

Hieff UNICON™ Robust Hotstart Taq DNA Polymerase

(20 U/μL)

Product description

This product is a hot-start DNA polymerase that utilizes two proprietary antibodies for dual blocking. It effectively inhibits both the 5'→3' polymerase activity and the 5'→3' exonuclease activity of Taq DNA polymerase. Upon heating at the pre-denaturation temperature for 30 seconds, the blocking antibodies are completely inactivated, releasing both the DNA polymerase and exonuclease activities.

This dual-blocking feature offers a dual advantage: it not only prevents non-specific amplification caused by mismatches or primer-dimers but also inhibits the degradation of probes that can lead to a decrease in fluorescent signal. This dual protection ensures greater stability of in vitro diagnostic reagents during transportation or when used at room temperature.

Specifications

Cat.No.	14325ES76 / 14325ES80 / 14325ES92 / 14325ES93
Size	1,000 U / 10,000 U / 25,000 U / 100,000 U

Components

Components No.	Name	14325ES76 (1,000 U)	14325ES80 (10,000 U)	14325ES92 (25,000 U)	14325ES93 (100,000 U)
14325	Robust Hotstart Taq (20 U/μL)	50 μL	500 μL	1.25 mL	5 mL

Storage

This product should be stored at -25~-15°C for 2 years.

Instructions

1. Reaction System (Examples of 25 μL and 50 μL)

Cat. NO.	Component Name	Volume (μL)	Volume (μL)
/	5× PCR Buffer	5	10
/	MgCl ₂ (250 mM)	0.35	0.7
/	dNTP Mix (25 mM each)	0.25	0.5
14325	Robust Hotstart Taq (20 U/μL)	0.15	0.3
/	Primer (10 μM)	0.4	0.8
/	Probe Mix (10 μM)	0.2	0.4
/	Template DNA	1-20	1-30
/	RNase Free H ₂ O	to 25 μL	to 50 μL

[Note]: Mix thoroughly before use and avoid vigorous shaking to prevent the formation of excessive bubbles.

1) dNTP concentration: It is recommended that the final dNTP concentration be 0.2–0.5 mM. If necessary, the optimal concentration can be determined by titration using the 25 mM each dNTP Mix in increments of 0.05–0.1 mM.

2) MgCl₂ concentration: It is recommended that the final Mg²⁺ concentration be 1.5–5 mM. If necessary, the optimal concentration can be determined by titration using the 250 mM MgCl₂ in increments of 0.2–0.5 mM.

3) Primer concentration: The Primer Mix contains multiple primer pairs. Typically, the final concentration of each primer can be adjusted between 0.1–1.0 μM according to the specific requirements.

4) Probe concentration: The Probe Mix contains multiple probes with different fluorescent signals. The concentration of each probe can be adjusted between 50–300 nM according to the specific requirements.

5) Template dilution: qPCR is highly sensitive. It is recommended to use diluted templates to ensure Ct values fall within the optimal range of 20–35.

6) Reaction volume: A reaction volume of 25 μL or 50 μL is recommended to ensure the efficiency and reproducibility of target gene amplification.

7) System preparation: Prepare the mixture in a biosafety cabinet using nuclease-free pipette tips and reaction tubes. Filter tips are recommended. Avoid cross-contamination and aerosol contamination.

2. Reference Amplification Protocol

2.1 Standard Protocol

Cycle Step	Temperature	Time	Cycles
Pre-denaturation	95°C	5 min	1
Denaturation	95°C	15 sec	45
Annealing/Extension	60°C ^a	30 sec ^b	

[Note]

a. Amplification Reaction: The amplification temperature should be adjusted based on the T_m value of the designed primers.

b. Fluorescence Signal Acquisition: The required time for fluorescence signal acquisition varies among different qPCR instruments; please set the parameters according to the minimum time limit specified for your instrument.

2.2 Fast Protocol

Cycle Step	Temperature	Time	Cycles
Pre-denaturation	95°C	30 sec	1
Denaturation	95°C	5 sec	45
Annealing/Extension	60°C	20 sec ^c	

[Note]: c. Fast Protocol: Please confirm via a preliminary experiment whether the qPCR instrument actually in use supports fast amplification.

3. Compatible Instruments

ABI 5700, 7000, 7300, 7700, 7900HT Fast, StepOne™, StepOne Plus™, ABI 7500, 7500 Fast, ViiA™7, QuantStudio™ 3 and 5, QuantStudio™ 6,7,12k Flex;

Stratagene MX3000P™, MX3005P™, MX4000P™;

Bio-Rad CFX96™, CFX384™, iCycler iQ™, iQ™5, MyiQ™, MiniOpticon™, Opticon®, Opticon® 2, Chromo4™;

Eppendorf Mastercycler® ep realplex, realplex 2 s;

Qiagen Corbett Rotor-Gene® Q, Rotor-Gene® 3000, Rotor-Gene® 6000;

Roche Applied Science LightCycler® 480, LightCycler® 2.0; Lightcycler® 96;

Thermo Scientific PikoReal Cyclers; Cepheid SmartCycler®; Illumina Eco qPCR; SLAN 96S, 96P.

Notes

1. Please use RNase-free consumables during the experiment.
2. Please operate with lab coats and disposable gloves, for your safety.
3. This product is for research use only.