DNase I recombinant, grade I
From bovine pancreas expressed in *Pichia pastoris*
Lyophilizate

**Cat. No. 04 536 282 001**
2 × 10,000 Units

1. What this Product Does

**Form**
Lyophilizate 2 × 10,000 Units

**Storage/Stability**
When stored at +2 to +8°C, the enzyme is stable until the expiration date printed on the label.

**Application**
DNase I, recombinant, grade I may be used for:
- eliminating DNA during protein isolation procedures (2)
- analysis of chromatin structure (3)
- eliminating DNA during sample preparation (4)

**Activity**
> 10,000 U/bottle according to Kunitz (25°C; DNA as substrate).

One unit according to Kunitz (1) is the enzyme activity that, under assay conditions, causes an absorbance increase at 260 nm of 0.001/min in 1 ml.

**Quality Control**

**Absence of RNases**
MS2 RNA (5 μg) is incubated with 2 U DNase I for 4 h at 37°C in 25 μl of buffer [25 mM Tris-HCl, 5 mM MgCl₂, 0.1 mM EDTA, pH 7.2].

2 U DNase I does not detectably degrade MS2 RNA during this assay.

**Absence of Proteases**
Resorufin-labeled casein (200 μg) is incubated with 50 U DNase I for 17 h at 37°C in 40 mM Tris-HCl, 4 mM CaCl₂, pH 7.8. The reaction is stopped by addition of trichloroacetic acid (TCA) and then centrifuged. The pH of the supernatant is adjusted with 500 mM Tris-HCl, pH 8.8. The absorbance of the supernatant is then measured at 574 nm to determine if the supernatant contains any resorufin-labeled peptides that were not precipitated by TCA.

In this assay, 50 U DNase I recombinant, grade I, does not produce a detectable signal at 574 nm.

2. How to Use this Product

**Assay Conditions**

Volume activity is determined in 0.1 M sodium acetate, 5 mM MgSO₄, pH 5.0.

Assay mixture: Calf thymus DNA (100 μg) is incubated with 20 – 50 U DNase I at 25°C.

The increase in absorbance is measured at 260 nm.

These conditions are used for the determination of activity according to Kunitz (1) since the Kunitz assay gives optimal reproducibility and sensitivity under these conditions. However, if you are using this enzyme for normal experimental purposes, we recommend using incubation buffers that are appropriate for a given application, e.g., as mentioned in the standard literature (Sambrook et al., *Curr. Prot. Mol. Biol.*, etc.).

**Procedure**

Current Protocols in Molecular Biology (5) suggests the following reaction conditions:

**For 100-μl reaction**
- 50 mM Tris-HCl, pH 7.5
- 10 mM MgCl₂ (for single-strand nicks; to create double-strand breaks, replace with 10 mM MnCl₂)
- 2 μg DNA
- 50 μg/ml BSA

1 μl DNase I (The concentration depends on the application.)

Incubate at 37°C for 1 to 30 min, depending upon the amount of digestion desired. Stop the reaction by adding 5 μl of 0.5 M EDTA. For nick translation, perform the DNase I reaction simultaneously with the DNA polymerase I reaction.

**Long-term Storage of the Dissolved Enzyme**
The solvent generally recommended for DNase I is water. When DNase I is reconstituted in water, the solution can be kept for 2 days at +4°C and for 1 month at −20°C. For best results, prepare appropriate aliquots and avoid repeated freezing and thawing.

However, for long-term storage, carefully dissolve DNase I in one of the buffers listed below. In buffer 1, the enzyme will be stable for several weeks. The enzyme will be stable for at least 18 months if it is dissolved in buffer 2, 3, or 4 and stored at the recommended temperatures.

1) 20 mM Tris-HCl, 20 mM MgCl₂, 5 mM CaCl₂, 0.1 mM dithioerythritol, 0.1 mM EDTA, 50% (v/v) glycerol, pH 8. Store this solution at −20°C; it will not freeze at this temperature and will be stable for several weeks.

2) 20 mM Tris-HCl, 50 mM NaCl, 1 mM dithioerythritol, 100 μg/ml BSA, 50% glycerol (v/v), pH 7.6. Store this solution at −20°C for up to 18 months; it will not freeze at this temperature.

3) 20 mM Tris-HCl, 1 mM MgCl₂, 50% (v/v) glycerol, pH 7.5. Store this solution at −20°C for up to 18 months; it will not freeze at this temperature.

4) 20 mM Tris-HCl, 1 mM MgCl₂, pH 7.5. Aliquot in appropriate amounts (e.g., 10 μl), freeze the aliquots quickly on dry ice, and store at −70°C for up to 18 months. Thaw only the amount needed for each experiment. Do not refreeze. Discard any leftover thawed solution.
3. Additional Information on this Product

3.1 How this Product Works

Background Information

DNase I from bovine pancreas, recombinant in *Pichia pastoris*, is a glycoprotein with a molecular weight of ~39 kDa. The recombinant enzyme is produced without using any animal cells or other materials derived from animals.

DNase I is a DNA-specific endonuclease that hydrolyzes ds or ss DNA to a mixture of oligo- and mononucleotides. The enzyme requires divalent cations for maximal activity (1).

The specificity of the reaction depends on the nature of the cation used. In the presence of Mg²⁺ DNase I causes single-stranded nicks in dsDNA, while in the presence of Mn²⁺ the enzyme produces double-stranded breaks.

References


4. Supplementary Information

Ordering Information

<table>
<thead>
<tr>
<th>Product</th>
<th>Pack size</th>
<th>Cat. No.</th>
</tr>
</thead>
<tbody>
<tr>
<td>DIG RNA Labeling Kit (SP6/T7)</td>
<td>1 kit (2 x 10 labeling reactions)</td>
<td>11 175 025 910</td>
</tr>
<tr>
<td>DIG Northern Starter Kit</td>
<td>10 labeling/detection reactions</td>
<td>12 039 672 910</td>
</tr>
<tr>
<td>High Pure RNA Isolation Kit</td>
<td>1 kit (50 reactions)</td>
<td>11 828 665 001</td>
</tr>
<tr>
<td>High Pure RNA Paraffin Kit</td>
<td>1 kit (100 isolations)</td>
<td>03 270 289 001</td>
</tr>
<tr>
<td>High Pure RNA Tissue Kit</td>
<td>1 kit (50 isolations)</td>
<td>12 033 674 001</td>
</tr>
<tr>
<td>Transcriptor Reverse Transcriptase</td>
<td>2000 U (43 x 500 U) for 200 reactions</td>
<td>03 531 295 001</td>
</tr>
<tr>
<td></td>
<td>500 U for 50 reactions</td>
<td>03 531 295 001</td>
</tr>
<tr>
<td></td>
<td>250 U for 25 reactions</td>
<td>03 531 317 001</td>
</tr>
<tr>
<td>Transcriptor First Strand cDNA Synthesis Kit</td>
<td>50 reactions</td>
<td>04 379 012 001</td>
</tr>
<tr>
<td>Transcriptor High Fidelity cDNA Synthesis Kit</td>
<td>50 reactions</td>
<td>05 081 955 001</td>
</tr>
<tr>
<td>DNA Polymerase I, endonuclease-free</td>
<td>250 units</td>
<td>10 642 711 001</td>
</tr>
<tr>
<td></td>
<td>1,000 units</td>
<td>10 642 720 001</td>
</tr>
<tr>
<td>Protector RNase Inhibitor</td>
<td>10,000 U (5 x 2,000 U) for 2,000 U</td>
<td>03 335 402 001</td>
</tr>
<tr>
<td></td>
<td>2,000 U</td>
<td>03 335 399 001</td>
</tr>
<tr>
<td>Complete</td>
<td>20 tablets</td>
<td>11 697 498 001</td>
</tr>
<tr>
<td></td>
<td>3 x 20 tablets</td>
<td>11 856 145 001</td>
</tr>
<tr>
<td>Tris-HCl</td>
<td>500 g</td>
<td>10 812 846 001</td>
</tr>
<tr>
<td>High Pure FFPE RNA Micro Kit</td>
<td>1 kit (up to 50 isolations)</td>
<td>04 823 125 001</td>
</tr>
<tr>
<td>High Pure PCR Cleanup Micro Kit</td>
<td>1 kit (up to 50 purifications)</td>
<td>04 983 955 001</td>
</tr>
<tr>
<td></td>
<td>1 kit (up to 200 purifications)</td>
<td>04 983 912 001</td>
</tr>
</tbody>
</table>

5. Supplementary Information

Changes to Previous Version

Editorial changes.

Text Conventions

To make information consistent and understandable, the following text conventions are used in this document:

<table>
<thead>
<tr>
<th>Text Convention</th>
<th>Use</th>
</tr>
</thead>
<tbody>
<tr>
<td>Asterisk *</td>
<td>Denotes a product available from Roche Diagnostics</td>
</tr>
</tbody>
</table>

Symbols

Symbols are used in this document to highlight important information:

<table>
<thead>
<tr>
<th>Symbol</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>❧</td>
<td>Information Note: Additional information about the current topic or procedure.</td>
</tr>
</tbody>
</table>

Trademarks

HIGH PURE, COMPLETE, and GENOPURE are trademarks of Roche. All third party product names and trademarks are the property of their respective owners.

Regulatory Disclaimer

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

Disclaimer of License

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

Contact and Support

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