Recombinant DNase-I

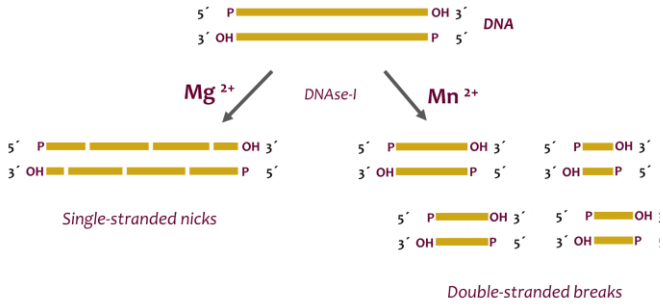
(RNase free)

Ordering info

TBZ0320. Recombinant DNase-I, 1000 units

Description

Recombinant DNase-I is an endonuclease from bovine pancreas produced in *Pichia pastoris*. It is a common reagent used to remove DNA in RNA and protein extraction protocols. The enzyme splits phosphodiester linkages within DNA releasing a mixture of oligo- and mononucleotides. DNase-I acts on single stranded DNA (ssDNA), double stranded DNA (dsDNA) and chromatin. DNase-I generates double-stranded breaks in Mn^{2+} presence, while in presence of Mg^{2+} the enzyme produces single-stranded nicks in dsDNA.



This kit can be used for both DNase digestion in solution as well as "in-column" digestion in combination with Tiaris Biosciences RNA Purification Kits.

Features

- Recombinant protein of bovine pancreas DNase I gene.
- Strictly dependent on Ca^{2+}
- Activated by divalent metals: Mg^{+2} and Mn^{+2}.

Applications

- DNA removal from RNA and protein samples prior downstream applications.
- Molecular evolution by DNA Shuffling.
- DNA Foot printing.
- Digestion of DNA in damaged cells in tissue culture.

Kit Components

Components	TBZ0320
Recombinant DNase-I	500 µL
DNase-I Buffer	5 mL
STOP Solution (EDTA 100 mM)	1 mL

Recombinant DNase-I (2 U/ µL): in 10 mM Tris-HCl (pH 7.5), 2 mM $CaCl_2$, 50% Glycerol (v/v).
DNase-I Buffer: 100 mM Tris-HCl (pH 7.5), 25 mM $MgCl_2$, 1 mM $CaCl_2$.
Recommended concentration of $MnCl_2$ in reaction buffer is 10 mM.

Storage

Store at -20°C.

DNase-I is Stable at room temperature for up to 7 days.

Unit Definition

One unit of the enzyme (Kunitz units) is defined as the amount of enzyme required for the complete degradation of 1 µg of plasmid DNA at 37°C in 10 minutes.

Quality Control

- Specific activity tested.
- Purity determined by SDS-PAGE.
- RNase activity: not detected (incubation of RNA transcript with DNase I).

Material related (not included)

- Water, nuclease free (TBB0300).
- DEPC-treated water (TBB0306).

PROTOCOL

I. Removal of genomic DNA from RNA samples

1. Thawing all components and spin them.
2. Prepare a mix of the following components,

Reaction Components	Volume
Recombinant DNase-I (2 U/ µL)	0.5-1 µL
Sample (RNA)	1 µg
DNase-I Buffer	Up 20 µL

3. Incubate at 37°C for 10-15 minutes.
4. To stop the reaction, add 1 µL STOP Solution and heating up 65-75°C for at least 10 min. If EDTA is not added, the RNA will undergo chemical cleavage when heated up.
5. Alternatively, use a High-Q™ Spin-column RNA purification method to isolate your RNA.