Product Description SALSA® MLPA® Probemix P417-B3 BAP1

To be used with the MLPA General Protocol.

Version B3. As compared to B2, one flanking probe and two reference probes have been replaced, one flanking probe is added and one reference probe is removed. Additionally, several probes have a change in length but no change in the sequences detected. For complete product history see page 8.

Catalogue numbers:
- **P417-025R:** SALSA MLPA Probemix P417 BAP1, 25 reactions.
- **P417-050R:** SALSA MLPA Probemix P417 BAP1, 50 reactions.
- **P417-100R:** SALSA MLPA Probemix P417 BAP1, 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.

General information: The SALSA MLPA Probemix P417 BAP1 is a **research use only (RUO)** assay for the detection of deletions or duplications in the *BAP1* gene, which is associated with BAP1 tumour predisposition syndrome.

The *BAP1* (BRCA1 associated protein 1) gene is a tumour suppressor gene that functions in the *BRCA1* growth control pathway and has an important role controlling in cell proliferation and growth inhibition. The *BAP1* gene locates in the 3p21 region, which is frequently affected by LOH or deletions in several cancer types including lung, breast and ovarian cancer, but also in uveal melanoma and mesothelioma.

BAP1 tumour predisposition syndrome (OMIM# 614327) is inherited in an autosomal dominant manner. Individuals carrying heterozygous *BAP1* mutations are at high-risk for the development of a variety of tumours, including benign melanocytic tumours as well as several malignant tumours, including uveal melanoma, cutaneous melanoma, malignant mesothelioma on exposure to asbestos, and other cancer types, such as renal cell carcinoma, basal cell carcinoma, lung adenocarcinoma and meningioma (Wiesner et al. 2011, Abdel-Rahman et al. 2011, Boru et al. 2019).

In addition, research suggests that uveal melanoma with monosomy of chromosome 3 (frequency 50-60% of all uveal melanomas) represents a distinct pathological entity as compared to uveal melanoma with normal disomy 3. The putative target gene on the 3p arm is *BAP1*, as inactivating somatic mutations of *BAP1* are identified in >80% of patients with metastasizing uveal melanoma (Harbour et al. 2010). Moreover, *BAP1* is commonly inactivated by somatic mutations and 3p21.1 losses in malignant pleural mesothelioma (Testa et al. 2011, Bott et al. 2011).

More information about *BAP1* tumour predisposition syndrome is available at:
https://www.ncbi.nlm.nih.gov/books/NBK390611/

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/

Exon numbering: The *BAP1* exon numbering used in this P417-B3 BAP1 product description is the exon numbering from the LRG_529 sequence. The exon numbering and NM_ sequence used have been retrieved on 02/2020. As changes to the NCBI database can occur after release of this product description, exon numbering may not be up-to-date.

Probemix content: The SALSA MLPA Probemix P417-B3 BAP1 contains 42 MLPA probes with amplification products between 132 and 395 nucleotides (nt). This includes 17 probes for the *BAP1* gene and 10 flanking probes included for the determination of the extent of the deletion or duplication. In addition, 14 reference probes are included that detect target relatively copy number stable regions in various cancer types including melanocytic tumours and mesothelioma. 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-fragments (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 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 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.

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 ACC-203 (SK-N-MC, ACC-512 (ARH-77) and NA04127 from the Leibniz Institute DSMZ and Coriell Institute have been

tested with this P417-B3 BAP1 probemix at MRC-Holland and can be used as a positive control samples, see 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 P417-B3	Expected CNA
SK-N-MC	DSMZ	3p12.3-p22.2	*BAP1* and 3p flanking genes	Heterozygous deletion
ARH-77	DSMZ	3p12.3-p22.2	*BAP1* and 3p flanking genes	Heterozygous deletion
NA04127	Coriell Institute	3p21.31-p22.2	*MLH1, RBM5, RASSF1* and *ZMYND10*	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 P417-B3 BAP1 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 and, additionally, the dosage quotient (DQ) of each individual reference probe in the patient samples should be between 0.80 and 1.20, when analysis is performed on germline samples. When these criteria are fulfilled, the following cut-off values for the 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. When analysis is performed on germline samples, inclusion of parental samples may be necessary for correct interpretation of complex results.
- 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. 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.

P417 specific notes:
- 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 *BAP1* are small (point) mutations, most of which will not be detected by using SALSA MLPA Probemix P417 BAP1.
- 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.

***BAP1* mutation database:** https://databases.lovd.nl/shared/genes/BAP1 We strongly encourage users to deposit positive results in the Leiden Open Variation Database (LOVD). Recommendations for the nomenclature to describe deletions/duplications of one or more exons can be found on http://varnomen.hgvs.org/.

Please report false positive results due to SNPs and unusual results (e.g., a duplication of *BAP1* exons 4 and 6 but not exon 5) to MRC-Holland: info@mlpa.com.

Table 1. SALSA MLPA Probemix P417-B3 BAP1

Length (nt)	SALSA MLPA probe	Chromosomal position (hg18)[a]			
		Reference	***BAP1*** (3p21.1)	Flanking probes	Location (hg18) in kb
64-105	Control fragments – see table in probemix content section for more information				
132	Reference probe 00797-L21698	5q31			05-132.038
137	Reference probe 06972-L14818	17p13			17-006.271
142	Reference probe 10699-L11281	6p12			06-052.053
149	**BAP1 probe** 17400-L21109		Exon 7		03-052.416
155	Reference probe 04566-L03955	16q13			16-055.482
160	**BAP1 probe** 17401-L21110		Exon 2		03-052.419
166	Reference probe 12741-L21552	21q22			21-042.050
173	**BAP1 probe** 17402-L21111		Exon 3		03-052.419
179	**BAP1 probe** 17403-L21112		Exon 11		03-052.414
184 ¬	**RASSF1 probe** 12125-L21391			3p21.31	03-050.353
190	**BAP1 probe** 17404-L21113		Exon 15		03-052.412
196	Reference probe 09763-L10178	15q21			15-042.740
202 * ¬	**DNAH1 probe** 22655-L31877			3p21.1	03-052.408
208	**BAP1 probe** 17405-L21114		Exon 5		03-052.417
214 ¬	**ZMYND10 probe** 03206-L13082			3p21.31	03-050.358
221 ¬	**HESX1 probe** 07223-L21127			3p14.3	03-057.207
227	**BAP1 probe** 16643-L19177		Exon 4		03-052.418
232	**BAP1 probe** 17406-L21392		Exon 1		03-052.419
238 *	Reference probe 19768-L31764	12q12			12-041.074
244	**BAP1 probe** 17407-L21116		Exon 13		03-052.413
251 *	Reference probe 08811-L31763	2p13			02-071.620
257	**BAP1 probe** 17408-L21394		Exon 6		03-052.416
263	**BAP1 probe** 21243-L29765		Exon 14		03-052.412
270 ¥	**BAP1 probe** 17398-L32045		Exon 9		03-052.415
276 ¥ ¬	**ROBO1 probe** 06439-L32044			3p12.3	03-079.071
282 ¥	**BAP1 probe** 21244-L32082		Exon 16		03-052.412
287 ¥	**BAP1 probe** 17411-L32043		Exon 10		03-052.415
293 ¥	Reference probe 08936-L32083	11p15			11-020.586
299	**BAP1 probe** 17412-L21397		Exon 8		03-052.416
308	**BAP1 probe** 17413-L21122		Exon 12		03-052.414
317 ¬	**FHIT probe** 04710-L01787			3p14.2	03-059.883
325	Reference probe 16275-L21395	19p13			19-012.782
332	**BAP1 probe** 17414-L21123		Exon 17		03-052.411
338	Reference probe 12785-L15496	2q13			02-108.972
346 ¬	**MITF probe** 10794-L11434			3p14.1	03-070.081
352 * ¬	**PHF7 probe** 22656-L31878			3p21.1	03-052.430
359 ¥	Reference probe 11614-L31880	12p13			12-004.894
364 ¬	**RBM5 probe** 15897-L18094			3p21.31	03-050.105
372	Reference probe 06016-L21128	19q13			19-059.319
378 ¬	**CPOX probe** 14836-L21403			3**q**11.2	03-099.792
385 ¬	**MLH1 probe** 16176-L21699			3p22.2	03-037.057
395	Reference probe 05914-L21838	18p11			18-013.724

a) See above section on exon numbering for more information.

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

Table 2a. *BAP1* and flanking probes arranged according to chromosomal location

Length (nt)	SALSA MLPA probe	Gene (exon[a])	Location / Ligation site	Partial sequence[b] (24 nt adjacent to ligation site)	Distance to next probe
385 ¬	16176-L21699	MLH1	3p22.2	CGTGGGCTGTGT-GAATCCTCAGTG	13.0 **M**b
364 ¬	15897-L18094	RBM5	3p21.31	ATATGATGACTA-CCGAGACTATGA	248.6 kb
184 ¬	12125-L21391	RASSF1	3p21.31	CAAAGCCAGCGA-AGCACGGGCCCA	4.9 kb
214 ¬	03206-L13082	ZMYND10	3p21.31	CCGCACTGCGCA-GGCGCGGCTAAC	2.0 **M**b
202 ¬	22655-L31877	DNAH1	3p21.1	CTTGAGTGCCTT-GAGTAGGTCTTG	3.2 kb
			Telomeric		
***BAP1*, at 3p21.1.** Ligation sites are indicated according to NM_004656.4.					
		stop codon	*2318-2320 (ex 17)*		
332	17414-L21123	**BAP1, ex 17**	2483-2484	TCCATCGTGCCC-TGAGGCCTGACA	0.6 kb
282	21244-L32082	**BAP1, ex 16**	2117-2118	TCCTACAGATTG-ATGACCAGAGAA	0.2 kb
190	17404-L21113	**BAP1, ex 15**	2042-2043	CACTGCTGAAGT-GTGTGGAGGCTG	0.3 kb
263	21243-L29765	**BAP1, ex 14**	2014-2015	GAGAAATACTCA-CCCAAGGTGAGC	0.4 kb
244	17407-L21116	**BAP1, ex 13**	1717-1718	CACATCTCCAAG-GTGCTTTTTGGA	1.0 kb
308	17413-L21122	**BAP1, ex 12**	1291-1292	AGCCGAGTTCCA-GTCCGCCCACCC	0.6 kb
179	17403-L21112	**BAP1, ex 11**	1245-1246	GTCCCCCATGCA-GGTAAGCTGGGA	0.8 kb
287	17411-L32043	**BAP1, ex 10**	960-961	CCACAAGTCTCA-AGAGTCACAGCT	0.5 kb
270	17398-L32045	**BAP1, ex 9**	826-827	AACCTGATGGCA-GTGGTGCCCGAC	0.5 kb
299	17412-L21397	**BAP1, ex 8**	782-783	TCGGCCTCGCCA-CTGCAGGGTAAG	0.4 kb
149	17400-L21109	**BAP1, ex 7**	613-614	GGCCTTAGTGCA-GTGCGGACCATG	0.2 kb
257	17408-L21394	**BAP1, ex 6**	513-514	GTAGAGCAAAGG-ATATGCGATTGG	0.6 kb
208	17405-L21114	**BAP1, ex 5**	386-387	TGGTTTCACAGC-TGATACCCAACT	0.4 kb
227	16643-L19177	**BAP1, ex 4**	344-345	ATACGTCCGTGA-TTGATGATGATA	1.1 kb
173	17402-L21111	**BAP1, ex 3**	224-225	AAGTGGAGGAGA-TCTACGACCTTC	0.2 kb
160	17401-L21110	**BAP1, ex 2**	188-187 reverse	ACCGAAATCTTC-CACGAGCAGGGT	0.1 kb
232	17406-L21392	**BAP1, ex 1**	141-140 reverse	GCTCCAGCCAGC-CCTTATTCATCT	11.1 kb
		start codon	*131-133 (ex 1)*		
			Centromeric		
352 ¬	22656-L31878	PHF7	3p21.1	CAACAGAGTGTT-GAGAACATCCAG	4.8 **M**b
221 ¬	07223-L21127	HESX1	3p14.3	ATCGATATTAGA-GAAGACTTAGCT	2.7 **M**b
317 ¬	04710-L01787	FHIT	3p14.2	TCTCCAGCCTTC-CTGGGAAGAACA	10.2 **M**b
346 ¬	10794-L11434	MITF	3p14.1	TCTTTATGGAAA-CCAAGGTCTGCC	9.0 **M**b
276 ¬	06439-L32044	ROBO1	3p12.3	CCACCTCGCATT-GTTGAACACCCT	20.7 **M**b
			– Centromere –		
378 ¬	14836-L21403	CPOX	3**q**11.2	GTGCTATGGGCG-TGAGCTCTGTTA	-

a) See above section on exon numbering for more information.

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

¬ 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.

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)	Location (hg18) in kb
251	08811-L31763	DYSF	2p13	TACCACCTACCT-GAGTATGTCGAA	02-071.620
338	12785-L15496	EDAR	2q13	CCCAGAACTGGA-TGGTACCTGACT	02-108.972
132	00797-L21698	IL4	5q31	ATCGACACCTAT-TAATGGGTCTCA	05-132.038
142	10699-L11281	PKHD1	6p12	AGGGTCTGTACT-TCCTGGAAGCAT	06-052.053
293	08936-L32083	SLC6A5	11p15	TGTTTGCCTCCT-TTGTGTCTGTAC	11-020.586
359	11614-L31880	KCNA1	12p13	AACTAAACCAAT-TGATTTAATAGT	12-004.894
238	19768-L31764	PPHLN1	12q12	CAAGTGGGCTGC-TGAAAAGCTAGA	12-041.074
196	09763-L10178	SPG11	15q21	AACCATTTTGTA-TAGCTGTAGTAG	15-042.740
155	04566-L03955	SLC12A3	16q13	TGTGTCATGAGG-ATGCGGGAGGGA	16-055.482
137	06972-L14818	AIPL1	17p13	ATCGGCTCTTCA-AGCTGGGCCGCT	17-006.271
395	05914-L21838	RNMT	18p11	TACAATGAACTT-CAGGAAGTTGGT	18-013.724

Length (nt)	SALSA MLPA probe	Gene	Location (hg18)	Partial sequence (24 nt adjacent to ligation site)	Location (hg18) in kb
325	16275-L21395	RNASEH2A	19p13	GGTCAAGGCCAA-AGCAGATGCCCT	19-012.782
372	06016-L21128	PRPF31	19q13	ACAAGTGCAAGA-ACAATGAGAACC	19-059.319
166	12741-L21552	RIPK4	21q22	AAGCCAAGAAGA-TGGAGATGGCCA	21-042.050

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

Related SALSA MLPA probemixes

- **P027 Uveal melanoma:** Contains probes for chromosomes 1p, 3, 6p and 8q
- **P226 SDH:** Contains probes for all exons of the *SDHB, SDHC, SDHD, SDHAF1* and *SDHAF2* genes.
- **P419 CDKN2A/2B-CDK4:** Contains probes for *CDKN2A/2B* and *CDK4* genes and flanking probes on chromosome 9.
- **P429 SDHA-MAX:** Contains probes for most exons (10 of 15 exons) of *SDHA* gene and all five exons of *MAX* gene.
- **ME024 9p21 CDKN2A/2B region:** Contains probes for *CDKN2A/2B* region at 9p21.3 and for the surrounding genes, both for copy number and methylation status determination.

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P417 Product history	
Version	*Modification*
B3	Two flanking probes and two reference probes have been replaced and one reference probe is removed. Additionally, several probes have a change in length but no change in the sequences detected.
B2	Two reference probes have been added and two target probes have a minor change in length.
B1	First unrestricted release. New probes for *BAP1* gene have been included for exons that were not covered in the previous version. In addition, all reference probes have been replaced.
A1	First restricted release.

Implemented changes in the product description

Version B3-01 — 26 March 2020 (02P)
- Product description adapted to a new product version (version number changed, changes in Table 1 and Table 2a/b) and adapted to a new template.
- Ligation sites of the probes targeting the *BAP1* gene updated according to new version of the NM_ reference sequence.
- For uniformity, the chromosomal locations and bands in this document are now all based on hg18 (NCBI36).
- Related probemixes section updated on page 7.
- Selected publications list updated on page 7-8.

Version 07 – 24 April 2019 (T08)
- New references added on page 2.
- Small changes of probe lengths in Table 1 and 2 in order to better reflect the true lengths of the amplification products.
- Minor textual changes throughout the document.

Version 06 – 23 December 2016 (T08)
- Product description adapted to a new lot (lot number added, small changes in Table 1 and Table 2, new picture included).
- New references added on page 2.
- Various minor textual changes on page 1.
- Various minor layout changes.
- Small changes of probe lengths in Table 1 and 2 in order to better reflect the true lengths of the amplification products.

Version 05 – 08 January 2016 (T08)
- Product description adapted to a new lot (lot number added, small changes in Table 1 and Table 2, new picture included).
- Related SALSA MLPA probemix added on page 1.
- New references added on page 2.
- MV locations adjusted for several probes in Table 1.
- Ligation sites updated for BAP1 probes according to NM_004656.3 in Table 2b.
- Various minor textual changes.

Version 04 – 7 May 2015 (54)
- New references added on page 2.
- Electropherogram picture using the old MLPA buffer removed.

Version 03 (48)
- Electropherogram pictures using the new MLPA buffer (introduced in December 2012) added.

Version 02 (48)
- Product description adapted to a new lot (lot number added, small changes in Table 1 and Table 2, new

picture included). - Various minor textual changes on the whole document. - Data analysis method has been modified. *Version 01 (46)* Not applicable, new document.

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