

mRNex® Deoxyribonuclease I (DNase I) GMP-grade (2 U/μL)

Cat. no. LD10611

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Product Information

Product name	mRNex® Deoxyribonuclease I (DNase I) GMP-grade (2 U/μL)
Cat.No.	LD10611
Size	500 U/2000 U/10000 U/10 KU
Storage and shipping	Shipped on dry ice. Store at -15 °C to -25 °C for up to one year.
Applications	Widely used in the preparation of DNA-free RNA Removal of template DNA after in vitro transcription Preparation of DNA-free RNA before RT-PCR and RT-qPCR In combination with DNA Polymerase I for DNA labeling through nick translation DNA fragmentation for library construction

2. Product Description

DNase I is an endonuclease that can digest single-stranded and double-stranded DNA to produce single deoxynucleotides or single-stranded or double-stranded oligodeoxynucleotides. It can hydrolyze the phosphodiester bond to produce monodeoxynucleotides and oligodeoxynucleotides containing 5'-phosphate groups and 3'-OH groups. The average digestion product is the smallest polytetranucleotide.

DNase I can catalyze many forms of DNA, such as single-stranded DNA, double-stranded DNA, and even chromatin (its cutting rate is affected by histones). The optimum pH range is 7–8. The activity of DNase I depends on Ca^{2+} and can be activated by divalent metal ions such as Co^{2+} , Mn^{2+} , Zn^{2+} , etc. 5 mM Ca^{2+} can protect the enzyme from being hydrolyzed.

In the presence of Mg^{2+} , the enzyme can recognize and cut any site on any strand of DNA randomly.

In the presence of Mn^{2+} , it can recognize two strands of DNA simultaneously and cut at almost the same site to form blunt ends, or sticky ends with 1–2 nucleotides protruding.

This product is produced in accordance with GMP process requirements, and is provided in liquid form.

3. Product Properties

Source	Recombinant E. coli with DNase I gene
Optimum Temperature	37°C
Storage Buffer	10 mM Tris-HCl (pH 7.6), 2 mM CaCl ₂ , 50% (v/v) Glycerol
Unit Definition	One activity unit (Kunitz Unit) is defined as the amount of enzyme required to increase the absorbance at 260 nm of the reaction solution by 0.001 within 1 minute at 25 °C and pH 5.0, using calf thymus DNA as the substrate. Reaction buffer composition: 10 mM Tris-HCl (pH 7.6), 2.5 mM MgCl ₂ , 0.5 mM CaCl ₂ , and 1 µg plasmid DNA.

4. Experimental Methods

4.1. Plasmid template digestion

4.1.1. Reaction system:

Use the RNase-free centrifuge tube and pipette tip to prepare the following reaction system:

Component	Volume
10× DNase I Buffer*	1 µL
DNase I	1 µL
RNA	x µL
RNase-free ddH ₂ O	Up to 10 µL

*Note: 1× DNase I Buffer: 10 mM Tris-HCl, 2.5 mM MgCl₂, 0.5 mM CaCl₂, pH 7.6 @ 25 °C

4.1.2. Reaction conditions:

Incubate at 37 °C for 15–30 minutes. Then, add EDTA solution to a final concentration of 2.5 mM and mix well. Heat at 65 °C for 10 minutes.

The processed template can be used in subsequent reactions such as capping reaction.

4.2. DNase I inactivation or inhibition

After adding EDTA to a final concentration of 2.5 mM, heating at 65°C for 10 min can inactivate DNase I. Phenol and chloroform extraction can also inactivate DNase I. The following conditions all have significant inhibitory effect on DNase I: Metal ion chelating agents, zinc ions with a concentration of millimoles/liter, 0.1% SDS, DTT, mercaptoethanol and other reducing agents, the salt concentrations above 50–100 mM

5. Notes

5.1. Enzymes should be stored in an ice box or on an ice bath when used, and should be stored at -20°C immediately after use.

5.2. For your safety and health, please wear personal protective equipment (PPE), such as laboratory coats and disposable gloves, when operating with this product