

2 x GoldStar MasterMix

Item No. G665843 (5 ml)

G665843 (25 ml)

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

Product content

individual parts making up a compound	G665843 5 ml	G665843 25 ml
2 x GoldStar MasterMix	5 x 1 ml	5 x 5 ml
ddH2O	5 x 1 ml	5 x 5 ml

Product Introduction

2×GoldStar MasterMix is a premixed system consisting of GoldStar DNA Polymerase, PCR Buffer, Mg²⁺, dNTPs and PCR stabilizers and enhancers. The pre-mixed PCR mixture makes the operation easier and faster, and minimizes the human error and contamination. The GoldStar DNA Polymerase contained in this product is a chemically modified, new high-efficiency Taq DNA Polymerase, the activity of the enzyme is completely blocked at room temperature, so that the enzyme is inactive at low or room temperature, thus effectively avoiding the non-specific amplification caused by the non-specific binding of the primer and the template or the dimerization of the primer under the condition of room temperature, the activation of the enzyme must be at 95°C, and the enzyme is activated at 95°C. To activate the enzyme, incubate at 95°C for 10 minutes. The unique buffer system allows the enzyme to be used in a wide range of applications, enabling efficient amplification of templates with high GC content, complex secondary structures, and low-copy templates. The unique MasterMix formula makes the whole reaction system more stable. PCR amplification with this product has an "A" base at the 3' end of the PCR product, which can be directly used for T/A cloning. The product does not contain any dye, and can be used for electrophoresis after the PCR program is finished by adding appropriate amount of sample buffer as needed. It can be used directly for downstream cloning or microarray hybridization after PCR amplification without the need of gel recovery to remove heterogeneous bands. It is mainly used in routine PCR, RT-PCR, multiplex PCR and gene chip detection, especially suitable for PCR reactions with high

specificity requirements.

Quality control

No exogenous nuclease activity detected; no host DNA residue detected by PCR; can effectively amplify single-copy genes in the human genome; stored at 2-8° C for three months, no obvious change in activity.

Usage

The following is an example of a PCR reaction system and reaction conditions for amplifying a 1 kb fragment of human genomic DNA as a template, which should be improved and optimized according to the structure of the template primers and the size of the target fragment in practice.

1. PCR reaction system

reagents	50 μ l reaction system	final concentration
2 x GoldStar MasterMix	25 μ l	1 \times
Forward Primer, 10 μ M	2 μ l	0.4 μ M
Reverse Primer, 10 μ M	2 μ l	0.4 μ M
Template DNA	<0.5 μ g	<0.5 μ g/50 μ l
ddH ₂ O	up to 50 μ l	/

Note: Please use the final concentration of 0.1-1.0 μ M as a reference for setting the range of primer concentration. If the amplification efficiency is not high, the primer concentration can be increased; if a non-specific reaction occurs, the primer concentration can be decreased to optimize the reaction system.

2. PCR reaction conditions

move	temp	timing	/
premutability	95° C	10 min	/
denaturation	95° C	30 s	30-40 cycles
annealing (metallurgy)	55-65° C	30 s	30-40 cycles

reach	72° C	60 s	30-40 cycles
ultimate extension	72° C	5 min	/

Attention:

(1) In general, the annealing temperature is 5°C lower than the melting temperature of the amplification primer T_m , and the annealing time is generally 30-60 s. When the desired amplification efficiency cannot be obtained, the annealing temperature should be lowered appropriately; and when a non-specific reaction occurs, the annealing temperature should be raised to optimize the reaction conditions.

(2) The extension time should be set according to the size of the amplified fragments. The amplification efficiency of GoldStar DNA Polymerase included in this product is 1-2 kb/min.

3) 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 amount will be insufficient; if the number of cycles is too large, the chance of mismatch will increase and the non-specific background will be serious. Therefore, the number of cycles should be minimized under the premise of ensuring the product yield.

(4) The enzyme should be activated under the condition of pre-denaturation at 95°C for 10 min.