Pfu DNA polymerase

Ver. 1.02

Cat. No. 503 - 025

Storage at : - 20 ℃

Description

Pfu DNA polymerase is purified from *E. coli* carrying a vector with the gene encoding *Pyrococcus furiosus* DNA polymerase. It has an intrinsic 3' to 5' exonuclease activity. It is a highly accurate DNA polymerase than Taq DNA polymerase (about 10 fold).

Components

10X Pfu reaction buffer	1 vial (800 ㎕)
dNTP mix (2.5 mM each)	1 vial (500 ㎕)
HQ buffer	1 vial (500 ㎕)

Purity

protease activity	None detected
SDS-PAGE	single band

Unit definition

One unit is the amount of Pfu DNA polymerase required to incorporate 10 nmol of dNTP into acid-insoluble product in 30 minutes at 72 ℃

Storage buffer

50 mM	Tris-HCl (pH 7.9)
50 mM	KCl
0.1 mM	EDTA
1 mM	DTT
0.5 mM	PMSF
50 %	glycerol (v/v)

Thermal PCR condition

95 ℃	2 min	
95 ℃	20 sec	30 - 35 cycles
A ℃	10 sec	
72 ℃	**B** min	
72 ℃	2 - 5 min	

A : The value is 4 ~ 6 lower than Tm of primers
Tm = 2 (A+T) + 4 (G+C)

B : below 2 kb 0.5 - 1 min/kb
more than 2 kb 1 - 2 min/kb

Reaction mixture

10X Pfu reaction buffer	5 ㎕
(optional : HQ buffer	5 - 20 ㎕)
dNTP mix (2.5 mM each)	4 ㎕
primer 1	5 - 10 pmol
primer 2	5 - 10 pmol
template	- ㎕
Pfu (2.5 U/㎕)	0.5 - 1 ㎕
DW	up to 50 ㎕

HQ buffer

-. In GC-rich reaction, HQ buffer increases the activity of Pfu DNA polymerase.
-. HQ buffer removes a hair-pin structure of GC-rich region.
-. The dilution factor of HQ buffer is variable, 0.5x - 2x, depending on a case by case basis.
-. We recommend to use of HQ buffer in PCR reaction of long-size target.