# 2X ACE Taq-Plus Master Mix (Blue)

**Cat# EP1003 – 5ml / EP1004 – 15ml**

**Storage at -20 ℃**

## INTRODUCTION

**2X ACE Taq-Plus Master Mix** contain Taq-Plus DNA Polymerase, dNTP, and an optimized buffer system. Taq-Plus DNA polymerase is a mixture of Taq DNA polymerase and an enzyme containing 3' → 5' exonuclease activity. Its fidelity is 6 times greater than that of Taq DNA Polymerase. Compared with Taq DNA Polymerase, Taq-Plus DNA polymerase has stronger amplification performance, higher sensitivity, and is more tolerant of impurities within 5 kb amplifying range.

The amplification can start only with the addition of primer and template, thereby easing PCR setup and improving reproducibility. It can amplify up to 10 kb from human genomic DNA or up to 15 kb from λ DNA. Protective agents in the **2X ACE Taq-Plus Master Mix** enable the resistance to repeated freeze-thaw cycles. Also, **2X ACE Taq-Plus Master Mix** contain blue loading dye, which enables direct loading PCR products onto agarose gel.

The obtained PCR products are compatible with **ACE One Step Cloning kit (ACE Biolabs, EC1001)**. The PCR products contain A at the 3'-end and can be directly cloned into T-Vectors.

## CONTENTS

| No | Component | **EP1003 – 5 ml** | **EP1004 – 15 ml** |
|---|---|---|---|
| AA | 2X ACE Taq-Plus Master Mix (Blue) | 1 ml x 5 | 1ml x 15 |

## UNIT DEFINITION

One unit (U) is defined as the amount of enzyme that incorporates 10 nmol of dNTPs into acid-insoluble products in 30 minutes at 74℃ with activated salmon sperm DNA as the template / primer.

## PROTOCOL

### 1. Mix the following components

| | |
|---|---|
| 2X ACE Taq-Plus Master Mix (Blue) | 25 μl |
| Primer 1 (10 uM) | 2 μl |
| Primer 2 (10 uM) | 2 μl |
| Template DNA / cDNA * | Optional |
| $ddH_2O$ | To 50 μl |

*Optimal reaction concentration varies from templates. In a 50 μl system, there commended template usage is as follows: 0.1-1 μg Human genomic DNA, 10-100 ng Bacterial genomic DNA, 0.5-5 ng λDNA, 0.1-10 ng Plasmid DNA.

**2. Place the sample in a PCR instrument and run the following program for PCR:**

| Stage | Temp. | Time | Cycle |
|---|---|---|---|
| Pre-Denaturation | 94℃ | 5 min | 1 |
| Denaturation | 94℃ | 30 s | 30-35 |
| Annealing* | 55℃ | 30 s | |
| Extension | 72℃ | 60 s/ kb | |
| Final Extension | 72℃ | 7 min | 1 |
| Hold | 4℃ | | |

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

## PRIMERS DESIGN 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. Tm 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. Tm and GC content of forward and reverse primes should be as similar as possible.

## PRODUCT USE LIMITATION

These products are intended for research use only.