

## New England Biolabs Certificate of Analysis

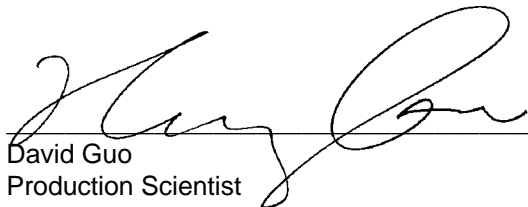
**Product Name:** WarmStart® RTx Reverse Transcriptase  
**Catalog Number:** M0380L  
**Concentration:** 15,000 U/ml  
**Unit Definition:** One unit is defined as the amount of enzyme that will incorporate 1 nmol of dTTP into acid-insoluble material in 20 minutes at 50°C.  
**Packaging Lot Number:** 10061431  
**Expiration Date:** 07/2021  
**Storage Temperature:** -20°C  
**Storage Conditions:** 10 mM Tris-HCl , 100 mM KCl , 1 mM DTT , 0.1 mM EDTA , 50 % Glycerol, (pH 7.4 @ 25°C)  
**Specification Version:** PS-M0380S/L v2.0

| WarmStart® RTx Reverse Transcriptase Component List |   |            |                      |
|---|---|------------|----------------------|
| NEB Part Number                                     | Component Description                           | Lot Number | Individual QC Result |
| M0380LVIAL  | WarmStart® RTx Reverse Transcriptase            | 10047020   | Pass                 |
| B1003SVIAL  | Magnesium Sulfate (MgSO <sub>4</sub> ) Solution | 10042724   | Pass                 |
| B0537SVIAL  | Isothermal Amplification Buffer                 | 10035085   | Pass                 |

| Assay Name/Specification  | Lot # 10061431 |
|---|----------------|
| <p><b>Endonuclease Activity (Nicking)</b><br/>           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 incubated for 4 hours at 37°C results in &lt;10% conversion to the nicked form as determined by agarose gel electrophoresis.</p>   | Pass           |
| <p><b>Exonuclease Activity (Radioactivity Release)</b><br/>           A 50 µl reaction in Isothermal Amplification Buffer containing 1 µg of a mixture of single and double-stranded [<sup>3</sup>H] E. coli DNA and a minimum of 15 units of RTx Reverse Transcriptase incubated for 4 hours at 37°C releases &lt;0.1% of the total radioactivity.</p>   | Pass           |
| <p><b>Non-Specific DNase Activity (16 Hour)</b><br/>           A 50 µl reaction in NEBuffer 2 containing 1 µg of T3 DNA in addition to a reaction containing Lambda-HindIII DNA and a minimum of 15 units of WarmStart® RTx Reverse Transcriptase incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.</p> | Pass           |

| Assay Name/Specification  | Lot # 10061431 |
|---|----------------|
| <p><b>Protein Purity Assay (SDS-PAGE)</b><br/>RTx Reverse Transcriptase is <math>\geq 99\%</math> pure as determined by SDS-PAGE analysis using Coomassie Blue detection.</p>   | <b>Pass</b>    |
| <p><b>qPCR DNA Contamination (E. coli Genomic)</b><br/>A minimum of 15 units of WarmStart<sup>®</sup> RTx Reverse Transcriptase is screened for the presence of E. coli genomic DNA using SYBR<sup>®</sup> 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 <math>\leq 1</math> E. coli genome.</p> | <b>Pass</b>    |
| <p><b>RNase Activity Assay (4 Hour Digestion)</b><br/>A 10 <math>\mu</math>l reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 1 <math>\mu</math>l of WarmStart<sup>®</sup> RTx Reverse Transcriptase is incubated at 37°C. After incubation for 4 hours, &gt;90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.</p>  | <b>Pass</b>    |

This product has been tested and shown to be in compliance with all specifications.



David Guo  
Production Scientist  
22 Jul 2019



Michael Tonello  
Packaging Quality Control Inspector  
11 Dec 2019