

# AmpliSeq™ for Illumina Comprehensive Cancer Panel

Fast and accurate investigation of > 400 genes with known associations to cancer.

## Highlights

- Relevant Gene Content**  
 Target all-exon coverage of 409 cancer-associated genes
- Fast, Streamlined Workflow**  
 Prepare sequencing-ready libraries in a single day from as little as 1 ng high-quality DNA, or 10 ng DNA from FFPE tissue
- Accurate Data**  
 Detect somatic mutations down to 5% frequency using local or cloud-based analysis

## Introduction

The AmpliSeq for Illumina Comprehensive Cancer Panel provides a targeted resequencing solution for analyzing somatic mutations across 409 genes with known associations to multiple cancer types, including lung, colon, breast, ovarian, melanoma, and prostate. (Table 1). The Comprehensive Cancer panel is part of a streamlined workflow that includes PCR-based library preparation, Illumina sequencing by synthesis (SBS) chemistry and next-generation sequencing (NGS) technology, and automated analysis. It requires as little as 1 ng high-quality DNA or 10 ng from DNA from low-quality samples per pool, making it compatible with various sample types, including formalin-fixed, paraffin-embedded (FFPE) tissues. The high gene content of the panel and low DNA input requirement empower a single, streamlined workflow for the quick assessment of cancer-related genetic variations, affording researchers the potential to unlock a wealth of genomic information from many tumor types.

## Relevant Gene Content

Content for the AmpliSeq for Illumina Comprehensive Cancer Panel was selected based on expert curation of the scientific literature and other high-quality databases and includes over 50% of the Wellcome Trust Sanger Institute Cancer Gene Census. It was designed to profile exonic regions of known cancer driver genes, drug targets, signaling cascade genes, apoptosis genes, DNA repair genes, transcription regulators, inflammatory response genes, and growth factor genes. This ready-to-use panel saves researchers the time and effort of identifying targets, designing amplicons, and optimizing performance.



Access a complete list of genes on the AmpliSeq for Illumina Comprehensive Cancer Panel at [www.illumina.com/products/by-type/sequencing-kits/library-prep-kits/ampliseq-comprehensive-cancer-panel.html](http://www.illumina.com/products/by-type/sequencing-kits/library-prep-kits/ampliseq-comprehensive-cancer-panel.html)

**Table 1: AmpliSeq for Illumina Comprehensive Cancer Panel At A Glance**

Parameter	Specification
No. of Genes	409
Targets	Full-exon coverage for oncogenes and tumor suppressor genes
Cumulative Target Size	1.7 Mb
Variant Types	SNVs, indels <sup>a</sup>
Amplicon Size	109 bp on average
No. of Amplicons	15,992
Input DNA Requirement	1–100 ng (10 ng recommended per pool)
No. of Pools per Panel	4
Supported Sample Types	FFPE tissue, blood
Percent Targets Covered at Minimum 500x at Recommended Throughput	> 95%
Coverage Uniformity (percent of targets with > 0.2x mean coverage)	> 90%
Percent On-Target Aligned Reads	> 90%
Total Assay Time	6 hours <sup>b</sup>
Hands-On Time	< 1.5 hours
DNA-to-Data Time	2.5 days

a. SNVs: single nucleotide variants; indels: insertions/deletions  
 b. Time represents library preparation only and does not include library quantification, normalization, or pooling

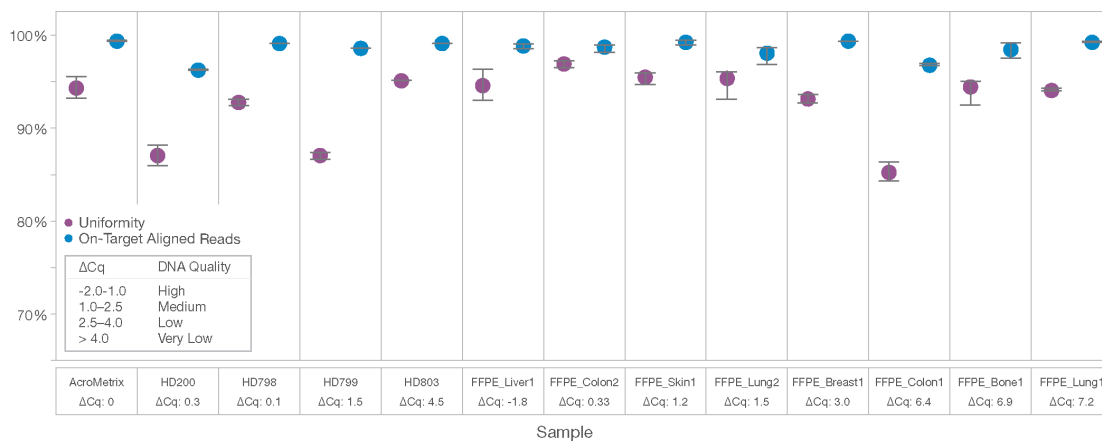
Data on file at Illumina, Inc. 2017

## Simple, Streamlined Workflow

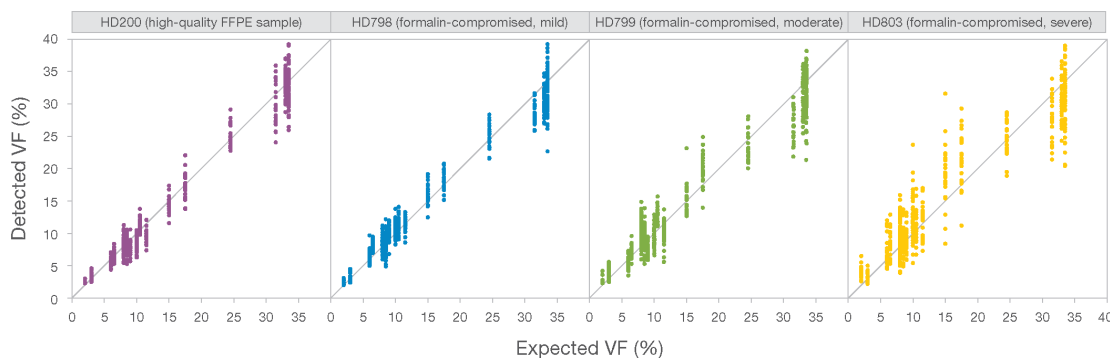
The AmpliSeq for Illumina Comprehensive Cancer Panel is part of a DNA-to-variant solution that offers streamlined content, easy-to-perform library preparation, push-button sequencing systems, and simplified data analysis.

Library preparation follows a straightforward, PCR-based protocol that can be completed in as little as 6 hours, with < 1.5 hours hands-on time. Resulting libraries can be normalized, pooled, and then loaded on to a flow cell for sequencing. Prepared libraries are sequenced on the NextSeq™ System (Table 2).

Resulting data can be analyzed locally using Local Run Manager or easily streamed into BaseSpace™ Sequence Hub. BaseSpace Sequence Hub and Local Run Manager can access the DNA Amplicon analysis workflow for alignment and variant calling. BaseSpace Sequence Hub can access BaseSpace Variant Interpreter, which assists with turning variant call data into annotated results.



**Figure 1: High Coverage Uniformity and On-Target Alignment**—DNA extracted from FFPE and HD samples of varying quality was prepared using the AmpliSeq for Illumina Comprehensive Cancer Panel and sequenced on the NextSeq System. Error bars indicate variability of technical replicates. ΔCq is an indicator of the quality of DNA isolated from FFPE tissues.



**Figure 2: High Concordance Between Expected and Detected Variant Frequency**—DNA from HD samples was prepared using the AmpliSeq for Illumina Comprehensive Cancer Panel and sequenced on the NextSeq System. Results show that 100% of expected SNVs were detected. ΔCq values are listed in Figure 1.

**Table 2: Illumina Sequencing Systems Recommended for Use with the AmpliSeq for Illumina Comprehensive Cancer Panel**

Instrument	Samples per Run	Run Time
NextSeq System (mid output)	4	26 hours
NextSeq System (high output)	12	29 hours

### Accurate Data

With the ability to assess hundreds of genes per sample, the AmpliSeq for Illumina Comprehensive Cancer Panel provides a high level of sample multiplexing, while maintaining excellent specificity and uniformity. To demonstrate assay capabilities, an AcroMetrix control sample, Horizon Discovery (HD) samples, and FFPE samples were evaluated using the AmpliSeq for Illumina Comprehensive Cancer Panel and the NextSeq™ System. Results showed high coverage uniformity and on-target percentage of aligned reads, even with varying sample quality and tissue type (Figure 1). In addition, HD samples of varying quality were evaluated for variant calling accuracy. Data showed high concordance between expected and detected variant frequency (VF) (Figure 2).

### Ordering Information

Order AmpliSeq for Illumina products online at [www.illumina.com](http://www.illumina.com)

Product	Catalog No.
AmpliSeq for Illumina Comprehensive Cancer Panel (24 reactions)	20019160
AmpliSeq for Illumina Library PLUS (24 reactions)	20019101
AmpliSeq for Illumina Library PLUS (96 reactions)	20019102
AmpliSeq for Illumina Library PLUS (384 reactions)	20019103
AmpliSeq for Illumina CD Indexes Set A (96 indexes, 96 samples)	20019105

### Learn More

To learn more about the AmpliSeq for Illumina Comprehensive Cancer Panel, [www.illumina.com/products/by-type/sequencing-kits/library-prep-kits/ampliseq-comprehensive-cancer-panel.html](http://www.illumina.com/products/by-type/sequencing-kits/library-prep-kits/ampliseq-comprehensive-cancer-panel.html)

To learn more about the AmpliSeq for Illumina targeted resequencing solution, read the overview at [www.illumina.com/content/dam/illumina-marketing/documents/products/datasheets/ampliseq-for-illumina-targeted-resequencing-solution-data-sheet-770-2017-022.pdf](http://www.illumina.com/content/dam/illumina-marketing/documents/products/datasheets/ampliseq-for-illumina-targeted-resequencing-solution-data-sheet-770-2017-022.pdf)