Chlamydia pneumoniae in Community-Acquired Pneumonia

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Abstract

Chlamydia pneumoniae has been established recently as an important human respiratory pathogen. The aim of this study was to define the prevalence of *C. pneumoniae* in community-acquired pneumonia. We prospectively investigated adult patients who were treated as inpatients and outpatients. Acute and convalescent serum samples were obtained from each patient. Serological diagnosis of *C. pneumoniae* infection was determined by enzyme-linked immunosorbent assay (ELISA). Eighty paired sera were tested for *C. pneumoniae*– specific IgM, IgG and IgA. Twenty-one patients (26.2%) had serological results compatible with acute *C. pneumoniae* infection. Eighteen (85.7%) of these infected patients were *C. pneumoniae*–specific IgM positive, three had a seroconversion of IgA and two had a four-fold or greater increase in *C. pneumoniae*-specific IgG antibody titer. The most common clinical manifestations of community-acquired pneumonia due to *C. pneumoniae* were fever (100%), cough (100%), chest pain (47.6%) and shortness of breath (42.9%). Physical examination revealed crackle in 85.7 per cent of the cases. These findings suggest that *C. pneumoniae* is a common cause of community-acquired pneumonia in Thailand.

Key word : Community-Acquired Pneumonia, *Chlamydia Pneumoniae*

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Chlamydia pneumoniae, the third member of the genus chlamydia, is gram negative bacteria which is an obligate intracellular organism of eukaryotic cells. Most respiratory infections caused by *C. pneumoniae* are mild or asymptomatic (1-4). Similar to *Mycoplasma pneumoniae* infec-

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tions, *C. pneumoniae* can cause recurrent or secondary lower respiratory tract infections since antibodies due to previous infection are detectable in serum[1]. Infection with *C. pneumoniae* occurs worldwide, resulting in a 40 to 90 per cent prevalence of serum antibody to the species [8,17]. *C. pneumoniae* causes both upper and lower respiratory tract infections such as sinusitis, pharyngitis, otitis media, bronchitis and pneumonia [9,10]. It has been reported that *C. pneumoniae* was responsible for 6-20 per cent of all community-acquired pneumonia (CAP)[11-13]. In Thailand, Nunthapisud et al reported that 2.5 per cent of children with respiratory tract infection was caused by *C. pneumoniae*[14]. However, the role of *C. pneumoniae* as the cause of CAP in adult patients is unknown. To evaluate the prevalence of *C. pneumoniae* in adult patients with CAP, we conducted a prospective study at three hospitals in Thailand.

PATIENTS AND METHOD

Patients

This prospective study was done at three participating hospitals (Phramongkutklao Hospital, Deja Hospital and Bangkok-Prapradang Hospital) from September 1998 to October 1999. The study population included all hospitalized patients and outpatients with community-acquired pneumonia. Inclusion criteria of the study were adapted from those used by Fang and colleagues as follows[10]: (a) adult patients more than 18 years of age with a putative diagnosis pneumonia made within 24 hours of visiting physicians, (b) a new pulmonary infiltration on chest radiograph, and (c) confirmatory clinical findings of one major criteria of either cough, sputum production, or temperature more than 37.8°C, or two minor criteria of pleuritic chest pain, dyspnea, altered mental status, pulmonary consolidation by physical examination, or white blood cell count more than 12,000 cell/ mm^3. Patients who were transferred from another hospital or hospitalized within 3 weeks before admission were excluded. Patients were also excluded if chest roentgenograms did not reveal a new infiltration or if radiographic abnormalities were attributable to a noninfectious etiology (e.g. pulmonary or septic emboli, pulmonary edema, malignancy). The study was approved by the committee on clinical investigation of the Phramongkutklao Hospital.

Serum samples

A serum sample was obtained at the time of enrollment for serological testing. Convalescent sera were obtained at 4-6 weeks intervals at follow-up appointment. All serum samples were separated immediately and stored at –30°C until tested.

Serology

The serum samples were tested for *C. pneumoniae* - specific IgM, IgG and IgA antibodies by enzyme-linked immunosorbent assay (ELISA Sero CP; Savyoun Diagnostics, Ashdol, Israel). In this test *C. pneumoniae* purified elementary bodies were used as antigen. Each serum sample was mixed thoroughly before processing according to the manufacturer's instructions. Initial sera to be tested were diluted 1:105. Results were spectrophotometrically determined and expressed as optical density units. Positive and negative results were determined according to COV (cut-off value) and COI (cut-off index). COI $\geq$ 1.1 indicated positive for *C. pneumoniae*–specific IgM, IgG and IgA antibodies. For all the patients with positive for *C. pneumoniae*–specific IgG or IgA antibodies in the first serum, the convalescent sera were tested at a pre–dilution of 1:4. A two-fold serially diluted standard serum was used to calculate the titre of patients' samples. Serological diagnosis of an acute chlamydia infection was based on the ELISA results with the following criteria: positive IgM antibody in one serum sample, or IgA or IgG antibody not detectable in the acute serum and became positive in convalescent serum (seroconversion), or a four-fold or greater increase in *C. pneumoniae*–specific IgG or IgA antibody titer if the first sera were positive for *C. pneumoniae*–specific IgG or IgA antibody.

RESULTS

Eighty paired sera were tested for *C. pneumoniae*– specific IgM, IgG and IgA. The mean age of the patients was 42.80 years (range 18 - 87 years). Forty four (55.0%) of the patients were male and 36 (45%) were female. Thirty four (42.5%) were admitted to one of three hospitals. Twenty-one (26.25%) showed evidence of acute *C. pneumoniae* infection. The results are summarized in Table 1. Out of the twenty one proven *C. pneumoniae*-infected patients, 18 cases (85.70%) were positive for IgM antibody,

Table 1. Results of serological test from 21 patients with *C. pneumoniae* pneumonia.

Diagnostic criteria	patients	percentage
IgM Ab positive	18	85.70
Seroconversion of IgA Ab	3	14.30
Seroconversion of IgA Ab only	1	4.70
A four-fold or greater increase in IgG Ab titer	2	9.50
A four-fold or greater increase in IgG Ab titer only	1	4.70
Seroconversion of IgA and A four-fold or greater increase in IgG Ab titer	1	4.70
Total	21	100

Table 2. Characteristics of 21 patients with community-acquired pneumonia due to *C. pneumoniae*.

Characteristic	Value
Demographic	
Mean age in year (range)	43.4 (18-86)
Male (%)	47.6
Female (%)	52.4
Patients with a comorbid illness (COPD, IHD, DM, HT, renal failure)(%)	23.8
Symptom (% of patients)	
Fever	100
Chill	28.6
Cough (productive)	100 (57.1)
Shortness of breath	42.9
Headache	14.3
Chest pain	47.6
Rhinitis	0
Diarrhea	10
Hoarseness	4.8
Change mental status	4.8
Duration of illness (d) before visiting physician	
Mean	7.1
Range	2-21
Finding	
Mean temperature (°C)	38.1
Patients with Tem ≤ 37.8°C (%)	38
Patients with Tem ≥ 38.9°C (%)	19
Patients with	
Respiratory rate > 30 breath/min (%)	9.5
Heart rate > 130 (%)	0
Systolic BP < 90 mmHg (%)	0
Bronchial breath sound (%)	23.8
Rales (%)	85.7
Wheezing (%)	19.0
Rhonchi (%)	4.8
Lymphadenopathy (%)	4.8
Laboratory finding	
Mean WBC count per mm^3 (range)	11,952 (3,100-38,800)
Patients with WBC > 10,000 cell/mm^3 (%)	57.1

3 had seroconversion of IgA and 2 had a four-fold or greater increase in *C. pneumoniae*-specific IgG antibody titer. Specific IgM antibodies were detected in 85.7 per cent of the patients, suggesting that the majority of these cases represented primary infection. Three cases (14.30%) were considered to have serological evidence of reinfection.

The characteristics of twenty-one patients with CAP due to *C. pneumoniae* are shown in Table 2. In general, the most common clinical manifestations were fever (100% of cases), cough (100% of cases, productive in 57.10%), shortness of breath (42.90%), and chest pain (47.60%). Chest auscultation revealed pneumonic rale in 85.70 per cent of the cases followed by bronchial breath sound (23.80%), wheezing (19%), and rhonchi (4.80%). One patient showed cervical lymphadenopathy which had disappeared at follow-up appointment.

Initial empiric antimicrobial treatment after visiting physicians comprised monotherapy (n = 12) and dual combination therapy (n = 9). Only 81 per cent of the patients had received appropriate antimicrobial treatment (Table 3). One patient who was treated with inappropriate antibiotics died from respiratory failure, whereas the other patients survived.

DISCUSSION

C. pneumoniae is recognized as a common cause of both endemic and epidemic pneumonia throughout the world[15,16]. Studies on seroprevalence indicated a worldwide incidence in the range of 40-90 per cent among the adult population[8,17]. In Thailand, the antibody detection rate of *C. pneumoniae* in healthy adults was 70 per cent for IgG and 43 per cent for IgA[8]. The reason behind the high seroprevalence rate in Thailand is unknown. It might indicate that Thailand has recently experienced an epidemic of *C. pneumoniae* similar to previously documented outbreaks in Scandinavia and Rhode Island[17-20].

Serology is the most commonly used diagnostic tool for the detection of respiratory *Chlamydia* infections in routine clinical practice. In the early 1970s, a sensitive micro-immuno-

Table 3. Antimicrobial treatment in patients with *C. pneumoniae* pneumonia.

Antibiotics	n	%
Monotherapy	12	57.1
Amoxicillin/clavulanic acid	4	19
Doxycycline	1	4.8
Roxithromycin	2	9.5
Azithromycin	1	4.8
Sparfloxacin	3	14.3
Levofloxacin	1	4.8
Appropriate treatment*	17	81
Dual combination therapy	9	42.9
Erythromycin + Amoxicillin/clavulanic acid	4	19
Roxithromycin + Ampicillin/sulbactam	1	4.8
Roxithromycin + 2nd/ 3rd gen. CS	3	14.3
Sparfloxacin + penicillin	1	4.8

* Erythromycin, roxithromycin, doxycycline, azithromycin, sparfloxacin, or levofloxacin; 2nd/ 3rd gen. CS = second/ third-generation cephalosporin.

fluorescence (MIF) assay was developed which proved to be suitable for routine diagnosis[21,22]. In this MIF test, purified elementary bodies are used to detect specific chlamydia antibodies in the IgM, IgG and IgA serum fractions. The MIF test is generally accepted as the gold standard for the serological diagnosis of acute *C. pneumoniae* infection. However, interpretation of specific and non-specific fluorescence patterns requires experience and skill. The interpretation of high titers (≥ 1:512) is very difficult and subjective. Our study used the ELISA method carried out with an automated instrument. The strips were coated with intact *C. pneumoniae* purified-elementary bodies as used in the MIF method. It is highly sensitive and specific for the detection of *C. pneumoniae* antibodies and easy to perform.

The serologic response to acute *C. pneumoniae* infection is characterized by two different patterns. Primary infection shows an IgM antibody titer rise within 2 to 4 weeks followed by IgA and IgG antibody increase within 6 to 8 weeks, whereas, reinfection shows IgG and IgA antibody titer rises quickly without increase of IgM antibody. The prevalence of *C. pneumoniae* as the cause of CAP has ranged from 3 per cent to 22 per cent[11-13,23] and could be as high as 43 per cent during seasonal outbreaks[12]. In the present study, 26.25 per cent of patients with CAP were due to *C. pneumoniae*. Among the 21 proven *C. pneumoniae*-infected patients, 18 cases (85.70%) were positive for IgM antibodies which suggested that the majority of these patients represent primary infection. Three patients had a seroconversion of IgA and 2 had a four-fold or greater increase in *C. pneumoniae*-specific IgG antibody titer.

C. pneumoniae pneumonia often presents initially with sore throat, hoarseness, and headache as important nonclassic pneumonic findings. Cough with scanty sputum is prominent which may last if not treated early and effectively. From studies that characterized the clinical manifestations of patients with *C. pneumoniae* as the sole cause of CAP (excluding the effects of copathogens), a subacute course is common and fever is low grade [6,7]. However, no distinguishing clinical characteristics identify cases of *C. pneumoniae* infection. In one study comparing *C. pneumoniae* infection to all other causes of pneumonia, no significant differences in symptoms, physical examination findings, or routine laboratory results were apparent[2]. Some common features include gradual onset of symptoms, associated pharyngitis, and often hoarseness[2]. In our study, the most common clinical manifestations were fever, cough, shortness of breath and chest pain. The results suggested that patients with CAP due to *C. pneumoniae* had characteristics similar to those of patients with pneumonia caused by other etiologies. Although no sign or symptoms appear to be unique to pneumonia due to *C. pneumoniae,* several conclusions may be made from our results as well as these of previous studies[2,6,7]. A subacute course is common and fever is usually low grade. Although only 81 per cent had received the appropriate antimicrobial treatment, all the patients except one fully recovered. Reports from Finland and North America have also mentioned that patients with *C. pneumoniae* pneumonia recovered without adequate antibiotic treatment[6,26]. Current guidelines for CAP stress the importance of *C. pneumoniae* as an etiological agent and prescribing macrolides or new fluoroquinolones which are also effective against other pathogens that cause CAP such as *Streptococcus pneumoniae, Mycoplasma pneumoniae and Legionella pneumophila* [24,25]. From our studies, *C. pneumoniae* is a common cause of CAP. In Thailand, we suggest that empirical treatment with macrolides or new fluoroquinolones is important in adult patients with CAP[5,24,25].

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การศึกษาความชุกของเชื้อ*คลามีเดีย นิวโมเนอี* ในผู้ป่วยโรคปอดบวมที่เกิดจากการติดเชื้อนอกโรงพยาบาล

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คณะวิจัยได้ทำการศึกษาหาความชุกของเชื้อ*คลามีเดีย นิวโมเนอี* ในผู้ป่วยโรคปอดบวมที่เกิดจากการติดเชื้อนอกโรงพยาบาล ได้ผู้ป่วยโรคปอดบวมที่ได้รับการรักษาในโรงพยาบาล และนอกโรงพยาบาลจำนวน 80 คน การวินิจฉัย*คลามีเดีย นิวโมเนอีใช้* serology โดยวิธี ELISA ผู้ป่วยทุกรายได้รับการเจาะเลือดสองครั้ง ครั้งแรกในขณะที่ได้รับการวินิจฉัยว่าเป็นโรคปอดบวม และหลังจากนั้นอีก 4–6 สัปดาห์ ซีรั่มจะนำไปตรวจ IgM, IgG และ IgA แอนติบอดี เกณฑ์การวินิจฉัยการติดเชื้อ*คลามีเดีย* ได้แก่ พบ IgM แอนติบอดี, seroconversion ของ IgG หรือ IgA แอนติบอดี, มี IgG หรือ IgA แอนติบอดี เพิ่มขึ้นมากกว่าหรือเท่ากับ 4 เท่า เมื่อเทียบกับ IgG หรือ IgA แอนติบอดีในซีรั่มครั้งแรก พบการติดเชื้อ*คลามีเดีย นิวโมเนอี* 21 คน (26.25 เปอร์เซ็นต์), 18 คน (85.70 เปอร์เซ็นต์) พบ IgM แอนติบอดี, 3 คนมี seroconversion ของ IgA แอนติบอดี และ 2 คนที่มี IgG แอนติบอดีเพิ่มขึ้นมากกว่า 4 เท่า อาการของผู้ป่วยโรคปอดบวมที่เกิดจากเชื้อ*คลามีเดีย นิวโมเนอี*ที่พบบ่อยได้แก่ ไข้ (100 เปอร์เซ็นต์) ไอ (100 เปอร์เซ็นต์) เจ็บหน้าอก (47.60 เปอร์เซ็นต์) และหอบเหนื่อย (42.90 เปอร์เซ็นต์) การตรวจร่างกายพบเสียง crackle (85.70 เปอร์เซ็นต์) จากผลการวิจัยพบว่าในประเทศไทยเชื้อ*คลามีเดีย นิวโมเนอี*เป็นสาเหตุที่พบบ่อยของโรคปอดบวมที่เกิดจากการติดเชื้อนอกโรงพยาบาล

คำสำคัญ : โรคปอดบวมที่เกิดจากการติดเชื้อนอกโรงพยาบาล, *คลามีเดีย นิวโมเนอี*

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