EFFECTS OF GLUTAMINE ON INFLAMMATORY CASCADE, SERUM HIGH MOBILITY GROUP PROTEIN 1 AND HEAT SHOCK PROTEIN IN PATIENTS WITH SEVERE PANCREATITIS

Le Zhang[1], Fei Li[1,*], Yuekui Bai[2], Kaidong Liu[2], Ming Liu[2]
[1]Department of General Surgery, Xuanwu Hospital Capital Medical University, Beijing, PR China - [2]Department of General Surgery, Beijing Haidian Hospital, Beijing, PR China

ABSTRACT

Objective: *To investigate the effect of glutamine on inflammatory cascade, serum high mobility group protein 1 (HMGB1) and heat shock protein (HSP) in patients with severe pancreatitis.*

Methods: *From June 2016 to November 2018, 90 patients with severe pancreatitis were randomly selected from the general surgery department of our hospital. According to the random number table method, they were randomly divided into the research group and the control group, with 45 patients in each group. Patients in the control group were given routine treatment, such as fasting, nutritional support, antispasmolysis and pain relief, control of pancreatic secretion and anti-infection. Patients in the study group were treated with glutamine on the basis of the control group. All patients received continuous treatment for two weeks. To observe the clinical efficacy of the two groups of patients, inflammation [tumour necrosis factor α(TNF-α), interleukin 8 (IL-8) and interleukin 1β (IL-1β), interleukin 6 (IL-6) and C-reactive protein (CRP)], serum levels of HMGB1, HSP, length of hospital stay, compare two groups of patients' serum amylase recovery time and duration of abdominal pain relief.*

Results: *After treatment, the clinical efficacy of the study group was 93.33%, significantly higher than the 77.78% of the control group (P<0.05). Compared with before treatment, TNF-a, IL-8, IL-1β, IL-6 and CRP levels in both groups were significantly reduced after treatment, while the levels of all indicators in the study group were significantly reduced (P<0.05). After treatment, serum HMGB1 and HSP levels in the two groups were significantly lower than before treatment, and serum HMGB1 and HSP levels in the study group were significantly lower than those in the control group (P<0.01). Compared with the control group, the hospitalization time, serum amylase recovery time and abdominal pain relief time were significantly reduced in the study group (P<0.01).*

Conclusion: *Glutamine has obvious clinical efficacy in the treatment of severe pancreatitis, can significantly reduce the inflammatory cascade reaction of patients, reduce serum HMGB1 and HSP levels, and has good clinical application value.*

Keywords: *Glutamine, severe pancreatitis, inflammatory cascade, high mobility group protein 1, heat shock protein.*

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Introduction

Acute pancreatitis is an inflammatory response that causes digestion, oedema, bleeding and even necrosis of pancreatic tissue after various causes activate pancreatic enzymes. Clinical manifestations include acute epigastric pain, nausea, vomiting, fever, and elevated blood trypsin. Most patients with pancreatic oedema have a good prognosis, while most patients with severe pancreatic haemorrhage and necrosis often have secondary infection, peritonitis and shock, with a high mortality rate, which is called severe pancreatitis[(1)]. Many studies have found that the occurrence of severe pancreatitis is closely related to systemic inflammatory response syndrome caused by a large number of cytokine cascade reactions[(2)]. If the disease is not appropriately and effectively controlled, it will eventually develop into secondary organ failure, and even lead to death. It has been found that the release of tumour necrosis factor-α (TNF-α), interleukin-6 (IL-6), nuclear transcription factor ϰB (NF-ϰB), C-reactive protein (CRP) and other inflammatory mediators can trigger an inflammatory cascade, which is the vital link in the control of severe pancreatitis[(3)]. Serum high mobility group protein 1 (HMGB1) is a newly discovered late-stage pro-inflammatory medi-

um, which has the characteristics of late release and long maintenance time, and plays an important role in the development of severe pancreatitis[4]. Heat shock proteins (HSP), also known as heat shock proteins, are a class of functional related proteins, and the synthesis of HSP is a protective mechanism of cell survival in the stress state. Studies have found that HSP is closely related to inflammatory diseases, such as colitis and pancreatitis[5]. Glutamine is an important energy substance for intestinal mucosal epithelial cells and immune cells. It has been reported that glutamine can promote nitrogen balance, maintain intestinal mucosal integrity, and enhance the immune function of the body[6]. This study mainly investigated the effect of glutamine on inflammatory cascade, HMGB1 and HSP in patients with severe pancreatitis.

Data and methods

Basic information

From June 2016 to November 2018, 90 patients with severe pancreatitis were randomly selected from the general surgery department of our hospital. According to the random number table method, they were randomly divided into the research group and the control group, with 45 patients in each group.

Criteria for inclusion:

• All patients met the criteria for diagnosis and classification of severe acute pancreatitis in clinical research on the diagnosis and classification of severe acute pancreatitis[7];

• The patient is in a good mental state and can cooperate with treatment;

• The study was approved by the hospital committee, and the informed consent was signed by the patients' families.

Exclusion criteria:

• Severe liver and kidney function, cardiac insufficiency;

• Infectious diseases;

• A drug allergy history;

• Lactation or pregnancy patients. Among them, there were 30 males and 15 females in the study group, aged from 37 to 52 years old, with an average age of 48.66±4.16 years old and an average course of disease of 1.29±0.24 days.

In the control group, there were 27 males and 18 females, aged from 39 to 55 years old, with an average age of 49.49±3.53 years old and an average course of disease of 1.27±0.22 days. There was no significant difference in age and other fundamental data between the two groups (P>0.05). See Table 1.

Group	n	Average Age (year)	Gender (n)		The Average Duration (d)
			Male	Female	
Research Group	45	48.66±4.16	30	15	1.29±0.24
Control Group	45	49.49±3.53	27	18	1.27±0.22
t/χ^2		1.021	0.431		0.412
P		0.311	0.512		0.681

Table 1: Comparison of basic data between the two groups.

Treatment methods

Patients in the control group were given routine treatment, such as fasting, nutritional support, antispasmolysis and pain relief, control of pancreatic secretion and anti-infection. The patients in the study group were treated with glutamine (Huarui Pharmaceutical co., LTD., approval number: Chinese medicine approval number H20173409, specification: 100ml: 20g) on the basis of the control group. The glutamine injection was mixed with compatible amino acid solution and injected with carrier solution, 2ml/kg/d. All patients received continuous treatment for two weeks.

Statistical methods

Statistical data were analysed by SPSS21.0 software package and measurement data were compared by independent sample t-test. Chi-square test was used to compare the counter data. Ridit test was used to compare hierarchical data. P<0.05 was considered statistically significant.

Results

Comparison of clinical efficacy between the two groups

After treatment, the total effective rate was 93.33% in the research group and 77.78% in the control group, which was significantly higher than that in the control group (P<0.05). This is shown in Table 2.

Group	n	Cure	The Basic Cure	Improve	Invalid	Total Effective Rate
Research Group	63	21 (46.67)	13 (28.89)	8 (17.78)	3 (6.67)	42 (93.33)
Control Group	63	15 (33.33)	12 (26.67)	8 (17.78)	10 (22.22)	35 (77.78)
χ^2						4.406
P						0.036

Table 2: Comparison of clinical efficacy between the two groups (%).

Note: total effective rate= $\frac{(\text{cure + basic cure + improvement})}{n} \times 100\%$.

Comparison of inflammatory cytokines between the two groups

After treatment, the levels of TNF-a, IL-8, IL-1β, IL-6, CRP and other inflammatory factors in the two groups were significantly reduced compared with those before treatment, while the above indicators in the study group were significantly lower than those in the control group (P<0.05). See Table 3.

Group	n	Time	TNF-α (ng/L)	IL-8 (ng/L)	IL-1β (ng/L)	IL-6 (ng/L)	CRP (ng/L)
Research Group	45	Before treatment	47.68±7.36	34.79±3.43	0.33±0.04	21.43±3.19	6.72±1.32
		After treatment	7.49±2.14[ab]	11.55±2.48[ab]	0.12±0.03[ab]	4.34±1.25[ab]	0.65±0.53[ab]
Control Group	45	Before treatment	48.22±7.43	33.86±3.57	0.32±0.06	21.53±2.47	6.73±1.29
		After treatment	19.39±5.18[a]	18.48±2.57[a]	0.23±0.03[a]	9.36±2.29[a]	2.14±1.23[a]

Table 3: Comparison of inflammatory cytokines between the two groups ($\bar{x}\pm s$).

Note: b means compared with the same group before treatment aP<0.05, and b means compared with the control group after treatment bP<0.05.

Comparison of serum HMGB1 and HSP levels between the two groups

After treatment, serum HMGB1 and HSP levels in both groups were significantly lower than before treatment, while the decrease was more significant in the study group (P<0.01). See Table 4.

Group	n	HMGB1 (ng/mL)		*t*	*P*	HSP (ng/mL)		*t*	*P*
		Before Treatment	After Treatment			Before Treatment	After Treatment		
Research Group	45	9.98±1.22	2.33±0.16	24.064	<0.001	6.35±0.69	2.15±0.22	38.903	<0.001
Control Group	45	9.95±1.18	4.88±0.73	24.511	<0.001	6.39±0.66	3.57±0.52	22.514	<0.001
t		0.119	22.889			0.281	16.879		
P		0.906	<0.001			0.779	<0.001		

Table 4: Comparison of serum HMGB1 and HSP levels between the two groups ($\bar{x}\pm s$).

Comparison of hospital stay, blood amylase recovery time and abdominal pain relief time between the two groups

It was found that the hospital stay, serum amylase recovery time and abdominal pain relief time of the study group were significantly lower than those of the control group (P<0.01), as shown in Table 5.

Group	n	Hospital Stays (d)	Serum Amylase Recovery Time (d)	Abdominal Pain Relief Time (d)
Research Group	45	9.43±1.04	6.27±1.78	4.26±1.08
Control Group	45	11.27±1.42	9.61±1.86	5.45±1.17
t		7.013	8.703	5.014
P		<0.001	<0.001	<0.001

Table 5: Comparison of hospital stay, serum amylase recovery time and abdominal pain relief time between the two groups ($\bar{x}\pm s$).

Discussion

Severe pancreatitis is a common acute and severe disease characterised by multiple organ functions. Moreover, severe pancreatitis is an extremely reactive disease of the pancreas with many complications, poor prognosis and high mortality[8]. Some studies have found that when patients develop severe pancreatitis, various trypsin activities show a significant increase trend that are closely related to the severity of the disease[9]. Systemic inflammatory response mediated by cytokines or inflammatory mediators plays an important role in the occurrence and development of severe pancreatitis. Glutamine is a non-essential amino acid that acts as a conditional amino acid that regulates protein synthesis when the body is stimulated, such as by infection. It is the only energy source of the small intestinal mucosal cells and the most critical nutrient in the process of intestinal mucosal self-repair. When severe pancreatitis occurs, the utilisation rate of glutamine by intestinal epithelial cells and immune cells in patients is significantly increased[10]. In this study, after treatment, the clinical efficacy of the study group was 93.33%, significantly higher than that of the control group (77.78%, P<0.05).

Conclusion: glutamine can significantly improve the therapeutic effect of severe pancreatitis.

Inflammatory cytokines play an important role in systemic inflammation caused by severe acute pancreatitis, and their accumulation and correlation will further lead to vascular leakage, multi-system organ failure and hypovolemic crisis[11]. Studies have found that the occurrence and development of severe pancreatitis is closely related to the level of inflammatory factors[12]. TNF-α is an essential pro-inflammatory factor in the body, which can activate the release of IL-6 and other inflammatory factors, triggering an inflammatory response, leading to apoptosis and necrosis. As an acute phase protein, CRP is mainly produced by liver cells and is an im-

portant inflammatory factor in the body, which is extremely sensitive to inflammation or infection[13]. IL-6 can induce liver cells to synthesize CRP, leading to defence and tissue damage, thus stimulating macrophages to gradually proteinase. IL-8 is a chemokine that mediates the inflammatory damage of IL-6 and TNF-α. IL-1β, mainly produced by activated monocytes/macrophages, and is a lymphocyte stimulating factor that induces the release of IL-6 and TNF-α secretion by neutrophils[14]. The results showed that, compared with before the treatment, the levels of inflammatory factors such as TNF-α, IL-8, IL-1β, IL-6 and CRP in both groups were significantly reduced after treatment, and the above indicators in the study group were significantly lower than those in the control group (P<0.05). The results showed that glutamine had an obvious anti-inflammatory effect. HMGB1, a member of the high mobility group protein superfamily, is a non-histone chromosome binding protein, mainly found in the eukaryotic cells. Studies have found that HMGB1 is characterised by late release, long maintenance peak time, and maintenance and amplification of inflammation[15]. As well, HMGB1 is a late inflammatory factor, which can aggravate the disease of patients by inducing the expression of TNF-α and other factors. It has been found that HSP is closely related to acute pancreatitis. Furthermore, HSP is an important stress protein in the body. When the body is stimulated by high temperature, oxidative stress, inflammation, tumour, etc., HSP level is drastically increased[16]. Jin et al.[17] found in a study of rats that serum HSP level was significantly increased after rats induced severe pancreatitis and glutamine could significantly inhibit the expression or release of serum HSP in patients with severe pancreatitis. In this study, serum HMGB1 and HSP levels in both groups were considerably lower after treatment than before treatment, while the decrease was more significant in the study group (P<0.01). The hospital stay, serum amylase recovery time and abdominal pain relief time of the study group were significantly lower than those of the control group (P<0.01). Conclusion: glutamine can significantly reduce serum HMGB1 and HSP levels, control disease progression and shorten recovery time of patients. This is similar to the research results of Yin et al.[18]. In summary, glutamine is an effective drug for the treatment of severe pancreatitis, which can significantly reduce the inflammatory cascade reaction, reduce serum HMGB1 and HSP levels, and promote the recovery of patients with significant clinical effects.

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Corresponding Author:
Fei Li
Email: qnu4gm@163.com
(China)