NEW ENGLAND BioLabs® Inc.

240 County Road
Ipswich, MA 01938-2723

Tel 978-927-5054
Fax 978-921-1350

www.neb.com
info@neb.com

New England Biolabs
Product Specification

Product Name:	*WarmStart® RTx Reverse Transcriptase (Glycerol-free)*
Catalog #:	*M0439L*
Concentration:	*75,000 units/ml*
Unit Definition:	*One unit is defined as the amount of enzyme that will incorporate 1 nmol of dTTP into acid-insoluble material in 20 minutes at 50°C.*
Shelf Life:	*24 months*
Storage Temp:	*-80°C*
Storage Conditions:	*10 mM Tris-HCl, 100 mM KCl, 1 mM DTT, 0.1 mM EDTA, (pH 7.4 @ 25°C)*
Specification Version:	*PS-M0439L v1.0*
Effective Date:	*03 Jan 2024*

Assay Name/Specification (minimum release criteria)

Endonuclease Activity (Nicking) - A 50 µl reaction in Isothermal Amplification Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 15 units of RTx Reverse Transcriptase (Glycerol-free) incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.

Exonuclease Activity (Radioactivity Release) - A 50 µl reaction in Isothermal Amplification Buffer containing 1 µg of a mixture of single and double-stranded [^{3}H] *E. coli* DNA and a minimum of 15 units of RTx Reverse Transcriptase (Glycerol-free) incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.

Functional Testing (RT-LAMP) - A 25 µl RT-LAMP reaction with 7.5 units of WarmStart® RTx Reverse Transcriptase (Glycerol-free), 10 ng of genomic RNA and 1X LAMP fluorescent dye results in a threshold time of ≤ 20 minutes as determined by fluorescent detection.

Non-Specific DNase Activity (16 Hour) - A 50 µl reaction in NEBuffer 2 containing 1 µg of T3 or T7 DNA in addition to a reaction containing Lambda-HindIII DNA and a minimum of 15 units of WarmStart® RTx Reverse Transcriptase (Glycerol-free) incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.

Protein Purity Assay (SDS-PAGE) - RTx Reverse Transcriptase (Glycerol-free) is ≥ 99% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.

qPCR DNA Contamination (*E. coli* Genomic) - A minimum of 15 units of RTx Reverse Transcriptase (Glycerol-free) is screened for the presence of *E. coli* genomic DNA using SYBR® Green qPCR with primers specific for the *E. coli* 16S rRNA locus. Results are quantified using a standard curve generated from purified *E. coli* genomic DNA. The measured level of *E. coli* genomic DNA contamination is ≤ 1 *E. coli* genome.

240 County Road
Ipswich, MA 01938-2723

Tel 978-927-5054
Fax 978-921-1350

www.neb.com
info@neb.com

New England Biolabs
Product Specification

Assay Name/Specification (minimum release criteria)
RNase Activity (Extended Digestion) - A 10 µl reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 15 units of WarmStart® RTx Reverse Transcriptase (Glycerol-free) is incubated at 37°C. After incubation for 4 hours, >90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.

Date 03 Jan 2024

Lauren Brown
Quality Approver