CRISPR Technology in Neurodegenerative Disease Research: A Focus on Alzheimer's

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Abstract

Neurodegenerative diseases, among which is Alzheimer's disease, are among the biggest challenges for modern medicine due to their progressive nature and lack of curative treatment approaches. Recent advances in CRISPR-Cas9 technology introduce new possibilities for editing of disease-associated genetic risk factors in iPSCs, thereby offering a therapeutic solution. The review summarizes the contemporary understanding of CRISPR-based gene editing in iPSCs in relation to their usage for generating neurons resistant to neurodegeneration. Key targets, methodologies, challenges, and future directions are discussed, highlighting the potential of CRISPR in research and therapy for neurodegenerative diseases.

Introduction

Genetic research has become increasingly significant and, the breakthrough CRISPR technology, has opened up possibilities for the study and treatment of neurodegenerative diseases. Alzheimer's disease is a neurodegenerative disease responsible for gradual memory decline and cognitive dysfunction, and a significant public health problem. Genetic editing technologies, such as CRISPR-Cas9, can help progress the understanding of genetic disorders. CRISPR technology can edit disease- related genes, which allow developing a targeted approach to therapeutic treatment of neurodegenerative diseases such as Alzheimer's. The environmental and social impact of CRISPR on the progress of neurodegenerative disease research can be significant.

Neurodegenerative Diseases and Alzheimer's

Neurodegenerative diseases are becoming a major threat to global health and society. Alzheimer's disease is a neurodegenerative disorder that is characterized by progressive loss of memory, cognitive ability and behavioral disorders. Alzheimer's disease is one of the most common neurodegenerative diseases that accounts for a significant amount of the population in the world. Neurodegenerative disorders place a huge burden on society by increasing medical costs, loss of productivity, and decreased quality of life. Gaining insights into the genetic and environmental causes of neurodegenerative diseases may help in the development of targeted therapeutic strategies for the patients. One such breakthrough technology that would aid in

dissecting the disease model and therapeutic development is CRISPR technology (Sen and Thummer 1597-1623). Use of CRISPR based technologies in understanding and treating Alzheimer's disease and other neurodegenerative disorders can have a major impact on global disease burden.

Genetic Factors in Neurodegenerative Diseases

Genetic factors are actively involved in the development and progression of neurodegenerative disease, such as Alzheimer's, which emphasizes the significance of the genetic dimension of research in neurodegenerative diseases. The presence of genetic mutations in particular genes, such as APP, PSEN1 and PSEN2, seems to be linked to familial forms of Alzheimer disease that affect the production and aggregation of amyloid-β (Barman et al. 419-434). Furthermore, genetic mutations can harm the normal functioning of certain cellular processes that eventually cause neuronal dysfunction and degeneration. Therefore, the identification of such genetic factors can allow researchers to come up with targeted interventions that can address the affected molecular pathways during the pathogenesis of the disease (Konstantinidis et al. 450-461). The application of the CRISPR technology in such studies can strengthen the genetic dimension of research as genetic mutations are sometimes implicated in neurodegenerative diseases. Being able to perform accurate genetic mutations, CRISPR technology is proven to be a promising approach to study genetic aspects and potential treatment options of neurodegenerative diseases.

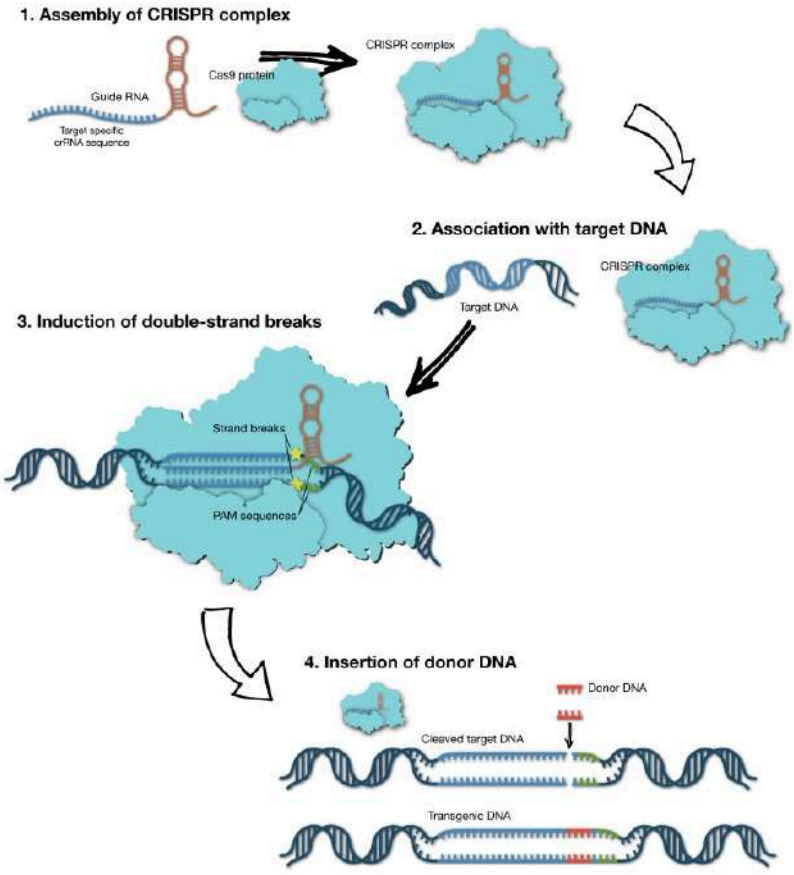
CRISPR Technology and iPSCs

Alzheimer's can benefit from CRISPR's introduction into iPSCs as they allow for gene editing of known pathogenic mutations present in the genome. iPSCs are stem cells made from somatic cells that have pluripotency, making them the ideal target cells for studying gene-editing's effect on cellular function in iPSCs (Sen and Thummer 1597-1623). The possibility of studying the effect of gene edits of known genes found in Alzheimer's disease such as mutations in APP, PSEN1/2, and APOE (Valadez-Barba et al. 332-339) may present promising results that propel discovery of targeted therapeutic interventions.

CRISPR-Cas9 Mechanism

This technology is a genome-editing tool based on a CRISPR-Cas9 system for precise and efficient gene targeting. This technique employs a guide RNA, which directs for a nuclease (named Cas9) the specific location on the genome where double-strand breaks will be performed. The cellular mechanisms will then repair these double-strand breaks and editing of the gene occurs (Barman et al. 419-434). This advances neurodegenerative diseases research

through the gene-editing of induced pluripotent stem cells to model genetic mutations that are believed to affect neuronal function (Valadez-Barba et al. 332-339). Resulting from the ability to generate iPS-derived neurons reflecting specific genetic modification meant for specific neurodegenerative diseases, such as Alzheimer's, offers a model for investigation of this disease's mechanisms and exploratory therapeutic approach (Sen and Thummer 1597-1623). Functionally characterizing variants that are found exploring their role in neuron degeneration and neurodegeneration influences, such as neuronal communication and synaptic development and stability, can offer further understanding of neurodegenerative diseases. This could lead to novel research-guided treatment approaches, including drug-discovery programs.



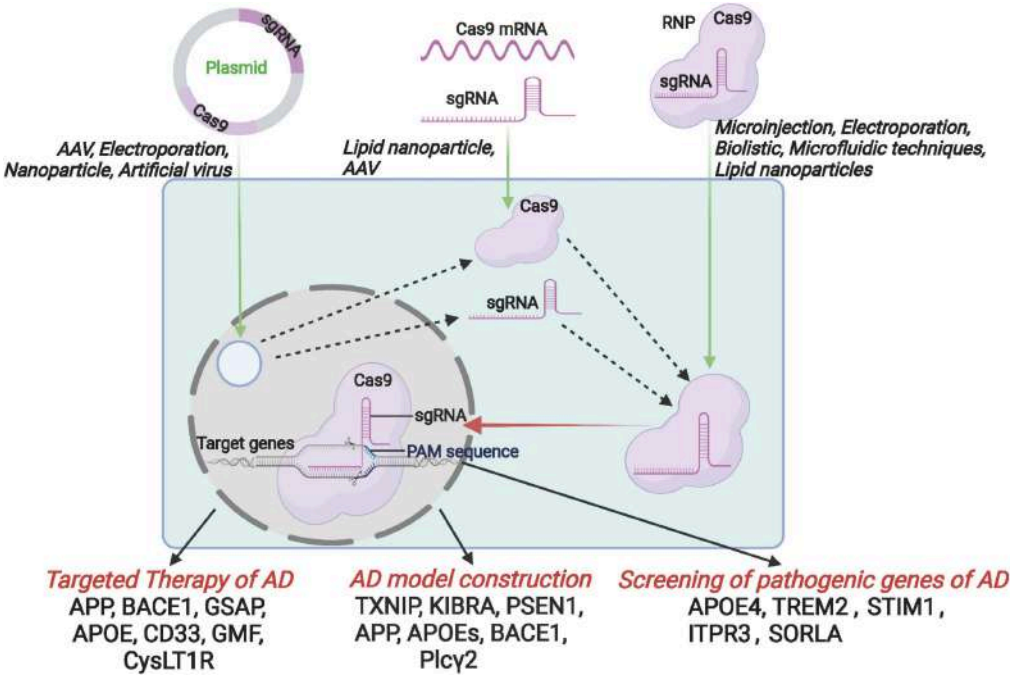
Key Genetic Targets for Alzheimer's

Alzheimer's disease is a genetically complex disorder and there are imperative target genes associated with the progression of this type of dementia such as amyloid precursor protein (APP), presenilin 1 and 2 (PSEN1/2) and apolipoprotein E (APOE). The mutation or alteration in APP is reported to cause amyloid precursor overproduction, leading to the aggregation of amyloid-beta peptide and consequent amyloid plaques (Konstantinidis et al. 450-461). The pathogenic presenilin (PSEN1/2) mutations interfere with the activity of

gamma-secretase complex enzymes and enhance the overproduction of amyloid-beta peptides, accelerating neurodegeneration. The polymorphism in the APOE gene is reported to be linked with the severity of early-onset and late-onset Alzheimer's (Barman et al. 419-434). The usage of CRISPR technology with respect to these genetic risk determinants can provide a therapeutic approach for Alzheimer's disease to modulate the risk through genetic correction of pathogenic mutations.

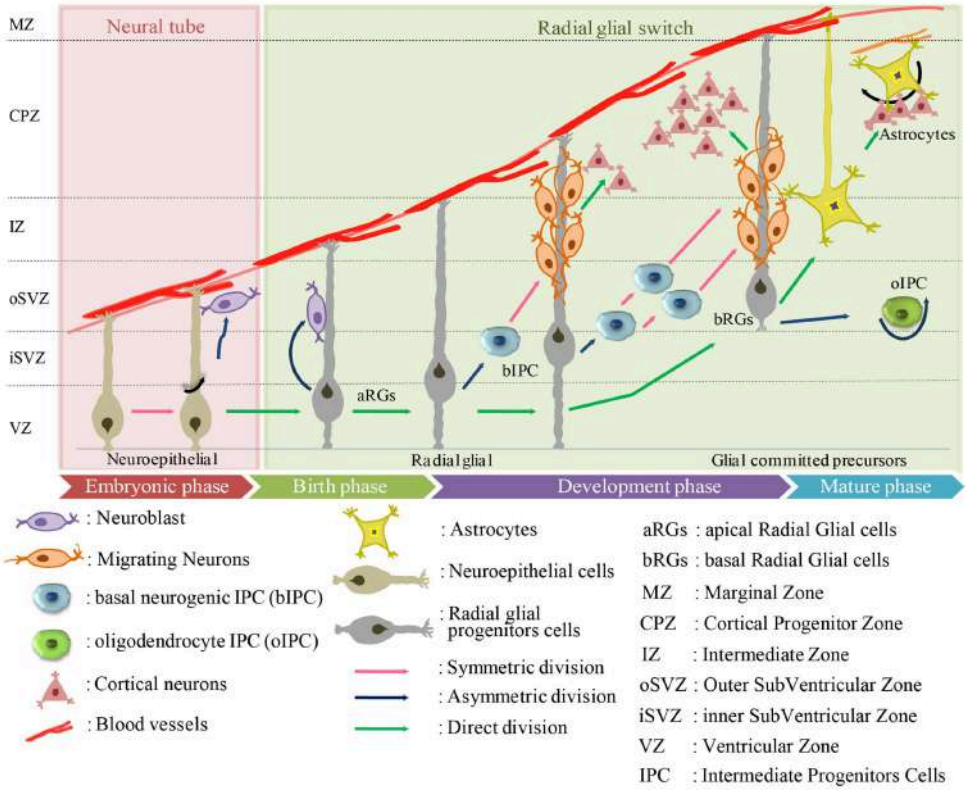
Methodologies in CRISPR Editing

To enable CRISPR editing to neuro-degenerative diseases, it is essential to design effective molecular delivery approaches to facilitate precise cutting. Various viral vectors, such as lentiviruses and adeno-associated viruses (AAVs), are generally used due to their excellent efficiency in transduction and integration of the CRISPR components into the target cells (Raikwar et al. 608-641). These vectors successfully transduced the CRISPR- Cas9 system into the iPSC, targeting the Alzheimer-related genes, such as APP and PSEN1 (Sen and Thummer 1597-1623). Host defense mechanisms against viral infections raise safety and efficacy concerns of using viral vectors. Therefore, non-viral delivery approaches such as electroporation and lipid nanoparticles are evaluated (Valadez-Barba et al. 332-339). Thus, optimizing molecular delivery mechanisms will further develop precise CRISPR editing and therapeutic advances in neurodegenerative diseases.



The CRISPR validation techniques play a crucial role to determine the efficiency of CRISPR editing on neurodegenerative diseases. One method involves the next-generation sequencing (NGS) technique to identify the off-target and on-target effects. It is most important to validate whether the CRISPR editing targets the specific regions like APP and PSEN1 genes (Konstantinidis et al. 450-461). The quantitative PCR (qPCR) technique is also used to validate the target genes and determine their expression activities. In addition to it, the fluorescence-activated cells sorting (FACS) technique was used to compare the edited cells with the unedited cells (Sen and Thummer 1597-1623). These techniques help to validate the efficient CRISPR editing target and regulate methods and their applications to process drug discovery targeting Alzheimer's and other neurodegenerative diseases.

Differentiation of edited iPSCs into neurons is crucial in studying Alzheimer's disease mechanisms and developing potential therapeutic strategies; thus, iPSCs are cultured under specialized conditions that facilitate their maturation into neurons that can establish synapses (Valadez-Barba et al. 332-339). Neurons created from edited iPSCs can provide insights into the roles of the target genes in neuron biology because the CRISPR technology can be used to characterize the effects of pathogenic genes on neuronal function and survival (Sen and Thummer 1597-1623). In turn, elucidating the genetic basis of cognitive decline in Alzheimer's disease will guide the development of targeted gene therapies that may help improve clinical outcomes in patients with cognitive impairment.



Challenges in CRISPR Editing

However, the utilization of CRISPR technology in neurodegenerative disease may pose challenges, such as the off-target effects accompanying genome editing. First, off-target effects refer to the undesired alteration to the genomic sequence, resulting in uncertainty in the phenotypic manifestation or could even aggravate the disease itself (Barman et al. 419-434). Off-target effects carry intrinsic risks in CRISPR therapies, hence necessitate the establishment and execution of validation procedures with high precision. In addition, researchers still need to examine the long-lasting effects of the editing processes, as it would either defeat the purpose of the treatment or lead to any adverse effects (Raikwar et al. 608-641). Therefore, CRISPR remains an exciting yet challenging option to explore in the field of neurodegenerative diseases.

In the context of CRISPR-based therapies for neurodegenerative diseases, gene stability is conceptualized as a major factor that affects the long-term safety and effectiveness of the treatment. Gene stability is defined as the ability of edited genes to remain unchanged for an extended period of time during treatment (Raikwar et al. 608-641). For neurodegenerative disorders such as Alzheimer's disease, the stability of target genes (e.g., APP and PSEN1) influences the effectiveness of gene-editing approaches because stable edits can prevent the development of pathological processes as well as neurons from impaired functions due to changes in their genome. Nonetheless, stability of edited genes has not yet been ensured during treatment since the processes involved in gene editing may have different outcomes, leading to a loss of stability (Sen and Thummer 1597-1623). Therefore, to develop gene-editing approaches that are ADN stable, more robust and highly efficient methods, as well as validation techniques capable of assessing the gene stability of genetically modified cells, should keep improving.

The ethical implications of the CRISPR technology debate on neurodegenerative disease research continue to be a contentious issue both in the scientific community and to the public at large. By manipulating genes, concerns regarding long-term effects, modification of human genetic material, and the ramifications involved were emerging a whole new ethical climate (D'SOUZA et al.). Genetic equity is one concept the CRISPR poses where only a certain population will benefit from the technology advancement; hence, the ethical concern requires consideration. The ecological impact where all elements would not be moving in balance can also spin from the negligence of the ethical implications that the CRISPR technology had posed (Barman et al. 419-434). Given the escalating possibility of CRISPR in germline editing, an effect is prominent in carrying on to the next generation; hence, the ethical debate would acquire consideration. Evaluating the implications of this research will heighten the possibilities of promising consideration to the issues entailed.

Future Directions in CRISPR Research

Future directions of CRISPR technology include its continual integration with other new technologies to further allow its advancement in neurodegenerative disease research. Improving organoids and in vitro models with CRISPR technology can allow for more understanding of mechanisms and pathways involved in disease onset and progression of neurodegenerative disorders at a cellular level and provide a greater understanding into developing therapeutics (Valadez-Barba et al. 332-339). In vivo testing and validation of CRISPR edits would be another important step forward in directions of research as this would allow for the determination of therapeutic potential and at what suitable levels and safety of genome editing in live subjects. Being able to demonstrate a disease model while continuously researching therapeutics directly in vivo would be an important addition to neurodegenerative discoveries in the lab as this can potentially allow findings in the lab to be translated for applications into clinical trials (Sen and Thummer 1597-1623). As these advances are made further towards the integration and establishment of the above-mentioned complementary methods and innovations with CRISPR technology, it can allow neurodegenerative targeted therapy to continue to develop towards discovery of a treatment method for neurodegenerative diseases fostered by the CRISPR technology revolution.

CRISPR technology is finding its way into the clinical setting, creating a new horizon for the treatment of neurodegenerative disorders, such as Alzheimer's. CRISPR has the potential to induce precise changes in DNA, allowing the correction of pathogenic mutations within key disease-causing genes (i.e., APP, PSEN1) involved in the pathogenesis of Alzheimer's disease (Barman et al. 419-434). These unique properties pave the way towards personalized therapeutic approaches that can halt or revert neurodegenerative disease progression. In addition, CRISPR technology combined with novel delivery methods improves its clinical implementation (Raikwar et al. 608-641). With steady efforts towards such clinical applications, CRISPR has the potential for real-life translation from the laboratory to the clinic in the near future, shedding light on innovative possibilities for neurodegenerative disease patients.

Conclusion

Undoubtedly, CRISPR technology is a milestone-based breakthrough in the development and research of neurodegenerative disorders and in particular Alzheimer disease. CRISPR allows precise genetic manipulation and addressing the major genetic targets APP, PSEN1, and APOE that are critical for the etiology of Alzheimer's disease. CRISPR-based methodologies including advanced gene delivery and gene editing technologies along with iPS cell developments have revolutionized the field and allowed easier and broad applications for this highly potential technology. Although there are limitations including but not limited to off-target effects and ethical concerns for the application of CRISPR, there are ongoing remarkable efforts

to provide and improve not only the efficacy but also the safety of CRISPR strategies. In the near future, we believe that CRISPR will dramatically change the potential and efficacy of neurodegenerative diseases research and its treatment approaches for further applications for drug and treatment development procedures.

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