2×Super Kfx MasterMix

Catalog Number:

S665683 (1 mL)
S665683 (5 mL)

Storage conditions: -20°C. If frequent use is required, store at 2-8°C.

Products content

Component	1 mL	5 mL
2×Super Kfx MasterMix	1 mL	5 x 1 mL
ddH2O	1 mL	5 x 1 mL

Products Introduction

This product is a premixed system consisting of Super Kfx DNA Polymerase, Mg2+, dNTPs, PCR stabilizers and enhancers at a concentration of 2×. Super Kfx DNA Polymerase is a fast, high-fidelity DNA polymerase with high amplification efficiency, which possesses 5′-3′DNA polymerase activity and 3′-5′ exonuclease activity. With 5′-3′ DNA polymerase activity and 3′-5′ exonuclease activity, the enzyme has the advantages of strong amplification ability, high fidelity and high specificity, etc. 2×Mix has added unique amplification enhancement factors and extension factors, and the unique formula makes the whole reaction system very stable and easy to operate, which is suitable for the amplification of various fragments and templates. The product is suitable for gene cloning, second generation library amplification, targeted gene mutation, SNP amplification experiments.

quality control

No exogenous nuclease activity, can effectively amplify various templates; stored at 2-8℃ for one month, no obvious activity change.

Usage

The following examples are conventional PCR reaction systems and conditions, which should be improved and optimized according to the template, primer structure and target fragment size.

1. PCR reaction system

All operations should be carried out on ice, please mix the components thoroughly after thawing, after use, please promptly return to -20 ℃ for storage.

reagents	50 μL reaction system	final concentration
2×Super Kfx MasterMix	25μL	1×
Forward Primer, 10 μM	2 μL	0.4 μM
Reverse Primer, 10 μM	2μL	0.4 μM
Template DNA, moderate amount	appropriate amount	<500 ng/50 μL
ddH2O	up to 50 μL	

2. PCR reaction system

move	temp	timing	
premutability	98°C	30 s-3 min	
denaturation	98°C	10-30 s	25-35 min cycle
annealing (metallurgy)	According to the primer Tm	15-30 s	25-35 min cycle
reach	72°C	4-6 kb/min	25-35 min cycle
ultimate extension	72°C	5 min	

take note of

1) Priority is given to three-step amplification; if the three-step method fails to amplify the target product or if the primer Tm value is greater than 68° C, try the two-step method.

2) Denaturation: pre-denaturation of simple templates 98° C, 30s-1min, for complex templates, the pre-denaturation time can be extended to 3min.

3) Annealing: In general, the annealing temperature is 3-5℃ lower than the Tm value of the primers. If the desired amplification efficiency cannot be obtained, the annealing temperature should be changed in a gradient to optimize the results; if a non-specific reaction occurs, the annealing temperature should be increased appropriately.

4) Extension: The extension time should be set according to the length of the amplified fragments and the complexity of the template, the amplification efficiency of this product is 4-6kb/min, for long fragments and templates with high complexity it is recommended that 2-4kb/min.

5) Cycling times: the number of cycles can be set according to the downstream application of the amplified product. If the number of cycles is too small, the amplification will be insufficient, and if the number of cycles is too large, the chance of mismatch will be increased, so the number of cycles should be minimized under the premise of guaranteeing the yield of the product.