

DFS-“Hot” Taq DNA Polymerase

Cat.-No.: N150 , 500 units

Optimized mixture of DFS-Taq Plus DNA and monoclonal Antibodies

Description:

Hot Start Taq DNA Polymerase is the optimized mixture of DFS-Taq Polymerase and Anti-Taq monoclonal antibodies. Antibodies block polymerase activity during set-up of the PCR reactions at ambient temperature (20-22 °C). The inhibition of Taq DNA polymerase is completely reversed when the temperature is above 70 °C. The PCR products obtained with Hot Start Taq DNA Polymerase are free from unspecific products and from primer-dimers.

Features:

- reliable and reproducible quantification in qPCR
- perfect for real time PCR
- especially for diagnostic purposes
- reaction set-up at room temperature
- activation of enzyme during first heating
- no change or optimization of protocol necessary
- high specificity, reduced primer mismatch or dimers

Applications:

- PCR with bacterial DNA
- Hot start PCR
- Real time PCR
- Amplification of complex genomic and cDNA templates
- Multiplex PCR
- High specificity PCR

Concentration: 5 U/μl.

Unit Definition: One unit of enzyme catalyses incorporation of 10 nanomoles of deoxyribonucleotides into acid-insoluble polynucleotide fraction in 30 min at 70C.

Activity assay: 50 mM Tris-HCl (pH 8.0 at 25°C), 50 mM NaCl, 10 mM MgCl₂, 200 μM dATP, 200 μM dCTP, 200 μM dGTP, 50 μM [3H] dTTP, 0,25 mg/ml activated calf thymus DNA.

Storage conditions: -20°C in 50 mM Tris-HCl (pH 8.0 at 25°C), 50 mM NaCl, 0.1 mM EDTA, 1 mM DTT, 50% glycerol and 1% triton X-100.

Reaction Buffers provided:

Ammonium-Reaction buffer (10X) “incomplete”

Ammonium-Reaction buffer (10X) “complete” with 25mM MgCl₂

MgCl₂ (100 mM)

Quality control: Endo-, exodeoxyribonucleases, ribonucleases free. Free of bacterial DNA-traces

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Components	Volume per reaction
10X reaction buffer	5 µl
100 mM MgCl ₂	optional
dNTP-Mix (40mM)	1.0 µl
Up-stream primer (10 µM stock)	0,5-2.5 µl
Down-stream primer (10µM stock)	0.5-2,5 µl
Template DNA	0.1-15 ng/ml plasmid DNA 1-10 µg/ml genomic DNA
DFS- Hot Taq DNA (5 u/µl)	0.2 - 1.0 µl
Sterile dest. Water (molecular grade)	up to 50 µl total reaction volume

Note:

- vortex all solutions carefully before using
- add the enzyme after Template DNA
- may you have to optimize the MgCl₂ concentration for best result

General Thermo-Cycler protocol:

Step	Time	Temperature
Initial denaturation	2-5 min	94-95°C
25-30 Cycles:		
Denaturation	10-25 sec	94-95°C
Annealing	10-25 sec	55-65°C
Extension	60 sec	72°C per 1kb
Final extension	5 min	72°C

Note:

- In case of low amount of DNA template, additionally cycles may be used

Cat.-no	Description	Amount
N150	DFS-"HOT" Taq DNA Polymerase	500 units
N152	DFS-"HOT" Taq DNA Polymerase	5x500 units
N154	DFS-"HOT" Taq DNA Polymerase	20x500 units

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