Active Recombinant E.coli Exonuclease III

Cat. No. Exonuclease III-26E **Lot. No.** (See product label)

SPECIFICATION

Product Overview	Exonuclease III is expressed in E.coli and derived from the Exo III gene of E.coli. It can act on dsDNA, gradually releasing 5'-monophosphate nucleotides from the 3'-OH end and generating ssDNA fragments. However, it is inactive against ssDNA and nucleotides linked by phosphorothioate. RNase Activity: ≤10% Endonuclease Activity: ≤10% Nonspecific DNase Activity: Negative Residual gDNA: ≤1 E.coli genome
Species	E.coli
Source	E.coli
Description	The optimal substrates of Exo III are dsDNA with blunt ends or 3'-recessed ends. It can also excise Nicks on dsDNA into Gaps. DsDNA with a 3'-protruding end can prevent Exo III from degrading it, and the degree of resistance depends on the length of the 3'-protruding end. When there are 4 or more protruding bases at the 3'-end, it can completely prevent Exo III from enzymatic cleavage. In addition, Exo III also has 3'-phosphatase activity, RNase H activity and AP-endonuclease activity.
Purity	≥ 95% by SDS-PAGE
Bio-activity	100 U/μL
Unit definition	One unit refers to the amount of enzyme required to catalyze the production of 1nmol of total acid-soluble nucleotides from 0.15 mM [3H]-labeled double-stranded DNA (dsDNA) that has been sonicated within 30 minutes at 37 centigrade in a total reaction volume of 50

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	μL.
Usage	Exonuclease III can effectively degrade nicked dsDNA and linear dsDNA (blunt ends or 5' overhangs) in the 3' to 5' direction, leaving supercoiled dsDNA intact. The following steps can be referred to for the enzymatic digestion reaction: 1. Prepare the reaction system on ice: DNA: Most 5 μg 10× Exo III Reaction Buffer: 5 μL Exonuclease III (E.coli): 0.5 μL (50U) Nuclease-free: Water Up to 50 μL 2. Incubate at 37 centigrade for 30 minutes. 3. Add EDTA to a final concentration of at least 11 mM to terminate the reaction. 4. Incubate at 70 centigrade for 30 minutes to inactivate the enzyme. 5. The following methods can be used for purification. Nucleic acid purification column; DNA gel extraction; After phenol/chloroform purification, precipitate with ethanol.
Applications	1. Non-directional nested deletion 2. Site-directed mutagenesis 3. Strand-specific probe preparation 4. Preparation of single-stranded substrates for dideoxy sequencing
Note	1. Exonuclease III cannot cleave phosphorothioate bonds. However, one end of the DNA molecule can be protected by introducing an α-phosphorothioate nucleotide, thus enabling unidirectional cleavage; 2. To obtain more accurate experimental results, it is recommended to conduct a preliminary experiment on the amount of enzyme used before the experiment; 3. Try to avoid freeze-thaw cycles of this product after receipt; 4. Please wear lab coat and disposable gloves when using; 5. This product should not be used directly for clinical diagnosis and treatment.
Stability	This product should be stored at -20 centigrade and can be stored for at least 24 months.
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Storage Buffer	5mM K3PO4, 200mM KCl, 5mM β-ME, 0.05mM EDTA, 200 μg/ml BSA, 50% Glycerol, @ 25 centigrade, pH 6.5 1X Exo III Reaction Buffer: 10mM Bis-Tris-Propane-HCl, 10mM MgCl2, 1mM DTT, pH 7.0 @ 25 centigrade
Shipping	Dry Ice

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