Evaluating Age Dependent Metabolic Interactions with Amyloid beta in *Caenorhabditis elegans* as a Novel Approach for Modeling Multi-Factor Based Neurodegeneration in Alzheimer's Disease Year II

Riya Reddy and Dr. Kirstin Cutshaw

Eastern Florida State College

Background

ALZHEIMER'S DISEASE

❖Alzheimer's disease is a progressive neurodegenerative disorder and the leading cause of dementia

➢Accounts for 60-80% of cases [1]

➢Symptoms include memory loss, difficulty with cognitive function, Impaired judgement, behavior, & personality changes

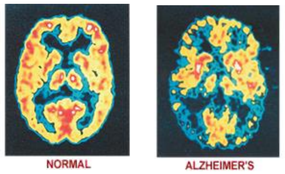


Figure 1: PET scans show which parts of the brain are active. When you compare a healthy person's scan (Left image) with the scan of someone who has Alzheimer's (Right image), the Alzheimer's brain has more dark patches. Those patches are areas where the brain isn't working as much because of the disease. *Source: https://www.alzinfo.org/what-happens-to-the-brain-in-alzheimers-disease/*

❖~50 million people worldwide were living with Alzheimer's disease in 2020; cases are estimated to increase significantly to 152 million by 2050 [2].

➢ Florida has one of the highest numbers of Alzheimer's disease cases in the U.S [3]

❖Research for Alzheimer's is ongoing; no cure or definite cause has been discovered

❖ Two categories of risk factors: genetics and lifestyle

➢Mutations in genes APP, PSEN1, and PSEN2 increase Amyloid β protein production

➢Lifestyle factors include aging, smoking, diet, & stress

AMYLOID BETA (aβ)

❖ Dominating theory: Amyloid cascade hypothesis developed in 1980s

➢Alzheimer's is triggered by misfolding aβ peptide

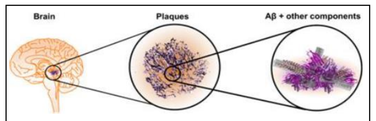


Figure 2: A defining feature of the Alzheimer's brain is the presence of senile plaques outside neurons. These plaques are protein deposits made mainly of amyloid-beta (Aβ), along with a variety of other components. Source: *https://doi.org/10.1186/s13024-021-00465-0*

❖Misfolded Aβ plaques disrupt neuronal communication

❖FDA approved treatments focus on anti-amyloid approaches

❖Genetic mutations & lifestyle factors lead to increased levels of aβ

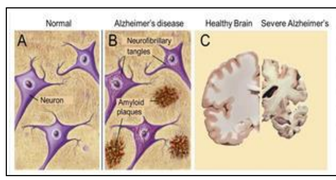


Figure 3: Neuropathology of Alzheimer's disease (AD). (A) Healthy brain tissue, with no plaques or tangles. (B) AD brain tissue showing the two main pathological hallmarks: extracellular amyloid plaques and intracellular neurofibrillary tangles. (C) Brain slices showing widespread neuronal loss and tissue shrinkage (atrophy) in late-stage AD. *Image sources: (A, B) BrightFocus Foundation, "Alzheimer's Disease: Amyloid Plaques and Neurofibrillary Tangles." (C) Beddows, C. (2016). "The neural basis of Alzheimer's disease," Elsevier SciTech Connect, via Wikimedia Commons.*

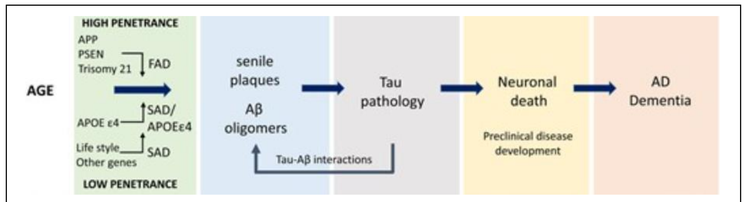


Figure 4: The amyloid cascade hypothesis and its revision. The classic hypothesis describes a fixed sequence: amyloid buildup, then tau pathology, then neurodegeneration and cognitive decline. Frisoni et al. (2022) proposed a *probabilistic* model in its place. In that model, AD comes in three variants: autosomal dominant, APOE ε4-related sporadic, and APOE ε4-unrelated sporadic. Moving through those three, penetrance goes down, the amyloid cascade matters less, and environmental and lower-risk genetic factors matter more. Source: https://doi.org/10.1093/brain/awad159

Research Objective

To investigate the individual and combined effects of metabolic stress, aging, and amyloid beta expression using *Caenorhabditis elegans* as a model organism

❖ Recent studies and failed clinical trials have raised doubts about the amyloid cascade hypothesis

❖ Multi-year project aimed to investigate the contributing factors of Alzheimer's to better understand its complex progression

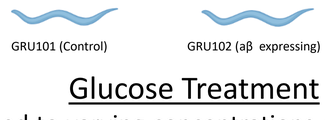
❖ Current focus on investigating the interaction between multiple factors in disease progression: metabolic stress, amyloid beta, and aging

➢Expands Year I findings by turning towards multi-factor hypothesis

Methodology

Strains

C. elegans strains GRU101 and GRU102 used to study the interaction between metabolic stress, aging, and its effect on phenotypes. Worms will be grown on Nematode Growth Medium agar (NGM) supplemented with glucose on petri dishes seeded with *Escherichia coli* OP50



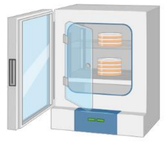
Glucose Treatment

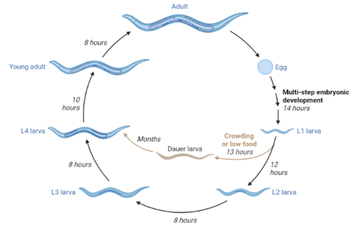
Worms will be exposed to varying concentrations of glucose (0 mM, 20 mM, 60 mM, 100 mM) to induce different levels of metabolic stress



Aging Intervals

Worms will be studied at different age intervals (Days 1, 5, & 10 after incubation) in order examine age-dependent changes in behavioral and morphological phenotypes





Phenotype Observations

Behavioral (locomotion, movement speed, thrashing) and morphological (body length & size) changes will be observed under microscope to analyze the individual vs combined effects of metabolic stress, aging, and aβ accumulation



Images created by R.Reddy using BioRender

Hypotheses

❖GRU102 (aβ strain) will display more pronounced behavioral and morphological changes than GRU101 (wildtype strain)

❖Increasing glucose concentrations will increase metabolic stress and potentially worsen phenotypes

➢ GRU102 worms treated with high glucose concentrations will display greater changes compared to GRU101 worms under the same conditions

❖Older worms will exhibit greater age-dependent changes, which will increase the severity of metabolic stress and aβ expression

➢Combination of high glucose concentrations, increased age, and aβ expression will produce the most significant phenotypic changes

Limitations

❖Limitations include a different nervous system and structure from humans

❖Incorporating glucose into the NGM agar may influence bacterial growth and nutrient availability, potentially affecting experimental outcomes.

➢ Possible that *E. coli* OP50 will consume the glucose in the agar before the *C. elegans* strains, reducing the concentration

❖ Data obtained from the experiment may model changes in the gut-brain axis and gut microbiome of the worms after consuming the *E. coli* and glucose

Future Directions

❖Establish relationship between metabolic stress, oxidative stress, aging, & amyloid beta

❖ Test a wider range of glucose concentrations to identify threshold where metabolic stress begins to influence behavior or morphology of *C. elegans*

❖ Continue studying the other known factors of Alzheimer's and interactions with each other (Tau tangles, neuroinflammation, genetic mutations, etc.)

➢Continue looking towards a multi factor approach

❖Integrate CRISPR-based gene editing to investigate correlation between elevated levels of aβ and metabolic stress at the genetic level

❖Study mutations in APP, PSEN1 & 2, APOE ε4 and their relationship to other metabolic or aging factors

❖Compare findings from Year I research to understand the relationship between cellular stress responses and Alzheimer's disease

Clinical Application

❖Provide insight between metabolic stress, aging, & aβ changes

❖Model can be extended for studying amyloid-targeting antibodies and other therapeutic approaches

➢Ongoing clinical trials / research for developing more efficient treatments

❖Results can be applied to understanding pathways of stress response and lifestyle in diseases: Alzheimer's, prion diseases, and Lewy body dementia

❖Basis for studying genetic aspects of the disease and other molecular factors involved in neurodegeneration

Key Citations

[1] McGirr, S., Venegas, C., & Swaminathan, A. (2020). Alzheimer's Disease: A Brief Review. *Journal of Experimental Neurology*, *1*(3), 89–98. https://doi.org/10.33696/neurol.1.015

[2] Breijyeh, Z., & Karaman, R. (2020). Comprehensive Review on Alzheimer's Disease: Causes and Treatment. *Molecules*, *25*(24), 1–28. https://doi.org/10.3390/molecules25245789

[3] Dhana, K., Beck, T., Desai, P., Wilson, R. S., Evans, D. A., & Rajan, K. B. (2023). Prevalence of Alzheimer's disease dementia in the 50 US states and 3142 counties: A population estimate using the 2020 bridged-race postcensal from the National Center for Health Statistics. *Alzheimer's & Dementia*, *19*(10), 4388–4395. https://doi.org/10.1002/alz.1308