

Difference Between T4 and T7 DNA Ligase

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Key Difference – T4 vs T7 DNA Ligase

DNA ligase is an important **enzyme** used in molecular biology techniques. It functions as a molecular glue to join nucleotides by forming phosphodiester bonds between **nucleotides**. The phosphodiester bonds are formed between the 3' **hydroxyl** end of the sugar moiety and the 5' end **phosphate** group. DNA ligase is vital in the replication process to join the Okazaki fragments of the lagging strand and during DNA repair mechanisms and *in vitro* cloning experiments to join the desired gene of interest to the **vector** genome. There are two main DNA ligases used currently by molecular biologists: T4 and T7 DNA ligases. **T4 DNA ligase is one of the first enzymes to be isolated from the T4 bacteriophage. T7 DNA ligase, which is a smaller protein, is an enzyme isolated from T7 bacteriophage.** This is the key difference between T4 and T7 DNA ligases.

What is the Action of DNA Ligase?

DNA ligation occurs in a three-step reaction. The first step is a nucleophilic attack on the alpha-phosphorus of **ATP** generating a covalent enzyme-adenylate intermediate in which AMP is linked to the terminal side-chain nitrogen of lysine. In the second step, a nucleophilic attack occurs on the phosphorus of the enzyme-adenylate by the 5'-phosphate-terminated DNA substrate, releasing **lysine** and forming DNA-adenylate. In the final step, DNA-adenylate is attacked by 3'-OH of another DNA strand, releasing AMP and joining the polynucleotides.

What is T4 DNA Ligase?

T4 Ligase was the first ligase to be isolated and identified by Meselson, Weigle, and Kellenberger and was the first ever commercial ligase produced. T4 DNA ligase is an ATP dependent enzyme which catalyzes the phosphodiester bond formation. It is a single **polypeptide** chain with 487 amino acid residues and has a molecular weight of about 77 kDa. The optimum pH for its activity lies in the range of 7.5 – 8.

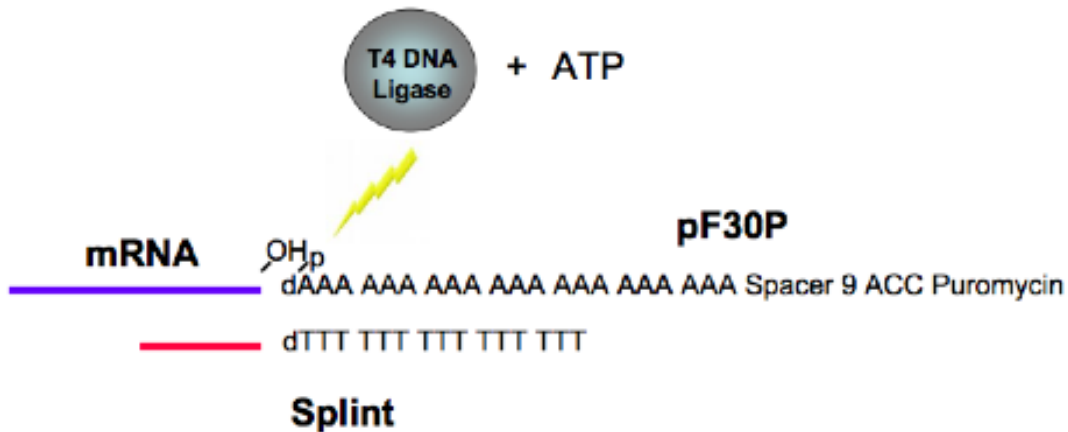


Figure 01: T4 DNA Ligase

T4 DNA Ligase, as its name suggests, is present in bacteriophage T4 and was isolated when phage produced the enzyme upon infection with *E. coli*. The enzyme ligates double stranded DNA duplexes with blunt ends and the fragments to be bound should be in close proximity to each other.

What is T7 DNA Ligase?

T7 ligase is 41 kDa protein produced by T7 Bacteriophage and is isolated by infecting *E. coli* cells. The main function of T7 ligase is to ligate DNA duplexes through phosphodiester linkages and join nearby fragments of DNA during cloning experiments. The optimum pH for T7 DNA ligase lies in the range of 7.0 – 7.2. T7 ligase is ATP dependent. T7 DNA ligases are also capable of annealing DNA and RNA hybrids, giving rise to abnormal conformations during transcription; however, this phenomenon is still under research by *in vitro* studies.

What are the similarities between T4 and T7 DNA Ligase?

- T4 and T7 DNA Ligase participate in a ligation reaction by annealing DNA duplexes through phosphodiester bond formation between 3' OH end and the 5' phosphate end of the DNA fragment.
- Both enzymes ligate blunt ended DNA duplexes which are in close proximity to each other.
- Both enzymes are ATP dependent.
- These enzymes are isolated by infecting *E. coli* cells with the particular bacteriophage.
- Applications of both enzymes include:

- Molecular cloning – to ligate the gene of interest to the vector
- DNA repair – to repair double stranded niches
- In vitro DNA replication
- Ligase chain reaction

What is the difference between T4 and T7 DNA Ligase?

T4 vs T7 DNA Ligase	
T4 DNA Ligase type of DNA ligase isolated from the bacteriophage T4 and is involved in ligating adjacent DNA fragments via phosphodiester bond formation.	T7 DNA Ligase type of DNA ligase isolated from the bacteriophage T7 and is involved in ligating adjacent DNA fragments via phosphodiester bond formation.
Size of the Protein	
Size of T4 DNA ligase is 77 kDa – larger in size compared to T7 DNA ligase	Size of T7 DNA ligase is 41 kDa.
Optimum pH	
pH range which works T4 DNA ligase is 7.5 – 8.0.	pH range which works T7 DNA ligase is 7.0 – 7.2.

Summary – T4 vs T7 DNA Ligase

DNA ligases are an important class of enzymes involved in molecular cloning to produce recombinant DNA molecules containing the gene of interest. Ligation reaction is an energy utilizing reaction and there are novel research studies underway to improve the accuracy via modulating the available ligases. T4 and T7 DNA ligases are two types of important DNA ligase enzymes used in molecular biological techniques. T7 DNA ligase is also a result of research which was based on the primary ligase isolated from T4 infected *E. coli*. T7 is portrayed more efficiency and accuracy in carrying out its function due to its smaller size. This is the difference between T4 and T7 ligase.

References:

1. “DNA Ligase, T4.” DNA Ligase, T4 – Worthington Enzyme Manual, [Available here](#). Accessed 22 Aug. 2017.
2. Doherty, Aidan J., et al. “Bacteriophage T7 DNA Ligase OVEREXPRESSION, PURIFICATION, CRYSTALLIZATION, AND CHARACTERIZATION.” Journal of Biological Chemistry, 10 May 1996, [Available here](#). Accessed 22 Aug. 2017.

Image Courtesy:

1. "Figure 3. Splint Aid Single Stranded mRNA-DNA T4 DNA Ligase Ligation." [\(CC BY 3.0\)](#) via [Commons Wikimedia](#)

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