240 County Road Ipswich, MA 01938-2723 Tel 978-927-5054 Fax 978-921-1350 www.neb.com info@neb.com

New England Biolabs Certificate of Analysis

Product Name: ProtoScript® II Reverse Transcriptase

Catalog #: M0368S/L/X
Concentration: 200,000 units/ml

Unit Definition: One unit is defined as the amount of enzyme that will incorporate 1 nmol of dTTP into an acid-insoluble form in 10 minutes at

37°*C*.

 Lot #:
 0041507

 Assay Date:
 07/2015

 Expiration Date:
 07/2017

 Storage Temp:
 -20°C

Storage Conditions: 20 mM Tris-HCl, 100 mM NaCl, 1 mM DTT, 0.1 mM EDTA, 0.01 % IGEPAL® CA-630, 50 % Glycerol, (pH 7.5 @

25°C)

Specification Version: PS-M0368S/L/X v1.0

Effective Date: 22 Sep 2016

| Assay Name/Specification (minimum release criteria) | Lot #0041507 |
|--|--------------|
| Endonuclease Activity (Nicking) - A 50 μ l reaction in ProtoScript® II Reverse Transcriptase Reaction Buffer containing 1 μ g of supercoiled PhiX174 DNA and a minimum of 200 units of ProtoScript® II Reverse Transcriptase incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis. | Pass |
| Exonuclease Activity (Radioactivity Release) - A 50 μl reaction in ProtoScript® II Reverse Transcriptase Reaction Buffer containing 1 μg of a mixture of single and double-stranded [³ H] <i>E. coli</i> DNA and a minimum of 200 units of ProtoScript® II Reverse Transcriptase incubated for 4 hours at 37°C releases <0.1% of the total radioactivity. | Pass |
| Non-Specific DNase Activity (16 Hour) - 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 200 units of ProtoScript® II 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. | Pass |
| Protein Purity Assay (SDS-PAGE) - ProtoScript® II Reverse Transcriptase is ≥ 99% pure as determined by SDS-PAGE analysis using Coomassie Blue detection. | Pass |







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| qPCR DNA Contamination (<i>E. coli</i> Genomic) - A minimum of 200 units of ProtoScript® II Reverse Transcriptase is screened for the presence of <i>E. coli</i> genomic DNA using SYBR® Green qPCR with primers specific for the <i>E. coli</i> 16S rRNA locus. Results are quantified using a standard curve generated from purified <i>E. coli</i> genomic DNA. The measured level of <i>E. coli</i> genomic DNA contamination is ≤ 1 <i>E. coli</i> genome. | Pass |
| RNAse Activity Assay (4 Hour Digestion) - A 10 µl reaction in ProtoScript® II Reverse Transcriptase Reaction Buffer containing 40 ng of a 300 base single-stranded RNA and a minimum of 1 µl of ProtoScript® II Reverse Transcriptase 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. | Pass |

Authorized by Denisa Gilaj 22 Sep 2016

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Inspected by Denisa Gilaj 13 Jul 2015