

Product Description SALSA® MLPA® Probemix P137-C1 SCN1A

To be used with the MLPA General Protocol.

Version C1. As compared to version B3, probes for SCN1A exon 3 and 15 and two reference probes have been replaced. For complete product history see page 9.

Catalogue numbers:

- **P137-025R:** SALSA MLPA Probemix P137 SCN1A, 25 reactions.
- **P137-050R:** SALSA MLPA Probemix P137 SCN1A, 50 reactions.
- **P137-100R:** SALSA MLPA Probemix P137 SCN1A, 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.

Intended use: The SALSA MLPA Probemix P137 SCN1A is an in vitro diagnostic (IVD)¹ or research use only (RUO) semi-quantitative assay² for the detection of deletions or duplications in *SCN1A* gene in genomic DNA isolated from human peripheral whole blood specimens. P137 SCN1A is intended to confirm a potential cause for and clinical diagnosis of Dravet syndrome and other *SCN1A*-related seizure disorders, and for Familial hemiplegic migraine 3 (FHM3).

Copy number variations (CNVs) detected with P137 SCN1A should be confirmed with a different technique. In particular, CNVs detected by only a single probe always require confirmation by another method. Most defects in the *SCN1A* gene are point mutations, none of which will be detected by MLPA. It is therefore recommended to use this assay in combination with sequence analysis.

Assay results are intended to be used in conjunction with other clinical and diagnostic findings, consistent with professional standards of practice, including confirmation by alternative methods, parental evaluation, clinical genetic evaluation, and counselling, as appropriate. The results of this test should be interpreted by a clinical molecular geneticist or equivalent.

This device is not intended to be used for standalone diagnostic purposes, pre-implantation or prenatal testing, population screening, or for the detection of, or screening for, acquired or somatic genetic aberrations, e.g. from DNA extracted from formalin-fixed paraffin embedded (FFPE) or fresh tumour materials.

¹Please note that this probemix is for In Vitro Diagnostic (IVD) use in the countries specified at the end of this product description. In all other countries, the product is for Research Use Only (RUO).

²To be used in combination with a SALSA MLPA Reagent Kit and Coffalyser.Net analysis software.

Clinical background: *SCN1A*-related seizure disorders include at the severe end of the spectrum Dravet syndrome (DS) and intractable childhood epilepsy with generalized tonic-clonic seizures (ICE-GTC), and at the mild end simple febrile seizures (FS) and generalized epilepsy with febrile seizures plus (GEFS+). Myoclonic-astatic epilepsy (MAE or Doose syndrome), Lennox-Gastaut syndrome (LGS), infantile spasms, and vaccine-related encephalopathy and seizures are the less commonly observed phenotypes.

SCN1A-related seizure disorders are inherited in an autosomal dominant manner. Pathogenic variants may be inherited or *de novo*. The percentage of cases with *de novo* mutations increases as the severity of the phenotype increases. The *SCN1A*-related seizures phenotype varies even among family members with the same pathogenic variant.

Dravet syndrome (DS; OMIM # 607208, also known as severe myoclonic epilepsy in infancy (SMEI), early infantile epileptic encephalopathy 6 (EIEE6), and polymorphic myoclonic epilepsy in infancy (PMEI)) is a rare lifelong form of epilepsy that begins in the first year of life. DS is associated with heterozygous mutations in the *SCN1A* gene. The frequency of *SCN1A* mutations in DS patients is approximately 70-80%, most of the mutations being *de novo* (Marini et al. 2009). Apparent *de novo* sporadic mutations may also be the result of germline mosaicism in apparently unaffected parents (de Lange et al. 2018). Somatic mosaic deletions of the *SCN1A* gene have also been identified in individuals with DS (Nakayama et al. 2018). DS is defined by prolonged and frequent seizures that do not remit, and normally evolve to include myoclonic seizures. Other symptoms of DS are behavioural and developmental delay, movement and balance problems, hyperactivity and sleep difficulties.

Intractable childhood epilepsy with generalized tonic-clonic seizures (ICE-GTC) is characterized by generalized seizures including absence seizures and generalized tonic-clonic seizures with onset in infancy or childhood. Children with frequent generalized tonic-clonic seizures often develop cognitive impairment. Febrile seizures (FS) are characterized by childhood seizures that occur only in association with fever (higher than 38°C). The age of onset is 6 months, and the seizures resolve by the age of 5 years old. Generalized epilepsy with febrile seizures plus (GEFS+) refers to findings in a family instead of an individual. The seizure phenotypes tend toward the mild end of the spectrum. Affected individuals within a family with GEFS+ often have febrile seizures in early childhood, followed by occasional tonic, clonic, myoclonic, or absence seizures which respond to medication and remit by late childhood or early adolescence.

More information is available at <https://www.ncbi.nlm.nih.gov/books/NBK1318/>.

Familial hemiplegic migraine (FHM) is a rare autosomal dominant subtype of migraine with aura. Familial hemiplegic migraine type 3 (FHM3) is caused by mutations in the *SCN1A* gene. More information is available at <https://www.ncbi.nlm.nih.gov/books/NBK1388/>.

Gene structure: The *SCN1A* gene spans ~139 kb on chromosome 2q24.3. The *SCN1A* LRG_8 is identical to the NCBI NG_011906.1 sequence. The LRG sequence is available at <http://www.lrg-sequence.org/>.

Transcript variants: *SCN1A* has multiple transcript variants. The NM_006920.6 sequence (coding sequence 479-6475; <https://www.ncbi.nlm.nih.gov/gene/6323>) represents transcript variant 2 of the *SCN1A* gene. The NM_001202435.3 sequence (coding sequence 257-6286) represents the transcript variant 4 of the *SCN1A* gene, which contains alternate 5' untranslated exons.

Exon numbering: The exon numbering used in this P137-C1 *SCN1A* product description is based on the exon numbering of the LRG_8, which uses the RefSeq transcript NM_06920.4, and has 26 exons. NM_006920.6 is available at NCBI, and has the same 26 coding exons and three additional non-coding exons at the beginning of the *SCN1A* gene. MRC-Holland follows the LRG sequence for exon numbering, but NM_006920.6 to indicate probe ligation sites in Table 2. The exon numbering and NM sequence used are from 10/2019.

Probemix content: The SALSA MLPA Probemix P137-C1 *SCN1A* contains 40 MLPA probes with amplification products between 142 and 454 nucleotides (nt). This includes 30 probes for the *SCN1A* gene, covering all 26 coding exons of *SCN1A*, two probes for the first non-coding exon of NM_006920.6 (exon hB), and two probes for the upstream region of *SCN1A* (exon hA). In addition, ten 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.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).

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 from peripheral blood, 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 unrelated individuals who are from families without a history of *SCN1A*-Related Seizure Disorders or FHM3. 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 NA10401 and NA10607 from the Coriell Institute have been tested at MRC-Holland and can be used as positive control samples to detect a duplication and a heterozygous deletion of the complete *SCN1A* gene, respectively. The quality of cell lines can change; therefore samples should be validated before use.

Performance characteristics: Approximately 2-3% of all mutations in Dravet syndrome are expected to be deletions/duplications (Marini et al. 2009), which can be detected with the P137-C1 probemix. In *SCN1A*-point mutation negative patients this percentage is approximately 16% (Madia et al. 2006, Wang et al. 2008, <https://www.ncbi.nlm.nih.gov/books/NBK1318/>). The percentage of deletions/duplications for other *SCN1A*-related seizures disorders and FHM3 is unknown. However, the association between these diseases and the *SCN1A* gene is well established. Analytical performance for the detection of deletions/duplications in the *SCN1A* gene is very high and can be considered >99% (based on a 2006-2019 literature review).

Analytical performance can be compromised by: SNPs or other polymorphisms (e.g. indels) in the DNA target sequence, impurities in the DNA sample, incomplete DNA denaturation, the use of insufficient or too much sample DNA, the use of insufficient or unsuitable reference samples, problems with capillary electrophoresis or a poor data normalisation procedure and other technical errors. The MLPA General Protocol contains technical guidelines and information on data evaluation/normalisation.

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 expected results for the *SCN1A* specific MLPA probes are allele copy numbers of 2 (normal), 1 (heterozygous deletion), or 3 (heterozygous duplication). Copy numbers of 4 (heterozygous triplication/homozygous duplication) or 0 (homozygous deletion) may occur, but are extremely rare.

The standard deviation of each individual probe over all the reference samples should be ≤ 0.10 and the dosage quotient (DQ) 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 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

- 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. 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.

Notes *SCN1A* results

- According to the updated NM_006920.6 sequence, three non-coding exons are described in the upstream region of the *SCN1A* gene. The P137-C1 probemix has two probes termed as exon hB, which correspond to exon 1 of NM_006920.6, and two probes termed as exon hA targeting the upstream region. The clinical significance of mutations/copy number changes in these non-coding exons is not fully known. However, microdeletions in exons hA and hB have been found in two DS patients, and a point mutation in exon hA has been described in a patient with partial epilepsy with antecedent febrile seizures (Gao et al. 2017, Nakayama et al. 2010).

- The *SCN1A* gene is located in a complicated region, since several highly homologous genes are present in this region (*SCN2A*, *SCN3A*, *SCN7A* and *SCN9A*). In rare cases, apparent duplications might therefore be due to sequence changes in the other similar genes.
- Mosaicism has been reported in individuals with DS (de Lange et al. 2018, Nakayama et al. 2018). Mosaic *SCN1A* deletions obtained with the P137 *SCN1A* probemix must be confirmed by analysis of a second, independently collected DNA sample or a different technique, in order to exclude a false positive mosaic result.

Limitations of the procedure:

- In most populations, the major cause of genetic defects in the *SCN1A* gene are small (point) mutations, most of which will not be detected by using SALSA MLPA Probemix P137 *SCN1A*.
- 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.

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.

***SCN1A* mutation databases:** LOVD Database (<https://databases.lovd.nl/shared/genes/SCN1A>) and *SCN1A* database (<http://www.caae.org.cn/gzneurosci/scn1adatabase/index.php>). We strongly encourage users to deposit positive results in the above mentioned 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 SNPs and unusual results (e.g., a duplication of *SCN1A* exons 6 and 8 but not exon 7) to MRC-Holland: info@mlpa.com.

Table 1. SALSA MLPA Probemix P137-C1 SCN1A

Length (nt)	SALSA MLPA probe	Chromosomal position (hg18) ^a	
		Reference	SCN1A
64-105	Control fragments – see table in probemix content section for more information		
142	Reference probe 09258-L11422	7q22	
149	SCN1A probe 04540-L03929		Exon 19
154	SCN1A probe 04531-L05030		Exon 10
160	SCN1A probe 04523-L18517		Exon 1
166	SCN1A probe 04524-L03913		Exon 2
172 ±	SCN1A probe 04541-L03930		Exon 20
178	SCN1A probe 04532-L03921		Exon 11
183	SCN1A probe 15940-L18066		Exon hA
190	Reference probe 06743-L06347	8q12	
196 *	SCN1A probe 22541-L31721		Exon 3
202	SCN1A probe 04542-L03931		Exon 21
211	SCN1A probe 04533-L03922		Exon 12
221	Reference probe 07223-L21127	3p14	
229	SCN1A probe 04526-L03915		Exon 4
236	SCN1A probe 15941-L18067		Exon hA
243	SCN1A probe 04543-L18518		Exon 22
250	SCN1A probe 04534-L30856		Exon 13
256 *	Reference probe 10808-L27953	4q25	
265	SCN1A probe 15942-L18068		Exon hB
274	SCN1A probe 04527-L04899		Exon 5
280	SCN1A probe 04544-L04900		Exon 23
292	SCN1A probe 04535-L03924		Exon 14
301	Reference probe 08313-L08182	11q22	
310	SCN1A probe 04528-L03917		Exon 6
319	SCN1A probe 04545-L18535		Exon 24
328 *	SCN1A probe 22542-L31769		Exon 15
337	SCN1A probe 07367-L07014		Exon 9
346 *	Reference probe 05982-L05407	20p12	
355	SCN1A probe 04529-L03918		Exon 7
364	SCN1A probe 17380-L04745		Exon 25
373	SCN1A probe 04537-L03926		Exon 16
382	Reference probe 04278-L23577	12q12	
391	SCN1A probe 04530-L03919		Exon 8
400	SCN1A probe 04538-L03927		Exon 17
409	SCN1A probe 15943-L18069		Exon hB
418	Reference probe 11057-L11726	14q31	
427	SCN1A probe 04539-L03928		Exon 18
436	Reference probe 14775-L16472	1q23	
445	SCN1A probe 04547-L18519		Exon 26
454	Reference probe 14954-L16687	6q22	

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

* New in version C1.

± SNP rs149579028 influences probe signal. In case of apparent deletions, it is recommended to sequence the region targeted by this probe.

Table 2. *SCN1A* probes arranged according to chromosomal location

Length (nt)	SALSA MLPA probe	SCN1A exon ^a	Ligation site NM_006920.6	Partial sequence ^b (24 nt adjacent to ligation site)	Distance to next probe
		<i>start codon</i>	479-481		
236 [⊖]	15941-L18067	Exon hA	14-15 in NM_001202435.3	GTGTAGGAGACA-CACTGCTGGCCT	0.1 kb
183 [⊖]	15940-L18066	Exon hA	108-107 reverse in NM_001202435.3	TTACCCCAAAA-GATGTAGTAAAA	21.1 kb
409 [⊖]	15943-L18069	Exon hB	37-38	CCGAGAGGATAC-TGCAGAGGTCTC	0.2 kb
265 [⊖]	15942-L18068	Exon hB	218-219	GATGCTGTTCCT-CACTGCAGATGG	54.3 kb
160	04523-L18517	Exon 1	565-566	GAAAGACGCATT-GCAGAAGAAAAG	14.9 kb
166	04524-L03913	Exon 2	779-780	AGGCCATCTTCC-GGTTCACTGCCA	2.2 kb
196 #	22541-L31721	Exon 3	916-915 reverse	GGGTTACTCATT-GTCATAAACACA	1.8 kb
229	04526-L03915	Exon 4	1037-1038	TTACTTTCCTTC-GGGATCCATGGA	1.7 kb
274	04527-L04899	Exon 5	1090-1091	AGGTACGTCACA-GAGTTTGTGGAC	1.0 kb
310 #	04528-L03917	Exon 6	1266-1267	CGTATTGCTCT-AATTGGGCTGCA	3.0 kb
355	04529-L03918	Exon 7	1470-1471	GGAGGGTTTTTT-AGATGCCTACT	1.2 kb
391	04530-L03919	Exon 8	1546-1547	AAAGCTGGTAGA-AATCCCAATTAT	1.0 kb
337	07367-L07014	Exon 9	1848-1849	GCAACAGGAGGC-AGCTCAGGTAAA	1.5 kb
154	04531-L05030	Exon 10	1903-1904	GAGCCAGTGCA-GCAGGCAGGCTC	1.4 kb
178	04532-L03921	Exon 11	2293-2294	GAGAGCCGTAGA-GATTCTTGTT	1.5 kb
211	04533-L03922	Exon 12	2517-2518	TGAAATGAGAAA-GAGAAGGTCAAG	1.0 kb
250	04534-L30856	Exon 13	2702-2703	ACTGTTCTCCAT-ATTGGTTAAAAG	1.9 kb
292	04535-L03924	Exon 14	2988-2989	GGTAGAACTTGG-ACTCGCCAATGT	1.4 kb
328	22542-L31769	Exon 15	3121-3120 reverse	ACGAGGGTTAAA-TTCCCAGAGCC	1.9 kb
373	04537-L03926	Exon 16	3795-3796	GACTGTACCAAT-TGCTGTAGGAGA	20.5 kb
400	04538-L03927	Exon 17	3948-3949	AGAACAGCCCGT-AGTGGAACCTGA	1.9 kb
427	04539-L03928	Exon 18	4079-4080	GAAGGACGTGTT-TCCGAATAGTTG	1.7 kb
149 #	04540-L03929	Exon 19	4274-4275	CATATGGCTATC-AAACATATTTCA	2.4 kb
172 ±	04541-L03930	Exon 20	4393-4394	AAATCTCTCAGG-ACACTAAGAGCT	7.2 kb
202	04542-L03931	Exon 21	4588-4589	AACACCACAACCT-GGTGACAGGTTT	2.9 kb
243	04543-L18518	Exon 22	4737-4738	ATAGGCCACATT-CAAGGATGGAT	1.6 kb
280	04544-L04900	Exon 23	4825-4826	CTGTACATGTAT-CTTTACTTTGTT	2.1 kb
319	04545-L18535	Exon 24	5005-5006	AAAAAACCGCAA-AAGCCTATACCT	1.8 kb
364	17380-L04745	Exon 25	5178-5179	ACGCATCAATCT-GGTGTTCAATTGT	2.5 kb
445	04547-L18519	Exon 26	5970-5971	TCTCAATCTGCC-ACAACCAAACAA	
		<i>stop codon</i>	6473-6475		

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.

⊖ The significance of deletions/duplications of exons hA and hB is not clear as these exons are non-coding. However, microdeletions in exons hA and hB have been found in two DS patients, and a point mutation in exon hA has been described in a patient with partial epilepsy with antecedent febrile seizures (Gao et al. 2017, Nakayama et al. 2010).

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.

± SNP rs149579028 influences probe signal. In case of apparent deletions, it is recommended to sequence the region targeted by this probe.

Related SALSA MLPA probemixes

- P138 SLC2A1-STXBP1 *GLUT1* deficiency syndrome, *STXBP1*-Encephalopathy with epilepsy, Ohtahara syndrome, contains probes for the *SLC2A1* and *STXBP1* genes.
- P166 KCNQ2 Benign familial neonatal convulsions, contains probes for the *KCNQ2* gene.
- P197 KCNQ3 Familial neonatal convulsions type 2, Epilepsy benign neonatal type 2, contains probes for the *KCNQ3* gene.
- P267 Dandy-Walker Dandy-Walker malformations, contains probes for the *ZIC1*, *ZIC4* and *VLDLR* genes.
- P408 ADLTE-LGI1 Autosomal dominant lateral temporal lobe epilepsy, contains probes for *LGI1*, *ADAM22*, *GPR98*, *KCNA1*, *KCNA4*, *KCNAB1* and *PDYN*.

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P137 Product history

Version	Modification
C1	Probes for SCN1A exon 3 and 15 and two reference probes have been replaced.
B3	Two reference probes have been replaced and two reference probes have been removed. In addition, several probe lengths have been adjusted.
B2	The 88 and 96 control fragments have been replaced (QDX2).
B1	Four probes for two new upstream exons have been included. In addition, several reference probes have been replaced.
A2	DNA denaturation control fragments (D-fragments) and X-, and Y- specific control fragments have been included.
A1	A probe for <i>SCN1A</i> exon 9 has been added. One control probe has been replaced.
A	First release.

Implemented changes in the product description

Version C1-01 – 19 November 2019 (02P)

- Product description rewritten and adapted to a new template.
- Product description adapted to a new product version (version number changed, changes in Table 1 and Table 2).
- Ligation sites of the probes targeting the *SCN1A* gene updated according to new versions of the NM reference sequences.
- P137-C1 is now CE marked.

Version B3-03 – 09 July 2019 (01P)

- Warning in Table to and 2 for More variable probe was updated. Aberrant results should be treated with

<p>caution was replaced by Results should be treated with caution.</p> <p><i>Version B3-02 – 18 January 2019 (01P)</i></p> <ul style="list-style-type: none"> - The sections general information and positive control DNA samples were updated. - Ligation sites in Table 2 adjusted according to the updated NM sequence. - Warning added to Table 1 and 2 for SNP rs149579028 that influences the probe signal: 172 nt probe 04541-L03930. - Warnings added for probe specificity relying on a single nucleotide difference between target gene and related gene or pseudogene to interpretation of results section and Table 2. - Warning added to Table 1 and Table 2 for more variable probe, 328 nt probe 04536-L03925. - Several references using the P137 probemix have been removed and added. - Various minor textual changes throughout the document. <p><i>Version B3-01 – 08 March 2018 (01P)</i></p> <ul style="list-style-type: none"> - 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). - Various minor textual or layout changes. <p><i>Version 15 – 11 January 2017 (55)</i></p> <ul style="list-style-type: none"> - Warning added in Table 1, 383 nt probe 03091-L07348. <p><i>Version 14 – 04 December 2015 (55)</i></p> <ul style="list-style-type: none"> - Product description adapted to a new lot (lot number changed, small changes in Table 1 and Table 2, new picture included). - References updated. - Small changes of probe lengths in Table 1 and 2 in order to better reflect the true lengths of the amplification products. <p><i>Version 13 – 22 July 2015 (54)</i></p> <ul style="list-style-type: none"> - Figure based on the use of old MLPA buffer (replaced in December 2012) removed. - Various minor textual changes throughout the document.
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*comprising EU (candidate) member states and members of the European Free Trade Association (EFTA).
The product is for RUO in all other European countries.