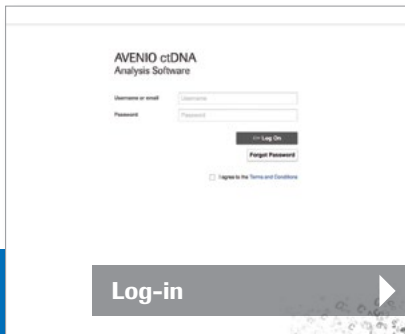


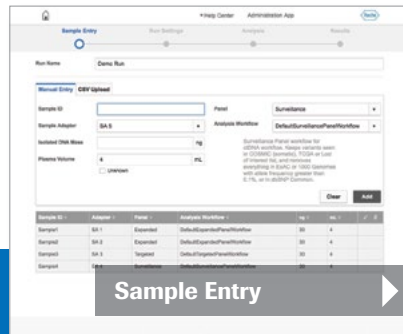
AVENIO ctDNA Analysis Software



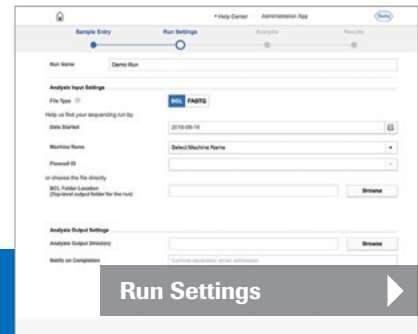
EXCEPTIONAL ACCURACY
MINIMAL EFFORT



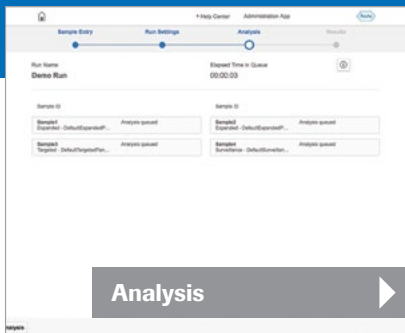
Log-in



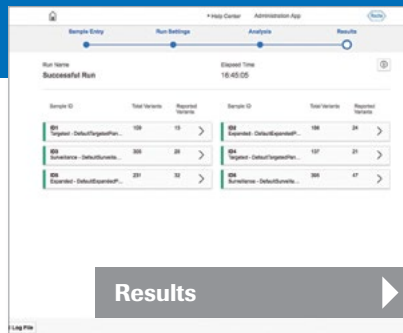
Sample Entry



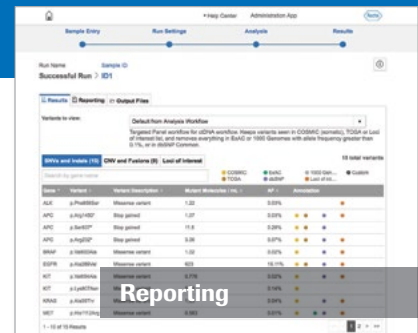
Run Settings



Analysis



Results



Reporting



Set up and run the analysis with ease

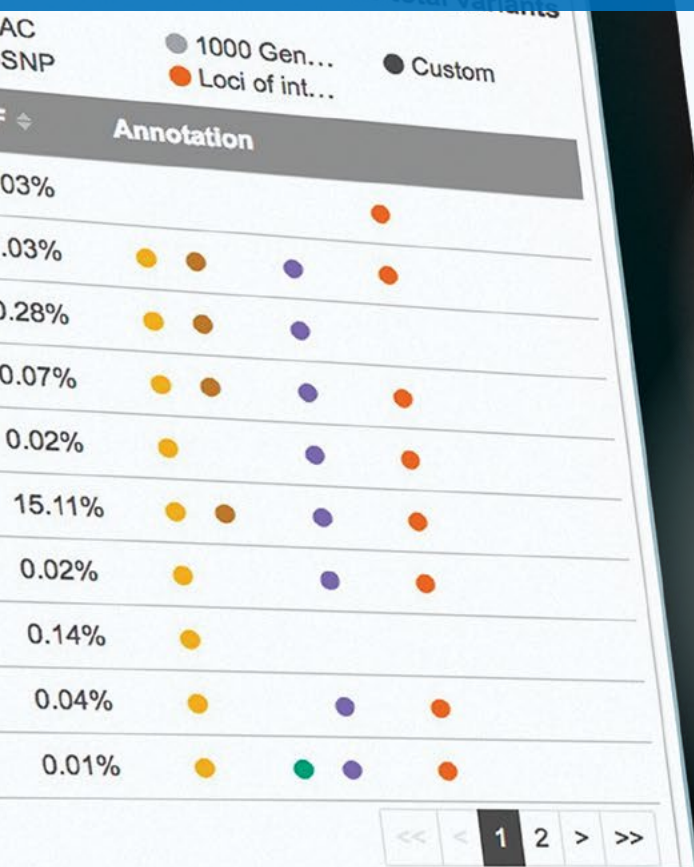
The AVENIO ctDNA Analysis Software requires minimal expertise to set up and run an analysis on ctDNA samples. This allows laboratory and bioinformatics professionals to focus more of their time on the findings and insights, and less on managing information systems.

Intuitive user interface:

- Offers two user settings (Admin, Lab User)
- Guides step-by-step workflow
- Provides real-time tracking of analysis
- Enables filtering and custom annotation
- Provides reports in PDF format, as well as variant results in VCF and BAM file formats.

With its ability to process approximately 800 million reads in less than a day, the AVENIO ctDNA Analysis Software can be set up to run in the evening, delivering results ready for review in the morning. The software can analyze multiple samples at once, saving time, effort and resources.

AVENIO ctDNA Analysis Software



The AVENIO ctDNA Analysis Software processes sequencing data and applies a powerful bioinformatic pipeline to profile tumors. The software generates clear information and reports from samples run using the AVENIO ctDNA Analysis Kits.

It fosters insights derived from oncology sequencing data while minimizing complexity and effort. Its interface guides the user confidently through the post-sequencing workflow. Intelligent bioinformatics with advanced proprietary algorithms and error suppression strategies deliver comprehensive results with proven accuracy and reproducibility.^{1,2}

Powerful, yet accessible and intuitive, the AVENIO ctDNA Analysis Software offers simplicity, efficiency and accuracy.

**For Research Use Only.
Not for use in diagnostic procedures.**



SNV
Single
Nucleotide
Variant



Indel
Insertion
or Deletion



Fusion



CNV
Copy Number
Variant

AVENIO ctDNA Analysis Software Variant Report

SAMPLE ID		ANALYSIS START DATE	
ID1		Jan 13, 2017 17:23:37 PST	
ANALYSIS WORKFLOW	DefaultTargetedPanelWorkflow	PAIRED	
ADAPTER		Targeted	
SA 1		FLUORESET	
ISOLATED DNA NAME	DefaultHeader		
REFSeq		SAMPLE VOLUME	4.0 mL

Results Summary

Gene	Variant	Variant Description	Allele Fraction	No. of Mutant Molecules per mL
NRAS	p.Arg54Gly	Missense variant	0.08%	3.27
NRAS	p.Arg59Thr	Missense variant	0.04%	1.53
TP53	p.Arg273Gln	Missense variant	0.35%	14.3
TP53	c.578-1G>A	Splice acceptor variant & intron variant	0.07%	2.86
TP53	c.543-1G>A	Splice acceptor variant & intron variant	0.34%	14.2
TP53	p.Pro228Leu	Missense variant		
TP53	p.Pro125Leu	Missense variant		
TP53	c.378-1G>A	Splice acceptor variant & intron variant	0.18%	6.47
TP53	p.Lys114Lys	Splice region variant & Synonymous variant	0.03%	1.22
ALK	p.Phe568Ser	Missense variant	0.02%	0.776
KIT	p.Val554Ala	Missense variant	0.14%	5.80
KIT	p.Lys927Asn	Missense variant	0.07%	2.96
APC	p.Arg32P	Stop gained	0.28%	11.5
APC	p.Ser93P	Stop gained	0.03%	1.07
EGFR	p.Asp299Val	Missense variant	<0.11%	693
MET	p.His1112Arg	Missense variant	0.01%	0.583
RRF1	p.Val508Asp	Missense variant	0.02%	1.02

Run Details

Run Name	NUMBER OF SAMPLES
Successful Run	1

Sample ID: ID1 Page 1 of 23

Detect all four mutation classes

The software's bioinformatic algorithms and checkpoints have been optimized with clinical samples to enable accurate variant calls across all four mutation classes.¹

Performance metrics

Mutant Allele Frequency	SNVs		Indels	
	0.5%*		1.0%	
AVENIO ctDNA Kit	Sensitivity	PPV	Sensitivity	PPV
Targeted	>99%	>99%	>99%	>99%
Expanded	>99%	>98%	>99%	>99%
Surveillance	>99%	>99%	>99%	>99%

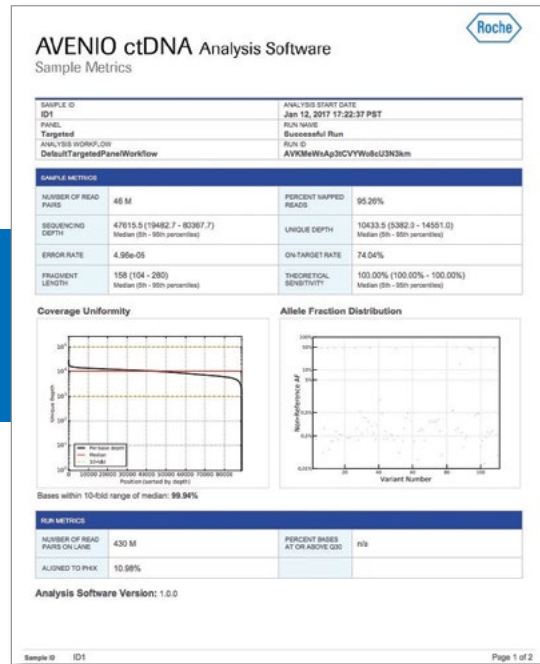
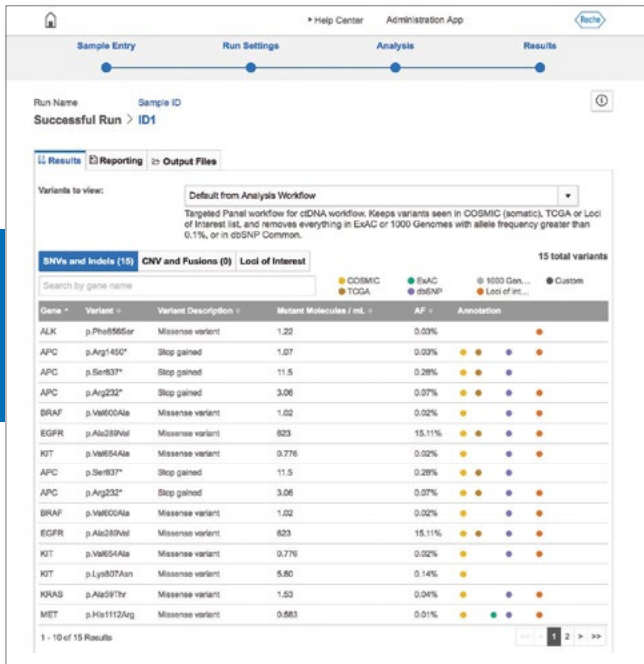
Mutant Allele Frequency/ Copy Number	Fusions		CNVs	
	1.0%		2.3 copies**	
AVENIO ctDNA Kit	Sensitivity	PPV	Sensitivity	PPV
Targeted	>96%	>99%	>99%	>99%
Expanded	>99%	>99%	>99%	>99%
Surveillance	>99%	>99%	>99%	>96%

* Detects variants down to 0.1%. **10% ctDNA at 5 copies in tumor. Performance samples - cell line mixes, cfDNA 10ng-50ng input. Sensitivity and Positive Predictive Value (PPV) metrics based on typical product performance. Sensitivity and PPV performance is reported per variant. SNV performance data based on hotspot calls; CNV performance based on MET variant. Results above were tested at the stated mutant allele frequencies. AVENIO ctDNA Analysis Kits also achieve >99.99% per base specificity across each of the panels. Performance data based on 40 million reads per sample for Targeted Kit, and 60 million reads per sample for Expanded and Surveillance Kits. Sequencing performed on an Illumina NextSeq 500.

Detect low frequency alleles with exceptional sensitivity and specificity

The AVENIO ctDNA Analysis Software employs integrated digital error suppression (iDES) strategies, combining molecular bar codes with in silico error suppression techniques. This proprietary method enables detection of low frequency alleles down to 0.1% with exceptional sensitivity and specificity.^{1,2}

For more details about integrated digital error suppression (iDES), please refer to the article, "Integrated digital error suppression for improved detection of circulating tumor DNA" by Newman, Lovejoy, Klass, et al, published in Nature Biotechnology in May 2016.



Call mutations with confidence

Five leading oncology databases, a curated loci of interest list and a customizable annotation database are integrated into the AVENIO ctDNA Analysis Software. Use it for quick and easy access to trusted public resources to cross-reference and confirm results:

- COSMIC
- TCGA
- ExAC
- dbSNP
- 1000 Genomes

Contextualized for variants, these annotation databases can help users call mutations with increased confidence.

Control quality across the workflow

The AVENIO ctDNA Analysis Software provides a sequencing quality report with key metrics such as sequencing depth, number of reads, on-target rate, coverage uniformity, and error rate to confirm quality at different stages in the workflow.

The software also provides access to VCF and BAM files as needed to facilitate further downstream analysis.

1. Data on file

2. Newman AM, Lovejoy AF, Klass DM, et al. Integrated digital error suppression for improved detection of circulating tumor DNA. Nature Biotechnology. 2016;34(5):547-555. doi:10.1038/nbt.3520.

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