EasyTaq® DNA Polymerase for PAGE

Cat. No. AP112
Concentration 5 units/μl
Storage: at -20°C for two years

Description

EasyTaq® DNA Polymerase for PAGE is purified from *E. coli* expressing a cloned DNA polymerase from Thermus aquaticus. The enzyme consists of a single polypeptide with a molecular weight of approximately 94 kDa. *EasyTaq*® DNA Polymerase for PAGE has 5′-3′ DNA polymerase activity and 5′-3′ exonuclease activity. lt lacks 3′-5′ exonuclease activity. This enzyme is supplied with unique buffer, and its PCR product is suitable for SDS-PAGE and agarose gel electrophoresis.

Highlights

- Extension rate is about 1-2 kb/min.
- Unique buffer system compatible with PAGE.
- Template-independent "A" can be generated at the 3′ end of the PCR product. PCR products can be directly cloned into *pEASY*®-T vectors.
- Amplification of genomic DNA fragment up to 3 kb.

Application

Short fragment PCR

Unit Definition

One unit of *EasyTaq*® DNA Polymerase for PAGE incorporates 10 nmol of deoxyribonucleotide into acid-precipitable material in 30 minutes at 74°C.

Quality Control

EasyTaq® DNA Polymerase for PAGE has passed the following quality control assays: functional absence of double- and single-strand endonuclease activity; >99% homogeneous measured by SDS-PAGE. Each batch of *EasyTaq*® DNA Polymerase for PAGE has been assayed for amplification efficiency to amplify p53 gene from 10 ng of human genomic DNA.

Storage Buffer

20 mM Tris-HCl (pH 8.0), 0.1 mM EDTA, 1 mM DTT, 100 mM KCl, 50% glycerol, stabilizers

10×*EasyTaq*® Buffer for PAGE (with Mg^{2+})

200 mM Tris-HCl (pH 8.3), 200 mM KCl, 100 mM $(NH_4)_2SO_4$, 20 mM $MgSO_4$, others

Kit Contents

Component	AP112-01/11	AP112-02/12
EasyTaq® DNA Polymerase for PAGE	2500 U	2500 U×4
10×*EasyTaq*® Buffer for PAGE	1.2 ml ×5	1.2 ml×20
2.5 mM dNTPs	- / 800 μl×5	- / 800 μl×20
6×DNA Loading Buffer	1 ml	1 ml ×4

Reaction Components

Component	Volume	Final Concentration
Template	Variable	as required
Forward Primer (10 μM)	1 μl	0.2 μM
Reverse Primer (10 μM)	1 μl	0.2 μM
10×*EasyTaq*® Buffer for PAGE	5 μl	1×
2.5 mM dNTPs	4 μl	0.2 mM
EasyTaq® DNA Polymerase for PAGE	0.5-1 μl	2.5-5 units
Nuclease-free Water	Variable	-
Total volume	50 μl	-

Thermal cycling conditions

94°C	2-5 min	
94°C	30 sec	30-35 cycles
50-60°C	30 sec	
72°C	1-2 kb/min	
72°C	5-10 min	

Notes

- A final concentration of 2 mM $MgSO_4$ is sufficient for most targets amplification. For some targets, more Mg^{2+} may be required.
- For optinal results, we recommend to use the 100 mM $MgSO_4$ stock to prepare a titration from 2 mM to 4 mM (final concentration) in 0.25 mM increments.
- 0.5 μl (2.5 units) enzyme is enough for per 50 μl reaction. For better amplification, up to 1 μl (5 units) enzyme can be used.

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