SYBR Green I for PCR

Technical literature is available at: www.mesgenbio.com. E-mail MesGen Technical Services if you have questions on use of this system: tech@mesgenbio.com

MESGEN INNOVATION BIOTECHNOLOGY

Catalog Number : MPY6155 **Lot Number :** Refer to vial **Packaging Size :** 1000μL☐5000μL☐

Ex (nm) : 497 **Em (nm) :** 525 **Solvent :** DMSO

Background

SYBR Green I, is a very sensitive dye for the detection of double stranded DNA (dsDNA), So it has been widely used in non-specific detection of amplification in realtime qPCR experiments. The double-strand DNA-specific SYBR Green I fluorescent reporter offers distinct advantages. SYBR Green I dye is inexpensive, easy to use, and sensitive. Well-designed primers must be used in SYBR Green quantitative RT-PCR reactions because SYBR Green I dye will detect nonspecific products, resulting in an overestimation of the target concentration.

Storage instruction

2-8°C & Protect from light

1. The flowing table 1 is our SYBR Green I RT-PCR Reagents Kit recipe for reference only. Please optimize it by yourself.

Reagent	Final concentration in the mix
dNTP	0.25mM
Tween20	1%
BSA	0.1% vol
Tris(pH8.4)	50mM
Hot-start Taq DNA polymerase	1.25 u per reaction
NH4Cl	10mM
KCl	20mM
MaCl2	2.5mM
Sybr Green	2X

2. On ice, prepare a 2x master mix containing no DNA, by mixing the components in the following order : water, DMSO, Taq polymerase buffer, dNTPs, MgCl2, Sybr Green, Taq polymerase.

3. Transfer 2x SYBR Green PCR Master Mix to tubes or plates. Add RNase-free water. Mix the individual solutions.

4. Prepare a reaction mix according to Table 2.

Due to the hot start, it is not necessary to keep samples on ice during reaction setup or while programming the real-time cycler.

PCR Reaction Setup: **Table 2**

DNA	Template DNA (<500 ng/reaction)
SYBR Mix	25.0 μL
Primer 1	2ul（5uM)
Primer 2	2ul（5uM)
dd H_2O	Added to 50.0 μL

Program your real-time cycler according to the program outlined in **Table 3**

PCR initial activation step	Heating cycling Number of cycles 40
95 °C 5min	96 °C 10s 60 °C 30s

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