NEW ENGLAND BioLabs Inc.
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New England Biolabs
Certificate of Analysis

Product Name:	*NEBridge® Ligase Master Mix*
Catalog Number:	*M1100L*
Concentration:	*3 X Concentrate*
Packaging Lot Number:	*10205049*
Expiration Date:	*08/2025*
Storage Temperature:	*-20°C*
Specification Version:	*PS-M1100S/L v1.0*
Composition (1X):	*Proprietary*

NEBridge® Ligase Master Mix Component List

NEB Part Number	Component Description	Lot Number	Individual QC Result
M1100LVIAL	NEBridge® Ligase Master Mix	10202242	**Pass**

Assay Name/Specification	Lot # 10205049
Functional Testing (Assembly) A 15 µl reaction containing 75 ng pGGAselect (Golden Gate destination plasmid, CamR), 75 ng each of 5 plasmids carrying fragments of a gene encoding laclZ, 1 µl of BsaI-HF v2 and 5 µl NEBridge Ligase Master Mix is incubated for 30 cycles of 37°C for 1 minute, 16°C for 1 minute, and then at 60°C for 5 minutes to linearize any remaining plasmid. Successfully assembled fragments result in laclZ gene in the pGGAselect vector and yield blue colonies on Cam/XGAL/IPTG agar plates. Transformation of T7 Express Competent E. coli (High Efficiency, NEB #C2566) with 2 µl of the assembly reaction yields >250 colonies and > 80% blue colonies when 5% of transformation is plated.	**Pass**
Functional Testing (Ligation and Transformation, Blunt Ends) After a 15 minute ligation of linearized, dephosphorylated LITMUS 28 containing blunt EcoRV ends and a mixture of compatible insert fragments, transformation into chemically competent NEB 5-alpha competent E. coli (high efficiency) cells yields a minimum of 106 recombinant transformants per µg plasmid DNA.	**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 10 µl of NEBridge™ Ligase Master Mix incubated for 16 hours at 37ºC results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.	**Pass**

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

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