DNA cleavage

Defined DNA cleavage was made possible by the discovery of restriction endonucleases (restrictases). These enzymes cleave dsDNA into very specific centrally symmetrical sequences of nucleotides called **palindromes** (see table). Restrictases are natural enzymes of bacteria, where they are part of the so-called **restriction-modification system**. The same sequence is methylated in the own DNA by this system, which prevents restrictases from degrading the own DNA. The system is therefore aimed at the degradation of foreign DNA whose target sequences are not protected. If the phage DNA enters the bacterium, it is mostly degraded - the phage undergoes restriction. Only exceptionally is some of the phage DNA methylated by the methylation system, the phage survives, has been modified and can reproduce in the given bacterial strain.

Target sequences of DNA restrictases

enzyme	origin	target sequence
EcoRI	*Escherichia coli*	G↓AATTC CTTAA↑G
HindIII	*Haemophilus influenzae*	A↓AGCTT TTCGA↑A
BsuI	*Bacillus subtilis*	GG↓CC CC↑GG

Specificity of restrictases has been used in DNA enzymology. Around a hundred such enzymes have been described, they are called by abbreviations of the names of the bacteria in which they were discovered (EcoRI - *E. coli*, HaeIII - *Haemophilus influenzae*). Each restrictase has a specific target sequence in which it cleaves either opposing phosphodiester bonds of dsDNA or bonds symmetrically several nucleotides away from the center of symmetry (see table). In the second case, **cohesive** ("sticky") ends of the resulting restriction fragments are formed, which can reassociate, even with fragments obtained by cleavage of other DNA with the same restrictase. This option is very advantageous when recombining fragments and constructing new genes and genomes. Today, however, it is not a problem to artificially create any cohesive ends on any dsDNA fragment.

Another biotechnologically important enzyme is DNA-ligase, discovered during the study of natural DNA replication. It covalently links the 3'-OH end of the chain to the 5'-P end of the DNA.

Links

Related Articles

- Genetic Engineering Biochemistry
- Separation of DNA fragments by electrophoresis
- Identification of restriction fragments
- Artificial DNA Synthesis
- Multiplication and expression of an isolated gene in a host cell

References

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