Version: 03/2024

ii. If the reverse transcription efficiency is low, or the secondary structure of RNA template is complex and GC content is high, the following steps are recommended:

1. Thaw the RNA templates, Primer Mix, dNTP Mix, SuperRT Buffer, SuperRT and RNase-free Water, and place on ice for later use.
2. Prepare the reaction system according to the following table, the total volume is 15 μL.

Reagent	20 μL System	Final Concentration
dNTP Mix，2.5 mM Each	4 μL	500 μM Each
Primer Mix	2 μL	
RNA Template	X μL	50 pg-5 μg
RNase-Free Water	to 15 μL	

Primer Mix is the combination of Oligo (dT) and Random Primer. According to the experimental needs, Oligo-dT Primer or Gene Specific Primer can be used.

3. Incubate at 70 ℃ for 10 minutes and take a quick ice bath for 2 minutes.
4. Centrifuge briefly to collect the solution from the wall to the bottom of the tube.
5. Add following reagents to the above reaction solution.

Reagent	20 μL System	Final Concentration
5×SuperRT Buffer	4 μL	1 ×
SuperRT, 200 U /μL	1 μL	

Note:1)If the initial amount of RNA is less than 50 ng, RNA enzyme inhibitor (RNasin) is recommended. It is not provided in this kit, if necessary, you can order it separately from our company. Cat.No.: CW0596.

6. Incubate at 42 ℃ for 30-50 minutes and at 85 ℃ for 5 minutes.
7. After the reaction, centrifuge briefly and cool on ice.
8. The reverse transcription products can be directly used for PCR reaction and fluorescence quantitative PCR reaction, or stored at -20 ℃ for long term storage.

SuperRT cDNA Synthesis Kit

Cat. No. : CW0741S (25 rxns)
CW0741M (100 rxns)

Storage Condition: -20℃

Components

Component	CW0741S 25 rxns	CW0741M 100 rxns
SuperRT, 200 U/μL	25 μL	100 μL
5×SuperRT Buffer	120 μL	500 μL
Primer Mix	60 μL	240 μL
dNTP Mix，2.5 mM Each	120 μL	500 μL
RNase-Free Water	1 mL	1 ml

Introduction

This product is a first-strand cDNA synthesis kit designed for the first step of two-step RT-PCR. The reverse transcriptase used in the kit is a novel high-efficiency reverse transcriptase derived from E. coli. The RNase H activity is eliminated to enhance the thermal stability, and this kit can be used with as little as pg-level of total RNA or mRNA. SuperRT reverse transcriptase has strong affinity with RNA. It can read through the RNA template with high GC content and complex secondary structure, and obtain high yield cDNA.

The kit is equipped with all reverse transcription reagents, including Super RT high efficiency reverse transcriptase, reaction buffer, primers, dNTP, etc., and is easy to use. It is suitable for the subsequent RT-PCR, qPCR, and DNA polymerase of various PCR reactions.

Features

1. Efficient reverse transcription efficiency: High affinity with RNA templates, reverse transcriptional efficiency of up to 90%, pg-level templates can be recognized.
2. Free to deal with complex templates: Even templates with high GC content and complex secondary structure can get good results without high temperature denaturation.

Features

1. RNase contamination should be avoided during operation to prevent RNA degradation or cross-contamination in experiments, and it is recommended that operators wear masks and disposable gloves, change gloves frequently, and use specialized instruments and consumables.
2. Disposable plasticware is preferred for experiments whenever possible. If glassware is used, it should be treated with 0.1% DEPC (diethyl pyrocarbonate) solution at 37 ℃ for 12 hours and autoclaved at 120 ℃ for 30 minutes, or dry heat sterilized at 180 ℃ for 60 minutes before use. The sterile water used in the experiment should be treated with 0.1% DEPC and then autoclaved.

3. Before use, please mix all reagents in this kit and gently to avoid foaming, and use after a short centrifugation. The enzymes involved should be put back to -20 ℃ as soon as possible after use to avoid repeated freeze-thaw.
4. If the initial amount of RNA is less than 50 ng, RNA enzyme inhibitor (RNasin) is recommended. It is not provided in this kit, if necessary, you can order it separately from our company. Cat.No.: CW0596.

Protocol

Note: 20 μL reaction system can for established for 1 ng-5 μg total RNA, if the total RNA amount is more than 5 μg, please scale up the reaction system.

i. Reverse transcription procedure:

1. Thaw the RNA templates, Primer Mix, dNTP Mix, SuperRT Buffer, SuperRT and RNase-free Water, and place on ice for later use.
2. Prepare the reaction system according to the following table, the total volume is 20 μL.

Reagent	20 μL System	Final Concentration
dNTP Mix，2.5 mM Each	4 μL	500 μM Each
Primer Mix	2 μL	
RNA Template	X μL	50 pg-5 μg
5×SuperRT Buffer	4 μL	1 ×
SuperRT, 200 U /μL	1 μL	
RNase-Free Water	to 20 μL	

Note: 1)If the initial amount of RNA is less than 50 ng, RNA enzyme inhibitor (RNasin) is recommended. It is not provided in this kit, if necessary, you can order it separately from our company. Cat.No.: CW0596.
2)Primer Mix is the combination of Oligo (dT) and Random Primer. According to the experimental needs, Oligo-dT Primer or Gene Specific Primer can be used. It is recommended to use Oligo-dT Primer 50 pmol, or Gene Specific Primer 2 pmol for 20 μl reaction system.

3. Vortex to mix, briefly centrifuge to collect the solution from the tube walls to the bottom of the tube.
4. Incubate at 42 ℃ for 30-50 minutes and at 85 ℃ for 5 minutes. After the reaction, centrifuge briefly and cool on ice.
5. The reverse transcription products can be directly used for PCR reaction and fluorescence quantitative PCR reaction, or stored at -20 ℃ for long term storage.