

Product Description

SALSA® MLPA® Probemix P308-B3 MET

To be used with the MLPA General Protocol.

Version B3

As compared to the previous B2 lot, the probe length of one target probe has been adjusted, there is no change in the sequence detected. For complete product history see page 9.

Catalogue numbers:

- **P308-025R:** SALSA MLPA Probemix P308 MET, 25 reactions.
- **P308-050R:** SALSA MLPA Probemix P308 MET, 50 reactions.
- **P308-100R:** SALSA MLPA Probemix P308 MET, 100 reactions.

To be used in combination with a SALSA MLPA reagent kit and Coffalyser.Net data analysis software. MLPA reagent kits are either provided with FAM or Cy5.0 dye-labelled PCR primer, suitable for Applied Biosystems and Beckman/SCIEX capillary sequencers, respectively (see www.mrcholland.com).

Certificate of Analysis

Information regarding storage conditions, quality tests, and a sample electropherogram from the current sales lot is available at www.mrcholland.com.

Precautions and warnings

For professional use only. Always consult the most recent product description AND the MLPA General Protocol before use: www.mrcholland.com. It is the responsibility of the user to be aware of the latest scientific knowledge of the application before drawing any conclusions from findings generated with this product.

General information

The SALSA MLPA Probemix P308 MET is a **research use only (RUO)** assay for the detection of deletions or duplications in the *MET*, *PTEN* and *LRRK2* genes.

The receptor tyrosine kinase MET, also known as hepatocyte growth factor receptor (HGFR), is frequently amplified in human cancers. Amplification results in high cell surface densities and constitutive activation of HGFR even in the absence of growth factor stimulation by its endogenous ligand, hepatocyte growth factor (HGF). The *MET* gene is essential for survival and long-distance migration of epithelial and myogenic precursors during embryogenesis. Cancer cells hijack this biological process for invasion and metastasis. Multiple mechanisms can lead to aberrant MET activation, which occurs in many types of cancers.

MET gene amplifications leading to constitutive activation of the MET receptor are detected in a wide range of tumour types e.g. brain tumours, gastric cancer and lung tumours (Peters and Adjei 2012). Patients with papillary renal carcinomas carry both germline and somatic point mutations in *MET* (Schmidt et al. 1997; Schmidt et al. 1999). The amplification of leucine-rich repeat kinase-2 (*LRRK2*) is required for oncogenic MET signalling in papillary renal and in thyroid carcinomas and acts downstream of MET activation (Looyenga et al. 2011). Loss of tumour suppressor protein PTEN together with amplification of *MET* can be an alternative mechanism for primary resistance for tyrosine kinase inhibitors (EGFR-TKIs) (Engelman et al. 2007; Sos et al. 2009).

This SALSA MLPA probemix is not CE/FDA registered for use in diagnostic procedures. Purchase of this product includes a limited license for research purposes.

Gene structure and transcript variants:

Entrez Gene shows transcript variants of each gene: <http://www.ncbi.nlm.nih.gov/sites/entrez?db=gene>
For NM_ mRNA reference sequences: <http://www.ncbi.nlm.nih.gov/sites/entrez?db=nucleotide>
Locus Reference Genomic (LRG) database: <http://www.lrg-sequence.org/>
Matched Annotation from NCBI and EMBL-EBI (MANE): <https://www.ncbi.nlm.nih.gov/refseq/MANE/>
Tark – Transcript Archive: <http://tark.ensembl.org/>

Exon numbering

The *MET*, *PTEN* and *LRRK2* exon numbering used in this P308-B3 MET product description is the exon numbering from the MANE project (release version 1.0) based on MANE Select transcripts as indicated in Table 2. The exon numbering used in previous versions of this product description can be found in between brackets in Table 2. As changes to the databases can occur after release of this product description, the NM_ sequence and exon numbering may not be up-to-date. Please note that exon numbering for the same genes might be different from literature and in other MRC Holland product descriptions, where other resources used for exon numbering are indicated.

Probemix content

The SALSA MLPA Probemix P308-B3 MET contains 48 MLPA probes with amplification products between 130 and 493 nucleotides (nt). This includes 23 probes for the *MET* gene and three flanking probes for *MET* gene. This probemix also includes four probes for *PTEN* gene and four probes for *LRRK2* gene. In addition, 14 reference probes are included detecting autosomal chromosomal locations, which are relatively copy number stable regions in various cancer types. The identity of the genes detected by the reference probes is available in Table 3. Complete probe sequences are available online (www.mrcholland.com).

This probemix contains nine quality control fragments generating amplification products between 64 and 105 nt: four DNA Quantity fragments (Q-fragments), two DNA Denaturation fragments (D-fragments), one Benchmark fragment, and one chromosome X and one chromosome Y-specific fragment (see table below). More information on how to interpret observations on these control fragments can be found in the MLPA General Protocol and online at www.mrcholland.com.

Length (nt)	Name
64-70-76-82	Q-fragments (only visible with <100 ng sample DNA)
88-96	D-fragments (low signal indicates incomplete denaturation)
92	Benchmark fragment
100	X-fragment (X chromosome specific)
105	Y-fragment (Y chromosome specific)

MLPA technique

The principles of the MLPA technique (Schouten et al. 2002) are described in the MLPA General Protocol (www.mrcholland.com). More information on the use of MLPA in tumour applications can be found in Hömig-Hölzel and Savola (2012).

MLPA technique validation

Internal validation of the MLPA technique using 16 DNA samples from healthy individuals is required, in particular when using MLPA for the first time, or when changing the sample handling procedure, DNA extraction method or instruments used. This validation experiment should result in a standard deviation ≤ 0.10 for all probes over the experiment.

Required specimens

Extracted DNA, which includes DNA derived from paraffin-embedded tissues, free from impurities known to affect MLPA reactions. For more information please refer to the section on DNA sample treatment found in the MLPA General Protocol. More information on the use of FFPE tissue samples for MLPA can be found in Atanesyan et al. (2017).

Reference samples

A sufficient number (≥ 3) of reference samples should be included in each MLPA experiment for data normalisation. All samples tested, including reference DNA samples, should be derived from the same tissue type, handled using the same procedure, and prepared using the same DNA extraction method when possible. Reference samples should be derived from different unrelated individuals who are from families without a

history of cancer. More information regarding the selection and use of reference samples can be found in the MLPA General Protocol (www.mrcholland.com).

Positive control DNA samples

See the section “Positive samples” on the P308 product page on our website.

Data analysis

Coffalyser.Net software should be used for data analysis in combination with the appropriate lot-specific MLPA Coffalyser sheet. For both, the latest version should be used. Coffalyser.Net software is freely downloadable at www.mrcholland.com. Use of other non-proprietary software may lead to inconclusive or false results. For more details on MLPA quality control and data analysis, including normalisation, see the Coffalyser.Net Reference Manual.

Interpretation of results

The standard deviation of each individual probe over all the reference samples should be ≤ 0.10 and for germline analysis the final ratio (FR) of each individual reference probe in the patient samples should be between 0.80 and 1.20. When these criteria are fulfilled, the following cut-off values for the FR of the probes can be used to interpret MLPA results for autosomal chromosomes or pseudo-autosomal regions:

Copy number status	Final ratio (FR)
Normal	$0.80 < FR < 1.20$
Homozygous deletion	$FR = 0$
Heterozygous deletion	$0.40 < FR < 0.65$
Heterozygous duplication/gain	$1.30 < FR < 1.65$
Heterozygous triplication/homozygous duplication/gain	$1.75 < FR < 2.15$
Ambiguous copy number	All other values

Note: The term “dosage quotient”, used in older product description versions, has been replaced by “final ratio” to become consistent with the terminology of the Coffalyser.Net software. (Calculations, cut-offs and interpretation remain unchanged.) Please note that the Coffalyser.Net software also shows arbitrary borders as part of the statistical analysis of results obtained in an experiment. As such, arbitrary borders are different from the final ratio cut-off values shown here above.

Please note that these above mentioned final ratios are only valid for germline testing. Final ratios are affected both by percentage of tumour cells and by possible subclonality.

- Arranging probes according to chromosomal location facilitates interpretation of the results and may reveal more subtle changes such as those observed in mosaic or subclonal cases. Analysis of parental samples may be necessary for correct interpretation of complex results in germline level.
- False positive results: Please note that abnormalities detected by a single probe (or multiple consecutive probes) still have a considerable chance of being a false positive result. Sequence changes (e.g. SNVs, point mutations) in the target sequence detected by a probe can be one cause. Incomplete DNA denaturation (e.g. due to salt contamination) can also lead to a decreased probe signal, in particular for probes located in or near a GC-rich region. The use of an additional purification step or an alternative DNA extraction method may resolve such cases. Additionally, contamination of DNA samples with cDNA or PCR amplicons of individual exons can lead to an increased probe signal (Varga et al. 2012). Analysis of an independently collected secondary DNA sample can exclude these kinds of contamination artefacts.
- Normal copy number variation in healthy individuals is described in the database of genomic variants: <http://dgv.tcag.ca/dgv/app/home>. Users should always consult the latest update of the database and scientific literature when interpreting their findings.
- Not all abnormalities detected by MLPA are pathogenic. In some genes, intragenic deletions are known that result in very mild or no disease (as described for *DMD* by Schwartz et al. 2007). For many genes, more than one transcript variant exists. Copy number changes of exons that are not present in all transcript

variants may not have clinical significance. Duplications that include the first or last exon of a gene (e.g. exons 1-3) might not result in inactivation of that gene copy.

- Copy number changes detected by reference probes or flanking probes are unlikely to have any relation to the condition tested for.
- False results can be obtained if one or more peaks are off-scale. For example, a duplication of one or more exons can be obscured when peaks are off-scale, resulting in a false negative result. The risk on off-scale peaks is higher when probemixes are used that contain a relatively low number of probes. Coffalyser.Net software warns for off-scale peaks while other software does not. If one or more peaks are off-scale, rerun the PCR products using either: a lower injection voltage or a shorter injection time, or a reduced amount of sample by diluting PCR products.

P308 specific note

- In samples from tumour tissues, reference probes are more prone to have deviating copy number results as compared to blood derived germline samples. When regions targeted by reference probes are affected by copy number alterations, it can help to turn the slope correction off in Coffalyser.Net analysis to get the correct copy number interpretation on the target region.

Limitations of the procedure

- In most populations, the major cause of genetic defects in the *MET*, *PTEN* and *LRRK2* genes are small (point) mutations, most of which will not be detected by using SALSA MLPA Probemix P308 MET.
- MLPA cannot detect any changes that lie outside the target sequence of the probes and will not detect copy number neutral inversions or translocations. Even when MLPA did not detect any aberrations, the possibility remains that biological changes in that gene or chromosomal region *do* exist but remain undetected.
- Sequence changes (e.g. SNVs, point mutations) in the target sequence detected by a probe can cause false positive results. Mutations/SNVs (even when >20 nt from the probe ligation site) can reduce the probe signal by preventing ligation of the probe oligonucleotides or by destabilising the binding of a probe oligonucleotide to the sample DNA.
- MLPA analysis on tumour samples provides information on the *average* situation in the cells from which the DNA sample was purified. Gains or losses of genomic regions or genes may not be detected if the percentage of tumour cells is low. In addition, subclonality of the aberration affects the final ratio of the corresponding probe. Furthermore, there is always a possibility that one or more reference probes *do* show a copy number alteration in a patient sample, especially in solid tumours with more chaotic karyotypes.

Confirmation of results

Copy number changes detected by only a single probe always require confirmation by another method. An apparent deletion detected by a single probe can be due to e.g. a mutation/polymorphism that prevents ligation or destabilises the binding of probe oligonucleotides to the DNA sample. Sequence analysis can establish whether mutations or polymorphisms are present in the probe target sequence. The finding of a heterozygous mutation or polymorphism indicates that two different alleles of the sequence are present in the sample DNA and that a false positive MLPA result was obtained.

Copy number changes detected by more than one consecutive probe should be confirmed by another independent technique such as long range PCR, qPCR, array CGH or Southern blotting, whenever possible. Deletions/duplications of more than 50 kb in length can often be confirmed by FISH.

LOVD and COSMIC mutation databases

LOVD database for *MET* gene: <https://databases.lovd.nl/shared/genes/MET>; LOVD database for *PTEN* gene: <https://databases.lovd.nl/shared/genes/PTEN> and LOVD database for *LRRK2* gene: <https://databases.lovd.nl/shared/genes/LRRK2>; COSMIC database: <http://cancer.sanger.ac.uk/cosmic>. We strongly encourage users to deposit positive results in these above mentioned LOVD and COSMIC Databases. Recommendations for the nomenclature to describe deletions/duplications of one or more exons can be found on <http://varnomen.hgvs.org/>.

Please report copy number changes detected by the reference probes, false positive results due to SNVs and unusual results (e.g., a duplication of *MET* exons 10 and 12 but not exon 11) to MRC Holland: info@mrcholland.com.

Table 1. SALSA MLPA Probemix P308-B3 MET

Length (nt)	SALSA MLPA probe	Chromosomal position (hg18) / Exon ^a				Location (hg18) in kb
		Reference	MET	PTEN	LRRK2	
64-105	Control fragments – see table in probemix content section for more information					
130	Reference probe 00797-L21056	5q31				05-132,038
139	Reference probe 10913-L31828	2p25				02-001,396
148	MET probe 18448-L23950		Exon 21			07-116,223
155	MET probe 18449-L23949		Exon 5			07-116,168
162	MET probe 10327-L24422		Exon 17			07-116,206
168	MET probe 00453-L23587		Exon 2			07-116,126
176	MET probe 10317-L23588		Exon 7			07-116,185
182	Reference probe 08731-L23593	9q21				09-078,101
189	MET probe 22585-L23584		Exon 11			07-116,190
197 ±	PTEN probe 06729-L23164			Intron 2		10-089,644
203	MET probe 18450-L24423		Exon 14			07-116,199
211	Reference probe 04533-L03922	2q24				02-166,607
218	MET probe 10320-L23586		Exon 10			07-116,187
224	LRRK2 probe 04282-L23585				Exon 41	12-039,020
229	Reference probe 13569-L23952	19p13				19-013,425
236	MET probe 10329-L23575		Exon 19			07-116,211
241 ~	CFTR probe 03839-L23953		7q31.2			07-116,907
249	MET probe 18451-L23954		Exon 18			07-116,209
255	MET probe 10323-L23583		Exon 13			07-116,199
261	Reference probe 07819-L23623	1q31				01-194,726
268 ~	IKZF1 probe 13873-L31794		7p12.2			07-050,412
275	MET probe 18435-L23582		Exon 16			07-116,205
283	MET probe 18452-L23628		Exon 3			07-116,159
289	MET probe 22614-L31904		Exon 8			07-116,185
294	Reference probe 11900-L23408	6p12				06-052,028
303	MET probe 18453-L31792		Exon 21			07-116,225
310	MET probe 10316-L23581		Exon 6			07-116,183
318	Reference probe 16438-L23633	18q21				18-045,645
325	MET probe 18454-L23630		Exon 4			07-116,167
332	MET probe 10325-L24169		Exon 15			07-116,202
338	Reference probe 13423-L31795	12p13				12-006,103
346	MET probe 18455-L23631		Exon 12			07-116,197
355	PTEN probe 18694-L24425			Intron 3		10-089,675
363	Reference probe 14981-L16717	6q22				06-129,461
373 #	PTEN probe 03638-L22839			Exon 5		10-089,683
382	LRRK2 probe 04278-L23577				Exon 1	12-038,905
393	MET probe 10312-L22033		Exon 1			07-116,100
400	Reference probe 09433-L09682	11q13				11-066,150
409	MET probe 10330-L23955		Exon 20			07-116,223
417	MET probe 18456-L23632		Exon 2			07-116,127
427 ~	CAV2 probe 04707-L02740		7q31.2			07-115,928
436	LRRK2 probe 04279-L23590				Exon 10	12-038,932
444	PTEN probe 17395-L21062			Exon 9		10-089,715
454	Reference probe 12790-L31796	2q13				02-108,880
463	LRRK2 probe 04280-L03684				Exon 15	12-038,955
474 ¥	MET probe 10319-L32155		Exon 9			07-116,186
483	Reference probe 06676-L06254	11p15				11-006,369
493	Reference probe 09772-L22978	15q21				15-042,706

^a See section Exon numbering on page 2 for more information.

¥ Changed in version B3. Minor alteration, no change in sequence detected.

± SNV rs146326040 (probe at 197 nt) could influence the probe signal. In case of apparent deletions, it is recommended to sequence the region targeted by this probe.

– Flanking probe. Included to help determine the extent of a deletion/duplication. Copy number alterations of only the flanking or reference probes are unlikely to be related to the condition tested.

This probe's specificity relies on a single nucleotide difference compared to a related gene or pseudogene. As a result, an apparent duplication of only this probe can be the result of a non-significant single nucleotide sequence change in the related gene or pseudogene.

SNVs located in the target sequence of a probe can influence probe hybridization and/or probe ligation. Please note: not all known SNVs are mentioned in the tables above. Single probe aberration(s) must be confirmed by another method.

Table 2. MET, PTEN and LRRK2 probes arranged according to chromosomal location

Length (nt)	SALSA MLPA probe	Gene exon ^a	Location (hg18) / Ligation site	Partial sequence ^b (24 nt adjacent to ligation site)	Distance to next probe
MET gene at 7q31.2. Indicated ligation sites are in NM_000245.4 (MANE Select).					
268 ~	13873-L31794	<i>IKZF1</i>	7p12.2	TACGAATGCTTG-ATGCCTCGGGAG	65.5 Mb
427 ~	04707-L02740	<i>CAV2</i>	7q31.2	CAGCAAATACGT-AATGTACAAGTT	172.2 kb
<i>Centromeric</i>					
393	10312-L22033	MET exon 1	366-367	TTCTCCACTGGT-TCCTGGGCACCG	26.6 kb
		<i>start codon</i>	397-399 (exon 1)		
168	00453-L23587	MET exon 2	446-447	GCTCCTGTTTAC-CTTGGTGCAGAG	0.5 kb
417	18456-L23632	MET exon 2	962-963	TTCATCTGTAAA-GGACCGGTTTCAT	32.1 kb
283	18452-L23628	MET exon 3	1699-1700	TGGGTCAATTCA-GCGAAGTCTCT	8.2 kb
325	18454-L23630	MET exon 4	1830-1831	CCTCATGTGAAT-TTTCTCTGGAC	1.0 kb
155	18449-L23949	MET exon 5	2040-2039 reverse	AGGCATTCTCC-GATCGCACACAT	14.4 kb
310	10316-L23581	MET exon 6	2144-2145	AAGGCTGACCAT-ATGTGGCTGGGA	2.1 kb
176	10317-L23588	MET exon 7	2280-2281	ACAGTTGGTCCT-GCCATGAATAAG	0.2 kb
289	22614-L31904	MET exon 8	2391-2392	ATTTCGCCGAAA-TACGGTCTATG	0.9 kb
474	10319-L32155	MET exon 9	2608-2609	GCATCTTCAGTT-ACCGTGAAGATC	0.8 kb
218	10320-L23586	MET exon 10	2679-2680	AGCACAATAACA-GGTGTTGGGAAA	3.7 kb
189	22585-L23584	MET exon 11	2789-2790	TAATTCAGAGAT-AATCTGTTGTAC	6.7 kb
346	18455-L23631	MET exon 12	3084-3085	TTATGCACGGTC-CCCAATGACCTG	1.8 kb
255	10323-L23583	MET exon 13	3233-3234	AACAGCACTGTT-ATTACTACTTGG	0.3 kb
203	18450-L24423	MET exon 14	3387-3388	GAAATGGTTTCA-AATGAATCTGTA	3.1 kb
332	10325-L24169	MET exon 15	3573-3574	AGTGCTCTAAAT-CCAGAGCTGGTC	2.4 kb
275	18435-L23582	MET exon 16	3708-3709	GATGGCAAGAAA-ATTCAGTGTGCT	1.4 kb
162	10327-L24422	MET exon 17	3797-3798	CATGAAAGATTT-TAGTCATCCCAA	3.3 kb
249	18451-L23954	MET exon 18	4019-4020	CTTGGCTGCAAG-AACTGTATGTA	1.3 kb
236	10329-L23575	MET exon 19	4149-4150	AAGTGGATGGCT-TTGGAAAGTCTG	12.2 kb
409	10330-L23955	MET exon 20	6 nt before exon 20	TGTTCTTGTTT-TACTAGTGGTCC	0.5 kb
148	18448-L23950	MET exon 21	4559-4560	CTCCTTCTGGGA-GACATCATAGTG	1.7 kb
		<i>stop codon</i>	4567-4569 (exon 21)		
303	18453-L31792	MET exon 21	6242-6243	TACACCCACCC-TCATTACATCAT	682.2 kb
<i>Telomeric</i>					
241 ~	03839-L23953	<i>CFTR</i>	7q31.2	AGAAAAAGGGTT-GAGCGGCAGGCA	-
PTEN gene at 10q23.31. Indicated ligation sites are in NM_000314.8 (MANE Select).					
		<i>start codon</i>	846-848 (exon 1)		
197 ±	06729-L23164	PTEN intron 2 (exon 2)	217 nt after exon 2 reverse	TATCACATAAGT-ACCTGATTATGT	31.0 kb
355	18694-L24425	PTEN intron 2 (exon 3)	145 nt before exon 3	GGGGTATTTGTT-GGATTATTTATT	7.8 kb
373 #	03638-L22839	PTEN exon 5	1250-1251	GGTGTAATGATA-TGTGCATATTTA	32.1 kb
444	17395-L21062	PTEN exon 9	6 nt before exon 9	TAAATTTCTTT-CTCTAGGTGAAG	-

		stop codon	2055-2057 (exon 9)		
LRRK2 gene at 12q12. Indicated ligation sites are in NM_198578.4 (MANE Select).					
		start codon	136-138 (exon 1)		
382	04278-L23577	LRRK2 exon 1	230-231	AGGAAAAACAGAT-AGAAACGCTGGT	26.3 kb
436	04279-L23590	LRRK2 exon 10	1286-1285 reverse	TCTTCTCATGTA-AACTGTTTTGGT	23.2 kb
463	04280-L03684	LRRK2 exon 15	1904-1905	GGATTCAGTGCT-TCACACACTGCA	65.6 kb
224	04282-L23585	LRRK2 exon 41	6106-6107	CAGCCATGATTA-TATACCGAGACC	-
		stop codon	7717-7719 (exon 51)		

^a See section Exon numbering on page 2 for more information.

^b Only partial probe sequences are shown. Complete probe sequences are available at www.mrcholland.com. Please notify us of any mistakes: info@mrcholland.com.

± SNV rs146326040 (probe at 197 nt) could influence the probe signal. In case of apparent deletions, it is recommended to sequence the region targeted by this probe.

– Flanking probe. Included to help determine the extent of a deletion/duplication. Copy number alterations of only the flanking or reference probes are unlikely to be related to the condition tested.

This probe's specificity relies on a single nucleotide difference compared to a related gene or pseudogene. As a result, an apparent duplication of only this probe can be the result of a non-significant single nucleotide sequence change in the related gene or pseudogene.

SNVs located in the target sequence of a probe can influence probe hybridization and/or probe ligation. Please note: not all known SNVs are mentioned in the tables above. Single probe aberration(s) must be confirmed by another method.

Table 3. Reference probes arranged according to chromosomal location.

Length (nt)	SALSA MLPA probe	Gene	Chromosomal band (hg18)	Partial sequence (24 nt adjacent to ligation site)	Location (hg18) in kb
261	07819-L23623	<i>KCNT2</i>	1q31	CCTTACCATTCA-TTTCCTTGTA	01-194,726
139	10913-L31828	<i>TPO</i>	2p25	GAGTGGCTGTAA-TTTGGGCCATTA	02-001,396
454	12790-L31796	<i>EDAR</i>	2q13	AGAATCAAGGCT-TTTGTGATATGT	02-108,880
211	04533-L03922	<i>SCN1A</i>	2q24	TGAAATGAGAAA-GAGAAGGTCAAG	02-166,607
130	00797-L21056	<i>IL4</i>	5q31	ATCGACACCTAT-TAATGGGTCTCA	05-132,038
294	11900-L23408	<i>PKHD1</i>	6p12	TGCTCTCTGGAT-TCAAGACTGAAA	06-052,028
363	14981-L16717	<i>LAMA2</i>	6q22	TGCCAAAGATGA-TGAGGTCATCTG	06-129,461
182	08731-L23593	<i>PCSK5</i>	9q21	GTCCTGGATTCT-ATGGTGACCAAG	09-078,101
483	06676-L06254	<i>SMPD1</i>	11p15	CTGCTGAAGATA-GCACCACCTGCC	11-006,369
400	09433-L09682	<i>RBM14</i>	11q13	CACCAACTGTCT-TCTTCTCTCGAC	11-066,150
338	13423-L31795	<i>VWF</i>	12p13	AAGGGCCTCCAT-TTCTCATTCTG	12-006,103
493	09772-L22978	<i>SPG11</i>	15q21	TTTCTTCAGGAT-TGATAGTCATTC	15-042,706
318	16438-L23633	<i>MYO5B</i>	18q21	CGTGGCCGACCA-AGCCACGCAGAA	18-045,645
229	13569-L23952	<i>CACNA1A</i>	19p13	GCTCCTACTTGA-GGAATGGCTGGA	19-013,425

Complete probe sequences are available at www.mrcholland.com.

Related products

For related products, see the [product page](#) on our website.

References

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Selected publications using SALSA MLPA Probemix P308 MET


- Sebai M et al. (2022). Novel germline MET pathogenic variants in French patients with papillary renal cell carcinomas type I. *Hum Mutat*. 43:316-27.

P308 product history	
Version	Modification
B3	As compared to the previous B2 lot, the probe length of one target probe has been adjusted, there is no change in the sequence detected.
B2	One flanking probe has been added, five reference probes have been replaced, and several probes have a change in length but no change in the sequence targeted.
B1	First release.

Implemented changes in the product description
<p>Version B3-02 – 19 May 2025 (04P)</p> <ul style="list-style-type: none"> - Description of the position of the 197 nt and 355 nt probes changed (no change in actual target sites). - Positive control DNA samples section: information moved to product page on website. - Related SALSA MLPA products section replaced with reference to product page on website. <p>Version B3-01 – 02 October 2023 (04P)</p> <ul style="list-style-type: none"> - Product description adapted to a new product version (version number changed, changes in Table 1 and Table 2) and to a new template. - Various minor textual or layout changes. - Ligation sites of the probes targeting the <i>MET</i> gene updated according to new version of the NM_000245.4 MANE Select sequence. <p>Version B2-01 – 05 February 2020 (02P)</p> <ul style="list-style-type: none"> - Product description adapted to a new product version (version number changed, changes in Table 1 and Table 2). - Various minor textual or layout changes. - Information about positive control DNA samples added to page 2. - Ligation sites of the probes targeting the <i>MET</i> gene updated according to new version of the NM_001127500.3, <i>PTEN</i> gene according to the NM_000314.8 and <i>LRRK2</i> gene according to the NM_198578.4 reference sequence. - Warning added to Table 1 and 2a for probe specificity relying on a single nucleotide difference between target gene and related gene or pseudogene. - Added probemixes P105, P315 and P458 in the “Related SALSA MLPA probemixes” section on page 7.

- For uniformity, the chromosomal locations and bands in this document are now all based on hg18 (NCBI36).

More information: www.mrcholland.com; www.mrcholland.eu

	MRC Holland bv; Willem Schoutenstraat 1 1057 DL, Amsterdam, The Netherlands
E-mail	info@mrcholland.com (information & technical questions) order@mrcholland.com (orders)
Phone	+31 888 657 200