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Authors: McFadden, Katherine W., Wade, Susan E., Dubovi, Edward J., and Gompper, Matthew E.

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## A Serological and Fecal Parasitologic Survey of the Critically Endangered Pygmy Raccoon (*Procyon pygmaeus*)

Katherine W. McFadden,<sup>1</sup> Susan E. Wade,<sup>2</sup> Edward J. Dubovi,<sup>3</sup> and Matthew E. Gompper<sup>4,5</sup> <sup>1</sup> Department of Ecology, Evolution and Environmental Biology, Columbia University, 1200 Amsterdam Ave., MC 5557, New York, NY 10027, USA; <sup>2</sup> New York State Animal Health Diagnostic Center, Parasitology Section, College of Veterinary Medicine, Cornell University, Ithaca, NY 14853, USA; <sup>3</sup> New York State Animal Health Diagnostic Center, Virology Section, College of Veterinary Medicine, Cornell University, Ithaca, NY 14853, USA; <sup>4</sup> Department of Fisheries and Wildlife Sciences, 302 Anheuser-Busch Natural Resources Building, University of Missouri, Columbia, MO 65211, USA; <sup>5</sup> Corresponding author (email: gompper@missouri.edu)

**ABSTRACT:** The pygmy raccoon (*Procyon pygmaeus*) of Cozumel Island, Mexico, is among the most endangered carnivores in the world, and causes of its decline are unclear. During 2002 and 2003, we sampled approximately 10% of the remaining population to survey exposure to viral and parasitic pathogens that may have contributed to population decline. We found evidence of exposure to infectious canine hepatitis, canine distemper, feline panleukopenia virus, and *Toxoplasma gondii*. The latter is suggestive of spillover from domestic cats, which have only recently been introduced to the island. Additional parasites identified include *Eimeria nutalli*, *Placoconus lotoris*, *Capillaria procyonis*, *Physaloptera* sp., a mite in the family Listrophoridae, and a trematode in the family Heterophyidae. Several of these are typical of the parasite community of the common raccoon (*Procyon lotor*).

**Key words:** Cozumel Island, Mexico, disease spillover, endoparasites, *Procyon pygmaeus*, raccoon, viral pathogens.

The pygmy raccoon (*Procyon pygmaeus*) is one of three Carnivora endemic to 486 km<sup>2</sup> Cozumel Island (20°16' to 20°26'N and 86°44' to 87°02'W) in southeastern Mexico. With a recommended IUCN threat status of critically endangered, these taxa are among the most threatened of the world's carnivores (Cuarón et al., 2004; McFadden, 2004); recent surveys suggest a total population size of <200 mature individuals for the pygmy raccoons and far less for the other species (the dwarf coati, *Nasua nelsoni*, and an unnamed taxon of fox, *Urocyon* sp.). The parasites and diseases of Cozumel carnivores have not been surveyed, although disease-related concerns are well recognized as critical for carnivore conservation, in part due to the potential for large populations

of domestic cats (*Felis catus*) and dogs (*Canis familiaris*) to act as reservoirs for generalist pathogens (Funk et al., 2001; Fiorello et al., 2004). As part of a larger study (McFadden, 2004) on the evolution and ecology of pygmy raccoons, we collected blood samples (for genetic studies) and fecal samples (for dietary studies), which allowed for an initial serological and fecal parasitologic survey.

Cozumel has significant populations of both feral and privately owned unvaccinated dogs and cats. Therefore, our serology survey was designed to assess for the presence of pathogens that may have been influenced by the sympatric domestic carnivore populations as well as pathogens that are recognized as conservation concerns for carnivores. We tested for antibodies to canine distemper virus (CDV), infectious canine hepatitis (ICH; =canine adenovirus), feline panleukopenia virus (FPV; =feline or raccoon parvovirus), and the protozoan *Toxoplasma gondii*. Each of these has been reported, sometimes at relatively high prevalence, in the common raccoon (*Procyon lotor*), and the viruses may be important causes of raccoon mortality (Roscoe, 1993; Barker and Parrish, 2001; Woods, 2001). Positive diagnosis of *T. gondii* may indicate spillover from domestic cats, as there is no recent or historic record of wild felid species on Cozumel (Hamblin, 1984). For fecal-borne parasites, we expected few endoparasitic species given the long (ca. 50,000 yr; McFadden, 2004) isolation of these animals from mainland Yucatan congeners. Island host populations are often depauperate in

numbers of endoparasitic species due to small population sizes of hosts, lack of intermediate hosts, as well as historic bottlenecks (Dobson, 1988; Pisanu et al., 2001).

Pygmy raccoons were live-trapped between 2002 and 2003 in the northwestern mangrove forests of Cozumel, where it appears the last sizable populations of the species occur. Details on trapping and handling are discussed in McFadden (2004). In brief, captured animals were anesthetized with ketamine hydrochloride (Fort Dodge Laboratories, Fort Dodge, Iowa, USA; 10 mg/kg) and xylazine (Vedco, Inc., St Joseph, Missouri, USA; 2 mg/kg), after which individuals were sexed and aged by size, dentition, reproductive characters, and capture history. Blood was collected from 28 individuals (13 adult males, seven adult females, three immature males, five immature females) by jugular venipuncture using a 21-gauge needle and both additive-free and heparin-containing vacutainer tubes. Blood samples were immediately centrifuged and the serum fraction frozen ( $-80^{\circ}\text{C}$ ) until laboratory analysis. Serologic testing for antibodies to infectious canine hepatitis and CDV was done using serum neutralization (SN), testing for FPV antibodies was done by hemagglutination-inhibition (HAI), and *T. gondii* antibody titers were determined by an indirect hemagglutination (IHA). Details on serologic test methodologies for viruses are given by Kimber et al. (2000). Fecal samples were collected from 25 individuals (11 adult males, five adult females, three immature males, six immature females) and preserved and stored in 10% formalin at room temperature until analysis. Fecal-borne parasites were prepared for microscopy using standard centrifugation concentration flotation techniques with zinc sulfate and sugar flotation media and identified to taxon based on morphometric criteria (Georgi et al., 1990).

McFadden (2004) estimated an island-wide population of pygmy raccoons of 300 animals based on extensive mark-recap-

ture efforts. Thus, our sampling represents approximately 8–10% of the global population of *P. pygmaeus*. Antibodies were detected to each of the pathogens examined. Prevalence of antibodies (#seropositive/#examined) for CDV was 4% due to a single weakly positive (titer = 8) adult male. Given this single, weak-positive individual, further examination of the prevalence of CDV in native and domestic carnivores is necessary to confirm the presence and importance of this virus on Cozumel. Three adult males (11%) were positive (two at 8; one at 32) for antibodies to ICH. Antibodies to raccoon parvovirus (FPV) were detected (7%) in two adult males (10; 1280). Antibodies to *T. gondii* were identified in 11% of individuals: two adult females (2048; 1024) and one adult male (1024). Multiple seropositive results occurred in one adult male who was positive for antibodies to ICH, FPV, and *T. gondii*.

Based on fecal analyses, mean number of parasitic species per host was 2.0 (SD=0.65; range=1–3). Five endoparasitic species were identified: *Eimeria nutalli* (36% prevalence), *Placoconus* (= *Arthrocephalus*) *lоторis* (96%), *Capillaria procyonis* (16%), a digenean fluke in the family Heterophyidae (40%), and a Spirurida species probably in the genus *Physaloptera* (4%). A fur mite in the family Listrophoridae was also identified in the feces of two individuals (8%). For all species, there were no significant relationships (Fisher exact tests;  $P \geq 0.05$ ) between prevalence and sex or prevalence and age. These fecal-borne parasites are typical representatives of the parasite fauna of the common raccoon (*P. lotor*), and their presence indicates that, despite the small host population size and the island environment, a relatively diverse fauna of directly (*P. lotoris*, *C. procyonis*) and indirectly transmitted (flukes) parasites have been maintained.

Pathogens such as ICH, FPV, and CDV commonly occur in *P. lotor* populations, and it is unclear if the exposure of pygmy raccoons is recent or historic. Nonetheless,

given the critically endangered status of the pygmy raccoon population, coupled with the ability of these pathogens to persist in reservoir populations of unvaccinated domestic dogs and cats, any evidence of current viral exposure is problematic. *Toxoplasma gondii* is likely novel to the Cozumel carnivore fauna, which historically lacked felids. Domestic and wild felids are the only definitive hosts for this protozoan parasite, so its presence in the raccoon population indicates probable spillover from introduced domestic cats. Such spillover of new disease-causing agents, coupled with the large reservoir populations of domestic carnivores, may be contributing (Cuarón et al., 2004) to the decline and apparent lack of recovery of the pygmy raccoon as well as other endemic Cozumel carnivores.

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