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New England Biolabs Certificate of Analysis

Product Name: Thermostable 5' App DNA/RNA Ligase

Catalog Number: M0319S
Concentration: 20 µM
Lot Number: 10043714
Expiration Date: 04/2021
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	10043715	Pass	
B7001SVIAL	NEBuffer™ 1	10041625	Pass	
B0787AVIAL	MnCl ₂	10017992	Pass	

Assay Name/Specification	Lot # 10043714
RNase Activity (Extended Digestion) A 10 µl reaction in NEBuffer 1 containing 40 ng of a 300 base single-stranded RNA 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.	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
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
Phosphatase Activity (pNPP) A 200 µl reaction in 1M Diethanolamine, pH 9.8, 0.5 mM MgCl2 containing 2.5 mM	Pass



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Assay Name/Specification	Lot # 10043714
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.	
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

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

Bo Wu

Production Scientist

22 Aug 2018

Michael Tonello

Packaging Quality Control Inspector

11 Jul 2019



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