Product Description
SALSA® MLPA® Probemix P116-B2 SGC

To be used with the MLPA General Protocol.

Version B2
For complete product history see page 9.

Catalogue numbers:
- **P116-025R:** SALSA MLPA Probemix P116 SGC, 25 reactions.
- **P116-050R:** SALSA MLPA Probemix P116 SGC, 50 reactions.
- **P116-100R:** SALSA MLPA Probemix P116 SGC, 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 P116 SGC is a **research use only (RUO)** assay for the detection of deletions or duplications in the *SGCA*, *SGCB*, *SGCD*, *SGCG* and *FKRP* genes, which are associated with Limb-Girdle Muscular Dystrophy. This probemix can also be used to detect the presence of the *FKRP* L276I point mutation.

Limb-Girdle Muscular Dystrophy (LGMD) is characterised by loss of muscle bulk and strength in patients. The distal muscles are affected late in LGMD, if affected at all. LGMD is typically an inherited disorder, though it may be inherited as a dominant, recessive or X-linked genetic defect. The muscle cells of patients with LGMD cannot properly form the proteins needed for normal muscle function. Defects of different proteins are involved in LGMD, each related to a specific type of muscular dystrophy.

Autosomal recessive LGMD is a genetically heterogeneous disorder. Of the many genes that can result in this disorder, the following genes are present in the P116 SGC probemix:

Gene	Number of exons	Number of probes	Length	Location	Distance from p-telomere	LGMD type
SGCA	10 exons	11 ¬	9.9 kb	17q21.33	~46 Mb	LGMD2D
SGCB	6 exons	6	17.6 kb	4q12	~52 Mb	LGMD2E
SGCD	9 exons	9	441 kb	5q33.3	~156 Mb	LGMD2F
SGCG	8 exons	8	144 kb	13q12.12	~23 Mb	LGMD2C
FKRP	4 exons	5 §	12.5 kb	19q13	~52 Mb	LGMD2I

¬ This includes a flanking probe located downstream of the *SGCA* gene.
§ This includes a probe specific for the *FKRP* L276I mutation. This probe will only generate a signal when the mutation is present.

More information is available at https://www.ncbi.nlm.nih.gov/books/NBK1408/.

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 *SGCA* exon numbering used in this P116-B2 SGC product description is the exon numbering from the LRG_203 sequence. The *SGCB* exon numbering is the exon numbering from the LRG_204 sequence. The *SGCD* exon numbering is the exon numbering from the LRG_205 sequence. The *SGCG* exon numbering is the exon numbering from the LRG_207 sequence. The *FKRP* exon numbering is the exon numbering from the LRG_761 sequence. The exon numbering of the NM_ sequence that was used for determining a probe's ligation site does not always correspond to the exon numbering obtained from the LRG sequences. 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.

Probemix content
The SALSA MLPA Probemix P116-B2 SGC contains 48 MLPA probes with amplification products between 130 and 492 nucleotides (nt). This includes ten probes for the *SGCA* gene (one for each exon), one flanking probe downstream of *SGCA*, six probes for the *SGCB* gene (one for each exon), nine probes for the *SGCD* gene (one for each exon), eight probes for the *SGCG* gene (one for each exon) and four probes for the *FKRP* gene (one for each exon). Furthermore, this probemix contains a probe specific for the *FKRP* L276I mutation which will only generate a signal when the mutation is present. In addition, nine reference probes are included that detect autosomal chromosomal locations. Complete probe sequences and the identity of the genes detected by the reference probes 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).

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, 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.

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 LGMD. 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
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/) have diverse collections of biological resources which may be used as positive control DNA samples in your MLPA experiments. The quality of cell lines can change; therefore samples should be validated before use.

SALSA Binning DNA SD030
The SD030 Binning DNA provided with this probemix can be used for binning of all probes including the mutation-specific probe 11373-L13479 (*FKRP* L276I mutation). SD030 Binning DNA is a mixture of genomic DNA from healthy individuals and plasmid DNA that contains the target sequence detected by the above mentioned probe. Inclusion of one reaction with 5 µl SD030 Binning DNA in initial MLPA experiments is essential as it can be used to aid in data binning of the peak pattern using Coffalyser.Net software. Furthermore, Binning DNA should be included in the experiment whenever changes have been applied to the set-up of the capillary electrophoresis device (e.g. when capillaries have been renewed). Binning DNA should never be used as a reference sample in the MLPA data analysis, neither should it be used in quantification of mutation signal(s). It is strongly advised that all samples tested are extracted with the same method and derived from the same source of tissue. For further details, please consult the SD030 Binning DNA product description, available online: www.mrcholland.com. **This product is for research use only (RUO).**

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 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	1.30 < FR < 1.65
Heterozygous triplication/homozygous duplication	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.

- Arranging probes according to chromosomal location facilitates interpretation of the results and may reveal more subtle changes such as those observed in mosaic cases. Analysis 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. 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 or in or near the *FKRP* gene. 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.

Limitations of the procedure

- In most populations, the major cause of genetic defects in the LGMD-related genes are small (point) mutations, most of which will not be detected by using SALSA MLPA Probemix P116 SGC.
- 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.

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.

SGCA, _SGCB_, _SGCD_, _SGCG_ and _FKRP_ mutation databases

https://databases.lovd.nl/shared/genes/SGCA
https://databases.lovd.nl/shared/genes/SGCB
https://databases.lovd.nl/shared/genes/SGCD
https://databases.lovd.nl/shared/genes/SGCG
https://databases.lovd.nl/shared/genes/FKRP

We strongly encourage users to deposit positive results in the Leiden Open Variation Database. 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 SGCA exons 6 and 8 but not exon 7) to MRC Holland: info@mrcholland.com.

Table 1. SALSA MLPA Probemix P116-B2 SGC

Length (nt)	SALSA MLPA probe	Chromosomal position (hg18)[a]					
		Reference	*SGCA*	*SGCB*	*SGCD*	*SGCG*	*FKRP*
64-105	Control fragments – see table in probemix content section for more information						
130	Reference probe 00797-L13645	5q					
136	**SGCA probe** 11367-L12092		**Exon 8**				
142	**SGCG probe** 03387-L02780					**Exon 1**	
148	**SGCA probe** 03372-L02765		**Exon 2**				
154	Reference probe 15163-L16938	3q					
160	**SGCB probe** 04611-L02767			**Exon 3**			
172	**SGCA probe** 03373-L13241		**Exon 9**				
178	**SGCG probe** 03388-L20658					**Exon 2**	
184	**SGCB probe** 03375-L20659			**Exon 5**			
190	**SGCB probe** 11368-L12093			**Exon 6**			
196	**SGCB probe** 17268-L20690			**Exon 1**			
202	**SGCD probe** 03376-L12512				**Exon 1**		
208	**SGCD probe** 11369-L12094				**Exon 2**		
214	Reference probe 08940-L09035	11p					
222 «	**FKRP probe** 11370-L20320						**Exon 2**
228	**SGCG probe** 03390-L02783					**Exon 4**	
232	**SGCD probe** 11371-L13242				**Exon 8**		
238	**SGCD probe** 03377-L02770				**Exon 3**		
245	Reference probe 08677-L08689	13q					
252	**SGCA probe** 11372-L12899		**Exon 6**				
259 § «	**FKRP probe** 11373-L13479						**L276I**
265	**SGCG probe** 03391-L12902					**Exon 5**	
274	**SGCD probe** 03378-L02771				**Exon 4**		
280 ¥	**SGCD probe** 21678-L31529				**Exon 6**		
285	Reference probe 05387-L21105	12p					
292	**SGCA probe** 11374-L12099		**Exon 1**				
301 ±	**SGCG probe** 03392-L02785					**Exon 6**	
310	**SGCD probe** 03379-L02772				**Exon 5**		
320	**SGCA probe** 11375-L12901		**Exon 4**				
328	Reference probe 09571-L10025	22q					
337	**SGCG probe** 03393-L13243					**Exon 7**	
343	**SGCB probe** 11376-L12101			**Exon 2**			
355	**SGCA probe** 11377-L12102		**Exon 5**				
363 ¬	COL1A1 probe 07983-L07764		down-stream				
373	**SGCG probe** 03394-L02787					**Exon 8**	
381	**SGCD probe** 03380-L04677				**Exon 7**		
388	**SGCD probe** 03381-L04694				**Exon 9**		
396	**SGCA probe** 11378-L20660		**Exon 10**				
403	Reference probe 04960-L20661	1p					
409 «	**FKRP probe** 11379-L12104						**Exon 3**
418 «	**FKRP probe** 11380-L12105						**Exon 4**
427	**SGCA probe** 11381-L12106		**Exon 7**				
432 ¥	**SGCB probe** 21891-L12107			**Exon 4**			
445	Reference probe 10370-L09644	6q					
454	**SGCA probe** 11383-L12108		**Exon 3**				
471 «	**FKRP probe** 11384-L21239						**Exon 1**
481	**SGCG probe** 11620-L20322					**Exon 3**	
492 *	Reference probe 18547-L24044	9q					

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

* New in version B2.
¥ Changed in version B2. Minor alteration, no change in sequence detected.
§ Mutation-specific probe. This probe will only generate a signal when the L276I mutation is present. It has been tested on artificial DNA **but not on positive human samples!**
± SNP rs114160429 could influence the probe signal. In case of apparent deletions, it is recommended to sequence the region targeted by this probe.
« 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.
¬ 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.

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. P116-B2 probes arranged according to chromosomal location

Table 2a. *SGCA* gene

Length (nt)	SALSA MLPA probe	*SGCA* exon[a]	Ligation site NM_000023.4	Partial sequence[b] (24 nt adjacent to ligation site)	Distance to next probe
		start codon	*37-39 (Exon 1)*		
292	11374-L12099	Exon 1	60-61	CTCTTCTGGACT-CCTCTCCTCGTG	1.4 kb
148	03372-L02765	Exon 2	155-156	TGTGCACACCTT-GGACCATGAGAC	0.3 kb
454	11383-L12108	Exon 3	323-324	TGCCACCCCAGA-AGATCGTGGGCT	0.3 kb
320	11375-L12901	Exon 4	377-378	GGACAGCTTTGA-TACCACTCGGCA	0.5 kb
355	11377-L12102	Exon 5	492-493	CTGCCCTCAACA-CCTGCCAGCCGC	0.8 kb
252	11372-L12899	Exon 6	739-740	CTTGCTACGACA-CCTTGGCACCCC	1.1 kb
427	11381-L12106	Exon 7	902-903	CTTCTTGGTGGA-TGCTCTGGTCAC	0.4 kb
136	11367-L12092	Exon 8	1011-1012	AGAGACCTGGCT-ACCTCCGAGTGA	4.7 kb
172	03373-L13241	Exon 9	1129-1130	CCATGTTCAATG-TGCACACAGGTG	0.4 kb
396	11378-L20660	Exon 10	1239-1238 *reverse*	GAGAAGGGAGGA-TGAAGTCAGGGC	9.3 kb
		stop codon	*1198-1200 (Exon 9)*		
363 ¬	07983-L07764	*COL1A1* gene		AAGACACAGGAA-ACAATGTATTGT	

Table 2b. *SGCB* gene

Length (nt)	SALSA MLPA probe	*SGCB* exon[a]	Ligation site NM_000232.5	Partial sequence[b] (24 nt adjacent to ligation site)	Distance to next probe
		start codon	*41-43 (Exon 1)*		
196	17268-L20690	Exon 1	183 nt before exon 1 *reverse*	GGCGCGTTGTAT-TGCACAGGGGCC	5.0 kb
343	11376-L12101	Exon 2	180-181	ATACATTCCGAT-TGATGAAGATCG	3.8 kb
160	04611-L02767	Exon 3	376-377	CGATTTAAGCAA-GTATCTGACATG	0.9 kb
432	21891-L12107	Exon 4	513-514	TGTAGAAAACAA-CAAAACTTCTAT	0.9 kb
184	03375-L20659	Exon 5	744-745	TGTATTCATTAT-GGGCAAAACCAT	4.0 kb
190	11368-L12093	Exon 6	928-929	GGGACGCTCTTC-AAGGTGCAAGTA	
		stop codon	*995-997 (Exon 6)*		

Table 2c. *SGCD* gene

Length (nt)	SALSA MLPA probe	*SGCD* exon[a]	Ligation site NM_000337.6	Partial sequence[b] (24 nt adjacent to ligation site)	Distance to next probe
		start codon	*113-115 (Exon 2)*		
202	03376-L12512	Exon 1	58 nt before exon 1	CTGACTGGGGCA-GCTTCTGAGCGC	2.5 kb
208	11369-L12094	Exon 2	1 nt after exon 2	AGGTGGAGATGG-TGAGTAATTCCC	15.0 kb
238	03377-L02770	Exon 3	157-158	AGCACCATGCCT-GGCTCTGTGGGG	164 kb
274	03378-L02771	Exon 4	397-398	AAAGAAATCCAG-TCCCGACCAGTA	80.6 kb
310	03379-L02772	Exon 5	439-440	TCTGCCAGAAAT-GTTACAGTGAAC	5.7 kb
280	21678-L31529	Exon 6	579-578 *reverse*	CTACTACCACTT-CATTATTGTCTG	52.5 kb
381	03380-L04677	Exon 7	646-647	CCTAAATCTATA-GAAACACCTAAT	110 kb
232	11371-L13242	Exon 8	759-760	AGAAGCTGGCAA-TATGGAAGCCAC	1.6 kb
388	03381-L04694	Exon 9	895-896	CAGAAGGTCTTC-GAGATCTGCGTC	
		stop codon	*983-985 (Exon 9)*		

Table 2d. *SGCG* gene

Length (nt)	SALSA MLPA probe	*SGCG* exon[a]	Ligation site NM_000231.3	Partial sequence[b] (24 nt adjacent to ligation site)	Distance to next probe
		start codon	*98-100 (Exon 2)*		
142	03387-L02780	Exon 1	51-52	TGGTAGAGCTCG-GGCCAGCTGTAG	22.8 kb
178	03388-L20658	Exon 2	215-216	TCTACTTGTTTG-TTCTTCTTTTAC	30.9 kb
481	11620-L20322	Exon 3	346-347	TTGGAAGGGGAA-TCAGAATTTTTA	16.0 kb
228	03390-L02783	Exon 4	439-440	GTGACTGTAAAT-GCGCGCAACTCA	28.7 kb
265	03391-L12902	Exon 5	530-531	AGATCAACTCCA-ACGACGGCAAGC	16.0 kb
301 ±	03392-L02785	Exon 6	631-632	TTTGAACATTCA-GTGGAGACACCC	25.2 kb
337	03393-L13243	Exon 7	705-706	GAGTCTAAGCAT-GGATGCCCCAAG	3.7 kb
373	03394-L02787	Exon 8	840-841	ACCCAAGCTGGT-GCAGGGGACGTG	
		stop codon	*971-973 (Exon 8)*		

Table 2e. *FKRP* gene

Length (nt)	SALSA MLPA probe	*FKRP* exon[a]	Ligation site NM_024301.5	Partial sequence[b] (24 nt adjacent to ligation site)	Distance to next probe
		start codon	*287-289 (Exon 4)*		
471 «	11384-L21239	Exon 1	128 nt after exon 1	TCGTGCTGGATA-AAGTGCAGGATC	1.8 kb
222 «	11370-L20320	Exon 2	71-72	TGCCCTCCTGGA-ACTCCCCCAGCC	0.5 kb
409 «	11379-L12104	Exon 3	128-127 *reverse*	CTGGGTCTGAGT-TGCGATTTGGCC	7.7 kb
259 § «	11373-L13479	Exon 4	1112-1111 *reverse*	CCAGCTCACTAT-GCGGATGCCCAG	0.9 kb
418 «	11380-L12105	Exon 4	1981-1982	CCAGATTTATCA-AATGGTCATGCC	
		stop codon	*1772-1774 (Exon 4)*		

[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.

§ Mutation-specific probe. This probe will only generate a signal when the L276I mutation is present. It has been tested on artificial DNA **but not on positive human samples!**
± SNP rs114160429 could influence the probe signal. In case of apparent deletions, it is recommended to sequence the region targeted by this probe.
« 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.
¬ 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.

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.

Related SALSA MLPA probemixes

- P034/P035 DMD: Contains probes for *DMD*, involved in Duchenne and Becker muscular dystrophy.
- P048 LMNA/MYOT: Contains probes for *LMNA*, *MYOT* and *CAV3*.
- P061 Lissencephaly: Contains probes for *POMT1* and *POMGNT1,* involved in LGMD2K and LGMD2O.
- P176 CAPN3: Contains probes for *CAPN3,* involved in LGMD2A.
- P268 DYSF: Contains probes for *DYSF,* involved in LGMD2B.
- P436 ANO5: Contains probes for *ANO5*, involved in LGMD2L.

References

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- Ten Dam L et al. (2020). Elucidation of the Genetic Cause in Dutch Limb Girdle Muscular Dystrophy Families: A 27-Year's Journey. *J Neuromuscul Dis*. 1-12.
- Marinakis NM et al. (2021). Phenotype-driven variant filtration strategy in exome sequencing toward a high diagnostic yield and identification of 85 novel variants in 400 patients with rare Mendelian disorders. *Am J Med Genet A*.
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P116 product history	
Version	*Modification*
B2	One reference probe has been replaced and several probe lengths have been adjusted.
B1	One extra probe each for *SGCB*, *SGCD* and *FKRP* have been included. Five reference probes and the 88 nt and 96 nt control fragments have been replaced.
A1	First release.

Implemented changes in the product description
Version B2-02 – 12 January 2022 (04P) - Product description rewritten and adapted to a new template. - Ligation sites of the probes targeting the *SGCA*, *SGCB*, *SCGD*, *SGCG* and *FKRP* genes updated according to new versions of the NM_ reference sequences. - Small changes of probe lengths in Table 1 and 2 in order to better reflect the true lengths of the amplification products. *Version B2-01 – 10 January 2019 (01P)* - Product description restructured and adapted to a new template. - Product description adapted to a new product version (version number changed, changes in Table 1 and Table 2, new picture included).

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