

Taq DNA Ligase

1 Packing list

Components	HY-KE8009-1000 U
Taq DNA Ligase (40 U/ μ L)	25 μ L
10 \times Taq Ligase Reaction Buffer	500 μ L

2 Introduction

This enzyme is a DNA ligase cloned from *Thermus aquaticus* HB8. It is expressed in *E. coli* using genetic recombination technology and purified and isolated multiple times. Taq DNA ligase catalyzes the formation of phosphodiester bonds, connecting the 5'-phosphate termini and 3'-hydroxy termini of two conforming oligonucleotide strands hybridized to the same complementary target DNA strand through phosphodiester bonds. This ligation reaction can only occur when the two oligonucleotide strands are completely paired with the complementary target DNA and there is no gap between the two oligonucleotide strands. Therefore, it can be used to detect single base substitutions. Taq DNA ligase is active in the range of 45 to 65°C.

3 Unit definition

In 50 μ L reaction system, the amount of enzyme required to ligate 50% of the 12 bp sticky end fragments in 15min under 45°C reaction conditions. The 12 bp sticky ends were derived from BstEII digestion of 1 μ g λ DNA.

4 General Protocol

Commonly used reaction systems (50 μ L)

Components	Adding amount
DNA	up to 1 μ g
10 \times Taq Ligase Reaction Buffer	5 μ L
Taq DNA Ligase (40 U/ μ L)	2 μ L
ddH ₂ O	up to 50 μ L

Reaction conditions: incubate at 45°C for 15min. Add stop stain solution (50% glycerol, 50 mM EDTA and bromophenol blue) to terminate the reaction.

5 Storage

-20°C, 1 years

6 Precautions

1. This enzyme requires NAD⁺ as a cofactor, and 10×Taq Ligase Reaction Buffer already contains NAD⁺. The buffer should be stored at -70°C to extend the half-life of NAD⁺.
2. 1× Taq Ligase Reaction Buffer: 20 mM Tris-HCl (pH 7.6, 25°C), 25 mM KAc, 10 mM Mg(Ac)₂, 10 mM DTT, 1 mM NAD and 0.1% Triton X-100, incubate at 45°C.
3. Taq DNA Ligase cannot replace T4 DNA ligase.
4. This product is for R&D use only, not for drug, household, or other uses.
5. For your safety and health, please wear a lab coat and disposable gloves to operate.