

New England Biolabs Certificate of Analysis

Product Name: *RtcB Ligase*
Catalog Number: *M0458S*
Concentration: *15 µM*
Unit Definition:
Lot Number: *10013752*
Expiration Date: *06/2020*
Storage 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	10012670	Pass
M0458SVIAL	RtcB Ligase	10009681	Pass
B1761SVIAL	10mM MnCl ₂	10009388	Pass
B0790AVIAL	10X RtcB Reaction Buffer	10012675	Pass

Assay Name/Specification	Lot # 10013752
Protein Purity Assay (SDS-PAGE) RtcB Ligase is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
Functional Testing (RtcB Ligase) A 20 µl reaction in 1X RtcB Reaction Buffer supplemented with 0.1 mM GTP and 1 mM MnCl ₂ 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
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
Endonuclease Activity (Nicking) A 50 µl reaction in RtcB Reaction Buffer containing 1 µg of supercoiled PhiX174 DNA	Pass

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<p>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.</p> <p>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.</p>	<p>Pass</p>

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



Alicia Bielik
Production Scientist
18 Jun 2018



Michael Tonello
Packaging Quality Control Inspector
20 Jun 2018