Proceedings of the 3rd International Conference on Modern Medicine and Global Health
DOI: 10.54254/2753-8818/2025.24201

Alzheimer's Disease and Adult Hippocampal Neurogenesis: Tracking Aβ and Immature Neuron Markers

Zhengyang Xu

Department of Chemistry, University of Washington, Seattle, USA
zx062504@uw.edu

Abstract: From the work of Moreno-Jimenez et al., it is known that the generation of new neurons dramatically declines in patients with Alzheimer's disease compared with mentally healthy individuals, marked by DCX^+ cells (immature neurons) failing to produce other structures characteristic of mature neurons. The same researchers also observed that such a phenomenon worsened as Alzheimer's disease progressed but did not offer any explanation. This research proposal introduces an experiment that could reveal the mechanism behind the decrease of adult hippocampal neurogenesis in patients with Alzheimer's disease. Focusing on Aβ1-42, a hallmark of Alzheimer's disease, this paper examines its role in reducing the number of new neurons produced. Techniques such as immunofluorescence, chromatin immunoprecipitation sequencing, and RNA sequencing are employed.

Keywords: Alzheimer's disease, Aβ, Adult hippocampal neurogenesis, Neuron markers.

1. Introduction

Alzheimer's disease (AD) is a common neurodegenerative disorder that could result in profound memory loss and cognitive decline, among other issues. To have a better picture of what might have contributed to these symptoms, researchers examined the brain tissues of patients who died of AD and identified senile plaques, which consist of Aβ and neurofibrillary tangles (NFTs) [1]. In addition, the researchers noticed the impairment of adult hippocampal neurogenesis (AHN) in AD patients, marked by the decline of neural stem cells that exhibit structures unique to mature neurons, and such impairment exacerbates as AD becomes more severe [2]. Researchers found that the amount of neural stem cells does not change and that there is no significant difference in the post-mortem brain samples of AD patients and healthy individuals [3], so the symptoms seen in AD patients are likely not due to population shifts in neural stem cells. A different mechanism may be at play. Since developing neural stem cells in AD patients lack mature neuron-specific markers, it is likely that genes that encode these structures are downregulated by a structure, which interrupts the maturation of these cells and AHN. Able to enter the nucleus [4] and absent in the brains of healthy individuals [2], Aβ1-42, a common Aβ species seen in AD [5], stands out as a candidate for the above-mentioned structure that interacts with genes and slows down AHN. Given this, an experiment is designed that investigates the interplay between Aβ and DNA and its results on AHN. The hypothesis is that higher concentrations of extracellular Aβ1-42 could increase the amount of nuclear Aβ1-42, bind to more sites on DNA, and inhibit more genes that encode mature neuron markers.

DOI: 10.54254/2753-8818/2025.24201

2. Experimental design

The experiment consists of three parts, with the first investigating the amount of Aβ1-42 in the nucleus, the second looking for the interactions between Aβ1-42 and DNA, and the third examining the expression levels of mature neuron markers. The effects of Aβ1-42 on the focus of each part will be investigated.

2.1. Part 1

In the first part of the experiment, four groups of human neural stem cells (Creative Bioarray, Cat. No.: PCELL-0297) will be set up, each treated with a different Aβ1-42 concentration, as Table 1 describes. Group A simulates neural stem cells in healthy humans, so no Aβ1-42 is added, and the cells can undergo undisturbed maturation. Groups B, C, and D demonstrate AHN in separate phases of AD. To approximate the brain of AD patients where Aβ is detected at low nanomolar concentrations [6], 250, 750, and 1000 nM of Aβ1-42 will be assigned to Groups B, C, and D, respectively. A higher Aβ1-42 concentration is correlated with more severe Alzheimer's disease [1, 2]. In the experiment, Aβ1-42 is tagged with a fluorescent dye. Cells will be looked at under a fluorescence microscope for the intensity of fluorescent signals.

Table 1: Various amounts of Aβ1-42 are given to each group

Groups	Aβ1-42 Concentrations (nM)
A	0
B	250
C	750
D	1000

2.2. Part 2

To substantiate the interactions between DNA and Aβ1-42, the second part of the experiment is performed using chromatin immunoprecipitation sequencing (ChIP-Seq). The four groups will receive the same amount of Aβ1-42 as in Part 1, but Aβ1-42 is no longer fluorescent.

After being added to cells, Aβ1-42 will undergo possible binding to DNA. Afterward, cells in each group will be treated following the protocol for ChIP-seq [7].

DNA will then be fragmented, and antibodies for Aβ1-42 will be added to immunoprecipitate DNA fragments bound by Aβ1-42, which will then be sequenced and examined for Aβ1-42 binding sites.

2.3. Part 3

Following the identification of all potential binding sites for Aβ1-42 on DNA, the third part of the experiment looks for the result of Aβ1-42 binding of various amounts on the expression of products encoded by genes near those sites. For simplicity, this experiment only reports the expression of four products (mature neuron markers): CB, βIII-tubulin, Prox1, and PSA-NCAM [1].

Repeating the procedure set up earlier, the four groups will receive the same concentrations of Aβ1-42 as before. To study the influence of Aβ1-42 on gene expression, RNA sequencing (RNA-seq) will be utilized to count the levels of mRNA that encode mature neuron-specific structures. To prepare the cells for sequencing, they will be further treated according to RNA-seq protocol [8].

DOI: 10.54254/2753-8818/2025.24201

3. Expected results

3.1. Part 1

Various levels of fluorescent intensities among the groups could be observed under the fluorescent microscope, which suggests various amounts of nuclear Aβ1-42.

Group A is treated with no Aβ1-42, so there could not be any of it in the nucleus. Therefore, neural stem cells in Group A could display no fluorescent signals.

Group B contains a trace amount of Aβ1-42, and there could be some of it in the nucleus, as characterized by weak fluorescent signals around the organelle.

Group C has more Aβ1-42 than the previous two groups, so even more Aβ1-42 could enter the nucleus and give off stronger fluorescent signals.

Group D is given the highest amount of Aβ1-42, so the group could surpass others in the amount of nuclear Aβ1-42, as indicated by the strongest fluorescent signals.

Table 2: Possible findings of this part of the experiment

Groups	Fluorescence Intensity
A	None
B	Weak
C	Stronger
D	Strongest

To visualize the possible results, Figure 1 is taken from another work [9], showing varying intensities of fluorescence that correspond to different amounts of protein expressed.

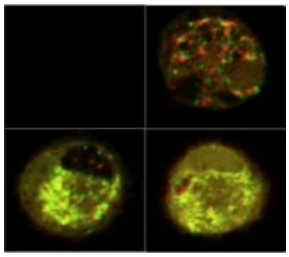

Figure 1: From top to bottom, left to right: possible fluorescence intensity seen in Groups A, B, C, and D. Reproduced from Akyuva et al (2020)

3.2. Part 2

ChIP-seq could identify different amounts of Aβ1-42 binding sites in each group.

From the expected results of Part 1, cells in Group A lack nuclear Aβ1-42. Therefore, no DNA fragments bound by Aβ1-42 could be found and the binding sites for Aβ1-42 could not be identified.

In Group B, there is likely some Aβ1-42 binding to DNA because of the low concentration of extracellular Aβ1-42. Given this, there could be a small number of binding sites sequenced.

In Group C, there could be more sites on DNA bound by Aβ1-42, as its extracellular concentration is higher. Accordingly, more binding sites could be identified by sequencing.

In Group D, cells are exposed to the highest amount of external Aβ1-42, which could lead to more DNA sites bound by Aβ1-42 than all other groups. This could be reflected by the greatest number of binding sites sequenced.

DOI: 10.54254/2753-8818/2025.24201

Table 3: Potential discoveries of this part of the experiment

Groups	Number of Binding Sites Sequenced
A	None
B	Some
C	More
D	Most

3.3. Part 3

RNA-seq could reveal varying mature neuron marker expression levels in the four groups.

Based on the expected results of Part 2, Aβ1-42 does not bind to the DNA of cells in Group A, so they could exhibit undisturbed maturation due to the absence of Aβ1-42, and all markers investigated in the experiment could be expressed at high levels, as shown by high levels of marker mRNA detected.

In Group B, the maturation of neural stem cells could be hindered because of the slight amount of Aβ1-42 binding present. All markers in this group could still be expressed, but less abundantly than in Group A. Hence, RNA-seq could identify lower amounts of marker mRNA.

In Group C, neural stem cells could experience further retardation in development since more sites on their DNA are bound by Aβ1-42, so there are likely fewer markers produced, leading to even fewer amounts of marker mRNA discovered by RNA-seq.

In Group D, neural stem cells might encounter the most profound degree of maturation impairment because the group could have the highest number of DNA sites bound by Aβ1-42. Markers could be expressed at a level lower than all other groups. RNA-seq could find no marker mRNA at all.

Table 4: Likely outcomes of this part of the experiment

Groups	Mature Neuron Markers' Expression Levels	Amounts of mRNA of Interest
A	Strongest	Highest
B	Weak	Low
C	Weaker	Lower
D	Weakest	Lowest

4. Discussion and summary

The strength of this design is its careful use of technologies in examining and quantifying biological structures that are at the center of AD. By using ChIp-seq in Part 2, the specific locations where Aβ1-42 binds to DNA could be learned, validating the claim that Aβ1-42 interacts with DNA. This compensates for the insufficiency of results from Part 1 to establish this feature. Furthermore, the method of transcriptional analysis described herein provides abundant information. RNA-seq identifies all RNA molecules present in the cell and reports their amount, allowing comparison between the effect of multiple Aβ1-42 concentrations on the expression of mature neuron markers among the four groups. This technique also provides insight into the expression of previously overlooked RNA molecules, offering potential targets for future research.

However, the experiment also comes with some limitations. The pathological roles of NFTs [10] are not considered in this experiment. NFTs could be intensifying the toxicity of Aβ1-42 and vice versa, so the experiment might provide an underestimate of the gene expression inhibition that takes place in AD patients. Similarly, it has been reported that synthetic Aβ leads to less damage than Aβ derived from brain samples [4]. This may also lead to an insufficient representation of pathological factors in the experiment.

DOI: 10.54254/2753-8818/2025.24201

Overall, the experiment outlined in this article is creative in explaining why AHN is impaired in AD patients. It could be established that Aβ1-42 plays an integral role in AHN disruption by entering the nucleus and binding to several sites on DNA, which inhibits the expression of relevant genes. A higher concentration of Aβ1-42 outside the cell could result in more of it in the nucleus, binding to more sites on DNA and downregulating more genes that encode mature neuron structures. The experiment could attribute AHN impairment in AD patients to the genetic interference of Aβ1-42.

References

[1] *Breijyeh, Z., & Karaman, R. (2020). Comprehensive review on Alzheimer's disease: Causes and treatment. Molecules, 25(24), 5789. https://doi.org/10.3390/molecules25245789*

[2] *Moreno-Jiménez, E. P., Flor-García, M., Terreros-Roncal, J., Rábano, A., Cafini, F., Pallas-Bazarra, N., Ávila, J., & Llorens-Martín, M. (2019). Adult hippocampal neurogenesis is abundant in neurologically healthy subjects an d drops sharply in patients with Alzheimer's disease. Nature Medicine, 25(4), 554–560. https://doi.org/10.1038/s4 1591-019-0375-9*

[3] *Cao, Y., Liu, P., Bian, H., Jin, S., Liu, J., Yu, N., Cui, H., Sun, F., Qian, X., Qiu, W., & Ma, C. (2023). Reduced neu rogenesis in human hippocampus with Alzheimer's disease. Brain Pathology, 34(3). https://doi.org/10.1111/bpa.1 3225*

[4] *Barucker, C., Harmeier, A., Weiske, J., Fauler, B., Albring, K. F., Prokop, S., Hildebrand, P., Lurz, R., Heppner, F. L., Huber, O., & Multhaup, G. (2014). Nuclear translocation uncovers the amyloid peptide AB42 as a regulator of gene transcription*. Journal of Biological Chemistry, 289(29), 20182–20191. https://doi.org/10.1074/jbc.m11 4.564690*

[5] *Hampel, H., Hardy, J., Blennow, K., Chen, C., Perry, G., Kim, S. H., Villemagne, V. L., Aisen, P., Vendruscolo, M., Iwatsubo, T., Masters, C. L., Cho, M., Lannfelt, L., Cummings, J. L., & Vergallo, A. (2021). The amyloid-B pathway in Alzheimer's disease. Molecular Psychiatry, 26(10), 5481–5503. https://doi.org/10.1038/s41380-021-01249-0*

[6] *Hayden, E. Y., & Teplow, D. B. (2013). Amyloid β-protein oligomers and Alzheimer's disease. Alzheimer S Research & Therapy, 5(6), 60. https://doi.org/10.1186/alzrt226*

[7] *Texari, L., Spann, N. J., Troutman, T. D., Sakai, M., Seidman, J. S., & Heinz, S. (2021). An optimized protocol for rapid, sensitive and robust on-bead ChIP-seq from primary cells. STAR Protocols, 2(1), 100358. https://doi.org/1 0.1016/j.xpro.2021.100358*

[8] *Koch, C. M., Chiu, S. F., Akbarpour, M., Bharat, A., Ridge, K. M., Bartom, E. T., & Winter, D. R. (2018). A Beginner's Guide to analysis of RNA sequencing data. American Journal of Respiratory Cell and Molecular Biology, 59(2), 145–157. https://doi.org/10.1165/rcmb.2017-0430tr*

[9] *Akyuva, Y., Nazıroğlu, M., & Yıldızhan, K. (2020). Selenium prevents interferon-gamma induced activation of TRPM2 channel and inhibits inflammation, mitochondrial oxidative stress, and apoptosis in microglia. Metabolic Brain Disease, 36(2), 285–298. https://doi.org/10.1007/s11011-020-00624-0*

[10] *Monteiro, A. R., Barbosa, D. J., Remião, F., & Silva, R. (2023). Alzheimer's disease: Insights and new prospects in disease pathophysiology, biomarkers and disease-modifying drugs. Biochemical Pharmacology, 211, 115522. https://doi.org/10.1016/j.bcp.2023.115522*