

# TwinStrand DuplexSeq™ AML MRD Solution



Next Generation Sequencing is a powerful emerging method for detecting residual acute myeloid leukemia (AML) after treatment. However, current methods produce an abundance of sequencing errors which obscure low-frequency leukemia-defining mutations.

The TwinStrand Duplex Sequencing™ AML MRD assay is orders of magnitude more sensitive and specific than other NGS based MRD assays, offering unprecedented opportunities for clinical research and drug development.

## Duplex Sequencing AML MRD

>100-fold higher resolution than other error-corrected NGS methods, revealing more clinically significant mutations

The 29 gene, 58kb panel encompasses loci mutated in >92% of adult AML patients

Assay enables sub-1/100,000 limit of detection anywhere in target region

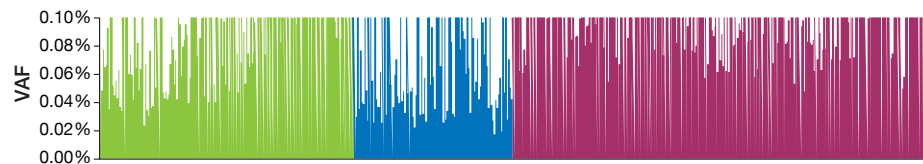
## Duplex Sequencing MRD Technical Performance

Cell line DNA mixture models patient with MRD

Gene	Mutation	Predicted VAF
TP53	Q136fs	1x10 <sup>-2</sup>
TP53	R249S	2x10 <sup>-3</sup>
TP53	R248Q	1x10 <sup>-3</sup>
NRAS	Q61R	2x10 <sup>-4</sup>
TP53	G245S	1x10 <sup>-4</sup>
NRAS	Q61L	4x10 <sup>-5</sup>
TP53	R175H	2x10 <sup>-5</sup>
KRAS	G12D	1x10 <sup>-5</sup>
KRAS	G12V	1x10 <sup>-5</sup>

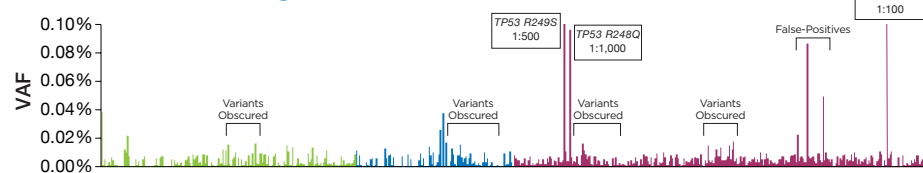
Sequence to >1M Duplex molecular depth

### No Error-Correction



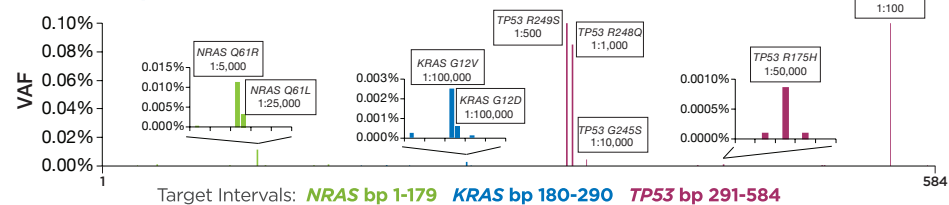
Background at all positions, all variants obscured

### Standard Single-Strand UMI Error-Correction



Many false positives from 10<sup>-3</sup> – 10<sup>-4</sup>

### Duplex Error-Correction



All mutations detected to below 10<sup>-4</sup>

The TwinStrand Duplex Sequencing AML MRD assay was used to sequence a serially-diluted mixture of AML mutation-containing DNA to a Duplex depth >1,000,000x. Target clones were identified to levels <1/100,000 with 100% sensitivity and specificity.

# AML MRD Panel Overview

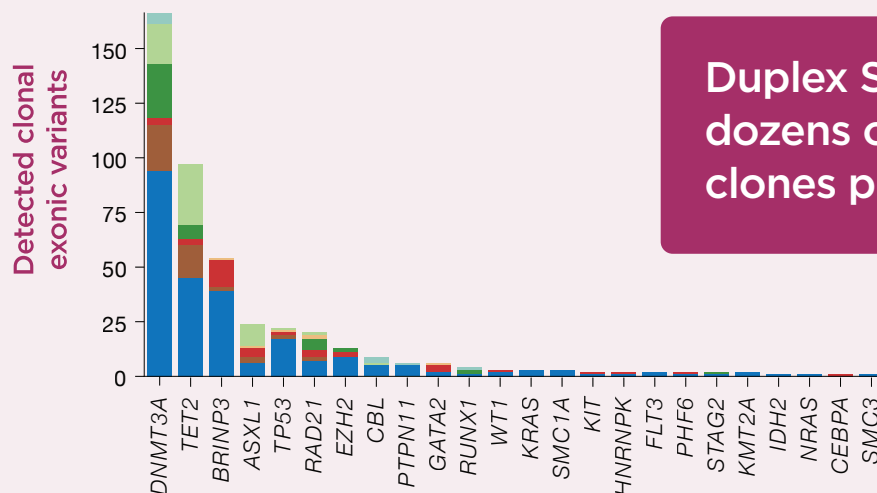
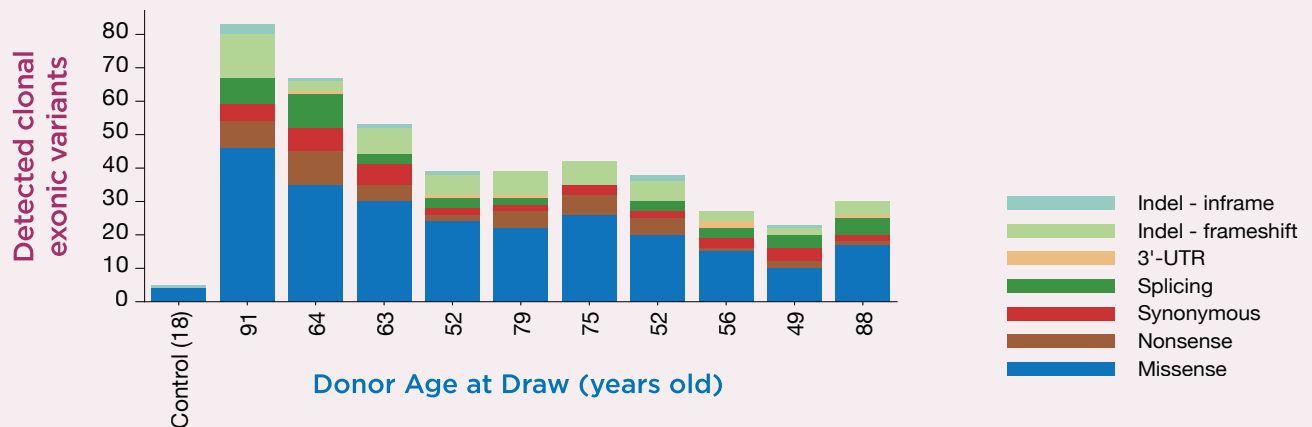


## Panel Overview

The AML panel encompasses the whole coding region or hotspots in 29 genes recurrently mutated in AML, MDS (myelodysplastic syndrome), and CHIP (clonal hematopoiesis).

Genes in Panel					
ASXL1	FAM5C (BRINP3)	IDH2	NPM1	RAD21	TET2
CBL	FLT3	KIT	NRAS	RUNX1	TP53
CEBPA	GATA2	KRAS	PHF6	SMC1A	U2AF1
DNMT3A	HNRNPK	MLL (KMT2A)	PTEN	SMC3	WT1
EZH2	IDH1	MYH11-CBFB	PTPN11	STAG2	

## Clonal Hematopoiesis



Duplex Sequencing detects dozens of simultaneous CHIP clones per individual with aging.