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## Certificate of Analysis

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## T7 RNA Polymerase

Catalog No: 1290

**Lot No:** See Product Label

**Package Size:** See Product Label

**Concentration:** See Product Label

**Protein (Bradford):** 0.199 mg/ml

**Specific Activity:** 126,016 units/mg

**Storage Conditions:** Store at -20°C

### Description

- DNA-dependent RNA polymerase which has stringent specificity for T7 phage promoters sequence (1)
- Ultrapure recombinant enzyme

### Applications

- Efficiently synthesizes in vitro transcripts from almost any DNA that is downstream from a T7 promoter (2)
- Suitable for preparing labeled single-stranded RNA probes of high specific activity (3)
- Transcripts can be used as hybridization probes, templates for in vitro translation, substrates in RNA processing systems, or exon and intron mapping of genomic DNA

### Unit Definition

One unit is the amount of enzyme required to incorporate 1 nmol of labeled UTP into acid-insoluble material in 60 minutes at 37°C.

### Assay Conditions

40 mM Tris-HCl (pH 7.9)  
8 mM MgCl<sub>2</sub>  
10 mM dithiothreitol  
4 mM Spermidine-(HCl)<sub>3</sub>  
1 µg pGEM-7Zf(+) plasmid  
0.4 mM each of ATP, CTP, GTP and [α-<sup>32</sup>P]UTP  
Reaction volume is 25 µl

### Storage Buffer

20 mM Potassium Phosphate (pH 7.5)  
100 mM NaCl  
1 mM dithiothreitol  
1 mM EDTA  
50% glycerol (w/v)

### Quality Control

**DNase, double-stranded:** Incubation of 50, 100, 200 units of enzyme with 0.015 µg of [<sup>32</sup>P] Lambda DNA for 1 hour at 37°C resulted in ≤0.4 slope of %-end label released per unit of enzyme. Reaction volume of 25 µl.

**DNase, single-stranded:** Incubation of 50, 100, 200 units of enzyme with 0.015 µg of heat denatured [<sup>32</sup>P] Lambda DNA for 1 hour at 37°C resulted in ≤0.4 slope of %-end label released per unit of enzyme. Reaction volume 25 µl.

#### 5'Exonuclease/5' Phosphatase:

Incubation of 50, 100, 200 units of enzyme with 0.25 µg of [5'-<sup>32</sup>P] Lambda/Hae III DNA fragments for 1 hour at 37°C resulted in a ≤0.4 slope of %-end label released per unit of enzyme. Reaction volume of 50 µl.

**RNase:** Incubation of 50, 100, 200 units of enzyme with 0.015 µg of [<sup>32</sup>P] RNA transcript for 1 hour at 37°C resulted in ≤0.4 slope of %-end label released per unit of enzyme. Reaction volume of 50 µl.

**Purity:** Approximately ≥90% pure, as judged by SDS-polyacrylamide gel electrophoresis.

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For more information or to re-order, please visit [www.chimerx.com](http://www.chimerx.com)

## **References**

- (1) Chamberlin, M. and Ring, J. (1973) *J. Biol. Chem.* 248, 2235-2244 (2) Tabor, S. and Richardson, C.C. (1985) *Proc. Natl. Acad. Sci. USA* 82, 1074-1078 (3) Sambrook, J., Fritsch, E.F. and Maniatis, T. (1989) *Mol. Cloning: A Lab. Manual, Second Ed.*, pp. 10.27-10.37