



2X Hi Fi mix

Cat No	Pack size	conc
HFM0500	5ml	2x
HFM2500	5x5ml	2x

Description :Hi Fi polymerase synthesizes higher yields of product from genomic DNA, cDNA, bacterial cultures. It is contain a 2.5 hours half life at 96°C and easy to amplify PCR product at G-C rich and secondary structure.

2X Hi Fi Mix is optimized mixture contain of Hi Fi polymerase, reaction buffer, dNTP And enhancer as 2-fold concentration. 2x Hi Fi mix is designed to allow the user for quick ,easy preparation of reaction mixture. The 2x Hi Fi mix can be amplification PCR products up to 10-15 kb and the products can be directly cloning into T-vector.

storage conditions: long time at -20°C
short time at 4 °C

Template

2 x Hi Fi mix is suitable for amplifying targets up to 3 kb from the following templates:

Genomic DNA: 10–200 ng

Plasmid DNA : 1–5 ng

cDNA : ~100 ng starting total RNA

Primers

Use 0.3 μ M 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 μ M per primer may improve yield.

Annealing Temperature

The annealing temperature is slightly higher than with typical PCR. The optimal annealing temperature should be ~2°C lower than the T_m 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.

PCR Protocol:

1. Thaw the 2x Hi Fi mix at room temperature. Vortex the 2x Hi Fi mix and then spin it briefly in a micro centrifuge to collect the material in the bottom of the tube.
2. Prepare one of the following reaction mixes on ice:

Component	Volume
2x Hot start mix	12.5 ul
Primer1 (20 pmol)	1-2 ul
Primer2 (20 pmol)	1-2 ul
template	1-10 ul
ddH ₂ O	Up to 25 ul
Total	25 ul

3. If necessary you can scale up your volume

1. Program the thermal cycler as follows:

Step	Temperature	Time	Cycle
Initial denaturation	94-96°C	0.5-2mins	1
Denaturation	94-96°C	0.2-2mins	15-30
Annealing	50-68	0.2-2mins	
Extension	68-75	1min/1kb	
Final extension	68-75	1-10mins	1

Step

After cycling, maintain the reaction at 4°C. Samples can be stored at -20°C until use.

Analyze products using standard agarose gel electrophoresis.