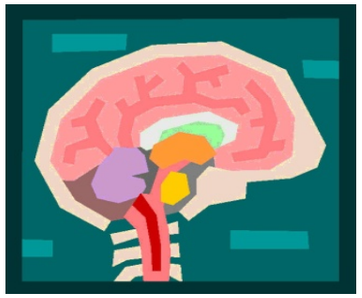

Generating mouse models of sporadic tauopathies using pathological tau purified from human brains

October 14, 2016

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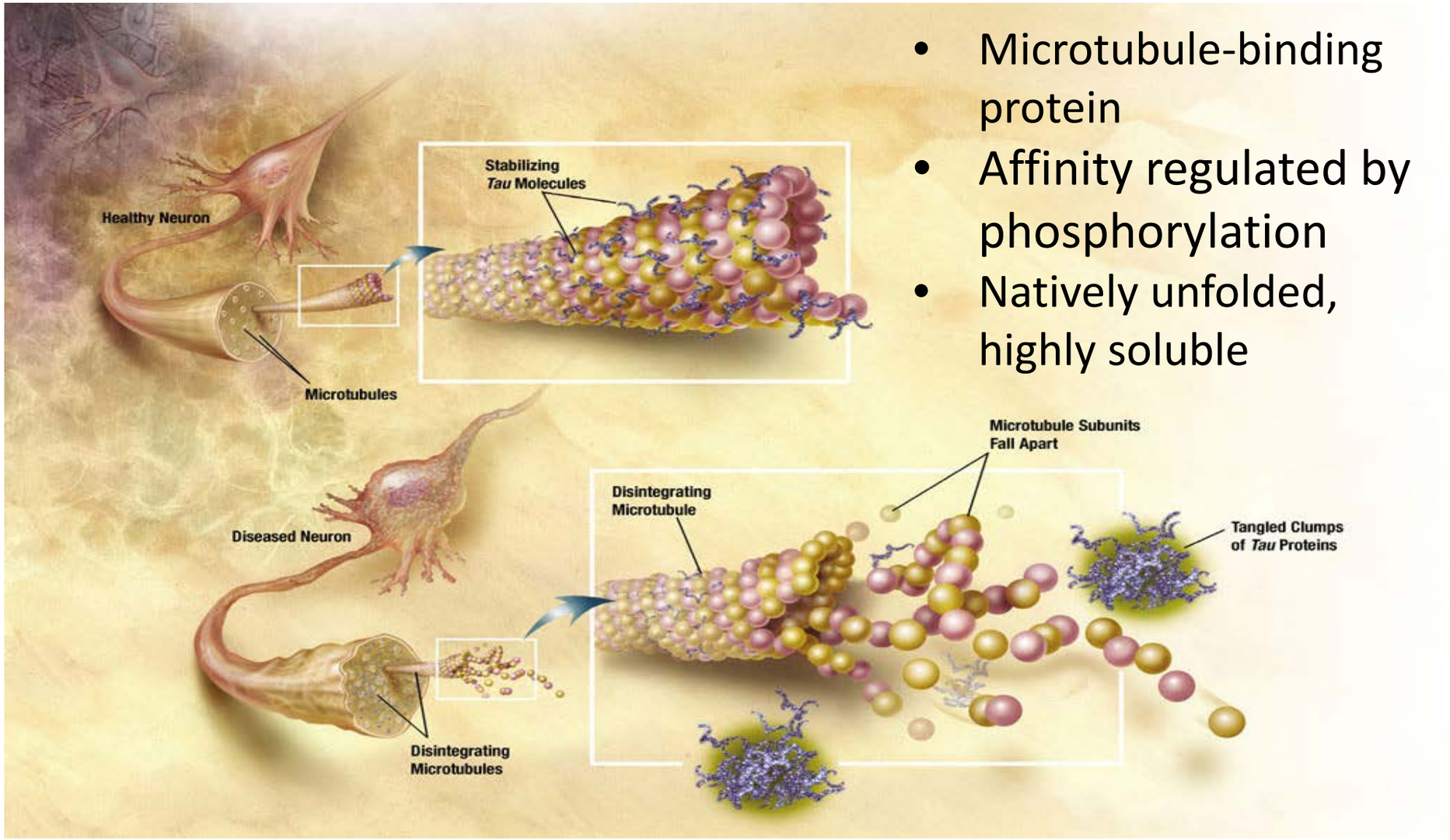
Center for Neurodegenerative Disease Research
University of Pennsylvania

Pathophysiology of tau

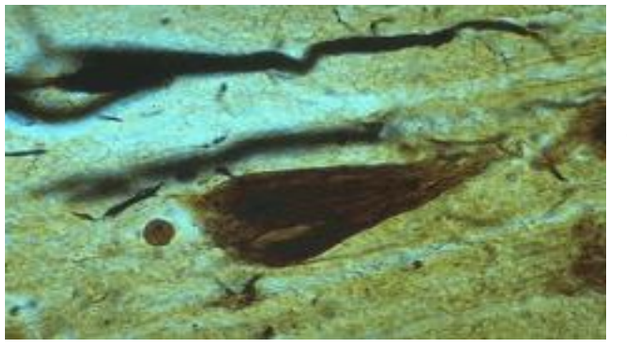


- Microtubule-binding protein
- Affinity regulated by phosphorylation
- Natively unfolded, highly soluble

Alzheimer's disease education and referral, NIA

Tau pathology

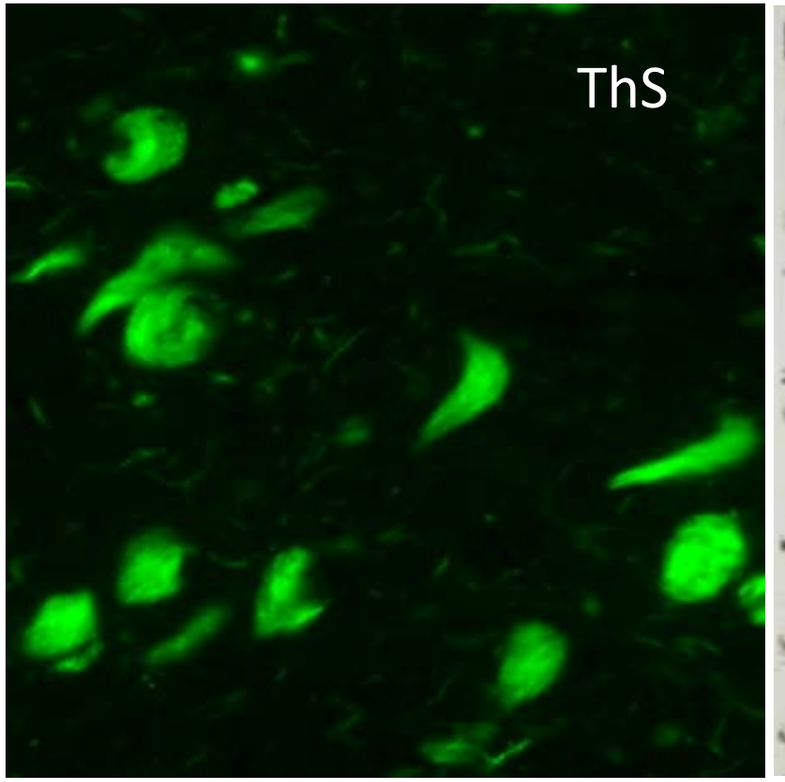
Tauopathies: neurodegenerative diseases characterized by intracellular aggregation of hyperphosphorylated tau, e.g. Alzheimer's disease (AD), corticobasal degeneration (CBD), progressive supranuclear palsy (PSP).



Neurofibrillary tangles (NFT)

Neurofibrillary tangles in AD:

- Enriched β-pleated sheets
- Stained by thioflavin S (ThS), Congo red
- Paired helical filaments with a diameter of ~8-20nm



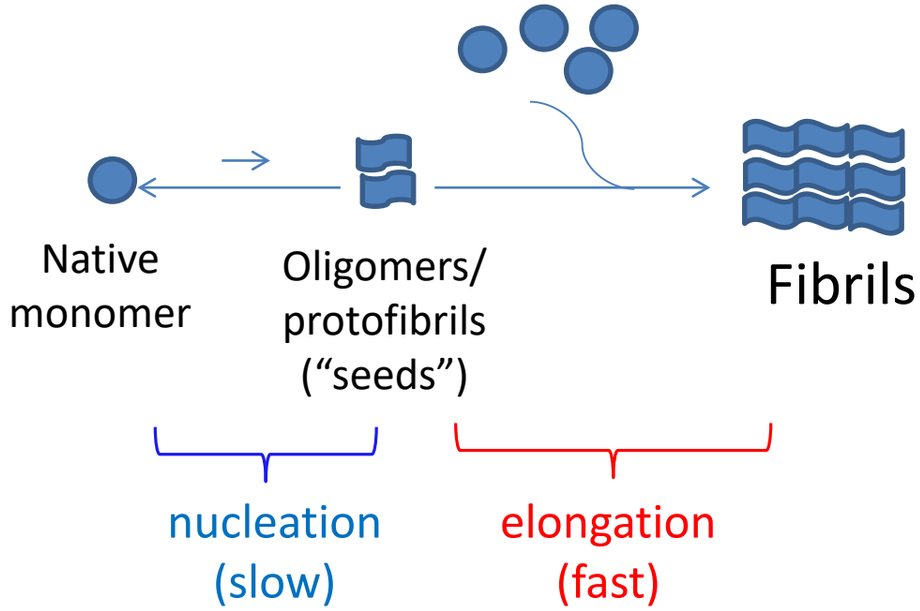
By Josh Daniels

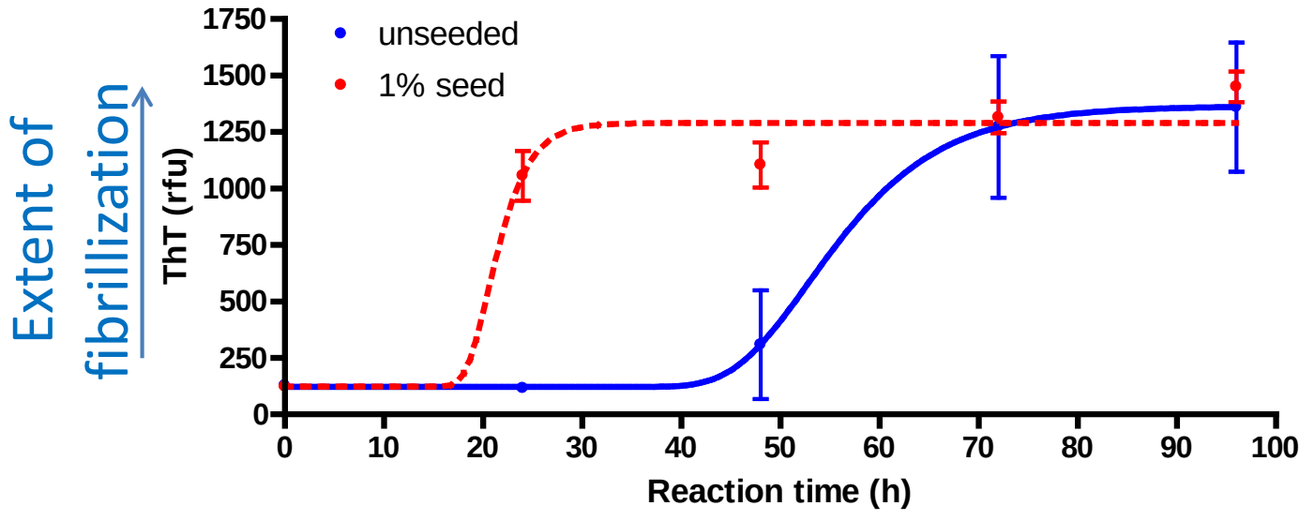
Lee et al., 1991

Developing cellular and mouse models with tau aggregation

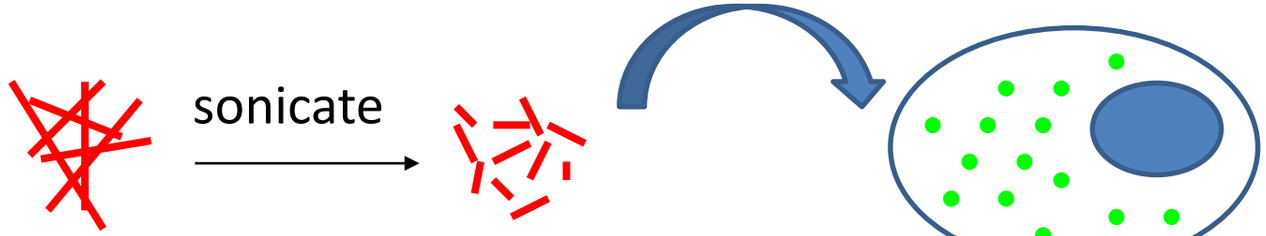
- Rationale: Tau pathology is a key mediator of neurodegeneration.
- NFTs correlate with neuron loss and cognitive decline better than Aβ plaques
- >40 tau mutations found in patients with frontotemporal dementia and Parkinsonism linked to chromosome 17
- Use of models:
- For elucidating mechanisms underlying the onset and progression of the diseases (?)
- For understanding mechanisms of tau-mediated neuronal dysfunction/neurodegeneration
- For preclinical testing of potential therapies
- Challenge: Tau is a highly soluble protein. Overexpression of even mutant tau does not lead to aggregation in cells.

Nucleation-dependent fibril assembly

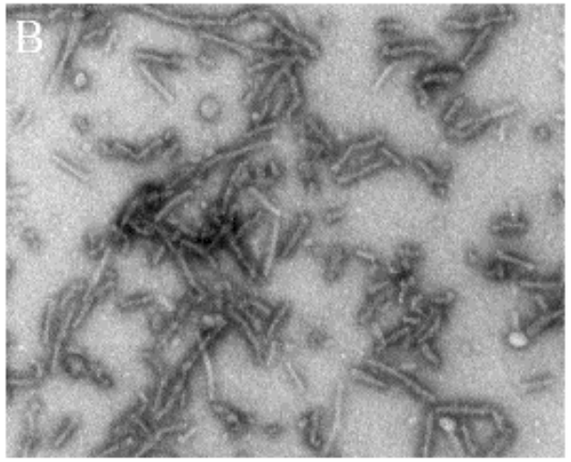




Seeded fibrillization in cultured cells



Preformed tau fibrils (**tau pffs**)



Heparin-induced

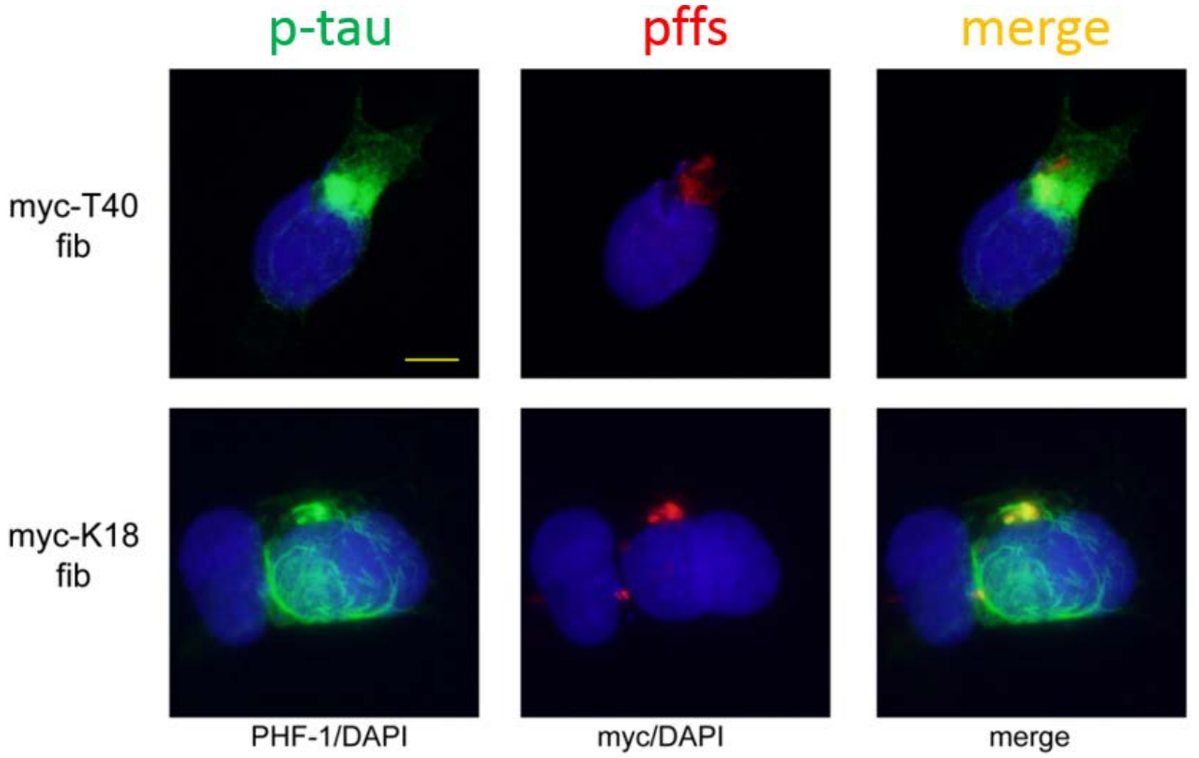
Cells overexpressing tau

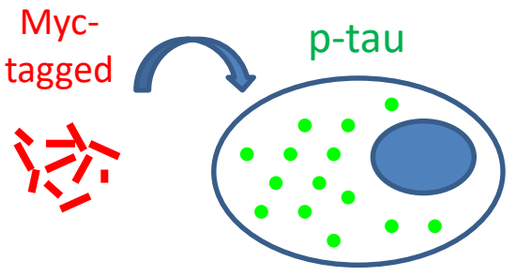
Internalization of tau pffs

Soluble tau recruited by pffs into aggregates

Intracellular tau fibrillization seeded by exogenous pffs in cultured cells

QBI 293 cells transiently transfected with WT T40:





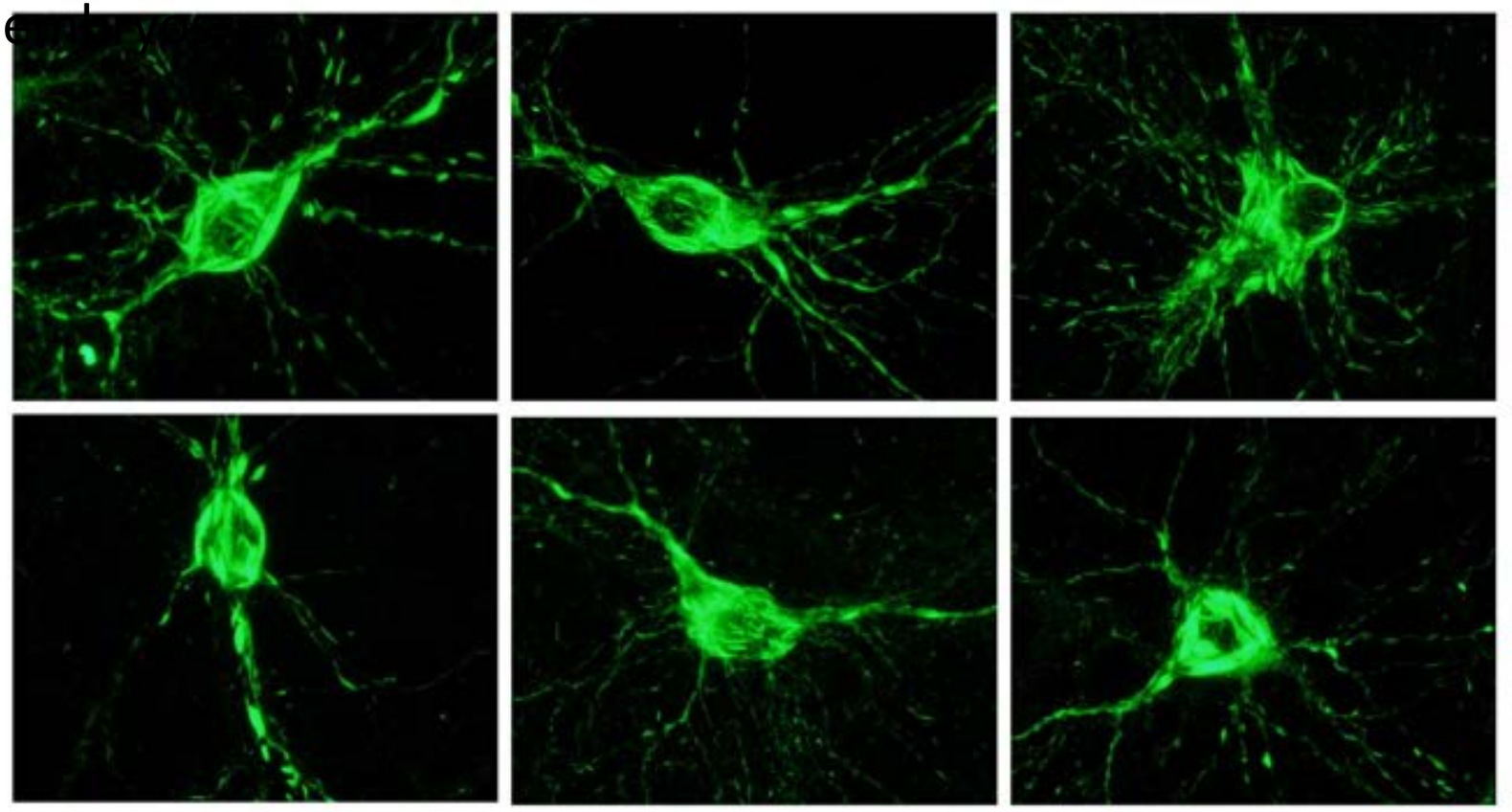
1% Triton-X extraction during fixing (remove soluble proteins)

- Internalization of small quantities of tau pffs is sufficient to template the fibrillization of soluble tau.
- Tau pffs can spontaneously enter cells through endocytosis.

Guo and Lee, *JBC*, 2011

Tau pffs induce tangle-like aggregates in primary neurons

Primary hippocampal neurons from PS19 (P301S tau) mouse

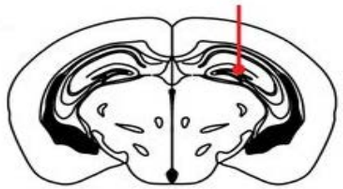


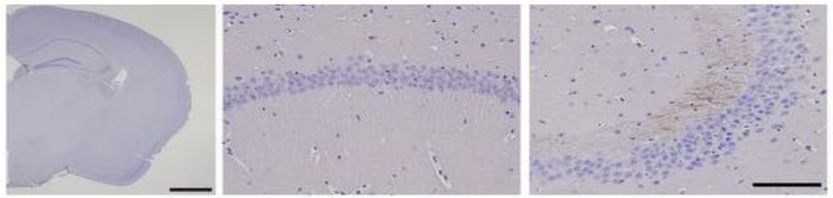
Guo and Lee, *FEBS Letters*, 2013

Seeding by preformed tau fibrils (tau pffs) *in vivo*

Hippocampal injection of tau pffs into 2-3 mo old PS19 (P301S) mice

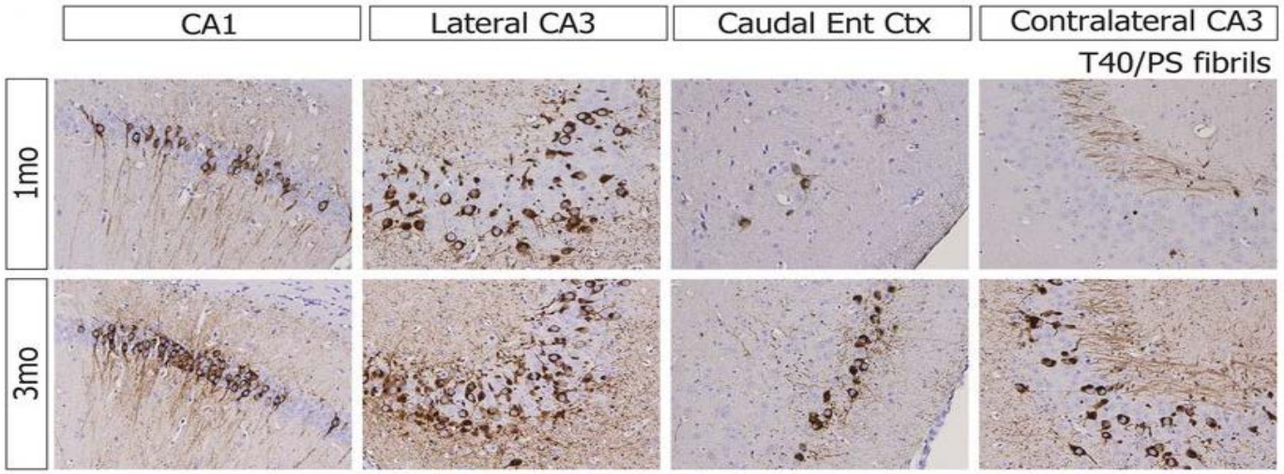
AT8 (p-tau)



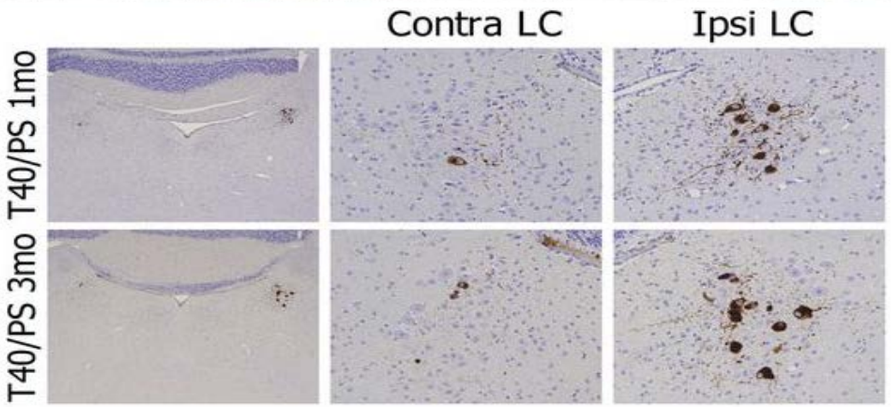


PBS control

Tau pff injected



Untreated PS19: appreciable pathology seen in 12 mo old mice

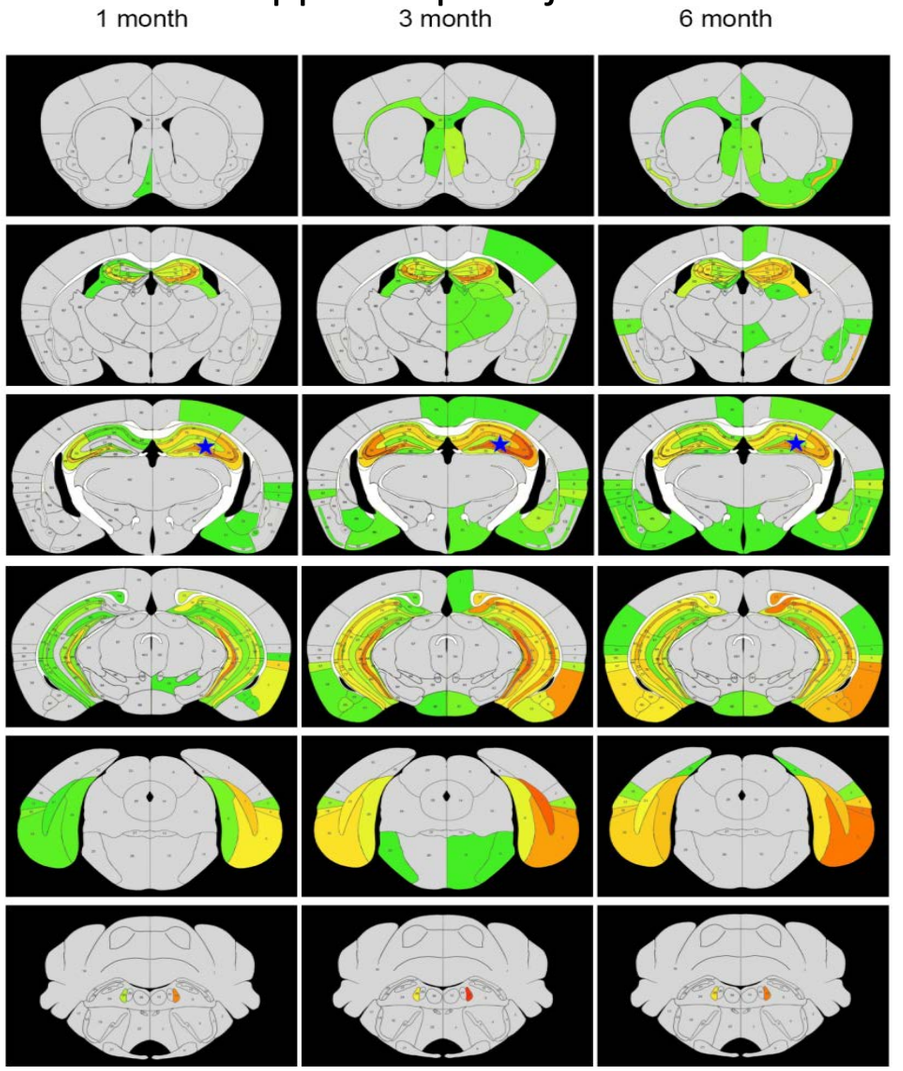


LC: locus coeruleus

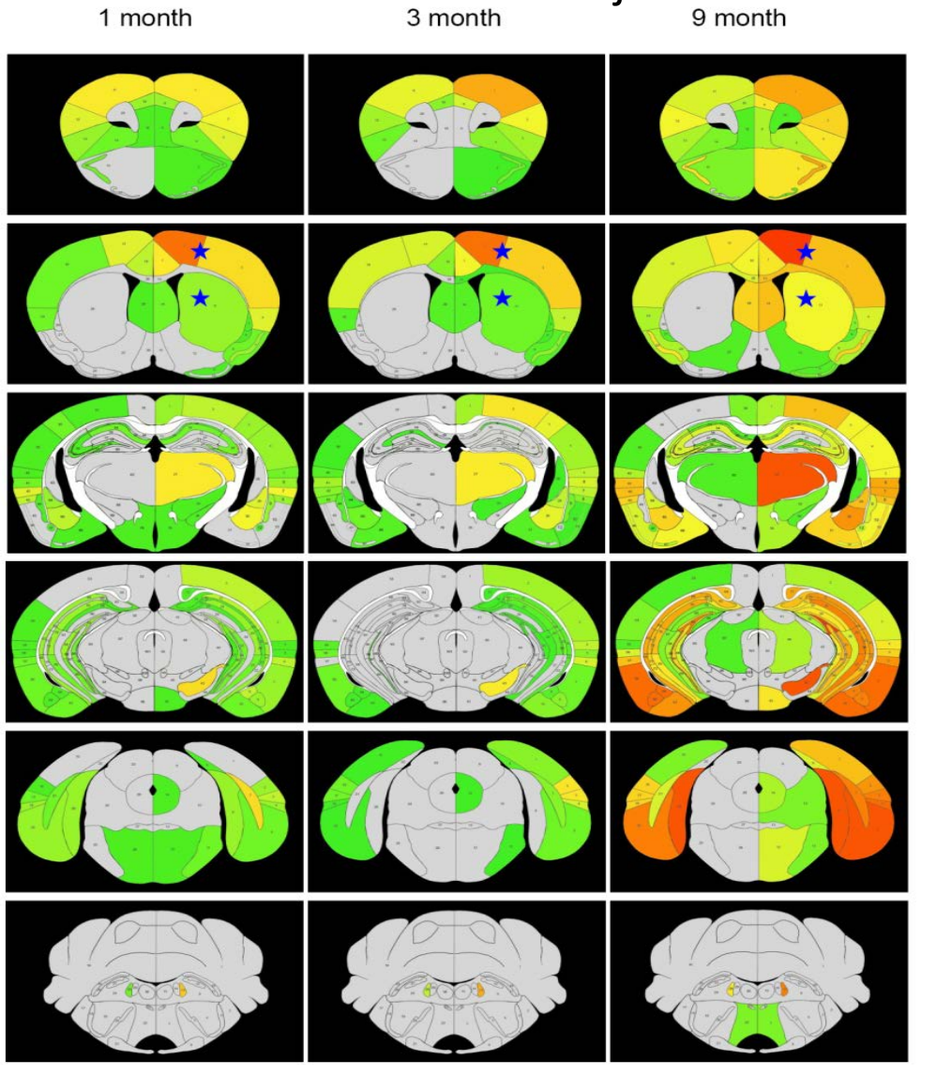
Iba, Guo, et al., *J. of Neuroscience*, 2013

Distinct patterns of tau pathology induced by different injection sites

Hippocampal injection

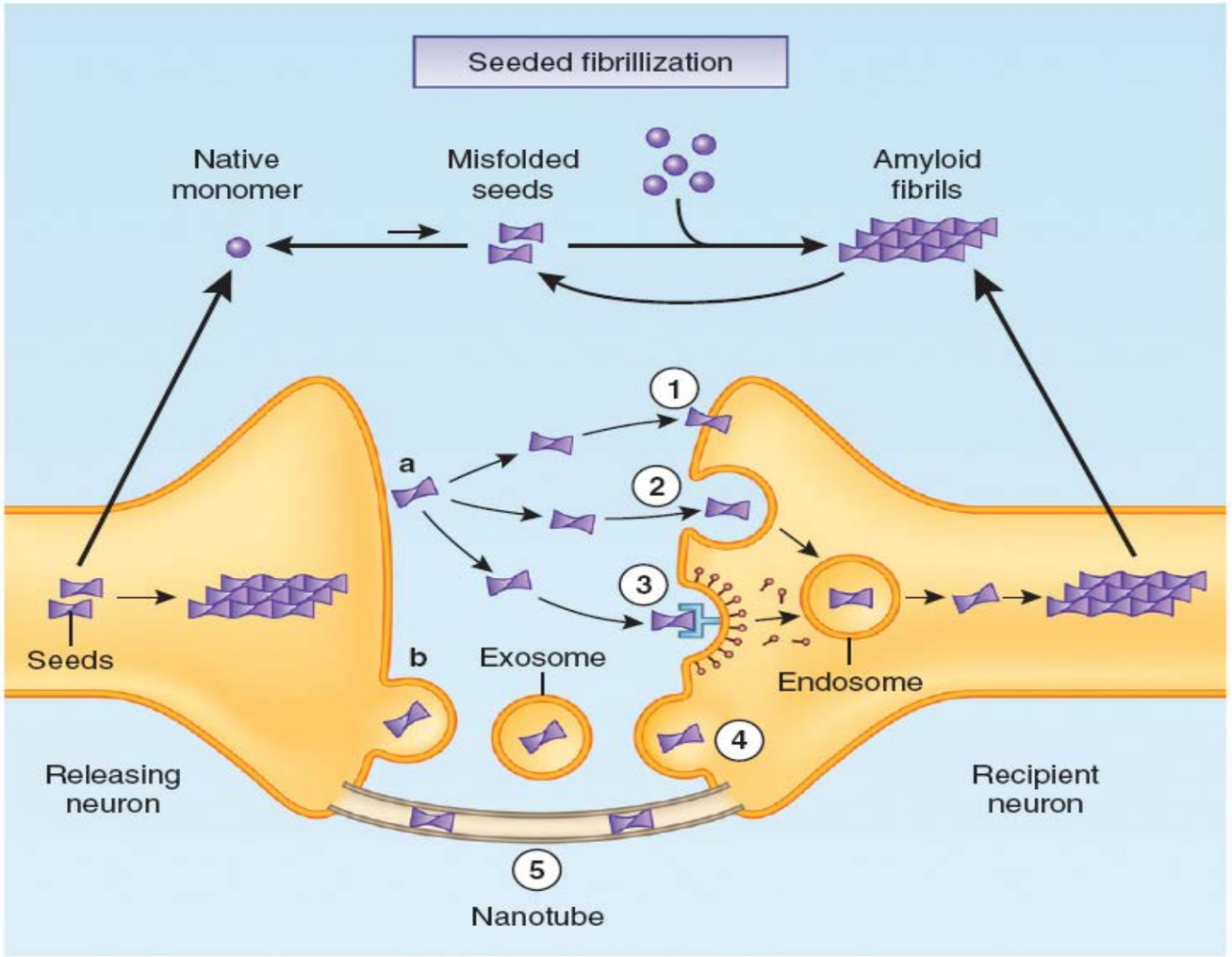


Striatum and cortex injection



Iba, Guo, et al., *J. of Neuroscience*, 2013

The “prion-like” cell-to-cell transmission of disease-associated protein aggregates



Guo and Lee, *Nat. Med*, 2013

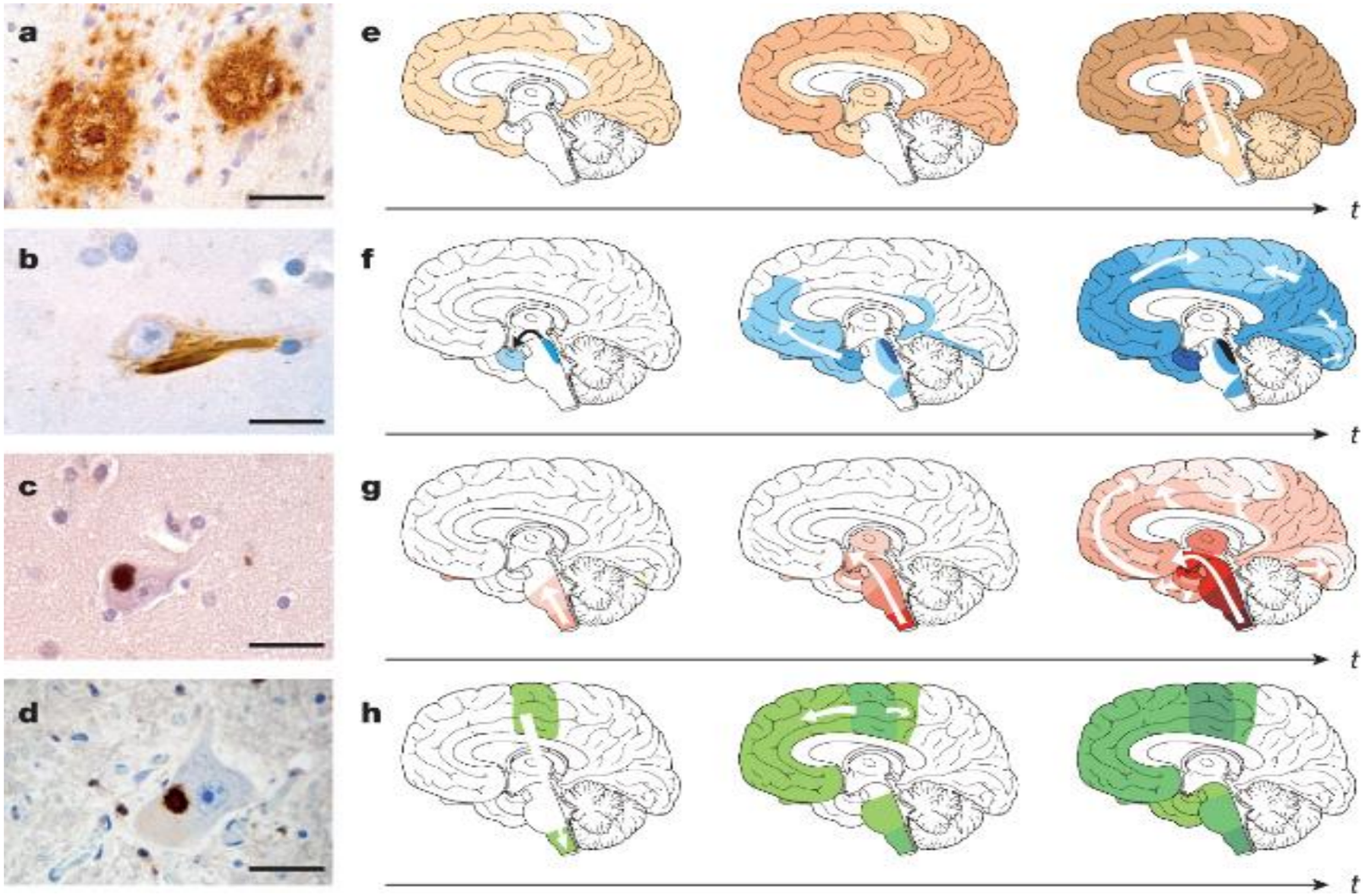
Stereotypical spreading of disease pathology as shared phenomenon among neurodegenerative diseases

Aβ plaques

Tau tangles

α-syn Lewy bodies

TDP-43 inclusions



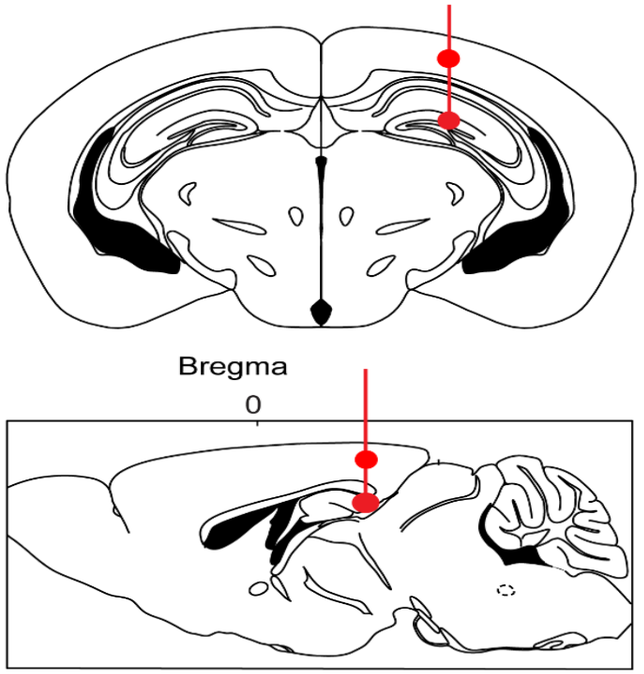
Jucker and Walker, *Nature*, 2013

Outline

A. Generating a mouse model of sporadic tauopathies using AD brain-derived tau aggregates
B. Exploring the effects of Aβ plaques on tau pathology in plaque-bearing mice
C. Characterizing pathology induced by tau aggregates purified from different tauopathies

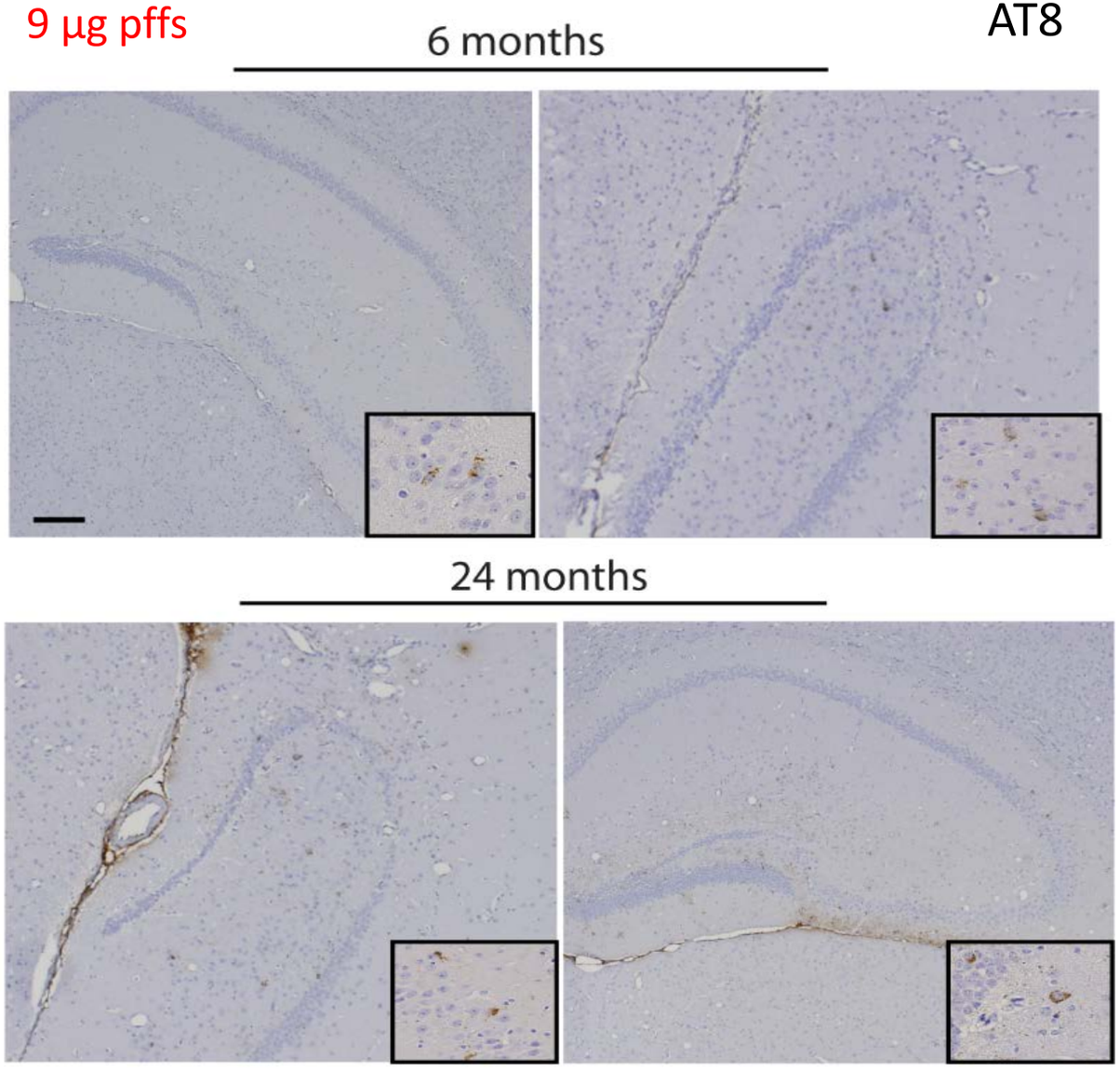
Injection of synthetic tau pffs into WT mice

2-3 mo old WT mice



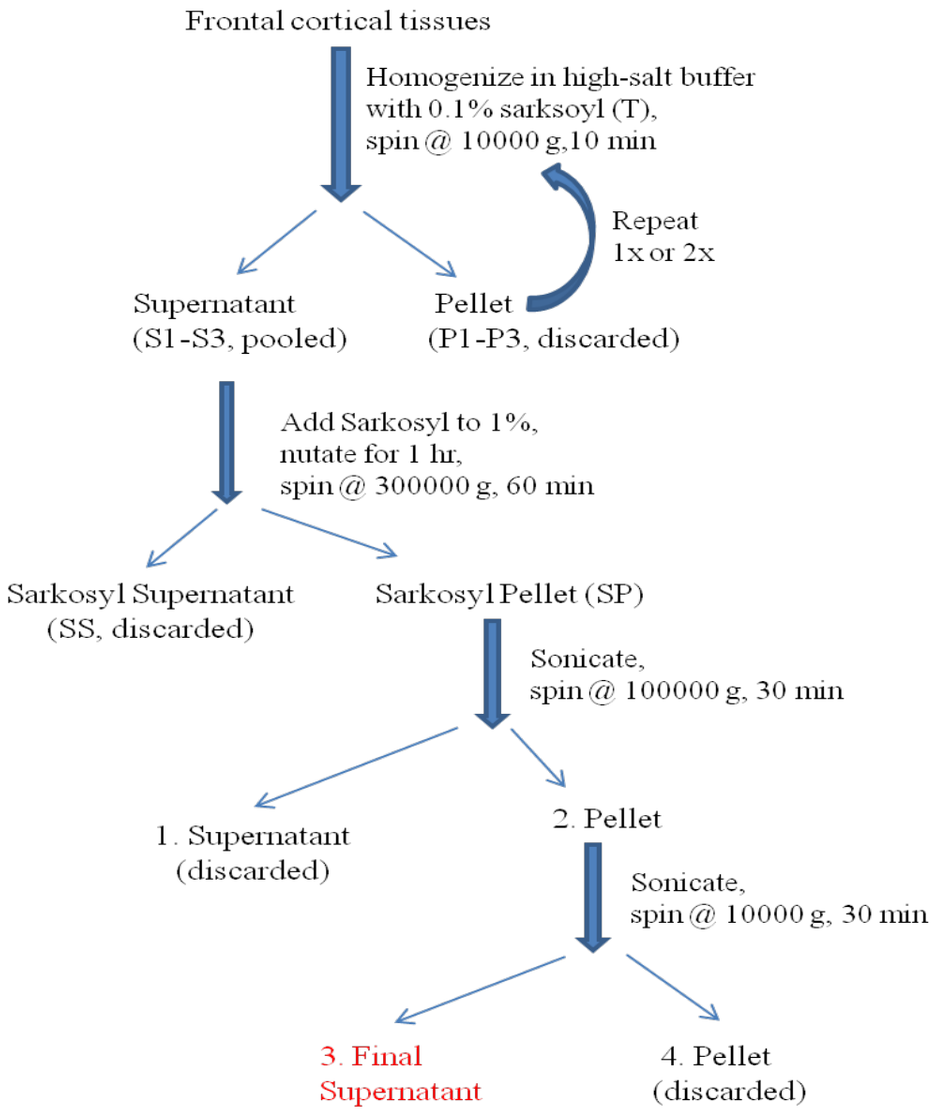
9 µg pffs

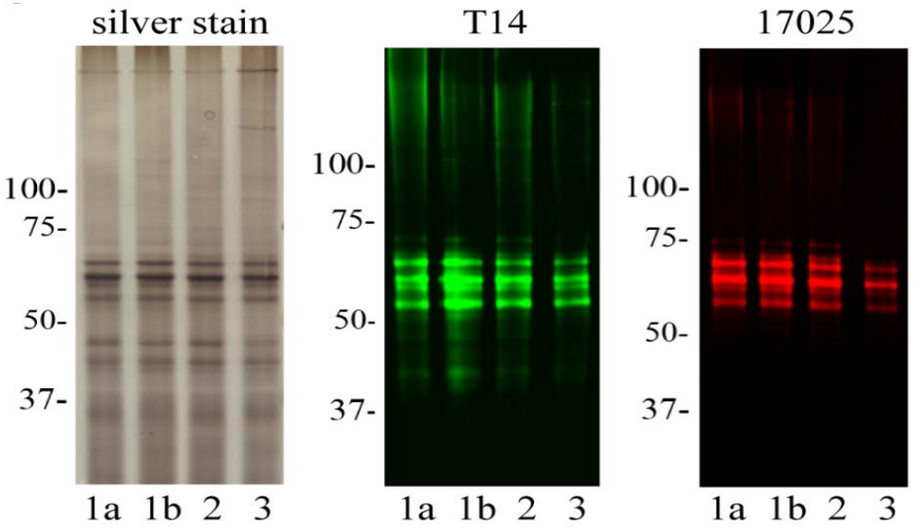
AT8

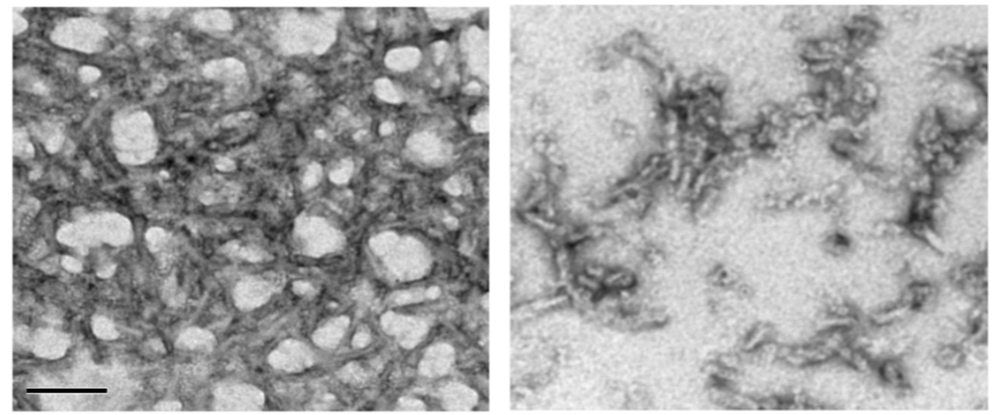


Guo et al., *JEM*, in press

Purification of pathological tau from AD brains



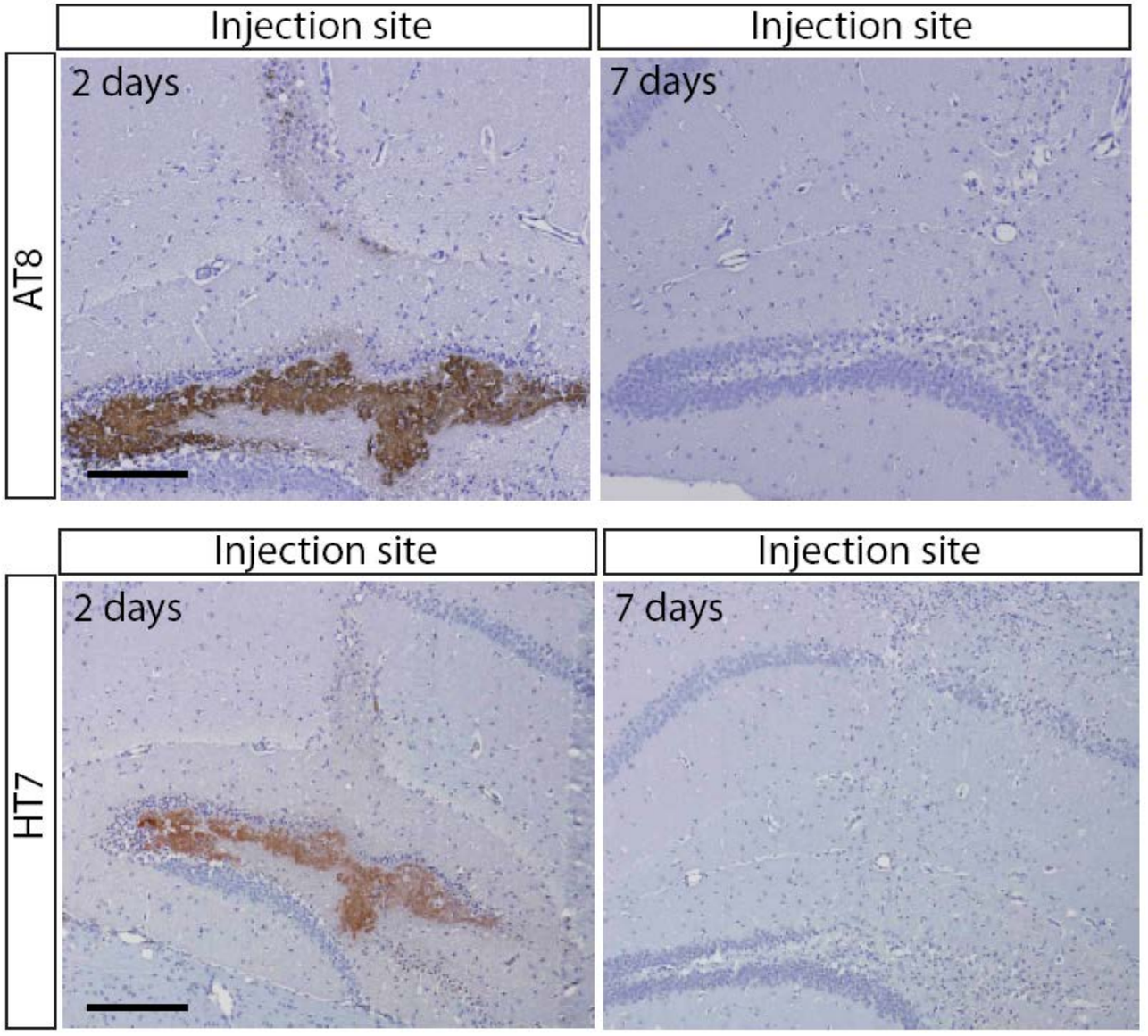




15-30 µg tau in final supernatant (**AD-tau**)/g gray matter
(sucrose gradient method: 1-5 µg/g gray matter)
Purity: 10-28%

Guo et al., *JEM*, in press

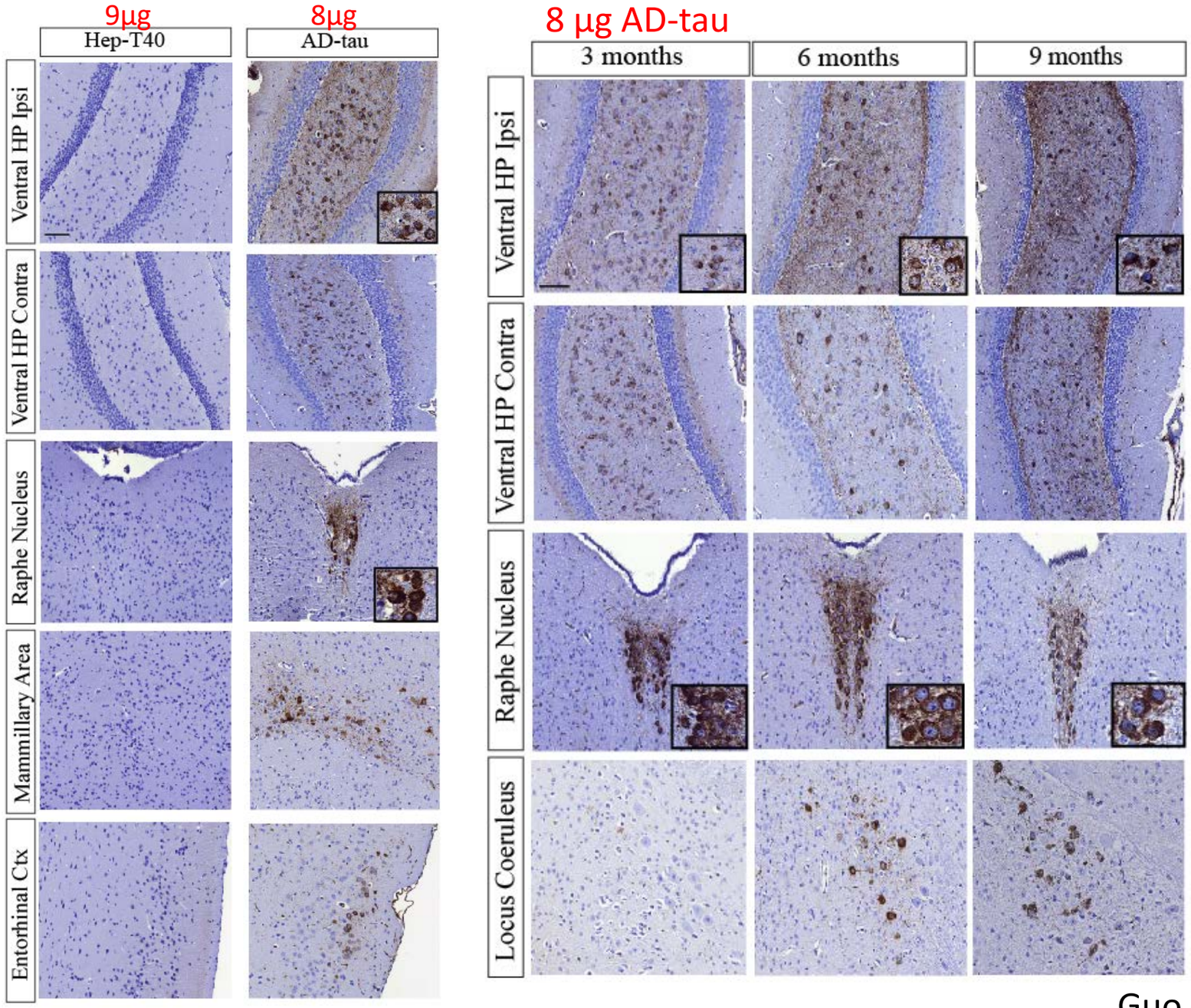
Rapid clearance of injected AD-tau



Guo et al., *JEM*, in press

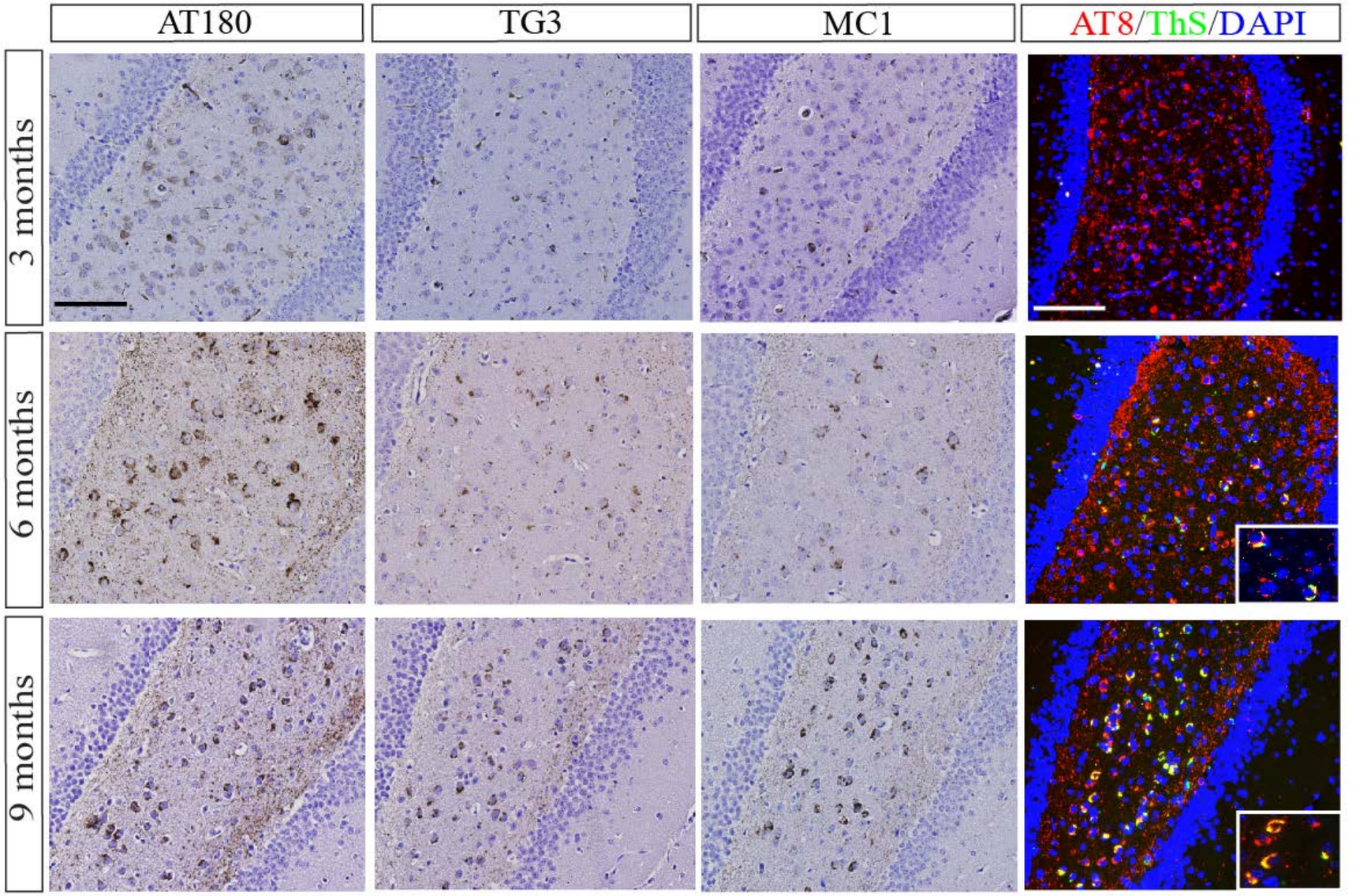
AD-tau induces and propagates tau pathology in WT mice

AT8



Guo et al., *JEM*, in press

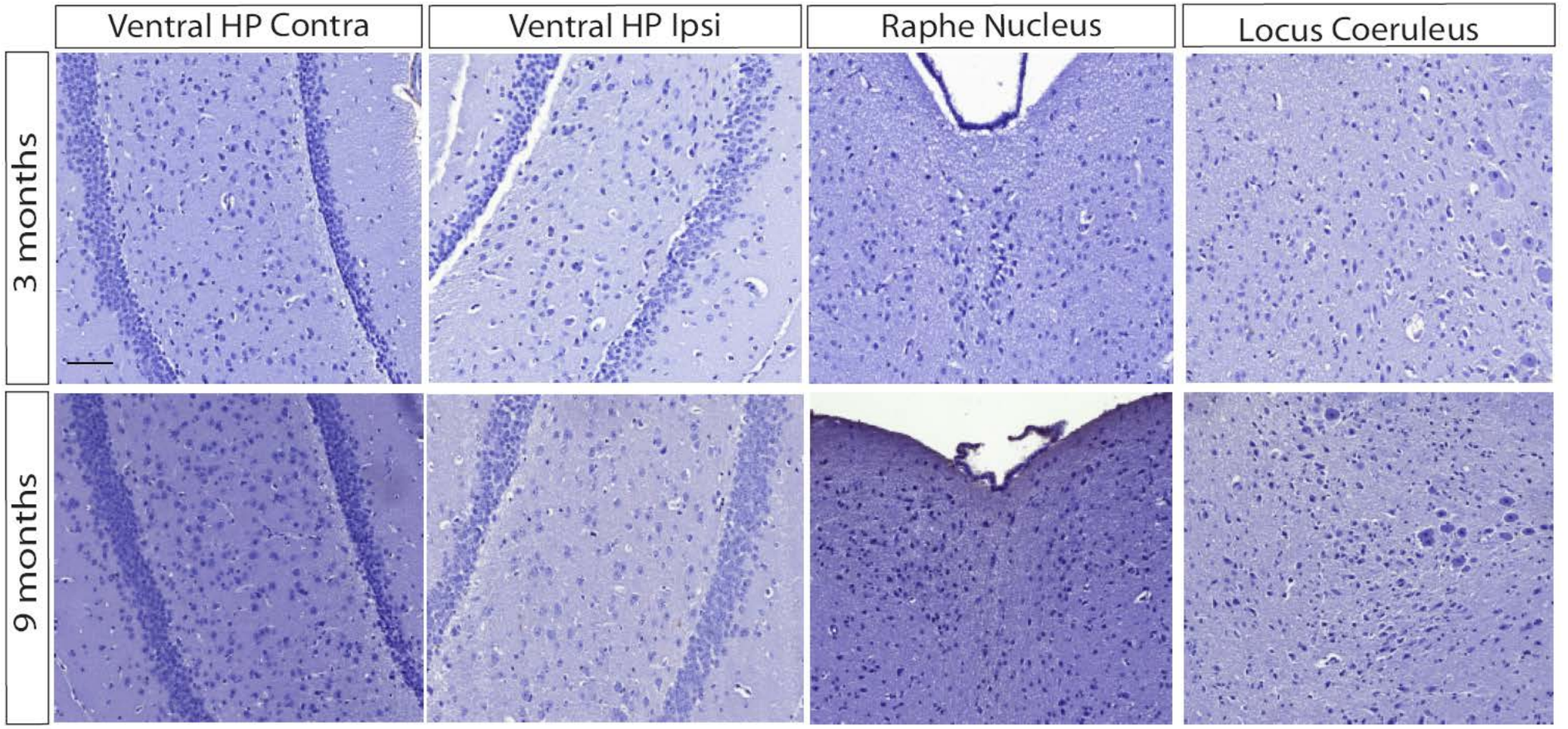
Time-dependent maturation of induced tau aggregates in WT mice



Guo et al., *JEM*, in press

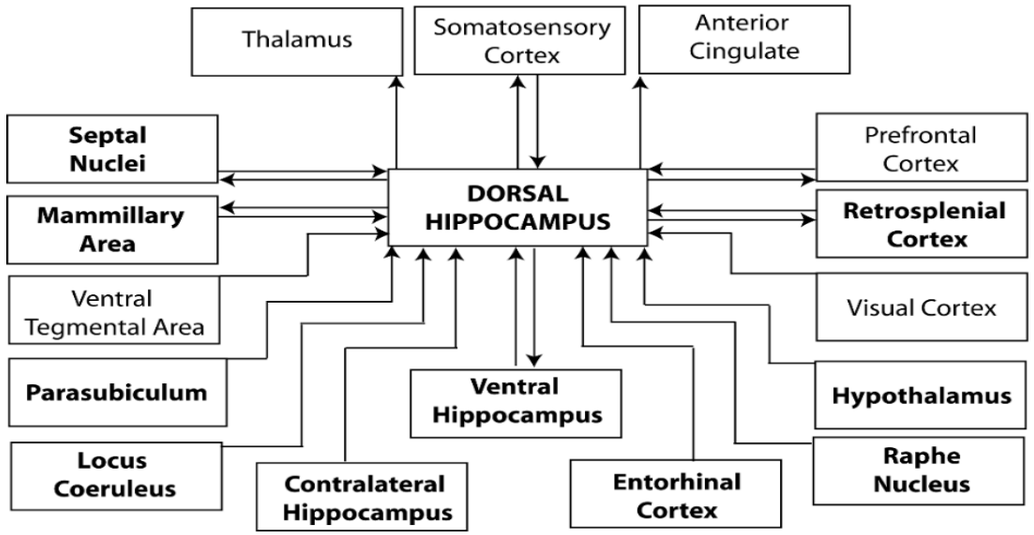
Injection of control brain extracts does not induce tau pathology

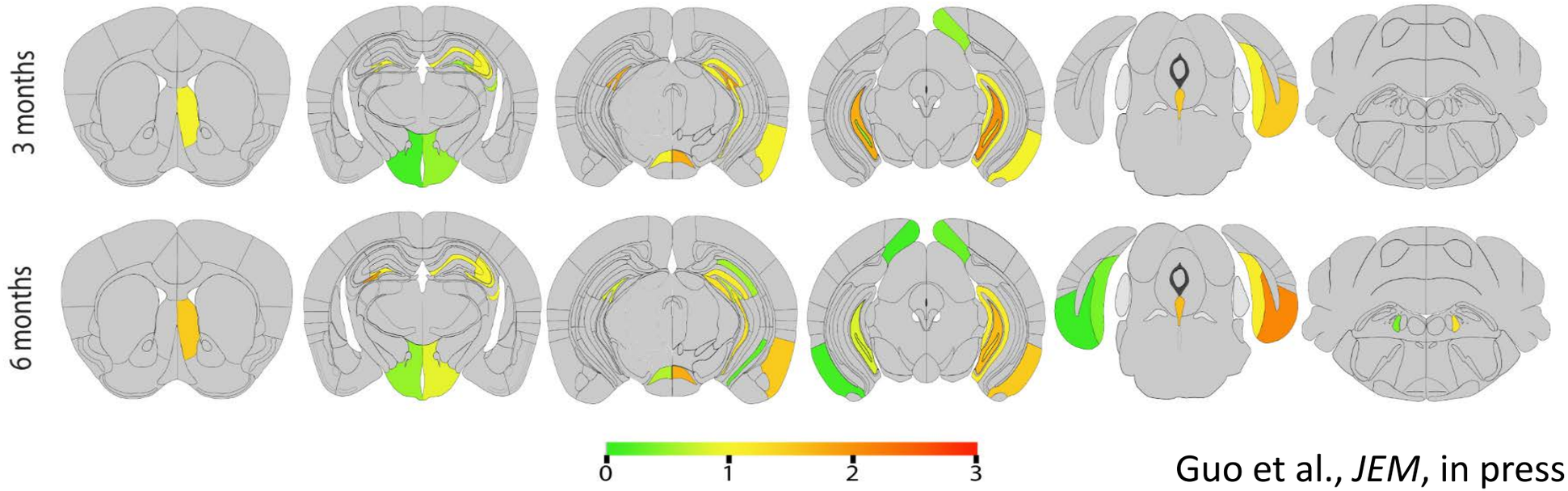
AT8



Guo et al., *JEM*, in press

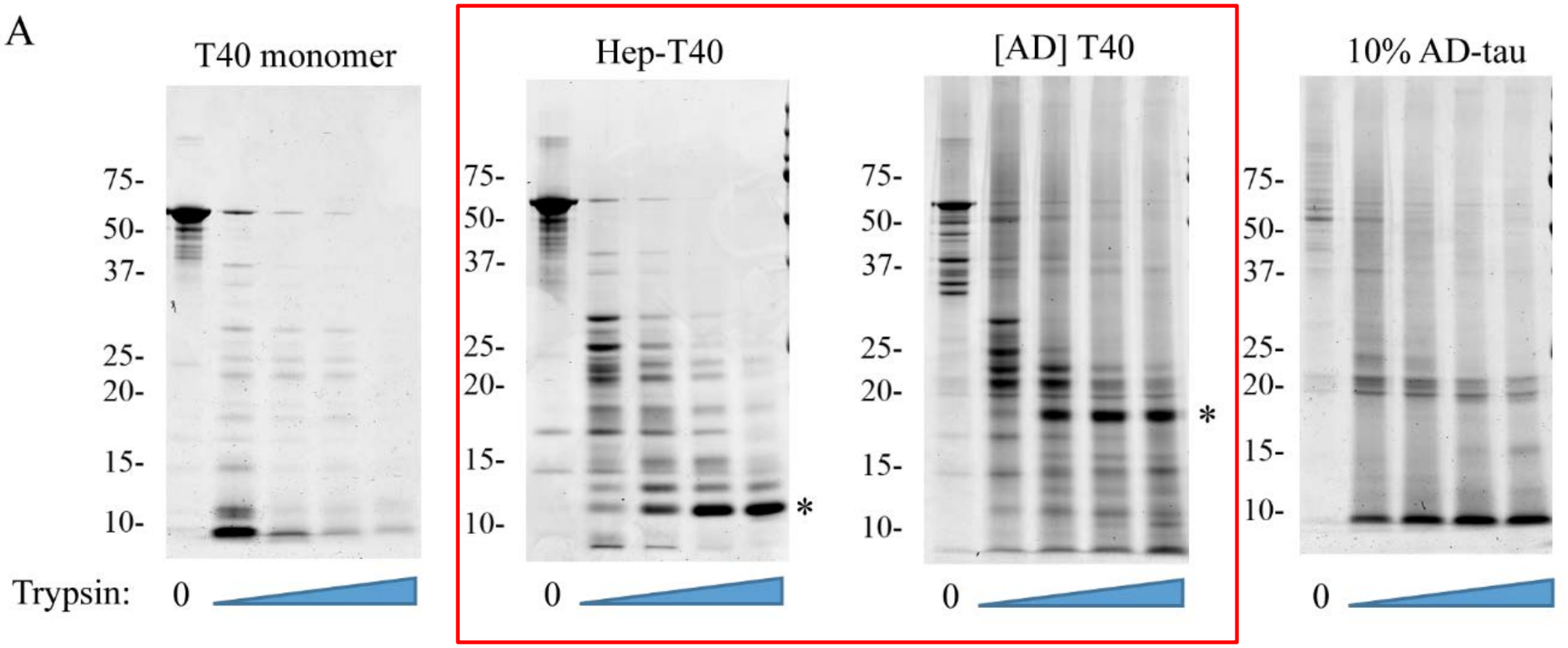
Propagation of tau pathology to anatomically connected brain regions





Guo et al., *JEM*, in press

Conformational difference between synthetic tau fibrils and AD-tau seeded fibrils



Hep-T40: Heparin-induced tau fibrils

[AD]T40: recombinant tau fibrils seeded by 10% AD-tau in vitro

Guo et al., *JEM*, in press

Summary for part A

- Tau fibrils developed in AD brains are different from synthetic tau fibrils.

→ Intracellular milieu? Post-translational modifications? Selection pressure?

- Induction and propagation of tau pathology in WT mice provide stronger support for the transmission hypothesis.
- Relatively restricted progression of tau pathology in WT mice.

→ Other factors are promoting pathological tau transmission in diseased brains? E.g. age, Aβ plaques, genetic risk factors…

Outline

A. Generating a mouse model of sporadic tauopathies using AD brain-derived tau aggregates

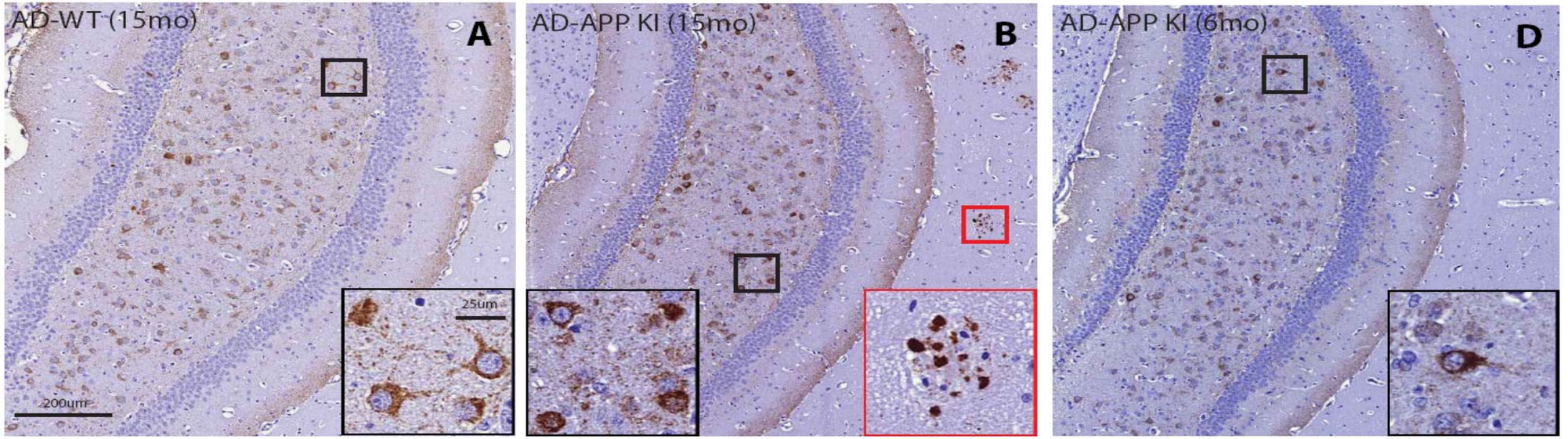
B. Exploring the effects of Aβ plaques on tau pathology in plaque-bearing mice

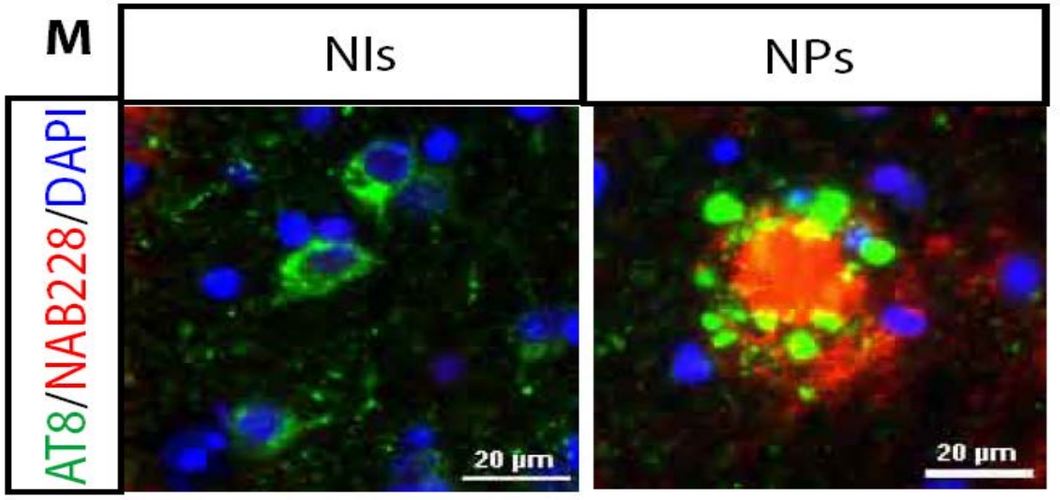
C. Characterizing pathology induced by tau aggregates purified from different tauopathies

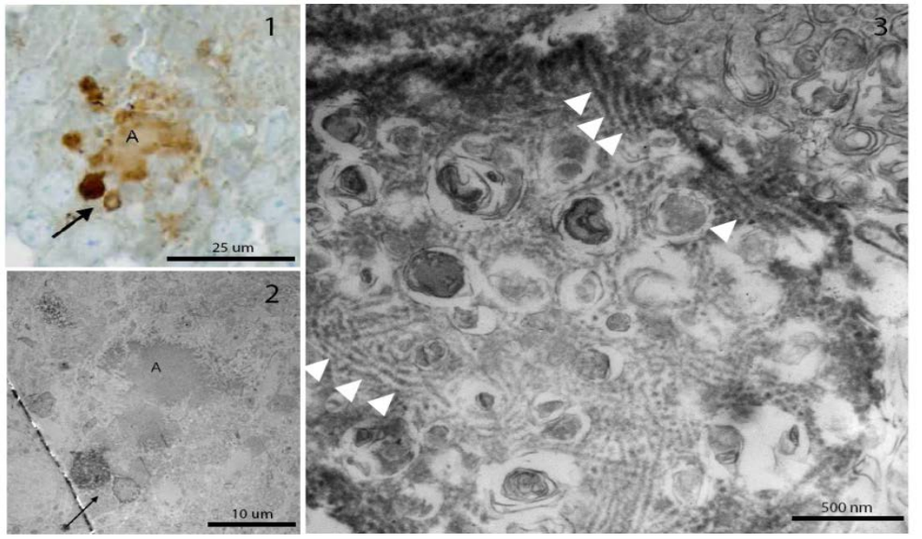
Two types of tau pathology: neuronal inclusions (NIs) and neuritic plaque-like tau aggregates (NPs)

3 months

AT8







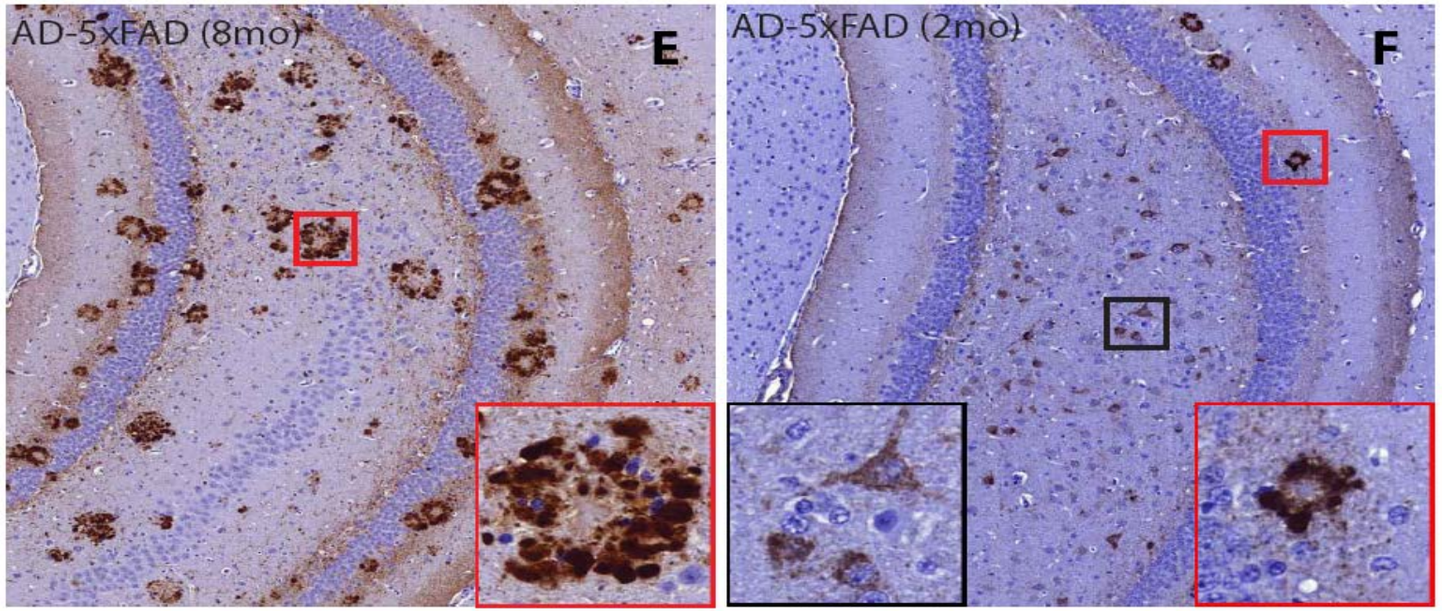
2 µg AD-tau injected in this study.

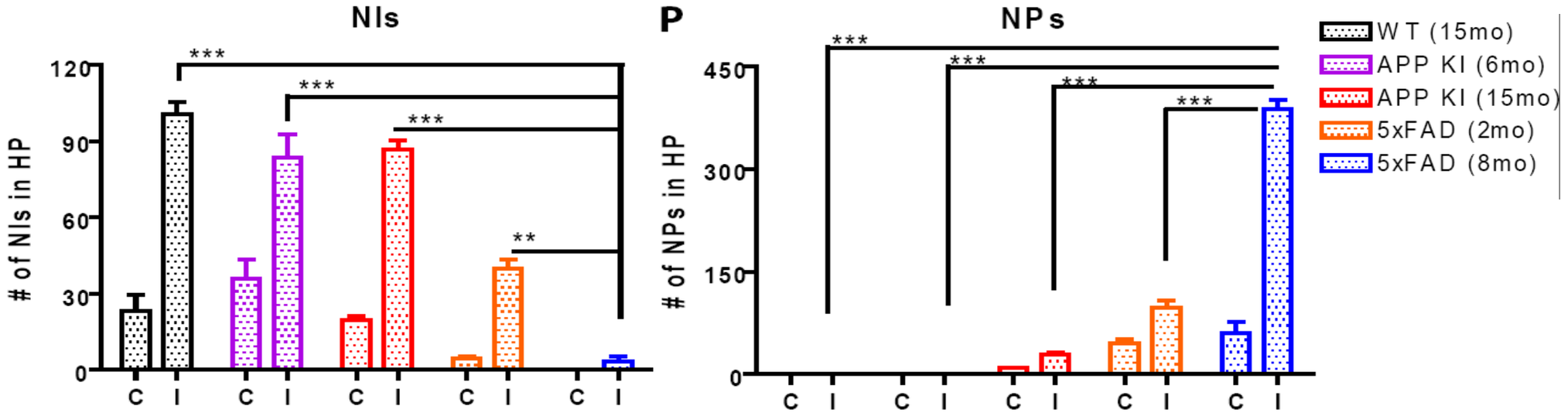
He, Guo, et al., submitted

Competition in the formation of NIs and NPs mediated by plaques

3 months

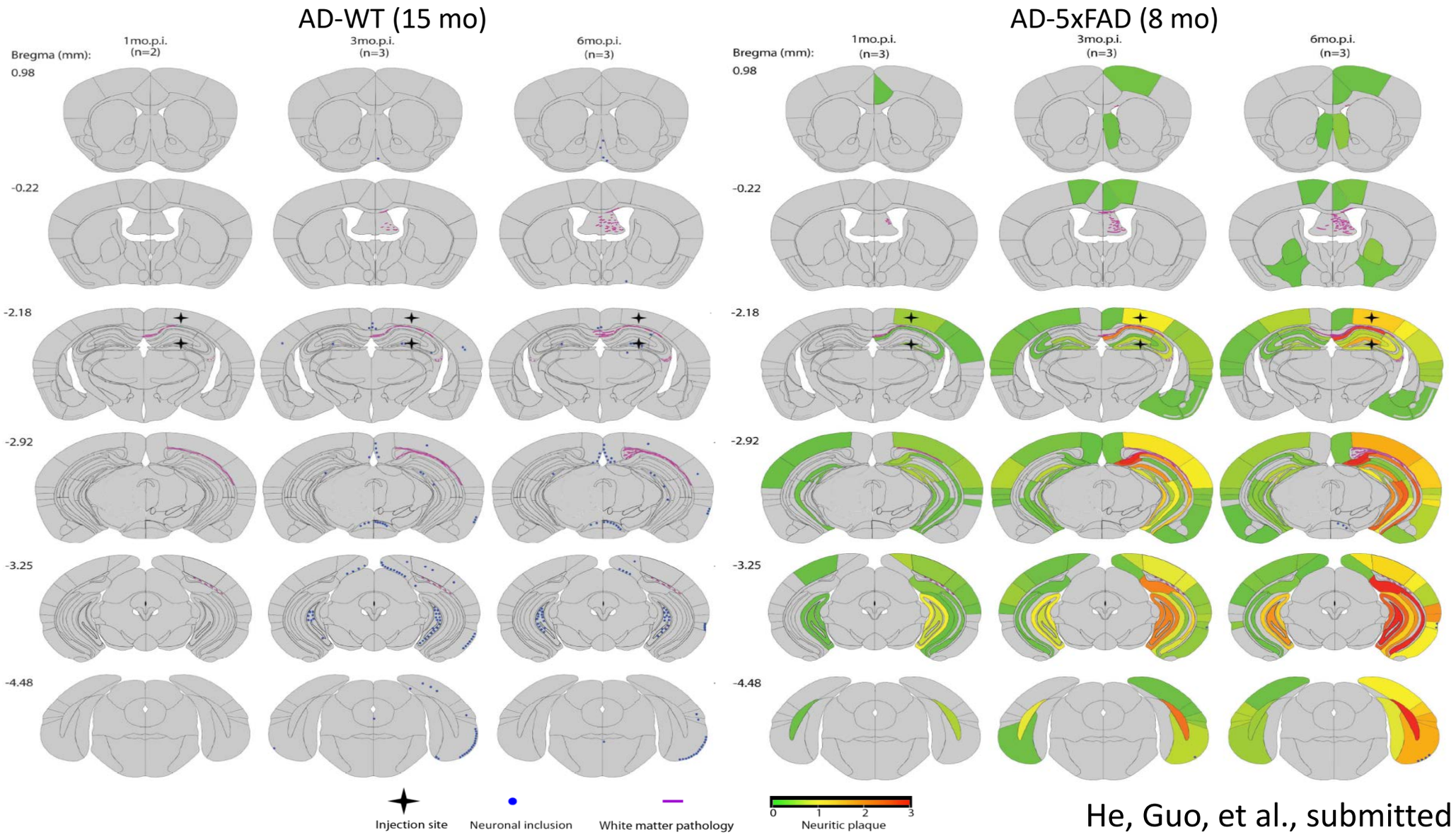
AT8





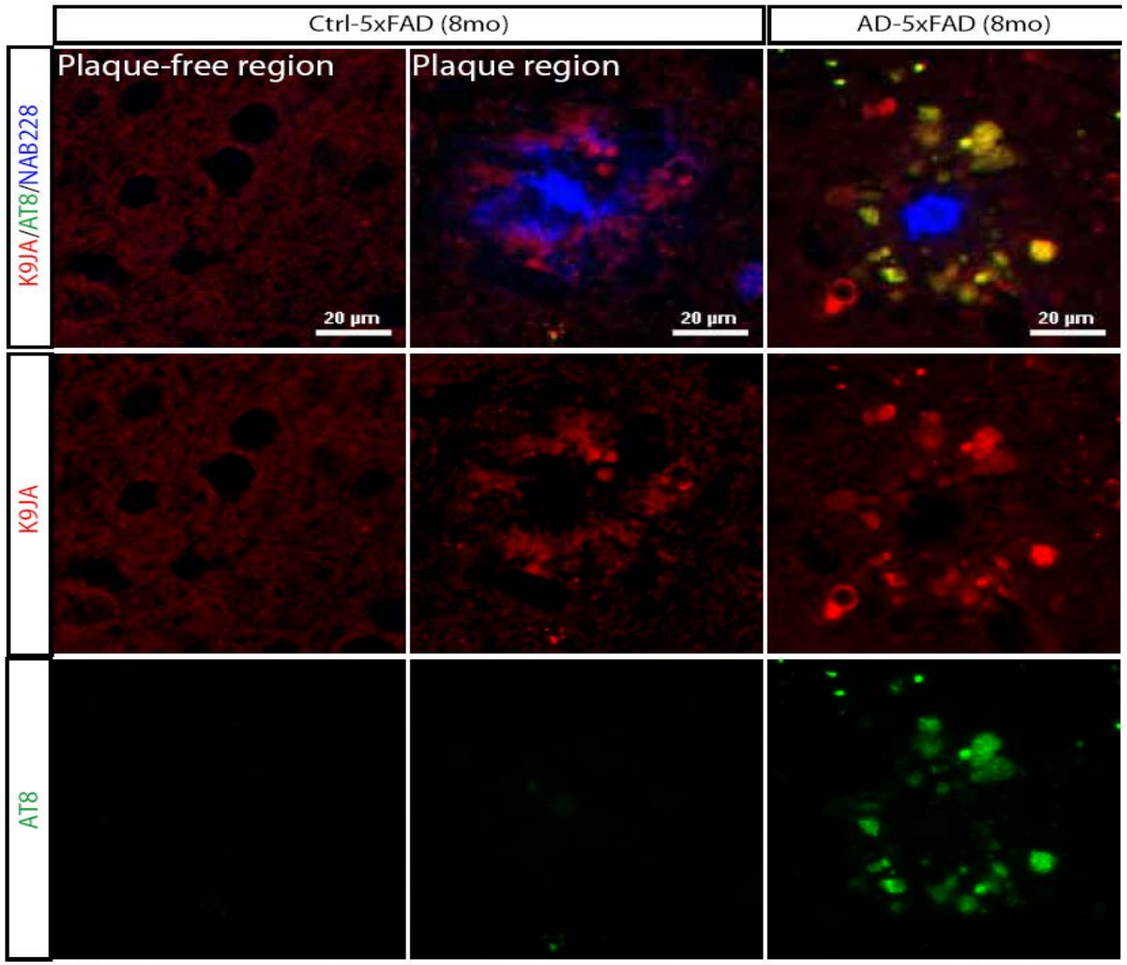
He, Guo, et al., submitted

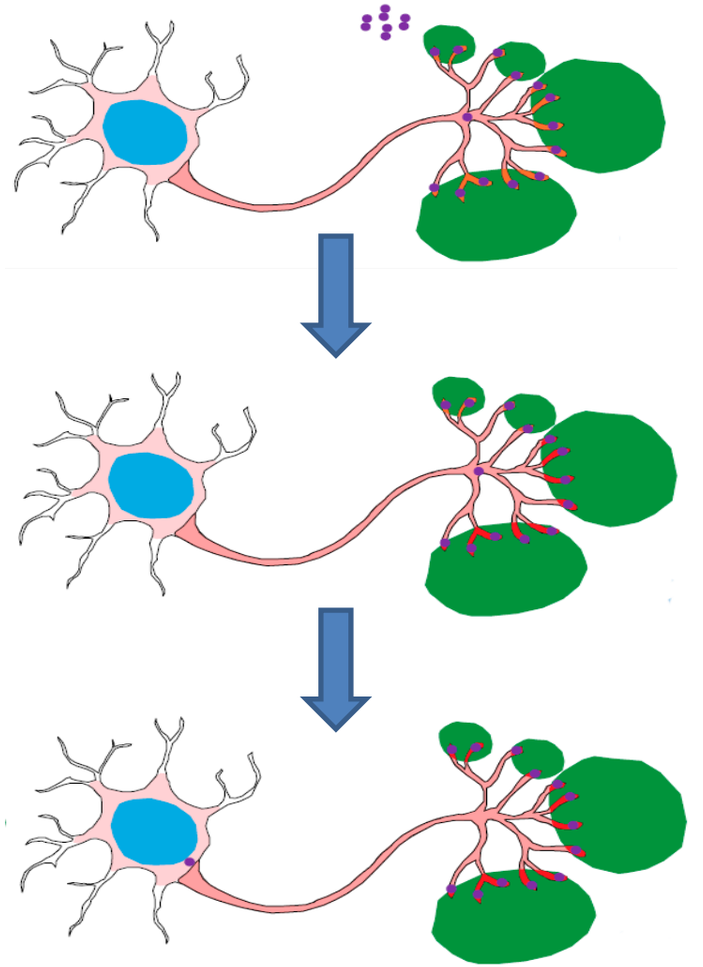
Spatiotemporal development of NIs and NPs



He, Guo, et al., submitted

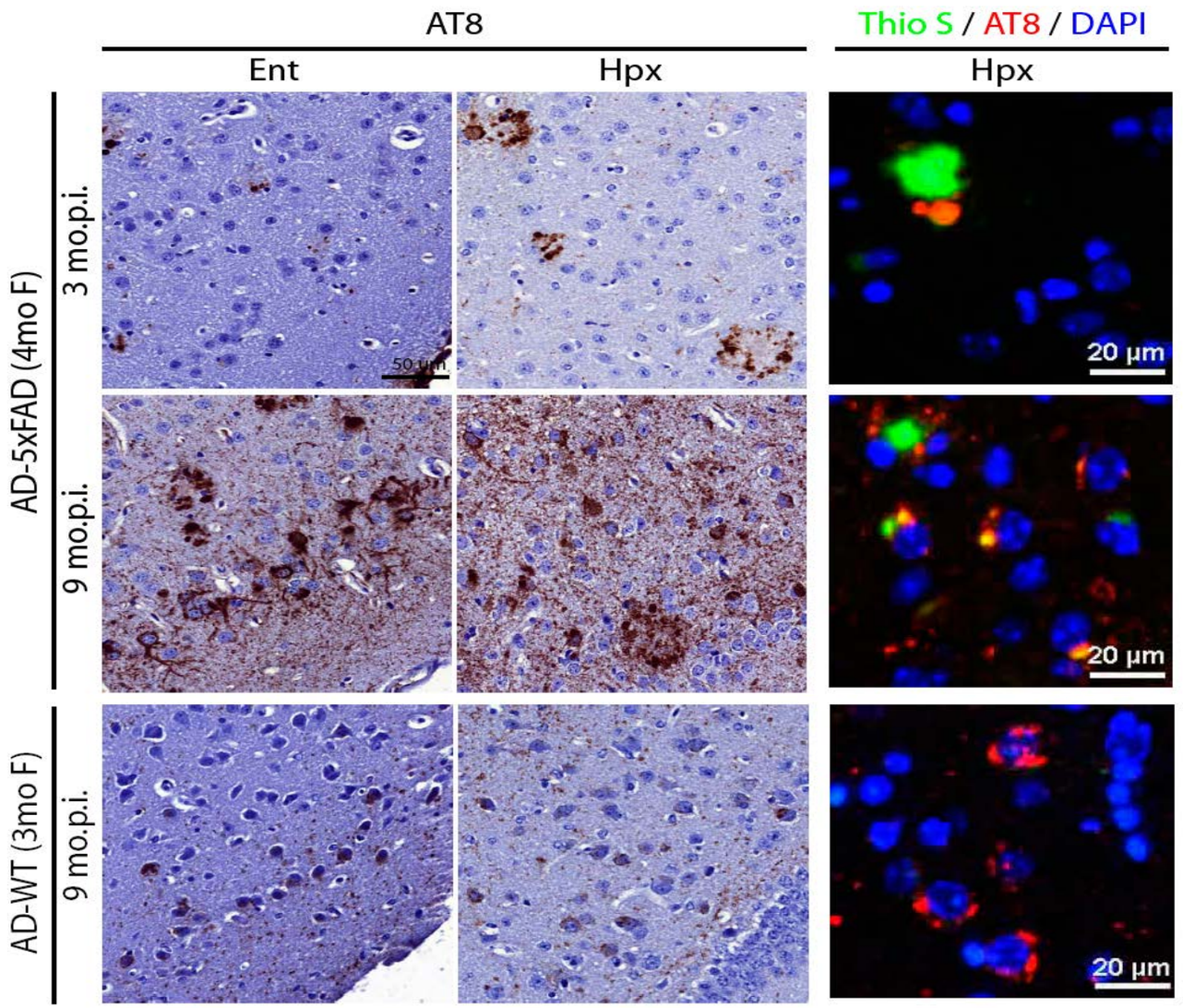
Primed tau aggregation at the peri-plaque dystrophic neurites

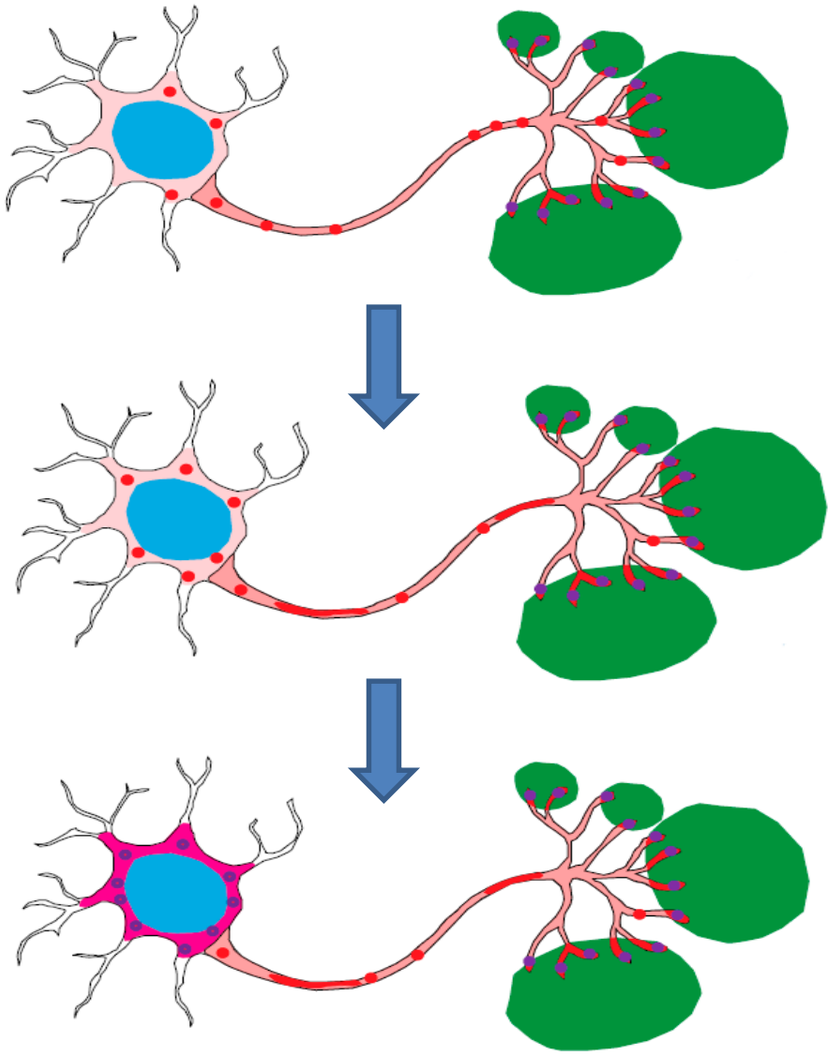




He, Guo, et al., submitted

Indirect enhancement of NIs and neuropil threads by Aβ plaques





He, Guo, et al., submitted

Summary for part B

- Aβ plaques promote the induction, maturation and propagation of tau pathology in animals with endogenous expression levels of tau.
- Consistent with neuropathological data and recent imaging studies
- Support a modified version of amyloid cascade hypothesis
- Two-step mechanism of Aβ-mediated tau pathogenesis :

(1) Enhanced seeded aggregation of tau in the peri-plaque dystrophic neurites → NP formation.

(2) NPs promote neuropil threads and NFT-like tau pathology through secondary seeding.

Outline

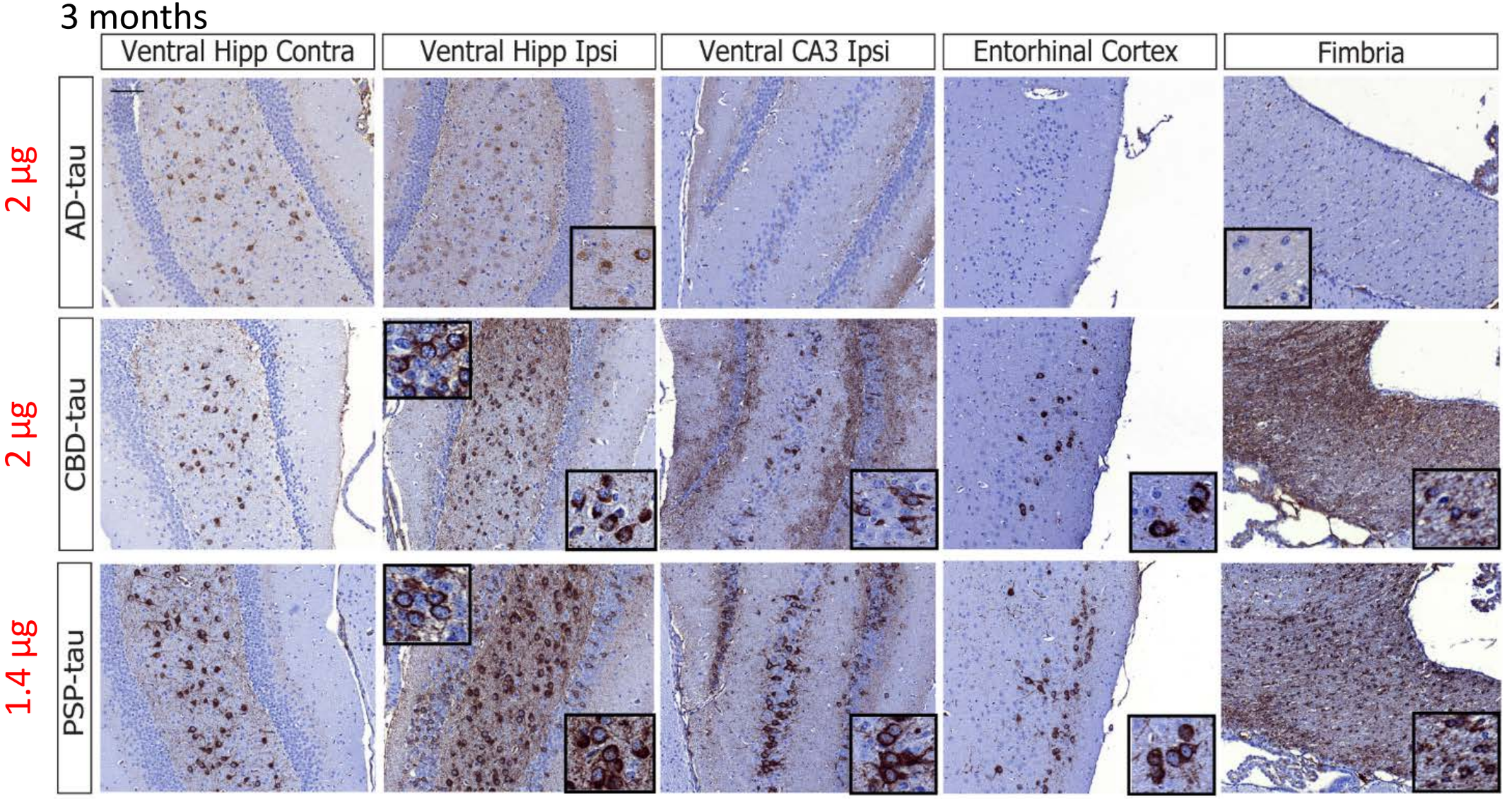
A. Generating a mouse model of sporadic tauopathies using AD brain-derived tau aggregates
B. Exploring the effects of Aβ plaques on tau pathology in plaque-bearing mice
C. Characterizing pathology induced by tau aggregates purified from different tauopathies

Heterogeneities in tauopathies

	Isoform composition of tau aggregates	Brain region affected	White matter pathology	Cell type involvement	Major symptoms
AD	3R and 4R tau at 1:1	Mainly cortical	No	Neurons	Cognitive impairment
CBD	Predominantly 4R tau	Cortical and subcortical	Extensive	Neurons + glia	Motor with associated cortical dysfunction
PSP	Predominantly 4R tau	Mainly subcortical	Moderate	Neurons + glia	Motor deficits

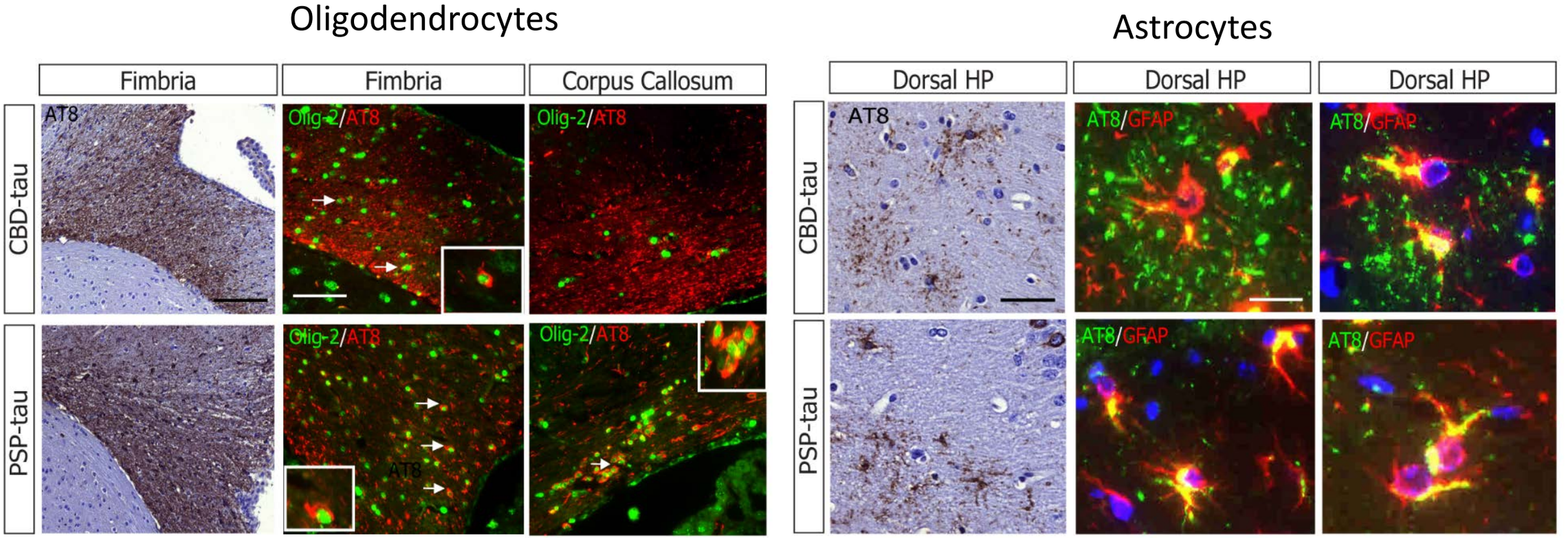
Caused by different strains of tau aggregates developed in different tauopathies?

Distinct patterns of tau pathology induced in WT mice by pathological tau purified from different tauopathies



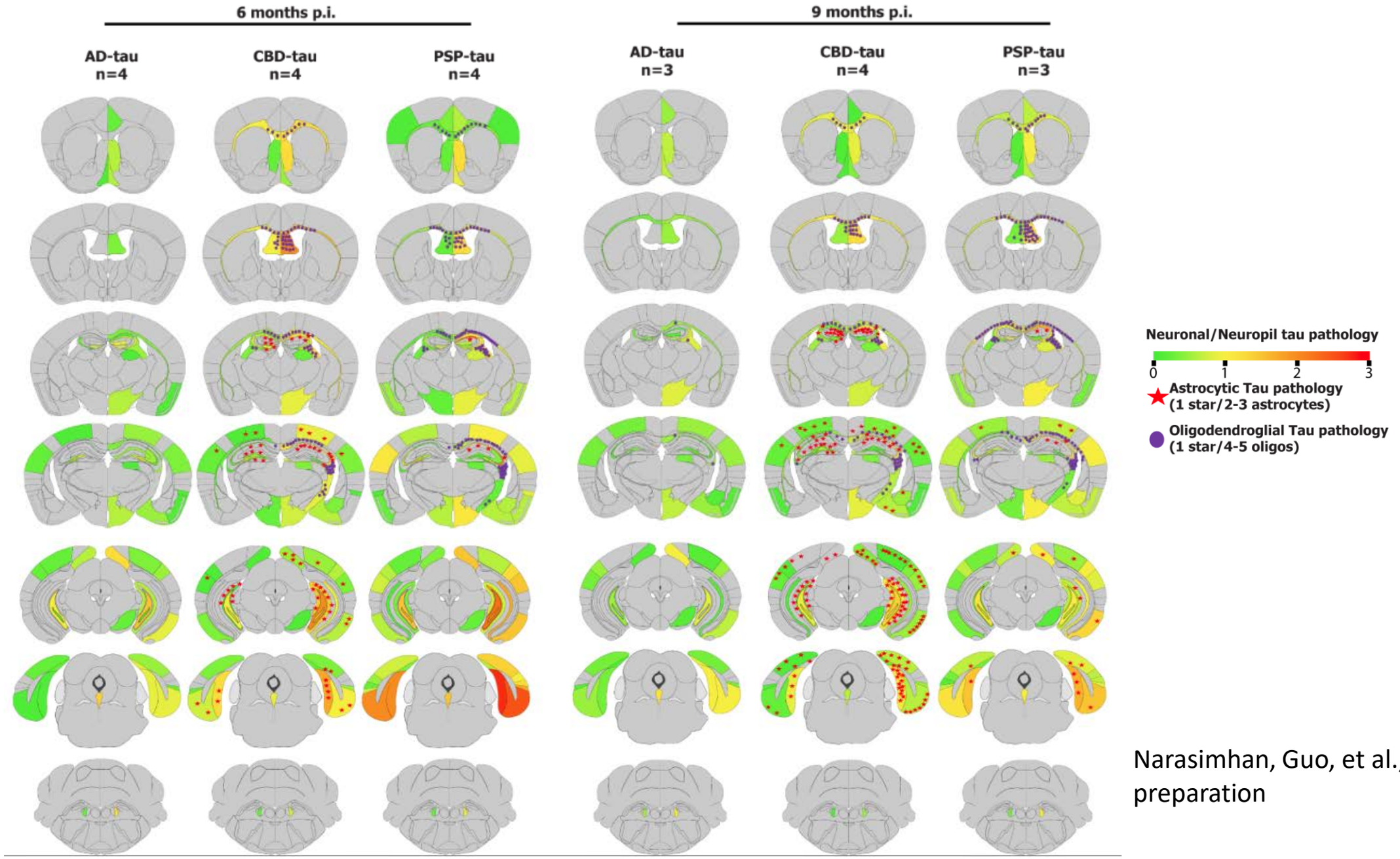
Narasimhan, Guo, et al., in preparation

Double-labeling for glial tau pathology



Narasimhan, Guo, et al., in preparation

Distribution of different types of tau pathology



Narasimhan, Guo, et al., in preparation

Summary for part C

- Tau aggregates developed in different tauopathies are of different strains.
- Cell type-specific development of tau pathology is dictated by pathological tau strains.
- Distribution of neuronal tau pathology is primarily determined by initiation sites and their connectome.

Acknowledgment

- Virginia Lee and John Trojanowski
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- Terry Schuck
- Bryan Zoll
- Joshua Daniels
- Soo-Jung Kim
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