



EconoScript™ RT, RNase H–

Reverse Transcriptase | Long cDNA synthesis

Contents:

EconoScript reverse transcriptase is provided at a concentration of 200 U/μL with 10X EconoScript reaction buffer

Background

EconoScript reverse transcriptase is engineered to reduce RNase H activity and provide increased thermal stability. EconoScript can synthesize cDNA at a temperature optimum of 42° C, providing increased specificity, higher yields of cDNA, and more full-length product. Because EconoScript RT is not significantly inhibited by ribosomal and transfer RNA, it can be used to synthesize cDNA from total RNA.

Features of this enzyme

- Thermostability – between 40 - 50 °C with optimal activity at 42 °C
- Length of cDNA – can be used to synthesize first-strand cDNA up to 7 kb
- Applications – synthesis of first-strand cDNA, primer extension, sequencing dsDNA, cDNA libraries, RT-PCR, and RT LAMP

Application Notes

EconoScript RT can be used for first strand synthesis of complementary DNA (cDNA) from RNA or single-stranded DNA templates. It can be used with RT-qPCR assays, RT-LAMP, or cDNA library construction.

**These products are intended for research use only, not for diagnostic use. The safety and efficacy of these products in diagnostic or other clinical uses has not been established.*

Shipping & Storage

EconoScript RT is stored at -20 °C in 50% glycerol, 50 mM Tris-HCl, 50 mM KCl, 1 mM DTT, 1 mM EDTA, pH 7.5. *Can be supplied in a glycerol-free buffer as a custom order.*

EconoScript™ RT is shipped on dry or blue ice. On arrival store at -20 °C for optimum stability. Repeated freeze/thaw cycles should be avoided.

Quality Control

- EconoScript™ RT Unit activity: A known reverse transcriptase is used to create a standard curve with a reverse-transcription quantitative PCR assay against which the activity of this enzyme is measured.
- Purity: >95% as determined by SDS-PAGE analysis
- EconoScript™ RT is free of detectable RNase and DNase (exo- and endonuclease).
- <0.05 ng contaminating host DNA per 200 U.

1X RT Reaction Buffer

50 mM Tris-HCl
75 mM KCl
3 mM MgCl₂
10 mM DTT
pH 8.3 at 25 °C

General Protocol for First Strand Synthesis of cDNA

1) In RNase- and DNase-free PCR tubes mix:

Component	Volume
RNA (1 ng-5 µg)	n µL
10 mM dNTP mix	1 µL
50 µM Oligo(dT) ₁₂₋₁₈ or 60 µM gene-specific primer	2 µL
Nuclease-free water	To 10 µL

2) Incubate the RNA/primer mixture at 65 °C for 5 minutes, then place on ice or 4 °C

3) Then add the following components

Component	Volume (1X)
10X EconoScript™ Reaction Buffer	2 µL
EconoScript™ (200 U/µL)	1 µL
RNase Inhibitor (40 U/µL)	0.2 µL
Nuclease-Free water	6.8 µL

1) Incubate the reaction mix at 42 °C for 60 minutes.

2) Terminate reaction by incubating at 70 °C for 15 minutes.

3) Cool reaction on ice or at 4 °C.

4) Collect reaction via centrifugation.

5) cDNA can be stored at -20 °C or used immediately for PCR. The cDNA product should not exceed 1/10th of the PCR reaction volume.