

## New England Biolabs Product Specification

*Product Name:* LunaScript™ RT SuperMix  
*Catalog #:* M3010S/L  
*Concentration:* 5X Concentrate  
*Shelf Life:* 24 months  
*Storage Temp:* -20°C  
*Composition (1X):* Proprietary  
*Specification Version:* PS-M3010S/L v1.0  
*Effective Date:* 14 Feb 2018

### Assay Name/Specification (minimum release criteria)

**Endonuclease Activity (Nicking)** - A 50 µl reaction in ThermoPol® Reaction Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 100 units of Luna® Reverse Transcriptase incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.

**Phosphatase Activity (pNPP)** - A 200 µl reaction in 1M Diethanolamine, pH 9.8, 0.5 mM MgCl<sub>2</sub> containing 2.5 mM *p*-Nitrophenyl Phosphate (pNPP) and a minimum of 100 units of Luna® Reverse Transcriptase incubated for 4 hours at 37°C yields <0.0001 unit of alkaline phosphatase activity as determined by spectrophotometric analysis.

**Protein Purity Assay (SDS-PAGE)** - Luna® Reverse Transcriptase is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.

**qPCR DNA Contamination (E. coli Genomic)** - A minimum of 1 µl of LunaScript™ RT SuperMix 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.

**RNase Activity Assay (4 Hour Digestion)** - A 10 µl reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 800 units of Luna® 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.

**Single Stranded DNase Activity (FAM-Labeled Oligo)** - A 50 µl reaction in CutSmart® Buffer containing a 20 nM solution of a fluorescent internal labeled oligonucleotide and a minimum of 800 units of Luna® Reverse Transcriptase incubated for 16 hours at 37°C yields <5% degradation as determined by capillary electrophoresis.



Date 14 Feb 2018

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Director of Quality Control

