Bio Academia
Manufacturer

AS ONE

Pfu DNA Polymerase (with dNTPs), Economy

02-021 200 U 2.5 U/µl # 02-021-05 5 x 200 U

Pyrococcus furiosus DNA polymerase (*Pfu* DNA polymerase) gene was expressed in *E. coli* in large quantities and highly purified. The enzyme has thermostable DNA polymerase activity and 3' →5' exonuclease (proofreading) activity. The MW is 90 kDa, same as that of the natural *Pfu* DNA polymerase.

- *Pfu* DNA polymerase is thermostabe and has low error rates.
- It is suitable for PCR and primer extension reactions that require high fidelity synthesis.
- *Pfu* DNA polymerase-generated PCR fragments are blunt-ended.

Applications

1) Cloning
2) DNA expression
3) site-directed mutagenesis

General composition of PCR reaction mixture (total 50 ul)	
Pfu DNA polymerase (2.5 units/ul)	0.5 ul
10 x Reaction Buffer (*Pfu*)	5 ul
2.5mM (each) dNTPs	4 ul
Template	<500ng
Primer 1	0.2～1.0uM (final conc.)
Primer 2	0.2～1.0uM (final conc.)
Sterile distilled water	up to 50ul

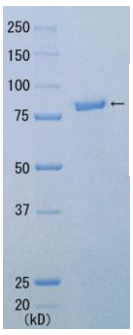
Specification

Storage Conditions: 50mM Tris-HCl (pH 8.2), 0.1mM EDTA, 1mM DTT, 50% glycerol, 0.1% Tween20, 0.1% Igepal CA-630
Store at -20°C
Concentration: 2.5 units/ul, where one unit is defined as the amount of enzyme that can incorporate 10 nmols of dNTPs into an acid-insoluble material in 30 minutes at 72°C when activated salmon sperm DNA was used as template/primer.

Quality Assurance: Greater than 95% of protein determined by SDS-PAGE (CBB staining) (Fig.1)
The absence of endonucleases and exonucleases was confirmed.
PCR Test**:** Good amplification result was obtained in PCR reaction using λDNA as a template (Fig. 2).

Reagents Supplied with Enzyme:

10 x Reaction Buffer (*Pfu*): 200mM Tris-HCl (pH 8.8), 100mM KCl, 100mM $(NH_4)_2SO_4$, 20mM $MgSO_4$, 1% TritonX-100, 1 mg/ml BSA
2.5mM (each) dNTPs



Pfu DNA polymerase

PCR condition
98°C 10sec
55°C 30sec
72°C 10min
} 30cycles
(2min in the case of 2kb DNA)

lane
M : marker
1 : 2 kb
2 : 4 kb
3 : 6 kb
4 : 8 kb

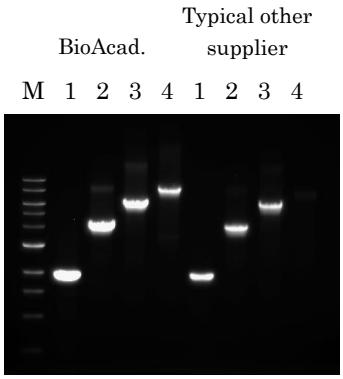


Fig. 1 SDS-PAGE of *Pfu* DNA polymerase

Fig. 2 Amplification of λ DNA