VIVIAN LINDMAYER FERREIRA

Epidemiology of *Chlamydia psittaci* in pet birds associated with psittacosis cases in humans

São Paulo

2016

VIVIAN LINDMAYER FERREIRA

Epidemiology of *Chlamydia psittaci* in pet birds associated with psittacosis cases in humans

Tese apresentada ao Programa de Pós-Graduação em Patologia Experimental e Comparada da Faculdade de Medicina Veterinária e Zootecnia da Universidade de São Paulo para obtenção do título de Doutor em Ciências

Departamento:

Patologia

Área de Concentração:

Patologia Experimental e Comparada

Orientador:

Profa. Dra. Tânia de Freitas Raso

São Paulo

2016

DADOS INTERNACIONAIS DE CATALOGAÇÃO NA PUBLICAÇÃO

(Biblioteca Virginie Buff D'Ápice da Faculdade de Medicina Veterinária e Zootecnia da Universidade de São Paulo)

T.3384
FMVZ

Ferreira, Vivian Lindmayer
Epidemiology of *Chlamydia psittaci* in pet birds associated with psittacosis cases in humans / Vivian Lindmayer Ferreira. -- 2016.
74 f. : il.

Título traduzido: Epidemiologia da *Chlamydia psittaci* em aves de companhia associada aos casos de psitacose em humanos.

Tese (Doutorado) - Universidade de São Paulo. Faculdade de Medicina Veterinária e Zootecnia. Departamento de Patologia, São Paulo, 2016.

Programa de Pós-Graduação: Patologia Experimental e Comparada.

Área de concentração: Patologia Experimental e Comparada.

Orientador: Profa. Dra. Tânia de Freitas Raso.

1. Clamidiose aviária. 2. Psitacose. 3. Vigilância epidemiológica. 4. Zoonose. I. Título.

UNIVERSIDADE DE SÃO PAULO

FACULDADE DE MEDICINA VETERINÁRIA E ZOOTECNIA

Comissão de Ética no uso de animais

CERTIFICADO

Certificamos que o Projeto intitulado "Epidemiologia da *Chlamydophila psittaci* em aves de companhia associada aos casos de psitacose em humanos", protocolado sob o nº2870, utilizando material biológico de aves silvestres, sob a responsabilidade da Profa. Dra. Tânia de Freitas Raso, está de acordo com os princípios éticos de experimentação animal da "Comissão de Ética no uso de animais" da Faculdade de Medicina Veterinária e Zootecnia da Universidade de São Paulo e foi aprovado em reunião do dia seis de fevereiro de 2013.

We certify that the Research "Epidemiology of *Chlamydophilia psittaci* in pet birds associated with psittacosis cases in humans", protocol number 2870, under the responsibility Profa. Dra. Tânia de Freitas Raso, agree with Ethical Principles in Animal Research adopted by "Ethic Committee in the use of animals" of the School of Veterinary Medicine and Animal Science of University of São Paulo and was approved at the meeting on 6 February of two thousand and thirteen.

São Paulo, 14 de fevereiro de 2013.

Denise Tabacchi Fantoni
Presidente

Av. Prof. Dr. Orlando Marques de Paiva, nº87
Cidade Universitária "Armando de Salles Oliveira"
São Paulo/SP – Brasil
05508-270

Fone: + 55 11 3091-7671/7676
Fax: +55 11 3032-2224
E-mail: fmvz@usp.br
http://www.fmvz.usp.br

Ministério do Meio Ambiente - MMA
Instituto Chico Mendes de Conservação da Biodiversidade - ICMBio
Sistema de Autorização e Informação em Biodiversidade - SISBIO

Autorização para atividades com finalidade científica

Número: 37364-4	Data da Emissão: 29/02/2016 08:31	Data para Revalidação*: 30/03/2017

* De acordo com o art. 28 da IN 03/2014, esta autorização tem prazo de validade equivalente ao previsto no cronograma de atividades do projeto, mas deverá ser revalidada anualmente mediante a apresentação do relatório de atividades a ser enviado por meio do Sisbio no prazo de até 30 dias a contar da data do aniversário de sua emissão.

Dados do titular

Nome: TÂNIA DE FREITAS RASO	CPF: 737.841.846-34
Título do Projeto: Epidemiologia da Chlamydophila psittaci em aves de companhia associada aos casos de psitacose em humanos	
Nome da Instituição : Faculdade de Medicina Veterinária e Zootecnia USP	CNPJ: 63.025.530/0019-33

Cronograma de atividades

#	Descrição da atividade	Início (mês/ano)	Fim (mês/ano)
1	COLETA E TRANSPORTE DE MATERIAL BIOLÓGICO DE AVES SILVESTRES E EXÓTICAS	03/2013	03/2016

Observações e ressalvas

1	As atividades de campo exercidas por pessoa natural ou jurídica estrangeira, em todo o território nacional, que impliquem o deslocamento de recursos humanos e materiais, tendo por objeto coletar dados, materiais, espécimes biológicos e minerais, peças integrantes da cultura nativa e cultura popular, presente e passada, obtidos por meio de recursos e técnicas que se destinem ao estudo, à difusão ou à pesquisa, estão sujeitas a autorização do Ministério de Ciência e Tecnologia.
2	Esta autorização NÃO exime o pesquisador titular e os membros de sua equipe da necessidade de obter as anuências previstas em outros instrumentos legais, bem como do consentimento do responsável pela área, pública ou privada, onde será realizada a atividade, inclusive do órgão gestor de terra indígena (FUNAI), da unidade de conservação estadual, distrital ou municipal, ou do proprietário, arrendatário, posseiro ou morador de área dentro dos limites de unidade de conservação federal cujo processo de regularização fundiária encontra-se em curso.
3	Este documento somente poderá ser utilizado para os fins previstos na Instrução Normativa ICMBio nº 03/2014 ou na Instrução Normativa ICMBio nº 10/2010, no que especifica esta Autorização, não podendo ser utilizado para fins comerciais, industriais ou esportivos. O material biológico coletado deverá ser utilizado para atividades científicas ou didáticas no âmbito do ensino superior.
4	A autorização para envio ao exterior de material biológico não consignado deverá ser requerida por meio do endereço eletrônico www.ibama.gov.br (Serviços on-line - Licença para importação ou exportação de flora e fauna - CITES e não CITES).
5	O titular de licença ou autorização e os membros da sua equipe deverão optar por métodos de coleta e instrumentos de captura direcionados, sempre que possível, ao grupo taxonômico de interesse, evitando a morte ou dano significativo a outros grupos; e empregar esforço de coleta ou captura que não comprometa a viabilidade de populações do grupo taxonômico de interesse em condição in situ.
6	O titular de autorização ou de licença permanente, assim como os membros de sua equipe, quando da violação da legislação vigente, ou quando da inadequação, omissão ou falsa descrição de informações relevantes que subsidiaram a expedição do ato, poderá, mediante decisão motivada, ter a autorização ou licença suspensa ou revogada pelo ICMBio, nos termos da legislação brasileira em vigor.
7	Este documento não dispensa o cumprimento da legislação que dispõe sobre acesso a componente do patrimônio genético existente no território nacional, na plataforma continental e na zona econômica exclusiva, ou ao conhecimento tradicional associado ao patrimônio genético, para fins de pesquisa científica, bioprospecção e desenvolvimento tecnológico. Veja maiores informações em www.mma.gov.br/cgen.
8	Em caso de pesquisa em UNIDADE DE CONSERVAÇÃO, o pesquisador titular desta autorização deverá contactar a administração da unidade a fim de CONFIRMAR AS DATAS das expedições, as condições para realização das coletas e de uso da infra-estrutura da unidade.

Equipe

#	Nome	Função	CPF	Doc. Identidade	Nacionalidade
1	VIVIAN LINDMAYER FERREIRA	Coleta, transporte e processamento do material biológico	281.964.788-01	297500843 SSPSP-SP	Brasileira

Locais onde as atividades de campo serão executadas

#	Município	UF	Descrição do local	Tipo
1	SAO PAULO	SP	Estado de São Paulo	Fora de UC Federal

Atividades X Táxons

#	Atividade	Táxons
1	Coleta/transporte de amostras biológicas ex situ	Passeriformes, Psittaciformes, Columbiformes

Este documento (Autorização para atividades com finalidade científica) foi expedido com base na Instrução Normativa nº 03/2014. Através do código de autenticação abaixo, qualquer cidadão poderá verificar a autenticidade ou regularidade deste documento, por meio da página do Sisbio/ICMBio na Internet (www.icmbio.gov.br/sisbio).

Código de autenticação: 94689979

PROTOCOLO DE PESQUISA Nº 01/2014
TÍTULO: "CHLAMYDOPHILA PSITTACI EM AVES E HUMANOS: ESTUDO DE UMA ZOONOSE EMERGENTES"
PESQUISADOR RESPONSÁVEL NO IIER: MARCOS VINICIUS DA SILVA
COLABORADORES : TÂNIA DE FREITAS RASO, VIVIAN LINDMAYER FERREIRA, BIL RANDERSON BASSETTI, MARILEIDE JANUÁRIA DE VASCONCELOS.

AUTORIZAÇÃO PARA INÍCIO DO ESTUDO

Devidamente aprovado pela Comissão Científica, Comitê de Ética em Pesquisa e Diretoria Técnica de Departamento deste Instituto, e regular quanto às informações sobre financiamento do projeto, o protocolo de pesquisa acima está **AUTORIZADO** para ter início.

Registre-se. Comunique-se.

São Paulo, 23 de maio de 2014.

Profa. Dra. Maria Aparecida Telles Guerra
Chefe De Saúde II da Seção de Pesquisas e Trabalhos Científicos

FOLHA DE AVALIAÇÃO

Autor: FERREIRA, Vivian Lindmayer

Título: Epidemiology of *Chlamydia psittaci* in pet birds associated with psittacosis cases in humans

Tese apresentada ao Programa de Pós-Graduação em Patologia Experimental e Comparada da Faculdade de Medicina Veterinária e Zootecnia da Universidade de São Paulo para obtenção do título de Doutor em Ciências

Data: ____/____/____

Banca Examinadora

Prof. Dr. ____

Instituição: ______________________________ Julgamento: ___________________

Prof. Dr. ____

Instituição: ______________________________ Julgamento:______________________

Prof. Dr. ____

Instituição: ______________________________ Julgamento:______________________

Prof. Dr. ____

Instituição: ______________________________ Julgamento:______________________

Prof. Dr. ____

Instituição: ______________________________ Julgamento:______________________

AGRADECIMENTOS

- À Irene Lindmayer e Roberto Eugênio Lindmayer (*in memoriam*), por estarem sempre ao meu lado, pelos valores que me transmitiram e por me ensinarem a ser fiel a eles.

- Ao meu marido André Luiz Cisi, pela cumplicidade e companheirismo, pelos sorrisos largos nas horas agradáveis e pelo incentivo nas horas difíceis. Amo você com todo meu coração!

- À Gabriela e Bruno Cisi, por me incentivarem a ser cada dia melhor.

- À toda minha querida família Lindmayer pelo apoio incondicional e compreensão pelos momentos que não pude estar presente durante o desenvolvimento deste trabalho.

- À Mônica Lindmayer e Laércio Veloso por estarem sempre por perto. À Mônica pelas incontáveis vezes em que me auxiliou em traduções/correções de manuscritos, documentos de intercâmbio em alemão e inglês; inclusive na revisão desta tese.

- À professora e orientadora Tânia de Freitas Raso pela confiança e ensinamentos ao longo do desenvolvimento deste trabalho. Confesso que não somente durante o desenvolvimento desta tese; foram 7 anos de aprendizados, grandes desafios, muitas lutas, e também grandes conquistas!

- Ao professor Fábio Gregori por abrir meus olhos para o mundo da Educação, por me mostrar o universo cativante que é a docência e as práticas de ensino/aprendizagem.

- Ao professor Dr. Marcus Vinicius da Silva e toda à equipe do Instituto de Infectologia Emílio Ribas pela parceria e oportunidade de pesquisa.

- À Fabiola Setim Prioste pela amizade sincera, por sempre estar presente nos momentos importantes!

- À Faculdade de Medicina Veterinária e Zootecnia da Universidade de São Paulo (FMVZ/USP) e ao departamento de Patologia, por ter sido minha casa nesses anos de pós-graduação.

- Ao Sátiro Márcio do Centro de Informações Estratégicas em Vigilância em Saúde – CIEVS pelo auxílio na análise dos resultados.

- À Fundação de Amparo à Pesquisa do Estado de São Paulo (FAPESP) pela bolsa de doutorado (Processo 2012/25067-8), e pelos demais auxílios financeiros concedidos para a realização deste trabalho e estágio no exterior (Processos 2012/25043-1, 2015/03811-5).

- A todos aqueles que de alguma forma contribuíram para a realização dessa pesquisa, ninguém constrói nada sozinho e fica difícil mencionar todas as pessoas que me marcaram nessa caminhada, foram tantos os aprendizados! Sinto uma profunda gratidão por todos aqueles que participaram da minha formação, e apesar do caminho longo que tenho pela frente, certamente saí muito mais rica em experiências profissionais e pessoais, do que quando comecei essa jornada! Muito obrigada!

“Há perguntas a serem feitas insistentemente por todos nós e que nos fazem ver a impossibilidade de *estudar por estudar*. De *estudar* descomprometidamente como se misteriosamente, de repente, nada tivéssemos que ver com o mundo, um lá fora e distante mundo, alheado de nós e nós dele.

Em favor de *que* estudo? Em favor de *quem*? Contra *que* estudo? Contra *quem* estudo? ”

Paulo Freire

RESUMO

FERREIRA, V. L. **Epidemiologia da *Chlamydia psittaci* em aves de companhia associada aos casos de psitacose em humanos.** [Epidemiology of *Chlamydia psittaci* in pet birds associated with psittacosis cases in humans]. 2016. 74 f. Tese (Doutorado em Ciências) - Faculdade de Medicina Veterinária e Zootecnia, Universidade de São Paulo, São Paulo, 2016.

As zoonoses representam a maior parte das doenças infecciosas emergentes, as quais tem ocorrência variável de acordo com fatores biológicos, ambientais e sócio-econômico-culturais. No que tange aos fatores sócio-culturais, uma prática crescente no Brasil é a manutenção de espécies silvestres como animais de estimação. Estes podem ter significante papel na disseminação de agentes patogênicos com potencial zoonótico, tal como *Chlamydia psittaci*, agente etiológico da clamidiose em aves e da psitacose em seres humanos. Os Psittaciformes representam a principal Ordem de aves acometida pela *C. psittaci*, sendo também a mais comumente mantida como pet. A clamidiose aviária é endêmica no Brasil, contudo, são raros os estudos direcionados a avaliação do seu potencial zoonótico. Em humanos a psitacose pode desencadear um quadro severo de pneumonia atípica, no entanto, devido à dificuldade relacionada ao diagnóstico laboratorial e pelo relativo desconhecimento da doença pelos profissionais de saúde, sua prevalência no país é ainda desconhecida. Dentro desse contexto, o presente trabalho teve como objetivo determinar a ocorrência da *C. psittaci* em pacientes suspeitos de psitacose atendidos no Ambulatório de doenças tropicais e zoonoses do Instituto de Infectologia Emílio Ribas (IIER); estabelecer o vínculo epidemiológico com aves realizando o diagnóstico nestas, assim como avaliar os fatores de risco relacionados com essa zoonose. Para tanto, amostras de sangue de pacientes com quadros suspeitos de psitacose foram coletadas para a investigação de anticorpos anti-*C. psittaci* IgA, IgM e IgG. Paralelamente, amostras biológicas de quaisquer espécies de aves relacionadas com os casos suspeitos de psitacose foram coletadas para a pesquisa molecular de *C. psittaci*. Entre os pacientes elegíveis deste estudo, 27% (10/37) foram classificados como casos confirmados de psitacose; 13,5% (5/37) como prováveis e 59,5% (22/37) como descartados. Pneumonia ($p = 0.004$), tosse ($p = 0.002$) e calafrio ($p = 0.011$) foram estatisticamente significantes quando comparado com os pacientes nos quais a psitacose foi descartada. Quanto ao vínculo epidemiológico com aves, 73% (11/15) dos casos prováveis e confirmados de psitacose relataram exposição domiciliar com aves e em 27% (4/15) a exposição foi ocupacional. Adicionalmente, 47% (7/15) dos pacientes tiveram contato com aves nas quais a infecção por *C. psittaci* foi comprovada laboratorialmente. Em 47% (7/15) dos casos não foi possível obter material biológico das aves relacionadas com os casos e em 6% (1/15) dos casos *C. psittaci* não foi detectada nas aves avaliadas. Ainda, os casos prováveis e confirmados de psitacose relataram manter contato próximo com suas aves, como pega-lás na mão (100%, 15/15), mantê-las no ombro (67%, 10/15), beijá-las (40%, 6/15) e dividir alimento com elas (13%, 2/15). Ressalta-se que essas práticas facilitam a transmissão do patógeno. Profissionais da saúde tanto humana quanto animal têm um papel importante a desempenhar na identificação de fatores que afetam a saúde de seus pacientes e devem, portanto, trabalhar juntos. Esforços mútuos contribuiriam no conhecimento de doenças com potencial zoonótico e certamente contribuiriam para medidas mais eficazes de prevenção e controle.

Palavras chave: Clamidiose aviária. Psitacose. Vigilância epidemiológica. Zoonose.

ABSTRACT

FERREIRA, V. L. Epidemiology of *Chlamydia psittaci* in pet birds associated with psittacosis cases in humans. [**Epidemiologia da *Chlamydia psittaci* em aves de companhia associada aos casos de psitacose em humanos**]. 2016. 74 f. Tese (Doutorado em Ciências) - Faculdade de Medicina Veterinária e Zootecnia, Universidade de São Paulo, São Paulo, 2016.

Zoonosis represent the majority of emerging infectious diseases, which have variable occurrence according to biological, environmental, socio-economic and cultural factors. With respect to socio-cultural factors, a growing practice in Brazil is keeping exotic animals as pets. These can have significant role in the spread of pathogens with zoonotic potencial, such as *Chlamydia psittaci*, etiologic agent of chlamydiosis in birds and psittacosis in humans. The Psittaciformes are the main order of birds affected by *C. psittaci* and is the most commonly kept as a pet. Avian chlamydiosis is endemic in Brazil; however, few studies have been conducted regarding its zoonotic potential. In human, psittacosis can lead to atypical pneumonia, however, due to difficulties related to laboratory diagnosis and the relative lack of knowledge by health professionals about this disease, its prevalence in the country is still unknown. In this context, this study aimed to determine the occurrence of *C. psittaci* in patients with psittacosis symptoms attended at the Ambulatory of Tropical Diseases and Zoonosis of the Infectology Institute Emilio Ribas (IIER). In addition, establishing the epidemiological link with birds and evaluate risk factors related to this zoonosis. Therefore, serum samples from eligible patients were collected in order to be tested for *C. psittaci* IgA, IgM and IgG antibodies. At the same time, biological samples from any species of birds related to suspected cases of psittacosis were collected for molecular analysis of *C. psittaci*. Among the eligible patients in this study, 27% (10/37) were classified as confirmed cases of psittacosis; 13.5% (5/37) as probable and 59.5% (22/37) as discarded. Pneumonia ($p = 0.004$), cough ($p = 0.002$) and chills ($p = 0.011$) were statistically significant when comparing with those patients in which psittacosis was discarded. With reference to the epidemiological link with birds, 73% (11/15) of the confirmed/probable cases had domiciliary contact with birds and 27% (4/15) had occupational contact. In addition, 47% (7/15) patients had contact with infected birds in which *C. psittaci* was laboratorial confirmed; from another 47% (7/15) of the cases, biological samples of the birds related to the patient could not be obtained, and in 6% (1/15) of the cases *C. psittaci* was not detected in the bird hosts evaluated. In addition, several confirmed/probable cases reported having close contact with the birds as handling (100%, 15/15), keeping the bird in the shoulder (67%, 10/15), kissing the bird (40%, 6/15) and sharing the food with the bird (13%, 2/15), practices that facilitate diseases transmission. Human and animal health care providers have an important role to play in identifying specific factors affecting the health of their patients and should work together. Conjointly efforts would increase the understanding on zoonotic disease and would, ultimately, improve prevention and control strategies.

Key words: Avian chlamydiosis. Epidemiological surveillance. Psittacosis. Zoonosis.

SUMÁRIO

1 INTRODUCTION 15

1.1 CHAPTERS PRESENTATION.......... 17

2 HISTORY ASPECTS AND THE RELEVANCE OF AVIAN CHLAMYDIOSIS IN BRAZILIAN CAGE BIRDS.......... 19

2.1 INTRODUCTION.......... 20

REFERENCES.......... 25

3 EPIDEMIOLOGICAL SURVEILLANCE OF PSITTACOSIS: THE USEFULNESS OF THE MULTIDISCIPLINARY APPROACH.......... 29

3.1 INTRODUCTION.......... 31

3.2 MATERIAL AND METHODS.......... 33

3.3 RESULTS.......... 35

3.4 DISCUSSION.......... 42

REFERENCES.......... 46

4 INTERSECTORAL ACTION FOR HEALTH: PREVENTING PSITTACOSIS SPREAD AFTER ONE REPORTED CASE.......... 50

4.1 INTRODUCTION.......... 51

4.2 EPIDEMIOLOGICAL SURVEILLANCE DESCRIPTION.......... 52

4.3 DISCUSSION.......... 56

REFERENCES.......... 59

5 PSITTACOSIS ASSOCIATED WITH PET BIRD OWNERSHIP: A CONCERN FOR PUBLIC HEALTH.......... 62

5.1 INTRODUCTION.......... 63

5.2 CASE REPORT.......... 63

5.3 DISCUSSION.......... 65

REFERENCES.......... 70

6 FINALS CONSIDERATIONS.......... 72

REFERENCES.......... 73

1 INTRODUCTION

The family Chlamydiaceae includes coccoid, non-motile, obligate intracellular organisms of 0.2 up to 1.5 µm diameter that reside in vacuole-like inclusions of eukaryotic cells, where they parasitize and multiply in a unique developmental cycle. These microorganisms are pathogens of both mammals and birds and infect many hosts, with variable tissue tropism causing a multiplicity of acute and chronic diseases (BEECKMAN; VANROMPAY, 2009; SACHSE et al., 2015). *Chlamydia* is the only currently defined genus in the Chlamydiaceae and is composed of the current species *Chlamydia trachomatis* (humans); *C. pneumoniae* (mainly humans), *C. psittaci* (birds), *C. muridarum* (rodents), *C. pecorum* (mainly cattle and koala), *C. suis* (swine), *C. avium* (birds), *C. gallinacea* (birds), *C. abortus* (sheep, goat, cattle), *C. caviae* (guinea pig), and *C. felis* (cats) (SACHSE et al., 2015). Significant species of the genera representing sources of infection for humans are *C. psittaci*, *C. abortus* and *C. felis*. (RODOLAKIS; MOHAMAND, 2010).

Nonetheless, undoubtly the most important animal chlamydioses with zoonotic potential is caused by *Chlamydia psittaci*, which is the causative agent of human psittacosis and avian chlamydiosis. Psittacosis occurs mostly after exposure to infected sources. Infection usually happens when a person inhales the aerosolized organism from dried faeces or respiratory secretions, which can occur through mouth-to-beak contact or handling of infected birds. Zoonotic transmission may result in subclinical infection or manifest as ‘flu-like’ illness or a potentially fatal interstitial pneumonia. The onset of the illness usually follows an incubation period of 5–14 days. As the disease is rarely fatal in properly treated patients, early diagnosis is important (NASPH, 2010; SACHSE et al., 2015).

In humans, psittacosis is usually diagnosed using a combination of clinical signs and serology. The most common confirmatory test is a fourfold rising titer (immunoglobulin G) to *C. psittaci* in paired sera with the microimmunofluorescence test. For epidemiological surveillance, the CDC (Center for Diseases Control and Prevention) has established case definitions (NASPH, 2010), nonetheless psittacosis confirmed cases might be difficult to determine. Some causes include cross-reactivity with other *Chlamydia* species infecting humans and empirical therapy for community-acquired-pneumonia, which may blunt antibody response to *C. psittaci*. Therefore, history of bird contact is a valuable clue in disease investigation, since exposure to birds is reported in 85% of the psittacosis cases (JUNG; GRAYSON, 1988; BEECKMAN; VANROMPAY, 2009).

In birds, transmission of *C. psittaci* primarily occurs from one infected bird to another susceptible bird in close proximity. The agent is excreted intermittently in faeces and exudates, and the primary route of infection is through the respiratory tract, followed by the oral route. The most common visible clinical sign of avian chlamydiosis involves the respiratory or gastrointestinal systems of birds. Clinical signs include lack of appetite, weight loss, depression, diarrhea, conjunctivitis, discharge from the eyes or nares, or even death. In other cases, birds actively infected with *C. psittaci* may be only mildly affected or show no signs of illness (VANROMPAY et al., 1995; SACHSE et al., 2015).

All over the world, at least 465 avian species were found to be infected with this zoonotic agent. *C. psittaci* is highly prevalent in Psittacidae, such as macaws, cockatoos, cockatiels, parrots, parakeets and lories (KALETA; TADAY, 2003); those are also de main bird species kept as pets.

It should be emphasized that birds are among the most popular pets around the world. For example, in Brazil, 37 million birds are kept as companion animals (ABINPET, 2015). In general, the top sources for acquiring a pet are friend/acquaintance, pet shop, recommended breeder, and private advertisement (HALSBY, 2014). Concerning zoonosis pathogens, it is well establish that *Chlamydia psittaci* is endemic among birds from different origins in the country (RASO, 2014). With reference to pet birds, there are also studies examining *C. psittaci* infections in birds from pet shops and breeders, revealing high prevalences (RASO et al., 2011; SANTOS et al., 2014) but there is little exploration of human infections arising from these facilities. In this scenario, how to recognize when birds may be effectively serving as a source of infection of *C. psittaci* to human beings? This query can only be clarified through a collaborative effort between veterinary and human health professionals, with mutual approaches in which the disease or infection can be observed simultaneously in both the human and the animal patient. For this reason, this study aimed continuous epidemiological investigations of *C. psittaci* infection in birds that played a role as a potential source of *C. psittaci* infection for human patients. In order to reach both susceptible hosts (human and animal), this research was developed by our team (Veterinary Faculty of the University of São Paulo), along with a partnership with the medical team of the Ambulatory of Tropical Diseases and Zoonosis of the Infectology Institute Emilio Ribas (IIER). The results of this investigation are pioneering in the national medical literature and can be verified in the chapters below.

1.1 CHAPTERS PRESENTATION

This thesis is written in chapters, including this introduction (chapter 1). Each chapter discloses relevant issues on avian chlamydiosis/human psittacosis disease, such as critical points on the interface of the bird´s pathogen and the clinically ill person, the gaps in understanding the animal management determinants for the disease occurrence and the lack of awareness among medical health professionals about this underestimated zoonosis.

Chapter 2 entitled *History Aspects and the Relevance of Avian Chlamydiosis in Brazilian Cage Birds* provides past aspects of avian chlamydiosis, on how since the beginning, i.e. the first case description, it has been considered a public health threat. Moreover, discuss the relevance of the disease in Brazilian pet birds, and why we do keep seeing the same failures in disease detection and prevention.

Chapter 3, *Epidemiological Surveillance of Psittacosis: the Usefulness of the Multidisciplinary Approach*, describe a thoroughgoing epidemiological study on human psittacosis at an Infectology Reference Center in the city of São Paulo. In Brazil, authors have performed surveys in occupational settings as well as investigating important psittacosis outbreaks (RASO et al., 2010; RASO et al., 2014); nevertheless, none studies have so far addressed the occurrence of the prevalence of type-specific antibodies (IgA, IgM and IgG) against *Chlamydia psittaci* in a selected hospital population. Furthermore, upon intensive investigation of each psittacosis case, attempts to reach the bird that potentially act as the source of infection was carried out. This is a pioneering work accomplished by veterinarians and physicians conjointly, and highlights the importance of multidisciplinary teams working on zoonotic diseases that can actually lead to the implementation of daily actions and strategies that bring the nexus of human and animal health into better focus to ensure positive and real health impact. This study will be submitted to a Public Health scientific jornal with the following co-authors; Dr. Marcos Vinícius da Silva, Sátiro Márcio and Dr. Tânia Freitas Raso.

Chapters 4, *Intersectoral Action for Health: preventing psittacosis spread after one reported case* and Chapter 5, *Psittacosis associated with Pet Bird Ownership: a Concern for Public Health*, accurately describe two case reports scrupulously selected from the epidemiological study described in Chapter 3. Each of the reports attempts to outline different hidden scenarios behind psittacosis cases; to be precise: lack of sanitary practices in pet birds breeding and trade, the role of pet shops as a connection point for zoonotic disease, unrestricted use of antibiotics by bird owners and very limited public awareness about the risks of contracting zoonotic diseases from pet birds. On the other hand, the difficulties related to human psittacosis diagnosis and surveillace. The lack of consciousness about this zoonosis by physicians, the struggle in acquiring paired serum sample for proper diagnosis and the lack of feedback by health professionals regarding the patient´s bird contact after the patient's successful diagnosis and treatment. Chapter 4 is going to be submitted to an Infectious Diseases scientific journal and published with the following co-authors, Dr. Marcos Vinícius da Silva, Dr. Bill Randerson Bassetti, Dr. Alessandra Pellini and Dr. Tânia Freitas Raso. In turn, chapter 5 is already published as follows: Ferreira, V.L.; Silva, M.V.; Nascimento, R.D.; Raso, T.F. Psittacosis associated with pet bird ownership: a concern for public health. Journal of Medical Microbiology Case Reports (DOI 10.1099/jmmcr.0.000085).

Lastly Chapter 6 brings finals considerations on the studies presented in this thesis and further thoughts of research perspectives concerning human psittacosis and avian chlamydiosis in Brazil.

2 HISTORY ASPECTS AND THE RELEVANCE OF AVIAN CHLAMYDIOSIS IN BRAZILIAN CAGE BIRDS

ABSTRACT

Avian chlamydiosis is a zoonotic disease of birds caused by the intracellular bacterium *Chlamydia psittaci*. In humans, this disease is called psittacosis and most infections are typically acquired from exposure to psittacines birds. *C. psittaci* infection is relatively common in caged birds and total eradication of *Chlamydia* from bird's patients is probably an unattainable goal. However, the clinical significance of this disease can greatly reduce if a common sense approach it used toward its control. Here we review historical aspects of avian chlamydiosis and the relevance of this disease in Brazilian pet birds.

Key words: *Chlamydia psittaci*. Psittacosis. Psittacine birds. Zoonosis

2.1 INTRODUCTION

Nowadays, birds are among the most popular animals maintained as pets. They have been kept for company for hundreds of centuries. Historically, during the Age of Exploration in the 15th and 16th centuries, Europeans were first introduced to the plentiful bird life of the New World. In fact, parrots were the first animals exported to the Old World (POLLOCK, 2013).

The first description of human psittacosis, a zoonotic disease caused by the intracellular bacterium *Chlamydia psittaci* also occurred in the Old World in 1879. By that time, the disease was not well known and Jakob Ritter, a physician, named the disease pneumotyphus. He wrote an article on infectious disease entitled, "*Contribution to the Question of Pneumotyphus.*" In this article, Ritter describes a small epidemic reporting the death of three individuals out of seven cases of psittacosis caused by imported parrots and finches caged maintained as pets in his brother's house in Switzerland. Ritter considered the birds as the source of infection, but made no final conclusions (RITTER, 1879; HARRIS; WILLIAMS, 1985).

Subsequently, a number of outbreaks were reported in Europe. The most important one occurred in Paris during the early 1890s. By then, the French physician Antonin Morange had observed several cases of flu-like symptoms in humans having contact with imported parrots, thus naming the disease psittacosis, which derived from the Greek word for parrot (*psittakus*) (RAMSAY, 2003; HARKINEZHAD et al., 2009; POSPISCHIL, 2009).

The Parisian outbreak had a 33% mortality rate and resulted in the first reported efforts to determine the cause of psittacosis. Edmond Nocard, a French veterinarian suggested a gram-negative bacterium, cultured off the dried wings of parrots that had died during a transatlantic shipment (RAMSAY, 2003). It is interesting to notice that all those reports were written in countries where Psittaciformes do not occur naturally, since they are found mostly in tropical and subtropical regions (GRAHL, 1990). Keeping imported exotic birds as pets was a common practice, first among aristocrats, later on merchants and other individuals started keeping these birds and their popularity crossed class lines as well as racial and ethnic barriers (POLLOCK, 2013).

One of the most important events in the recorded history of psittacosis disease - the outbreak of 1929-1930 - was attributed to Amazon parrots imported from South America. In

July 1929, the physician Enrique Barros informed the Medical Society of Córdoba, Argentina, about the appearance of over 100 cases of a serious pneumonia among inhabitants of the province. Dr. Barros described that a large consignment of 5.000 psittacines birds was imported into Argentina from Brazil and that coincidently a severe epidemic amongst these birds was going on. This gave him the clue to the real diagnosis (LANCET, 1930; MEYER, 1942).

The disease spread throughout Argentina and the trade of parrots was completely forbidden. Nevertheless, some unscrupulous dealers, wishing to minimize their losses, sold their flocks at the port and the infected birds were conveyed to at least 12 different countries (MEYER, 1942). During the worldwide outbreak (1929-1930), overall 766 human psittacosis cases with 112 fatalities were reported, most of them from Germany (215 cases), Argentina (180 cases), USA (169 cases) and the United Kingdom (125 cases) (POSPISCHIL, 2009).

In the USA, the public health report stressed that cases were associated with "newly acquired tropical birds". Data from 1929 indicate that about 350.000 to 410.000 birds were brought into the country. The role of animal importers and pet shops as places of bird-to-bird transmission gained considerable attention by American public health officials (RAMSAY, 2003). Thus, the urgent need for a study of psittacosis was brought to Karl Friedrich Meyer, one of the world's most prodigious investigators in animal diseases and public health at that time. He was a veterinarian and professor at the University of California (UCSF, 2016; HONIGSBAUM, 2014).

Before Meyer, no one assessed the extent to which the disease was spread also by psittacines birds bred in North American aviaries including in birds not displaying signs of illness. These silent infections were a particular problem in California where, during the Depression (1930s), many people supplemented their incomes by breeding budgerigars in backyard aviaries. To control the diseases in the birds, Meyer proposed to the breeders to sacrifice 20% of their stock, in return he would undertake inoculation studies at the University and certify aviaries that were found to be disease-free. In the following two years, he had tested nearly 30.000 parakeets and certified 185 Californian aviaries as chlamydiosis-free (HONIGSBAUM, 2014).

In South America, scientists were also aware of the disease. In the 1930s in Brazil, researchers at the Instituto Biológico in São Paulo, Genésio Pacheco and Otto Bier, conducted studies on Amazon parrots in order to investigate their diseases. They published a report on a highly lethal epizootic disease among different species of parrots. The etiologic agent had different characteristics from the one causing avian chlamydiosis. Further, it was established that it was a herpes virus (PACHECO, 1932; KALETA; DOCHERTY, 2007).

It was almost 67 years later that *Chlamydia psittaci* was confirmed in the country in a study conducted in different breeder collections of Amazon parrots (RASO, 1999). By this time, Amazon parrots already represented one of the most popular groups of pet birds in Brazil. Considering the zoonotic potential of this disease and the high number of infected birds found in the study, the authors recommended that diagnostic tests and prophylactic measures should be instituted to control *C.psittaci* among Brazilian psittacines flocks (RASO et al., 2002). Even so, far too little was done by health authorities to prevent the disease, the main efforts came from the scientific community that continued to do research in different avian groups.

Chlamydia psittaci was detected in populations of free-living birds in several parts of the country. In Central-western Brazil, molecular analysis demonstrated a prevalence of 6% in blue fronted parrots (*Amazona aestiva*) and 9% in Hyacinth macaws (*Anodorhynchus hyacinthinus*), all apparently healthy birds (RASO et al., 2006). Another epidemiology survey conducted in Southeast Brazil revealed a prevalence of 1.2% in a red-tailed Amazon (*Amazona brasiliensis*) population, also in birds with no clinical signs of diseases (RIBAS et al., 2014).

On the contrary, higher prevalence of *C. psittaci* and even outbreaks of avian chlamydiosis with huge mortality rates were observed in captive birds. In the São Paulo State, 58 blue-fronted Amazon parrot nestlings, recovered from the illegal trade, became ill at a wildlife rehabilitation center; mortality rate was 96.5% despite treatment (RASO et al., 2004). In the largest seizure of Hyacinth macaws designated for the illegal trade in Brazil, a 65% prevalence of *C. psittaci* infection was detected (RASO et al., 2013). In the Brazilian Southeastern region, ECO et al. (2009) also reported a chlamydiosis outbreak in psittacine birds recovery from the illegal trade highlingting that more attention must be given to this disease.

As already mentioned by Meyer in 1957, "*it is not surprising that the best-known segment of the natural history of avian chlamydiosis is the infection in an unnatural niche: captivity. When the bird is bred and raised in captivity in large numbers under conditions that differ radically from those of his natural habitat, the host-parasite relationship undergoes some changes. The pathogen strains isolated from acutely infected cage birds have been distinctly virulent.*" (MEYER, 1957). Not different, Dr. Barros when investigating what supposed to be the largest avian chlamydiosis outbreak in Argentina in the 1930s, made the following affirmation "*the organism is habitually present in the intestine of birds of the parrot tribe, and it becomes virulent under conditions whith lower resistance, thus producing the avian epidemic. Such epidemics do not arise in the habitat of the birds, but only when they are brought in large numbers into new and unfavourable conditions.*" (LANCET, 1930).

Barros and Meyer observed these features in former times, when modern scientific tools were not available as they are today. Nowadays, we do know that asymptomatic carriers are common and may carry the pathogen for years and then emerge with clinical signs following some stressful episode (FLAMMER, 1997); and still we keep seeing the same management problems leading to high prevalence of *C. psittaci* infection and avian chlamydiosis in birds flocks. Above, it was mentioned studies on animals maintained illegally in captivity, but many times the conditions are not very different from the ones in which the birds are maintained or raised legally in breeding aviaries and pet shops.

Raso et al. (2011) evaluate the presence of *C. psittaci* in cockatiels (*Nymphicus hollandicus*) from different breeding aviaries. The birds were intended to pet shop trade in São Paulo. Incredibly, 90% of the birds were infected with *C. psittaci*. The hidden scene behind this ratio is the lack of veterinary medical assistance, good husbandry practices and proper clean and disinfection measures. Santos et al. (2014) conducted a study on risk factors associated with *C. psittaci* infection in psittacines birds sold in pet shops and in psittacines birds kept in households in Bahia. Birds kept in households were far less frequently positive (3%) than those at pet markets (17%). Among the factors analysed in the epidemiology of the disease, population density and cage hygiene in birds at pet markets were significantly associated with *C. psittaci* infection.

This reality is not only detrimental to the avian species infected, but also lead to an issue of public health concern. Raso et al. (2015) report a case in which a 36-year-old patient was admitted to a hospital with acute respiratory distress syndrome, requiring treatment in the Intensive Care Unit for many days. Health authorities conducted epidemiological surveillance and reached the pet shop where the patient had bought a pet cockatiel (*N. hollandicus*). Several birds at the pet shop were found to be *C. psittaci* positive.

As to the legislation, so far sanitary rules have been focusing mainly the poultry industry (BRASIL, 2009). Nonetheless, in 2014 the Brazilian Federal Council of Veterinary Medicine (Conselho Federal de Medicina Veterinária) (CFMV, 2014) released an ordinance regulating the general guidelines concerning technical responsibility in shops selling or even donating animals. Such ordinance emphasizes that the technician in charge must ensure the health aspects of the establishment with special attention to avoid the presence of animals with potential risk of zoonosis transmission or diseases easily transmitted among the species involved. This is of particular relevance when taken into account the number of birds maintained as pets in the country, 37 million, according to the Brazilian Association of Pet Product Industries (Associação Brasileira da Indústria de Produtos para Animais de Estimação) (ABINPET, 2015).

It is important to keep in mind that a sick/infected bird in a pet shop can potentially spread the illness to other animals within the shop, and to a large number of geographically distributed owners as newly purchased pets are taken home. Pet shops can therefore act as a nexus point for zoonotic disease (HALSBY et al., 2014).

Prophylactic measures are of paramount importance to avoid such situations. Pet shops should purchase their birds from sources that routinely test for avian chlamydiosis and guarantee the health of their birds; separate birds according to species and age (utilize barriers between cages) to prevent bird to bird contact and cross contamination; minimize stress (relocation, unnecessary handling, chilling, overheating). Also, keeping accurate records of all birds for at least one year; records should include bird identification, species, source, date acquired, date of illness, clinical signs of disease, treatment, and bird deaths. Protect staff by wearing protective clothing when handling potentially infected birds or cleaning their cages. Clean and disinfect all cages, food and water bowls daily and whenever moving birds. Wash with detergent, rinse with water, then apply disinfectant (allow appropriate contact time), and rinse with water and finally test new birds upon arrival and before boarding or selling. Bird testing and treatment should be conducted or supervised always by an experienced avian veterinarian (NASPHV, 2010).

In addition, worth mention that for avian chlamydiosis successful treatment, antibiotic therapy is equally as important as reinfection prevention. That is, treatment of infected birds requires not only administration of the therapeutic dose of drug. Frequent cleaning to eliminate infected dust and disinfecting of the aviary/cage is beneficial in eliminating chlamydiae from the environment and crucial in preventing reinfection (VANROMPAY et al., 1995).

The National Association of State Public Health Veterinarians of the USA provides many recommendations on avian chlamydiosis control (NASPHV, 2010). Nonetheless, as in other countries, here in Brazil authorities have power to impose conditions on the licensing of pet shops and breeding establishments. In addition, apply rules including taking all reasonable precautions to prevent outbreaks and spreads of disease minimizing the hazards for animals and for people in contact with them.

REFERENCES

ASSOCIAÇÃO BRASILEIRA DA INDÚSTRIA PET (ABINPET). **Evolução de Mercado Anual**. 2015. Disponível em: <http://www.agricultura.gov.br/arq_editor/file/camaras_tematicas/Insumos_agropecuarios/79RO/IBGE_PAEB.pdf>. Acesso em: 21 jun. 2016.

BRASIL. Ministério da Agricultura, Pecuária e Abastecimento. **Manual de Legislação – Programas Nacionais de Saúde Animal do Brasil**. Brasília, DF, 2009. 440 p.

CONSELHO FEDERAL DE MEDICINA VETERINÁRIA (CFMV). **Manual de Legislação do Sistema CFMV/CRMVs.** Brasília, DF: CFMV, 2009. Publicado no DOU de 12/01/2015, seção, 1p. 56.

ECO, R.; PREIS, I. S.; MARTINS, N. R.; VILELA, D. A. R.; SHIVAPRASAD, H. L. An outbreak of chlamydiosis in captive psittacines. **Brazilian Journal of Veterinary Pathology,** v.2, n.2, p.85 – 90, 2009.

FLAMMER, K. Chlamydia. In: ALTMAN, R.; CLUBB, S.; DORRESTEIN, G. M.; QUESENBERRY, K. **Avian medicine and surgery**. Philadelphia: Saunders, 1997. p.364-378.

GRAHL, W. D. **Papageien:** Lebensweise, Arten, Zucht. 9th ed. Germany: Verlag Eugen Ulmer,1990, 293 p.

HALSBY, K. D.; WALSH, A. L.; CAMPBELL, C.; HEWITT, K.; MORGAN, D. Healthy Animals, healthy people: zoonosis risk from animal contact in pet shops, a systematic review of the literature. **PLoS One,** v.26, n.9, p.e89309, 2014.

HARKINEZHAD, T.; GEENS, T.; VANROMPAY, D. *Chlamydophila psittaci* infections in birds: A review with emphasis on zoonotic consequences. **Veterinary Microbiology**, v.16, n.135, p. 68-77, 2009.

HARRIS, R. L.; WILLIAMS, T.W. "Contribution to the question of pneumotyphus": A Discussion of the Original Article by J. Ritter in 1880. **Reviews of Infectious Diseases**. v.7, n.1, p.119-122, 1985.

HONIGSBAUM, M. In search of sick parrots: Karl Friedrich Meyer, disease detective. **The Lancet**, v.383, p.1880-1881, 2014.

KALETA, E. F.; DOCHERTY, D. E. Avian herpesviruses. In: THOMAS, N. J.; HUNTER, D. B.; ATKINSON, C. T. **Infectious diseases of wild birds.** Iowa: Blackwell Publishing, 2007. p.63-86.

LANCET. Psittacosis in Argentina. **Lancet**, v.215, p.472-473, 1930.

MEYER, K. F. The ecology of psittacosis and ornithosis**. Medicine**, v.21, n.2, p.175, 1942.

MEYER, K. F. The natural history of plague and psittacosis. **Public Health Rep,** v.72, n.8, p.705-719, 1957.

NATIONAL ASSOCIATION OF STATE PUBLIC HEALTH VETERINARIANS (NASPHV). **Compendium of Measures to Control *Chlamydia psittaci* (Avian Chlamydiosis).** 2010. Disponível em: <http://www.NASPHVv.org/Documents/Psittacosis.pdf>. Acesso em: 13 maio 2016.

PACHECO, G. Investigações sobre doenças de psittacideos. **Memórias do Instituto Oswaldo Cruz**, v.26, n. , p.169-233, 1932.

POLLOCK, C. G. Companion Birds in Early America. **Journal of Avian Medicine and Surgery**, v.7, n.2, p.148-151, 2013.

POSPISCHIL, A. From disease to etiology: historical aspects of chlamydia-related diseases in animals and humans. **Drugs of Today**, v.45, Suplement B, p.141-146, 2009.

RAMSAY, E. C. The psittacosis outbreak of 1929-1930. **Journal of Avian Medicine and Surgery**, v.17, n.4, p.235-237, 2003.

RASO, T. F. **Detecção de infecção por *Chlamydia psittaci* em papagaios do gênero Amazona mantidos em cativeiro**. 1999. 61 f. Dissertação (Mestrado em Medicina Veterinária - Faculdade de Ciências Agrárias e Veterinárias, Universidade Estadual Júlio de Mesquita Filho, Jaboticabal, SP.

RASO, T. F.; BERCHIERI JR., A.; PINTO, A. A. Evidence of *Chlamydophila psittaci* infection incaptive amazon parrots in Brazil. **Journal of Zoo and Wildlife Medicine**, v.33, n. 2, p.118–121, 2002.

RASO, T. F.; FERREIRA, V. L.; BASSETTI, B. R.; SZTAJNBOK, J.; KHOURY, Z.; NASCIMENTO, R. D.; SILVA, M. V. Psittacosis causing acute respiratory distress syndrome (ARDS). **J Pulm Respir Med**, v.5, n.4, p.1-5, 2015 doi: 10.4172/2161-105X.1000283

RASO, T. F.; GODOY, S. N.; MILANELO, L.; DE SOUZA, C.A.; MATUSCHIMA, E. R.; ARAÚJO JÚNIOR, J. P.; PINTO, A. A. An outbreak of chlamydiosis in captive blue-fronted Amazon parrots (*Amazona aestiva*) in Brazil. **Journal of Zoo and Wildlife Medicine** v.35, n.1, p. 94-96, 2004.

RASO, T. F.; GRESPAN, A.; FERREIRA, V. L.; PINHEIRO, S. R. *Chlamydophila psittaci* infection in pet cockatiels (*Nymphicus hollandicus*) in Brazil. In: INTERNATIONAL CONGRESS ON PATHOGENS AT THE HUMAN-ANIMAL INTERFACE (ICOPHAI): IMPACT, LIMITATIONS AND NEEDS IN DEVELOPING COUNTRIES, 1., 2011, Addis Abeba. **Proceedings**….Addis Abeba, 2011, p.41.

RASO, T. F.; SEIXAS, G. H. F.; GUEDES, N. M. R.; PINTO, A. A. *Chlamydophila psittaci* in free-living Blue-fronted Amazon parrots (*Amazona aestiva*) and Hyacinth macaws (*Anodorhynchus hyacinthinus*) in the Pantanal of Mato Grosso do Sul, Brazil. **Veterinary Microbiology**, v.117, p.235–241, 2006.

RASO, T. F.; TEIXEIRA, R. H.; CARRASCO, A. O.; ARAÚJO JÚNIOR, J. P., PINTO, A. A. *Chlamydophila psittaci* infections in hyacinth macaws (*Anodorhynchus hyacinthinus*) confiscated in Brazil. **Journal of Zoo and Wildlife Medicine**., v.44, n.1, p.169-72, 2013.

RIBAS, J. M.; SIPINSKI, E. A. B.; SERAFINI, P. P., FERREIRA, V. L., RASO, T. F, PINTO, A. A. *Chlamydophila psittaci* assessment in threatened red-tailed Amazon (*Amazona brasiliensis*) parrots in Paraná, Brazil. **Ornithologia,** v.6, n.2, p.144-147, 2014.

RITTER, J. Beitrag zur Frage des Pneumotyphus. Eine Hausepidemie in Uster (Schweiz) betreffend. **Dtsch. Arch. Klin. Med**. n.25, p.53-96, 1879.

SANTOS, F.; LEAL, D. C.; RASO, T. F.; SOUZA, B. M.; CUNHA, R. M.; MARTINEZ, V. H.; BARROUIN-MELO, S. M.; FRANKE, C. R. Risk factors associated with *Chlamydia psittaci* infection in psittacine birds. **Journal of Medical Microbiology**, v.63, n.3, p.458-63, 2014.

VANROMPAY, D.; DUCATELLE, R.; HAESEBROUCK, F. *Chlamydia psittaci* infections: a review with emphasis on avian chlamydiosis. **Veterinary Microbiology**, v.45, p.93-119, 1995.

UNIVERSITY OF CALIFORNIA SAN FRANCISCO (UCSF) [2016?]. **A history of UCSF**. Disponível em: http:<//history.library.ucsf.edu/meyer.html>. Acesso em: 23 Junho 2016.

3 EPIDEMIOLOGICAL SURVEILLANCE OF PSITTACOSIS: THE USEFULNESS OF THE MULTIDISCIPLINARY APPROACH

ABSTRACT

Human psittacosis is a zoonotic disease caused by the bacterium *Chlamydia psittaci*. Transmission of the pathogen usually originates from close contact with infected birds frequently in the context of companion animals. In general, communication between the veterinarian caring for the family pets and the human health care providers treating the human household members is limited. Similarly, there may be limited contact between public health professionals and their animal health counterparts. The growing recognition of the links among human and animal health requires new efforts for cooperation and communication between professionals working in these sectors. In this respect, in the present study selected patients with previous bird contact were investigated for psittacosis disease at the Ambulatory of Tropical Diseases and Zoonosis of the Infectology Institute Emilio Ribas (IIER), Brazil. Serum samples from eligible patients were tested by microimmunofluorescent (MIF) test for *C. psittaci* IgA, IgM and IgG antibodies. Seropositive patients were scrutinised and attempts to reach the avian host that potentially act as the source of infection were made by a veterinarian team. Thus, biological samples from birds related to the patients were acquired and tested by polymerase chain reaction (PCR). Furthermore, questionnaires in respect of potentially risk factors concerning birds contact were applied. Among the eligible patients in this study, 27% (10/37) were classified as confirmed cases of psittacosis; 13.5% (5/37) as probable and 59.5% (22/37) as discarded. Pneumonia ($p = 0.004$), cough ($p = 0.002$) and chills ($p = 0.011$) were statistically significant when comparing with those patients in which psittacosis was discarded. With reference to the epidemiological link with birds, 73% (11/15) of the confirmed/probable cases had domiciliary contact with birds and 27% (4/15) had occupational contact. In addition, 47% (7/15) patients had contact with infected birds in which *C. psittaci* was laboratorial confirmed; from another 47% (7/15) of the cases, biological samples of the birds related to the patient could not be obtained, and in 6% (1/15) of the cases *C. psittaci* was not detected in the bird hosts evaluated. Moreover, several confirmed/probable cases reported having close contact with the birds as handling (100%, 15/15), keeping the bird in the shoulder (67%, 10/15), kissing the bird (40%, 6/15) and sharing the food with the bird (13%, 2/15), practices that facilitate diseases transmission. Human and animal health care

providers have an important role to play in identifying specific factors affecting the health of their patients and should work together. Conjointly efforts would increase the understanding on zoonotic disease and would, ultimately, improve prevention and control strategies.

Key words: *Chlamydia psittaci*. Pneumonia. Pet birds. Zoonosis

3.1 INTRODUCTION

The relationship between human and animal health is becoming increasingly complex and includes several biological and social factors. Approximately 60% of all infectious pathogens of human beings are zoonotic in origin and their occurrence in humans relies on the human-animal interface, defined as the continuum of contacts between humans and animals, their environments, or their products (WOOLHOUSE; GOWTAGE-SEQUERIA, 2005). As an example, nowadays domestication in our global society has taken unprecedented proportions and the rising numbers of household pets means that an increasing number of people are exposed to the risk of acquiring zoonotic disease from companion animals (DAMBORG et al., 2015).

Concerning pet birds, a major bacterial zoonosis threat is avian chlamydiosis, known as psittacosis in human beings, and caused by the intracellular bacterium *Chlamydia psittaci*. It has been isolated from more than four hundred avian species and is most commonly identified in psittacine birds (e.g. parrots, macaws, cockatiels, and parakeets). Among non-psittacine birds, infection with *C. psittaci* occurs most frequently in pigeons and doves (KALETA; TADAY, 2003; NASPHV, 2010). The time between exposure to *C. psittaci* and the onset of illness in caged birds ranges from three days to several weeks. However, latent infection are common and active disease may appear years after exposure. Shipping, crowding, chilling, breeding and other stress factors may activate shedding of the infectious agent among birds with latent infection. Birds may appear healthy but may be carriers of *C. psittaci* and shed the organism intermittently. When shedding occurs, the organism is excreted in feces and nasal/ocular discharges of infected birds, is resistant to drying, and can remain infective for several months (NASPHV, 2010).

In humans, transmission occurs either by inhalation of aerosolized organisms in dried feces or respiratory tract secretions, or by direct bird contact. Consequently, those at great risk are individuals with leisure or occupational exposure to birds, including pet bird owners, veterinarians, pet shop employees, and poultry processing plant employees. As a result, cases of psittacosis can range from a sporadic case in a pet bird owner to an outbreak affecting several birds in a commercial flock and multiple infected workers (GAEDE et al., 2008; RASO et al., 2015).

Usually, in humans, 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 a mild, non-specific illness to a systemic illness with severe pneumonia. Humans with symptomatic infections typically have an abrupt onset of fever, chills, headache, malaise, and myalgia. A nonproductive cough is usually present and can be accompanied by breathing difficulty and/or chest tightness. Clinical presentation and positive antibodies G against *C. psittaci* in acute and convalescent-paired sera using MIF methods establish most diagnoses. Acute-phase serum specimens should be obtained as soon as possible after the onset of symptoms, and convalescent-phase serum specimens should be obtained at least two weeks after the first specimen (NASPHV, 2010).

Although notification is mandatory in most countries, the impact of psittacosis on human health is difficult to determine. This disease probably occurs more often than reported because individuals with mild cases may not seek medical attention and physicians may not inquire about bird exposure when evaluating patients. Additionally, antimicrobials employed empirically for the therapy of community-acquired pneumonia may prevent an accurate diagnosis (CDC, 1992).

In Brazil, *C. psittaci* infection in birds in endemic (RASO, 2014), despite that, concerning the infection in humans, with exception of few occupational surveys or occasional psittacosis case reports published in the medical literature, none epidemiological studies in communities have been described (RASO et al., 2010; RASO et al., 2014; FERREIRA et al., 2015). As in other countries, these psittacosis cases described in Brazil occurred mainly after exposure to infected pet birds. Nonetheless, communication between the veterinarian caring for the family pets and the human health care providers treating the human household members is currently limited. Similarly, there may be limited contact between public health professionals and their animal health counterparts. The growing recognition of the links among human health, animal health, and the environment requires new efforts for cooperation and communication between professionals working in these sectors (RABINOWITZ; CONTI, 2010). In this respect, in the present study selected patients with previous bird contact were investigated for psittacosis disease at a Reference Infectious Diseases Hospital in São Paulo, Brazil. When proven, seropositive patients were scrutinised and attempts to reach the avian host that potentially act as the source of infection were made by a veterinarian team. Furthermore, risk factors concerning birds contact were also analysed.

3.2 MATERIAL AND METHODS

Human clinical cases and data collection

Eligible participants were patients attended at the Ambulatory of Tropical Diseases and Zoonosis of the Infectology Institute Emilio Ribas (IIER), who had clinical disease compatible with psittacosis, i.e. mild flu-like illness to a systemic illness with severe pneumonia as well as history of bird contact. Moreover, contacts of index cases of psittacosis disease. The patients evaluated either searched the IIER service spontaneously for medical consultation; or were directed to the IIER after searching the Psittacosis Research Group at the School of Veterinary Medicine and Animal Science of the University of São Paulo (FMVZ/USP) due to avian chlamydiosis in their pet birds. This study was conducted from 2011 to 2015. Exclusion criteria were the following: lack of epidemiological link with birds, presence of immunosuppression defined as chemotherapy, known HIV infection, immunosuppressive therapy after organ or bone marrow transplant, or active tuberculosis. All patients gave written informed consent and their names were replaced by an identification number to ensure data security. The study is registered at the Brazilian Committees of Ethics in Research (CEP) as well as at the National Committee for Ethics in Research (CONEP) of the National Health Council.

Upon consultation, participants received a questionnaire from physicians designed to assess information on professional and non-professional activities, smoking habits, general health, use of medications, and occupations/non-occupational contact with birds. In the case of contact with birds, additional data has been obtained; i.e. as how long the bird has been acquired, clinical signs suggestive of avian chlamydiosis, hygiene measures, use of personal protective equipment when cleaning cage, and kind of contact with the birds. When the questionnaire was not filled, health and epidemiological available data were taken from the patients' medical record.

According to CDC (1997) guidelines for psittacosis with adaptations, a patient was considered to have a confirmed case of psittacosis when clinical illness was compatible with psittacosis and the case was laboratory confirmed by MIF with a 4-fold or greater increase in IgG against *C. psittaci*. Also if there was presence of immunoglobulin M against *C. psittaci* to a reciprocal titre of ≥10. A patient was considered to have a probable case of psittacosis if clinical illness was compatible with psittacosis and the patient was epidemiologically linked to a confirmed case of psittacosis or the patient has supportive serology, i.e. single IgG titer of ≥ 32 detected by MIF. Despite not mentioned in the CDC guidelines, Ig A anti-*C.psittaci* in the serum samples was also evaluated in this study.

Human sample processing

Health professionals of the IIER collected the blood samples from the eligible patients placing the samples in Serum Separator Tubes. Next, the samples conditioned on ice (4°C) were immediately sent to the Psittacosis Research Group at FMVZ/USP. Upon arrival, samples were given a unique identifier number, aliquoted (400 µL) into 1.0 mL conical tubes with screw caps and processed within 24 hours, remaining aliquots were stored at -80°C. Serum samples were tested for immunoglobulin IgA, IgM and IgG against *C. psittaci* by microimmunofluorescent (MIF) test kit (Focus Diagnostics, CA, USA) according to the manufactures' protocol. Serum were screened at 1:16 dilution for IgA and IgG; 1:10 for IgM and titre to the end point. For IgA and IgG, seropositive subjects were defined as those with immunoglobulins titres of ≥ 16 and seronegative subjects as those whose titre was < 16, the same for IgM but with titres of ≥ 10 or < 10. To rule out the presence of cross-reactive antibody, testing was also performed for *C. trachomatis* and *C. pneumoniae*. When reacting with the other chlamydial types serum samples were considered positive for *C. psittaci* if they showed the highest grade of reaction with *C. psittaci* antigen.

Birds' cases, biological sample collection and processing

Upon epidemiological surveillance, biological samples of birds directly or indirectly related with the eligible participants attended at the IIER were obtained. Therefore, cloacal and/or oropharyngeal swab samples were taken and placed in tubes containing sterile PBS (Phosphate Buffer Saline) and immediately frozen (-80°C) until the moment of diagnostic testing.

For *C. psittaci* diagnosis, DNA was extracted from the swabs samples with Nucleid Acid and Protein Purification Kit® (Macherey-Nagel, GmbH & Co. KG, Germany), according to the manufacture's protocol. Brazilian strain from monk parakeets (*Myiopsitta monachus*)

(Cpsi/Mm/BR01, GenBank number JQ926183.1) and ultrapure water were used as positive and negative controls, respectively. PCR was based on primers targeting a conserved region of *pmp* gene sequences as designed by Laroucau et al. (2001). Primers sequences were: CpsiA (5'ATGAAACATCCAGTCTACTGG3') and CpsiB (5'TTGTGTAGTAATATTATC AAA3'). Samples were analyzed by electrophoresis on 1.5% agarose gel (Uniscience®, Brazil), stained with Gel RedTM (Uniscience®, Brazil) 0.5µg/10 mL and run at 80 volts/60 min. Samples showing a 300 base pair DNA fragment under UV light were considered to be positive for *C. psittaci*.

All procedures performed in this study were consistent with the Bioethics Commission of the School of Veterinary Medicine of São Paulo University (Number 2870) and authorized by ICMbio/SISBIO (Brazilian Chico Mendes Institute for Biodiversity Conservation/Biodiversity Authorization and Information System – Number 373644).

Statistical analyses

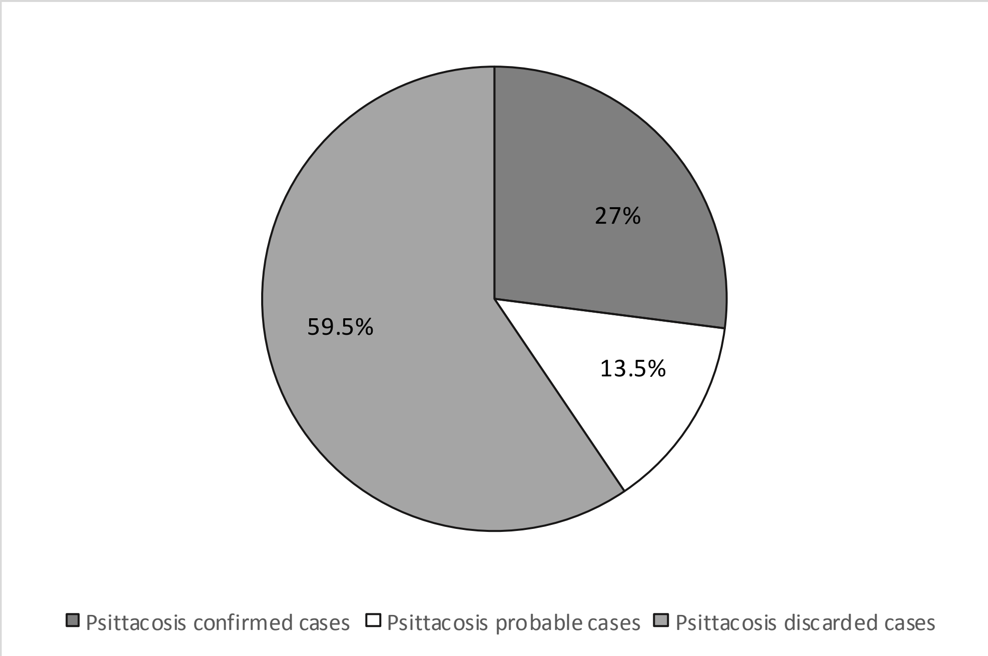
Statistical analysis was performed using Epi Info version 7.3.2. In the analysis, characteristics of potential risk factors reported by patients, as well as by cases controls were compared. Univariate analysis was performed using chi-square tests. When the expected cell counts were < 5, Fisher's exact p-value 2 sides was used.

3.3 RESULTS

Human clinical cases and laboratorial results

Overall, 37 participants were eligible according to the case definition. Serum samples were collected from all eligible participants. Antibodies against *C. psittaci* were detected in 46% (17/37) of the patients. From these, in 10 cases, psittacosis was considered confirmed, 5 was considered probable cases and in 2 psittacosis was discarded. In total, among all eligible patients in this study, 27% (10/37) were classified as confirmed cases of psittacosis; 13.5% (5/37) as probable and 59.5% (22/37) as discarded (Figure 1). Titres of antibodies ranged from 10 to 1024 (Table 1).

Figure 1 - Psittacosis confirmed, probable and discarded cases in 37 eligible participants attended at the Medical Institute of Infectious Diseases Emilio Ribas, São Paulo, Brazil.



Font: (FERREIRA, V. L., 2016)

With respect of age and sex of the confirmed cases, the ages ranged from 19 to 63, with a mean of 38 years. Thirty percent (3/10) of the patients were young with ages between 19 and 25; 60% (6/10) were adults with ages between 26 and 59 and 10% (1/10) were elderly people older than 60 years. There were 50% (5/10) male and 50% (5/10) female patients.

Concerning the clinical features, the main predominant syndrome was a respiratory tract infection with constitutional symptoms (Table 2). The most common clinical findings were cough (100%), fever (67%), pneumonia (60%) and chill (40%). Furthermore, pneumonia ($p = 0.004$), cough ($p = 0.002$) and chills ($p = 0.011$) were statistically significant when comparing with those patients in which psittacosis was discarded. With reference to the epidemiological link with birds, 73% (11/15) of the confirmed/probable cases had domiciliary contact with birds and 27% (4/15) had occupational contact. In addition, 47% (7/15) patients had contact with infected birds in which *C. psittaci* was laboratorial confirmed; from another 47% (7/15) of the cases, biological samples of the birds related to the patient could not be obtained, and in 6% (1/15) of the cases *C. psittaci* was not detected in the bird hosts evaluated.

Table 1 - Information of confirmed and probable psittacosis cases and of the birds epidemiologically related to them during the study conducted at the Medical Institute of Infectious Diseases Emilio Ribas, São Paulo, Brazil.

(to be continued)

		Patients first serum sample			Patients second serum sample			Data of the bird (s) epidemiologically related to the case				
		Antibody tires (MIF test)										
Case	Type of exposure	IgG	IgM	IgA	IgG	IgM	IgA	Bird species	*C. psittaci* DNA detection	Clinical signs in birds	Treatment	Outcome
1	Domiciliary	**1024**	N	N	-	-	-	Cockatiel	Positive	Diarrhea, anorexia, prostration	Unkown	Death
2	Domiciliary	**256**	**80**	N	-	-	-	Cockatiel	Positive	Blepharitis	Doxycycline	Recovery
3	Domiciliary	**32**	N	N	N	N	N	Cockatiel	Positive	Prostation, emaciation	Doxycycline	Death
4 (Contact case 3)	Domiciliary	**32**	**10**	N	-	-	-	Cockatiel	Positive	Prostation, emaciation	Doxycycline	Death
5	Domiciliary	**32**	N	N	-	-	-	Budgerigar	Not performed	Unkown	Unkown	Unkown
6	Domiciliary	**32**	N	N	**64**	N	N	Cockatiels	Positive	Asymptomatic	Oxytetracycline	Recovery
7 (Contact case 6)	Domiciliary	**64**	**20**	N	-	-	-	Cockatiels	Positive	Asymptomatic	Oxytetracycline	Recovery
8	Occupational*	**128**	**20**	N	-	-	-	Various	Not performed	Various	Various	Unkown
9	Domiciliary	**32**	N	N	-	-	-	Blue fronted parrot	Not performed	Unkown	Unkown	Unkown

(Conclusion)

		Patients first serum sample			Patients second serum sample			Data of the bird (s) epidemiologically related to the case				
		Antibody tires (MIF test)										
Case	**Type of exposure**	**IgG**	**IgM**	**IgA**	**IgG**	**IgM**	**IgA**	**Bird species**	***C. psittaci* DNA detection**	**Clinical signs in birds**	**Treatment**	**Outcome**
10	Occupational*	**512**	N	**20**	-	-	-	Various	Not performed	Various	Various	Unkown
11	Domiciliary	N	**20**	N	N	**20**	N	Blue fronted parrot	Positive	Unkown	Unkown	Death (euthanasia)
12	Occupational*	**16**	**10**	N	**512**	**80**	**80**	Various	Negative	Various	Various	Unkown
13	Domiciliary	**256**	**20**	N	**1024**	**20**	N	Cockatiel	Positive	Diarrhea, prostration	Not performed	Death
14	Domiciliary	**128**	**20**	N	**512**	**40**	N	Cockatiel	Positive	Diarrhea, prostration	Not performed	Death
15	Occupational*	**32**	**40**	N	**32**	**20**	**20**	Various	Not performed	Various	Various	Unkown

Font: (FERREIRA, V. L., 2016)

*Occupational cases were veterinary practitioners working exclusively with birds, thus, they were constantly exposed to several birds species with different clinical symptoms and submitted to different treatments.
(-) sample not collected.

Table 2 - Clinical presentation of 15 patients with confirmed/probable psittacosis attended at the Medical Institute of Infectious Diseases Emilio Ribas, São Paulo, Brazil.

Symptoms/features	Number of patients	Percentage (%)	P value *
Respiratory			
Cough	15	100	0.002*
Pneumonia	9	60	0.004*
Dyspnea	7	47	0.242
Chest pain	4	27	0.392
Constitutional			
Fever	10	67	0.140
Chill	6	40	0.011*
Malaise	5	33	0.736
Body ache	5	33	0.095
Neurological			
Headache	5	33	0.736
Gastrointestinal			
Vomiting	1	7	0.554
Diarrhea	1	7	>0.999
Dermatological			
Rash	1	7	0.392

Font: (FERREIRA, V. L., 2016)

*p-values ≤ 0.005 were considered statistically significant

Detailed information on the bird’s management in confirmed/probable and discarded psittacosis cases are described in table 3. From the total number of confirmed/probable cases several patients reported having close contact with the birds as handling (100%, 15/15), keeping the bird in the shoulder (67%, 10/15), kissing the bird (40%, 6/15) and sharing the food with the bird (13%, 2/15).

Table 3 - Management information of birds epidemiologically related to probable, confirmed and discarded psittacosis cases attended at the Medical Institute of Infectious Diseases Emilio Ribas, São Paulo, Brazil

(to be continued)

Information on patients type of exposure and management of the bird	**Patient outcome**		**p value**
	Probable and Confirmed	**Discarded**	
	total (%)	**total (%)**	
Kind of exposure			0.345
Environmental	0 (0)	2 (9.0)	
Domiciliary	11 (73)	16 (72.9)	
Occupational	4 (27)	3 (13.6)	
Unkown	0 (0)	1 (4.5)	
***C.psittaci* PCR**			0.345
Positive	7 (46.7)	9 (41.0)	
Negative	1 (6.6)	6 (27.2)	
Unkown	7 (46.7)	7 (31.8)	
Bird with symptoms			0.406
Yes	11 (73)	11 (50.0)	
No	2 (13.5)	6 (27.2)	
Unkown	2 (13.5)	5 (22.8)	
Bird died			0.612
Yes	12 (80)	14 (63.6)	
No	1 (6.6)	3 (13.6)	
Unkown	2 (13.4)	5 (22.8)	
Time of ownership			0.317
≤ 30 days	4 (26.6)	4 (18.2)	
≥ 1 year	3 (20)	9 (41.0)	
Numerous *	4 (26.6)	3 (13.6)	
Unkown	4 (26.6)	6 (27.2)	
Cage cleaning frequency			0.254
Daily	5 (33.2)	4 (18.2)	
Every other day	2 (13.4)	2 (9.0)	
Triweekly	2 (13.4)	8 (36.4)	
Not at all	0 (0)	2 (9.2)	
Unkown	6 (40)	6 (27.2)	

(conclusion)

Information on patients type of exposure and management of the bird	Patient outcome		
	Probable and Confirmed	Discarded	p value
	total (%)	total (%)	
Wash hands after cage cleaning			0.520
Yes	9 (60)	14 (63.6)	
No	0 (0)	2 (9.2)	
Unkown	6 (40)	6 (27.2)	
Keep the bird on the shoulder			0.253
Yes	10 (66.6)	8 (36.4)	
No	3 (20)	8 (36.4)	
Unkown	2 (13.4)	6 (27.2)	
Kiss the bird			0.242
Yes	6 (40)	4 (18.2)	
No	6 (40)	12 (54.6)	
Unkown	3 (20)	6 (27.2)	
Share food with the birds			>0.999
Yes	2 (13.4)	2 (9.2)	
No	10 (6.6)	14 (63.6)	
Unkown	3 (20)	6 (27.2)	
Contact with the bird's feces			>0.999
Yes	9 (60)	12 (54.4)	
No	4 (26.6)	5 (22.8)	
Unkown	2 (13.4)	5 (22.8)	
Sleep with the bird			
Yes	1 (6.6)	0 (0)	0.392
No	10 (66.6)	17 (77.2)	
Unkown	4 (26.6)	5 (22.8)	

Font: Font: (FERREIRA, V. L., 2016)

* Occupational cases were veterinary practitioners working exclusively with birds, thus, they were constantly exposed to several birds species with different clinical symptoms and submitted to different treatments.

3.4 DISCUSSION

The history of contact between animals and humans has always involved infectious diseases. In fact, the majority of emerging infectious diseases in the past three decades are zoonotic. Therefore, the control and prevention of these diseases can be accomplished only to improving approaches to reducing disease transmission among humans and other animals. Epidemiologic characterization is the first step in defining a disease. It is the epidemiologic features, as prevalence, incidence, transmission route, and susceptible populations that are of paramount importance in developing control programs (RABINOWITZ; CONTI, 2010; NELSON, 2014). With reference to psittacosis, determination on the disease burden in humans is still lacking.

To provide prevalence and incidence rate estimates that are both reliable and generalizable, studies must include a sample large enough to capture most cases and sufficiently distributed both geographically and sociologically, to be representative of the general population (WARD, 2013). However, with uncommon diseases as psittacosis, the challenge is multiplied because cases are fewer and harder to find.

Usually confirmation of psittacosis may only be sought in moderate to severe cases, resulting in many unrecognized mild and asymptomatic infections (SILLIS; LONGBOTTOM, 2011). In addition, even in severe cases, diagnosis might not be conclude because symptoms caused by the *C. psittaci* infection are similar to those of other respiratory pathogens and it is often not included in routine microbiological diagnostic panels for pneumonia (BEECKMAN; VANROMPAY, 2009; SPOORENBERG et al., 2016). In many countries, as in Brazil, psittacosis is not notifiable, thus limiting important epidemiological data assessment.

In the present study psittacosis was evaluated at an Infectious Diseases Reference Hospital, with a 27% (10/37) occurence of confirmed cases in five years, with periodic peaks. Veterinarians aided in this research counselling pet bird's owners in contact with birds with chlamydiosis to get medical assistance at the IIER. Nonetheless, in general, only few studies have evaluated prevalence of *C. psittaci* in human hospitals. Spoorenberg et al. (2016) conducted a survey in two Dutch hospitals in order to detected psittacosis on community-acquired pneumonia (CAP) patients during 3.2 years, finding a prevalence of 4.8% (7/148). In the other hand, on a similar study in Germany, Dumke et al. (2015) found a prevalence of 2.1% (17/783) between 2011 and 2012.

In the studies mention above, eligible patients were those with confirmed CAP, therefore psittacosis cases not leading to pneumonia were not considered. In the presented study, pneumonia ($p = 0.004$), cough ($p = 0.002$) and chills ($p = 0.011$) were statistically significant when comparing with those patients in which psittacosis was discarded. Nonetheless, curiously, 40% of the probable/confirmed psittacosis cases did not presented pneumonia; eligible patients were those who were sick, but necessarily in contact with birds, since exposure to birds is reported in the majority of psittacosis cases (YUNG; GRAYSON, 1988), and achieving the patients source of infection was one of the goals of this work.

In general, surveillance systems fail to capture cases at distinct levels of the surveillance pyramid, one of them is from the community since not all cases seek healthcare (GIBBONS et al., 2014). In the present study, serosurvey in contacts from 3-index psittacosis cases were carried out, in two of them one confirmed and one probable case were established. Contact tracing is an important part of epidemiologic investigation in order to increase case detection (CDC, 1992). As regards to psittacosis disease, such action would improve the estimation of the real number of cases, which in turn could mobilize governmental support in order to increase attentiveness to human psittacosis among health practitioners and local community.

Currently in Brazil, 37 million birds are maintained as pets (ABINPET, 2015) and the community are not necessarily aware of the widespread occurrence and the zoonotic nature of avian chlamydiosis. In this study, 47% patients had contact with infected birds in which *C. psittaci* was laboratorial confirmed. In 6% of the cases that *C. psittaci* was not detected, the birds evaluated were under antibiotic therapy due to different medical disorders, which may result in false negative findings in laboratorial diagnosis (FLAMMER, 1997). In this case, veterinarian prescribed the antibiotics, even so it should be mentioned that pet owners frequently use tetracyclines for any case of respiratory disease without veterinary advice, or even prophylactically. Most pet bird owners are unaware of the dangerous situation that they might create by using these drugs frequently as they could generate resistant strains of *C. psittaci* and other bacteria (HARKINEZHAD et al., 2007; NASPHV, 2010).

Veterinarians have responsibilities in instructing correctly pet owners, they must also inform them of common human symptoms of the disease and direct the animal owner to his/her physician if such symptoms have been noted in the human beings sharing the animal’s environment or if any persons in the home are at increased risk for the disease (RABINOWITZ; CONTI, 2010). In the present study, in respect of human psittacosis, all person that searched assistance at the FMVZ/USP with the veterinarian team were directed to the medical group of the IIER. It should be emphasized that veterinarians must have caution to not overstep professional boundaries. According to Rabinowitz and Conti (2010), in some instances, veterinarians have been asked to treat people or provide medical advice and medications that could be used for human beings as well as other animals, especially when dealing with zoonotic diseases. Veterinary health professionals need to be aware of the concept of professional scope of practice and the need not to exceed professional bounds in such situations.

Another issue that worth be cited is that finding the source of infection of a human psittacosis case that is the infected bird, is of utmost importance in avoiding further cases. For example, an infected animal recent acquired from a pet shop and linked to a psittacosis case, could have spread the illness to other animals within the shop, and to a large number of owners as newly purchased pets had been taken home (HALSBY, 2014; RASO et al. 2015). Usually, for the public health professional, stablishing the epidemiological features, as source of infection and susceptible population are the most important tools for developing prophylactic measures. In contrast, a clinician, whose primary role is to treat a disease, may be more concerned with the clinical symptoms or pathophysiology of the disease (NELSON, 2014). For instance, an infectious agent that causes CAP will be treated empirically with antibiotic therapy and symptomatic management of the pathophysiology, irrespective of how the infection was acquired or what the infectious organism is (NELSON; STEINHOFF, 2014). Linking persistent flu-like symptoms, respiratory distress, fever, chills, headache, weakness or fatigue to human psittacosis through a enquiry with regards to the patient medical history and professional and leisure occupations can bring contact with birds to light. At the healthcare-level, this may lead to a more adequately report symptomatic cases that have sought medical advice (BEECKMAN; VANROMPAY, 2009; GIBBONS et al., 2014). That means that physicians attending the community may aid veterinarians and public health workers in achieving the infected avian host, thus avoiding disease propagation.

On the other hand, physicians could become more aware of the routine contributions made by practicing veterinarians to human health, especially in educating person in contact with animals about the risks of acquiring zoonotic diseases. In this research, questionnaires were applied to identify possible risk factors husbandry practices of the owner towards their birds that could be involved in the epidemiology of psittacosis. Due to the small sample size of human psittacosis cases, the power of the statistical analysis was limited. Nonetheless, many pet owners reported having extremely close contact with their birds through mouth to beak contact, as kissing and sharing food with them, thus, all eligible participants were advised about preventive management to reduce chances of becoming infected with *C. psittaci* when dealing with or acquiring pets.

Some limitations of this study must be mentioned. First, this was a pioneering research including physicians and veterinarians on psittacosis/avian chlamydiosis in one human hospital. This represented a great initial shared strategy in order to maximize human and animal health, but it is unknown whether the results can be generalised. Second, in 57% of the patients, paired serum samples to a more accurately serological diagnosis could not be obtained. As previously mentioned, epidemiologists have established case definitions for psittacosis epidemiologic surveillance, nonetheless, these definitions are not used as the sole criteria for establishing clinical diagnoses and treatment (NASPHV, 2010). Many times, especially when not hospitalized, upon consultation patients seemed to understand the importance and agreed in returning for a second sample collection at first place, but after receiving suitably antibiotic therapy and getting better, they did not return to the hospital for revaluation and convalescent-serum sample gathering. In other cases, patients or their parents did not allow a second serum sample collection even when they were hospitalized, and their rights were respected. Third, from 47% of the psittacosis cases, biological samples of the birds related to the patient could not be obtained. At times, the bird died and was discarded before the patient became sick. Other times, obtaining the necessary follow-up information on the patient’s birds was not possible after the patient’s consultation; furthermore, possibly many owners do not truly believe that their pets might represent a health risk, once they do not show any signs of illness. Particularly for *C. psittaci*, it is not rare that infected birds are asymptomatic (FLAMMER, 1997).

Finally, it is clear that public health challenges concerning both human and animal health approaches still need to be overcome. However, as observed in this research direct communication and cooperation among veterinarians, physicians and other health care providers can truthfully improve knowledge of disease threats aiding in effective prophylactic measures for disease control.

REFERENCES

ASSOCIAÇÃO BRASILEIRA DA INDÚSTRIA PET (ABINPET). **Evolução de Mercado Anual**. 2015. Disponível em: <http://www.agricultura.gov.br/arq_editor/file/camaras_tematicas/Insumos_agropecuarios/79RO/IBGE_PAEB.pdf>. Acesso em: 21 jun. 2016.

BEECKMAN, D. S. A.; VANROMPAY, D. C. G. Zoonotic *Chlamydophila psittaci* infections from a clinical perspective. **Clinical Microbiology and Infection**, v. 15, n. 1, p. 11-17, 2009.

CENTER FOR DISEASES CONTROL AND PREVENTION (CDC). Case definitions for infectious conditions under public health surveillance. **Morbidity and Mortality Weekly Report**, v. 46, p. 1-55, 1997.

CDC, CENTER FOR DISEASES CONTROL AND PREVENTION. Human psittacosis linked to a bird distributor in Mississippi – Massachusetts and Tennessee, 1992. **Morbidity and Mortality Weekly Report**, v. 41, p. 794-797, 1992.

DAMBORG, P.; BROENS, E. M.; CHOMEL, B. B.; GUENTHER, S.; PASMANS, F.; WAGENAAR, J. A.; WEESE, J. S.; WIELER, L. H.; WINDAHL, U.; VANROMPAY, D.; GUARDABASSI, L. Bacterial zoonoses transmitted by household pets: state-of-the-art and future perspectives for targeted research and policy actions. **Journal of comparative pathology**, v. 55, supplement 1, 2015. doi http://dx.doi.org/10.1016/j.jcpa.2015.03.004, 2015.

DUMKE, R.; SCHNEE, C.; PLETZ, M. W.; RUPP, J.; JACOBS, E.; SACHSE, K. ROHDE, G.; CAPNETZ STUDY GROUP. *Mycoplasma pneumoniae* and *Chlamydia* spp. infection in community-acquired pneumonia, Germany, 2011–2012. **Emerging Infectious Disease**., v. 21, n. 3, p.426-434, 2015.

FERREIRA, V. L.; SILVA, M. V.; NASCIMENTO, R. D.; RASO, T. F. Psittacosis associated with pet bird ownership: a concern for public health. **JMM Case Reports**, v. 2, n. 5, 2015. DOI 10.1099/jmmcr.0.000085.

FLAMMER, K. Chlamydia. In: ALTMAN, R.; CLUBB, S.; DORRESTEIN, G. M.; QUESENBERRY, K. **Avian medicine and surgery**. Philadelphia: Saunders, 1997. p.364-378.

GAEDE, W.; RECKLING, K. F.; DRESENKAMP, B.; KENKLIES, S.; SCHUBERT, E.; NOACK, U.; IRMSCHER, H. M.; LUDWIG, C.; HOTZEL, H.; SACHSE, K. *Chlamydophila psittaci* infections in humans during an outbreak of psittacosis from poultry in Germany. **Zoonoses Public Health**, v. 55, n. 4, p.184-188, 2008.

GIBBONS, C. L.; MANGEN, M. J. J.; PLASS, D.; HAVELAAR, A.; BROOKE, R. J.; KRAMARZ, P.; PETERSON, K. L.; STUURMAN, A. L.; CASSINI, A.; FÈVRE, E. M.; KRETZSCHMAR, M. E. E. Measuring underreporting and under-ascertainment in infectious disease datasets: a comparison of methods. **BMC Public Health**, v. 14, n. 147, p. 1-17, 2014.

HALSBY, K. D.; WALSH, A. L.; CAMPBELL, C.; HEWITT, K.; MORGAN, D. Healthy Animals, healthy people: zoonosis risk from animal contact in pet shops, a systematic review of the literature. **PLoS One,** v. 26, n. 9, p.e89309, 2014.

HARKINEZHAD, T.; VERMINNEN, K.; VAN DROOGENBROECK, C.; VANROMPAY, D. *Chlamydophila psittaci* genotype E/B transmission from African grey parrots to humans. **J Medical Microbiology**. v. 56, p. 1097-100, 2007.

KALETA, E. F.; TADAY, E. M. A. Avian host range of *Chlamydophila* spp. based on isolation, antigen detection and serology. **Avian Pathology**, v. 32, n. 5, p. 435-462, 2003.

LAROUCAU, K.; SOURIAU, A.; RODOLAKIS, A. Improved sensitivity of PCR for *Chlamydophila* using *pmp* genes. **Veterinary Microbiology**, v. 82, p. 155-164, 2001.

NATIONAL ASSOCIATION OF STATE PUBLIC HEALTH VETERINARIANS (NASPHV). **Compendium of Measures to Control *Chlamydia psittaci* (Avian Chlamydiosis).** 2010. Disponível em: <http://www.NASPHVv.org/Documents/Psittacosis.pdf>. Acesso em: 13 maio 2016.

NELSON, K. E. Epidemiology of infectious disease: general principles. In: NELSON, K.E.; WILLIAMS, C.M. **Infectious disease epidemiology, theory and practice**. Burlington: Jones & Bartlett, 2014. p. 19-45.

NELSON, K. E.; STEINHOFF, M. C. The epidemiology of acute respiratory infections. In: NELSON, K. E.; WILLIAMS, C. M. **Infectious disease epidemiology, theory and practice.** Burlington: Jones & Bartlett, 2014. p. 561-610.

RABINOWITZ, P. M.; CONTI, L. A. **Human-animal medicine, clinical approaches to zoonoses, toxicants and other shared health risks**. Missouri: Saunders Elsevier, 2010, 411 p.

RASO, T. F. Clamidiose – Novas abordagens diagnósticas e terapêuticas. In: CUBAS, Z. S.; SILVA, J. C.; CATÃO-DIAS, J. L. **Tratado de animais selvagens: medicina veterinária**. 2 ed., São Paulo: Roca, 2014. p. 1369-1381.

RASO, T. F.; CARRASCO, A. O.; SILVA, J. C.; MARVULO, M. F.; PINTO, A. A. Seroprevalence of antibodies to *Chlamydophila psittaci* in zoo workers in Brazil. **Zoonoses Public Health**, v. 57, n. 6, p. 411-416, 2010.

RASO, T. F.; FERREIRA, V. L.; BASSETTI, B. R.; SZTAJNBOK, J.; KHOURY, Z.; NASCIMENTO, R. D.; SILVA, M. V. Psittacosis causing acute respiratory distress syndrome (ARDS). **J Pulm Respir Med**, v.5, n.4, p. 1-5, 2015 doi: 10.4172/2161-105X.1000283

RASO, T. F.; FERREIRA, V. L.; TIMM, L. N.; ABREU, M. F. T. Psittacosis domiciliary outbreak associated with monk parakeets (*Myiopsitta monachus*) in Brazil: need for surveillance and control. **JMM Case Reports**, v. 1, p. 1-4, 2014.

SILLIS, M.; LONGBOTTOM, D. Chlamydiosis. In: PALMER, S.; SOULSBY, L.; TORGERSON, P. R.; BROWN, D. W. G**. Zoonosis, clinical practice, and public health control**. Oxford: Oxford University Press, 2011. p. 146- 157.

SPOORENBERG, S. M.; BOS, W. J.; VAN , E. J.; DIJKSTRA, F.; HEDDEMA, E. R.; VAN VELZEN-BLAD, H.; HEIJLIGENBERG, R.; GRUTTERS, J. C.; DE JONGH, B. M.; OVIDIUS STUDY GROUP. *Chlamydia psittaci*: a relevant cause of community-acquired pneumonia in two Dutch hospitals. **Netherlands Journal of Medicine**, v. 74, n. 2, p. 75-81, 2016.

WARD, M. M. Estimating disease prevalence and incidence using administrative data: some assembly required**. The Journal of Rheumatology**., v. 40, n. 8, p. 1241-1243, 2013.

WOOLHOUSE, M. E.; GOWTAGE-SEQUERIA, S. Host range and emerging and reemerging pathogens. **Emerging Infectious Diseases**. v. 11, n. 12, p. 1842-1847, 2005.

YUNG, A. P.; GRAYSON, M. L. Psittacosis: a review of 135 cases. The **Medical Journal of Australia**, v. 148, p. 228–233, 1988.

4 INTERSECTORAL ACTION FOR HEALTH: PREVENTING PSITTACOSIS SPREAD AFTER ONE REPORTED CASE

ABSTRACT

Zoonotic diseases are a significant health threat for humans and animals. In the past, the epidemiology, etiology and pathology of infectious agents affecting humans and animals have mostly been investigated in separate studies. However, it is evident, that combined approaches are needed to understand geographical distribution, transmission and infection biology of zoonotic agents. Herein we report a severe case of psittacosis contracted from a infected pet bird. The epidemiological surveillance conducted by a multidisciplinary team was crucial to establish the infection dynamic across the human and bird hosts, i.e. the infection in the human patient, in her pet bird and in birds from the store where the pet was adquired. Thus, avoiding disease propagation. Reliable data about zoonotic diseases can only be generated through the application of multidisciplinary approaches which take into account the epidemiological factors and interactions of humans, animals and their environments as an integrated system.

Keywords: Birds disease. Pet. *Chlamydia psittaci*. Surveillance. Zoonosis

4.1 INTRODUCTION

Psittacosis is a zoonotic disease caused by an obligate intracellular Gram-negative bacterium *Chlamydia psittaci*. Transmission of *C. psittaci* usually originates from close contact with infected birds, particularly psittacine birds such as parrots, cockatiels, parakeets and lovebirds, mostly in the context of pet birds. Human infection with *C. psittaci* usually occurs when a person inhales organisms that have been aerosolized from dried feces or respiratory tract secretions from a diseased bird or asymptomatic carrier (NASPHV, 2010). Hence, handling the plumage and tissues of infected birds, cleaning cages and mouth to beak contact represent a zoonotic risk (BEECKMAN; VANROMPAY, 2009).

In humans, 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 a mild, non-specific illness to a systemic illness with severe pneumonia. Humans with symptomatic infections typically have an abrupt onset of fever, chills, headache, malaise, and myalgia. A nonproductive cough is usually present and can be accompanied by breathing difficulty and/or chest tightness. Most diagnoses are established by clinical presentation and positive antibodies against *C. psittaci* in paired sera using MIF methods (NASPHV, 2010).

In most of the countries, psittacosis is a notifiable disease and must be reported within 48 h (BOECK et al. 2016). In Brazil, psittacosis is not a notifiable disease (BRASIL, 2016), this is one factor that might contributes to the difficulty in determine its impact in human health. This is of particular relevance when taken into account the number of birds maintained as pets, 37 million, according to the Brazilian Association of Industries of Pet Products (Associação Brasileira da Indústria de Produtos para Animais de Estimação) (ABINPET, 2015). Furthermore, it is well established that *C. psittaci* infection in birds as well as avian chlamydiosis is endemic in the country, being reported in several species of captive and free-living birds (RASO et al., 2002; RASO et al., 2006; RASO et al., 2011, SANTOS et al.; 2014 FERREIRA et al., 2016).

In the past, the epidemiology, etiology and pathology of infectious agents affecting humans and animals have mostly been investigated in separate studies. However, it is evident that combined approaches are needed to understand geographical distribution, transmission and infection biology of zoonotic agents (REGIER et al., 2016). Successful zoonosis control requires coordinated action across the traditional professional lines of veterinary and human medicine as well as science (PALMER, 2011). The current paper describes the epidemiological surveillance of a severe psittacosis case, since the clinical suspicion of the human case until detecting *Chlamydia psittaci* infection in the patient´s pet. From that point on, also reaching the store and surviyng the birds were the infected pet was purchased. Moreover, it discusses the importance of a multidisciplinary working team in order to promptly halt the spread of the disease.

4.2 EPIDEMIOLOGICAL SURVEILLANCE DESCRIPTION

A female patient, 19 years, was admitted to the Infectology Institute Emilio Ribas (IIER), São Paulo, Brazil, presenting acute respiratory distress syndrome. The patient lived with her parents in São Paulo city. There was no history of recent travels. No significant past medical history regarding infectious diseases, smoking, illicit drug use or exposure to suspected ill individuals could be determined. The patient had been well until one week prior to her admission, when she started with dry cough, fever (38-39°C) and progressive dyspnea. Upon admission blood arterial gas saturation was 89% in room air conditions, thus she needed instantly respiratory support remaining in the Intensive Care Unit for 8 days and another 7 days in infirmary.

When talking to the patient’s mother, physicians find out that about four previous weeks she had bought a pet cockatiel (*Nymphicus hollandicus*), with which the patient maintained close interaction including mouth-to-beak contact. Thereby psittacosis was suspected. The epidemiological service of the IIER contacted the Psittacosis Research Group at the Pathology Department of São Paulo University (FMVZ/USP). Therefore, a patient’s serum sample was collected and sent to this Institution. The sample was tested for immunoglobulin IgM and IgG against *C. psittaci* by microimmunofluorescent (MIF) test kit (MIF, Foccus®, USA) according to the manufactures’ protocol. Antibodies against *C. psittaci*, with IgM serum titer of 80 and IgG 1:256 was revealed. A second serum sample from the patient could not be obtained.

Once human psittacosis was confirmed, a veterinarian of the FMVZ/USP evaluated the pet cockatiel from the patient. According to the family, the bird was in good health condition, nonetheless, while seen by a veterinarian, it was shown that the bird presented mild blepharitis, a clinical sign commonly observed in cockatiels with chlamydiosis. *C. psittaci* infection in the pet bird was determined after cloacal and ocular swab samples were collected and investigated by polymerase chain reaction (PCR) according to Laroucau et al. (2001). The bird was submitted to treatment with doxycycline during 45 days.

Next, the epidemiological service of the IIER contacted veterinarians from the Zoonosis Control Center of São Paulo Municipality (CCZ/SP), as well as health professionals from the Central Coordination of Health Surveillance (Covisa), and from the Epidemiological Surveillance Center of São Paulo State (CVE). In order to trace the source of infection, the official public health authorities found out the place where the cockatiel was purchased. It was a small, unventilated illegal establishment in an underprivileged neighbourhood. Over there, chickens (*Gallus gallus*), guineafowl (*Numida meleagris*) and muscovy ducks (*Cairina moschata*) were kept, slaughtered and sold clandestinely (Figures. 1 and 2). Moreover, a few individuals of others avian species such as cockatiels, pigeons (*Columba livia*) and budgerigars (*Melopsittacus undulatus*) were kept to be sold as pets. All birds were maintained in small enclosures in insalubrious conditions (Figure 3).

Veterinarians collected cloacal swabs samples from, pigeons (07), budgerigars (03), cockatiels (02), chickens (10), muscovy ducks (02) and guineafowl (01) in order to be evaluated for *C. psittaci*. The samples were processed by PCR and *C. psittaci* DNA was detected in all pigeons (07) and cockatiels (02). These infected birds were asymptomatic and intended to be sold as pets.

The local Official Veterinary Service demanded the closure of the establishment since the place had no license and permits requirements to raise or sell animals or derivate products. They also requested that all birds raised for consumption were euthanized at the place. In this case, the major public health concern was food poisoning from consumption of food items produced (meat and eggs), once the animals were maintained in unacceptable unhygienic conditions, without any kind of biosecurity measures to prevent diseases. Furthermore, blood samples were collected from some of the poultry with clinical signs suggestive of avian mycoplasmosis, which were seropositive for *Mycoplasma gallisepticum* when tested with Rapid serum agglutination test (Myco-Galli teste Biovet®, São Paulo, Brazil) as designed by OIE (2008). Despite not being a zoonotic disease, avian micoplasmosis represents a major threat to the to commercial poultry flocks, which is one of the main activities in Brazilian agribusiness with rigorous laws applying to it (BRASIL, 2009).

The birds (pigeons, cockatiels and budgerigars) intended to be traded as pets were taken to the Zoonosis Control Center of São Paulo Municipality (CCZ/SP) and treated with doxycycline for 45 days by a veterinarian specialized in avian medicine. The birds were retested upon antibiotic therapy completion and *C. psittaci*'s DNA was not detected in the molecular test performed. Besides, both the patient with psittacosis and her cockatiel had a prompt and uneventful recovery from the disease.

Figure 1 - A- Front of the store where birds infected with *Chlamydia psittaci* were sell as pets. **B -** Back of the store where chickens (*Gallus gallus*), guineafowl (*Numida meleagris*) and muscovy ducks (*Cairina moschata*) were kept, slaughtered and sold clandestinely. Psittacine birds found to be infected with *Chlamydia psittaci* were maintained to be sold as pets (arrow)

Font: (FERREIRA, V. L., 2016)

Figure 3 Conditions of birds maintained in an illegal store closed by official public health professionals. **A** – Guineafowl (*Numida meleagris*) seropositive for *Mycoplasma gallisepticum*, with swollen infraorbital sinus. **B, C -** Several species of birds maintained together in high densities under unsanitary conditions. **D** –Pigeons infected with *C. psittaci* intended for the trade of pets kept together with poultry intended for consumption. **E** – Illegal poultry meat processed in unhealthy environments intended for commerce

Font: (FERREIRA, V. L., 2016)

4.3 DISCUSSION

An integrated zoonotic disease surveillance system needs to detect disease emergence in human or animal populations at the earliest time possible. Multidisciplinary teams of professionals that have relevant expertise and field experience would identify populations at risk and causes and risk factors of infection, and then rapidly and widely disseminate this information so that immediate and longer-term disease prevention and control interventions can be implemented. The goal of these interventions would be to control the extent and geographic scope of the outbreak and to minimize morbidity, mortality, and economic losses in both human and animal populations (CHAN, 2009).

In the present case, a multidisciplinary team composed by veterinarians, physicians and other health professionals from different official services and institutions worked together in order to trace the origin of a severe human psittacosis case. At any rate, this investigation had this repercussion, with extensive alerts released by the epidemiological team of the hospital in which the patient was admitted, probably due to the severity of the patient's condition.

The disease notification, as well as the epidemiological surveillance work carried out, that is, the ongoing systematic collection, recording, analysis and interpretation of data, in this study is not the most common seen in psittacosis cases. From 2005 through 2009, 66 human cases of psittacosis were reported to the Centers for Disease Control and Prevention in USA (NASPHV, 2010). These cases are possibly an underestimation of the true incidence, given that even brief exposure can lead to systemic infection, and testing for *C. psittaci* is often not included in routine microbiological diagnostic panels for pneumonia (SPOORENBERG et al., 2016). Besides, it is important to keep in mind that individuals with mild cases may not seek medical attention contributing to the lack of epidemiological information of this disease (CDC, 1992).

Note that the IIER is one of the largest infectology centers in the country, whose medical staff is a reference in tropical medicine. Over there, psittacosis cases have been recognised, and thus reported to health surveillances agencies, after critically ill patients had undergone several other medical services without definitive diagnosis (FERREIRA et al., 2015; RASO et al., 2015). A possible explanation is that practitioners are not necessarily aware of the widespread occurrence and the zoonotic nature of avian chlamydiosis (BEECKMAN; VANROMPAY, 2009) neither of the populariry of birds as pets. In addition, even when they are, psittacosis might not be brought to light in first place because contact with birds is not suspected or because the symptoms caused by the *C. psittaci* infection are similar to those of other respiratory pathogens (BEECKMAN; VANROMPAY, 2009).

Unlike other countries, in Brazil psittacosis is not present in the list of notifiable diseases. However, there is a specification in that list declaring that any epizootic/ or animal death that could lead to any occurrence of disease in humans should be reportable (BRASIL, 2016). Thus, despite some states' efforts to increase disease reporting by health care providers, some public health experts believe that underreporting by providers is still a problem. Many health care providers do not fully understand their role in infectious disease surveillance, including the importance of prompt reporting of clinical information to relevant public health authorities (GAO, 2004).

As observed in the present report, promt notification is important to facilitate timely public health action. At the same time, through the combined efforts of veterinary and human health communities, the effectiveness of all surveillance for zoonotic diseases is truly enhanced (CROM, 2002). In this case, physicians assisted in the control of psittacosis through proper clinical diagnosis and treatment of the human patient, as well as notifying the disease, thus giving raise to the epidemiological investigation. In turn, veterinarians played a crucial role in evaluating, achieving the diagnosis and treating properly the patient's pet cockatiels. Conjointly, investigating *C. psittaci* infection and treating the birds from the illegal establishment, thus acting directly at the source of infection. Particularly in this investigation, veterinarians also helped to avoid spreading avian mycoplasmosis and potential food borne diseases of public health significance, since their action led to the closure of an illegal commerce of poultry and derivate products that were processed and sold without any kind of sanitary inspection.

The work carried out in this study highlights an important practical implication, that is, physicians and veterinarians are the key professionals to recognize and report zoonotic events (KAHN, 2007). It might sound obvious, but in practice, there is a lack of communication between these two health care fields. Enhanced communication between hospital epidemiologists, and local medical and animal public health officials would not only help to expedite a local response, but also help to identify whether unusual diseases or outbreaks involving animals and humans were related or separate events. This is of paramount importance to avoid diseases dissemination, since without integrated approaches, diseases of people and animals can move more quickly (KAHN, 2006).

In this case, additional possible psittacosis cases in owners of birds purchased from the retailer could not be identified. Neither could the birds be tracked to the respective distributors, since the retailer could not provide any of this information. Ideally, for accurate identification of owners and suppliers of infected birds, retailers and suppliers should maintain records identifying the origin and destination of birds (CDC, 1992). Even so, the current investigation was effective in preventing psittacosis propagation, once infected birds intended for the pet commerce were tested, retained, treated and retested.

Although case reports are based on an infection in a single patient, they may yield important epidemiologic information regarding the disease occurrence (GANGE; GOLUB, 2014). The present study brings alertness about a potential fatal zoonosis, which occurrence in Brazil is certainly underestimated. Taking into account the number of pet birds maintained in the country, 37 million (ABINPET, 2015), along with the lack of regulations and inspection concerning pet birds breeding and trade, how many people might buy infected birds and get sick?

Lastly, it is important to think out that surveillance and diagnosis are indispensable, however, they do not stop disease - they only identify it. Veterinarians and physicians need to become involved in all aspects of the agent/host/environment causation triad (KAHN, 2006; REGIER et al., 2016). Only in this way they can support and develop logical evaluations, design intervention strategies and ultimately truly effective prevention strategies.

REFERENCES

ASSOCIAÇÃO BRASILEIRA DA INDÚSTRIA PET (ABINPET). **Evolução de Mercado Anual**. 2015. Disponível em: <http://www.agricultura.gov.br/arq_editor/file/camaras_tematicas/Insumos_agropecuarios/79RO/IBGE_PAEB.pdf>. Acesso em: 21 jun. 2016.

BEECKMAN, D. S. A.; VANROMPAY, D. C. G. Zoonotic *Chlamydophila psittaci* infections from a clinical perspective. **Clinical microbiology and infection**, v. 15, n. 1, p. 11-17, 2009.

BOECK, C.; DEHOLLOGNE, C.; DUMONT, A.; SPIERENBURG, M.; HEIJNE, M.; GYSSENS, L.; VAN DER HILST, J.; VANROMPAY, D. Managing a cluster outbreak of psittacosis in Belgium linked to a pet shop visit in The Netherlands. **Epidemiology and Infection**, v.144, n.8, p. 1710-1716, 2016.

BRASIL. Ministério da Agricultura, Pecuária e Abastecimento. **Manual de Legislação – Programas Nacionais de Saúde Animal do Brasil**. Brasília, DF, 2009. 440 p.

BRASIL. Portaria n. 204, 17 de fevereiro de 2016. **Diário Oficial da União**, Brasília, 18 Fev. 2016. Sec. 1.

CDC, CENTER FOR DISEASES CONTROL AND PREVENTION. Human psittacosis linked to a bird distributor in Mississippi – Massachusetts and Tennessee, 1992. **Morbidity and Mortality Weekly Report**, v. 41, p. 794-797, 1992..

CHAN, M. Achieving an effective zoonotic disease surveillance system . In: KEUSCH, G. T.; PAPPAIOANOU, M.; GONZALEZ; M. C.; SCOTT, K.; TSAI; P. **Sustaining global surveillance and response to emerging zoonotic diseases.** Washington DC: The National Academy Press, 2009. p. 115-163.

CROM, R. L. Veterinary surveillance for zoonotic diseases in the United States. In: BURROUGHS, T.; KNOBLER, S.; LEDERBERG, J. **The emergence of zoonotic diseases:** understanding the impact on animal and human health**.** Washington, DC: National Academy of Sciences, 2002. p. 90-101.

FERREIRA, V. L.; SILVA, M. V.; NASCIMENTO, R. D.; RASO, T. F. Psittacosis associated with pet bird ownership: a concern for public health. **JMM Case Reports**, v. 2, n. 5, 2015. DOI 10.1099/jmmcr.0.000085.

FERREIRA, V. L.; DIAS, R. A.; RASO, T. F. Screening of feral pigeons (*Columba livia*) for pathogens of veterinary and medical importance. Revista Brasileira de Ciência Avícola, v.19, 2016. no prelo.

UNITED STATE GOVERNMENT ACCOUNTABILITY OFFICE. EMERGING INFECTIOUS DISEASES (GAO). Emerging infectious diseases: review of state and federal diseases survaillence efforts. Washington, DC.: Government Accountability Office, 2004. Report to the Chairman, Permanent Subcommittee on Investigations, Committee on Governmental Affairs, U.S. Senate. Disponível em: < http://www.gao.gov/products/GAO-04-877 >. Acesso em 28 Jul 2016.

GANGE, S. J.; GOLUB, E. T. Study design. In: NELSON, K. E.; WILLIAMS, C. M. Infectious disease epidemiology, theory and practice. Burlington: Apha Press, 2014. Cap. 3, p.45-72.

KAHN, L. H. Managing zoonotic disease risks: need for greater physician and veterinarian collaboration. **Journal of Chinese clinical medicine**, v.2, n.2, p.258-264, 2007.

KAHN, L. H. Confronting zoonoses, linking human and veterinary medicine. **Emerging Infectious Disease**. v. 12, n. 4, p. 556–561, 2006.

LAROUCAU, K.; SOURIAU, A.; RODOLAKIS, A. Improved sensitivity of PCR for *Chlamydophila* using *pmp* genes. **Veterinary Microbiology**, v. 82, p. 155-164, 2001.

NATIONAL ASSOCIATION OF STATE PUBLIC HEALTH VETERINARIANS (NASPHV). **Compendium of Measures to Control *Chlamydia psittaci* (Avian Chlamydiosis).** 2010. Disponível em: <http://www.NASPHVv.org/Documents/Psittacosis.pdf>. Acesso em: 13 maio 2016.

WORLD ORGANIZATION FOR ANIMAL HEALTH (OIE). Avian Mycoplasmosis**.** **Manual of Diagnostic Tests and Vaccines for Terrestrial Animals**. 2008. Disponível em < http:// www.oie.int/fileadmin/Home/eng/Health_standards/tahm/2.03.05_%20AVIAN_MYCO.pdf> Acesso em: 21 Jun 2016.

PALMER, S. The global chalenge of disease control. In: PALMER, S.; SOULSBY, L.; TORGERSON, P. R.; BROWN, D. W. G**. Zoonosis, clinical practice, and public health control**. Oxford: Oxford University Press, 2011. p. 3-12.

RASO, T. F.; FERREIRA, V. L.; BASSETTI, B. R.; SZTAJNBOK, J.; KHOURY, Z.; et al. Psittacosis causing acute respiratory distress syndrome (ARDS). **J Pulm Respir Med**, v.5, n.4, p. 1-5, 2015 doi: 10.4172/2161-105X.1000283

RASO, T. F.; GRESPAN, A.; FERREIRA, V. L.; PINHEIRO, S. R. *Chlamydophila psittaci* infection in pet cockatiels (*Nymphicus hollandicus*) in Brazil. In: INTERNATIONAL CONGRESS ON PATHOGENS AT THE HUMAN-ANIMAL INTERFACE (ICOPHAI): IMPACT, LIMITATIONS AND NEEDS IN DEVELOPING COUNTRIES, 1., 2011, Addis Abeba. **Proceedings**….Addis Abeba, 2011, p.41.

RASO, T. F.; SEIXAS, G. H. F.; GUEDES, N. M. R.; PINTO, A. A. *Chlamydophila psittaci* in free-living Blue-fronted Amazon parrots (*Amazona aestiva*) and Hyacinth macaws (*Anodorhynchus hyacinthinus*) in the Pantanal of Mato Grosso do Sul, Brazil. **Veterinary Microbiology**, v. 117, p. 235–241, 2006.

RASO, T.F.; BERCHIERI, JR., A.; PINTO, A. A. Evidence of *Chlamydophila psittaci* infection incaptive amazon parrots in Brazil. **Journal of Zoo and Wildlife Medicine**, v. 33, n. 2, p. 118–121, 2002.

REGIER, Y.; ROURKE, O.; KEMPF, V. A. Bartonella spp. - a chance to establish One Health concepts in veterinary and human medicine. **Parasit Vectors**, v. 9, n.1, p. 261, 2016. DOI 10.1186/s13071-016-1546-x.

SANTOS, F.; LEAL, D. C.; RASO, T. F.; SOUZA, B. M.; CUNHA, R. M.; MARTINEZ, V. H.; BARROUIN-MELO, S. M.; FRANKE, C. R. Risk factors associated with *Chlamydia psittaci* infection in psittacine birds. **Journal of medical microbiology**, v. 63, n. 3, p. 458-63, 2014.

SPOORENBERG, S. M.; BOS, W. J.; VAN , E. J.; DIJKSTRA, F.; HEDDEMA, E. R.; VAN VELZEN-BLAD, H.; HEIJLIGENBERG, R.; GRUTTERS, J. C.; DE JONGH, B. M.; OVIDIUS STUDY GROUP. *Chlamydia psittaci*: a relevant cause of community-acquired pneumonia in two Dutch hospitals. **Netherlands Journal of Medicine**, v. 74, n. 2, p. 75-81, 2016.

5 PSITTACOSIS ASSOCIATED WITH PET BIRD OWNERSHIP: A CONCERN FOR PUBLIC HEALTH

ABSTRACT

Psittacosis is a zoonotic infectious disease caused by *Chlamydia psittaci* and most cases involve avian contact history. In humans, psittacosis induces symptoms ranging from mild ‘flu-like’ symptoms to serious atypical pneumonia. Unless specifically thought of, the diagnosis of psittacosis can be missed and the disease is usually treated as atypical pneumonia. Here, we detail cases of psittacosis related to pet birds. A 16-year-old male was admitted with fever and persistent cough. The patient reported previous treatment with broad-spectrum antibiotics that led to limited improvement of his condition, and owning pet birds; thus, psittacosis was suspected. Serum samples from the patient were obtained and tested using a microimmunofluorescence assay, revealing an IgG titre of 64. An epidemiological investigation was conducted in five family members related to the patient; two possessed anti-*C. psittaci* antibodies (IgG titre 64 and IgM titre 20; IgG titre 128 and IgA titre 20). Additionally, *C. psittaci* DNA was detected by PCR carried out on the family’s pet birds. Psittacosis probably occurs more often than reported. Individuals with milder cases may not seek medical attention, and physicians may not inquire about bird exposure. There is a need for awareness campaigns directed at health workers and birds owners.

Key words**:** Atypical pneumonia. *Chlamydia psittaci*. Chlamydiosis. Psittacine. Zoonosis.

*Article published as follows: FERREIRA, V. L.; SILVA, M. V.; NASCIMENTO, R. D.; RASO, T. F. Psittacosis associated with pet bird ownership: a concern for public health. **JMM Case Reports**, v. 2, n. 5, 2015. DOI 10.1099/jmmcr.0.000085.

5.1 INTRODUCTION

Psittacosis is a zoonotic disease caused by the obligate intracellular bacterium *Chlamydia psittaci*. Clinical psittacosis in humans varies in severity from mild “flu-like” symptoms with fever, headache, sore throat and photophobia to a more serious atypical pneumonia with a dry cough and painful breathing. Infections primarily occur in bird owners, pet shop employees, breeders, poultry and wildlife workers who inhale aerosols from the feces and secretions of infected birds (VANROMPAY et al., 1995; KIRCHNER, 1997; NASPHV, 2010). In humans, psittacosis is typically diagnosed using a combination of clinical signs and serological tests. The most common test to confirm psittacosis is a microimmunofluorescence test (MIF) to determine if there has been an increase in the anti-*C. psittaci* antibody titer (NASPHV, 2010).

In birds, the disease resulting from *C. psittaci* infection is called chlamydiosis. Avian chlamydiosis signs are usually non-specific and include ruffled feathers, anorexia, discharge, and diarrhea. Infected birds usually remain asymptomatic and may intermittently shed the agent in respiratory secretions and feces, which is the source of infection for other avian species and humans (VANROMPAY et al., 1995).

Although notification is mandatory in most countries, the impact of psittacosis on human health is difficult to determine. This disease likely occurs more often than reported because individuals with mild cases may not seek medical attention and physicians may not inquire about bird exposure when evaluating patients. Additionally, antimicrobials employed empirically for therapy of community-acquired pneumonia may prevent an accurate diagnosis (CDC, 1992). This disease significance is becoming a more significant public health concern because of the popularity of pet birds and the placement of birds in childcare facilities, garden centers and rest homes (MATSUI et al. 2008; HARKINEZHAD et al. 2009). In this report, we present mild and severe cases of psittacosis in members of a family that owns pet birds.

5.2 CASE REPORT

A sixteen years old male patient was attended at the Ambulatory of Tropical Diseases and Zoonosis of the Infectology Institute Emilio Ribas, São Paulo, Brazil with chest pain and a cough that had persisted for 3 weeks. When he first sought medical assistance, he received treatment with azithromycin (500 mg once a day for 3 days) and prednisone (20 mg, once a day for 4 days), followed by treatment with levofloxacin (500 mg once a day for 7 days). His symptoms, including a fever (38.4ºC), returned after the antibiotic therapy was completed. A chest x-ray was performed, which revealed increased hilar bronchovascular markings, mild hyperlucency at the lung bases and subtle peri-hilar linear opacities in the middle lung fields, more conspicuous on the right side. The laboratory tests are summarized in Table 1.

The patient lived with his family (a mother, two brothers and one sister) in an urban area, and he reported no recent travel. He did not smoke or use illicit drugs, and he had not been exposed to individuals with a contagious illness. He did report having close contact with several psittacine birds every other weekend in his father's residence, which is a small apartment located in downtown São Paulo (Figure 1). The epidemiological profile associated with the patient's clinical symptoms led to a hypothesized diagnosis of psittacosis. To confirm the diagnosis, paired serum samples were collected with a 4-week interval in between samples and evaluated for anti-*C. psittaci* antibodies (IgG, IgM and IgA) using immunofluorescence (Chlamydia MIF Serology-Focus Diagnosis). The results revealed an IgG anti-*C. psittaci* titer of 32 in the first serological sample evaluated and a titer of 64 in the second sample. In both samples, IgM and IgA titers were not detected. The patient recovered completely following antibiotic therapy with doxycycline (100 mg twice a day for 14 days).

Because of the zoonotic nature of this disease, four family members who had contact with the pet birds as well as the patient's mother, who had no contact with the birds, were clinically evaluated. Serum samples were also collected from the family members for *C. psittaci* serological testing with MIF. Two family members possessed anti-*C. psittaci* antibodies. The patient's father, 51 years old, had an IgG titer of 64 and an IgM titer of 20; the patient's brother, 14 years old, had an IgG titer of 128 and an IgA of 20 (Table 2). Informed consent was obtained from all individuals who participated in this study.

The patient's father revealed that a few months earlier he had developed a persistent nonproductive cough that lasted for several weeks. He did not seek medical assistance because he attributed his condition to his smoking habits. The other patient's relatives did not report previous illnesses.

Because the pet birds were the potential sources of *Chlamydia psittaci* infection, a veterinarian went to the family's home to evaluate the birds. Cloacal swabs samples were collected from all of the birds, including 12 lovebirds (11 *Agapornis personata* and 1 *Agapornis roseicollis*), 21 cockatiels (*Nymphicus hollandicus*) and 2 yellow-chevroned parakeets (*Brotogeris chiriri*) (Figure 2). To determine if the birds were carrying *C. psittaci*, genomic DNA was extracted from the cloacal swab samples with a Nucleic Acid and Protein Purification Kit® (Macherey-Nagel, GmbH & Co. KG, Germany) according to the manufacturer's protocol. Our standard laboratory strain of *C. psittaci* from monk parakeets (*Myiopsitta monachus*) (Cpsi/Mm/BR01, GenBank number JQ926183.1) and ultrapure water were used as positive and negative controls, respectively. The PCR primers targeted a conserved region of pmp gene sequences that were reported previously (LAROUCAU et al., 2001). *C. psittaci* DNA was detected in the samples from one lovebird (*A. personata*) and two cockatiels (*N. hollandicus*). All of the birds presented no signs of disease.

5.3 DISCUSSION

Psittacosis in humans is considered to be a rare but potentially severe disease. Acute psittacosis cases, some with fatal outcomes, have been described in the literature (VERWEIJ et al., 1995; PETROVAY; BALLA, 2008; FRAEYMAN et al., 2010).

However, these cases are most likely only the tip of the iceberg. What goes undetected are less severe infections, which are either asymptomatic or misdiagnosed because the symptoms triggered by the infection are similar to those of other respiratory pathogens (HARKINEZHAD et al., 2007). In the current study, milder cases of psittacosis were diagnosed only after a patient with more severe symptoms sought medical assistance. Additionally, the third medical team that cared for the patient was the first team to raise the possibility of psittacosis and conduct a more thorough inquiry into the patient's history of bird exposure.

Concerning laboratory tests for psittacosis, the CDC (Centers for Disease Control and Prevention) has established case definitions for psittacosis. A patient has a confirmed case of psittacosis if the clinical illness is compatible with symptoms of the disease and there is a 4-fold or greater increase in antibodies against *C. psittaci* between paired acute and convalescent-phase serum samples. A case of psittacosis can also be confirmed if there are immunoglobulin M antibodies against *C. psittaci* with a reciprocal titer of ≥ 16 detected by MIF (CDC, 1997; NASPHV, 2010). According to this classification, the patient's father, who presented with a milder case of psittacosis, was considered to be a confirmed case (Table 2). A 4-fold increase in antibodies against *C. psittaci* between paired serums samples was not observed in the index case. Nevertheless, a delay in gathering the serum samples after the onset of symptoms may have affected the serological tests. Furthermore, treatment with antibiotics 2 weeks prior to testing may have inhibited the patient's antibody response, which has been reported previously in the literature (FRAEYMAN et al., 2010). The other case, the patient's brother, was likely an asymptomatic infection because the brother reported no illness even though he had anti-*C. psittaci* IgG and IgA titers as well as close contact with the infected pet birds.

Note that the CDC case definitions were established primarily for epidemiological purposes; these definitions should not be used as the sole criteria for establishing a clinical diagnosis (NASPHV, 2010). As observed in this study, psittacosis diseases can be highly variable with the severity of the disease ranging from asymptomatic infection or a mild non-specific illness to a more severe pneumonia (NASPHV, 2010; CHENG et al., 2013). This variability makes it challenging to confirm a psittacosis diagnosis. A history of bird contact is a valuable piece of information that should be collected during diagnosis because exposure to birds has been reported in 85% of psittacosis cases (YUNG; GRAYSON, 1988). The epidemiological investigation conducted in this study was very important for confirming the diagnosis because the source of the infection was determined through molecular detection of *C. psittaci* in the infected pet birds.

It should be emphasized that birds are among the most popular pets around the world. For example, in the USA, 8.3 million birds are kept as companion animals (AVMA, 2012). Nevertheless, public awareness about the risks of contracting zoonotic diseases from pet birds is very limited. Psittacine (e.g., macaws, parrots, budgerigars, cockatiels) birds are among the most popular birds kept as pets. These birds are also the main chlamydia-positive avian species (KALETA; TADAY, 2003; EVANS, 2011) and as observed in this study, they may represent a major risk for humans to contract psittacosis. The risk of infection with further disease development was enhanced by the 35 birds living in the family's apartment balcony. The risk was also enhanced for their neighbors because they were likely unaware of this potential source of infection and the possibility of acquiring a zoonosis. Therefore, this family's living situation poses a public health risk to the individuals living in the neighboring apartments.

Another concern is that only 12.3% of bird-owning households currently seek veterinary advice for their animals (AVMA, 2012). Birds infected with *C. psittaci* can be asymptomatic, and testing programs conducted by veterinarians could help protect humans from acquiring this zoonosis. In this report, the birds presented no evident clinical signs of disease, and the owners had never taken their pets to a veterinarian. Successful infection prevention requires that individuals in contact with animals be aware of the disease risks. If the family had been informed earlier about the zoonotic nature of avian chlamydiosis, effective prophylactic measures could have been taken earlier. Many of the disease risks associated with pet contact can be reduced through simple measures, such as proper animal selection and changes in animal contact (STULL et al., 2012). *C. psittaci* resists drying and may remain infectious for months, so the cages of infected birds must be disinfected thoroughly. Sick birds should be treated, and their handlers should wear protective clothing and a high-efficiency respirator (N95 rating) (CFSPH, 2009; NASPHV, 2010).

Improved zoonotic disease education is needed for pet-owning households. Pet birds should be bought from reputable suppliers and examined by a veterinarian when they are first acquired. Birds and cages should be kept in a well-ventilated area to prevent the accumulation of infectious dust. Cages should be cleaned regularly to prevent the build-up of waste, and they should be treated first with a cleaning solution to reduce aerosolization (CFSPH, 2009). Furthermore, increased communication between professions is needed to improve overall knowledge of zoonotic diseases and develop the optimal approaches for reducing pet-associated pathogen transmission (STULL et al., 2012). Therefore, additional effort from physicians and veterinarians is required.

Figure 1 - Cockatiels (*N. hollandicus*) related to psittacosis cases and kept as pet birds on a balcony´s apartment in São Paulo city. Note the high density of birds in a small place

Font: Picture provided by the pet bird owner, 2015

Figure 2 - Psittacine pet birds related to psittacosis cases and kept on a balcony´s apartment in São Paulo city. Note inappropriate management: cages arranged irregularly according to the space, feeders under perches allowing the presence of feces in them and dirt accumulation

Font: (FERREIRA, V.L, 2015)

Table 1 - Laboratorial tests for the sixteen year-old male patient with psittacosis

Leucogram	%	Results	Reference values
Leukocytes		13.7 x 10^3 cel/ mm^3	4-11 x 10^3 cel/ mm^3
Neutrophils	73,7%	10.1 x 10^3 cel/ mm^3	1.6- 7 x 10^3 cel/ mm^3
Eosinophils	2,5%	0.3 x 10^3 cel/ mm^3	0.05-0.5 x 10^3 cel/ mm^3
Basophils	0.6%	0.1 x 10^3 cel/ mm^3	0 - 0.2 x10^3 cel/ mm^3
Lymphocytes	15.1%	2.1 x 10^3 cel/ mm^3	0.9 - 3.4 x10^3 cel/ mm^3
Monocytes	8.1%	1.1 x 10^3 cel/ mm^3	0.2 – 0.9 x10^3 cel/ mm^3
C-reative protein		18,2 mg/L	< 5 mg/L

Font: (FERREIRA et al., 2015)

Table 2 - Clinical features and serology results for six individuals connected to the index case of psittacosis

Patient	Gender	Age (years)	Clinical features	Contact with *C. psittaci* infected birds	Antibody titers – MIF test first sample			Antibody titers – MIF test second sample		
					IgG	IgM	IgA	IgG	IgM	IgA
1	M	16	chest pain, cough, fever	yes	**32**	<10	<16	**64**	< 10	< 16
2	M	14	none	yes	**128**	<10	**20**	--	--	--
3	M	51	persistent cough	yes	**64**	**20**	<16	--	--	--
4	M	12	none	yes	< 16	< 10	<16	--	--	--
5	F	39	none	no	< 16	< 10	<16	--	--	--
6	F	10	none	yes	< 16	< 10	<16	--	--	--

-- Serum sample not collected

Font: (FERREIRA et al., 2015)

REFERENCES

AMERICAN VETERINARY MEDICAL ASSOCIATION (AVMA). **Pet Ownership & Demographics Sourcebook**. 2012. Disponível em: https:<//www.avma.org/KB/Resources/Statistics/Pages/Market-research-statistics-US-pet-ownership.aspx>. Acesso em: 21 jun 2016.

CDC, CENTER FOR DISEASES CONTROL AND PREVENTION. Human psittacosis linked to a bird distributor in Mississippi – Massachusetts and Tennessee, 1992. **Morbidity and Mortality Weekly Report**, v. 41, p. 794-797, 1992.

CENTER FOR DISEASES CONTROL AND PREVENTION (CDC). Case definitions for infectious conditions under public health surveillance. **Morbidity and Mortality Weekly Report**, v. 46, p. 1-55, 1997.

CFSPH. **Avian Chlamydiosis**. 2009. Disponível em: http:<//www.cfsph.iastate.edu/Factsheets/pdfs/chlamydiosis_avian.pdf> Acesso em: 21 jun 2016.

CHENG, Y. J.; LIN, K. Y.; CHEN, C. C.; HUANG, Y. L.; LIU, C. E.; LI, S. Y. Zoonotic atypical pneumonia due to *Chlamydophila psittaci*: First reported psittacosis case in Taiwan. **Journal of Formosan Medical Association**, v.112, p.430-433, 2013.

EVANS, E. E. Zoonotic diseases of common pet birds: psittacine, passerine, and columbiform species. **Veterinary Clinics of North America**, v. 14, n. 3, p. 457-76, 2011.

FRAEYMAN, A.; BOEL, A.; VAN VAERENBERGH, K.; DE BEENHOUWER, H. Atypical pneumonia due to *Chlamydophila psittaci*: 3 case reports and review of literature. **Acta Clinica Belgica**, v. 65, p. 192–196, 2010.

HARKINEZHAD, T.; VERMINNEN, K.; BUYZERE, M. D.; RIETZSCHEL, E.; BEKAERT S.; VANROMPAY, D. Prevalence of *Chlamydophila psittaci* infections in a human population in contact with domestic and companion birds. **Journal of Medical Microbiology**, v. 58, p. 1207-1212, 2009.

HARKINEZHAD, T.; VERMINNEN, K.; VAN DROOGENBROECK, C.; VANROMPAY, D. *Chlamydophila psittaci* genotype E/B transmission from African grey parrots to humans. **Journal of Medical Microbiology,** v.56, p.1097-100, 2007.

KALETA, E. F.; TADAY, E. M. Avian host range of *Chlamydophila* spp. based on isolation, antigen detection and serology. **Avian Pathology**, v. 32, p. 435–461, 2003.

KIRCHNER J. Psittacosis, is contact with birds causing your patients pneumonia? **Postgraduate Medicine**, v. 102, n. 2, p. 181-2, 187-8, 193-4, 1997.

LAROUCAU, K.; SOURIAU, A.; RODOLAKIS, A. Improved sensitivity of PCR for *Chlamydophila* using pmp genes. **Veterinary Microbiology**, v. 82, n. 2, p. 155-64, 2001.

MATSUI, T.; NAKASHIMA, K.; OHYAMA, T.; KOBAYASHI, J.; ARIMA, Y.; KISHIMOTO, T.; OGAWA, M.; CAI, Y.; SHIGA, S.; ANDO, S.; KURANE, I.; TABARA, K.; ITAGAKI, A.; NITTA, N.; FUKUSHI,H.; MATSUMOTO, A.; OKABE, N. An outbreak of psittacosis in a bird park in Japan. **Epidemiology and Infection**, v.136, n. 4, p. 492-5, 2008.

NATIONAL ASSOCIATION OF STATE PUBLIC HEALTH VETERINARIANS (NASPHV). **Compendium of Measures to Control *Chlamydia psittaci* (Avian Chlamydiosis).** 2010. Disponível em: <http://www.NASPHVv.org/Documents/Psittacosis.pdf>. Acesso em: 13 maio 2016.

PETROVAY, F., BALLA, E. Two fatal cases of psittacosis caused by *Chlamydophila psittaci*. **J Med Microbiol**. v.57, p.1296-1298, 2008.

STULL, J. W.; PEREGRINE, A. S.; SARGEANT, J. M.; WEESE, J. S. Household, knowledge, attitudes and practices related to pet contact and associated zoonoses in Ontario, Canada. **BMC Public Health**, v. 12, p. 553, 2012.

VANROMPAY, D.; DUCATELLE, R.; HAESEBROUCK, F. *Chlamydia psittaci* infections: a review with emphasis on avian chlamydiosis. **Veterinary Microbiology**. v. 45, p. 93-119, 1995.

VERWEIJ, P.E.; MEIS, J.F.; EIJK, R.; MELCHERS, W. J.; GALAMA, J. M. Severe human psittacosis requiring artificial ventilation: case report and review. **Clinical Infectious Disease** v. 20, n. 2, p. 440-2, 1995.

YUNG, A. P.; GRAYSON, M. L. Psittacosis: a review of 135 cases. The **Medical Journal of Australia**, v. 148, p. 228–233, 1988.

6 FINALS CONSIDERATIONS

In Brazil, in the last decade, considerable evidence indicating that *Chlamydia psittaci* is endemic among birds has been obtained (RASO, 2014). However, zoonotic transmissions as well as the impact of *C.psittaci* infections on human health needs to be better understood. The fact that the overall number of reported cases remained generally low is, to some extent, due to the absence of this pathogen in most routine diagnostic schemes. The symptoms in affected individuals are mainly non-specific and influenza-like, but severe pneumonia are not uncommon (KNITTLER et al. 2014). The present research demonstrates human psittacosis occurrence of 27% (10/37) in patients with previous contact with birds. According to Spoorenberg et al. (2016), doctors should be aware of the possibility of psittacosis in any case of community-acquired pneumonia.

In addition, efforts to control avian chlamydiosis should be enhanced by veterinarians and public health authorities. In the current research, psittacoses patients reported having contact with birds in which *C. psittaci* was laboratorial confirmed. In previous studies, Santos et al. (2014) found a 17% *C.psittaci* infection prevalence in birds from pet shops and Raso et al. (2011) 90% prevalence in cockatiels intended to the pet commerce. This provides an opportunity to anticipate disease in the human population and prevent its appearance by implementing appropriate control measures.

Unquestionably, surveillance is of little use if not shared with other groups or individuals who can act on the information to prevent or diagnose disease. In this context, veterinary-human medical partnerships can expand the horizons of medical care (RABINOWITZ; CONTI; 2010). Yet professional interaction of this type is often limited, it is possible, brings many benefits and should be encourage. As described in these thesis chapters, a multisciplinary team composed by veterinarian and physician allowed the first epidemiological study on human psittacosis in a hospital setting in which the source of infection was also investigated.

Hopefully in the future researches, diagnosticians, laboratory scientists and practicioners will take a more holistic view of health; and change the many overlaps between human and animal medicine and health.

REFERENCES

ASSOCIAÇÃO BRASILEIRA DA INDÚSTRIA PET (ABINPET). **Evolução de Mercado Anual**. 2015. Disponível em: <http://www.agricultura.gov.br/arq_editor/file/camaras_tematicas/Insumos_agropecuarios/79RO/IBGE_PAEB.pdf>. Acesso em: 21 jun. 2016.

BEECKMAN, D. S. A.; VANROMPAY, D. C. G. Zoonotic *Chlamydophila psittaci* infections from a clinical perspective. **Clinical Microbiology and Infection**, v. 15, n. 1, p. 11-17, 2009.

HALSBY, K. D.; WALSH, A. L.; CAMPBELL, C.; HEWITT, K.; MORGAN, D. Healthy Animals, healthy people: zoonosis risk from animal contact in pet shops, a systematic review of the literature. **PLoS One,** v. 26, n. 9, p.e89309, 2014.

KALETA, E. F.; TADAY, E. M. A. Avian host range of *Chlamydophila* spp. based on isolation, antigen detection and serology. **Avian Pathology**., v. 32, n. 5, p. 435-462, 2003.

KNITTLER, M. R.; BERNDT, A.; BÖCKER, S.; DUTOW, P.; HÄNEL, F.; HEUER, D.; KÄGEBEIN, D.; KLOS, A.; KOCH, S.; LIEBLER-TENORIO, E.; OSTERMANN, C.; REINHOLD, P.; SALUZ, H. P.; SCHÖFL, G.; SEHNERT, P.; SACHSE, K. *Chlamydia psittaci*: New insights into genomic diversity, clinicalpathology, host–pathogen interaction and anti-bacterial immunity. **International Journal of Medical Microbiology**, v. 304, .n. 7, p.877-93, 2014.

NATIONAL ASSOCIATION OF STATE PUBLIC HEALTH VETERINARIANS (NASPHV). **Compendium of Measures to Control *Chlamydia psittaci* (Avian Chlamydiosis).** 2010. Disponível em: <http://www.NASPHVv.org/Documents/Psittacosis.pdf>. Acesso em: 13 maio 2016.

RABINOWITZ, P. M.; CONTI, L. A. **Human-animal medicine, clinical approaches to zoonoses, toxicants and other shared health risks**. Missouri: Saunders Elsevier, 2010, 411p.

RASO, T. F. Clamidiose – Novas abordagens diagnósticas e terapêuticas. In: CUBAS, Z. S.; SILVA, J. C.; CATÃO-DIAS, J. L. **Tratado de animais selvagens: medicina veterinária**. 2 ed., São Paulo: Roca, 2014. p. 1369-1381.

RASO, T. F.; CARRASCO, A. O.; SILVA, J. C.; MARVULO, M. F.; PINTO, A. A. Seroprevalence of antibodies to *Chlamydophila psittaci* in zoo workers in Brazil. **Zoonoses Public Health**, v. 57, n. 6, p. 411-416, 2010.

RASO, T. F.; FERREIRA, V. L.; TIMM, L. N.; ABREU, M. F. T. Psittacosis domiciliary outbreak associated with monk parakeets (*Myiopsitta monachus*) in Brazil: need for surveillance and control. **JMM Case Reports**, v. 1, p. 1-4, 2014.

RASO, T. F.; GRESPAN, A.; FERREIRA, V. L.; PINHEIRO, S. R. *Chlamydophila psittaci* infection in pet cockatiels (*Nymphicus hollandicus*) in Brazil. In: INTERNATIONAL CONGRESS ON PATHOGENS AT THE HUMAN-ANIMAL INTERFACE (ICOPHAI): IMPACT, LIMITATIONS AND NEEDS IN DEVELOPING COUNTRIES, 1., 2011, Addis Abeba. **Proceedings**….Addis Abeba, 2011, p.41.

RODOLAKIS, A.; MOHAMAD, K. Zoonotic potential of *Chlamydophila*.**Veterinary Microbiology**, v. 140, n. 3, p. 382-391, 2010.

SACHSE, K.; BAVOIL; P.; KALTENBOECK, B.; STEPHENS, R.; KUO, C.; ROSSELLÓ-MÓRA, R.; HORN, M. Emendation of the family Chlamydiaceae: proposal of a single genus, *Chlamydia*, to include all currently recognized species. **Systematic and applied microbiology**, v. 38, n. 2, p. 99-103, 2015.

SANTOS, F.; LEAL, D. C.; RASO, T. F.; SOUZA, B. M.; CUNHA, R. M.; MARTINEZ, V. H.; BARROUIN-MELO, S. M.; FRANKE, C. R. Risk factors associated with *Chlamydia psittaci* infection in psittacine birds. **Journal of medical microbiology**, v. 63, n. 3, p. 458-63, 2014.

SPOORENBERG, S. M.; BOS, W. J.; VAN , E. J.; DIJKSTRA, F.; HEDDEMA, E. R.; VAN VELZEN-BLAD, H.; HEIJLIGENBERG, R.; GRUTTERS, J. C.; DE JONGH, B. M.; OVIDIUS STUDY GROUP. *Chlamydia psittaci*: a relevant cause of community-acquired pneumonia in two Dutch hospitals. **Netherlands Journal of Medicine**, v. 74, n. 2, p. 75-81, 2016.

VANROMPAY, D.; DUCATELLE, R.; HAESEBROUCK, F. *Chlamydia psittaci* infections: a review with emphasis on avian chlamydiosis. **Veterinary Microbiology**, v. 45, p. 93-119, 1995.

YUNG, A. P.; GRAYSON, M. L. Psittacosis: a review of 135 cases. The **Medical Journal of Australia**, v. 148, p. 228–233, 1988.