

## New England Biolabs Certificate of Analysis

**Product Name:** T7 RNA Polymerase (High Concentration)  
**Catalog Number:** M0460T  
**Concentration:** 1,000,000 U/ml  
**Unit Definition:** One unit is defined as the amount of enzyme that will incorporate 1 nmol ATP into acid-insoluble material in a total reaction volume of 50 µl in 1 hour at 37°C in 1X RNA Polymerase Reaction Buffer.  
**Packaging Lot Number:** 10075393  
**Expiration Date:** 05/2022  
**Storage Temperature:** -20°C  
**Storage Conditions:** 100 mM NaCl , 50 mM Tris-HCl (pH 7.9), 1 mM EDTA , 20 mM BME , 0.1 % Triton X-100 , 50 % Glycerol  
**Specification Version:** PS-M0460T v1.0

T7 RNA Polymerase (High Concentration) Component List			
NEB Part Number	Component Description	Lot Number	Individual QC Result
M0460TVIAL	T7 RNA Polymerase (High Concentration)	10075392	Pass
B9012SVIAL	RNAPol Reaction Buffer	10073837	Pass
B2534AVIAL	MgCl <sub>2</sub> Solution	10057487	Pass

Assay Name/Specification	Lot # 10075393
<b>Endonuclease Activity (Nicking)</b> A 50 µl reaction in RNAPol Reaction Buffer containing 1 µg of supercoiled PhiX174 DNA and a minimum of 150 units of T7 RNA Polymerase incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.	Pass
<b>Exonuclease Activity (Radioactivity Release)</b> A 50 µl reaction in RNAPol Reaction Buffer containing 1 µg of a mixture of single and double-stranded [ <sup>3</sup> H] E. coli DNA and a minimum of 150 units of T7 RNA Polymerase incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.	Pass
<b>Protein Purity Assay (SDS-PAGE)</b> T7 RNA Polymerase is ≥ 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
<b>Non-Specific DNase Activity (16 Hour)</b>	Pass

Assay Name/Specification	Lot # 10075393
<p>A 50 µl reaction in RNAPol Reaction Buffer containing 1 µg of Lambda DNA and a minimum of 250 units of T7 RNA Polymerase incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.</p>	
<p><b>Promoter Specificity</b> A 50 µl reaction in RNAPol Reaction Buffer in the presence of 2 mM NTPs containing 1 µg of Lambda DNA as a template and a minimum of 200 units of T7 RNA Polymerase incubated for 1 hour at 37°C results in &lt;1.5% of the amount of product incorporated as compared to a control reaction using T7 DNA as a template.</p>	<b>Pass</b>
<p><b>RNase Activity (Extended Digestion)</b> A 10 µl reaction in RNAPol Reaction Buffer containing 40 ng of a 300 base single-stranded RNA and a minimum of 50 units of 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.</p>	<b>Pass</b>

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

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