WizPure™ FX-Taq DNA Polymerase

(For Long & Fidelity PCR)

- W1341ES **250 U** -
- W1341E **500 U** -
- W1341E-5 **2,500 U** -
- W1341S **250 U** dNTP
- W1341 **500 U** dNTP
- W1341-5 **2,500 U** dNTP

Description

WizPure™ FX-Taq DNA Polymerase is a thermally stable, processive, 5'→ 3' DNA polymerase and 3'→5' proofreading function. The FX-Taq DNA polymerase optimized for PCR amplification of genomic DNA templates up to 20 kb and lambda DNA up to 30 kb. With its enhanced processivity, yield, speed and excellent 3'→5' exonuclease and 3'→5' proofreading activity, this enzyme is able to consistently deliver accurate and reliable amplification of long templates. This product is the ideal choice for long DNA templates unable to be amplified in conventional PCR, and is highly suitable for multiple downstream applications including complex cloning and genotyping experiments. The PCR product amplified with this mixture has one A added at 3'-end, so the product can be directly used for TA cloning.

Kit Contents

Contents	W1341S	W1341	W1341-5
FX-Taq DNA Polymerase (2.5U/µl)	250 U	500 U	2,500 U
10X Reaction Buffer	1 ml	2 ml	10 ml
dNTP mix (2.5 mM each)	0.5 ml	1 ml	5 ml
25mM MgCl2	0.5 ml	1 ml	5 ml

Applications

- Long range PCR
- High-fidelity PCR and primer-extension reactions
- Genotyping
- Library construction
- High GC amplification
- Next-generation DNA sequencing
- TA cloning

Storage Buffer

20mM Tris-HCl, 1mM dithiothreitol, 0.1mM EDTA, 100mM NaCl, Stabilizer, 50% glycerol, pH 8.0 (25°C).

10X Reaction Buffer

Contained 15mM MgCl2.

Unit Definition

1 unit is defined as the amount of enzyme that will incorporate 10 nmol of dNTP into acid-insoluble material in 30 minutes at 75°C.

Storage Conditions

Store all components at -20ºC in a non-frost-free freezer.

Quality Control

No endonuclease activity, nicking activity, exonuclease activity, or priming activity has been detected.

Quality Authorized by : Jamie Ahn 

Protocol

This standard protocol applies to a single reaction where only template, primers, and water need to be added to the FX-Taq DNA Polymerae. For multiple reactions, scale-up volume of reaction components proportionally. All reagents should be thawed on ice, gently mixed and briefly centrifuged before use.

1. The following table shows recommended component volumes:
   On ice, prepare each of following master mixes, combine, and place in heated (to 95°C) thermal cycler:

For 50µl PCR Reaction	Volume	Final Conc.
FX-Taq DNA Polymerase (2.5U/µl)	1 µl	1.0 ~ 2.5 U
10X Reaction Buffer [1)]	5 µl	1 X
dNTP mix (2.5 mM each)	4 µl	200 µM each
25mM MgCl2 (Optional) [2)]	1 ~ 3 ul	1.5 ~ 3.0 mM
Template	< 500 ng	< 500 ng
Forward Primer	5 ~ 50 pmol	0.1 ~ 1 µM
Reverse Primer	5 ~ 50 pmol	0.1 ~ 1 µM
Distilled water	up to 50 µl	

NOTE:
1) 10X Reaction Buffer contains 15mM MgCl2.
2) For more higher Mg2+ ion, please add 25mM MgCl2.
3) In general, use greater than 0.5 µM primers for sensitivity and less than 0.5 µM for specificity.
4) Recommended amount of template per PCR reaction:
   - < 50 ng plasmid or
   - < 500~1000ng genomic DNA or
   - 2µl of a 100µl single plaque eluate or
   - one single bacterial colony

2. Ensure reactions are mixed thoroughly by pipetting or gentle vortexing followed by a brief spin in a microcentrifuge.
3. Optional-Overlay reactions with one-half volume PCR-grade mineral oil when not using heated lid on thermal cycler.
4. Transfer tubes on ice into a thermal cycler pre-warmed. The following table shows recommended cycling conditions:

Step	Temp (°C)	Time	Cycle
Initial Denaturation	95	5 min.	1
Denature	95	10 ~ 30 sec.	25 ~ 40
Anneal	50~65	10 ~ 30 sec.	
Extend	72	1 min./kb	
Final Extension	72	5 min.	1

IMPORTANT: Annealing temperature should be 2-6ºC lower than the primer melting temperature. Elongation time should be ~1 min/1 kb.

NOTE: Cycling conditions may need to be optimized, depending on different primer and template combinations. For example, raise the annealing temperature to prevent non-specific primer binding, increase extension time to generate longer PCR products.

5. After cycling, maintain the reactions at 4°C or store at -20°C until ready for analysis.

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