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Gray fox (*Urocyon cinereoargenteus*) parasite diversity in central Mexico

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ABSTRACT

Mexico has a long history of parasitological studies in communities of vertebrates. However, the mega diversity of the country makes fauna inventories an ongoing priority. Presently, there is little published on the parasite fauna of gray foxes (*Urocyon cinereoargenteus* Schereber, 1775) and this study provides new records of parasites for gray foxes in central Mexico. It is a continuation of a series of previous parasitological studies conducted with this carnivore in Mexico from 2003 to the present. A total of 24 foxes in the Parque Nacional El Cimatario (PANEC) were trapped, anaesthetized, and parasites recovered. The species found were *Dirofilaria immitis*, *Ctenocephalides canis, C. felis, Euhoplopsillus glacialis affinis* (first report for gray foxes in Mexico) *Pulex simulants,* and *Ixodes* sp. Three additional gray fox carcasses were necropsied and the parasites collected were adult nematodes *Physaloptera praeputialis* and *Toxocara canis*. The intensive study of the gray fox population selected for the 2013–2015 recent period allowed for a two-fold increase in the number of parasite species recorded for this carnivore since 2003 (nine to 18 parasite species), mainly recording parasitic arthropods, *Dirofilaria immitis* filariae and adult nematodes. The parasite species recorded are generalists that can survive in anthropic environments; which is characteristic of the present ecological scenario in central Mexico. The close proximity of the PANEC to the city of Santiago de Queretaro suggests possible parasite transmission between the foxes and domestic and feral dogs. Furthermore, packs of feral dogs in the PANEC might have altered habitat use by foxes, with possible impacts on transmission.

1. Introduction

The study of the parasite fauna in terrestrial vertebrates of Mexico has a history of over 80 years, but it is estimated that the parasites of only 21% of the total number of vertebrate species in the country have been studied (1145 of 5488 recorded species). Of these, parasites of mammals are one of the least-studied groups, with 121 host species studied of 535 recorded (Pérez-Ponce de León, 2001; Pérez-Ponce de León and García-Prieto, 2001; Pérez-Ponce de León et al., 2011). This situation is a reflection of the general lack of information of the global parasite fauna of terrestrial vertebrates, in part as a consequence of the biological properties of each host taxon (e.g., abundance, distribution, location). Additionally, legal issues related to conservation (Poulin and Morand, 2000; Pérez-Ponce de León and García-Prieto, 2001, Sarukhán et al., 2009) are sometimes an issue, such as is the safety of field researchers in areas where there is criminal activity.

The generation of new basic information on parasite species richness, including new species, in Mexico is vital for biodiversity information but also for the application of such information in the

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development of new methodological tools within the conceptual framework of ecosystem health (Karr, 1996; Rapport et al., 1998a, 1998b; Lafferty, 2012). This paper is an analysis of previous and new data obtained during twelve years of study of a gray fox (*Urocyon cinereoargenteus* Schreber, 1775) population in a protected natural area located to the South of the city of Santiago de Querétaro, in central Mexico. This protected natural area is a remaining fragment of tropical deciduous forest, surrounded by an anthropic environment. There are few studies focused on the parasites of the gray fox in México (Pineda-López, 1984; Hernández-Camacho et al., 2010, 2011, 2012a and 2015), and the majority of the information available for this carnivore covers traditional aspects concerning feeding habits (Guerrero et al., 2002), spatial ecology (González-Pérez et al., 1992; Hernández-Camacho and López-González, 2009), or evolution (Arroyo-Cabrales and Carranza-Castañeda, 2009). In the United States, however, there is considerable information on the parasites of this host (Buechner, 1944; Simmons et al, 1980; Dyer, 1984; Rogers, 1984; Davidson et al., 1992a and b; Steelman et al., 1998, 2000; Allen et al., 2011; Oates et al., 2012; Rosypal et al., 2013, and Ubelaker et al., 2015). The objective of our study was to generate basic information of the diversity of the parasite community of *Urocyon cinereoargenteus* in a remaining fragment of tropical highland deciduous forest in the state of Querétaro.

2. Materials and methods

2.1. Study area

The study was conducted in the Parque Nacional El Cimatario (PANEC) (20° 28′ 30″ and 20° 33′ 23″ N and 100° 19′ 37″ and 100° 23′ 12″ W), located south of the city of Santiago de Queretaro. The park has an area of 25 km^2 with thornscrub as the most abundant vegetation in the area, mixed scrubland, dry tropical deciduous forest and portions with two human-induced grasslands and reforested areas with introduced tree species (Baltasar et al., 2004; Hernández-Camacho et al., 2011).

2.2. Capture and handling of hosts

Due to the logistical difficulties of trapping during the rainy season, all foxes were captured during the dry seasons (December to May) of 2003, 2005, 2011, 2013, 2014, and 2015, with coil spring traps Duke traps #3 (Duke Company, West Point, MS, USA) and Victor soft catch #3 (Oneida Victor Inc. Ltd., Euclid, OH, USA) for medium-sized canids. Each captured individual was immobilized using zolazepam hydrochloride and tiletamine hydrochloride. The dosage depended on the weight of the animal (10 mg/kg), and administration was intramuscular, in the hindquarters. The animal was released from the trap as soon as the anesthetic took effect and placed in a recumbent position to be externally examined and its physical condition assessed. Meristic data were obtained including total length measured from the nostrils to the last caudal vertebrae, weight, sex, and estimation of age through tooth enamel color and gum recession. The animal's temperature and respiration were monitored constantly to detect any negative responses to the anesthetic. Once the animal recovered from the anesthesia, it was released in the same capture area. During 2013–2015, we also had access to three gray fox carcasses, which were processed by specialized veterinary staff from the Department of Natural Sciences at the Universidad Autonoma de Querétaro.

The procedure for capture, handling, and chemical restraint was carried out in accordance with the guidelines of the American Society of Mammalogists for the handling and restraint of wild mammals (Kreeger and Arnemo, 2007; Hernández-Camacho and López-González, 2009; Gannon et al., 2011) and with permits by Secretaria of Medio Ambiente y Recursos Naturales (SEMARNAT) for the corresponding years.

2.3. Sample processing

Parasites were processed according to the traditional techniques described by Lamothe (1997). Adult nematodes were fixated with warm ethanol 70% and processed in lactic acid for identification. Ecto parasites, such as fleas and ticks, were collected with forceps and fixated in ethanol 70%, fleas were processed with potassium hydroxide 10% and lactic acid and mounted in Canada Balsam. Blood smears were fixated with methanol 100% and dyed using Giemsa protocol (Lamothe, 1997). The parasites collected were deposited in the Colección Nacional de Helmintos [CNHE] at the Instituto de Biología and the Colección de Siphonaptera [SIPHO] at the Museo de Zoología, both of the Universidad Nacional Autónoma de México. A Box-Cox transformation was used to normalize parasite abundance data (MINITAB® 17 Statistical Software, 2010; Wang et al., 2008; Osborne, 2010), chosen for its robustness to stabilize variance and bearing in mind potential differences in the various types of sampling (feces, trappings and necropsies) during a decade of field work. Following the transformation, we further analyzed the data for parasite diversity (PAST statistical software version 2.17c; Hammer et al., 2001) using species richness indices Menhinick (D_{Mn}) and Margalef (D_{Mg}), Shannon's diversity index (H′), Simpson's diversity index (D) and Berger-Parker dominance index (d). These indices were selected for their varied and widely used role in biological diversity analysis (Magurran, 2004; Magurran and McGill, 2011).

3. Results

From December 2003 to February 2015, 24 gray foxes (14 females, 10 males) were caught in the PANEC and necropsies were performed on three additional animals (one female, two males) killed by feral dogs. All animals were within the range of weight and height for the species as described by Fritzell and Haroldson (1982) (mean weight and SD of 3.8 ± 0.27 kg and mean length and SD of 1020.62 ± 35.7 mm for females and a mean weight and SD of 4.23 ± 0.25 kg and mean length and SD of 1063 ± 11.54 mm for males). All individuals were considered as adults.

A total of six species of parasite arthropods and 11 species of endo parasites were found in the foxes. The species richness indices were $D_{Mn} = 1.137$ and $D_{Mg} = 2.836$, the diversity indices were$H' = 1.78$ and $D = 0.7398$, but with a dominance Berger-Parker index of $d = 0.4161$. These results reflect the dominance of *Uncinaria stenocephala* as the most abundant parasite found in these foxes including parasites species previously recorded to 2013 (Hernández-Camacho et al. (2011). From 2013 to 2015, the number of parasite species in gray foxes increased almost two-fold with the detection of two additional species of nematode: *Dirofilaria immitis* filariae (first detected in 2014; CNHE access number 9905), *Toxocara canis* adult nematodes (CNHE access number 9906) previously recorded in feces analysis in Hernández-Camacho et al. (2011) and *Physaloptera praeputialis* adult nematodes (CNHE access number 9907), and six species of arthropods: *Pulex simulans* (first detected 2013, SIPHO access number 8832), *Echidnophaga gallinacea* (2014, SIPHO 8825, 8826), *Ctenocephalides canis* (2014; SIPHO 8828, 8829), *C. felis* (2014; 8830, 8831), and *Euhoplopsyllus glacialis affinis* (2015, SIPHO 8824); and *Ixodes* sp. (2014, species identification in progress) possibly representing a new record for Mexico.

4. Discussion

Our study represents one of the first long-term studies on the gray fox parasite community in Mexico. Although we used a very simple processing program for diversity analysis, the basic information provided on parasites diversity in these animals, which live in a protected area surrounded by an anthropic environment, is relevant to future management programs in both these habitat types in Mexico. Our results on the parasite community show that the diversity of parasites of the gray fox in the PANEC comprises primarily generalist species, of which, most of them have a direct life cycle not requiring intermediate hosts, with life cycles with several transmission strategies as paratenic hosts, autoinfection or transplacental transmission such as in *U. stenocephala* or *Strongyloides stercoralis* life cycles (Anderson, 2000). Our data differs from those in studies conducted in the United States regarding the parasite community of *Urocyon cinereoargenteus*, where many of the parasite species have complex life cycles, requiring at least two species of intermediate hosts (Conti, 1984; Dyer, 1984; Anderson, 2000; Ubelaker et al., 2015).

All flea species found during this study except *E. glacialis affinis*, were new records for *U. cinereoargenteus* in Querétaro, and *E. glacialis affinis* is a new record for gray foxes in Mexico (Ayala-Barajas et al., 1988). *Pulex simulans* is the most common flea in gray foxes in the United States and may indicate that this wild canid may come into contact with domestic dogs in the study area (Dobler and Pfeffer, 2011). The PANEC is a protected natural area with human presence for the past 33 years. Historically, El Cimatario has been impacted by agricultural and extractive production since the 16t^{h} century, by both the indigenous population and the Spanish conquistadors in the Colonial period (Escobar-Ledesma, 2002; Valencia-Cruz, 2010). These early impacts may have resulted in the extirpation of parasite communities with complex life cycle as a consequence of significant ecosystem modification.

It is also important to note that the flea species reported in this study, are potential vectors of arbovirus or bacteria such as *Rickettsia* spp. (Dobler and Pfeffer, 2011; Acosta-Gutiérrez, 2014); although the actual human health scenario in Mexico is still, in general, unknown. Presently, there is no record of any micro parasite species using these recorded parasitic arthropod species as vectors, despite analyses performed in a laboratory specialized in the diagnosis of *Erlichia canis* and *Babesia canis* (Hernández-Camacho, unpublished data) in these foxes. However, this possibility is not ruled out and regular monitoring of the *U. cinereoargenteus* population is recommended in order to avoid possible epizootic and zoonotic diseases of importance in the medium or long term.

Recently, there have been changes in the policy of access to the PANEC, allowing people to enter the park since 2005, with a corresponding increase in the presence of feral dog packs as a consequence of domestic dogs neglect. These packs subject the community of mesocarnivores to pressure (Reed and Merenlender, 2011), causing the wild canids to avoid paths and trails that they used to travel according to the records of feces observed in transects surveyed in the study conducted by Hernández-Camacho et al. (2011), compared to 2015 data showing only 10% of the amount of feces observed in the previous study. This situation may have an impact on the gray fox distribution in the park, from 2 individuals/km^2, estimated in Hernández-Camacho, and López-González (2009), the species richness of fleas reported in the current paper might be related to this change, and perhaps also to shifts in contact between the foxes and domestic y feral dogs.

The detection of *Dirofilaria immitis* filariae in traditional blood smears of a gray fox was a notable discovery, considering the low prevalence of this species in gray foxes in the United States (Simmons et al., 1980; King and Bohning, 1984; Wixsom et al., 1991), based on necropsy and the reduced probability of recording the filariae in blood because of low infections (Simmons et al., 1980; Anderson, 2000), considering then this is a one-time opportunity for blood sampling, although adult nematodes have been not registered in the necropsied canids and not through indirect method such as blood smears as. For Querétaro, adult heartworms had previously been recorded in stray dogs, in 1.3% of 378 stray dogs (Cantó et al., 2011), in a road-killed coyote near a landfill west of the city, in which adult nematodes were found in the heart (Hernández-Camacho and Pineda-López, 2012b), and in gray foxes from another population located 20 km north of the PANEC, diagnosed by blood smear examination (Hernández-Camacho et al., 2015).

5. Conclusions

The results reported here, together with those reported previously, constitute one of the first long-term studies of parasite communities in Mexican carnivores. The study provides new information of the species richness and diversity of the parasite community of *Urocyon cinereoargenteus* in a remaining fragment of tropical highland deciduous forest habitat, surrounded by an anthropic environment in central Mexico. Data from this study suggest an almost two-fold increase in the number of parasites reported for this host in Mexico, and provide guidance for the future management of carnivores and their parasites in protected natural areas in this country; these due to inclusion of flea records. We consider that this study may be an example of the potential scenario in parasite community of carnivores living in an anthropic environment in Mexico and it will be useful for future management strategies in natural protected areas in the country.

Conflict of interests

None.

Acknowledgments

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