



# 2x qPCR Probe Master Mix - low ROX

Article	Content
SL-9802R-smp	1 ml, 100 rxn × 20 µl
SL-9802R-5ML	5 x 1 ml, 500 rxn × 20 µl
SL-9802R-10ML	10 x 1 ml, 1000 rxn × 20 µl
SL-9802R-20ML	20 x 1 ml, 2000 rxn × 20 µl



Long-Term Storage at -20°C in the dark

Short-Term Storage at 4°C in the dark

#### DESCRIPTION

Our **primaQUANT PROBE 2x qPCR Master Mix** is an optimized ready-touse mixture for probe-based assays such as Taqman®, Beacons and MGBs. It contains a modified HotStart DNA Polymerase, as well as dNTPS and MgCl<sub>2</sub>. The sophisticated buffer system provides fast kinetics and target amplification even for difficult templates.

The **primaQUANT 2x Master Mix** contains all components - you just need to add primers and template DNA/cDNA.

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### **DID YOU KNOW?**

- primaQUANT PROBE is also offered as a blue-colored mix for better handling.
- This Master Mix contains low ROX at a final concentration of 50 nM and is also compatible with all QuantStudio™, Aria™, Q-Tower™ and Rotor-Gene™ real-time systems.
- For qPCR cyclers requiring other concentrations of ROX, primaQUANT PROBE is also available with high concentrations of ROX or without ROX.



# **Recommended Reaction Mixture per Well**

### **BEFORE YOU START**

- After thawing, please invert the Master Mix tube 6-8 times.
- **Do not vortex** the Master Mix to avoid damaging the enzyme.

Component	Stock Concentration	20 µl Reaction	10 µl Reaction	Final Concentration
2x primaQUANT Master Mix	2x	10 µl	5 µl	1x
Forward Primer	4 µM	1µl	0.5 µl	200 nM (100 - 400 nM recommended)
Reverse Primer	4 µM	1µl	0.5 µl	200 nM (100 - 400 nM recommended)
Probe	8 µM	1µl	0.5 µl	400 nM (200 - 600 nM recommended)
Template DNA	-	variable	variable	0.1 - 10 ng per reaction
Sterile Water	-	adjust to 20 µl	adjust to 10 µl	-



### -Ù- NOTE

For maximum efficiency and specificity, adjustments of annealing temperature as well as extension time, primer/probe concentration and template concentration may be needed.

### CALCULATOR TOOL

Please feel free to download our Excel sheet calculator to calculate the necessary volumes: calculator.steinbrenner-laborsysteme.de.





# **Standard Protocol**



- For the majority of qPCR assays, standard cycling conditions can be applied for the majority of qPCR assays out-of-the box.
- However, cycling conditions strongly depend on the primer, probe, amplicon and input material and thus some of these factors might need adjustments.

#### **3-STEP PROTOCOL**

Step	Time	Temperature	
Initial Denaturation	1 - 3 minutes	92°C - 95°C	
Denaturation	5 - 10 seconds	92°C - 95°C	
Annealing	1 - 5 seconds	60°C depending on primer	25 - 40 cycles
Extension	10 - 20 seconds	72°C	

#### **2-STEP PROTOCOL**

Step	Time	Temperature	
Initial Denaturation	1 - 3 minutes	92°C - 95°C	
Denaturation	5 - 10 seconds	92°C - 95°C	
Annealing / Extension combined	10 - 20 seconds	60°C depending on primer	25 - 40 cycles



# **Ultra-fast Protocol**



- Ultra-fast cycling conditions highly depend on the ramping rate of your qPCR cycler, primer, probe, amplicon and input material and thus might need adjustments.
- Ultra-fast cycling conditions can be applied for the majority of qPCR assays out-of-the box, provided that your primer/probe sets do not show unspecific binding.

#### **3-STEP PROTOCOL**

Step	Time	Temperature		
Initial Denaturation	1 minute	92°C - 95°C	A CEE	
Denaturation	1 - 5 seconds	92°C - 95°C		
Annealing	1 - 5 seconds	60°C depending on primer	25 - 40 cycles	
Extension	1 second	72°C		

#### **2-STEP PROTOCOL**

Step	Time	Temperature	
Initial Denaturation	1 minute	92°C - 95°C	
Denaturation	1 second	92°C - 95°C	
Annealing / Extension combined	1 - 5 seconds	60°C depending on primer	25 - 40 cycles

### MANUAL



### S primaQUANT PROBE

# **Applications**

Probe-based quantitative PCR

- TaqMan® Probes
- Any kind of Dual-Labeled Hydrolysis Probe
- Molecular Beacons
- Scorpion Probes

DNA Genotyping DNA SNP Analysis RNA and miRNA Expression Multiplexing (up to 4 colors) Transcript Variant Analysis

#### QUALITY CONTROL PROCEDURE

Our **primaQUANT PROBE 2x qPCR Master Mix** undergoes stringent quality controls. Each lot is tested in a probe-based qPCR with cDNA and lambda DNA input.

Enzyme purity and homogenity of > 98 % is validated using a Bioanalyzer SDS protein electrophoresis.

All **primaQuant** Master Mixes are free of detectable endonuclease- & exonuclease activity:

- Incubation of 1 µg of plasmid DNA with 5 U for 4h at 37°C and 72°C
- Incubation of 1 μg of a DNA size standard with 5 U for 4h at 37°C and 72°C

For more information, please visit our website: www.steinbrenner.de



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# **Further Products**

Products that may also interest you

#### **REVERSE TRANSCRIPTION KIT**

S primaREVERSE RT-KT

For the efficient cDNA synthesis out of total RNA extractions try the primaREVERSE RT-Kit with article number SL-9540.