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ORIGINAL RESEARCH

Clinical Characteristics of *Chlamydia psittaci* Infection Diagnosed by Metagenomic Next-Generation Sequencing: A Retrospective Multi-Center Study in Fujian, China

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Objective: This study aimed to describe and compare the epidemiological, demographic, clinical, laboratory and radiological characteristics as well as the complications, treatments, and outcomes of these patients.

Methods: We retrospectively investigated clinical data of patients with *C. psittaci* infection (psittacosis) in eight Grade IIIA hospitals of Fujian. Metagenomic next-generation sequencing (mNGS) was used identify *C. psittaci* in clinical samples of all included patients.

Results: A total of 74 patients (39 severe/35 non-severe) was diagnosed with psittacosis, 25 (33.8%) of whom had history of poultry exposure. Common symptoms included high fever (98% [37/74]), fatigue (52.7% [39/74]), and dyspnea (51.4% [38/74]). Common manifestations in imaging included consolidation (89.2%), pleural effusion (77.0%), and air bronchogram (66.2%). Common complications included acute respiratory distress syndrome (55.4% [41/74]), type I respiratory failure (52.7% [39/74]), acute liver injury (41.9% [31/74]), and secondary infection (27.0% [20/74]). The in-hospital mortality rate was 8.11% (6/74).

Conclusion: C. *psittaci* infection is represents an underestimated cause of CAP. For SCAP patients with poultry and bird contact history, specimens were encouraged to be sended for mNGS test in time. *C. psittaci* infection can lead to severe, multiple system involvement, and several complications. mNGS facilitate timely diagnosis of *C. psittaci* infection.

Keywords: *Chlamydia psittaci*, metagenomic next-generation sequencing, community-acquired pneumonia, bronchoalveolar lavage fluid

Introduction

Community-acquired pneumonia (CAP) is a severe infection of the lower respiratory tract that frequently results in admission to the intensive care unit (ICU),[1] and severe CAP (SCAP) is associated with particularly high mortality rates,

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reaching 23–47%.[2] SCAP, which represents 15–28% of all CAP cases, is commonly caused by atypical pathogens, such as *Legionella pneumophila, Chlamydia pneumoniae*, or *Chlamydia psittaci*.[3] C.*psittaci* is a zoonotic intracellular bacterial pathogens that can infect a broad range of animal hosts and occasionally, humans.[4] *C. psittaci* infection in humans, also known as psittacosis, is usually believed to be an uncommon and occurs sporadically worldwide, typically presenting as CAP.[5] *C. psittaci* has been reported as the causative agent in 1.03% of all CAP cases, and 7.3–7.5% of all SCAP, constituting a public health risk.[6–9]

Disease severity ranged from mild to severe, and severe cases can present with fulminant sepsis, acute respiratory distress syndrome (ARDS), multiple organ dysfunction syndrome (MODS) and even death.[10] Psittacosis-related mortality can range from 0% to 20% among different geographic regions and clinics,[6–8] and *C. psittaci* is classified as a biothreat agent by the United States Centers for Disease Control and Prevention (CDC) due to its potentially life-threatening symptoms (https://www.cdc.gov/nndss). Since its symptoms are relatively non-specific and current testing methods are limited, psittacosis is easily under-diagnosed, misdiagnosed, or diagnosed late. Moreover, standard tests in CAP patients do not typically include *C. psittaci* infection.[11] Currently, there are culture-based methods, PCR-based assays, and various serological methods available to test for *C. psittaci* infection.[12] However, serological tests for *C. psittaci* are prone to cross-reaction with other chlamydial species, false negatives in the early acute phase, and delays between symptom onset and laboratory-based diagnosis in the interval before convalescent samples are available. These cumulative factors have subsequently hindered efforts to determine its precise incidence and prevalence, resulting in underestimation of psittacosis incidence globally.[13,14]

Metagenomic next-generation sequencing (mNGS) has emerged as an efficient, high throughput method for simultaneous identification of different pathogens, including bacteria, fungi, viruses, eukaryotic parasites, and novel pathogens in clinic, and has led to increased detection of rare and unexpected pathogens.[15] Thus, mNGS can provide a relatively rapid diagnosis to expedite antibiotic treatments and improve patient prognosis. mNGS has been used in the confirmed diagnosis of *C. psittaci* infection in recent years, which limited to a few case reports and a small case series.[16–18] mNGS seemed superior to the traditional methods in diagnosis of *C. psittaci* infection. Till now, there are no reports of CP colonization in humans. Moreover, CP is not common microbial contaminants present in the clinical sample. Positive mNGS results using specimens from sputum, blood, or BALF can serve as diagnostic criteria of *C. psittaci* infection. Furthermore, there are no detail data in China concerning on *C. psittaci* infection. The objective of this study was to describe clinical, laboratory, and radiological characteristics, treatment, and outcomes of patients with *C. psittaci* infection diagnosed by mNGS and to compare the clinical features between severe ill and non-severe patients.

Methods

Study Design and Setting

This retrospective study was performed by collecting data from medical records of confirmed *C. psittaci* infection patients referred to six hospital between April 1, 2021 and March 30, 2022. *C. psittaci* infection was confirmed by mNGS in serial samples of sputum, endotracheal aspirate (ETA), broncho-alveolar lavage fluid (BALF), or serum. The study was approved by the ethical review board of each participating clinical center. Written informed consent was waived for this retrospective study. The present study was performed in accordance with the Helsinki Declaration. Specimens were placed in a sterile sputum container, stored at −20°C, and then sent to Jieyi Genomics Institute (Hangzhou, China) for detection. The workflow of mNGS for sample collection and processing includes nucleic acid extraction, library construction, sequencing, and sequence data analysis, as previously described.[16] Severe pneumonia was diagnosed according to guidelines of the American Thoracic Society/Infectious Disease Society of America.[11] Demographics data, clinical symptoms and signs, laboratory tests, radiological characteristics, treatments, and outcomes data were retrieved.

Radiological Assessment

Two radiologists independently interpreted all chest CT scans and were blinded to the clinical information of each patient. The major CT indications were described using international standard nomenclature defined by the Fleischner Society glossary.[19]

Statistical Analysis

Categorical data was presented as counts and percentages, and continuous data were expressed as mean ± SD if the data were normally distributed, or expressed as median with interquartile range (IQR) values. Means for continuous variables were compared using independent group Students t tests when the data were normally distributed and the Mann–Whitney test in cases of non-normal distribution. Proportions for categorical variables were compared using the $\chi 2$ test. A P value less than 0.05 was considered statistically significant. All statistical analyses were done with SPSS version 20.

Results

Demographic Data of Included Patients

In total, 74 patients with *C. psittaci* infection diagnosed by mNGS were enrolled in this study. The full demographic and clinical characteristics of the patients are presented in Table 1. All cases were community acquired. Of these, 39 patients required ICU admission, and were therefore classified as severe cases. The large majority of our sample were obtained

Table 1 Demographics and Clinical Characteristics of Patients with *C. psittaci* Infection

Variables	All Patients (n=74)	Severe Group (n=39)	Non-Severe Group (n=35)	*p* value
Age (years), median (IQR)	64.5 (55.7, 74.0)	66.0 (53.0, 74.0)	62.0 (56.0, 73.0)	0.729
Men, n (%)	51 (68.9%)	27 (69.2%)	24 (68.6%)	0.951
Live in rural area, n (%)	66 (89.2%)	36 (92.3%)	30 (85.7%)	0.464
Exposure history, n (%)	34 (46.0%)	24 (61.5%)	10 (28.6%)	0.004
History of contact with avian, n (%)	9 (12.2%)	4 (10.3%)	5 (14.3%)	0.727
History of contact with poultry, n (%)	25 (33.8%)	20 (51.3%)	5 (14.3%)	0.001
Samples for mNGS				
BALF, n (%)	71 (96.0%)	37 (94.9%)	34 (97.1%)	1.000
Blood samples, n (%)	4 (5.4%)	4 (10.3%)	0 (0.0%)	0.117
Tracheal aspirate, n (%)	1 (1.4%)	0 (0.00%)	1 (2.9%)	0.473
CSF samples, n (%)	1 (1.4%)	1 (2.6%)	0 (0.0%)	1.000
No underlying diseases, n (%)	40 (54.1%)	18 (46.2%)	22 (62.9%)	0.150
Hypertension, n (%)	20 (27.0%)	12 (30.8%)	8 (22.9%)	0.444
Diabetes, n (%)	14 (18.9%)	8 (20.5%)	6 (17.1%)	0.712
Malignancy, n (%)	4 (21.6%)	1(2.6%)	3 (8.6%)	0.339
Chronic liver disease, n (%)	2 (2.7%)	1(2.6%)	1 (2.9%)	1.000
SOFA, median (IQR)	2 (1, 5)	5 (3, 7)	1 (0, 2)	<0.001
APACHE II, median (IQR)	11 (8, 14)	13.7±4.6	8.1±3.9	<0.001
CURB-65, mean (SD)	1.9±1.6	3.1±1.2	0.5±0.6	<0.0001
CRB-65, mean (SD)	1.0±1.1	1.7±1.04	0.2±0.5	<0.0001
Time from illness onset to hospital admission (days), median (IQR)	5.5 (2.7, 7)	4 (3, 7)	6 (2, 10)	0.249
Time from illness onset to confirmed diagnosis (days), median (IQR)	9.5 (8, 12.3)	9 (8, 11)	10 (8, 15)	0.064
Time from admission to confirmed diagnosis (days), median (IQR)	4 (3, 6)	4 (3, 6)	5 (3, 6)	0.155

(*Continued*)

Table 1 (Continued).

Variables	All Patients (n=74)	Severe Group (n=39)	Non-Severe Group (n=35)	*p* value
Symptoms				
Productive cough n (%)	37 (50.0%)	22 (56.4%)	15 (42.9%)	0.244
Dry cough, n (%)	28 (37.8%)	16 (41.0%)	12 (34.3%)	0.551
High Fever n (%)	64 (86.5%)	35 (89.7%)	29 (82.9%)	0.502
Moderate Fever n (%)	7 (9.5%)	4 (10.3%)	3 (8.6%)	1.000
Low fever, n (%)	3 (4.1%)	0 (0.0%)	3 (8.6%)	0.101
Dyspnea n(%)	38 (51.4%)	31 (79.5%)	7 (20.0%)	<0.0001
Hemoptysis n (%)	5 (6.8%)	3 (7.7%)	2 (5.7%)	1.000
Chill, n (%)	18 (24.3%)	10 (25.6%)	8 (22.9%)	0.780
Myalgia, n (%)	8 (10.8%)	5 (12.8%)	3 (8.6%)	0.714
Chest pain, n (%)	21 (28.4%)	13 (33.3%)	8 (22.9%)	0.318
Fatigue, n (%)	39 (52.7%)	31 (79.5%)	8 (22.9%)	<0.0001
Headache, n (%)	10 (13.5%)	6 (15.4%)	4 (11.4%)	0.740
Delirium, n (%)	15 (20.3%)	13 (33.3%)	2 (5.7%)	0.003
Nausea, n (%)	4 (5.4%)	3 (7.7%)	1 (2.9%)	0.617
Vomit, n (%)	3 (4.1%)	3 (7.7%)	0 (0.0%)	0.242
Abdominal pain, n (%)	4 (5.4%)	3 (7.7%)	1 (2.9%)	0.617
Diarrhea, n (%)	5 (6.8%)	3 (7.7%)	2 (5.7%)	1.000
Pharyngalgia, n (%)	4 (5.4%)	2 (5.1%)	2 (5.7%)	1.000
Signs				
Relative bradycardia	38 (51.4%)	22 (56.4%)	16 (45.7%)	0.358
Moist rale, n (%)	47 (63.5%)	30 (76.9%)	17 (48.6%)	0.011
Dry rale, n (%)	8 (10.8%)	3 (7.7%)	5 (14.3%)	0.464

from the respiratory system, with 71 (96.0%) from BALF, four from serum (5.4%), and three from cerebrospinal fluid (CSF, 8.4%). The mean turnaround time for mNGS ranged from 24 to 48 h after receipt by sequencing facility.

The median age of these patients (51 males and 23 females) was 64.5 (55.7, 74.0) years. Sex distribution and age did not differ significantly between the severe and non-severe groups ($p = 0.951$ and $p = 0.729$, respectively). Forty (54.1%) had coexisting conditions, including hypertension (27.0%), diabetes (18.9%), and malignancy (5.4%). Underlying diseases did not significantly differ between the two groups ($p = 0.150$). There was a distinct seasonality to the cases with 33/74 (44.6%) presenting in the 3-month period from October to December (Figure 1). The monthly distribution of onset peaked in November and December, with relatively few cases occurring in warmer months (Figure 1).

Sixty-six (89.2%) patients lived in rural areas, and 46.0% of the patients had a history of environmental exposure. Severe patients had more contact history than non-severe patients (61.5% vs 28.6%, $p = 0.004$). History of avian exposure was similar between the two severity groups (10.3% vs.14.3%, $p = 0.727$). Seven patients worked as either poultry slaughterers or breeders. Ten patients had slaughtered live poultry for cooking. Eleven patients had raised ducks or chickens at home privately. Nine patients had raised birds (pet parrots [n = 4], pigeon [n = 3], *Garrulax canorus* [n = 1], peacock [n = 1]).

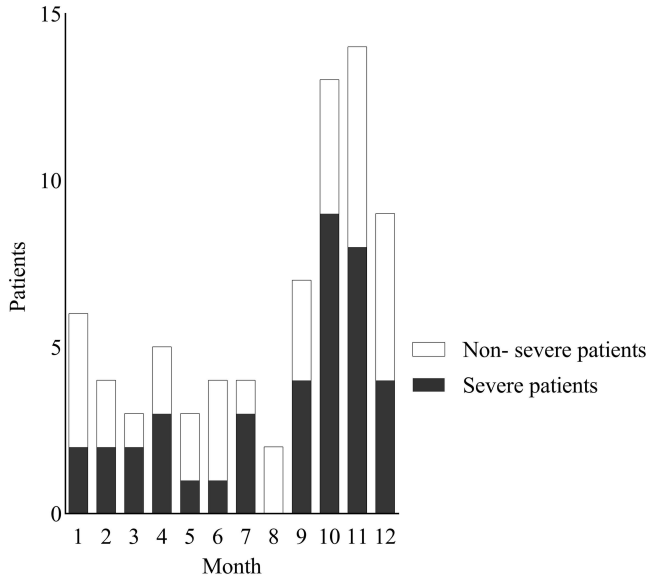

Figure 1 Cumulative hospital admissions of psittacosis per month.

Disease Severity

Duration from onset of symptoms to admission ranged from 1 to 15 days, with a median of 5.5 days. The median time from hospitalization to confirmed diagnosis was 4.0 days. The duration from onset of symptoms to confirmed diagnosis and hospitalization to confirmed diagnosis was similar (9.0 days vs 10.0 days, $p = 0.064$, and 4.0 days vs 5.0 days, $p = 0.155$). Severe patients had higher CURB65, sepsis-related organ failure assessment (SOFA), and APACHE II than non-severe patients (all $p<0.05$).

Clinical Symptoms and Signs

The most common symptoms on admission were high fever (64/74 [86.5%]), productive cough (50.00% [37/74]) and dyspnea (51.4% [38/74]); less common symptoms included dry cough (11/74 [28%]), headache (13.5% [10/74]), haemoptysis (6.8% [5/74]), vomit (4.1% [3/74]), and diarrhea (6.8% [5/74]) (Table 1). Compared with non-severe patients, severe patients were more likely to report dyspnea (79.5% [31/39] vs 20.0% [7/35] in severe vs non-severe respectively; $p<0.0001$), and delirium (13/39 [33.3%] vs 2/35 [5.7%]; severe vs non-severe respectively; $p = 0.003$) at presentation. Relative bradycardia was found in 38 patients (51.4% [38/74]). Relative bradycardia and dry rale did not differ between groups (all $p>0.05$).

Laboratory Testing

Laboratory findings between the two groups were shown in Table 2. The levels of creatinine, urea nitrogen, procalcitonin, D-dimer on admission were significantly higher in the severe group compared with the non-severe group (all $p<0.05$), while lymphocyte, platelet counts, and albumin were all lower upon admission in the severe group than in the non-severe group (all $p<0.05$). Other laboratory parameters, including white blood cell counts, neutrophil counts, and lactate dehydrogenase levels were not significantly different between groups (all $p>0.05$). Lymphocytopenia was identified in around 60% of the total cohort and in 81.5% of the severe patients, and 29.1% of all patients had thrombocytopenia. Lymphocytopenia, proteinuria and hematuresis were more common in the severe group compared with the non-severe group (all $p<0.05$).

CT Findings on Admission

On admission, abnormalities in chest CT images were detected among all patients (Table 2). In the full cohort, 64.9% (48/74) of patients presented with bilateral lung involvement while 74.3% (55/74) presented with multi-lobe involvement. Multiple lobular involvement was more common in severe patients than in non-severe patients (84.6% [33/39] vs 62.9% [22/35], $p = 0.032$). Subpleural lesions were more common than central lung lesions (63.5% vs 12.2%). The most

Table 2 Laboratory Findings and Chest CT Findings Upon Hospital Admission with *C. psittaci* Infection

Variables	Total (n=74)	Severe Group (n=39)	Non-Severe Group (n=35)	*p* value
White blood cell count (×109/L), median (IQR)	8.3 (5.9, 11.2)	9.1 (6.6, 11.2)	7.7 (5.8, 11.4)	0.530
Neutrophil count (×109/L), median (IQR)	7.1 (4.6, 9.9)	7.8 (5.1, 9.9)	6.6 (4.3, 9.8)	0.148
Lymphocyte count (×109/L), median (IQR)	0.6 (0.4,1.0)	0.5 (0.3, 0.7)	0.8 (0.5, 1.1)	<0.001
Lymphopenia, n (%)	54 (73.0%)	35 (89.7%)	19 (54.3%)	0.001
Platelet count (×109/L), median (IQR)	188.0 (123.8, 252.3)	143.0 (99.0, 240.0)	212.0 (172.0, 256.0)	0.013
Thrombocytopenia, n (%)	12 (16.2%)	10 (25.6%)	2 (5.7%)	0.020
C-reactive protein (CRP, g/L), mean (SD)	180.6±79.2	207.2±74.8	150.9±74.1	0.002
Procalcitonin (ng/mL), median (IQR)	0.8 (0.3, 2.0)	1.6 (0.7, 9.2)	0.36 (0.2, 1.0)	<0.001
Albumin (ALB, g/L), median (IQR)	29.7 (26.3, 33.8)	28.2 (25.0, 30.0)	32.4 (29.6, 36.6)	<0.001
Creatine kinase (CK, U/L), median (IQR)	256.0 (85.0, 831.8)	567.0 (165.0, 1388.0)	107.0 (61.0, 275.0)	<0.001
Lactate dehydrogenase (LDH, U/L), median (IQR)	434.5 (266.3, 685.0)	463.0 (293.0, 879.0)	351.0 (235.0, 551.0)	0.086
D-dimer (μg/L), median (IQR)	1.1 (0.5, 2.7)	1.3 (0.7, 5.0)	0.8 (0.4, 1.7)	0.024
Proteinuria, n (%)	48 (64.9%)	31 (79.5%)	17 (48.6%)	0.005
Hematuresis, n (%)	34 (46.0%)	23 (59.0%)	11 (31.4%)	0.018
Chest CT findings				
Bilateral lungs involved, n (%)	48 (64.9%)	29 (74.4%)	19 (54.3%)	0.071
Multiple lobular involved, n (%)	55 (74.3%)	33 (84.6%)	22 (62.9%)	0.032
Consolidation, n (%)	66 (89.2%)	36 (92.3%)	30 (85.7%)	0.464
Ground-glass opacity, n (%)	15 (20.3%)	7 (18.0%)	8 (22.9%)	0.600
Air bronchogram, n (%)	49 (66.2%)	29 (74.4%)	20 (85.7%)	0.118
Pleural effusion, n (%)	57 (77.0%)	32 (82.1%)	25 (71.4%)	0.278
Bilateral pleural effusion, n (%)	32 (43.2%)	22 (56.4%)	10 (28.6%)	0.016
Pericardial effusion, n (%)	5 (6.8%)	3 (7.7%)	2 (5.7%)	1.000
Lesions close to the pleura, n(%)	47 (63.5%)	25 (64.1%)	22 (62.9%)	0.912
Central location, n (%)	9 (12.2%)	3 (7.7%)	6 (17.1%)	0.292
Random distribution, n (%)	18 (24.3%)	11 (28.2%)	7 (20.0%)	0.411

common findings of chest CT images were consolidation (89.2% [66/74]), pleural effusion (77.0% [57/74]), and air bronchogram (66.2% [49/74]) (Figures 2 and 3). In contrast, ground-glass opacity (GGO, 20.2% [15/74]) and pericardial effusion (6.8% [5/74]) were less common in CT imaging of the full cohort. Bilateral pleural effusion was more common in severe patients than in non-severe patients (56.4 [22/39]% vs 28.6% [10/35]; $p = 0.016$).

Complications

During the hospital stay, 81.2% (69/74) of patients presented with common complications involving multiple vital organs, including ARDS (55.4% [41/74]), followed by type I respiratory failure (52.7% [39/74]), acute liver injury

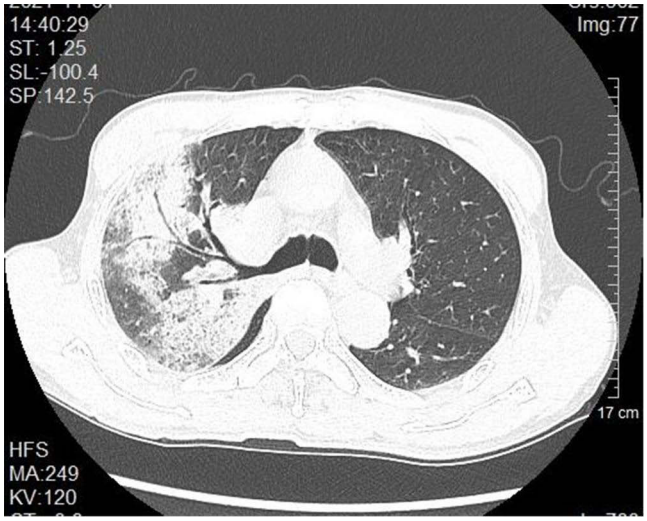

Figure 2 Chest imaging of a 67-yr-old man with C. *psittaci* infection show multiple consolidations in the right upper and lower lobe, with air bronchogram in the upper lobe.

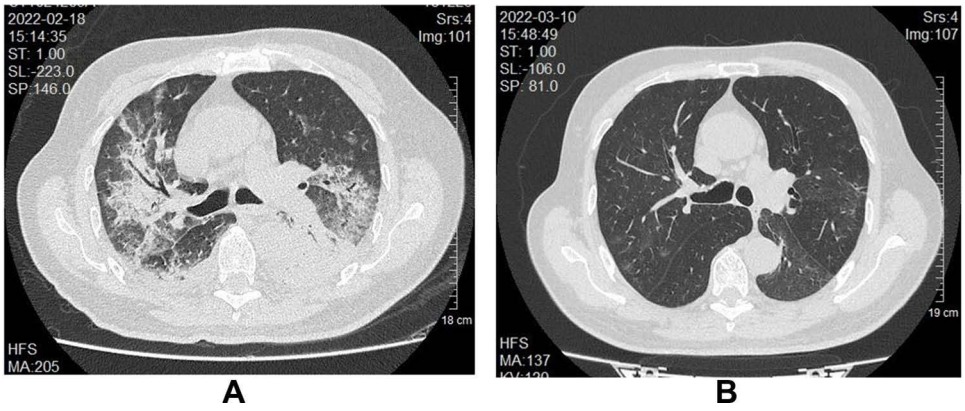

Figure 3 Chest imaging of a 78-yr-old man with C. psittaci infection before and after treatment. (**A**) Pre-treatment computed tomography (CT) scan showing bilateral large consolidation with air bronchogram in right lung and bilateral pleural effusion. (**B**) Follow-up CT scan after combination therapy for 20 days showing the consolidation and pleural effusion disappeared.

(41.9% [31/74]), secondary infection (27.03% [20/74]), anemia (25.7% [19/74]), acute myocardial injury (18/74 [24.3%]), and acute heart failure (21.6% [16/74]) (Table 3). The less common complications included acute or chronic heart failure (6.76% [5/74]), rhabdomyolysis (6.76% [5/74]), diffuse intravascular coagulation (6.76% [5/74]), and gastrointestinal bleeding (4.05% [3/74]). 16.2% (12/74), 18.9% (14/74). The median time from admission to ARDS in severe patients was 24.0 (7.5, 42.0) hours. 24.3% [18/74] of all patients progressed to multi-organ dysfunction (MODS). Time from admission to MODS was 36.0 (24.0, 48.0) hours.

Treatments

Most patients were treated with doxycycline (73.0%) or moxifloxacin (73.0%). Forty-three patients (58.1%) did not receive any initial targeted treatment with activity against *C. psittaci*. Compared with the severe group, more patients received broad-spectrum antibiotic treatment than in the non-severe group (61.5% vs 34.3%; p=0.019). More severe patients than non-severe patients received combination therapy (64.1% vs 37.1%; p = 0.035). 31.1% patients (23/74) received high-flow nasal cannula therapy and 39.2% (29/74) received non-invasive ventilation. Compared with non-severe patients, severe patients were more likely to receive mechanical ventilation, either invasively or non-invasively. Seventeen of the severe patients (43.6%) received invasive mechanical ventilation, one of whom received extracorporeal membrane oxygenation (ECMO) as a rescue therapy, while six died during the ICU stay. The duration of invasive

Table 3 Complications, Treatments and Outcomes of Patients with *C. Psittaci* Infection

Complications	Total (n=74)	Severe Group (n=39)	Non-Severe Group (n=35)	*p* value
Acute respiratory distress syndrome, n (%)	41 (55.4%)	36 (92.3%)	5 (14.3%)	<0.0001
Mild acute respiratory distress syndrome, n (%)	12 (16.2%)	8 (20.51%)	4 (11.4%)	0.290
Moderate acute respiratory distress syndrome, n (%)	14 (18.9%)	13 (33.3%)	1 (2.9%)	0.001
Severe acute respiratory distress syndrome, n (%)	15 (20.3%)	15 (38.5%)	0 (0.0%)	<0.0001
Time from admission to ARDS (hours), median (IQR)	24.0 (12.5, 48.0)	24.0 (7.5, 42.0)	36 (24.0, 66.0)	0.148
Type I respiratory failure, n (%)	39 (52.7%)	33 (84.6%)	6 (17.1%)	<0.0001
Type II respiratory failure, n (%)	3 (4.1%)	3 (7.7%)	0 (0.0%)	0.242
Acute cardiac injury, n (%)	18 (24.3%)	17 (43.6%)	1 (2.9%)	<0.0001
Acute heart failure, n (%)	16 (21.6%)	14 (35.9%)	2 (5.7%)	0.002
Acute on chronic heart failure, n (%)	5 (6.8%)	3 (7.7%)	2 (5.7%)	1.000
Acute kidney injury (AKI), n (%)	13 (17.6%)	12 (30.8%)	1 (2.9%)	0.002
Anemia, n (%)	19 (25.7%)	14 (35.9%)	5 (14.3%)	0.034
Rhabdomyolysis, n (%)	5 (6.8%)	5 (12.8%)	0 (0.0%)	0.056
Disseminated intravascular coagulation, n (%)	5 (6.8%)	5 (12.8%)	0 (0.0%)	0.056
Gastrointestinal bleeding, n (%)	3 (4.1%)	2 (%)	0 (0.0%)	0.495
Acute liver injury, n (%)	31 (41.9%)	20 (51.3%)	11 (31.4%)	0.084
Secondary infection, n (%)	20 (27.0%)	17 (%)	3 (8.6%)	<0.0001
Septic Shock, n (%)	12 (16.2%)	12 (%)	0 (0.0%)	<0.0001
Multiple organ dysfunction syndrome (MODS), n (%)	18 (24.3%)	18 (46.2%)	0 (0.0%)	<0.0001
Time from admission onset to MODS (hours), median (IQR)	36.0 (24.0, 48.0)	36.0 (24.0, 48.0)	–	–
Treatment				
Doxycycline, n (%)	54 (73.0%)	28 (71.8%)	26 (74.3%)	0.810
Azithromycin, n (%)	7 (9.5%)	5 (12.8%)	2 (5.7%)	0.435
Moxifloxacin, n (%)	54 (73.0%)	30 (76.9%)	24 (68.6%)	0.419
Combination therapy, n (%)	38 (51.4%)	25 (64.1%)	13 (37.1%)	0.021
Empirical antibiotic coverage for atypical pathogens, n (%)	43 (58.1%)	19 (48.7%)	24 (68.6%)	0.084
Using broad-spectrum antibiotics, n (%)	36 (48.6%)	24 (61.5%)	12 (34.3%)	0.019
Glucocorticoid therapy, n (%)	20 (27.0%)	14 (35.9%)	6 (17.1%)	0.070
Fluid resuscitation, n (%)	10 (13.5%)	10 (25.4%)	0 (0.0%)	0.001
Vasoactive drugs, n (%)	11 (14.9%)	11 (28.2%)	0 (0.0%)	0.001
Respiratory Support at highest level				
Nasal cannula oxygen therapy, n (%)	10 (13.5%)	0 (%)	10 (28.6%)	<0.0001
Mask oxygen therapy, n (%)	14 (18.9%)	2 (5.1%)	12 (34.3%)	0.001
Non-invasive ventilation, n (%)	18 (24.3%)	10 (25.6%)	8 (22.9%)	0.780

(*Continued*)

Table 3 (Continued).

Complications	Total (n=74)	Severe Group (n=39)	Non-Severe Group (n=35)	*p* value
High-flow nasal cannula, n (%)	16 (21.6%)	11 (28.2%)	5 (14.3%)	0.146
Invasive mechanical ventilation, n (%)	16 (21.6%)	16 (41.0%)	0 (0%)	<0.0001
Duration of invasive mechanical ventilation (hours)	212.0 (166.5, 240.0)	212.0 (166.5, 240.0)	–	–
ECMO, n (%)	1 (1.4%)	1 (2.6%)	0 (0.0%)	1.000
Initial respiratory support failure (%)				
NIV failure, n /total (%)	10/29 (%)	9 (43.4%)	1 (10%)	0.015
HFNC failure, n/total (%)	8/23 (%)	8 (44.4%)	0 (0.0%)	0.006
IMV failure, n /total (%)	6/17 (%)	6 (35.3%)	0 (0.0%)	0.026
Continuous renal replacement therapy, n (%)	4 (5.4%)	4 (10.3%)	0 (0.0%)	0.117
Outcome				
Death, n (%)	6 (8.1%)	6 (15.4%)	0 (0.0%)	0.026
ICU length of stay (days), median (IQR)	7.0 (0.0, 10.0)	7.0 (0.0, 10.0)	–	–
Hospitalization duration (days), median (IQR)	14.0 (10.0, 18.3)	17.0 (11.0, 21.0)	13.0 (9.0, 14.0)	0.001

mechanical ventilation in severe patients was 212.0 (166.5, 240.0) hours. Four patients received CRRT as salvage therapy.

Outcomes

The in-hospital mortality of patients was 8.11% (6/74) (Table 3). No patients in the non-severe group died during their hospital stay. The average length of hospital stay for severe patients was significantly longer than for non-severe patients (17 days vs 13 days, $p = 0.001$). The average length of ICU stay for severe patients was 7.0 (0.0, 10.0) days.

Discussion

To our knowledge, the present study represents the largest case series in China to date describing the demographic, clinical, laboratory, and radiological characteristics as well as the complications, treatments, and outcomes of these patients with *C. psittaci* infection. We found that *C. psittaci* infection does not appear to be rare in China. *C. psittaci* infection is an important emerging etiology of SCAP, but is not well described in epidemiologic studies of CAP. Our study also compared clinical characteristics between severe and non-severe patients to facilitate accurate diagnosis and treatment.

C. psittaci has been reported in relatively few case studies of CAP. Dumke et al developed a real-time qPCR assay to specifically detect members of family Chlamydiaceae in pharyngeal swabs of CAP patients, and found *C. psittaci* in 1.4% of CAP cases.[20] Sun and co-workers used mNGS to detect etiologies of SCAP in Beijing, China and reported that *C. psittaci* is responsible for ~9.0% (4 of 44) of SCAP.[8] Similarly, Wu and colleagues et al found a prevalence of 7.3% (24 of 329) *C. psittaci* infection in mNGS-based diagnosis of BALF samples in a multicenter, prospective study of 329 adult SCAP patients.[9] A nationwide, multicenter study of SCAP etiology in China that employed a combination of traditional culture-based assays, antigen tests, PCR assays, and mNGS diagnostics revealed that *C. psittaci* infection contributed to 6.8% (15/222) of the SCAP cases.[10] mNGS can facilitate *C. psittaci* detection in the relatively easily acquired BALF samples of CAP patients.[9,10] In this study, Psittacosis was detected in serum samples of three patients in the severe group, presumably due to high bacterial load, though possibly attributable to detection of circulating bacterial

DNA fragments. Other studies reported *C. psittaci* in stool and CSF specimens detected by PCR, suggesting that these specimens could also facilitate mNGS-based diagnosis.[13]

Most cases of the reported *C. psittaci* infection were from southern China.[10] Behavioural and environmental factors, such as altitude, temperature, and developed poultry breeding industry contribute to the higher prevalence of psittacosis in southern China than that in northern China, and thus warrant further study. Fujian province has a population of 39.41 million, and the number of cases reported from this province annually is higher than the total number reported from several countries, including Japan and the United State.[13,14] However, it remains unknown whether *C. psittaci* infection is endemic to Fujian or if a high genetic susceptibility among Han Chinese contributes to this rate. The epidemiology of *C. psittaci* infection in our study follows seasonal clustering and more than three quarters of the patients came from rural areas. It is speculated that the temperature is related to the pathogen growth. It should also be noted that males comprised a predominant proportion of these cases. Although the reason is unclear why psittacosis patients in this study were more likely to be older, it is reasonable to speculate that older people have a greater likelihood of entering close contact with poultry in rural China. *C. psittaci* can be found in many species of poultry or other birds, sheep, and horses; and in the United States and Japan, domesticated psittacine birds (*eg*, parrots and cockatoos) have been identified as the most common source of psittacosis.[21] *C. psittaci* typically enters hosts through the respiratory tract via inhalation of aerosolised bacteria in bird secretions, droppings, or feathers.[4] These studies suggest that the live poultry in markets and small breeding operations prevalent throughout rural China likely serve as the largest reservoirs for psittacosis. Thus, we regarded contact with poultry as the greatest risk factor for *C. psittaci* infection in our study, and especially in severe cases. Aggressive intervention to limit further animal-to-person transmission in live poultry markets warrants careful consideration. Many of the cases in previous reports involved a history of avian contact, particularly with psittacines and racing pigeons. Diagnosis and treatment of SCAP should include a thorough enquiry into the patients poultry and avian exposure history.

Notably, a greater proportion of the psittacosis patient cohort in our study was identified as severe, which aligned well with previous studies.[13,17] Indeed, another study from China reported mortality rates as high as 20% among adult patients with severe *C. psittaci* infection.[10] Disparities in fatality rates among studies are potentially attributable to ethnicity, pathogen genotype, and pathogen exposure level. We found that abrupt onset of symptoms was frequent in this study cohort, with fever representing the most common symptom, followed by dyspnea, and cough, which aligned with other reports.[17,18] Some patients displayed non-specific flu-like symptom, including pharyngeal congestion or sore throat. Dyspnea was significantly higher in the severe group than in the non-severe group, which is likely a manifestation of impaired cardiopulmonary function. Unlike typical bacterial pneumonia in which the clinical findings are confined to the lungs, multi-system symptoms involving gastrointestinal and neurological presentations were not uncommon in this series. Psittacosis is often mistaken for gastroenteritis or meningitis in the absence of respiratory symptoms, and without exposure history. In addition, no association has yet been established between symptoms of *C. psittaci* infection and pathogen genotype. Various genotypes are known to exhibit differences in host preference and virulence characteristics, which can overlap, thereby confounding identification. In this cohort, lobar consolidation accompanied by air bronchogram in chest CT scans was the main finding, which agreed well with findings by Qu and Su et al.[10,17] Complications were frequent among severe patients in this study. The pathophysiology leading to multi-organ injury in *C. psittaci* infection is poorly understood. ARDS was the most common complication among severe cases. *C. psittaci* infects pulmonary epithelial cells where it then proliferates, leading to alveolar and endothelial injury.[22,23] Damage to the alveolar-capillary barrier, inflammation, and the accumulation of detritus and protein-rich fluid in alveolae can cause ARDS. Compromising the alveolar-capillary membrane can allow pathogen into the bloodstream, resulting in sepsis.[22,23] In severe cases, *C. psittaci* has been shown to infect the kidneys, liver, and central nervous system,[19,24] and the detection of *C. psittaci* DNA in CSF suggested that this pathogen could directly invade the central nervous system,[13] indicating encephalitis. Our series recorded one case *C. psittaci* in cerebrospinal fluid. It is possible that the acute kidney injury observed in our study may have been related to direct effects of *C. psittaci*, hypoxia, and shock. Cardiac injury has been associated with other respiratory diseases such as avian flu and COVID-19, and is potentially lethal if it develops into fulminant myocarditis. We observed cardiac injury in almost 70% of severe patients in our study, in sharp contrast with previous studies that reported relatively few cases of *C. psittaci* endocarditis and myocarditis. The pathogenesis of

cardiac injury and exacerbated congestive heart failure associated with *C. psittaci* infection remains unclear. Co-infection was common in our study, especially in severe cases, and could possibly reflect impaired immunity and barrier function, and significant lymphocyte reduction, although this phenomenon has not been described in other studies.

CAP management guidelines suggest that severe patients should be treated with a combination therapy that includes quinolone or on admission, and severe cases in this study were given the most effective antibiotic based on clinical suspicion without waiting for laboratory confirmation. However, empirical treatment with doxycycline, macrolide, and quinolone antibiotics, which covers this rare, should be considered for an avian or poultry source of infection indicated by contact history. Given the high mortality of psittacosis, we suggest that severe patients receive double or triple drug combination therapies. There is a prominent lack of data concerning drug resistance among *Chlamydiae* species, and randomized, controlled clinical trials could validate or substantially improve interventions for severe patients. However, no significant conclusions can be drawn from this study on the efficacy of particular therapeutic interventions. Future work should also explore the use of corticosteroid therapies in severe cases, in light of the role of inflammation in exacerbating pulmonary dysfunction. Corticosteroids were shown to reduce mortality for severe COVID-19 patients, but should be administered cautiously in treatment of severe *C. psittaci* infection due to potential side effects.

Some limitations of the study are listed below: First, data were collected retrospectively and recall bias could have influenced data. Because there are many confounding factors due to retrospective study conducted in different hospitals, we did not perform risk factor analysis in current study. Second, clinical follow-up data were not obtained from patients after discharge, and future work will assess the long-term effects or outcomes of *C. psittaci* infection. Third, the institutions included in this study are major hospitals in Fujian may have introduced bias in group selection, so caution should be exercised in the interpretation of our results between severe and non-severe cases possibly related to selection bias. Fourth, verification with serological tests and/or cultures should be included in future work to support the mNGS-based diagnoses.

Conclusion

Our study provides physicians with a better understanding of the demographic, clinical, laboratory and radiological characteristics of *C. psittaci* infection, as well as possible complications, treatment strategies, and outcomes of patients with severe infection. These findings are especially relevant for clinicians in China, where the prevalence appears higher than that reported in other countries. Clinicians should be aware of such disease entity in CAP. Early detection of *C. psittaci* infection is critical to informing clinical interventions and appropriately targeted antibiotic. We also propose mNGS as an effective and quick diagnostic method that can improve accuracy and reduce delays in identifying psittacosis. Further study of the pathogenesis of psittacosis will improve its management, epidemic control, and pandemic preparedness.

Data Sharing Statement

The datasets generated and analyzed during the current study are available from the corresponding author upon reasonable request.

Ethics Statement

The ethics committees of the First Affiliated Hospital of Fujian Medical University, Mindong Hospital, Fuqing General Hospital, Fuzhou First Hospital, Zhangzhou Affiliated Hospital of Fujian Medical University and LongYan First Hospital jointly approved the study. Because it is a retrospective study, the ethics Committees approved it without informed consent. All research data are anonymous. This study follows the Helsinki Declaration.

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Disclosure

The authors report no potential conflicts of interest in this work.

References

1. Torres A, Chalmers JD, Dela Cruz CS, et al. Challenges in severe community-acquired pneumonia: a point-of-view review. *Intensive Care Med*. 2019;45(2):159–171. doi:10.1007/s00134-019-05519-y
2. Cilloniz C, Dominedo C, Garcia-Vidal C, Torres A. Community-acquired pneumonia as an emergency condition. *Curr Opin Crit Care*. 2018;24(6):531–539. doi:10.1097/MCC.0000000000000550
3. Cillóniz C, Torres A, Niederman M, et al. Community-acquired pneumonia related to intracellular pathogens. *Intensive Care Med*. 2016;42(9):1374–1386. doi:10.1007/s00134-016-4394-4
4. Radomski N, Einenkel R, Müller A. Chlamydia-host cell interaction not only from a bird's eye view: some lessons from Chlamydia psittaci. *FEBS Lett*. 2016;590(21):3920–3940. doi:10.1002/1873-3468.12295
5. Beeckman DS, Vanrompay DC. Zoonotic Chlamydophila psittaci infections from a clinical perspective. *Clin Microbiol Infect*. 2009;15:11–17. doi:10.1111/j.1469-0691.2008.02669.x
6. Hogerwerf L, Gier DE, Baan B. Chlamydia psittaci (psittacosis) as a cause of community-acquired pneumonia: a systematic review and meta-analysis. *Epidemiol Infect*. 2017;145(15):3096–3105. doi:10.1017/S0950268817002060
7. Spoorenberg SM, Bos WJ, van Hannen EJ, et al. Chlamydia psittaci: a relevant cause of community-acquired pneumonia in two Dutch hospitals. *Neth J Med*. 2016;74(2):75–81.
8. Sun T, Wu X, Cai Y, et al. Metagenomic next-generation sequencing for pathogenic diagnosis and antibiotic management of severe community-acquired pneumonia in immunocompromised adults. *Front Cell Infect Microbiol*. 2021;11:661589. doi:10.3389/fcimb.2021.661589
9. Wu X, Li Y, Zhang M, et al. Etiology of severe community-acquired pneumonia in adults based on metagenomic next-generation sequencing: a prospective multicenter study. *Infect Dis Ther*. 2020;9(4):1003–1015. doi:10.1007/s40121-020-00353-y
10. Qu J, Zhang J, Chen Y, et al. Aetiology of severe community acquired pneumonia in adults identified by combined detection methods: a multi-centre prospective study in China. *Emerg Microbes Infect*. 2022;11(1):556–566. doi:10.1080/22221751.2022.2035194
11. Metlay JP, Waterer GW, Long AC, et al. Diagnosis and Treatment of Adults with Community-acquired Pneumonia. An Official Clinical Practice Guideline of the American Thoracic Society and Infectious Diseases Society of America. *Am J Respir Crit Care Med*. 2019;200(7):e45–e67. doi:10.1164/rccm.201908-1581ST
12. Nieuwenhuizen AA, Dijkstra F, Notermans DW, van der Hoek W. Laboratory methods for case finding in human psittacosis outbreaks: a systematic review. *BMC Infect Dis*. 2018;18(1):442. doi:10.1186/s12879-018-3317-0
13. McGovern OL, Kobayashi M, Shaw KA, et al. Use of real-time PCR for Chlamydia psittaci detection in human specimens during an outbreak of Psittacosis - Georgia and Virginia, 2018. *MMWR Morb Mortal Wkly Rep*. 2021;70(14):505–509. doi:10.15585/mmwr.mm7014a1
14. Kozuki E, Arima Y, Matsui T, et al. Human psittacosis in Japan: notification trends and differences in infection source and age distribution by gender, 2007 to 2016. *Ann Epidemiol*. 2020;44:60–63. doi:10.1016/j.annepidem.2020.03.001
15. Simner PJ, Miller S, Carroll KC. Understanding the promises and hurdles of metagenomic next-generation sequencing as a diagnostic tool for infectious diseases. *Clin Infect Dis*. 2018;66(5):778–788. doi:10.1093/cid/cix881
16. Fischer N, Rohde H, Indenbirken D, et al. Rapid metagenomic diagnostics for suspected outbreak of severe pneumonia. *Emerg Infect Dis*. 2014;20(6):1072–1075. doi:10.3201/eid2006.131526
17. Zhang Z, Zhou H, Cao H, et al. Human-to-human transmission of Chlamydia psittaci in China, 2020: an epidemiological and aetiological investigation. *Lancet Microbe*. 2022;3(7):e512–e520. doi:10.1016/S2666-5247(22)00064-7
18. Su S, Su X, Zhou L, et al. Severe Chlamydia psittaci pneumonia: clinical characteristics and risk factors. *Ann Palliat Med*. 2021;10(7):8051–8060. doi:10.1080/22221751.2021.1948358
19. Zhang H, Zhan D, Chen D, et al. Next-generation sequencing diagnosis of severe pneumonia from fulminant psittacosis with multiple organ failure: a case report and literature review. *Ann Transl Med*. 2020;8(6):401. doi:10.21037/atm.2020.03.17
20. Dumke R, Schnee C, Pletz MW, et al. Mycoplasma pneumoniae and Chlamydia spp. infection in community-acquired pneumonia, Germany, 2011-2012. *Emerg Infect Dis*. 2015;21(3):426–434. doi:10.3201/eid2103.140927
21. Balsamo G, Maxted AM, Midla JW, et al. Compendium of measures to control Chlamydia psittaci infection among humans (Psittacosis) and pet birds (Avian Chlamydiosis), 2017. *J Avian Med Surg*. 2017;31(3):262–282. doi:10.1647/217-265
22. Branley JM, Weston KM, England J, Dwyer DE, Sorrell TC. Clinical features of endemic community-acquired psittacosis. *New Microbes New Infect*. 2014;2(1):7–12. doi:10.1002/2052-2975.29
23. Knittler MR, Berndt A, Böcker S, Jiang Y. Chlamydia psittaci: new insights into genomic diversity, clinical pathology, host-pathogen interaction and anti-bacterial immunity. *Int J Med Microbiol*. 2014;304(7):877–893. doi:10.1016/j.ijmm.2014
24. Wu HH, Feng LF, Fang SY. Application of metagenomic next-generation sequencing in the diagnosis of severe pneumonia caused by Chlamydia psittaci. *BMC Pulm Med*. 2021;21(1):300. doi:10.1186/s12890-021-01673-6