Product Name: SplintR® Ligase
Catalog Number: M0375L
Concentration: 25,000 U/ml
Unit Definition: One unit is defined as the amount of enzyme needed to ligate (to 50% completion) 2 picomoles of a tripartite FAM-labeled DNA:RNA hybrid substrate in a 20 µl reaction at 25°C in 15 minutes in 1X SplintR® Ligase Reaction Buffer.

Packaging Lot Number: 10159592
Expiration Date: 07/2024
Storage Temperature: -20°C
Storage Conditions: 10 mM Tris-HCl, 300 mM NaCl, 1 mM DTT, 0.1 mM EDTA, 50 % Glycerol, (pH 7.4 @ 25°C)
Specification Version: PS-M0375S/L v1.0

### SplintR® Ligase Component List

<table>
<thead>
<tr>
<th>NEB Part Number</th>
<th>Component Description</th>
<th>Lot Number</th>
<th>Individual QC Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>M0375LVIAL</td>
<td>SplintR® Ligase</td>
<td>10156776</td>
<td>Pass</td>
</tr>
<tr>
<td>B0375SVIAL</td>
<td>10X SplintR® Ligase Reaction Buffer</td>
<td>10156815</td>
<td>Pass</td>
</tr>
</tbody>
</table>

### Assay Name/Specification

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</thead>
</table>

**Endonuclease Activity (Nicking)**
A 50 µl reaction in SplintR® Ligase Reaction Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 125 units of SplintR® Ligase incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.

**Exonuclease Activity (Radioactivity Release)**
A 50 µl reaction in SplintR® Ligase Reaction Buffer containing 1 µg of a mixture of single and double-stranded [³H] E. coli DNA and a minimum of 125 units of SplintR® Ligase incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.

**Protein Purity Assay (SDS-PAGE)**
SplintR® Ligase is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.

**RNase Activity (Extended Digestion)**
A 10 µl reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA
Assay Name/Specification

and a minimum of 25 units of SplintR® 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.

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<tr>
<th>Assay Name/Specification</th>
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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](http://www.neb.com/trademarks) for additional information.

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31 Aug 2022

Erin Varney  
Packaging Quality Control Inspector  
31 Aug 2022