AmpFi HS DNA Polymerase

DM-AmpHS400

Store at -20°C.

Description

AmpFi HS DNA Polymerase is a Taq Polymerase that has rapid extension rates and contains a proprietary antibody that blocks polymerase activity at low temperatures. HotStart allows for a reaction set-up at room temperature without non-specific amplification and primer dimer formation. This polymerase provides increased processivity, yields, and sensitivity, and reduced reaction times by up to 70%, compared to wild-type Taq DNA polymerase when conditions are optimized.

The antibody dissociates from the DNA polymerase during the initial denaturation step, restoring enzyme activity. Hence, reducing non-specific amplification and competition for reagent availability. This specificity increase improves the yield of PCR products. AmpFi HS DNA Polymerase has 5'-3' polymerase and 5'-3' exonuclease activities, lacks 3'-5' exonuclease activity, and produces 3'-dA-tailed amplicons. PCR products made with the polymerase can be used with TA cloning vectors.

Product Component	Quantity
AmpFi HS DNA Polymerase	400 rxn (200 µl)
AmpFi HS Buffer[1]	2 x 1.0 ml

*Buffer contains 1.5 mM Mg^{2+}

Protocol

1. Mix individual components before use and assemble reaction.

Component	Volume
AmpFi HS Buffer	5 µl
dNTP Mix (10 mM)	0.5 µl
Forward Primer (10 µM)	0.5 µl
Reverse Primer (10 µM)	0.5 µl
Template DNA	Variable (100 ng genomic DNA)
AmpFi HS DNA Polymerase	0.5 µl **
Nuclease-free H2O	Up to 25 µl

**Reaction volumes of 25 µl are recommended with 0.5 µl AmpFi HS DNA Polymerase. For difficult targets or crude samples, increase to 1 µl.

2. Gently mix the reaction components, and briefly centrifuge. Run thermocycling conditions for standard PCR:

Step	Temperature	Time
Initial Denaturation	95°C	10 min
25 – 35 Cycles	95°C 60°C *** 72°C	15 sec 15 sec 15 sec/kb
Final Extension	72°C	1 min

*** AmpFi HS Buffer allows for primer annealing at 60°C for most primers and adjust only if needed.

3. After PCR, maintain the reaction at 4°C or store at -20°C until use.
4. Analyze the amplification products by agarose gel electrophoresis.
5. Visualize by ethidium bromide or applicable staining.