Ion AmpliSeq Cancer Hotspot Panel v2
Just one tube. Just 10ng of DNA. Just one day.

The **Ion AmpliSeq™ Cancer Hotspot Panel v2** allows translational and disease researchers to fast-track oncology research by surveying hotspot regions of 50 oncogenes and tumor suppressor genes, with wide coverage of the *KRAS*, *BRAF* and *EGFR* genes.

This research panel, with improved primer design, contains 207 primer pairs in a single tube and requires as little as 10ng of DNA, enabling researchers to sequence challenging samples such as formalin-fixed, paraffin-embedded (FFPE) tissue. The convenient predesigned panel allows researchers to focus on data generation and analysis, not on the labor-intensive primer design and target selection steps. While hybridization-based target selection methods require 7 to 72 hours to complete both target selection and library preparation, Ion AmpliSeq™ technology makes it possible to complete the entire process in about 3.5 hours using simple PCR reactions. Additionally, Ion AmpliSeq™ library construction steps are automated using standard 96-well plate–based protocols, further simplifying the workflow and allowing large projects to be rapidly completed without additional sample transfer steps.

"Sequencing gene panels with Ion Torrent™ technology is complete. You have the **Ion AmpliSeq™ gene selection technology**, the Ion PGM™ sequencing system and also the Ion Reporter™ analysis software.
For me, it is important to have a whole integrated solution, so researchers can advance from the tumor to results."

**Pierre Laurent-Puig, M.D., Ph.D.
Paris-Descartes University
Medical School, Paris, France**

Dataset for this panel is available at **thermofisher.com/ioncommunity**

Ion AmpliSeq Cancer Hotspot Panel v2	
Targets	Hotspot regions, including ~2,800 COSMIC mutations of 50 oncogenes and tumor suppressor genes, with wide coverage of the *KRAS*, *BRAF* and *EGFR* genes
Amplicon length	111–187 bp, average 154 bp
Primer pool size	207 primer pairs in 1 tube
Input DNA required	As little as 10ng per DNA sample
Time-to-results	Single-day workflow from DNA to annotated variants (run time varies by read length and chip type)
Sample multiplexing	Ion 314™ Chip: 2 samples, ~1,400X average coverage Ion 316™ Chip: 8 samples, ~1,400X average coverage Ion 318™ Chip: 16 samples, ~1,400X average coverage

	Specification	Observed performance (Ion 314 Chip)
Coverage uniformity [†]	>95%	>98%
On-target reads[‡]	>90%	>96%
Average depth of coverage	NA	>2,000X
SNP detection sensitivity	NA	98% detection rate for 5% variant frequency at positions with average sequencing coverage from 1,000X to 4,000X

† Coverage uniformity = percentage of bases covered at ≥20% of the mean coverage.
‡ On-target reads = percentage of reads that mapped to target regions, out of total mapped reads per run.

Thermo Fisher
SCIENTIFIC

Ideal for FFPE sequencing — requires as little as 10ng of DNA

Ion AmpliSeq technology enables the use of as little as 10ng of input DNA for the entire Ion AmpliSeq™ cancer research panel. This breakthrough technology is designed to deliver accurate sample representation even from FFPE samples, which are typically available only in small amounts and often exhibit variable quality. In addition, the primers in this panel are designed to produce, on average, 154 bp amplicons, so that even degraded samples can be used to generate reliable data. Target selection is completed using standard PCR equipment.

DNA to annotated variants in a single day

The single-day workflow from DNA to annotated variants allows you to address time-sensitive assays. No other target selection method enables you to go from FFPE sample to annotated variants in a single day using simple PCR-based technology. Construct libraries using the Ion AmpliSeq™ Library Kit Plus, then set up the Ion Chef™ System for automated template preparation in just 15 minutes of hands-on time. After sequencing on the Ion PGM System, automated analysis is then performed with Torrent Suite™ Software on the Ion PGM™ Torrent Server. The final step, using Ion Reporter™ Software, is optional and can be used to automate bioinformatics analysis including variant annotations. This software is ideal for use with routine DNA research assays (Figure 1).
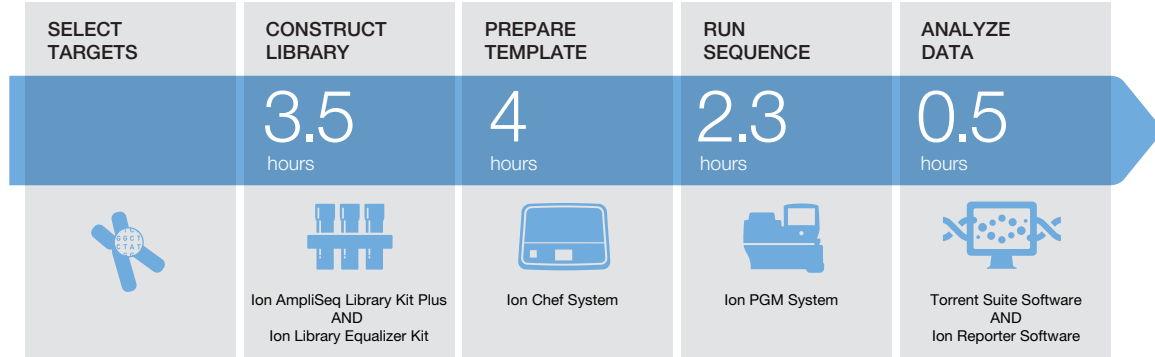
The Ion AmpliSeq Cancer Hotspot Panel v2 targets 50 genes

ABL1	EGFR	GNAS	KRAS	PTPN11
AKT1	ERBB2	GNAQ	MET	RB1
ALK	ERBB4	HNF1A	MLH1	RET
APC	EZH2	HRAS	MPL	SMAD4
ATM	FBXW7	IDH1	NOTCH1	SMARCB1
BRAF	FGFR1	JAK2	NPM1	SMO
CDH1	FGFR2	JAK3	NRAS	SRC
CDKN2A	FGFR3	IDH2	PDGFRA	STK11
CSF1R	FLT3	KDR	PIK3CA	TP53
CTNNB1	GNA11	KIT	PTEN	VHL

Applied Biosytems™ TaqMan™ Mutation Detection Assays are available for the genes listed above.

Confirm variants using TaqMan Assays

Integrated with the search portal for TaqMan Assays, Torrent Suite Software enables direct submission of detected variants for orthogonal confirmation experiments. Choose either TaqMan Mutation Detection Assays powered by castPCR™ technology on Applied Biosystems™ QuantStudio™ 6, 7 or 12K Flex Real-Time PCR Systems, or Applied Biosystems™ TaqMan™ SNP Genotyping Assays using digital PCR on the Applied Biosystems™ QuantStudio™ 3D Digital PCR System. Accurate and reliable results are achievable in about three hours.

Find out how to confirm variants at
thermofisher.com/ordertaqman

SELECT TARGETS	CONSTRUCT LIBRARY	PREPARE TEMPLATE	RUN SEQUENCE	ANALYZE DATA
	3.5 hours	**4** hours	**2.3** hours	**0.5** hours
	Ion AmpliSeq Library Kit Plus AND Ion Library Equalizer Kit	Ion Chef System	Ion PGM System	Torrent Suite Software AND Ion Reporter Software

Figure 1. Ion AmpliSeq panel workflow using an Ion 314 Chip and a 2.3-hour, 1 x 200-base sequencing run.

Ordering information

Product	Cat. No.
Ion AmpliSeq Cancer Hotspot Panel v2 (primer pool)	4475346
Ion AmpliSeq Library Kit 2.0 (8, 96 or 384 reactions for both PCR amplification and library construction)	4475345, 4480441, 4480442
Ion Xpress Barcode Adapters Kits	4474517, 4471250, 4474009, 4474518, 4474519, 4474520, 4474521
Additional Ion AmpliSeq products	
Ion AmpliSeq Comprehensive Cancer Panel (primer pool)	4477685
Ion AmpliSeq Custom Panels can be ordered via Ion AmpliSeq Designer. Go to **thermofisher.com/ampliseqcustom**	

Find out how to transform your oncology research at
thermofisher.com/cancergenomics

Thermo Fisher
SCIENTIFIC