

RNase H

Cat.No. LH101

Storage: at -20°C for two years

Concentration: 5,000 units/ml

Description

Ribonuclease H (RNase H) specifically degrades the RNA strand in RNA-DNA hybrids. It does not hydrolyze the phosphodiester bonds within single-stranded and double-stranded DNA and RNA. This product is purified from *E. coli* expressing the recombinant rnhA gene on a plasmid with 17 kDa molecular weight and it can be inactivated by heating at 65°C for 20 minutes.

Application

- Removal of mRNA prior to synthesis of second strand cDNA.
- Identification of RNA-DNA Hybrid.

Kit Contents

Component	LH101-01	LH101-02
RNase H	100 units	5×100 units
10×RNase H Buffer	1 ml	5×1 ml

Definition of Activity Unit

One unit is defined as the amount of RNase H that solubilizes 1 nmol RNA in the 3 tagged poly(rA)·poly(dT) hybrid chain in 20 minutes at 37°C in 50 µl system.

Quality Control

RNase A activity: In 50 µl reaction system, 40 units of enzyme are incubated with 1 µg of RNA at 37°C for 1 hour, agarose gel electrophoresis shows that RNA is not degraded.

ssDNA exonuclease activity: In 50 µl reaction system, 40 units of enzyme are incubated with 1 µg of Oligo DNA at 37°C for 0.5 hour, ssDNA is not degraded.

Endonuclease activity: In 50 µl reaction system, 40 units of enzyme are incubated with 1 µg of pBR322 DNA at 37°C for 4 hours, the ratio of RF I to RF II is no more than 10%.

Storage Buffer

25 mM Tris-HCl pH 7.4, 100 mM KCl, 0.1 mM EDTA, 1.5 mM DTT, 200 µg/ml BSA, 50% Glycerol

10×RNase H Buffer

200 mM Tris-HCl pH 8.3, 150 mM DTT, 1 M KCl, 45 mM MgCl₂

Working Condition (Take RT-PCR as an example)

Add 1 µl of RNase H at the end of reverse transcription in the 0.1-1 µg reverse transcription system, then incubate at 37°C for 15-30 minutes.

Precautions

Fully and gently vortex 10×RNase H Buffer before use.

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