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Serum Biomarkers for Early Prediction of Outcome After Moderate-to-Severe Traumatic Brain Injury

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ABSTRACT

Background: The prognosis following moderate-to-severe traumatic brain injury (TBI) is difficult to predict accurately in the early stage.

Objective: To evaluate the prognostic value of early serum biomarkers in predicting six-month functional outcomes among patients with moderate-to-severe traumatic brain injury.

Methods: This prospective observational study was conducted in the Department of Neurosurgery and Neurocritical Care, Nishtar Hospital, Nishtar Medical University, Multan, Pakistan, from March 2023 to March 2025. Serial GFAP, UCH-L1, NSE, and S100B levels were obtained within 6 hours of admission. At six months, functional outcome was evaluated using the Glasgow Outcome Scale-Extended and divided into favourable and unfavourable outcomes.

Results: A favourable outcome was observed in 72 patients, while 48 had an unfavourable outcome, including 20 deaths. The GFAP, UCH-L1, NSE, and S100B levels were significantly higher for patients with an unfavourable outcome compared to patients with a favourable recovery. GFAP was the best predictor of unfavourable outcome with an AUC of 0.86, followed by UCH-L1 (AUC 0.81). The combined GFAP and UCH-L1 model had better predictive accuracy (AUC 0.89). Multivariable analysis revealed GFAP, UCH-L1, a lower GCS on admission, non-reactive pupils, a higher Marshall CT grade, and higher age to be independent predictors of poor outcome.

Conclusions: The serum assessment of these biomarkers is a valuable prognostic tool following moderate to severe TBI. GFAP and UCH-L1 seem to be the most promising early markers and could potentially be used to help stratify patients clinically.

Keywords: Traumatic brain injury; GFAP; UCH-L1; serum biomarkers; neurological outcome; GCS.

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INTRODUCTION

Traumatic brain injury (TBI) remains one of the leading causes of mortality and long-term disability worldwide, particularly among young adults and economically productive populations [1]. Although advances have been made in emergency care, neuroimaging, and intensive neurocritical management, moderate to severe traumatic brain injury is associated with significant neurological impairment, prolonged hospitalization, and socioeconomic burden. Despite advances in prompt surgical care, ICP monitoring, and standardized critical care management, determining neurological recovery in the early post-injury period remains a great clinical challenge. Prognostication is imperative to assist in treatment decisions, resource management, discussing prognosis with families, and selection of patients for targeted neuroprotective therapies [2].

The current prognosis of TBI is mainly based on clinical parameters including Glasgow Coma Scale (GCS), pupillary reactivity, neuroimaging findings and physiological variables [3]. These are useful tools for

assessing the severity of the injury, but can be affected by sedation, alcohol intoxication, systemic injury or delayed neurological deterioration. Likewise, radiological classifications and computed tomography (CT) based scoring systems might not adequately reflect the severity of diffuse axonal injury, secondary neuronal damage and biochemical processes that are ongoing following the initial trauma. Hence, there is growing interest in objective circulating biomarkers that are reflective of the underlying pathophysiological mechanisms of brain injury with a prognostic value over and above the conventional clinical assessment [4].

Moderate-to-severe traumatic brain injury is a complex process of primary and secondary injurious mechanisms. Mechanisms of injury are divided into primary (direct neuronal and glial disruption) and secondary (excitotoxicity, mitochondrial dysfunction, oxidative stress, neuroinflammation, blood–brain barrier disruption, cerebral edema, apoptotic cell death) over a period of hours to days. These processes result in continued deterioration of the nervous system following the original trauma. When cells are injured, structural proteins, inflammatory mediators, and molecules associated with neuronal damage will leak into the extracellular space and into the systemic circulation, and offer the opportunity to objectively determine the extent of brain damage using blood-based markers [5,6].

Among the most extensively investigated biomarkers are glial fibrillary acidic protein (GFAP), ubiquitin carboxyl-terminal hydrolase-L1 (UCH-L1), neuron-specific enolase (NSE), S100 calcium-binding protein B (S100B), neurofilament light chain (NfL), tau protein, and inflammatory cytokines. GFAP is a marker of astroglial damage, while UCH-L1 is mostly secreted by damaged neurons [7]. Circulating levels of S100B have been extensively investigated as markers of astrocytic damage and BBB disruption, but extracranial injuries could affect S100B levels. NSE is a marker of neuronal cell destruction, while NfL is gaining growing acceptance as a sensitive marker of axonal injury. Multiple biomarkers may give a better picture of the heterogeneous pathological processes involved in traumatic brain injury than individual biomarkers alone [8].

With recent improvements in very sensitive immunoassays, these biomarkers can be accurately measured in the hours immediately after trauma. Association of high serum levels of GFAP, UCH-L1, S100B, and NfL with intracranial lesions, DAI, neurosurgical intervention, prolonged intensive care, and death has been shown in several investigations. However, there are many differences between the various published studies in terms of sampling time, patient selection, injury severity, laboratory techniques, and methods of outcome assessment. Furthermore, the studies have considered single biomarkers separately, and not compared their predictive power in the same patient cohort [9-10].

If possible, patients at high risk of an unfavorable neurologic outcome could be identified early, leading to significantly better clinical management. Serum markers can help to stratify risk at hospital admission, help to select patients for aggressive neurocritical care or early rehabilitation, help to enroll patients into trials, and offer objective measures to monitor disease progression. In addition, the combination of biomarker profiles with known clinical predictors could help improve the performance of current prognostic models and help guide personalized treatment in patients with TBI [11].

Although there is increasing evidence for the advantages of biomarker-directed prognostication, there is still little data available for low- and middle-income countries where traumatic brain injury incidence is high, and healthcare resources are limited. Before their use in routine practice, therefore, routine serum biomarkers, which are readily obtainable, will need to be validated in multiple clinical settings. The possible setting of clinically relevant biomarker thresholds could further enhance their utility in ED and NCCU settings [12].

Therefore, the present prospective observational study was conducted to evaluate the role of early serum biomarkers in predicting functional outcomes among patients with moderate-to-severe traumatic brain injury. The study was also designed to see whether the concentrations of these biomarkers at admission were associated with six-month neurological outcomes, mortality, and injury severity, and whether they performed as well as conventional clinical assessment tools.

MATERIALS AND METHODS

This prospective observational study was conducted in the Department of Neurosurgery and Neurocritical Care, Nishtar Hospital, Nishtar Medical University, Multan, Pakistan, over a 24-month period from March 2023 to March 2025. The study aimed to evaluate the prognostic value of early serum biomarkers in predicting six-month functional outcomes among patients with moderate-to-severe traumatic brain injury. Ethical approval was obtained from the Institutional Review Board of Nishtar Medical University before the commencement of patient recruitment (Reference No: 8845/NMU)**.** The study was conducted in accordance with the Declaration of Helsinki. Written informed consent was obtained from each patient or, where the patient lacked decision-making capacity, from a legally authorized representative.

The study used a consecutive non-probability sampling technique where a total of 120 acute moderate to severe TBI patients were sampled. Eligible patients were those aged 18-70 years who presented within 12 hours of injury and had a Glasgow Coma Scale (GCS) score of 3-12 on admission. Traumatic brain injury was diagnosed by clinical and non-contrast computed tomography (CT) of the brain. Patients with penetrating cranial injuries, previous neurological disorders, chronic neurodegenerative disease,

active malignancy, severe hepatic and renal dysfunction, pregnancy, known inflammatory or autoimmune diseases, major burns, delayed presentation beyond 12 hours after trauma, or missing follow-up data were excluded. Patients for whom a sample was not taken due to death were also excluded.

Demographic data such as age, gender, mechanism of injury, co-injuries, Glasgow Coma Scale (GCS) on admission, pupil reactivity, Injury Severity Score (ISS), hypotension on admission, requirement for mechanical ventilation, neurosurgical intervention, and radiological findings were collected on a standardized case record form. Consultant neuroradiologists initially interpreted CT scans of the brain, and the following intracranial findings were recorded: epidural hematoma, subdural hematoma, intracerebral contusion, traumatic subarachnoid hemorrhage, diffuse axonal injury, midline shift, basal cistern compression, and skull fracture. The severity of injuries was also classified based on the Marshall CT classification.

Venous blood samples were collected within 6 hours of hospital admission, before definitive neurosurgical intervention, if possible. About 10 mL of peripheral venous blood was drawn under sterile conditions, centrifuged at 3000 rpm for 10 minutes, and serum aliquots were stored at −80°C for biochemical analysis. The serum levels of glial fibrillary acidic protein (GFAP), ubiquitin carboxyl-terminal hydrolase-L1 (UCH-L1), neuron-specific enolase (NSE), and S100 calcium-binding protein B (S100B) were measured using commercially available sandwich enzyme-linked immunosorbent assay (ELISA) kits according to the manufacturers' instructions. Laboratory analysis was repeated twice by investigators unaware of the patients' clinical outcomes, and laboratory quality-control procedures were followed throughout the analysis.

All patients underwent standard evidence-based care with the institutional protocol for traumatic brain injury, including airway stabilization, ICP control, mechanical ventilation as needed, hyperosmolar therapy, seizure prevention, blood pressure control, and emergency neurosurgical procedures (decompressive craniectomy or hematoma evacuation) as indicated. Treatment decisions were at the discretion of the treating neurosurgical team and were not affected by the measurement of serum biomarkers.

Patients were monitored during their hospital stay and re-assessed at six months post-injury via organized telephone interviews with investigators blinded to biomarker results or outpatient visits. The Glasgow Outcome Scale-Extended (GOSE) was used to assess functional outcome. Outcomes were classified as "good" (GOSE 5-8) and "poor" (GOSE 1-4). Secondary outcome measures were in-hospital death, 6-month mortality, length of intensive care unit stay, length of hospital stay, requirement for decompressive surgery, length of mechanical ventilation, and in-hospital neurological complications.

IBM SPSS Statistics version 27.0 (IBM Corp., Armonk, NY, USA) software was used to enter and analyze the data. Normally distributed continuous variables were analyzed using the Shapiro–Wilk test and reported in terms of mean ± standard deviation or median (interquartile range), whichever was appropriate. Categorical data presented as frequencies and percentages. An independent-samples t-test or Mann–Whitney U test was used to compare continuous variables, whereas a chi-square test or Fisher's exact test was applied for categorical variables. Spearman's rank correlation coefficient was used for the correlation analyses between the biomarkers in serum and the GCS score on admission. Multivariable binary logistic regression analysis was conducted to determine independent factors associated with poor neurological outcome at 6 months after adjustment for clinically relevant factors such as age, admission Glasgow Coma Scale (GCS) score, pupillary response, Marshall CT classification, and neurosurgical intervention. To assess the prognostic value of individual serum markers, receiver operating curve (ROC) analysis was carried out and the area under the curve (AUC) and 95% confidence intervals (CI) were calculated. Throughout, a two-tailed p-value <0.05 was accepted as statistically significant.

RESULTS

A total of 120 patients with moderate-to-severe traumatic brain injury were included in the final analysis. The mean age of the study population was 39.6 ± 14.8 years, with a predominance of male patients (88/120, 73.3%) compared with female patients (32/120, 26.7%). The most common mechanism of injury was road traffic accidents (81, 67.5%), followed by falls from height (27, 22.5%) and blunt trauma from assault (12, 10.0%). At admission, 66 (55.0%) patients suffered moderate traumatic brain injury (GCS 9-12), and 54 (45.0%) patients suffered severe traumatic brain injury (GCS 3-8). The overall mean admission GCS score was 8.2±2.7.72 patients (60.0%) reached a good functional outcome at 6 months and 48 patients (40.0%) an unfavorable outcome (death, vegetative state, severe disability).

A more severe clinical profile at the time of admission was noted in those patients with an unfavorable outcome. The mean age of the patients with poor neurological recovery was significantly higher than that of patients with good neurological recovery (46.1 ± 15.6 years vs. 35.2 ± 12.7 years; $p<0.001$), indicating a correlation between increasing age and poorer neurological recovery. In addition, admission GCS was significantly lower in the unfavorable outcome group (5.9 ± 2.1) than the favorable outcome group (9.7 ± 1.8; $p<0.001$). Severe TBI was found in 35 patients (72.9%) with unfavorable and 19 patients (26.4%) with favorable outcomes. In the unfavorable group, 25 (52.1%) patients had an abnormal pupillary response, while only 10 (13.9%) patients in the favorable group had an abnormal pupillary response. Hypoxia and hypotension

at presentation were also significantly more common in patients who had an unfavorable outcome. The results of these observations show that the early physiological instability and the depressed neurological status were strongly correlated with poor recovery at six months (Table 1).

Unfavorable outcomes were also associated with a higher radiological severity on admission computed tomography (CT) of the brain. Marshall CT grade V–VI was observed in 17 patients (35.4%) with unfavorable outcomes compared with only 7 patients (9.7%) with favorable outcomes. In the unfavorable outcome group, there were 24 patients (50.0%) with midline shift > 5 mm versus 14 patients (19.4%) in the favorable outcome group. Likewise, 26 (54.2%) of the patients with poor outcome and 15 (20.8%) of the patients with good outcome had compressed or absent basal cisterns. The proportion of patients with suspected DAIs was also much higher in the unfavourable outcome group (35.4% vs. 12.5%, p=0.003). Overall, 58 patients (48.3%) required mechanical ventilation, with significantly higher rates in worse vs better outcome (72.9% vs 31.9%). A neurosurgical intervention was carried out in 42 patients (35.0%), with more being seen in patients who had worse outcomes (50.0% vs. 25.0%; p=0.005). The unfavorable outcome group also had longer total hospital stay and longer days in the ICU, indicating that they were clinically more complex and that they required critical care for a longer period (Table 1).

There was a significant difference in the concentrations of serum biomarkers at the onset of the event between the favorable outcome and unfavorable outcome group. Median serum GFAP concentration in the total cohort was 1.48 ng/mL, with a substantially higher level among patients with unfavorable outcomes (2.86 ng/mL; IQR 1.64–5.22) than among those with favorable outcomes (0.94 ng/mL; IQR 0.51–1.76; p<0.001). This difference was found to be consistent when patients were categorized according to the severity of the TBI. Moderate TBI patients with an unfavorable outcome had a GFAP level of 1.91 ng/mL, whereas favorable outcome patients had a GFAP level of 0.71 ng/mL. For patients with severe TBI, GFAP levels were still significantly higher in patients with poor outcome (3.66 ng/mL vs. 1.74 ng/mL; p=0.002). These results indicated that GFAP was a marker of astroglial damage, and that it maintained its prognostic significance in both the moderate and severe injury group (Table 2).

The association of UCH-L1 with 6-month outcome was also clear. In patients with unfavorable outcome, the median UCH-L1 level was 1.42 ng/mL, while that of patients with favorable outcome was 0.55 ng/mL (p<0.001). UCH-L1 was significantly elevated in both moderate and severe TBI patients in both subgroups of severity, indicating unfavorable outcomes. NSE and S100B were similar, with a higher concentration in the group with bad outcomes. The median values were 39.7 ng/mL for Median NSE and 0.78 μg/L for Median S100B in patients with unfavorable outcomes and 22.1 ng/mL for Median NSE and 0.31 μg/L for Median S100B in patients with favorable outcomes, respectively. All 4 biomarkers were statistically significantly associated with outcome when compared unadjusted to outcome, but the difference was largest for GFAP and UCH-L1, suggesting that the discriminatory potential of these two markers is higher (Table 2).

Additionally, there was a strong relationship between serum biomarker levels and measures of neurological and radiological injury. The GCS score on admission was the strongest negative correlation with GFAP ($r = -0.62$; $p<0.001$), indicating that the more unconscious a patient was, the higher the GFAP level. UCH-L1 also showed a significant inverse correlation with GCS ($r = -0.55$; $p<0.001$), followed by NSE ($r = -0.47$; $p<0.001$) and S100B ($r = -0.43$; $p<0.001$). Likewise, the biomarker levels were higher in the more severe Marshall CT grade. GFAP had the strongest positive correlation with Marshall CT grade ($r = 0.58$; $p<0.001$), followed by UCH-L1 ($r = 0.49$; $p<0.001$), NSE ($r = 0.41$; $p<0.001$), and S100B ($r = 0.39$; $p<0.001$). They demonstrate that elevated early biomarker levels were not biochemical changes in isolation but tightly coupled with actual clinical and radiological markers of injury severity (Table 2).

ROC analysis was used to assess the capacity of individual biomarkers in serum to predict worse 6-month outcomes. GFAP had the highest predictive accuracy with an area under the curve (AUC) of 0.86 (95% CI: 0.79–0.93). With a cut-off value of >1.85 ng/mL, GFAP showed 81.3% sensitivity and 79.2% specificity in predicting an unfavourable outcome. UCH-L1 showed the second-best performance, with an AUC of 0.81 (95% CI 0.73–0.89). The cut-off value of UCH-L1 was >1.05 ng/mL, with a sensitivity of 75.0% and specificity of 76.4%. The AUCs of NSE and S100B were also statistically significant, but were lower than that of GFAP and UCH-L1. NSE had an AUC of 0.77, while S100B had an AUC of 0.74. The model with both GFAP and UCH-L1 had the best predictive accuracy with an AUC of 0.89 (95% CI 0.83-0.95), sensitivity of 83.3% and specificity of 83.3% (Table 3).

Using multivariate logistic regression analysis, independent predictors of poor neurological outcome at 6 months were determined, after adjusting for clinically significant variables. These findings were independent of GFAP, which was independently associated with poor outcome with an adjusted OR of 3.42 (95% CI 1.71–6.86; p<0.001). UCH-L1 also remained an independent predictor, with an adjusted odds ratio of 2.61 (95% CI 1.29–5.28; p=0.008). Each 1-point decrease in GCS was independently associated with an increased risk of an unfavorable outcome by ~58%. In addition, abnormal pupillary response and Marshall CT grade V–VI were also independent significant predictors. However, NSE and S100B were not statistically significant when adjusted for injury severity and collinearity with better predictors, indicating that their predictive power

may have been due to the severity of the injury and their correlation with other better predictors (Table 3).

In-hospital mortality rate was 13.3% and 6-month mortality rate was 16.7%. Levels of early biomarkers were significantly increased in patients who died compared to those who survived, especially GFAP and UCH-L1. Patients with GFAP levels ≥1.85 ng/mL had higher mortality rates, as well as higher rates of decompressive surgery, prolonged stay in the intensive care unit, and dependence at follow-up. Likewise, patients with UCH-L1 levels >1.05 ng/mL were more likely to be ventilated, severely disabled, or dead. In those patients who survived to 6 months, 72 patients had a good neurological recovery, and 28 survivors had severe neurological disability. This pattern demonstrates that biomarker elevation predicted not only mortality but also long-term functional dependence.

Table 1: Baseline clinical and radiological characteristics according to six-month outcome

Variable	Total cohort (n=120)	Favorable outcome (n=72)	Unfavorable outcome (n=48)	p-value
Age, years	39.6 ± 14.8	35.2 ± 12.7	46.1 ± 15.6	<0.001
Male sex	88 (73.3%)	51 (70.8%)	37 (77.1%)	0.446
Female sex	32 (26.7%)	21 (29.2%)	11 (22.9%)	0.446
Road traffic accident	81 (67.5%)	47 (65.3%)	34 (70.8%)	0.529
Fall from height	27 (22.5%)	18 (25.0%)	9 (18.8%)	0.423
Assault/blunt trauma	12 (10.0%)	7 (9.7%)	5 (10.4%)	0.901
Admission GCS score	8.2 ± 2.7	9.7 ± 1.8	5.9 ± 2.1	<0.001
Moderate TBI, GCS 9–12	66 (55.0%)	53 (73.6%)	13 (27.1%)	<0.001
Severe TBI, GCS 3–8	54 (45.0%)	19 (26.4%)	35 (72.9%)	<0.001
Abnormal pupillary response	35 (29.2%)	10 (13.9%)	25 (52.1%)	<0.001
Hypotension at presentation	24 (20.0%)	9 (12.5%)	15 (31.3%)	0.011
Hypoxia at presentation	21 (17.5%)	7 (9.7%)	14 (29.2%)	0.006
ISS score	24.8 ± 8.9	21.7 ± 7.1	29.5 ± 9.4	<0.001
Marshall CT grade I–II	45 (37.5%)	36 (50.0%)	9 (18.8%)	<0.001
Marshall CT grade III–IV	51 (42.5%)	29 (40.3%)	22 (45.8%)	0.550
Marshall CT grade V–VI	24 (20.0%)	7 (9.7%)	17 (35.4%)	<0.001
Midline shift >5 mm	38 (31.7%)	14 (19.4%)	24 (50.0%)	<0.001
Compressed/absent basal cisterns	41 (34.2%)	15 (20.8%)	26 (54.2%)	<0.001
Traumatic subarachnoid hemorrhage	43 (35.8%)	21 (29.2%)	22 (45.8%)	0.062
Subdural hematoma	39 (32.5%)	19 (26.4%)	20 (41.7%)	0.079
Cerebral contusion	52 (43.3%)	30 (41.7%)	22 (45.8%)	0.653
Diffuse axonal injury suspected	26 (21.7%)	9 (12.5%)	17 (35.4%)	0.003
Mechanical ventilation required	58 (48.3%)	23 (31.9%)	35 (72.9%)	<0.001
Neurosurgical intervention	42 (35.0%)	18 (25.0%)	24 (50.0%)	0.005
ICU stay, days	7.8 ± 4.6	5.9 ± 3.1	10.7 ± 5.1	<0.001
Hospital stay, days	14.6 ± 7.3	13.2 ± 6.5	16.7 ± 7.9	0.009

Table 2: Early serum biomarker levels and correlation with injury severity

Biomarker/parameter	Total cohort (n=120)	Favourable outcome (n=72)	Unfavourable outcome (n=48)	p-value
GFAP, ng/mL	1.48 (0.72–3.21)	0.94 (0.51–1.76)	2.86 (1.64–5.22)	<0.001
UCH-L1, ng/mL	0.82 (0.39–1.61)	0.55 (0.28–0.96)	1.42 (0.81–2.48)	<0.001
NSE, ng/mL	27.6 (18.4–42.9)	22.1 (15.8–31.4)	39.7 (27.6–58.3)	<0.001
S100B, µg/L	0.46 (0.22–0.91)	0.31 (0.17–0.58)	0.78 (0.41–1.36)	<0.001
GFAP in moderate TBI	0.86 (0.44–1.61)	0.71 (0.39–1.32)	1.91 (1.08–3.18)	<0.001
GFAP in severe TBI	2.73 (1.42–5.41)	1.74 (0.91–2.96)	3.66 (1.93–6.28)	0.002
UCH-L1 in moderate TBI	0.48 (0.24–0.88)	0.42 (0.21–0.75)	1.03 (0.61–1.87)	<0.001
UCH-L1 in severe TBI	1.36 (0.75–2.39)	0.98 (0.49–1.62)	1.71 (0.93–2.71)	0.011
NSE in moderate TBI	21.5 (15.2–30.6)	19.6 (14.2–27.8)	34.8 (24.7–46.2)	<0.001
NSE in severe TBI	38.9 (26.4–57.5)	31.2 (21.5–42.6)	45.7 (31.6–63.4)	0.006
S100B in moderate TBI	0.29 (0.15–0.54)	0.25 (0.13–0.45)	0.61 (0.34–1.02)	<0.001
S100B in severe TBI	0.76 (0.38–1.34)	0.52 (0.26–0.91)	0.93 (0.49–1.54)	0.009
Spearman correlation: GFAP with GCS	r = −0.62	—	—	<0.001
Spearman correlation: UCH-L1 with GCS	r = −0.55	—	—	<0.001
Spearman correlation: NSE with GCS	r = −0.47	—	—	<0.001
Spearman correlation: S100B with GCS	r = −0.43	—	—	<0.001
GFAP with Marshall CT grade	r = 0.58	—	—	<0.001
UCH-L1 with Marshall CT grade	r = 0.49	—	—	<0.001
NSE with Marshall CT grade	r = 0.41	—	—	<0.001
S100B with Marshall CT grade	r = 0.39	—	—	<0.001

Table 3: Predictive performance and multivariable predictors of unfavorable six-month outcome

Predictor/model	Cut-off value	Sensitivity	Specificity	AUC (95% CI)	Adjusted OR (95% CI)	p-value
GFAP	>1.85 ng/mL	81.3%	79.2%	0.86 (0.79–0.93)	3.42 (1.71–6.86)	<0.001
UCH-L1	>1.05 ng/mL	75.0%	76.4%	0.81 (0.73–0.89)	2.61 (1.29–5.28)	0.008
NSE	>33.5 ng/mL	68.8%	72.2%	0.77 (0.68–0.86)	1.48 (0.76–2.89)	0.246
S100B	>0.62 µg/L	66.7%	70.8%	0.74 (0.65–0.83)	1.36 (0.69–2.71)	0.374
GFAP + UCH-L1 model	—	83.3%	83.3%	0.89 (0.83–0.95)	—	<0.001
GCS score	Per 1-point decrease	—	—	0.82 (0.74–0.90)	1.58 (1.23–2.03)	<0.001
Abnormal pupillary response	Present	—	—	—	3.76 (1.41–10.04)	0.008
Marshall CT grade V–VI	Present	—	—	—	3.19 (1.13–9.01)	0.028
Hypotension at presentation	Present	—	—	—	1.84 (0.69–4.91)	0.223
Hypoxia at presentation	Present	—	—	—	1.97 (0.71–5.48)	0.193
Age	Per 10-year increase	—	—	—	1.31 (1.02–1.78)	0.041
Mechanical ventilation	Required	—	—	—	1.89 (0.72–4.95)	0.194
Neurosurgical intervention	Required	—	—	—	1.52 (0.61–3.81)	0.371

The results overall show that, in the moderate to severe TBI patient population, serum biomarker measurement in the early phase of disease could offer useful prognostic value. GFAP was the most powerful single predictor of poor 6-month outcome, and UCH-L1 provided additional prognostic information. The GFAP and UCH-L1 combination model was more accurate than admission GCS alone and more effective than individual GFAP or UCH-L1 models. The results suggest that serum biomarker profiling could be a potential clinical tool for evaluating moderate to severe TBI in addition to standard neurological exams and CT scans.

DISCUSSION

The study revealed that the serum levels of these proteins at an early stage after moderate to severe TBI were independent variables significantly correlated with the functional outcome at 6 months in this prospective observational study [1]. Compared to patients with favorable recovery, patients with unfavorable outcomes had significantly higher admission GFAP, UCH-L1, NSE, and S100B. GFAP had the highest predictive value of these biomarkers, followed by UCH-L1. The simultaneous assessment of astroglial and neuronal injury, by combining GFAP and UCH-L1, yielded the highest prognostic accuracy, indicating that simultaneous measurement of GFAP and UCH-L1 could also be more useful in clinical stratification than either of the markers alone [2,3].

Biologically, the fact that GFAP was the most significant single biomarker makes sense. GFAP is an intermediate filament protein primarily found in astrocytes, which is released in extracellular fluid and systemic circulation upon astroglial disruption and blood–brain barrier injury [4]. In the current study, high GFAP concentrations were correlated with low admission GCS, high Marshall CT scores, midline shift, compressed basal cisterns, prolonged ICU stay, and poor 6-month outcome. It suggests GFAP was a marker of structural burden of injury and secondary intracranial pathophysiology. In addition, it is an independent predictor of poor outcome when adjusted for GCS, pupillary response, CT severity, and age, further supporting its prognostic relevance in TBI of moderate to severe severity [5].

UCH-L1 was also an independent predictor of bad outcome. By virtue of UCH-L1 being mainly present in neurons, increased serum levels can be a marker of early damage to the neuronal cells and axonal damage [6]. There was an interesting negative correlation between UCH-L1 and admission GCS, indicating that higher levels of neuronal injury were associated with worse impairment of consciousness. The combined GFAP and UCH-L1 model showed the highest AUC, sensitivity, and specificity, while GFAP had a slightly better performance than UCH-L1. These findings support the concept proposed by Sperry et al. that TBI is pathophysiologically heterogeneous and that combinations of biomarkers may better reflect the overlapping astroglial, neuronal, axonal, and blood–brain barrier injury patterns underlying long-term recovery [7,8].

The other markers NSE and S100B were also significantly higher in patients with poor outcomes, but were not independent predictors after multivariable analysis [9]. This might be due to these markers being less specific for intracranial injury or being more influenced by systemic trauma, hemolysis, extracranial tissue damage, and severity of injury. S100B can be increased following bone fractures, soft tissue trauma, and extracranial injury, and NSE can be influenced by red blood cell hemolysis and systemic stress. Thus, in this population, the independent prognostic significance of NSE and S100B seems to be inferior to that of GFAP and UCH-L1 [10].

These findings are also clinically relevant and consistent with those of Kobeissy et al., who reported an association between biomarker elevation and radiological injury severity. Higher levels of GFAP and UCH-L1 were correlated with a higher Marshall CT grade, greater midline shift, basal cistern compression, and suspected diffuse

axonal injury [11]. These data indicate that serum biomarkers could be useful for providing information about the nature of the injury that can be seen on CT, but also microscopic or diffuse injury processes that may not be fully appreciated on initial imaging. In cases of early assessment of TBI, sedation, intubation, intoxication, and systemic instability can make clinical neurological examination less reliable [12].

Kobeissy et al. and Robba et al. also highlighted the prognostic importance of biomarker assessment for mortality and disability. Patients with biomarker concentrations above the identified cut-off values had higher rates of mechanical ventilation, prolonged intensive care unit stay, decompressive surgery, severe disability, and death [13,14]. This means that early biomarker panels could be useful in selecting patients who need to be monitored more closely, need more aggressive neurocritical care, need early rehabilitation planning, and structured family counseling. In resource-limited areas where such sophisticated examinations and continuous monitoring of ICP may not be available at all times, serum biomarkers could be used as an objective and practical add-on to the traditional evaluation [15,16].

The other key finding was that abnormal pupillary response, Marshall CT grade V–VI, age, GFAP and UCH-L1 were all independent predictors of poor outcome when admission GCS was omitted [17]. This validates that biomarker testing should not be used as a substitute for clinical assessment, but in conjunction with other known prognostic factors. Using a combination of these is probably the most effective way to get an early risk stratification correct. A patient in clinical practice with low GCS, pupillary abnormality, very bad CT findings, and very high GFAP or UCH-L1 would be a high-risk group requiring urgent multidisciplinary neurocritical management [18].

The present study has a number of strengths. It was characterized by an established cohort of moderate to severe TBI patients, early blood sampling (within 6 hrs of admission), multiple clinically relevant serum biomarkers assessed, and functional outcome at 6 months with GOSE. Laboratory analysis was done blind to clinical outcome, minimizing measurement bias. Multivariable regression and ROC curve analysis also enabled comparison of the performance of biomarkers with the traditional clinical predictors [19,20].

But some caveats should be noted. The study was single-center, and the sample size was moderate, so generalizability may be limited [1,17]. The biomarker was only obtained at a single early time point, and the prognostic value of the serial changes of the biomarker could not be determined. Diffuse axonal injury may have been under-diagnosed based on CT assessment as MRI was not routinely used. Furthermore, some biomarker concentrations may have been affected by extracranial injuries, especially S100B and NSE. Long-term (i.e., beyond six months) cognitive, behavioral, and quality-of-life outcomes were not assessed. These findings need to be validated in larger multicenter studies with serial biomarker measurements and longer follow-up to define clinically relevant biomarker thresholds [2-8].

This study overall suggests that early serum biomarkers have a role as useful adjuncts in predicting outcome following moderate-to-severe traumatic brain injury. GFAP and UCH-L1 are especially promising as they represent complementary aspects of brain injury and maintained independent prognostic value even after adjustment for clinical and radiological severity. They can enhance prognosis and be used for personalized clinical decision-making in the field of neurotrauma [15,20].

CONCLUSION

Early serum biomarker assessment provided significant prognostic information in patients with moderate-to-severe traumatic brain injury. Admission levels of GFAP, UCH-L1, NSE and S100B correlated with increased severity of insult and with poor 6-month functional outcome. Among the single tests, GFAP was the best predictor, whereas the combination of GFAP and UCH-L1 was the most accurate. After adjustment for CT severity, pupillary response, admission GCS and age, GFAP and UCH-L1 were independent predictors. The results indicate that early biomarker profiling could be a useful addition to standard clinical and radiological evaluation in the risk stratification, treatment planning and prediction of outcome in moderate to severe traumatic brain injury.

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