

**Description** SuperHot Master Mix (2x) is an optimized ready-to-use PCR mixture of *Taq* DNA Polymerase, antibodies for *Taq* DNA polymerase, PCR buffer, MgCl<sub>2</sub> and dNTPs. 2x PCR Master Mix contains all components for PCR, except DNA template and primers. The mixture was shown to be effective for Real Time PCR. SuperHot Master Mix (2x) contains SuperHot *Taq* DNA Polymerase of Bioron and has a high sensitivity and specificity.

### Content

Ref No.	S119102	119102	119110	color
<b>SuperHot Master Mix (2x) *</b>	<b>Sample size</b>	<b>200 reactions</b>	<b>5x200reactions</b>	<b>white</b>
<b>MgCl<sub>2</sub> 100 mM</b>	<b>1 mL</b>	<b>1 mL</b>	<b>5x 1 mL</b>	<b>green</b>
<b>PCR Water</b>	<b>1.8 mL</b>	<b>2x 1.8 mL</b>	<b>10x 1.8 mL</b>	<b>transparent</b>
<b>Datasheet</b>	<b>1</b>	<b>1</b>	<b>1</b>	<b>--</b>

\* Contains Antibody blocked Hotstart *Taq* DNA Polymerase (recombinant), NH<sub>4</sub>-PCR Buffer with 3 mM MgCl<sub>2</sub> and dNTP 400 μM each.

**Applications:** SuperHot Master Mix (2x) is suitable for a wide range of PCR methods like qPCR, Real-Time PCR and classic PCR. It can be used for regular PCR with a fragment-size up to 5 kb.

**Concentration:** 2x

**Sensitivity:** high

**Unit definition:** One unit of activity is defined as the amount of enzyme required to incorporate 10 nmoles of dNTP into acid-insoluble DNA fraction in 30 minutes at 72 °C.

**Additionally provided:** 1 tube MgCl<sub>2</sub> (100 mM)

**Recommended MgCl<sub>2</sub> concentration:** 1.5 mM – 6 mM

### Quality control

- 98% protein homogeneity in 10% SDS-PAGE
- No detectable exo-/endonuclease activities
- PCR amplification tests with different templates

**Storage condition:** -20 °C

**Pipetting scheme and thermocycler protocol:**

Components	Volume /25 $\mu$ L PCR-Reaction	Final concentration
2X PCR Master Mix	12.5 $\mu$ L	1X
Forward Primer	variable	0.1 – 1 $\mu$ M
Reverse Primer	variable	0.1 – 1 $\mu$ M
Template DNA	variable	100 pg – 1 $\mu$ g
Sterile dest. water	up to 25 $\mu$ L	-

Due to the inhibition of polymerase activity at room temperature by Anti Taq DNA polymerase antibodies all reactions may be settled-up at room temperature, it will not result in increase of unspecific product or primer-dimers formation.

If MgCl<sub>2</sub> is not added to the reaction mixture, final concentration of MgCl<sub>2</sub> in the reaction mixture will be 2.25 mM.

**Thermocycler protocol**

step	time	temperature
initial denaturation	2 minutes	94 °C
Number of cycles: 25 - 35		
denaturation	10 - 30 seconds	94 °C
annealing	20 - 30 seconds	55 - 68 °C *
extension	1 minute	72 °C

\* Usually the optimal annealing temperature is 5 °C below the melting temperature of the primers

**Notes:**

Program the cycler according to the manufacturer's instructions.

Each program should start with an initial denaturation step at 94 °C for 2 to max. 5 min.

Recommended elongation time is 1 min per 1 kb of target.

For maximum yield and specificity, temperatures (annealing) and cycling times should be optimized for each new template target or primer pair.