

RNA Fragmentation Reagents

Store at room temperature.

Catalog #:	AM8740
Components:	10X Fragmentation Reagent - 200 μ L of a buffered zinc solution Stop Solution - 200 μ L of 200 mM EDTA pH 8.0
Appearance:	Clear solutions
Storage Conditions:	Store at room temperature.

USER INFORMATION

Product Description: Ambion[®] RNA Fragmentation Reagents are designed to fragment RNA (both labeled and unlabeled) to sizes between 60–200 nucleotides. Fragmentation of amplified RNA is commonly recommended prior to hybridization on printed oligonucleotide microarrays. RNA fragmentation improves the hybridization kinetics and signal produced on oligonucleotide microarrays.

Applications: **For fragmentation of 2–20 μ g of RNA, the recommended reaction volume is 10 μ L.**

1. Bring the RNA sample volume to 9 μ L with Nuclease-free Water (Ambion Cat #AM9938).
2. Add 1 μ L of the 10X Fragmentation Buffer to the RNA sample.
3. Mix, spin briefly, and incubate at 70°C for 15 min in a heating block, thermocycler, or water bath.
4. Add 1 μ L of the Stop Solution, place it on ice until use, or store it at –80°C.

For fragmentation of 20–50 μ g of RNA, the recommended reaction volume is 20 μ L.

1. Bring the RNA sample volume to 18 μ L with Nuclease-free Water (Ambion Cat #AM9938).
2. Add 2 μ L of the 10X Fragmentation Buffer to the RNA sample.
3. Mix, spin briefly, and incubate at 70°C for 15 min in a heating block, thermocycler, or water bath.
4. Add 2 μ L of the Stop Solution, place it on ice until use, or store it at –80°C.

Note: The reaction volume and the incubation time are important to obtain RNA fragments with preferred size range.

RELATED PRODUCTS

MessageAmp[™] RNA Amplification Kits

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Generate high yields of amplified and labeled cRNA from low input amounts of total RNA, for microarray-based expression profiling. Applied Biosystems offers a full line of Ambion[®] MessageAmp Kits tailored for different array analysis applications.

QUALITY CONTROL

Nonspecific Endonuclease Activity:	Meets or exceeds specification when a sample is incubated for 14–16 hr with 300 ng supercoiled plasmid DNA and analyzed by agarose gel electrophoresis.
Exonuclease Activity:	Meets or exceeds specification when a sample is incubated for 14–16 hr with 40 ng ³² P-labeled <i>Sau3A</i> fragments of pUC19 and analyzed by PAGE.
RNase Activity:	Meets or exceeds specification when a sample is incubated for 14–16 hr with 25 ng ³² P-labeled RNA and analyzed by PAGE.
Functional Testing:	Following the protocol above, 5 μ g of a total RNA sample is fragmented. RNA products of fragmentation reaction are required to be smaller than 200 nucleotides as measured on an Agilent bioanalyzer RNA 6000 Nano LabChip.

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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For research use only. Not for use in diagnostic procedures.

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