New England Biolabs
Product Specification

Product Name: NEBuilder® HiFi DNA Assembly Cloning Kit
Catalog #: E5520S
Kit Components:
- NEBuilder® High-Fidelity Master Mix (M5520) — Store at -20°C
- NEBuilder® Positive Control (N2611) — Store at -20°C
- NEB® 5-alpha Competent E. coli (High Efficiency) (C2987) — Store at -80°C
- SOC Outgrowth Medium (B9020) — Store at 4°C
- pUC19 Vector (N3041) — Store at -20°C

Shelf Life: 12 months
Storage Temp: Multi-temperature
Specification Version: PS-E5520S v1.0
Effective Date: 09 Apr 2018

<table>
<thead>
<tr>
<th>Assay Name/Specification (minimum release criteria)</th>
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<tbody>
<tr>
<td>Functional Testing (NEBuilder® HiFi DNA Assembly) - 10 µl of 2X NEBuilder® HiFi DNA Assembly Cloning Kit was incubated with 0.05 pmol each of 6 DNA fragments (4 fragments of 1,000 bp, one fragment of 1,152 bp with 80 bp overlap, and a vector of 3,373 bp with a 20 bp overlap) in a final volume of 20 µl at 50°C for 60 minutes. NEB® 5-alpha Competent E. coli (High Efficiency) were transformed with 2 µl of the assembled products. Successfully assembled fragments produce an intact lacZ gene in the pACYC184 vector, and yield blue colonies on an IPTG/Xgal/Chloramphenicol plate when incubated overnight at 37°C after transformation. Greater than 100 blue colonies were observed when 1/10 of the outgrowth (500 µl) was spread on a plate.</td>
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* Individual Product Component Note - Standard Quality Control Tests are performed for each component included in NEBuilder® HiFi DNA Assembly Cloning Kit and meet the designated specifications.

Derek Robinson
Director of Quality Control

Date 09 Apr 2018