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Product Information Sheet

Product Information	
***Taq* DNA Ligase**	
Part Number	L6060L 280260
Concentration	40,000 U/mL
Unit Size	20,000 U

Product Specification	
Storage Temperature	-25°C to -15°C
TEST:	**SPECIFICATION:**
Purity (SDS-PAGE)	>99%
Specific Activity	400,000 U/mg
SS Exonuclease	400 U <5.0% released
DS Exonuclease	400 U <1.0% released
DS Endonuclease	400 U = No conversion
***E.coli* DNA Contamination**	400 U <10 copies

Product Description:

Taq DNA Ligase catalyzes the formation of a phosphodiester bond in duplex DNA containing adjacent 5'-phosphoryl and 3'-hydroxyl termini, using NAD^+ as a cofactor.

Source of Protein

A recombinant *E. coli* strain carrying the cloned *Taq* DNA Ligase gene.

Supplied In

10 mM Tris-HCl
50 mM KCl
1 mM DTT
0.1 mM EDTA
0.1% Tween-20
50% glycerol
pH 7.5 @ 25°C

Supplied with

B6060 (10X *Taq* DNA Ligase Buffer)

10X *Taq* DNA Ligase Buffer (B6060):

200 mM Tris-HCl
250 mM KCl
100 mM $MgCl_2$
5 mM NAD
0.1% Triton X-100
pH 7.6 @ 25°C

Unit Definition

1 unit is defined as the amount of *Taq* DNA Ligase required to join 50% of 1 µg of the 12-base cohesive ends of Lambda DNA cut with Hind III in 50 µl 1X *Taq* DNA Ligase Buffer following a 10 minute incubation at 45°C.

Quality Control Analysis:

Unit Characterization Assay

Unit activity was measured using a 2-fold serial dilution method. Dilutions of enzyme batch were made in 1X Taq DNA Ligase Reaction Buffer ($[Taq\ DNA\ Ligase]_f$ = 0.1-0.0008 µg/µL) and added to 50 µL reactions containing 1.0 µg DNA and 1X *Taq* DNA Ligase Reaction Buffer. Reactions are incubated for 10 minutes at 45°C, stopped, and analyzed on a 0.8% agarose gel stained with ethidium bromide.

SDS-Page (Physical Purity Assessment) and Specifications

2.0 µL of concentrated enzyme solution was loaded on a denaturing 4-20% Tris-Glycine SDS-PAGE gel flanked by a broad-range MW marker and 2.0 µL of a 1:100 dilution of the same enzyme species. Following electrophoresis, the gel was stained and the samples compared to determine physical purity. The acceptance criteria for this test requires that the aggregate mass of contaminant bands in the concentrated sample do not exceed the mass of the protein of interest band in the dilute sample, confirming greater than 99% purity of the concentrated sample.

Contamination Tests:

Single-Stranded Exonuclease Activity

A 50 µL reaction containing 11,000 cpm of a radiolabeled single-stranded DNA substrate and 10 µL of enzyme solution incubated for 4 hours at 37°C resulted in less than 5.0% release of TCA-soluble counts.

Double-Stranded Exonuclease Activity
A 50 µl reaction containing 5,000 cpm of a radiolabeled double-stranded DNA substrate and 10 µL of enzyme solution incubated for 4 hours at 37°C resulted in less than 1.0% release of TCA-soluble counts.

Double-Stranded Endonuclease Activity
A 50 µL reaction containing 0.5 µg of pENZuC DNA and 10 µL of enzyme solution incubated for 4 hours at 37°C resulted in no visually discernible conversion to nicked circular DNA as determined by agarose gel electrophoresis.

E. coli 16S rDNA Contamination test
Replicate 5 µL samples of enzyme solution were denatured and screened in a TaqMan qPCR assay for the presence of contaminating *E.coli* genomic DNA using oligonucleotide primers corresponding to the 16S rRNA locus. The acceptance criterion for the test is the threshold cycle count (C_t) produced by the average of 3 replicate no template control samples. Based on the correlation between the no template control C_t values and standard curve data, the detection limit of this assay is <10 copies genome/sample.

Legal Disclaimer:

Certain applications in which this product can be used may be covered by patents issued and applicable in the United States and abroad. Purchase of this product does not include a license to perform any patented application; therefore it is the sole responsibility of users of this product to determine whether they may be required to engage a license agreement depending upon the particular application in which the product is used.

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