VioTaq[™] DNA Polymerase

Description:

Recombinant VioTaq DNA Polymerase is expressed and purified from *E. coli* to a very high homogeneity without nuclease contamination. It consists of a single polypeptide with a molecular weight of about 92 kD. VioTaq is thermostable at 95 °C with a half-time of 30 minutes and replicates DNA at 70-74 °C. This enzyme has 5'-3' exonuclease and extra A addition activity without 3'-5' exonuclease activity. It has a remarkable DNA polymerization activity with a very low error frequency (~10⁵). VioTaq DNA polymerase is most suitable for large-scale PCR screening at a low cost.

Product Contents:

VioTaq DNA Polymerase (stored at -20 $^\circ\!\!\!C$), 500 U (2.5 U/µl); 10X PCR Buffer, containing 20 mM MgCl_2

Storage Buffer:

20 mM Tris-HCl, pH 8.0; 0.1 mM EDTA; 1 mM DTT; 1.0% Triton X-100; 50% glycerol

Unit Definition:

One unit incorporates 10 nmole of dNTP into acid-precipitable material in 30 minutes at 74 $^\circ\!C.$

Basic PCR protocol:

1. Suggested set-up of a PCR reaction

Components	Volume	Final Conc.
10X PCR buffer plus	5 µl	1X (with 2.0 mM
MgCl ₂		MgCl ₂)
10 mM each dNTP	1 µl	0.2 mM each
10 μM 5' Primer	2.5 μl	0.5 μΜ
10 µM 3' Primer	2.5 μl	0.5 μM
Template DNA	1-10 μl	< 0.5 µg
VioTaq (2.5 U/µl)	1 µl	2.5 U
Add sterile ddH ₂ O to	50 μl (Fina	l volume)

- Mix contents and overlay with 50 μl of mineral oil. When using a thermal cycler with a heated lid, do not need to add mineral oil.
- 3. Suggested PCR amplification cycles

Initial Denaturation: 94 °C for 3-5 min.

Run 25-35 cycles of the following steps:

*Denature at 94 $^\circ\!C\,$ for 30-60 sec.

*Anneal at Temp (start as 5 $^\circ\!\! C\,$ below Tm) for 1 min.

*Elongate at 72 °C for 1 min. (for 1-2 kb product)

(Adjust elongation time based on product size,

1-2 kb per 1 min.)

Final Elongation: 72 $^\circ\!\mathrm{C}$ for 10 min.

Maintain at 4 °C or store at -20 °C.