

SYBR Green PCR SuperMix

Description

Catalog No.	Specification	SYBR Green PCR SuperMix	
100EQ	20µL—500 rxns	4 x 1.25mL	1n

Advantage

- 1) Quickly get results, saving up to 50% of the time
- 2) Optimized ready-to-use master mix for rapid PCR reactions
- 3) Accurate detection of various starting amounts of templates, stable amplification, quantitative results with high repeatability
- 4) Balanced K⁺ and NH₄⁺ ion ratios, as well as stand-alone ROX Reference Dye packaging for all real-time PCR instruments

Introduction

SYBR Green PCR SuperMix is an optimized 2x real-time PCR master mix containing HotStarTaq DNA Polymerase, SYBR Green® fluorescent dye, dNTP and Mg²⁺. In addition, the balanced K⁺ and NH₄⁺ ion ratios in the buffer promote specific primer annealing. To ensure a highly sensitive and specific PCR reaction, the reaction can be initiated by simply adding the primer and cDNA template to the ready-to-use PCR master mix. The unique PCR buffer ensures sensitive qPCR on all real-time PCR instruments without optimization.

Kit Components

Component	Character	Advantage
HotStarTaq DNA Polymerase	Heating at the pre-denaturation temperature for 30s will completely inactivate the blocking antibody and release the DNA polymerase activity.	Effectively suppresses non-specific amplification caused by primer-dimer annealing
SYBR Green qPCR Buffer	Suitable for all real-time PCR instruments	qPCR run time is reduced, results are obtained faster, PCR reactions can be completed in one day

SYBR Green I dye	Strong fluorescence signal when combined with DNA duplex	High sensitivity amplification provides a wide-area linear range, a Ct value of 5-35 and a sensitivity for single-digit detection, and is suitable for melting curve analysis.
ROX dye	Calibration of fluorescent signals on ABI and Agilent PCR machines	Calibration of PCR machines require ROX dyes does not affect reaction results

Kit principle

SYBR Green PCR SuperMix provides a wide range of specific, sensitive assays for standard and rapid PCR machines. The SYBR Green I dye in the master mix can analyze multiple target nucleic acids without the need to synthesize sequence-specific probes. The special fast PCR buffer can greatly shorten the denaturation, annealing and extension time, and has good applicability to complex templates, templates with more PCR inhibitor residues (such as soil and fecal DNA), and long-range amplification. In addition, HotStarTaq DNA Polymerase can be activated by heating at 95°C for 30sec, requiring a strict hot start to avoid nonspecific products.

Kit application

SYBR Green PCR SuperMix Can be used for gene expression analysis of cDNA, plasmids, gDNA, absolute quantitative analysis. It is suitable for various real-time PCR machines, including ABI, Bio-Rad, Eppendorf, Roche and Agilent PCR machines.

Attention

1. Template

cDNA: For two-step quantitative qPCR, Use 10¹µL of cDNA reverse transcribed from total RNA (10pg to 1ng).

In the 20¹µL reaction system, the amount of cDNA template used is generally not more than 100 ng.. It should be noted that when detecting high-abundance genes in undiluted cDNA, the Ct value in quantitative PCR results may be too low, which may affect the accuracy of quantification. Gradient dilution of the cDNA template results in more accurate results.

Plasmid and genomic DNA: 100pg to 1ng of genomic DNA or 10⁻¹⁰ copies of plasmid DNA can be used in a 20¹µL system.

1. Transportation and storage

- Ice bag, dry ice
- Store at -20 °C in the dark. This product contains the fluorescent dye SYBR® Green I. When storing or formulating the reaction system, avoid strong light. Please mix it upside down before
- For your safety and health, please wear a lab coat and wear disposable gloves when performing the

Reaction System

A reaction system as described below was established. To perform multiple reactions, prepare a premix of the common components, add a suitable volume to each tube or well, and then add a special reaction component (eg, template)

96 wells

Composition

	50 µL reaction system	20 µL reaction system
2 x SYBR Green PCR Master Mix	25 µL	10 µL
PCR Forward Primer (10 µM)	1 µL	0.4 µL
PCR Reverse Primer (10 µM)	1 µL	0.4 µL
Template		
*50 x ROX Dye(Optional)	1 µL	0.4 µL
RNase-Free ddH ₂ O	to 50 µL	to 20 µL

1. It is recommended to use a 20 µL or 50 µL system to ensure the validity and repeatability of the amplification of the gene of
2. Cover or seal the reaction tube/PCR plate and mix gently. It can be centrifuged slightly to ensure that all components are at the bottom of the
3. Place the reaction system in a real-time PCR instrument, collect data and analyze the results. Set up your PCR instrument as shown in the table Optimum temperature

The incubation time can be determined by the specific situation.

ROX dye

The fluorescent signal in the reaction system can be standardized by adding a ROX dye to the reaction system according to the selected instrument. The table below lists the amount of ROX required per unit of operation (per 50 µL of reaction system):

Instrument	The amount of ROX required
ABI7300 • 7900HT • StepOne etc.	5 µL

ABI7500, 7500Fast, ViiA7, Stratagene Mx3000, Mx3005P

and Mx4000 etc.

1 µL

Roche, Bio-Rad, Eppendorf etc.

No need to add

Two-step amplification procedure:

Stage	Number of cycles	Temperature	Time
Pre-denaturation	1x	95 °C	30 sec
Denaturation	1	95 °C	5 sec
Annealing/extension	35-40x	60 °C	30 sec
Melting Curve stage			

Three-step amplification procedure:

Stage	Number of cycles	Temperature	Time
Pre-denaturation	1x	95 °C	30 sec
Denaturation	1	95 °C	5 sec
Annealing	35-40x	50-60 °C	30 sec
extension		72 °C	30 sec
Melting curve analysis(Melting Curve stage)			

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