DNase I (RNase-free)

NEW ENGLAND BioLabs Inc.
1-800-632-7799
info@neb.com
www.neb.com

M0303S 014150417041

M0303S

RR Yes

1,000 units **2,000 U/ml** **Lot: 0141504**
RECOMBINANT **Store at –20°C** **Exp: 4/17**

Description: DNase I (RNase-free) is an endonuclease that nonspecifically cleaves DNA to release di-, tri- and oligonucleotide products with 5´ -phosphorylated and 3´ -hydroxylated ends (1,2). DNase I acts on single- and double-stranded DNA, chromatin and RNA:DNA hybrids.

Source: An *E. coli* strain that carries an MBP fusion clone of Bovine Pancreatic DNaseI.

Now Recombinant

Applications:

- Degradation of DNA template in transcription reactions
- Removal of contaminating genomic DNA from RNA samples
- DNase I footprinting
- Nick Translation

Supplied in: 2 mM $CaCl_2$, 10 mM Tris-HCl (pH 7.6) and 50% glycerol.

Reagents Supplied with Enzyme:
10X DNase I Reaction Buffer.

Reaction Conditions: 1X DNase I Reaction Buffer. Incubate at 37°C.

1X DNase I Reaction Buffer:
10 mM Tris-HCl
2.5 mM $MgCl_2$
0.5 mM $CaCl_2$
pH 7.6 @ 25°C

Unit Definition: One unit is defined as the amount of enzyme which will completely degrade 1 µg of pBR322 DNA in a total reaction volume of 50 µl in 10 minutes at 37°C.

Complete degradation is defined as the reduction of the majority of DNA fragments to tetranucleotides or smaller.

Protocol

Typical Reaction:

1. Resuspend 10 µg RNA in 1X DNase I Reaction Buffer to a final volume of 100 µl.
2. Add 2 units of DNase I, mix thoroughly and incubate at 37°C for 10 minutes.
3. Add 1 µl of 0.5 M EDTA (to a final concentration of 5 mM).
4. Heat inactivate at 75°C for 10 minutes.

Quality Assurance: Free of Detectable RNases.

Quality Control Assays

RNase Activity: Incubation of 100 units of DNase I with 10 µg of double-stranded RNA Ladder for 2 hours at 37°C resulted in the same electrophoretic profile as untreated RNA. Incubation of 2 units of DNase I with 10 µg of single-stranded RNA Ladder for 1 hour at 37°C resulted in the same electrophoretic profile as untreated RNA.

Heat Inactivation: 75°C for 10 minutes.

Note: EDTA should be added to a final concentration of 5 mM to protect RNA from being degraded during enzyme inactivation (3).

References:

1. Kunitz, M. (1950) *J. Gen. Physiol.* 33, 349–362.
2. Vanecko, S. and Laskowski, M. (1961) *J. Biol. Chem.* 236, 3312–3316.
3. Huang, Z. (1996) *Biotechniques* 20, 1012–1020.

CERTIFICATE OF ANALYSIS

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