

## Therminator™ DNA Polymerase



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



M0261S 018140916091

# M0261S



200 units 2,000 U/ml Lot: 0181409

RECOMBINANT Store at -20°C Exp: 9/16

**Description:** Therminator DNA Polymerase is a 9°N™ DNA Polymerase variant with an enhanced ability to incorporate modified substrates such as dideoxynucleotides, ribonucleotides and acyclo-nucleotides (1,2).

**Source:** An *E. coli* strain that carries the 9°N (D141A / E143A / A485L) DNA Polymerase gene, a genetically engineered form of the native DNA polymerase from *Thermococcus species* 9°N-7.

Supplied in: 100 mM KCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM dithiothreitol and 50% glycerol.

### Applications:

- DNA sequencing by partial ribosubstitution (3)
- DNA sequencing using dideoxy (4) or acyclo (5) chain terminators
- SNP analysis with dideoxy or acyclo chain terminators (6)

### Reagents Supplied with Enzyme:

10X ThermoPol® Reaction Buffer.

**Reaction Conditions:** 1X ThermoPol Reaction Buffer, DNA template, primer, 200 µM dNTPs and 0.5–2 units of Therminator DNA polymerase in a total reaction volume of 100 µl.

### 1X ThermoPol Reaction Buffer:

20 mM Tris-HCl  
10 mM (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>  
10 mM KCl  
2 mM MgSO<sub>4</sub>  
0.1% Triton® X-100  
pH 8.8 @ 25°C

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

**Unit Assay Conditions:** 1X ThermoPol Reaction Buffer, 200 µM dNTPs including [<sup>3</sup>H]-dTTP and 15 nM primed single-stranded M13mp18.

**Heat Inactivation:** No

### Quality Control Assays

**Exonuclease Activity:** Incubation of a 50 µl reaction in ThermoPol Reaction Buffer containing a minimum of 20 units of Therminator DNA Polymerase and 1 µg of a mixture of single and double-stranded [<sup>3</sup>H] *E. coli* DNA for 4 hours at 75°C releases < 0.1% of the total radioactivity.

**Endonuclease Activity:** Incubation of a 50 µl reaction in ThermoPol Reaction Buffer containing a minimum of 20 units of Therminator DNA Polymerase with 1 µg of supercoiled φX174 DNA for 4 hours at 37°C results in < 10% conversion to the nicked form as determined by agarose gel electrophoresis.

### References:

1. Gardner, A. F. and Jack, W. E. (1999) *Nucleic Acids Research* 27, 2545–2555.
2. Gardner, A. F. and Jack, W. E. (2002) *Nucleic Acids Research* 30, 605–613.
3. Barnes, W. F. (1978) *J. Mol. Biol.* 119, 83–99.
4. Sanger, F., Nicklen, S. and Coulson, A. R. (1977) *Proc. Natl. Acad. Sci. USA*, 74, 5463–5467.
5. Trainor, G. L. (1996) U.S. Patent # 5, 558, 991.
6. Haff, L. A. and Simirnov, I. P. (1997) *Genome Methods* 7, 378–388.

### Companion Products Sold Separately:

Magnesium Sulfate (MgSO<sub>4</sub>) Solution  
#B1003S 6.0 ml

ThermoPol Reaction Buffer Pack  
#B9004S 6.0 ml

ThermoPol II (Mg-free) Reaction Buffer Pack  
#B9005S 6.0 ml

ThermoPol DF (Detergent-free) Reaction Buffer Pack  
#B9013S 6.0 ml

(see other side)

CERTIFICATE OF ANALYSIS

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#B9005S 6.0 ml

ThermoPol DF (Detergent-free) Reaction Buffer Pack  
#B9013S 6.0 ml

(see other side)

CERTIFICATE OF ANALYSIS

Deoxynucleotide Solution Set  
#N0446S 25 µmol each

Deoxynucleotide Solution Mix  
#N0447S 8 µmol each  
#N0447L 40 µmol each

Acyclonucleotide Set  
#N0460S 0.5 µmol each



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TRITON® is a registered trademark of Union Carbide Corporation.

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Deoxynucleotide Solution Mix  
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#N0447L 40 µmol each

Acyclonucleotide Set  
#N0460S 0.5 µmol each



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