



Product Information

RIBONUCLEASE A

Product Number **R5125, R4875, R5503, R5000, R5250 and R5500**

Storage Temperature Freezer

CAS #: 9001-99-4

EC# 3.1.27.5

Synonyms: RNase A, Pancreatic RNase, Ribonuclease I, Endoribonuclease I, and Ribonuclease 3'-pyrimidinooligonucleotidohydrolase.

Product Description

Appearance: White to white with a yellow cast powder
Molecular weight: 13,700 based upon the amino acid sequence¹

Extinction coefficient: The E1% is 7.0 when measured at 280 nm²

Isoelectric point: The isoelectric point (PI) for this enzyme is 9.6³

Ribonuclease A (RNase A) is a single chain polypeptide containing 4 disulfide bridges. In contrast to RNase B it is not a glycoprotein.⁴ RNase A can be inhibited by alkylation of the histidine-12 or histidine-119 which are present in the active site of the enzyme.⁵ Activators of RNase A include potassium and sodium salts. The optimal temperature for activity is 60°C, although the enzyme does exhibit activity from 15-70°C. The pH optimum is 7.6, with an activity range of 6-10⁶. The highest activity is exhibited with single stranded RNA.⁷ RNase A is a very stable enzyme and can withstand temperatures up to 100°C. At 100°C, RNase A is most stable between pH 2.0 and 4.5⁸.

RNase A is an endoribonuclease that attacks at the 3' phosphate of a pyrimidine nucleotide. The sequence of pG-pG-pC-pA-pG will be cleaved to give pG-pG-pCp and A-pG. The highest activity is exhibited with single stranded RNA.⁷

Components

RNase A is isolated from bovine pancreas. A method of preparation is described in Crestfield, A.M. et.al.⁸ This product is supplied as an essentially protease and salt-free lyophilized powder.

Disclaimer/Precautions

RNase A is stable to both heat and detergents. In addition, it adsorbs strongly to glass. Scrupulous precautions are necessary to insure that residues of RNase A do not cause artifacts in processes requiring intact RNA.

Preparation Instructions

When Sigma tests the activity of RNase A a stock solution is prepared in water at 1 mg/mL. Prior to usage, if the DNase impurity is a concern, one can use Sigma DNase free RNase that can be used directly (R6513 and R4642). Boiling of these products is not required prior to use.

RNase A can be made free of DNase by boiling. According to the following method described by Sambrook, J. Fritsch, E.F. and Maniatis, T.⁹, prepare a 10 mg/mL stock solution in 10 mM sodium acetate buffer (pH 5.2). Heat to 100°C for 15 minutes, allow to cool to room temperature, and then adjust to pH 7.4 using 0.1 volumes of 1 M TRIS/HCl pH 7.4. Aliquot and store at -20°C. If RNase A is boiled at a neutral pH, precipitation will occur. When boiled at a low pH, some precipitation may occur because of protein impurities that are present.

Storage/Stability

RNase A is recommended to be stored in a freezer location (- 0°C). If properly stored, this product should have an approximate shelf life of 2 years. Stock solutions stored in frozen aliquots are stable for at least 6 months.

Procedure

A major application for RNase A is the removal of RNA from preparations of plasmid DNA. For this application, DNase free RNase A is used at a final concentration of 10 ug/mL.¹⁰

Another use for RNase A is that can be used to test for complementarity between RNA:DNA hybrids.

References

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