

Taq DNA Polymerase Technical Manual No. 0213

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II	Introduction	1
III	Key Features	1
IV	Ship and Storage	1
V	General PCR Protocol Using Taq DNA Polymerase	1
VI	Order Information	2

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.

1



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