

RNase I_f



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M0243S 013130615061

M0243S



5,000 units **50,000 U/ml** **Lot: 0131306**
RECOMBINANT **Store at -20°C** **Exp: 6/15**

Description: Ribonuclease I_f (RNase I_f) is a single strand specific RNA endonuclease which will cleave at all RNA dinucleotide bonds leaving a 5' hydroxyl and 2', 3' cyclic monophosphate (1). RNase I_f is a recombinant protein fusion of RNase I (from *E. coli*) and maltose-binding protein. It has identical activity to RNase I.

Source: An *E. coli* strain containing a genetic fusion of the RNase I gene (*rna*) from *E. coli* and the gene coding for maltose-binding protein (MBP)(2)

Supplied in: 100 mM NaCl, 10 mM Tris-HCl (pH 8.0), 0.5 mM EDTA, 1 mM dithiothreitol and 50% glycerol.

Applications:

- Degradation of single-stranded RNA to mono-, di- and trinucleotides (3)
- Used in ribonuclease protection assays

Reagents Supplied with Enzyme:

10X NEBuffer 3.

Reaction Conditions:

1X NEBuffer 3.
Incubate at 37°C.

1X NEBuffer 3:

100 mM NaCl
50 mM Tris-HCl
10 mM MgCl₂
1 mM dithiothreitol
pH 7.9 @ 25°C

Unit Definition: One unit is defined as the amount of enzyme required to fully digest 1 picomole of synthetic ssRNA 33-mer in a total reaction volume of 10 µl in 15 minutes in 1X NEBuffer 3 as visualized on a 20% acrylamide gel (40:1 Bis) stained with SYBR Gold®.

Unit Assay Conditions: 1X NEBuffer 3 containing 3.3 µM of synthetic ssRNA 33-mer in a total reaction volume of 10 µl.

Quality Control Assays

ss DNA Exonuclease Activity: Incubation of 50 units of enzyme with 1 µg sonicated and denatured [³H] DNA (10⁵ cpm/µg) for 30 minutes at 37°C in 50 µl reaction buffer released < 1% radioactivity.

ds DNA Exonuclease Activity: Incubation of 50 units of enzyme with 1 µg sonicated [³H] DNA (10⁵ cpm/µg) for 30 minutes at 37°C in 50 µl reaction buffer released < 1% radioactivity.

Endonuclease Activity: Incubation of 50 units of enzyme with 1 µg φX174 RF I DNA for 1 hour at 37°C in 50 µl reaction buffer resulted in < 10% conversion to RF II.

Heat Inactivation: 70°C for 20 minutes.

Note: RNase I_f will not degrade DNA. It has a strong preference for single-stranded RNA over double-stranded RNA.

References:

1. Spahr, P. F. and Hollingworth, B. R. (1961) *J. Biol. Chem.* 236, 823–831.
2. Meador, J. III and Kennel, D. (1990) *Gene* 95, 1–7.
3. Meador, J. III Cannon, B., Cannistraro, V. J. and Kennel, D. (1990) *Eur J. Biochem.* 187, 549–553.

CERTIFICATE OF ANALYSIS

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