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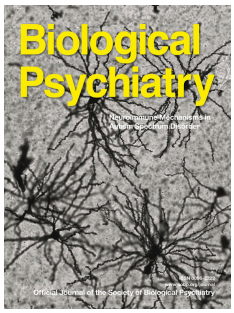

Neurochemical markers of traumatic brain injury – relevance to acute diagnostics, disease monitoring, and neuropsychiatric outcome prediction

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Neurochemical markers of traumatic brain injury – relevance to acute diagnostics, disease monitoring, and neuropsychiatric outcome prediction

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Abstract

Considerable advancements have been made in the quantification of biofluid-based biomarkers for traumatic brain injury (TBI), which provide a clinically accessible window to investigate disease mechanisms and progression. Methods with improved analytical sensitivity compared with standard immunoassays are increasingly utilized, which has opened for the use of blood tests in the diagnosis, monitoring, and outcome prediction of TBI. Most work to date has focused on acute TBI diagnostics, whilst the literature on biomarkers for long-term sequelae is relatively scarce. In this review, we give an update on the latest developments in biofluid-based biomarker research in TBI and discuss how acute and prolonged biomarker changes can be used to detect and quantify brain injury and predict clinical outcome and neuropsychiatric sequelae.

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Introduction

Traumatic brain injury (TBI) is a major cause of death and disability worldwide (1). Clinically, TBI is classified as mild, moderate, or severe based on loss of consciousness, post-traumatic amnesia, and structural damage on head CT or MRI. Most of the TBI cases are concussive or mild. The current clinical imaging techniques, although useful for diagnosis of moderate to severe TBI, are not sensitive enough to detect subtle brain injury. Biofluid-based biomarkers may be complementary and/or alternative methods to detect and quantify the amount of injury to different cell types and structures of the brain, as well as tissue reactions to and recovery processes following a TBI. Recently, the United States Food and Drug Administration (FDA) cleared blood glial fibrillary acidic protein (GFAP) and ubiquitin C-terminal hydrolase L1 (UCH-L1) for prediction of absence of intracanial injuries on head CT (2-4). Additionally, the Scandinavian Neurotrauma Committee (SNC) has proposed serum S100 calcium-binding protein B (S100B) for detection of intracranial findings following head trauma (5, 6). These blood-based biomarkers have been shown to be useful diagnostic tools, and may reduce the usage of CT scans in the Emergency Department (ED) setting (3, 7).

The outcome following TBI is highly variable (8, 9). The traditional view is that most individuals who sustain a concussive or mild TBI recover within days to weeks (10). In about 15% of individuals, post-concussive symptoms persist for more than a year, which is referred to as post-concussive syndrome (PCS) (11, 12). The symptoms of PCS can be generally categorized into four domains: physical, cognitive, emotional, and sleep. PCS, albeit an outdated term due to lack of granularity, still gives an indication of the long-term impact of a mild TBI. Biofluid-based biomarkers that correlate with or predict physical, cognitive, emotional, and sleep outcomes following TBI would be very useful, especially in the clinical setting.

In this review, we give an update on the latest developments in biofluid-based biomarker research in TBI, and discuss how acute and prolonged biomarker changes can be used to detect and quantify brain injury and predict clinical outcome.

Disease mechanisms in TBI of relevance to fluid biomarkers

TBI is characterized by multifaceted post-injury acute and chronic processes that may contribute to recovery and neurodegeneration. Acute TBI results in axonal injury with release of cytoskeletal proteins, disrupted axonal transport of proteins like amyloid β (Aβ),

phosphorylated tau (P-tau), TAR DNA-binding protein 43 (TDP-43) and α-synuclein that may build-up in the brain tissue and contribute to neurodegenerative processes, along with neuroinflammatory responses, including microglial and astrocytic activation, as well as injury to oligodendrocytes and cellular and structural components of the neurovascular unit.

Fluid biomarkers for axonal injury

One of the most well-studied axonal protein is tau, a microtubule-associated protein predominantly expressed in short cortical unmyelinated axons (Table 1) (13). Increased concentrations of cerebrospinal fluid (CSF) total tau (T-tau) have been found in acute samples from patients with moderate to severe TBI, where the initial levels correlated with 1-year functional outcome (14). In a study of Olympic boxers who underwent repeated lumbar punctures (LPs), CSF T-tau increased 7-10 after a bout and normalized after three months of rest (15). With the advances in immunoassay technology, T-tau could also be quantified in blood with high analytical sensitivity (16). Plasma T-tau was measured in blood samples from professional ice hockey players within hours after a sports-related concussion (SRC) and the levels were increased compared with the preseason baseline (17, 18). In the context of chronic SRC, a recent study found no difference in plasma T-tau in National Football League players with a history of repetitive head trauma compared with controls (19).

Another axonal protein that has garnered a lot of attention is neurofilament light (NfL). NfL is a component of the axonal cytoskeleton and is primarily expressed in large-caliber myelinated axons (Table 1) (20). CSF NfL is a sensitive biomarker of neuroaxonal damage (21), and has been validated in several neurodegenerative disorders (22-28). In 2016, the ultrasensitive assay for quantification of NfL in serum or plasma was first tested in a TBI context (29). In patients with acute moderate to severe TBI, serum NfL could distinguish these patients from healthy controls with area under the receiver operating characteristic curve (AUROC) of 0.98-1.0 (29). Furthermore, serum NfL showed moderate to strong correlations with both CSF and ventricular CSF from the same individuals (29). In subsequent studies, serum NfL measured within 48 hours of injury could also distinguish patients with CT findings from those with normal CT with high accuracy (2, 30, 31). In a recent study conducted in clinic-based patients with a history of mild, moderate or severe TBI who were followed with serial blood samples from 30 days up to five years, serum NfL could distinguish patients with mild, moderate, or severe TBI from each other as well as controls (32). Serum NfL concentration at 1-year correlated with global brain volumes measured at the same time point, as well as with

diffusion tensor imaging measures of white matter integrity (32). NfL has also been measured in professional ice hockey players with acute concussion, where higher levels in serum were seen in players with prolonged return to play (RTP) (18). Furthermore, serum NfL performed better than plasma T-tau in distinguishing concussed athletes from controls (18). In the context of subacute and chronic repetitive head impact, NfL measured 7-10 days after a bout was elevated compared with controls and the levels decreased after 3 months of rest (33). Furthermore, serum NfL correlated with the corresponding CSF values (r=0.86), as well as with number of hits received to head (33). In a recent study of professional athletes with a history of repetitive SRC who underwent LP and blood assessment months after the most recent SRC, serum NfL correlated with CSF (r=0.76), and serum NfL could distinguish concussed athletes from controls with high accuracy (32).

Both plasma T-tau and serum NfL have been assessed in relation to strenuous exercise or body trauma (Table 1). Plasma T-tau has been shown to be more sensitive to body trauma, while serum NfL is not affected by body trauma or strenuous exercise (18).

In regards to the temporal profile of T-tau and NfL quantified in blood, T-tau seems to be an acute biomarker, especially in concussion, while NfL in serum peaks 7-10 days after a head trauma and may be detectable months to years after injury (18, 32, 34) (Table 1).

Fluid biomarkers for astrocytic activation

S100B is a protein primarily expressed by astrocytes and was the first biomarker to be proposed for clinical use by the SNC (5, 6). In a metanalysis of 2466 patients with TBI, S100B had a pooled sensitivity, specificity, and negative predictive value of 97%, 40%, and 99%, respectively in predicting CT findings (35). These findings have been confirmed in additional studies (6, 36). Furthermore, S100B led to a 32% reduction in unnecessary head CTs compared to either the Canadian CT Head Rule or the New Orleans Criteria. In another study, a secondary elevation of S100B following TBI was shown to significantly predict secondary pathological CT/MRI findings (mainly ischemic-like lesions) in mild to severe TBI patients with high sensitivity and specificity (37). This was superior to common clinical features (pupil response, Glasgow Coma Scale, admission CT findings, intracranial pressure, and hemoglobin levels) used to predict secondary pathological findings (37).

In the context of SRC, serum S100B increased 1 hour after a concussion compared with preseason control results (18). However, when compared to NfL and tau, S100B had lower

diagnostic and prognostic value (18). A major limitation of S100B is that it significantly increased after strenuous exercise (18). Similarly, other studies have reported exercise-related elevation in S100B (38-40). The increase in S100B observed in these studies may be due to the fact that S100B expression is found in adipocytes, skeletal tissue and various other organs (41-44).

Another biomarker of astrocyte reactivity is GFAP, which is an intermediate filament protein (Table 1) (45). Serum GFAP has been shown to distinguish TBI patients with intracranial findings on head CT from those without with high accuracy (2-4). Recently GFAP and UCH-L1 (a protein abundantly found in neurons) were cleared by the FDA for detection of intracranial injury on head CT following TBI (4). In the context of acute TBI, serum GFAP and UCH-L could distinguish patients with intracranial lesions on CT from those without with high accuracy. In the largest study of GFAP and UCH-L1 to date, including 1959 patients with mild to moderate TBI, serum GFAP and UCH-L measured within 12 hours of inury had high sensitivity and negative predictive value for the detection of traumatic intracranial injury on head CT (46). Several studies have assessed the combination of GFAP and UCH-L1 for predicting CT fidings following acute TBI. The combination of GFAP and UCH-L1 performed better than either biomarker alone in predicting intracranial injuries on head CT following TBI (46-48). Recent studies have found that GFAP alone may perform similarly as the GFAP and UCH-L1 combination for predicting CT findings following mild TBI (4, 49, 50). These studies also found that GFAP or the GFAP and UCH-L1 combination outperformed S100B for predicting CT findings (48, 49, 51). Several studies have also compared GFAP and UCH-L1 with NfL and T-tau in predicting intracranial pathology on head CT or brain structural MRI. In one of these studies, serum GFAP performed similary or slightly better than NfL, while T-tau perfomed worse in detecting MRI findings following a mild TBI and UCH-L1 had variable levels (2). In another study, serum GFAP, UCH-L1 and NfL had almost similar performance, while T-tau perfomed worse in detecting CT pathology associated with TBI (52).

In the context of SRC, increased levels of GFAP measured within 48 hours after a concussion have been seen in collegiate athletes (53, 54). In a recent study, concussed collegiate athletes had increased concentrations of serum GFAP, NfL, and UCH-L1 measured within 24-48 hours after a concussion compared with preseason baseline, with the highest concentrations in concussed athletes with loss of consciousness or post-traumatic amnesia (55). The levels of

GFAP and NfL remained elevated for several days in these types of concussions. In another recent study, GFAP, NfL, and UCH-L1 increased in United States cadets who sustained a concussion, as well as in cadets who participated in the same combative training excercise but did not incur a concussion (56). These recent studies provide support for potential utility of blood biomarkers, especially GFAP and NfL, for SRC or military concussion.

Serum S100B is an acute biomarker that peaks within hours after injury (Table 1). The utiliy of S100B beyond the acute phase and in relation to TBI severity is yet to be examined in detail. Serum GFAP has been shown to increase acutely after injury (Table 1). Recently, we found that GFAP is detectable in serum following mild, moderate, or severe TBI even months to years fter head trauma (57). A drawback of GFAP as a biomarker of intracranial injury on head CT following TBI is that it seems to perform worse in older patients (58) (Table 1).

Fluid biomarkers for injury to oligodendrocytes

Myelin basic protein (MBP) is a marker of oligodendrocytes (Table 1), which is detectable in blood and indicates potential disruption in myelin (59-61). In animals exposed to various degrees of blast TBI, MBP was elevated in serum (59). Elevated serum MBP was also seen in patients with severe TBI (59-61). The marker has not been examined in mild TBI.

Fluid biomarkers for microglial activation

Microglia are found throughout the CNS, where their main function is to clear damaged cells and synapses, as well as infectious agents (62). Following TBI, microglia can clear cell debris and orchestrate neurorestorative processes that are beneficial to neurological recovery. Microglia can also produce pro-inflammatory and cytotoxic mediators that hinder CNS repair and further contribute to neuronal dysfunction and cell death. The shift between these two opposite functions is not well-understood. Triggering receptor expressed on myeloid cells 2 (TREM2) is a receptor mainly expressed on the surface of the microglia (63). Recently TREM2 has been found to play a role in the In Alzheimer's disease (AD). CSF soluble TREM2 (sTREM2) has been found to be increased in patients with AD as compared to controls (64). The availability of the CSF assay for sTREM2 also opens a window of opportunity for assessing the potential role of microglia in human TBI.

Fluid biomarkers for disruption of the neurovascular unit/BBB

TBI causes disruption of the BBB integrity (65). Clinically, CSF:serum albumin ratio is commonly used as a surrogate marker of BBB integrity (66). In the context of TBI, CSF:serum albumin ratio was measured in a study of professional athletes with a history of repetitive heads trauma (67), where the levels of CSF:serum albumin ratio was unaltered. A plausible explanation could be that CSF:serum albumin ratio may not be a sensitive enough measure of BBB integrity, or that BBB integrity may be disrupted in the acute phase of the injury but not in the chronic phase as this study was performed.

Another biomarker of BBB leakage is soluble platelet-derived growth factor receptor (sPDGFRβ), a protein highly expressed in pericytes of the vasculature (68, 69). Increased CSF sPDGFRβ has been reported in patients with AD compared to controls, where the levels of CSF sPDGFRβ correlated with CSF:serum albumin ratio (70). In the context of TBI, sPDGFRβ is yet to be examined.

Fluid biomarkers for TBI-related proteinopathies

TBI may also cause tangle pathology, which consists predominantly of P-tau (71) (Table 1). Recently, phosphorylated tau (P-tau; using antibody that specifically recognizes phosphothreonine-231) and T-tau were measured in plasma samples from 217 TBI patients, where P-tau and P-tau to T-tau ratio demonstrated perfect discrimination of mild TBI from controls (AUROC of 1.0) (72). The ratio of P-tau to T-tau also showed strong ability to predict positive CT findings (AUROCs 0.921 and 0.923, respectively) (72). In another study, P-tau and GFAP together performed significantly better for predicting CT findings than either biomarker individually (AUROC 0.96) (58). A recent meta-analysis found that the most promising biomarkers for predicting CT findings in TBI were GFAP in combination with UCH-L1, although P-tau was comparable while S100B was significantly lower (AUROC 0.98, 0.92, 0.72, respectively) (73). In the context of SRC, P-tau was measured in CSF of 16 professional athletes with a history of repetitive concussion and 15 healthy controls and there was was no significant difference in the levels of CSF P-tau between the groups (74), suggesting that the marker may not detect long-term P-tau changes, although more studies are needed.

Experimental and post-mortem studies suggest that athletes who have had repetitive head trauma may develop brain amyloid deposition (seen in 43% of cases) (75-77). The amyloid

deposition or plaques seen in TBI are predominantly composed of 42 amino acid-long and aggregation-prone amyloid β (Aβ42) (Table 1), which are also seen in AD (78, 79). In a study, Aβ40 and Aβ42 were measured in CSF from professional athletes with a history of repetitive concussions, and both CSF Aβ40 and Aβ42 were decreased with the highest effect size seen for Aβ42 (74), suggestive of potential brain amyloid pathology. Altered Aβ42 has also been observed in CSF and plasma of patients with acute severe TBI. In a study, decreased CSF Aβ42 concentration was seen in 12 patients with severe TBI compared with 20 controls when measured acutely (80). In another study, Aβ42 was measured in plasma collected at 24 hours, 30, and 90 days following TBI from 34 TBI patients and 69 healthy volunteers, where the levels of Aβ42 were significantly increased at all measured time points (81).

In addition to the classic pathologies of tangles and amyloid plaques observed in some individuals with TBI, especially those with chronic traumatic encephalopathy, TBI is also associated with TDP-43 inclusions and less commonly with α-synuclein inclusions (82). Currently, there are no reliable fluid assays to quantify TDP-43 or α-synuclein inclusions in individuals with TBI. With advances in the detection of misfolded seeds of α-synuclein in biofluids using real-time quaking-induced conversion or protein misfolding cyclic amplification (similar technologies to qualitatively detect trace amounts of diffusible misfolded α-synuclein, through its ability to induce aggregation of added recombinant α-synuclein in CSF over time, using thioflavin T fluorescence), brain α-synuclein pathology can be reliably detected in lumbar CSF from patients with Parkinson's disease and other synucleinopathies (83). While so-called real-time-induced has been used to quantify TDP-43 in CSF of patients with amyotrophic lateral sclerosis and frontotemporal dementia (84), this technique is yet to be utilized for quantification of TDP-43 or α-synuclein inclusions in individuals with TBI.

Novel candidate fluid biomarkers

A recent TBI biomarker avenue of research has been quantifying CNS-derived proteins contained in extracellular vesicles (EVs). There are several potential advantages to quantifying proteins in EVs: (1) EVs protect their content from degradation by endogenous proteases that are common in blood (85, 86); (2) EVs can easily cross the BBB (87); and (3) EVs are found to be more biologically active than proteins found within circulating blood

(88). In a study of veterans with a history of remote, elevated EV NfL was seen in those with history of multiple mild TBIs and elevated chronic neurobehavioral symptoms (89). Similarly, significantly increased EV tau and EV P-tau were found in veterans with a history of multiple mild TBIs compared to controls (88). In a recent study of civilians with a history of TBI, EV NfL and EV GFAP measured at 1-year after injury were elevated in patients with moderate to severe TBIs compared to controls, with EV GFAP performing better than EV NfL in distinguishing patients with moderate to severe TBIs from controls (90).

EVs may also contain microRNAs (miRNAs) released from injured neurons (91). miRNAs are found throughout the body and is particular essential to neuronal injury and repair (92, 93). Similar to the other established proteins measured in EVs, miRNAs due to their small size can transverse the BBB easily and make them attractive as potential biomarkers of TBI. Furthermore, miRNAs have been implicated in both the primary (91) and secondary damage responses to TBI (94). Several studies have investigated the role of miRNAs as biomarkers for TBI. A study compared EV RNA in the CSF of 11 severe TBI patients and 17 controls and found that most of the RNA packaged in CSF microparticles was non-coding RNA, and that two of these non-coding RNAS (miR-9 and miR-451) were differentially expressed in severe TBI patients (95).

Which of these biomarkers predict neuropsychiatric sequelae?

As mentioned earlier, TBI (even a mild one) may cause long-term neuropsychiatric symptoms, including cognitive and emotional symptoms and sleep disturbances (11, 12). In civilian patients hospitalized for an orthopedic injury, presence of comorbid mild TBI was associated with an increased risk of post-traumatic stress disorder (PTSD) and depression 3 to 6 months after injury (96). In another study of 91 patients with TBI and 27 patients with multiple traumas but without evidence of brain damage, major depressive disorder was significantly more frequent among patients with TBI than among the controls during the first year after sustaining a TBI (97). In military TBI, there is an increased risk of post-traumatic stress and depressive symptoms that may worsen over time (98). In the context of sports-related TBI, several studies suggest that symptoms of depression, anxiety and emotional lability are higher in concussed athletes, especialy those with a history of repetitive head trauma (99, 100). Emerging studies indicate that chronic symptoms of PTSD, depression, and neurobehavioral following mild TBI are associated with increased concentrations of neuronal injury proteins in peripheral blood. For example, increased PTSD symptoms in service

members have been associated with increased plasma tau (101). In another study, it was found that PTSD, depression, and neurobehavioral symptoms following TBI were associated with increased tau and NfL but not Aβ40 or -42 (102). In a recent study of veterans and service members with remote history of repetive mild TBI, higher concentrations of serum and EV NfL correlated with increased neurobehavior, PTSD, and depressive symptoms (89). In another study, we found increased concentrations of serum NfL to correlate with poor sleep and lower executive function socres following a remote mild TBI (103). In the context of SRC, we found higher concentrations of NfL correlated with Rivermead Post-Concussion Symptoms Questionnaire Scores both in athletes who have had an acute concussion (18) as well as those who developed chronic PCS (32). These studies, depite their caveats, suggest that axonal injury as measured by serum T-tau or NfL may underlie the severity of neuropsychiatric symptoms such as depression, neurobehavioral and PTSD-related symptoms.

Neuropsychiatric symptoms such as anxiety and depression go hand in hand with functional outcome following TBI. In a recent study, depressive and anxiety symptoms correlated strongly with function and disability measures in daily life (104). Currently, there are several candidate biomarkers that have shown promising prognostic utility (73). In a recent study, NfL and GFAP measured within 24 hours predicted unfavourable outcome (AUROC 0.75 and 0.82, respectively) (105). In a study of professional Swedish ice hockey players, low serum NfL predicted a more favorable functional outcome and lower risk of PCS (18, 33). In the same cohort, initial level of plasma T-tau predicted RTP, but had lower predictive value than NfL (18, 33), while S100B showed no associations with RTP. S100B has shown mixed results in predicting outcome in severe TBI. In a recent study, serum NfL measured at an average of 1 year injury correlated with functional outcome assessed at the same time, while no relationship with functional outcome was seen for GFAP, T-tau, or UCH-L1 (57). In one study of severe TBI, serum S100B measured within two weeks of injury could discriminate patients who would had unfavorable outcome (defined as severe disability, vegetative state, or death based on the Glasgow Outcome Scale [GOS]) from favorable outcome (moderate, mild disability or no disability based on the GOS) at 12 months (106). In the same study, S100B was compared with UCH-L1, GFAP, and NfL, and NfL and GFAP added the most independent information in predicting functional outcome, while S100B was the least useful (106). In another study, S100B was not associated with outcome at 12 months, while serum NfL measured within 24 hours after injury was associated with outcome at 12-month (29).

Synthesis and conclusion: what additional research is needed?

Several of the existing large-scale biofluid-based biomarker studies have been focused on distinguishing TBI from controls. In the ED setting, GFAP, UCH-L1, and S100B have been shown to be useful in distinguishing patients with trauma-related cranial CT findings from those without. Several recent studies have found that a panel of biomarkers may outperform individual biomarkers, especially with regard to diagnostic or predictive value (4, 47). For example, a combination of GFAP and UCH-L1 performs better than individual values in predicting the presence of intracranial pathology (4, 47). There is a scarcity of literature assessing these biomarkers in the subacute or chronic phase of TBI and longitudinally, which is an important topic for future research.

NfL has been shown to be an excellent biomarker for assessing axonal injury following various TBI severity and over months to years after TBI (32). However, serum NfL reflects one aspect of the TBI pathophysiology, and there is a need for assessing other pathologies such as tangles, amyloid deposition, astrogliosis, microglial activation, and BBB disruption. Therefore, we may need a panel of biomarkers for TBI. Blood assays for several of these pathologies are under development or refinement, however, the are few studies that have assessed these assays in TBI.

Finally, recent studies have assessed the relationship between GFAP, T-tau, NfL, and Aβ42, where higher levels of T-tau, GFAP and NfL with elevated neuropsychiatric symptoms following TBI (101, 102) or worse functional outcome (18, 29, 33, 57, 105, 106). Although these recent studies show promise for utility of these biomarkers for further understaning of the impact of neuropsychiatric symptoms, larger longitudinal studies are needed to adress whether initial levels these biomaerker would predict neuropsychiatric outcomes.

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Conflicts of interest

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Table 1. Commonly studied fluid biomarkers of TBI				
Biomarker	**Pathology**	**Measurement source**	**Acute or chronic TBI**	**CNS specificity**
T-tau	Expressed in unmyelinated cortical axons.	Blood and CSF. The correlation between blood and CSF is weak.	Elevated in acute and chronic brain injury.	To certain degree, also expressed in peripheral nerves and renal tubules (107).
NfL	Expressed in myelinated subcortical axons.	Blood or CSF. Strong correlation between blood and CSF levels.	Elevated in acute, subacute, or chronic brain injury. Remains elevated up to five years after TBI.	Mainly specific to brain but also found in peripheral nervous system (107).
S100B	Expressed in astroglia cells.	Blood and CSF.	Elevated mainly in acute TBI.	No, also expressed in skeletal muscles and adipocytes (41-44, 107).
GFAP	Expressed in astroglia cells.	Blood and CSF	Acute and subacute; Peaks at ~20 hours, thereafter declines. In a recent study declined but rerose at ~ 6 months (57).	Yes.
MBP	Expressed in oligodendroglia and Schwann cells.	Blood and CSF.	Acute.	To certain degree; also expressed also in peripheral nervous system.
UCH-L1	Expressed in neuronal cells.	Blood and CSF.	Acute; peaks at ~ 8 hours.	To certain degree; also expressed in distal renal tubules and islets of Langerhans (107).

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P-tau	Expressed in neurofibrillary tangles	Blood and CSF	Acute and subacute	Likely yes.
Aβ42	Expressed in amyloid plaque and synapses	Blood and CSF. Weak correlation between the blood and CSF.	In blood and CSF elevated acutely after TBI. In CSF elevated in chronic concussion (67).	Likely yes.
CSF:serum albumin ratio	Surrogate marker of BBB function.	Requires paired blood and CSF and studies, which are very few. No increases in chronic concussion (67).	Not yet assessed in acute concussion and not significantly altered in chronic concussion.	
Abbreviations: T-tau = total tau; NfL = neurofilament light; S100B = S100 calcium-binding protein B; GFAP = glial fibrillary acidic protein; UCH-L1 = ubiquitin c-terminal hydrolase L1; MBP = myelin basic protein; P-tau = phosphorylated tau; Aβ42 = amyloid- β 42; CSF = cerebrospinal fluid; BBB = blood-brain barrier				

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