WIRAGEN
Enseignement et Recherche

TECHNICAL SHEET:

2x SYBR-Green PCR Mastermix

Ref : PW05-01

Specification: 50T

Storage: Store at a constant temperature of -20°C, valid for one year.

Product Description:

This system is a premixed system for real-time fluorescence quantification of the dye method (SYBR Green I). The product contains components such as HotStart Taq DNA Polymerase, dNTPs, Mg2+, reaction buffer and stabilizer at optimized concentrations. It is mainly used for the detection of genomic DNA target sequence and cDNA target sequence after RNA reverse transcription, such as gene expression analysis, copy number analysis, SNP genotype analysis, etc., and is suitable for different types of fluorescent quantitative PCR.

This system is a 2× premixed real-time fluorescence quantitative PCR reaction system. When using it, you only need to add templates, primers and water to make the appropriate concentration 1×, and the reaction can be carried out. It has the advantages of fast and simple, high sensitivity, strong specificity, and good stability, which can minimize human errors, save PCR experiment operation time, and reduce the chance of contamination.

The ROX reference dye is provided in separate tube and can be added if using a cycler that require ROX as a passive reference dye.

Steps:

Standard operating procedure:

1. Prepare the reaction system according to the following table

Component	25 µl Volume	50 µl Volume	Final concentration
2x SYBR-GREEN PCR Master mix	12.5 µl	25 µl	1 x
Primer 1 (10 µM)	0.5 – 2.5 µl	1 – 5 µl	0.2 ~ 1.0 µM
Primer 2 (10 µM)	0.5 – 2.5 µl	1 – 5 µl	0.2 ~ 1.0 µM
Template DNA	5 µl	10 µl	-
ddH2O	-	-	-
Total Volume	25 µl	50 µl	-

Téléphone : 041 40 42 27 / 06 59 19 60 33
Facebook/Instagram : Wiragen Academy
Site : www.wiragen.com
E-mail : contact@wiragen.com / wiragen.academy@gmail.com
Siège social : Local 01 lotissement Si Redouane 02 N°16 Senia
Siège formation : 12 Rue de Nancy, Résidence Col.Othmane, Oran

PCR Cycling Protocol

- **Two Step Cycling**

Pre-denaturation	Denature	Anneal / Extend
1 CYCLE	35-45 CYCLES	
95°C	95°C	60°C
10 min	10 – 20 S	20 – 60 S

- **Three Step Cycling**

Pre-denaturation	Denature	Anneal	Extend
1 CYCLE	35-45 CYCLES		
95°C	95°C	56 – 64°C	72°C
10 min	10 – 20 S	10 – 30 S	10 – 60 S

Note :

1- For Roche Light Cycler480, the pre-denaturation time suggest 10min, ABI7500 suggest 5mins.
2- If the annealing temperature is lower or exceed 200bp, please use the three-step method.
3- Use Special areas and pipettes before and after amplification. Wear gloves and change them frequently. Do not open the reaction tube after the completion of PCR reaction to minimize contamination of PCR products to samples