For life science research only.
Not for use in diagnostic procedures.

DNase I recombinant, RNase-free

from bovine pancreas, expressed in *Pichia pastoris*

Version: 05
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Deoxyribonuclease I
Enzyme solution with 10x Incubation Buffer.

Cat. No. 04 716 728 001 10,000 U
10 U/µl

Store the product at −15 to −25°C.

1. General Information

1.1. Contents

Vial / bottle	Label	Function / description	Content
1	DNase I recombinant, RNase-free	Solution, 10 U/µl Enzyme storage buffer: 20 mM Tris-HCl, 50 mM NaCl, 2 mM $CaCl_2$, 2 mM $MgCl_2$, 1 mM DTE, 0.1 mg/ml Pefabloc SC*, 50% glycerol (v/v), pH 7.6 (+4°C).	1 vial, 10,000 U
2	DNase I recombinant, RNase-free, Incubation Buffer, 10x conc.	Incubation Buffer: 400 mM Tris-HCl, 100 mM NaCl, 10 mM $CaCl_2$, 60 mM $MgCl_2$, pH 7.9.	5 vials, 1 ml each

1.2. Storage and Stability

Storage Conditions (Product)

When stored at −15 to −25°C, the product is stable through the expiry date printed on the label.

Vial / bottle	Label	Storage
1	DNase I recombinant, RNase-free	Store at −15 to −25°C.
2	Incubation Buffer, 10x conc.	

1.3. Additional Equipment and Reagent required

For preparation of Enzyme dilution buffer

See section, **Working Solution** *for information on preparing solution.*

- Tris-HCl*
- Glycerol

For digestion of DNA

- Water, RNase-free

For digestion of genomic DNA in RNA samples

- Water, RNase-free
- Protector RNase Inhibitor* (optional)
- 0.2 M EDTA, pH 8.0.

1.4. Application

DNase I recombinant, RNase-free may be used to degrade DNA in applications that are sensitive to the presence of RNase:

- Remove genomic DNA from RNA preparations prior to RT-PCR.
- Isolate DNA-free RNA after *in vitro* transcription reactions.
- Perform nick translations.
- Map DNase-sensitive regions in eukaryotic DNA.

2. How to Use this Product

2.1. Before you Begin

Working Solution

Enzyme dilution buffer: 25 mM Tris-HCl*, 50% glycerol (v/v), pH 7.6 (+4°C).

2.2. Protocols

Digestion of DNA

1. Prepare the following reaction mixture:

Reagent	Volume/Amount
DNA	1 µg
Incubation Buffer, 10x conc.	2 µl
DNase I, recombinant, RNase-free	1 – 2 U
Water, RNase-free	up to 20 µl
Total Volume	**20 µl**

2. Incubate at +25 to +37°C for 10 minutes.

3. Use the sample for further analysis.

Digestion of genomic DNA in RNA samples

1. Prepare the following reaction mixture:

Reagent	Volume/Amount
Total RNA	10 – 50 µg
Incubation Buffer, 10x conc.	5 µl
DNase I, recombinant, RNase-free	2.5 – 10 U
Protector RNase Inhibitor* (optional)	10 U
Water, RNase-free	up to 50 µl
Total Volume	**50 µl**

2. Incubate at +25 to +37°C for 15 to 20 minutes.

3. Stop the reaction by adding 2 µl 0.2 M EDTA, pH 8.0 to a final concentration of 8 mM.
   – Heat to +75°C for 10 minutes.
   The concentration of EDTA must be considered for all subsequent applications.

2.3. Parameters

Activator

Requires divalent cations for maximal activity.

Biological Activity

10×10^3 U/ml

EC-Number

EC 3.1.21.1

Inactivation

One unit DNase I recombinant, RNase-free is heat-inactivated by 10 minutes incubation at +75°C.

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

Molecular Weight

Approximately 39 kD glycoprotein.

Unit Definition

One unit is the enzyme activity that effects an absorbance increase of 0.001/minute under assay conditions in 1 ml at 260 nm.

Assay conditions

Volume activity is determined according to the following assay mixture. 100 µg calf thymus DNA is incubated in 2.5 ml 1x Incubation Buffer with 40 to 70 units DNase I recombinant, RNase-free at +25°C. The absorbance increase is measured at 260 nm.

3. Additional Information on this Product

3.1. Test Principle

Preparation

DNase I recombinant, RNase-free is produced without using any animal cells or other materials derived from animals. DNase I is a DNA-specific endonuclease that hydrolyzes the phosphodiester linkages of double-stranded or single-stranded DNA to a mixture of oligo- and mononucleotides.

3.2. Quality Control

For lot-specific certificates of analysis, see section, **Contact and Support**.

4. Supplementary Information

4.1. Conventions

To make information consistent and easier to read, the following text conventions and symbols are used in this document to highlight important information:

Text convention and symbols	
Information Note: Additional information about the current topic or procedure.	
Important Note: Information critical to the success of the current procedure or use of the product.	
① ② ③ etc.	Stages in a process that usually occur in the order listed.
❶ ❷ ❸ etc.	Steps in a procedure that must be performed in the order listed.
* (Asterisk)	The Asterisk denotes a product available from Roche Diagnostics.

4.2. Changes to previous version

Layout changes.
Editorial changes.

4.3. Ordering Information

Product	Pack Size	Cat. No.
Reagents, kits		
Tris hydrochloride	500 g	10 812 846 001
Protector RNase Inhibitor	custom fill	03 335 429 103

4.4. Trademarks

All product names and trademarks are the property of their respective owners.

4.5. License Disclaimer

For patent license limitations for individual products please refer to:
List of biochemical reagent products.

4.6. Regulatory Disclaimer

For life science research only. Not for use in diagnostic procedures.

4.7. Safety Data Sheet

Please follow the instructions in the Safety Data Sheet (SDS).

4.8. Contact and Support

To ask questions, solve problems, suggest enhancements or report new applications, please visit our **Online Technical Support Site**.

To call, write, fax, or email us, visit **sigma-aldrich.com**, and select your home country. Country-specific contact information will be displayed.

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