Libyan Journal of Applied and Contemporary Sciences
ISSN
E: 3136-3709
L: 3136-3695
Vol 1: No 1

Serum Neuron-Specific Enolase and Associated Biochemical Markers in Traumatic Spinal Cord Injury: A Case-Control Study

Mustafa Alburhan [1]* ; Razzaq.A. Sachit [1]; Ameer AA aldafaay [2]

[1] Department of Medical Chemistry, College of Medicine, Al-Mustansiriyah University, Baghdad, Iraq
[2] Department of Clinical Laboratory Sciences, Faculty of Pharmacy, Al-Turath University, Baghdad, Iraq

Email corresponding author's: dr.mustafa.al_burhan@uomustansiriyah.edu.iq

Received: 22/5/2026 Accepted: 28/6/2026 Published: 30/6/2026

Abstract

Background: Traumatic spinal cord injury (tSCI) is associated with early neural tissue disruption, systemic inflammation, metabolic stress, and variable neurological outcomes. Although neurological examination and imaging remain central to diagnosis, blood-based biomarkers may provide an additional biochemical window into the acute injury state. Neuron-specific enolase (NSE) has been proposed as a candidate marker of neuronal injury; however, its interpretation is strengthened when assessed alongside inflammatory, enzymatic, hematological, and metabolic indicators.**Objective:** This study aimed to evaluate serum NSE and selected biochemical markers in patients with TSCI compared with healthy controls, and to examine their discriminatory performance using comparative statistics, correlation analysis, logistic regression, and receiver operating characteristic (ROC) analysis. **Methods:** This case-control study included 60 patients with clinically and radiologically confirmed TSCI and 60 healthy controls. Blood samples were obtained during the early post-admission period. Serum NSE, creatine kinase (CK), lactate dehydrogenase (LDH), C-reactive protein (CRP), glucose, white blood cell (WBC) count, and hemoglobin (Hb) were analyzed. Group comparisons were performed according to data distribution, and diagnostic discrimination was assessed by ROC analysis using Youden-derived cut-off values. **Results:** The TSCI and control groups were comparable with respect to age, body mass index, and sex distribution. Patients with TSCI demonstrated a distinct acute biochemical profile, with significantly higher NSE, LDH, CRP, CK, WBC count, and glucose levels, together with significantly lower Hb concentration. ROC analysis showed excellent discriminatory performance for all evaluated biomarkers. LDH achieved the highest area under the curve (AUC = 0.997; cut-off >245 U/L; sensitivity 98.3%; specificity 98.3%), followed by NSE (AUC = 0.978; cut-off >30.0 ng/mL; sensitivity 96.7%; specificity 95.0%). CK and CRP also showed strong discrimination, with AUC values of 0.928 and 0.936, respectively. Within the TSCI group, NSE correlated positively with LDH, WBC, CRP, and CK, and negatively with Hb, suggesting that the neurochemical signal occurred within a broader inflammatory and systemic injury response. particularly when interpreted alongside complementary biochemical markers, including LDH, CRP, CK, WBC, Hb, and glucose. **Conclusion**: Serum NSE is a promising neurochemical biomarker for differentiating patients with traumatic TSCI from healthy controls, particularly when interpreted alongside complementary biochemical markers includes LDH, CRP, CK, WBC, Hb, and glucose. However, because the comparison group consisted of healthy individuals rather than trauma controls, the high ROC performance should be interpreted as case-control discrimination rather than evidence of trauma-specific diagnostic accuracy. validation in larger cohorts, trauma-control groups, serial sampling designs, and outcome-linked models is required before routine clinical translation.

Keyword : Traumatic spinal cord injury; Neuron-specific enolase; NSE; LDH; CRP; CK; Biomarkers; ROC analysis; Case-control study.
Doi : https://doi.org/10.64943/ljacs.2026.010106

INTRODUCTION

Traumatic spinal cord injury is one of the most disabling injuries to the nervous system, resulting in motor and sensory impairment, systemic disorders, and high treatment and rehabilitation costs. Its epidemiological burden remains significant worldwide, with traffic accidents and falls being among the most common causes [1-3].

The clinical challenge in this field is not only to prove the existence of the injury, but also to estimate its severity early on, anticipate the accompanying tissue burden, and distinguish between true neurological damage and general systemic responses to trauma. The current practice relies primarily on neurological examination and imaging, which are essential, but there is still a need for easy-to-measure, quick-to-obtain, and repeatable blood biomarkers to support early evaluation, triage, and prognosis [4,5].

In this context, neuro-specific enolase (NSE) has received increased attention because it may reflects neuronal damage, and clinical and experimental studies have shown that its level may rise after central nervous system injuries, including spinal cord injury [4-7].

This issue is particularly important in case-control studies because this design allows for direct comparison of differences between patients and controls, as well as the evaluation of the potential diagnostic value of the biomarker within a clear statistical framework. Nevertheless, there is still a practical gap in the literature as there are only a few studies comparing NSE with an appropriate control group, as well as other more common serum markers, including CRP, WBC, LDH, CK, and Hb. These indicators are then incorporated into regression models and ROC analysis to assess their discriminative power more stringently [5-8].

Aim of study

This study aimed to evaluate the differences in NSE levels and other basic serum markers between TSCI patients and healthy control subjects within a case-control design, while assessing their discriminatory performance using group comparisons, logistic regression, and ROC curve analysis.

METHODS

Study Design and Clinical Setting

This prospective observational controlled study aimed to assess the diagnostic and discriminatory power of some serum neurochemical, inflammatory, enzymatic, hematological and metabolic markers in acute traumatic spinal cord injury (TSCI) patients. This study was carried out in Dr. Saad Al-Witry Neurosciences Hospital and Al-Imamain Al-Kadhimain Medical City, Baghdad, Iraq, from January 2026 to April 2026. All measurements were made according to a standard clinical and laboratory protocol, and the same analytical approach was followed in all study groups.

Study Population and Group Definition

The final analytical cohort included 120 adult participants divided into two groups: 60 patients with acute TSCI and 60 apparently healthy controls. Patients were assigned to the TSCI group when the diagnosis was supported by clinical neurological examination and confirmed by appropriate spinal imaging, including X-ray, computed tomography, and/or magnetic resonance imaging when clinically indicated. Neurological classification was based on the International Standards for Neurological Classification of Spinal Cord Injury

(ISNCSCI). Healthy controls were age- and sex-comparable individuals with no recent trauma, no known neurological disease, and no active inflammatory or major systemic illness.

Eligibility Criteria

Eligible TSCI patients were adults aged 20-60 years who presented with acute spinal cord injury and were available for early venous blood sampling after hospital admission. Control participants were adults within the same age range who had no history of spinal cord injury, brain injury, active inflammatory disease, or clinically relevant systemic disorder likely to influence the investigated laboratory markers.

Exclusion Criteria

Participants were excluded if they had a previous history of spinal cord injury, traumatic brain injury, stroke, pre-existing neurological disease, chronic neurodegenerative disease, diabetes mellitus, chronic hypertension, severe renal or hepatic disease, active infection, autoimmune disease, malignancy, or pregnancy. Patients receiving anticoagulants, antidiabetic medications, antihypertensive medications, corticosteroids, immunomodulatory therapy, or any acute pharmacological intervention likely to alter inflammatory or neural biomarker concentrations before baseline sampling were also excluded. These criteria were applied to reduce potential confounding and to preserve the biological interpretability of the measured serum markers.

Clinical and Neurological Assessment

At presentation, all patients with suspected TSCI were clinically evaluated and neurological examination was performed using standard motor and sensory examination principles. For the case group, the location of injury, neurological signs/symptoms, location of pain or pressure, and category of clinical severity were recorded. Imaging was reviewed to support the clinical diagnosis, define the level of injury and to rule out other neurological conditions. Neurological severity was assessed at admission by a specialist physician according to the ISNCSCI/AIS classification. For the present analysis, patients with AIS grades A or B were categorized as severe, whereas those with AIS grades C or D were categorized as non-severe.

Blood Sampling and Pre-Analytical Processing

All subjects had venous blood samples taken at the beginning of the post-admission period (6-12 hours after the traumatic injury). Venous blood was drawn in a standard manner for serum analysis. Samples were transferred for laboratory processing within approximately 10 minutes of collection. Following the formation of the clot, the samples were centrifuged. The samples were centrifuged at 2400 rpm for 5 minutes, and the serum was separated and kept at -20 C for approximately one month, until batch analysis. Samples were not subjected to freeze–thaw cycles before analysis. Blood samples for complete blood count were collected in EDTA tubes and analyzed in the routine laboratory procedures. Group information was not used in analytical processing of laboratory measurements wherever possible.

Laboratory Measurements

NSE was quantified using a commercially available Human NSE ELISA kit (Elabscience®, Cat. No. E-EL-H1047) and read using a GloMax Discover multimode microplate reader (Promega, USA) at 450 nm. According to the manufacturer, the assay has a detection range of 2.34–150 ng/mL and a sensitivity of 1.4 ng/mL, with intra-assay and inter-assay coefficients

of variation below 10%. NSE concentrations were calculated using the standard calibration curve provided with the assay, and sample dilution was performed when required according to the manufacturer's instructions. A subset of 10 TSCI serum samples was analyzed in duplicate as an internal analytical check, while the remaining samples were measured as single determinations. Serum samples showing evidence of hemolysis were excluded before NSE analysis.

Study Outcomes

The primary outcome was the discriminatory performance of serum NSE and associated biochemical markers in differentiating patients with TSCI from healthy controls. Secondary outcomes included between-group differences in demographic and laboratory variables, correlations between NSE and selected clinical or laboratory parameters within the TSCI group, and the association of individual markers with TSCI status using logistic regression analysis.

Statistical Analysis

Statistical analysis was performed using SPSS version 27 and GraphPad Prism version 10. The distribution of continuous variables was assessed using the Shapiro-Wilk test. Normally distributed variables were summarized as mean +/- standard deviation, whereas non-normally distributed variables were summarized as median and interquartile range. Categorical variables were expressed as frequencies and percentages. Between-group comparisons were performed using the Mann-Whitney U test for non-normally distributed continuous variables, Welch's t-test for normally distributed variables with unequal variance where appropriate, and the Chi-square test for categorical variables. Effect sizes were reported using Cliff's delta for non-parametric comparisons and Cohen's d for parametric comparisons. Correlations between NSE and selected demographic or laboratory variables in the TSCI group were assessed using Spearman's rank correlation coefficient. Receiver operating characteristic (ROC) curve analysis was used to evaluate biomarker discrimination between TSCI patients and controls. The area under the curve (AUC), 95% confidence interval, optimal cut-off value, sensitivity, specificity, and Youden index were reported. The ROC-derived cut-off values were selected and evaluated within the same study dataset and should therefore be considered exploratory and internally derived rather than clinically validated thresholds. Continuous variables were standardized as z-scores before univariate logistic regression analysis, therefore, the reported odds ratios represent the change in odds of TSCI associated with a one standard-deviation increase in each variable. All statistical tests were two-sided, and a p value < 0.05 was considered statistically significant.

Use of Artificial Intelligence Assisted Tools

ChatGPT (OpenAI, San Francisco, CA, USA) was utilized only to help in editing the manuscript in English, correcting grammar, and enhancing sentence clarity when writing the manuscript. The study design, participant recruitment, data collection, laboratory measurements, statistical analysis, generation of research data, interpretation of the results, and formulation of the scientific conclusions did not involve the use of the tool. The author critically reviewed, verified, and revised all AI-assisted text and holds full responsibility of the accuracy, originality and integrity of the manuscript.

Ethical Considerations

The study protocol was reviewed and approved by the Ethics Committee of the Iraqi Ministry of Health, Department of Research, Training, and Development on 12 February 2026 (approval number 48/2026). The study was conducted in accordance with accepted ethical principles for human research. Written informed consent was obtained from all participants or, when a patient was unable to provide consent at presentation, from a legally authorized representative or next of kin. Participant confidentiality was preserved throughout data collection, laboratory analysis, and statistical reporting.

Results

The present analysis compared the demographic characteristics and selected laboratory biomarkers between patients with spinal cord injury and healthy controls. The two groups were similar in age, BMI and sex distribution, minimizing the chance of demographic imbalance contributing to the differences in biomarkers. There were 51 males and 9 females (85.0% and 15.0%, respectively) in the TSCI group and 49 males and 11 females (81.7% and 18.3%, respectively) in the control group. There was no statistically significant difference in sex distribution between the groups (p = 0.807) by Chi-square test. Patients with TSCI, however, showed a different biochemical profile with elevated levels of CK, glucose, WBC, LDH, CRP and NSE and decreased Hb concentration. The pattern indicates that there is a combination of systemic inflammation, tissue injury, metabolic stress, anemia or blood loss due to trauma, and neuronal damage in the TSCI group.

Table 1. Comparison of demographic characteristics and laboratory biomarkers between patients with spinal cord injury (TSCI) and healthy controls

Variable	TSCI (*n* = 60)	Control (*n* = 60)	Statistical test	Effect size	*p* value
Age (years)	32.93 ± 11.77	33.33 ± 6.85	*Mann–Whitney U*	Cliff's δ = -0.09	0.377
BMI (kg/m²)	26.10 ± 3.43	26.20 ± 3.57	*Mann–Whitney U*	Cliff's δ = -0.01	0.937
CK (U/L)	(471.25 [320.40–565.00])	(156.54 [130.93–174.18])	*Mann–Whitney U*	Cliff's δ = 0.86	<0.001
Glucose (mg/dL)	(188.00 [97.75–241.29])	(97.50 [84.75–116.75])	*Mann–Whitney U*	Cliff's δ = 0.58	<0.001
WBC (×10⁹/L)	(15.70 [12.18–20.74])	(6.95 [5.58–8.20])	*Mann–Whitney U*	Cliff's δ = 0.96	<0.001
Hb (g/dL)	(9.70 [8.90–10.50])	(15.00 [14.00–16.00])	*Mann–Whitney U*	Cliff's δ = -0.97	<0.001
LDH (U/L)	(361.60 [325.80–424.80])	(144.00 [119.75–180.75])	*Mann–Whitney U*	Cliff's δ = 1.00	<0.001
CRP (mg/L)	(17.29 [14.85–20.82])	(4.80 [3.26–12.55])	*Mann–Whitney U*	Cliff's δ = 0.87	<0.001
NSE (ng/mL)	(43.28 [42.09–44.89])	(18.30 [15.95–21.36])	*Mann–Whitney U*	44Cliff's δ = 0.8	<0.001

Data are presented as mean ± standard deviation (SD) or median [interquartile range (IQR)] shown in parentheses. Continuous variables were compared using the Mann–Whitney U test. Statistical significance was defined as p < 0.05. Effect sizes for Mann–Whitney U comparisons were reported using Cliff's delta (δ).

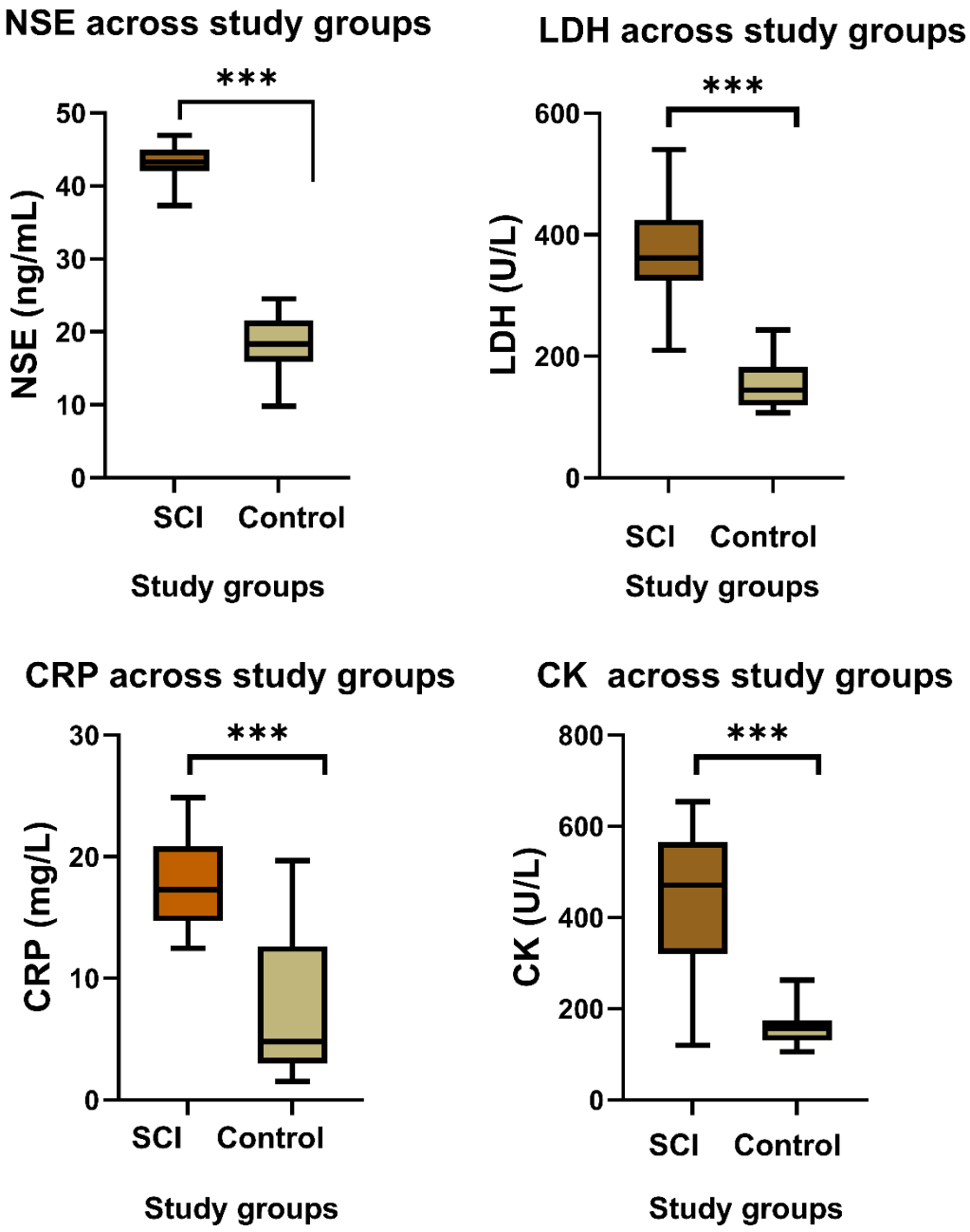

Figure 1. Comparative Distribution of Serum Neurochemical and Inflammatory Biomarkers between TSCI Patients and Healthy Controls

Figure 1. Box-and-whisker plots showing the distribution of serum NSE, LDH, CRP, and CK levels in patients with spinal cord injury (TSCI) and healthy controls. Boxes represent the interquartile range, horizontal lines indicate the median, and whiskers represent the data range. Statistical comparisons between groups were performed using the Mann–Whitney U test. ***$p < 0.001$.

Table 2. Clinical characteristics of the case group

Domain	Category	n	%
Site of injury	Cervical	38	63.3
Site of injury	Thoracic	13	21.7
Site of injury	Lumbar	6	10
Site of injury	Sacral	3	5
Tingling distribution	Hands and feet	40	66.7
Tingling distribution	Feet	20	33.3
Pain/pressure location	Head	23	38.3
Pain/pressure location	Back	21	35
Pain/pressure location	Neck	16	26.7
TSCI severity	Severe	50	83.3
TSCI severity	Non-severe	10	16.7

Shows the clinical distribution within the case group only. Severe TSCI included AIS grades A–B, whereas non-severe TSCI included AIS grades C–D.

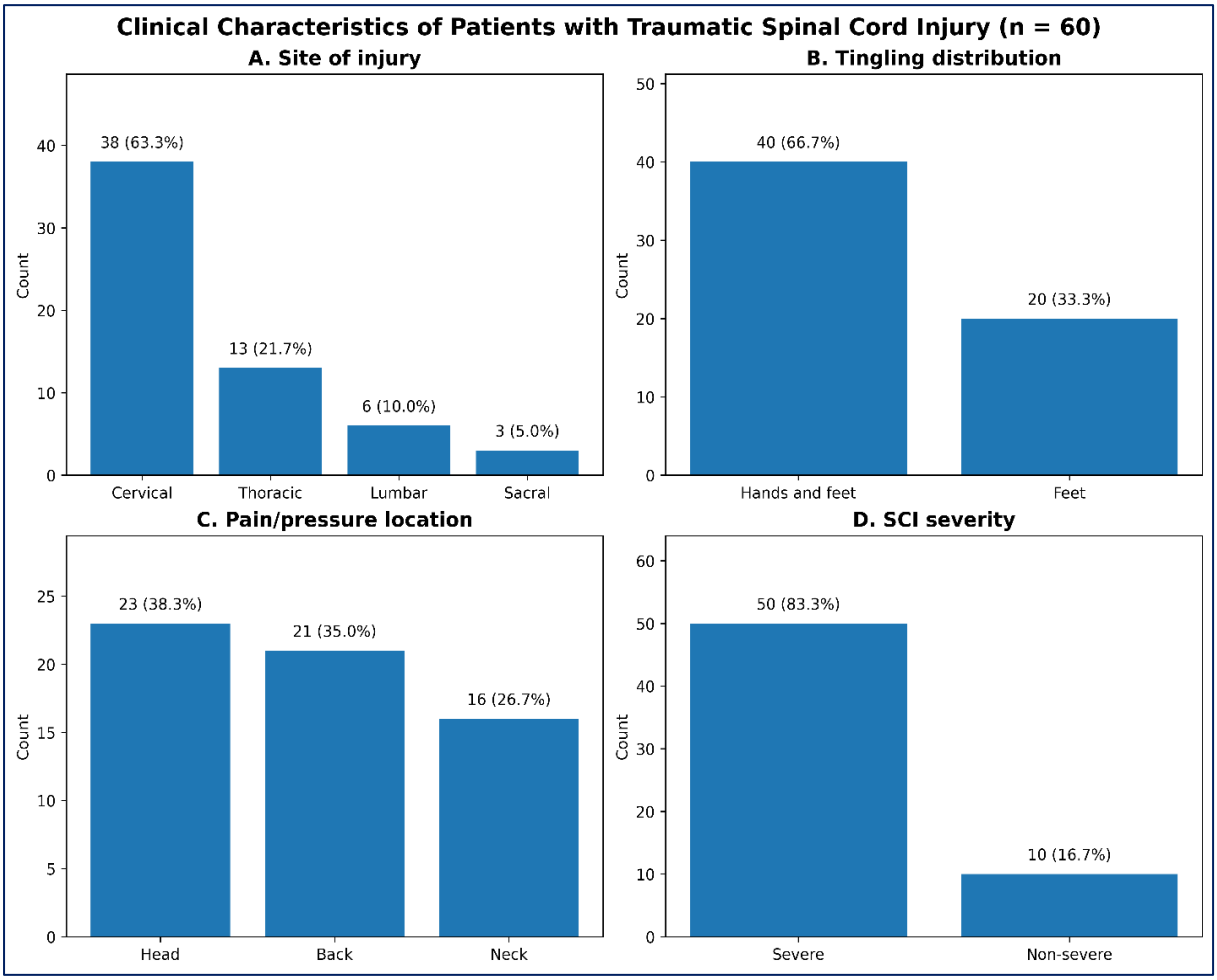

Figure 2. Clinical characteristics of patients with traumatic spinal cord injury (tSCI). Panels show (A) site of injury, (B) tingling distribution, (C) pain/pressure location, and (D) SCI severity. Data are presented as frequencies and percentages (n = 60). No inferential statistical comparisons were performed for these descriptive clinical characteristics.

Table 3. Receiver Operating Characteristic (ROC) Analysis of Serum Biomarkers

Biomarker	AUC	Std. Error	95% CI	cut-off	Sensitivity (%)	Specificity (%)	Youden index	*P* value
CK	0.928	0.028	0.874–0.981	>250 U/L	88.3	91.7	0.8	<0.0001
LDH	0.997	0.0027	0.992–1.00	>245 U/L	98.3	98.3	0.966	<0.0001
CRP	0.936	0.022	0.894–0.978	>13.0 mg/L	86.7	90	0.767	<0.0001
NSE	0.978	0.0099	0.958–0.997	>30.0 ng/mL	96.7	95	0.917	<0.0001

Abbreviations: AUC, area under the receiver operating characteristic curve; CI, confidence interval; CK, creatine kinase; LDH, lactate dehydrogenase; CRP, C-reactive protein; NSE, neuron-specific enolase; TSCI, spinal cord injury. Cut-off values were determined using the Youden index. Sensitivity and specificity are presented as percentages. AUC values closer to 1.0 indicate better discriminatory performance. A two-sided *P* value < 0.05 was considered statistically significant. The reported cut-off values were derived from the present dataset using the Youden index and should be interpreted as exploratory thresholds requiring external validation.

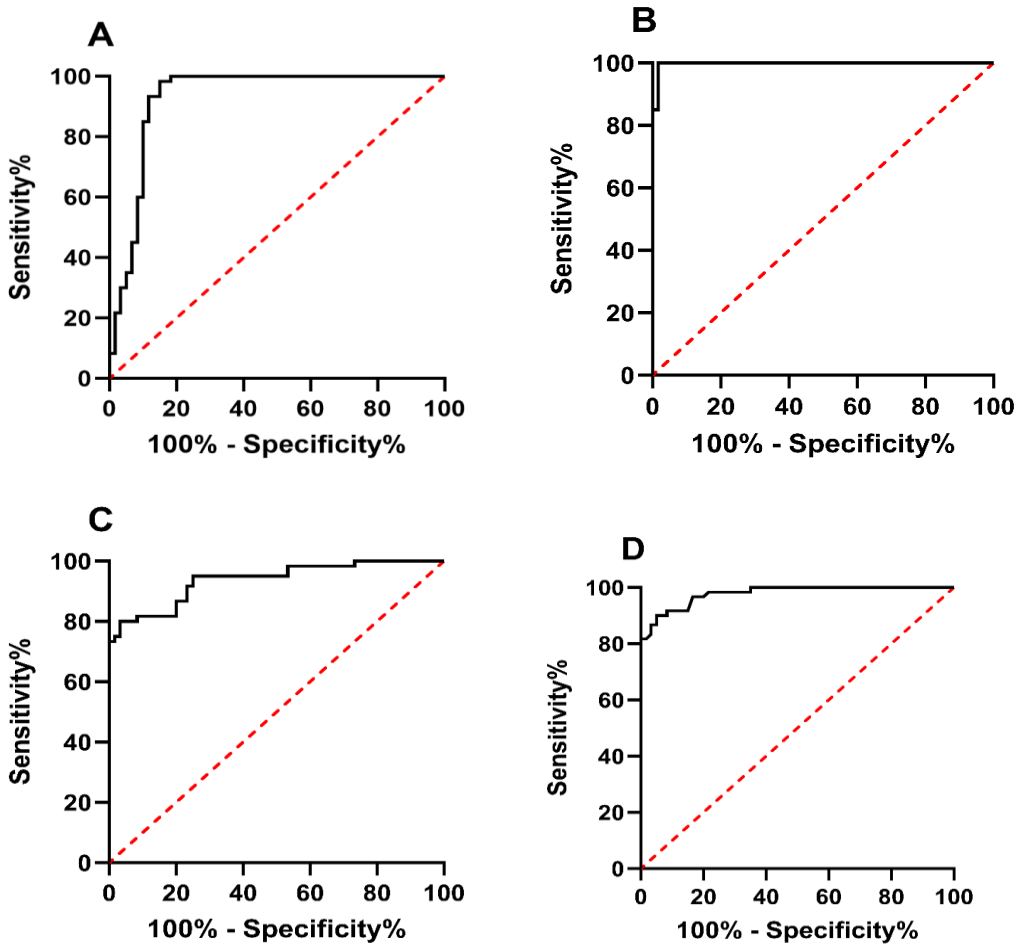

Figure 3. Receiver Operating Characteristic Curves of Serum Biomarkers for Discriminating TSCI Patients from Healthy Controls

Figure 3. Receiver operating characteristic (ROC) curves showing the diagnostic performance of serum biomarkers in differentiating patients with spinal cord injury (TSCI) from healthy controls. The panels represent: A, CK; B, LDH; C, CRP; and D, NSE. The red dashed diagonal line represents the line of no discrimination. Higher areas under the curve indicate better discriminatory performance.

Table 4. Spearman Correlation of Serum NSE with Demographic and Laboratory Parameters in the TSCI Group

Variable	Spearman's rho (ρ) with NSE	Strength of correlation	*p* value
Age	-0.059	Very weak negative	0.660
BMI	0.040	Very weak positive	0.755
CK	0.627	Strong positive	<0.001
Glucose	0.375	Weak positive	0.003
WBC	0.672	Strong positive	<0.001
Hb	-0.697	Strong negative	<0.001
LDH	0.750	Strong positive	<0.001
CRP	0.641	Strong positive	<0.001

Spearman's rank correlation coefficients (ρ) were calculated among patients with spinal cord injury (TSCI). Correlation strength was interpreted according to the absolute value of ρ as follows: very weak (<0.20), weak (0.20–0.39), moderate (0.40–0.59), strong (0.60–0.79), and very strong (≥0.80). A two-sided *p* value < 0.05 was considered statistically significant.

Variable	Age	BMI	CK	Glucose	WBC	Hb	LDH	CRP	NSE
Age	1.000	0.074	-0.118	-0.096	-0.086	0.182	-0.082	-0.091	-0.059
BMI	0.074	1.000	0.112	0.183	0.096	-0.211	0.129	0.141	0.040
CK	-0.118	0.112	1.000	0.344	0.512	-0.579	0.646	0.584	0.627
Glucose	-0.096	0.183	0.344	1.000	0.318	-0.401	0.412	0.398	0.375
WBC	-0.086	0.096	0.512	0.318	1.000	-0.604	0.689	0.631	0.672
Hb	0.182	-0.211	-0.579	-0.401	-0.604	1.000	-0.712	-0.655	-0.697
LDH	-0.082	0.129	0.646	0.412	0.689	-0.712	1.000	0.759	0.750
CRP	-0.091	0.141	0.584	0.398	0.631	-0.655	0.759	1.000	0.641
NSE	-0.059	0.040	0.627	0.375	0.672	-0.697	0.750	0.641	1.000

Figure 4. Spearman correlation matrix showing the relationships among demographic characteristics and laboratory biomarkers in patients with spinal cord injury (TSCI).

Blue colors indicate positive correlations, whereas red colors indicate negative correlations. Color intensity reflects the magnitude of the Spearman correlation coefficient (r), ranging from −1 (strong negative correlation) to +1 (strong positive correlation).

Table 5. Univariate Logistic Regression Analysis of Standardized Variables Associated with Spinal Cord Injury

Variable	Standardized OR	95% CI	*p* value
Age	0.96	0.67–1.37	0.821
BMI	0.98	0.70–1.37	0.905
CK	2.70	1.79–4.07	<0.001
Glucose	1.85	1.25–2.74	0.002
WBC	3.50	2.27–5.39	<0.001
Hb	0.30	0.19–0.47	<0.001
LDH	3.20	2.12–4.83	<0.001
CRP	2.30	1.55–3.40	<0.001
NSE	4.80	3.06–7.53	<0.001

Abbreviations: BMI, body mass index; CK, creatine kinase; WBC, white blood cell count; Hb, hemoglobin; LDH, lactate dehydrogenase; CRP, C-reactive protein; NSE, neuron-specific enolase; OR, odds ratio; CI, confidence interval. Continuous variables were standardized as z-scores before univariate logistic regression analysis. Standardized odds ratios represent the change in odds of TSCI associated with a one-standard-deviation increase in each variable. An OR > 1 indicates a positive association with TSCI status, whereas an OR < 1 indicates an inverse association. All models were univariate and should not be interpreted as demonstrating independent predictive effects. A two-sided p value < 0.05 was considered statistically significant.

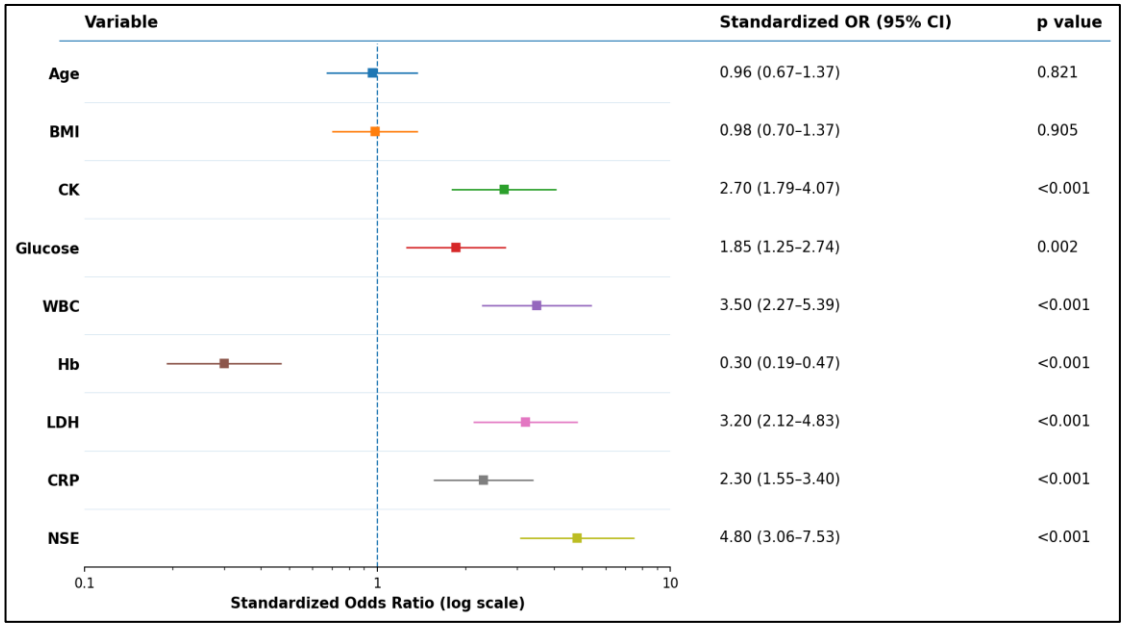

Figure 5. Univariate Standardized Odds Ratios of Demographic and Serum Biomarker Variables Associated with Spinal Cord Injury

Figure 5. Forest plot of univariate logistic regression analysis showing standardized odds ratios (ORs) and 95% confidence intervals (CIs) for variables associated with TSCI status. The dashed vertical line represents the null value (OR = 1), and the x-axis is presented on a logarithmic scale. A two-sided *p* value < 0.05 was considered statistically significant. Standardized ORs represent the change in odds of TSCI associated with a one-standard-deviation increase in each variable.

DISCUSSION

The present case-control study showed that the serum NSE, LDH, CRP, CK, WBC count and glucose levels were significantly elevated and the hemoglobin level was significantly decreased in patients with spinal cord injury as compared to healthy controls. The age, BMI and sex distribution of the two groups were comparable, which further supports the interpretation that the observed laboratory differences are more likely due to the injury state rather than to baseline demographic imbalance. This pattern is biologically feasible in the context of acute TSCI, which is a condition known to involve primary neural tissue disruption, secondary inflammatory cascades, skeletal and soft-tissue trauma, metabolic stress, and systemic physiological burden. The distribution of the case group by sex (male predominance) and the high percentage of cervical injuries are also similar to that seen in global and population-based studies of TSCI [1-3].

Among the studied biomarkers, NSE occupied a central position because of its closer biological relationship to neuronal and neuroendocrine tissue injury. In the current sample, serum NSE was substantially higher in TSCI patients than in controls, and the magnitude of separation between groups was large. This finding supports the concept that disruption of neural elements after spinal cord trauma may be reflected in peripheral blood during the early post-injury period. Previous biomarker literature has emphasized that blood-based markers may complement neurological examination and imaging by providing a measurable biochemical signal of tissue injury, although their clinical usefulness depends on timing, assay reliability, and the clinical comparison group used [4]. NSE has been specifically proposed as a candidate marker in acute central nervous system injury and spinal cord trauma because cellular injury may facilitate its release into extracellular fluid and subsequently into serum [5-8]. Therefore, the present findings are in line with the biological rationale for NSE elevation after TSCI, while also adding value by examining it alongside inflammatory, enzymatic, hematological, and metabolic variables within the same case-control framework.

The ROC analysis confirms the marked biochemical discrimination between TSCI cases and healthy controls. NSE demonstrated excellent discriminatory performance, with a high AUC, sensitivity, and specificity within this case-control comparison. Importantly, these findings should not be interpreted as evidence of trauma-specific diagnostic accuracy. LDH showed the highest AUC in this dataset, approaching perfect discrimination; however, LDH is a nonspecific marker of tissue injury and cellular damage. Its strong performance is therefore likely influenced by the marked biological contrast between acutely injured patients and healthy individuals. NSE has greater biological relevance to neural injury, but its ability to distinguish TSCI from other acute traumatic injuries remains to be established in trauma-controlled cohorts. Independent validation in external trauma cohorts is required before these thresholds can be considered for clinical application.

The very high AUC values found in this study should therefore be interpreted in the context of the design of the investigation. A case-control comparison with healthy controls is suitable to determine if measurable biochemical differences exist between acute TSCI patients and healthy controls, but may overestimate diagnostic performance in routine clinical practice. Healthy controls do not have the inflammatory and enzymatic background of trauma patients without spinal cord involvement. This restriction does not affect the internal findings, but rather sets their boundaries. The present results suggest that there is a strong serum biochemical signature associated with TSCI, but further research is needed to determine if the same markers can differentiate TSCI from other types of trauma, predict neurological outcome, and stratify severity within the injured population. This transition from case-control

discrimination to prognostic and trauma-controlled validation has been emphasised in the TSCI biomarker field many times before [9-11].

The correlation analysis offers another layer of interpretation. In the TSCI group, NSE strong positive correlations with LDH, WBC, CRP, and CK, a weak positive correlation with glucose and strong negative correlation with Hb. These associations indicate that the increase in serum NSE occurs within a broader biological response involving inflammation, tissue injury, and hematological changes. The close association with LDH is especially relevant as it connects the neurochemical signal with a general indicator of cellular injury. The NSE signal is also present in an acute inflammatory environment, as shown by the correlations with WBC and CRP. This does not imply that inflammation is the direct cause of the elevation of NSE, but it does indicate that patients with a greater biological injury burden will have higher values in multiple marker systems. This pattern supports the complementary interpretation of multiple biomarkers rather than reliance on a single laboratory measurement.

The inflammatory component of the findings is clinically relevant. The levels of CRP and WBC were significantly higher in TSCI patients, indicating a possible early systemic inflammatory response after trauma. Although specific to spinal cord tissue, CRP can potentially may reflect the extent of systemic inflammation associated with primary and secondary injury mechanisms. Previous research has correlated early CRP elevations with subsequent clinical evolution following TSCI and with intramedullary changes on MRI [12]. Based on more recent data, it has also been proposed that CRP-derived indices, such as the CRP-to-albumin ratio, may have prognostic value in TSCI [13]. In the current study, the group separation of CRP was good, the AUC was high, and the correlation with NSE was positive. These results confirm its complementary marker status. It should not be used as a biomarker of neural injury, but rather as a useful inflammatory marker to better biologically define the acute state of TSCI.

Also, the haematological results should be noted. Hb was significantly reduced in the TSCI group and strongly negatively correlated with NSE. This observation may be due to trauma-induced blood loss, hemodilution, severity of systemic injury, or early physiological stress. While not specific to TSCI, previous clinical studies have reported anemia during the acute phase following spinal cord injury, and after TSCI [14,15]. The inverse association between Hb and NSE in the present analysis may indicates that patients with a more severe signal of neurochemical injury may also have a more severe systemic injury. This relationship should be viewed with caution as many factors can affect hemoglobin levels, such as bleeding, fluid resuscitation, baseline nutritional status, and associated injuries. However, it is not ignored when developing a wider early assessment model because it is consistently different between groups and it has a correlation pattern.

Both CK and LDH were significantly elevated in the TSCI group, consistent with muscle injury, tissue disruption and cellular damage following trauma. They are likely to be found in a population with acute spinal injury, particularly if there is associated musculoskeletal injury, immobilization, or tissue compression. Their clinical significance is however different from that of NSE. CK and LDH are very good markers of injury burden, but not specific to neuronal damage. The high performance of LDH in the ROC analysis may be due to the marked difference between the acutely injured cases and healthy controls. Thus, LDH is useful in the current model as a component of an injury signature, but should not be interpreted as a specific marker of parenchymal damage to the spinal cord. The same holds true for CK, which was well discriminated and well correlated with NSE, but is a marker with wide tissue origins.

The glucose level was also elevated in the TSCI group. Trauma-induced acute hyperglycemia can result from release of catecholamines, cortisol response, pain, stress, systemic inflammation and metabolic dysregulation. Glucose was moderately correlated with NSE, and significantly associated with TSCI status in the univariate model in the present study. This association is clinically plausible but glucose is a non-specific stress marker. It could be more useful when combined with inflammatory and tissue injury markers. The difference was observed in patients without diabetes and without antidiabetic medication; therefore, the difference is not likely due to known chronic diabetic status, but may be due to stress hyperglycemia or unrecognized metabolic variation.

The logistic regression results were consistent with the descriptive and ROC results. In the standardized univariate analysis showed that NSE, WBC, LDH, CK, CRP, glucose and Hb were significantly associated with TSCI status, whereas age and BMI were not. This pattern is consistent with the comparability of the groups at baseline and with the pathophysiology of acute trauma. The high odds ratio for NSE may indicates that there is a strong association between the increase of NSE and the presence of TSCI in this data set. Concurrently, the importance of WBC, LDH, CRP, CK, glucose, and Hb may indicates that the systemic response is broader for TSCI status. Many of these biomarkers are biologically related and statistically correlated, so multivariable interpretation should be done with caution. Adjustment for inflammatory and tissue injury markers may be influenced by strong collinearity between these markers, leading to wider confidence intervals and apparent independence moving from one marker to another. Therefore, these findings support further evaluation of combined biomarker approaches in future studies rather than a single-marker interpretation. The significance of this study is that it is not an isolated study of NSE, but rather a study that is put into a practical clinical context. The current research on TSCI biomarkers increasingly focuses on panels of neural injury markers, inflammatory markers, and systemic laboratory markers. Different serum and CSF markers have been shown to may reflect various biological aspects of the same injury, such as neuronal damage, glial injury, inflammation, and severity of tissue disruption [16]. Similarly, more general reviews of epidemiology and biomarkers have emphasized the need for reproducible timing, standardized assays, validated cut-offs, and outcome-dependent interpretation of TSCI biomarkers [17,18]. The present study is in this direction as it shows that NSE is a good marker when used in combination with other laboratory markers such as LDH, CRP, CK, WBC, Hb, and glucose and not as a single laboratory marker.

There are a few caveats to be noted. First, the sample size was adequate for an initial case-control biomarker study, but was not large enough to produce stable cut-offs for clinical use. Second, the control group was healthy, not trauma, which could overestimate the apparent diagnostic performance of the nonspecific injury markers (e.g., LDH, CK, CRP, WBC). Third, sampling was done in an early post-admission window; however, no evaluation of biomarker kinetics over time was done. Serial measurements may give a better idea of peak levels, clearance and relationships with secondary injury. Fourth, the study was not powered for detailed subgroup analyses according to neurological severity or injury level. The marked imbalance between severe and non-severe cases and the small numbers in some anatomical injury-level subgroups limited reliable biomarker comparisons across these categories. Future studies with larger and more balanced cohorts should examine the relationships between serum biomarkers, neurological severity, and injury level. Lastly, assay information such as assay characteristics and analytical precision should be clearly reported since the interpretation of biomarkers is highly dependent on the reproducibility of the measurement.

The results have clinical and research significance, despite these shortcomings. The data indicate that there is a measurable serum signature associated with acute TSCI that includes neurochemical, inflammatory, enzymatic, hematological and metabolic components. The NSE seems to be a more relevant marker as it is biologically related to neuronal injury and was very discriminatory between cases and controls. LDH, CRP, CK, WBC, Hb, and glucose provide complementary information regarding systemic injury burden, inflammation, tissue damage, anemia, and stress response. In the clinical setting such a panel may one day be used to facilitate early assessment, triage and risk stratification if validated against trauma controls and clinical outcomes. The findings therefore suggest that NSE may be more informative when interpreted alongside complementary markers of inflammation, tissue injury, hematological change, and metabolic stress.

CONCLUSION

The present study supports the role of serum NSE as a promising neurochemical biomarker for discriminating TSCI cases from healthy controls while showing that its biological interpretation is strengthened when considered alongside LDH, CRP, CK, WBC, Hb, and glucose. The high ROC performance observed in this case-control setting demonstrates clear biochemical separation between the two groups but should not be interpreted as evidence of trauma-specific diagnostic accuracy. Because healthy rather than trauma controls were used, further validation in cohorts including patients with acute trauma without TSCI is required. Serial sampling and assessment of neurological severity and functional recovery will also be important in defining the future clinical utility of these biomarkers.

Statements and Declarations

Funding

The author declares that no funds, grants, or other financial support were received during the preparation, conduct, analysis, or writing of this manuscript.

Competing Interests

The author declares that he has no relevant financial or non-financial interests to disclose.

Figure Originality Statement

All figures included in this manuscript are original and were generated from the original study data and subsequently designed and formatted by the author using Adobe Photoshop. None of the figures has been reproduced, adapted, or modified from any previously published material.

Author Contributions

Mustafa Alburhan conceived the study, developed the main research idea, designed the study protocol, supervised the research work, contributed to data interpretation, prepared the manuscript, and critically revised the final version for important intellectual content .**Razzaq A. Sachit** contributed to participant recruitment, clinical coordination, data collection, and interpretation of the study findings, and critically reviewed the manuscript .**Ameer A. A. Aldafaay** contributed to participant recruitment, clinical assessment, data collection, and coordination of the research procedures, and critically reviewed the manuscript .All authors read and approved the final manuscript and agreed to be accountable for all aspects of the work.

Data Availability

The datasets generated and/or analyzed during the current study are available from the corresponding author upon reasonable request. The data are not publicly available due to participant privacy and confidentiality considerations.

Ethics Approval

This study involved human participants and was conducted in accordance with the ethical principles of the Declaration of Helsinki and its later amendments. The study protocol was reviewed and approved by the Ethics Committee of the Iraqi Ministry of Health, Department of Research, Training, and Development on 12 February 2026, Iraq (approval number: 48/2026).

Acknowledgements

The author would like to acknowledge the medical, laboratory, and administrative personnel that assisted in the clinical and laboratory facets of this study. The author also admits that ChatGPT (OpenAI, San Francisco, CA, USA) was used only to edit the text in English, polish grammar, and enhance the clarity of the text. The author independently reviewed and approved all scientific content, data interpretation, conclusions, and final revisions.

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إنولاز الخلايا العصبية النوعي في المصل والمؤشرات الحيوية المرتبطة به في إصابات الحبل الشوكي الرضية: دراسة حالة-مراقبة

الملخص

الخلفية: ترتبط إصابات الحبل الشوكي الرضية (tSCI) بتلف مبكر في الأنسجة العصبية، والتهاب جهازي، وإجهاد أيضي، ونتائج عصبية متفاوتة. على الرغم من أن الفحص العصبي والتصوير يظلان أساسيين في التشخيص، إلا أن المؤشرات الحيوية في الدم قد توفر نافذة حيوية إضافية لفهم حالة الإصابة الحادة. وقد اقتُرح إنولاز الخلايا العصبية النوعي (NSE) كمؤشر محتمل لتلف الخلايا العصبية؛ ومع ذلك، فإن تفسيره يصبح أكثر دقة عند تقييمه جنبًا إلى جنب مع المؤشرات الالتهابية والإنزيمية والدموية والأيضية. الهدف: هدفت هذه الدراسة إلى تقييم إنولاز الخلايا العصبية النوعي في المصل ومؤشرات حيوية مختارة لدى مرضى إصابات الحبل الشوكي الرضية مقارنةً بمجموعة ضابطة من الأصحاء، ودراسة قدرتها على التمييز باستخدام الإحصاءات المقارنة، وتحليل الارتباط، والانحدار اللوجستي، وتحليل منحنى خصائص التشغيل (ROC). المنهجية: شملت هذه الدراسة 60 مريضًا مصابًا بإصابات الحبل الشوكي الرضية المؤكدة سريريًا وشعاعيًا، و60 شخصًا سليمًا كمجموعة ضابطة. تم الحصول على عينات الدم خلال الفترة المبكرة بعد دخول المستشفى. وجرى تحليل مستويات إنزيم NSE، وإنزيم الكرياتين كيناز (CK)، وإنزيم نازعة هيدروجين اللاكتات (LDH)، والبروتين المتفاعل CRP) C)، والجلوكوز، وعدد خلايا الدم البيضاء (WBC)، والهيموجلوبين (Hb) في المصل. وأُجريت مقارنات بين المجموعات وفقًا لتوزيع البيانات، وقُيِّم التمييز التشخيصي باستخدام تحليل منحنى ROC مع قيم القطع المشتقة من طريقة يودن. النتائج: كانت مجموعتا إصابات الحبل الشوكي الرضية (TSCI) والمجموعة الضابطة متقاربتين من حيث العمر، ومؤشر كتلة الجسم، وتوزيع الجنس. أظهر مرضى إصابات الحبل الشوكي الرضية (TSCI) نمطًا كيميائيًا حيويًا حادًا مميزًا، مع ارتفاع ملحوظ في مستويات إنزيم NSE، وإنزيم LDH، والبروتين المتفاعل C، وإنزيم CK، وعدد خلايا الدم البيضاء، والجلوكوز، بالإضافة إلى انخفاض ملحوظ في تركيز الهيموجلوبين. وأظهر تحليل منحنى ROC أداءً تمييزيًا ممتازًا لجميع المؤشرات الحيوية التي تم تقييمها. حقق إنزيم نازعة هيدروجين اللاكتات (LDH) أعلى مساحة تحت المنحنى (AUC = 0.997؛ القيمة الحدية > 245 وحدة/لتر؛ الحساسية 98.3%؛ النوعية 98.3%)، يليه إنزيم نيورون سبيسيفيك إينولاز (AUC = 0.978) (NSE؛ القيمة الحدية > 30.0 نانوغرام/مل؛ الحساسية 96.7%؛ النوعية 95.0%). كما أظهر كل من إنزيم الكرياتين كيناز (CK) وبروتين سي التفاعلي (CRP) تمييزًا قويًا، بقيم AUC بلغت 0.928 و0.936 على التوالي. ضمن مجموعة إصابات الحبل الشوكي الرضية (TSCI)، ارتبط إنزيم NSE إيجابيًا مع كل من LDH، وعدد كريات الدم البيضاء (WBC)، وCRP، وCK، وسلبيًا مع الهيموجلوبين (Hb)، مما يشير إلى أن الإشارة الكيميائية العصبية تحدث ضمن استجابة التهابية وجهازية أوسع نطاقًا، لا سيما عند تفسيرها جنبًا إلى جنب مع المؤشرات الكيميائية الحيوية التكميلية، بما في ذلك LDH، وCRP، وCK، وعدد كريات الدم البيضاء، والهيموجلوبين، والجلوكوز. الخلاصة: يُعدّ إنزيم NSE في مصل الدم مؤشرًا حيويًا عصبيًا واعدًا للتمييز بين مرضى إصابات الحبل الشوكي الرضية والأفراد الأصحاء، لا سيما عند تفسيره جنبًا إلى جنب مع مؤشرات حيوية كيميائية تكميلية تشمل LDH وCRP وCK وعدد خلايا الدم البيضاء والهيموجلوبين والجلوكوز. مع ذلك، ولأن مجموعة المقارنة كانت تتألف من أفراد أصحاء وليس من مرضى مصابين بإصابات رضية، ينبغي تفسير الأداء العالي لمنحنى ROC على أنه تمييز بين الحالات والضوابط وليس دليلًا على دقة تشخيصية خاصة بالإصابات الرضية. يلزم التحقق من صحة هذا المؤشر في مجموعات أكبر، ومجموعات مقارنة بين المصابين والضابطين، وتصاميم أخذ عينات متسلسلة، ونماذج مرتبطة بالنتائج قبل تطبيقه سريريًا بشكل روتيني.

الكلمات المفتاحية: إصابة الحبل الشوكي الرضية؛ إنزيم NSE؛ LDH؛ CRP؛ CK؛ المؤشرات الحيوية؛ تحليل ROC؛ دراسة الحالات والشواهد.