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Survey of Vector-borne Comorbidities in a Leishmania-endemic Canine Population from Paraguay

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Introduction

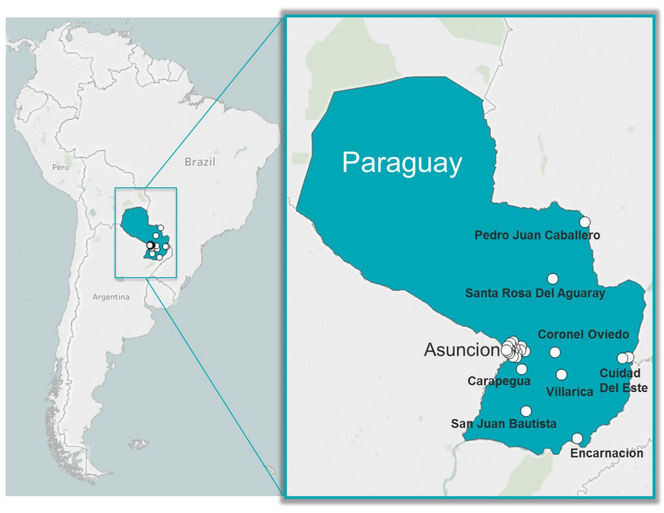
Canine Visceral Leishmaniasis (CVL) is an endemic parasitic disease in Paraguay with a majority of seropositive dogs observed to be asymptomatic for clinical signs consistent with CVL. Little is known about the full landscape of canine vector-borne diseases (CVBD) infecting dogs in Paraguay and the role concurrent infections may play in the onset of CVL. The objective of this study was to assess the incidence of Leishmania and other CVBD agents in a population of pet dogs from Leishmania-endemic regions of Paraguay.

Methods

A total of 100 serum samples and 400 whole blood samples were collected. The samples originated from 32 unique postal codes distributed across the eastern region of Paraguay. Nucleic acid was extracted from the whole blood samples and real-time PCR was run to detect the following pathogens: *Leishmania*; *Ehrlichia canis*; *Ehrlichia chaffeensis*; *Ehrlichia ewingii*; *Anaplasma platys*; *Anaplasma phagocytophilum*; *Babesia vogeli*; *Babesia gibsoni*; *Babesia canis*; *Rickettsia spp*.; *Bartonella spp*.; *Mycoplasma haemocanis*; *Candidatus Mycoplasma haematoparvum*; *Hepatozoon canis*; *Hepatozoon americanum*; *Dirofilaria immitis*; and *Trypanosoma cruzi*. ELISAs were performed on serum to detect the presence of antibodies to *Leishmania, Ehrlichia spp., E. canis, E. ewingii, E. chaffeensis, Anaplasma spp., A. platys, A. phagocytophilum, Babesia gibsoni*, *Babesia canis/vogeli*, and *Borrelia burgdorferi*, as well as antigen from *D. immitis* and *Angiostrongylus vasorum*.

Results

Serology Results (n=100)		
CVBD Pathogen	Pos, n	% Pos, n
Leishmania	**46**	**46%**
Ehrlichia spp.	**51**	**51%**
Ehrlichia canis	**51**	**51%**
E. chaffeensis	5	5%
E. ewingii	3	3%
Anaplasma spp.	**46**	**46%**
A. platys	**23**	**23%**
A. phagocytophilum	3	3%
Babesia canis / B. vogeli	**25**	**25%**
B. gibsoni	1	1%
Borrelia burgdorferi	0	0%
Dirofilaria immitis	1	1%
Angiostrongylus vasorum	0	0%
Leishmania + other CVBD	**37**	**37%**



Occurrence of CVBD co-infections Leish-positive samples by whole blood PCR (n=71)		
CVBD Pathogen	Pos, n	% Pos, n
None	43	60.6%
Ehrlichia canis	**20**	**28.2%**
Anaplasma platys	**5**	**7.0%**
Babesia vogeli	**4**	**5.6%**
M. haemocanis	2	2.8%
Cand. M. haematoparvum	2	2.8%
Hepatozoon canis	1	1.4%

PCR Results (n=400)		
CVBD Pathogen	Pos, n	% Pos, n
Leishmania	**71**	**17.8%**
Trypanosoma cruzi	0	0.0%
Ehrlichia canis	**110**	**27.5%**
E. chaffeensis	0	0.0%
E. ewingii	0	0.0%
Anaplasma platys	**45**	**11.3%**
A. phagocytophilum	0	0.0%
Babesia vogeli	**37**	**9.3%**
B. canis	0	0.0%
B. gibsoni	0	0.0%
Rickettsia spp.	**1**	**0.3%**
Bartonella spp.	0	0.0%
Mycoplasma haemocanis	**15**	**3.8%**
Cand. M. haematoparvum	**12**	**3.0%**
Hepatozoon canis	**6**	**1.5%**
H. americanum	0	0.0%
Dirofilaria immitis	0	0.0%
Leishmania + other CVBD	**28**	**7.0%**

Discussion

Real-time PCR detected Leishmania infections in this population at a rate of 18%. Additional CVBD pathogens detected by PCR included *E. canis* (28%), *A. platys* (11%), *B. vogeli* (9%), *M. haemocanis* (4%), *Cand. M. haematoparvum* (3%), and *H. canis* (2%). All of these pathogens are known to be transmitted by *Rhipicephalus sanguineus* ticks. Overall, 7% of samples were PCR positive for *Leishmania* and at least one other CVBD pathogen with *E. canis* being the most common co-infecting agent. Of the serum samples tested by ELISA, 46% were seropositive for *Leishmania*. High seropositive rates were also observed for *Ehrlichia spp*. (51%), *E. canis* (51%), *Anaplasma spp*. (46%), *A. platys* (23%), and *Babesia spp*. (26%). Overall, 37% of samples were seropositive to *Leishmania* and at least one additional CVBD pathogen. These findings indicate that co-infections with *Leishmania* and other CVBD pathogens, particularly those transmitted by *Rhipicephalus sanguineus* ticks, are relatively common in Paraguayan dogs. The potential for co-infection with other CVBD pathogens should be taken into consideration when diagnosing and treating CVL.