MRD and impact on Center Performance Measures

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Primary Questions

Should we incorporate MRD measures into Center performance?

1 year survival post Allogeneic HCT

How should data collection questions be modified to improve precision and prepare for future analyses?

Which diseases? Acute Leukemia

but recognize ALL and AML differently

- How are centers collecting data currently? Wael Saber
- MRD and technique sensitivity. ALL and AML. Stella Davies, Bart Scott
- Differing techniques for molecular testing & CHIP Chris Hourigan
- How should we use it in Center Performance Score
- Recommendation on:
  How should we revise the data collection forms &
  How should we use the data in the Center Specific Analysis of Outcome

What do we collect now and on which form in ALL/AML

- Molecular data/cytogenetic data at 3 time points: dx, between dx and HCT, and at HCT
- Single time point: flow cytometry to test for MRD at HCT only (if CR is achieved).
- No sensitivity threshold is asked
- In AML, molecular panel asked now includes: FLT3-ITD, FLT3-TKD, IDH1/2, CEBPA, KIT, NPM1, Others
- In ALL, molecular panel includes: BCR/ABL, TEL-AML/AML1, Others
- Disease classification form (f2402)

MRD testing according to center volume

	High volume	Low volume	P Value
MRD testing by center volume, AML in CR1/CR2			
No. of patients	6107	1666	
MRD testing			0.03[a]
No	407 (7)	86 (5)	
Yes	5700 (93)	1580 (95)	
MRD testing by center volume, ALL in CR1/CR2			
No. of patients	2734	828	
MRD testing			0.39[a]
No	110 (4)	39 (5)	
Yes	2624 (96)	789 (95)	

Hypothesis testing: [a] Pearson chi-square test

TED; first all for all indications; US only; 2017-2019

MRD testing according to center volume

	High volume	Low volume
AML in CR1/CR2		
No. of patients	5700	1580
MRD testing method		
Flow only	2547 (45)	788 (50)
NGS/PCR only	358 (6)	28 (2)
Both	2795 (49)	764 (48)
ALL in CR1/CR2		
No. of patients	2624	789
MRD testing method		
Flow only	1430 (54)	474 (60)
NGS/PCR only	78 (3)	17 (2)
Both	1116 (43)	298 (38)

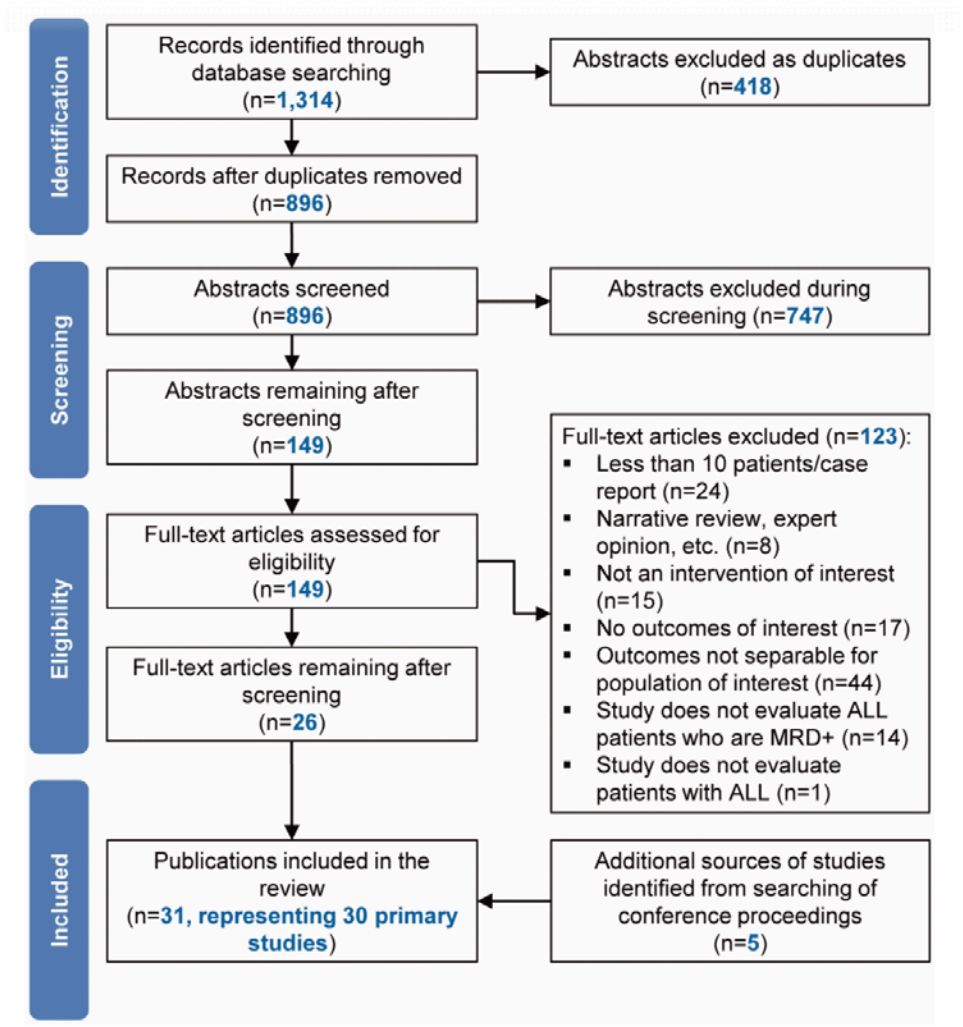
LEUKEMIA & LYMPHOMA
2020, VOL. 61, NO. 5, 1052-1062
https://doi.org/10.1080/10428194.2019.1709834

Taylor & Francis
Taylor & Francis Group

A systematic review of outcomes after stem cell transplantation in acute lymphoblastic leukemia with or without measurable residual disease

Shweta Shah[a], Amber Martin[b], Monica Turner[b], Ze Cong[a], Faraz Zaman[a], and Anthony Stein[c]

[a] Amgen Inc., Thousand Oaks, CA, USA; [b] EVIDERA, Evidence, Synthesis, Modeling, and Communications, Waltham, MA, USA; [c] City of Hope National Medical Center, Duarte, CA, USA



Hazard Ratios for OS in Adults with ALL With and Without MRD

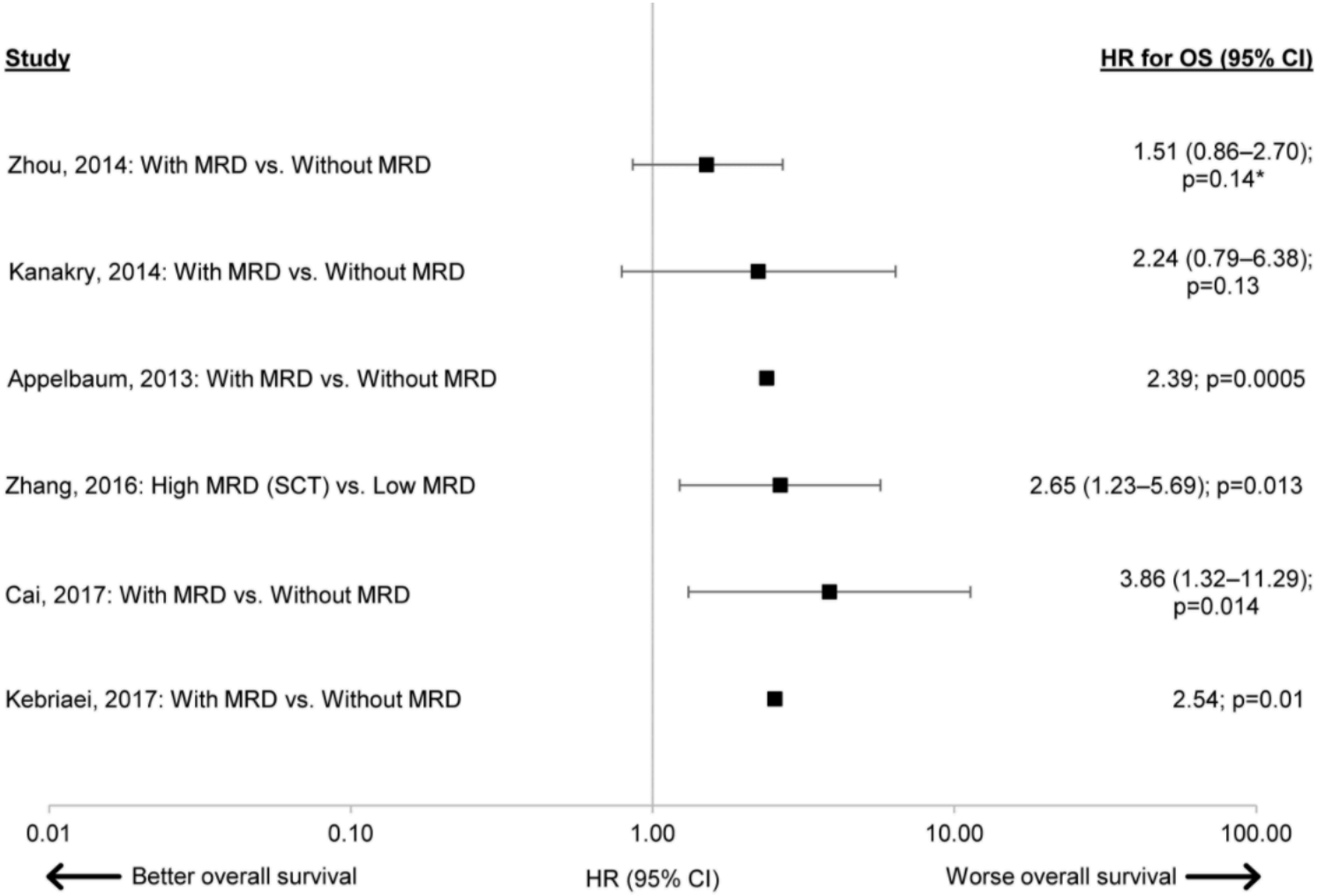


Figure 2. Available hazard ratios for overall survival. CI: confidence interval; HR: hazard ratio; MRD: measurable residual disease; OS: overall survival; SCT: stem cell transplantation.

Shah et al, 2020

Outcome Of Allo HSCT for Adults with ALL in CR1

Time point	Measure	No. of studies	Range in patients with MRD	Range in patients without MRD
Median	Median OS	One [15]	1.98 months	Not reached
	Median RFS	One [23]	6.5 months	Not reached
	Median DFS	One [15]	1.16 months	Not reached
2-year results	OS rate	Three [16,17,19]	37–57.7%	68–81.9%
	RFS rate	Two [16,19]	40.2–57%	61–70.3%
	DFS rate	Two [24,25]	54%	52–66%
3-year results	OS rate	Two [14,21]	27–64%	68–82%
	DFS rate	One [21]	27%	73%
5-year results	OS rate	Three [15,18,20]	33–53%	58–75%
	DFS rate	Three [15,20,25]	10–41%	47–72%
10-year results	DFS rate	One [26]	30%	35%

CR1: first complete remission; DFS: disease-free survival; MRD: measurable residual disease; OS: overall survival; RFS: relapse-free survival.

Shah et al, 2020

Outcome of Allo HSCT for Adults with ALL in CR2

Time point	Measure	No. of studies	Range in patients with MRD	Range in patients without MRD
Mean survival	DFS	One [28]	36–52 months	35–82 months
Median	Median OS	Four [29–31,34]	8–17 months	7 months to not reached
	Median RFS	One [34]	10.5 months	51 months
	Median EFS	Two [29,31]	6–7 months	5–18 months
2-year results				
	DFS rate	One [37]	61.2%	74.4%
	EFS rate	Two [29,31]	0–19%	7–46%
	PFS rate	One [32]	28%	47%
3-year results				
	PFS rate	One [27]	29.6%	28.9%
	DFS rate	Three [28,36,38]	27–50%	40–73.9%
6-year results	DFS rate	One [39]	24%	74%
10-year results	DFS rate	One [26]	23%	32%

Shah et al, 2020

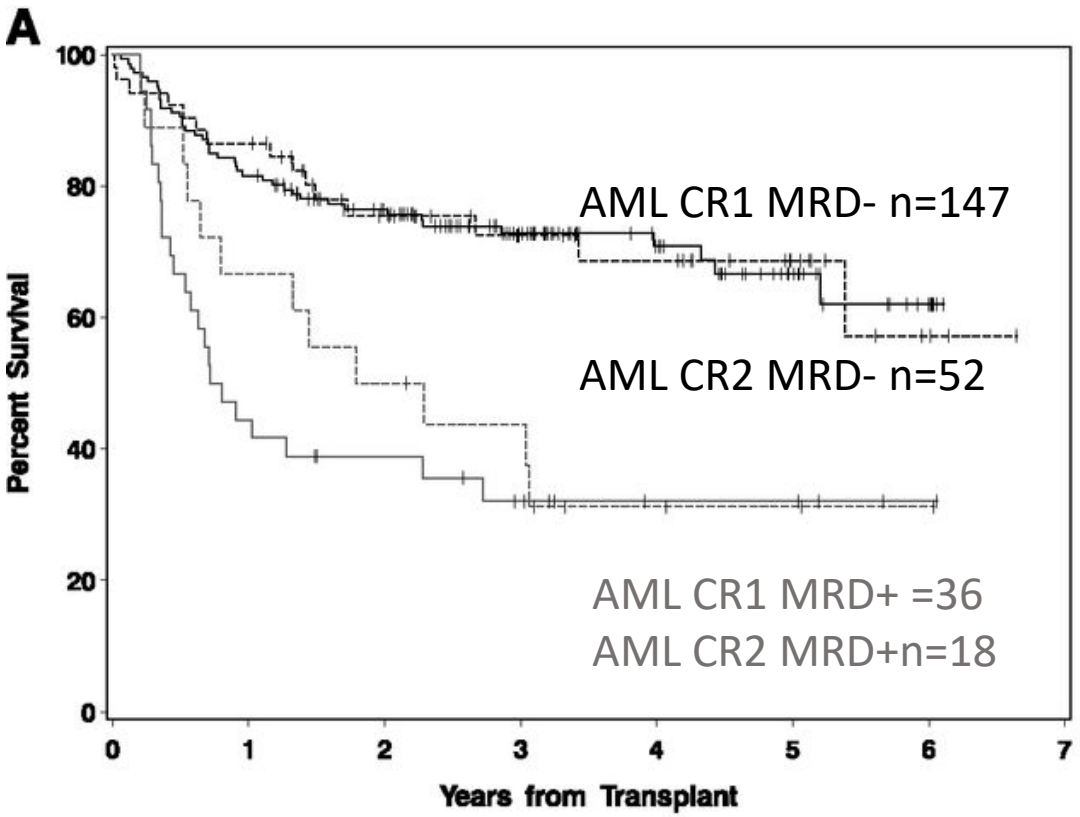
Pediatric ALL

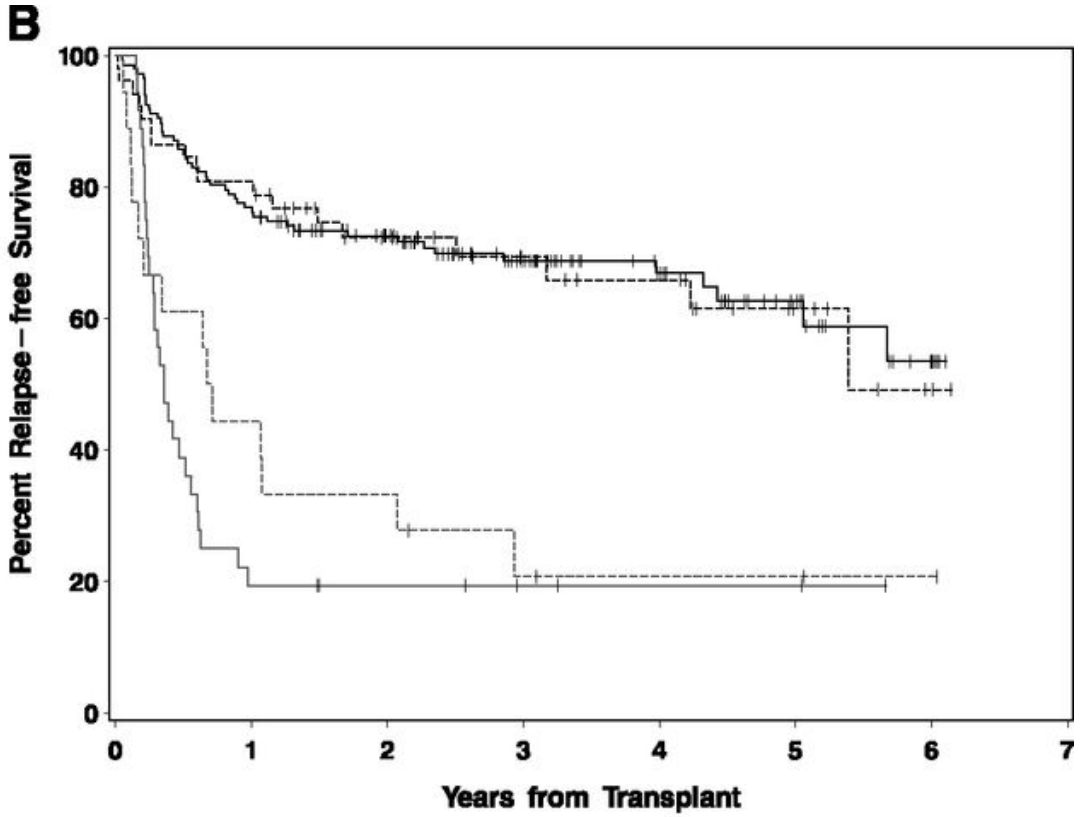
Adam Lamble , Rachel Phelan and Michael Burke, 2017

Table 2. Studies supporting the prognostic significance of MRD prior to HSCT.

Author	Year	Study Type	Technique	Sensitivity	*N*	Age, Years, Median (Range)	Remission	Results
Knechtli [32]	1998	R	PCR	$<10^{-3}$–10^{-5}	64	<18	CR1, CR2	2-year EFS 73% MRD− vs. 0% MRD+ $p < 0.001$
Van der Velden [33]	2001	R	PCR	$<10^{-4}$	17	<15	CR1, CR2	5-year RFS 80% MRD− vs. 33% MRD+
Sanchez [34]	2002	P	MCF	$<10^{-4}$	24	18 (3–49)	≥CR1	2-year RFS 73% MRD− vs. 33% MRD+ $p = 0.03$
Bader [35]	2002	R	PCR	$<10^{-4}$	41	9.8 (1.5–17.8)	≥CR1	5-year EFS 78% MRD− vs. 32% MRD+ $p = 0.011$
Krejci [36]	2003	R	PCR	$<10^{-4}$	140	<19	≥CR1	5-year EFS 75.2% MRD− vs. 29.8% MRD+
Imashuku [37]	2003	P	PCR	$<10^{-4}$	95	9 (0.3–20)	Not remission, ≥CR1	Available data in 19 relapses, all 19 were MRD+
Goulden [38]	2003	R	PCR	$<10^{-4}$	64	Pediatric	≥CR1	3-year EFS 73% MRD− vs. 17% MRD+ $p < 0.001$
Sramkova [39]	2007	P	PCR	$<10^{-4}$	25	1.1–19	Partial remission, CR1, CR2	EFS 94% MRD− vs. 13% MRD+ $p < 0.001$
Paganin [40]	2008	P	PCR	$<10^{-4}$	60	5 (0.6–17)	CR2	3-year EFS 73% MRD− vs. 19% MRD+ $p < 0.05$
Bader [41]	2009	P	PCR	10^{-4}	91	11.1 (3–22.6)	CR2, CR3	3-year EFS 60% MRD− vs. 27% MRD+
Elorza [42]	2010	P	MCF	10^{-4}	31	7 (<1–16)	≥CR1	2-year EFS 74% MRD− vs. 20% MRD+
Leung [43]	2012	R	MFC	10^{-4}	64	11.3 (0.6–25.1)	≥CR1	5-year OS 87.5% MRD− vs. 48.5% MRD+
Ruggeri [44]	2012	R	PCR/MFC	10^{-3-5}	170	6.5 (<1–17)	CR1,CR2, CR3	4-year CIR 24% MRD− vs. 39% MRD+
Bachanova [45]	2012	P	MFC	10^{-3}	86	20 (6–63)	CR1, CR2, CR3	2-year RR 26% MRD− vs. 30% MRD+
Shah [46]	2014	R	MFC	10^{-4}	34	<21	CR2	RR 35% MRD− vs. 64% MRD+
Balduzzi [47]	2014	P	PCR	10^{-4}	82	8 (<1–20)	CR1, CR2, CR3	5-year EFS 77.7% MRD− vs. 30.8% MRD+ $p < 0.001$
Bar [48]	2014	R	MCF	10^{-3}–10^{-4}	153 (62 ped)	24.6 (0.6–61.8)	≥CR1	3-year EOR 17% MRD− vs. 38% MRD+

Pre-HCT MRD in AML



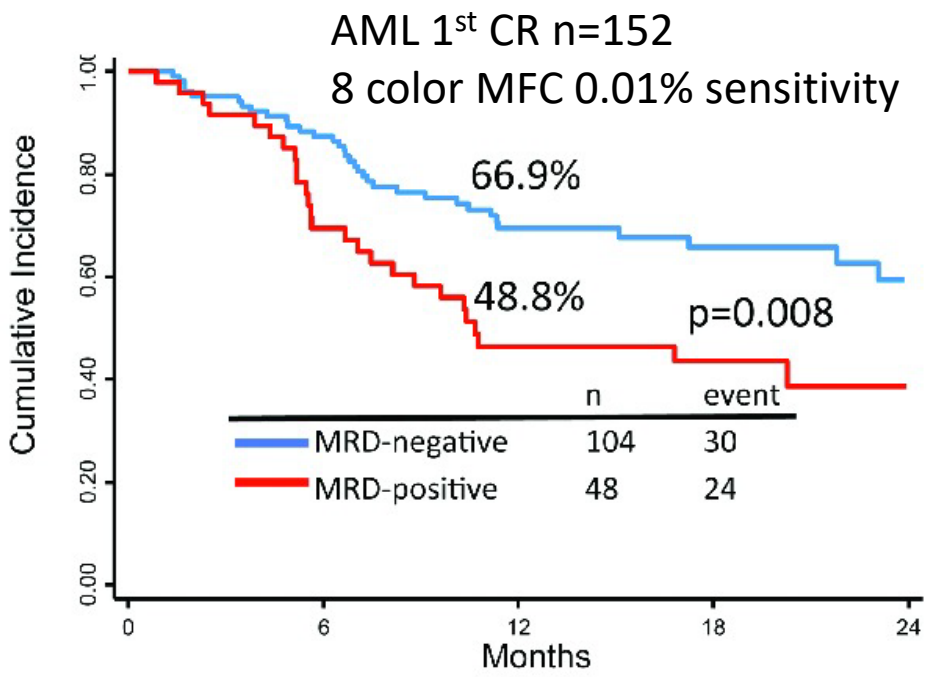


MAC: Bu4, H-TBI, Treo, RAB

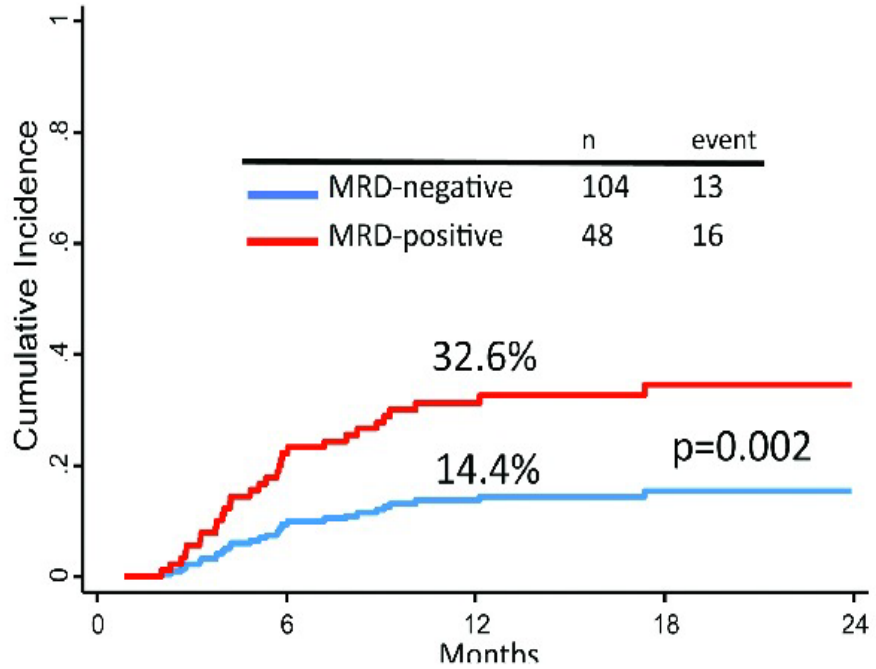
Walter et al. *Blood* 2013;122:1813-1821

Pre-HCT MRD in AML

Overall Survival

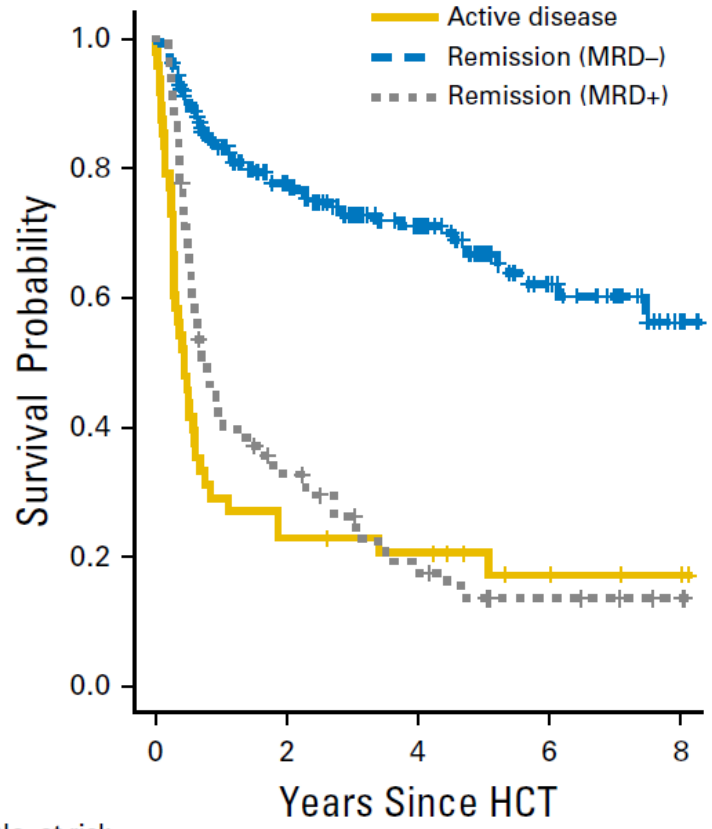


Relapse

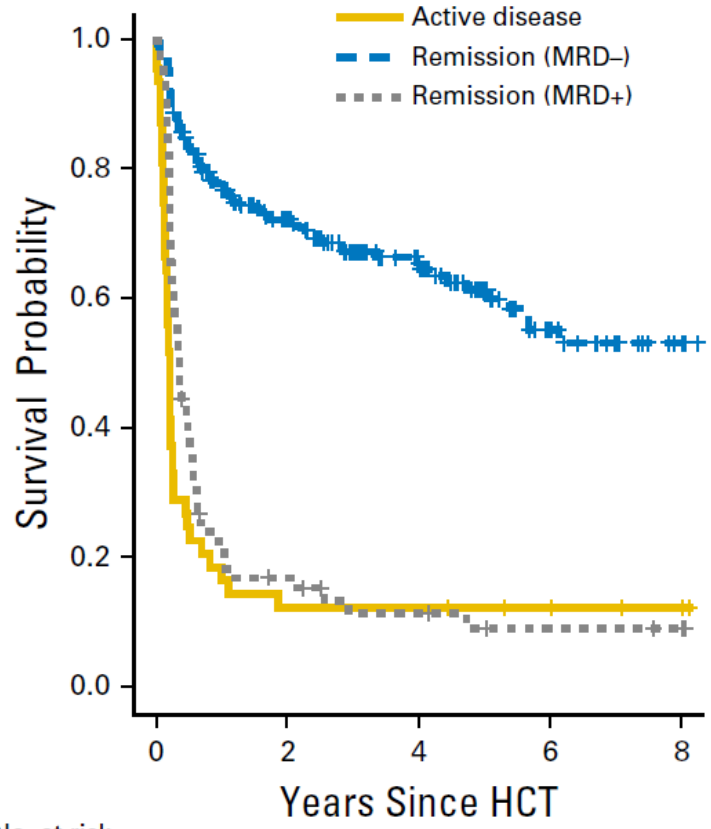


Oran et al. *Haematologica* 2017;102

Pre-HCT MRD in AML



No. at risk					
Active disease	48	11	9	4	2
Remission (MRD–)	235	136	80	34	8
Remission (MRD+)	76	22	11	5	2

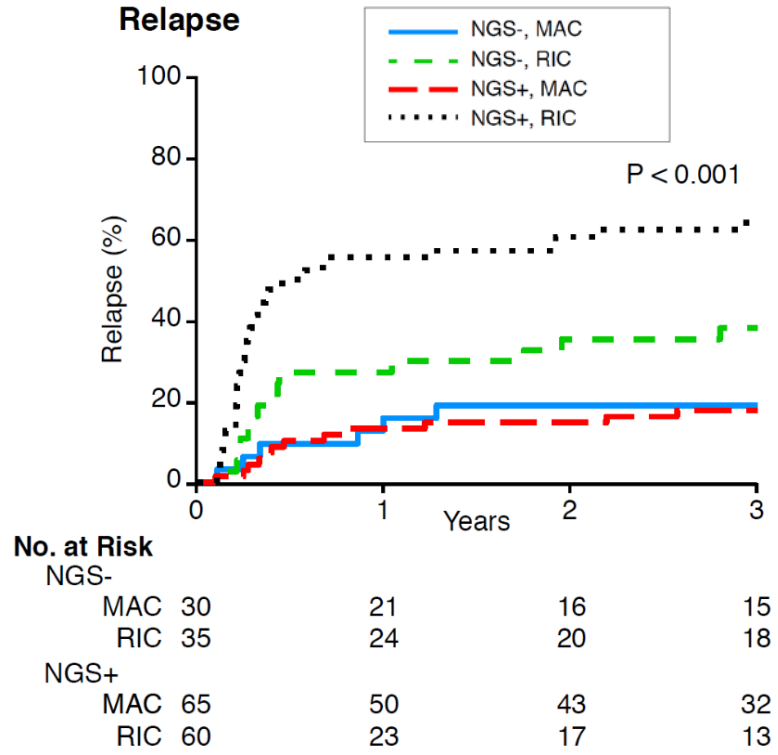


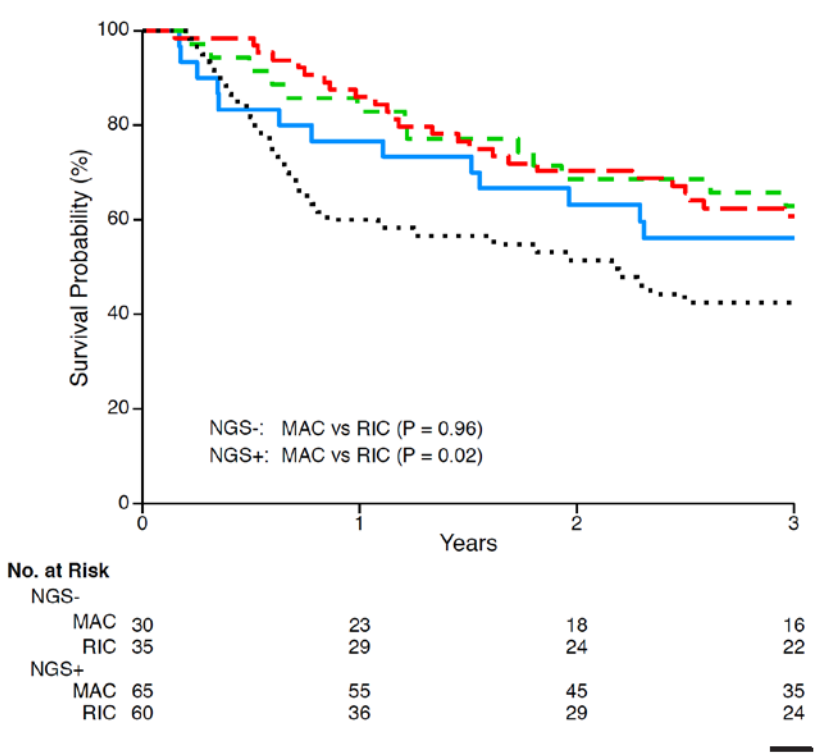
No. at risk					
Active disease	48	6	6	4	2
Remission (MRD–)	235	127	73	29	7
Remission (MRD+)	76	11	6	3	2

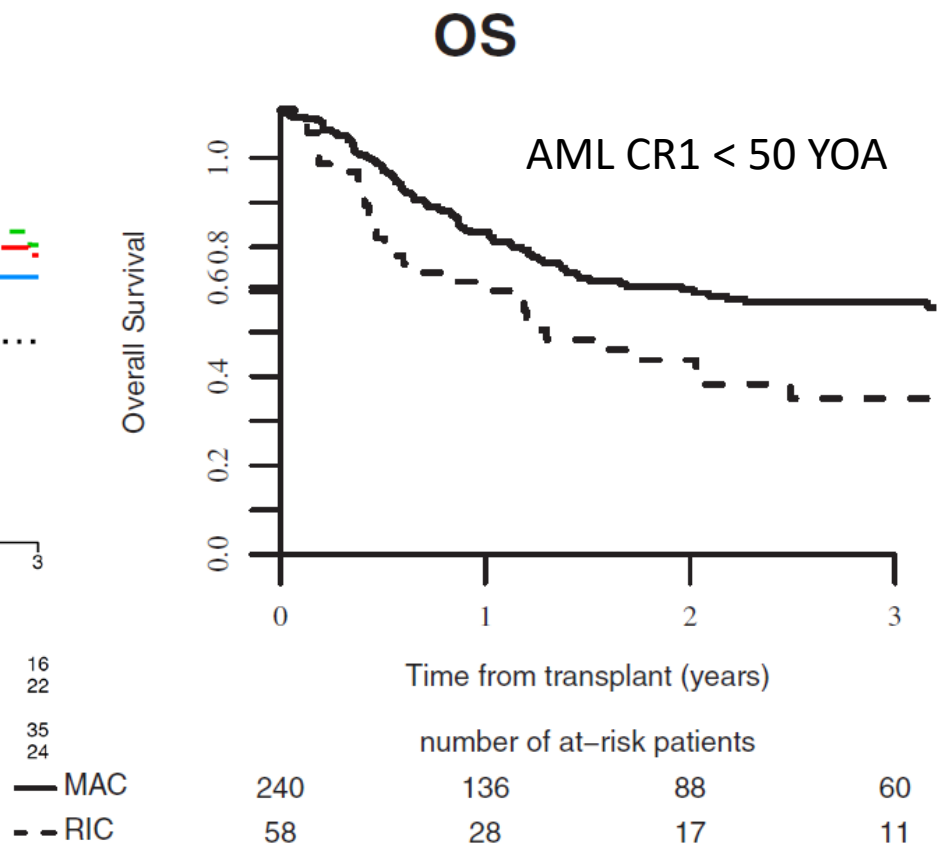
359 AML MAC HCT 2006-2014
Bu4, H-TBI, Treo, RAB

Araki et al. *J Clin Oncol* 2015;34:329-336

MRD Modifies Effect of Conditioning Intensity



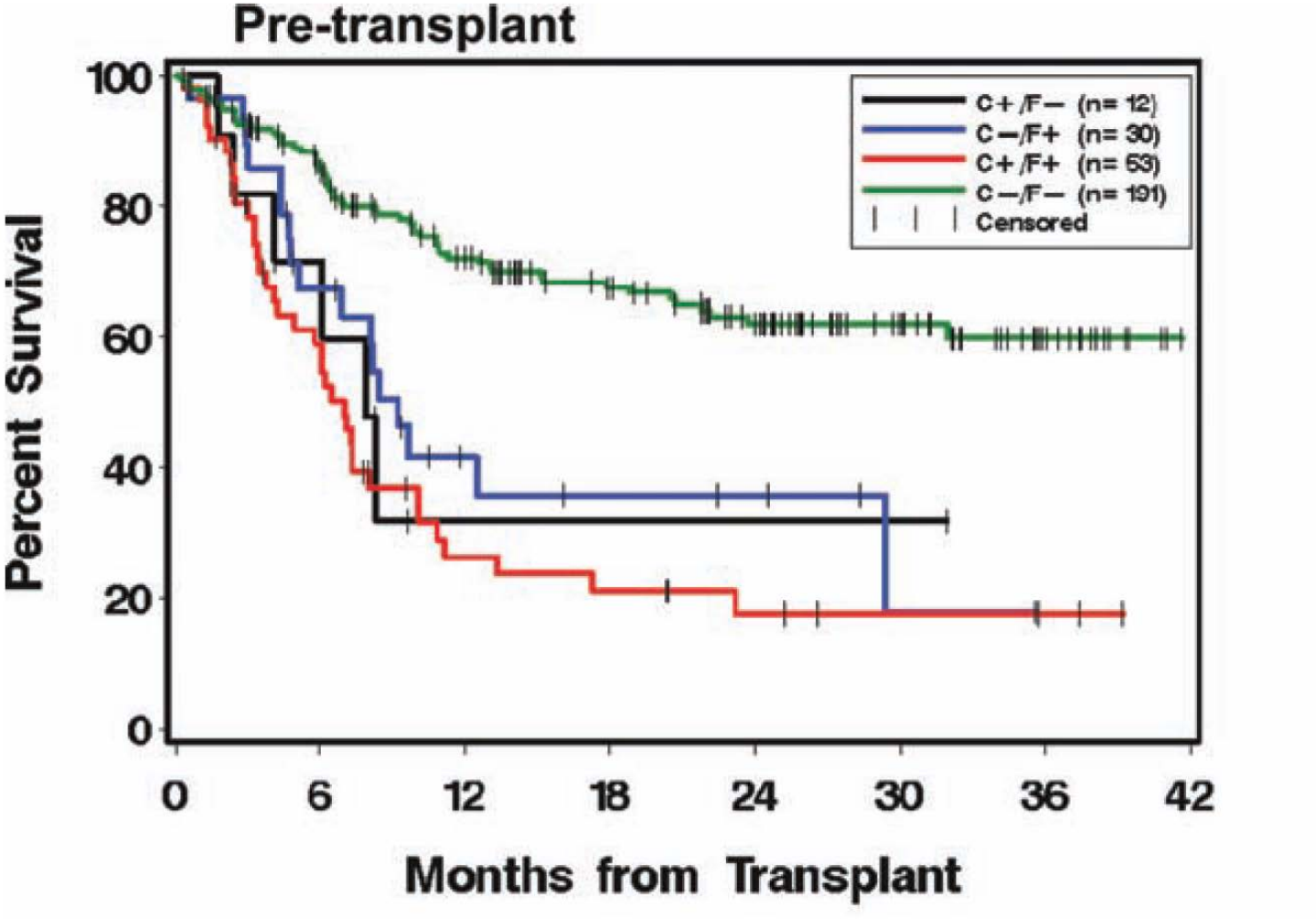




190 of 218 AML patients
51kB multiplex PCR targeting 13 genes
VAF as low as 0.1% (1/1000), or
0.02% (1/5000) for insertions in mutated *NPM1* and *FLT3*-ITD.

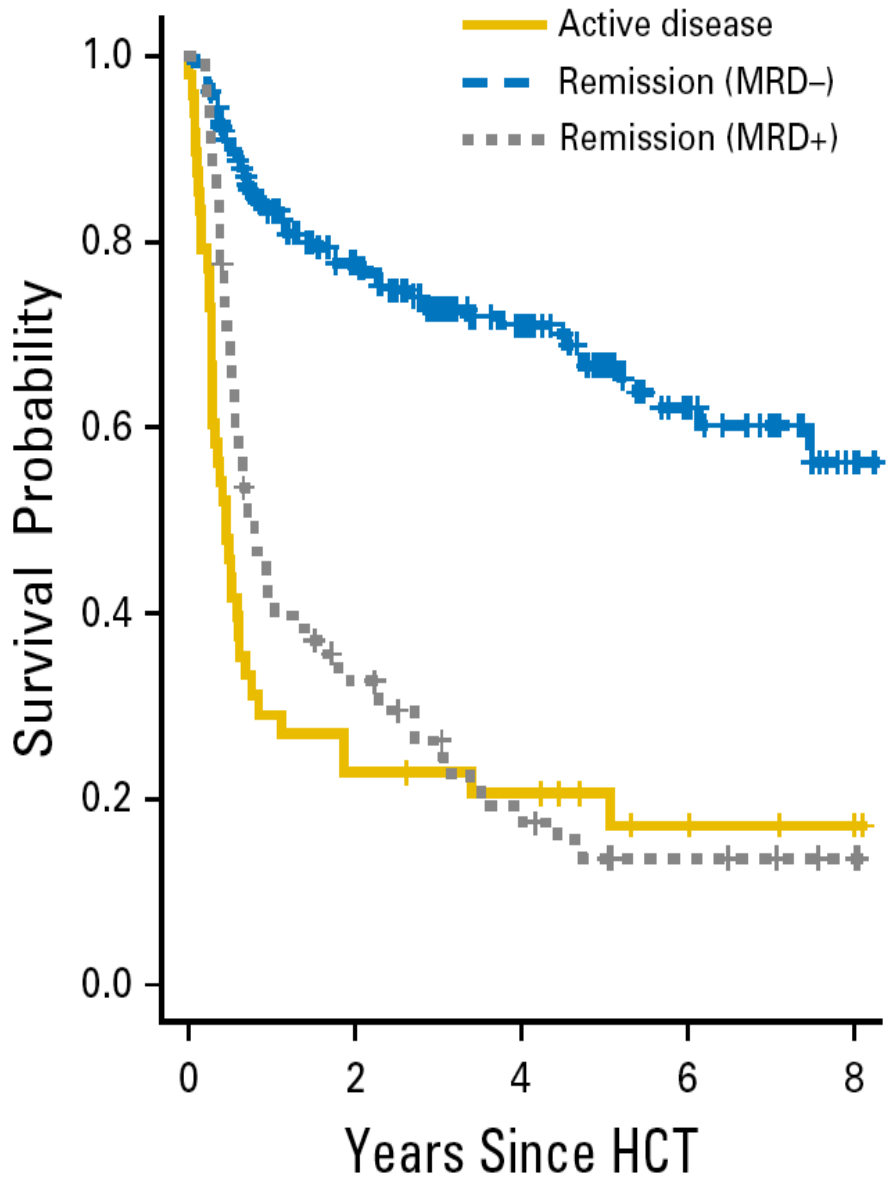
Hourigan et al. *J Clin Oncol*. 2019;38:1273-1283
Gilleece et al. *Am J Hematol*. 2018;93:1142-1152

Does the Method of Detection of MRD Matter?

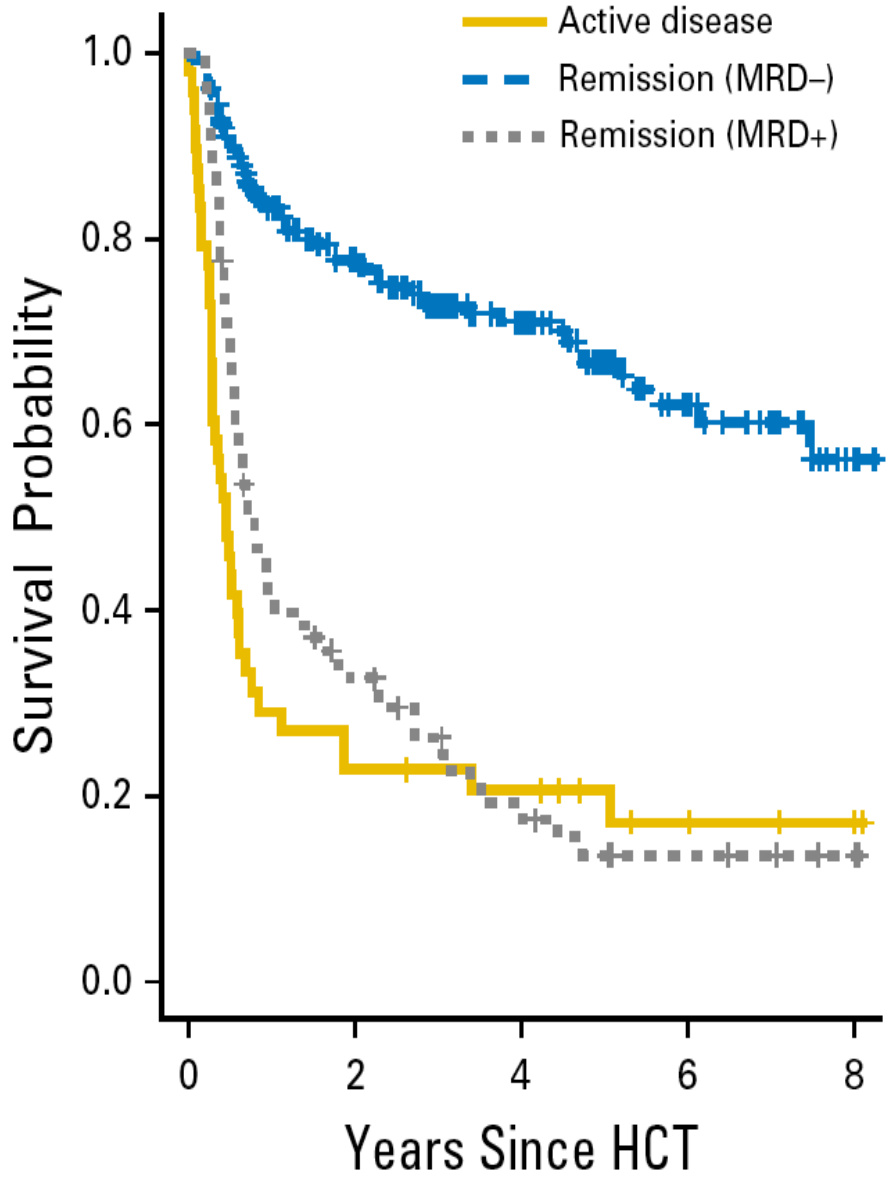


286 AML in CR
62% MAC
38% RIC
C=standard karyotype and AML FISH probe
F=10 color MFC

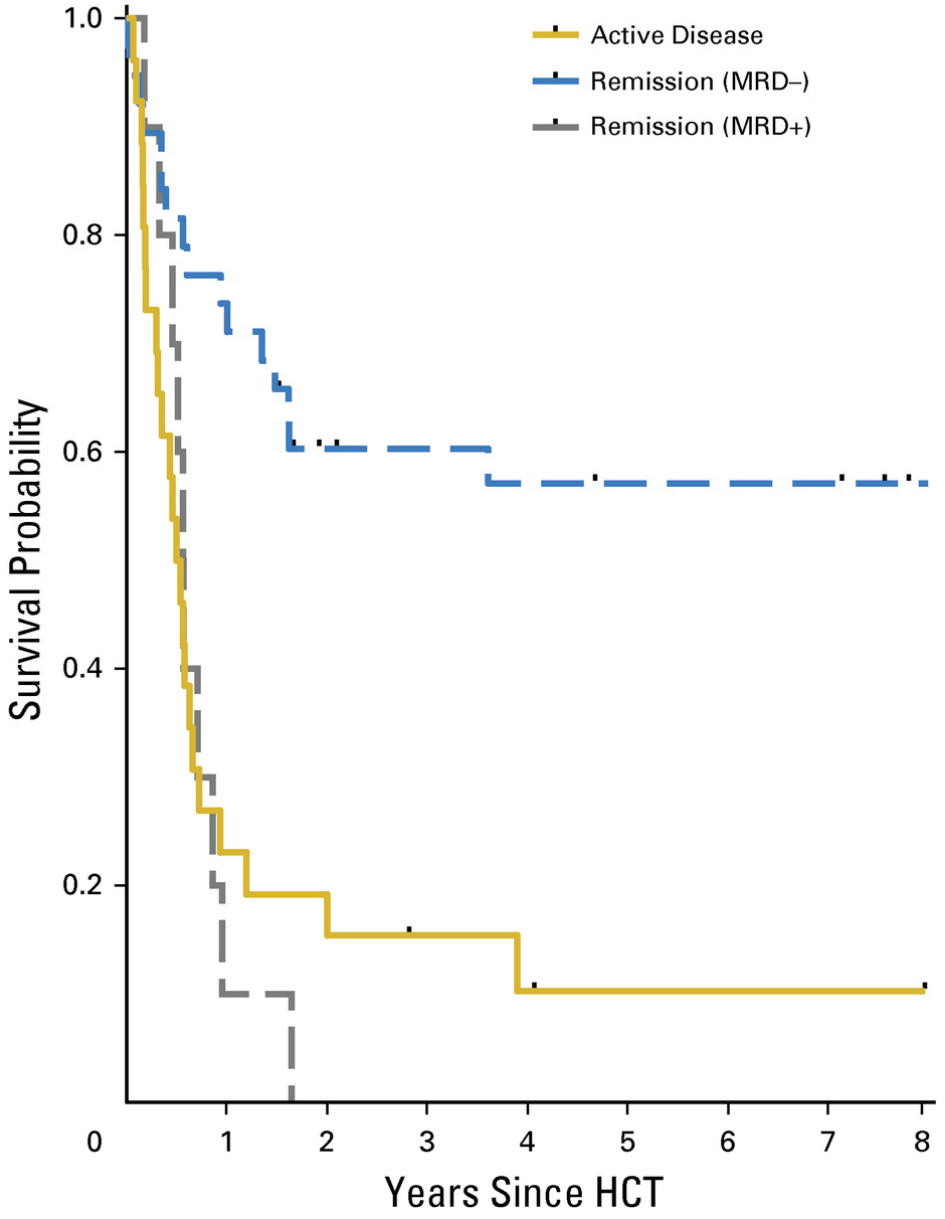
Fang et al. *Cancer* 2012;118:2411-1419



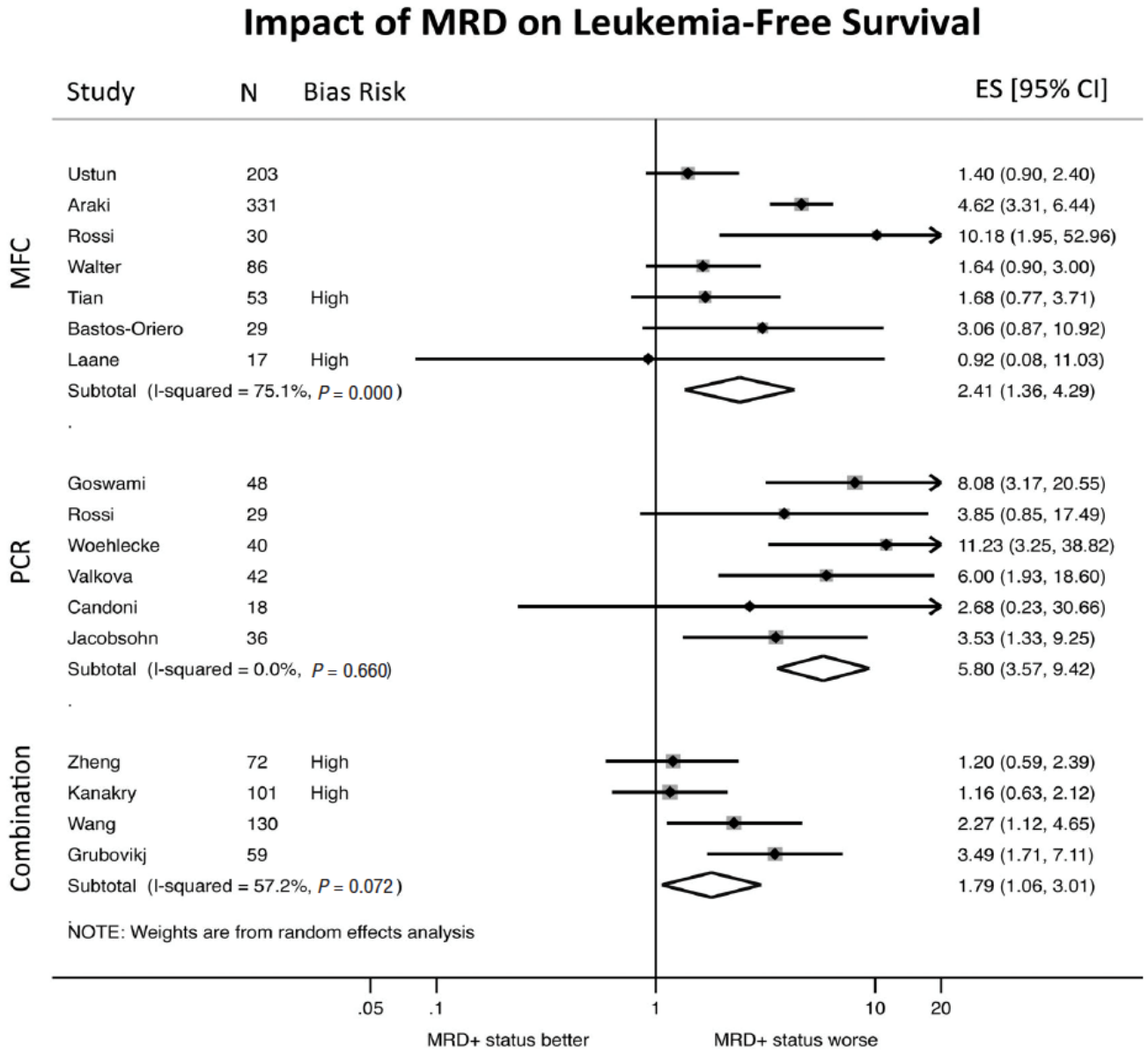
Araki *et. al.*, JCO 2016



Araki *et. al.*, JCO 2016



Hourigan *et. al.*, JCO 2016



Regardless of test used:

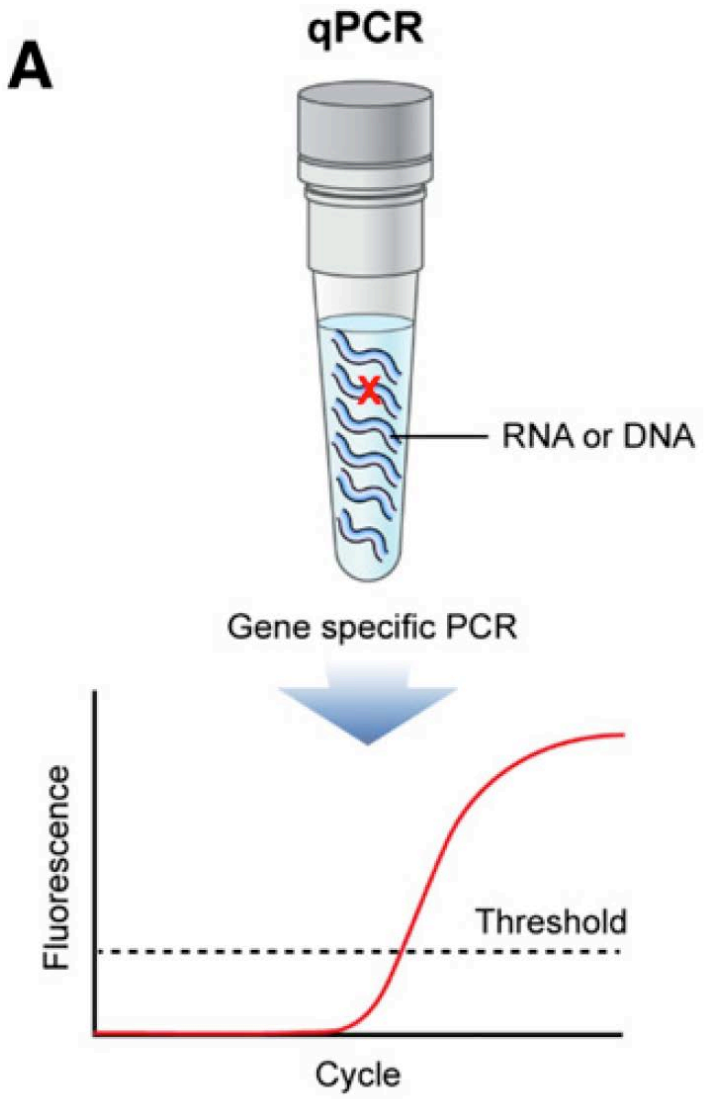
AML MRD in CR *before* Allo-HCT

=

worse survival *after* transplant.

Buckley *et. al.*, Haematologica, 2017

qPCR



Uses:

CBF (Inv16, t8,21)
NPM1mut (A, B and D)
BCR-ABL1

Advantages:

Ubiquitous presence in most clinical labs
Fast turnaround time
High sample throughput
Broad dynamic range.

Disadvantages:

Limited number of suitable targets/assays
Relative lack of multiplexing ability
Need to validate each target/assay individually
Limited ability to quantify at v.low MRD

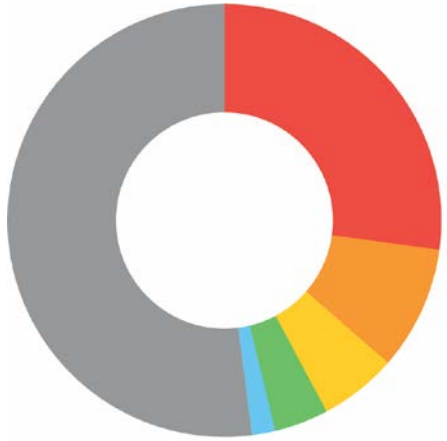
Hourigan and Freeman, ASH Educational, 2019

ELN Consensus Summary 2018 - MOLECULAR

- **Real-time qPCR** *…high sensitivity … therefore currently considered the gold standard….limited to … ~40% of AML patients*

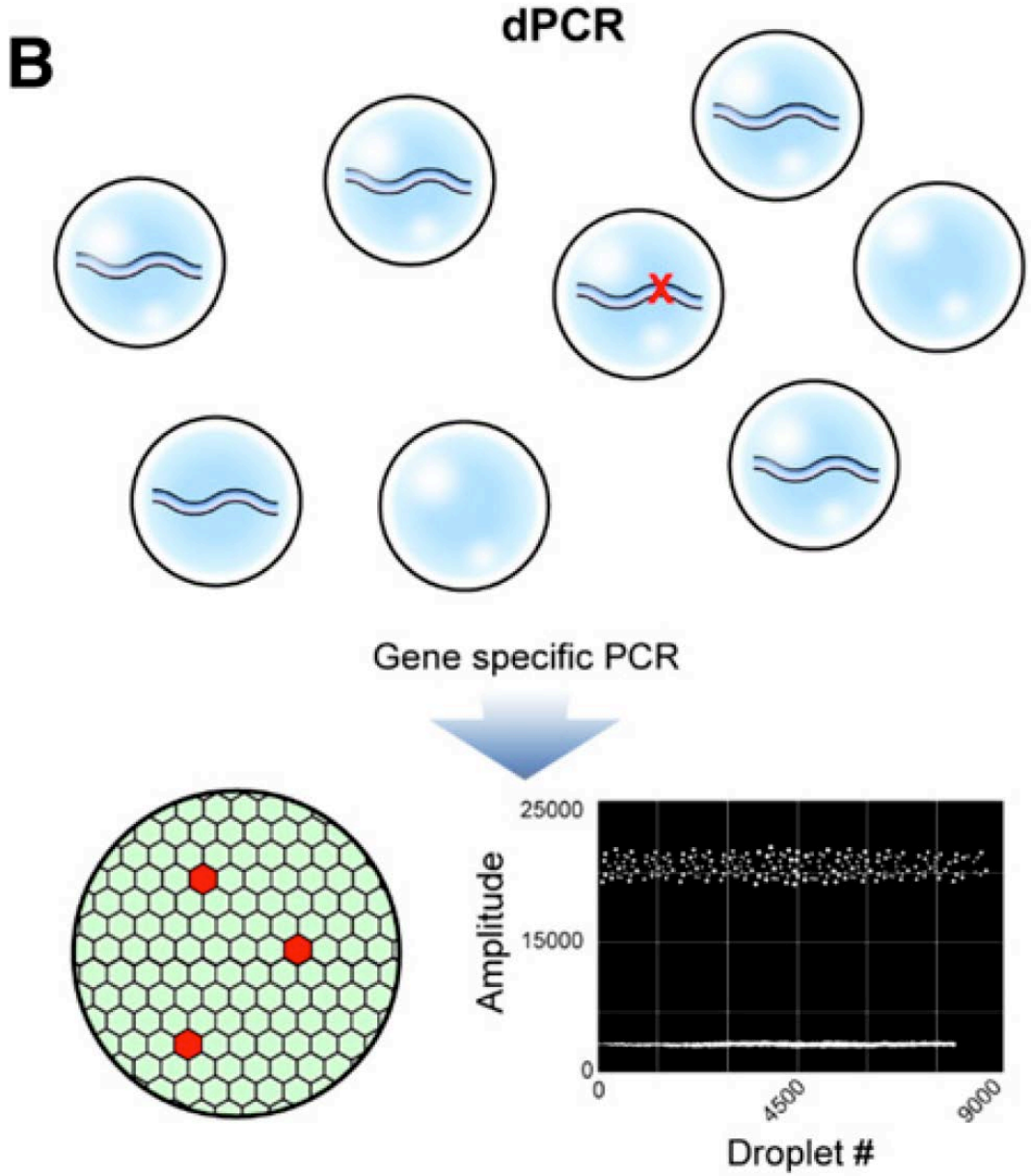
- 100ng cDNA/rxt (10K ABL1 copy)
- Run Triplicates
- EAC assays/criteria
- Ref. standards, pos. and no template control

Target	Classification	Target	Classification
NPM1	Insertion mutation	*NPM1*	Insertion mutation
PML-RARA	Fusion transcript	*PML-RARA*	Fusion transcript
CBFB-MYH11	Fusion transcript	*CBFB-MYH11*	Fusion transcript
RUNX1-RUNX1T1	Fusion transcript	*RUNX1-RUNX1T1*	Fusion transcript
BCR-ABL1	Fusion transcript	*BCR-ABL1*	Fusion transcript



- **Bone Marrow** (5-10ml, first pull, EDTA or Heparin okay) **AND Blood**

- **Complete molecular remission:** Must be in morphological CR. Two successive MRD negative samples obtained within interval of ≥ 4 weeks at a sensitivity level of at least 1 in 1000.
- **Molecular Relapse:** ↑MRD level of 1 $\log^{10}$ between 2 positive samples (4wk) in a patient who previously tested negative.
- **Molecular Persistence:** *<100-200 copies/10^4 ABL copies corresponding to <1% to 2% of target to reference gene or allele burden.* ***Progression:*** *↑MRD level of 1 $\log^{10}$ any 2 positive samples.*



Digital PCR

Uses:

As qPCR:

CBF (Inv16, t8,21)

NPM1mut (A, B and D)

BCR-ABL1

Advantages:

Absolute quantification – good for low MRD

Doesn't need standard curve

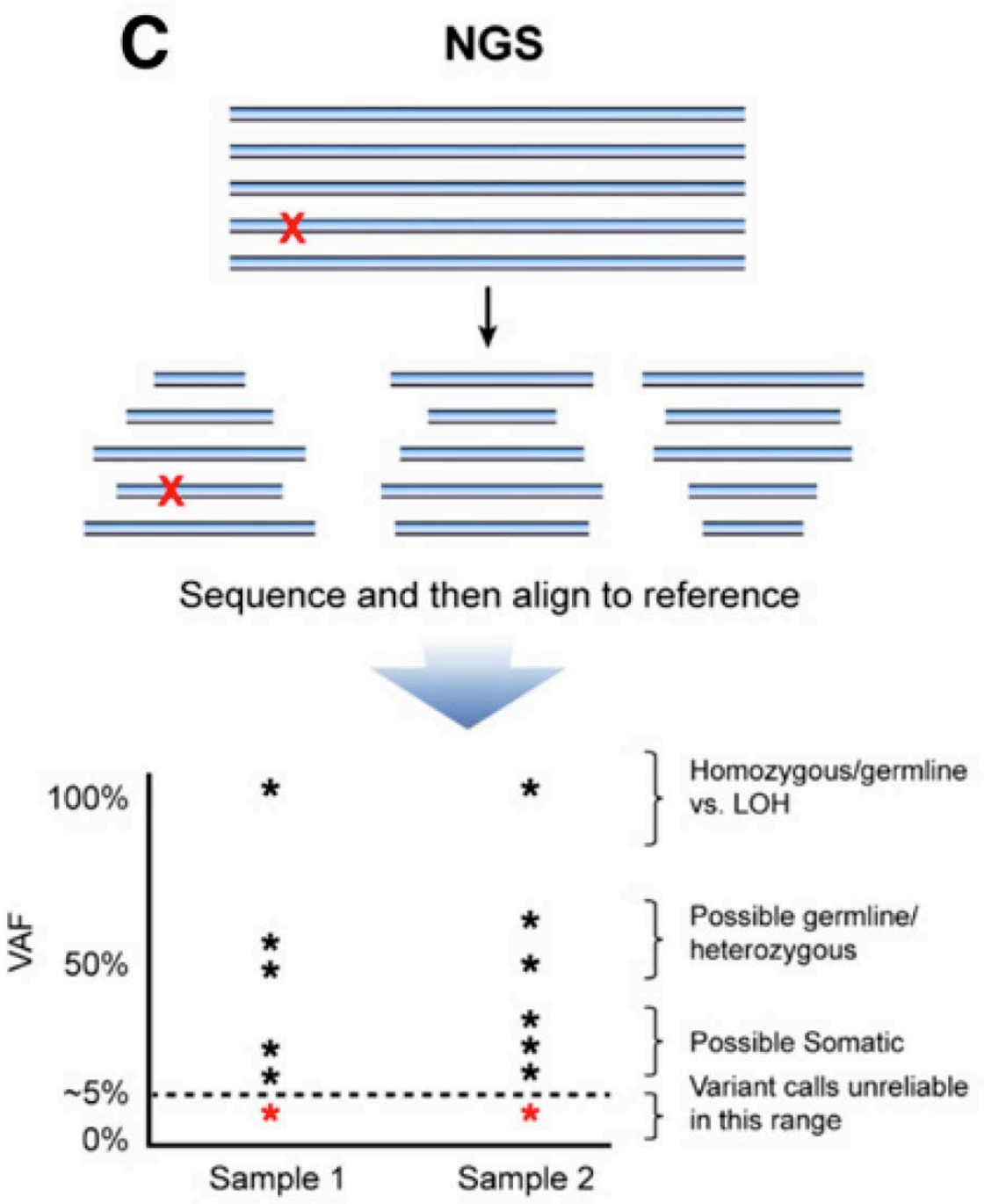
Disadvantages:

Technology not in common clinical use

Assays not clinically validated (unlike qPCR)

Cost >qPCR

Hourigan and Freeman, ASH Educational, 2019



NGS – diagnostic

aka: "myeloid panel"

Uses:

Genetic profiling of AML when blasts >5%

Not for measurable residual disease

Typical gDNA input 20-200ng

Advantages:

Broad panel = lots of targets

Disadvantages:

Very high **false positive** rate for variants <5%

Very high **false negative** rate for variants <5%

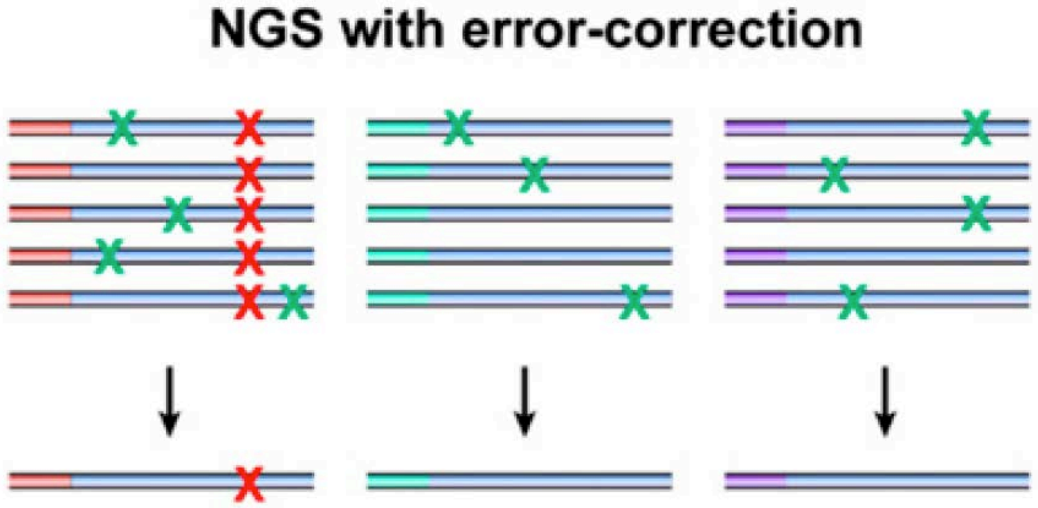
Hourigan and Freeman, ASH Educational, 2019

Ivey *et al.*
NEJM. 2016

Type		No. of Patients	Percent
A	GCTATTCAAGATCTCTG TCTG GCAGTGGAGGAAGTCTCTTTAAGAAAATAG	257	74
B	GCTATTCAAGATCTCTG CATG GCAGTGGAGGAAGTCTCTTTAAGAAAATAG	22	6
D	GCTATTCAAGATCTCTG CCTG GCAGTGGAGGAAGTCTCTTTAAGAAAATAG	31	9
	GCTATTCAAGATCTCTG CTTG GCAGTGGAGGAAGTCTCTTTAAGAAAATAG	5	
	GCTATTCAAGATCTCTG TATG GCAGTGGAGGAAGTCTCTTTAAGAAAATAG	5	
	GCTATTCAAGATCTCTG TCGG GCAGTGGAGGAAGTCTCTTTAAGAAAATAG	3	
	GCTATTCAAGATCTCTG CAGG GCAGTGGAGGAAGTCTCTTTAAGAAAATAG	2	
	GCTATTCAAGATCTCTG TAAG GCAGTGGAGGAAGTCTCTTTAAGAAAATAG	2	
	GCTATTCAAGATCTCTG CGTG GCAGTGGAGGAAGTCTCTTTAAGAAAATAG	1	
	GCTATTCAAGATCTCTG TTTG GCAGTGGAGGAAGTCTCTTTAAGAAAATAG	1	
	GCTATTCAAGATCTCTG CAAA GCAGTGGAGGAAGTCTCTTTAAGAAAATAG	1	
	GCTATTCAAGATCTCTG TAGG GCAGTGGAGGAAGTCTCTTTAAGAAAATAG	1	
	GCTATTCAAGATCTCTG CTCG GCAGTGGAGGAAGTCTCTTTAAGAAAATAG	1	
	GCTATTCAAGATCTCTG CAGA GCAGTGGAGGAAGTCTCTTTAAGAAAATAG	1	
	GCTATTCAAGATCTCTG CCGG GCAGTGGAGGAAGTCTCTTTAAGAAAATAG	1	10
	GCTATTCAAGATCTCTG TCAG GCAGTGGAGGAAGTCTCTTTAAGAAAATAG	1	
	GCTATTCAAGATCTCTG TTCG GCAGTGGAGGAAGTCTCTTTAAGAAAATAG	1	
	GCTATTCAAGATCTCTG CCGTT CAGTGGAGGAAGTCTCTTTAAGAAAATAG	1	
	GCTATTCAAGATCTCTG TCAAGACTTTCTTA AAGTCTCTTTAAGAAAATAG	1	
	GCTATTCAAGATCTCTG TCGGAGTCTCGGCGGAC TCTCTTTAAGAAAATAG	1	
	GCTATTCAAGATCTC ACAA TGGCAGTGGAGGAAGTCTCTTTAAGAAAATAG	1	
	GCTATTCAAGATCTC TCCATGCTCC TGGAGGAAGTCTCTTTAAGAAAATAG	1	
	GCTATTCAAGATCTCTGGCAG CGGA TGGAGGAAGTCTCTTTAAGAAAATAG	1	
	GCTATTCAAGATCTCTGGCAG AGGC TGGAGGAAGTCTCTTTAAGAAAATAG	1	
	GCTATTCAAGATCTCTGGCAGT CTTTCGCTCAC GTCTCTTTAAGAAAATAG	1	
	GCTATTCAAGATCTCTGGCAGTG TTTTGCTC AAGTCTCTTTAAGAAAATAG	1	
	GCTATTCAAGATCTCTGGCAGTG TTTTTCCC AAGTCTCTTTAAGAAAATAG	1	
		346	Total No. of Patients

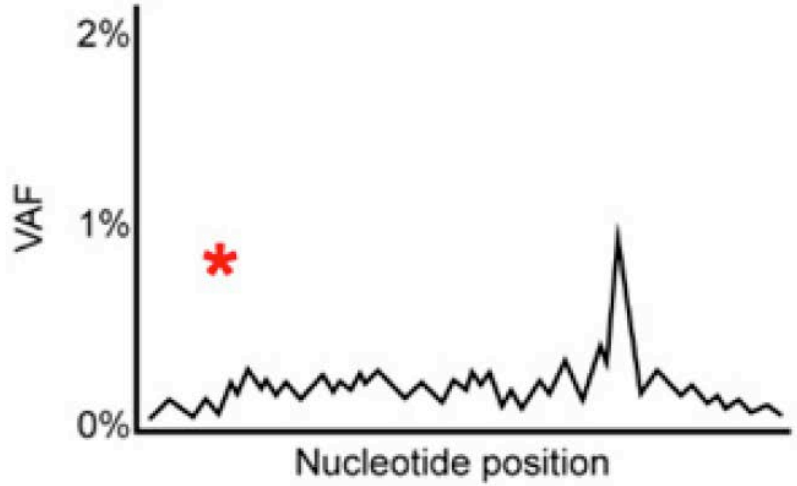
NGS – MRD Depth

externally validated test not yet available clinically



UMI based consensus clustering

And/Or



Background error model

Uses:

Research *(clinical soon hopefully)*

200ng to 2ug gDNA input

Advantages:

Broad panels – can track lots of variants

Detection down to to 0.001 or below

Potential for patient personalization

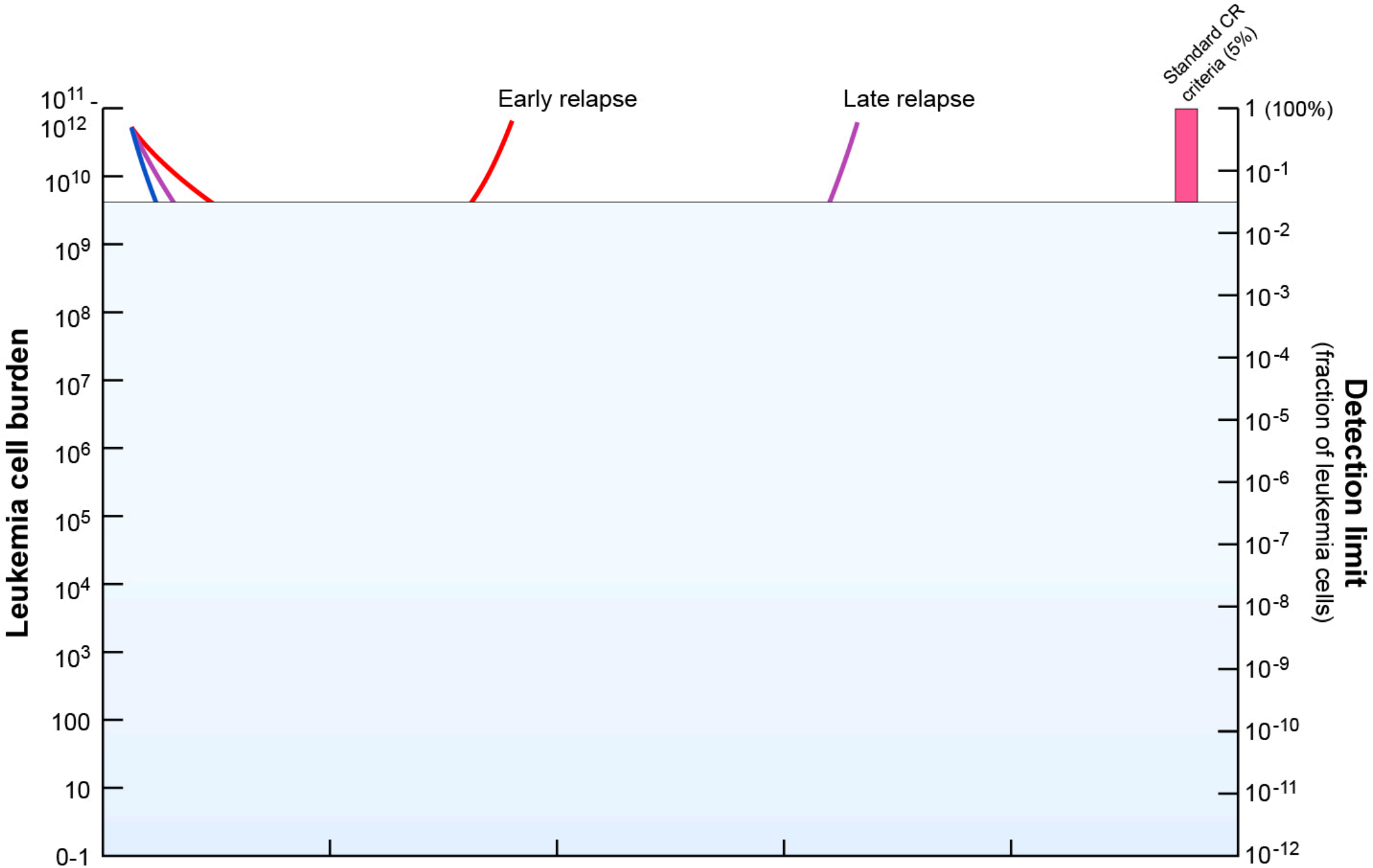
Disadvantages:

Cost

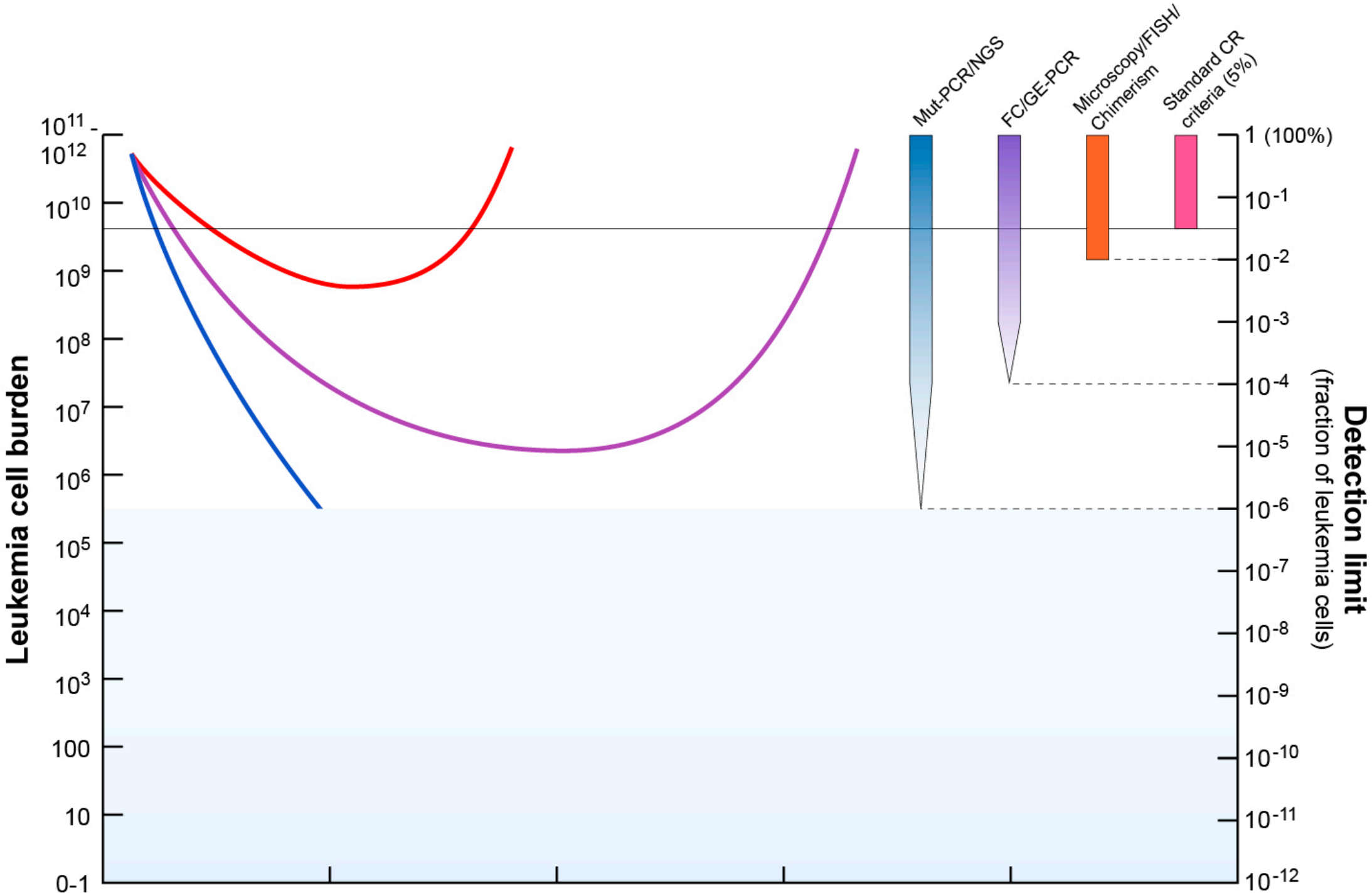
Clinical utility of detected variants unknown

Clinical utility of VAF thresholds unknown

Hourigan and Freeman, ASH Educational, 2019



Hourigan and Karp, Nature Reviews Clinical Oncology, 2013

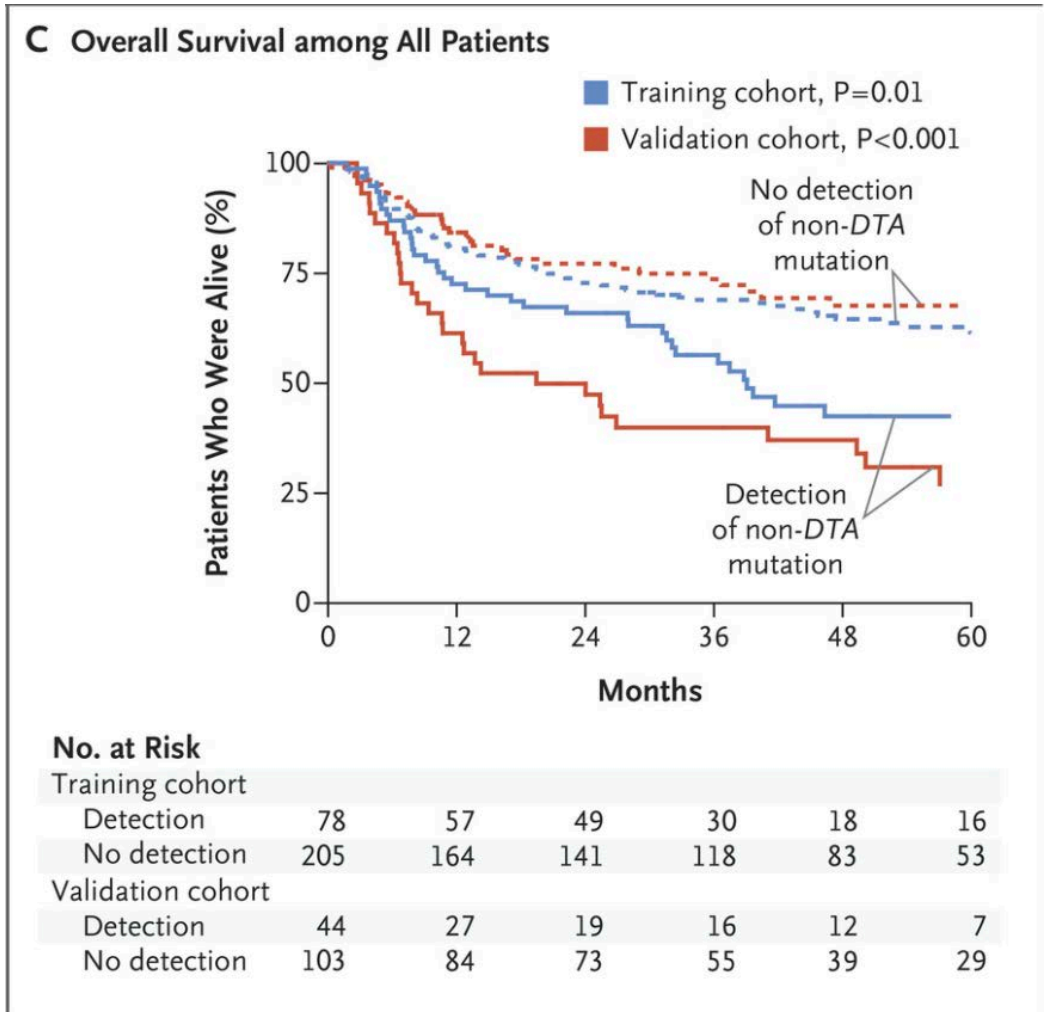


Hourigan and Karp, Nature Reviews Clinical Oncology, 2013

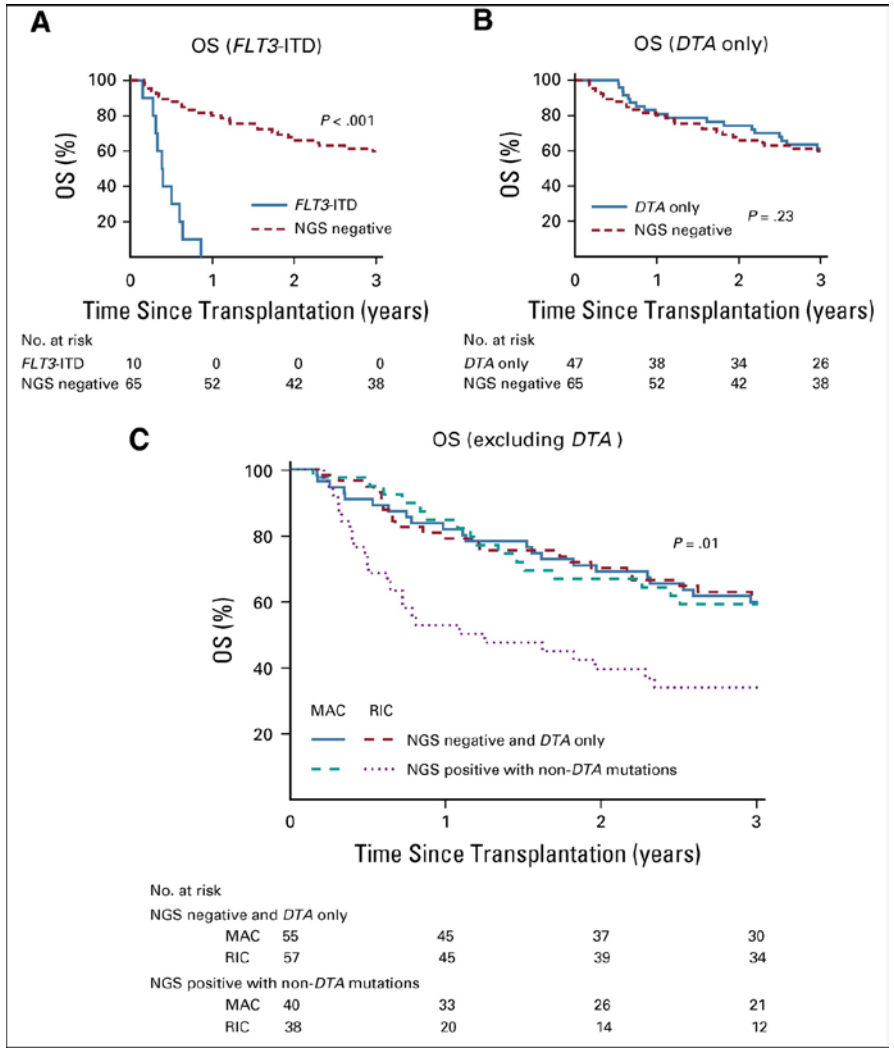
Take-home messages on molecular MRD

- ELN recommendation is currently only for qPCR (CBF, NPM1, BCR-ABL)
- Cytos and FISH low sensitivity (not MRD) but may be helpful if positive in cytomorphological remission
- ~80% of flow+ cases post induction will be deep NGS+. Also many flow- cases.
- Diagnostic NGS “myeloid panels” <u>insufficient</u> to test for MRD negativity
- “Late” mutations (FLT3, RAS, KIT) often lost at relapse = helpful if positive
- “early” mutations (DTA) often persist in cured patients = ?helpful if negative

Not all mutations are cancer – example: “DTA”



Mojca Jongen-Lavrencic, NEJM, 2018



Hourigan, JCO, 2020

Questions?

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Does MRD always matter?

Yes for AML and ALL CR1 MRD + or –
CR2 MRD + or –
CR3+ OK to ask but we do not know if matters for CR3+

MRD+ is as risky as morphologic disease pre transplant

Centers without high sensitivity MRD testing (and thus MRD unknown) are including patients with higher risk of relapse.

Recomendations: **Revise the questions** to ask the following:

For ALL, AML and MDS (consider the same questions for CLL, myeloma)

Pre-transplant

1. In Morphologic CR, was MRD assessed? y/n.

2. If Flow was tested
   Was an original leukemia immunophenotype used for detection? y/n
   Was an aberrant phenotype used for detection? y/n
   What is the lower limit of detection?

3. Was molecular assay (PCR or NGS) used for MRD detection? y/n
   Was MRD detected? y/n

4. Were cytogenetic assays (Metaphase or FISH) used for MRD detection? y/n
   Was MRD detected? y/n

Recommendations

For the Outcomes Analysis of 1 year survival.

Include these changes only for ALL, AML

Use modified pre-transplant disease status definitions:

CR1 (or CR2 or later CR) without MRD

CR with MRD+

and

CR with no high sensitivity testing for MRD

How complete is molecular data is (AML as example)?

- Selection: first alloHCT for AML since F2402R2 (July 2017, when time point of between dx and HCT are added)
- Select molecular/cytogenetic abnormalities (7- by FISH, CEBPA, FLT3-TKD, FLT3-ITD, NPM1)
- Data complete across all 3 time points in only 10%
- Data complete across two time points (dx and at HCT):
  - CEBPA 12%
  - FLT-TKD 17%
  - FLT-ITD 23%
  - NPM1 19%
  - 7 by FISH 12%