

T4 DNA ligase

Cat. No. CMC12 **Lot. No.** (See product label)

SPECIFICATION

Product Overview

Source	E.coli
Description	T4 DNA Ligase is a recombinant enzyme formulated and tested for high activity in blunt- and cohesive-end ligations for cloning and linker addition. The enzyme covalently joins 5"-phosphate and 3"-hydroxyl termini in duplex DNA or RNA. It is free of detectable exo- or endonuclease activities.
Applications	Cloning of restriction fragments; Joining linkers and adapters to blunt-ended DNA.
Unit Definition (Cohesive End Ligation Unit)	One unit is defined as the amount of enzyme required to give 50% ligation of Hind III fragments of lambda DNA (5" DNA termini concentration of 0.12μM, 300μg/ml) in a total reaction volume of 20μl in 30 minutes at 16°C in 1× T4 DNA Ligase Reaction Buffer. One Cohesive End Ligation Unit equals 0.015 Weiss (ATP-PP exchange) units, or one Weiss unit equals 67 Cohesive End Ligation Units.
Concentration	400,000 units/ml.
Quality Control Tests	Activity, SDS-PAGE (purity), DNase, endonuclease/nickase, RNase.
Storage	T4 DNA ligase in 10mM Tris-HCl, 50mM KCl, 1mM dithiothreitol, 0.1mM EDTA, 0.2 mg/ml BSA, 50% glycerol, pH 7.4 (25°C) should be stored at -20°C.
Heat Inactivation	65°C for 10 minutes.

 Tel: 1-631-559-9269 1-516-512-3133

 Email: info@creative-biomart.com  Fax: 1-631-938-8127

 45-1 Ramsey Road, Shirley, NY 11967, USA

1× T4 DNA Ligase Reaction Buffer	50mM Tris-HCl, 10mM MgCl ₂ , 1mM ATP, 10mM dithiothreitol, 25µg/ml BSA, pH7.5 (25°C)).
Reaction Conditions	1× T4 DNA Ligase Reaction Buffer, incubate at 16°C. For convenience, ligations may be done at room temperature (20-25°C). For cohesive (sticky) ends, use 1µl of T4 DNA Ligase in a 20µl reaction for 10 minutes. For blunt ends, use 1µl of T4 DNA Ligase in a 20µl reaction for 2 hours.

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