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Biomarkers of Acute Brain Injury in the Emergency Department

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<http://dx.doi.org/10.5772/64222>

Abstract

The diagnosis of acute brain injury in the acute care setting is based on neurological examination and neuroimaging tools such as computed tomography (CT) scanning and magnetic resonance imaging (MRI). However, there are limitations to both CT and MRI scanning. The lack of objective, noninvasive and readily accessible clinical tools to detect injury has left clinicians with uncertainty about how to best identify and treat these conditions. It is also very difficult for patients and their families who struggle to better understand the deficits they deal with on a daily basis. There have been many studies exploring many promising biomarkers during the last decade. Despite the large number of published studies there is still a lack of any Food and Drug Administration (FDA)-approved biomarkers for brain injury in adults and children. Given all of these researches, there is now an important need to validate and introduce them into the clinical setting. This chapter reviews commonly studied biomarkers for acute brain injury in humans, with an emphasis on traumatic brain injury and stroke.

Keywords: biomarkers, acute, brain injury, traumatic brain injury, stroke, ischemic stroke, hemorrhagic stroke, subarachnoid hemorrhage, emergency department, neurosurgery, neuroimaging, neurosurgical intervention, migraine, diagnosis, prognosis, blood test, serum, cerebrospinal fluid, glial fibrillary acid protein, ubiquitin C-terminal hydrolase, S100B, tau, spectrin breakdown products, neurofilaments, neuron specific enolase, CT scan, magnetic resonance imaging (MRI)

1. Introduction

1.1. Epidemiology of acute neurological diseases and the evolution of acute brain injury biomarkers

Neurological biomarkers have considerable diagnostic and prognostic promise given their variety, range, and specificity, though despite the potential advantages of their use and the

array of conditions to which they apply, biomarkers have yet to be broadly employed in the clinical setting [1–4]. Before assessing the roles of biomarkers in the diagnosis and prognosis of various acute neurological conditions, some consideration should first be given to the epidemiology of these conditions. Traumatic brain injury (TBI) results from external blunt force trauma to the head – most often through motor vehicle accidents, falls, and sports injuries – and is marked by cognitive and motor deficiency, the severity of which is determined via the Glasgow Coma Scale (GCS). TBI may be associated with bleeding (hematoma), brain swelling (cerebral edema), hydrocephalus, herniation, increased intracranial pressure, and microscopic damage to the neuronal and astrocyte network. An estimated 40% of acute injury deaths in the United States are attributed to TBI, with mortality rates highest among those aged 15–24 or 65 and older; individuals who are part of ethnic minority groups and possess lower socioeconomic status also suffer increased risk for TBI.

Stroke, comparatively, is characterized by either an ischemic and a hemorrhagic insult and is both the third most common cause of mortality in the United States and, along with ischemic heart disease, the leading cause of mortality worldwide. Moreover, stroke is also the second leading cause of disability among the global population, with resulting complications ranging from vision impairments and aphasia to varying degrees of paralysis, short-term memory loss, dementia, and difficulty concentrating and learning novel information. Aneurysmal subarachnoid hemorrhage (SAH) is a degenerative sub-condition of stroke, inciting hydrocephalus, vasospasm, and continuous bleeding in 26–73% of patients. While the incidence of SAH in the United States ranged, as of 2010, from roughly 0.015 to 0.287% of the population, the mortality rate of SAH sufferers varies from 40 to 60%. Both age-controlled stroke incidence and subsequent medical and economic complications have especially intensified in both developing nations and lower-income communities. The symptomology and neuroimaging data of SAH in particular can often resemble that of acute migraine, for which it is mistaken in roughly 12–51% of diagnoses. Acute migraine afflicts approximately 18% of women and 6% of men in the United States. Despite the widespread prevalence of migraine and the significant physical and financial burden it poses to sufferers, the condition is often underdiagnosed.

In light of practitioners' concerns with timeliness, accuracy, and risk aversion in the diagnostic and treatment processes, serum biomarkers with reliable specificity for and sensitivity to various categories of brain injury are appetizing diagnostic and monitoring tools. This is especially so in contexts haunted by indeterminate, unavailable, or simply untimely neuroimaging, hence why biomarker panels would be of particular utility in more rural settings and non-hospital environments where rapid triage is especially critical. Although promising cerebrospinal fluid (CSF) biomarkers for hemorrhage, migraine, and stroke have been studied, serum biomarkers are needed. Having a blood test would eliminate the need for invasive procedures such as lumbar puncture, which tends to be a lengthy and physically uncomfortable process with potential complications. Biomarkers have also shown potential in evaluating injury severity such as brain infarction and in predicting post-stroke prognosis, thereby aiding health care workers in their assessment of the level of post-stroke care required by individual patients. It must be noted, however, that while both serum and CSF biomarkers are examined in the following chapter, the promptness with which serum samples can be collected and

analyzed far exceeds that of CSF biomarkers; in addition, serum assays have the advantage of being able to be collected in the field or in settings where access to technical equipment is limited.

This chapter reviews some of the most widely studied biomarkers for acute brain injury in the clinical setting, with an emphasis on traumatic brain injury and stroke.

2. Biomarkers of astroglial injury

2.1. S100 β

S100 β is found in astrocytes and is a low-affinity calcium-binding protein [5] that helps to regulate intracellular levels of calcium. It is considered a marker of astrocyte injury or death. It can also be found in cells that are not neuronal such as adipocytes, chondrocytes, and melanoma cells, making it non-brain specific [6, 7]. S100 β is one of the most extensively studied biomarkers for brain injury [2, 3, 8]. Elevated S100 β levels in serum have been associated with increased incidence of post-concussive syndrome and problems with cognition and MRI abnormalities [9–13]. However, there are also a number of studies negating these findings [14–17]. Since many of these results have not been consistently reproduced, the clinical value of S100 β in TBI, particularly mild TBI and concussion, is still controversial. A number of studies have found correlations between elevated serum levels of S100 β and CT abnormalities in adults and children [3, 18]. Unfortunately, its utility in the setting of polytrauma remains controversial, because it is also elevated in trauma patients with peripheral trauma who had no direct head trauma [19–21].

S100B seems an equally suspect diagnostic tool for acute ischemic stroke. Several findings have placed peak S100B elevation at days 2–4 following acute ischemic stroke onset, rendering its measurement somewhat fruitless in the majority of ischemia cases in which recombinant tissue plasminogen activator (rt-PA) generally needs to be performed within the first three or, more rarely, 4.5 h, post-infarction onset. However, concerns with S100B's specificity are potentially less problematic when distinguishing hemorrhagic from ischemic stroke. Because morphological damage tends to be more immediate in cases of hemorrhaging than in ischemia, S100B concentrations have been shown to peak earlier in cases of hemorrhage, at roughly 24 h post-infarction onset. This timescale, however, might still be too prolonged to render S100B a viable diagnostic marker, although it should be noted that CAT scans are routinely given 24 h after ischemia onset in order to rule out hemorrhage. Moreover, research has tended to focus on the potential association between S100B and ischemia rather than hemorrhage.

Instead, S100B offers more promise as a prognostic tool in the evaluation of infarction severity in coordination with MRI results and, relatedly, patient outcome and functionality. S100B concentrations within the first 2–10 days after onset have been found to be predictive of infarct volume, potentially more so than NSE levels are, and moreover have correlated well with assessments of neurological functionality as measured by the National Institutes of Health Stroke Scale (NIHSS) [22]. The prognostic value of S100B is also exhibited in its potential to

predict hemorrhagic transformation in patients who have suffered ischemia within 24 h. Moreover, Fassbender et al. found that ischemia resulting in lesions greater than 5 cm in volume was correlated with significantly greater concentrations of S100B 10, 24, and 72 h post onset [22].

S100B has also been studied as a possible means by which to distinguish aneurysmal subarachnoid hemorrhage from acute migraine. Aneurysmal SAH is a degenerative condition, inciting continuous bleeding in 26–73% of patients [23, 24], as well as hydrocephalus and vasospasm [25]. It is not uncommon for SAH to be initially mistaken for migraine, especially since headache is a prominent symptom of both conditions, with the frequency of overall SAH misdiagnosis between 12 and 51% [26–30]. Indeed, physical symptoms of SAH and severe, acute-onset migraine are remarkably similar to one another. CT and MRI scans remain the most reliable means by which SAH can be diagnosed, the former more accurate in detection within the first 24 h following attack and the latter more accurate after 24 h have elapsed since attack onset. However, vasospasm can render imaging of the aneurysm through magnetic resonance angiography Magnetic resonance angiography (MRA) difficult. The error rate of such scans, especially due to their qualitative nature, may perhaps be improved upon by the use of diagnostic biomarkers. S100B concentrations have been shown to rise in response to acute migraine, peaking 2–4 days after onset during the “pain-free period,” but detectable within 2–3 h of onset [31, 32]. Because S100B concentration time courses in SAH and migraine are similar, biomarkers such as NSE, which are characterized by more distinctive patterns in acute migraine, are likely more useful diagnostic tools.

2.2. Glial fibrillary acid protein (GFAP)

Glial fibrillary acidic protein (GFAP) is a protein found in astroglial skeleton and is found in both white and gray brain matter. It was first isolated by Eng et al. [33] and appears to be strongly upregulated during astrogliosis [34]. Serum GFAP has been shown to be elevated with various types of brain damage including neurodegenerative disorders [35, 36], stroke [37], and severe traumatic brain injury [19, 38–43]. In particular, GFAP has become a very promising brain-specific glial-derived biomarker for mild TBI in adults and children [14, 20, 21, 44–47]. GFAP is released into serum following a mild TBI within an hour of injury and remains elevated for several days after injury [21, 44, 47]. Unlike S100 β , GFAP is elevated in mild TBI patients with axonal injury as evidenced by MRI at 3 months post injury [14]. In adults and children, serum GFAP levels distinguish mild TBI patients from trauma patients without TBI and detect intracranial lesions on CT with a sensitivity of 94–100% [20, 21, 44, 46, 47]. Moreover, GFAP outperforms S100 β in detecting CT lesions in the setting of multiple trauma when extracranial fractures are present [21, 46]. GFAP also predicts the need for neurosurgical intervention in patients with mild TBI [44, 47]. The temporal profile of GFAP was evaluated in a large cohort of 584 trauma patients seen at the emergency department. GFAP performed consistently over 7 days in identifying concussion and mild to moderate TBI, detecting traumatic intracranial lesions on head computed tomography (CT), and predicting neurosurgical intervention [47].

GFAP promises to be an especially potent biomarker, perhaps providing the opportunity to reliably distinguishing ischemic and hemorrhagic stroke. The necessity of expeditiously distinguishing between the two subsets of stroke poses a particular difficulty because of the manner in and extent to which treatment paradigms vary between the stroke classes. While the securing of endovascular coils or surgical implantation of clips at the site of the aneurysm are viable treatment options for hemorrhage, rt-PA, a recombinant form of an endogenous serine protease used for thrombolysis, offers the only Food and Drug Administration (FDA)-approved treatment for ischemic stroke beyond supportive care. Studies have shown that the risk-benefit ratio of rt-PA administration is favorable for patients treated within 3 h of stroke onset, although some institutions place the threshold to 6 h [24, 48–51]. Mechanical thrombectomy, an endovascular procedure in which the offending clot is excised via stent retriever, is becoming an increasingly explored treatment method viable up to 6 h within the onset of stroke symptom presentation [52–54]. Even this treatment strategy for ischemic stroke, however, is not usually undertaken without having first administered rt-PA to the patient [55].

For both ischemic and hemorrhagic stroke, patient outcomes improve with more rapid treatment administration. Although swift use of rt-PA would benefit patients suffering from ischemic stroke, which comprises roughly 87% of stroke cases, clinical practitioners must be circumspect in their use of the treatment because of its detrimental effects on the 13% of individuals presenting with hemorrhagic stroke [56]. The administration of rt-PA in response to ischemic stroke, however, can only be as prompt as proper diagnosis. Healthcare providers are thus left with the difficult task of both rapidly confirming the diagnosis of stroke to ensure eligibility for treatment, and rapidly confirming that a patient is not at risk for significant hemorrhage [24, 48–51]. CT and MRI scans are currently the primary diagnostic means at a physician's disposal, though assessment also includes a focused medical history, physical exam, and blood work. However, CT scans are more readily available in most settings than MRI [25]. In some communities, portable CT scanners are available for use by emergency responders, though this particular technology is neither widely available nor cost-effective. MRI, in comparison, can be more sensitive than CT scans in determining the presence of both intracerebral hemorrhage and the degree of ischemia [26, 49]. However, recent studies have shown that even MRI scans can miss roughly 17% of strokes [28–30, 57, 58]. CT scans are even less effective. Studies indicate that less than half of patients with ischemic stroke will show characteristic changes on CT scan within 3 h of symptom onset [58]. Even more advanced imaging techniques such as perfusion-diffusion mismatch models have received criticism for their ability to accurately predict lesion growth. Hence, current imaging techniques, despite providing arguably our most reliable stroke diagnoses, may temporally limit the diagnosis of stroke and delay the provision of timely treatment. Stroke biomarkers have been suggested by many investigators as an opportunity to improve our ability to diagnose and treat stroke in a timely and safe manner [59].

Recent studies have indicated that the pathophysiological kinetics of serum GFAP, an intermediate filament protein expressed in astroglial cells, may render GFAP a promising contender in the search for acute biomarkers [47]. Trauma or disease-induced cellular necrosis in the brain and spinal cord are known to augment GFAP levels released into the plasma, and

numerous findings have suggested that the more immediate structural damage to the artery and blood-brain barrier caused by hemorrhage in comparison with ischemic stroke renders the subsequent surge in GFAP levels a reliable indicator of hemorrhage specifically [60–63]. Within the first 6 h of stroke onset, significantly higher levels of serum GFAP have been found in patients suffering from hemorrhage than in those with ischemic stroke [62, 63]. Indeed, both GFAP and other biomarkers generally indicative of stress and morphological trauma, such as S100B, do not seem to peak until 2–4 days following ischemic stroke onset [63, 64]. In cases of hemorrhage, these levels decline roughly 6–12 h after symptom onset, within a time window before which biomarker surges are observed in ischemic stroke [63]. It should be noted, however, that S100B may not be as reliable and sensitive an indicator of hemorrhage as GFAP, as S100B levels are not necessarily as elevated within the first few hours of SAH as they are in intracerebral hemorrhage. Thus, diagnostic strategies employing GFAP would be expected to overlook fewer cases of hemorrhage. Moreover, serum GFAP levels have also accorded well with observational features, with this protein concentration directly proportional to acute stroke severity (as measured by NIHSS) and Intracranial Hemorrhage (ICH) volume and inversely proportional to functionality as long as 3 months after stroke onset. Cutoff points for serum GFAP level significance, however, have ranged from 0.29 to 2.9 $\mu\text{g/L}$ [62, 63].

3. Biomarkers of neuronal injury

3.1. Neuron specific enolase (NSE)

Neuron specific enolase (NSE), an isozyme of the glycolytic enzyme enolase, is found in central and peripheral neuronal cell bodies. It increases in serum following cell injury [65] and has a biological half-life of 48 h. Notably, it is also present in erythrocytes and endocrine cells [66]. NSE is passively released into the extracellular space only under pathological conditions during cell destruction. Several studies have been published examining serum NSE following mild TBI [65, 67–70]. Many of these reports contained inadequate control groups and noted that serum NSE had limited utility as a marker of neuronal damage after trauma. Early levels of NSE concentrations have been correlated with outcome in children, particularly those under 4 years of age [71–74]. In the setting of diffuse axonal injury (DAI) in severe TBI, levels of NSE at 72 h of injury have shown an association with unfavorable outcome [75]. One of the limitations of NSE is the occurrence of false positive results in the setting of hemolysis [76, 77].

Data surrounding potential correlations between NSE concentration and ischemia are somewhat mixed and thus require continued research. Missler et al. found that serum concentrations of NSE in patients suffering from acute ischemia did increase, peaking on day 1.9 after onset and were significantly correlated with infarction volume as measured by CT [78]. NSE levels, however, were not found to correlate with outcome as determined by the GCS during either discharge or 6 months post onset [78]. Martens [79] studied NSE levels in patients who had lapsed into unconsciousness following acute global ischemia and found significant differences in serum and CSF NSE concentrations between those who regained consciousness and those who died or slipped into a vegetative state. Infants suffering from hypoxic ischemic

encephalopathy (HIE) were marked by significantly increased concentrations of serum NSE 4–48 h and 5 days after birth, in comparison to healthy controls, and moreover expressed higher levels of NSE when suffering from the more severe stage III HIE rather than stages I or II, with NSE levels predicting poor outcome [80]. Jung et al., however, reported that while CSF NSE levels rose in response to SAH, they failed to correlate with the resulting cerebral vasospasm as glycine, glutamate, histidine, and glutamine did. Additionally, serum NSE levels failed to correlate with vasospasm development and ischemia in general [81]. In cases of hemorrhage, results about serum NSE were similarly mixed. Moritz et al. found that among patients who had suffered spontaneous SAH, both mean and peak concentrations of CSF but not serum NSE sampled within 8 days of onset predicted high or low performance on the GCS and predicted cerebral infarction and intracranial hypertension, but not vasospasm [82]. While Kuroiwa et al. did not observe a correlation between SAH and intracerebral hemorrhage patients' serum NSE concentrations and initial state of consciousness and neurological profile at admission, serum NSE levels tended to be higher in those with higher Fisher CT scores of 3 or 4 [83]. Moreover, those who were found to have vasospasm via cerebral angiography tended to experience peak NSE levels between days 5 and 15 since onset. Moreover, a correlation was observed between serum NSE level and hematoma size in those whose hematoma was 5 cm or greater [83]. Similarly, Oertel et al. found that SAH patients had higher levels of serum NSE within 3 days of onset if they had received Fisher CT scores of 4. However, unlike S100B, NSE was not found to reliably predict or be correlated with anything else, including vasospasm or GCS-determined outcome [84]. In terms of distinguishing between SAH and acute onset migraine, serum NSE levels were found to be significantly reduced in those with acute benign migraine in comparison to healthy controls, though they did not necessarily serve to indicate neurological injury [85].

3.2. Ubiquitin C-terminal hydrolase (UCH-L1)

A promising candidate biomarker for TBI currently under investigation is ubiquitin C-terminal hydrolase-L1 (UCH-L1). This protein is involved in the addition and removal of ubiquitin from proteins that are destined for metabolism [86] and therefore, has an important role in the removal of excessive, oxidized, or misfolded proteins in neurons [87]. UCH-L1 was previously used as a histological marker for neurons [88]. Clinical studies in humans with severe TBI have confirmed that the UCH-L1 protein is significantly elevated in human CSF and is detectable very early after injury [89, 90]. It remains significantly elevated for at least 1 week post injury [90] and there is very good correlation between CSF and serum levels [91]. Serum UCH-L1 is also elevated in children with moderate and severe TBI [92]. Most recently, UCH-L1 was detected in the serum of mild TBI patients within an hour of injury [47, 93]. Serum levels of UCH-L1 discriminated concussion patients from uninjured and non-head-injured trauma control patients who had orthopedic injuries or motor vehicle trauma without head injury [47, 93]. A handful of studies have shown serum UCH-L1 levels to be significantly higher in those with intracranial lesions on CT than those without lesions [45, 47, 93, 94] and to be much higher in those eventually requiring a neurosurgical intervention [47, 93]. The temporal profile of UCH-L1 was evaluated in a large cohort of 584 trauma patients seen at the emergency

department. UCH-L1 rose rapidly and peaked at 8 h after injury and declined rapidly over 48 h [47].

Despite UCH-L1's capacity to identify concussive TBI, it is perhaps best employed prognostically, like S100B, in the assessment of stroke. Because increased serum and CSF concentrations of UCH-L1 are symptomatic of blood-brain barrier disruption, the deubiquinating enzyme is particularly useful in assessing both brain damage severity (i.e., infarction volume and extent of vasospasm) and the resulting outcome in patients who have suffered hemorrhage or ischemia. Indeed, studies investigating the role of UCH-L1 in stroke have noted its potential to measure neurodegenerative injury. Individuals who suffered acute ischemic episodes within 12–24 h following an aortic aneurysm repair were found to have elevated CSF concentrations of UCH-L1 [95]. Despite the confounders of surgical distress and cardiopulmonary or circulatory complication, the study concluded that UCH-L1 levels were reliably associated with neurological damage. The 12–24 h timescale restricts the use of UCH-L1 to monitoring, even though ischemic apoptosis has been noted to peak at 24–48 h post onset [47–95]. UCH-L1 holds therapeutic promise for ischemia patients to the extent that it and similar deubiquinating enzymes have been found to reduce infarction in cases of rapid ischemic tolerance following brief ischemia [96]. Interestingly, polyubiquitinated protein buildups in hippocampal synapses have been reported in response to global ischemia [97], raising the question of how well such results might coordinate with MRI imaging data. The potential role of UCH-L1 in cases of hemorrhage is perhaps clearer. Lewis et al. found that individuals suffering from aneurysmal (SAH) had consistently higher concentrations of UCH-L1 in the CSF 2 weeks after post-aneurysmal rupture, which moreover were significantly associated with poor recovery [98]. Furthermore, patients in whom CSF S100B levels reduced experienced improved recovery when UCH-L1 concentrations dropped as well. Siman et al. similarly found that CSF concentrations of UCH-L1, among an array of seven other biomarkers including NSE and S100B, taken over a 10-day period since aneurysmal rupture, rose significantly and predicted severity of infarction, vasospasm, and outcome [95]. However, they found mixed results for whether UCH-L1 levels peaked on the first day, the seventh day, or remained relatively consistent throughout the measured time course.

4. Biomarkers of axonal injury

4.1. Alpha-II spectrin breakdown products

Alpha-II-spectrin is a 280-kDa protein that is an important structural component of the cortical membrane cytoskeleton, particularly abundant in axons and presynaptic terminals [99, 100]. It serves as a key substrate for both calpain-2 and caspase-3 cysteine proteases [101, 102]. These proteases (caspase-3 and calpain-2) cleave cytoskeletal α II-spectrin [103, 104] into spectrin breakdown products (SBDPs). These SBDPs have been reported in CSF from adults with severe TBI and they have shown a significant relationship with severity of injury and clinical outcome [105–112]. The time course of calpain-mediated SBDP150 and SBDP145 (markers of necrosis) differs from that of caspase-3-mediated SBDP120 (marker of apoptosis)

and have been shown to correlate with severity of injury, CT scan findings, and outcome at 6 months post injury [111, 112]. These findings were similar in children with moderate to severe TBI [92]. More recently, serum levels of SBDP150 measured in mild TBI patients have shown a significant association with acute measures of injury severity, such as GCS score, intracranial injuries on CT, and neurosurgical intervention [113]. In this study, serum SBDP150 levels were much higher in patients with concussion than other trauma patients who did not have a head injury [113].

In patients suffering from aneurysmal subarachnoid hemorrhage, CSF concentrations of calpain-mediated and caspase-mediated SBDPs have been found to be significantly elevated within 72 h post onset and up to 12 h pre-cerebral arterial vasospasm due to the role of necrotic proteolysis in hemorrhage and vasospasm-induced neurodegeneration [114, 115]. Research on the potential role of SBDPs in human ischemia has been comparatively more sparse, though rat studies have indicated an association between caspase-mediated spectrin breakdown products and ischemia-induced apoptosis [116, 117], and the overstimulation of mammalian calpain 1 and calpain 2 has been understood to be involved with the pathophysiology of acute stroke [118]. Moreover, alpha-spectrin-related insights into treatment potential for both cerebral ischemia and TBI have surfaced in the form of caspase cascade inhibitors, which have been able to arrest processes of apoptosis in the aftermath of the aforementioned acute neurological disorders [119, 120]. Biomarker panel including assays of caspase-3 and D-dimer has potential in delineating stroke from ischemic stroke mimics, such as acute migraine [121].

4.2. Tau protein

Tau is an intracellular, microtubule-associated protein that is highly enriched in axons and is involved with assembling axonal microtubule bundles and participating in anterograde axoplasmic transport [122]. Tau lesions are apparently related to axonal disruption such as in trauma or hypoxia [123, 124]. After release, it is proteolytically cleaved at the N- and C-terminals. The C-tau has been investigated as a potential biomarker of CNS injury.

Initial elevated CSF C-tau levels in severe TBI patients have been shown to predict elevations in intracranial pressure and to be associated with poor clinical outcome [125]. In a study by Shaw et al., an elevated level of C-tau was associated with a poor outcome at hospital discharge and with an increased chance of an intracranial injury on head CT [126]. However, these findings were not reproducible when C-tau was measured in peripheral blood in mild TBI [127]. Two additional studies showed that C-tau was a poor predictor of CT lesions and a poor predictor of post-concussive syndrome [15, 128]. Total tau (T-tau) has also been found to be correlated with severity of injury in severe TBI [129–132]. Ost et al. found that tau measured in CSF on days 2 to 3 discriminated between TBI and controls with (normal pressure hydrocephalus) and also between good and bad outcome at 1 year per dichotomized Glasgow Outcome Scale (GOS) score [131]. Unfortunately, T-tau was not detected in serum throughout the study. Phosphorylated -tau (P-Tau) is also being examined following head trauma [133].

The hyperphosphorylation of tau in the development of apoptosis-related neurofibrillary tangles has been explored in relation to neurodegeneration induced by transient cerebral ischemia [134]. Dewar et al. suggested the role of cytoskeletal breakdown in both cerebral focal

ischemia and Alzheimer's-induced impairment, having found that permanent focal cerebral ischemia resulted in modification of the protein tau [135]. Results have been more prolific for hemorrhage, perhaps due to the more immediate severity and thus morphological damage it tends to entail. In accordance with the aforementioned time scale for TBI, Hu et al. observed that serum tau levels in patients with intracerebral hemorrhage were significantly predictive of 3-month mortality, with these prognoses achieving greater predictive accuracy than the NIHSS [136]. Augmented CSF levels of tau were reported in patients following severe episodes of aneurysmal subarachnoid hemorrhage in comparison with more moderate episodes, also correlating with the motor score on the GCS and proving more elevated in those patients with fatal outcomes [137].

4.3. Neurofilaments

Neurofilaments are heteropolymeric components of the neuron cytoskeleton that consist of a 68-kDa light neurofilament subunit (NF-L) backbone with either 160 kDa medium (NF-M) or 200 kDa heavy subunit (NF-H) side arms [138]. They provide structural support for the axon. It is postulated that after a TBI, calcineurin (a calcium-dependent phosphatase) dephosphorylates neurofilament side arms, and contributes to axonal injury [139]. Phosphorylated NF-H in CSF has been found to be elevated in adult patients with severe TBI compared to controls [89]. Hyperphosphorylated NF-H has also been correlated with severity of brain injury in children [140]. In this study, NF-H levels taken on the second to fourth day remained significantly higher in patients with poor outcome in comparison to patients with good outcome and in those children with DAI on initial CT scan [140]. Vajtr et al. also found elevated serum NF-H in patients with DAI over 10 days after admission with highest levels from day 4 to 10 [141].

Serum concentrations of phosphorylated NFL-H (pNFL-H) sampled from patients with acute ischemia have been shown to correlate with CT scan assessment of ischemia upon admission and at 7 days post onset as determined by the Alberta Stroke Program Early CT Scale, NIHSS, and GCS [142]. Sellner et al. also found significantly higher serum pNFL-H concentrations in ischemic patients 24 h after onset [143]. However, exploration of the timely diagnostic value of pNFL-H for ischemia has again been mixed. While Singh et al. observed elevation of serum pNFL-H in ischemic patients, levels did not reach significance or predict patient outcome or infarct volume until 3 weeks post onset [144]. In cases of hemorrhagic stroke, neurofilaments show promise as prognostic markers, with elevated CSF levels assayed within 10–14 days of aneurysmal SAH onset and correlating with GCS performance as 1 year post onset [145]. Assays of pNFL-H were also found to correlate with early neurological deterioration and survival rates 6 months post onset with accuracy comparable to that of the NIHSS [146].

5. Conclusion

There is a great need to validate brain injury biofluid biomarkers in the acute care setting such as in the emergency department. Biomarkers measured through a simple blood test have the potential to provide invaluable information about the management of acute brain injury for

conditions such as TBI and stroke. Biomarkers could potentially facilitate diagnosis and risk stratification of these patients. Biomarkers could provide timely information about the pathophysiology of injury to allow for monitoring and assessment of progression and recovery. Biomarkers could provide major opportunities for drug target identification and guide the conduct of clinical research as surrogate outcome measures. Although research in the field of brain injury biomarkers has increased significantly over the last decade, clinical studies have not been adequately powered with enough patients to validate them. Large clinical studies are underway that will change this and will bring a blood test closer to the bedside.

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