DIAGNOSTIC VALUE OF pTAU AND NF-L LEVELS IN CEREBROSPINAL FLUID AND PLASMA IN PATIENTS WITH TRAUMATIC BRAIN INJURY

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Abstract

Severe traumatic brain injury (TBI) is among the leading causes of death worldwide, and survivors often suffer permanent cognitive and neurological impairment. Early diagnosis and treatment can greatly improve outcomes and recovery.

The purpose of this study is to investigate the diagnostic value of the biochemical markers pTau and NF-L in the early diagnosis of TBI in patients by analysing their concentration in cerebrospinal fluid and blood plasma.

The study includes 20 cases of patients who suffered severe TBI. Within 24 hours of the injury, cerebrospinal fluid and plasma were collected and compared to identical samples obtained from a control group of 11 non-traumatic cadavers.

The results from the present study show significant rise in cerebrospinal pTau and plasma NF-L levels in the control group, and insignificant rise of the levels of plasma pTau in TBI patients. In conclusion, the investigated molecules pTau and NF-L are not reliable for early diagnosis and monitoring of TBI.

Key words: *biomarkers, brain injury, TAU, NF, TBI*

Introduction

Traumatic brain injury (TBI) is a condition that usually occurs as a result of a direct blow to the head, penetrating injuries to the cranial cavity, or acceleration-deceleration forces [1, 2, 3, 4]. Severe TBI are among the leading causes of death worldwide, and survivors often suffer permanent cognitive and neurological impairment [5, 6]. Early diagnosis and treatment can greatly improve outcomes and recovery. Currently, TBI is diagnosed based on clinical findings, Glasgow Coma Scale (GCS), neurological examination, and computed tomography (CT) imaging. Each of them has its limitations and problems. GCS is an effective way to assess neurological status, but factors such as the patient's use of alcohol and drugs, as well as the presence of trauma to other parts of the body, can affect its accuracy. On the other hand, CT is rarely an effective method for proving mild to moderate TBI, exposes patients to ionizing radiation and is a relatively expensive diagnostic method [7, 8]. Modern medicine strives to optimize diagnostic methods and minimize invasive and harmful diagnostic methods. It is for this reason that, in the last decade, the attention has been focused on research related to the isolation of biochemical indicators with statistical significance in the diagnosis and prediction of brain injuries [9, 10, 11, 12, 13, 14]. Among these are Tau-protein and neurofilaments (NF).

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The aim of the present study is to investigate the value of the biochemical markers Tau-protein and NF-L in the early diagnosis of TBI by examining their concentration in cerebrospinal fluid and plasma in patients with severe traumatic brain injury.

Materials and methods

The study includes 20 cases of adult patients with isolated severe traumatic brain injury admitted for treatment at the Anesthesiology and Intensive Care Clinic of University Multi-profile Hospital for Active Treatment "St. George" Plovdiv. After obtaining written informed consent from the patients and their relatives, samples of cerebrospinal fluid obtained from the lateral ventricles as well as plasma from peripheral venous blood were collected from each of them. Samples were collected 24 hours after injury.

As a control group, 11 people who died as a result of non-traumatic causes (drowning, hanging and cardiovascular accidents) were selected, subject to a forensic medical examination in the Department of Forensic Medicine of University Multi-profile Hospital for Active Treatment "St. George" Plovdiv. From controls, CSF samples were collected from the lateral ventricles, as well as blood plasma from the subclavian arteries. The samples were collected before the 24th hour after death.

The concentration of pTau and NF-L in the cerebrospinal fluid and blood plasma of the cases and the control group was investigated using the ELISA method. The obtained results were processed by SPSS v26.0 statistical analysis software and subjected to the Mann-Whitney U-test.

Results

The studied group of patients consisted of 20 victims of closed craniocerebral trauma caused by a fall, traffic accident or beating. Of them, 4 are female and 16 are male. The control group consisted of 11 who died as a result of drowning, hanging, or cardiovascular accident. 3 of them are female and 9 are male. The average age for the patients who died with a brain injury was 51 ± 18 years, and for the control group the average age was 64 ± 18 years.

To assess the difference in the concentration of pTau and NF-L in CSF and plasma, the Mann-Whitney U Test was performed. Results were visualized using histograms.

The test showed statistically significant differences in the concentration of pTau in cerebrospinal fluid in the control group (Median=0.000, n= 11) and patients with traumatic brain injury (Median= 0.000, n=20), U= 60 , z= 3.224, p= 0.001.

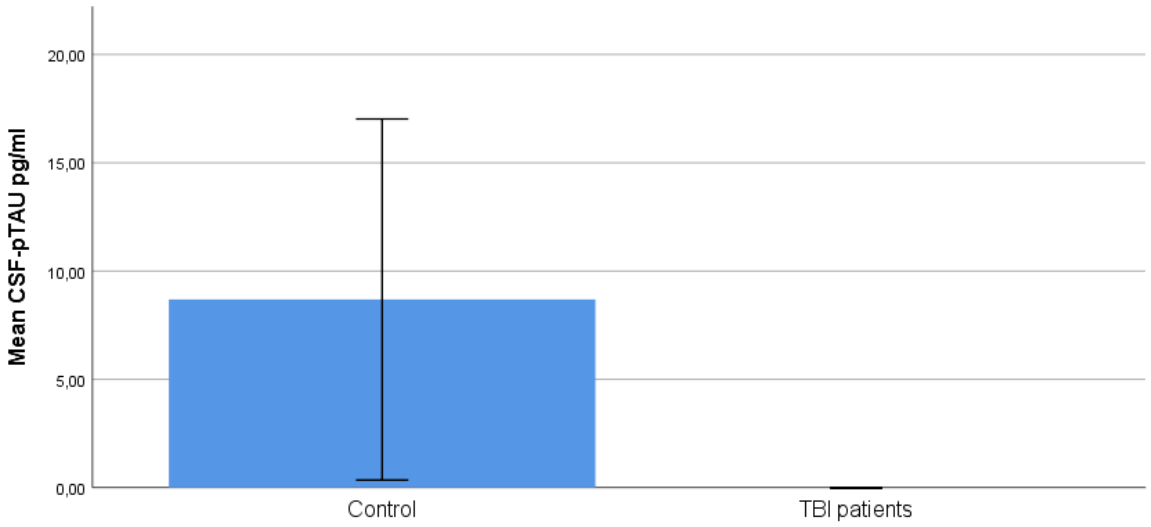

Figure 1 Graphic representation of the concentration of pTAU in the cerebrospinal fluid of the control and study groups.

There were insignificant differences in the concentration of pTau in plasma in the control group (Median=0.000, n= 11) and patients with craniocerebral trauma (Median= 0.000, n=20), U= 104.5 , z= 0.743, p= 0.458.

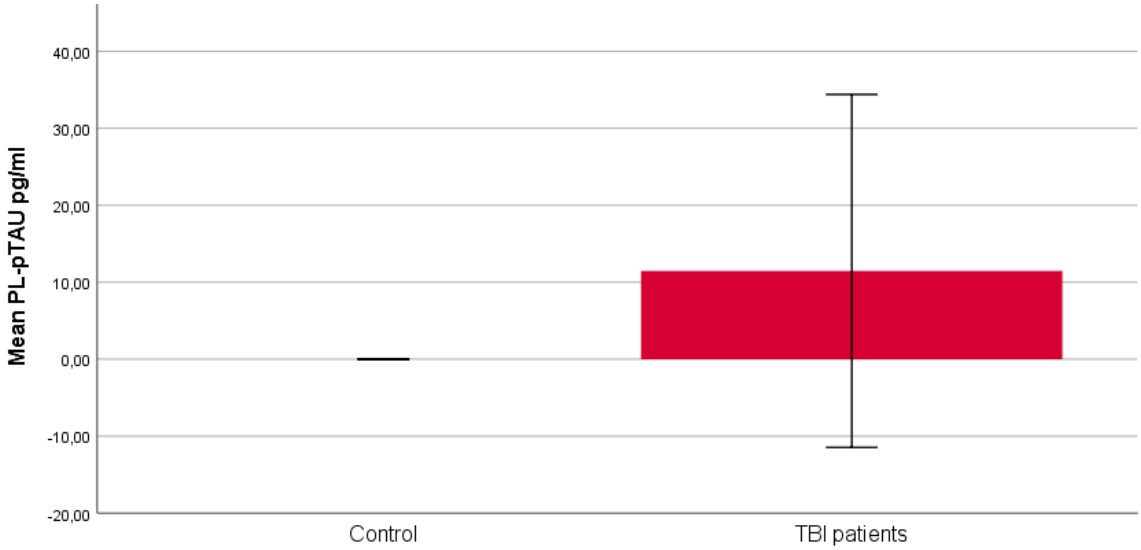

Figure 2 Graphic representation of pTAU concentration in plasma of control and study group.

Statistically insignificant differences in the concentration of NF-L in cerebrospinal fluid from the control group (Median=0.000, n= 11) and patients with craniocerebral trauma (Median= 0.000, n=20), U= 82, z= 1.676, p = 0.094.

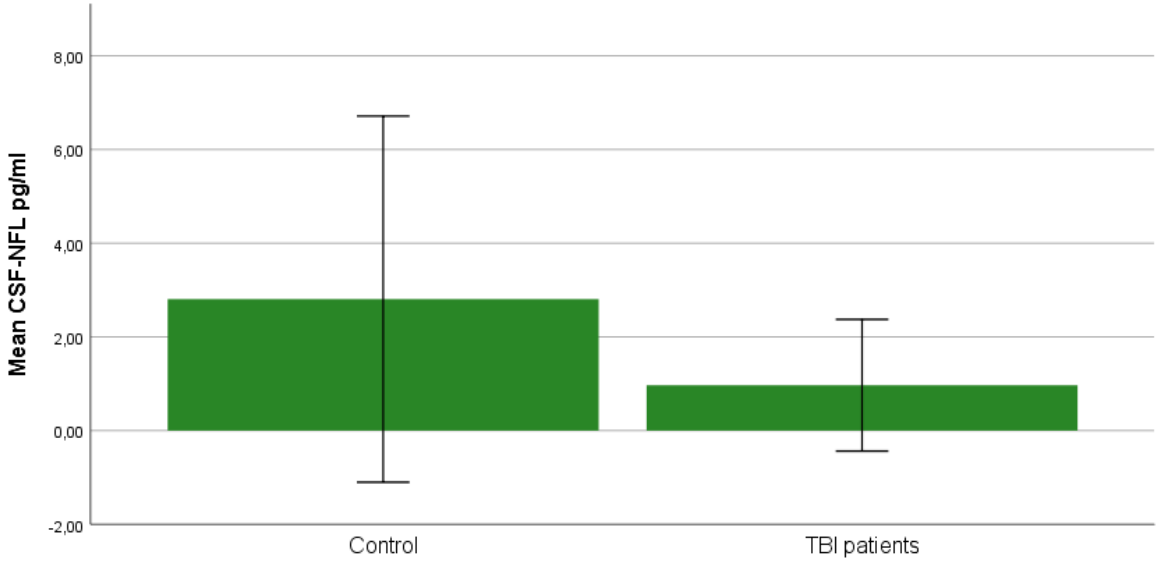

Figure 3 Graphic representation of NF-L concentration in cerebrospinal fluid of the control and study groups.

Statistically significant differences in the concentration of NF-L in plasma from the control group (Median=1.1600, n= 11) and patients with brain injury (Median= 0.000, n=20), U= 58 , z= 2.790, p = 0.005.

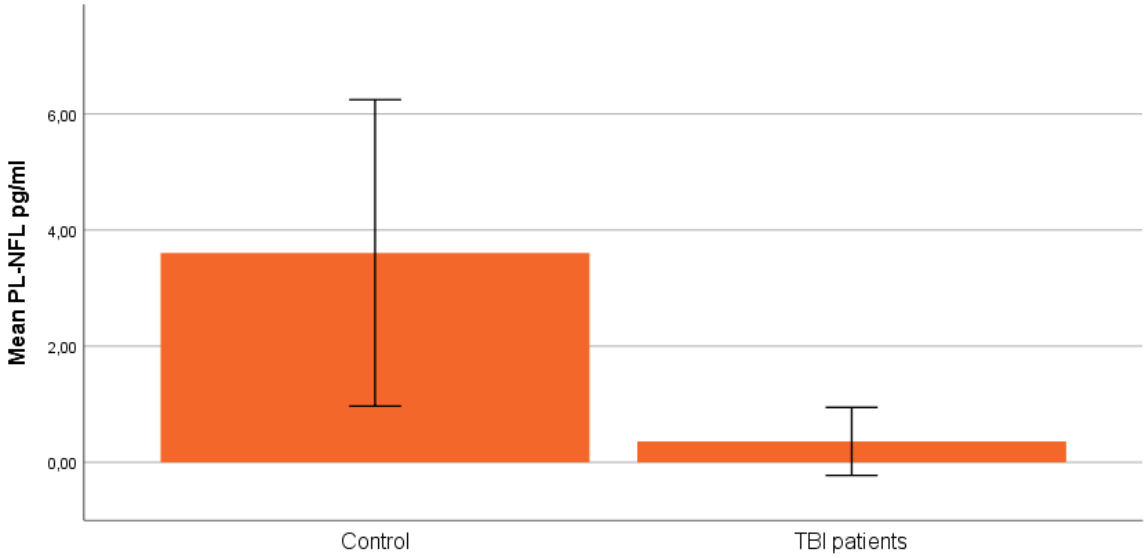

Figure 4 Graphic representation of NF-L concentration in plasma of the control and the studied group.

Of the studied group of patients, 8 survived and 12 died in the first 6 months after the TBI. No statistically significant correlation was observed between the levels of the studied indicators in CSF and plasma and the mortality of patients.

Discussion

The study conducted showed a statistically significant increase in the concentration of p-TAU in cerebrospinal fluid (p=0.001) in the control group, which was not found in any other similar studies. A statistically significant increase in plasma NF-L concentration was found in the control group (p=0.005). An increased concentration of CSF and NF-L was reported in the control group,

although the results were not statistically significant. While not statistically significant (p=0.458), plasma pTau was increased in TBI patients, which correlated to the results from other similar studies [15] [16]. The low levels of NF-L in CSF and plasma in TBI patients less than 24 hours after the injury correlates with the data from literature [13], [17], proving NF-L to be a late, rather than early predictor for TBI. The obtained results indicate that the investigated biomarkers are most likely not suitable for early diagnosis of craniocerebral injuries. It is necessary to carry out studies on a larger scale, in order to clarify the reported dependencies.

Conflict of interests

There is no conflict of interest.

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References

1. C. S. Hill, M. P. Coleman, and D. K. Menon, "Traumatic Axonal Injury: Mechanisms and Translational Opportunities," *Trends in Neurosciences*, vol. 39, no. 5. Elsevier Ltd, pp. 311–324, May 01, 2016. doi: 10.1016/j.tins.2016.03.002.
2. S. Kleiven, "Why Most Traumatic Brain Injuries are Not Caused by Linear Acceleration but Skull Fractures are," *Front Bioeng Biotechnol*, vol. 1, 2013, doi: 10.3389/fbioe.2013.00015.
3. D. A. Crooks, C. L. Scholtz, F. G. Vowles, S. Fimls, and S. Greenwald, "Axonal Injury in Closed Head Injury 109."
4. H. Xiao-sheng, Y. Sheng-yu, Z. Xiang, F. Zhou, Z. Jian-ning, and Y. Li-sun, "Diffuse axonal injury due to lateral head rotation in a rat model," 2000.
5. J. Haarbauer-Krupa, M. J. Pugh, E. M. Prager, N. Harmon, J. Wolfe, and K. Yaffe, "Epidemiology of Chronic Effects of Traumatic Brain Injury," *J Neurotrauma*, vol. 38, no. 23, pp. 3235–3247, Dec. 2021, doi: 10.1089/neu.2021.0062.

A. R. Rabinowitz and D. H. Smith, "Traumatic Brain Injury and Rationale for a Neuropsychological Diagnosis of Diffuse Axonal Injury," in *Genes, Environment and Alzheimer's Disease*, Elsevier Inc., 2016, pp. 267–293. doi: 10.1016/B978-0-12-802851-3.00009-7.

6. P. Angelova, I. Kehayov, A. Davarski, and B. Kitov, "Contemporary insight into diffuse axonal injury," *Folia medica*, vol. 63, no. 2. NLM (Medline), pp. 163–170, Apr. 30, 2021. doi: 10.3897/folmed.63.e53709.
7. S. H. Jang, "Diagnostic problems in diffuse axonal injury," *Diagnostics*, vol. 10, no. 2. MDPI AG, 2020. doi: 10.3390/diagnostics10020117.
8. D. B. Hier *et al.*, "Blood biomarkers for mild traumatic brain injury: a selective review of unresolved issues," *Biomarker Research*, vol. 9, no. 1. BioMed Central Ltd, Dec. 01, 2021. doi: 10.1186/s40364-021-00325-5.
9. R. Tomaiuolo, M. Zibetti, C. Di Resta, and G. Banfi, "Challenges of the Effectiveness of Traumatic Brain Injuries Biomarkers in the Sports-Related Context," *Journal of Clinical Medicine*, vol. 12, no. 7. MDPI, Apr. 01, 2023. doi: 10.3390/jcm12072563.
10. H. S. Ghaith *et al.*, "A Literature Review of Traumatic Brain Injury Biomarkers," *Molecular Neurobiology*, vol. 59, no. 7. Springer, pp. 4141–4158, Jul. 01, 2022. doi: 10.1007/s12035-022-02822-6.
11. S. Yokobori, K. Hosein, S. Burks, I. Sharma, S. Gajavelli, and R. Bullock, "Biomarkers for the clinical differential diagnosis in traumatic brain injury-A systematic review," *CNS Neuroscience and Therapeutics*, vol. 19, no. 8. pp. 556–565, Aug. 2013. doi: 10.1111/cns.12127.

12. F. Al Nimer *et al.*, "Comparative assessment of the prognostic value of biomarkers in traumatic brain injury reveals an independent role for serum levels of neurofilament light," *PLoS One,* vol. 10, no. 7, Jul. 2015, doi: 10.1371/journal.pone.0132177.
13. K. Nishimura, J. G. Cordeiro, A. I. Ahmed, S. Yokobori, and S. Gajavelli, "Advances in Traumatic Brain Injury Biomarkers," *Cureus,* Apr. 2022, doi: 10.7759/cureus.23804.
14. R. Rubenstein *et al.*, "Comparing plasma phospho tau, total tau, and phospho tau–total tau ratio as acute and chronic traumatic brain injury biomarkers," *JAMA Neurol*, vol. 74, no. 9, pp. 1063–1072, Sep. 2017, doi: 10.1001/jamaneurol.2017.0655.
15. J. Gill, K. Merchant-Borna, A. Jeromin, W. Livingston, and J. Bazarian, "Acute plasma tau relates to prolonged return to play after concussion," 2017.
16. S. Neselius, H. Brisby, A. Theodorsson, K. Blennow, H. Zetterberg, and J. Marcusson, "Csf-biomarkers in olympic boxing: Diagnosis and effects of repetitive head trauma," *PLoS One,* vol. 7, no. 4, Apr. 2012, doi: 10.1371/journal.pone.0033606.