Atypical Pneumonia
Updates on *Legionella*, *Chlamydophila*, and *Mycoplasma* Pneumonia

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KEYWORDS

• Community-acquired pneumonia (CAP) • Walking pneumonia • *Legionella* • Legionnaires' disease • Pontiac fever • *Chlamydophila* • *Mycoplasma*

KEY POINTS

- The clinical diagnosis of atypical pneumonia remains elusive but recent advances in rapid diagnostic platforms show promise of earlier identification of the infectious organism.
- Macrolides and respiratory fluoroquinolones remain the antibiotics of choice for atypical pneumonia but there are several new antibiotics currently under development or clinical trials.
- Both *Chlamydophila* and *Mycoplasma* have been associated with chronic diseases, but *Legionella* seems to occur sporadically and is not associated with chronic diseases.

INTRODUCTION

Pneumonia is a common cause of hospital admission and mortality and is categorized based on the clinical context in which a patient develops symptoms of infection. These categories include community-acquired pneumonia (CAP), CAP with risk factors of resistant organisms, hospital-acquired pneumonia, and ventilator-associated events. CAP is defined as contracting pneumonia with minimal or no recent contact with the healthcare system CAP is one of the most common infectious diseases and is caused by various infectious pathogens, including viruses, typical bacteria, and atypical pathogens. This article reviews the clinical considerations of atypical causes of CAP that include *Legionella*, *Mycoplasma*, and *Chlamydophila* and discusses current controversies surrounding the diagnosis and treatment of atypical CAP.

LEGIONELLA PNEUMOPHILA
Clinical Presentation

Legionella infections are manifested mainly in 2 forms:

1. Legionnaires' disease, which is a severe form of pneumonia due to infection with *Legionella*. Legionnaires' disease can manifest as a multisystem disease most commonly involving the lungs and gastrointestinal tract and is associated with significant mortality.[1]
2. Pontiac fever, which is a mild and self-resolving flu-like disease. The characteristics of Pontiac fever are mild fever, chills, myalgia, and

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Clin Chest Med 38 (2017) 45–58
http://dx.doi.org/10.1016/j.ccm.2016.11.011
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headache that lasts 2 to 5 days and often resolves itself without significant mortality.[2]

Legionella mostly affects people above 50 years of age but cases have been reported in infants and neonates.[3] Legionnaires' disease is hard to distinguish from pneumonia caused by other pathogens because it presents similar clinical symptoms; however, presence of diarrhea and elevated creatinine kinase levels can be indicators of infection by *Legionella*.[4] Pneumonia due to *Legionella* is usually found in clusters that are not associated with person-to-person transmissions but is related to exposure to the same source of infection. Most of the *Legionella* infections are acquired by contaminated water or soil. Rainfall, high humidity, and work in gardens with compost are risk factors for acquiring *Legionella* disease.[5–7] Most of the cases of legionnaires' disease are associated with *Legionella pneumophila*, but many other bacterial species have been found to cause *Legionella* lung infections.[7,8]

Diagnostic Considerations

Because many manifestations of *Legionella* are similar to other typical and atypical pneumonias, clinical symptoms or radiologic evidences are of little value for diagnostic purposes. The Centers for Disease Control and Prevention defines confirmation of infection if *Legionella* can be cultured from sputum or bronchoalveolar lavage, a positive urine antigen test, or a 4-fold increase in antibodies specific to *Legionella*.[9,10] Details about these tests are summarized in **Table 1**. Polymerase chain reaction (PCR)-based diagnostic tests are being tested and some of them show specificity and sensitivity, although these tests are yet to be approved by Food and Drug Administration (FDA). Other tools, such as direct immunostaining, are used to detect the presence of bacterium but frequently require invasive procedures to collect tissue for testing.[11]

Prognosis

Legionnaires' disease has significant mortality rates if untreated or if there is delay in administrating appropriate antibiotic therapy. The risk factors associated with mortality are acquiring the infection in nosocomial settings, diabetes, immunosuppression, and malignancies.[12,13] Complete recovery from the infection in these susceptible populations might be prolonged and signs of stress and trauma might persist for years.[14]

Treatment

Antibiotics are the first-line therapy for *Legionella* pneumonia. Failure to administer appropriate antimicrobial therapies at early stage of infection is associated with high mortality rates.[15,16] The correct choice of antibiotic depends not only on its in vitro bactericidal or bacteriostatic activity but also on its ability to penetrate the cell membrane of host tissues because *Legionella* resides within host tissue cells. Fluoroquinolones and macrolides are the 2 most commonly used and highly effective antibiotics to treat patients with legionnaires' disease. Including these agents in initial treatment regimen is prudent if *Legionella* infection is suspected based on an ongoing outbreak in the area, travel history, or extrapulmonary symptoms.[17]

It was found during the first reported outbreak of legionnaires' disease that tetracycline and erythromycin are more effective than other antibiotics, such as β-lactam antibiotics, whereas the use of steroids has been associated with unfavorable outcome.[1] Erythromycin has been the antibiotic

Table 1
Diagnostic tests for *Legionella* species

Test	Sensitivity (%)	Advantages	Limitations
Culture	20–80	Detects all the *Legionella* species	Takes technical expertise, longer duration >5 d
Urinary antigen	70–100	Quick, same-day results, not affected by antibiotic treatment	Kits available are limited mostly to *Legionella pneumophila*; other species may go undetected
Serology	80–90	Little effect of antibiotic treatment	Paired samples are required
Direct fluorescence assay	25–75	Performed on pathologic tissue	Technically difficult

of choice for the treatment of legionnaires' disease that is highly effective but has been associated with significant side effects, especially when used intravenously.[16,18–20] Azithromycin, another macrolide, has been shown highly effective in treating patients with *Legionella* infection, with minor side effects.[21] Azithromycin has been successfully used to treat *Legionella* infection not responding to erythromycin and is frequently chosen to treat patients infected with *Legionella*.[22] Other antibiotics that are effective against *Legionella* are clarithromycin, rifampin, ciprofloxacin, and doxycycline, and these are used either alone or with erythromycin.[18] In a prospective study, it has been shown that fluoroquinolones are at least as effective as erythromycin in treating patients with legionnaires' disease.[23] Levofloxacin, either 500 mg for 10 days or 750 mg for 5 days, can cure most of the patients (>95%) and is becoming the antibiotic of choice for legionnaires' disease.[24] Use of levofloxacin is increasing to treat *Legionella* infection and is associated with early clinical response and shorter hospital stay.[25] A meta-analysis by Burdet and colleagues[26] revealed quinolones may be superior to macrolides in treating the *Legionella* infection.

The usual duration of therapy for most of the antibiotics is 5 to 10 days and is often sufficient to completely treat patients with *Legionella* infection, but duration of therapy up to 3 weeks may be considered in immunocompromised patients.[17] The route of administration used for the antibiotics depends on the severity of the infection, with parenteral therapy preferred for severe infections. If intravenous therapy is initiated early in infection, then therapy can be transitioned to oral route to complete therapy once a desirable response is observed. Treatment options are outlined in **Table 2**.

Acquired antibiotic resistance among *Legionella* species can be seen in vitro but is rarely reported in vivo, although a recent report has shown the presence of fluoroquinolone resistance in *Legionella* in patients who are treated with these antibiotics.[27,28] These reports warrant special attention toward ineffectiveness or relapse of disease during ongoing antibiotic therapy.

Table 2
Antibiotic therapy for *Legionella*, *Chlamydophila*, and *Mycoplasma* community-acquired pneumonia

Medication	Dose
Azithromycin	1.5 g over 5 d (500 mg on day 1 followed by 250 mg for 4 d)
Clarithromycin	500 mg PO bid for 10 d
Doxycycline	100 mg bid for 7–21 d
Tetracycline	250 mg qid for 7–21 d
Levofloxacin	750 mg PO/IV for 5–10 d or 500 mg PO/IV daily for 7–14 d
Moxifloxacin	400 mg daily for 10 d
Nemonoxacin[a]	500 mg daily for 7 d or 750 mg daily for 7 d
Slorithromycin[a]	800 mg on day 1 followed by 400 mg daily for 4 d

[a] Nemonoxacin and slorithromycin remain in the trial phase and are not yet FDA approved. Nemonoxacin treatment was associated with clinical in all patients with *C pneumoniae* identified as etiologic pathogen between 22 phase II clinical trials (n = 9). Slorithromycin shows in vitro activity against *C pneumoniae* but has not been specifically tested in vivo.
Data from Refs.[60,62,66]

Conflicts and Controversies

Most cases of legionnaires' disease reported are due to *Legionella* pneumophila serotype-1 (80%).[29] This might reflect a diagnosis bias because most of the commercial kits available detect *Legionella* serotype-1 antigen in urine samples but not for other species. Efforts are under way to develop rapid diagnostic test for *Legionella* species, such as multiplex PCR assays, and may be more efficacious than detection of *Legionella* pneumophila serotype-1 antigen in patients' urine.[11,30]

To date, there are few reported cases of *Legionella* species that are resistant to conventional antibiotics resistance and there is little evidence that combination therapy is superior to monotherapy.[31,32] *Legionella* resistant to ciprofloxacin has been reported. It was unclear if the strain of *Legionella* was resistant at the presentation of disease or developed resistance during treatment because the patient was treated with ciprofloxacin and clinically improved from severe infection.[27] Regardless, several new antibiotics are under development that target intracellular organisms, such as *Legionella*, either by favoring a low pH enthronement or by inhibiting bacterial protein synthesis.[33–35] Currently, these therapies are not available for clinical use.

Person-to-person transfer is usually not considered a route of transmission for *Legionella*; however, reports are emerging showing person-to-person transfer.[36,37] Despite these reports, person-to-person contact seems to be the exception. The best means of preventing disease is by thwarting the contamination of water supplies. Water temperature, pipe age, and pipe configuration have been

shown to play a role in the contamination of water supplies with *Legionella*.[38,39] Current recommendations to prevent *Legionella* contamination include maintaining water temperature outside the optimal temperature for *Legionella* growth, preventing stagnation, superheat-and-flush or point-of-use filters, UV irradiation, and chemical disinfection.[40] Currently there are no clear recommendations as to optimal combination of preventative measures; therefore, despite the method of prevention utilized, the World Health Organization recommends quarterly water testing.[41]

CHLAMYDOPHILA PNEUMONIAE

Clinical Presentation

Chlamydophila pneumoniae has been implicated in upper respiratory infections, acute bronchitis, and pneumonia.[42] The common symptoms of *C pneumoniae* pneumonia and their frequencies are presented in **Table 3**. Classically, pneumonia due to *C pneumoniae* presents as a mild illness predominated by fever and cough, often preceded by upper respiratory symptoms of rhinitis and sore throat. In a 2013 study of an outbreak by Conklin and colleagues,[43] duration of cough ranged from 1 to 64 days with a mean of 21 days. Although the classic presentation is associated with nonproductive cough, approximately 70% of patients presented with sputum production in outbreaks of *C pneumoniae* infection in 2006 and 2013. The presentation is especially difficult to distinguish from pneumonia due to *Mycoplasma pneumoniae* or respiratory viruses. Despite previous suggestions that hoarseness and laryngitis are more common in infection from *C pneumoniae* than from *M pneumoniae*, comparison of clinical features of both causes have shown the opposite.[44,45] Punji and colleagues[45] demonstrated that cough, rhinitis, and hoarseness were significantly more common in *M pneumoniae* infection than in *C Pneumoniae* infection. In the same study, C-reactive protein and aspartate aminotransferase elevations were significantly greater in *C pneumoniae* infection than in *M pneumoniae* infection. Other clinical symptoms and laboratory findings due to the 2 pathogens were not significantly different. C-reactive protein and white blood cell values have been previously shown significantly lower in both *C pneumoniae* and *M pneumoniae* pneumonia than in pneumonia due to *Streptococcus pneumoniae*.[44] No single symptom, laboratory finding, or collection of findings can reliably distinguish pneumonia due to *C pneumoniae* from pneumonia due to other respiratory pathogens. Additionally, *C pneumoniae* infection may occur concomitantly with other pathogens, which may influence clinical presentation.[44]

Table 3
Major symptoms encountered in *Chlamydophila pneumoniae* community-acquired pneumonia

	Frequency (%)
Constitutional	
Fever	68.1–97.8
Myalgias/arthralgias	37.5–40.5
Confusion	7.5
Upper respiratory/ear, nose and throat	
Headache	25–60
Rhinorrhea	6.7–72.9
Sinus pain	43.2
Sore throat	9–72.9
Hoarseness	15.7
Lower respiratory	
Cough	82–98
Sputum production	67.5–68.8
Dyspnea	25–58.3
Wheezing	58.7
Chest pain	9–17.5
Hemoptysis	7.5
Gastrointestinal	
Nausea ± vomiting	5–19.1
Diarrhea	5–12.5

Data from Refs.[43–45]

Imaging

A list of roentgenographic manifestations of *C pneumoniae* is presented in **Table 4**. On initial chest radiograph, a unilateral pattern of alveolar infiltrates or bronchopneumonia predominates. Findings are usually confined to a single lobe with lower lobe involvement more frequent than middle or upper lobe involvement.[46–48] A pattern of interstitial pneumonia is comparatively rare. Up to a quarter of patients may demonstrate a small to moderate-size pleural effusion. Hilar or mediastinal lymphadenopathy is an uncommon finding on chest radiograph. Findings may depend on the timing of imaging in the course of the illness, the method of diagnosis, and whether concomitant infection with another respiratory pathogen is excluded. In 1 review of 17 patients classified as having primary infection, admission chest radiographs showed predominantly unilateral findings with repeat chest radiographs taken an average of 3.8 days later showing predominantly bilateral findings.[46]

Table 4
Major imaging findings in *Chlamydophila pneumoniae* community-acquired pneumonia

Imaging Type	Chest Radiograph (%)	CT Scan (%)
Distribution		
Unilateral	42–75	50
Bilateral	24–25	50
Involvement of only 1 lobe	62–86	33
Lower lobe	88	71
Middle lobe	25	46
Upper lobe	21	67
Chest radiograph patterns		
Bronchopneumonia	88	—
Alveolar infiltrates	29–86	—
Interstitial infiltrates	0–4	—
Air bronchogram	57	—
CT parenchymal findings		
Consolidation	—	83
Bronchovascular bundle thickening	—	71
Reticular or linear opacity	—	62
Ground-glass opacities	—	54
Pulmonary emphysema	—	46
Airway dilatation	—	38
Lymphadenopathy	0–17	33
Pleural effusion	14–38	25

Data from [*Chest radiograph*] Kauppinen MT, Lahde S, Syrjala H. Roentgenographic findings of pneumonia caused by Chlamydia pneumoniae. A comparison with streptococcus pneumonia. Arch Intern Med 1996;156(16):1851–6; Boersma WG, Daniels JM, Löwenberg A, et al. Reliability of radiographic findings and the relation to etiologic agents in community-acquired pneumonia. Respir Med 2006;100(5):926–32; and [*CT scan*] Nambu A, Saito A, Araki T, et al. Chlamydia pneumoniae: comparison with findings of Mycoplasma pneumoniae and Streptococcus pneumoniae at thin-section CT. Radiology 2006;238(1):330–8.

In a retrospective review of thin-section CT scans of 24 patients serologically diagnosed with *C pneumonia* CAP, Nambu and colleagues[49] found a significant increase in airway dilatation compared with patients with pneumonia due to *S pneumoniae* or *M pneumoniae* as well as an increased rate of pulmonary emphysema compared with *M pneumoniae* but not *S pneumoniae*. The study speculated that the increased rate of airway dilatation and pulmonary emphysema may reflect obstructive lung disease as a predisposing risk factor for *C pneumoniae* pneumonia and may not be caused by the infection itself. Despite the statistically significant increase in airway dilatation and/or pulmonary emphysema, neither these findings nor any other on CT was able to reliably distinguish pneumonia from *C pneumoniae* from pneumonia due to other pathogens. Overall, findings in *C pneumoniae* on CT scan were widely variable. Involvement of more than 1 lobe, usually upper and/or lower lobe involvement, with consolidation and bronchovascular bundle thickening were the predominant findings. Bilateral lung involvement was seen in half of patients. Ultimately, the imaging findings on either radiograph or CT scan are nonspecific for *C pneumoniae* and cannot be reliably used to identify the pathogen in the etiology of pneumonia.[46–48]

Diagnostic Considerations

Accepted techniques for identifying *Chlamydophila* infection include serologic studies and culture or PCR of respiratory tract samples. Historically, diagnosis of *Chlamydophila* infection has relied on serologic studies, requiring a 4-fold rise in IgG or IgA levels between acute and convalescent serum samples. Serologic methods in general are cumbersome because patients must return 4 to 6 weeks after initial presentation to retrospectively confirm the diagnosis. Moreover,

the retrospective nature of diagnosis means serologic results have little effect on treatment decisions. Alternative serologic criteria allowing diagnosis on initial presentation, such as a serum IgM antibody titer of 1:16 or greater, rely on the timing of sample collection, because a rise in titers may not be observed early in the course of acute infection or reinfection.[50,51] Relying solely on initial serologic samples for diagnosis (that is, forgoing retrospective confirmation with convalescent serum samples) risks missing 25% to 33% of infections.[52] Additionally, initial serologic testing may take days to result, further limiting their use in initial management decisions. Serologic techniques are limited in specificity by potential cross-reactivity between *C pneumoniae* antigens and antigens of other *Chlamydia* species.

Microimmunofluorescence is considered the reference standard for serologic diagnosis.[42,51] ELISA is also available and may be less technically demanding and more objectively interpretable than microimmunofluorescence.[51] Complement fixation is not a recommended diagnostic technique owing to a limited sensitivity and specificity.[42,52]

Although considered specific due to a low rate of asymptomatic carriage, the sensitivity of culture is markedly limited by the fastidious and slow growth of *Chlamydophila*, which may require weeks.[42,50,53] Previous studies have shown a very low frequency of growth in culture, even from specimens where infection is identified by serology and/or PCR.[52] In a 2010 study, She and colleagues[50] recommended against the routine use of culture for diagnosis after failing to identify any positive culture results from 6981 specimens from patients with respiratory symptoms despite a rate of *Chlamydophila* as the cause of CAP and other respiratory infections of 5% to 22%.

Given the limitations of serology and culture, PCR of respiratory tract specimens has emerged as the favored method of diagnosis. Specimens may be assessed with multiplex PCR, allowing for the detection of multiple potential respiratory pathogens without significant diminishment in sensitivity compared with singleplex PCR testing.[54] In 2012, the FDA approved the FilmArray Respiratory Panel (BioMérieux, France), which uses multiplex PCR for the detection of *C pneumoniae* in addition to *M pneumoniae*, *Bordetella pertussis*, and 17 respiratory viruses on nasopharyngeal swab (NPS) specimens.[55] PCR remains limited in specificity, however, by asymptomatic carriage, which approaches 5% in healthy adults.[53] Specificity is further limited by a pattern of persistence of *Chlamydophila* identified on respiratory swabs well after resolution of clinical symptoms in some patients. In a recent outbreak, approximately 80% of patients who were positive for *Chlamydophila* infection by PCR of respiratory samples remained positive for up to 8 weeks after resolution of symptoms.[43] Patients may continue to harbor the pathogen in the absence of symptoms for up to 11 months, even after appropriate antibiotic therapy.[56] Positive PCR results in patients with a history of *C pneumoniae* infection may, therefore, be challenging to attribute definitively to reinfection, persistent infection or ongoing asymptomatic carriage with other potential pathogens causing new symptoms.[57] Furthermore, the identification of *Chlamydophila* in respiratory samples does not rule out coinfection with other pathogens, which has been noted to occur in multiple studies and may affect clinical presentation.[44,46,47,52,53]

Alternative methods of detection include identification of circulating *Chlamydophila* lipopolysaccharide in serum, *C pneumoniae* presence in circulating phagocytes or atheromas, and seroresponse to *C pneumoniae* antigens. These methods are technically demanding, however, and currently used only in research settings.[51]

Prognosis

Compared with infection with typical bacterial respiratory pathogens, such as *Streptococcus, Klebsiella,* and *Pseudomonas* species, the course and outcomes for pneumonia due to *C pneumoniae* are thought to be benign. Outcomes are typically reported for patients with atypical pneumonias as a group, however, and there are few data available on outcomes specific to *C pneumoniae*.

A 2012 study of etiologic agents in CAP and their effect on outcomes by Capelastegui and colleagues[58] identified 151 patients with pneumonia due to atypical pathogens, 37 of whom (or 24%) had *C pneumoniae*.[49] Atypical pneumonia had a hospitalization rate of 25.8%, an ICU admission rate of 0.7%, and a mechanical ventilation rate of 0.7%. With the exception of mechanical ventilation, these rates were significantly lower for atypical pneumonias than for pneumonia due to typical bacteria; 30-day mortality was 1.3% compared with 4.3% for pneumonia due to typical bacteria, although this difference was not statistically significant. Outcomes more specific to *C pneumoniae* were not reported. The mortality rate of *C pneumoniae* pneumonia is likely low, with 30-day mortality rates for atypical pneumonias in general ranging from 0% to 2.2%.[59] In the 2013 outbreak studied by Conklin and colleagues[43] no deaths were reported among 52 patients. However, 22 of these patients had persistently positive oropharyngeal swabs (OPSs)

for *C pneumoniae* up to 8 weeks after the outbreak, and many of these patients experienced cough symptoms for several weeks after completion of antibiotic treatment. Patients should be advised that cough could persist even after completion of an appropriate antibiotic course.

Treatment

Recommendations for antibiotic treatment of *C pneumoniae* are limited by an absence of standardized diagnostic criteria and the use of serology alone for diagnosis in most previous studies. Infectious Diseases Society of America (IDSA) guidelines from 2007 note a lack of strong evidence to recommend specific antibiotic therapy for the pathogen.[17] Treatment recommendations continue to rely on expert opinion. Given a pattern of reappearance of symptoms after a standard course of therapy, longer courses of antibiotics have been recommended when *Chlamydophila* is identified.[42] A list of antibiotics, their doses, and treatment courses as recommended by expert opinion is given in **Table 2**.[60]

Because *C pneumoniae* is an obligate intracellular microbe, antibiotics must achieve intracellular penetration to achieve efficacy. Antibiotics that interfere with DNA and protein synthesis, including macrolides, tetracyclines, and fluoroquinolones, demonstrate in vitro activity against the pathogen and are the recommended drug classes for clinical treatment. Ciprofloxacin, however, demonstrates a higher minimum inhibitory concentration than other fluoroquinolones and may, therefore, be less efficacious. *C pneumoniae* is resistant to trimethoprim, sulfonamides, aminoglycosides, and glycopeptides. Penicillin and amoxicillin have demonstrated in vitro activity against *Chlamydia* species but are not recommended as part of routine therapy against *C pneumoniae*.

Resistance to the recommended therapies is considered rare and does not seem to play a role in either treatment failure or in the persistence of *C pneumoniae* identified on respiratory samples after completion of therapy because isolates obtained from patients after appropriate therapy demonstrate in vitro sensitivity.

Three novel antibiotics, nemonoxacin, slorithromycin, and AZD0914, have all demonstrated in vitro activity against *Chlamydophila* but are currently in trial stages and have not yet received FDA approval for treatment.[61–63] Nemonoxacin is a novel fluoroquinolone with in vitro activity comparable to azithromycin, doxycycline, and levofloxacin.[62] In 2 phase II clinical trials of 256 and 192 patients with mild to moderately severe CAP, nemonoxacin led to clinical treatment success in all patients identified as having *C pneumoniae*, although this totaled only 9 patients between the 2 trails.[64,65] Slorithromycin is a novel fourth-generation macrolide with in vitro activity against *Chlamydophila* that demonstrated noninferiority to moxifloxacin for the treatment of CAP in a recent phase III clinical trial.[66] No patients with *Chlamydophila* were specifically identified in the study. AZD0914 is a bacterial DNA gyrase/topoisomerase inhibitor that demonstrates high activity against *Chlamydophila* and other respiratory pathogens in vitro but is not yet under clinical investigation for treatment of respiratory infections.[63]

Conflicts and Controversies

C pneumoniae infection has been identified as a possible contributing factor in a multitude of chronic conditions. A 2013 meta-analysis by Orrskog and colleagues[67] identified *C pneumoniae* infection as potentially linked with 26 chronic conditions, most strongly with conditions of the circulatory system. Research interest into a causal link between *Chlamydophila* infection and atherosclerosis has been intense since 1988, when Saikku and colleagues[68] identified a higher rate of serologic evidence of infection in patients with a history of coronary heart disease. Subsequently, *C pneumoniae* was identified by culture, PCR, and immunohistochemical methods in macrophages, endothelial cells, and smooth muscle cells in atherosclerotic vessel walls. Each of these techniques has been criticized, however, given that isolation in culture is rare and inconsistent, PCR identification is widely variable and potentially prone to contamination, and immunohistochemical staining is plagued by cross-reactivity with human proteins.[69] Furthermore, identification of *C pneumoniae* in atherosclerotic lesions has not correlated well with seropositivity. It has been suggested that the initially identified serologic markers, such as elevations in IgG, may be more reflective of atherosclerotic processes other than persistent *C pneumoniae* infection, such as smoking and inflammation.[70] In recent meta-analyses, elevated titers of IgG or IgA to *C pneumoniae* have been associated with increased stroke risk and increased inflammatory markers.[71,72]

The connection between *C pneumoniae* infection and atherosclerosis has been most strongly shaken by disappointing results in studies of antibiotic therapy. A 2005 meta-analysis of 11 randomized controlled trials, including 19,217 patients with established coronary artery disease, showed that antibiotic therapy had no effect on rates of myocardial infarction or all-cause mortality.[73] The CLARICOR trial, which demonstrated

an unexpected increase in long-term mortality after short-term treatment with clarithromycin in patients with stable coronary heart disease, further contributed to doubt in the association.[70] The failure of antibiotic therapies to influence cardiovascular outcomes may reflect a lack of an association but could also result from the limited efficacy of antibiotics to penetrate atherosclerotic plaques or eradicate infection. Alternatively, the initiation of atherosclerosis may depend on transient *C pneumoniae* infection rather than chronic infection. Ultimately, the hypothesized association remains to be definitively demonstrated.[74]

Definitively implicating persistent *C pneumoniae* infection in the pathogenesis of chronic diseases will first require a method of reliably identifying persistent infection. No standardized method yet exists, but potential methods have been investigated.[51] In a 2008 study by Bunk and colleagues[75] using proteomics, 12 *C pneumoniae* antigens were identified that produce a serologic IgG antibody response in patients shown to have persistent infection by PCR of either circulating phagocytes or atheromas. Two antigens, Cpaf-c and RpoA, produced the strongest response and could potentially be used in the future as evidence of chronic infection. The possibility that *C pneumoniae* infection, however, may play an initiating role in the pathogenesis of chronic conditions that does not require chronic infection remains.

MYCOPLASMA PNEUMONIAE

Clinical Manifestations

Pneumonia due to *M pneumoniae* can often have a misleading clinical picture with its mild and indistinct symptoms, such as myalgias, cervical adenopathy, nonproductive cough, and fatigue, making it difficult to distinguish from other upper respiratory infections caused by viruses and other atypical bacterium.[76–78] The age group most often affected by *M pneumoniae* include school-aged children and young adults with outbreaks typically occurring during the autumn season.[76–79] Outbreaks occur among close contacts and members within the same household or confined spaces.[80] Apart from its atypical symptoms, *M pneumoniae* presentations can vary dramatically ranging from the mild upper respiratory symptoms to pneumonia and other extrapulmonary manifestations in absence of pneumonia,[6] including dermatologic, cardiovascular, and central nervous system findings.[81] The extrapulmonary manifestations of *M pneumoniae* are outlined in **Table 5**.

Imaging characteristics of *M pneumoniae* infections also follow along with its indistinct nature. The chest radiograph often shows diffuse interstitial patterns sometimes out of proportion to a patient's physical findings. On CT of the chest, the interstitial changes seen in the chest radiograph show up as tree-in-bud formation.[82] In 2016, Gong and colleagues[82] completed a prospective study that looked at a population of 1280 pediatric cases with *M pneumoniae* pneumonia between the years 2010 to 2014 and found that there were a high proportion of the patients with extensive patchy infiltrates both unilaterally and bilaterally indicating that the diagnosis of pneumonia could not be made on imaging characteristics alone and should occur with clinical findings. Other findings found on CT chest imaging include bronchial wall thickening and ground-glass consolidation.

Table 5
Extrapulmonary manifestations of *Mycoplasma pneumoniae*

Skin	Erythema Nodosum, Cutaneous Leukocytoclastic Vasculitis, Stevens-Johnson Syndrome
Gastrointestinal	Acute hepatitis
Central nervous system	Encephalitis, aseptic meningitis
Cardiovascular	Cardiac thrombi, Kawasaki disease

Diagnostic Considerations

The diagnosis of pneumonia has long been considered a clinical diagnosis as encouraged by the IDSA where a patient should have suggestive symptoms and associated imaging findings correlating with pneumonia and other associated diagnostic techniques have remained controversial due to frequent low yield results.[17] For an overview of diagnostic techniques, see **Table 6**.

Confirmatory diagnostic testing plays an important role in delineating epidemiology of infection and antibiotic resistance patterns. Traditionally diagnosis of *M pneumoniae* has come from cultures and serology where isolation via culture was considered the gold standard. Given the fastidious nature of *M pneumoniae* it is not routinely cultured anymore because it is slow growing and culture results are often inconsistent and provide poor clinical utility given the length of time the organism takes to grow.[77,79]

Alternative methods of diagnosing *M pneumoniae* include serologic studies using ELISA to quantify expression of antibodies to the bacteria,

Table 6
Diagnosis of *Mycoplasma pneumoniae*

Diagnostic Test	Sample Type	Advantages/Disadvantages of Test
Culture	Sputum	Advantages • If positive, 100% specific and considered the gold standard Disadvantages • Long growth period that provides limited clinical utility
Serology	Serum	Advantages • Test has ability to quantify expression amount Disadvantages • Poor sensitivity and specificity • Requires paired sera (acute and convalescent phases) leading to retrospective results High false-positive rate likely due to carrier state
Molecular	Sputum, NPA, NPS, OPS	Advantages • Readily available with fast results; high specificity Disadvantages • Expensive commercial kits • Improved standardization among kits required to determine optimal sample specimen

microparticle agglutination studies and complement fixation assays. For a definitive diagnosis in the serologic studies paired sera were needed to demonstrate a significant 4-fold elevation of IgG or a subsequent seroconversion of IgG in the sera collected 3 to 4 weeks later.[83–86] Due to the delay in antibody production during initial infection and the time needed to allow for seroconversion, the serologic tests also have poor utility in diagnosing acute *M pneumoniae* infections in clinical practice and functioned more as a retrospective confirmation for epidemiologic studies.[79,83–85] With the many disadvantages of culture and serology in diagnosing *M pneumoniae* infections, diagnostics are evolving toward more rapid molecular techniques including nucleic acid amplification techniques.

Molecular diagnostic techniques allow for a timely diagnosis of *M pneumoniae* infections and are quickly becoming the mainstay for diagnosis in clinical practice with the development of a vast repertoire of laboratory techniques including nucleic acid amplification techniques, multilocus variable number tandem-repeat analysis, multilocus sequence typing, among many others.[79] These tests have quickly become preferential with their ability to produce fast results with high specificity and sensitivity.[79,83] Many of the new tests undergo real-time PCR to look at specific gene regions of *M pneumoniae* as the regions encoding 16S ribosomal RNA, P1 gene, ATPase operon, and the community-acquired respiratory distress syndrome (CARDS) toxin.[79,83–86] This technology allowed for the development of multiplex PCR, which often allow for the detection of several atypical pathogens, including *C pneumoniae*, *C psittaci*, and *Legionella* species, among other respiratory viruses.[54,79] There still remains some debate over which sample type has the best sensitivity and specificity while performing these assays, with current studies showing that sputum samples yield more positive results than both nasopharyngeal aspirates (NPAs) and NPSs as well as OPSs.[85,87]

Prognosis

The clinical course of *M pneumoniae* infections is usually mild and self-limiting in nature and resolves within 2 to 4 weeks regardless of treatment.[77,78,83,84] There have been cases of severe infections, however, resulting in acute respiratory distress syndrome and severe neurologic complications that are associated with increased morbidity and mortality.[88]

Treatment

Infection from *M pneumoniae* is often underdiagnosed, where patients tend to not seek treatment given the subacute nature of their symptoms.[76–79] The bacterium has a long incubation of approximately 3 weeks with prolonged bacterial shedding where symptoms can last up to 4 months; however, most cases resolve naturally within 2 to 4 weeks without treatment.[77,79,83]

When patients present for clinical care, treatment is often guided by the IDSA guidelines for CAP based on a patient's symptoms and imaging

results.[17] *M pneumoniae*, as a small, self-replicating bacteria that lacks a cell wall, is inherently resistant to the family of β-lactam type of antimicrobials but is routinely covered in the empiric treatment of CAP treatment with macrolide therapy, usually without a formal laboratory diagnosis. Treatment with such antimicrobials can shorten the course of the illness by using a 5-day to 2-week course of antibiotic therapy dependent on the choice of antibiotic in infected individuals.[89,90] Because *M pneumoniae* often affects children and young adults, macrolides have become the treatment of choice because both tetracyclines and fluoroquinolones have unfavorable side-effect profiles that can occur in the younger patient population, such as discoloration of dentition with tetracyclines and tendinitis with fluoroquinolones.[90]

The treatment of extrapulmonary symptoms or complicated *M pneumoniae* pneumonia remains unclear apart from the administration of antibiotics. In patients with extrapulmonary conditions associated with *M pneumoniae*, it is important to understand the inflammatory nature of the bacteria where, through pathways associated with Toll-like receptor 2, the bacteria are able to induce proinflammatory cytokines and inflammasome activity.[91] This partially helps explain why the symptoms are present more often in young adults who express a more robust immune response rather than infants or geriatric patients who are unable to mount the same level of response.[92] In patients with central nervous system syndromes from *M pneumoniae*, such as encephalitis and stroke, case reports have suggested the use of steroids and immunoglobulin therapy may be of benefit, although this has not been validated in clinical trials.[5,93] Similar reports have been made for patients with severe *M pneumoniae* pneumonia resulting in acute respiratory distress syndrome, suggesting possible benefit from extracorporeal membrane oxygenation and steroids.[5,84,88] Antimicrobial options are summarized in **Table 2**.

Conflicts and Controversies

Infections with *M pneumoniae* are usually mild, which can make it a difficult diagnosis; however, complications can occur with severe infections that sometimes correlate with macrolide-resistant strains and reiterate the importance of therapy guidelines.

With its mild clinical presentation, *M pneumoniae* can be a challenging clinical diagnosis as one that often mimics mild respiratory viruses; or, patients fail to present for evaluation due to their low-grade symptoms, making it an underdiagnosed infection. With the development of many novel molecular diagnostic techniques, it is becoming faster and easier for clinicians to make a formal diagnosis; however, with the many new techniques, there is still no standardized test recommended by IDSA guidelines. Several barriers that may arise in the primary care settings are that many of these molecular tests are expensive and many of these techniques require specialized laboratory equipment. There have been several assays developed that allow for the convenience of testing for multiple pathogens, with current tests approved for clinical use, including the Bioscience USA illumigene assay (Meridian Bioscience, USA) approved by the FDA in the United States and the FilmArray Respiratory Panel (BioMérieux, France) approved in parts of Europe.[83,87] These multiplex assays can often detect a positive result, which may not always correlate with the presence of disease because many patients may be a carrier, have a coinfection, or have overcome the clinical infection but still are undergoing a prolonged period of bacterial shedding.[87,94] It remains unclear whether the asymptomatic carriage of *M pneumoniae* or colonization can be differentiated from active infection with the new diagnostic techniques. Such results can cause confusion, make interpretation of results difficult, and may lead to unnecessary treatment with antibiotics and increased health care resources based on initiation of respiratory precautions in hospitalized patients.

Macrolide resistance in *M pneumoniae* has been a rapidly emerging phenomenon with reports of increasing resistance in Asia, Europe, and the United States.[79,95–97] Countries in Asia have shown a large amount of macrolide resistance; in Beijing it has been reported that as many as 98% of certain populations infected with *M pneumoniae* between 2008 and 2012 are resistant to macrolide therapy.[95] The emerging resistance patterns have also been found in the United States, where up to 13% to 27% of *M pneumoniae* infections have been resistant to macrolide therapy.[96,98] Resistance to macrolides can come by various mechanisms, including the most common, a single-nucleotide polymorphism at one of the residues around the binding site of the peptidyl transferase loop of the 23s ribosomal RNA subunit preventing binding, which ultimately can inhibit protein synthesis.[99] It remains unclear as to how the emerging resistance patterns are going to affect clinical prescribing patterns in the near future in the United States; however, at this time, there are no formal recommendations for macrolide prophylaxis in close contacts of infected individuals. The mainstay of preventing infection spread remains handwashing and respiratory droplet isolation to limit transmission of the bacteria.

There have also been studies linking *M pneumoniae* to asthma, supporting that the presence of the bacteria can precede the onset of asthma and also cause acute exacerbations in those with preexisting asthma. Biscardi and colleagues[100] showed that 20% of pediatric patients requiring hospitalizations due to acute exacerbations of asthma were positive for *M pneumoniae* and 50% of those patients were having their initial exacerbation. A similar study in adult patients showed that 18% of the patients hospitalized for acute asthma exacerbations were positive for *M pneumoniae*.[101] Chronic stable asthmatics have been found to have *M pneumoniae* present in their airways significantly more than control patients and this may help explain some of the chronic inflammation that asthmatics experience and decreased forced expiratory volume in the first second of expiration (FEV_1) due to the IgE-mediated hypersensitivity effect that *M pneumoniae* has on the airways.[102] Treatment with macrolides, such as clarithromycin, can improve FEV_1, it is suspected that either the antimicrobial aspect of macrolides on *M pneumoniae* or their ability to modulate inflammation may be responsible for this improvement.[103]

SUMMARY

CAP due to *Legionella*, *Chlamydophyla*, or *Mycoplasma* continues to be a diagnostic challenge due to the nonspecific clinical and radiographic presentations. The vague clinical presentations of atypical CAP contribute to its underdiagnosis and under-reporting. Advancements in diagnostic techniques bring hope to rapid and accurate diagnosis of atypical CAP. Macrolides and respiratory fluoroquinolones are currently the antibiotics of choice, but this may change in the near future as more antibiotics resistance patterns emerge for atypical CAP. Several controversies still exist in atypical CAP, underscoring the need for continued investigation of preventing atypical CAP and determine its association with chronic lung diseases.

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