

## BlasTaq™ HotStart 2X PCR MasterMix

Cat. No. G598

Store at -20 °C.

## **Product Description**

BlasTaq™ HotStart 2X PCR MasterMix is a ready-to-use MasterMix containing abm's BlasTaq™ HotStart DNA Polymerase in a uniquely-formulated buffer with gel loading dye. This polymerase is a strategically-engineered, next generation Taq Polymerase that has rapid extension rates, robust performance, and contains a proprietary antibody that blocks polymerase activity at low temperatures. HotStart allows for a convenient reaction set-up at room temperature without non-specific amplification and primer dimer formation. With specialized reaction conditions, this polymerase provides increased processivity, yields, and sensitivity, while shortening reaction times by up to 70%, compared to wild-type Taq DNA polymerase.

During the initial denaturation step, the antibody dissociates from the DNA polymerase and restores enzyme activity. This feature significantly reduces non-specific product formation that would otherwise compete for reagent availability **offering higher specificity and improved yield of PCR products**. BlasTaq $^{TM}$  has 5'-3' polymerase and 5'-3' exonuclease activities, lacks 3'-5' exonuclease activity, and produces 3'-dA-tailed amplicons. PCR products made with BlasTaq $^{TM}$  can be used with TA cloning vectors.

| Product Component                               | Quantity          | Part No. |
|---|-------------------|----------|
| BlasTaq™ HotStart 2X PCR MasterMix <sup>1</sup> | 800 rxn (10.0 ml) | G598     |

<sup>&</sup>lt;sup>1</sup> Buffer contains 1.5 mM Mg<sup>2+</sup>.

## Protocol

1. Mix individual components before use and assemble reaction on ice.

| Product Component                  | Volume                        |  |
|------------------------------------|-------------------------------|--|
| BlasTaq™ Hotstart 2X PCR MasterMix | 12.5 µl                       |  |
| Forward Primer (10 µM)             | 1 µl                          |  |
| Reverse Primer (10 μM)             | 1 µl                          |  |
| Template DNA                       | Variable (100 ng genomic DNA) |  |
| Nuclease-free H <sub>2</sub> O     | Up to 25 µl                   |  |

Gently mix the reaction components, and briefly centrifuge. Run thermocycling conditions for standard PCR:

| Step                 | Temperature | Time      |
|----------------------|-------------|-----------|
| Initial Denaturation | 95 °C       | 10 min    |
|                      | 95 °C       | 15 sec    |
| 25 – 35 Cycles       | 60 °C²      | 15 sec    |
|                      | 72 °C       | 15 sec/kb |
| Final Extension      | 72 °C       | 1 min     |

<sup>&</sup>lt;sup>2</sup> The BlasTaq<sup>TM</sup> HotStart buffer allows for primer annealing at 60°C for most primers and adjust only if needed.

- 3. After PCR, maintain the reaction at 4 °C or store at -20 °C until use.
- 4. Analyze the amplification products by agarose gel electrophoresis.
- 5. Visualize by ethidium bromide or SafeView™ (Cat No. **G108**) staining.

## **General Notes**

- Optimized buffer for enhanced yields, sensitivity, and specificity, surpassing wild-type Tag polymerase.
- Reduce reaction times by up to 70% with a specialized protocol.
- For low yields, increase the reaction volume to 50 µl.
- For low yields or smearing, verify the quality of the template.