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ORIGINAL ARTICLE

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Prospective multicenter study of the causative organisms of community-acquired pneumonia in adults in Japan

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Abstract Although a wide variety of recognized pathogens causes community-acquired pneumonia, the precise etiology in Japan remains unknown. We prospectively investigated the etiology in 232 patients with community-acquired pneumonia who visited 20 community-general hospitals. New diagnostic methods, using polymerase chain reaction (PCR) assays and urinary antigen tests were employed, in addition to conventional methods. The frequency of identification of causative pathogens was high (73.3%), and the leading organism was *Streptococcus pneumoniae* (24.6%), followed by *Haemophilus influenzae* (18.5%), viruses (16.4%), *Chlamydia pneumoniae* (6.5%), *Mycoplasma pneumoniae* (5.2%), and *Legionella* spp. (3.9%). *S. pneumoniae* and *M. pneumoniae* were the most prevalent pathogens in younger patients, and *S. pneumoniae* and *H. influenzae* were the most prevalent in elderly patients. Multiple or mixed infections were found in 25.9% of all patients and in 35.3% with a causal diagnosis. The results have important practical implications for the initial treatment of adult patients with community-acquired pneumonia.

Key words Community-acquired · Pneumonia · Etiology · Adult

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Introduction

Community-acquired pneumonia has been a common and important illness despite the availability of potent new antimicrobial agents and the development of effective antimicrobial chemotherapy. In Japan, pneumonia is the fourth leading cause of death, and from 57 to 70 persons per 100000 population died per year of this disease in the past decade.

The initial management of community-acquired pneumonia usually starts empirically because of the unknown detailed etiology at the time. Physicians should refer to the frequency of causative agents when they start empiric therapy, for it is one of the critical factors in the initial management. Many prospective studies of the etiology of community-acquired pneumonia in hospitalized patients have been reported in Western countries, and guidelines for the management of community-acquired pneumonia have been published on the basis of the findings.[1–4]

A guideline for the management of community-acquired pneumonia was also published in 2000 in Japan.[5] Unfortunately, only two prospective studies of the etiology of community-acquired pneumonia have been reported in Japan.[6,7] These two study groups showed a few differences in the frequency distribution from that in Western countries (e.g., lower frequency of *Legionella*), and the frequency may vary among different geographical regions, because the

studies were conducted in a limited geographic area. A more precise prospective study has been strongly desired to elucidate the etiology, and to improve the management of community-acquired pneumonia.

The present study was a prospective multicenter study of the etiology of community-acquired pneumonia in several regions in Japan. The characteristics of this study are: (i) the enrollment of all patients with pneumonia accessing hospitals of the investigators, including both hospitalized patients and outpatients, and (ii) atypical agents, including *Legionella*, *Mycoplasma*, *Chlamydia*, *Coxiella*, and viruses were identified in investigators' laboratories that specialized in the identification of each pathogen, using several suitable diagnostic tools and tests to increase accuracy for such identification.

Patients and methods

Patients

Two hundred and thirty-nine patients with community-acquired pneumonia were enrolled in this study; they accessed the outpatient clinics of 20 community general hospitals in eight prefectures (6 hospitals in Okinawa; 4 hospitals each in Fukuoka and Nagasaki; 2 hospitals in Okayama; and 1 hospital each in Miyagi, Yamagata, Fukushima, and Shizuoka) between December 1999 and March 2000. Community-acquired pneumonia was defined as the recognition of new infiltration on chest X-ray when the patients suffered from symptoms suggesting pneumonia, including cough, fever, dyspnea, sputum production, and/or pleuritic chest pain. Patients with mild pneumonia were treated by the internal medicine staff and/or the investigators at the outpatient clinics of the hospitals, and those with moderate and severe pneumonia were admitted to the hospitals and treated by them. Patients were excluded from this study if their abnormalities on the chest X-ray were attributed to other causes, such as congestive heart failure, pulmonary infarction, obstructive pneumonia due to lung tumors, interstitial lung disease, or sarcoidosis. All patients gave their informed consent to participate in this study.

Microbiological laboratory studies

If purulent expectorated sputum or deep-cough specimens were obtained, a gram stain test and a quantitative or semiquantitative culture of the sputum were performed as routine sputum tests at each hospital. Sputum data were evaluated only when the gram stain test revealed numerous leukocytes (>25 in a 100 × microscopic field) and few epithelial cells (<10 in a 100 × microscopic field).[8] The gram-stain readings in all patients were rechecked by one of the investigators, a specialist in clinical microbiology, because the validity of the results was related to the experience of the interpreter.[9] Invasive diagnostic tests, including bronchoscopy and transthoracic needle aspiration, were performed to obtain uncontaminated lower respiratory secretions in several patients who were suspected to have anaerobic infection. Expectorated purulent sputum or deep-cough specimens were also used for cultures of *Legionella* and *Chlamydia*; and for polymerase chain reaction (PCR) of *Legionella*, *Chlamydia*, *Mycoplasma pneumoniae*, and viruses. The cultures for *Legionella* and *Chlamydia* were performed as previously described; the specimens were plated on buffered charcoal-yeast extract alpha and modified Wadowsky Yee (MWY) agars,[10] and in cycloheximide-treated HEp-2 cells grown in 24-well cell culture plates,[11] respectively. Blood cultures were obtained on admission for the hospitalized patients.

Serum samples were also obtained from all patients for serological tests. Second and third (convalescent) serum samples were obtained from patients, usually at follow-up appointments 2 weeks and 4 weeks after the first sampling. All serum samples were separated immediately and stored at −70°C until tested. We tested the sera for the presence of specific antibodies to *Chlamydia*, *M. pneumoniae*, *Legionella*, *Coxiella burnetii*, and the *Streptococcus milleri* group.

Chlamydia pneumoniae and *Chlamydia psittaci* were determined by a microimmunofluorescence (MIF) method, utilizing *C. pneumoniae* KK-pn15 and TW-183, *C. trachomatis* L2/434/Bu, and *C. psittaci* 6BC strains as antigens to detect IgG and IgM antibodies to chlamydial species.[11,12] *M. pneumoniae* was determined by microparticle agglutination (PA) or complement fixation (CF) methods. *Legionella*, *C. burnetii*, and the *S. milleri* group were detected using an indirect fluorescence antibody (IFA) test.[13,14] Formalin-fixed bacteria served as antigens: standard strains of *Legionella*, including *L. pneumophila* serogroup 1–6, *Legionella bozemanii*, *Legionella dumoffii*, and *Legionella micdadei*, Nine Mile strain phase II of *C. burnetii*, and standard strains of the *S. milleri* group such as *S. anginosus*, *S. constellatus*, and *S. intermedius* were used. Viral antibody titer was assessed by CF for adenovirus and respiratory syncytial (RS) virus, and by hemagglutination inhibition for influenza A and B viruses and parainfluenza virus.

The PCR assays of *Legionella*, *Chlamydia*, *Mycoplasma pneumoniae*, and viruses including influenza A and B viruses, parainfluenza virus, adenovirus, and RS respiratory syncytial virus were performed according to the methods described in previous reports.[10,15,16–20]

Urinary antigen test for *Legionella* species was detected using commercial kits, Binax Now (Binax, ME, USA) and Biotest (Biotest, Dreieich, Germany). Influenza viruses A and B were also detected with several commercial kits used at each clinic or hospital.

The culture, PCR assay, serological test, and urinary antigen test for *Legionella* and serological test for the *S. milleri* group were performed by the investigator's laboratory at the University of the Ryukyus, the culture, PCR assay, and serological test for *Chlamydia*, by that at Kawasaki Medical School; the PCR assay for *M. pneumoniae* by that at Nagasaki University; the serological test for *C. burnetti* by that at Kagoshima University; and the PCR and serological test for viruses were performed by the

Table 1. General characteristics of study population (n = 232)

Mean age (years)	60.2	(range, 17 to 99)
Sex (M/F)	134/98	
No. of patients with underlying diseases	140	(60.3%)
Hypertension	25	(10.8%)
Diabetes mellitus	20	(8.6%)
Chronic obstructive pulmonary disease	19	(8.2%)
Old pulmonary tuberculosis	14	(6.0%)
Cardiac diseases	11	(4.7%)
Bronchial asthma	11	(4.7%)
Malignancy	10	(4.3%)
Chronic liver diseases	9	(3.9%)
Arrhythmia	9	(3.9%)
Bronchopulmonary diseases	7	(3.0%)
Rheumatoid arthritis	7	(3.0%)
Digestive ulcer	6	(2.6%)
Postgastrectomy or esophagectomy	5	(2.2%)
Cerebrovascular diseases	4	(1.7%)
Alcoholism	4	(1.7%)
Hypothyroidism	4	(1.7%)
Psychological disorder	3	(1.3%)
Miscellaneous	33	(14.2%)

Kitasato Otsuka Virus Assay Laboratory and that of Kurume University.

Criteria for determination of microbial etiology

A confirmed causative diagnosis of community-acquired pneumonia required one of the following:

1. Recovery of a likely pulmonary pathogen from an uncontaminated specimen source, including blood, pleural fluid, or transthoracic lung aspirates.
2. Detection of an organism in sputum culture – and/or detection of specific DNA or antigen of an organism by PCR or urinary antigen assay – that is a likely pulmonary pathogen and does not colonize the upper or lower airways in the absence of disease, such as *Mycobacterium tuberculosis*, *Legionella* species, pathogenic fungi, influenza virus, RS virus, adenovirus, and *Pneumocystis carinii*. *C. pneumoniae* detected using a PCR assay was considered definitive only when results showed an increase in antibody titers between paired serum samples, because it has occasionally been recovered from healthy adults, although only rarely. *M. pneumoniae* and viruses detected using PCR assays were also considered definitive when the clinical course of patients was compatible with pneumonia caused by the organisms, as judged by the investigators.
3. An organism showing positive serological tests of a fourfold or greater increase in the antibody titer level in paired serum samples. An organism was also considered as a presumptive pathogen when an antibody titer for *M. pneumoniae* of at least 1:160 was detected using the PA assay, that for *Chlamidia* species showed an IgM titer of ≥16, that for *Legionella* species showed at least 1:256, that for *C. burnetii* showed at least 1:80, and that for the *S. milleri* group showed at least 1:512.
4. Bacteria recovered from expectorated sputum at high concentrations (≥10^7 colony-forming units [CFU]/ml) of a predominant), or at moderate concentrations (10^5 to 10^6 CFU/ml) with the finding of phagocytosed bacteria compatible with the culture results on cytological screening by a gram stain test.

Data analysis

The χ^2 test was used to determine the significance of differences in proportions between groups.

Results

Patient characteristics

Two hundred and thirty-two patients (134 male and 98 female) were finally analyzed in the present study. Seven patients were subsequently excluded: 2 with acute eosinophilic pneumonia, the others with obstructive pneumonia, lung cancer, hypersensitive pneumonia, drug-induced pneumonia, and wrong diagnosis with other disease, respectively. The mean age of the patients was 60.2 years (range, 17 to 99 years).

One hundred and forty patients (60.3%) carried at least one underlying diseases (Table 1). The major diseases were hypertension (10.8%), diabetes mellitus (8.6%), chronic obstructive pulmonary disease (COPD; 8.2%), and old pulmonary tuberculosis (6.0%). The number of patients with underlying diseases was 8 under 40 years old (21.0%), 47 between 40 and 64 years old (58.8%), and 85 over 64 years old (74.6%). There were no patients with AIDS and no organ transplant recipients. Fifty-four patients (23.3%) had received prior antimicrobial therapy for the diagnosis of bronchitis or upper respiratory tract infections before they were enrolled in this study.

Two hundred patients were hospitalized when they were diagnosed with pneumonia. Thirty-two patients received

Table 2. Causative agents identified in 232 patients with community-acquired pneumonia, and their distribution by age

Etiology	*n* (%)	Age (years)		
		<40 (*n* = 38)	40–64 (*n* = 80)	≥65 (*n* = 114)
Streptococcus pneumoniae	57 (24.6)	6 (15.8)	19 (23.8)	32 (28.1)
Streptococcus milleri group	5 (2.2)	0	0	5 (4.4)
Streptococcus spp.	2 (0.9)	0	0	2 (1.8)
Staphylococcus aureus	8 (3.4)	1 (2.6)	3 (3.8)	4 (3.5)
Corynebacterium spp	2 (0.9)	1 (2.6)	0	1 (0.9)
Moraxella catarrhalis	5 (2.2)	0	1 (1.3)	4 (3.5)
Haemophilus influenzae	43 (18.5)	4 (10.5)	16 (20.0)	23 (20.2)
Haemophilus parainfluenzae	1 (0.4)	0	0	1 (0.9)
Klebsiella pneumoniae	3 (1.3)	0	0	3 (2.6)
Pseudomonas aeruginosa	1 (0.4)	0	0	1 (0.9)
Legionella spp.	9 (3.9)	1 (2.6)	5 (6.3)	3 (2.6)
Anaerobes[a]	9 (3.9)	0	4 (5.0)	5 (4.4)
Mycoplasma pneumoniae	12 (5.2)	6 (15.8)	4 (5.0)	2 (1.8)
Chlamydia pneumoniae	15 (6.5)	3 (7.9)	4 (5.0)	8 (7.0)
Chlamydia psittaci	5 (2.2)	0	2 (2.5)	3 (2.6)
Coxiella burnetii	2 (0.9)	1 (2.6)	0	1 (0.9)
Cryptococcus neoformans	1 (0.4)	1 (2.6)	0	0
Viruses[b]	38 (16.4)	6 (15.8)	17 (21.3)	15 (13.2)
Unknown	62 (26.7)	11 (28.9)	23 (28.8)	28 (24.6)

[a] Including 2 *Peptostreptococcus* spp., 4 *Prevotella* spp., 1 *Bacteroides fragilis*, 1 *Fusobacterium nucleatum*, and 1 *Eubacterium limoso*

[b] Including 31 influenza A, 2 parainfluenza virus, 1 respiratory sincytial virus, 3 adenovirus, and 1 *Varicella zoster* virus

antimicrobial therapy at an outpatient clinic, and 1 of them was admitted to hospital later because of deterioration of status.

Etiology of pneumonia

Table 2 shows the frequencies of the various causal agents for community-acquired pneumonia among the 232 patients and their distribution by age. Causative pathogens were identified in 170 patents (73.3%). The most frequently identified pathogen was *Streptococcus pneumoniae,* which was identified in 57 (24.6%) of the patients, including 4 (1.7%) complicated with bacteremia. The next most common agent was *Haemophilus influenzae*, which was identified in 43 patients (18.5%), followed by *C. pneumoniae* in 15 (6.5%), and *M. pneumoniae* in 12 (5.2%). The frequencies of other agents were 3.9% for *Legionella* spp. (9 patients), 3.4% for *Staphylococcus aureus* (8 patients, including methicillin-resistant *S. aureus* in a patient), and 2.2 % for *C. psittaci, S. milleri* group, and *Moraxella catarrhalis*, respectively (5 patients). The frequency of "atypical" pathogens, including *M. pneumoniae*, *C. pneumoniae*, *Legionella* spp., and *C. burnetii* reached 16.5%, in addition to viruses whose frequency was 16.4% (including Influenza A, 13.4%).

The commonest causal agents in younger patients (less than 40 years of age) were *S. pneumoniae* and *M. pneumoniae* (both 15.8%), followed by *H. influenzae* (10.5%) and *C. pneumoniae* (7.9%). In the middle aged group (40–64 years) and the older group (over 65), *S. pneumoniae* (23.8% and 28.1%, respectively) was the most common pathogen, followed by *H. influenzae* (20.0% and 20.2%, respectively). *Legionella* was the third most common in patients aged 40–64 years, and *C. pneumoniae* was third most common in patients above the age of 65. The frequency of atypical pathogens such as *M. pneumoniae*, *C. pneumoniae*, *C. psittaci*, and *C. burnetii* was significantly higher in the younger group than in the other two groups (26.3% vs 12.5% and 12.3%: $P < 0.001$), while that of *H. influenzae*, as well as anaerobes, was higher in older groups ($P < 0.001$). Gram-negative bacilli and streptococci, including to *S. milleri* group, were especially frequent in patients above the age of 65.

Two hundred and seventy-five different causal agents were identified in 170 patients, implying a high rate of multiple causes (1.6 agents per patient). A single agent was found in 110 patients (47.4% of all patients and 64.7% with a causal diagnosis), while two to two causal agents were found in 60 patients (25.9% and 35.3%, respectively). Various causal agents were revealed in mixed infections (Table 3), and an association between *S. pneumoniae* and atypical pathogens was implied.

The yield of detection of causative organisms from patients who had not previously received antibiotics was 74.7% (133 of 178 patients), while it was 64.8 % (35 of 54 patients) from patients with prior antibiotic therapy ($P = 0.094$).

Blood cultures were performed in 150 patients (64.7%); bacteria were detected in only 5 (2.2%). Isolates were *S. pneumoniae* in 4 patients and *Staphylococcus capitis* in 1 patient, and the organism in this latter patient was considered as normal flora of the skin from which the blood sample was taken.

Table 3. Distribution of associations between the various causal agents in mixed infections

	S. pneumoniae	*S. milleri* group	*S. aureus*	*M. catarrhalis*	*H. influenzae*	*H. parainfluenzae*	*Legionella* sp.	Anaerobes	*M. pneumoniae*	*C. pneumoniae*	*C. psittaci*	*C. burnetii*	Viruses	Single case (%)[a]
Streptococcus pneumoniae	–	1	1	1	3	0	1	0	2	2	1	1	9	37 (15.9)
Streptococcus milleri group	1	–	0	0	0	0	0	2	0	0	0	0	0	3 (1.3)
Staphylococcus aureus	1	0	–	0	0	0	0	0	0	0	0	0	2	5 (2.2)
Moraxella catarrhalis	1	0	0	–	1	0	1	0	0	2	0	0	0	2 (0.9)
Haemophilus influenzae	3	0	0	1	–	0	2	0	1	2	1	1	3	30 (12.9)
Haemophilus parainfluenzae	0	0	0	0	0	–	0	2	0	0	0	0	0	0
Legionella spp.	1	0	0	1	2	0	–	0	0	0	1	0	0	5 (2.2)
Anaerobes	0	2	0	0	0	2	0	–	0	0	0	0	0	1 (0.4)
Mycoplasma pneumoniae	2	0	0	0	1	0	0	0	–	1	0	0	0	8 (3.4)
Chlamydia pneumoniae	2	0	0	2	2	0	0	0	1	–	0	0	0	9 (3.9)
Chlamydia psittaci	1	0	0	0	1	0	1	0	0	0	–	0	1	1 (0.4)
Coxiella burnetii	1	0	0	0	1	0	0	0	0	0	0	–	0	0
Viruses	9	0	2	0	3	0	0	0	0	0	1	0	–	22 (9.5)
Total[b]	22	3	3	5	12	2	5	4	4	7	4	2	15	

[a] Percentage of all patients infected with a single agents
[b] The number was counted as in each when more than two agents were identified in one patient

Severity of the infection

The severity of pneumonia was categorized as three illness statuses: "mild", "moderate", and "severe", using the criteria established previously by a report from the Committee for the Respiratory System, Japan Society of Chemotherapy.[21] In brief, "mild" and "severe" pneumonia were defined as illness meeting three or more of the following four criteria: body temperature (<37.5°C, and ≥38.6°C), infiltration score on chest X ray (<4 and ≥6), white blood cell count in peripheral blood (<10000/mm^3 and ≥15000/mm^3), and C-reactive protein (<4.0 mg/dl and 10.0 mg/dl). "Moderate" pneumonia was defined as that excluded from the categories of "mild" and "severe". When underlying diseases or complications in the patients were considered to be severe in terms of effects on the severity of pneumonia, disease status was defined as "severe", even though the pneumonia, itself, was mild or moderate. Thirty-three patients (14.2%) were classified as having mild pneumonia, 158 (68.1%) were classified as moderate, and 40 (17.2%) were classified as severe (severity in 1 patient was unknown because of insufficient laboratory data). Six patients underwent mechanical ventilation.

The leading causative agent in patients with severe pneumonia was *S. pneumoniae* (27.3%), followed by *C. pneumoniae* (12.1%), *H. influenzae* (9.1%), and influenza virus A (9.1%). Mixed infections (15.2%) were detected in five patients: *S. pneumoniae* and influenza virus A, *S. pneumoniae* and *Staphylococcus aureus*, *S. aureus* and influenza virus A; *C. pneumoniae* and *M. catarrhalis*; and *S. milleri* group and two anaerobes, respectively. On the other hand, the main agents in patients with mild pneumonia were *S. pneumoniae* (15%), *M. pneumoniae* (7.5%), and *C. pneumoniae* (7.5%) in addition to influenza virus A (17.5%).

Four patients of advanced age (79 to 99 years; 1.7%) died of pneumonia and complications (interstitial pneumonia or perforation of digestive ulcer), and the status of their pneumonias was considered severe in one and moderate in three at the time of admission. Causative agents were detected in three patients: *S. pneumoniae* and influenza virus A, *Pseudomonas aeruginosa*, and *M. catarrhalis*, respectively.

Discussion

The present study is the first multicenter investigation of the etiology of community-acquired pneumonia in Japan, enrolling all patients with pneumonia accessing hospitals. Most studies thus far on the etiology of community-acquired pneumonia have targeted only patients who required hospitalization, and the whole picture of the etiology of community-acquired pneumonia is unclear.

The protocol in the present study resulted in a high rate of identification of the causative agents, i.e., 73.3%, which was higher than that in previous studies,[6,7,22–25] but lower than that in a study from Israel,[26] which showed over 80%.

In most studies, the responsible pathogens were not defined in as many as 50% of the patients, even when several diagnostic tests were performed. This is likely a reflection of several factors, including the effect of prior antimicrobial therapy, the presence of unusual pathogens that go unrecognized, the presence of viral infection, and the presence of pathogens that are currently not identified or recognized, as has been discussed.[6,22–25] In the present study, the presence of atypical and viral infections was detected clearly by specialized techniques, which may have contributed to the higher frequency of identification of the pathogens, even when patients had received prior antimicrobial therapy; in 64.8% of these patients, the responsible pathogens were identified.

The distributions of the main etiologic groups and the five most frequent pathogens were comparable to those in several previous series. A review of published studies from North America showed that *S. pneumoniae* was consistently the leading pathogen in community-acquired pneumonia (20% to 60% of all episodes); followed by *H. influenzae*, accounting for 3% to 10% of episodes; *C. pneumoniae*, accounting for 4% to 6%; *M. pneumoniae*, accounting for 1% to 6%; *Legionella*, accounting for 2% to 8%; and viruses, accounting for 2% to 8%.[27] The frequencies of detection of *S. pneumoniae*, *C. pneumoniae*, *M. pneumoniae*, and *Legionella* in the present study were 24.6%, 6.5%, 5.2%, and 3.9%, respectively, while we found that the frequencies of *H. influenzae* and viruses at 18.5% and 16.4%, respectively) were relatively higher. *H. influenzae* was identified exclusively by sputum examination, mostly in patients with COPD or chronic bronchopulmonary diseases, especially those aged above 40 years. Because *H. influenzae* can more easily colonize these patients' airways,[28,29] the etiology of this pathogen in a pneumonia episode may be questioned. However, all patients with the infection in the present study expectorated a valid sputum sample with the finding of gram-negative short bacilli phagocytosed by neutrophils, and 30 of the 43 patients with *H. influenzae* infection had no evidence of other pathogens. Therefore, we listed it here as, at least, a probable pathogen of pneumonia.

The frequency of *Legionella* in community-acquired pneumonia in Japan has been reported as less than 1%,[6,7] which was lower than that in studies from Europe to North America, in which it ranged from 2% to 15%.[1–4,27] In the present study, the frequency was 3.9%, which was higher than that in the two previous studies in Japan. The frequency found may be due to the method of laboratory detection used including urinary antigen assay and PCR with reliable and rapid detection.[30]

One of our important results is the finding of multiple or mixed infections in 60 patients, representing 25.9% of all patients and 35.3% with a causal diagnosis. These are relatively higher percentages compared to those that in previous reports with high rates of mixed infection.[26,31] Several studies have reported that some patients with community-acquired pneumonia have mixed infections, involving bacterial agents and atypical and/or viral agents,[26,32] mainly combinations of *S. pneumoniae* and *C. pneumoniae*,[32–34] or of *S. pneumoniae* and viruses.[35,36] Such findings are similar to those in the present study (Table 3), but in our study, anaerobes were often detected with several other anaerobes, followed by the *S. milleri* group and *H. parainfluenzae*. To summarize, almost all possible combinations of causal agents have been identified in all studies reporting a high rate of mixed infections.

A unique pathogen detected in three studies from Japan, including the present one, was the *S. milleri* group, whose frequency ranged from 2.0% to 3.6%.[6,7] This group has been recently recognized as a new pathogen of community-acquired pneumonia.[6,37,38] This pneumonia is found in patients aged above 40, and easily progresses to lung abscess and/or empyema.[37,39] In the present study, all five patients with this infection were over 65 years old, and two of them had a complication with lung abscess or empyema that developed after the pneumonia.

Our study has a limitation in that it was performed in a winter season, and this seasonal factor may have led to an etiological bias. This is probably the case with the high frequency of Influenza A virus, and this high frequency may also be due to the use of several techniques for identification, including antibody titers in pared sera, the antigen detection assay, and the PCR assay. When the incidence of pathogens was compared in an epidemic versus nonepidemic year for influenza, only *S. aureus* was significantly increased.[40] The frequency of *S. aureus* (3.4%) in our study was similar to that in previous studies performed over all seasons.

In conclusion, the results of our study have important practical implications for the initial treatment of adult patients with community-acquired pneumonia in Japan. The etiology of the disease was not markedly different from that in Western countries. *S. pneumoniae* and *M. pneumoniae* were the most prevalent pathogens in younger patients, and *S. pneumoniae* and *H. influenzae* were the most prevalent in the elderly. The probability of mixed infections involving bacterial agents and atypical and/or viral agents should be considered in the initial treatment.

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Appendix

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