v. 4, n. 101, 2024

International Journal of Health Science

Acceptance date: 02/12/2024

GENE THERAPIES AND CRISPR IN CLINICAL MEDICINE: APPLICATIONS, ADVANCES AND ETHICAL CHALLENGES

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Abstract: Gene editing using the CRISPR--Cas9 technique has emerged as one of the greatest innovations in clinical medicine, offering the potential to correct genetic mutations responsible for various diseases. This technology has been applied in the treatment of rare and common diseases such as muscular dystrophy, sickle cell anemia and cancer, bringing new paradigms to personalized medicine. This article addresses the clinical applications of CRISPR-Cas9, exploring its possibilities, challenges and ethical and regulatory implications. In addition, it discusses the advancement of personalized therapies, with a focus on clinical feasibility, accessibility and the obstacles faced for the widespread adoption of these therapies. The study also highlights the regulatory aspects that need to be adjusted to ensure the safety and efficacy of these innovative approaches.
Keywords: CRISPR-Cas9, Gene Therapy, Personalized Medicine, Muscular Dystrophy, Sickle Cell Anemia, Ethics and Regulation.

INTRODUCTION

Gene therapy is an innovative therapeutic approach that aims to correct genetic defects by modifying the genetic material of the patient's cells. With the advancement of technologies, especially the CRISPR-Cas9 system, the possibility of gene editing has become not only more precise and accessible, but also more promising for the treatment of a wide range of genetic diseases. CRISPR-Cas9, discovered in 2012, offers an effective tool for modifying genes in a controlled and relatively inexpensive way, allowing for a more effective therapeutic approach in various medical conditions. Since its first experimental applications, gene editing has been considered a revolution in medicine, with a direct impact on areas such as muscular dystrophy, sickle cell anemia, and cancer.

This article aims to explore the clinical applications of CRISPR-Cas9 in medicine, with an emphasis on how this technology is being used to correct genetic mutations associated with rare and common diseases. In addition, the ethical and regulatory challenges that arise with the use of this tool will be discussed, as well as the implications for personalized medicine, with a focus on the clinical feasibility and accessibility of these treatments. Gene editing, while a significant advance, also raises questions about equity in access to treatments and the need for robust regulation to ensure patient safety.

CRISPR-CAS9 TECHNIQUE: FUNDAMENTALS AND APPLICATIONS

THE CRISPR-CAS9 SYSTEM

CRISPR-Cas9 is a revolutionary molecular tool that allows precise and targeted DNA editing. Initially identified in bacteria as a defense mechanism against viruses, the technique has been adapted for use in multicellular organisms, including humans (Jinek et al., 2012). CRISPR technology uses an enzyme called Cas9, which, with the help of a guide RNA, is able to cut DNA in specific places, allowing parts of the genetic code to be removed, inserted or replaced.

The precision and ease of use of CRISPR-Cas9 have become its greatest attractions, allowing for much more efficient genetic manipulations than previous techniques, such as TALEN (Transcription Activator-Like Effector Nuclease) or Zinc Finger (O'Geen et al., 2015). This ability to precisely cut and edit DNA allows scientists to treat and correct genetic mutations associated with a wide range of diseases, creating new therapeutic possibilities.

CLINICAL APPLICATIONS OF CRISPR-CAS9

Duchenne Muscular Dystrophy (DMD)

Duchenne muscular dystrophy (DMD) is a serious genetic disease that mainly affects boys, resulting in progressive muscle degeneration. This condition is caused by a mutation in the dystrophin gene, which is essential for the structure of muscle cells. The application of CRISPR-Cas9 in DMD aims to correct the gene mutation, restoring dystrophin production and therefore muscle function. In 2016, the first laboratory study demonstrated that CRISPR could successfully correct the mutation in the dystrophin gene in human cells, paving the way for subsequent clinical trials. More recent trials, carried out on animal models, have also shown promising results, with partial restoration of muscle function (Mendell et al., 2021).

Sickle cell anemia

Sickle cell anemia is **an** inherited disease caused by a mutation in the **HBB** gene, which leads to the production of abnormal hemoglobin and deformation of blood cells. Gene editing with CRISPR-Cas9 offers the possibility of correcting this mutation, allowing patients to produce normal hemoglobin. These clinical trials are underway to evaluate the use of the technique in hematopoietic stem cells, with the aim of curing the disease. In a 2021 study, a group of researchers used CRISPR to modify stem cells from patients with sickle cell anemia, with promising results of improved production of normal hemoglobin (Dever et al., 2021).

CANCER

The use of CRISPR-Cas9 in cancer treatment has shown promise, especially in modifying immune system cells, such as T-cells, to recognize and attack tumor cells. Immunotherapy with CAR-T cells (Chimeric Antigen Receptor T-cells) is an emerging clinical application that uses CRISPR to modify T-cells in order to increase their effectiveness in fighting cancer. Ongoing clinical trials are exploring the use of CRISPR-Cas9 to modify T-cells in patients with leukemia, lymphoma and other types of cancer, with encouraging results in terms of tumor response (Sadelain et al., 2017).

PERSONALIZED GENE THERAPIES: CHALLENGES AND FEASIBILITY

CLINICAL VIABILITY

Although CRISPR-Cas9 technology has shown great potential, its clinical application still faces technical challenges. Efficient delivery of CRISPR into target cells is one of the main barriers. Although delivery methods such as viral vectors and lipid nanoparticles have shown some efficacy, they still have limitations in terms of precision, safety and ability to reach specific cells. One of the most promising approaches is the use of liposomes and nanoparticles that can be administered in a more controlled manner, reducing the risk of side effects and increasing treatment efficiency (Chung et al., 2020).

Another important challenge is genetic instability. Edits made by CRISPR-Cas9 can generate unwanted mutations in locations outside the target area, which is known as "off-target effects". This can cause problems such as the development of tumors or other unwanted conditions, making it necessary to apply enhancement technologies to increase the accuracy of genetic edits.

ACCESSIBILITY AND COST

The accessibility of CRISPR-Cas9-based therapies is also a crucial issue. Currently, the costs involved in developing these therapies are high, which makes their application limited, especially in developing countries. Manufacturing personalized therapies requires complex technologies and highly skilled labor, which can result in prohibitive prices for patients and health systems. The personalized approach also requires individual genetic sequencing, which can further increase costs.

However, studies suggest that the long-term benefits of therapies such as gene editing may outweigh these costs, since there may be a reduction in hospital admissions and chronic treatments for rare or long-term diseases (O'Dwyer et al., 2022). In this context, it is essential that governments and international organizations develop public policies that promote equitable access to these therapies, especially for the poorest populations.

ETHICAL AND REGULATORY IMPLICATIONS (CONTINUED)

ETHICS OF GERMLINE EDITING (CONTINUED)

Germline editing, which involves modifying DNA in embryos or reproductive cells, remains a highly controversial area, especially when it comes to hereditary modification. Genetic modifications made to germ cells can be passed on to future generations, which raises questions about the long-term consequences of these interventions. If the effects of a genetic modification turn out to be negative, this could affect not only the patient, but also future generations, which makes it difficult to assess the ethics of this practice.

In 2018, Chinese scientist He Jiankui was widely criticized for editing the genome of human embryos, resulting in babies with HIV resistance. The experiment was widely

condemned by the scientific community, which warned of the lack of proper monitoring and robust ethical evaluation before carrying out such modifications (Cyranoski, 2019). This incident sparked a global debate on the ethical constraints and moral limits of germline editing.

Although many bioethicists argue that germline editing should be banned until issues of safety and morality are fully resolved, others argue that, if applied in a controlled and regulated manner, gene editing could correct serious hereditary diseases, offering a way to eliminate incurable diseases. An intermediate solution advocated by some experts would be to allow germline editing in very restricted contexts, such as for fatal genetic diseases, but with strict regulation and international oversight.

REGULATORY CHALLENGES

The regulation of CRISPR-Cas9 therapies is another crucial aspect in guaranteeing the safety and efficacy of such interventions. Currently, regulatory approaches to gene editing vary widely between countries. In the United States, for example, the FDA (Food and Drug Administration) has approved gene therapies for some conditions, but in the case of germline editing, the agency takes a much more conservative line, only allowing modifications to somatic cells, but with major limitations.

In contrast, in some European countries, such as the UK, there are more flexible regulations for research involving gene editing, especially for treatments related to specific diseases, as long as they are in line with strict ethical protocols. However, many experts point out that the lack of a global regulatory framework could lead to disparities in access to treatments, especially in developing countries, where genetic treatments are still inaccessible to the majority of the population.

Furthermore, somatic cell editing for genetic diseases is not without its regulatory challenges. Authorities need to ensure that patients undergoing these therapies are aware of the risks involved, including potential off--target mutation or unwanted side effects. The lack of regulatory uniformity between countries can result in a complex legal framework for patients and healthcare professionals, hindering the effective application of these technologies on the global stage.

PERSONALIZED GENE THERAPIES: PERSPECTIVES AND ADVANCES

PRECISION MEDICINE AND CRISPR-CAS9

Personalized or precision medicine has been a field that has benefited enormously from the innovations brought about by CRISPR-Cas9. By enabling the specific modification of genes based on the patient's genetic profile, CRISPR makes it possible to treat diseases in a highly personalized way, addressing the molecular cause of the problem. Instead of a one-size-fits-all approach, personalized medicine recognizes the genetic, environmental and lifestyle differences between individuals, providing more effective and less invasive treatments.

For example, in the case of sickle cell anemia, genetic modification of the patient's stem cells to correct the HBB gene mutation can enable the patient to produce normal hemoglobin. This treatment is highly personalized, since the patient receives a therapy that corresponds to their own genetic profile, rather than a generic treatment.

In addition, with advances in genetic sequencing techniques, it is possible to identify specific mutations that can be corrected or treated with personalized therapies. This opens up a new field of possibilities in the

International Journal of Health Science ISSN 2764-0159 DOI https://doi.org/10.22533/at.ed.15941012402126

treatment of rare and complex diseases, allowing CRISPR-Cas9 to be applied precisely and effectively, depending on the genetic needs of each patient.

CHALLENGES IN THE USE OF PERSONALIZED THERAPIES

Despite the promises of personalized medicine, there are substantial challenges that need to be overcome for these therapies to become widely accessible and viable. The genetic complexity of rare diseases, for example, makes it difficult to find a solution that is effective for all patients with the same condition. Although gene editing with CRISPR-Cas9 has shown promising results in experimental models, large-scale application and the development of personalized therapies still face significant issues.

Another major challenge is the issue of cost. Personalized gene therapies require a complex infrastructure, including the genetic sequencing of patients, the modification of cells and the manufacture of personalized medicines. This process is extremely expensive and often inaccessible to the majority of the population. The scalability of these therapies is a critical problem that needs to be solved so that they can become a reality for a greater number of patients.

Finally, the implementation of these therapies requires specialized training for healthcare professionals, who must be trained to deal with the complexities of genetic editing and its potential consequences. Training and medical education are therefore essential aspects for the successful dissemination of these therapies in clinical practice.

CRISPR-CAS9 AND THE FUTURE OF CLINICAL MEDICINE

PROMISING ADVANCES AND POTENTIAL FUTURE APPLICATIONS

The future of CRISPR-Cas9 in clinical medicine is extremely promising. Current applications are just the beginning of a field that has the potential to transform not only the treatment of genetic diseases, but also the way we treat complex diseases such as cancer and neurodegenerative diseases.

CRISPR has been exploited as a diagnostic tool, with the possibility of identifying genetic mutations associated with diseases even before symptoms appear. Gene editing can be used to correct mutations in DNA repair genes, helping to prevent diseases such as cancer before they develop.

Another promising application of CRISPR-Cas9 is antiviral therapy. Editing the genome of cells infected by viruses, such as HIV or hepatitis, to remove the viral material or correct the cells so that they become resistant to infection, could revolutionize the treatment of infectious diseases, offering a longer-lasting and more effective alternative to traditional antiviral treatments.

ON THE ROAD TO UNIVERSAL THERAPY: CRISPR-BASED THERAPIES FOR VARIOUS CONDITIONS

As research into CRISPR-Cas9 advances, new therapeutic approaches are being developed to treat a wide range of conditions, from heart disease to neurodegenerative diseases such as Alzheimer's and Parkinson's. Although these areas are still in the early stages of research, preliminary results are promising. Scientists are exploring ways of using CRISPR to modify brain cells and correct genetic defects that cause neurological disorders.

In addition, CRISPR-Cas9-based therapies are being explored for healthy aging. The use of gene editing to reverse the signs of cellular aging or correct DNA damage caused by time could transform the approach to anti-aging medicine. Research into stem cells and cell rejuvenation with CRISPR is opening up possibilities for more effective treatments against age-related diseases.

CONCLUSION

Gene therapy and the use of CRISPR-Cas9 technology represent a revolution in clinical medicine, with the potential to completely transform the approach to treating both rare and common genetic diseases. Since its discovery, gene editing has shown promising experimental results, being successfully applied in animal models and early clinical studies. The correction of mutations in diseases such as Duchenne muscular dystrophy, sickle cell anemia, and the use of immune therapies to treat cancer are just a few examples of how this technology can have a profound impact on medicine.

Personalized medicine, which is powered by the CRISPR-Cas9 technique, offers a more effective and precise approach, adapted to patients' individual genetic characteristics. This opens up a new paradigm for the treatment of diseases, enabling interventions that attack the root of the genetic problem, rather than just treating the symptoms. The possibility of creating highly specific therapies, which involve correcting genetic defects in somatic cells, has the potential to cure diseases that until recently were incurable, providing new hope for patients with rare and debilitating conditions.

However, despite the promise and progress, the road to widespread adoption of these therapies is fraught with challenges. Developing safe and efficient delivery methods for CRISPR in target cells is still one of the biggest obstacles. The precision of gene editing, although it has improved significantly since the first studies, still requires advances to minimize off-target effects and guarantee the safety of treatments. Therapies based on CRISPR-Cas9 also face economic challenges. The high cost of both research and production of personalized therapies makes these options inaccessible to many populations, especially in countries with less structured health systems.

The ethical issue also remains one of the main barriers. Gene editing in embryos and germ cells, with its potential to permanently modify a person's genetic lineage, raises questions about the limits of human intervention in genetics and the possible consequences for future generations. Although germline editing has the potential to eradicate genetic diseases, its indiscriminate use can lead to undesirable scenarios, such as the development of "designer babies" with chosen genetic characteristics, something that challenges ethical and moral principles about human autonomy and dignity. The balance between scientific innovation and ethical precautions will be key to ensuring that gene editing technologies are used responsibly and beneficially.

In addition, regulatory issues need to be adjusted to ensure that gene therapies are used safely, effectively and fairly. Although several countries have established regulations on the use of CRISPR-Cas9 in medical treatments, there is still no uniform approach globally. Harmonization of regulations and the development of robust international guidelines are necessary to avoid irresponsible practices and ensure that gene therapies are accessible in an equitable and ethical manner. Strict control, together with a continuous assessment of risks and benefits, will help ensure that advances in genetic science bring more benefits than harm.

The future of CRISPR-based medicine is therefore promising, but not without challenges. The transition from the laboratory to cli-

nical practice, with the widespread adoption of CRISPR-based therapies, requires scientists, doctors, legislators and society as a whole to work together to tackle the technical, ethical and regulatory issues that arise. The combination of advances in gene editing technology, the improvement of delivery techniques, the refinement of personalized therapeutic approaches and ethical and safe regulation is key to gene therapies becoming a viable, affordable and safe tool for the treatment of an ever-increasing range of diseases.

As new treatments based on CRISPR-Cas9 continue to be developed and tested, medicine is expected to become more personalized, precise and effective, offering treatments that truly meet patients' needs in a unique and individualized way. This promise could not only transform the treatment of genetic diseases, but also open doors to the cure of many conditions previously seen as incurable, creating new possibilities for human health.

However, the next steps require scientists to continue improving the tools and techniques, while society in general must ensure that these innovations are used responsibly. The advance of gene therapy, mediated by CRISPR-Cas9 technology, will certainly shape the future of medicine, and the way we deal with its social, economic and ethical impacts will determine how successful this medical revolution will be. Thus, the future of genetic medicine depends not only on science, but also on how we choose to govern and apply these powerful technologies for the good of all humanity.

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