



## ***E. coli* Ribonuclease H (RNase H), Recombinant**

### **BACKGROUND**

**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
- Size:** 1,000 units
- 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 3H labeled M13 DNA/RNA hybrid to acid-soluble ribonucleotides in 20 min at 37°C.
- Quality Assurance:** Greater than 95% protein determined by SDS-PAGE (CBB staining) (Fig.1). 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:** 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)
- Data Link:** UniProtKB/Swiss-Prot [P0A7Y4](#) (RNH\_ECOLI)
- Storage:** Store at -20°C
- 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

**\*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.

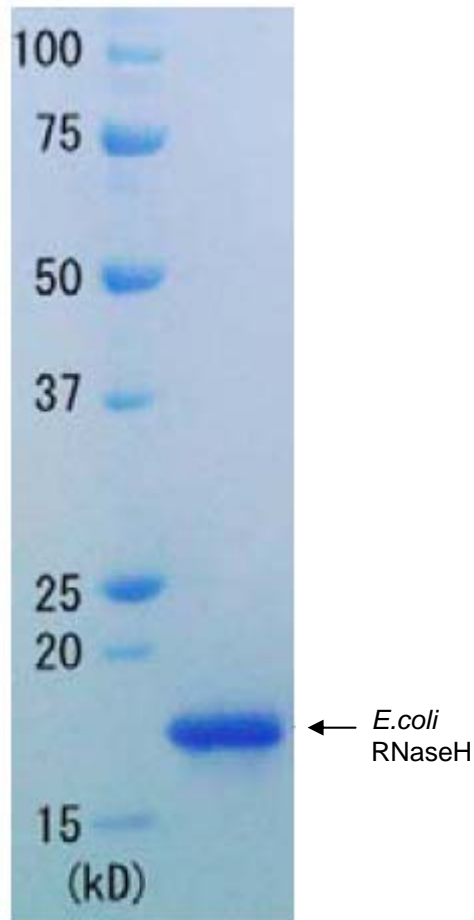


Fig.1 SDS-PAGE of *E. coli* RNaseHE.

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E-mail: [export@cosmobio.co.jp](mailto:export@cosmobio.co.jp)

Phone : +81-3-5632-9617

FAX : +81-3-5632-9618