

Product Components

Components	Component number	Concentration	Size-1 1,000 U	Size -2 10,000 U
T4 RNA Ligase 2, truncated	RM21509	200,000 U/mL	5 µL	50 µL
10X T4 RNA Ligase Reaction Buffer	RM20146	10X	500 µL	500 µL
50% PEG 8000	RM20133	50%	500 µL	1.25 mL

Product Description

T4 RNA Ligase 2, truncated (T4 Rnl2 truncated) 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 Rnl2 truncated 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 Rnl2 truncated is unable to adenylate the 5' end of the substrate, and as a result it cannot ligate the phosphorylated 5' end of RNA or DNA to the 3' end of RNA. 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 adenylated primers.

Application Features

- ◆ 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 creation
- ◆ Join a single stranded adenylated primer to RNA for strand-specific cDNA library construction

Product Source

An *E. coli* strain that carries the cloned truncated T4 RNA Ligase 2 gene.

Unit Definition

200 units is defined as the amount of enzyme required to give 80% ligation of a 31-mer RNA to the pre-adenylated end of a 17-mer DNA Universal miRNA Cloning Linker in a total reaction volume of 20 µl in 1 hour at 25°C.

Storage Buffer

10 mM Tris-HCl, 100 mM NaCl, 0.1 mM DTT, 0.1 mM EDTA, 50% Glycerol, pH 7.5 @ 25°C

Storage Temperature

-20°C.

Reaction Conditions

1X T4 RNA Ligase Reaction Buffer, Incubate at 25°C

1X T4 RNA Ligase Reaction Buffer

50 mM Tris-HCl, 10 mM MgCl₂, 1 mM DTT, pH 7.5 @ 25°C

Heat Inactivation

65°C for 20 minutes.