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RealHi DNA Polymerase

Accurate Amplification

Store at -20°C

Cat. No. RT003

500 units, with dNTP

RealHi DNA Polymerase (5 U/µl): 100 µl

10X Reaction Buffer (with 20mM Mg^{2+}): 2 ml

10 mM dNTPs Mix: 200 µl

Cat. No. RT033

500 units, without dNTP

RealHi DNA Polymerase (5 U/µl): 100 µl

10X Reaction Buffer (with 20mM Mg^{2+}): 2 ml

Recombinant	✓
5′ to 3′ Exonuclease	✓
3′ to 5′ Exonuclease	✓
Endonuclease Free	✓

Description

RealHi DNA Polymerase is ideal for amplification of DNA fragments, especially for high-fidelity applications. High fidelity is provided by a mixture of RealTaq DNA Polymerase and a proofreading (3′–5′ exonuclease activity) enzyme, RealPfu DNA Polymerase. This formulation achieves greater yields with higher fidelity than standard DNA polymerase.

Quality Control

Nuclease activity is not detected after incubation of 1 µg Lambda/Hind III DNA with 5 units RealHi DNA Polymerase in 50 µl reaction volume in supplied reaction buffer for 18 hours at 37°C.

Storage Buffer

50 mM Tris-HCl (pH 9.0), 100 mM NaCl, 0.1 mM EDTA, 1% Triton X-100, 5 mM DTT, 50% Glycerol, Stabilizers

10X Reaction Buffer

100 mM KCl, 20 mM $MgSO_4 \cdot 7H_2O$, 200 mM Tris-HCl (pH 8.8), 1% Triton X-100, 100 mM $(NH_4)_2SO_4$, 1 mg/ml BSA.

This reaction buffer is supplied as a 10X concentrate and should be diluted for use.

Unit Definition

One unit of enzyme catalyzes the incorporation of 10 nmol of dNTP into acid-insoluble form in 30 minutes at 70°C.

General Reaction Conditions

The optimal conditions for the concentration of RealHi DNA Polymerase, primers and template DNA will depend on the system being utilized. It may be necessary to determine the optimal conditions for each individual component.

1. Add the following components to a sterile microtube on ice :

Components	Volume	Final Concentration
10X Reaction Buffer	5 µl	1X
10 mM dNTP Mix	0.5 µl	0.1 µm
Primer Mix (10 µM each)	1 µl	0.2 µm
Template DNA	0.5~10 µl	n/a
RealHi DNA Polymerase(5U/µl)	0.25 µl	1.25 units
D.W.	to 50 µl	n/a

2. Suggested Reaction Parameters for RealHi DNA Polymerase :

Segment	Number of Cycles	Temperature	Duration
1	1	94°C	1~3 minutes
2	25~35	94°C (Denature)	30 seconds~1 minute
		Primer Tm-5°C (Anneal)	30 seconds~1 minute
		72°C (Extend)	1 minute/Kbp
3	1	72°C	7 minutes
		4°C	

3. Analyze the amplification products by agarose gel electrophoresis and visualize by ethidium bromide staining.

Note: For research use only. Not for use in diagnostic or therapeutic procedures.

RBC
It's the Joy of Discovery

Real Biotech Corporation