

*n*Tag-Tenuto

Size Cat # Size P225B 250 units 5 units/µl P250B 500 units 5 units/ul

Store at -20℃

Supplied with: 10X nTag-Tenuto Buffer (Mg2+ free)

dNTP Mixture (2 mM each)

GC Melt I GC Melt II 25mM MqCl₂ Sterile water

India Contact:

Life Technologies (India) Pvt. Ltd. 306, Aggarwal City Mall, Opposite M2K Pitampura,

Delhi - 110034 (INDIA).

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Fax: +91-11-42208444

Email: customerservice@lifetechindia.com

Web: www.lifetechindia.com

Product description

nTag-Tenuto is nTag (Cat.# P025, P050) supplemented with 3'→5' proofreading activity and a PCR enhancing factor for improved efficiency and fidelity. nTag-Tenuto DNA Polymerase can be used to amplify DNA longer than 10 kb, which is difficult with common Tag polymerases alone. Thus, this product is improved in both fidelity (> 2 fold) of PCR products and amplification efficiency of longer PCR products. 10X nTag-Tenuto Buffer contains 20 mM Mg²⁺ and loading dyes,

Characteristics

- Molecular weight: 94 kDa
- Error rate: 3 0 X 10-6
- Thermal stability: Half life of 40 min at 95℃
- A-tail formation at 3' ends of amplified DNA products.

Applications

- Amplification of long DNA fragments (>5~15 kb)
- Amplification of high-complexity template DNA such as cDNA and genomic DNA
- Primer extension
- Colony PCR
- Multiplex PCR
- Labeling of DNA fragments with radioactive-isotopes
- Nucleotide sequencing

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

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°C in a 50-µl reaction mixture (20 mM Tris-HCl/pH 8.8, 50 mM KCl, 2.5 mM MaCl2, 10 mM β-mercaptoethanol, 12.5 μg of calf thymus DNA).

Storage buffer

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

10X nTag-Tenuto Buffer (Mg2+ free)

Ma2+ free buffer: Ma2+ free

Life Technologies™

Your Molecular & Cell Technology Partner

Standard PCR conditions 10X nTag-Tenuto Buffer

- PCR mixture^a

(Mg ²⁺ free/Dve plus)	2 μΙ
nTaq-Tenuto DNA Polymeraseb (5 units/µl)	0.2 µl
dNTP mixture (2 mM each, final conc., 200 μM each)	2 μΙ
MgCl ₂ ^c (25 mM)	Xμl
Template DNAd (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 Assemble the reaction mixture on ice	
^b Add the PCR polymerase at the final step	
cAdjust Mg ²⁺ to 1.5~5 mM with 25 mM MgCl ₂	
dPlasmid DNA, 0.1 ng~30 ng; genomic DNA, 5	0 ng~500 ng

- PCR cvcle

95℃	2 min	
95℃	30 sec	
55℃~65℃	30~60 sec	
72℃	1 min/kb	
25~35 times		
72℃	5 min	
When cycles are over, keep the reaction mixture at 4°C; may add 10 mM EDTA until use to prevent DNA degradation.		
^a Recommended annealing temperatures is 5 to 10℃ below the lower Tm of the two primers used.		
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Mg2+ free buffer: Mg2+ free

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Standard PCR conditions

- PCR mixture

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Primer 2 (5 pmole/µl)	1 µl
Sterile water	up to 20 μl
^a Assemble the reaction mixture on ice	
bAdd the PCR polymerase at the final step	
cAdjust Mg ²⁺ to 1.5~5 mM with 25 mM MgCl ₂	
dPlasmid DNA, 0.1 ng~30 ng; genomic DNA, 50	ng~500 ng

- PCR cvcle

	Initial denaturation	95℃	2 min
	Denaturation	95℃	30 sec
	Annealing ^a	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°C; may			

add 10 mM EDTA until use to prevent DNA degradation. ^aRecommended annealing temperatures is 5 to 10℃ below

the lower Tm of the two primers used.