



Effect of azithromycin combined with fat-soluble vitamin on serum inflammatory cytokines and chemokines as well as lung function in children with mycoplasma pneumonia

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ABSTRACT

Objective: To study the effect of azithromycin combined with fat-soluble vitamin on serum inflammatory cytokines and chemokines as well as lung function in children with mycoplasma pneumonia. **Methods:** 56 children with mycoplasma pneumonia admitted in our hospital between May 2012 and May 2016 were collected, the treatment methods and laboratory test results were retrospectively analyzed, and then they were divided into the control group ($n=26$) who received azithromycin treatment alone and the observation group ($n=30$) who received fat-soluble vitamin combined with azithromycin treatment. Before treatment and after 1 week of treatment, enzyme-linked immunosorbent assay (ELISA) was used to detect the serum levels of inflammatory cytokines and chemokines; lung function monitor for children was used to detect lung function index levels. **Results:** Before treatment, differences in serum inflammation index and chemokine contents as well as lung function indexes were not statistically significant between two groups of patients ($P>0.05$). After treatment, serum inflammation indexes IL-4 and IL-13 contents of observation group were lower than those of control group while IL-10 and IL-12 contents were higher than those of control group ($P<0.05$); serum chemokines MCP-4, MDC and CysLTs contents of observation group were lower than those of control group ($P<0.05$); lung function indexes V-T, FEV1, PEF and MMEF_{25%-75%} levels of observation group were higher than those of control group ($P<0.05$). **Conclusion:** Fat-soluble vitamin combined with azithromycin can reduce the systemic inflammatory response and optimize the lung function in children with mycoplasma pneumoniae infection.

1. Introduction

Mycoplasma pneumoniae is a pediatric mycoplasma pneumonia pathogen, it mostly occurs in infants and children, and the basic pathological changes are interstitial pneumonia and capillary bronchitis[1]. The prognosis of mycoplasma pneumonia is good in general, but disease protraction may occur in some children with poor resistibility and even affect their normal growth and development, and active measures need to be taken. Azithromycin is the main antibiotic for treatment of respiratory tract infection and

has been recommended as the first-line drug for respiratory tract infections in children in many countries and areas, but with the increased application of azithromycin, some children can develop drug resistance, and the clinical treatment effect is limited[2]. Fat-soluble vitamin is one of six major nutrients that can maintain the normal function and metabolism of the body, current studies have shown that fat-soluble vitamins can promote the immunity and resist the inflammation, and therefore, many scholars suggest it for the adjuvant treatment of infantile pneumonia[3,4]. In spite of this, there is no clear report about the effect of fat-soluble vitamin on serum inflammation indexes and pulmonary function indexes in children with mycoplasma pneumoniae infection. In the following study, the effect of azithromycin combined with fat-soluble vitamin on serum inflammatory cytokines and chemokines as well as lung function in children with mycoplasma pneumonia was analyzed.

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2. Materials and methods

2.1. Clinical information

56 children with mycoplasma pneumonia admitted in our hospital between May 2012 and May 2016 were included in the study, the treatment methods and laboratory test results were retrospectively analyzed, and then they were divided into the control group ($n=26$) who received azithromycin treatment alone and the observation group ($n=30$) who received fat-soluble vitamin combined with azithromycin treatment. Control group included 14 male cases and 12 female cases, they were 1–10 years old and the course of disease was 3–9 d; observation group included 16 male cases and 14 female cases, they were 1–10 years old and the course of disease was 2–8 d. The two groups of children were not statistically different in distribution of gender, age and course of disease ($P>0.05$), the families of children signed the informed consent, and the research process was approved by the hospital ethics committee.

Inclusion criteria were as follows: (1) 1–10 years old; (2) not associated with severe congenital diseases; (3) not associated with asthma, variant rhinitis and other chronic upper and lower respiratory tract diseases; (4) without history of lung inflammation 3 months prior to admission. Exclusion criteria were as follows: (1) taking antibiotics and other treatments before admission; (2) with vitamins, antibiotics and other allergy; (3) with severe heart, liver and kidney dysfunction; (4) dropping out of the treatment, and with incomplete clinical data.

2.2. Treatment methods

Both groups received positive symptomatic treatment according to the actual conditions. On the basis of symptomatic treatment, control group of patients received azithromycin treatment specifically as follows: intravenous drip of azithromycin injection (Emeishan Tonghui Pharmaceutical Co., LTD., approved by H20066565) in saline for injection (Beijing Tiantan Biological Products Co., LTD., approved by S10870001), 10 mg/(kg · d), once/d for 5 d in a row, then stopping it for 4 d, switching to oral administration of azithromycin tablets (Zhuhai Rundu Pharmaceutical Co., LTD., approved by H20100102) on an empty stomach, once/d for 3 d in a row, and then stopping it. On the basis of symptomatic treatment, observation group of children received fat-soluble vitamin combined with antibiotic therapy, specifically as follows: intravenous drip of fat-soluble vitamin I for injection (Chengdu Tiantaishan Pharmaceutical Co., LTD., approved by H20052571), once/d for 1 week in a row. Azithromycin usage and dosage were the same as those of control group.

2.3. Serum indexes

Before treatment and after 1 week of treatment, 1 mL of fasting peripheral venous blood was extracted from two groups of children, added in sodium citrate (Sichuan Nigale Biotech Co., LTD., approved by H20046067) for anticoagulation and then centrifuged for 15 min at 2 500 r/min to get supernatant, and the detection indexes were as follows: (1) inflammation indexes: ELISA was used to detect interleukin-4 (IL-4), interleukin-10 (IL-10), interleukin-12 (IL-12) and interleukin-13 (IL-13) levels; (2) chemokines: ELISA was used to determine monocyte chemoattractant protein-4 (MCP-4), macrophage-derived chemokine (MDC) and cysteinyl leukotrienes (CysLTs) levels.

2.4. Lung function

Before treatment and after 1 week of treatment, lung function monitor for children (Sichuan Sikeda Technology Co., LTD., model S-980A I) was used to detect lung function indexes, including tidal volume (V), forced expiratory volume in one second (FEV1), peak expiratory flow (PEF) and maximal mid-expiratory flow velocity (MMEF_{25%-75%}).

2.5. Statistical methods

SPSS15.0 software was used for statistical processing, measurement data was in terms of $\bar{x}\pm s$, comparison before and after treatment was by paired *t* test, comparison between groups after treatment was by routine *t* test and $P<0.05$ indicated statistical significance in differences.

3. Results

3.1. Inflammation indexes

Comparison of serum inflammation indexes IL-4, IL-10, IL-12 and IL-13 contents between two groups of patients was as follows: before treatment, differences in serum inflammation indexes IL-4, IL-10, IL-12 and IL-13 contents were not statistically significant between two groups of patients ($P>0.05$); after 1 week of treatment, serum inflammation indexes IL-4 and IL-13 contents of both groups were lower than those before treatment while IL-10 and IL-12 contents were higher than those before treatment, and differences within same group were statistically significant ($P<0.05$). After 1 week of treatment, serum inflammation indexes IL-4 and IL-13 contents of observation group were lower than those of control group while IL-10 and IL-12 contents were higher than those of control group, and differences between groups were statistically significant ($P<0.05$), shown in Table 1.

Table 1Comparison of serum inflammation index contents before and after treatment ($\bar{x}\pm s$).

Groups	n	IL-4 (pg/mL)		IL-10 (pg/mL)		IL-12 (ng/L)		IL-13 (ng/L)	
		Before treatment	After treatment						
Observation group	30	32.18±4.92	11.53±1.78	12.19±1.76	22.98±3.14	240.17±25.88	357.28±40.91	273.18±30.56	120.36±14.28
Control group	26	33.05±4.51	21.45±2.59	12.07±1.89	17.61±2.07	241.58±26.95	294.72±34.23	274.65±31.19	193.28±25.75
t		0.182	7.394	0.163	6.092	0.271	8.394	0.154	9.232
P		>0.05	<0.05	>0.05	<0.05	>0.05	<0.05	>0.05	<0.05

3.2. Chemokines

Comparison of serum chemokines MCP-4, MDC and CysLTs contents between two groups of patients was as follows: before treatment, differences in serum chemokines MCP-4, MDC and CysLTs contents were not statistically significant between two groups of patients ($P>0.05$); after 1 week of treatment, serum chemokines MCP-4, MDC and CysLTs contents of both groups were lower than those before treatment, and differences within same group were statistically significant ($P<0.05$). After 1 week of treatment, serum chemokines MCP-4, MDC and CysLTs contents of observation group were lower than those of control group, and differences between groups were statistically significant ($P<0.05$), shown in Table 2.

3.3. Lung function

Comparison of lung function indexes V-T, FEV1, PEF and MMEF_{25%-75%} levels between two groups of patients was as follows: before treatment, differences in lung function indexes V-T, FEV1, PEF and MMEF_{25%-75%} levels were not statistically significant between two groups of patients ($P>0.05$); after 1 week of treatment, lung function indexes V-T, FEV1, PEF and MMEF_{25%-75%} levels of both groups were higher than those before treatment, and differences

within same group were statistically significant ($P<0.05$). After 1 week of treatment, lung function indexes V-T, FEV1, PEF and MMEF_{25%-75%} levels of observation group were higher than those of control group, and differences between groups were statistically significant ($P<0.05$), shown in Table 3.

4. Discussion

Children's immune system development is not perfect, they are prone to infection after pathogenic bacteria infection, and the *Mycoplasma pneumoniae* is the most common clinical pediatric pneumonia pathogen[5]. Antibiotics are the main drugs for treatment of pediatric mycoplasma pneumonia, but *Mycoplasma pneumoniae* is without cell wall, and penicillin, cephalosporin and other antibiotics that target the cell wall are invalid for it. Azithromycin is a new macrolide antibiotic, it can both disinfect and enhance immunity, it is the clinically recognized common drug for mycoplasma pneumoniae infection at present, and the curative effect is outstanding. Fat-soluble vitamins include vitamins A, D, E and K, and they have been widely used in clinical treatment of infectious diseases[6,7]. Latest study has shown that the occurrence of mycoplasma pneumonia in children may be directly related to some vitamin deficiency, so the exogenous vitamin supplementation during treatment is expected

Table 2Comparison of serum chemokine contents before and after treatment (pg/mL, $\bar{x}\pm s$).

Groups	n	MCP-4		MDC		CysLTs	
		Before treatment	After treatment	Before treatment	After treatment	Before treatment	After treatment
Observation group	30	115.39±18.76	54.21±5.93	1209.37±151.95	540.26±59.71	1402.64±171.53	409.23±51.18
Control group	26	114.72±19.58	79.83±8.64	1215.83±148.64	892.53±95.82	1453.58±154.32	874.38±97.23
t		0.183	7.293	0.164	9.283	0.172	15.385
P		>0.05	<0.05	>0.05	<0.05	>0.05	<0.05

Table 3Comparison of lung function index levels before and after treatment ($\bar{x}\pm s$).

Groups	n	V-T (mL/kg)		FEV1 (L)		PEF (L)		MMEF _{25%-75%} (L/s)	
		Before treatment	After treatment	Before treatment	After treatment	Before treatment	After treatment	Before treatment	After treatment
Observation group	30	8.62±0.91	12.19±1.76	1.21±0.15	2.76±0.39	3.17±0.39	4.89±0.56	1.41±0.18	2.54±0.31
Control group	26	8.71±0.95	9.84±1.06	1.23±0.17	1.79±0.25	3.25±0.38	3.91±0.42	1.43±0.19	1.81±0.24
t		0.183	7.394	0.163	6.394	0.174	6.092	0.142	6.485
P		>0.05	<0.05	>0.05	<0.05	>0.05	<0.05	>0.05	<0.05

to accelerate disease rehabilitation. In the study, children with pneumonia mycoplasma infection received fat-soluble vitamin combined with azithromycin and were then compared with the children who received azithromycin alone to clarify the reliability of the auxiliary fat-soluble vitamin treatment.

Children with mycoplasma pneumonia are mainly characterized by local and systemic inflammatory response, a large number of inflammatory mediators are released into the circulating blood, and severe cases can lead to other tissue and viscera dysfunction[8]. Imbalance of pro-inflammatory/anti-inflammatory factor expression is the basic mechanism of infectious disease, IL-4 and IL-13 are the pro-inflammatory factors that play an important role in a variety of infectious diseases, and they are mainly secreted by Th2 cells and can induce airway hyperresponsiveness and small airway structure reconstruction[9]. IL-10 and IL-12 are anti-inflammatory factors and are secreted by Th1 cytokines, and they have inhibiting effect on the generation of Th2 cytokines and reduce airway inflammation[10]. Many studies have shown that there is Th1/Th2 cytokine imbalance in patients with lung inflammation, it is mainly characterized by the increased pro-inflammatory factors and the decreased anti-inflammatory factor expression and activity, and therefore, the contents of pro-inflammatory and anti-inflammatory factors can objectively reflect the clinical therapeutic effect[11]. It was found in the study that compared with control group, observation group were with lower serum IL-4 and IL-13 contents, and higher IL-10 and IL-12 contents after 1 week of treatment, confirming that fat-soluble vitamin combined with azithromycin therapy can effectively inhibit the secretion of pro-inflammatory factors and also recover the pro-inflammatory/anti-inflammatory balance.

The massive generation and accumulation of inflammatory factors in the lungs are directly related to the abnormal expression of chemokines, MCP-4 is mainly from the mononuclear cells and epithelial cells, and can selectively recruit the Th2 cells to the inflammatory sites and trigger the immune inflammatory injury[12,13]. MDC is mainly from the mononuclear macrophages, its receptor CCR4 is expressed in the Th2 cells, and it can recruit eosinophils into the airway and release a variety of toxic proteins and cytokines, ultimately prompting airway stenosis and airway limitation[14]. CysLTs is arachidonic acid metabolite, is the strongest bronchoconstrictor, and can also induce high mucus secretion, increase microvascular permeability and aggravate wheezing and coughing. In the study, the contents of above chemokines were detected, and it was found that compared with control group, observation group were with lower serum MCP-4, MDC and CysLTs contents after 1 week of treatment, confirming that after adding adjuvant fat-soluble vitamin therapy, the chemokine secretion reduces, and it may be directly associated with the effect of enhancing the body's resistibility.

The direct consequence of massive generation of inflammatory mediators and chemokines is the continuous airway function deterioration in children with bronchial pneumonia, which is specifically characterized by decreased ventilation capacity and slowed ventilation flow rate[15,16]. V-T, FEV1, PEF and MMEF_{25%-75%} are the visual indicators that reflected large airway and small airway function, and the V-T, FEV1 and PEF levels decrease when the ventilation function declines; MMEF_{25%-75%} level can reflect the small airway function, there is capillary bronchial inflammation in children with bronchopneumonia, and MMEF_{25%-75%} levels may decrease in severe cases[17,18]. In the study, the levels of above lung function indexes were detected, and it was found that compared with control group, observation group were with higher lung function indexes V-T, FEV1, PEF and MMEF_{25%-75%} levels, confirming that after fat-soluble vitamin therapy was added, the pulmonary function in children with bronchial pneumonia is improved significantly, and it further confirms the superior curative effect of combined therapy.

To sum up, it is concluded that fat-soluble vitamin combined with azithromycin has positive clinical value for the treatment of children with mycoplasma pneumoniae infection, can reduce systemic inflammatory response and optimize lung function, and is worth popularization and application in clinical practice in the future.

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