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T7 RNA Polymerase, GMP-Grade

Catalog #GMP-T7P-EE101

Storage Condition -20°C ± 5°C for 24 months. Avoid repeated freeze/thaw cycles.
Form Liquid
Source An *E. coli* strain that carries the gene for bacteriophage T7 RNA Polymerase
Storage Buffer 50 mM Tris-HCl, 100 mM NaCl, 1 mM EDTA, 20 mM 2-mercaptoethanol, 0.1% Triton X-100, 50% Glycerol, pH 7.9
Concentration 50U/µL
Unit Definition One unit is defined as the amount of enzyme required to incorporate 1nmol ATP into acid-soluble material in a total reaction volume of 50µL in 1 hour at 37°C.

Product Contents

- T7 RNA Polymerase (50U/µL
- 5X Transcription Buffer-1 (Tris-Acetate, $Mg(OAc)_2$, NaOAc, Spermidine, DTT, pH 8.1)

Product Description

T7 RNA Polymerase is a DNA-dependent RNA polymerase that is highly specific for the T7 phage promoters. T7 RNA Polymerase catalyzes *in vitro* mRNA synthesis in the 5' to 3' direction and accepts modified nucleotides into the transcripts. The mRNA products can be used for many downstream applications.

Applications

- mRNA for *in vitro* translation
- Radiolabeled RNA probe
- Non-isotopic RNA labeling
- RNA vaccines preparation
- Guide RNA preparation
- Antisense RNA preparation
- Capped mRNA synthesis with cap analog

Quality Control Statement

KACTUS manufactures this product according to GMP guidelines and performs stringent quality control testing before release. It is suitable for mRNA therapeutics and manufacturing of mRNA vaccines. Regulatory support documents are available. Please contact support@kactusbio.us for more information.

Quality Control Release Criteria

Assay	Criteria
Activity (Probe Incorporation)	≥ 50kU/mL
Purity (SEC-HPLC)	≥ 95%
Residual Endonuclease	Negative
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Endotoxin	≤ 10EU/mL
Residual DNase	Negative
Residual RNase	Negative
Residual Protease	Negative
Residual Host Cell Protein	≤ 20ng/mg
Residua	≤ 100pg/mL
Residual Heavy Metal	≤ 10ppm
Bioburden	≤ 1CFU/10mL

Protocol for *In Vitro* Transcription

1. Prepare the following reaction mixture.

Reagent	Quantity
5X Transcription Buffer-1 (included with T7 RNA Polymerase)	4µL
CTP/GTP/ATP/UTP (100mM each)	2µL each
Murine RNase Inhibitor (40U/µL)	1µL
Pyrophosphatase, Inorganic (0.1U/µL)	1µL
T7 RNA Polymerase (50U/µL)	2µL
Template DNA	1µg
RNase-free Water	Up to 20µL

2. Incubate at 37°C for 1-2 hours.
3. After transcription, add 2U DNase I to remove DNA template for 15 minutes at 37°C.
4. Inactivate DNase I by phenol/chloroform extraction.

Notes

- Murine RNase Inhibitor is added to protect RNA from possible RNase contamination.
- The reaction mixture should be prepared at room temperature as DNA may precipitate in the presence of spermidine at 4°C.
- The reaction may be scaled up as needed.