

T5 Exonuclease



Catalog #	3374
Package Size	5,000 units
Volume	500 µl
Concentration	10 units/µl

Description

T5 Exonuclease is a highly efficient 5'→3' exonuclease for either ssDNA or dsDNA. It also has ssDNA specific endonuclease activity in the presence of magnesium ions. However, the enzyme does not degrade supercoiled dsDNA⁽¹⁾. The mode of action of T5 Exonuclease *in vivo* may be analogous to that of the 5'→3' exonuclease activity of *E. coli* DNA Polymerase I^(1,2).

Protein Purity

The physical purity of this enzyme is ≥98% as assessed by SDS-PAGE with Coomassie® blue staining (see figure below).

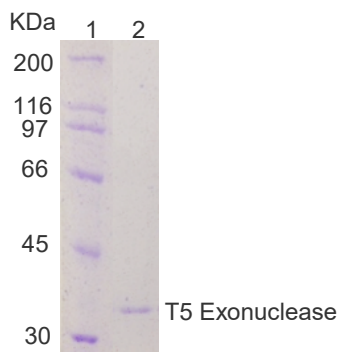


Figure: Lane 1. Protein Marker
Lane 2. T5 Exonuclease

Product Source

E. coli strain expressing the T5 phage D15 gene.

Applications

- Plasmid mutagenesis methods.
- Oligonucleotide site-directed mutagenesis.
- Generation of plasmid-sequencing templates.
- Generation of 3'-overhang for improved cloning procedures.

Storage Temperature

-20 °C

Product Includes

- 1) T5 Exonuclease
- 2) 10x T5 Exonuclease reaction buffer

Storage Buffer

50 mM Tris-HCl
50 mM KCl
1 mM DTT
0.1 mM EDTA
50% Glycerol
pH 7.5 @ 25 °C

10x T5 Exonuclease Reaction Buffer

330 mM Tris-acetate (pH 7.5)
660 mM potassium acetate
100 mM magnesium acetate
5.0 mM DTT

Unit Definition

One unit of T5 Exonuclease catalyzes the release of 1 nmol of acid-soluble nucleotides from double-stranded calf thymus DNA in 30 minutes at 37 °C under standard assay conditions.

Quality Control

T5 Exonuclease is free from detectable RNase or contaminating DNA endonuclease activities.

Protocol

1. Set-up the reaction as follows:

DNA	x µl (up to 1 µg)
10X Buffer	5.0 µl
T5 Exonuclease	1.0 µl
H ₂ O up to	50.0 µl

2. Incubate at 37 °C for 15-30 minutes.
3. Add 10 mM EDTA to stop the reaction.
4. Clean-up treated samples by column purification or phenol/chloroform extraction followed by ethanol precipitation.

References

1. Sayers, J.R. and Eckstein, F. (1990) *J. Biol. Chem.* 265, 18311.
2. Sayers, J.R. et al. (1991). *Nucleic Acids Res.* 19, 4127-4132.

Related Products

- Exonuclease III (Cat.# 3415)
- T4 DNA Ligase (Cat.# 3212)
- Hot Start Taq DNA Polymerase (Cat.# 3293)
- Taq DNA Polymerase 2x Premix (Cat.# 3249)
- Taq DNA Polymerase (Cat.# 3243)

Technical Support

Intact Genomics is committed to supporting the worldwide scientific research community by supplying the highest quality reagents. Each new lot of our products is tested to ensure they meet the quality standards and specifications designated for the product. Please follow the instructions carefully and contact us if additional assistance is needed. We appreciate your business and your feedback regarding the performance of our products in your applications.