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ORIGINAL RESEARCH

Utility of Serum Occludin as a Prognostic Biomarker of Severe Traumatic Brain Injury and Mediation Role of Acute Lung Injury: A Two-Center Prospective Cohort Study

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Background: Occludin is a crucial biomarker of blood-brain barrier disruption. Here, we investigated the association between serum occludin levels and poor neurological outcomes after severe traumatic brain injury (sTBI) and the mediating effect of acute lung injury (ALI).

Methods: In this two-center prospective cohort study of 246 patients with sTBI and 100 controls, the serum occludin levels were measured. In-hospital ALI and six-month post-sTBI Glasgow Outcome Scale (GOS) scores were documented. Independent factorial relationships between severity and prognosis were determined.

Results: Patients showed notably enhanced serum occludin levels compared to controls. Serum occludin levels were independently associated with Glasgow coma scale (GCS) scores, and Rotterdam computed tomography (CT) scores were linearly related to the likelihood of ALI and poor prognosis (GOS scores of 1–3), as well as independently associated with ALI, poor prognosis, ordinal GOS scores, and continuous GOS scores. The association of serum occludin levels with ALI and poor prognosis was not moderated by age, sex, hypertension, diabetes, alcohol consumption, or tobacco smoking. ALI partially mediates the relationship between serum occludin level and poor prognosis. As confirmed via a series of statistical approaches, prediction models of poor prognosis and ALI incorporating serum occludin levels and their respective independent predictors performed satisfactorily.

Conclusion: A significant increase in serum occludin levels following sTBI is closely correlated with trauma severity, ALI, and poor prognosis. This may partially elucidate the link between serum occludin levels and poor prognosis, thereby strengthening serum occludin as an acceptable prognostic biomarker of sTBI.

Keywords: traumatic brain injury, occludin, severity, prognosis, acute lung injury, mediation role, biomarkers

Introduction

Severe traumatic brain injury (sTBI) is one of the most lethal forms of trauma and is characterized by high mortality and disability rate.[1] Mechanistically, brain tissues are attacked by external forces, leading to irreversible primary brain injury and subsequent secondary brain injury.[2] The latter has intricate pathophysiological mechanisms, including an excessive inflammatory response, mitochondrial dysfunction, and oxidative imbalance, subsequently leading to blood-brain barrier destruction and exacerbation of brain edema, finally causing neuronal death, neurological impairments, and even death of patients.[3,4] Acute lung injury (ALI) is a complication of high-frequency sTBI management that can easily lead to poor prognosis in patients.[5] The Glasgow Coma Scale (GCS) and Rotterdam computed tomography (CT) classification are the

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two favorite approaches for severity estimation of sTBI.[6,7] Concerning assessment of neurological status after sTBI, the Glasgow Outcome Scale (GOS) is accepted as a usable avenue in clinical practice.[8] Given that blood biomarkers are easy to obtain, they have been extensively studied in recent decades with regard to the prognostic significance of sTBI during recent decades.[9–11]

The blood-brain barrier (BBB) is a paramount structure of the central nervous system, and compromised BBB structure or function is a pathological hallmark of various neurological diseases.[12] Tight junction (TJ)-associated proteins maintain integrity and function of blood-brain barrier (BBB).[13] Occludin was the first identified transmembrane TJ protein and massively contributes to BBB integrity.[14] Blood occludin may be a biochemical marker of BBB destruction and brain edema.[15–18] Moreover, higher serum occludin levels after acute ischemic stroke are independently associated with early neurological deterioration, hemorrhage transformation, and poor prognosis.[19,20] These findings suggest that serum occludin is a biomarker of brain injury. In addition, occludin is abundant in alveolar cells and essential for maintaining normal alveolar barrier integrity in ALI as a result of ventilation or acute respiratory distress syndrome,[21,22] meaning that occludin may be an indicator of ALI. Currently, several neuron- or glia- specific biomarkers, such as neuron specific enolase, S100-beta protein, glial fibrillary acidic protein, neurofilament light, tau and ubiquitin carboxyl hydrolase L1, have been fully studied regarding their prognostic values in sTBI.[23–25] However, occludin is different from the preceding biomarkers, because it functions as a TJ protein with apparent participation in both brain injury and lung injury,[19–22] therefore proffering a notable insight into clinical study in relation to ALI and neurological prognosis following sTBI. Here, serum occludin levels were measured in humans with sTBI in an effort to discern its relevance to trauma severity, ALI, and neurological outcome, while assessing the mediating role of ALI on its association with poor prognosis.

Materials and Methods

Study Design. Subject Enrollments and Ethical Statements

This observational analytic study was conducted between September 2021 and October 2023 at two comprehensive hospitals: Shengzhou People's Hospital (Shengzhou, China) and Longquan People's Hospital (Longquan, China). Intrinsically, two sub-studies, namely the cross-sectional analysis and prospective cohort assessment, composed this clinical investigation. First, controls and patients with sTBI were employed to unravel the evolutionary course of serum occludin levels subsequent to sTBI. In the latter part, patients were used to determine the role of serum occludin as a prognostic metric for sTBI. Patients were required to be at least 18 years old, stay in a state of pretraumatic GOS score of 1, be admitted to the hospital within post-trauma twelve hours on account of blunt, isolated head trauma, and experience post-resuscitation GCS scores from 3 to 8. Subsequently, we excluded patients with certain neurological illnesses, such as cerebrovascular accidents, brain neoplasms, and head trauma, as well as those with specific conditions or severe diseases, such as the use of steroid hormones, infections within the past month, pregnancies, malignancies, heart failure, and ascites. Controls comprised volunteers who were free from some chronic illnesses, such as hypertension, diabetes, and dyslipidemia, and had normal results in some routine tests, such as blood erythrocyte counts, blood hemoglobin levels, blood leukocyte counts, blood neutrophil counts, and blood potassium levels. This study was performed in accordance with the Declaration of Helsinki and follow-up updates. The study protocol was approved by the Institutional Review Committees of the Shengzhou People's Hospital (No. 2021–039-01) and Longquan People's Hospital (No. 2018–023). The lawful representatives of patients and controls were familiarized with the study details and then independently provided written informed consent forms.

Data Collection and Neurological Functional Assessment

In two centers, operational modalities and other therapeutic methods were adopted to treat patients based on the protocol specified in the guidelines for the management of sTBI.[26] All patients were consecutively enrolled and their admission time since trauma was documented. At the emergency center, we inquired about their age, sex, tobacco smoking, alcohol consumption, hypertension, and diabetes mellitus. Traumatic causes were dichotomized into traffic accidents and other types. Readings were obtained following non-invasive measurements of arterial blood pressure at the emergency center.

So as to mirror the severity of the sTBI, post-resuscitation GCS scores at admission were recorded by trained assessors, who were blinded to the other clinical materials. Based on head CT imaging, positive radiological hemorrhagic manifestations were categorized into two types: mass and non-mass lesion bleeding. The former encompasses hematomas localized in the intracerebral, epidural, and subdural cavities, while the latter consists of bleeding located in the intraventricular and subarachnoid spaces. The non-bleeding appearance included brain contusions and pneumocephalus. Rotterdam CT scale scores were computed in accordance with radiological data. Operation within 24 hours and seizure onset subsequent to sTBI were registered. In-hospital ALI was diagnosed according to previous criteria as follows: acute emergences of clinical symptoms with the ratio of partial pressure of arterial oxygen to fractional inspired oxygen equal to or below 300, findings of bilateral infiltrates via chest radiograph and exclusion of left arterial hypertension.[27] Via the structured interviews, GOS scores at six-month mark post-sTBI were documented by the experienced clinician through a telephone visit, in order to reflect neurological function, and scores of 1–3 is indicative of a poor prognosis.[28]

Immune Test

All standardized procedures were strictly performed in two centers according to pre-established specifications. Blood samples were collected via venous access from the patients and controls. A rapid action was done to put blood samples in 5 mL gel-prefilled biochemistry tubes, and after centrifugation 2000 × g for 10 minutes, the supernatant was extracted with transferal into Eppendorf tubes for storage at −80°C conditions awaiting measurements. Within three months of preservation, serum specimens must be thawed for quantification of occludin levels. Serum occludin levels were detected in duplicate by the same professional personnel using a sandwich enzyme-linked immunosorbent assay kit (item number, CSB-EL016263HU; CUSABIO, China). As for this kit, the detection range was from 23.5 to 1500 pg/mL, the minimum detectable limit was 5.8 pg/mL, and the inter- and intra-assay precision coefficients of variation were below 10%.

Statistical Analysis

The SPSS statistical package (version 20.0; SPSS Inc., Chicago, Illinois, USA) was used for statistical analyses. The data were divided into categorical and continuous variables. The former was expressed in the form of frequencies (proportions). For the latter, the Shapiro–Wilk test or Kolmogorov–Smirnov test was performed to identify normality. Normally distributed variables were reported as means (standard deviations, SDs) if non-normally distributed, and as medians (percentiles 25th-75th). To compare the data between two groups or among multiple groups, the independent *t*-test, Mann–Whitney *U*-test, Pearson's Chi-square test, Fisher's exact test, and Kruskal–Wallis test were employed as needed. Bivariate correlations were investigated using the Spearman's test. Here, five dependent variables were selected, including continuous GOS scores, serum occludin levels, ALI, poor prognosis and ordinal GOS scores, and many kinds of multivariate statistical methods were employed, covering the multivariate linear regression analysis, binary logistic regression analysis and ordinal regression analysis. Significantly distinct factors in the univariate analyses were included in the multivariate models to identify the independent variables. Discrimination efficiencies were estimated from the receiver operating characteristic (ROC) curve using MedCalc statistical software version 17.4 (MedCalc Software, Mariakerke, Belgium). The R software (version 3.5.1; https://www.r-project.org), variance inflation factor (VIF) was calculated, and the nomogram, restricted cubic spline, decision curve, forest plot, and graphs of mediation and subgroup analyses were obtained. Scatter plots, bar plots, and ROC curves were plotted using the GraphPad Prism 7.01 (GraphPad Software, Inc., San Diego, California, USA). Two-sided statistical significance was defined as $P < 0.05$.

Results

Participant Features

After consecutive recruitment, 295 patients with sTBI met the inclusion criteria. Subsequently, we excluded 49 patients, including 18 with previous neurological diseases, 21 with some specific conditions or severe diseases, 3 rejected study entry, 2 lost to follow-up, 2 provided inadequate information, and 3 had unqualified blood samples. Ultimately, 246 patients were included in this analysis. The baseline patient characteristics are shown in Table 1. One hundred controls were aged from 20 to 71 years (mean, 44.8 years; SD, 13.3 years), the male to female was 1.33 (57/43), and included 34

Table 1 Baseline Characteristics and Correlativity Analyses in Severe Traumatic Brain Injury

Variables	All Patients	Bivariate Correlation Analyses by the Spearman Test			
		Serum Occludin Levels		Six-Month GOS Scores	
		ρ	P value	ρ	P value
Gender (male/female)	142/104	0.080	0.210	−0.102	0.110
Age (years)	43.7±12.6	0.114	0.075	−0.184	0.004
Cigarette smoking	75 (30.5%)	−0.032	0.620	0.053	0.410
Alcohol drinking	73 (29.7%)	−0.046	0.473	0.083	0.196
Hypertension	60 (24.4%)	0.013	0.845	−0.003	0.964
Diabetes mellitus	38 (15.4%)	0.081	0.206	−0.154	0.016
Admission time (h)	5.9 (4.7–7.0)	0.067	0.295	−0.026	0.687
Blood-drawing time (h)	6.9 (5.7–8.6)	0.031	0.629	0.028	0.663
Traffic accidents	137 (55.7%)	−0.115	0.072	0.060	0.347
GCS scores	5 (4–7)	−0.679	<0.001	0.611	<0.001
Systolic arterial pressure (mmHg)	131.5 ± 23.1	0.078	0.223	0.064	0.314
Diastolic arterial pressure (mmHg)	77.9 ± 13.2	0.058	0.365	−0.029	0.654
Rotterdam CT scores	4 (4–5)	0.544	<0.001	−0.608	<0.001
Abnormal cisterns	190 (77.2%)	0.322	<0.001	−0.253	<0.001
Midline shift above 5 mm	148 (60.2%)	0.352	<0.001	−0.453	<0.001
Epidural hematoma	128 (52.0%)	0.043	0.499	−0.067	0.296
Subdural hematoma	139 (56.5%)	0.093	0.148	−0.069	0.282
Subarachnoid hemorrhage	160 (65.0%)	0.153	0.016	−0.202	0.001
Intraventricular hemorrhage	26 (10.6%)	0.055	0.386	−0.053	0.406
Intracerebral hematoma	128 (52.0%)	0.092	0.151	−0.154	0.015
Brain contusion	136 (55.3%)	0.131	0.040	−0.064	0.320
Pneumocephalus	88 (35.8%)	−0.040	0.535	−0.024	0.704
Operation within 24 hours	107 (43.5%)	−0.070	0.271	−0.045	0.482
Acute lung injury	71 (28.9%)	0.394	<0.001	−0.363	<0.001
Seizure	69 (28.0%)	−0.018	0.781	−0.107	0.093
Blood glucose levels (mmol/l)	8.5 (6.7–11.7)	0.232	<0.001	−0.227	<0.001
Blood WBC count ($\times 10^9$/l)	7.2 (5.8–9.2)	0.094	0.143	−0.200	0.002
Serum occludin levels (ng/mL)	8.8 (4.6–14.1)	-	-	−0.576	<0.001

Abbreviations: CT, computed tomography; GCS, Glasgow coma scale; GOS, Glasgow outcome scale; WBC, white blood cell.

tobacco smokers and 33 alcohol drinkers. From a statistical perspective, the above four variables were not significantly different between controls and patients (all P>0.05).

Serum Occludin Levels and sTBI-Related Severity

Serum occludin levels were markedly higher in the patients than in the controls (P<0.001; Figure 1). In subgroups with GCS scores spanning from 3 to 8, significant differences were existent in terms of serum occludin levels among six subgroups, with the highest levels in subgroup of GCS score 3, followed by those with the scores from 4 to 6 and the lowest levels in that of GCS score 8 (P<0.001; Figure 2); consistently, serum occludin levels were significantly in opposite proportion to GCS scores (P<0.001; Figure 3). Alternatively, Serum occludin levels substantially increased in the order of Rotterdam CT scores, varying from 3 to 6 (P<0.001; Figure 4), and serum occludin levels were markedly enhanced with increasing Rotterdam CT scores (P<0.001; Figure 5). To be continued, aside from GCS scores and Rotterdam CT scores, other factors of notable correlation with serum occludin levels were abnormal cisterns, midline shift above 5 mm, subarachnoid hemorrhage, brain contusion, ALI and blood glucose levels (all P<0.05; Table 1). Excluding ALI as a subsequent event and three components of the Rotterdam CT scoring system, namely abnormal cisterns, midline shift > 5 mm, and subarachnoid hemorrhage, the remaining four variables were forced into the multiple-factorial model. As a result, Serum occludin levels were independently correlated with GCS score [beta (β), −2.530; 95%

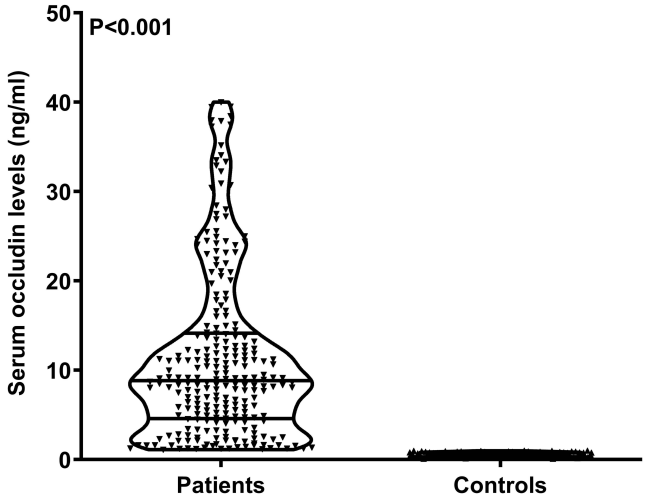

Figure 1 Variation of serum occludin levels subsequent to severe traumatic brain injury. There was a substantial increase in serum occludin levels in patients with severe traumatic brain injury compared to controls (P<0.001).

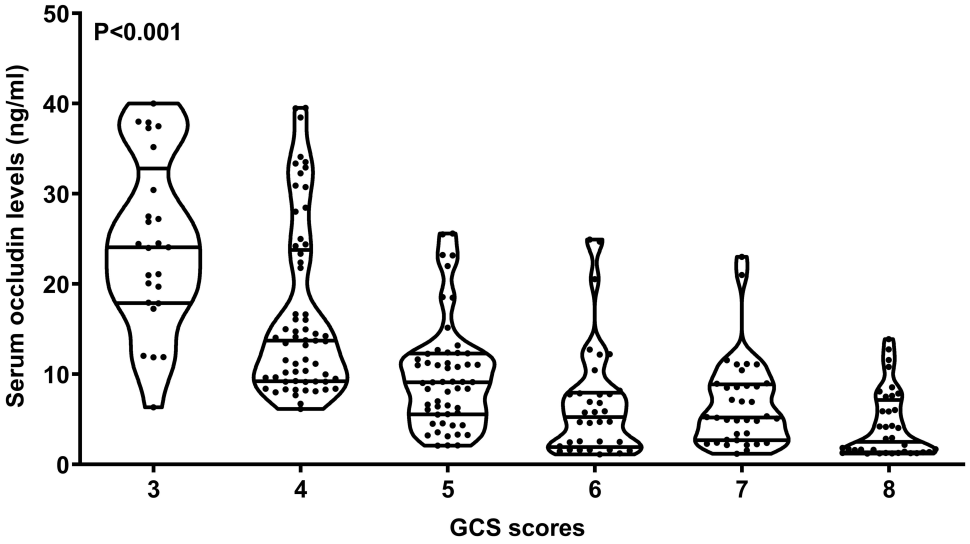

Figure 2 Disparities of serum occludin levels at a basis of Glasgow coma scale scores following severe traumatic brain injury. Serum occludin levels of patients were notably distinct among the six subgroups subdivided based on Glasgow coma scale scores post-severe traumatic brain injury, manifested with the highest levels in the subgroup with a score of 3, the medium levels in those with scores from to 4–7 and the lowest levels in those with a score of 8 (P<0.001).
Abbreviation: GCS, Glasgow coma scale.

confidence interval (CI], −3.241–1.819; VIF, 1.119; P=0.001) and Rotterdam CT score (β, 2.427; 95% CI, 1.232–3.622; VIF, 1.538; P=0.001).

Serum Occludin Levels and 6-Month Neurological Functional Status Following sTBI

As outlined in Figure 6, six-month GOS scores were intimately correlated with the serum occludin levels (P<0.001; Figure 6). In addition, the GOS scores were closely related to age, diabetes mellitus, GCS scores, Rotterdam CT scores, abnormal cisterns, midline > 5 mm, subarachnoid hemorrhage, intracerebral hemorrhage, ALI, blood glucose levels, and blood leukocyte counts (all P<0.05; Table 1). Similar to the preceding interpretation, abnormal cisterns, midline > 5 mm, and subarachnoid hemorrhage were excluded from the multivariate model. Therefore, the model was inclusive of the rest, leading to the revelation that GOS scores were independently associated with GCS scores (β, 0.224; 95% CI; 0.120–0.327; VIF, 1.991; P=0.001), Rotterdam CT scores (β, −0.491; 95% CI, −0.659–0.322; VIF, 1.793; P=0.002),

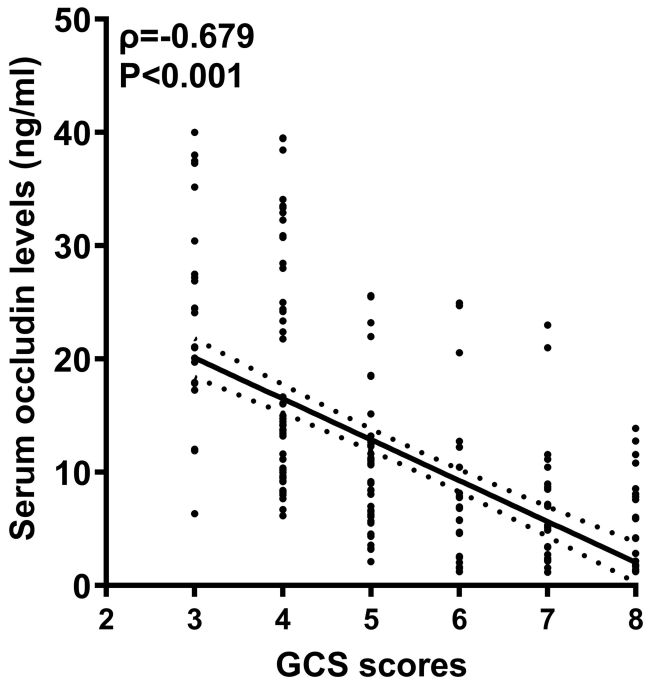

Figure 3 Relationship between serum occludin levels and Glasgow coma scale scores of humans diseased of severe traumatic brain injury. Serum occludin levels were inversely proportional to the Glasgow Coma Scale scores among individuals with severe traumatic brain injury (P<0.001).
Abbreviation: GCS, Glasgow coma scale.

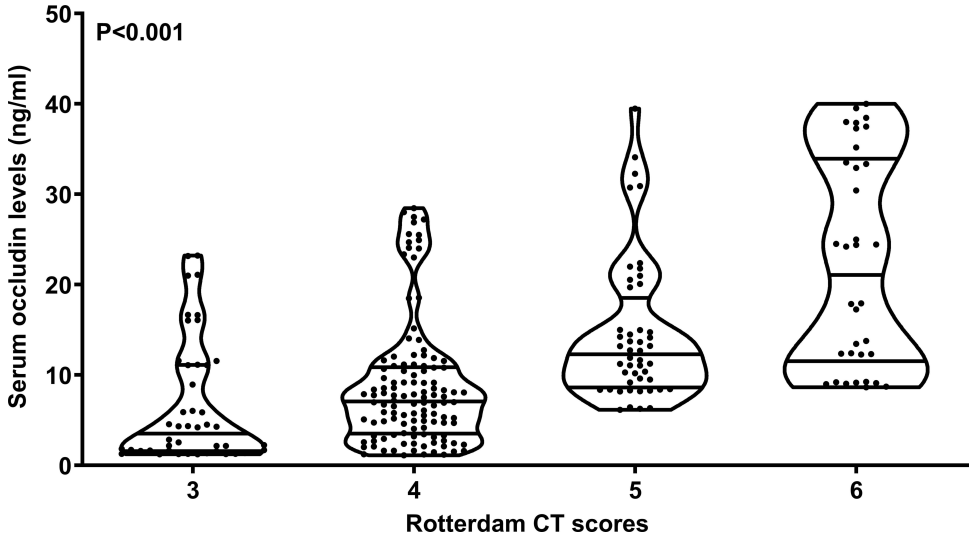

Figure 4 Distinction of serum occludin levels according to Rotterdam computed tomography scores in patients experiencing severe traumatic brain injury. Serum occludin levels of cases showed significant differences among the four subgroups categorized in accordance with Rotterdam computed tomography scores after severe traumatic brain injury, marked by the highest levels in the subgroup with score 6, the medium levels in those with scores 4–5 and the lowest levels in those with score 3 (P<0.001).
Abbreviation: CT, computed tomography.

ALI (β, −0.351; 95% CI, −0.682–0.020; VIF, 1.267; P=0.038), and serum occludin levels (β, −0.024; 95% CI, −0.041–0.007; VIF, 1.843; P=0.006).

Five subgroups based on GOS scores had an exhibition of pronouncedly distinct serum occludin levels, marked by the highest levels in the subgroup with a score of 1, a subsequent gradual decline in the levels with increasing scores from 2 to 4, and the lowest levels in the subgroup with a score of 5 (P<0.001; Figure 7). Among the five subgroups, there were ten variables of substantial disparity: age, GCS score, Rotterdam CT score, abnormal cisterns, midline > 5 mm,

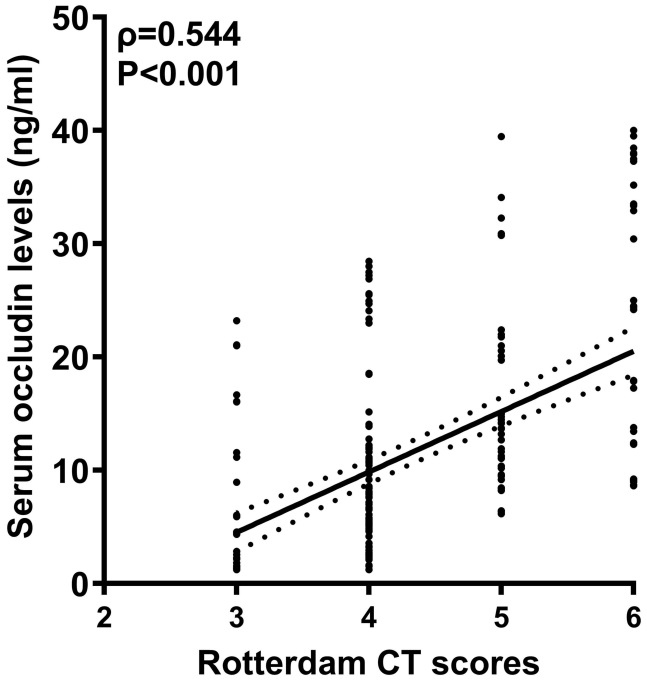

Figure 5 Relation of serum occludin levels to Rotterdam computed tomography scores among individuals suffering from severe traumatic brain injury. Serum occludin levels were positively correlated with Rotterdam computed tomography scores after severe traumatic brain injury (P<0.001).
Abbreviation: CT, computed tomography.

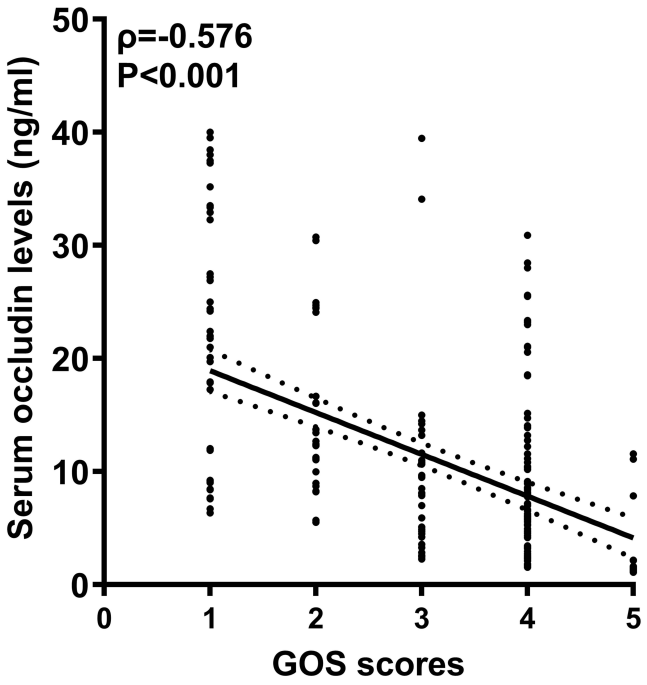

Figure 6 Correlation of serum-based occludin levels with Glasgow outcome scale scores in subjects diseased of severe traumatic brain injury. Serum-based occludin levels were inversely correlated with the Glasgow outcome scale scores of patients with severe traumatic brain injury (P<0.001).
Abbreviation: GOS, Glasgow outcome scale.

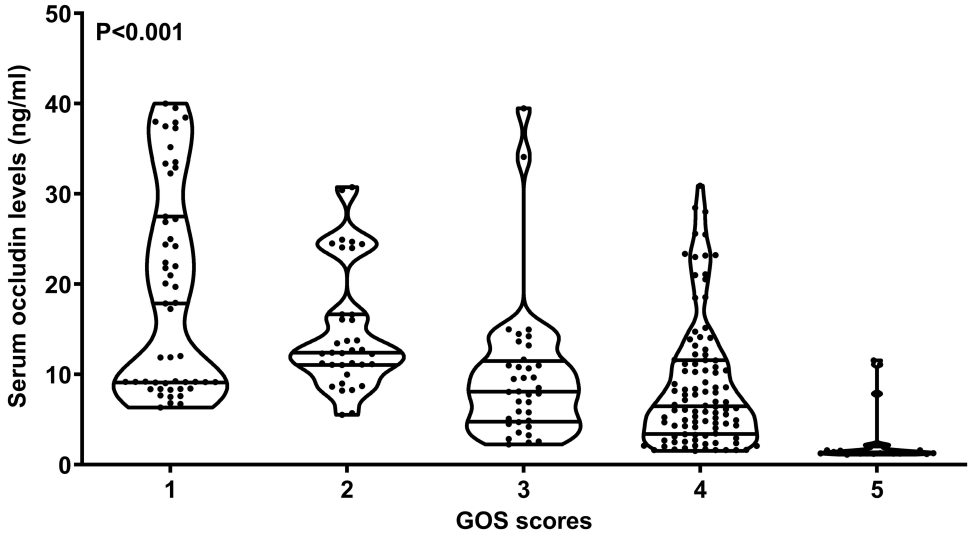

Figure 7 Differences of serum-based occludin levels among subgroups with distinct Glasgow outcome scale scores post-severe traumatic brain injury. Serum-based occludin levels were substantially higher in patients with a Glasgow outcome scale score of 1, followed by those with scores of 2–4, and were markedly lower in those with a score of 5, following severe traumatic brain injury (P<0.001).
Abbreviation: GOS, Glasgow outcome scale.

subarachnoid hemorrhage, serum occludin levels, ALI, blood glucose levels, and blood leukocyte counts (all P<0.05; Table 2). Considering that the Rotterdam CT classification is composed of abnormal cisterns, midline > 5 mm, and subarachnoid hemorrhage, these three factors were not included in the multivariate model. The other seven variables

Table 2 Relevant Factors to Ordinal Glasgow Outcome Scale Scores After Severe Traumatic Brain Injury

Variables	Six-month Glasgow Outcome Scale Scores					P value
	1	2	3	4	5	
Gender (male/female)	34/17	20/15	22/14	54/46	12/12	0.558
Age (years)	52 (38–62)	44 (36–54)	41 (38–46)	39 (35–50)	43 (37–47)	0.012
Cigarette smoking	15 (29.4%)	6 (17.1%)	13 (36.1%)	34 (34.0%)	7 (29.2%)	0.392
Alcohol drinking	14 (27.5%)	8 (22.9%)	11 (30.6%)	29 (29.0%)	11 (45.8%)	0.414
Hypertension	15 (29.4%)	5 (14.3%)	10 (27.8%)	23 (23.0%)	7 (29.2%)	0.515
Diabetes mellitus	14 (27.5%)	5 (14.3%)	6 (16.7%)	10 (10.0%)	3 (12.5%)	0.109
Admission time (h)	5.9 (5.4–7.0)	5.9 (3.1–6.8)	6.4 (5.0–8.3)	5.8 (5.0–6.6)	6.1 (4.3–7.0)	0.150
Blood-drawing time (h)	6.7 (6.1–8.7)	6.9 (3.9–8.4)	7.7 (6.1–9.7)	6.8 (6.0–8.0)	7.4 (5.3–9.0)	0.120
Traffic accidents	27 (52.9%)	17 (48.6%)	21 (58.3%)	57 (57.0%)	15 (62.5%)	0.827
GCS scores	4 (3–4)	5 (4–5)	5 (4–6)	6 (5–7)	8 (7–8)	<0.001
Systolic arterial pressure (mmHg)	129 (113–164)	130 (124–135)	130 (116–142)	132 (119–140)	133 (130–164)	0.628
Diastolic arterial pressure (mmHg)	79 (72–95)	80 (78–86)	75 (71–87)	77 (70–85)	82 (76–90)	0.115
Rotterdam CT scores	5 (4–6)	5 (4–6)	4 (4–5)	4 (4–4)	3 (3–4)	<0.001
Abnormal cisterns	50 (98.0%)	30 (85.7%)	26 (72.2%)	66 (66.0%)	18 (75.0%)	<0.001
Midline shift above 5 mm	48 (94.1%)	26 (74.3%)	20 (55.6%)	52 (52.0%)	2 (8.3%)	<0.001
Epidural hematoma	27 (52.9%)	18 (51.4%)	24 (66.7%)	49 (49.0%)	10 (41.7%)	0.341
Subdural hematoma	28 (54.9%)	23 (65.7%)	26 (72.2%)	48 (48.0%)	14 (58.3%)	0.097
Subarachnoid hemorrhage	43 (84.3%)	23 (65.7%)	25 (69.4%)	54 (54.0%)	15 (62.5%)	0.007
Intraventricular hemorrhage	6 (11.8%)	3 (8.6%)	8 (22.2%)	6 (6.0%)	3 (12.5%)	0.103
Intracerebral hematoma	33 (64.7%)	18 (51.4%)	20 (55.6%)	49 (49.0%)	8 (33.3%)	0.126
Brain contusion	27 (52.9%)	19 (54.3%)	26 (72.2%)	56 (56.0%)	8 (33.3%)	0.061

(*Continued*)

Table 2 (Continued).

Variables	Six-month Glasgow Outcome Scale Scores					P value
	1	2	3	4	5	
Pneumocephalus	22 (43.1%)	8 (22.9%)	14 (38.9%)	36 (36.0%)	8 (33.3%)	0.411
Operation within 24 hours	24 (47.1%)	15 (42.9%)	19 (52.8%)	37 (37.0%)	12 (50.0%)	0.454
Acute lung injury	31 (60.8%)	11 (31.4%)	9 (25.0%)	19 (19.0%)	1 (4.2%)	<0.001
Seizure	22 (43.1%)	7 (20.0%)	9 (25.0%)	25 (25.0%)	6 (25.0%)	0.107
Blood glucose levels (mmol/l)	9.6 (8.3–15.2)	8.9 (6.9–12.4)	7.7 (6.7–11.4)	8.4 (6.2–11.7)	7.8 (6.8–9.2)	0.003
Blood WBC count ($\times 10^9$/l)	8.5 (6.3–11.2)	6.7 (5.5–9.3)	6.9 (5.8–9.2)	7.1 (5.8–8.8)	6.5 (5.0–8.2)	0.013
Serum occludin levels (ng/mL)	17.9 (9.1–27.4)	12.4 (11.1–16.7)	8.1 (4.8–11.3)	6.5 (3.4–11.6)	1.4 (1.2–1.9)	<0.001

Notes: Frequencies (percentages) and medians (lower-upper quartiles) were reported for showing categorical and continuous variables separately. Where applicable, the Pearson's Chi-square test and Kruskal–Wallis *H*-test were implemented for data multigroup comparisons.
Abbreviations: CT, computed tomography; GCS, Glasgow coma scale; WBC, white blood cell.

were entered into the ordinal multivariate model, and then it was confirmed that GOS scores were independently associated with GCS scores (β, 0.539; 95% CI; 0.307–0.770; VIF, 1.959; P=0.003), Rotterdam CT scores (β, −0.960; 95% CI, −1.325–0.596; VIF, 1.738; P=0.005), ALI (β, −0.763; 95% CI; −1.340–0.186; VIF, 1.352; P=0.010), and serum occludin levels (β, −0.047; 95% CI, −0.084–0.010; VIF, 1.813; P=0.013).

As depicted in Figure 8, serum occludin levels were significantly higher in patients with a poor prognosis than in those with a good prognosis (P<0.001). Meanwhile, as shown in Figure 9, serum occludin levels displayed strong discrimination efficiency for likelihood of poor prognosis and the optimal value was selected as 6.6 ng/mL, which anticipated poor prognosis with 86.9% sensitivity and 58.9% specificity (the maximum Youden index at 0.458). Figure 10 shows the presence of a linear correlation between serum occludin levels and the possibility of a poor prognosis (P for nonlinear >0.05). The results of the univariate analyses are outlined in Table 3, showing that several factors, such as age, diabetes mellitus, GCS scores, Rotterdam CT scores, abnormal cisterns, midline > 5 mm, subarachnoid hemorrhage, subdural hematoma, ALI, serum occludin levels, blood glucose levels, and blood leukocyte counts, were substantially different between patients with and without poor prognosis (all P<0.05). Thereafter, other significantly different parameters, but not the three components of the Rotterdam CT assessment system, such as abnormal cisterns, midline > 5 mm, and subarachnoid hemorrhage, were integrated into the binary multivariate model. A further finding was that serum occludin levels [odds ratio (OR), 1.051; 95% CI, 1.006–1.098; VIF, 1.611; P=0.026],

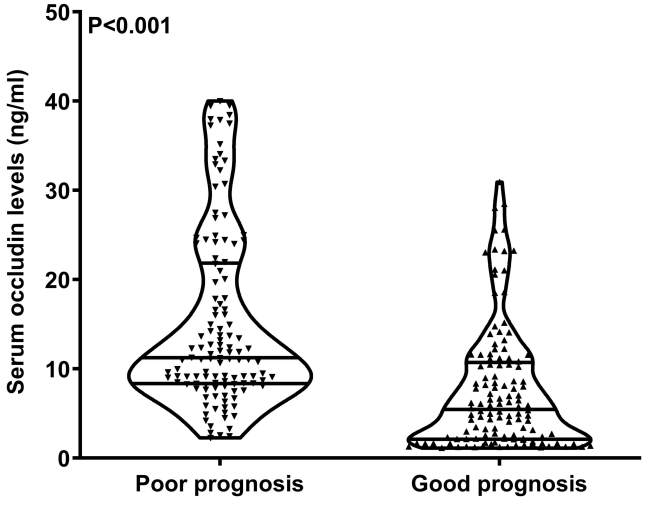

Figure 8 Serum occludin levels between patients with the development of poor prognosis and those without such a condition after severe traumatic brain injury. Occludin levels in the serum of patients with severe traumatic brain injury were significantly higher in patients with poor prognosis than in those without such adverse events (P<0.001).

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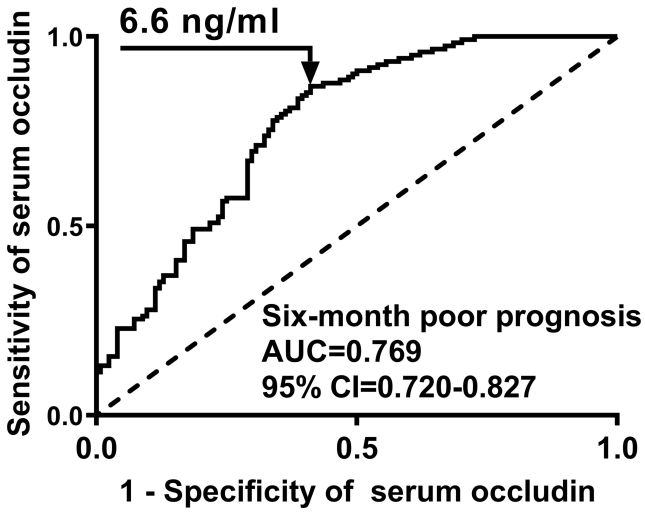

Figure 9 Receiver operating characteristic curve as to serum occludin levels for predicting poor prognosis subsequent to severe traumatic brain injury. Serum occludin levels were at a state of effective discrimination ability for poor prognosis, and the optimal value of the levels was designated as 6.6 ng/mL, yielding the maximum Youden index for predicting prognosis in severe traumatic brain injury. Arrow indicates threshold value of serum occludin levels.
Abbreviations: AUC, area under curve; 95% CI, 95% confidence interval.

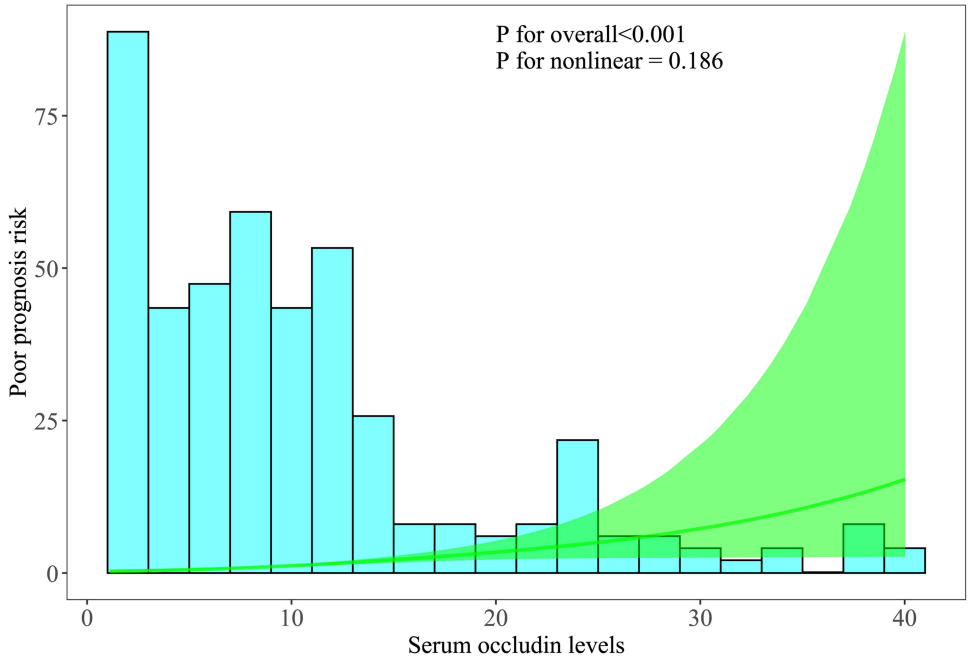

Figure 10 Restricted cubic spline for assessment of linear relationship between serum occludin levels and probability of poor prognosis following severe traumatic brain injury. Serum occludin levels exhibited a linear relationship with the risk of poor prognosis after severe brain injury (P for nonlinear > 0.05).

GCS scores (OR, 0.621; 95% CI, 0.467–0.827; VIF, 1.431; P=0.001), ALI (OR, 2.926; 95% CI, 1.564–5.474; VIF, 2.128; P=0.015) and Rotterdam CT scores (OR, 2.665; 95% CI, 1.639–4.334; VIF, 2.116; P=0.002) remained as the four independent predictors of poor prognosis after sTBI. Moreover, several conventional variables such as age, sex, hypertension, diabetes, alcohol consumption, and cigarette smoking negligibly moderated the relationship between serum occludin levels and the possibility of poor prognosis in patients with sTBI (all P for interaction >0.05; Figure 11). The preceding four factors were consolidated to create a combined model to forecast poor prognosis. The

Table 3 Pertinent Variables to Six-month Functional Outcome and Acute Lung Injury After Severe Traumatic Brain Injury

Variables	Six-Month Neurological Status			Acute Lung Injury		
	Poor Prognosis	Good Prognosis	*P* value	Presence	Absence	*P* value
Gender (male/female)	76/46	66/58	0.150	40/31	102/73	0.779
Age (years)	45.7 ± 13.1	41.8 ± 11.9	0.014	47.0 ± 13.6	42.4 ± 12.0	0.008
Cigarette smoking	34 (27.9%)	41 (33.1%)	0.376	22 (31.0%)	53 (30.3%)	0.914
Alcohol drinking	33 (27.0%)	40 (32.3%)	0.371	21 (29.6%)	52 (29.7%)	0.983
Hypertension	30 (24.6%)	30 (24.2%)	0.942	13 (18.3%)	47 (26.9%)	0.157
Diabetes mellitus	25 (20.5%)	13 (10.5%)	0.030	16 (22.5%)	22 (12.6%)	0.050
Admission time (h)	6.0 (4.6–7.5)	5.8 (4.9–6.8)	0.532	5.9 (4.7–7.7)	5.9 (4.8–7.0)	0.416
Blood-drawing time (h)	7.2 (5.6–8.9)	6.8 (5.9–8.3)	0.914	6.8 (5.9–9.2)	7.0 (5.7–8.4)	0.395
Traffic accidents	65 (53.3%)	72 (58.1%)	0.450	34 (47.9%)	103 (58.9%)	0.117
GCS scores	4 (4–5)	6 (5–8)	<0.001	4 (4–5)	6 (5–7)	<0.001
Systolic arterial pressure (mmHg)	131.5± 24.2	131.4 ± 22.0	0.976	133.3 ± 26.4	130.7 ± 21.6	0.465
Diastolic arterial pressure (mmHg)	78.9 ± 14.5	76.9 ± 11.7	0.231	78.4 ± 15.2	77.6 ± 12.3	0.706
Rotterdam CT scores	5 (4–6)	4 (3–4)	<0.001	5 (4–6)	4 (3–4)	<0.001
Abnormal cisterns	106 (86.9%)	84 (67.7%)	<0.001	64 (90.1%)	126 (72.0%)	0.002
Midline shift above 5 mm	94 (77.0%)	54 (43.5%)	<0.001	54 (76.1%)	94 (53.7%)	0.001
Epidural hematoma	69 (56.6%)	59 (47.6%)	0.159	39 (54.9%)	89 (50.9%)	0.562
Subdural hematoma	77 (63.1%)	62 (50.0%)	0.038	48 (67.6%)	91 (52.0%)	0.025
Subarachnoid hemorrhage	91 (74.6%)	69 (55.6%)	0.002	54 (76.1%)	106 (60.6%)	0.021
Intraventricular hemorrhage	17 (13.9%)	9 (7.3%)	0.089	11 (15.5%)	15 (8.6%)	0.110
Intracerebral hematoma	71 (58.2%)	57 (46.0%)	0.055	43 (60.6%)	85 (48.6%)	0.088
Brain contusion	72 (59.0%)	64 (51.6%)	0.243	39 (54.9%)	97 (55.4%)	0.943
Pneumocephalus	44 (36.1%)	44 (35.5%)	0.924	25 (35.2%)	63 (36.0%)	0.907
Operation within 24 hours	58 (47.5%)	49 (39.5%)	0.204	29 (40.8%)	78 (44.6%)	0.593
Acute lung injury	51 (41.8%)	20 (16.1%)	<0.001	-	-	-
Seizure	38 (31.1%)	31 (25.0%)	0.283	14 (19.7%)	55 (31.4%)	0.064
Blood glucose levels (mmol/l)	8.8 (7.2–13.5)	8.0 (6.3–10.7)	0.009	9.2 (7.2–14.0)	8.4 (6.7–11.1)	0.026
Blood WBC count ($\times 10^9$/l)	7.5 (5.9–10.0)	7.0 (5.7–8.6)	0.025	7.8 (6.0–9.7)	6.8 (5.7–8.9)	0.031
Serum occludin levels (ng/mL)	11.3 (8.4–21.8)	5.5 (2.1–10.6)	<0.001	12.1 (9.1–24.4)	7.5 (2.8–11.9)	<0.001

Notes: Counts (proportions), medians (lower-upper quartiles) and mean ± standard deviation were shown for variables. The Pearson's Chi-square test, Mann–Whitney *U*-test and independent *t* test was in utilization for data comparisons.
Abbreviations: CT, computed tomography; GCS, Glasgow coma scale; WBC, white blood cell.

model was described in the form of a nomogram (Figure 12), had a good fit (Figure 13), possessed satisfactory clinical benefits (Figure 14), and had a substantially higher prognostic capability than its four components under the ROC curve (all P<0.05; Figure 15).

Subgroup	Total	OR(95% CI)	P	P interaction
Age				0.193
≥ 50 years	73	1.083 (1.011-1.159)	0.023	
< 50 years	173	1.064 (1.010-1.121)	0.019	
Gender				0.074
Male	142	1.102 (1.030-1.180)	0.005	
Female	104	1.035 (0.973-1.101)	0.269	
Hypertension				0.622
Yes	60	1.076 (0.975-1.187)	0.143	
No	186	1.068 (1.017-1.121)	0.008	
Diabetes mellitus				0.184
Yes	38	1.102 (0.897-1.354)	0.354	
No	208	1.067 (1.021-1.114)	0.004	
Cigarette smoking				0.615
Yes	75	1.090 (0.998-1.190)	0.056	
No	171	1.059 (1.007-1.113)	0.025	
Alcohol drinking				0.422
Yes	73	1.051 (0.994-1.112)	0.081	
No	173	1.060 (1.005-1.118)	0.034	

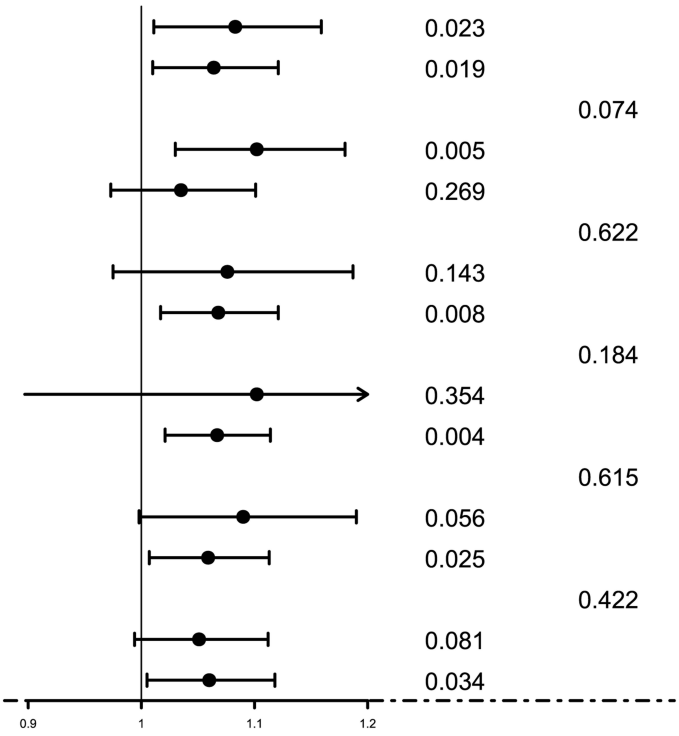

Figure 11 Forest plot assessing subgroup analysis with respect to serum occludin levels' association with poor prognosis following severe traumatic brain injury. The relationship between serum occludin levels and poor prognosis after severe brain injury was not moderated by the conventional variables in the graph (all P interaction > 0.05).
Abbreviations: OR, odds ratio; 95% CI, 95% confidence interval.

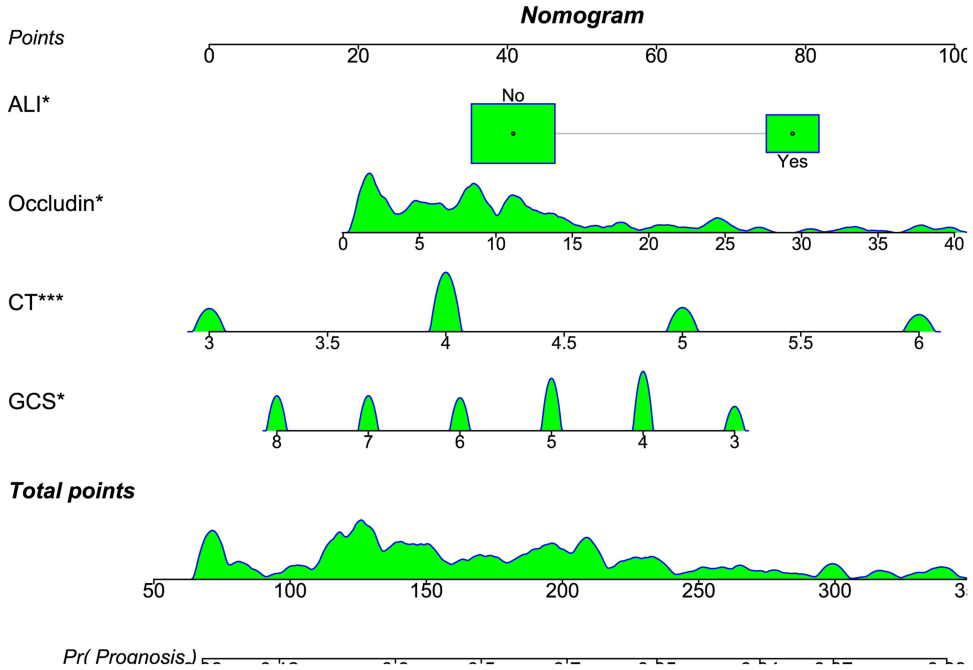

Figure 12 Nomogram evaluating combined model of prognosis prediction in severe traumatic brain injury. The model was composed of four independent predictors of poor prognosis after severe traumatic brain injury. Each variable corresponded to a separate score, and the aggregative scores of the four factors pointed to a risk of poor prognosis.
Abbreviations: ALI, acute lung injury; GCS, Glasgow Coma Scale; CT, Rotterdam computed tomography.

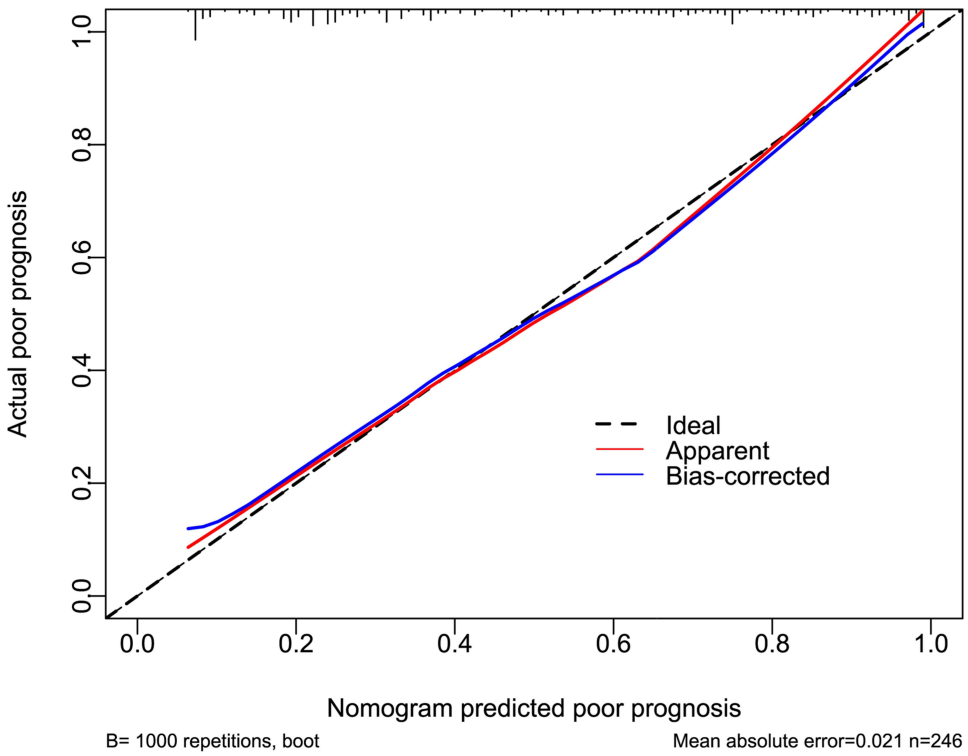

Figure 13 Calibration curve showing model fit of prognosis anticipation following severe traumatic brain injury. The model was constructed based on four independent prognostic factors after severe brain injury. The model showed good fit.

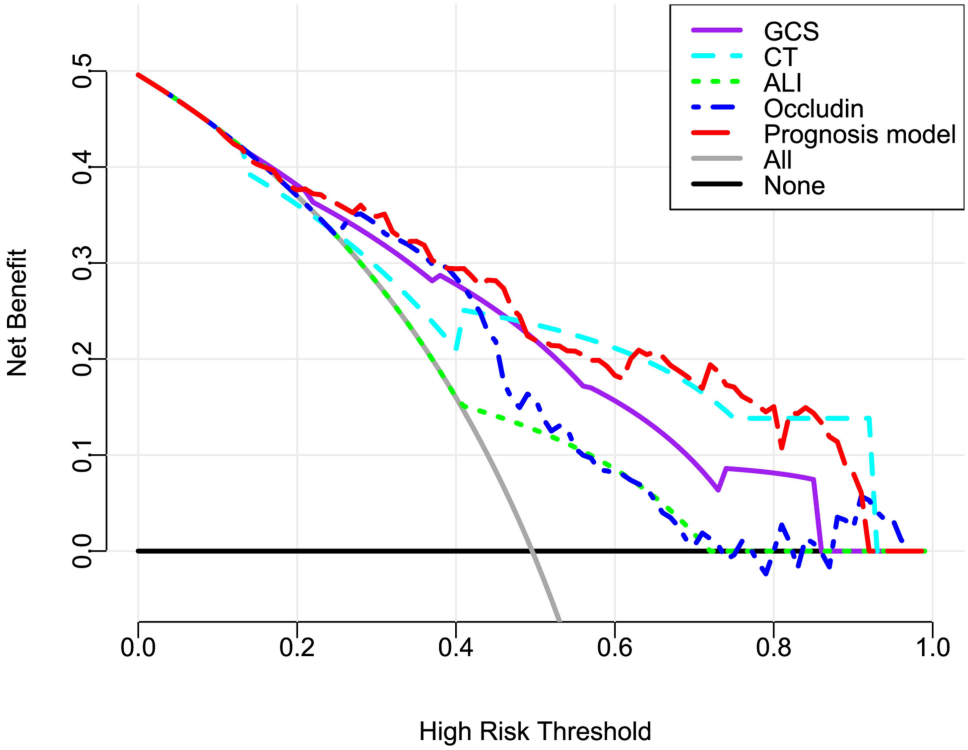

Figure 14 Decision curve displaying clinical validities of prognosis prediction of prognosis model and its components after severe traumatic brain injury. The model was configured based on four predictive factors of poor prognosis following severe traumatic brain injury. This model is clinically valid for severe traumatic brain injury.
Abbreviations: ALI, acute lung injury; GCS, Glasgow Coma Scale; CT, Rotterdam computed tomography.

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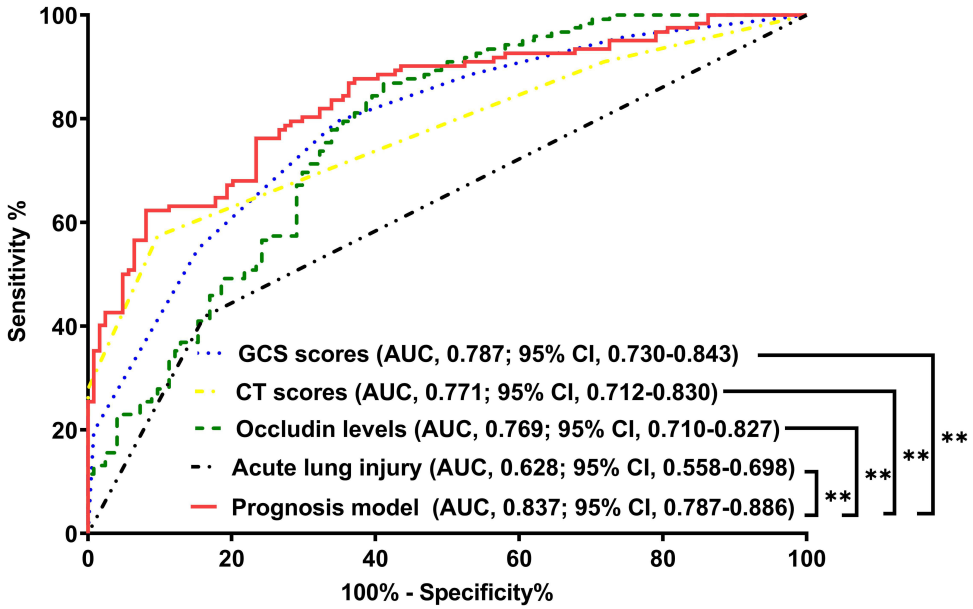

Figure 15 Receiver operating characteristic curve demonstrating prognostic efficiency of merged model and its members following severe traumatic brain injury. The model was established by integrating the four predictors of poor prognosis after severe traumatic brain injury. The model possessed substantially higher predictive ability than the other four independent variables (**P<0.01).
Abbreviations: GCS, Glasgow Coma Scale; CT, Rotterdam computed tomography classification; AUC, area under curve; 95% CI, 95% confidence interval.

Serum Occludin Levels in Relation to ALI and Mediation Role of ALI

As shown in Figure 16, serum occludin levels were markedly enhanced in ALI patients (P<0.001). Simultaneously, as displayed in Figure 17, serum occludin levels exhibited effective predictive value for ALI probability, and a value more than 7.7 ng/mL distinguished ALI risk with 87.3% sensitivity and 51.4% specificity (the maximum Youden index of 0.387). Serum occludin levels linearly correlated with the likelihood of ALI (P for nonlinear >0.05; Figure 18). As shown in Table 3, age, GCS score, Rotterdam CT score, abnormal cisterns, midline > 5 mm, subarachnoid hemorrhage, subdural hematoma, serum occludin levels, blood glucose levels, and blood leukocyte counts were significantly different between the ALI groups (all P<0.05). Abnormal cisterns, midline > 5 mm, and subarachnoid hemorrhage were not included in the

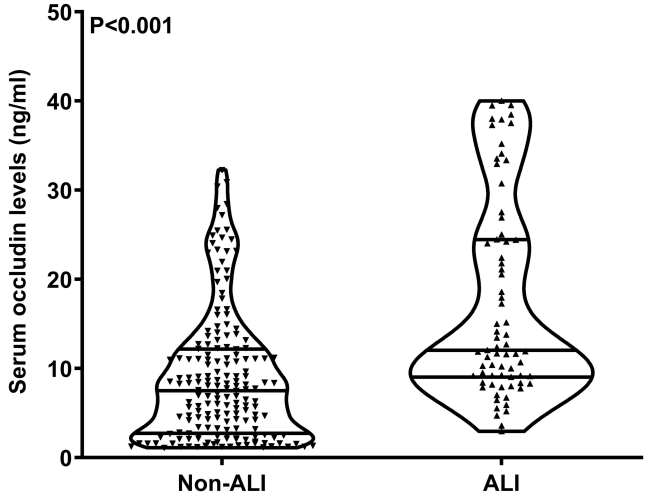

Figure 16 Serum occludin levels between patients experiencing acute lung injury and those not suffering from this condition following severe traumatic brain injury. Occludin levels in the serum of patients diagnosed with severe traumatic brain injury were significantly higher in those with acute lung injury than in those without acute lung injury (P<0.001).
Abbreviation: ALI, acute lung injury.

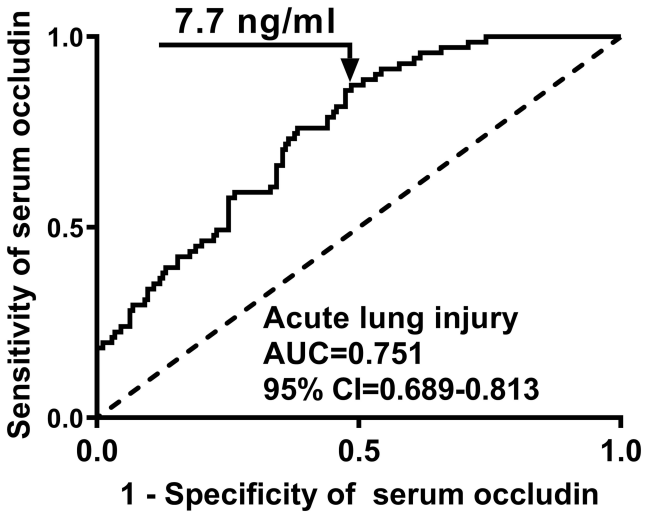

Figure 17 Receiver operating characteristic curve assessing predictive ability of serum occludin levels for acute lung injury secondary to severe traumatic brain injury. Serum occludin levels had a strong discrimination capability for the development of acute lung injury following severe traumatic brain injury, and its optimal value was chosen to be 7.7 ng/mL for forecasting acute lung injury in severe traumatic brain injury with the maximum Youden index. Arrow indicates threshold value of serum occludin levels.
Abbreviations: AUC, area under curve; 95% CI, 95% confidence interval.

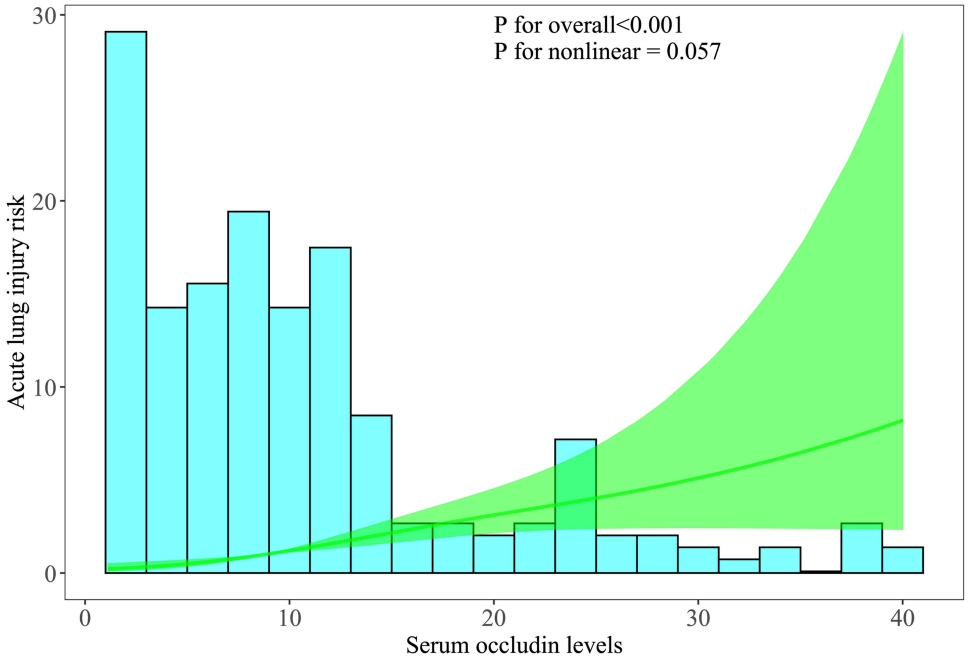

Figure 18 Restricted cubic spline verifying linear correlation of serum occludin levels with likelihood of acute lung injury post-severe traumatic brain injury. Serum occludin levels were linearly correlated with the risk of acute lung injury after severe traumatic brain injury (P for nonlinear > 0.05).

multivariate model because of their contribution to the Rotterdam CT classification. The other factors were added to the binary multivariate model, and subsequently, it was proved that serum occludin levels (OR], 1.058; 95% CI, 1.015–1.102; VIF, 1.778; P=0.008), GCS scores (OR, 0.673; 95% CI, 0.495–0.916; VIF, 1.911; P=0.004), and Rotterdam CT scores (OR, 2.048; 95% CI, 1.334–3.142; VIF, 1.639; P=0.001) kept independently associated with post-sTBI ALI. Age, sex, hypertension, diabetes, alcohol consumption, and cigarette smoking did not significantly interact with serum occludin levels in association with ALI (all P for interaction >0.05; Figure 19). The independent predictive variables for ALI were merged to form a combined model for ALI prediction. A nomogram was used to visualize the model (Figure 20). The model showed good stability in ALI prediction (Figure 21), exhibited acceptable clinical validity

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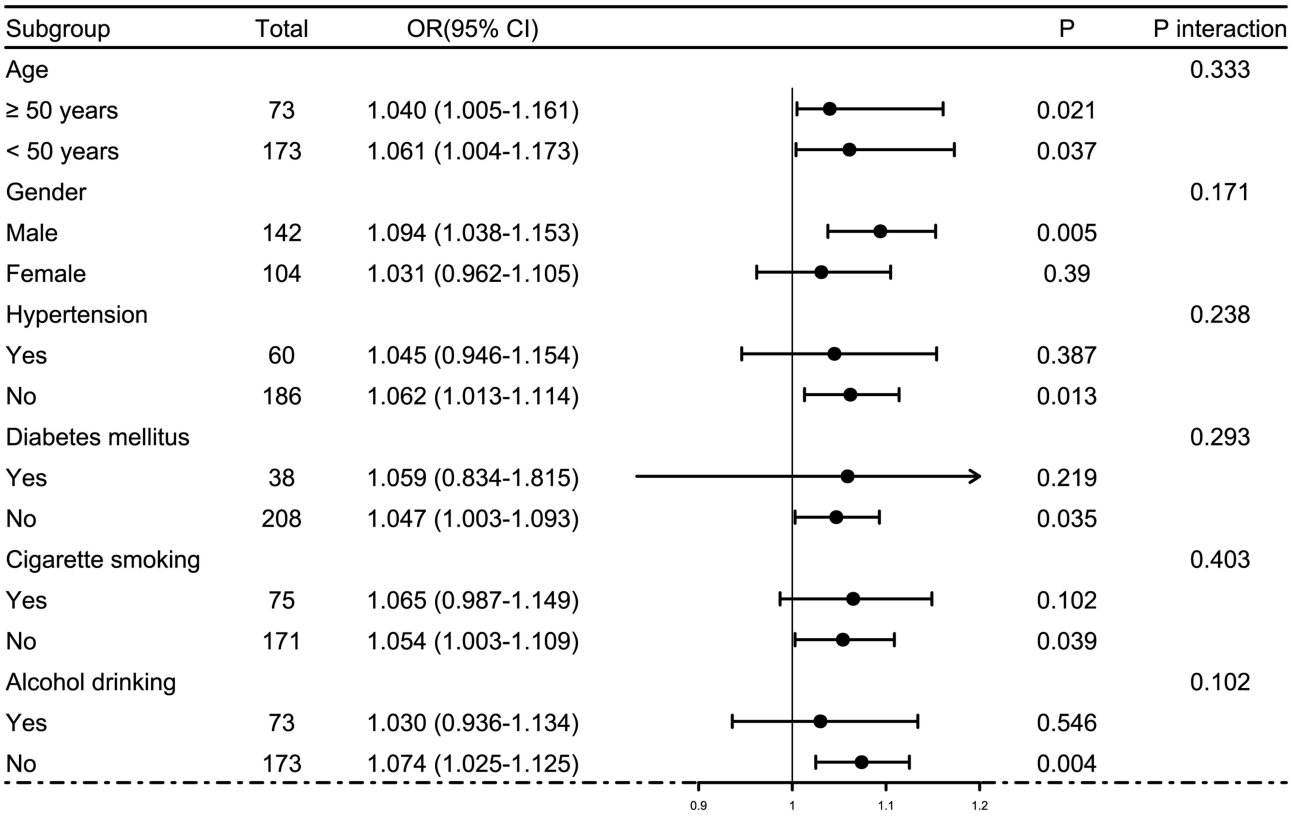

Figure 19 Forest plot describing subgroup assessment of association of serum occludin levels with acute lung injury secondary to severe traumatic brain injury. Serum occludin levels had no significant interactions with the common variables in prognosis prediction after severe brain injury (all P > 0.05).
Abbreviations: OR indicates odds ratio; 95% CI, 95% confidence interval.

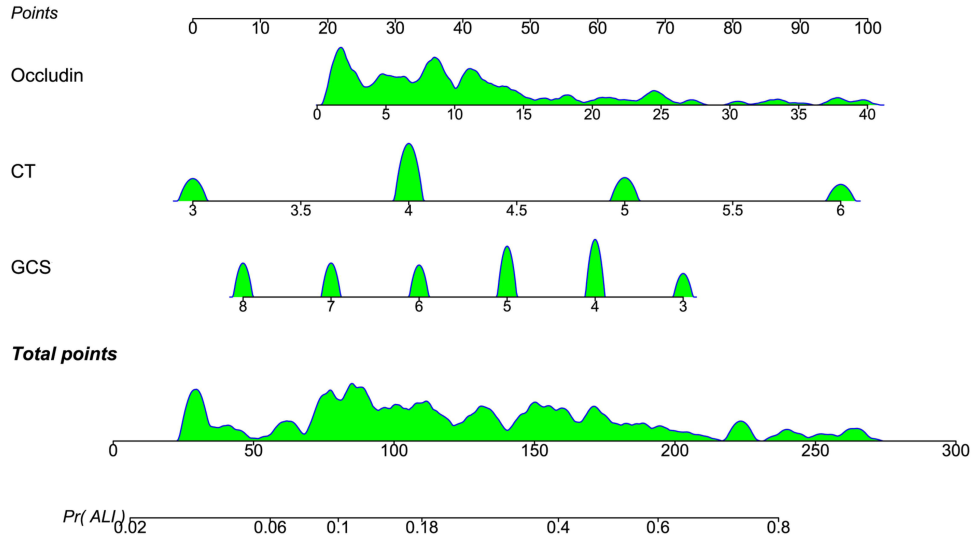

Figure 20 Nomogram outlining combined model for predicting acute lung injury subsequent to severe traumatic brain injury. The model was composed of three independent predictors of acute lung injury following severe traumatic brain injury. Each variable corresponded to its respective score, and the cumulative scores of these factors were related to the possibility of acute lung injury.
Abbreviations: GCS, Glasgow coma scale; CT, Rotterdam computed tomography classification.

(Figure 22), and outperformed any of its three members in ALI prognostication under the ROC curve (all P<0.05; Figure 23). ALI in part mediated association between serum occludin levels and poor prognosis, and its indirect effects constituted 26.0% of the total (Figure 24) with effectiveness of comparative stability (Figure 25).

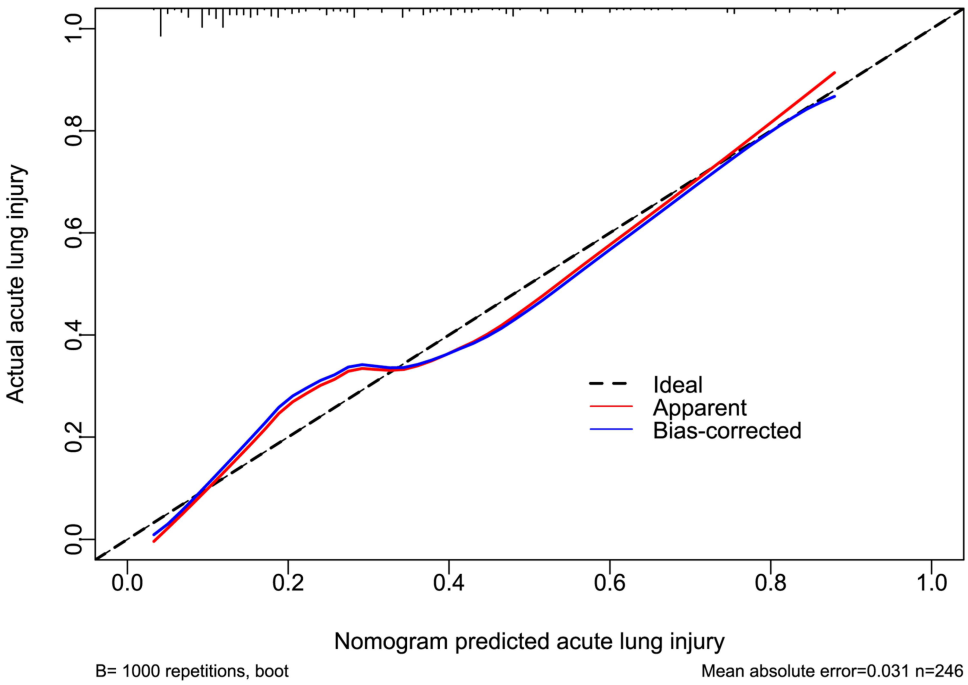

Figure 21 Calibration curve portraying model fit of acute lung injury secondary to severe traumatic brain injury. The model was created using three independent factors of acute lung injury severe brain injury. The model was stably run in the context of severe traumatic brain injury.

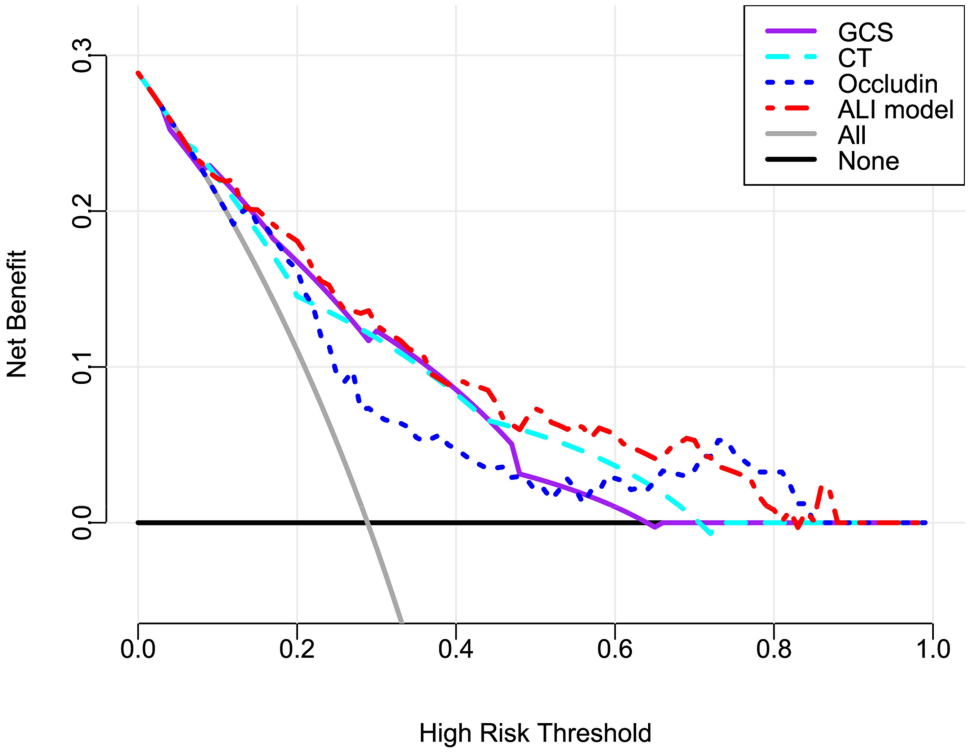

Figure 22 Decision curve depicting clinical benefits of combined model and its components in predicting acute lung injury after severe traumatic brain injury. The model comprises three predictive factors for acute following a severe traumatic brain injury. This model is clinically beneficial for the treatment of severe traumatic brain injury. **Abbreviations**: ALI, acute lung injury; GCS, Glasgow Coma Scale; CT, Rotterdam computed tomography.

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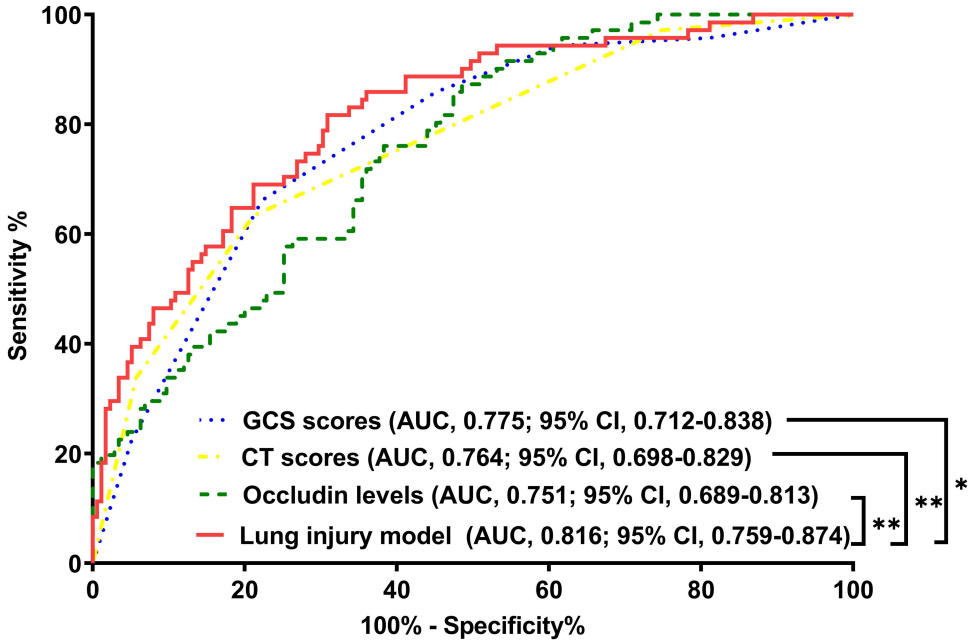

Figure 23 Receiver operating characteristic curve delineating effectiveness of merged model and its members in prognosticating acute lung injury following severe traumatic brain injury. The model was developed by incorporating three predictors of acute lung injury following severe traumatic brain injury. The model had a markedly higher predictive capability than the other three independent predictors (*P<005; **P<0.01).
Abbreviations: GCS, Glasgow Coma Scale; CT, Rotterdam computed tomography classification; AUC, area under curve; 95% CI, 95% confidence interval.

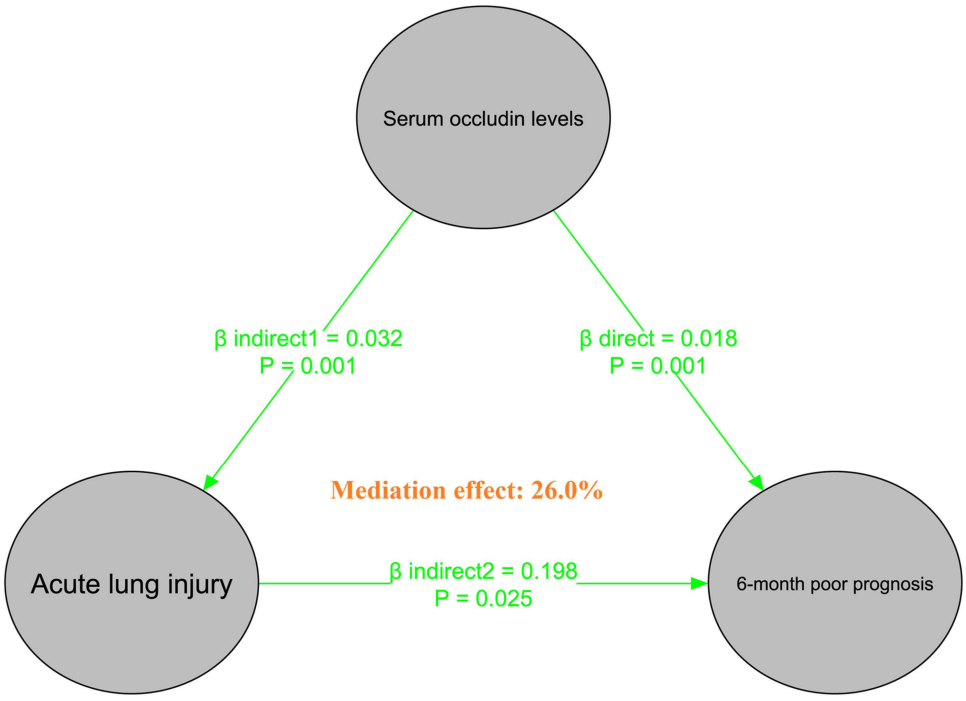

Figure 24 Mediation role of acute lung injury on association of serum occludin levels with poor prognosis after severe traumatic brain injury. Prognosis association of serum occludin levels was partially mediated via acute lung, and its mediation effect accounted for 26.0% of all.

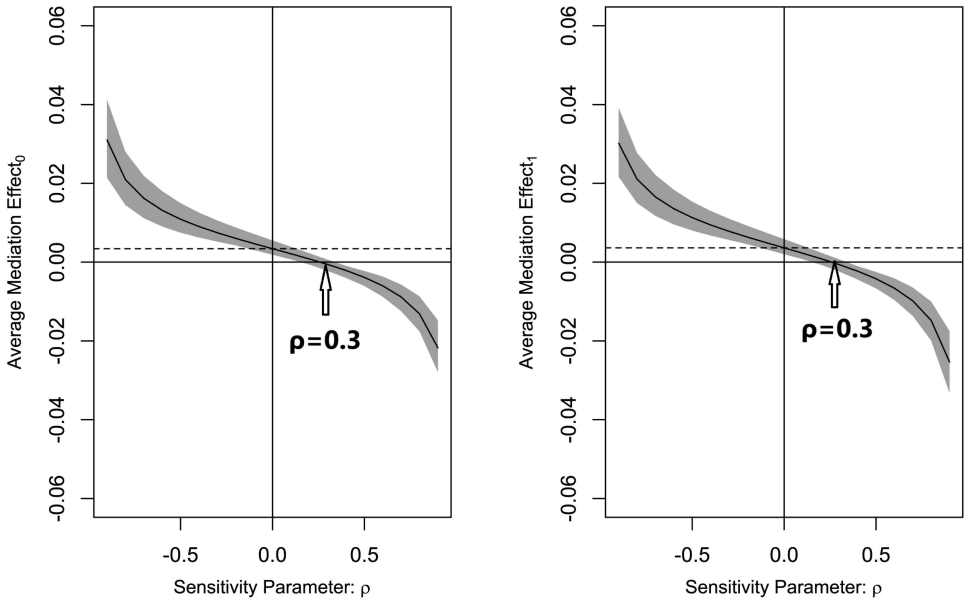

Figure 25 Sensitivity analysis of mediation effect of acute lung injury on prognosis association of serum occludin levels following severe traumatic brain injury. Under both control and treatment conditions, there was statistically satisfactory robustness in the mediation effect of acute lung injury on the prognostic association of serum occludin levels in severe traumatic brain injury, because sensitivity analysis showed that both ρ values amounted to 0.3.

Discussion

To the best of our knowledge, this is the first study to investigate the prognostic implications of serum occludin level in patients with sTBI. First, serum occludin levels were significantly increased after sTBI. Second, serum occludin levels were independently associated with severity indicators such as GCS scores and Rotterdam CT classification. Third, serum occludin levels were independently associated with poor prognosis and ALI. Fourth, serum occludin levels were combined with other independent predictors of poor prognosis and ALI to form two models of satisfactory clinical efficacy. Finally, ALI may mediate the association between serum occludin levels and poor prognosis in this cohort of patients with sTBI. Overall, the above evidence supports the hypothesis that serum occludin may act as a prospective biochemical marker for the assessment of severity and anticipation of prognosis during sTBI management.

Acute brain injury, including sTBI, is characterized by BBB disruption.[29] BBB destruction is attributable to complex processes involving inflammation, oxidation, and amino acid toxicity reactions, and its damage further results in pathophysiological injury to the brain, encompassing cerebral edema, neuronal death, and even neurological impairments.[30] Therefore, maintaining BBB integrity is critical. Occludin, a member of the TJ protein family, is an essential protein that structurally composing the BBB.[14] Under certain pathological circumstances, occludin dissolution or anomalous occludin enhances BBB permeability.[29–32] To date, exploring occludin-targeting medications has become a method for improving BBB permeability, thereby restoring neuronal function and the subsequent recovery of neurological function in acute brain injury diseases.[33–35]

In rats with cerebral ischemia from middle cerebral artery occlusion, blood occludin levels were elevated in a time-dependent fashion, with a prompt enhancement at 4.5 hours following ischemia, which concorded with the loss of occludin from ischemic cerebral microvessels and obvious BBB leakage at 4.5 hours following ischemia.[16] In another similar experiment in rats subjected to middle cerebral artery occlusion, the temporal trends of blood occludin levels were consistent with their loss from ischemic cerebral microvessels.[17] BBB breakdown is strongly linked to the development.[36] In a recent study, serum occludin was verified as an identifiable biomarker of perihematomal edema after human intracerebral hemorrhage in the framework of a multivariate analysis.[37] These data suggest that blood occludin is a potential metric for early BBB damage.

Serum occludin levels were significantly higher in patients diagnosed of intracerebral hemorrhage than in controls,[37] and patients with acute ischemic stroke versus controls with pseudo-strokes had profoundly increased serum occludin

levels.[20] Additionally, blood occludin levels were notably higher in mice with TBI than in those under normal conditions, as well as in 20 patients with TBI than in 20 healthy controls.[29] Consistently, serum occludin levels were substantially elevated after sTBI in this cohort of 246 subjects compared with 100 controls. In response to acute brain injury, occludin from cerebral microvessels can be released into the blood circulation;[16,17] therefore, serum occludin levels may increase, shedding light on the possibility of serum occludin as a prognostic biomarker of acute brain injury.

Regarding the relationship between serum occludin levels and sickness severity in acute brain injury diseases, a study of acute ischemic stroke using univariate analysis showed that serum occludin levels were highly correlated with cerebral infarct volume and National Institutes of Health Stroke Scale (NIHSS) scores.[19] Nonetheless, another study reported that plasma occludin levels were not related to NIHSS scores following acute ischemic stroke.[38] Considering that the former cohort had median NIHSS scores of 14,[19] while the latter cohort had scores of 3,[38] a large severity disparity may be an interpretation of the result inconsistency. In the current study of 246 patients with sTBI, serum occludin levels were independently associated with the GCS and Rotterdam CT scores. Thus, this study may provide a new clue to solidify the concept that serum occludin levels mirror the magnitude of sTBI trauma.

Among two cohorts of patients with acute ischemic stroke, serum occludin levels were reported to be independently associated with several outcome variables such as early neurological deterioration, hemorrhagic transformation, and poor ninety-day prognosis.[19,20] In our study of sTBI humans, post-trauma six-month neurological prognosis indicated by continuous GOS, binary GOS, and ordinal GOS scores, together with ALI, were designated as the outcome variables of interest. Intriguingly, serum occludin levels were independently associated with all the outcome variables. Furthermore, restricted cubic splines were plotted, and subgroup analyses were performed to enhance the robustness of the results that serum occludin may have the opportunity to be a prognostic indicator of poor prognosis and ALI subsequent to sTBI.

Serum occludin levels, along with NIHSS scores, were the two independent predictors of hemorrhage transformation following reperfusion in patients with acute ischemic stroke, and the two indicators were combined to create a model with a markedly higher area under the ROC curve over both.[39] In another study of acute ischemic stroke, serum occludin levels combined with other clinical risk factors, such as the Alberta Stroke Program Early CT scores and endovascular treatment, which were all independent predictors of hemorrhagic transformation, substantially enhanced the accuracy of the predictive effect.[40] In our study, the four independent predictors of poor prognosis, namely, serum occludin levels, GCS scores, Rotterdam CT scores and ALI, were merged to build a prognosis model; analogously, the three independent predictive factors of ALI, ie, serum occludin levels, GCS scores and Rotterdam CT scores, to establish an ALI model. The two models were validated with a good fit, satisfactory clinical validity, and high predictive efficiency using several statistical tools. Taken together, serum occludin may be an auxiliary prognosticator of sTBI with an increasing ability to predict ALI and neurological function.

Occludin strongly preserves alveolar barrier integrity, with extensive distribution in alveolar cells,[21,22] reinforcing serum occludin as a possible indicator of ALI. ALI has been shown to be a determinant of poor prognosis following sTBI, and concurrently, serum occludin levels have been found to be an independent predictor of ALI. ALI may be a link between serum occludin levels and poor prognosis following sTBI. ALI partially mediated the association between serum occludin levels and poor prognosis in patients with sTBI in the present statistics. In other words, ALI may elucidate the link between serum occludin levels and a poor prognosis after sTBI. However, the intricate interplay between these factors warrants further investigation.

Several limitations or prospects must be mentioned here. First, although the potential clinical utility of serum occludin as a prognostic biomarker of sTBI may be recognized, clinical implementation challenges, such as assay standardization and cost- effectiveness, must be overcome in future. Second, partial mediation of ALI on prognostic association of serum occludin levels were found in this host of patients with sTBI, while the limitations of mediation analysis should be acknowledged given possible existence of inadequately large sample size in observational data could affect robustness of results by mediation analysis. Third, serum occludin levels were gauged merely at admission and therefore their temporal trends were not reported in the present study. To finish this work in future may be informative in clinical practice. Finally, the current study had an adequate sample size based on statistical analysis, but it is a single-cohort observational analysis after all; so, before routine clinical application of serum occludin as a prognostic biochemical indicator of sTBI, a larger

and even multi-center cohort study with diverse populations and settings is in future necessitated for the sake of better enhancing reliability and scientificity of the conclusions.

Conclusions

There was a notable increase in serum occludin levels following sTBI, which was independently associated with severity, ALI, and six-month poor prognosis. Moreover, the association between serum occludin levels and poor prognosis is partly mediated by ALI. In summary, serum occludin level may be a prognostic predictor of clinical prospects, and ALI may partially explain the association between serum occludin level and poor prognosis after sTBI.

Data Sharing Statement

The datasets generated and/or analyzed during the current study are not publicly available because they are personal data, but are available from the corresponding author upon reasonable request.

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Disclosure

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References

1. Khan SI, Karim R, Khan SI, et al. Factors affecting severity and prognosis of traumatic brain injury among Bangladeshi patients: an institution based cross-sectional study. *Traffic Inj Prev*. 2024:1–9. doi:10.1080/15389588.2024.2363470
2. Capizzi A, Woo J, Verduzco-Gutierrez M. Traumatic brain injury: an overview of epidemiology, pathophysiology, and medical management. *Med Clin North Am*. 2020;104(2):213–238. doi:10.1016/j.mcna.2019.11.001
3. Kursancew ACS, Faller CJ, Bortoluzzi DP, et al. Neuroinflammatory response in the traumatic brain injury: an update. *Neurochem Res*. 2024;50(1):64. doi:10.1007/s11064-024-04316-4
4. Podell JE, Morris NA. Traumatic brain injury and traumatic spinal cord injury. *Continuum*. 2024;30(3):721–756. doi:10.1212/CON.0000000000001423
5. Chacón-Aponte AA, Durán-Vargas ÉA, Arévalo-Carrillo JA, et al. Brain-lung interaction: a vicious cycle in traumatic brain injury. *Acute Crit Care*. 2022;37(1):35–44. doi:10.4266/acc.2021.01193
6. Bodien YG, Barra A, Temkin NR, et al. Diagnosing level of consciousness: the limits of the Glasgow Coma Scale total score. *J Neurotrauma*. 2021;38(23):3295–3305. doi:10.1089/neu.2021.0199
7. Mohammadifard M, Ghaemi K, Hanif H, Sharifzadeh G, Haghparast M. Marshall and Rotterdam computed tomography scores in predicting early deaths after brain trauma. *Eur J Transl Myol*. 2018;28(3):7542. doi:10.4081/ejtm.2018.7542
8. Lu J, Marmarou A, Lapane KL. IMPACT Investigators. Impact of GOS misclassification on ordinal outcome analysis of traumatic brain injury clinical trials. *J Neurotrauma*. 2012;29(5):719–726. doi:10.1089/neu.2010.1746
9. Luo LF, Weng JF, Cen M, et al. Prognostic significance of serum translocator protein in patients with traumatic brain injury. *Clin Chim Acta*. 2019;488:25–30. doi:10.1016/j.cca.2018.10.035
10. Yan T, Shan H, Wang Z, et al. Temporal change of serum xanthine oxidase levels and its relation to clinical outcome of severe traumatic brain injury: a prospective cohort study. *Neurosurg Rev*. 2023;46(1):320. doi:10.1007/s10143-023-02233-8
11. Jin C, Huang X, Hu Y, Xu B, Ma J. A prospective longitudinal cohort study of serum stanniocalcin-1 as a potential prognostic biomarker of severe traumatic brain injury. *Ther Clin Risk Manag*. 2024;20:341–361. doi:10.2147/TCRM.S463955
12. Zapata-Acevedo JF, Mantilla-Galindo A, Vargas-Sánchez K, González-Reyes RE. Blood-brain barrier biomarkers. *Adv Clin Chem*. 2024;121:1–88. doi:10.1016/bs.acc.2024.04.004
13. Fanning AS, Mitic LL, Anderson JM. Transmembrane proteins in the tight junction barrier. *J Am Soc Nephrol*. 1999;10(6):1337–1345. doi:10.1681/ASN.V1061337
14. Petty MA, Lo EH. Junctional complexes of the blood-brain barrier: permeability changes in neuroinflammation. *Prog Neurobiol*. 2002;68(5):311–323. doi:10.1016/s0301-0082(02)00128-4
15. Shi S, Cheng J, Zhang C, et al. Peripheral blood occludin level as a biomarker for perioperative cerebral edema in patients with brain tumors. *Dis Markers*. 2020;2020:8813535. doi:10.1155/2020/8813535
16. Pan R, Yu K, Weatherwax T, Zheng H, Liu W, Liu KJ. Blood occludin level as a potential biomarker for early blood brain barrier damage following ischemic stroke. *Sci Rep*. 2017;7:40331. doi:10.1038/srep40331

17. Shi S, Qi Z, Ma Q, et al. Normobaric hyperoxia reduces blood occludin fragments in rats and patients with acute ischemic stroke. *Stroke*. 2017;48(10):2848–2854. doi:10.1161/STROKEAHA.117.017713
18. Ren W, Jing G, Shen Q, et al. Occludin and connexin 43 expression contribute to the pathogenesis of traumatic brain edema. *Neural Regen Res*. 2013;8(29):2703–2712. doi:10.3969/j.issn.1673-5374.2013.29.002
19. Li W, Yuan S, Sui X, et al. Higher serum occludin after successful reperfusion is associated with early neurological deterioration. *CNS Neurosci Ther*. 2022;28(7):999–1007. doi:10.1111/cns.13830
20. Li W, Qi Z, Kang H, et al. Serum occludin as a biomarker to predict the severity of acute ischemic stroke, hemorrhagic transformation, and patient prognosis. *Aging Dis*. 2020;11(6):1395–1406. doi:10.14336/AD.2020.0119
21. Lin X, Bai H, Barravecchia M, et al. Occludin is essential to maintain normal alveolar barrier integrity and its protective role during ARDS progression. *Int J Mol Sci*. 2024;25(21):11595. doi:10.3390/ijms252111595
22. Liu M, Gu C, Wang Y. Upregulation of the tight junction protein occludin: effects on ventilation-induced lung injury and mechanisms of action. *BMC Pulm Med*. 2014;14:94. doi:10.1186/1471-2466-14-94
23. Wang KK, Yang Z, Zhu T, et al. An update on diagnostic and prognostic biomarkers for traumatic brain injury. *Expert Rev Mol Diagn*. 2018;18(2):165–180. doi:10.1080/14737159.2018.1428089
24. Ghaith HS, Nawar AA, Gabra MD, et al. A literature review of traumatic brain injury biomarkers. *Mol Neurobiol*. 2022;59(7):4141–4158. doi:10.1007/s12035-022-02822-6
25. Singh AK, Asif S, Pandey DK, Chaudhary A, Kapoor V, Verma PK. Biomarkers in acute traumatic brain injury: a systematic review and meta-analysis. *Cureus*. 2024;16(6):e63020. doi:10.7759/cureus.63020
26. Carney N, Totten AM, O'Reilly C, et al. Guidelines for the management of severe traumatic brain injury, Fourth Edition. *Neurosurgery*. 2017;80(1):6–15. doi:10.1227/NEU.0000000000001432
27. Bernard GR, Artigas A, Brigham KL, et al. Report of the American-European consensus conference on ARDS: definitions, mechanisms, relevant outcomes and clinical trial coordination. The Consensus Committee. *Intensive Care Med*. 1994;20(3):225–232. doi:10.1007/BF01704707
28. Lalla LT, Czorlich P, Fischer M, et al. Dose-dependent association of hyperoxia and decreased favorable outcomes in mechanically ventilated patients with traumatic brain injury, a retrospective cohort study. *Eur J Trauma Emerg Surg*. 2025;51(1):75. doi:10.1007/s00068-024-02730-5
29. Robinson BD, Tharakan B, Lomas A, et al. Exploring blood-brain barrier hyperpermeability and potential biomarkers in traumatic brain injury. *Proc Bayl Univ Med Cent*. 2020;33(2):199–204. doi:10.1080/08998280.2020.1727706
30. Li W, Pan R, Qi Z, Liu KJ. Current progress in searching for clinically useful biomarkers of blood-brain barrier damage following cerebral ischemia. *Brain Circ*. 2018;4(4):145–152. doi:10.4103/bc.bc_11_18
31. Lochhead JJ, McCaffrey G, Quigley CE, et al. Oxidative stress increases blood-brain barrier permeability and induces alterations in occludin during hypoxia-reoxygenation. *J Cereb Blood Flow Metab*. 2010;30(9):1625–1636. doi:10.1038/jcbfm.2010.29
32. Sugiyama S, Sasaki T, Tanaka H, et al. The tight junction protein occludin modulates blood-brain barrier integrity and neurological function after ischemic stroke in mice. *Sci Rep*. 2023;13(1):2892. doi:10.1038/s41598-023-29894-1
33. Hu Q, Zhang R, Dong X, Yang D, Yu W, Du Q. Huperzine A ameliorates neurological deficits after spontaneous subarachnoid hemorrhage through endothelial cell pyroptosis inhibition. *Acta Biochim Biophys Sin*. 2024;56(4):645–656. doi:10.3724/abbs.2024037
34. Zhang Y, Zhang P, Shen X, et al. Early exercise protects the blood-brain barrier from ischemic brain injury via the regulation of MMP-9 and occludin in rats. *Int J Mol Sci*. 2013;14(6):11096–11112. doi:10.3390/ijms140611096
35. Wen J, Qian S, Yang Q, Deng L, Mo Y, Yu Y. Overexpression of netrin-1 increases the expression of tight junction-associated proteins, claudin-5, occludin, and ZO-1, following traumatic brain injury in rats. *Exp Ther Med*. 2014;8(3):881–886. doi:10.3892/etm.2014.1818
36. Klatzo I. Pathophysiological aspects of brain edema. *Acta Neuropathol*. 1987;72(3):236–239. doi:10.1007/BF00691095
37. Yuan S, Ma Q, Hou C, et al. Association of serum occludin levels and perihematomal edema volumes in intracranial hemorrhage patients. *CNS Neurosci Ther*. 2024;30(3):e14450. doi:10.1111/cns.14450
38. Lasek-Bal A, Kokot A, Gendosz de Carrillo D, et al. Plasma levels of occludin and claudin-5 in acute stroke are correlated with the type and location of stroke but not with the neurological state of patients-preliminary data. *Brain Sci*. 2020;10(11):831. doi:10.3390/brainsci10110831
39. Yuan S, Li W, Hou C, et al. Serum occludin level combined with NIHSS score predicts hemorrhage transformation in ischemic stroke patients with reperfusion. *Front Cell Neurosci*. 2021;15:714171. doi:10.3389/fncel.2021.714171
40. Yuan S, Ma Q, Hou C, et al. The combination model of serum occludin and clinical risk factors improved the efficacy for predicting hemorrhagic transformation in stroke patients with recanalization. *Heliyon*. 2024;10(2):e25052. doi:10.1016/j.heliyon.2024.e25052