# National Association of State Public Health Veterinarians, Inc.

TO: State Public Health Veterinarians
State Epidemiologists
State Veterinarians
Interested Pet Bird Professionals

FROM: Kathleen A. Smith, DVM MPH
Chair, Psittacosis Compendium

RE: *Compendium of Measures To Control Chlamydophila psittaci Infection Among Humans (Psittacosis) and Pet Birds (Avian Chlamydiosis), 2009*

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On behalf of the National Association of State Public Health Veterinarians, I am pleased to provide you with a copy of the *Compendium of Measures to Control Chlamydophila psittaci Infection Among Humans (Psittacosis) and Pet Birds (Avian Chlamydiosis), 2009*. The Compendium committee and consultants believe these updates and revisions will aid public health officials, physicians, veterinarians, and the pet bird industry to control this disease in birds and in people.

This Compendium updates the 2008 Compendium. Notable changes in the 2009 Compendium are as follows:

Infection in Humans, Laboratory Testing

- At the time of this report, microimmunofluorescence (MIF) testing kits will no longer be available through FOCUS Diagnostic Laboratories. As a result, the committee recommends that the individual laboratories listed in Table 1 be contacted directly to ascertain the testing options they provide for human specimens.

Appendix 1: Testing Methods for *C. psittaci* in Birds:

- Elementary Body Agglutination (EBA) testing is no longer being offered by Texas Veterinary Medical Diagnostic Laboratory. It is still described under the section titled "Tests for antibodies" as other laboratories may conduct this test in the future

Appendix 2: Treatment Options for Birds with Avian Chlamydiosis:

- The "Recommendations for Treating and Caring for Infected and Exposed Birds" section was moved from the Prevention and Control Recommendation section in the main document to the beginning of Appendix 2. These recommendations are critical for

treatment success and placing them in the treatment section ensures a greater likelihood that they will be reviewed.

- A statement was added that it is important for avian facility managers to provide employees with simple, concise written treatment procedures to ensure treatment success.

- More emphasis is placed on recommending doxycycline over other tetracycline antibiotics for the treatment of avian chlamydiosis

- Routine prophylactic antibiotic treatment is now discouraged as it may cause adverse effects and could generate resistant strains of *C. psittaci* and other bacteria.

The final, and probably the most significant, update is that the Centers for Disease Control and Prevention (CDC) and the University of Georgia College of Veterinary Medicine have collaborated on the development of a new diagnostic test for *C. psittaci*.[i]

Genetic studies have indicated that there are at least seven avian genotypes of *C. psittaci*. Distinguishing these serotypes currently requires multiple confirmatory tests and is very time consuming. A real-time polymerase chain reaction (PCR) assay, targeting the ompA gene has been developed which uses Light Upon extension (LUX $^{TM}$) chemistry and high resolution melt (HRM) analysis that can accurately and quickly differentiate these genotypes. This test may become a valuable epidemiologic tool to evaluate human outbreaks and link human/avian transmission in the future. The CDC is interested in applying this diagnostic tool in select cases or outbreaks. Therefore, if you have suspect human cases and are willing to submit samples for molecular testing, please contact:

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Email: dvj3@cdc.gov

To help control this disease and protect public health, the Compendium committee and its consultants encourage you to distribute this 2009 version of the Compendium to health officials, veterinarians, and the pet bird industry in your state and actively promote the document as a standard. We would also like to request that if you update any web links to this document that you please delete any previous Compendiums, as we want to ensure that interested people access the most current version.

This document will be reviewed and updated on an as needed basis. The most recent version, along with sample case report forms and associated client materials can be accessed on the National Association of State Public Health Veterinarians website at http://www.nasphv.org.

[i] Mitchel, S.L. Wolf, B.J. et al, Genotyping of Chlamydophila psittaci using Real-Time PCR and High Resolution melt Analysis. *J Clin Microbiol* 2009;47:175-181

# Compendium of Measures To Control *Chlamydophila psittaci* Infection Among Humans (Psittacosis) and Pet Birds (Avian Chlamydiosis), 2009

## National Association of State Public Health Veterinarians (NASPHV)

## SUMMARY

Psittacosis, also known as parrot fever and ornithosis, is a bacterial infection of humans that can cause severe pneumonia and other serious health problems. It is caused by *Chlamydophila psittaci*, formerly known as *Chlamydia psittaci***.** From 2002 through 2007, 91 human cases of psittacosis were reported to the Centers for Disease Control and most resulted from exposure to infected pet birds, usually cockatiels, parakeets, parrots, and macaws. In birds, *C psittaci* infection is referred to as avian chlamydiosis. Infected birds shed the bacteria through feces and nasal discharges, and humans become infected from exposure to these materials. This compendium provides information about psittacosis and avian chlamydiosis to public health officials, physicians, veterinarians, the pet bird industry, and others concerned with controlling these diseases and protecting public health. The recommendations in this compendium provide standardized procedures for controlling avian chlamydiosis in birds, a vital step to protecting human health. This document will be reviewed and revised as necessary.

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# Compendium of Measures To Control *Chlamydophila psittaci* Infection Among Humans (Psittacosis) and Pet Birds (Avian Chlamydiosis), 2009

**National Association of State Public Health Veterinarians (NASPHV)**

## INTRODUCTION

*Chlamydophila psittaci* is a member of the family *Chlamydiaceae.* Currently there are eight serovars and nine genotypes described which in the future may prove of importance in the epidemiology of the disease in animals and humans[1]. In some cases, these obligate intracellular bacteria can be transmitted from birds to humans. In humans, the resulting infection is referred to as psittacosis (also known as parrot fever and ornithosis). Psittacosis typically causes influenza-like symptoms and can lead to severe pneumonia and nonrespiratory health problems. With appropriate treatment, the disease is rarely fatal. From 2002 to 2007, 91 human cases of psittacosis were reported to the CDC (mean 15, range 12-21)[2]. This is likely an underrepresentation of the actual number as milder cases may not seek medical attention or be reported. Persons at risk include those exposed to pet birds, pigeons, and poultry and in specific occupations such as laboratory and wildlife workers. Human infection can result from brief exposure to infected birds or their contaminated excretions or secretions.

In this compendium, *C psittaci* infection in birds is referred to as avian chlamydiosis. Chlamydial organisms have been isolated from over 460 bird species from 30 orders[3] but are most commonly identified in psittacine (parrot-type) birds, especially cockatiels and budgerigars (also called parakeets or budgies). Among caged, nonpsittacine birds, infection with *Chlamydiaceae* organisms occurs most frequently in pigeons and doves. Avian chlamydiosis can occur but is infrequently diagnosed in canaries and finches.[4] The recommendations in this compendium provide standardized procedures for controlling avian chlamydiosis in the pet bird population, an essential step in efforts to control psittacosis among humans. This compendium is intended to guide public health officials, physicians, veterinarians, the pet bird industry, and others concerned with the control of *C psittaci* infection and the protection of public health.

## INFECTION IN HUMANS (PSITTACOSIS)

### Transmission

The disease resulting from *C psittaci* infection in humans is called psittacosis and most infections are typically acquired from exposure to psittacine birds. Transmission has also been documented from poultry and free-ranging birds, including doves, pigeons, birds of prey and shore birds. Infection with *C psittaci* usually occurs when a person inhales organisms that have been aerosolized from dried feces or respiratory tract secretions of infected birds. Other means of exposure include mouth-to-beak contact and handling infected birds' plumage and tissues. Even brief exposures can lead to symptomatic infection; therefore, certain patients with psittacosis might not recall or report having any contact with birds. Currently, pet birds are thought to pose a low risk to immunocompromized persons.[5] Person-to-person transmission has been suggested but not proven.[6] Standard infection-control precautions are sufficient for humans with psittacosis, and specific isolation procedures (e.g., private room, negative pressure air flow, and masks) are not indicated.

### Clinical Signs and Symptoms

The onset of illness typically follows an incubation period of 5 to 14 days, but longer periods have been reported. The severity of the disease ranges from inapparent illness to systemic illness with severe pneumonia. Before antimicrobial agents were available, 15% to 20% of humans with *C psittaci* infection died. Currently, mortality is extremely rare. Humans with symptomatic infections typically have abrupt onset of fever, chills, headache, malaise, and myalgia. They usually develop a nonproductive cough that can be accompanied by

breathing difficulty and chest tightness. A pulse-temperature dissociation (fever without increased pulse rate), enlarged spleen, and nonspecific rash are sometimes observed. Auscultatory findings may underestimate the extent of pulmonary involvement. Radiographic findings may include lobar or interstitial infiltrates. The differential diagnosis of pneumonia caused by psittacosis includes infection with *Coxiella burnetii, Histoplasma capsulatum*, *Mycoplasma pneumoniae*, *Legionella* spp*, C. pneumoniae* or other *Chlamydiaceae*, and respiratory viruses such as influenza. *Chlamydophila psittaci* can affect organ systems other than the respiratory tract, resulting in endocarditis, myocarditis, hepatitis, arthritis, keratoconjunctivitis, encephalitis, and more recently, ocular adnexa lymphoma.[7]. Severe illness with respiratory failure, thrombocytopenia, and hepatitis has been reported.

**Case Definition**

The CDC and the Council of State and Territorial Epidemiologists (CSTE) have established case definitions for epidemiologic surveillance.[8] These definitions should not be used as the sole criteria for establishing a clinical diagnosis or determining medical management. A patient is considered to have a confirmed case of psittacosis if clinical illness is compatible with psittacosis and the case is laboratory confirmed by one of three methods:

- *C psittaci* is cultured from respiratory secretions,
- there is a 4-fold or greater increase in antibody against *C psittaci* by complement fixation (CF) or microimmunofluorescence (MIF) to a reciprocal titer of ≥32 between paired acute- and convalescent-phase serum samples, or
- there is presence of immunoglobulin M antibodies against *C psittaci* by MIF to a reciprocal titer of ≥16.

A patient is considered to have a probable case of psittacosis if clinical illness is compatible with psittacosis and the patient is epidemiologically linked to a confirmed case (either an avian case or has common exposure with another human case) or the patient has supportive serology (e.g., a single antibody titer of ≥32, detected by CF or MIF in at least one serum sample obtained after onset of symptoms).

**Diagnosis**

Most diagnoses are established by clinical presentation and positive antibodies against C. psittaci in paired sera using MIF. The MIF is more sensitive and specific than the previously used CF tests; however, there is still some cross-reactivity with other chlamydiae, such as *C. pneumoniae*, *C. trachomatis*, and *C. felis*. Acute-phase serum specimens should be obtained as soon as possible after onset of symptoms, and convalescent-phase serum specimens should be obtained at least two weeks after the first specimen. Because antimicrobial treatment can delay or diminish the antibody response, a third serum sample 4-6 weeks after the acute sample might help confirm the diagnosis. Acute and convalescent sera should be tested simultaneously at the same laboratory.

The infectious agent can also be isolated from the patient's sputum, pleural fluid, or clotted blood during acute illness and before treatment with antimicrobial agents; however, culture of *C. psittaci* is performed by few laboratories because of technical difficulty and safety concerns. Certain polymerase chain reaction (PCR) assays can be used to document organism nucleic acid in clinical samples to distinguish *C psittaci* from other chlamydial species and to genotype *C. psittaci* amplicons.[9] Because proper sample collection techniques and handling are critical for obtaining accurate test results, clinical laboratories should be contacted for specifics on specimen submission.

**Laboratories that Test Human Specimens for *Chlamydiaceae***

Information about laboratory testing is available from state public health departments. Few commercial laboratories have the capability to differentiate chlamydial species by MIF. Certain laboratories accept human specimens to confirm *C. psittaci* infection **(**Table 1**)**. Other sources might be available.

**Table 1: Laboratories that test human specimens for *Chlamydophila psittaci***

| Laboratory | Tests Performed | Telephone Number<br>Website |
|---|---|---|
| Focus Diagnostics (Quest subsidiary) Cypress, CA | MIF (IgM, IgA, IgG) Culture | (800) 445-4032<br>www.focusdx.com |
| Laboratory Corp of America Burlington, NC | MIF (IgM, IgG)Culture | (800) 222-7566<br>www.labcorp.com |
| Specialty Labs, Santa Monica, CA | MIF (IgM, IgG, IgA)<br>PCR (pleural fluid only) | (800) 421-4449<br>www.specialtylabs.com |
| Viromed Minnetonka, MN | MIF (IgG, IgM)Culture | (800) 582-0077<br>www.viromed.com |
| Response and Surveillance Laboratory, Respiratory Diseases Branch, CDC Atlanta, GA** | MIF (requires paired sera),PCR, Culture, genotyping (multiple specimen types) | (404) 639-4921 |

*MIF = microimmunofluorescence, PCR = polymerase chain reaction assay
**CDC is a reference laboratory and samples must be submitted through State Health Departments

**Treatment**

Tetracycline antibiotics are the drug of choice for *C. psittaci* infection in humans. Mild to moderate cases can be treated with oral doxycycline (100 mg every 12 hours) or tetracycline hydrochloride (500 mg every six hours) for a minimum of 10 days. Severely ill patients should be treated with IV doxycycline hyclate (4.4mg/kg/day divided into two infusions, maximum 100 mg/dose). Antibiotic therapy should be continued for at least 10-14 days after fever abates. Most *C. psittaci* infections are responsive to antibiotics within 1-2 days, however relapses can occur. Although in-vivo efficacy has not been determined, macrolide antibiotics are considered the best alternative agents in patients for whom tetracyclines are contraindicated (e.g. children <8 years of age, pregnant women, and persons allergic to tetracyclines).[10]

## INFECTION IN BIRDS (AVIAN CHLAMYDIOSIS)

**Transmission**

*C. psittaci* is excreted in the feces and nasal discharges of infected birds. The organism is environmentally labile but can remain infectious for over a month if protected by organic debris (e.g., litter and feces). Some infected birds can appear healthy and shed the organism intermittently. Shedding can be exacerbated by stress factors, including reproductive activities, relocation, shipping, crowding and chilling.

**Clinical signs**

The usual duration between exposure to *C. psittaci* and onset of illness ranges from 3 days to several weeks. However, active disease can appear with no identifiable exposure. Whether the bird has acute or chronic signs of illness or dies depend on the species of bird, virulence of the strain, infectious dose, stress factors, age and extent of treatment or prophylaxis.

Signs of avian chlamydiosis are non-specific and include lethargy, anorexia and ruffled feathers. Other signs include serous or mucopurulent ocular or nasal discharge, diarrhea and excretion of green to yellow-green urates. Severely affected birds may become anorectic and produce sparse, dark green droppings, followed by emaciation, dehydration and death.

**Case Definitions**

Clinical signs may not always be evident in all infected birds. A confirmed case of avian chlamydial infection is defined on the basis of one of the following:

- isolation of *C. psittaci* from a clinical specimen,

- identification of chlamydial antigen by use of immunofluorescence (fluorescent antibody) on the bird's tissues,
- a ≥ 4-fold change in serologic titer in two specimens from the bird obtained at least two weeks apart and assayed simultaneously at the same laboratory, or
- identification of *Chlamydiaceae* within macrophages in smears or tissues (e.g. liver, conjunctival, spleen, respiratory secretions) stained with Gimenez or Macchiavello stain. Clinical signs may not be evident.

A probable case of avian chlamydial infection is defined as compatible illness and one of the following:
- a single high serologic titer as defined by the laboratory in a specimen obtained after onset of signs or
- *Chlamydiaceae* antigen (identified by use of enzyme-linked immunosorbent assay [ELISA], PCR, or fluorescent antibody) in feces, a cloacal swab specimen or respiratory tract or ocular exudates.

A suspected case of avian chlamydial infection is defined as:
- a compatible illness that is not laboratory confirmed but is epidemiologically linked to a confirmed case in a human or bird,
- a bird with no clinical signs and a single high serologic titer or detection of chlamydial antigen,
- compatible illness with positive results from a nonstandardized test or a new investigational test, or
- compatible illness that is responsive to appropriate therapy.

**Diagnosis**

Several diagnostic methods are available for identifying avian chlamydiosis in birds (see Appendix 1)

**Treatment**

Treatment should be supervised by a licensed veterinarian (see Appendix 2).

## PREVENTION AND CONTROL RECOMMENDATIONS

Aviary and pet shop owners are encouraged to implement recommendations such as those described in the Model Aviary Program.[11] Such programs encourage disease prevention and improve animal health and the human-animal bond. To prevent transmission of *C. psittaci* to humans and birds, specific control measures are recommended:

- **Educate persons at risk.** Inform all persons in contact with birds or bird-contaminated materials about the zoonotic nature of the disease. By the time infection is recognized in a group of birds, a critical period for pathogen accumulation and dissemination has already occurred. Bird caretakers with respiratory or influenza-like symptoms should seek medical attention and inform their health care provider about bird contact.
- **Protect persons at risk.** When cleaning cages or handling potentially infected birds, caretakers should wear protective clothing, which includes gloves, eyewear, a disposable surgical cap, and an appropriately fitted respirator[12] with N95 or higher rating. Surgical masks might not be effective in preventing transmission of *C. psittaci*. In addition, necropsies of potentially infected birds should be performed in a biological safety cabinet. Wet the carcass with detergent and water to prevent aerosolization of infectious particles.
- **Maintain accurate records of all bird-related transactions for at least one year to aid in identifying sources of infected birds and potentially exposed persons.** Records should include the date of purchase, species of birds purchased, individual bird identification, source of birds, and any identified illnesses or deaths among birds. In addition, the seller should record the name, address, and telephone number of the customer and individual bird identification (e.g., band or microchip number).
- **Avoid purchasing or selling birds that have signs consistent with avian chlamydiosis.** Signs are nonspecific and may include lethargy, ocular or nasal discharge, diarrhea, ruffled feathers or low body weight.

- **Avoid mixing birds from multiple sources**. To prevent epornitics (disease outbreaks) and pathogen transmission to humans, additional control and prevention methods (e.g. health screening, extended quarantine) may be required when birds from multiple sources are mixed.
- **Quarantine newly acquired or exposed and isolate ill birds.** Isolation should include housing in a separate air space from other birds and noncaretakers. Quarantine birds, including those that have been to shows, exhibitions, fairs, and other events for at least 30 days and test before adding them to a group.
- Test birds before they are to be boarded or sold on consignment. House them in a room separate from other birds pending test results (see Appendix 1).
- **Screen birds with frequent public contact (e.g., bird encounters, long term care facilities, schools)**. Such testing may be used to reduce potential human exposure from birds. Specific protocols should be established in consultation with a veterinarian, recognizing that some birds may demonstrate persistent IgG antibodies in the absence of active infection (see Appendix 1).
- **Practice preventive husbandry.** Position cages to prevent the transfer of fecal matter, feathers, food, and other materials from one cage to another. Do not stack cages and be sure to use solid-sided cages or barriers if cages are adjoining. The bottom of the cage should be made of a wire mesh. Substrate/litter that will not produce dust (e.g., newspapers) should be placed underneath the mesh. Clean all cages, food bowls, and water bowls daily. Soiled bowls should be emptied, cleaned with soap and water, rinsed, placed in a disinfectant solution, and rinsed again before reuse. Between occupancies by different birds, cages should be thoroughly scrubbed with soap and water, disinfected, and rinsed in clean running water. Exhaust ventilation should be sufficient to prevent accumulation of aerosols and prevent cross contamination of rooms.
- **Control the spread of infection.** Isolate birds requiring treatment. Rooms and cages where infected birds were housed should be cleaned immediately and disinfected thoroughly. Workers should wear appropriate protective clothing. When the cage is being cleaned, transfer the bird to a clean cage. Thoroughly scrub the soiled cage with a detergent to remove all fecal debris, rinse the cage, disinfect it (most disinfectants require 5-10 minutes of contact time) and rerinse the cage to remove the disinfectant. Discard all items that cannot be adequately disinfected (e.g., wooden perches, ropes, nest material, substrate/litter). Minimize the circulation of feathers and dust by wet-mopping the floor frequently with disinfectants and preventing air currents and drafts within the area. Reduce contamination from dust by spraying the floor with a disinfectant or water before sweeping it. A vacuum cleaner or pressure washer may aerosolize infectious particles and should be used with caution. Frequently remove waste material from the cage (after moistening the material), and burn or double-bag the waste for disposal. Care for healthy birds before handling isolated or sick birds. There is no documented transmission of *C. psittaci* via ventilation systems from pet bird aviaries or pet stores to humans, nor are there any studies specific for *C. psittaci* viability in these systems. Properly maintained ventilation systems are at low risk of harboring *C. psittaci*.[13] Theoretically, desiccation from forced air movement may reduce viability of the organism. Use of a high efficiency particulate air (HEPA) filter on air system return may be an option to reduce particulate matter in the air.
- **Use disinfection measures.** All surfaces should be thoroughly cleaned of organic debris before disinfection**.** *C. psittaci* is susceptible to many disinfectants and detergents as well as heat; however, it is resistant to acid and alkali. Examples of effective disinfectants include 1:1,000 dilution of quaternary ammonium compounds (e.g., Roccal®, Zephiran®), 1% Lysol® or freshly prepared 1:32 dilution of household bleach (½ cup/gallon). Many disinfectants are respiratory irritants for both humans and birds and should be used in a well-ventilated area. Avoid mixing disinfectants with any other product.

**Recommendations for Treating and Caring for Infected and Exposed Birds**

All birds with confirmed or probable avian chlamydiosis should be isolated and treated, preferably under the supervision of a veterinarian (Appendix 2).

**Responsibilities of bird owners, physicians, and veterinarians**

Humans exposed to birds with avian chlamydiosis should seek medical attention if they develop influenza-like symptoms or other respiratory tract illnesses. The physician should consider psittacosis in ill patients exposed to birds and collect specimens for laboratory analysis if indicated. Psittacosis in humans is a Nationally Notifiable Disease[14] and most states require physicians to report cases of psittacosis to the

appropriate state or local public health authorities. Early and specific treatment for psittacosis should be initiated. Timely diagnosis and reporting can help identify the source of infection and control the spread of disease. Local and state public and/or animal health authorities may conduct epidemiologic investigations and institute additional disease control measures. Birds that are suspected sources of human infection should be referred to veterinarians for evaluation and treatment.

Veterinarians should consider a diagnosis of avian chlamydiosis for any lethargic bird that has nonspecific signs of illness, especially if the bird was recently purchased. If avian chlamydiosis is suspected, the veterinarian should submit appropriate laboratory specimens to confirm the diagnosis. Laboratories and attending veterinarians should follow local and state regulations or guidelines regarding case reporting. Veterinarians should work closely with authorities on investigations and inform clients that infected birds should be isolated and treated. In addition, they should educate clients about the public health hazard posed by *C. psittaci* and the appropriate precautions that should be taken to avoid the risk for transmission.

**Local and state epidemiologic investigations**

Local health authorities should report suspected cases to their state health department.[15] Public health and animal health authorities at the local or state level may need to conduct cooperative epidemiologic investigations to control the transmission of *C psittaci* among humans and birds. An epidemiologic investigation should be initiated if a bird with confirmed or probable avian chlamydiosis was either:

- procured from a pet store, breeder, or dealer within 60 days of the onset of signs of illness;
- linked to a person with confirmed or probable psittacosis; or
- associated with several other suspect avian cases from the same source.

Other situations can be investigated at the discretion of the appropriate local or state public health department or animal health authorities.

Investigations involving recently purchased birds should include a visit to the site where the infected bird is located and identification of the location where the bird was originally procured (e.g., pet shop, dealer, breeder, or quarantine station). Authorities should document the number and types of birds involved, the health status of potentially affected persons and birds, locations of facilities where birds were housed, relevant ventilation-related factors and any treatment protocol.[16] Suspect birds should be tested as recommended (Appendix 1). Examination of sales records for follow up of other birds that had contact with the infected bird may be considered.

**Quarantine of birds**

Depending on the state's regulatory authority, animal or public health officials may issue a quarantine for all affected and exposed birds on premises where *C. psittaci* infection has been identified. The purpose of imposing a quarantine is to prevent further pathogen transmission. Reasonable options should be made available to the owners and operators of pet stores. Preferably, the owner of quarantined birds should treat the birds in a separate quarantine area to prevent exposure to the public and other birds. Alternatively, and with the approval of authorities, the owner can sell the birds after at least 7 days of treatment, provided that the new owner agrees in writing to continue the quarantine and treatment and is informed of the disease hazards. After completion of the treatment or removal of the birds, quarantine can be lifted after the premises are thoroughly cleaned and disinfected. Environmental testing can be valuable in evaluating the effectiveness of cleaning and disinfection. The area can then be restocked with birds.

**Bird importation regulations**

Large-scale commercial importation of psittacine birds from foreign countries ended in 1993 with the implementation of the Wild Bird Conservation Act.[17] Limited importation of personal pets and avicultural specimens is permitted at this time. Illegally imported (smuggled) birds are a potential source of *C. psittaci* infection to domestic birds and people. The United States Department of Agriculture, Animal Plant Health and Inspection Service, Veterinary Services still regulates the legal importation of pet birds to ensure that exotic poultry diseases are not introduced into the United States.[18] These regulations are set forth in the Code of Federal Regulations, Title 9, Chapter 1. Current minimum treatment protocols under these regulations are not always sufficient to resolve infection in all birds.

## Appendix 1

## TESTING METHODS FOR *C. PSITTACI* IN BIRDS

Bacteria are classified as *Chlamydophila psittaci* on the basis of shared biochemical characteristics and genome composition. The individual chlamydial organisms that meet these classification criteria are not identical and represent life forms that have evolved, and continue to evolve, through infection of both ancient and naïve hosts. Diversity in the organism, the level of exposure, and the host response may cause spurious test results in some individual animals.

Diagnosis of avian chlamydiosis can be difficult, especially in the absence of clinical signs. A single testing method might not be adequate. Therefore, use of a combination of culture, antibody-detection and antigen-detection methods is recommended, particularly when only one bird is tested. Although there is no epidemiologic evidence of increased risk to young, elderly, or immunocompromised humans, more rigorous testing should be considered for birds in contact with these individuals. Consultation with an experienced avian veterinarian may help when selecting tests and interpreting results. Because proper sample collection techniques and handling are critical for obtaining accurate test results, clinical laboratories should be contacted for specifics on specimen submission.

**Pathologic diagnosis**

In birds with avian chlamydiosis, cloudy air sacs and enlargement of the liver and spleen may be observed, but no specific gross lesion is pathognomonic. Chromatic or immunologic staining of tissue or impression smears can be used to identify organisms in necropsy and biopsy specimens.

**Bacteriologic culture**

Use of culture is recommended to avoid limitations associated with other tests. Tissue specimens from the liver and spleen are the preferred necropsy specimens. In live birds, combined conjunctival, choanal and cloacal swab specimens or liver biopsy specimens are ideal for diagnosis. Live birds being screened for *C psittaci* might not shed the microorganism daily. Therefore, to optimize recovery, serial fecal specimens should be collected for 3 to 5 consecutive days and pooled for submission as a single sample.

*Chlamydophila* species are obligate intracellular bacteria that must be isolated in tissue culture or embryonating chicken eggs. Specialized laboratory facilities and training are necessary for reliable identification of chlamydial isolates and adequate protection of microbiologists. The diagnostic laboratory should be contacted for specific procedures required for collection and submission of specimens. The proper handling of specimens is critical for maintaining the viability of organisms for culture, and a special transport medium is required. Following collection, specimens should be refrigerated and sent to the laboratory packed in ice but not frozen.

**Tests for antibodies**

A positive serologic test result is evidence that the bird was infected by *Chlamydiaceae* at some point, but it might not indicate that the bird has an active infection. False-negative results can occur in birds that have acute infection when specimens are collected before seroconversion. Treatment with an antimicrobial agent can diminish the antibody response. However, IgG titers may persist following successful treatment.

When specimens are obtained from a single bird, serologic testing is most useful when signs of disease and the history of the flock or aviary are considered and serologic results are compared with white blood cell counts and serum activities of liver enzymes. A >4-fold increase in the titer of paired samples or a combination of a titer and antigen identification is needed to confirm a diagnosis of avian chlamydiosis.

- **Elementary-body agglutination (EBA) -** The elementary body is the infectious form of *C psittaci*. Elementary-body agglutination is commercially available and detects IgM antibodies, an indicator of early infection. Titers >10 in budgerigars, cockatiels, and lovebirds and titers >20 in larger birds are frequently detected in cases of recent infection. However, increased titers can persist after treatment is completed.

- **Indirect Fluorescent Antibody Test (IFA) -** Polyclonal secondary antibody is used to detect host antibodies (primarily IgG). Sensitivity and specificity varies with the immunoreactivity of the polyclonal antibody to various avian species. Low titers may occur because of non-specific reactivity.

- **Complement fixation (CF**) - Direct CF is more sensitive than agglutination methods. False-negative results are possible in specimens from parakeets, young African gray parrots, and lovebirds. High titers can persist after treatment and complicate interpretation of subsequent tests. Modified direct CF is more sensitive than direct CF.

**Tests for antigen**

Tests for antigen detect the organism. These tests give rapid results and do not require live, viable organisms; however, false-positive results from cross-reacting antigens can occur. False-negative results can occur if there is insufficient antigen or if shedding is intermittent. As with all nonculture tests, results must be evaluated in conjunction with clinical findings.

- **Enzyme-Linked Immunosorbent Assay** (**ELISA) -** ELISA tests were originally developed for identification of *Chlamydia trachomatis* in humans. The exact sensitivity and specificity of these tests for identifying other *Chlamydiaceae* are not known. They are now occasionally used to identify suspected *C. psittaci* in birds. If a bird has a positive ELISA result but is healthy, the veterinarian should attempt to verify that the bird is shedding antigen via isolation of the organism. When a clinically ill bird has a negative ELISA result, a diagnosis of avian chlamydiosis cannot be excluded without further testing (e.g., culture, serologic testing or polymerase chain reaction [PCR] assay).

- **Fluorescent Antibody Test (FA) -** Monoclonal or polyclonal antibodies, fluorescein staining techniques and fluorescent microscopy are used to identify the organism in impression smears or other specimens. These tests have similar advantages and disadvantages to ELISA. This test is utilized by some state diagnostic laboratories.

**Tests for DNA**

Numerous laboratories offer diagnostic testing using polymerase chain reaction assay (PCR). PCR amplification can be sensitive and specific for detection of target DNA sequences in collected specimens (e.g., combined conjunctival, choanal and cloacal swab specimens and blood). Results differ between laboratories because there are no standardized PCR primers and laboratory techniques and sample handling may vary. Because of the sensitivity of the assay, samples for PCR must be collected using techniques to avoid contamination from the environment or other birds. PCR does not differentiate between viable and nonviable microorganisms. Test results must be interpreted in light of clinical presentation and other laboratory tests.

**Additional tests**

Additional diagnostic techniques are in use or under development. Readers are encouraged to research peer-reviewed reports on such tests before use.

**Laboratories that test avian specimens for *C psittaci***

Table 2 lists government and university laboratories that perform chlamydial diagnostic tests. There are numerous private laboratories that provide similar services. Inclusion in Table 2 does not imply endorsement by the National Association of State Public Health Veterinarians or constituent institutions.

**Table 2: Laboratories that test avian specimens for Chlamydiaceae**

| Laboratory | Tests Performed* | Telephone Number<br>Website |
|---|---|---|
| Diagnostic Center for Population and Animal Health, Michigan State University, East Lansing, MI | Culture, PCR | (517) 353-2296<br>www.dcpah.msu.edu |
| Comparative Pathology Laboratory, University of Miami, Miami, FL | ELISA (antigen), IFA PCR | (305)585-6303<br>www.pathology.med.miami.edu |
| Infectious Diseases Laboratory (IDL), University of Georgia College of Veterinary Medicine, Athens, GA | Culture PCR, IFA | (305) 585-6303<br>www.vet.uga.edu/sams/idl |
| Veterinary Medical Diagnostic Laboratory (VMDL) College Station, TX | Culture, PCR, DCF | (979) 845-3414<br>http://tvmdlweb.tamu.edu/ |
| Diagnostic Virology Lab, National Veterinary Service Laboratory (NVSL), VS, APHIS,USDA, Ames IA** | CF, Culture | (515) 663-7551<br>http://www.aphis.usda.gov/animal_health/lab_info_services/about_dvl.shtml |

*CF = Complement fixation, EBA = Elementary body agglutination, ELISA= Enzyme-linked immunosorbent assay, IFA = Immunofluorescent antibody, PCR =Polymerase chain reaction assay, DCF – Direct compliment fixation, DCF = Direct florescent antibody

**NVSL is a USDA reference laboratory and samples must be submitted through State Veterinary Diagnostic Laboratories

## Appendix 2

## TREATMENT OPTIONS FOR BIRDS WITH AVIAN CHLAMYDIOSIS

Routine prophylactic antibiotic treatment is highly discouraged as it may cause adverse affects and could generate resistant strains of *C. psittaci* and other bacteria

Treatment of avian chlamydiosis can be difficult. Although treatment protocols are usually successful, knowledge is evolving and no protocol ensures safe treatment or complete elimination of infection in every bird. Therefore, treatment for avian chlamydiosis should be supervised by a licensed veterinarian after consultation with an experienced avian veterinarian.

### General Recommendations for Treating and Caring for Infected and Exposed Birds

- The recommended treatment period for avian chlamydiosis has historically been 45 days, except in budgerigars where 30 days of treatment can be effective.
- Avian facility managers should provide employees with simple, concise written treatment procedures to assure treatment success.
- Sick birds may consume inadequate amounts of medicated food or water, so they should be initially treated with drugs delivered directly by mouth or injection.
- Protect birds from undue stress (e.g., chilling, relocation), poor husbandry, and malnutrition. These problems reduce the effectiveness of treatment and promote the development of secondary infections with other bacteria or yeast.
- Observe the birds daily, and weigh them every 3 to 7 days. If the birds are not maintaining weight, have them reevaluated by a veterinarian.
- Remove oyster shell, mineral blocks, and cuttlebone during treatment. High dietary concentrations of calcium and other minerals inhibit the absorption of tetracyclines. In hand-fed neonates where dietary calcium is required, the calcium and tetracycline should be given at least 4 to 6 hours apart
- Isolate birds that are to be treated in clean and uncrowded cages.
- Good husbandry practices should be followed to prevent opportunistic infections:
    - clean up all spilled food promptly
    - wash food and water containers daily.
    - provide appropriate vitamins daily.
- Continue medication for the full treatment period to avoid incomplete resolution of the infection.
- Birds may have reduced chlamydial shedding within days of treatment initiation.
- Treated birds can be reinfected; therefore contaminated aviaries should have a final thorough cleaning and disinfection several days before treatment ends.
- Post-treatment testing should be conducted no sooner than two weeks after treatment is completed.

### Treatment Using Doxycycline

Doxycycline is presently the drug of choice for treating birds with avian chlamydiosis. It is better absorbed and more slowly eliminated than other tetracyclines. This allows doxycycline to be effective with lower drug doses (improving palatability with food or water-based administration) or administered less frequently (improving ease of treatment). Treated birds should be monitored for signs of doxycycline toxicosis. Toxicosis can cause general signs of illness (signs of depression, inactivity and decreased appetite), green- or yellow-stained urine and altered results of hepatic tests (high serum activities of aspartate aminotransferase and lactate dehydrogenase and high serum concentration of bile acids). If toxicosis occurs, administration should be stopped and supportive care provided until the bird recovers. Treatment with a different regimen or lower doxycycline dose can be started at a later date. Below are several options for treatment. Options should not be combined in the same day.

- **Doxycycline medicated feed for budgerigars and cockatiels**—It is critical to use the recommended doxycycline formulation and dietary ingredients to achieve safe and effective results. The following medicated diet[19] can be used to treat avian chlamydiosis:

    1. Mix 1 part cracked steel-cut oats with 3 parts hulled millet (measured by volume).
    2. To each kilogram of oat-seed mixture, add 5 to 6 mL of sunflower oil. Mix thoroughly to coat all seeds.
    3. Add 300 mg of doxycycline hyclate (from capsules) per kilogram of oat-seed-oil mixture, and mix thoroughly to ensure that oats and seeds are evenly coated.

    Prepare fresh medicated oat-seed-oil mixture daily because doxycycline stability in this diet is unknown. Feed as the sole diet. The oats and hulled millet seed are available at health food stores. Small-sized millet should be selected. Sunflower oil is available in grocery stores. Doxycycline hyclate capsules are available in 50- and 100-mg sizes.

- **Doxycycline medicated water**—Results of pharmacologic studies indicate that doses of 200 to 400 mg of doxycycline hyclate/L of water for cockatiels, 400 to 600 mg/L for Goffin's cockatoos, and 800 mg/L for African gray parrots will maintain therapeutic concentrations.[20,21] Research data are lacking for other species, but empiric use of 400 mg/L of water has been successful for many psittacine birds. Medicated water should be prepared daily and provided in clean bowls, rather than water bottles. Do not use medicated water for budgerigars as it will not maintain therapeutic concentrations.[16]

- **Orally administered doxycycline**—Doxycycline is the drug of choice for oral administration; either the monohydrate or calcium-syrup formulations can be used. Dosage recommendations are as follows: 25 to 35 mg/kg every 24 hours for cockatiels, 25-50 mg/kg for Senegal parrots, blue-fronted and orange-winged Amazon parrots; and 25 mg/kg every 24 hours for African gray parrots, Goffin's cockatoos, blue and gold macaws and green-winged macaws. Precise dosages cannot be extrapolated for other species; however, 25 to 30 mg/kg every 24 hours is the recommended starting dosage for cockatoos and macaws, and 25 to 50 mg/kg every 24 hours is recommended for other psittacine species. If the bird regurgitates or refuses the drug, another treatment method should be used.

- **Injectable doxycycline**— The only suitable doxycycline formulation for intramuscular injection is Vibramycin SF IV[a] (Vibrovenos®), [a] a specific European formulation that can be imported into the U.S. (Table 3). It is effective if administered at doses of 75 to 100 mg/kg, IM, every 5 to 7 days for the first 4 weeks and subsequently every 5 days for the duration of treatment. This formulation can cause irritation at the injection site, but it is usually tolerated. Other injectable doxycycline hyclate formulations may cause severe tissue reactions if given IM.

**Alternative treatment regimens**

- **Injectable Oxytetracycline** - Limited information exists to guide the use of an injectable, long-acting oxytetracycline product LA-200.[b] Current dosage recommendations are as follows: SC injection of 75 mg/kg every 3 days in Goffin's cockatoos, blue-fronted and orange-winged Amazon parrots, and blue and gold macaws. [22] This dosage might be suitable for other species but has not been tested. This product causes irritation at the site of injection and is best used to initiate treatment in ill birds or those that are reluctant to eat. After stabilization with oxytetracycline treatment, the birds should receive another form of treatment to reduce the irritation that is caused by repeated oxytetracycline injection.

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a. Pfizer Laboratories, London, England
b. Pfizer Laboratories, Exton, Penn.

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- **Chlortetracycline (CTC) Medicated Feed** - Chlortetracycline medicated feed has historically been used for flock treatment, however doxycycline regimens are preferred. If used, CTC medicated feed should be the only food provided to the birds during the entire treatment. Birds' acceptance of medicated feed is variable. Thus, food consumption should be monitored. Acceptance can be enhanced by first adapting the birds to a similar, nonmedicated diet. Treatment begins when the birds accept the medicated feed as the sole food in their diet. The following options are available:
  - Medicated mash diets (i.e., >1% CTC with <0.7% calcium) prepared with corn, rice, and hen's scratch.[23]
  - Pellets and extruded products containing 1% CTC can be used. They are available and appropriate for use with pet birds. Select a pellet size appropriate for the size of bird being treated.[24, 25]
  - A special diet might be necessary for lories and lorikeets, which feed on nectar and fruit in the wild.[26]

**Treatment Methods Not Recommended**

Use of water medicated with chlortetracycline (Aureomycin), oxytetracycline (Terramycin) or other tetracycline products (except doxycycline) is not recommended. These products may reduce water consumption, are not likely to be effective and may interfere with disease testing.

**Sources of Medications**

The following sources (Table 3) are not listed as an endorsement of the companies or products. Other sources might be available.

**Table 3: Sources of medication for avian chlamydiosis**

| Contact | Product | Telephone Number<br>Website |
|---|---|---|
| | **DOXYCYCLINE** | |
| Local pharmacies | 1.Docycycline hyclate capsules 50 & 100 mg<br>2. Doxycycline calcium oral suspension<br>3. Doxycycline monohydrate oral suspension<br>4. Vibramycin 50 & 100 mg capsules | |
| Dr. Gerry M. Dorrestein<br>Wilhelminalaan 19A<br>5512BJ Vessem<br>The Netherlands | Vibramycin SF I.V.* | Tel: 000 316 11057602<br>Fax: 000 313 02533131<br>dorresteingm@planet.nl |
| | **MEDICATED FEED** | |
| Avi-Sci Inc., St. Johns, MI | Chlortetracycline, 1% | Tel: 800.942.3438<br>mike@avi-sci.com |
| Roudybush, Paso Robles, CA | Chlortetracycline, 1% | Tel: 800.326.1726<br>www.roudybush.com |
| Ziegler Brothers Inc.<br>Gardners, PN | Chlortetracycline, 1%<br>(special order, 50# minimum) | Tel: 800.841.6800<br>www.zeiglerfeed.com |
| | **CHLORTETRACYCLINE POWDER** | |
| Fort Dodge Animal Health<br>Fort Dodge, IA | Aureomycin, (chlortetracycline hydrochloride) soluble powder concentrate, 4 oz packets | Tel: 800.685.5656<br>fdorder@FDAH.com |
| Phibro Animal Health<br>Fairfield, NJ | CLTC 100 (Chlortetracycline hydrochloride), 22% (100 gm/lb) | Tel: 888.403.0074<br>www.phibroah.com |
| Agrilaboratories Inc.<br>St. Joseph, MO | CTC (Chlortetracycline hydrochloride), soluble powder, 25.6 oz. packet (102 g CTC) | Tel: 800.542.8916<br>www.agrilabs.com |
| | **TETRACYCLINE POWDER** | |
| Agrilaboratories Inc.<br>St. Joseph, MO | Tetra – bac 324 (Tetracycline hydrochloride), soluble powder, 324 g/lb concentration | Tel: 800.542.8916<br>www.agrilabs.com |
| | **OXYTETRACYCLINE POWDER** | |
| Agrilaboratories Inc.<br>St. Joseph, MO | Agrimycin – 343 (Oxytetracycline hydrochloride), soluble powder, 343 g/lb concentration | Tel: 800.542.8916<br>www.agrilabs.com |

*Investigational New Animal Drug Application (INADA) is no longer required, Contact the FDA at (301) 594-0796 about obtaining a personal import letter.

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[15] Sample Human Psittacosis Case Report http://www.nasphv.org/documentsCompendia.html

[16] Sample Avian Chlamydiosis Case Report; http://www.nasphv.org/documentsCompendia.html

[17] Wild Bird Conservation Act of 1992, Title I of PL 102–440. 16 US Code 4901–4916. Available at: http://international.fws.gov/permits/web%20list%20wbca.htm.

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## Additional Resources

### General Public (Fact Sheets)

Medline Medical Encyclopedia: Psittacosis; US National Library of Medicine and National Institute of Health
http://www.nlm.nih.gov/medlineplus/ency/article/000088.htm

Psittacosis, Technical Information; Center for Disease Control and Prevention
http://www.cdc.gov/ncidod/dbmd/diseaseinfo/psittacosis_t.htm

Psittacosis in Birds and People Public Health Fact Sheet; Massachusetts Department of Public Health
http://www.mass.gov/Eeohhs2/docs/dph/cdc/factsheets/psittacosis_birds_people.pdf

Psittacosis Fact Sheet: Public Health: Seattle and King Counties
**http://www.kingcounty.gov/healthservices/health/communicable/diseases/psittacosis.aspx**

### Medical and Public Health Professionals

Ohio Dept. of Health Infectious Disease Control Manual http://www.odh.ohio.gov/pdf/IDCM/psitta.pdf

Psittacosis Control Guidelines for Local Health Departments; Virginia Dept of Health
http://www.vdh.virginia.gov/epidemiology/DEE/otherzoonosis/documents/Psittacosis/Psittacosis%20for%20LHD%20revApr06%20e.pdf

WebMD's "emedicine" site http://www.emedicine.com/med/topic1951.htm

**Occupational Health and Safety**

Hazard Information Bulletin on psittacosis; Occupational Safety and Health Association
http://www.osha.gov/dts/hib/hib_data/hib19940808.html

Safety Services; Occupational Health; Animal Care and Use Occupational Health Program, University of California, Davis http://safetyservices.ucdavis.edu/occupational-health-services/acu/educational-materials/psittacosis/?searchterm=psittacosis

Psittacosis: Bioterrorism Agent Profiles for Health Care Workers; Arizona Department of Health http://www.azdhs.gov/phs/edc/edrp/es/pdf/psittacosisset.pdf

Psittacosis; Canadian Center for Occupational Health and Safety http://www.ccohs.ca/oshanswers/diseases/psittacosis.html

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Psittacosis; State of New Jersey Department Of Agriculture http://www.state.nj.us/agriculture/divisions/ah/diseases/psittacosis.html

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