vivantis

PRODUCT INFORMATION
Deoxyribonuclease I (DNase I)

Product: Deoxyribonuclease I (DNase I)

Grade: Ultra Pure Grade

Code: PC0719-50kU; PC0719-100kU

Molecular Weight: 39kDaltons

Concentration: Refer to the product label

Lot No.:

Expiry Date:

Description

DNase I is a recombinant form of DNase I which is RNase-free, originally isolated from bovine pancreas, is a recombinant enzyme expressed in *Pichia pastoris*. It is a DNA-specific endonuclease that hydrolyzes the phosphodiester linkages of double- and single-stranded DNA to a mixture of mono- and oligonucleotides.

DNase I manufactured using state-of-the-art processes yielding animal-free material. The enzyme is highly purified and rigorously tested for contaminating RNase and protease activity of RT-PCR. It is an important tool for all applications requiring DNA-free RNA templates and achieving reliable results with undegraded and stable RNA.

Application:
DNase I used for isolation of DNA-free RNA in diagnostic and therapeutic applications:

- To ensure that RT-PCR templates are free of genomic DNA
- To remove DNA templates after in vitro of RNA

Unit Definition

One unit according to Kunitz produces an increase in absorbance of 0.001/minute under assay conditions in 1ml at 260nm.

Volume activity (calf thymus DNA):	9-14kU/mL
Volume activity (calf thymus DNA, modified buffer system):	No limit
Proteases (up to 50U resorufin-marked casein after 17 hrs; 37°C):	Not detectable
Ribonucleases (up to 10U with MS UU RNA/ 4hrs / 37°C):	Not detectable

Storage Temperature

Store at -15 to -25°C within specification range for 24 months. Avoid exposure to frequent temperature changes. See the expiration date on the stickers of product item.

Storage Buffer

20mM Tris-HCl, 50mM NaCl, 2mM $CaCl_2$, 2mM $MgCl_2$, 50% glycerol and enhancers.
Concentration of stock solutions will vary depending on application.

Dilution Buffer

25mM Tris-HCl pH7.6, 50% glycerol

Recommended 1ml of 10x Reaction Buffer

400mM Tris-HCl, 100mM NaCl, 60mM $MgCl_2$, 10mM $CaCl_2$, pH7.9
$MgCl_2$ in the above buffers may also be substituted with 10mM $MnCl_2$ if it is desired to cleave both strands of DNA at the same site.
Note: DNase I is sensitive to physical denaturation. Gently mix preparations by inversion; do not vortex.

Heat Inactivation

Incubate samples at 75°C for 10 minutes. To protect RNA from being degraded during DNase I inactivation, add EDTA to a final concentration of 5mM.

- Alternatively, DNase I recombinant, RNase-free can be inactivated and removed by phenol extraction according to standard protocol.

Inhibitors

EGTA; EDTA; SDS, salt concentrations>100mM will reduce DNase activity

Suggested Procedure

1. For complete digestion on DNA, prepare the reaction mix below:

Components	Volume
10X Reaction Buffer	2µl
DNase I Solution (1U/µL)	1 – 2U
DNA	1µg
Nuclease-free Water	Top up to 20µl

2. Incubate at 25 – 37°C for 10 minutes.
3. Use the sample for further analysis.

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1. For digestion of genomic DNA in RNA sample, prepare the reaction mix below:

Components	Volume
10X Reaction Buffer	5µl
DNase I Solution (1U/µL)	2 - 10U
Total RNA	10 - 50µg
*Optional: RNase Inhibitor	10U
Nuclease-free Water	Top up to 50µl

2. Incubate at 25 – 37°C for 10 minutes.
3. Stop the reaction by adding 2µl of 0.2M EDTA, pH8.0 to a final concentration of 8mM and heating to 75°C for 10 minutes. The concentration of EDTA has to be taken into account for all subsequent applications.