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New England Biolabs Certificate of Analysis

Product Name:RtcB LigaseCatalog Number:M0458SConcentration:15 μMPackaging Lot Number:10201913Expiration Date:02/2025Storage Temperature:-20°C

Storage Conditions: 50 mM NaCl, 10 mM Tris-HCl, 1 mM DTT, 0.1 mM EDTA, 50 % Glycerol,

(pH 7.4 @ 25°C)

Specification Version: PS-M0458S v1.0

RtcB Ligase Component List				
NEB Part Number	Component Description	Lot Number	Individual QC Result	
N2080AVIAL	GTP	10176039	Pass	
M0458SVIAL	RtcB Ligase	10200642	Pass	
B1761SVIAL	10mM MnCl2	10167824	Pass	
B0790AVIAL	10X RtcB Reaction Buffer	10176038	Pass	

Assay Name/Specification	Lot # 10201913
Endonuclease Activity (Nicking) A 50 µl reaction in RtcB Reaction Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 15 pmol of RtcB 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 RtcB Reaction Buffer containing 1 µg of a mixture of single and double-stranded [³H] E. coli DNA and a minimum of 15 pmol of RtcB Ligase incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
Functional Testing (RtcB Ligase) A 20 μl reaction in 1X RtcB Reaction Buffer supplemented with 0.1 mM GTP and 1 mM MnCl2 containing 0.5 μM of a 17 mer 5' FAM-labeled 3' phosphorylated RNA, 0.5 μM of a 30 mer 5' OH RNA and 0.75 μM RtcB Ligase incubated for 1 hour at 37°C results in ≥ 80% ligation as determined by capillary electrophoresis.	Pass
Protein Purity Assay (SDS-PAGE) RtcB Ligase is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass



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Assay Name/Specification	Lot # 10201913
RNase Activity (Extended Digestion) A 10 µl reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 15 pmol of RtcB 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

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

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Jessica Cane Production Scientist

01 Mar 2023

Josh Hersey

Packaging Quality Control Inspector

27 Jul 2023



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