TaqMan Master Mix

Description

TaqMan Master Mix (2x) is an optimized ready-to-use PCR mixture for TaqMan Assays. The master mix contains a HotStart Taq DNA Polymerase, optimized PCR buffer, $MgCl_2$ and dNTPs. Only DNA template, primers and TaqMan probe have to be added by the customer. The master mix may also be suited for other probe based Real Time PCR assays.

Content

Ref No.	S119104	119104	119108	color
Taq Master Mix (2x) *	**Sample size**	**200 reactions**	**5x 200 rcts**	**white**
$MgCl_2$ 100 mM	**1 mL**	**1 mL**	**5x 1 mL**	**green**
PCR Water	**1.8 mL**	**2x 1.8 mL**	**10x 1.8 mL**	**transparent**
Datasheet	**1**	**1**	**1**	**--**

* Contains HotStart Taq DNA Polymerase (recombinant), PCR Buffer with 8 mM $MgCl_2$ and dNTPs 400 µM each.

Applications: TaqMan Master Mix (2x) is suitable for all probe based Real-Time PCR applications 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 $MgCl_2$ (100 mM)

Recommended $MgCl_2$ 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

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Pipetting scheme and thermocycler protocol:

Components	Volume / 25 µL PCR-Reaction	Final concentration
2X PCR Master Mix	12.5 µL	1X
Forward Primer	variable	0.1 – 1 µM
Reverse Primer	variable	0.1 – 1 µM
Template DNA	variable	100 pg – 1 µg
Sterile dest. water	up to 25 µL	-

It's strongly recommended to settle-up all reactions on ice to avoid formation of unspecific products or primer-dimers formation.

If $MgCl_2$ is not added to the reaction mixture, final concentration of $MgCl_2$ in the reaction mixture will be 4 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.