

## 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 highly 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)

## Specifications

Form: 50 units/ul in 20mM Tris-HCl (pH 7.5), 100mM KCl, 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 <sup>3</sup>H labeled M13 DNA/RNA hybrid to acid-soluble ribonucleotides in 20 min at 37°C.

Storage: at -20°C

Quality Assurance: Greater than 95% protein determined by SDS-PAGE (CBB staining) (Fig.1). Endoand 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: RNaseH Reaction Buffer (10 X): 100 mM Tris-HCl (pH 8.0), 100 mM MgCl<sub>2</sub>, 500 mM NaCl, 10 mM DTT, 500 ug/ml BSA (Bovine Serum Albumin)

\*Caution; To avoide 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: 3309563
- 2. Sambrook J & Russell DW (2001) *Molecular Cloning*, Chapter 11 "Preparation of cDNA Libraries and Gene Identification". CSHL Press
- 3. Vournakis JN *et al* (1975) "Electrophoretic patterns of deadenylylated chorion and globin mRNAs." *Proc.Natl.Acad.Sci.USA* **72**: 2959-2963 PMID: 1059086
- 4. Donis-Keller H (1979) "Site specific enzymatic cleavage of RNA." Nucleic Acids Res. 7: 179-192 PMID: 386279

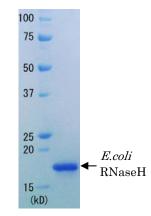


Fig.1 SDS-PAGE of *E.coli* RNaseH