PRODUCT INFORMATION SHEET

T4 DNA Ligase

PRODUCT SPECIFICATIONS

Product Name: Recombinant T4 DNA Ligase, RUO Grade

Product number/code: T4 DNA Ligase enzyme, His purified, 267-126

Product Description: T4 DNA Ligase is prepared using CrisBio®, a novel expression system that utilizes a recombinant baculovirus carrying a T4 Bacteriophage gene to inoculate *Trichoplusia ni* chrysalises

Source of Protein: Engineered Baculovirus carrying the T4 Bacteriophage gene inoculated in *Trichoplusia ni*

Concentration: 400,000 U/ml and 2,000,000 U/ml.

Unit Availability: 400,000 U and 100,000 U, available in both concentrations. Supplied with 10X Reaction Buffer

Unit Definition: One unit (U) is defined as the amount of enzyme required to ligate ≥ 90% of 3 µg of Lambda DNA-*Hind*III Digest in 20 min at 16°C

Shelf Life: 2 years from manufacturing date. Shelf life currently under assessment with stability studies

Storage Temp: -20°C

Limitations of Use:
For research and development use only. Not suitable for human or animal therapeutic or clinical diagnostic use. SDS sheets relevant to this product are available upon request.

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Date revised: September 2025 D-PS-T4DNALigase_v01

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Storage Buffer: 10 mM Tris-HCl pH 7.4, 50 mM KCl, 1 mM DTT, 0.1 mM EDTA and 50% Glycerol

Assay	SDS-PAGE Purity	SDS-PAGE Western Blot Identity[1]	Enzyme Activity	DNase Activity	Residual gDNA
Specification	≥ 95%	Specific Band Detection	≥ 2,000,000 U/ml; ≥ 400,000 U/ml	< RFU of 0.02 U control	≤ 100 pg/mg

[1] Performed during manufacture process

Assay	RNase Activity	Endonuclease Activity
Specification	< RFU of 3E-06 U control	Nicked band intensity ≤ low positive control

QUALITY CONTROL ANALYSIS

Enzyme Activity is determined in a 20 µl reaction in 1X Reaction Buffer containing 3 µg of Lambda DNA-*Hind*III Digest, incubated for 20 minutes at 16°C, resulting in ≥ 90% ligation of the DNA fragments as determined by agarose gel electrophoresis.

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Protein Purity is evaluated by SDS-PAGE of concentrated and diluted enzyme solutions followed by Coomassie stain. Purity is assessed by comparing the aggregate mass of non-specific bands in the concentrated sample to the mass of the protein of interest band in the diluted sample.

DNase Contamination is detected by Fluorometric Assay:
Use of a fluorogenic substrate that emits fluorescence after cleavage by the presence of DNases.

Residual (genomic) DNA Contamination is evaluated by quantitative PCR:
Amplification of specific genomic sequence of *T ni*.

RNase Activity is detected by Fluorometric Assay:
Use of a fluorogenic substrate that emits fluorescence after cleavage by the presence of RNases.

Endonuclease activity (nicking)
A 50 µl reaction containing 0.5 µg of supercoiled DNA and a minimum of 40 µg of T4 DNA Ligase incubated for 4 hours at 37°C results in less nicked product than the lower positive control, determined by agarose gel electrophoresis.

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