2× Taq Master Mix

Catalog # P111 / P112

Version 5.1

Vazyme biotech co., ltd.

Introduction

Taq DNA Polymerase is a thermostable DNA polymerase that possesses a 5 '→3 'polymerase activity and a 5 'flap endonuclease activity.

2× Taq Master Mix contains Taq DNA Polymerase, dNTP, and an optimized buffer system. The amplification can start only with the addition of primer and template, thereby easing PCR setup and improving reproducibility. Protective reagents in the 2× Taq Master Mix enable the resistance to repeated freeze-thaw cycles.

2× Taq Master Mix also provides another edition with dyes which enable direct loading PCR products onto agarose gels. The obtained PCR products are compatible with ClonExpress II One Step Cloning Kit series (Vazyme, Cat.No. #C112, #C113). The PCR products contain A at the 3'-end and can be directly cloned into T-Vectors.

Package Information

Components	P111-01 5 ml	P111-02 15 ml	P111-03 50 ml
2× Taq Master Mix	5 ml	15 ml	50 ml

Components	P111-w1 5 ml	P111-w2 15 ml	P111-w3 50 ml
2× Taq Master Mix	5 ml	15 ml	50 ml
ddH_2O	5 ml	15 ml	50 ml

Components	P112-01 5 ml	P112-02 15 ml	P112-03 50 ml
2× Taq Master Mix (Dye Plus)	5 ml	15 ml	50 ml

Components	P112-w1 5 ml	P112-w2 15 ml	P112-w3 50 ml
2× Taq Master Mix (Dye Plus)	5 ml	15 ml	50 ml
ddH_2O	5 ml	15 ml	50 ml

Storage

Store at -20℃.

Quality Control

Exonuclease Activity: The product is tested in a reaction containing 10 U of enzyme and 0.6 μg of λ-Hind III. After incubation at 37℃ for 16 hours, there is no visually discernible change to DNA bands determined by agarose gel electrophoresis.

Endonuclease Activity: The product is tested in a reaction containing 10 U of enzyme and 0.6 μg of Supercoiled pBR322. After incubation at 37℃ for 4 hours, there is no visually discernible change to DNA band determined by agarose gel electrophoresis.

Functional Assay: The α-1-antitrypsin gene is amplified for 30 cycles in a 50 μl system using 100 ng human genomic DNA as template. A single DNA band of 360 bp is detected by agarose gel electrophoresis.

Protocol

1. General reaction mixture for PCR:

ddH_2O	to 50 μl
2× Taq Master Mix	25 μl
Template DNA*	Optional
Primer 1 (10 μM)	2 μl
Primer 2 (10 μM)	2 μl

*The recommended amount of DNA template for a 50 μl reaction system is as follows:

Human Genomic DNA	0.1 - 1 μg
Bacterial Genomic DNA	10 - 100 ng
λ DNA	0.5 - 5 ng
Plasmid DNA	0.1 - 10 ng

2. Thermocycling conditions for a routine PCR:

94℃	5 min (Pre-denaturation)	
94℃	30 sec	} 30 - 35 cycles
55℃*	30 sec	
72℃	60 sec / kb	
72℃	7 min (Final extension)	
4℃	Hold	

*The optimal annealing temperature should be 1-2℃ lower than the T_m of the primers used.

Handling Notes

Taq DNA Polymerase also shows polymerase activity at room temperature. Therefore, it is recommended to set up reaction systems on ice and then immediately start the reaction in PCR amplifier, so as to reduce nonspecific amplification during preparation and get better PCR results.

Primers Designing Notes

1. Choose C or G as the last base of the 3'-end of the primer;
2. Avoid continuous mismatching at the last 8 bases of the 3'-end of the primer;
3. Avoid hairpin structure at the 3'-end of the primer;
4. T_m of the primers should be within the range of 55℃ - 65℃;
5. Additional sequence should not be included when calculating Tm of the primers;
6. GC content of the primers should be within the range of 40% - 60%;
7. T_m and GC content of forward and reverse primes should be as similar as possible.