Advancements in Genetic Engineering

Editorial

Advancement in the Field of Gene Cloning

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Cloning is a term that most people are familiar with. A clone is a genetically identical duplicate. It may be a gene copy, a cell, or an entire organism. Even a human being. However, while human cloning raises many ethical concerns and is prohibited in most parts of the world, gene cloning has been going on for well over 30 or 40 years, with animal cloning happening more recently. Gene cloning, also known as molecular cloning, is the method of isolating a DNA sequence of interest in order to replicate it several times. Clones are close copies. Stanley Cohen and Herbert Boyer developed techniques for creating recombinant DNA, a form of synthetic DNA, in 1973. Recombinant DNA is generated by fusing two or more DNA strands together, resulting in DNA sequences that would not normally occur together. To put it another way, selected DNA (or "interest DNA") is inserted into an established organismal genome, such as bacterial plasmid DNA or another vector.

After that, the recombinant DNA can be injected into another cell, such as a bacterial cell, for amplification and probably protein development. The uptake, absorption, and expression of foreign genetic material causes a cell's genetic alteration, which is referred to as transformation. The discovery of restriction endonucleases allowed recombinant DNA technology to be developed.Restriction enzymes, also known as restriction endonucleases, are prokaryotic enzymes that recognise and cut DNA at restricted sequences. They are thought to have developed as a protection against foreign DNA, such as viral DNA. There are over 3,000 restriction enzymes known. The table below shows some of the more common restriction enzymes, with up and down arrows indicating cleavage sites. .

Restriction enzymes are given their names after the prokaryotic organism from which they were isolated. Those isolated from E. coli, for example, will start with Eco. EcoRI cleaves between the G and A on both strands, resulting in overlapping "sticky" ends. SmaII restriction enzyme cleavage, on the other hand, results in "blunt" ends. On both strands, the enzyme cleaves between the G and C.

The DNA fragment that will be cloned must first be separated. This DNA of interest may be a gene, a fragment of a gene, a promoter, or another piece of DNA, and it's usually extracted using PCR or restriction enzyme digestion. A restriction enzyme, as mentioned previously, is an enzyme that cuts double-stranded DNA at a particular sequence. Without destroying the nitrogenous bases, the enzyme makes two incisions, one into each strand of the double helix. This results in either overlapping or blunt ends (also known as sticky ends). The discovery of restriction endonucleases won Daniel Nathans and Hamilton Smith the Nobel Prize in Medicine in 1978. The manipulation of E. coli bacteria to generate human insulin for diabetics was the first practical application of their work.

After isolating the DNA of interest, ligation is needed to inject the amplified fragment into a vector and generate the recombinant DNA molecule. If the sticky ends of the restriction fragments (or a fragment and a plasmid/vector) are complementary, they can be spliced together. It's even possible to ligate the blunt end. Restriction enzymes are used to digest the plasmid or vector (which is normally circular), allowing the target DNA to be inserted. The sticky ends of the isolated DNA of interest and the plasmid or vector would be complementary if they are digested with the same restriction enzyme.