

PF0212

Recombinant Pfu DNA Polymerase

Catalog No.	CSI13232	Quantity:	100 U
	CSI13233		500 U
	CSI13234		2500 U

Alternate Names: DNA polymerase, EC 2.7.7.7, Pfu polymerase, Pfu-DNA Polymerase.

Description: Pfu DNA polymerase enzyme is found in the hyperthermophilic archaeon *Pyrococcus furiosus*, where it functions *in vivo* to replicate the organism's DNA. *In vitro*, Pfu is used to swiftly amplify DNA in the Polymerase Chain Reaction, where the enzyme serves the central function of copying a new strand of DNA during each extension step. Pfu DNA polymerase has superior thermostability and 'proofreading' properties compared to other thermostable polymerases. Unlike Taq DNA polymerase, Pfu DNA polymerase possesses 3' to 5' exonuclease proof reading activity, meaning that it works its way along the DNA from the 5' end to the 3' end and corrects nucleotide misincorporation errors. Pfu DNA polymerase-generated PCR fragments will have fewer errors than Taq-generated PCR inserts. As a result, Pfu is more commonly used for molecular cloning of PCR fragments than the historically popular Taq. Pfu DNA polymerase is superior for techniques that require high-fidelity DNA synthesis, but can also be used in conjunction with Taq polymerase to obtain the fidelity of Pfu with the speed of Taq polymerase activity.

Pfu DNA Polymerase is a thermo-stable enzyme having a Mw of about 90 kDa. Pfu DNA Polymerase is derived from *E. coli* that and cloned from *Pyrococcus furiosus* strain Vc1 DSM3638. Pfu DNA Polymerase replicates DNA at 75°C, catalyzing the polymerization of nucleotides into duplex DNA in the 5' to 3' direction in the existence of magnesium. Pfu DNA Polymerase possesses 3' to 5' exonuclease (proofreading) activity. Base misinsertions that take place during polymerization are swiftly removed by the proofreading activity of the polymerase. Therefore, Pfu DNA Polymerase is suggested for use in PCR and primer extension reactions that require high-fidelity synthesis. Pfu DNA Polymerase-generated PCR fragments are blunt-ended.

Physical Appearance: Sterile liquid formulation.

Gene ID: 1468044

Protein Accession No: P61875

Source: *E. coli*

Formulation: 50 mM Tris-HCl, pH 8.2 + 1 mM DTT + 0.1 mM EDTA + 0.05% CHAPS and 50% glycerol.

PRC Protocol: Add the following components to a amplify 1 kb DNA template: 5 µl 10 x buffer with MgSO₄. 4 µl 2.5 mM dNTPs. 1 µl Primers mix (10 µM each). 0.2 µl Pfu-DNA Polymerase. and 38 µl Water. Amplify using the following cycling parameters: Heat Soak: 1 cycle at 94°C/4 min. Denaturation: 30 cycles at 94°C/30 sec. Annealing: 30 cycles at 58°C /30 sec. Extension: 30 cycles at 72°C /90 sec. Final: 1 cycle at 72°C /5 min.

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Unit Definition: 1 U of enzyme catalyzes the incorporation of 10 nmol of dNTP into acid-insoluble product in 30 minutes at 75°C.

Applications:

1. Ideal for high-fidelity amplification.
2. 3'-5' exonuclease activity provides a low error rate.
3. One of the most thermostable DNA polymerases known.
4. Lack of extendase activity means no unwanted 3' overhangs.
5. Optimal for blunt-end PCR cloning.
6. Optimum temperature near 75°C.
7. 95% active after 1-hour incubation at 98°C.

Storage & Stability: Pfu DNA Polymerase although stable at 10°C for 5 days, should be stored desiccated below -18°C.

Please prevent freeze-thaw cycles.

NOT FOR HUMAN USE. FOR RESEARCH ONLY. NOT FOR DIAGNOSTIC OR THERAPEUTIC USE.

