Tinzyme Co., Limited

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T4 Polynucleotide Kinase

Product Number: T4K01

Shipping and Storage

-20°C.

Components

Components	Volume
T4 PNK（10U/μl）	20μl
10mM ATP	50μl
10×T4 PNK Reaction Buffer	1ml

Description

T4 PNK is a polynucleotide 5 'hydroxy kinase that catalyzes ATP γ- Transfer and exchange of phosphonic acid to double stranded/single stranded DNA or RNA, as well as single nucleotides with 3 'phosphate groups at 5' - hydroxyl groups: 5 '- OH+NTP5' ↔ P+NDP; This enzyme also has 3 '- phosphatase activity, which hydrolyzes the 3' - phosphate group from the 3 '- phosphate end of the oligonucleotide, deoxygenated 3' - monophosphate nucleoside, and deoxygenated 3 '- diphosphate nucleoside.

Application

1. Phosphorylation of the 5 'end of primers or PCR products for linkage reactions.
2. Phosphorylation of the 5 'end of the synthesized DNA connector (Linker) for connection reaction.
3. Labeling of the 5 'end of DNA and RNA, used as an oligonucleotide probe.

Unit Definition

The amount of enzyme required to mix 1 nmol of [γ-32P]ATP with acid insoluble precipitate is defined as 1 active unit, using Micrococcal Nuclease treated calf thymus DNA as substrate, at 37 ℃, pH 7.6, within 30 minutes.

Quality control

After multiple column purification, only a clear and single target band was visible in SDS-PAGE gel detection. The PCR method detected no residual Escherichia coli DNA, and no contamination of nucleic acid endonucleases, exonucleases, phosphatases, and RNA enzyme activities.

Suggestions

1. 1×T4 PNK reaction buffer does not contain ATP, and ATP with a final concentration of 1mM needs to be added to the reaction system. Alternatively, the T4 DNA ligase reaction solution can be used instead.
2. Ammonium ions strongly inhibit the activity of T4 PNK, so DNA should be dissolved in a solution without ammonium salts.
3. DTT oxidation can cause a decrease in enzyme activity. When the buffer is not fresh, additional DTT needs to be added.