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: T4 RNA Ligase 2, truncated

Catalog #: M0242S/L
Concentration: 200,000 units/ml

Unit Definition: 200 units is defined as the amount of enzyme required to give 80% ligation of a 31-mer RNA to the pre-adenylated end of a 17-

mer DNA in a total reaction volume of 20  $\mu$ l in 1 hour at 25°C.

 Lot #:
 0091704

 Assay Date:
 04/2017

 Expiration Date:
 4/2019

 Storage Temp:
 -20°C

Storage Conditions: 10 mM Tris-HCl, 100 mM NaCl, 0.1 mM DTT, 0.1 mM EDTA, 50 % Glycerol, (pH 7.5 @) 25 °C)

Specification Version: PS-M0242S/L v1.0
Effective Date: 12 Feb 2018

Assay Name/Specification (minimum release criteria)	Lot #0091704
<b>Endonuclease Activity (Nicking)</b> - A 50 μl reaction in T4 RNA Ligase Reaction Buffer containing 1 μg of supercoiled PhiX174 DNA and a minimum of 200 units of T4 RNA Ligase 2, truncated incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	Pass
<b>Exonuclease Activity (Radioactivity Release)</b> - A 50 $\mu$ l reaction in T4 RNA Ligase Reaction Buffer containing 1 $\mu$ g of a mixture of single and double-stranded [ $^3$ H] <i>E. coli</i> DNA and a minimum of 200 units of T4 RNA Ligase 2, truncated incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
Phosphatase Activity (pNPP) - A 200 $\mu$ l 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 200 units of T4 RNA Ligase 2, truncated incubated for 4 hours at 37°C yields <0.0001 unit of alkaline phosphatase activity as determined by spectrophotometric analysis.	Pass
<b>Protein Purity Assay (SDS-PAGE)</b> - T4 RNA Ligase 2, truncated is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
RNase Activity (Extended Digestion) - A 10 $\mu$ l reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 200 units of T4 RNA Ligase 2, truncated is incubated at 37°C. After incubation for 16 hours, >90% of the substrate RNA remains intact as determined by gel electrophoresis using polyacrylamide gel electrophoresis.	Pass

Authorized by Derek Robinson 12 Feb 2018







Inspected by Priscilla Hong 11 Jul 2017