# Product Manual

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|---|---|
| **Product Name** | Pfu II High-Fidelity DNA Polymerase |
| **Source** | Recombinant expression in E. coli |
| **Catalog Number** | CSB-DEM043 |
| **Physical Form** | Liquid |
| **Enzyme Activity** | 1U/μL |
| **Storage Conditions** | -20 ±5°C |
| **Storage Buffer** | 10 mM Tris-HCl, 100 mM KCl, 1 mM DTT, 0.1 mM EDTA, 0.5% Tween 20, 0.5% NP-40, 50% Glycerol |
| **Activity Definition** | The amount of enzyme required to incorporate 10 nmol of deoxynucleotides into acid-insoluble material in 30 minutes at 75°C is defined as 1 unit (U). |
| **Quality Control** | No detectable exonuclease and endonuclease activities |
| **Shelf Life** | 24 months |

## Product Description

Pfu II High-Fidelity DNA Polymerase is a fusion of a proofreading domain and an enhanced processivity domain, resulting in increased fidelity and speed. It can be used as a standalone enzyme or provided in a master mix format, enabling highly specific amplification of various templates. It is an ideal choice for cloning and can be used for long or challenging amplicons. Compared to other DNA polymerases, Pfu II High-Fidelity DNA Polymerase offers stable performance, shorter experimental protocols, and higher yield with lower enzyme usage.

## Product components

| Component No. | Component Name | Specifications | | |
|---|---|---|---|---|
| 1 | 5×Pfu buffer | 0.6mL | 3mL | 6mL |

WUHAN HUAMEI BIOTECH CO.,LTD

No.818 Gaoxin Avenue, Wuhan Hi-tech Medical Devices Park, Donghu High-tech Development Zone 430206, Wuhan City, Hubei Province, P.R. China.

www.cusag.cn / www.cusagivd.com cusag@cusag.cn +86-27-65521556/+86-27-87196282 Ext 853 +86-27-87196150

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|---|---|---|---|---|
| 2 | Pfu Ⅱ high fidelity DNA polymerase | 100U | 500U | 1000U |

**Operating instructions**

**Recommended Reaction System**

| Composition | Addition amount |
|---|---|
| $ddH_2O$ | Up to 30 μL |
| 5×Pfu Buffer | 6 μL |
| 10 mM dNTPs | 1 μL |
| Upstream Primer (10 μM) | 0.5 μL |
| Downstream Primer (10 μM) | 0.5μL |
| Template DNA | X μL |
| Pfu Ⅱ high fidelity DNA polymerase | 1 μL |

**Note:**

**a. Primer concentration:**

Generally, a primer concentration of 0.2 μM in the reaction system yields good results. Adjust the primer concentration within the range of 0.1 μM to 1.0 μM if the reaction performance is poor.

**b. Template concentration:**

Template Concentration: Genomic DNA (animal and plant) 0.1-1 μg, E. coli genomic DNA 10-100 ng, λDNA 0.1-10 ng, plasmid DNA 0.1-10 ng. If using undiluted cDNA stock as the template, the volume should not exceed 1/10 of the total qPCR reaction volume.

**c. Polymerase concentration:**

Adjust the enzyme usage between 0.25 - 1 μL. Increasing the enzyme usage generally improves amplification yield but may reduce specificity.

**d. Mg2+ and Additives:**

The Mg2+ concentration in most PCR reaction systems should be within the range of 1.0-5.0 mM. However, for challenging samples such as high-GC DNA, additives like DMSO or formamide may need to be added to the PCR reaction system.

**Recommended PCR Reaction Program**

| Temperature | Time | Cycling |
|---|---|---|
| 98°C | 1 min | 1 |
| 98°C | 10 s | 30-35 |
| 45-68°C | 20 s | |
| 72°C | 1 kb/10-15s | |
| 72℃ | 5min | 1 |

**Note:**

**e. Annealing Temperature and Time:**

Annealing Temperature and Time: Adjust the annealing temperature based on the primer's Tm value. Generally, setting it 3-5°C below the primer's Tm value is sufficient. The recommended annealing time is 20 seconds, which can be adjusted within the range of 10-30 seconds.