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Polysaccharide from *Spirulina platensis* improves sepsis-induced acute lung injury by alleviating inflammatory response and down-regulating endocan expression in rats

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Abstract

To explore the favourable functions of polysaccharide from *Spirulina platensis* (PSP) on sepsis-originated acute lung injury, together with the corresponding mechanisms. 60 male SD rats were randomly separated into sham, sepsis, and sepsis+PSP group with 20 rats in each group. Sepsis rat model was constructed by cecal ligation and puncture (CLP). The mortality and sepsis-related symptoms were used to judge the model success. CLP successfully established the sepsis-caused acute lung injury rat model. In sepsis rats, systemic inflammation and pulmonary inflammation, as well as endocan protein level were increased. PSP treatment improved sepsis-induced acute lung injury and systemic inflammation, and reduced the endocan protein level. Lung injury biomarkers were positively correlated with serum pro-inflammatory factors/serum endocan protein, while serum pro-inflammatory factors were positively associated with serum endocan level. PSP ameliorates acute lung injury via relieving inflammatory response and down-regulating endocan expression in sepsis rat.

Keywords: acute lung injury; polysaccharide; spirulina platensis; inflammatory response; endocan

Practical Application: Polysaccharide from *Spirulina platensis* (PSP) ameliorates acute lung injury via relieving inflammatory response and down-regulating endocan expression in sepsis rat, suggesting that PSP might be used for the treatment of inflammation related lung injury.

1 Introduction

Sepsis, as a common critical illness, is a systemic inflammatory reaction syndrome triggered by various factors, which can result in systemic and multi-organ damages, and is the main fatal factor of patients in critical ward (Carvalho et al., 2013). Among them, lung is often the most susceptible organ, and acute lung injury usually can be caused by sepsis in the early stage, which is progressing rapidly with no specific treatment method, and the mortality is high (Miyashita et al., 2016). Therefore, how to effectively prevent and treat acute lung injury caused by sepsis has become the focus of clinical research.

It is found that excessive inflammatory response takes a critical effect on the occurrence and progression of sepsis-induced acute lung injury. Moreover, the dysfunction of endothelial cells and exorbitant release of pro-inflammatory cytokines are the keys of pulmonary alveolar capillary membrane injury while pulmonary alveolar capillary membrane injury is a representative feature of lung damage. As a larvaceous marker of endothelial cells, endocan exerts a significant role in a crowd of endothelial relevant pathophysiological illnesses (Fujishima et al., 2016). A study showed that intraperitoneal injection of endocan can effectively inhibit the level of proinflammatory factors and apoptosis of pulmonary epithelial cells in endotoxin-induced acute lung injury model in mice, thus reducing endotoxin-caused acute lung injury (Zhang et al., 2018). endocan, also named endothelial cell-specific molecule-1(ESM-1), is specifically secreted from pulmonary vascular endothelial cells, mainly expressed in alveolar capillary endothelial cells, pulmonary venules and arterioles, and bronchial and submucosal glandular epithelial cells under physiological conditions. It is a circulating protein polysaccharide, one of the biomarkers associated with vascular endothelial injury and a new marker of dysfunction in endothelial cells. Generally, the expression level of serum endocan in healthy crowds is very low, but it is significantly higher in sepsis patients and closely related to the severity, thus, it is used as a diagnostic and prognostic indicator of sepsis (Pierrakos & Vincent, 2010; Scherpereel et al., 2006). As a result, inflammatory response and endocan expression changes exert a central effect in acute lung injury caused by sepsis.

Polysaccharide from Spirulina platensis (PSP) is an active substance extracted from Spirulina (a natural monocyte plant). In recent years, the pharmacological effects of PSP have been widely concerned by researchers, making it become a hot spot in the research and development of marine drugs at home and abroad. Studies have shown that PSP is an acid heteropolysaccharide, basically non-toxic, these bioactive substances have diversiform special physiological roles on the organism, can prevent and

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cure diversiform diseases, and also has a significant dual role in enhancing the body's immunity. More and more attention has been paid to the research and development of the pharmacological actions of PSP, including anti-tumor, anti-various cardiovascular diseases, anti-inflammatory, anti-peptic ulcer, anti-coagulation, anti-oxidation, anti-fatigue, anti-radiation, anti-thrombotic, anti-toxic damage, anti-edema, hypoglycemic, anti-aging, prolonging life span, enhancing the body immunity and promoting DNA synthesis in recent years (Nishino et al., 1991; Schwartz et al., 1976). However, there is no study about the treatment of PSP in sepsis and lung injury. Thereby, in this investigation, we will explore whether PSP plays a beneficial effect in acute lung injury triggered by sepsis and whether it regulates inflammatory response and endocan expression.

2 Materials and methods

2.1 Animals and groups

60 clean and healthy SD male rats (rat age: 3~4 months, body mass: 300 ± 50 g) were purchased from Shanghai Shrek Experimental Animal Co., Ltd and would be randomly split into sham, sepsis, and sepsis+PSP group, and each group included 20 rats. The rats were treated according to the ethical guidelines of the animal center of Shanghai Shrek Experimental Animal Co., Ltd.

2.2 Sepsis model and drug administration

Sepsis model in rats was established by adopting CLP method as previously depicted (Dejager et al., 2011; Hubbard et al., 2005). All rats were subjected to preoperative fasting without water for 12 h, then, they were anesthetized by i.m. 60 mg/kg of ketamine (Ketaset,Zoetis) and 10 mg/kg of xylazine (AnaSed, Akorn, Lake Forest, IL) followed by conventional iodophor disinfection on abdomens, skin preparation and spreading the sterile gauze hole towel, and about 1 cm of abdominal wall skin was carved open along the abdominal middle line of the lower abdominal part of the cartilago ensiformis. For the rats in sepsis group and sepsis+PSP group, when the caecum was ascertained by sterile forceps, most of the caecum which distance from the place of 1/2 of the ileocecal part was ligated by surgical suture. 20 G of sterile needles were used for perforating at the head and tail end of the ligated caecum, then the caecum was squeezed, and the content was squeezed out about 0.3 mL along the perforation site. The caecum and the extruded contents were sent back to the abdominal cavity and sutured the abdominal wall incision layer by layer. Following CLP, the rats in the sepsis+PSP group were performed a intragastric administration of 80 $mg.kg^{-1}$ of PSP while the rats in the sham and sepsis groups were given equal volume normal saline for intragastric administration. After CLP operations, the animals were performed a subcutaneous resuscitation by injection of 3 mL/100 g of normal saline.

2.3 General observation

After CLP for 24 h, the mortality rates were counted, and sepsis-related symptoms were observed and recorded at 6 h after CLP. The sepsis-related symptoms included mental state, postoperative activity, vertical hair, eyelid secretion, anal stool adhesion condition, and urine turbidity condition. Moreover, the abdominal cavity turbid exudate, intestinal adhesion and cecal necrosis were also been observed after the animals were sacrificed to break the abdominal cavity.

2.4 Sample collection

After CLP for 24 h, the animals were anesthetized via intraperitoneally injection of 100 mg/kg Ketamine. Then, 5 mL of blood was collected through abdominal aorta and centrifuged by centrifuge to collect supernatant to obtain serum, and the subnatant to obtain plasma. Meanwhile, the lung tissues and bronchoalveolar lavage fluids were acquired. For collection of bronchoalveolar lavage samples, the rats were ligated at right lobe and inserted a puncture needle at the upper end of the trachea followed by 3 times of rinse by 0.3 mL of normal saline, and each rinse was preformed 3~5 times. The irrigating solution was retrieved and centrifuged at 1500 g for 10 min at 4 °C, then, the supernatant was absorbed into new EP tube and stored at -20 °C. The serum and plasma samples, and lung tissues were stored at -80 °C.

2.5 W/D ratio of lung tissues

The middle lobes of right lung tissues were resected. After the surface liquid was dried by filter paper, the W weight was weighed on the electronic balance and then placed in the oven at 65°C. After 72 h of drying, the D weight was weighed again, and the W/D ratio of lung was measured.

2.6 Determination of pulmonary myeloperoxidase (MPO) activity

The upper lobes of right lung tissues were excised, and the activity of MPO was determined by utilizing colorimetry as previously described (Harkin et al., 2004).

2.7 Pulmonary histopathological estimate

The lower lobe of right lung was abscised and fixed for 24 h in 4% polyformaldehyde solution for preparing paraffin blocks. After sectioning, hematoxylin and eosin (HE) staining was conducted for pathological examination and lung morphologic alterations were observed with 40× light microscopy. The pathological score of lung damage was performed according to a previously scoring system described in Table 1 (Kiyonari et al., 2000). The higher the score, the worse the lung injury.

Table 1. The degree of pathological lung injury scoring system.

Types of lesion	Values of different lesion ranges				
	0	0~24%	25%~49%	50%~74%	75%~100%
Alveolar congestion or bleeding	0	1	2	3	4
Inflammatory cells infiltration	0	1	2	3	4
Alveolar structural changes	0	1	2	3	4
Edema	0	1	2	3	4

2.8 Enzyme-linked immunosorbent assay (ELISA)

The concentrations of the lung injury biomarkers of SP-D, CC16 and sRAGEs in plasma (Engels et al., 2016), the systemic inflammation biomarkers of TNF-α, IL-6 and IL-8 in serum and bronchoalveolar lavage specimens, and endocan in serum and bronchoalveolar lavage samples were measured by ELISA referring to the manufacturer's guides of Kits (Shanghai Enzymatic Company, China). Proteins concentrations were expressed as ng/mL in liquid.

2.9 qRT-PCR

The proinflammatory factors mRNA levels in left lung tissues of rats were estimated by qRT-PCR. Resumptively total RNA was extracted by using Trizol reagent (Invitrogen, USA) under the manufacturer's directions. After cDNAs were created through applying high-capacity cDNA Reverse Transcription Kit (Applied Biosystems, USA), cDNAs were amplified utilizing the following primers sequences: TNF-α: forward, 5'-TCAAAGGGGAACGGACATAG-3', reverse, 5'-ACCAGGATTCTGTGGCAATC-3'; IL-6: forward, 5'-GAAATCGTGGAAATGAG-3', reverse, 5'-TAGGTTTGCCGAGTAGA-3'; IL-8: forward, 5'-AAGATTGTCCAAAAGATGCTAA-3', reverse, 5'-ATCGGTGCAATCTATCTTCTTT-3'. GAPDH was adopted as an internal control, and the primers sequences of GAPDH were: forward, 5'-GCCTTCCGTGTTCCTACC-3', reverse, 5'-AGAGTGGGAGTTGCTGTTG-3'. The relative quantification was measured by the ΔΔ CT way. The PCR assays were performed for three times.

2.10 Spearman correlation coefficient analysis

Spearman correlation coefficient method was used for analysis of the relations between the lung injury biomarkers, inflammatory biomarkers, and endocan protein which involved in both sepsis and PSP treatment.

2.11 Statistical analysis

SPSS 16.0 software was adopted for statistical differential analysis between groups. The outcomes were exhibited as means ± SD and rate (%). For comparisons of quantitative data between groups, one-way ANOVA was performed. Comparisons of percentage/rate data between groups was carried out utilizing the X^2 test, non-parametric Mann–Whitney U test, and Fisher's exact test. For relationships analysises, the linear regression equation was conducted. $P < 0.05$ was deemed as a statistical difference.

3 Results

3.1 PSP treatment reduced the death rate and alleviated the sepsis-related symptoms

At 24h after CLP, the mortality rate was recorded while the sepsis-related symptoms were observed at 6 h after CLP. The outcomes displayed that there was no death in the sham group with no abnormal performance. In sepsis group, 10 rats were died with 50% of mortality rate, and at 6 h after CLP surgery, the rats emerged mental depression, shiver, hair erection, slow action, reduced activity, increased eyelid secretion, stool adhesion at anus, turbid urine. After breaking the abdomen, the abdominal cavity could be seen turbid exudate, intestinal sticky, and partly caecum necrosis, which satisfy the successful judgment indicators of sepsis model. After PSP treatment, the above-mentioned discomfort triggered by sepsis was significantly alleviated, at 24 h after CLP, 4 rats were died (20% of mortality), and compared with the sepsis group, the mortality showed statistically different ($P < 0.01$).

3.2 PSP treatment mitigated sepsis-induced lung injury

Lung injuries statuses were reflected by W/D ratio of lung, pulmonary MPO activity, the pathological score of lung injury and the lung injury biomarkers of SP-D, CC16 and sRAGEs. The outcomes displayed that compared with the sham rats, the W/D ratio of lung of sepsis rats was significantly increased ($P < 0.01$), which indicated that sepsis caused pulmonary edema. After PSP treatment, the W/D ratio of lung was remarkably reduced compared with the sepsis rats ($P < 0.01$) but higher than in sham group with no statistical difference ($P > 0.05$) (Figure 1A). Compared with the sham group, the lung MPO was increased in sepsis ($P < 0.001$) and sepsis+PSP group ($P < 0.05$),, indicating that sepsis caused pulmonary neutrophil infiltration, and compared to the sepsis rats, the lung MPO was remarkably decreased in sepsis+PSP rats ($P < 0.01$) (Figure 1B). Analogously, compared to the sham rats, lung pathological score was remarkably increased in sepsis group and sepsis+PSP rats ($P < 0.001$), and compared to the sepsis rats, the lung MPO was increased in sepsis+PSP group ($P < 0.05$) (Figure 1C). The find of lung injury biomarkers protein levels exhibited that compared with the sham group, the concentrations of SP-D, CC16 and sRAGEs in plasma were increased in sepsis group ($P < 0.01$), and compared to the sepsis rats, plasma CC16 ($P < 0.05$) and sRAGEs ($P < 0.01$) concentrations were decreased in sepsis+PSP group (Figure 1D).

3.3 PSP treatment alleviated sepsis-induced systemic and pulmonary inflammatory reaction

To explore the effetcs of PSP on inflammatory response implicated in sepsis induced acute lung injury, the expression levels of TNF-α, IL-6, and IL-8 protein in bronchoalveolar lavage fluid and serum were detected by ELISA while the mRNA levels of them in lung tissues were measured with qRT-PCR. The results showed that sepsis triggered abormal increases of TNF-α, IL-6, and IL-8 protein levels in bronchoalveolar lavage fluid and serum samples while they were significantly reversed after PSP treatment (Figure 2A, 2B). Similar to former analysis, in lung tissues, the mRNA expression levels of TNF-α, IL-6, and IL-8 were all observably upregulated in sepsis group compared with in sham group, however, they were remarkably reduced with PSP treatment (Figure 2C).

3.4 PSP treatment inhibited the sepsis-induced increase of systemic and pulmonary endocan expression

ELISA was conducted to estimate the protein expression levels of endocan in bronchoalveolar lavage fluid and serum. The results displayed that the expression of endocan protein in bronchoalveolar lavage fluid in sepsis rats was observably higher than in sham rats ($P < 0.01$), and in sepsis+PSP rats was higher than in sham rats ($P < 0.05$) (Figure 3A). Compared with the sham rats, the serum endocan protein in sepsis rats was markedly ascending ($P < 0.01$), and the serum endocan protein was declined in the sepsis rats who received PSP treatment ($P < 0.05$) (Figure 3B).

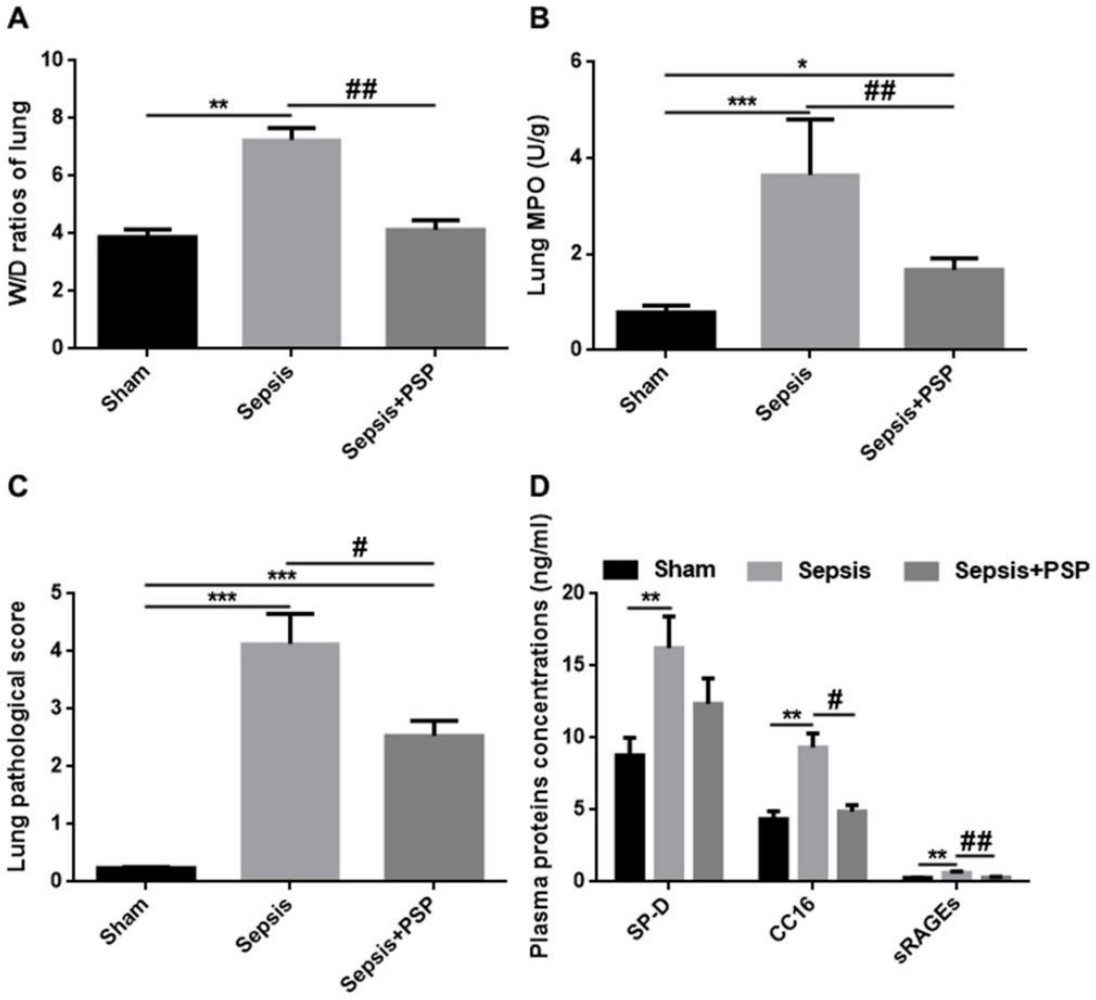

Figure 1. PSP treatment mitigated sepsis-triggered lung injury at 24 h after CLP. (A) W/D weight ratio of lung tissues; (B) Pulmonary myeloperoxidase (MPO) activity determined by using colorimetry; (C) The pathological score of lung injury was evaluated by previously scoring system after pulmonary histopathological examination; (D) The concentrations of the lung damage signs of SP-D, CC16, and sRAGEs in plasma detected by ELISA. W, Wet; D, dry; MPO, myeloperoxidase; SP-D, surfactant protein D; CC16, Clara cell 16 kD protein; sRAGEs, soluble receptor for advanced glycation endproducts. *P < 0.05; **P < 0.01; ***P < 0.001 vs sham group ; #P < 0.05; ##P < 0.01 vs sepsis group, .

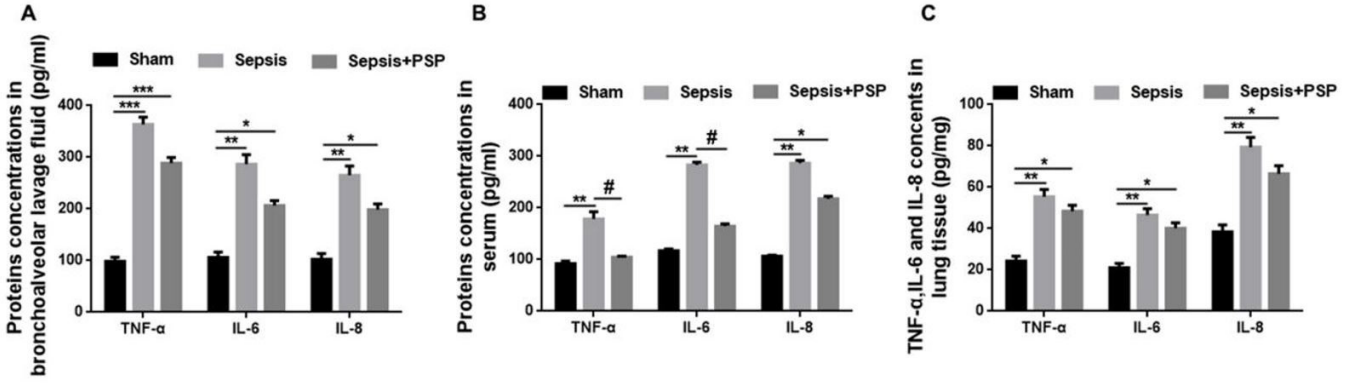

Figure 2. PSP treatment alleviated sepsis-induced systemic and pulmonary inflammatory reaction at 24h after CLP. (A, B) The protein levels of pro-inflammatory factors of TNF-α, IL-6, and IL-8 in bronchoalveolar lavage fluid and serum by ELISA; (C) The mRNA levels of TNF-α, IL-6, and IL-8 in lung tissues detected by qRT-PCR. TNF-α, tumor necrosis factor-α; IL-6, interleukin-6; IL-8, interleukin-8. *P < 0.05; **P < 0.01; ***P < 0.001 vs sham group; #P < 0.05 vs sepsis group.

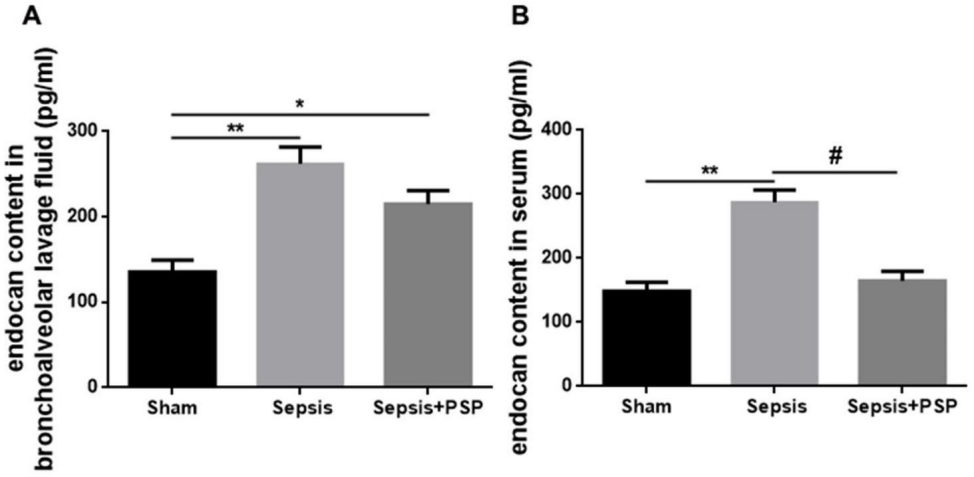

Figure 3. PSP treatment inhibited the sepsis-induced increase of systemic and pulmonary endocan expression at 24h after CLP. (A, B) The protein levels of endocan in bronchoalveolar lavage fluid and serum by ELISA. *P < 0.05; **P < 0.01 vs sham group; #P < 0.05 vs sepsis group.

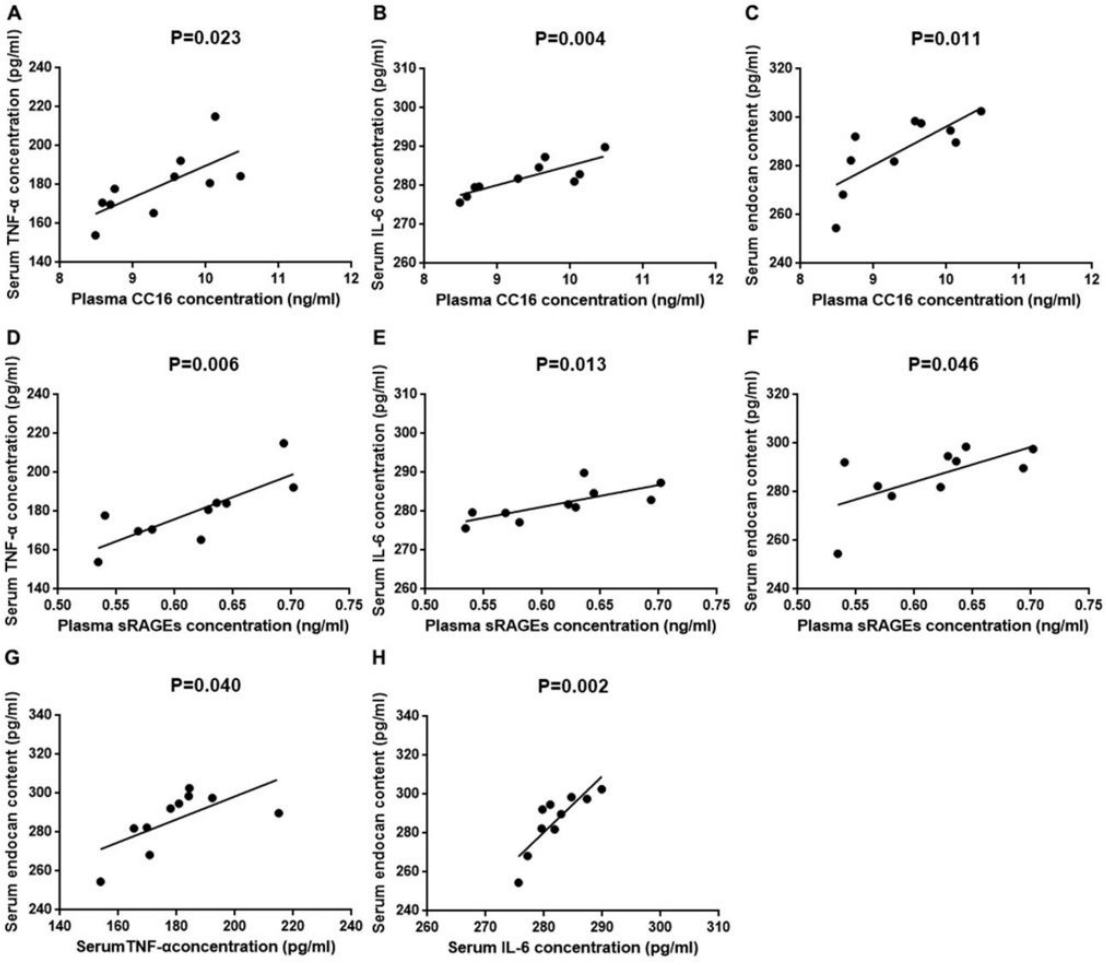

Figure 4. The relations between plasma lung injury biomarkers, serum pro-inflammatory factors, and serum endocan protein level at 24 h after CLP. (A-C) The relationships between the plasma CC16 and serum TNF-α/IL-6/endocan which produced changes/played roles in both sepsis and PSP treatment from sepsis group by Spearman correlation coefficient method; (D-F) The relationships between the plasma sRAGEs and serum TNF-α/IL-6/endocan which produced changes/played roles in both sepsis and PSP treatment from sepsis group by Spearman correlation coefficient method; (G, H) The relationships between the serum endocan and serum TNF-α/IL-6 which produced changes/played roles in both sepsis and PSP treatment from sepsis group by Spearman correlation coefficient method. CC16, Clara cell 16 kD protein; sRAGEs, soluble receptor for advanced glycation endproducts; TNF-α, tumor necrosis factor-α; IL-6, interleukin-6.

3.5 Plasma lung injury biomarkers were positively correlated with serum proteins levels of pro-inflammatory factors/ serum endocan protein, while serum proteins levels of pro-inflammatory factors were positively associated with serum endocan protein level

The lung injury biomarkers of CC16 and sRAGEs, pro-inflammatory markers TNF-α and IL-6, and endocan protein which produced changes/played roles in both sepsis and PSP treatment from sepsis group were performed correlation analysis by using Spearman correlation coefficient method. The outcomes showed that there were positive correlations between plasma CC16/sRAGEs concentration and serum TNF-α/IL-6/endocan concentration, between serum TNF-α/IL-6 concentration and serum endocan content ($P < 0.05$ or $P < 0.01$) (Figure 4).

4 Discussion

Our main discoveries were that PSP treatment could effectively reduce mortality, relieved sepsis-related symptoms, improve sepsis-caused acute lung injury, systemic and pulmonary inflammatory response and inhibit sepsis-induced increased endocan expression in sepsis rats. Moreover, we found that the release of proinflammatory factors induced by sepsis was positively associated with the level of biomarker proteins of lung injury in plasma and the change of serum endocan expression, and the level of biomarker proteins of lung injury in plasma was positively related to serum endocan expression.

Sepsis as a common serious complication of burn, trauma, surgery and ischemia-reperfusion injury, is an important factor leading to death (Chen et al., 2017b). It has been proved that sepsis-triggered uncontrollable systemic inflammation and ultimately multiple organ damnification are the main causes of early death (Liu et al., 2016). Lung is the most vulnerable critical organ in sepsis. Although various diagnosis and treatment techniques have been improved in recent years, acute lung injury caused by sepsis is still the chief causation of death (Ho et al., 2015), which is a difficult problem that critical medicine needs to solve at present. In our study, we adopted a classic CLP method according to previous literature (Dejager et al., 2011; Hubbard et al., 2005) to construct sepsis-caused acute lung injury model in rats, and we found at 24 h after CLP, the mortality of sepsis rats was arrived to 50%, and sepsis-relevant symptoms such as mental depression, slow action, reduced activity, etc., at 6 h after CLP surgery, which indicated the CLP-induced sepsis rats model was successfully built. Moreover, at 24 h after CLP, pulmonary edema (reflected by increased W/D ratio of lung (Joyce et al., 2001)), pulmonary neutrophil infiltration (reflected by increased lung MPO (Gong et al., 2012)), remarkably increased lung pathological score, and increased lung injury biomarkers protein levels of plasma SP-D, CC16, and sRAGEs (Engels et al., 2016) were appeared in the sepsis rats, which indicated that acute lung injury rats model caused by CLP/sepsis was successfully constructed. Besides, at 24 h after CLP surgery, the proinflammatory factors of TNF-α, IL-6, and IL-8 protein levels from bronchoalveolar lavage fluid and serum, as well as their mRNA levels in lung tissues were markedly increased in sepsis rats, which satisfied the most common characteristics of sepsis. Meanwhile, sepsis led to a increase of endocan expression in serum and bronchoalveolar lavage fluid, which clarified that endocan may involved in sepsis. The previous studies found endocan was significantly increased in the serum of sepsis patients (Scherpereel et al., 2006), which was consistent with our findings.

It has been known that Spirulina comprises functional compounds, for instance, phenolics, phycocyanins, and polysaccharides, which makes Spirulina possesses multifarious functions, such as antioxidant, anti-inflammatory, and immunostimulating effects (Finamore et al., 2017; Grom et al., 2020; Ficagna et al., 2020; Dionisio et al., 2020; Hwang et al., 2019). PSP, extracted from Spirulina, is a marine natural product with almost no side effects, has various favourable effects on the human body, including anti-tumor, anti-inflammatory, anti-edema, anti-oxidation, anti-toxic damage, enhancing the body's immunity, etc (Nishino et al., 1991; Schwartz et al., 1976). In this work, compared to the sepsis rats, the mortality of sepsis rats was decreased and sepsis-related symptoms were significantly improved after receiving PSP treatment, and the W/D ratio of lung (pulmonary edema), lung MPO (pulmonary neutrophil infiltration), lung injury biomarkers protein levels of plasma CC16 and sRAGEs, lung pathological score, as well as the proinflammatory factors proteins of TNF-α and IL-6 in serum, and serum endocan protein levels were reduced in sepsis + PSP group. These findings effective supported the effects of anti-inflammatory, anti-edema, and anti-toxic damage of PSP which have been previously reported (Nishino et al., 1991; Schwartz et al., 1976). Here, we firstly attested that PSP played a protective role in sepsis-caused acute lung damage in rats, which might be related to the effects of anti-inflammatory, anti-edema, and anti-toxic damage of PSP, as well as the inhibitory effect of PSP on sepsis-caused increased endocan protein expression in bronchoalveolar lavage fluid and serum. PSP playing roles is mainly through the peripheral circulatory system rather than directly acting on the lung tissue.

Inflammatory damage is a vital factor of the pathogenesis of septic and histionic injuries (Huang et al., 2019). Hyperinflammation exerts a critical role in the development and progression of acute pulmonary damage induced by sepsis (Fujishima et al., 2016). TNF-α, IL-6, and IL-8 as the main proinflammatory factors in the body, are positively related to the degree of intracorporal inflammatory reaction, and can respond to the level of inflammatory reaction in the body (Chen et al., 2017a; Wang et al., 2019). We found sepsis-induced increased serum TNF-α/IL-6 protein level was positively related to lung damage biomarkers protein CC16/sRAGEs concentration in plasma in rats with sepsis-caused acute pulmonary damage. Previous research has shown that increased plasm CC16 concentration is associated with the enhancive permeability of the alveolar-capillary membrane (Michel et al., 2005). sRAGEs, mainly sourced from alveolar type I cells, plays an anti-inflammatory effect (Engels & van Oeveren, 2015), and as a lung damage sign, increased plasm sRAGEs concentration is related to a higher pulmonary leak index, refracting enhancive permeability of the alveolar-capillary membrane (Tuinman et al., 2013). In addition, excessive release of proinflammatory cytokines is the key to alveolar capillary membrane injury, which is a representative feature of lung damage (Zhang et al., 2018). So, sepsis-induced inflammatory reaction will result in sRAGEs secretion in alveolar

type I cells to resist pro-inflammatory products, then cause a enhancive permeability of the alveolar-capillary membrane, which further aggravate lung damage. Scherpereel, et al. study discovered that the endocan level in serum in sepsis patients was about 4 times higher than that in healthy persons and patients with systemic inflammatory reaction syndrome. The endocan level in septic shock sufferers was markedly higher than that in severe sepsis patients, and the endocan level in death patients was remarkably higher than that in survival patients (Scherpereel et al., 2006). These finds indicated that endocan was participated in sepsis and positively related to the severity of sepsis. Most of the studies related to endocan are collection of serum samples, in our study, in addition to collecting serum samples, bronchoalveolar lavage fluid which can directly reflect lung lesions in early stage was also used to detect endocan expression, and we found that sepsis induced increased endocan protein expression in both bronchoalveolar lavage fluid and serum samples, after treatment with PSP, the serum endocan level was declined. Our study not only testified that endocan involved in sepsis, but also participated in sepsis-caused acute pulmonary damage. Former study has proved that up-regulation of endocan level can lighten paraquat-triggered lung damage in mice (Yang et al., 2018), and endocan has anti-inflammatory property by decreasing the proinflammatory factors levels of TNF-α, IFN-γ, IL-1β, and IL-6, and can relieve acute lung damage caused by LPS (pulmonary epithelium cell apoptosis) (Zhang et al., 2018). In this work, based on the PSP treatment effectively inhibited the inflammatory response and alleviated acute pulmonary damage induced by sepsis, so, the endocan protein level in serum was downregulated in sepsis rats after treating by PSP. Furthermore, there were positive correlations between plasma CC16/sRAGEs concentration and serum endocan concentration, between serum TNF-α/IL-6 concentration and serum endocan content.

In conclusion, sepsis-caused acute lung injury may be connected with enhanced systematic inflammatory reaction and increased endocan expression in serum in sepsis rats. PSP ameliorates acute lung damage through inhibiting inflammatory response and down-regulating endocan expression in sepsis rat model. Moreover, the release of pro-inflammatory factors may lead to the enhancement of acute lung injury caused by sepsis, for the sake of inhibiting the inflammatory reaction, endocan in alveolar capillary endothelial cells is released into blood, then the expression of endocan in bronchoalveolar lavage fluid and serum is increased and followed by relieving acute lung injury caused by sepsis.

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