

## 2×Taq Master Mix (Dye)

### T665590

Storage at -20°C . Avoid freeze/thaw cycle

#### Introduction:

Taq Master Mix is a 2 × concentrated solution of Taq Master Mix,  $Mg^{2+}$ , dNTPs and all other components required for PCR, except DNA template and primers. This pre-mixed formulation saves time and reduces contamination due to a reduced number of pipetting steps required for PCR set up. The mix is optimized for efficient and reproducible PCR. Its application for routine PCR with high reproducibility and generation of PCR products for TA. The original formula makes the reaction system stable. At the same time, complex DNA templates can be effectively amplified, and operational error and contamination can be minimized to the possible. This product has been added with dye (blue) and can be tested directly after the reaction is complete.

#### Ordering Information:

| Cat No.  | Component               | T665590-5mL | T665590-25mL | Storage                            |
|----------|-------------------------|-------------|--------------|------------------------------------|
| T665590A | 2 × Taq Master Mix(Dye) | 1mL×5       | 5mL × 5      | -20°C .<br>Avoid freeze/thaw cycle |
| T665590B | dd H <sub>2</sub> O     | 1mL×5       | 5mL × 5      | -20°C .                            |

Notes: 2 × Taq Master Mix contains Taq DNA Polymerase, 3mM  $MgCl_2$  and 400μM each dNTP

#### Protocol:

Gently vortex and briefly centrifuge 2 × Taq Master Mix after thawing. Place a thin-walled PCR tube on ice and add the following components for each 50μL reaction:

PCR reaction:

| Components            | Total volumn:50μL | Concentration |
|-----------------------|-------------------|---------------|
| 2×Taq Master Mix      | 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/50 μl |
| ddH <sub>2</sub> O    | up to 50 μL       |               |

Notes: The recommended concentration range of the PCR primers is 0.1-1  $\mu$ M. Excessive primer concentrations increase the probability of mispriming and generation of non-specific PCR products.

PCR thermal cycling conditions :

| Step                 | Temperature | Time  | Number of cycles |
|----------------------|-------------|-------|------------------|
| Initial denaturation | 94°C        | 2 min |                  |
| Denaturation         | 94°C        | 30s   | 25-35            |
| Annealing            | 55-65°C     | 30s   |                  |
| Extension            | 72°C        | 30s   |                  |
| Final Extension      | 72°C        | 2 min |                  |

Notes :

- 1) The annealing temperature should be 5°C lower than the melting temperature ( $T_m$ ) of the primers. Annealing for 30 seconds is normally sufficient. If non-specific PCR products appear, the annealing temperature should be optimized stepwise in 1-2°C increments.
- 2) The optimal extension temperature for Taq DNA polymerase is 70-75°C. The recommended extension step is 2 min/kb at 72°C for PCR.
- 3) If less than 10 copies of the template are present in the reaction, about 40 cycles are required. For higher template amounts, 25-35 cycles are sufficient.