Report for 2022 Research Grant "Assessing Prophylactic and Therapeutic Efficacy of a Cellulose Ether Compound TC-5RW on CJD"

Zerui Wang, Case Western Reserve University, Cleveland, OH, USA, 44106

Title: Assessing Prophylactic and Therapeutic Efficacy of a Cellulose Ether Compound TC-5RW on CJD

Project Objective: The primary objective of this study is to develop effective prevention and treatment strategies for human prion diseases, specifically targeting Creutzfeldt-Jakob Disease (CJD). We are investigating the potential prophylactic and therapeutic effects of the cellulose ether compound TC-5RW.

*We hypothesized that small molecules, such as cellulose ethers, could provide a novel approach to therapeutic intervention. TC-5RW was identified as a candidate due to its minimal cytotoxicity and ability to cross the blood-brain barrier. Initial studies in prion-diseased mouse models showed that TC-5RW could prolong survival when administered as a pre-treatment.

Summary of Accomplishments to Date

**In Vitro* Studies:

Inhibition of Prion Aggregation: TC-5RW was found to inhibit the seeding activity of the infectious human prion protein (PrP^{Sc}). This compound also decreased the levels of proteinase K (PK)-resistant PrP^{Sc} when incubated with brain homogenates from patients with different types of CJD directly at 37°C.

Dose-Dependent Effects: Our tests showed that the amount of PK-resistant PrP^{Sc} decreased in a dose-dependent manner. Lower concentrations of TC-5RW were effective against the most common subtypes of sporadic and genetic CJD, except for fatal familial insomnia (FFI), which required higher concentrations.

*Animal Studies:

<u>Prophylactic Efficacy in Tg40h Mice:</u>

Increased Survival Time: In transgenic humanized prion mouse model, Tg40h mice, which mimic acquired CJD, prophylactic administration of TC-5RW increased survival times significantly (215.7 days post-inoculation vs. 205.6 days in controls).

Reduced PrP^{Sc} Levels: The amount of brain PK-resistant PrP^{Sc} was significantly lower in treated mice compared to controls.

<u>Therapeutic Efficacy in Tg40h Mice:</u>

No Significant Improvement: When administered after prion exposure, TC-5RW did not significantly improve survival times or reduce PrP^{Sc} levels.

<u>Prophylactic Efficacy in TgMHu2ME199K Mice:</u>

Reduced Disease Severity: In TgMHu2ME199K mice, which mimic genetic CJD, early prophylactic treatment with TC-5RW significantly reduced disease severity scores and PK-resistant PrP^{Sc} levels.

Therapeutic Efficacy in TgMHu2ME199K Mice:

Limited Improvement: Later therapeutic treatment showed some reduction in disease severity but was less effective in reducing PrP^{Sc} levels.

Conclusion

Our research demonstrates that TC-5RW Show protective effect in **prophylactic group in humanized transgenic mice in both sCJD and E200K models,** which indicates that TC-5RW has strong potential as a preventive measure against human prion diseases, significantly increasing survival times and reducing pathological markers when administered early. However, its therapeutic efficacy post-infection needs further improvement. Future studies will focus on optimizing the compound's delivery and exploring combination therapies to enhance its protective effects.

Key Findings and Implications for the Prion Disease Field

Prophylactic Potential: TC-5RW shows strong potential as a prophylactic agent. Early administration before prion exposure can significantly increase survival times and reduce pathological markers in prion diseases.

Challenges in Therapeutic Application: The compound's effectiveness as a therapeutic agent after prion infection is limited, indicating that early intervention is crucial.

Mechanism of Action: TC-5RW likely disrupts the aggregation process of prion proteins, which is critical in the progression of prion diseases.

Next Steps in Our Work

Optimize Administration: To enhance the compound's efficacy, we will explore alternative routes of administration, such as:

Subcutaneous Implantation: This method may provide a sustained release of the compound, increasing its duration of action.

Intracerebral Injection: Direct administration to the brain may improve the compound's therapeutic effects.

Investigate Mechanisms: We aim to identify the specific binding targets or pathways through which TC-5RW interacts with prion proteins, providing insights into its mechanism of action.

Synergistic Compounds: We will search for other small molecules that could work synergistically with TC-5RW to enhance its therapeutic effects.

Further *In Vivo* Studies: Continued animal studies will help refine dosing and administration schedules to maximize both prophylactic and therapeutic outcomes.

Acknowledgement: Thanks for the Strides for CJD Grant and funding from CJD Foundation, and the donors listed below: The Garry Buttermann, IV Memorial Research Grant, contributed by Jill Canuel. The Dora Middendorf Memorial Grant contributed by Renee Middendorf and Family. The Lynda Morris Memorial Grant, contributed by David Morris and Family. The Eugene A. Riedel Memorial Research Grant, contributed by Jacqueline Riedel. The Dr. Robert Brooks Memorial Grant, contributed by Loving Family &

Friends. The Paula Smart Memorial Research Grant, contributed by Jeff Moxie. The Strides for CJD Grant and CJD Foundation Grant