

# T4 RNA Ligase II

## Catalog # TRL-BE103

Product Component	Sizes
T4 RNA Ligase II (10U/μL)	1000U / 10kU
10X T4 Rnl2 Reaction Buffer	500μL / 5mL

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

**Form** Liquid (10U/μL)

**Source** *E. coli*

**Concentration** 10U/μL

**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 MgCl<sub>2</sub>, 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.

### 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.

### 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 <sub>2</sub> O	Up to 20 μL
10X T4 Rnl2 Reaction Buffer	2 μL
Nicked dsRNA substrate	2 μL
T4 RNA Ligase II	1 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.