

Product Description

AcuGenix™ Hot Start Taq DNA Polymerase-Low DNA (Glycerol-Free) is a specialized enzyme optimized and formulated to meet the specific requirements of lyophilized reagents. This enzyme uses chemical modification to completely block Taq enzyme activity at room temperature, effectively suppressing non-specific amplification caused by non-specific primer annealing or primer-dimers under low-temperature conditions, thereby improving the specificity and sensitivity of PCR. It can also operate in a "Time release" manner, gradually releasing enzyme activity during the temperature ramp of PCR cycling, which further increases the specificity and sensitivity for amplifying low-copy-number templates. The main advantages of this enzyme in fluorescent PCR are high sensitivity, high fluorescence signal, and high specificity. This product is processed with a special technology to ensure no DNA residue, making it suitable for the development of detection reagents for biopharmaceuticals.

Size

5 U/μL

Components

1. 5 U/μL AcuGenix™ Hot Start Taq DNA Polymerase-Low DNA (Glycerol-Free)
2. 10×PCR Buffer II (Mg²⁺ free) (optional)
3. 25 mM MgCl₂ (optional)

*10×PCR Buffer II (Mg²⁺ free) does not contain dNTPs or Mg²⁺. Please add dNTPs and MgCl₂ when preparing reaction systems.

Unit Definition

One unit (U) of activity is defined as the amount of enzyme required to incorporate 10 nmol of deoxynucleotides into acid-insoluble material using activated chum salmon sperm DNA as template/primer within 30 minutes at 74 °C.

Storage Buffer

20 mM Tris-HCl (pH 8.0), 100 mM KCl, 1 mM EDTA, 1 mM DTT, Stabilizer.

Storage

Store at -20±5°C. Vortex before use. Avoid freeze-thaw cycles.

Quality Control

1. SDS-PAGE electrophoresis purity no less than 98%.
2. Amplification sensitivity, between-batch difference, and stability.
3. No exogenous nuclease activity, no exogenous endo/exonuclease contamination.

Prepare Reaction Mix

Components	Volume per Reaction	Final Concentration
10×PCR Buffer II (Mg ²⁺ free) ¹	5 μL	1×
dNTPs (10 mM each)	1 μL	200 μM
25 mM MgCl ₂	2-8 μL	1-4 mM
5 U/μL AcuGenix™ Hot Start Taq DNA Polymerase-Low DNA (Glycerol-Free)	0.25-0.5 μL	1.25-2.5U/Reaction
25×Primer Mix ²	2 μL	1×
Template	--	<1 μg/Reaction
ddH ₂ O	To 50 μL	--

1. This Buffer does not contain dNTP or Mg²⁺. dNTPs and MgCl₂ must be added before use.
2. If used for qPCR/qRT-PCR, fluorescent probes need to be added to the reaction system. The final concentration of primers is usually 0.2 μM for better results. when the reaction performance is poor, the primer concentration can be adjusted within the range of 0.2-1 μM. The probe concentration is usually optimized in the range of 0.1-0.3 μM. Concentration gradient experiments can be performed to find the best combination of primers and probes.

Reaction Programe

Two-step			
Procedure	Temperature	Time	Cycle
Hot Start	95°C	5-10 min	1
Denaturation	95°C	10-20 s	40-50
Annealing and Extension	56-64°C	20-60 s	

Three-step			
Procedure	Temperature	Time	Cycle
Hot Start	95°C	5-10 min	1
Denaturation	95°C	10-20 s	40-50
Annealing	56-64°C	10-30 s	
Extension	72°C	10-60 s	

Notes

1. Due to differences in ramp rates among thermal cycles and the intrinsic properties of this enzyme, if a 5-minute hot-start step results in poor amplification, it is recommended to optimize the hot-start time within the range of 5 to 15 minutes.
2. With high specificity and sensitivity, it is suitable for multiplex PCR reactions.
3. The DNA polymerase has 5'-3' polymerase activity, 5'-3' exonuclease activity, lacks 3'-5' exonuclease activity or proofreading function.
4. Suitable for conventional PCR, RT-PCR, and detection methods such as probe-based assays, fluorescent dye-based assays, and gene chip assays.
5. The PCR product has an A at the 3' end and can be directly cloned into T vectors.
6. For primers with low annealing temperatures or amplicon fragments longer than 200 bp, it is recommended to use a three-step method for amplification.