Instructions for use

HS Q-Taq, 5 U/µl

40 µl (200 units)

Recombinant Hot Start Taq DNA Polymerase, antibody blocked

1. Description

HotStart version of the recombinant heat stable Taq DNA polymerase from the thermophilic bacterium *Thermus aquaticus* in storage buffer, plus additional 10x concentrated PCR reaction buffer and 10x concentrated PCR reaction buffer with red gel loading dye.

For research use only. Not approved for use in clinical or *in vitro* diagnostics.

2. Applications

The ***HS Q-Taq*** polymerase is the optimal choice for all highly specific PCR amplifications as well as standard Taq-based cycling protocols. The antibody-mediated blocking of the DNA polymerase is released only at the initial denaturation step, hence resulting in highly specific amplification of the target sequence without production of unwanted side products caused by unspecific primer annealing. In combination with our unique buffers ***HS Q-Taq*** polymerase delivers specific PCR amplification of good yield with a wide range of PCR templates. ***HS Q-Taq*** is able to amplify PCR products up to at least 3 kb with genomic DNA and up to at least 5 kb in size with Lambda DNA and is appropriate for use in the amplification of DNA from genomic, viral, and plasmid templates. ***HS Q-Taq*** DNA polymerase possesses a 5´→ 3´ polymerase- as well as a 5´-flap endonuclease activity and generates a 3'dA (adenine)-overhang which may well be used for TA-cloning purposes.

3. Set contents

HS Q-Taq DNA polymerase in storage buffer containing 50 % glycerol.
PCR buffer (10x) with 20 mM $MgCl_2$.
PCR buffer RED (10x) with 20 mM $MgCl_2$ and 0.1 % cresol red (ready-to-load).
Filled in colour coded tubes.

Reagent	Amount	Lid colour
HS Q-Taq DNA polymerase, 5 U/µl	1 tube, 40 µl	red
10x PCR buffer	1 tube, 1 ml	blue
10x PCR buffer RED	1 tube, 1 ml	violet

The 10x PCR buffer RED contains a red dye which functions as a loading dye. The buffer has sufficient density for direct loading of PCR reactions onto an agarose gel for PCR product analysis. The red dye migrates in a 1% agarose gel at the same rate as a 1kb DNA fragment. The dye turns yellow at an acidic pH. The use of the colourless PCR reaction buffer is adequate for all general PCR applications and is particularly recommended when direct fluorescence or absorbance readings are required or any further proceedings.

4. Storage Buffer

50 mM Tris-HCl (pH 8.0), 100 mM KCl, 0.1 mM EDTA, 0.5 % IGEPAL CA-630, 0.5 % Tween-20, 1 mM DTT, 50 % glycerol

5. Enzyme activity

5 units/µl enzyme solution

6. Unit definition

One unit of activity is defined as the amount of enzyme required to incorporate 10 nmoles of dNTP into an acid-insoluble DNA fraction in 30 minutes at 72 °C.

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7. Suggested pipetting scheme

Set up can be done at room temperature.

Components	Apply for PCR reaction of 20 µl volume	Final concentration (recommended)
PCR buffer (10x)	2 µl	1x
dNTP-Mix (2 mM)	2 µl	800 µM (200 µM each)
Forward primer (e.g. 5 pmol/µl)	variable (e.g. 1 µl)	0.1-0.5 µM
Reverse primer (e.g. 5 pmol/µl)	variable (e.g. 1 µl)	0.1-0.5 µM
Template DNA	variable	0.01-10 ng / reaction
HS Q-Taq polymerase (5 U/µl)	variable (i.e. 0.2 µl)	0.5-1.5 U
Sterile dest. water	adjust to 20 µl final volume	

8. Basic amplification protocol

Step	Time	Temperature
Initial denaturation	2 minutes	92-95 °C
25-35 cycles		
Denaturation	2-10 seconds	92-95 °C
Annealing	2-10 seconds	55-68 °C
Extension	variable, depends on the length of product	72 °C

9. Notes

For maximum yield and specificity, annealing temperatures and annealing time as well as extension time and cycle numbers should be optimized for each template target and primer pair. Usually the optimal annealing temperature is 2-5 °C below the melting temperature of the primers. Recommended elongation time is 20-40 seconds per 1 kb of target. Elongation times of 30 seconds per 1 kb may be sufficient but longer elongation times may be necessary depending on the complexity of the template DNA.

10. Recommended $MgCl_2$ concentration

2 mM (final)

In case the $MgCl_2$ concentration has to be adjusted, use a separate $MgCl_2$ solution (10 mM) in PCR quality and add in appropriate amounts according to the scheme below. We recommend doing PCR with a $MgCl_2$ gradient in order to find the optimal concentration.

Pipetting scheme for additional $MgCl_2$

Final $MgCl_2$ conc. in mM	2.5	3	3.5	4
Add 10 mM $MgCl_2$ solution in following amounts to 20 µl reaction volume	1 µl	2 µl	3 µl	4 µl

11. Storage conditions

Store the enzyme at -20°C. However, short term storage (few hours) of the enzyme may be done at ± 0°C (wet ice). The enzyme is also stable at room temperature for at least 3 days.
The buffer should be stored at -20°C, but may also be stored at +4 °C for several weeks.

Product is not covered by pending or issued patents or may have certain limitations. To our best knowledge, this product does not provide any conflict with pending or issued patents.

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