

Therminator™ γ DNA Polymerase



1-800-632-7799
info@neb.com
www.neb.com



M0334S 002120914091

M0334S



200 units 2,000 U/ml Lot: 0021209

RECOMBINANT Store at -20°C Exp: 9/14

Description: Therminator γ (gamma) DNA Polymerase is a 9°N™ DNA polymerase variant with an enhanced ability to incorporate gamma-phosphate labeled dNTPs.

Source: Therminator γ DNA Polymerase is purified from a strain of *E. coli* that carries the 9°N (D141A / E143A / W355A / L408W / R460A / Q461S / K464E / D480V / R484W / A485L) DNA Polymerase gene, a genetically engineered form of the native DNA polymerase from *Thermococcus* species 9°N-7. Developed by LI-COR® Biosciences and derived from Therminator.

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Supplied in: 300 mM KCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM dithiothreitol and 50% glycerol.

Applications:

- Incorporation of modified nucleotides into DNA.
- DNA sequencing using gamma-phosphate modified nucleotides.

Reagents Supplied with Enzyme:
10X Therminator γ Reaction Buffer.

Reaction Conditions: 1X Therminator γ Reaction Buffer, DNA template, primer, 200 μ M dNTPs and 0.5–2 units of Therminator γ DNA Polymerase in a total reaction volume of 100 μ l.

1X Therminator γ Reaction Buffer:

50 mM KCl
20 mM Tris-HCl
5 mM MgSO₄
0.02% IGEPAL® CA-630
(pH 9.2 @ 25°C)

Supplied in: 300 mM KCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM dithiothreitol and 50% glycerol.

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Unit Definition: One unit is defined as the amount of enzyme that will incorporate 10 nmol of dNTPs into acid insoluble material in 30 minutes at 75°C.

Unit Assay Conditions: 1X Therminator γ Reaction Buffer, 200 μ M dNTPs including [³H]-dTTP and 15 nM primed M13mp18.

Enzyme Properties:

3' → 5' Exonuclease: No
5' → 3' Exonuclease: No
Strand Displacement: Yes

Molecular Weight: 89,580 Daltons (theoretical)

Specific Activity: 8,500 units/mg

Quality Control Assays

Exonuclease Activity: Incubation of a 50 μ l reaction in Therminator γ Reaction Buffer containing a minimum of 20 units of Therminator γ DNA Polymerase and 1 μ g of a mixture of single and double-stranded [³H] *E. coli* DNA for 4 hours at 37°C releases < 0.1% of the total radioactivity.

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Endonuclease Activity: Incubation of a 50 μ l reaction in Therminator γ Reaction Buffer containing a minimum of 20 units of Therminator γ DNA Polymerase with 1 μ g of supercoiled ϕ X174 DNA for 4 hours at either 37°C or 75°C results in < 10% conversion to the nicked form as determined by agarose gel electrophoresis.

Companion Products:

Therminator γ Reaction Buffer
#B0334S 6 ml

Deoxynucleotide Solution Set
#N0446S 25 μ mol of each

Deoxynucleotide Solution Mix
#N0447S 8 μ mol each
#N0447L 40 μ mol each

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CERTIFICATE OF ANALYSIS

Endonuclease Activity: Incubation of a 50 μ l reaction in Therminator γ Reaction Buffer containing a minimum of 20 units of Therminator γ DNA Polymerase with 1 μ g of supercoiled ϕ X174 DNA for 4 hours at either 37°C or 75°C results in < 10% conversion to the nicked form as determined by agarose gel electrophoresis.

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