

Product Description SALSA® MLPA® Probemix P301-B1 Medulloblastoma mix 1

To be used with the MLPA General Protocol.

Version B1. As compared to version A2, several reference probes have been replaced and several probes have a change in length but no change in the sequence detected. For complete product history see page 7.

Catalogue numbers:

- **P301-025R:** SALSA MLPA Probemix P301 Medulloblastoma mix 1, 25 reactions.
- **P301-050R:** SALSA MLPA Probemix P301 Medulloblastoma mix 1, 50 reactions.
- **P301-100R:** SALSA MLPA Probemix P301 Medulloblastoma mix 1, 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.mlpa.com).

Certificate of Analysis: Information regarding storage conditions, quality tests, and a sample electropherogram from the current sales lot is available at www.mlpa.com.

Precautions and warnings: For professional use only. Always consult the most recent product description AND the MLPA General Protocol before use: www.mlpa.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.

This SALSA MLPA probemix is for basic research and intended for experienced MLPA users only!

This probemix enables the quantification of genes or chromosomal regions in which the occurrence or relevance of copy number changes is not yet well-established. Interpretation of results may be complicated, and MRC Holland may only be able to provide basic support.

General information: The SALSA MLPA Probemix P301 Medulloblastoma mix 1 is a **research use only (RUO)** assay for the detection of deletions or duplications in the chromosomes 6, 14q, 16 and 17, which are thought to be associated with medulloblastoma.

Medulloblastoma (MB) is the most common paediatric primary central nervous system (CNS) tumour and accounts for between 15% and 20% of CNS tumours in patients under the age of 20. It is a highly invasive embryonal neuroepithelial tumour that arises in the cerebellum and has a tendency to disseminate throughout the CNS early in its course. Overall survival is 50-60% at five years, although this decreases to 30% in the longer term due to local recurrence and/or metastasis. There are four distinct molecular subtypes of MB (WNT, sonic hedgehog (SHH), Group 3, and Group 4) which can be used for patient risk stratification and that have the potential to identify new therapeutic strategies for the treatment of MB (Taylor et al. 2012). These molecular subtypes of MB include also characteristic and recurrent copy number alterations, which are covered by the P301, P302 and P303 Medulloblastoma probemixes.

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.

Probemix content: The SALSA MLPA Probemix P301-B1 Medulloblastoma mix 1 contains 47 MLPA probes with amplification products between 126 and 492 nucleotides (nt). This includes 35 probes for the chromosomes 6, 14q, 16 and 17. In addition, 12 reference probes are included that target relatively copy number stable regions in various cancer types including medulloblastoma. Partial/complete probe sequences and the identity of the genes detected by the reference probes are available in Table 2b and online (www.mlpa.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.mlpa.com.

Length (nt)	Name
64-70-76-82	Q-fragments (only visible with <100 ng sample DNA)
88-96	D-fragments (low signal of 88 nt and 96 nt fragment 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.mlpa.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 reference 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 healthy individuals without a history of cancer. More information regarding the selection and use of reference samples can be found in the MLPA General Protocol.

Positive control DNA samples: MRC-Holland cannot provide positive DNA samples. Inclusion of a positive sample in each experiment is recommended. Coriell Institute (<https://catalog.coriell.org>) and Leibniz Institute DSMZ (<https://www.dsmz.de/home.html>) have a diverse collection of biological resources which may be used as a positive control DNA sample in your MLPA experiments. Sample ID numbers NA12721, NA01221, NA09367, NA07994, NA06802, NA05966, NA06226, NA02325, NA09687, NA13284, NA08039, NA05875, NA12074, NA06047, NA13476 and NA16445 from the Coriell Institute have been tested with this P301-B1 probemix at MRC-Holland and can be used as a positive control samples to detect deletions and duplications in multiple genomic regions as described in the table below. The quality of cell lines can change; therefore samples should be validated before use.

Sample name	Source	Chromosomal position of CNA*	Altered target genes in P301-B1	Expected CNA
NA12721	Coriell Institute	6p22.3	<i>E2F3, SOX4</i>	Heterozygous duplication
NA01221	Coriell Institute	6q15	<i>MAP3K7</i>	Heterozygous duplication
NA09367	Coriell Institute	6q23.2-q23.3	<i>SGK1, MYB</i>	Heterozygous duplication
NA07994	Coriell Institute	6q23.3-q26	<i>MYB, MYCT1, MAP3K4</i>	Heterozygous duplication
NA06802	Coriell Institute	6q26	<i>MAP3K4</i>	Heterozygous deletion
NA05966	Coriell Institute	14q23.1-q24.3	<i>OTX2, MLH3</i>	Heterozygous duplication
NA06226/NA02325	Coriell Institute	16p13.3	<i>AXIN1, MEFV</i>	Heterozygous duplication
NA09687	Coriell Institute	16p13.3	<i>AXIN1</i>	Heterozygous deletion
		16q22.3-q24.3	<i>ZFH3, FANCA</i>	Heterozygous duplication
NA13284	Coriell Institute	16p13.3	<i>AXIN1</i>	Heterozygous duplication
NA08039	Coriell Institute	16p13.3	<i>MEFV</i>	Heterozygous duplication
NA05875	Coriell Institute	16p11.2	<i>TGFB111</i>	Heterozygous deletion
NA12074	Coriell Institute	16q22.3	<i>ZFH3</i>	Heterozygous deletion
NA06047	Coriell Institute	17p13.2-p13.3	<i>HIC1, PAFAH1B1, ATP2A3</i>	Heterozygous deletion
NA13476	Coriell Institute	17p11.2	<i>PRPSAP2</i>	Heterozygous deletion

Sample name	Source	Chromosomal position of CNA*	Altered target genes in P301-B1	Expected CNA
NA16445	Coriell Institute	17q25.3	<i>TK1, BIRC5, ARHGDI1, RAC3</i>	Heterozygous duplication

* Indicated chromosomal bands accommodate genes targeted by MLPA probes, however, the whole extent of copy number alteration (CNA) present in this cell line cannot be determined by this P301-B1 Medulloblastoma mix 1 probemix.

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.mlpa.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 . When this criterion is fulfilled, the following cut-off values for the dosage quotient (DQ) of the probes can be used to interpret MLPA results for autosomal chromosomes or pseudo-autosomal regions:

Copy number status	Dosage quotient
Normal	$0.80 < DQ < 1.20$
Homozygous deletion	$DQ = 0$
Heterozygous deletion	$0.40 < DQ < 0.65$
Heterozygous duplication	$1.30 < DQ < 1.65$
Heterozygous triplication/Homozygous duplication	$1.75 < DQ < 2.15$
Ambiguous copy number	All other values

Please note that these above mentioned dosage quotients are only valid for germline testing. Dosage quotients 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 cases.
- 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. Incomplete DNA denaturation (e.g. due to salt contamination) can lead to a decreased probe signal, in particular for probes located in or near a GC-rich region or in or near the *SOX4, MYB, AXIN1, HIC1, CDK5R1, ARHGDI1 and RAC3* genes. 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.
- When running MLPA products, the capillary electrophoresis protocol may need optimization. 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: lower injection voltage / injection time settings, or a reduced amount of sample by diluting PCR products.

Limitations of the procedure:

- In most populations, the majority of alterations in the chromosomal regions (chromosomes 6, 14q, 16 and 17) included in this probemix are small (point) mutations, which will not be detected by using SALSA MLPA Probemix P301 Medulloblastoma mix 1.
- 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. SNPs, point mutations, small indels) in the target sequence detected by a probe can cause false positive results. Mutations/SNPs (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.

Please report false positive results due to SNPs and unusual results to MRC-Holland: info@mlpa.com.

Table 1. SALSA MLPA Probemix P301-B1 Medulloblastoma mix 1

Length (nt)	SALSA MLPA probe	Reference	Chromosomal position (hg18)			
			Chr 6	Chr 14q	Chr 16	Chr 17
64-105	Control fragments – see table in probemix content section for more information					
126 †	Reference probe 18709-L21698	5q31				
131 ‹	RAC3 probe 11724-L14159					17q25.3
142	Reference probe 08143-L08022	5p12				
148	ERBB2 probe 00675-L00572					17q12
154	ADGRG1 probe 10195-L10655				16q13	
166	MEFV probe 05451-L04888				16p13.3	
173 #	PAFAH1B1 probe 04175-L03579					17p13.3
178 †	RARA probe 10209-L32138					17q21.2
184 *	Reference probe 04857-L32200	5p13				
190	OTX2 probe 10204-L13655			14q23.1		
196	TK1 probe 10217-L10687					17q25.3
202	TGFB1I1 probe 13121-L10718				16p11.2	
208 †	NLK probe 21533-L32170					17q11.2
212 ‹	HIC1 probe 13122-L00949					17p13.3
220	MLH3 probe 02107-L14737			14q24.3		
226	MYCT1 probe 10201-L10663		6q25.2			
232 † ‹	AXIN1 probe 21510-L10649				16p13.3	
243 *	Reference probe 19134-L25333	21q22				
250 ‹	ARHGDI1 probe 08097-L12510					17q25.3
257	TOP2A probe 01055-L00628					17q21.2
265 ‹	SOX4 probe 10214-L10681		6p22.3			
274	ATP2A3 probe 10186-L10646					17p13.2
283	RPS6KB1 probe 08617-L08629					17q23.1
293 ‹	MYB probe 10245-L00359		6q23.3			
301 ‡	TP53 probe 02379-L13860					17p13.1
310	AXIN2 probe 00440-L14744					17q24.1
317 *	Reference probe 06580-L30649	2q24				
325 †	BIRC5 probe 03717-L32161					17q25.3
332 †	PRPSAP2 probe 01454-L14433					17p11.2
337	PPM1D probe 03195-L02652					17q23.2
346	SGK1 probe 10212-L10679		6q23.2			
355	Reference probe 06711-L06315	15q24				
364	MAP3K4 probe 10196-L14739		6q26			
373	MMP2 probe 04765-L04113				16q12.2	
381	VEGFA probe 12579-L11003		6p21.1			
391 † ‹	CDK5R1 probe 22788-L10650					17q11.2
400	FANCA probe 04181-L14740				16q24.3	
409 *	Reference probe 09720-L32156	12q24				
418	MAP3K7 probe 10198-L10658		6q15			
424	RCVRN probe 10211-L10677					17p13.1
432 †	Reference probe 21891-L12107	4q12				
445	ZFX3 probe 04738-L04086				16q22.3	
454	Reference probe 07607-L07292	15q26				
465 *	Reference probe 12460-L32162	22q12				
472	E2F3 probe 12029-L12891		6p22.3			
481 *	Reference probe 08614-L32159	12p12				
492 *	Reference probe 17001-L18577	20q11				

* New in version B1.

† Changed in version B1. Minor alteration, no change in sequence detected.

‡ This probe may be non-specific. Apparent deletions/gains of this TP53 probe always need to be confirmed with another probemix (e.g. P056 TP53) or with another technique.

‹ Probe located in or near a GC-rich region. A low signal can be caused by salt contamination in the DNA sample leading to incomplete DNA denaturation, especially of GC-rich regions.

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.

Table 2a. P301-B1 probes arranged according to chromosomal location

Length (nt)	SALSA MLPA probe	Gene	Location (hg18)	Partial sequence ⁺ (24 nt adjacent to ligation site)	Distance to next probe
Chromosome 6					
472	12029-L12891	E2F3	6p22.3	CTTTGCCTGTAA-TAAGAAAGTGCG	1,1 Mb
265 «	10214-L10681	SOX4	6p22.3	CCAGCATTCGAG-AAACTCCTCTCT	22,1 Mb
381	12579-L11003	VEGFA	6p21.1	GTGGATTTTGGA-AACCAGCAGAAA	47,5 Mb
418	10198-L10658	MAP3K7	6q15	GTGTTTACAGTG-TTCCCAAGGAGT	43,2 Mb
346	10212-L10679	SGK1	6q23.2	GAGGATGGGTCT-GAACGACTTTAT	1,0 Mb
293 «	10245-L00359	MYB	6q23.3	ATGCGTCGGAAG-GTCGAACAGGAA	17,5 Mb
226	10201-L10663	MYCT1	6q25.2	CACTTCCCACAG-TCTGAGCCGTCC	8,3 Mb
364	10196-L14739	MAP3K4	6q26	AAACGCATGTCA-ACCAAACATCAG	-
Chromosome 14q					
190	10204-L13655	OTX2	14q23.1	TCCTGCATGCAG-AGGTCCTATCCC	18,2 Mb
220	02107-L14737	MLH3	14q24.3	TGCCGCCTTATT-GAAGCTCTGTCC	-
Chromosome 16					
232 «	21510-L10649	AXIN1	16p13.3	GATCATCGGCAA-AGTGGAGAAGGT	3,0 Mb
166	05451-L04888	MEFV	16p13.3	GGCCTCACTGGA-GGACGTGGGCCA	28,2 Mb
202	13121-L10718	TGFB111	16p11.2	CAGGAACCTAAT-GCCACTCAGTTC	22,7 Mb
373	04765-L04113	MMP2	16q12.2	TGCAACCTGTTT-GTGCTGAAGGAC	2,2 Mb
154	10195-L10655	ADGRG1	16q13	GATTGTGGTACA-GAACACCAAAGT	15,3 Mb
445	04738-L04086	ZFH3	16q22.3	ATTCTTAGCAAT-AAGAACATCTCC	16,8 Mb
400	04181-L14740	FANCA	16q24.3	CATGTTGCTGTG-GACATGTACTTG	-
Chromosome 17					
212 «	13122-L00949	HIC1	17p13.3	AGAGTGTGCGGA-AAGCGCGGCGGG	615 kb
173	04175-L03579	PAFAH1B1	17p13.3	CTGTTCTGCAGA-TATGACCATTAA	1,3 Mb
274	10186-L10646	ATP2A3	17p13.2	GAGGTGTTTCGAG-TCACGCTTCCCC	3,7 Mb
301 ‡	02379-L13860	TP53	17p13.1	ACTGCCCAACAA-CACCAGCTCCTC	2,2 Mb
424	10211-L10677	RCVRN	17p13.1	CTTCCAGACGAT-GAAAACACGCCG	9,0 Mb
332	01454-L14433	PRPSAP2	17p11.2	TAGAAACCAAGA-TGAACATAACCA	4,8 Mb
208	21533-L32170	NLK	17q11.2	AGAATTAGATGA-ATCCCCTCATAT	4,3 Mb
391 «	22788-L10650	CDK5R1	17q11.2	TCATGAGCTCAA-AGATGCTGCAGA	7,3 Mb
148	00675-L00572	ERBB2	17q12	GGTGCAGGGCTA-CGTGCTCATCGC	644 kb
178	10209-L32138	RARA	17q21.2	TCTCTGGACATT-GACCTCTGGGAC	61 kb
257	01055-L00628	TOP2A	17q21.2	AAGCCCTTCAAT-GGAGAAGATTAT	19,5 Mb
283	08617-L08629	RPS6KB1	17q23.1	CAGGAGTGTTTG-ACATAGACCTGG	730 kb
337	03195-L02652	PPM1D	17q23.2	TGTGGTCATCAT-TCGGGGCATGAA	4,9 Mb
310	00440-L14744	AXIN2	17q24.1	CCCGAAGCTCTT-GTGAAGTGTCTT	12,7 Mb
196	10217-L10687	TK1	17q25.3	ATCTTTCACCAA-GATGGGTGGCAC	40 kb
325	03717-L32161	BIRC5	17q25.3	CTCTACATTCAA-GAACTGGCCCTT	3,7 Mb
250 «	08097-L12510	ARHGDI1	17q25.3	CAGGTTAACCGA-GAGATAGTGTCC	165 kb
131 «	11724-L14159	RAC3	17q25.3	GGCAGGATCCTG-TCCTCTGTGCCG	-

‡ This probe may be non-specific. Apparent deletions/gains of this TP53 probe always need to be confirmed with another probemix (e.g. P056 TP53) or with another technique.

« Probe located in or near a GC-rich region. A low signal can be caused by salt contamination in the DNA sample leading to incomplete DNA denaturation, especially of GC-rich regions.

Table 2b. Reference probes arranged according to chromosomal location

Length (nt)	SALSA MLPA probe	Gene	Location (hg18)	Partial sequence + (24 nt adjacent to ligation site)	Distance to next probe
317	06580-L30649	SCN2A	2q24	AACTTGGTTTGG-CAAATGTGGAAG	-
432	21891-L12107	SGCB	4q12	TGTAGAAAACAA-CAAACCTCTAT	-
184	04857-L32200	NIPBL	5p13	CTGCAATGTTGC-AAAAATCCTAGA	7,3 Mb
142	08143-L08022	FGF10	5p12	GATGCTGCCAAT-TCAAGGTTTGTG	87,7 Mb
126	18709-L21698	IL4	5q31	ATCGACACCTAT-TAATGGGTCTCA	-
481	08614-L32159	H2AFJ	12p12	ACTCAGGACCAA-GTTCTGGGAAGA	101,4 Mb
409	09720-L32156	NOS1	12q24	AGAATATGACAT-TGTGCACCTGGA	-
355	06711-L06315	HEXA	15q24	AGGCACTCCACT-TCCTCCTCGAGC	26,8 Mb
454	07607-L07292	IGF1R	15q26	CATGGTAGCCGA-AGATTTACAGT	-
492	17001-L18577	SAMHD1	20q11	CCCTGTCACCTC-AAGTTTGAGGAT	-
243	19134-L25333	KCNJ6	21q22	CTCGAAGCTCCT-ACATCACCAGTG	-
465	12460-L32162	LARGE1	22q12	AGGAATAGCTGC-ACCTTCGAACCT	-

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

Related SALSA MLPA probemixes

- **P302 Medulloblastoma mix 2:** Contain probes for chromosomes 2, 3, 7 and 9.
- **P303 Medulloblastoma mix 1:** Contain probes for chromosomes 1, 4q, 5q, 8, 10 and 20.
- **P251-P252-P253 Neuroblastoma:** Contain probes for multiple chromosomal regions that frequently show copy number changes in neuroblastoma tumours (chromosomes 1, 2, 3, 4, 7, 9, 11, 12, 14 and 17).

References

- Atanesyan L et al. (2017). Optimal fixation conditions and DNA extraction methods for MLPA analysis on FFPE tissue-derived DNA. *Am J Clin Pathol.* 147:60-8.
- Hömig-Hölzel C and Savola. (2012). Multiplex ligation-dependent probe amplification (MLPA) in tumor diagnostics and prognostics. *Diagn Mol Pathol.* 21:189-206.
- Schouten JP et al. (2002). Relative quantification of 40 nucleic acid sequences by multiplex ligation-dependent probe amplification. *Nucleic Acids Res.* 30:e57.
- Schwartz M et al. (2007). Deletion of exon 16 of the dystrophin gene is not associated with disease. *Hum Mutat.* 28:205.
- Taylor MD et al. (2012) Molecular subgroups of medulloblastoma: the current consensus. *Acta Neuropathol.* 123:465-72.
- Varga RE et al. (2012). MLPA-based evidence for sequence gain: pitfalls in confirmation and necessity for exclusion of false positives. *Anal Biochem.* 421:799-801.

Selected publications using SALSA MLPA Probemix P301 Medulloblastoma mix 1

- Goschzik T et al. (2015) Molecular stratification of medulloblastoma: comparison of histological and genetic methods to detect Wnt activated tumours. *Neuropathol Appl Neurobiol.* 41:135-44.
- Łastowska M et al. (2015) Contrast enhancement pattern predicts poor survival for patients with nonWNT/SHH medulloblastoma tumours. *J Neurooncol.* 123:65-73.
- Pietsch T et al. (2014) Prognostic significance of clinical, histopathological, and molecular characteristics of medulloblastomas in the prospective HIT2000 multicenter clinical trial cohort. *Acta Neuropathol.* 128:137-49.
- Trubicka J et al. (2016) Identification of a novel inherited ALK variant M1199L in the WNT type of medulloblastoma. *Folia Neuropathol.* 54:23-30.
- Yehia M et al. (2019) Association of Aggresomes with Survival Outcomes in Pediatric Medulloblastoma. *Sci Rep.* 9:12605.

P301 Product history	
Version	Modification
B1	Several reference probes have been replaced and several probes have a change in length but no change in the sequence detected.

A2	Control fragments (QDX) have been optimized, one reference probe has been replaced and one added.
A1	First release.

Implemented changes in the product description

Version B1-01 — 16 September 2020 (02P)

- Joint product description for P301, P302 and P303 probemixes is now divided into separate product descriptions.
- Product description adapted to a new product version and to a new template (version number changed, changes in Table 1 and Table 2).
- Various minor textual or layout changes.
- Name of *GPR56* gene is updated in Tables 1 & 2a to *ADGRG1* according to the new HUGO nomenclature.
- For uniformity, the chromosomal locations and bands in this document are now all based on hg18 (NCBI36).

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

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