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HEALTH EVALUATION OF FREE-RANGING AND CAPTIVE PICHIS (*ZAEDYUS PICHIY*; MAMMALIA, DASYPODIDAE), IN MENDOZA PROVINCE, ARGENTINA

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ABSTRACT: The health of free-ranging and captive pichis (*Zaedyus pichiy*) was assessed in Mendoza Province, Argentina, between November 2001 and December 2006. Postmortem examinations of 150 confiscated and vehicle-killed pichis and clinical examinations of 139 wild-caught individuals suggest that the wild populations are currently in good health. Lesions and scars were observed in a large proportion of wild-caught pichis. The most common lesions were associated with parasitism or parasite larva migration. *Sarcocystis* cysts were relatively common in the skeletal muscle, and *Besnoitia* cysts were observed in the lungs of 24 evaluated animals. Elevated ambient humidity levels often caused moist dermatitis with epidermal detachment in captive pichis. This report constitutes the first health evaluation of free-ranging and captive *Z. pichiy*. It will be a starting point for future health studies and will be beneficial for the captive management of this species.

Key words: Besnoitia, histopathology, parasitology, pathology, serology, *Trypanosoma cruzi*, wildlife health.

INTRODUCTION

The pichi (Zaedyus pichiy) is a small (approximately 1 kg body mass) semifossorial armadillo species endemic to Argentina and Chile that enters hibernation during winter months (Superina and Boily, 2007; Superina, 2008). Pichis are diurnal and opportunistic omnivores with a preference for insects. Although neither historic nor current census data are available, lower encounter rates in Mendoza Province, in central west Argentina, suggest that wild populations have decreased considerably in the past years. This depletion of wild populations has recently led to the inclusion of Z. pichiy in the International Union for Conservation of Nature (IUCN) Red List of Threatened Species, where it is listed as "near threatened" (Fonseca and Aguiar, 2004). The apparent population declines may have been partially caused by poaching and disease epidemics. There have been unconfirmed reports of disease outbreaks of unknown etiology severely affecting wild populations in some areas of Mendoza, but the potential role of diseases in these population reductions remains undefined. Although wild specimens of other armadillo species have been screened for specific pathogens and parasites, such as Leptospira (Carillo et al., 1972), Salmonella (Quevedo et al., 1978), or Sarcocystis (Lindsay et al., 1996), only one study (Mazza et al., 1935) included pichis. The purpose of this study was to describe the lesions affecting wild and captive pichis of Mendoza Province, Argentina. These baseline data are a starting point for future health studies and will contribute to the investigation of the causality between human encroachment into wildlife habitat and disease occurrence. Furthermore, the information presented here will be beneficial for the captive management of this species.

MATERIALS AND METHODS

Project location and sample and data collection

Samples from free-living pichis were collected across vehicle, walking, and horseback transects of varying length in randomly chosen areas of Mendoza Province, Argentina $(32^{\circ}00' - 37^{\circ}30' \text{S}, 66^{\circ}30' - 70^{\circ}00' \text{W}).$ The southern part of Mendoza Province belongs to the Patagonian steppe ecoregion (Olson et al., 2001). The arid climate and the poor volcanic soil, covered with sands of variable depth, together determine the scarce vegetation (Candia et al., 1993), which is composed of grasslands with isolated shrubs, areas with open bush communities, and basaltic steps (Candia and Dalmasso, 1995). Trees are absent from this region. The arid northeastern part of Mendoza is part of the Low Monte ecoregion (Olson et al., 2001). The dry climate and sandy grounds favor the growth of the characteristic monte vegetation, which consists of small trees, shrubs, and scarce pasture (Peralta de Galmarini and Martinez Carretero, 1995).

Transect sampling was performed in all seasons and at all times of day between November 2001 and December 2006. In total, 139 wild-caught pichis (87 males, 52 females) were captured by hand. Capture sites were registered with a handheld global positioning system (GPS), and environmental temperature was measured with a digital thermometer. Thirty-three animals were considered juveniles, 22 were classified as yearlings, and 84 were adults based on their body mass, carapace length and width, the presence and absence of scars, and capture date. Physical examinations and sample collections were carried out under manual restraint. Clinical examinations focused on body condition, visible external lesions and scars, signs of pathologic processes, ectoparasite load, and reproductive status. The animals were measured, weighed with a spring scale (Pesola AG, Baar, Switzerland), and their rectal temperature was registered with a digital thermometer (TES Electrical Electronic Corp., Taipei, Taiwan). Ticks and fleas were collected and stored in 96% ethanol for identification. Fecal samples were collected for parasitology and kept refrigerated. Blood samples were obtained by venipuncture of the medial coccygeal vein. Due to the small size of this vein and rapid coagulation, blood was collected directly from the needle into heparin-coated and uncoated microcapillaries (Biocap S.A., Buenos Aires, Argentina), which were subsequently sealed with plasticine (Critoseal, Oxford Labware, St. Louis, Missouri, USA). Thin smears of fresh blood were air-dried. Coated microcapillaries were stored in a cool box with cold packs and transported to the laboratory, while uncoated microcapillaries were first kept at 30 C until coagulation and retraction of the blood clot occurred, after which they were transferred to the cool box. Rectal temperature was measured no later than 5 min after capture, and blood samples were extracted within 15 min. All pichis were released at their capture site within 45 min.

Captive pichis included wild-caught and captive-born pichis that were maintained 1-26 mo in a private facility in Luján de Cuyo, Mendoza, Argentina (33°01'S, 68°55'W). The enclosure consisted of individual pens made of wire mesh and sheet metal, $2 \times 1.5 \times 2.5$ m each; soil to a depth of 2 m was provided as a natural substrate for digging. Food, consisting of a varying mixture of fruits, vegetables, meat, dry cat food, rice, and a vitamin-mineral supplement, was offered once daily; water was provided ad libitum. Pichis were visually inspected at feeding time; individuals presenting lesions or signs of disease were removed from the enclosure and maintained separately until their recovery.

Dead wild pichis were obtained from collaborating inspectors and rangers of the Mendoza Department of Natural Renewable Resources; confiscated carcasses were refrigerated prior to submission. Necropsies were performed on vehicle-killed animals (12 males, four females) and confiscated pichis (58 males, 82 females, five of unknown sex), as well as on captive pichis that died during the study period (seven males, five females). The animals were classified as juveniles, yearlings, or adults based on morphologic features, such as carapace length and width, and the presence or absence of scars. They were examined for macroscopic lesions, and representative tissue samples of reproductive organs, heart, lungs, liver, kidney, diaphragm, skeletal muscle, skin, intestine, lymph nodes, adrenal glands, and any macroscopic lesion were collected when available. Not all organs could be sampled from all individuals because they often were eviscerated at the time of confiscation. Tissues were stored in 10% neutral-buffered formalin.

Sample and data analysis

Hematologic and serum chemistry analyses were performed in collaboration with Laboratorio Mera, Mendoza, Argentina, using routine laboratory techniques as described by Superina and Mera y Sierra (2008). All examined pichis were considered clinically healthy, and blood values were used to establish reference values (Superina and Mera y Sierra, 2008). Microbiologic samples from captive pichis were inoculated in sheep blood agar, MacConkey Agar, and Sabouraud Agar and cultured aerobically at 37 C. Identification of isolates was done with routine microbiologic techniques (Finegold and Baron, 1986). Commercial test kits based on indirect hemagglutination were used to test for antibodies to Toxoplasma gondii and Trypanosoma cruzi (Toxotest HAI, Wiener Laboratory, Rosario, Argentina; and Chagas HAI, Polychaco S.A.I.C., Buenos Aires, Argentina, respectively). Samples were considered positive if agglutination was present at dilutions $\geq 1:16$. Ectoparasites and fecal samples were submitted to the Centro de Parasitología y Vectores, La Plata, Argentina, for analysis and were identified based on morphologic characteristics. Fecal samples were analyzed by means of the modified McMaster method and Willis flotation technique with Sheather solution (Hawken, 1983; Hendrix, 1998). Tissues for histopathologic analysis were fixed in Bouin's fluid, embedded in paraffin, sectioned at 5 µm, and stained with hematoxylin and eosin (HE). Histologic slides were screened by light microscopy for signs of pathologic alterations and presence of pathogens.

Data analyses were performed with a statistical software program (SPSS, version 11.0, SPSS Inc., Chicago, Illinois, USA). The ectoparasite load of male and female pichis, and of individuals of different age classes, was compared with an analysis of variance (AN-OVA). Fisher's exact and chi-square tests were used to compare the prevalence of histopathologic lesions per affected organ in juveniles, yearlings, and adults, and in males and females, respectively. A Kruskal-Wallis test was used to compare the prevalence of scars among age classes. Calculated P values ≤ 0.05 were considered statistically significant.

This project was approved by the Institutional Animal Care and Use Committee of the University of New Orleans and the Dirección de Recursos Naturales Renovables of Mendoza Province, Argentina. Histologic slides were shipped to the US with export permits from the Dirección de Fauna Silvestre of Argentina and under a US Fish and Wildlife Service Import License.

RESULTS

Live wild-caught pichis

The average rectal temperature outside of the hibernation season was 35.2 ± 1.2 C. Rectal temperature was highly variable (range 32.2–38.3 C, n=89), but this variability was not related to environmental temperature or age. Although rectal temperature was measured as soon as possible after the animals were captured, it is possible that it was elevated because of stress related to capture and handling. The temperature of one adult male sampled during hibernation was 18.9 C, similar to the temperature measured within its burrow (18 C).

Clinical signs were observed in 25 of 139 animals and included emaciation, apathy, dehydration, decalcified carapace border, pale mucosa, inflammation and abscesses on the penis, and infected wounds. Lesions were observed on 31 pichis and included skin lesions, missing or worn out scutes, or fractured scutes. Males and females were equally affected (23% vs. 21%; Mann-Whitney U-test, P > 0.05). The prevalence of lesions was not significantly different in adults, yearlings, and juveniles (24%, 36%, and 9%, respectively; Kruskal-Wallis test, P > 0.05). Scars were observed in 64 animals and affected mainly the bands, head shield, carapace border, and tail. Forty-six percent of all males, and an identical percentage of females, had one or more scars. No significant difference in the prevalence of scars was observed among age classes (51% adults; 50% yearlings; 30% juveniles; Kruskal-Wallis test, P > 0.05). Four individuals had leukocytosis with neutrophilia.

Fecal analyses were performed on 53 animals, and intestinal parasites were detected in 93% of the samples. Aspidodera spp. were detected in 79% of these animals, followed by Eimeria oocysts (64%), Trichostrongylidea eggs (26%), Mathevotaenia sp. (9%), Cyclobulura sp. (6%), and *Trichuris* sp. (4%). Fleas of the species Malacopsylla grossiventris or Phthiropsylla agenoris were found on 84 of 139 examined armadillos. Amblyomma *pseudoconcolor* was found on a single pichi from northern Mendoza. Males had similar burdens of ticks and fleas as females, but significantly more yearlings than juveniles were infested (ANOVA, P=0.045). No blood parasites were observed. All of the 24 tested wild pichis, and all of the 11 evaluated captive pichis, were seronegative for *Toxoplasma gondii*. Two out of 25 tested wild individuals were seropositive for *Trypanosoma cruzi*, with titers of 16 and 64.

Captive pichis

Health problems were observed in only a few captive pichis. One captive adult male was affected by a severe acute hepatitis with subcutaneous hemorrhages and anemia. Antibiotic treatment was supplemented with intramuscular (IM) injections of 2 mg/kg menadione (Mestil-Ka, Fada Pharma, Buenos Aires, Argentina) for 5 days. Two males required isolation and antibiotic treatment of infected lesions during the reproductive season. Six animals that had been temporarily maintained in 200-l barrels filled with sand developed reddened, moist cutaneous lesions and skin detachment on paws, abdomen, and face. The affected animals were kept in a dry environment during treatment with antiseptics and antibiotics. Menadione was administered IM for 5 days due to prolonged bleeding after IM injections. All affected animals fully recovered and were released in their enclosures after 2 to 3 wk of treatment.

Dead pichis

Tissue samples were collected from 173 dead pichis; 18 of these were not analyzed histologically due to advanced autolysis. The gender of five individuals could not be determined because reproductive organs had been removed; these were excluded from further analyses. Consequently, results are reported for 150 pichis (87% of sampled dead animals). Of these, 120 had been confiscated from poachers, 10 were hit by cars, 10 drowned in captive conditions after intense rainfall that flooded their enclosures, and 10 died for other reasons, including lack of adaptation to captive conditions (one juvenile male), hypothermia (one adult female), and unknown causes (eight individuals).

Animals were not examined for macroscopic external lesions because most pichis

had extensive destruction of the carapace due to dog bites (poached individuals) or because they were hit by a car. Many skin samples had burn artifacts, such as edema and clots, associated with poaching; poachers often hold eviscerated pichis over an open flame to avoid decomposition. Nevertheless, these burn artifacts did not significantly impede the microscopic interpretation of skin samples. Macroscopic lesions on internal organs were rarely observed. Several lactating females and one adult male had enlarged adrenal glands. The liver was yellowish or ocher in five adults and one yearling. An enlarged, sometimes granulated spleen was observed in seven animals. Three adult males and one juvenile female had macroscopic lesions on the heart consisting of a whitish apex or a whitish spot on the ventricle. The heart was dilated in three adults. One juvenile male, which had been rescued from a cattle guard, had a circular lesion of 0.9×1.3 cm with myiasis on the right leg. One juvenile female had a subcutaneous abscess caudal to the right elbow that was associated with *Klebsiella* and filamentous bacteria related to a penetrating wound. Finally, one adult female had a cecal perforation that led to peritonitis, lymphadenitis of the mesenteric lymph node, and sepsis.

Histopathologic analysis revealed microscopic lesions in almost all studied pichi lungs (98% of studied animals) and over 75% of all examined skeletal muscles, hearts, and gastrointestinal tracts, while female reproductive organs and spleens rarely had microscopic lesions (Table 1). In general, the prevalence of affected organs was similar in male and female pichis, as well as in individuals of different age classes. Lesions of the adipose tissue were more common in adult males than in females of the same age class (P < 0.0001, Fisher's exact test), while the latter had significantly more hepatic lesions than the former (P < 0.05). Hepatic lesions were also more common in yearling females than in males of the same age class

Affected organ system	Number of analyzed samples ^a	Number of samples with histopathologic lesions	Predominant lesions	Number of individuals affected by predominant lesions ^b
Skin	89	45	Dermatitis	39
			Abscess	1
Skeletal muscle	145	111	Rhabdomyolysis	97
			Sarcocystis cysts	15
			Inflammation	9
Adipose tissue	141	5	Inflammation	3
Heart	129	97	Necrosis	87
			Myocarditis	11
Lungs	132	129	Inflammation	110
			Acute pulmonary shock	105
			Microgranuloma	27
			Besnoitia cysts	24
Lymph node	59	8	Hemosiderosis	4
			Lymphadenitis	2
Liver	106	73	Periportal lymphocytic inflammation	65
Kidney	130	39	Pyelonephritis	18
			Tubular mineralization	11
			Chronic interstitial nephritis	5
Ureter	53	36	Inflammation	35
Spleen	56	2	Besnoitia cysts	1
Tongue	55	11	Inflammation	9
			Sarcocystis cysts	1
Stomach	15	3	Gastritis	3
Intestine	33	27	Enteritis	27
Colon	27	20	Colitis	19
Adrenal glands	84	8	Shock	3
			Nodular hyperplasia	2
Testes	30	5	Orchitis	4
Ovary	42	0		
Uterus	47	3	Endometritis	2

TABLE 1. Histopathologic lesions by system in wild pichis Zaedyus pichiy.

^a Note that not all organs were available for sampling in all 150 examined individuals.

^b Note that one individual may have had several lesions in one organ.

(P < 0.05), and their prevalence was significantly different among age classes of female pichis, where yearlings were more frequently affected than adults and juveniles had the lowest prevalence (P < 0.05, chi-square). The most common microscopic lesions were hypersensitivity dermatitis; chronic lymphoplasmacytic inflammation of the gastrointestinal tract attributed to parasitism; varying degrees of pulmonary granulomatous inflammation and peribronchiolar lymphocytic inflammation attributed to Besnoitia sp. infection; myocardial necrosis and pulmonary congestion, edema, hemorrhage, and atelectasis attributed to acute pulmonary shock; and rhabdomyolysis of the skeletal muscle attributed to stress (Table 1).

Capture stress or trauma, such as hunting dog bites, was associated with acute pulmonary shock in 105 evaluated animals (Table 1). In the skeletal muscle and tongue, the acute stress response was manifested as rhabdomyolysis, while mild myocardial necrosis related to stress or shock was the most common finding in the cardiac muscles (87 pichis). The acute shock related to capture and killing was manifested as renal tubular necrosis caused by hypoxia in three adult pichis and mild congestion or hemorrhage in the adrenal glands of three adults.

Microgranulomas associated with migrating nematode larvae were noted in sections of skeletal muscle, adipose tissue, heart, tongue, lungs, liver, kidney, periureteral region, and reproductive organs. Several other organ lesions may have been related to parasite migration, such as mild periportal lymphocytic inflammation (61% of evaluated livers), inflammation of the ureter and surrounding connective tissue (66% of evaluated ureters), lymphocytic pyelonephritis (14% of evaluated kidneys), and eosinophilic inflammation of the testicular and spermatic cord tunics (10% of evaluated testes). Other types of hepatitis were rare and affected only four animals; they were probably associated with low-grade sepsis or parasite migrations. No signs of infection with T. cruzi or T. gondii were observed in any histologically evaluated pichi, including one of the individuals that was seropositive for T. cruzi.

DISCUSSION

This study constitutes the first report of a health evaluation in free-ranging and captive populations of Z. pichiy. In general, the studied wild populations were considered to be in good health; they were mainly affected by the consequences of external and internal parasitism. Several organs had lesions that were attributed to the stress and shock caused by the capture of pichis by poachers and their dogs. The collected tissues of some individuals had varying stages of autolysis because of an excessively long interval between the time of death and confiscation and sampling; not all samples were appropriate for histologic analysis. Nevertheless, the sampling method is considered to be an ethically justifiable means to perform population studies on endangered species that require extraction of tissue samples. Furthermore, it is often the only legally acceptable means to obtain samples from protected wildlife. Two caveats should be considered, however, when using poached animals for population studies. First, the sampled animals may not constitute a representative subsample of a wild population because the reproductive status or presence of disease may have changed their activity or altered their behavior, thus predisposing them to predation or poaching. Second, the total quantity and temporal distribution of samples may depend on poaching activity, as well as the frequency and success of antipoaching patrols.

Lesions and scars related to trauma were observed in a large proportion of wild-caught pichis. In males, skin and carapace lesions and lesions and abscesses on the penis may have been caused by intraspecific fights during the reproductive season. Territorial fights have been observed both in wild and captive male armadillos and have led to severe injuries of captive pichis. In our study, captive males were maintained in individual pens, while other males lived in contiguous enclosures. While interactions between these captive males were rare outside the breeding season, gonadally competent males often reached through the mesh fence with their forelegs in an attempt to fight with a potential rival. Nevertheless, because males did not have more lesions or scars than females, territorial fights are probably not the main cause of these trauma-related lesions. Some lesions may have been inflicted by predators; the osseous carapace does not provide complete protection against predators, and both the teeth of mammalian predators and the claws of raptors can easily fracture it. Lesions also may have been related to flight behavior as thorny areas are utilized for both shelter and for burrow entrances. Although thorns do not penetrate the carapace, they can cause lesions between the carapace bands. Indeed, 20% of the lesions and over 25% of scars were located on carapace bands or skin folds between bands. Finally, a large proportion of scars consisted of missing or worn-out scutes on the head shield and its superior border

(18%) or on the carapace border (15%). When foraging or digging, pichis will move the soil with their head. Sustained contact of the head shield with sand and stones may lead to abrasion and the worn appearance of scutes. Injuries or fracture of scutes on the loose upper border of the head shield and the sharply pointed hooklike scutes of the carapace border may occur when these structures get caught in a root or another obstacle while the animal is backing up.

Infected wounds were rare, although lesions (often covered in soil) were relatively common. Other than the abscess noted on the base of the penis of an adult male, only one adult male had infected lesions; this individual had been severely injured by a hunting dog. Most of the other observed lesions were superficial and probably healed rapidly. Only four individuals had leukocytosis with neutrophilia, indicating infection. No external lesions were observed in three of these animals, while the fourth only had a superficial wound on the left front leg; the infections thus probably affected one or more internal organs. It is possible, however, that infections occur more frequently in wild pichis than observed in this study. Infection may induce behavioral changes that reduce the probability of capture or such animals may be rapidly removed from the population by predators.

Both excessive humidity and a reduced availability of water in the pichi's environment have been found to cause negative effects in wild pichis. The state of dehydration observed in one adult male and the tubular mineralization of the kidneys of 11 adult animals that were evaluated histologically may be explained by the low precipitation rates and absence of open water sources in the natural habitat of pichis. Elevated ambient humidity, on the other hand, has been reported to cause disease in wild populations and has caused cutaneous lesions in captive pichis. Several captive individuals developed moist dermatitis with epidermal detachment and infection of the lesions with bacteria that are commonly found in soil, such as Pseudomonas, Citrobacter, and Escherichia coli. The temporary maintenance of these pichis in 200-l barrels, in which the substrate may have retained humidity due to reduced exposure to sunlight and suboptimal ventilation, seems to have led to an increased permeability of the skin's stratum corneum. Skin abrasions may then have occurred during intense contact of the compromised stratum corneum with the abrasive substrate. Such lesions were not observed in wild pichis during this study, but several inhabitants of rural areas in Mendoza Province have reported that a disease locally known as "pichi plague," characterized by red spots on the skin covering large abscesses, has led to local extinctions. The locals consistently associated this disease with extended periods of rainfall. Consequently, it is possible that the "pichi plague" is caused by opportunistic bacterial colonization of the skin compromised by the effects of excessive moisture.

The majority of observed enteroparasites were nematodes, primarily trichostrongylids; protozoans and cestodes also were observed. The most common species were *Aspidodera fasciata* and *Aspidodera scoleciformis* (Heterakoidea). Adult specimens of this genus, which are commonly found in armadillos (Navone, 1986; Fujita et al., 1995), were also regularly observed in fecal matter of *Z. pichiy* collected for other studies.

Many histologic lesions were associated with parasitism or parasite larva migration. The most prominent skin lesions involved perivascular inflammation consistent with hypersensitivity that sometimes extended to the adipose tissue, and these were most likely due to parasitism by fleas, ticks, and mites. Fleas were commonly observed on wild pichis, while *Amblyomma pseudoconcolor* was observed for the first time on a pichi during this study (Superina et al., 2004). Only a single intrafollicular mite was found, but these ectoparasites can be difficult to demonstrate histologically, especially when they are present in low numbers. Parasitism was the most probable cause of gastritis and the mild to moderate, chronic enteritis and colitis observed in 94% of all pichis. Occasionally, this process was associated with protozoa, mainly coccidian, or nematodes. Lesions associated with migrating nematode larvae were found in a wide range of tissues. Aspidodera spp. are the most common intestinal parasites of pichis, but larval migration has not been reported in this genus, and none of the other Heterakoidea species studied to date seems to have a larval phase outside the gut (Read and Skorping, 1995). Larval migration occurs in some, but not all trichostrongylid nematodes (Read and Skorping, 1995), but further studies are needed to determine whether the life cycle of the trichostrongylids that parasitize armadillos includes a migratory tissue phase.

Seven juveniles and eight adults had Sarcocystis cysts in the skeletal muscle, which were sometimes associated with mild lymphocytic inflammation in the muscle, presumably due to rupture of the cysts. It is probable that the mild lymphocytic inflammation observed in the tongue and the myocardium of nine and 11 animals, respectively, was caused by rupture of protozoan cysts, although cysts were only noted in the tongue of an adult female and in the heart muscle of an adult male. Several Sarcocystis species have been identified in the skeletal muscle and tongue of North and South American armadillos (Howells et al., 1975; Lindsay et al., 1996; Tanhauser et al., 2001), and the prevalence can be as high as 100% (Lindsay et al., 1996). The nine-banded armadillo has been identified as a naturally infected intermediate host of Sarcocystis neurona (Cheadle et al., 2001; Tanhauser et al., 2001). The zoonotic potential of the Sarcocystis species affecting wild pichis is considered to be low because hunters and consumers usually eat well-cooked rather than raw or undercooked pichi meat. Furthermore, the intermediate as well as the definitive hosts of each *Sarcocystis* species are considered to be specific or limited to closely related species (Fayer, 2004).

Besnoitia cysts were present in the lungs of 24 wild pichis. No other report on Besnoitia infection of any armadillo species or of a wild mammal species of Argentina has been found in the literature; the only previous report of *Besnoitia* from Argentina involved domestic rabbits (Venturini et al., 2002). This study therefore documents for the first time besnoitiosis in an armadillo and in a wild mammal native to Argentina. Although less severe, besnoitiosis was generally associated with acute and chronic inflammatory processes resembling the lesions of pulmonary besnoitiosis in maras (Dolichotis patagonum; Juan-Salles et al., 2004), and this protozoan was the likely cause for the high prevalence of chronic inflammation in the lungs. *Besnoitia* cysts were also present in the spleen of one adult pichi. The identification of *Besnoitia* in a relatively large proportion of the studied wild pichis requires further research to evaluate its clinical significance and to identify its definitive host.

In conclusion, this study suggests that the wild pichi populations of Mendoza Province, Argentina, have multiple forms of parasitism but generally are in good health. The results reported here lay the foundation for future health assessments of wild pichi populations.

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LITERATURE CITED

- CANDIA, R., AND A. D. DALMASSO. 1995. Dieta del guanaco (*Lama guanicoe*) y productividad del pastizal en la Reserva La Payunia, Mendoza (Argentina). Multequina (Mendoza, Argentina) 4: 5–15.
- —, S. PUIG, A. DALMASSO, F. VIDELA, AND E. MARTINEZ CARRETERO. 1993. Diseño del plan de manejo para la Reserva Provincial La Payunia (Malargüe, Mendoza). Multequina (Mendoza, Argentina) 2: 5–87.
- CARILLO, C. G., D. M. MEYERS, AND B. SZYFRES. 1972. Bataviae group Leptospirae isolated from armadillos in Argentina. Tropical and Geographical Medicine 24: 377–381.
- CHEADLE, M. A., S. M. TANHAUSER, J. B. DAME, D. C. SELLON, M. HINES, P. E. GINN, R. J. MACKAY, AND E. C. GREINER. 2001. The nine-banded armadillo (*Dasypus novemcinctus*) is an intermediate host for *Sarcocystis neurona*. International Journal for Parasitology 31: 330–335.
- FAYER, R. 2004. Sarcocystis spp. in human infections. Clinical Microbiology Reviews 17: 894–902.
- FINECOLD, S. M., AND E. J. BARON. 1986. Bailey and Scott's diagnostic microbiology. 7th Edition. The C. V. Mosby Company, St. Louis, Missouri, 914 pp.
- FONSECA, G. A. B., AND J. M. AGUIAR. 2004. The 2004 Edentate Species Assessment Workshop. Edentata 6: 1–26.
- FUJITA, O., N. ABE, Y. OKU, L. SANABRIA, A. INCHAUSTTI, AND M. KAMIYA. 1995. Nematodes of armadillos in Paraguay: A description of a new species Aspidodera esperanzae (Nematoda: Aspidoderidae). Journal of Parasitology 81: 936– 941.
- HAWKEN M. L. (ed.). 1983. A manual of laboratory techniques. 2nd Edition. British Small Animals Veterinary Association, Cheltenham, UK, 107 pp.
- HENDRIX, C. M. 1998. Diagnostic veterinary parasitology. 2nd Edition. Mosby Publication, St. Louis, Missouri, 321 pp.
- HOWELLS, R. E., A. D. V. CARVALHO, M. N. MELLO, AND N. M. RANGEL. 1975. Morphological and histochemical observations on *Sarcocystis* from the nine-banded armadillo, *Dasypus novemcinctus*. Annals of Tropical Medicine and Parasitology 69: 463–474.

- JUAN-SALLES, C., G. R. HERNANDEZ, M. M. GARNER, AND B. C. BARR. 2004. Pulmonary besnoitiasis in captive maras (*Dolichotis patagonum*) associated with interstitial pneumonia. Veterinary Pathology 41: 408–411.
- LINDSAY, D. S., R. MCKOWN, S. J. UPTON, C. T. MCALLISTER, M. A. TOLVIO-KINNUCAN, J. K. VEATCH, AND B. L. BLAGBURN. 1996. Prevalence and identity of *Sarcocystis* infections in armadillos (*Dasypus novemcinctus*). Journal of Parasitology 82: 518–520.
- MAZZA, S., S. MIYARA, AND H. E. SANJURJO. 1935. Comprobación de animales domésticos y de nuevas especies de mamíferos silvestres portadores de Schizotrypanum cruzi en los alrededores de la Ciudad de Mendoza. In Novena Reunión de la Sociedad Argentina de Patología Regional, Mendoza, 1–4 October 1935, pp. 548– 558.
- NAVONE, G. T. 1986. Estudios parasitológicos en edentados argentinos. II. Nematodes parásitos de armadillos: Aspidodera fasciata (Schneider, 1866); Aspidodera scoleciformis (Diesing, 1851) y Aspidodera vazi Proença, 1937 (Nematoda-Heterakoidea) Neotropica 32: 71–79.
- OLSON, D. M., E. DINERSTEIN, E. D. WIKRAMANAYAKE, N. D. BURGESS, G. V. N. POWELL, E. C. UNDERWOOD, J. A. D'AMICO, I. ITOUA, H. E. STRAND, J. C. MORRISON, C. J. LOUCKS, T. F. ALLNUTT, T. H. RICKETTS, Y. KURA, J. F. LAMOREUX, W. W. WETTENGEL, P. HEDAO, AND K. R. KASSEM. 2001. Terrestrial ecoregions of the world: A new map of life on Earth. BioScience 51: 933–938.
- PERALTA DE GALMARINI, I., AND E. MARTINEZ CARRETERO. 1995. Guías botánicas para la Provincia de Mendoza. II. Reserva Natural Telteca. INCA Editorial, Mendoza, Argentina, 60 pp.
- QUEVEDO, F., J. LASTA, AND B. MARTÍNEZ. 1978. The armadillo as a reservoir host for Salmonella. In The armadillo as an experimental model in biomedical research. Pan American Health Organization, Caracas, Venezuela, pp. 115–117.
- READ, A. F., AND A. SKORPING. 1995. The evolution of tissue migration by parasitic nematode larvae. Parasitology 111: 359–371.
- SUPERINA, M. 2008. The ecology of the pichi Zaedyus pichiy in western Argentina. In The biology of the Xenarthra, S. F. Vizcaíno and W. J. Loughry (eds.). University Press of Florida, Gainesville, pp. 313–318.
 - , AND P. BOILY. 2007. Hibernation and daily torpor in an armadillo, the pichi (*Zaedyus pichiy*). Comparative Biochemistry and Physiology–Part A 148: 893–898.
 - ——, AND R. MERA Y SIERRA. 2008. Hematology and serum chemistry values in captive and wild pichis, *Zaedyus pichiy* (Mammalia, Dasypodidae). Journal of Wildlife Diseases 44: 902–910.

- —, A. A. GUGLIELMONE, A. J. MANGOLD, S. NAVA, AND M. LARESCHI. 2004. New distributional and host records for *Amblyomma pseudoconcolor* Aragão, 1908 (Acari: Ixodidae: Amblyomminae). Systematic & Applied Acarology 9: 41–43.
- TANHAUSER, S. M., M. A. CHEADLE, E. T. MASSEY, B. A. MAYER, D. E. SCHROEDTER, J. B. DAME, E. C. GREINER, AND R. J. MACKAY. 2001. The ninebanded armadillo (*Dasypus novemcinctus*) is

naturally infected with *Sarcocystis neurona*. International Journal for Parasitology 31: 325–329.

VENTURINI, L., M. PETRUCCELLI, M. PISCOPO, J. M. ÚNZAGA, M. C. VENTURINI, D. BACIGALUPE, W. BASSO, AND J. P. DUBEY. 2002. Natural *Besnoitia* sp. infection in domestic rabbits from Argentina. Veterinary Parasitology 107: 273–278.

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