

A View of Brain Injury through the Eyes of the Clinical Pathology Laboratory

Frederick G. Strathmann, PhD

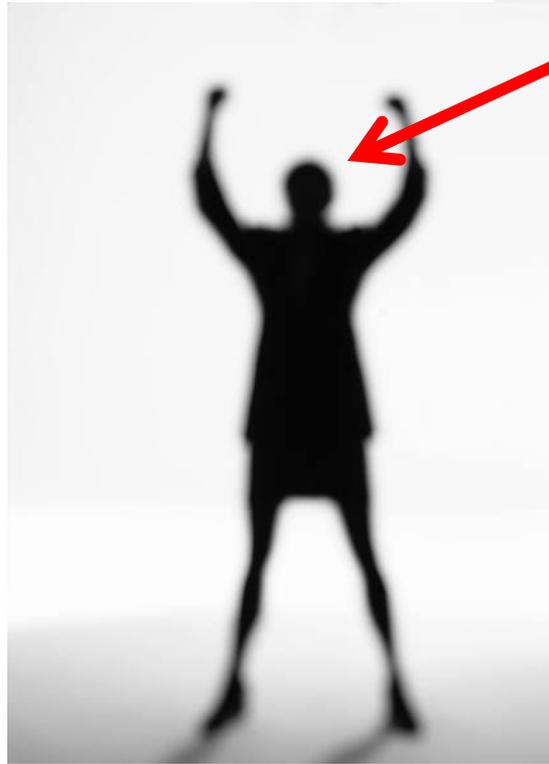
David J. Petron, MD

Pathology Grand Rounds

This is the brain:



It is in your head.



I want to study it.



Disclosures - FStrathmann

- Collaboration for POC device development in TBI:
 - 3PDx
 - PLC Diagnostics Inc.

Objectives

- Describe the clinical workflow for TBI assessment
- State the underlying primary and secondary pathology in TBI
- Discuss the benefits and limitations of existing TBI biomarkers

Outline

Part 1 (Petron)

- Clinical Workflow for TBI assessment
- Presentation of cases

Part 2 (Strathmann)

- TBI Pathology
- Current biomarkers for TBI
- Direction of TBI biomarker research

Concussions

Feb 15, 2013

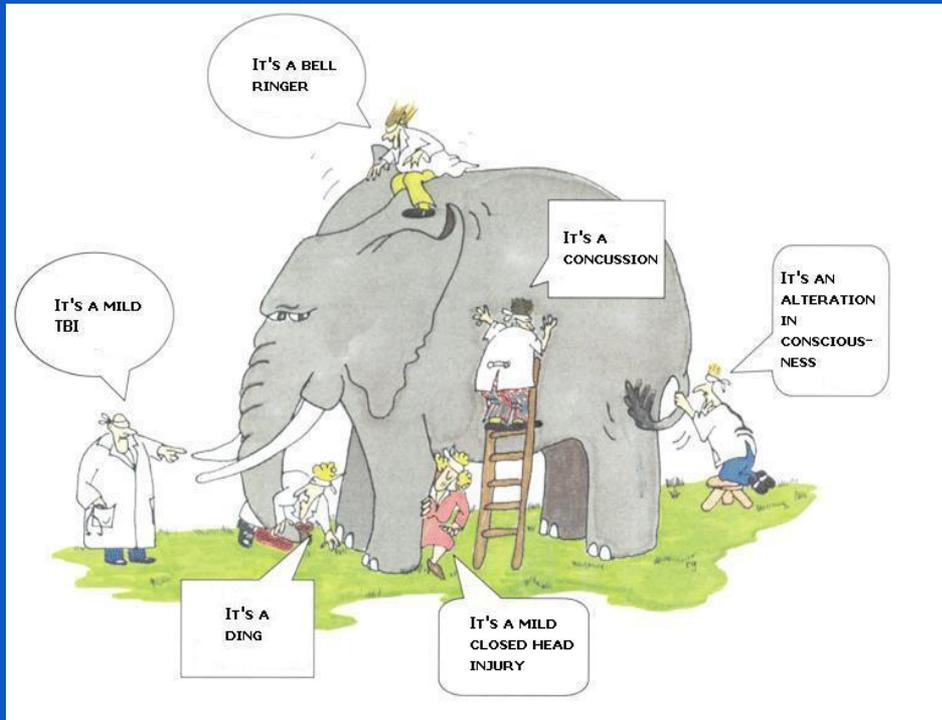
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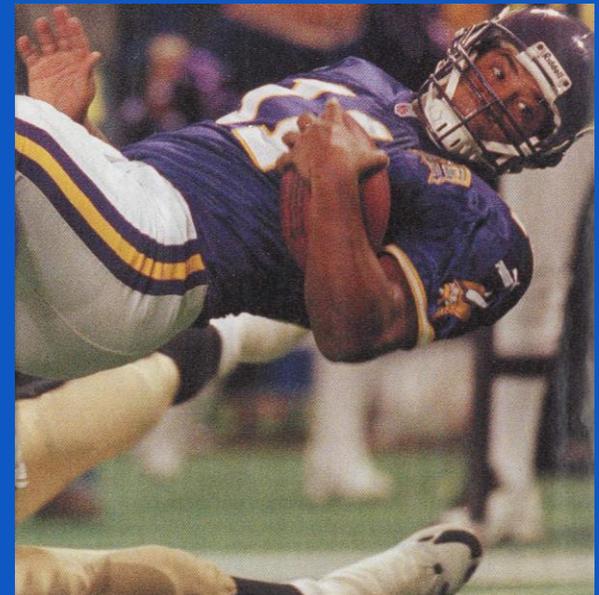
What is a Concussion?



- Japanese Proverb:
- A problem clearly identified is half the solution

Closed Head Trauma without Structural Defects: Concussions

- Definition: “A trauma-induced alteration in mental status that may or may not be accompanied by loss of consciousness”
- **No structural defects** present on neuro-imaging
- Defects may be present on psychological exam

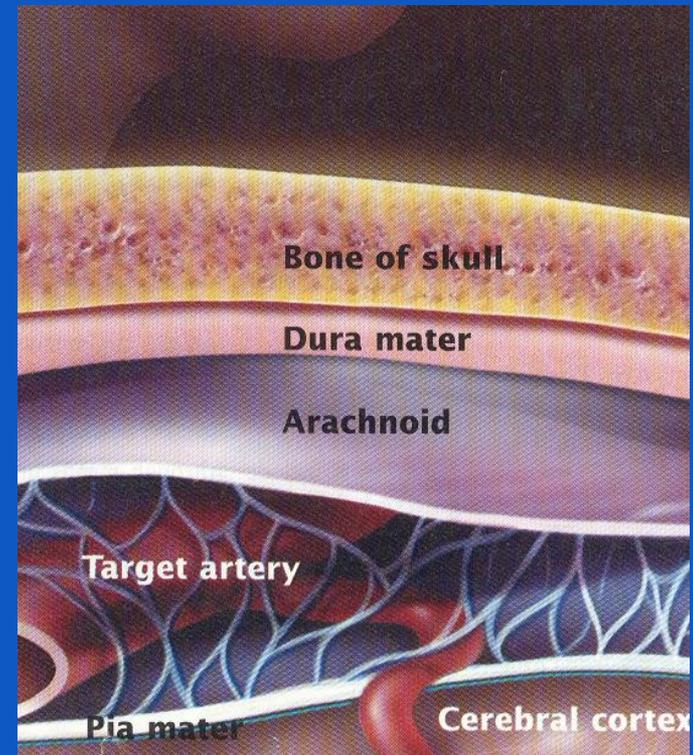


Why this matters...

- House Bill 204 “Protection of Athletes with Head Injuries Act”
 - Requires removal of a child from a sporting event when the child is suspected of sustaining a concussion or head injury
 - Prohibits a child from returning to play until they have received medical clearance from a “qualified healthcare provider trained in the evaluation and management of concussion”
-

Closed Head Injury with Structural Defect

- Structural defects are lesions that can be seen on neuro-imaging
- Structural defects are synonymous with brain damage
- Structural defects = no return to play
- Not “concussions”



Mechanisms of injury

- **Coup injuries:** a direct blow to a stationary skull that compresses/injures underlying brain at the point of impact
- **Contracoup injuries:** injury to brain tissue on the opposite side of a blow or force as a result of a moving brain rebounding against the skull or from tearing of fragile subarachnoid vessels
- ****Rotational injuries:** injury to brain tissue as a result of shearing forces between the skull and brain tissue

Teenage Concussions in the News

The New York Times

Sports

High School Football Player Dies

By [MICHAEL S. SCHMIDT](#) and [DAVE CALDWELL](#)

Published: October 16, 2009

A Montclair High School football player who died Wednesday told a teammate that he still had postconcussion symptoms after he was cleared by doctors to return to play, the school's interim principal said Thursday.

Ryne Dougherty, a junior linebacker, died Wednesday night. He was 16. He had a brain hemorrhage Monday while making a tackle during a junior varsity game. Doctors cleared Dougherty to return Oct. 6, roughly three weeks after he sustained a concussion in a practice Sept. 18. He first returned to action Friday, participating in one play for the varsity team....

....Dougherty is **the third teenager to die in New Jersey in the last three months** after participating in football activities; he is the **second to die because of a brain injury**. Dougherty is also at least the **fourth high school player in the United States to die this year because of a head injury**....



Public Misconceptions about MTBI

Guilmette & Paglia 2004

60% agree....How quickly a person recovers from a head injury depends primarily on how hard they work at recovering.

50% agree....Sometimes a second blow to the head can help a person remember things they had forgotten.

Invisible Injury...The person looks the same with no medical evidence of injury.

Expectation from self and others to 'get over it' and 'get back into the game'.

Scenario:

“Sprained Ankle:



“Sprained Brain”



Overview

– Features:

- 1. Can be caused by direct blow to head, neck, or anywhere on body (forces transmitted to head)
- 2. Typically results in rapid onset of neurologic impairment that resolves spontaneously and quickly, though may be prolonged



Overview



- 3. Acute clinical symptoms *typically* reflect a functional, rather than structural, disturbance (?)
- 4. Resolution typically follows a sequential course
- 5. Typically associated with normal neuroimaging (CT/MRI)

Macro to Micro

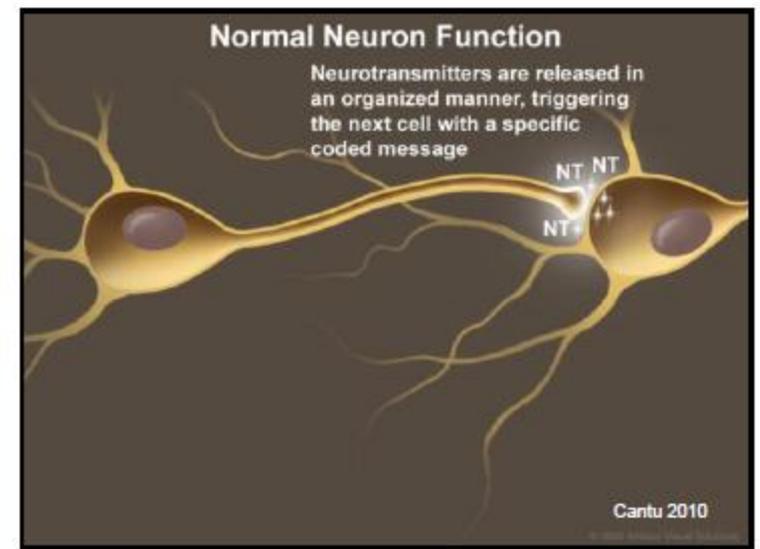
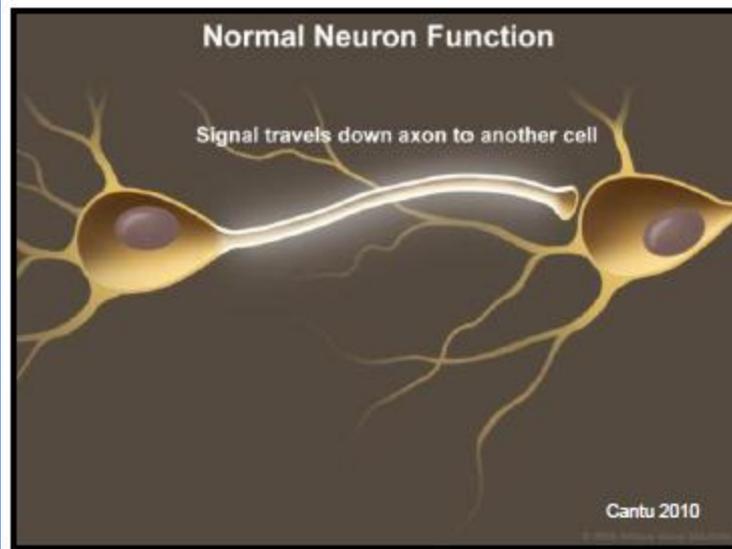
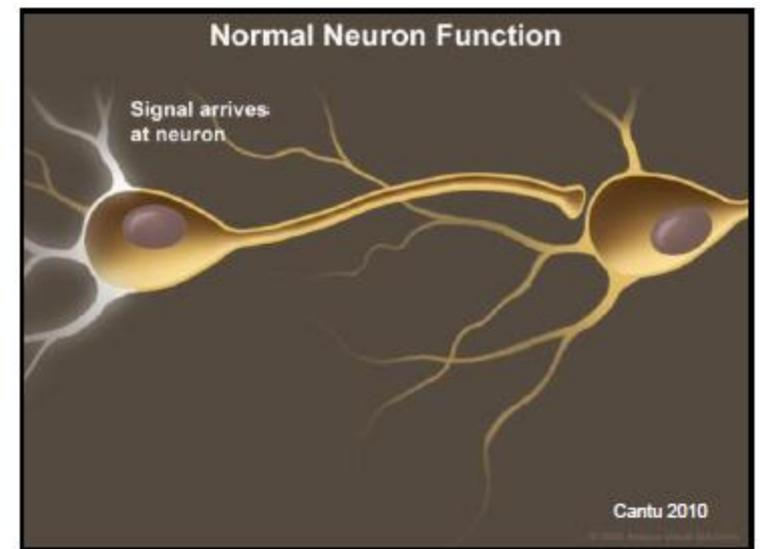
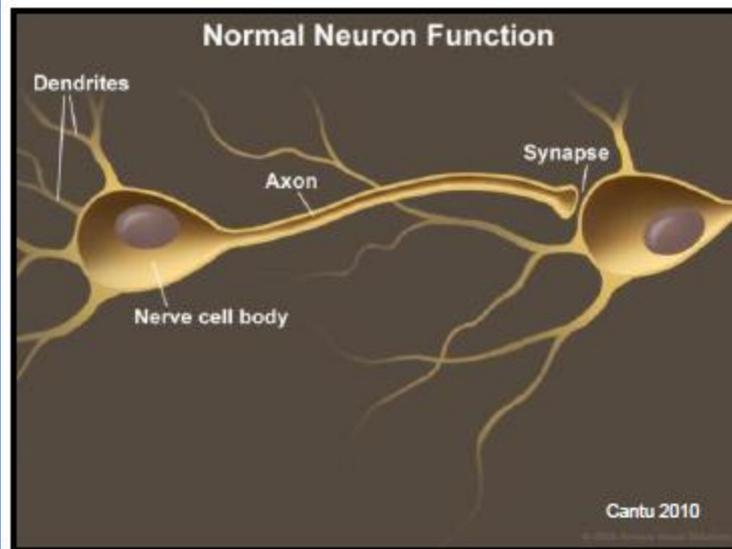
- Cellular Events
 - “A metabolic injury”
 - “Metabolic Mismatch”
 - “An energy crisis in the brain”
 - Axonal Events
 - Spectrum of Diffuse Axonal Injury
-



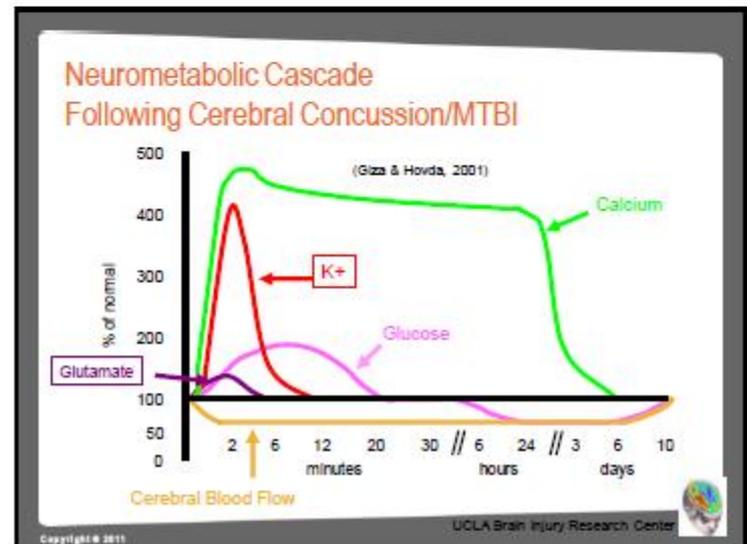
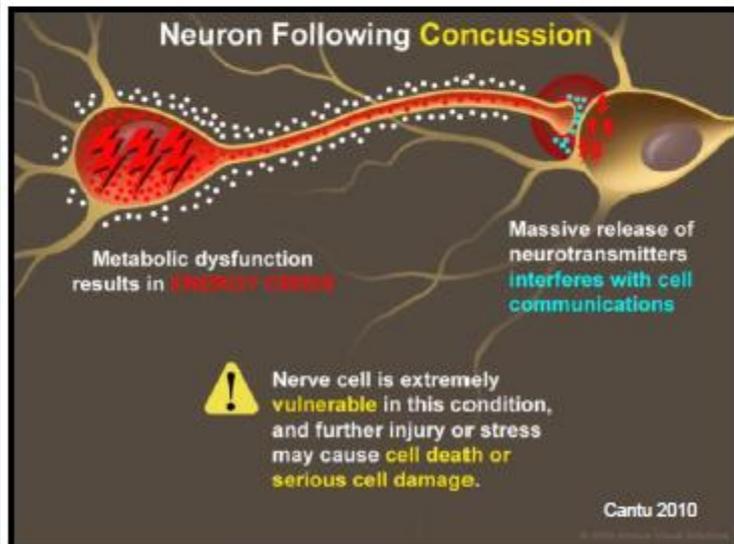
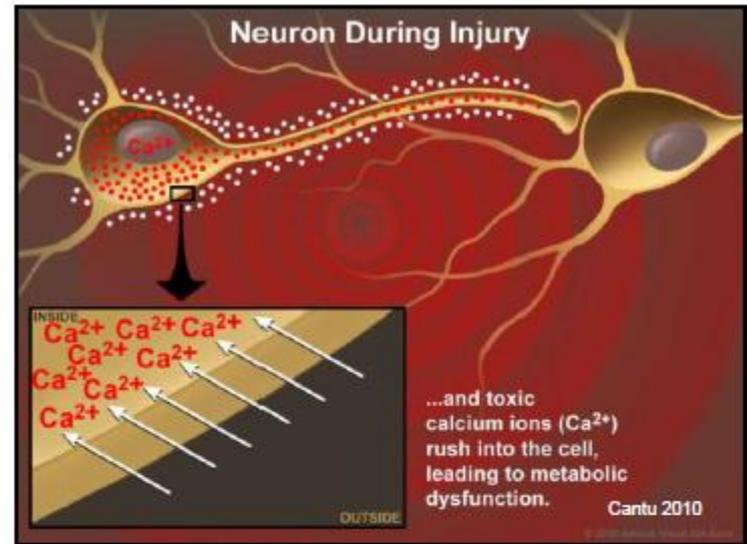
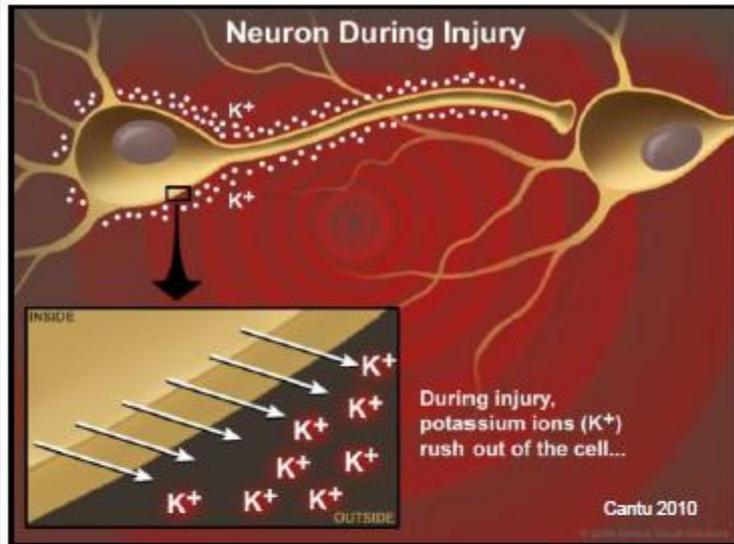
Pathophysiology – Cellular Events

- Axonal stretching may lead to both changes in ionic permeability and injury to the cytoskeleton
- Release of excitatory neurotransmitters and uncontrolled ionic fluxes
- Energy dependent ion pumps activated (Na/K ATPase) in attempt to restore homeostasis
- Glucose demand/utilization increases acutely and then is diminished for up to 2-4 weeks
- Concurrently, local/regional decreases in cerebral blood flow
- Calcium influx and mitochondrial sequestration leads to impairment of oxidative metabolism and subsequent increase in Lactate

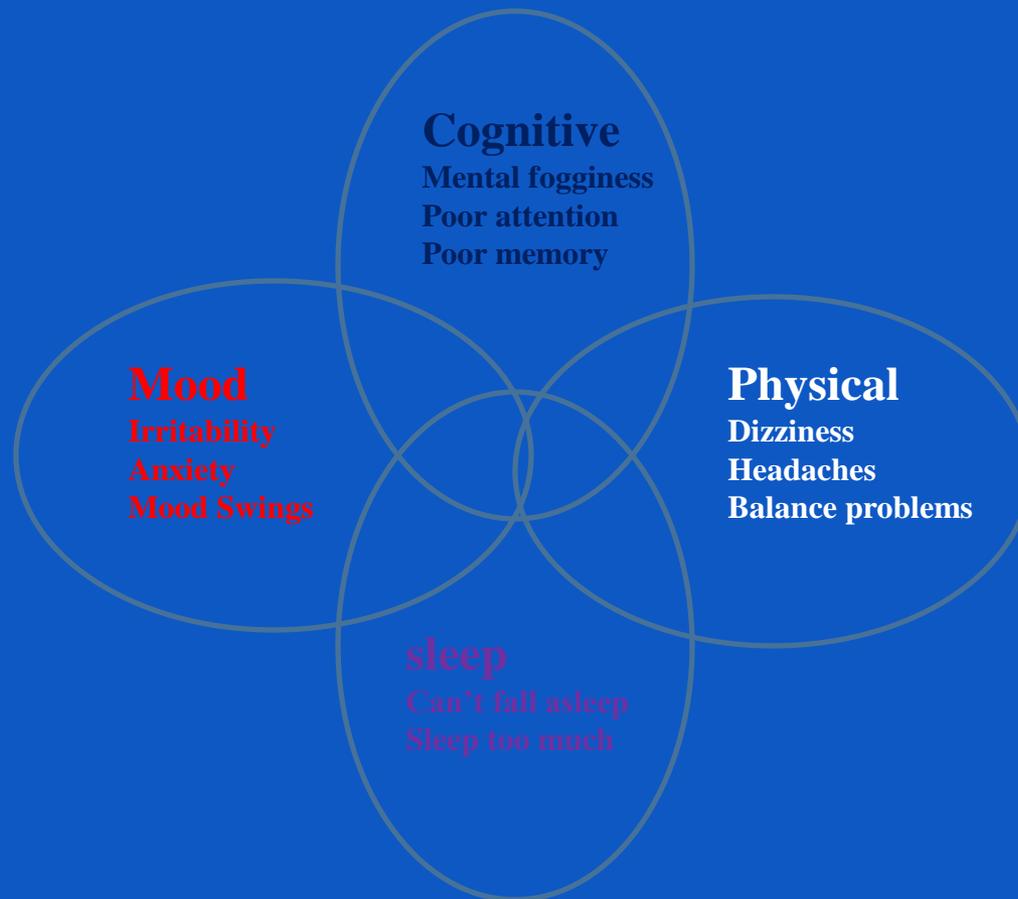
How the Brain's Messaging System Works



Communication Breakdown

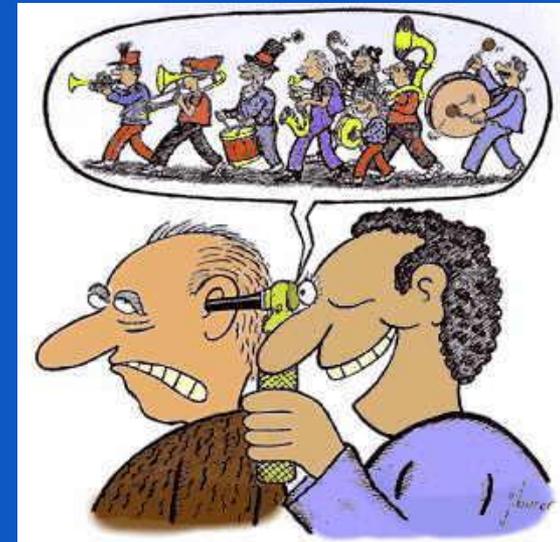


Post Concussion Symptoms



Common Physical Symptoms

- Headache
- Dizziness and lightheadness
- Balance problems
- Blurry vision and light sensitivity
- Tinnitus and noise sensitivity
- Nausea
- Numbness



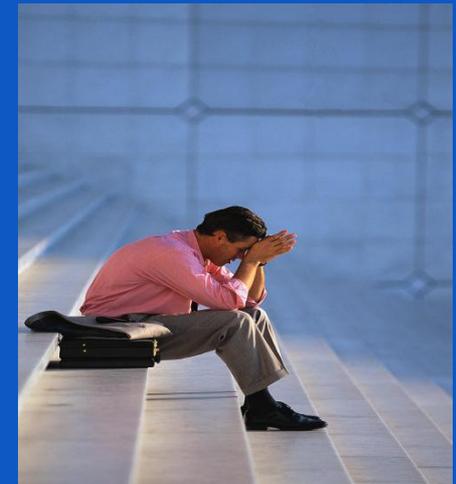
Common Cognitive Symptoms

- Feeling “dazed” or “foggy”
- Easily distracted
- Trouble doing more than one thing at a time
- Poor learning and memory
- Poor reading comprehension
- Poor mental stamina
- Problems with expression



Common Mood Symptoms

- Irritability
- Anxiety
- Mood swings
- Easily overwhelmed
- Emotional and behavioral outburst
- Lack of motivation



Exertional Effects

- Symptoms are worsened by:
 - ❖ Mental effort
 - ❖ Environmental stimulation
 - ❖ Emotional stress
 - ❖ Physical activity



At a football game...

- 16yo HS FB player presents to your office 3 days after sustaining frontal, helmet to helmet blow that produced 10 sec LOC, 10 min period of retrograde amnesia, 30 min period of post-traumatic amnesia. Evaluated at ER; CT neg. Now c/o fatigue, mod to severe bifrontal HA, bilat blurred vision, nausea, fogginess & difficulties with short-term memory.

According to recent published data, which acute or sub-acute symptom is most predictive of experiencing a protracted recovery (less than 3 wks)?

1. Acute Loss of consciousness
2. Presentation of retrograde amnesia
3. Subacute symptom of bilateral blurred vision
4. Subacute symptom of fogginess
5. None of the above, there are no defined prognostic indicators of outcome

Clinical Management Issues

ACUTE MANAGEMENT

- Rule out more serious intracranial pathology
 - CT, MRI, neurologic examination primary diagnostic tests

POST INJURY MANAGEMENT

- Prevent Second Impact Syndrome
- Prevent Cumulative Effects of Injury
- Prevent Post Concussion Syndrome

Determination of asymptomatic status is essential for reducing repetitive and chronic morbidity of injury.

- *Heads UP Booklet/CDC Report, 2007*

How Long is Long Enough?

Preliminary animal and human studies suggest:



-There is **significant benefit from complete rest for several days** to reduce the secondary negative pathophysiological effects on recovery

-Begin a program of **gradual exertion** before starting back to work/school/playing full time

-**Symptom free waiting period**

What Should We Do During Early Recovery

- **Sleep:** It is common for people to need more sleep in recovering from a concussion. 12-15 hours a day is not unusual.
- **Follow a routine:** get up and do a low demand activity during the day. Go to bed at the same time each night; take a nap during the day if needed.
- **Listen to your body:** If you feel tired or your symptoms worsen take a break. If you “push through” your symptom recovery will be prolonged. Reduce demand to match your available energy.

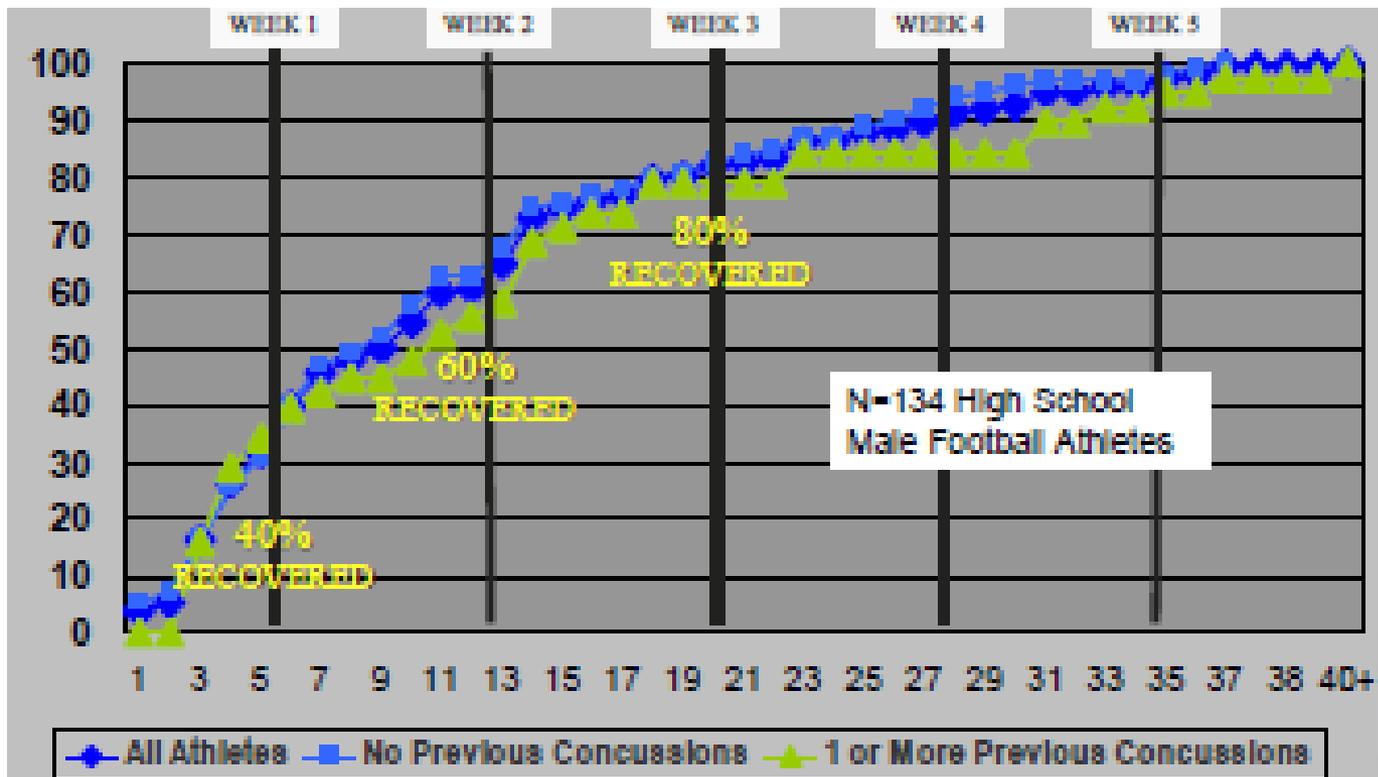
What Should We Avoid During Early Recovery

- **Avoid high risk activities:** Because of reduced reaction time the potential for further injury increases dramatically so, sports, skiing, snow or long boarding, cycling etc. should be avoided until cleared.
- **Avoid high stimulus environments:** Going to a busy shopping mall, noisy restaurant, rock concert, ballgame, party, full day of school and other high stimulus events will slow down recovery and increase symptoms. If you must do something demanding arrange an exit point if you become fatigued.

Real Life Application of Recovery

	Clinical and Physiological Recovery	Patient Experience
Super Acute (~first 5 days)	Symptoms, cognitive dysfunction can be severe, disrupt daily activities. Brain in neurometabolic crisis- Inability to recruit resources	Symptoms, cognitive dysfunction render person unable to perform daily functions, return to work, etc. Exertion may negatively impact recovery
Acute (~6-30 days)	Gradual improvement in symptoms, cognitive functions; full recovery in ~90% of cases. Brain on course back to normal metabolic state. Over recruitment of resources.	Gradual return to full function at work/school/home that requires more effort than customary to meet normal demands; fatigue present.
Chronic (>30 days)	Small %age with persistent symptoms (PCS). Brain returned to normal state; co-morbidities influence recovery	Resume all normal activities without restriction, complications or accommodations.

Individual Recovery From Sports MTBI: How Long Does it Take?



Collins et al., 2008, Neurosurgery

On Field Markers Summary

- Athletes with on-field retrograde amnesia were 10x more likely to have a “poor outcome” at three days post concussion.
- Athletes with on-field anterograde (PTA) amnesia were 4.2x more likely to have a “poor outcome” at three days post concussion.
- Brief LOC is not predictive of outcome.

On Field Symptom Summary

- Brief LOC is not predictive of subacute or protracted outcomes following concussion.
- **Amnesia** is important for acute presentation, but may not be predictive of a protracted recovery.
- Post-injury dizziness and mental foginess is the best predictor of protracted recovery and “post-concussion syndrome.
- Etiology of dizziness?
 - Migraine variant?
 - Central Vestibular Dysfunction?
 - Peripheral Vestibular Dysfunction?
 - Cervicogenic?
 - Psychologic?
- There is a need for better clinical tools to assess dizziness construct

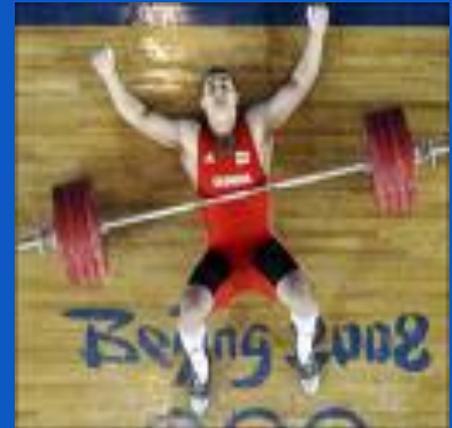
Impact of Multiple Concussions

- 3+ concussions increase the risk of re-injury by 400%
- 3-4 concussions increase the risk of depression/mood disorders by 100%
- 5+ concussion increase the risk of permanent memory problems by 300%
- 75% of repeat concussions happen within 7 days
- 92% of repeat concussions happen within 10 days
- Lesser impact will produce concussion symptoms



Risk Factors for a Complicated Recovery: Exceptions to the Rule?

- History of headaches
- History of multiple concussions
- History of LD/ADD
- Re-injury before symptom free period
- Psychiatric history
- Psychosocial stressors at the time of injury, lower psychological resilience
- Substance abuse
- Litigation/blaming others
- Limited social support



Concussion Management

- Every concussion should be managed individually
 - No two concussions are the same, even in the same patient!!!
- Guidelines, not hard fast rules
- Trend is to more conservative
- Medico legal issues

ALWAYS

- Perform an *exercise challenge* before returning a player to competition
- Arrange for adequate *follow up* before leaving the locker room
- Always *document* your recommendations

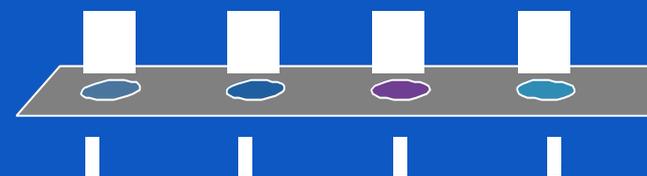
At a gymnastics meet...

- A gymnast fell off the uneven bars during a competition and hit her head on the ground. She had a 2-minute loss of consciousness and had posttraumatic amnesia for 2 hours, which completely resolved. She was sent to the emergency room and had a CT scan of the head, which was negative for any intracranial bleeding. She was discharged to home. She now comes 2 days later to your office complaining of headache and the inability to concentrate in class.

This patient has signs and symptoms of:

1. postconcussive syndrome
2. continuing symptoms of a concussion
3. second-impact syndrome
4. epidural hematoma
5. malingering

Answer Now



10

Countdown
Timer
On Slide

THE END



CONCUSSION

LOOK ON THE BRIGHT SIDE. FOR ONE BRIEF, GLORIOUS
MOMENT, YOU FORGOT YOU WERE ON THE CUBS.

TBI: Pathophysiology and Key Biomarkers

Frederick G. Strathmann, PhD

Brain Injury Statistics

- 1.7 million people (US) require TBI-related medical treatment annually
- 16% of TBI cases require hospitalization
- 3% of TBI cases result in death
- Estimated 25% of TBI cases never seek medical care

Leading Causes of TBI

- Accidental injury
 - Automobile accidents
 - Sports
 - Falls
 - Violence/firearms
- Shaken Baby Syndrome
- Estimated \$50 billion in economic costs (US) annually
- Military
 - Signature injury of Iraq and Afghanistan wars
- 20% of deployed personnel suffer TBI
 - 212,000 (2000 to 2011)
- \$192 million on care in 2010

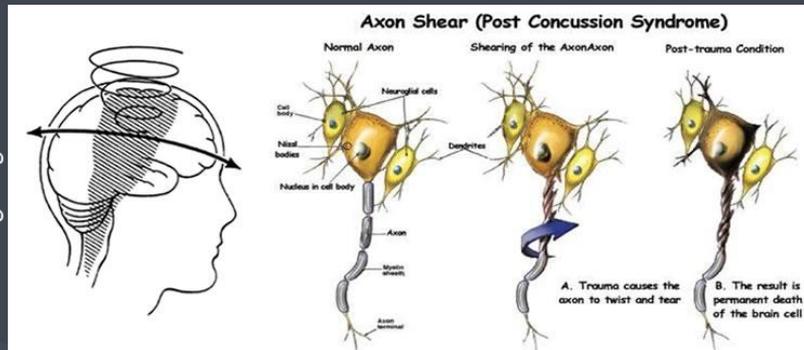
Pathophysiology of TBI

Primary Injury

- Direct tissue damage
- Contusions
- Lacerations
- Shearing damage

Secondary Injury

- Biochemical, cellular and physiological components
- Minutes to years
- Considerable symptom overlap
 - Parkinson's, Alzheimer's, Huntington's, ALS



Long Term Consequences of TBI

- Unprovoked seizures
- Premature death

Penetrating TBI

- Unemployment
- Aggressive Behavior
- Depression
- Alzheimer's disease-type dementia
- Neurodegenerative hallmarks of dementia

Moderate
to
Mild
TBI

Secondary Injury: A Closer Look

Vasculature

- Perfusion ↓
 - hypoxia/ischemia
- BBB breakdown
 - Damage vs. response



sammybutler.blogspot.com

Metabolism

- Oxygen availability ↓
 - Metabolic crisis
 - Neurometabolic cascade
- Brain Temperature ↓↑
- Toxic accumulation

TBI Injury: Key Characteristics for Current Biomarkers

- Blood-brain barrier disruption
- Vasculature disruption
- Intermixing of peripheral blood with CNS
- Diffuse axonal injury (DAI)

TBI Biomarker “Wish List”

Characteristics

- Sensitive
- Specific
- Present in blood
- Appropriate detection window

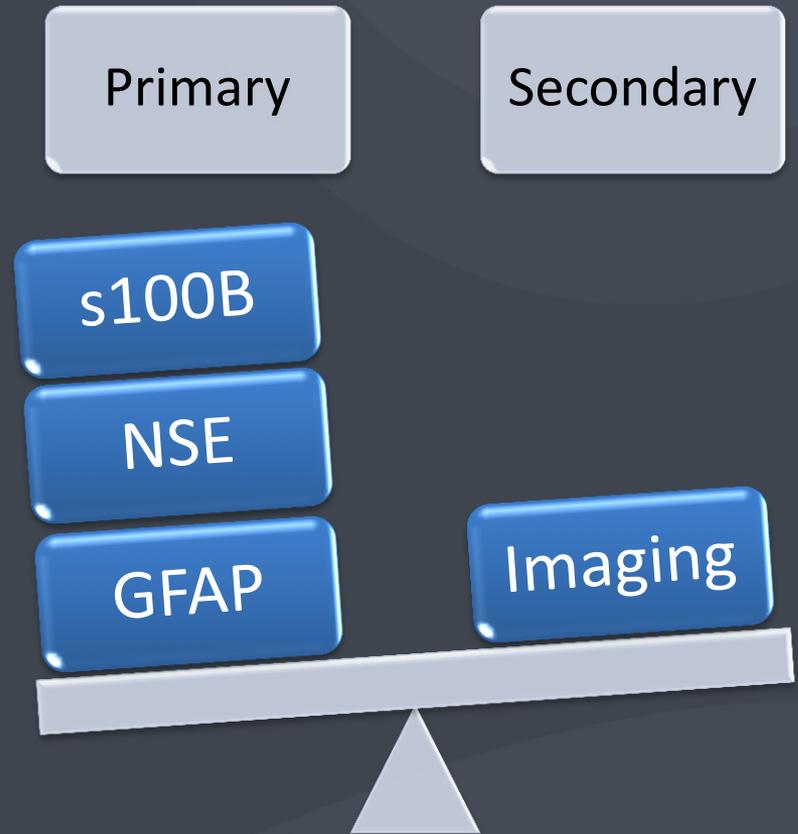
Comparison

- Liver
 - AST/ALT
 - Bilirubin, Coag, TP
- Heart
 - Cardiac Troponins
 - BNP

Prominent Biomarkers of TBI

Common Features

- Venipuncture
- CSF
- Markers of Necrosis
- BBB disruption necessary (?)



Cell Types in the CNS

Neurons

- Transmission of electrical and chemical signals
 - GABAergic
 - Dopaminergic
 - Etc.

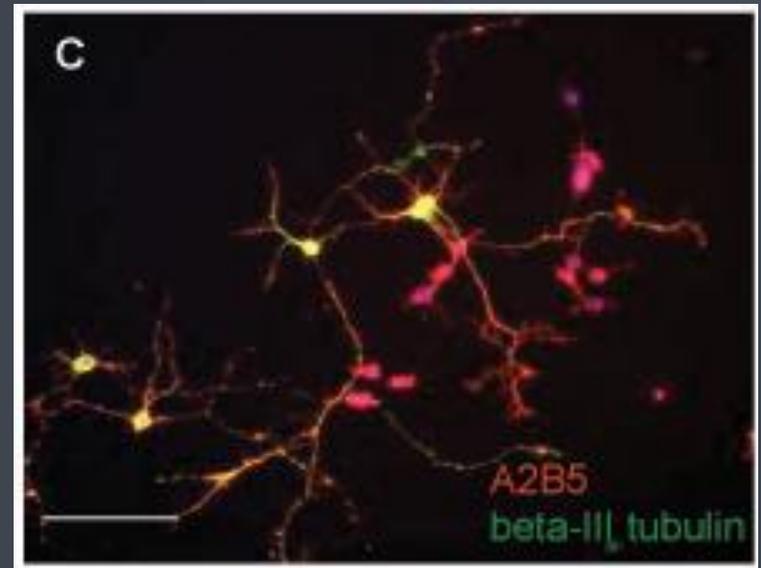
Glia

- Microglia
 - MØ of the CNS
- Oligodendrocytes
 - Myelination
- Astrocytes
 - Everything else

TBI Biomarkers by Cell Type

Neurons

- Neuron specific enolase
 - Glycolysis
- Tau (p)
 - Filament
- Neurofilament light protein

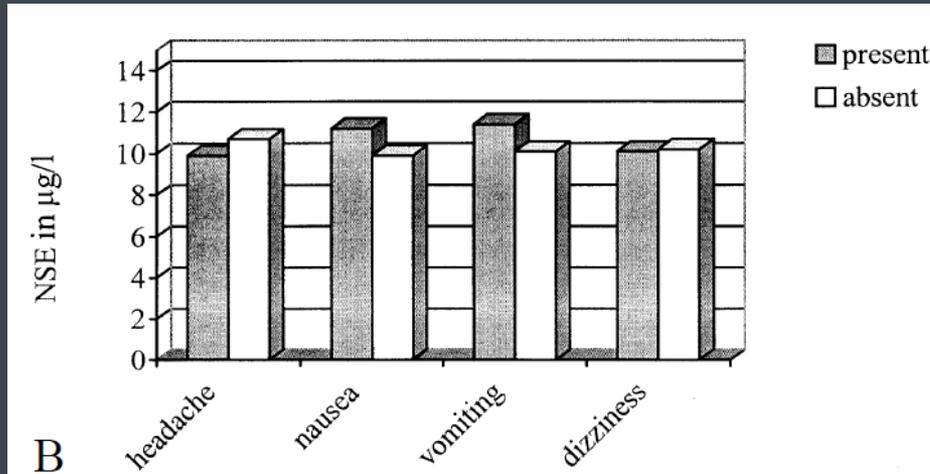


Neuronal Markers of TBI: NSE

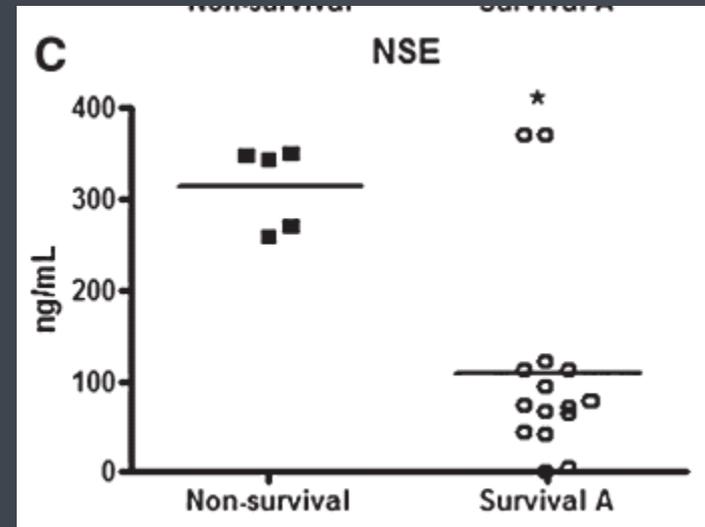
- Dimeric enolase
 - Glycolysis
- Present within hours
- $t_{1/2}$ of 20 – 40 hours
- Released with cell necrosis
- Measured by Immunoassay
- Increases detection window
- Reports of low sensitivity & specificity

NSE Utility in TBI

Mild



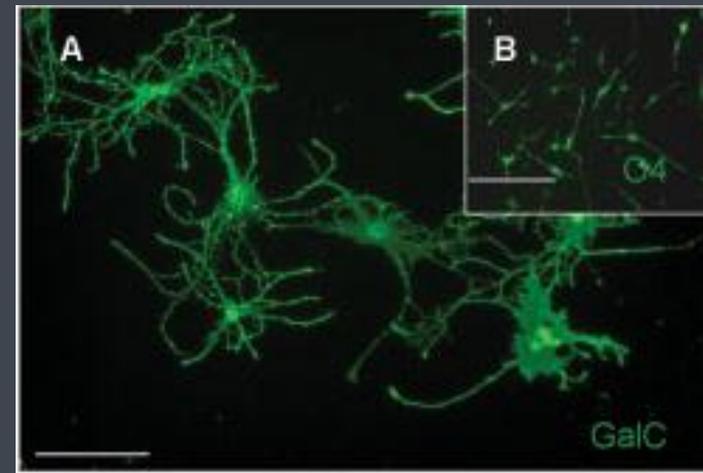
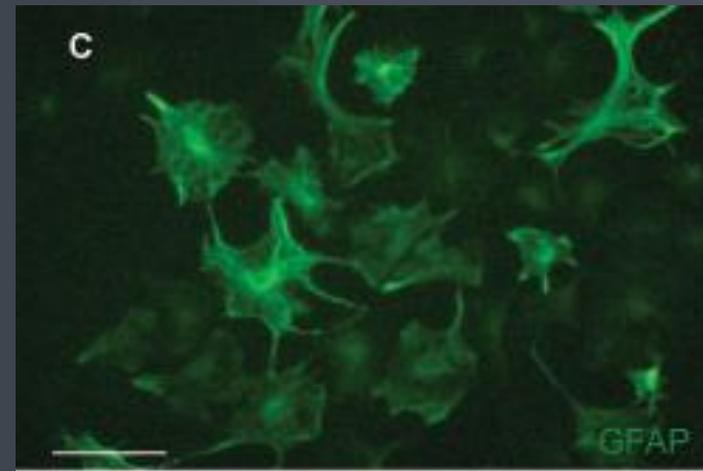
Moderate - Severe



TBI Biomarkers by Cell Type

Macroglial

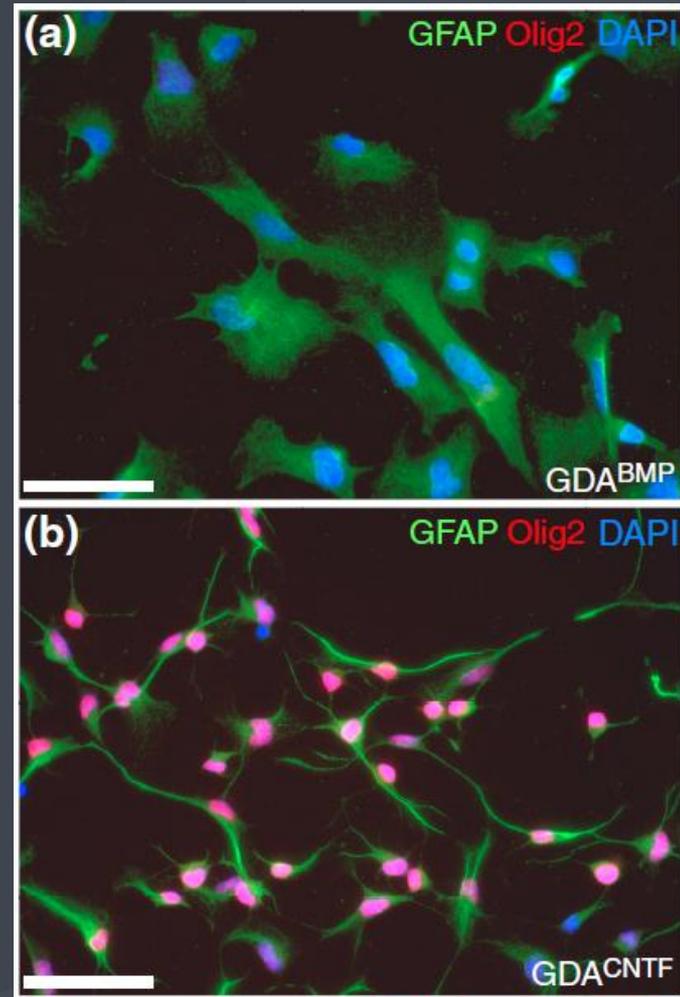
- Astrocyte
 - S100B
 - GFAP
- Oligodendrocyte
 - Myelin Basic Protein



Astrocyte Populations

Phenotype

- Distribution in the brain
 - White matter astrocytes
 - Gray matter astrocytes
 - Others
- Developmental differences
- Lineage differences

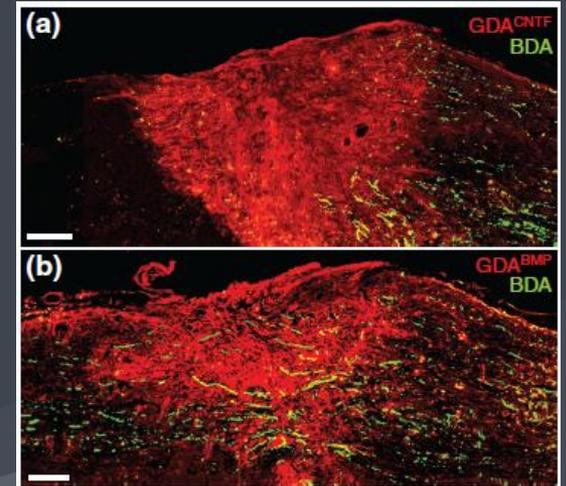


Early Events in Astrocyte Activation

- Transient
- Occurs distal to primary injury site where minimal tissue disruption is found
- Increase in size
- Nuclear hypertrophy
- Morphology changes to a stellate shape
- Increase production of:
 - Cytosolic enzymes
 - AO
 - Structural proteins
 - Organelles
 - Tropic and growth factors
 - Enhance adjacent cell survival

Late Events in Astrocyte Activation: Reactive Gliosis

- Cell hypertrophy
 - *IF upregulation*
 - GFAP & S100B
 - Extension, interdigitation
 - Glial scar
 - Inhibit neurite outgrowth
- “Reactive Astrocytes”
 - TNF-alpha
 - AA
 - NO
 - ROS



Astrocytic Markers of TBI: S100B

- Intracellular Ca binding
- Detected within 6 hours post-injury
- $t_{1/2} \approx 2$ hours
- Measured by immunoassay
- Concerns
 - Present in many non-CNS cell types
 - Widely used biomarker for malignant melanoma
 - Specificity diminished with multiple traumas

The Role of S100B in Injury

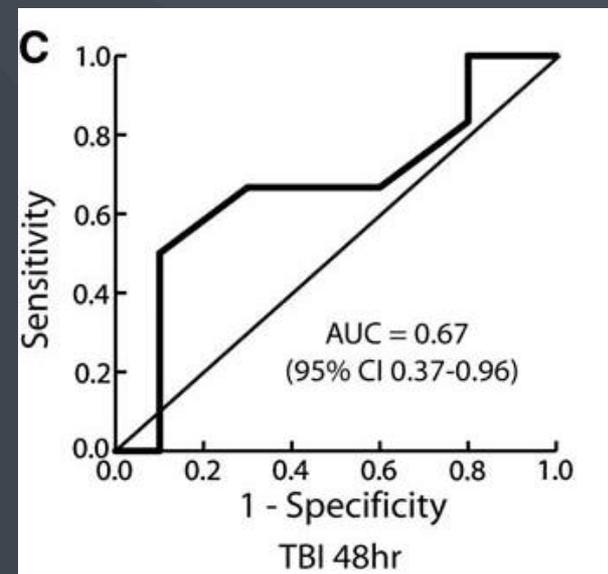
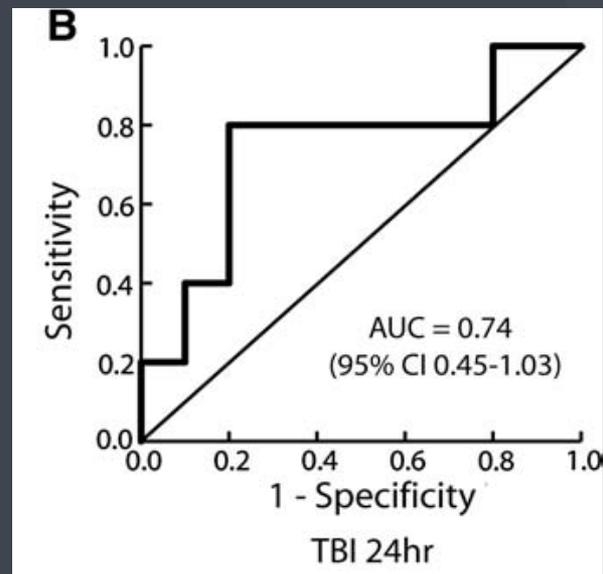
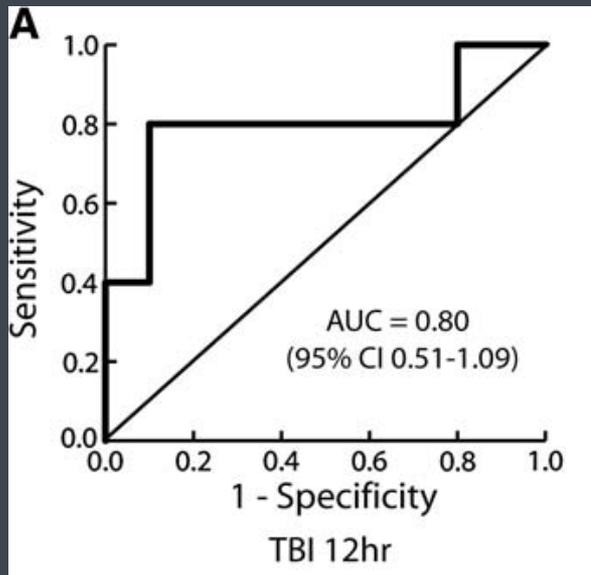
Low Concentration

- Promote neurite extension
- Promote neuron survival
- Regulate muscle development and regeneration

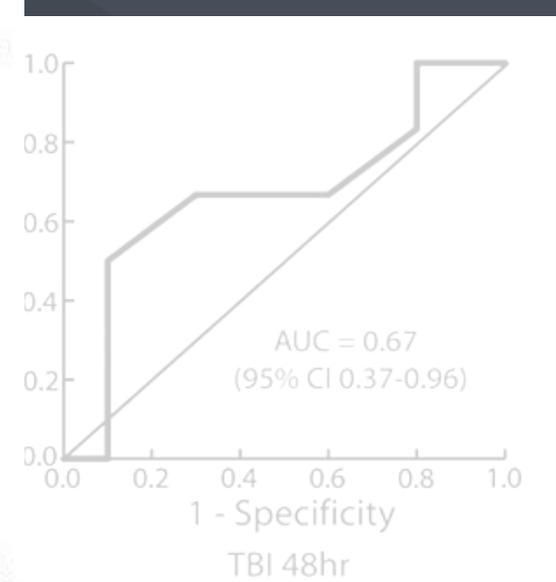
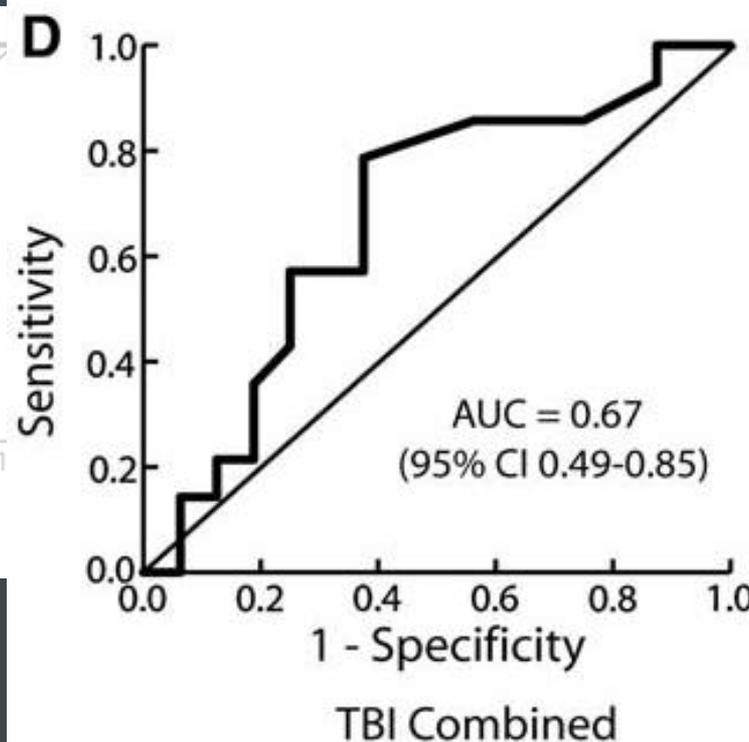
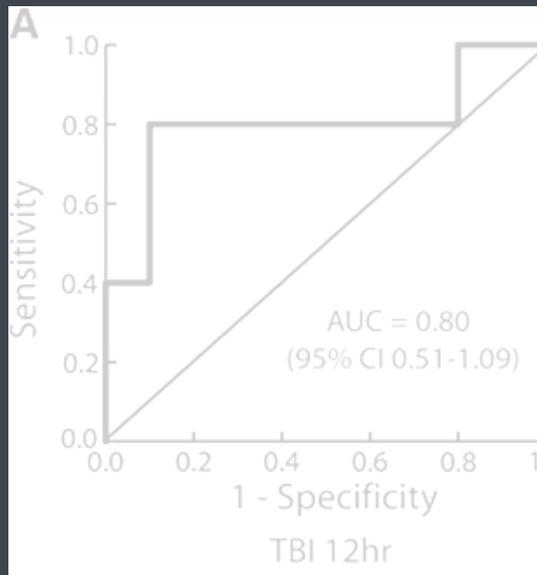
High Concentration

- Upregulates NOS, COX-2
- NO release and cell death
- Increase ROS production
- Perturbs Lipid homeostasis and cell cycle arrest

S100B Performance: Time Course



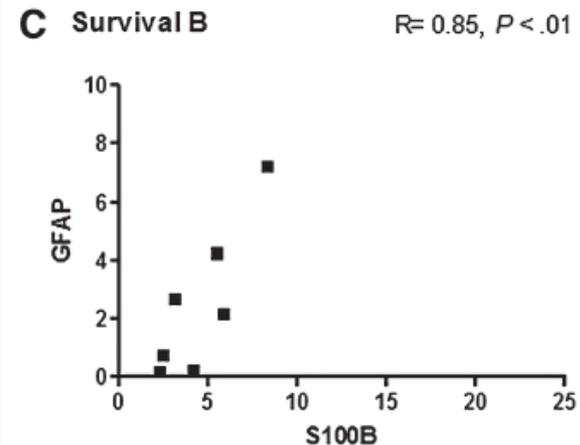
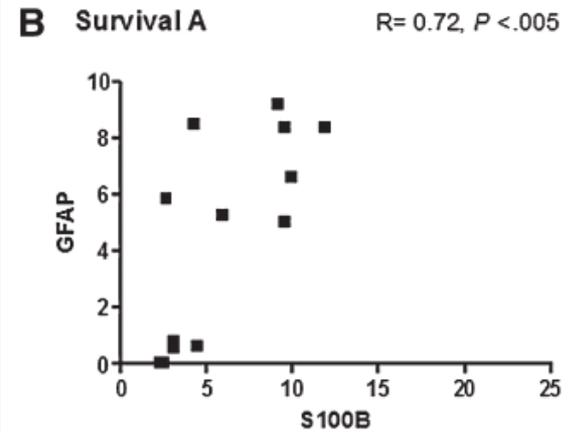
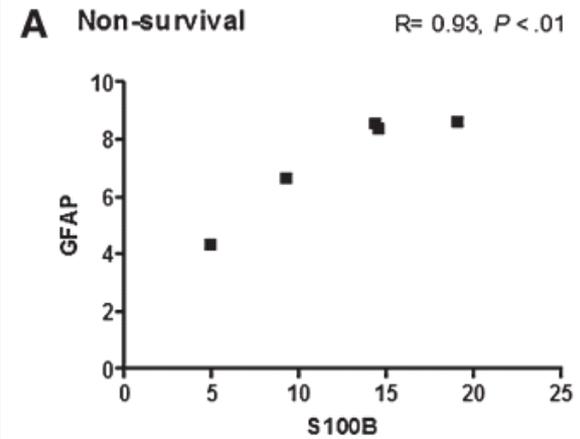
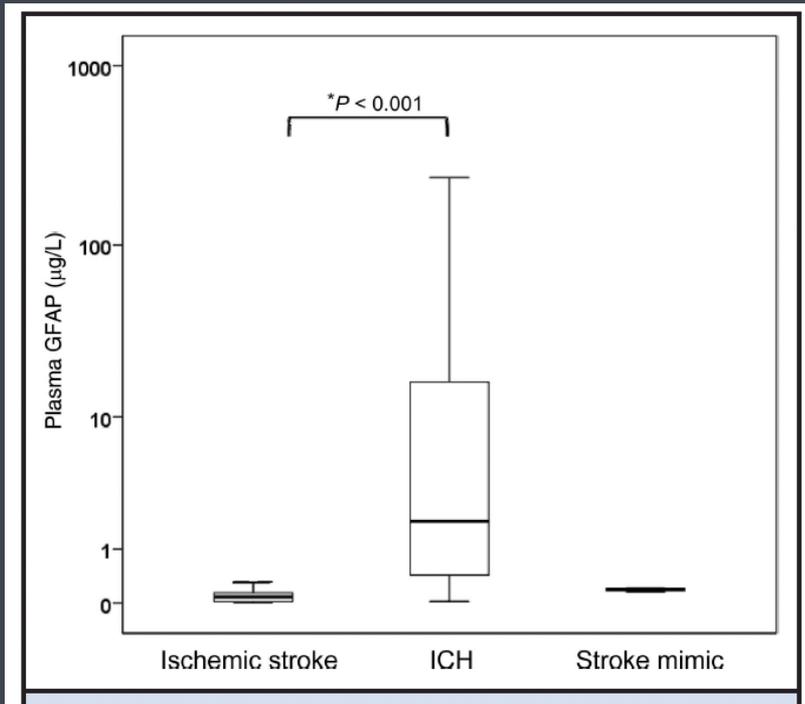
S100B Performance: Time Course



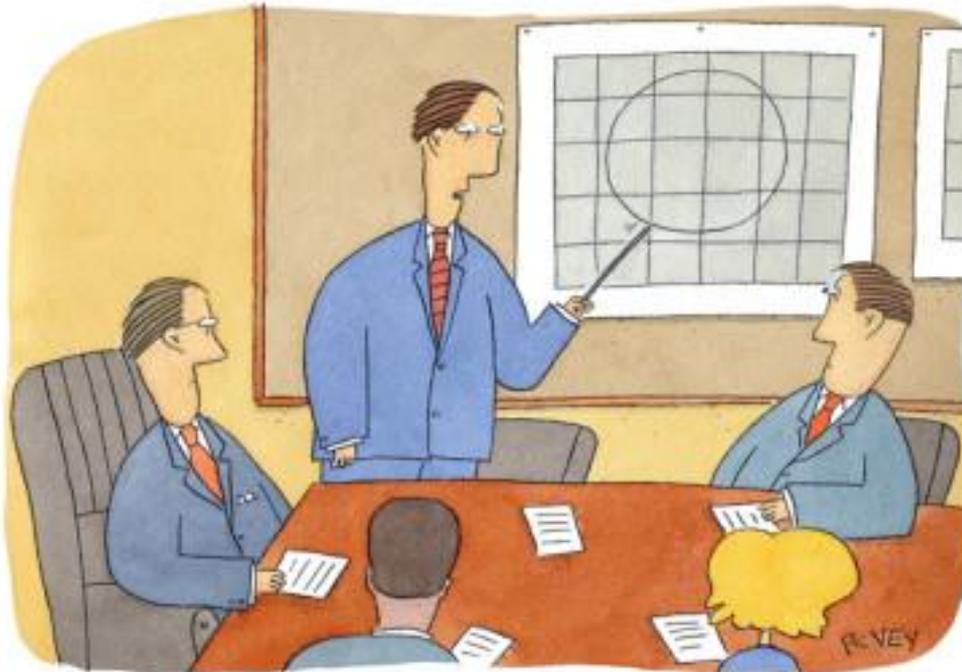
Astrocytic Markers of TBI: GFAP

- Intermediate filament
- Time of detection thought to indicate type and severity of injury
- $t_{1/2}$ not well defined
- Released after cell necrosis
- Measured by WB or immunoassay (clinical?)
- Exclusive to CNS
- Isoforms exist
 - Consequence?

GFAP Performance



Do TBI Biomarkers Help?



"...and it's at this point when we began to be unsure where it all began and where it's all going to end."

What else can biomarkers do?

- S100B
 - Window of opportunity for drug delivery to the brain
- Combinations
 - Determine injured cell types
 - Possibly indicate severity
- Tie-breaker for clinically ambiguous cases

TBI Injury with the University of Utah Football Team

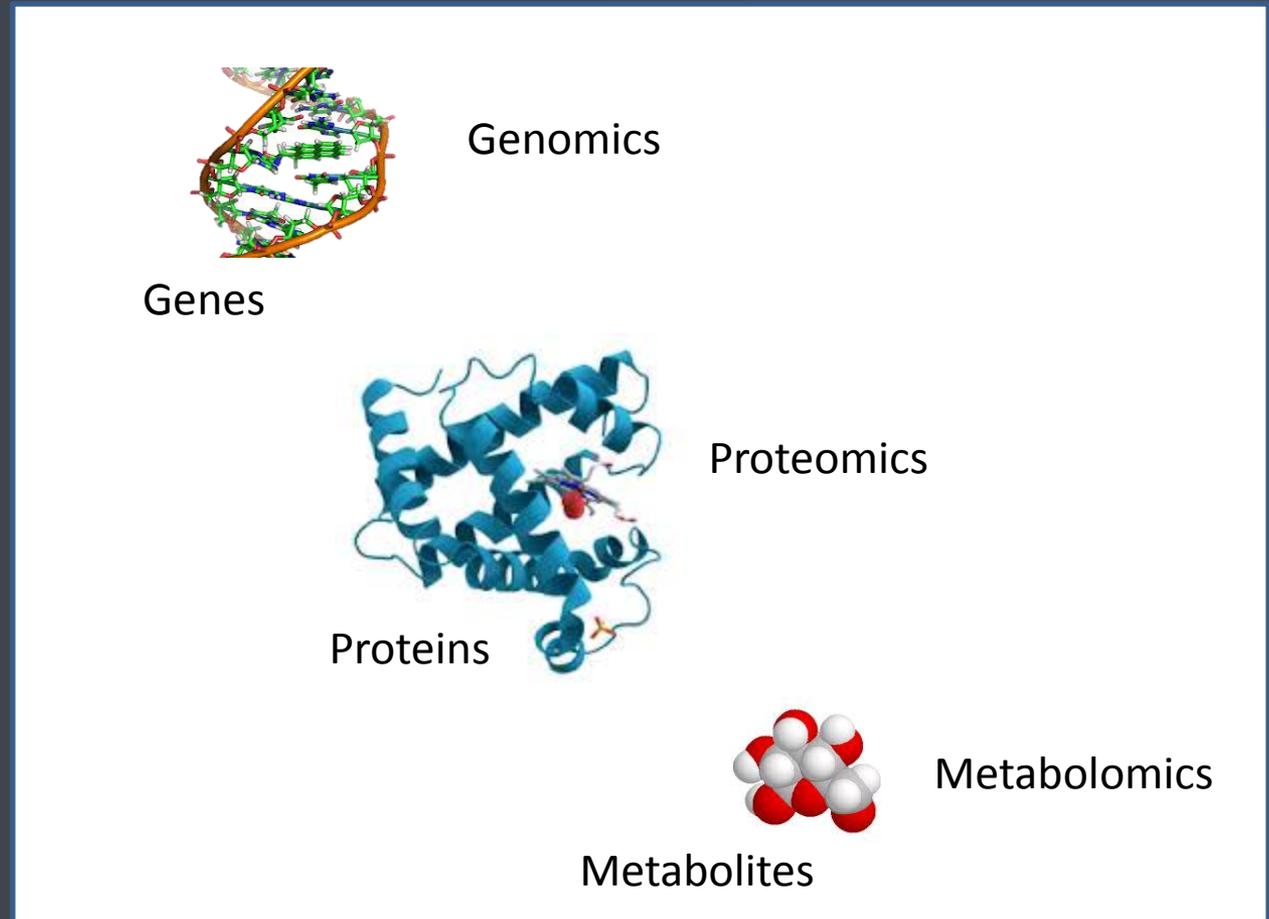


- Can the use of S100B and NSE add to the diagnostic accuracy or efficacy in sports related TBI?
- Dr. David Petron
- Dr. Bradley Hayes
- Stefanie Schulte
- UofU Sports Medicine

New Trends in TBI-related Biomarker Research

- ApoE
- UCH-L1
- miRNA

- POC devices
- Imaging



Biomarker Discoveries in TBI

Classification	Examples
Neuron Injury	NSE, tau, APP
Glial cell injury	s100B, GFAP, MBP
Cell death or Degeneration	NR2A/2B autoantibodies
Metabolic Alterations	Lactate, pyruvate, etc.
Physiological Monitoring	CBF, Brain temperature
Genomic, Proteomic	ApoE, miRNAs

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