

GoldStar DNA Polymerase

Item No. G665840 (2500 U)

Storage condition: -20°C

Product content

individual parts making up a compound	G665840 2500 U
GoldStar DNA Polymerase, 5 U/ μ l	5 $\times$ 100 μ l
5 $\times$ GoldStar PCR Buffer	5 x 1.9 ml

Note: The 5 $\times$ GoldStar Taq PCR Buffer for this product contains 8.5 mM magnesium ions.

Product Introduction

GoldStar DNA Polymerase is a chemically modified, new and highly efficient Taq DNA Polymerase. The activity of the enzyme is completely blocked at room temperature, making the enzyme inactive at low or room temperature, thus effectively avoiding non-specific amplification caused by the non-specific binding of the primer to the template or primer dimerization at room temperature, and the enzyme has to be incubated at 95°C for 10 minutes to activate. The activation of the enzyme must be incubated at 95°C for 10 minutes. The unique buffer system enables the enzyme to be used in a wider range of applications, allowing high GC content, complex secondary structures and low-copy templates to be amplified efficiently. 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 specificity of this product is strong, and it can be used directly for downstream cloning or microarray hybridization experiments without the need of gel recovery to remove heterobands after PCR amplification. It is mainly used in routine PCR, RT-PCR, Real-time PCR, multiplex PCR, gene chip analysis and SNP detection, and is especially suitable for PCR reactions with high specificity requirements.

Unit definitions

The amount of enzyme required to dope 10 nmol of deoxyribonucleotide into acid-insoluble material was defined as 1 activity unit (U) at 74° C for 30 min using activated salmon sperm DNA as template/primer.

Quality control

The purity of SDS-PAGE is more than 99%; no exogenous nuclease activity is detected; no residual host DNA is detected by PCR; it can effectively amplify single-copy genes in the human genome. It can effectively amplify single-copy genes in the human genome. No obvious activity change after one month of storage at room temperature.

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 template, primer structure and size of the target fragment in actual operation.

1. PCR reaction system

reagents	50 μ l reaction system	final concentration
5 $\times$ GoldStar PCR Buffer	10 μ l	1 $\times$
dNTP Mix, 10 mM each	1 μ l	200 μ M each
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
GoldStar DNA Polymerase, 5 U/ μ l	0.5 μ l	2.5 U/50 μ l
ddH ₂ O	up to 50 μ l	/

Note: For primer concentration, please use the final concentration of 0.1–1.0 μ M as a reference. 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 Taq 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 low, the amount of amplification will be insufficient; if the number of cycles is too high, 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.