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THE MOST EXCITING INNOVATION IN GENE THERAPY

Medicine

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ABSTRACT

Gene therapy is viable, available and reliable. Gene therapy or human gene transfer is the process of replacing defective genes in a cell with healthy genes. The defective genes produce genetic diseases. People with genetic disorders such as Alzheimer's disease, Huntington's' disease and Parkinson's disease are among those who require gene therapy. The key objective a modern gene therapy involves. Replacing defective (mutated) gene/s that causes disease with a healthy copy of the gene/s. Introducing a new functional copy of gene/s into the cells to help fight a disease . Inactivating, or "knocking out," (silencing; gene silencing) a mutated gene that is functioning improperly. In the present scenario gene therapy is not only limited to therapeutic delivery of gene/s but also regulation of gene functioning as well.

KEYWORDS

Gene silencing ,Multiple Sclerosis, Parkinson's disease, Traumatic Brain Injury, Spinal Cord Injury, Dystopia, Chronic Regional Pain Syndrome, Motor Neuron Disease/Amyotrophic Lateral Sclerosis

Introduction to Gene Therapy

The gene therapy is defined as the therapeutic delivery of nucleic acid (gene/s) via artificial means to the cells precisely nucleus to cure diseases (1). The gene therapy is an experimental technique provides a platform to edit and or replace a defective gene responsible for onset and progression of a disease/disorder (2). In the present medical innovations, gene therapy stands a novel therapeutic technology enable us to edit and or replace a defective gene/s responsible for the disease (3). The nucleic acid located in the nucleus is the center for genetic information which transcribed and translated into meaningful outcomes via various molecules including RNA and proteins. The flow of genetic information from nucleic acid to protein referred as central dogma and essential for vital metabolic pathways (4). There are several possibilities and instances where the flow of genetic information becomes abnormal at DNA, RNA and at protein level result a defective metabolic pathway triggers the onset of a diseases/disorder. Hence, to find a cure for such diseases, one must focus on the defective gene/s (5). Further, there are several diseases including cancer are the result of abnormal over expression of multiple genes need regulation. (6). The proteins are translated products of nucleic acid and involve in physiology at a different level, one as a structural unit and second as functional. The proteins are highly dynamic molecules in the biological world and associated with numerous vital functions including signaling, acting as an enzyme, catalyzing metabolic pathways and structural components as well (6). The functionality of a protein molecule depends on the gene itself and flow of genetic information. (7). There are several diseases/disorders which are the result of defective gene/s and translated proteins. (8) In several instances, it becomes essential to remove a defective gene/s and or control gene activity as well (9). Here, DNA and RNA delivered to a cell have different mechanism; DNA replaces a defective gene with the fresh copy while RNA used for gene silencing (10). It is often reported that during cancer and other immunological disorders several genes switch to over-expression resulting expression of proteins trigger diseases. (11). Hence, gene therapy is not limited to therapeutic delivery but also negative regulation of various genes responsible for diseases/disorders. Targeted Diseases and disorder The targeted diseases and disorders for gene therapy can be broadly classified as metabolic disorders, genetic disorders, neurological disorders, and cancer (12). Here, nucleic acid alone (antisense DNA and RNA) and or in conjugation with other biomolecules including proteins and vaccine, ribozymes, etc. (13). In the present scenario due to innovations in cutting-edge biomedical technologies gene therapy had a wide spectrum of targeted diseases. Here is a list of diseases/disorders where gene therapy finds an opportunity for cure and management (14). There is an increasing list of genetic diseases need immediate attention and development in gene therapy based treatments. The high-risk genetic diseases are hemophilia, SCIDs, sickle cell anemia, cystic fibrosis, muscular dystrophy, phenylketonuria, type 2 diabetes (15), Ataxia and Turner syndrome, etc. Here, both in-vivo and ex-vivo gene therapies are required for complete management of genetic disorders (16). The neurological disorders need more sophisticated treatment option as viral tissue lack capacity of regeneration. Here, gene therapy can surely play a vital role in the management of several prevailing neurological disorders including Multiple Sclerosis, Parkinson's disease, and Traumatic Brain Injury. Spinal Cord Injury, Dystonia, Chronic Regional Pain Syndrome, Motor Neuron Disease/Amyotrophic Lateral Sclerosis (17). 3. Metabolic diseases/disorders Gene therapy has recently shown great promise as an effective treatment for some metabolic diseases caused by genetic defects in both animal models and human clinical trials (18). The metabolic disorders phenylketonuria, inborn error metabolism, insulin resistance, mitochondrial diseases, myopathy, encephalopathy, lactic acidosis, etc. The metabolic disorders have multiple inputs and difficult to cure via conventional therapeutics need an alternate, and here gene therapy could be an option (19). Apart from genetic and metabolic disorders, gene therapy had shown increasing potential in finding a cure for infectious diseases as well. (20).Mostly, diseases caused by viruses including HIV, hepatitis virus (B, C), influenza; H1N1, H5N1, human papillomavirus, human T-cell lymphotropic virus, Herpes Simplex Virus, EpsteinBarr Virus, Cytomegalovirus, and Mycobacterium tuberculosis are a major target under gene therapy for infectious diseases (21). There are several approaches to gene therapy correcting defective gene/s associated with particular diseases. Somatic gene therapy is the transfer of genes into the somatic cells of the patient, such as cells of the bone marrow, and hence the new DNA does not enter the eggs or sperm (22).

Adenoviruses contain DNA as genetic material infects a cell, lose their protein coat, and transfer DNA into the nucleus, where it is transcribed (23).

This DNA does not integrate into the host genome, and thus, its effects are transient. Hence, multiple administrations of the vector are usually required (24). The advantages of the herpes simplex virus (HSV) are its large size, the wide spectrum of action and the continuous expression of genes from long-lived infection. (25)

SIGNIFICANT GAP IN RESEARCH

Advantage of Gene Therapy Comparing with conventional therapies gene therapy offers a cure for targeted diseases and disorders rather than symptomatic relief. The goal of gene therapy replaces a defective and malfunctioning gene with a fresh and functional copy. The gene therapy offers therapeutic gene delivery to a large number of human diseases including genetic, neurological, infectious and metabolic (26). The present cutting-edge molecular biology techniques are capable of harvesting therapeutic gene, molecular cloning, designing ideal delivery vehicles and targeted therapeutic delivery. (27). The real-time PCR allows us to determine the level of gene expression which further relates functionality of targeted gene delivered for targeted gene therapy (28).

WHERE THE RESEARCH GO NEXT

Now, nanotechnology is playing a crucial role in the delivery of therapeutic gene/s to the sub cellular compartment of the cell where other delivery vehicles often get fail. The nanoparticles are comparatively easy to synthesize and allow maximum cargo up to 20 kb of nucleic acid (DNA and RNA) offer promise for the disease caused by the failure of multiple genes (29). At the same time, several reports demonstrated the cytotoxic effect of nanoparticle and viral vehicle used for therapeutic delivery of gene/s. Here, liposome emerged as a novel tool for the gene delivery with negligible side effects (30). Liposomes are also providing an ease in the integration with biological membrane and release of cargo to cell and sub cellular compartment (31). The incorporation of biocompatible liposome minimizes cellular and tissue-specific toxicity caused by nanoparticles. Additionally, the use of liposome as delivery vehicle further combat the risk of oncogenic and other inflammatory consequences caused by viral vehicles (32).

CURRENT DEBATE

With the available technology gene therapy is aiming new heights in modern medicine and upcoming cutting-edge biological innovations further enhance the efficiency of therapy. With these innovations and incorporation of multidisciplinary approaches, gene therapy seems more feasible and effective. Disadvantage of Gene Therapy Since the beginning of gene therapy, it was believed that technology provides a complete cure for targeted disease and disorder rather than symptomatic relief as conventional medicine do. In gene therapy, DNA encoding a therapeutic protein is packaged within a "vector," which transports the DNA inside cells within the body. Despite the several clinical trials many diseases such as X-linked SCID, Parkinson's disease, multiple myeloma, chronic and acute lymphocytic leukemia, and adrenoleukodystrophy failed for approved by FDA . The major challenge in gene therapy is the delivery of therapeutic gene/s to targeted cells. The available delivery vehicles for gene delivery are either viral vector or non-viral vector, and both are associated with several limitations. The issue with viral vehicles (retroviruses, adenovirus, and adeno-associated viruses) is their nature itself (33).

The viral vehicles are the first choice for gene therapy for a large number of disease and their pathogenic nature remain a major hurdle. The viral vehicles do have a great capacity to hold nucleic acid cargo and higher transfection capacity (34). But at the same time, the design of virus into delivery vehicle remains associated with a great risk of infection. On the contrary, non-viral vehicles which are less pathogenic but have limited DNA carrying capacity and transfection efficiency. Further, hunting of a therapeutic functional copy of the gene is itself a major challenge in gene therapy. The design of recombinant cassette for gene therapy requires a proper orientation of gene (downstream to the promoter region), and its replacement with the defective gene in the nucleus need a precise nucleotide sequence (35). The delivery of foreign gene/s with viral and non-viral vehicle often activates immune reaction which also remains a major challenge for gene therapy. To counter immune complications, a massive dose of several immunosuppressive drugs remain in clinical use (36). There are difficulties in real-time monitoring of gene therapy and functionality of delivered gene with the cell. Despite a long history of more than 30 years, very limited numbers of clinical trials gain attention for FDA approval which also questions the success of the technology. Considering clinical aspect of gene therapy, somatic gene therapy is quite feasible, but germ line therapy is associated with several ethical issues (37).

MAJOR ADVANCES AND DISCOVERIES

The viral vehicles are largely associated with cellular toxicity and result switching various inflammatory, metabolic pathways. Further, gene therapy involving viral vehicles are associated with oncogenic properties and turn on various oncogenic genes. Multi-gene disorders such as heart disease, high blood pressure, Alzheimer's disease, arthritis, and diabetes are quite difficult to treat through this therapy as conditions or disorders that arise only from mutations in a single gene are the best candidates for gene therapy (38). It is also reported that most of the gene therapies are short-lived, and one of the reasons for the failure of previous clinical trials studies carried out in the past. The gene therapy for infectious disease requires a significant genetic rearmament of virulence genes and delivery vehicle raised safety concern in gene therapy. Non-viral vehicles including liposome, nanoparticle, and polysomes are associated with limitations of gene delivery as well (39). The management of genetic disorders require gene therapy in germ line cell, and it's very difficult to use a human in the laboratory for clinical trials. Further, there are increasing ethical issues in the design of viral particles into a delivery vehicle for gene therapy as well (40). The gene therapy requires a great deal of scientific expertise and hence cost factor of such therapy itself a big challenge in the present scenario. All these challenges need to resolve to bring technology for clinical applications. Future Prospects The ability to deliver a functional copy of gene/s to a cell provides a new avenue to modern therapeutics. The cutting-edge molecular biology techniques are playing a vital role in defining precision and accuracy in delivery of therapeutic gene/s. Despite the earlier failure in gene therapy new advancements in molecular biology established several milestones in disease management. As a result, Human gene therapy (HGT) emerged as a new therapeutic approach utilizing molecular biology and biotechnology revolution. However, still, we are in beginning era of gene therapy as the efficacy of technology depends on several factors including an ideal carrier, specific gene delivery and functional aspect of therapeutic gene/s in a living system. The most crucial task of gene therapy in present time is their costs which need to resolve for large-scale clinical studies. The experimental aspect of gene therapy is quite complex and involves several molecular events including hunting of functional therapeutic gene/s, molecular cloning cascade, finding an ideal vehicle and loading on a delivery vehicle, transfection and precise delivery to a targeted cell. The real-time monitoring of functionality of delivered gene/s defines the success of gene therapy.

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