ER Stress Modulation as a Target for Ameliorating Effects of Blast Induced Traumatic Brain Injury

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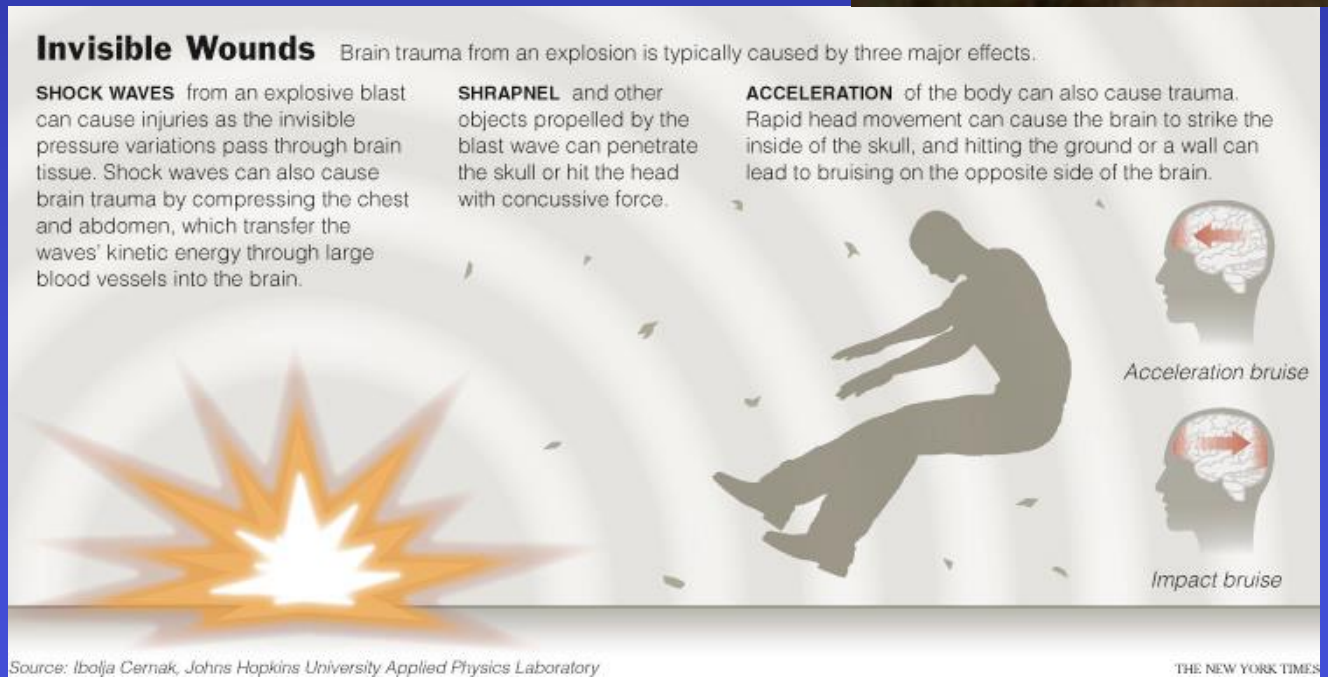
Disclosures

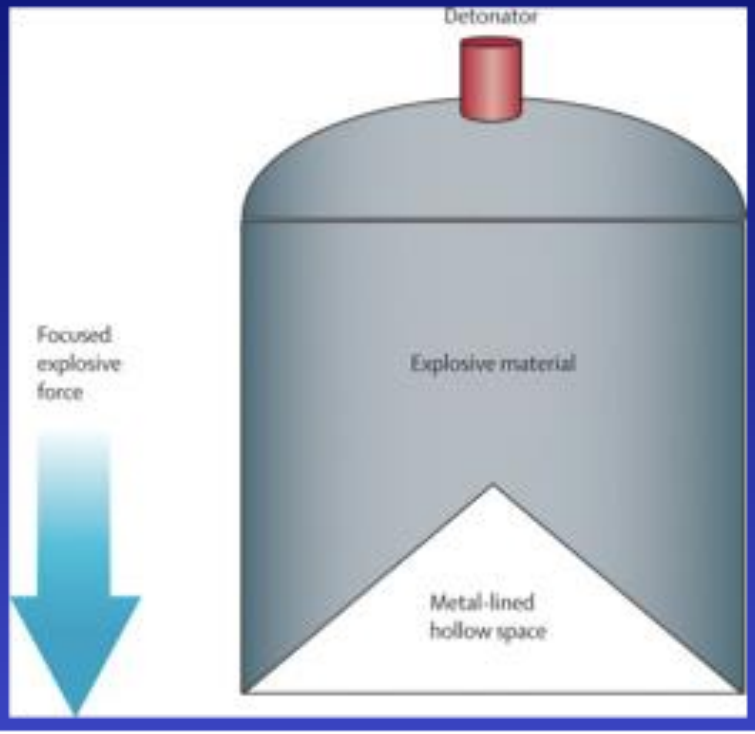
- Grants
  - NIH
  - NIOSH
  - Synthes
  - BHR Pharma
  - Hyperbranch Medical technologies

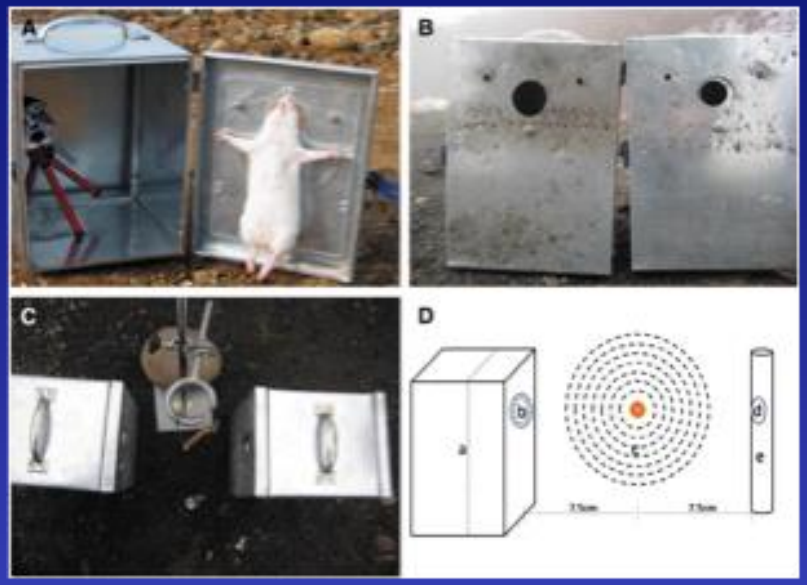
Blast

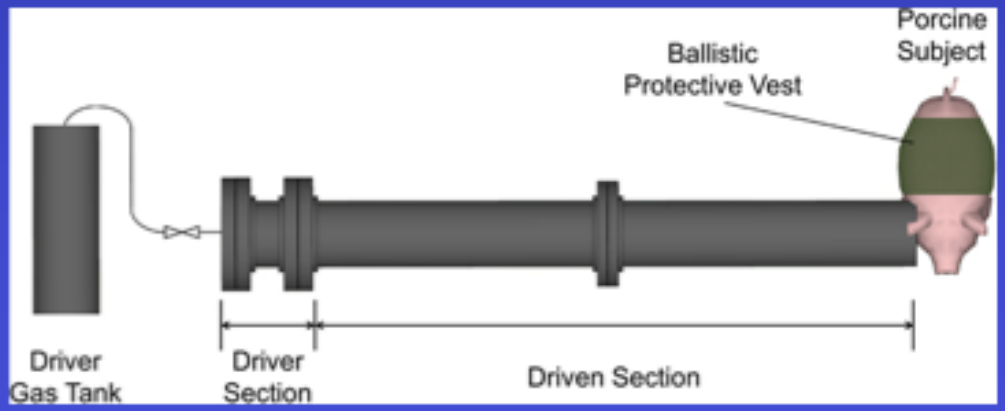
- "Signature injury of modern warfare"
  - 320,000 soldiers
  - Association with long-term neurodegenerative disease: CTE

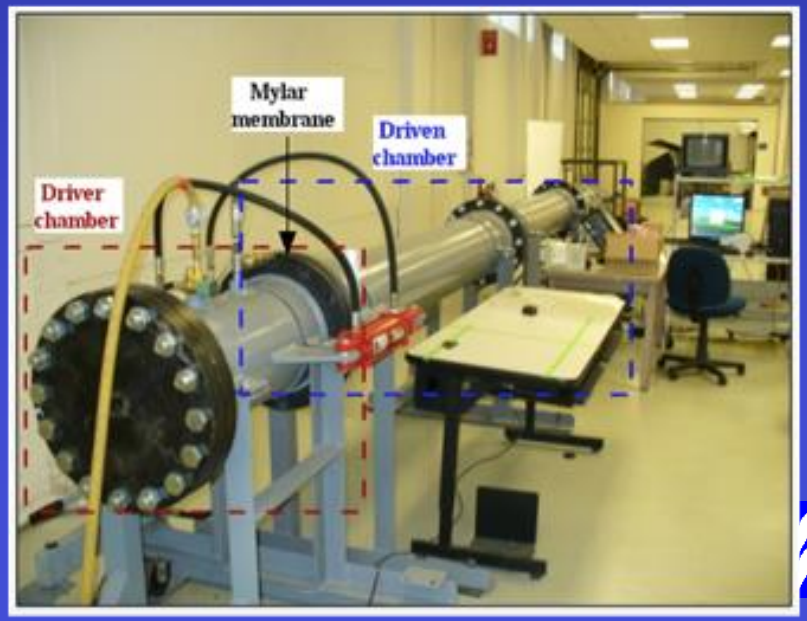


Many Models of Blast









Important Model Parameters

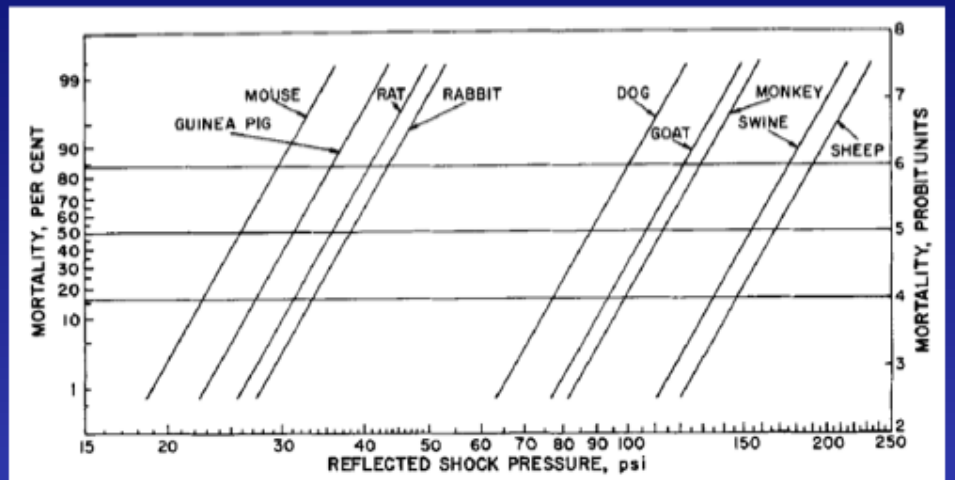


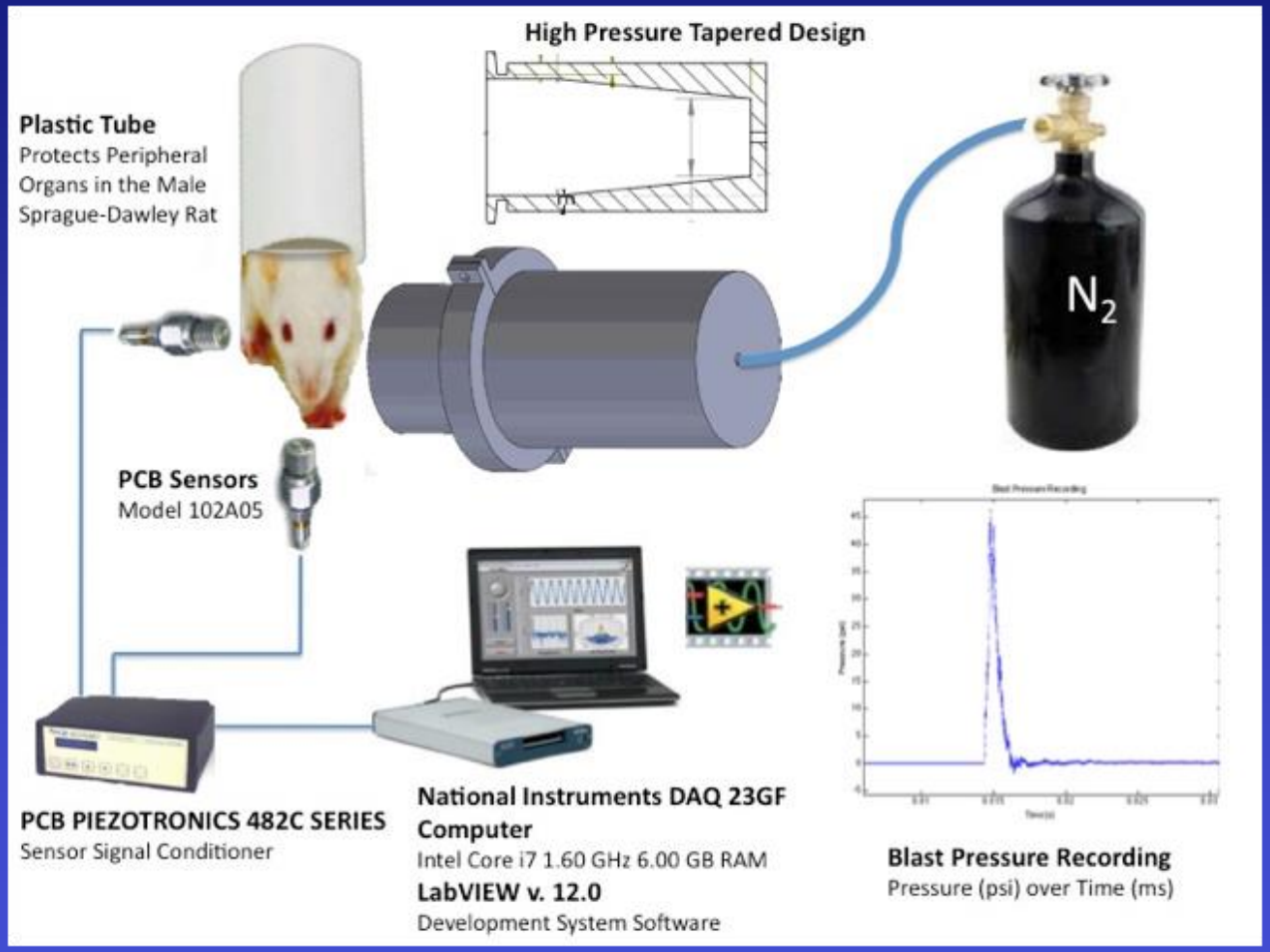
FIGURE 4. Mortality curves for animals exposed to "short"-duration reflected pressures from high-explosive charges detonated overhead while mounted prone on a concrete pad. Probit regression equation: $y = a + b \log x$; where y is the percent of mortality in probit units, a and b the intercept and slope constants, and x the pressure. Data taken from References 3, 6, and 7; all measurements were made at ambient pressure of 12 psia.

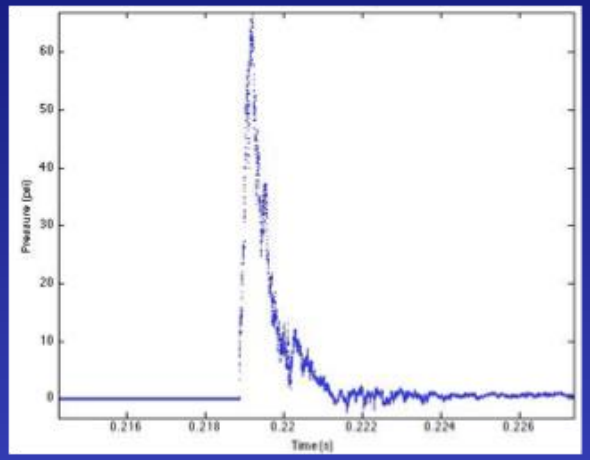
- Peak Overpressure
- Duration
- Impulse (area under curve)
- Sensor Orientation (Reflected vs. Incident)

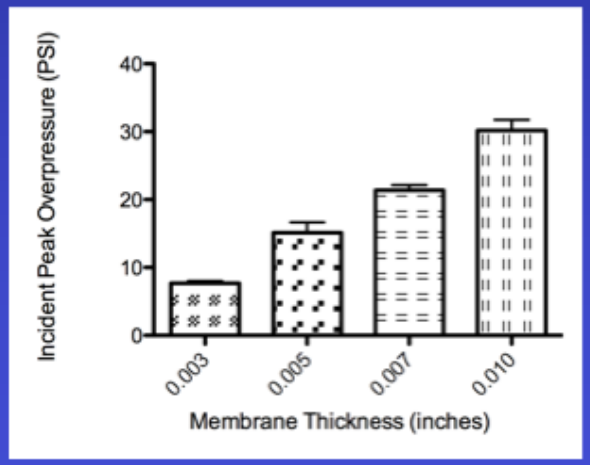
What is clinically relevant?

  – Humans: ~7-145 PSI; 2-10 ms duration

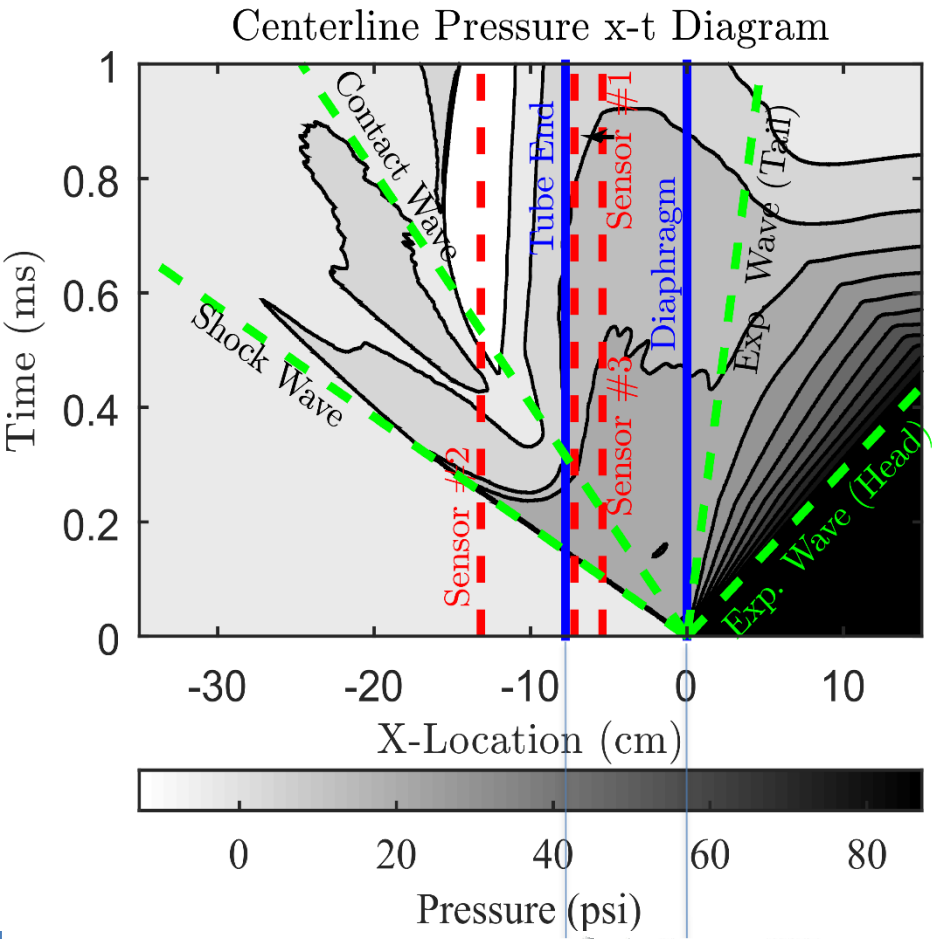
Our Set Up

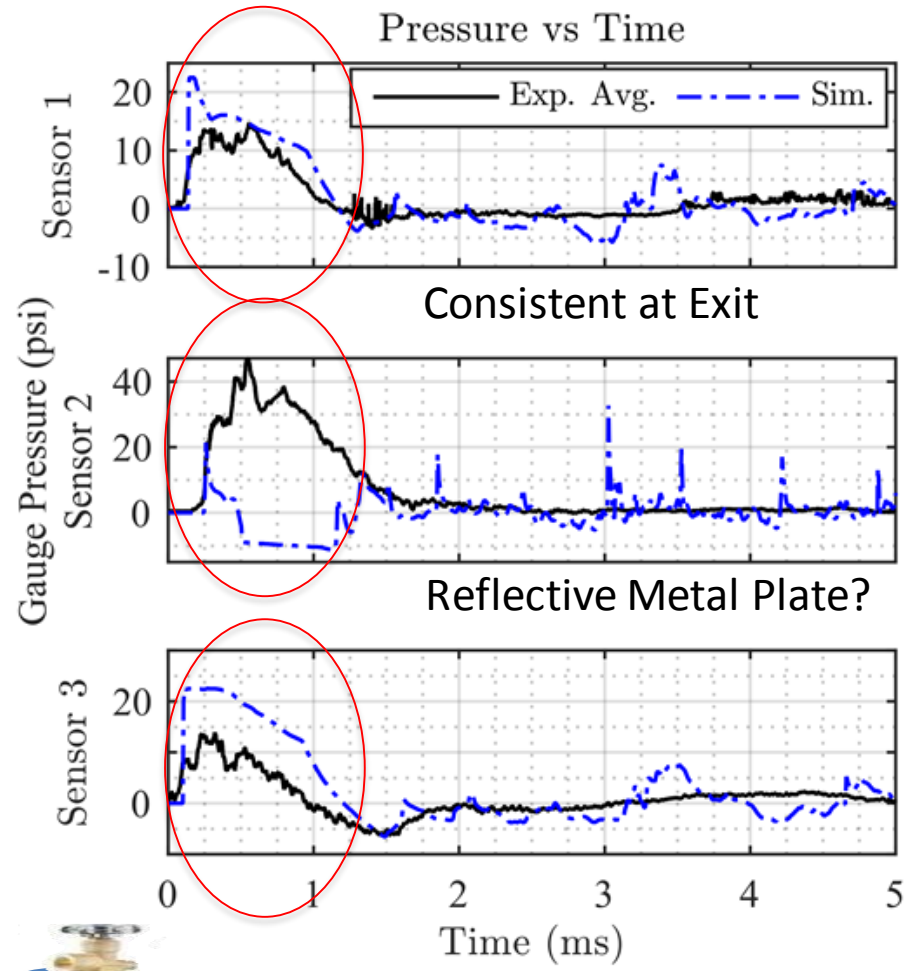

High Pressure Tapered Design
Plastic Tube
Protects Peripheral Organs in the Male Sprague-Dawley Rat
N_2
PCB Sensors
Model 102A05
PCB PIEZOTRONICS 482C SERIES
Sensor Signal Conditioner
National Instruments DAQ 23GF
Computer
Intel Core i7 1.60 GHz 6.00 GB RAM
LabVIEW v. 12.0
Development System Software
Blast Pressure Recording
Pressure (psi) over Time (ms)


Pressure (psi)
Time (s)


Incident Peak Overpressure (PSI)
Membrane Thickness (inches)
0.003
0.005
0.007
0.010

Compare to Mathematical Blast Model

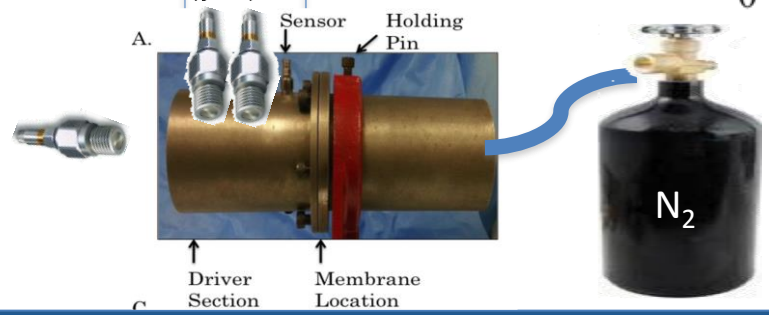




Consistent at Exit

Reflective Metal Plate?

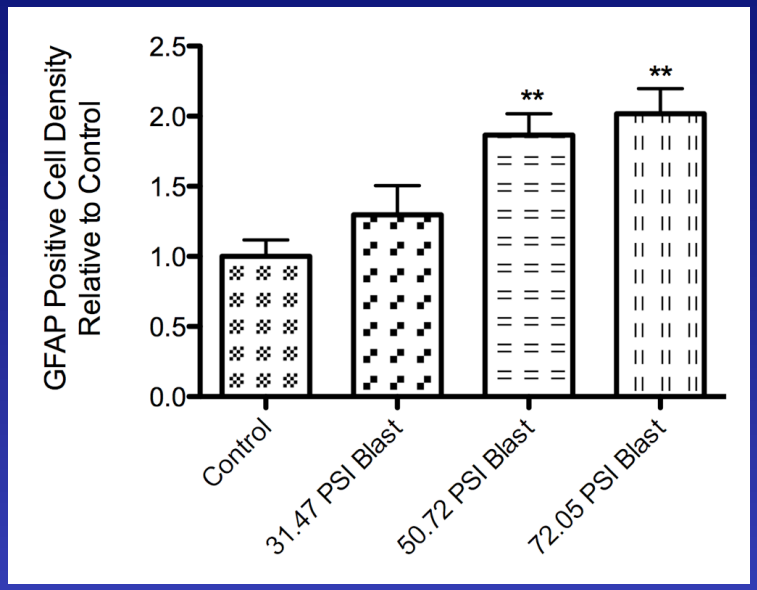
Gradual Increase
Not Sharp?

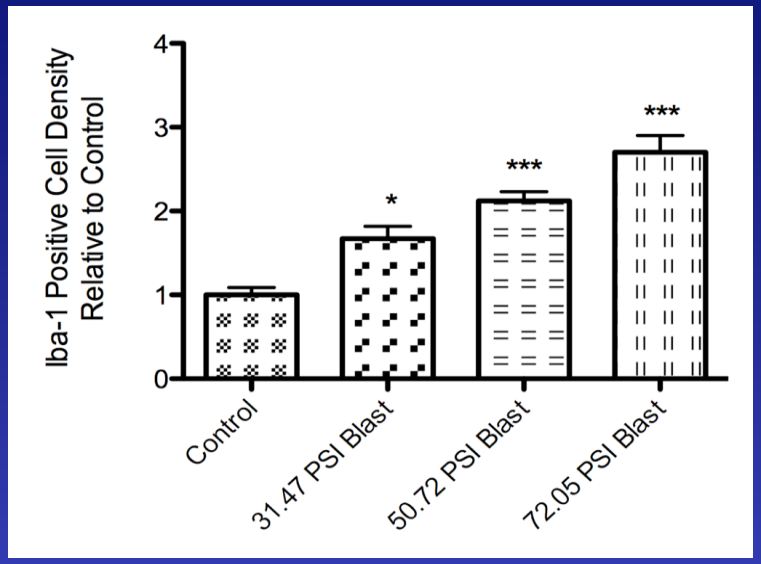


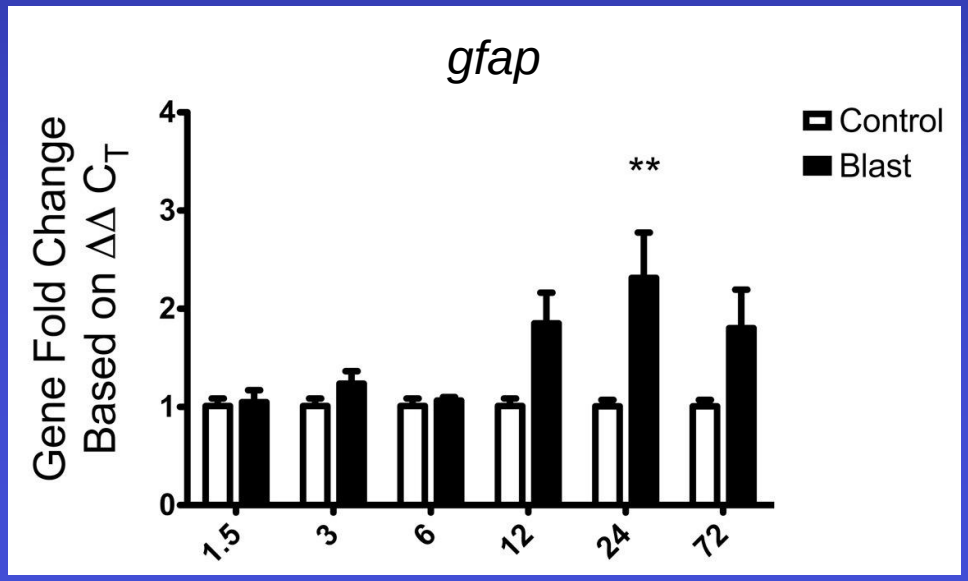
Map Blast Wave Velocity and Shape

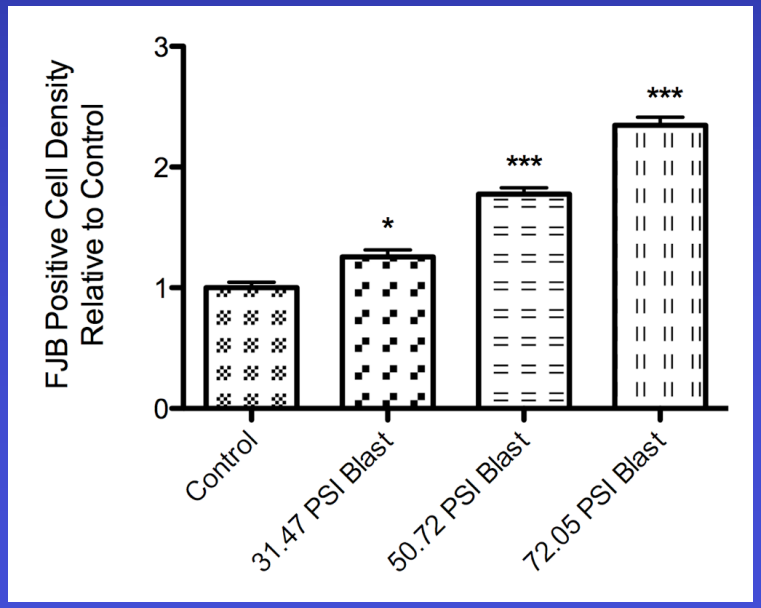
Regele and Philips, Iowa

Injury Response Validated



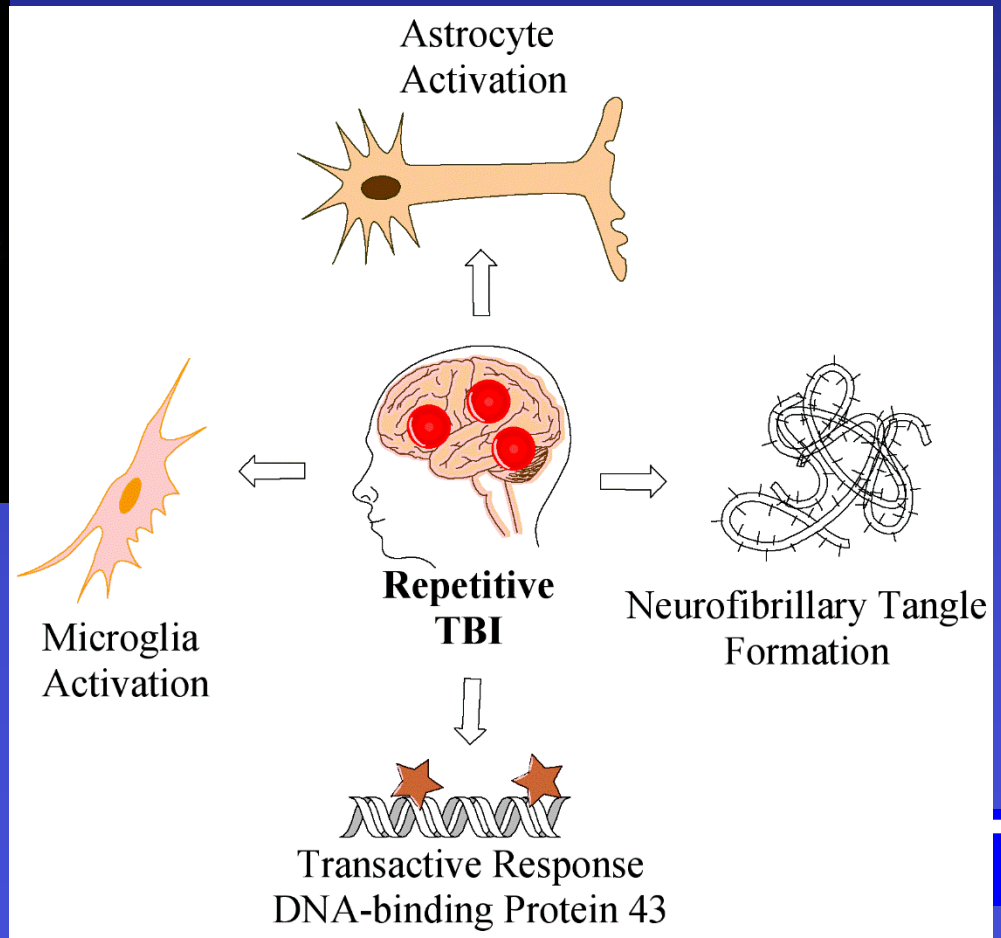




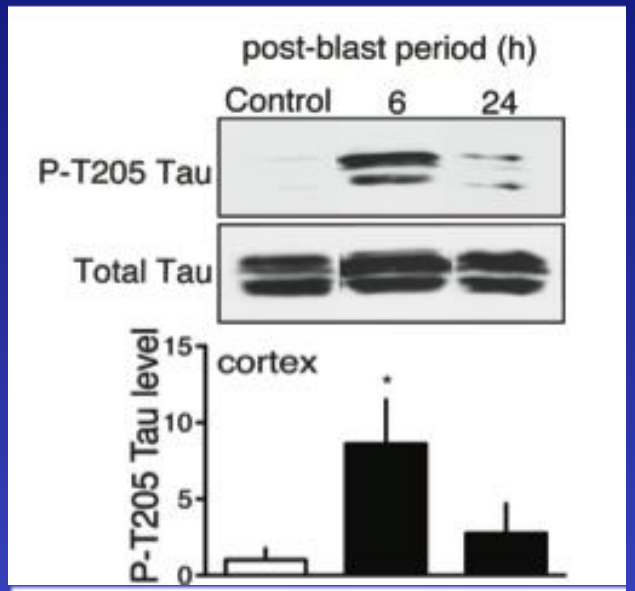


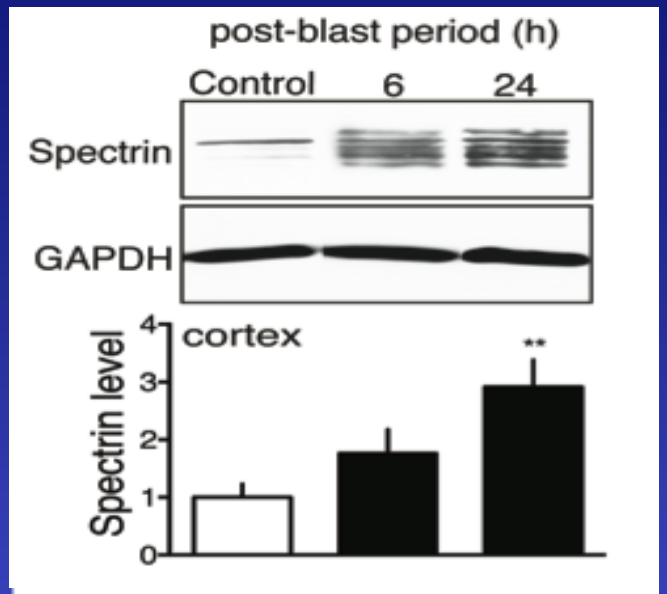
Are We Modeling CTE

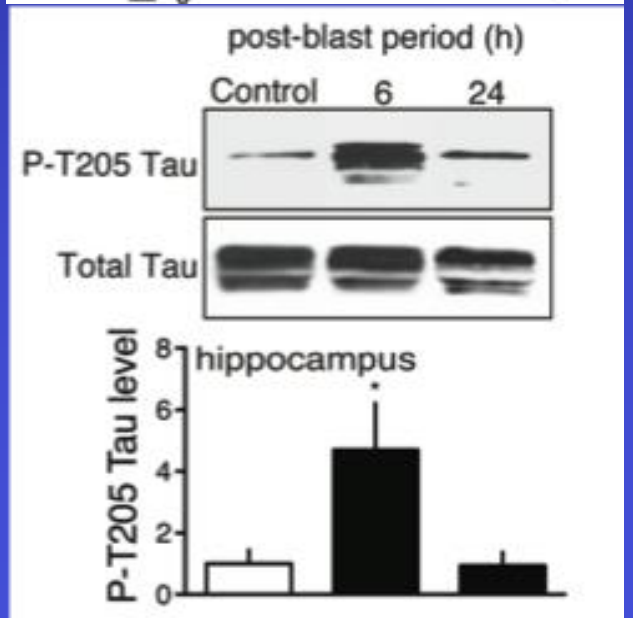
- Poor Impulse Control
- Executive Dysfunction
- Memory Impairment
- Suicidal Ideation

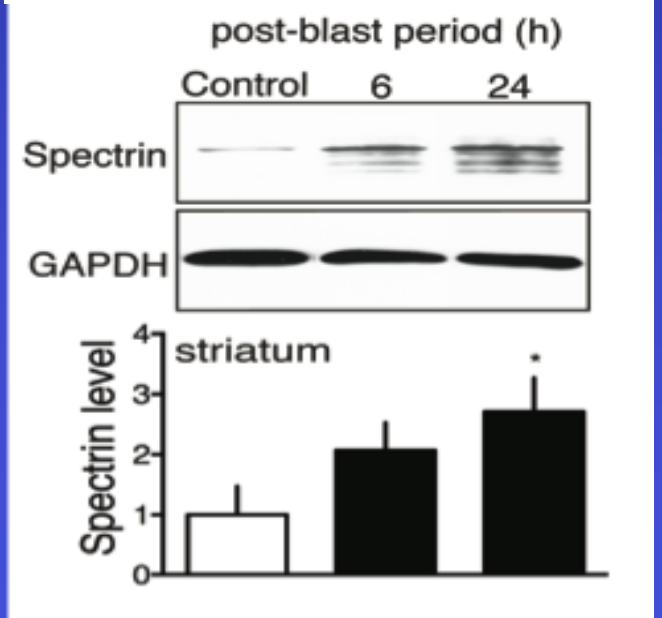


Blast Leads to Tau Phosphorylation

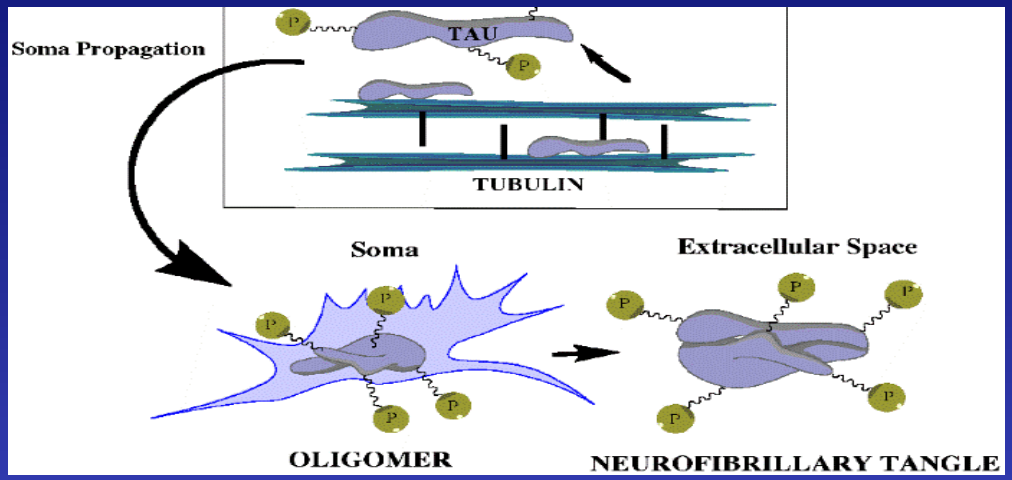








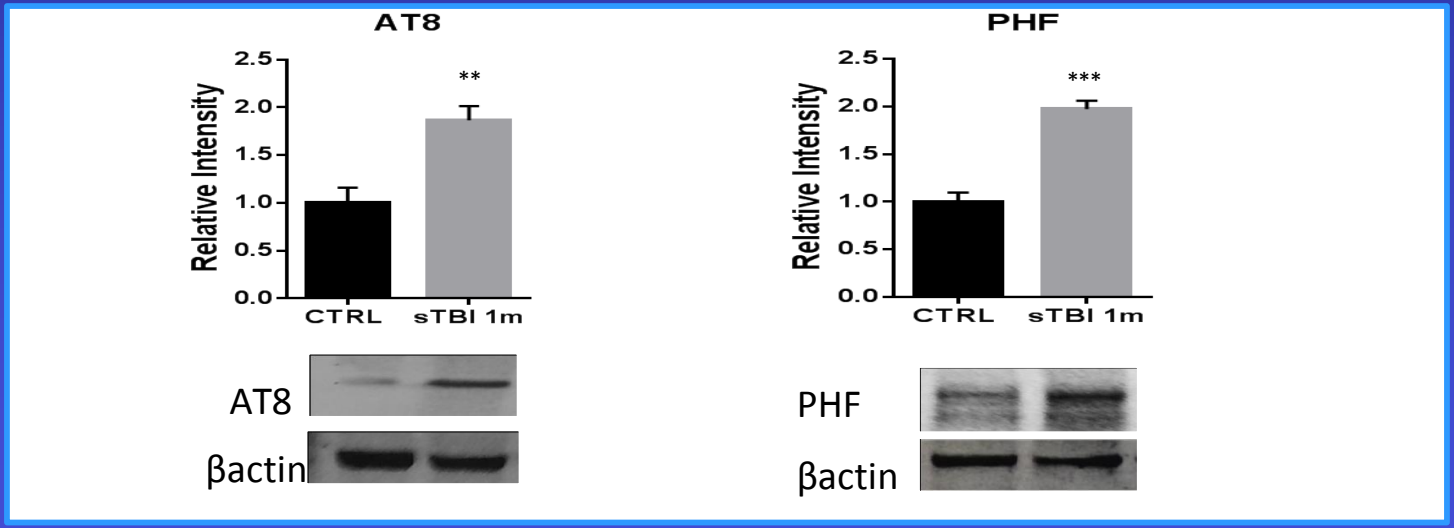
Tau Phosphorylation Persists after TBI



Adapted from
Lucke-wold et al., 2014

Tau Phosphorylation Site Threonine 205

Tau Conformational Change Marker



**p<0.01; ***p<0.001; Student's *t* test

What Causes Tauopathy

Journal of Alzheimer's Disease 28 (2012) 839–854
DOI 10.3233/JAD-2011-111037
IOS Press

Endoplasmic Reticulum Stress Induces Tau Pathology and Forms a Vicious Cycle: Implication in Alzheimer's Disease Pathogenesis

Journal of Pathology
J Pathol 2012; **226:** 693–702
Published online in Wiley Online Library
(wileyonlinelibrary.com) DOI: 10.1002/path.3969

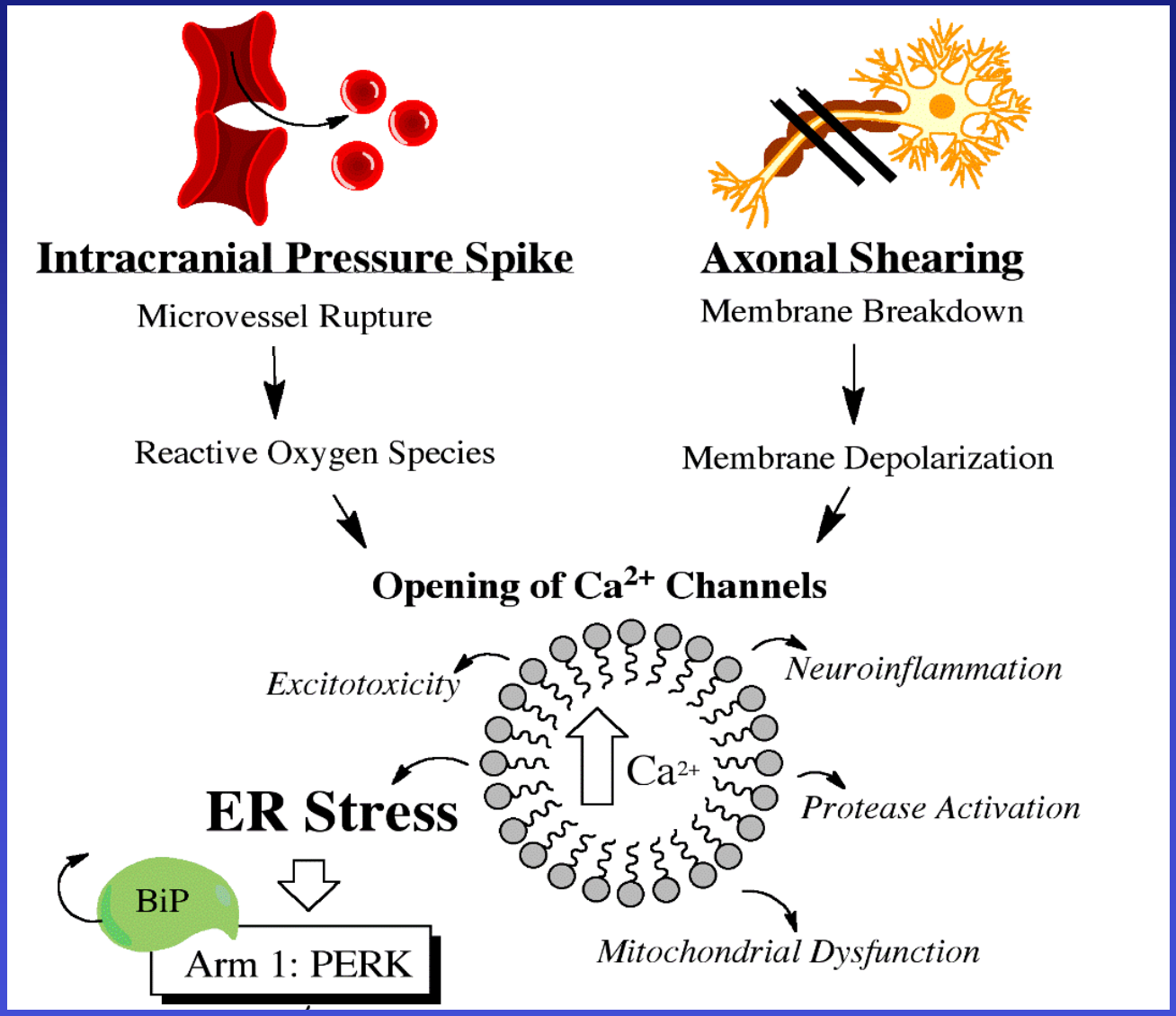
ORIGINAL PAPER

The unfolded protein response is associated with early tau pathology in the hippocampus of tauopathies

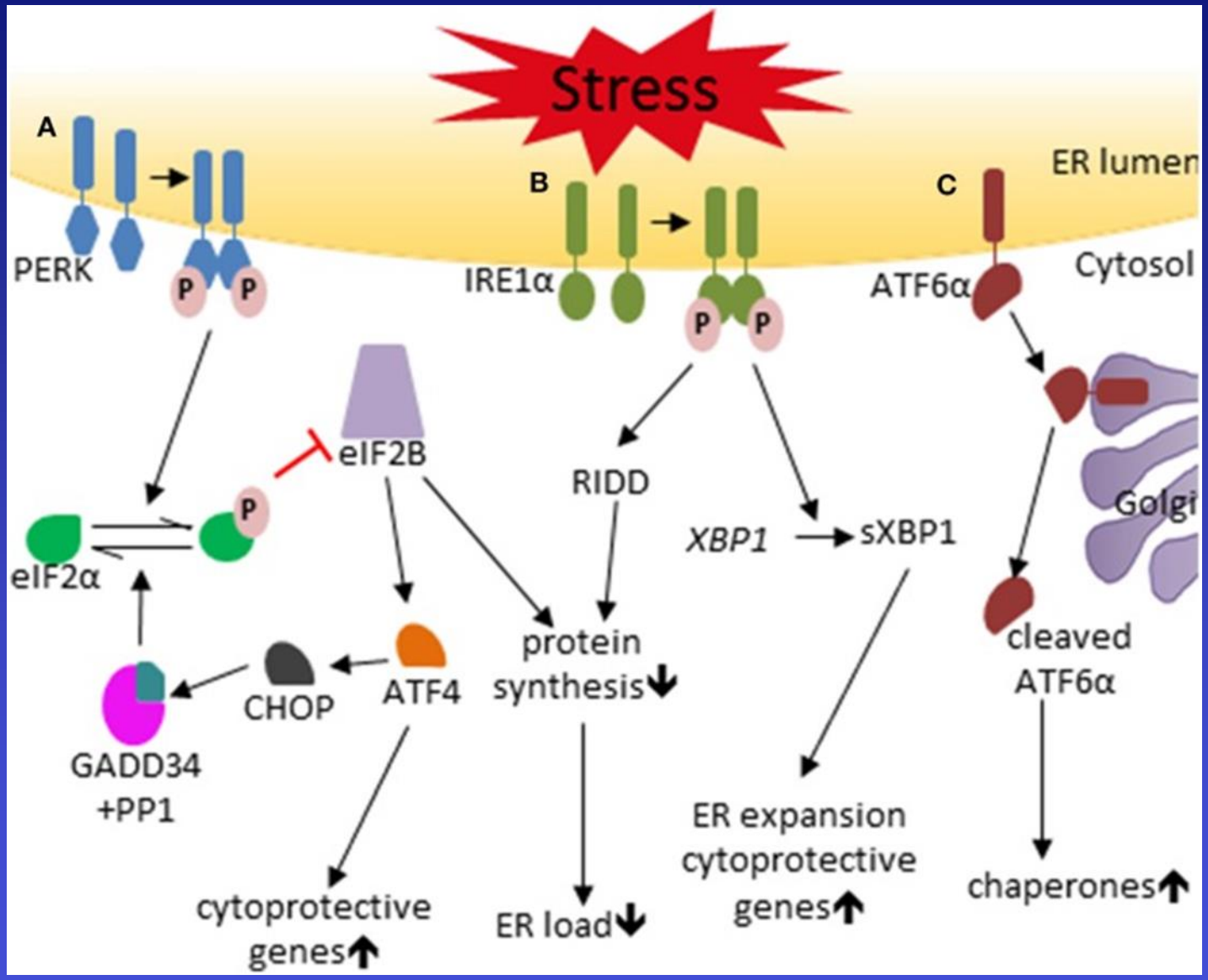
Cell Injury, Repair, Aging and Apoptosis

The Unfolded Protein Response Is Activated in Pretangle Neurons in Alzheimer's Disease Hippocampus

Activation/Modulation of ER Stress

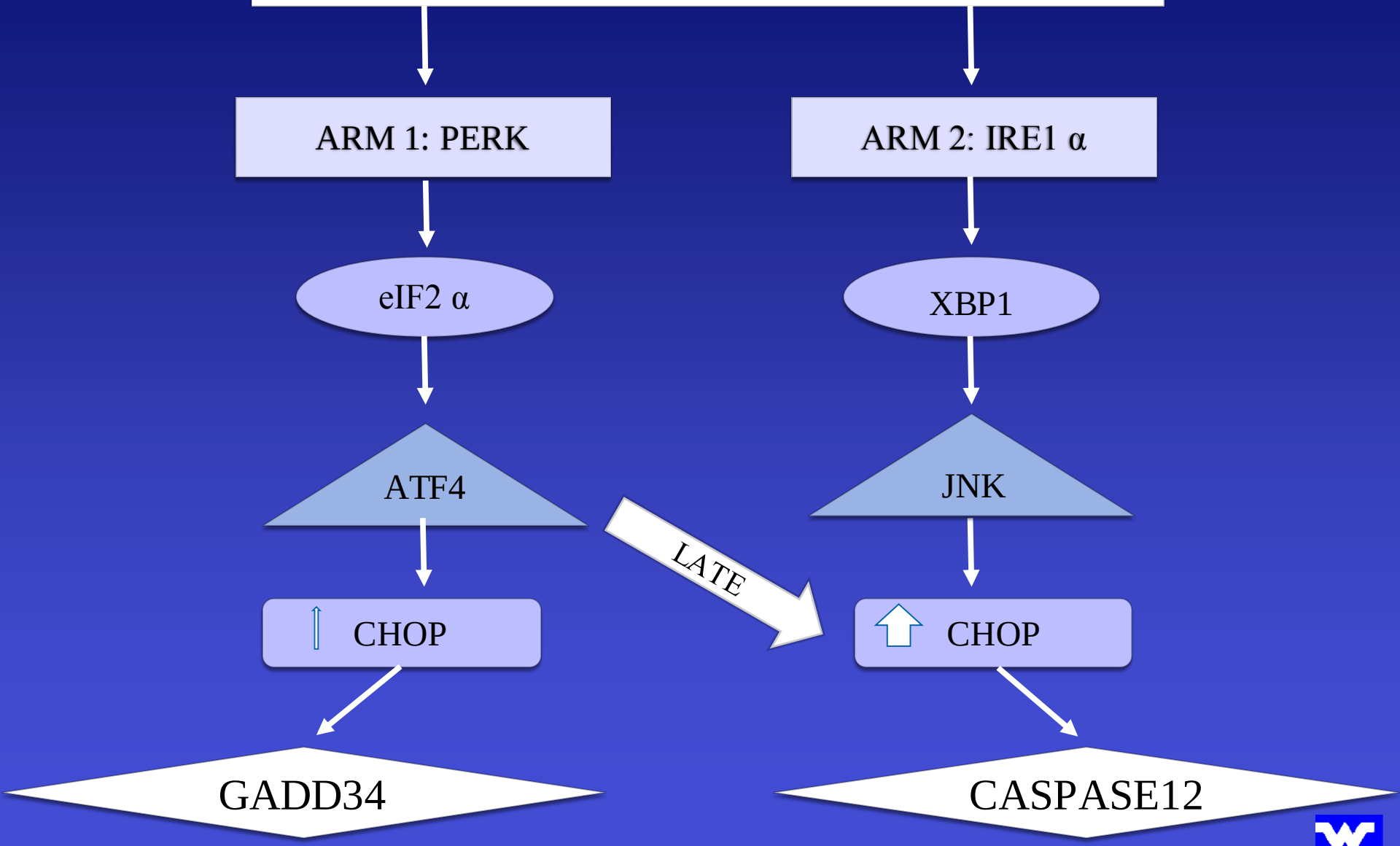


What is ER Stress

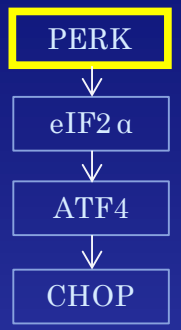


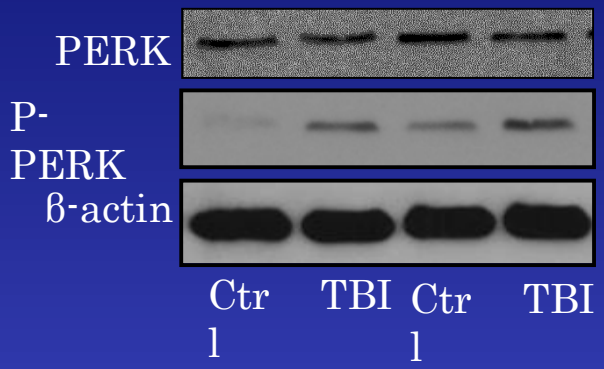
Stone and Lin, Frontiers in Neuroscience. 2015.

ER Stress Pathway



PERK Phosphorylation

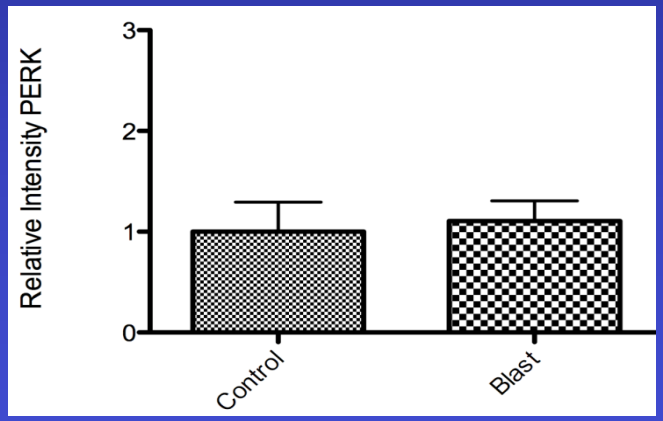


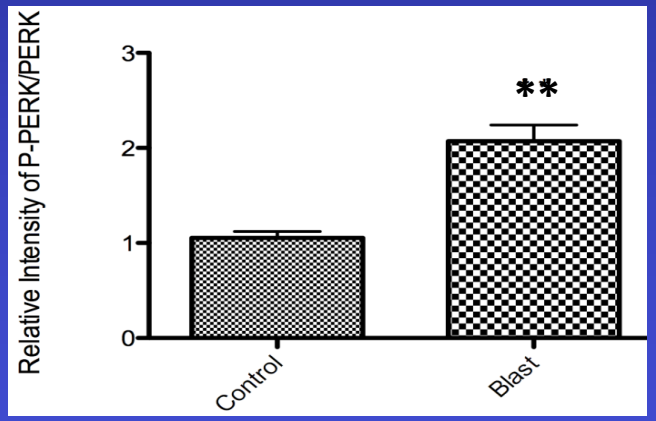


30 min post-TBI

Left Pre-Frontal Cortex

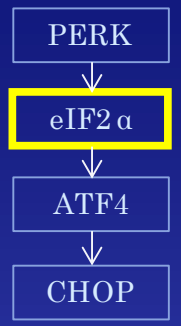
Image J analysis (n=4)

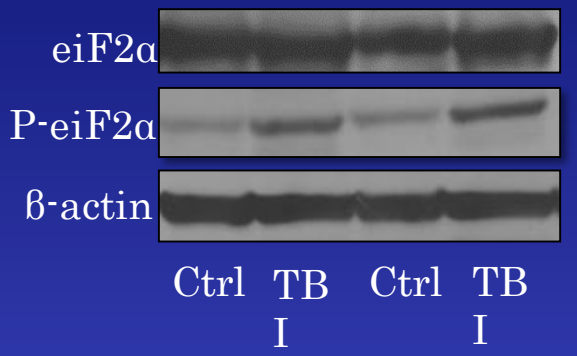




**=
p<0.01

eIF2α Phosphorylation

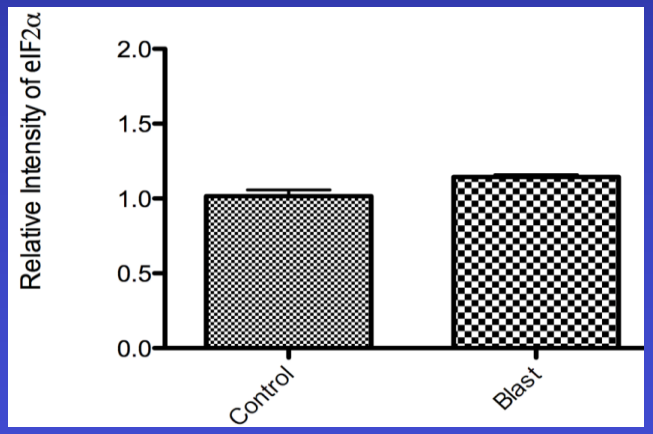


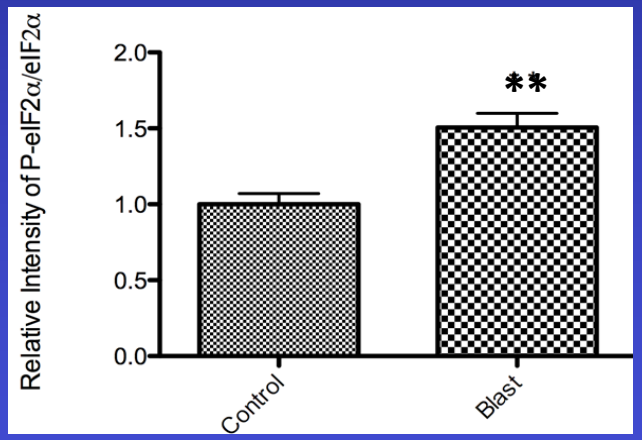


30 min post-TBI

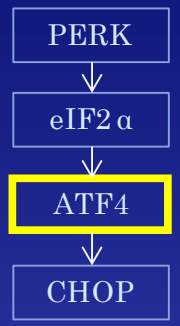
Left Pre-Frontal Cortex

Image J analysis (n=4)

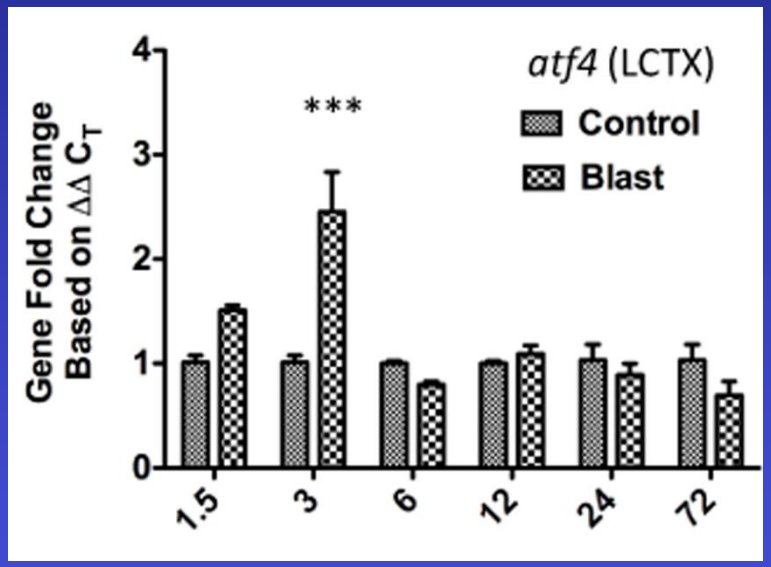


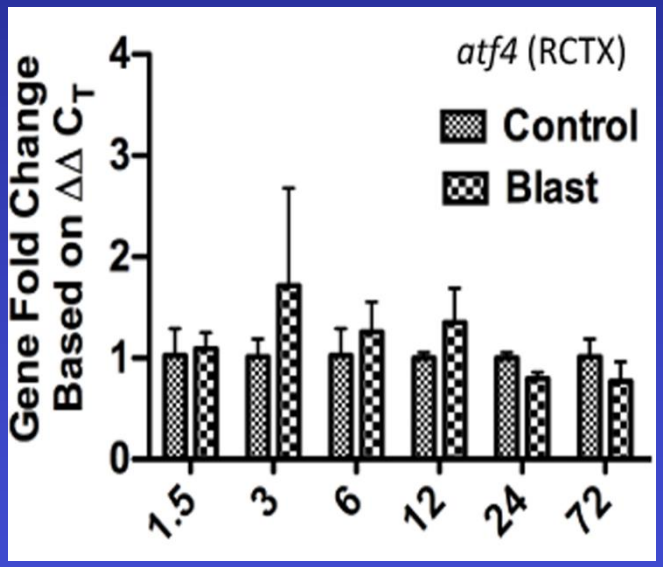


**= $p<0.01$

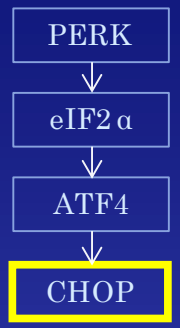


atf4 Gene Expression Time Course

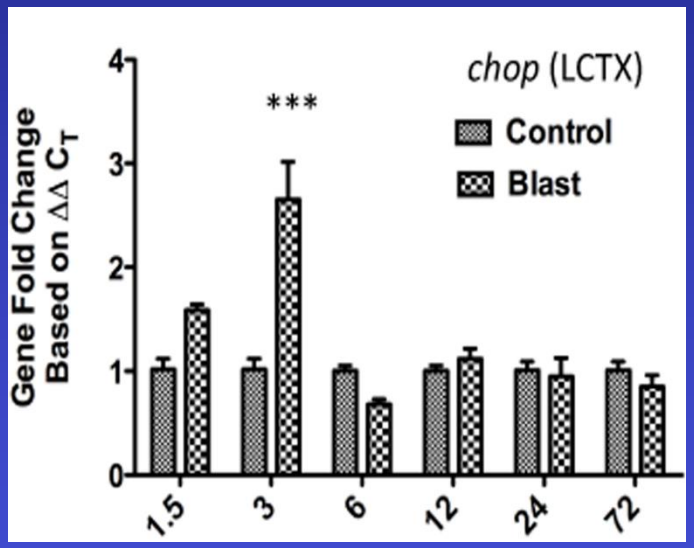


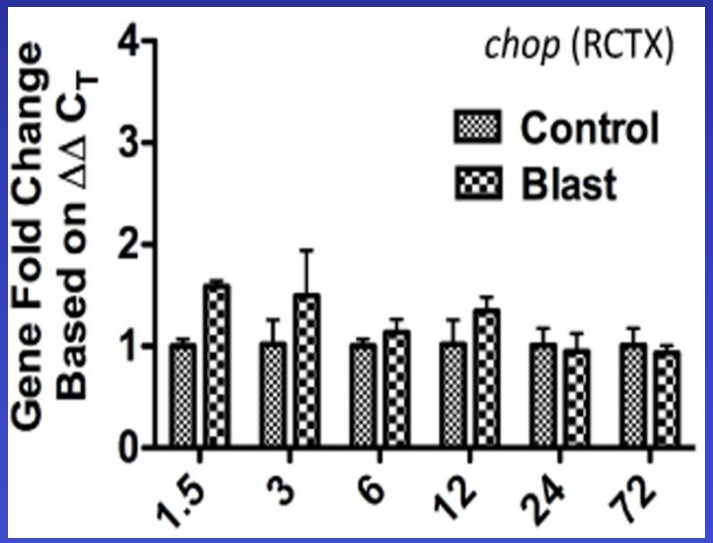


***= p<0.001



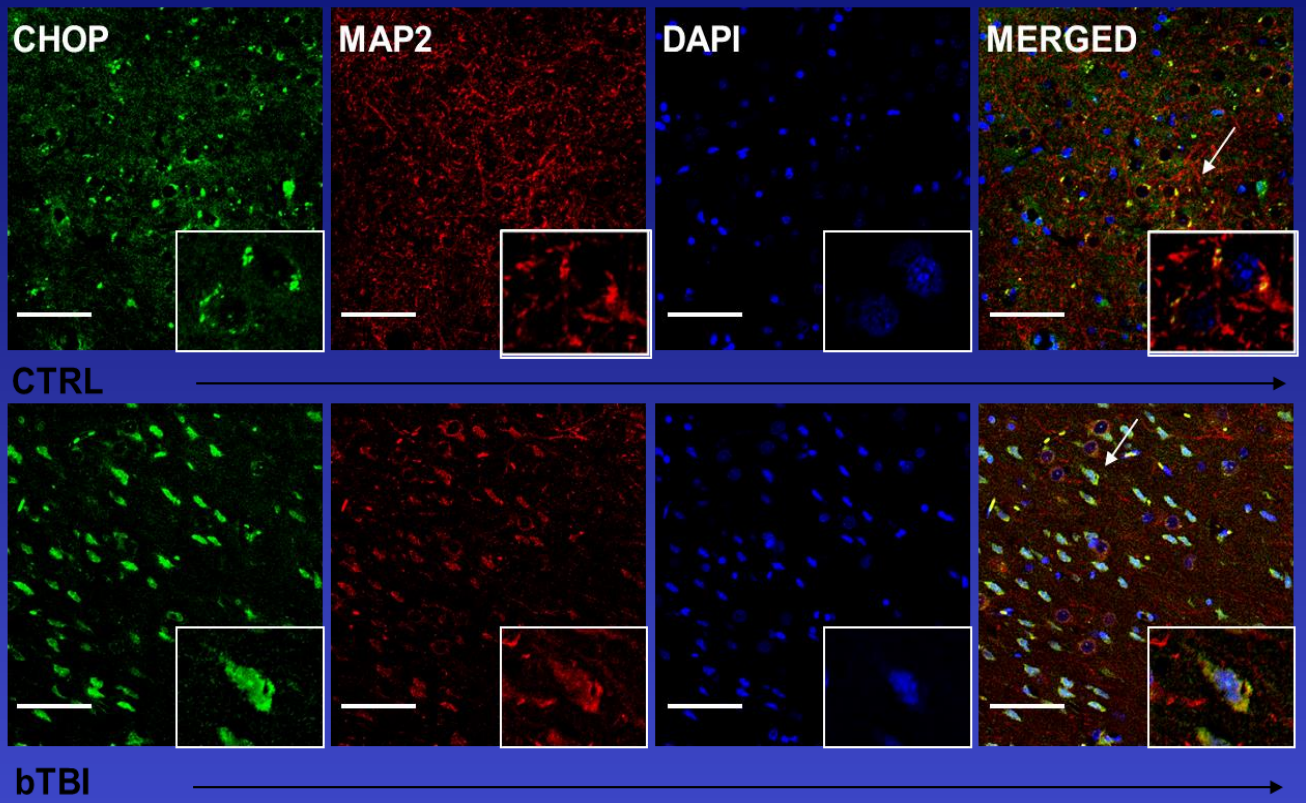
chop Gene Expression Time Course

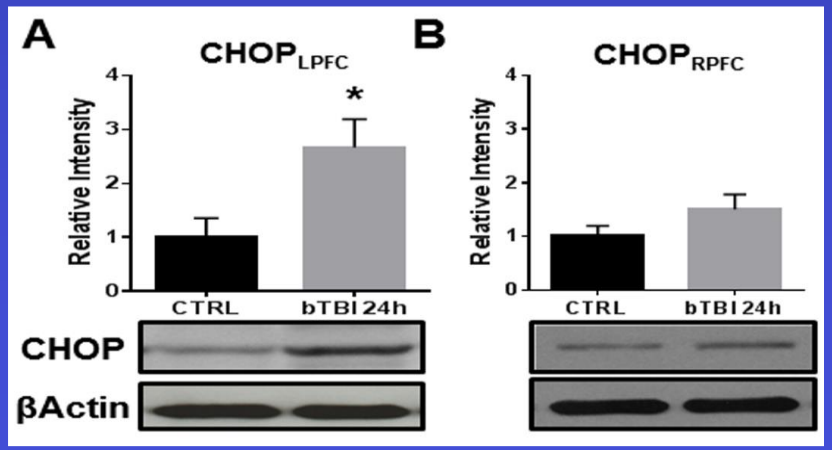




***=
p<0.001

ER stress in Left Cortical Neurons after Blast





Modeling CTE-like phenotype

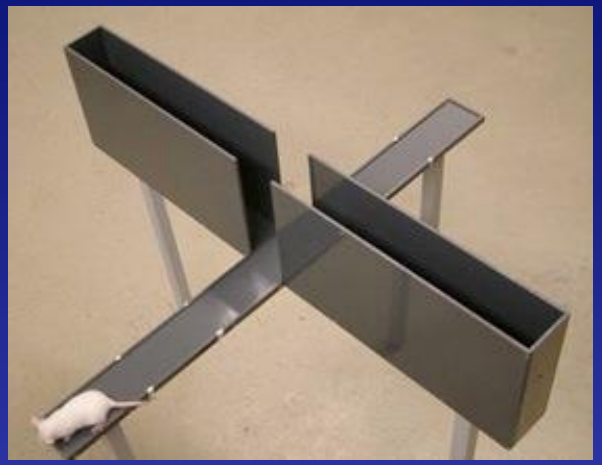
Elevated Plus Maze

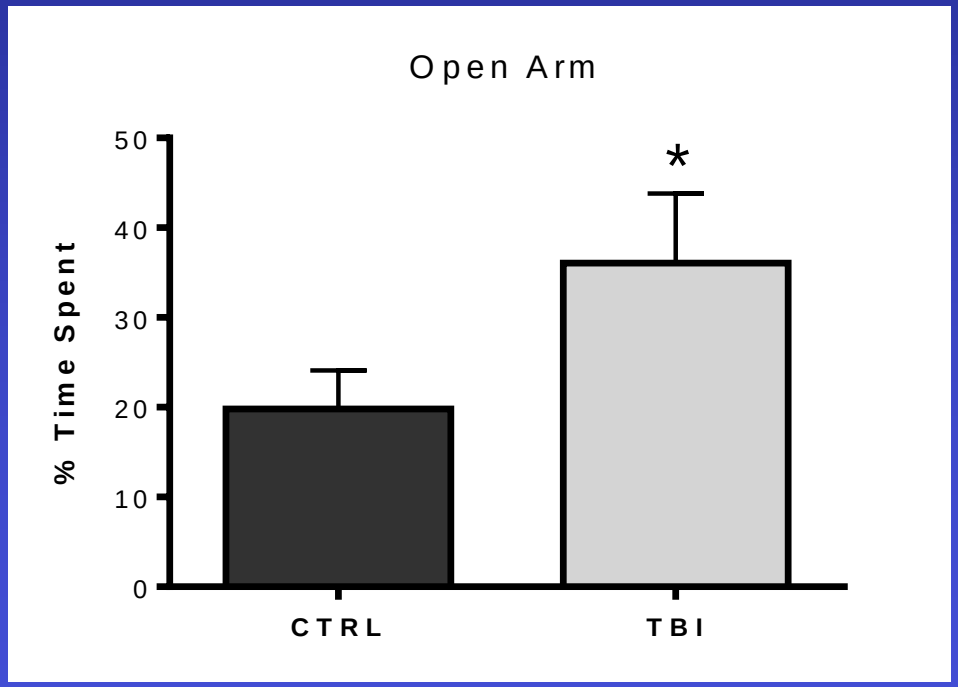
Impulsive-like behavior

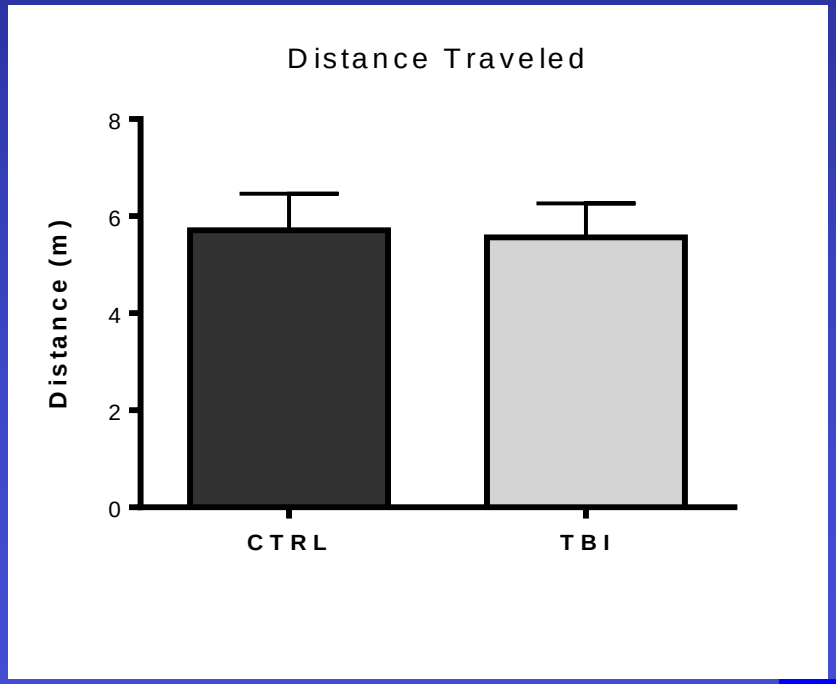
Morris Water Maze

Spatial memory deficits

Elevated Plus Maze

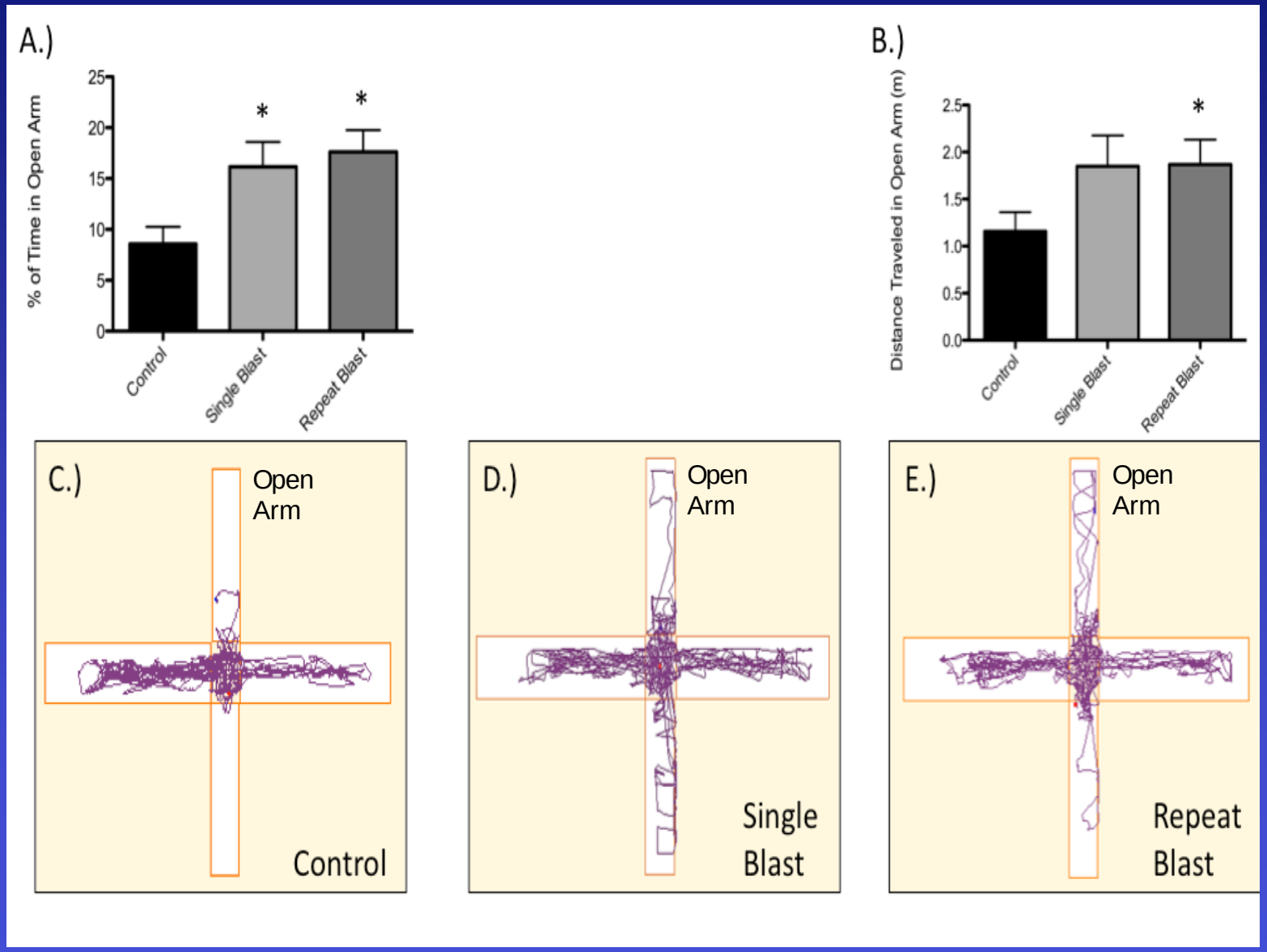




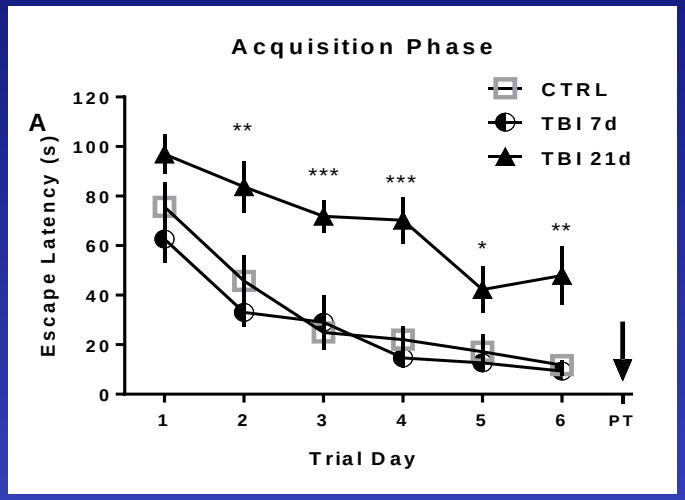


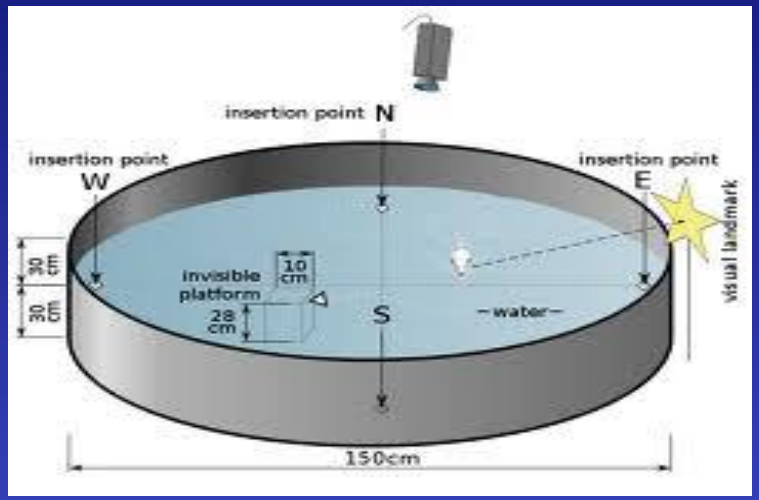
*= p<0.05

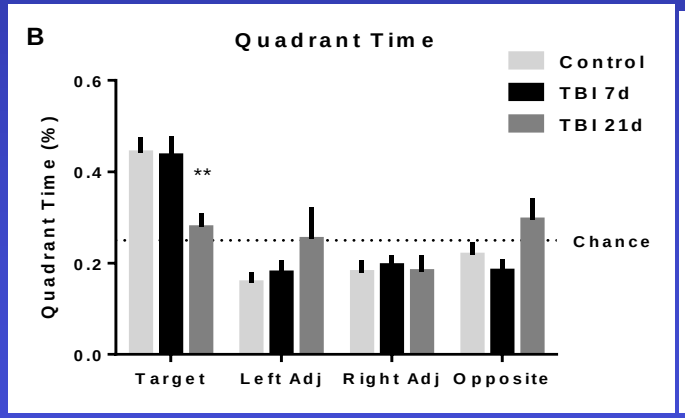
Elevated Plus Maze

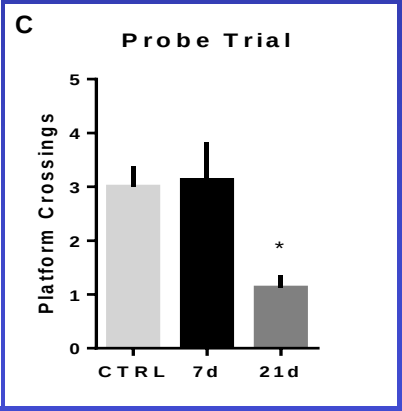


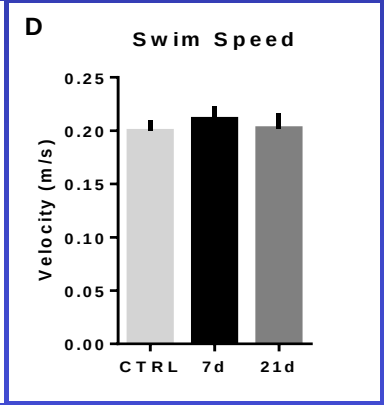
MORRIS WATER MAZE







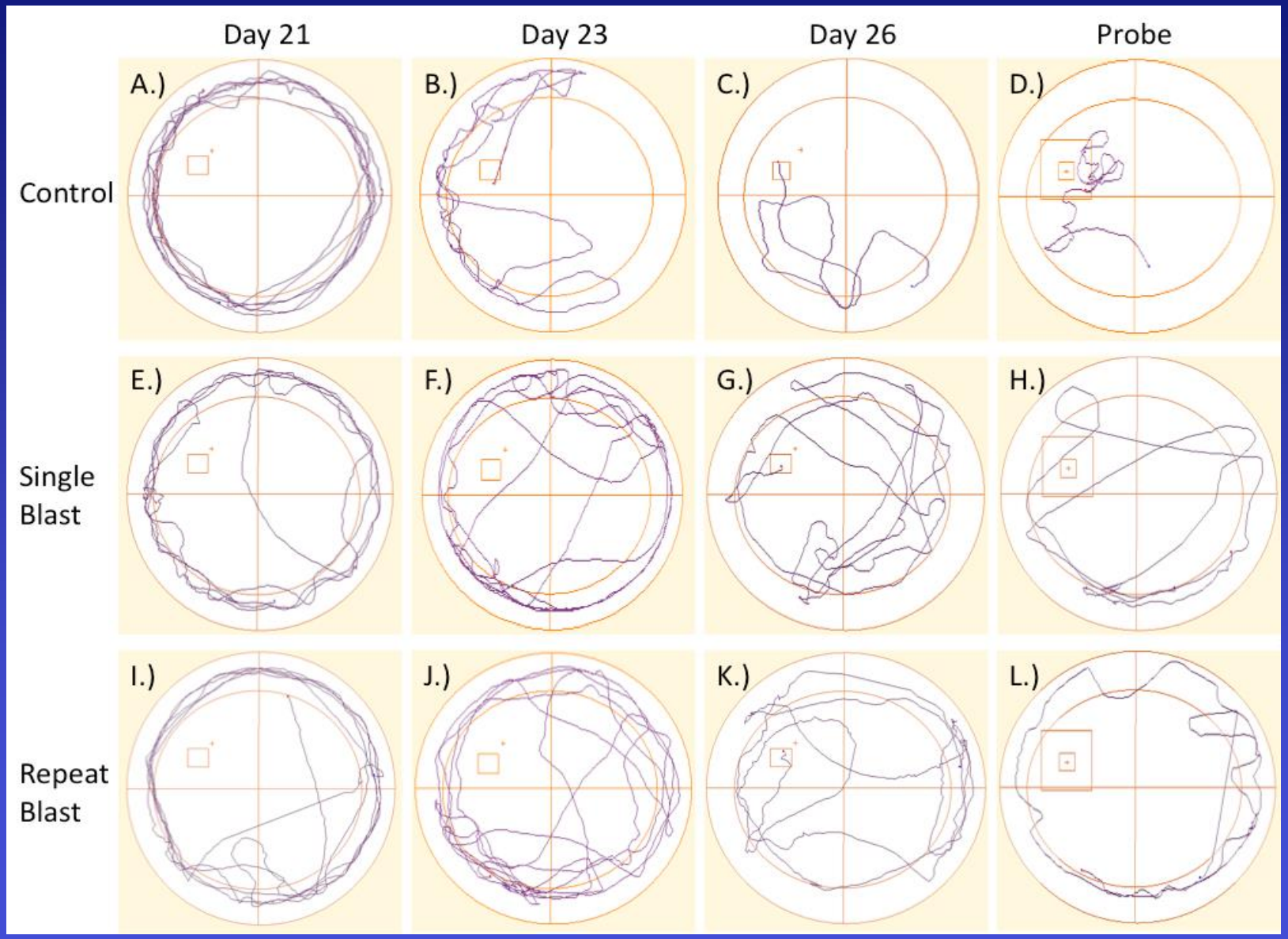




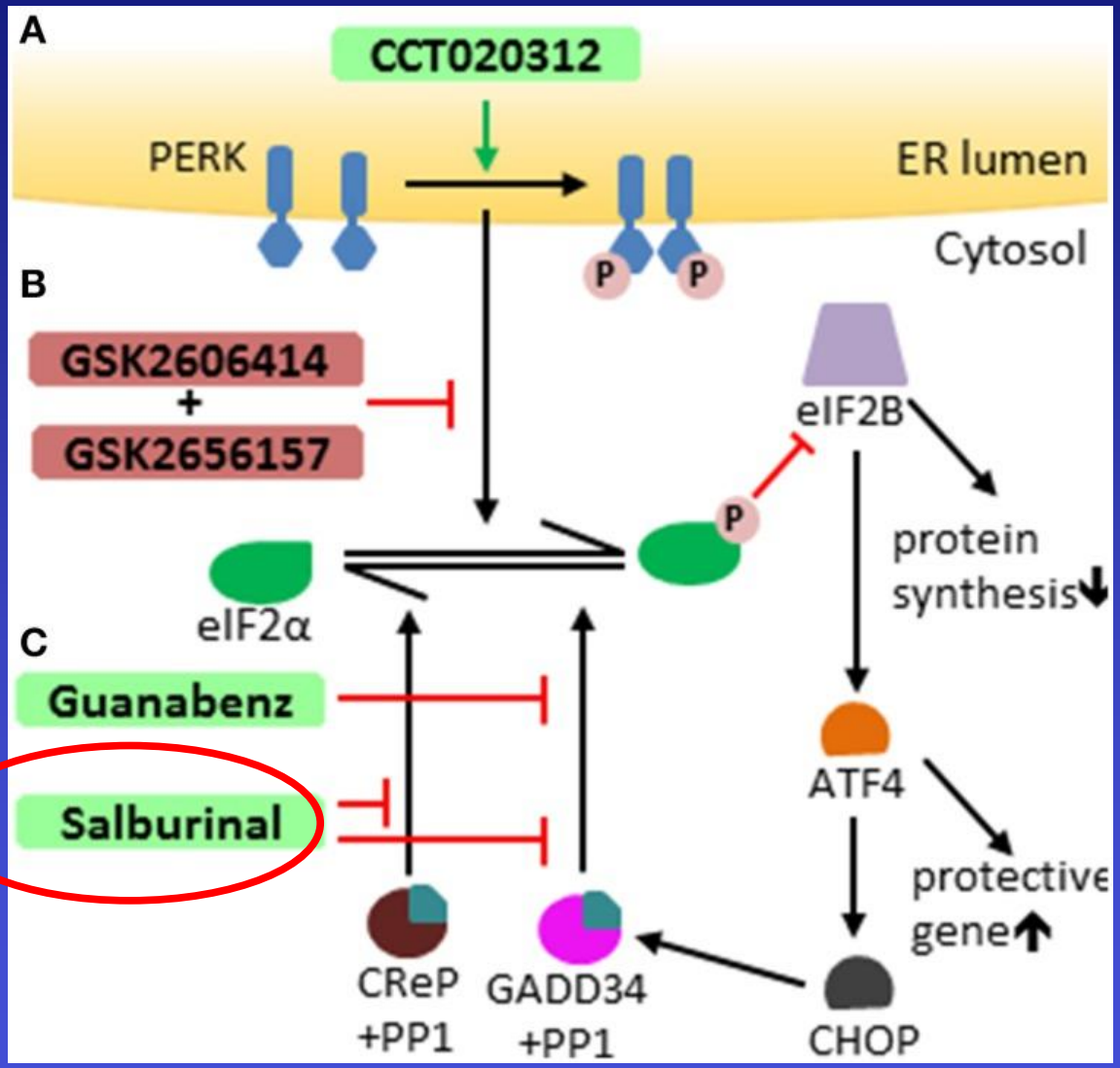
n = 8; *p<0.05, **p<0.01, ***p<0.001

Logsdon, Tan & Hernandez, 2014, *In progress*

MORRIS WATER MAZE

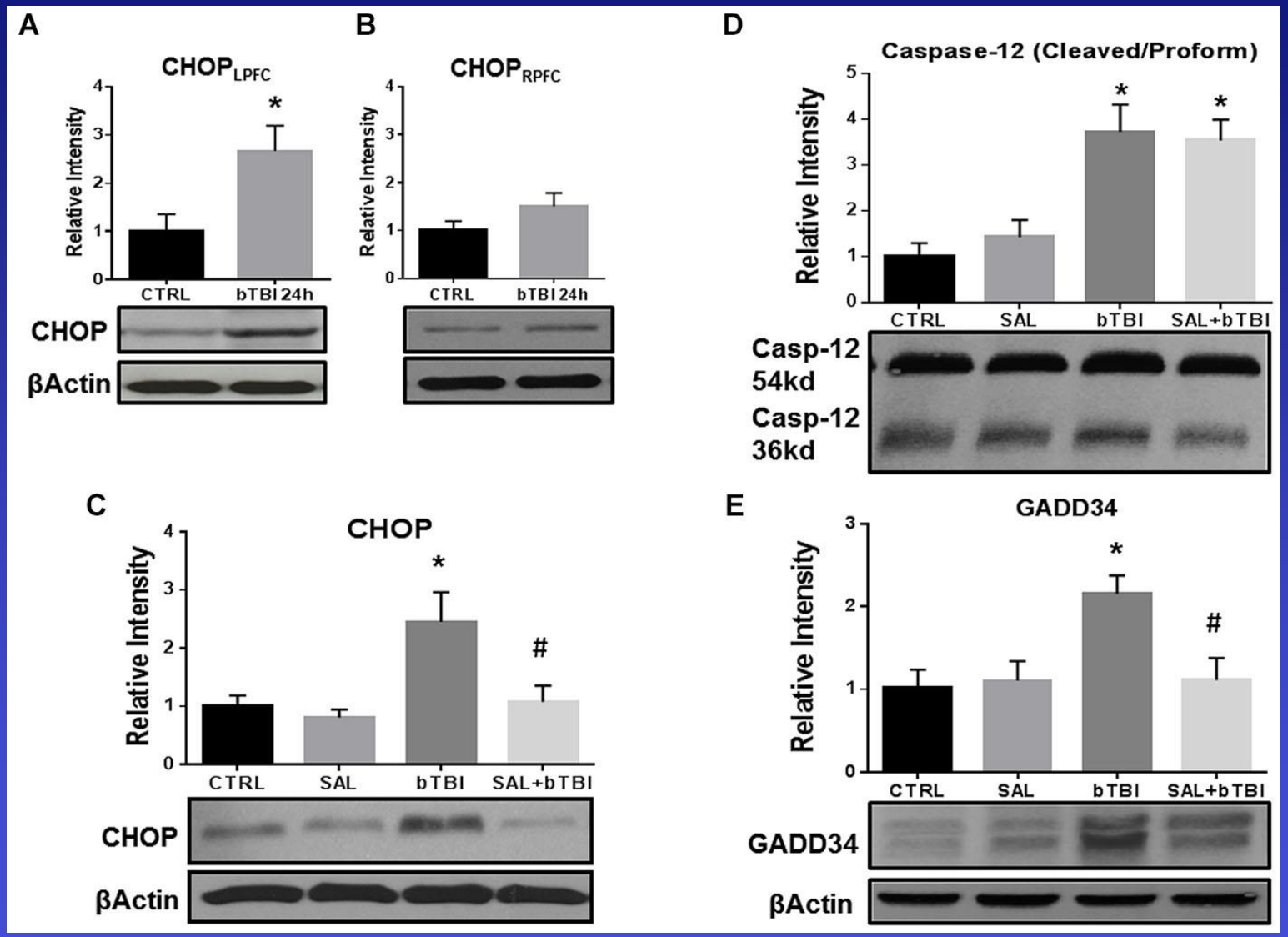


Inhibit PERK Cascade

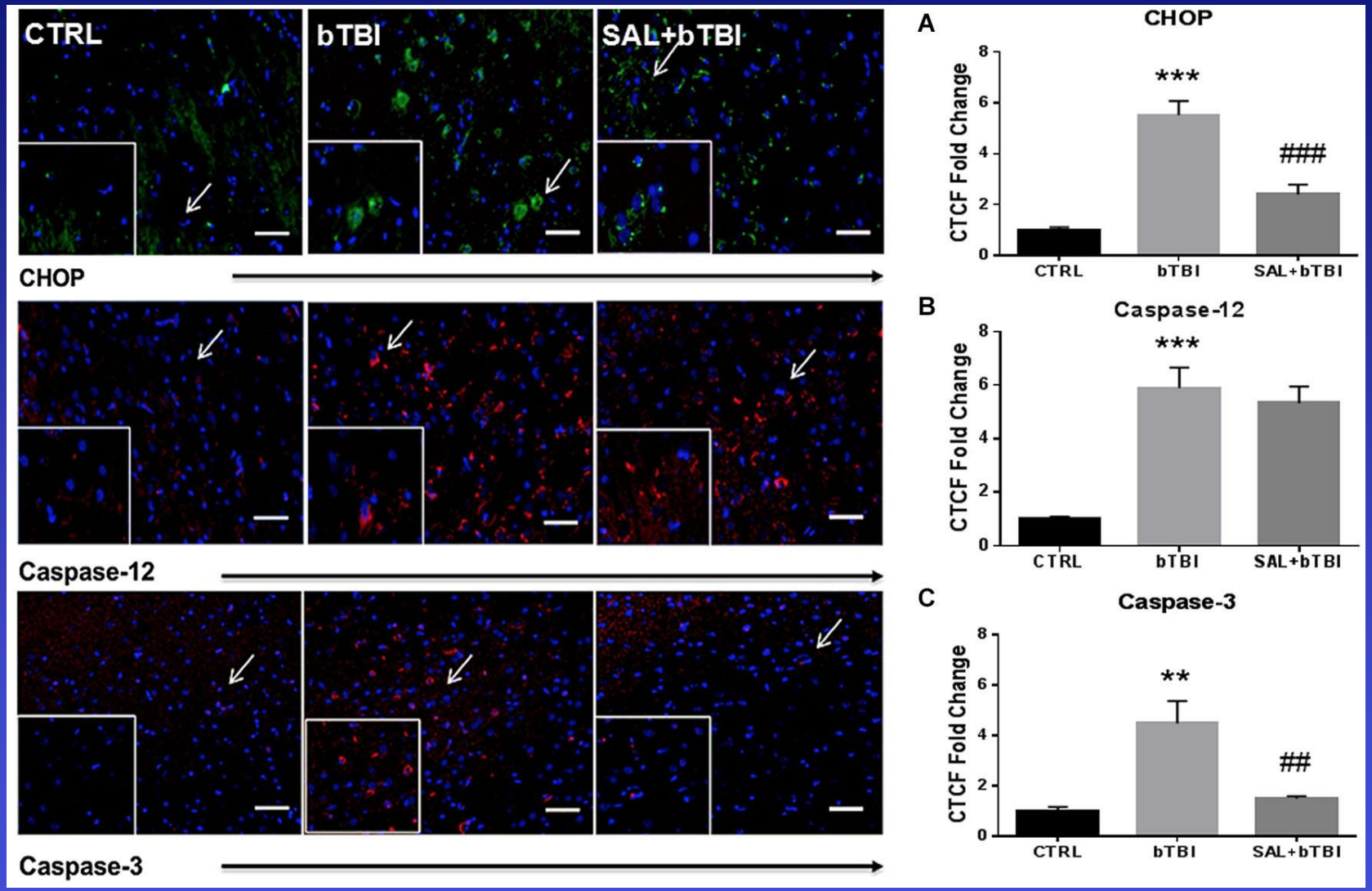


Stone and Lin, Frontiers in Neuroscience. 2015.

Inhibit PERK Cascade

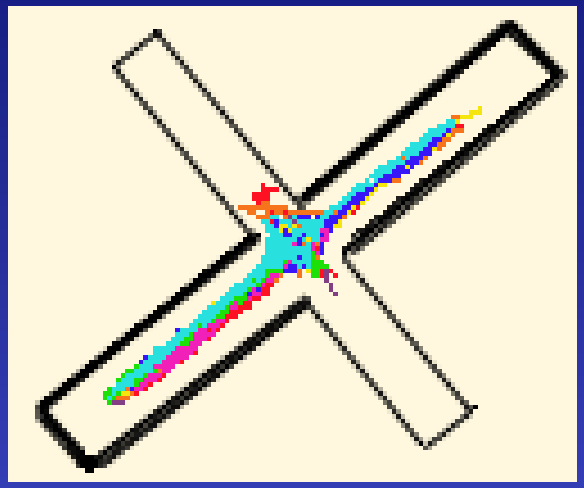


Inhibit PERK Cascade

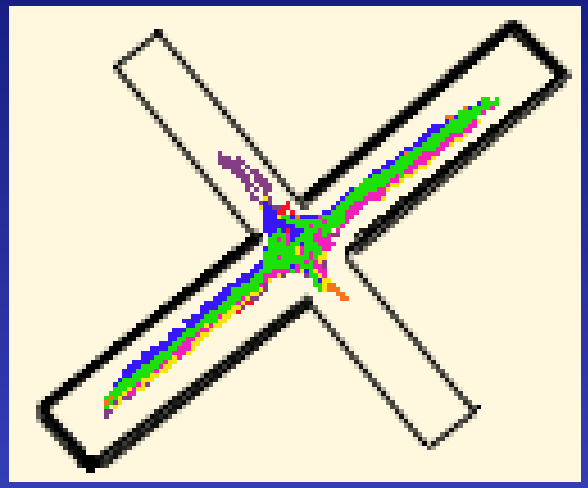


The Behavior Follows

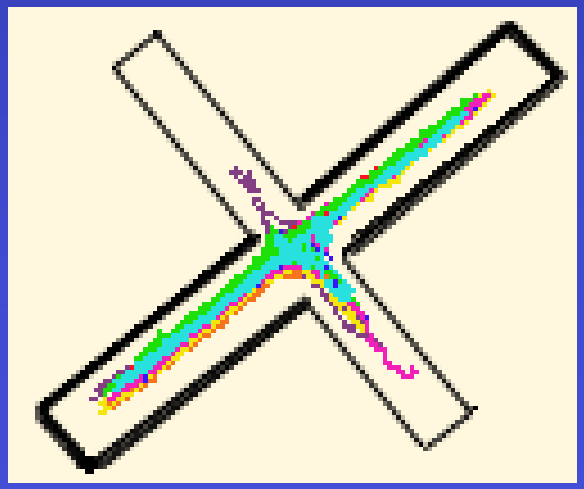
Control



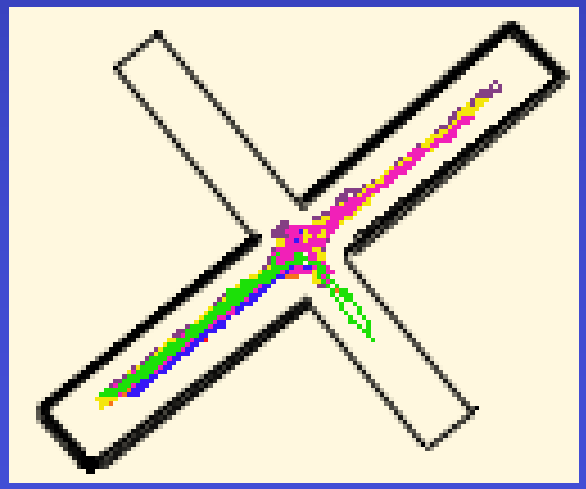
Salubrinal



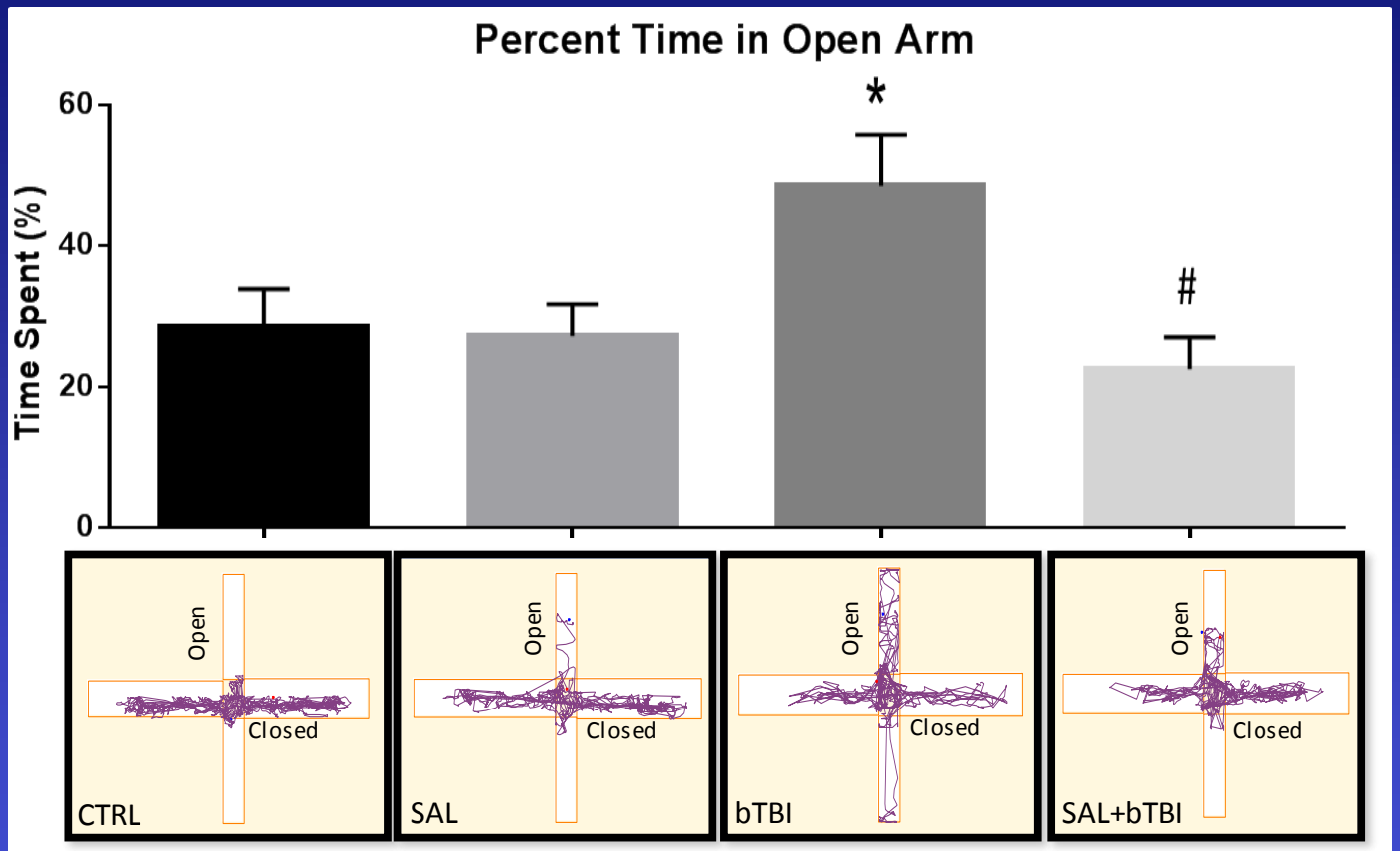
TBI



TBI+ Salubrinal



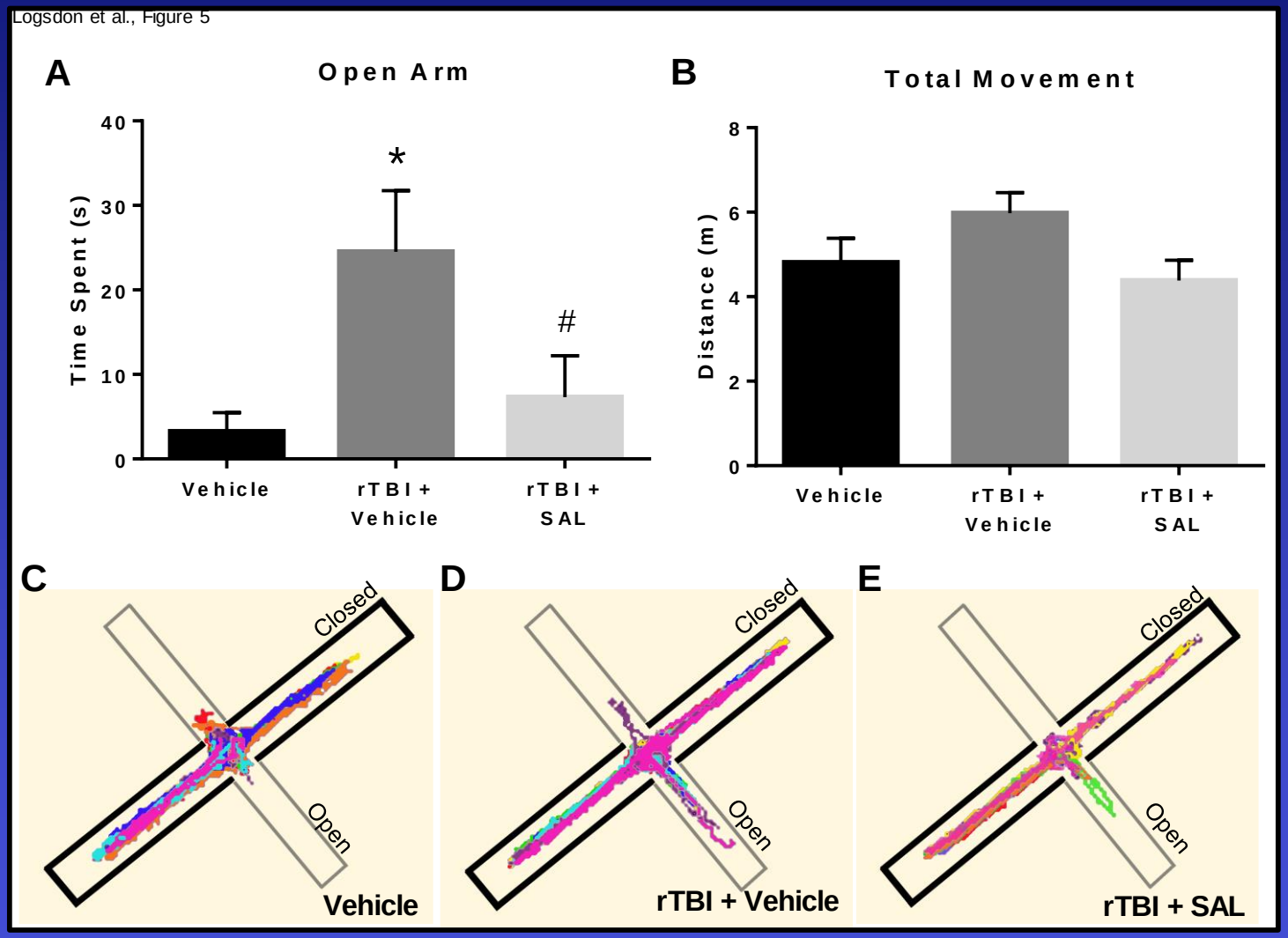
The Behavior Follows



Elevated Plus Maze, n = 12; *p < .05, #p < 0.05

Blast injury to the prefrontal cortex exhibits impulsive-like behavior in rat;
Salubrinal attenuates this behavior by balancing CHOP, GADD34, & Caspase-3 (Cellular Fate)

Even for Repeat Blast



Conclusions

- For the first time, we have shown that ER stress is involved in bTBI.
- We propose that BBB disruption correlates with ER stress activation (data not shown today).
- CHOP was pushed beyond threshold and localized to neurons but not astrocytes.
- The ER stress response occurred prior to and may possibly play a role in astrocyte activation.
- bTBI (single and multiple) significantly increases impulsive-like behavior within 7 days.
- bTBI produces spatial memory deficits at 21 days.
- Modification of the ER stress response may prove beneficial in promoting neuronal protection and ameliorating abnormal behavior.

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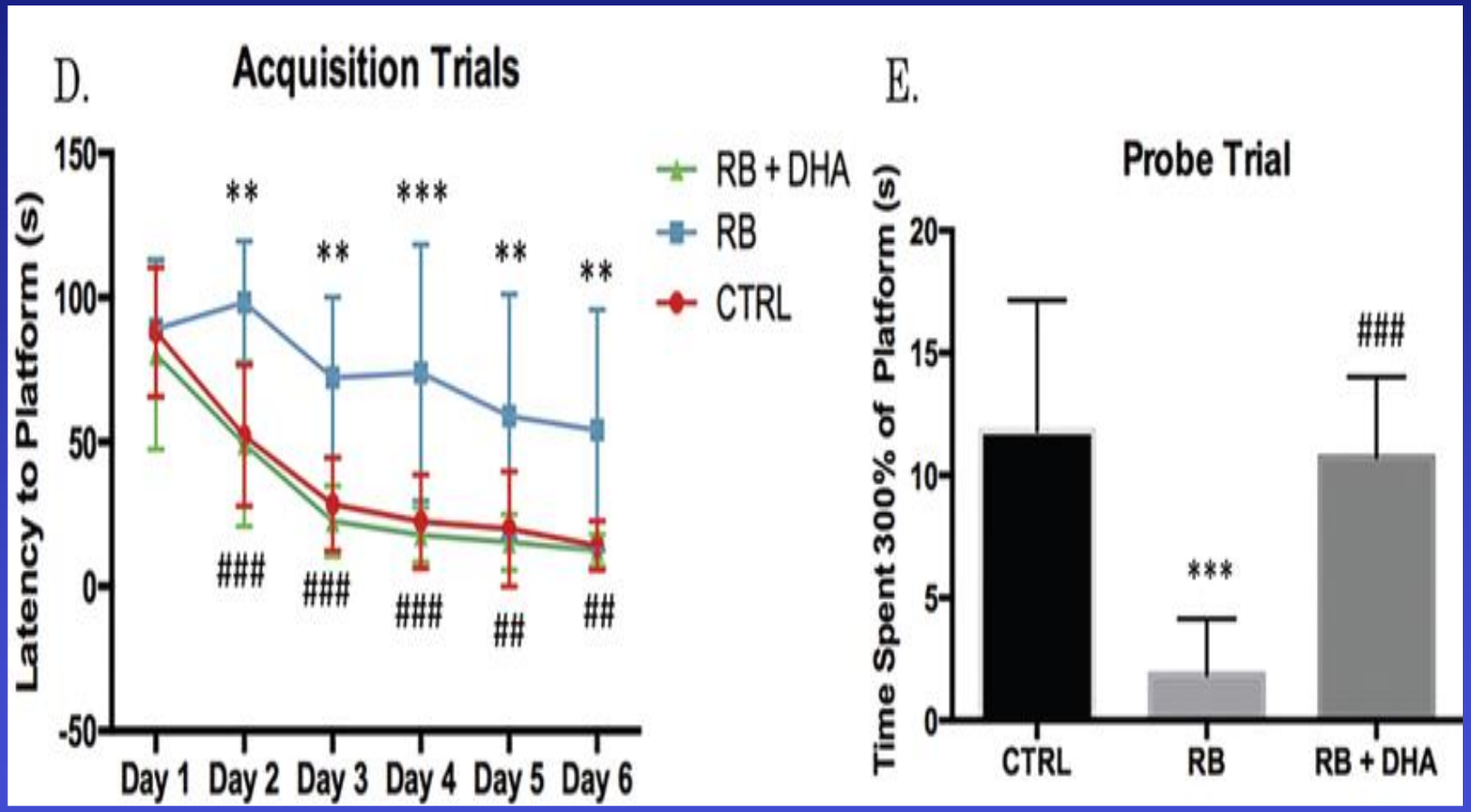
- **Aric Logsdon**
- **Brandon Lucke-Wold**
- Xinlan Li, MD
- Zhenjun Tan, MD
- Melissa Torres-Altoro
- Michael Philips

Questions?

Extra Slides

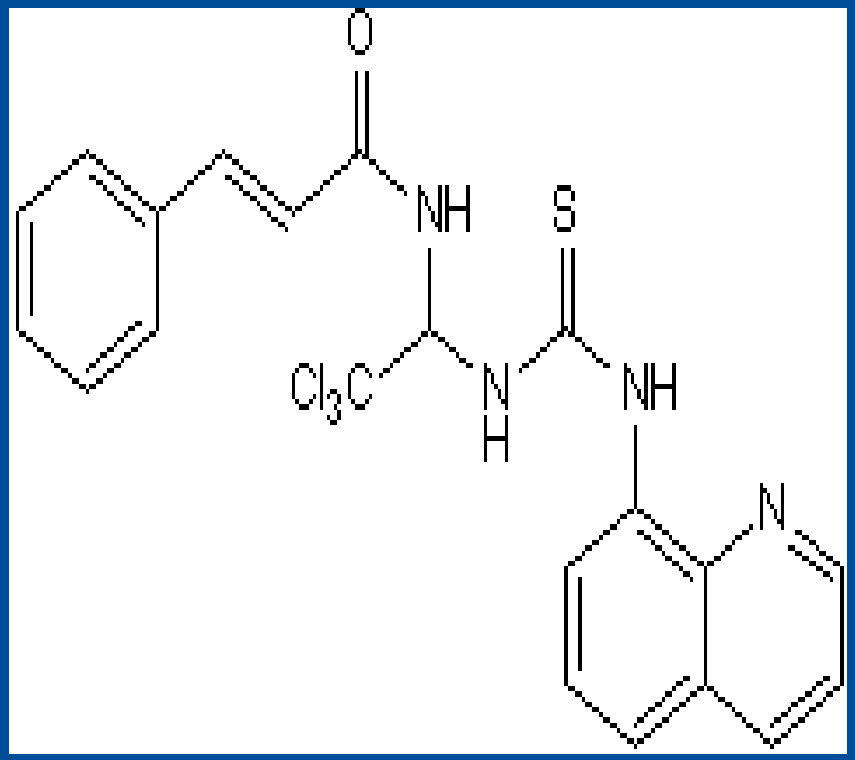
ER stress Modulation with DHA Improves Cognitive Performance



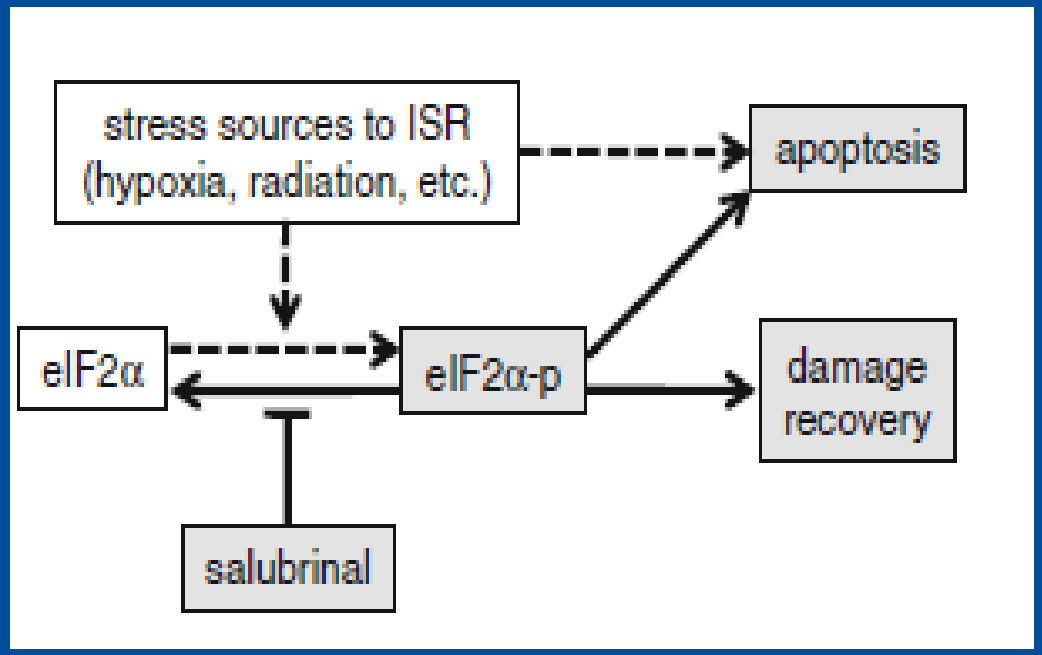
Pharmacological Manipulation

Salubrinal

3-Phenyl-*N*-[2,2,2-trichloro-1-[[(8-quinolinylamino)thioxomethyl]amino]ethyl]-2-propenamide

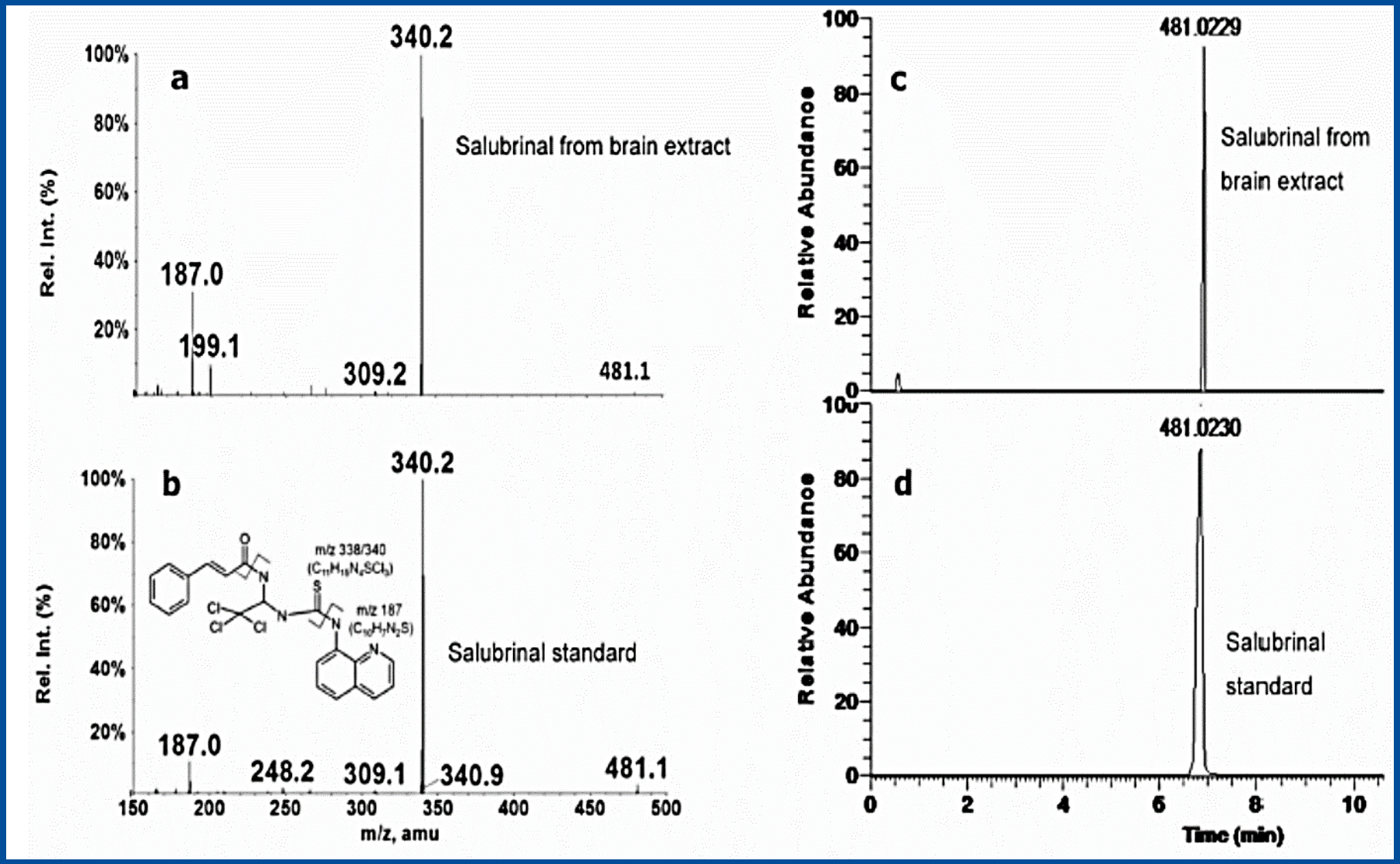


Injection: I.P. *30 mins prior to TBI
Vehicle: 10% DMSO in Saline
Half-Life: ~1.2 h
* (Sokka et al., J. Neuro 2007)



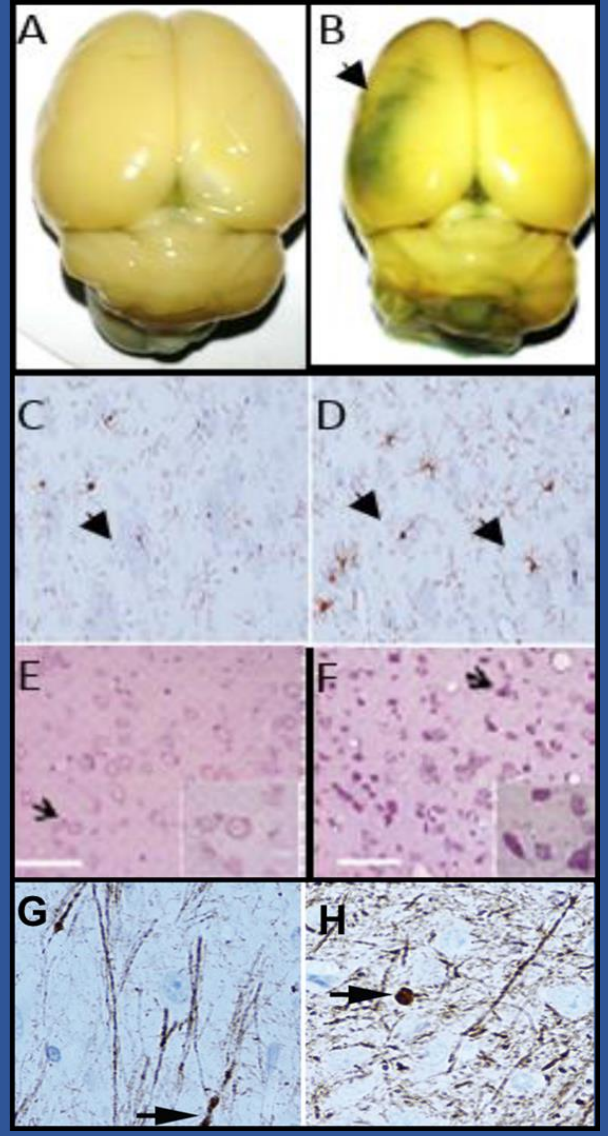
Zhang et al., J. Bone Min. Metab., 2012

Salubrinal Gets into the Brain



LC-MS confirmation of salubrinal in mouse brain tissue following intraperitoneal administration

Moreno et al., Nature 2012



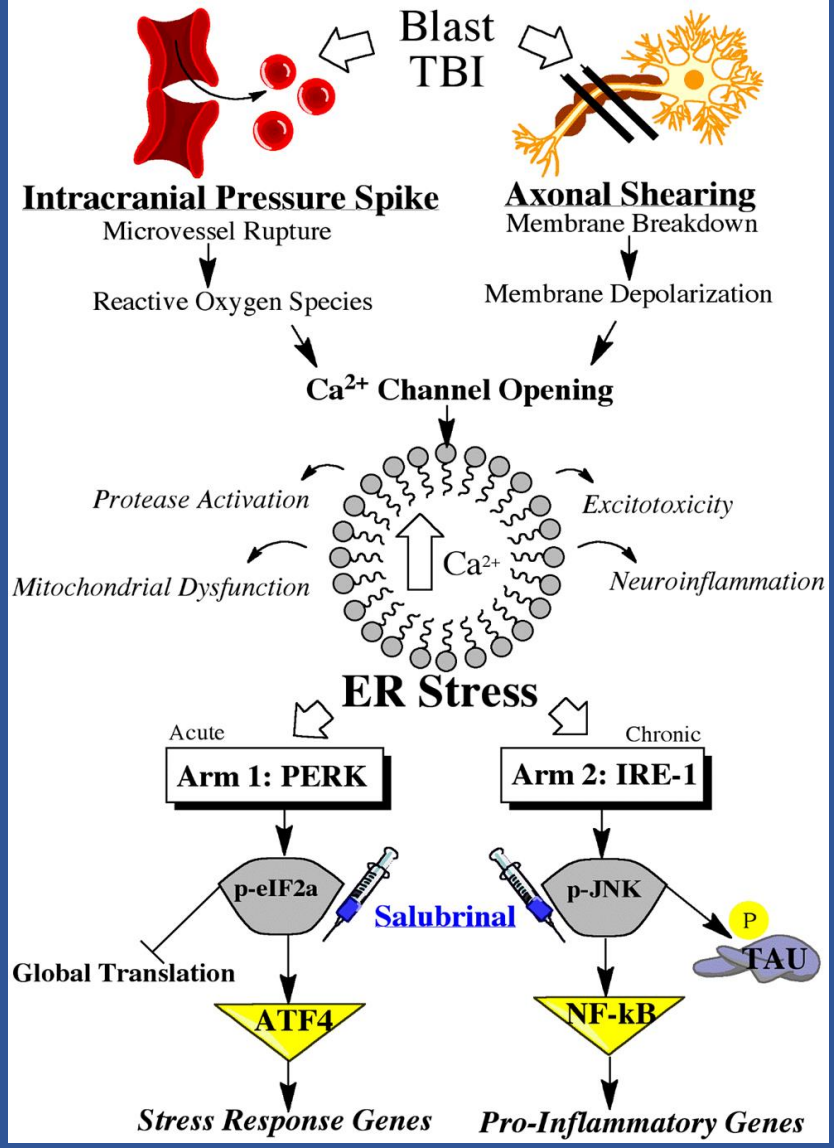


FIGURE 2 | (A) Evan's blue-albumin extravasation in left brain parenchyma of CTRL, **(B)** and at 6h post-bTBI. **(C)** Staining for active astrocytes (GFAP) in CTRL, **(D)** and at 72 h post-bTBI. **(E)** Free iron staining in CTRL, **(F)** and at 24 h post-bTBI. **(G)** Axonal injury (pSMI-31) seen at 24 h post-bTBI shows beaded dystrophic axons, **(H)** and axonal bulb formation **(H).** Schematic shows how blast exposure leads to Ca^{2+} perturbation and the ER stress mechanism