Taq DNA Polymerase

POLYMERASE CHAIN REACTION (PCR)

Description

Taq DNA Polymerase is a single-subunit enzyme purified from the thermophilic bacterium *Thermus aquaticus*. It polymerizes DNA from a primer annealed to a DNA template in the presence of deoxyribonucleoside triphosphates. *Taq* DNA Polymerase has a temperature optimum around 75°C and can survive repeated incubations at 95°C. It also lacks intrinsic nuclease activity (1, 2). The recombinant form of the native enzyme is expressed in *E. coli* and provides excellent reproducibility between lots.

Applications

Amplification of template molecules for PCR

Taq DNA Polymerase, licensed for use in PCR, is extensively tested for contaminating nickase, single- and double-stranded exonuclease and endonuclease activities. When used with the supplied 10X PCR Buffer, the enzyme can successfully amplify single-copy genes from genomic DNA and can yield specific PCR products exceeding 2 kb in length (Fig 1).

DNA sequencing by the dideoxynucleotide method

Taq DNA Polymerase is ideal for sequencing templates that have a high degree of secondary structure. Due to its high temperature optimum, sequencing reactions can be performed at 70°C, where templates show minimal secondary structure and the stringency of primer hybridization is high (3).
Taq DNA Polymerase also lacks intrinsic nuclease activity and therefore gives uniform bands in autoradiograms.

Properties

Unit Definition: One unit catalyzes the incorporation of 10 nmol of total nucleotide into acid-insoluble product in 30 min at 70°C utilizing M13mp18 DNA as a template.

Molecular Weight: 86 to 90 kDa.

Activators: Requires divalent cations, with Mg^{2+} preferred over Mn^{2+}.

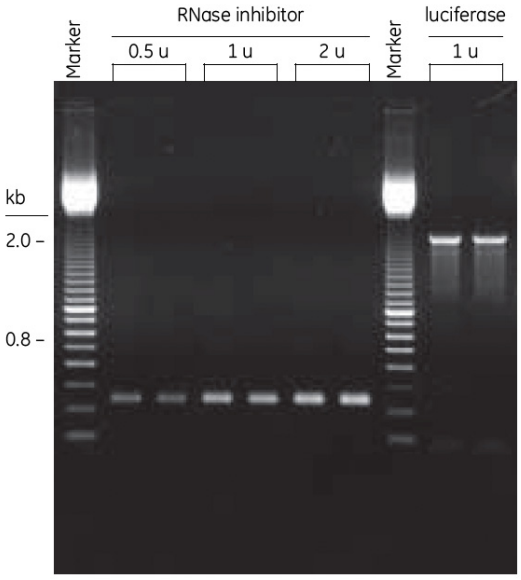

Fig 1. Amplification of a single-copy gene (RNase inhibitor; ~250 bp PCR product) from 5 ng human genomic DNA and a 2.2 kb PCR product (luciferase gene) from 10 pg plasmid. Amplifications were performed using the recommended conditions and the amounts of *Taq* DNA Polymerase shown. Marker = 100 Base-Pair Ladder (27-4001-01).

Components

***Taq* DNA Polymerase:** Enzyme is supplied at a concentration of 5 units/µL in 50 mM Tris-HCl pH 7.5, 5 mM DTT, 0.1 M EDTA, 50% glycerol and stabilizers.

10X PCR Buffer: 100 mM Tris-HCl (pH 9.0), 15 mM $MgCl_2$, 500 mM KCl.

25 mM $MgCl_2$ solution: included with 27-0798-04/5/6

Quality Control

PCR: Functionally tested for PCR by amplification of a PCR product from human genomic DNA using primers for the p53 gene.

Nickase: At least 90% of φX-174 DNA remains as Form I when 2 µg DNA is incubated with at least 10 units of *Taq* DNA Polymerase in a 30 µL reaction mixture for 1 h at 65°C.

DNase: Less than 1% of [3H]-DNA is hydrolyzed when 100 ng [3H]-DNA (HphI/AluI restricted M13 DNA) is incubated with 10 units of *Taq* DNA Polymerase in a 40 µL reaction mixture for 1 h at 65°C.

Restriction endonuclease: No contaminating restriction endonuclease is detected by agarose gel electrophoresis when 1 µg λ DNA is incubated with at least 10 units of *Taq* DNA Polymerase in a 50 µL reaction mixture for 18 h at 65°C under mineral oil.

Storage

Store at –20°C.

References

1. *Chien, A. et al., J. Bacteriol.* **127**, 1550 (1976).
2. Kaledin, A. S. *et al., Biokhimiya* (English translation) **45**, 494 (1980).
3. Innis, M. A. *et al., Proc. Natl. Acad. Sci. USA* **85**, 9436 (1988).

Ordering information

Product	Code Number
Taq DNA Polymerase (cloned) 250 units	27-0798-04*
Taq DNA Polymerase (cloned) 4 × 250 units	27-0798-05*
Taq DNA Polymerase (cloned) 10 × 250 units	27-0798-06*

*Supplied with 10 × PCR Buffer containing 100 mM Tris-HCl, pH 9.0, 15 mM $MgCl_2$ and 500 mM KCl.

Related products

PCR

Product	Code Number
PuReTaq™ Ready-To-Go™ PCR Beads	see catalog for full range
FideliTaq™ DNA Polymerase	see catalog for full range
FideliTaq PCR Master Mix (2X)	E71182
FideliTaq PCR Master Mix Plus	E71183
Amersham™ Hot Start Master Mix	25-1500-01
Amersham Hot Start Mix RTG	see catalog for full range

RT-PCR

Ready-To-Go RT-PCR Beads	see catalog for full range
RT-PCR Master Mix (2X)	E78370
FideliTaq RT-PCR Master Mix (2X)	E71185

First-strand synthesis

First-Strand cDNA Synthesis Kit	27-9261-01
Ready-To-Go You-Prime First-Strand Beads	27-9264-01
Ready-To-Go T-Primed First-Strand Kit	27-9263-01
TimeSaver™ cDNA Synthesis Kit	27-9262-01

Nucleotides

Amersham dNTPs	see catalog for full range

mRNA and total RNA Purification

mRNA Purification Kit	27-9258-01
mRNA Purification Kit	27-9258-02
Amersham RNAspin Isolation Kits	see catalog for full range
QuickPrep Micro mRNA Purification Kit	27-9255-01
QuickPrep mRNA Purification Kit	27-9254-01

DNA purification

ExoSAP-IT™	see catalog for full range
GFX™ PCR DNA and Gel Band Purification Kit	28-9034-70
MicroSpin™ S-400 HR Columns	27-5140-01
100 Base-Pair Ladder	27-4007-01

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