Knowledge about bacterial and viral pathogens present in wild mammals in Chile: a systematic review

Sebastián Llanos-Soto[1] and Daniel González-Acuña[2]

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This study organizes all available information about viral and bacterial pathogens of wild mammals in Chile. This was done in order to identify pathogens that have been well-documented and recognize those that have not been properly studied, determine the number of articles that have been published annually about this topic and identify regions in Chile that concentrate the highest and lowest number of studies concerning viral and bacterial pathogens. A total of 67 scientific articles published in peer-reviewed journals from 1951 to 2018 were selected for revision. Results indicate that the number of publications has increased per decade but there are years in which no articles were published. Most studies addressed *Leptospira,* rabies, hantavirus, *Mycobacterium avium paratuberculosis* (MAP) and distemper. Rodentia, Carnivora, Chiroptera and Cetartiodactyla were the most studied mammal orders. Information about presence/absence of pathogens was found for 44 wild mammal species. Research was mainly carried out in central and southern Chile and the most commonly employed methods for pathogen diagnosis were serology and molecular techniques. Overall, research in wild mammals has been directed towards the evaluation of zoonotic diseases, while vector-borne and non-zoonotic diseases have been mostly neglected by the scientific community over the years.

Keywords: Bacteria; Chile; mammals; virus; zoonoses.
Palabras clave: Bacteria; Chile; mamíferos; virus; zoonosis.

[1]Laboratorio de Enfermedades y Parásitos de Fauna Silvestre. Departamento de Ciencia Animal. Facultad de Ciencias Veterinarias, Universidad de Concepción, Chile.
[2]Laboratorio de Vida Silvestre. Departamento de Ciencia Animal. Facultad de Ciencias Veterinarias, Universidad de Concepción, Chile.

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Corresponding author:
Daniel González Acuña
Facultad de Ciencias Veterinarias
Universidad de Concepción
Av. Vicente Méndez 595
Chillán, Chile.
Phone: +56 422208739
Fax: +56 422208739
danigonz@udec.cl

Introduction

Knowledge about the presence of infectious diseases in wildlife is pivotal to understand the potential consequences that they might have on species conservation and their threat to human health[1]. Anthropogenic factors, such as the introduction of alien species, climate change, habitat loss and fragmentation and human encroachment in natural areas might increase the risk of disease transmission from wild reservoirs to domestic animals and humans[2,3].

Diseases that are transmitted between animals and humans are called zoonoses and cause both economical and social losses, especially in underdeveloped and developing countries[3]. Zoonotic pathogens, such as rabies and hantavirus, have their origin in mammal reservoirs and are considered extremely important for public health systems because of their consequences on human health[4,5]. In this context, affected individuals may have their health compromised by zoonotic diseases, and in many cases, they might be wrongly characterized as common infections or even go unnoticed to health care institutions[6]. Urbanization and industrial activities, such as agriculture and forestry, have intensified in Chile during recent years and they will probably continue to do so in the future[7]. These factors may lead to habitat fragmentation, ecosystem disruption and over-exploitation of species, which added to the expansion of human and domestic animal populations in areas close to natural habitats, might contribute to the transmission of infectious diseases from free-living wild animals to domestic animals and humans[8-11].

The objectives of this review were to (1) gather and organize all the information available in articles published in peer-reviewed journals involving the assessment of viral and bacterial infections in Chilean wild mammals, (2) identify which pathogens have been prioritized by the local scientific community and which have received little to no attention, (3) evaluate the number of articles published annually about the prevalence of viral and bacterial pathogens in wild mammal hosts and (4) recognize the number of studies developed in this topic in the different regions of Chile.

Materials and Methods

Peer-reviewed scientific publications evaluating the prevalence of viral and bacterial pathogens in Chilean wild

mammals were searched and listed following the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) declaration guidelines[12]. Articles published in Spanish or English from January 1950 through July 2018 were selected for revision. Data from unpublished literature (grey literature) was excluded from this study (*i.e.*, abstracts, books, local bulletins and presentations at scientific conferences) based on the fact that these types of scientific documents do not often undergo a rigorous peer-review process prior to publication. This means that the accuracy, reliability and quality of the findings being presented in these documents cannot be ensured. Viral and bacterial agents were considered pathogenic when there was information available in the literature indicating their ability to cause disease in animals, humans or both. The revision considered pathogens present in free-living native and introduced mammalian species, with the exception of domestic animals and individuals maintained in captivity in zoos, farms and exhibition centers. Publications about pathogens found in mammals from the Chilean Antarctic Territory were also excluded from the review, as they have been already reviewed in another scientific article[13].

Google Scholar (https://scholar.google.cl/), Scielo Scientific Library (http://www.scielo.cl/) and PubMed (https://www.ncbi.nlm.nih.gov/pubmed/) databases were used to conduct an extensive search for publications. Keywords "bacteria", "bacterial", "*Brucella*", "coronavirus", "*Corynebacterium*", "distemper", "hantavirus", "herpesvirus", "infectious disease", "*Leptospira*", "*Mycobacterium*", "*Mycoplasma*", "parvovirus", "pathogen", "picornavirus", "rabies", "retrovirus", "*Salmonella*", "vector-borne", "viral", "virus", "zoonosis" AND "Carnivora", "Cetartiodactyla", "Chiroptera", "Didelphimorphia", "Lagomorpha", "mammal", "Microbiotheria", "Paucituberculata", "Rodentia", "wildlife", "Xenarthra" AND "Chile" were inputed in independent searches. These same keywords were also employed to perform searches in Spanish to account for publications in local journals. Articles that were not available for download online were physically searched in the libraries from Universidad de Chile, Universidad Austral, Pontificia Universidad Católica de Chile and Universidad de Concepción. Information about prevalence for each pathogen was organized and listed in a supplementary table using Microsoft® Excel 2010. The table included data about mammal host, pathogen characterization (*e.g.*, serotype, genetic lineage, class), region in Chile where the study was developed and technique used for diagnosis. Data from selected publications was analyzed using line graphs to evaluate a potential trend in the number of articles published per decade since 1951 and identify the most studied mammal orders in Chile.

Data was searched for a total of 150 mammal species from eight orders: Didelphimorphia (2), Paucituberculata (1), Microbiotheria (1), Chiroptera (11), Xenarthra (3), Rodentia (63), Cetartiodactyla (47), Lagomorpha (2) and Carnivora (20). Three species were excluded from the review because their distribution was restricted to Antarctica (*Ommatophoca rossii*) or their presence in Chile has yet to be confirmed (*Stenella attenuate* and *Stenella longirostris*). Research studies that included previously diagnosed cases of viral and bacterial infections in their analysis (*e.g.*, positive cases of rabies diagnosed by the Chilean National Institute of Public Health, ISP in Spanish) were also included in this review. Pathogens were listed from highest to lowest in the discussion depending on the number of articles available in the literature related to that specific pathogen.

Results

A total of 67 publications about viral and bacterial pathogens in Chilean mammals were included in this review. From the literature assessed, 34 studies evaluated the prevalence of viral pathogens and 35 studies concerned bacteria. Information about presence/absence of pathogens was found for 44 species from the Rodentia (15), Carnivora (10), Chiroptera (9), Cetartiodactyla (8), Didelphimorphia (1) and Lagomorpha (1) orders (Figure 1). Details about the results of this review are indicated in the Supplementary Table 1.

Overall, the number of studies addressing the infection or exposure to viral or bacterial pathogens in wild mammals has increased per decade (Figure 2), however, during the last decade there are years in which the number of articles published ranges from 0 to 2 (*i.e.*, 2010, 2012 and 2016). Publications dedicated to each pathogen varied in number, most studies were related to *Leptospira* (16 studies), rabies virus (12 studies), hantavirus (10 studies), *Mycobacterium avium paratuberculosis* (8 studies) and canine distemper virus (6 studies).

The first study to record the presence of pathogens in wild mammals in Chile was performed by Neghme et al. in 1951 and involved the evaluation of *Leptospira* in brown rats (*Rattus norvegicus*) captured in a slaughterhouse from the Metropolitana region[14]. Rodentia was the most studied mammal order with 27 publications assessing the presence of viral and bacterial pathogens. A high number of articles were dedicated to investigate the long-tailed pygmy rice rat (*Oligoryzomys longicaudatus*), with 19 scientific publications involving the evaluation of *Leptospira* and hantavirus in this species. Order Carnivora (16), Chiroptera (12) and Cetartiodactyla (12) have also received attention from the scientific community. Only two studies involved a lagomorph species and a single article concerned a member of the Didelphimorphia order. No study determined infection with viral or bacterial

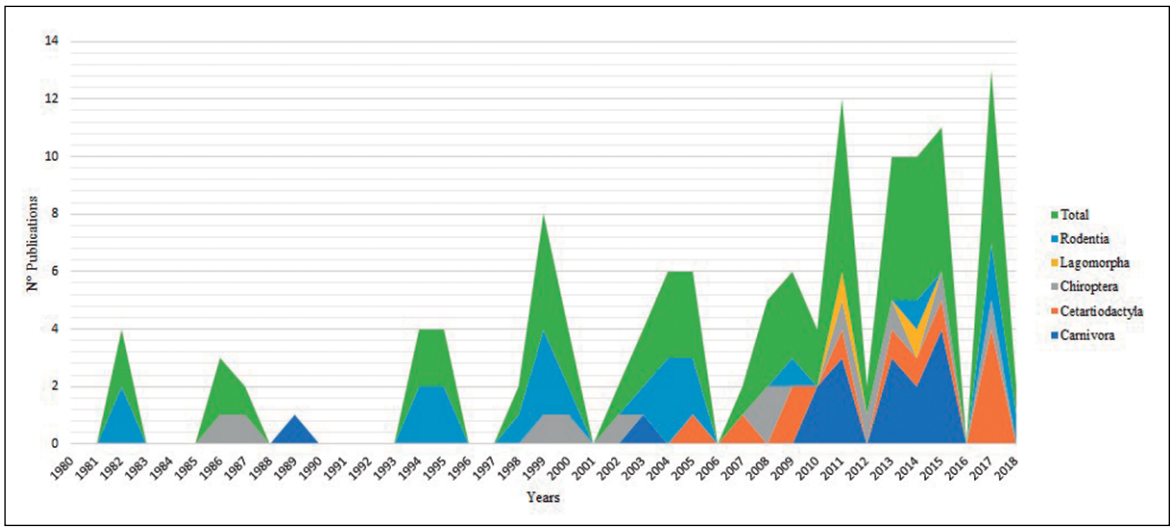

Figure 1. Number of scientific studies regarding viral or bacterial infections in different mammal orders from 1980 to 2018.

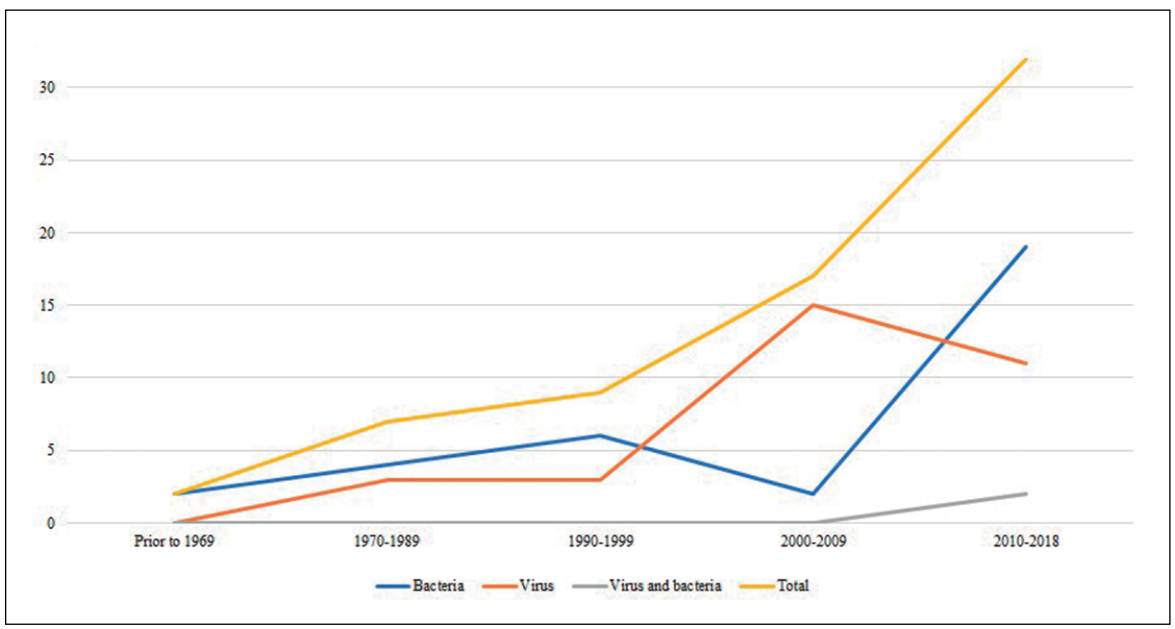

Figure 2. Number of scientific studies assessing the prevalence of viral or bacterial infections in wild mammals in Chile from 1951 to 2018.

pathogens in members of the orders Paucituberculata, Microbiotheria and Xenarthra.

A high number of studies of viral and bacterial infections were focused in southern and central Chile, particularly from the Coquimbo to the Los Lagos regions. The most studied regions were the Los Ríos region with 24 studies, the Metropolitana region with 14 studies and the Los Lagos region with 13 studies. Only three studies in rabies and a single one in hantavirus were carried out in the Maule Region. Research studies including mammal species from the northern regions of Chile are lacking, with two studies in the Antofagasta region and a sigle one in the Tarapacá and the Atacama regions. Research has not been carried out in the Arica y Parinacota region. A total of 7 and 4 studies have been developed in the regions of Aysén and Magallanes, respectively.

Most studies applied serological methods for pathogen diagnosis in mammal hosts, such is the case of the distemper virus and parvovirus and the bacterium *Brucella*, which have been only assessed using serology. These methods were also commonly applied in research studies involving hantavirus in wild rodents. The application of molecular methods to detect pathogens in wild mammals has raised in the last two decades in Chile, which has

been reflected in an increase in the use of these type of techniques for diagnosing viruses and bacteria in wild mammals. The use of other methods, such as direct immunofluresence, bacterial culture and histopathology has been mostly restricted for the specific diagnosis of certain pathogens.

Discussion

Wild mammals have played a crucial role as reservoirs of infectious diseases in developing countries and have been involved in the occurrence of spill-over epizootic events in human populations[3]. These events negatively impact public health systems in these countries and result in important economic losses[3,4]. In Chile, most scientists and governamental institutions have destined their efforts in studying those zoonotic agents considered as a serious threat to human health, such as *Leptospira*, rabies virus and hantavirus. Pathogens restricted to animal hosts and which do not represent a threat to humans (*e.g.*, canine distemper virus) have only recently began to receive scientific attention and, in consequence, information available about them is much more limited.

Leptospira

Leptospirosis is a cosmopolitan zoonotic disease of great relevance for human health, particularly in developing and low-income countries, where sanitary conditions and resources destined for disease diagnosis and prevention are limited[15]. The agent responsible for causing this disease is *Leptospira*, a bacterial pathogen capable of infecting a variety of mammal hosts and survive for several months in the environment in areas with warm and humid climate conditions[16].

Rodents play an important role in the maintenance and dissemination of pathogenic and non-pathogenic *Leptospira* to other mammal species in urban and rural areas of Chile, including humans and domestic animals[17,18]. To this date, evidence of infection by this pathogen have been shown in nine rodent species present in the country; the degu (*Octodon degus*), olive-colored akodont (*Abrothrix olivaceus*), long-haired akodont (*Abrothrix longipilis*), Darwin's leaf-eared mouse (*Phyllothis darwini*), long-tailed pygmy rice rat (*O. longicaudatus*), black rat (*Rattus rattus*), brown rat (*R. norvergicus*), house mouse (*Mus musculus*) and long-clawed mole mouse (*Geoxus valdivianus*)[17-27]. *Rattus norvegicus* and *R. rattus* are particularly relevant reservoirs due to the high prevalence of *Leptospira* documented in these species in rural areas of the country[18,27]. This bacterium has been described in wild mammals in south-central areas of Chile, and more specifically in the Metropolitana, Los Ríos, Los Lagos and Aysén regions.

Reports indicating the presence of *Leptospira* in other mammal orders are scarce in comparison to the information available for rodents. This bacterium has been documented in only two carnivore species; the South American sea lion (*Otaria flavescens*) and the American mink (*Neovison vison*)[9,28]. The interaction between wild animals and domestic reservoirs (*i.e.,* dogs and livestock) can act as an important mechanism for the ocurrence of leptospirosis cases in human inhabited environments[29]. Future studies dedicated to evaluate the role of wild species in the maintainance and transmission of Leptospira will make an important contribution to the understanding about the dynamics of this pathogens in natural and urban areas of the country.

Rabies virus

Rabies is a cosmopolitan zoonotic disease of great importance for public health worldwide[30]. All mammals are susceptible to rabies virus but only chiropterans and carnivores are capable of successfully maintain and transmit the infection in the long-term[30]. In Chile, confirmed cases of rabies are characterized by the ISP in different antigenic variants using specific monoclonal antibodies. Each variant represents an assemblage of viruses within a serotype that possesses defined antigenic properties[31]. This method of antigenic characterization of rabies has been widely applied in the country to study the geographical and temporal distribution of the virus[32,33].

The first case of human rabies in Chile was reported in 1879 and surveillance in domestic and wild species has been performed since 1929 by the ISP (at the time called "Instituto Bacteriológico de Chile")[34,35]. The efforts of the ISP allowed to identify wildlife and domestic species prevalent to the virus, amongst them livestock, lagomorphs, rodents, carnivores and chiropterans[33,34]. However, active surveillance of rabies began following the first oubreak of this disease in bats, more specifically, in the brazilian free-tailed bat (*Tadarida brasiliensis*) in 1985[33]. After this event, rabies cases have been mainly reported in bats[33,36].

To this date, five antigenic variants have been identified in Chile, including the canine antigenic variant AgV2 and four variants associated with insectivorous bats; the antigenic variant AgV4 *Tadarida*, antigenic variant AgV6 *Lasiurus*, antigenic variant AgV3 *Myotis chiloensis* and an antigenic variant AgV not typed for *Histiotus*[33,37,38]. The most common variant registered in Chile from 2008 to 2013 has been AgV4[39]. Antigenic variants are described to be host-specific [38,40], but recent studies suggest that there could be cross-species spillover transmission of rabies variants in bats[32,38,41,42].

Six of the eleven species of bats distributed in the country have been described to be infected with the virus; the big-eared brown bat (*Histiotus macrotus*), small big-

eared brown bat (*Histiotus montanus*), mouse-eared bat (*M. chiloensis*), eastern red bat (*Lasiurus borealis*), hoary bat (*Lasiurus cinereus*) and Brazilian free-tailed bat (*T. brasiliensis*)[32,33,37,43]. Considering the available scientific information, *T. brasiliensis* and *L. cinereus* remain the most relevant reservoirs of bat rabies in Chile[32,36,37,41,43,44]. Escobar et al. (2013) indicate that both *T. brasiliensis* and *L. cinereus,* and their respective antigenic variants of rabies (AgV4 and AgV6), share similar ecological niches in Chile and their distribution is limited by the presence of natural ecological barriers, such as the Andes Mountains to the east and the Pacific Ocean to the west[32]. Bat rabies seems to follow a seasonal pattern with peaks of positivity during hot season (October-March) and decrease in colder months[33,39]. This could be related to a reduction in bat activity during winter in the Southern hemisphere and therefore less chance of rabies transmission among bats and from bats to other susceptible species[33,39,45].

The increase in population size and density of dogs, added to the presence of bat (mainly *T. brasiliensis*) in urban areas, have raised the risk of rabies transmission events from bats to dogs across the country[46,47]. This is particularly important for central Chile, which possesses a high number of bat-related antigenic variants and an increased richness of chiropteran species[33]. Highly populated human settlements, such as the large cities located in the Metropolitana, Maule, Biobío and Valparaíso regions, posses an elevated risk of rabies transmission between animals and humans and concentrate the higher number of human cases[33,39,47]. Rabies cases have been reported in wild carnivores in Chile in years prior to 1990[48], however, insectivorous bats are currently considered the most important reservoir of rabies in the country with the 97.31% (1339/1376) of positive cases reported by the ISP from 1985 to 2012[32]. The importance of bats as rabies reservoirs and the increasing population of stray dogs in in urban areas rise concern about the current risk of rabies transmission from bats to dogs and, ultimately, humans beens[47].

Hantavirus

Seven rodent species have been found to be exposed or infected with the Andes virus in Chile; the olive-colored akodont (*A. olivaceus*), long-haired akodont (*A. longipilis*), Darwin's pericote (*P. darwini*), Southern big-eared mouse (*Loxodontomys micropus*), black rat (*R. rattus*), brown rat (*R. norvergicus*) and long-tailed pygmy rice rat (*O. longicaudatus*)[49-57]. Reports of hantavirus belong to areas in south and central Chile, particularly from the Coquimbo to Magallanes regions. To this date, the presence of hantavirus in the country has been restricted solely to rodents and has not been possible to identify the virus in wild species from other orders, such as the case of marsupials and chiropterans[54].

Oligoryzomys longicaudatus is the most important reservoir of the Andes virus in Chile, being responsible of disseminating the hantavirus to humans along its distributional ranged from the Copiapó to the Magallanes regions[54,57-59]. *Oligoryzomys longicaudatus* has been characterized as a high mobility species, which increases the risk of encountering humans throughout its wide home range (320-4800m^2)[60]. This species can be found in close proximity to urban areas, inhabiting humid environments and areas covered by bushes near water sources[60]. Increase in wild rodent population density in peri-urban areas has been linked to an elevation in the number of cases of hantavirus cardiopulmonary syndrome (HCPS) in humans[51]. It has been suggested that the explosive increase in the number of rodents during years of synchronized flowering of native bamboo, particularly *Chusquea quila*, might be implicated in the occurrence of hantavirus outbreaks in humans due to an increased available of food provided by these plants[51,52]. Most cases of hantavirus infection in Chile occur near towns in rural and peri-urban areas where *O. longicaudatus* is present and people are constantly exposed to become infected with the virus[55,61,62].

Mycobacterium

Mycobacterium avium subp. *paratuberculosis* (MAP) is an important pathogen of cattle and small ruminants, responsible for causing important economic losses in animal production worldwide[63]. In Chile, native ungulates are at continuous risk of becoming infected with MAP due to the high prevalence of this pathogen reported in livestock, particularly in southern areas of the country[64]. The guanaco (*Lama guanicoe*), the Southern pudu (*Pudu puda*), the Chilean Huemul (*Hippocamelus bisulcus*) and the European hare (*Lepus europaeus*) have been reported to carry this bacterium in Chile[65-69]. MAP-infected guanacos did not display any health-related issues and European hares did not present microscopic or macroscopic lesions associated with the infection[65,68]. In the case of the huemul, the bacterium presented similar molecular characteristics to MAP isolates commonly reported in livestock in Chile, suggesting that the latter are spreading the infection to huemul populations[70]. The situation is concerning for this endangered deer species, particularly in central Chile, where the population of huemuls is facing severe conservation issues and its habitat is being disturbed by the presence of domestic animals[71]. Similarly, MAP infections were documented in three Southern pudus found in areas commonly occupied by dairy cattle[67]. Currently, there is no information about pathological findings of MAP infection in free-ranging pudúes, however, this bacterium was indicated as the cause of death for an individual maintained in captive settings[66].

Infections with MAP have also been reported in introduced deer species in southern Chile, such as the red deer

(*Cervus elaphus*) and fallow deer (*Dama dama*), which could be transmitting the pathogen to livestock and vice versa[72,73]. The European wild boar (*Sus scrofa*) is currently distributed in rural and protected areas of south-central Chile and is considered as a carrier of MAP in Europe[74,75]. Currently, there have not been reports of MAP infection in this species in the country. The role that introduced ungulates species are playing in the transmission of MAP and other pathogens to native wildlife and livestock is a topic that still needs to be explored in Chile.

Canine distemper virus

Distemper is a viral disease prevalent in dogs all over the world and capable causing severe illness in wild carnivores[76]. A wide range of species from different families, such as the Canidae, Felidae, Hyaenidae, Mustelidae, Ursidae, Vivirridae and Procyonidae have been reported to be exposed or infected to canine distemper virus (CDV), in some cases, with major population declines[76,77]. CDV currently possesses an endemic status in urban and rural populations of dogs in Chile, with seroprevalences that range from 51 to 73%[10,78]. Dogs have been indicated as the source of CDV outbreaks in populations of the South American gray fox (*Lycalopex griseus*) in central Chile[10,79,81] and may be a threat to populations of the endangered Darwin's fox (*Lycalopex fulvipes*) in south-central Chile[82].

To this date, there are no study that applied methods to directly detect distemper virus in Chilean wildlife, however, serological surveys have found exposure to the virus in the American mink (*N. vison*), South American sea lion (*O. flavescens*), South American gray fox (*L. griseus*) and Andean fox (*Lycalopex culpaeus*)[8,9,79,80,81]. There is no information about infection or exposure to CDV in wild felids and indigenous mustelids in the country.

Neglected pathogens and wild mammal species

This section includes pathogens that have received little attention from the scientific community in Chile and information about their presence in natural hosts is currently lacking or inexistent.

Parvoviruses have been detected in a wide range of wild carnivores around the world, belonging to the Canidae, Felidae and Mustelidae families[83]. In Chile, canine (CPV) and feline parvoviruses (FPV) are recognized affections of dogs and cats, respectively[84]. Serological assessments of CPV in *L. culpaeus*, *L. griseus* and *O. flavescens* have found past exposure to this virus[9,10,80], meanwhile, there is no information about the exposure or infection with FPV in wild species. Both CPV and FPV, have shown to cause gastrointestinal affections in carnivores elsewhere[83], but infections and potential pathological consequences of these viruses on Chilean species are still undetermined.

Like parvoviruses, information about retroviruses in Chilean wildlife is lacking. Mora et al. (2015) found that guignas in Chiloé were infected with feline leukemia (FeLV) and feline immunodeficiency viruses (FIV) with no apparent clinical signs[85]. Nucleotide sequences obtained from FeLV and FIV in guignas were almost identical to those found in domestic cats, suggesting that cats may be playing an important role in the transmission of retroviruses to wild felids. Some feline species, such as the guigna, inhabit in areas close to human settlements and they ocassionally predate on poultry[86], which increases the risk of interacting with infected domestic cats. Retroviruses have been detected in the cougar and other large felids in North America[87], however, the presence of these viral agents in populations of cougars and most of the native wild felids species in Chile still needs to be addressed.

Information about viral and bacterial pathogens in native ruminants is very scarce. Bovine viral diarrhea virus (BVDV) was detected in the southern pudu and two Chilean huemuls were found to be exposed to the virus[88,89]. Similar to MAP, BVDV isolates from the southern pudu share molecular characteristics with viruses circulating in cattle, suggesting that livestock are acting as disseminators of pathogens to native wildlife in Chile[70,88]. Other pathogens present in livestock, such as bovine rhinotracheitis virus (BoHV-1) and *Brucella spp.*, were assessed in the Chilean huemul using serological methods, but no individual were found to be exposed[89].

There is no information about infections with viral or bacterial pathogens in aquatic carnivores, such as members of the Otariidae, Phocidae and Mustelidae families, except for the South American sea lion (*O. flavescens*) and the invasive American mink (*N. vison*). Furthermore, cetaceans have been mostly overlooked by the local scientific community, with only one viral disease being identified in Chile[90,91]. The Chilean dolphin (*Cephalorhynchus eutropia*), black porpoise (*Phocoena spinipinnis*), bottle-nosed dolphin (*Tursiops truncatus*) were found to present marks on their bodies typical of tattoo skin disease, an affection that might lead to neonatal mortality and have negative consequences on host population dynamics[90,91]. No other pathogens have been reported in cetaceans distributed in Chile, despite the fact that exposure to cetacean morbillivirus and *Brucella* sp. was described in species living along the Peruvian section of the Pacific Ocean[92-94].

Tick-borne pathogens

Ticks are hematophagous ectoparasites of almost every terrestrial vertebrate and play an important role as vector of pathogens[95]. In Chile, viral and bacterial tick-borne pathogenic agents have been neglected and information available about the presence of *Anaplasma platys*[96], *Ehrlichia canis*[97], "*Candidatus* Rickettsia andeanae"[98] and *Rickettsia felis*[99] is restricted to domestic mammals.

However, a recent study identified "*Candidatus* Neoehrlichia chilensis" in wild rodent species from southern Chile using molecular methods[100].

Recently, *Borrelia burgdorferi* was reported in Brazil, Mexico, Canada, Chile, Costa Rica, Colombia and Venezuela[101], however, most cases have been diagnosed based only on clinical and serological evidence, without a molecular characterization and isolation of the agent[101]. This has only been done by Ivanova et al. (2014), who reported *Borrelia chilensis* VA1, a new spirochete species from the Lyme group[102]. Additionally, Verdugo et al. (2017), found infection with *B. chilensis* in *Ixodes stilesi* ticks collected from the native southern pudu deer and suggests that *I. stilesi* may be playing a role in the maintainance of the spiroquete[103]. Further studies are necessary to properly understand the mechanisms of natural transmission of this bacterium and the risks of infection for domestic animals and humans.

Pathogen transmission between wildlife and domestic animals

Pathogen transmission between wild species and livestock is bidirectional and affect both animal production and species conservation all over the world[104]. Factors, such as human encroachment into wildlife inhabited areas and the expansion and intensification of animal production systems to natural areas, can increase the risk for contact and pathogen transmission at the livestock-wildlife interface[105]. The interaction among livestock and wild species not only occurs in anthropogenically disturbed zones, but also in protected natural areas of Chile[71,106]. Pathogens, such as MAP, BVDV and *Corynebacterium pseudotuberculosis*, are being transmitted from farm animals to wild ungulates facing conservation issues[70,88,107]. For this reason, it is extremely relevant to understand the consequences that these infectious agents might have on the health of the affected species and the existent mechanisms for the transmission of diseases between livestock animals and wildlife.

Dogs are another threat for Chilean wildlife due to their predatory behaviour over native species and for their role as carriers of infectious pathogens[108]. Dogs inhabiting natural areas have been linked with outbreaks of viral diseases in carnivore populations that have resulted in mass mortality events of wild animals in the past[109]. Dogs populations have increased in size and density over the years in urban and rural areas of Chile, which might increase the possibility of encounters between wildlife and domestic dogs and the transmission of pathogenic organisms[10,46,78,79,110].

Conclusions

To date, most publications have involved the study of zoonotic viral and bacterial pathogens in Chilean wild mammals. Non-zoonotic and vector-borne pathogens have been neglected by the local scientific community, despite their importance for wildlife conservation and public health, respectively. It is also concerning that a large number of studies have been performed in southern and central regions of Chile, while the development of research studies in the northern areas of the country have been limited. Research about viral and bacterial pathogens in Chilean wild mammals is still very scarce and further studies are necessary in order to properly understand the role that certain species might be playing as reservoir of infectious agents. The information gathered in future investigations dedicated to evaluate the presence of infection in wild mammals will establish the basis for more complex studies destined to understand the epidemiology and ecology of zoonotic and non-zoonotic infectious diseases in the country.

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Annexed. Summary of peer-reviewed studies published from 1951 to 2017 evaluating the prevalence of bacterial and viral infections in Chilean wildlife

Pathogen	Host Order	Host	Pathogen characterization	Prevalence/ Number of positives (percentage)	Location	Diagnostic technique	Reference
Rabies (Rhabdoviridae)	Chiroptera	Brazilian free-tailed bat *Tadarida brasiliensis*	Not determinded	12/73 (14.1%)	Metropolitana (9), Valparaíso (2), O'Higgins region (1)	Direct immunofluorescence and mouse inoculation tests	36
			Not determined	3/3 (100%)	Metropolitana region	Direct immunofluorescence and mouse inoculation test	111
			Not determined	1/619 (0.16%)	Metropolitana region	Direct immunofluorescence	36
			Antigenic variant 4 (AgV4)	104 positives	Metropolitana (60), Valparaíso (13), O'Higgins (13), Maule (8), Biobío (7), Coquimbo (4), Araucanía (2) and Los Lagos (1) regions	Mouse inoculation test and indirect immunofluorescence	112
			Genetic lineage B	1 positive	Not specified	Mouse inoculation test and RT/PCR	41
			Genetic lineage C	2 positives	Metropolitana and Valparaíso regions	Mouse inoculation test and RT/PCR	41
			Genetic lineage D	82 positives	Coquimbo to Los Lagos regions[a]	Mouse inoculation test and RT/PCR	41
			Genetic lineage E	3 positives	Metropolitana region	Mouse inoculation test and RT/PCR	41
			Genetic lineage A Antigenic variant 4 (AgV4)	2 positives	Not specified	Mouse inoculation test, indirect immunofluorescence and RT/PCR	113
			Genetic lineage B Antigenic variant 4 (AgV4)	4 positives	Not specified	Mouse inoculation test, indirect immunofluorescence and RT/PCR	113
			Genetic lineage C Antigenic variant 4 (AgV4)	27 positives	Not specified	Mouse inoculation test, indirect immunofluorescence and RT/PCR	113
			Antigenic variant 4 (AgV4)	672 positives	Metropolitana (260), Biobío (158), Valparaíso (88), O'Higgins (48), Maule (45), Los Lagos (44), Coquimbo (17), Araucanía (10) and Atacama regions (2)	Direct immunofluorescence	37
			Not determined	297 positives	Metropolitana region	Direct immunofluorescence	43
			Antigenic variant 4 (AgV4)	568 positives	Not specified	Mouse inoculation test and Direct immunofluorescence	38
			Antigenic variant 9 (AgV9)	4 positives	Not specified	Mouse inoculation test and Direct immunofluorescence	38
			Cluster I	64 positives	Not specified	RT/PCR and DNA sequencing	38
			Cluster III	1 positive	Not specified	RT/PCR and DNA sequencing	38
			Cluster IV	1 positive	Not specified	RT/PCR and DNA sequencing	38
			Antigenic variant 4 (AgV4)	910 positives	From Coquimbo to Los Ríos regions	Direct immunofluorescence	32
			Not determined	1243/23868 (4.95%)	Not specified	Direct immunofluorescence	33
			Not determined	856 positives	Not specified	Direct immunofluorescence	39

Annexed. Continuation

Pathogen	Host Order	Host	Pathogen characterization	Prevalence/ Number of positives (percentage)	Location	Diagnostic technique	Reference
	Chiroptera	*Histiotus* sp.	Antigenic variant not typed	13 positives	Metropolitana (3), Valparaíso (3), Biobío (3) and Magallanes regions (4)	Direct immunofluorescence	37
	Chiroptera	Big-eared brown bat *Histiotus macrotus*		0/4	Not specified	Direct immunofluorescence and mouse inoculation test	36
			Genetic lineage A Antigenic variant 4 (AgV 4)	1 positive	Not specified	Mouse inoculation test, indirect immunofluorescence and RT/ PCR	113
			Genetic lineage A Antigenic variant not typed	9 positives	Not specified	Mouse inoculation test, indirect immunofluorescence and RT/ PCR	113
			Not determined	3 positives	Metropolitana region	Direct immunofluorescence	43
			Cluster III	9 positives	Not specified	RT/PCR and DNA sequencing	38
			Not determined	24/188 (11.32%)	Not specified	Direct immunofluorescence	33
			Not determined	14 positives	Not specified	Direct immunofluorescence	39
	Chiroptera	Small big-eared brown bat *Histiotus montanus*	Not determined	1/7 (12.50%)	Not specified	Direct immunofluorescence	33
	Chiroptera	*Lasiurus* sp.	Genetic lineage E	1 positive	Biobío region	Mouse inoculation test and RT/PCR	41
			Genetic lineage B Antigenic variant 4 (AgV4)	5 positives	Not specified	Mouse inoculation test, indirect immunofluorescence and RT/ PCR	113
			Antigenic variant 6 (AgV6)|	27 positives	Metropolitana (19), Valparaíso (1), O'Higgins (2) and Biobío regions (5)	Direct immunofluorescence	37
	Chiroptera	Eastern Red Bat *Lasiurus borealis*		0/8	Not specified	Direct immunofluorescence and mouse inoculation test	36
			Not specified	4 positives	Metropolitana region	Direct immunofluorescence	43
			Antigenic variant 6 (AgV6)	4 positives	Not specified	Direct immunofluorescence and mouse inoculation tests	38
			Cluster IV	4 positives	Not specified	RT/PCR and DNA sequencing	38
			Not determined	14/81 (14.74%)	Not specified	Direct immunofluorescence	33
			Not determined	9 positives	Not specified	Direct immunofluorescence	39
	Chiroptera	Hoary bat *Lasiurus cinereus*	Not determined	19 positives	Metropolitana region	Direct immunofluorescence	43
			Antigenic variant 6 (AgV6)	14 positives	Not specified	Direct immunofluorescence and mouse inoculation tests	38
			Cluster IV	11 positives	Not specified	RT/PCR and DNA sequencing	38
			Antigenic variant 6 (AgV6)	52 positives	From Metropolitana to Los Ríos regions	Direct immunofluorescence	32
			Not determined	44/131 (25.14%)	Not specified	Direct immunofluorescence	33
			Not determined	37 positives	Not specified	Direct immunofluorescence	39

Annexed. Continuation

Pathogen	Host Order	Host	Pathogen characterization	Prevalence/ Number of positives (percentage)	Location	Diagnostic technique	Reference
	Chiroptera	Southern Red Bat *Lasiurus blossevillii*		0/1	Not specified	Direct immunofluorescence	33
	Chiroptera	*Myotis* sp.	Genetic lineage D Antigenic variant 3 (AgV3)	2 positives	Not specified	Mouse inoculation test, indirect immunofluorescence and RT/PCR	113
	Chiroptera	Mouse-eared bat *Myotis chiloensis*	Genetic lineage A	1 positive	Valparaíso region	Mouse inoculation test and RT/PCR	41
			Antigenic variant 3 (AgV3)	7 positives	Metropolitana (2), Atacama (1), Valparaíso (1), Araucanía (1) and Los Lagos (2) regions	Direct immunofluorescence	37
			Not determined	2 positives	Metropolitana region	Direct immunofluorescence	43
			Antigenic variant 4 (AgV4)	2 positives	Not specified	Direct immunofluorescence and mouse inoculation tests	38
			Antigenic variant 3 (AgV3)	5 positives	Not specified	Direct immunofluorescence and mouse inoculation tests	38
			Antigenic variant 8 (AgV8)	2 positives	Not specified	Direct immunofluorescence and mouse inoculation tests	38
			Cluster II	6 positives	Not specified	RT/PCR and DNA sequencing	38
			Not determined	13/1210 (1.06%)	Not specified	Direct immunofluorescence	33
			Not determined	9 positives	Not specified	Direct immunofluorescence	39
	Chiroptera	Vampire bat *Desmodus rotundus*		0/3	Not specified	Direct immunofluorescence	33
	Chiroptera	Kalinowski's Mastiff Bat *Mormopterus kalinowskii*		0/8	Not specified	Direct immunofluorescence	33
	Carnivora	South American gray fox *Lycalopex griseus*	Not determined	5/58 (8.62%)	Magallanes region: Bernardo O'Higgins (1), San Gregorio (1), Morro Chico (1), Porvenir (2)	Direct immunofluorescence and mouse inoculation tests	48
	Carnivora	*Lycalopex* sp.		0/120	Not specified	Direct immunofluorescence	33
	Carnivora	Molina's Hog-nosed skunk *Conepatus chinga*		0/5	Not specified	Direct immunofluorescence	33
	Carnivora	Lesser grison *Galictis cuja*		0/4	Not specified	Direct immunofluorescence	33
	Carnivora	South American sea lion *Otaria byronia*		0/3	Not specified	Direct immunofluorescence	33
	Carnivora	Guiña *Leopardus guigna*		0/3	Not specified	Direct immunofluorescence	33
	Carnivora	Pampas cat *Leopardus colocolo*		0/1	Not specified	Direct immunofluorescence	33
	Rodentia	Coypu *Myocastor coypus*		0/2	Not specified	Direct immunofluorescence	33
	Carnivora	Mountain lion *Puma concolor*		0/1	Not specified	Direct immunofluorescence	33

Annexed. Continuation

Pathogen	Host Order	Host	Pathogen characterization	Prevalence/ Number of positives (percentage)	Location	Diagnostic technique	Reference
Distemper (CDV)	Carnivora	*Lycalopex* spp.	Not determined	14/33 (42%)	Coquimbo region	Microneutralization assay	10
	Carnivora	South American gray fox *Lycalopex griseus*	Not determined	1 positive	Biobío region	ELISA	83
			Not determined	13/28 (46.4%)	Coquimbo region	Microneutralization test and Cytopathic effect in cell culture	79
	Carnivora	Andean fox *Lycalopex culpaeus*	Not determined	1/5 (20%)	Coquimbo region	Microneutralization test and Cytopathic effect in cell culture	79
			Not determined	8/16 (50%)	Metropolitana (7) and O'Higgins region (1)	Indirect ELISA and seroneutralization test	80
	Carnivora	South American Sea Lion *Otaria byronia*	Not determined	2/3 (66.7%)	Los Ríos region	Seroneutralization test	9
	Carnivora	American mink *Neovison vison*	Not determined	9/23 (39.1%) or 5/23 (21.7%)[b]	Los Ríos region	Microneutralization test	10
Hantavirus-Andes	Rodentia	Long-tailed Pygmy Rice Rat *Oligoryzomys longicaudatus*	Not determined	13/102 (12.74%)	Aysén region	ELISA	49
			Not determined	12.7%[c]	Aysén region	Serology[d]	50
			Not determined	24 positives	Aysén (11), O'Higgins (5), Biobío (2), Araucanía (3), Los Ríos (1), Los Lagos (1) and Metropolitana regions (1)	Serology[d]	51
			Not determined	18/59 (13.51%)[e]	Los Ríos (10) and Los Lagos regions (4)	ELISA	52
			Not determined	2 positives	Biobío region	ELISA	53
			Not determined	20/209 (9.57%)	Los Lagos region	ELISA	59
			Not determined	5/48 (10.4%)	Biobío (2), Valparaíso (1), O'Higgins (1), Maule (1), Araucanía, Los Ríos and Los Lagos regions	Strip immunoblot assay (SIA)	54
			Not determined	1/69 (1.44%)	Magallanes region	Strip immunoblot assay (SIA) and RT-PCR	57
				0/3	Metropolitana region	ELISA	56
	Rodentia	Olive-colored akodont *Abrothrix olivaceus*	Not determined	6/80 (7.5%)	Aysén region	ELISA	49
			Not determined	7.50%[c]	Aysén region	Serology[d]	50
			Not determined	4/547 (0,73%)	Metropolitana, Biobío, O'Higgins, Araucanía, Los Lagos and Aysén (4) regions	Serology[d]	51
				No positives[f]	Los Lagos and Los Ríos regions	ELISA	52
				0/98	Los Lagos region	ELISA	59
				0/127	From Valparaíso to Los Lagos regions including the Metropolitana region	Strip immunoblot assay (SIA)	54

Annexed. Continuation

Pathogen	Host Order	Host	Pathogen characterization	Prevalence/ Number of positives (percentage)	Location	Diagnostic technique	Reference
	Rodentia	Long-haired akodont *Abrothrix longipilis*	Not determined	1/36 (2.78%)	Aysén region	ELISA	49
			Not determined	2.70%	Aysén region	Serology[d]	50
			Not determined	12/300 (4%)	Los Ríos (3), Biobío (2), Araucanía and Aysén regions (2)	Serology[d]	51
			Not determined	4/43 (9.3%)	Los Ríos (4) and Los Lagos regions	ELISA	52
			Not determined	2/44 (4.6%)	Biobío region	ELISA	53
			Not determined	3/163 (1.84%)	Los Lagos region	ELISA	59
				No positives[g]	Valparaíso, O'Higgins, Maule, Biobío, Los Ríos and Los Lagos regions	Strip immunoblot assay (SIA)	54
				0/29	Metropolitana region	ELISA	56
	Rodentia	Sanborn's akodont *Abrothrix sanborni*		No positives[g]	Los Ríos region	Strip immunoblot assay (SIA)	54
	Rodentia	Darwin's leaf-eared mouse *Phyllotis darwini*	Not determined	2/61 (3.3%)	Metropolitan region	Serology[d]	51
				No positives[g]	O'Higgins region	Strip immunoblot assay (SIA)	54
	Rodentia	Southern big-eared mouse *Loxodontomys micropus*	Not determined	1 positive	Biobío region	Serology[d]	51
				0/8	Los Lagos region	ELISA	59
			Not determined	1/19 (5.3%)	Biobío region	ELISA	53
				No positives[g]	Los Lagos region	Strip immunoblot assay (SIA)	54
	Rodentia	House mouse *Mus musculus*		No positives[g]	Valparaíso, Araucanía and Metropolitana regions	Strip immunoblot assay (SIA)	54
				0/24	Antofagasta and Metropolitana regions	ELISA and PCR	55
	Rodentia	Black rat *Rattus rattus*		No positives[g]	Valparaíso, Maule, Biobío and Los Lagos regions	Strip immunoblot assay (SIA)	54
			Not determined	1/57 (1.75%)	Coquimbo (1), Valparaíso, Metropolitana, Araucanía and Los Lagos regions	ELISA and PCR	55
				0/2	Metropolitana region	ELISA	56
	Rodentia	Brown rat *Rattus norvegicus*		No positives[g]	Valparaíso, O'Higgins, Maule, Biobío and Metropolitana regions	Strip immunoblot assay (SIA)	54
			Not determined	2/80 (2.5%)	Metropolitana region	ELISA and PCR	55
			Not determined	1/6 (16.66%)	Metropolitana region	ELISA, RT-PCR and sequencing	56
	Didelphimorphia	Elegant Fat-tailed opossum *Thylamys elegans*		No positives[g]	Valparaíso and O'Higgins regions	Strip immunoblot assay (SIA)	54
	Rodentia	Degu *Octodon degus*		0/25	Metropolitana region	Serology[d]	51
	Rodentia	Chilean climbing mouse *Irenomys tarsalis*		0/2	Los Ríos region	ELISA	59
	Rodentia	Pearson's long-clawed mouse *Geoxus annectens*		0/1	Los Ríos region	ELISA	59

Annexed. Continuation

Pathogen	Host Order	Host	Pathogen characterization	Prevalence/ Number of positives (percentage)	Location	Diagnostic technique	Reference
Hantavirus - Seoul	Rodentia	Brown rat *Rattus norvegicus*	Not determined	2 positives	Not specified	Not specified	55
Canine parvovirus (CPV)	Carnivora	*Lycalopex* spp.	Not determined	16/33 (49%)	Coquimbo region	Haemagglutination inhibition test (HAI)	10
	Carnivora	Andean fox *Lycalopex culpaeus*	Not determined	1/16 (6.25%)	Los Cipreses National Reserve (O'Higgins region)	ELISA and haemagglutination inhibition test (HAI)	80
	Carnivora	South American Sea Lion *Otaria byronia*	Not determined	3/3 (100%)	Los Ríos region	Hemagglutination inhibition test (HAI)	9
Feline leukemia virus (FeLV)	Carnivora	Guigna *Leopardus guigna*	Not determined	3/15 (20%)	Chiloé Island (Los Lagos region)	PCR amplification and sequencing	85
Feline immunodeficiency virus (FIV)	Carnivora	Guigna *Leopardus guigna*	Not determined	2/15 (13.3%)	Chiloé Island (Los Lagos region)	PCR amplification and sequencing	85
Herpesvirus (Gammaherpesvirus)	Carnivora	Darwin's fox *Lycalopex fulvipes*	Not determined	4/28 (14.29%)	Chiloé Island (Los Lagos region)	PCR	114
Bovine rhinotracheitis (BoHV-1)	Cetartiodactyla	Chilean Huemul *Hippocamelus bisulcus*		0/18	Aysén region	Serological neutralization test	89
Foot-and-mouth disease virus	Cetartiodactyla	Southern Pudu *Pudu puda*	Not determined	1 negative	Biobío region	Serological neutralization test	88
Bovine viral diarrhea virus (BVDV)	Cetartiodactyla	Chilean Huemul *Hippocamelus bisulcus*	Not determined	2/18 (11.1%)	Aysén region	Serological neutralization test	89
	Cetartiodactyla	Southern Pudu *Pudu puda*		1 positive	Biobío region	Serological neutralization test, reverse-transcriptase PCR and DNA sequencing	88
Tattoo skin virus (*Poxvirus*)	Cetartiodactyla	Chilean Dolphin *Cephalorhynchus eutropia*	Not determined	4/13 (30.8%)	Chilean Northern Patagonia	Visual inspection of lesions	90
	Cetartiodactyla	Black Porpoise *Phocoena spinipinnis*	Not determined	3/3 (100%)	Punta de Choros (Coquimbo region)	Visual inspection of lesions	90
			Not determined	3/4 (75%)	Punta de Choros (Coquimbo region)	Visual inspection of lesions	91
	Cetartiodactyla	Bottle-nosed Dolphin *Tursiops truncatus*	Not determined	1/1 (100%)	Isla de Choros (Coquimbo region)	Visual inspection of lesions	90

Annexed. Continuation

Pathogen	Host Order	Host	Pathogen characterization	Prevalence/ Number of positives (percentage)	Location	Diagnostic technique	Reference
***Leptospira* spp.**	Carnivora	American mink *Neovison vison*	Not determined	31/57 (55.6%)	Los Ríos (5), Los Lagos (10) and Aysén regions (16)	PCR	28
	Rodentia	Degu *Octodon degus*	Not determined	26/260 (10%)	Metropolitana region	PCR	27
			Not determined	7/144 (4.86%)	Metropolitana region	Nested PCR	18
	Rodentia	Olive-colored akodont *Abrothrix olivaceus*	Not determined	3 positives	Los Ríos region	Renal biopsy	19
			Serovar Poi (3) Hardjo (3), Pomona (3), Copenhageni (2), Medanensis (1), Icterohaemorrhagiae (1), Icterohaemorrhagiae-Medanensis (1), Sejroe	19/41 (46.3%)	Los Ríos region	Microscopic agglutination test (MAT) and renal biopsy	20
			Not determined	8/14 (57.1%)	Los Ríos region	Indirect immunofluorescence, immunoperoxidase, dark-field microscopy and Levaditi's staining	23
			Not determined	35/83 (42.2%)	Los Ríos region	Indirect immunofluorescence and immunoperoxidase	24
			Not determined	91/206 (44.2%)	Los Ríos region	Serology, immunochemical diagnostic and microscopic agglutination test (MAT)[h]	17
			Not determined	6/53 (11.3%)	Los Ríos region	Bacterial culture, dark-field microscopy and endonuclease restriction enzyme	25
			Serovar Icterohaemorrhagiae	1/10 (10%)	Metropolitana region	Microscopic agglutination test (MAT)	26
			Not determined	21/187 (11.23%)	Metropolitana region	Nested PCR	18
	Rodentia	Long-haired akodont *Abrothrix longipilis*	Serovar Sejroe (5), Poi (2) Copenhageni (1), Medanensis (1), Hardjo (1), Copenhageni, Pomona, Icterohaemorrhagiae	13/22 (59.09%)	Los Ríos region	Microscopic agglutination test (MAT) and renal biopsy	20
			Not determined	9/16 (56.3%)	Los Ríos region	Indirect immunofluorescence, immunoperoxidase, dark-field microscopy and Levaditi's staining	23
			Not determined	62/126 (49.2%)	Los Ríos region	Indirect immunofluorescence and immunoperoxidase	24
			Not determined	87/175 (49,7%)	Los Ríos region	Serology, immunochemical diagnostic and microscopic agglutination test (MAT)[h]	17
				0/9	Los Ríos region	Bacterial culture and dark-field microscopy	25

Annexed. Continuation

Pathogen	Host Order	Host	Pathogen characterization	Prevalence/ Number of positives (percentage)	Location	Diagnostic technique	Reference
	Rodentia	Darwin's pericote *Phyllotis darwini*	Not determined	4/68 (5.9%)	Metropolitana region	PCR	27
			Not determined	3/62 (4.8%)	Metropolitana region	Nested PCR	18
	Rodentia	Long-tailed Pygmy Rice Rat *Oligoryzomys longicaudatus*	Not determined	2 positives	Los Ríos region	Renal biopsy	19
			Serovar Copenhageni (2), Poi (3), Pomona (3), Icterohaemorrhagiae-Copenhageni (1), Copenhageni-Poi (1), Sejroe, Medanensis, Hardjo	15/36 (41.7%)	Los Ríos region	Microscopic agglutination test (MAT) and renal biopsy	20
			Not determined	15/36 (41.7%)	Los Ríos region	Indirect immunofluorescence, immunoperoxidase, dark-field microscopy and Levaditi's staining	23
			Not determined	25/89 (28.1%)	Los Ríos region	Indirect immunofluorescence and immunoperoxidase	24
			Not determined	77/191 (40.3%)	Los Ríos region	Serology, immunochemical diagnostic and microscopic agglutination test (MAT)[h]	17
			Not determined	16/76 (21.1%)	Los Ríos region	Bacterial culture, dark field microscopy and endonuclease restriction enzyme	25
			Not determined	2/45 (4.44%)	Metropolitana region	Nested PCR	18
	Rodentia	*Rattus* sp.	Serovar Icterohaemorrhagiae	2 positives	Metropolitana region	Guineae pig inoculation	115
	Rodentia	Black rat *Rattus rattus*	Serovar Copenhageni (1), Medanensis (1), Sejroe, Hardjo, Pomona, Poi, Icterohaemorrhagiae	3/5 (60%)	Los Ríos region	Microscopic agglutination test (MAT) and renal biopsy	20
			Not determined	7/17 (41.8%)	Los Ríos region	Indirect immunofluorescence, immunoperoxidase, dark-field microscopy and Levaditi's staining	23
			Not determined	9/34 (26.5%)	Los Ríos region	Indirect immunofluorescence and immunoperoxidase	24
			Not determined	18/85 (21.2%)	Los Ríos region	Serology, immunochemical diagnostic and microscopic agglutination test (MAT)[h]	17
				0/15	Los Ríos region	Bacterial culture and dark-field microscopy	25
			Serovar Icterohaemorrhagiae	1/3 (33.33%)	Metropolitana region	Microscopic agglutination test (MAT)	26
			Not determined	51/246 (20.7%)	Los Ríos region	PCR	116
			Not determined	5/84 (5.95%)	Metropolitana region	Nested PCR	18

Annexed. Continuation

Pathogen	Host Order	Host	Pathogen characterization	Prevalence/ Number of positives (percentage)	Location	Diagnostic technique	Reference
	Rodentia	Brown rat *Rattus norvegicus*	Not determined	63/100 (63%)	Metropolitan region	Direct observation with ultramicroscope	14
				0/2	Los Ríos region	Indirect immunofluorescence, immunoperoxidase, dark-field microscopy and Levaditi's staining	23
				0/8	Los Ríos region	Indirect immunofluorescence and immunoperoxidase	24
			Not determined	2/27 (7.4%)	Los Ríos region	Serology, immunochemical diagnostic and microscopic agglutination test (MAT)[h]	17
			Not determined	2/14 (14.3%)	Los Ríos region	Bacterial culture, dark-field microscopy and endonuclease restriction enzyme	25
			Serovar Icterohaemorrhagiae	3/9 (33.3%)	Metropolitana region	Microscopic agglutination test (MAT)	26
			Not determined	3/29 (10.3%)	Los Ríos region	PCR	116
			Not determined	24/63 (38.1%)	Metropolitana region	Nested PCR	18
	Rodentia	House mouse *Mus musculus*		0/2	Los Ríos region	Microscopic agglutination test (MAT) and renal biopsy	20
			Not determined	1/8 (12.5%)	Los Ríos region	Indirect immunofluorescence, immunoperoxidase, dark-field microscopy and Levaditi's staining	23
			Not determined	2/26 (7.7%)	Los Ríos region	Indirect immunofluorescence and immunoperoxidase	24
			Not determined	20/97 (20.6%)	Los Ríos region	Serology, immunochemical diagnostic and microscopic agglutination test (MAT)[h]	17
			Not determined	7/31 (22.58%)	Los Ríos region	Bacterial culture, dark-field microscopy and endonuclease restriction enzyme	25
				0/13	Metropolitan region region	Microscopic agglutination test (MAT)	26
			Not determined	18/80 (22.5%)	Los Ríos region	PCR	116
			Not determined	6/47 (12.8%)	Metropolitana region	Nested PCR	18
	Rodentia	Long-clawed mole mouse *Geoxus valdivianus*		0/1	Los Ríos region	Indirect immunofluorescence and immunoperoxidase	24
			Not determined	1/2 (50%)	Los Ríos region	Serology, immunochemical diagnostic and microscopic agglutination test (MAT)[h]	17
	Rodentia	Southern big-eared mouse *Loxodontomys micropus*		0/1	Los Ríos region	Indirect immunofluorescence and immunoperoxidase	24
				0/1	Los Ríos region	Serology, immunochemical diagnostic and microscopic agglutination test (MAT)[h]	25

Annexed. Continuation

Pathogen	Host Order	Host	Pathogen characterization	Prevalence/ Number of positives (percentage)	Location	Diagnostic technique	Reference
Leptospira interrogans	Carnivora	American mink *Neovison vison*	Not determined	5 positives[i]	Aysén region	Ribosomal RNA gene sequencing	28
	Carnivora	South American Sea Lion *Otaria byronia*	Serovar Bratislava and Pomona (1), Hardjo, Icterohaemorrhagiae, Copenhageni, Canicola	1/3 (33.33%)	Los Ríos region	Immunohistochemestry (IHC)	9
	Rodentia	Degu *Octodon degus*	Serovar Bratislava (2)	26/260 (10%)	Metropolitana region	Microscopic agglutination test (MAT) and PCR	27
			Not determined	7/144 (4.9%)	Metropolitan region	Nested PCR	18
	Rodentia	Darwin's pericote *Phyllotis darwini*		0/68	Metropolitana region	Microscopic agglutination test (MAT)	27
	Rodentia	Olive-colored akodont *Abrothrix olivaceus*	Serovar Hardjo (1), Javanica (5), Icterohaemorrageae (1), Pomona	8/31 (25.8%)[j]	Los Ríos region	Microscopic agglutination test (MAT)	21
			Not determined	12/33 (36.4%)	Los Ríos region	Serology, microscopy or bacterial culture[d]	22
	Rodentia	Long-haired akodont *Abrothrix longipilis*	Serovar Pomona (7), Hardjo (2), Canicola (1), Hardjo-Pomona (2), Icterohaemorrhageae	12/53 (22.6%)	Los Ríos region	Microscopic agglutination test (MAT)	21
			Not determined	29/60 (48.3%)	Los Ríos region	Serology, microscopy or bacterial culture[d]	22
	Rodentia	Darwin's pericote *Phyllotis darwini*		0/68	Metropolitana region	Microscopic agglutination test (MAT)	27
	Rodentia	Long-tailed Pygmy Rice Rat *Oligoryzomys longicaudatus*		0/8	Los Ríos region	Microscopic agglutination test (MAT)	21
				0/9	Los Ríos region	Serology, microscopy or bacterial culture[d]	22
	Rodentia	Brown rat *Rattus norvegicus*	Serovar Icterohaemorrhageae-Javanica (1), Pomona, Hardjo, Canicola	1/4 (25%)	Los Ríos region	Microscopic agglutination test (MAT)	21
				0/4	Los Ríos region	Serology, microscopy or bacterial culture[d]	22
	Rodentia	Black rat *Rattus rattus*	Serovar Pomona (3), Hardjo (2), Javanica, Canicola, Icterohaemorrhageae	5/7 (71.4%)	Los Ríos region	Microscopic agglutination test (MAT)	21
			Not determined	4/7 (57.1%)	Los Ríos region	Serology, microscopy or bacterial culture[d]	22
	Rodentia	House mouse *Mus musculus*		0/2	Los Ríos region	Microscopic agglutination test (MAT)	21
				0/7	Los Ríos region	Serology, microscopy or bacterial culture[d]	22
	Rodentia	Long-clawed mole mouse *Geoxus valdivianus*		0/1	Los Ríos region	Microscopic agglutination test (MAT)	21
			Not determined	1/1 (100%)	Los Ríos region	Serology, microscopy or bacterial culture[d]	22

Annexed. Continuation

Pathogen	Host Order	Host	Pathogen characterization	Prevalence/ Number of positives (percentage)	Location	Diagnostic technique	Reference
Leptospira borgpetersenii	Rodentia	Degu *Octodon degus*	Serovar Ballum	2/260 (0.77%)	Metropolitana region	Microscopic agglutination test (MAT)	27
	Rodentia	Darwin's pericote *Phyllotis darwini*		0/68	Metropolitana region	Microscopic agglutination test (MAT)	27
	Carnivora	American mink Neovison vison	Not determined	4 positives[i]	Los Ríos (2), Los Lagos (1) and Aysén regions (1)	Ribosomal RNA gene sequencing	28
Leptospira kirschneri	Rodentia	Degu *Octodon degus*		0/260	Metropolitana region	Microscopic agglutination test (MAT)	27
	Rodentia	Darwin's pericote *Phyllotis darwini*		0/68	Metropolitana region	Microscopic agglutination test (MAT)	27
	Carnivora	South American Sea Lion *Otaria byronia*	Serovar Grippotyphosa	0/3	Los Ríos region	Immunohistochemestry (IHC)	9
Leptospira biflexa	Rodentia	Degu *Octodon degus*		0/260	Metropolitana region	Microscopic agglutination test (MAT)	27
	Rodentia	Darwin's pericote *Phyllotis darwini*		0/68	Metropolitana region	Microscopic agglutination test (MAT)	27
	Carnivora	South American Sea Lion *Otaria byronia*	Serovar Patoc	2/3 (66.67%)	Los Ríos region	Immunohistochemestry (IHC)	9
Leptospira santarosai	Rodentia	Degu *Octodon degus*		0/260	Metropolitana region	Microscopic agglutination test (MAT)	27
	Rodentia	Darwin's pericote *Phyllotis darwini*		0/68	Metropolitana region	Microscopic agglutination test (MAT)	27
Yersinia enterocolitica	Rodentia	Olive-colored akodont *Abrothrix olivaceus*	Not determined	2/117 (1.7%)	Los Ríos region	Bacterial culture	117
	Rodentia	Long-haired akodont *Abrothrix longipilis*	Not determined	5/32 (15.6%)	Los Ríos region	Bacterial culture	117
	Rodentia	Long-tailed Pygmy Rice Rat *Oligoryzomys longicaudatus*	Not determined	2/106 (1.9%)	Los Ríos region	Bacterial culture	117
	Rodentia	Brown rat *Rattus norvegicus*	Not determined	3/15 (20%)	Los Ríos region	Bacterial culture	117
***Mycobacterium avium* paratuberculosis**	Lagomorpha	European hare *Lepus europaeus*	Not determined	48/380 (12.6%)	Los Ríos region	Mycobacteria growth indicator tube (MGIT) and Real-time PCR	68
			Not determined	62/92 (67.4%)	Los Ríos region	Bacterial culture and Real-time PCR	69
	Cetartiodactyla	Southern Pudu *Pudu puda*	Not determined	1/1 (100%)	Biobío region	Histopathology	66
			Not determined	3/3 (100%)	Los Ríos region	Mycobacteria detection system and real-time PCR	67
	Cetartiodactyla	Guanaco *Lama guanicoe*	Not determined	21/501 (4.2%)	Magallanes region	Bacterial culture and PCR	65
	Cetartiodactyla	Red deer *Cervus elaphus*	Not determined	4/4 (100%)	Los Lagos region	Histopathology, bacterial culture and PCR	72
			Not determined	14/14 (100%)	Los Ríos and Los Lagos regions	Histopathology	73
	Cetartiodactyla	Fallow deer *(Dama dama)*	Not determined	9/9 (100%)	Los Ríos and Los Lagos regions	Histopathology	73
	Cetartiodactyla	Chilean huemul *(Hippocamelus bisulcus)*	Not determined	6/14 (42.8%)	Aysén and Magallanes regions (Bernardo O'Higgins National Park)	Mycobacteria detection system and PCR	70

Annexed. Continuation

Pathogen	Host Order	Host	Pathogen characterization	Prevalence/ Number of positives (percentage)	Location	Diagnostic technique	Reference
***Mycoplasma* sp.**	Carnivora	Darwin's fox *Lycalopex fulvipes*	Not determined	17/30 (56.67%)	Chiloé Island (Los Lagos region)	Real-time PCR and DNA sequencing	118
Mycoplasma haemocanis	Carnivora	Darwin's fox *Lycalopex fulvipes*	Not determined	8 positives[i]	Chiloé Island (Los Lagos region)	DNA sequencing	118
Mycoplasma haemofelis	Carnivora	Darwin's fox *Lycalopex fulvipes*	Not determined	1 positive[i]	Chiloé Island (Los Lagos region)	DNA sequencing	118
Coxiella burnetti	Carnivora	Darwin's fox *Lycalopex fulvipes*		0/30	Chiloé Island (Los Lagos region)	RReal-time PCR	118
***Borrelia* sp.**	Carnivora	Darwin's fox *Lycalopex fulvipes*		0/30	Chiloé Island (Los Lagos region)	RReal-time PCR	118
	Cetartiodactyla	Southern Pudu *Pudu puda*		0/2	Los Ríos region	PPCR and DNA sequencing	103
***Bartonella* sp.**	Carnivora	Darwin's fox *Lycalopex fulvipes*		0/30	Chiloé Island (Los Lagos region)	RReal-time PCR	118
***Rickettsia* sp.**	Carnivora	Darwin's fox *Lycalopex fulvipes*	Not determined	1/30 (3.3%)	Chiloé Island (Los Lagos region)	Real-time PCR and DNA sequencing	118
Candidatus Neoehrlichia chilensis	Rodentia	*Abrothrix* sp.		4/5 (80%)	Los Ríos region	cPCR and DNA sequencing	100
	Rodentia	House Mouse *Mus musculus*		1/5 (20%)	Los Ríos region	cPCR and DNA sequencing	100
Salmonella enterica	Carnivora	South American Sea Lion *Otaria byronia*	Serovar Havana (1), Newport (1)	2/13 (15.38%)	Antofagasta region	Bacterial culture and invA gene detection by PCR	119
***Brucella* sp.**	Carnivora	South American Sea Lion *Otaria byronia*		0/3	Los Ríos region	Plaque agglutination	9
Brucella canis	Carnivora	South American Sea Lion *Otaria byronia*		0/3	Los Ríos region	Plaque agglutination	9
Brucella abortus	Carnivora	South American Sea Lion *Otaria byronia*		0/3	Los Ríos region	Bengal rose	9
	Cetartiodactyla	Chilean Huemul *Hippocamelus bisulcus*		0/18	Aysén region	Rose Bengal test	89

Annexed. Continuation

Pathogen	Host Order	Host	Pathogen characterization	Prevalence/ Number of positives (percentage)	Location	Diagnostic technique	Reference
Escherichia coli	Carnivora	South American Sea Lion *Otaria byronia*	DAEC (Diffusely-adherent *Escherichia coli*) (1), EPEC (Entero-pathogenic *E. coli*)	1/15[k] (6.7%)	Tarapacá region	Bacterial culture and PCR	120
Campylobacter insulaenigrae	Carnivora	South American Sea Lion *Otaria byronia*	Strain OFI	1/5[k] (6.7%)	Los Ríos region	Bacterial culture and amplified fragment length polymorphism analysis	121
Edwarsiella tarda	Carnivora	South American Sea Lion *Otaria byronia*	Not determined	22/30[l] (73.3%)	Los Ríos region	Bacterial culture	122
Klebsiella pneumoniae	Rodentia	Long-tailed chinchilla *Chinchilla lanigera*	Not determined	13/53 (24.5%)	Coquimbo region	Bacterial culture	123
	Carnivora	South American Sea Lion *Otaria byronia*	Not determined	1/30 (3.3%)	Los Ríos region	Bacterial culture	137
Proteus mirabilis	Carnivora	South American Sea Lion *Otaria byronia*	Not determined	2/30 (6.7%)	Los Ríos region	Bacterial culture	137
	Rodentia	Long-tailed chinchilla *Chinchilla lanigera*	Not determined	1/53 (1.9%)	Coquimbo region	Bacterial culture	138
Corynebacterium pseudo-tuberculosis	Cetartiodactyla	Chilean Huemul *Hippocamelus bisulcus*	Ovine genotype	2/2 (100%)	Aysén region	Bacterial culture and PCR	107
Morganella morganii	Carnivora	South American Sea Lion *Otaria byronia*	Not determined	2/30 (6.7%)	Los Ríos region	Bacterial culture	137
	Rodentia	Long-tailed chinchilla *Chinchilla lanigera*	Not determined	1/53 (1.9%)	Coquimbo region	Bacterial culture	138
Staphylococcus aureus	Rodentia	Long-tailed chinchilla *Chinchilla lanigera*	Not determined	4/53 (7.5%)	Coquimbo region	Bacterial culture	138
Pseudomona auriginosa	Rodentia	Long-tailed chinchilla *Chinchilla lanigera*	Not determined	2/53 (3.8%)	Coquimbo region	Bacterial culture	138

[a]Regions were not specified for each positive sample. [b]9 seropositive samples considering a titer cut-off of 1:8 and 5 positives with a titer cut-off of 1:16. [c]Only the prevalence is indicated but there is no information about number of positive cases nor total number of individuals studied. [d]The specific serological method for analysis was not indicated in the study. [e]The study specified the collection site for only 14 positive samples. [f]Only the number of captured individuals is specified not the number of analyzed samples. [g]The total number of individuals examined in the study was not specified in the methodology. [h]The specific serological and immunohistochemical method for analysis were not indicated in the study. [i]DNA/RNA sequencing was performed in samples confirmed in the same study. [j]The detail of number of positive samples presented in this study does not match with the total number of positive samples. [k]The study does not indicate if each fecal sample collected was from a unique individual or samples were taken more than one time from the same individual. [l]The study does not indicate if each sample was collected from a unique individual.

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