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Original Article

Association Between Acute Inflammatory Cells and Mutation in ICAM-1 Gene With Injury Severity and Outcome Among Traumatic Cerebral Hemorrhagic Contusion

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Background: Acute inflammation in the brain after trauma is mediated by acute inflammatory cells (neutrophils) that contributes to ischemic brain damage, neurological deterioration, and poor outcome. Migration of neutrophils to brain is mediated by intercellular adhesion molecule-1 (ICAM-1). The aim was to determine an association between neutrophils counts, ICAM-1 level and mutation in ICAM-1 gene with injury severity and patient's outcome.

Methods: Cross-sectional study was conducted for 90 Sudanese patients presented with traumatic cerebral hemorrhagic contusion to the National Center for Neurological Sciences, Khartoum, Sudan from December 2015 to January 2018. Non-Sudanese patients and hemorrhagic contusion associated with other type of brain bleeding were excluded in this study. Moreover, 90 apparently healthy individuals were participated as control.

Results: Most patients were males (93.3%), their ages ranged from 25 to 44 years, 11.1% of the patients had severe brain injury, 22.2% had brain edema and the mortality rate was 8.9%. Circulatory levels of leukocytes, neutrophil and, ICAM-1 among patients who sustained trauma were significantly elevated compared with controls ($p = 0.000$). The high level of leukocytes and neutrophils counts were significantly associated with ICAM-1 pg/mL circulatory level. High levels of leukocytes, neutrophils, and ICAM-1 were documented in severe brain injuries. High level of ICAM-1 was observed among patients admitted with brain edema. Leukocytes and neutrophils counts were significantly associated with patient outcome. High level of ICAM-1 (304.88 pg/mL) was observed among patients with poor outcome compared to survivals (263.93 pg/mL). The highest circulatory level of ICAM-1 (280.75 pg/mL) was observed among patient having adenine-adenine (AA) mutant homozygous alleles, followed by (272 pg/mL) for guanine-guanine (GG) homozygous alleles, then (245.12 pg/mL) for guanine-adenine (GA) heterozygous alleles.

Conclusions: Mutation in ICAM-1 gene and increased levels of leukocytes, neutrophils, and ICAM-1 constitutes important markers for injury severity and patient's outcome.

Key words: *cerebral contusion, inflammation, ICAM-1, neutrophils*

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Introduction

Infiltration of peripheral leukocytes in central nervous system (CNS) can be caused by direct rupture of vessels in the primary insult or by secondary disruption of the blood brain barriers (BBB) due to neuro-inflammation or up regulation of adhesion molecule on the endothelial surface.[1] Neutrophils are rapidly recruited following CNS injury, present at the time of neuronal death, and can trigger tissue damage through generation of toxic free radicals, proteolytic enzymes and pro-inflammatory cytokines such as interleukin 1β (IL-1β) and tumor necrosis factor α (TNF-α).[2] Intercellular adhesion molecule-1 (ICAM-1) is a member of the immunoglobulin superfamily mainly expressed by endothelial cells, mediates leukocytes passage across the vascular endothelial cell layer to the sites of tissue injury by promoting leukocytes firm adhesion and transmigration.[3] The G > A 5498 polymorphism in exon 6 of ICAM-1 gene has been shown to influence the binding of ICAM-1 on endothelial cells and Leukocyte function antigen and Macrophage one antigen on leucocytes, mediating leukocytosis and its migration in an inflammatory environment.[4] This domain is essential for the structure and function of ICAM-1 protein.[5] Recent genome wide association studies have demonstrated a strong correlation between rs5498 and adhesion molecules level.[6] Genetic variants in ICAM-1 gene have been shown to regulate the expression level of ICAM-1 and have been widely studied for possible genetic association with a range of degenerative and inflammatory diseases.[7,8] Traumatic brain injuries (TBIs) including cerebral hemorrhagic contusion induced disturbances in the cerebrum-vasculature and may cause endothelial cell activation that alters expression of ICAM-1.9 All types of TBIs including hemorrhagic cerebral contusion disrupt BBB leading to infiltration of cells and subsequent inflammation and neuro-degeneration. Mechanism of how the immune response affects the TBIs outcome is controversy and different studies reported different results. The aim of this study was to find out possible association between neutrophils counts, ICAM-1 level and mutation in ICAM-1 gene (G > A 5498) with injury severity and patient's outcome.

Methods

Ethical Approval

The ethical approval was obtained from Ethical Review Board of National Center for Neurological Sciences (NCNS).

Study Populations

This is a prospective, cross-sectional study conducted at the NCNS from December 2015 to 2018. Non-Sudanese patients and hemorrhagic contusion associated with other type of brain bleeding were excluded. Epidemiological and clinical data were collected using questionnaire.

Laboratory Assay

Two blood samples were collected from all participants on entry, one in ethylenediamin tetra-acetic-acid for leukocytes counts and molecular study. The second in plain container for ICAM-1 measurement. Leukocyte counts were done using Mindary BC-3200 auto hematology analyzer (Mindray-biomedical Co., Ltd., Shenzhen, China). Serum was separated by centrifugation (Hettich Zenterfuge EBA200, Kirchlengern, Germany) at 2,000 RPM for 5 minutes (= 460.616 g) and then stored at -20°C. The level of ICAM-1 was measured in serum using enzyme-linked Immunosorbent assay system (TECAN Group Ltd., Mannedorf, Switzerland) based on the kits protocol (Kono Biotech Co-Ltd, Zhejiang, China).

Molecular Genetic Analysis

Deoxyribonucleic acid (DNA) was extracted from whole blood using QIAGEN®(Biotechnology industry, Venlo, Netherlands, Germany) commercial DNA extraction kits (vacuum protocol). DNA quantity and quality was measured using Nanodrop spectrophotometer and gel electrophoresis (Cleaver Gel Documentation system, Cleaver Scientific Ltd., Rugby, Warwickshire, UK). The following primers were used to amplify the target region in ICAM-1 gene (5'-GGAACCCATTGCCCGAGC-3' forward, and 5'GGTGAGGATTGCATTAGGTC-3' reverse). Polymerase chain reaction (PCR) (TC-3000 Thermal Cycler-PCR, Barloworld Scientific, Staffordshire, UK) was conducted in to 20 μL reaction volume that contains 4 μL of 5× Firepol® Master Mix (Solis BioDyne, Tartu, Estonia), 1 μL (200 ng) forward primer, 1 μL (200 ng) reverse primer, 1 μL (200 ng) DNA and 13 μL distilled water. The PCR reaction condition includes initial denaturation 94°C/3 minutes, denaturation 94°C/30 minutes, annealing 58°C/30 minutes,

extension 72°C/30 minutes and final extension 72°C/5 minutes for 30 cycles. The PCR product was then loaded in 2% agarose gel electrophoresis and visualized under ultraviolet light. PCR product was digested with BstUI restriction enzyme (NEW ENGLAND BioLabs®, Ipswich, MA, USA), and separated on 2% agarose gel electrophoresis stained with ethidium bromide and visualized under ultra violet light (Fig. 1).

Statistical Analysis

Data were analyzed using statistical package for social sciences (SPSS) version 19 (IBM Corp., Armonk, NY, USA).

Results

Ninety patients and equally healthy individuals were enrolled in the study. Males constituted 84 patients (93.3%), their age ranged from 25 to 44 years. The Glasgow Coma Scale (GCS) was < 8 (severe) in 11.1% of the patients, GCS 8–12 (moderate disability) was reported in 30.0% of patients and in 58.9% of the patients the GCS was 13–15 (mild). Brain edema was observed among 22.2% of the patients, and the mortality rate among the patients was 8.9% (Table 1). The mean concentration of circulatory levels of leukocytes 12.27 10^9/L and neutrophils count 10.42 10^9/L were significantly higher in patients compared with controls 5.34 × 10^9/L and 3.13 × 10^9/L respectively ($p = 0.000$).

ICAM-1 level was significantly elevated among patients sustained trauma ($p = 0.000$) compared to normal healthy individuals (Table 2). Significant

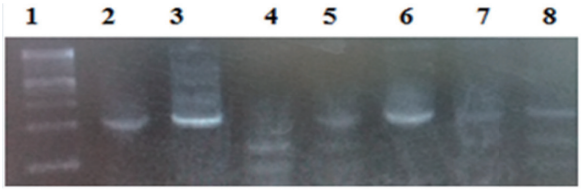

Fig. 1. Agarose gel electrophoresis of polymerase chain reaction products of intercellular adhesion molecule-1 (ICAM-1) gene, digested with BstUI restriction enzyme. Lane 1: deoxyribonucleic acid ladder. Land 2, 3, 5, 6, and 7: guanine-guanine (GG) homozygous alleles (band at 223 bp). Land 8: guanine-adenine (GA) heterozygous alleles (band at 223, 136, and 87 bp). And Land 4: adenine-adenine (AA) homozygous alleles (band at 136 and 87 bp).

Table 1. Epidemiological and clinical features of trauma patients

Parameter	N (%)
Gender	
Male	84 (93.3)
Female	6 (6.7)
Age	
5–14	19 (21.1)
15–24	23 (25.6)
25–44	29 (32.2)
45–65	14 (15.6)
More than 65	5 (5.6)
GCS	
Mild injury (13–15)	53 (58.9)
Moderate injury (8–12)	27 (30.0)
Sever injury (< 8)	10 (11.1)
Presence of brain edema	
Yes	20 (22.2)
No	70 (77.8)
Outcome	
Discharge	82 (91.1)
Death	8 (8.9)

GCS: Glasgow Coma Scale.

correlation was found between leukocytes and neutrophils counts with ICAM-1 pg/mL circulatory level. The mean concentrations of leukocytes, neutrophils, and ICAM-1 were high in severe brain injures compared with moderate and mild one. High circulatory level of ICAM-1 was observed among patients admitted with brain edema. However, leukocytes and neutrophils counts show no significant association with presence or absence of brain edema. leukocytes ($p = 0.026$) and neutrophils ($p = 0.017$) counts were significantly associated with patient outcome. In contrast no statistical significant difference was seen between high ICAM-1 level and mortality, although the high (304.88 pg/mL) ICAM-1 level was observed among patients developed poor outcome compared to survivals (263.93 pg/mL) (Table 3). The polymerase chain reaction - restriction fragment polymorphism results of ICAM-1 gene genotype showed that, guanine-guanine (GG) was representing 76.7% of the patients, guanine-adenine (GA) 18.9% and adenine-adenine (AA) 4.4%. The highest circulatory level of ICAM-1

Table 2. The levels of leukocytes, neutrophils Interleukin one beta, and intercellular adhesion molecule-1 among case and control

Clinical feature	Mean concentration		
	TWBCs 10^9/L	Neutrophils 10^9/L	ICAM-1 pg/mL
GCS groups			
Mild injury	11.70	9.72	271.57
Moderate injury	12.70	10.93	243.70
Sever injury	14.10	12.80	311.10
p-value	0.311	0.138	0.402
Brain edema			
Yes	12.29	10.30	300.40
No	12.20	10.46	258.19
p-value	0.845	0.897	0.230
Outcome			
Death	15.63	14.13	304.88
Discharge	11.94	10.06	263.93
p-value	0.026	0.017	0.082

Values represent mean ± standard deviation.
GCS: Glasgow Coma Scale; ICAM-1: intercellular adhesion molecule one; pg: pictogram; TWBCs: total white blood cells.

Table 3. Association of leukocytes, neutrophils, and intercellular adhesion molecule-1 with clinical features of patients

Parameter	Group	Mean	Std. deviation	Ratio case/control	*p*-value
Leukocytes 10^9/L	Case	12.27	4.87	2.3	0.000
	Control	5.34	1.53		
Neutrophils 10^9/L	Case	10.42	4.76	3.3	0.000
	Control	3.13	1.15		
ICAM-1 pg/mL	Case	267.57	137.97		0.000
	Control	192.80	36.26	1.4	

ICAM-1: intercellular adhesion molecule-1; pg: picogram.

(280.75 pg/mL) was observed among patients having AA mutant homozygous alleles, followed by (272.33 pg/mL) for GG homozygous alleles, and then (245.12 pg/mL) for GA heterozygous alleles (Fig. 2). However, ICAM-1 genotypes were not associated with patient clinical features and outcome (Table 4).

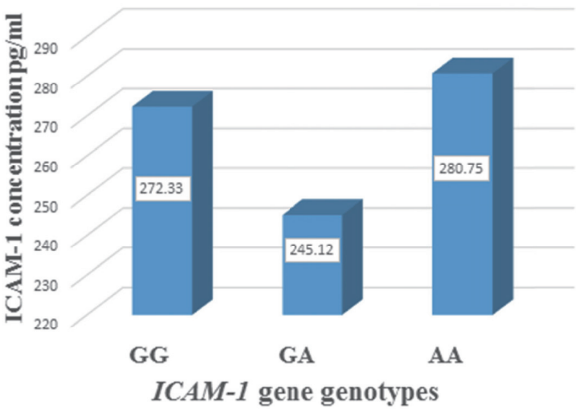

Fig. 2. Association between intercellular adhesion molecule-1 (ICAM-1) gene genotypes with ICAM-1 circulatory levels (*p*-value of ANOVA test = 0.757).

Discussion

Recruitment of leukocytes, specifically neutrophil is characteristic of the early inflammatory response following human TBIs. Recent data have shown that neutrophils infiltrate into the injured brain more rapidly than other types of peripheral inflammatory cells and are known as the first responders.[10]

Table 4. Association between intercellular adhesion molecule-1 gene genotypes with clinical features of patients

Clinical features	ICAM-1 genotypes			*p*-value
	GG	GA	AA	
GCS groups				
Mild injury, n (%)	39 (56.5)	11 (64.7)	3 (75.0)	0.675
Moderate injury, n (%)	23 (33.3)	3 (17.6)	1 (25.0)	
Sever injury, n (%)	7 (10.1)	3 (11.8)	0 (0.0)	
Brain edema				
Yes, n (%)	15 (21.7)	4 (23.5)	1 (25.0)	1.000
No, n (%)	54 (78.3)	13 (76.5)	3 (75.0)	
Outcome				
Death, n (%)	6 (8.7)	2 (11.8)	0 (0.0)	0.763
Discharge, n (%)	63 (91.3)	15 (88.2)	4 (100.0)	

AA: adenine-adenine; GA: guanine-adenine; GCS: Glasgow Coma Scale; GG: guanine-guanine; ICAM-1: intercellular adhesion molecule-1.

Our finding demonstrates the significant elevation of leukocytes and neutrophils among traumatic cerebral hemorrhagic contusion patients compared to normal healthy individuals.[11] Increased concentration of ICAM-1 was previously reported in patients with cardiovascular disease, tumors, auto-immune disease, and other diseases with an inflammatory reaction.[12] Adhesion molecules play a pathophysiologic role in cerebrovascular diseases.[13] Up regulation of ICAM-1 has been reported in cerebral micro-vessels following TBIs in rats.[14] ICAM-1 was used as a marker of the activation of cerebral vascular endothelial cells after TBIs in experimental animals.[15] In humans the level of ICAM-1 was measured previously in many ischemic cerebrovascular diseases, and was shown to be significantly elevated in patients with subarachnoid hemorrhage over control subjects and directly correlated with cerebral vasospasm.[16] ICAM-1 was also expressed in the brain after cortical contusion injuries.[17] In the present study serum ICAM-1 was significantly elevated among cerebral hemorrhagic contusion TBIs patients compared with the normal healthy individuals. In addition, ICAM-1 level was highly correlated with leukocytes. Similarly, the infiltration and accumulation of leukocytes in the contused injured brain area was significantly associated with increased level of ICAM-1 and was correlated with second brain injury.[4] The expression of ICAM-1 was reported previously to produce an accumulation of neutrophils, this in turn, cause cell damage.[18] It is hypothesized that, patients with significant injury should have a higher degree of leukocytosis compared to patients with minor injurie.[19] However, leukocytes as predictive marker for injury severity is conflicting; some researcher reported significant association,[11] while others reported non-significant association.[20] Neutrophils that are recruited to sites of brain trauma were typically correlated with severity of injury.[21] Although no statistical significant correlation between leukocytes and neutrophils counts with injury severity in our study, the highest count was seen among patients who sustained severe injuries, compared to moderate and mild injuries. A previous study demonstrated that high ICAM-1 concentration correlated well with tissue and BBB damage, and giving an indication of the degree of immunologic activation in the injured CNS.[22]

Over expression of ICAM-1 was reported to be associated with severe inflammatory processes and tissue injury.[23] Increased ICAM-1 further induces adhesion between vascular endothelial cells and leukocytes, which impairs vascular endothelial cells and increases the permeability of capillaries, and eventually leads to brain damage.[24] Our finding revealed that although no statistically significant correlation between ICAM-1 level and injury severity; the highest concentration of ICAM-1(311.10 pg/mL) was observed among patients who presented with severe injury compared to moderate (243.70 pg/mL) and mild (271.57 pg/mL). However, ICAM-1 level was influenced by various biological factors such as age, sex, smoking, insulin resistance, IL-6 level, alkaline phosphatase, and aspartate transaminase activities.[25]

Brain swelling occurring after head trauma is

probably an inflammatory response due to intracerebral cytokine production and increased leukocytes adhesion as a result of a direct effect on vascular permeability and leucocytes activation.[26] Accumulation of leukocytes leads to secondary brain injury by lowering cerebral blood flow, increased edema and elevated intracranial pressure.[27] In experimental rats with traumatic cerebral contusions, the infiltration and accumulation of leukocytes in the contused injured brain area was significantly associated with brain edema and was correlated with second brain injury.[4] Emerich et al. have suggested an important role for neutrophils in edema formation in the CNS following ischemic brain insult.[2] In a very recent study high neutrophils count was used as predictive marker for cerebral edema formation among cerebral infarction patients.[28] Mckee and Lukens correlated neutrophils with edema formation through BBB breakdown, neuro-degeneration, and subsequent neuronal death.[21] Morancho et al. stated that neutrophils can promote early pathogenesis including brain swelling through the release of free radicals' proteases and pro-inflammatory cytokines.[29] Depletion of neutrophils significantly decreases tissue edema.[30] However, Whalen et al. claimed that there is no direct relationship between brain edema and leukocytes accumulation.[31] The connection between neutrophil infiltration and brain edema seems to be a time-dependent issue.[32] Our finding demonstrated no significant correlation between brain edema and high leukocytes/neutrophils counts. Leukocytosis is associated with a worse outcome, particularly during focal ischemia or hypoxia, which are frequently found in patients with severe head injury.[33] High leukocytes and neutrophils counts were significantly associated with death in our study. It is well documented that neutrophils attempt to clear cell debris by phagocytosis, but also contribute to ongoing damage by releasing toxic mediators such as reactive oxygen species, proteases, and pro-inflammatory cytokines which have the potential to adversely affect the integrity of the BBB and contribute to secondary brain injury.[34] In contrast, Roth et al. study does not fully support that more neutrophils' recruitment leads to more severe brain injury and worse outcome.[35] The discrepancy between different results was justified by several studies which suggest the presence of two subpopulations of neutrophils with opposite roles. In a stroke model, it was shown that rosiglitazone, one of the agonists for peroxisome proliferator-activated receptor-γ, could promote the infiltration of N2-like neutrophils into the ischemic core and protect neuron damage concomitantly through facilitating the dissolution of inflammatory responses.[36] Cuartero et al. proved that N2 phenotype of neutrophils is beneficial for the brain injury. Despite the compelling evidence for the separate subsets of neutrophils, there are no specific markers to identify and distinguish them, which warrants more investigation in the future. Their roles in the CNS diseases may depend on pro/anti-inflammatory (N1, N2) phenotypes that are regulated by specific environmental cues in the brain after injury. Further understanding of these cues and the outcomes associated with particular phenotypes may allow neutrophils to serve as disease-modifying factors in the CNS.[36] Mckeating et al. reported that serum level of ICAM-1 significantly correlated with outcome after TBIs.[22]

Although no significant association was found between ICAM-1concentration and outcome in our study, patients admitted with high concentration of ICAM-1 (304.88 pg/mL) develop poor outcome compared to survival (263.93 pg/mL).

In the present study no association between ICAM-1 G > A 5498 gene polymorphism and mortality, however, all patients (100%) who carry AA mutant homozygous alleles have shown poor outcome. The G > A 5498 in ICAM-1 gene change the amino acid sequence of the immunoglobulin-like domain 5. This domain is crucially important for the activity of the ICAM-1 protein.[4] ICAM-1 G >A 5498 gene polymorphism was associated with an increased expression of ICAM-1,[37] and with high ICAM-1 circulatory levels.[38] Positive correlation was found between ICAM-1 G > A 5498 gene polymorphism with ICAM-1 circulatory levels among patients suffering from cerebrovascular disorder.[39] Ataya et al. stated that ICAM-1 is involved in endothelial injury and atherosclerosis; synthesis and genetic basis of this molecule may be therapeutic target for the complications of TBIs.[40] Our finding demonstrates negative association between ICAM-1 gene polymorphism G > A 5498 with ICAM-1 circulatory levels although the highest concentration was observed among AA mutant homozygous alleles carriers. These conflicting results may be explained by the size, the heterogeneity of TBIs and the ethnic background variety of different populations. However, genetic association studies require large sample size, biological evidence and functional significance that the risk variant is implicated in the pathogenesis of disease.

Conclusions

Mutation in the inflammatory ICAM-1 gene, increased levels of serum ICAM-1 and highly neutrophils are involved in the pathogenesis after traumatic cerebral hemorrhage. Further studies that defining inflammatory pathways' influence traumatic cerebral hemorrhage pathogenesis will improve understanding brain injury etiology and will aid in the identification novel immune-based treatment strategies.

Conflict of Interest Statement

Non-declared.

Acknowledgments

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References

1. Lenzlinger PM, Morganti-Kossman MC, Laurer HL, McIntosh TK. The duality of the inflammatory response to traumatic brain injury. *Mol Neurobiol* 2001;24:169-181. doi:10.1385/MN:24:1-3:169
2. Emerich DF, Dean RL 3rd, Bartus RT. The role of leukocytes following cerebral ischemia: pathogenic variable or bystander reaction to emerging infarct? *Exp Neurol* 2002;173:168-181. doi:10.1006/exnr.2001.7835
3. Knorr R, Dustin ML. The lymphocyte function-associated antigen 1 domain is a transient binding module for intercellular adhesion molecule (ICAM)-1 and ICAM-3 in hydrodynamic flow. *J Exp Med* 1997;186:719-730. doi:10.1084/jem.186.5.719
4. Ma Q, Chen S, Klebe D, Zhang JH, Tang J. Adhesion molecules in CNS disorders: biomarker and therapeutic targets. *CNS Neurol Disord Drug Targets* 2013;12:392-404. doi:10.2174/18715273113120300 12
5. Miller J, Knorr R, Ferrone M, Houdei R, Carron CP, Dustin ML. Intercellular adhesion molecule-1 dimerization and its consequences for adhesion mediated by lymphocyte function associated-1. *J Exp Med* 1995;182:1231-1241. doi:10.1084/jem.182.5.1231
6. Paré G, Ridker PM, Rose L, et al. Genome-wide association analysis of soluble ICAM-1 concentration reveals novel associations at the NFKBIK, PNPLA3, RELA, and SH2B3 loci. *PLoS Genet* 2011;7:e1001374. doi:10.1371/journal.pgen.1001374
7. Papa A, Danese S, Urgesi R, et al. Intercellular adhesion molecule 1 gene polymorphisms in inflammatory bowel disease. *Eur Rev Med Pharmacol Sci* 2004;8:187-191.
8. Ji YN, Wang Q, Zhan P. Intercellular adhesion molecule 1 gene K469E polymorphism is associated with coronary heart disease risk: a meta-analysis involving 12 studies. *Mol Biol Rep* 2012;39:6043-6048. doi:10.1007/s11033-011-1418-6
9. Balabanov R, Goldman H, Murphy S, et al. Endothelial cell activation following moderate traumatic brain injury. *Neurol Res* 2001; 23:175-182. doi:10.1179/016164101101198514
10. Li P, Gan Y, Mao L, Leak R, Chen J, Hu X. The critical roles of immune cells in acute bain injuries. In: Chen J, Hu X, Stenzel-Poore M, Zhang J, eds. *Immunological Mechanisms and Therapies in Brain Injuries and Stroke*. New York, NY: Springer; 2014:9-25. doi:10.1007/978-1-4614-8915-3_2
11. Subramanian A, Agrawal D, Pandey RM, Nimiya M, Albert V. The leukocyte count, immature granulocyte count and immediate outcome in head injury patients. In Agrawal A, ed. *Brain Injury—Pathogenesis, Monitoring, Recovery and Management*. Rijeka, Croatia: InTech; 2012:139-152. doi:10.5772/27020
12. Witkowska AM, Borawska MH. Soluble intercellular adhesion molecule-1(sICAM-1): an overview. *Eur Cytokine Netw* 2004;15:91-98.
13. Kraus J, Gerriets T, Leis S, Stolz E, Oschmann P, Heckmann JG. Time course of VCAM-1 and ICAM-1 in CSF in patients with basal ganglia haemorrhage. *Acta Neurol Scand* 2007;116:49-55. doi:10.1111/j.1600-0404.2006.00790.x
14. Chen G, Shi J, Hu Z, Hang C. Inhibitory effect on cerebral inflammatory response following traumatic brain injury in rats: a potential neuroprotective mechanism of N-aetylcysteine. *Mediators Inflamm* 2008;2008:716458. doi:10.1155/2008/716458
15. Wang KW, Wang HK, Chen HJ, et al. Simvastatin combined with antioxidant attenuates the cerebral vascular endothelial inflammatory response in a rat traumatic brain injury. *Biomed Res Int* 2014;2014:910260. doi:10.1155/2014/910260
16. Frijns CJ, Kappelle LJ. Inflammatory cell adhesion molecules in ischemic cerebrovascular diseases. *Stroke* 2002;33:2115-2122. doi:10.1161/01.STR.0000021902.33129.69
17. Hang CH, Chen G, Shi JX, Zhang X, Li JS. Cortical expression of nuclear factor kappaB after human brain contusion. *Brain Res* 2006;1109:14-21. doi:10.1016/j.brainres.2006.06.45
18. Kira S, Daa T, Kashima K, Mori M, Noguchi T, Yokoyama S. Mild hypothermia reduces expression of intracellular adhesion molecule-1 (ICAM-1) and the accumulation of neutrophils after acid-induced lung injury in the rat. *Acta Anaesthesiol Scand* 2005;49:351-359. doi:10.1111/j.1399-

6576.2005.00593.x
19. Santucci CA, Purcell TB, Mejia C. Leukocytosis as a predictor of severe injury in blunt trauma. *West J Emerg Med* 2008;9:81-85.
20. Paladino L, Subramanian AR, Bonilla E, Sinert RH. Leukocytosis as prognostic indicator of major injury. *West J Emerg Med* 2010;11:450-455.
21. McKee CA, Lukens JR. Emerging Roles for the immune system in traumatic brain injury. *Front Immunol* 2016;7;556. doi:10.3389/fimmu.2016.00556
22. Mckeating EG, Andrews PJ, Mascia L. Leukocyte adhesion molecules profiles and outcome after traumatic brain injury. *Acta Neurochir Suppl* 1998;71:200-202. doi:10.1007/978-3-7091-6475-4_57
23. Mitsui A, Tada Y, Shibata S, et al. Deficiency of both L-selectin and ICAM-1 exacerbates imiquimod-induced psoriasis-like skin inflammation through increased infiltration of antigen presenting cells. *Clin Immunol* 2015;157:43-55. doi:10.1016/j.clim.2014.12.011
24. Yilmaz G, Granger DN. Leukocyte recruitment and ischemic brain injury. *Neuromolecular Med* 2010;12:193-204. doi:10.007/s12017-009-8074-1
25. Ponthieux A, Herbeth B, Droesch S, Lambert D, Visvikis S. Age-and sex related reference values for serum adhesion molecules concentrations in healthy individuals: Intracellular adhesion molecule-1 and E-, P-, and L-Selectin. *Clin Chem* 2003;49:1544-1546. doi:10.1373/49.9.1544
26. Gürkanlar D, Lakadamyali H, Ergun T, Yilmaz C, Yücel E, Altinörs N. Predictive value of leukocytosis in head trauma. *Turk Neurosurg* 2009;19: 211-215.
27. Schwarzmaier SM, Kim SW, Trabold R, Plesnila N. Temporal profile of thrompogenesis in the cerebral microcirculation after traumatic brain injury in mice. *J Neurotrauma* 2010;27:121-130. doi:10.1089/neu.2009.1114
28. Zhang Y, Liu G, Wang Y, Su Y, Leak RK, Cao G. Procalcitonin as a biomarker for malignant cerebral edema in massive cerebral infarction. *Sci Rep* 2018;8:993. doi:10.1038/s41598-018-19267-4
29. Morancho A, Rosell A, Garcia-Bonilla L, Montaner J. Metalloproteinase and stroke infarct size: role for anti-inflammatory treatment? *Ann N Y Acad Sci* 2010;1207:123-133. doi:10.1111/j.1749-6632.2010.05734.x
30. Lu Q, Harrington EO, Newton J, et al. Adenosine protected against pulmonary edema through transporter- and receptor A2-mediated endothelial barrier enhancement. *Am J Physiol Lung Cell Mol Physiol* 2010;298:L755-L767. doi:10.1152/ajplung.00330.2009
31. Whalen MJ, Carlos TM, Dixon CE, et al. Reduced brain edema after traumatic brain injury in mice deficient in P-selectin and intercellular adhesion molecule-1. *J Leukoc Biol* 2000;67:160-168. doi:10.1002/jlb.67.2.160
32. Zheng H, Chen C, Zhang J, Hu Z. Mechanism and therapy of brain edema after intracerebral hemorrhage. *Cerebrovasc Dis* 2016;42:155-169. doi:10.1159/000445170
33. Rovlias A, Kotsou S. Classification and regression tree for prediction of outcome after severe head injury using simple clinical and laboratory variables. *J Neurotrauma* 2004;21:886-893. doi:10.1089/0897715041526249
34. Gyoneva S, Ransohoff R. Inflammatory reaction after traumatic brain injury: therabutic potential of targeting cell-cell communication by chemokines. *Trends Pharmacol Sci* 2015;36:471-480. doi:10.1016/j.tips.2015.04.003
35. Roth TL, Nayak D, Atanasijevic T, Koretsky AP, Latour LL, McGavern DB. Transcranial amelioration of inflammation and cell death after brain injury. *Nature* 2014;505:223-228. doi:10.1038/nature12808
36. Cuartero MI, Ballesteros I, Moraga A, et al. N2 neutrophils, novel players in brain inflammation after stroke: modulation by the PPARγ agonist rosiglitazone. *Stroke* 2013;44:3498-3508. doi:10.1161/STROKEAHA.113.002470
37. Popović D, Starčević JN, Letonja MŠ, et al. Polymorphism rs5498 of the ICAM-1 gene affects the progression of carotid atherosclerosis in patients with type 2 diabetes mellitus. *Lipids Health Dis* 2016;15:79. doi:10.1186/s12944-016-0247-y
38. Wang MY, Bai DC, Zhu P, Fu Y, Bu DF, Zhang Y. Effects of ICAM-1 gene K469E, K56M polymorphisms on plasma sICAM-1 expression levels in Chinese Yugur, Tibetan and Han Nationalities. *Zhongguo Shi Yan Xue Ye Xue Za Zhi* 2012;20:1205-1211. [in Chinese]
39. Singh A, Singh AK, Singh SK, Paliwal VK, Gupta RK, Prasad KN. Association of ICAM-1 K469E polymorphism with neurocysticercosis. *J Neuroimmunol* 2014;276:166-171. doi:10.1016/j.jneuroim.2014.07.018
40. Atay AE, Esen B, Akbas H, et al. Serum ICAM-1 level and ICAM-1 gene 1462A>G (K469E) polimorphism on microalbuminuria in nondiabetic, nonhypertensive and normolipidemic obese patients: genetical background of microalbuminuria in obesity. *Nefrologia* 2017;37:381-388. doi:10.1016/j.nefro.2016.11.025