Version: 03
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DATASHEET

# DNase I

**Cat. No.:** E00053-100; E00053-500; E00053-1000
**Size:** 100 U/ 500 U/ 1000 U/ 5000 U/ 10000 U

## Product Introduction

Deoxyribonuclease I (DNase I) is DNA-specific endonuclease that cleaves both single-stranded DNA,double-stranded DNA and DNA-RNA hybrids, yielding 5'-phosphate-terminated polynucleotides with a free hydroxyl group on position 3'-producing tetranucleotides. The activity of DNase I is strictly dependent on $Ca^{2+}$ and can be activated by divalent metal ions such as $Mg^{2+}$ or $Mn^{2+}$. In the presence of $Mg^{2+}$, DNase I nonspecifically recognizes and cleaves a double-stranded DNA at anysite on either strand, and in the presence of $Mn^{2+}$, it recognizes and cleaves almost the same sites on both strands of the DNA to produce DNA fragments with blunt ends or sticky ends with 1~2 nucleotide overhangs.

GenScript is offering DNase I produced by expression in a *P. pastoris* strain carrying a plasmid encoding the bovine DNase I.

**Source:** Recombinant DNase I expressed by yeast.
**Species:** Bovine
**Molecular Weight:** 35-40 kDa, on SDS-PAGE under reducing conditions.
**Unit Definition:** One unit is the amount of the enzyme that increases the absorbance at 260 nm by 0.001 per minute at 25 °C, pH 5.0, with calf thymus DNA as the substrate.
**Optimal active temperature:** 37 °C
**Formulation:** Supplied as a solution of 20 mM sodium acetate, 5 mM $CaCl_2$, 0.1 mM PMSF, 50% (v/v) glycerol, pH 6.5 at 25 °C.
**Storage & Stability:** This product remains stable up to 12 months at -20 °C. Avoid repeated freeze-thaw cycles.
**Inactivation:** Add EDTA with a final concentration of 2.5 mM and heat the solution at 65 °C for 10 min can inactivate DNase I.
**Application:**

- DNA template digestion following *in vitro* transcription
- Genomic DNA digestion prior to RT-PCR
- Preparation of DNA-free RNA samples
- Nick-translation
- Studies of DNA-protein interactions by DNase I, RNase-free footprinting
- Prevent cell clumping without affecting cell viability

## Quality Control Specifications

| Assay | Specifications |
|---|---|
| Appearance | Clear, colorless liquid |
| Purity | ≥ 95% as analyzed by SDS-PAGE |
| Enzyme Activity | ≥ 2 U/μl |
| Endotoxin Level | ≤ 0.1 EU/μg of protein by gel clotting method |

| Residual RNase | Non-detectable |
|---|---|

## Reagents Supplied:

| Components | Amount | | | | | Storage |
|---|---|---|---|---|---|---|
| DNase I | 100 U | 500 U | 1000 U | 5000 U | 10000 U | -20 °C |
| DNase I 10× Reaction Buffer | 1 ml*1 vial | 1 ml*1 vial | 1 ml*2 vials | 6 ml*1 vial | 6 ml*2 vials | -20 °C |

## Typical protocol for removal of template DNA after *in vitro* transcription

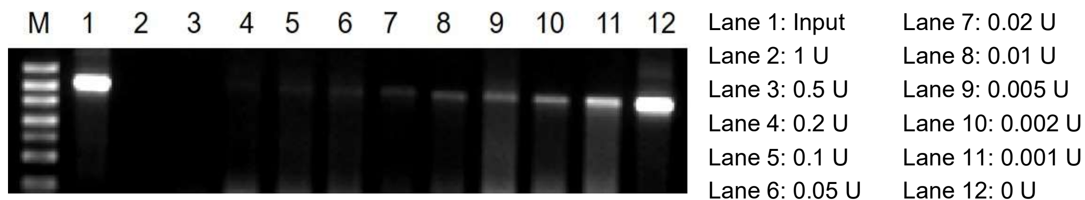
- Add 2 U of DNase I (E00053) to the transcription system for 0.2-1 μg DNA.
- Mix thoroughly, incubate at 37 °C for 30 min.
- Inactivate DNase I by phenol/chloroform extraction.

## Typical protocol for removal of genomic DNA from RNA sample

Assemble the reaction in a nuclease-free microcentrifuge tube or PCR strip tube on ice with the following order:

| Components | Volume |
|---|---|
| DNase I (E00053) | 10 U |
| DNase I 10 × Reaction Buffer | 10 μl (1 ×) |
| RNA | ~10 μg |
| RNase free water | to 100 μl |
| Mix thoroughly, incubate at 37 °C for 30 min. | |
| Add EDTA with a final concentration of 5 mM and heat the solution at 65 °C for 10 min to inactivate DNase I. Alternatively, DNase I can be inactivated by phenol/chloroform extraction. | |
| Use the prepared RNA as the template for RT-PCR. | |

## Data Images




Incubate with 100 ng plasmid DNA at 37 °C for 30 min, tiny amount of GenScript DNase I can completely digest the DNA.

**GenScript USA, Inc.**
860 Centennial Ave. Piscataway, NJ 08854
Tel: 1-732-885-9188

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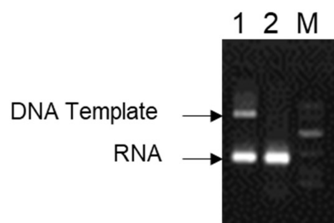


Lane 1: IVT RNA (untreated)
Lane 2: IVT RNA + 2 U DNase I

Add 2 U of DNase I (E00053) to 20 μl *in vitro* transcription system, incubate at 37 °C for 30 min, GenScript DNase I can completely digest the DNA template.

## References


1. Kienzle, N., et al. "DNaseI treatment is a prerequisite for the amplification of cDNA from episomal-based genes." *BioTechniques* 20.4 (1996): 612-616.
2. Anderson, Stephen. "Shotgun DNA sequencing using cloned DNase I-generated fragments." *Nucleic acids research* 9.13 (1981): 3015-3027.
3. Green, Michael R., Tom Maniatis, and D. A. Melton. "Human β-globin pre-mRNA synthesized in vitro is accurately spliced in Xenopus oocyte nuclei." *Cell* 32.3 (1983): 681-694.


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