

PuroScript™ Universal One-Step RT-qPCR Kit

Product information

Product Description

The **PuroScript™ Universal One-Step RT-qPCR Kit** enables direct real-time quantitative analysis of target RNA sequences using intercalating fluorescent dye (SYBR Green-type) detection. RNA is first reverse transcribed into cDNA and then amplified by qPCR in the same tube, avoiding repeated tube opening and reducing the risk of cross-contamination between samples.

The PuroScript™ RT Enzyme Mix contains an engineered reverse transcriptase with significantly improved thermal stability for efficient reverse transcription of complex templates, together with an RNase inhibitor that protects RNA from degradation.

The 2X Universal Blue One-Step RT-qPCR Mix contains hot-start Taq DNA polymerase, dNTPs, Mg²⁺ and optimized buffer components, as well as a unique ROX passive reference dye that makes the kit compatible with a wide range of real-time PCR instruments. The mix also contains a blue tracer dye that makes it easy to confirm that the reaction has been set up; its spectrum does not overlap with qPCR fluorescent dyes and does not affect results.

Storage

Ship on wet ice. Store at -20 °C protected from light, valid for 12 months.

Safety Information:

Always wear appropriate lab coat, gloves and goggles. Please consult the Material Safety Data Sheet (MSDS) for more information.

Product Components:

Component	RS3345-100
PuroScript™ RT Enzyme Mix	100 µL
2X Universal Blue One-Step RT-qPCR Mix	1 mL
Nuclease-Free Water	1 mL

Product Description	Catalog No.
PuroScript™ Universal One-Step RT-qPCR Kit (100 reactions)	RS3345-100

PuroScript™ Universal One-Step RT-qPCR Kit

Product information

RT-qPCR reaction preparation (20 µL)

Component	Volume	Final Conc.
2X Universal Blue One-Step RT-qPCR Mix	10 µL	1X
PuroScript™ RT Enzyme Mix	1 µL	–
Forward Primer (10 µM) ^a	0.8 µL	0.4 µM
Reverse Primer (10 µM) ^a	0.8 µL	0.4 µM
Template RNA ^b	Variable	< 1 µg total RNA
Nuclease-Free Water	Add to 20 µL	–

Recommended RT-qPCR cycling conditions

Step	Temperature	Time	Cycles
Reverse Transcription	55 °C	10 min	1
Pre-denaturation	95 °C	1 min	1
Denaturation	95 °C	15 s	40
Annealing/Extension ^c	60 °C	1 min	
Melt Curve	Instrument default		1

a: Final primer concentration can range from 0.2 to 1.0 µM; 0.4 µM is recommended.

b: The optimal template amount depends on the copy number of the target; test a dilution series to find it. Recommended input is 0.1 ng–100 ng.

c: For a three-step protocol, use annealing at 55–65 °C for 20 s and extension at 72 °C for 30 s. To improve specificity, use the two-step protocol or raise the annealing temperature; to improve efficiency, use the three-step protocol or lengthen the extension.

Notes:

1. Gently invert the 2X Universal Blue One-Step RT-qPCR Mix to mix before use. Do not vortex, to avoid bubbles.
2. Keep all reagents on ice while preparing the reactions.
3. The mix contains fluorescent dye; avoid strong light while preparing reactions.
4. Use new disposable pipette tips when preparing and dispensing reactions to avoid cross-contamination between samples.
5. Use high-quality RNA templates. Repeated freeze-thaw cycles affect amplification; aliquot RNA that will be used in several experiments.
6. Compatible with common real-time PCR instruments, including Applied Biosystems (StepOne™, 7500, ViiA 7™, QuantStudio™), Bio-Rad (CFX96™, CFX384™), Roche LightCycler™, Qiagen Rotor-Gene®, Agilent/Stratagene Mx3000P® and others.