

x-VITA™ Hot Start Taq Polymerase

TAQP-H01-001

Description

x-VITA™ Taq DNA polymerase is a Hot Start DNA polymerase designed to minimize unspecific amplification improving PCR specificity. Taq DNA polymerase bound to a proprietary antibody that blocks polymerase activity until denaturation step occurs. The heat labile antibodies are rapidly inactivated by raising the temperature (4 minutes at 95-97°C). This prevents or minimizes primer-dimer and nonspecific products. Like the Taq polymerase, the enzyme has 5'→3' polymerase activity and a weak 5'→3' exonuclease activity but no 3'→5' exonuclease activity (proofreading). Before enzyme activation none of enzyme activities are detectable.

Features and applications

- ✓ Adds extra nucleotides (preferentially adenine)
- ✓ without template at 3' ends leaving 3' overhangs PCR fragments. This fact allows the popular TA-cloning or GC cloning.
- ✓ Amplifies from a femptograms of DNA targets. Amplification from a limited DNA template or low copy number genes
- ✓ Real time PCR, RT-PCR and quantitative RT-PCR
- ✓ Genotyping with Taqman probes
- ✓ PCR fragments amplification for TA or GC cloning (preferably use a proofreading polymerase for cloning purpose and a blunt cloning vector)

Assay conditions

25 mM Tris-HCl pH 9.0 at 25 °C, 50 mM KCl, 2 mM MgCl₂, 0.1 mg/mL gelatine, 200 μM dATP, dGTP, dTTP, 100 μM [α 32-P] dCTP (0.05 μCi/nmol) and 12.5 μg activated salmon sperm DNA.

Unit definition

One unit is defined as the amount of enzyme required to catalyse the incorporation of 10 nanomoles of dNTPs into acid-insoluble material in 30 minutes at 74°C.

Quality Certifications

- ✓ Functionally tested in PCR
- ✓ Undetected bacterial DNA (by PCR)
- ✓ Undetectable nucleases activity (endo-, exo- and ribonucleases)

Storage

Store at -20°C.

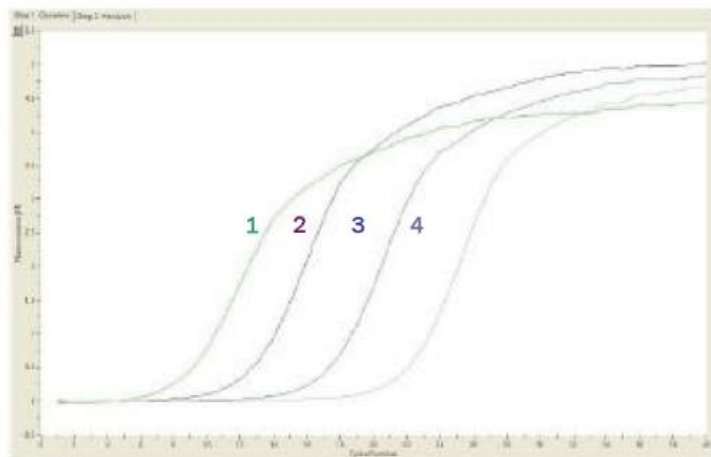
Product use limitation

This product is developed, designed and sold exclusively for research purposes and use only. The product is not intended for diagnostics or drug development, nor is it suitable for administration to humans or animals.

Recommended PCR assay (20 µl assay)

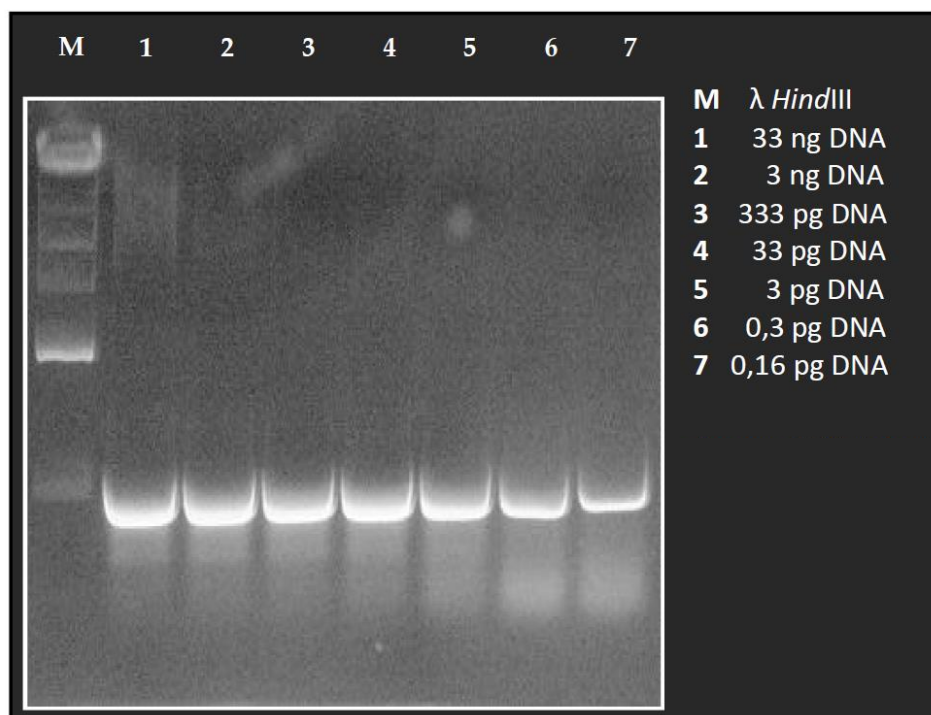
Components	Volume	Final concentration
10X PCR buffer	2 µL	1X
MgCl ₂ 25 mM	2 µL	2.5 mM
dNTPs 8 mM mixture	2 µL	0.8 mM
Primer Forward (15 mM)	1 µL	0.75 µM
Primer Reverse (15 mM)	1 µL	0.75 µM
Template DNA	0.2-10 µL	1.75-2.5 ng/µL
Taq DNA polymerase (5 U/µL)	0.2 µL	0.05 U/µL
PCR grade H ₂ O	to 20 µL	-

Cycling instructions: 94 °C 5:00, 40x (94 °C 0:35, Tm 0:35, 72 °C 1'/kb), 72 °C 7:00, 4 °C ∞



- 1 1000 pg DNA/ Ct=8,576
- 2 50 pg DNA/ Ct=12,52
- 3 2,5 pg DNA/ Ct=17,12
- 4 0,125 pg DNA/ Ct=21,87

Real time PCR in Light cycler (Roche) using x-VITA™ Hot Start Taq DNA polymerase



- M λ HindIII
- 1 33 ng DNA
- 2 3 ng DNA
- 3 333 pg DNA
- 4 33 pg DNA
- 5 3 pg DNA
- 6 0,3 pg DNA
- 7 0,16 pg DNA

Amplification of up 160 fg DNA using x-VITA™ Hot Start DNA polymerase