

nPfu-Special

Cat.#	Size	Size
P100S	200 units	2 units/μl
P100M	400 units	2 units/μl
P100L	1,000 units	2 units/μl

Store at -20°C

Supplied with: 10X nPfu-Special A Buffer
10X nPfu-Special B Buffer
100% DMSO
dNTP Mixture
Sterile water

Product description

nPfu-Special was cloned from *Pyrococcus furiosus* and genetically modified for high processivity. nPfu-Special DNA polymerase is highly efficient in amplifying longer DNA than 10 kb in relatively short period of time (15-30 sec/kb). It has lower error rate of 4.4×10^{-7} , 50 and 6 times lower than Taq and wild type Pfu DNA polymerase, respectively.

Characteristics

- Low error rate : 4.4×10^{-7}
Amplify blunt-end PCR products

Applications

- DNA amplification for cloning purpose
- DNA amplification for blunt-end vector cloning
- Amplification of longer DNA than 5 kb ~ 10 kb
- DNA base sequence analysis

For Research Use Only. Not for use in diagnostic procedures.

ISO9001 ISO14001 ISO13485

Quality control

- Purity: >99% on SDS-PAGE
- Endonuclease free
- Exonuclease free

Unit definition

One unit is defined as the amount of enzyme required to incorporate 10 nmol of dNTPs into acid insoluble materials in 30 min at 74°C in a 50 μl reaction mixture using, 12.5 μg of DNase I-activated calf thymus DNA as template.

Storage buffer

20 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM DTT, 100 mM KCl, 200 μg/ml BSA, 50% glycerol

nPfu-Special Buffer

- 10X nPfu-Special A Buffer
- 10X nPfu-Special B Buffer
• Buffer A is recommended for a regular PCR, but buffer B can be used when PCR is not successful with buffer A in absence and presence of DMSO

Quality control

- Purity: >99% on SDS-PAGE
- Endonuclease free
- Exonuclease free

Unit definition

One unit is defined as the amount of enzyme required to incorporate 10 nmol of dNTPs into acid insoluble materials in 30 min at 74°C in a 50 μl reaction mixture using, 12.5 μg of DNase I-activated calf thymus DNA as template.

Storage buffer

20 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM DTT, 100 mM KCl, 200 μg/ml BSA, 50% glycerol

nPfu-Special Buffer

- 10X nPfu-Special A Buffer
- 10X nPfu-Special B Buffer
• Buffer A is recommended for a regular PCR, but buffer B can be used when PCR is not successful with buffer A in absence and presence of DMSO

Product description

nPfu-Special was cloned from *Pyrococcus furiosus* and genetically modified for high processivity. nPfu-Special DNA polymerase is highly efficient in amplifying longer DNA than 10 kb in relatively short period of time (15-30 sec/kb). It has lower error rate of 4.4×10^{-7} , 50 and 6 times lower than Taq and wild type Pfu DNA polymerase, respectively.

Characteristics

- Low error rate : 4.4×10^{-7}
Amplify blunt-end PCR products

Applications

- DNA amplification for cloning purpose
- DNA amplification for blunt-end vector cloning
- Amplification of longer DNA than 5 kb ~ 10 kb
- DNA base sequence analysis

For Research Use Only. Not for use in diagnostic procedures.

Standard PCR conditions

- PCR mixture^a

10X nPfu-Special Buffer	2 μl
nPfu-Special DNA polymerase ^b (2 units/μl)	0.5 μl
dNTP mixture (2 mM each)	2 μl
Template DNA ^c	1 μl
Primer 1 (5 pmole/μl)	1 μl
Primer 2 (5 pmole/μl)	1 μl
Sterile water	up to 20 μl
^a Assemble the reaction mixture on ice	
^b Add the PCR polymerase at the final step	
^c Plasmid DNA, 0.1 ng~30 ng; genomic DNA, 50 ng~500 ng	

- PCR cycle

Initial denaturation	95°C	2 min
Denaturation	95°C	10 sec
Annealing ^a	55°C~65°C	5~30 sec
Elongation	72°C	15~30 sec/kb
Number of cycles	25~35 times	
Final elongation	72°C	5 min

※ Two-step PCR is recommended for longer PCR products than 5 kb. Use 68°C for both elongation and annealing temperatures. Reduce the elongation time or amount of enzyme if smeared PCR bands appear. Try reduced amounts of enzyme if the template amount is low. Use DMSO (3-10%) to amplify difficult template targets (GC-rich).

Standard PCR conditions

- PCR mixture^a

10X nPfu-Special Buffer	2 μl
nPfu-Special DNA polymerase ^b (2 units/μl)	0.5 μl
dNTP mixture (2 mM each)	2 μl
Template DNA ^c	1 μl
Primer 1 (5 pmole/μl)	1 μl
Primer 2 (5 pmole/μl)	1 μl
Sterile water	up to 20 μl
^a Assemble the reaction mixture on ice	
^b Add the PCR polymerase at the final step	
^c Plasmid DNA, 0.1 ng~30 ng; genomic DNA, 50 ng~500 ng	

- PCR cycle

Initial denaturation	95°C	2 min
Denaturation	95°C	10 sec
Annealing ^a	55°C~65°C	5~30 sec
Elongation	72°C	15~30 sec/kb
Number of cycles	25~35 times	
Final elongation	72°C	5 min

※ Two-step PCR is recommended for longer PCR products than 5 kb. Use 68°C for both elongation and annealing temperatures. Reduce the elongation time or amount of enzyme if smeared PCR bands appear. Try reduced amounts of enzyme if the template amount is low. Use DMSO (3-10%) to amplify difficult template targets (GC-rich).

India Contact:

Life Technologies (India) Pvt. Ltd.
306, Aggarwal City Mall, Opposite M2K Pitampura,
Delhi – 110034 (INDIA).
Ph: +91-11-42208000, 42208111, 42208222
Mobile: +91-9810521400
Fax: +91-11-42208444
Email: customerservice@lifetechindia.com
Web: www.lifetechindia.com

nPfu-Special

Cat.#	Size	Size
P100S	200 units	2 units/μl
P100M	400 units	2 units/μl
P100L	1,000 units	2 units/μl

Store at -20°C

Supplied with: 10X nPfu-Special A Buffer
10X nPfu-Special B Buffer
100% DMSO
dNTP Mixture
Sterile water

India Contact:

Life Technologies (India) Pvt. Ltd.
306, Aggarwal City Mall, Opposite M2K Pitampura,
Delhi – 110034 (INDIA).
Ph: +91-11-42208000, 42208111, 42208222
Mobile: +91-9810521400
Fax: +91-11-42208444
Email: customerservice@lifetechindia.com
Web: www.lifetechindia.com