



THE ENZYME-COMPANY

2x dU-HoT Master Mix (2x)

Cat: 119202 100 rcs (delivered in 2 x 1.25 ml)

Description

2x dU-HoT Master Mix is an optimized ready-to-use PCR mixture of *Taq* DNA Polymerase, antibodies to *Taq* DNA polymerase, PCR buffer, $MgCl_2$ and dNTPs. dUTP in the Mixture provides the tool for carry-over protection of the sample with UDG (cat#111025) treatment. 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.

Features

- High sensitivity & specificity
- SuperHOTTM *Taq* DNA Polymerase of Bioron included (*Taq* Polymerase with antibodies versus *Taq* polymerase)
- dUTP to provide carry-over protection

Applications

Real-Time qPCR assays
PCR with End point analysis

Components and Storage condition

dU-HoT Master Mix (2x) 2 x 1.25ml for 100 rcs in 50 μ l or 200 rcs in 25 μ l
 $MgCl_2$ (100mM) 1ml
Store at -20°C. Protect from light. Avoid repeated freeze/thawing. Shipped on blue-ice.

Performance and purity tests

Tested for the absence of endodeoxyribonucleases and exodeoxyribonucleases.

The dU-HOT 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 μ l of 2x SuperHot PCR Master Mix with 1 μ g of pUC19 DNA in 50 μ l for 4 hours neither at 37°C nor at 70°C

Ordering Information

Catalog #	Pack size
119202	100 rcs (2x1,25ml)
119210	500 rcs (10x1,25ml)

Bioron GmbH

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Protocol for PCR with dU-HOT 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:

50µl reaction volume			25µl reaction volume	
Component	Volume	Final concentration	Volume	Final concentration
2X PCR Master Mix	25µl	1X	12.5µl	1X
Forward Primer	variable	0.1-1µM	variable	0.1-1µM
Reverse Primer	variable	0.1-1µM	variable	0.1-1µM
Template DNA	variable	10pg-1µg	variable	10pg-1µg
Sterile Deionized Water	up to 50µl	-	up to 25µl	-

- 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 a PCR program

Real-time PCR amplification is done on ABI@PRISM 7700, iQ5 Real Time PCR System (BioRad) or other appropriate machines for Real-time PCR suitable for use of intercalating dye chemistry. All samples are run in triplicate with the appropriate single PCR controls (no template, no primers). Always prepare 2 Master Mixes for gene of interest and control gene to be sure in experiment-to-experiment consistency.

Real-Time Cycler Conditions

step	time	temperature
initial denaturation	5 minutes	95 °C
40 cycles		
denaturation	15 seconds	95 °C
annealing	1 minute	60 °C

Version FH06.01.09

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