Tinzyme Co., Limited

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Taq DNA ligase

Product Number: LG08

Shipping and Storage

-20℃ can be stored for 2 years to avoid repeated freeze-thaw cycles

Components

Component	LG08	LG08
Taq DNA Ligase	2000U	10000U
5×NAD DNA Ligase Buffer	0.3ml	1.5ml

Description

DNA ligase catalyzes the formation of phosphodiester bonds between 5 '- phosphate and 3' - hydroxyl groups in double stranded DNA, using NAD+as a coenzyme as the energy source for the reaction. Connection only occurs when oligonucleotides perfectly pair with complementary target DNA and there is no gap between them. Not active on single stranded DNA or RNA and flat terminal DNA. Therefore, single base mutations can be detected. High thermal stability allows for precise detection of SNPs under high-density hybridization conditions, with high specificity and rigor.

Our company's Tth Taq Ligase is a recombinant protein expressed and purified through multiple steps.

Concentration

40U/μl

Features

Good heat resistance

Application

1. Used for Gibson assembly method
2. dsDNA splicing repair
3. Ligase chain reaction can be used, LCR,Ligase detection reaction, detecting allele specificity

Unit definition

1 active unit refers to the amount of enzyme required to connect 50% of 1ug of λ DNA fragments (12bp sticky end) digested by BstEII within 15 minutes in a 1×Taq DNA ligase reaction buffer system at 45℃.

Activity determination conditions

20mM Tris-HCl, 25mM KAc, 10mM Mg(Ac)2, 10mM DTT, 1mM NAD 和 0.1% Triton X-100（pH 7.6 ,25℃）, 45℃ incubation。

Storage buffer

10mM Tris-HCl， 50mM KCl， 1mM DTT， 0.1 mM EDTA， 200μg/ml BSA， 50% Glycerol， pH 7.4, 25°C

Quality control

After strict quality control testing, it is ensured that the product has the highest activity and purity.

Note

1. Taq DNA ligase can effectively connect complementary fragments of 12bp, but cannot connect complementary fragments of

4bp (typical restriction enzyme digestion products).

2. Reaction conditions: Incubate DNA and enzymes in a 1×Taq DNA Igase Buffer at 45℃ or in a thermal cycler for 15 minutes, and terminate the reaction with a mixture of 50% glycerol, 50 mM EDTA, and bromophenol blue.
3. 1×Taq DNA ligase reaction buffer requires NAD+as a cofactor. $NAD^+$is provided in a 10×Taq DNA ligase buffer; The buffer should be stored at -80°C to extend the half-life of NAD+cofactors.

Protocol

Connection reaction

1. Prepare the reaction system according to the following table

Component	Volume/μl
5×NAD DNA Ligase Buffer	4
Substrate: DNA with complementary sticky ends>8nt	100ng-1μg
Taq DNA ligase (40U/μl)	1-2
H_2O	Variable
Total	20

2. 45-50°C connection for 15 minutes
3. For longer double stranded DNA substrates, agarose electrophoresis is used to detect the connecting products; For short DNA fragments shorter than 100 bp, polyacrylamide gel electrophoresis was used to detect the connecting products.