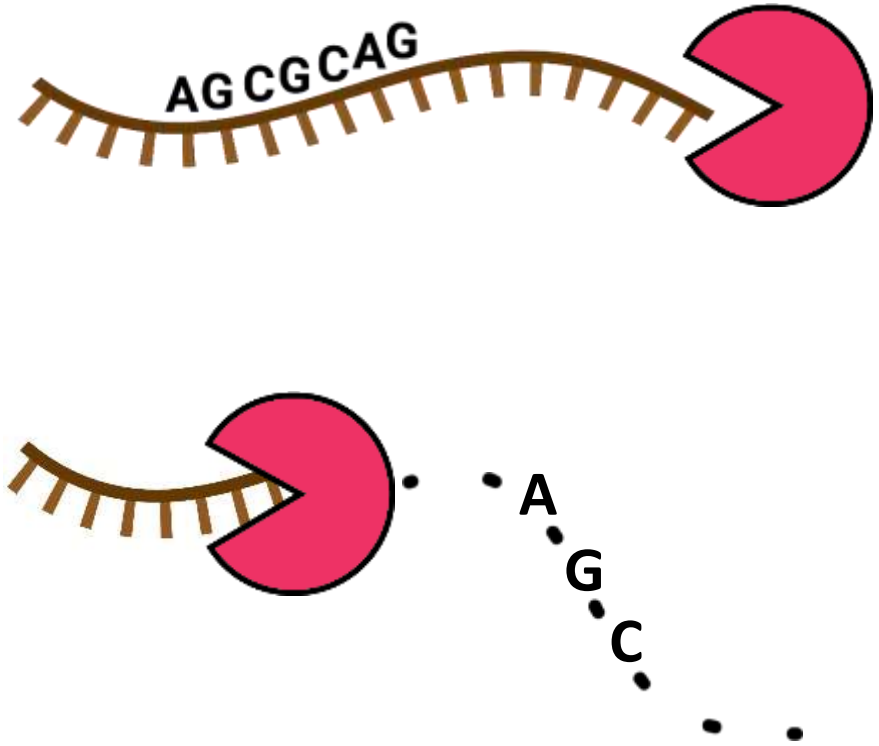
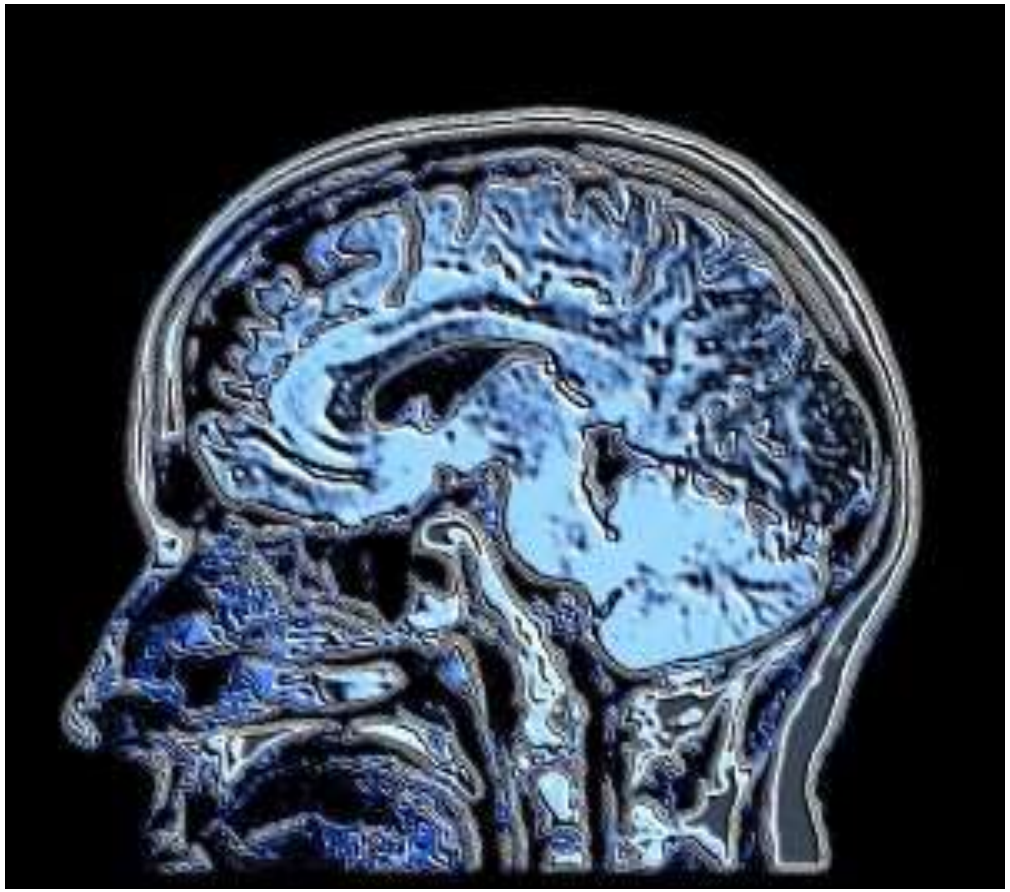
How RNAs are degraded and how that impacts human disease and possible therapies.

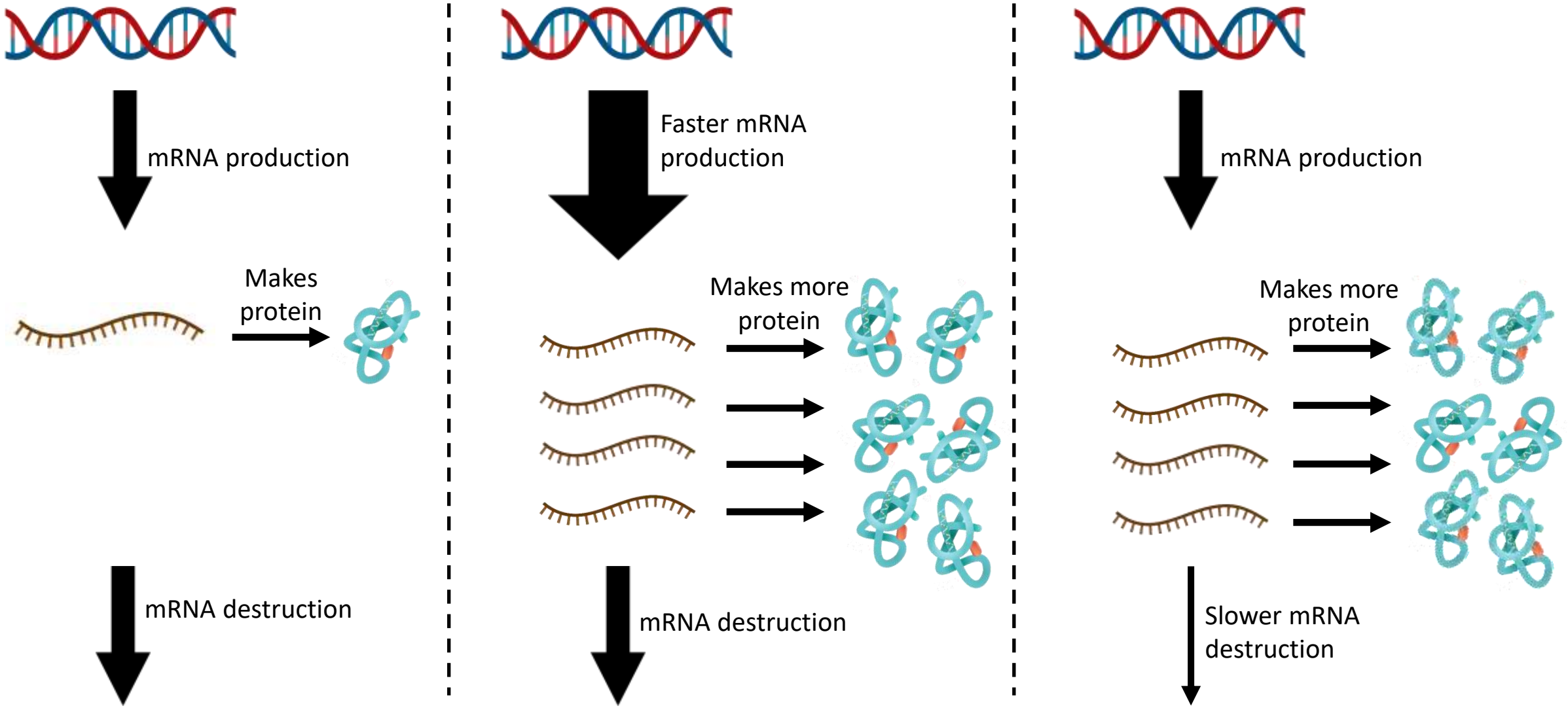


RNA biology and its connection to some neurodegenerative diseases



Founder and Consultant of Faze Medicines

Controlling Amount of Messenger RNAs Determines How Much Protein is Made

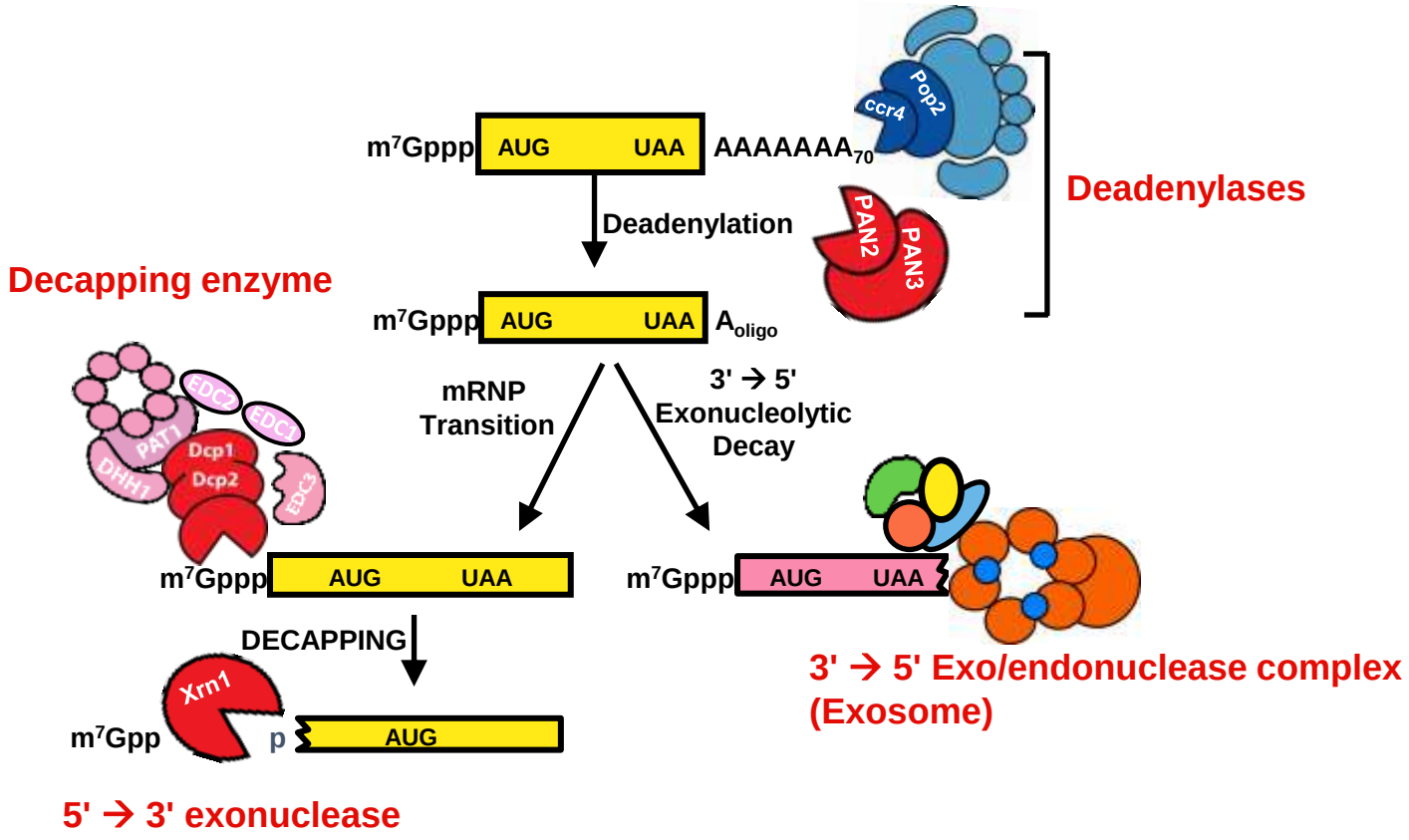


Key Question:

How do RNAs normally get degraded in cells?

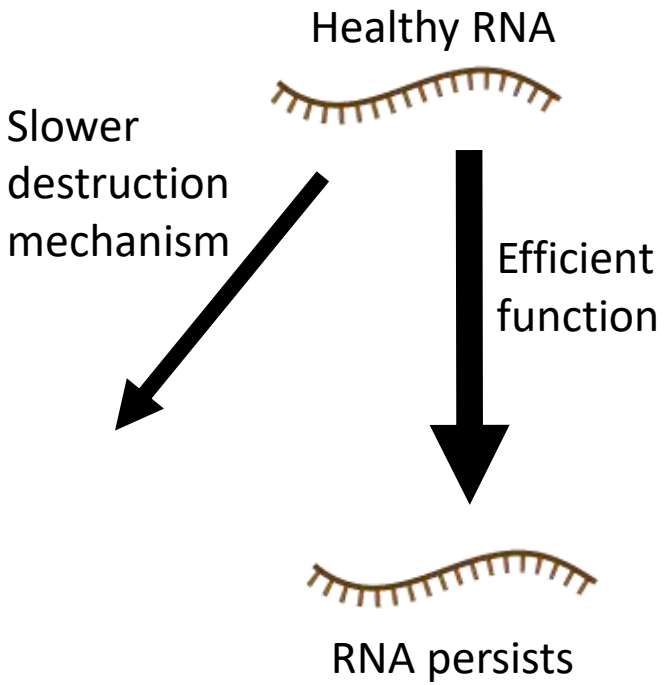
The Pathways, Nucleases and Regulators for mRNA Degradation

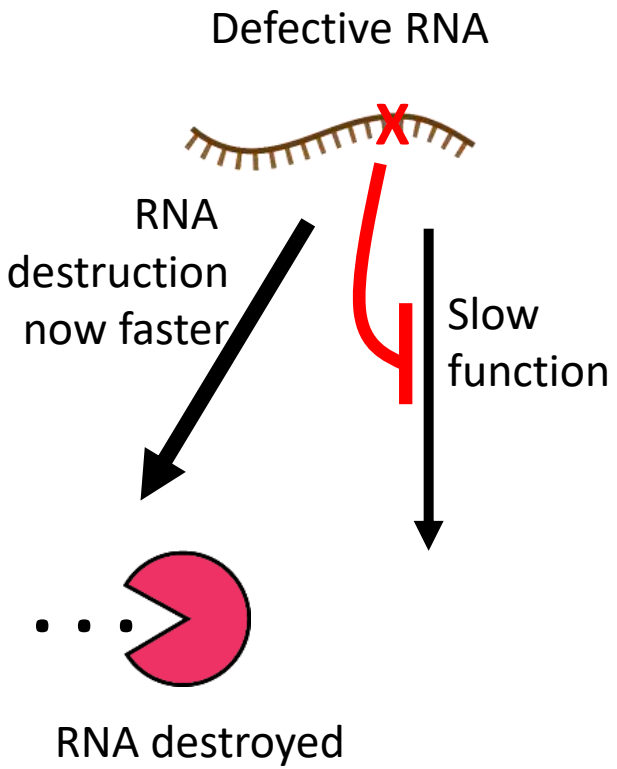
(100 people-years: One Cartoon)



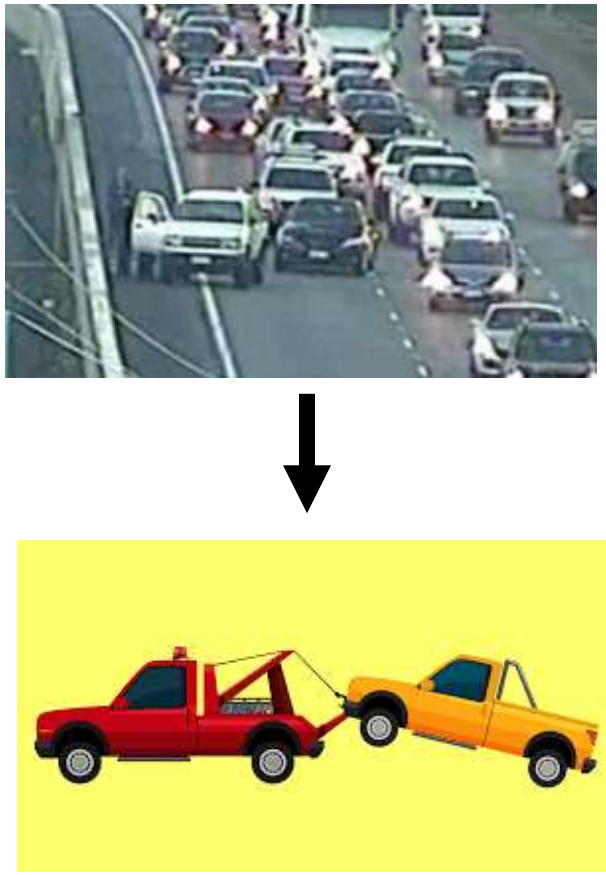
Understanding RNA Decay Revealed the Basis for Some Human Diseases

Principle #1: Cells clean up after themselves and degrade defective RNAs





Keeps cells functioning



Keeps traffic flowing

Consider a City with Unlimited Predatory Tow Trucks

30 seconds

Off to car crusher

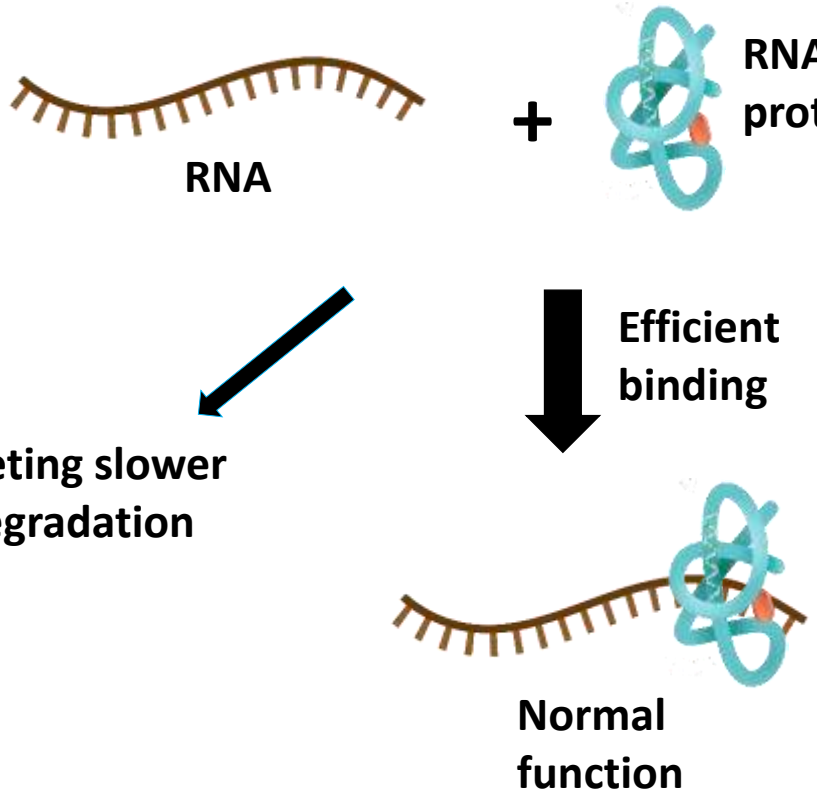
15 minutes

On the road again

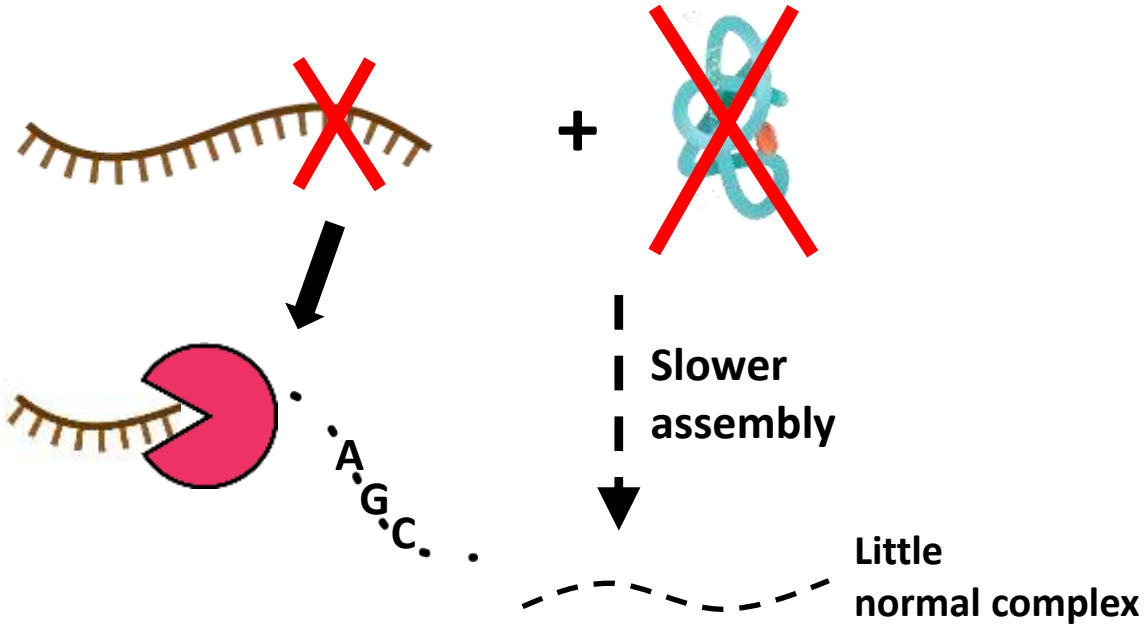
Leads to destruction of potentially functional machines

RNA Hyper-degradation Diseases: Defects in Protein Binding RNAs Leads to their Degradation and Subsequent Disease.

Normal RNA-Protein Interaction



RNA Hyper-degradation Disease



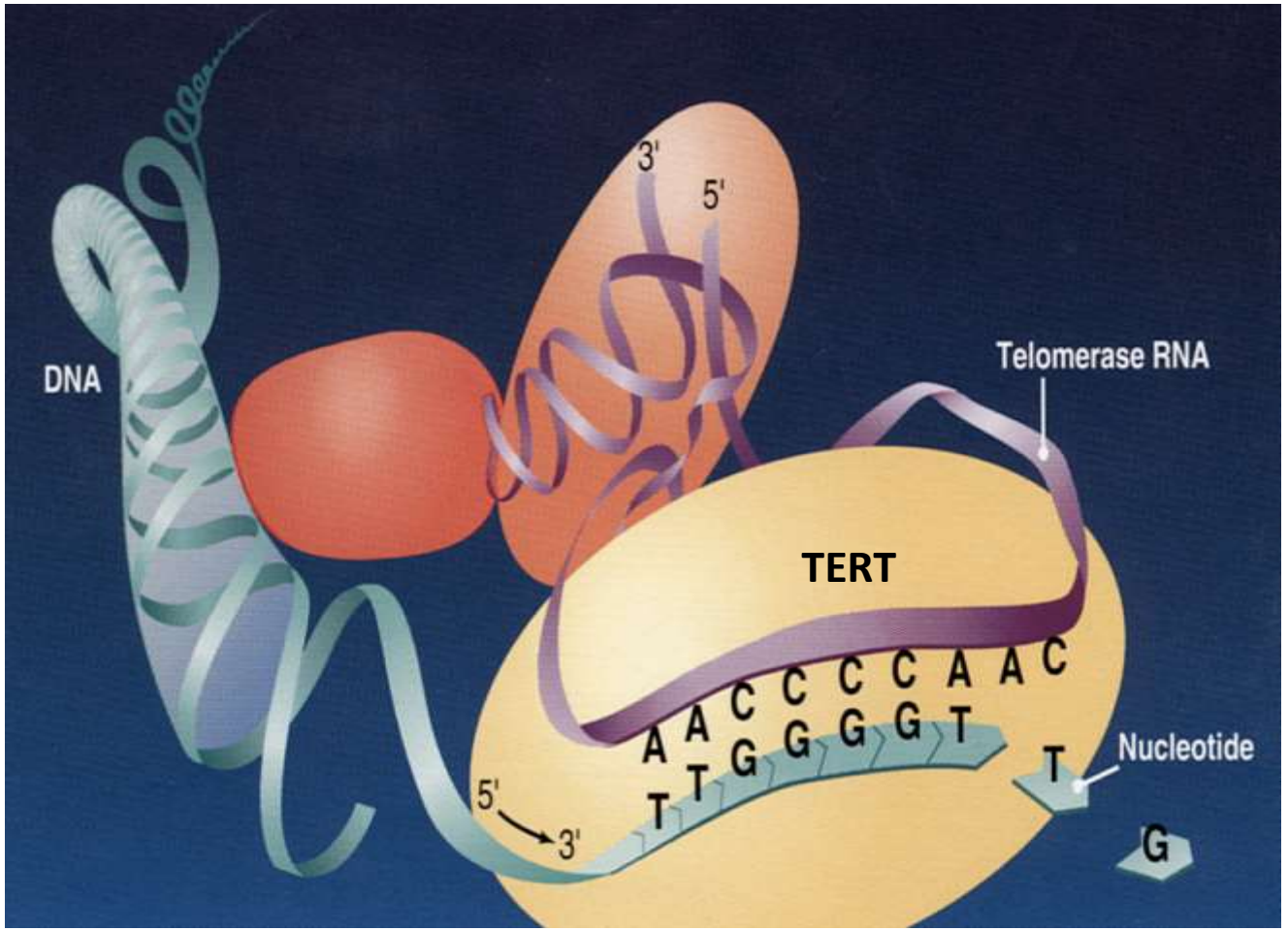
No function leads to disease

Some Examples:

Spinal Muscular Atrophy

Dyskeratosis congenita: Loss of telomerase RNA and/or telomerase function

Telomerase – Protein & RNA Machine that extends chromosome ends during cell division



Most important in cells that divide frequently. E.G.

- **Bone marrow –> produces all our blood cells**
- **Skin**

Patients have:

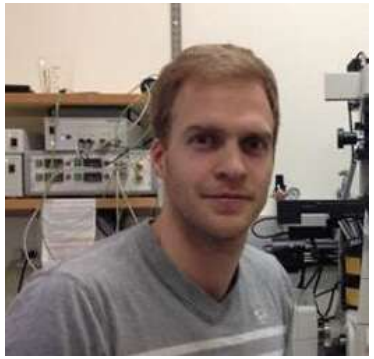
- **Bone marrow failure (usually early 20's)**
- **Skin disease and other issues**

No effective treatments

Sid Shukla

CU BIOCHEMISTY
PHD STUDENT

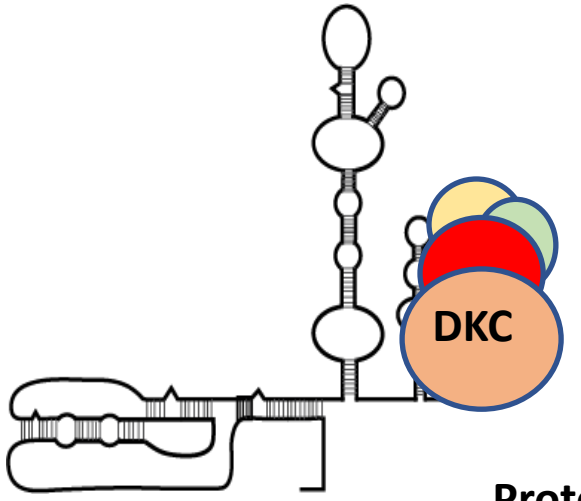
Sid's PhD: How is the telomerase RNA degraded?
If we block that degradation, can we treat the disease?



Jens Schmidt

Tom Cech

Healthy Individual



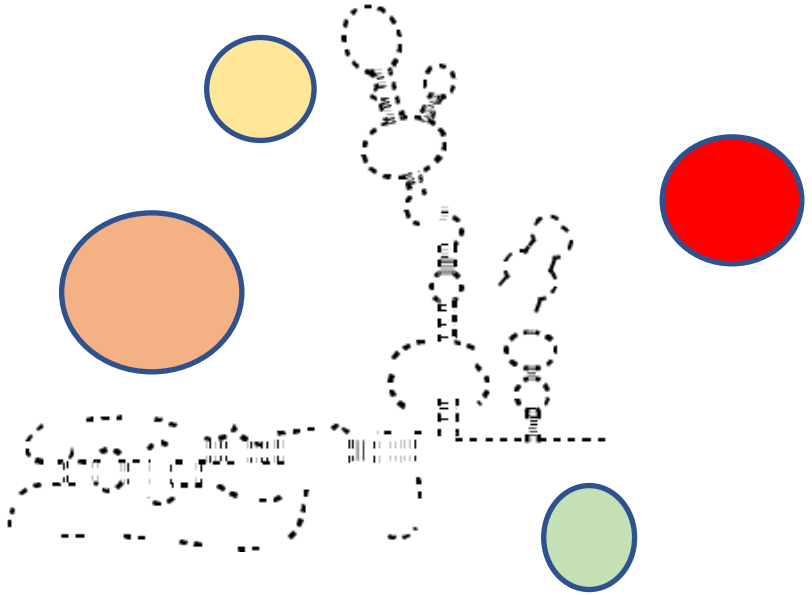
Telomerase RNA

Proteins bound To Telomerase RNA

NORMAL FUNCTION

?

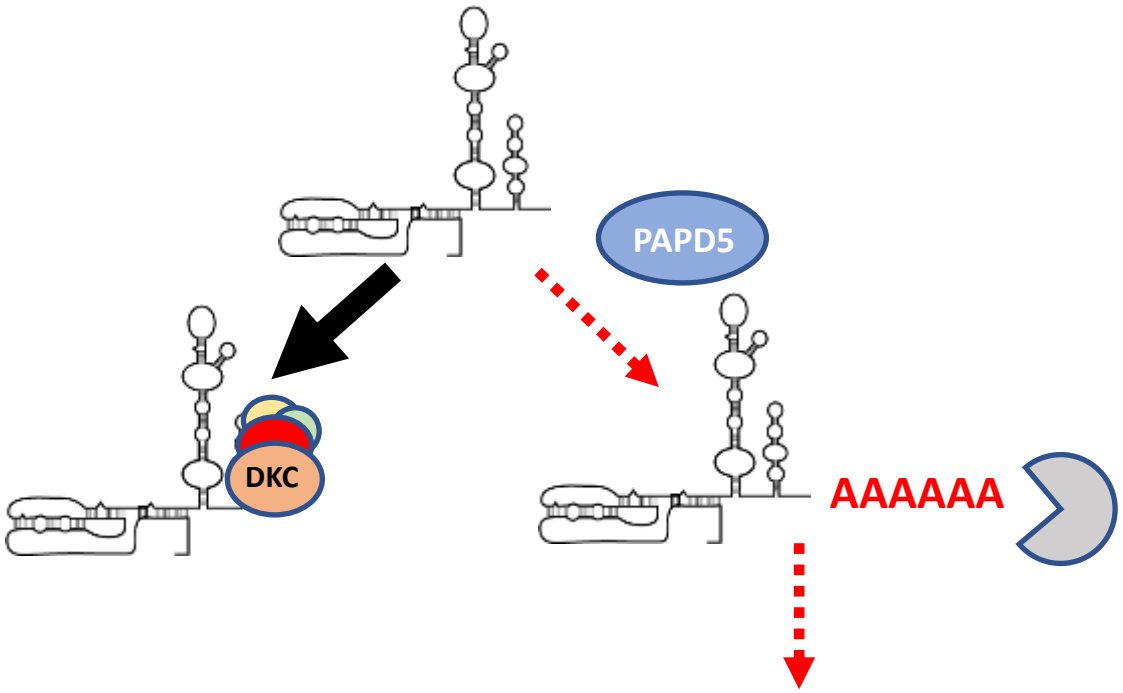
Dyskeratosis Congenita Individual



DISEASE

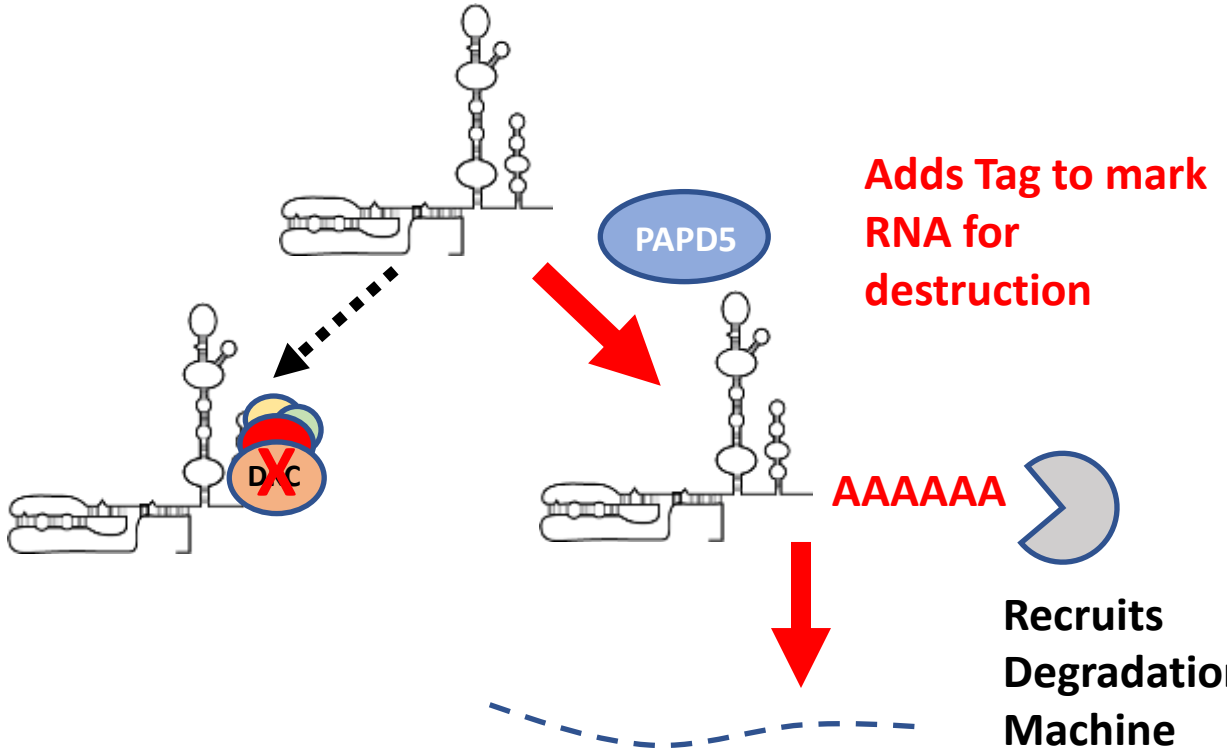
Dyskeratosis Congenita is often Caused by Hyper-degradation of the Telomerase RNA

Normal People



Telomerase Complex is functional

Dysteratosis Congenita patients

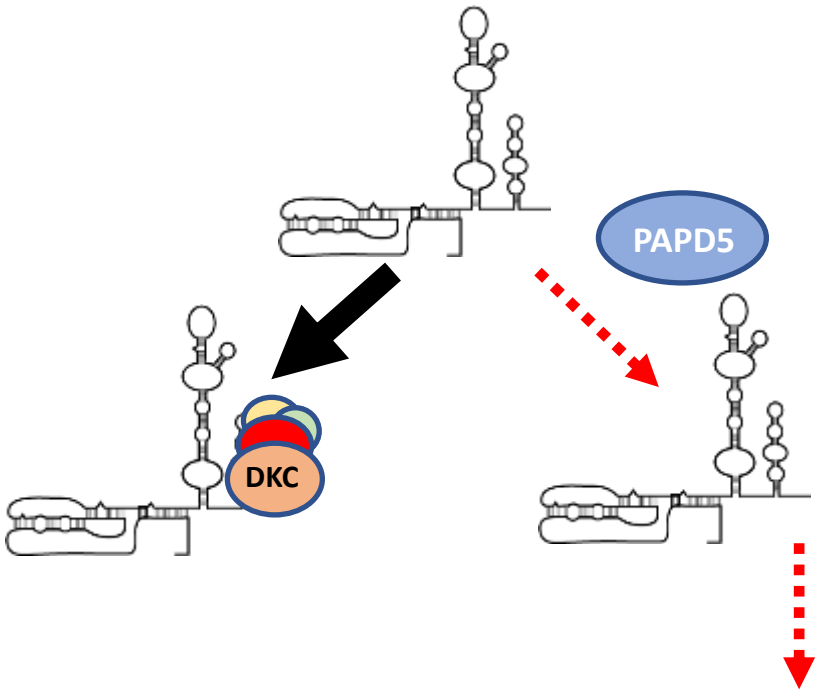


Telomerase RNA degraded:

- **Bone marrow failure**
- **Skin conditions**

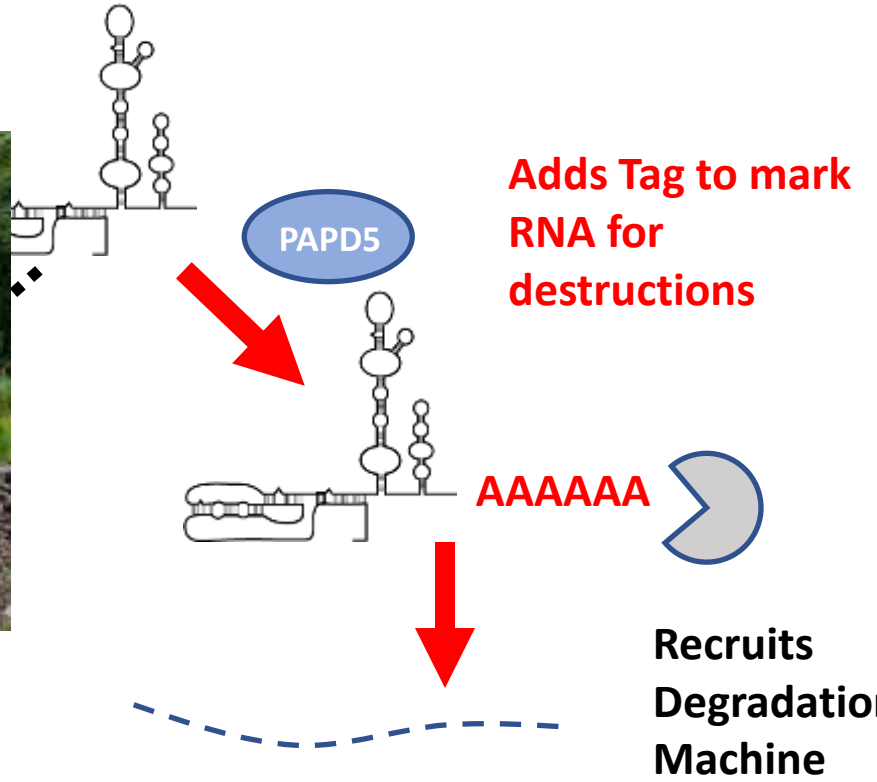
Dyskeratosis Congenita is often Caused by Hyper-degradation of the Telomerase RNA

Normal People



Telomerase Complex is functional

Dysteratosis Congenita patients

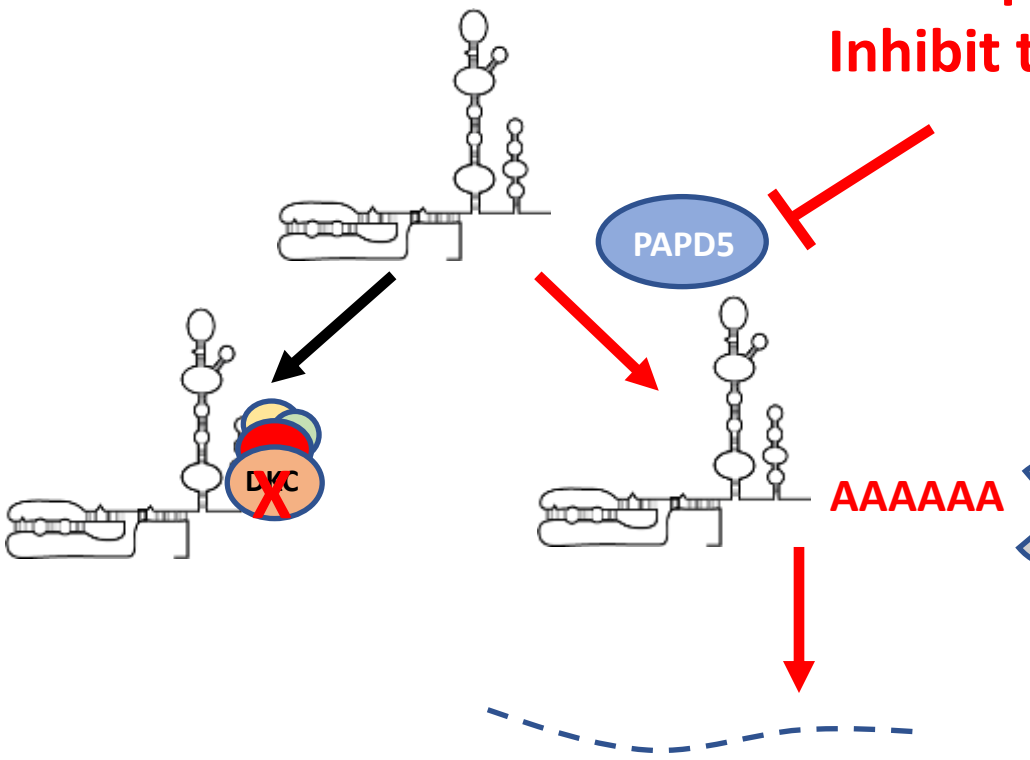


Telomerase RNA degraded:

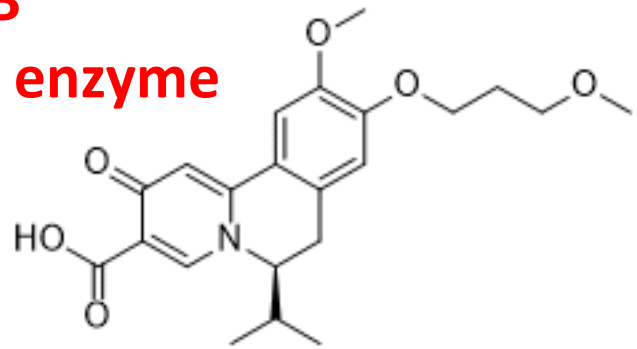
- **Bone marrow failure**
- **Skin conditions**

Could blocking telomerase RNA degradation rescue bone marrow failure?

Mutations in patients slow formation of proper complex



Drugs developed
For Hepatitis B
Inhibit tagging enzyme



And restores normal levels of telomerase RNA in cells from dyskeratosis patients

In the Lab, We Can Rescue Bone Marrow Function in Cells from Dyskeratosis Patients by Stopping Telomerase RNA Degradation

Human Embryo Stem Cells

Normal cells

(Differentiate in lab)

Production of normal levels of types of blood cells

Cells with mutation causing dyskeratosis congenita

(Differentiate in lab)

Greatly reduced production of blood cells

Add drug blocking telomerase RNA degradation

Cells with mutation causing dyskeratosis congenita

(Differentiate in lab)

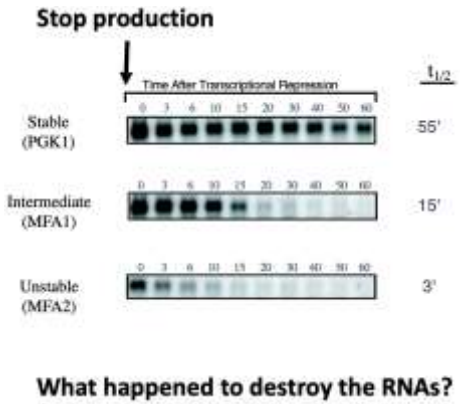
Production restored

Luis Batista, PhD
Washington University in St Louis

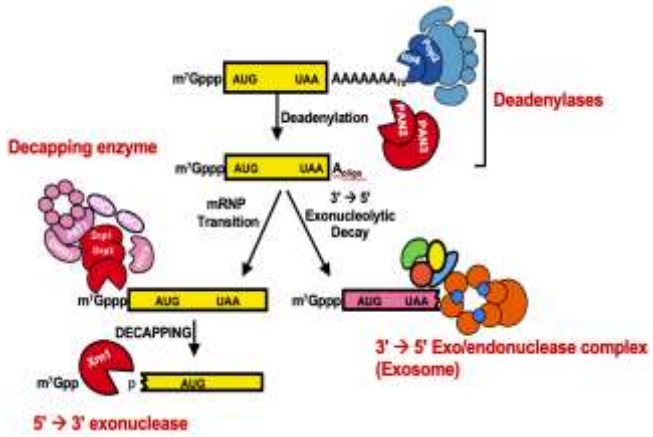
Similar rescue can be observed in mouse model (Agarwal lab)

Extending this work to other human diseases:

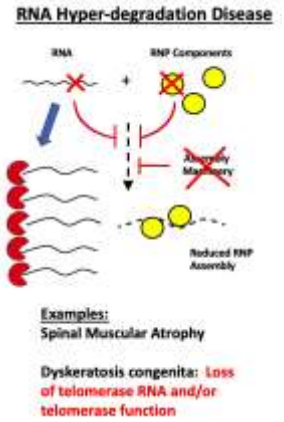
Question: how do RNAs get degraded



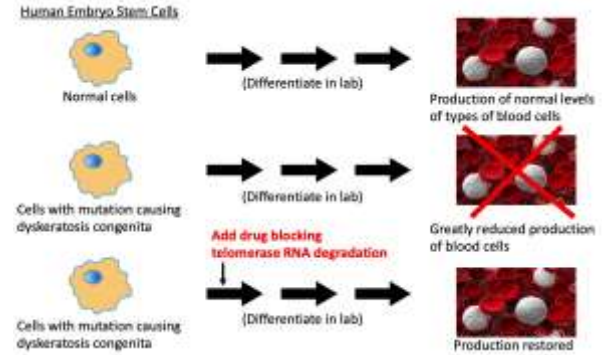
Understood mechanism of RNA degradation



Gained insight into some human disease



Developed therapeutic ideas



- **Continue to work on other RNA hyper-degradation diseases (Poikilodermia with neutropenia: hyper-degradation of miRNAs leading to bone marrow failure.)**

- **Now studying RNA hypo-degradation diseases (where RNA escapes degradation and accumulates to toxic levels), which may cause forms of muscular dystrophy, ALS**

A scientific mid-life crisis…….

RNA Degradation understood (to first level)

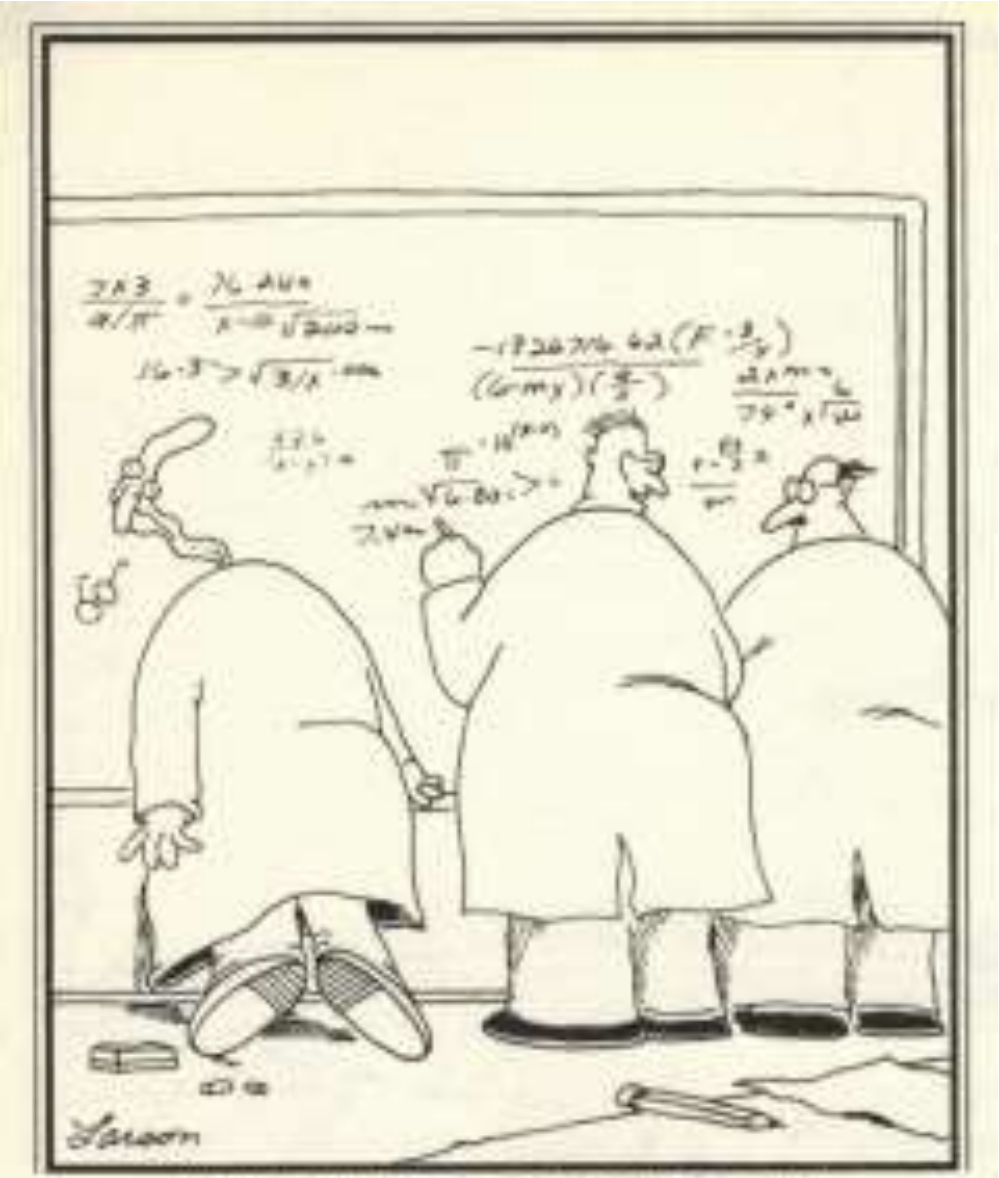
Getting Older……Shift my research to neurodegenerative disease

Why neurodegenerative diseases?

1) **Major societal issue…Getting worse.**

2) **Poorly understood biology.**

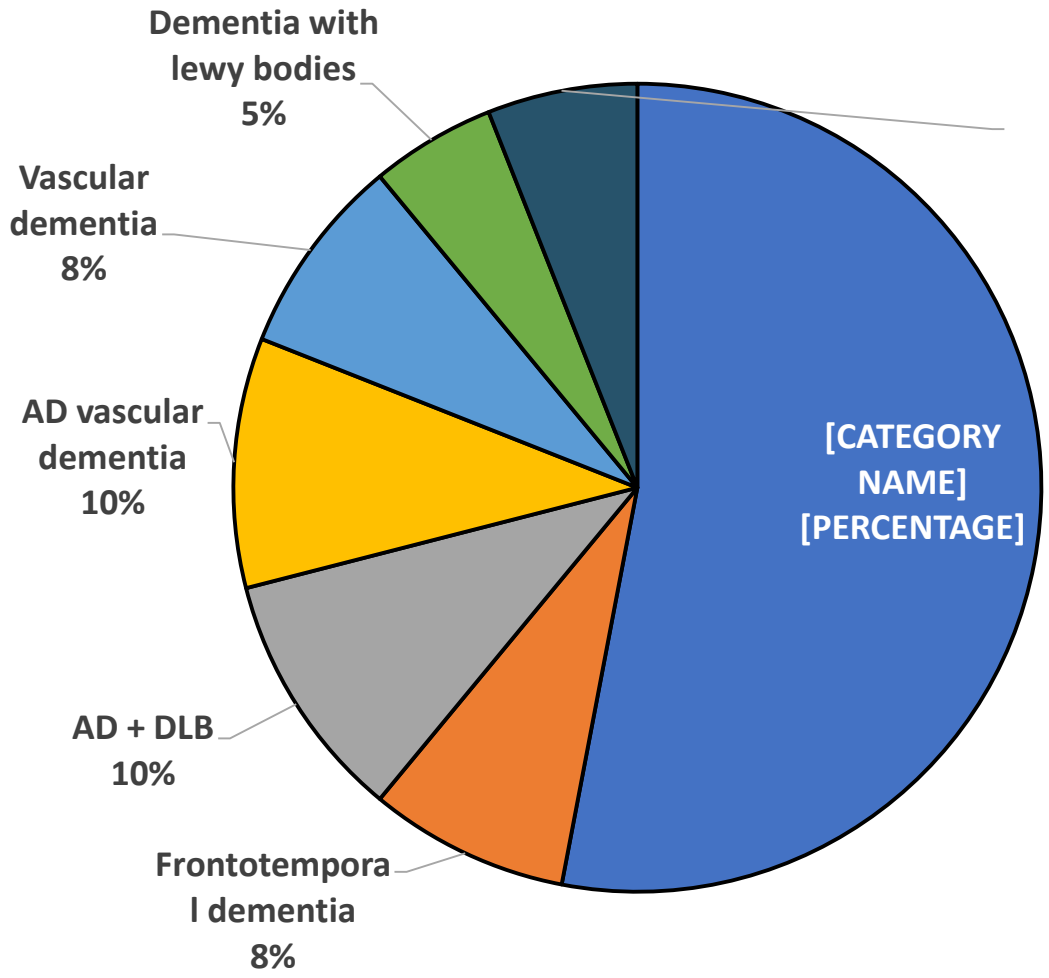
3) **Unstudied connection to RNA biology.**



"Look, Parker's blown his cerebral cortex on RNA degradation"

Neurodegeneration/Dementia is a big problem

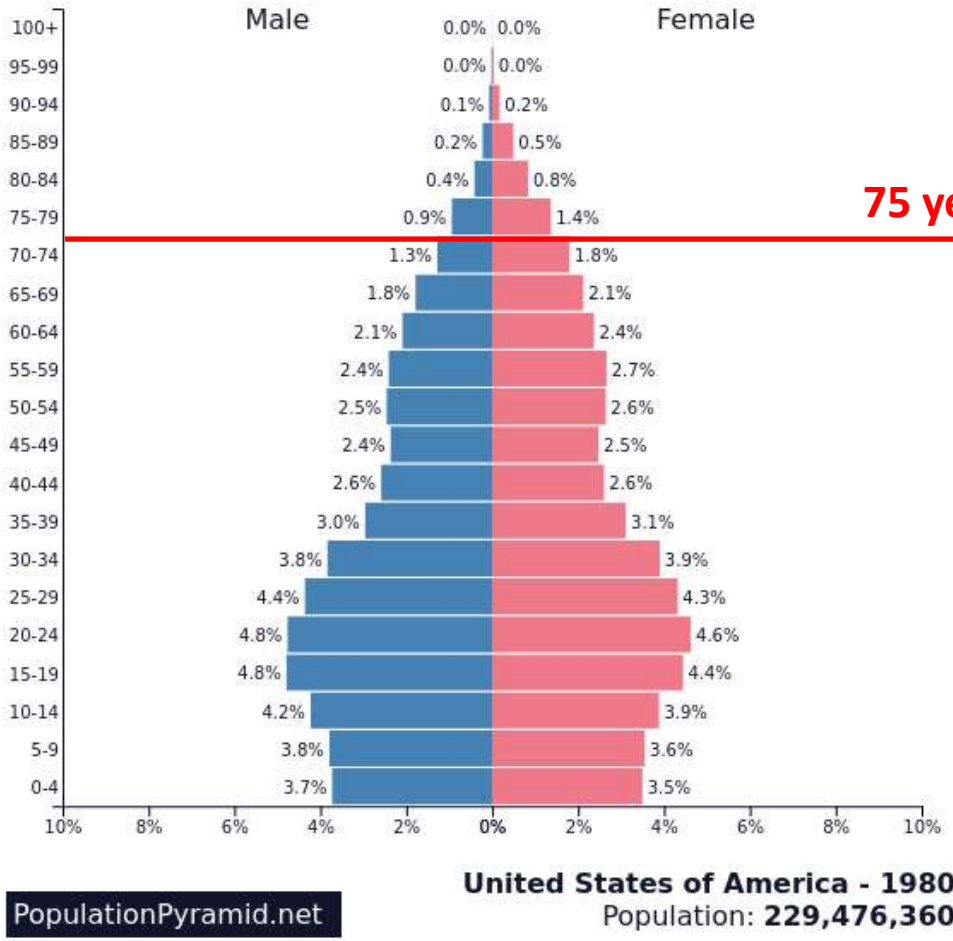
- Dementia refers to a set of symptoms, such as memory loss, difficulty completing familiar tasks, and disorientation
- Often caused by neurodegenerative diseases
- 6th leading cause of death (6.2 mil cases) (Just Alzheimer's disease)
- $355 billion in 2020 (could rise to $1.1 trillion by 2050)
- Large emotional impact on families & caregivers



(Petersen et. Al., 2016)

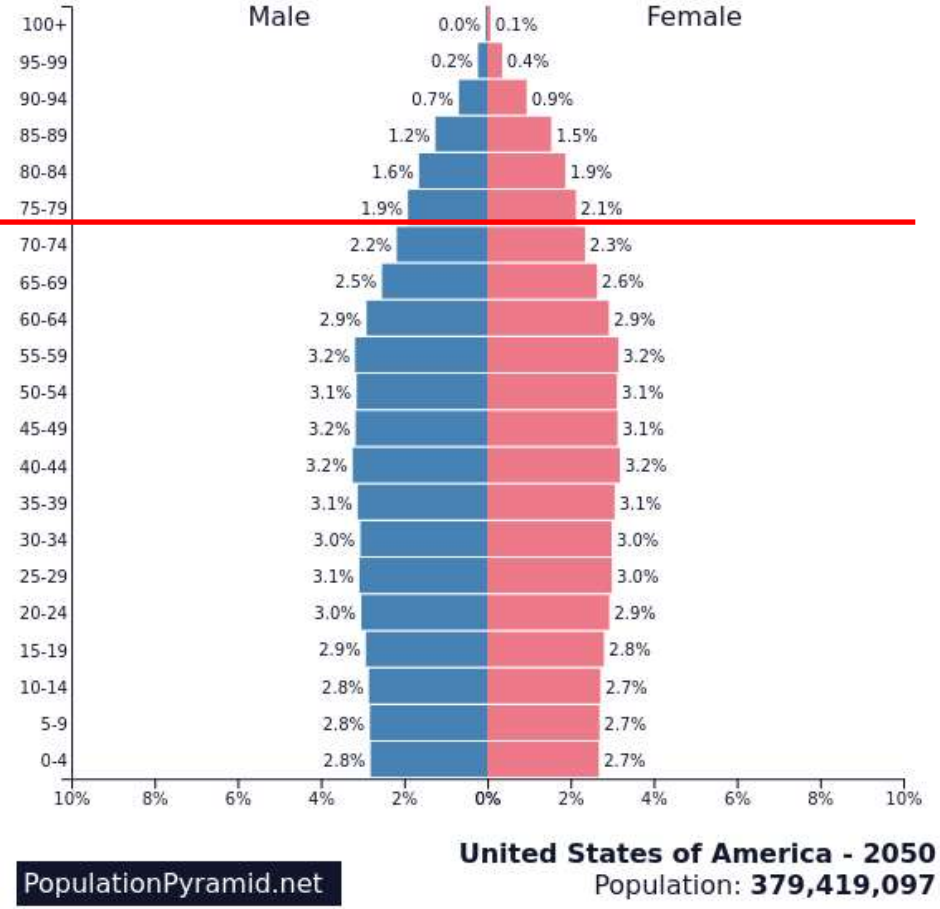
Number 1 Risk Factor for Neurodegenerative Diseases is Age.
And our population is getting older.

11.5% >65 years old
4% > 75 years old



75 years old

22.1% >65 years old
12.5% >75 years old



1980

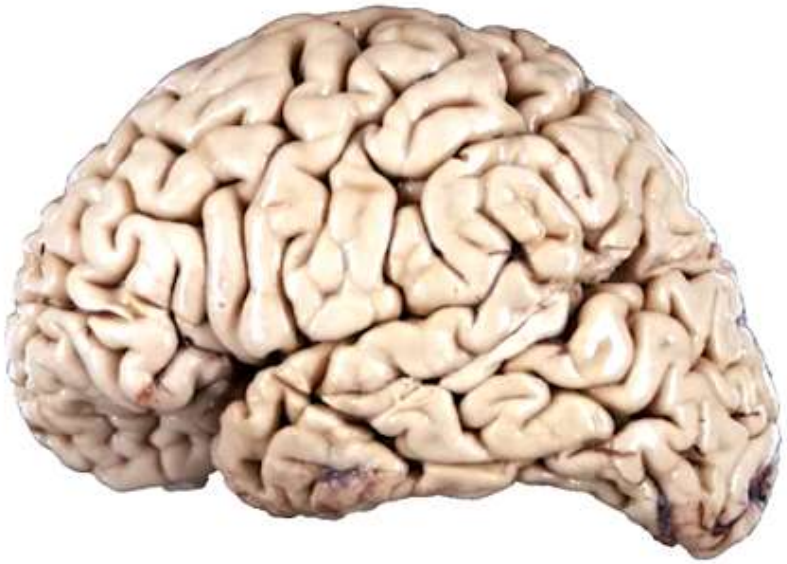
2050

Neurodegenerative diseases are complex.

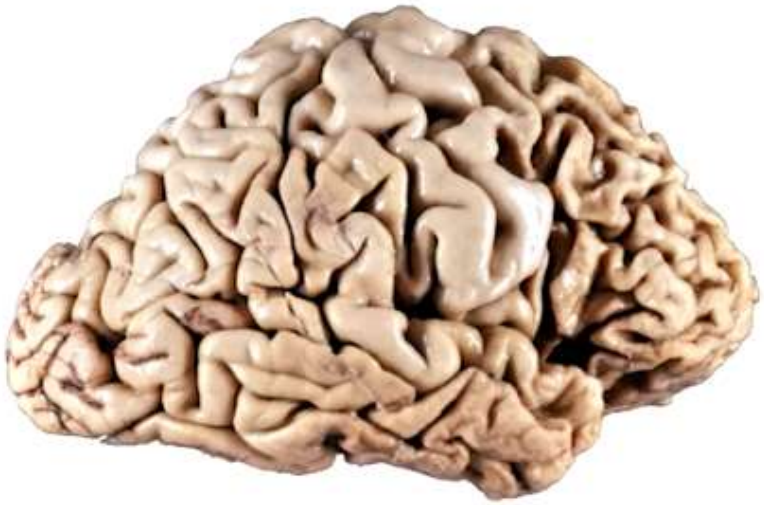
Affected by:

- **Neuroinflammation and the immune system.**
- **Interplay between different cell types in the brain.**
- **Cardiovascular Health.**
- **Genetic and Environmental inputs.**
- **……..**

Neurodegenerative diseases result in neuron death



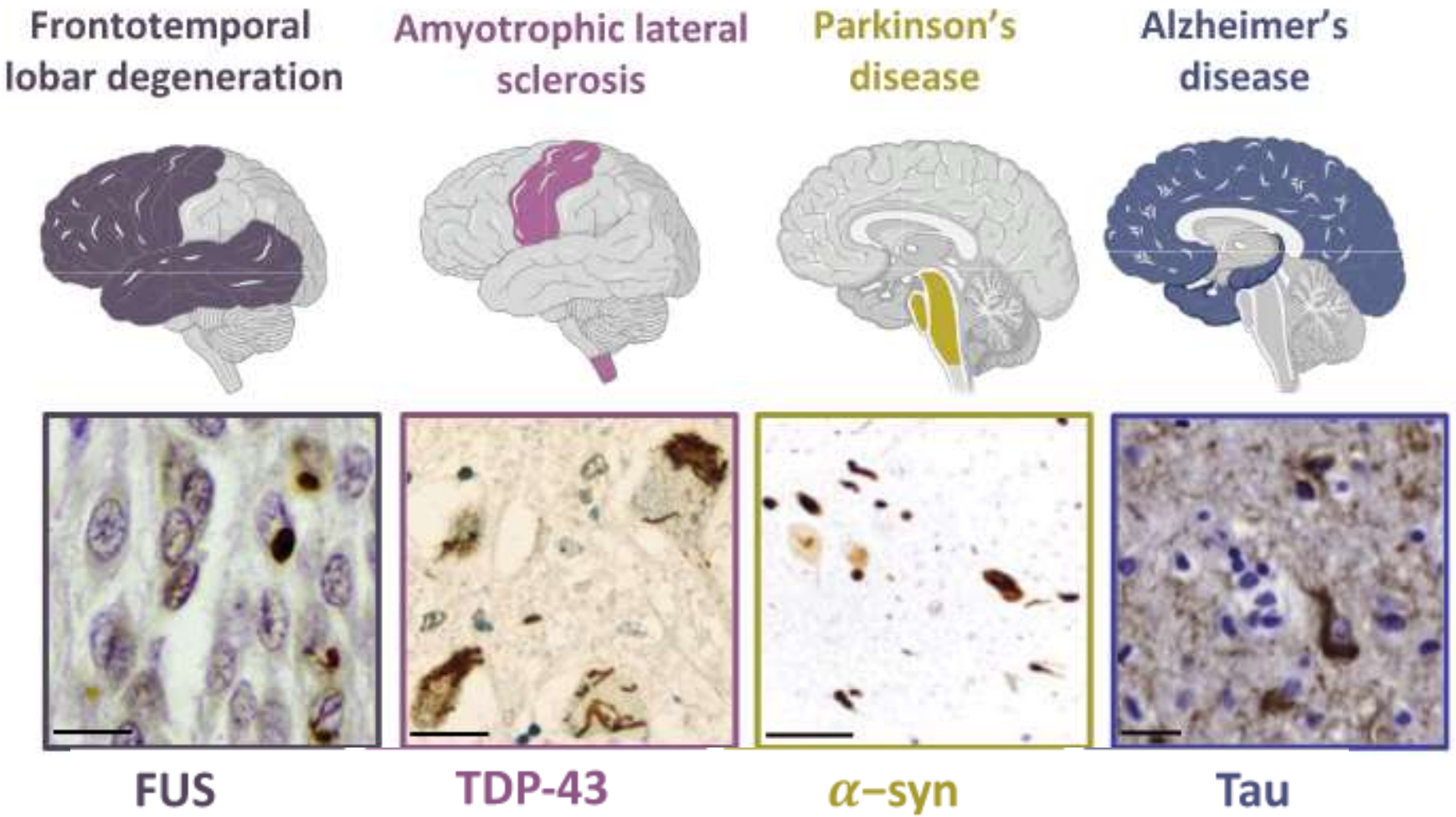
HEALTHY BRAIN



BRAIN WITH A
NEURODEGENERATIVE DISEASE

Humans essentially do not replace brain neurons.
Lose enough neurons and brain function is compromised.

Neurons die in neurodegenerative diseases due to the build up of aberrant protein aggregates

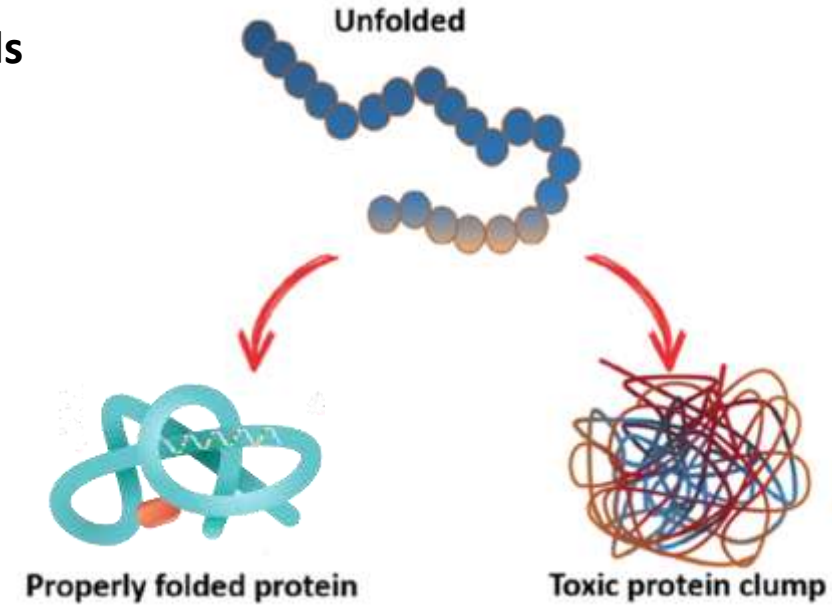


Different diseases have different proteins forming aggregates and affect different regions of the brain.

(But often co-pathology: e.g. Tau aggregates with α-synuclein disease)

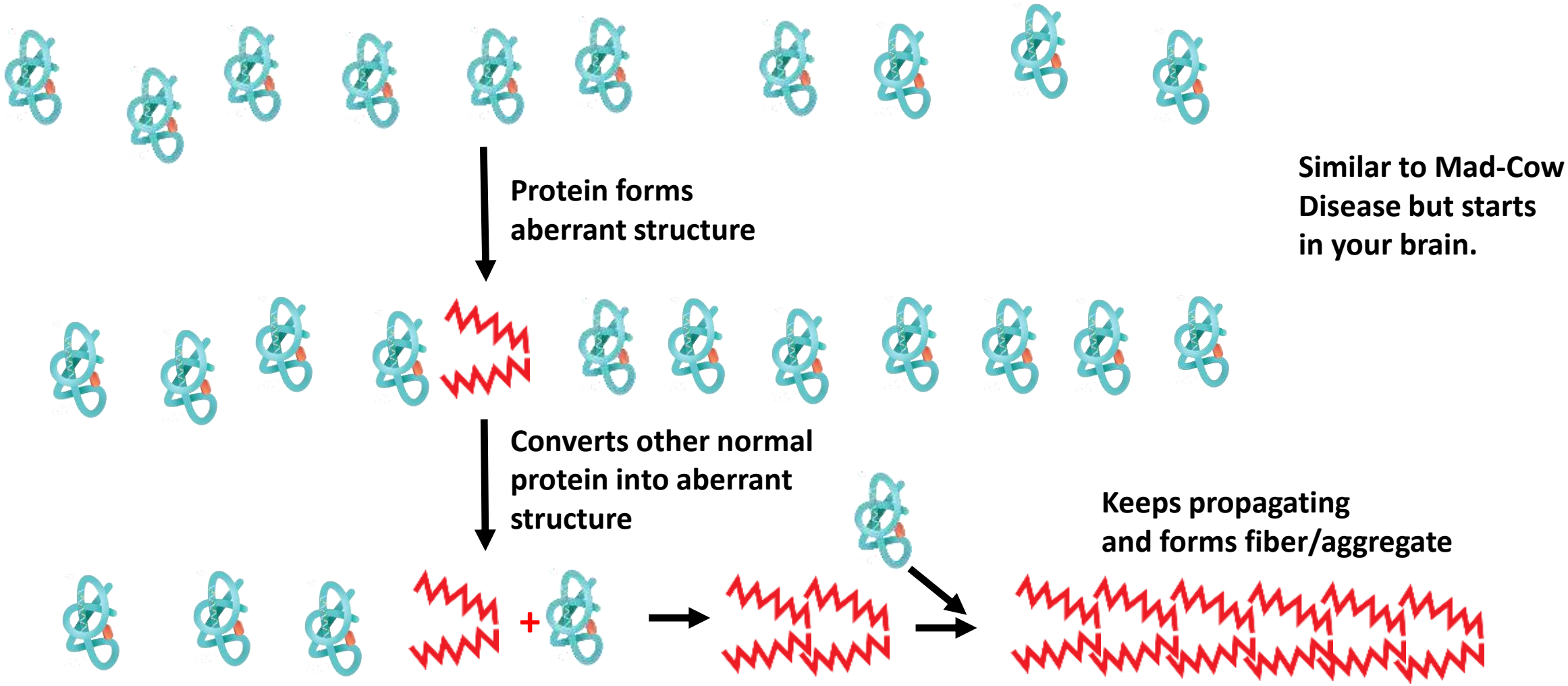
Neurodegenerative diseases are a class of protein folding diseases

Proteins are strings of amino acids that fold up into a structure that allows them to perform chemical reactions.



Of misfolded proteins

Misfolded protein aggregates involved in neurodegeneration can spread within brains by a "prion" mechanism



"Prion"—like mechanisms can spread toxic ideas in human cultures.

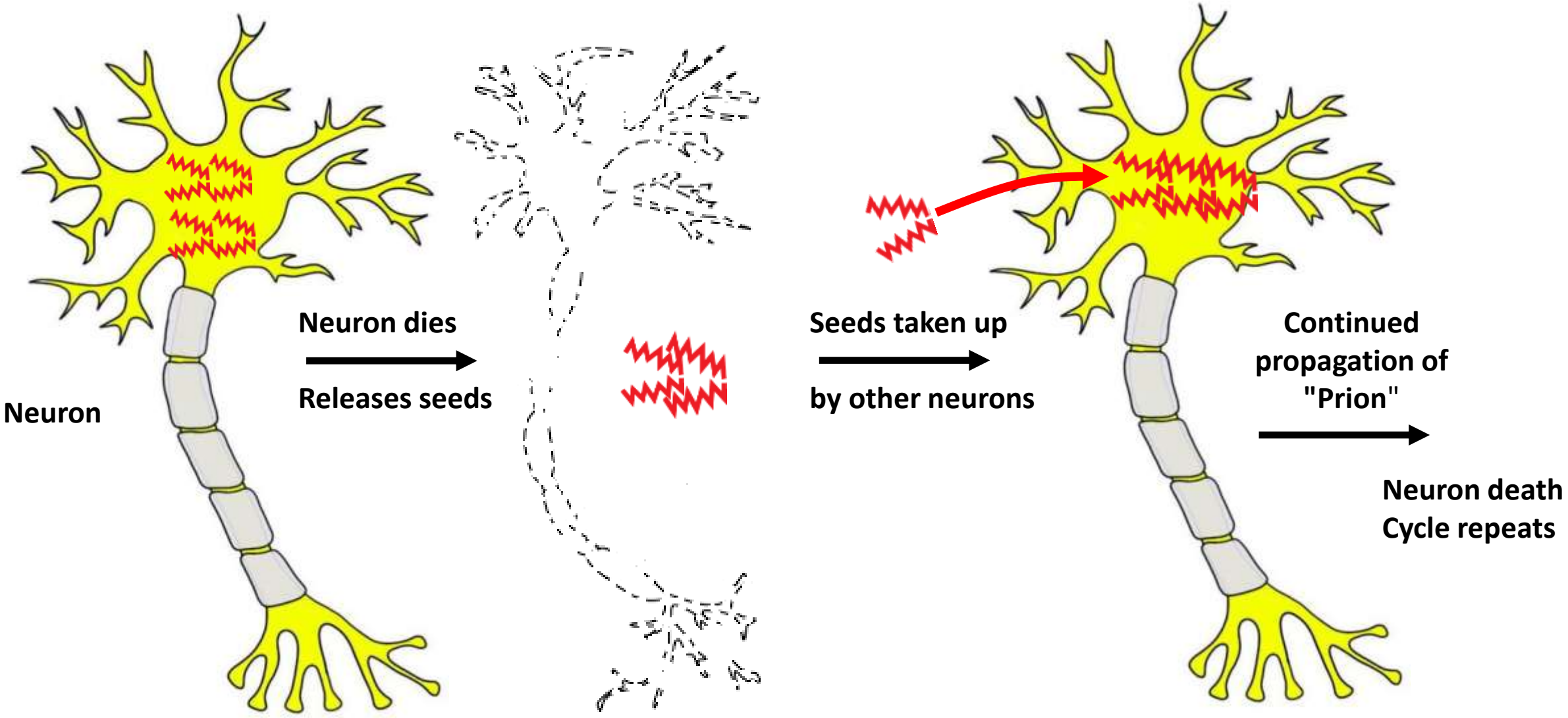
Politician enters abnormal state

Triggers other politicians to enter abnormal state

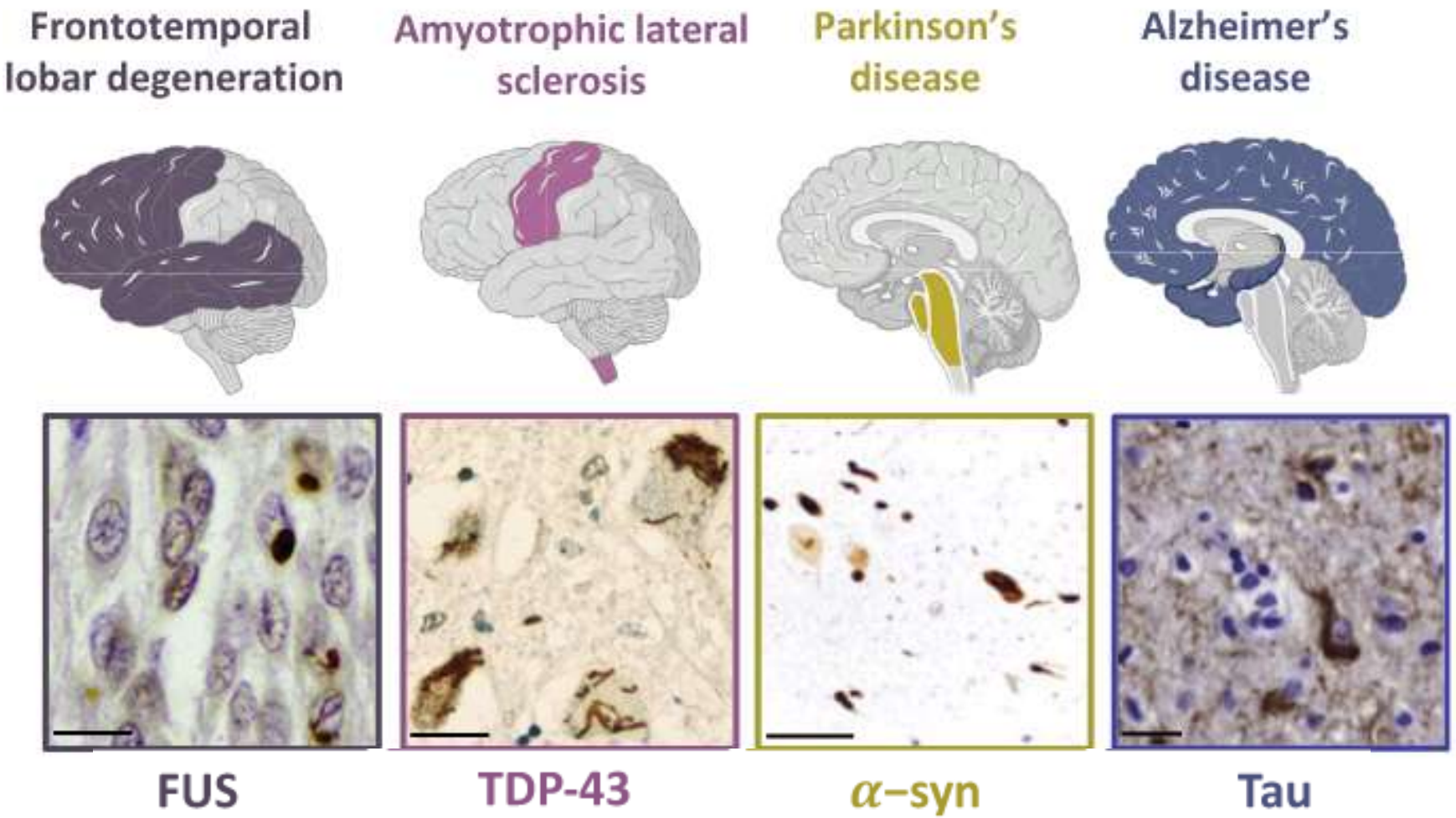
Keeps propagating

Damage Occurs.

Prions can spread between neurons

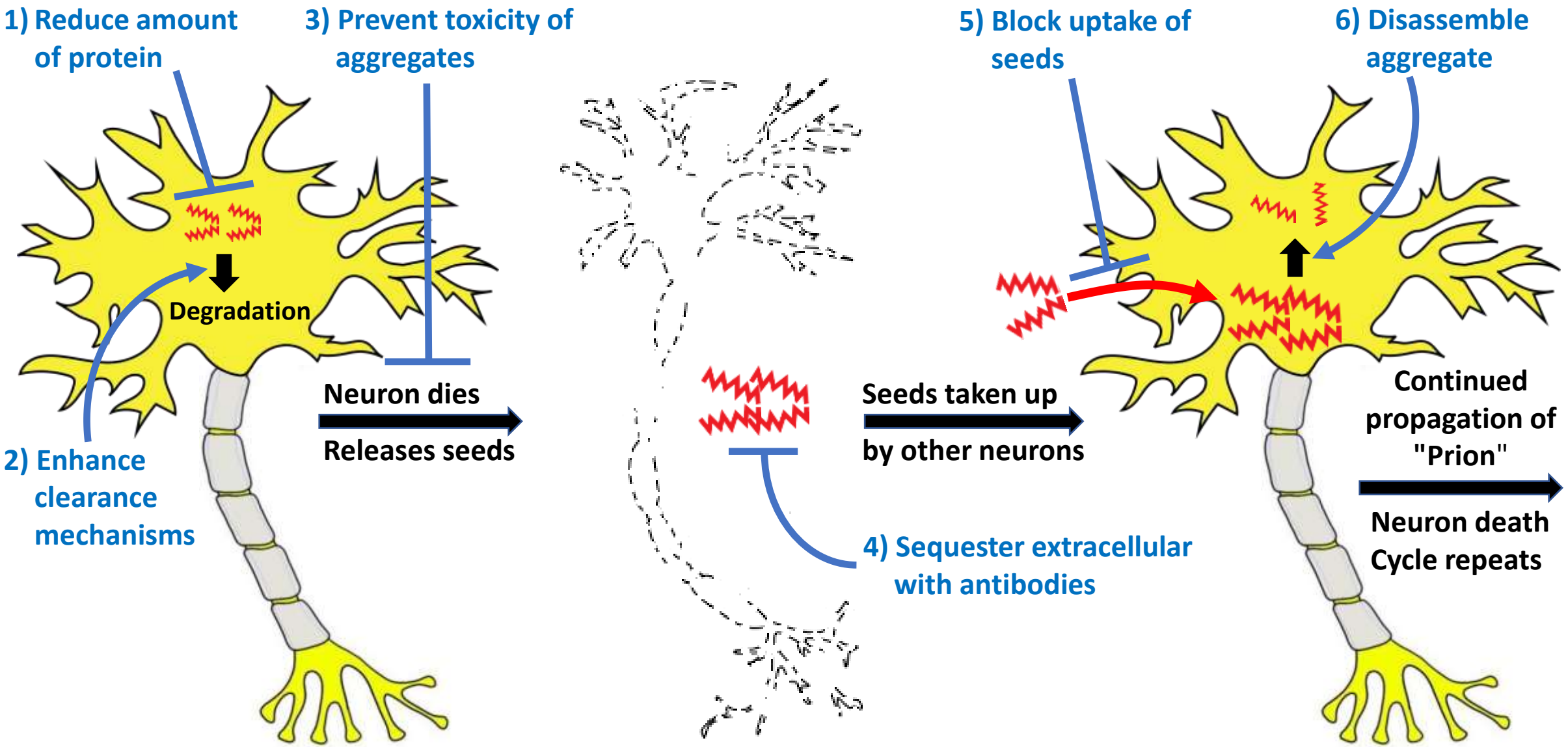


All of these proteins/diseases can spread by prion-like mechanisms



Spreading mechanisms are critical to neurodegenerative diseases.

Possible interventions to limit “prion-like” spread



Possible interventions to limit “prion-like” spread

1) Reduce amount of protein

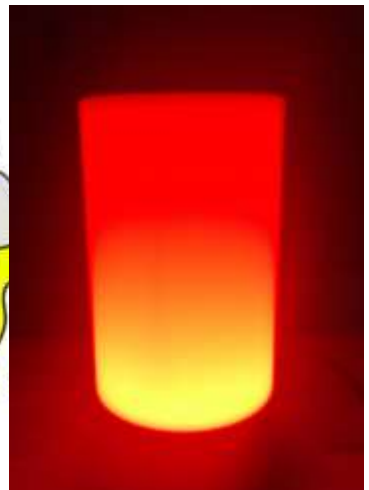
3) Prevent toxicity of aggregates

Degradation

2) Enhance clearance mechanisms

Neuron dies

Releases seeds



GENUS THERAPY
(Li-Huei Tsai)
Light/sound induced Gamma-Oscillations reduce protein aggregates in mice

4) Sequester extracellular with antibodies

Seeds taken up by other neurons

5) Block uptake of seeds

6) Disassemble aggregate

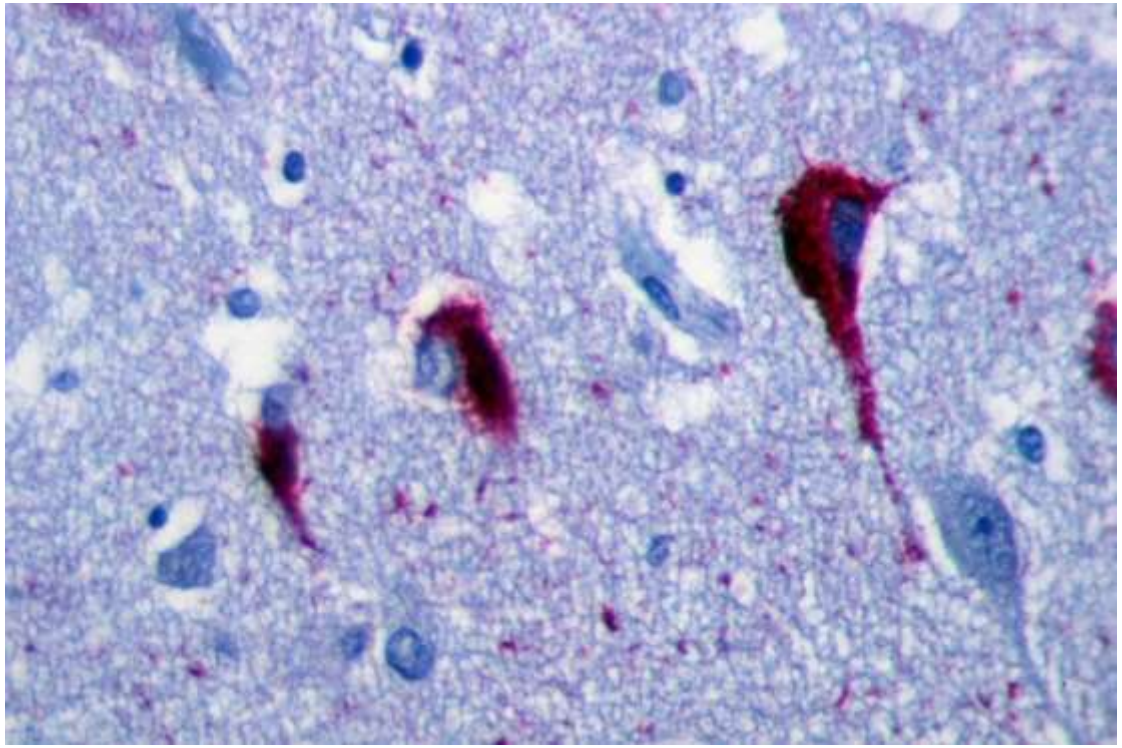
Continued propagation of "Prion"

Neuron death
Cycle repeats

Tau aggregates are central to >25 different neurodegenerative diseases and cause ~80% of cases of dementia (referred to as tauopathies)

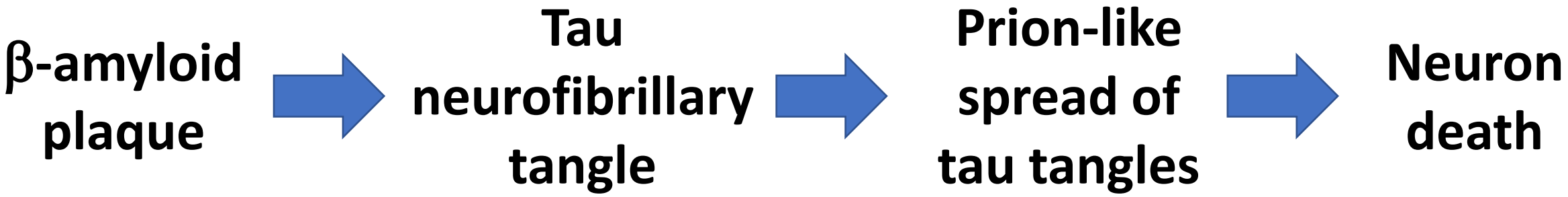
AB Plaques
(Alzheimer's Disease)

Repetitive Head Trauma
(Chronic traumatic encephalopathy: CTE)



Measles Infections (1/500-1000 infections)
(Subacute sclerosing Panencephalitis: SSPE)

b-amyloid (Abeta) and tau are involved in Alzheimer's disease progression



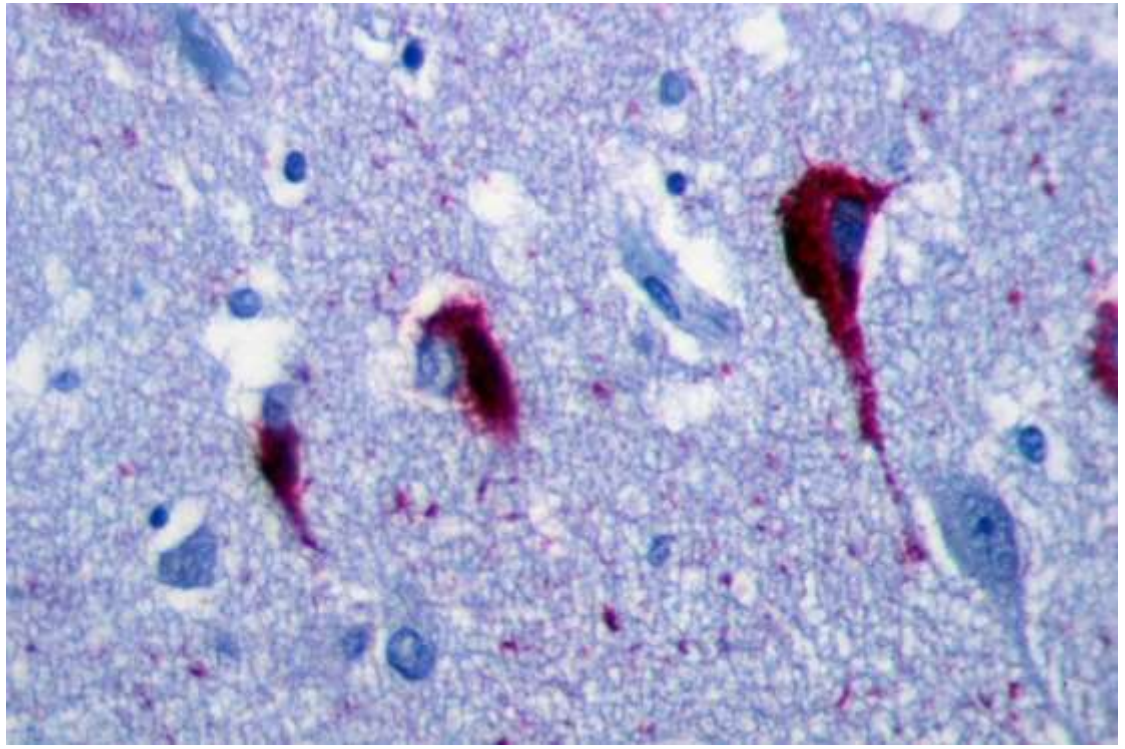
- Best current interpretation: β-amyloid plaques promote tau aggregates (also called tangles), which lead to neuronal death and Alzheimer's disease

- Key Question: How do β-amyloid plaques promote tau aggregates ?

Tau aggregates are central to multiple neurodegenerative diseases

Aβ Plaques
(Alzheimer's Disease)

Repetitive Head Trauma
(Chronic traumatic encephalopathy)



Biological Questions:

What are tau aggregates?
Where/How do Tau aggregates form?
What controls that process?
How do they kill neurons?

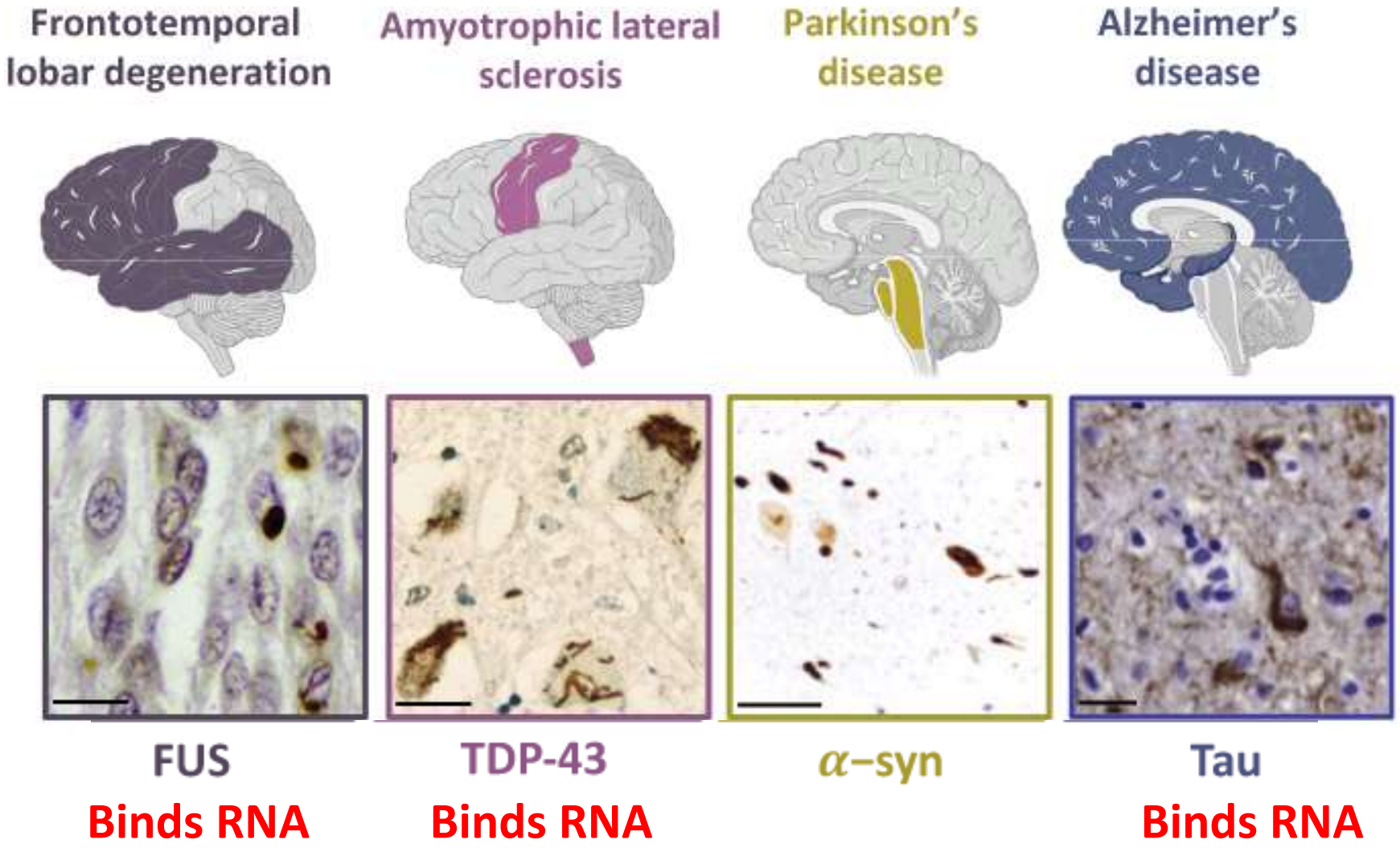
Clinical Questions:

Can one reduce the toxicity of Tau aggregates (or toxic species)?

Or their formation?

Or their spread?

Why my lab?



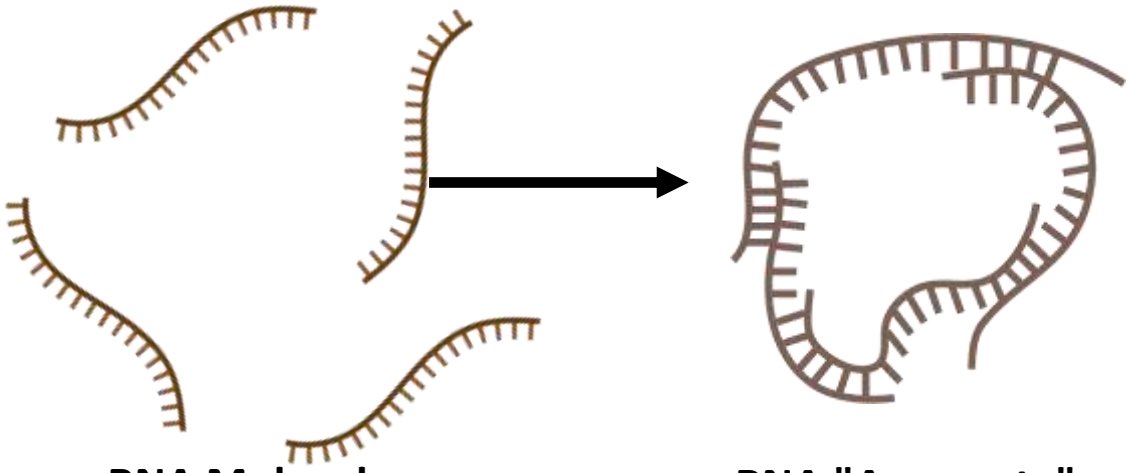
And unstudied connection to RNA biology and RNA-protein granules.

Could RNA Granules and ”aggregating” RNAs be involved in diseases of tau aggregation ?

Our Logic:

Many of the proteins that aggregate in neurodegenerative disease are RNA binding proteins.

RNA is also good at forming “aggregates”.



Evan Lester (MD/PhD student from Anschutz)

Evan’s PhD:

Are there RNAs in tau aggregates?

Could they explain some of toxicity?

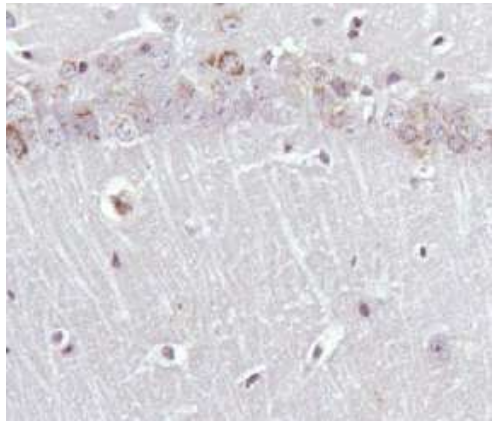
Multiple systems are used to study tau aggregation

Mouse Models of Disease

Normal Mouse

With Age

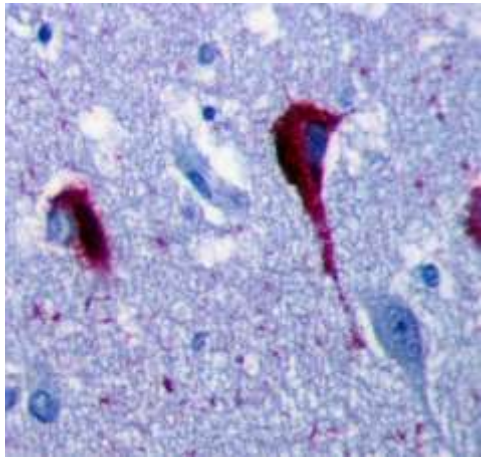
Mouse Brain stays healthy



Mouse genetically engineered to get tauopathy disease

With Age

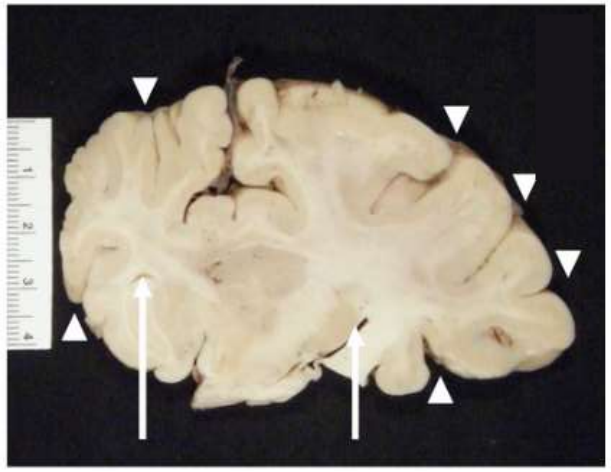
Mouse Brain develops tau aggregates



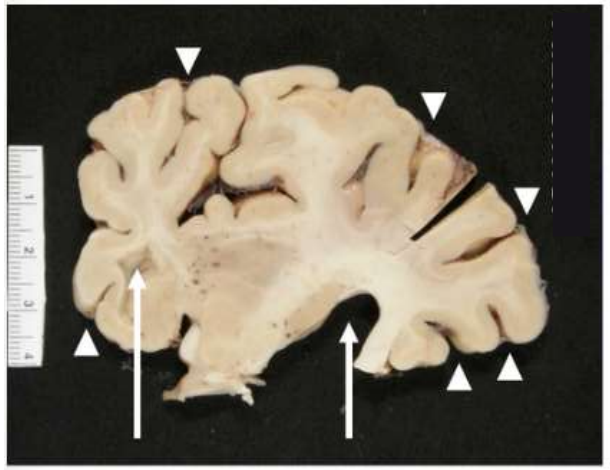
Verifying new insights in human tissue is critical

Post-mortem Samples from Patients

Normal



Alzheimer's Disease



Stan Prusiner
(UCSF)

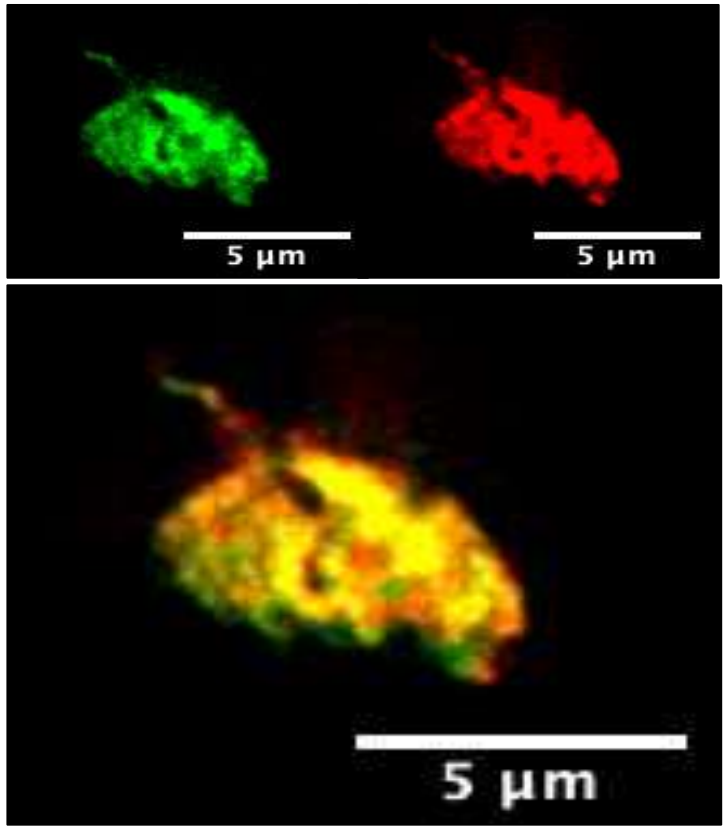
Nadine Bakkar
(Barrow Neurological Institute)

Is what we learn in animal models or cell lines occurring in human disease?

Evan learned:

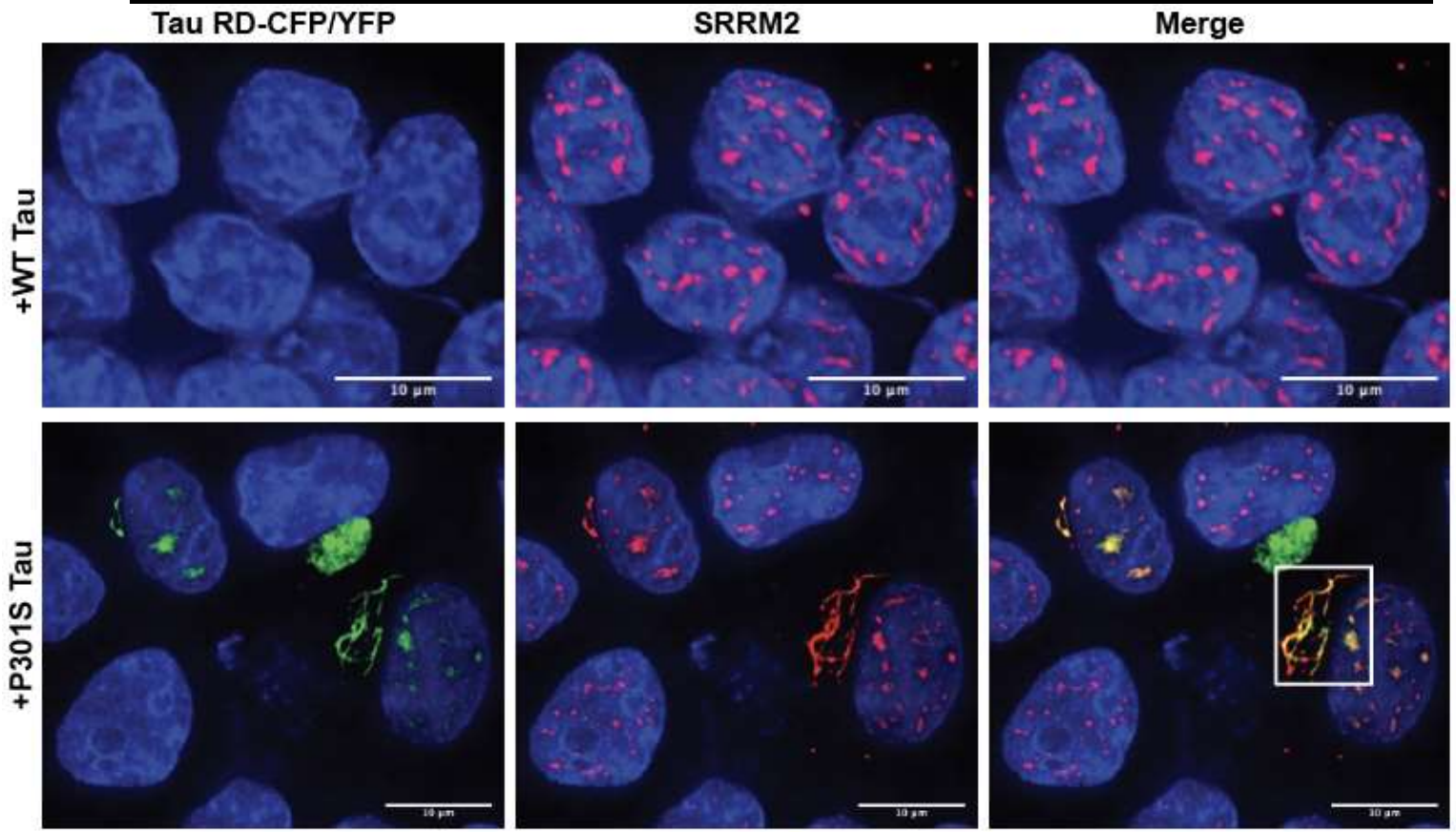
Tau aggregates contain RNA ……and RNA binding proteins (SRRM2)

Green = Tau Red = RNA



Tau Aggregate from mouse model

Green = Tau Red = SRRM2 Yellow = MERGE

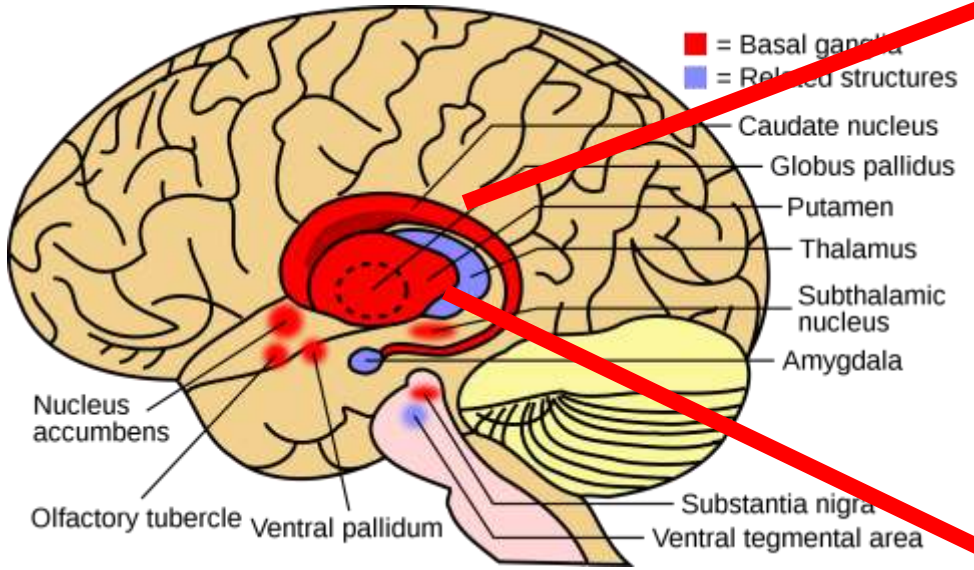


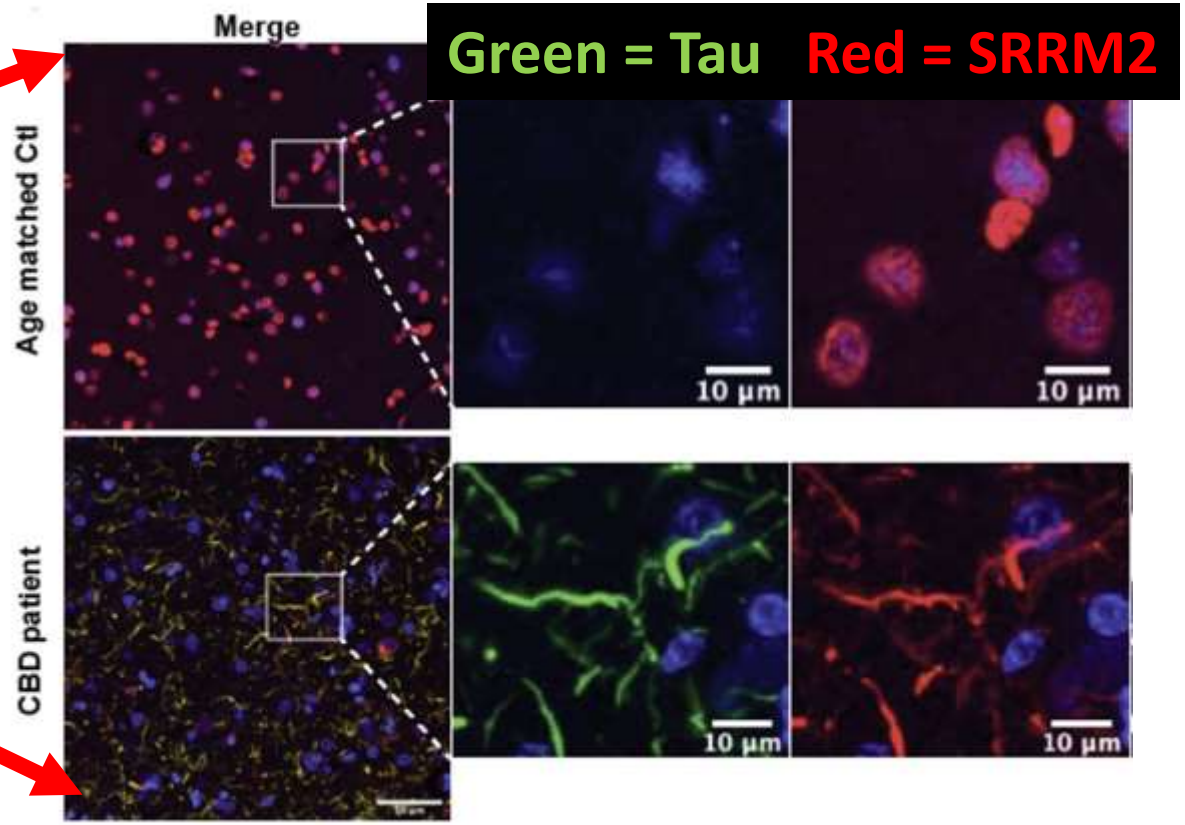
Tau Aggregate in cell line model

Most enriched in snRNAs and snoRNAs.

SRRM2 accumulates in tau aggregates in human disease.

Experiment: Examine location of SRRM2
Protein in normal and disease brain samples
From patients who had tauopathies



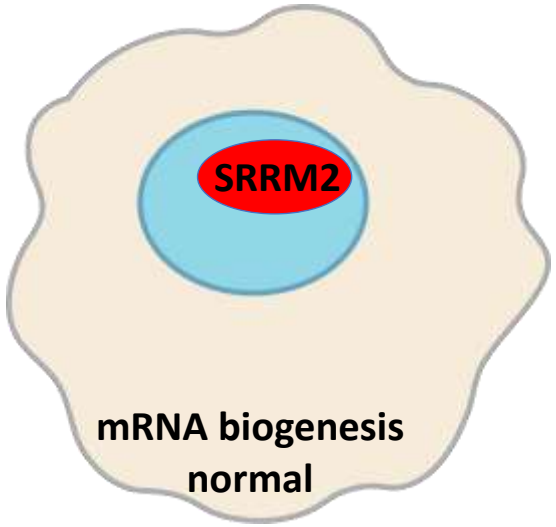


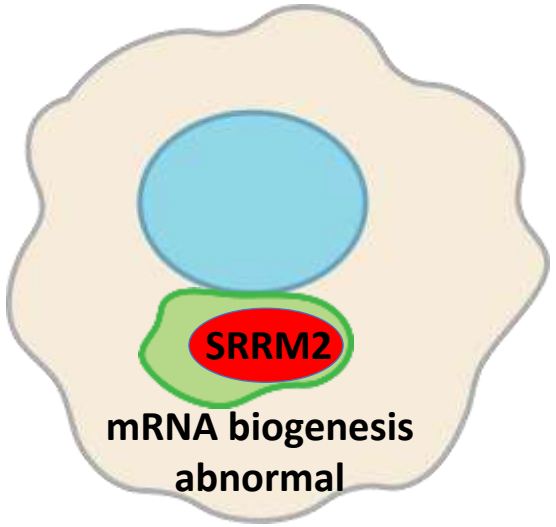
Patients with corticobasal degeneration
Develop a form of tauopathy

Similar results seen in brains from Alzheimer's patients

Why do we care that tau aggregates contain RNA and RNA binding proteins?

May contribute to mechanisms by which tau aggregates kill cells

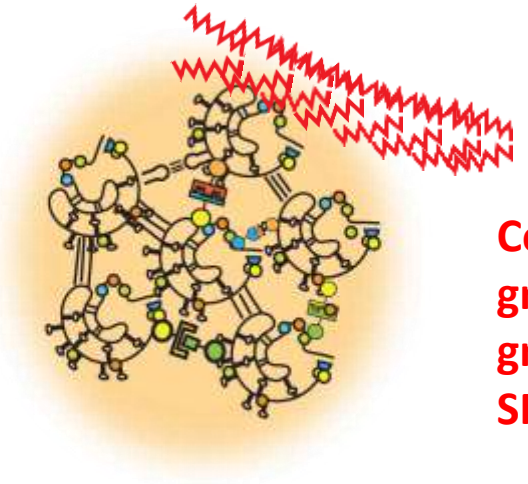




RNAs and RNA binding proteins trapped in tau aggregates are involved in mRNA biogenesis

Post-mortem analyses of patients with tau aggregates show related alterations in mRNA biogenesis

May reveal how tau aggregates grow within cells



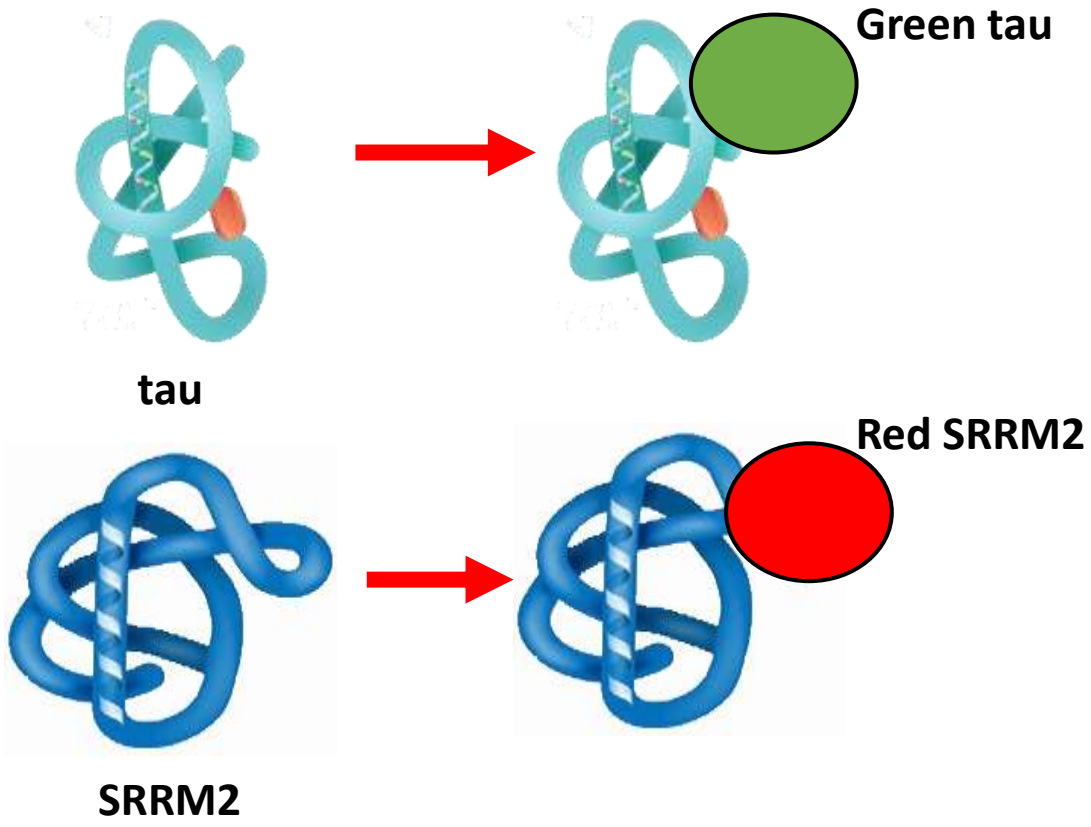
Could tau aggregates grow off RNA-protein granules that contain SRRM2?

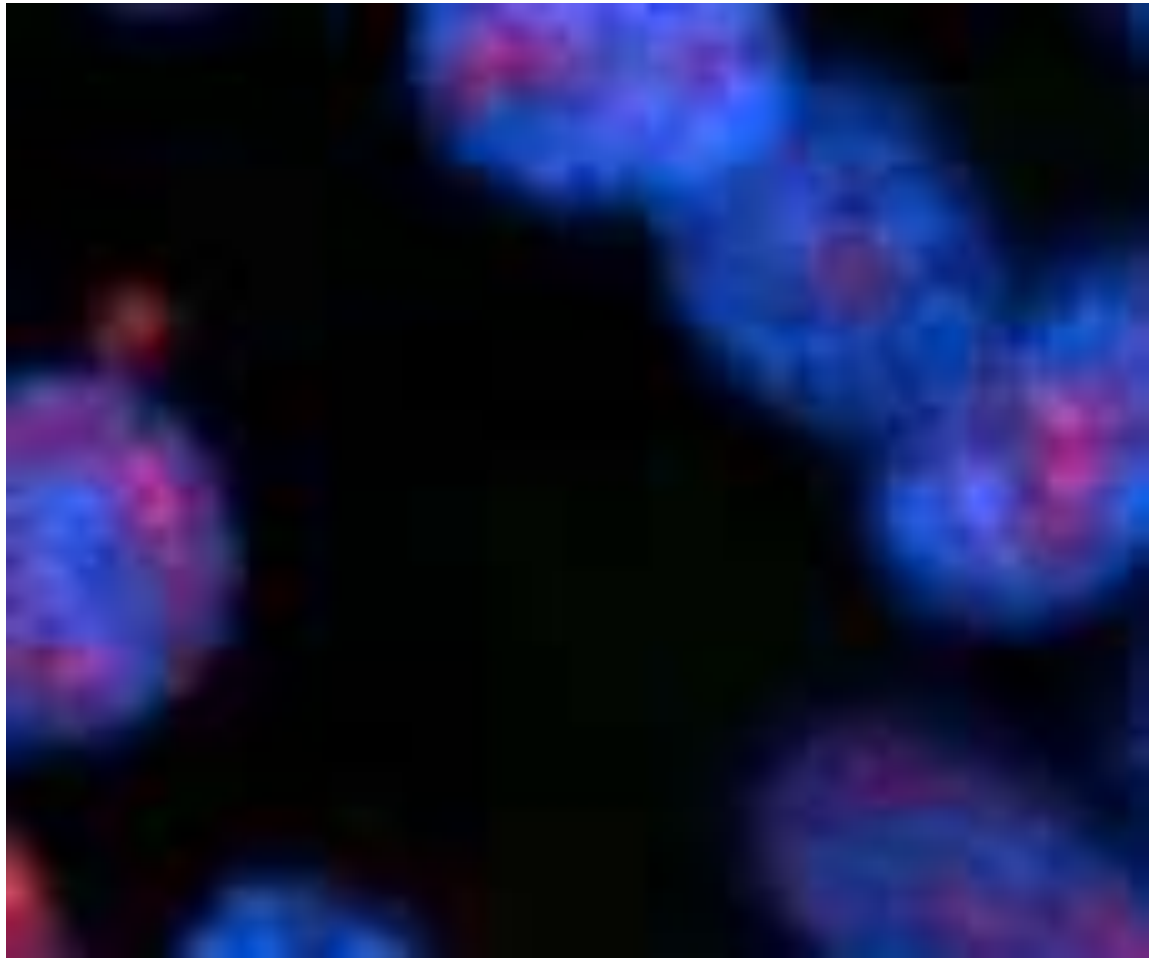
If we know how/where tau aggregates grow in cells, identifies possible ways to inhibit that process and reduce toxicity

Making movies of cellular events is powerful way to study biology

Tau aggregates preferentially grow off surface of cytosolic RNA-protein granules containing SRRM2 and some RNAs.

Genetically engineering colors onto proteins





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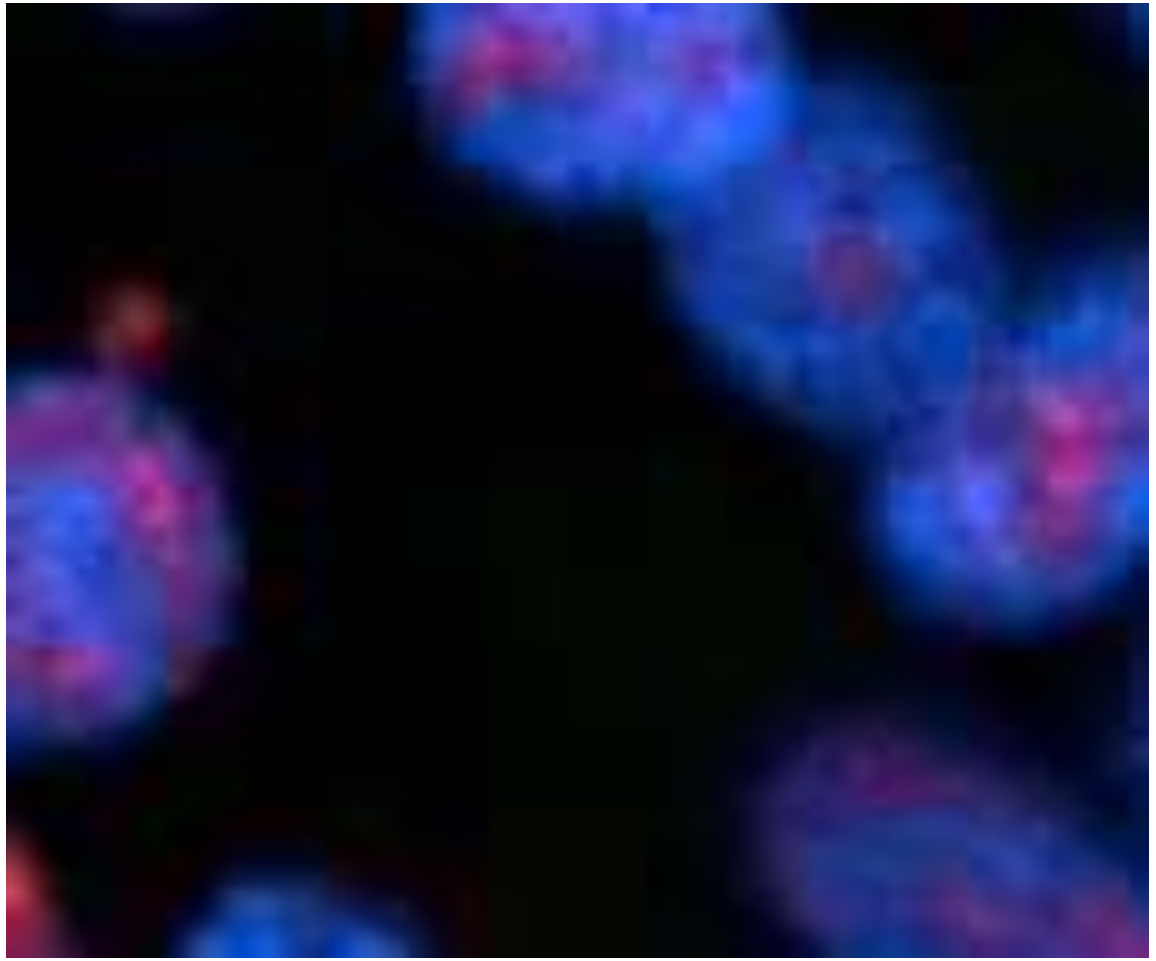
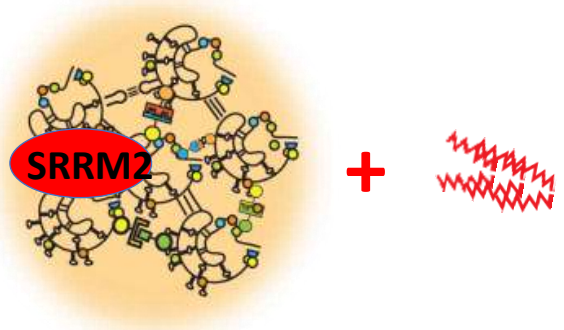
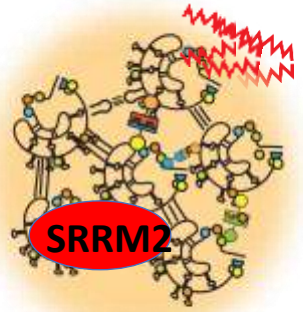
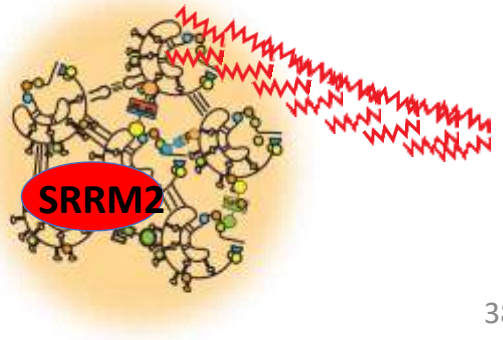




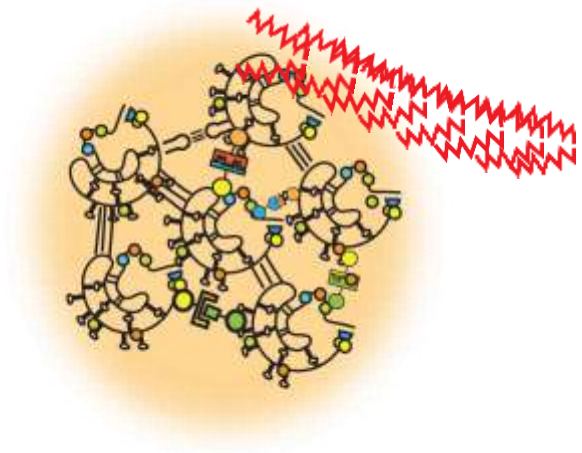
Tau seed interacts with surface of RNA-protein granule (Mitotic Interchromatin Granule (MIG))



Surface of RNA-protein granule (MIG) creates ideal Place for aggregate to grow



Is this Relevant to Disease?

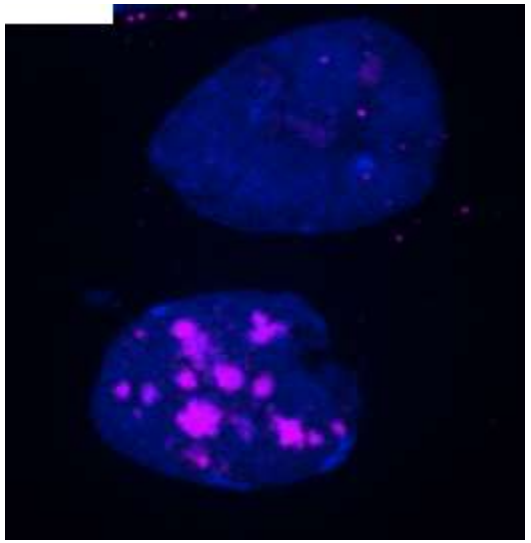


1) Cytosolic SRRM2 assemblies (MIG-like?)are seen in post-mortem neurons from Alzheimer's disease patients. (Tanaka et al., 2018). (Relationship to tau not examined).

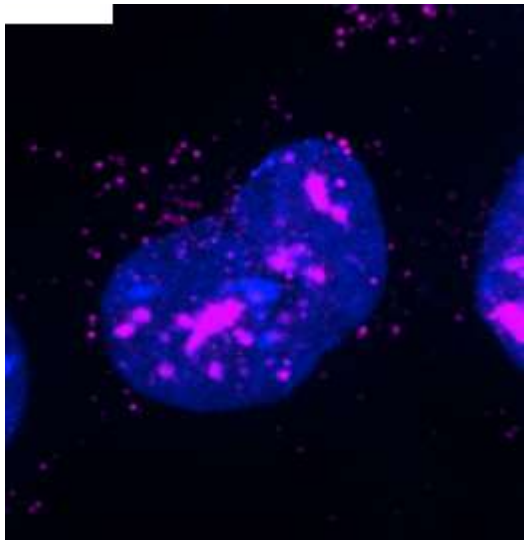
2) Cytosolic SRRM2 assemblies (MIG-like) are triggered in neurons in the lab by markers of neuroinflammation. (No tau here)

Spoorthy Reddy

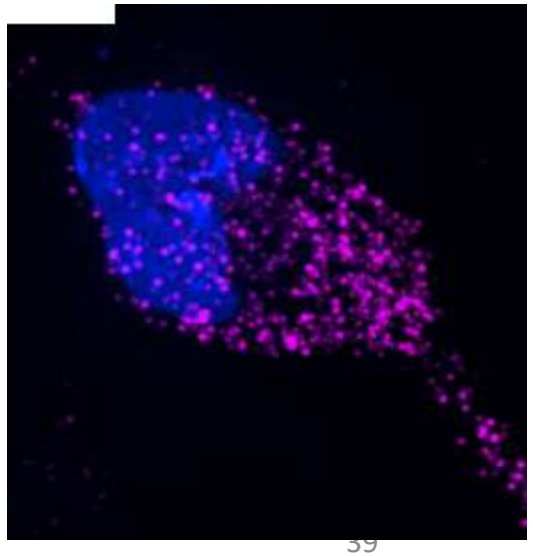
Normal Neuron



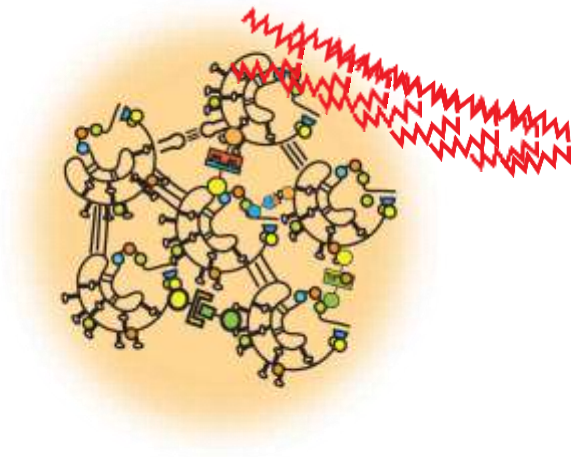
+ Prostaglandin PJD2
(increases in Neuroinflammation)



+ Prostaglandin PJD2
(Extreme Example)



Tau aggregates preferentially grow off surface of cytosolic RNA-protein granules containing SRRM2 and some RNAs.



This is important since it identifies a specific biochemical site within cells where tau aggregation preferentially occurs.

What is actually going on?

What is the biochemical property that promotes tau aggregation?

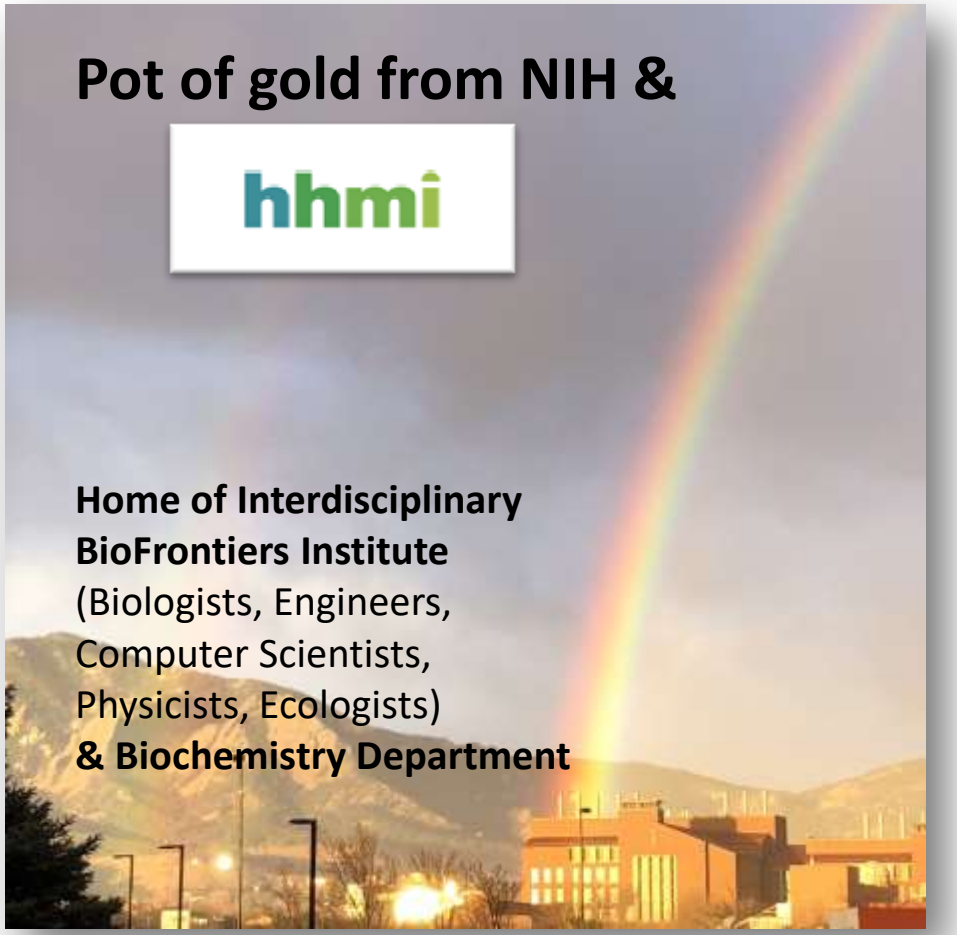
Can we alter it to change tau aggregation?

What is the role of RNA in tau aggregates?

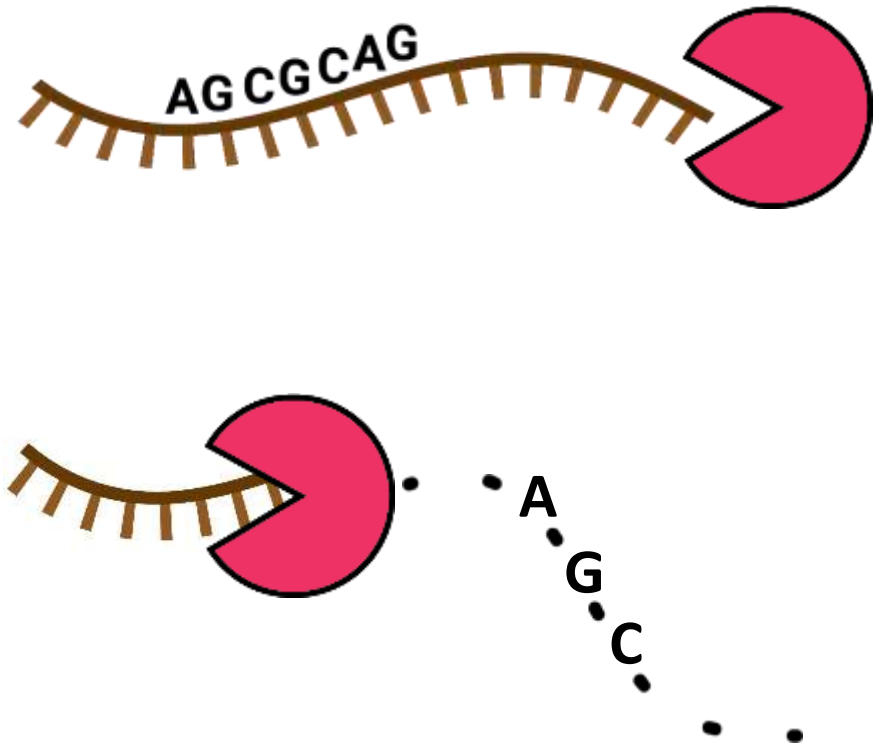
Do RNA and RNA binding proteins in tau aggregates contribute to neuronal death?

Thank you for invitation and listening.

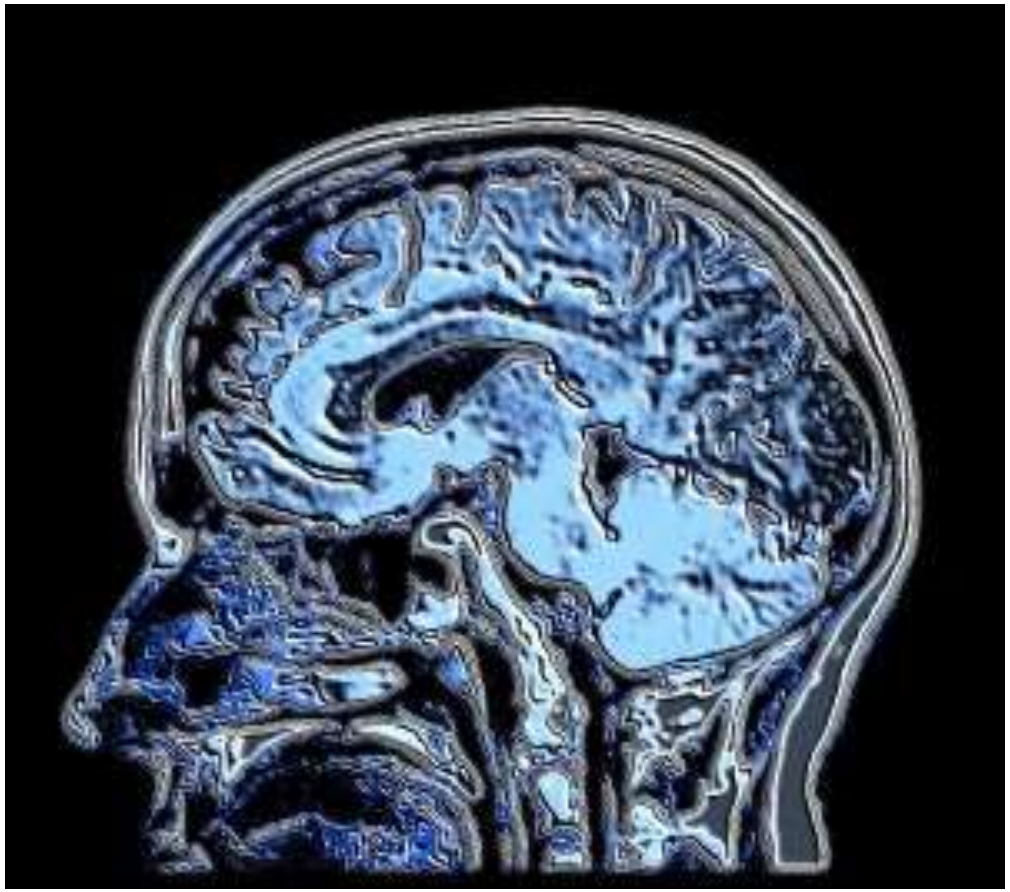
Parker Lab at University of Colorado Boulder



How RNAs are degraded and how that impacts human disease And possible therapies.

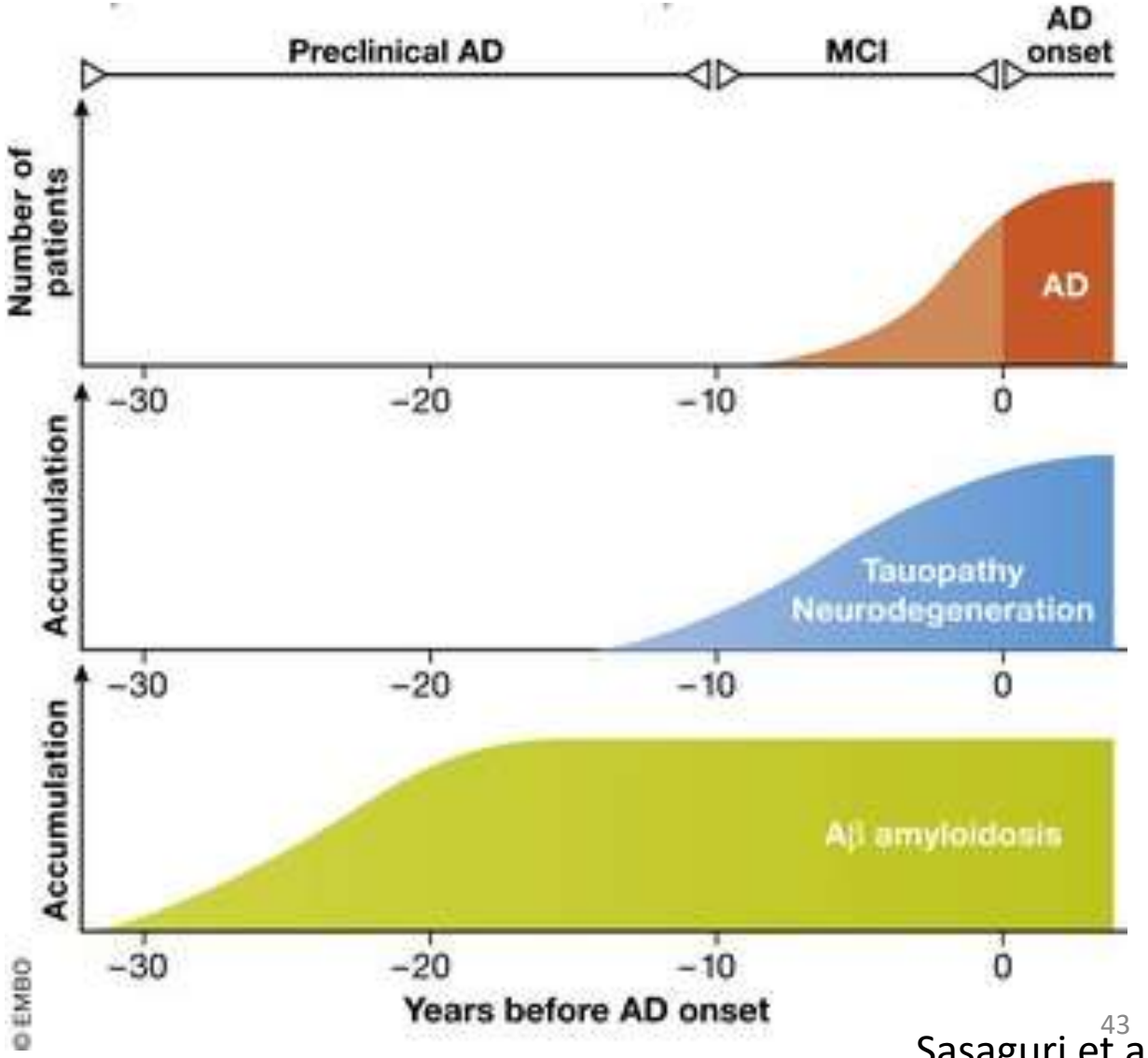


RNA biology and its connection to some neurodegenerative diseases



β-amyloid plaques form first in Alzheimer's disease so they may initiate the disease

A. β-amyloid plaques can be observed decades prior to symptoms
B. Neurofibrillary tau tangles can be seen years before symptoms
C. Mild Cognitive Impairment (MCI) and later Alzheimer's disease symptoms



Sasaguri et al., 2017

Mad Cow Disease (and Creutzfeldt-Jacob Disease) are “prion” diseases that can be transmitted by eating an animal with the prion disease

Cow with ”Mad Cow Disease”

Gets into food chain

Spreads as prion: Causes neuron death and disease