



Applied Biological Materials Inc

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Taq Plus DNA Polymerase

Store at -20°C

Cat. No.	Description	Concentration	Quantity
G012	Taq Plus DNA Polymerase	5U/ μ l	250U
G040	Taq Plus DNA Polymerase	5U/ μ l	1000U

Product Description

Taq Plus DNA Polymerase is a mixture of Taq DNA Polymerase and proofreading DNA Polymerase, which allows for the amplification of long templates, up to 20kb, with high fidelity. The two enzymes act synergistically during PCR to generate more accurate and longer PCR products with greater yields compared to Taq DNA Polymerase alone. PCR products, amplified up to 20kb in length with Taq Plus DNA Polymerase, contain a mixture of blunt ends and single base (A) 3' overhang. The error rate of this PCR amplification is 7.5×10^{-5} per nucleotide per cycle.

The products can be used for direct T/A cloning, but its efficiency is not as high as PCR products amplified with Taq polymerase alone.

Product Components	250U	1000U
Taq Plus DNA Polymerase (5U/ μ l)	50 μ l	200 μ l
10X PCR buffer, without Mg ²⁺	1ml	3ml
25mM MgSO ₄	1ml	1ml

Storage Buffer Components

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

10X PCR Buffer Components

200mM Tris-HCl (pH 8.3), 200mM KCl and 100mM (NH₄)₂SO₄ and PCR enhancers.

Unit Definition

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

Shipping and Storage

Upon arrival, Taq Plus DNA Polymerase should be stored at -20°C. Avoid repeated freeze-thaw cycles of all Taq Plus components to retain maximum performance. All Taq Plus 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 Plus DNA Polymerase, primers, MgSO₄ and template DNA) vary and need to be optimized.

PCR reactions should be assembled in a DNA-free environment. DNA sample preparation, reaction mixture assemblage and the PCR process, in addition to the subsequent reaction analysis, should be performed in separate areas.

A control reaction, omitting template DNA, should always be performed to confirm the absence of contamination.

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

Components	Volume	Final Concentration
Template DNA	~100ng	~2ng/μl
Forward primer (10μM)	1-2μl	0.2- 0.4μM
Reverse primer (10μM)	1-2μl	0.2- 0.4μM
10X PCR buffer, without Mg ²⁺	5μl	1X
25mM MgSO ₄	2-3μl	1-1.5mM
10mM dNTP	1μl	0.2mM
Taq Plus DNA Polymerase (5U/μl)	0.5- 1μl	2.5-5U
ddH ₂ O	up to 50μl	

- *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 30-40 cycles of PCR amplification as follows:
 - Denature:** 94°C for 30 sec
 - Anneal:** 55°C for 30 sec
 - Extend:** 72°C for 1min/1kb 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.