January 2024

# Determination of Antibody Affinity for the Alzheimer's Amyloid-β Protein

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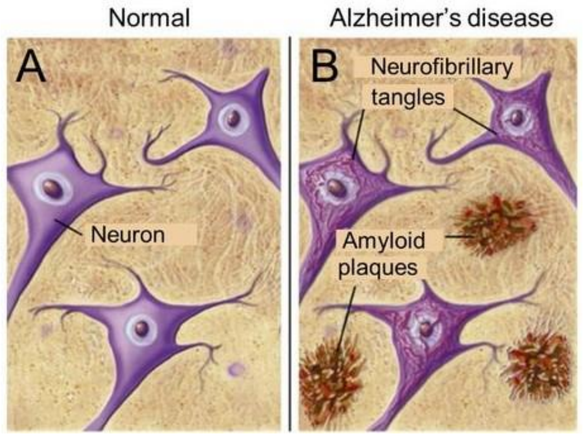


**Figure 1** shows a comparison of a healthy brain and neurons that have been affected by Alzheimer's Disease. Image from (*1*).

## Introduction

One of the pathological hallmarks of Alzheimer's Disease (AD) is the accumulation of Amyloid-β (Aβ) in the brain as senile plaques. AD is one of the most common types of dementia resulting in loss of memory and motor function, ultimately leading to death. Aβ monomers are naturally occurring and nonpathogenic, while aggregated protofibrils are more neurotoxic and neuroinflammatory. This project is investigating two antibodies that bind to different regions of Aβ. Ab5 binds to the N-terminus of the peptide, while Ab2.1.3 binds to the C-terminus (*5*). The objective is to determine which antibody has a higher affinity for Aβ and whether aggregation status impacts antibody binding.

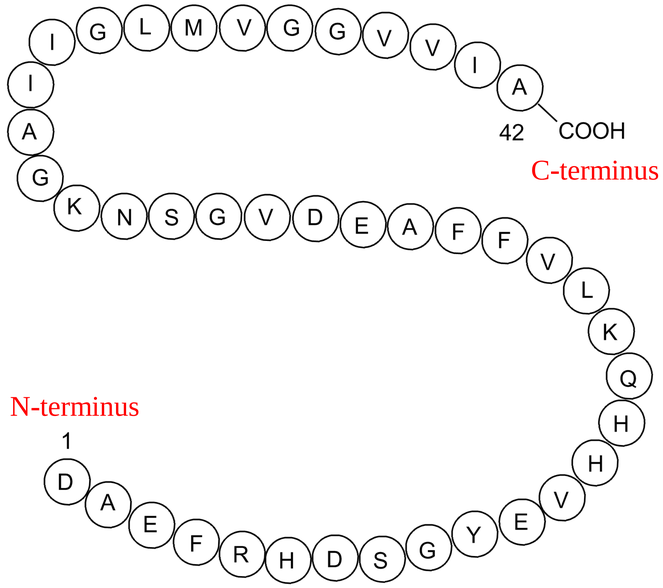


**Figure 2** shows the amino acid sequence of Amyloid-β-42. The N-terminus is marked by the number 1, while the C-terminus is represented as a COOH. Image from (*2*).

## Methodology

**Size Exclusion Chromatography (SEC).** Aβ protofibrils and monomers were purified in the Nichol's Lab for these experiments.

**Indirect Enzyme Linked Immunosorbent Assay (ELISA).** Amyloid protofibrils or monomers were plated, and then antibody 5 or 2.1.3 were used as primary antibodies. These antibodies were raised in inoculated rabbits. Then, anti-rabbit HRP is used as a secondary antibody. HRP undergoes a reaction to give a signal that is read using a spectrophotometer.

## Research Findings

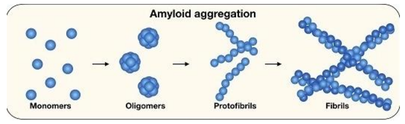


Figure 3 shows the different aggregation intermediates. Image from Source 3.

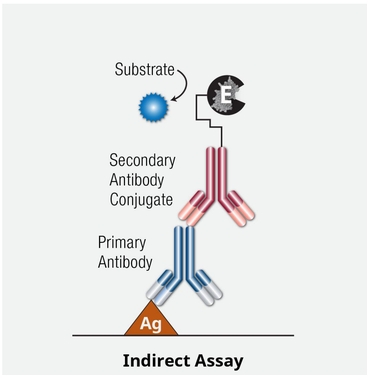


**Figure 4** shows an overview of the Indirect ELISA procedure. Image from Source 4.

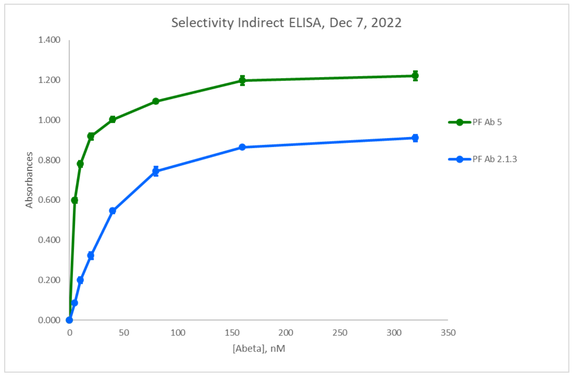


**Figure 5** shows that Ab5 has a higher inherent affinity for protofibrils and monomers than Ab2.1.3.

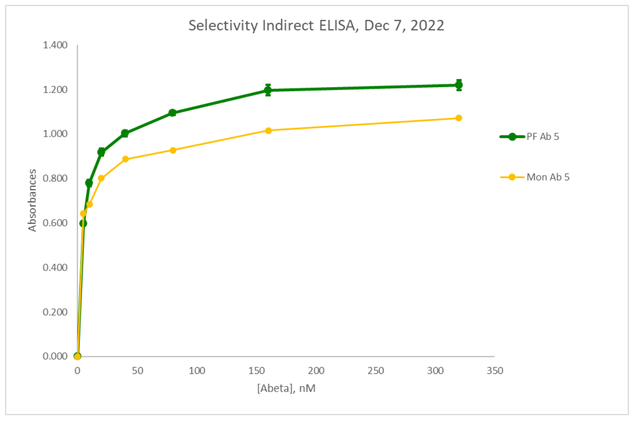


**Figure 6** shows that Ab5 has a similar affinity for the protofibrils and monomers. This result indicates that the N-terminus is exposed on both species.

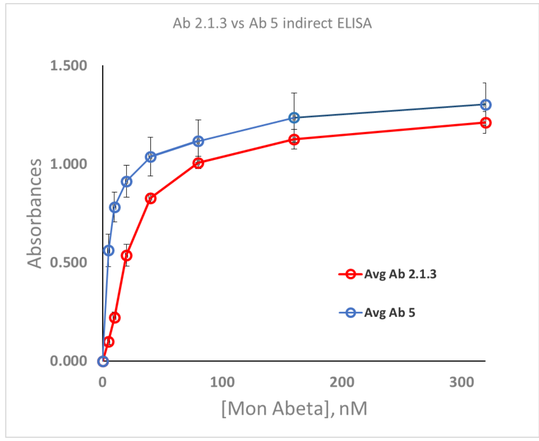


**Figure 7** shows that Ab 5 has a higher affinity towards monomers than Ab 2.1.3.

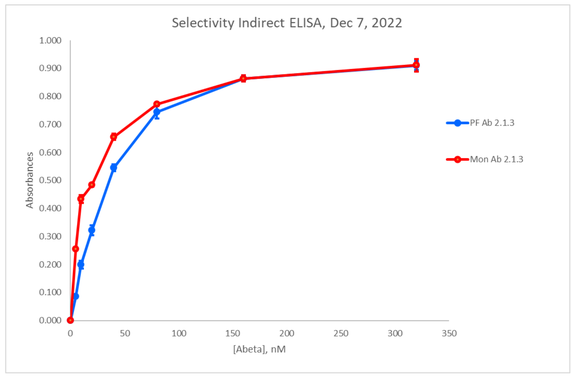


**Figure 8** indicates that the C-terminus is exposed on both protofibrils and monomers.

## Conclusions and Future Directions

-We see that both the C-terminus and N-terminus of peptide are available for antibody binding, but Ab5 antibody has a higher affinity for both protofibrils and monomers

-These results indicate that both antibodies are able to bind the protofibrils and monomers, with Ab5 showing a higher affinity

## References

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