

| Product Information | |
|---------------------|----------------|
| WGS Ligase | |
| Part Number | L6030-W-L |
| Reaction Size | 24 Reactions |
| Volume | 0.24 mL |
| Storage Temperature | -25°C to -15°C |
| Lot Number | |
| Reference Number | |

Product Description: WGS Ligase is optimized for ligation following WGS Fragmentation. It catalyzes the formation of a phosphodiester bond between the terminal 5' phosphate and a 3' hydroxyl groups of duplex DNA or RNA. The enzyme efficiently joins blunt and cohesive ends and repairs single stranded nicks in duplex RNA, RNA, or DNA/RNA hybrid (1).

| Product Specifications | | | | | | |
|------------------------|------------|-------------------|------------------|------------------|-----------------|---------------------------|
| L6030 | | | | | | |
| Assay | SDS Purity | Specific Activity | SS Exonuclease | DS Exonuclease | DS Endonuclease | E. coli DNA Contamination |
| Units Tested | n/a | n/a | 6,000 U | 6,000 U | 6,000 U | 6,000 U |
| Specification | >99% | 300,000 U/mg | < 1.0 % Released | < 1.0 % Released | No Conversion | < 10 copies |

Source of Protein: A recombinant *E.coli* strain carrying the cloned T4 DNA Ligase gene

Unit Definition: 1 unit is defined as the amount of DNA Ligase required to join 50% of 100 ng of DNA fragments with cohesive termini in 50 µl 1X DNA Ligase Buffer following a 30 minute incubation at 23°C

Molecular weight: 55,292 Daltons

Quality Control Analysis:

Unit Activity is measured using a 2-fold serial dilution method. Dilutions of enzyme batch were made in 1X DNA Ligase Reaction Buffer and added to 20 µL reactions containing double stranded DNA fragments and 1X DNA Ligase Reaction Buffer. Reactions are incubated for 30 minutes at 23°C, stopped, and analyzed on a 1% agarose gel stained with ethidium bromide.

Protein Concentration is determined by OD₂₈₀ 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.

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. MSDS sheets relevant to this product are available upon request.

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 dithiothreitol, 0.1 mM EDTA, 50% Glycerol, pH 7.5 @ 25°C

Supplied with: B9020: 330mM Tris-HCl, 50mM MgCl₂, 5mM DTT, 5mM ATP, 30% PEG 6000, pH 7.6 @ 25°C

References:

1. Engler, M.J. and Richardson, C.C. (1982) P.D. Boyer (Eds.), *The Enzymes*, 5, pp. 3. San Diego: Academic Press.

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. MSDS sheets relevant to this product are available upon request.