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 regulating the expressions of target genes, desired traits might be improved in live organisms. However, in the recent age, this sector has exhibited distinctive impacts in terms of human life improvement. Crucial proteins necessary for health issues and nutritional needs may be generated securely, inexpensively, and in adequate quantities thanks to this technology. This technique offers a wide range of uses and has the potential to improve essential elements of life, such as health, food security, and resistance to a variety of harmful environmental impacts. Genetically engineered plants, particularly in agriculture, have boosted resistance to hazardous agents, increased product production, and demonstrated higher flexibility for better survival. Furthermore, recombinant medicines are now being utilised with confidence, and commercial clearances are being obtained quickly. Bioremediation and the treatment of severe illnesses are additional common uses of recombinant DNA technology, gene therapy, and genetic alterations. Because of the enormous development and wide variety of applications in the field of recombinant DNA technology, this review article focuses mostly on its significance and potential application in daily life. The obstacles of enhancing products at the gene level can be tough to overcome, but they must be overcome for the future of recombinant DNA technology to be brighter.***

KEYWORDS: ***Crop Improvement, Gene Therapy, Molecular, Recombinant DNA Technology.***

1. INTRODUCTION

Three variables have a significant impact on human life food scarcity, health difficulties, and environmental concerns. Aside from a clean and safe environment, food and health are essential human needs. Human food requirements are quickly growing as the world's population grows at a faster rate. Humans demand food that is both safe and affordable. Several human-related health concerns cause a high number of deaths across the world. Noncommunicable as well as communicable illnesses, including such cardiovascular disease, cancer, diabetes, AIDS/HIV, TB, and malaria, kill about 36 million people each year. Despite significant efforts, present global food production falls well short of human needs, and health-care facilities in third-world nations are considerably worse. Rapid industrialization has increased environmental contamination, and industrial wastes are permitted to mingle directly with water, affecting aquatic marine life and, indirectly, humans[1]. As a result, current technology must be used to overcome these challenges. Unlike traditional approaches to overcoming agriculture, health, and environmental issues through breeding, traditional medicines, as well as pollutants degradation through existing method, genetic engineering makes use of modern tools and approaches, including such molecular cloning and transformation, which take less time and produce more reliable results. In contrast to traditional breeding, which transmits a large number of both particular and nonspecific genes to the recipient, gene editing simply delivers a small block of desired genes to the target using different methods such as biolistic as well as Agrobacterium-mediated transformation. Homologous cell gene targeting or nuclease-mediated site-specific genome editing are both used to change plant genomes. Site-specific genome integration mediated by recombinases and oligonucleotide-directed mutagenesis are additional options[2].

1.1.Recombinant DNA technology:

Changing genetic material outside of an organism to achieve improved and desired traits in live creatures or as their products is referred to as recombinant DNA technology. This method entails inserting DNA fragments from a number of sources into a suitable vector with a desired gene sequence. Manipulation of an organism's genome can be done by adding one or more new genes as well as regulatory elements, or by recombining genes and regulatory elements to reduce or inhibit the expression of indigenous genes. Using restriction endonucleases for specific target sequence DNA sites, enzyme cleavage is used to get 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.

Paul Berg, Herbert Boyer, Annie Chang, as well as Stanley Cohen of Stanford University & University of California San Francisco created the first recombinant DNA (rDNA) molecules in 1973. Regulation or safe use of rDNA technology were considered in 1975 at "The Asilomar Conference." Recombinant DNA technologies to boost agricultural and medicine development took longer than planned due to unforeseen challenges and impediments to produce good 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[3].

1.2.Recombinant DNA Technology's Applications.

There are several applications of recombinant DNA technology such as crop improvement, gene therapy, in food, in Medicine show in Figure 1.

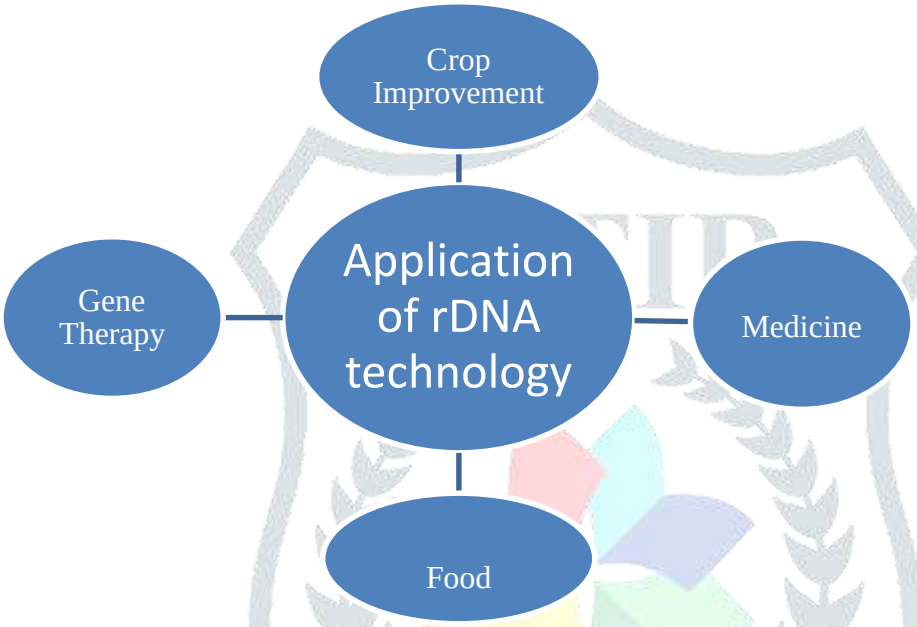

Figure 1: Illustrate the major application of recombinant DNA technology.

1.2.1. Recombinant DNA technology in Crop Improvement: Several possible uses of genetic engineering in crop enhancement are listed below:

1.2.1.1.Advance of Transgenic Plants:

Transgenic plants are genetically altered plants that carry foreign genes. This dna Recombinant technique can help with disease resistance, insect and pest resistance, herbicide and pesticide tolerance, drought tolerance, metal toxicity tolerance, male sterility induction for plant breeding, and quality enhancement. A good example is BT-cotton, which is resistant to bollworms.

1.2.1.2.Root Nodule Formation in Cereal Crops:

Leguminous plants have nitrogen-fixing microorganisms Rhizobium in their root nodules. In the root nodules, this bacterium transforms free air nitrogen to nitrates. Through genetic engineering techniques, the bacterial genes responsible for nitrogen fixation may now be transmitted to cereal crops such as wheat, rice, maize, barley, and others, making these plants capable of fixing atmospheric nitrogen as well[4].

1.2.1.3.Developments of C4 Plants:

Crop plants' photosynthetic efficiency can be improved, resulting in increased production. Converting C3 plants to C4 plants, which can be accomplished by protoplasm fusion or recombinant DNA technologies, can boost photosynthetic rate. C4 plants have a larger biomass output potential than C3 plants. Tropical or subtropical zones are where the majority of C4 plants (sorghum, sugarcane, maize, and certain grasses) are cultivated.

1.2.2. Applications in Medicine:

1.2.2.1.Manufacture of Antibiotics:

Penicillin & streptomycin are produced in large quantities using the fungus Penicillium as well as Streptomyces. To substantially improve the production of these antibiotics, genetically efficient strains of these fungus have been created.

1.2.2.2.Development of Hormone Insulin:

Insulin is a hormone derived from the pancreas of cows and pigs and is used by diabetics. The structure of this insulin differs somewhat from that of human insulin. As a result, around 5% of people experience allergic responses. The human gene for insulin synthesis has been inserted into bacterial DNA, and these genetically modified bacteria are utilised to produce insulin on a massive scale. This insulin does not trigger an allergic reaction[5].

1.2.2.3.Production of Vaccines:

Vaccines are currently made by infecting disease-causing microorganisms with antigen-coding genes. Antibodies like this prevent you against being infected with the same bacterium or virus.

1.2.2.4.Interferon production:

Virus-induced proteins generated by virus-infected cells are known as interferons. Interferon is an antiviral that acts as a first line of defense against viruses that cause severe illnesses such as breast cancer as well as lymphoma. Human blood cells generate natural interferon in extremely low quantities. As a result, it is also quite expensive. Interferon can now be produced at a considerably lower cost using recombinant DNA technology.

1.2.3. Food & 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 roles and uses in the food industry, several essential enzymes such as lipases and amylases are accessible for specific manufacturing. Another significant breakthrough 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 production. Certain fungus strains have been altered in order to limit their capacity to produce hazardous compounds. Lysozymes are efficient bacteria-killing agents in the food industry. They keep microbial organisms from colonizing. It's a good agent for storing foods including fruit, vegetable, cheese, and meat since it extends their shelf life. Immobilized lysozyme in polyvinyl alcohol films and cellulose can be used to prevent food spoilage bacteria from growing. Diseases as well as Health Recombinant DNA technology has a wide range of applications in the treatment of illnesses and the improvement of health. The sections that follow detail the major advances in recombinant DNA technology that have improved human health[6].

1.2.4. Gene Therapy.

Gene therapy is a cutting-edge medical procedure with therapeutic promise. The first successful report in the field of gene therapy for the treatment of a genetic condition established 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 obstacles, such as maintaining patients on PEGylated ADA during gene therapy and directing gene transfer to T-lymphocytes, were causes for poor outcomes at the start of this method. Later, successful results were achieved by targeting haematopoietic stem cells (HSCs) using an enhanced gene transfer procedure as well as a myeloablative conditioning regime.

1.2.5. The Metabolism of Drugs: An Investigation:

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

recombinant DNA methods.

1.2.6. Vaccines or 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 approach of transferring adenovirus vectors encoding pathogen antigens, as well as a fast and protection-sustaining strategy against mucosal infections. This operates as a pharmacological vaccination, inducing an anti-influenza condition in the airway via transgenic expression. Human follicle-stimulating hormone (FSH) may now be produced in vitro using recombinant DNA technology. FSH is a complicated heterodimeric protein, and a specific cell line from eukaryotes has been chosen to express it. Recombinant DNA technology has made it possible to treat assisted reproduction by promoting follicular development. r-FSH is being used to treat a large number of individuals. Most notably, recombination of r-FSH and Luteinizing Hormone (LH) proved successful in enhancing ovulation and conception.

1.2.7. Berries Contain Medically Important Compounds.

The function gene has been used to improve the nutritional content 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 diverse components in straw-berries, such as proanthocyanin, l-ascorbate, flavonoid, polyphenols, and flavonoid, are crucial for genetic transformation to improve the component of interest.

1.2.8. Energy Applications.

Hydrogen generation is mediated by several microorganisms, including cyanobacteria, which is an environmentally beneficial energy source. The specific production is maintained by appropriately using the needed 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 achieve with traditional energy sources that emit CO2 and other dangerous pollutants. Cyanobacteria can also be genetically modified to convert CO2 into reduced fuel molecules. This strategy has worked for a wide range of commodity compounds, primarily energy carriers like short and medium chain alcohols[7].

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 contains genes that code for harmful proteins. In clinical trials, human gene therapy is mostly used to treat cancer. The major focus 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.

2. LITERATURE REVIEW

Amy Y. Shen et al. studied the creation of an appropriate cell line is required for the production of recombinant proteins in animal cell culture for research, clinical development, or commercial reasons. The type of the protein to be expressed, the quantity of material required, as well as the timeframe during which continuous production is required all influence the choice of expression system and mode of expression. Large, heavily disulfide bonded or glycosylated proteins are typically produced using mammalian expression methods. The type of the protein to be expressed, the quantity of material required, as well as the timeframe during which continuous production is required all influence the choice of expression system and mode of expression. Large, heavily disulfide bonded or glycosylated proteins are typically produced using mammalian expression methods[8].

Valmik K. Vyas et al. discussed about Candida albicans is a pathogenic yeast that causes high-mortality mucosal and systemic infections. The lack of simple molecular genetics has been a key roadblock in pathogenesis research. Genetic engineering is difficult due to the lack of meiosis and plasmids, especially for

crucial tasks and gene families. We describe a CRISPR method for C. albicans that overcomes several of the organism's genetic engineering challenges. CRISPR-induced mutations may be directed to target genes with a high frequency, making homozygous gene knockouts straightforward to isolate even without selection. Furthermore, the method allows strains with mutations in numerous genes, gene families, and genes that encode critical activities to be created. This CRISPR method also works in a new clinical isolate with an unknown ploidy. Our technique revolutionizes the capacity to modify Candida's DNA and opens up a new window into the pathogen's biology[9].

Peter T. Lomedico et al. discussed about A fast technique to assess the stages along the gene expression pathway is required to employ recombinant DNA technology to functionally examine mutations incorporated into cloned eukaryotic genes. Because cloned rat insulin genes are not successfully transcribed after transfection into diverse cell lines, I wondered if the insulin gene might be placed into a transcriptional unit that operates in all mammals to drive expression. This recombinant plasmid regulates the synthesis of properly spliced and polyadenylated insulin mRNA that acts in the production and secretion of rat proinsulin, according to tests performed immediately after its introduction into mammalian cells. This method allows for the fast examination of cloned in vitro-engineered mutations as well as the programming of eukaryotic cells to produce proteins that they would not ordinarily produce[10].

3. DISCUSSION

Recombinant DNA technology is a significant advancement in science that has made life considerably simpler for humans. It has expanded techniques for biological applications such as cancer therapy, genetic illnesses, diabetes, and a variety of plant problems, including viral or fungal resistance, in recent years. The importance of recombinant DNA technology in cleaning up the environment (phytoremediation as well as microbial remediation) and improving plant resilience to many unfavorable conditions (drought, pests, and salt) has long been acknowledged. Three variables have a significant impact on human life: food scarcity, health difficulties, and environmental concerns. Aside from a clean and safe environment, food and health are essential human needs. Human food requirements are quickly growing as the world's population grows at a faster rate. Humans demand food that is both safe and affordable. Several human-related health concerns cause a high number of deaths across the world. Noncommunicable or communicable illnesses, such as cardiovascular disease, cancer, diabetes, AIDS/HIV, TB, malaria, and a variety of others, kill around 36 million people each year. It made tremendous advances not just in people, but also in plants and microbes. The obstacles of enhancing products at the gene level can be tough to overcome, but they must be overcome for the future of recombinant DNA technology to be brighter.

4. CONCLUSION

Recombinant DNA technology is a significant advancement in science that has made life considerably simpler for humans. It has expanded techniques for biological applications such as cancer therapy, genetic illnesses, diabetes, and a variety of plant problems, including viral or fungal resistance, in recent years. The importance of recombinant DNA technology in cleaning up the environment (phytoremediation and microbial remediation) and improving plant resilience to many unfavorable conditions (drought, pests, & salt) has long been acknowledged. It made tremendous advances not just in people, but also in plants and microbes. The obstacles of enhancing products at the gene level can be tough to overcome, but they must be overcome for the future of recombinant DNA technology to be brighter. Pharmaceuticals, in particular, have severe 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 because many variables might interfere and hinder it from succeeding. The obstacles of enhancing products at the gene level can be tough to overcome, but they must be overcome for the future of recombinant DNA technology to be brighter.

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