RNase A

1.2018

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RNase A

The Ribonuclease A (RNase A) is a 13.7 kDa (monomer) endoribonuclease isolated from bovine pancreas, which selectively cleaves single-stranded RNA 3′ next to pyrimidine residues (cytosine, uracil). It degrades RNA to cyclic nucleotide monophosphates to 5′-OH and 2′-, 3′-cyclic monophosphate. The enzyme exhibits no endonuclease or exonuclease activity towards DNA substrates. The RNase A is used to remove RNA during the isolation procedures of plasmid and genomic DNA.

The enzyme is very active under a wide range of reaction conditions and difficult to inactivate. At low salt concentrations (up to 100 mM NaCl), the RNase A cleaves single- and double-stranded RNA as well as an RNA strand in RNA-DNA hybrids. However, under high salt concentrations (>300 mM NaCl), the RNase A specifically cleaves single-stranded RNA.

Features

- Enzyme activity: > 80 Kunitz units/mg protein
- Supplied in a salt-free, freeze dried form
- **DNase-free**
- Protease-free

Applications

- Purification of RNA-free DNA
- Plasmid and genomic DNA isolation
- Removal of RNA during recombinant proteins preparations
- RNA protection assays
- Mapping of single-base mutations in DNA or RNA

Usage

- → Stock solutions should be prepared to a final concentration 1–10 mg/ml by resuspending in 10 mM Tris-HCl (pH 7.5), 15 mM NaCl, 50% (v/v) glycerol or in TE buffer.
- → The recommended working solution concentration depends on application.
  - For removal of RNA from plasmid preparations use 10 µg/ml working solution and incubate sample for 1 hour at room temperature.
  - For preparation of „blunt ends" of double-stranded cDNA use 100 ng/ml working solution.

Additional information

- → The RNase A has a high affinity to glass surfaces.
- → At neutral pH & high concentrations (>10 mg/ml) the enzyme will precipitate.
- → The enzyme is inhibited by diethyl pyrocarbonate (DEPC), guanidinium salts (4 M GuSCN), β-mercaptoethanol, heavy metals and RNase-inhibitors like ***RIBO****PROTECT Hu* (RT35).
- → In order to remove the enzyme from a sample, perform a separation with spin columns or several phenol/chloroform extractions.

Quality control

The activity of the enzyme, absence of deoxyribonucleases and proteases has been confirmed in appropriate quality tests.

Unit definition

One unit of activity is defined as that amount of enzyme which causes the hydrolys is of RNA to yield a velocity constant, k=1, at 25°C and pH 5.0 (Kunitz-Unit).

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Component	RP14	RP145
RNase A	10 mg	50 mg

Storage & shipping

Storage conditions
Keep at -20°C (lyophilized or in a 50% glycerol solution) for long-term storage or at +4°C for up to several weeks. When stored at -20°C (lyophilized or in a glycerol solution), the enzyme remains stable for several years.

Shipping conditions
Shipping at ambient temperature.

ⓘ For research use only

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