

The therapeutic effect of combined therapy of hemopoietin and methylprednisolone in treatment of spinal cord ischemia-reperfusion injury of patient with cervical spondylotic myelopathy and its influence to the level of serum IL-1 β , IL-1Ra and IL-8

Purpose: To study the therapeutic effect of combined therapy of hemopoietin and methylprednisolone in treatment of spinal cord ischemia-reperfusion injury and explore the its influence to the level of IL-1 β , IL-1Ra and IL-8.

Method: 80 patients who had been accepted for cervical spondylotic myelopathy in our hospital during March 2014 to September 2017 were selected as research objects. They were randomly divided into control group and observation group, each containing 40 patients. Both groups of patients were subjected to anterior cervical discectomy and fusion internal fixation. However, the difference was that the control group was subjected to Rapid intravenous injection of 30 mg/kg methylprednisolone 30 min prior to spinal cord decompression, while the observation group was subjected to intravenous injection of both 30mg/kg methylprednisolone and 3000U/kg hemopoietin 30 min before spinal cord decompression. The neurological function of both groups of patients before treatment and 3 months after treatment was evaluated using JOA score (Japanese orthopedic association score) and spinal cord function rating method (40-point rating method). ELISA was carried out to measure the level of S-100 β , neurone specific enolase (NSE), IL-1 β , IL-1Ra, IL-8 of two groups of patients; The life quality of patients in both groups 3 months after treatment was evaluated using WHOQOL-100.

Results: After 3 months of treatment, the JOA score and score of 40-point rating method for observation group was (18.33 \pm 2.71) point and (39.12 \pm 3.97) point, respectively, which were significantly higher than that (P=0.025,0.019) of control group; the level of S-100 β and NSE in observation group was (0.08 \pm 0.02)g/L and (9.51 \pm 0.45)g/L, respectively, which were significantly higher than values (P=0.031,0.022) of control group; the level of IL-1 β and IL-8 in observation group was (21.73 \pm 3.55)ng/L and (356.97 \pm 32.26)ng/L, respectively, both of which were lower than the values (P=0.016,0.018) of control group; while the level of L-1Ra in observation was (21.63 \pm 1.14)ng/L, which was higher than that (P=0.021)of control group; the WHOQOL-100 scores in physiology, psychology, independence, social relation, environment, spirit and overall life quality of observation group were all higher than that of control group, indicating the intergroup difference was of statistical significance (all P<0.001).

Conclusion: The combined therapy of hemopoietin and methylprednisolone achieved significant therapeutic effect in treatment of spinal cord ischemia-reperfusion injury. Moreover, this combined therapy had effective function in reducing S-100 β and NSE, inhibiting IL-1 β , increasing IL-8 and IL-1Ra, and thus realized protection of spinal cord nerve function and improved the life quality of patients. The combined therapy of hemopoietin and methylprednisolone is worth of being promoted in clinics.

Keywords: hemopoietin • methylprednisolone • cervical spondylotic myelopathy • spinal cord ischemia-reperfusion injury • IL 1 β

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Introduction

Cervical spondylotic myelopathy refers to nerve function deficit induced by spinal compression ischemia caused by degenerative pathological change of cervical vertebra, which is a typical clinical symptom [1]. The pathogenesis of cervical spondylotic myelopathy is degenerative pathological change of cervical vertebra. Due to the emergence of soft tissue outside the spinal cord and peripheral bone compression, the spinal cord nerve function is affected and location changing of anatomical structure is caused [2,3]. Cervical spondylotic myelopathy may lead to spinal cord blood supply disorder and venous reflux disorder, cause myelomalacia and myelonecrosis and result in abnormal spinal nerve function [4,5]. Although surgery can effectively relieve spinal cord compression, it may also cause ischemia reperfusion injury in certain degree, thus affecting the recovery of nerve function after decompression [6,7]. Spinal cord ischemia reperfusion injury refers to non-improvement or even aggravation of nerve function after recovery of spinal cord blood supply. In some extremely serious cases, the spinal nerve cloud may suffer irreversible delayed neuronal death [8,9]. Methylprednisolone is an effective drug for treating spinal ischemia reperfusion. However, with the clinical use of methylprednisolone, it is gradually recognized that large dosage of methylprednisolone may cause complications such as gastrointestinal bleeding and osteonecrosis [10]. Hemopoietin determines the oxygenation state of tissues in large degree. The hemopoietin is highly expressed in central nervous system (CNS) cell, which exerts neurotropic effect, anti-neuron apoptosis, anti-oxidation and anti-inflammation functions via approach of autocrine and paracrine [11]. Currently, very few researches have been reported on the therapeutic effect

of combined therapy of hemopoietin and methylprednisolone in treatment of spinal cord ischemia-reperfusion injury of patients with cervical spondylotic myelopathy. To this end, this study analyzes therapeutic effect of combined therapy of hemopoietin and methylprednisolone in treatment of spinal cord ischemia-reperfusion injury of patients with cervical spondylotic myelopathy and its influence to serum IL-1 β , IL-1Ra and IL-8 level, which is expected to provide reference for spinal cord ischemia-reperfusion injury of patients with cervical spondylotic myelopathy.

Data and method

General data

80 patients who had been accepted for cervical spondylotic myelopathy in our hospital during March 2014 to September 2017 were selected as research objects.

Inclusion criteria

Patients who meet the diagnosis standard of cervical spondylotic myelopathy; under 75 years old in age; single section or two adjacent-section degeneration of lumbar intervertebral disc upon MRI test, significant signal indicating vascular myelopathies; no peripheral vascular diseases which affect upper limb function; no heart, liver, kidney and lung diseases; all selected patients enjoyed the right to know and accepted this study.

Exclusion criteria

Patients refuse to join in this test; having mental diseases; equal to or over 75 years old in age; having heart and kidney diseases; affect upper limb function by vascular disease. All 80 patients were randomly divided into control group and observation group, each containing 40. There was no significant difference in gender, age, body weight, height, disease course, site of lesion (single section/two adjacent-section) between two groups ($P > 0.05$), as shown in **TABLE 1**.

Table 1. Comparison of general data between two groups

Groups	n	Gender (male / female)	Age (years)	Body weight (kg)	Height (cm)	Disease course (years)	Site of lesion (single section/two adjacent-section)
Observation group	40	22/18	57.43 \pm 3.29	66.07 \pm 3.72	166.14 \pm 9.27	1.78 \pm 0.31	24/16
Control group	40	21/19	57.35 \pm 2.86	66.11 \pm 3.63	165.08 \pm 8.65	1.75 \pm 0.42	23/17
t/X ²		0.763	0.454	0.771	1.095	0.696	0.993
P		0.121	0.139	0.106	0.328	0.244	0.087

Therapeutic method

Both groups of patients were subjected to anterior cervical discectomy and fusion internal fixation for single-section or two adjacent-section intervertebral disk. The surgery was performed by the same group of surgeon for two groups. 30 min prior to spinal cord decompression, the control group was subjected to rapid intravenous injection of 30 mg/kg methylprednisolone (finished within 15 min), while the observation group was subjected to intravenous injection of both 30 mg/kg methylprednisolone and 3000U/kg hemopoietin (finished within 15 min). After surgery, each patient from both groups was orally administrated with 80 mg of methylprednisolone, once a day, for 2 consecutive weeks.

Observation indexes

Neurological functional status

The neurological function of both groups of patients before treatment and 3 months after treatment was evaluated using JOA score (Japanese orthopaedic association score) and spinal cord function rating method. The larger the score is, the better the neurological function status is.

Biochemical index

ELISA was carried out to measure the level of S-100 β , neurone specific enolase (NSE), IL-1 β , IL-1Ra, IL-8 of two groups of patients before treatment and 3 months after treatment; The ELISA kits were all purchased from Shanghai Runyu Biotechnology Co. Ltd.

Life quality

The life quality of patients in both groups 3 months after treatment was evaluated using WHOQOL-100. WHOQOL-100 includes 6 dimensions related to life quality, i.e. physiology, psychology, independence, social relation, environment and spirit. Furthermore, the WHOQOL-100 includes 24 items and each item includes 4 questions.

Plus 4 additional questions on overall health condition and overall life quality, there is totally 100 questions included in the WHOQOL-100. The higher the score in each dimension is, the better the life equality is.

Statistical treatment

SPSS 22.00 software was adopted for statistical analysis. The measurement data was expressed in the form of mean \pm standard error ($\bar{x} \pm s$) and subjected to independent t-test; enumeration data was subjected to chi-square test, $P < 0.05$ represents the different is of statistical significant.

Results

Comparison of neurological function score between two groups

Before treatment, there is no significant difference in JOA score and score of 40-point rating method between two groups ($P > 0.05$); After 3 months of treatment, the JOA score and score of 40-point rating method for observation group was (18.33 ± 2.71)point and (39.12 ± 3.97) point, respectively, which were significantly higher than that ($P = 0.025, 0.019$) of control group, as shown in **TABLE 2**.

Comparison of S-100 β and NSE level between two groups

Before treatment, there was no significant difference in level of S-100 β and NSE between two groups ($P = 0.552, 0.031$); after 3 months of treatment, the level of S-100 β and NSE in observation group was (0.08 ± 0.02) g/L and (9.51 ± 0.45)g/L, respectively, which were significantly higher than values ($P = 0.031, 0.022$) of control group, as shown in **TABLE 3**.

Comparison of level of IL-1 β , IL-1Ra and IL-8 between two groups

Before treatment, there was no significant difference in level of IL-1 β , IL-1Ra and IL-8 between two groups ($P = 0.252, 0.377, 0.144$); after 3 months of treatment, the level of IL-1 β and IL-8 in observation group was

Table 2. Comparison of neurological function score between two groups (Score, $\bar{x} \pm s$)

Groups	n	JOA score		40-point rating method	
		Before treatment	After 3 months of treatment	Before treatment	After 3 months of treatment
Observation group	40	10.15 \pm 1.62	18.33 \pm 2.71	30.94 \pm 3.25	39.12 \pm 3.97
Control group	40	10.21 \pm 1.95	14.96 \pm 2.83	30.87 \pm 3.01	35.56 \pm 4.03
t		0.194	5.327	0.393	6.008
P		0.428	0.025	0.316	0.019

(21.73±3.55) ng/L and (356.97±32.26) ng/L, respectively, both of which were lower than the values (P=0.016,0.018)of control group; while the level of L-1Ra in observation was (21.63±1.14) ng/L, which was higher than that (P=0.021)of control group, as shown in **TABLE 4**.

Comparison of life quality between two groups

After 3 months of treatment, the WHOQOL-100 scores in physiology, psychology, independence, social relation, environment, spirit and overall life quality of observation group were all higher than that of control group, indicating the intergroup difference was of statistical significance (all P<0.001), as shown in **TABLE 5**.

Summary

After 3 months of treatment, the JOA score and score of 40-point rating method for observation group was (18.33 ± 2.71) point and (39.12 ± 3.97) point, respectively, which were significantly higher than that (P=0.025, 0.019) of control group; the level of S-100β and NSE in observation group was (0.08 ± 0.02)g/L and (9.51 ± 0.45)g/L, respectively, which were significantly higher than values

(P=0.031, 0.022) of control group; the level of IL-1β and IL-8 in observation group was (21.73 ± 3.55)ng/L and (356.97 ± 32.26) ng/L, respectively, both of which were lower than the values (P=0.016, 0.018) of control group; while the level of L-1Ra in observation was(21.63 ± 1.14) ng/L, which was higher than that (P=0.021) of control group; the WHOQOL-100 scores in physiology, psychology, independence, social relation, environment, spirit and overall life quality of observation group were all higher than that of control group, indicating the intergroup difference was of statistical significance (all P<0.001).

Discussion

Cervical spondylotic myelopathy is a clinically common disease. Operative decompression has long been regarded as a dominant approach to treat such disease by relieving the compression applied on spinal cord and maintaining the stability of spine. However, since the compressed spine has already shown ischemic anoxic changes, a sudden increase of blood supply after operative decompression may cause ischemia reperfusion injury [12,13]. At present, there has been no specific

Table 3. Comparison of S-100β and NSE level between two groups (g/L, $\bar{x} \pm s$)

Groups	n	S-100β		NSE	
		Before treatment	After 3 months of treatment	Before treatment	After 3 months of treatment
Observation group	40	0.19 ± 0.12	0.08 ± 0.02	15.62 ± 1.33	9.51 ± 0.45
Control group	40	0.18 ± 0.16	0.12 ± 0.04	15.54 ± 1.47	12.03 ± 0.86
t		0.196	5.082	0.304	6.017
P		0.552	0.031	0.411	0.022

Table 4. Comparison of level of IL-1β, IL-1Ra and IL-8 between two groups (ng/L, $\bar{x} \pm s$)

Groups	n	IL-1β		IL-1Ra		IL-8	
		Before treatment	After 3 months of treatment	Before treatment	After 3 months of treatment	Before treatment	After 3 months of treatment
Observation group	40	45.25 ± 4.16	21.73 ± 3.55	16.17 ± 2.02	21.63 ± 1.14	843.66 ± 57.82	356.97 ± 32.26
Control group	40	45.08 ± 3.74	30.94 ± 2.18	16.04 ± 1.97	19.49 ± 1.05	841.25 ± 60.17	417.78 ± 41.33
t		0.634	8.044	0.122	6.705	0.698	6.916
P		0.252	0.016	0.377	0.021	0.144	0.018

Table 5. Comparison of life quality between two groups after 3 months of treatment (Score, $\bar{x} \pm s$)

Groups	n	Physiology	Psychology	Social relation	Independence	Environment	Spirit	Overall life quality
Observation group	40	73.26 ± 7.85	81.74 ± 11.81	77.13 ± 8.54	87.19 ± 12.55	83.11 ± 8.72	83.41 ± 10.78	68.09 ± 7.52
Control group	40	67.34 ± 9.03	81.15 ± 10.96	69.02 ± 6.35	79.14 ± 11.28	71.65 ± 9.03	75.36 ± 7.61	59.43 ± 6.41
t		10.635	12.146	13.893	17.095	19.414	18.006	17.172
p		<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

therapy for spinal cord ischemia-reperfusion injury. Methylprednisolone is a synthetic glucocorticoid, which has therapeutic effect to spinal cord ischemia-reperfusion injury owe to its function of inhibiting lipid peroxidase and inflammatory response after trauma. This drug has strong anti-inflammatory action and is regarded as a standard therapy for acute spinal cord injury. Relevant literatures show [14] a large dosage of methylprednisolone may cause various untoward effects, which is unbeneficial for the improvement of neurological function. Researches show that [15] hemopoietin and its receptor are widely expressed in organs and tissues of human body, which have potential cytoprotection. Hemopoietin has neurotrophic activity, which can improve the brain tissue's tolerance to ischemia-hypoxia and exerts neuroprotective effect via anti-cell apoptosis, anti-oxidation, anti-inflammation. This study showed that after 3 months of treatment the JOA scores and scores of 40-point rating method in observation group were both significant higher than that control group ($P=0.025,0.019$), indicating the combined therapy of hemopoietin and methylprednisolone can well promote the recovery of neurological function of patients with cervical spondylotic myelopathy.

Some researches show that [16] hemopoietin has similar function with platelet-derived growth factor (PDGF), both of which can improve the activity of antioxidant enzymes such as glutathion peroxidase; in addition, hemopoietin can inhibit the increase of thiobarbituric acid after ischemia-reperfusion injury, realizing the function of anti-brain lipid peroxidation. Serum neuron and astrocyte specific protein can effectively reflect the disease status of traumatic brain injury, cerebral infarction and spinal cord injury. The S-100 β contained in hemopoietin is a type of Ca-binding protein, which has certain correlation with some calcium ions exerting their own function by relying on cells. When the contents of S-100 β and cerebrospinal fluid (CSF) are increased, the damage level of nervous centralis is proportion to the contents of S-100 β and CSF [17]. NSE, as a potential marker in diagnosis of cerebral spinal cord injury, has attracted increasing attention. Previous researches [18] have proved that the increase of peripheral blood NSE level is positively correlated with the damage level of nervous centralis, while negatively

related with prognosis. Some researches [19] show that serum NSE level can be regarded as a specific marker for acute stage of spinal cord injury. This research showed that after 3 months of treatment the level of S-100 β and NSE in observation group was significantly higher than that of control group ($P=0.031, 0.022$), indicating the combined therapy of hemopoietin and methylprednisolone can effectively reduce the level of S-100 β and NSE in serum, and thus relieving the spinal cord ischemia-reperfusion injury.

Research [20] shows that IL-1 β is involved in the early development course of spinal cord ischemia reperfusion injury. The increase of expression of IL-1 β after reperfusion injury is a main molecular basis accounting for blood spinal cord barrier lesion. Research shows [21] that IL-1Ra, IL-1R's antagonist, can competitively combine with IL-1 β receptor to block biological effects of IL-1, thus defending the pathological damage caused by IL-1 β and relieving ischemic injury. Positive expression or generation of IL-8 will not occur in normal spinal cord tissue, but only occur in the process of ischemia reperfusion injury (specifically, around endothelial cells), indicating IL-8 is mainly derived from endothelial cells. Hirose K et al. [22] studied the role of antithrombin in reducing ischemia reperfusion-induced inflammation of rats and relieving spinal cord injury and found that antithrombin could significantly inhibit the expression of IL-8. This research showed that after 3 months of treatment the level of IL-1 β and IL-8 in observation group was lower than that in control group ($P=0.016,0.018$); while the level of IL-1Ra in observation group was higher than that in control group ($P=0.021$). This indicates the combined therapy of hemopoietin and methylprednisolone has advantages in inhibiting IL-1 β and IL-8 and promoting IL-1Ra, and thus achieves better protection of spinal cord nerve function. Furthermore, this research shows that the combined therapy of hemopoietin and methylprednisolone can improve life quality of patients more significantly compared with single application of methylprednisolone.

Conclusion

The combined therapy of hemopoietin and methylprednisolone achieved significant therapeutic effect in treatment of spinal cord ischemia-reperfusion injury. Moreover, this combined therapy had effective function in

reducing S-100 β and NSE, inhibiting IL-1 β , increasing IL-8 and IL-1Ra, and thus realized protection of spinal cord nerve function and improved the life quality of patients. The combined therapy of hemopoietin and methylprednisolone is worth of being promoted in clinics.

References

- Park MS, Ju YS, Moon SH *et al.* Reoperation rates after anterior cervical discectomy and fusion for cervical spondylotic radiculopathy and myelopathy: a national population-based study. *J. Spine.* 41(20), 1593-1599 (2016).
- Abodeiyamah KO, Stoner KE, Grossbach AJ *et al.* Effects of brain derived neurotrophic factor Val66Met polymorphism in patients with cervical spondylotic myelopathy. *J. Clin. Neurosci.* 24, 117-121 (2016).
- Levine DN. Pathogenesis of cervical spondylotic myelopathy. *J. Neurol. Neurosurg. Psych.* 62(4), 334-340 (1997).
- Talekar K, Poplawski M, Hegde R *et al.* Imaging of spinal cord injury: acute cervical spinal cord injury, cervical spondylotic myelopathy, and cord herniation. *Seminars. Ultrasound. CT. MRI.* 37(5), 431-447 (2016).
- Miura J, Doita MK, Marui T *et al.* Dynamic evaluation of the spinal cord in patients with cervical spondylotic myelopathy using a kinematic magnetic resonance imaging technique. *J. Spinal. Disorders. Tech.* 22(1), 8-13 (2009).
- Nardone R, Höller Y, Brigo F *et al.* The contribution of neurophysiology in the diagnosis and management of cervical spondylotic myelopathy: a review. *Spinal. Cord.* 54(10), 756 (2016).
- Zuozhang Y, Lin X, Hongpu S *et al.* A patient with lung cancer metastatic to the fifth thoracic vertebra and spinal cord compression treated with percutaneous vertebroplasty and I-125 seed implantation. *Diag. Interv. Rad.* 17(4), 384-387 (2011).
- Bell MT, Smith PD, Jr J CC *et al.* Attenuation of spinal cord ischemia-reperfusion injury with treatment of alpha-2 agonist dexmedetomidine. *J. Vasc. Surg.* 54(6), 1862-1862 (2011).
- Wang Y, Su R, Lv G *et al.* Supplement zinc as an effective treatment for spinal cord ischemia/reperfusion injury in rats. *Brain. Res.* 1545(4), 45-53 (2014).
- Boyaci MG, Eser O, Kocogullari CU *et al.* Neuroprotective effect of alpha-lipoic acid and methylprednisolone on the spinal cord ischemia/reperfusion injury in rabbits. *Br. J. Neurosurg.* 29(1), 1-6 (2014).
- Diller GP, Lammers AE, Haworth SG *et al.* A modelling study of atrial septostomy for pulmonary arterial hypertension, and its effect on the state of tissue oxygenation and systemic blood flow. *Cardiol. Young.* 20(1), 25-32 (2010).
- Hamamoto Y, Ogata T, Morino T *et al.* Real-time direct measurement of spinal cord blood flow at the site of compression: relationship between blood flow recovery and motor deficiency in spinal cord injury. *Spine.* 32(18), 1955-1962 (2007).
- Chang HS, Nejo T, Yoshida S *et al.* Increased flow signal in compressed segments of the spinal cord in patients with cervical spondylotic myelopathy. *Spine.* 39(26), 2136-2142 (2014).
- Jongen PJ, Stavrakaki I, Voet B *et al.* Patient-reported adverse effects of high-dose intravenous methylprednisolone treatment: a prospective web-based multi-center study in multiple sclerosis patients with a relapse. *J. Neurol.* 263(8), 1641-1651 (2016).
- Iacobucci I, Li Y, Roberts KG *et al.* Truncating erythropoietin receptor rearrangements in acute lymphoblastic leukemia. *Cancer. Cell.* 29(2), 186-200 (2016).
- Yin X, Xu J, Shi J *et al.* Immunohistochemical detection of erythropoietin, platelet-derived growth factor and their receptors in ameloblastomas. *J. Hard. Tiss. Biol.* 18(1), 19-26 (2009).
- Albuerne M, Mammola CL, Naves FJ *et al.* Immunohistochemical localization of S100 proteins in dorsal root, sympathetic and enteric ganglia of several mammalian species, including man. *J. Peripheral. Nervous. Sys. Jpn.* 3(4), 243-253 (1998).
- Chai QL, Pediatrics DO. Correlation of serum NSE and S100 β levels with inflammatory response and immune response in children with hand-foot-mouth disease complicated by encephalitis. *J. Hainan. Med. Univ.* 23(13), 90-93 (2017).
- Cao F, Yang XF, Liu WG *et al.* Elevation of neuron-specific enolase and S-100 β protein level in experimental acute spinal cord injury. *J. Clin. Neurosci.* 15(5), 541-544 (2008).
- Yamamoto S, Tanabe M, Wakabayashi G *et al.* The role of tumor necrosis factor- α and interleukin-1 β in ischemia-reperfusion injury of the rat small intestine. *J. Surg. Res.* 99(1), 134-141 (2001).
- Merhisoussi F, Berti M, Wehrlehaller B *et al.* Intracellular interleukin-1 receptor antagonist type 1 antagonizes the stimulatory effect of interleukin-1 alpha precursor on cell motility. *Cytokine.* 32(3-4), 163-170 (2005).
- Hirose K, Okajima K, Uchiba M *et al.* Antithrombin reduces the ischemia/reperfusion-induced spinal cord injury in rats by attenuating inflammatory responses. *Thromb. Haemost.* 91(1), 162-170 (2004).