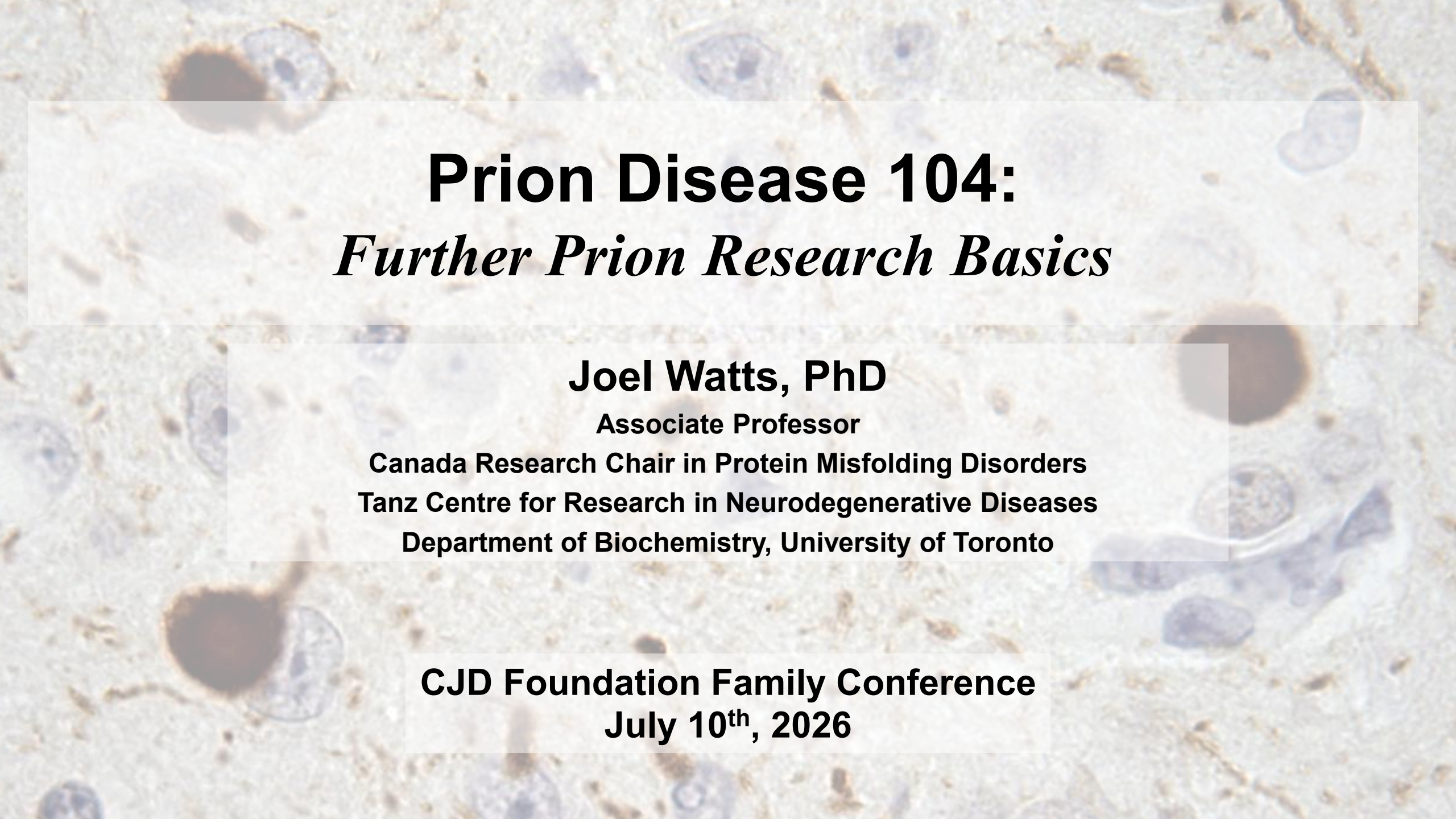




Prion Disease 104:

Further Prion Research Basics

Joel Watts, PhD

Associate Professor

Canada Research Chair in Protein Misfolding Disorders

Tanz Centre for Research in Neurodegenerative Diseases

Department of Biochemistry, University of Toronto

CJD Foundation Family Conference
July 10th, 2026

A Quick Biology Review



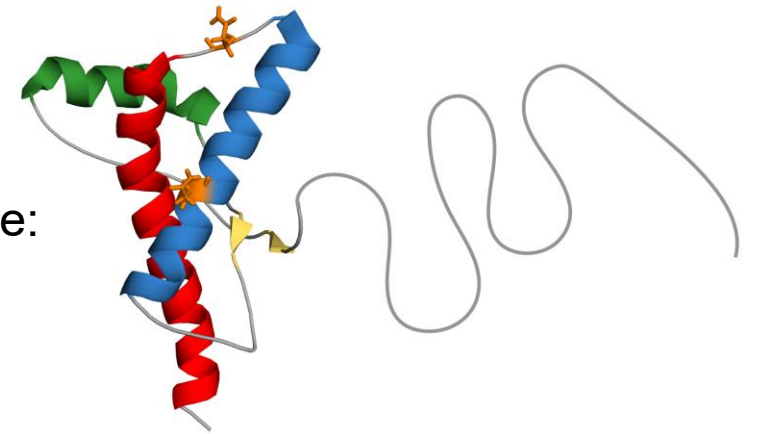
Prion (*PRNP*) Gene Sequence

```
ATGGCGAACCTTGGCTGCTGGATGCTGGTTCTCTTTGTGGCCACATGGAGTGACCTGGGC
CTCTGCAAGAAGCGCCCGAAGCCTGGAGGATGGAACACTGGGGGCAGCCGATACCCGGGG
CAGGGCAGCCCTGGAGGCAACCGCTACCCACCTCAGGGCGGTGGTGGCTGGGGGCAGCCT
CATGGTGGTGGCTGGGGGCAGCCTCATGGTGGTGGCTGGGGGCAGCCCCATGGTGGTGGC
TGGGGACAGCCTCATGGTGGTGGCTGGGGTCAAGGAGGTGGCACCCACAGTCAGTGGAAC
AAGCCGAGTAAGCCAAAAACCAACATGAAGCACATGGCTGGTGGCTGCAGCAGCTGGGGCA
GTGGTGGGGGGCCTTGGCGGCTACATGCTGGGAAGTGCCATGAGCAGGCCCATCATA CAT
TTCGGCAGTGACTATGAGGACCGTTACTATCGTGAAAACATGCACCGTTACCCCAACCAA
GTGTACTACAGGCCCATGGATGAGTACAGCAACCAGAACAACCTTTGTGCACGACTGCGTC
AATATCACAATCAAGCAGCACACGGTCACCACAACCACCAAGGGGGGAGAACTTCACCGAG
ACCGACGTTAAGATGATGGAGCGCGTGGTTGAGCAGATGTGTATCACCCAGTACGAGAGG
GAATCTCAGGCCTATTACCAGAGAGGATCGAGCATGGTCCTCTTCTCCTCTCCACCTGTG
ATCCTCCTGATCTCTTTCCTCATCTTCCTGATAGTGGGATGA
```

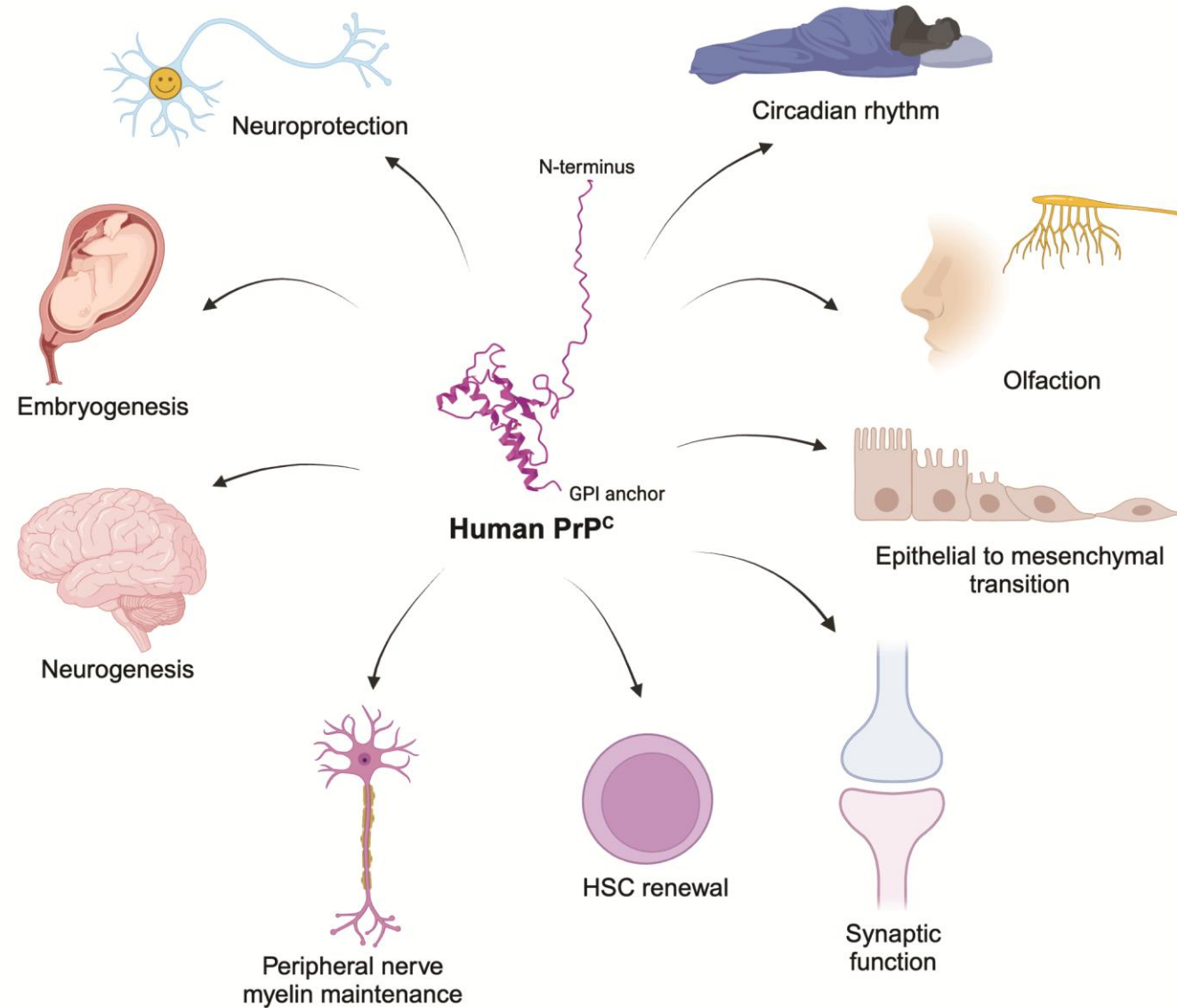
Prion Protein (PrP) Sequence

```
MANLGCWMLVLFVATWSDLGLCKKRPKPGGWNTGGSRYPGQGSPPGGNRYPPQGGGGWGQP
HGGGWGQPHGGGWGQPHGGGWGQPHGGGWGQGGGTHSQWNKPSKPKTNMKHMAGAAAAGA
VVGGLGGYMLGSAMSRPIIHFGSDYEDRYRENMHRYPNQVYYRPMDEYSNQNNFVHDCV
NITIKQHTVTTTTTKGENFTETDVKMMERVVEQMCITQYERESQAYYQRGSSMVLFSPPV
ILLISFLIFLIVG
```

PrP^C Structure:

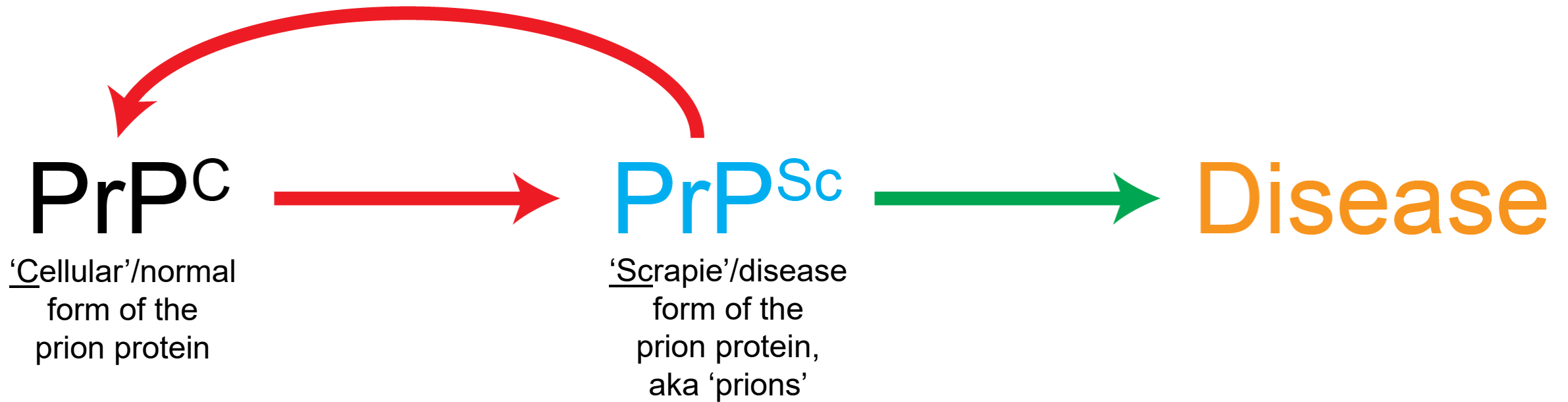


Why Do We Have PrP^C To Begin With??



The 'Prion Principle'

PrP^{Sc} acts as a 'seed' that converts
PrP^C into additional PrP^{Sc}



What is a Prion?

PrP^C



Normal protein movements

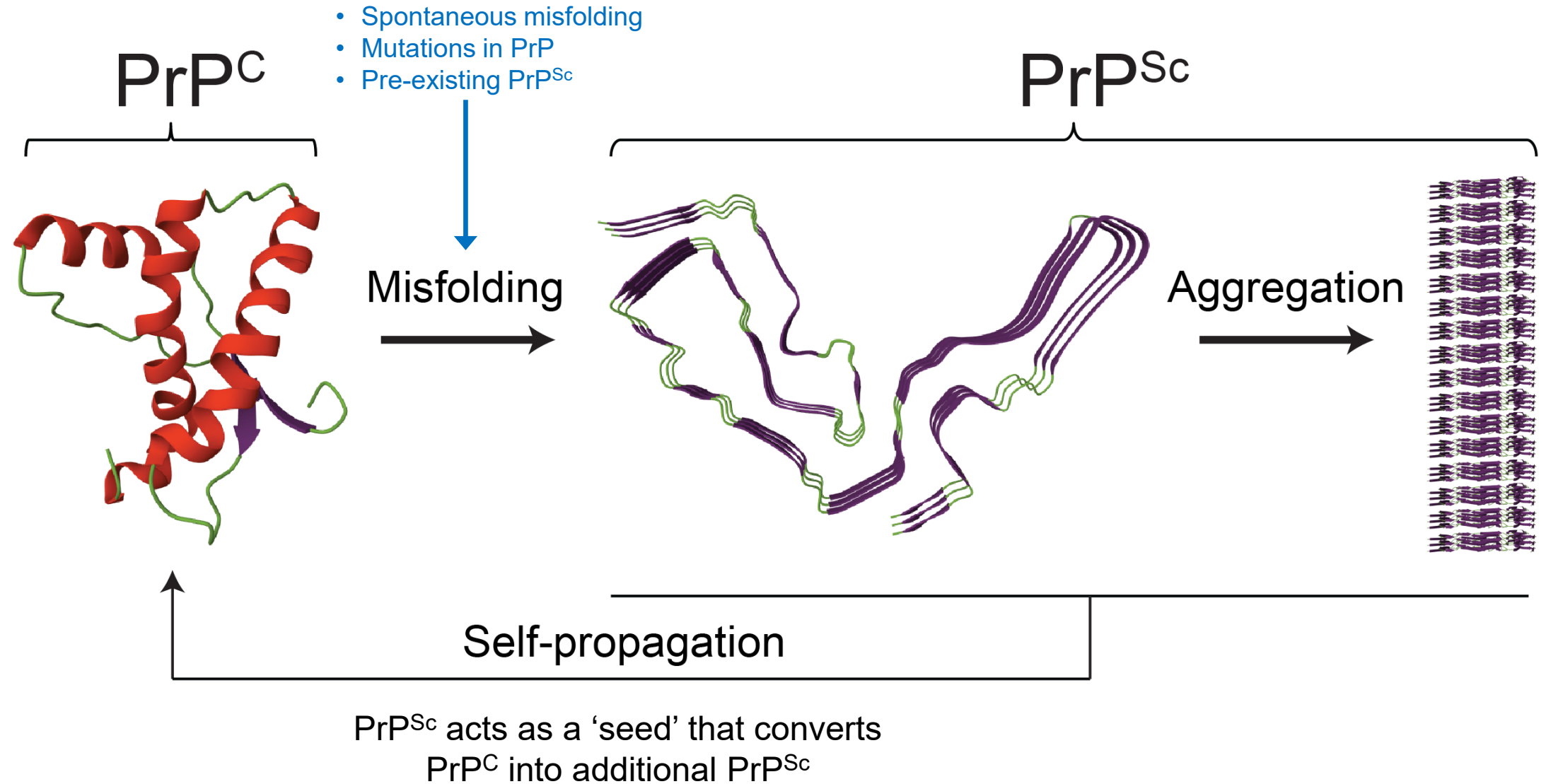


PrP^{Sc}

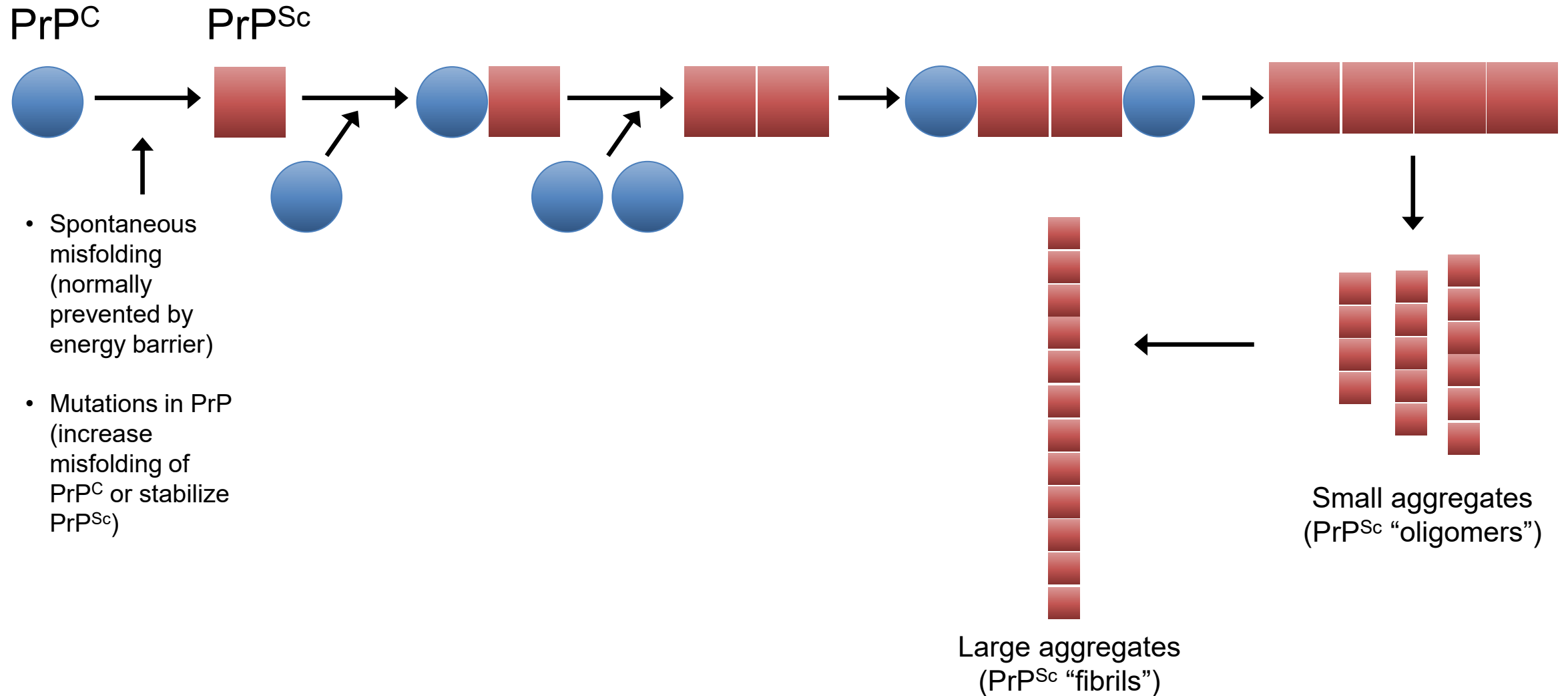


Conversion into a dangerous shape

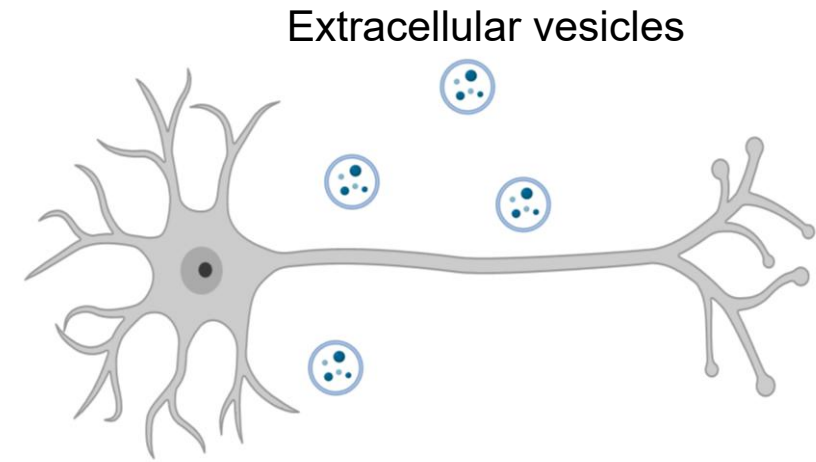
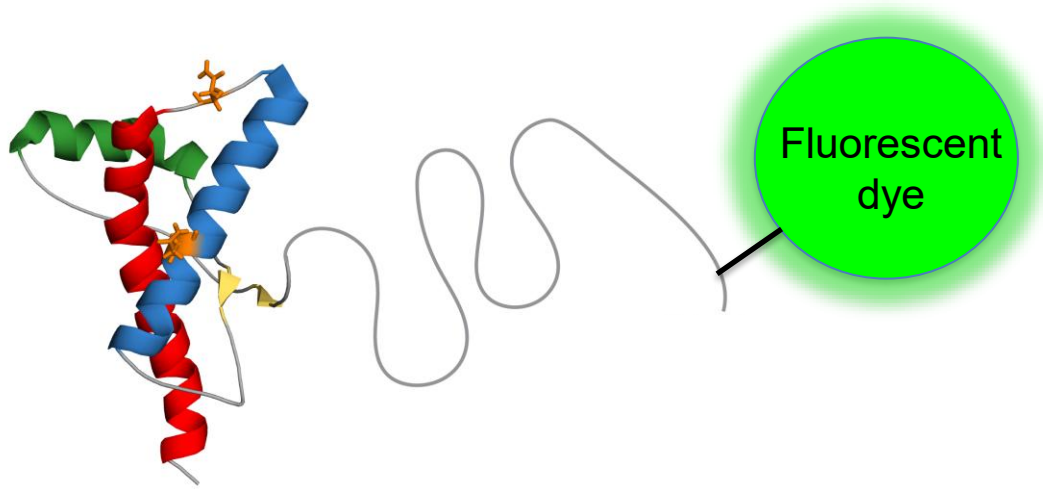
The 'Prion Principle'



Prion Replication and Propagation



Visualizing Prions and Prion Replication in Cells



Dr. Szymon Manka/Ellie Hanzi

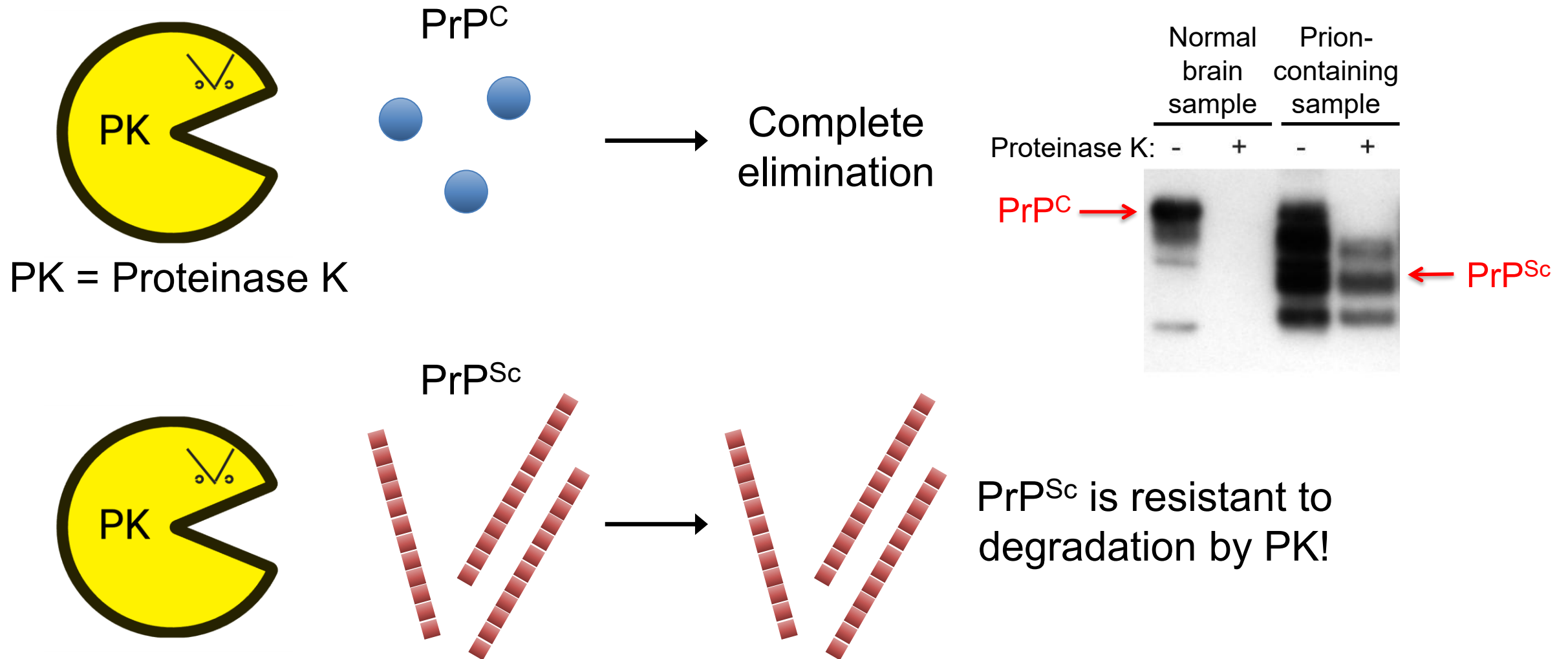
“Engineering human cells to propagate fluorescent prions for live monitoring of prion infection and high-resolution *in situ* prion structure determination”



Dr. Jakub Soukup

“Higher-resolution visualization of prion fibrils in neurons and extracellular vesicles using state-of-the-art Cryo-TEM and FIB-SEM imaging”

How do Researchers Distinguish Between PrP^C and PrP^{Sc}?



Prions Are Infectious Proteins

Inoculate mouse prions
intracerebrally



Mice

Inoculate hamster prions
intracerebrally



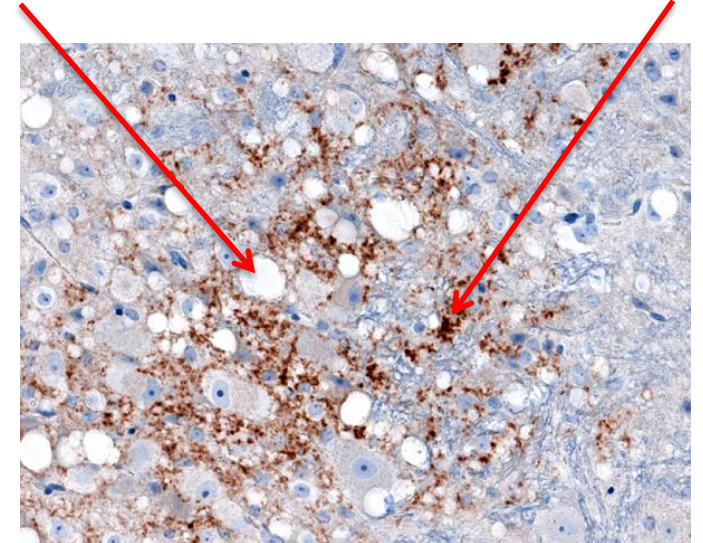
Hamsters

Incubation periods

- Mouse prions: ~120-150 days
- Hamster prions: ~70-80 days

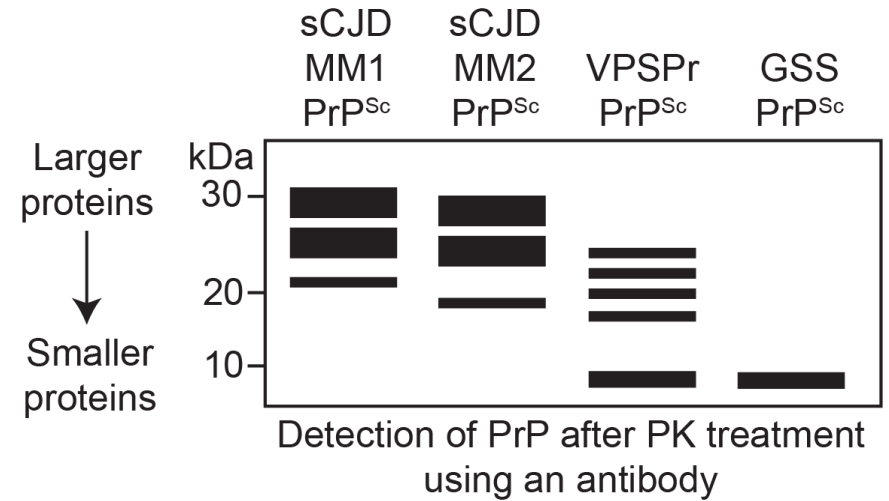
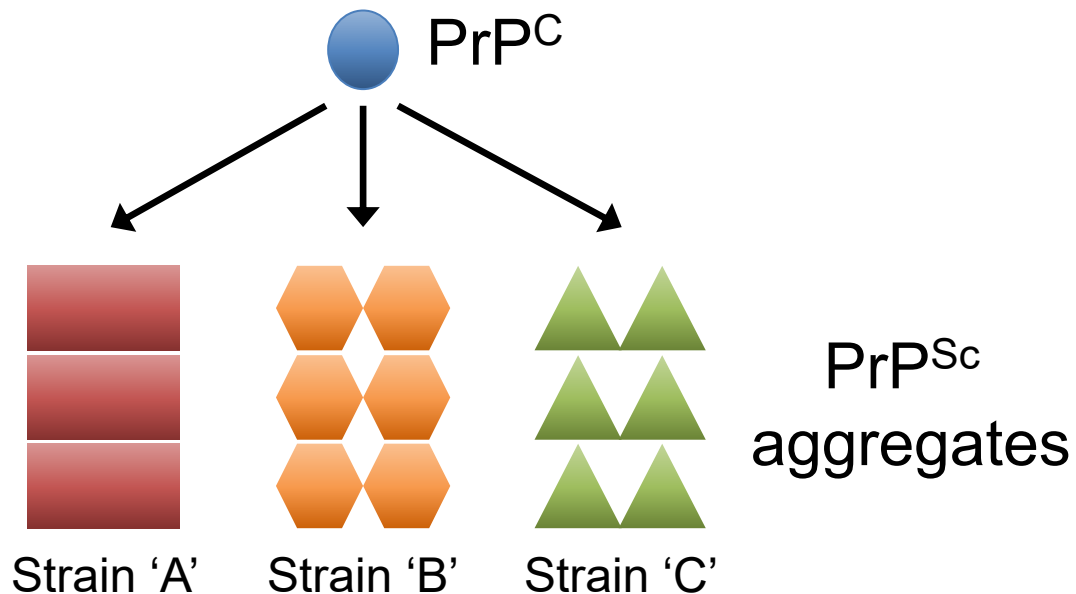
Spongiform change
(vacuolation)

PrP^{Sc} deposits

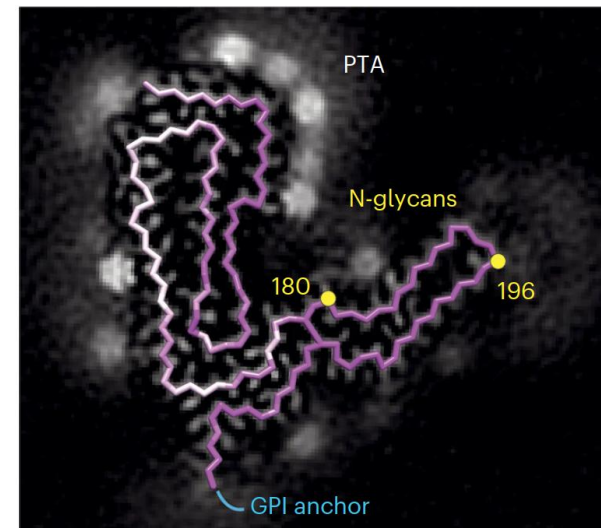


Prion Strains

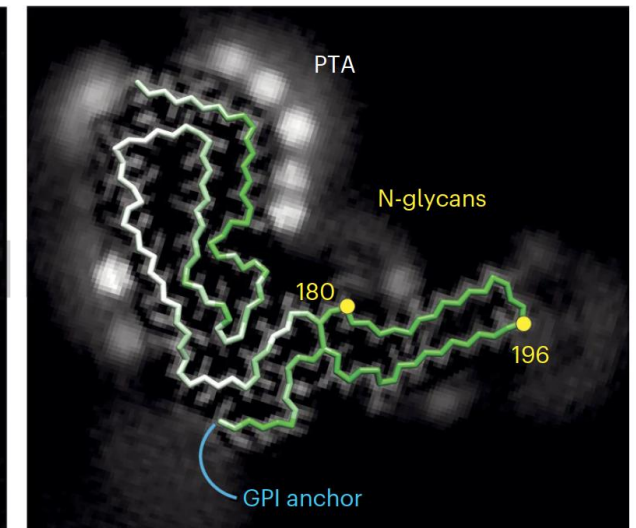
- Clinical and pathological variability in the prion diseases (both in laboratory animals and humans) can be explained by the existence of distinct 'strains' of prions
- Prion strains represent distinct structures of PrP^{Sc} aggregates



ME7 mouse prion strain



RML mouse prion strain



Manka *et al.*, *Nature Chemical Biology*, 2023

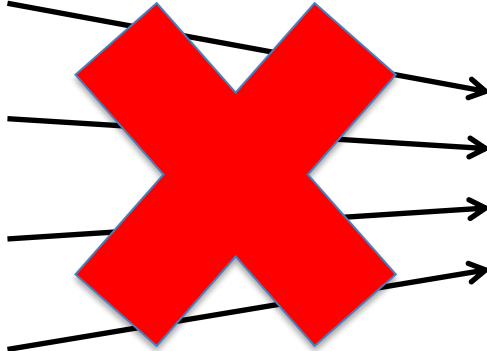
Prion Replication and the Species Barrier

Hamster prions

Human prions

Deer prions

Sheep prions



Mice

Poor Transmission
(Low transmission rates,
long incubation periods)

"Species Barrier"

Inoculate CJD
brain extract



Efficient Transmission

No Species Barrier

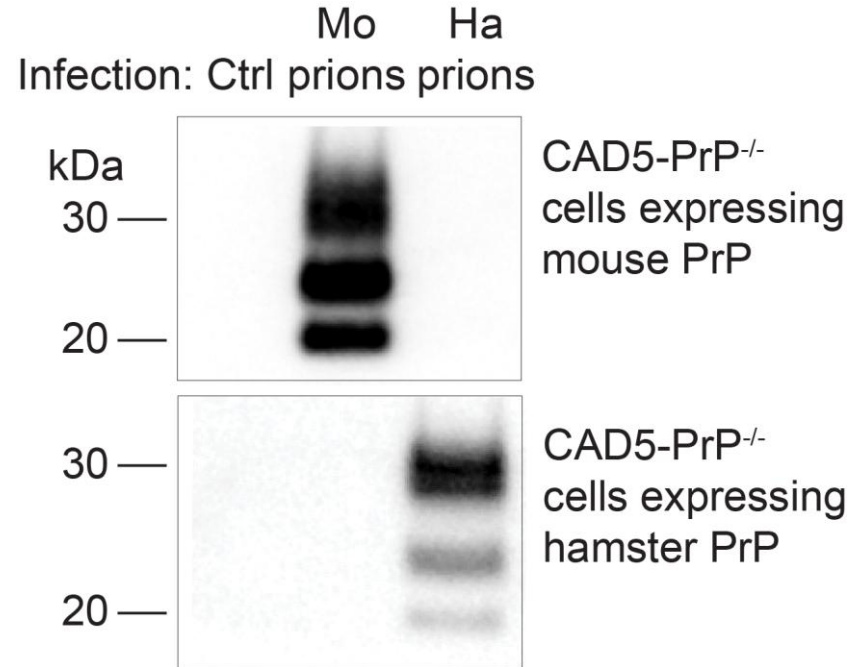
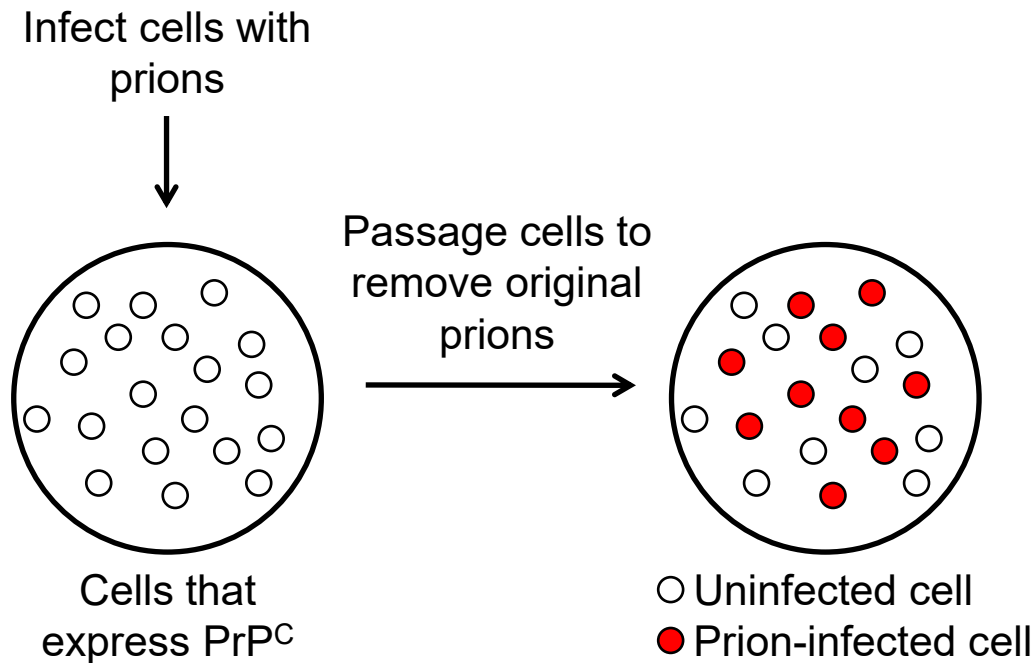
Transgenic Mouse
Expressing Human PrP^C



Dr. Alyssa Block

"Refined transgenic approaches to investigate
selective adaptation of human prions during
intracranial and peripheral transmissions"

Prion Replication in Cultured Cells



+ PK

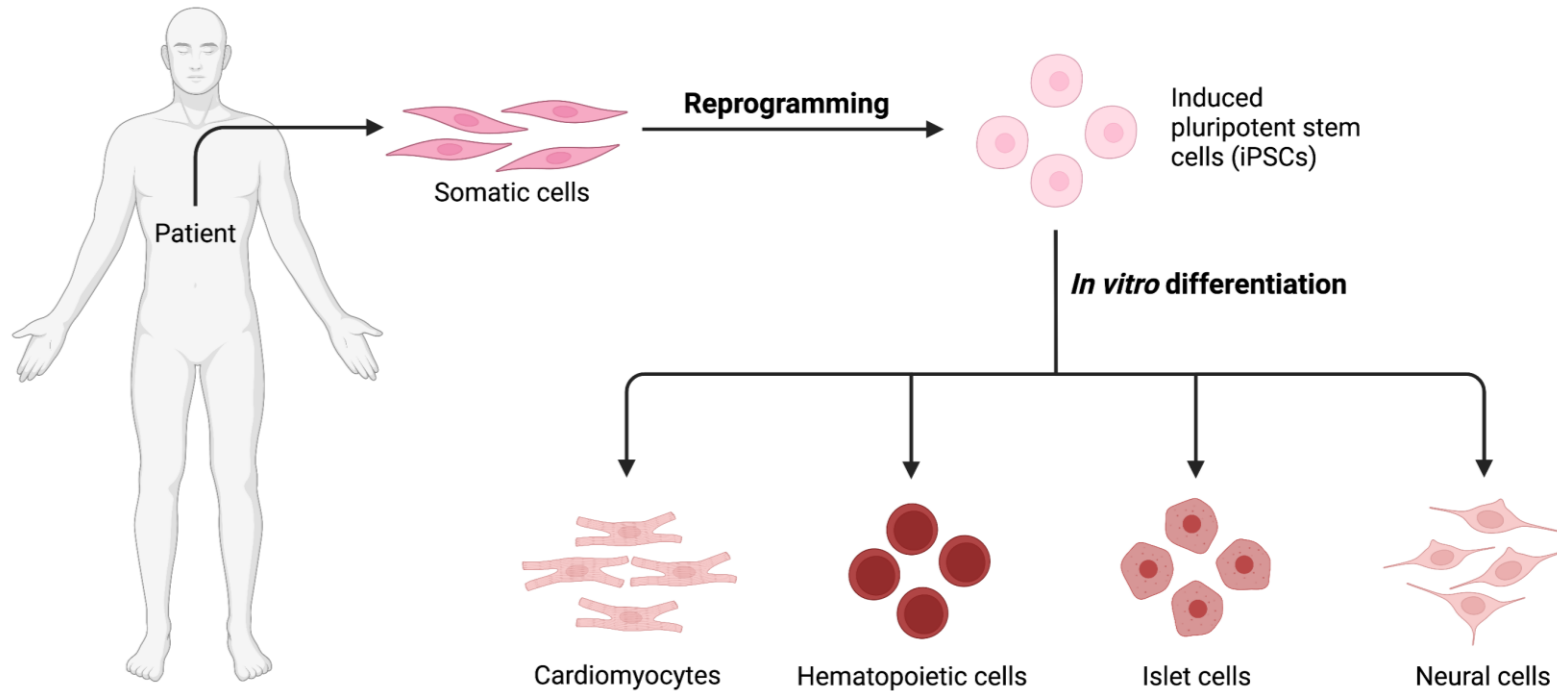
Arshad *et al.*, *J Neurochem*, 2023



Dr. Joel Watts

“Engineered biosensors for improved prion detection”

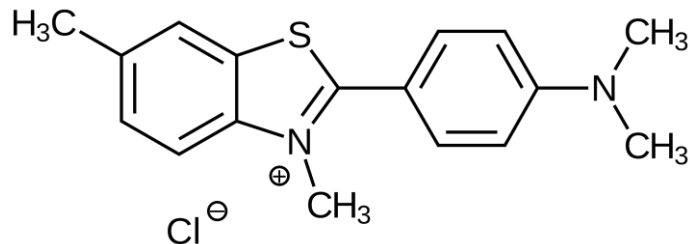
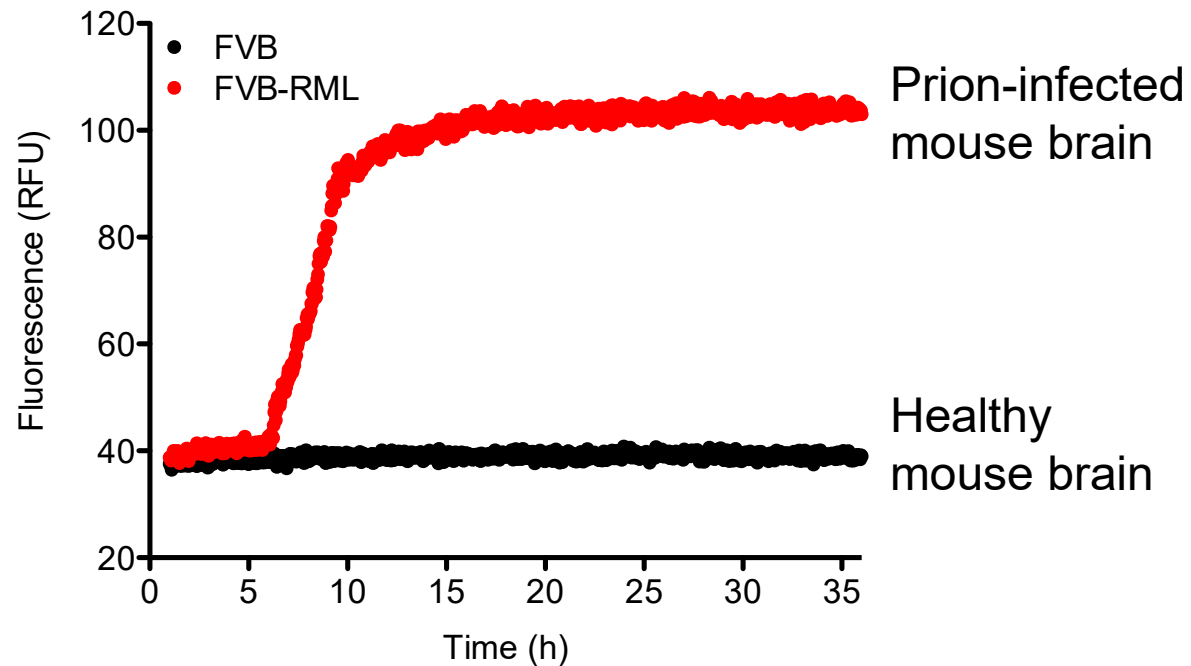
Using Patient-Derived Cells to Study Mechanisms of Prion Toxicity



Dr. Jose Andres Alepuz Guillen

“Characterization of synaptic abnormalities in iPSC-derived neurons from E200K carriers”

Prion Detection Using RT-QuIC



Thioflavin T: Becomes fluorescent when bound to protein aggregates

- Addition of PrP^{Sc} to recombinant PrP^C and incubation at 37°C with shaking results in the production of PrP aggregates that can be detected using Thioflavin T fluorescence
- RT-QuIC is extremely sensitive: it can detect minute quantities of prions in the CSF, olfactory swabs, and skin from sporadic CJD patients

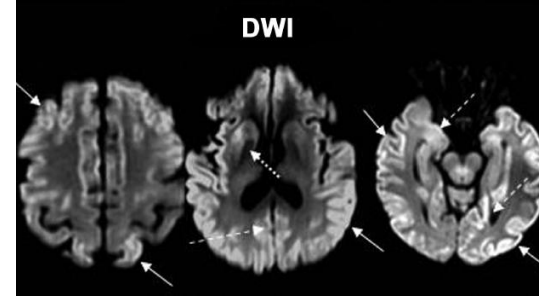


Dr. Gianluigi Zanusso

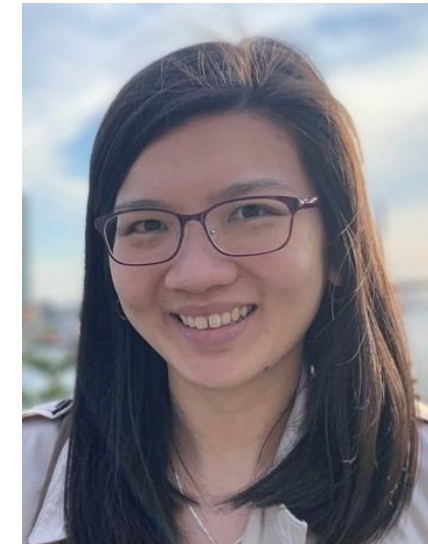
“Multi-center assessment of olfactory swab procedure performed by non-otolaryngologists for improving and simplifying human prion disease diagnosis”

Prion Disease Diagnosis and Biomarkers

- Imaging biomarkers:
 - Magnetic resonance imaging (MRI)
 - Electroencephalogram (EEG)
- CSF biomarkers:
 - *Not-so-specific for prion disease:*
 - Tau protein levels
 - 14-3-3 protein levels
 - Neurofilament light chain (NfL) levels
 - *Specific for prion disease:*
 - RT-QuIC positivity
 - PrP levels (for PrP-lowering therapies)



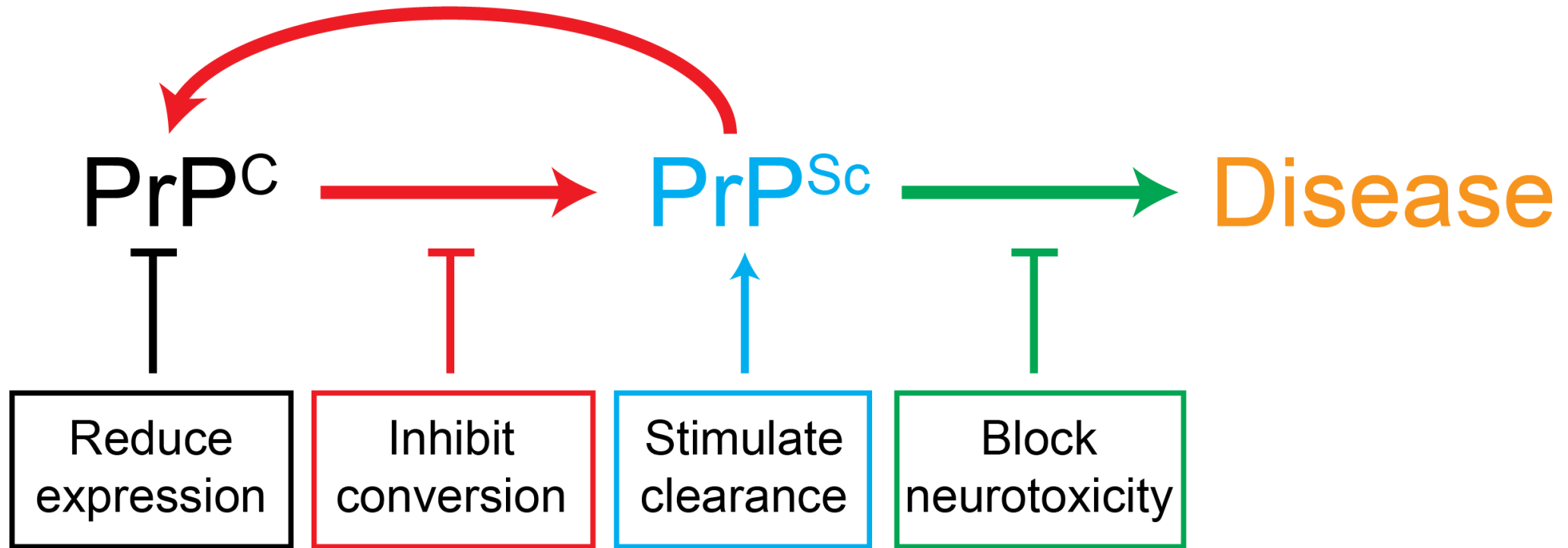
Vitali *et al.*, *Neurology*, 2011



Dr. Pei Ying Lee

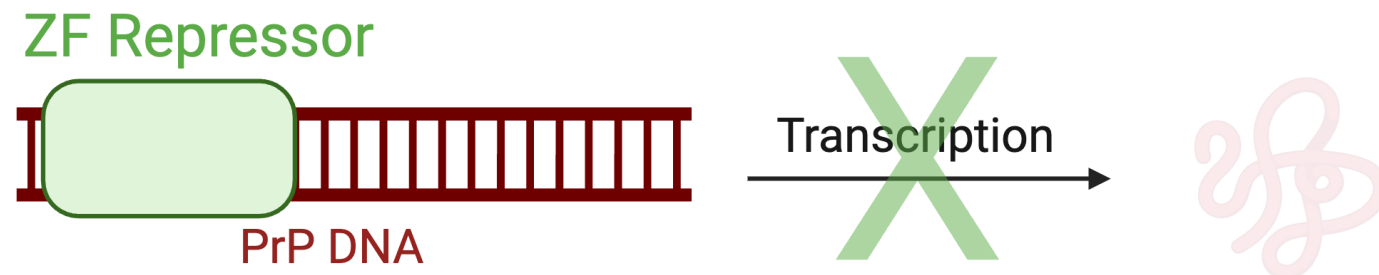
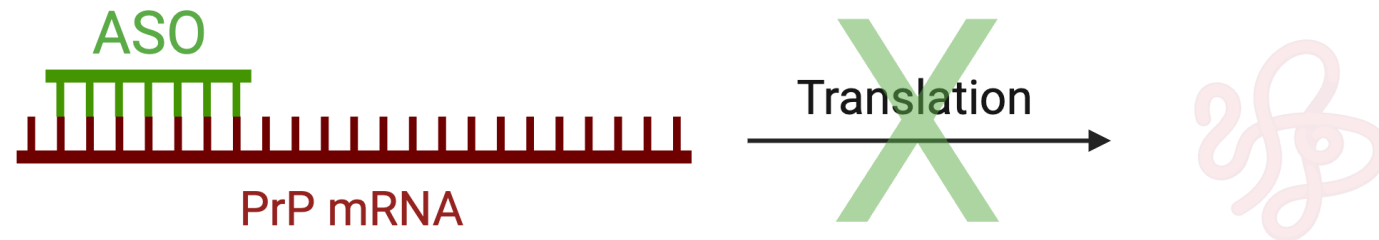
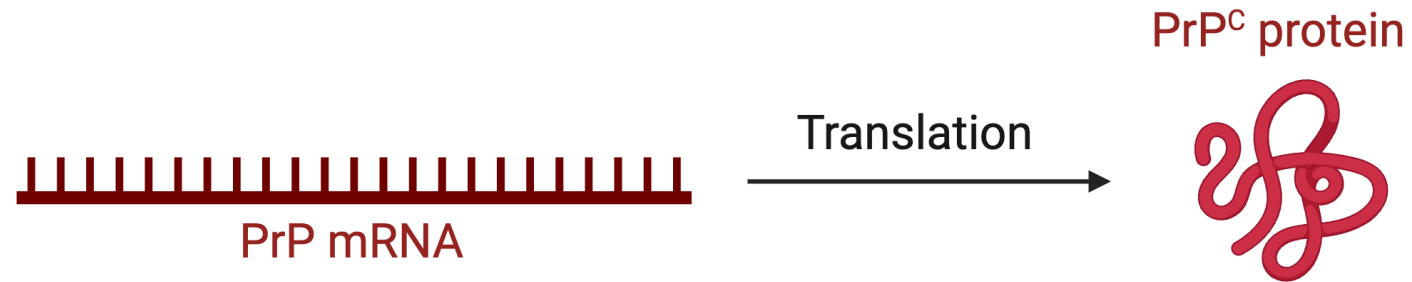
“Assessing retinal function as a potential early biomarker for Creutzfeldt-Jakob Disease (CJD)”

The Search for Prion Disease Therapeutics

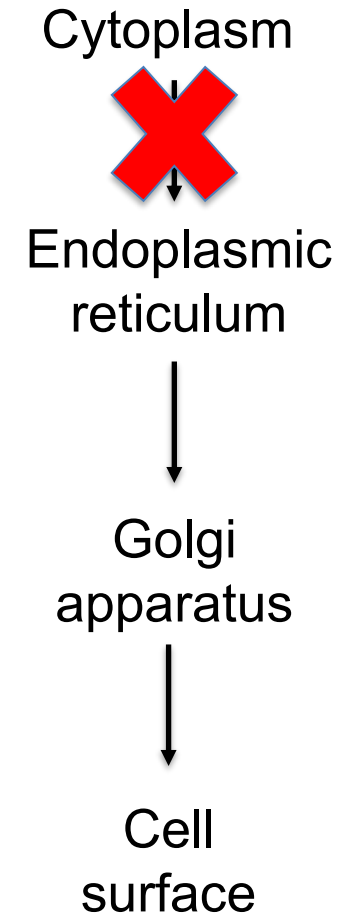
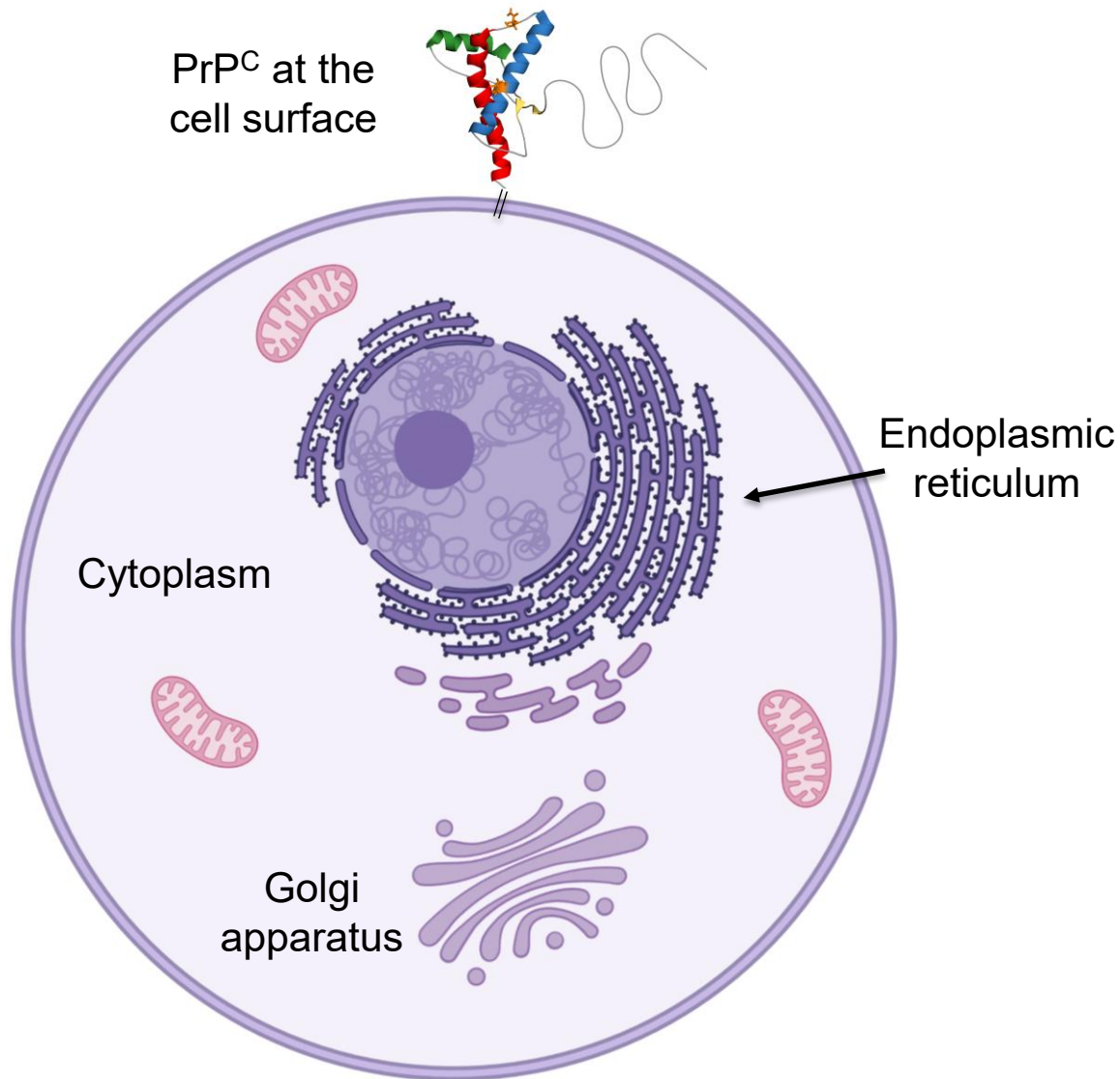


Strategies for Reducing PrP^C Levels

DNA $\xrightarrow{\text{Transcription}}$ mRNA $\xrightarrow{\text{Translation}}$ Protein



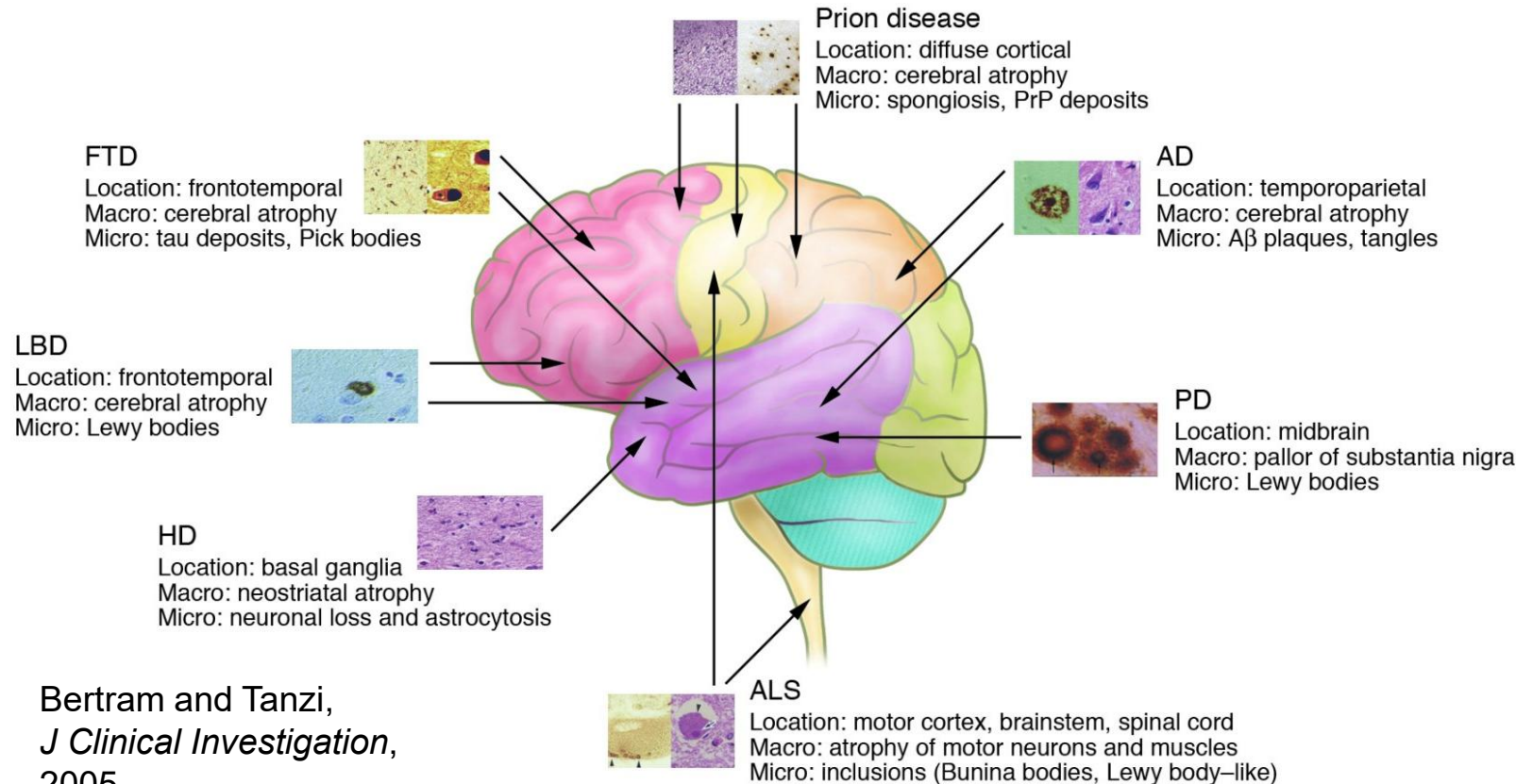
Blocking PrP^C Trafficking to the Cell Surface



Dr. Nina Oberbeck

“Assessing efficacy of a PrP-lowering Molecular Gate as a therapeutic for prion disease”

Neurodegenerative Diseases

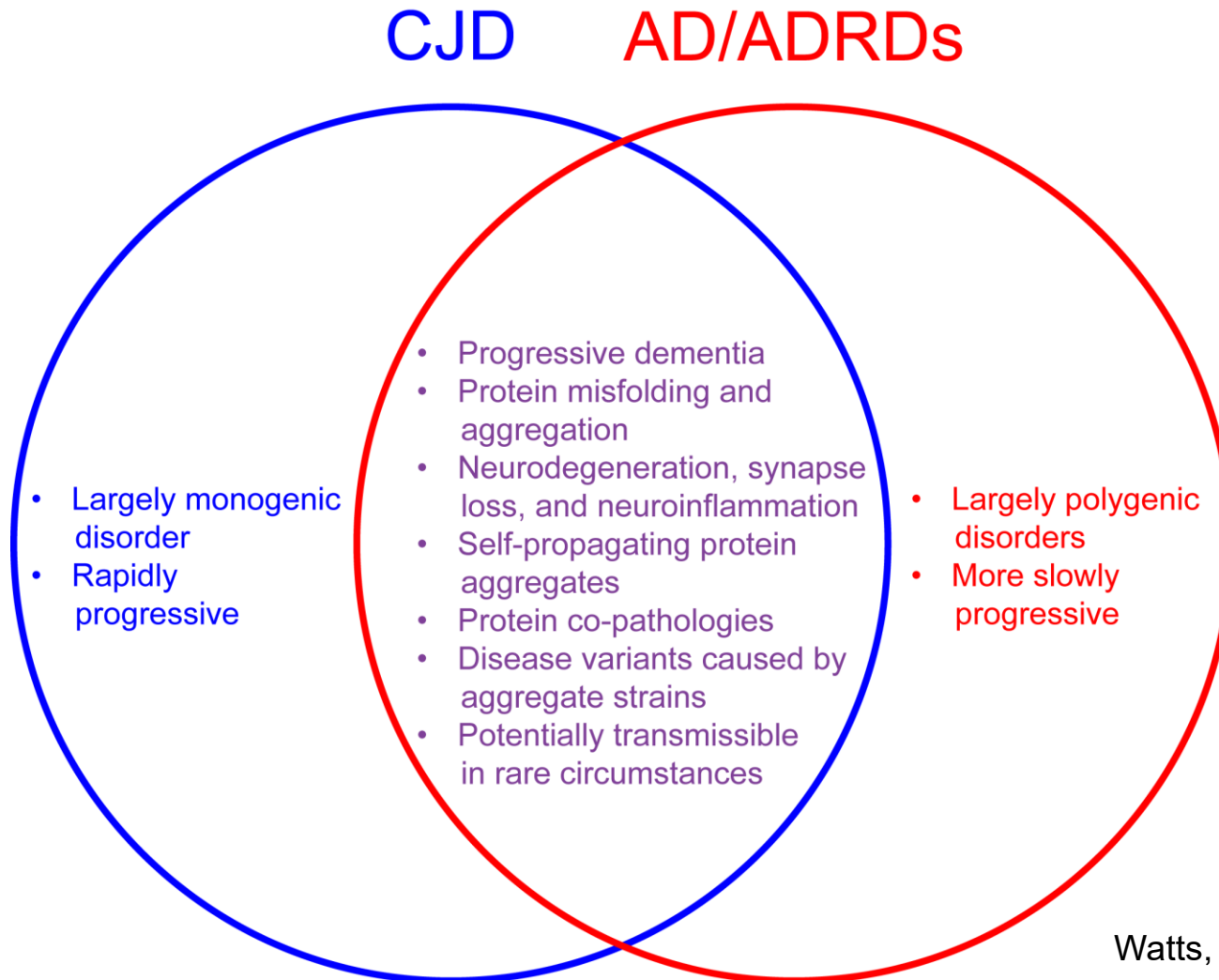


Dr. Ignazio Cali

“Investigating prion protein seeding activity in non-prion disease dementia”

- Common themes in neurodegenerative diseases:
 - Loss of neurons in the brain or peripheral nervous system
 - Protein misfolding and aggregation
 - Protein co-pathologies

Overlaps Between CJD and Other Neurodegenerative Diseases



Fundamental prion disease research can lead to major advances in our ability to understand, diagnose, and treat other human neurodegenerative diseases!

ANY QUESTIONS??