



CERTIFICATE OF ANALYSIS

T4 Polynucleotide Kinase



Source:

An *E.coli* strain that carries the cloned Polynucleotide kinase gene from bacteriophage T4.

T4 polynucleotide Kinase catalyzes the transfer and exchange of Pi from the γ position of ATP to the 5' hydroxyl terminus of polynucleotides (double- and single-stranded DNA and RNA) and nucleoside 3' - monophosphates. The enzyme also catalyzes the removal of 3' - phosphoryl groups from 3' - phosphoril polynucleotides, deoxynucleoside 3' - monophosphates and deoxynucleoside 3' - diphosphates.

Storage Conditions:

10 mM Tris-HCl (pH 7.5), 50 mM NaCl, 0.1 mM EDTA, 50% glycerol, 1mM DTT.
Store at - 20^o C.

1X SE-T4 Polynucleotide Kinase Buffer:

50 mM Tris-HCl, (pH 7.6@ 25^o C), 10 mM MgCl₂, 5 mM DTT.

Applications:

- end-labeling DNA or RNA for probes and DNA sequencing;
- addition of 5' - phosphates to oligonucleotides to allow subsequent ligation;
- removal of 3' - phosphoril groups.

Unit Definition:

One unit is defined as the amount of enzyme catalyzing the incorporation of 1 nmol of acid-insoluble [³²P] in 30 minutes at 37^oC.

Unit Assay Conditions:

1 x T4 Polynucleotide Kinase Buffer, 66 μ M [γ -³²P] ATP (5 x 10⁶ cpm/ μ mol) and 0.26 mM 5' - hydroxyl - terminated salmon sperm DNA.

Quality Control Assays:

Nonspecific endonuclease assay:

No appearance of nicked DNA was detected after incubation of 1 μ g supercoiled pUC19 DNA with 10 units of enzyme for 4 hours at 37^o C.

No alteration of the pattern of DNA bands was detected after incubation of 1 μ g λ /HindIII DNA fragments with 10 units of enzyme in 50 μ l of reaction mixture for 16 hours at 37^o C.

Test for detection of RNase contaminants:

No fluorescence increasing was detected after incubation of 10 units of enzyme with 0.1 mM of fluorescent-labeled deoxyribooligonucleotide including ribonucleotide for 30 min at 37^o C.

(B.R.Kelemen, T.A. Klink, M.A. Behlke, S.R.Eubanks, P.A. Leland, R.T. Raines. 1999. *Nucleic Acids Res.*, 27, P. 3696-3701).

Reagents Supplied with Enzyme:

10X SE-T4 Polynucleotide Kinase Buffer.

S E311
500 units
10,000 u/ml

Lot: 24

Exp: 07/20

Store at -20C

For more details
scan the code



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