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New England Biolabs
Certificate of Analysis

Product Name:	*T4 RNA Ligase 2 (dsRNA Ligase)*
Catalog #:	*M0239S/L*
Concentration:	*10,000 units/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.*
Lot #:	*0041802*
Assay Date:	*02/2018*
Expiration Date:	*2/2020*
Storage Temp:	*-20°C*
Storage Conditions:	*10 mM Tris-HCl, 50 mM KCl, 0.1 mM DTT, 0.1 mM EDTA, 35 mM $(NH_4)_2SO_4$, 50 % Glycerol, (pH 7.5 @ 25°C)*
Specification Version:	*PS-M0239S/L v1.0*
Effective Date:	*13 Feb 2018*

Assay Name/Specification (minimum release criteria)	**Lot #0041802**
Endonuclease Activity (Nicking) - 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 <10% conversion to the nicked form as determined by agarose gel electrophoresis.	**Pass**
Exonuclease Activity (Radioactivity Release) - A 50 µl reaction in T4 RNA Ligase Reaction Buffer containing 1 µg of a mixture of single and double-stranded [^{3}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 <0.1% of the total radioactivity.	**Pass**
Phosphatase Activity (pNPP) - A 200 µl reaction in 1M Diethanolamine, pH 9.8, 0.5 mM $MgCl_2$ 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.	**Pass**
Protein Purity Assay (SDS-PAGE) - T4 RNA Ligase 2 (dsRNA Ligase) is ≥ 99% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	**Pass**
RNase Activity (Extended Digestion) - 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.	**Pass**

Authorized by
Derek Robinson
13 Feb 2018

Inspected by
Bo Wu
13 Feb 2018

ISO 9001 Registered Quality Management
ISO 14001 Registered Environmental Management
ISO 13485 Registered Medical Devices