

  
Simplifying Genomics

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# T4 RNA Ligase 2, Deletion Mutant

Cat. Nos. LR2D1132K and LR2D11310K

## 1. Introduction

T4 RNA Ligase 2, Deletion Mutant, also known as T4Rnl2(1-249), is used to ligate single-stranded adenylated DNA or RNA oligonucleotides to small RNAs for cloning or next-generation RNA sequencing. The preadenylated 5' ends of DNA or RNA are ligated to the 3' ends of RNA. Unlike the full-length enzyme, T4Rnl2(1-249) is unable to adenylate the 5' end of the substrate in the presence of ATP. However, it can use a preactivated donor (App-DNA or App-RNA) and join it to the 3' end of an acceptor; thus, performing the ligation reaction in the absence of ATP prevents circularization and other undesirable bimolecular reactions.

The enzyme is active in a pH range of 6.0 to 8.0.

## 2. Product Designations and Kit Components

Product	Kit Size	Catalog Number	Reagent Description	Part Numbers	Volume
T4 RNA Ligase 2, Deletion Mutant	2,000 Units	LR2D1132K	T4 RNA Ligase 2 Deletion Mutant (200 U/μl)	E0132-200D1	10 μL
			10X T4 RNA Ligase 2 Deletion Mutant Reaction Buffer	SS001310-D1	40 μL
	10,000 Units	LR2D1132K	T4 RNA Ligase 2 Deletion Mutant (200 U/μl)	E0132-200D2	50 μL
			10X T4 RNA Ligase 2 Deletion Mutant Reaction Buffer	SS001310-D1	200 μL

## 3. Product Specifications

**Storage:** Store only at  $-20^{\circ}\text{C}$  in a freezer without a defrost cycle.

**Storage Buffer:** T4 RNA Ligase 2, Deletion Mutant, is supplied in a 50% glycerol solution containing 50 mM Tris-HCl (pH 7.5), 0.1 M NaCl, 0.1 mM EDTA, 1 mM dithiothreitol (DTT), and 0.1% Triton® X-100.

**Unit Definition:** One unit is the amount of enzyme required to give 50% ligation of a 22-mer RNA to the preadenylated end of a 17-mer DNA when both oligos are annealed to a complementary 39-mer DNA strand in 30 minutes at  $37^{\circ}\text{C}$  under standard assay conditions.

**Activity Assay:** The unit definition assay is performed in a reaction containing 50 mM Tris-HCl (pH 7.5), 2 mM  $\text{MgCl}_2$ , 1 mM DTT, and 0.4 μg of an equimolar mix of the 22-mer, 17-mer, and 39-mer oligonucleotides, and varying amounts of enzyme.

**T4 RNA Ligase 2, Deletion Mutant, 10X Reaction Buffer:** 500 mM Tris-HCl (pH 7.5), 20 mM  $\text{MgCl}_2$ , and 10 mM DTT

**Contaminating Activity Assays:** T4 RNA Ligase 2, Deletion Mutant, is free of detectable DNA exo- and endonuclease, and RNase activities.

## 4. Applications

- Preparing cDNA libraries for small-RNA transcriptome analysis.
- Providing optimal linker ligation for miRNA cloning.

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