DOD State-of-the-Science (SoS) Meeting

Cis P-tau directly links TBI to CTE and AD, but can be blocked by antibody

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Beth Israel Deaconess Medical Center

Conflict of Interest Disclosure

Drs. Ping Lu and Xiao Zhou are inventors of Pin1 technology, which was licensed by BIDMC to Pinteon. Both Dr. Lu and Dr. Zhou own equity in, and consult for, Pinteon. Their interests were reviewed and are managed by BIDMC in accordance with its conflict of interest policy.

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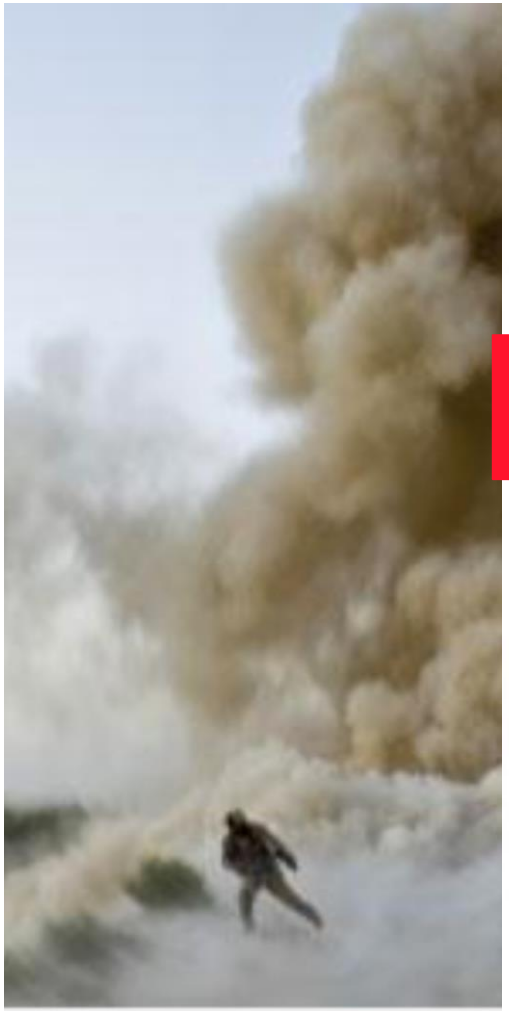
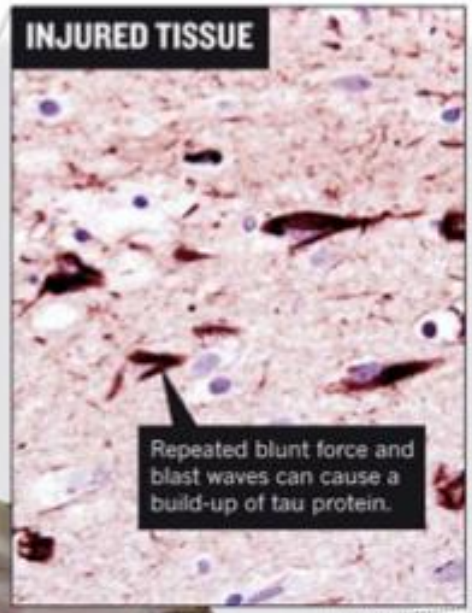
Tau tangles are a hallmark of CTE brains in veterans

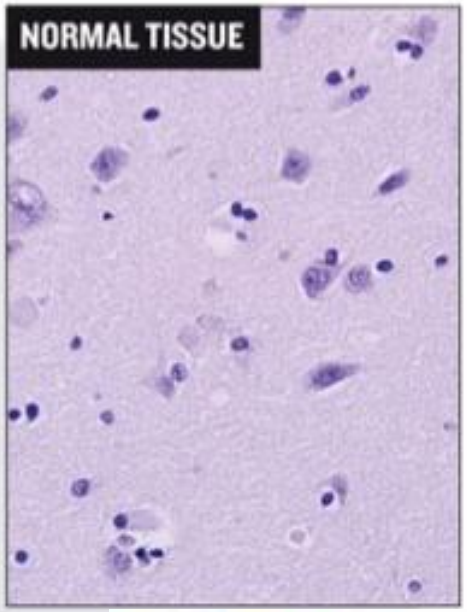


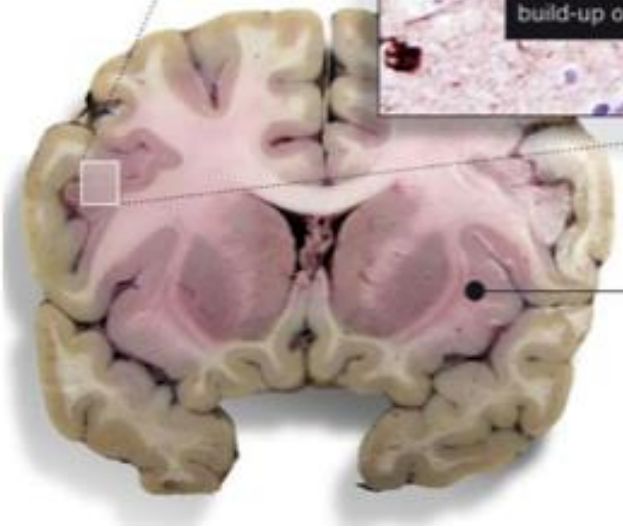
TRAUMA IN THE BRAIN

Brains affected by blunt force and blast waves can show few outward signs of injury. But a microscope reveals neurological

similar to those found in Alzheimer's disease.







The brain shows swelling, but no gross deformities.

Weinberger, 2011, Nature
Mckee, 2013, Brain

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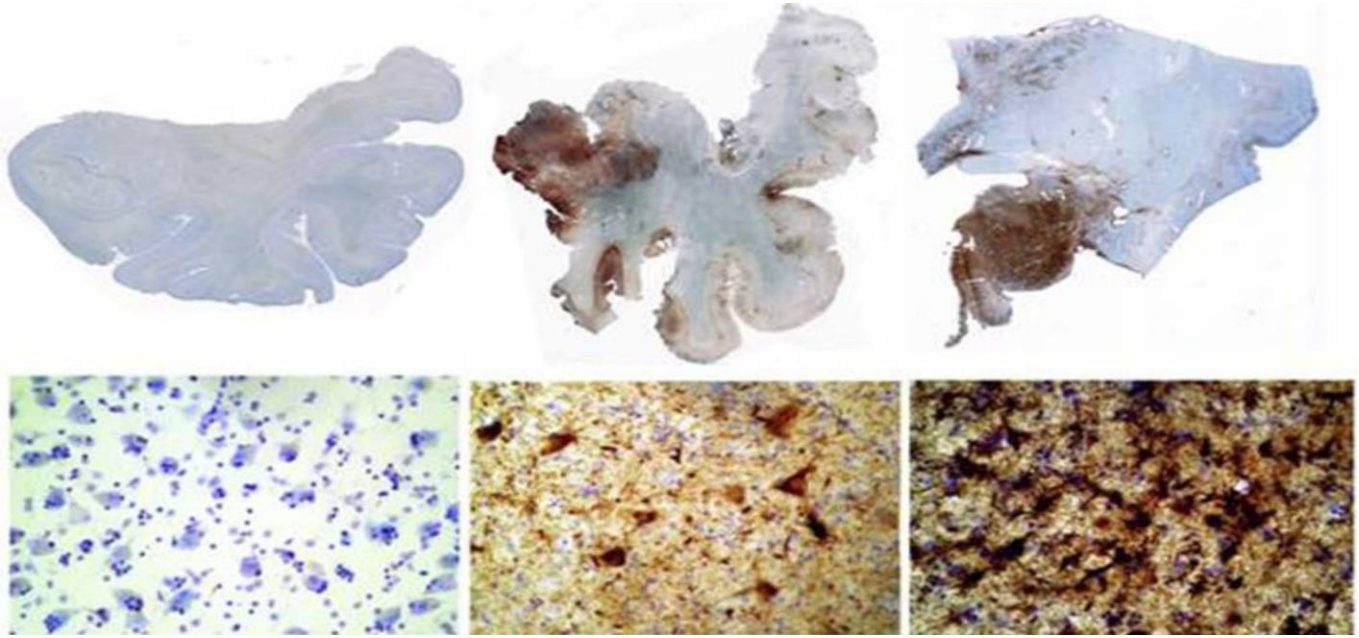
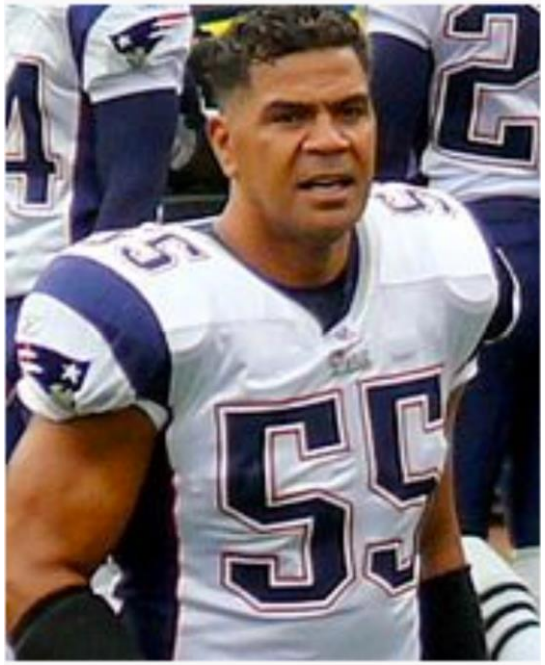
Tau tangles are a hallmark of CTE brains in athletes

Healthy Brain **NFL player** **Boxer** **Junior Seau**





Mckee et al., 2009, J. Neuropathol Exp Neurol

Star in Chargers and Pats
Suicide due to CTE

- **Molecular profiles of tau tangles from boxer CTE and AD are the same (Schmidt et al., Acta Neuropathol, 2001)**

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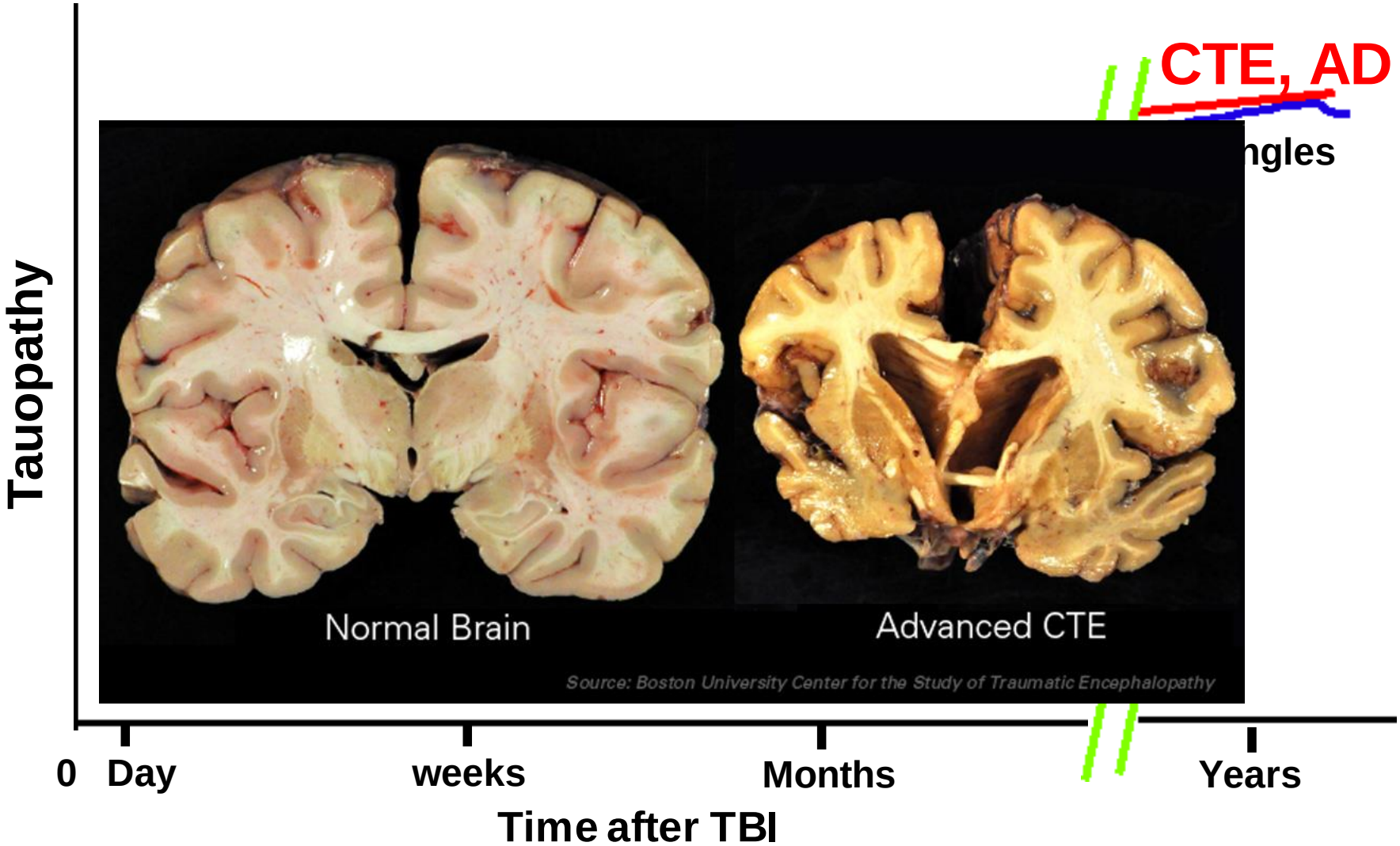
However, no tau pathology in acute TBI brains
No direct evidence linking TBI to AD and CTE



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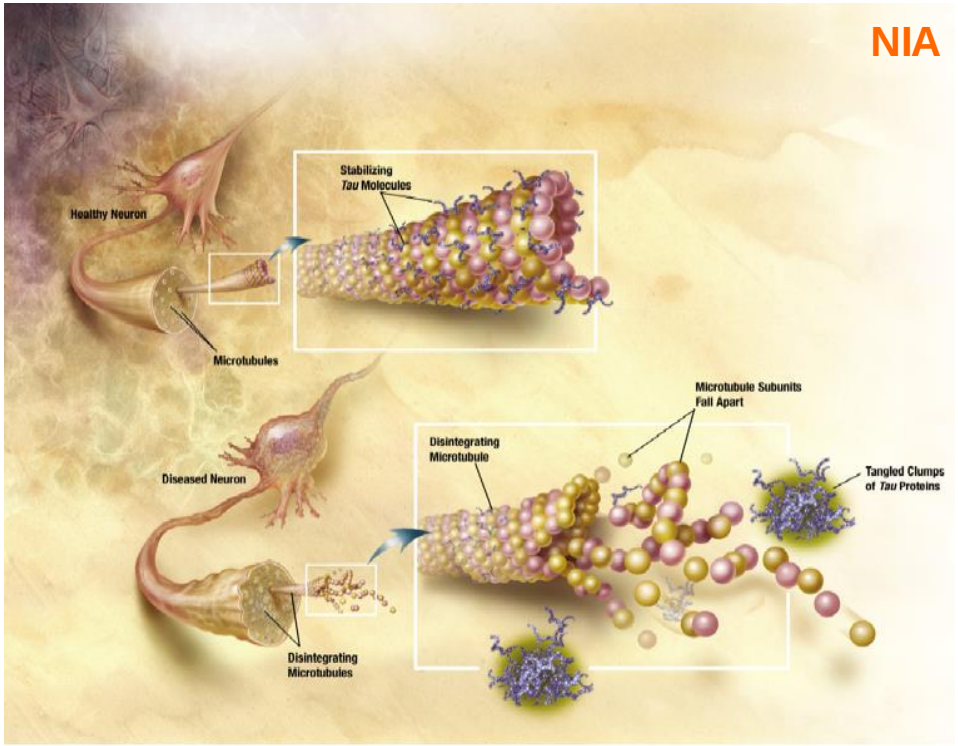
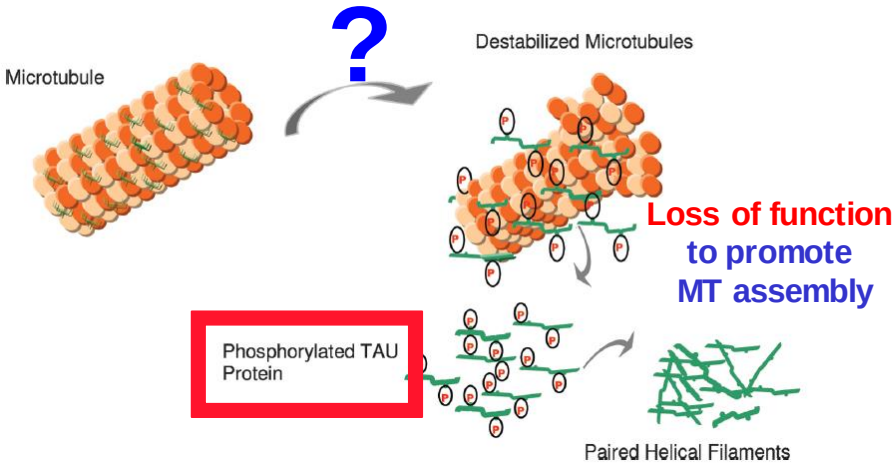
Kondo et al., 2015, Nature

WHAT DO WE LEARN FROM STUDYING TAU IN ALZHEIMER'S DISEASE?

Tau is critical for normal brain function, but misshapen in AD





Loss of function to promote MT assembly

Toxic gains-of-function (p-tau aggregates and PHFs)
1) Sequester normal tau
2) Obstacles to cargo transport

Ballatore et al., 2007, Nature Rev Neurosci;
Paula et al., 2009, Dement & Neuropsychol

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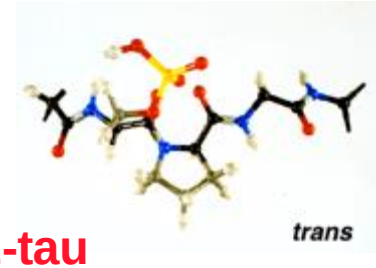
cis p-tau is the early pathogenic species, a promising biomarker and a therapeutic target in AD

trans **(biological)**
(Internal control)

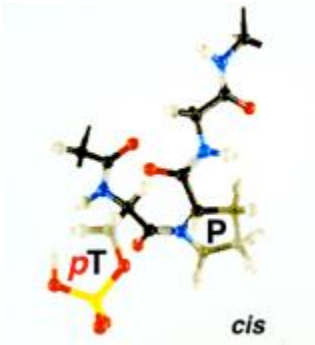


pT231-tau

AD with aging →

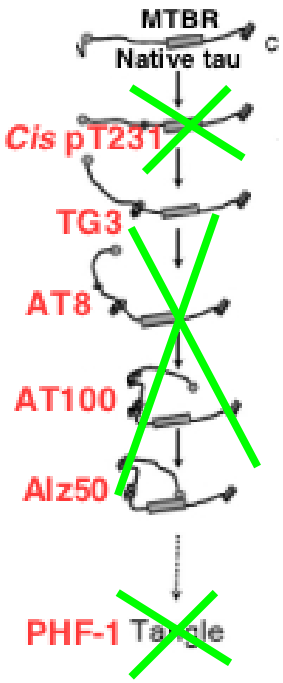
← **+Pin1**

cis **(pathogenic)**
(Biomarker)



cis **Antibody**

→ **Vaccine**



Microtubule assembly	Yes	No
Dephosphorylation	Yes	No
Protein degradation	Yes	No
Protein aggregation	No	Yes
Early appearance	No	Yes
Pathogenic localization	No	Yes
Neurodegeneration	No	Yes
Toxic p-tau spread	No	Yes?

↓ **Healthy neuron**

↓ **AD**

↓ **Treat AD at early stages**

Nakamura et al., Cell, 2012
Lim et al., JCI, 2008
Pastorino et al., Nature, 2006
Liou et al., Nature, 2003
Lu et al., Nature, 1999

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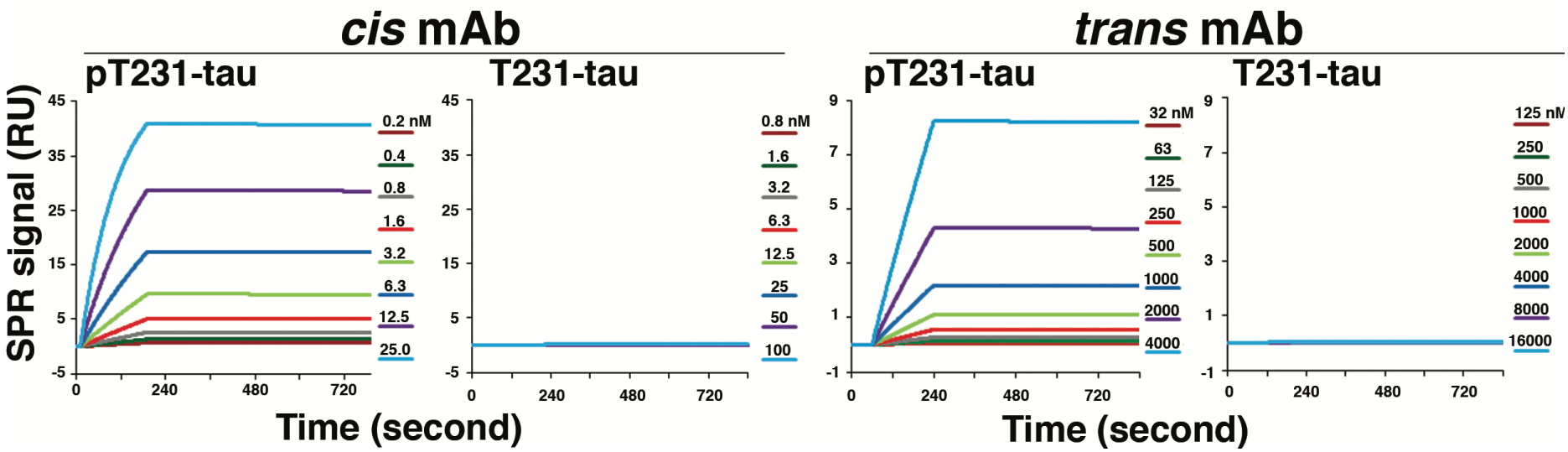
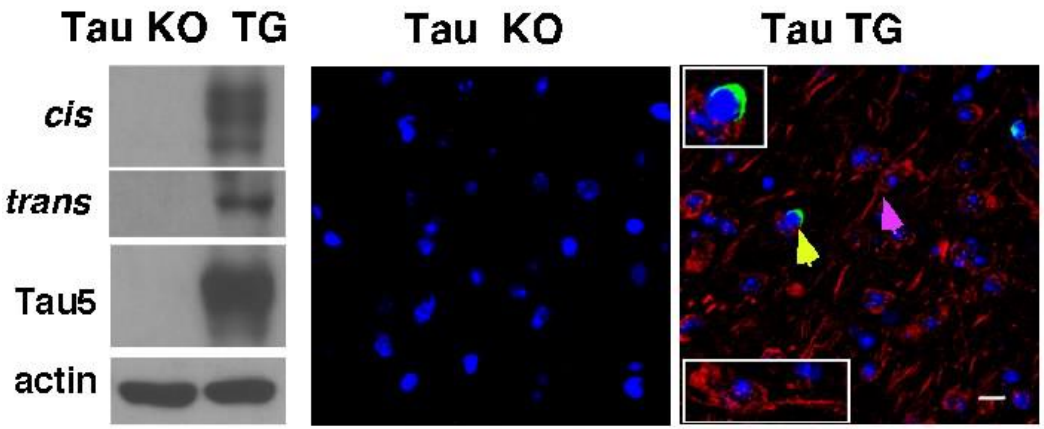
Luna-Munoz et a., JAD, 2007

Development of *cis* and *trans* P-tau mAbs



mAb	Peptide	Ka (Ms^{-1})	Kd (s^{-1})	K_D (nM)
cis	pT231-tau	40700	1.10×10^{-5}	0.27
cis	T231-tau	0.17	1.00×10^{-5}	58820
trans	pT231-tau	250	1.05×10^{-5}	42.0
trans	T231-tau	4.5	1.95×10^{-5}	4333



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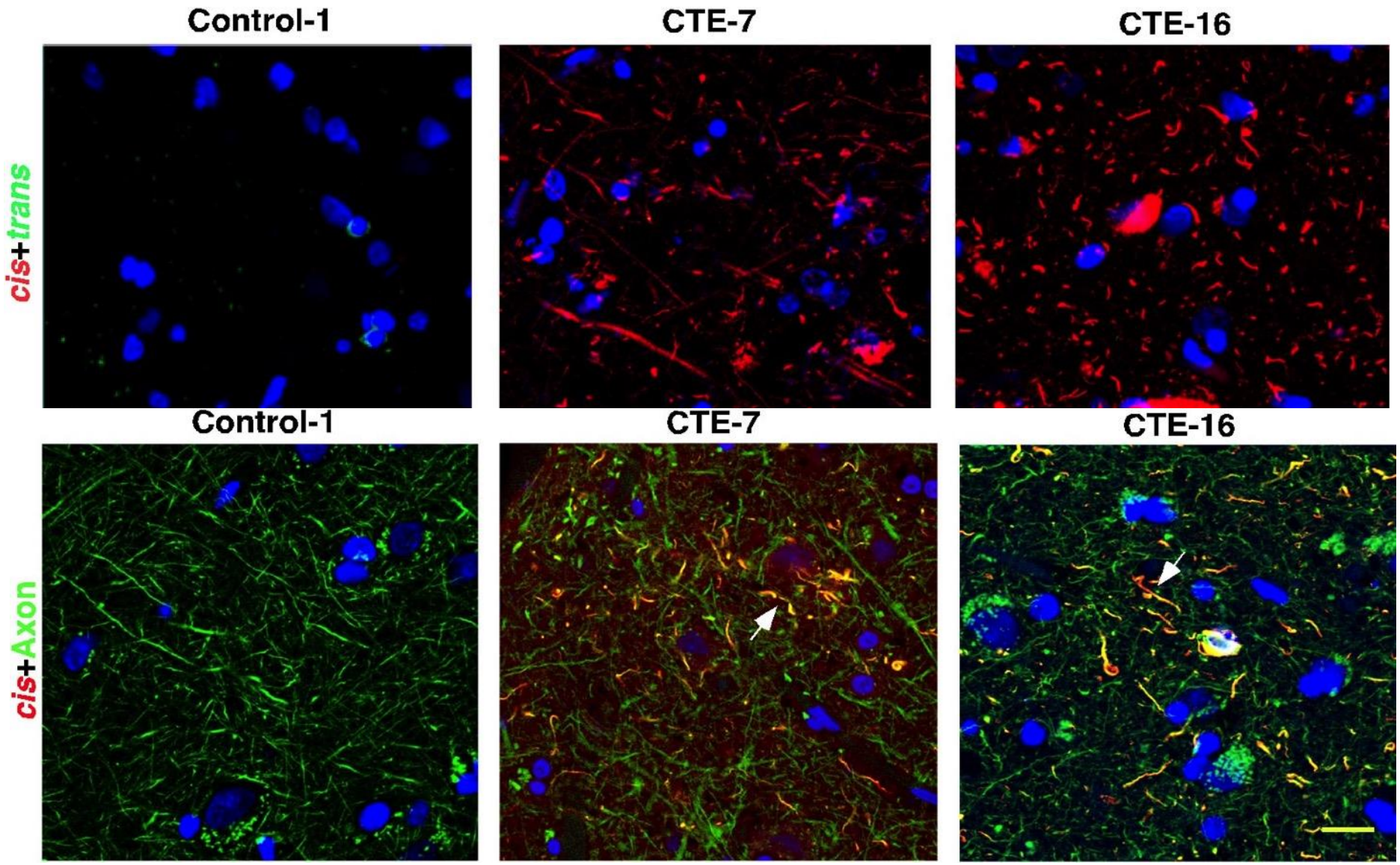
Kondo et al., 2015, Nature

Robust *cis* P-tau in human AD and CTE brains



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Kondo et al., 2015, Nature

When does *cis* P-tau appear after TBI?

- **Experimental models of TBI in mice to model most common TBI**
  - **Closed and open field blast-induced TBI to mimic military TBI**
  - **Impact-induced TBI to mimic sport TBI (range from mild to severe)**

- **Experimental models of TBI in cell culture**
  - **Neuron stress**

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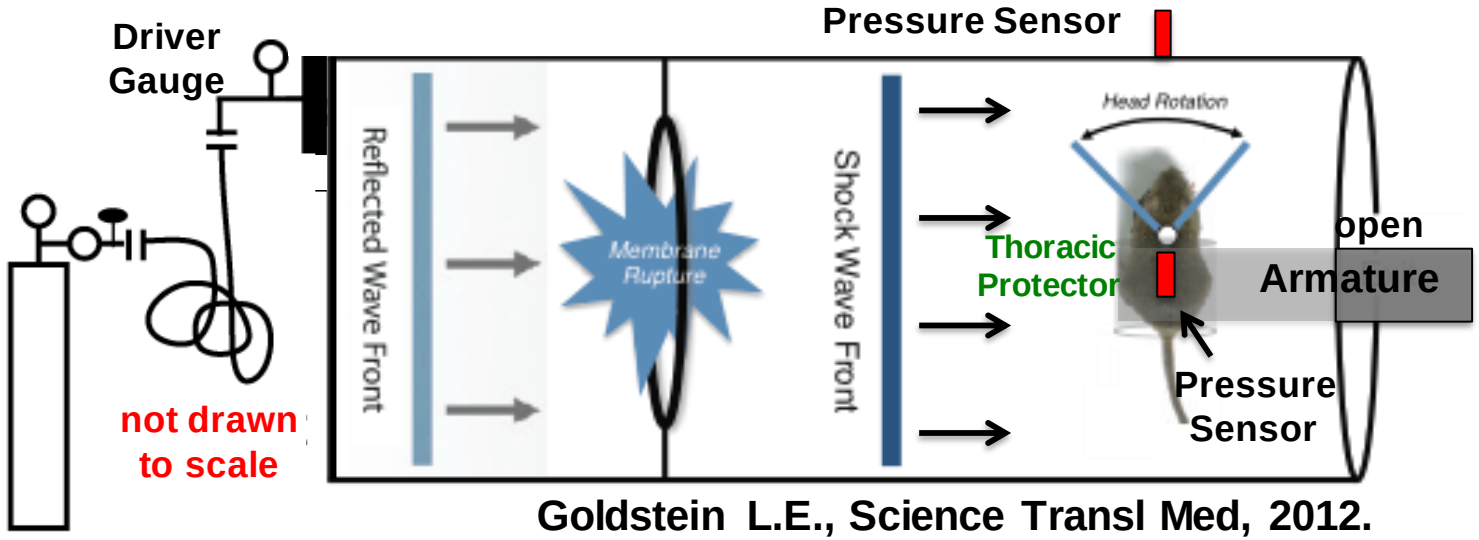
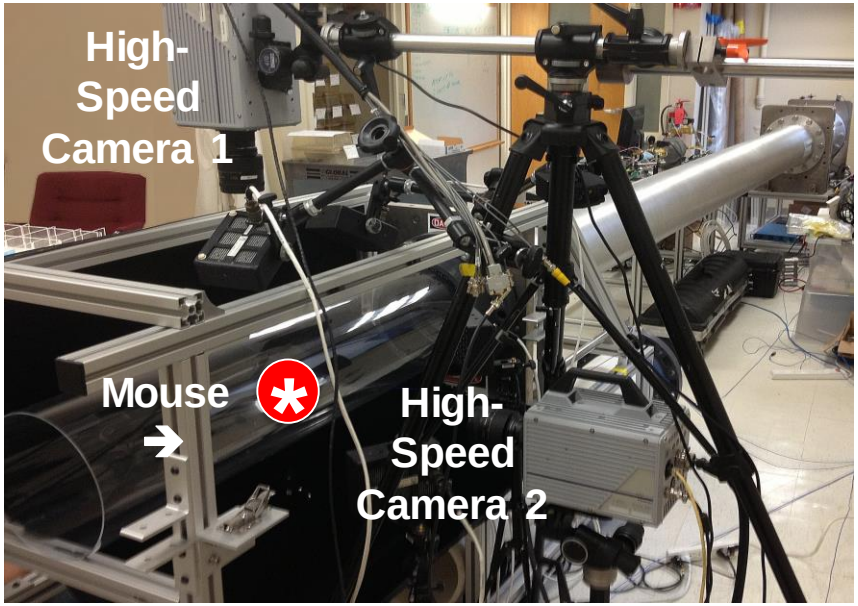
Kondo et al., 2015, Nature

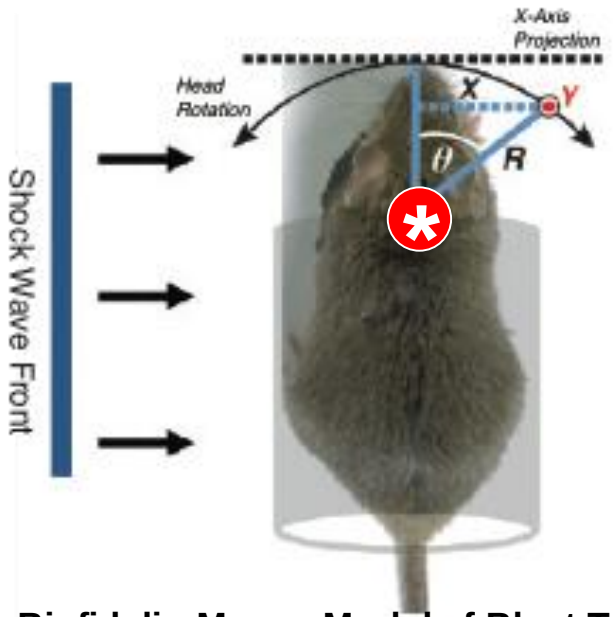
High pressure blast TBI model to mimic military TBI



Goldstein L.E., Science Transl Med, 2012.

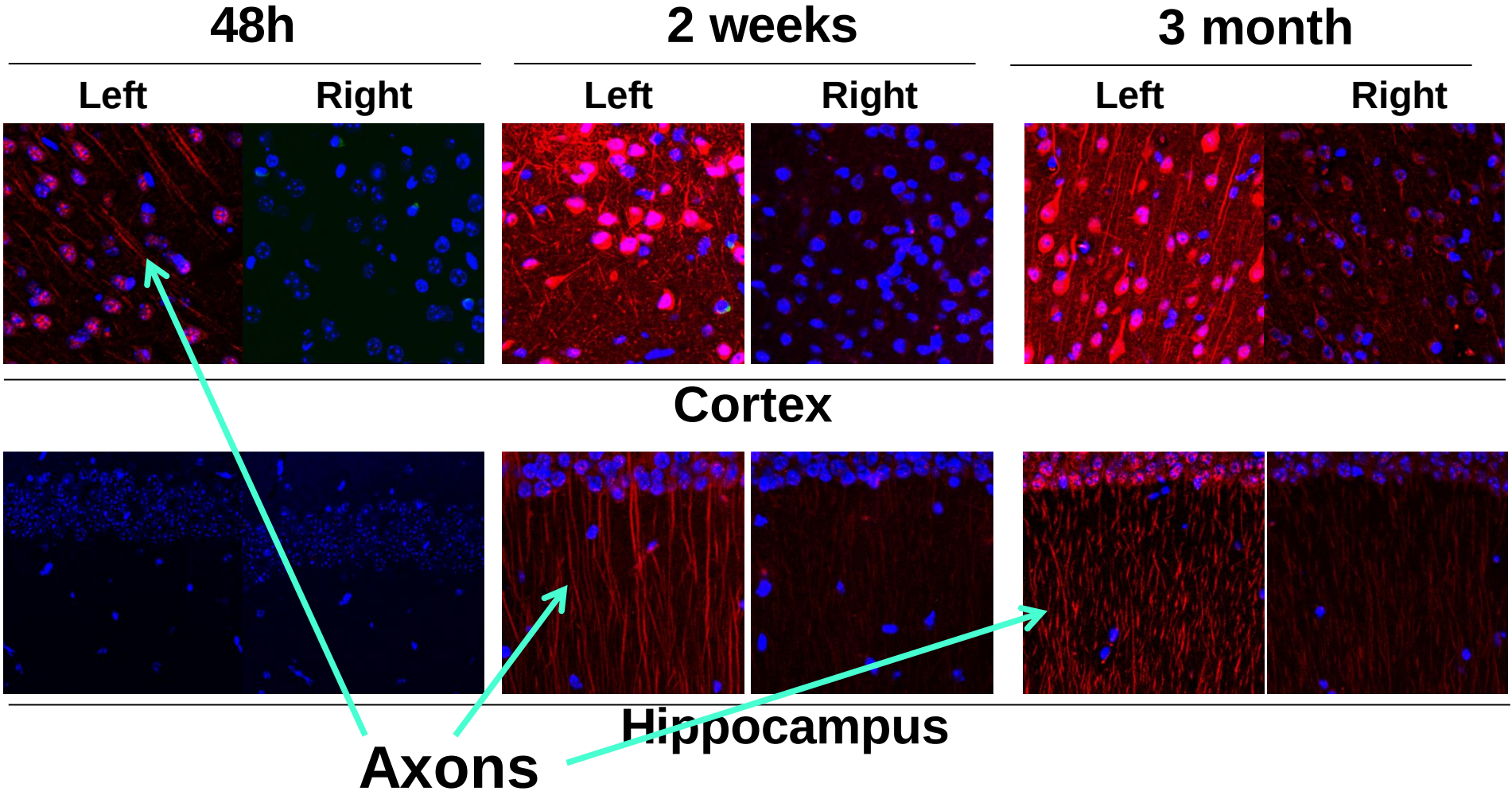


Blast Tube + High-Speed Videography t =1 μsec



Biofidelic Mouse Model of Blast TBI-CTE

High pressure blast induces profound, persistent and spreadable *cis* P-tau



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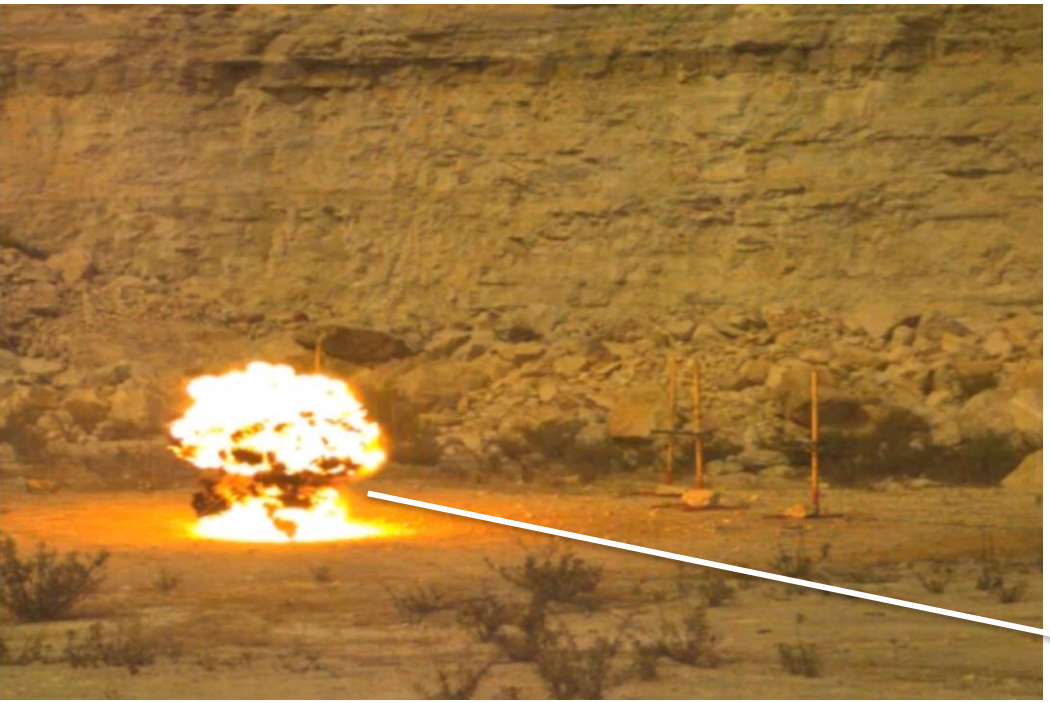
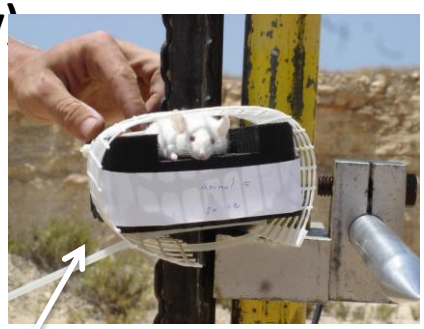
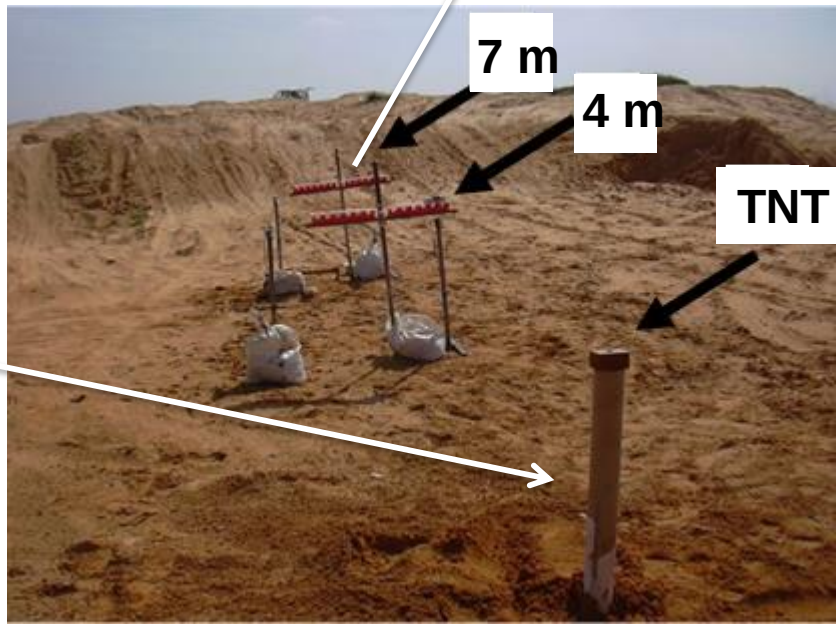
Kondo et al., 2015, Nature

Open Field Blast TBI

Anesthetized mice are positioned 4 and 7 m distance from a controlled open field detonation (500 g TNT generating 5.5 and 2.5 PSI pressure, respectively)







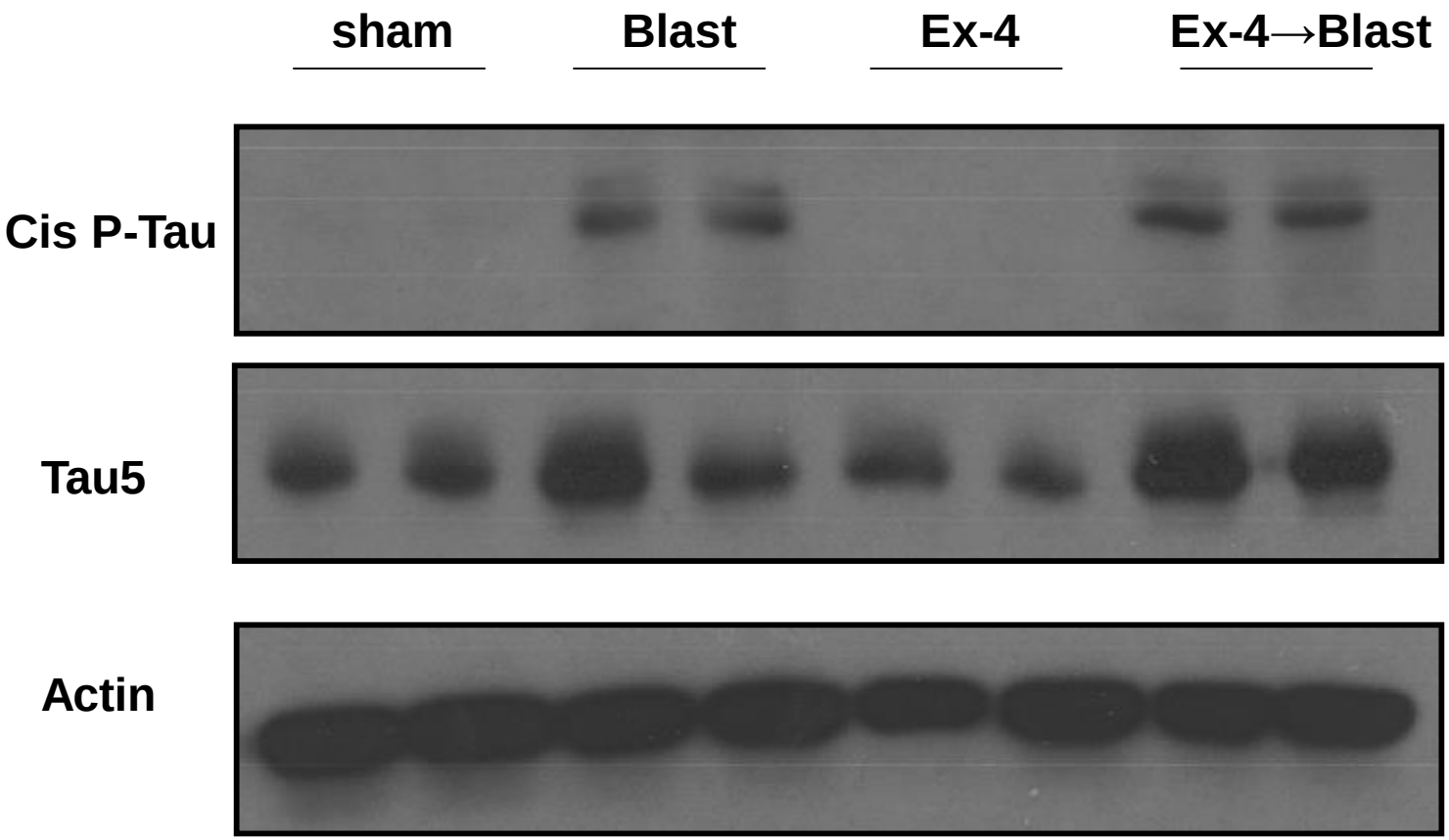
Rubovitch et al., *Exp Neurol*. 232:280-9, 2011
Tweedie et al., *Alzheimers Dement* 2015 [Epub ahead of print]

- No acute alterations in basic neurological assessment or brain gross pathology.
- 72 h Immunohistochemistry: early inflammation and diffuse neuronal cell loss throughout brain
- 72 h Gene array: oxidative stress, inflammation
- 7 & 30 days: cognitive and behavioral impairments at 7 and 30 days post blast.
- 14 days Gene array: Alzheimer's and dementia related pathways up regulated
- 30 days: MRI changes altered BBB permeability evident

Collaboration with
Chagi Pick, Tel Aviv Univ
Nigel Greig, NIH

Cis P-tau is robustly induced in open field blast TBI

48h later the Blast with TNT



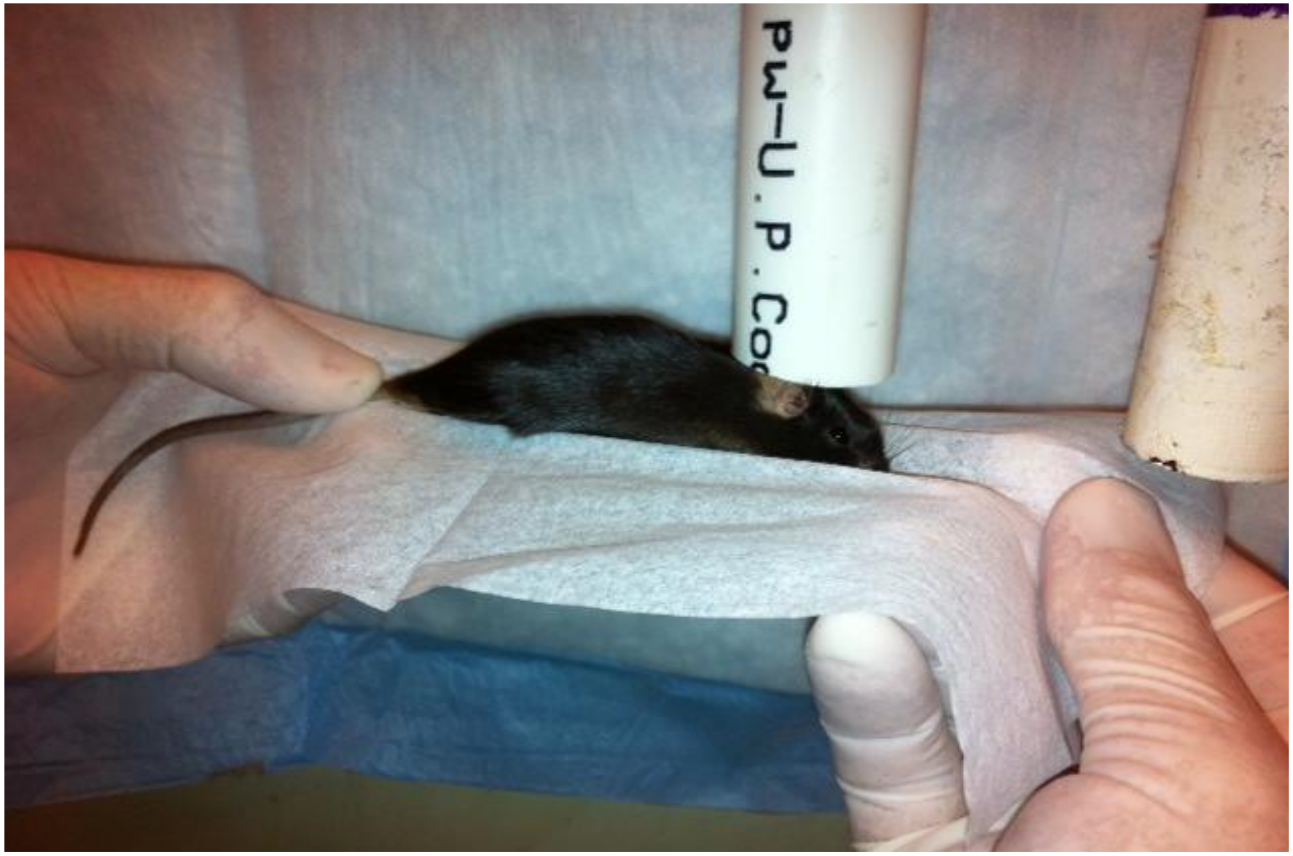
Ex-4 is a peptide and work as a Glp-1 receptor agonist.

Collaboration with
Chagi Pick, Tel Aviv Univ
Nigel Greig, NIH

Low speed impact TBI model to mimic sport TBI

Low speed impact • No skull deformation • No brain hemorrhage

• Injury severity dependent on weight drop and its height



Developed by Mannix and Meehan at Children's Hospital (Ann Neurol, 2013)

Flier et al., Nature Protocols, 2009

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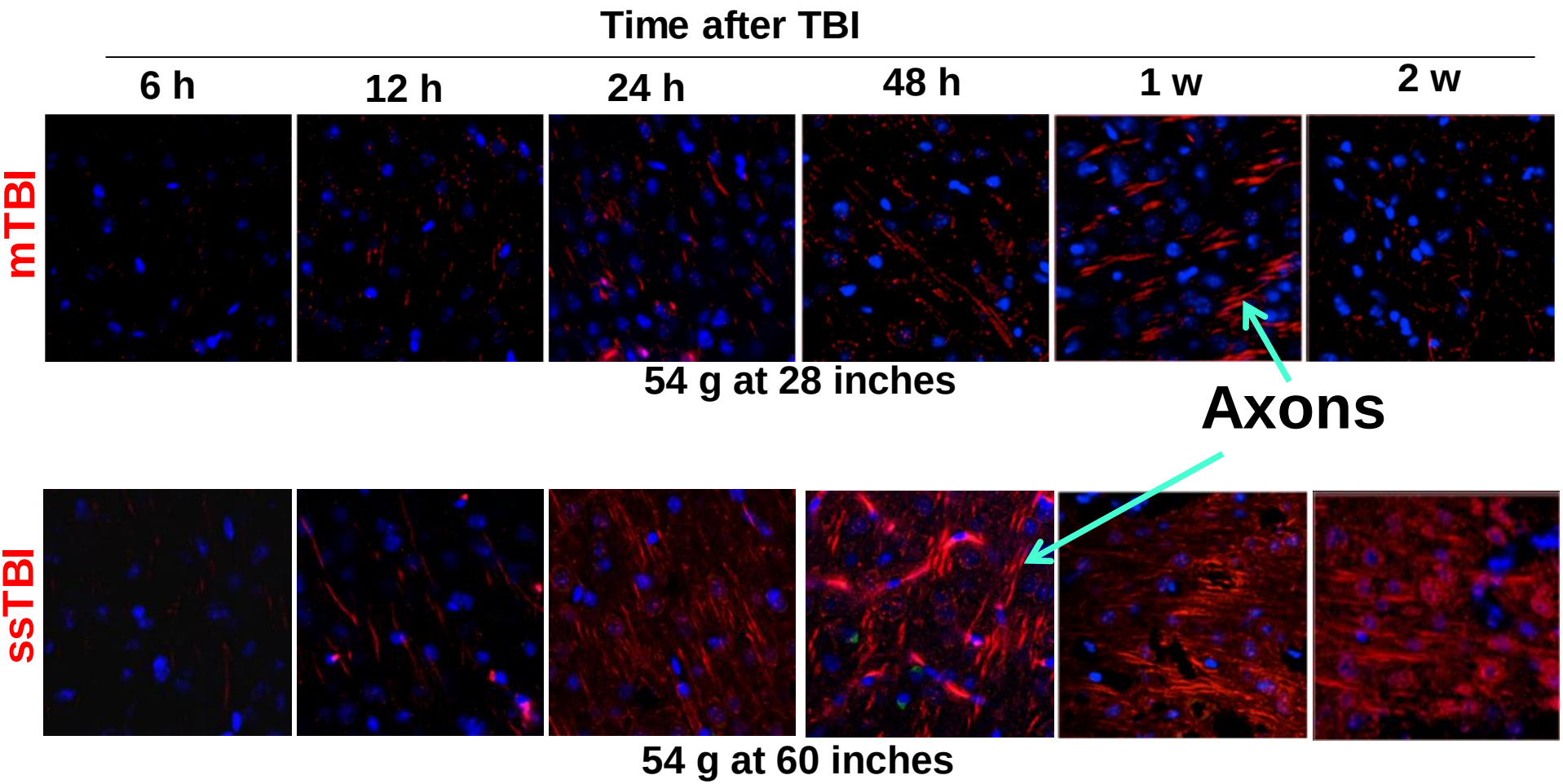
Whereas mTBI moderately and transiently, ssTBI robustly and persistently induces *cis* P-tau



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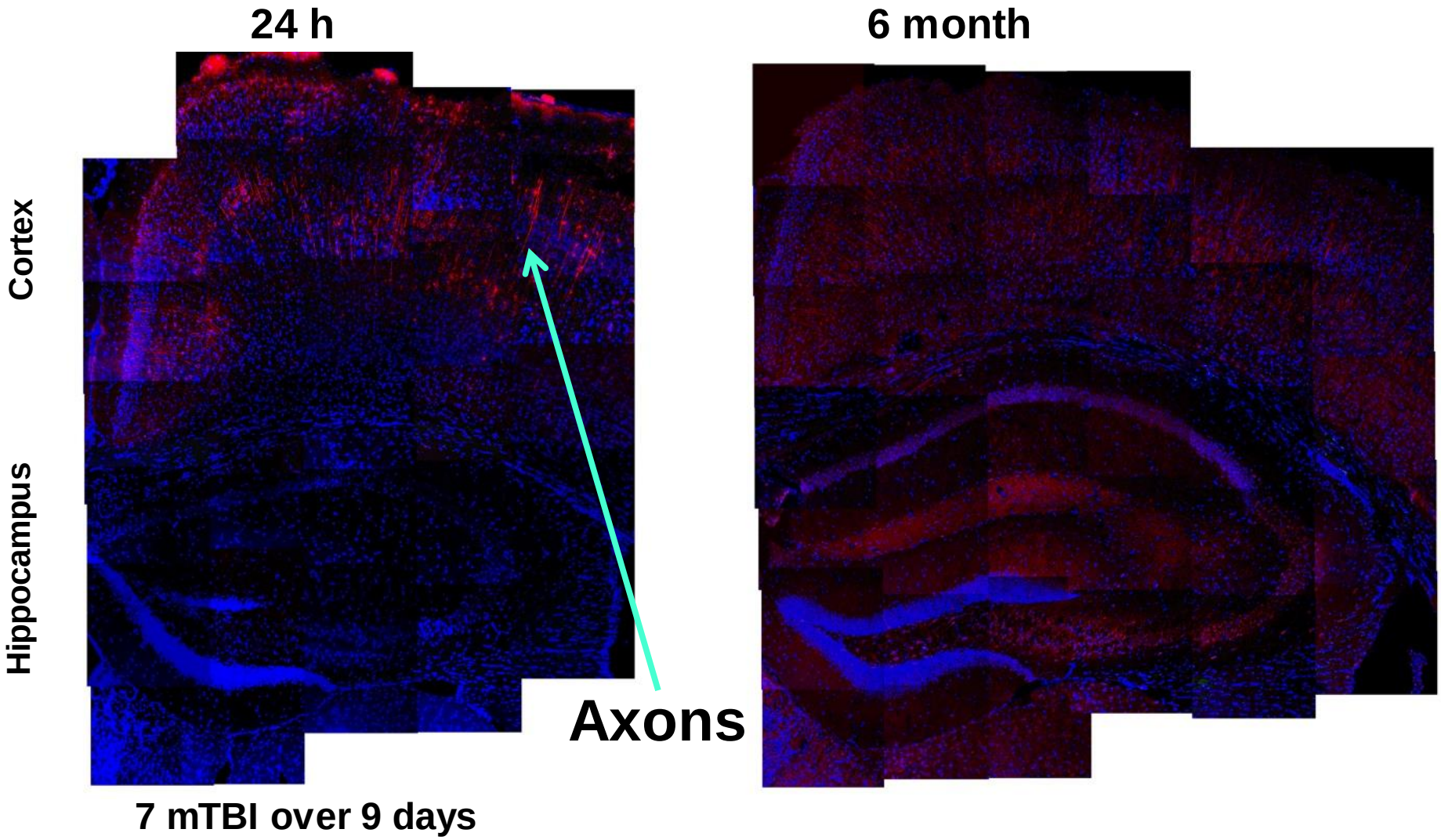
Kondo et al., 2015, Nature

Repetitive mTBI robustly and persistently induces *cis* P-tau, spreading to deeper brain regions with time



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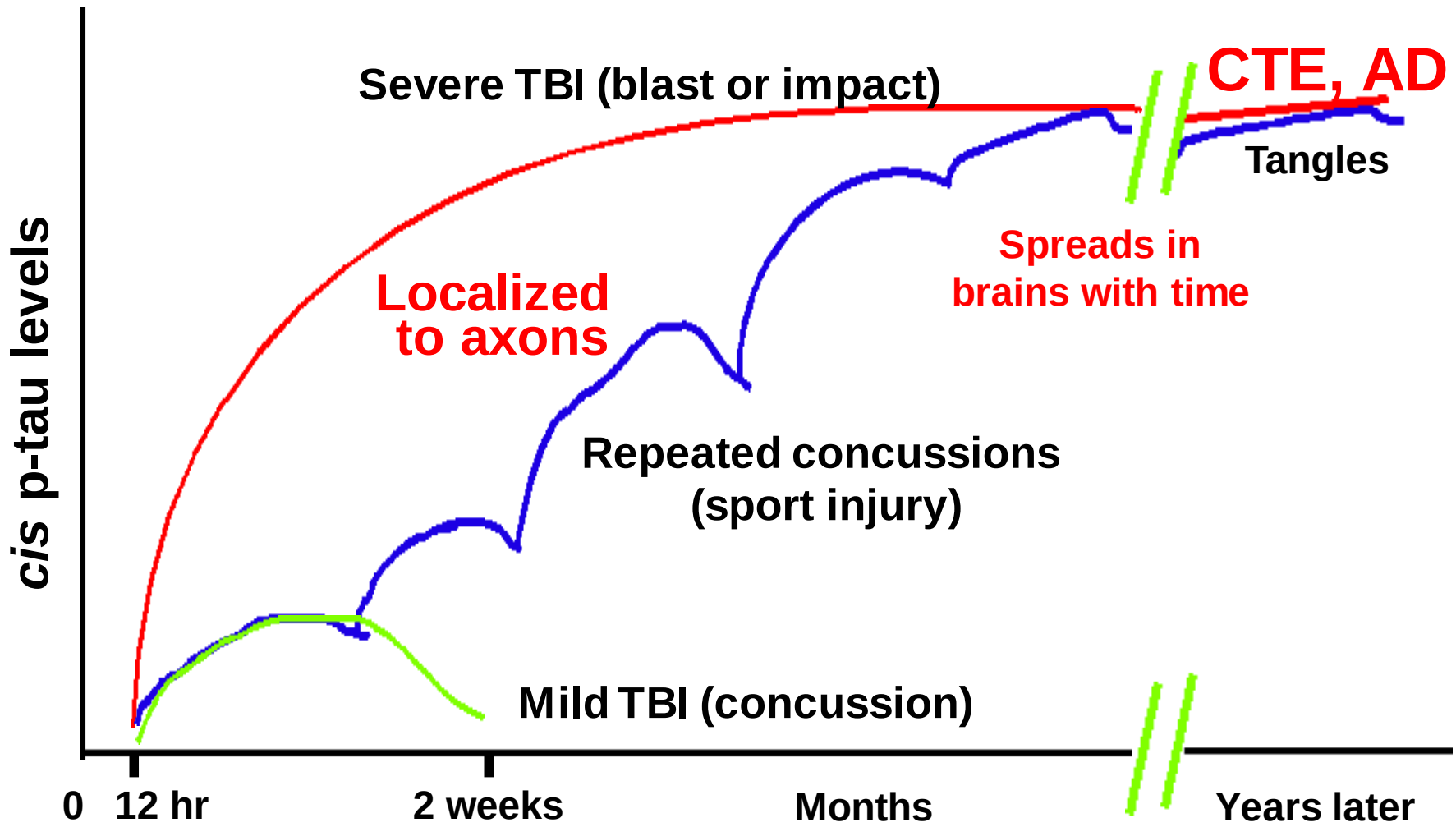
Kondo et al., 2015, Nature

Kinetics of *cis* p-tau induction after different TBIs



"Over the past 70 years, diffuse axonal injury (DAI) has emerged as one of the most common and important pathological features of traumatic brain injury (TBI)"
(Douglas Smith, 2012, Exp Neurol)

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Kondo et al., Nature, 2015

IS *CIS* P-TAU NEUROTOXIC? IS *CIS* P-TAU STOPPABLE? (IN CELL CULTURE)

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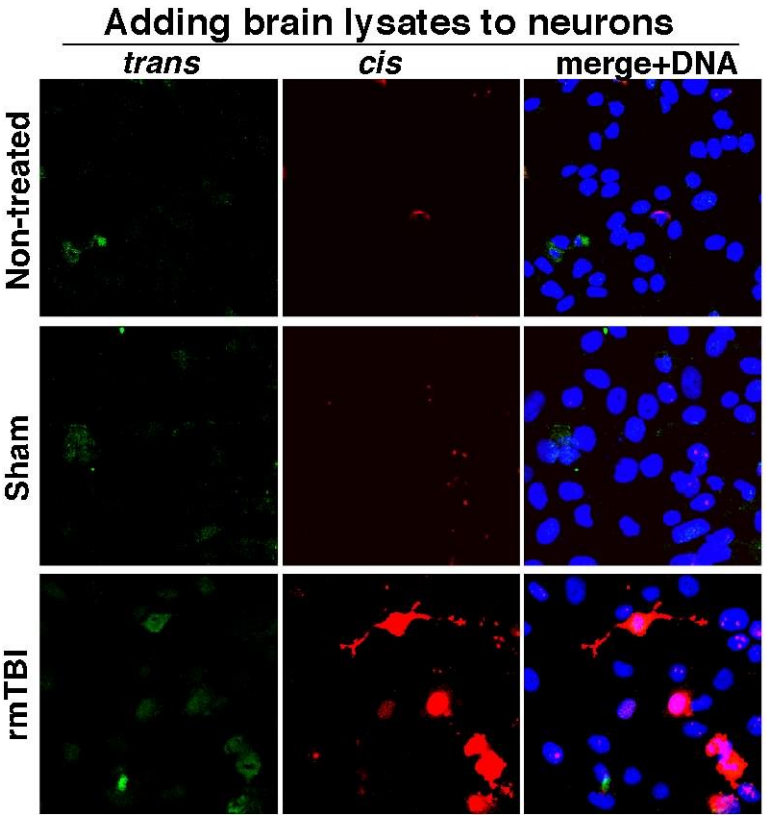
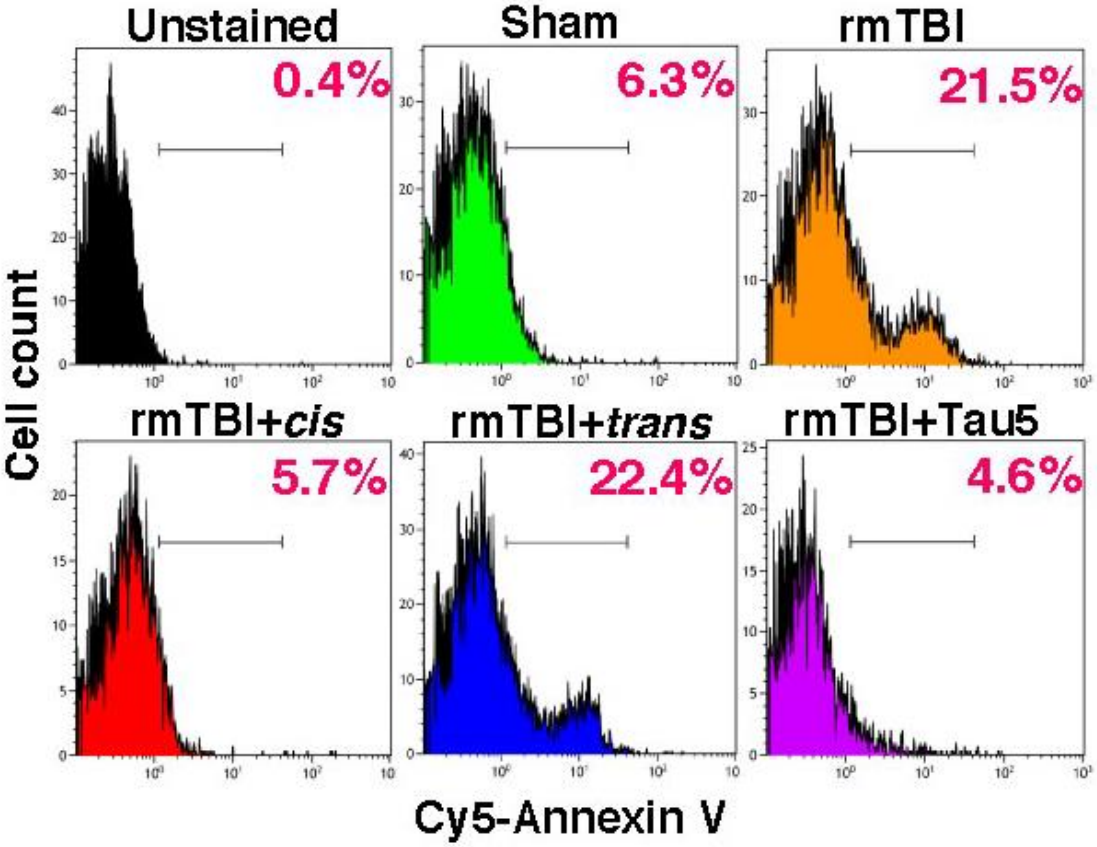
TBI brain extracts kill neurons by apoptosis, which is fully stopped by *cis*, but not *trans*, mAb





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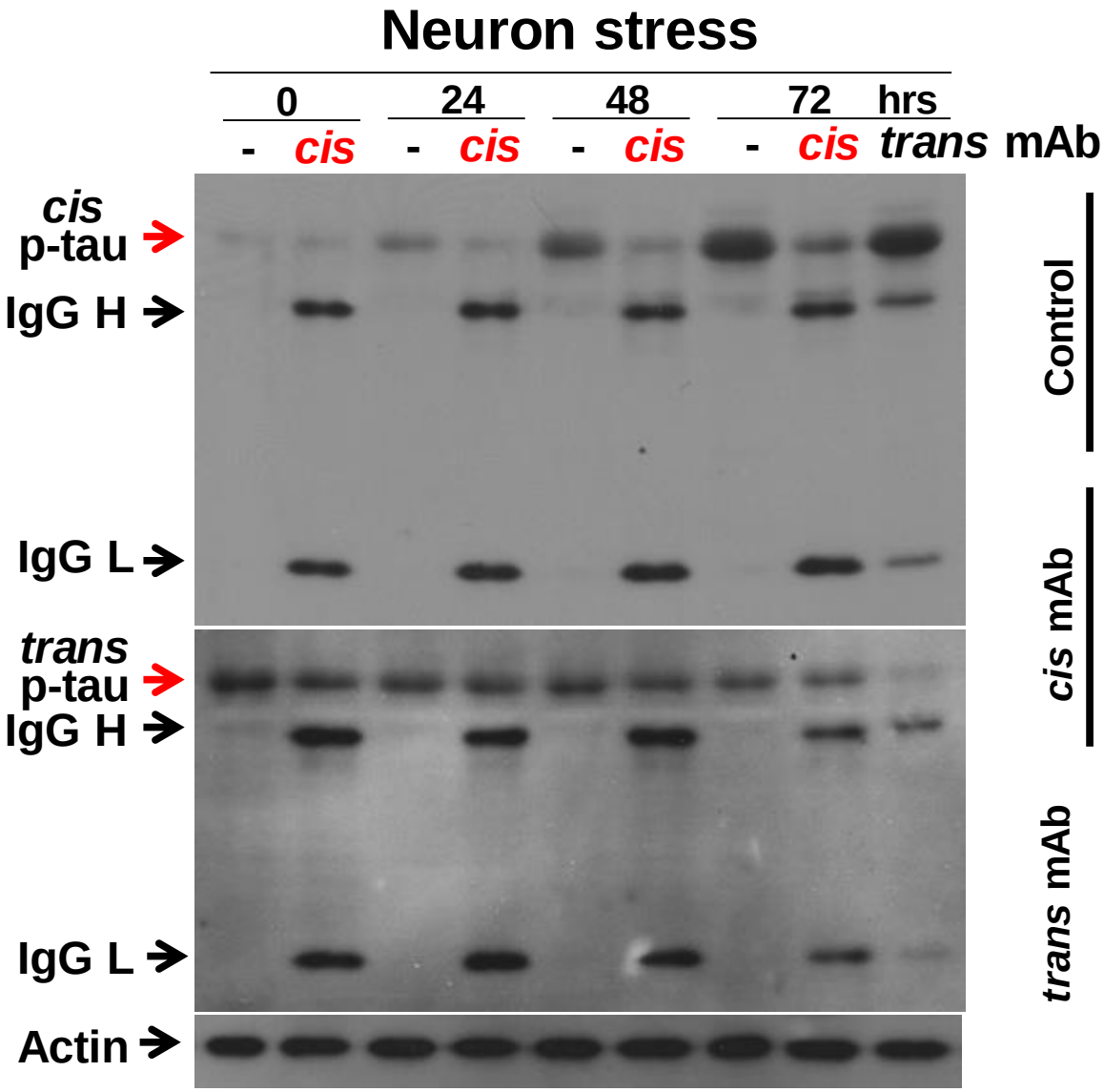
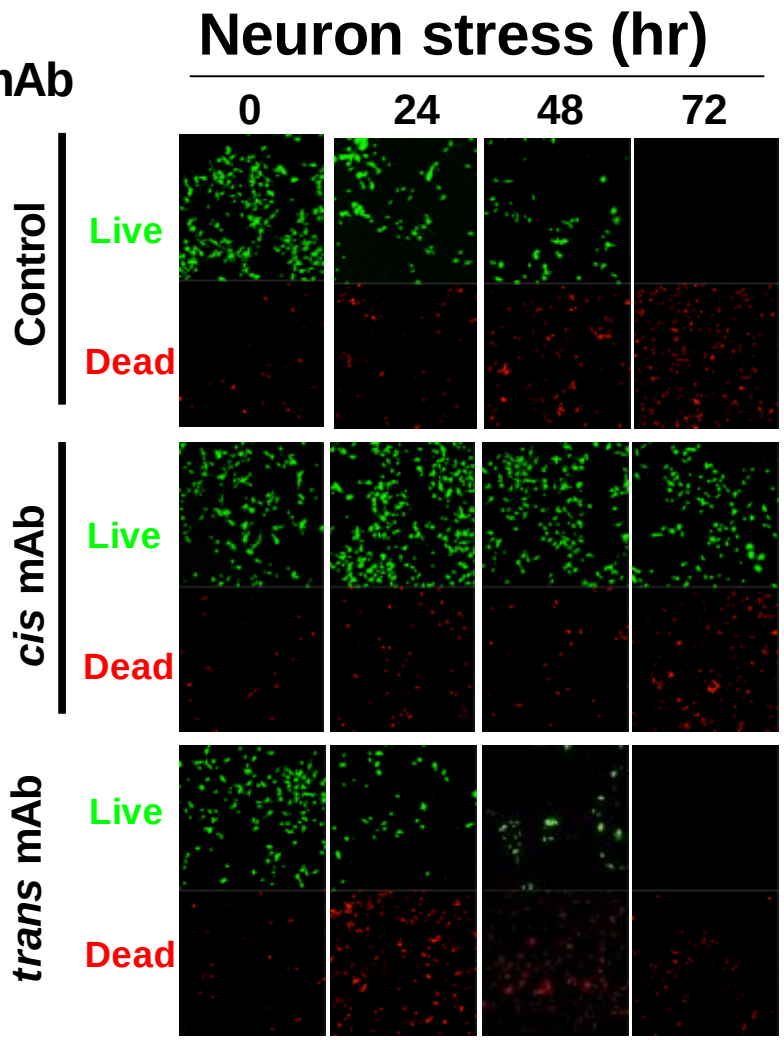
cis mAb potently stops the ability of serum depletion to induce *cis* p-tau and apoptosis





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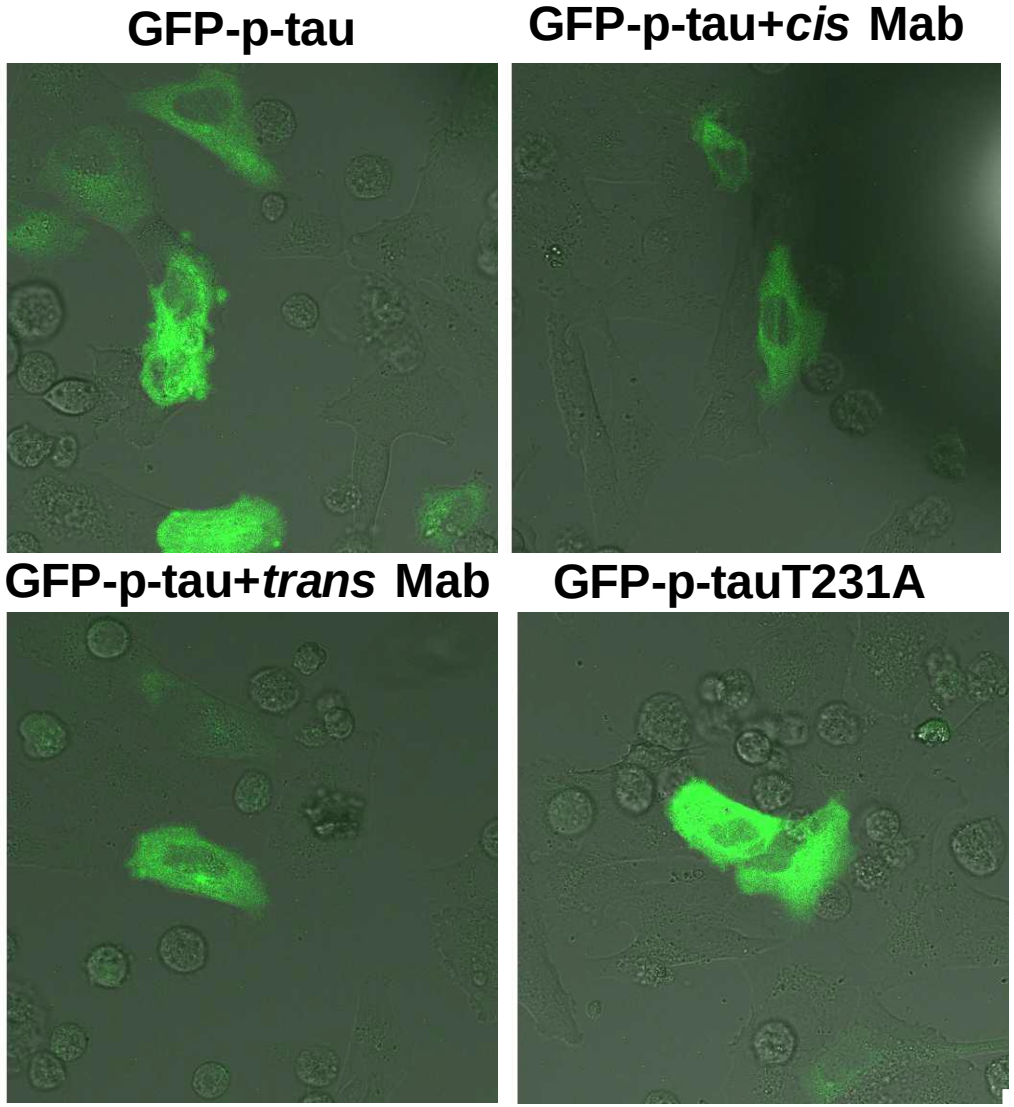
Kondo et al., 2015, Nature

cis pT231-tau is both necessary and sufficient to induce neurotoxicity, but is fully stopped by cis mAb



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Kondo et al., 2015, Nature

Treating ssTBI or rmTBI mice with *cis* P-tau mAb stops brain damage and prevents CTE

- Eliminates toxic *cis* P-tau and spread
- Restores brain histopathology
- Restores neuron ultrastructure, electrophysiology
- Prevents widespread tau tangle formation
- Prevent widespread neuron loss and brain atrophy
- Restore clinically relevant functional outcomes

  including risk-taking behavior, urinary dysfunction, sensorimotor imbalance, other neurological problems.

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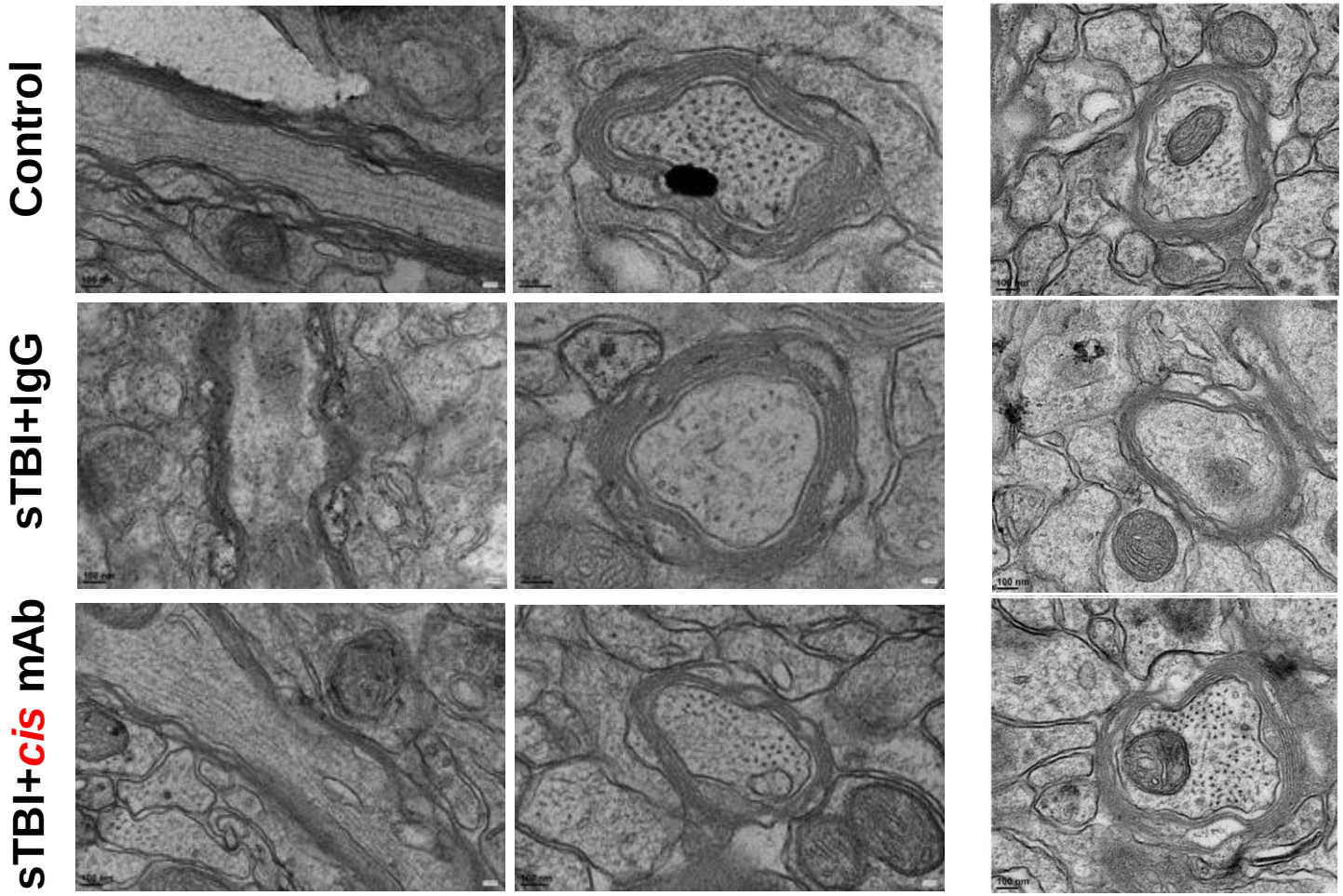
Kondo et al., 2015, Nature
Lu & Zhou, unpublished data

cis mAb potently restores axonal microtubules and mitochondrial defects after severe TBI in mice

2 week after injury

Microtubules **Mitochondria**



Kondo et al., 2015, Nature

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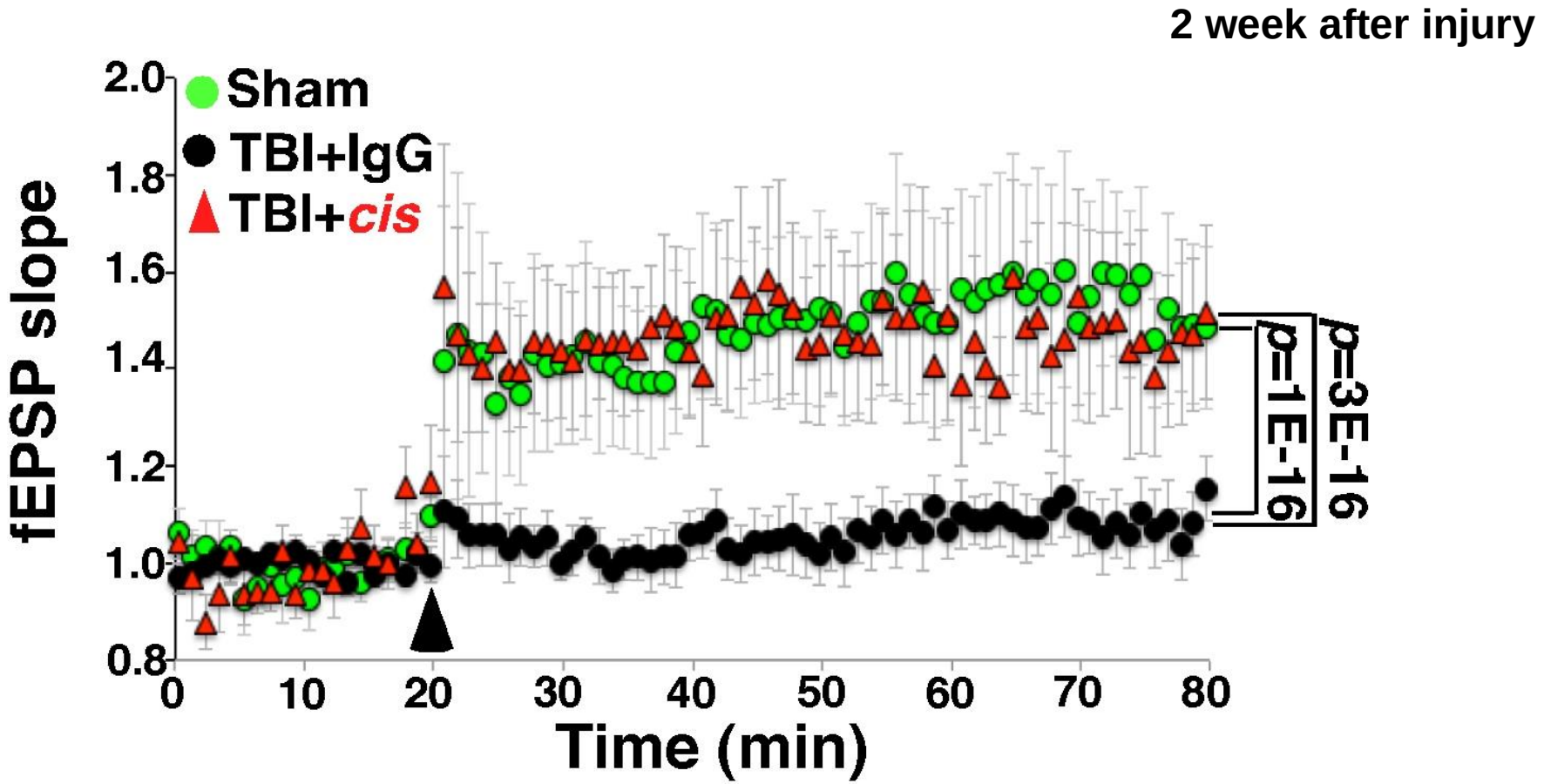
Cis mAb potently restores cortical long-term potentiation (LTP) defects after ssTBI in mice



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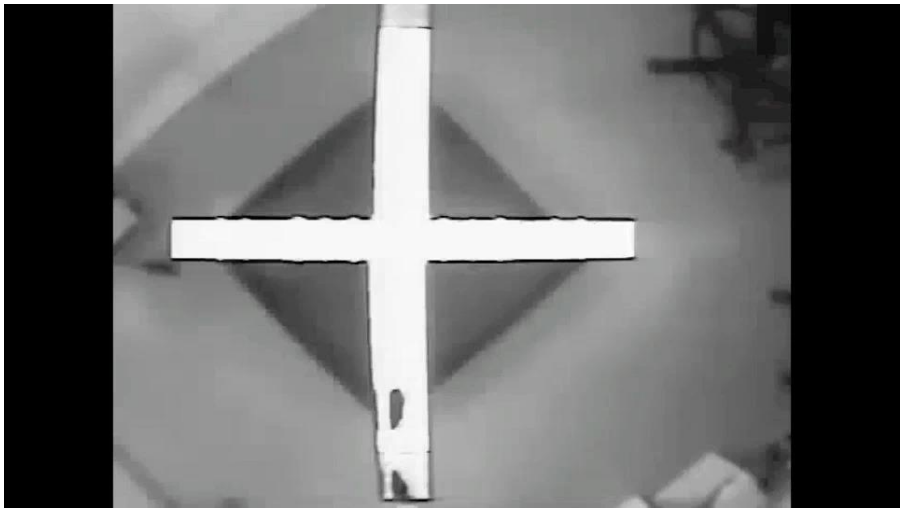
Kondo et al., 2015, Nature

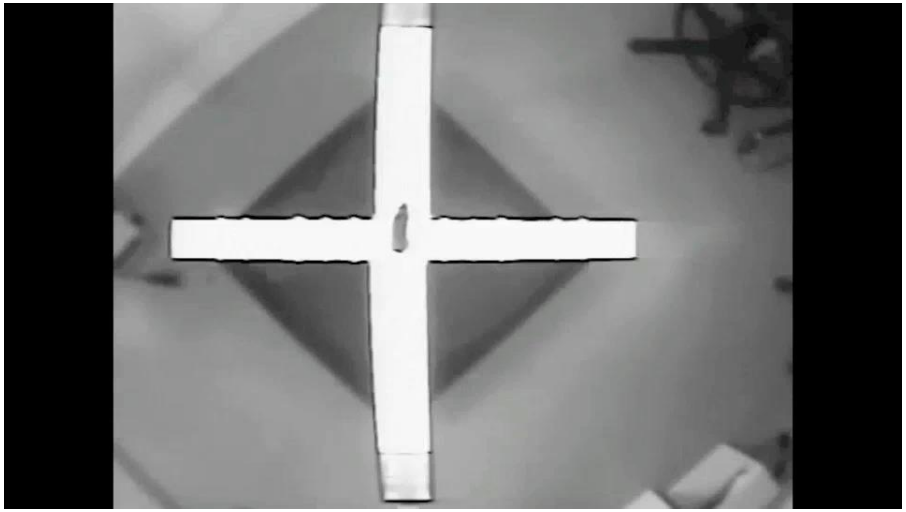
Cis mAb potently restores anxiety/risk-taking behavior after severe TBI

Walf and Frye, 2007, Nature Protocol, 2: 322

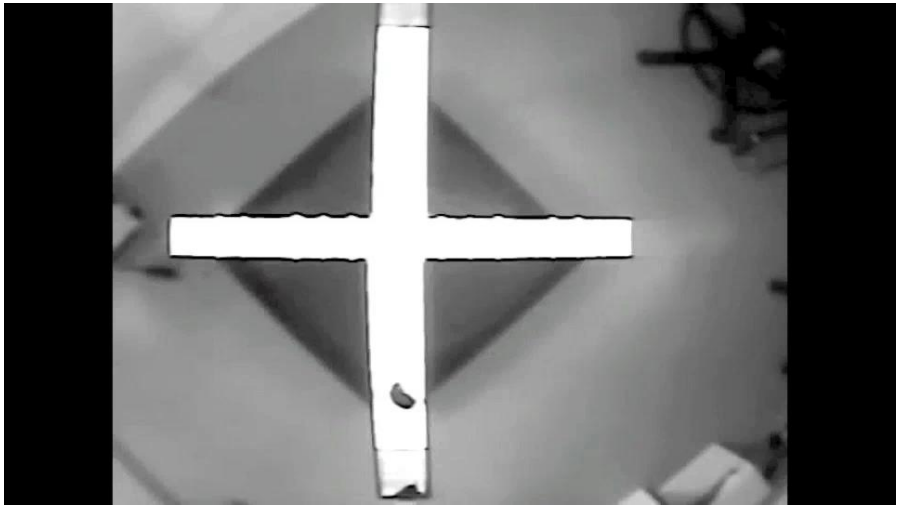
Sham

2 M after injury





TBI+IgG



TBI+*cis* mAb

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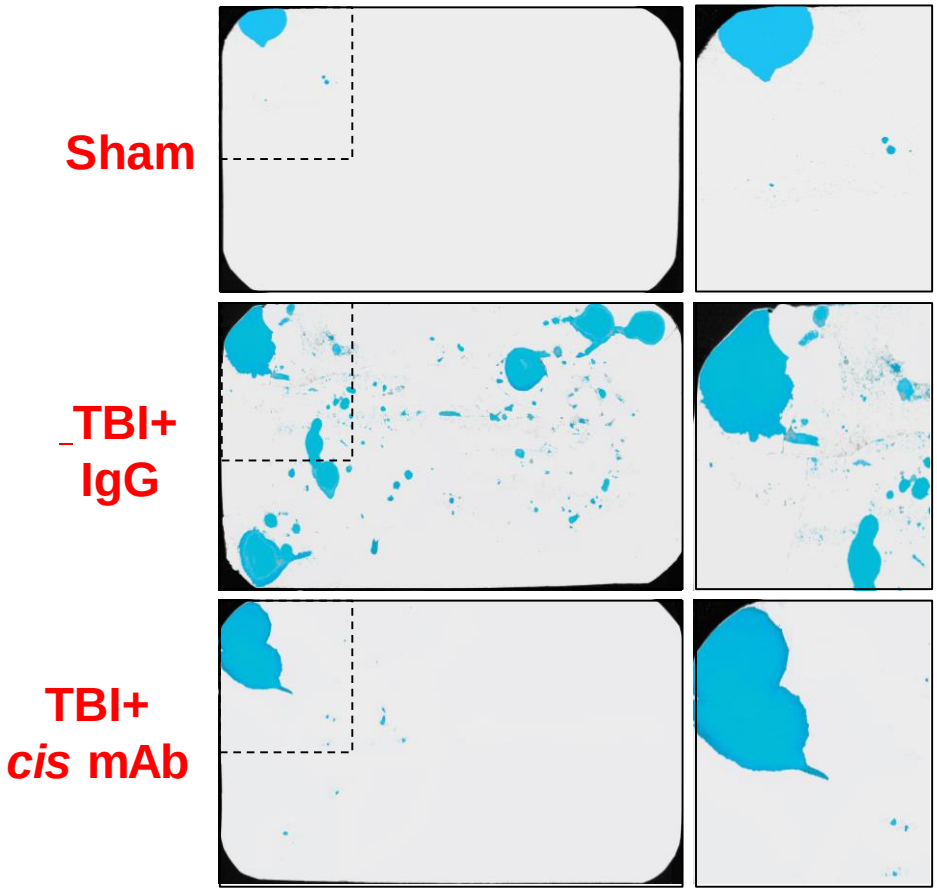
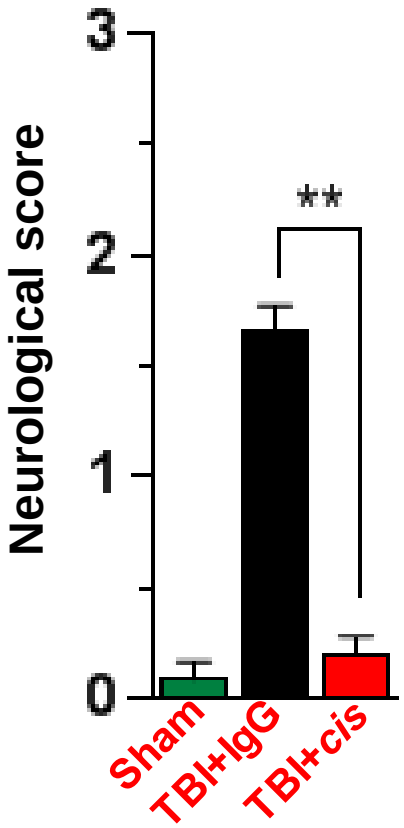
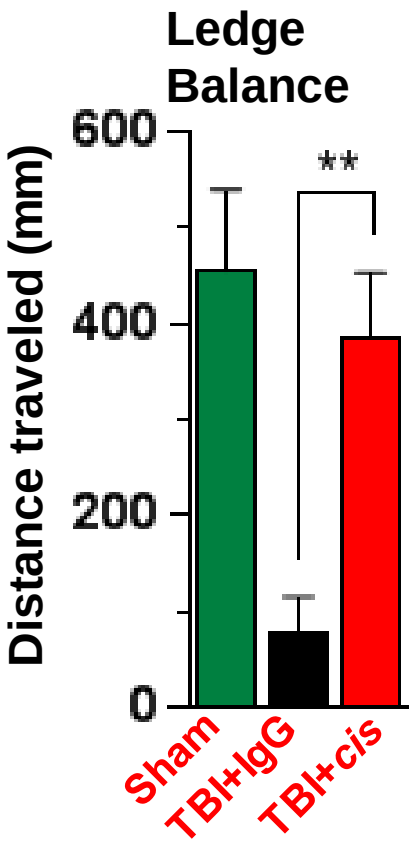
Kondo et al., 2015, Nature

Cis antibody treatment restores urinary incontinence, other neurological defects after severe TBI in mice

5 months after injury







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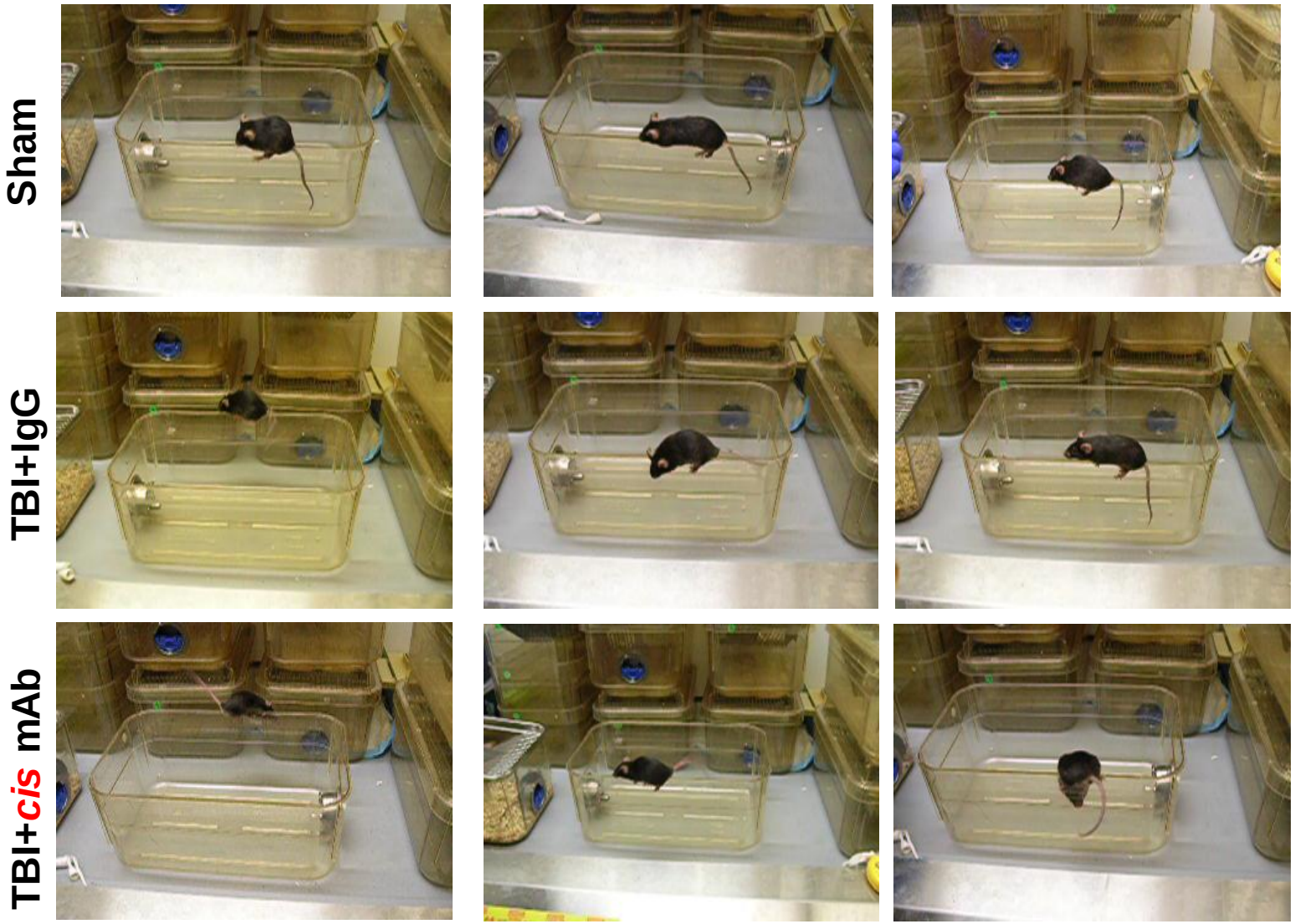
Lu, Zeidel et al., unpublished

Cis mAb restores sensorimotor/balance dysfunction after severe TBI, a major clinical problem

5 month after injury



Ledge test
Rangroo et al. 2013, Nat Med 19, 12

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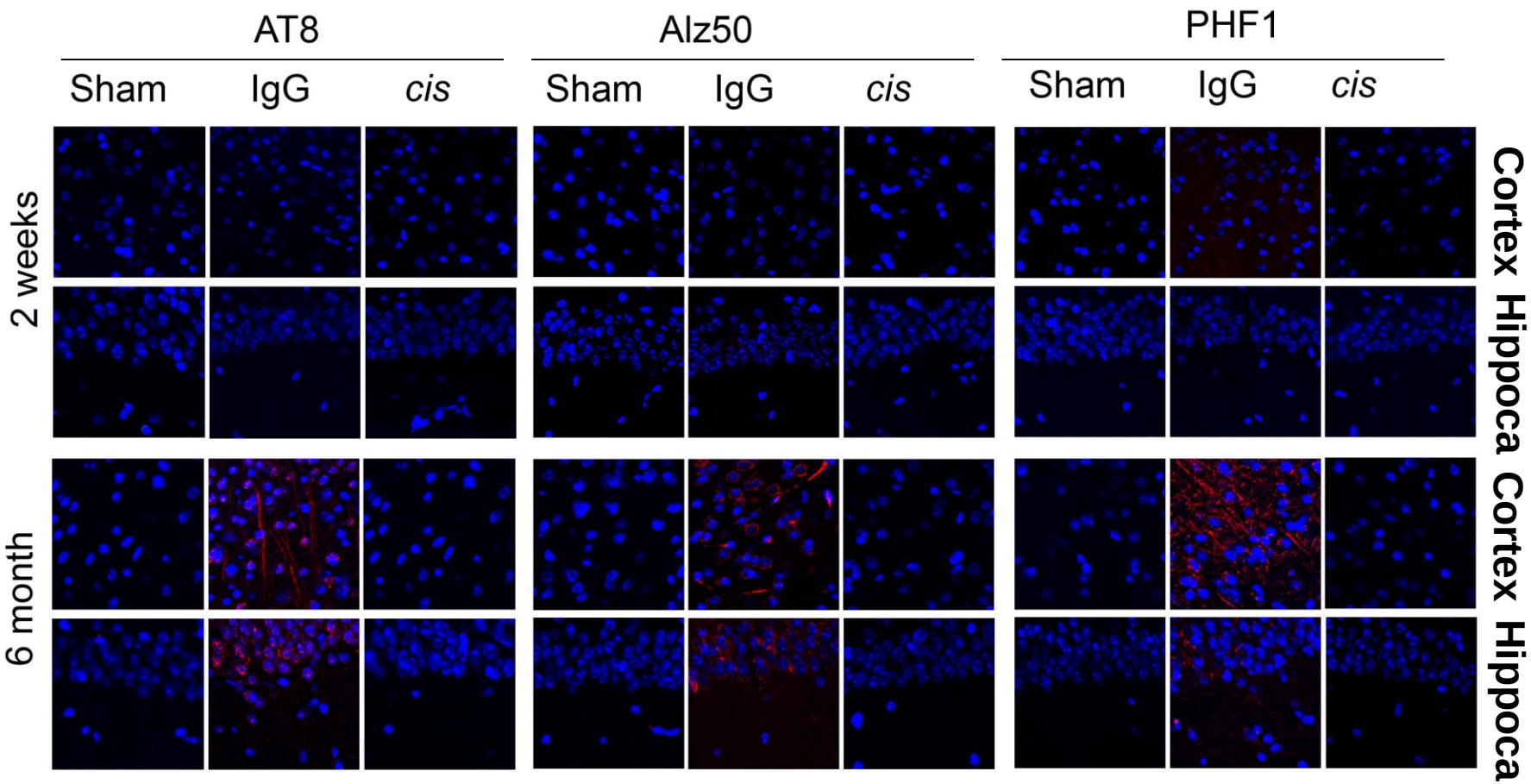
Lu et al., unpublished

Cis mAb prevents the development and spread of tauopathy after TBI, a hallmark of CTE



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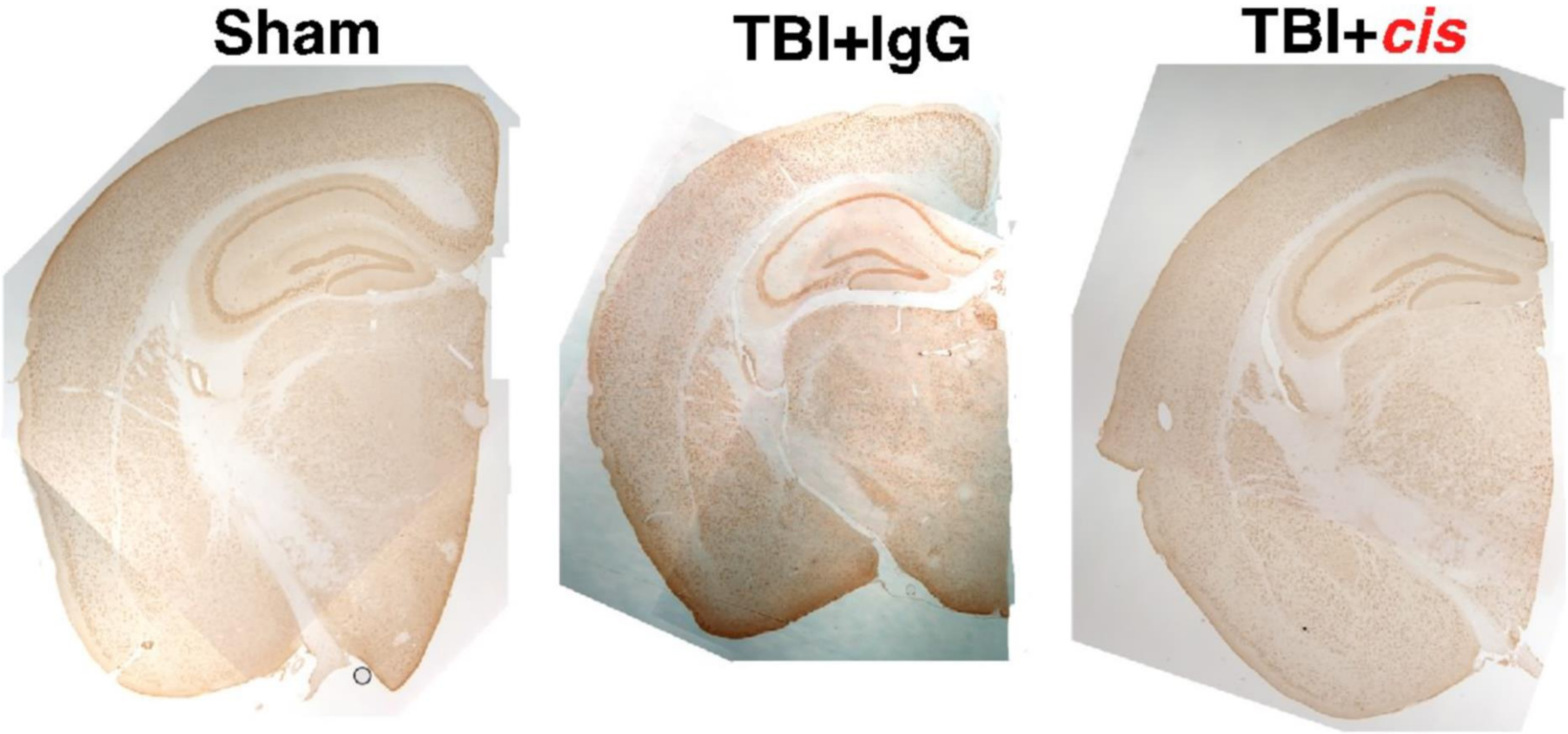
Kondo et al., 2015, Nature

Cis antibody prevents widespread brain atrophy after severe TBI, a major feature of CTE and AD

6 month after injury



Kondo et al., 2015, Nature

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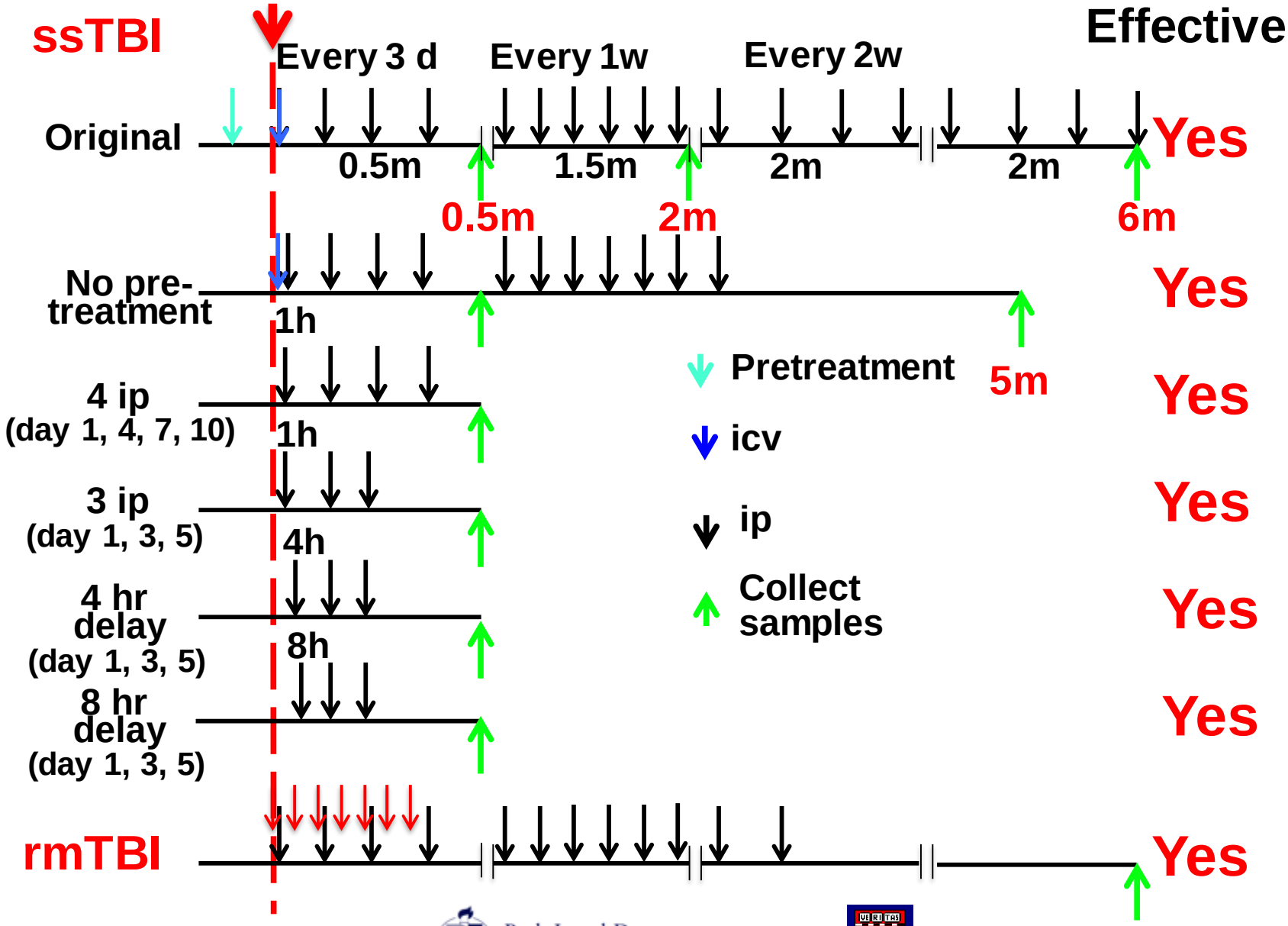
Cis mAb treatment regimens and their effectiveness



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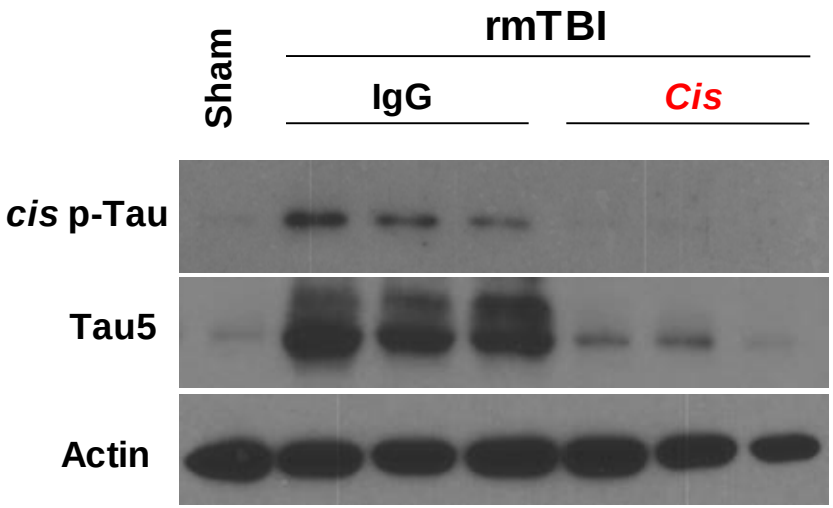
Kondo et al., Nature, 2015;
Lu et al., unpublished

Cis mAb treatment effectively eliminates *cis* P-tau in repetitive mild TBI mice

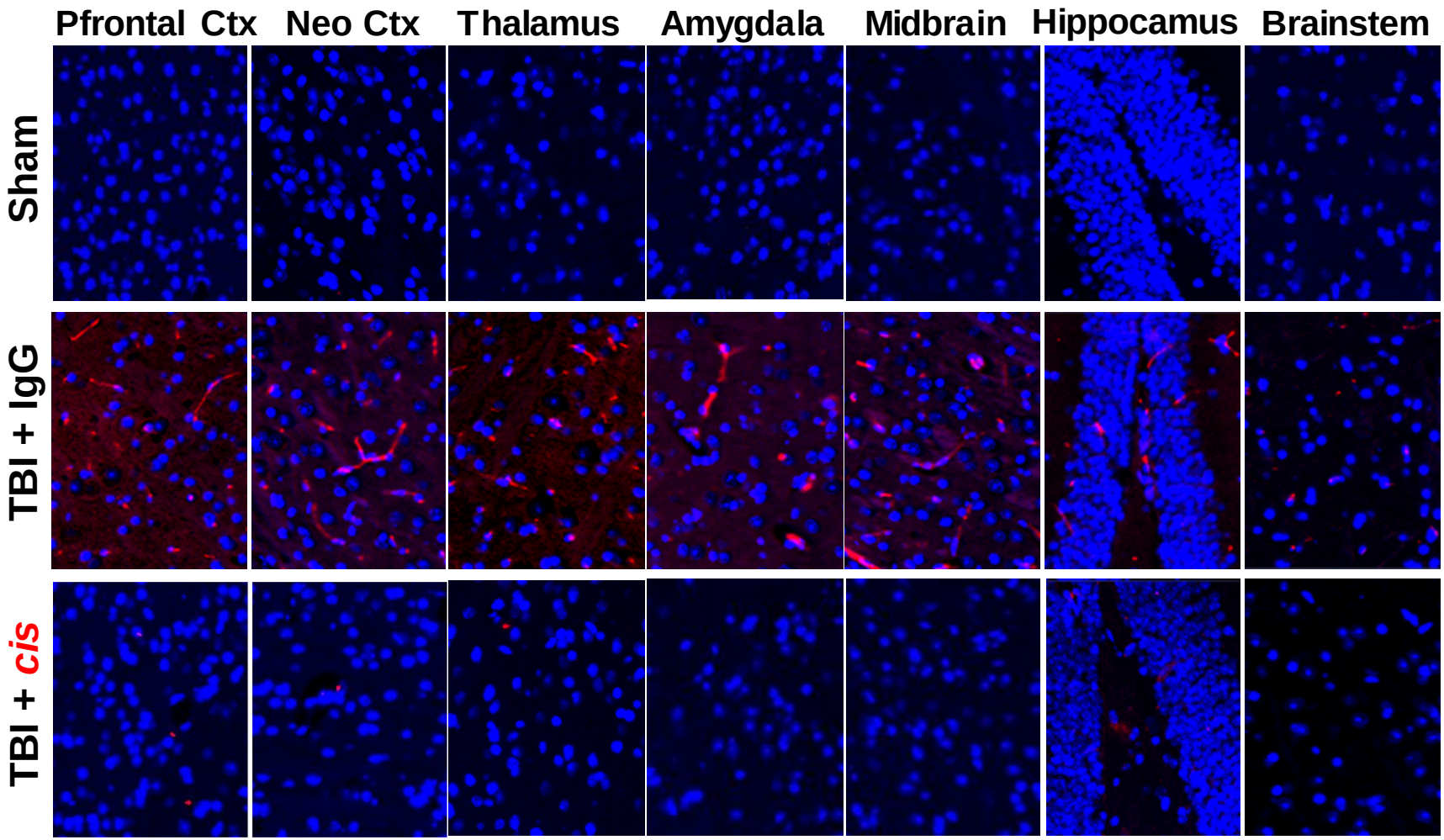


Beth Israel Deaconess Medical Center

Lu, Zhou, et al., unpublished

Cis mAb treatment prevents tauopathy development and spread in repetitive mild TBI mice

AT8



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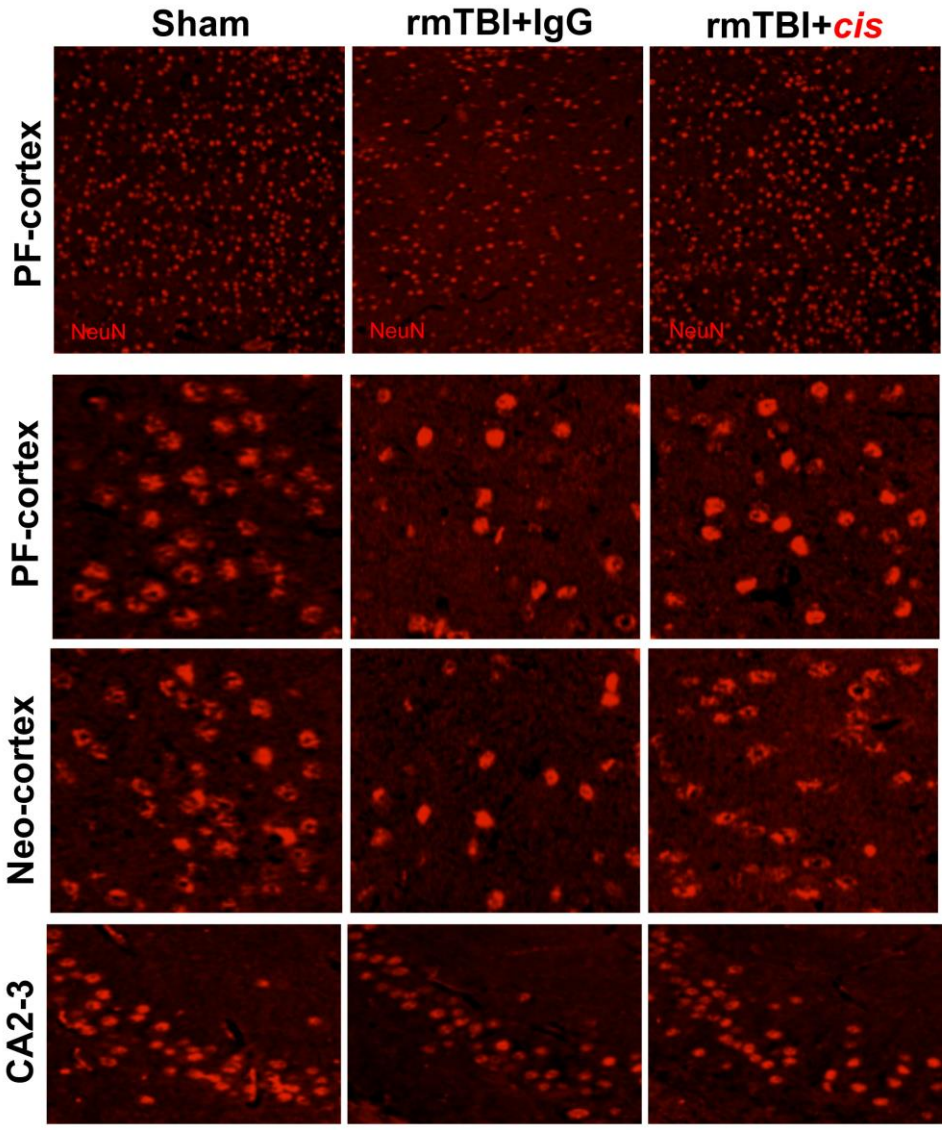
Lu, Zhou, et al., unpublished

Cis mAb treatment prevents neuron loss after rmTBI in mice



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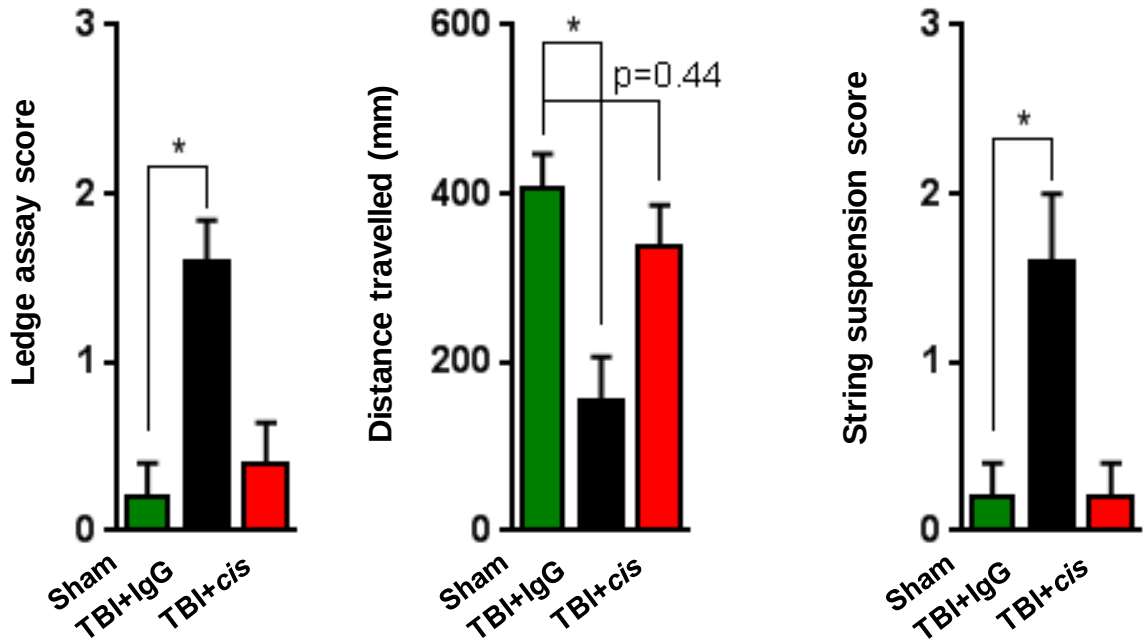
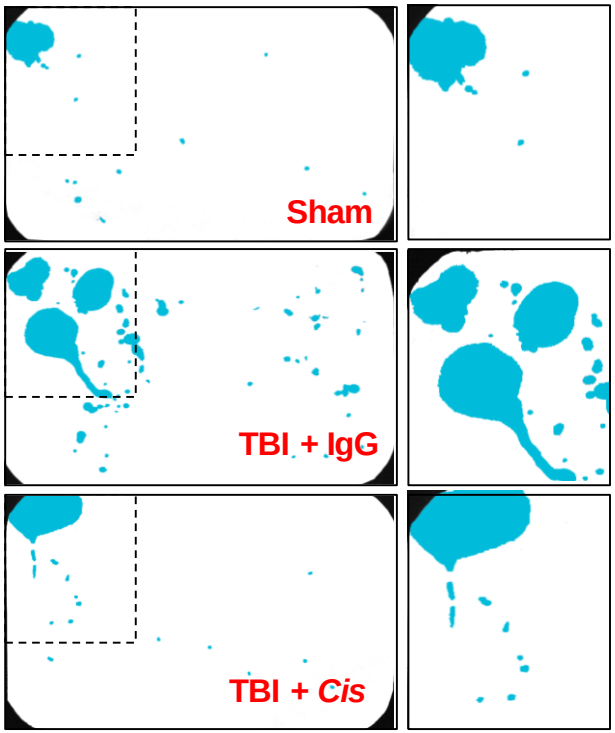
Lu, Zhou, et al., unpublished

Cis mAb treatment restores neurological and behavioral defects after rmTBI in mice





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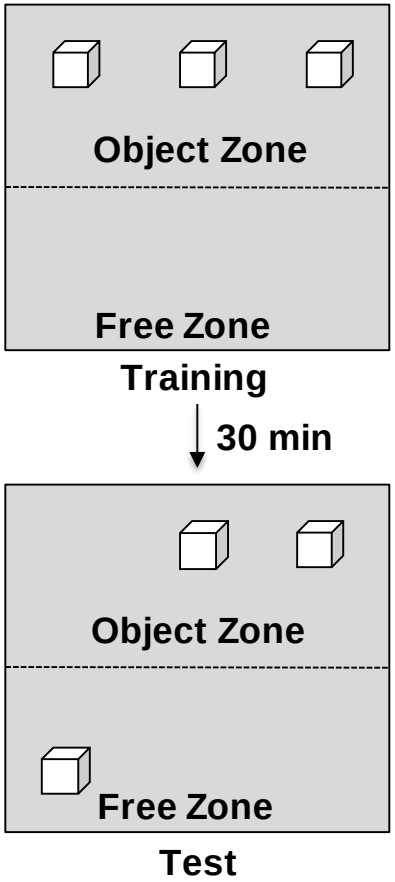
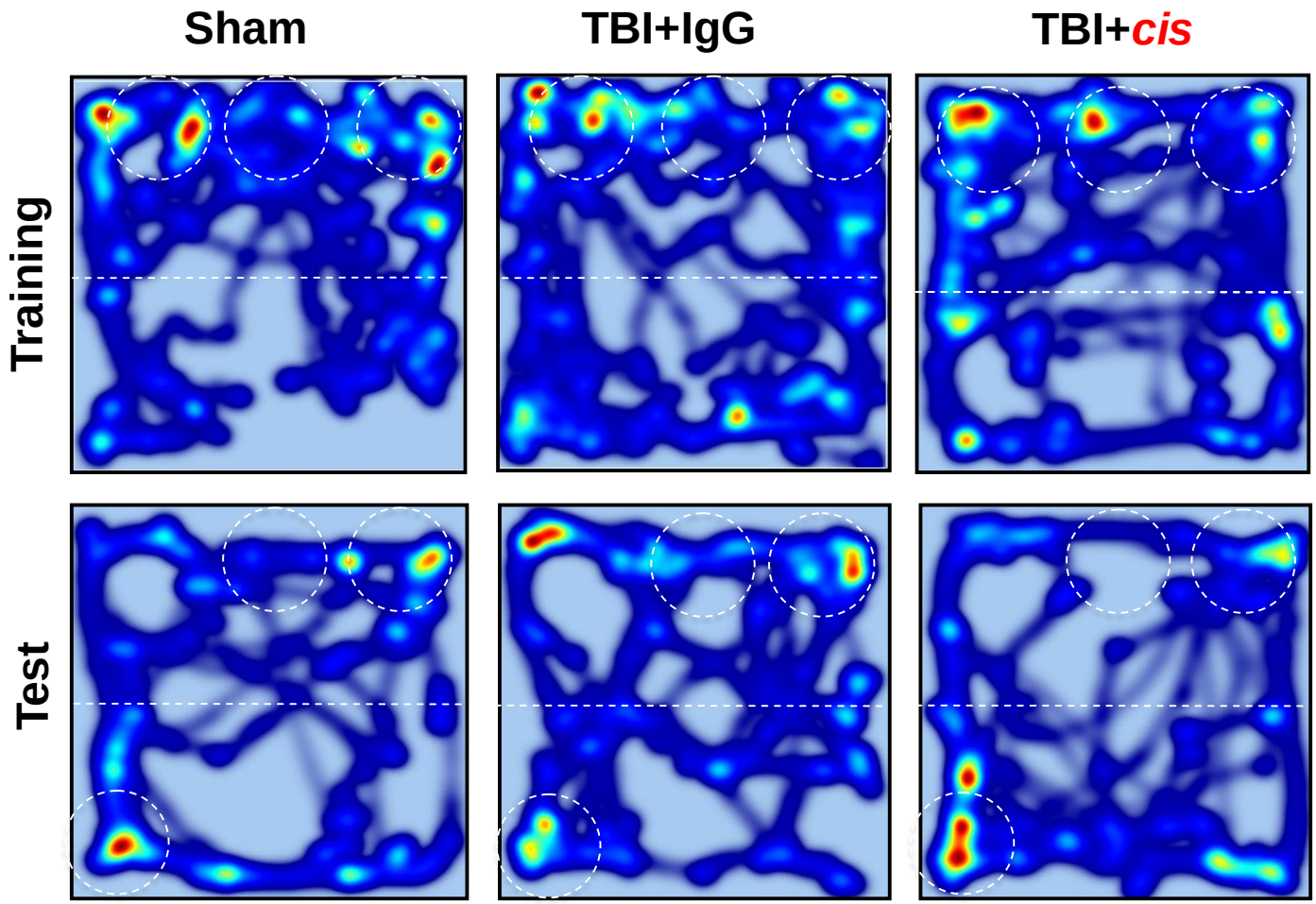
Lu, Zeidel, Zhou, et al., unpublishe

Cis mAb treatment prevents learning and memory loss in repetitive mild TBI mice





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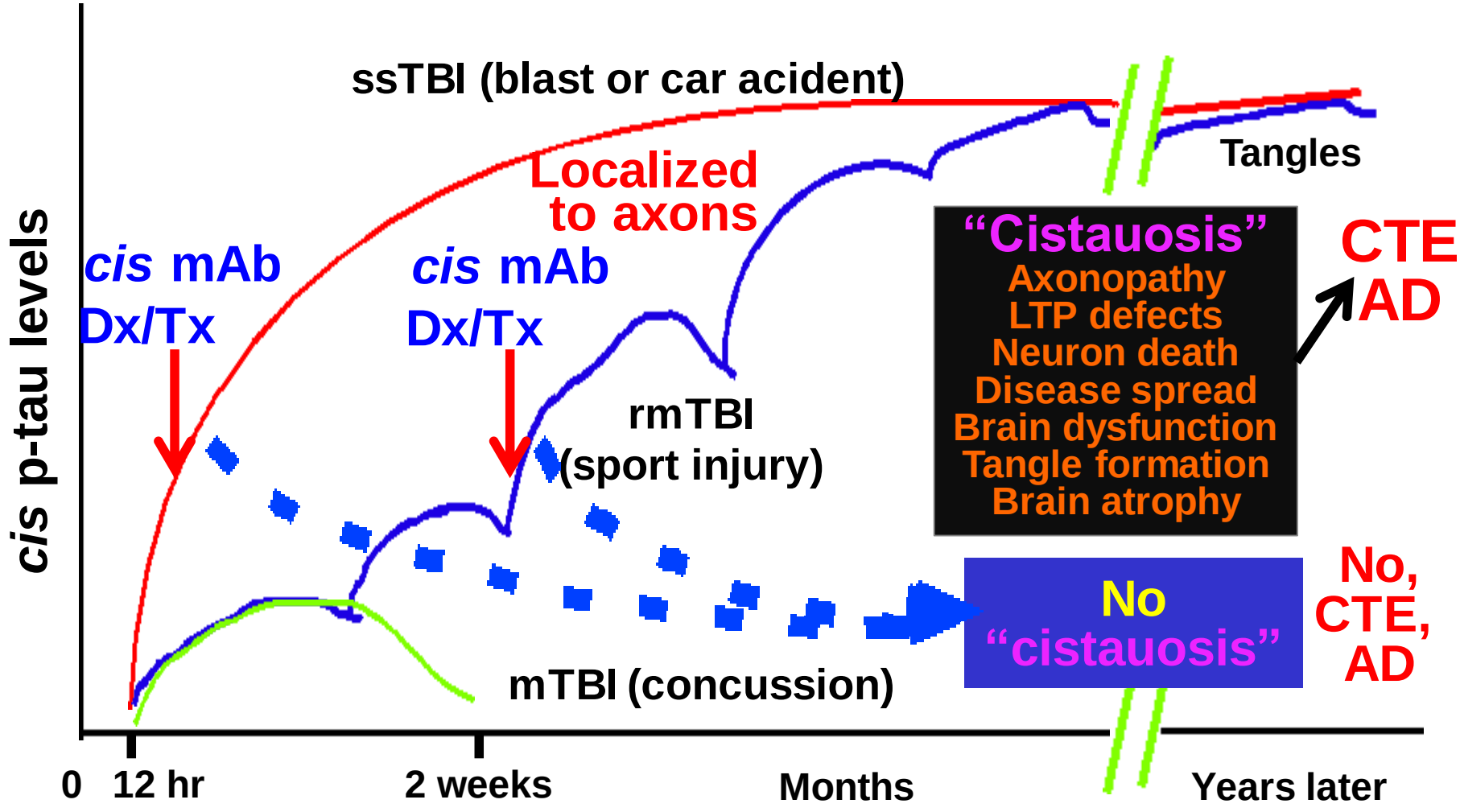
Lu, Zhou, et al., unpublished

Early disease mechanism, diagnosis and treatment of TBI, AD and CTE



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Kondo et al., 2015, Nature

Lu/Zhou laboratories

Pin1 in cancer

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Xiaolian Lian
Howie Chen
Yu-Min Lin
Manli Luo
Adrian Tun-Kyi
Sang Min
Tae Ho Lee
Prudent Lam
Futoshi Suizu
Akihide Ryo
Yih-Cherng Liou
Gerburg Wulf

Pin1 fundamentals

Morris Nechama
Yusuke Kawabata
Kazuhiro Nakamura
Oliver Kops
Gerlind Stoller
Pei-Jung Lu

Pin1 in Alzheimer, TBI

Asami Kondo
Onder Albayram
Howie Chen
Jenny Driver
Koorosh Shaphafd
Kazuhiro Nakamura
Lucia Pastorino
Tae Ho Lee
Suk Ling Ma
Jormay Lim
Anyang Sun
Yih-Cherng Liou
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Pin1 Inhibitors

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(Former lab members in blue)

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(Collaborators in AD and TBI)

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Linda Nicholson
Virginia Lee
Skip Binder
Rebekah Mannix
Bill Meehan
Lee Goldstein
Ann Mckee
Chagi Pick
Nigel Greig

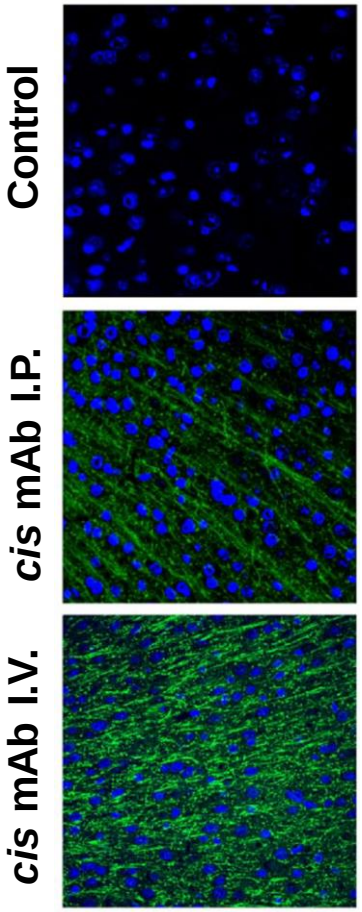
Antibodies and Vaccines

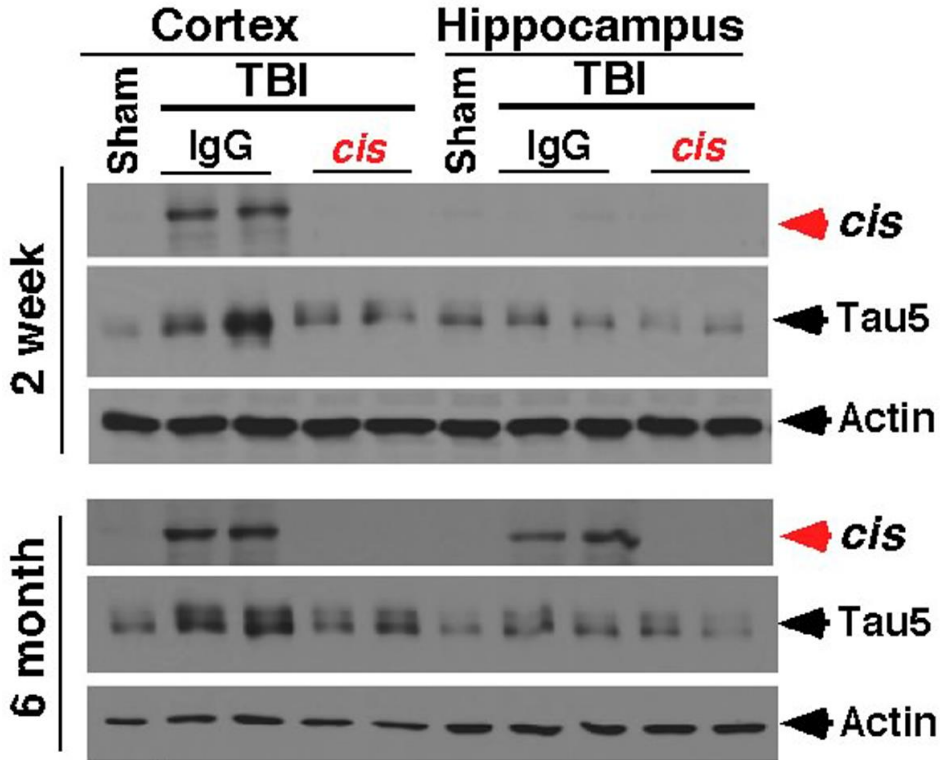
Asami Kondo
Onder Albayram
Koorosh Shaphafd

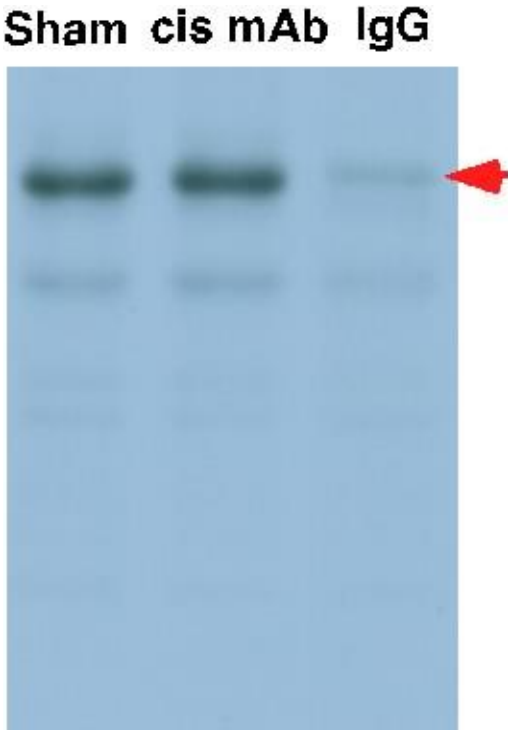
PinX1

Chenyu Li

Cis mAb blocks *cis* P-tau induction, spread and neurotoxicity after ssTBI in mice







PARP cleavage

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Kondo et al., 2015, Nature