Poly(U) Polymerase

M0337S

60 units 2,000 U/ml Lot: 0031301
RECOMBINANT Store at –20°C Exp: 1/15

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

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 3H 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
   - UTP 0.5 mM final
   - RNA >100 pmol
   - RNase Inhibitor* (40 units/µl) 1 µl
   - Poly(U) Polymerase (2 units/µl) 1 µl
   - H2O to 25µl
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 µg of supercoiled plasmid for 4 hours at 37°C resulted in <10% conversion to nicked molecules as determined by agarose gel electrophoresis.

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

(see other side)
References:

Companion Products:
Murine RNase Inhibitor
#M0314S 3,000 units
#M0314L 15,000 units

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