NEW ENGLAND BioLabs Inc.
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New England Biolabs
Certificate of Analysis

Product Name:	*Quick Ligation™ Kit*
Catalog Number:	*M2200S*
Unit Definition:	*N/A*
Packaging Lot Number:	*10118338*
Expiration Date:	*07/2023*
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-M2200S/L v1.0*

Quick Ligation™ Kit Component List			
NEB Part Number	**Component Description**	**Lot Number**	**Individual QC Result**
M2200SVIAL	Quick Ligation™ Kit	10113966	**Pass**
B2200SVIAL	Quick Ligation™ Reaction Buffer	10109465	**Pass**

Assay Name/Specification	**Lot # 10118338**
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**
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 37ºC. After incubation for 16 hours, >90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.	**Pass**
qPCR DNA Contamination (E. coli Genomic) 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**
Protein Purity Assay (SDS-PAGE) Quick Ligase is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue	**Pass**

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detection.	
Exonuclease Activity (Radioactivity Release) A 50 µl reaction in NEBuffer 1 containing 1 µg of a mixture of single and double-stranded [^{3}H] E. coli DNA and a minimum of 2000 units of Quick Ligase incubated for 4 hours at 37ºC 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 2000 units of Quick Ligase incubated for 4 hours at 37ºC results in <10% conversion to the nicked form as determined by agarose gel 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**
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**
Non-Specific DNase Activity (16 Hour) A 50 µl reaction in NEBuffer 1 containing 1 µg of CIP-treated Lambda-HindIII DNA and a minimum of 2000 units of Quick Ligase incubated for 16 hours at 37ºC results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	**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**

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Ligation and Recutting (Terminal Integrity, Digested DNA) A 20 µl reaction in 1X T4 DNA Ligase Reaction Buffer containing 2 µg of Lambda 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.	**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.

Ana Egana
Production Scientist
02 Sep 2021

Michael Tonello
Packaging Quality Control Inspector
02 Sep 2021