

## New England Biolabs Certificate of Analysis

**Product Name:** T4 RNA Ligase 2  
**Catalog Number:** M0239L  
**Concentration:** 10,000 U/ml  
**Unit Definition:** One unit is defined as the amount of enzyme required to ligate 0.4 µg of an equimolar mix of a 23-mer and 17-mer RNAs in a total reaction volume of 20 µl in 30 minutes at 37°C.  
**Packaging Lot Number:** 10158106  
**Expiration Date:** 06/2024  
**Storage Temperature:** -20°C  
**Storage Conditions:** 10 mM Tris-HCl, 50 mM KCl, 0.1 mM DTT, 0.1 mM EDTA, 35 mM (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>, 50 % Glycerol, (pH 7.5 @ 25°C)  
**Specification Version:** PS-M0239S/L v1.0

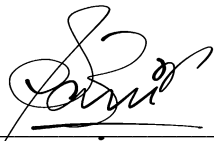
| T4 RNA Ligase 2 Component List |                                 |            |                      |
|--------------------------------|---------------------------------|------------|----------------------|
| NEB Part Number                | Component Description           | Lot Number | Individual QC Result |
| M0239LVIAL                     | T4 RNA Ligase 2 (dsRNA Ligase)  | 10153955   | Pass                 |
| B0239SVIAL                     | T4 RNA Ligase 2 Reaction Buffer | 10156729   | Pass                 |

| Assay Name/Specification  | Lot # 10158106 |
|---|----------------|
| <b>Protein Purity Assay (SDS-PAGE)</b><br>T4 RNA Ligase 2 (dsRNA Ligase) is ≥ 99% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.   | <b>Pass</b>    |
| <b>Phosphatase Activity (pNPP)</b><br>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 T4 RNA Ligase 2 (dsRNA Ligase) incubated for 4 hours at 37°C yields <0.00001 unit of alkaline phosphatase activity as determined by spectrophotometric analysis.                      | <b>Pass</b>    |
| <b>RNase Activity (Extended Digestion)</b><br>A 10 µl reaction in T4 RNA Ligase Reaction Buffer containing 40 ng of a 300 base single-stranded RNA and a minimum of 10 units of T4 RNA Ligase 2 (dsRNA Ligase) is incubated at 37°C. After incubation for 4 hours, >90% of the substrate RNA remains intact as determined by gel electrophoresis using agarose gel electrophoresis. | <b>Pass</b>    |
| <b>Exonuclease Activity (Radioactivity Release)</b><br>A 50 µl reaction in T4 RNA Ligase Reaction Buffer containing 1 µg of a mixture of  | <b>Pass</b>    |

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|---|--------------------|
| <p>single and double-stranded [<sup>3</sup>H] E. coli DNA and a minimum of 100 units of T4 RNA Ligase 2 (dsRNA Ligase) incubated for 4 hours at 37°C releases &lt;0.1% of the total radioactivity.</p> <p><b>Endonuclease Activity (Nicking)</b><br/>A 50 µl reaction in T4 RNA Ligase Reaction Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 100 units of T4 RNA Ligase 2 (dsRNA Ligase) incubated for 4 hours at 37°C results in &lt;10% conversion to the nicked form as determined by agarose gel electrophoresis.</p> | <p><b>Pass</b></p> |

This product has been tested and shown to be in compliance with all specifications.

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