

2×AMPQX® Ultra-Rapid II Hot Start PCR Master Mix

Cat. no. LM201006-3

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Product Information

Product name	2 × AMPQX® Ultra-Rapid II Hot Start PCR Master Mix
Cat.No.	LM201006-3
Size	1 mL / 5×1 mL / 50×1 mL / 100×1 mL
Storage and shipping	The product is shipped with dryice and can be stored at -25°C ~ -5 °C for 2 year.

2. Product description

2×AMPQX® Ultra-Rapid II HotStart PCR Master Mix contains antibody-modified thermostable Taq DNA Polymerase, added with strong extension factors and optimized buffer system, with super high amplification efficiency, and is very suitable for PCR amplification of most colonies such as Escherichia coli, Agrobacterium, yeast, etc.

The extension speed of this product for complex template amplification such as genome within 3kb can reach 1 sec/kb, the amplification speed of 3-6 kb fragments can reach 3 sec/kb, the amplification speed of 6-10 kb fragments can reach 5 sec/kb, and the amplification speed of fragments above 10 kb can reach 10 sec/kb. The amplification speed of simple templates such as plasmids within 6 kb can reach 1 sec/kb, which can greatly save PCR reaction time.

At the same time, The Mix contains dNTPs and Mg²⁺, and only primers and templates need to be added for amplification.

In addition, The Mix contains red tracer dye, which can be directly electrophoresed after the reaction is completed, making it easy to use.

The protective agent added to the system allows this product to maintain stable activity after repeated freezing and thawing. The 3' end of the PCR product has A, which can be easily cloned into the T vector.

3. Instructions

3.1. Recommended PCR Reaction System

Components	Volume μL	Volume μL	Final Conc.
2 $\times$ AMPQX® Ultra-Rapid II Hot Start PCR Master Mix(a)	25	12.5	1 $\times$
Template(b)	x	x	-
Forward Primer (10 μM)(c)	2	1	0.4 -0.5 μM
Reverse Primer (10 μM)	2	1	0.4 -0.5 μM
ddH ₂ O	Up to 50	Up to 25	-

[Note]:

- a) In 1 $\times$ Master Mixes containing 2 mM Mg²⁺ and 200 μM dNTPs, thaw and mix thoroughly before use.
- b) E.coli and Agrobacterium can be directly loaded with the bacterial liquid or by picking colonies. For yeast, it is recommended to boil the bacterial liquid or colonies for 5 minutes before loading. Ensure that the bacterial liquid samples are mixed thoroughly by shaking before loading. The recommended volume for loading bacterial liquid is 2-4 μL (0.5-2.5 OD).

Recommended usage amounts for different templates:

Types of templates	Recommended usage amounts (for a 50 μL reaction system)
Genomic DNA	10-1,000 ng
Plasmid or λ DNA	0.5-50 ng
E. coli culture	0.5-2.5 OD

- c) The final concentration range of primers in the PCR reaction system is 0.2-1 μM , with a recommended concentration of 0.4 μM .

3.2. Reaction program

Cycle step	Temp.	Time	Cycles
Initial denaturation	95 °C	30min	1
Denaturation	95 °C	15 sec	30-35
Annealing(a)	60 °C	20 sec	
Extension(b)	72 °C	1-10 sec/kb	
Final extension	72 °C	5 min	1

[Note]:

- a) Recommended annealing temperature: 60°C, a temperature gradient can be set up to find the optimal temperature for primer annealing. The recommended annealing time is set to 20 sec and can be adjusted from 10-30 sec. Too long annealing time may result in diffuse amplification products on the gel.
- b) For complex templates such as genomic DNA and E. coli up to 3 kb, set the extension time to 1 sec/kb. For complex templates up to 6 kb, set the extension time to 3 sec/kb. For most complex templates up to 10 kb, set the extension time to 5 sec/kb. For complex templates longer than 10 kb, set the extension time to 10 sec/kb.

For simpler templates such as plasmids, set the extension time as follows:

Up to 6 kb: 1 sec/kb

6-10 kb: 3 sec/kb

Longer than 10 kb: 5-10 sec/kb

If higher yield is needed, you can appropriately extend the extension time, but it should not exceed 30 sec/kb.

4. Notes

1. This product is for research use only!
2. Please operate with lab coats and disposable gloves for your safety.