

## New England Biolabs Product Specification

*Product Name:* NEBNext<sup>®</sup> Q5U<sup>™</sup> Master Mix  
*Catalog #:* M0597S/L  
*Concentration:* 2X Concentrate  
*Shelf Life:* 12 months  
*Storage Temp:* -20°C  
*Composition (1X):* Proprietary  
*Specification Version:* PS-M0597S/L v1.0  
*Effective Date:* 15 Mar 2019

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

**Non-Specific DNase Activity (16 hour, Buffer)** - A 50 µl reaction in 1X NEBNext<sup>®</sup> Q5U<sup>™</sup> Master Mix containing 1 µg of T3 or T7 DNA in addition to a reaction containing Lambda-HindIII DNA incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.

**PCR Amplification (dU Bypass)** - A 25 µl reaction in 1X NEBNext<sup>®</sup> Q5U<sup>™</sup> Master Mix with 10 ng of genomic DNA and 0.5 µM primers containing dU residues for 30 cycles of PCR results in the expected 720 bp product.

**qPCR DNA Contamination (E. coli Genomic)** - A minimum of 1 µl of NEBNext<sup>®</sup> Q5U<sup>™</sup> Master Mix 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 ≤ 1 *E. coli* genome.

**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 1 µl of NEBNext<sup>®</sup> Q5U<sup>™</sup> Master Mix is incubated at 37°C. After incubation for 16 hours, >90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.



Date 15 Mar 2019

Derek Robinson  
Director of Quality Control

