T7 Exonuclease



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M0263S **₩** RR **BB 25°** M87







10,000 U/ml 1,000 units Lot: 0021205 RECOMBINANT Store at -20°C Exp: 5/14

Description: T7 Exonuclease acts in the 5' to 3' direction, catalyzing the removal of 5' mononucleotides from duplex DNA. T7 Exonuclease is able to initiate nucleotide removal from the 5' termini or at gaps and nicks of double-stranded DNA (1). It will degrade both 5' phosphorylated or 5´ dephosphorylated DNA. It has been also reported to degrade RNA and DNA from RNA/DNA hybrids in the 5' to 3' direction but is unable to degrade either double-stranded or single-stranded RNA (2). The protein is the product of T7 gene 6.

Source: Purified from an *E. coli* strain containing a TYB12 intein fusion

Supplied in: 10 mM Tris-HCI (pH 8.0), 0.1 mM EDTA, 5 mM DTT and 50% glycerol.

Reagents Supplied with Enzyme:

10X NEBuffer 4.

Reaction Conditions: 1X NEBuffer 4. Incubate at 25°C.

1X NEBuffer 4:

50 mM potassium acetate 20 mM Tris-acetate 10 mM magnesium acetate 1 mM DTT pH 7.9 @ 25°C

Unit Definition: One unit is defined as the amount of enzyme to produce 1 nmol of acid soluble deoxyribonucleotide from double-stranded DNA in a total reaction volume of 50 ul in 30 minutes at 25 °C

Unit Assay Conditions: 50 mM potassium acetate, 20 mM Tris-acetate, 10 mM magnesium acetate, 1mM dithiothreitol (pH 7.9) and 0.15 mM sonicated duplex [3H] DNA.

Quality Control Assays

SS DNA Exonuclease Activity: Incubation of 10 units of enzyme with 1 µg sonicated and denatured [3H]-DNA (105 cpm/µg) for 30 minutes at 25°C in 50 ul reaction buffer released < 1.5% radioactivity.

Endonuclease Activity: Incubation of 400 units of enzyme with 1 µg ϕ X174 RF I DNA for 1 hour at 25°C in 50 µl 1X reaction buffer resulted in < 10% conversion to RF II.

RNase Activity (RNA/DNA Hybrid): Incubation of 10 units of enzyme with 15.2 nmol [3H]poly(A)-poly(dT) hybrid polymer for 1 hour at 37°C in a 50 µl reaction buffer released 9.7 nmol adenosine - 5'-monophosphate.

Heat Inactivation: No

References:

- 1. Kerr, C. and Sadowski, P. D. (1972) J. Biol. Chem. 247. 305-318.
- 2. Shinozaki, K. and Okazaki, T. (1978) Nucleic Acids Res. 5, 4245-4261.

CERTIFICATE OF ANALYSIS

T7



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Exonuclease

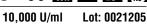
M0263S **₩** RR MEA 25° M67

1,000 units









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