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Terminal Deoxynucleotidyl Transferase (20 U/μL)

Product description

Terminal Deoxynucleotidyl Transferase (TdT) is a template-independent DNA polymerase that catalyzes the repetitive addition of deoxynucleotides to the 3'-hydroxyl termini of oligonucleotides, single-stranded, and double-stranded DNA. The TdT reaction requires short sequences of at least three bases to act as primers. When using RNA as a substrate, TdT activity is highly dependent on the tertiary structure and nucleotide composition at the 3'-end of the acceptor RNA. In general, TdT exhibits lower efficiency with RNA templates compared to DNA templates.

Specifications

Cat. No.	10302ES76 / 10302ES86 / 10302ES99 / 10302ES98
Size	500 U / 2,500 U / 180,000 U / 333,000 U
Concentration	20 U/μL
Source	Recombinant expression in <i>E.coli</i>
Unit Definition	One unit (U) is defined as the amount of enzyme required to incorporate 1 nmol of deoxyribonucleotide into polydeoxynucleotides in 60 minutes at 37°C.
Inhibitors	Metal chelators, ammonium, chloride, iodide, and phosphate ions.
Inactivation	Add EDTA and heat at 70°C for 10 min.

Components

Components No.	Name	10302ES76	10302ES86	10302ES99	10302ES98
10302-A	Terminal Deoxynucleotidyl Transferase (20 U/µL)	25 μL	125 μL	9 mL	16.65 mL
10302-B	5× Reaction Buffer	400 μL	2×1 mL	40 mL	74 mL

Storage

This product should be stored at -25~-15°C for 1 year.

Instructions

DNA 3' Tailing Protocol

1. Prepare the following reaction mixture and mix thoroughly (for N-nucleotide tailing):

Component	Amount	
5× Reaction Buffer	4 μL	
DNA fragments	1 pmol of 3'-ends	
dATP or dTTP	~1.1N pmol	
dGTP or dCTP	~2N pmol	
Terminal Deoxynucleotidyl Transferase (20 U/μL)	25–40 U	

www. yeasenbio.com Page 1 of 2



Component	Amount
H ₂ O	to 20 μL

- 2. Incubate at 22-37 °C for 15-30 minutes.
- 3. Terminate the reaction by either heating at 70 °C for 10 minutes or adding 2 μ L of 0.5 M EDTA solution.

DNA and Oligonucleotide 3' End Labeling Protocol

1. Prepare the following reaction mixture and mix thoroughly:

Component	Amount
5× Reaction Buffer	10 μL
Linear DNA	10 pmol
Radiolabelled ddATP (10 TBq/mmol)	1–2 MBq
Terminal Deoxynucleotidyl Transferase (20 U/μL)	40 U
H₂O	to 50 μL

- 2. Incubate at 37 °C for 15 minutes.
- 3. Terminate the reaction by either heating at 70 °C for 10 minutes or adding 2 μ L of 0.5 M EDTA solution.

Notes

- 1. The 5× Reaction Buffer contains CoCl₂, which may not be compatible with downstream applications. It is recommended to purify the reaction mixture by spin column chromatography, phenol/chloroform extraction followed by ethanol precipitation to remove CoCl₂.
- 2. This product is intended for research use only.
- 3. For your safety and health, wear a lab coat and disposable gloves when handling this reagent.

www. yeasenbio.com Page 2 of 2