RNA Marker RL6000

Cat No. : R1041

Size: 5×20μL

Storage: Store at-70°C or -20°C (up to 6 months).

Contents:

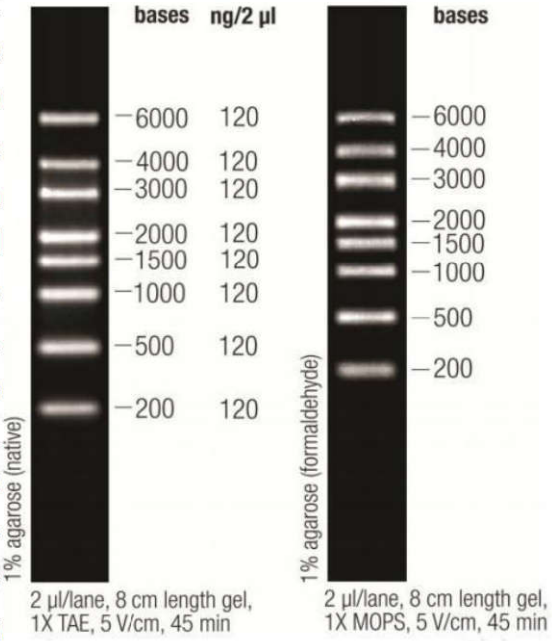
Contents	Amount
RNA Marker RL6000	100 (5×20)μL (for 50 applications)
2X RNA Loading Dye	1 mL

Storage Buffer: 1 mM EDTA (pH 6.0).

2X RNA Loading Dye: 95% formamide, 0.025% SDS, 0.025% bromophenol blue, 0.025% xylene cyanol FF, 0.025% ethidium bromide, 0.5 mM EDTA.

Product Description:

RNA Marker RL6000 is a mixture of eight chromatography-purified single-stranded RNA transcripts (in bases): 6000, 4000, 3000, 2000, 1500, 1000, 500 and 200 The Marker is designed for qualitative and quantitative analysis of RNA on agarose gels stained with ethidium bromide or SYBR Green II. The Marker is free of degraded RNA and NTP's. Therefore, spectrophotometric measurements provide accurate values of RNA concentration in each ladder band. Due to this feature, the RNA Marker could be used for approximate RNA quantification on gels. The RNA Marker RL6000 is recommended for electrophoresis in the following: native 1% agarose with TAE buffer, denaturing formaldehyde agarose with MOPS buffer and denaturing glyoxal/DMSO agarose with sodium phosphate buffer.



Protocol:

Note:

- RNA Markers, as any RNA, are extremely sensitive to degradation by ribonucleases. To avoid RNA degradation, use protective gloves and prepare fresh gels and electrophoresis buffers just before use. Plastic ware, tips and solutions should be treated with diethyl pyrocarbonate.
- Use the supplied 2X RNA Loading Dye both for sample RNA and RNA Marker.
- Mix equal volumes of the 2X RNA Loading Dye and RNA sample, heat at 70ºC for 10min, chill on ice and load.
- Loading of equal volumes of the sample and the Marker is recommended. The required volumes can be obtained by diluting samples with the 2X RNA Loading Dye and Water,

nuclease-free.

- The 2X RNA Loading Dye contains a denaturing agent formamide. When samples are treated with this agent, RNA molecules separate according to their size both on native and denaturing agarose gels.
- For more precise RNA analysis and for Nothern blots, denaturing electrophoresis is recommended.

I. Marker preparation for loading*

(1) Thaw the Marker on ice.

(2) Mix the contents well by pipetting or by gentle vortexing, as concentration gradients may form in frozen products over time.

(3) Use a 0.25μL aliquot of the Marker per 1mm of the gel lane width.

(4) Prepare the following for 8mm width of gel lane :

–2μL of 2X RNA Loading Dye,

–2μL of RNA Marker RL6000.

(5) Vortex briefly and spin down.

(6) Heat at 70ºC for 10min. Chill quickly on ice and load on gel.

*The prepared probe is suitable for electrophoresis both in native agarose with TAE buffer, and in denaturing formaldehyde agarose with MOPS buffer.

II. RNA visualization

- The 2X RNA Loading Dye allows for RNA visualization without additional staining of denaturing agarose gels. If RNA fragments are separated on native agarose gels, additional staining with ethidium bromide is recommended.
- When visualizing a gel under UV light, an additional dark zone of ethidium bromide can sometimes be observed. However, this has no influence on the quality of RNA separation.
- Avoid long exposure to the UV light, as this may cause RNA degradation.
- For Northern blots, perform electrophoresis in denaturing formaldehyde agarose with MOPS buffer. A 2μL aliquot of the RNA Marker is well visible after being transferred on Hybond-N^+ membrane from 1% formaldehyde gel, whereas the same amount of RNA Marker is less visible when transferred from 1% native agarose.
- The ethidium bromide present in the sample and/or in the gel does not interfere neither with the RNA transfer onto the membrane, nor with RNA hybridization with the probe.