



THE ENZYME-COMPANY

2x dU-HoT Master Mix 100rx

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, MgCl₂ 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.

2x dU-HoT 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 (NH₄)₂SO₄
- 130mM TrisHCl, pH 8.8 at 25°C
- 0,02% Tween-20
- 3 mM MgCl₂
- dNTPs (dATP, dCTP, dGTP, dTTP): 0.4mM of each
- dUTP – 0,6mM

Pack size 2x 1,25ml – sufficient for 100 HotStart PCR reactions in 50 μl reaction volume **10x 1,25ml** – sufficient for 500 HotStart PCR reactions in 50 μl reaction volume. **Supplied with the enzyme:** 1ml tube MgCl₂ (100mM)

Performance and purity tests

Tested for the absence of endodeoxyribonucleases and exodeoxyribonucleases.

The 2x 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 25μl of 2x dU-HoT Master Mix with 1μg of pUC19 DNA in 50μl for 4 hours neither at 37°C nor at 70°C

Protocol for PCR with HotStart 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:

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.

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

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