

2 × CYF8 FastLong PCR MasterMix

Product Description	Size	
	CYPC8001	CYPC8002
2×CYF8 FastLong PCR Master Mix	1 ml	5 ml

Store: -20°C, 2 years

Product Description

The kit contains CYF8 FastLong DNA Polymerase, dNTPs and optimized reaction buffer at a concentration of 2×. Before reaction, DNA/cDNA template, specific primer, and ddH₂O should be prepared to mix the final concentration of 1×.

The Mix contains red tracer dye for quicker electrophoresis. And the PCR amplification products are with polyA end, which can be directly employed to the clone reaction with our pTOPO vector. Of course, after purified, the amplification products also can be used for other follow-up experiments, such as digestion, ligation and fluorescence sequencing.

Product Features

- High yield;
- Fast and long amplification: 5s-10s/kb, about 10kb;
- Super Fidelity: Up to 3–6 times that of traditional Taq DNA polymerase;
- Powerful amplification capacity: suitable for the amplification of complicated structure or high GC content template;

Recommended PCR system settings:

Component	25 µl Reaction	50 µl Reaction	Final Concentration
2×CYF8 PCR MasterMix	12.5 µl	25 µl	1 ×
Forward Primer (10 µM)	0.5 µl	1 µl	0.2 µM
Reverse Primer (10 µM)	0.5 µl	1 µl	0.2 µM
Template DNA	as required	as required	
ddH ₂ O	up to 25 µl	up to 50 µl	

Reference template dosage (50 µl reaction system)

Plasmid: 0.1–10 ng; Bacterial genome: 10–100 ng; Human genome: 50–150 ng;

cDNA: 1–5 µl from RT reaction;

Proposed PCR cycle conditions

Step	Temperature	Time	Cycle Number
Initial denaturation	94°C	2-3 minutes	
Denaturation	94°C	10 seconds	

Annealing	55°C	10-15 seconds	25-35 cycles
Extension	72°C	10 seconds/kb	
Final Extension	72°C	1-5 minutes	
	4-8°C	Hold	

Precautions:

1. Due to CYF8 super fast amplification capacity, 5s/kb extension and less cycles are recommended for simple or plasmid templates; 15s-20s/kb extension and more cycles for complicated template is recommended;
2. For templates with high GC content, the pre-denaturation and denaturation temperatures can be increased to 98°C, and/or extending the denaturation time; But for the long fragments, raising the denaturation temperature is not allowed to avoid the DNA damage, whereas extending pre-denaturation time (for example, 5 min) and denaturation time (e.g. 5s-10s) is recommended.
3. If the amplification template GC content is high or the complex template amplification effect is poor, the DMSO can be added to the reaction mixture with the final concentration of 1-8%, according to 1% gradient increasing to look for the best concentration. Or adding betaine to the final concentration of 1.0-1.7 M. And at the same time, using the Touchdown PCR.

Example Amplification

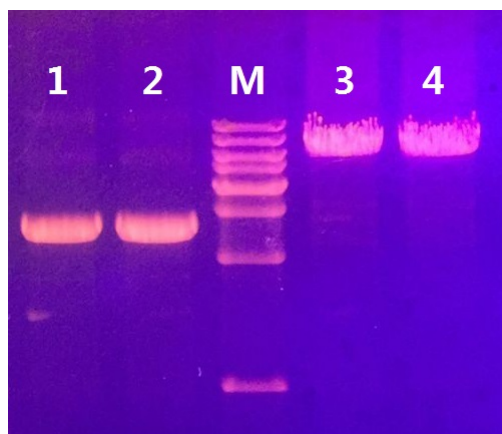


Figure 1. Amplification of DNA fragment from 50 ng human genomic DNA in 50 μ l reaction mixture.
 Lane M: 1kb ladder;
 Lane 1-2: H α 1AT, 2.6Kb, extension 10 sec/kb;
 Lane 3-4: β -globin, 6kb, extension 15 sec/kb.

This product is furnished for LABORATORY RESEARCH USE ONLY.
Not for diagnostic or therapeutic use.