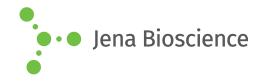
DATA SHEET





■ T7 P&L RNA Polymerase HC

Highly concentrated and modified T7 RNA Polymerase for set-up of high yield in vitro transcription reactions

Cat. No.	Amount
RNT-018	50 μl (2 μg/μl)

For general laboratory use.

Shipping: shipped on gel packs
Storage Conditions: store at -20 °C

Additional Storage Conditions: avoid freeze/thaw cycles

Shelf Life: 12 months

Purity: ≥ 95 % (SDS-PAGE)

Form: liquid

Concentration: $2.0 - 2.2 \mu g/\mu l$ (A280, $\epsilon = 140 L mmol^{-1} cm^{-1}$ [1] in T7

RNA Polymerase HC Storage Buffer)

Description:

Bacteriophage T7 RNA Polymerase is a DNA-dependent RNA polymerase (99 kDa) that catalyzes *in vitro* RNA synthesis from a DNA sequence containing a T7 phage promoter.

The highly concentrated version and modified T7 RNA Polymerase HC is ideally suited for set-up and optimization of high yield *in vitro* transcription reactions. Proline 266 has been replaced by leucine (P266L) which has been associated with

- decreased abortive transcription^[1].
- increased 5' homogeneity of transcripts synthesized from A-initiating phi2.5 promoter $^{[2]}$
- increased 5' incorporation efficiency of GTP analogs^[3]

The provided T7 IVT Set-up Buffer, $MgCl_2$ and DTT solutions enable testing of different reaction conditions such as nucleotide/ $MgCl_2$ concentrations. Recommendation for assay set-up with minimal optimization: HighYield T7 P&L RNA Synthesis Kit (#RNT-201).

Activity: 1 μ l T7 P&L RNA Polymerase HC generates >/= 30 μ g RNA in 10 min at 37°. This is comparable to 1000 U of NEB's T7 RNA Polymerase (High Concentration), #M0460T (activity assay conditions: 1x T7 IVT Set-up Buffer supplemented with 10 mM DTT, 12 mM MgCl₂, 3 mM NTPs, 1 μ l T7 RNA Polymerase HC and 1 μ g DNA template (1.4 kpb RNA transcript) in 20 μ l. RNA yield was determined with a fluorescence microplate assay.)

Content:

T7 P&L RNA Polymerase HC

1x 50 μl (2 μg/μl)

50 mM Tris-HCl, 1 mM EDTA, 100 mM NaCl, 5 mM DTT, 0.1 % Triton-X-100, 50 % Glycerol (v/v), pH 8.0

T7 IVT Set-up Buffer

1x 200 µl (10x)

400 mM Tris-HCl (pH 8.0), 20 mM Spermidin

DTT

1x 100 µl (100 mM)

MgCl₂

1x 200 µl (200 mM)

Related Products:

HighYield T7 RNA Synthesis Kit, #RNT-101 RNAse Inhibitor - recombinant, #PCR-392 NTP bundle, #NU-1014

Selected References:

[1] Guillerez et al. (2005) A mutation in T7 RNA polymerase that facilitates promoter clearance. Natl. Acad. Sci. U.S.A102:5958.

