

Manual

Thermo PNK

Thermostable polynucleotide kinase. Concentration 10 U/μl.

catalog #	size
1016-50	500 U

For research use only.

Guarantee

A&A Biotechnology provides guarantee on this product.

The company does not guarantee correct performance of this kit in the event of:

- not adhering to the supplied protocol
- use of not recommended equipment or materials
- use of other reagents than recommended or which are not a component of the product
- use of expired or improperly stored product or its components

Description

Thermo PNK is a recombined enzyme produced by the *E.coli* strain carrying a plasmid with a cloned gene encoding of thermostable polynucleotide kinase from *Pyrococcus woesei*. Enzyme is thermostable and exhibits maximum activity at 70-75 °C. The activity is 50% inhibited when a concentration of salt ions exceeds 100 mM. The presence of dNTPs does not reduce the activity of this enzyme.

Application

- enzyme catalyzes transfer of the terminal phosphate group of ATP to the 5'-hydroxyl terminus of DNA or RNA.
- in low pH it is also capable of removing 3'-phosphates from RNA or DNA without altering their 5' ends
- labeling of DNA and RNA probes for hybridization
- phosphorylation of PCR primers and PCR products
- synthesis of genes on the inserted complementary oligonucleotides

Contents

	1016-50	storage
PNK kinase	500 U (10 U/μl)	-20 °C
10 mM ATP	1 x 200 μl	-20 °C
Thermo PNK buffer	1 x 1.25 ml	-20 °C
10x Thermo PNK reaction buffer: 100 mM Tris-HCl, pH 8,8, 100 mM KCl, 100 mM (NH ₄) ₂ SO ₄ , 25 mM MgCl ₂ , 1% Triton X-100		
ultrapure water	1 x 1.5 ml	-20 °C – +25 °C

DNA phosphorylation kinase

1. Thaw and mix all components and add:

component	reaction volume
	20 µl
ds DNA or oligonucleotide (probe)	0.2-2 µg 5 µM
Thermo PNK buffer	2 µl
10 mM ATP	2 µl
PNK kinase	10 U
ultrapure water	up to 20 µl

2. Incubate for 5-30 min at temp. 70-75 °C.

Notes

- To remove Thermo PNK activity it's recommended using Clean-Up kits (np. # 021-50, 021-250).
- Phosphorylated DNA fragments can be purified using Gel-Out kits (np. # 023-50, 023-250).
- In the presence of RNA and DNA, Thermo PNK has a higher affinity to catalyze the phosphorylation reaction of the 5' end of RNA.
- Thermo PNK can be used with thermostable ligases during *in vitro* gene synthesis reactions based on complementary oligonucleotides.



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