M-MLV Reverse Transcriptase

[Cat. No.] E-3121, E-3122

Introduction

Moloney Murine Leukemia Virus (*M-MLV*) Reverse Transcriptase (RTase) is an RNA-dependent DNA polymerase. It can synthesize complementary DNA by using RNA molecules as a template. *M-MLV* RTase is isolated from a *E.coli* strain containing a recombinant clone and it can synthesize cDNA up to 9 kb. Furthermore, by using the RNase H+ of *M-MLV* RTase, template RNA is removed after the cDNA synthesis to minimize the effect of residual template RNA in PCR. This product can be applied for first-strand cDNA synthesis for RT-PCR and qRT-PCR.

Applications

- First-strand cDNA synthesis from RNA
- RT-PCR and qRT-PCR

Components

Components	E-3121	E-3122
M-MLV Reverse Transcriptase	10,000 U (50 µl)	50,000 U (50 µl x 5)
5X Reaction buffer	0.5 ml	0.5 ml x 5
10 mM dNTPs	0.2 ml	0.2 ml x 5
100 mM DTT	0.3 ml	0.3 ml x 5

*** Note:** For research use only. Not for use in diagnostic or therapeutic procedures.

Specifications

M-MLV Reverse Transcriptase	
5' to 3' exonuclease activity	No
3' to 5' exonuclease activity	No
3'–A overhang	No
Fragment size	Up to 9 kb

Buffer Composition

5X Reaction buffer	150 mM Tris-HCl, 250 mM KCl, 10 mM $MgCl_2$, pH 8.1

Storage Buffer

M-MLV Reverse Transcriptase is supplied in 50% (v/v) glycerol containing 20 mM Tris-HCl, 150 mM NaCl, 0.1 mM EDTA, 1 mM DTT, 50 mM $(NH_4)_2SO_4$, and stabilizer, pH 7.6.

Unit Definition

One unit is defined as the amount of enzyme that incorporates 1 nmole of dTTP into acid-insoluble products in 10 min at 37°C using poly(A)·oligo(dT) as template primer.

Quality Control

- Nuclease Contamination Assay: Nuclease activity is not detected after incubation of 1 µg of DNA and RNA with 200 U of *M-MLV* Reverse Transcriptase at 37-42°C for 3 hrs.

Storage

Store at -20°C. If stored in the recommended temperature, this product will be stable until the expiration date printed out on the label.

Online Resources

Korean **English**

Visit our **product page** for additional information and protocols.

Ordering Information

Description		Cat. No
M-MLV Reverse Transcriptase	10,000 U (50 rxn)	E-3121
	50,000 U (250 rxn)	E-3122

Notice

BIONEER corporation reserves the right to make corrections, modifications, improvements and other changes to its products, services, specifications or product descriptions at any time without notice.

Explanation of Symbols

LOT Batch Code · REF Catalog Number · Caution · Consult Instructions For Use · Contains Sufficient for <n> tests · RUO Research Use Only · Temperature Limitation · Use-by Date

Experimental Procedures

Step 1

Primer annealing

1. Mix template RNA and primers in a sterile tube (not provided) indicated as below.

- Amount of template RNA and primers

Components		20 µl reaction
Template RNA	Total RNA	1 µg
	RNA	5-100 ng
Primers	Oligo dT or Random primer	10-100 pmol
	Sequence specific	10-30 pmol

2. Incubate the mixture at 65°C for 10 min and place it on ice directly.

Step 2

Preparation of reaction mixture

3. Transfer the incubated mixture to other sterile tubes (not provided) and mix with 5X Reaction buffer, 10 mM dNTPs, 100 mM DTT, RNase inhibitor (not provided), *M-MLV* Reverse Transcriptase, and nuclease-free water to a total volume of 20 µl.

- Preparation of reaction mixture

Components	20 µl reaction
Step 1 mixture	Variable
5X Reaction buffer	4 µl
10 mM dNTPs	2 µl or Variable
100 mM DTT	2 µl
RNase inhibitor (Not provided)	Variable
M-MLV Reverse Transcriptase (200 U/ µl)	200 U
Nuclease-free water	Variable
Total volume	20 µl

Step 3

cDNA synthesis

4. Perform the reaction under the following conditions.

Step	Temperature	Time
cDNA synthesis	37-42°C	1 hr
Heat inactivation	95°C	5 min

5. After the reaction, maintain the reaction mixture at 4°C. The samples can be stored at -20°C until use.