Taq DNA Ligase

Instruction for Use

Cat. No./Spec.: E1012-A/1,000 U; E1012-B/2,000 U; E1012-C/10,000 U

Concentration: 40 U/μL

Product Description

Taq DNA Ligase is a heat-resistant ligase that catalyzes the formation of phosphodiester bonds at the 5´ -phosphate and 3´ -hydroxyl ends of two adjacent DNA strands. This reaction can occur only when the two oligonucleotide chains are perfectly paired with the complementary target DNA and there is no gap between the two oligonucleotide chains. Therefore, it can be used to detect single nucleotide variants. Taq DNA ligase uses NAD as a cofactor. Taq DNA ligase was active in the range of 37-75°C.

Components

Component	*E1012-A*	*E1012-B*	*E1012-C*
Taq DNA Ligase (40 U/μL)	*25 μL*	*50 μL*	*250 μL*
10X Taq DNA Ligase Buffer	*250 μL*	*250 μL*	*1.25 ml*

Storage Condition & Shelf Life

Store at -20°C. The product is valid for 2 years.

Source

Recombinant E. coli strain containing ligase gene cloned from Thermus aquaticus HB8.

Unit Definition

1 unit is the amount of enzyme required to bind 50% of 1 μg BstEII enzyme to the 12-base pair sticky end of λ DNA for 15 minutes at 45 ° C at a total reaction volume of 50 μl.

Scope of Application

• Using ligase chain reaction (LCR) and ligase detection reaction (LDR) to specifically detect alleles

• Incorporating phosphorylated oligonucleotides during primer extension amplification for mutation detection

Protocol

1. Prepare reaction system.

Component	*Amount*
DNA	*Up to 1 μg*
Taq DNA Ligase (40 U/μL)	*5 μL*
10X Taq DNA Ligase Buffer	*2 μL*
ddH_2O	*To 50 μL*

2. Incubate at 45°C for 15 min.

Notes

1. Reaction conditions: Incubate DNA and enzyme in 1X Taq DNA Ligase Buffer at 45°C for 15 minutes; Or incubate DNA and enzymes in a thermal circulator. The reaction was terminated with a mixture of 50% glycerol, 50 mM EDTA, and bromophenol blue.

2.1X Taq DNA Ligase Buffer requires NAD+ as a cofactor. NAD+ has been added to the 10X Taq DNA Ligase Buffer. To prolong the half-life of NAD+ cofactors, the buffer should be stored at -80°C.

3.Taq DNA Ligase does not connect the short 4-base overlaps (typical restrictive endonuclease digestion), whereas it efficiently connects the 12-base pair overlaps.

This product is for research use only.