

## 2 ×SuperTaq PCR Mastermix

**Code No. :** NG011S 10 ml

NG011M 80 ml

**Storage :** Store at -20 °C for up to one year and avoid repeated freezing and thawing.

### Introduction :

2 ×SuperTaq PCR mastermix is a 2 × concentrated, optimized mixture composed of superTaqDNA Polymerase, dNTP Mixreaction buffer and other PCR reaction enhancer. Suitable for ultra-fast PCR reactions and large-scale genetic testing, it demonstrates strong amplification capability for target fragments with high GC content and complex secondary structures. For short fragments or simple templates such as plasmids, a 5 s/kb extension speed and fewer cycles can be attempted to further shorten PCR time; for long fragments  $\geq 3$  kb or complex templates with low yields, a 15-30 s/kb extension speed or more cycles can be used. PCR reaction system can be carried out only by adding template, primer, water and mix to the reaction buffer, which simplifies the experimental operation process and reduces the pollution in the operation process. PCR products have an A-tail at the 3' end and can be directly used for TA cloning after purification. This product is a dye-containing PCR premix; after the reaction, electrophoresis can be performed directly without adding DNA loading buffer. It can also be purified for subsequent operations such as restriction digestion, ligation, and fluorescent sequencing. If frequently used, this product can be stored at 4 °C.

### Product Specifications :

1. Efficient and stable buffer system.
2. High sensitivity: low abundance template can be effectively amplified.
3. Wide applicability: The reaction system can effectively expand the complex template structure and high GC content template.

### Operation Procedure:

#### 1. Preparation of PCR reaction system

| content                       | 25 $\mu$ l       | 50 $\mu$ l       | Final Conc. |
|-------------------------------|------------------|------------------|-------------|
| 2 ×SuperTaq PCR mastermix     | 12.5 $\mu$ l     | 25 $\mu$ l       | 1 ×         |
| Forward Primer ( 10 $\mu$ M ) | 1-2.5 $\mu$ l    | 2-5 $\mu$ l      | 400-800 nM  |
| Reverse Primer ( 10 $\mu$ M ) | 1-2.5 $\mu$ l    | 2-5 $\mu$ l      | 400-800 nM  |
| Template                      |                  |                  | pg-ng       |
| ddH <sub>2</sub> O            | Up to 25 $\mu$ l | Up to 50 $\mu$ l |             |

#### 2. PCR reaction conditions

| No. of Cycles | Temperature | Time   | Step                 |
|---------------|-------------|--------|----------------------|
| 1             | 95 °C       | 2 min  | Initial denaturation |
|               | 95 °C       | 15s    | Denaturation         |
| 25-35         | 50-72 °C    | 15s    | Annealing            |
|               | 72 °C       | 15s/kb | Extension            |
| 1             | 72 °C       | 5 min  | Extension            |

3. detection : Take 2-5  $\mu$ l of the reaction mixture for electrophoresis to observe the results; products containing



dye can be directly subjected to agarose gel electrophoresis.

**Note:**

1. Template amount: 50-100 ng genomic DNA, 1-30 ng plasmid, or 1-2  $\mu$ l cDNA from RT-PCR reaction.
2. For simple templates such as plasmids, when amplifying DNA fragments  $\leq 3$  kb, the extension time can be set to 5 s/r. When amplifying 3-6 kb fragments, the amplification rate is 10-15 s/kb. For genomic or cDNA templates, the amplification rate is 15-20 s/kb; for amplification of high GC or templates with complex secondary structures, the recommended extension time is 30 s/kb.
3. Actual reaction conditions vary depending on the structure of the template, primers, etc. Optimal reaction conditions must be set based on the characteristics of the template, primers, and target fragment, and the reaction volume should be scaled up or down proportionally.

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