



T4 Polynucleotide Kinase

Code No. PNK-111

Lot No. *****

Storage Store at -20°C

Size 1,500units

- Source : *Escherichia coli* JM109 which is carrying T4 pse T gene.
- Reaction : $(\gamma\text{-}^{32}\text{P})\text{-ATP} + 5'\text{-OH-polynucleotide} \rightarrow \text{ADP} + (5'\text{-}^{32}\text{P})\text{polynucleotide}$
- Concentration : *** units/ μl
- Unit Definition : One unit is the amount of enzyme activity that incorporates 1 nmole of $(\gamma\text{-}^{32}\text{P})\text{-ATP}$ into micrococcal nuclease-treated calf thymus DNA in 30 minutes at 37°C.
- Assay Condition : 50 mM Tris-HCl(pH7.6)
100 μM $(\gamma\text{-}^{32}\text{P})\text{-ATP}$
10 mM MgCl_2
10 mM 2-mercaptoethanol
0.2 mg/ml micrococcal nuclease-treated calf thymus DNA
- Storage Buffer : 50 mM Tris-HCl(pH7.5)
1 mM DTT
0.1 mM EDTA
50 mM KCl
0.1 μM ATP
50 % Glycerol
- 10 × protruding end kinase buffer : 0.5 M Tris-HCl(pH8.0)
0.1 M MgCl_2
50 mM DTT
- 10 × blunt end or recessed end kinase buffer : 0.5 M Tris-HCl(pH9.5)
0.1 M MgCl_2
50 mM DTT
- Denaturation Buffer : 20 mM Tris-HCl(pH9.5)
1 mM Spermidine
0.1 mM EDTA
- Contaminant Assay
1. Nonspecific Endonuclease : When 10 units of enzyme were incubated with 1 μg of *Hind*III digest of λ -DNA for 20 hours at 37°C in 50 μl reaction volume, no degradation of the DNA fragments is observed after agarose gel electrophoresis.
2. Nonspecific Exonuclease : 30 units of enzyme, when incubated with 1 μg of *E. coli* ^3H -DNA for 4 hours at 37°C in 50 μl reaction volume, will release less than 0.01 % acid soluble counts.
3. Nicking Activity : After incubation of 1 μg of $\Phi\text{X174DNA(RFI)}$ with 25 units of this enzyme for 4 hours at 37°C in 50 μl reaction volume, the supercoiled structure is observed after agarose gel electrophoresis.



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Inspiration for Life Science

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