

HotTaq DNA Polymerase

Cat. No.	Pack size	Conc.	Supplied buffer
E031	500 u	5 u/μl	x10 Taq Buffer, 25mM Mg ²⁺ x10 Taq Buffer, without Mg ²⁺ 25 mM MgCl ₂ solution
E032	5'000 u	5 u/μl	x10 Taq Buffer, 25mM Mg ²⁺ x10 Taq Buffer, without Mg ²⁺ 25 mM MgCl ₂ solution

Description:

HotTaq Polymerase is a chemically modified version of Taq polymerase. HotTaq polymerase specific feature is that it allows to carry out a so called PCR with a "hot start" (Hot Start PCR). Using HotTaq polymerase makes it possible to increase the sensitivity and specificity of a PCR. It can increase yield, reduce nonspecific amplification and give a chance to achieve a result, if the product is not obtained in a normal PCR conditions. HotTaq Regular and HotTaq Ultimate are two alternative versions of chemically modified Taq polymerase.

HotTaq Regular polymerase has properties closer to the polymerases with antibodies. This version can be used in most cases where the hot start is wanted. Read distance with the use of this polymerase does not differ from one with non-modified polymerase.

HotTaq Ultimate polymerase provides a more strong hot start. We recommend using this polymerase for a hot start PCR with extreme conditions. If you look for more than 1 kb fragment, we do not recommend using this enzyme.

We recommend using HotTaq Regular polymerase in the RealTime PCR in the first place.

Taq polymerase is a thermostable enzyme with mass of approx. 94kDa. The source of the enzyme is a strain of *E.coli* with plasmid encoding the sequence of Taq DNA polymerase. The enzyme has activity maximum at 70-74 °C (although the enzyme is able to work at lower temperatures of about 25 °C). Enzyme's half-life at 94 °C is at least 45 min. The enzyme catalyzes DNA synthesis in 5'→3' direction in the presence of Mg²⁺ and at the appropriate pH. It also has a 5'→3' exonuclease activity (this activity is realized only on double-stranded DNA). The presence of the 5'→3' exonuclease activity makes the enzyme suitable for use in RealTime PCR.

Taq polymerase adds extra adenines at the 3'-end of PCR products. It does not create interferences with cloning. If it is necessary to obtain PCR products without extra adenines we recommend using Pfu polymerase. The maximum length of PCR products that can be obtained using Taq polymerase is approx. 5,000 bp.

Applications:

- Hot Start PCR
- RealTime PCR

Buffers:

- **x10 HotTaq Buffer, 25 mM Mg²⁺**
700 mM Tris-HCl, pH 8.3 at 25 °C,
166 mM (NH₄)₂SO₄,
25 mM MgCl₂
- **x10 HotTaq Buffer, without Mg²⁺**
700 mM Tris-HCl, pH 8.3 at 25 °C,
166 mM (NH₄)₂SO₄

Storage buffer:

50% glycerol (v/v),
20 mM Tris-HCl, pH 7.5 at 25 °C,
100 mM NaCl,
0.1 mM EDTA,
2 mM DTT,
0.2% Tween 20

Concentration:

5 u/μl

Unit definition:

One unit is defined as the amount of enzyme required to catalyze the incorporation of 10 nmol of dNTPs into an acid-insoluble form in 30 minutes at 70 °C.

Quality control:

The enzyme is tested for the absence of nicking activities, of endo-, exodeoxyribonucleases and ribonucleases. Activity and thermostability tested in PCR.

Storage conditions:

Store at -20 °C

Shipping conditions:

No special conditions required.

Safety and precautions:

Avoid contact with eyes, skin and clothing.

In case of contact with eyes, rinse immediately with plenty of water for at least 15 minutes and seek medical advice. Make the rinsing more thorough by separating the eyelids with fingers. If symptoms such as redness and irritation persist, seek medical attention.

In case of contact with skin, immediately wash the skin with soap and plenty amounts of water.

Recommended PCR protocol:

(the protocol can be modified according to use of another components or volume)

Mix the following components
(total volume - 25 µl):

x10 Taq Buffer, 25mM Mg ²⁺ or x10 Taq Buffer, without Mg ²⁺ 25 mM MgCl ₂	2.5 µl 2.5 µl 2.5 µl
2.5 mM dNTP Mix	2.5 µl
Primer 1 (10 pmol/µl)	0.5 µl
Primer 2 (10 pmol/µl)	0.5 µl
Template DNA (10 ng)	2.0 µl
HotTaq polymerase (5 u/µl)	0.5 µl
ddH ₂ O	up to 25 µl

PCR cycles:

Step	Temp.	Time
Initial denaturation	94 °C	3 min
Denaturation	94 °C	10 sec
Annealing	60 °C	10 sec
Elongation	72 °C	10 sec

30 cycles

Note: Annealing temperature, step duration and cycles number can be modified according to the actual tasks.