

T7 RNA Polymerase

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Cat. No. 18033-019**Lot No. _____ 2,500 units; 50 U/μl****Exp. Date: _____. Store at -20°C (not frost-free)**Description:

T7 RNA Polymerase is a DNA-dependent RNA polymerase which has been isolated from *E. coli* expressing the T7 RNA polymerase gene on a plasmid (1). The enzyme has an extremely high specificity for T7 promoter sequences (2) and will synthesize large quantities of RNA from a DNA fragment inserted downstream from a promoter. A strong class III promoter (3) has been used to construct various cloning vectors, and inserts into the multiple cloning site of these vectors can be transcribed to generate discrete RNA's.

Components:

18033-019	T7 RNA Polymerase	Lot No.
Y90108	5X T3/T7 Buffer	Lot No.
Y00147	0.1 M DTT	Lot No.

Unit Definition:

One unit incorporates 1 nmol of labeled nucleotide into acid-precipitable material in 1 hour at 37°C.

Storage Buffer:

20 mM potassium phosphate
(pH 7.7)
0.1 M NaCl
0.1 mM EDTA
1 mM DTT
50% (v/v) glycerol
0.01% (w/v) Triton® X-100

5X T3/T7 Buffer:

0.2 M Tris-HCl (pH 8.0)
40 mM MgCl₂
10 mM spermidine-(HCl)₃
125 mM NaCl
Refer to Functional Assay
Conditions on reverse side for
further details.

Quality Control Assays: See Back Page

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This product is distributed for laboratory research use only. CAUTION: Not for diagnostic use. The safety and efficacy of this product in diagnostic or other clinical uses has not been established.

For technical questions about this product, call the Life Technologies TECH-LINE™ (800) 828-6666.

Quality Control Assays:

This product has passed the following quality control assays: functional absence of exonuclease, endo-ribonuclease and DNA nicking activities; performance in a transcription reaction.

The enclosed buffers were assayed with the enzyme and met quality control specifications.

Functional Assay Conditions:

2 μ l 5X T3/T7 Buffer
70 μ M [α -³²P]UTP (280 μ Ci of 400 Ci/mmol)
0.4 mM each ATP, CTP, GTP
5 mM DTT
0.1 μ g linearized template DNA
50 units T7 RNA Polymerase
Reaction Volume: 10 μ l
Incubation: 10 minutes at 37°C

NOTE: The reaction is not set up on ice due to potential precipitation of DNA in the presence of spermidine.

References:

1. Davanloo, P., Rosenberg, A. H., Dunn, J. J., and Studier, F. W. (1984) *Proc. Natl. Acad. Sci. USA* 81, 2035.
2. Chamberlin, M., McGrath, J., and Waskell, L. (1970) *Nature* 228, 227.
3. Studier, F. W., and Dunn, J. J. (1983) *Cold Spring Harbor Symposia on Quantitative Biology XLVII*, 999.