

T4 DNA Ligase

02-050 20,000 U (400U/μl), 02-050-5 5 X 20,000 U (400U/μl)

Bacteriophage T4 derived DNA ligase catalyzes the formation of phosphodiester bonds between 3'-OH termini and 5'-P termini in duplex DNA or RNA (1). This enzyme will join blunt end and cohesive end termini as well as repair single stranded nicks in duplex DNA, RNA or DNA/RNA hybrids.

T4 DNA ligase was expressed in *E.coli* in large quantities and highly purified. MW is 55.3 kDa.

Applications:

- 1) Insertion of DNA fragment into a vector
- 2) Linker (or Adaptor) ligation with DNA fragment

Storage conditions:

10mM Tris-HCl (pH 7.6), 50mM KCl, 0.1mM EDTA, 1mM dithiothreitol, 50% glycerol Store at -20°C

Concentration:

400 U/µl, where one unit is the amount of enzyme that ligates more than 90% of 6 µg of λ DNA-HindIII fragments in a 20µl mixture in 30 minutes at 16°C.

Quality Assurance:

Greater than 95% protein determined by SDS-PAGE (CBB staining)

The absence of endonucleases and exonucleases was confirmed.

Reagents Supplied with Enzyme:

10 x T4 Ligase Reaction Buffer (T4-Lig): 500mM Tris-HCl (pH 7.6), 100mM MgCl₂, 10 mM ATP, 100mM dithiothreitol (02-T4b 1.25ml)

Data Link: Swiss-Prot P00970

References:

 Weiss, B. et al. (1968) "Enzymatic breakage and joining of deoxyribonucleic acid." J. Biol. Chem. 243: 4543-4555 PMID: 4879167

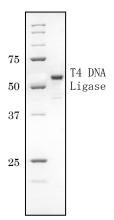


Fig.1 SDS-PAGE of T4 DNA ligase protein



Fig.2 DNA ligation activity Ligation of Hind III fragments of λ DNA using 400 unit of T4 DNA ligase Incubation at 16°C for 0, 10, 20, 30, min.