

Instrument	Final ROX Conc.
ABI 7000, 7300, 7700, 7900HT	500 nM
ABI 7500, Stratagene Mx3005P, Mx4000	50nM

Pack size:

2x 1,25 ml - sufficient for 100 HotStart PCR reactions in 50 µl reaction volume

Performance and purity tests :

Tested for the absence of endodeoxyribonucleases and exodeoxyribonucleases. The 2x HotRox Master Mix is tested in the amplification of a single-copy gene of mouse genomic DNA.

Endodeoxyribonuclease Assay:

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

Application:

- PCR
- Primer extension
- Multiplex PCR
- Low-copy targets PCR
- Real-time PCR

Associated activities:

Endonuclease and exonuclease activities were not detectable after 2 hours incubation of the mixture with 0.22 mg of EcoR I digested lambda DNA at 72°C in the presence of 15-20 units of enzyme in HotRox Master MIX

Storage conditions:

Store HotRox Master MIX frozen at -20°C and protect from light.

Protocol for PCR with SuperHot PCR Master Mix

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.

Add in a thin walled PCR tube:

component	50 µl reaction volume		25 µl reaction volume	
	volume	final conc.	volume	final conc.
2x PCR Master Mix	25 µl	1x	12.5 µl	1x
Forward Primer	variable	0.1-1 µl	variable	0.1-1 µl
Reverse Primer	variable	0.1-1 µl	variable	0.1-1 µl
Template DNA	variable	10 pg-1µg	variable	10 pg-1 µg
Sterile Deionized Water	up to 50 µl	-	up to 25 µl	-

- Gently vortex the sample and briefly centrifuge to collect all drops to the bottom of the tube.
- Overlay the sample with mineral oil or add an appropriate amount of wax if the thermal cycler is not equipped with a heated lid.
- Place the samples in a thermocycler and start the optimal PCR program.

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