Serosurvey of Pathogens in Domestic Dogs on the Border of Noël Kempff Mercado National Park, Bolivia

Authors: Bronson, Ellen, Emmons, Louise H., Murray, Suzan, Dubovi, Edward J., and Deem, Sharon L.

SEROSURVEY OF PATHOGENS IN DOMESTIC DOGS ON THE BORDER OF NOËL KEMPFF MERCADO NATIONAL PARK, BOLIVIA

Ellen Bronson, Med. Vet., Louise H. Emmons, Ph.D., Suzan Murray, D.V.M., Dipl. A.C.Z.M., Edward J. Dubovi, Ph.D., and Sharon L. Deem, D.V.M., Ph.D., Dipl. A.C.Z.M.

Abstract: The threat of disease transmission from domestic animals to wildlife has become recognized as an increasing concern within the wildlife community in recent years. Domestic dogs pose a significant risk as reservoirs for infectious diseases, especially for wild canids. As part of a multifaceted ecologic study of maned wolves and other canids in the large, remote Noël Kempff Mercado National Park (NKMNP) in northeastern Bolivia, 40 domestic dogs in two villages and at two smaller settlements bordering the national park were sampled for exposure to canine diseases. High levels of exposure were found to canine distemper virus and canine parvovirus, both of which are known to cause mortality in maned wolves and other carnivores. Moderate to high levels of exposure were found to rabies virus, *Ehrlichia canis*, and *Toxoplasma gondii*, as well as significant levels of infection with *Dirofilaria immitis*. This study reports evidence of exposure to several diseases in the domestic dogs bordering the park. Contact between wild carnivores and dogs has been documented in the sampled villages, therefore dogs likely pose a substantial risk to the carnivores within and near NKMNP. Further measures should be undertaken to decrease the risk of spillover infection from domestic animals into the wild species of this region.

Key words: Bolivia, domestic dog, domestic-wildlife interface, maned wolf, Noël Kempff Mercado National Park, seroprevalence.

INTRODUCTION

The threat of disease transmission from domestic animals to wildlife has become recognized as an increasing concern among wildlife conservationists.[12,32] Domestic dogs pose a significant risk as reservoirs for infectious diseases, especially to wild canids, because of their abundance and ability to travel long distances into the habitat of wild carnivores.[19,20] Domestic dogs have been implicated in epidemics due to spillover infections in African lions (*Panthera leo*),[10] Ethiopian wolves (*Canis simensis*),[27] and African wild dogs (*Lycaon pictus*).[1,21] Knowledge of the disease prevalence in the domestic dog, as well as wild carnivore populations, is essential for conservation management of ecosystems where these species coexist.[32]

Noël Kempff Mercado National Park (NKMNP) is a 1.5 million ha park in the northeastern corner

From Department of Animal Health, Smithsonian's National Zoological Park, 3001 Connecticut Avenue, NW, Washington, DC 20008, USA (Bronson, Murray, Deem); Department of Zoology, NHB 390, MRC 108, Smithsonian Institution, P.O. Box 37012, Washington, D.C. 20013-7012, USA (Emmons); and the Department of Population Medicine and Diagnostic Science, College of Veterinary Medicine, Cornell University, Ithaca, NY 14853, USA (Dubovi). Present address (Bronson): Maryland Zoo in Baltimore, Druid Hill Park, Baltimore, Maryland 21217, USA. Correspondence should be directed to Dr. Bronson (ellen.bronson@marylandzoo.org).

of Santa Cruz Department, Bolivia. The park is diverse in habitat including high ground semi-deciduous forests, seasonally flooded and riverside humid forests, savanna wetlands, and dry upland savannas. Because of its large area and pristine state, NKMNP is a World Heritage site, which is a designation by the United Nations of a place of significant cultural or natural importance to humanity. In comparison with other ecosystems in South America that are being destroyed at an accelerated rate, the majority of this isolated park remains untouched by human activities. It hosts one of the largest protected maned wolf (*Chrysocyon brachyurus*) populations, estimated at 118 breeding pairs fragmented among six isolated savannas within 3,600 km^2 of suitable habitat in the park, or one quarter of the park's area.[38] Numerous other carnivore species inhabit the park, including crab-eating foxes (*Cerdocyon thous*), pampas foxes (*Lycalopex gymnocercus*), bush dogs (*Speothos venaticus*), jaguars (*Panthera onca*), pumas (*Puma concolor*), ocelots (*Leopardus pardalis*), jaguarundis (*Puma yagouaroundi*), margays (*Leopardus wiedii*), pampas cats (*Leopardus braccatus*), and Geoffroy's cats (*Leopardus geoffroyi*). All of these canid and felid species are listed in Appendix I or II of the Convention on International Trade in Endangered Species (CITES).[35,37] There is minimal human presence within the park and the borders are well enforced, but multiple large estancias, or ranches, and villages border the park to the west in Bolivia. The east-
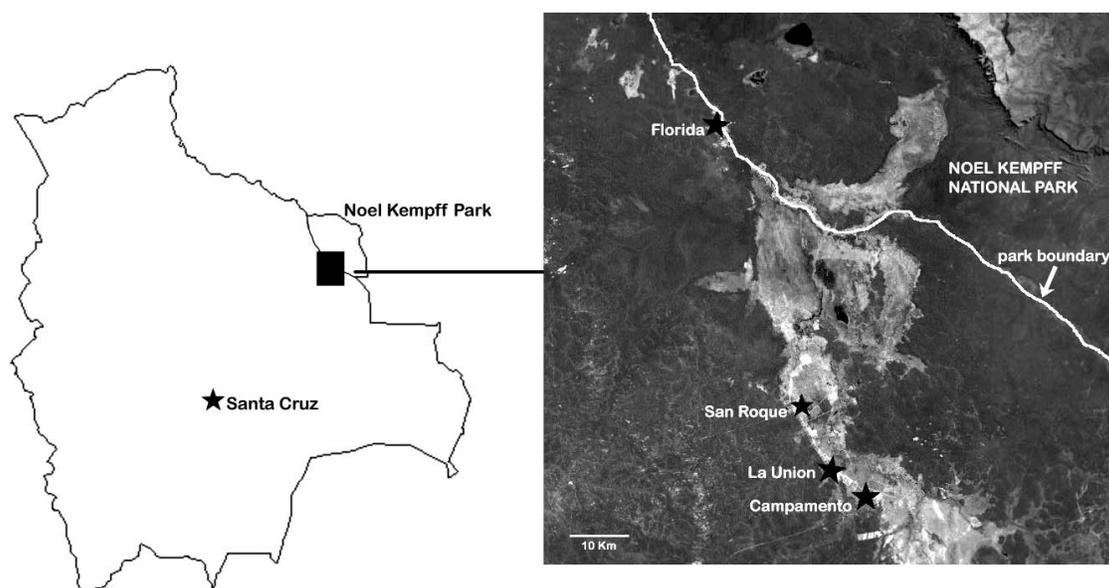
Figure 1. Map of Noël Kempff Mercado National Park in northeast Bolivia. Inset shows study area near the southwestern park boundary and stars indicate areas sampled in the study. Bar = 10 km.

ern border of the park is separated from densely populated areas in the Brazilian states of Rondônia and Mato Grosso by the Río Iténez.

A health survey of the maned wolves in NKMNP was initiated in 2000, and initial results revealed exposure to multiple canine pathogens, most notably canine distemper virus (CDV) and canine parvovirus (CPV).[14] To more accurately evaluate the risk that domestic dogs represent to the health of the maned wolves, the study was broadened to include a serosurvey of domestic dogs that inhabit villages and ranches bordering the park. These dogs are free roaming, in fair health, and largely unvaccinated. Direct contact, as well as indirect contact, with feces, urine, and other body fluids, likely exists between dogs and crab-eating foxes and potentially exists with other carnivores such as the maned wolf. Crab-eating foxes are common in the park and surrounding areas and share territories with maned wolves. The foxes are frequently seen within the villages (Emmons, Deem, and Bronson, unpubl. data) and could therefore be a link in the disease ecology of the carnivores in the NKMNP. Similar links have been proven or suspected between carnivores in other ecosystems.[2,3,10]

The objective of this study was to determine the prevalence of exposure to select infectious and parasitic agents of the domestic dogs surrounding the park in order to evaluate the disease risk they may pose for the maned wolves and other carnivores residing within NKMNP. This information will be used to make recommendations to the administration of the park to better protect these endangered species.

MATERIALS AND METHODS

All procedures performed for this study were approved by and in accordance with the Institutional Animal Care and Use Committee of the National Zoological Park's Conservation and Research Center (Front Royal, Virginia, USA).

Forty serum samples from domestic dogs at four sites were collected during two field seasons as part of a larger study of the ecology of maned wolves of NKMNP, in northeastern Bolivia (Fig. 1). In October 2004, seventeen dogs from a total estimated population of 30 adult dogs were sampled in the village of Florida, an isolated town of approximately 120 inhabitants on the Río Paraguá, on one of the few roads leading into the park. The inhabitants of this village make a living mostly from subsistence farming and hunting, but a small tourist industry is growing, and many of the village men are NKMNP park guides. In October 2005, eighteen dogs in the larger village of Campamento (estimated 500 residents and 60–100 adult dogs) were sampled. There are no veterinarians within 200 km of either of these villages. Dogs at two estancias between the park and the village of Campamento were also sampled in October 2005. All adult dogs present at San Roque (*n* = 3) and two of four adult dogs at La Union were included in the study. Each

estancia maintains several hundred head of cattle on savanna habitat.

A village resident accompanied us to each of the locations, and permission was obtained from each municipality before sampling. The authors went from house to house to discuss the project and obtain consent from each owner. A brief survey was conducted with owners to determine the age, vaccination history, and health history of the dogs sampled. When the owners could be identified, they were also questioned about the frequency their dogs entered the forest and how often they saw large carnivores near the villages. These interviews were conducted most intensely in the second year of sampling, which included the two estancias and the village of Campamento, and to a lesser degree, Florida.

Dogs at all sites were manually restrained and muzzles applied before sampling. Blood was drawn from the jugular or saphenous vein into serum separator tubes (Corvac Sherwood Medical, St. Louis, Missouri 63103, USA) and cooled on ice for 1–4 hr until return to the research base. Only those dogs known to be >4 mo old were included in order to decrease the likelihood of interference by maternal antibodies.[22] Ticks were collected from the skin of infested dogs and preserved in 70% ethanol. After sampling, and with the permission of each owner, all dogs present at each house, regardless of age, were vaccinated with a killed rabies vaccine (Imrab 3, Merial, Athens, Georgia 30601, USA) and a modified live combination vaccine against CDV, CPV, canine adenovirus Type 2 (CAV), and canine parainfluenza virus (Duramune Max 5, Fort Dodge Animal Health, Overland Park, Kansas 66225, USA). All animals were dewormed with pyrantel pamoate (Strongid T, Pfizer Animal Health, Exton, Pennsylvania 19341, USA). One dose of injectable ivermectin (Ivomec, Merial, Athens, Georgia 30601, USA) was given to dogs <6 mo old to avoid complications with possible *Dirofilaria immitis* infestations in older animals.

On return to the research base, the sera were separated by centrifugation (Mobilespin, Vulcan Technologies, Grandview, Missouri 64040, USA) at 3,000 *g* for 15–20 min and stored in liquid nitrogen (5DS, Taylor-Wharton Cryogenics, Theodore, Alabama 36590, USA) or a battery-powered freezer. Samples were transported to the United States 3–5 days later and stored at −70°C until analyzed. Serologic testing for antibodies to CAV, canine coronavirus (CCV), CDV, canine herpesvirus (CHV), CPV, *Toxoplasma gondii*, *Brucella canis*, and *Leptospira interrogans* (serovars *L. pomona*, *L. hardjo*, *L. grippotyphosa*, *L. icterohaemorrhagiae*, and *L.*

canicola), as well as for antigen of *D. immitis* were performed at the New York State Veterinary Diagnostic Laboratory as previously described.[14,19] Serologic testing for rabies virus antibodies was performed at Kansas State Veterinary Diagnostic Laboratory using the rapid fluorescent focus inhibition test. The dogs from Campamento, La Union, and San Roque were also tested for antibodies to *Ehrlichia canis*, *Borrelia burgdorferi*, and *Rickettsia rickettsii* at the Texas Veterinary Medical Diagnostic Laboratory using the immunofluorescence assay (IFA).

RESULTS

The average age of the dogs in Florida was 1.93 yr ± standard deviation (SD) 1.55 yr with a range of 6 mo to 5 yr. The average age of the dogs in Campamento was 3.02 yr ± SD 2.16 yr with a range of 4 mo to 7 yr.

Based on informal interviews with owners, a portion of the dogs sampled had been previously vaccinated against rabies; none of the dogs at the four sites had been previously vaccinated against diseases other than rabies. In the more isolated village of Florida, only the two oldest animals had been vaccinated by a state-mandated rabies program 4 yr previously. All of the dogs at each of the estancias and 10 of the 18 dogs in the village of Campamento had been previously vaccinated against rabies.

Tables 1 and 2 summarize each disease agent tested, methods used, positive cutoff values, and test results for each site. Over 85% of all dogs tested were seropositive for CDV and CPV, as well as for *E. canis*. The majority of the dogs positive for *E. canis* were also positive for *R. rickettsii* and negative for *B. burgdorferi*. Antibodies for CHV were found in 70% of the dogs tested, and CAV and CCV antibodies were found in less than one fifth and one tenth of the dogs, respectively. Positive titers to *T. gondii* were found in 80% of dogs tested. Positive rabies titers were found in all of the dogs in the village of Florida, only two of which had been previously vaccinated. Five dogs at San Roque, La Union, and Campamento had positive rabies titers; all of these animals had been reported by the owners to have been vaccinated against rabies. Antigen of *D. immitis*, the filarid parasite that causes canine heartworm disease, was found in 41% of the dogs in Florida and 22% of the dogs in Campamento. Positive titers to *B. canis* were only seen in the town of Campamento and in less than one quarter of these dogs. Positive titers to *L. interrogans* were identified in eight of the 40 dogs tested, with mostly low titers to the serovars *L.*

Table 1. Number of domestic dogs serologically positive for each viral pathogen per total number of dogs at each site sampled near Noël Kempff Mercado National Park, Bolivia.

	Canine adenovirus	Canine coronavirus	Canine distemper virus	Canine herpesvirus	Canine parvovirus	Rabies virus
Test method[a]	SN	SN	SN	SN	HAI	RFFIT
Positive titer	1:4	1:8	1:8	1:8	1:10	1:5
Site *n* (%)						
Florida	2/17 (12)	2/17 (12)	15/17 (88)	11/17 (65)	16/17 (94)	17/17 (100)
San Roque	0/3 (0)	0/3 (0)	3/3 (100)	1/3 (33)	2/3 (67)	0/2 (0)
La Union	0/2 (0)	0/2 (0)	2/2 (100)	2/2 (100)	2/2 (100)	2/2 (100)
Campamento	5/18 (28)	1/18 (6)	17/18 (94)	14/18 (78)	14/18 (78)	3/18 (17)
Total	7/40 (18)	3/40 (8)	37/40 (93)	28/40 (70)	34/40 (85)	22/39 (56)

[a] SN, serum neutralization; HAI, hemagglutination inhibition; RFFIT, rapid fluorescent focus inhibition test.

hardjo, *L. canicola*, *L. grippotyphosa*, and *L. icterohaemorrhagiae*.

DISCUSSION

In this survey of free-roaming domestic dogs on the border of the NKMNP in northeastern Bolivia, the seroprevalence (e.g., antibodies) to multiple canine pathogens, was examined. The findings documented exposure, but not necessarily current infection to these agents at the time of testing. Paired serum titers several weeks apart, or further diagnostic procedures (e.g., molecular testing, virus isolation) are generally required in order to accurately diagnose active infection, which is impractical in a remote field survey.

The results of this study suggest that domestic dogs living near NKMNP have been exposed to several canine pathogens. Of greatest concern are the high levels of exposure to CDV and CPV in the dogs from all of the sampled sites. The majority of dogs at each site had positive titers to distemper

and parvovirus; in fact in the entire study only three and six dogs, respectively, were seronegative for CDV and CPV. Villagers in Campamento described canine epidemics in the past year characterized by swollen eyes, noses, and mouths with serous or hemorrhagic discharge, all signs of distemper virus infection, as well as multiple young dogs dying of hemorrhagic diarrhea, suggestive of parvovirus infection.

All dogs negative for CDV were 1 yr or younger, and the majority of the highest titers (>1:1,024) were seen in animals 1–2 yr of age. These high titers likely indicate recent infection. The dogs in Florida had low CDV titers (1:8 to 1:24) with the exception of one 1-yr-old dog with a high titer of 1:768. The dogs in the other areas had much higher titers, with 10/23 dogs with a titer >1:256. This finding implies that a more recent epidemic likely occurred in dogs in the Campamento area, and less recently in the Florida dogs, although most dogs, even those <1 yr, had been exposed.

Table 2. Number of domestic dogs serologically positive for each parasitic, bacterial, or rickettsial pathogen per total number of dogs at each site sampled near Noël Kempff Mercado National Park, Bolivia.

	Dirofilaria immitis	*Toxoplasma gondii*	*Brucella canis*	*Leptospira serovars*	*Ehrlichia canis*	*Borrelia burgdorferi*	*Rickettsia rickettsii*
Test method[a]	Occult HW	IHA	Slide Aggl./ AGID II	Microaggl.	IFA	IFA	IFA
Positive titer	—	1:64	—	1:100	—	—	—
Site *n* (%)							
Florida	7/17 (41)	13/17 (76)	0/17 (0)	1/17 (6)	—	—	—
San Roque	1/3 (33)	2/3 (67)	0/3 (0)	3/3 (100)	2/2 (100)	0/2 (0)	2/2 (100)
La Union	1/2 (50)	2/2 (100)	0/2 (0)	0/2 (0)	2/2 (100)	0/2 (0)	2/2 (100)
Campamento	4/18 (22)	15/18 (83)	4/18 (22)	4/18 (22)	15/18 (83)	0/18 (0)	15/18 (83)
Total	13/40 (33)	32/40 (80)	4/40 (10)	8/40 (20)	19/22 (86)	0/22 (0)	19/22 (86)

[a] Occult HW, occult heartworm; IHA, indirect hemagglutination; Slide Aggl./AGID II, slide agglutination/agar gel immunodiffusion test II; Microaggl., microagglutination; IFA, immunofluorescence assay.

Canine distemper virus causes morbidity and mortality in numerous carnivore species.[15] The disease has caused serious population declines in several species, and spillover infection stemming from domestic dogs has been proven or suspected in African lions,[10] African wild dogs,[1] and giant pandas.[29] Maned wolves are susceptible to CDV, and mortalities have been reported in captivity.[28] CDV is most commonly transmitted via aerosol or direct contact,[24] so close contact between dogs and maned wolves would be required to cause infection, which has not been documented to date in this park as it has in other maned wolf habitats. A further possibility would be transmission of CDV from the infected dogs to the more gregarious and common crab-eating fox, a species known (according to residents) to enter the sampled villages while simultaneously sharing territories with the maned wolves of NKMNP. The crab-eating fox could be a potential link in the spread of CDV from dogs to other carnivores, including the maned wolf. Similar links have been suspected or proven in carnivores in CDV outbreaks in Africa.[2,3,10]

The high prevalence of elevated titers for CPV was found in the dogs at all sites. All of the negative animals were <12 mo old and among the youngest dogs sampled. Parvovirus is transmitted via feces and persists for several months in the environment,[7] so transmission of CPV from domestic dogs roaming or hunting in or near the park to wild canids is a significant concern. Maned wolves in captivity are highly susceptible to this virus, which causes mortality especially in pups.[28,30] Parvovirus is also reported to be pathogenic in crab-eating foxes.[30]

A serosurvey of dogs near Madidi National Park in northwestern Bolivia found a similarly high seroprevalence of CDV and CPV in dogs.[19] In contrast, a survey of crab-eating foxes in contact with village dogs in the Amazon basin of Brazil revealed no foxes with positive titers to these two viruses, likely due to low prevalence of the disease in the domestic dogs.[11] The difference in these surveys may reflect regional differences in the endemic diseases of domestic dogs or sampling at different stages of epidemic cycles.

All of the dogs in the village of Florida had moderate to high rabies titers, although only the two oldest animals in the town had been previously vaccinated. Among dogs at the other sites, positive titers were found only in dogs that had been reported to have been vaccinated, and these titers were low. Many of the dogs reported to have been vaccinated did not have titers. The low or absent titers in vaccinated dogs may have resulted from waning titers due to insufficient vaccination frequency, improper vaccine storage prior to use, inadequate immune response owing to poor health, or inaccurate information concerning vaccination status given by owners. Rabies virus is endemic in domestic dog populations across much of South America. The disease is frequent in free-roaming dogs in the large city of Santa Cruz de la Sierra, Bolivia, located 400 km from NKMNP, where it is most prevalent in 1- to 2-yr-old animals and occurs in 5–6 yr cycles.[40] Data are scarce on the prevalence of rabies in dogs in rural, isolated regions such as the villages of this survey. Positive rabies titers in the dogs of Florida may indicate past infection with survival, which is rare but has been reported in dogs.[16,18] Cross-reaction or a nonspecific reaction is also possible, although such reactions are seldom observed with titers >1:25,[13] which were seen in seven of the 17 dogs in Florida. None of the dogs showed signs consistent with rabies at the time of exam, but current infection and sampling during the incubation period of this viral disease cannot be excluded. Of interest, two of the eight maned wolves sampled by the authors in the NKMNP to date also had low-positive titers to rabies virus,[9,14] so a dog-strain of rabies stemming from this or other surrounding villages cannot be ruled out. Rabies epidemics originating from domestic dog strains are known to have caused population declines in Ethiopian wolves[39] and African wild dogs.[21] There is also a report of an oncilla (Leopardus tigrinus) from Bolivia, with a high-positive rabies titer, which is thought to have survived previous exposure to rabies.[13]

Exposure to other canine viruses in the dog populations surrounding NKMNP includes a 70% seroprevalence of canine herpesvirus. Close contact is necessary for transmission of this virus, and it does not persist in the environment, so the risk to other canids in the region is likely limited. Low prevalence was found for CAV (18%) and CCV (8%) in the dogs tested, although very high titers to CAV have been observed in every sample taken from the maned wolves of NKMNP,[9,14] including an increasing titer in a female that has been sampled yearly from 2003–2005. CAV is transmitted via contact with infected body fluids and is stable for days to months in the environment. Infected dogs can excrete the virus in the urine for at least 6–9 mo.[23] CAV has been reported to cause pup mortalities in captive maned wolves.[6] Based on the lower seroprevalence of the dogs compared with maned wolves sampled for CAV, two possible explanations are 1) that the maned wolves are not currently being exposed from domestic dogs, and the virus (or an antigenically related virus) is endemic in the

maned wolf population in NKMNP, or 2) that dogs or other canids other than those sampled are transmitting the virus. Additional sampling of dogs and wild canids in the park is needed to understand the epidemiology of this disease in NKMNP.

Evidence of previous infection was found at low prevalence for the bacterial organisms *B. canis* and *L. interrogans*. Leptospirosis is considered to be a re-emerging disease in humans in parts of South America.[8] Results similar to those in this study, with few seropositive dogs, were found in a survey in northwestern Bolivia.[19] Eight of the 40 dogs in this present study had positive titers to the *Leptospira* serovars tested, all of them low, except for one dog at the estancia San Roque, which had high titers to two serovars, *L. canicola* and *L. icterohaemorrhagiae*, likely indicating recent infection. This study tested for five of the most common serovars in dogs; however, many other serovars exist and may be of potential importance but would not be reflected in the results.

In the 2005 season, exposure to the tick-borne rickettsial agent *E. canis* was examined. Many urban domestic dogs in South America are infected and become symptomatic with this pathogen, which causes canine monocytic ehrlichiosis. In a survey of mostly urban dogs in Brazil, 19.8% were seropositive for *E. canis*.[26] Few studies of *E. canis* seroprevalence in rural, free-roaming dogs have been reported from South America. *E. canis* is transmitted primarily by the brown dog tick, *Rhipicephalus sanguineus*, although *Amblyomma* and *Dermacentor* tick spp. are also known to be involved in transmission.[5] Two of the dogs tested in this study had two engorged adult *R. sanguineus* ticks on exam. Most of the dogs sampled (86%) had positive titers, many of them high, to this agent. However, infection with closely related rickettsial pathogens can cross-react with the immunofluorescence assay (IFA) test,[33] many of which have not been characterized for tropical regions. All dogs tested negative for exposure to *B. burgdorferi*, the causative agent of Lyme disease, which is common in the Northern hemisphere but has been rarely identified serologically and not yet isolated from South America.[26,36] The majority of dogs that tested positive to *E. canis* also had positive titers to *R. rickettsii* (84%), which causes Brazilian spotted fever in South America and is primarily transmitted by the tick *Amblyomma cajennense*. Horses, tapirs (*Tapirus terrestris*), and capybara (*Hydrochoerus hydrochaeris*) are the main hosts of this tick; however, when tick loads are high, *A. cajennense* will readily feed on dogs, wild carnivores, and less often, humans.[25] No ticks of this species were found on dogs in this study,

but the dogs have contact with horses and likely other mammals that are known to harbor this tick. Of interest but not surprising, *A. cajennense* were identified among ticks collected from three of the 11 maned wolves examined by the authors (Bronson, Deem, Emmons, unpubl. data). A subset of the maned wolves have been tested for the same rickettsial agents and show similar results but lower titers than the dogs. Results suggest that many of the dogs and maned wolves are concurrently infected with *E. canis* and *R. rickettsii*, although cross-reactions with closely related apathogenic agents within each of these genera could cause false-positive reactions. Future testing may elucidate the significance of potential infection with rickettsial organisms and whether dogs are transmitting this agent to the maned wolves of NKMNP.

Among the parasitic agents tested, the antigen of *D. immitis* was found in several of the dogs. Seven of the 17 dogs in Florida were positive, indicating current infection, and six of these dogs had high-positive results. Six of the 23 dogs at the other sites had antigen to *D. immitis*. Half of the maned wolves tested to date have been antigen-positive for heartworm, two of them with high titers,[9,14] raising concerns of possible dog–mosquito–wild carnivore cycles or endemic disease among the maned wolves that may have originated from domestic or wild canids. In wild canids, parasite loads are generally low and disease is usually subclinical, although one survey in Illinois, USA, found lower reproductive success in a small percentage of infected coyotes.[34] The role domestic dogs play in canine heartworm disease in wild carnivores is unknown, but it is assumed that transmission from domestic dogs via mosquitoes can occur.[4]

A high prevalence of toxoplasmosis was found in the dogs of this study, similar to findings in a dog survey in northwestern Bolivia, where many dogs had titers suggestive of current infection.[19] There are few domestic cats in these communities, so dogs are likely being exposed by ingestion of infected small mammals or undercooked meat given to them by their owners. Toxoplasmosis in dogs is typically subclinical, although neurologic or generalized disease may occur.[17] The significance of this finding for the wild carnivores in the park is unknown.

The results of this health survey provide evidence of exposure to multiple pathogens among the domestic dogs around the NKMNP. Based on observations by the authors, it is known that crab-eating foxes enter villages and also share territories with maned wolves, including water holes and walking paths, as evidenced by scats, prints, camera

trap images, and direct observations. Although the populations of maned wolves within the park are geographically distant from most of the surrounding villages, there are continuous populations of foxes as well as maned wolves that live between the estancias and the border of NKMNP. Feral or uncontrolled and unvaccinated domestic dogs entering the park may come into contact with the wild carnivores and transmit pathogens that cause disease. Owners in this survey reported that many of their dogs hunt with or without their owners outside of the villages several times a month. In the more detailed survey performed in the second year of the study in the larger village of Campamento, dog owners reported that 10/18 (55%) adult dogs sampled hunted one to four times per month. Half of the owners questioned (6/12) in this region reported seeing foxes regularly in and around the village, and wild felids in the wooded areas. None of the villagers reported seeing maned wolves near the settlements. This limited information supports the theory that dogs enter the protected areas either to hunt or while roaming free around the villages. Feces and urine are likely deposited in the vicinity of wild carnivores and are a possible means of transmission for infectious diseases. Dogs may come in direct contact with wild carnivores while hunting in the park or when foxes enter the villages. Although the demographics of exposure were beyond the scope of this study, indirect exposure to the wild carnivores in the park, including the maned wolves, is likely. Direct exposure between dogs and crab-eating foxes is occurring and is possible between dogs and other carnivores such as the maned wolves of NKMNP.

Preliminary results of ongoing studies of the maned wolves and sympatric canids in NKMNP indicate that domestic dogs and maned wolves share exposure to multiple canine diseases, most notably CDV, CPV, rabies, *D. immitis*, *E. canis*, and *R. rickettsii*, and transmission from dogs to maned wolves may be occurring directly or via foxes or other sylvatic hosts.[14] During sampling, all village dogs encountered were vaccinated by the authors against rabies virus, distemper, parvovirus, canine adenovirus, and parainfluenza virus. There was a close working relationship with members of the villages, and cooperation of the residents was outstanding. The villagers had great interest in this study and in receiving vaccinations for their dogs. Occasional rabies vaccination campaigns are carried out in Bolivia by the government, but due to lack of funding and remoteness of the villages bordering this isolated national park, these pivotal sites are often overlooked. Lessons learned from national parks in Africa and North America, where domestic dogs have been proven or suspected of transmitting disease to wild carnivores and causing significant population declines,[1,10,21,27,31] should motivate conservation managers to prevent such epidemics from crossing the domestic–wild animal interface in this region. Enforcing park borders and keeping domestic dogs from entering national parks such as NKMNP are paramount as are ongoing vaccination campaigns to reduce the prevalence of disease among the domestic dogs living in communities that border protected areas. Several studies of disease exposure in domestic dogs have been carried out in Africa,[1,10,27] but this represents one of few published from South America,[11,19,20] despite the high number of endangered carnivores on this continent. The value of incorporating such surveys into broad-based ecologic studies is fundamental to learning more about the disease risks faced by wild species, in order to optimize their protection in the future.

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