



E. coli Ribonuclease H (RNase H), Recombinant

02-060 1,000 units, # 02-060-5 5 x 1,000 units

Ribonuclease H (RNase H) is an endoribonuclease which specifically degrades the RNA strand of an RNA/DNA hybrid, leaving the DNA strand and unhybridized RNA intact. *E. coli* RNase H (RNaseHI) was over-expressed in *E. coli* as a recombinant protein and the protein then purified. MW is 17.6 kDa.

Applications

1. Removal of mRNA in DNA/RNA hybrid prior to the synthesis of the second strand of cDNA (1, 2)

2. Removal of poly (A) tails from mRNA after hybridization with oligo (dT) (3)

3. Oligodeoxyribonucleotide-directed site-specific cleavage of RNA (4)

Specification

Form: 50 units/ul in 20mM Tris-HCI (pH 7.5), 100mM KCI, 1mM DTT, 50% Glycerol Specific Activity: 100,000 units/mg protein Unit Definition: 1 unit is defined as the amount of the enzyme that hydrolyzes 1 nmol of the RNA in ³H labeled M13 DNA/RNA hybrid to acid-soluble ribonucleotides in 20 min at 37°C. Storage: -20°C

Quality: Greater than 95% protein determined by SDS-PAGE (Fig. 1, CBB staining). Endo- and exo-DNase activities and RNase activity were not detected with 100 U/ml RNaseH in 50 ul reaction at 37°C.

Reagents Supplied with Enzyme: 10X RNaseH Reaction Buffer: 100 mM Tris-HCl (pH 8.0), 100 mM MgCl₂, 500 mM NaCl, 10 mM DTT, 500 ug/ml BSA (Bovine Serum Albumin)

Caution: To avoid contamination of trace amounts of nucleic acids in BSA, use reaction buffer that does not contain BSA and use RNaseH at higher concentrations.

Data Link: UniProtKB/Swiss-Prot POA7Y4 (RNH_ECOLI)

References:

- 1. Gubler U (1987) "Second-strand cDNA synthesis: mRNA fragments as primers." *Method Enzymol* 152: 330-335 PMID: <u>3309563</u>
- 2. Sambrook J & Russell DW (2001) *Molecular Cloning*, Chapter 11 "Preparation of cDNA Libraries and Gene Identification". CSHL Press
- Vournakis JN *et al* (1975) "Electrophoretic patterns of deadenylylated chorion and globin mRNAs." *Proc Natl Acad Sci USA* 72: 2959-2963 PMID: <u>1059086</u>
- Donis-Keller H (1979) "Site specific enzymatic cleavage of RNA." Nucleic Acids Res. 7: 179-192 PMID: <u>386279</u>



Fig.1 SDS-PAGE of E. coli RNaseH

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