Spotlight 59 oncology panel

All-in one solution for high sensitivity NGS from circulating tumor cells and cfDNA

FLUXION

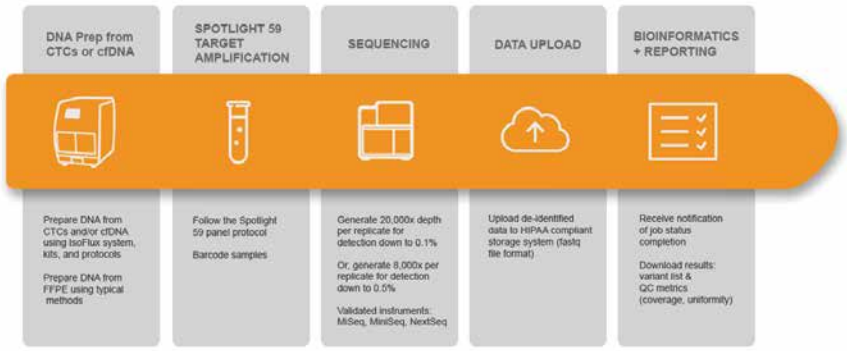
Designed specifically for detection of mutations from liquid biopsy samples, the **Spotlight 59** oncology panel is a turnkey solution for Illumina sequencers, offering ultra-sensitive variant detection from low input quantities of DNA. The high sensitivity of **Spotlight 59** makes it ideal for CTCs, cfDNA, and other challenging sample types such as FFPE. Combined with ERASE-Seq, Fluxion's statistically powered variant caller, **Spotlight 59** delivers 100% specificity at 0.2% allele frequency (no false positives across 27.5kb panel). Used in conjunction with Fluxion's IsoFlux Liquid Biopsy System, **Spotlight 59** creates a complete sample-to-answer assay for high sensitivity station detection of solid tumor cancers from blood samples.

The **Spotlight 59** oncology panel offers comprehensive and hotspot coverage of 59 clinically-relevant oncology-related genes, using a 277-amplicon design to generate multiplex libraries compatible with Illumina sequencing platforms. This product is a complete kit that includes all elements necessary for generating ready-to-sequence libraries, including primer pairs and indexed sequencing adapters.

The **Spotlight 59** oncology panel utilizes Illumina-compatible dual-indexed adapter sequences and has been validated on the MiniSeq, MiSeq, and HiSeq platforms. It is designed to work with ERASE-Seq, Fluxion's statistically powered variant caller that is available free of charge to Spotlight users.

Features

- Detection to 0.1% allele frequency
- No false positives across 27.5kb panel (average specificity >99.999%)
- Turnkey solution for library generation in a single kit, including sequencing adapters and sample barcodes
- Ready-to-sequence libraries in just 2 hours
- Starting input quantities as low as 200 cells (using IsoFlux NGS kit with WGA), or 20 ng DNA
- Average amplicon size of 140 bp for compatibility with cfDNA and FFPE
- ERASE-Seq variant calling included with kit purchase
- No MID (molecular IDs) required; increases sensitivity, reduces bias and assay complexity
- No primer trimming required



The **Spotlight 59** liquid biopsy workflow for ultra-accurate detection of rare variants starts with isolation of CTCs and/or cfDNA using the IsoFlux system, kits, and protocols. This is followed by targeted amplification and sequencing at 20,000X per sample. Fluxion's ERASE-Seq bioinformatics solution is accessed via secure upload to a HIPAA-compliant storage solution. Statistical comparison to a well-characterized normal control specific to the **Spotlight 59** panel is performed, allowing accurate variant calling to 0.1% allele frequency. Variant calls are available for download within 24 hours. The analysis service is provided free of charge for Spotlight 59 customers.

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Product specifications

Feature	Specification
Input DNA required	20-100 ng
Minimum starting cell number	200 total (CTCs plus background)
Starting sample	CTC, cfDNA, FFPE
Time required	2 hours
Number of amplicons	277 (Spotlight 59) + 104 (sample ID)
Amplicon size	Average 140 bp
Number of genes covered	59
Total target size	25.75 kb
On target percentage	> 90%
Coverage uniformity at > 20% of mean	> 90%
Limit of detection (at 40 ng input, for base substitutions)	0.1%, 0.3% using WGA
Validated sequencers	MiniSeq, MiSeq, HiSeq
Multiplexing on MiSeq v3 at 20,000X average depth	5 samples
ERASE-Seq bioinformatics	HIPAA-compliant cloud-based analysis included

Genes represented

Genes represented in the Spotlight 59 oncology panel and number of amplicons

ABL1	5	*DDR2*	1	*FLT3*	4	*IDH2*	2	*MSH6*	4	*RET*	6
AKT1	2	*DNMT3A*	1	*FOXL2*	1	*JAK2*	2	*NOTCH1*	3	*STK11*	5
ALK	2	*EGFR*	9	*GNA11*	2	*JAK3*	3	*NPM1*	1	*SMAD4*	10
APC	9	*ERBB2*	4	*GNAQ*	2	*KDR*	9	*NRAS*	3	*SMARCB1*	4
ATM	19	*ERBB4*	8	*GNAS*	2	*KIT*	14	*PDGFRA*	4	*SMO*	5
BRAF	2	*EZH2*	1	*H3F3A*	2	*KRAS*	3	*PIK3CA*	11	*SRC*	1
CDH1	3	*FBXW7*	6	*HIST1H3B*	4	*MAP2K1*	5	*PIK3R1*	8	*TP53*	21
CDKN2A	2	*FGFR1*	2	*HNF1A*	4	*MET*	6	*PTEN*	14	*TSC1*	1
CSF1R	2	*FGFR2*	4	*HRAS*	2	*MLH1*	1	*PTPN11*	2	*VHL*	3
CTNNB1	1	*FGFR3*	6	*IDH1*	1	*MPL*	1	*RB1*	12		

The **Spotlight 59** oncology panel includes both clinically relevant hotspot loci and regions of contiguous coverage, depending on the allele distribution across each target gene. The table depicts the genes represented, followed by the number of amplicons for each gene.

Contiguous, overlapping coverage is included for APC, ATM, EGFR, FBXW7, FGFR3, H3F3A, HNF1A, KIT, MSH6, PIK3CA, PTEN, SMAD4, and TP53.

Comprehensive coding exon coverage is included for TP53.

IsoFlux

Highly sensitive enrichment and detection of CTCs and protein markers from blood samples

The system combines the power of antibody-based magnetic bead separations with the precision of flow cytometry. Its superior performance is proven daily in labs around the world, setting standards for CTC isolation efficiency, purity, and ease of use.

Multiple kits available: EpCAM, EpCAM/EGFR, EpCAM/EGFR/Mesenchymal, and user-defined.

ACCELA

accela s.r.o., Služeb 4, 108 00 Prague 10, Czech Republic
Tel.: +420 255 700 886, Fax: +420 272 700 882
accela@accela.eu, www.accela.eu