J Infect Chemother 21 (2015) 153–160

Contents lists available at ScienceDirect

Journal of Infection and Chemotherapy

journal homepage: http://www.elsevier.com/locate/jic

Original article

Setting a standard for the initiation of steroid therapy in refractory or severe *Mycoplasma pneumoniae* pneumonia in adolescents and adults

Naoyuki Miyashita [a, *], Yasuhiro Kawai [a], Norikazu Inamura [b], Takaaki Tanaka [b], Hiroto Akaike [b], Hideto Teranishi [b], Tokio Wakabayashi [b], Takashi Nakano [b], Kazunobu Ouchi [b], Niro Okimoto [a]

[a] Department of Internal Medicine 1, Kawasaki Medical School, Okayama, Japan
[b] Department of Pediatrics, Kawasaki Medical School, Okayama, Japan

ARTICLE INFO

Article history:
Received 22 July 2014
Received in revised form
10 October 2014
Accepted 15 October 2014
Available online 19 December 2014

Keywords:
Mycoplasma pneumoniae
Refractory pneumonia
Severe pneumonia
Interleukin 18
Lactate dehydrogenase
Steroid therapy

ABSTRACT

Serum interleukin (IL)-18 level was thought to be a useful as a predictor of refractory or severe *Mycoplasma pneumoniae* pneumonia, and steroid administration is reported to be effective in this situation. The serum levels of IL-18 correlated significantly with those of lactate dehydrogenase (LDH). The purpose of this study was to set a standard for the initiation of steroid therapy in *M. pneumoniae* pneumonia using a simple serum marker. We analyzed 41 adolescent and adult patients with refractory or severe *M. pneumoniae* pneumonia who received steroid therapy, and compared them with 108 patients with *M. pneumoniae* pneumonia who responded to treatment promptly (control group). Serum LDH levels were significantly higher in the refractory and severe group than in the control group at the initiation of steroid therapy (723 vs 210 IU/L, respectively; $p < 0.0001$). From receiver operating characteristic curve analysis, we calculated serum LDH cut-off levels of 364 IU/L at initiation of steroid therapy and 302 IU/L at 1–3 days before the initiation of steroid therapy. The administration of steroids to patients in the refractory and severe group resulted in the rapid improvement of symptoms and a decrease in serum LDH levels in all patients. Serum LDH level can be used as a useful parameter to determine the initiation of steroid therapy in refractory or severe *M. pneumoniae* pneumonia. A serum LDH level of 302–364 IU/L seems to be an appropriate criterion for the initiation of steroid therapy.

1. Introduction

Mycoplasma pneumoniae is one of the major causative pathogens of community-acquired pneumonia (CAP) in children and younger adults [1]. During 2010 and 2012, large epidemics of *M. pneumoniae* infection occurred throughout Japan [2]. Although pneumonia due to *M. pneumoniae* is usually a benign, self-limited disease, some cases are known to develop into refractory or severe, life-threatening pneumonia [3–10]. The pathogenesis of refractory or severe *M. pneumoniae* infections is closely related to an excessive immune response against the pathogen, such as highly activated cell-mediated immune responses and vigorous expression of cytokines [1,11–15]. Thus, immunosuppressive therapy, such as with corticosteroids, downregulates the cell-mediated immune response and has shown a profound beneficial effect by reducing the immune-mediated pulmonary injury seen in mycoplasmal infections [3–10,14–17]. However, the appropriate timing for initiation of steroid therapy has not been investigated.

Interleukin (IL)-18 is believed to be associated with the severity of pneumonia because its level increases particularly in the acute phase and it induces the expression of various cytokines [12–15]. Thus, serum IL-18 level was thought to be a useful as a predictor of refractory or severe *M. pneumoniae* pneumonia [12–15]. However, IL-18 cannot currently be measured in most hospitals and clinics in a timely fashion. In our preliminary study, we found a significant correlation between serum IL-18 and lactate dehydrogenase (LDH) levels [15]. The purpose of this study was to determine whether there was LDH level that could be used to determine which patients will require steroid therapy in *M. pneumoniae* pneumonia. We analyzed serum LDH levels consecutively in adolescent and adult patients with refractory or severe *M. pneumoniae* pneumonia and

* Corresponding author. Department of Internal Medicine 1, Kawasaki Medical School, 2-1-80 Nakasange, Kita-ku, Okayama City, Okayama 700-8505, Japan. Tel.: +81 86 225 2111; fax: +81 86 232 8343.
E-mail address: nao@med.kawasaki-m.ac.jp (N. Miyashita).

http://dx.doi.org/10.1016/j.jiac.2014.10.008
1341-321X/

compared them with levels in *M. pneumoniae* pneumonia patients who responded to antibiotic therapy promptly.

2. Materials and methods

2.1. Study population

This study was conducted by 25 institutions from January 2010 to December 2013. We enrolled adolescent and adult patients with CAP who had suspected *M. pneumoniae* pneumonia, according to the scoring system of the Japanese Respiratory Society (JRS) CAP guidelines [18], and patients with refractory or severe CAP who were initially admitted and treated in other hospitals and transferred to our hospital before initiation of steroid therapy. The diagnosis was based on clinical signs and symptoms (cough, fever, productive sputum, dyspnea, or chest pain) and radiographic pulmonary abnormalities that were at least segmental and not caused by pre-existing or other known causes. Exclusion criteria included immunosuppressive illness (*i.e.*, HIV positive, neutropenia secondary to chemotherapy, use of >20 mg/day prednisone or other immunosuppressive agents, and history of organ transplant); hospitalization in the preceding 30 days; residence in a nursing home or extended care facility; and active tuberculosis. Informed consent was obtained from all patients, and the study protocol was approved by the Ethics Committee at Kawasaki Medical School.

2.2. Definition of refractory or severe M. pneumoniae pneumonia

Refractory *M. pneumoniae* pneumonia was defined as follows: 1) prolonged fever for 7 days or more or 2) increasing cough and infiltrates in chest radiograph despite administration of appropriate antibiotics. Patients with severe *M. pneumoniae* pneumonia who required intensive care unit (ICU) admission were defined by Infectious Diseases Society of America/American Thoracic Society criteria for severe CAP [19]. The 2 major criteria-mechanical ventilation with endotracheal intubation and septic shock requiring vasopressors-are indications for admission to ICU. Patients who met the three or more of the nine minor criteria-respiratory rate $\geq$30 breaths/min, arterial oxygen pressure/fraction of inspired oxygen ratio $\leq$250, multilobar infiltrates, confusion/disorientation, Blood urea nitrogen level $\geq$20 mg/dL, white blood cell count <4000/mm^3, platelet count <100,000/mm^3, core temperature <36 °C, and hypotension requiring aggressive fluid resuscitation-are indications for admission to ICU.

2.3. Study protocol

Nasopharyngeal swab or sputum specimens, serum samples, and urine were collected for microbiological tests at admission. Bronchoscopic examination was employed to obtain specimens in patients with endotracheal intubation. Blood tests for peripheral white blood cell count, C-reactive protein, total protein, LDH, alanine aminotransferase, and aspartate aminotransferase were performed on admission, at 2–4 days after antibiotic therapy, at 5–7 days after antibiotic therapy, at the end of antibiotic therapy, and at 30 days after antibiotic therapy. The serum level of IL-18 was measured on admission with a Human IL-18 ELISA kit (MBL Co. Ltd, Nagoya, Japan) according to the supplier's instructions. Clinical information including laboratory data in patients with refractory or severe *M. pneumoniae* pneumonia before initiation of steroid therapy was collected from former hospitals. Blood tests including LDH were performed in all patients at 1–3 days and 4–7 days before steroid therapy.

After admission, minocycline was administered intravenously twice daily at doses of 100 mg. If adverse events were observed or clinical symptoms and signs had not improved after 3 days minocycline administration, antibiotic therapy was changed to quinolones by the attending physicians. If minocycline was administered before admission to our hospitals, antibiotic was changed to quinolones or β-lactams plus macrolides by the attending physicians. Steroid was administered to patients who met the criteria for refractory or severe *M. pneumoniae* pneumonia.

2.4. Microbiological laboratory tests

Microbiological tests such as Gram stain, cultures for the detection of bacteria, *Legionella* species, *Chlamydia* species, and *M. pneumoniae*, urinary antigen tests for the detection of *Streptococcus pneumoniae* and *L. pneumophila* and serological tests for the detection of viruses, *Legionella* species, *Chlamydia* species, *Coxiella burnetii*, and *M. pneumoniae* were performed as described previously [20,21]. Cultivation of *M. pneumoniae* was carried out using pleuropneumonia-like organism broth (Difco, Detroit, MI, USA). DNA then was extracted by using a QIAamp DNA Mini Kit (QIAGEN K. K., Tokyo, Japan) in accordance with the manufacturer's instructions. *M. pneumonia* DNA was detected by real-time polymerase chain reaction (PCR) targeting a conserved part of the gene encoding P1 adhesin. Antibodies to *M. pneumoniae* were measured using a particle agglutination test (Serodia-Myco II kit, Fujirebio, Tokyo, Japan). The microbial etiology was classified as “definitive”, “presumptive”, or “unknown” as described previously [20,21].

A search for mutations at sites 2063, 2064, and 2617 in the *M. pneumoniae* 23S rRNA domain V gene region was performed using a direct sequencing method in samples with a positive culture or PCR result and the minimum inhibitory concentration (MIC) of 11 antimicrobial agents for the *M. pneumoniae* isolates was determined by micro-dilution methods as described previously [22,23].

2.5. Statistical analysis

Statistical analysis was performed using Stat View version 5.0. (SAS Institute Inc, Cary, NC, USA). The incidence of underlying conditions was analyzed using Fisher's exact test. Serum LDH values are presented as the median ± interquartile range. Mean age of patients and laboratory data were compared using Mann–Whitney's *U* test. Serum LDH cut-off level for the initiation of steroid therapy was calculated by employing a receiver operating characteristic (ROC) curve [24].

3. Results

3.1. Patient characteristics

Cases of *M. pneumoniae* pneumonia mixed with other microorganisms were excluded in this study. During the study period, 108 CAP cases who met all parameters out of six of the JRS scoring system [18] were assessed using the microbiological tests described in the Materials and Methods. The diagnostic sensitivity and specificity of the JRS scoring system for the presumptive diagnosis of *M. pneumoniae* pneumonia were 28–35% and 99–100%, respectively [18,20,21]. All CAP cases were diagnosed as *M. pneumoniae* pneumonia; fifty-four cases were PCR positive, 10 cases were culture positive, and 106 cases demonstrated a four-fold increase in antibody titer. All 108 cases of *M. pneumoniae* pneumonia were responded to minocycline promptly and no relapse was observed after 5–7 days minocycline administration. Finally, we analyzed 108 cases of *M. pneumoniae* pneumonia as a control group.