Pharmacological Therapeutic Interventions in Anatomical Aging and Molecular Imaging for STUB1 Ataxias

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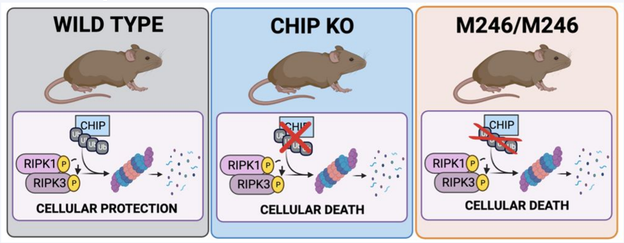
Introduction

Key Points

- **Necroptosis** is a programmed cell death pathway, regulated by receptor-interacting protein kinase 1 and 3 (**RIPK1** and **RIPK3**) carboxyl terminus of heat shock 70-interacting protein (**CHIP**).
- RIPK1/3 are substrates of CHIP ubiquitin ligase domain which targets and degrades them.
- CHIP polymorphisms result in a hereditary form of ataxia known as **STUB1 ataxias**. Major issues in ataxias include anatomical aging with a decrease in growth, as well as molecular aging with decreased neuron density, specifically with Purkinje cells.
- Genotypic changes that affect CHIP expression and related proteins can affect necroptotic activity:



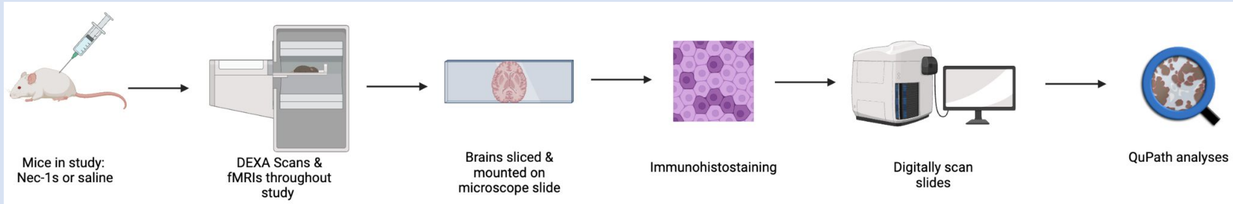
Hypothesis

- **The pharmacological therapeutic intervention of Nec-1s will decrease anatomical aging through reduced growth and will show increased neuronal cell count (particularly Purkinje cell count) in molecular imaging data.**

Methodologies

- **Anatomical Aging:**
  - **DEXA Scans** reveals information about anatomical aging and bone composition.
- **Molecular Imaging:**
  - **Immunohistostaining** allows to view structures in the brain and reveals information about molecular , being able to analyze effects of treatments → Cresyl Violet (**CV**), Hematoxylin and Eosin (**H&E**), **Madder**, **Calbindin**, & Neuronal Nuclei (**NeuN**) staining.

Experimental Design



Results

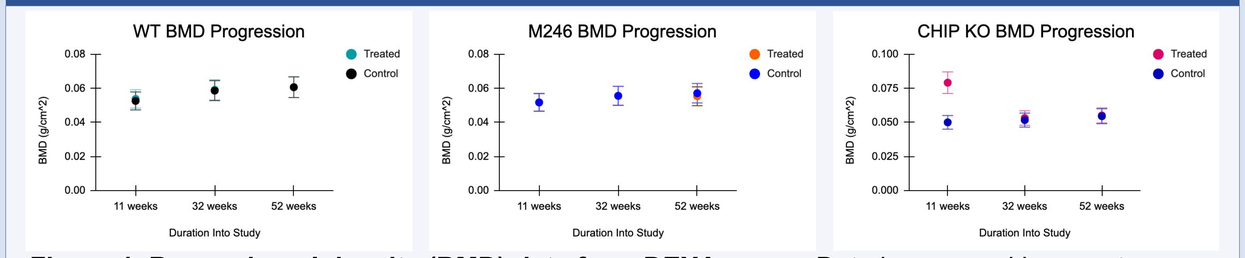


Figure 1. Bone mineral density (BMD) data from DEXA scans. Data is arranged by genotype and treatment. Data has not been normalized.

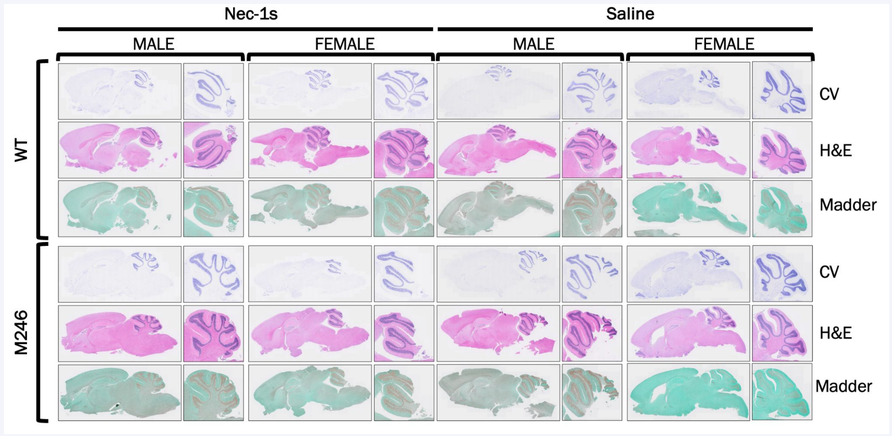


Figure 2. Histology of WT vs. M246. The figure contains the stained microscope slide images of brain slices from male & female, which received either Nec-1s or saline treatments (n = 16). Slides were stained with CV, H&E, and Madder stains.

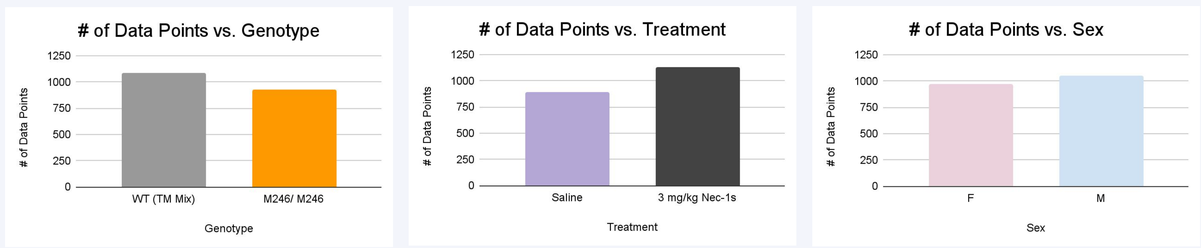


Figure 3. QuPath analyses data on Madder stained brain slices. Data points were recorded for Purkinje cells in the cerebellar regions of the brain slices.

Conclusion

- **Anatomical Imaging**
  - The increase in change of bone mass density between the time points decreases, suggesting a decrease in growth, and potential increased anatomical aging, as the mice get older and reaching the end of the study duration.
  - WT mice have the highest range of BMD values throughout the study duration, followed by M246 and CHIP KO.
  - There does not seem to be a clear correlation between treatment mice and increased BMD values in comparison to the control mice; therefore, more data values need to be assessed in the future.
- **Molecular Imaging**
  - M246 mice had decreased neuronal cell count from QuPath analyses compared to WT mice.
  - The treatment group mice that received Nec-1s had higher neuronal cell counts from QuPath analyses compared to the control group mice that received saline.
  - Males and females across the groups had similar neuronal cell counts, with females having slightly fewer cell counts on average compared to males.
- **Overall, pharmacological changes can serve as therapeutic interventions for STUB1 ataxias by reducing anatomical and molecular aging!**

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