

mTaq DNA Polymerase

Product Number: PC04

Shipping and Storage

-20°C。

Components

Component	PC04M 2500U
mTaq DNA Polymerase, 5 U/μl	5×100μl
mTaq PCR Buffer, 10×	5×1.8ml

Note: mTaq PCR Buffer contains 30mM MgCl₂.

Description

mTaq DNA Polymerase is a new type of DNA Polymerase which is modified by deletion of an amino acid segment in the N terminal of Taq DNA polymerase and mutation. Modified to tolerate inhibitors present in whole blood, the product can directly amplify DNA in whole blood samples of humans and mice without prior genome extraction and purification. This product can be used directly for T/A cloning because the 3' end of the amplified PCR product has an "A" base.

Activity Definition

After column purification, the purity was more than 99% according to SDS-PAGE. No exogenous nuclease activity was detected. No host residual DNA was detected by PCR. Can effectively amplify single copy genes in human genome; Stored at room temperature for one week, no obvious change in activity.

Protocol

- Reverse the mTaq DNA Polymerase repeatedly until it is thoroughly mixed before use.
- Put the PCR thin-walled tube on ice and add the following reagents except the whole blood. 1. PCR Reaction System

Reagent	50μL reaction system	Final Concentration
mTaq DNA Polymerase	1μl	
mTaq PCR Buffer, 10×	5μl	1×
dNTP Mix, 2.5mM each	4μl	200μM each
Forward Primer, (10μM)	2μl	0.4μM
Reverse Primer, (10μM)	2μl	0.4μM
*Whole Blood	≤10%	
RNase-Free water	xμl	
Total	50μl	

Note: 1) * Before adding the whole blood, suck up and down repeatedly and mix all kinds of reagents thoroughly.

2) DNA template: Whole blood can be treated with heparin sodium, Na-EDTA, K-EDTA, or sodium citrate. A whole blood level of 5-10% is usually recommended. High concentrations of blood are not recommended. For templates with high GC content, 10%DMSO was added.

3) Primers: The length of oligonucleotide primers usually contains 20-30nucleotides, and the optimal GC content is 40-60% and evenly distributed in the primers. In conventional PCR reactions, primer concentrations should be 0.1-1.0μM as a reference for the set range.

- Finally, add the whole blood to the bottom of the tube.
- PCR reaction conditions



Tinzyme Co., Limited

Email: sales@tinzyme.com

Website: www.tinzyme.com

Tel: +86-755-86134126

WhatsApp/Facebook/Twitter: +86-189-22896756

Step	Temperature	Time	
Pre-denaturation	95°C	5min	
Denaturation	95°C	30s	} 35-40cycles
Annealing	50-68°C	30s	
Extend	72°C	250-500bp/min	
Final extension	72°C	10min	

Note: 1) Preheat the PCR instrument to 94-95°C, place the sample on the PCR instrument and start the cycle.

- 2) mTaq improves cold sensitivity and has some hot start characteristics. Reaction ingredients can usually be prepared on ice, and finally Nonspecific products were avoided by adding polymerase and preheating the thermocycler to denaturation temperature (95°C).
 - 3) Denaturation temperature and time: In order to fully lysate blood cells and release/denature DNA prior to PCR cycle, the initial denaturation is required to be 95°C for 5 minutes.
 - 4) Annealing temperature and time: the annealing time is usually 30 seconds -1 minute. The annealing temperature can be 5°C lower than the theoretical annealing temperature (Tm) and optimized by gradient PCR.
 - 5) Elongation time: the elongation reaction is usually carried out at 72°C. Generally, the stretch time is 1 minute every 250-500 bp. The final extension is recommended at 72°C for 10 minutes.
 - 6) Usually 35-40 cycles can achieve optimal amplification.
5. Result detection: After the reaction, 5µL reaction product was taken and electrophoresis buffer was added to detect the result.