An Overview on Recombinant DNA Technology and Its Applications

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ABSTRACT: Recombinant DNA technology was only a dream a century ago, when it was thought that by manipulating the expressions of target genes, desirable qualities could be improved in living beings. However, in the recent era, this area has proven unique effects in terms of human life progress. Crucial proteins essential for health concerns and dietary needs can be generated securely, affordably, and in adequate quantities thanks to this technology. This technique offers a wide range of uses and has the potential to improve crucial areas of life, such as health, food security, including resistance to a variety of detrimental environmental impacts. Genetic modifications, particularly in agriculture, have enhanced resistance to toxic compounds, increase in product yield, and demonstrated increased ability to adapt for better survival. Furthermore, recombinant pharmaceuticals are now being used with confidence, as well as commercial authorizations are being obtained quickly. Bioremediation and the treatment of serious diseases are also common uses of recombinant DNA technology, gene therapy, as well as genetic modifications. Because of the tremendous development and wide range of applications in the field of recombinant DNA technology, this review article focuses primarily on its significance and potential applications in daily life.

KEYWORDS*: *Agriculture, Disease, Genetic Engineering, Recombinant, DNA Technology.

1. INTRODUCTION

Three elements have a significant impact on human life: food scarcity, health problems, as well as environmental concerns. Aside from a clean and safe environment, food and health are essential human needs [1]. Human food needs are quickly rising as the world's population grows at a faster pace. Humans need food that is both safe and affordable. Several human-related health problems cause a significant number of fatalities throughout the world. About 36 million people die each year from non-communicable and communicable illnesses such as cardiovascular diseases, cancer, diabetes, AIDS/HIV, TB, malaria, and other disorders. Despite significant efforts, present global food production falls well short of human needs, and health-care facilities in third-world nations are much worse[2].

Rapid industrialisation has increased environmental contamination, and industrial wastes are permitted to mingle directly with water, affecting aquatic marine life and, indirectly, humans. As a result, contemporary technology must be used to solve these problems. Unlike conventional methods to overcoming agriculture, health, and environmental concerns through breeding, medicinal products, as well as air pollutant degradation though the conventional methods, genetic engineering makes use of modern tools or approaches, such as molecular cloning & transformation, which are faster and produce more reliable results. In contrast to traditional breeding, which transmits a large number of both particular and nonspecific genes to the recipient, genetic engineering simply delivers a small block of desired genes to the target via different methods such as biolistic and Agrobacterium-mediated transformation [3].

Homologous recombination-dependents gene targeting or nuclease-mediated site-specific genome editing are both used to change plant genomes. Site-specific genome integration mediated by recombinases or oligonucleotide-directed mutagenesis are other options. By creating novel vaccinations and medicines, recombinant DNA technology is helping to improve health conditions. Diagnostic tests, monitoring equipment, and novel therapy methods are all being developed to enhance treatment tactics. One of the most prominent instances of genetic engineering in health is the creation of new kinds of experimental mutant mice for research purposes and the synthesis of synthetic human insulin and erythropoietin by genetically engineered bacteria. Environmental problems such as turning wastes into biofuels and bioethanol, cleaning oil spills, carbon, and other hazardous wastes, and detecting arsenic and other pollutants in drinking water have all been addressed using genetic engineering techniques. Bio mining and bioremediation may both benefit from genetically engineered microorganisms.

Recombinant DNA Technology is a kind of recombinant DNA that is changing genetic material outside of an organism to acquire improved and desired traits in live creatures or as their products is referred to as recombinant DNA technology. This technique entails inserting DNA fragments from a number of sources into a suitable vector with a desired gene sequence. Manipulation of an organism's genome may be done by adding one or more new genes and regulatory elements, or by recombining genes and regulatory elements to reduce or inhibit the expression of indigenous genes. Using restriction endo-nucleases for particular target sequence DNA sites, enzymatic cleavage is used to acquire various DNA fragments, which are then joined using DNA ligase activity to fix the desired gene in vector. After that, the vector is injected into a host organism, which is cultured in culture to generate multiple copies of the inserted DNA fragment, and then clones containing a relevant DNA fragment are chosen and collected.

Regulation and safe use of rDNA technology were addressed in 1975 at "The Asilomar Conference." Recombinant DNA techniques to promote agricultural and medicine development took longer than planned due to unanticipated challenges and hurdles to obtain acceptable results, contrary to scientists' expectations at the time of Asilomar. Since the mid-1980s, however, an increasing number of goods such as hormones, vaccinations, therapeutic agents, and diagnostic tools have been created to enhance health [4].

1.1.Applications of Recombinant DNA Technology:

1.1.1. Food and Agriculture:

Recombinant DNA technology offers a wide range of applications, including the development of new enzymes that are appropriate for certain food processing conditions. Because of their unique functions and uses in the food industry, many essential enzymes such as lipases and amylases are accessible for specific manufacturing. Another significant accomplishment made feasible by recombinant DNA technology is the creation of microbial strains. A variety of microbial strains have been created that generate enzymes as a result of specialized engineering for protease synthesis. Certain fungus strains have been genetically engineered to decrease their capacity to produce hazardous compounds. Lysozymes are efficient bacteria-killing agents in the food industry. They prevent microbial organisms from colonizing. It's a good agent for storing foods including fruits, vegetable, cheese, or meat since it extends their shelf life. Immobilized lysozyme in polyvinyl alcohol films and cellulose may be used to prevent food spoilage bacteria from growing. The addition of lysozyme to fish skin gelatin gels extends the shelf life of food and inhibits the development of germs that degrade food. DspB, which was created from T7, can hydrolyze exopolysaccharides from Staphylococcus and E. coli. The bacterial population declines as a result of DspB's capabilities. The combined activity of serine proteases and amylases may be used to eliminate biofilms in the food industry. Diseases and Health: Recombinant DNA technology offers a broad range of applications in the treatment of illnesses and the improvement of health. The following sections detail some of the most significant advances in recombinant DNA technology for human health:

1.1.2. Gene Therapy is a kind of genetic therapy that involves the use:

Gene therapy is a cutting-edge medical procedure with therapeutic promise. The first successful report in the area of gene therapy for the treatment of a genetic illness offered a more secure path toward treating the most lethal hereditary disorders. This approach has shown to be effective in treating adenosine deaminase deficiency (ADA-SCID), a primary immunodeficiency. Several difficulties, such as maintaining patients on PEGylated ADA (PEG-ADA) during gene therapy and directing gene transfer to T-lymphocytes, were causes for poor outcomes at the start of this technique. Later, good results were achieved by targeting haematopoietic stem cells (HSCs) using an enhanced gene transfer technique and a myeloablative conditioned regime.

1.1.3. Antibodies or Their Derivatives Production:

Recently, plant systems have been utilized to produce and create various antibodies or their derivatives. Most significantly, out of a large number of antibodies and antibody derivatives, seven have met all of the criteria. CaroRx, Streptococcus mutants, an oral pathogen that causes tooth decay, may be detected by this antibody. T84.66 is a monoclonal antibody that may detect antigen carcinoembryonic, which is still regarded an affectively defined marker in epithelial malignancies. In transgenic soybean and Chinese Hamster Ovary (CHO) cells, a full-length humanized IgG1 known as anti-HSV and anti-RSV, which can act as a recognizing agent for herpes simplex virus (HSV)-2- glycoprotein B, has been produced. Antibodies from both sources have been proven to inhibit vaginal HSV-2 transmission in mice after topically applying them; if they functioned similarly in people, it would be a cheap and effective way to prevent infections spread via sexual encounters.

The idio type of malignant B cells in the well-characterized mouse lymphoma cell line 38C13 inspired the development of the 38C13 scFv antibody. Antibody administration to mice resulted in the development of anti-idiotype antibodies that identify 38C13 cells, assisting in the protection of mice against lymphoma cells, which is a fatal challenge. Transgenesis as well as agroinfiltration in tobacco transformed transiently enabled the synthesis of full-length monoclonal antibodies, scFv, and diabody derivatives in plants. Each of these antibodies may block testosterone synthesis induced by stimulated hCG in cells grown by LEYDIG, and uterine weight growth can be slowed in mice whose hCG activity is monitored. Antibodies may be used to diagnose cancers and treat them [5].

1.1.4. The Metabolism of Drugs is being investigated.

The importance of investigating the complex system of drug metabolizing enzymes involved in drug metabolism is critical for the appropriate effectiveness and effects of medicines. Heterologous expression, in which the enzyme's genetic information is generated in vitro or in vivo through gene transfer, has lately played a role in recombinant DNA methods.

Vaccines and recombinant hormones are being developed. In comparison to recombinant vaccinations, conventional vaccines have poorer effectiveness and specificity. Nasal transfer of adenovirus vectors encoding pathogen antigens is a fearless and painless way of transferring adenovirus vectors encoding pathogen antigens, as well as a fast and protection-sustaining strategy against mucosal infections. This works as a pharmacological vaccination, inducing an anti-influenza condition in the airway through transgenic expression. Traditional Chinese Medicines: Traditional Chinese Medicines have a significant role in diagnostics and treatments as an important component of alternative medicine. These medications are linked to ideas that are, to some degree, compatible with the concept of gene therapy. These medicines may be used to carry therapeutic genes and as co-administered medications. Along with the Ri plasmid, the transgenic root system offers significant potential for introducing other genes. It is usually carried in A. rhizogenes vector systems with modified genes to improve properties for particular uses. The cultures proved to be a useful tool for studying metabolic pathway biochemical characteristics and gene expression profiles. The turned cultures may reveal intermediates and important enzymes involved in the production of secondary metabolites[6].

1.1.5. Berries Contain Medically Important Compounds.

The rolC gene has resulted in an increase in the nutritional value of strawberries. This gene boosts sugar content as well as antioxidant action. Anthocyanin glycosylation necessitates the use of two enzymes: glycosyl-transferase and transferase. Some nutrition-related genes for various components in strawberries, such as proanthocyanidin, l-ascorbate, flavonoid, polyphenols, and flavonoid, are essential for genetic transformation to improve the component of interest. The anthocyanin components are controlled by the bHLH and FRUITE4 genes in raspberry, whereas flavonol is controlled by ERubLRSQ072H02. These genes may increase output and quality by undergoing certain transformations. All of the chemicals listed above have medicinal properties [7].

1.2.Environment:

Environmental problems may be solved through genetic engineering in a variety of ways. Working together, the University of Tennessee and Oak Ridge National Laboratory were the first to release genetically modified microorganisms for bioremediation purposes in the wild, such as the Pseudomonas fluoresces strain designated HK44. The modified strain has a transposon-based bioluminescence-producing lux gene linked inside a promoter, which resulted in enhanced naphthalene breakdown and a concomitant bioluminescent response. HK44 is a bioluminescence signaling reporter for naphthalene bioavailability and biodegradation, and it may be utilized as an online tool for in situ monitoring of bioremediation processes. Fibre optics and photon counting modules may be used to detect bioluminescent signal generation.

1.3.Plant Resistance Development and Phytoremediation:

For the detection and absorption of pollutants in drinking water and other samples, genetic engineering has been extensively utilized. The insertion of the AtPHR1 gene into the garden plants Torenia, Petunia, and Verbena, for example, altered their capacity to absorb Pi. The increased Pi absorption capacity of AtPHR1 transgenic plants may help with efficient phytoremediation in contaminated aquatic settings. A portion of the AtPHR1 gene was introduced into pBinPLUS, a binary vector with an improved cauliflower mosaic virus 35S promoter. This plasmid was given the name pSPB1898 and it was used to convert Petunia and Verbena using Agrobacterium tumefaction. Although AtPHR1 is successful in other plant species such as Torenia, Petunia, and Verbena, overexpression of AtPHR1 may impede posttranscriptional alteration of the endogenous AtPHR1 counterpart [8].

Energy-related applications Hydrogen generation is mediated by many microorganisms, including cyanobacteria, which is an environmentally beneficial energy source. The specific production is maintained by correctly using the necessary enzymes, which play an important part in product creation. However, sophisticated methods such as genetic engineering, nutritional and growth environment manipulation, mixed culture, metabolic engineering, and cell-free technologies have showed promise in increasing hydrogen generation in cyanobacteria and other biofuels. The commercialization of this energy source will help to keep the environment clean, which is impossible to do with traditional energy sources that emit CO_2 and other harmful pollutants. Cyanobacteria may also be genetically modified to convert CO_2 into reduced fuel molecules [9].

1.4.As a result, the following will be the current challenges and future prospects:

The fact that microbial cells are often employed in the manufacture of recombinant pharmaceuticals shows that they face a number of challenges that prevent them from effectively generating functional proteins, but they may be overcome by making changes to the cellular systems. Posttranslational modifications, activation of cell stress responses, instability of proteolytic activities, poor solubility, and resistance to expressing additional genes are all common challenges that must be overcome. Human genetic mutations result in protein synthesis deficits, which may be altered/treated by incorporating foreign genes to fill in the gaps and restore normal levels. In recombinant DNA technology, the utilization of Escherichia coli serves as a biological framework that enables producers to operate in regulated ways to create the necessary molecules using cost-effective methods.

By allowing for the study and modification of yeast genes not only in the test tube but also in yeast cells, recombinant DNA research holds tremendous potential for furthering our knowledge of yeast biology. Most significantly, by transforming yeast with DNA and cloning the genes using a number of selectable marker systems created specifically for this purpose, it is now feasible to return to yeast. These advances have combined to make true molecular as well as traditional genetic manipulation and analysis possible in yeast. The biological issues that recombinant DNA technology has most successfully solved are those that revolve on the structure and organization of individual genes. Recombinant DNA technology has lately seen significant advancements, resulting in significant shifts in research orientations and the opening of new avenues for sophisticated and exciting biosynthetic pathway study via genetic modification.

As a method of gene therapy, recombinant DNA technology provides a source of prevention and treatment for acquired genetic diseases in general. The creation of DNA vaccines is a novel method to provide protection against a variety of illnesses. The DNA transferred in this procedure includes genes that code for harmful proteins. In clinical trials, human gene therapy is mostly used to treat cancer. The primary emphasis of research has been on high transfection effectiveness in relation to gene delivery system design. The use of transfection for cancer gene therapy with low toxicity, such as in the cases of brain cancer, breast cancer, lung cancer, and prostate cancer, is currently being researched. Gene therapy is also being considered for kidney transplantation, Gaucher disease, hemophilia, Alport disease, renal fibrosis, as well as a few other illnesses [10].

2. DISCUSSION

Recombinant DNA technology is a significant advancement in science that has made life considerably simpler for humans. It has improved methods for biological applications such as cancer therapy, genetic illnesses, diabetes, and a variety of plant problems, including viral and fungal resistance, in recent years. The problems of enhancing products at the gene level may be tough to overcome, but they must be overcome for the future of recombinant DNA technology to be brighter. Pharmaceuticals, in particular, have significant challenges in producing high-quality goods since the changes made to a gene are not recognized by the body. Furthermore, growing product is not always a good thing since many variables may interfere and hinder it from succeeding. In terms of health, recombinant technology is assisting in the treatment of many illnesses that cannot be treated under regular circumstances, despite the fact that immune reactions obstruct excellent outcomes.

The emergence of genetically modified plants and other goods has raised many concerns. For example, it is self-evident that genetically modified plants may cross-breed with wild plants, damaging our biodiversity by spreading their "designed" DNA into the ecosystem. Furthermore, there are worries that genetic engineering may have harmful health consequences. To overcome such problems and address the concerns of ordinary people, more comprehensive study is needed in this area. Genetic engineering methods face a number of challenges that must be addressed by more precise gene augmentation tailored to the organism's DNA. This necessitates sequence similarity between the bacterial genome and the incoming DNA. Plasmid maintenance as well as reconstitution may be made simple. The transfer of genetic materials from one source into another has disastrous consequences for both safety and biodiversity

3. CONCLUSION

Recombinant DNA technology is a significant advancement in science that has made life considerably simpler for humans. It has improved methods for biological applications such as cancer therapy, genetic illnesses, diabetes, and a variety of plant problems, including viral and fungal resistance, in recent years. The importance of recombinant DNA technology in cleaning up the environment (phytoremediation or microbial remediation) and improving plant resistance too many unfavourable conditions (drought, pests, and salt) has long been acknowledged. It made tremendous advances not just in people, but also in plants and microbes. The problems of enhancing products at the gene level may be tough to overcome, but they must be overcome for the future of recombinant DNA technology to be brighter. Pharmaceuticals, in particular, have significant challenges in producing high-quality goods since the changes made to a gene are not recognized by the body. Furthermore, growing product is not always a good thing since many variables may interfere and hinder it from succeeding. In terms of health, recombinant technology is assisting in the treatment of many illnesses that cannot be treated under regular circumstances, despite the fact that immune reactions obstruct excellent outcomes.

Genetic engineering methods face a number of challenges that must be addressed by more precise gene augmentation tailored to the organism's DNA. A RecA-dependent mechanism would be used to integrate incoming single-stranded DNA into the bacterial chromosome. This necessitates sequence similarity between the bacterial genome and the incoming DNA. Plasmid maintenance as well as reconstitution may be made simple. The transfer of genetic materials from one source into another has disastrous consequences for both safety and biodiversity. The emergence of genetically modified plants as well as other goods has raised many concerns. For example, it is self-evident that genetically modified plants may cross-breed with wild plants, damaging our biodiversity by spreading their "designed" DNA into the ecosystem. Furthermore, there are worries that genetic engineering may have harmful health consequences. To overcome such problems and address the concerns of ordinary people, more comprehensive study is needed in this area.

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