

Protocol Cat. No. K1088

APEX BI

# **DNase I (RNase-free)**



DNase I (RNase-free) is an endonuclease that digests single- or double- stranded DNA, generating dinucleotide, trinucleotide, and oligonucleotide products with 5'-phosphorylated and 3'-hydroxylated ends. The activity of DNase I (RNase-free) dependent on Ca<sup>2+</sup> and can be activated by Mg<sup>2+</sup> or Mn<sup>2+</sup>. In the presence of Mg<sup>2+</sup>, DNase I (RNase-free) can randomly cleaves arbitrary sites of double-stranded DNA; in the presence of Mn<sup>2+</sup>, DNase I (RNase-free) can simultaneously recognize both strands of DNA and cleaves at nearly same sites.

DNase I (RNase-free) can act on single stranded DNA, double stranded DNA, chromatin, and RNA: DNA hybrid strands. DNase I (RNase-free) is suitable for RNA extraction, in vitro transcription, and removal of DNA for RT-PCR experiments.

### **Components and Storage**

Components	1000 U	5000 U	10000 U
DNase I (RNase-free) (2 U/µI)	500 µl	500 µl x 5	1 ml x 5
10X DNase I Buffer	1.5 ml	1.5 ml	1.5 ml x 2
Store the components at -20°C.		E Cana	

# Protocol

1. Place the RNase-free PCR tube on ice and prepare the reaction system according to the following table:

Components	100 µl Reaction		
RNA	~10 µg		
10X DNase I Buffer	10 µl		
DNase I (RNase-free)	1 µl		
Nuclease-free H <sub>2</sub> O	To 100 μl		

- 2. Mix thoroughly. Incubate for 10 min at 37°C.
- 3. Add 1 µl of 0.5 M EDTA (final concentration of EDTA is 5 mM).
- 4. Heat inactivation at 75°C for 30min.

## Product Information

#### Unit Definition

One unit is defined as the amount of enzyme required to completely degrade 1 µg of pBR322 DNA in a 100 µl reaction system at 37°C in 10 min. Completed decomposition refers to the degradation of most DNA fragments into tetranucleotides or shorter nucleotides.

#### Storage Buffer

10 mM Tris-HCl (pH 7.6 at 25°C), 2 mM CaCl<sub>2</sub>, 50% Glycerol

#### **Reaction Buffer**

10 mM Tris-HCl (pH 7.6 at 25°C), 2.5 mM MgCl<sub>2</sub>, 0.5 mM CaCl<sub>2</sub>

### Note

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





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