Robustart Taq
DNA Polymerase-Low DNA

Cat:E16-NA

V01

Product Description

Robustart Taq DNA Polymerase-Low DNA is a hot-start Taq enzyme product developed by BIORI. This product effectively suppresses nonspecific reactions caused by nonspecific primer annealing or primer-dimer formation during PCR setup and amplification, providing excellent specificity and more efficient amplification of low-concentration templates. It is suitable for multiplex PCR amplification and shows strong system compatibility, delivering stable amplification performance across different types of PCR reactions. The product is processed using a special manufacturing procedure and contains no residual DNA, making it suitable for the development of detection reagents related to biopharmaceutical applications.

Specification

5 U/μL

Components

1. 5 U/μL Robustart Taq DNA Polymerase-Low DNA
2. 10× PCR Buffer II (Mg^{2+} free) (optional)
3. 25 mM $MgCl_2$ (optional)

*10× PCR Buffer II (Mg^{2+} free) does not contain dNTPs or Mg^{2+} Add dNTPs and $MgCl_2$ when preparing the reaction mixture.

Activity Definition

One unit (U) is defined as the amount of enzyme required to incorporate 10 nmol of deoxynucleotides into acid-insoluble material using activated salmon sperm DNA as template/primer at 74℃ for 30 min.

Storage

Store at -20±5℃. Mix thoroughly before use and avoid repeated freeze-thaw cycles.

Quality Control

1. SDS-PAGE purity is greater than 98%.
2. Amplification sensitivity, lot-to-lot consistency, and stability are tested.
3. No exogenous nuclease activity and no contamination with exogenous endonucleases or exonucleases.

PCR Reaction Setup

Component	Volume per Reaction	Final Concentration
10× PCR Buffer II (Mg^{2+} free) [1]	5 μL	1×
dNTPs (10 mM each dNTP)	1 μL	200 μM
25 mM $MgCl_2$	2-8 μL	1-4 mM
5 U/μL Robustart Taq DNA Polymerase-Low DNA	0.25-0.5 μL	1.25-2.5 U
25× Primer Mix [2]	2 μL	1×
Template	—	<1 μg/reaction
ddH_2O	To 50 μL	—

1. This buffer does not contain dNTPs or Mg^{2+}; dNTPs and $MgCl_2$ must be added separately.

2. For qPCR or RT-qPCR applications, add a fluorescent probe to the reaction mixture. A final primer concentration of 0.2 μM usually gives good results. If reaction performance is suboptimal, adjust the primer concentration within 0.2-1 μM. Probe concentration can usually be optimized within 0.1-0.3 μM. Gradient experiments can be performed to determine the optimal primer and probe combination.

Reaction Conditions

Two-step Program			
Step	Temperature	Time	Cycles
Hot Start	95℃	1-5 min	1
Denaturation	95℃	10-20 s	35-50
Annealing/Extension	56-64℃	20-60 s	35-50
Annealing/Extension	56-64℃	20-60 s	35-50

Three-step Program			
Step	Temperature	Time	Cycles
Hot Start	95℃	1-5 min	1
Denaturation	95℃	10-20 s	35-50
Annealing	56-64℃	10-30 s	35-50
Extension	72℃	10-60 s	35-50

Notes

1. The enzyme supports rapid hot-start activation at 95℃ or 94℃ for 1-5 min.

2. It has strong system compatibility and provides higher specificity and sensitivity.

3. It is suitable as a high-sensitivity PCR detection reagent and can be used for multiplex PCR amplification.

4. It has 5′-3′ polymerase activity and 5′-3′ exonuclease activity; it has no 3′-5′ exonuclease activity and no proofreading function.

5. It is suitable for qualitative and quantitative detection by conventional PCR and RT-PCR.

6. PCR products carry an A overhang at the 3′ end and can be directly used for T-vector cloning.

7. A three-step program is recommended for primers with low annealing temperatures or for amplification of fragments longer than 200 bp.