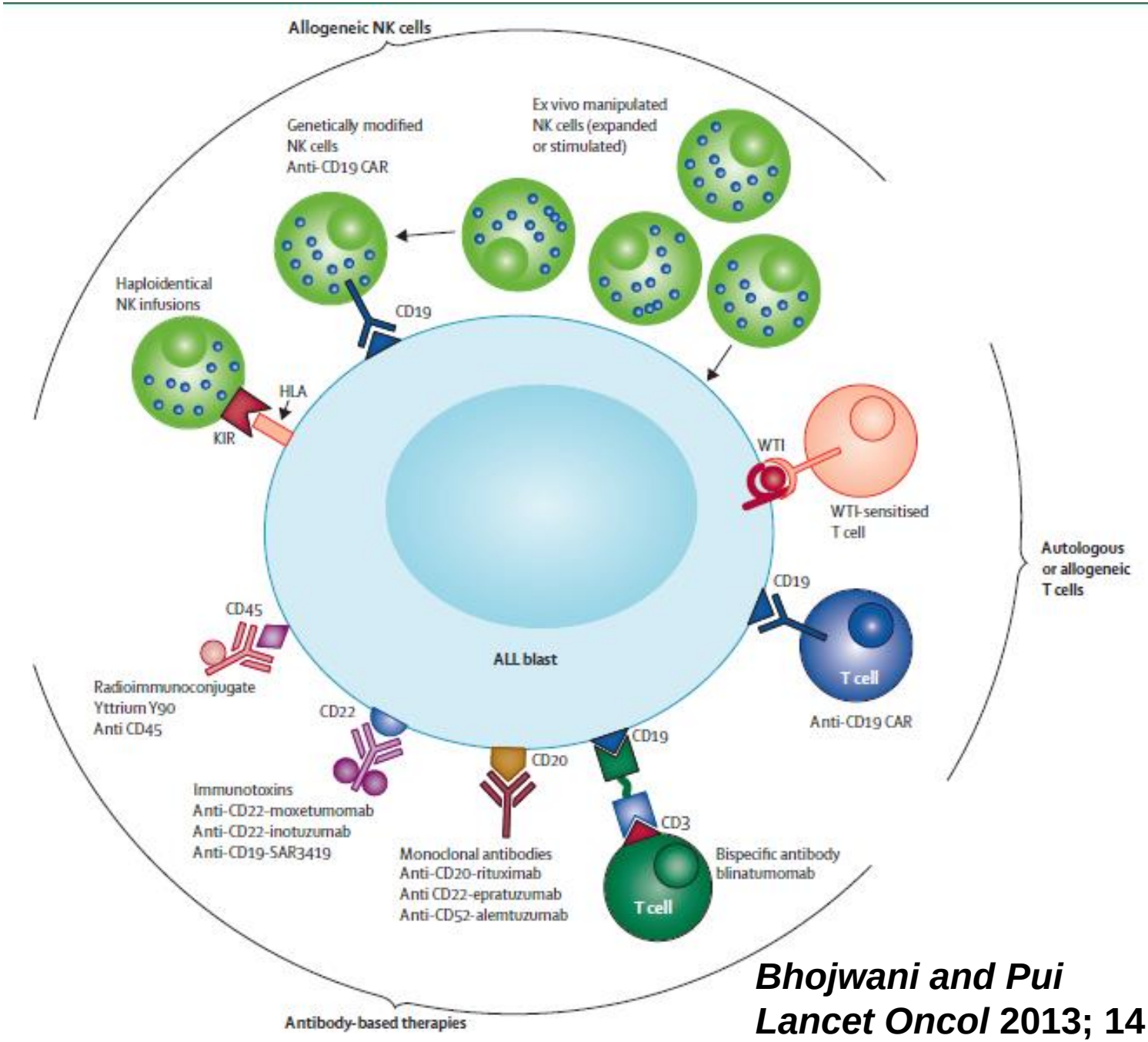
AIEOP-BFM-ALL 2000:
Increased Relapse After Lowering Intensity of Delayed Intensification in Negative MRD After Remission Induction

	4-yr DFS	4y DFS <10y	4y DFS >10y
III	91.8%	90.7	81.6
II	95.8%	92.5	90.3
P=	0.04	0.26	0.04

Schrappe et al. 2016 ASH

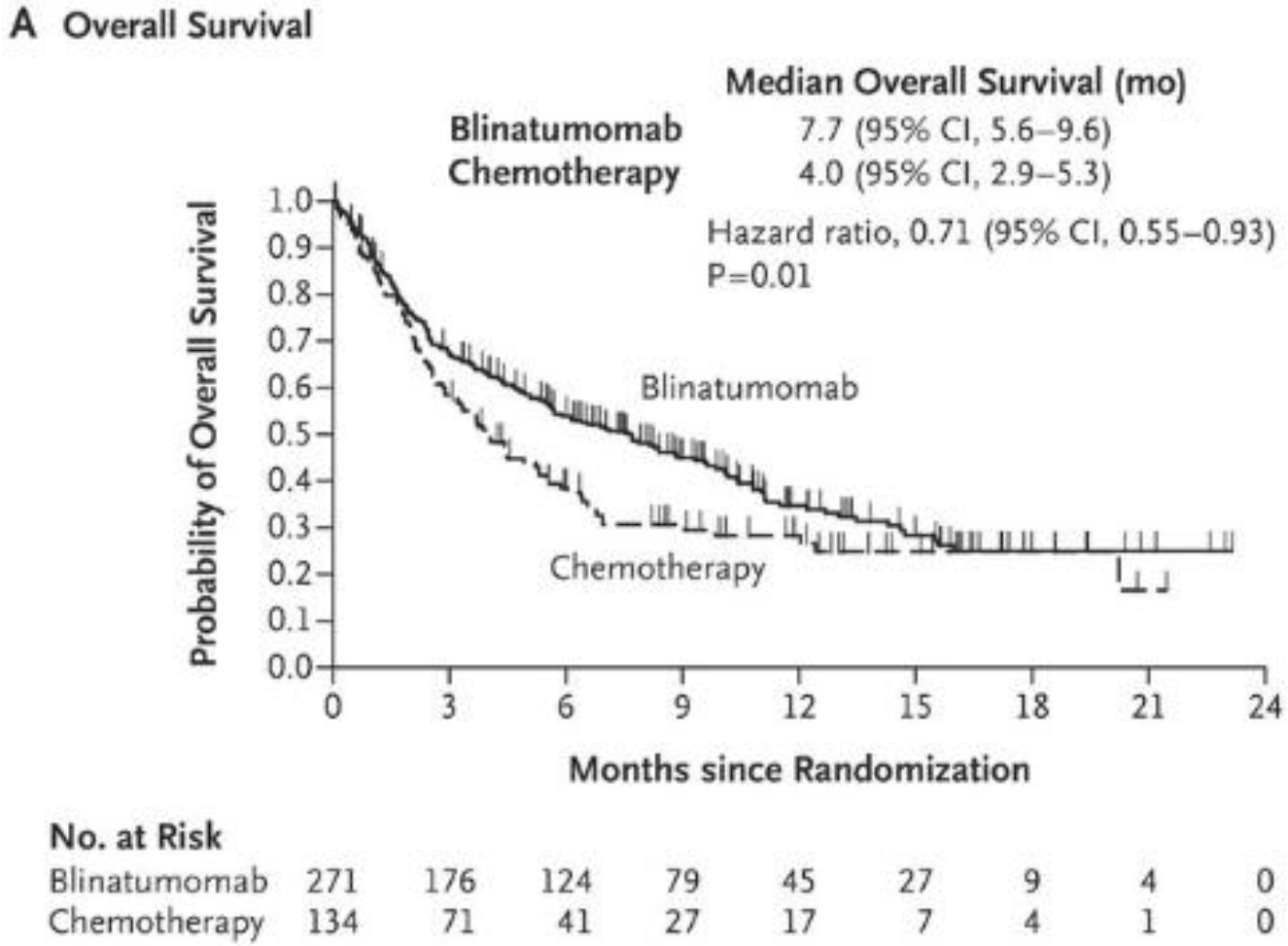
Figure 19: Event-Free Survival ALL 2003 vs 2010 by Ma-Spore 2010 Risk Stratification (SR)





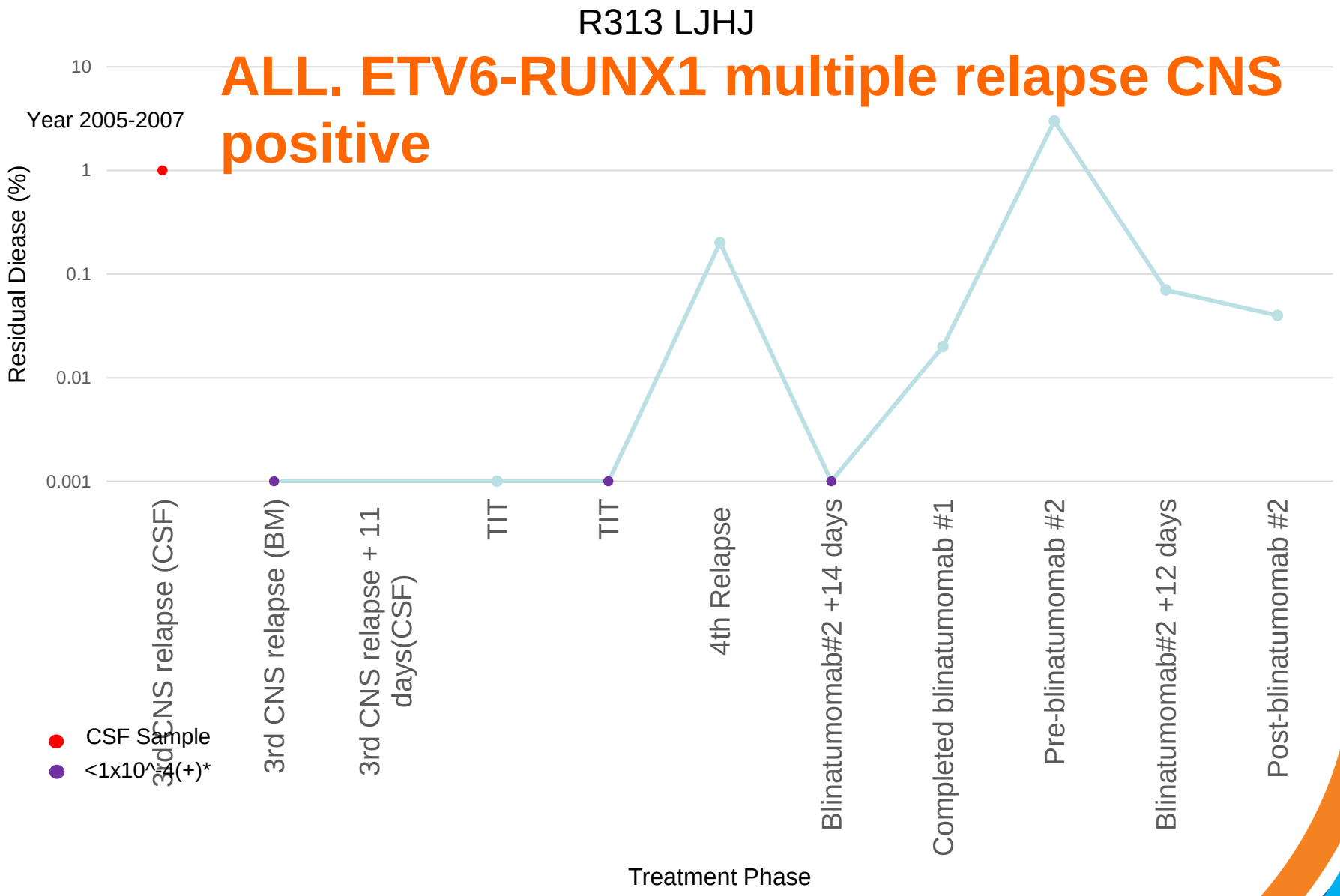
Bhojwani and Pui
***Lancet Oncol* 2013; 14: e205–17**

Outcome after Blinatumomab relapsed ALL

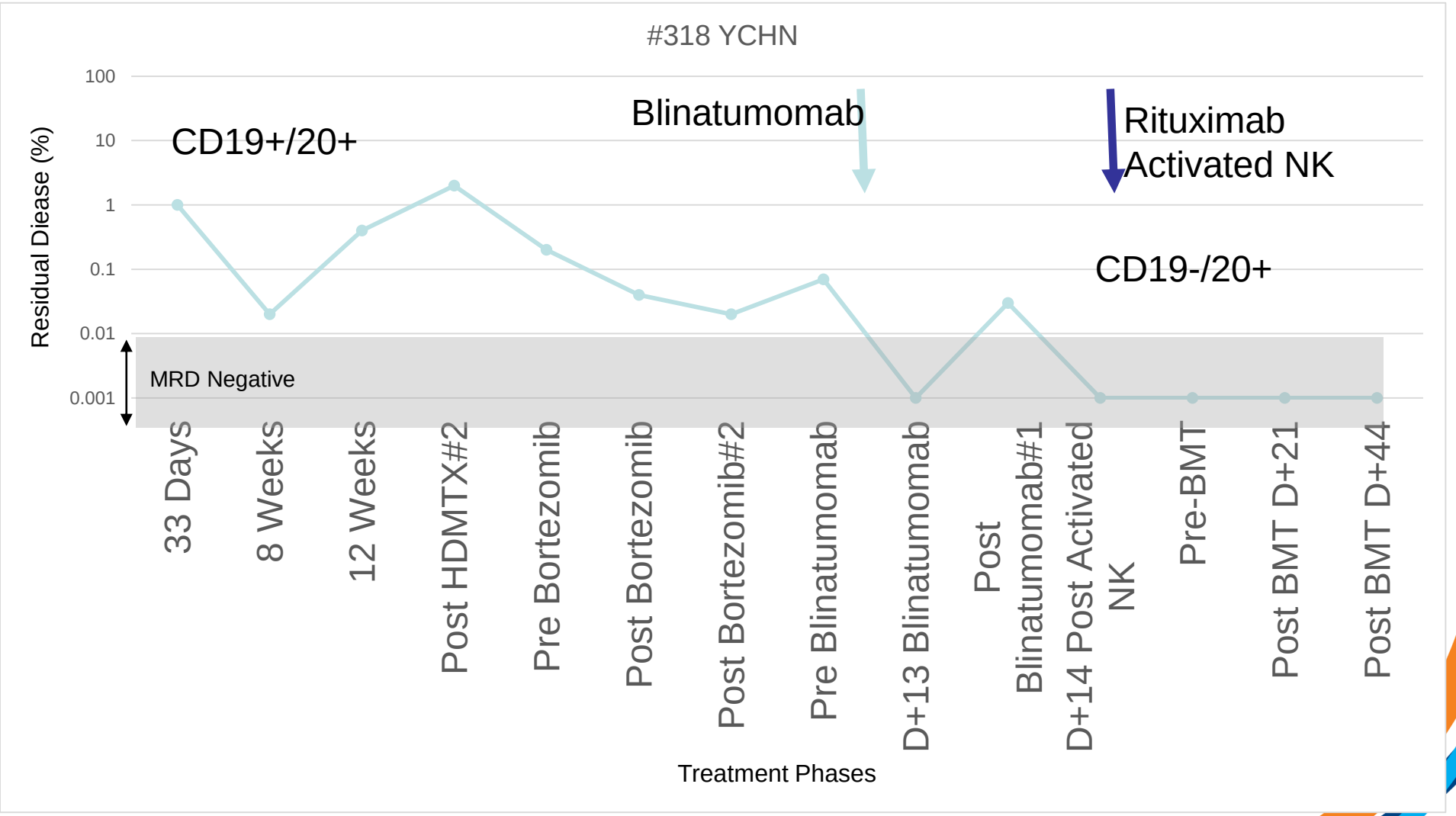


Kantarjian et al. New Engl J Med 2017

ALL. ETV6-RUNX1 multiple relapse CNS positive

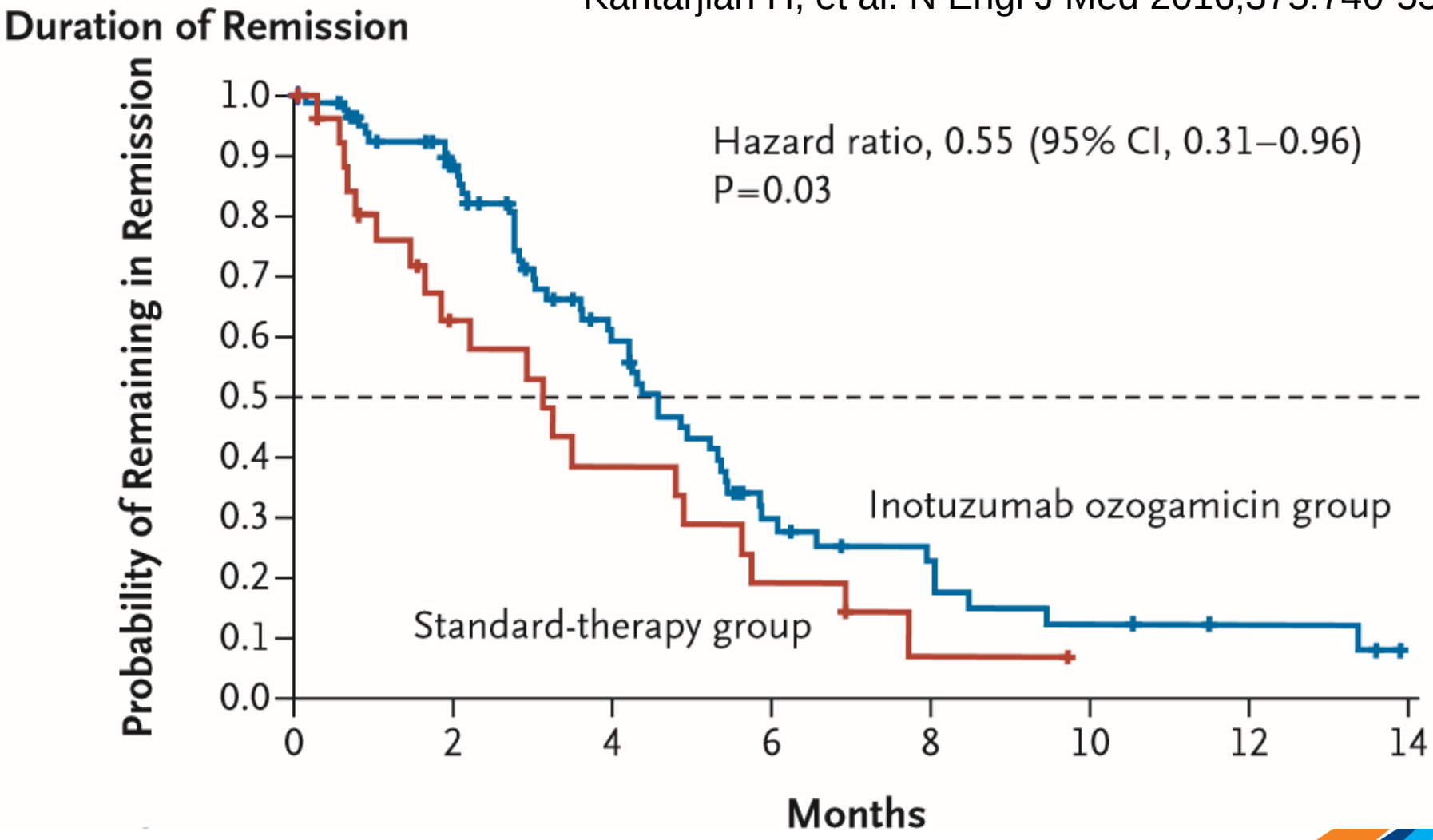


NCI HR, hypodiploid ALL, ?Li Fraumeni



Inotuzumab Ozogamicin in R/R ALL

Kantarjian H, et al. N Engl J Med 2016;375:740-53.



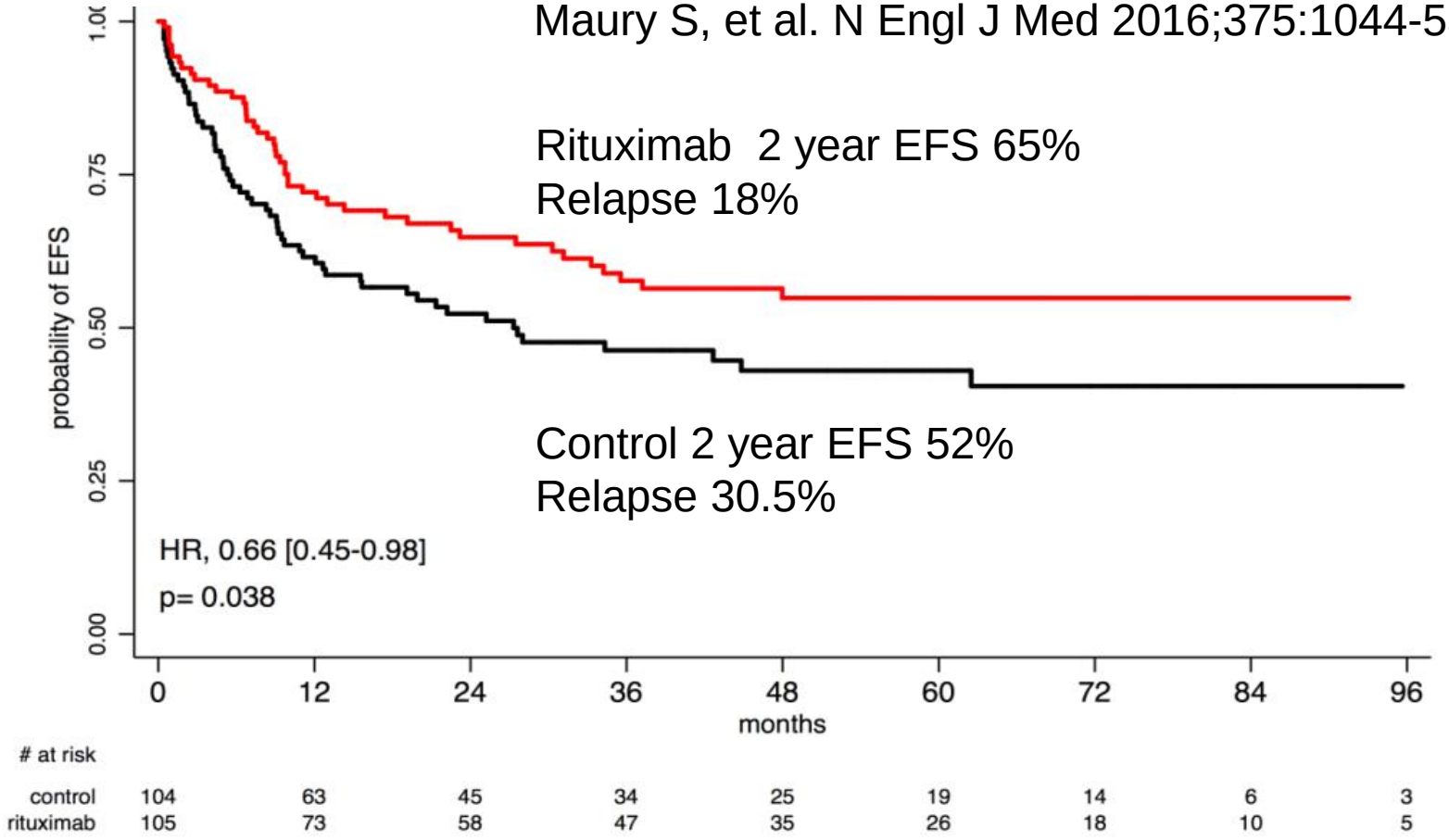
Rituximab Ph-ve/CD20+ ALL
GRAAL 2005

Ritux given in Induction, Consolidation, DI, Maintenance 1st yea
Total 16-18 doses

Maury S, et al. N Engl J Med 2016;375:1044-53.

Rituximab 2 year EFS 65%
Relapse 18%

Control 2 year EFS 52%
Relapse 30.5%



Every body love CARs

Responses to CAR T Cells in Acute Lymphoblastic Leukemia

Study	Pts. enrolled	Pts. with CR	Pts. with negative MRD
Davila et al. *Science Transl Med* 2014	16	14	12
Maude et al. *NEJM* 2014	30	27	22*
Lee et al. *Lancet* 2014	20	14	12
Total	**66**	**55 (83%)**	**46 (70%)**

*MRD studies not performed in 2 of the 27 patients who achieved CR

10 relapses, 5 CD19-negative

Immunotherapy anti CD19

Effective CAR – more than parts
CAR-T persistence - a few months
T-cell anergy
Quality of T-cell
– after multiple chemotherapy difficult
CAR-T grow faster &sustained > tumour
– Effector: Target
US$500k per dose

Personalised Medicine

Current

Morphology
+ cytochemistry
Flow cytometry
B vs T
Cytogenetics
Hyperdiploid > 50
Hypodiploid < 44

OFT
BCR-ABL1
MLL-AF4
E2A-PBX1
ETV6-RUNX1

Future

Real-time PCR
MRD
Multiparametric flow
Flow MRD

Pharmacogenomics
TPMT, NUDT15
Microarray GEP
Next generation sequencing
RNA seq
Exome profiling
Whole genome seq

Thank you

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