

Reverse transcription

Material

PrimeScript™RT reagent Kit with gDNA Eraser (TaKaRa Code No. RR047A)

Procedure

1. Genomic DNA elimination reaction

- 1) Prepare the genomic DNA elimination reaction solution on ice.
- 2) Add RNA template with the suitable amount of the master mix to a PCR tube.

Reagent	Amount
5X gDNA Eraser Buffer	2.0 µl
gDNA Eraser	1.0 µl
Total RNA	1.0 µl (Up to 1 µg of total RNA)
RNase Free dH ₂ O	Up to 10.0 µl

- 3) Run the program:
42°C 2 min

2. 4°C Reverse-transcription reaction

- 1) Prepare the reverse-transcription reaction solution on ice.
- 2) Add Reaction solution from Step 1 with the suitable amount of the master mix to a PCR tube.

Reagent	Amount
Reaction solution from Step 1	10.0 µl
5X PrimeScript Buffer 2 (for Real Time)	4.0 µl
RT Primer Mix	4.0 µl*
PrimeScript RT Enzyme Mix I	1.0 µl
RNase Free dH ₂ O	4.0 µl

- 3) Run the program:
37°C 15 min
85°C 5 sec
4°C