

GenePharmaTM SBS Polymerase, Terminator III

Catalog: J01001-500

J01001-1000

Store at -20°C

Description:

GenePharmaTM SBS Polymerase, Terminator III is a 9°N DNA Polymerase variant with an enhanced ability to incorporate modified nucleotides such as 3'-amino-dNTPs, 3'-azido-dNTPs and other nucleotide analogs modified at the 3' ribose position.

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

Supplied in: 10mM Tris-HCl (pH7.5 @ 25°C), 100 mM KCl,1 mM dithiothreitol, 0.1 mM EDTA, 50% glycerol.

Applications:

•Incorporation of nucleotide analogs with 3'- OH substitutions.

•DNA sequencing using 3'-azido-ddNTP chain terminators.

•SNP analysis with 3'-azido-ddNTP chain terminators.

•DNA labeling with 3'-amino-ddNTP.

Reagents Supplied with Enzyme: 10×Terminator III Buffer.

Reaction Conditions: 1×Terminator III Buffer, DNA template, primer, 200 μ M dNTPs and 0.5–2units of GenePharmaTM SBS Polymerase, Terminator III in a total reaction volume of 100 μ l.

1×Terminator III Buffer:

20 mM Tris-HCl; 10 mM (NH₄)₂SO₄; 10 mM KCl; 2 mM MgSO₄; 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 dNTPs into acid insoluble material in 30 minutes at 75°C.

Heat Inactivation: No

Enzyme Properties: $3' \rightarrow 5'$ Exonuclease: No $5' \rightarrow 3'$ Exonuclease: No Strand Displacement: Yes

Molecular Weight: 90,000 Daltons.

Quality Control Assays

Exonuclease Activity: Incubation of a 20µl reaction in 1×Terminator III Buffer containing a minimum 2 unit SBS Polymerase, Terminator III with 1µg human genomic DNA for 1 hour at either 37°C or 75°C without obvious degrade band by agarose gel electrophoresis releases.

Endonuclease Activity: Incubation of a 20 μ l reaction in 1×Terminator III Buffer containing a minimum 2 unit SBS Polymerase, Terminator III with 1 μ g of supercoiled PUC57 DNA for 1hour at either 37°C or 75°C results in < 10% conversion to the nicked form as determined by agarose gel electrophoresis.

