

## ***RNase-Free DNase I***

Cat. Nos. **D9902K, D9905K, and D9910K**

**RNase-Free DNase I** is an endonuclease that efficiently hydrolyzes double- (ds) or single-stranded DNA to a mixture of short oligo- and mononucleotides. In the presence of  $Mg^{2+}$ , cleavage of each strand of a dsDNA substrate proceeds independently.<sup>1</sup> In contrast, in the presence of  $Mn^{2+}$ , the enzyme cleaves both strands of DNA at approximately the same site to generate molecules with blunt ends or 1- or 2-base overhangs<sup>1</sup> that can be blunted with T4 DNA Polymerase.

EPICENTRE's RNase-Free DNase I from bovine pancreas is the highest quality DNase I commercially available. Provided at a concentration of 1 MBU/ $\mu$ l, RNase-Free DNase I is available in 2,500-, 5,000-, and 10,000-MBU sizes and is suitable for use in each of the following applications:

- Elimination of the DNA template following *in vitro* RNA synthesis with T7, T3, or SP6 Phage RNA Polymerases.
- Characterization of DNA:protein interactions by of "DNase I footprinting".<sup>1,2</sup>
- Treatment of RNA prior to RT-PCR.<sup>3</sup>
- Radiolabeling of DNA by nick translation.<sup>1,4</sup>

### **Product Specifications**

**Storage:** Store only at  $-20^{\circ}C$  in a freezer without a defrost cycle.

**Storage Buffer:** RNase-Free DNase I is supplied in a 50% glycerol solution containing 10 mM Tris-HCl (pH 7.5), 10 mM  $CaCl_2$ , and 10 mM  $MgCl_2$ .

**Unit Definition:** One Molecular Biology Unit (MBU) of RNase-Free DNase I produces an increase in the  $A_{260}$  of a solution of dsDNA, of 0.001 per minute at  $25^{\circ}C$ . Functionally, 1 MBU completely digests 1  $\mu$ g of pUC19 DNA to oligonucleotides in 10 minutes at  $37^{\circ}C$ .

**Quality Control:** RNase-Free DNase I is function-tested in two assay systems. A hyperchromicity assay is performed in a 10-ml reaction containing 50  $\mu$ g/ml native calf thymus DNA, 0.1 M sodium acetate (pH 5.0), 5 mM  $MgCl_2$ , 2 mM  $CaCl_2$ , and varying amounts of enzyme. A digestion assay is performed in a 50- $\mu$ l reaction containing 33 mM Tris-acetate (pH 7.8), 66 mM potassium acetate, 10 mM magnesium acetate, 0.5 mM dithiothreitol, 2 mM  $CaCl_2$ , 1.0  $\mu$ g of pUC19 DNA, and varying amounts of enzyme.

**Contaminating Activity Assays:** RNase-Free DNase I is free of detectable RNase activities as assayed by PAGE analysis of 1  $\mu$ g of a synthetic RNA transcript following incubation with enough RNase-Free DNase I to completely digest 1000  $\mu$ g of DNA.

### **References:**

1. Sambrook, J. *et al.*, (1989) in: *Molecular Cloning: A Laboratory Manual (2nd ed.)*, Cold Spring Harbor Laboratory Press, New York.
2. Galas, D.J. and Schmitz, A. (1978) *Nucl. Acids Res.* **5**, 3157.
3. Kienzle, N. *et al.*, (1996) *BioTechniques* **20**, 612.
4. Rigby, P.W.J. *et al.*, (1977) *J. Mol. Biol.* **113**, 237.

**Related Products:** The following products are also available:

- AmpliScribe<sup>™</sup> T7-Flash<sup>™</sup> Transcription Kits
- AmpliScribe<sup>™</sup> T7, T3, and SP6 High-Yield Transcription Kits
- DuraScribe<sup>®</sup> T7 and SP6 Transcription Kits
- RiboScribe<sup>™</sup> RNA Probe Synthesis Kits
- T7, T3, and SP6 Phage RNA Polymerases
- MasterAmp<sup>™</sup> RT-PCR Kits
- Plasmid-Safe<sup>™</sup> ATP-Dependent DNase
- Exonuclease I
- Exonuclease III
- Mung Bean Nuclease
- OmniCleave<sup>™</sup> Endonuclease

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