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DNase I ,lyophilized powder

Product Number: RNK4501LY

Shipping and Storage

Freeze dried powder is stored at 4 °C, and after reconstitution, the solution is stored at -20 °C

Components

| Component | RNK4501LY |
|---------------------------------|-----------|
| | 2000U |
| DNase I ,lyophilized powder | 6 |
| Reaction Buffer(with MgCl2),10× | 6×1mL |
| 200 mM EDTA | 6×1mL |
| RNase Free Water | 12×1mL |

Description

DNase I is a deoxyribonuclease that requires a divalent cation and can be used to degrade single stranded or double stranded DNA. The principle is that DNase I hydrolyzes phosphate diester bonds to produce single nucleotides or oligonucleotides with 5 '- phosphate groups and 3' - OH groups. Both Mg^{2+} and Mn^{2+} can activate the activity of DNase I, while Ca^{2+} concentration directly affects enzyme activity. When Mg^{2+} is present, it can randomly generate incisions on each single strand of double stranded DNA; In the presence of Mn^{2+} , double stranded DNA can be broken and fragmented. Used for the preparation of RNA without DNA contamination, reverse transcription, and in vitro transcription experiments.

Preparation and important precautions before the experiment

- 1. Because DNase I is often used in DNA digestion experiments that require maintaining RNA integrity, it minimizes RNase contamination during the enzyme preparation process and can be safely used for RNA extraction. However, as the enzyme does not contain RNase inhibitors, caution should be taken to prevent contamination from exogenous RNase during use.
- 2. DNase I is greatly affected by shear force. Before use, the centrifuge tube can be inverted and mixed evenly to avoid vortex oscillation.
- 3. The dosage of DNase I should not exceed 1U for every 1µg RNA processed.

Protocol

1. Freeze dried powder reconstitution:

Add 1mL RNase Free Water to the freeze-dried powder, invert it upside down to dissolve it, and dilute the DNase I enzyme activity to 2U/µL.

 Taking the preparation of RNA samples for RT-PCR as an example, prepare a 10µL reaction system as shown in the table below:

| Component | Volume | Final concentration |
|---|--------|---------------------|
| RNA | XμL | 1 µg |
| Reation Buffer(with MgCl ₂),10× | 1 µL | $1 \times$ |
| DNaseI(RNase Free) | 0.5 µL | 1 U |
| RNase Free Water | YμL | Up to 10µL |

- 3. Incubate at 37 °C for 15 minutes.
- 4. Add 1 μ L of 200 mM EDTA and incubate at 65 °C for 10 minutes to inactivate DNase I and terminate the reaction.
- 5. The processed RNA can be used for RT-PCR.

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