

DF Taq DNA Polymerase Basic kit, 1000 U (Cat No: PD010S)

Description:

DF Taq DNA polymerase is a thermostable DNA polymerase manufactured with the highest standards. It is certified as DNA free (DF) as well as RNase and DNase free, that guarantee reproducible and efficient PCR reactions.

Taq DNA Polymerase is a thermostable enzyme that catalyzes 5'→3' synthesis of DNA. The enzyme has no detectable 3'→5' proofreading exonuclease activity but possesses low 5'→3' exonuclease activity. DF Taq DNA Polymerase has high processivity up to 5 kb, while it can reliably amplify a substrate as low as 6-12 copies per reaction e.g. 16 S rRNA fragment is consistently detected by PCR containing 1.25 units of DF Taq DNA polymerase in reactions where 10 fg of *E.coli* genomic DNA standard is added

Source:

The Enzyme is purified from an *E. coli* strain carrying a plasmid with Taq DNA polymerase gene from *Thermus aquaticus* YT-1.

Unit definition:

One unit is defined as the amount of enzyme required to catalyze the incorporation of 10 nmoles of dNTPs into acid insoluble material in 30 minutes at 72°C.

Reagents supplied:

PD010S kit		
Cat No	Description	Tubes No.
PD011S	DF Taq DNA polymerase, 5u/μl, 1000u, 0.2 ml (50 mM Tris-HCl (pH 7.9 @ 25oC), 50 mM KCl, 0.1 mM EDTA, 50% glycerol, 0.5% IGEPAL, 1 mM DTT, 0.5% Tween-20)	1
BR006-1.5	10x Taq DNA polymerase Buffer with 15mM MgCl ₂ (100 mM Tris-HCl (pH 8.3), 500 mM KCl, 15mM MgCl ₂)	1
BR007-1.5	10x Taq DNA polymerase Buffer w/o MgCl ₂ (100 mM Tris-HCl (pH 8.3), 500 mM KCl)	1
BR008-1.5	25mM MgCl ₂	1

PCR Guidelines:

PCR recommended reaction:

Components	25µl assay	Final Concentration/ Quantity (25µl assay)	50µl assay	Final Concentration/ Quantity (50µl assay)
10x Taq pol. Buffer with 15mM MgCl ₂	2.5µl	1x	5µl	1x
dNTP mix 10mM each	0.5µl	0.2 mM each	1µl	0.2 mM each
10µM forward primer	0.5µl	0.2-0.5 µM	2.5µl	0.2-0.5 µM
10µM reverse primer	0.5µl	0.2-0.5 µM	2.5µl	0.2-0.5 µM
Template DNA	Variable	10 fg-500 ng*	Variable	100 fg-500 ng*
DF Taq DNA pol. (5u/µl)	0.25µl	1.25 u/25 µl reaction (0.05 u/µl)	0.25-0.5µl	1.25-2.5 u/50 µl reaction (0.025-0.05 u/µl)
Sterile ultrapure water	Up to 25 µl		Up to 50 µl	

* Substrate quantity is mostly dependent on the complexity and the purity of substrate and should be defined after testing for each primer set/substrate combination. For highly pure DNA substrates (OD260/OD280 ~ 2 and OD260/OD230 ~ 2,2) dissolved in water for molecular biology, 5-10 copies of specific target substrate are enough for end product gel visualization.

PCR recommended conditions:

Step	Temperature	Time
Initial denaturation	95°C	5 min
25-35 cycles		



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Denaturation	95°C	30sec
Annealing	45-68°C*	20sec
Extension	72°C	1min/kb
Final extension	72°C	5 min
Hold	4°C	Indefinitely

*Annealing temperature depends on primers' Tm

Functional Quality Control:

DF Taq DNA Polymerase is tested for performance in the polymerase chain reaction (PCR) using 1.25 units of enzyme to amplify:

- a 400-bp region of *E.coli* ribosomal DNA using 100 fg of *E.coli* gDNA as substrate in a 25µl reaction. The resulting PCR product is visualized as a single band on agarose gel.
- a 5000-bp region of Lambda DNA using 20ng as substrate in a 50µl reaction. The resulting PCR product is visualized as a single band on agarose gel.

Other Quality Controls:

Certified as DNase-RNase free using fluorescently labeled RNase and DNase probes. DNA free contamination is tested extensively through PCR for the detection of *E.coli* 16S ribosomal DNA. More details concerning quality controls could be found in respective Certificate of analysis.

Shipping

Shipped on blue ice or dry ice

Storage conditions

Store at -20°C ± 5°C

Shelf life:

12 months upon production