enzymatics a QIAGEN company

***Product Specifications** L6060L Rev 02*

Product Information	
***Taq* DNA Ligase**	
Part Number	L6060L
Concentration	40,000 U/mL
Unit Size	20,000 U
Storage Temperature	-25°C to -15°C
Lot Number	
Reference Number	

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 (1).

Product Specifications						
L6060						
Assay	SDS Purity	Specific Activity	SS Exonuclease	DS Exonuclease	DS Endonuclease	*E. coli* DNA Contamination
Units Tested	**n/a**	**n/a**	**400**	**400**	**400**	**400**
Specification	>99%	400,000 U/mg	<5.0% Released	<1.0% Released	No Conversion	<10 copies

Source of Protein: Recombinant *E. coli* strain carrying the cloned *Taq* DNA Ligase gene.

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 SmaI and SalI in 50 µl 1X *Taq* DNA Ligase Buffer following a 10 minutes incubation at 45°C.

Molecular weight: 76.9 kDa

Quality Control Analysis:

Unit Activity is measured using a 2-fold serial dilution method. Dilutions of enzyme batch were made in 1X Taq DNA Ligase Reaction Buffer and added to 50 µL reactions containing λ Hind III digested 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.

Protein Concentration (OD_{280}) is determined by OD_{280} absorbance.

Physical Purity is evaluated by SDS-PAGE of concentrated and diluted enzyme solutions followed by silver stain detection. Purity is assessed by comparing the aggregate mass of contaminant bands in the concentrated sample to the mass of the protein of interest band in the diluted sample.

Single-Stranded Exonuclease is determined in a 50 µL reaction containing a radiolabeled single-stranded DNA substrate and 10 µL of enzyme solution incubated for 4 hours at 37°C.

Double-Stranded Exonuclease is determined in a 50 µL reaction containing a radiolabeled double-stranded DNA substrate and 10 µL of enzyme solution incubated for 4 hours at 37°C.

Double-Stranded Endonuclease is determined in a 50 µL reaction containing 0.5 µg of plasmid DNA and 10 µL of enzyme solution incubated for 4 hours at 37°C.

***E. coli* 16S rDNA Contamination** is evaluated using 5 µL replicate samples of enzyme solution 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.

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 at 25°C)

Supplied with:
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 at 25°C)

Usage Instructions: Nick ligation in double-stranded DNA

1. Set up the following reaction mixture in a total volume of 50 µL:

Components	Final Concentration	Volume
Nuclease free water	N/A	X µL
10X Taq DNA Ligase Buffer (B6060)	1X	5 µL
DNA	up to 1µg	X µL
Taq DNA Ligase (L6060L)	80 U	2 µL
	Total Volume =	50 µL

2. Incubate at 45°C for 10 minutes.

References:
1. Takahashi, M. et al. (1984). J. Biol. Chem. 259, 10041-10047.

Disclaimer:
Use of this enzyme in certain applications may be covered by patents and may require a license. Purchase of this product does not include a license to perform any patented application; therefore, it is the sole responsibility of the users of the product to determine whether they may be required to engage in a license agreement depending upon the particular application in which the product is used.

Limitations of Use
This product was developed, manufactured, and sold for *in vitro* use only. The product is not suitable for administration to humans or animals. SDS sheets relevant to this product are available upon request.