Add-Probe RT-PCR Master (2x Conc.)

Research Use Only

Product Code

74201

Component

1. Add-Probe RT-PCR Master (2x conc.) 1.0 ml

Storage Condition

Store at -20℃

Description

Add-Probe RT-PCR Master (2x) provides sensitive and easy-to-use components which contain all the reagents for first strand cDNA synthesis and PCR reaction in one-tube by using TaqMan®, and is designed for high sensitivity and specificity on various real-time instruments.

Quality Control.

The performance of Add-Probe RT-PCR Master (2x) is tested in an RT and PCR one-tube reaction using human total RNA with specific primers and TaqMan® probes. The sensitivity of the kit is verified by the detection of GAPDH and Actin transcript in 10 pg total RNA after 30 cycles.

Storage and Stability

Add-Probe RT-PCR Master is stable for 2 years when stored in a constant temperature freezer at less than -20℃

Made in KOREA This product was manufactured through ISO 9001 & 13485 system.

Nucleic Acid Amplification Protocol

1. Add the following components to a thin-walled PCR tube:

Nuclease-Free H_20	x µl
Add-Probe RT-PCR Master (2x conc.)	10.0 µl
Forward primer (10 µM)	0.25~2.0 µl
Reverse primer (10 µM)	0.25~2.0 µl
TaqMan Probe (10 µM)	0.25~2.0 µl
(Optional) 50x ROX dye	x µl
RNA template	x µl
Total reaction volume	20 µl

* Recommendation for template RNA concentration in a 20 µl reaction volume
1) total RNA: 100 fg ~ 1 µg
2) mRNA: 10 fg ~ 1 µg

2. PCR cycling

cDNA synthesis	50℃, 20 min	
Initial denaturation	95℃, 10 min	
PCR cycling (30 – 40 cycles)	95℃, 10 sec	
	60℃, 30 - 60 sec	

[Note] 50x ROX dye
ROX dye can be included in the reaction to normalize the fluorescent reporter signal, for instruments which are compatible with that option.
50x ROX is a 25 µM concentration. Use the following table to determine the amount of ROX to use with a particular instrument.

Instrument	Final ROX concentration
AB 7000, 7300, 7700, 7900HT, 7900 Fast, StepOne and StepOnePlus	500 nM
AB 7500, 7500 Fast, Stratagene Mx3000P, Mx3005P and Mx4000	50 nM

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