Product Information

Green-2-Go 1-Step TaqProbe RT-qPCR Mastermix-No Dye

Product information for QPCR008-NODYE:

Kit Components:

Components	100 Reactions (20 µl Per Reaction)
TaqProbe qPCR MasterMix - NODYE	1.25 ml
qRT-PCR Enzyme Mix (50X)	40 µl
Nuclease-free H2O	1 ml

Product Description
A complete qPCR system, the Green-2-Go 1-Step TaqProbe RT-qPCR Mastermix-no Dye Kit contains all reagents necessary for both Reverse Transcription (RT) and TaqProbe based qPCR amplification to occur in a single qPCR reaction tube. The Green-2-Go 1-Step TaqProbe RT-qPCR Mastermix-no Dye kit is an amalgamation of two key formulations; the qRT-PCR Enzyme Mix and the TaqProbe 2X qRT-PCR MasterMix within a proprietary blend of stabilizers and enhancers to enable a seamless coupling of two separate reactions into a real-time “single step” procedure.

Storage Conditions
Store all components at -20 °C in a non-frost-free freezer. All components are stable for 1 year from the date of shipping when stored and handled properly.

Protocol
RT-PCR should be assembled in a nuclease-free environment. RNA sample preparation, reaction mixture assembly, PCR and subsequent reaction analysis should be performed in separate areas. The use of “clean”, automatic pipettors designated for PCR and aerosol resistant barrier tips are recommended.

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1. Prepare the following reaction mixture in a qPCR tube on ice:

Components	Reaction Volume			Concentration
	10 µl	20 µl	50 µl	
Total RNA or Poly(A) + mRNA	Variable	Variable	Variable	5 pg - 1 µg/rxn 0.05 pg - 20 ng/rxn
TaqProbe qPCR MasterMix – No Dye	5 µl	10 µl	25 µl	1X
qRT-PCR Enzyme Mix (50X)	0.2 µl	0.4 µl	1 µl	1X
Forward Primer (10 µM)	0.3 µl	0.6 µl	1.5 µl	300 nM
Reverse Primer (10 µM)	0.3 µl	0.6 µl	1.5 µl	300 nM
TaqProbe	Variable	Variable	Variable	100-300nM
Nuclease-free H2O	Up to 10 µl	Up to 20 µl	Up to 50 µl	-

Note: *Sequence specific primers should be used.*

2. Gently mix and ensure all the components are at the bottom of the amplification tube. Centrifuge briefly if needed.
3. Program the thermal cycler so that cDNA synthesis is followed immediately by qPCR amplification.

Step	Temperature	Duration	Cycle(s)
cDNA Synthesis	42°C	15 mins	1
Pre-Denaturation	95°C	10 mins	1
Denaturation	95°C	15 secs	40
Annealing	60°C	60 secs	

Recommendations for Optimal Results

- Aliquot reagents to avoid contamination and repeating freeze-thaw cycles.
- TaqProbe qPCR MasterMix components are light sensitive; avoid prolonged exposure to intense light.
- Start PCR as soon as the reaction mixture is prepared and always keep the reaction mixture chilled in an ice box prior to qRT-PCR reaction.

For laboratory research only. Not for clinical applications.

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