T7 RNA Polymerase, 50 U/ µl

ORDERING INFORMATION

CAT. No.	SIZE	PACKAGE CONTENT
BR1101201	1,000 Units	T7 RNA Polymerase, 50 U/µl 10X T7 RNA Polymerase Reaction Buffer
BR1101202	5,000 Units	T7 RNA Polymerase, 50 U/µl 10X T7 RNA Polymerase Reaction Buffer

COMPONENT	COMPOSITION
T7 RNA Polymerase, 50 U/µl	T7 RNA Polymerase, 50 U/µl, in storage buffer containing 50 % (v/v) glycerol
10X T7 RNA Polymerase Reaction Buffer	Optimized T7 RNA Polymerase reaction buffer

STORAGE	Store at −20 °C until expiry date.
STORAGE BUFFER COMPOSITION	10 mM K3PO4, 200 mM KCl, 0.1 mM EDTA, 0.1% Tween® 20, 30 mM β-ME, 50% Glycerin - pH 7.9 at 25 °C

FEATURES

- Exceptionally pure enzyme
- Rigorously tested for DNA and RNase contamination to prevent unwanted background transcription
- Exceptional specificity for the T7 promoter

APPLICATIONS

- *in vitro* RNA synthesis from DNA template
- Radiolabeled RNA probes
- Non-isotopic RNA labeling
- Preparation of RNA vaccines
- Guide RNA for gene targeting
- mRNA for *in vitro* translation and micro injection
- RNA structure, processing and catalysis studies
- RNA amplification
- Anti-sense RNA for gene expression experiment

DESCRIPTION

biotechrabbit™ T7 RNA Polymerase is a highly specific, DNA-dependent RNA polymerase renowned for its ability to selectively transcribe RNA from DNA sequences that are under the control of T7 phage promoters. With a molecular weight of approximately 99 kDa, this enzyme plays a crucial role in the *in vitro* synthesis of RNA, offering a reliable solution for precise and efficient RNA production. The enzyme's exceptional specificity for the T7 promoter ensures that RNA transcription is not only highly regulated but also incredibly efficient, making it an indispensable tool in the generation of high-quality RNA.

Produced through recombinant expression in *Escherichia coli*, biotechrabbit's T7 RNA Polymerase is manufactured under stringent quality control conditions to guarantee the highest level of purity and activity. This includes rigorous testing for DNA and RNase contamination. Ensuring the absence of contaminating DNA is critical because it prevents unwanted background transcription that could compromise the accuracy of experimental results. Similarly, testing for RNase contamination is essential to safeguard the integrity of the RNA produced, as RNases can degrade RNA, leading to the loss of valuable samples and data. By eliminating these potential contaminants, biotechrabbit provides a product that is highly reliable, consistent, and suitable for even the most sensitive applications.

The enzyme is ideally suited for a variety of applications, including the synthesis of RNA for *in vitro* translation, the production of labeled RNA probes, the generation of antisense RNA, and various RNA structural and functional studies. In addition to its use in basic research, T7 RNA Polymerase is also a valuable tool in synthetic biology, where it facilitates the production of RNA transcripts for advanced applications such as CRISPR guide RNA synthesis, ribozyme production, and RNA aptamer generation. The enzyme's reliability and consistency, derived from rigorous quality checks during production, ensure reproducible results across a wide range of experimental setups, making biotechrabbit's T7 RNA Polymerase a trusted choice for researchers seeking high-performance RNA transcription solutions.

PROTOCOL

Prevention of contamination

When assembling the reactions, care should be taken to eliminate the possibility of contamination with undesired RNA, DNA and nucleases.

- Use separate clean areas for preparation of samples and reaction mixtures.
- Wear fresh gloves. Use sterile tubes and pipette tips with aerosol filters for assay setup.
- Use only water and reagents that are free of RNA, DNA and nucleases.
- With every assay setup, perform a contamination control reaction that does not include a template.

BASIC PROTOCOL

COMPONENT	VOLUME/CONCENTRATION
10X T7 RNA Polymerase Reaction Buffer	10 µl
Nucleotides (ATP, GTP, CTP, UTP)	3.75 mM each
RNAse Inhibitor (optional)	100 U
Template DNA	2 – 5 µg
T7 RNA Polymerase (50 U/µl)	2 – 4 µl
Nuclease free water	add up to 100 µl
Total volume	100 µl

- Mix and centrifuge briefly to collect the liquid in the bottom of the tube.
- Incubate for 2 hours at 37°C.
- Stop the reaction by adding 5 µl 0.5 M EDTA

RELATED PRODUCTS

RNase Inhibitor, 40 U/µl (BR040090X)

CERTIFICATE OF ANALYSIS

Unit definition

One unit is defined as the amount of enzyme needed to catalyze the incorporation of 5 nmol rCTP in acid-precipitable product within one hour at 37°C in a total volume of 100 µl.

Quality Control

Exonuclease assay

Linearized lambda/HindII fragments are incubated with T7 RNA Polymerase in a 50 µl reaction mixture for 4 h at 37°C. No degradation of DNA was observed.

Endonuclease assay

lambda DNA is incubated with T7 RNA Polymerase in a 50 µl reaction mixture for 4 h at 37°C. No degradation of DNA was observed.

Nick Activity

Supercoiled plasmid DNA is incubated with T7 RNA Polymerase in a 50 µl reaction mixture for 4 h at 37°C. No conversion of covalently closed circular DNA to nicked DNA was detected.

RNase Assay

A sample of the enzyme is incubated with a RNA template. RNase activity was not observed after agarose gel.

E.coli DNA contamination assay

A sample of the denatured DNA Polymerase is analyzed with specific primers targeting the 16S rRNA gene in qPCR for the presence of contaminating *E. coli* DNA. No *E. coli* DNA was detectable.

Purity

SDS-PAGE followed by Coomassie® blue staining confirms the purity of the enzyme.
The physical purity of T7 RNA Polymerase is ≥ 90%

Quality confirmed by: Head of Quality Control

SAFETY INSTRUCTIONS

For safety instructions please see Safety Data Sheets (SDS)
Sicherheitshinweise finden Sie in den Sicherheitsdatenblättern (SDB) unter
www.biotechrabbit.com/support/documentation.html

USEFUL HINTS

- Visit Applications at www.biotechrabbit.com for more products and product selection guides.
- Most biotechrabbit products are available in custom formulations and bulk amounts.
- In case any customization is required, please contact biotechrabbit via oem@biotechrabbit.com.

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Certified Quality-System mdc ISO 13485 ISO 9001

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