

Tumor Hotspot MASTR Plus

Research Application

For the use of somatic and germline variant detection of selected target regions in 26 frequently mutated genes in:

- FFPE-derived DNA
- FFT- and blood-derived DNA

A molecular research panel for the identification of SNVs in the hotspots of 26 frequently mutated genes in solid tumors. This NGS assay is designed with input from selected INCa centers in France.

Assay Characteristics

Genes with hotspots included			
<i>AKT</i>	<i>ERBB2</i> (HER2)	<i>IDH1</i>	<i>PDGFRA</i>
<i>ALK</i>	<i>ERBB4</i>	<i>IDH2</i>	<i>PIK3R1</i>
<i>BRAF</i>	<i>FGFR2</i>	<i>KIT</i>	<i>PIK3CA</i>
<i>CDKN2A</i> (p16-INK4A, p14-ARF)	<i>FGFR3</i>	<i>KRAS</i>	<i>PTEN</i> (full gene coverage)
<i>CTNNB1</i> (β-catenin)	<i>H3F3A</i> (Histone H3, F3A)	<i>MEK1</i> (MAP2K1)	<i>STK11</i> (LKB1) (full gene coverage)
<i>DDR2</i>	<i>HIST1H3B</i> (Histone H1, 3B)	<i>MET</i>	
<i>EGFR</i>	<i>HRAS</i>	<i>NRAS</i>	

Assay Characteristics Cont.

Genomic region analyzed	25.7 kb
Number of amplicons	252
Amplicon length	128-245 bp
Number of plexes	4
Verified with NGS system	Illumina MiSeq
Designed to be compatible with	Illumina NextSeq, MiniSeq and Ion Torrent NGS Systems
Complete variant spectrum	SNVs

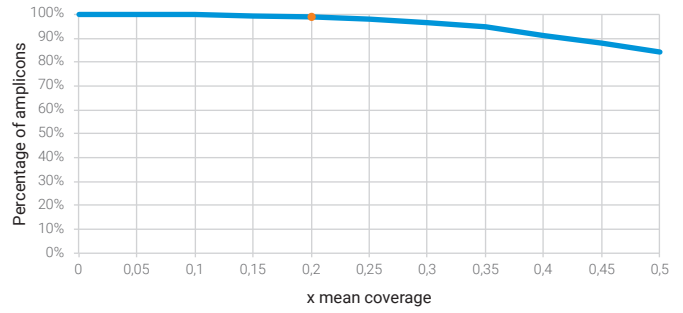
	Illumina MiSeq reagent kit v2	Illumina MiSeq reagent kit v3
Sequencer capacity Total reads	12,000,000	22,000,000
# samples/run @ 5% VAF _{sample} 20 reads per allele**	17	31
# samples/run @ 50% VAF _{sample} 20 reads per allele**	170	308*

*only 192 MID combinations available

** Number of samples per run for Illumina & IonTorrent NGS Systems can be calculated via the [sequencing calculator](#).

Performance

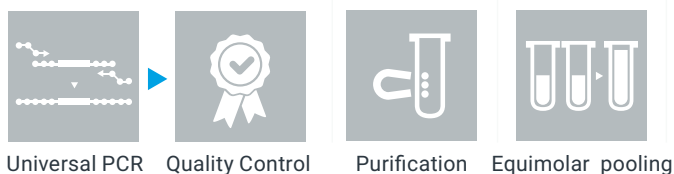
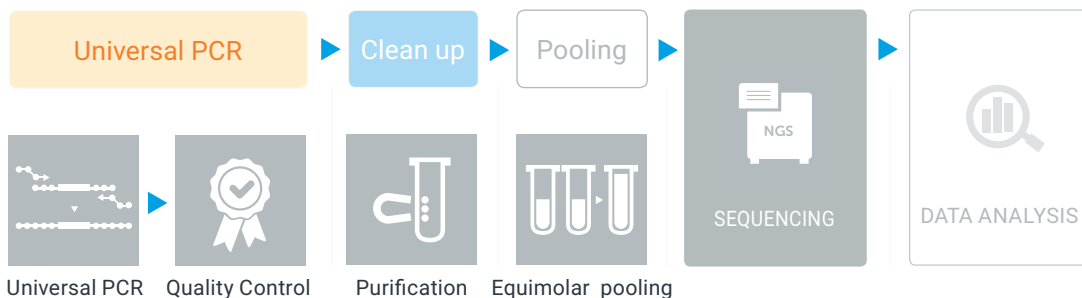
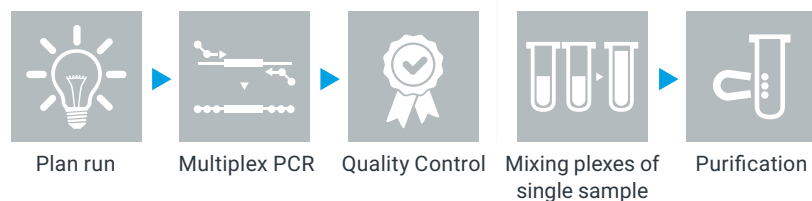
Uniformity of amplification (0.2x mean coverage)	98.8 %
On target read count	>97 %
DNA input	as low as ng per plex



Graph presenting the read counts of Tumor Hotspot MASTR Plus amplicons, showing their uniform representation.

Gene	Coverage and included hotspot mutations	Coverage						
		Lungs	Colon	Breasts/ovarians	Skin	Stomach	Blood	Pancreas, thyroid, prostate, glioblastoma, and others
<i>AKT</i>	Exon 4, includes mutations in the PH domain affecting Glu17, Phe35	✓	✓	✓		✓		✓
<i>ALK</i>	Exon 20 to 29, includes mutations in kinase domain affecting Ile1171, Phe1174, Leu1196, Phe1245, Gly1269, Arg1275 and Tyr1278	✓						✓
<i>BRAF</i>	Exon 11 and 15, includes mutations in kinase domain affecting Gly466, Gly469, Asp494, Val600 and Lys601	✓	✓		✓		✓	✓
<i>CDKN2A</i> (p16-INK4A, p14-ARF)	Full exon coverage	✓		✓				✓
<i>CTNNB1</i> (β-catenin)	Exon 3 includes mutations affecting Asp32, Ser33, Gly34, Ser37, Thr41 and Ser45	✓	✓				✓	✓
<i>DDR2</i>	All coding exons from exon 4 to 19	✓						
<i>EGFR</i>	Exon 18 to 21, spanning the kinase domain that includes mutations affecting Glu709, Gly719, Glu746-Pro753, Ser768 and Leu858	✓	✓	✓		✓		✓
<i>ERBB2</i> (HER2)	Exon 19 to 21, spanning the kinase domain that includes mutations affecting Leu755, Gly776, Val777 and Val842	✓		✓		✓		✓
<i>ERBB4</i>	Exon 10 and 12	✓				✓		✓
<i>FGFR2</i>	Exon 7, 12 and 14, including mutations affecting Ser252, Asn549 and Lys659			✓	✓	✓		✓
<i>FGFR3</i>	Exon 7, 9, 14 and 16, including mutations affecting Arg248, Ser249, Tyr373 and K650							✓
<i>H3F3A</i> (Histone H3, F3A)	Exon 2, including mutations affecting Lys28							✓
<i>HIST1H3B</i> (Histone H1, 3B)	Exon 1, including mutations affecting Lys28							✓
<i>HRAS</i>	Exon 2-4, including mutations affecting Gly12, Gly13 and Gln61							✓
<i>IDH1</i>	Exon 4 including mutations affecting Arg132							✓
<i>IDH2</i>	Exon 4 including mutations affecting Arg140 and Arg173							✓
<i>KIT</i>	Exon 8 to 11, 13, 14, 17 and 18, including mutations affecting aa417-419, 557-560, Leu576 and kinase domain mutations				✓	✓	✓	✓
<i>KRAS</i>	Exon 2 to 4, including mutations affecting Gly12 and Gly13	✓	✓					✓
<i>MEK1</i> (MAP2K1)	Exon 2 to 3, including mutations affecting Lys57 and mutations in Pro124 in the kinase domain	✓			✓			✓
<i>MET</i>	Exon 2, 10, 14 to 20, including mutations Glu168, Thr1010 and the kinase domain including Tyr1253	✓	✓					
<i>NRAS</i>	Exon 2-4, including mutations affecting Gly12, Gly13 and Tyr61	✓			✓			✓
<i>PDGFRA</i>	Exon 12, 14 and 18, including mutations in the kinase domain affecting Asp852					✓	✓	✓
<i>PIK3R1</i>	Exon 11-13, including mutations affecting aa 452 to 464		✓	✓				✓
<i>PIK3CA</i>	Exon 2, 3, 10, 11 and 21, including mutations affecting Glu39, Arg88, Glu542, Glu545 and His1047	✓	✓	✓				✓
<i>PTEN</i>	Full exon coverage	✓	✓	✓		✓		✓
<i>STK11</i> (LKB1)	Full exon coverage			✓				✓

MASTR Plus Workflow



Ordering Information

Cat. No.	Product Name	Sample
MR-0200.024	Tumor Hotspot MASTR Plus	24

* MID (Molecular Identifiers) kits are necessary to complete the workflow

www.agilent.com

Not for EU
genomics@agilent.com

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

This information is subject to change without notice.

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