

2×Taq MasterMix (Dye)

Cat. No. : CW0682L (25 mL)

Components

| Component | CW0682L 25 mL |
|-----------------------|------------------|
| 2×Taq MasterMix (Dye) | 5×5 mL |
| ddH ₂ O | 5×5 mL |

Note: 2×Taq MasterMix contains Taq DNA polymerase, 3 mM MgCl₂, and 400 μM dNTP mix.

Introduction

2×Taq MasterMix is a convenient premixed 2×concentrated solution for PCR which include Taq DNA Polymerase, PCR Buffer, Mg²⁺, dNTPs, PCR Stabilizer and PCR Intensifier. As the unique MasterMix makes the whole reaction system quite stable, more than 98% PCR amplification and complicated templates can be performed. It is easy to use and can maximatily avoid the personal error and contamination. As dye (blue) is contained in the mixture, it is recommended to electrophoresis directly without adding loading buffer to the PCR product. Taq MasterMix catalyzes the non-template directed addition of an adenine residue to the 3'-end of both strands of DNA molecules to make it suitable for TA cloning.

Applications

- Routine PCR
- DNA sequence detection

Key Features

- Convenient, you just add template, primers and water to the 2×MasterMix
- Stability and repeatability

Protocol

The following basic protocol serves as a general guideline and a starting point for any PCR amplification. Optimal reaction conditions (incubation times and temperatures, concentration of Taq MasterMix, primers, and template DNA) vary and need to be optimized according to the template, primer structure and target fragment size.

1. Prepare the reaction mix to 50 µL according to the following table.

| Reagent | 50 µL PCR reaction | Final Concentration |
|-----------------------|--------------------|---------------------|
| 2×Taq MasterMix (Dye) | 25 µL | 1× |
| Forward Primer, 10 µM | 2 µL | 0.4 µM |
| Reverse Primer, 10 µM | 2 µL | 0.4 µM |
| Template DNA | <0.5 µg | <0.5 µg/reaction |
| RNase-Free Water | Up to 50 µL | |

Note: The recommended primer concentration for PCR is between 0.1-1.0 µM of each primer. The use of higher concentrations of primers can have for the higher amplification effect. Low primer concentration generally ensures cleaner product and lower background.

2. PCR reaction conditions.

| Procedure | Temperature | Time |
|------------------|-------------|-------|
| Pre-denaturation | 94°C | 2 min |
| Denaturation | 94°C | 30 s |
| Annealing | 55-65°C | 30 s |
| Extension | 72°C | 30 s |
| Final extension | 72°C | 2 min |

Note:

1) The recommended annealing temperature is about 5°C below T_m. If extra bands are observed, higher annealing temperatures should be considered. The absence of product can indicate the need for a lower annealing temperature.

2) PCR extension time is depended on the target gene sequence and CWBIO Taq DNA polymerase is approximately 1 kb DNA/30 s.

3) The number of PCR cycles will basically depend on the downstream application of the PCR product.

3. Detection of results:

The dye (blue) is contained in the mixture; after the end of the reaction, 5 µL reaction products electrophoresis directly without adding loading buffer to the PCR product.

Shipping and Storage

Store at -20°C for up to 3 years. For frequently used, Store at 2-8°C for short-term (1 week), and avoid frequent freeze-thawing.