

## 2x HotStart *Taq* PCR MasterMix

**Cat. no.** KTT-BB02

**Storage:** For long term storage, store at  $-20^{\circ}\text{C}$ ; 2x HotStart *Taq* PCR MasterMix retains full activity after repeated freezing and thawing. For regular use, store at  $4^{\circ}\text{C}$ .

### Product Size

Components	KTT-BB02	KTT-BB02M
2x HotStart <i>Taq</i> PCR MasterMix	1 mL	1 mL
ddH <sub>2</sub> O	1 mL	1 mL
Loading dye in MasterMix	Yes	No

## Introduction

2x HotStart *Taq* PCR MasterMix is a 2x concentrated, optimized mixture composed of a HotStart *Taq* DNA polymerase, dNTPs, MgCl<sub>2</sub>, a reaction buffer, a PCR reaction enhancer, an optimizer, and a stabilizer. The advantages of 2x HotStart PCR MasterMix include high convenience, sensitivity, specificity, and stability. It minimizes human errors during the PCR operating process. The HotStart inhibitor blocks the substrate binding site of HotStart *Taq* DNA polymerases in a temperature-dependent manner. Inactive polymerase-inhibitor complexes are formed at temperatures  $< 40^{\circ}\text{C}$ , where the affinity of the HotStart inhibitor for the HotStart *Taq* DNA polymerase is higher than the binding affinity of the template DNA. Between 40 and  $55^{\circ}\text{C}$ , the HotStart inhibitor competes with the template DNA for binding to the *Taq* DNA polymerase, thereby shifting the binding equilibrium toward complex formation with only target-specific primed template DNA. This minimizes nonspecific amplification in PCR and ensures high sensitivity and specificity.

2x HotStart *Taq* PCR MasterMix is designed for quick and easy preparation of reaction mixture, which minimizes contamination during the PCR operating process. For PCR reaction set-up, users only need to pipet an aliquot part of 2x HotStart *Taq* PCR MasterMix and dilute the MasterMix to 1x by adding templates, primers, and water up to the reaction volume. There are two types of this product: MasterMix with loading dye (blue) and MasterMix without loading dye (colorless). PCR products produced using MasterMix with loading dye can be loaded directly without extra loading buffer.

### Product Components (2x)

0.1 U/ $\mu\text{l}$  HotStart *Taq* DNA polymerase

500  $\mu\text{M}$  dNTP each

50 mM Tris-HCl (pH 8.3)

20 mM KCl

4 mM MgCl<sub>2</sub>

Stabilizer and enhancer for the storage buffer

## Applications

- Gene detection: 2× HotStart *Taq* PCR MasterMix is especially suitable for techniques such as large-scale gene detection, semi-quantitative PCR, and detection of small amounts of DNA.
- Highly specific DNA amplification: This is suitable for techniques such as highly sensitive amplification of genomic DNA with a high background (e.g., specific gene sites or detection of an exogenous virus in genomic DNA), DNA sequencing, Multiplex PCR, T-A cloning.

## Protocol

1. To set up a 25-μL PCR reaction system: A 1-kb fragment of human genomic DNA was amplified using 2×HotStart *Taq* PCR MasterMix (if using a different reaction system, proportionally increase or reduce the amount of reaction components).

Template	<1 μg
Primer 1 (10 μM)	1 μL
Primer 2 (10 μM)	1 μL
2x Master Mix	12.5 μL
ddH <sub>2</sub> O	Up to 25 μL

2. PCR cycle set-up :

94°C 3 min	
94°C 30 sec	} 30 cycles
55°C 30 sec	
65°C 1 min	
65°C 5 min	

3. Result detection: Load 5 μL of PCR products to agarose gel for detection.

---

Note: The example is only for reference; users must set up an optimal reaction system according to different reaction conditions such as different templates or primers.

---

This product is for research only, not for diagnostic and clinical use.