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T4 RNA Ligase II (High Concentration)

Catalog # TRL-BE104

Product Name	Catalog	Size
T4 RNA Ligase 2 (High Concentration)	TRL-BE104-A	5000 U
	TRL-BE104-B	50 kU

Product Description

T4 RNA Ligase II, also known as T4 Rnl2, is an ATP-dependent RNA ligase catalyzing the inter- and intramolecular RNA strand joining activity via phosphodiester bond formation. Unlike T4 RNA Ligase I, T4 RNA Ligase II is much more active in joining nicks on double-stranded RNA (dsRNA) than joining the ends of single-stranded RNA. The enzyme can also ligate the 3´ OH of RNA to the 5´ phosphate of DNA in a double-stranded structure. The enzyme requires an adjacent 5´ phosphate and 3´ OH for ligation.

Specifications

Components	TRL-BE104-A (5000 U)	TRL-BE104-B (50 kU)
T4 RNA Ligase 2 (High Concentration) (50 U/µl)	TRL-BE104-A1 (100 µl)	TRL-BE104-B1 (1 ml)
10×T4 Rnl2 Reaction Buffer	TRL-BE104-A2 (500 µl)	TRL-BE104-B2 (5 ml)

Storage/Transport Store at -20°C ± 5°C for up to 24 months. Avoid repeated freeze/thaw cycles. Transport on dry ice.

Form Liquid (50U/µL)

Source *E. coli*

Storage Buffer 10 mM Tris-HCl, 100 mM NaCl, 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, pH 7.5

10X T4 Rnl2 Reaction Buffer 500 mM Tris-HCl, 100 mM MgCl2, 50 mM DTT and 10 mM ATP, pH 7.6

Unit Definition One unit is defined as the amount of enzyme required to ligate 0.4 µg of an equimolar mix of a 23-mer and 17-mer RNAs in a total reaction volume of 20 µL in 30 minutes at 37°C.

Applications

- Joining nicks on double-stranded RNA
- Joining 5’ phosphate of oligodeoxyribonucleotides and 3’OH of oligoribonucleotides

Recommended protocol for joining nicks on dsRNA

1. *Preparation of nicked dsRNA substrates.* Heat the RNA mixtures (at equal molar ratio) at 65°C for 3 minutes. Then incubate on ice bath for 2 minutes.
2. *Nick ligation in dsRNA.* Prepare the following reaction mixture:

Reagent	Volume
Nuclease-free H_2O	Up to 20 µL
10X T4 Rnl2 Reaction Buffer	2 µL
Nicked dsRNA substrate	2 µL
T4 RNA Ligase II (High Concentration)(50 U/µl)	0.2 µL

   a. Mix gently and incubate at 25°C for 1 hour.
   b. Add Protease K or EDTA to stop the reaction.

Notes

1. It is not recommended to heat inactivate T4 RNA Ligase II at 85°C for 5 minutes, as this may denature dsRNA.