Product Description SALSA® Binning DNA SD016-S01

Version S01.

Catalogue number: SD016: SALSA® Binning DNA**,** six reactions

To be used with the following SALSA MLPA probemix: P282-A3 RYR1 mix 2 in combination with a SALSA® MLPA® reagent kit, available for various number of reactions. MLPA reagent kits are either provided with FAM or Cy5.0 dye-labelled PCR primer, suitable for Applied Biosystems and Beckman capillary sequencers, respectively (see www.mlpa.com).

Precautions and warnings: For professional use only. Always consult the most recent product description AND the corresponding probemix product description AND the MLPA General Protocol or the MS-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: This SD016 DNA can be used as Binning DNA sample for the MLPA probemix version as specified above and in Table 1. Binning and filtering are the processes of linking a signal to its probe identity by use of the probe length. The Binning DNA can also be used as an artificial positive control for the specific point mutations. See Table 1 and the corresponding probemix product description for more details on mutation-specific probe targets present.
Please note that this Binning DNA is a mixture of female genomic DNA from healthy individuals and artificial DNA of 50-80 nt length covering probe target sequences and not covering the whole exon.
This product is for research use only (RUO).

Experimental set up: MLPA reactions for binning purposes should be performed with 5 µl of Binning DNA, properly mixed. Inclusion of one reaction with SALSA Binning DNA SDX016 in the initial MLPA experiment is essential as it can 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 a different polymer type is used).

Data analysis: Coffalyser.Net software must be used for analysis of MLPA experiments. When performing the fragment analysis step in Coffalyser.Net, select SD016 in the *bin smpl* –column. By selecting the SD016 sample as your binning sample, probes will be correctly identified in the peak pattern across all patient samples. Coffalyser.Net software is available free of charge on www.mlpa.com.

Warning: 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), as for this purpose true mutation/SNP positive patient samples or cell lines should be used. It is strongly advised to use sample and reference DNA extracted with the same method and derived from the same source of tissue.

Binning DNA content: MRC-Holland is unable to provide mutation positive human DNA samples. As an alternative, we have prepared a mixture of female genomic DNA from healthy individuals and a titrated amount of plasmid DNA that contains the target sequences recognised by the mutation-specific probes present in the MLPA probemix version as specified above and in Table 1.

The plasmid DNA included in the SD016 DNA contains partial sequences of the RYR1 gene. These sequences include 15 different mutations which will be detected by MLPA probes that are present in the aforementioned probemix version (for details, see Table 1) and will generate mutation-specific signals for these probes.
Please note that the plasmid DNA contains the target sequences detected by the probes specified in Table 1 and the sequence of the 105 nt chromosome Y specific control fragment. The amount of plasmid in this Binning DNA (relative to the genomic DNA) results in a relative probe signal for the 105 nt probe on this female DNA which is similar to the relative probe signal obtained on male DNA samples. As a result, the 100 and 105 nt control fragments indicate the presence of two copies chromosome X and one copy chromosome Y.

Storage and stability: Upon arrival, Binning DNA must be stored between -25 °C and -15 °C, in the original packaging. When stored under the recommended conditions, a shelf life of at least 1 year is guaranteed, also after opening. The expiry date is mentioned on the label of the vial.

Table 1. Mutation-specific probe targets in SD016-S01 Binning DNA

Probemix	Gene/Exon	Probe length	Probe ID	Present in probemix version	Details
P282	RYR1 exon 6	142 nt	07563-L07263	A3	c.488G>T
	RYR1 exon 9	148 nt	07564-L17041	A3	c.742G>A
	RYR1 exon 17	286 nt	03665-L03137	A3	c.1841G>T
	RYR1 exon 39	292 nt	03668-L03140	A3	c.6487C>T
	RYR1 exon 39	154 nt	07566-L07266	A3	c.6488G>A
	RYR1 exon 40	214 nt	04054-L07267	A3	c.6617C>G
	RYR1 exon 43	164 nt	07568-L07269	A3	c.7025A>G
	RYR1 exon 44	171 nt	07569-L07270	A3	c.7063C>T
	RYR1 exon 44	189 nt	07570-L17043	A3	c.7124G>C
	RYR1 exon 45	300 nt	03671-L03144	A3	c.7300G>A
	RYR1 exon 46	310 nt	07740-L17046	A3	c.7361G>A
	RYR1 exon 98	178 nt	07571-L17042	A3	c.14209C>T
	RYR1 exon 100	196 nt	07572-L07273	A3	c.14471T>C
	RYR1 exon 101	202 nt	07574-L07275	A3	c.14545G>A
	RYR1 exon 104	220 nt	07575-L07276	A3	c.14918C>T

Note: Mutation nomenclature and exon numbering used here may differ from literature! Please notify us of any mistakes: info@mlpa.com. Please consult the respective probemix product description to find corresponding gene transcripts.

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

Implemented Changes – compared to the previous SD016 product description versions

Version S01-02 – 27 May 2017 (15)

- Product description adjusted to a new product description format. No changes in content of SD.
- Various textual changes.