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Taq DNA Polymerase

Store at -20°C

Cat. No.	Description	Concentration	Quantity
G008	Taq DNA Polymerase	5 U/µl	5000 U
G009	Taq DNA Polymerase	5 U/µl	1000 U
G126	Taq DNA Polymerase	5 U/µl	10000 U

Product Description
Taq DNA Polymerase is a highly thermostable DNA Polymerase isolated from the thermophilic bacterium *Thermus aquaticus*. Taq DNA Polymerase catalyzes the 5' to 3' synthesis of DNA. The enzyme has no detectable 3' to 5' proofreading exonuclease activity, and possesses low 5' to 3'exonuclease activity. PCR products, amplified up to 6 kb in length with Taq DNA Polymerase, contain a single base (A) 3' overhang.

Product Components	1000 U	5000 U	10000 U
Taq DNA Polymerase (5 U/µl)	200 µl	1 ml	2 ml
10X PCR Buffer, with Mg^{2+}	2 ml	10 ml	20 ml
25 mM $MgSO_4$	1 ml	1 ml	1 ml

Storage Buffer Components
50 mM Tris-HCl (pH 8.0), 100 mM NaCl, 0.1 mM EDTA, 5 mM DTT, 50 % glycerol and 1.0 % Triton®X-100.

Unit Definition
One unit of the enzyme catalyzes the incorporation of 10 nmol of deoxyribonucleotides into a polynucleotide fraction in 30 mins at 74°C.

Shipping and Storage
Upon arrival, Taq DNA Polymerase should be stored at -20°C. Avoid repeated freeze-thaw cycles of all Taq components to retain maximum performance. All Taq components are stable for 1 year from the date of shipping if stored and handled properly.

Protocol
The following basic protocol serves as a general guideline and starting point for any PCR amplification. Optimal reaction conditions (incubation times, temperatures, concentration of Taq DNA Polymerase, primers, $MgSO_4$ and template DNA) may vary and need to be optimized for each specific PCR.

All PCR experiments should be assembled in a nuclease-free environment. In addition, DNA sample preparation, reaction set-up and subsequent reaction(s) should be performed in separate areas to avoid cross contamination.

A negative control reaction (omitting template DNA) should always be performed in tandem with sample PCR to confirm the absence of DNA contamination.

1. Add the following components to a sterile 0.2 ml PCR tube sitting on ice.

Components	Volume	Final Concentration
Template DNA	~100 ng	~2 ng/µl
Forward primer (10 µM)	1 - 2.5 µl	200 - 500 nM
Reverse primer (10 µM)	1 - 2.5 µl	200 - 500 nM
10X PCR buffer, with Mg^{2+}	5 µl	1X
25 mM $MgSO_4$ (optional)*	0 - 3 µl	1.5 - 3 mM
dNTP Mix (10 mM)	1 µl	200 µM
Taq DNA Polymerase (5 U/µl)	0.5 - 1 µl	2.5 - 5 U
Nuclease-free H_2O	up to 50 µl	-

* Optimal Mg^{2+} concentration is specific to each DNA template-primer set and can only be determined experimentally.

• *We recommend preparing a mastermix for multiple reactions to minimize reagent loss and enable accurate pipetting.*

2. Mix contents of tube and centrifuge briefly.
3. Incubate tube in a thermal cycler at 94°C for 3 mins to completely denature the template.
4. Perform 30 - 35 cycles of PCR amplification as follows:

   Denature: 94°C for 30 sec

   Anneal: 45 - 72°C for 30 sec

   Extend: 72°C for 1 min/1 kb template

5. Incubate for an additional 5 mins at 72°C and maintain the reaction at 4°C. The samples can be stored at -20°C until use.
6. Analyze the amplification products by agarose gel electrophoresis and visualize by ethidium bromide or SafeView™ (Cat No. G108) staining. Use appropriate molecular weight standards.