[Cat. No.] Please refer to the Ordering Information

Introduction

HotStart Tag DNA Polymerase is designed to perform Hotstart PCR. The HotStart Taq DNA Polymerase is inhibited at lower temperature, but is fully activated at temperature above 70°C. This prevents the formation of mis-primed products and primer-dimers during the reaction setup process, resulting in improved PCR specificity.

Applications

- · Real-time quantification of DNA and cDNA targets using dual probe and dsDNA binding dve
- HotStart PCR
- Multiplex PCR
- Automated PCR

Components

Components	E-2017	E-2017-2	E-2017-1
HotStart <i>Taq</i> DNA Polymerase	250 U (50 µl)	500 U (50 µl x 2)	1,000 U (50 µl x 4)
10X Reaction buffer	0.5 ml	0.5 ml x 2	0.5 ml x 4
10 mM dNTPs	0.5 ml	0.5 ml x 2	0.5 ml x 4
Dilution buffer	0.5 ml	0.5 ml x 2	0.5 ml x 4
Components	E-2017-3	6 E	E-2017-4
Components HotStart <i>Taq</i> DNA Polymerase	E-2017-3 250 U (50 µl)	6 E	E-2017-4 1,000 U (50 μl x 4)
Components HotStart <i>Taq</i> DNA Polymerase 10X Reaction buffer	E-2017-3 250 U (50 μl) 0.5 ml	6 E	-2017-4 1,000 U (50 μl x 4) .5 ml x 4
Components HotStart Taq DNA Polymerase 10X Reaction buffer 10 mM dNTPs	E-2017-3 250 U (50 μl) 0.5 ml	в Е (О	5 ml x 4
Components HotStart Taq DNA Polymerase 10X Reaction buffer 10 mM dNTPs Dilution buffer	E-2017-3 250 U (50 μl) 0.5 ml - 0.5 ml	<mark>і Е</mark> (О	E-2017-4 1,000 U (50 μl x 4) .5 ml x 4 - .5 ml x 4

Specifications

HotStart Taq DNA Polymerase				
5' to 3' exonuclease activity	Yes			
3' to 5' exonuclease activity	No			
3'-A overhang	Yes			
Fragment size Up to 10 kb				

Buffer Composition

10X Reaction buffer	100 mM Tris-HCl, 450 mM KCl, 15 mM MgCl₂, pH 9.0
Dilution buffer	20 mM Tris-HCl, 0.5 mM EDTA, 1 mM DTT, 100 mM KCl, stabilizer, 50% glycerol, pH 8.0

Storage Buffer

HotStart Tag DNA Polymerase is supplied in 50% (v/v) glycerol containing 20 mM Tris-HCl, 0.5 mM EDTA, 1 mM DTT, 100 mM KCI, and stabilizer, pH 8.0.

Unit Definition

One unit is defined as the amount of enzyme that incorporates 10 nmole of dNTP into acid-insoluble products in 30 min at 72°C.

Quality Control

 Nuclease Contamination Assay: Nuclease activity is not detected after incubation of 1 µg of substrate DNA (supercoiled plasmid and Lambda/Hind III DNA) with 5 U of HotStart Tag DNA Polymerase in 50 µl reaction volume at 37°C and 70°C for 18 hrs.

Storage

Store at -20°C. If stored in the recommended temperature, this product will be stable until the expiration date printed out on the label.

Online Resources



Korean



English

Visit our product page for additional information and protocols.

Ordering Information

	Description		Cat. No
250 U	10X Reaction buffer with 15 mM MgCl ₂	10 mM dNTP	E-2017
			E-2017-3
500 U		10 mM dNTP	E-2017-2
1,000 U		10 mM dNTP	E-2017-1
			E-2017-4

Notice

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Explanation of Symbols





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Experimental Procedures

Steps Procedure Details						
1	Thaw reagents	 Thaw 10X Reaction buffer and 10 mM dNTPs on ice and mix thoroughly before use. Then, briefly spin down all components including template DNA, HotStart <i>Taq</i> DNA Polymerase, primers and nuclease-free water. 				
		2. Add all components into PCR tubes (not provided) to a total volume of 20 µl or 50 µl.				
		Preparation of reaction mixture				
		Componen	ts	20 µl reaction	50 µl reaction	
		Template DNA*		Variable	Variable	
		Forward primer (10 pmol	/µI)	1-2 µl	2.5-5 µl	
		Reverse primer (10 pmol	//μl)	1-2 µl	2.5-5 µl	
2	0	10X Reaction buffer		2 µl	5 µl	
		10 mM dNTPs		2 µl or Variable	5 µl or Variable	
	December of	HotStart Taq DNA Polym	erase (5 U/µl)	0.5-1 U	1-2.5 U	
	Preparation of	Nuclease-free water		Variable	Variable	
		Total volume		20 µl	50 µl	
		DNA, > 10 pg; human genomic DNA, > 1 ng. 3. Mix the reaction mixture by tapping or pipetting, and briefly spin down.				
		Step	Temperature	Time	Cycles	
		Pre-denaturation	94°C	15 min	1 cycle	
		Denaturation	94°C	0.5-1 min	·	
3		Annealing	45-65°C [†]	0.5-1 min	25-35 cycles	
		Extension	72°C	1 min/kb		
	Incubate reactions in a thermal cycler	Final extension	72°C	5-10 min	1 cycle	
		Note: For maximum yield and specificity, temperatures and cycling times should be optimized for each new template DNA or primers. Set the annealing temperature to 3-5°C lower than the Tm of the primers.				
4	Analyze with gel electrophoresis	 5. After the reaction, maintain the reaction mixture at 4-8°C. The samples can be stored at -20°C until use. 6. Load samples on agarose gel with adding a loading-dye mixture, and perform gel electrophoresis for analysis. 				

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