

PRODUCT INFORMATION

RNase A, DNase and Protease-free

Pub. No. MAN0012003

Rev. Date 09 January 2017 (Rev. B.00)

Lot: _ **Expiry Date:** _

Store at -20 °C

Components	#EN0531
RNase A, DNase and Protease-free, 10 mg/mL	10 mg

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Description

RNase A is an endoribonuclease that specifically degrades single-stranded RNA at C and U residues. It cleaves the phosphodiester bond between the 5'-ribose of a nucleotide and the phosphate group attached to the 3'-ribose of an adjacent pyrimidine nucleotide. The resulting 2', 3'-cyclic phosphate is hydrolyzed to the corresponding 3'-nucleoside phosphate (1, 2).

Applications

- Plasmid and genomic DNA preparation (3, 4).
- Removal of RNA from recombinant protein preparations.
- Ribonuclease protection assays. Used in conjunction with RNase T1 (3).
- Mapping single-base mutations in DNA or RNA (5, 6).

Source

Bovine pancreas.

Molecular Weight

13.7 kDa monomer.

Concentration

Protein concentration is determined by measuring the absorbance at 278 nm using molar absorption coefficient $\epsilon=9800 \text{ M}^{-1}\text{cm}^{-1}$ (7).

Definition of Activity Unit

One unit of the enzyme causes an increase in absorbance of 1.0 at 260 nm when yeast RNA is hydrolyzed at 37 °C and pH 5.0. Fifty units are approximately equivalent to 1 Kunitz unit (8).

Specific activity

≥5000 U/mg protein (≥100 Kunitz units/mg protein).

Storage Buffer

The enzyme is supplied in: 50 mM Tris-HCl (pH 7.4) and 50% (v/v) glycerol.

Inhibition and Inactivation

- Inhibitors: the most potent inhibitor is a mammalian ribonuclease inhibitor, e.g., Thermo Scientific RiboLock RNase Inhibitor (#EO0381).
Other inhibitors:
uridine 2',3'-cyclic vanadate, 5'-diphosphoadenosine 3'-phosphate and 5'-diphosphoadenosine 2'-phosphate (2), SDS, diethyl pyrocarbonate, 4 M guanidinium thiocyanate plus 0.1 M 2-mercaptoethanol and heavy metal ions.
- Not inactivated by heating, reliably removed by spin column or phenol/chloroform extraction.

Note

- Recommended concentration of RNase A is 1-100 µg/mL depending on the application.
- The enzyme is active under a wide range of reaction conditions. At low salt concentrations (0 to 100 mM NaCl), RNase A cleaves single-stranded and double-stranded RNA as well the RNA strand in RNA-DNA hybrids. However, at NaCl concentrations of 0.3 M or higher, RNase A specifically cleaves single-stranded RNA (9).

CERTIFICATE OF ANALYSIS

Endodeoxyribonuclease Assay

No detectable degradation was observed after incubation of supercoiled plasmid DNA with RNase A, DNase and Protease-free.

Labeled Oligonucleotide (LO) Assay

No detectable degradation after incubation of single-stranded or double-stranded radiolabeled oligonucleotides with RNase A.

Protease Assay

No detectable degradation of protease substrate after incubation of FTC-casein with RNase A.

Quality authorized by:



Jurgita Zilinskiene

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References

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