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# 2× PCR Master Mix (with Dye)

# 1 Components

Contents	HY-K0531-1 mL	HY-K0531-5 mL	HY-K0531-20 mL
2× PCR Master Mix (with Dye)	1 mL	1 mL ×5	(1 mL ×5)×4

## 2 Introduction

MCE 2× PCR Master Mix (with Dye) is a convenient, ready-to-use kit for PCR. The 2× PCR Master Mix contains all the reagents necessary for routine PCR, including Taq DNA Polymerase, dNTPs Mix, MgCl<sub>2</sub> and optimized reaction buffer. With the primers and template added, the optimized system will provide sensitive and reliable DNA synthesis. After the PCR reaction, the PCR products can be loaded directly onto an agarose gel. It saves preparation time, reduces the risk of contamination from multiple pipetting steps, and provides consistent reaction-to-reaction performance. The obtained PCR products contain polyA at the 3'-end and can be directly cloned into T-Vectors.

## 3 Protocol

#### 1. Prepare PCR reaction mixture

To obtain reliable PCR reaction results, the suggested template amount is 50 ng to 200 ng for genomic DNA or 0.1 ng to 10 ng for plasmid DNA. Please prepare the PCR reaction solution according to the list below (all reagents should be placed on ice).

Reagent	Volume
2× PCR Master Mix	25 μL
PCR Forward Primer (10 μM)	2 μL
PCR Reverse Primer (10 μM)	2 μL
DNA	x μL
ddH <sub>2</sub> O	to 50 μL

#### 2. Common cycling parameters for a routine PCR

Temperature	Time		Cycles
94°C	5 min		1
94°C	30 sec	)	
50-60°C	30 sec		30-35
72°C	30-60 sec/kb		
72°C	10 min		1

### Notes:

a. The product provides 1.5 mM  $Mg^{2+}$  in  $1 \times$  concentration.

b. The PCR reaction conditions should be set according to the specific conditions such as template amount, target fragment size, base sequence and primer length in actual operation.

- c. The annealing temperature can be set 2-5°C lower than the theoretical value of T<sub>m</sub>.
- d. For higher yields, the recommended extension time for molecular identification is 30 sec/kb and for gene cloning is 60 sec/kb.

# 4 Storage Conditions

Store at -20°C for 2 years. 4°C for short-term storage (up to 3 months).

Avoid repetitive freeze-thaw cycles while using.

# 5 Precautions

- a. Gently invert the tube upside down several times before use. DO NOT vortex. Brief centrifugation prior to use is recommended.
- b. It is recommended to set up reaction systems on ice. Taq DNA Polymerase also shows polymerase activity at room temperature, so as to reduce nonspecific amplification during preparation and get better PCR results.
- c. The PCR product is not suitable for polyacrylamide gel electrophoresis.
- d. This product is for R&D use only, not for drug, household, or other uses.
- e. For your safety and health, please wear a lab coat and gloves while handling.

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