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Version: 04/2024

SuperPro Multiplex PCR Mix

Cat. No. : CW3314S (1 mL)

Storage Condition: -20 ℃, avoid repeated freeze-thaw.

Components

Component	CW3314S 1 mL
2.5×SuperPro Multiplex PCR Mix	1 mL
ddH_2O	1 mL

Introduction

SuperPro Multiplex PCR Mix is a premixed system suitable for various types of multiplex PCR, with a concentration of 2.5×. It contains DNA polymerase, PCR Buffer, dNTPs, Mg^{2+}, stabilizer and enhancer and other components. This product only needs to add primers and templates for amplification. The operation is simple and convenient, reduces the probability of contamination, improves the detection flux and reproducibility.

The DNA polymerase contained in SuperPro Multiplex PCR Mix is a genetically engineered recombinant enzyme with 5'-3'DNA polymerase activity and no 5'-3' exonuclease activity. DNA polymerase is an antibody-modified hot start enzyme. It can effectively reduce the non-specific amplification caused by the non-specific binding of primer and template or primer dimer at room temperature. Meanwhile, it has the advantages of short activation time, strong amplification ability, high sensitivity and good stability. The combination of the unique PCR buffer system and the thermal starter enzyme significantly improves the amplification efficiency of PCR with higher sensitivity and stronger inhibitor tolerance.

SuperPro Multiplex PCR Mix has a wide range of applications and is suitable for various types of multiple PCR, such as microsatellite analysis, amplicon library preparation, genotyping and SNP detection.

Notes

1. Before use, please gently mix the product upside down after it is completely melted, and use it after a short centrifugation.
2. Avoid repeated freeze-thaw, which may degrade product performance. This product can be stored at -20 ℃ for longer storage.

Protocol

1. PCR reaction system

   Extracted DNA amplification reaction system:

Reagent	25 μL System	50 μL System	Final Concentration
2.5×SuperPro Multiplex PCR Mix	10 μL	20 μL	1×
5×Primer Mix	5 uL	10 μL	1×
Template DNA	X μL	X μL	
ddH_2O	to 25 μL	to 50 μL	

Note:When designing primers, the difference between Tm of each primer should be minimized, and the difference should be controlled within 5 ℃.The final concentration of each primer pairs of 0.05-0.2 μM can be set as a reference for setting range. The primer concentration can be increased when the amplification efficiency is not high, and the primer concentration can be reduced in the case of non-specific reactions, so that the reaction system can be optimized. In order to achieve the best amplification effect, it is recommended that the primer mixture be used after vortex for 10 s and short centrifugation.

2. PCR reaction program

Step	Temperature	Time	Cycle Number
Pre-denaturation	95 ℃	2 min	1
Denaturation	95 ℃	10 s	30-40
Annealing	55-65 ℃	30 s	
Extension	72 ℃	1 kb/min	
Terminal extension	72 ℃	5 min	1

Note:1) In general experiments, the annealing temperature is 5 ℃ lower than the melting temperature (Tm) of the amplification primer, and the annealing temperature should be appropriately reduced when the ideal amplification efficiency cannot be obtained. When nonspecific reactions occur, increase the annealing temperature to optimize reaction conditions

2) The number of cycles can be set according to the downstream application of the amplification product. If the number of cycles is too low, there is insufficient amplification. If the number of cycles is too high, the mismatch rate increases, leading to significant nonspecific background. Therefore, the number of cycles should be minimized on the premise of ensuring the yield of the product.

3) PCR products are easy to produce aerosol contamination, which leads to inaccurate and unreliable results. It is recommended to physically separate PCR reaction system preparation area and PCR reaction area, use special pipette and other equipment, and clean each experimental area regularly (0.5% sodium fluoride or 10% bleaching agent should be used to clean) to ensure the reliability of the experimental results.