Tinzyme Co., Limited

Email: sales@tinzyme.com Website: www.tinzyme.com
Tel: +86-755-86134126 WhatsApp/Facebook/Twitter: +86-189-22896756

FastStar DNA Polymerase (Glycerol-free)

Product Number: PC16F

Shipping and Storage

-20±5°C.

Components

Component	PC16F 500U	PC16F 50KU
FastStar DNA Polymerase (Glycerol-free)(5U/μL)	100μL	10×1mL

Description

FastStar DNA Polymerase (Glycerol free) is a mixture of glycerol free 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 (Glycerol free) has 5'→3' DNA polymerase activity and 5'→3' exonuclease activity, without 3'→5' exonuclease activity. The enzyme extends at a 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℃ and below. Heating at 95℃ 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 freeze-drying experiments such as single enzyme freeze-drying, multiple amplification, and DNA sequencing.

Notes

1. After taking out this product from -20℃, please melt it at 4℃ or room temperature. Do not cover it with your hands;
2. After taking out this product, centrifuge it instantaneously before use. This product can be operated at room temperature. If the experimental environment temperature is higher than 25℃ or the experimental time is too long, please place the enzyme on ice;
3. Avoid repeated freeze-thaw of this product, as repeated freeze-thaw (freeze-thaw times ≤ 10 times) may cause a decrease in product performance. This product can be stored for a long time at -20±5℃;
4. This product is recommended for small portion packaging.

Quality Control

1. Protein purity: The purity detected by HPLC is close to 99%;
2. Detection of Exonuclease Residues:10U of proenzyme and 0.6μg λ-Hind III was incubated at 37℃ for 16 hours, and the electrophoresis bands of DNA did not change.
3. Endonuclease residue detection:10U of proenzyme and 0.6μg Supercooled pBR322 DNA was incubated at 37℃ for 4 hours, and the electrophoresis bands of the DNA did not change.
4. RNase residue detection:10U of proenzyme and 1μg HeLa cell total RNA at 37℃ for 1 hour, and the electrophoresis band of RNA remains unchanged.

Protocol

The following are examples of PCR reaction conditions and reaction systems for amplifying 1kb fragments using human

genome DNA as a template. In practical operations, corresponding improvements and optimizations should be made based on the template, primer structure, and target fragment size.

1. PCR reaction system

Reagent	50μL reaction system	Final Concentration
PCR Buffer	x μL	1×
dNTP Mix,10mM each	1 μL	200 μM each
Forward Primer, 10 μM	1 μL	0.2 μM
Reverse Primer, 10 μM	1 μL	0.2 μM
Template DNA	< 0. 5 μL	< 0.5 μg/50 μL
FastStar DNA Polymerase (Glycerol-free)(5U/μL)	0.25-0.5 μL	1.25-2.5U/50 μL
ddH_2O	Up to 50 μL	

Note:The reaction system can be configured at room temperature.

2. PCR reaction condition

Step	Temperature	Time	cycles
Predenaturation	95℃	5-60s	1
Denaturation	95℃	5-15s	35-45
Annealing/Extend	55-65℃	30s	

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

2)In general experiments, the annealing temperature is 5℃ lower than the melting temperature Tm 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.