New England Biolabs
Product Specification

Product Name: Hi-T7™ RNA Polymerase
Catalog #: M0658S
Concentration: 50,000 units/ml
Unit Definition: One unit is defined as the amount of enzyme required to incorporate 1 nmol ATP into acid-insoluble material in 1 hour at 50°C.
Shelf Life: 24 months
Storage Temp: -20°C
Storage Conditions: 50 mM Tris-Cl, 100 mM NaCl, 1 mM EDTA, 1 mM DTT, 0.1% Triton® X-100, 50% Glycerol, (pH 7.9 @ 25°C)
Specification Version: PS-M0658S v1.0
Effective Date: 20 Jun 2018

<table>
<thead>
<tr>
<th>Assay Name/Specification (minimum release criteria)</th>
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<tbody>
<tr>
<td><strong>Endonuclease Activity (Nicking)</strong> - A 50 µl reaction in NEBuffer 4 containing 1 µg of supercoiled PhiX174 DNA and a minimum of 150 units of Hi-T7™ RNA Polymerase incubated for 4 hours at 37°C results in &lt;10% conversion to the nicked form as determined by agarose gel electrophoresis.</td>
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<td><strong>Exonuclease Activity (Radioactivity Release)</strong> - A 50 µl reaction in NEBuffer 4 containing 1 µg of a mixture of single and double-stranded [³H] E. coli DNA and a minimum of 150 units of Hi-T7™ RNA Polymerase incubated for 4 hours at 37°C releases &lt;0.1% of the total radioactivity.</td>
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<td><strong>Protein Purity Assay (SDS-PAGE)</strong> - Hi-T7™ RNA Polymerase is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.</td>
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<td><strong>RNase Activity (Extended Digestion)</strong> - A 10 µl reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 50 units of Hi-T7™ RNA Polymerase is incubated at 37°C. After incubation for 4 hours, &gt;90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.</td>
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Derek Robinson
Director of Quality Control

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