Invitrogen
life technologies

T7 RNA Polymerase

Cat. No. 18033-019 **Size: 2,500 units**
Conc.: 50 U/µl **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
Y90108	5X T3/T7 Buffer
Y00147	0.1 M DTT

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

Storage Buffer:
20 mM Tris-HC1 (pH 7.5)
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 $MgCl_2$
10 mM spermidine-$(HCl)_3$
125 mM NaCl
Refer to Functional Assay Conditions on reverse side for further details.

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Quality Control:
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 µl 5X T3/T7 Buffer
70 µM [α-^{32}P]UTP (280 µCi of 400 Ci/mmol)
0.4 mM each ATP, CTP, GTP
5 mM DTT
0.1 µg linearized template DNA
50 units T7 RNA Polymerase
Reaction Volume: 10 µl
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. Acid. 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.