



Data Sheet (01.06.2005)

# mi-Hot Taq Mix

## 2x Hot start PCR Master Mix (for real-time PCR)

Source: *Thermus aquaticus*, strain YT-1

Cat.-No.	Size
mi-E8011	100 rxs

For research use!

2x 1.25 ml – for 100 Hot Start PCRs in a 50 µl volume.

### 2x Hot start PCR Master Mix Composition

- Taq DNA Polymerase (recombinant) in reaction buffer: 0.1 units/µl
- Antibodies to Taq DNA polymerase, concentration adjusted for the effective inhibition of polymerase activity at 37°C
- 32 mM (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>
- 130 mM TrisHCl, pH 8.8 at 25°C
- 0.02% Tween-20
- 3 mM MgCl<sub>2</sub>
- dNTP (dATP, dCTP, dGTP, dTTP; 0.4 mM each)

supplied with the mix: 1 ml tube MgCl<sub>2</sub> (100 mM)

### Unit definition for Taq polymerase

One unit is defined as the amount of the enzyme required to catalyze the incorporation of 10 nmol of dNTP into an acid-insoluble form in 30 minutes at 72°C.

### Endodeoxyribonuclease Assay

No detectable conversion of covalently closed circular DNA to nicked DNA was observed after incubation of 1 µg of pUC19 DNA with 25 µl of 2x mi-Hot Taq Mix in 50 µl for 4 hours neither at 37°C nor at 70°C.

Store at -20°C

### Description

2x mi-Hot Taq PCR Master Mix is an optimized ready-to-use PCR mixture of Taq DNA Polymerase, antibodies to Taq DNA polymerase, PCR buffer, MgCl<sub>2</sub> and dNTPs. The antibodies block polymerase activity during set-up of the PCR at room temperature (20-22°C). The inhibition of Taq DNA polymerase is completely reversed at a temperature above 70°. 2x Hot start PCR Master Mix contains all components for PCR, except DNA template and primers. It is tested for the absence of endodeoxyribonucleases and exodeoxyribonucleases.

### Applications

mi-Hot Taq Mix is tested for the amplification of a single-copy gene of mouse genomic DNA and recommended for complex genomic DNA or cDNA templates, low copy number of targets, large numbers of thermal cycles and multiplex PCR.

The mixture was shown to be effective for real-time PCR.

### Protocol for PCR with mi-Hot Taq Mix

Due to the inhibition of polymerase activity by anti Taq DNA polymerase antibodies, all reactions may be set up at room temperature, without increase of unspecific products or primer-dimer formation.

50 µl reaction volume:	Volume	Final concentration
2x mi-Hot Taq Mix	25 µl	1x
Forward Primer	variable	0.1-1 µM
Reverse Primer	variable	0.1-1 µM
Template DNA	variable	10 pg - 1 µg
Sterile deionized water	up to 50 µl	

Mix thoroughly and centrifuge briefly down.

Overlay the sample with mineral oil or add an appropriate amount of wax if the thermocycler is not equipped with a heated lid. Place the samples in a thermocycler and start the PCR program.