ClearPeak 2x SYBR Master Mix, No ROX

100 rxn, 500 rxn

Cat No: SMMN1, SMMN5

Shipping : Ship with blue ice.
Storage : Store at -20°C. Avoid freeze and thaw cycles. *ClearPeak* 2x SYBR Master Mix, No ROX can be stably stored for 6 months under dark conditions at 2~8°C after thawing.

General Information

ClearPeak 2x SYBR Master Mix, No ROX# is a specialized premix for real-time fluorescence quantitative qPCR reactions using the dye method (SYBR Green I). The core component *EcoTaq* DNA Polymerase is an antibody based hot start DNA polymerase that can be restored by heating at 95°C. It has many advantages such as strong specificity and high detection sensitivity, and is paired with an optimal buffer optimized for qPCR. The unique qPCR buffer system of this product, combined with hot start enzymes, effectively inhibits the production of non-specific products and significantly improves the amplification efficiency of qPCR. It is very suitable for high specificity and sensitivity qPCR reactions. This product is also suitable for qPCR rapid reaction program. Good standard curves can be obtained within a wide quantitative range, with accurate, reproducible, and reliable quantification of target genes.

 # Instruments do not require ROX correction: LightCycler® 480, LightCycler® 96, LightCycler® 2.0, Bio-Rad iCyler iQ, iQ5, CFX96, etc.

SYBR PCR Setup

Component	Amount
ClearPeak 2x SYBR Master Mix, No ROX	10 µl
Forward Primer, 10 µM*	0.4 µl
Reverse Primer, 10 µM*	0.4 µl
Template cDNA**	2-5 µl
ddH_2O	up to 20 µl
Total	20 µl

*200 nM of primer final concentration is applicable for most cases. The concentration can be adjusted within 0.1~1.0 µM when amplification efficiency is not satisfactory.

**Too much or too little template used may lead to inaccuracy of quantitative result. A range of 1-100 ng is recommended to result in a good Ct value (15<Ct<35). If template is stocked at high concentrations, dilute it prior to loading to prevent possible loading errors.

PCR Reaction Condition

Two Step

Temperature	Time	Cycles
95°C	5-10 min***	
95°C	15 sec	40
60°C	30-60 sec	

***Please note that the hot-start polymerase in this system needs to be activated at 95°C for 5 minutes prior to amplification. If the amplicon sequence is GC-rich, the time for pre-denaturation/enzyme activation can be prolonged to 10 minutes.

For further information;
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Three Step

Temperature	Time	Cycles
95°C	5-10 min	
95°C	15 sec	
50°C-60°C	30 sec	40
72°C	30 sec	

Important Notes:

- Please gently mix the tube upside down before use and avoid foaming as much as possible. Avoid repeated freeze-thaw, as repeated freeze-thaw may cause a decrease in product performance.
- Due to the presence of fluorescent dye SYBR Green I in this product, it is necessary to store it away from light to avoid strong light exposure when preparing the reaction.
- In practical operation, corresponding improvements and optimizations should be made to PCR Setup and PCR Reaction Conditions based on different templates, primer structures, and target fragment sizes.
- Usually, the amount of DNA templates is based on 10-100ng genomic DNA or 1-10ng cDNA. Due to the different copy numbers of target genes contained in templates of different species, gradient dilution can be performed on the templates to determine the optimal template usage.
- Typically, the final concentration of the primer is 0.2μM can achieve successful results, where a final concentration of 0.1-1.0μM primer may serve as a reference for setting the range. In the case of low amplification efficiency, the concentration of primers can be increased; When non-specific reactions occur, the concentration of primers can be reduced to optimize the reaction.
- Due to the high sensitivity nature of the qPCR reaction, contamination of air or aerosols may lead to reaction failure or result inaccuracy. Please set up the qPCR reaction in a clean environment using filtered tips, and sterilized tubes and pipette sets.