



FastStar DNA Polymerase

Product Number: PC16

Shipping and Storage

-20°C.

Components

Component	PC16	PC16	PC16
	500U	5KU	50KU
FastStar DNA Polymerase (5 U/μL)	100μL	1ml	10ml

Description

FastStar DNA Polymerase is a mixture of anti Taq enzyme monoclonal antibodies and Taq DNA Polymerase suitable for Hot Start PCR. When using Taq enzyme antibodies for PCR amplification, the binding of Taq enzyme antibodies to Taq enzyme inhibits DNA polymerase activity before high-temperature denaturation, which can effectively inhibit non-specific annealing of primers and non-specific amplification caused by primer dimers under low temperature conditions. Taq enzyme antibodies undergo denaturation in the initial DNA denaturation step of PCR reaction, and polymerase activity is restored, achieving a hot start effect. This product does not require special inactivation of Taq enzyme antibodies and can be used under conventional PCR reaction conditions.

FastStar DNA Polymerase has 5'→3' DNA polymerase activity and 5'-3' exonase activity, without 3'-5' exonase activity. The enzyme has an elongation rate of 2kb/min and can amplify fragments up to 5kb in length. The amplified PCR product has an "A" base attached to its 3'end, making it suitable for direct T/A cloning. The blocking rate of polymerase activity reaches over 95% at temperatures of 55°C and below. Heating at 95°C for 5 seconds can restore DNA polymerase activity. This product has the characteristics of fast extension speed and high amplification efficiency, and is mainly suitable for experiments such as PCR amplification of DNA fragments and DNA sequence determination.

Unit definition

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

Quality Control

After multiple column purification, the purity was detected by SDS-PAGE to be over 99%; no exogenous nuclease activity was detected. Closed at 55°C for 30 minutes without polymerase activity; 95°C for 5 seconds, fully activated.

Protocol

The following example is a PCR reaction system and reaction conditions for amplifying a 1 kb fragment using human genome DNA as a template. In practical operation, corresponding improvements and optimizations should be made based on the template, primer structure, and target fragment size.

1. The addition amount of enzymes in the 50μL PCR reaction system is 0.25-0.5μL as well as 1.25-2.5 U/50μL
2. PCR reaction conditions

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

} 25-35cycles



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Note:1)The hot start enzyme used in this product needs to be incubated at 95°C for at least 5 seconds to activate the enzyme.

2)In general experiments, the annealing temperature is 5°C lower than the melting temperature T_m of the amplification primer, and when ideal amplification efficiency cannot be achieved, the annealing temperature should be appropriately reduced.

3)The extension time should be set based on the size of the amplified fragment.

4)The number of cycles can be set based on the downstream application of the amplification product. If the number of cycles is too small, the amplification amount is insufficient; If there are too many cycles, the probability of mismatch will increase, and the non-specific background will be severe. So, while ensuring product yield, the number of cycles should be minimized as much as possible.