Version: 01
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**Datasheet**

# SP6 RNA Polymerase

**Cat. No.:** E00067-2; E00067-5; E00067-10
**Size:** 2 kU/ 5 kU/ 10 kU

## Product Introduction

Bacteriophage SP6 RNA polymerase is a DNA-dependent RNA polymerase with high affinity and specificity for SP6 phage promoter, limiting the SP6 RNA synthesis to DNA cloned downstream of an SP6 promoter.. Like T7 RNA Polymerase, the SP6 RNA Polymerase is also widely used for the synthesis of specific transcripts, as well as being a suitable model for studying the mechanisms of transcription. The RNA produced by SP6 RNA Polymerase is catalyzed from 5'→3' and is suitable for many downstream applications.

GenScript is offering SP6 RNA Polymerase produced by expression in an *E. coli* strain carrying a plasmid encoding the SP6 RNA Polymerase.

| | |
|---|---|
| **Source:** | Recombinant SP6 RNA Polymerase expressed by *E.coli* |
| **Species:** | *Salmonella typhimurium LT2* |
| **Molecular Weight:** | ~98.5 kDa |
| **Unit Definition:** | One unit is defined as the amount of enzyme required to incorporate 1 nmol ATP into an acid-insoluble material in 1 hour at 37 °C. |
| **Optimal active temperature:** | 37 °C. |
| **Formulation:** | Supplied as a solution of 50 mM Tris, 100 mM NaCl, 1 mM EDTA, 3 mM DTT, 0.1% Triton X-100, 50% glycerol. |
| **Storage & Stability:** | This product remains stable for up to 12 months at -20 °C. Avoid repeated freeze-thaw cycles. |
| **Application:** | • Synthesis of the single-strand RNA<br>• Synthesis of highly labeled RNA probes<br>• Synthesis of precursors of siRNA<br>• Synthesis of precursors for RNA splicing<br>• Synthesis of capped mRNA when a cap analog is used as a primer |

## Quality Control Specifications

| Assay | Specifications |
|---|---|
| Appearance | Clear, colorless liquid |
| Purity | ≥ 95% as analyzed by SDS-PAGE |
| Enzyme Activity | ≥ 20 U/μl |

| | |
|---|---|
| Endotoxin Level | ≤ 0.1 EU/μg of protein as analyzed by gel clotting method |
| Residual Endonuclease | Non-detectable |
| Residual Exonuclease | Non-detectable |
| Residual RNase | Non-detectable |

## Reagents Supplied

| Components | Amount | | | Storage |
|---|---|---|---|---|
| SP6 RNA Polymerase | 2 kU | 5 kU | 10 kU | -20 °C |
| 10 × Transcription Buffer | 1 ml | 1 ml | 2 ml | -20 °C |

## Protocols for *in vitro* transcription

Assemble the reaction in a nuclease-free microfuge tube or PCR strip tube on ice with the following order:

| Components | Volume |
|---|---|
| SP6 RNA Polymerase (E00067) | 40 U |
| 10 × Transcription Buffer | 2 μl |
| NTP Mix, 10 mM each, nuclease free (C01731) | 4 μl |
| Linearized template DNA | 0.2-1 μg |
| RNase Inhibitor (E00070) | 20 U |
| DTT (optional) | 5 mM final |
| RNase free water | to 20 μl |
| Incubate at 37 °C for 1 hour. | |
| We recommend removing template DNA with DNase I. Add 2 U of DNase I (E00053) into the reaction solution, mix thoroughly, and incubate at 37 °C for 30 min. | |

## Data Images

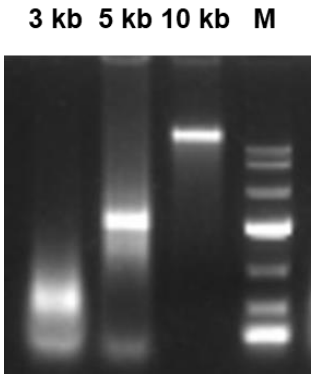


Fig 1. *In vitro* transcription of RNA with the recommended reaction conditions, the transcribed RNA can be up to 10 kb.

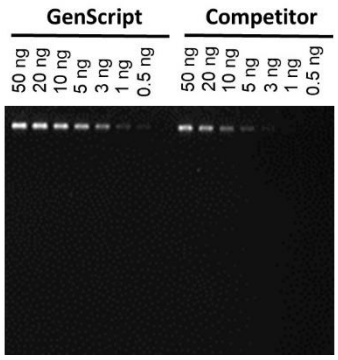


Fig 2. *In vitro* transcription of RNA with tiny amount of DNA template, GenScript's SP6 RNA Polymerase performs better than Competitor's.

## References


1. Butler, Eugene T., and M. J. Chamberlin. "Bacteriophage SP6-specific RNA polymerase. I. Isolation and characterization of the enzyme." Journal of Biological Chemistry 257.10 (1982): 5772-5778.
2. Kotani, Hirokazu, et al. "Nucleotide sequence and expression of the cloned gene of bacteriophage SP6 RNA polymerase." Nucleic acids research 15.6 (1987): 2653-2664.