

CORRELATION BETWEEN SERUM EHSP70 LEVEL AND SERUM ANG-2, ISS SCORE AND APACHE II IN TRAUMA PATIENTS

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ABSTRACT

Objective: To study the correlation between serum extracellular heat shock protein 70 (eHSP70) level and serum angiopoietin 2 (Ang 2), acute physiology and chronic health evaluation II (APACHE II), and injury severity score (ISS) in trauma patients.

Methods: Forty-eight trauma patients who were treated in our hospital from June 2018 to December 2019 were collected and divided according to ISS score into a mild trauma group (ISS score ≤ 16 points, 18 cases), a moderate trauma group (16 points < ISS score < 25 points, 16 cases), and a severe trauma group (ISS score ≥ 25 points, 14 cases). Another 22 adults under physical examination in our hospital were selected as the normal control group. The peripheral blood of each group was drawn for testing, and the serum eHSP70 and Ang 2 levels, APACHE II, and ISS score were compared. According to the prognosis of trauma patients, they were divided into survival group and death group, and the serum eHSP70 and Ang 2 levels of the subjects in each group were compared. The correlation between the serum eHSP70 level, Ang 2 level, ISS score, and APACHE II of trauma patients was further analyzed.

Results: The APACHE II and ISS score of patients in the mild trauma group, moderate trauma group, and severe trauma group were significantly higher than those in the normal control group ($P < 0.05$). Moreover, the APACHE II and ISS scores in the mild and moderate trauma groups were significantly lower than those in the severe trauma group ($P < 0.05$). No significant difference was revealed in APACHE II between the mild trauma group and the moderate trauma group ($P > 0.05$). The ISS score in the moderate trauma group was significantly higher than that in the mild trauma group ($P < 0.05$). The levels of eHSP70 and Ang 2 in the mild trauma group, moderate trauma group, and severe trauma group were significantly higher than those in the normal control group ($P < 0.05$). The levels of eHSP70 and Ang 2 in the mild and moderate trauma groups were significantly lower than those in the severe trauma group ($P < 0.05$). No significant difference was seen in the levels of eHSP70 and Ang 2 between the mild trauma group and the moderate trauma group ($P > 0.05$). The eHSP70 and Ang 2 levels in the survival group were significantly higher than those in the normal control group ($P < 0.05$). The eHSP70 and Ang 2 levels in the death group were significantly higher than those in the survival group ($P < 0.05$). eHSP70 was taken as the dependent variable, and Ang 2, ISS score, and APACHE II were taken as independent variables to analyze the correlation. Results showed that eHSP70 was positively correlated to Ang 2, ISS score, and APACHE II ($P < 0.05$).

Conclusion: The serum eHSP70 and Ang 2 levels, ISS score, and APACHE II of trauma patients are significantly increased, and the serum eHSP70 level is positively correlated to Ang 2 level, ISS score, and the APACHE II of trauma patients.

Keywords: Trauma, eHSP70, Ang 2, ISS, APACHE II.

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Introduction

Trauma caused by falls and traffic has become one of the public hazards that threatens human life and health in modern society. Trauma can cause a series of related biological effects in the body, including endothelial cell dysfunction in severe cases, which leads to subsequent infections and multiple organ tissue disorders, possibly endangering

the lives of patients^(1, 2). At present, the study of the pathogenesis of trauma and intervention methods is not deep enough, so treatment has become the focus and challenge of clinical practice.

Clinical reports have shown that tissue muscle cells will secrete relevant endogenous substances under injury and stimulation that can bind to pattern-recognition receptors and cooperate with pathogen-related molecules to further activate the innate

immune system, which ultimately leads to increased tissue and organ damage⁽³⁾. Extracellular heat shock protein70 (eHSP70) is an endogenous substance with an immune activity regulation function, which acts as a “molecular chaperone” in cells⁽⁴⁾. Clinical studies have shown that eHSP70 can participate in anti-apoptosis, anti-oxidative stress, and other links, which also have immune regulation functions⁽⁵⁾.

Clinical statistics have suggested that the expression level of angiotensin-2 (Ang 2) of patients increased significantly in the early stage of trauma without factors such as bacteria and infection; therefore, it can be believed that in addition to bacteria and infection, there is also a class of substances that will act to increase Ang 2⁽⁶⁾. The acute physiology and chronic health evaluation II (APACHE II) and injury severity score (ISS) can predict the severity and prognosis of critically ill patients, creating treatment conditions for medical staff to timely and effectively improve the rescue success rate⁽⁷⁾.

However, there are few studies on the relevance of the above serum indicators and scores in trauma patients. Therefore, this study aims to explore these relationships, and the results are as follows.

Materials and methods

General data

Forty-eight trauma patients who were treated in our hospital from June 2018 to December 2019 were selected.

Inclusion criteria for the study comprised the following factors:

- Patients with sudden acute trauma;
- Patients who had 2 or more anatomical parts or organ injuries, and one of them was a fatal injury;
- The patient and family members signed an informed consent form;
- The study had submitted an application to the ethics committee of our hospital and obtained approval.

Exclusion criteria for the study comprised:

- Those with acute cerebrovascular disease and malignant tumors;
- Patients who had used drugs that affect immune function in the past 3 months;
- Those who drank a lot of alcohol before the injury or took sedative drugs;
- Those with heart, liver, or kidney dysfunction.

According to the ISS score, 48 trauma patients were divided into a mild trauma group (ISS score

≤16 points), a moderate trauma group (16 points < ISS score < 25 points), and a severe trauma group (ISS score ≥25 points). Eighteen patients were in the mild trauma group, including 10 male patients and 8 female patients, with an average age of 36.15 (±5.49) years. Sixteen patients were in the moderate trauma group, with 8 male and 8 female patients and an average age of 35.01 (±5.96) years.

Finally, 14 patients were in the severe trauma group, 8 male and 6 female patients, with an average age of 37.86 (±4.89) years. In addition, 22 adults who received a physical examination in our hospital during the same period were collected as the normal control group, including 12 males and 10 females, with an average age of 35.16 (±5.19) years. No statistically significant difference in the general data of subjects in each group was identified after statistical software testing ($P > 0.05$).

Method

The peripheral blood of subjects in each group was separately extracted into a centrifuge tube and placed in a tabletop centrifuge at 3000 r·min⁻¹ for 5 min. After centrifugation, the supernatant was taken and placed in an ultra-low temperature refrigerator at -80°C for experimentation.

Observation indicators

- The levels of eHSP70 and Ang 2 in the serum of each group of experimental subjects were measured by enzyme-linked immunosorbent assay (ELISA).
- The APACHE II, ISS score, and statistical analysis were performed on all subjects in each group.
- According to the prognosis of trauma patients, they were divided into a survival group or death group, and the serum eHSP70 and Ang 2 levels of subjects in each group were compared.
- Pearson's correlation coefficient was used to analyze the correlation between serum eHSP70 level and Ang 2 level, ISS score, and APACHE II in trauma patients.

Statistical methods

All data in this study are processed and analyzed using SPSS23.0. Measurement data with a normal distribution and uniform variance were expressed as ($\bar{x} \pm s$), and the two independent samples of normal distribution were assessed using a t-test. Single-factor analysis of variance was used for multi-sample comparisons. A correlation analysis between

serum eHSP70 level, Ang 2 level, ISS score, and APACHE II of trauma patients was processed by Pearson's correlation, and $P < 0.05$ was regarded as a statistically significant difference.

Results

Comparison of APACHE II and ISS score in patients with different trauma levels

The APACHE II and ISS scores of patients in the mild trauma group, moderate trauma group, and severe trauma group were significantly higher than those in the normal control group ($P < 0.05$). Levels in the mild and moderate trauma groups were significantly lower than those in the severe trauma group ($P < 0.05$). No significant difference was demonstrated in APACHE II between the mild trauma group and the moderate trauma group ($P > 0.05$). The ISS score of patients in the moderate trauma group was significantly higher than that in the mild trauma group ($P < 0.05$). See Table 1.

Groups	n	APACHE II (points)	ISS (points)
Normal control group	22	0.00	0.00
Mild trauma group	18	9.41±3.81 [△]	10.01±3.02 [△]
Moderate trauma group	16	9.22±3.91 [△]	18.23±1.21 ^{*△}
Severe trauma group	14	20.81±5.81 [*]	30.22±4.51 [*]

Table 1: Comparison of APACHE II and ISS scores in patients with different trauma levels ($\bar{x} \pm s$).

Note: Compared with the normal control group, ^{*} $P < 0.05$; compared with the mild trauma group, [#] $P < 0.05$; compared with the severe trauma group, [△] $P < 0.05$.

Comparison of serum eHSP70 and Ang 2 levels in patients with different trauma levels

The levels of eHSP70 and Ang 2 in the mild trauma group, moderate trauma group, and severe trauma group were significantly higher than those in the normal control group ($P < 0.05$). The levels in the mild and moderate trauma groups were significantly lower than those in the severe trauma group ($P < 0.05$). There was no significant difference in the levels of eHSP70 and Ang 2 between the mild trauma group and the moderate trauma group ($P > 0.05$).

Comparison of eHSP70 and Ang 2 levels between the survival group and the death group

The eHSP70 and Ang 2 levels in the survival group were significantly higher than those in the normal control group ($P < 0.05$). The eHSP70 and Ang 2 levels in the death group were significantly higher than those in the survival group ($P < 0.05$). See Table 3.

Groups	n	eHSP70 (ng/mL)	Ang 2 (pg/mL)
Normal control group	22	1.44±0.39	455.91±136.12
Mild trauma group	18	4.40±0.40 [*]	2130.11±1025.36 [*]
Moderate trauma group	16	4.95±0.56 [*]	3380.25±1589.87 [*]
Severe trauma group	14	13.04±3.92 [*]	8990.12 ± 3102.46 [*]

Table 2: Comparison of serum eHSP70 and Ang 2 levels in patients with different trauma levels ($\bar{x} \pm s$).

Note: Compared with the normal control group, ^{*} $P < 0.05$; compared with the mild trauma group, [#] $P < 0.05$; compared with the severe trauma group, [△] $P < 0.05$.

Groups	n	eHSP70 (ng/mL)	Ang 2 (pg/mL)
Normal control group	22	1.44±0.39	455.91±136.12
Survival group	37	4.49±1.23 [*]	965.48±201.15 [*]
Death group	11	13.04±2.49 ^{*#}	8825.15±3025.13 ^{*#}

Table 3: Comparison of eHSP70 levels between the survival group and the death group at different time periods ($\bar{x} \pm s$).

Note: Compared with the normal control group, ^{*} $P < 0.05$; compared with the survival group, [#] $P < 0.05$.

Correlation analysis of serum eHSP70 level and Ang 2 level, ISS score, and APACHE II in trauma patients

eHSP70 was taken as the dependent variable, and Ang 2, ISS score, and APACHE II were taken as independent variables to analyze the correlation. eHSP70 was positively correlated to Ang 2, ISS score, and APACHE II ($P < 0.05$). See Table 4.

Indicators	eHSP70	
	r	P
Ang2	0.576	0.015
ISS	0.459	0.042
APACHE II	0.323	0.039

Table 4: Correlation analysis between serum eHSP70 level and Ang 2 level, ISS score, and APACHE II score.

Discussion

At present, the risk of severe trauma in humans is increasing daily, and the mortality of patients is also increasing. Therefore, it is particularly important to explore methods that can effectively diagnose and treat trauma patients.

Clinical studies have shown that when predicting the prognosis of patients with multiple injuries in the emergency department, a combination of physiological-based quick scoring measures and ISS scores can be selected⁽⁸⁾. Therefore, this

study used APACHE II and ISS scores to evaluate the patient's condition. The results found that the severe trauma group had the highest APACHE II and ISS scores. eHSP70 is the current hot spot for molecular research on injury-related patterns and can be detected in the serum of people with pregnancy, hypertension, and rheumatoid arthritis⁽⁹⁾. Clinical reports have shown that the release of cell necrosis and the active secretion of the cell are the main pathways for the formation of eHSP70. Some scholars have proposed that recombinant HSP70 can activate macrophages, thereby promoting intracellular calcium levels and cytokine secretion, ultimately aggravating the damage⁽¹⁰⁾. Other scholars found that eHSP70 can act on the contractile function of cardiomyocytes to a certain extent and play an important role; in addition, eHSP70 can also promote cancer cell growth and inhibit cell apoptosis⁽¹¹⁾. The above results indicate that the increase of serum eHSP70 level can participate in the pathophysiology of trauma. Many reports have confirmed that eHSP70 can be used as a sign of death in male patients with severe head trauma, and its increased level is expected to become an important reference indicator for evaluating severe head injury⁽¹²⁾. Scholars have shown that eHSP70 is significantly increased in the serum of children who died of craniocerebral injury, which is an important marker for evaluating the mortality of children with severe traumatic craniocerebral injury. However, the sample size of this report is too small and insufficient to confirm this conclusion⁽¹³⁾.

Ang 2 can be used as an important reference for the dysfunction of vascular endothelial cells that exists in the Weibel–Palade body. Under the induction of many factors, the Weibel–Palade body can secrete Ang 2 in exocellular form, leading to the destruction of the endothelium, and then playing an important role in the physiological and pathological process of septic shock and other diseases⁽¹⁴⁾. Human sepsis-related reports showed that the plasma Ang 2 level in patients with septic shock is significantly higher than that in a normal control group. In addition, the blood circulation Ang 2 level is closely related to the severity of the disease and the risk of death in patients with sepsis⁽¹⁵⁾. Animal experiments confirmed that the level of Ang 2 in the sepsis group was significantly increased. After in-depth research, it was concluded that recombinant Ang 2 can cause damage to the endothelial barrier function and structure. The above results all indicate that the destruction of endothelial cells in trauma patients

could lead to an increase in Ang 2 levels. Previous reports have suggested that Ang 2 can be highly expressed in sepsis caused by cells and fungi, and its level is positively correlated with APACHE II and the sequential organ failure score, which can be used as an independent risk factor for the severity of patients with sepsis. Other scholars have found that changing the expression level of the Toll-like receptor 2 can lead to changes in the endothelial cell Weibel–Palade body exocytosis, which in turn leads to an increase in Ang 2 levels. This study showed that the levels of eHSP70 and Ang 2 in the severe trauma group increased significantly compared with the other two groups. Moreover, the levels increased after trauma, indicating that severe trauma patients suffered more damage, tissue cells suffered greater damage, and the stress response of patients was strong. Therefore, both eHSP70 and Ang 2 can be used as biological markers for evaluating the condition of trauma patients. We found that the eHSP70 and Ang 2 levels in the death group were significantly higher than those in the survival group ($P < 0.05$), showing that eHSP70 and Ang 2 levels can be used as important reference indicators for evaluating the prognosis of trauma patients. In addition, we used the Pearson correlation coefficient to analyze the correlation between serum eHSP70 levels and Ang 2 levels, ISS scores, and APACHE II scores.

We found that serum eHSP70 in trauma patients was positively correlated with Ang 2, ISS scores, and APACHE II scores ($P < 0.05$).

In summary, the serum levels of eHSP70, Ang 2, ISS score, and APACHE II of trauma patients were significantly increased in comparison to the control group. In addition, the serum eHSP70 level was positively correlated with the Ang 2 level, ISS score, and APACHE II scores of trauma patients.

References

- 1) Brown CVR, Alam HB, Brasel K, Hauser CJ, de Moya M, et al. Western Trauma Association Critical Decisions in Trauma: Management of Renal Trauma. *J Trauma Acute Care Surg* 2018; 85: 1021-1025.
- 2) Kinder F, Giannoudis PV, Boddice T, Howard A. The Effect of an Abnormal BMI on Orthopaedic Trauma Patients: A Systematic Review and Meta-Analysis. *J Clin Med* 2020; 9: 1302-1308.

- 3) Allen BK, Callaway DW, Gibbs M, Noste E, West K, et al. Regarding the Joint statement from the American College of Surgeons Committee on Trauma (ACS COT) and the American College of Emergency Physicians (ACEP) regarding the clinical use of Resuscitative Endovascular Balloon Occlusion of the Aorta (REBOA). *Trauma Surg Acute Care Open* 2018; 3: 168.
- 4) Aboutaleb N, Faezi M, Nasser Maleki S, Nazarinia D, Razavi Tousi SMT, et al. Conditioned medium obtained from mesenchymal stem cells attenuates focal cerebral ischemia reperfusion injury through activation of ERK1/ERK2-BDNF signaling pathway. *J Chem Neuroanat* 2019; 97: 87-98.
- 5) Cesa LC, Shao H, Srinivasan SR, Tse E, Jain C, et al. X-linked inhibitor of apoptosis protein (XIAP) is a client of heat shock protein 70 (Hsp70) and a biomarker of its inhibition. *J Biol Chem* 2018; 293: 2370-2378.
- 6) Mueller T, Freystein J, Lucas H, Schmoll H. Efficacy of a Bispecific Antibody Co-Targeting VEGFA and Ang 2 in Combination with Chemotherapy in a Chemoresistant Colorectal Carcinoma Xenograft Model. *Molecules* 2019; 24: 2865-2869.
- 7) Shi YJ, Zheng GH, Qian LY, Qsman RA, Li GG, et al. Longitudinal Analysis of Risk Factors for Clinical Outcomes of Enterobacteriaceae Meningitis/Encephalitis in Post-Neurosurgical Patients: A Comparative Cohort Study During 2014-2019. *Infect Drug Resist* 2020; 13: 2161-2170.
- 8) Docherty AB, Sim M, Oliveira J, Adlam M, Ostermann M, et al. Early troponin I in critical illness and its association with hospital mortality: a cohort study. *Crit Care* 2017; 21: 216.
- 9) Tsuboyama K, Tadakuma H, Tomari Y. Conformational Activation of Argonaute by Distinct yet Coordinated Actions of the Hsp70 and Hsp90 Chaperone Systems. *Molecular Cell* 2018; 70: 722-729.
- 10) Radons J. The human HSP70 family of chaperones: where do we stand? *Cell Stress Chaperones* 2016; 21: 379-404.
- 11) Zaiter SS, Huo Y, Tiew FY, Gestwicki JE, McAlpine SR. Designing de Novo Small Molecules That Control Heat Shock Protein 70 (Hsp70) and Heat Shock Organizing Protein (HOP) within the Chaperone Protein-Folding Machinery. *J Med Chem* 2019; 62: 742-761.
- 12) Zhou Y, Wang BL, Yang M, He L, Qin L, et al. Construction of Mycoplasma ovipneumoniae pcDNA3.1-TBP30-Hsp70 Fusion Plasmid and Its Effect on Cellular Immune Response in Mice. *China Animal Husbandry Veterinary Med* 2019; 3: 15-21.
- 13) Zhai E, Liang W, Lin Y, Huang LL, He X, et al. HSP70/HSP90-Organizing Protein Contributes to Gastric Cancer Progression in an Autocrine Fashion and Predicts Poor Survival in Gastric Cancer. *Cell Physiol Biochem* 2018; 47: 879-892.
- 14) Li WX, Wei D, Xie XM, Liang JY, Song KP, et al. DL-3-n-Butylphthalide regulates the Ang-1/Ang 2/Tie-2 signaling axis to promote neovascularization in chronic cerebral hypoperfusion. *Biomed Pharmacother* 2019; 113: 108757.
- 15) O'Brien K, Saravanabavan S, Zhang JQJ, Wong ATY, Munt A, et al. Regression of Peritubular Capillaries Coincides with Angiogenesis and Renal Cyst Growth in Experimental Polycystic Kidney Disease. *Int J Nephrol Renovasc Dis* 2020; 13: 53-64.

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