

RNA Millennium™ Markers

Store below -70°C.

Catalog # (P/N):	AM7150
Amount:	25 lanes
Concentration:	1.0 µg/µL
Volume:	50 µL
Storage Conditions:	Store below -70°C. Avoid multiple freeze-thaw cycles. The product may be stored short-term at -20°C.
Storage Buffer:	0.1 mM EDTA

USER INFORMATION

Product Description:	RNA Millennium™ Markers are a set of 10 transcripts, 0.5, 1, 1.5, 2, 2.5, 3, 4, 5, 6 and 9 kb in length. Each transcript contains a 20 base poly(A) tail. They are designed for use as RNA size standards in Northern blots and can be visualized by ethidium bromide staining and UV fluorescence, or by autoradiography using the Ambion® Millennium Marker Probe (P/N AM7785; Figure 1).
Handling Instructions:	RNA is very sensitive to degradation by exogenous ribonucleases introduced during handling. Wear gloves when handling this product. Use RNase-free reagents, tubes, and barrier pipette tips. Thawing Instructions Thaw just to completion at 37°C, vortex for a few seconds when fully thawed, and place on ice. Aliquot the RNA, if necessary, to minimize freeze-thaw cycles (≤5).
Applications:	RNA Millennium Markers are designed for use on denaturing agarose gels for Northern analysis. Before use, thaw RNA Millennium Markers and vortex for approximately 15 sec, to ensure homogeneity and consistent staining. Two microgram (2 µg) of RNA Millennium Markers should be enough to visualize within the gel, however, 4 µg may be needed to see all ten bands on the membrane after transfer. Formaldehyde Gels Use 2 µg (2 µL) aliquots directly by mixing them with 3X volume of NorthernMax® Formaldehyde Load Dye (P/N AM8552) and 50 µg/mL ethidium bromide (final concentration in the sample). Alternatively, dilute 2 µL aliquots of the markers with 3 µL of Nuclease-free water and mix with 3X volume (15 µL) NorthernMax Formaldehyde Load Dye. Vortex samples briefly in NorthernMax Formaldehyde Load Dye, centrifuge briefly, heat to 65°C for 15 min, and place on ice. A 1 % denaturing agarose gel is suitable for separation of these markers (Figure 1). Note: Gel parameters can greatly affect the appearance of RNA Millennium Markers. Using less than optimal conditions, total RNA may look intact while the RNA Millennium Markers may appear somewhat smeared. Optimal gel conditions include a freshly prepared denaturing gel, utilizing high quality reagents such as those found in the Ambion NorthernMax Kit (P/N AM1940). The gel should not become hot during electrophoresis. We recommend 4 volts/cm, measured between the electrodes, and running the bromophenol blue to within 3 cm of the bottom of the gel. Overloading the markers will also decrease the resolution. Ethidium Bromide Staining The markers may be visualized by several methods. <ol style="list-style-type: none">Add ethidium bromide to the sample at 10–50 µg/mL final concentration before loading.Incorporate ethidium bromide into the gel or running buffer at 0.5 µg/mL.Stain post-electrophoresis using 0.5 µg/mL ethidium bromide in gel running buffer (e.g., 1 µL of 10 mg/mL ethidium in 20 mL of buffer). Note: For methods B or C, destain for 5–10 min in buffer without ethidium bromide.

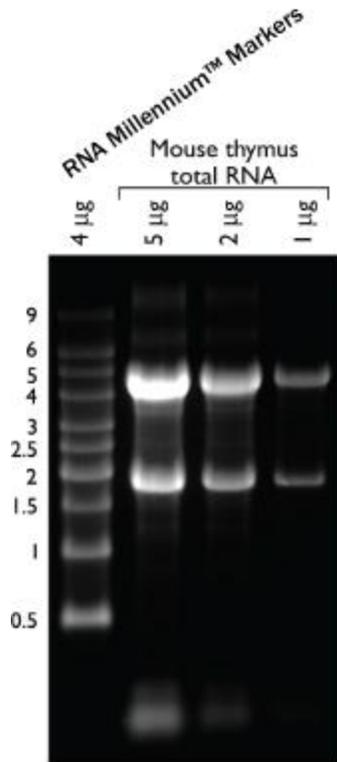


Figure 1: RNA Millennium™ Markers (4 µg) along with total RNA were electrophoresed on a 1% denaturing agarose gel and stained with ethidium bromide.

QUALITY CONTROL

Functional Testing: 2 µg (2 µL) of RNA Millennium Markers generates 10 distinct bands on a 1% denaturing agarose gel with ethidium bromide staining.

The markers remain intact when incubated at 37°C overnight.

OTHER INFORMATION

Material Safety Data Sheets: Material Safety Data Sheets (MSDSs) can be printed or downloaded from product-specific links on our website at the following address: www.ambion.com/techlib/msds. Alternatively, e-mail your request to MSDS_Inquiry_CCRM@appliedbiosystems.com. Specify the catalog or part number(s) of the product(s), and we will e-mail the associated MSDSs unless you specify a preference for fax delivery. For customers without access to the internet or fax, our technical service department can fulfill MSDS requests placed by telephone or postal mail. (Requests for postal delivery require 1–2 weeks for processing.)

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