AmpFi 2X qPCR MasterMix

DM-AmpqMM500

Store at -20°C.

Description

AmpFi 2X qPCR MasterMix provides analysis of DNA samples for quantitative real-time in a convenient, reliable setup.This qPCR MasterMix contains Taq Polymerase and DNA Polymerase, providing rapid extension rates. This polymerase provides increased yields, and sensitivity, while optimizing reaction times by up to 70%, compared to wild-type Taq DNA polymerase.

AmpFi 2X qPCR MasterMix has 5'-3' polymerase and 5'-3' exonuclease activities, lacks 3'-5' exonuclease activity, and produces 3'-dA-tailed amplicons. qPCR products made with AmpFi 2X qPCR MasterMix can be used with TA cloning vectors. The MasterMix contains dye comparable to SYBR Green™ and EvaGreen™.

Product Component	Quantity
AmpFi 2X qPCR MasterMix	500 rxn (4 x 1.25 ml)
ROX Reference Dye	50 µl

Protocol

The suggested quantity of ROX Reference Dye to incorporate into the MasterMix can differ based on the specific type of qPCR machine being used.

- No ROX equipment: Not needed.
- Low ROX equipment: 1 µl/1.25 ml or 22.5 µl/25 ml MasterMix.
- High ROX equipment: 11.5 µl/1.25 ml or 225 µl/25 ml MasterMix

1. Mix individual components before use and assemble reaction on ice.

Component	Volume
AmpFi 2X qPCR MM*	10 µl
Forward Primer (10 µM)	0.5 µl
Reverse Primer (10 µM)	0.5 µl
Template DNA	Variable (100 ng genomic DNA)
Nuclease-free H2O	Up to 20 µl

*Reaction buffer contains 1.5 mM Mg2+

2. Gently mix the reaction components, and briefly centrifuge. Use thermocycling conditions:

Step	Temperature	Duration (Standard)	Duration (Fast)	Cycles
Enzyme Activation	95°C	3 min	3 min	1
Denaturation	95°C	15 sec	1 sec	40
Annealing/Extension	60°C	1 min	10 sec	
Melting Curve	Refer to specific guidelines for the instrument used			

Notes

- This specialized buffer enhances yields, sensitivity, and specificity beyond what is achievable with wild-type Taq polymerase.
- Begin the qPCR immediately after preparing the reaction mixture. If immediate start is not feasible, store the mixture on ice until you commence the qPCR.
- Employ standard thermocycling conditions for miRNA cDNA templates or other suitable applications.