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roboklon

DNase I - RNase free

Deoxyribonuclease I

DNase I
Endonuclease

Nonspecific deoxyribonuclease that degrades both double-stranded and single-stranded DNA endonucleolytically releasing 5'-phosphorylated di-, tri-, and oligonucleotide products (2).

Cat. No.	Size
E1345-01	1.000 units
E1345-02	5.000 units

Unit Definition:
One unit is the amount of enzyme required to completely degrade 1 µg of plasmid DNA in 10 min at 37°C.
One functional DNase I unit is approximately equivalent to 0.3 Kunitz units (1)

Storage Conditions:
Store at -20°C

Quality Control:
Functionally tested for digesting of template - plasmid DNA. The absence of RNase confirmed by appropriate quality test utilizing spectrophotometry assays of RNA sample concentration before and after incubation with an excess of enzyme.

Note 1: This DNase solution does not contain RNase inhibitor. Please handle with care to avoid RNase contamination.
Ribonuclease inhibitor is available as a separate product (Cat. No. E4210).

Note 2: DNase I is sensitive to physical denaturation. Therefore, do not vortex solutions containing DNase I. Mix by gently flipping the tube or by pipetting.

Applications:

- Preparation of DNA-free RNA (degradation of contaminating DNA after RNA isolation) (3)
- Preparation of DNA-free RNA prior to RT-PCR and RT-qPCR (4)
- Removal of template DNA following *in vitro* transcription.
- Studies of DNA-protein interactions (footprinting).
- DNA labeling by nick-translation.
- Production of random fragments (generation of libraries) (5)

Enzyme activity:

DNase I requires Ca^{2+} and Mg^{2+} for hydrolyzing double-stranded DNA. In the presence of Mg^{2+}, DNase I cleaves each strand of double-stranded DNA independently in a statistically random fashion (recommended Reaction Buffer I). In the presence of Mn^{2+}, the enzyme cleaves both DNA strands at approximately the same site, producing DNA fragments with blunt-ends or with overhang termini of only one or two nucleotide (recommended Reaction Buffer II) (5).

10 x Reaction Buffer I:

100 mM Tris-HCl, 25 mM $MgCl_2$, 100 mM $CaCl_2$, pH 7.4 @ 25°C.

10 x Reaction Buffer II:

20 mM Tris-HCl (pH 7.5 at 22°C), 300 mM KCl, 0.1 mM dithiothreitol, 7 mM EDTA, 20 mM magnesium acetate, 200 µg/ml bovine serum albumin and 50% [v/v] glycerol.

Inactivation:

Inactivated by heating at 65°C for 10 min in the presence of EDTA or EGTA.

Inhibitors:

Metal chelators (EGTA, EDTA), transition metals, SDS, reducing agents (DTT, β-mercaptoethanol).

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. *Although not required for most applications please see the additional DNase I digestion conditions in Manual for GeneMATRIX UNIVERSAL DNA/RNA/Protein Purification Kit (E3597) and GeneMATRIX UNIVERSAL RNA Purification Kit (E3598).*
4. *Sanyal, A., et al., An effective method of completely removing contaminating genomic DNA from an RNA sample to be used for PCR. Mol. Biotechnol., 8, 135-137, (1997).*
5. *Sambrook, J., Fritsch, E.F. and Maniatis, T. (1989) Molecular Cloning: A Laboratory Manual 2nd ed., Cold Spring Harbor Laboratory, Cold Spring Harbor, New York.*