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### Abstract

Following a large outbreak of community-acquired psittacosis in 2002 in residents of the Blue Mountains, New South Wales, Australia, we reviewed new cases in this area over a 7-year period from 2003 to 2009. Using the 2010 criteria from the Centers for Disease Control National Notifiable Diseases Surveillance System, 85 patients with possible psittacosis were identified, of which 48 were identified as definite or probable infection. Clinical features of these cases are summarized. In addition to Chlamydia-specific serology, specimens, where available, underwent nucleic acid testing for chlamydial DNA using real-time PCR. Chlamydia *psittaci* DNA was detected in samples from 23 patients. Four of 18 specimens were culture positive. This is the first description of endemic psittacosis, and is characterized in this location by community-acquired psittacosis resulting from inadvertent exposure to birds. The disease is likely to be under-diagnosed, and may often be mistaken for gastroenteritis or meningitis given the frequency of non-respiratory symptoms, particularly without a history of contact with birds. Clinical characteristics of endemic and outbreak-associated cases were similar. The nature of exposure, risk factors and reasons for the occurrence of outbreaks of psittacosis require further investigation.

### Keywords

Atypical pneumonia, Chlamydia *psittaci*, community-acquired pneumonia, medical geography, ornithosis, psittacosis

### Disciplines

Medicine and Health Sciences | Social and Behavioral Sciences

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## Clinical features of endemic community-acquired psittacosis

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### Abstract

Following a large outbreak of community-acquired psittacosis in 2002 in residents of the Blue Mountains, New South Wales, Australia, we reviewed new cases in this area over a 7-year period from 2003 to 2009. Using the 2010 criteria from the Centers for Disease Control National Notifiable Diseases Surveillance System, 85 patients with possible psittacosis were identified, of which 48 were identified as definite or probable infection. Clinical features of these cases are summarized. In addition to *Chlamydia*-specific serology, specimens, where available, underwent nucleic acid testing for chlamydial DNA using real-time PCR. *Chlamydomphila psittaci* DNA was detected in samples from 23 patients. Four of 18 specimens were culture positive. This is the first description of endemic psittacosis, and is characterized in this location by community-acquired psittacosis resulting from inadvertent exposure to birds. The disease is likely to be under-diagnosed, and may often be mistaken for gastroenteritis or meningitis given the frequency of non-respiratory symptoms, particularly without a history of contact with birds. Clinical characteristics of endemic and outbreak-associated cases were similar. The nature of exposure, risk factors and reasons for the occurrence of outbreaks of psittacosis require further investigation.

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### Introduction

Among members of the chlamydiaceae family, there is now a single genus (*Chlamydia*) that includes three major human pathogens: *Chlamydomphila pneumoniae*, *C. psittaci* and *C. trachomatis* [1]. *Chlamydomphila psittaci* is the agent of a multisystem disease called 'psittacosis' [2]. This zoonotic infection most commonly occurs in people with a history of exposure to birds in either the setting of occupational or companion animal exposure [3, 4]. It causes community-acquired pneumonia with a frequency of less than 2% [5, 6]. The features of psittacosis

were extensively reviewed prior to the identification of *C. pneumoniae* as an alternative diagnosis of chlamydial pneumonia [7, 8]. It is recognized now that human respiratory infection can be caused by both *C. pneumoniae* and *C. psittaci*.

Micro-immunofluorescence (MIF) serology together with rapid ELISA screening has for many years been the mainstay of diagnosis of psittacosis and is reported to be more specific than complement fixation testing (CFT) [9]. Initial ELISA testing for *Chlamydia*-specific IgG and IgA may be used to screen specimens prior to the more laborious MIF testing, which is often reserved for testing acute and convalescent specimens in parallel. Recently, diagnostic criteria for psittacosis have been published by the USA Centers for Disease Control (CDC) National Notifiable Disease Surveillance System (NNDSS), classifying cases as confirmed or probable according to culture, species-specific nucleic acid testing (NAT) and serology (<http://www.cdc.gov/nndss>, under psittacosis).

The Blue Mountains region is west of Sydney, New South Wales, Australia, and is a heavily forested area with an altitude gradient from sea level to 1000 m. The human population resides close to the major roads and railway lines that traverse the mountains. Similar numbers of the population live below an altitude of 360 m (lower mountains) and above this altitude (upper mountains). Despite the presence of similar bird species populations in both regions the majority of cases of psittacosis occur in the upper mountains. The factors responsible for this remain unclear but may pertain to the environmental survival of *C. psittaci*. The entire Blue Mountains region abounds in bird life, particularly in psittacine birds, such as the crimson rosella (*Platycercus elegans*), the king parrot (*Alisterus scapularis*) and the sulphur-crested cockatoo (*Cacatua galerita*).

In 2002, an outbreak of 59 cases of serologically diagnosed (and hence probable) community-acquired psittacosis occurred [10]. Epidemiological risk factors included residence in the upper mountains, males between the ages of 40 and 60 years, recent sighting of birds and mowing lawns without a grass-catcher. It was suggested that mowing lawns and other selected outdoor activities may generate infective aerosols of bird material and secretions, an explanation also given for an outbreak of 16 cases in Bright, Victoria, Australia, in 1997 [11]. Both these Australian reports were criticized because they did not provide conclusive evidence that the infective agent was *C. psittaci*. In this study, we describe endemic community-acquired psittacosis and summarize the clinical features of confirmed and probable cases, using the 2010 CDC NNDSS definitions as the basis for diagnosis.

## Methods

The Blue Mountains Local Government Area (BMLGA) was chosen as the location for this study. Psittacosis is notifiable to public health authorities in New South Wales, with cases consistently reported from the BMLGA. Notified cases were reviewed and classified as confirmed, probable or possible. Cases were classified as confirmed if the organism was cultured, or there was a fourfold rise in species-specific antibody (using MIF). Cases were classified as probable if there was a positive, species-specific, NAT result (using PCR) or a single serology result with a MIF titre equal to or greater than 128 (one dilution higher than the manufacturer's recommendation). Enzyme immunoassay (EIA) seropositivity was used as the trigger for performing MIF. To avoid low-level seropositive results, patients with MIF titres of 1:64 or less were regarded as possible cases only, and were excluded. Patients with only EIA evidence (cut-off ratio of 2.2) of chlamydial infection without species-specific confirmation by MIF or PCR were

regarded as possible cases and not analysed further. Absence of alternative diagnoses and lack of positive species-specific PCR for other chlamydial species was required for inclusion.

Active surveillance for clinical cases of community-acquired psittacosis was implemented in 2002. People presenting to the Blue Mountains Hospital Emergency Department with community-acquired pneumonia or an unexplained febrile illness had throat swabs collected for chlamydial PCR and culture, as well as other specimens for serology and PCR. Chlamydial PCR was performed using a Corbett real-time platform with genus-specific [12, 13] and two species-specific primers [14–16]. More recently, an additional primer sequence based on the *INC A* gene was used [17]. PCR tests for *C. pneumoniae* and *C. trachomatis* DNA were performed in parallel, using primers based on the published species-specific sequences, to exclude inadvertent species cross-reactions [15, 16].

Limited numbers of specimens were available for culture: because of the potential laboratory risks, only those specimens that were PCR-positive were cultured. Specimens were cultured in a monkey green kidney cell line using two passages as described [14]. Chlamydial cultures were confirmed using both the genus-specific direct fluorescence assay *Pathfinder* (Biorad, Hercules, CA, USA) and the species-specific PCR.

Initial (acute) serology was performed using an EIA screen for chlamydial IgG and IgA (Medac, Wedel, Germany). The manufacturer's cut-off of 1.0 was raised to 2.2 following an in-house assessment that the specificity was increased at this level (data not shown). EIA testing, if positive, was followed by MIF IgG testing (Savyon, Ashod, Israel). Where acute and convalescent specimens were available, MIF assays were performed in parallel in one laboratory. *C. pneumoniae* and *C. trachomatis* MIFs were performed concurrently.

Follow-up public health surveys and reviews of inpatient medical records were used to determine clinical presentation and risk factors of cases. Where possible, patients were interviewed soon after the diagnosis and questioned about possible risk exposures to birds. Convalescent serology was requested 6 weeks after symptom onset. Re-presentations to hospital were recorded.

## Results

Between January 2003 and December 2009, 84 cases of psittacosis in the BMLGA were notified to the regional public health unit. Twenty-one patients did not meet the diagnostic criteria of proven or probable psittacosis and were excluded. Four patients were not available for follow-up. Five patients who lived outside the BMLGA were excluded. Five patients had an alternative diagnosis (three with *C. trachomatis* and one

each with *C. pneumoniae* and *Legionella pneumophila* infection). One patient had a second episode of possible psittacosis and this episode was excluded. Forty-eight patients met the diagnostic criteria and lived in the Blue Mountains. The total number of all presentations of pneumonia in this period was 1184, giving a crude prevalence of 35/1184 (2.9%).

Of the 48 cases included in this study, 14 were definite (four culture positive and 10 sero-conversions) and 34 probable (18 diagnosed by serology alone and 16 were PCR-positive and/or serology positive). In total, there were 23 patients with positive PCR results (six PCR-positive only, nine PCR-positive plus serology, four PCR-positive plus sero-conversion, two PCR-positive plus culture, and two PCR-positive plus culture plus serology, one sero-conversion).

Eight patients gave a history of direct bird contact (handled birds or kept birds at home); four, of occupational exposure (veterinary: *n* = 3, wildlife rescue: *n* = 1); and nine patients fed birds in their garden. No records were available for two patients. The majority of patients did not have a history of direct bird contact (*n* = 29, 60%). Indirect contact with birds (seeing birds and excreta in the environment) was universal. A history of lawn-mowing was available for 19 patients with 12/19 (63%) either mowing lawns or using blower devices in the month prior to illness.

There was a striking seasonality to the cases with 31/45 (69%) presenting in a 3-month period of March to May. There were no cases in the warmer months of October to December.

The demographics of residential area, age and sex were similar to the 2002 outbreak in the same region [10]. There was a male predominance and a geographic clustering of cases in the upper mountains.

**Clinical and laboratory features**

Clinical details were available in 42 of the 48 cases. Thirty-six patients were hospitalized, with an average length of hospitalization of 4.5 days. Forty-two patients reported general symptoms (including fever, rigors, myalgia), 28 patients had respiratory symptoms, 26 patients had neurological symptoms, and 15 patients had gastrointestinal symptoms (Table 1). Two patients had a rash; one was an itchy truncal maculopapular rash, possibly related to antibiotic use. In the second case a rash on the torso was described as a non-urticarial maculopapular rash, which lasted 48 h and resolved spontaneously.

The majority, but not all, patients had evidence of pneumonia (*n* = 35, 83%). All patients had a fever or a history of febrile illness. Their average temperature on presentation was 38.3°C with a peak fever of 39°C and the mean time to fever resolution was 2.8 days. Headache was common (52%). Two patients underwent a lumbar puncture; *C. psittaci* DNA

**TABLE 1. Clinical presentations of patients with proven and probable psittacosis**

Symptom	Total ( <i>n</i> = 44), <i>n</i> (%)
Fever	42 (95)
Cough	25 (57)
Headache	23 (52)
Myalgia	10 (23)
Nausea	8 (18)
Diarrhoea	6 (14)
Rigors	6 (14)
Sputum	5 (11)
Chest pain	5 (11)
Dyspnoea	3 (7)
Abdominal pain	2 (5)
Haemoptysis	1 (2)

was detected by PCR in the cerebro-spinal fluid (CSF) of one patient. Peripheral blood leucocyte counts were generally normal (mean  $9.4 \times 10^9/L$ ) with a mean neutrophil count of  $6.3 \times 10^9/L$ . Serum sodium was mildly decreased in 16 of the 48 cases (mean 131.7, range 126 to 134 mmol/L). In four patients renal function was mildly abnormal and returned to normal with recovery.

Liver function tests (LFTs) on presentation were abnormal in 24 (60%) of 40 patients tested, with no result available in nine (19%) patients, and normal in 15 (31%). Seventeen (35%) patients had LFTs less than twice the upper limit of normal (ULN), six (12.5%) had tests between 2 and 5 × ULN, and one patient had >5 × ULN. The predominant abnormality was transaminitis (*n* = 21, 44%). Clinical features and co-morbidities at presentation are summarized in Tables 1 and 2.

***Chlamydomphila psittaci*-specific MIF tests**

Thirty-eight (of 48) patients were tested. Ten had fourfold increases in titre, 26 had a single high titre, one had a falling titre and one remained negative. Single titre results ranged from 64 to 1024 with a mode of 128.

**TABLE 2. Comorbidities in patients with proven and probable psittacosis**

Comorbidity	Total ( <i>n</i> = 33), <i>n</i> (%)
Hypertension	5 (15)
Hyperlipidaemia	4 (12)
Ischaemic heart disease	4 (12)
Cancer (prostate and transitional cell cancer and squamous cell cancer)	3 (9)
Diabetes	3 (9)
Pneumonia	3 (9)
Mental health problems	2 (6)
Chronic airflow limitation	2 (6)
Thyroid disease	2 (6)
Cardiomyopathy	1 (3)
HIV	1 (3)
Asthma	1 (3)

## PCR

Twenty-three patients had at least one specimen positive by *C. psittaci* PCR; throat swab in 16 and sputum in three; other positives included CSF, blood and urine. Eight confirmed and 15 probable cases were PCR positive. Eighteen patients had a specimen available for culture; four (22%) were culture positive. All four positive cultures were from throat swabs collected prior to commencement of antibiotics.

## Radiology

Thirty-eight patients presented with an abnormal chest X-ray; in 32 there was evidence of consolidation or opacification suggestive of pneumonia. One patient had X-ray features suggestive of left ventricular failure and five had non-specific changes, such as peri-bronchial thickening. The lower lobes were involved most frequently (24/38, 63%), followed by the upper lobes in seven (18%) cases and the middle or lingular lobes in 5 (13%). Two patients had bilateral abnormalities. The right lower lobe was affected most commonly, in 15 (39%) patients. There were five patients with pleural effusions (four unilateral and one bilateral). In one patient fibrotic changes were noted in the upper lobes, probably unrelated to psittacosis.

## Treatment and follow-up

Most patients were treated with doxycycline (25 of 44 patients; 56%) or roxithromycin (17 out of 44 patients; 38%). One patient received no effective antibiotics and recovered and one was treated successfully with a quinolone. One patient (2%) who required invasive ventilatory support died of respiratory failure on day 2 of admission. All other patients made a good recovery with few complications. Six patients re-presented to the hospital with respiratory symptoms in the year after their episode of psittacosis. Two of these patients had chronic lung disease and four had further infective episodes of respiratory illness. Others presented with migraine headaches ( $n = 2$ ), back pain ( $n = 2$ ), gastrointestinal problems ( $n = 2$ ) and prostate, sexual health problems and cerebellar ataxia (one each).

## Discussion

Endemic psittacosis has not been described previously. The term has been used in two other studies where the disease has a high prevalence [18, 19] but these did not use strictly defined diagnostic criteria. We now report that, following a large outbreak in 2002 in the Blue Mountains region of New South Wales, Australia [10], psittacosis, as defined by the NNDSS guidelines, is endemic in this region.

*Chlamydophila psittaci* has been described as a sporadic cause of community-acquired pneumonia in up to 2% of cases, with a

frequent corresponding history of bird exposure. Amongst our population the percentage of all pneumonias due to this pathogen is 2.9% but periodically rises to 10–15%. The results presented here may be an under-representation of the incidence of the disease in the region, as less severe or sub-clinical cases managed in the primary care setting are unlikely to be tested and thus unreported to the public health authorities.

Direct bird contact is identified only in a minority of cases reported from the area, although environment exposure to psittacine birds in general is largely unavoidable in the region due to the significant numbers of resident species. Cases occur every year and are seasonal (predominantly March to May). We have proven that these cases are due to *C. psittaci* by culture, molecular and serological diagnosis. Hence, in our region, community-acquired psittacosis is endemic and often occurs without a history of direct bird contact. Further work is required to identify other risk factors. The high, endemic prevalence of psittacosis is presumably due to excessive environmental contamination with native bird material and prolonged survival of the organism in the environment. Additional behavioural and environmental factors, such as altitude, temperature and lawn-mowing, are worthy of further study.

Many of the clinical descriptions of psittacosis predate accurate molecular and serological diagnosis. The two largest case series were reported before *C. pneumoniae* was recognized as a separate pathogen [7,8]. Presented here is a large series of patients with psittacosis, whose clinical presentation is similar to the previous outbreak in this location. We have confirmed the multi-system nature of clinical presentations and with frequent headache, myalgia and gastrointestinal symptoms as well as the major, previously described, laboratory abnormalities of hypernatraemia, abnormal LFTs and normal white cell count (WCC). Chest X-rays revealed a high frequency of unilateral and lobar pneumonia. Pre-morbid conditions including HIV appeared not to influence outcomes adversely.

In this series, non-respiratory symptoms of headache and neurological presentation were common, mirroring previous studies [6,7,19,20]. Moreover, we record a case of psittacosis with a positive PCR result from CSF. This patient had a high degree of direct bird exposure but lacked a CSF cellular response and had normal CSF protein and glucose measurements. To our knowledge a positive PCR result from CSF has only previously been described on one occasion [21]. Clinicians unfamiliar with psittacosis may misdiagnose it as viral illness, meningitis or gastroenteritis, especially in the absence of suspicion or history of bird exposure [22].

Limited follow-up of patients through a search of admission data revealed the need for further admission to hospital with respiratory symptoms in only six (12%) cases of which two had

pre-existing lung disease. It should be noted that at least two patients with possible psittacosis died before a complete set of diagnostic tests could be performed.

In this cohort of patients all three modalities of diagnosis—serology, PCR and culture—may be useful. However we advise against routine culture because of the infectivity of *C. psittaci*. The suggested serological NNDSS criteria for diagnosis of psittacosis are stringent. Our laboratory no longer uses CFT serology as the use of both ELISA and MIF is more reliable [23]. By increasing the recommended cut-off of both serological tests by at least one dilution above the manufacturer's recommendation, we ensured that the patients included in this study were rigorously selected.

The use of genus- and species-specific PCRs that detect both major respiratory species (*C. psittaci* and *C. pneumoniae*) is useful and could be included in multiplex approaches to the diagnosis of community-acquired pneumonia.

In our experience molecular diagnosis for confirmation of *C. psittaci*-specific PCR with a second or third primer set using a different target, as well as using three species real-time PCR in order to exclude infection with *C. pneumoniae* or *C. trachomatis* has been useful. Additionally, we suggest a diagnosis could be confirmed if there is both molecular and serological evidence of infection in the context of an appropriate clinical presentation and the absence of alternate diagnosis.

This study was conducted in the setting of routine clinical care and many patients did not have the requested follow-up serology or appropriate and timely specimens at follow-up, limiting the opportunities to confirm all cases. The retrospective nature of the symptom and follow-up analysis of patients is by its nature limited by the available documentation. Due to the attainment of cases through hospital presentation we may have excluded mild cases from consideration and underestimated the incidence of disease in this environment.

The incidence of *C. pneumoniae* infection was low in the present series. Despite using chlamydial serology, PCR and culture, we were only able to identify two patients with infection due to *C. pneumoniae* during this period, suggesting this pathogen is rare in this geographical area. This is consistent with other Australian studies showing low prevalence of *C. pneumoniae* [5, 24–26].

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## Conflict of Interest

None declared.

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