

## **Product Description SALSA® Binning DNA SD033-S01**

Version S01.

Catalogue number: SD033: SALSA® Binning DNA, six reactions

To be used with the following SALSA MLPA probemix: P436-A1 ANO5, 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 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 SD033 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 mutation. See Table 1 and the corresponding probemix product description for more details on the mutation-specific probe target 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 the probe target sequence 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  $\mu$ l of Binning DNA, properly mixed. Inclusion of one reaction with SALSA Binning DNA SD033 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 SD033 in the *bin smpl*–column. By selecting the SD033 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 <a href="https://www.mlpa.com">www.mlpa.com</a>.

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, 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 synthetic DNA that contains the target sequence recognised by the mutation-specific probe present in the MLPA probemix version as specified above and in Table 1.

The synthetic DNA included in the SD033 DNA contains a partial sequence of the ANO5 gene. This sequence includes one mutation which will be detected by the MLPA probe that is present in the aforementioned probemix version (for details, see Table 1) and will generate a mutation-specific signal for this probe. Please note that the synthetic DNA contains the target sequence detected by the above mentioned probe and the sequence of the 105 nt chromosome Y specific control fragment. The amount of synthetic DNA 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 in SD033-S01 Binning DNA

Probemix	Gene/Exon	Probe length	Probe ID	Present in probemix version	Details
P436	ANO5 exon 5	198 nt	18658-SP0690- L24012	A1	c.191dupA

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

More information: www.mlpa.com; www.mlpa.eu		
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## Implemented Changes – compared to the previous SD033 product description versions

Version S01-02 - 04 October 2017 (15)

- Product description completely rewritten and restructured.
- Change in contact details.

Version 01 (01)

- Not applicable, new document.