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New England Biolabs Certificate of Analysis

Product Name:	T7 RNA Polymerase
Catalog #:	M0251S/L
Concentration:	50,000 units/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 μ l in 1 hour at 37°C in 1X RNA Polymerase Reaction Buffer.
Lot #:	0091405
Assay Date:	05/2014
Expiration Date:	5/2016
Storage Temp:	-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-M0251S/L v1.0
Effective Date:	15 May 2014

Assay Name/Specification (minimum release criteria)	Lot #0091405
Endonuclease Activity (Nicking) - A 50 μ l reaction in RNAPol Reaction Buffer containing 1 μ g 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
Exonuclease Activity (Radioactivity Release) - A 50 μ l reaction in RNAPol Reaction Buffer containing 1 μ g of a mixture of single and double-stranded [³ H] <i>E. coli</i> 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
Non-Specific DNase Activity (16 Hour) - 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.	Pass
Promoter Specificity - A 50 μ l reaction in RNAPol Reaction Buffer in the presence of 100 μ M NTPs containing 1 μ g of Lambda DNA as a template and a minimum of 200 units of T7 RNA Polymerase incubated for 4 hours at 37°C results in <1.5% of the amount of product incorporated as compared to a control reaction using T7 DNA as a template.	Pass
Protein Purity Assay (SDS-PAGE) - T7 RNA Polymerase is \geq 95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.	Pass
RNase Activity (Extended Digestion) - A 10 μ l 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, >90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.	Pass

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Authorized by Derek Robinson 15 May 2014



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Inspected by Dongxian Yue 15 May 2014