

PRODUCT INFORMATION

Lambda Exonuclease

Pub. No. MAN0012006
Rev. Date 03 May 2016 (B.00)

Lot: __ **Expiry Date:** __

Components	#EN0561 1000 U	#EN0562 5000 U
Concentration	10 U/ μ L	10 U/ μ L
10X Reaction Buffer	1 mL	5 x 1 mL

Store at -20 °C

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Description

Lambda Exonuclease is a highly processive 5'→3' exodeoxyribonuclease. It selectively digests the 5'-phosphorylated strand of double-stranded DNA. The enzyme exhibits low activity on single-stranded DNA and non-phosphorylated DNA, and has no activity at nicks and limited activity at gaps in DNA (1, 2).

Applications

- Generating single-stranded PCR products for use in:
 - DNA sequencing (3);
 - analysis of DNA single-strand conformation polymorphism (SSCP) (4);
 - rolling circle amplification.
- Producing single-stranded DNA from double-stranded DNA fragments.
- Cloning of PCR products (5).

Source

E.coli cells with a cloned *exo* gene of phage lambda.

Definition of Activity Unit

One unit of the enzyme catalyzes the release of 10 nmol of acid soluble reaction products from double-stranded substrate in 30 min at 37 °C.

Storage Buffer

The enzyme is supplied in: 25 mM Tris-HCl (pH 8.0), 0.1 mM EDTA, 1 mM DTT, 50 mM NaCl, 0.1% (v/v) Triton X-100 and 50% (v/v) glycerol.

10X Reaction Buffer

670 mM glycine-KOH (pH 9.4), 25 mM MgCl₂, 0.1% (v/v) Triton X-100.

Inhibition and Inactivation

- Inhibitors: salt at relatively low concentration (0.2 M KCl, 0.1 M NaCl), p-chloromercuribenzoate.
- Inactivated by heating at 80 °C for 15 min.

Reaction Conditions (5)

For 50 µL reaction mixture:

10X Reaction Buffer	5 µL
DNA	2 µg
Lambda Exonuclease	10 U

Incubate at 37 °C for 1 to 30 min, depending upon extent of digestion required. Stop reaction by the addition 2 µL of 0.5 M EDTA or by heating at 80 °C for 10 min.

CERTIFICATE OF ANALYSIS

Endodeoxyribonuclease Assay

Incubation of supercoiled plasmid DNA with enzyme.

Functional Assay

Lambda Exonuclease was tested in generation of single-stranded DNA from a PCR product prepared using two primers one of which was 5'-phosphorylated.

Quality authorized by:

 Jurgita Zilinskiene

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References

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3. Higuchi, R.G., Ochman, H., Production of single-stranded DNA templates by exonuclease digestion following the polymerase chain reaction, *Nucleic Acids Res.*, 17, 5865, 1989.
4. Schwieger, F., Tebbe, C.C., A new approach to utilize PCR-single-stranded-conformation polymorphism for 16S rRNA gene-based microbial community analysis, *Appl. Environ. Microbiol.* 64, 4870-4876, 1998.
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