

PRODUCT INFORMATION

Thermo Scientific ABsolute qPCR Capillary Mix

#AB-1283/B 16 x 1.25 mL

Lot _ Expiry Date _

Ordering Information

Component	#AB-1283/B 2000 rxns of 20 µL
ABsolute qPCR Capillary Mix	16 x 1.25 mL

Store at -20°C



Description

Thermo Scientific ABsolute qPCR Capillary Mix has been developed to quantify DNA and cDNA. With the exception of primers and template, this 2X mix contains all the components required to perform a rapid, sensitive and reproducible qPCR reaction:

- Thermo Scientific Thermo-Start DNA Polymerase, a chemically modified hot-start version of Thermo Scientific ThermoPrime *Taq* DNA Polymerase, which prevents non-specific amplification during the reaction set-up. **This enzyme requires an activation step at 95°C for 15 minutes.** Thermo-Start™ has 5' to 3' polymerization and exonuclease activity but lacks 3' to 5' exonuclease activity (proofreading).
- Proprietary reaction buffer which provides highly sensitive, specific and consistent fluorescence readings for real-time and end-point analysis. This buffer has been optimized for MgCl₂ and enhancers to improve amplification across a wide range of templates including plant DNA and GC rich fragments.
- dNTP's, including dTTP to improve reaction sensitivity and efficiency compared to dUTP.

Cycler & Probe Compatibility

ABsolute™ qPCR Capillary Mix is compatible for use with any capillary format cyclers, including Roche Lightcycler® 2.0.

Storage Conditions

Store at -20°C until ready for use. ABsolute qPCR Capillary Mix can be stored at 4 °C for up to 1 month. Avoid repeated freeze thawing.

Additional Info

The use of disposable gloves, RNase and DNase free filter tips and plastics is recommended.

For optimal results, the recommended amplicon length is in the range of 60 to 300 bp.

As best performance is achieved with dTTP, the ABsolute QPCR Capillary Mix contains a nucleotide mix with dTTP instead of dUTP.

Protocol

Thaw the reagents on ice, mix the solutions and spin down before use to recover the maximum amount. Do not vortex the ABsolute qPCR Capillary Mix.

Briefly centrifuge to avoid bubbles within the wells, as these will interfere with the fluorescence. Always include a no template control (NTC).

Example of Reaction Mix preparation for a 20 µL final reaction:

	Volume	Final Concentration
ABsolute qPCR Capillary Mix (2X)	10 µL	1X
Forward primer (10 µM)*	0.8 µL	400 nM
Reverse primer (10 µM)*	0.8 µL	400 nM
Probe		100-250 nM
Template (DNA or cDNA)**	1-5 µL	< 250 ng/rxn
Water, nuclease-free (#R0581)	To 20 µL	
Total volume	20 µL	

*For optimization, a primer titration should be performed from 100 nM to 500 nM final concentration. Scale up or down the volume and concentration as appropriate.

**The volume of template to add to the qPCR reaction can be adjusted as required. For standard templates only 1 µL should be added to reduce carryover of PCR inhibitors. This volume can be increased up to 5 µL for low copy number templates.

Example of qPCR thermal cycling protocol:

	Temp.	Time	Number of cycles
Enzyme activation	95 °C	15 min	1 cycle
Denaturation	95 °C	15 s	40 cycles
Annealing/Extension*	60 °C	60 s	

*Separate annealing (50–60°C for 30 s) and extension steps (72 °C for 30 s) may be necessary with some probe systems (e.g. Molecular Beacons), as the optimal temperature for detecting fluorescence may be different.

CERTIFICATE OF ANALYSIS

ABsolute qPCR Capillary Mix is tested functionally using qPCR. The product must demonstrate linearity of amplification over a specified serial dilution of human genomic DNA.

Quality authorized by:

 Jurgita Zilinskiene

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