ESPECIALLY CELLULAR IMMUNITY IN PATIENTS WITH TRAUMATIC BRAIN INJURY OF DIFFERENT SEVERITY IN ACUTE PERIOD


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OSOBEHNOSTI KLETOKHNOGO IMMUNITETA U PAKhIENTOV C CHEREPNO-MOZGOVNOY TRAUMY RAZLICHNOY STEPENI TAJESTVI V OSTROM PERIODE ZABOLYVANIIA

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Currently, it is known that in response to brain damage a reaction from the immune system is triggered, but its role in the formation of clinical manifestations remains a little-studied problem to date. Using multicolor cytometric analysis, a study was conducted to determine the number of Th1-, Th2-, Tfh- and Th17-cells among CD45RA-negative CD3 + CD4 + cells in a group of patients with mild TBI (M-TBI, n = 20) and with moderate to severe TBI (MS-TBI, n = 16) and a group of conditionally healthy donors (n = 30). The imbalance between Th2 cells and Th17 cells was found in patients with TBI in the acute period, which probably predetermines the course of the disease and the development of complications.

Keywords: flow cytometry; traumatic brain injury (TBI); CD45RA-negative (memory) CD3 + CD4 + lymphocytes.

Introduction. The use of structural neuroimaging techniques often does not allow timely assessment of the severity of the brain injury that occurred. This is characteristic, first of all, for traumatic brain injury (TBI) of mild severity, at which a submicroscopic level of damage is observed. This provision determines the need to find new methods for differential diagnosis of severe TBI and predict the risk of development of consequences. One of these promising areas is the study of the immune status or subpopulation composition of peripheral blood lymphocytes of patients in the acute period of injury. Currently, it is known that the response of the immune system is triggered in response to brain damage, but its role in the formation of individual clinical manifestations remains a problem that has not been sufficiently studied [1]. Reactions from the immune system, which develop after a brain injury and, possibly, are directed against their own antigens, in the early period of the disease are related to damage to the nervous tissue. However, according to the latest available data, they are subsequently able to stimulate the processes of repair and regeneration in the brain tissue [2]. To analyze the nature of changes during TBI, flow cytofluorometry was...
used to examine T-lymphocyte subpopulations in patients with brain injury.

**Material and methods.** Blood samples in 16 patients with diagnosis mild TBI (M-TBI) and 20 patients with moderate to severe TBI (MS-TBI) between the ages of 18 and 45 years. The severity of TBI was assigned according to established criteria [3].

The combination of monoclonal antibodies, approach design and “gating strategy” allowed to identify main Th subsets in patients and healthy control peripheral blood were described previously [4]. Sample acquisition was performed using a Navios flow cytometer (Beckman Coulter, Inc., USA). Th subsets size (the percentage) within CD45RA-negative (memory) CD3⁺ CD4⁺ lymphocytes are given in medians and quartile ranges (Med (Q₂₅; Q₇₅)).

The statistical comparisons of data between TBI patients and healthy controls were performed using the Mann-Whitney U-test. Differences were considered significant when \( p \) values were < 0.05. All of the statistical analysis of data was carried out with GraphPad Prizm 6 (GraphPad Software, USA).

**Result and discussion.** The results shown in figure. The comparison of these different Th subsets between patients and control group indicated significantly lower Th1 (\( p = 0.019; p = 0.003 \)) and significantly higher Th17 (\( p < 0.001; p < 0.001 \)) with patients of M-TBI and M-TBI if compared with healthy controls. Also, we did observe significant lower in Th2 cells with patients of M-TBI if compared with healthy controls. As for the Thh-cells, they were change only slightly, but there are significant changes (\( p = 0.039 \)) between the groups with M- and MS-TBI.

The revealed changes in the CD45RA-negative CD3⁺CD4⁺ cell population in patients with TBI allowed to raise the question of the significance of determining these cells subpopulations, which may be a predictor of the course of TBI in the acute period.

Scatter plots a, b, c and d showing the percentages Th1 (CXCR5⁻CXCR3⁺CCR6⁻CCR4), Th2 (CXCR5⁻CXCR3⁻CCR6⁻CCR4⁺), Th17(CXCR5⁻CXCR3⁻CCR6⁺CCR4⁻) and Thh (CXCR5⁺CXCR3⁻CCR6⁻CCR4⁻), respectively, in the peripheral blood samples for TBI and healthy control subjects. Numbers represent the percentage of the indicated Th subset among total CD45RA-negative CD3⁺CD4⁺ cell population. Each dot represents individual subjects, and horizontal bars represent the group medians and quartile ranges (Med (Q₂₅; Q₇₅)).

**References**