molecular biology GENESPIN DATA SHEET

> T4 DNA Ligase

T4 DNA Ligase (isolated and purified from an E. coli strain) catalyzes the formation of a phosphodiester bond between juxtaposed5'-phosphate and 3'-hydroxyl groups in duplex DNA or RNA. This enzyme is capable of joining adjacent nucleotides in either a blunt-ended or cohesive ended configuration, as well as repairing single-stranded nicks in duplex DNA, RNA or DNA/RNA hybrids.

cat. no.	*Weiss units*	*CE units*	*amount*	*note*
STS-TDL 100	250 Weiss units	50000 units	100ul	10X T4 Ligase Buffer
STS-TDL 1000	1250 Weiss units	250000 units	500ul	10X T4 Ligase Buffer

FOR RESEARCH USE ONLY

UNIT DEFINITION
5,9 Weiss UNITS of T4 Ligase is the amount of the enzyme required to catalyze the ligation of greater than 95% of 1µg of λ/ HindIII fragments at 16°C in 20 min.

SHIPPING
Shipped in green ice.

STORAGE
Store at -20C°

SHELF LIFE
12 months

CONCENTRATION
2.5 Weiss units/µl (500 CE units/µl)

Assay Set-Up:
Before starting, vortex all components thoroughly to ensure homogeneity.
Prepare a premix for the number of assays you need according to the following protocol:

Optimal ligation occurs at 16°C for up to 16h in 1X T4 DNA Ligase Reaction Buffer.
As little as 10 min or 2h are usually needed for ligating cohesive ends and blunt ends, respectively, when reaction is performed @ RT. Recommended 5'-DNA termini concentration is 0.1-1 µM; 1-10 µg/ml total DNA (insert plus vector) should be used for efficient ligation. Insert/vector molar ratios between 2 and 6 are optimal for single insertions. Ratios below 2/1 result in lower ligation efficiency, whereas ratios above 6/1 promote multiple inserts.

0.1 - 1 Weiss Units of T4 DNA Ligase are used
T4 DNA Ligase can be inactivated @ 65°C for 10 min.