Received: 27 February 2024 | Revised: 22 April 2024 | Accepted: 29 April 2024
DOI: 10.48309/JMPCR.2024.450111.1160
Journal of Medicinal and Pharmaceutical Chemistry Research
SAMI

FULL PAPER

Correlation of Neuron Specific Enolase protein levels with the severity of traumatic brain injury as measured by the Glasgow Coma scale and marshall classification

Wibowo Artho Sutrisno[a] |Hamzah Hamzah[a] |Dhania Anindita Santosa[a,*] |Prananda Surya Airlangga[a] |Prihatma Kriswidyatomo[a] |Bambang Pujo Semedi[a] |Anggraini Dwi Sensusiati[b] |Atika Atika[c]

[a]*Department of Anesthesiology and Reanimation, Faculty of Medicine, Airlangga University, Dr Soetomo General Hospital, Surabaya, Indonesia*

[b]*Department of Radiology, Faculty of Medicine, Airlangga University, Surabaya, Indonesia*

[c]*Department of Public Health and Preventive Medicine, Faculty of Medicine, Airlangga University, Surabaya, Indonesia*

***Corresponding Author:**
Dhania Anindita Santosa
Email: dhaniasantosa@gmail.com
Tel.: +628123011861

Traumatic Brain Injury (TBI) is a trauma with high morbidity and mortality. The degree of TBI severity so far uses the Glasgow Coma Scale (GCS). Marshall classification of traumatic brain injury from head CT scans has been shown to predict outcome in patients with traumatic brain injury. Neuron Specific Enolase (NSE), a potential biomarker of brain damage for prognostic evaluation, is a structural protein of the central nervous system, which will be regulated to maintain homeostasis in the event of axonal damage. This study aims to find a relationship between Neuron Specific Enolase protein levels and the severity of traumatic brain injury as measured by the Glasgow Coma Scale and Marshall classification. Observational, cross-sectional study with a sample of 51 subjects who were diagnosed with traumatic brain injury and admitted to the emergency room. The subject was resuscitated, a blood sample was taken to check NSE levels and a head CT scan was examined. The research was conducted at Dr Soetomo Hospital, Surabaya from August to October 2023. Fifty-one subjects in this study, consisting of 36 men and 15 women, with an average age of 43 years. The results show that the lower GCS is strongly correlated with the higher NSE value with the correlation test results $r = -0.467$, $p < 0.001$. The results show that the higher the Marshall classification is strongly correlated with the higher the NSE value with the correlation test results $r = 0.475$, $p < 0.001$. There is a significant relationship between NSE levels and GCS levels and also with higher the NSE level, the GCS is decreased. Likewise, there is a significant relationship between the NSE level and the Marshall classification, the higher the NSE level, the higher the Marshall classification.

KEYWORDS

Neuron Specific Enolase; traumatic brain injury; Glasgow Coma Scale; Marshall classification.

Introduction

Traumatic brain injury, or intracranial injury, is an injury to the brain caused by external forces [1]. TBI is classified based on severity (mild, moderate, and severe), mechanism (closed or penetrating), and other features (such as the area of the brain affected or the

name of the brain part) [2]. Mechanical damage that occurs during impact is called primary injury, and then secondary injury occurs where sequential pathological processes occur with delayed clinical symptoms, such as cerebral ischemia and intracranial hypertension, which are sensitive to therapeutic intervention [3,4].

Increasingly advanced technology, both in the world of medicine and biochemistry, has encouraged many discoveries to support clinical trials. Biomarkers of brain damage have been known for a long time and their use is starting to become widespread, one of which is Neuron Specific Enolase (NSE). NSE is a structural protein of the central nervous system, located mainly in the cytoplasm of neurons, which participates in slow axoplasmic transport [5]. Under normal circumstances, NSE is not secreted, but if axons are damaged, NSE is regulated to maintain homeostasis, so it can be used directly to assess functional damage to neurons [6].

The severity of traumatic brain injury is measured based on structural features, loss of consciousness, changes in mental status, post-traumatic amnesia, and the Glasgow Coma Scale, into three categories, namely mild, moderate and severe traumatic brain injury [7,8]. The Glasgow Coma Scale itself is a practical method for assessing decreased consciousness in response to specified stimuli, and is used in clinical practice to this day [9]. Assessment is based on eye opening which is given a score of 1-4 (ranging from not opening the eyes at all, opening after a pressure stimulus, opening after a sound stimulus, and opening spontaneously), verbal reaction with a score of 1-5 (ranging from no verbal reaction at all, only groans, only utters a few words, confused, and speaks fluently), and movement reactions with a score of 1-6 (no movement at all, abnormal extension, abnormal flexion, normal flexion with stimulation, localizes pain, and obeys order) [10].

The Marshall system is a patient selection management based on the severity of non-contrast head CT scan findings. The principle is the degree of swelling and the presence of bruising or bleeding. The classification is: 1) diffuse injury I = no visible intracranial pathology, 2) diffuse injury II = midline shift of the brain up to 5 mm, basal cisterns remain visible, no lesion > 25 cm^3, 3) diffuse injury III = line shift middle of the brain up to 5 mm, basal cisterns compressed or missing, no lesions > 25 cm^3, 4) diffuse injury IV = midline shift of the brain > 5 mm, no lesions > 25 cm^3, 5) mass lesion evacuated V = action has been taken surgery to evacuate the lesion, 6) non-evacuated mass lesion VI = lesion > 25 cm^3, not evacuated surgically [11,12].

Experimental

This study was observational with a cross-sectional design, aiming to determine the comparison between NSE protein levels and the severity of TBI as measured by GCS and Marshall classification. This research was conducted in the emergency room of Dr Soetomo Hospital Surabaya from August 2023 to October 2023. The inclusion criteria were: 1) all TBI patients with any GCS who came to the emergency room, 2) patients aged 17-65 years. Meanwhile, the exclusion criteria are: 1) the patient's family refuses to be included in the study, 2) patients with a history of Alzheimer's disease, DM, melanoma, multiple sclerosis, Down syndrome, and epilepsy.

Patients who met inclusion underwent a GCS examination after initial resuscitation, blood serum samples were taken for NSE examination, and a head CT scan was performed. The blood serum taken was processed in the clinical laboratory at Dr Soetomo Hospital Surabaya, and the NSE levels were checked using the ELISA method. The subject's head CT scan was assessed by a radiologist and his Marshall classification recorded.

Results

Fifty-one subjects were obtained in this study. Subjects were patients diagnosed with traumatic brain injury, either due to traffic accidents, household accidents, or work accidents. Subject characteristics can be seen in Table 1.

TABLE 1 Research characteristics

Characteristics	N	%
Sex		
Male	36	70.6%
Female	15	29.4%
Age (mean±SD) = 43.4±16.0 years		
< 20 years	5	9.9 %
20-29 years	7	13.8 %
30-39 years	10	19.6 %
40-49 years	10	19.6 %
50-59 years	6	11.8 %
60-69 years	13	25.5%
Action		
Craniotomy surgery	32	62.5 %
Non craniotomy surgery	19	37.5 %
End care's outcome		
Survival	36	70.6 %
Non survival	15	29.4 %
Classification of TBI based on GCS		
Mild – moderate (GCS > 8)	27	52.94 %
Severe (GCS ≤ 8)	24	47,06 %
Marshall Classification		
Low – intermediate (Diffuse Injury I – IV)	14	27.5 %
High (Evacuated Mass V – non evacuated mass VI)	37	72.5 %

Blood serum taken from 51 patients was examined for NSE using the ELISA method, and a mean value of 2.62 ng/mL with an SD of 0.89 was obtained, as presented in Table 2.

TABLE 2 NSE research result

Parameter	Mean	Median	Mode	SD	Value Range
NSE (ng/mL)	2.62	2.62	1.68	0.89	1.10- 4.66

The classification of TBI in this study was divided into two categories based on GCS, namely mild-moderate TBI (GCS > 8) and severe TBI (GCS ≤ 8). From these two categories, the Shapiro-Wilk method statistical normality test was carried out and the p-values were 0.685 and 0.828, respectively, indicating that the distribution was normal. The t test for each category shows significant differences (Figure 1), with the mild-moderate TBI category or GCS > 8 (N = 27) having a mean ± SD = 2.25 ± 0.72, while the severe TBI category or GCS ≤ 8 has a mean value ± SD = 3.04 ± 0.89, with a difference test having a p-value of 0.044. The Pearson correlation test for the relationship between NSE levels and TBI severity based on GCS had $r = -0.467$ with $p < 0.05$ (Table 3).

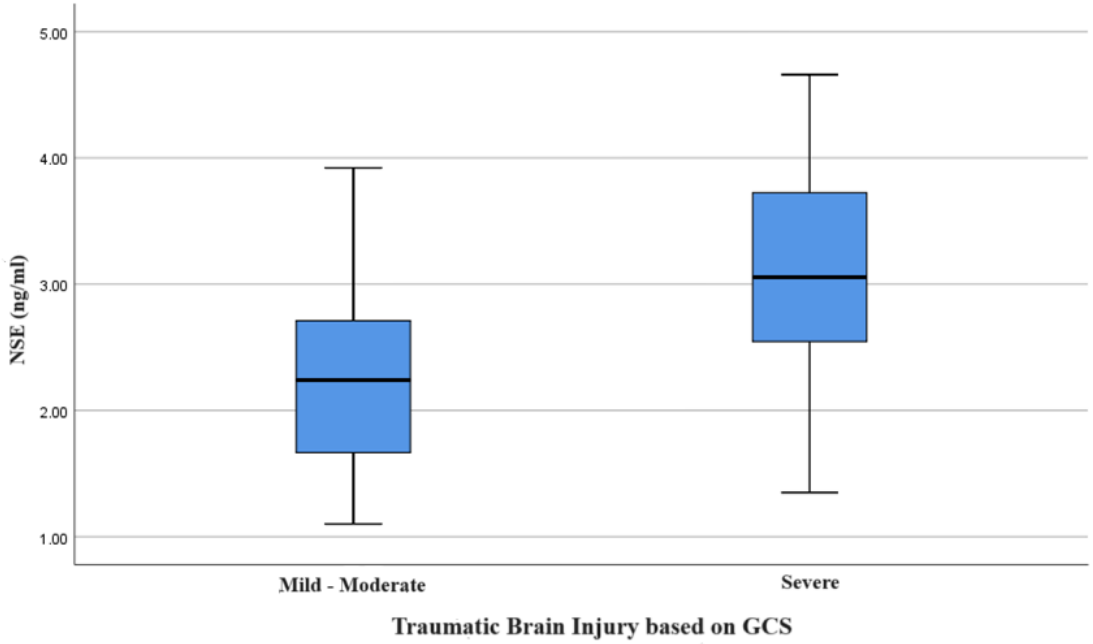

FIGURE 1 Comparison of NSE levels based on the severity of traumatic brain injury from GCS values

TABLE 3 Statistical test for the relationship between NSE levels and GCS

GCS	Mild – moderate TBI	Severe TBI
	Normality test	
P-value (Saphiro-Wilk)	0.685	0.828
	Difference test (p-value 0.044	
Mean ± SD (N)	2.25 ± 0.72 (27)	3.04 ± 0.89 (24)
	Pearson correlation test, R = - 0.467 , p value 0.001	

The TBI classification based on the Marshall classification in this study is divided into two, namely the low- intermediate Marshall classification (diffuse injury I-IV) and the high Marshall classification (evacuated mass V – non evacuated mass VI). From these two categories, the Shapiro-Wilk method statistical normality test was carried out and the p-values were 0.077 and 0.909, respectively, indicating that the distribution was normal. The t test for each category shows significant differences (Figure 2), with the Marshall classification category low - intermediate or diffuse injury I - IV (N = 14) having a mean ± SD = 2.11 ± 0.91, while the High Marshall classification or evacuated mass V – non evacuated mass VI has a mean value ± SD = 2.82 ± 0.82, with a difference test having a p-value of 0.010. The Pearson correlation test for the relationship between NSE levels and Marshall classification has r = 0.475 with $p < 0.05$ (Table 4).

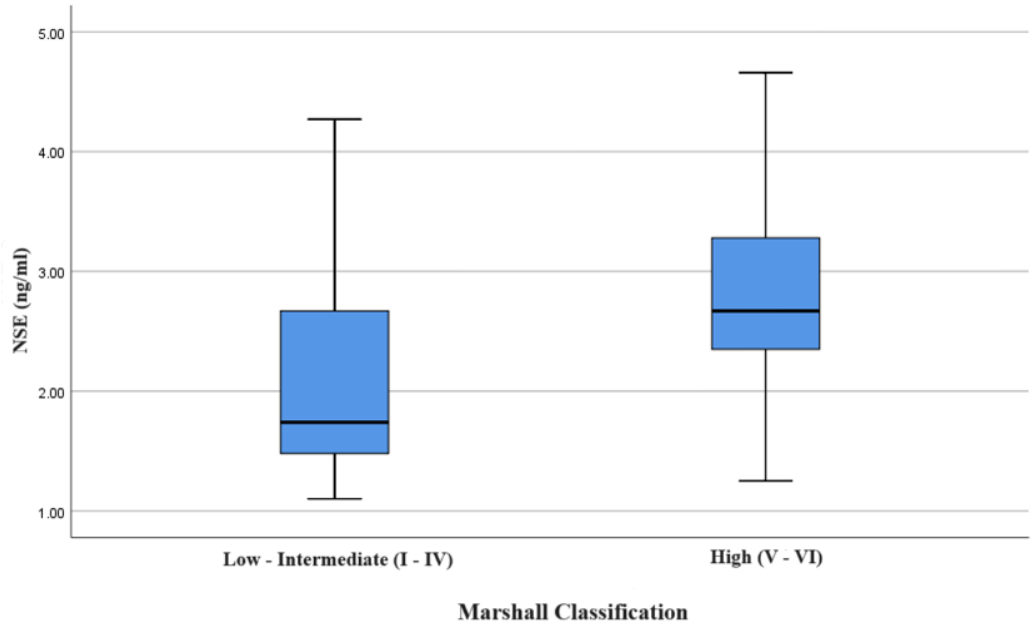

FIGURE 2 Comparison of NSE levels based on Marshall classification

TABLE 4 Statistical test for the relationship between NSE levels and Marshall classification

Marshall Classification	Low - intermediate	High
	Normality test	
p-value (Saphiro-Wilk)	0.077	0.909
	Difference test (p-value 0.010)	
Mean ± SD (N)	2.11 ± 0.91 (14)	2.82 ± 0.82 (37)
Pearson correlation test, R = 0.475 , p value 0.000		

Discussion

Fifty-one subjects in this study showed that there was an increase in NSE levels in traumatic brain injury with levels of 2.62 ng/ml. Cheng's research in 2014 showed an increase in NSE in traumatic brain injury patients with levels of 0.96- 2.32 ng/ml [13].

This study shows that there is a correlation between the NSE level and the patient's GCS at admission (after initial resuscitation), namely the higher the NSE level, the lower the GCS. Zaheer *et al.* found something similar, but in acute ischemic stroke patients, where they concluded that serum NSE levels were useful as a marker for predicting stroke severity and early functional outcomes [14].

Richter et al in 2022 conducted research on patients with injuries resulting from trauma and found that a high Marshall classification was associated with increased NSE levels [15]. This research concludes the same thing, where the higher the NSE level, the higher the Marshall classification. NSE as a protein in the cytoplasm of neuronal cells, and S100B, correlate with radiological measurements of infarct volume in the first week of stroke.

Biomarkers of brain damage have been widely discovered, starting from Neuron Specific Enolase, S100B, Glial Fibrillary Acidic Protein, Myelin Basic Protein, and tau protein [16]. In the early phase of acute stroke, biomarker determination is cheaper and easier to measure tissue damage and evaluate treatment effects than other modalities, such as radiology [17]. The hope for TBI is that biomarkers of brain damage can also show the same thing, especially in improving diagnosis and evaluation.

The limitation of this study is that it did not examine post-procedure NSE and carry out periodic NSE evaluations so that we could see trends in NSE during treatment. Multivariate

analysis could not be carried out because the determining factor for increased NSE was only due to trauma, there were no other variables such as comorbidities, history of previous trauma, type of trauma whether isolated brain injury or accompanied by other extra-cranial injuries.

Conclusion

This study found that there was a significant relationship between NSE levels and GCS in traumatic brain injury, namely that the higher the NSE level, the lower the GCS. The Marshall classification in this study has a significant relationship with the NSE level, namely the higher the NSE level, the higher the Marshall classification. It is hoped that in further research, serial evaluations will be carried out to see trends in NSE values before and after surgery. Multivariate analysis needs to be carried out to see the relationship between various factors and NSE levels, such as comorbid data, type of head trauma, and number of traumas.

Acknowledgements

The authors would like to thank the Emergency Room, Clinical Pathology Unit, and Radiology Unit, in dr Soetomo General Hospital Surabaya.

Funding

There was no specific funding from governmental, commercial, or non-profit entities for this work.

Authors' Contributions

All authors contributed to data analysis, article preparation, and paper revision and have collectively assumed responbility for all aspects of this work.

Conflict of Interests

The authors have stated their absence of any conflicts of interest regarding this study.

Ethics Clearance Statement

This study was approved by the Ethics Committee of Dr. Soetomo Surabaya Hospital with no. 0684/KEPK/VI/2023 which was issued in July 2023.

Orcid:

Wibowo Artho Sutrisno:
https://orcid.org/0009-0008-6781-0389
Hamzah Hamzah:
https://orcid.org/0000-0002-5665-3567
Dhania Anindita Santosa:*
https://orcid.org/0000-0001-7237-3218
Prananda Surya Airlangga:
https://orcid.org/0000-0002-4674-6751
Prihatma Kriswidyatomo:
https://orcid.org/0000-0002-2840-2226
Bambang Pujo Semedi:
https://orcid.org/0000-0003-4499-3481
Anggraini Dwi Sensusiati:
https://orcid.org/0000-0002-9841-1769
Atika Atika:
https://orcid.org/0000-0001-8319-4135

References

[1] D. Padmaja, A. Luthra, R. Mitra, Neurotrauma, *Essentials of Neuroanesthesia*, **2017**, 535-585. [Crossref], [Google Scholar], [Publisher]

[2] C. Werner, K. Engelhard, Pathophysiology of traumatic brain injury, *British Journal of Anaesthesia*, **2007**, *99*, 4-9. [Crossref], [Google Scholar], [Publisher]

[3] S. Saleh, Sinopsis neuroanestesia klinik, Surabaya: Universitas Airlangga Pers, **2023**, 201. [Crossref], [Publisher]

[4] S. Syahrul, I. Imran, N. Fajri, Clinical characteristics of traumatic brain injury patients in Dr. Zainoel Abidin public hospital Banda Aceh, Indonesia, *Bali Medical Journal*, **2020**, *9*, 194-200. [Crossref], [Google Scholar], [Publisher]

[5] B. Stefanović, O. Đurić, S. Stanković, S. Mijatović, K. Doklestić, B. Stefanović, B. Jovanović, N. Marjanović, N. Kalezić, Elevated serum protein S100B and neuron specific

enolase values as predictors of early neurological outcome after traumatic brain injury, *Journal of Medical Biochemistry,* **2017**, *36*, 314-321. [Crossref], [Google Scholar], [Publisher]

[6] M. Alvin, P.S. Airlangga, E. Kusuma, P. Kriswidyatomo, P. Lestari, Y. Setiawati, The role of neuron-specific enolase (NSE) and S100B protein in the incidence of acute postoperative cognitive dysfunction (POCD) in geriatric patients receiving general anesthesia, *Bali Medical Journal*, **2022**, *11*, 1822-1827. [Crossref], [Google Scholar], [Publisher]

[7] N.M. Rehatta, E. Hanindito, A.R. Tantri, Anestesiologi dan Terapi Intensif*, Buku Teks Kati-Perdatin. Gramedia Pustaka Utama*, **2019**. [Google Scholar], [Publisher]

[8] A. Rofik, U. Ambarsari, H.A. Sodali, M. Munir, The relationship between leukocytes numbers and consciousness level of craniotomy patients at The Jemursari Islamic Hospital Surabaya in 2018-2019, *Indonesian Journal of Medical Laboratory Science and Technology*, **2021**, *3*, 81-89. [Crossref], [Google Scholar], [Publisher]

[9] P.S. Airlangga, P. Rahardjo, Buku ajar anestesiologi dan terapi intensif fisiologi pernapasan, *Airlangga University Press*, **2022**. [Google Scholar], [Publisher]

[10] G. Teasdale, Glasgow Coma scale assessment, **2015**. [Publisher]

[11] B. Botz, marshall classification of traumatic brain injury, **2022**. [Publisher]

[12] M.W. Asih, E.D. Martadiani, N.S. Laksminingsih, Y.W. Sulaiman, I.G.A. Trisnawati, Reliability of cerebral edema interpretation on head computed tomography scan in mild and moderate traumatic brain injury, *Bali Medical Journal*, **2021**, *10*, 1067-1069. [Crossref], [Google Scholar], [Publisher]

[13] F. Cheng, Q. Yuan, J. Yang, W. Wang, H. Liu, The prognostic value of serum neuron-specific enolase in traumatic brain injury: systematic review and meta-analysis, *PloS One*, **2014**, *9,* 106680. [Crossref], [Google Scholar], [Publisher]

[14] A. Haque, R. Polcyn, D. Matzelle, N.L. Banik, New insights into the role of neuron-specific enolase in neuro-inflammation, neurodegeneration, and neuroprotection, *Brain Sciences,* **2018**, *8*, 33. [Crossref], [Google Scholar], [Publisher]

[15] S. Richter, E. Czeiter, K. Amrein, A. Mikolic, J. Verheyden, K. Wang, A.I.R. Maas, E. Steyerberg, A. Büki, D.K. Menon, V.F.J. Newcombe, Prognostic value of serum biomarkers in patients with moderate-severe traumatic brain injury, differentiated by Marshall computer tomography classification, *Journal of Neurotrauma*, **2023**, *40*, 2297-2310. [Crossref], [Google Scholar], [Publisher]

[16] W. Chen, G. Wang, C. Yao, Z. Zhu, R. Chen, W. Su, R. Jiang, The ratio of serum neuron-specific enolase level to admission glasgow coma scale score is associated with diffuse axonal injury in patients with moderate to severe traumatic brain injury, *Frontiers in Neurology*, **2022**, *13*, 887818. [Crossref], [Google Scholar], [Publisher]

[17] M. Li, Z. Zhao, G. Yu, J. Zhang, Epidemiology of traumatic brain injury over the world: a systematic review, *General Medicine: Open Access*, **2016**, *4*, 275-275. [Crossref], [Google Scholar], [Publisher]

How to cite this article: Wibowo Artho Sutrisno, Hamzah, Dhania Anindita Santosa, Prananda Surya Airlangga, Prihatma Kriswidyatomo, Bambang Pujo Semedi, Anggraini Dwi Sensusiati, Atika, Correlation of Neuron Specific Enolase protein levels with the severity of traumatic brain injury as measured by the Glasgow Coma scale and marshall classification. *Journal of Medicinal and Pharmaceutical Chemistry Research*, 2024, 6(10), 1512-1518. **Link:** https://jmpcr.samipubco.com/article_195359.html