

New England Biolabs Product Specification

Product Name:	<i>T4 RNA Ligase 1 (ssRNA Ligase), High Concentration</i>
Catalog #:	<i>M0437M</i>
Concentration:	<i>30,000 units/ml</i>
Unit Definition:	<i>One unit is defined as the amount of enzyme required to convert 1 nanomole of 5'-[³²P] rA16 into a phosphatase-resistant form in 30 minutes at 37°C.</i>
Shelf Life:	<i>24 months</i>
Storage Temp:	<i>-20°C</i>
Storage Conditions:	<i>10 mM Tris-HCl, 50 mM KCl, 1 mM DTT, 0.1 mM EDTA, 50 % Glycerol, (pH 7.4 @ 25°C)</i>
Specification Version:	<i>PS-M0437M v1.0</i>
Effective Date:	<i>05 Feb 2018</i>

Assay Name/Specification (minimum release criteria)

Endonuclease Activity (Nicking) - A 50 µl reaction in T4 RNA Ligase 1 Reaction Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 30 units of T4 RNA Ligase 1 (ssRNA Ligase), High Concentration 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 T4 RNA Ligase 1 Reaction Buffer containing 1 µg of a mixture of single and double-stranded [³H] *E. coli* DNA and a minimum of 30 units of T4 RNA Ligase 1 (ssRNA Ligase), High Concentration incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.

Protein Purity Assay (SDS-PAGE) - T4 RNA Ligase 1 (ssRNA Ligase), High Concentration is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.

qPCR DNA Contamination (E. coli Genomic) - A minimum of 10 units of T4 RNA Ligase 1 (ssRNA Ligase), High Concentration 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 (Extended Digestion) - A 10 µl reaction in NEBuffer 4 containing 40 ng of RNA transcript and a minimum of 30 units of T4 RNA Ligase 1 (ssRNA Ligase), High Concentration is incubated at 37°C. After incubation for 16 hours, >90% of the substrate RNA remains intact as determined by gel electrophoresis using gel electrophoresis.



Date 05 Feb 2018

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Director of Quality Control

