

Technical Datasheet

TERMINAL DEOXYNUCLEOTIDYL TRANSFERASE

(Calf Thymus)

Catalogue No:	CA-1390-01 CA-1390-02	Pack Size:	300U 1500U
Specific Activity:	_____ U/mg	Lot No:	_____
Protein (Bradford):	_____ mg/ml	Concentration:	_____ U/ μ l

Unit Definition: One unit is the amount of enzyme required to transfer 1nmol of dAMP from dATP to the 3'-OH terminus of the oligonucleotide initiator d(A)₅₀ in 1 hour at 37°C.

Assay Conditions: 100mM K-Cacodylate, (pH 7.2)
0.1mM DTT
1mM CoCl₂
8mM MgCl₂
16.67 μ M oligonucleotide d(A)₁₅
2.5mM dATP
0.4 μ Ci [³²P] dATP
Reaction volume 60 μ l.

Storage Buffer: 50mM Kpi (pH 7.4)
1mM BME
50% glycerol (w/v)

Storage Conditions: Store at -20°C.

Important Note: All reactions should be run in polypropylene tubes.



QUALITY CONTROL

Endonuclease: incubation of 5, 10 and 20U of enzyme with 1.0µg pBR322 DNA at 37°C for 1 hour in a reaction volume of 10µl resulted in <10% conversion of RFI to RFII DNA.

3'-Exonuclease: incubation of 12, 24 and 48U of enzyme with 0.13µg 3'-ends of λ/Taq I fragments (3'-labelled with T4 DNA Polymerase [³H]dCPT), incubated for 1 hour at 37°C in a reaction volume of 50µl resulted in a 0.017 slope of %-end label released per unit of enzyme.

5'-Exonuclease: incubation of 12, 24 and 48U of enzyme with 0.05µg of 5'-ends of [5' ³³P] λ/Hae III DNA fragments for 1 hour at 37°C in a reaction volume of 50µl resulted in a 0.259 slope of %-end label released per unit of enzyme.

DNase, double-stranded: incubation of 12, 24 and 48U of enzyme with 0.015µg [³²P] λ DNA for 1 hr at 37°C in a reaction volume of 20µl resulted in a slope of 0.005 for %-end label released per unit of enzyme.

DNase, single-stranded: incubation of 12, 24 and 48U of enzyme with 0.015µg [³²P] λ DNA for 1 hour at 37°C in a reaction volume of 20µl resulted in a slope of 0.047 for %-end label released per unit of enzyme.

RNase: incubation of 12, 24 and 48U of enzyme with 0.015µg of [³²P] transcript pPVI/Pvu II for 1 hour at 37°C in a reaction volume of 50µl resulted in a slope of 0. for %-end label released per unit of enzyme.