DNA Ligase

(*Escherichia coli*)

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Cat. No.	Size
E1065-01	500 units
E1065-02	2 500 units

Unit Definition: One unit is defined as the amount of enzyme required to yield 50% ligation of Hind III fragments of lambda DNA. Incubation is at 16°C in 20 µl of assay mixture with a DNA terminus concentration of 0.02 µM (50 µg/ml).

Storage Conditions:
Store at -20˚C

DNA Ligase (*E.coli*) catalyzes the formation of a phosphodiester bond between juxtaposed 5 phosphate and 3 hydroxyl cohesive termini in duplex DNA.

Description:

- Catalyzes the formation of a phosphodiester bond between duplex DNA fragments with cohesive ends.
- Condensation of the 5'-phosphoryl group with an adjacent 3'-hydroxyl group is coupled with the hydrolysis of NAD^+.
- Suitable for high-efficiency cloning of full-length cDNA (1, 2).

Reaction buffer:

30 mM Tris-HCl (pH 8.0 at 22°C), 1 mM dithiothreitol, 4 mM $MgCl_2$, 26 µM NAD^+, 50 µg/ml bovine serum albumin.
Optimal ligation occurs at 16°C.

Storage Buffer:

10 mM Tris-HCl (pH 7.4 at 22°C), 50 mM KCl, 0.1 mM EDTA, 10 mM ammonium sulfate, 1 mM dithiothreitol and 50% (v/v) glycerol.

Ligation Assay Conditions:

30 mM Tris-HCl (pH 8.0 at 22°C), 4 mM $MgCl_2$, 1.2 mM EDTA, 1 mM dithiothreitol, 0.026 mM NAD^+, 50 µg/ml bovine serum albumin and Hind III fragments of lambda DNA. Incubation is at 16°C for 30 min.

Quality Control:

All preparations are tested for contaminating endonuclease and exonuclease activities, along with functional testing in the ligation reaction.

References:

1. *Okayama, H. and Berg, P. (1982) Mol. Cell. Biol. 2, 161-170.*
2. *Gubler, U and Hoffman, B.J. (1983) Gene 25, 263-269.*