Keystone: Traumatic Brain Injury—Epidemiology and Characteristics

Alzforum thanks Sam Gandy, Soong Ho Kim, and Effie Mitsis at Mount Sinai School of Medicine for preparing this meeting summary, edited by Tom Fagan.

Over the last decade, scientists have realized that brain injury, including single traumatic events and multiple mild concussions, can precipitate similar pathologies to those found in people with neurodegenerative conditions such as Alzheimer's, frontotemporal dementia, and amyotrophic lateral sclerosis. This has led to intense study into the nexus between brain injury and neurodegeneration. At Clinical and Molecular Biology of Acute and Chronic Traumatic Encephalopathies, a Keystone symposium held 26 February-2 March 2012, scientists gathered to discuss the latest research advances in this field. Alzforum thanks meeting co-organizer Samuel Gandy, Mount Sinai Medical Center, New York, and colleagues Soong Ho Kim and Effie Mitsis for preparing brief summaries of the meeting. We also thank the many speakers for their input and for providing their slides for our readers to peruse.

The meeting started with **Steven DeKosky**, University of Virginia, Charlottesville. DeKosky reviewed the current understanding of the causes, and the short- and long-term consequences, of traumatic brain injury (TBI). Long-term outcomes in TBI differ depending on age at TBI and whether there was a single or multiple trauma. Pathologies of long-term TBI include chronic neurological and cognitive disorder, chronic traumatic encephalopathy (CTE), and Alzheimer's disease (AD). CTE is characterized by neurofibrillary tangles and TDP-43 inclusions, noted DeKosky. Acute, single-incident, severe injuries (e.g., a car accident) increase risk for AD, whilst less severe repetitive injuries (e.g., boxing) lead to neurofibrillary tangle-based CTE. Long-term effects of TBI are often manifested later in life, and ApoE4 carriers have a 10-fold increased risk for dementia after TBI, said DeKosky.

Introducing a broad definition of TBI that encompasses all causes, **Andrew Maas**, Antwerp University Hospital, Belgium, suggested it would help reporting, comparing, and interpreting worldwide TBI studies. He suggested that epidemiology of TBI is changing in Europe; the overall incidence fell due to fewer traffic accidents, but more falls and contusions occur in older age groups. The consequences of this shift in demographics include different pathologies and comorbidities, and concern over the use of anticoagulants, which aggravate TBI. However, no change in clinical outcome was observed, despite the change in the epidemiology. Standardized epidemiologic monitoring should be considered essential to achieve targeted prevention goals and efficient trauma management, suggested Maas (see slides).

Kevin Guskiewicz, University of North Carolina, Chapel Hill, highlighted the diversity of sports-related concussions, which requires individual treatment strategies. Athletes with prior concussions have increased susceptibility to further concussions, which are associated with chronic symptoms. After a concussion, it is critical to carry out a serial assessment of acute symptoms (somatic, neurobehavioral, and cognitive) before they resolve, usually within about seven days, said Guskiewicz. Longitudinal population-based studies are needed to

understand long-term effects of repetitive mild TBI (mTBI) and sub-concussive events, he said. One such study demonstrated that retired football players exhibit atrophy of white and grey matter, and suffer from memory problems, mild cognitive impairment (MCI), and clinical depression.

In that vein, **Victoria Johnson** found that chronic inflammation persists in the corpus callosum (CC) for years following a single TBI in humans. Johnson, from the University of Pennsylvania, Philadelphia, noted that on postmortem, CR3/43- and CD68-positive amoeboid microglia were found in the CC of more than one-third of cases with moderate (two weeks to one year) and long-term (one to 47 years) survival, but none in healthy controls or the acute TBI group. The thickness of the CC was dramatically reduced in those surviving TBI over one year.

Jenna Ziebell, from the Spinal Cord & Brain Injury Research Center, Lexington, Kentucky, reported that rod-like microglia were observed in the somatosensory barrel cortex of rats after midline fluid percussion injury (FPI). Iba1-positive rod microglia were elongated, and they aligned themselves with neuronal dendrites and axons. These changes were evident from day 1 to 28 post-injury; however, they were most prominent at day 7. Rod microglia were labeled with Iba1 as well as CD68 and OX6, but not CD11b or CD11C.

Ramesh Nayak outlined the possibility of using CNS antigens in re-circulating phagocytes as a source of biomarkers for TBI at different disease stages. Nayak, from the diagnostics company MSDx in Tucson, Arizona, compared two biomarkers, tau and hippocalcin 1, between relapsing remitting multiple sclerosis (RRMS) patients and healthy controls. After using appropriate cutoff values, he detected both tau and hippocalcin 1 in one of 12 control lysates and seven of 18 RRMS lysates. Comparing the two groups for either biomarker did not yield a statistically significant difference, but a comparison between controls and RRMS patients who were positive for either tau or hippocalcin 1 (one out of 12, nine out of 18, respectively) provided a statistically significant difference (see slides).

On the therapeutic front, **Sarah Hellewell**, Monash University, Melbourne, Australia, tested if erythropoietin (EPO) aids in a diffuse traumatic axonal injury (TAI) and hypoxia rat model. EPO administration one hour and 24 hours post-injury decreased the number of axonal bulbs in the CC, rescued MAP2 loss in the cortex and caudate nucleus, and reduced the number of CD68-positive macrophages/microglia in the brain. In addition, hypoxia rapidly upregulated EPO receptor expression.

Sports-Related Injury and Chronic Traumatic Encephalopathy

Increasingly, sports-related head injuries, which can lead to chronic traumatic encephalopathy, are seen as a major public health concern. As was evident at Clinical and Molecular Biology of Acute and Chronic Traumatic Encephalopathies, a Keystone symposium held 26 February-2 March 2012, scientists are beginning to get a better understanding of the forces involved in brain injuries and what kind of damage they create at the molecular, cellular, and organ level. As repeated blows to the head lead to physiological changes in the brain, questions to be addressed include, How do sub-concussive blows contribute to impairment, and how may that be detected? Several theories surround

concussion biomechanics, such as rotational and pressure gradients (whiplash, head motion), and the frontal lobe is a critical region of interest in both biomechanics and CTE. In terms of football collisions, typical blows generating 50-100 times acceleration due to gravity result in delivery of 100-200 pounds of force to the helmet, head, and neck. If the player is braced for the hit, that force is higher. For example, the helmet-first collision documented in three NFL players (Everett, Stingley, and LeGrand) resulted in more than 5,000 pounds of force on the neck.

Thomas Talavage and his team at Purdue University, West Lafayette, Indiana, initiated a study in high school sports players using cognitive tests (impact), structural and functional MRI, MRS in the pre- and post-season, and adding head collision monitoring (HITS, X2Impact) in season. Using these techniques, the researchers found that temporal and spatial distributions of hits link to distinct functional outcomes (see slides). Head contacts typically seen in sports affect neurophysiology as indicated by fMRI and MRS (the metabolic changes are sport and gender dependent), and metabolic changes in asymptomatic football players are consistent with TBI. Therefore, Talavage and colleagues conclude that concussion represents a physical injury with functional consequences.

Jeffrey Barth, University of Virginia, Charlottesville, focused on mild TBI cases entering the TBI study at UVA in the 1980s (55 percent of a total of 1,248 cases). It was found early on in this study that nearly 34 percent of people with mild TBI had not returned to work more than three months post-injury. Studies by Barth of mild head injury at the UVA in the early 1980s, aided by The Wall Street Journal article that described mild TBI as the silent epidemic, gave impetus to the early characterization of diffuse axonal injury and the neurochemical mechanisms involved in mild TBI. The Sports as a Laboratory Assessment Model (SLAM), used to study of football players at UVA, was among the first that focused on sports concussion as a vehicle for clinical research (to be applied to the general population) and for sports medicine. Barth (see slides) showed differences in cognitive functions, such as attention and complex problem solving pre- and post-concussion, and an inability to benefit from the practice effect on neurocognitive testing. The natural recovery curve from these sports concussions was between three and 10 days. Acceleration-deceleration mTBI/concussion sideline assessments (Standardized Assessment of Concussion [SAC] for civilians; Military Acute Concussion Evaluation [MACE] for military) and brief computerized neuropsychological assessments evolved from this work.

Based upon these studies, several critical issues in sports-related concussion were identified as important in terms of outcome (see slides): 1) severity of the injury; 2) when is it safe to return to play; and 3) the effects of multiple, timing, and latency of concussions. It is important to strike a balance, when considering return-to-play issues, between what is known scientifically (evidence based) and what we observe and hope to eventually understand, said Barth. Several negative outcomes have been identified as associated with early return-to-play and multiple concussions and sub-concussive blows: 1) Second Impact syndrome; 2) CTE; 3) emotional disturbance (depression); and 4) acute and chronic cognitive deficits. Individual vulnerability is associated with recovery. Barth said there are several important findings in the concussion literature: Recovery from a single sports

concussion usually takes three to 10 days; one sports concussion increases the risk for subsequent concussion; multiple concussions increase the severity and duration of cognitive deficits; and children appear to recover from concussion more slowly than adults. Barth concluded that the controversy and complexities of the concussion issue should be treated as a challenge to scientific and clinical skills.

Ann McKee, Boston University, summarized our current understanding of chronic traumatic encephalopathy (CTE). CTE is a neurodegenerative disorder characterized by tauopathy and TDP-43 proteinopathy, but no Aβ accumulation. Once triggered, CTE progresses slowly over decades and spreads through many brain structures. CTE is often found in the brains of professional sports players, military veterans, and young people who had multiple mTBIs. The symptoms of CTE are often insidious and begin in midlife with personality and behavioral changes in addition to memory impairment, said McKee (see slides). **Theresa Currier Thomas**, University of Kentucky, Lexington, reported that in a rat model of CTE, thrombospondin-1 (TSP1)-mediated synaptogenic events are ongoing at one month post-diffuse brain injury (midline fluid percussion injury: FPI). TSP1 gene expression increased 30-fold between days 5-7 post-injury, while synaptic gene expression (SYN and GAP43) initially decreased, then rebounded, over 28 days. This evidence of post-traumatic circuit reorganization is accompanied by late onset of sensory sensitivity, increase in evoked glutamate release and altered neuron morphology at 28 days post-injury in the thalamus of FPI rats (see slides).

Metabolic and Axonal Dysfunction in Traumatic Brain Injury

There is now convincing evidence that traumatic brain injury can lead to pathologies, including deposition of amyloid-β, that are akin to those seen in certain neurodegenerative disorders. But what are the underlying molecular and cellular events that precipitate such pathology? This was one of the topics discussed at Clinical and Molecular Biology of Acute and Chronic Traumatic Encephalopathies, a Keystone symposium held 26 February-2 March 2012.

Stephen Ahlers, Naval Medical Research Center, Silver Spring, Maryland, discussed blast-induced brain neurotrauma (BINT) in military personnel and in an animal model of acute or repeated exposure to trauma, which simulates the multiple exposures of military personnel deployed to war zones (i.e., Iraq and Afghanistan). Ahlers (see slides) noted overlap between PTSD and neurotrauma, and the relationship of BINT to chronic traumatic encephalopathy (CTE) and Alzheimer's disease. He focused on mild TBI, considered to be the most pervasive form of brain injury in the military, indicating that the greatest challenges currently in TBI research are in identifying whether mild BINT is similar to classic types of concussion, where the borderline between BINT and PTSD lies, and whether BINT is a new problem entirely, since previously the symptoms associated with artillery battles were considered to be PTSD rather than organic. Ahlers discussed the findings of the Breacher Study, which was conducted in instructors who are exposed to blast on multiple occasions, and students, who are not exposed to blast as frequently as are instructors. Breachers are military or civilian personnel who are trained to use explosives to blast through buildings or walls, and are thus exposed to multiple blast and are at risk for BINT. The study found no effects in students, but instructors exhibited signs

of cognitive impairment and showed changes on neuroimaging. The take-home message is that the number of blast exposures over time is important, and that future experiments need to be developed to characterize the impairment from blast. Regarding animal studies, the goal is to elucidate the natural history of repeated exposure to blast overpressure (BOP) on brain function and physiology. Ahlers characterized the BOP threshold for disruption, identified the importance of orientation of the animal to the BOP wave, and reported on pathologic outcomes, including Aβ changes in brain, and the effect of BOP on learning and memory. In a paper in Frontiers in Neurology (see Ahlers et al., 2012), Ahlers and colleagues classify two types of consequences of blast brain injury and associated parameters (blast frequency, intensity, physical forces, clinical manifestations, onset, radiology/pathology, and biomarkers).

Nicholas Tustison, also at UVA, emphasized that TBI is a complex disease process involving mechanical disruption of brain tissue and activation of secondary injury cascades, which culminate in widespread axonal disconnection, neuronal cell death, and loss of function. Diffusion tensor imaging affords quantification of microstructural white matter injury, while high-resolution T1-weighted sequences allow for computation of cortical thickness and density maps that could help characterize TBI, said Tustison (see slides). Although advances in equipment and sequence design have yielded dramatic improvements in image quality in recent years, image analysis is critical to accurately detecting subtle alterations that may reflect clinically significant disease. Beginning with a description of white matter changes in TBI, followed by a discussion of specific metrics used in DTI analysis (fractional anisotropy and mean diffusivity), and the types of DTI analytic possibilities (i.e., manual and automated regions of interest, tractography and tract-based spatial statistics [TBSS], and voxel-based morphometry), Tustison asked, What is the best method for identifying effects of blast TBI in the military? In sum, he reviewed current approaches for the computational analysis of DTI and cortical maps in TBI, and by presenting work conducted in their lab at UVA, discussed how to optimize their use.

Meeting co-organizer **Samuel Gandy**, Mount Sinai Medical Center, New York, discussed the Alzheimer's "nexus," focusing on three main questions:

1. Can our understanding of late-onset AD pathology inform our understanding of post-trauma AD?

2. Can post-traumatic AD be distinguished from AD without a history of TBI?

3. How can our understanding of AD be applied to experimental therapeutics for post-traumatic AD?

Gandy reviewed recent findings from transgenic mouse studies and clinical trials that indicate pre- and post-amyloid vaccination in mice and passive immunotherapy in humans did not lead to cognitive improvement, despite a lowering of amyloid burden (see slides). These findings raise the question of the "right target" in AD, and whether treating a single target will ever be sufficient, said Gandy. He then turned to the role of TBI in neurodegeneration, and potential pathways that may be involved. He emphasized the role of signal transduction in

Aβ regulation, and noted the value of mGluR antagonists as a potential treatment for neurodegenerative dementia. He reported that mGluR signaling may be involved in regulating Aβ42 metabolism at the synapse. He showed that mGluR2/3 antagonists lower levels of various Aβ conformers in the hippocampus and cortex, and that they correct Aβ-induced contextual memory deficits, improve novel object recognition, and decrease anxiety in APP transgenic mice. Gandy revealed that BCI-632, a neurogenic compound, improves outcomes following experimental TBI. He suggested that mGluR2/3 antagonists and pro-neurogenic/pro-autophagic compounds may be useful in preventing or treating late neurodegenerative sequelae of TBI. Gandy then questioned the role of ApoE4, noting that an important issue for clarification is whether ApoE4 exacerbates tau pathology independently of effects on Aβ42. Gandy wondered, for example, if bapineuzumab infusion should be tested during acute post-TBI phase, and if ApoE4 alters the structure and/or conformation of *both* plaques and tangles visualized using luminescent conjugated oligothiophenes.

Christopher Giza, University of California, Los Angeles, described two potential ways to treat developmental TBI—a ketone diet and D-cycloserine. The ketogenic diet reduced lesion volume in immature rats after controlled cortical impact (CCI), a model of TBI that causes cellular energy crisis due to disrupted mitochondrial function. CCI induces upregulation of monocarboxylate transporter 2, a transporter of ketone bodies, which helped the animals to utilize more ketones in the diet. D-cycloserine, a partial agonist for NMDA receptors, restored hippocampal NMDA NR2A subunits in P19 rat pups that were diminished by fluid percussion injury (FPI). FPI reduced NMDAR postsynaptic currents, which led to impaired memory tasks in these animals. This decreased network activation was also observed in children who suffered TBI during a spatial working memory task. D-cycloserine also activated CaMKII levels, and improved spatial and object recognition memory in the P19 pups. Lastly, using a repeat closed head injury model in juvenile P35 rats, Giza showed that a second injury occurring 24 hours after the first (during the period of post-TBI hypometabolism) actually worsened the metabolic crisis, while a second injury sustained five days after the first (at a time of recovery from hypometabolism) did not show additive/worsened metabolic consequences (see slides).

TBI—Learning From Markers, Models, and Diseases

Injury to the brain, even what might be considered mild, can have devastating consequences on brain physiology. But how can scientists probe how the brain responds to injury? At Clinical and Molecular Biology of Acute and Chronic Traumatic Encephalopathies, a Keystone symposium held 26 February-2 March 2012, researchers showed how they can use animal models, and both imaging and fluid markers, to get a better handle on the pathological changes evoked by brain injury.

Andrew Mayer, Mind Research Network, Albuquerque, New Mexico, asked if biomarkers might help map recovery from traumatic brain injury. He showed that the term "mild" to describe some types of TBI may be inadequate (see slides). He noted that there are many diffuse injury mechanisms, but the pathophysiology underlying mild TBI and how these injuries change as a function of time remain unclear. While DTI holds promise for in-vivo characterization of white matter

pathology, said Mayer, the direction and magnitude of anisotropic diffusion abnormalities continue to be debated. Findings in the field are varied, with increased, decreased, or no difference in fractional anisotropy reported. Mayer presented an independent replication (using 28 subjects) of previous findings of increased FA during the semi-acute phase of injury in a cohort of 22 individuals. He has also carried out a prospective study on the putative recovery of diffusion abnormalities among 26 volunteers. He applied novel analytic strategies to capture spatially heterogeneous white matter injuries. Results indicated that a general pattern of high anisotropic diffusion/low radial diffusivity in various white matter tracts in both the replication or original cohorts, but this was only consistently observed in the genu, or anterior end, of the corpus callosum across both samples. Mayer identified a greater number of localized clusters (i.e., lesions) having increased anisotropic diffusion across both cohorts, confirming heterogeneity in white matter injury. Finally, evidence that lesions recover in patients re-examined across a four-month interval correlated with a reduction in self-reported post-concussive symptoms. Mayer concluded that diffusion abnormalities are associated with cytotoxic edema secondary to mechanical damage, resulting in changes in ionic homeostasis, and alterations in the ratio of intracellular and extracellular water.

Kathryn Saatman, University of Kentucky, Lexington, reported acute activation of calpain in the cortex and hippocampus in rodent models of contusion TBI, while axonal calpain activation was seen in diffuse or mild repetitive TBI (see slides). Rats exposed to TBI, then treated with the calpain inhibitor AK295, more quickly recovered motor function and had improved memory, though the inhibitor was not neuroprotective when given prophylactically. Mice overexpressing the human endogenous calpain inhibitor, calpastatin, exhibited reduced proteolysis of cortical α-spectrin and sodium channels after contusion TBI. Motor and cognitive deficits were less severe in the transgenic mice after injury. Similar approaches, using calpain inhibitor treatment or genetic overexpression of calpastatin, have been successful in reducing cognitive impairment, tau phosphorylation, and amyloid-β plaque formation in transgenic mouse models (APP/PS1) of Alzheimer's disease.

David Brody, Washington University School of Medicine, St. Louis, Missouri, showed that interstitial fluid (ISF) amyloid-β (Aβ) levels are dynamic in humans and can change up to eightfold during hours and days after brain injury (see slides). Increased ISF Aβ levels over time after injury correlate with improved global neurological status (as reflected in better Glasgow Coma Score). In contrast, mean ISF tau levels in the initial 12 hours after injury inversely correlate with clinical outcomes at six months. In PDAPP, Tg2576, and wild-type mice, controlled cortical impact TBI decreased PBS-soluble Aβ and acutely reduced ISF Aβ, the latter being correlated with reduced neuronal activity. In comparison, Brody detected increased insoluble Aβ in injured axons of 3xTg-AD and APP/PS1 mice.

Keeping with the Aβ theme, **Douglas Smith**, University of Pennsylvania, Philadelphia, reviewed current knowledge of diffuse axonal injury (DAI) and changes in Aβ metabolism. DAI is caused by stretch injury, which breaks microtubules and partially interrupts axonal transport, leading to axonal swelling

and APP accumulation. Repetitive mild stretch of axons (repetitive mTBI) induced Ca2+ influx and upregulation of Na+ channels, noted Smith. He observed long-term axonal pathology in human tissue up to six months postmortem, where Aβ, APP, tau, and neurofilaments colocalized with PS1 and BACE1. More thioflavin S-positive Aβ plaques were detected in TBI patients postmortem, compared to diffuse plaques in controls. He detected acute Aβ plaque formation in 30 percent of TBI patients, which he attributed to a polymorphism of neprilysin, an Aβ-degrading enzyme.

To map amyloid deposition in TBI patients in vivo, **David Menon**, University of Cambridge, U.K., used PET-PIB imaging to create non-displaceable-binding potential (BPND) maps using a simplified reference tissue model. Compared to controls, diffuse high PIB BPND signals increased in both gray and white matter in TBI brains during eight weeks post-injury. To avoid nonspecific binding and provide detailed assessment of temporal patterns, PIB BPND was measured only in cerebral cortex regions of interest, and results from each patient plotted against time post-TBI. PIB binding in the region rose above the normal range in the second week and normalized after four weeks. Using autoradiography, he also measured titrated PIB in postmortem brains from patients who died between three hours and eight weeks post-TBI. PIB robustly and specifically bound to the cortical tissue. **Rachel Bennett**, also from WashU, created three lines of AD-ApoE mice by crossing 3xTg-AD animals with those carrying knocked-in human ApoE2, ApoE3, or ApoE4 alleles. Bennett gave a single controlled cortical impact (CCI) to six- to eight-month-old AD-ApoE mice and then measured APP, Aβ40, and tau levels. APP and Aβ40 levels increased after the injury. AD-ApoE4 mice exhibited more APP than AD-ApoE2 and AD-ApoE3. In contrast, Aβ40 increased equally in all AD-ApoE mice. No pre-/post-TBI difference in tau level emerged, but AD-ApoE4 mice accumulated more tau than AD-ApoE2 and AD-ApoE3 mice.

Andrei Irimia, University of California, Los Angeles, discussed patient-tailored quantification of brain atrophy in TBI and CTE using multimodal neuroimaging. Multimodal imaging (MMl) can aid in the formulation of therapies to accelerate recovery from brain injury and improve quality of life. Longitudinal quantification of brain atrophy due to CTE and TBI provides significant neurobiological and clinical insight into the progression of these conditions. Irimia and colleagues hypothesized that their MMI framework allows for the detection and characterization of significant gray matter (GM) and white matter (WM) atrophy occurring between acute and chronic stages of TBI/CTE (see slides). The group acquired structural imaging at 1.5 Tesla, both acutely (three days post-injury) and chronically (180 days post-injury), using T1, fast spin echo, gradient echo, long tau inversion recovery, DTI, diffusion-weighted imaging, and susceptibility-weighted imaging techniques. Gray/white matter pathology metrics and segmentations of acute lesions, hemorrhage, and edema were obtained. Cortical parcellation yielded brain morphometrics and volumetrics, and DTI-based white matter fiber tracking was conducted to render structural connectivity matrices. The group identified individual fiber tracts exhibiting statistically significant atrophy, and they correlated volumetric atrophy measures against computed bi-frontal, bi-caudate, ventricular, Evan's, and Huckman's indices. The presence of gray/white matter atrophy was identified in all subjects, and detailed

descriptive metrics computed for each cortical region and fiber tract involved. The atrophy quantification framework was more comprehensive compared to using only traditional atrophy indices because, in addition to relevant metrics, it offered the ability to map atrophy and identify significantly affected white matter fibers. These strategies may help clinicians and healthcare providers design improved methods for patient monitoring and rehabilitation.

Linda Van Eldik, at the University of Kentucky, Lexington, showed that a novel, orally active blood-brain barrier-penetrant compound, MW151, suppressed production of proinflammatory cytokines (IL-1β, TNF-α, S100B) in Aβ-induced brain injury, which helped to maintain synaptic integrity and recover hippocampus-dependent cognitive performance. MW151 treatment also helped in two animal TBI models of diffuse axonal injury (cortical impact and midline fluid percussion) when administered a few hours post-injury to mimic the time gap of injury to trauma center (see slides). Post-TBI, MW151 also prevented the typical increase in seizure susceptibility, glial activation, and cognitive deterioration after seizure induction with a second "hit" by electroconvulsive shock.

Diagnosis and Model Treatments for Traumatic Brain Injury

Identifying injuries that might lead to chronic traumatic encephalopathy or other neurodegenerative pathologies, and then treating those injuries, are two major challenges for those in the traumatic brain injury field. Both were addressed at Clinical and Molecular Biology of Acute and Chronic Traumatic Encephalopathies, a Keystone symposium held 26 February-2 March 2012. **Kaj Blennow**, University of Gothenburg, Sweden, has performed studies on CSF biomarkers to identify and monitor neuronal injury in amateur boxers. He reported that after a bout, CSF levels of neurofilament light (NFL), tau, GFAP, and S100B increased in boxers who received more than 15 punches, reflecting acute axonal and glial damage. This was despite the boxers wearing head protection, not being knocked out, and sparring only a small number of rounds. The CSF NFL reached five times higher after a bout than after over three months' rest, and the severity correlated with the number of blows suffered. Interestingly, Blennow and colleagues detected no change in the levels of these CSF biomarkers in amateur soccer players after 15-30 headings or in Swedish military officers after repeated blast exposure from the firing of heavy weapons during training (see slides).

Kevin Wang, University of Florida, Gainesville, showed examples of candidate blood-based protein biomarkers for TBI. He and his colleagues selected UCH-L1 (cell body injury), GFAP (glial injury), and SBDP150 (axonal injury), among others, as potential acute markers, while testing SBDP120 (axonal injury), MBP-fragment (demyelination), and MAP2 (dendritic injury) as sub-acute markers. They detected UCH-L1 in the CSF and serum of TBI patients, and their levels in the first 24 hours post-injury correlated with clinical outcome. UCH-L1 stayed elevated beyond 24 hours in CSF, but normalized in serum. UCH-L1 can be used as a biomarker of neuronal loss in aneurismal subarachnoid hemorrhage, based on a previous study, said Wang (see Lewis et al., 2010). Wang's data suggest that tau is vulnerable to calpains and caspase-3 under acute and sub-acute TBI conditions, and tau fragments might be potential contributors to CTE.

Using a human Aβ knock-in mouse model, which does not overexpress APP, **Milos Ikonomovic**, University of Pittsburgh, Pennsylvania, showed that simvastatin treatment after CCI injury lowered Aβ levels, suppressed microglial activation, reduced hippocampal synaptic and neuronal loss, and restored hippocampus-dependent cognitive function (see slides). **Mark Burns**, Georgetown University, Washington, D.C., reported that APP accumulated in damaged axons as soon as 30 minutes post-TBI (CCI). Additionally, TBI triggered APP processing and production of Aβ monomers and soluble Aβ oligomers. DAPT (a γ-secretase inhibitor) treatment of TBI mice blocked abnormal increases in Aβ40 at 24 hours post-injury, dramatically reduced lesion volume, and rescued spatial learning and fine motor coordination at one month post-injury. However, CCI also caused global dendritic spine loss 24 hours after injury, even on the uninjured side of the brain, and DAPT treatment did not suppress it.

Fernando Gomez-Pinilla, University of California, Los Angeles, showed that FPI mice treated with a curcumin derivative exhibited more BDNF production, reduced oxidative stress (less 4-HNE), and increased availability of ATP (enhanced AMPK activity) compared with untreated FPI mice. A DHA supplement also counteracted the effects of FPI by restoring BDNF and CamKII levels and hippocampus-dependent cognitive function, said Gomez-Pinilla. In contrast, low DHA diet or diets high in calories (fructose or saturated fats) attenuated BDNF function and triggered more anxiety-like behavior in both sham and FPI mice, compared to DHA-fed controls. Besides a proper diet, exercise may benefit TBI since voluntary exercise caused an epigenetic change on the BDNF gene in rodents, which led to more BDNF mRNAs and protein production in the hippocampus (see slides).