

Technology of Recombinant DNA



Divya shrivastava, Archana Rautela*, Nidhi Semwal and Deepika Joshi

Gyani Inder Singh Institute of Professional Studies, Dehradun, India

Submission: October 13, 2021; **Published:** December 21, 2021

***Corresponding author:** Archana Rautela, Gyani Inder Singh Institute of Professional Studies, Dehradun, India

Abstract

Biotechnology, often known as genetic engineering or recombinant DNA (rDNA), is an industrial technique that applies DNA research to practical applications. rDNA is artificial DNA created by combining or inserting one or more DNA strands. By modifying microbes, animals, and plants to generate therapeutically valuable chemicals, it opened up new possibilities for medical genetics and biomedicine. Recombinant DNA technology helps improve health conditions by creating new vaccines and medicines.

Keywords: Chimeric DNA; Restriction enzymes; Transgenic Plants; Gene Therapy; Future prospects

Introduction

There are three variables that can affect human life: food scarcity, health problems, and environmental concerns. Food, health, and a safe and clean environment are essential for human survival. Global population growth is outpacing human food needs. Humans demand safe and affordable food. The number of deaths worldwide is largely related to human health concerns. There are 36 million deaths every year caused by noncommunicable and communicable diseases, such as cancer, diabetes, AIDS/HIV, TB, and malaria. In third-world countries, health care facilities are significantly worse than those in the host country. Today, global food production far exceeds human needs. Industrial waste is allowed to mix directly with water during a time of rapid industrialization, affecting aquatic life and humans indirectly. Technology must be used to overcome these challenges. Gene engineering involves modern techniques, such as molecular cloning and transformation, which are faster and more effective than traditional methods of addressing issues in agriculture, the environment, and health. Genetic engineering differs from conventional breeding, which transfers both specific and nonspecific genes to the recipient via a number of approaches, including biolistics and *Agrobacterium*. In addition, genetic engineering strategies have also been used to address environmental issues such as converting wastes into biofuels and bioethanol [1-7], cleaning up oil spills, carbon, and other toxic wastes, and detecting arsenic and other contaminants in drinking water via erium-mediated transformation [1].

A plant's genome is altered either through homologous recombination-dependent gene targeting or through nuclease-mediated site-specific genome modification. You can also use recombinase-mediated site-specific genome integration and oligonucleotide-directed mutagenesis [3]. Recombinant DNA technology plays a significant role in improving health conditions by developing new vaccines and pharmaceuticals. New diagnostics, monitoring devices, and therapeutic approaches are developed also to improve the treatment strategies. Gene manipulation in the process of producing synthetic human insulin and erythropoietin is one of the most notable uses of genetic engineering to improve health [3] and can also produce new types of mutant mice. In addition, genetic engineering strategies have also been used to address environmental issues such as converting wastes into biofuels and bioethanol [4-7], cleaning up oil spills, carbon, and other toxic wastes, and detecting arsenic and other contaminants in drinking water. Microbes modified with genetic engineering can also be used for biomining and bioremediation. DNA recombinant technology contributed to the progress of biology and contributed to a number of significant developments. Through the modification of bacteria, animals, and plants to produce medically important compounds, a large range of therapeutic commodities with rapid impact in medical genetics and biomedicine have been created [8,9].

Biotechnology pharmaceuticals are mostly recombinant in nature, which is crucial in fighting lethal diseases. As a result of

recombinant DNA technology, pharmaceutical products changed human life to such an extent that the U.S. In 1997, the FDA approved more recombinant drugs than in the previous several years combined, including anaemia, AIDS, cancers (Kaposi's sarcoma, leukaemia, and colorectal, kidney, and ovarian cancers), and hereditary disorders (cystic fibrosis, familial hypercholesterolemia, Gaucher's disease, haemophilia A, severe combined immunodeficiency disease, and Turner's syndromes). Because plants grow multigame transfer, advanced technologies like as site-specific integration and precisely regulated gene expression are critical [10]. Transcriptional regulation of endogenous genes, their efficiency in new environments, and precise control of transgenic expression are all key difficulties in plant biotechnology that must be addressed [11]. A method of recombining (joining together) DNA segments. Recombinant DNA molecules are made up of fragments of two or more DNA molecules. DNA molecules made from recombinant DNA may enter a cell and multiply under specific conditions, either by themselves or after integrating into chromosomes. Recombinant DNA molecules (rDNA) can be composed of genetic material from various sources (via molecular cloning) in the laboratory to create a sequence that would not naturally exist in the human genome. Herbert Boyer, at the University of California, San Francisco, and Stanley Cohen, at Stanford University, produced recombinant DNA for the first time in 1973. Plasmids can be inserted with foreign DNA using *E. coli* restriction enzymes [12].

Recombinant DNA is DNA created by combining at least two strands. Recombinant DNA is possible because DNA molecules from all organisms share the same chemical structure and differ only in nucleotide sequence within that same overall structure. Recombinant DNA molecules are sometimes called chimeric DNA since they can be made from two different species, like the mythical chimera. The R-DNA technology uses palindromic sequences, which results in blunt and sticky ends. DNA sequences from any species can be used to make recombinant DNA molecules. Plant DNA is linked with DNA from fungi, whereas bacteria DNA is linked with human DNA. Further, DNA sequences that do not exist in nature can be synthesized chemically and incorporated into recombinant molecules. In recombinant DNA technology, any DNA sequence can be created using synthetic DNA and injected into a variety of living organisms. When recombinant proteins are made using recombinant DNA, they are produced within live cells. When recombinant DNA encodes a protein, it is not always translated into a recombinant protein [13]. The expression of foreign proteins usually requires specialised expression vectors and substantial rearranging by the foreign codons. Recombinant DNA and genetic recombination differ in that the former is created in a test tube, while the latter results from the mixing of existing DNA sequences in almost every species [14].

Recombinant DNA is also known as rDNA. We need to understand DNA before we can get to the "r" part.

The DNA provides everything necessary for reproducing an organism. DNA consists of a sugar base, a phosphate base, and a nitrogen base. The nitrogen base adenine (A) is combined with thymine (T), guanine (G), and cytosine (C). There are two nitrogen bases, A & T and G & C. Nitrogen nuclei are able to assemble in an infinite number of ways, and they form a structure known as the "double helix," which is illustrated below. Deoxyribose is the sugar used in DNA. All organisms have the same four nitrogen bases. Diversity is determined by the sequence and number of bases. DNA does not create organisms, only proteins do. RNA is translated into mRNA, which is then translated into protein, which in turn forms the organism. The way a protein is formed changes when the DNA sequence changes. This results either in a new protein or in an inactive protein. The recombinant comes into play now that we know what DNA is. When a piece of DNA is combined with another, it is called recombinant DNA. Thus, recombinant! Recombinant DNA can be created by combining two or more different strands of DNA. In most cases, recombinant DNA is generated by combining two different organisms' DNAs. Recombinant DNA comes into play now that we know what DNA is. Recombinant DNA is produced by mixing a portion of one DNA strand with another. The term recombinant was subsequently coined. Chimeras are recombinant DNA molecules. By connecting two strands of DNA, scientists can create a new strand.

Recombinant DNA can be made using three different techniques. There are three methods. Three methods. Transformation, Phage introduction, and non-bacterial transformation are the three methods. Three methods. Here is an overview of each separately. DNA fragments are selected to be inserted into a vector as part of the transformation process. This part of the DNA is cut with a restriction enzyme, and the DNA insert is ligated with DNA Ligase. Selectable markers contained in the insert can be used to identify recombinant molecules. When a host cell without a vector is exposed to an antibiotic, the host with the vector dies, but the host without the vector lives since it is resistant. During a process called transformation, vectors are introduced into host cells. *E. Coli* is an example of a possible host cell. In order for the host cells to accept foreign DNA, they must first be specially prepared. Depending on their properties, different vectors can be used for different purposes. Different characteristics may distinguish transformed hosts from their untransformed counterparts. Several properties can be observed, including symmetrical cloning sites, large sizes, and high copy numbers.

It is somewhat similar to the process described above, Transformation. A main difference between bacterial and non-bacterial is that bacteria, such as *E. coli*, do not serve as hosts. In microinjection, DNA is injected directly into the nucleus of the host cell being converted. In biolistics, the host cells are sprayed with high-velocity micro projectiles such as gold or tungsten particles coated with DNA.

Virus introduction is similar to transfection, except that instead of bacteria, phages are used. Packaging of a vector in vitro is used. Viruses such as lambda or M13 are used to build recombinant phages. The recombinants that are generated are selectively selected by various selection procedures. Recombinant protein is produced by the host cell when recombinant DNA is present. In the absence of expression factors, this genome is not able to produce significant amounts of recombinant proteins. In order for a protein to be expressed, a gene must be surrounded by signals that provide instructions for transcription and translation by the cell. Promoters, binding sites for ribosomes, and terminators provide these signals. These signals can be found in expression vectors, which are used to introduce foreign DNA into cells. Each species has its own signal. *E. Coli* is unlikely to be able to recognize signals from human promoters and terminators, so these signals must come from *E. Coli* signals. If the gene has introns or signals that are terminators of the bacterial host, problems arise. Recombinant proteins might not be digested, folded, or destroyed properly if the process is prematurely terminated. Recombinant proteins are generally made by yeast and filamentous fungi in eukaryotic systems. Due to needs and the need for support, animal cells are difficult to use. There are, however, some proteins that eukaryotes cannot make, so bacteria are needed [15].

In the past decade, it has also gained considerable importance in the fields of transgenic animals, pest-resistant crops, as well as genetically modified foods and drinks. Some of the areas where this technology is having a significant impact are as follows: [16]:

The use of transgenic animals as experimental models in biomedical research.

In biomedical research, transgenic fruit flies (*Drosophila melanogaster*) are used as model organisms to develop better crops (resistant to insects, pests, herbicides, and harsh environmental conditions such as heat).

- a. A plant that produces its own insecticide.
- b. Improve product shelf life by cropping.
- c. Increased nutritional value of crops.
- d. Virus-resistant crops.
- e. The hepatitis B vaccine (recombinant)
- f. The prevention and treatment of sickle cell anemia.
- g. Treatment and prevention of cystic fibrosis.
- h. Detection and prevention of clotting factors.
- i. Production of insulin.
- j. Recombinant pharmaceutical production.
- k. A genetic therapy based on germ line cells or somatic cells [16].

- l. Technology for recombinant DNA synthesis [17]
- m. These tools include mainly the following:

Polymerases help synthesize, polymerases help cut, and ligases help bind. In recombinant DNA technology, restriction enzymes are used to determine the position of the desired gene within the vector genome. These enzymes come in two forms: exonucleases and endonucleases. Endonucleases cut the ends of DNA strands, whereas exonucleases remove the middles of DNA strands. Restriction endonucleases are sequence-specific and cut DNA at specific points. DNA is measured for length and a specific site called a restriction site is used to make the cut. This results in sticky ends in the sequence. By cutting the desired genes and the vectors with the same restriction enzymes, complementary sticky notes are created, which makes the ligases' job easier to bind the desired gene to the vector.

Recombinant DNA technology would be incomplete without these tools because they are the ultimate means through which a gene is introduced into a host organism. Plasmids and bacteriophages are the most commonly used vectors in recombinant DNA technology due to their high copy number. The origin of replication consists of a sequence of nucleotides from which replication begins; the selectable marker, the antibiotic resistance gene, and DNA cloning sites, the places included in restriction enzymes.

Recombinant DNA technology relies on the host to incorporate the desired DNA using enzymes. There are a number of methods for incorporating recombinant DNA into the host.

- a. The selection of cloning vectors
- b. DNA insert into vector to form rec DNA molecule
- c. A suitable host is introduced with the rDNA molecule.
- d. Host cells that have been transformed.
- e. The expression and multiplication of DNA-inserted into the host.

Cloning DNA segments of interest from DNA segments of interest is the first step in rDNA technology. Enzymatically, this DNA segment can then be isolated. The segment of DNA which is of interest is known as a foreign insert, target insert or clone. Cloning vectors are self-replicating molecules, into which the DNA insert is to be integrated. The next step in rec DNA technology is to select the best cloning vector. Plasmids and bacteriophages are the most commonly used vectors.

The cleaved endonucleases [in step(i)] have been ligated (joined) to the vector DNA by the enzyme ligase to form what is known as an insert-cloning molecule. A suitable host cell is selected and the rec DNA molecule formed [in step (iii)] is introduced into this target cell. Recombinant DNA enters host cells by undergoing transformation. In most cases, selected hosts are

bacterial cells such as *E. coli*, but yeast and fungi can also be used. A transformed cell (or recombinant cell) is one that has taken up a recDNA molecule. The transformed cells are separated from the non-transformed cells in this step using various methods that make use of marker genes. The foreign DNA should also be tested for expression in host cells before being inserted into vector DNA. Finally, the transformed host cells should be multiplied to obtain sufficient numbers of copies. Genes may also be transferred and expressed in another organism if necessary [18]. There are many uses for recombinant DNA technology, including the development of enzymes applicable to certain food-processing conditions. Many essential enzymes are accessible in specific manufacturing processes in the food industry due to different enzyme roles and functions. The creation of microbial strains was another important breakthrough enabled by recombinant DNA techniques. Specialized engineering for protease synthesis resulted in the creation of different microbial strains that produce enzymes. Some fungi strains have been modified to limit their production of hazardous compounds [19].

In the food industry, lysozymes are effective bacteria-killers. They prevent the colonization of microorganisms. Since it extends

the shelf life of foods like fruits, vegetables, cheese, and meat, it is a good agent for storing them. It is possible to prevent food spoilage by immobilizing lysozyme in polyvinyl alcohol films and cellulose. Lysozyme can also be used to improve the shelf life of fish skin gel and inhibit the growth of bacteria that degrade food [20–22]. *E. coli* and *Staphylococcus exopolysaccharides*. Hydralization of coli can be achieved using DSPB, the engineered version of T7. This ability of DspB results in a decrease in bacteria population [22]. The combination of serine proteases and amylases can be used to remove biofilms related to the food industry [23]. *S. Salmonella infantis*, *Clostridium perfringens*, *B. Cereus*, *Campylobacter jejuni*, and *L. monocytogenes*, *Yersinia enterocolitica*, and some other food spoiling microorganisms can be inhibited by glucose oxidase. It is also considered A wide variety of foodborne pathogens can be killed with this enzyme [22]. A factory that creates recombinant proteins for use in medicine recently went into operation, and more are in the works to create similar essential proteins for medical use. There have been numerous recombinant proteins expressed in various plant species for use as enzymes; many protein research projects rely on proteins found in milk, and some are used in industries and in medicine [24] (Figure 1).

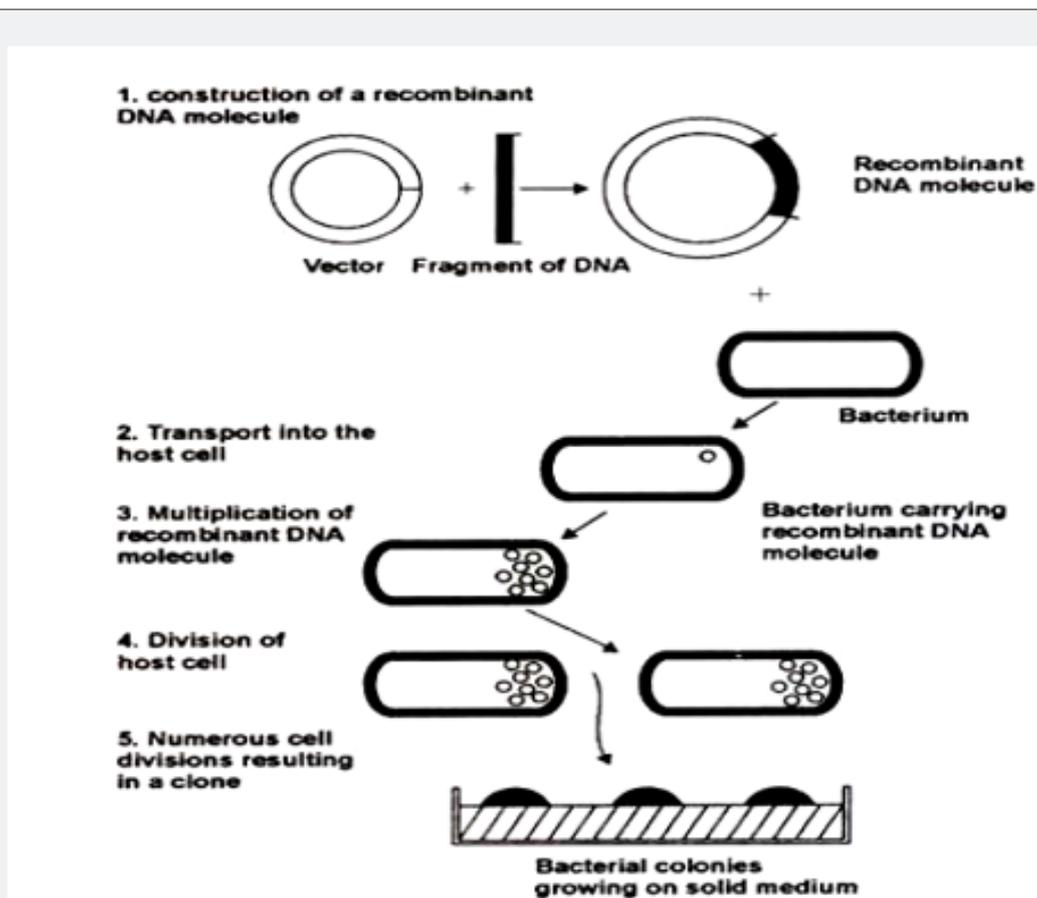


Figure 1: The basic steps of rec DNA Technology using the bacterial plasmid as cloning vector.

There are many applications of recombinant DNA technology in treating diseases and improving health conditions. The following sections describe the significant breakthroughs in recombinant DNA technology that have improved human health. A cutting-edge medical procedure, gene therapy has therapeutic potential. The first report of gene therapy treating a genetic disorder paved the way for treating the deadliest hereditary diseases. In [25,26], there is a question about adoption. Efficacy of the method has been demonstrated in the treatment of adenosine deaminase deficiency (ADA-SCID), one of the most common primary immunodeficiencies. There were several obstacles, including the use of PEGylated ADA (PEG-ADA) during gene therapy and targeting a T-lymphocyte-specific gene transfer, contributed to poor results when the method was introduced [27,28]. there are two places to put these. In a later study, hematopoietic stem cells (HSCs) were targeted via enhanced gene transfer and myeloablative conditioning [29]. There are genes associated with adrenoleukodystrophy (X-ALD) and X-linked disorders which appear through lentivirus transmission based on HIV-1. According to [30], the presence of XALD suggests that true HSCs are genetically corrected.

In the first human genetic disease to be treated with a lentiviral vector, a lentiviral vector proved highly effective [31]. In 2006, immunotherapy was used to treat metastatic melanoma by triggering the production of certain proteins. A new avenue for research into the medical use of immunotherapy to treat fatal diseases has opened up as a result of this accomplishment. According to [32]. In two patients, high levels of cells designed for tumour detection in the blood using a retrovirus expressing a T-cell receptor led to a significant reduction of metastatic melanoma lesions up to a year after infusion. In subsequent studies, this technique was applied to metastatic synovial cell carcinoma patients. [33]. Cytotoxic T-cells were genetically modified to express a Chimeric Antigen Receptor (CAR) with specificity for the B-cell antigen CD19 to treat chronic lymphocytic leukemia. The *in vivo* selection of disease pathogenesis, regardless of the fact that genetically modified cells only repair a few progenitors, results in the selection for diseases like SCID-X1 and ADASCID. Human HSCs can be protected from chemo by combining the gene and pharmacological treatment during chemotherapy for glioblastoma [34]. Many antibodies and their derivatives have recently been produced using plants. In contrast, seven antibodies or antibody derivatives met all the requirements out of a large number. This antibody, which is made by transgenic tobacco plants, is a type of chimeric secretory IgA/G. *Streptococcus* mutants, a cause of tooth decay, can be detected by this antibody.

The monoclonal antibody T84. 66 detects antigen carcinoembryonic, which is still considered an affectively defined marker in epithelia malignancies. [35,36]. In transgenic soybean cells and Chinese Hamster Ovary (CHO) cells, humanised full-

length IgG1 have been synthesized that can recognize the herpes simplex virus (HSV)-2. A glycoprotein B was produced. In mice, antibodies from both sources inhibited vaginal transmission of HSV-2 after topically applying They could easily prevent infections spread during sexual encounters if they worked the same in people [37,38]. The 38C13 scFv antibody is based on the idiotype of malignant B cells in the well-characterized mouse lymphoma cell line 38C13. In mice, antibody administration resulted in the development of anti-idiotypic antibodies that identified 38C13 cells, which assisted in the protection of mice against lymphoma cells [39,40]. A monoclonal antibody known as PIPP recognizes human chorionic gonadotropin. The agroinfiltration and transgenesis of tobacco plants enabled them to synthesize full-length monoclonal antibodies, scFvs, and diabody derivatives [41]. For medicines to function correctly, it is critical to investigate the complex system of enzymes and metabolic processes involved in their metabolism. Recombinant DNA methods are increasingly using heterologous expression, which involves expressing the enzyme's genetic information *in vitro* or *in vivo* via gene transfer [42,43].

The effectiveness and specificity of conventional vaccines are less than those of recombinant vaccines. The nasal delivery of adenovirus vectors which contain antigens of disease-causing organisms provides a painless, fast and sustainable approach to transmitting adenovirus vectors which contain pathogen antigens. As a pharmacological vaccine, it induces anti-influenza activity in the airways through transgene expression. [44] There is now a possibility to produce human growth hormone (FSH) using recombinant DNA. A targeted eukaryotic cell line has been selected to express FSH since it is a highly complicated heterodimeric protein. Technology that uses recombinant DNA promotes follicle development for treating assisted reproductive. Large numbers of people are receiving treatment with r-FSH. Recombination FSH and Luteinizing Hormone (LH), in particular, improved ovulation [45,46].

In recent years, human follicle-stimulating hormone (FSH) has been produced using recombinant DNA technology. A specific cell line from eukaryotes is used to express this complex heterodimeric protein. By promoting the development of follicles, recombinant DNA can be used for assisted reproductive. Large numbers of people are receiving treatment with r-FSH. Recombination of r-FSH and Luteinizing Hormone (LH), in particular, was successful in enhancing ovulation and conception. Biochemical properties and gene expression profiles of metabolic pathways were studied using the cultures. In turn cultures, intermediates and key enzymes involved in the biosynthesis of secondary metabolites can be identified [47,48]. Activating the *rolC* gene increased strawberry nutrition. The sugar content of this gene is increased as well as antioxidant activity. Two enzymes are needed for anthocyanin glycosylation: glycosyl-transferase and transferase. A

number of nutritionally-related genes discovered in strawberries are useful for genetic transformation of the strawberries to improve one component. The anthocyanin components are controlled by the bHLH and FRUITE4 genes in raspberry, whereas flavonol is controlled by the ERubLRSQ072H02 gene. Certain transformations are able to increase output and quality from these genes. [49].

A collaboration, the University of Tennessee and Oak Ridge National Laboratory released genetically modified microorganisms for bioremediation purposes, such as strain HK44 of *Pseudomonas fluorescens*. [50,51]. A transposon-based bioluminescence-producing lux gene was linked to the promoter of the modified strain, resulting in increased breakdown of naphthalene bioluminescent response. In situ monitoring of bioremediation can be achieved using [52]. HK44 detects naphthalene availability and biodegradation using bioluminescence signaling [53]. In recent years, genetic engineering has been widely used to detect and absorb contaminants in drinking water and other products. In *Torenia*, *Petunia*, and *Verbena*, insertion of the AtPHR1 gene altered their ability to absorb Pi. In contaminated aquatic environments, AtPHR1 transgenic plants may contribute to effective phytoremediation. [The 54A portion of the AtPHR1 gene has been introduced into pBinPLUS, a binary vector with an improved cauliflower mosaic virus 35S promoter. The plasmid used for transformation was pSPB1898 [54,55] using *Agrobacterium tumefaciens* in *Petunia* and *Verbena* [56]. The relevance of plant metabolic mechanisms in the remediation of environmental contaminants has been demonstrated. Decomposition or digestion can be difficult for some compounds. TNT is only partially digested, so the nitrogen reacts with oxygen to produce deadly superoxide.

The monodehydroascorbate reductase gene is knocked out, strengthening the plant's resistance to TNT. By fine-tuning enzyme activity and using knock-out engineering, plants respond more effectively to hazardous metals. An enzyme that produces heavy metal binding peptides, phytochelatin synthase, was attenuated enzymatically to improve heavy metal tolerance [57]. Metabolic mechanisms of plants are known to play a critical role in the remediation of pollutants in the environment. Decomposition and digestion of the compounds are difficult. When TNT is only partially digested, nitrogen reacts with oxygen to form deadly superoxide. The monodehydroascorbate reductase in plants is knocked out to increase their tolerance to TNT. By enhancing enzyme activity with knockout engineering, plants can more effectively deal with hazardous metals. To improve heavy metal tolerance by inhibiting an enzyme that produces heavy metal binding peptides, phytochelatin synthase, enzymatic activity was attenuated [58]. A BRASSINOSTEROL (BR) is responsible for regulating physiological and developmental processes in plants. BRs action is the result of phosphorylation and dephosphorylation cascades [59].

In the process of producing hydrogen, cyanobacteria break down carbon dioxide, a powerful greenhouse gas. A specific production process is maintained by correctly using the enzymes necessary to create the product. However, advanced techniques such as genetic engineering, nutrition manipulation, and metabolic engineering are now being used to create cell-free systems [60–62]. The commercialization of this energy source will help to keep the environment clean, which is impossible to achieve with traditional energy sources that emit CO₂ and other harmful substances [63]. Genetically modified cyanobacteria can also convert CO₂ into reduced fuel molecules. As a result, carbon energy sources will no longer be harmful to the environment. In general, this strategy has been successful for a wide range of commodity compounds, primarily energy carriers, such as short and medium chain alcohols [64]. *G. sulfurreducens* conductive biofilms are promising sources in the fields of renewable energy, bioremediation, and bioelectronics. The electron acceptor CL-1 produced biofilms that were six times more conductive than wild-type biofilms when cultured with electrode. When the PilZ gene producing proteins was deleted from the *G. sulfurreducens* genome became more active compared to wild-type. CL-1Ln has been isolated from a strain in which the gene GSU1240 has been removed. The strain has a high conductivity and a low formal potential, reducing potential losses in microbial fuel cells. Lower losses increase potential energy [65].

The fact that many recombinant pharmaceuticals are manufactured with microbial cells suggests that several obstacles come into the way of their production of functional proteins, but these can be overcome by altering cellular mechanisms. The main obstacles to gene expression are posttranslational modification, the activation of cell stress responses, and the instability of proteasomal activity. Oftentimes, human genetic mutations lead to insufficiencies in protein synthesis, which can be rectified by incorporating genetic material to close these gaps and bring levels back to normal. *Escherichia coli* is being used in recombinant DNA technology as a platform for producers to use controlled processes to produce the required molecules at an affordable cost [66,67]. In addition to allowing the analysis and manipulation of yeast genes in the test tube, recombinant DNA research offers considerable potential for studying yeast biology in greater depth. Most importantly, yeast is now able to be turned into cloned genes by transforming them with DNA and selecting selectable markers developed for this purpose. Molecular and traditional genetic manipulations of yeast are now both possible because of technological advancements. Recombinant DNA has proved most effective in addressing biological issues centered around the structure and organization of individual genes [68,69].

Recent breakthroughs in recombinant DNA technology have brought about profound changes in the research area and opened up interesting and advanced avenues for biosynthetic pathway research. ACTIOMYCETES are used for pharmaceutical

productions, such as the production of nutrients and drugs for health science research. A significant proportion of the biosynthetic compounds comes from these sources and as such has been considered in the design of recombinant drugs. Their compounds were found to be more effective against a wide range of bacteria and other pathogenic microbes in clinical trials. Additionally, these compounds have shown antitumor and immunosuppressive properties [70]. Technology that makes use of recombinant DNA can prevent and treat genetic disorders.

The development of DNA vaccines is an innovative method of providing immunity against several diseases. This process involves the delivery of DNA that contains pathogenic genes. In clinical trials, human gene therapy is mostly used to treat cancer. Transfection efficiency has been studied in relation to the design of gene delivery systems in recent years. The idea of using transfection for cancer gene therapy with minimal side effects is still being investigated, including for cases of brain, breast, lung, and prostate cancer. There are also considerations of gene therapy for renal transplantation, Gaucher disease, hemophilia, Alport syndrome, and renal fibrosis [71].

References

1. Kumar S, Kumar A (2015) Role of genetic engineering in agriculture. *Plant Archives* 15(1): 1-6.
2. Cardi T, Stewart CN Jr (2016) Progress of targeted genome modification approaches in higher plants. *Plant Cell Rep.* 35(7): 1401–1416.
3. Lomedico PT (1982) Use of recombinant DNA technology to program eukaryotic cells to synthesize rat proinsulin: a rapid expression assay for cloned genes. *Proc Natl Acad Sci U S A* 79(19): 5798-5802.
4. Ullah MW, Khattak WA, Ul-Islam M, Khan S, Park JK (2015) Encapsulated yeast cell-free system: a strategy for cost effective and sustainable production of bio-ethanol in consecutive batches. *Biotechnology and Bioengineering* 20(3): 561-575.
5. Ullah MW, Khattak WA, Ul-Islam M, Khan S, Park JK (2014) Bio-ethanol production through simultaneous and fermentation using an encapsulated reconstituted cell-free enzyme system. *Biochemical Engineering journal* 91: 110–119.
6. Khattak WA, Ul-Islam M, Ullah MW, Yu B, Khan S, et al. (2014) Yeast cell-free enzyme system for bio-ethanol production at elevated temperatures. *Process Biochemistry* 49(3): 357-364.
7. Khattak WA, Ullah MW, Ul-Islam M, Shaikat K, Minah Kim, et al. (2014) Developmental strategies and regulation of cell-free enzyme system for ethanol production: a molecular prospective. *Appl Microbiol Biotechnol* 98(23): 9561-9578.
8. Galambos L, Sturchio JL (1998) Pharmaceutical firms and the transition to biotechnology: a study in strategic innovation. *Business History Review* 72(2): 250–278.
9. Steinberg FM, Raso J (1998) Biotech pharmaceuticals and biotherapy: an overview. *J Pharm Pharm Sci* 1(2): 48-45.
10. Liu W, Yuan JS, Stewart CN Jr (2013) Advanced genetic tools for plant biotechnology. *Nat Rev Genet* 14(11): 781–793.
11. Venter M (2007) Synthetic promoters: genetic control through cis engineering. *Trends in Plant Sci* 12(3): 118-124.
12. (2018) GNN- Genetics and Genomics Timeline. www.genomenetwork.org.
13. Rosano, Germán L, Ceccarelli, Eduardo A (2014) Recombinant protein expression in *Escherichia coli*: advances and challenges. *Front Microbiol* 5: 172.
14. Promoters used to regulate gene expression (2018).
15. www.rpi.edu/dept/chem-eng/Biotech-Environ/Projects00/rdna/rdna.html.
16. Chaudhuri Keya (2013) recombinant DNA technology (text book), the energy and resource institute publishers, India.
17. byjus.com/biology/recombinant-dna-technology.
18. <http://www.biologydiscussion.com/dna/recombinant-dna-technology/recombinant-dna-technology-with-diagram/1778>
19. ZS Olempska-Ber, RI Merker, MD Ditto, MJ DiNovi (2006) Food-processing enzymes from recombinant microorganisms—a review. *Regul Toxicol Pharmacol* 45 (2): 144–158.
20. ZX Lian, ZS Ma, J Wei, H Liu (2012) Preparation and characterization of immobilized lysozyme and evaluation of its application in edible coatings. *Process Biochemistry* 47(2): 201–208.
21. SH Bang, A Jang, J Yoon, Pil Kim, Jong Soo Kim, et al. (2011) Evaluation of whole lysosomal enzymes directly immobilized on titanium (IV) oxide used in the development of antimicrobial agents, *Enzyme and Microb Technol* 49(3): 260–265.
22. Thallinger B, EN Prasetyo, GS Nyanhongo, GM Guebitz (2013) Antimicrobial enzymes: an emerging strategy to fight microbes and microbial biofilms. *Biotechnology J* 8(1): 97–109.
23. CE Torres, G Lenon, D Craperi, R Wilting, A Blanco (2011) Enzymatic treatment for preventing biofilm formation in the paper industry. *Appl Microbiol Biotechnol* 92 (1): 95–103.
24. JK C Ma, PM W Drake, P Christou (2003) The production of recombinant pharmaceutical proteins in plants. *Nature Reviews Genetics* 4(10): 794-805.
25. M Cavazzana-Calvo, S Hacein-Bey, G De Saint Basile, F Gross, E Yvon, et al. (2000) Gene therapy of human severe combined immunodeficiency (SCID)-X1 disease. *Science* 288 (5466): 669–672.
26. S Hacein-Bey-Abina, F Le Deist, F Carlier, Christophe Hue, Jean-Pierre De Villartay, et al. (2002) Sustained correction of X-linked severe combined immunodeficiency by ex vivo gene therapy. *N Engl J Med* 346 (16): 1185–1193.
27. SJ Howe, MR Mansour, K Schwarzwald, Cynthia Bartholomae, Michael Hubank, et al. (2008) Insertional mutagenesis combined with acquired somatic mutations causes leukemogenesis following gene therapy of SCID-X1 patients. *J Clin Invest* 118(9): 3143–3150.
28. RM Blaese, KW Culver, AD Miller, CS Carter, T Fleisher, et al. (1995) T lymphocyte directed gene therapy for ADA-SCID: initial trial results after 4 years. *Science* 270(5235): 475–480.
29. A Aiuti, S Vai, A Mortellaro, Giulia Casorati, Francesca Ficara, et al. (2002) Immune reconstitution in ADA-SCID after PBL gene therapy and discontinuation of enzyme replacement. *Nature Medicine* 8(5): 423–425.
30. N Cartier, S Hacein-Bey-Abina, CC Bartholomae, Gabor Veres, et al. (2009) Hematopoietic stem cell gene therapy with a lentiviral vector in X-linked adrenoleukodystrophy. *Science* 326(5954): 818–823.

31. E Montini, A Biffi, A Calabria (2012) Integration site analysis in a clinical trial of lentiviral vector based haematopoietic stem cell gene therapy for meatchromatic leukodystrophy. *Human Gene Therapy* 23 article A13.
32. RA Morgan, ME Dudley, JR Wunderlich, Marybeth S Hughes, James C Yang, et al. (2006) Cancer regression in patients after transfer of genetically engineered lymphocytes. *Science* 314(5796): 126–129.
33. PF Robbins, RA Morgan, SA Feldman, James C Yang, Richard M Sherry, et al. (2011) Tumor regression in patients with metastatic synovial cell sarcoma and melanoma using genetically engineered lymphocytes reactive with NY-ESO-1. *J Clin Oncol* 29(7): 917–924.
34. JE Adair, BC Beard, GD Trobridge, Tobias neff, Jason k Rockhill, et al., (2012) Extended survival of glioblastoma patients after chemoprotective HSC gene therapy. *Science Translational Medicine* 4(133): 133ra57.
35. E Stoger, C Vaquero, E Torres, M Sack, L Nicholson, et al. (2000) Cereal crops as viable production and storage systems for pharmaceutical scFv antibodies. *Plant Mol Biol* 42 (4): 583–590.
36. Vaquero C, Sack M, Schuster F, Finnern R, Drossard J, et al. (2002) A carcinoembryonic antigen-specific diabody produced in tobacco. *The FASEB Journal* 16(3): 408-410.
37. Karrer E, Bass SH, Whalen R, Patten PA (2012) US. Patent No. 8,252,727.
38. Ionescu RM, Vlasak J, Price C, Kirchmeier M (2008) Contribution of variable domains to the stability of humanized IgG1 monoclonal antibodies. *J Pharm Sci* 97(4): 1414-1426.
39. Adams CW, Allison DE, Flagella K, Presta L, Clarke J, et al. (2006) Humanization of a recombinant monoclonal antibody to produce a therapeutic HER dimerization inhibitor, pertuzumab. *Cancer Immunology, Immunotherapy* 55(6): 717-727.
40. McCormick AA, Reddy S, Reiln SJ, Cameron TI, Czerwinski DK, et al. (2008) Plant-produced idiotype vaccines for the treatment of non-Hodgkin's lymphoma: safety and immunogenicity in a phase I clinical study. *Proceedings of the National Academy of Sciences of the United States of America* 105(29): 10131-10136.
41. Kathuria S, Sriraman R, Nath R, Sack M, Pal R, et al. (2002) Efficacy of plant produced recombinant antibodies against HCG. *Human Reproduction* 17(8): 2054-2061.
42. Rostami-Hodjegan A, Tucker GT (2007) Simulation and prediction of in vivo drug metabolism in human populations from in vitro data. *Nat Rev Drug Discov* 6(2): 140-148.
43. Nicholson JK, Holmes E, Wilson ID (2005) Gut microorganisms, mammalian metabolism and personalized health care. *Nat Rev Microbiol* 3(5): 431-438.
44. Zhang J, Tarbet EB, Toro H, Tang D-CC (2011) Adenovirus vectored drug-vaccine duo as a potential driver for conferring mass protection against infectious diseases. *Expert Rev Vaccines* 10(11): 1539-1552.
45. Fan QR, Hendrickson WA (2005) Structure of human follicle stimulating hormone in complex with its receptor. *Nature* 433(7023): 269-277.
46. Assidi M, Dufort I, Ali A, Mélanie Hamel, Omran Algriany, et al. (2008) Identification of potential markers of oocyte competence expressed in bovine cumulus cells matured with follicle-stimulating hormone and/or phorbol myristate acetate in vitro. *Biology of Reproduction* 79(2): 209-222.
47. Hu ZB, Du M (2006) Hairy root and its application in plant genetic engineering. *Journal of Integrative Plant Biology* 48(2): 121-127.
48. Ling CQ, Wang LN, Wang Y, Yuan-hui Z, Zi-fei Y, et al. (2014) The roles of traditional Chinese medicine in gene therapy. *J Integr Med* 12(2): 67-75.
49. Mazzoni L, Perez-Lopez P, Giampieri F, Alvarez-Suarez JM, Gasparri M, et al. (2016) The genetic aspects of berries: from field to health. *Journal of the Science of Food and Agriculture* 96(2): 365-371.
50. Ripp S, Nivens DE, Ahn Y, Werner C, Jarrell J, et al. (2000) Controlled field release of a bioluminescent genetically engineered microorganism for bioremediation process monitoring and control. *Environmental Science and Technology* 34(5): 846-853.
51. Saylor GS, Cox CD, Burlage R, Ripp S, Nivens DE, et al. (1999) Field application of a genetically engineered microorganism for polycyclic aromatic hydrocarbon bioremediation process monitoring and control. In *Novel Approaches for Bioremediation of Organic Pollution* Springer, New York, NY, USA, pp: 241-254.
52. King JMH, Digrazia PM, Applegate B, Sanseverino J, Dunbar P, et al. (1990) Rapid, sensitive bioluminescent reporter technology for naphthalene exposure and biodegradation. *Science* 249(4970): 78-781.
53. Chatterjee J, Meighen EA (1995) Biotechnological applications of bacterial bioluminescence (lux) genes. *Photochemistry and Photobiology* 62(4): 641-650.
54. Matsui K, Togami J, Mason JG, Chandler SF, Tanaka Y (2013) Enhancement of phosphate absorption by garden plants by genetic engineering: a new tool for phytoremediation. *BioMed Research International*, 182032, p. 13.
55. Horsch RB, Fry JE, Hoffmann NL, Eichholtz D, Rogers SG, et al. (1985) A simple and general method for transferring genes into plants. *Science* 227(4691): 1229-1230.
56. Tamura M, Togami J, Ishiguro K, Nakamura N, Katsumoto Y, et al. (2003) Regeneration of transformed verbena (*verbena × hybrida*) by *Agrobacterium tumefaciens*. *Plant Cell Reports* 21(5): 459-466.
57. Jez JM, Lee SG, Sherp AM (2016) The next green movement: plant biology for the environment and sustainability. *Science* 353(6305): 1241-1244.
58. Clemens S, Ma JF (2016) Toxic heavy metal and metalloid accumulation in crop plants and foods. *Annu Rev Plant Biol* 67(1): 489-512.
59. Kim EJ, Youn JH, Park CH, et al. (2016) Oligomerization between BSU1 family members potentiates brassinosteroid signaling in *Arabidopsis*. *Molecular Plant* 9(1): 178- 181.
60. Ullah MW, Ul-Islam M, Khan S, Kim Y, Park JK (2016) Structural and physico-mechanical characterization of biocellulose produced by a cell-free system. *Carbohydr Polym* 136: 908-916.
61. Ullah MW, Ul-Islam M, Khan S, Kim Y, Park JK (2015) Innovative production of bio-cellulose using a cell-free system derived from a single cell line. *Carbohydr Polym* 132: 286-294.
62. Ullah MW, Khattak WA, Ul-Islam M, Khan S, Park JK (2016) Metabolic engineering of synthetic cell-free systems: strategies and applications. *Biochemical Engineering Journal* 105: 391-405.
63. Tiwari A, Pandey A (2012) Cyanobacterial hydrogen production-a step towards clean environment. *International Journal of Hydrogen Energy* 37(1): 139-150.
64. Savakis P, Hellingwerf KJ (2015) Engineering cyanobacteria for direct biofuel production from CO₂. *Current Opinion in Biotechnology* 33: 8-14.

65. Leang C, Malvankar NS, Franks AE, Nevin KP, Lovely DR (2013) Engineering *Geobacter sulfurreducens* to produce a highly cohesive conductive matrix with enhanced capacity for current production. *Energy and Environmental Science* 6: 1901-1908.
66. Ferrer-Miralles N, Domingo-Espin J, Corchero J, Vázquez E, Villaverde A (2009) Microbial factories for recombinant pharmaceuticals. *Microb Cell Fact* 8: 7.
67. Vajo Z, Fawcett J, Duckworth WC (2001) Recombinant DNA technology in the treatment of diabetes: insulin analogs. *Endocr Rev* 22(5): 706-717.
68. DeJong JM, Liu Y, Bollon AP, Robert M Long, Stefan Jennewein, et al. (2006) Genetic engineering of taxol biosynthetic genes in *Saccharomyces cerevisiae*. *Biotechnol Bioeng* 93(2): 212-224.
69. Walker GM (2009) *Yeasts* in Desk Encyclopedia of Microbiology, Elsevier, 2nd edition.
70. Mendez C, Weitnauer G, Bechthold A, Salas JA (2000) Structure alteration of polyketides by recombinant DNA technology in producer organisms prospects for the generation of novel pharmaceutical drugs. *Current Pharmaceutical Biotechnology* 1(4): 355-395.
71. Misra A (2010) *Challenges in Delivery of Therapeutic Genomics and Proteomics*, Elsevier, Amsterdam, Netherlands.



This work is licensed under Creative Commons Attribution 4.0 License
DOI: [10.19080/AIBM.2021.16.555947](https://doi.org/10.19080/AIBM.2021.16.555947)

Your next submission with Juniper Publishers will reach you the below assets

- Quality Editorial service
- Swift Peer Review
- Reprints availability
- E-prints Service
- Manuscript Podcast for convenient understanding
- Global attainment for your research
- Manuscript accessibility in different formats
(Pdf, E-pub, Full Text, Audio)
- Unceasing customer service

Track the below URL for one-step submission
<https://juniperpublishers.com/online-submission.php>