T4 DNA Ligase

ArcticZymes T4 DNA Ligase is an ATP and Mg^{2+} dependent dsDNA ligase which catalyses the formation of a phosphodiester bond between 3'-hydroxyl and 5'-phosphate termini in duplex DNA, duplex RNA, and some DNA/RNA hybrids. The enzyme is recombinantly produced in *E. coli*.

T4 DNA Ligase is active on both blunt-end and cohesive end substrates. It is also completely inactivated by incubating at 70°C for 10 minutes.

T4 DNA Ligase is extensively tested for contaminating DNase and RNase activities as well as residual host-cell gDNA.

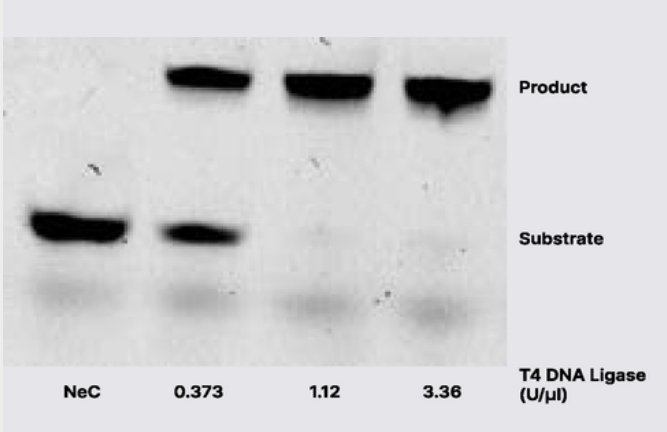

Fig 1. T4 DNA Ligase displays good nick-joining activity of dsDNA

T4 DNA Ligase activity on various substrates. *In vitro* assays of ligation activity were performed using three oligos hybridised into a nicked 20 bp dsDNA substrate. Efficient DNA ligation was observed after 30 min at 25°C when using 1.12 U/µl T4 DNA Ligase. At 0.373 U/µl T4 DNA Ligase showed about 70% substrate turnover.

Quality control

Optimal reaction conditions	50 mM Tris-HCl (pH 7.5 at 25°C), 10 mM DTT, 5 mM $MgCl_2$ and 1 mM ATP.
Storage buffer	10 mM Tris-HCl pH 7.5 at 25°C 50 mM KCl 1 mM DTT 0.1 mM EDTA 50% (v/v) Glycerol
Stability	The enzyme is stable at -20°C for two years in the supplied storage buffer. The enzyme tolerates a minimum of four freeze-thaw cycles (-80°C) without loss of activity

Quality control

dsDNA endonuclease activity	10 000 U T4 DNA Ligase was incubated with a supercoiled plasmid (1 µg) for 4 hours at 37°C. Agarose gel electrophoresis did not reveal any transformation of closed circular DNA to nicked DNA.
ssDNA endonuclease activity	10 000 U T4 DNA Ligase was incubated with M13 ssDNA (0.5 µg) for 4 hours at 37°C. Agarose gel electrophoresis did not reveal any visible signs of ssDNA degradation.
Exonuclease activity	10 000 U T4 DNA Ligase was incubated with either 3H-dATP labelled ds or ssDNA (0.5 µg, 500 bp) for 4 hours at 37°C. Acid soluble radioactivity from labelled DNA was not significantly over blank test for either substrate.
RNase activity	5000 U T4 DNA Ligase was incubated with a 2 kb RNA transcript (1 µg) for 4 hours at 37°C. Agarose gel electrophoresis did not reveal any visible signs of RNA degradation.
***E. coli* gDNA contamination**	5000 U T4 DNA Ligase was analysed in a probe-based qPCR assay detecting the 23S ribosomal subunit in *E. coli*. No *E. coli* gDNA could be detected (LOD: < 3 *E. coli* genomic copies.).

Ordering Information

	Article no.	Pack Size*	Concentration
T4 DNA Ligase	71800-202	250 kU	≥ 5000 U/µl
	71800-100	Custom	Custom

* 0.1 units is defined as the amount of enzyme that is needed to convert 1 pmol (of 18 pmol) of nicked DNA substrate in 20 minutes at 25°C in a 20 µl reaction volume in a buffer consisting of 62.5 mM Tris-HCl pH 7.5, 10 mM DTT, 1 mM ATP, 0.05 mg/ml BSA and 25 mM KCl. One Weiss Unit is equivalent to approximately 500 ArcticZymes Units.

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Quality

ArcticZymes is dedicated to the quality of its products and is certified according to ISO 13485:2016. ArcticZymes offers the convenience of providing standard bulk enzymes as off the shelf products. In addition, ArcticZymes offers enzymes in customized formats. For additional information, please contact us.

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Additional Information

We are pleased to provide data and information relating to ArcticZymes T4 DNA Ligase. Available data includes stability, buffer storage conditions, pH, and specific activity data. For more information, please check our website www.arcticzymes.com.

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