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Certificate of Analysis

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RNase I (E. coli)

Catalog No: 1330

Lot No: See Product Label
Package Size: See Product Label
Concentration: See Product Label
Protein (Lowry): 0.004 mg/ml
Specific Activity: 1,261,231 units/mg
Storage Conditions: Store at -20°C

Applications

- Hybridization of a synthetic DNA oligomer to a complementary single-stranded region of a RNA molecule can be used to create a site that can be cleaved by RNase H (1)
- Used to remove RNA strand before second strand cDNA synthesis (2, 3)
- Detects RNA-DNA regions in naturally occurring double-stranded DNA (4)
- Used to analyze in vitro polyadenylation reaction products (5)

Unit Definition

One unit is the amount of enzyme required to produce 1 nmol of acid soluble ribonucleotide from [32P]poly(A).poly(dT) at 37°C in 20 minutes

Assay Conditions

20 mM HEPES-KOH (pH 8.0)
50 mM KCl
10 mM $MgCl_2$
1 mM dithiothreitol
0.6 nmoles [^{32}P]poly(A)-poly(dT)
Reaction volume of 25µl

Storage Buffer

20 mM Tris-HCl (pH 7.5 at 4^{o}C)
300 mM KCl
1.0mM DTT
7 mM EDTA
20 mM Mg Acetate
50% glycerol (v/v)

Quality Control

Endonuclease: Incubation of 30, 60, and 120 units of enzyme with 1.0 µg of pBR322 DNA at 37°C for 1 hour resulted in ≤5% conversion of RFI to RFII. Reaction volume of 50 µl.

DNase, double-stranded: Incubation of 30, 60, and 120 units of enzyme with 0.015 µg of [^{32}P] lambda DNA for 1 hr at 37°C resulted in the release of ≤0.4 slope of %-end label released per unit of enzyme. Reaction volume of 50 µl.

DNase, single-stranded: Incubation of 30, 60, and 120 units of enzyme with 0.015 µg of heat-denatured [^{32}P] Lambda DNA for 1 hour at 37^{o}C resulted in ≤0.4 slope of %-end label released per unit of enzyme. Reaction volume of 50 µl.

RNase: Incubation of 30, 60, and 120 units of enzyme with 0.015 µg of [^{32}P] RNA transcript for 1 hr at 37^{o}C resulted in ≤0.4 slope of %-end label released per unit of enzyme. Reaction volume of 50 µl.

Purity: Approximately ≥90% pure, as judged by SDS-polyacrylamide gel electrophoresis.

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References

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(5) Goodwin, E.C. and Rottman, F.M. (1982) Nucl. Acids Res. 20, 916
(6) Hillenbrand, G. and Staudenbauer, W.L. (1982) Nucl. Acids Res. 10, 833-852