Poly(U) Polymerase

NEW ENGLAND BioLabs Inc.
1-800-632-7799
info@neb.com
www.neb.com

M0337S 003161218121

M0337S

RR NEB2 37° 65°

60 units	**2,000 U/ml**	**Lot: 0031612**
RECOMBINANT	**Store at –20°C**	**Exp: 12/18**

Description: Poly(U) Polymerase catalyzes the template independent addition of UMP from UTP or AMP from ATP to the 3´ end of RNA.

Source: An *E. coli* strain that carries the cloned poly(U) polymerase gene of *Schizosaccharomyces pombe* Cid1.

Applications:
- Labeling of RNA with UTP
- Poly(U) tailing of RNA for cloning
- Studying effects of poly(U) tailing on stability and translation of RNA transferred into eukaryotic cells
- Poly(A) tailing of 2´O-Me modified 3´ ends

Supplied in: 10 mM Tris-HCl (pH 7.5), 100 mM NaCl, 0.1 mM EDTA, 1.0 mM DTT and 50% glycerol.

Reagents Supplied with Enzyme:
10X NEBuffer 2

Reaction Conditions: 1X NEBuffer 2 supplemented with 1 mM UTP.* Incubate at 37°C.

Note: UTP is not included in the buffer.

1X NEBuffer 2:
50 mM NaCl
10 mM Tris-HCl
10 mM $MgCl_2$
1 mM DTT
pH 7.9 @ 25°C

Unit Definition: One unit is defined as the amount of enzyme that incorporates 1 nmol of UMP into RNA in a 50 µl volume in 10 minutes at 37°C.

Unit Assay Conditions: 1X NEBuffer 2, 0.5 mM ^{3}H UTP and 5 µg yeast RNA are combined in a 50 µl reaction incubated at 37°C for 10 minutes.

Protocol for a Typical Tailing Reaction:

1. Combine the following in a sterile microcentrifuge tube:

10X NEBuffer 2	2.5 µl
UTP	0.5 mM final
RNA	>100 pmol
RNase Inhibitor* (40 units/µl)	1 µl
Poly(U) Polymerase (2 units/µl)	1 µl
H_2O	to 25 µl

2. Incubate at 37°C for 10 minutes.

*RNase Inhibitor is recommended but not required.

Quality Assurance: Poly(U) Polymerase contains no detectable DNAses, and RNAses. The purified protein contains no detectable DNA or RNA.

Quality Control Assays

RNase Assay: Incubation of a 10 µl reaction containing 2 units of Poly(U) Polymerase with 40 ng of RNA transcript for 16 hours at 37°C resulted in no detectable degradation of the RNA as determined by gel electrophoresis.

DNA Exonuclease Activity: Incubation of a 50 µl reaction containing 10 units of Poly(U) Polymerase with 1 cg of a mixture of single and double-stranded ^{3}H *E.coli* DNA for 3 hours at 37°C released < 0.1% of the total radioactivity.

DNA Endonuclease Activity: Incubation of a 50 µl reaction containing 10 units of Poly(U) Polymerase with 1 µg of supercoiled plasmid for 4 hours at 37°C resulted in < 10% conversion to nicked molecules as determined by agarose gel electrophoresis.

(see other side)

CERTIFICATE OF ANALYSIS

Poly(U) Polymerase

NEW ENGLAND BioLabs Inc.
1-800-632-7799
info@neb.com
www.neb.com

M0337S 003161218121

M0337S

RR NEB2 37° 65°

60 units 2,000 U/ml Lot: 0031612
RECOMBINANT Store at –20°C Exp: 12/18

Description: Poly(U) Polymerase catalyzes the template independent addition of UMP from UTP or AMP from ATP to the 3´ end of RNA.

Source: An *E. coli* strain that carries the cloned poly(U) polymerase gene of *Schizosaccharomyces pombe* Cid1.

Applications:
- Labeling of RNA with UTP
- Poly(U) tailing of RNA for cloning
- Studying effects of poly(U) tailing on stability and translation of RNA transferred into eukaryotic cells
- Poly(A) tailing of 2´O-Me modified 3´ ends

Supplied in: 10 mM Tris-HCl (pH 7.5), 100 mM NaCl, 0.1 mM EDTA, 1.0 mM DTT and 50% glycerol.

Reagents Supplied with Enzyme:
10X NEBuffer 2

Reaction Conditions: 1X NEBuffer 2 supplemented with 1 mM UTP.* Incubate at 37°C.

Note: UTP is not included in the buffer.

1X NEBuffer 2:
50 mM NaCl
10 mM Tris-HCl
10 mM $MgCl_2$
1 mM DTT
pH 7.9 @ 25°C

Unit Definition: One unit is defined as the amount of enzyme that incorporates 1 nmol of UMP into RNA in a 50 µl volume in 10 minutes at 37°C.

Unit Assay Conditions: 1X NEBuffer 2, 0.5 mM ^{3}H UTP and 5 µg yeast RNA are combined in a 50 µl reaction incubated at 37°C for 10 minutes.

Protocol for a Typical Tailing Reaction:

1. Combine the following in a sterile microcentrifuge tube:

10X NEBuffer 2	2.5 µl
UTP	0.5 mM final
RNA	>100 pmol
RNase Inhibitor* (40 units/µl)	1 µl
Poly(U) Polymerase (2 units/µl)	1 µl
H_2O	to 25 µl

2. Incubate at 37°C for 10 minutes.

*RNase Inhibitor is recommended but not required.

Quality Assurance: Poly(U) Polymerase contains no detectable DNAses, and RNAses. The purified protein contains no detectable DNA or RNA.

Quality Control Assays

RNase Assay: Incubation of a 10 µl reaction containing 2 units of Poly(U) Polymerase with 40 ng of RNA transcript for 16 hours at 37°C resulted in no detectable degradation of the RNA as determined by gel electrophoresis.

DNA Exonuclease Activity: Incubation of a 50 µl reaction containing 10 units of Poly(U) Polymerase with 1 cg of a mixture of single and double-stranded ^{3}H *E.coli* DNA for 3 hours at 37°C released < 0.1% of the total radioactivity.

DNA Endonuclease Activity: Incubation of a 50 µl reaction containing 10 units of Poly(U) Polymerase with 1 µg of supercoiled plasmid for 4 hours at 37°C resulted in < 10% conversion to nicked molecules as determined by agarose gel electrophoresis.

(see other side)

CERTIFICATE OF ANALYSIS

Notes: Poly(U) Polymerase in NEBuffer 2 will incorporate UMP or AMP from UTP or ATP into RNA. Tailing length of poly(U) varies with UTP. Poly(U) Polymerase is highly processive under low primer concentrations (< 100 pmol)

References:

1. Wickens, M. and Kwak, J.E. (2008) *Science* 319, 1344.
2. Kwak, J.E. and Wickens, M. (2008) *RNA* 13, 860.
3. Rissland, O.O.S., Mikulasova, A. and Norbury, C.J. (2007) *Molecular and Cell Biology* 27, 3612.

Companion Products:

RNase Inhibitor, Murine
#M0314S 3,000 units
#M0314L 15,000 units

RNase Inhibitor, Human Placenta
#M0307S 2,000 units
#M0307L 10,000 units

Notes: Poly(U) Polymerase in NEBuffer 2 will incorporate UMP or AMP from UTP or ATP into RNA. Tailing length of poly(U) varies with UTP. Poly(U) Polymerase is highly processive under low primer concentrations (< 100 pmol)

References:

1. Wickens, M. and Kwak, J.E. (2008) *Science* 319, 1344.
2. Kwak, J.E. and Wickens, M. (2008) *RNA* 13, 860.
3. Rissland, O.O.S., Mikulasova, A. and Norbury, C.J. (2007) *Molecular and Cell Biology* 27, 3612.

Companion Products:

RNase Inhibitor, Murine
#M0314S 3,000 units
#M0314L 15,000 units

RNase Inhibitor, Human Placenta
#M0307S 2,000 units
#M0307L 10,000 units

NEW ENGLAND BIOLABS® is a registered trademark of New England Biolabs, Inc.

This product is intended for research purposes only. This product is not intended to be used for therapeutic or diagnostic purposes in humans or animals.

Page 2 (M0337)