AddGreen RT-qPCR Kit and Master (2x conc.)

Product Information

Product Code 71202, 71302

Provided with

Cat. No. 71202: AddGreen RT-qPCR Kit 1. 20x Enzyme Solution 0.1 mL 2 5x Reaction Buffer 0.4 ml

Cat. No. 71302: AddGreen RT-qPCR Master (2x conc.) 1.0 mL

Storage Conditions -10°C ~ -30°C

Stability Stable for 1 year from manufacturing date.

Components of AddGreen RT-qPCR Kit and Master

AddStart Taq DNA Polymerase, Reverse Transcriptase, RNase Inhibitor, Tris-HCl (pH8.5), Potassium Chloride, Ammonium Sulfate, MgCl₂, DTT, Protein stabilizers, Intercalating dye (AddGreen I) and dNTP mixture.

Description

AddGreen RT-qPCR Kit and Master provides sensitive and easy-to-use components which contain all the reagents for first strand cDNA synthesis and PCR reaction in one-tube reaction, and is designed for high sensitivity and specificity on various real-time instruments. Especially, thermostable MMLV RTase (RNase H-), AddStart Taq DNA Polymerase and RNase Inhibitor are included in 20x Enzyme Solution. AddGreen RT-PCR Master with UDG (2x conc.) is a ready-to-use mixture included all components for one-step RT-qPCR in one tube.

Applications

- Quantitative real-time PCR
- Gene expression analysis
- Genetic variation analysis

Manufacture

애드바이오메디텍

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Nucleic Acid Amplification Procedure

1-1. Add the following components to a thin-walled PCR tube for AddGreen RT-qPCR Kit:		
Nuclease-Free Water	xμl	
5x Reaction Buffer	4.0 µl	
Forward primer (10 μM)	0.25 ~ 2.0 μl	
Reverse primer (10 µM)	0.25 ~ 2.0 µl	
(Optional) 50x ROX dye	0.25 ~ 2.0 µl	
RNA template	×μl	
20x Enzyme Solution	1.0 μΙ	
Total reaction volume	20 μΙ	

1-2. Add the following components to a thin-walled PCR tube for AddGreen RT-qPCR Master:

Nuclease-Free Water	xμl
2x AddGreen RT-qPCR Master (2x conc.)	10.0 μl
Forward primer (10 μM)	0.25 ~ 2.0 μl
Reverse primer (10 µM)	0.25 ~ 2.0 μl
(Optional) 50x ROX dye	xμl
RNA template	xμl
Total reaction volume	20 µl

2. PCR cycling

z. PCR Cycling	
cDNA synthesis	50°C, 20 min
Initial denaturation	95°C, 10 min
PCR cycling	95°C, 15 sec
(30 ~ 40 cycles)	60°C, 1 min
Melting analysis	60°C → 90°C

[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 μ M 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