The Role of Tau and the RNA-Binding Protein Nab2 on Cholinergic Degeneration in *GAL4-UAS Drosophila melanogaster* Models

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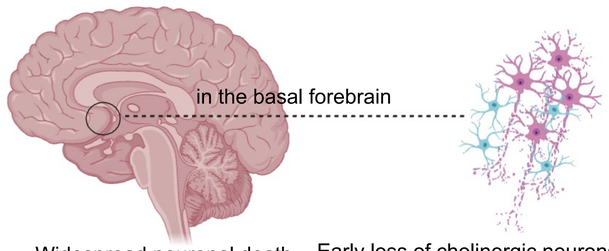
Introduction

Cholinergic degeneration in Alzheimer's Disease (AD)



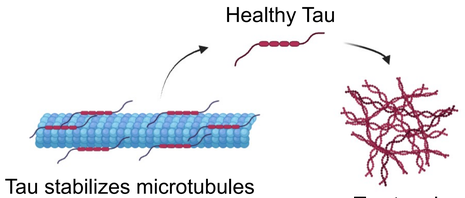
Widespread neuronal death in AD

Early loss of cholinergic neurons affects vision, sleep, and motor movements

Acetylcholine produced by cholinergic neurons is synthesized by ChAT (cholineacetyltransferase) and broken down by AChE (acetylcholinesterase).

Neurofibrillary tangle formation

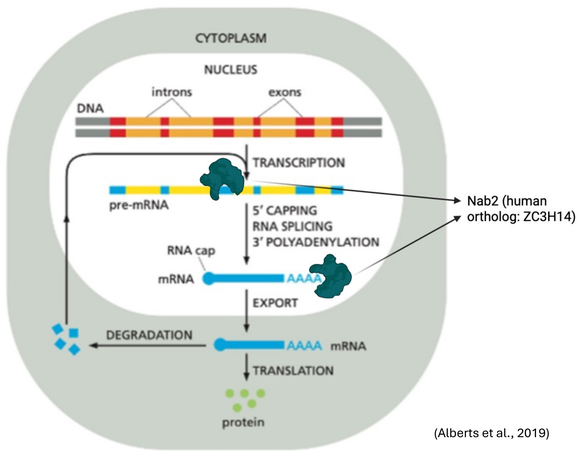
Pathological tau forms NFTs and contributes to neuronal loss, synaptic dysfunction, and cognitive decline.



Tau stabilizes microtubules

Tau tangles

mRNA Processing



(Alberts et al., 2019)

RNA-Binding proteins are associated with many neurodegenerative diseases, including ZC3H14, which modulates tau aggregations and causes neuronal death when overexpressed. However, Nab2 overexpression lowers tau levels.

Goals

Does tau contribute to early cholinergic degeneration in AD by affecting ChAT mRNA and protein levels? Can Nab2 reverse these changes and prevent cholinergic degeneration?

Drosophila GAL4-UAS Models can be used to study neurodegeneration

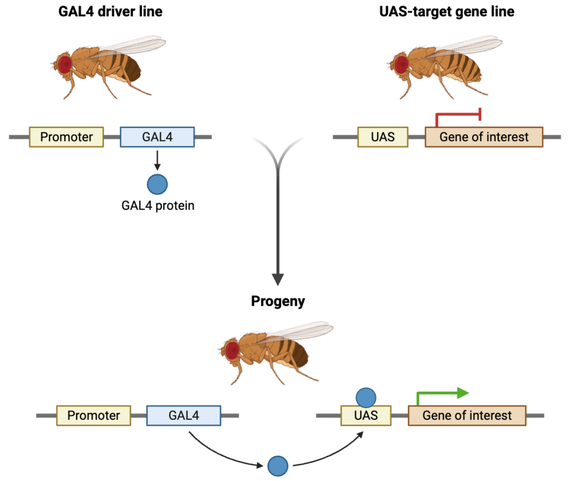


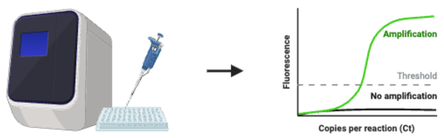
Figure 1: The GAL4-UAS System. *Drosophila* carrying the gene encoding the GAL4 protein are crossed with *Drosophila* carrying the UAS promoter next to the gene of interest to produce progeny in which the GAL4 gene drives transcription of the gene of interest.

Drosophila mushroom bodies have significant cholinergic extensions

Methodology

18-21-day-old female *Drosophila* overexpressing tau and Nab2

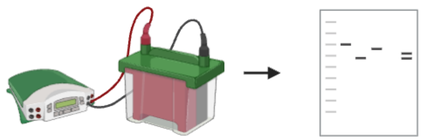
1. mRNA expression analysis



RT-qPCRs using synthesized cDNA

Ct values used for analysis

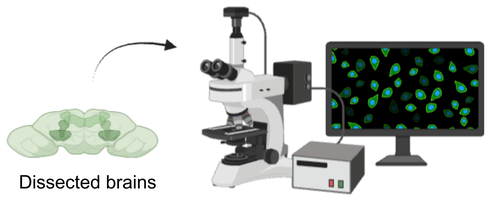
2. Protein expression analysis



Western Blots

Antibody binding on nitrocellulose membrane

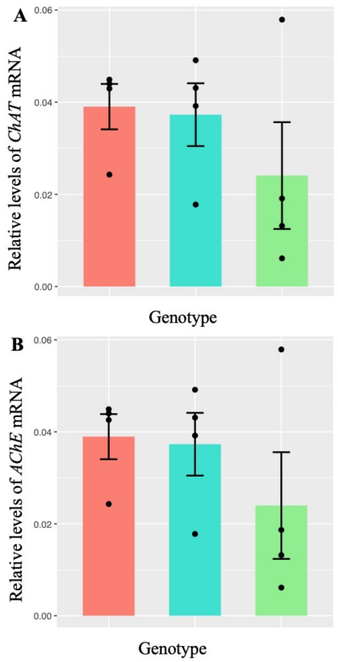
3. Visualizing ChAT-GFP neurons



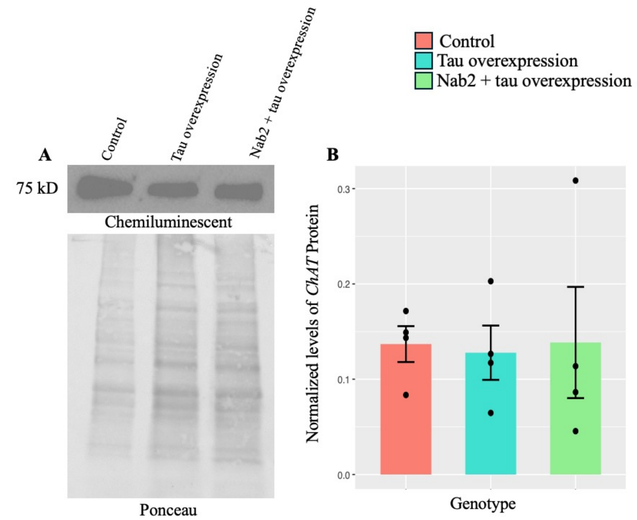
Dissected brains

Results

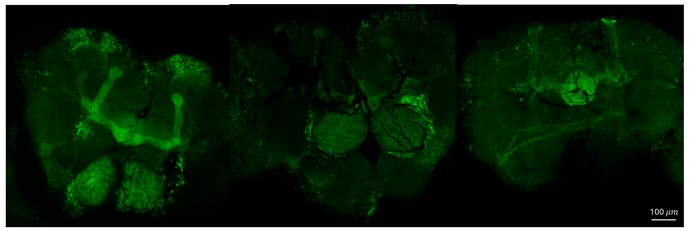
Tau and Nab2 overexpression does not affect ChAT or AChE mRNA levels



Tau and Nab2 overexpression does not affect ChAT protein levels



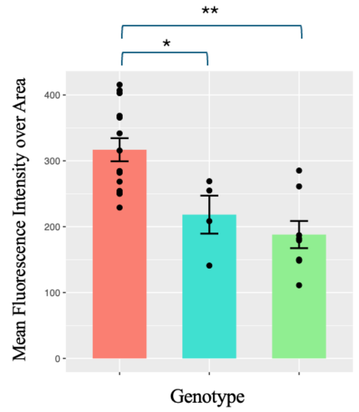
Tau and Nab2 overexpression significantly decreases GFP intensity in *Drosophila* mushroom bodies



Control

Tau overexpression

Nab2 + Tau overexpression



Conclusions and Future Directions

- Nab2 does not rescue tau-induced degeneration of the mushroom bodies.
- Tau and Nab2 overexpression do not affect mRNA and protein levels of ChAT and AChE.
- Nab2 may not be a potential candidate to address cholinergic degeneration.

➢Analyze the degree of cholinergic degeneration in mushroom bodies via single-cell RNA sequencing.
➢Clarify the exact connections between tau and Nab2.
➢Using older flies with severe degeneration to investigate changes in ChAT and AChE expression.
➢Validate Nab2 and tau overexpression.

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