enzynomics

*n*Taq-Pure HOT (purified from insect cell)

Cat.#	Size	Conc.
P725PI	250 units	5 units/μl
P750PI	500 units	5 units/μl

Store at -20℃

Supplied with: 10X *n*Taq-HOT Buffer
dNTP Mixture(2 mM each)
Sterile water

India Contact:

Life Technologies (India) Pvt. Ltd.
306, Aggarwal City Mall, Opposite M2K Pitampura,
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Email: customerservice@lifetechindia.com
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Product description

*n*Taq-Pure HOT (purified from insect cell) is a highly purified polymerase especially for PCR reactions where freedom from bacterial genomic DNA is essential. A gene encoding the *Thermus aquaticus* (Taq) DNA polymerase was cloned and expressed in Spodoptera frugiperda (Armyworm), and the enzyme was purified to homogeneity. *n*Taq-Pure HOT (purified from insect cell) is a hot start PCR polymerase which remains inactive at temperatures lower than 75℃. Therefore, a heat activation step is required to activate *n*Taq-Pure HOT (purified from insect cell). Activation is accomplished at denaturation steps of PCR cycles and, thus, *n*Taq-Pure HOT (purified from insect cell) is active only after initiation PCR cycles. Nonspecific elongation of incorrectly annealed primers before initiation of PCR reaction is one major cause of nonspecific DNA amplification. For this reason, amplification of nonspecific bands can be effectively prevented by using *n*Taq-Pure HOT (purified from insect cell), thus increasing the specificity of target DNA amplification. In addition, *n*Taq-Pure HOT (purified from insect cell) was purified by state of the art technology for minimizing the *E. coli* genomic DNA contamination. PCR product was not detected after 40 cycles of amplification using *E. coli* 16S RNA primer.

Characteristics

- Absolutely free from E. coli genomic DNA contamination
- Molecular weight: 94 kDa
- Error rate: 2.4 X 10^{-5}
- Thermal stability: Half life of 40 min at 95℃
- A-tail formation at 3' ends of amplified duplex DNA
- No activity at temperatures lower than 75℃. Heating up to 95℃ results in activation of enzyme

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

ISO9001 ISO14001 ISO13485

Quality control

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

Applications

- High specific amplification of DNA fragments shorter than 3 kb.
- Amplification of cDNA and genomic DNA.
- Amplification of template DNA with secondary or higher- ordered structure that is resistant to PCR amplification
- Well-suited for an automated PCR machines, for which PCR reaction mixtures are prepared at room temperature
- Primer extension
- Multiplex PCR

Unit definition

One unit is defined as the amount of enzyme required to incorporate 10 nmol of dNTP into acid insoluble materials in 30 min at 74℃ in a 50 μl reaction mixture (20 mM Tris-HCl/pH 8.8, 50 mM KCl, 2.5 mM MgCl2, 10 mM β-mercaptoethanol, 12.5 μg of calf thymus DNA).

Storage buffer

20 mM Tris-HCl (pH 7.9), 100 mM KCl, 0.1 mM EDTA, 1 mM DTT, 0.5% NP-40, 0.5% Tween-20, 50% glycerol.

10X *n*Taq-HOT Buffer

Containing 15 mM $MgCl_2$

Cautions

When nTaq-Pure HOT DNA is used, initial denaturation of 10 min is required to ensure effective activation of the enzyme.

LIFE TECH Life Technologies™
Your Molecular & Cell Technology Partner

Standard PCR conditions

- PCR mixture

10X *n*Taq-HOT Buffer	2 μl
*n*Taq- Pure HOT(purified from insect cell)[a] (5 units/μl)	0.2 μl
dNTP Mixture (2 mM each)	2 μl
Template DNA[b] (0.1~500 ng/μl)	1 μl
Primer 1 (5 pmole/μl)	1 μl
Primer 2 (5 pmole/μl)	1 μl
Sterile water	up to 20 μl

[a]Add the PCR polymerase at the final step
[b]Plasmid DNA, 0.1 ng~30 ng; genomic DNA, 50 ng~500 ng

- PCR cycle

Initial denaturation[a]	95℃	10 min
Denaturation	95℃	30 sec
Annealing[b]	55℃~65℃	30~60 sec
Elongation	72℃	1 min/kb
Number of cycles	25~35 times	
Final elongation	72℃	5 min

When cycles are over, keep the reaction mixture at 4℃; may add 10 mM EDTA until use to prevent DNA degradation.
[a]At least 10 min of initial denaturation time is required to fully activate the chemically modified PCR DNA polymerase
[b]Recommended annealing temperatures is 5 to 10℃ below the lower Tm of the two primers used.

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