Pfu DNA Polymerase

User's Instruction

Description

Pfu DNA polymerase is isolated and purified from *E. coli* which is cloned the gene of *Pyrococcus furiosis* DNA polymerase. It catalyzes the polymerization of nucleotides into duplex DNA in the 5′→3′ direction in the presence of magnesium and exhibits 3′→5′ exonuclease (proofreading) activity. Pfu DNA polymerase is recommended for use in PCR and primer extension reactions that require high fidelity.

Kit Contents

	250U
1. Pfu DNA Polymerase (2.5U/μl)	100 μl
2. 10 × Pfu Buffer	1 ml

Note

- **Pfu DNA Polymerase Storage Buffer:** 20mM Tris-HCl (pH8.0), 0.1 mM EDTA, 1mM DTT, Stabilizers, 50% glycerol.
- **10 x Pfu Buffer:** 200 mM Tris-HCl (pH8.8), 100 mM KCl, 100 mM $(NH_4)_2SO_4$, 20 mM $MgSO_4$, etc.

Protocol

1. Set up PCR amplification reaction as the following table (take 50 μl per well as an example):

Component	Volume
Pfu DNA Polymerase (2.5U/μl)	1 μl
10 × Pfu Buffer	5 μl
dNTP Mixture (2.5 mM each)	4 μl

Template	<0.5 μg
Forward Primer (10 μM)	1 μl
Reverse Primer (10 μM)	1 μl
ddH_2O	Up to 50 μl

2. Thermocycling Conditions

	Temperature	Time
Initial Denaturation	94°C	2-5 min
30 Cycles	94°C	30 sec
	50-60°C	30 sec
	72°C	2 min/kb
Final Extension	72°C	5-10 min

Storage

Store at -20°C for 1 year. Avoid multiple freeze-thaw cycles.