



# 2x Blue qPCR Master Mix with SYBRGreen - high ROX

Article	Content
SL-9902HRB-smp	1 ml, 100 rxn × 20 µl
SL-9902HRB-5ML	5 ml, 500 rxn × 20 μl
SL-9902HRB-10ML	10 ml, 1000 rxn × 20 µl
SL-9902HRB-20ML	20 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 CYBR Blue 2x qPCR Master Mix** is an optimized readyto-use mixture for SYBRGreen-based assays. It contains a modified HotStart DNA Polymerase, SYBRGreen dye, 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. The **blue color does not interfere** with the qPCR reaction but helps you to track wells already filled during pipetting steps.

# Ŋ. DID YOU KNOW?

- This Master Mix contains high ROX at a final concentration of 500 nM.
- primaQUANT CYBR is also available with low concentrations of or without ROX.



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## **Recommended Reaction Mixture per Well**

### ပ်ဴ- 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)
Template DNA	-	variable	variable	0.1 - 10 ng per reaction
Sterile Water	-	adjust to 20 µl	adjust to 10 µl	-



### 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.







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## **Standard Protocol**



- Standard cycling conditions can be applied for the majority of qPCR assays out-of-the box.
- However, cycling conditions highly depend on the primer, probe, amplicon and input material and thus might require adjustments.

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

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



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# **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 require 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		9	
Initial Denaturation	1 minute	92°C - 95°C		
Denaturation	1 - 5 seconds	92°C - 95°C		
Annealing	1 - 5 seconds	60°C depending on primer	25 - 40 cycles	
Extension	5 seconds	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	5 - 10 seconds	60°C depending on primer	25 - 40 cycles

### MANUAL



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# **Applications**

Generally dye-based quantitative PCRs DNA Genotyping DNA SNP Analysis RNA Expression Single-Color Multiplexing Telomerase Length Assays miRNA Expression Analysis Transcript Variant Analysis

### QUALITY CONTROL

Our **primaQUANT CYBR 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 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

#### FURTHER INFORMATION

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.