Can Gene Editing Be The Key to Treating Alzheimers?

Saani Kulkarni

Commonly known as the most complex part of the human body, the brain contains millions of neurons, each with the ability to send and receive unique messages utilizing electrochemical signals. However, in individuals with Alzheimers and other neurodegenerative diseases, the typical function of neurons is impaired greatly. The lifespan of a neuron is dependent upon 3 primary factors: communication, metabolism, and its ability to repair itself. Without proper signaling, neurons are unable to access their target cells and gain access to necessary trophic factors. Moreover, neurons require adequate nutrients and chemicals to operate, gaining their energy through oxygen and glucose from the blood; the lack of these necessities will result in the death of the neuron. They must also be able to maintain a healthy state throughout their lifespan; while other human body cells may die quickly, neurons are observed to be able to live beyond 100 years of age in the human body.

While in a healthy human brain neurons may possess all these qualities, the opposite can be said for the brain in regards to Alzheimers; eventually, many neurons stop functioning and die out. Due to this, essential connections at the synapse may be broken down, impairing an individual from receiving necessary signals to carry out tasks. While researchers continue to investigate all causes behind Alzheimers, the beta-amyloid protein is commonly known as a major factor in the development of the disease. The cause of the cleavage of the amyloid precursor protein (APP), the presence of the beta-amyloid protein is originally beneficial, as it plays a significant part in neural growth and neural repair. However, a form of beta-amyloid known as beta-amyloid peptide 42 has been proven to be significantly toxic to the human body. Almost plaque-like, beta-amyloid 42 begins to gather in large quantities between neurons, damaging their functioning and breaking down their connections. Alongside b-amyloid 42, a protein known by tau that functions as a stabilizer for microtubules by attaching to them is often found in the human body as well. However, tau can separate and connects to other tau molecules instead, creating numerous knots and disruptions inside the neuron and ultimately affecting their ability to communicate. Researchers have found a positive correlation between an increased presence of tau and that of beta-amyloid, hinting at the fact that there are multiple mechanisms intertwined in the progression of Alzheimers.

Once neuronal networks begin to disappear, the memory of the individual, with parts of the brain such as the hippocampus and entorhinal cortex, is the first to be affected. As the disease spreads, the cerebral cortex of the brain is next, and ultimately individuals may be unable to carry out many basic behaviors independently. With such an extreme increase in severity, researchers have attempted to devise methods that will be able to treat Alzheimers and be able to reduce the symptoms gained as a result of it.

While originally researchers believed that a type of anti-beta amyloid drug would be a viable solution, it was soon understood that the state of the disease was already at a level of severity where simply treating it by affecting the amount of beta-amyloid protein in the brain would not necessarily result in a major change of quality of life nor health. In order to target the protein, the protein accumulation is removed as efficiently and quickly as possible in order to preserve the remaining parts of the brain.

Due to the precision of gene editing and the nature of the disease, teams worldwide have been examining the potential to treat Alzheimers using a variety of gene editing techniques. A significant positive towards gene editing would be the failure of other treatments to surpass the blood-brain barrier; however, gene therapy has the ability to surpass such obstructions. Despite this, many experiments regarding gene editing have not yet resulted in effects that would be considered noteworthy to the point of a fixed treatment. Researchers wish to continue experimentation by altering the type of viral vector, target for the therapeutic gene, and the route through which the vector should take. Viral vectors are considered one of the most effective ways for gene delivery to any target cells. When done ex-vivo, the target cells are drawn out from the organism's body, then introduced to the therapeutic gene, and planted back into the organism; in-vivo requires the process to be done within the organism itself. This can result in either gene silencing or overexpression, all of which is dependent on the target cell itself. Considering the wide range in virus shapes and structures, many types of viral vectors are utilized in order to attain precision and efficacy.

In treatments for Alzheimers, researchers have traditionally experimented with adeno-associated virus (AAV) vectors, due to their non-toxic and non-pathogenic nature. Taking into account the possibility of mutagenesis in the body, it is essential that the viral vector must possess these two qualities. Researchers identified around 100 AV variants with a range of 13 serotypes; although multiple have been deemed adequate for experimentation, AAV2 is used most frequently due to its high level of safety and sustained expression of the therapeutic gene within the neuron. Along with the identified AAVs, a genetically engineered AAV capsid (AAV-PHP.B) has been utilized in studies and expresses the ability to deliver a greater capacity of AAV

genomes to the central nervous system (CNS) and convert more than 50% of astrocytes and neurons (Chen, W., Hu, Y., & Ju, D, 2020).

While the selection of a vector is critical, researchers globally have experimented with AAV vectors and various targets in the body to treat Alzheimers; without choosing the proper target, the therapeutic gene may simply diminish symptoms slightly while possessing no contribution to the treatment and root cause of Alzheimers. Endoplasmic reticulum (ER) stress and the unfolded protein response (UPR) has been extensively studied, as the majority of neurodegenerative diseases are correlated with an accumulation of misfolded proteins, which contributes to stress and eventual breakdown of the ER. Additionally, beta-amyloid oligomers are a source of destabilization to the calcium of the ER and its homeostasis, resulting in the death of neurons. By potentially being able to reduce the amount of stress affecting the ER, researchers have begun to experiment with a method to target the unfolded protein response (UPR) signaling in the hopes that the signaling pathway may be amplified for improvement in protein folding through gene editing. Notably, a positive correlation between the overregulation of the UPR pathway and glioblastoma invasion has been discovered, therefore being a safety concern when experimenting with this pathway. Another potential target for researchers has been the mTOR pathway, which deals with the regulation of mammalian metabolism and has also been known to play a role in neurodegeneration; when being abnormally regulated, protein accumulation is unable to be removed. In 2017, Chen et. al previously demonstrated that the delivery of mTOR positive regulators to the retina via the AAV vector resulted in a decrease of ganglion cell death and CNS axon regeneration; a variety of other studies have also reported positive effects on neuron regeneration in mouse models, highlighting the potential for the mTOR pathway to be an ideal target. Researchers have also devised methods for autophagy to be used through gene editing; in 2012, Gorbatyuk et. al demonstrated that the overexpression of a kinase through AAV2 resulted in the autophagy of dysfunctional mitochondria. In doing so, mitochondrial function that was lost as a result of b-amyloid oligomers was regained (Chen et.al, 2020). Similarly, microglia and astrocyte function has been brought to light as potential targets, considering the great importance of microglia within the neuroimmune system, and the damaging of their functions resulting in the neuron destruction witnessed in Alzheimers. The re-regulation of microglia function has been considered a potential treatment; multiple studies have reviewed the ability of the overexpression of the receptor TREM2 on myeloids to assist beta amyloid destruction and microglial movement. Through this process, the plaque formation and eventual disruption of neurons caused by b-amyloids is reduced, and spatial memory abilities are increased.

In recent years, studies with clustered, regularly interspaced short palindromic repeats and the CRISPR-associated protein 9 (CRISPR-Cas9) have increased significantly, and yielded optimistic results. Targeted to specific genes and highly precise, gene-editing techniques and therapies have improved greatly as a result of this technology. Comprised of a singular enzyme (Cas9) and a single guide RNA (sgRNA), the target DNA sequence is registered and recognized by the sgRNA, and cleaved by the Cas9 endonuclease, resulting in either a replacing of the mutated sequence or the insertion and deletion of sequences for inactivation. Considering that genetic mutations only comprise around 1% of familial Alzheimer's cases, CRISPR's role in helping patients with AD may be limited to changing symptom expression by manipulating b-amyloid metabolism (Bhardwaj, S., Kesari, K. K., Rachamalla, M., Mani, S, et. al, 2021).

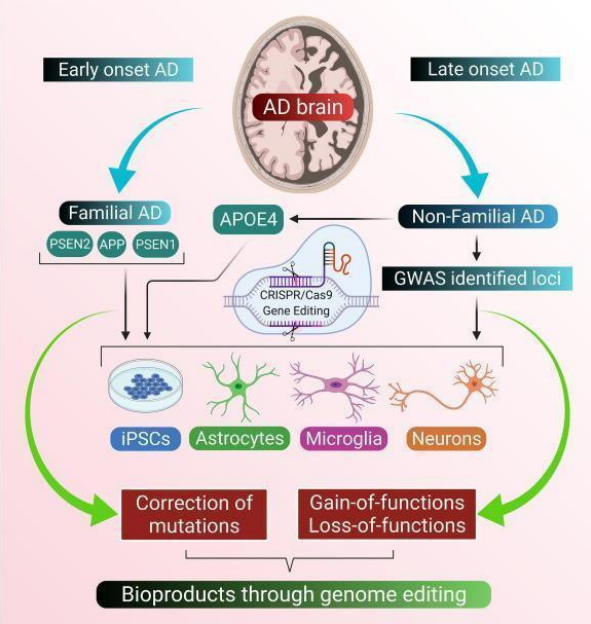

Figure 1. Proposed CRISPR Cas-9 gene editing approaches to types of Alzheimer's Disease. Adapted from "CRISPR-Cas9 gene editing : New hope for Alzheimer's disease therapeutics," by Bhardwaj, S. et al, 2021, Journal of Advanced Research (https://doi.org/10.1016/j.jare.2021.07.001). CC BY 4.0.

While the variety of findings in regards to gene editing and the treatment of Alzheimer's disease are promising, much work is needed to ensure the safety of procedures on the human brain and their efficacy. Host responses to viral vectors may prove to be dangerous, as is the concern of patients with high-functioning immune systems creating antibodies with the purpose of neutralizing the inserted vectors. Additionally, the ethics and viability of a supposed treatment must be taken into consideration; factors such as cost, effectiveness, and availability all must be regarded. Improved knowledge of the mechanisms behind Alzheimer's and their connection to various parts of the human body will undoubtedly improve the potential prospects for treatment. In combination with the continued efforts to find suitable vectors and outline target genes, many scientists and doctors are hopeful that gene-editing techniques may be a potential means to find a conclusive treatment to a disease that has affected millions of people worldwide.

References

1. Bhardwaj, S., Kesari, K. K., Rachamalla, M., Mani, S., Ashraf, G. M., Jha, S. K., Kumar, P., Ambasta, R. K., Dureja, H., Devkota, H. P., Gupta, G., Chellappan, D. K., Singh, S. K., Dua, K., Ruokolainen, J., Kamal, M. A., Ojha, S., & Jha, N. K. (2021). CRISPR/Cas9 gene editing: New hope for alzheimer's disease therapeutics. Journal of Advanced Research. https://doi.org/10.1016/j.jare.2021.07.001
2. Chen, W., Hu, Y., & Ju, D. (2020). Gene therapy for neurodegenerative disorders: Advances, insights and prospects. Acta Pharmaceutica Sinica B, 10(8), 1347–1359. https://doi.org/10.1016/j.apsb.2020.01.015
3. Gaj, T., Sirk, S. J., Shui, S.-lan, & Liu, J. (2016). Genome-editing technologies: Principles and applications. Cold Spring Harbor Perspectives in Biology, 8(12). https://doi.org/10.1101/cshperspect.a023754
4. Warnock, J. N., Daigre, C., & Al-Rubeai, M. (2011). Introduction to viral vectors. Methods in Molecular Biology, 1–25. https://doi.org/10.1007/978-1-61779-095-9_1
5. U.S. Department of Health and Human Services. (n.d.). What happens to the brain in alzheimer's disease? National Institute on Aging. Retrieved April 25, 2022, from https://www.nia.nih.gov/health/what-happens-brain-alzheimers-disease