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## Health Evaluation of Pampas Deer (*Ozotoceros bezoarticus celer*) at Campos del Tuyú Wildlife Reserve, Argentina

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ABSTRACT: Samples from 14 free-ranging pampas deer (Ozotoceros bezoarticus celer) were collected in 1995 and 1998, at Campos del Tuyú Wildlife Reserve, Buenos Aires, Argentina. Hematology, serum chemistries, minerals and metals, and fecal parasites were analyzed. In addition, fecal ova and parasites were evaluated seasonally during 1998-2000. Serology for infectious diseases included bluetongue, brucellosis, bovine respiratory syncytial virus infection, bovine viral diarrhea/mucosal disease, infectious bovine rhinotracheitis, Johne's disease (paratuberculosis), foot and mouth disease (FMD), leptospirosis (eight serovars), epizootic hemorrhagic disease, and parainfluenza-3 (PI-3). Three (21%) pampas deer had antibodies to Leptospira spp. and six (43%) to PI-3 virus. Serologic results for all other infectious agents were negative. Domestic cattle (n=27) included in this study for comparison had antibodies to Leptospira, infectious bovine rhinotracheitis virus, bovine viral diarrhea virus, and PI-3 virus (74-100% of tested animals) and one animal (4%) to *Brucella* sp. All cattle had antibodies to FMD virus attributable to vaccination. This study provides the first data on the health status of the southernmost subspecies of pampas deer.

*Key words:* Argentina, disease, hematology, *Ozotoceros bezoarticus celer*, pampas deer, parasites, serology, survey.

The southernmost subspecies of pampas deer (*Ozotocerus bezoarticus celer*) was the most abundant herbivore of the Argentinean pampas grasslands until the 19th century (Cabrera, 1943; Jackson and Langguth, 1987). This deer is considered a rare species in South America (Holloway, 1975), and is one of the most endangered species in Argentina (García Fernández et al., 1997). Only two isolated populations of this subspecies remain in the wild (Jackson and Langguth, 1987). The dramatic decrease in pampas deer populations has been attributed to commercial and game overhunting (Daguerre, 1970; Thornback and Jenkins, 1982), habitat alteration by agriculture and livestock (Vigglizo, 1994), and diseases introduced by cattle (Sáenz, 1967). Diseases affecting this subspecies have not been studied and reports from the past century are occasional and inaccurate (Jackson and Langguth, 1987).

Identifying factors affecting health of endangered species populations is essential for management and for development of captive breeding and reintroduction and repopulation programs (Wolff and Seal, 1993; Karesh and Cook, 1995). As part of a long term study to evaluate factors limiting pampas deer recovery, we conducted health evaluations in 1995 and 1998. The objective of this study was to establish baseline health parameters for pampas deer and evaluate their exposure to common cattle pathogens.

The study area was located in General Lavalle County (36°15′S, 56°55′W), Buenos Aires, Argentina. The Campos del Tuyú Wildlife Reserve (CDT; 3,025 ha), is separated from surrounding cattle ranches by natural barriers, which restrict cattle movements but allow pampas deer to move freely in and outside the reserve. The area is known for low livestock productivity (stocking rates of 0.5 animals per ha and 86% calving success). Disease control in CDT neighboring ranches is limited to mandatory vaccination programs for foot and mouth disease (FMD) and brucellosis (BR) (SENASA, 1997). Some ranchers occasionally immunize for clostridial diseases and randomly deworm their cattle.

Disease	Test procedure <sup>a</sup>	Number positive/number tested (%)		
	(positive titer)	Pampas deer	Domestic cattle	
Bluetongue <sup>b</sup>	AGID	0/14	0/27	
0	(NA)			
Infectious bovine rhinotracheitis <sup>b</sup>	SN	0/14	20/27	
	(1:8)		(74%)	
Bovine viral diarrhea <sup>b</sup>	SN	0/14	20/27	
	(1:8)		(74%)	
Brucellosis <sup>b</sup>	Agg	0/14	1/27	
	(1:50 deer; 1:100 cattle)		(4%)	
Epizootic hemorrhagic disease <sup>b</sup>	AGID	0/7	Not tested	
	(NA)			
Foot-and-mouth disease <sup>b</sup>	c-ELISA	0/14	27/27 <sup>c</sup>	
	(1:10)		(100%)	
Johnes disease <sup>b</sup>	ELISA	0/14	0/27	
	(NA)			
Leptospirosis <sup>b,d</sup>	MAT	3/14	24/27	
	(1:25 deer; 1:200 cattle)	(21%)	(89%)	
Parainfluenza-3 <sup>b</sup>	HI	6/14	20/27	
	(1:5)	(43%)	(74%)	
Bovine respiratory syncytial virus	SN	0/7	Not tested	
infection <sup>e</sup>	(1:4)			
Bovine leucocis <sup>b</sup>	AGID	Not tested	0/27	
	(NA)			

TABLE 1. Serologic tests performed, methods used, and results of testing pampas deer (*Ozotoceros bezoarticus celer*) and domestic cattle for infectious diseases in Argentina.

<sup>a</sup> AGID=agar gel immunodiffusion; cELISA=competitive enzyme-linked immunodiffusion assay; ELISA=enzyme-linked immunodiffusion assay; Agg=tube agglutination test; SN=serum neutralization; MAT=microscopic agglutination test; HI=hemagglutination inhibition.

<sup>b</sup> Tests performed at Pathobiology Laboratory of C.I.C.V., INTA (Instituto Nacional de Tecnología Agropecuaria), Castelar, CC 77, Moron, Buenos Aires, Argentina.

<sup>c</sup> Vaccination titers.

<sup>d</sup> Leptospira interrogans serovars ballum, castellonis, canicola, grippotyphosa, icterohaemoragiae, copenhageni, pomona, pyrogenes, sejroe, wolffi, tarassovi.

e Tests performed at Diagnostic Laboratory, Oklahoma State University, Stillwater, Oklahoma, USA.

Seven female and seven male deer were immobilized in December 1995 and April 1998 at CDT and neighboring ranches. Blood samples were collected from the jugular vein and feces were collected manually from the rectum. Additionally, 37 deer feces were collected seasonally during 1998–2000, either fresh from the ground (11%) or after the animals were observed defecating (89%). To establish background information on cattle pathogen exposure in the area, 27 blood samples were collected at four neighboring ranches from March to June 1996.

Basic hematology was conducted at the field site. Deer and cattle sera were harvested after centrifugation of blood and frozen in liquid nitrogen. Table 1 summarizes the serologic tests performed, the methods used, and prevalence estimates for infectious disease exposure in deer and cattle. Table 2 describes parasitologic techniques used for fecal analysis and results. Serum chemistries were processed on a wet automated analyzer (Hitachi 747 Wet Chemistry Analyzer, Hitachi) at Wild-Life laboratory, Buenos Aires, Argentina. Chlorinated pesticides and polychlorinated biphenyls were analyzed at the Animal Health Diagnostic Laboratory (Michigan State University, East Lansing, Michigan, USA) as described by Price et al. (1986). Though limited by small sample size, a Kruskal-Wallis one-way ANOVA was used

Parasites	Number positive/ number tested (%)	Test procedure
		Å
Unknown nematode ova	10/37	Sugar flotation <sup>a</sup>
	(27%)	
Capillaria spp. ova	2/37	Sugar flotation
	(5%)	
Moniezia spp. ova	5/37	Sugar flotation
	(14%)	-
Coccidia	1/37	Sugar flotation
	(2%)	0
Ostertagia spp.	30/33	Third stage larvae culture <sup>b</sup>
	(91%)	0
Cooperia spp.	7/33	Third stage larvae culture
	(21%)	
Oesophagostomun spp.	13/33	Third stage larvae culture
	(39%)	
Trichostrongylus spp.	20/33	Third stage larvae culture
	(61%)	Third stage failvae culture
Haemonchus spp.	1/33	Third stage larvae culture
	(3%)	Tillitu stage laivae culture
Dyctiocaulus spp.	3/37	Concentration of first store
		Concentration of first stage
	(8%)	larvae <sup>c</sup>
Unknown 1st stage larvae <sup>d</sup>	35/37	Concentration of first stage
	(95%)	larvae

TABLE 2. Results of fecal examinations of pampas deer for parasites, 1998–2000.

<sup>a</sup> Qualitative sugar flotation methods were used to determine the presence of gastrointestinal parasite eggs (Bembrook, 1965).
 <sup>b</sup> Third stage nematode larvae were cultured by method of Henriksen and Korsholm (1983) and later identified using the keys for bovine larvae of Dr. Roman Niec (Fiel et al., 1998).

 $^{\rm c}$  Concentration and identification of first stage larvae of lung parasites was done by Baermann technique (Baermann, 1917).  $^{\rm d}$  Unidentified first stage "S" shaped tail larvae with a dorsal spine.

to determine if test results differed among individuals of each sex (P < 0.05) (Zar, 1996).

All deer examined in this study (n=14) were in good physical condition and no abnormalities of clinical significance were found. Weights ranged from 26–33 kg (mean=29 kg, SD=2.49, n=7) for females and from 24.5–38 kg (mean=32.6 kg, SD=4.30, n=7) for males.

Results for hematology, serum chemistries, enzymes, and minerals are shown in Table 3. Total solids and percent of monocytes were higher in males than in females (P < 0.05) while alkaline phosphatase (ALK) was higher in females (P < 0.05). For seven deer sampled in 1995, values for chromium and boron were below detectable limits (<1 ppm and <0.1 ppm, respectively). For the same individuals, chlorinated pesticides and polychlorinated biphenyls were also under the test threshold (0.001–0.007 ppm and 0.05 ppm respectively).

Baseline hematology values have not been previously described for this subspecies. White blood cell and differential counts were similar to those reported for O. bezoarticus leucogaster captured in Brazil (Duarte et al., 1993). However, these authors report lymphocyte predominating in the differential as in cattle (Duncan et al., 1997). Those data are in contrast to our findings and to data from other Neotropical deer (Santos, 1999). Even so, a high variability in leukocyte counts for deer has been described by several authors (Duarte et al., 1993; Kay, 1994). Many of these changes are attributed to age and sex as well as nutrition, health, and behavioral status of sampled animals (Kitchen, 1986; Kay, 1994). Our data show higher mean

Test (units)	Mean	Median	STD	Min	Max	n
Hematocrit (%)	43.2	42.0	5.5	36.5	55.0	13
Total solids (g/dl)	5.9	5.8	0.5	5.2	6.8	13
White blood cells (WBC)						
$(\text{cells/ml}^3 \times 10^3)$	6.20	5.35	2.50	2.80	11.20	12
Neutrophils (% of WBC)	48.6	44.0	16.4	29.0	78.0	13
Lymphocytes (% of WBC)	34.5	35.0	16.8	8.0	68.0	13
Monocytes (% of WBC)	4.1	4.0	2.1	0.0	8.0	13
Eosinophils (% of WBC)	12.2	3.0	15.9	0.0	48.0	13
Basophils (% of WBC)	0.5	0.0	0.9	0.0	2.0	13
Glucose (mg/dl)	169	153	63	72	279	13
Creatinine (mg/dl)	1.4	1.3	0.4	1.1	2.2	8
Blood urea nitrogen (mg/dl)	34	39	13	6	48	9
Total protein (g/dl)	5.3	5.5	0.9	3.3	6.8	13
Albumin (g/dl)	2.4	2.4	0.3	2.0	2.8	13
Globulins (g/dl)	3.0	3.1	0.9	1.2	4.4	13
Alb./Glob.	0.9	0.8	0.3	0.5	1.8	13
Alkaline phosphatase (IU/l)	116	108	39	75	183	8
Alanine aminotransferase						
(IU/l)	15	15	3	12	18	8
Lactate dehydrogenase (IU/l)	274	261	51	195	354	8
Creatine kinase (IU/l)	126	54	149	30	429	7
Calcium (mg/dl)	9.3	9.0	0.9	7.8	10.7	13
Phosphorus (mg/dl)	4.8	4.8	1.8	1.8	7.5	13
Sodium (mEq/l)	144	148	12	121	156	13
Potassium (mEq/l)	4.8	4.8	0.5	4.1	5.8	13
Chloride $(mEq/l)$	99	108	17	75	113	13
Copper (mcg/ml)	0.8	0.8	0.2	0.5	1.1	6
Iron (mcg/ml)	1.8	1.8	0.6	0.9	2.4	6
Magnesium (mg/dl)	2.4	2.4	0.5	1.7	3.3	13
Zinc (mg/l)	0.4	0.4	0.0	0.3	0.4	6
Selenium (ng/ml)	16	14	8	8	26	6

TABLE 3. Hematology, serum chemistries, enzymes, minerals and metal values for pampas deer at Campos del Tuyú Wildlife Reserve, Argentina.

packed cell volume, but within normal ranges, than those described for captive marsh deer (*Blastocerus dichotomus*) from Brazil (Santos, 1999) and for white-tailed deer (*Odocoileus virginianus*) (Kitchen, 1986; International Species Inventory System, undated).

To our knowledge, data reported here are the first published serum chemistry, mineral, and metal values for free-ranging pampas deer. Most of these values were similar or lower than those reported for captive marsh deer and white-tailed deer (Kitchen, 1986; Santos, 1999; International Species Inventory System, undated). However, glucose tended to be higher in our study. Increase in glucose within 10– 60 min after chemical immobilization has been reported for deer restrained with xylazine and etorphine (Mautz et al., 1980). Therefore, use of carfentanil citrate (Wildnil, Wildlife Pharmaceuticals, Fort Collins, Colorado, USA) and xylazine (Cervizine, Wildlife Pharmaceuticals) to immobilize pampas deer in our study could be responsible for the increased values (Uhart, unpubl. data). Likewise, significant variations in all serum components have been attributed to biological, genetic, and environmental factors, as well as capture methods used and excitement of the animal (Kitchen, 1986). Values presented in this paper are based on a small sample size and should be interpreted considering the influence of these factors.

Pampas deer had antibodies against two

of 11 infectious agents, whereas cattle were positive for six of nine pathogens. Serology results are detailed in Table 1. Deer and cattle were seropositive to *Leptospira*; evidence of two serovars found in deer, pomona and wolffi, were also found in cattle. Of those pampas deer with positive antibody titers to *Leptospira interrogans*, one had a 1:100 titer to serovar wolffi and the others had titers of 1:100 and 1:400 to serovar pomona. Leptospiral antibody titers in cattle included serovars castellonis (37%), tarassovi (26%), wolffi (22%), and pomona (4%) and ranged from 1:200 to 1: 1,600. Antibody titers above 1:400 were only found for serovars wolffi and tarassovi.

Leptospirosis occurs regularly in domestic ungulates and has been reported from a wide range of wild mammals. Antibody titers in pampas deer in our study were similar to those reported for other wild ungulates (Goyal et al., 1992; New et al., 1993). Mathias et al. (1999) reported 24% of pampas deer sampled in Brazil seropositive for serovars hardjo, wolffi, and mini.

Antibodies to parainfluenza 3 (PI-3) virus were found in six of 14 deer (43%) and in 20 of 27 cattle (74%) tested. Infections with PI-3 virus in wild ungulates are usually subclinical, though they can cause interstitial pneumonia (Van Campen and Early, 2001). High prevalences (80-100%) to PI-3 have been reported for other deer species, as reviewed by Van Campen and Early (2001). Nevertheless, the significance of PI-3 for this deer population is unknown. Serologic tests used for deer samples were those of standard application for domestic cattle (OIE, 1996) but were not validated for this species. Therefore, results should be interpreted with caution (Hietala and Gardner, 1999). Additionally, our small sample size limits our interpretation of disease prevalence within this population.

Little information has been published on infectious diseases in pampas deer. Mathias et al. (1999) and Duarte et al. (1993) reported negative serological findings for FMD, brucellosis, and bluetongue in *O. bezoarticus leucogaster* from Brazil. Morbidity and mortality due to FMD-like disease was reported for *O. bezoarticus celer* from Samborombón (Bianchini and Luna Pérez, 1972; Jackson, unpubl. data) living in proximity to axis deer (*Axis axis*), fallow deer (*Dama dama*), and cattle.

Six parasite genera were found to infect pampas deer, with slight prevalence changes occurring seasonally. Ova of gastrointestinal parasites were present in the feces of four of 14 immobilized deer, including unidentified nematode ova in four deer and *Moniezia* spp. in one animal. Parasite ova were found in 18/37 (49%) samples collected seasonally. Detailed findings are summarized in Table 2.

Overall, the endoparasites found in our study are widely distributed and have been described for other deer species and domestic ruminants worldwide (Duarte et al., 2001; Hoberg et al., 2001). Amongst gastrointestinal parasites found in our study, only Ostertagia spp. and Haemonchus spp. are recognized as potential primary pathogens for deer (Hoberg et al., 2001). Lungworms identified in our deer are considered pathogenic for deer (Munro, 1994b), but have not been mentioned for South American species (Duarte et al., 2001). Almost all seasonal samples collected for this study were positive for unidentified first stage larvae with a dorsal spine. Because of the unknown significance of this finding, further investigations are warranted.

*Fasciola hepatica* has been reported in pampas deer from Uruguay (Hernández Russo et al., 1996). Its presence can not be excluded in our study because sedimentation methods were not used. Duarte et al. (1993, 2001) have described many of the parasites found in our study in pampas deer from Brazil.

This study provides the first data on the health status of the southernmost freeranging subspecies of pampas deer. Longterm conservation of this species cannot be achieved without integrating health aspects into future management actions.

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