

SuperFastStar DNA Polymerase

Cat. No. : CW3316S (500 U)
CW3316M (5 kU)
CW3316L (50 kU)

Storage Condition: -20°C

Components

Component	CW3316S 500 U	CW3316M 5 kU	CW3316L 50 kU
SuperFastStar DNA Polymerase (5 U/μL)	100 μL	1 mL	10 mL

Introduction

SuperFastStar DNA Polymerase is a hybrid of anti-Taq enzyme monoclonal antibody and Taq DNA Polymerase for Hot Start PCR. When using Taq enzyme antibody for PCR amplification, the combination of Taq enzyme antibody and Taq enzyme inhibits the activity of DNA polymerase before high temperature denaturation, which could effectively inhibit the non-specific annealing of primer and the non-specific amplification caused by primer dimer at low temperature. The Taq enzyme antibody is denatured in the initial DNA denaturation step of PCR reaction, and the polymerase activity is restored to achieve the hot start effect. The use of this product does not require special inactivation of Taq enzyme antibodies, and can be used under conventional PCR reaction conditions.

SuperFastStar DNA Polymerase has 5'-3' DNA polymerase activity and 5'-3' exonuclease activity, but no 3'-5' exonuclease activity. Enzyme extension rate is 2 kb/min, and can amplify fragments up to 5 kb. The amplified PCR product has an "A" base attached to the 3' end and can therefore be used directly for T/A cloning. The blocking rate of polymerase activity at a temperature of 55 °C and below is more than 95%, and DNA polymerase activity can be restored by heating at 95 °C for 5 s. This product has the characteristics of fast extension speed and high amplification efficiency, and is mainly suitable for DNA amplification with PCR, DNA sequencing and other experiments.

Activity Definition

Using activated salmon sperm DNA as template/primer, the amount of enzyme required to incorporate 10 nmol deoxynucleotides into an acidic insoluble substance at 74 °C within 30 minutes is defined as 1 unit of activity (U).

Quality Control

After multiple column purifications, purity exceeding 99% is confirmed through SDS-PAGE analysis. No exogenous nuclease activity is detected. It exhibits closure at 55 °C for 30 minutes with no polymerase activity. Complete activation is achieved at 95 °C for 5 seconds.

Protocol

The following example uses human genomic DNA as a template to amplify PCR reaction conditions and reaction systems of 1 kb fragments, and should be improved and optimized according to the template, primer structure and target fragment size in practice.

1. The recommended amount of enzyme in a 50 μ L PCR reaction is 0.25-0.5 μ L, equivalent to 1.25-2.5 U/50 μ L.
2. PCR reaction condition

Step	Temperature	Time
Pre denaturation	95 °C	5-60 s
Denaturation	95 °C	30 s
Annealing	55-65 °C	30 s
Extension	72 °C	30 s
Final extension	72 °C	2 min

Note:

- 1) The hot-start enzyme used in this product needs to be incubated at 95 °C for at least 5 s to activate the enzyme.
- 2) In general experiments, the annealing temperature is 5 °C lower than the melting temperature (T_m) of the amplified primer. The annealing temperature can be appropriately reduced when the ideal amplification efficiency cannot be obtained.
- 3) The extension time should be set according to the size of the amplified fragment.
- 4) The number of cycles can be set according to the downstream application of the amplification product. If the number of cycles is too small, the expansion is insufficient. If the number of cycles is too many, the probability of mismatch will increase, and the non-specific background will be severe. Therefore, the number of cycles should be minimized under the premise of ensuring the yield of the product.

This product is for scientific research only, which shall not be used for clinical diagnosis or other purposes.