

## **HIFI DNA Polymerase**

**Cat No:** HF500 **Sise:** 500u 5u/ul

## Description

Hi Fidelity polymerase is a mixture of thermostable enzymes. It is specifically developed to synthesize length of PCR product up to 30 kb and with low error rate. Hi Fi polymerase synthesizes higher yields of product from genomic DNA, cDNA, and bacterial cultures. It has 2.5 hours half life at 96oC and easily amplify PCR product of G-C rich or secondary structure DNA by adding G-C rich buffer.

storage conditions: -20°C

**10X reaction buffer:** Buffer containing 25mM MgCl2

**Unit description:** One unit is defined as the amount of enzyme that will incorporate 10nmole of dNTP into acid-insoluble material in 30 minutes at 74 °C. The reaction conditions are: 50mM Tris-HCl pH8.8, 50mM NaCl, 5mM MgCl2, 200mM each of dATP, dCTP, dGTP, dTTP, 10mg activated calf thymus DNA and 0.1mg/ml BSA in a final volume of 50 ul.

**Template:** Hi Fidelity Polymerase is suitable for amplifying targets up to 15 kb from the following templates:

Genomic DNA: 10–200 ng Plasmid DNA: 1–5 ng

cDNA : ~100 ng starting total RNA

Amplification of longer targets (up to 15 kb) may be possible, but may require more template and longer elongation times.

**Primers:** Use 0.3 mM per primer as a general starting point. For larger amounts of template (e.g., 200 ng genomic DNA), increasing the concentration up to 0.5 mM per primer may improve yield.

**Annealing Temperature:** The annealing temperature is slightly higher than with Typical PCR. The optimal annealing temperature should be  $\sim$ 2°C lower than the Tm of the primers used. A range of 58–68°C is recommended.

**Extension Time:** As little as 30 seconds per kb is suitable for most targets. Use up to 60 seconds per kb for maximum yield.

## Program the thermal cycler

Step	Temperature	Time	Cycle
Initial denaturation	94-95 °C	1-3 mins	1
Denaturation	94-95 °C	10-60 sec	
Annealing	50-68 °C	10-30sec	25-35
Extension	72 °C	1min/1kb	
Final extension	72 °C	1-10 mins	1

*IMPORTANT: Annealing temperature should be 2-6 °C lower than the primer melting temperature* 

Shipping and Storage conditions Shipping and temporary storage at -20 °C and for up to 1 month at room temperature has no detrimental effects on the quality of Hi Fidelity DNA polymerase.

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