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## New England Biolabs Certificate of Analysis

Product Name: Quick Ligation™ Kit

Catalog Number: M2200S
Unit Definition: N/A

Lot Number: 10018036
Expiration Date: 06/2020
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-M2200S/L v1.0

Quick Ligation™ Kit Component List				
<b>NEB Part Number</b>	Component Description	Lot Number	Individual QC Result	
M2200SVIAL	Quick Ligation™ Kit	10009400	Pass	
B2200SVIAL	Quick Ligation™ Reaction Buffer	0011801	Pass	

Assay Name/Specification	Lot # 10018036
<b>qPCR DNA Contamination (E. coli Genomic)</b> A minimum of 2000 units of Quick Ligase is screened for the presence of E. coli genomic DNA using SYBR® Green qPCR with primers specific for the E. coli 16S rRNA locus. Results are quantified using a standard curve generated from purified E. coli genomic DNA. The measured level of E. coli genomic DNA contamination is ≤ 1 E. coli genome.	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 1 µl of Quick 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
Single Stranded DNase Activity (FAM-Labeled Oligo) A 50 µl reaction in CutSmart® Buffer containing a 20 nM solution of a fluorescent internal labeled oligonucleotide and a minimum of 10,000 units of Quick Ligase incubated for 16 hours at 37°C yields <5% degradation as determined by capillary electrophoresis.	Pass
<b>Ligation and Recutting (Terminal Integrity, Digested DNA)</b> A 20 μl reaction in 1X T4 DNA Ligase Reaction Buffer containing 2 μg of Lambda	Pass



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Assay Name/Specification	Lot # 10018036
DNA-HindIII Digest and a minimum of 4000 units of Quick Ligase incubated for 16 hours at 37°C results in >95% ligation of the DNA fragments as determined by agarose gel electrophoresis. Of these ligated fragments, >95% can be recut with HindIII.	
Non-Specific DNase Activity (16 Hour) A 50 $\mu$ I reaction in NEBuffer 1 containing 1 $\mu$ g of CIP-treated Lambda-HindIII DNA and a minimum of 2000 units of Quick Ligase incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	Pass
Protein Purity Assay (SDS-PAGE)  Quick Ligase is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
Functional Testing (Ligation and Transformation) After a five-minute ligation of linearized, dephosphorylated LITMUS 28 or pUC19 (containing either blunt [EcoRV] or cohesive [HindIII] ends) and a mixture of compatible insert fragments, transformation into chemically competent E. coli DH-5 alpha cells yields a minimum of 1 x 10e6 recombinant transformants per μg plasmid DNA.	Pass
DNase Activity (Labeled Oligo, 3' extension) A 50 µl reaction in CutSmart® Buffer containing a 20 nM solution of a fluorescent labeled double-stranded oligonucleotide containing a 3' extension and a minimum of 10,000 units of Quick Ligase incubated for 16 hours at 37°C yields <5% degradation as determined by capillary electrophoresis.	Pass
DNase Activity (Labeled Oligo, 5' extension) A 50 µl reaction in CutSmart® Buffer containing a 20 nM solution of a fluorescent labeled double-stranded oligonucleotide containing a 5' extension and a minimum of 10,000 units of Quick Ligase incubated for 16 hours at 37°C yields <5% degradation as determined by capillary electrophoresis.	Pass
Double Stranded DNase Activity (Labeled Oligo) A 50 μl reaction in CutSmart® Buffer containing a 20 nM solution of a fluorescent labeled double-stranded oligonucleotide containing a blunt end and a minimum of 10,000 units of Quick Ligase incubated for 16 hours at 37°C yields <5% degradation as determined by capillary electrophoresis.	Pass
Endonuclease Activity (Nicking) A 50 μl reaction in NEBuffer 1 containing 1 μg of supercoiled PhiX174 DNA and a minimum of 2000 units of Quick Ligase incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	Pass



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Assay Name/Specification	Lot # 10018036
Exonuclease Activity (Radioactivity Release)	Pass
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 2000 units of Quick Ligase	
incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	

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

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Mary Lorenzen Production Scientist

29 Jun 2018

Michael Tonello

Packaging Quality Control Inspector

27 Aug 2018

