

Instructions for use

Lambda Hind III-Marker

This marker was manufactured by using unmethylated Lambda-DNA and restriction endonuclease *Hind*III.

Classical standard for analysis of middlerange DNA fragments. Since the marker is produced by restriction digest of Lambda-DNA, fragments are mostly distributed in an equimolar way (with the exception of the 4361 bp fragment), additionally allowing rough quantification of the DNA analysed.

The following fragment sizes (in bp) are included:

23130 ¹⁾, 9416, 6557, 4361 ¹⁾, 2322, 2027, 564, 125

¹ Labelled fragments contain cohesive ends of bacteriophage Lambda which often hybridise, resulting in fragments/bands of higher molecular weight. We recommend heating the marker prior to use as described below, in order to separate those two fragments.

The whole Lambda phage genome contains 48.502 bps.

Content:

X910.1 - 2x 50 μg Lambda Hind III-Marker
0100.1 - 1x sample buffer ROTI[®]Load DNA (with Glycerol)

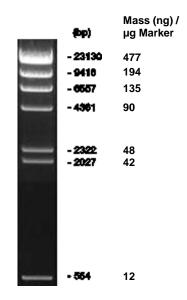


Fig.: 1 % agarose (NEEO, Art. No. 2267

Application:

Depending on the application, the marker can be dissolved directly in the supplied sterile-filtered 1 x sample buffer ROTI®Load DNA (with glycerol) (Art. No. 0100.1) or in TE- ROTI®Stock 100 x TE, Art. No. 1052.1).

The lyophilized DNA is dissolved for 15 minutes at room temperature in an appropriate volume (see below) under occasional stirring. Prior to use we recommend to dissolve the cohesive ends by incubating at 65 °C for 5 mins, followed by cooling at 4 °C or on ice.

This treatment is necessary to separate the 23130 bp fragment completely from the 4361 bp fragment in order to obtain clear bands. Otherwise, the 23130 bp fragment is overloaded and smears upwards, while the 4361 bp fragment is only faintly visible.

Sample application/concentration:

Standard loading for mini to midi gels per lane

- with bands visible in UV-light after ethidium bromide-staining:
 - $0.5 1.0 \mu g$ in 10 $\mu l = 1$ lane
- with detection after ethidium bromidestaining with signal-integrated camera systems:
 - $0.3 0.4 \mu g$ in 10 $\mu l = 1$ lane

Optimal are wide lanes (see figure).

<u>Please note:</u> The upper 3 fragments (see figure) have a significantly higher molecular mass than the lower fragments. With standard loading, they therefore appear slightly overloaded and smeared.

We recommend adjusting the application amount depending on whether the upper or the lower bands are to be well represented.

Storage:

Optimal storage temperature is -20 °C. Please avoid repeated thawing and freezing.

We recommend portioning in aliquots. Please mix the solution well before aliquoting to obtain evenly concentrated aliquots.

Lambda Hind III-Marker

X910.1

100 μg + sample buffer

Carl Roth GmbH + Co. KG

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