

Catalog No.: 7000

Expressed in: E. coli

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 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 and 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	Το 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:

Volume (1X)
2 μL
1 μL
0.2 μL
6.8 μL

- 1. Incubate the reaction mix at 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 $1/10^{th}$ of the PCR reaction volume.