GIBCOBRL

LIFE TECHNOLOGIES™

Terminal Deoxynucleotidyl Transferase Recombinant

Cat. No. 10533-065
Lot No. _____ 500 units; 15 U/µl
Exp. Date: _____. Store at -20°C (not frost-free).

Description:
Terminal Deoxynucleotidyl Transferase, recombinant (rTdT) is purified from a baculovirus clone. rTdT is a DNA polymerase that catalyzes the addition of dNTPs to the 3' hydroxyl terminus of the DNA. Protruding, recessed, or blunt-ended double or single-stranded DNA molecules with chain lengths of three or more nucleotides serve as a substrate for TdT. This enzyme can be used for labeling the 3' ends of DNA with cordycepin triphosphate or for adding homopolymer tails to the 3' ends of DNA.

Components:

10533-065	rTdT	Lot No.
Y95102	5X TdT Buffer	Lot No.

Unit Definition:
One unit incorporates 1 nmol dATP into acid-precipitable material in 1 hour at 37°C, using $d(pA)_{50}$ as a primer.

5X TdT Buffer:	Storage Buffer:
0.5 M potassium cacodylate (pH 7.2)	0.1 M potassium phosphate (pH 7.2)
10 mM $CoCl_2$	200 mM KCl
1 mM DTT	1 mM 2-mercaptoethanol
	50% (v/v) glycerol

Store 5X TdT Buffer at -20°C.

Note: 5X TdT Buffer contains cobalt chloride and potassium cacodylate. See Material Safety Data Sheet.

The 5X TdT Buffer may appear pink due to the cobalt chloride. It is recommended to avoid radioisotopes supplied in ≥ 1 mM DTT. The increased concentration of DTT from the radioisotope can cause the DTT to react with the cobalt chloride in the 5X TdT buffer causing a precipitate to form.

Doc. Rev.: 110100

Quality Control Assays:
This product has passed the following quality control assays: absence of detectable endonuclease, exonuclease, and phosphatase activities; performance in G-tailing; addition of 20-25 pmol of dGMP per pmol of 3' terminus using *Pst* I-cut pBR322 DNA.

The enclosed buffers were assayed with the enzyme and met quality control specifications.

Tailing Assay on Blunt-end DNA Fragments:
40 µl 5X TdT Buffer
5 pmol 3'ends (1.6 µg *Hae* III-cut λ DNA)
1 mM [^{3}H]dATP
30 units rTdT
Reaction Volume: 200 µl
Incubation: 0.5 to 1 hour at 37°C.

Tailing Assay on *Pst* I-cut pBR322 DNA:
10 µl 5X TdT Buffer
2 pmol 3'ends (3 µg *Pst* I-cut pBR322 DNA)
5-50 µM dGTP
2 µl [^{3}H]dGTP (5-20 Ci/mmol; 1 mCi/ml)
15 units rTdT
Reaction Volume: 50 µl
Incubation: 30 minutes at 37°C.

From quality control data, Life Technologies extrapolates the concentration of dGTP required for the addition of 20-25 dG residues/3' end. Under the above conditions, _____ µM gave the desired result.