NEW ENGLAND BioLabs Inc.
be INSPIRED drive DISCOVERY stay GENUINE
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:	*Thermostable 5' App DNA/RNA Ligase*
Catalog Number:	*M0319S*
Concentration:	*20 µM*
Packaging Lot Number:	*10116069*
Expiration Date:	*05/2023*
Storage Temperature:	*-20°C*
Storage Conditions:	*10 mM Tris-HCl , 50 mM KCl , 1 mM DTT , 0.1 mM EDTA , 50 % Glycerol, (pH 7.4 @ 25°C)*
Specification Version:	*PS-M0319S/L v1.0*

Thermostable 5' App DNA/RNA Ligase Component List

NEB Part Number	Component Description	Lot Number	Individual QC Result
M0319SVIAL	Thermostable 5' App DNA/RNA Ligase	10116068	**Pass**
B7001SVIAL	NEBuffer™ 1	10111607	**Pass**
B0787AVIAL	$MnCl_2$	10117459	**Pass**

Assay Name/Specification	Lot # 10116069
Functional Testing (Targeted Ligation) A 20 µl reaction in 1X NEBuffer 1 containing 20 pmol of 30 bp FAM-labeled single-stranded RNA, 200 pmol 17 bp 5' pre-adenylated single-stranded DNA linker, and 40 pmol Thermostable 5' App DNA/RNA Ligase incubated for 1 hour at 65°C results in ≥80% ligation of the substrate RNA as determined by capillary electrophoresis.	**Pass**
Phosphatase Activity (pNPP) A 200 µl reaction in 1M Diethanolamine, pH 9.8, 0.5 mM MgCl2 containing 2.5 mM p-Nitrophenyl Phosphate (pNPP) and a minimum of 100 pmol of Thermostable 5' App DNA/RNA Ligase incubated for 16 hours at 37°C yields <0.0001 unit of alkaline phosphatase activity as determined by spectrophotometric analysis.	**Pass**
Exonuclease Activity (Radioactivity Release) A 50 µl reaction in NEBuffer 1 containing 1 µg of a mixture of single and double-stranded [^3H] E. coli DNA and a minimum of 100 pmol of Thermostable 5' App DNA/RNA Ligase incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	**Pass**
RNase Activity (Extended Digestion) A 10 µl reaction in NEBuffer 1 containing 40 ng of a 300 base single-stranded RNA	**Pass**

NEW ENGLAND BioLabs Inc.
be INSPIRED drive DISCOVERY stay GENUINE
240 County Road
Ipswich, MA 01938-2723
Tel 978-927-5054
Fax 978-921-1350
www.neb.com
info@neb.com

Assay Name/Specification	Lot # 10116069
and a minimum of 100 pmol of Thermostable 5' App DNA/RNA Ligase 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.	
Endonuclease Activity (Nicking) A 50 µl reaction in NEBuffer 1 containing 1 µg of supercoiled PhiX174 DNA and a minimum of 100 pmol of Thermostable 5' App DNA/RNA Ligase incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	**Pass**

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

One or more products referenced in this document may be covered by a 3rd-party trademark. Please visit www.neb.com/trademarks for additional information.

Bhairavi Jani
Production Scientist
18 Oct 2021

Michael Tonello
Packaging Quality Control Inspector
18 Oct 2021

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