Relationship of plasma proadrenomedullin and cortisol levels with systemic inflammatory response and target organ damage in children with sepsis after burn

Xing Wei, Jie Ou−Yang✉, Chen Zhou

Anesthesiology Department, the Second Affiliated Hospital of Kunming Medical University in Yunnan Province, Kunming City, Yunnan Province, 650101, China

ARTICLE INFO

ABSTRACT

Objective: To study the relationship of plasma proadrenomedullin (pro-ADM) and cortisol (Cor) levels with systemic inflammatory response and target organ damage in children with sepsis after burn. **Methods:** A total of 30 children with sepsis after burn who were treated in the hospital between August 2014 and August 2016 were collected as observation group, and 30 normal children who received vaccination in the hospital during the same period were collected as normal control group. The pro-ADM and Cor levels in plasma as well as the levels of inflammatory factors, myocardial injury markers and intestinal barrier function indexes in serum of the two groups were determined. Pearson test was used to assess the correlation of plasma pro-ADM and Cor levels with systemic inflammatory response and target organ damage in patients with sepsis after burn. **Results:** Plasma pro-ADM and Cor levels in observation group were higher than those in normal control group. Serum inflammatory cytokines IL-1, IL-6, IL-10 and TNF- α levels in observation group were higher than those in normal control group; serum myocardial injury markers CK-MB, cTnⅠ and NT-proBNP levels were higher than those in normal control group; serum intestinal barrier function indexes ET, DAO and D-L levels were higher than those in normal control group. **Conclusion:** Plasma pro-ADM and Cor levels increase in patients with sepsis after burn, and are highly consistent with systemic inflammatory response and target organ injury.

1. Introduction

Sepsis after burn is a common complication after deep burn, which is generally with single bacterial infection in early stage and with mixed infection of a variety of bacteria in late stage, occurs within 2 weeks after burn, and is easily complicated with shock and multiple organ failure[1,2]. Clarifying burn sepsis severity is of great significance for the determination of the follow-up treatment, proadrenomedullin (pro-ADM) can indirectly reflect the adrenal medulla levels in the circulating blood, and studies show that it is better than acute physiology and chronic health evaluation

(APACHEⅡ) score in evaluating the prognosis in sepsis[3]. Cortisol (Cor) is a typical stress hormone, which is massively secreted under the stimulation of intense stressors such as burns and infections, and can be used to determine the severity of systemic stress[4,5]. In the study, plasma pro-ADM and Cor levels in children with burn sepsis were determined, and the inner link between their levels and the disease severity was further discussed in order to find sensitive markers for early judgment of burn sepsis, now reported as follows.

2. Information and methods

2.1 Case information

A total of 30 children with sepsis after burn who were treated in the hospital between August 2014 and August 2016 were selected as observation group, 30 normal children who received vaccination in

the hospital during the same period were selected as normal control group, and the families of research subjects signed informed consent. Observation group included 16 male cases and 14 female cases that were 2-9 years old; normal control group included 15 male cases and 15 female cases that were 1-7 years old. The gender and age distribution of the two groups were similar, and the hospital ethics committee approved the study.

2.2 Blood sample preparation

Immediately after admission, 2.0 mL of cubital venous blood was extracted from two groups of research subjects, anti-coagulated, then let stand at room temperature for stratification and centrifuged at 2 500-3 500 r/min for 10 min to separate upper serum and lower plasma, which were cryopreserved in cryogenic environment for test.

2.3 Plasma proadrenomedullin and cortisol levels

The proadrenomedullin (pro-ADM) and cortisol (Cor) levels in plasma were detected by immunochromatography kit.

2.4 Serum inflammatory factors

Enzyme-linked immunosorbent assay was used to determine serum interleukin-1 (IL-1), interleukin-6 (IL-6), interleukin-10 (IL-10) and tumor necrosis factor α (TNF-α) levels.

2.5 Target organ injury indexes

Automatic microparticle chemiluminescence enzyme immunoassay analyzer was used to detect serum myocardial injury markers creatine kinase (CK-MB), troponin Ⅰ (cTnⅠ) and N-terminal pro-brain natriuretic peptide (NT-proBNP) levels. The contents of endotoxin (ET), diamine oxidase (DAO) and D-lactate (D-L) in serum were measured by the chromogenic substrate limulus amebocyte lysate.

2.6 Statistical processing

pro-ADM, Cor, inflammatory factors, myocardial injury markers and intestinal barrier function indexes belonged to measurement data and were in terms of (Mean ± SD), and comparison between two groups was by grouping t test. Statistical software was SPSS 24.0, and statistics $P<0.05$ indicated statistical significance in differences.

3. Results

3.1 Plasma pro–ADM and Cor

Comparison of plasma pro-ADM (nmol/L) and Cor (pmol/L) levels between two groups of research subjects was as follows: plasma pro-ADM level in observation group was (1.46±0.17) nmol/L, Cor level

was (1093.27±146.19) pmol/L, and both were significantly higher than those in normal control group. Differences in plasma pro-ADM and Cor levels were statistically significant between two groups of research subjects ($P<0.05$), shown in Table 1.

Table 1.

Comparison of plasma pro-ADM and Cor levels between two groups of research subjects.

Groups	n	pro-ADM	Cor
Control group	30	0.54±0.08	342.14±39.66
Observation group	30	1.46±0.17	1 093.27±146.19
t		7.291	27.384
P		＜0.05	＜0.05

3.2 Serum inflammatory factors

Comparison of serum inflammatory factors IL-1, IL-6, IL-10 and TNF-α levels between two groups of research subjects was as follows: serum IL-1, IL-6, IL-10 and TNF-α levels in observation group were significantly higher than those in normal control group. Differences in serum inflammatory factors IL-1, IL-6, IL-10 and TNF-α levels were statistically significant between two groups of research subjects ($P<0.05$), shown in Table 2.

Table 2.

Comparison of serum inflammatory factor levels between two groups of research subjects (pg/mL).

Groups	n	IL-1	IL-6	IL-10	TNF-α
Control group	30	14.27±1.82	15.28±2.11	14.52±1.89	20.63±3.42
Observation group	30	22.13±2.75	24.36±3.05	34.74±4.16	40.28±5.11
T		9.182	10.482	15.282	20.651
P		＜0.05	＜0.05	＜0.05	＜0.05

3.3 Myocardial injury markers

Comparison of myocardial injury markers CK-MB (IU/L), cTnⅠ (ng/mL) and NT-proBNP (pg/mL) levels between two groups of research subjects was as follows: serum CK-MB, cTnⅠ and NT-proBNP levels in observation group were significantly higher than those in normal control group. Differences in serum CK-MB, cTnⅠ and NT-proBNP levels were statistically significant between two groups of research subjects ($P<0.05$), shown in Table 3.

Table 3.

Comparison of serum myocardial injury marker levels between two groups of research subjects.

Groups	n	CK-MB	cTnⅠ	NT-proBNP
Control group	30	9.75±1.63	1.16±0.14	602.77±68.95
Observation group	30	34.28±4.51	1.58±0.23	1 538.26±187.15
T		16.827	7.182	25.492
P		＜0.05	＜0.05	＜0.05

3.4 Intestinal barrier function indexes

Comparison of serum intestinal barrier function indexes levels between two groups of research subjects was as follows: serum ET, DAO and D-L levels in observation group were significantly higher

than those in normal control group. Differences in serum intestinal barrier function indexes ET, DAO and D-L levels were statistically significant between two groups of research subjects ($P<0.05$), shown in Table 4.

Table 4.

Comparison of serum intestinal barrier function index levels between two groups of research subjects.

Groups	n	ET	DAO	D-L
Control group	30	0.27±0.04	2.05±0.34	7.53±0.86
Observation group	30	1.05±0.21	11.73±1.95	26.47±3.51
T		7.287	9.271	12.371
P		<0.05	<0.05	<0.05

3.5 Correlation analysis

The correlation of plasma pro-ADM and Cor levels in children with sepsis after burn with systemic inflammatory response and target organ damage was as follows: Pearson test showed that plasma pro-ADM and Cor levels in children with sepsis after burn were positively correlated with serum inflammatory cytokines IL-1, IL-6, IL-10 and TNF-α levels, serum myocardial injury markers CK-MB, cTnⅠ and NT-proBNP levels, and serum intestinal barrier function indexes ET, DAO and D-L levels ($P<0.05$).

4. Discussion

Burn sepsis is a clinical critical condition, and it can cause short-term shock death especially in young children[6,7]. Early judgment of the disease severity is the basis to choose the reasonable means of intervention, and also the key to optimize the treatment outcome, hematology indexes have the advantage of simple and easy sampling and quick test results obtaining, and therefore, looking for sensitive circulating blood indexes for sepsis is the focus of current clinical research. Pro-ADM has high specificity in the diagnosis of sepsis, and it has immunomodulation, anti-infection and other effects. It has been found that the pro-ADM content increases in many infectious diseases[8]. There is generally increased Cor level in circulating blood of children with sepsis, which is mainly because that the toxic molecules stimulate the HPA axis and cause ACTH secretion to increase[9]. In order to define the pro-ADM and Cor value for early diagnosis of sepsis after burn, pro-ADM and Cor contents in plasma were compared between two groups of subjects in this study, and it was found that compared with normal control group, the observation group were with higher plasma pro-ADM and Cor contents, confirming that the highly expressed pro-ADM and Cor are the standard of burn sepsis. The correlation of pro-ADM and Cor contents with burn sepsis remains to be further studied below, which is specifically elaborated from the three aspects of systemic inflammatory response, myocardial injury and intestinal barrier function indexes.

Systemic inflammatory response is the most typical manifestation of patients with burn sepsis, and the pro-inflammatory mediators massively synthesized and secreted in the body will further induce neutrophil aggregation and stimulate continuous synthesis of pro-inflammatory factors, thus forming the inflammatory cascade reaction[10,11]. There is pro-inflammatory/anti-inflammatory imbalance in children with sepsis, the levels of IL-1, IL-6, TNF-α and other pro-inflammatory factors increase significantly, the levels of IL-10 and other anti-inflammatory factors increased reactively, but the increase is as much as that of pro-inflammatory factors, finally anti-inflammatory factors cannot completely neutralize the inflammatory effect of proinflammatory factors, and it leads to the occurrence and progression of systemic inflammatory reaction[12,13]. In the study, the serum levels of above inflammatory factors were compared between the two groups, and it was found that compared with normal control group, the observation group were with serum higher serum levels of pro-inflammatory factors IL-1, IL-6 and TNF-α as well as higher level of anti-inflammatory factor IL-10, confirming that that there are abnormally highly expressed pro-inflammatory and anti-inflammatory factors in children with burn sepsis, and they are involved in the occurrence of systemic inflammatory response together. Further Pearson test showed that plasma pro-ADM and Cor levels in children with sepsis after burn were positively correlated with inflammatory cytokines IL-1, IL-6, IL-10 and TNF-α levels, confirming that the highly expressed pro-ADM and Cor can directly increase systemic inflammatory response. Sustained progress in sepsis patients can lead to multiple organ dysfunction syndromes, the heart is the most easily involved important organ, and the great majority of children died from heart failure or severe myocarditis[14,15]. CK-MB and cTnⅠ are the typical myocardial enzyme spectrum indexes, which are highly expressed in serum early after myocardial injury; NT-proBNP is a specific indicator of cardiac failure, and the serum NT-proBNP content increases when the cardiac function of children with sepsis declines[16]. In this study, the serum levels of myocardial injury markers were compared between the two groups, and it was found that compared with normal control group, the observation group were with higher serum CK-MB, cTnⅠ and NT-proBNP contents, confirming that that there is obvious myocardial injury in children with burn sepsis. Further Pearson test showed that plasma pro-ADM and Cor levels in children with sepsis after burn were positively correlated with myocardial injury markers CK-MB, cTnⅠ and NT-proBNP levels, indicating that highly expressed pro-ADM and Cor can directly prompt myocardial injury occurrence, and is a sign of myocardial injury appearance.

Because of the massive toxin generation and blood circulation disorder, children with sepsis are mostly with intestinal barrier

dysfunction, which is characterized by the increase of serum ET content[17]. 95% of DAO exists in the villus cells on mammalian intestinal mucosa, and the DAO in the cells can be released into the blood when various pathological causes cause intestinal mucosal injury. D-L is the metabolite of intestinal bacterial fermentation and can enter the blood circulation when the intestinal permeability increases, so the D-L content can objectively reflect the change and damage degree of intestinal mucosal permeability[18,19]. In this study, serum levels of these intestinal barrier function indexes were compared between the two groups, and it was found that compared with normal control group, the observation group were with higher serum ET, DAO and D-L contents, confirming that there is intestinal mucosal barrier dysfunction in children with burn sepsis. Further Pearson test showed that plasma pro-ADM and Cor levels in children with sepsis after burn were positively correlated with intestinal barrier function indexes ET, DAO and D-L levels, indicating that the highly expressed pro-ADM and Cor can induce intestinal mucosal barrier injury.

The abnormally high plasma pro-ADM and Cor levels in children with burn sepsis can lead to increased systemic inflammatory response, myocardial injury and intestinal mucosa barrier dysfunction. Early detection of plasma pro-ADM and Cor contents is expected to be the reliability index for burn sepsis diagnosis and disease severity judgment, and helps to choose the reasonable treatment and optimize the final treatment outcome.

References

[1] Chen L, Meng K, Su W, Fu Y. The effect of continuous sedation therapy on immunomodulation, plasma levels of antioxidants, and indicators of tissue repair in post-burn sepsis patients. *Cell Biochem Biophys* 2015; **73**(2): 473-478.

[2] Wang SX, Liu QY, Li Y. Lentinan ameliorates burn sepsis by attenuating CD4+ CD25+ Tregs. *Burns* 2016; **42**(7): 1513-1521.

[3] Balcan B, Olgun S, Akbas T, Eryuksel E, Karakurt S. Level of adrenomedullin in cases with adrenal defficiency and its relation to mortality in patients with sepsis. *Tuberk Toraks* 2016; **64**(3): 191-197.

[4] Kertai MD, Fontes ML. Predicting adrenal insufficiency in severe sepsis: the role of plasma-free cortisol. *Crit Care Med* 2015; **43**(3): 715-716.

[5] Venkatesh B, Imeson L, Kruger P, Cohen J, Jones M, Bellomo R et al. Elevated plasma-free cortisol concentrations and ratios are associated with increased mortality even in the presence of statin therapy in patients with severe sepsis. *Crit Care Med* 2015; **43**(3): 630-635.

[6] Long H, Xu B, Luo Y, Luo K. Artemisinin protects mice against burn sepsis through inhibiting NLRP3 inflammasome activation. *Am J Emerg Med* 2016, **34**(5): 772-777.

[7] Xu X, Shi Z, Hu J, Yuan B, Huang H, Fang H, et al. Identification of differentially expressed genes associated with burn sepsis using microarray. *Int J Mol Med* 2015; **36**(6): 1623-1629.

[8] Oncel MY, Erdeve O, Uras N, Dilmen U. Is pro-adrenomedullin a more useful marker in hospitalized infants with sepsis. *Eur J Pediatr* 2014; **173**(1): 127-128.

[9] Koskela M, Ala-Kokko TI, Gaddnas F, Herzig KH, Karhu T, Oikarinen A, et al. Blister fluid and serum cytokine levels in severe sepsis in humans reflect skin dysfunction. *Acta Anaesthesiol Scand* 2017; **61**(1): 53-61.

[10] Virzi GM, Clementi A, Brocca A, de Cal M, Marcante S, Ronco C. Cardiorenal syndrome type 5 in sepsis: role of endotoxin in cell death pathways and inflammation. *Kidney Blood Press Res* 2016; **41**(6): 1008-1015.

[11] Lobo LA, Benjamim CF, Oliveira AC. The interplay between microbiota and inflammation: lessons from peritonitis and sepsis. *Clin Transl Immunol* 2016; **5**(7): e90.

[12] Mertens K, Lowes DA, Webster NR, Talib J, Hall L, Davies MJ, et al. Low zinc and selenium concentrations in sepsis are associated with oxidative damage and inflammation. *Br J Anaesth* 2015; **114**(6): 990-999.

[13] Escobar DA, Botero-Quintero AM, Kautza BC, Luciano J, Loughran P, Darwiche S, et al. Adenosine monophosphate-activated protein kinase activation protects against sepsis-induced organ injury and inflammation. *J Surg Res* 2015; **194**(1): 262-272.

[14] Bower G, Ashrafian H, Cappelletti S, Lee LM, Harling L, Ciallella C, et al. A proposed role for sepsis in the pathogenesis of myocardial calcification. *Acta Cardiol* 2017; **72**(3): 249-255.

[15] Wang H, Cui Z, Sun F, Ding H. Glucan phosphate inhibits HMGB-1 release from rat myocardial H9C2 cells in sepsis via TLR4/NF-κB signal pathway. *Clin Invest Med* 2017; **40**(2): E66-E72.

[16] Haileselassie B, Su E, Pozios I, Nino DF, Liu H, Lu DY, et al. Myocardial oxidative stress correlates with left ventricular dysfunction on strain echocardiography in a rodent model of sepsis. *Intensive Care Med Exp* 2017; **5**(1): 21.

[17] Wang L, Cui YL, Zhang Z, Lin ZF, Chen DC. Rhubarb monomers protect intestinal mucosal barrier in sepsis via junction proteins. *Chin Med J (Engl)* 2017; **130**(10): 1218-1225.

[18] Gu GS, Ren JA, Li GW, Yuan YJ, Li N, Li JS. Cordyceps sinensis preserves intestinal mucosal barrier and may be an adjunct therapy in endotoxin-induced sepsis rat model: a pilot study. *Int J Clin Exp Med* 2015; **8**(5): 7333-7341.

[19] Liu H, Liu Z, Zhao S, Sun C, Yang M. Effect of BML-111 on the intestinal mucosal barrier in sepsis and its mechanism of action. *Mol Med Rep* 2015; **12**(2): 3101-3106.