TransScript® Green One-Step qRT-PCR SuperMix

Please read the data sheet carefully prior to use.

Cat. No. AQ211

Storage: at -20°C away from light for two years

Description

TransScript® Green One-Step qRT-PCR SuperMix is designed for one-step qRT-PCR with high sensitivity, high synthesis efficiency and high amplification efficiency. This kit firstly synthesizes first-strand cDNA with RNA as templates using reverse gene-specific primers, and then performs qPCR with the synthesized cDNA as templates using both forward and reverse gene-specific primers to achieve one step from reverse transcription to qPCR in a single tube.

Highlights

• *TransScript*® Green One-Step RT/RI Enzyme Mix is used to efficiently synthesize first-strand cDNA from RNA and 2×*PerfectStart*™ Green One-Step qPCR SuperMix is used for qPCR, enabling simple procedures to minimize the chance of contamination.

• High sensitivity and high specificity to ensure accurate results.

Applications

• High-copy and low-copy gene detection

• Detection of RNA virus or trace amounts of RNA

Passive Reference Dye Compatibility with Different Instruments

• Passive Reference Dye I (50×)

ABI Prism 7000/7300/7700/7900, ABI Step One, ABI Step One Plus, ABI 7900HT, ABI 7900HT Fast

• Passive Reference Dye II (50×)

ABI Prism 7500, ABI Prism 7500 Fast, ABI QuantStudio Dx/3/5, ABI QuantStudio 6/7/12K Flex, ABI ViiA 7, Stratagene Mx3000P/Mx3005P/Mx4000

• No Passive Reference Dye

Roche LightCycler 480, Roche Light Cycler 96, MJ Research Chromo4, MJ Research Opticon 2, Takara TP-800, Bio-Rad iCycler iQ, Bio-Rad iCycler iQ5, Bio-Rad CFX96, Bio-Rad C1000 Thermal Cycler, Thermo Scientific Pikoreal 96, Qiagen Corbett Rotor-Gene 6000, Qiagen Corbett Rotor-Gene G, Qiagen Corbett Rotor-Gene Q, Qiagen Corbett Rotor-Gene 3000, Mastercycler ep realplex

Kit Contents

Component	AQ211-01	AQ211-02
TransScript® Green One-Step RT/RI Enzyme Mix	40 µl	160 µl
2×*PerfectStart*™ Green One-step qPCR SuperMix	1 ml	4×1 ml
Passive Reference Dye (50×)	40 µl	160 µl
RNase-free Water	1 ml	4×1 ml

Recommended qPCR Reaction Components and Conditions (20 µl reaction system)

Component	Volume	Final Concentration
RNA Template	1 pg~100 ng	as required
Forward Primer (10 µM)	0.4 µl	0.2 µM
Reverse primer (10 µM)	0.4 µl	0.2 µM
2×*PerfectStart*™ Green One-step qPCR SuperMix	10 µl	1×
TransScript® Green One-Step RT/RI Enzyme Mix	0.4 µl	-
Passive Reference Dye (50×) (optional)	0.4 µl	1×
RNase-free Water	Variable	-
Total volume	20 µl	-

qPCR (three-step)

45°C 5 min
94°C 30 sec
94°C 5 sec
50-60°C 15 sec* } 40-45 cycles
72°C 10 sec*
Dissociation Stage

qPCR (two-step)

45°C 5 min
94°C 30 sec
94°C 5 sec
60°C 30 sec* } 40-45 cycles
Dissociation Stage

For ABI qPCR instrument, we suggest using the following read time (Fluorescent signals can be collected during the annealing or extension stage for 3-step qPCR):

* For ABI Prism 7700/7900, set the read time to 30 seconds.
* For ABI Prism 7000/7300, set the read time to 31 seconds.
* For ABI Prism 7500, set the read time to 34 seconds.
* For ABI ViiA 7, set the read time to at least 19 seconds.

Two-step qPCR is more suitable for higher specificity assay.
Three-step qPCR is more suitable for higher amplification efficiency assay.

Notes

• Avoid RNase contamination.
• Use high-quality, intact RNA templates to ensure the success of qRT-PCR.
• Only gene-specific primers are compatible with this kit. Oligo(dT) or random primers cannot be used.

FOR RESEARCH USE ONLY