



Gene to Protein Pvt. Ltd.

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www.genetoprotein.com

info@genetoprotein.com

800 GENOME, 800 GENETIC

T4 RNA Ligase

Cat # ME349

Pack Size: 250U

Storage: -20°C



Introduction

T4 RNA Ligase is an enzyme that can join RNA molecules with either a 5'-phosphate or a 3'-hydroxyl group to RNA or DNA molecules with a 3'-hydroxyl and a 5'-phosphate end, respectively. Here is a data sheet for T4 RNA Ligase:

Storage buffer

The enzyme is supplied in a storage buffer containing 50 mM Tris-HCl, pH 7.5, 0.1 mM EDTA, 1 mM DTT, 50% glycerol.

Unit definition

One unit is defined as the amount of enzyme required to catalyze the ligation of 50% of the labeled RNA oligonucleotide substrate (5' -pCuApCpGpA-3') to an unlabeled RNA oligonucleotide (5' -OH-GUCUGUUCG-3') in a total reaction volume of 20 µl in 30 minutes at 25°C.

Reaction Condition

T4 RNA Ligase

RNA or DNA substrate with a 5'-phosphate or 3'-hydroxyl group

RNA or DNA substrate with a 3'-hydroxyl or 5'-phosphate end

Reaction buffer (50 mM Tris-HCl, pH 7.5, 10 mM MgCl₂, 1 mM ATP)

Incubation at 25°C for 10-60 minutes

Protocol

1. Thaw the T4 RNA Ligase on ice.
Prepare the reaction mix as follows:
Component Volume per reaction (µl)
RNA or DNA substrate variable
Reaction buffer 5
T4 RNA Ligase 1
Water variable
2. Mix the reaction thoroughly and incubate at 25°C for 10-60 minutes, depending on the amount and type of substrate used.
3. Inactivate the enzyme by heating the reaction at 65°C for 10 minutes.
4. Use the ligated RNA or DNA as required for downstream applications.
5. Note: The optimal reaction conditions may vary depending on the specific substrates and the desired application. The incubation time, temperature, and enzyme and substrate concentrations may need to be optimized for each specific reaction. The enzyme should be stored at -20°C and thawed on ice before use.

