Taq DNA Polymerase with Robust Buffer (- dNTPs)

Product code	02-012　200 U　　02-012-5　200 U x 5
Size	200 U
Storage	-20℃
Concentration	5 U/µl
Product Description	*Thermus aquaticus* DNA polymerase (Taq DNA polymerase) was expressed in *E. coli* in large quantities and highly purified. The enzyme has thermostable DNA polymerase activity and the MW is 94 kDa. This enzyme is suitable for PCR reactions; capable of amplifying DNA with various primers.
Definition of activity	One unit is defined as the amount of enzyme that can incorporate 10nmols of total dNTPs into an acid-insoluble material in 30 minutes at 74℃ when activated salmon sperm DNA was used as template / primer.
Purity	Greater than 95% purity as determined by SDS-PAGE (CBB staining) (Fig.1) The absence of endonucleases and exonucleases was confirmed.
PCR Test	Good amplification result was obtained in PCR reaction using λDNA as a template up to 14 kB (Fig.2)
Components	**Taq DNA polymerse (5U/µl)**: 20 mM Tris-HCl (pH 8.0), 100 mM KCl, 0.1 mM EDTA, 1 mM DTT, 50% glycerol, 0.5% Tween20, 0.5% Igepal CA-630 (02-Taq 40µl) **10 x Robust Buffer (Taq)** (02-Trb 1.0ml)
Application	1. High-throughput PCR 2. Colony PCR 3. Incorporation of dUTP, dITP, and fluorescence-labeled nucleotides 4. Primer extension 5. Addition of a single nucleotide (adenosine) at the 3'-blunt ends for cloning into TA vector. General composition of PCR reaction mixture (total 50µl) *Taq* DNA polymerase (5 U/µl) (02-Taq) ※0.25 µl 10x Robust Buffer (Taq) (02-Trb) 5 µl 2.5mM (each) dNTPs (02-Dnt) 4 µl Template <500 ng Primer 1 0.2~1.0 mM (final conc.) Primer 2 0.2~1.0 mM (final conc.) Sterile distilled water up to 50 µl ※ Use of excess amount is not recommended. **Cautions for using Robust Buffer (*Taq*)** Robust Buffer induces maximum enzymatic activity. To avoid production of undesirable smear bands in gel electrophoresis analysis, the optimal reaction time is recommended as follows: 1) about 5 to 10 seconds / kb elongation time for template up to 8 kb, and about 15 seconds / kb for up to 14 kb; 2) roughly the same elongation time is set with2-step PCR (shuttle PCR) and 3-step PCR; 3) extend the elongation time by short steps when amplification is not seen. Amplification can be detected more rapidly by adopting 2-step PCR.
Related product	02-001 Taq DNA Polymerase with Standard Buffer (+dNTPs) 02-002 Taq DNA Polymerase with Robust buffer (+dNTPs)
Please note: All products are FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES. NOT FOR MILITARY USE.	

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Data Images: 02-012 Taq DNA Polymerase with Robust Buffer (-dNTPs)

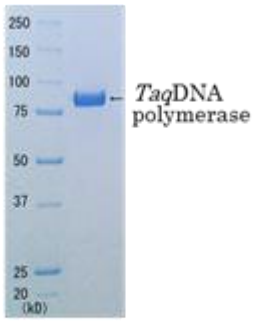


Fig.1 SDS-PAGE of *Taq*DNA polymerase

Protocols for PCR

Examples of PCR conditions for the amplification of various sizes of λDNA (results shown in Fig.2)

2 kb, 4 kb

94 °C	1 min	
95 °C	5 sec	} 25 cycles
65 °C	20 sec	

6 kb

94 °C	1 min	
95 °C	5 sec	} 25 cycles
65 °C	1 min	

8 kb

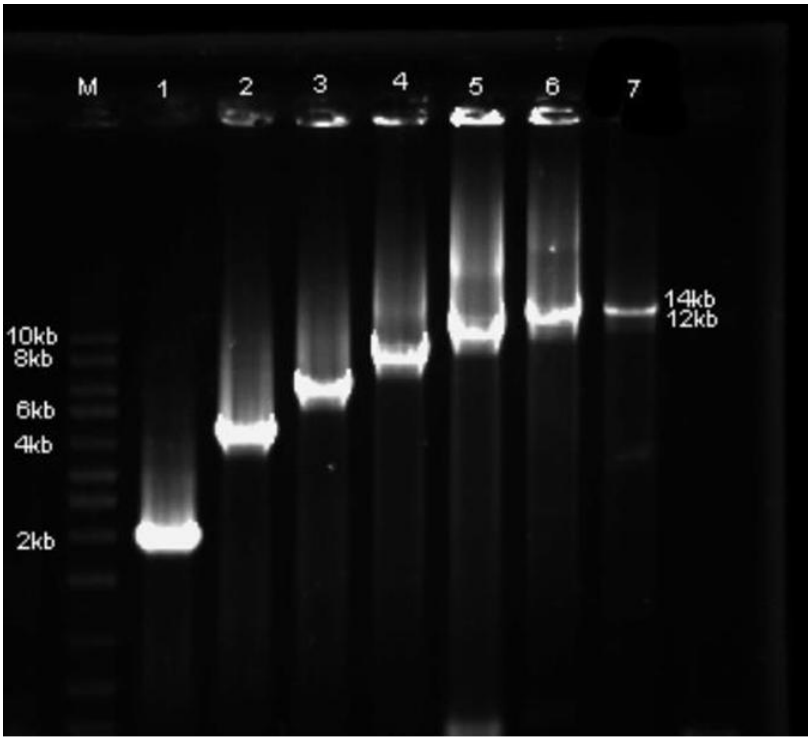
94 °C	1 min	
95 °C	5 sec	} 25 cycles
65 °C	1 min 20 sec	

10 kb, 12 kb

94 °C	1 min	
98 °C	5 sec	} 30 cycles
68 °C	3 min	
72 °C	3 min	

14 kb

94 °C	1 min	
98 °C	5 sec	} 30 cycles
68 °C	4 min	
72 °C	4 min	



M: marker,
lane1: 2kb, lane2: 4kb, lane3: 6kb, lane4: 8kb,
lane5: 10kb, lane6: 12kb, lane7: 14kb,

Fig.2 PCR products obtained by using Robust buffer (agarose gel electrophoresis)

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