

Taq DNA Polymerase Technical Manual No. 0213

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I INTRODUCTION

Taq DNA Polymerase is a thermostable DNA Polymerase isolated from an *E. coli* strain that carries the *Taq* DNA polymerase gene. *Taq* DNA polymerase is the most common polymerase used for PCR* reactions

II APPLICATIONS

Taq DNA Polymerase can be used in most applications including the following:

- PCR.
- > 3' A-tailing of blunt ends.
- > Primer extension.
- > DNA sequencing.

III KEY FEATURES

Key features of Taq DNA Polymerase:

- > Terminal transferase activity. *Taq* DNA Polymerase has terminal transferase activity which results in the addition of a single nucleotide (adenosine) at 3' end of the extension product.
- > High-purity. No contamination activity has been detected in standard test reactions.

IV SHIPPING AND STORAGE

This product is shipped on blue ice. Store the product at -20° C.

V GENERAL PCR PROTOCOL USING Taq DNA POLYMERASE

1. Thaw all the reagents for PCR on ice. Vortex to mix to remove concentration gradient and then spin down briefly.

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2. Set up 50 µl PCR reaction in a thin-wall PCR tube on ice by the following recipe:

5 μ I 10X Tag buffer solution containing Mg²⁺.

1 µl 10 mM dNTP stock

1 µl Forward primer (50 uM)

1 µl Reverse primer (50 uM)

2 µl Template (up to 100 ng/µl) sterile or filtered H₂O

39.5 µl sterile or filtered H₂O

0.5 µl Taq polymerase (5 units/µl)

3. Program PCR cycler as following and start:

Initial denaturing: 94°C for 3 minutes

Then 30 cycles of: 94°C for 30 seconds

55°C for 45 seconds

72°C for 60 seconds (about 1 kb/minute)

Extension: 72°C for 7 minutes

- 4. When the temperature of PCR cycler reaches 94°C, put PCR reaction tube in and continue the program.
- Analyze PCR fragments on a agarose or polyacrylamide gel. 5.

Note:

This is a basic protocol. One needs to optimize the reagent concentrations, conditions and parameters. 1.

This protocol is for PCR cycler with a hot lid. Otherwise, mineral oil needs to be added to prevent 2. evaporation.

3. 5% DMSO, 1M betaine, or both can be included in PCR reaction to improve the results when a GC-rich template is used.

VI ORDER INFORMATION

Tag DNA Polymerase, Cat. No. E00007-1000 Cat. No. E00007-50000 Green Taq DNA Polymerase, Cat. No. E00043

* The PCR process is covered by U. S. Patent numbers 4683195 and 4683202 issued to Cetus and owned by Hoffman-La Roche Inc. GenScript does not encourage or support the unauthorized use of the PCR process. Use of this product is recommended for persons that either have a license to perform PCR or are not required to obtain a license.

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> > 2



Taq DNA Polymerase without Mg²⁺

Cat. No.: E00008

Size: 1,000 U

Description:

Taq DNA Polymerase is a thermostable DNA Polymerase isolated from an *E. coli* strain that carries the *Taq* DNA polymerase gene. *Taq* DNA Polymerase is the most common polymerase used for PCR* reactions.

Key Feature:

Terminal transferase activity. Taq DNA Polymerase has terminal transferase activity which results in the addition of a single nucleotide (adenosine) at 3"end of the extension product.

High-purity. No contamination activity has been detected in standard test reactions.

Unit Definition:

One unit is the amount of enzyme that can incorporate 10 nmol of dNTP into acid-insoluble material in 30 minutes at $74^{\circ}C$.

10 X reaction Buffer (without Mg²⁺):

500 mM KCl, 100 mM Tris HCl (pH 9.0 at 25°C), 1% Triton X-100 Buffer. This buffer is optimized for use with 200 μ M dNTPs.

Example

PCR performance, activity, nuclease.



Lane	Таq	United Used		
1		0.1		
2	Leading Brand A	0.25		
3		0.5		
4	Leading Brood B	0.1		
5		0.25		
6		0.5		
7	Genscript	0.1		
8		0.25		
9	1	0.5		

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DATASHEET Version: 2013-08-30

Note: If the reaction is performed without this buffer, then add 0.1% Triton X-100 (final concentration) to ensure high activity.

Storage Buffer and Concentration:

The enzyme is delivered in 5 units/ μ l in 20 mM Tris HCl (pH 8.0), 0.1 mM EDTA, 1 mM DTT, 0.1% Triton X-100 and 50% glycerol.

Storage:

This product can be stored at -20°C for future using.

Formulation:

GenScript *Taq* DNA Polymerase has been formulated using GenScript's proprietary technology. The enzyme can be shipped at room temperature or even 37°C for seven days without any loss of activity.

Applications:

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Taq DNA Polymerase can be used in most applications including the following:

- PCR*
- 3" A-tailing of blunt ends
- Primer extension
- DNA sequencin



DATASHEET

Version: 2013-09-10

Taq DNA Polymerase, concentrated

Cat. No. E00012

Size: 2500 U

Description:

Tag DNA Polymerase is a thermostable DNA polymerase isolated from an E. coli strain that carries the Tag DNA Taq DNA Polymerase is the most polymerase gene. common polymerase used in PCR*. In some cases, such as RAPD PCR, adding large volume of general Taq DNA polymerase (5 U/µI), which has a high concentration of glycerol in its storage buffer, will increase the glycerol concentration in the reaction mix, interfering with PCR performance. The use of concentrated Tag DNA Polymerase (25 U/µI), with a far slimmer dose of glycerol, can prevent poor PCR efficiency. Note: Concentrated Tag DNA Polymerase (GenScript, E00012) is supplied with 10X reaction buffer containing 15 mM magnesium chloride. The dNTP (10 mM) mixture may be ordered separately (See related products).

Key Feature:

Terminal Transferase Activity: A single nucleotide
(adenosine) is added to the 3' end of the extension product.
High-Purity: No contamination activity has been detected in

standard test reactions.

Concentration:

Supplied in 25 units/µl in 20 mM Tris HCl (pH 8.0), 0.1 mM EDTA, 1 mM DTT, **0.1% Triton X-100** and 50% glycerol.

Unit Definition:

One unit is defined as the amount of enzyme that can incorporate 10 nmol of dNTP into acid-insoluble material in 30 minutes at 74°C.

10 X reaction Buffer (with Mg²⁺)

500 mM KCl, 100 mM Tris HCl (pH 9.0 at 25B0C), 15 mM MgCl2, 1% Triton X-100 Buffer. This buffer is optimized for use with 200 μ M dNTPs.

Important:

If another reaction buffers are used with *Taq* DNA Polymerase, Triton X-100 must be added to a final concentration of 0.1% to ensure high enzyme activity with *Taq* DNA Polymerase;

Storage:

Store the product at -20°C.

Formulation:

Genscript *Taq* DNA Polymerase has been formulated using GenScript's proprietary technology. The enzyme can be shipped at room temperature or stored at 37°C for seven days without any significant loss of activity.

Applications:

The applications of Taq DNA Polymerase are as follows:

- PCR*
- 3' A-tailing of blunt ends (T/A-cloning)
- Primer extension
- DNA labeling reactions

QC Tests

PCR performance, activity, nuclease.

		-	-		-		-			Lane	Taq	United Used
M	1	4	3	4	5	0		•	3	1		0.1
										2	Leading Brand A	0.25
								18. EK	3	Diana	0.5	
								1	4		0.1	
										5	Leading Brood B	0.25
								6		0.5		
				7		0.1						
										8	Genscript	0.25
										9		0.5

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