T4 Ligase

Shipping: On Dry/Blue Ice

Exp. Date: See vial

Batch No.: See vial

Catalog numbers

BIO-27026: 500u (5u/µl)

A Meridian Life Science® Company

Store at –20°C

Storage and stability:
T4 Ligase is shipped on Dry/Blue Ice. It should be stored at -20°C upon receipt. When stored under optimum conditions, the reagents are stable for a minimum of 12 months from date of purchase.

Storage and Dilution Buffer:
10mM Tris-HCl, pH 7.4, 100mM NaCl, 1mM DTT, 0.1mM EDTA and 50% glycerol.

Q/C Assay Conditions:
Complete ligation of cohesive-ended *Hind* III fragments was achieved using only 0.01 units of enzyme per mg of Lambda DNA when incubated for 30 minutes at 16°C. Efficient ligation of blunt -ended *Hae*III fragments was achieved using 1.0 units of enzyme per mg of DNA when incubated for 30 minutes at 16°C.

Unit Definition:
One Weiss unit is defined as the amount of enzyme required to catalyze the conversion of 1nmole of ^{32}P pyrophosphate into Norit-adsorbable material in 66mM Tris HCl (pH 7.6), 6mM $MgCl_2$, 1mM DTT, 0.066mM ATP and 3.3mM [^{32}PPi] at 37°C for 20 minutes (2). 0.014 Weiss unit of ligase is equivalent to 1 cohesive end unit which catalyzes greater than 95% ligation of 1mg of Lambda/*Hind*III fragments at 16°C in 20 minutes.

Notes:
For Research Use Only.

Description

T4 DNA Ligase catalyzes the joining of two strands of DNA between the 5'-phosphate and the 3'-hydroxyl groups of adjacent nucleotides in either a blunt-ended or cohesive-ended configuration. T4 DNA Ligase catalyzes the joining of RNA to either DNA or RNA strands in a duplex molecule but will not join single-stranded nucleic acids. T4 DNA Ligase is ATP dependent.

Features

- Catalyzes the joining of double-stranded DNA
- Supplied with 10x reaction buffer and ATP
- No loss of transformation efficiency

Applications

- Ligation of cohesive and blunt-ended DNA fragments for cloning
- Sealing nicks in double-stranded DNA
- Ligation of synthetic linkers to blunt-ended DNA

Components

Product Name	500 Units
T4 DNA Ligase	100µl
10x Reaction Buffer	1.2ml
ATP Solution	1.2ml

Reagent Specifications:
10x Reaction Buffer: 660mM Tris-HCl (pH 7.6), 50mM $MgCl_2$, 100mM DTT.
Separate ATP Solution: 10mM ATP in 50mM Tris-HCl (pH 7.5).

Reaction Conditions (for a 20µl volume)

10x T4 Ligase Reaction Buffer	2.0µl
10x ATP Solution	2.0µl
Vector	as required
Insert	as required
Enzyme	1.0–1.5µl
Water (ddH_2O)	up to 20µl

For blunt ended DNA, run the DNA ligation for 1-2 hours at room temperature. For 3'-dA ends, incubate the reaction for 16-24 hours at 12-14°C.

The Total DNA concentration should be no more than 100ng.

The recommended **Insert : Vector** ratio should be between 6 and 2. A smaller ratio will result in a less efficient ligation, whilst a higher ratio will incite multiple insertions.

This data is intended for use as a guide only; conditions will vary from reaction to reaction and may need optimization.

Associated products

Product Name	Pack size	Cat. No.
Quick-Stick Ligase	50 Reactions	BIO-27027
X-GAL	1g	BIO-37035
α-Select Bronze Competent Cells	2ml (10 x 200µl)	BIO-85025
BIO*Blue* Competent Cells	1ml (10 x 100µl)	BIO-85036

Citations:

1. Lopez, M., *et al. Am. J. Enol. Vitic.* **60(2)**, 215-222 (2009).
2. Junghwa An, *et al. Conservation Genetics USA* **5(1)**, 121-125 (2004).
3. Garaizar, J., *et al. Appl. & Environ Microbiol* **66(12)**, 5273-5281 (2000).
4. Normand-Sdiqui, N., Ahktar, S.,D *Int. J Pharmaceuticals* **163(1-2)**, 63-71 (1998).

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