Data sheet

HotBegan Taq DNA polymerase

Cat. No: P0028 500 U (5 U/µL)
(Included 25 mM MgCl2 and 10x Buffer with BSA)

Introduction

HotBegan Taq DNA polymerase is a hot start DNA polymerase designed to minimize unspecific amplification improving PCR specificity. HotBegan Taq DNA polymerase is a **Horse-Power-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

- Inactive at room temperature.
- 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.

Applications

- 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) (see pSpark® DNA Cloning System).
- Amplification from a limited DNA template or low copy number genes.

Assay conditions

25mM Tris-HCl pH9,0 at 25°C, 50mM KCl, 2mM MgCl2, 0,1mg/mL gelatine, 200 µM de dATP, dGTP, dTTP, 100µM[α32-P]dCTP (0,05µCi/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.

Concentration: 5 U/µL

Quality Certifications

- ✓ Functionally tested in PCR.
- ✓ Non detected bacterial DNA (by PCR).
- ✓ Not detectable activity of nucleases (endo-, exo, and ribo-).

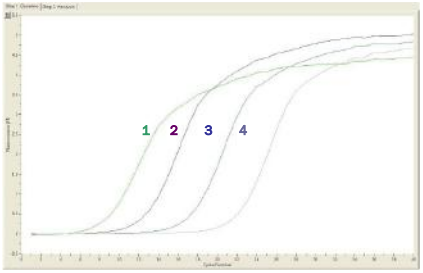
Storage: Upon receipt, store the entire kit at -20 °C

(Continued on reverse side)

Recomended PCR assay (20 µl assay)

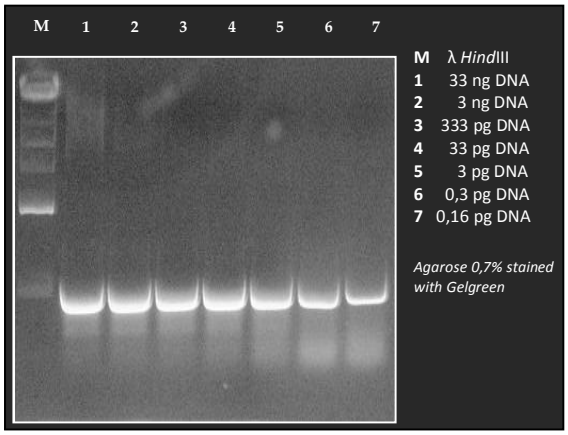
10X PCR buffer B	2µl (1X)
$MgCl_2$ (25 mM)	2µl (2,5mM)
dNTPmix (2 mM each dNTP)	2µl (200 µM each dNTP)
Forward Primer (15µM)	1µl (0,75 pmol/µL)
Reverse Primer (15µM)	1µl (0,75 pmol/µL)
Template DNA	plasmide: 30-75ng; gDNA: 100-500ng
Polymerase(5U/ µl)	0,2 µl (1U)
PCR grade H20	up to 20 µL

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



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 HotBegan Taq DNA polymerase.



Amplification of up 160 fg DNA using HotBegan DNA polymerase.

PRODUCT USE LIMITATION

This product is developed, designed and sold exclusively for research purposes and in vitro use only. The product was not tested for use in diagnostics or for drug development, nor is it suitable for administration to humans or animals. Please refer to www.canvaxbiotech.com for Material Safety Data Sheet of the product.

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