Sample Information

Sample ID	Analysis Date	DNA Input*	Test	Flowcell ID	Analysis ID	Lot Number
MAF02	2021-09-30	7154 GE	BC_RUO1	H3KT7AFX3	210930 LAB	D21001

*Genome Equivalents: number of amplifiable haploid genomic copies analyzed by the assay. 1 GE = 3.3 pg of DNA.
The DNA input is listed as not quantifiable if too much / not enough input DNA is present or due to failure of Positive Control.

Sample Validity

Run Validity				Sample Validity			
Positive control:	✓	No template control:	✓	Sequencing depth:	✓	Quantification:	✓

For detailed information about validity parameters please refer to the QC part of this report.
Mutation calls will only be reported as valid, if all controls and sample performance criteria are met.
Otherwise, the mutation calls will be marked as invalid.

Sample Mutation Status

AKT1	ERBB2	ESR1	KRAS	PIK3CA	TP53
3	2	0	1	2	18

Somatic Mutations Detected

Gene ID	Transcript	Coding DNA Change	Amino Acid Change	COSMIC ID	MAF	Mutant Molecules	Validity
AKT1	ENST00000554581	c.47-25G>T	p.?		0.112%	8	✓
AKT1	ENST00000554581	c.49G>A	p.E17K	COSV62571334	0.234%	17	✓
AKT1	ENST00000554581	c.69G>T	p.R23=		0.103%	7	✓
ERBB2	ENST00000269571	c.2313_2324dup	p.Y772_A775dup	COSV54062409	0.124%	9	✓
ERBB2	ENST00000269571	c.2360G>A	p.G787D		0.136%	10	✓
KRAS	ENST00000256078	c.35G>A	p.G12D	COSV55497369	0.356%	25	✓
PIK3CA	ENST00000263967	c.1633G>A	p.E545K	COSV55873239	0.122%	9	✓
PIK3CA	ENST00000263967	c.3140A>G	p.H1047R	COSV55873195	0.265%	19	✓
TP53	ENST00000269305	c.1002G>A	p.G334=		0.095%	7	✓
TP53	ENST00000269305	c.1017G>T	p.E339D		0.104%	7	✓
TP53	ENST00000269305	c.328C>T	p.R110C	COSV52684761	0.089%	6	✓
TP53	ENST00000269305	c.336C>A	p.G112=		0.089%	6	✓
TP53	ENST00000269305	c.372C>T	p.C124=		0.094%	7	✓
TP53	ENST00000269305	c.469G>A	p.V157I	COSV52676009	0.097%	7	✓
TP53	ENST00000269305	c.482C>T	p.A161V	COSV52678014	0.106%	8	✓
TP53	ENST00000269305	c.524G>A	p.R175H	COSV52661038	0.276%	20	✓
TP53	ENST00000269305	c.723del	p.C242Afs*5	COSV52760886	0.369%	26	✓
TP53	ENST00000269305	c.734G>A	p.G245D	COSV52667838	0.262%	19	✓
TP53	ENST00000269305	c.743G>A	p.R248Q	COSV52661580	0.369%	26	✓
TP53	ENST00000269305	c.818G>A	p.R273H	COSV52660980	0.386%	28	✓
TP53	ENST00000269305	c.827C>T	p.A276V	COSV52704969	0.274%	20	✓
TP53	ENST00000269305	c.848G>A	p.R283H	COSV52700134	0.089%	6	✓
TP53	ENST00000269305	c.851C>A	p.T284K		0.089%	6	✓
TP53	ENST00000269305	c.925C>A	p.P309T		0.085%	6	✓
TP53	ENST00000269305	c.925C>T	p.P309S	COSV52729434	0.094%	7	✓
TP53	ENST00000269305	c.984C>A	p.F328L		0.104%	7	✓

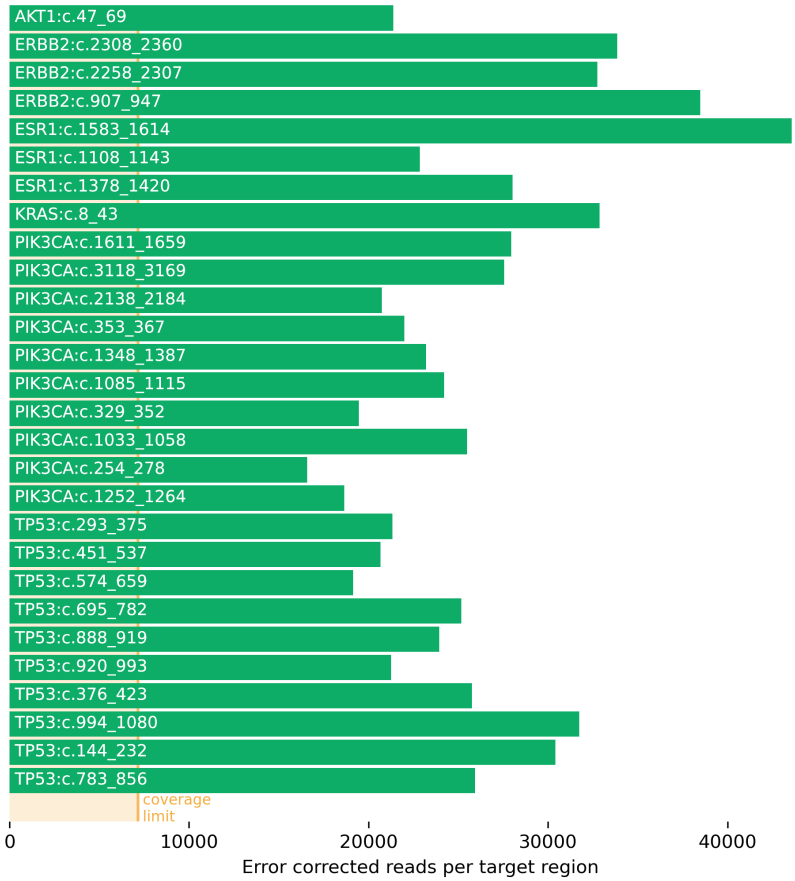
- No germline mutation calls detected for this sample.
  For a complete listing of mutation calls (including invalidated calls), please refer to the VCF output.

Sequencing Run Quality

Category	Acceptance Range	Value
Sequencer ID	- / -	NDX550262_RUO
Sequencing Kit	- / -	
Reads Passing Filter	≥ 80%	
Cluster Density [Clusters / mm^2]	NextSeq™ 500/550: ≤ 220k MiSeq™: ≤ 1400k	
Percentage >Q30	≥ 80%	

Sample Validity

Sample Coverage



Sequencing depth per target region: green bars indicate sufficient coverage while orange bars indicate insufficient target coverage.

Positive Control

	Limits	Result	
Depth of coverage	1.0	>1	✓
Quantification	500.0 GE<x GE<1500.0 GE	1410 GE	✓
Detection	10.0	≥10	✓

If the quantification of the positive control fails, mutant molecule values (MM) will not be reported per sample.

No Template Control

NTC ID	Avg. UID threshold	Avg. UID result	
NTC	15	0.0	✓

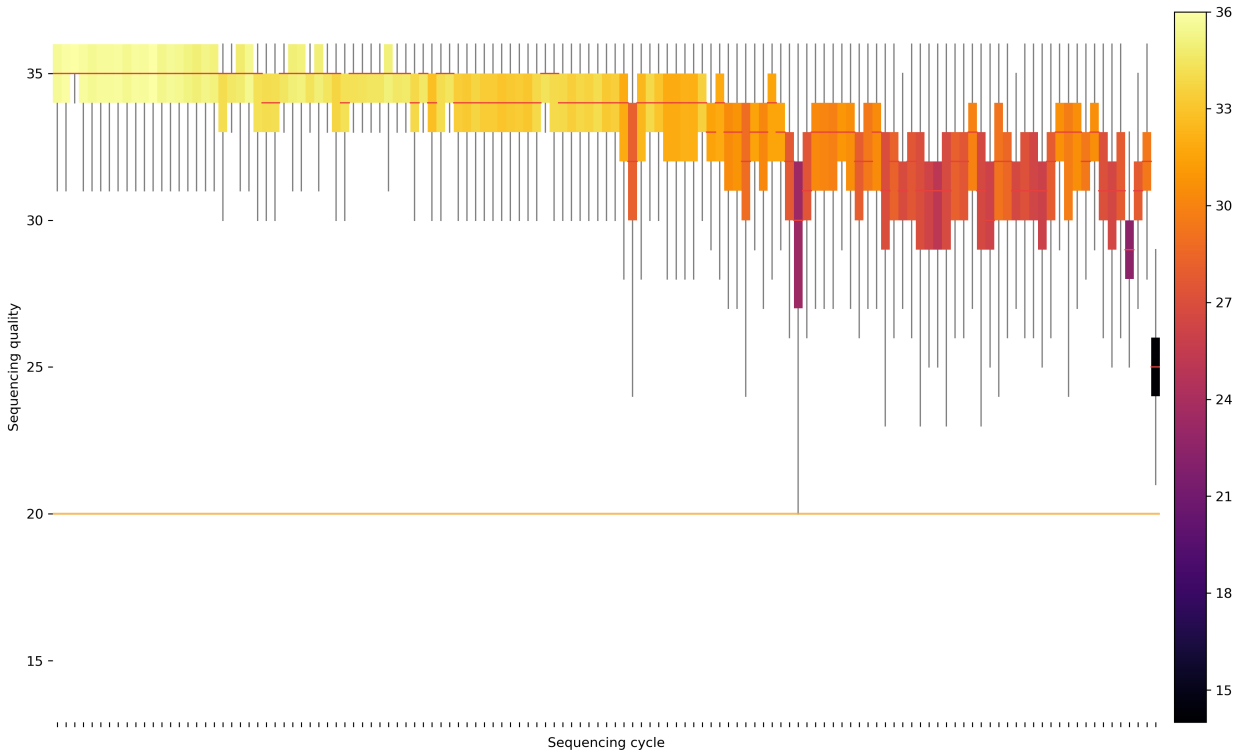
Sample Metrics

Read Overview

Category	Result
Reads imported from sequencer	14,896,267
Reads passing quality filter	14,769,485
Usable read rate	99.15%
Reads with sufficient UID size	14,311,470
Read rate with sufficient UID size	96.9%
Raw error corrected reads	818,829
Filtered error corrected reads	818,289
Filtered error corrected read rate	99.93%

FASTQ reads are initially filtered by the sequencing device (Illumnina NextSeq™ 500\550 or Illumina MiSEQ™). For the Plasma-SeqSensei™ assay, these reads are further inspected for low quality nucleotide calls in general and for the bar-code sequence (UID / UMI) in particular.
From this, the usable read rate is determined. Reads are grouped according to their bar-code sequence and bar-coded groups with at least 4 members are further processed (reads with sufficient UID size).
Error corrected reads are generated from these groups with a certain conversion rate.

Sequencing Quality



Distribution of QPhred quality values in error corrected reads. Shaded area indicates QPhred quality values excluded from mutation calling. A subset of 10,000 reads is used per FASTQ file of the sample to generate this plot.

Analysis Software and Database Information

Analyzed by Plasma-SeqSensei™ Software v1.1.0

Assay configuration BC_RUO1

Sequencing result annotation supported by:

- COSMIC Databse v92 (https://cancer.sanger.ac.uk/cosmic)
- The Ensembl database

The Plasma-SeqSensei™ Software is for research use only. Not for use in diagnostics procedures.

Date, Signature