

Product Manual

Product Name	One Step RT-qPCR Kit
Catalog Number	CSB-DKT031
Physical Form	Liquid
Storage Conditions	-20 ±5°C, avoid repeated freeze-thaw cycles; can be stored at 2-8°C for frequent use
Transportation Conditions	≤0°C; transport on dry ice
Product Components	Buffer system, dNTPs, Taq DNA Polymerase, UDG, M-MLV reverse transcriptase, RNase Inhibitor
Quality Control	All components have been tested and found to be free of nucleases, endonucleases, and residual RNases.
Shelf Life	12 months

Product Description

The One Step RT-qPCR Kit is a reagent kit used for quantitative PCR reactions with RNA as a template. In the experimental process, reverse transcription and quantitative PCR are carried out in the same reaction tube, simplifying the experimental procedure. The kit utilizes heat-resistant M-MLV reverse transcriptase to efficiently synthesize the first strand cDNA, and HS Taq DNA Polymerase for quantitative amplification. It is suitable for specific detection systems using fluorescent probes (one-step amplification).

Product Components

Label	Components	Specifications		
		50T	100T	500T
1	2×One-step buffer Mix	0.75mL	1.5mL	7.5mL
2	One Step RT-qPCR Taq Mix	50μL	100μL	500μL

Operating instructions

1. Perform the operation on ice until the 2×Taq PCR Mix is completely dissolved. After thorough mixing, centrifuge the solution and collect it at the bottom of the tube.
2. Recommended reaction system for one-step amplification:

Components	Volume
ddH ₂ O	Up to 30 μL
2×One-step buffer Mix	15μL

Forward Primer (10 μ M)	0.6 μ L
Reverse Primer (10 μ M)	0.6 μ L
Probe(10 μ M)	0.3 μ L
One Step RT-qPCR Taq Mix	1 μ L
Template RNA	50pg-1 μ g

[Notes]:

a. Gently mix the reaction system. If necessary, collect all reagents at the bottom of the tube by centrifugation to avoid excessive bubble formation caused by vigorous shaking.

b. Primer concentration: Generally, a final concentration of 0.2 μ M for primers in the reaction system yields good results. If the reaction performance is poor, adjust the primer concentration within the range of 0.1 μ M-1.0 μ M.

c. Template concentration: As the copy numbers of target genes in templates from different species vary, gradient dilution can determine the optimal template usage amount.

3. Recommended PCR reaction program:

Temperature	Time	number of cycles
50-65 $^{\circ}$ C	5-15min	1
95 $^{\circ}$ C	3min	1
95 $^{\circ}$ C	10s	40-45
45-68 $^{\circ}$ C	30s*	

[Notes]:

d.This product exhibits good reverse transcription efficiency between 50-65 $^{\circ}$ C, with a minimum reverse transcription time of 5 minutes.

e.The annealing temperature needs to be adjusted based on the T_m value of the primers, generally set 3-5 $^{\circ}$ C lower than the T_m value of the primers. [*]Collect fluorescence.

Precautions:

1. Please use RNase-free consumables during the experiment.
2. For your safety and health, wear lab coat and disposable gloves during operation.