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ATR-MED®

Product Information

DNase I, RNase-free

Catalogue Number: #36273-1mL

Store at -20 °C

Product Composition

Component of DNase I, RNase-free 1U/µL (#36273)	Volume 1000 U
10X Reaction Buffer with MgCl2 (#79664)	1.25 mL
10X Reaction Buffer without MnCl2 (#79655)	1 mL
100 mM MnCl2 (#66252)	1 mL
50 mM EDTA (#82506)	1 mL

Product Description

ATR-MED's DNase I, RNase-free is an endonuclease that digests single- and double-stranded DNA in the presence of Mg2+ ions. If Mn2+ ions are present, both DNA strands are cleaved at approximately the same site.
It hydrolyzes phosphodiester bonds producing mono- and oligodeoxyribonucleotides with 5'-phosphate and 3'-OH groups.
DNase I attacks each strand of DNA independently and the cleavage sites are random.

Highlights

- Protease – None Detected
- RNase – None detected
- There was no nonspecific nuclease activity
- Breaks down and eliminates unwanted DNA from samples
- Reduces the viscosity of bacterial lysates (protein extracts), making pipetting easier

Applications

- Preparation of DNA-free RNA prior to RT-PCR and RT-qPCR and *in vitro transcription*
- Generation of a library of randomly overlapping DNA inserts.
- Reaction buffer containing Mn2+ is used
- to treat nuclear lysate to obtain single nucleosomes in a study.
- Used for the removal of DNA from protein sample
- RNA purification by removing DNA
- Prepare DNA for nick translation
- Footprinting assays to determine DNA-protein interactions

Product Procedure

Removal of genomic DNA from RNA preparations

1. Add to an RNase-free tube:

RNA	1 µg
10X reaction buffer with MgCl2	1 µL
DNase I, RNase-free (#36273)	1 µL (1 U)
DEPC-treated Water	to 10 µL

2. Incubate at 37 °C for 30 min.
3. Add 1 µL 50 mM EDTA and incubate at 65 °C for 10 min. RNA hydrolyzes during heating with divalent cations in the absence of a chelating agent. Alternatively, use phenol/chloroform extraction.
4. Use the prepared RNA as a template for reverse transcriptase.

Note

- Do not use more than 1 U of DNase I, RNase-free per 1 µg of RNA.
- If using DNase, I, HC, enzyme can be diluted in 1X DNase reaction buffer just prior to use, or in storage buffer (not supplied see composition on reverse page) for longer storage.
- Volumes of the reaction mixture and 50 mM EDTA solution can be scaled up for larger amounts of RNA. The recommended final concentration of RNA is 0.1µg/µL.

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Removal of template DNA after *in vitro* transcription

1. Add 2 U of DNase I, RNase-free per 1 µg of template DNA directly to a transcription reaction mixture. In some cases, the amount of enzyme should be determined empirically.
2. Incubate at 37 °C for 15 minutes.
3. Inactivate DNase I by phenol/chloroform extraction

Molecular Weight

29 kDa monomer.

Definition of Activity Unit

One unit of the enzyme completely degrades 1 µg of plasmid DNA in 10 min at 37 °C.

Enzyme activity is assayed in the following mixture: 10 mM Tris-HCl (pH 7.5 at 25 °C), 2.5 mM $MgCl_2$, 0.1 mM $CaCl_2$, 1 µg of pUC19 DNA. One DNase I unit is equivalent to 0.3 Kunitz unit

Inhibition and Inactivation

- Inhibitors: metal chelators, transition metals (e.g., Zn) in millimolar concentrations, SDS (even at concentrations less than 0.1%), reducing agents (DTT and β-mercaptoethanol), ionic strength above 50-100 mM.
- Inactivated by heating at 65 °C for 10 min in the presence of EGTA or EDTA (use at least 1 mol of EGTA/EDTA per 1 mol of Mn^{2+}/Mg^{2+}

Storage/Stability

Store at -20°C in a tightly sealed container. The shelf life is 12 months.

Shipping

ATR-MED's DNase I, RNase-free is shipped on blue ice.

Precautions and Disclaimer

This product is for R&D use only, not for use in drug, diagnostic procedures, household, or other uses. When working with the product, always wear a suitable lab coat and disposable gloves, protective eyewear. For more information, please consult the appropriate material safety data sheets (MSDSs). These are available online as pdf-file or on request (info@atrmed.com). To the extent allowed by law, ATR-MED Inc. will not be liable for special, incidental, indirect, punitive, multiple, or consequential damages in connection with or arising from this document, including your use of it. By use of this product, you accept all the terms and conditions of ATR-MED products. All trademarks are the property ATR-MED unless otherwise specified.

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