



Just fine Molecular Biology

Exonuclease III

Description Exonuclease III is a 3'-5' exonuclease specific for double-stranded DNA. The enzyme catalyzes the stepwise removal of mononucleotides starting from a 3'-OH at nicks, blunt ends, recessed ends and 3'-overhangs of less than 4 bases, yielding nucleoside 5'-phosphates. A limited number of nucleotides are removed during each binding event, resulting in progressive deletions within the population of DNA molecules. Exonuclease III activity depends partially on helical structure and displays sequence dependence (C>A=T>G). Temperature, salt concentration and the ratio of enzyme to DNA greatly affect enzyme activity, requiring reaction conditions to be adjusted to specific applications. Exonuclease III degrades DNA from 3'-phosphate ends due to its intrinsic 3'-phosphatase activity. In addition, the enzyme has apurinic endonuclease activity and ribonuclease H activity. Exonuclease III is used in conjunction with S1 nuclease for unidirectional deletion of sequences from the termini of DNA fragments.

Source: Isolated from E.coli strain carrying the gene of the enzyme on a plasmid.

Concentration: 80000 units/ml.

Assay Condition: 50mM Tris-HCl (pH 7,6), 10mM MgCl₂, 1mM DTT, 0,15 mM sonicated pancreatic DNA.

10X Reaction Buffer: 500mM Tris-HCl (pH 7.6 at 30°C), 100mM MgCl₂

Storage Buffer: 10 mM Tris-HCl (pH 7.5); 50 mM KCl; 0,5 mM EDTA; 1 mM 2-mercaptoethanol; 50% glycerol

Storage: store at -20°C

Applications:

- Unidirectional nested deletions
- Site-directed mutagenesis
- Preparation of strand-specific probes
- Preparation of single-stranded substrates for dideoxy sequencing

Unit Definition: One unit is defined as the amount of enzyme required to produce 1 nmoles of acid-soluble nucleotides in 30 minutes at 37°C.

Catalog #	Pack size
405040	4000 u
405200	20000 u

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