

T4 RNA
Ligase

S

E349

1,000 units
10,000 u/ml

Lot: 10

Exp: 07/20

Store at -20C

Source:

An *E.coli* strain that carries the cloned RNA Ligase gene from bacteriophage T4.

T4 RNA Ligase catalyzes ligation of a 5' phosphoryl-terminated nucleic acid donor to a 3' hydroxyl-terminated nucleic acid acceptor through the formation of a 3'->5' phosphodiester bond, with hydrolysis of ATP to AMP and P_i. Substrates include single-stranded RNA and DNA as well as dinucleoside pyrophosphates.

Storage Conditions:

10 mM Tris-HCl (pH 7.4), 50 mM KCl, 1 mM DTT, 0,1 mM EDTA, 50% glycerol.
Store at -20° C.

1X SE-T4 RNA Ligase Buffer:

50 mM Tris-HCl, (pH 7.8@ 25° C), 10 mM MgCl₂, 10 mM DTT, 1 mM ATP.

Applications:

-labeling of 3'-termini of RNA with 5'-[³²P] pCp;
-inter- and intra -molecular joining of RNA and DNA molecules.

Unit Definition:

One unit is defined as the amount of enzyme required to convert 1 pmol of [³H]ATP in AMP-ligase4 complex in 15 minutes at 25° C.

Quality Control Assays:

Nonspecific endonuclease assay:
No appearance of nicked DNA was detected after incubation of 1µg supercoiled pUC19 DNA with 10 units of enzyme for 4 hours at 37° C.
No alteration of the pattern of DNA bands was detected after incubation of 1µg λ/HindIII DNA fragments with 10 units of enzyme in 50 µl of reaction mixture for 16 hours at 37° C.

Oligonucleotide Assay:

No detectable degradation of a single-stranded and double-stranded deoxyribooligonucleotides was observed after incubation with 10 units of enzyme for 3 hours at 37° C.

Test for detection of RNase contaminants:

No fluorescence increasing was detected after incubation of 10 units of enzyme with 0.1 mM of fluorescent-labeled deoxyribooligonucleotide including ribonucleotide for 30 min at 37° C.
(B.R.Kelemen, T.A. Klink, M.A. Behlke, S.R.Eubanks, P.A. Leland, R.T. Raines. 1999. *Nucleic Acids Res.*, 27, P. 3696-3701).

Reagents Supplied with Enzyme:

10X SE-T4 RNA Ligase Buffer.

