



Super Fidelity DNA Polymerase

Product Number: PC06

Shipping and Storage

Ice pack transportation, stored at -20°C, with a shelf life of 2 years, to avoid repeated freezing and thawing.

Components

Component	100U	1KU	5KU
Super Fidelity DNA Polymerase(5U/μl)	20μL	200μL	1mL
5×Reaction Buffer(with Mg ²⁺)	0.2mL	1mL	5mL

Description

Super Fidelity DNA Polymerase is a new generation of ultra fidelity DNA polymerase modified from Pfu DNA Polymerase, which has greatly improved its long segment amplification ability, amplification specificity, and amplification yield. By optimizing the reaction buffer and using simple templates such as lambda DNA and plasmids, fragments up to 40kb can be effectively amplified; Using complex templates such as genomic DNA, fragments up to 20kb can be amplified; The use of cDNA templates can effectively expand fragments up to 10kb in length. Its mismatch rate is 1/53 of that of ordinary Taq enzymes and 1/6 of that of Pfu enzymes, and the amplification speed can reach 15-30 seconds/kb. High fidelity and excellent amplification efficiency make Super Fidelity DNA Polymerase suitable for direct PCR of bacterial, fungal, plant tissue, animal tissue, or whole blood samples, with amplification products being flat ended.

Application

This product is suitable for PCR reactions using genomic DNA, cDNA, Plasmid DNA, and crude samples as templates.

1. High fidelity PCR and vector construction;
2. Gene cloning;
3. Gene directed mutagenesis;
4. High throughput PCR and sequencing.

Protocol

1. Reaction system:

Component	Volume
5×Reaction Buffer(with Mg ²⁺)*	10μl
dNTP Mix (10mM each)	1μl
Upstream Primers(10μM)	2μl
Downstream Primers(10μM)	2μl
Super Fidelity DNA Polymerase(5U/μl)	1μl
Template DNA	xμl
ddH ₂ O	up to 50μl

Note: The system already contains a final concentration of 2mM Mg²⁺. If necessary, explore the optimal concentration of Mg²⁺ at intervals of 0.2-0.5mM.

The optimal reaction concentration varies among different templates and can be adjusted according to the sample type.

2. Reaction program:

Step	Temperature	Time	Cycle Number
Pre-denaturation ^a	95°C	3minutes	1



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Denaturation ^b	95°C	15seconds	
Annealing ^c	55°C	15 seconds	25-35 cycles
Extension	72°C	30-60 sec/kb	
Final Extension	72°C	5-10 minutes	1

a. The recommended pre denaturation temperature for most templates is 95°C. If the amplicon exceeds 10kb, the pre denaturation temperature can be lowered to 92°C for no more than 2 minutes.

b. For most templates, a 95°C denaturation time of 5-10 seconds is sufficient. If the amplicon exceeds 10kb, the pre denaturation temperature can be lowered to 92°C and the denaturation time can be extended to 15 seconds.

c. Generally speaking, the annealing temperature should be set within the range of primer T_m value ± 3°C, and it is recommended to set the annealing time to 10 seconds. For some difficult templates, the annealing time can be adjusted between 10-30 seconds.

Note

1. Please use high-quality templates.
2. Do not use dUTP and primers and templates containing uracil.
3. If necessary for the experiment, the usage of Super Fidelity DNA Polymerase can be increased appropriately, but it is recommended not to exceed 2 U of enzyme in a 50µl system.
4. Super Fidelity DNA Polymerase has strong proofreading activity. Therefore, if amplification products require TA cloning, DNA purification must be performed before adding A.
5. To prevent the degradation of primers due to the proofreading activity of Super Fidelity DNA Polymerase, please add polymerase at the end when preparing the reaction system.
6. This product is only for scientific research purposes.
7. For your safety and health, please wear lab coats and disposable gloves when operating.