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Short Communications

**A serological and fecal parasitological survey of the critically endangered pygmy
raccoon (*Procyon pygmaeus*)**

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

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

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16 The pygmy raccoon (*Procyon pygmaeus*) is one of three Carnivora endemic to
17 486 km² Cozumel Island (20°16' to 20°26'N and 86° 44' to 87°02'W) in southeastern
18 Mexico. With a recommended IUCN threat status of critically endangered, these taxa are
19 among the most threatened of the world's carnivores (Cuaron et al. 2004; McFadden
20 2004), as recent surveys suggest a total population size of <200 adult individuals for the
21 pygmy raccoons and far less for the other species (the dwarf coati *Nasua nelsoni* and an
22 unnamed taxon of fox, *Urocyon* sp.). The parasites and diseases of Cozumel carnivores
23 have not been surveyed, although disease-related concerns are well recognized as critical

24 for carnivore conservation, in part due to the potential for large populations of domestic
25 cats (*Felis catus*) and dogs (*Canis familiaris*) to act as reservoirs for generalist pathogens
26 (Funk et al. 2001; Fiorello et al. 2004). As part of a larger study (McFadden 2004) on the
27 evolution and ecology of pygmy raccoons, we collected blood samples (for genetic
28 studies) and fecal samples (for dietary studies) which allowed for an initial serological
29 and fecal parasitological survey.

30 Cozumel has significant populations of both feral and privately owned,
31 unvaccinated dogs and cats. Therefore our serology survey was designed to assess for the
32 presence of pathogens which may have been influenced by the sympatric domestic
33 carnivore populations, as well as pathogens which are recognized as conservation
34 concerns for carnivores. We surveyed for antibodies to canine distemper virus (CDV),
35 infectious canine hepatitis (ICH; = canine adenovirus), feline panleukopenia virus (FPV;
36 = feline or raccoon parvovirus), and the protozoan *Toxoplasma gondii*. Each of these has
37 been reported, sometimes at relatively high prevalence, in the common raccoon (*P. lotor*),
38 and the viruses may be important causes of raccoon mortality (Roscoe 1993; Barker and
39 Parrish 2001; Woods 2001). Positive diagnosis of *T. gondii* may indicate spillover from
40 domestic cats, as there is no recent or historic record of wild felid species on Cozumel
41 (Hamblin 1984). For fecal-borne parasites, we expected few endoparasitic species given
42 the long (ca 50K yrs; McFadden 2004) isolation of these animals from mainland Yucatan
43 congeners. Island host populations are often depauperate in numbers of endoparasitic
44 species due to small population sizes of hosts, lack of intermediate hosts, as well as
45 historic bottlenecks (Dobson 1988; Pisanu et al. 2001).

46 Pygmy raccoons were live-trapped between 2002 and 2003 in the northwestern
47 mangrove forests of Cozumel where it appears the last sizable populations of the species
48 occur. Details on trapping and handling are discussed in McFadden (2004). In brief,
49 captured animals were anesthetized with ketamine hydrochloride (Fort Dodge
50 Laboratories, Fort Dodge, Iowa; 10 mg/kg) and xylazine (Vedco, Inc., St Joseph,
51 Missouri; 2 mg/kg), after which individuals were sexed and aged by size, dentition,
52 reproductive characters, and capture history. Blood was collected from 28 individuals (13
53 adult males, 7 adult females, 3 immature males, 5 immature females) by jugular
54 venipuncture using a 21g needle and both additive-free and heparin-containing vacutainer
55 tubes, and immediately centrifuged and the serum fraction frozen (-80°C) until laboratory
56 analysis. Infectious canine hepatitis and CDV challenge was assessed by serum
57 neutralization (SN) tests, FPV was assessed by hemagglutination-inhibition (HAI) tests,
58 and *T. gondii* titers were assessed by an indirect hemagglutination test (IHA). Details on
59 testing methodologies for viruses are given by Kimber et al. (2000). Fecal samples were
60 collected from 25 individuals (11 adult males, 5 adult females, 3 immature males, 6
61 immature females) and preserved and stored in 10% formalin at room temperature until
62 analysis. Fecal-borne parasites were prepared for microscopy using standard
63 centrifugation concentration flotation techniques with zinc sulfate and sugar flotation
64 media, and identified to taxon based on morphometric criteria (Georgi et al. 1990).

65 McFadden (2004) estimated an island-wide population of pygmy raccoons of ca
66 300 animals based on extensive mark-recapture efforts. Thus our sampling represents ca
67 8-10% of the global population of *P. pygmaeus*. Antibodies were detected to each of the
68 pathogens examined. Prevalence of infection (# infected/# examined) for CDV was 3.6%

69 due to a single weakly positive (titer positive at 1:8) adult male. Given the weakness of
70 this single positive individual, further examination of the prevalence of CDV in native
71 and domestic carnivores is necessary to confirm the presence and importance of this virus
72 on Cozumel. Three adult males (10.7%) were positive (two at 1:8; one at 1:32) to ICH.
73 Raccoon parvovirus (FPV) was diagnosed (7.1%) in two adult males (1:10; 1:1280).
74 Antibodies to *T. gondii* were identified in 10.7% of individuals: two adult females
75 (1:2048; 1:1024) and one adult male (1:1024). Multiple infections occurred in one adult
76 male who was positive for ICH, FPV, and *T. gondii*.

77 Based on fecal analyses, mean number of parasitic species per host was 2.0 (s.d. =
78 0.65; range = 1–3). Five endoparasitic species were identified: *Eimeria nutalli* (36%
79 prevalence), *Placoconus* (= *Arthrocephalus*) *lotoris* (96%), *Capillaria procyonis* (16%), a
80 digenean fluke in the family Heterophyidae (40%), and a Spirurida species probably in
81 the genus *Physaloptera* (4%). A fur mite in the family Listrophoridae was also identified
82 in the feces of two individuals (8%). For all species there were no significant
83 relationships (Fisher exact tests; $p \geq 0.05$) for any species between prevalence and sex, or
84 prevalence and age. These fecal-borne parasites are typical representatives of the parasite
85 fauna of the common raccoon (*P. lotor*), and their presence indicates that despite the
86 small host population size and the island environment, a relatively diverse fauna of
87 directly (*P. lotoris*, *C. procyonis*) and indirectly-transmitted (flukes) parasites have been
88 maintained.

89 Pathogens such as ICH, FPV, and CDV are typical of *P. lotor* populations, and so
90 it is unclear if the exposure of pygmy raccoons is recent or historic. Nonetheless, given
91 the critically endangered status of the pygmy raccoon population, coupled with the ability

92 of these pathogens to persist in reservoir populations of unvaccinated domestic dogs and
93 cats, any evidence of current viral exposure is problematic. *Toxoplasma gondii* is likely
94 novel to the Cozumel carnivore fauna, which historically lacked felids. Domestic and
95 wild felids are the only definitive hosts for this protozoan parasite, and so its presence in
96 the raccoon population indicates spillover from introduced domestic cats which are the
97 only felids on the island. Such spillover of new disease-causing agents, coupled with the
98 large reservoir populations of domestic carnivores which likely enhance the impact of
99 generalist viruses of carnivores, may be contributing (Cuarón et al 2001) to the decline
100 and apparent lack of recovery of the pygmy raccoon as well as other endemic Cozumel
101 carnivores.

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