

# Absolute Master Mix

Cat No: AM-M-001-1250

Size: 1.25ml

Stored at 25°C up to one week, at 4°C up to six months and at -20°C up to one year.

**MOLEQULE-ON**<sup>®</sup>

## Description

Absolute Master Mix is a ready-to-use PCR reaction mixture. It only requires addition of primers, DNA template and water to carry out polymerase chain reaction. Absolute Master Mix contains *Taq* DNA polymerase, PCR Buffer, dNTPs and loading dyes. Absolute Master Mix is supplied at the 2X concentration to allow approximately 50% of the final reaction volume to be used for the addition of primer and template solutions. Components of Absolute Master Mix are tested for the absence of DNase, RNase and exonuclease activities. Recombinant *Taq* DNA polymerase is tested for the absence of exonuclease, and double- and single-stranded endonuclease activities.

## Recommendations

Since PCR is a powerful technique capable of amplifying trace amounts of DNA, all appropriate precautions should be taken to avoid cross-contamination. Ideally, amplification reactions should be assembled in a DNA-free environment. Use of filter tips is recommended. Special care should be taken to avoid contamination with primers or template DNA between individual reactions. PCR products should be analyzed in an area separate from the reaction preparation area.

## Protocol

### *Standard PCR with Absolute Master Mix:*

1. For each 20 µl reaction, add the following materials in a 0.2 ml PCR tube on ice just prior to use:

Absolute Master Mix	10µl
Forward primer, 5-10 µM	1µl
Reverse primer, 5-10 µM	1µl
DNA template	1µl
Add ddH <sub>2</sub> O to make volume up to	20µl
2. Mix gently and briefly centrifuge.
3. Cap the tubes tightly to avoid expulsion and evaporation.
4. Place the tubes in thermal cycler and process according to recommended thermal cycler conditions; initial denaturation at 94°C for 3-5 minutes, 30-40 cycles of denaturation at 94°C for 30 seconds, annealing ( $T_m$  of primer – 5) for 1 minute, extension at 72°C for 2 minutes and 1 cycle of final extension at 72°C for 7-15 minutes.
5. After the PCR reaction, load the PCR product on agarose gel directly.
6. Use the UV or blue-light to visualize the gel.

## Caution

1. During operation, always wear a lab coat, disposable gloves, and protective equipment.
2. Not intended for any animal or human therapeutic or diagnostic uses.