T4 RNA Ligase 2, truncated

2,000 units 200,000 U/ml Lot: 0091501
RECOMBINANT Store at –20°C Exp: 1/17

Description: T4 RNA Ligase 2, truncated (T4 Rnl2tr) specifically ligates the pre-adenylated 5’ end of DNA or RNA to the 3’ end of RNA. The enzyme does not require ATP for ligation but does need the pre-adenylated substrate. T4 Rnl2tr is expressed from a plasmid in E. coli which encodes the first 249 amino acids of the full length T4 RNA Ligase 2. Unlike the full length ligase, T4 Rnl2tr cannot ligate the phosphorylated 5’ end of RNA or DNA to the 3’ end of RNA (1–3). This enzyme, also known as Rnl2 (1–249) has been used for optimized linker ligation for the cloning of microRNAs. This enzyme reduces background ligation because it can only use pre-adenylated primers oligonucleotides (4–5).

Source: An E. coli strain that carries the truncated T4 RNA Ligase 2 gene.

Applications:
- Ligate a pre-adenylated DNA or RNA sequence tag to any RNA 3’-end
- Join a single stranded adenylated primer to small RNAs for cDNA library construction
- Join a single stranded adenylated primer to RNA for strand-specific cDNA library construction

Reagents Supplied with Enzyme:
10X T4 RNA Ligase Reaction Buffer and 50% PEG 8000.

Reaction Conditions: 1X T4 RNA Ligase Reaction Buffer. Incubate at 25°C.

Molecular Weight: 28,284.33 daltons
Specific Activity: 500,000 U/mg
Molarity: 14 µM
Heat Inactivation: 65°C for 20 minutes

Quality Control Assays
RNase Assay: A 10 µl reaction in T4 RNA Ligase Reaction Buffer containing 40 ng of labeled RNA and 200 units of T4 Rnl2tr was incubated at 25°C.

DNA Endonuclease Activity: Incubation of a 50 µl reaction containing 200 units of T4 Rnl2tr with 1 µg of a mixture of single and double-stranded “H E. coli” DNA (200,000 cpmp/µg) for 4 hours at 37°C resulted in <10% conversion to RF II as determined by agarose gel electrophoresis.

Certificate of Analysis

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Certificate of Analysis
Phosphatase Activity: Incubation of 200 units of enzyme with 1 µg \( p \)-nitrophenyl phosphate (PNPP) in 50 µl T4 RNA Ligase Reaction Buffer for 3 hours at 37°C released less than 0.05 µmol inorganic phosphate.

References: