

T 4 DNA Ligase

Applications:

- Cloning of restriction fragments
- joining linkers and adapters to blunt-ended DNA
- gene (gene fragments) synthesis.

Description:

T4 DNA Ligase catalyzes the formation of a phosphodiester bonds between 5' phosphate and 3' hydroxyl termini in duplex DNA/RNA. This enzyme can join blunt end and cohesive end termini, repair single stranded nicks in duplex DNA, RNA, or DNA/RNA hybrids.

Concentration: 100-200 u/ μ l

Source:

Isolated from E.coli strain that carries the cloned DNA ligase gene from bacteriophage T4

Usage:

For most cohesive end ligations, a 30 minute incubation at 20°C is sufficient. Incubations at 16°C for 4-16 hours are routinely used for the majority of applications. Ligation of blunt ends and single-base pair overhang fragments requires more enzyme to achieve the same extent of ligation as cohesive end DNA fragments. Ligation may be enhanced by addition of PEG, or by reducing the rATP concentration. ATP is an essential cofactor for the reaction.

Storage buffer:

50 mM KCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM DTT, 50% glycerol.

Unit definition:

One unit is defined as the amount of enzyme required to give 50% ligation of Hind III fragments of lambda DNA in 30 minutes at 16°C at 5' termini concentration of 0.12 μ M (300 μ g/ml). One Cohesive End Ligation Unit equals 0.015 Weiss units. One Weiss unit equals 67 Cohesive End Ligation Units.

Reaction buffer (10X):

500 mM Tris HCL (pH 7,8), 100 mM MgCl₂, 100 mM DTT, 10 mM ATP.

Quality Assurance:

Free of contaminating exonuclease and endonuclease

Storage: shipped on blue ice, 24 months at -20°C

Ordering information:

Cat.-no	Description	Amount
402-002	T 4 DNA Ligase (weiss units)	2000 units
402-010	T 4 DNA Ligase (weiss units)	10.000 units

Related products: