Product Description SALSA® Binning DNA SD015-S01

Version S01.

Catalogue number: SD015: SALSA® Binning DNA**,** six reactions

To be used with the following SALSA MLPA probemix: P281-A3 RYR1, 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 SD015 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 SD015 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 SD015 in the *bin smpl* –column. By selecting the SD015 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 SD 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 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 SD015 DNA contains partial sequences of the RYR1 gene. These sequences include 18 different mutations which will be detected by MLPA probes that are present in the aforementioned probemix versions (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 above mentioned probes 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 target sequences in SD015 Binning DNA

Product	Gene/Exon	Probe length	Probe ID	Present in probemix version	Details
P281	RYR1 exon 6	142 nt	04052-L03453	A3	487C>T
	RYR1 exon 11	148 nt	04058-L03133	A3	1021G>A
	RYR1 exon 46	154 nt	04508-L03146	A3	7373G>A
	RYR1 exon 39	166 nt	03667-L03139	A3	6502G>A
	RYR1 exon 12	172 nt	04061-L03134	A3	1209C>G
	RYR1 exon 17	177 nt	03666-L03138	A3	1840C>T
	RYR1 exon 45	190 nt	03672-L03145	A3	7304G>A
	RYR1 exon 101	202 nt	03677-L03150	A3	14582G>A
	RYR1 exon 40	214 nt	04054-L03455	A3	6617C>T
	RYR1 exon 44	220 nt	03670-L03143	A3	7048G>A
	RYR1 exon 100	226 nt	04060-L03149	A3	14512C>G
	RYR1 exon 15	238 nt	04053-L03454	A3	1654C>T
	RYR1 exon 2	244 nt	04051-L03452	A3	103T>C
	RYR1 exon 102	250 nt	03678-L03151	A3	14693T>C
	RYR1 exon 14	262 nt	03663-L03135	A3	1565A>C
	RYR1 exon 100	265 nt	04056-L03457	A3	14387A>G
	RYR1 exon 43	274 nt	07567-L07268	A3	7007G>A
	RYR1 exon 100	284 nt	07573-L07274	A3	14477C>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 SD015 product description versions
Version S01-02 – 11 January 2018 (15)
- SD version and probemix version information adjusted on page 1, SD lot removed throughout document.
- Precautions and warnings and information about experimental set-up added to page 1.
- Information added to table 1.
- Mutation details for P281 214 nt probe adjusted in table 1.
- Various textual and layout changes.
Version 01 (07)
- Not applicable, new document.