TOOLS Quick RT Kit

For rapid first-strand cDNA synthesis and removal of gDNA

Contents

Introduction

The TOOLS Quick RT Kit is designed for rapid first-strand cDNA synthesis with high efficiency and for rapid removal of genomic DNA (gDNA). The kit is designed for 2-step quantitative reverse transcription polymerase chain reaction (RT-qPCR). The 5× TOOLS RT SuperMix contains buffer, dNTPs, TOOLScript Reverse Transcriptase, and RNase inhibitor as well as random primer/Oligo(dT)23 VN primer mix. It takes only 15 min (50°C) to complete the synthesis of first-strand cDNA by using the innovative RT enzyme. This product also has a high affinity for RNA, which enables the efficient and sensitive reverse transcription of any template, such as GC-rich and complicated secondary structures of template RNA, leading to high yields of cDNA.

Important Notes

1. The 4× gDNA Eraser and 5× TOOLS RT SuperMix contain glycerol. Therefore, before pipetting, collect the liquid through brief centrifugation.
2. Use RNase-free water to dissolve RNA. Do not use TE because the EDTA in TE inhibits the reverse transcription reaction.
3. The cDNA product can be used for qPCR, and it is not suitable for long-fragment PCR and molecular cloning.

Kit Contents

Contents	TTC-RB23
RNase-Free ddH_2O	2× 1 mL
4x gDNA Eraser	400 µL
5× TOOLS RT SuperMix[a]	400 µL
5× Control Mix[b]	40 µL

a. This mix contains buffer, dNTPs, random primer/Oligo(dT)23 VN primer mix, TOOLScript Reverse Transcriptase, and RNase inhibitor.

b. This mix is same formulated as 5× TOOLS RT SuperMix, but without TOOLScript Reverse Transcriptase.

Storage

The TOOLS Quick RT Kit can be stored at −20°C for up to 12 months.

Protocol

1. Removal of gDNA

Mix the following components thoroughly in an RNase-free PCR tube and incubate at 42℃ for 2 min.

Table 1. gDNA removal reaction components

Solution	Volume
RNase-Free ddH_2O	To 16 µL
4× gDNA Eraser	4 µL
Template RNA	Total RNA: 1 pg–1 µg

2. Add 4 µL of 5× TOOLS RT SuperMix to the mixture of Step 1 (16 µL) and mix thoroughly.
3. (Optional) If checking the contamination of genomic DNA is required, scale up and prepare the extra RNA sample in Step 1. Add 4 µl of 5× control Mix to the mixture of Step 1 (16 µL) and mix thoroughly.
4. Perform RT reaction according to Table 2.

Table 2. RT reaction

Temperature	Time
50°C*	15 min
85°C	5 sec

Note: For templates with a complex secondary structure or high GC content, the temperature can be increased to 55ulabenefitting the yield.

The products can be used for PCR immediately or be stored at −20°C. It is recommended to store the products at −80℃ and prepare aliquots to avoid repeated freezing and thawing.